Textbook for
MRCOG-1

Basic Sciences in Obstetrics & Gynaecology

As per the latest RCOG Modules

Includes more than 1000 SBAs

Richa Saxena

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Dedicated to

My mother Mrs Bharati Saxena
For always being there…..

"My mother was the most beautiful woman I ever saw. All I am I owe to my mother.
I attribute all my success in life to the moral, intellectual
and physical education I received from her."

-George Washington

Preface

"I wanted a perfect ending. Now I have learnt, the hard way, that some poems don't rhyme and some stories don't have a clear beginning, middle, and end.

Life is about not knowing, having to change, taking the moment and making the best of it, without knowing what is going to happen next.

It is a delicious ambiguity."

—**Gilda Radner**

Years ago, after acquiring the MD (Obstetrics and Gynaecology) degree, similar to many young Indian doctors, I too wanted to add a foreign qualification in my credentials. Though I had completed part of the process, I could not complete it in entirety because of some health-related issues which prevented me in pursuing my career as a surgeon. However, life has its own ways and here I am writing a book for the doctors wishing to obtain the degree "Membership of Royal College of Obstetricians and Gynaecologists", (MRCOG, UK). For more details related to the MRCOG examination, kindly refer to the Royal College of Obstetricians and Gynaecologists (RCOG) website, *https://www.rcog.org.uk/*. For details related to the Part 1 examination, kindly click on the link, *https://www.rcog.org.uk/en/careers-training/mrcog-exams/part-1-mrcog/format/*

This book, "Textbook for MRCOG-1" is intended for the doctors who are planning to appear in MRCOG Part 1 examination. The MRCOG examination is meant for those doctors (undergraduates as well as postgraduates) who wish to pursue their specialisation in obstetrics and gynaecology in the UK. This comprises of a two-part examination. Part 1 MRCOG is a written examination, which helps in the evaluation of basic and clinical sciences relevant to the subject.

Fundamental aspects of all the important subjects related to basic sciences in medicine have been covered in this book. This is inclusive of subjects such as anatomy, physiology, biochemistry and nutrition, pathology, microbiology and immunology, embryology, genetics, biophysics, epidemiology, endocrinology and pharmacology. There are also separate chapters on "Principles of Clinical Practice", "Obstetrics" and "Gynaecology". The text has been covered in accordance with the latest curriculum and examination format as described by the RCOG and has been written in an easy-to-understand manner, well-illustrated with pictures. Though it is not possible to cover the entire subject in a single chapter, most topics, which are important from the point of view of examination, have been adequately described.

According to the latest RCOG layout, the questions for the MRCOG examination would be in the "single best answer" or SBA format. For the purpose of self-assessment, a list of SBAs along with their answer keys has been provided at the end of each chapter. In total, approximately 1,000 SBAs are enlisted in this book. Therefore, the students preparing for this examination do not need to buy a separate book on SBAs.

Writing a book is a colossal task. It can never be completed without divine intervention and approval. Therefore, I have decided to end this preface with a small prayer of thanks to the Almighty, which I was taught in my childhood.

"Father, lead me day by day, ever in thy own sweet way.
Teach me to be pure and good and tell me what I ought to do."

—**Amen**

Simultaneously, I would like to extend my thanks and appreciation to all the related authors and publishers whose references have been used in this book. Book creation is teamwork, and I acknowledge the way the entire staff of M/s Jaypee Brothers Medical Publishers (P) Ltd., New Delhi, India, worked hard on this manuscript to give it a final shape. I believe that writing a book involves a continuous learning process. Though extreme care has been taken to maintain the accuracy while writing this book, constructive criticism would be greatly appreciated. Please e-mail me your comments at the e-mail address: *richa@drrichasaxena.com*. Also, please feel free to visit my website *www.drrichasaxena.com* for obtaining information related to various other books written by me and to make use of the free resources available for the doctors.

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Contents

Principles of Clinical Practice

Evidence-Based Medicine

The practice of evidence-based medicine combines clinical expertise and external evidence. This is an approach to medical practice, which aims at integrating individual clinical expertise with the best available external clinical evidence from systematic research in form of well-designed and conducted research trials. Clinical expertise implies the proficiency and judgment that the individual clinicians acquire through clinical experience and clinical practice. Health economic assessment is a central parameter in evidence-based medicine, especially while making judicious use of current best evidence to reach clinical decisions. Evidence-based medicine involves the conscientious, explicit and judicious use of current best evidence in making decisions about the care of individual patients. Evidence-based medicine is a guide only and we should not assume that all patients should be treated similarly according to the results of clinical trials. It is used to make decisions about the care of individual patients. Each patient is an individual, and the clinician must remember this while initiating treatment.

All types of clinical trials are included in the practice of evidence-based medicine. However, the methods must be critically appraised in order to assess the validity of the evidence. Objective measurements of disease outcome eliminate bias, are more scientific relative to subjective measures, and are therefore applicable to the practice of evidence-based medicine. Strongest degree of evidence coming from meta-analysis, systemic reviews and randomized controlled trials (RCTs) can yield the strongest recommendations, whereas evidence in form of case-control trials can yield only weak recommendations. Often an RCT will be conducted to assess the benefits or risks associated with a new, expensive treatment. Though RCTs reveal a strong degree of evidence, they are not the only trials that contribute to evidence-based medicine. Prospective trials, observational and cross-sectional studies all provide vital information that guides the process of daily decision-making. Grading criteria for various levels of evidence is described in **Table 1.1** and **Fig. 1.1**.

TABLE 1.1	Grading criteria for levels of evidence	
Levels of evidence	**Grading criteria**	**Grading of recommendations**
1a	Systematic review of RCTs including meta-analysis	A
1b	Individual RCT with narrow confidence interval	A
1c	All or none studies	B
2a	Systematic review of cohort studies	B
2b	Individual cohort studies and low quality RCT	B
2c	Outcome research studies	C
3a	Systematic review of case-control studies	C
3b	Individual case-control studies	C
4	Case series, poor quality cohort and case-control studies	C
5	Expert opinion	D

Abbreviation: RCT, randomized controlled trial

FIG. 1.1: Pyramid showing various levels of evidence

Consent

Informed Consent

Before undertaking any surgery, it is important for the doctor to take informed consent from the patient. Today, the informed consent is required for all operative procedures. The process involves counselling the patient about the various available surgical options so that the patient can select the best surgical procedure out of the various available options. In practice, the informed consent involves informing the patient about the diagnosis, degree of certainty regarding the diagnosis, the surgery that would be recommended in that case and possible alternatives along with their expected outcomes, risks and benefits. The patient outcome, if no therapy is administered must also be explained to the patient. The consent should be taken well in advance of surgery in a comfortable setting. The patient must be given adequate time to absorb the information, ask any questions if she feels so and then to make an informed decision. Effective communication between the patient and the surgeon is of utmost importance, while counselling the patient regarding various available treatment options. The surgeon may make use of written material (self-explanatory patient leaflets), visual aids (models), websites, etc. to explain the procedure to the patients. The patients must also be informed about the advantages, disadvantages, success and failure rates, and complications of the various procedures. The patient must be counselled even regarding the rare complications that are serious and may affect the individual's life. The patient should be given adequate time to interpret and absorb the information presented to him before making the final decision.

The informed consent requires the presence of following pieces of information: nature of the procedure; rationale of doing the procedure; advantages and disadvantages of doing the procedure; and availability of alternatives. The elements of informed consent are as follows:
❏ Disclosure of information
❏ Comprehension by the patient
❏ Voluntary transaction
❏ Validation

Disclosure of information: The patients must be explained about their diagnosis and also briefed about the various available treatment options, including no treatment and various medical, surgical and alternative therapies. Risks and benefits of each modality need to be explained in sufficient details so that a reasonable adult patient can understand the situation and make an informed choice.

Comprehension by the patient: The language and the descriptive material, which is used to explain the situation to the patient, must be appropriate to the patient's level of comprehension. The patients must be asked questions in between to ensure that they understand what they have been told.

Voluntariness: While making a decision, the patient must be free of coercion or constraints and must be able to choose freely. The patient should be mentally competent to be able to make a choice and there must be no evidence of limitation in her ability to understand the information. She must be in a condition to act independently on the basis of information that has been disclosed.

Validation: A written consent form must be given to the patient, which must be duly signed by her. Consent must be taken for each procedure, which is going to be performed even if they are being performed in a single setting. If an additional pathology is discovered at the time of surgery, the surgeon can legally operate on it, only if the condition is life-threatening. On the other hand, if the condition is not life-threatening, then the surgeon must finish the planned surgery and discuss the condition later with the patient.

Exceptions to the Informed Consent

There are four exceptions to the informed consent:
1. *Emergency situations*: If the relatives are unavailable, the patient is unconscious and is suffering from an emergency life-threatening condition. No consent is required from anyone if one feels that a criminal act has been perpetrated.
2. *Intentional relinquishing by the patient*: Waiver may be given by the patient in case of research projects or exploratory laparotomy.
3. *Mental illness*: The patient is mentally incompetent, i.e. the patient has been declared mentally unsound to be able to understand and take decisions appropriately. In this case, the court takes the responsibility for the patient.
4. *Therapeutic privilege*: In case the patient is unconscious or is in the state of confusion and there are no relatives, the physician can act in the patient's benefit without taking her consent.

Types of Consent

Implied Consent

Implied consent relates to situations in which the patient's behaviour indicates consent to what is proposed. For example, if a clinic appointment is sent to a patient and she duly attends, it can be assumed that she has given consent for being there.

Verbal Consent

In case of verbal consent, the patient gives a verbal approval for a proposed procedure. For example when the clinician tells the patient, "I am just going to take some blood from your arm", the patient gives a verbal consent by saying, "okay, doctor go ahead." Verbal informed consent is adequate for procedures such as blood investigations, cervical smear, etc. The procedure such as cervical smear should be preferably performed with a chaperone.

Written Consent

Implied and verbal consent are all right for the basics of daily practice. But as soon as the healthcare professional starts dealing with anything major, especially if there is any risk to the patient or her baby, legal backup with consent in writing would be required. Nowadays, standard forms for taking consent are available to make sure that all the legalities are covered.

Components of Consent

Consent has three main components:
1. *Capacity*: "Capacity" means the individual's ability to give consent.
2. *Information*: This requires provision of adequate, accessible information to enable a rational decision so that the patient is able to process the information and weigh up the pros and cons of the proposed treatment, the pros and cons of the other possible treatments and the pros and cons of having no treatment. The information, which patients may want to know, before deciding whether to consent to treatment or an investigation, may include the following:
   • Details of the diagnosis, prognosis, and the likely prognosis if the condition is left untreated
   • Uncertainties about the diagnosis including options for further investigation prior to treatment
   • Options for treatment or management of the condition, including the option not to treat
   • The purpose of a proposed investigation or treatment; details of the procedures or therapies involved, including subsidiary treatment such as methods of pain relief; how the patient should prepare for the procedure; and details of what the patient might experience during or after the procedure including common and serious side effects
   • For each option, explanations of the likely benefits and the probabilities of success and discussion of any serious or frequently occurring risks need to be done.
   • Advice about whether a proposed treatment is experimental
   • How and when the patient's condition and any side effects would be monitored or re-assessed
   • The name of the doctor who will have overall responsibility for the treatment and, where appropriate, names of the senior members of his or her team
   • Information regarding whether doctors in training will be involved in the care of the patient, and the extent to which students may be involved in an investigation or treatment
   • A reminder that patients can change their minds about a decision at any time
   • A reminder that patients have a right to seek a second opinion
   • Where applicable, details of costs or charges that the patient may have to meet

3. *Communication*: The individual must be able to let others know their decision. If they are unable to communicate the decision, they cannot give consent.

Gillick's Competence

In the early 1980s, the Department of Health issued a circular, which stated that a doctor could provide contraceptive advice or treatment to a girl under the age of 16 without parental knowledge or consent. Many parents were not happy about this because they thought that such policy might encourage their children to engage in sexual activity. One such parent was Victoria Gillick, who was a mother of 10, Roman Catholic and "pro-life activist". She sought assurances from her local health authority (West Norfolk and Wisbech) about her daughters. She wanted to know that no one would prescribe contraceptive advice or treatment for them without her consent. However, the Health Authority declined to provide assurances. Therefore, Mrs Gillick took them to court. She argued that a doctor providing contraception to an under-age girl would be "aiding and abetting" an unlawful act resulting in sexual intercourse with a minor. The local court found this in favour of the Health Authority. Mrs Gillick took the case to the Court of Appeal, which found it in her favour, stating that a child under the age of 16 could not give consent. The Department of health appealed to the House of Lords in 1985. The judgment was decided in favour of the Department, by a majority of the three judges who heard the case. The spokesman for the judges was Lord Fraser. Their view was that a child under the age of 16 could be competent to give consent. The concept of "Gillick competence" was derived from this, i.e. Gillick's competence can be described as the ability of an under-age child to give valid consent.

Fraser Guidelines

According to the Fraser guidelines, there are five conditions, which must be met for a child to be "competent". Fraser's competence is in preference to saying a child is "Gillick competent". This means that a doctor can provide contraceptive advice and treatment to a child under the age of 16 without parental consent. However, one of the following conditions needs to be fulfilled. These five conditions came to be known as the "Fraser Guidelines".
❑ The young person must understand the advice being given.
❑ The young person cannot be convinced to involve parents/carers or allow the medical practitioner to do so on their behalf.
❑ It is likely that the young person will begin or continue having intercourse with or without treatment/contraception.
❑ The young persons' physical or mental health (or both) is likely to suffer unless they receive treatment/contraception.

❑ The young person's best interests require administration of contraceptive advice, treatment or supplies without parental consent.

Gillick and Fraser originally related to contraception only. However, now they have tended to extend to cover other areas. In 1990, the Access to Health Records Act stated that a "Gillick competent" child could deny parental access to their health records.

Axon

"Axon" was a case relating to the provision of termination of pregnancy to the under-age child without parental involvement. This is likely to determine the law in relation to abortion services and the under-age girl for the foreseeable future.

Sue Axon from Wythenshawe in Manchester, went to court in 2005 in a case related to the ability of doctors to advise about or provide abortion services to under-age girls without the knowledge of the parents. Mrs Axon lost the case and decided not to pursue it further.

Bolam

"Bolam" is the term used for indicating whether the clinician had behaved in a reasonable way. It has risen from a legal case: Bolam v Friern Hospital Management Committee in 1957. From this came the "Bolam principle" relating to whether a doctor's actions had been reasonable. A doctor's behaviour would be judged legal if a substantial body of his/her peers would have behaved in the same way as the doctor had done. "Peers" means "equals", so that if you are a SpR, you would be compared with other SpRs; if you are a consultant, you shall be compared with other consultants. This meant that doctors' behaviour was used for defining what the reasonable behaviour by the doctors was. This was open to criticism.

In the "Bolitho case", the judge took the view that it was for the court to decide what was reasonable behaviour, not the medical profession. In other words, the court could dismiss the views and practices of this "substantial body of peers" as wrong.

The Mental Capacity Act 2005

The main aim of this act is to provide a legal framework for making decisions on behalf of those adults who lack the capacity for making a particular decision by themselves. Every possible step to confirm capacity must be taken before deciding that someone lacks capacity. If there is doubt about whether the patients have capacity or not, the health professional must get an expert opinion from consultant psychiatrist or psychologist having a background in dealing with patients having learning difficulties.

The legalities in such cases are wrapped up in the Mental Capacity Act 2005. A court order will be usually required to provide treatment in these cases. The court would normally expect to make a "one-off" decision relating to a particular treatment for an individual lacking capacity. If the court foresees that further decisions may be needed, it can appoint a "Deputy" to act on behalf of an individual who lacks capacity. The Deputy will have lasting power to make decisions on the patient's behalf over all matters, including medical care. In an emergency situation, treatment can be provided without a court order. However, in these cases it is sensible to get a second opinion to confirm that it is an emergency and that urgent treatment is necessary.

In these cases, relatives and carers are not able to give consent. However, the health professional in charge can use "consent form 4" from the Department of Health to authorise the investigation or treatment. The health professional must be acting only in the best interest of the patient by consulting the relatives, carers, etc. and the Trust's legal department. A second opinion should also be obtained from a colleague. There are a number of serious situations that must be referred to the Court for its judgement. For example, if it was felt that a young woman (who lacks capacity) would be incapable of rearing a child, the parents might wish her to be sterilised. The courts view removal of fertility as extremely serious. Any decision of this kind would have to come from the Court and it would be illegal for the health professional to use the consent form 4. However, in case of an adult woman who lacks capacity to give consent or withholds consent to treatment, it is alright for the health professional to carry out hysterectomy for dealing with menorrhagia by using the consent form 4 if he/she is able to demonstrate that they are acting in the patient's best interest even though the procedure would render the woman infertile.

The Mental Capacity Act (2005) also extends "powers of attorney" to cover medical matters. "Power of attorney" implies that individuals give someone else the legal power to make decisions on their behalf. For example, old persons may realise that their brain is beginning to fail. The "power of attorney" may be given to their children, but it could also be given to a trusted friend or lawyer. An individual can arrange for someone to have "lasting power of attorney" in the event of his/her losing capacity.

Rights of the Unborn and Newborn Children

Unborn babies have little by way of legal rights. In particular, a mother cannot be made to put herself at risk or through unpleasant or unwanted procedures just for the benefit of the child. A pregnant woman cannot be made to have treatment, e.g. caesarean section, even if this means that her baby will die or come to serious harm. Once a child is born, it acquires the same rights as others.

Refusal of the parents to give consent for treatment of their newborn child is dealt with in the Department of Health document. The key feature is that clinicians and parents may not always agree on what is best for a child. Usually, if parents refuse treatment for their child then treatment will not go ahead. However, if the clinicians and their colleagues believe that it is crucial for the child to have the treatment in question, for example, if they think that the child would die or suffer serious permanent injury without the treatment then the courts can be asked to decide what would be best in the child's interests. Applications to court can be made at short notice if necessary. If the emergency is such that there is no time to apply to court, any doubts should be resolved in favour of the preservation of life.

Audit

Definition

Audit is the process of quality improvement of the healthcare services, thereby improving the overall quality of life. It aims at improving the patient care and outcome by assessing, evaluating and improving the care of the patients. This is achieved through the systematic review of care against set criteria. Based on the findings of the review, the changes are identified and implemented. Where indicated, the identified changes are implemented at an individual, team or service level. Further monitoring is implemented to confirm if these changes result in an improvement towards the delivery of healthcare services. Difference between audit and research has been described in **Table 1.2**.

Steps of an Audit Cycle

A typical audit cycle is described in **Figure 1.2** and comprises of the following steps:
1. *Initial needs assessment*: The audit cycle comprises of an initial needs assessment where the requirements of the department/section/individual are determined and the actual audit itself is determined.
2. *Identification of standards*: Then what is to be audited is decided upon; it is important to identify the standards against which the audit will be compared. These can be national standards or clinical guidelines determined by the national bodies or comparisons can even be made within the department.
3. *Data collection*: Once the standards are set, data collection is undertaken, with selection of retrospective or prospective data followed by data analysis.
4. *Recommendations*: The results can then be presented, compared to the standards and from this, recommendations for improvements/implementation of change are made.
5. *Re-audit*: Finally, to assess how effectively these recommendations have been implemented, a re-audit is suggested for some stage in the future.
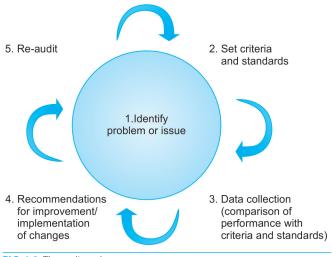
FIG. 1.2: The audit cycle

TABLE 1.2	Difference between audit and research	
Characteristic	**Research**	**Audit**
Definition	Discovers and defines the right thing to do	Determines whether the right thing is being done
Aims	Aims for the generalization of the findings	It is never possible to generalize the findings because each report deals with an individual situation
Special feature	Each project stands alone	Involves a cyclical series of reviews
Methodology	Collection of complex and unique data	Collection of routine data

Confidential Enquiry into Maternal Deaths

All maternal deaths in the UK and Ireland are investigated by the national programme, the Confidential Enquiry into Maternal Deaths (CEMD). These enquiries have been conducted in the UK since 1952. The committee directly responsible for the report was previously Confidential Enquiries into Maternal and Child Health (CEMACH). It was commissioned by National Institute of Clinical Excellence (NICE). CEMACH had been incorporated into Centre for Maternal and Child Enquiries (CMACE), which was the body primarily responsible for conducting these enquiries. Since June 2012, the CEMD has been carried out by the MBRRACE-UK (Mothers and Babies: Reducing Risk through Audits and Confidential Enquiries) collaboration. While the CMACE produced a report every triennium, analysing all maternal deaths from the previous 3 years divided into topic-specific chapters, the reports produced by the MBRRACE are now published on an annual basis, with each report focusing on a selection of chapters. Each MBRRACE-UK report now also contains "confidential enquiry into maternal morbidity" (CEMM) elaborating details of women who survived the problems related to pregnancy. The topic for 2014 CEMM was maternal sepsis.

Maternal death is defined by the International Classification of Diseases, Injuries and Causes of Death (ICD9/10)

as the death of a woman while pregnant or within 42 days of termination of pregnancy, from any cause related to or aggravated by pregnancy or its management, but not from accidental or incidental causes". It does not matter if the pregnancy lasted only for a few weeks, as in miscarriage. The idea is to limit the definition of maternal death both in time and causation to produce agreed international definitions. Pregnancy should have contributed to the death, i.e. she would not have died if she had not been pregnant. All maternal deaths are investigated in the confidential enquiries. Late deaths can be described as deaths occurring between 42 days and 1 year after pregnancy that are due to direct or indirect causes. Coincidental deaths are deaths from unrelated causes that happen to occur during pregnancy or the puerperium.

The latest CEMD was published in 2014 and focused on surveillance of all maternal deaths from the period 2010-12. The figures for the maternal mortality rate for the years 2006-08 and 2010-12 were 11 per 100,000 women and 10 per 100,000 women respectively. The reduction in mortality rates for the years 2010–2012 was related to reduction in deaths due to direct (obstetric causes). At the same time, there has been no significant change in the rate of indirect maternal deaths over the past 10 years. Actions are therefore urgently required to address deaths from indirect causes.

A "maternity" is any pregnancy going to 24 weeks or beyond or one resulting in a live birth before 24 weeks. The maternal mortality rate can be defined as the number of "direct" plus "indirect" deaths per 100,000 "maternities". Direct deaths are deaths resulting from obstetric complications of the pregnant state (pregnancy, labour and puerperium), from interventions, omissions, incorrect treatment or from a chain of events resulting from any of the above, e.g. bleeding, eclampsia, etc. Indirect deaths are deaths resulting from a previous existing disease, or disease that developed during pregnancy and which was not due to direct obstetric causes, but which was aggravated by the physiologic effects of pregnancy, e.g. cardiac disease.

According to the 2010-12 maternal mortality report published in 2014, two-thirds of the women died from indirect causes and almost three-quarters of all women who died had pre-existing medical and mental complications. Only one-third of the patients died due to direct complications of pregnancy such as bleeding. Almost a quarter of women who died had sepsis (severe infection). One in 11 of the women died from flu. The following key messages were given by this report:

❏ *Think sepsis*: The healthcare professional must keep the diagnosis of sepsis in mind, at an early stage, when an unwell pregnant patient or a recently pregnant woman presents. The key actions for diagnosis and management of sepsis are: early diagnosis, rapid antibiotics and review by senior doctors and midwives.

❏ *Influenza vaccine*: To avoid preventable deaths, the benefits of influenza vaccination (flu vaccine) to the pregnant women should be promoted and pregnant women at any stage of pregnancy should be offered vaccination.

❏ Women who have pre-existing medical and mental health problems require pre-pregnancy advice and multidisciplinary care comprising of the specialist and obstetric services.

Clinical Negligence Scheme for Trusts

Clinical Negligence Scheme for Trusts (CNST) is an option in risk management. Risks management can be defined as the identification, analysis, assessment, minimisation or elimination of unacceptable risks. The CNST has two main roles, first is running a scheme like an insurance scheme to help deal with clinical litigation claims, and secondly setting up standards to help improve the quality of services and risk management. They aim to improve clinical care and reduce the number of claims through an extensive risk management programme. If there are things that have gone wrong and generated claims, the CNST would want Trusts to be aware of them and to take steps to prevent their re-occurrence.

The CNST is like an insurance scheme for NHS hospital Trusts. The CNST covers the costs of clinical negligence claims. It is a voluntary scheme, but all NHS hospital Trusts are members. It is run by the NHSLA (NHS litigation authority). The NHSLA completely takes over the business of dealing with claims. Trusts pay an annual fee to CNST proportional to their risk of having claims against them. Its great attraction is that a paid-up member is fully indemnified against all clinical negligence claims. The scheme provides great reassurance, but at considerable cost. It is like a mutual scheme run by a group of clubs. All the contributors pay an agreed sum each year to cover the anticipated costs of all the claims that might be made against them. If a claim arises, the scheme deals with the cost and not the individual club. The clubs pay different fees according to the risk of them having a claim and the likely cost of settling it. The biggest burden on the CNST comes from maternity claims, so maternity services get particular attention.

The annual fee can be reduced by a Trust by implementing good risk management strategies. The implementation of good risk management is measured against criteria set by the CNST. There are three levels of risk management featured for the Trusts, each with different discounts. Level 1 is basic and includes the fundamentals such as someone in charge of risk management, a risk management committee, and provision of appropriate documentation, e.g. for protocols, etc. If the Trust meets these criteria, its contribution is reduced by 10%.

Levels 2 and 3 add more demanding measures to reduce risk and attract 20% and 30% reductions in contributions, respectively.

For level 2, the Trust has to show that it has implemented all the steps it did for level one, like its protocols. For level 3, it must actively monitor the implementation and deal with any problems. Since maternity services are a major problem for the Trusts, they have their own criteria and levels. There are five "standards" of CNST. The five "standards" are defined as organisation, clinical care, high-risk conditions, communication, and postnatal and neonatal care. Each "standard" has 10 "criteria" or subsections. For example, "Organisation" has 10 "criteria". Frequencies with which the CNST inspectors visit a Trust are described in **Table 1.3**.

If a Trust feels it is ready to move up a level, it can request an earlier inspection. On the other hand, a Trust that fails an assessment must be visited in the next financial year. The NHSLA employs a company called Det Norske Veritas to carry out the required assessments. The CNST only covers clinical claims. There are parallel schemes for non-clinical claims: the Liabilities to Third Parties Scheme (LTPS) and the Property Expenses Scheme (PES).

Impact on Obstetricians' Lives

Its impact is huge and wide-ranging. For example, it requires that a consultant should be present on the labour ward in cases of eclampsia, maternal collapse, caesarean section for major placenta praevia, post-partum haemorrhage (PPH) greater than 1.5 litres if the bleeding is continuing, a patient being taken back to the operating theatre, etc. Also, there should be an annual audit to ensure that the presence of consultants in the labour ward is in line with "safer childbirth". Similar annual audits are also required regarding the presence of other staff of the labour ward, from anaesthetists to labour ward assistants.

It lays down requirements for training in relation to antepartum haemorrhage, cord prolapse, early detection of severe illness, eclampsia, electronic foetal monitoring, post-operative care, PPH, maternal resuscitation, neonatal resuscitation, shoulder dystocia, vaginal breech delivery, etc.

Foetal Intrauterine Death

CEMACH defines this as death in utero from 24 weeks onwards. Intrauterine foetal death is a major disaster for the families. It is helpful for them to have supportive counselling from appropriately trained staff. The Department of Health now puts considerable emphasis on support for those bereaved and proper training for staff. There are also self-help groups, e.g. "Stillbirth and neonatal death support" (SANDS) and "The Child Bereavement Charity". Hospitals should have staff trained in bereavement counselling. It is important for the clinician to try to find the cause to be able to advise about future pregnancies. Nevertheless, most cases of foetal death remain unexplained, particularly in later gestations. Many cases are preceded by IUGR. Nowadays, more than 50% of cases are unexpected. Some likely causes for intrauterine foetal death are listed in **Table 1.4**.

TABLE 1.3	Frequencies with which the CNST inspectors visit a Trust
Level	**Assessment**
None	Every year
1	Every 2 years
2	At least once every 3 years
3	At least once every 3 years

TABLE 1.4	Causes for intrauterine foetal death

Foetal
- Anatomical: Cardiac, renal and other anomalies
- Chromosomal: Trisomy, etc.
- Infection, both viral and bacterial
  - Ascending infection following the rupture of membranes
  - Trans-placental spread of infection
- Foetal anaemia
  - Parvovirus infection
  - Rhesus incompatibility
  - Foeto-maternal transfusion
  - Alpha-thalassaemia
  - Bleeding from vasa praevia.

Maternal

Maternal characteristics
- Maternal age <20 and >40 years
- High BMI

Maternal disease
- IDDM and GDM
- Hypertension
- Renal disease
- SLE
- APS
- Thrombophilia
- Conditions causing high fever
- Major abdominal trauma
- Maternal death

Pregnancy conditions
- Obstetric cholestasis
- PIH and HELLP syndrome (haemolysis, elevated liver enzymes, low platelets)
- Prolonged pregnancy

Placental
- Placental abruption
- Failure of trophoblastic invasion of the spiral arteries
- Unexplained elevation of MSAFP in 2nd trimester

Multiple pregnancy
- Monochorionic twins
  - TTTS
  - Cord entanglement
- Triplets, quadruplets, etc.

Labour and delivery
- Precipitate labour
- Hypertonic contractions
- Uterine rupture

Contd...

Contd...
- Shoulder dystocia
- Breech with stuck head
- Bleeding from vasa praevia.

- Cord prolapse
- True knots
- Nuchal cord
  – Cord entanglement with monochorionic twins

Abbreviations: BMI, body mass index; SLE, systemic lupus erythematosus; IDDM, insulin-dependent diabetes mellitus; APS, anti-phospholipid syndrome; PIH, pregnancy-induced hypertension; MSAFP, maternal serum alpha foeto-protein; TTTS, twin-to-twin transfusion syndrome

Definition

All definitions of intrauterine foetal death require that the baby dies in the womb. According to the WHO, the baby must weigh at least 500 g for it to be classified as intrauterine death. As per CEMACH, intrauterine foetal death can be defined as foetal death in utero from 24 weeks onwards, with no specification of the weight, which fits with the accepted definition of stillbirth. Kindly refer to Chapter 10 for details related to stillbirths and perinatal mortality rate.

Diagnosis

The diagnosis of intrauterine foetal death is most often considered when the mother reports with the absence of foetal activity. Diagnosis is confirmed by the failure to find evidence of foetal heart activity on ultrasound scan.

Management

Modern management policy requires discussion with the mother regarding next step of management. Most patients opt for immediate induction, but some may wish to delay induction by a day or two so that they can come to terms with what has happened. Bromocriptine is the drug used for suppression of lactation in such women in the UK. It carries some risk and is no longer licensed in the USA for this purpose.

If the dead foetus is retained for more than a couple of weeks, disseminated intravascular coagulation may develop due to absorption of thromboplastins. In the rare situation of the woman who insists on awaiting a natural outcome, this would require the monitoring of coagulation parameters.

The great majority of women will wish to get on with the next pregnancy as soon as possible. They should be encouraged to get over the immediate grief. Counselling the bereaved parents is of prime importance. Some clinicians wait until the results of all the investigations have come to make sure there is no obvious recurring cause for intrauterine death.

Choose the Single Best Answer (SBA)

Q 1. Which of the following statement is true about evidence-based medicine?
A. Combines clinical expertise and external evidence
B. Does not involve health economic assessment
C. Is restricted to randomised placebo-controlled trials
D. Is used to cut down waiting lists
E. Tries to rely on subjective measurements of disease outcomes

Q 2. Which of the following statement is not true regarding the perinatal mortality rate?
A. It is usually expressed at the rate per thousand total births over one year
B. It is attributable to congenital malformations in 50% of cases
C. In England and Wales, it is higher in those whose mother was born in Pakistan than in those whose mother was born in the West Indies
D. The rate is marginally higher in boys
E. It is lowest in mothers aged between 20 and 29 years

Q 3. A surgical team presented their data demonstrating an increased rate of post-surgical wound infection following gastro-intestinal surgery compared with published standards from the Royal College of Surgeons. What is the most appropriate next step to be taken up by the team who is undertaking audit in this case?
A. Data analysis
B. Data collection
C. Identify standards
D. Implement change
E. Needs assessment

Q 4. A vascular team intends to compare their results for aortic aneurysm repair with national standards. What is the most appropriate next step to be taken up by the team who is undertaking audit in this case?
A. Data collection
B. Identify standards
C. Implement change
D. Needs assessment
E. Re-audit

1. A   2. B   3. D   4. A

Q 5. **A team wishes to audit their departmental results on the use of anticoagulation in patients with obstetric thromboembolic disease. What is the most appropriate next step to be taken up by the team who is undertaking audit in this case?**
- A. Data analysis
- B. Data collection
- C. Identify standards
- D. Implement change
- E. Needs assessment

Q 6. **An 82-year-old female who has dementia and is a resident in a nursing home is reviewed due to a vaginal discharge shown to be gonorrhoea. You suspect elder abuse and wish to contact the police. What is the most suitable form of consent, which should be obtained in this case?**
- A. Consent from carer
- B. Consent from court of law
- C. Consent from next of kin if possible
- D. No consent required
- E. Verbal consent required

Q 7. **Which of the following statement regarding "consent in clinical practice" is correct?**
- A. Parents of a mentally handicapped individual can give consent for her sterilisation.
- B. Parental consent is required for a girl of 14 to have termination of pregnancy.
- C. Jehovah's Witness parents can refuse blood transfusion for their children.
- D. A mother-to-be can refuse consent to Caesarean section, even if it means the child will die or sustain serious damage.
- E. An intoxicated woman who gets into bed with a man is, in effect, giving consent for sexual intercourse.

Q 8. **A surgical team assessing post-operative complications following surgery for vaginal hysterectomy has retrospectively collected data over the last 5 years on 133 patients. What is the most appropriate next step for the team undertaking audit in this case?**
- A. Data analysis
- B. Data collection
- C. Identify standards
- D. Implement change
- E. Needs assessment

Q 9. **At a recent directorate meeting, an obstetrician has been nominated to undertake the next clinical audit. What is the most appropriate next step for the team undertaking audit in this case?**
- A. Data analysis
- B. Data collection
- C. Needs assessment
- D. Identify standards
- E. Implement change

Q 10. **A team presented their audit of post-operative analgesia for pelvic surgery approximately 1 year ago from which a number of recommendations were made and changes implemented. What is the next best step which must be undertaken in this patient?**
- A. Data analysis
- B. Re-audit
- C. Data collection
- D. Needs assessment
- E. Identification of standards

Q 11. **You wish to perform karyotype analysis on a patient that you suspect has Turner's syndrome. What is the most suitable form of consent which must be obtained in this case?**
- A. Consent from carer
- B. Consent from court of law
- C. Consent from next of kin if possible
- D. Verbal consent required
- E. Written consent required

Q 12. **A 25-year-old female presents with postnatal depression and refuses treatment. What is the most suitable form of consent which must be obtained in this case?**
- A. Consent from carer
- B. Consent from court of law
- C. Consent from next of kin if possible
- D. Verbal consent required
- E. No consent required

Q 13. **Which of the following is true regarding foetal death in utero?**
- A. Is usually due to diabetes
- B. Can be prevented by proper obstetric management
- C. Induction of labour should be deferred until the cervix is favourable
- D. Conception should be discouraged for at least 6 months
- E. Danazol should be prescribed to suppress lactation

5. C   6. D   7. D   8. A   9. C   10. B   11. D   12. C   13. B

Anatomy

Blood Supply to the Brain

The arterial circulation to the brain mainly comprises of anterior cerebral circulation and posterior cerebral circulation. The anterior and posterior cerebral circulations form a part of an anastomotic ring, the circle of Willis **(Fig. 2.1)**, and are interconnected via anterior and posterior communicating arteries, present bilaterally. Circle of Willis is located at the base of the brain and helps in providing backup circulation to the brain in case of the occlusion of one of the vessels. However its exact structure is highly variable amongst individuals and often many people have inadequate arteries. These arteries may not be able to compensate in case of occlusion of a large vessel.
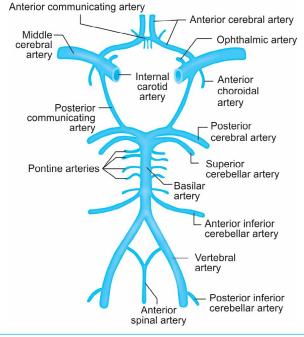
FIG. 2.1: Circle of Willis

Anterior Cerebral Circulation

This supplies blood to the anterior portion of the brain and is formed from the internal carotid arteries. The left and right internal carotid arteries arise from the common carotid arteries in the neck. The internal carotid artery branches into the anterior cerebral artery and continues as the middle cerebral artery. The two anterior cerebral arteries are connected by an anterior communicating artery.

Posterior Cerebral Circulation

This forms blood supply to the posterior portion of the brain, including the occipital lobes, cerebellum and the brain stem. It is supplied mainly by the vertebral arteries on the two sides. These are the branches of the subclavian arteries. The vertebral arteries fuse to form the basilar artery within the cranium. Before fusing, the vertebral arteries also give rise to the posterior inferior cerebellar vessels on the two sides. The basilar arteries supply the midbrain and the cerebellum and branch out to form the posterior cerebral artery. Other branches of the vertebral arteries help in supplying the midbrain and the cerebellum respectively.

Anatomy of Thorax

Diaphragm

The diaphragm is a large muscle that forms a partition between the cavities of the thorax and the abdomen. It also plays a crucial role in respiration.

Attachments of the Diaphragm

The diaphragm has a more or less circular origin from the thoracic outlet **(Fig. 2.2)**. The origin of the diaphragm can be divided into sternal, costal and vertebral parts.

❑ *The sternal part*: This consists of two slips, right and left, which arise from the back of the xiphoid process.

❑ *The costal part*: This consists of broad slips, one each from the inner surface of each of the lower six ribs (i.e. 7th–12th) and their costal cartilages. These slips interdigitate with those of an anterior muscle wall, the transversus abdominis.

❑ *The lumbar part*: This comprises of two crura, right and left. Each of the crura arises from the anterolateral aspects of the bodies of lumbar vertebrae and the lateral and medial arcuate ligaments.

The right crus is larger than the left. It arises from the bodies of vertebrae L1, L2, L3 and from the intervening intervertebral discs. On the other hand, the left crus arises from the vertebrae L1 and L2.

The medial margins of the two crura are joined to each other (at the level of the lower border of vertebra T12) to form the median arcuate ligament. The descending aorta passes from thorax to abdomen under cover of this ligament.

The lateral arcuate ligament represents a thickened band of the fascia over the quadratus lumborum, a muscle in the posterior wall of the abdomen. It is attached laterally to the 12th rib (about its middle) and medially to the transverse process of the first lumbar vertebra.

The medial arcuate ligament is a thickened band of the fascia covering the psoas major. It is attached laterally to the transverse process of the first lumbar vertebra. Medially, it becomes continuous with the lateral margin of the corresponding crus.

From its extensive origin, described previously, the muscular fibres of the diaphragm run upwards and converge to be inserted on the margins of a large, flat, central tendon, which is located just below the pericardium and heart. The central tendon is usually made up of three leaf-like parts (or folia) that are fused together.

The apex of anterior (triangular) leaf is apex is directed towards the xiphoid process and its base posteriorly, where it becomes continuous with two tongue-shaped posterior leaves. The apex of the anterior leaf receives the sternal fibres, while the sides of this leaf receive the anterior costal fibres. The posterior costal fibres reach the lateral sides of the posterior folia, while the fibres of the crura and those arising from the arcuate ligaments reach the apices and medial margins of the posterior folia. The upper convex part of the diaphragm is called its dome and it bulges considerably into the bony thorax.

Apertures in the Diaphragm

Many structures passing from thorax to abdomen (or vice versa) pass through apertures in (or around) the diaphragm. There are three large apertures, one each for the aorta, the oesophagus and the inferior vena cava, and several smaller ones.

The aortic aperture: It lies behind the median arcuate ligament, and in front of the disc between vertebrae T12 and L1. The aorta, therefore, passes behind the diaphragm rather than through it.

During inspiration, the pull of fibres of the muscle on the median arcuate ligament ensures that the aorta is not compressed. The aortic aperture also transmits the thoracic duct (which lies to the right side of the aorta) and sometimes the azygos and hemiazygos veins.

Central tendon of diaphragm

Diaphragm

Right crus

Vertebrae

Sternum

Vena cava passing through caval openig

Oesophagus passing through oesophageal hiatus

Aorta passing through aortic hiatus

12th (floating) ribs

Left crus

Left psoas major

Left quadratus lumborum

Diaphragm (inferior view)
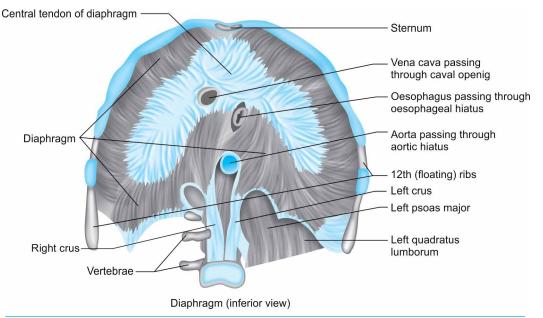
FIG. 2.2: Scheme showing attachments of the diaphragm

Aperture for the oesophagus: This is elliptical in shape. It is situated at the level of the 10th thoracic vertebra, usually an inch to the left of the midline. It is formed by splitting of the fibres of the right crus a little below their attachment to the central tendon. Since the oesophagus is surrounded by muscles, it is compressed during expiration. This prevents regurgitation of the contents of the stomach. Besides the oesophagus, the aperture also transmits the phreno-oesophageal ligament, the vagal trunks, the right and left gastric nerves which are continuations of the vagus nerves and the oesophageal branches of the left gastric artery, with their accompanying veins and lymphatics. The left gastric nerve is placed anteriorly and the right one posteriorly.

Aperture for the inferior vena cava: The inferior vena cava enters the thorax through the opening opposite the T8 vertebra just to the right of the midline.

Embryology

The diaphragm is partly derived from the cervical myotomes and the mesoderm. It is made up of structures arising from the septum transversum, pleuroperitoneal membranes, the dorsal mesentery and body wall. The septum transversum forms the central tendon.

Nerve Supply

The diaphragm receives a double nerve supply. The motor nerve supply arises from the right and left phrenic nerves. The sensory nerve supply to the peripheral part of the muscle is from the lower six intercostal nerves.

Blood Supply

The diaphragm is supplied by the right and left phrenic arteries, the intercostal arteries, and the musculophrenic branches of the internal thoracic arteries.

Venous drainage from the diaphragm occurs through the inferior vena cava and azygos vein on the right and the adrenal/renal and hemiazygos veins on the left.

The Pleura

The pleura comprises of two layers: the parietal and the visceral. The parietal layer is in contact with the chest wall, while the visceral layer is in close contact with the lungs. Apart from lining the surfaces of the lung, the visceral pleura dips into the fissures of the lungs, and lines the contiguous sides of the lobes. The parietal and visceral layers of pleura are in contact with each other being separated only by a potential space called the pleural cavity. The parietal pleura can be subdivided into the following parts:

❑ *The costovertebral pleura*: This lines the inner aspect of the ribs and intercostal spaces, part of the inner surface of the sternum, and the sides of thoracic vertebrae.

❑ *The diaphragmatic pleura*: This lines the upper surface of the diaphragm. However, not all parts of the diaphragm are covered by pleura.

❑ *The mediastinal pleura*: Mediastinal pleura is the portion of the parietal pleural membrane that lines the mediastinum. It is bounded by and is continuous with the anterior and posterior margins of the costovertebral pleura, the cervical pleura superiorly and the diaphragmatic pleura inferiorly. At the root of the lung on both sides, the mediastinal parietal pleura passes laterally along the structures of the root to merge with the visceral pleura. This region is the isthmus.

Despite the various divisions, pleura forms one continuous layer. The visceral pleura is relatively insensitive to pain. However, the parietal pleura is highly sensitive to pain. The diaphragmatic pleura is supplied by the phrenic nerve over the domes and the intercostal nerves over the periphery. The blood supply of the visceral pleura is derived from the bronchial and pulmonary arteries.

Nerves of the Thorax

Phrenic Nerve

The phrenic nerves are amongst the most important nerves in the body as they are the only motor supply to the diaphragm. Each nerve (right or left) arises from the (anterior primary rami of) spinal nerves C3, C4 and C5, with the contribution from C4 being the greatest **(Fig. 2.3)**. The nerve descends vertically through the lower part of the neck and then through the thorax to reach the diaphragm. Some terminal branches enter the abdomen. In the neck, the phrenic nerve descends vertically across the scalenus anterior muscle. Crossing the medial (or lower) border
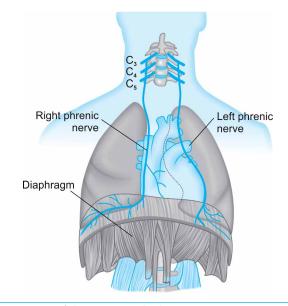
FIG. 2.3: Course of phrenic nerve in the thorax

of the muscle, it crosses in front of the first part of the subclavian artery. On the right side, however, the nerve is usually separated from the artery by a part of the scalenus anterior. Throughout its course in the neck, the nerve lies deep to the sternocleidomastoid muscle. On entering the thorax, the nerve passes medially crossing in front of the internal thoracic artery and comes into relationship with structures in the mediastinum. Subsequent relations are different on the right and left sides.

The left phrenic nerve passes inferiorly down the neck to the lateral border of scalenus anterior. Then it passes medially across the border of scalenus anterior parallel to the internal jugular vein which lies inferomedially. At this point it is deep to the prevertebral fascia, the transverse cervical artery and the suprascapular artery. It descends between the left subclavian and the left common carotid arteries and crosses the left surface of the arch of the aorta. It then courses along the pericardium, superficial to the left atrium and left ventricle, piercing the diaphragm just to the left of the pericardium. It carries sensory fibres from the pleura, pericardium and a small part of the peritoneum.

Relations of the Left Phrenic Nerve

The relations of the left phrenic nerve are as follows:

Above the arch of the aorta: Above the arch of aorta, the nerve lies in the interval between the left common carotid and left subclavian arteries. It, at first lies posterior and lateral to the vagus nerve, but crosses the latter superficially and comes to lie in front and medial to it.

The nerve then crosses the aortic arch lying on its anterolateral side. Here, the nerve crosses superficial to the left of superior intercostal vein.

Below the arch of aorta: Below the arch of the aorta, the phrenic nerve crosses in front of the structures comprising the root of the left lung and then descends across the heart (left ventricle) lying between the parietal pericardium and the mediastinal pleura.

Relations of the Right Phrenic Nerve

The relations of the right phrenic nerve are as follows:

After crossing the internal thoracic artery, the nerve reaches the right brachiocephalic vein. It runs downwards lateral to this vein and at its lower end the nerve passes onto the lateral side of the superior vena cava. Leaving the vena cava the nerve descends over the right side of the heart (right atrium) lying between the parietal pericardium and the mediastinal pleura. Just above the diaphragm, the nerve lies lateral to the inferior vena cava.

The Vagus Nerve

The vagus nerve arises from the brain (medulla oblongata). It descends vertically in the neck in close relationship to the internal or common carotid artery and the internal jugular vein. In the lower part of the neck, the nerve crosses anterior to the first part of the subclavian artery and enters the thorax.

Course and Relations of Vagus Nerve in the Thorax

Course of right vagus: In the superior mediastinum, the right vagus nerve lies on the right side of the trachea. Here it is posteromedial first, to the right brachiocephalic vein and then to the superior vena cava. The nerve passes deep to the azygos vein to reach the posterior side of the root of the right lung.

Course of left vagus: The left vagus nerve descends between the left common carotid and left subclavian arteries in the superior mediastinum. It passes behind the left brachiocephalic vein and then crosses the left side of the arch of the aorta to reach the posterior aspect of the root of the left lung. The nerve is related laterally to the left lung and pleura. Above the arch of the aorta the vagus is crossed by the left phrenic nerve. Over the arch of the aorta, it is crossed by the left superior intercostal vein.

Having reached the root of the lung, each vagus nerve (right or left) divides into a number of branches and therefore ceases to exist as distinct trunks. Recurrent laryngeal nerve is an important branch given by the vagus nerve in the thorax, which provides the motor supply to most of the intrinsic muscles of the larynx. The nerves also provide the sensory supply to the mucous membrane of the lower half of the larynx.

Anatomy of the Female Breast

The female breast/mammary gland is situated within the subcutaneous tissues and extends from the 2nd to the 6th rib in the midclavicular line, overlying the fascia over pectoralis major and serratus anterior. Beneath the breast tissue, there is a condensation of superficial fascia, which acts as a posterior capsule for the breast. The gland is normally mobile over this fascia. The parenchyma comprises of about 15–20 lobes, each of which is drained by a lactiferous duct. The various lactiferous ducts open on the nipple. The breast stroma comprises of adipose and fibrous tissue. The breast nipple is surrounded by an areola of pigmented skin, which darkens during pregnancy and then remains so. The areola contains accessory mammary glands, sweat glands, and sebaceous glands. These form Montgomery tubercles during pregnancy and lubricate the nipple during lactation. The upper outer quadrant of the breast containing a large amount of glandular tissue is the most common site of breast carcinomas.

Embryology

At the end of the first month of embryonic development, the mammary gland begins to develop as two vertical ectodermal thickenings in form of solid buds into the

underlying mesenchyme. These thickenings extend from the axilla to the inguinal region. The ventral part of each forms the nipple. The mammary glands develop from the nipples during foetal life. At the time of puberty, in the females, the breasts grow and there occurs the development of ducts and lobules. However, true secretory alveoli do not develop until pregnancy.

Blood Supply (Figs 2.4A and B)

Arterial Supply

The arteries supplying the breast are derived from axillary artery (via branches such as superior thoracic artery, pectoral branches of thoracoacromial artery, lateral thoracic artery, etc.), internal thoracic artery and intercostal arteries. Internal thoracic artery and its perforating branches supply medial part of the breast. Lateral thoracic artery supplies lateral part of the breast. A profound part is also supplied by intercostal arteries and their branches.

Venous Drainage

The corresponding veins (i.e. the axillary vein, internal thoracic vein and the intercostal veins) accompany the arteries supplying the breast. The veins draining the breast tissues form an anastomotic circle around the base of the nipple, called Haller circulus venosus. From this, large branches transmit blood from medial part of the breast into internal thoracic veins and from the lateral part of the breast into the lateral thoracic vein and intercostal veins. These eventually drain into the superior vena cava. Connections between the intercostal veins and the vertebral plexus result in metastatic deposits to bones and the nervous system in cases of breast carcinoma.

Lymphatic Drainage of the Breasts
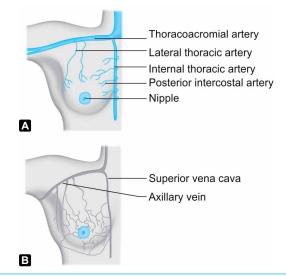
The lymph vessels of the breast are situated into two layers (superficial and deep layers), making subareolar plexus (superficial and deep) that are interconnected. Superficial lymph vessels transmit the lymph fluid into the axillary lymph nodes. Lymphatic drainage of various quadrants of breast is described in **Table 2.1** and **Figure 2.5**.

The majority of lymph drains into the subareolar plexus and then into the pectoral group of axillary lymph nodes. 75% of lymph drains to this group of lymph nodes. Lymph from the medial aspect of the breasts is most likely to drain through the intercostal spaces into the parasternal group of lymph nodes, while that from the lateral breasts is likely to drain into the axillary and infraclavicular nodes. Free communication exists between nodes below and above the clavicle and between the axillary and cervical group of lymph nodes. Internal mammary nodes communicate with the lymphatics across the midline. Therefore, cancer from one side can spread to the opposite breast.

The axillary nodes can be arbitrarily divided into five groups:
1. *The lateral nodes*: These lie behind the axillary vein and drain the upper limb.
2. *The pectoral nodes*: These lie at the inferior border of the pectoralis minor and drain most of the breast tissue.

TABLE 2.1	Lymphatic drainage of various quadrants of the breast
Quadrant	**Group of nodes to which drainage occurs**
Superolateral quadrant	Anterior, posterior axillary group of lymph nodes and supraclavicular group of lymph nodes
Superomedial quadrant	Internal mammary group, supraclavicular nodes
Inferomedial quadrant	Internal mammary group, supradiaphragmatic nodes
Inferolateral quadrant	Posterior intercostal nodes, subdiaphragmatic group

FIGS 2.4A AND B: Blood supply in the region of breast: (A) Arterial supply; (B) Venous supply
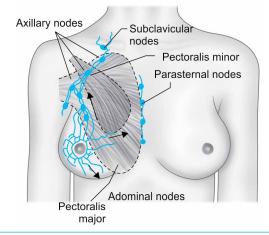
FIG. 2.5: Lymphatic drainage of the breast

3. *The posterior or subscapular nodes*: These are present in the posterior axillary fold and primarily drain the posterior shoulder.
4. *The central nodes*: These are present near the base of the axilla and receive lymph from the previously mentioned three groups. The central nodes belonging to the axillary group of lymph nodes form the group, which is most likely to be palpable against the lateral thoracic wall.
5. *The apical nodes*: These lie medial to the axillary vein and superior to the pectoralis minor. The apical nodes receive the lymph from all the other groups and sometimes directly from the breast. They eventually drain into the deep cervical group of lymph nodes.

Therefore, at the time of breast examination, it is important to carefully examine the axilla and to examine the supra- and subclavicular lymph nodes. The clavicular group, however, is not part of the axillary group of lymph nodes.

Nowadays, a simple nomenclature of classification of axillary nodes is adopted based on the relation of the nodes to the pectoralis minor muscle. Those lying below the muscle are the low nodes (Level 1); those lying behind the muscle are the middle group (Level 2). The nodes between the upper border of pectoralis minor and the lower border of the clavicle are the upper or the apical group (Level 3).

Nerve Supply

Nerve supply to the breasts is derived from the branches of 4th–6th intercostal nerves. They carry the sensory and sympathetic efferent fibres. Supply to the nipples is from T4. This forms an extensive plexus of nerves within the nipple, its sensory fibres terminating close to the epithelium in form of free endings such as Meissner's corpuscles and Merkel disc endings.

Anatomy of the Abdominal Wall

The part of the abdominal wall extending all the way from the midline to the lateral edge of the quadratus lumborum is referred to as the anterior abdominal wall. Therefore, the anterior abdominal wall is not only confined to the anterior aspect of the abdomen, but also includes the lateral sides. Schematic transverse section through the abdomen showing various muscles is described in **Figure 2.6**.

Muscles of the Anterior Abdominal Wall

The musculature of the abdominal wall is composed of two muscle groups. One group, comprising of the flat muscles, consists of three muscles: (1) the external oblique, (2) the
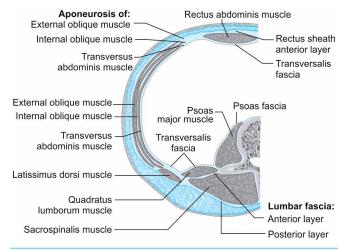
FIG. 2.6: Schematic transverse section through the abdominal wall

internal oblique and (3) the transversus abdominis. The second group is composed of two muscles that run vertically and comprise of the muscles, rectus abdominis and the pyramidalis. **Figures 2.7** and **2.8** illustrate the various muscles of the anterior abdominal wall.

External Oblique Muscle

The external oblique muscle is the largest and most superficial of the flat muscles of the anterolateral abdominal wall. The fibres of the external oblique muscle run forwards and downwards.

Origin: It arises from the external surface of the lower 8 ribs (ribs 5th–12th).
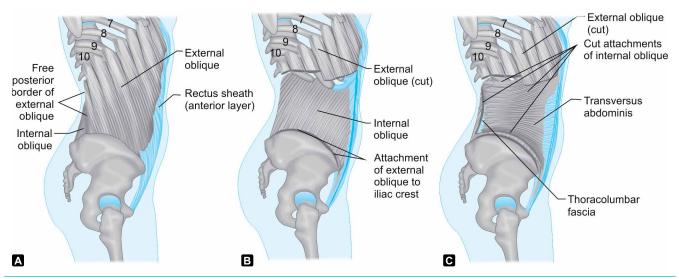
Insertion: The external oblique muscle courses diagonally anteriorly and inferiorly to get inserted upon the pubic tubercle, anterior half of iliac crests, and linea alba.

Internal Oblique Muscle

This muscle is intermediate amongst the three muscles of anterior abdominal wall. The fibres of the internal oblique muscle run forwards and upwards.

Origin: The internal oblique muscle arises from the thoracolumbar fascia, anterior two-thirds of the iliac crest, and the connective tissue deep to the lateral third of inguinal ligament.

Insertion: This muscle courses at a right angle to the fibres of the external oblique muscle and gets inserted on the inferior borders of 10th–12th ribs, linea alba and pecten pubis via the conjoint tendon. The aponeurosis of the internal oblique splits at the lateral edge of the rectus muscle into an anterior and posterior lamina to envelope the rectus abdominis muscle. The anterior layer blends with the aponeurosis of the external oblique. Posterior to the rectus muscle, this

FIGS 2.7A TO C: Flat muscles of the anterior abdominal wall: (A) Layer 1: External oblique muscle; (B) Layer 2: Internal oblique muscle; (C) Layer 3: Transversus abdominis muscle
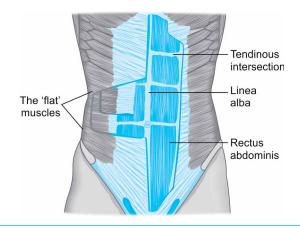
FIG. 2.8: Rectus abdominis muscle

aponeurosis blends with the aponeurosis of the transversus abdominis to form a portion of the posterior rectus sheath.

In most areas, the fibres of this muscle are perpendicular to the fibres of the external oblique, but in the lower abdomen, their fibres arch somewhat more caudally, and run in a direction similar to those of the external oblique.

Transversus Abdominis Muscle

The innermost of the flat muscles is the transversus abdominis and its fibres run more or less transversely.

Origin: This muscle arises from the internal surface of 7th–12th costal cartilages, thoracolumbar fascia, iliac crest, and connective tissue deep to the lateral third of the inguinal ligament.

Insertion: Coursing transversely to the midline, the upper three-fourths of the transversus aponeurosis lies behind the rectus muscle. The lower one-fourth of the aponeurosis

passes in front of the rectus muscle. The fibres of transversus abdominis gets inserted into the linea alba along with the aponeurosis of internal oblique, and into the pubic crest and pecten pubis via the conjoint tendon.

Between the muscle fibres of internal oblique and transversus abdominis, there is a neurovascular plane of the anterolateral abdominal wall, which contains the nerves and arteries supplying the anterolateral abdominal wall.

Rectus Abdominis Muscle

Rectus abdominis muscle belongs to the group of muscle, which runs vertically. It is the principal muscle of the vertical group. There are three tendinous inscriptions within each rectus abdominis muscle. These fibrous interruptions within the muscle help in firmly attaching it to the rectus sheath. This produces a six-pack appearance in athletic individuals.

These fibrous interruptions are usually confined to the region above the umbilicus, but sometimes can also be found below the umbilicus. When found below the umbilicus, the rectus sheath is attached firmly to the rectus muscle at the region of inscription. This may cause difficulty at the time of muscle separation during Pfannenstiel incision.

Origin: This muscle takes its origin from the pubic symphysis and the pubic crest.

Insertion: After taking their origin, the rectus muscle fibres run vertically to get inserted into the xiphoid process and the fifth, sixth, and seventh costal cartilages. The rectus muscle is surrounded by a sheath, comprising of the aponeuroses of the oblique muscles and the transversus abdominis. The rectus sheath has been described in details later in this chapter.

Pyramidalis

This muscle is absent in approximately 20% of the population and lies anterior to the inferior part of rectus abdominis. This muscle marks the midline and assists in the identification of the medial borders of the rectus muscle.

Origin: A small, vestigial, triangular-shaped muscle, the pyramidalis, arises from the pubic symphysis.

Insertion: It inserts on the anterior surface of the pubis and the anterior pubic ligament. It ends in the linea alba which is especially thickened for a variable distance superior to the pubic symphysis. The pointed insertion of the pyramidalis muscles into the linea alba can be used for locating the midline.

Blood Supply to the Anterior Abdominal Wall

The primary blood supply to the abdominal wall is from the superficial and deep blood vessels. The main blood vessels supplying the anterolateral abdominal wall are as follows:
❑ Superior epigastric vessels and the branches of musculophrenic artery
❑ Inferior epigastric and deep circumflex iliac arteries
❑ Superficial circumflex iliac and superficial epigastric arteries
❑ Posterior intercostal vessels of the 11th intercostal space and the anterior branches of the subcostal vessels.

The blood supply of the anterior abdominal wall is demonstrated in **Figure 2.9**. The superficial blood vessels originate from the femoral artery and include the superficial epigastric, the superficial circumflex, and the superficial external pudendal arteries. The deep vessels, on the other hand, originate from the external iliac and the internal thoracic artery. These include the inferior epigastric artery, the deep circumflex artery and the superior epigastric artery, which is the terminal branch of the internal thoracic artery. The internal thoracic artery also gives rise to the musculophrenic artery, which anastomoses with the deep circumflex artery. Anastomosis between the various vessels of abdominal wall helps in ensuring an excellent blood supply to all areas of the abdominal wall. The individual blood vessels would now be described.

Superior Epigastric Vessel

Superior epigastric vessel is the direct continuation of the internal thoracic artery. It enters the rectus sheath superiorly through its posterior layer and supplies the superior part of the rectus abdominis and anastomoses with the inferior epigastric artery in the umbilical region.

Inferior Epigastric Vessel

Inferior epigastric vessel arises from the external iliac artery just superior to the inguinal ligaments. It runs superiorly in
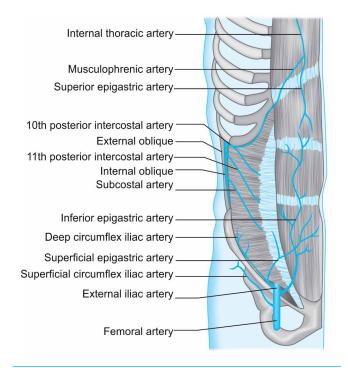
FIG. 2.9: Arteries of the anterolateral abdominal wall

the transversalis fascia to enter the rectus sheath below the arcuate line. It enters the lower part of the rectus abdominis and anastomoses with the superior epigastric artery.

Superficial Circumflex Iliac Artery

Superficial circumflex iliac artery is the branch of femoral artery, which runs in the subcutaneous tissue towards the umbilicus. It supplies the superficial abdominal wall of the inguinal region and the adjacent anterior thigh region.

Superficial Epigastric Artery

Superficial epigastric artery begins as a single artery that branches extensively and runs in the subcutaneous tissues towards the umbilicus. It supplies superficial abdominal wall of pubic and inferior umbilical regions.

Musculophrenic Artery

The musculophrenic artery originates from the internal thoracic vessels and descends along the costal margin. It supplies the superficial and deep abdominal walls of the epigastric and upper umbilical regions.

The 10th and 11th posterior intercostal arteries and subcostal arteries originate from aorta. They continue beyond the ribs to descend in the anterior abdominal wall between internal oblique and transversus abdominis muscles. They supply superficial and deep abdominal wall of lateral lumbar or flank region.

Lymphatic Drainage of the Anterior Abdominal Wall

Lymphatics in the region above the umbilicus drain into the axillary lymph nodes. Lymphatics in the region below the umbilicus drain into the superficial inguinal nodes. Superficial inguinal lymph nodes also receive lymph drainage from lower abdominal wall, buttocks, scrotum, penis, labium majus, and the lower parts of the vagina and anal canal. The efferent lymphatic vessels from the superficial inguinal group of lymph nodes primarily drain into the external iliac nodes and, ultimately, the lumbar (aortic) nodes, eventually reaching the cisterna chyli and the thoracic duct.

On the other hand, the deep inguinal lymph nodes receive most of the drainage from the lower limbs. Efferent lymphatic vessels from the deep inguinal group of lymph node, similar to the superficial group, drain into the external iliac, common iliac and lumbar group of lymph nodes, ultimately reaching the cisterna chyli and thoracic duct.

Nerve Supply of the Anterior Abdominal Wall

The major nerves supplying the anterior abdominal wall include the thoracoabdominal nerves, subcostal nerve, the ilioinguinal nerves, the iliohypogastric nerves and the lateral cutaneous branches of the thoracic spinal nerves **(Fig. 2.10)**. These nerves can be described as given below.

Thoracoabdominal Nerve

These are the distal, abdominal part of the anterior rami of the inferior five thoracic spinal nerves (T7–T11). The thoracoabdominal nerves travel caudad between the transversus abdominis and the internal oblique muscles.

These nerves innervate the flat muscles of the abdominal wall and the rectus muscle.

Iliohypogastric and Ilioinguinal Nerves

Both of these nerves are the terminal branches of the anterior ramus of the spinal nerve L1, with the iliohypogastric nerve being the superior terminal branch and the ilioinguinal nerve being the inferior one. Iliohypogastric nerve supplies the skin overlying the iliac crest, upper inguinal and hypogastric regions, internal oblique and transversus abdominis muscles. Ilioinguinal nerve, on the other hand, supplies the skin of lower inguinal region, mons pubis, anterior scrotum or labium majus and the adjacent medial thigh as well as inferior-most regions of the internal oblique and transversus abdominis.

Damage to these nerves may result in sensory changes in the mons pubis and the labia majora.
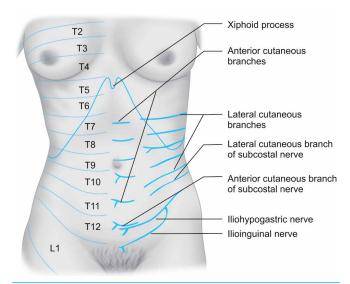
FIG. 2.10: Nerves of the anterior abdominal wall

Lateral Cutaneous Branches

These branches emerge from the musculature of the anterolateral abdominal wall and originate from the anterior rami of spinal nerves T7–T9. It then enters the subcutaneous tissues along the anterior axillary line in the form of anterior and posterior divisions.

Subcostal Nerve

It originates from the anterior ramus of spinal nerve T12. It passes between the second and third layers of the abdominal muscle and then traverses the inguinal canal.

Anterior Abdominal Cutaneous Branches of Thoracoabdominal Nerves

These supply the following areas:

Skin superior to the umbilicus: Supplied by T7–T9

Skin around the umbilicus: Supplied by T10

Skin below the umbilicus: Supplied by T11, and the cutaneous branches of the subcostal, iliohypogastric and ilioinguinal nerves.

Rectus Sheath

The rectus sheath is formed by the conjoined aponeuroses of the flat abdominal muscles. It is formed by the decussation and interweaving of the aponeurosis of these muscles. The aponeurosis of external oblique muscle contributes to the formation of the anterior wall of the sheath throughout its length. A concentric line, "arcuate line" lies midway between the umbilicus and pubic symphysis and demarcates the transition between the aponeurotic posterior wall of the sheath covering the superior three-fourths of the rectus and the transversalis fascia covering the inferior quarter.

Throughout the length of the sheath, the fibres of the anterior and posterior layer of the sheath interlace in the anterior median line to form the complex linea alba. The ventral rami of the lower seven thoracic nerves and the anastomosis between the superior and inferior epigastric vessels occur within the rectus sheath. When the pyramidalis muscle is present, it lies within the sheath, anterior to the rectus abdominis.

The composition of the rectus sheath above and below the arcuate line is described in **Figures 2.11A and B**.

Above the Arcuate Line

The superior two-thirds of the internal oblique apo-neurosis splits into two layers at the lateral border of rectus abdominis, with one lamina passing anterior to the muscle and the other posterior to it. The anterior lamina joins the aponeurosis of external oblique muscle to form the anterior layer of the rectus sheath. The posterior lamina of the internal oblique joins the aponeurosis of transversus abdominis to form the posterior layer of the rectus sheath.

Below the Arcuate Line

Below the arcuate line, the aponeuroses of the three flat muscles pass anterior to the rectus abdominis to form the anterior layer of the rectus sheath, leaving only the relatively thin transversalis fascia to cover the rectus abdominis muscle posteriorly.

Superior to the Costal Margin

The posterior layer of the rectus sheath is also deficient superior to the costal margin because the transversus abdominis is continued superiorly as the transversus thoracis, which lies internal to the costal cartilages, and the internal oblique attaches to the costal margin. Hence, superior to the costal margin, rectus abdominis muscle lies directly on the thoracic wall.
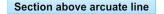
Importance for the Surgeon
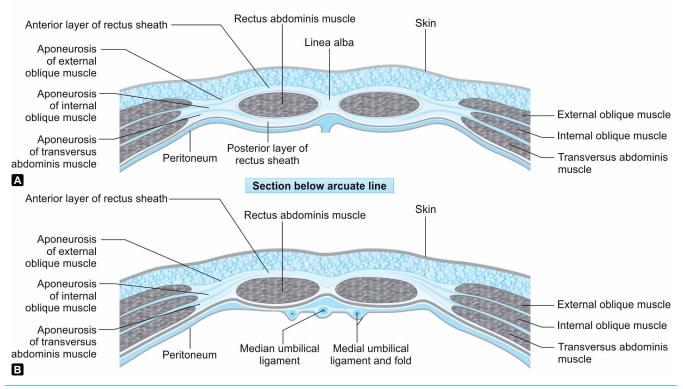
There are several specialised aspects of the rectus sheath that are important to the surgeon. In forming the rectus sheath, the conjoined aponeuroses of the individual flank muscles can be separated lateral to the rectus muscles, but as they reach the midline, they fuse and lose their separate directions. As a result of this midline fusion, these layers are usually incised together in the midline while giving a transverse fascial incision.

Posterior Abdominal Wall

The posterior abdominal wall is made up of the following structures:
❑ Lumbar vertebrae in the median plane
❑ Psoas major muscle lying along each side of the vertebral bodies
❑ Quadratus lumborum muscles which are present more laterally.

FIGS 2.11A AND B: The rectus sheath

Abdominal Cavity and its Contents

Abdominal Aorta

The thoracic aorta pierces the diaphragm at T12 to become the abdominal aorta. It ends by dividing into two common iliac arteries at the level of L4. Note that the bifurcation (union) of the inferior vena cava occurs at the level of L5 and therefore lies below the level of bifurcation of the aorta. Various branches of abdominal aorta are listed in **Table 2.2** and **Figure 2.12**. The ventral branches of the aorta include the coeliac artery and superior and inferior mesenteric arteries. Several anastomoses occur between the branches of these ventral vessels. These are as follows:

❑ Anastomosis between the branches of left gastric artery with the oesophageal branches (directly arising from the aorta) around the lower oesophagus

❑ Anastomosis between left gastric artery with the right gastric artery (branch of hepatic artery)
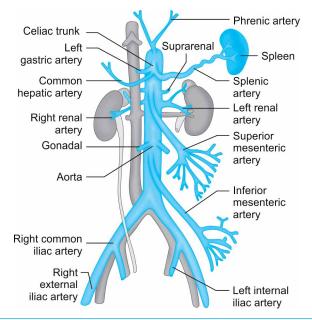
FIG. 2.12: Branches of abdominal aorta

TABLE 2.2	Branches of abdominal aorta				
Name of the branch	**Level of vertebra for origin**	**Paired or not**	**Anterior or posterior**	**Branches**	**Description**
Inferior phrenic a.	T12	Yes	Posterior		Originates just below the diaphragm, supplying it from below

Coeliac axis	Upper L1	No	Anterior	1. Left gastric a.
				2. Splenic a.
				  a. Short gastric arteries (6)
				  b. Splenic arteries (6)
				  c. Left gastroepiploic a.
				3. Common hepatic a.
				  a. Cystic a.
				  b. Right gastric a.
				  c. Gastroduodenal a.
				    i. Right gastroepiploic a.
				    ii. Superior pancreaticoduodenal a.
				  d. Right hepatic a.
				  e. Left hepatic a.
					Coeliac axis is the artery of the foregut and arises from the aorta between the right and the left crura of the diaphragm. It is 1 cm long and is surrounded by the coeliac plexus of nerves.

Superior mesenteric a.	Lower L1	No	Anterior	1. Jejunal and ileal arteries
				2. Inferior pancreaticoduodenal a.
				3. Middle colic a.
				4. Right colic a.
				5. Ileocolic a.
				  a. Anterior caecal a.
				  b. Posterior caecal
				  c. Appendicular a.
				  d. Ileal a.
				  e. Colic a.
					This is a large anterior branch arising just below coeliac trunk. It supplies the gastro-intestinal tract from the middle of the second part of the duodenum as far as the distal one-third of the transverse colon. In other word, this artery supplies the parts of the gut, which are derived from the midgut.

Middle suprarenal a.	L1	Yes	Posterior		To the adrenal glands
Renal a.	In between L1 and L2	Yes	Posterior		Large arteries, each arising from the side of the aorta and divide into several branches which supply the corresponding segment of each kidney

Contd...

Contd...

Name of the branch	Level of vertebra for origin	Paired or not	Anterior or posterior	Branches	Description
Gonadal a.	L2	Yes	Anterior		Ovarian artery in women and testicular artery in men
Lumbar a.	L1–L4	Yes	Posterior		Four on each side that supply the abdominal wall and the spinal cord
Inferior mesenteric a.	L3	No	Anterior	1. Left colic a. 2. Sigmoid arteries (2 or 3) 3. Superior rectal a.	The superior rectal artery is the continuation of the inferior mesenteric artery and descends in the base of the pelvic mesocolon. It supplies parts of the gut, which are derived from the hindgut.
Median sacral a.	L4	No	Posterior		This artery arises from the middle of the aorta at its lowest part
Common iliac a.	L4	Yes	Posterior	1. External iliac a. 2. Internal iliac a.	This is the end of abdominal aorta which bifurcates to supply blood to the lower limbs and the pelvis

Abbreviation: a., artery

- Anastomosis between anterior and posterior superior pancreaticoduodenal arteries (branches of coeliac trunk) with the inferior pancreaticoduodenal (superior mesenteric branch) around the head of the pancreas and second part of the duodenum
- The marginal artery anastomosis between the middle colic and the left colic
- Anastomosis between the superior rectal artery (branch of inferior mesenteric) with the middle rectal artery (branch of internal iliac) and/or the inferior rectal (branch of internal pudendal artery which arises from the internal iliac).

Peritoneal Reflections

The abdominal cavity and most of the viscera within it are lined by a serous membrane called the peritoneum. Since the peritoneum is a closed sac that is invaginated by viscera, it has a parietal layer lining the abdominal wall; and a visceral layer, which is closely applied to the viscera.

The pericardium, pleura, and peritoneum have a similar arrangement, having parietal and visceral layers, with a cavity between. The peritoneal cavity contains a thin film of fluid which allows free movement of the viscera against the abdominal wall and against each other.

Basic Arrangement of the Peritoneum Relative to the Viscera

Some abdominal organs are in contact with the posterior abdominal wall, and are only partly lined by peritoneum. Such viscera are described as being retroperitoneal and have limited mobility (e.g. bare area of liver, duodenum, ascending colon, descending colon, rectum, kidneys and ureters, adrenal glands, and major vessels, such as abdominal aorta, inferior vena cava and iliac vessels). In contrast to such viscera, there are other organs which are suspended from the abdominal wall by double-layered folds of peritoneum passing from the abdominal wall to the viscera, e.g. small intestine. The fold of peritoneum by which the small intestine (jejunum and ileum) is attached to the posterior abdominal wall is known as the mesentery. Some other similar folds are mesocolon (attached to the colon), and mesovarium (attached to the ovaries), etc. Blood vessels and nerves reach the concerned viscera through these folds. The peritoneal cavity is completely closed in the male. On the other hand, in the female, it communicates via the tubal ostia. Some peritoneal reflections are known as ligaments or folds, e.g. gastrohepatic ligament or rectouterine fold respectively. A broad peritoneal sheet or peritoneal reflection is termed as omentum. These include the lesser omentum and the greater omentum.

The abdominal cavity also comprises of a general peritoneal cavity (or the greater sac) and the omental bursa (or the lesser sac) which lies behind the stomach and its peritoneal attachments. The lesser sac communicates with the greater sac by the so-called epiploic foramen, which can be found by running a finger along the gall bladder to the free edge of the lesser omentum. The longitudinal section through the abdominal cavity illustrating various peritoneal reflections is shown in **Figure 2.13**.

Lesser Omentum

Lesser omentum is a broad peritoneal reflection, which connects the stomach to the liver.

Greater Omentum

The greater omentum is a double fold of peritoneum that connects the stomach to the posterior abdominal wall. It hangs down from the stomach and merges with the mesentery attached to the anterior aspect of transverse colon. Though it is fused behind with the transverse colon and mesocolon, it is separate from them.
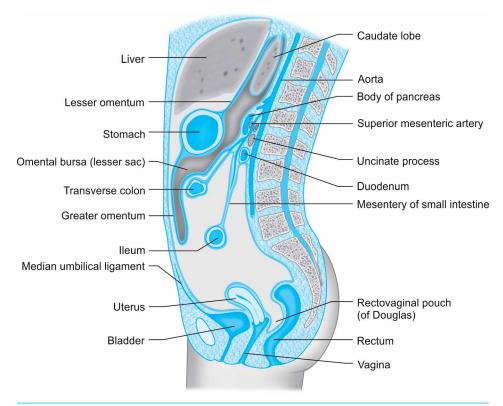
FIG. 2.13: Section through the abdominal cavity illustrating peritoneal reflections (the grey area shows the lesser sac, while the white area shows the greater sac)

Greater Sac

This extends from the diaphragm to the pelvic floor. It is the cavity in the abdomen that is inside the peritoneum but lies outside the lesser sac. It is further divided into two compartments by the transverse mesocolon:
1. *Supracolic compartment*: This lies above the transverse mesocolon and contains stomach, liver and spleen
2. *Infracolic compartment*: This lies below the transverse mesocolon and contains the small intestine, ascending and descending colon.

Lesser Sac

Relations

Anteriorly: The lesser omentum (superiorly), posterior surface of the stomach (centrally) and the anterior two layers of the greater omentum (inferiorly).

Posteriorly: (1) The peritoneum that covers the diaphragm, pancreas, left kidney and suprarenal gland, and duodenum and (2) the posterior two layers of the greater omentum which fuse with transverse mesocolon.

Superiorly: Gastrosplenic part of greater omentum (on the left side); caudate lobe of liver (on the right side).

Laterally: Limited on the left side by lienorenal ligament; on the right side opens into the greater sac through the epiploic foramen.

Epiploic Foramen (Foramen of Winslow)
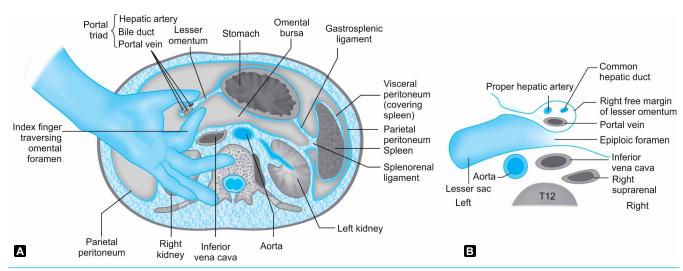
As previously described, the epiploic foramen is the passage of communication, or foramen, or an opening from the greater into the lesser sac. It lies immediately posterior to the free, right edge of the lesser omentum. A finger in the opening and a thumb in front of the omentum would encircle the bile duct (at the right), the hepatic artery (at the left), and the portal vein posterior and between them) **(Figs 2.14A and B)**. Boundaries of the epiploic foramen are described next.

Anterior: Free border of the lesser omentum, with the common bile duct, hepatic artery, and portal vein between its two layers

Posterior: The peritoneum covering the inferior vena cava

Superior: Peritoneum on the caudate lobe of the liver

Inferior: The peritoneum covering the commencement of the duodenum and the hepatic artery, with the latter passing forward below the foramen before ascending between the two layers of the lesser omentum.

FIGS 2.14A AND B: (A) Figure showing the examiner's fingers inserted in the epiploic foramen; (B) Transverse section at the level of T12 vertebra showing the epiploic foramen

Inguinal Region/Groin

Inguinal Canal

The inguinal canal in the adult is approximately 1.5 inches (4 cm) long and runs downwards and medially towards the superficial inguinal ring, starting from the deep inguinal ring **(Fig. 2.15)**. Therefore, the deep inguinal ring acts as the entrance point for the inguinal canal whereas the superficial inguinal ring acts as the exit point. The deep inguinal ring is situated in the transversalis fascia, midway between the anterior superior iliac spine and the symphysis pubis, and lies about 1.25 cm above the inguinal ligament and is lateral to the epigastric vessels. The inferior epigastric artery runs medial to the deep inguinal ring. Clinically, this has value in differentiating indirect (lateral to artery) from direct (medial to artery) inguinal hernias.

The superficial inguinal ring is a triangular slit in the external oblique aponeurosis just above and lateral to the pubic tubercle. Inguinal canal acts as a pathway through which the structures can pass from the abdominal wall to the external genitalia. It also acts as the potential area for the development of inguinal hernias.

Boundaries

Superior Wall (Roof)

- ❏ Medial crus of aponeurosis of external oblique
- ❏ Musculoaponeurotic arches of internal oblique and transversus abdominis
- ❏ Transversalis fascia.
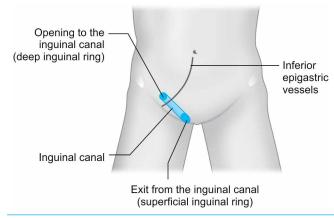
FIG. 2.15: Diagram showing the inguinal canal

Anterior Wall

- ❏ Aponeurosis of the external oblique (in the medial third)
- ❏ Fleshy part of internal oblique (lateral third of canal only).

Posterior Wall

- ❏ Transversalis fascia
- ❏ Medial-third of the posterior wall: Conjoint tendon (fused aponeuroses of the internal oblique and transversus abdominis), and inguinal falx (reflected part of inguinal ligament)
- ❏ Lateral-third of the posterior wall: Deep inguinal ring.

Inferior Wall (Floor)

- ❏ Inguinal ligament
- ❏ Lacunar ligament (medial third of canal only)
- ❏ Iliopubic tract (lateral third of canal only).

Contents of the Inguinal Canal

Contents of the inguinal canal are as follows **(Fig. 2.16)**:

Males
❑ Spermatic cord
❑ Ilioinguinal nerve (this nerve only passes through the superficial inguinal ring. It is not carried through the deep inguinal ring and therefore does not formally travel through the inguinal canal).

Females
❑ Round ligament (in the female the inguinal canal transmits the round ligament to the labium majus).
❑ Ilioinguinal nerve (this nerve only passes through the superficial inguinal ring. It is not carried through the deep inguinal ring).

Inguinal Ligament

The inguinal ligament is present at the upper end of the front of the thigh, i.e. at its junction with the anterior abdominal wall. The ligament is actually the thickened and folded lower edge of the aponeurosis of the external oblique muscle. It extends from the anterior superior iliac spine to the pubic tubercle in a curved line which folds posteriorly. Its medial attachment forms a narrow sling for support of the spermatic cord or round ligament of the uterus. The spermatic cord is present near the medial end of the inguinal ligament. It is seen to emerge through the superficial inguinal ring. Present a little below the medial end of the inguinal ligament is the saphenous opening. This is an oval aperture in the deep fascia of the thigh. The lateral and inferior margin of the opening is sharp and is known as the falciform margin.

Spermatic Cord

This is formed when testis passes through the inguinal canal descending into the scrotum. It has three coverings:
1. Internal spermatic fascia (derived from the transversalis fascia)
2. Cremasteric fascia (derived from internal oblique)
3. External spermatic fascia (derived from external oblique).

Contents: The contents of the cord are as follows:
❑ Vas deferens (ductus deferens)
❑ Three nerves:
  • Genital branch of genitofemoral nerve (supplies the cremaster muscle)
  • Ilioinguinal nerve (supplies the scrotum and the groin)
  • Sympathetic nerves.
❑ Three arteries:
  • Testicular artery (from the aorta)
  • Artery to the vas (from inferior vesical artery)
  • Cremasteric artery (from the inferior epigastric artery).
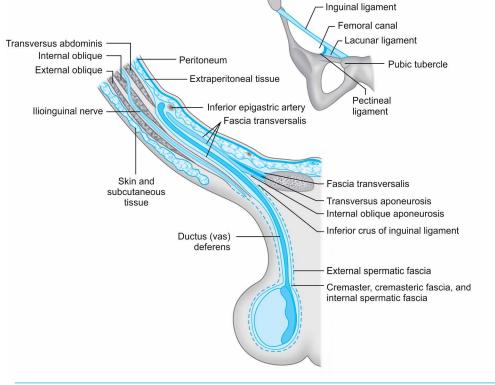
FIG. 2.16: Contents of the inguinal canal in a male (picture in the inset shows inguinal ligament and its modifications)

❏ Lymphatics (which drain to the para-aortic nodes)
❏ Pampiniform venous plexus
❏ Processus vaginalis (this is the obliterated peritoneal connection with the tunica vaginalis of the testes).

The inguinal ligament serves as a landmark for the following:
❏ The tendon of psoas major and the femoral branch of the genitofemoral nerve both pass under the inguinal ligament.
❏ The long saphenous vein terminates in the femoral vein about 3 cm below the inguinal ligament.
❏ The external iliac becomes the common femoral artery at the inguinal ligament.
❏ The superficial epigastric vein passes in front of the inguinal ligament.
❏ The midinguinal point lies halfway between the anterior superior iliac spine and pubic tubercle. The femoral artery crosses into the lower limb at this anatomical landmark.
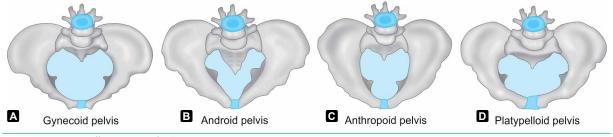
Anatomy of the Female Pelvis

The birth passage comprises of three parts, namely the pelvic inlet, pelvic cavity, and the pelvic outlet. The bony pelvis can be classified into four types: (1) gynaecoid, (2) android, (3) anthropoid, and (4) platypelloid **(Figs 2.17A to D** and **Table 2.3)**. Of these, the gynaecoid type of pelvis is the most common, with the diameters favorable for vaginal delivery. The anterior view of maternal gynaecoid pelvis is shown in **Figure 2.18**. Gynaecoid pelvis is an ideal type of pelvis and is characterised by the presence of the following features:
❏ The pelvic brim is almost round in shape, but slightly oval transversely
❏ Ischial spines are not prominent
❏ Subpubic arch is rounded and measures at least 90° in dimension

TABLE 2.3	Different pelvic types and their characteristics			
	Gynaecoid (40–50%)	**Anthropoid (20%)**	**Android (30%)**	**Platypelloid (2–5%)**
Pelvic inlet	Oval at the inlet with anterior-posterior diameter being just slightly less than the transverse diameter	Oval, long and narrow. The anterior-posterior diameter of the inlet exceeds the transverse diameter giving it an oval shape	Heart shaped/triangular with the base toward the sacrum. As a result, posterior segment is short, and anterior segment is narrow	Pelvic brim is flat and transverse kidney-shaped. Diameter is much larger than the anterior-posterior diameter
Sidewall	Straight	Straight	Convergent sidewalls (widest posteriorly)	Walls diverge downward
Subpubic arch	Wide and curved subpubic arch (subpubic angle is not <85°)	Subpubic arch is long and narrow; subpubic angle may be slightly narrowed	Long and straight subpubic arch; narrow subpubic angle	The subpubic arch is generally wide, and the subpubic angle is in the excess of 90°
Ischial spines	Ischial spines are not prominent	Ischial spines are not prominent	Ischial spines are prominent	Ischial spines are not prominent
Sacrum	Sacrum is well-curved, and sacral angle exceeds 90°	Sacrum is long and narrow with usual curve; sacral angle is >90°	Sacrum is inclined forward and straight; sacral angle is <90°	The sacrum is prominent and the sacral promontory tends to encroach upon the area of the hind pelvis; sacral angle is >90°
Bituberous diameter	Normal	Normal or short	Short	Wide
Sacro-sciatic notch	Wide and shallow	Wider and more shallow	Narrow and deep	Slightly narrow and small

A Gynecoid pelvis   **B** Android pelvis   **C** Anthropoid pelvis   **D** Platypelloid pelvis

FIGS 2.17A TO D: Different types of pelvis
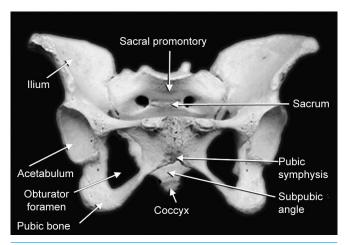
FIG. 2.18: Anterior view of maternal pelvis
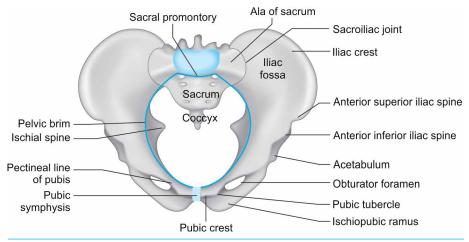
FIG. 2.19: Boundaries of the pelvic brim

❑ Obturator foramen is triangular in shape
❑ Sacrum is wide with average concavity and inclination
❑ Sacro-sciatic notch is wide.
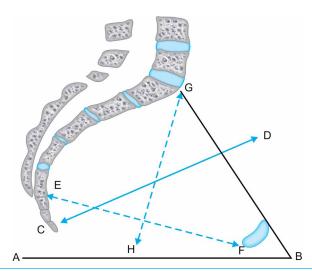
The pelvic brim **(Fig. 2.19)** divides the pelvis into false pelvis and true pelvis. The boundaries of the pelvic brim or inlet include the following: sacral promontory, sacral alae, sacroiliac joints, iliopectineal lines, iliopectineal eminence, upper border of superior pubic rami, pubic tubercles, pubic crest and upper borders of pubic symphysis.

❑ *False pelvis*: False pelvis lies above the pelvic brim and has no obstetrical significance.
❑ *True pelvis*: True pelvis lies below the pelvic brim and plays an important role in the childbirth and delivery. The true pelvis forms a bony canal through which the foetus passes at the time of labour. It is formed by the symphysis pubis anteriorly and sacrum and coccyx posteriorly. The true pelvis can be divided into three parts: (1) pelvic inlet, (2) cavity and (3) outlet.

Pelvic Inlet

Pelvic inlet is round in shape and is narrowest in antero-posterior dimension and widest in the transverse diameter. The foetal head enters the pelvic inlet with the longest diameter of the foetal head [anterior-posterior (AP) diameter] in the widest part of the pelvic inlet (transverse diameter).

The plane of the pelvic inlet (also known as superior strait) is not horizontal, but is tilted forwards. It makes an angle of 55° with the horizontal. This angle is known as the angle of inclination. Radiographically this angle can be measured by measuring the angle between the front of the vertebra L5 and plane of inlet and subtracting this from 180°. Increase in the angle of inclination has obstetric significance as this may result in delayed engagement of the foetal head and delay in descent of foetal head. Increase in the angle of inclination also favours occipitoposterior position. On the other hand, the reduction in the angle of inclination may not have any obstetric significance.

FIG. 2.20: Different planes and axes of the pelvis: AB—Horizontal line; GB—Plane of inlet; FE—Plane of obstetric outlet; DC—Axis of the inlet; GH—Axis of obstetrical outlet

FIG. 2.21: Medial view of maternal pelvis (from left)

The axis of the pelvic inlet is a line drawn perpendicular to the plane of inlet in the midline **(Fig. 2.20)**. It is in downwards and backwards direction. Upon extension, this line passes through the umbilicus anteriorly and through the coccyx posteriorly. For the proper descent and engagement of foetal head, it is important that the uterine axis coincides with the axis of inlet.

Diameters of the Pelvic Inlet (Anterior-Posterior Diameters) (Fig. 2.21)

Anterior-posterior diameter (true conjugate or anatomical conjugate = 11 cm): This is measured from the midpoint of sacral promontory to the upper border of pubic symphysis.

Obstetric conjugate (10.5 cm): The obstetric conjugate is measured from the midpoint of sacral promontory to the most bulging point on the back of symphysis pubis. This is the shortest AP diameter of the pelvic inlet and measures about 10.5 cm.

Diagonal conjugate (12.5 cm): It is measured from the tip of sacral promontory to the lower border of pubic symphysis.

Out of three AP diameters of the pelvic inlet, only diagonal conjugate can be assessed clinically during the late pregnancy or at the time of the labour. Obstetric conjugate can be calculated by subtracting 1.5–2 cm from the diagonal conjugate. Also the true conjugate can be inferred by subtracting 1.2 cm from the diagonal conjugate.

Measurement of the Diagonal Conjugate

After placing the patient in dorsal position and taking all aseptic precautions, two fingers are introduced into vagina. The clinician tries to feel the anterior sacral curvature with these fingers **(Fig. 2.22)**. In normal cases it will be difficult to feel the sacral promontory. The clinician may be required to depress the elbow and wrist while mobilising

Diagonal conjugate (12.5 cm)
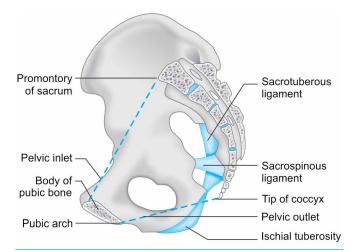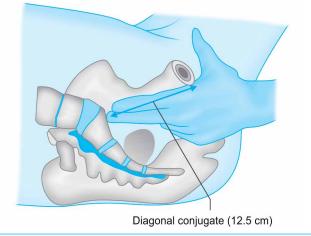
FIG. 2.22: Measurement of diagonal conjugate

the fingers upwards in order to reach the promontory. The point at which the bone recedes from the finger is sacral promontory. A marking is placed over the gloved index finger by the index finger of the other hand. After removing the fingers from the vagina, the distance between the marking and the tip of the middle finger is measured in order to obtain the measurement of diagonal conjugate. In clinical situations it may not always be feasible to measure the diagonal conjugate. In these cases if the middle finger fails to reach the sacral promontory or reaches it with difficulty, the diagonal conjugate can be considered as adequate. Under normal circumstances, an adequate pelvis would be able to allow an average-sized foetal head to pass through.

Transverse Diameter of Pelvic Inlet

Anatomical transverse diameter (13 cm): It is the distance between the farthest two points on the iliopectineal line **(Fig. 2.23)**. It is the largest diameter of the pelvic inlet and lies 4 cm anterior to the promontory and 7 cm behind the symphysis.
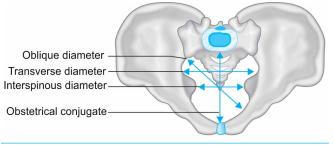
FIG. 2.23: Superior view of pelvic inlet

Obstetric transverse diameter: This diameter passes through the midpoint of true conjugate and is therefore slightly shorter than the anatomical transverse diameter.

Oblique Diameters of Pelvic Inlet

There are two oblique diameters, right and left (12 cm). The right oblique diameter passes from right sacroiliac joint to the left iliopubic eminence, whereas the left diameter passes from left sacroiliac joint to the right iliopubic eminence.

Pelvic Cavity

This is bounded above by the pelvic brim and below by the plane of least pelvic dimension, anteriorly by the symphysis pubis and posteriorly by sacrum. The plane of least pelvic dimension extends from the lower border of pubic symphysis to the tip of ischial spines laterally and to the tip of 5th sacral vertebra posteriorly.

Plane of Cavity (Plane of Greatest Pelvic Dimensions)

This plane passes between the middle of the posterior surface of the symphysis pubis and the junction between 2nd and the 3rd sacral vertebra. Laterally it passes through the centre of acetabulum and the upper part of greater sciatic notch. Since this is the roomiest plane of pelvis, it is also known as the plane of greatest pelvic dimensions. This is almost round in shape. Internal rotation of the foetal head occurs when the biparietal diameter of the foetal skull occupies this wide pelvic plane while the occiput is on the pelvic floor, i.e. at the plane of least pelvic dimensions.

Diameters of Pelvic Cavity

Anterior-posterior diameter (12 cm): It measures from the midpoint on the posterior surface of pubic symphysis to the junction of second and third sacral vertebra.

Transverse diameter (12 cm): It is the distance between two farthest points laterally. Since there are no bony landmarks, the diameter cannot be exactly measured and can be roughly estimated to be about 12 cm.

Pelvic Outlet

❑ *Anatomical outlet*: It is a lozenge-shaped cavity bounded by anterior border of symphysis pubis, pubic

arch, ischial tuberosities, sacrotuberous ligaments, sacrospinous ligaments and tip of coccyx.

❑ *Plane of anatomical outlet*: It passes along with the boundaries of the anatomical outlet and consists of two triangular planes with a common base, which is the bituberous diameter.

❑ *Anterior sagittal plane*: Its apex is at the lower border of the symphysis pubis.

❑ *Anterior sagittal diameter (6–7 cm)*: It extends from the lower border of the pubic symphysis to the centre of bituberous diameter.

❑ *Posterior sagittal plane*: Its apex lies at the tip of the coccyx.

❑ *Posterior sagittal diameter (7.5–10 cm)*: It extends from the tip of the sacrum to the centre of bituberous diameter.

❑ *Obstetric outlet*: It is bounded above by the plane of least pelvic dimensions, below by the anatomical outlet, anteriorly by the lower border of symphysis pubis, posteriorly by the coccyx and laterally by the ischial spines.

Diameters of Pelvic Outlet

Anterior-posterior diameters of pelvic outlet include the following:

Anatomical AP diameter (11 cm): It extends from tip of the coccyx to the lower border of symphysis pubis.

Obstetric AP diameter (13 cm): It extends from the lower border of symphysis pubis to the tip of coccyx (as it moves backwards during the second stage of labour).

Transverse diameter of the pelvic outlet includes the following:

Bituberous diameter (11 cm): It extends between the inner aspects of ischial tuberosities. The bituberous diameter is measured with the knuckles of the closed fist of the hand placed between the two ischial tuberosities. If the pelvis is adequate, the intertuberous diameter allows four knuckles.

Bispinous diameter (10.5 cm): It extends between the tips of ischial spines.

Measurement of various pelvic diameters is summarised in **Table 2.4**. A pelvis is contracted when one or more diameters are less than the minimal normal range.

TABLE 2.4	Summary of the measurement of the diameters of the pelvis		
Diameter	**Pelvic brim**	**Pelvic cavity**	**Pelvic outlet**
Anterior posterior	11 cm	12 cm	13 cm
Oblique	12 cm	12 cm	–
Transverse	13 cm	12 cm	11 cm

Pelvic Axis

Anatomical axis: This is an imaginary line joining the central points of the planes of inlet, cavity and outlet. This axis is C-shaped with concavity directed forwards. It has no obstetric significance.

Obstetric axis: It is an imaginary line, which represents the direction in which the head passes during the labour. It is J-shaped and passes downwards and backwards along the axis of the inlet till the ischial spines are reached after which it passes downwards and forwards along the axis of pelvic outlet **(Fig. 2.24)**.
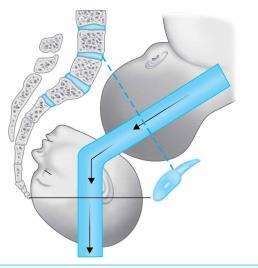
FIG. 2.24: Obstetric axis

Difference between the Male and the Female Pelvis

All diameters are absolutely greater in the female pelvis than in the male. In the male, the subpubic arch is about 50–60°. The sacral promontory is more prominent in the male, so that the male inlet is heart-shaped whereas the female inlet is more rounded, facilitating engagement of the foetal head. The obturator foramen is triangular in the female pelvis. The sacrum is shorter and wider in the female, and its upper part is straight. Difference between the male and female pelvis is tabulated in **Table 2.5** and illustrated in **Figures 2.25A and B**.
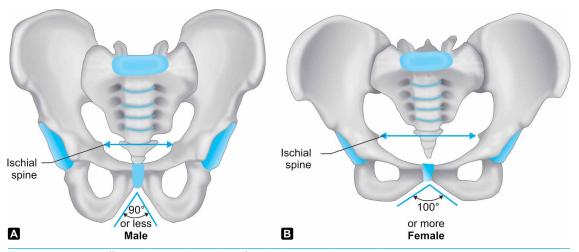
Sacrum

The sacrum is composed of five fused vertebral bodies. The lateral alae of sacrum articulate with the ilium to form the sacroiliac joints. Ventral rami of S1–S4 emerge from the pelvic sacral foramina. Two ligaments originate from the surface of sacrum, sacrospinous ligament and the sacrotuberous ligament.

❏ *Sacrospinous ligament*: Extends from the lateral margin of the sacrum and coccyx to the ischial spine.
❏ *Sacrotuberous ligament*: Extends from the sacrum to the ischial tuberosity.

The muscle piriformis originates from the anterior surface of the sacrum. The sacrum in a gynaecoid pelvis is broad, shallow and concave, as opposed to the flattened, narrow and long sacrum in the male pelvis.

TABLE 2.5	Difference between the male and female pelvis	
Parameter	**Female pelvis**	**Male pelvis**
General structure	Female pelvis is larger and broader	Male pelvis is taller (due to a higher iliac crest), narrower and more compact
Pelvic canal	Short and almost cylindrical	Long and tapered
Pelvic sidewalls	Are wide apart	Sidewall converge from the pelvic inlet towards the outlet
Shape of pelvic inlet	Large and oval	Heart shaped
Pelvic outlet	Comparatively large	Comparatively small
Subpubic arch	Subpubic arch is wider in females with the subpubic angle varying between 90° and 100°	Subpubic arch is narrower with the subpubic angle varying between 70° and 90°
Ischial spines and tuberosity	Less prominent	Heavier and project farther into the pelvic cavity in males
Sacrum	Shorter, wider and flatter. It is more curved posteriorly with a less pronounced promontory	Long, narrow, straighter with a pronounced sacral promontory
Articular surface of the sacrum	Articulates laterally with two sacral bodies; superiorly with L5; oval and occupies one-third of the alar surface	Articulates laterally with three sacral bodies; superiorly with L5 and occupies half of the alar surface
Obturator foramen	Triangular	Oval in shape
Acetabula	Wider and face medially	Narrow and face laterally

FIGS 2.25A AND B: Difference between the male and female pelvis: (A) Male; (B) Female

Pelvic Organs

Pelvic organs, which comprise the female internal genitalia in a woman, consist of vagina, uterus, fallopian tubes and ovaries. In a male, the pelvic organs, which form part of the internal male genitalia, include testes, epididymis, ductus deferens, seminal vesicles, ejaculatory ducts, bulbourethral glands, prostate. Pelvic organs, which form a part of the gastrointestinal tract, include rectum and anal canal in both males and females. **Figure 2.26** illustrates the median section of the female pelvis.

Vulva

An ill-defined area containing the next-mentioned external genital organs along with the perineum is known as the vulva. The external genital organs include the mons pubis, labia majora, labia minora, the vestibule, the hymen or its remnants, the ostia of the accessory glands (Bartholin's glands, Skene's glands and the vestibular glands) and clitoris. Anteriorly the vulva is bound by mons pubis, posteriorly by the perineum and on the two sides by the labia majora. The mons pubis is a rounded mass of fatty tissue that covers the pubic bone. During puberty, it becomes covered with hair. The mons pubis contains sweat glands and the oil-secreting (sebaceous) glands that release substances which are involved in sexual attraction (pheromones). The labia majora are relatively large, fleshy folds of tissue which pass anteriorly from mons veneris to end posteriorly in the skin over the perineal body. They are comparable to the scrotum in males. The inner surface of labia majora is softer, moister and not covered with hair in contrast to the outer surfaces. The labia majora contains sweat and sebaceous glands, which produce lubricating secretions. They also contain apocrine glands, secretions of which are responsible for the characteristic aroma of the vaginal secretions. After puberty, hair appears on the
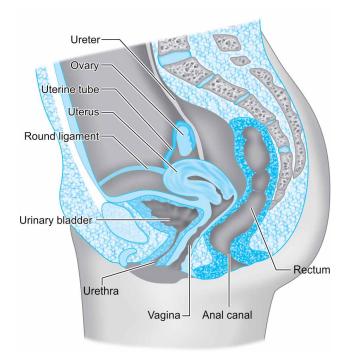
FIG. 2.26: Median section of the female pelvis

mons pubis and the outer surface of labia majora. The common skin lesions which can occur in this region include folliculitis, boils and sebaceous cysts. The labia minora are found just medial to the labia majora. They contain no hair follicles, sweat or sebaceous glands. Anteriorly they enclose the clitoris and posteriorly they join to form the fourchette. A rich supply of blood vessels gives the labia minora a pink colour.

During sexual stimulation, these blood vessels become engorged with blood, causing the labia minora to swell and become more sensitive to stimulation.

The vestibule is the part of the vulva lying between the two labia minora laterally and extends medially to the hymenal sulci. The Bartholin's glands are located in the vestibule on either side. They produce mucus and

participate in lubrication during sexual intercourse. Skene's glands are a pair of paraurethral glands, located on the anterior wall of vagina around the lower end of the urethra. They are homologous with the prostate glands in males. Besides these glands, there are minor vestibular glands located around the vestibule. The clitoris is an erectile organ, located between the labia minora at their upper end that corresponds to the penis in the male. It consists of a glans, covered with frenulum and prepuce, and a body. It is attached to the undersurface of pubic symphysis with the help of a suspensory ligament. The normal length of clitoris is about 1–1.5 cm and width is about 5 mm. Length of more than 3.5 cm and width of greater than 1 cm is termed as clitoromegaly. The clitoris, like the penis, is very sensitive to sexual stimulation and can become erect, resulting in an orgasm. It has a rich supply of blood vessels and nerves. Therefore, injury to clitoris can result in profuse haemorrhage and can be extremely painful.

The development of vulvar tissues occurs under the influence of hormone oestrogen. Following menopause, as a result of oestrogen deficiency, the vulvar skin becomes thinner and drier resulting in atrophic vulvitis and itching.

The external genital organs have three main functions: (1) Enabling sperm to enter the body, (2) protecting the internal genital organs from infectious organisms and (3) provision of sexual pleasure.

Blood Supply

Blood supply to the external genitalia is from the following vessels:

❑ *Superficial external pudendal artery*: The superficial external pudendal artery is one of three superficial branches of the femoral artery near the inguinal ligament. It supplies the skin and superficial fascia of the upper medial thigh, and skin of the pubic region.

❑ *Deep external pudendal artery*: The deep external pudendal artery usually derives from the femoral artery and supplies the labium majus. It anastomoses with the internal pudendal artery. Though deep external pudendal artery usually arises from the femoral artery, it may sometimes also arise from the medial circumflex femoral artery.

❑ *Internal pudendal artery*: The internal pudendal artery terminates in branches which supply the perineal and vulval structures, including the erectile tissue of the vestibular bulb and clitoris.

❑ *Inferior epigastric artery*: The mons pubis is supplied by the inferior epigastric artery, a branch of the external iliac artery.

Lymphatic Drainage

Vulva drains into the superficial inguinal lymph nodes. The lymphatics cross the midline in the anterior part of the vulva.

Nerve Supply

The innervation of the external genitalia is mainly by the pudendal nerve, which arises at S2–S4 levels and accompanies the pudendal vessels. This nerve branches anteriorly to form the perineal nerve and the dorsal nerve of clitoris, which innervate the perineal membrane, external genitalia and the clitoris. Posteriorly, another branch of the pudendal nerve, the inferior rectal nerve, supplies the ischiorectal fossa and the anal area. The perineal nerve gives the sensory supply to the vulva; it also innervates the anterior part of the external anal sphincter and levator ani, and the superficial perineal muscles. The dorsal nerve of the clitoris is sensory.

Two more nerves, the ilioinguinal (L1) and the genital branch of the genitofemoral (L1–L2), arising from the lumbar plexus, innervate the medial and lateral aspects of the vulvar skin respectively.

Female Internal Genitalia

Vagina

The vagina is a narrow, muscular but elastic organ, having a length of about 4–5 inches in an adult woman. It connects the external genital organs to the uterus. The vagina is the main female organ of sexual intercourse. It acts as the passageway for transportation of sperms and for the expulsion of the menstrual blood. It helps in the delivery of the baby. The lower end of vagina lies at the level of hymen and at this level, it is surrounded by erectile tissues, which correspond to the corpus spongiosum of males. The vaginal portion of the cervix projects into the upper part of the vagina, resulting in the formation of anterior, posterior and lateral fornices. The posterior fornix, which is the deepest, is related to the rectouterine pouch. The depth of the fornices depends upon the development of the portio vaginalis of the cervix. The attachment of the vagina to cervix is at a higher level on the posterior aspect in comparison to the other regions. As a result, the posterior vaginal fornix is the deepest and the posterior vaginal wall longest in comparison to the anterior or lateral walls. The posterior wall is about 4.5 inches long, whereas the anterior wall is about 3.5 inches. Therefore the posterior vaginal wall is longer than the anterior vaginal wall. The anterior and posterior vaginal walls are in contact with each other, not the lateral walls. There are three sulci in the anterior vaginal wall. One lies above the urethral meatus and is known as the submeatal sulcus. About 3.5 cm above this sulcus in the anterior vaginal wall, there is another sulcus called as transverse vaginal sulcus, which corresponds to the junction of urethra and bladder. Further upwards is the bladder sulcus, which is indicative of the junction of the bladder to the anterior vaginal wall. The vaginal mucosa is lined by stratified squamous epithelium. There are no glands in the vagina and the vaginal secretions are mainly derived from

the mucus discharge of the cervix and transudation through the vaginal epithelium. The vagina has no glands but the vestibular glands provide moisture for the vagina. The pubococcygeal muscles act as a sphincter for the vagina.

In the sexually mature woman, the epithelial squamous cells of the vagina can be classified into four distinct histologic zones:

1. *Basal cells*: This comprises of a single layer of cuboidal cells attached to the basement membrane. These cells are firmly attached to the basement membrane and do not exfoliate, so are not present in the vaginal smears. These are the least mature cells from which the regeneration of the epithelium is maintained.
2. *Parabasal cells*: This zone comprises of several rows of polyhedral cells superficial to the basal layer. These cells may be seen in Pap smears. Upon exfoliation, these cells lose their intercellular bridges and appear round or oval.
3. *Intermediate layer*: This zone comprises of several rows of slightly larger, flatter cells above the parabasal cellular layer. Upon exfoliation, these cells appear larger and less rounded than the parabasal cells. On histopathological examination, the lowermost parabasal cells stain bluish-green, whereas those near the surface take up the eosinophilic stain.
4. *Superficial zone*: This zone comprises of several layers of large, flat cells having dark pyknotic nuclei. In Pap smears, these cells appear large and polyhedral having a clear transparent cytoplasm. Under certain pathological conditions, these cells may lose their nuclei and become keratinised, thereby appearing orange coloured on Papanicolaou stain. The superficial squamous cells in this zone usually stain pink unless the vaginal pH becomes abnormal.

After puberty, during the reproductive phase, increased oestrogen production causes proliferation of the vaginal stratified cells and increases their glycogen content, thereby allowing the formation of lactic acid by Doderlein's bacilli from the glycogen present in the epithelial cells and increasing resistance to infection. This results in a vaginal secretion with a pH of about 4. The acidic pH inhibits the growth of other pathogenic organisms. During reproductive life, the vaginal pH remains on an average about 4.5. Before puberty and after menopause, this pH becomes about 7.

Altered oestrogen levels and conditions such as pregnancy, diabetes, prolonged antibiotic therapy, etc. may result in a reduction in the Doderlein's bacilli, thereby disrupting the natural vaginal flora. This may lead to the development of infection such as candidiasis and bacterial vaginosis.

Relations

Anteriorly: The vagina is related anteriorly to the cervix, ureters, and bladder and is fused with the urethra. **Figure 2.27** illustrates the peritoneal reflection of the vagina and the bladder.
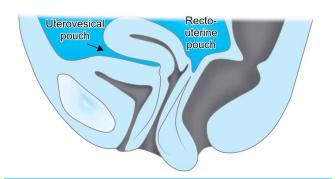
FIG. 2.27: Peritoneal reflection of the vagina and the bladder

Posteriorly: The rectouterine pouch (pouch of Douglas), the rectum, and the perineal body. The pouch of Douglas extends down up to the upper one-third of the posterior vaginal wall.

Laterally: Ureter and uterine artery.

Blood Supply

The vagina is supplied by the uterine and vaginal branches of the internal iliac artery. Other sources include inferior vesical and middle rectal arteries, which anastomose freely on the vaginal wall. The vaginal artery is replaced by the inferior vesical artery in males. The venous drainage of vagina is to the vaginal venous plexus with the vaginal vein draining into the internal iliac vein or the uterine vein.

Nerve Supply

The uterovaginal nerve plexus lying in the base of the broad ligament on the either side of the supravaginal part of the cervix gives rise to the parasympathetic and sympathetic nerves supplying the vagina. The sympathetic fibres are derived from lumbar splanchnic nerves, whereas the parasympathetic fibres are derived from the pelvic splanchnic nerves. The inferior fibres from the uterovaginal plexus supply the superior part of the vagina. These are derived from the inferior hypogastric plexus and the pelvic splanchnic nerves.

The nerve supply to the lower part of the vagina is from the branch of pudendal nerve called the deep perineal nerve.

Lymphatic Drainage

The lower one-third of the vagina drains to the superficial inguinal lymph nodes (similar to the vulval drainage), while the upper two-thirds drain into the external and internal iliac and sacral nodes (similar to that of cervix).

Fallopian Tubes

Also known as the oviduct or the uterine tube, each fallopian tube is about 2–3 inches long and extends from the upper edge of the uterus toward the ovaries. The two fallopian

tubes normally extend laterally from the uterine cornua and open into the peritoneal cavity near the ovaries by flaring into a funnel-shaped structure, infundibulum having finger-like projections (fimbriae). Thus, the tubes do not directly connect with the ovaries. When an oocyte is released from an ovary, the fimbriae guide it towards the infundibulum of the fallopian tube. Fallopian tube has a lining of ciliated cells interspersed with non-ciliated secretory cells ("peg" cells). The fallopian tube must be sufficiently mobile to assist the ovum onwards by peristalsis. Presence of cilia along with the muscles of the tubal wall helps in propelling an oocyte downwards through the fallopian tube into the uterine cavity where it ultimately implants in form of a blastocyst. A healthy fallopian tube is not palpable, but if it is thickened it may be felt. The fallopian tube runs in the upper margin of the broad ligament part of which is known as the mesosalpinx. This encloses the tube in such a way that it is completely covered with peritoneum except for a narrow strip along its inferior aspect. Starting from the lateral to medial side, the fallopian tube can be divided into four parts, which are as follows:

1. *Infundibulum*: This is the funnel-shaped distal end of the tube, which opens into the peritoneal cavity through the abdominal ostium. The finger-like projections of the fimbriated end of the infundibulum (fimbriae) spread over the medial surface of the ovary. One large ovarian fimbria is attached over the superior pole of the ovary.
2. *Ampulla*: This is the widest and the largest part of the tube, which lies medial to the infundibulum. Fertilisation of the oocyte usually occurs in the ampulla.
3. *Isthmus*: This is the thick-walled part of the tube, which enters the uterine cornu.
4. *Uterine part*: This includes the short intramural segment of the tube, which passes through the wall of the uterus and opens via the uterine ostium into the uterine cavity at the uterine cornu.

Ovaries

The ovaries are almond-shaped, pearl-coloured, female gonads responsible for producing the oocytes (female gametes or the germ cells). It is commonly situated on the lateral wall of the pelvis in the angle between the external iliac vein and the ureter, where it can be palpated upon bimanual examination. The normal ovary measures 4 cm × 2 cm (1.5 inches × 0.75 inch). In postmenopausal women, the ovaries become smaller and shrunken and are covered with scar tissue. In the woman of reproductive age group, the developing egg cells (oocytes) are contained by the fluid-filled cavities called follicles, present in the ovarian walls. Each ovary is suspended by a short fold of peritoneum known as the mesovarium, which arises from the broad ligament and convey the ovarian vessels **(Fig. 2.28)**. The ovaries lie above the pelvic brim at the time of birth and do not descend down until the cavity of the
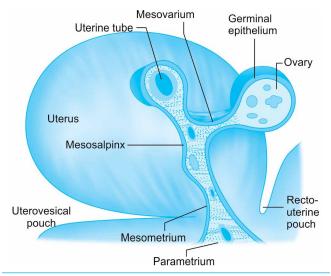
FIG. 2.28: Sagittal section of the left broad ligament showing its relationship to the ovaries and the uterine tubes

pelvis deepens during childhood. The enlarging uterus at the time of pregnancy is more likely to pull the ovaries into the abdominal cavity. The ovaries are sensitive to touch and pressure.

In the prepubertal woman, the connective tissue capsule over the surface of the ovary is covered by a smooth layer of ovarian mesothelium or surface germinal epithelium. This usually comprises of a single layer of cuboidal cells and is usually continuous at the hilum with the peritoneum and the mesovarium. These cubical cells lie on a dense layer of connective tissue, the tunica albuginea and give the ovarian surface a dull, greyish appearance. After puberty, the surface epithelium becomes progressively scarred and distorted because of the repeated rupture of the ovarian follicles and discharge of oocytes during ovulation. The Graafian follicles are interspersed throughout the stroma, and some of them may be seen bulging at the surface of the ovary; they vary in size according to their stage of development. Free surface of ovary has no peritoneal covering. The ovaries lie close to the lateral pelvic walls suspended from the posterior surfaces of the broad ligaments **(Fig. 2.29)**. Therefore, ovarian disease, which involves the parietal peritoneum at this site, may produce pain referred via the nerve to the medial side of the thigh. The suspensory ligament of the ovary (or infundibulopelvic ligament) extends from the ovary to the lateral pelvic wall. It is not considered a true ligament because it does not physically support any anatomical structure. The ovarian artery descends in the suspensory ligament and, by way of the broad ligament and mesovarium, enters the hilum of the ovary.

The ovarian ligament, on the other hand, connects the ovary to the body of the uterus, immediately posterior and inferior to the entrance of the uterine tube. It is a short ligament present medially within the mesovarium and lies beneath the posterior layer of the broad ligament. Together,
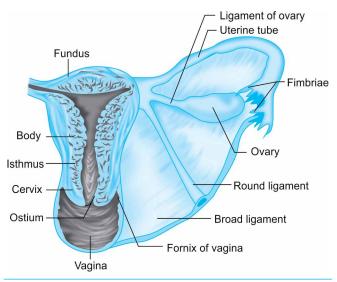
FIG. 2.29: Posterior view of the female reproductive tract

the ovarian and round ligaments are homologous with the gubernaculum testis of the male.

A baby girl is born with oocytes in her ovaries. No new oocytes develop after birth. Between 16 weeks and 20 weeks of gestation, the ovaries of a female foetus contain 6–7 million oocytes. Most of these oocytes gradually die away, leaving about 1–2 million oocytes to be present at birth. At puberty, only about 300,000 oocytes remain, of which only a small percentage mature into eggs. The many thousands of oocytes that do not mature undergo degeneration. Degeneration is usually complete by the time the woman attains menopause.

Since the ovary is suspended inside the peritoneal cavity and its surface is not covered with peritoneum, the oocyte expelled at the time of ovulation passes into the peritoneal cavity. However, its intraperitoneal life is short because it is soon trapped by the fimbriae of the infundibulum of the uterine tube. Eventually, the ovum is carried to the ampulla where it is fertilised. Each ovary has tubal and uterine ends, medial and lateral surfaces, and mesovarian (anterior) and free (posterior) borders.

Relations

The superior or tubal end: This is closely related to the uterine tube and is attached to the suspensory ligament of the ovary.

The inferior or uterine end: This is attached to the ovarian ligament.

Medial surface: This is related to the uterine tube and the ileum.

Lateral surface: Lateral surface of the ovary is in contact with the parietal peritoneum that lines the sidewall of the pelvis.

The anterior border: It is attached to the mesovarium, and contains hilum of the ovary.

The posterior border: It is related to the uterine tube and ureter.

Blood Supply

The ovary derives its blood supply directly from the abdominal aorta. The ovary is supplied by the ovarian artery, a direct branch of the abdominal aorta. The ovarian artery arises from the aorta at the level of first lumbar vertebra. After crossing the pelvic brim this vessel enters the broad ligament and divides into terminal branches within the mesovarium. Both the ovarian artery and the vein enter and exit the ovary at the hilum. The left ovarian vein drains into the left renal vein and the right ovarian vein drains directly into the inferior vena cava.

The ovarian artery arises from the aorta just below the renal artery and runs downwards on the anterior surface of the psoas muscle to the pelvic brim, where it crosses in front of the ureter and then passes into the infundibulopelvic fold of the broad ligament. As the ureter crosses the brim of the pelvis it lies in front of the bifurcation of the common iliac artery. It runs downwards and forwards on the lateral wall of the pelvis to reach the pelvic floor, and then passes inwards and forwards to pass beneath the uterine artery.

Relations of the Ovarian Artery

Right ovarian artery: Right ovarian artery crosses the inferior vena cava and is crossed by the following:
- Middle colic vessels
- Caecal veins
- Terminal ileal vein
- Ileocolic vein.

Left ovarian artery: The left ovarian artery is crossed by the following:
- The left colic and the sigmoid branches of the inferior mesenteric vessels
- Descending colon.

Nerve Supply

The nerve supply to the ovaries (the ovarian plexus) includes parasympathetic, postganglionic sympathetic and autonomic afferent fibres.

Lymphatic Supply

The lymphatics from the ovary drain into the para-aortic nodes on both sides of the midline. The iliac nodes are also sometimes involved.

Ovarian Fossa

The ovarian fossa is a depression in the lateral wall of the pelvis and is present between the external and internal

iliac vessels. The ovaries usually lie in this fossa. The ureter descends in the posterior boundary of the fossa while the obturator nerve and vessels cross its floor.

Relations

- *Superiorly*: External iliac vessels
- *Anteriorly*: Obliterated umbilical artery
- *Posteriorly*: Ureter and internal iliac artery.

Uterus

The uterus is a thick-walled, muscular, pear-shaped organ located in the middle of the pelvis, in which the development of foetus and embryo occurs. The adult uterus comprises of two main parts: body (uterine corpus) and cervix. The non-gravid uterus lies in the lesser pelvis between the urinary bladder and rectum. Uterus is formed by the 10th week of gestation by the union of two Müllerian ducts, with fusion beginning in the midline, then extending caudally and in cephalad directions. Uterine body is formed from mesoderm. Uterine cavity is formed as the septum dissolves slowly.

The uterus is anchored in its position by several ligaments. The uterus is a very dynamic structure, the size and proportions of which change during the various stages of life. The non-gravid uterus is approximately 7.5 cm long, 5 cm wide and 2 cm thick. It weighs about 90 g. In an adult woman, the uterine body is twice as long as the cervix, whereas the converse is true in the newborn.

The adult uterus is usually anteverted and anteflexed so that its mass lies over the bladder and the cervix. When the bladder is empty, the uterus lies in a transverse plane. In about one-fifth of normal women the uterus may be retroverted. Uncomplicated retroversion does not cause infertility, and in the absence of incarceration it will not cause miscarriage. If pregnancy occurs, the uterus nearly always rises up into the abdomen in the normal way at about 12th week, and after delivery it resumes its retroverted position. In early pregnancy, no attempt should be made to correct the position.

Peritoneal Relations of the Uterus

The uterus (except for the cervix) is covered anteriorly and superiorly by the peritoneum. Anteriorly, the uterine body is separated from the urinary bladder by the vesicouterine pouch where the peritoneum is reflected from the uterus onto the posterior margin of the superior surface of the bladder. Posteriorly, the peritoneum is reflected from the posterior aspect of the body of the uterus, cervix and the vaginal fornix on to the anterior surface of the rectum (rectouterine pouch). The relations of the uterus can be summarised as follows:

Anterior relation: The uterus is anteriorly related to the vesicouterine pouch and the superior surface of the bladder. The supravaginal part of the cervix is directly related to the bladder and is separated from it only by fibrous connective tissue.

Posterior relation: The uterus is posteriorly related to the rectouterine pouch containing loops of small intestine and the anterior surface of the rectum.

Lateral relation: The uterus is related laterally to the peritoneal broad ligaments on each side of the cervix and vagina and fascial cardinal ligaments on each side of the cervix and vagina.

Peritoneal Attachments of Uterus

Broad ligament: This is a double fold of peritoneum, which covers the uterus and continues up to the lateral pelvic wall. The broad ligament, similar to the round ligament has no supporting function to the uterus. It just helps maintain uterus in the position of anteversion. The fallopian tube lies between the two layers of the broad ligament. This part of the broad ligament is known as the mesosalpinx, whereas the part adjacent to the uterus is called the mesometrium. Besides the uterine tube, the broad ligament contains connective tissue (the parametrium), the uterine and ovarian vessels, the round and ovarian ligaments, and some embryonic remnants (e.g. the epoophoron, which consists largely of a duct parallel to and below the uterine tube). The posterior layer of the broad ligament adjacent to the ovaries forms the mesovarium.

Round ligament: This is a fibrous band attached to the uterus immediately below the entrance of the uterine tube. From here, it extends laterally and anteriorly, hooks around the inferior epigastric artery, travels in the inguinal canal, and eventually ends in the labium majus. The round ligament is usually accompanied in the foetus by a process of peritoneum, the processus vaginalis. Processus vaginalis can be occasionally present in the adults.

Supporting Ligaments of the Uterus

There are various ligaments, which support the uterus and are derived from the condensation of the parametrial tissues. **Figure 2.30** illustrates horizontal section of the pelvic viscera showing various uterine ligaments. On the other hand, ligaments formed as a result of peritoneal reflection and/or folding have minimal role in supporting the uterus.

Transverse cervical ligament: The visceral pelvic fascia on the lateral aspect of the cervix is thickened in form of the lateral (or transverse) cervical (or cardinal) ligament. It forms the uterosacral ligament on the posterior aspect.

Parts of the Uterus

Uterine Corpus

The uterine corpus forms the superior two-thirds of the uterus. The corpus of the uterus is a highly muscular
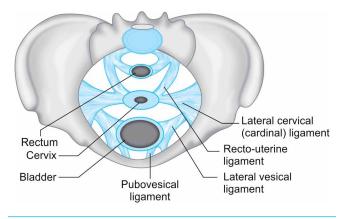
FIG. 2.30: Horizontal section of the pelvic viscera showing various uterine ligaments

structure, which can stretch to accommodate a growing foetus. Its muscular walls contract during labour to push the baby out through the cervix and the vagina. The uterine corpus includes the fundus of the uterus, the rounded part of the uterus, which lies superior to the uterine cornu. The remaining part of the body lies between the two layers of broad ligament and is freely movable. During the reproductive years, the corpus is twice as long as the cervix. After menopause, the reverse is true and the cervix is twice as long as the uterine corpus.

The uterine cavity is slit-like, which is approximately 6 cm in length and extends from the external os to the walls of the fundus. The uterine horns (cornu) are in the superolateral regions of the uterine cavity, where the uterine tubes enter. The uterine cavity continues inferiorly as the cervical canal. The uterine cavity, particularly the cervical canal constitutes the birth canal through which the foetus passes out at the end of gestation. The wall of the body of uterus comprises of three layers:

1. *Parametrium*: This is the serosa or the outer serous coat, which comprises of peritoneum.
2. *Myometrium*: This comprises of the middle coat of smooth muscles, which become greatly distended during pregnancy. The contraction of the myometrial muscles helps in the expulsion of the foetus and placenta.
3. *Endometrium*: This is the inner mucous coat, which firmly adheres to the underlying myometrium. The endometrium is actively involved in the menstrual cycle. If conception occurs, the blastocyst gets implanted in this layer. If the conception does not occur, the inner surface of this layer is shed, resulting in menstrual bleeding. There is no submucosa in the uterus. There-fore the endometrium is applied directly to the muscle.

Cervix

The cervix forms lower third of the uterus and is approximately 2.5 cm in length in an adult non-pregnant woman. The sperms enter the uterine cavity through the cervical canal. It also serves as a passage for the exit of menstrual blood. During labour, the cervical canal widens to form the lower uterine segment, the contractions of which help in the expulsion of the foetus. The cervix serves as a good barrier against bacterial infection, except around the time of ovulation, during the menstrual period, or during labour. Bacteria responsible for causing the sexually transmitted diseases can enter the uterus through the cervix at the time of sexual intercourse.

The cervix is composed of two parts: supravaginal part (between the uterine isthmus and the vagina) and the vaginal part (which protrudes into the vagina). The portion of cervix projecting into the vagina is known as ectocervix or portio vaginalis. The part of cervix within the uterine cavity is known as the endocervix. The opening of the ectocervix is known as the external os. In women who have not borne children, the external os is circular, but after labour it becomes a transverse slit. The external os of the cervix is normally on a level with the ischial spines.

The opening of the cervix inside the uterine cavity is known as the internal os. The passage between the external os and internal os is known as the endocervical canal. The epithelium of cervix is varied.

The ectocervix is composed of stratified squamous epithelium; whereas the endocervix, which lies within the uterus, is composed of simple columnar epithelium. The area adjacent to the junction of ectocervix and endocervix is known as the transformation zone. The location of squamo-columnar junction in relation to the external os is variable over a woman's lifetime. The squamo-columnar junction is under hormonal influence and so alters during puberty and the menopause. The squamo-columnar junction usually recedes within the endocervical canal after the menopause. At puberty the endocervical epithelium, which is simple columnar epithelium, extends distally into the vagina, forming an ectropion. The ectropion can sometimes be a cause of post-coital bleeding or mucoid vaginal discharge, and most commonly occurs during pregnancy or in the woman taking the combined oral contraceptive pills. Since the transformation zone is the most common site for cervical carcinoma, it is important to brush this site adequately when performing a cervical smear.

The endocervical glands are responsible for producing mucus, whose consistency varies during various phases of the menstrual cycle. This mucus is thick and impenetrable to sperms until just before ovulation. At ovulation, the consistency of the mucus changes and it becomes more thin and stretchable so that sperms can penetrate through it and fertilisation can occur. The cervix is mostly composed of fibrous tissue, which mainly comprises of collagen and a small amount of elastin. On the other hand, the uterine corpus has a largely muscular structure.

Peritoneal relations of the cervix: The anterior part of supravaginal portion of cervix is not covered by the peritoneum and is separated in front from the bladder by

fibrous tissue known as parametrium, which extends also on to its sides and laterally between the layers of the broad ligaments. The uterine arteries reach the margins of the cervix in this fibrous tissue. Posteriorly the peritoneum passes downwards from the uterine body to cover the posterior surface of the supravaginal cervix and the upper third of the posterior vaginal wall. The peritoneum is then reflected on to the rectum, resulting in the formation of the rectouterine pouch or the pouch of Douglas.

Lymphatic drainage of the cervix: The lymphatics from the cervix pass either laterally in the base of the broad ligament or posteriorly along the uterosacral ligaments to reach the sidewall of the pelvis. Most of the vessels drain to the internal iliac, obturator and external iliac nodes, but some vessels also pass directly to the common iliac and lower para-aortic nodes.

Innervation of the cervix: Pain from the cervix is carried by pelvic splanchnic nerves, hence there can be bradycardia during cervical dilatation.

Blood Supply to the Uterus

Figure 2.31 illustrates blood supply to the female internal genitalia. The main blood supply to the uterus is via the uterine artery (one on each side), which is a branch of the anterior trunk of the internal iliac artery. In addition, there is an anastomosis with the tubal branch of the ovarian artery, which contributes to the supply of the uterine fundus. The uterine artery passes inferiorly from its origin into the pelvic fascia. The uterine artery first runs downwards on the lateral wall of the pelvis, in the same direction as the ureter; then turns inwards and forwards, lying in the base of the broad ligament. By taking such a course, the uterine artery crosses above the ureter at a distance of about 2 cm from the uterus, at the level of the internal os. The ureter, therefore, passes under the uterine artery.

On reaching the wall of the uterus, the uterine artery turns upwards to run tortuously along the lateral uterine sidewalls to reach the upper part of the uterus near the entrance of fallopian tubes. It continues to move along the lower border of the fallopian tube where it ends by anastomosing with the ovarian artery, a direct branch of the abdominal aorta. In this part of its course, the uterine artery gives rise to branches, which run transversely and pass into the myometrium. These are known as the arcuate arteries. Blood supply to anterior and posterior walls is provided by the arcuate arteries, which run circumferentially around the uterus. From the arcuate vessels, branches known as the radial arteries arise at right angles. They reach the basal layers of endometrium where they are termed as the basal arteries. From the basal arteries, spiral and straight arterioles of the endometrium are derived. The arcuate artery to the cervix is also known as the circular artery of the cervix. The uterine artery gives off a small descending branch that supplies the cervix and the vagina. The uterine artery also supplies branches to the fallopian tube and ureter as it crosses it. Cervicovaginal branches anastomose with vaginal arteries to form the azygos arteries of the vagina.

The vaginal and ovarian arteries do not enlarge during pregnancy. Trophoblastic invasion of the spiral vessels during second trimester in normal pregnancy is responsible for a 10-fold increase in blood flow. Due to this trophoblastic invasion, the small muscular spiral arteries get converted into large vascular channels, which transform the uteroplacental circulation into a low-resistance system.

The uterine vein follows the uterine artery all along its course in the broad ligament and forms a uterine venous plexus on each side of the cervix. The uterine vein ultimately drains into the internal iliac vein. The uterine veins do not have surrounding sheaths unlike the veins of the arms and legs. The uterine venous plexus is connected with the superior rectal vein, thereby forming a portal systemic anastomosis.

Innervation of the Uterus

Sympathetic fibres of the uterus mainly arise from the uterovaginal plexus, which is largely formed from the
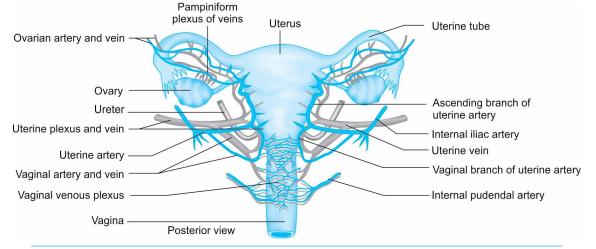
FIG. 2.31: Blood supply to the female internal genitalia

inferior hypogastric plexus. Parasympathetic fibres of the uterus are derived from the pelvic splanchnic nerves (S2–S4). The afferent fibres mainly ascend through the inferior hypogastric plexus to enter the spinal cord via T10–T12 and L1 nerve fibres. The uterus contains alpha-receptors, which cause contractions in the pregnant uterus, and beta-receptors, which cause relaxation in the non-pregnant uterus. Thus the uterine contraction and relaxation is under the control of sympathetic nervous system.

Hypogastric plexus is a plexus of nerves, which supplies the viscera of the pelvic cavity. It contributes branches to the uterine/vaginal plexus in females, vesical plexus in both males and females and to the prostatic plexus in the males.

The inferior hypogastric plexus is a paired structure, which lies between the pelvic viscera (vagina and rectum) and the pelvic wall in females and on the either side of the rectum in males. It extends into the base of the broad ligament of the uterus and lies between the two iliac vessels.

The sources of the inferior hypogastric plexus are as following:

❏ Hypogastric nerve, which is a continuation of the superior hypogastric plexus on the either side
❏ Sacral splanchnic nerves (postganglionic sympathetic axons)
❏ Pelvic splanchnic nerves (preganglionic parasympathetic axons from the ventral primary rami of spinal nerves S2–S4).

Lymphatic Drainage

The fundus and upper part of the uterine body drain into the lumbar (aortic) nodes, the lower part of the body into the external iliac, nodes, and the cervix into the external and internal iliac and the sacral nodes. The lymphatic drainage of the fundus of the uterus is into the para-aortic nodes (at the level of first lumbar vertebra). Nearly all the lymphatic vessels from the corpus uteri join those leaving the cervix and therefore reach similar groups of nodes. A few vessels at the fundus follow the ovarian channels, and there is an inconstant pathway along the round ligament to the superficial inguinal group of lymph nodes.

Male Internal Genitalia

Testes

Each testis (one on either side) is an oval-shaped structure about 4 cm in its longest (vertical) diameter and lies in the scrotal sac of each side respectively. The outer surface of the testis is formed by a dense fibrous membrane called the tunica albuginea. The substance of testis comprises of a large number of lobules, which are separated from each other by septa. Each lobule contains one or more highly convoluted seminiferous tubules. These tubules are lined by an epithelium, the cells of which are concerned with the production of spermatozoa. It has been estimated that each testis has about 200 lobules, and that each lobule has one to three seminiferous tubules. The total number of tubules is between 400–600.

The testes are responsible for producing sperms and male hormones (mainly testosterone) that regulate reproductive organ development.

The seminiferous tubules of testis produce spermatozoa, which acquire their mobility in the epididymis. Primary spermatocytes are formed from spermatogonium, which divide into two secondary spermatocytes, which eventually divide into four spermatids. The process of spermatogenesis takes 74 ± 4 days.

The predominant androgen product produced by the testes is testosterone. The main site of testosterone synthesis is the Leydig cell of testis. Luteinising hormone (LH) binds to Leydig cells, stimulating the production of testosterone. In the adult testis, Leydig cells express aromatase (P450arom) and actively synthesise oestradiol. Inhibin, which down regulates FSH synthesis and inhibits its secretion, is synthesised by the Sertoli cell.

Lymphatic drainage: The testicular lymphatics pass along with the arteries to the para-aortic group of lymph nodes in the region of the renal arteries. Lymph from the vulva (in females), scrotum (in males) and lower limbs (in both) passes to the superficial and then on to the deep inguinal nodes.

Pelvic Organs: Part of the Gastrointestinal Tract

Rectum

The rectum extends from the level of the body of the third sacral vertebra to the anorectal line. The rectum varies from 10 cm to 15 cm in length, while the circumference varies from 15 cm at the rectosigmoid junction, to 35 cm or more at its widest ampullary portion.

The structure of the rectum is different from that of the colon in regards to the following parameters:

❏ There are no taenia coli
❏ There are no sacculations
❏ There are no appendices epiploicae.

Its upper end is continuous with the sigmoid colon. The lower end of the rectum lies a little below and in front of the tip of the coccyx and becomes continuous with the anal canal. The lower part of the rectum, which is wider than the upper part, is called the ampulla. The third sacral vertebra corresponds to the termination of a definite mesentery. It also acts as a marker for the following anatomical changes in the rectum:

❏ The point at which there is a change in the blood supply
❏ The level at which the tinea of the sigmoid spread out to reinforce the longitudinal muscle coat
❏ It corresponds to the site where the rectum narrows to join the sigmoid
❏ It marks the change in colour, the capillary pattern and the rugosity of the rectal mucosa.

Curves of the Rectum

The rectum has an anteroposterior curve corresponding to that of the sacrum and coccyx. The upper part of the rectum is directed backwards, and the lower part is directed forwards. The rectum also has three lateral curves. While passing downwards from the midline, the rectum deviates first to the right, then to the left and again to the right, finally returning to the middle line at its lower end.

Folds in the Rectum

The mucous membrane of the rectum shows a number of transverse folds. Usually three folds are present. The circular and longitudinal muscle coats may extend into these folds. Longitudinal folds may also be present.

Peritoneal Relations

The upper part of the rectum is covered with peritoneum in front and at the sides; the middle part is covered in front only; the lower part lies below the level of the rectovaginal pouch and therefore is devoid of peritoneal covering.

Relations

Posteriorly: The rectum rests posteriorly on the lower part of the sacrum and the coccyx, the right and left piriformis muscles, the right and left coccygeus muscles and the right and left levator ani muscles. Several nerves and vessels intervene between the rectum and these structures.

Anteriorly: In a male, the rectum is related to the urinary bladder, the seminal vesicles, the ductus deferentia and the lower ends of the ureters and the prostate.

In the female the rectum is related anteriorly to the vagina and the lower part of the uterus.

In both sexes the upper part of the rectum may be related anteriorly to the sigmoid colon and/or coils of ileum.

Laterally: In the upper part, the rectum may be related to the sigmoid colon and/or coils of ileum. In the lower part, the rectum is related to the right and left coccygei and the right and left levator ani muscles.

Blood Supply

Blood supply to the rectum is from the inferior mesenteric artery through its rectal branches.

Nerve Supply

Its parasympathetic supply is derived from the hypogastric plexus of S234 origin.

Lymphatic Drainage

The rectum and the upper two-thirds of the anal canal drain into the internal and common iliac nodes, the sacral nodes and along the superior arteries in to the pre-aortic nodes.

Anal Canal

Anal canal is the lowest part of the alimentary canal. Superiorly, it is continuous with the lower end of the rectum and inferiorly it opens to the exterior at the anus. The anal canal is about 4 cm in length. Therefore, it is noticeably narrower than the rectum. There is a sudden change in direction of the alimentary canal at the junction of the rectum with the anal canal. While the lower part of the rectum is directed downwards and forwards, the anal canal is directed downwards and backwards. The anorectal junction lies at the level of the pelvic diaphragm, with the rectum lying above the pelvic diaphragm in the true pelvis, and the anal canal lying below it in the perineum.

Interior of the Anal Canal

The upper 15 mm or so of the anal canal is lined by mucous membrane. This mucous membrane shows six to ten longitudinal folds, which are known as anal columns (also known as the columns of Morgagni). The lower ends of the anal columns are united to each other by short transverse folds of mucous membrane, called anal valves (of Ball). The anal valves together form a transverse line that runs all round the anal canal, resulting in the formation of the pectinate line.

The next 15 mm or so of the anal canal is also lined by mucous membrane known as the pecten or the transitional zone. The anal columns are not present in this region. The lower limit of the pecten often has a whitish appearance because of which it is referred to as the white line (of Hilton). **Figure 2.32** illustrates anatomy of the anal canal. The interior of anal canal is divided by the pectinate line and the Hilton's line into three areas:
1. Upper 15 mm above the pectinate/dentate line
2. Intermediate 15 mm (between the pectinate and Hilton's line)
3. Lower 8 mm: Anal verge.
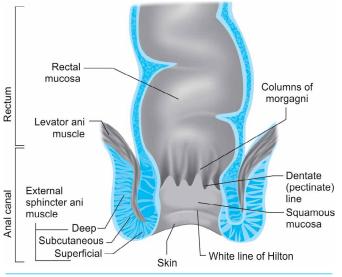
FIG. 2.32: Anatomy of the anal canal

The pectinate line has the following significance:
❏ Marks the mucocutaneous junction of the anal canal
❏ Corresponds with the position of anal valves
❏ Divides the anal canal into upper and lower areas, which are different in development, blood supply, lymph drainage and nerve supply **(Table 2.6)**.

On the other hand, the Hilton's white line has the following significance:
❏ It indicates the junction between non-keratinised stratified squamous epithelium in the upper part and the keratinised stratified squamous epithelium in the lower part.
❏ It serves as a colour contrast between the bluish pink area above and black skin below.
❏ It indicates lower end of the internal sphincter.
❏ Lunate fascia and anal fascia extend up to this line.
❏ Ischiorectal abscesses (when communicate with anal canal) open at or below the Hilton's line.

The Anal Musculature

The anal canal is surrounded by a number of sphincters, which help in keeping it closed except during defaecation. These sphincters keep the lateral walls of the anal canal approximated when the anal canal is empty, resulting in the formation of an anterior-posterior slit. These sphincters are as follows:

Internal anal sphincter: This is formed by thickening of the circular smooth muscle coat of the lower part of rectum. It surrounds the upper three-fourths of the anal canal and extends from the upper end of the anal canal up to the white line. Internally, it is separated from the mucous membrane by internal venous plexus. It is externally separated from the external sphincter muscle by a conjoint sheath derived from levator ani and the longitudinal muscles of the rectum.

Nerve supply: Superior hypogastric plexus and the pelvic splanchnic nerves.

Blood supply: Superior rectal artery (branch of inferior mesenteric artery); middle rectal artery (branch of internal iliac artery); and inferior rectal artery (branch of internal pudendal artery).

External anal sphincter: The external sphincter is a voluntary sphincter, which surrounds the entire length of the anal canal. This is made up of striated muscle, which comprises of the following three parts:
1. *Subcutaneous part*: This lies below the level of the white line, i.e. inferior to the level of the internal sphincter. The subcutaneous part forms a flat band around the anus. It is separated from the perianal skin by external venous plexus.
2. *Superficial part*: It is elliptical in shape and lies external to the lower part of the internal sphincter between the levels of the pectinate line and the white line. The fibres of this part are attached posteriorly to the coccyx and anococcygeal raphe, and anteriorly to the perineal body.
3. *Deep part*: This lies external to the upper half of the internal sphincter (above the level of the pectinate line). It is annular in shape and surrounds the anorectal junction. It has no bony attachment and is inserted into the perineal body.

Nerve supply to the external anal sphincter is by the inferior rectal branch of the pudendal nerve and perineal branch of the fourth sacral nerve.

Relations of the Anal Canal

Posteriorly: The anal canal is separated from the coccyx by a mass of fibromuscular tissue that is called the anococcygeal ligament (or body).

Anteriorly: In front of the anal canal there is another similar fibromuscular mass called the perineal body. The perineal body separates the anal canal from the membranous urethra and the bulb of the penis in the male; and from the vagina in the female.

TABLE 2.6	Distinction between the upper and lower areas of the anal canal	
Feature for distinction	**Above the pectinate line**	**Below the pectinate line**
Destination of the lymphatic drainage	Internal iliac group of lymph nodes via the pararectal group of lymph nodes	Below the Hilton's line, drainage is to the superficial inguinal group of lymph nodes
Epithelium	Columnar epithelium (this line represents the end of the part of the gut derived from the hindgut)	Stratified squamous epithelium (non-keratinised) until Hilton's line where the anal verge becomes continuous with the perianal skin containing keratinised epithelium
Embryological origin	Endoderm	Ectoderm
Artery	Superior rectal artery	Middle and inferior rectal artery
Vein	Superior rectal vein	Middle and inferior rectal vein
Haemorrhoid classification	Internal haemorrhoids (not painful)	External haemorrhoids (painful)
Nerves	Inferior hypogastric plexus Sympathetic fibres L1, L2 and parasympathetic fibres S2, S3, S4	Inferior rectal nerves

Laterally: Lateral to the anal canal on either side, there is a triangular depression called the ischiorectal fossa **(Fig. 2.33)**, which has a base directed to the surface of the perineum and its apex is at the line of meeting of the obturator and anal fascia. The boundaries of ischiorectal fossae are as follows:

Roof: The levator ani muscle forms the inner wall and roof of the ischiorectal fossae

Medially: The sphincter ani externus and the anal fascia

Laterally: The tuberosity of the ischium, the obturator fascia and the obturator internus muscle
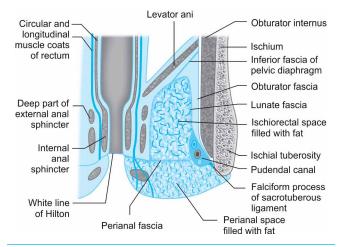
FIG. 2.33: Anatomy of the ischiorectal fossa

Anteriorly: The fascia of Colles covering the transversus perinei superficialis, and the inferior fascia of the urogenital diaphragm

Posteriorly: The gluteus maximus and the sacrotuberous ligament.

The contents of ischiorectal fossae are as follows:
- ❑ The inferior haemorrhoidal vessels and nerves (crossing the space transversely)
- ❑ The perineal and perforating cutaneous branches of the pudendal plexus (present at the back part)
- ❑ The posterior labial vessels and nerves (present in the forepart)
- ❑ The internal pudendal vessels and pudendal nerve (lying in Alcock's/pudendal canal on the lateral wall)
- ❑ The fossa is filled with fatty tissue across which, there are numerous fibrous bands extending from the sides.

Blood Supply to the Pelvis

Abdominal aorta divides at the level of the fourth lumbar vertebra into two common iliac vessels. These pass downwards and laterally and divide opposite the sacroiliac joint into two vessels: the internal iliac and the external iliac vessel. While the external iliac vessels mainly supply the lower extremity, the internal iliac vessels supply the pelvis.

The blood supply to the pelvis is mainly by the internal iliac artery, also known as the hypogastric artery **(Fig. 2.34)**. The internal iliac vessel arises at the bifurcation of the common iliac vessel opposite the lumbosacral articulation
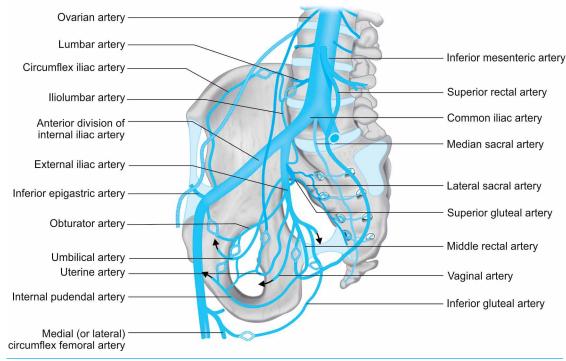
FIG. 2.34: Blood supply to the pelvis

and passes downwards to the upper margin of the greater sciatic foramen where it divides into two large trunks: anterior and posterior, both of which give rise to various branches **(Table 2.7)**. The place of division of hypogastric artery varies between the upper margin of sacrum and the upper border of the greater sciatic foramen.

Relations of the internal iliac vessels are as follows:
- ❏ *Anterior*: Ureter
- ❏ *Posterior*: Internal iliac vein, lumbosacral trunk and piriformis muscle
- ❏ *Lateral*: External iliac vein (near its origin); obturator nerve (lower down).

Other significant blood vessels supplying the pelvis are as follows:

Median Sacral Artery

The median sacral artery arises directly from the abdominal aorta at the point where it bifurcates into the two common iliac arteries. It descends over the L4 and L5 vertebrae as well as the sacrum and coccyx to supply the sacrum. At the level of the sacroiliac joint it is crossed anteriorly by the ureter.

Superior Rectal Artery and Vein

This artery is a branch of the inferior mesenteric artery and supplies the superior two-thirds of the rectum.

Ovarian Artery and Vein

These vessels are found only in the female. The analogous vessel in the male is the testicular artery. The ovarian artery is a branch of the abdominal aorta that supplies the ovary and the uterine tube.

External Pudendal Artery

The external pudendal artery arises from the femoral artery. It supplies the skin over the pubic symphysis (the mons pubis in the female) and gives off anterior scrotal and labial arteries and the dorsal artery of the penis and clitoris.

TABLE 2.7   Branches of the internal iliac artery

Anterior trunk		
Branch	**Sub-branch**	**Area of blood supply**
Superior vesical (umbilical) artery	Umbilical artery may give two vessels: artery to vas deferens and superior vesical artery. Sometimes, the superior vesical artery may branch out directly from the anterior trunk	Superior vesical artery supplies the bladder
Middle rectal artery	—	Rectum
Inferior vesical artery	—	Supplies the posterior bladder, seminal vesicle, and prostate
Obturator artery	—	Gives rise to anterior and posterior branches, which encircle the margin of the obturator foramen. They supply the medial thigh and hip
Internal pudendal artery	• Inferior rectal artery • Perineal artery • Posterior labial/scrotal branches • Artery to the bulb of vestibule/bulb of penis • Deep artery of the penis/clitoris • Dorsal artery of penis/clitoris • Urethral artery • Artery of the urethral bulb	These supply the anal canal, perineum and its muscles, scrotum (or labia), bulb of the penis (or vestibule), and urethra
Inferior gluteal artery	—	Supply the gluteal muscles
Uterine artery (or the artery of ductus deferens)	Vaginal branch	Uterus
Vaginal artery	—	Vagina
Posterior trunk		
Branch	**Sub-branch**	**Area of blood supply**
Iliolumbar vessels	Iliac branches Lumbar branches	Bones and muscles in the iliac fossa
Lateral sacral vessels	Superior and inferior branches	Branches to the sacrum and coccyx
Superior gluteal vessels	—	Supply the gluteal muscles

External Iliac Artery

The external iliac artery arises from the common iliac artery and gives off two branches: the inferior epigastric artery and the deep circumflex iliac artery. The deep circumflex iliac artery and the corresponding vein runs along the internal surface of the ala of the ilium to supply the muscles located there. At its origin it is crossed by the ovarian vessels in the female, and occasionally by the ureter. The external iliac artery passes obliquely downward and laterally along the medial border of the psoas major, from the bifurcation of the common iliac to a point beneath the inguinal ligament. The external iliac artery continues as the femoral artery below the inguinal ligament. At the upper part of its course, the external iliac vein lies partly behind it, but lower down lies entirely to its medial side.

Nerve Supply to the Pelvis and the Lower Limbs

Pelvic Nerves

The pelvis is innervated mainly by the sacral and coccygeal spinal nerves and the pelvic part of the autonomic nervous system **(Figs 2.35A and B)**. The piriformis and coccygeus muscles form a bed for the sacral and coccygeal nerve plexuses. The anterior rami of the S2 and S3 nerves emerge between the digitations of these muscles.

The sacral plexus **(Fig. 2.36)**, which lies in the front of piriformis, supplies most of the pelvic structures, buttocks and lower limbs. It is formed by the lumbosacral trunk, the ventral rami of S1–S3 and the upper division of S4. The lumbosacral trunk comprises of the anterior divisions of the lumbar, sacral nerves and the coccygeal nerve.
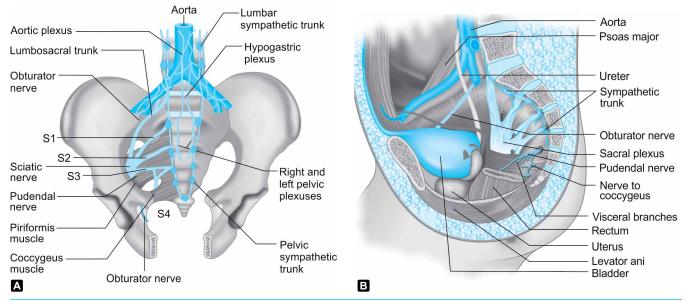
Most of the innervation of the perineum is by the pudendal nerve (S2–S4). It contains motor, sensory (pain and reflex), and postganglionic sympathetic fibres.

The pudendal nerve traverses the greater sciatic foramen below the piriformis, crosses the back of the ischial spine, and enters the perineum through the lesser sciatic foramen.

It traverses the pudendal canal in the lateral wall of the ischiorectal fossa, gives off the inferior rectal nerve, and divides into the perineal nerve and the dorsal nerve of the penis (or clitoris). The perineal nerve divides into a deep branch to perineal muscles and a superficial branch to the scrotum (or labium majus).

Pelvic Part of Autonomic Nervous System: Hypogastric Plexus

Sympathetic fibres reach the pelvis by downward continuations of the sympathetic trunk and of the aortic plexus. In front of the sacrum, the sympathetic trunks consist largely of preganglionic fibres and present three or four ganglia each. This forms the hypogastric plexus of nerves, which supplies the viscera of pelvic cavity. This plexus is situated in front of the last lumbar vertebra and sacral promontory. It is formed by the presacral nerve, which lies in front of sacral promontory and divides into two hypogastric nerves which pass downwards and laterally along the pelvic wall. They help to form the inferior hypogastric plexus, which is a diffuse plexus that lies in the region of uterosacral ligaments. The inferior hypogastric plexus is formed from the fibres of the sacral splanchnic nerves, pelvic splanchnic nerves and hypogastric nerves. Inferior hypogastric plexus supplies the viscera of the pelvis. This plexus also receives pelvic splanchnic fibres from parasympathetic system comprising of ventral primary

FIGS 2.35A AND B: Nerve supply to the pelvis: (A) Frontal view; (B) Lateral view
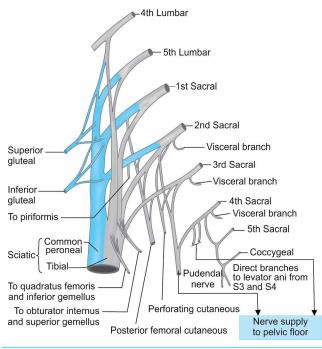
FIG. 2.36: The sacral plexus

rami of S2–S4. These fibres control micturition, defaecation and erection.

The hypogastric plexus divides into two lateral portions called the pelvic plexus. These are situated at the sides of rectum and vagina in females and supply the viscera of pelvis. The cervix is surrounded by a rich plexus of nerves called the Frankenhauser plexus. The ovaries derive their nerve supply from the coeliac and renal ganglia. The lower vagina, clitoris, posterior part of labia majora and the perineum are supplied by the pudendal nerve (S2–S4). The ilioinguinal nerve (L1) and the genital branch of the genitofemoral nerve (L1 and L2) supply mons pubis and upper and outer aspect of labia majora and perineum. The rectal plexus is formed from the posterior division of the hypogastric plexus.

The Pudendal Nerve

The pudendal nerve arises from the anterior rami of the second to fourth sacral roots. These form a trunk before leaving the pelvis via the greater sciatic foramen. It passes immediately behind the ischial spine and swings forward to enter the perineum via the lesser sciatic foramen. The nerve passes through the ischiorectal fossa where it gives off its terminal branches. The pudendal nerve and internal pudendal vessels are embedded in the pudendal canal in the lateral wall of the ischiorectal fossa. The pudendal canal runs medial to the obturator internus and is in close contact with the obturator fascia.

The pudendal nerve courses through the following five regions:
1. The sacral region where the three sacral segments fuse
2. Gluteal region

3. Pudendal canal which begins at the posterior border of the ischiorectal fossa and ends at the posterior edge of the urogenital diaphragm
4. The deep perineal space which is the fascial space between the superior and inferior fasciae of the urogenital diaphragm
5. The superficial perineal space.

The pudendal nerve divides into three branches:
1. *The dorsal nerve of the clitoris*: The dorsal nerve of the clitoris supplies the skin surrounding this structure. It supplies levator ani. Dorsal nerve of the clitoris and the inferior hypogastric plexus also provide sensory innervation for the peritoneum in the pouch of Douglas.
2. *The inferior haemorrhoidal/rectal nerve*: The inferior rectal nerve supplies the external anal sphincter and the perianal skin. On the other hand, the internal anal sphincter is supplied by the autonomic nervous system.
3. *The perineal nerves*: The perineal nerve innervates the sphincter urethrae and other muscles of the anterior compartment (ischiocavernosus, bulbospongiosus, superficial and deep transverse perineal) via a deep branch, and the skin over the posterior two-thirds of the labium majus and the mucous membrane of the labia minora via its superficial branch.

Clinical Application

❑ The pudendal nerve blocks interrupt the second to fourth sacral nerves and will be inadequate for a forceps delivery or to block the pain of the second stage of labour.
❑ Paracervical blocks should be avoided in pregnancy as they are associated with foetal bradycardias.

Lymphatic Drainage of Lower Limb and Pelvis

Lymph Nodes of Pelvis

Lymph nodes of the pelvic region, which drain the female genital organs, comprise of the groups as described in **Figure 2.37**. The cervix drains primarily into the external and internal iliac group of lymph nodes, whereas the body of the uterus drains mainly into the external iliac and lumbar nodes. Various lymph node groups which drain the pelvic region are as follow.

Inguinal Group

The inguinal group of lymph nodes comprises of a vertical and a horizontal group. The horizontal group is also known as the superficial inguinal group and receives afferent lymphatic vessels from perineum, buttocks, the big toe, feet, abdominal wall below the umbilicus, vulva and anus (below the pectinate line). This group of lymph nodes drains into the deep inguinal group.

The vertical group of lymph nodes is also known as the deep femoral group and follows the saphenous and femoral
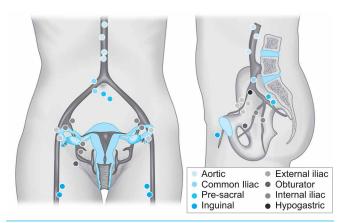
FIG. 2.37: Lymph nodes of the pelvic region

veins. The superior-most lymph node of this group is located under the inguinal ligament and is known as the Cloquet's node or the gland of Rosenmüller.

Hypogastric Group

The hypogastric group or the glands of parametrium drain the lymphatics from the cervix, bladder, upper-third of vagina and greater part of the body of the uterus. This group of glands may be extensively involved in the cases of cancer of cervix and vagina. These glands are present below the bifurcation of the common iliac group.

External Iliac Group

They lie above the pelvic brim along the external iliac vessels. They receive lymph from inguinal group of lymph nodes. They also receive lymph from the pelvic viscera, especially the upper parts of the pelvic organs placed anteriorly and in the middle. These lymph nodes eventually drain into the common iliac group of lymph nodes.

Internal Iliac Group

This group of lymph nodes is clustered around the anterior and posterior divisions of the internal iliac vessels and the origin of gluteal artery. This group of lymph nodes also receives drainage from the inferior pelvic viscera, deep perineum and gluteal region and ultimately drains into the common iliac group of lymph nodes.

Sacral Group

These lymph nodes lie in the concavity of the sacrum, adjacent to the median sacral vessels. They receive the lymph drainage from the posterior part of the pelvic viscera, particularly the uterine cervix and the upper-third of the vagina. They ultimately drain either into internal or common iliac group of lymph nodes. There are two types of sacral lymph nodes: (1) a medial group lying in front of the sacral promontory and (2) a lateral group lying lateral to the rectum.

Common Iliac Group of Lymph Nodes

They lie in the upper part of the pelvis and receive drainage from the three main groups of lymph nodes, i.e. (1) external iliac group, (2) internal iliac group and (3) the sacral group. This group of lymph nodes forms a common route for drainage of the pelvic organs. The lymphatic drainage passes from this group of lymph nodes to the lumbar (caudal/aortic group) lymph nodes. Sometimes, the common iliac group of lymph nodes may receive some direct drainage from pelvic organs such as neck of bladder and inferior vagina.

Pararectal Group of Lymph Nodes

This is minor group of lymph nodes which occupies the connective tissue along the branches of internal iliac vessels. Both the primary and the minor group of pelvic nodes are highly interconnected so that many nodes can be removed without disturbing drainage. These interconnections also allow the cancerous cells to spread in any direction in relation to the pelvic and abdominal viscera.

Lumbar Group

This comprises of mainly two groups of lymph glands:
1. The inferior group and
2. The superior group.

The inferior group lies in front of the aorta below the origin of inferior mesenteric artery. The superior group lies near the origin of ovarian artery and receives lymphatics from the ovaries and fallopian tubes as well as from the inferior lumbar group. The lymphatics from the uterine fundus also pass to this group via the ovarian lymphatics.

Anatomy of the Urinary Tract

Urinary Bladder

The urinary bladder acts as a reservoir of urine, which is formed continuously in the kidneys and is conveyed to the urinary bladder through the ureters. When the bladder is distended beyond a certain limit, the desire for micturition is felt. This limit is usually reached when the bladder contains about 300 mL of urine. The maximum capacity of the urinary bladder is about 500 mL.

In an adult the urinary bladder lies in the pelvis. However, when distended with urine, part of it extends above the level of the pubic symphysis and comes in contact with the anterior abdominal wall. It is important to note that as the distended bladder ascends the fold of peritoneum passing from the anterior abdominal wall, the superior surface of the bladder also rises so that no peritoneum intervenes

between a distended bladder and the anterior abdominal wall. In the infant, the bladder lies above the level of the pubic symphysis, i.e. it is an abdominal organ rather than a pelvic one. The bladder in a young child is separated from the anterior abdominal cavity by a fold of peritoneum. On the other hand, there is no peritoneal covering in the retropubic space, when the bladder lies in the pelvis.

The empty urinary bladder has four surfaces each of which is triangular, giving it shape of a tetrahedron **(Fig. 2.38)**. The posterior surface of the bladder is also called the base or fundus. The superior surface faces upwards. The right and left inferolateral surfaces face downwards, laterally and forwards. They meet the superior surface at the right and left lateral borders. Posteriorly they meet the lateral margins of the base. Anteriorly it narrows to form the apex of the bladder. The apex of the bladder is joined to the umbilicus by the remains of the urachus, which forms the middle umbilical ligament. The peritoneum is carried by the apex of the bladder on to the abdominal wall, resulting in the formation of the middle umbilical fold. The right and left ureters join the urinary bladder at its posterolateral angles. The lowest part of the bladder is called the neck. The urethra emerges from the bladder neck.

Relations

Superiorly: The superior surface of the bladder is separated by peritoneum from part of the sigmoid colon and from coils of small intestine.

Posteriorly: In the female, uterus and the upper part of the vagina lie behind the bladder. The bladder is separated from the anterior surface of the body of the uterus by the vesicouterine excavation, but below the level of this excavation it is connected to the front of the cervix uteri and the upper part of the anterior wall of the vagina by areolar tissue.

Laterally: The bladder is connected to the lateral pelvic wall by the fascia endopelvina.

Peritoneal Relations of the Bladder

Figure 2.39 illustrates median section of the female pelvis showing peritoneal reflection over the bladder. The superior surface of the urinary bladder is covered by peritoneum. Traced anteriorly this peritoneum becomes continuous with that lining the anterior abdominal wall. In the midline this peritoneum is raised into a fold called the median umbilical fold due to the presence of the median umbilical ligament here. Traced laterally the peritoneum of the superior surface is reflected on to the lateral pelvic wall. This peritoneum is referred to as the false lateral ligament of the bladder. Traced posteriorly, the peritoneum on the superior surface of the bladder passes on to the upper part of the base from where it is reflected on to the front of the uterus. The peritoneum lined depression between the urinary bladder and the uterus is known as the vesicouterine pouch. The inferolateral surfaces of the urinary bladder are not lined by peritoneum.

Embryology

The bladder is derived from two sources, the cloaca and mesonephric ducts. The primitive cloaca is divided by the urorectal septum into the urogenital sinus and rectum. The bladder largely develops from the vesicle part of the urogenital sinus. The urogenital sinus may be divided into three component parts: the first of these is the cranial portion, which is continuous with the allantois and forms the bladder proper. The pelvic part of the sinus forms the prostatic urethra and epithelium as well as the membranous urethra and bulbourethral glands in the male and the membranous urethra and part of the vagina in females. The mesonephric ducts are drawn into the floor of the bladder as it expands to form the trigone. The bladder epithelium is derived from the endoderm of the urogenital sinus, whereas the ureter is derived from mesoderm.

Blood Supply

It has a blood supply from the internal iliac artery (via the superior and inferior vesical arteries). In the female, the
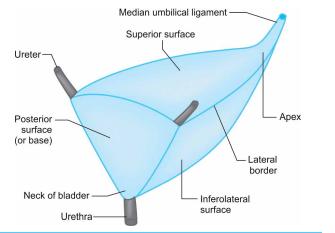
FIG. 2.38: Various surfaces of the urinary bladder
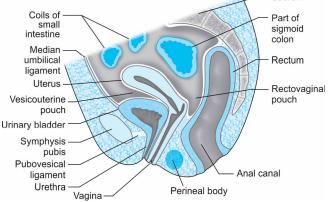
FIG. 2.39: Median section of the female pelvis showing peritoneal reflection

inferior vesical artery is replaced by the vaginal artery and the uterine artery also gives branches to the bladder.

Venous Drainage

Venous drainage is to the internal iliac veins. The vesical plexus drains into the internal iliac vein via the superior and inferior vesical plexuses.

Nerve Supply

The nerves of the bladder are derived from the following:
❏ Fine medullated fibres from the third and fourth sacral nerves, and
❏ Non-medullated fibres from the hypogastric plexus.

Sympathetic fibres of the bladder arise from the hypogastric plexus and nerves, whereas the parasympathetic fibres arise from pelvic splanchnic nerves and the inferior hypogastric plexus. Sympathetic fibres are derived from spinal segments T10 to L2.

Lymphatic Drainage

Lymphatic drainage is to the external iliac group of lymph nodes.

Ureter

The ureter (right or left) is a long tube that connects the lower end of the renal pelvis with the urinary bladder. Each ureter is about 25–30 cm (10–12 inches) long. The upper half of this length lies on the posterior abdominal wall and the lower half in the true pelvis. The ureter is composed of both longitudinal and circular muscle fibres. The longitudinal muscular fibres are only present in the part traversing the bladder wall. Rest of the ureter comprises of circular muscular fibres. Waldeyer's sheath is an investment of muscle surrounding the ureteral opening in the bladder wall. Ureter is lined by transitional epithelium. It is more dilated on the right side in pregnancy due to pressure at the pelvic brim and high progesterone levels. The ureter can be seen lying on the tips of the transverse processes of the lumbar vertebrae on an intravenous urogram.

Abdominal Part of the Ureter

The abdominal part of each ureter runs downwards (with a slight medial inclination). At the brim of the pelvis, the ureter crosses the upper end of the external iliac artery (and vein), and comes to lie on the lateral wall of the pelvis. Here, it runs backwards and laterally. Finally, it leaves the pelvic wall and turns medially and forwards to reach the posterolateral part of the urinary bladder.

Relationship of the Abdominal Part of the Ureter

The relations of the abdominal part of the ureter are different on the right and left sides **(Fig. 2.40)**.

Relations of Right Ureter

Anteriorly:
❏ The abdominal part of the right ureter is overlapped at its upper end by the descending part of the duodenum
❏ Lower down it is crossed by the terminal ileum and by the root of the mesentery
❏ Several smaller structures cross the right ureter from medial to lateral side. These include the following:
  • The testicular or ovarian vessels
  • The right colic and ileocolic branches of the superior mesenteric artery
  • The terminal part of the superior mesenteric artery, itself (in the root of the mesentery)
  • The arteries are also accompanied by the corresponding veins.

Posteriorly:
❏ Psoas major muscle
❏ The genitofemoral nerve

Medially: The inferior vena cava lies a short distance medial to the right ureter.

Relations of Left Ureter

Anteriorly:
❏ The abdominal part of the left ureter is crossed (near the brim of the pelvis) by the sigmoid colon. The left ureter then passes deep to the apex of the V-shaped attachment of the sigmoid mesocolon.
❏ It is crossed by the following structures from medial to lateral side:
  • The testicular or ovarian vessels
  • The left colic branches of the inferior mesenteric artery.

Medially: The inferior mesenteric vein is placed parallel to the left ureter, a little to its medial side.

Pelvic Part of the Ureter

Figure 2.41 illustrates the course and relationship of the pelvic part of the female ureter. At the brim of the pelvis the ureter crosses the upper end of the external iliac artery (and vein), and comes to lie on the lateral wall of the pelvis. Here it runs backwards and laterally. After entering the pelvis the ureter runs across the sacroiliac joint, and the anterior border of the greater sciatic notch to reach the ischial spine. It then runs medialward and forward on the lateral aspect of the cervix uteri and upper part of the vagina to reach the fundus of the bladder. In this part of its course it is accompanied for about 2.5 cm by the uterine artery, which then crosses in front of the ureter and ascends between the two layers of the broad ligament. The ureter is distant about 2 cm from the side of the cervix of the uterus. The point of termination of the bladder corresponds to the pubic tubercle.
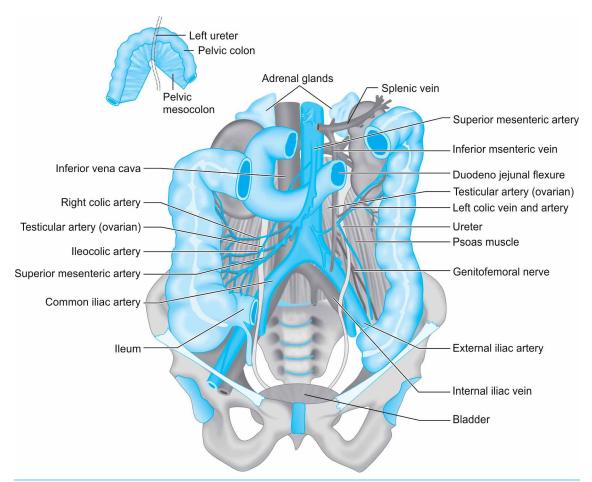
FIG. 2.40: Course and relationship of the abdominal part of the female ureter
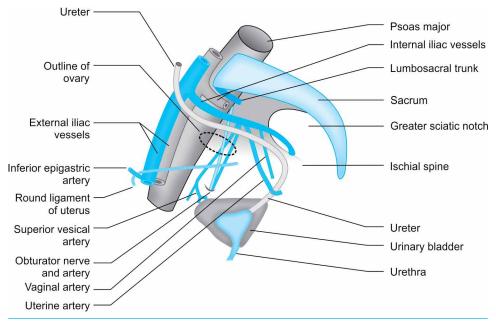
FIG. 2.41: Course and relationship of the pelvic part of the female ureter

Relations of Pelvic Part of Ureter

These are similar on the right and left sides.

Posteriorly:

1. As the ureter runs backwards and laterally on the lateral wall of the pelvis it lies on the fascia covering the obturator internus. Here the ureter crosses several structures that lie between it and the lateral pelvic wall. In the female, these structures include:
   - The superior vesical artery
   - The obturator nerve, artery and vein
   - The vaginal artery
   - The uterine artery.
2. In both the male and female, the ureter is related posteriorly to the internal iliac vessels that separate it from the lumbosacral trunk and from the sacroiliac joint.

Anterior: In the female, the ovary lies immediately in front of the ureter. Here the ureter forms the posterior wall of the ovarian fossa in which the ovary lies. As the ureter leaves the lateral pelvic wall and turns anteromedially, it lies over the levator ani, but is separated from it by a mass of fat. In the female, the ureter passes a short distance lateral to the cervix of the uterus (just above the lateral fornix of the vagina), and then passes anterior to the vagina to reach the urinary bladder. Lateral to the cervix, the ureter is crossed by the uterine artery and the broad ligament.

The terminal part of the ureter passes obliquely through the thickness of the wall of the urinary bladder to open into its posterior wall. The openings lie at the lateral angles of a triangular area on the posterior wall of the urinary bladder, which is known as the trigone.

Embryology

Ureter is of mesodermal origin; it is derived by a process of budding of the caudal end of the mesonephric duct.

Blood Supply

The blood supply to the ureters comes in an above downwards direction from the renal artery, abdominal aorta, ovarian artery, common and internal iliac, vesical and uterine arteries. The vein corresponds to the arteries.

Lymphatic Drainage

The lymphatic drainage of the ureter is as follows:

- ❏ The upper abdominal part drains to lateral aortic nodes
- ❏ The lower abdominal part drains to common iliac nodes
- ❏ The pelvic part drains into external iliac and internal iliac nodes.

Female Urethra

The female urethra is a narrow membranous canal about 4 cm long, extending from the internal to the external urethral orifice. This is much shorter in comparison to the male urethra, which is about 20 cm long. Throughout its length the urethra is closely related to the anterior wall of the vagina. Its lining is composed of stratified squamous epithelium, which becomes transitional near the bladder. The urethra consists of three coats: muscular, erectile, and mucous, the muscular layer being a continuation of that of the bladder.

Both in the male and in the female the urethra is surrounded by an internal sphincter, the sphincter vesicae; and by an external sphincter, the sphincter urethrae. The sphincter vesica is composed of a ring of smooth muscle surrounding the urethra at its junction with the bladder. This sphincter is involuntary and is supplied by autonomic nerves. On the other hand, the sphincter urethra surrounds the urethra as it passes through the deep perineal space. It is present between the superior and inferior fascia of the urogenital diaphragm and is composed of striated muscle fibres. It is voluntary in nature and is supplied by the perineal branch of the pudendal nerve.

Relations

Urethra is placed behind the symphysis pubis, embedded in the anterior wall of the vagina, and its direction is obliquely downward and forward; it is slightly curved with the concavity directed forward.

Adrenal Glands

The adrenal glands are paired retroperitoneal organs. As the name implies the right and left suprarenal glands lie in close relationship to the upper poles of the corresponding kidneys. They are enclosed with the kidney in the renal fascia, but lie outside the renal capsule. Each suprarenal gland is relatively flat and has an anterior and a posterior surface. When seen from the front the right suprarenal gland is triangular. It has medial, lateral and inferior borders. The left suprarenal gland is semilunar. It has a convex medial margin and a concave lateral margin. Each gland is about 50 mm in vertical diameter, about 30 mm from side to side, and about 10 mm from front to back. Each gland weighs about 5 g. Each gland is made up of a superficial layer, the cortex, and a deeper part called the medulla. The cortex forms nearly 80% of the volume of the gland. The volume of the medulla is about one-tenth of the cortex. Cells of adrenal cortex can be divided into three regions:

1. *Zona glomerulosa*: This region produces the hormone aldosterone, which helps in maintaining water and electrolyte balance in the tissues.
2. *Zona fasciculata*: This region produces hydrocortisone and related compounds which help in the maintenance of carbohydrate balance in the body.
3. *Zona reticularis*: This region produces sex hormones including oestrogens, progesterone and androgens.

Adrenal medulla contains chromaffin cells which secrete hormones such as adrenaline and noradrenaline into the blood. These cells of the suprarenal medulla are modified postganglionic sympathetic neurons.

Embryological Origin

The adrenal cortex has embryological origin from the mesoderm, whereas the adrenal medulla is derived from the neural crest cells.

Blood Supply

Each suprarenal gland receives three suprarenal arteries:
1. *Superior adrenal artery*: This arises from the corresponding inferior phrenic artery.
2. *Middle adrenal artery*: This arises from the abdominal aorta.
3. *Inferior adrenal artery*: This arises from the corresponding renal artery.

Venous Drainage

Each adrenal gland on either side is drained by one vein. On the right side, it drains into the inferior vena cava, and on the left side into the left renal vein.

Lymphatic Drainage

Lymphatics from the adrenal glands drain into the lateral aortic group of nodes.

Nerve Supply

The medulla of the suprarenal gland receives numerous preganglionic sympathetic nerves.

Anatomy of the Thigh

Femoral Triangle

The femoral triangle is the name given to an area of the anterior aspect of the thigh formed as different muscles and ligaments cross each other producing an inverted triangular shape. This is the region on the front of thigh, lying medial to the upper part of the sartorius.

This can be considered as a region of importance, because it contains several vessels and nerves.

Boundaries (Fig. 2.42)

Upper boundary or base: The inguinal ligament

Lateral boundary: The medial margin of the sartorius

Medial boundary: The medial margin of the adductor longus

Apex of the triangle: The apex of the triangle, directed inferiorly, lies where the medial and lateral borders meet.

Roof of the triangle: This is formed by the fasciae over the region, and the superficial structures lying within them, which include the following:
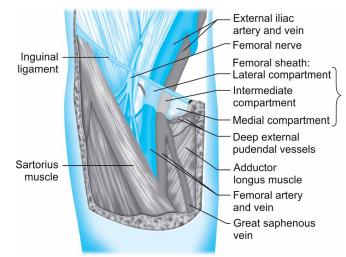
FIG. 2.42: Boundaries of the femoral triangle

❏ The saphenous opening
❏ The cribriform fascia
❏ The terminal part of the saphenous vein
❏ The superficial inguinal lymph nodes.

Contents of the Femoral Triangle

❏ *Femoral artery*: The artery running down the middle of the femoral triangle is the femoral artery. The femoral artery lies at the midinguinal point, which is midway between the pubic symphysis and anterior superior iliac spine and is the inferior extremity of the midclavicular line.
❏ *Femoral vein*: Femoral vein lies medial to the artery
❏ *Femoral nerve*: A short distance lateral to the artery is the trunk of the femoral nerve.

[Medial to lateral, these structures can be described with a mnemonic "van" (vein, artery, nerve)].

At the upper end of the femoral triangle, femoral vein lies medial to the femoral artery. However, at the apex of the femoral triangle the vein lies behind the artery. The great saphenous vein joins the femoral vein. Other tributaries of the femoral vein correspond to the branches of femoral artery.

Femoral Nerve

Femoral nerve is the largest branch of the lumbar plexus formed from the nerve roots L2 to L4. The nerve descends from the lumbar plexus in the abdomen through the psoas major muscle. It then travels through the pelvis to the midpoint of the inguinal ligament. It traverses behind the inguinal ligament into the thigh where it splits into anterior and posterior division. It then passes through the femoral triangle lateral to the femoral vessels (enclosed in the femoral sheath). The femoral nerve may be injured by surgeries to the femoral triangle (such as femoral embolectomy, femoro-popliteal bypass or femoral

aneurysm repair), massive haematoma within the thigh, traction during surgery or any form of trauma (e.g. gunshot wounds to the femoral triangle). Injury to the femoral nerve causes weakness of the quadriceps muscle. This may be associated with difficulty in extending the knees. The knee may give way on walking and the patient has difficulty climbing stairs. There is numbness over the anterior aspect of the thigh and medial region of the leg. The knee jerk is reduced or absent.

Branches of Femoral Nerve

After a short course, the femoral nerve divides into anterior and posterior divisions, each of which divides into a number of branches as follows:
- *Muscular branches*: The posterior division gives off several muscular branches. These are:
  - Branch to the rectus femoris
  - Branch to vastus lateralis
  - Branch to vastus intermedius
  - Branch to vastus medialis.
- *The intermediate cutaneous nerve of the thigh*: Anterior division of the femoral nerve gives off a stem that supplies the sartorius muscle and then continues as the intermediate cutaneous nerve of the thigh.
- *The medial cutaneous nerve of the thigh*: Anterior division of the femoral nerve also gives off the medial cutaneous nerve of the thigh. This nerve crosses anterior to the femoral vessels in the lower part of the femoral triangle.
- *The saphenous nerve*: This is a terminal, sensory branch of femoral nerve. It runs downwards lateral to the profunda femoris artery. It then continues with the femoral vein and artery through the adductor canal. The saphenous nerve runs down the medial side of the leg and supplies the medial side of the calf as far as the medial malleolus; it terminates in the region of the ball of the big toe and may supply the medial side of the dorsum of the foot.

Branches of Femoral Artery

The femoral artery gives off three branches, just below the inguinal ligament, as it lies in the femoral triangle. They pass through the cribriform fascia to become superficial and include the following:
- The superficial circumflex iliac artery
- The superficial epigastric artery
- The superficial external pudendal artery.

A short distance below the inguinal ligament, the femoral artery gives off two branches.
1. *The deep external pudendal artery*: This artery arises from the medial side of the femoral artery and runs medially over the floor of femoral triangle.
2. *The profunda femoris artery*: This is the largest branch of the femoral artery and is also the deep artery of the

thigh. This artery arises from the posterolateral aspect of the femoral artery, and runs downwards lateral to the femoral artery. The profunda femoris artery gives off two major branches: medial and lateral circumflex femoral arteries.

Small Structures in the Femoral Triangle

Some smaller structures present in the femoral triangle are as follows:
- *Nerve to pectineus*: It lies medial to the femoral nerve. This nerve arises from the femoral nerve within the pelvis and enters the thigh by passing deep to the inguinal ligament. It passes medially deep to the femoral artery to reach the pectineus.
- *Femoral branch of the genitofemoral nerve:* The genitofemoral nerve originates from the upper L1-2 segments of the lumbar plexus. It passes downwards and pierces the psoas major muscle to emerge on its anterior surface. It continues to run downwards and divides into two branches: the genital branch and the femoral branch. The genital branch passes through the deep inguinal ring to enter the inguinal canal, while the femoral branch runs on the external iliac artery to pass beneath the inguinal ligament. It eventually enters the femoral sheath, which it pierces to supply the skin over femoral triangle. Structures which cross the genitofemoral nerve are tabulated in **Table 2.8**.
- *Lateral cutaneous nerve of the thigh*: This nerve is seen near the lateral angle of the femoral triangle.

Femoral Sheath

Femoral sheath can be defined as the downward prolongation of the fascia lining the abdomen with fascia transversalis extending anteriorly in front of femoral vein and fascia iliaca posteriorly. The sheath assumes the form of a short funnel which is wide superiorly and inferiorly fuses with the fascial investment of femoral vein about 4 cm below the inguinal ligament. Lateral wall of the sheath is vertical and is perforated by the ilioinguinal nerve; medial wall is directed obliquely downwards and laterally and is pierced by the great saphenous vein and some lymphatic vessels.

The sheath is divided by two vertical partitions between its anterior and posterior wall.

TABLE 2.8	Structures crossing the genitofemoral nerve
Right side	**Left side**
Ureter	Ureter
Ovarian/testicular vessels	Ovarian/testicular vessels
Iliocolic artery	Left inferior colic artery
Mesentery of the small intestine	Inferior mesenteric vein
Right infracolic compartment	Left infracolic compartment
Lumbar plexus	

Contents

The lateral compartment: Femoral artery and the genital branch of genitofemoral nerve.

The intermediate compartment: Femoral vein

The medial compartment: Lymphatic vessels and a lymph gland embedded in a small amount of areolar tissue.

The medial compartment is the smallest and is also known as the femoral canal. The femoral canal is conical and measures about 1.25 cm in length. Its base is directed upwards and is named as the femoral ring.

Femoral Ring

Femoral ring is the abdominal opening of the femoral canal and includes the medial compartment of the femoral canal in the lower abdomen.

Boundaries of Femoral Ring

❑ *Anteriorly*: Inguinal ligament
❑ *Medially*: Crescentic base of lacunar ligament
❑ *Laterally*: Fibrous septum on the medial side of femoral vein
❑ *Posteriorly*: Pectineal ligament and pectineus muscle. It is not lined by peritoneum.

The contents of the femoral ring are Cloquet's node and lymphatics.

The femoral ring is the main site for a femoral hernia where the lining of the hernia would be peritoneum.

Adductor Canal

The space deep to the sartorius, over the middle one-third of the thigh, is called the adductor canal. It is also known as the subsartorial canal.

Boundaries

Anterior: The vastus medialis

Posterior: The adductor longus (above) and the adductor magnus (below)

Medial: A strong fibrous membrane lying deep to the sartorius.

Contents of the Canal

❑ *The femoral artery and vein*: The vein lies posterior to the femoral artery in the upper part of the canal, and lateral (deep) to it in the lower part. At the lower end of the canal, the femoral vessels pass through a large aperture in the aponeurosis of the adductor magnus (to reach the popliteal fossa).
❑ *The saphenous nerve*: The saphenous nerve runs along the femoral artery gradually crossing it from lateral to

medial side. At the lower end of the canal, the nerve pierces the fibrous roof to become superficial.
❑ *The subsartorial plexus of nerves*: This plexus lies over the fascia forming the roof of the adductor canal. This plexus also receives contributions from the medial cutaneous nerve of the thigh, the saphenous nerve, and the anterior division of the obturator nerve.

Femoral Artery

Femoral artery is the continuation of the external iliac artery (which is the terminal branch of the abdominal aorta). The branches of the femoral artery **(Fig. 2.43)** are as follows:
❑ Superficial circumflex iliac artery
❑ Superficial epigastric artery
❑ Superficial external pudendal artery
❑ Deep external pudendal artery
❑ Profunda femoris
❑ Descending genicular artery (is a small branch that arises from the femoral artery near its termination within the adductor canal).

The profunda femoris artery gives rise to the following three branches:
1. Perforating branches (three or four in number)
2. Lateral circumflex femoral artery
3. Medial circumflex femoral artery.

After exiting the femoral triangle, femoral artery continues down the anterior surface of the thigh via adductor canal. The adductor canal ends at an opening in the adductor magnus called the adductor hiatus. The femoral artery moves through this opening into the posterior compartment of the thigh proximal to the knee. Here the femoral artery is known as the popliteal artery.

Great Saphenous Vein

Figure 2.44 illustrates great saphenous vein and its tributaries. Each lower limb has two large superficial veins, the great (or long) saphenous vein; and the small (or short) saphenous vein. The great saphenous vein is a continuation of the medial marginal vein of the foot. It begins from the dorsal venous arch from where it passes anterior to the medial malleolus of the ankle and enters the medial side of the leg. Ascending on the medial side of the leg, it crosses the medial side of the knee joint, and ascends on the medial side of the thigh. Before passing over the medial epicondyle of the femur at the knee joint, it merges with several superficial veins of the leg. While passing through the thigh, the great saphenous vein turns anteriorly while merging with several superficial veins. It then travels superficially over the medial region of the thigh, remaining parallel to the medial edge of the sartorius muscle. In the femoral triangle, the long saphenous vein forms an arch as it penetrates into the depth of the thigh. It ultimately passes through the saphenous opening (fossa ovalis) of the fascia lata (cribriform fascia)

FIG. 2.43: Branches of the femoral artery

Labels (left side): Deep circumflex iliac artery; Superficial circumflex iliac artery; Lateral femoral circumflex artery; Deep artery of the thigh; Perforating branches; Superior lateral genicular artery; Patellar anastamoses; Inferior lateral genicular artery; Anterior tibial artery

Labels (right side): External iliac artery; Inferior epigastric artery; Superficial epigastric artery; Superficial external pudendal artery; Deep external pudendal artery; Obturator artery; Femoral artery; Medial circumflex femoral artery; Muscular branches; Adductor hiatus; Descending genicular artery; Superior medial genicular artery; Inferior medial genicular artery

FIG. 2.44: The great saphenous vein and its tributaries

Labels (left side): Inguinal ligament; Superficial circumflex iliac vein; Saphenous opening (fossa ovalis); Small saphenous vein; Dorsal metatarsal veins; Dorsal digital veins

Labels (right side): Superficial epigastric vein; Femoral vein; Superficial external pudendal vein; Accessory saphenous vein; Great saphenous vein; Great saphenous vein; Dorsal venous arch; Dorsal digital vein of medial side of great toe

and opens onto the anterior surface of the femoral vein 4 cm below the inguinal ligament. It enters the femoral vein at this junction, which then passes through the femoral canal.

Tributaries

The great saphenous vein receives numerous tributaries from the front and back of the leg, and from the front of the thigh. These include the following:

❑ *Veins accompanying the branches of femoral artery*: Just before it pierces the cribriform fascia, it receives the superficial epigastric, superficial circumflex iliac and external pudendal veins. These veins accompany the corresponding arteries.
❑ *Anterior cutaneous vein of the thigh*: It also receives the anterior cutaneous vein of the thigh, which drains the lower part of the front of the thigh.
❑ *Anterior vein of the leg*: Just below the knee it receives the anterior vein of the leg, and the posterior arch vein.
❑ *Medial marginal vein of the foot*: Over the dorsum of the foot the great saphenous vein receives the medial marginal vein of the foot.
❑ *Perforating veins*: The great saphenous vein is connected to the deep veins of the leg and thigh through a number of perforating veins. The valves of the perforating veins should prevent backflow of blood from the deep to the superficial system.

Nerve Supply of The Lower Limbs

Innervation of the lower limb is from the lumbosacral plexus. This is formed from the ventral rami of the spinal nerves, T12 to S4. The sciatic nerve is the nerve supplying posterior aspect of the thigh and is derived from L4 to S3 and contains fibres from anterior and posterior aspects of the lumbosacral plexus. Sciatic nerve gives rise to the tibial nerve and the common peroneal nerve. The tibial nerve is derived from the anterior division of L4–S3. It passes through the popliteal fossa and provides branches to the posterior aspect of the calf and the knee joint.

The femoral nerve arises from the dorsal division of the ventral rami of L2–L4. It passes beneath the inguinal ligament to enter the thigh where it divides into an anterior and posterior division. It provides innervation to the quadriceps and sartorius as well as gives rise to the anterior cutaneous branches.

Perineum

The perineum comprises of a urogenital triangle anteriorly and an anal triangle posteriorly **(Fig. 2.45)**. Anatomical borders of the perineum are as follows:
❑ *Anteriorly*: Pubic symphysis
❑ *Posteriorly*: The tip of the coccyx
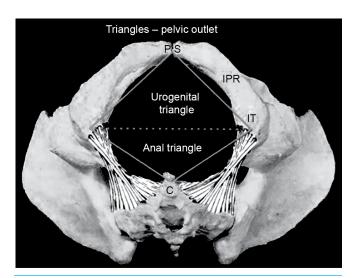
FIG. 2.45: Triangles of the pelvic outlet
Abbreviations: PS, pubic symphysis; IPR, ischiopubic rami; IT, ischial tuberosity

❑ *Laterally*: Inferior pubic rami, inferior ischial rami, and the sacrotuberous ligament
❑ *Roof:* The pelvic floor
❑ *Base:* Skin and fascia.

Perineum, which is the inferior part of the pelvic outlet, can be divided into two triangles by drawing an imaginary line between the two ischial tuberosities. The urogenital triangle, with the pubic symphysis being its apex lies anteriorly. On the other hand, the anal triangle with its apex at the coccyx lies posteriorly.

Anatomy of the Urogenital Region

The urogenital triangle lies superficial to the anterior pelvic diaphragm. It is bordered by a line joining the ischial tuberosities and the ischiopubic rami. The urogenital triangle is associated with the structures of the urogenital system—the external genitalia and the urethra. Unlike the anal triangle, the urogenital triangle has an additional layer of strong deep fascia, the perineal membrane. The perineal membrane is a tough fascial sheet, which attaches to the sides of the urogenital triangle. It is penetrated by the urethra and the external genitalia. The perineal membrane provides attachment for the muscles of the superficial external genitalia and helps in supporting the pelvic viscera. It has pouches on its superior and inferior surfaces, i.e. the superficial and deep perineal pouches respectively **(Fig. 2.46)**.

❑ *Superficial perineal pouch*: This is a potential space between the perineal membrane superiorly, and the perineal fascia inferiorly. It exists superficial to the perineal membrane. It contains the erectile tissues that form the penis and clitoris, and three muscles—the ischiocavernosus, bulbospongiosus (pierced by the vulva and Bartholin's glands in the females and surrounding the corpus spongiosum in males) and superficial transverse perineal muscles. The greater
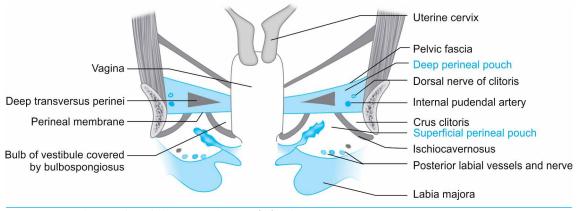
FIG. 2.46: Coronal section through the urogenital region of a female

vestibular glands (Bartholin's glands) are also located in the superficial perineal pouch.

❑ *Deep perineal pouch:* This is a potential space between the pelvic floor superiorly, and the perineal membrane inferiorly. In other words, deep perineal pouch is the space between the perineal membrane and the levator ani muscle. In females, it contains part of the urethra, the external urethral sphincter, the deep transverse perineal muscles and the areolar tissue. In males, it also contains the bulbourethral glands.

Urogenital Diaphragm

Urogenital diaphragm, also known as the triangular ligament can be described as the muscular sheath, which separates deep perineal pouch from the upper pelvis. Some people doubt the existence of the urogenital diaphragm. The muscles of the urogenital diaphragm include the following:

❑ Superficial transversus perinei
❑ Bulbospongiosus muscle
❑ Ischiocavernosus muscle
❑ Sphincter urethrae.

For detailed description of the muscles of the pelvic floor, pelvic diaphragm and the perineal body, kindly refer to Chapter 14.

Anal Triangle

The anal triangle (the posterior perineum) lies between the ischial tuberosities and the coccyx and comprises of the following:

❑ Anus and its sphincters (both internal and external anal sphincters)
❑ Levator ani and the median raphe
❑ Ischiorectal fossae.

Anatomy of the Foetus

Obstetrically, the head of foetus is the most important part, since an essential feature of labour is an adaptation between the foetal head and the maternal bony pelvis. Only a comparatively small part of the head of the foetus at term is represented by the face; the rest is composed of the firm skull, which is made up of two frontal, two parietal and two temporal bones, along with the upper portion of the occipital bone and the wings of the sphenoid. The bones are not united rigidly but are separated by membranous spaces, the sutures.

The foetal skull has five main sutures **(Fig. 2.47)**, which are as follows:

1. *Sagittal or longitudinal suture*: This suture lies longitudinally across the vault of the skull in midline between the anterior fontanelle and the posterior fontanelle. It lies between the two parietal bones.
2. *Coronal suture*: These sutures are present between the parietal and frontal bones, and extend transversely on either side from the anterior fontanelle.
3. *Lambdoid suture*: This suture separates the occipital bone from the two parietal bones and extends transversely both on the right and left side from the posterior fontanelle.
4. *Frontal/metopic suture*: This suture is present between the two halves of the frontal bone in the skull of infants and children and usually disappears by the age of 6 years.
5. *Squamosal suture*: Anteriorly, it begins backwards from the pterion. It then extends between the temporal squama and the lower border of the parietal bone. Posteriorly, it is continuous with the nearly horizontal parietomastoid suture.
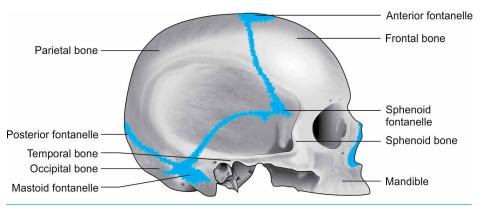
FIG. 2.47: Important sutures and bones of foetal skull

Where several sutures meet, an irregular space is formed, which is enclosed by a membrane and is designated as a fontanelle. There are six fontanelles at the edges of the parietal bones:

❑ One anterior or greater fontanelle
❑ Two anterolateral (sphenoidal) fontanelles
❑ Two posterolateral (mastoidal) fontanelles
❑ One posterior or lesser fontanelle.

The greater or anterior fontanelle is a lozenge-shaped space situated at the junction of the sagittal and coronal sutures **(Fig. 2.48)**. The lesser, or posterior fontanelle is represented by a small triangular area at the intersection of the sagittal and lambdoid sutures. Both may be felt readily during labour, and their recognition gives important information concerning the presentation and position of the foetus. The two main fontanelles having obstetric significance in the foetal head are anterior fontanelle (bregma) and posterior fontanelle (lambda).

Anterior fontanelle is formed by joining of four sutures: frontal suture (anteriorly); sagittal suture (posteriorly) and coronal sutures on the two sides (laterally). The palpation of anterior fontanelle on vaginal examination is of great obstetric significance **(Table 2.9)**. The anterior fontanelle closes by approximately 2 years of age.

On the other hand, the joining of three sutures forms posterior fontanelle: sagittal suture (anteriorly) and lambdoid sutures on the two sides.

Presenting Parts of Foetal Skull (Fig. 2.49)

These include the following:

Vertex: This is a quadrangular area bounded anteriorly by bregma (anterior fontanelle) and coronal sutures; posteriorly by lambda (posterior fontanelle) and lambdoid sutures; and laterally by arbitrary lines passing through the parietal eminences. When vertex is the presenting part, foetal head lies in flexion.

Face: This is an area bounded by the root of the nose along with the supraorbital ridges and the junction of the chin or floor of mouth with the neck. Foetal head is fully extended during this presentation.
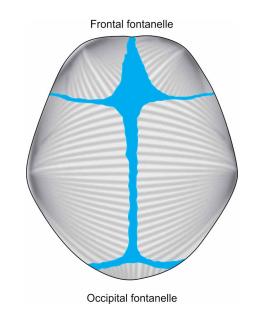
FIG. 2.48: Anterior and posterior fontanelles in foetal head

TABLE 2.9	Obstetric significance of the anterior fontanelle

• Palpation of anterior fontanelle indicates degree of flexion of foetal head
• It facilitates moulding of foetal head
• The membranous nature of anterior fontanelle helps in accommodating the rapid growth of brain during neonatal period
• Floor of the anterior fontanelle reflects the intracranial status. The floor may be depressed in case of dehydration and elevated in case of hydrocephalus or other conditions with raised intracranial tension

Brow: This is an area of forehead extending from the root of nose and supraorbital ridges to the bregma and coronal sutures. The foetal head lies midway between full flexion and full extension in this presentation.

Some other parts of foetal skull, which are of significance, include the following:

Sinciput: Area in front of the anterior fontanelle corresponding to the forehead.

Occiput: Area limited to occipital bone.

Mentum: Chin of the foetus.

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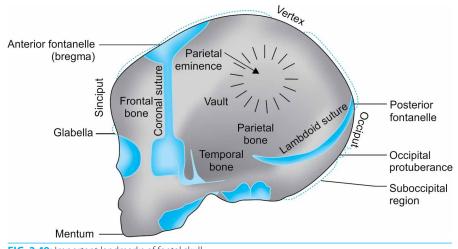
FIG. 2.49: Important landmarks of foetal skull

Parietal eminences: Prominent eminences on each of the parietal bones.

Subocciput: This is the junction of foetal neck and occiput, sometimes also known as the nape of the neck.

Submentum: This is the junction between the neck and chin.
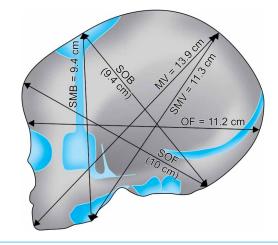
Important Diameters of Foetal Skull

Anterior-Posterior Diameters

The important AP diameters of the foetal skull are sub-occipitobregmatic (9.4 cm); suboccipitofrontal (10 cm); occipitofrontal (11.2 cm); mentovertical (13.9 cm); submentovertical (11.3 cm) and submentobregmatic (9.4 cm). These diameters are described in **Figure 2.50** and **Table 2.10**.

Transverse Diameters
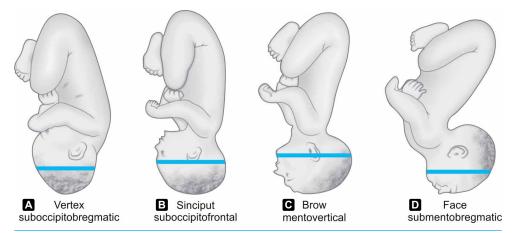
❑ *Biparietal diameter (9.5 cm)*: It extends between the two parietal eminences. This diameter nearly always engages.
❑ *Supersubparietal diameter (8.5 cm)*: It extends from a point placed below one parietal eminence to a point placed above the other parietal eminence of the opposite side.

FIG. 2.50: Diameters of foetal skull
Abbreviations: SMB, submentobregmatic; SOB, suboccipitobregmatic; OF, occipitofrontal; MV, mentovertical; SMV, submentovertical; SOF, suboccipitofrontal

❑ *Bitemporal diameter (8 cm)*: Distance between the anteroinferior ends of the coronal sutures.
❑ *Bimastoid diameter (7.5 cm)*: Distance between the tips of the mastoid process. This diameter is nearly incompressible.

TABLE 2.10	Anterior-posterior diameters of the foetal head which may engage			
Diameter	**Extent**	**Length**	**Attitude of head**	**Presentation**
Suboccipitobregmatic	Extends from the nape of the neck to the centre of bregma	9.4 cm	Complete flexion	Vertex
Suboccipitofrontal	Extends from the nape of the neck to the anterior end of anterior fontanelle or centre of sinciput	10 cm	Incomplete flexion	Vertex
Occipitofrontal	Extends from the occipital eminence to the root of nose (glabella)	11.2 cm	Marked deflexion	Vertex
Mentovertical	Extends from midpoint of the chin to the highest point on sagittal suture	13.9 cm	Partial extension	Brow
Submentovertical	Extends from the junction of the floor of the mouth and neck to the highest point on sagittal suture	11.3 cm	Incomplete extension	Face
Submentobregmatic	Extends from the junction of the floor of the mouth and neck to the centre of bregma	9.4 cm	Complete extension	Face

| A | Vertex suboccipitobregmatic | B | Sinciput suboccipitofrontal | C | Brow mentovertical | D | Face submentobregmatic |

FIGS 2.51A TO D: Engaging diameters of foetal head depending on the position of presenting part

TABLE 2.11	Plane of engagement of foetal head depending upon its attitude	
Attitude of head	**Plane of shape**	**Engagement**
Complete flexion	Biparietal-suboccipitobregmatic	Almost round
Deflexion	Biparietal-occipitofrontal	Oval
Incomplete extension	Biparietal-mentovertical	Bigger oval
Complete extension	Biparietal-submentobregmatic	Almost round

TABLE 2.12	Foetal denominators in relation to foetal presenting parts
Foetal presenting part	**Denominator**
Vertex	Occiput
Face	Mentum
Brow	Frontal eminence
Breech	Sacrum
Shoulder	Acromion

The foetal head is said to be engaged when maximum transverse diameter of foetal head can pass through the pelvic brim. The shape and the diameter of the circumference of the foetal skull varies with the degree of flexion and hence the presentation. A normal pelvis would be easily able to permit the engagement of the foetal skull in vertex and face presentations. This is so as in case of vertex and face presentations, the engaging AP diameters of foetal skull are respectively suboccipitobregmatic (9.4 cm) and submentobregmatic (9.4 cm) **(Table 2.11)**. However, the passage of the foetal head in brow presentation would not be able to take place in a normal pelvis as the engaging AP diameter of foetal skull is mentovertical (13.9 cm) in this case. Therefore, arrest of labour occurs when the foetal head is in brow presentation **(Figs 2.51A to D)**.

Obstetrically, the head of foetus is the most important part, since an essential feature of labour is an adaptation between the foetal head and the maternal bony pelvis. The foetal skull has three parts, the face, the base and the vault, of which only the cranial vault molds at the time of labour. The cranial vault is composed of the following bones: parietal, frontal, squamous temporal and the squamous part of the occipital bone. Bone formation in this region occurs primarily by way of intramembranous ossification. The cranial base consists of the following bones: basilar and lateral portions of the occipital bone, sphenoid, ethmoid and the petrous and mastoid part of the temporal bone. The bones that form the base of the skull are formed by endochondral ossification.

Denominator

Denominator can be described as an arbitrary fixed bony point on the foetal presenting part **(Table 2.12)**.

Choose the Single Best Answer (SBA)

Q 1. **A 26-year-old female presents with right iliac fossa pain and is taken to theatre for an appendectomy. An incision is made through the skin and onto muscle with fibres passing inferiorly in an oblique direction. Which muscle is cut in this scenario?**
A. Internal oblique
B. External oblique
C. Rectus abdominis
D. Transversus abdominis
E. Pyramidalis

Q 2. **Whilst examining the abdomen of a 21-year-old female with abdominal pain you notice a well-defined "six-pack". Which muscle is this?**
A. Rectus abdominis
B. Transversus abdominis
C. Cremaster
D. External oblique
E. Internal oblique

Q 3. **A 26-year-old female presents with right iliac fossa pain and is taken to theatre for an appendectomy. An incision is made through the skin and onto muscle with fibres passing superiorly in an oblique direction. Into which muscle has the incision been made?**
A. Transversus abdominis
B. Cremaster
C. External oblique
D. Internal oblique
E. Rectus abdominis

Q 4. **Regarding the blood supply of anterior abdominal wall, which is not true?**
A. Cutaneous branches from superior and inferior epigastric arteries supply the flanks.
B. Cutaneous branches from the superior and inferior epigastric arteries supply the area near the midline.
C. Branches from the intercostal and lumbar arteries supply the flanks.
D. All the above.
E. None of the above

Q 5. **Regarding the lymphatic drainage of the anterior abdominal wall, which of the following is true?**
A. The cutaneous lymph vessels above the umbilicus drain into the anterior axillary lymph nodes, while the vessels below this level drain into the superficial inguinal lymph nodes.
B. All lymphatic drainage occurs into the inguinal nodes.
C. The cutaneous lymph vessels above the umbilicus drain into the supraclavicular lymph nodes, while the vessels below this level drain into the deep inguinal lymph nodes.

D. All the above
E. None of the above

Q 6. **Which of the following characteristics about rectus sheath is correct?**
A. Below the arcuate line, the posterior layer of the rectus sheath is formed by the transversalis fascia.
B. Each rectus abdominis muscle is attached by a single tendon to the pubic bone.
C. Below the arcuate line, the posterior wall of the sheath is formed by internal oblique.
D. All the above
E. None of the above

Q 7. **Which of the following is not true regarding the branches of the aorta?**
A. The superior mesenteric artery supplies the gastrointestinal tract from the middle of the second part of the duodenum as far as the distal one-third of the transverse colon.
B. The coeliac trunk is surrounded by the coeliac plexus of nerves.
C. The superior mesenteric artery arises from the aorta at the level of the first lumbar vertebra.
D. The left colic artery is the larger terminal branch of the superior mesenteric artery.
E. The inferior mesenteric artery arises from the aorta at the inferior border of the third part of the duodenum at the level of the third lumbar vertebra.

Q 8. **Which of the following is not true regarding the abdominal aorta?**
A. Enters the abdomen at the level of the twelfth thoracic vertebra.
B. It divides into the two common iliac arteries in front of the fifth lumbar vertebra.
C. The common iliac arteries divide into external and internal iliac arteries in front of the sacroiliac joint.
D. The external iliac artery runs along the medial border of the psoas major muscle.
E. The superior mesenteric artery is a posterior visceral branch of the aorta.

Q 9. **Which of the following is true regarding the human rectum?**
A. Drains lymph to the pre-aortic nodes.
B. Has mesentery in the posterior third.
C. Is covered anteriorly by peritoneum along its whole length.
D. Has a blood supply from the terminal branches of the superior mesenteric artery.
E. Has appendices epiploicae.

---

1. B   2. A   3. D   4. A   5. A   6. A   7. D   8. E   9. A

Q 10. **Which of the following is not true regarding the boundaries of epiploic foramen?**

A. Anteriorly it is formed by the free border of the lesser omentum.
B. Inferiorly by the peritoneum covering the duodenum.
C. Posteriorly by inferior vena cava.
D. Posteriorly by superior vena cava.
E. Superiorly by the caudate lobe of liver.

Q 11. **Which of the following vessel is not a branch of the internal iliac artery?**

A. Iliolumbar artery
B. Lateral sacral artery
C. Middle rectal artery
D. Obturator artery
E. Ovarian artery

Q 12. **Which of the following is not true regarding the appendix?**

A. It is located in the retrocaecal recess.
B. The longitudinal coat of the appendix is derived from the three bands of taeniae coli.
C. Is supplied by branches of the superior mesenteric artery.
D. It is typically less than 10 cm in length in the adult.
E. McBurney's point lies two-thirds laterally from a line from umbilicus to the anterior superior iliac spine.

Q 13. **Which of the following is not true regarding the pancreas?**

A. Derives part of its blood supply from the splenic artery
B. Has parts in both the supracolic and infracolic compartments
C. Is pierced by the middle colic artery
D. Lies anterior to the left kidney
E. The junction of the splenic and superior mesenteric veins lies behind the gland.

Q 14. **Which of the following is not correct regarding the common bile duct?**

A. Lies anterior to the first part of the duodenum
B. Lies posterior to the portal vein
C. Lies in the free edge of the lesser omentum
D. Lies to the right of the hepatic artery
E. May open into the duodenum independent of the pancreatic duct

Q 15. **Which of the following structures does not pass through the female inguinal canal?**

A. Iliohypogastric nerve
B. Ilioinguinal nerve
C. Interior epigastric artery
D. Round ligament
E. Spermatic cord

Q 16. **Which of the following structures pass under the inguinal ligament?**

A. The long saphenous vein
B. The superficial femoral artery
C. The superficial epigastric vein
D. The genital branch of the genitofemoral nerve
E. The femoral branch of the genitofemoral nerve

Q 17. **Which of the following is true concerning the femoral ring?**

A. It is bounded laterally by the femoral artery.
B. It is bounded medially by the lacunar ligament.
C. It is lined by peritoneum.
D. It is not traversed by lymph vessels.
E. Passes deep to the inguinal ligament.

Q 18. **In the femoral triangle, which of the following is/are true of the femoral artery?**

A. Crossed by the superficial circumflex iliac vein
B. Lateral to the femoral nerve
C. Medial to the long saphenous vein
D. Posterior to the femoral branch of the genitofemoral nerve
E. Posterior to the femoral vein at the apex of the triangle

Q 19. **Which of the following is true regarding the femoral nerve?**

A. Has a branch which supplies the skin of the scrotum
B. Lies within the femoral sheath
C. Lies lateral to the femoral vein
D. May supply part of the foot
E. Does not have the same origin as the obturator nerve

Q 20. **Which of the following is a branch of the femoral artery?**

A. Peroneal artery
B. The ascending genicular artery
C. The deep epigastric artery
D. Medial circumflex iliac artery
E. Superficial circumflex iliac artery

Q 21. **Which of the following is not true regarding the femoral canal?**

A. It contains a superficial inguinal lymph node.
B. It contains the lymph node of Cloquet.
C. Has the inguinal ligament as its anterior border.
D. Has the lacunar ligament as its medial border.
E. Has the pectineal ligament as its posterior border.

Q 22. **Which of the following is true regarding the femoral nerve?**

A. It arises from the same nerve roots as the obturator nerve.
B. It supplies lateral side of the dorsum of foot.
C. It gives a branch to the scrotum.
D. It enters the femoral sheath.
E. Saphenous nerve arises from the anterior division of femoral nerve.

10. D   **11. E**   **12. E**   **13. C**   **14. B**   **15. E**   **16. E**   **17. B**   **18. D**   **19. C**   **20. E**   **21. A**   **22. A**

Q 23. Which of the following is true concerning the great (long) saphenous vein?
A. Ascends posterior to the medial malleolus
B. Passes through the femoral canal
C. Receives blood from the posterior tibial veins
D. Passes through the cribriform fascia
E. Receives the blood from deep external pudendal veins

Q 24. A 63-year-old man in the surgical ward is complaining of numbness over the anterior thigh and medial aspect of his right leg. He is unable to extend his right knee and the knee jerk is reduced. He had undergone a femoral aneurysm repair 3 days ago. Which is the most probable nerve damaged in this case?
A. Femoral nerve
B. Saphenous nerve
C. Femoral branch of genitofemoral nerve
D. Genital branch of genitofemoral nerve
E. Sciatic nerve

Q 25. A 33-year-old lady, who is 38 weeks pregnant, presents to her general practitioner with a 4-week history of pain and paraesthesia over the upper outer aspect of her right thigh. There is no restriction of movements in her hips or knees and her gait is normal. Which is the most likely nerve, which is affected in this case?
A. Lateral cutaneous nerve of thigh
B. Femoral nerve
C. Common peroneal nerve
D. Sciatic nerve
E. Tibial nerve

Q 26. Please choose the most appropriate answer regarding the innervations of various muscles from the given list.
A. Triceps is supplied by radial nerve.
B. Gastrocnemius is supplied by tibial nerve.
C. Opponens pollicis is supplied by the median nerve.
D. Deltoid muscle is supplied by axillary nerve.
E. All are correct.

Q 27. Which of the following regarding the vagina is correct?
A. The paramesonephric duct gives rise to both the uterus and the entire vagina.
B. Is 7–10 cm long.
C. The urogenital sinus gives rise to part of the vagina and the proximal part of the urethra.
D. Becomes canalised at 22 weeks of gestational age.
E. Vaginal fluid has a lower potassium concentration than plasma.

Q 28. Presence of vaginal septa is not associated with which of the following?
A. Dysmenorrhoea
B. Dyspareunia
C. Obstructed labour
D. Uterine abnormalities
E. Easy removal in most of the cases

Q 29. Which of the following statement is true regarding the vagina?
A. Contains mucus-secreting glands in its epithelium.
B. During reproductive life, has an acid pH.
C. Has an anterior wall longer than the posterior wall.
D. Is derived from the mesonephric duct.
E. Is related in its lower third to the bladder base.

Q 30. Which of the following is not true regarding the vulva?
A. The urethral fold fuses, as in the male, to develop into labia minora.
B. The nerve endings of the vestibule are mostly free, with few or no corpuscles.
C. The blood supply of the labia majora is mainly derived from the external and internal pudendal arteries.
D. The labia majora contain Ruffini, Meissner, Merkel and Pacini corpuscles.
E. The Bartholin's gland is normally not palpable.

Q 31. From which of the following is the nerve supply to the vulva derived?
A. Genitofemoral nerve
B. Ilioinguinal nerve
C. Pudendal nerve
D. All the above
E. None of the above

Q 32. Blood supply of the vulva is derived from which of the following?
A. Deep external pudendal artery
B. Internal pudendal artery
C. Superficial external pudendal artery
D. All the above
E. None of the above

Q 33. Which of the following is not true regarding the vagina?
A. Is covered anteriorly by peritoneum.
B. Is related anteriorly and posteriorly to the Wolffian (Gartner's) ducts.
C. It is made up of keratinized squamous epithelium.
D. The lateral vaginal walls are in contact with each other.
E. All the above.

23. D   24. A   25. A   26. E   27. B   28. A   29. B   30. A   31. D   32. D   33. E

Q 34. **The change, which occurs in the vagina at puberty, includes which of the following?**

A. An increase in the pH
B. Reduction in the number of Döderlein's bacilli
C. Exfoliation of superficial cells with pyknotic nuclei
D. Reduction in the glycogen content of the epithelium
E. The appearance of glands in the epithelium

Q 35. **Which of the following is true regarding the vagina?**

A. Has venous drainage to the external iliac vein
B. Is covered with peritoneum in its upper anterior aspect
C. Is lined by squamous epithelium
D. Is supplied in part by the pudendal nerve
E. Has an anterior wall that is longer than the posterior wall

Q 36. **The lymphatic drainage of the cervix does not go to which of the following?**

A. Directly to the para-aortic nodes
B. To the internal iliac nodes
C. To the obturator node
D. To the superficial inguinal nodes
E. None of the above

Q 37. **Which of the following statements about the cervix is correct?**

A. Nulliparous cervix is slit-shaped.
B. Peritoneum covers the upper part of the vagina posteriorly.
C. The isthmus is part of the cervix.
D. Cervix is lined by stratified squamous epithelium.
E. The squamocolumnar junction is found at the internal os.

Q 38. **Which of the following statement concerning the uterus is correct?**

A. It is formed from the mesonephric ducts.
B. Pain from the body of the uterus is carried by the pelvic splanchnic nerves.
C. The uterine artery is a branch of the internal iliac artery.
D. The uterine artery passes below the ureter.
E. The uterus is supported only by the levator ani muscles.

Q 39. **Which of the following is not true regarding the uterus and the cervix?**

A. The body of the uterus and cervix are relatively insensitive to cutting and burning.
B. The cavity of the uterus, as in the cervix is fusiform in shape.
C. The endometrial lining and not the cervical lining is shed at the time of menstruation.
D. The lymphatics of the cervix drain to the external and internal iliac nodes.
E. In pregnancy the internal os is located in the cervix rather than the isthmus.

Q 40. **Which of the following statement is true concerning retroversion of the uterus?**

A. It is a common cause of subfertility.
B. May be corrected by a Fothergill operation.
C. Should be corrected with a Hodge pessary in early pregnancy.
D. It is caused by heavyweight lifting.
E. It may occur in 20% of normal women.

Q 41. **Which of the following is true regarding the innervation of the uterus and birth canal?**

A. A paracervical nerve block may be used for a forceps delivery.
B. The pudendal nerve block provides good pain relief for the second stage of labour.
C. The parasympathetic nervous system causes contraction of the pregnant uterus.
D. The uterus contains alpha and beta receptors.
E. Pudendal nerve arises from S2 to S3 nerve roots.

Q 42. **Which of the following is not true regarding the ovary?**

A. Has lymphatic drainage to the para-aortic nodes
B. It is not covered by peritoneum
C. Is sensitive to pressure
D. Lies posterior to the broad ligament
E. Receives its blood supply from a branch of the internal iliac artery.

Q 43. **Which of the following regarding the ovary is correct?**

A. The ovaries do not descend into the pelvic cavity until childhood.
B. The ovarian artery arises from the aorta at the level of the third lumbar vertebra.
C. The ureter passes in front of the ovary at the level of the bifurcation of the iliac arteries.
D. The nerve supply to the ovary is derived exclusively from parasympathetic fibres.
E. During pregnancy the enlarged uterus pushes the ovaries down into the pelvis.

Q 44. **Which of the following does not contribute to the boundaries of the ovarian fossa?**

A. External iliac vein
B. Internal iliac artery
C. Internal pudendal artery
D. Obliterated umbilical artery
E. Ureter

Q 45. **The outermost portion of the ovary is covered by which of the following?**

A. Adipocytes
B. Cuboidal cells
C. Intercalary cells
D. Mesothelial cells
E. Smooth muscle cells

34. C   35. D   36. D   37. B   38. C   39. B   40. E   41. D   42. E   43. A   44. C   45. B

Q 46. Which of the following is not true concerning ovarian function?

A. Circulating inhibin concentrations are a marker of granulosa cell function.
B. Granulosa cells secrete oestradiol.
C. Insulin-like growth factor (somatomedin C) is not secreted by the ovary.
D. Oestradiol is derived from androgen precursors.
E. Progesterone is the major steroid of the developing follicle.

Q 47. Which of the following organs does not respond to oestrogens?

A. Breasts
B. Cervical glands
C. Fallopian tube
D. Vaginal epithelium
E. None of the above

Q 48. Which of the following is not true regarding the fallopian tube?

A. Has a thick muscle layer in the isthmus
B. Is actively motile
C. Is covered by peritoneum
D. Lies anterior to the round ligament
E. Possesses a cilial lining

Q 49. Which of the following is true regarding the right ovary?

A. Is covered by peritoneum in the adult
B. Receives its blood supply from the internal iliac artery
C. Has the ovarian ligament attached to its medial pole
D. Its venous drainage is to the right renal vein
E. Has lymphatic drainage to the internal iliac nodes

Q 50. Which of the following is true regarding the anal sphincter?

A. The internal anal sphincter is attached to the outer longitudinal fibres of the rectum.
B. Deep external anal sphincter merges with puborectalis.
C. The internal anal sphincter is supplied by inferior rectal nerve.
D. The external anal sphincter is supplied by the inferior hypogastric plexus.
E. The superficial external anal sphincter is not attached to the coccyx.

Q 51. Which of the following is not true regarding the ischio-rectal fossa?

A. The two fossae communicate with each other.
B. Contains the perineal nerve in its posterior part.
C. Lies inferior to the levator ani.
D. The anal canal and the sloping levator ani muscles form the medial wall of each fossa.
E. Contains the middle rectal artery.

Q 52. Which of the following is not true regarding the pudendal canal?

A. Contains the pudendal artery and vein
B. Does not contain the pudendal nerve
C. Runs superior to the sacrotuberous ligament
D. Runs medial to the obturator internus
E. Passes medial to the ischial spines

Q 53. Which of the following is not a branch of the pudendal nerve?

A. Dorsal nerve of the penis
B. Genitofemoral nerve
C. Inferior rectal nerve
D. Perineal nerve
E. None of the above

Q 54. Which of the following is true concerning the pudendal nerve?

A. Arises from the anterior rami of S2–S4
B. Supplies the levator ani muscle
C. Supplies the clitoris
D. Leaves the pelvis through the greater sciatic foramen
E. All the above

Q 55. Which of the following nerve does the pudendal nerve not carry?

A. Motor fibres to the external anal sphincter
B. Motor fibres to the internal anal sphincter
C. Parasympathetic fibres to the anal canal
D. Sensory fibres from the perineum
E. Sensory fibres to the clitoris

Q 56. Which of the following is not true regarding the uterine vessels?

A. The uterine artery arises from the anterior division of internal iliac artery.
B. The uterine artery crosses above the ureter and reaches the cervix at the level of the internal os.
C. The uterine artery, along with the vaginal and ovarian arteries, enlarges during pregnancy.
D. The uterine veins do not have surrounding supporting sheaths.
E. The uterine vein drains into the internal iliac vein.

Q 57. Which of the following is not true regarding the external iliac artery?

A. At its origin is crossed by the ovarian vessels
B. At its origin is crossed by the ureter
C. Enters the thigh posterior to the inguinal ligament
D. Gives rise to the deep external pudendal artery
E. Lies medial to the external iliac vein at its proximal end.

Q 58. Which of the following is not true regarding the inferior hypogastric (pelvic) plexus?

A. Contains parasympathetic fibres
B. Extends into the base of the broad ligament of the uterus
C. Gives rise to the vesical plexus
D. It lies between the two iliac vessels
E. Is situated on the side of the anal canal

46. E   47. E   48. D   49. C   50. B   51. E   52. B   53. B   54. E   55. B   56. C   57. D   58. E

Q 59. Which of the following is true regarding the nerves supplying the pelvic structures?

A. The phrenic nerve arises from the anterior rami of C3–C5.
B. The femoral nerve supplies psoas major and iliacus.
C. The phrenic nerve has a contribution to the suprarenal glands via the coeliac plexus.
D. The levator ani muscle is innervated by S2–S4.
E. All the above.

Q 60. Which of the following statements about the lymphatic drainage of the genital tract is true?

A. Drainage from the corpus uteri goes partly to the superficial inguinal nodes.
B. Drainage from the oviducts is mainly via the para-aortic nodes.
C. Ovarian drainage is directly to the para-aortic nodes.
D. The lower-third of the vagina drains to the superficial inguinal nodes.
E. All the above.

Q 61. Within lymph nodes, which of the following is not true?

A. B lymphocytes predominate in the follicles of the cortex.
B. Lymph flows from the hilum of the node outwards to the marginal sinus.
C. T lymphocytes predominate in the paracortex.
D. Primary follicles are aggregates of B cells.
E. Secondary follicles develop following antigenic stimulation.

Q 62. Which of the following regarding the female urinary bladder is true?

A. Is connected laterally to the tendinous arch of the pelvic fascia
B. Is not in contact with the supravaginal uterine cervix
C. Is separated from the posterior surface of the pubis by peritoneum
D. Is not joined to the umbilicus by the urachus
E. Receives visceral afferent innervation from the pudendal nerve

Q 63. Which of the following is true regarding the female urinary bladder?

A. The urinary bladder is not connected to the umbilicus
B. Receives visceral afferent innervation from the pudendal nerve
C. Is separated from the posterior surface of the pubis by peritoneum
D. Is connected medially to the tendinous arch of the pelvic fascia
E. Is in contact with the supravaginal uterine cervix.

Q 64. Which of the following statements is correct concerning the ischiorectal fossa?

A. It is separated from the perianal space by perianal fascia
B. The fossa is crossed transversely by the inferior haemorrhoidal veins
C. The levator ani muscle forms the floor of the fossa
D. The obturator internus muscle lies in its medial wall
E. The pudendal nerve lies within the fat of the fossa

Q 65. Which of the following does the right ureter not lie in close relationship with?

A. Bifurcation of the right common iliac artery
B. Inferior mesenteric artery
C. Infundibulopelvic ligament
D. Uterine artery
E. None of the above

Q 66. Which of the following is not true concerning the ureters in the female?

A. Both cross below the uterine arteries near the cervix.
B. Both cross near the bifurcation of the common iliac artery.
C. Both have three sites of anatomic constriction.
D. Both pass anterior to the ovarian vessels.
E. Both run on the anterior surface of the psoas major muscle.

Q 67. Which of the following is true concerning the ureter?

A. Has the genitofemoral nerve lies anterior to it
B. Is seen lying on the tips of the transverse processes of the thoracic vertebrae
C. It is surrounded by Waldeyer's sheath as it passes through the bladder wall
D. Lies anterior to the renal artery at the hilum of the kidney
E. Passes into the pelvis over the bifurcation of the internal iliac artery.

Q 68. Which of the following is true regarding the ureter?

A. The muscular layer of the ureter consists of longitudinal and circular layers throughout its whole length.
B. Has a squamous epithelium.
C. It is 10–15 cm in length.
D. The early splitting of the ureteric bud may result in partial or complete duplication of the ureter.
E. It is of endodermal origin.

Q 69. Which of the following is not true concerning the female urethra?

A. Corresponds developmentally to the membranous urethra in the male
B. Has a muscular layer continuous with that of the bladder
C. Has an external sphincter supplied by the obturator nerve
D. Is lined throughout by transitional epithelium
E. Transverses the perineal membrane.

59. E   60. E   61. B   62. A   63. E   64. B   65. B   66. D   67. C   68. D   69. D

Q 70. Which of the following organs is derived from the ectodermal neural crest cells?

A. Inner medulla of adrenal glands
B. Outer cortex of adrenal glands
C. Liver
D. Spleen
E. Pancreas

Q 71. In the human testis, which of the following is correct?

A. Inhibin is synthesised by the Sertoli cell.
B. Testosterone synthesis is stimulated by follicle stimulating hormone (FSH).
C. The main site of testosterone synthesis is the Sertoli cell.
D. The predominant androgen product is androstenedione.
E. It also secretes prolactin.

Q 72. Spermatozoa acquire the ability to become mobile in which of the following?

A. Seminiferous tubules
B. Epididymis
C. Vas deferens
D. None of the above
E. All the above

Q 73. Which of the following is not a part of axillary group of lymph nodes?

A. Central group
B. Clavicular group
C. Lateral group
D. Pectoral group
E. Subscapular group

Q 74. Which of following concerning the lymphatic drainage of the female breast is not true?

A. Approximately 90% of the lymph passes to the posterior infraclavicular nodes.
B. Axillary lymph nodes receive more than half the lymph from the breast.
C. Lymph from the medial part of breast may drain to the parasternal nodes.
D. There is a subareolar lymph plexus.
E. Female breast extends from 2nd rib to 6th rib.

Q 75. Which of the following is not true regarding the female breast?

A. Contains lactiferous ducts in the neonate
B. Is drained by the internal thoracic vein
C. Lies between the 2nd rib and 7th rib
D. Lies entirely in the superficial fascia
E. It is derived embryologically from endoderm

Q 76. Which of the following is not true regarding the diaphragm?

A. Is partly derived from the pleuroperitoneal membranes
B. Has an origin from the xiphoid
C. The vagal trunks enter the diaphragm through the oesophageal opening

D. Is supplied by the musculophrenic branch of the internal thoracic artery
E. An opening in the central tendon transmits the left phrenic nerve

Q 77. Which of the following is not true concerning the pleura?

A. Extends into the neck above the first rib
B. It does not invaginates between the lobes of the lungs
C. Is in contact with parietal and visceral structures
D. Lies posterior to the upper pole of the right kidney
E. Over the diaphragm is supplied by the phrenic nerve

Q 78. Which of the following is not true concerning the vagus nerve?

A. Carries C1 fibres to the infrahyoid muscles
B. Carries motor fibres to the palate muscles
C. Emerges from the medulla oblongata
D. Gives off the recurrent laryngeal nerve on the right as it passes over the subclavian artery
E. Supplies the cricothyroid muscle

Q 79. Which of the following is not true regarding the left phrenic nerve?

A. Carries sensory afferents from the pleura and the pericardium
B. Is anterior to the left scalenus anterior muscle
C. Is anterior to the termination of the thoracic duct
D. Is posterior to the internal jugular vein
E. Is posterior to the prevertebral fascia

Q 80. In the female pelvis all the following are true except:

A. All diameters are greater in comparison to the male pelvis.
B. The sub-pubic arch is about 80–85 degrees.
C. The sacral promontory is more prominent.
D. The obturator foramen is triangular.
E. The sacrum is shorter and wider in the female pelvis.

Q 81. Which of the following concerning measurement of the pelvic size is not correct?

A. An obstetric conjugate of less than 10 cm indicates contracted pelvis.
B. The obstetric conjugate is usually shorter than the anatomical conjugate.
C. The obstetric conjugate is the shortest anterior posterior distance at the inlet.
D. The pelvis of a Chinese woman is usually smaller than that of a European.
E. The transverse diameter is the largest distance between the lateral walls.

Q 82. Which of the following is true concerning the pelvic surface of the sacrum?

A. Gives origin to the levator ani muscle
B. Gives origin to the piriformis muscle
C. Is broader in the male than in the female
D. Is in contact with the anal canal
E. Transmits the dorsal rami of sacral nerves

70. A   71. A   72. B   73. B   74. A   75. E   76. E   77. B   78. A   79. C   80. C   81. D   82. B

Q 83. What is not true regarding the female pelvis?
  A. The inlet is an oval whose longest diameter lies transversely.
  B. All diameters in the mid-strait are 12 cm.
  C. The transverse diameter at the level of the ischial spines is 10.5 cm.
  D. The true conjugate is the anteroposterior diameter of the brim and measures about 11.5 cm.
  E. The sacrum is flattened, narrow and long in females.

Q 84. Which of the following is not true regarding the foetal skull?
  A. Moulding in labour only affects the vault.
  B. The vertex is delineated by the anterior and posterior fontanelles and the frontal eminences.
  C. The anterior fontanelle is at the junction of the sagittal and coronal sutures.
  D. The anterior fontanelle closes in the first 6 months of infancy.
  E. The smallest anterior-posterior diameter of foetal head is suboccipitobregmatic.

Q 85. Which of the following structures do not take part in the formation of the anterior fontanelle in the foetal skull?
  A. Frontal suture
  B. Glabella
  C. Coronal sutures
  D. Sagittal suture
  E. None of the above

Q 86. Which of the following skull bones is not derived from intramembranous ossification?
  A. Occipital bone
  B. Parietal bone

  C. Temporal bone
  D. Sphenoid bone
  E. All the above

Q 87. Which of the following structures are not attached to the perineal body?
  A. External anal sphincter
  B. External urethral sphincter
  C. Levator ani
  D. Pubovaginalis
  E. Deep transverse perineal muscles

Q 88. Which of the following pierce the urogenital diaphragm?
  A. Obturator nerve
  B. Rectum
  C. Ureters
  D. Urethra
  E. Uterus

Q 89. Which of the following is not true regarding the urogenital diaphragm?
  A. It is formed by the sphincter urethrae and deep transverse perineal muscles, which are enclosed between two fascial layers.
  B. It is situated in the anterior part of the perineum.
  C. Posteriorly the two layers of fascia fuse with the perineal body.
  D. The closed space between the superficial and deep fascia is called the superficial perineal pouch.
  E. The inferior layer of fascia is called the perineal membrane.

83. E   84. D   85. B   86. D   87. B   88. D   89. D

Physiology

Haematology

Blood

Blood is a connective tissue in fluid form. In human bodies, blood makes up about 7% of body weight. Blood contains the blood cells that are called formed elements, and the liquid portion is known as plasma. Three types of cells present in the blood are as follows:
1. Red blood cells or erythrocytes
2. White blood cells or leucocytes
3. Platelets or thrombocytes

Blood volume forms a lower percentage of body weight in fat in comparison to thin people. Blood volume can be calculated by the following formula:

$$BV = \frac{PV}{1 - HC}$$

Where BV = blood volume; PV = plasma volume; HC = haematocrit

Blood volume rises after water is drunk because the water is absorbed into the blood. Blood expresses serum when it clots. Serum is plasma minus its clotting factors.

Red Blood Cells (Erythrocytes)

Red blood cells (RBCs) are the non-nucleated formed elements in the blood. Red blood cells are also known as erythrocytes. It is a biconcave, non-nucleated disc-like structure containing haemoglobin. Central portion of the RBC is thinner and periphery is thicker. The half-life of RBC is 120 days. Red colour of the red blood cell is due to the presence of the pigment, haemoglobin. An adult human on an average has 200 g of haemoglobin in the circulating blood. RBCs play a vital role in transport of respiratory gases. It contains the enzyme carbonic anhydrase, which catalyses the combination of H_2O with CO_2 to produce carbonic acid (H_2CO_3). RBCs are larger in number compared to the other two blood cells, namely white blood cells and platelets. Erythrocytes are responsible for the major part of blood viscosity. The blood viscosity rises exponentially with the haematocrit. Red blood cells in humans are nonnucleated. Due to the absence of nucleus in human RBC, the DNA as well as the organelles such as mitochondria and Golgi apparatus also are absent in the RBC. Due to the absence of mitochondria, the energy in the RBCs is produced from glycolytic process. Red cell does not have insulin receptor and so the glucose uptake by RBCs is not controlled by insulin. During the process of haematopoiesis in the bone marrow, the immature red blood cells may be nucleated. Reticulocytes, the most immature circulating RBCs, may show an intracellular network pattern if appropriately stained with certain dyes. However, nucleated red cells are not normally seen in peripheral blood. While flowing through the vessels, red blood cells form an axial stream away from the vessel wall. They travel at slower velocity in capillaries in comparison to the venules because the capillary bed has a greater total cross-sectional area than the venular bed.

The walls of the erythrocyte deform easily to squeeze through capillaries. Normal red cells are approximately 7 microns in diameter. They become bullet-shaped as they pass through capillaries having a diameter of 5 microns. RBCs are the blood component most frequently used for transfusion. A transfusion of RBCs increases the amount of oxygen that can be carried to the tissues of the body.

RBCs that have been separated from the liquid plasma (packed RBCs) should be administered to patients who have anaemia or who have blood loss because the plasma contains the clotting factors which may not be required in these patients.

Breakdown of Erythrocytes in the Body

The normal erythrocyte lifespan is 16–18 weeks (120 days). Breakdown of erythrocytes takes place in the reticuloendothelial system. Breakdown yields iron, which is retained for further use. At the time of erythrocyte breakdown, haemoglobin is degraded into iron, globin and porphyrin. Erythrocyte breakdown also yields bilirubin that is carried by plasma protein to the liver.

Haematocrit Value

Haematocrit or the packed cell volume (PCV) can be defined as the volume percentage of red blood cells in blood. Haematocrit may be obtained by centrifugation of blood because red cells are heavier than plasma. For this purpose, blood is collected in a haematocrit tube along with a suitable anticoagulant and centrifuged for 30 minutes at a speed of 3,000 revolutions per minute (rpm). During this process, the red blood cells settle down at the bottom, whereas the clear plasma is present on top. Plasma forms 55% and red blood cells form 45% of the total blood. In between the plasma and the red blood cells, there is a thin layer of white buffy coat, which is formed by the aggregation of white blood cells and platelets. The appearance of centrifuged blood could be suggestive of various clinical conditions as shown in **Table 3.1**.

Haematocrit may also be calculated by multiplying the mean cell volume by the red cell count. However, this gives a slightly lower value in comparison to centrifugation because a little amount of plasma is trapped between cells during centrifugation. Haematocrit rises in a patient who sustains widespread burns due to the loss of plasma and interstitial fluid. It falls following injections of aldosterone due to an increase in the extracellular fluid and hence plasma volume. Haematocrit also falls in cases of macrocytic megaloblastic anaemia such as pernicious (B_{12} deficiency) anaemia because though individual RBCs are large, total red cell mass is decreased.

Erythrocyte Sedimentation Rate

Erythrocyte sedimentation rate (ESR**)** can be defined as the rate at which the red blood cells sediment in 1 hour. It is a non-specific measurement of inflammation. It is measured using a Westergren tube and is measured as mm/hour.

TABLE 3.1   **Appearance of centrifuged blood**

Appearance of centrifuged blood	Clinical disorder
Reduced percentage of red blood cells	Anaemia
Cloudy or milky appearance	High plasma lipid levels
Yellow appearance	Jaundice
Red appearance	Haemolysis
Greatly thickened buffy coat	Leukaemia

Increased ESR

Factors increasing ESR are as follows:
- ❑ Large cells
- ❑ Lower amount of cells
- ❑ More proteins
- ❑ Inflammatory disease processes such as:
  - Rheumatoid arthritis
  - Systemic lupus erythematosus
  - Polymyalgia rheumatica
  - Malignancy (myeloma)
- ❑ It increases with age (roughly, normal ESR is half of age) and tends to be higher in females.

Reduced ESR

Factors, which may reduce ESR, include the following:
- ❑ Dysfibrinogenaemia
- ❑ Hypogammaglobulinaemia
- ❑ Low-molecular-weight dextran
- ❑ Polycythaemia vera
- ❑ Secondary conditions, which feature abnormal red blood cells:
  - Sickle cell anaemia
  - Hereditary spherocytosis
  - Acanthocytosis
- ❑ Microcytosis, e.g. haemoglobin disease, iron deficiency, etc. produces an artefactual decrease in ESR
- ❑ Hypofibrinogenaemia:
  - Disseminated intravascular coagulation
  - Massive hepatic necrosis
- ❑ Excessive anticoagulation
- ❑ High white blood cell count
- ❑ Cachexia
- ❑ Heart failure

White Blood Cells

White blood cells (WBCs) or leucocytes are the colourless and nucleated formed elements of blood. WBCs differ from RBCs in many aspects as described in **Table 3.2**.

TABLE 3.2   **Difference between red blood cells and white blood cells**

Characteristic	Red blood cells	White blood cells
Colour	Red	Colourless
Shape	Disc-shaped and biconcave	Irregular
Nucleus	Absent	Present
Granules	Absent	Present in some types
Types	Only one type	Many types (e.g. neutrophils, baso-phils, eosinophils, monocytes, lymphocytes, etc.)
Lifespan	Longer (120 days)	Shorter (12 hours to 15 days)

Classification of White Blood Cells

Some of the WBCs have granules in the cytoplasm. Based on the presence or absence of granules in the cytoplasm, the leucocytes are classified into two groups **(Fig. 3.1)**:

❑ Granulocytes (which have granules): These can be further classified into three types:
  • *Neutrophils*: These WBCs have granules that take up both acidic and basic stains.
  • *Eosinophils*: These WBCs have granules that take up acidic stain.
  • *Basophils*: These WBCs have granules that take up basic stain.
❑ Agranulocytes (which do not have granules): These can be further classified into two types:
  • Monocytes
  • Lymphocytes

Lifespan of different types of WBCs is summarised in **Table 3.3**.

Neutrophils

Neutrophil granulocytes are the most common leucocytes in normal blood. Also known as polymorphs, these white blood cells have fine or small granules in the cytoplasm. These cells comprise 60–70% of circulating leucocytes. They contain proteolytic enzymes. Their granules contain enzymes, which along with toxic oxygen metabolites can kill and digest the bacteria they engulf. Nucleus of these cells is

TABLE 3.3	Lifespan of different types of white blood cells
Type of white blood cell	**Lifespan (days)**
Neutrophils	2 to 5
Eosinophils	7 to 12
Basophils	12 to 15
Monocytes	2 to 5
Lymphocytes	½ to 1

multi-lobed. The number of lobes in the nucleus depends upon the age of cell. In younger cells, the nucleus is not lobed, while the older neutrophils may have 2 to 5 lobes in their nucleus. The neutrophils are amoeboid in nature. They contain actin and myosin microfilaments, which are responsible for their amoeboid motility. They are present in high concentration in pus. Since the pus mainly comprises of dead neutrophils, there will not be much pus formation in case of deficiency of neutrophils.

Production of neutrophils increases following tissue damage. Reduction in the neutrophil count may result in the development of throat ulcers because neutrophils are not available to kill bacterial invaders. Glucocorticoids can cause suppression of lymphocytes and eosinophils, not neutrophils.

Basophils
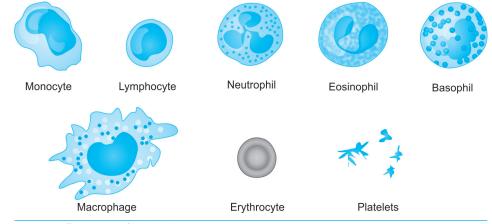
Basophils also have coarse granules in the cytoplasm, which stain purple-blue with methylene blue. Basophilic granules contain histamine and heparin.
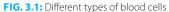
Eosinophils

Eosinophils comprise nearly 1–4% of white cells. Eosinophils have coarse (larger) granules in the cytoplasm, which stain pink or red with eosin (eosinophilic). Nucleus is bilobed and spectacle-shaped. Eosinophils attack parasites and produce leukotrienes. They are also involved in mucosal immunity. Therefore, they are relatively common in the mucosa of the respiratory, urinary and alimentary tracts. They release cytokines, particularly interleukin-4 and platelet-activating factor (PAF). Their number increases in parasitic infections and allergic conditions.

Monocytes

Monocytes originate from stem cells in bone marrow. Activated T cells release GMCSF (granulocyte/macrophage

Monocyte   Lymphocyte   Neutrophil   Eosinophil   Basophil

Macrophage   Erythrocyte   Platelets

FIG. 3.1: Different types of blood cells

colony-stimulating factor), which stimulates monocyte stem cells to proliferate. Monocytes can increase in number when their parent cells are stimulated by factors released from activated lymphocytes. The half-life of monocytes in blood is approximately 72 hours. However, its half-life in tissues is unknown (maybe 3 months). Monocytes are the largest amongst all the leucocytes having a diameter varying between 14 μ and 18 μ. The cytoplasm is clear without granules. Nucleus is round, oval, horseshoe-shaped, bean-shaped or kidney-shaped and is placed either in the centre of the cell or pushed to one side. Therefore, a large amount of cytoplasm is seen. After 4–6 days in the circulation, monocytes migrate out to become tissue macrophages. Monocytes can transform into large multi-nucleated cells in certain chronic infections, e.g. the "giant cells" seen in tissues affected by tuberculosis and leprosy. The primary host response to bacterial infections is usually dependent on mononuclear phagocytes and neutrophils.

Lymphocytes

Similar to the monocytes, lymphocytes also do not have granules in the cytoplasm. Depending upon the size, lymphocytes are divided into two types, large lymphocytes (having a diameter of 10–12 μ) and small lymphocytes (having a diameter of 7–10 μ). Depending upon the function, lymphocytes are divided into two types: T lymphocytes (concerned with cellular immunity) and B lymphocytes (concerned with humoral immunity due to production of immunoglobulins). Immunoglobulins are made by ribosomes in lymphocytes. T lymphocytes comprise majority of circulating lymphocytes in plasma. After birth, some lymphocytes are formed in the bone marrow, but most are formed in the lymph nodes, thymus and spleen from precursor cells that originally came from the bone marrow. For details related to lymphocytes, kindly refer to chapter 6.

Blood Coagulation

Platelets

Platelets or thrombocytes are the formed elements of blood. Platelets are small colourless, non-nucleated and moderately refractive bodies. Platelets are membrane-encapsulated fragments of megakaryocytes. They are more numerous than the white cells by a factor of 20 or more. They do not contain any nucleus. However, the cytoplasm contains electron dense granules, lysosomes and mitochondria. Two types of granules are present in cytoplasm of platelets: alpha granules and the dense granules. Substances present in these granules are summarized in **Table 3.4**. Platelets are about half the diameter of red cells, which in turn are smaller than white cells. Aspirin is a drug having no impact on the coagulation cascade. It just causes a decline in the platelet count.

TABLE 3.4	Substances present in the platelet granules
Alpha granules	**Dense granules**
• *Clotting factors*: Fibrinogen, factors V and XIII • Platelet-derived growth factor • Vascular endothelial growth factor • Basic fibroblast growth factor • Endostatin • Thrombospondin	• Nucleotides • Serotonin • Phospholipids • Calcium • Lysosomes

Although platelets have no nucleus, they are metabolically active and are able to express membrane receptors and release stored substances when triggered. However, due to the absence of nucleus, they are unable to produce new proteins. Therefore, aspirin and other antiplatelet drugs are likely to affect their functioning for the remainder of the platelet lifespan. Lack of platelets is known as thrombocytopenia. Platelet lifespan is approximately 9–10 days in normal individuals. There is an increase in the number of platelets after injury and surgery because this increases the tendency of blood to clot. When in contact with collagen, the platelets develop pseudopodia, thereby adhering to the collagen and to one another. Platelets are capable of producing nitric oxide, prostaglandins and thromboxane, but not prostacyclin. Normal platelet count varies between 200,000 and 400,000/mm³. Platelets have a role in blood clotting. Platelets are responsible for the formation of intrinsic prothrombin activator, which is responsible for the onset of blood clotting **(Fig. 3.2)**. They help in clot retraction and in haemostasis or prevention of blood loss immediately following injury by causing constriction of the blood vessels and sealing the site of injury by forming a temporary platelet plug.

Coagulation Pathway

Blood coagulation or clotting is a process in which the blood changes its form from liquid to a gel, resulting in formation of a clot. Coagulation of blood occurs through a series of reactions due to the activation of a group of substances (clotting factors), which are essential for clotting. Thirteen clotting factors (factors I to XIII) are identified. In general, blood clotting occurs in three stages:
1. Formation/activation of prothrombin activator
2. Conversion of prothrombin into thrombin
3. Conversion of fibrinogen into fibrin

Activation of the prothrombin activator occurs via two pathways, extrinsic and intrinsic pathway **(Fig. 3.2)**. In the intrinsic pathway, endothelial damage caused by rupture of blood vessels during injury results in the exposure of collagen. When factor XII comes in contact with collagen, a sequential series of events occurs eventually resulting in the formation of prothrombin activator.

In the extrinsic pathway, formation of prothrombin activator is initiated by tissue thromboplastin derived from the injured tissues.
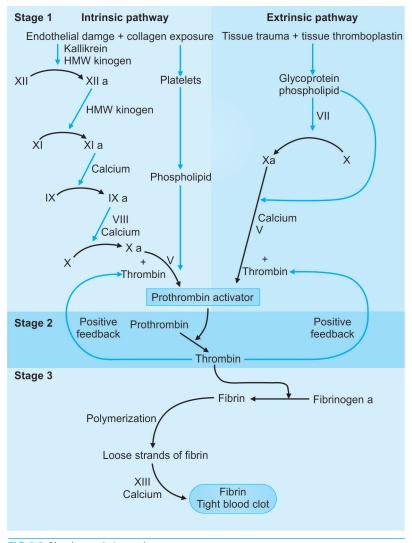
FIG. 3.2: Blood coagulation pathway
Abbreviation: HMW, high molecular weight

Prothrombin activator formed as a result of either intrinsic or extrinsic pathway cause conversion of prothrombin to thrombin in the presence of calcium (factor IV). Thrombin causes conversion of fibrinogen to fibrin. Fibrin monomers then polymerise with other monomer molecules to form locally arranged strands of fibrin. This is converted into a well-aggregated meshwork of stable clot in the presence of fibrin-stabilising factor XIII in the presence of calcium ions. Presence of calcium ions is essential for the clotting of blood. Removal of calcium ions prevents clotting. Vitamin K is required for blood clotting. It is required by the liver for synthesis of prothrombin and other factors.

Prevention of initial blood loss after injury: Bleeding from a small cut in the skin is normally diminished by local vascular spasm due to the effects of tissue damage and serotonin on vascular smooth muscles. Bleeding ceases within about 5 minutes in normal people. This is the upper limit of the normal "bleeding time". Bleeding from a small cut in the skin is greater from warm skin than from cold skin because warmth dilates blood vessels in the skin. Bleeding is reduced if the affected limb is elevated because intravascular pressure is reduced in an elevated limb.

Tests of Coagulation

Bleeding Time

Bleeding time (BT) is the time interval beginning from oozing of blood after a cut or injury till arrest of bleeding. Bleeding time is determined by platelets and by vascular contraction. Normal duration of bleeding time varies between 3 minutes and 6 minutes. It is prolonged in cases of purpura. Purpura is caused by capillary or platelet disorders. Widespread purpura occurs due to failure of platelet plugging of capillaries and may be due to a low platelet count or to capillary abnormality. These cases could be characterised by a low platelet count. Platelet count below $20–40 \times 10^9$ per litre accounts for serious bleeding.

The spontaneous bleeding from the gums, etc. seen in vitamin C deficiency (e.g. scurvy) is due to capillary abnormality and is not a clotting defect.

Clotting Time

Clotting time (CT) is the time interval from oozing of blood after a cut or injury till the formation of clot. Its normal duration varies between 3 and 8 minutes. It is prolonged in cases of haemophilia.

Prothrombin Time

The prothrombin time (PT) measures the clotting time from the activation of factor VII, through the formation of fibrin clot. It is the time taken by blood to clot after addition of tissue thromboplastin to it. This test measures the integrity of the extrinsic and common pathways of coagulation. Since the prothrombin time assesses the extrinsic pathway, it is prolonged in cases of abnormalities of factors VII, X, V or II. The normal prothrombin time is 16–18 seconds. Causes of prolonged PT include warfarin therapy, unfractionated heparin (not low molecular weight), disseminated intravascular coagulation (DIC) and liver diseases (e.g. cirrhosis).

The coumarin/warfarin interferes with vitamin K metabolism, which inhibits the γ-carboxylation of factors II, VII, IX and X and thus prolongs the prothrombin time. Vitamin K restores a normal prothrombin time and is used therapeutically if the PT is prolonged or the international normalised ratio (INR) is very high.

Activated Partial Thromboplastin Time

Activated partial thromboplastin time (APTT) is the time taken for the blood to clot following the addition of an activator such as phospholipid, along with calcium to it. It is also called activated partial prothrombin time. Therefore, the activated partial thromboplastin time measures the integrity of the intrinsic and common pathways of coagulation. Normal duration of partial prothrombin time is 30–45 seconds. Apart from detecting abnormalities in blood clotting, measurement of APTT is also used for monitoring the treatment effects with heparin, a commonly used anti-coagulant during pregnancy. APTT is prolonged in cases of heparin or warfarin therapy as well as deficiency or inhibition of factors II, V, VIII, IX, X, XI and XII.

Heparin has an immediate effect on coagulation by potentiation of the formation of irreversible complexes between anti-thrombin and activated serine protease coagulation factors and thus has minimal effects on the prothrombin time. Low-molecular-weight heparin, on the other hand, specifically acts on factor Xa and thus is not monitored through either APTT or PT.

Haemophilia A

Haemophilia A is due to a recessive abnormality of the X chromosome and is associated with the deficiency of factor VIII. Haemophilia, therefore, affects the intrinsic pathway. Deficiency of factor VIII increases the clotting time. In these cases, abnormal bleeding does not occur until the factor VIII level falls below 50%. Haemophilia does not interfere with initial haemostasis due to vascular closure, so the bleeding time is normal as in this case. However, when the vascular spasm wears off, failure of clotting is revealed as a persistent ooze of blood. Treatment is by supplying the missing factor VIII.
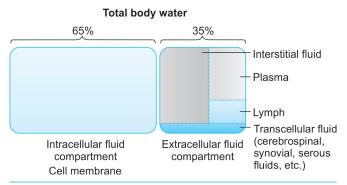
Homeostasis and the Fluid Balance

Body Fluids

In a normal healthy woman, roughly 70% of the total body weight comprises of water, which is distributed into two major compartments **(Fig. 3.3)**:

A. *Intracellular fluid (ICF)*: It comprises nearly two-third of the total body water.
B. *Extracellular fluid (ECF)*: It comprises nearly one-third of the total body water. Extracellular fluid can be divided into the following 5 categories:
   1. Interstitial fluid and lymph
   2. Plasma
   3. Fluid in bones
   4. Fluid in dense connective tissues like cartilage
   5. *Transcellular fluid*: This includes different body fluids such as cerebrospinal fluid, intraocular fluid, digestive juices, intrapleural fluid, pericardial fluid and peritoneal fluid, synovial fluid in joints and fluid in urinary tract.

Of the ECF, approximately 75–80% is interstitial and 20–25% is plasma. In a normal woman, plasma volume is about 2.5–3 L having a pH of 7.4. The normal plasma osmolality is approximately 300 mOsmol/L. Proteins present in the plasma account for only 1% of its osmolality. Plasma has the tonicity of a normal saline solution (0.9% sodium chloride). Normal sodium concentration of plasma is approximately 135–145 mmol/L. In the obese patients,

FIG. 3.3: Distribution of body fluids

ECF is relatively contracted. The following substances are found in higher concentrations intracellularly than extracellularly:

- Potassium
- Magnesium
- Phosphate
- Alpha-fetoprotein (AFP)
- Adenosine diphosphate (ADP)
- Adenosine monophosphate (AMP)

Sodium and chloride are in higher concentrations extracellularly.

Transport Mechanisms

Various transport mechanisms exist in the body, which account for the movement of substances within the cells and across the cell membranes. Some of the mechanisms for transport include diffusion, solvent drag, filtration, osmosis, non-ionic diffusion, carrier-mediated transport and phagocytosis.

Osmosis

Osmosis describes the movement of solvent from a region of low solute concentration to a region of high solute concentration across a semi-permeable membrane. Osmotic water movements ensure that the osmolality of the ICF becomes the same as that of ECF. The microvascular endothelium is not freely permeable to water and relies upon a number of processes including osmosis. The intracellular sodium concentration is exquisitely sensitive to the extracellular sodium with osmosis resulting in shifts in water between the compartments. The process of osmosis is opposed by the hydrostatic pressure. The hydrostatic pressure, which would stop osmosis from occurring, is the osmotic pressure of the solution, which can be calculated with help of the formula:

$$P = nRT/V$$

Where P = osmotic pressure; n = number of osmotically active particles; R = gas constant; T = absolute temperature; and V = volume

1 Osmol = molecular weight in grams/number of osmotically active particles in solution Therefore, for an ideal solution of glucose, which does not ionise in the solution:

1 Osmol = molecular weight/1 = 180 g

However, in case of a substance which dissociates to form ions in the solution, e.g. NaCl, which dissociates into two ions, 1 Osmol = molecular weight/2 = 58.5/2 = 29.2 g.

Freezing point depression of a solution is also caused by the number of osmotically active particles. Greater the concentration of the osmotically active particles, greater is the freezing point depression. In an ideal solution with no interaction, 1 mole of osmotically active particles per

litre depresses the freezing point by 1.86°C. Therefore, an aqueous solution that depresses the freezing point by 1.86°C is defined as containing 1 osmol/L. The major osmotic components of plasma are the cations, sodium and potassium and their accompanying anions along with glucose and urea. Concentration of sodium in plasma is 140 mmol/L. Therefore, sodium and accompanying anions contribute to an osmolality of 280 mOsmol/L. Concentration of potassium is 4 mmol/L. Therefore, potassium and accompanying anions contribute to an osmolality of 8 mOsmol/L. Glucose and urea contribute to an osmolality of 5 mOsmol/L each. Therefore, total plasma osmolarity accounts to 300 mOsmol/L. During pregnancy, this osmolarity may fall to at least 290 mOsmol/L.

At the arterial end of the capillary, the hydrostatic forces acting outwards are greater than the osmotic forces acting inwards. Therefore, there is a net movement of fluid out of the capillaries. At the arteriolar end of the capillary, the hydrostatic pressure is 37 mm of Hg; interstitial pressure is 1 mm of Hg and intravascular pressure is 25 mm of Hg. Therefore, the net force driving the water out is 37 – 1 – 25 = 11 mmHg.

On the other hand, at the venous end of the capillary, the hydrostatic forces acting outwards are less than the osmotic forces acting inwards. Therefore, there is a net movement of fluid inwards inside the capillaries. The capillary membrane is only permeable to water and small solutes. It is impermeable to the plasma proteins. At the venous end, the hydrostatic pressure is 17 mmHg, whereas the interstitial and intravascular pressure is same as that at the arteriolar end. The net driving force into the capillaries is 25 + 1 – 17 = 9 mmHg. Therefore, the fluid enters the capillaries at the venous end.

Diffusion

Non-ionised diffusion is a process in which there is a preferential transfer of the substance in non-ionised form. Cell membranes comprise of a lipid bilayer with specific transporter proteins embedded in it. Lipid soluble drugs, e.g. propranolol can cross the lipids of blood-brain-barrier or the placenta by non-ionised diffusion.

Carrier-Mediated Transport

This implies transport of substances across the cell membrane with the help of specific carriers. This could be of two types: facilitated transport or active transport.

Facilitated transport: In these cases, the transport of the substances is down the concentration gradient, from an area of high concentration to an area of low concentration aided by specific carrier molecule, e.g. uptake of glucose by the muscle cells facilitated by insulin.

Active transport: In these cases, the transport is up the concentration gradient, from an area of low concentration to

an area of high concentration, e.g. removal of sodium from the muscle cells by the ATPase dependent sodium pump.

Acid-Base Balance

pH is expressed as the negative logarithm of the [H$^+$] in moles/litre.

$$pH = \log\frac{1}{H^+}$$

Hydrogen ion (H$^+$) contains only a single proton (positively charged particle), which is not orbited by any electron. Therefore, it is the smallest ionic particle. An increase in H$^+$ ion concentration decreases the pH resulting in the development of acidosis. On the other hand, a reduction in H$^+$ concentration increases the pH, resulting in alkalosis. pH of the arterial blood normally ranges between 7.36 and 7.44. The normal H$^+$ concentration in the ECF is 38 to 42 nM/L. An increase in pH by onefold requires a tenfold decrease in H$^+$ concentration. In a healthy person, the pH of the ECF is 7.40 and it varies between 7.38 and 7.42. The maintenance of acid-base status is very important for homeostasis, because even a slight change in pH may cause serious threats to many physiological functions. PCO$_2$ raises [H$^+$] levels and hence lowers pH. [HCO$_3^-$] lowers [H$^+$] by buffering and hence raises pH. pH of urine is usually less than 7 because the normal diet leaves acidic, rather than alkaline, residues.

Determination of Acid-Base Status

It is difficult to determine the acid-base status in the ECF by direct methods. So, it is determined by an indirect method using Henderson-Hasselbalch equation by measuring the concentration of bicarbonate ions (HCO$_3^-$) and the CO$_2$ dissolved in the fluid. The pH is calculated as follows:

$$pH = pK + \log\frac{HCO_3^-}{CO_2}$$

Where pK is constant with pH of 6.1. Thus,

$$pH = 6.1 + \log\frac{HCO_3^-}{CO_2}$$

Compensatory Mechanisms

Whenever there is a change in pH beyond the normal range, some compensatory changes occur in the body to bring the pH back to normal level. The body has three different mechanisms for regulating the acid-base status:
1. Acid-base buffer system, which binds free H$^+$
2. Respiratory mechanism, which eliminates CO$_2$
3. Renal mechanism, which excretes H$^+$ and conserves the bases (HCO$_3^-$)

Amongst the three mechanisms, the acid-base buffer system is the fastest one and readjusts the body pH within seconds. The respiratory mechanism takes a few minutes, whereas the renal mechanism is slower and it takes few hours to few days to bring the pH back to normal. However, the renal mechanism is the most powerful amongst the three and helps in maintaining the acid-base balance of the body fluids.

Regulation of Acid-Base Balance by Acid-Base Buffer System

An acid-base buffer system is a mechanism that helps in regulating the acid-base mechanism via combination of a weak acid (protonated substance) with a base or the salt (unprotonated substance). Buffer system is the one that acts immediately to prevent the changes in pH.

Types of Buffer System

The following three types of buffer systems are present in the body:
1. Bicarbonate buffer system
2. Phosphate buffer system
3. Protein buffer system

Bicarbonate buffer system: Bicarbonate buffer system is present in ECF (plasma). It consists of the protonated substance, carbonic acid (H$_2$CO$_3$) which is a weak acid and the unprotonated substance, HCO$_3^-$, which is a weak base. HCO$_3^-$ is present in the form of salt, i.e. sodium bicarbonate (NaHCO$_3$). Carbon dioxide spontaneously converts to carbonic acid (H$_2$CO$_3$), and some of the carbonic acid spontaneously converts to bicarbonate (HCO$_3^-$) plus hydrogen ions (H$^+$).

Bicarbonate buffer system prevents the fall of pH in a fluid on addition of a strong acid like hydrochloric acid (HCl). Normally, when HCl is mixed with a fluid, pH of that fluid decreases quickly because the strong HCl dissociates into H$^+$ and Cl$^-$. However, upon addition of the bicarbonate buffer system (NaHCO$_3$) to the fluid with HCl, the pH is not altered much because the H$^+$ ions dissociated from HCl combine with HCO$_3^-$ ions (from NaHCO$_3$) to form a weak acid, H$_2$CO$_3$. This H$_2$CO$_3$ in turn dissociates into CO$_2$ and H$_2$O. This reaction can be summarised by the equation below:

$$HCl + NaHCO_3 \rightarrow H_2CO_3 + NaCl$$

Bicarbonate buffer system also prevents the increase in pH in a fluid to which a strong base like sodium hydroxide (NaOH) is added. Normally, when a base (NaOH) is added to a fluid, pH increases. It is prevented by adding H$_2$CO$_3$, which dissociates into H$^+$ and HCO$_3^-$ ions. The hydroxyl group (OH) of NaOH combines with H$^+$ and forms H$_2$O. At the same time, Na$^+$ combines with HCO$_3^-$ ions to form NaHCO$_3$. NaHCO$_3$ is a weak base and it prevents the increase in pH. This reaction is summarised by the below-mentioned equation:

$$NaOH + H_2CO_3 \rightarrow H_2O + NaHCO_3$$

Bicarbonate buffer system is not powerful like the other buffer systems. However, it plays an important role in maintaining the pH of body fluids in comparison to the other buffer systems because the concentration of two components (HCO_3^- and CO_2) of this buffer system is regulated separately by two different mechanisms. Concentration of HCO_3^- is regulated by kidney and the concentration of CO_2 is regulated by the respiratory system. Normal concentration of bicarbonate (HCO_3^-) is about 20–27 mM. It is increased with persistent vomiting but decreased in renal failure (acidosis) and severe diarrhoea.

Clinical Evaluation of Disturbances in Acid-Base Status—Anion Gap

Anion gap is an important measurement for the clinical evaluation of disturbances in acid-base status. Only few cations and anions are measured during routine clinical investigations. Commonly measured cation is sodium (136 mEq/L) and the unmeasured cations are potassium, calcium and magnesium. The prinipical anions, which are measured, are chloride (100 mEq/L) and bicarbonate (24 mEq/L) ions. The unmeasured anions are phosphate, sulphate, proteins in anionic form such as albumin and other organic anions like lactate. Difference between concentrations of unmeasured anions and unmeasured cations is called anion gap and is calculated as follows:

Anion gap = $[Na^+] - [HCO_3^-] - [Cl^-]$
= 136 – 24 – 100 mEq/L = 12 mEq/L

Normal value of anion gap ranges between 8 and 16 mEq/L. It increases when concentration of unmeasured anions (e.g. protein, lactate, etc.) increases and decreases when concentration of unmeasured cations decreases. An increase in the anion gap implies the presence of more unmeasured anions than usual. This can occur in situations such as ketoacidosis, lactic acidosis, hyperosmolar acidosis, and poisoning with salicylates, methanol, ethylene glycol, paraldehyde and hypoalbuminaemia. A reduced anion gap can occur in cases such as bromide poisoning and multiple myeloma.

Abnormalities of Acid-Base Balance

Acidosis

The abnormalities of acid-base balance can be categorized into acidosis (pH less than 7.36) and alkalosis (pH greater than 7.44). Acidosis is produced by two main mechanisms:
1. Increase in partial pressure of CO_2 in the body fluids, particularly in arterial blood
2. Decrease in HCO_3^- concentration

Since the partial pressure of CO_2 (pCO_2) in arterial blood is controlled by lungs, the acid-base disturbances produced by the changes in arterial pCO_2 are called the respiratory disturbances. Increased pCO_2 causes increased production of H_2CO_3. Dissociation of H_2CO_3 results in an augmented

TABLE 3.5	Values of pH and pCO_2 in acidosis and alkalosis	
Disorders	**pH**	**pCO_2 (mm of Hg)**
Normal	7.36–7.44	36–44
Respiratory acidosis	<7.36	>44
Respiratory alkalosis	>7.44	<36
Metabolic acidosis	<7.36	<44
Metabolic alkalosis	>7.44	>36

production of H^+ ions resulting in the development of respiratory acidosis. In the acidosis of the respiratory origin, primary abnormality is in the control of PCO_2. On the other hand, the disturbances in acid-base status produced by the change in HCO_3^- concentration are generally called the metabolic disturbances. Depending on the underlying primary abnormality, acidosis can be further classified as either respiratory or metabolic acidosis. Values of pH and pCO_2 in different types of acidosis and alkalosis are tabulated in **Table 3.5**.

Respiratory Acidosis

Respiratory acidosis is characterised by low pH and high PCO_2 levels. The basic abnormality is the failure of excretion of CO_2 from the lungs. Carbon dioxide in the blood dissolves to form carbonic acid in presence of the enzyme carbonic anhydrase. Carbonic acid in turn dissociates to form hydrogen and bicarbonate ions. In the long term, compensation of respiratory acidosis is done by kidneys by retaining the bicarbonate ions that increases the pH towards normal. Respiratory acidosis may be associated with conditions such as breath holding, excessive sedation, cerebrovascular accident affecting medulla oblongata, respiratory tract obstruction and obstructive airway disease.

Metabolic Acidosis

In these cases, the pH is low but the PCO_2 is not elevated. This occurs due to the following causes:
- *Excessive acid production*: Excessive acid production can occur in conditions such as diabetic ketoacidosis and starvation (due to the production of ketone bodies). Methanol poisoning in which methanol is metabolised to formaldehyde which subsequently forms formic acid is another cause of excessive acid production.
- *Impaired acid excretion*: Failure of acid excretion occurs in conditions such as chronic renal failure. This specifically occurs in cases of renal tubular acidosis where the patients are not initially uraemic but uric acid excretion by the kidneys is impaired. Uraemia is a well-recognised cause for metabolic acidosis, although the precise mechanism remains unclear. It may be related to the diversion of glutamate metabolism to the liver and the consequent bicarbonate consuming effect of hepatic ureagenesis. Acetazolamide is a diuretic drug that inhibits ammonia formation within the kidneys, thereby causing metabolic acidosis.

Excessive loss of alkali: Excessive loss of alkali occurs in patients with pancreatic fistula or prolonged diarrhoea since in these cases, the body fluids lost are alkaline in nature.

Alkalosis

Alkalosis is the increase in pH (decrease in H^+ concentration) above the normal range. Alkalosis can be produced due to the following causes:

- Decrease in partial pressure of CO_2 in the arterial blood
- Increase in HCO_3^- concentration

Depending on the underlying primary abnormality, alkalosis can be further classified as either respiratory alkalosis or metabolic alkalosis. In the alkalosis of the respiratory origin, primary abnormality is in the control of PCO_2.

Respiratory Alkalosis

This is a condition that is characterised by high pH and low PCO_2. This is induced by hyperventilation. The commonest clinical presentation of respiratory alkalosis is anxiety (due to an acute fall in the concentration of H^+ ions), paraesthesiae, tetany, etc. Tetany occurs because more plasma protein is ionised when the pH is high. This protein binds more calcium, lowering the ionised levels of calcium. In pregnancy, there is hyperventilation. However, there is no change in the pH because the kidneys excrete sufficient amounts of bicarbonate to compensate for the fall in CO_2. When ascending to high altitude, the decreased oxygen concentration stimulates respiration resulting in respiratory alkalosis.

Metabolic Alkalosis

In these cases, the pH is high, but PCO_2 is not reduced. This commonly occurs due to the loss of acidic fluid commonly due to prolonged vomiting. Pyloric stenosis is also an important cause of metabolic alkalosis. It can also occur due to excess alkali ingestion, observed in patients taking antacids for peptic ulceration. Metabolic alkalosis frequently accompanies hypokalaemia. Aldosterone causes reabsorption of sodium ions and secretion of hydrogen ions promoting alkalosis. Thiazide diuretics can also cause metabolic alkalosis.

Vomiting of gastric contents alone leads to a loss of hydrochloric acid and hence causes alkalosis; however, vomiting of contents from the lower gastrointestinal tract results in a loss of chloride ions and an increase in bicarbonate in the ECF. Vomiting therefore causes metabolic alkalosis.

Electrolyte Imbalance

Hyponatraemia

Hyponatraemia is the commonest biochemical abnormality occurring in up to 10% of hospitalised patients. Causes of hyponatraemia include the following:

- Syndrome of inappropriate antidiuretic hormone or SIADH (as in subarachnoid haemorrhage and pneumonia)
- Hypothyroidism
- Addison's disease
- Diuretic therapy
- Liver disease
- Congestive cardiac failure
- Bronchial carcinoma with SIADH

Though ecstasy use is not strictly a cause of hyponatraemia, hyponatraemia has been responsible for deaths in association with ecstasy use due to excessive fluid consumption. Based on the assessment of the volaemic status of the patient, the causes of hyponatraemia can be sub-divided into three categories:

1. *Euvolaemia*: For example, SIADH, high water intake, hypothyroidism, etc. SIADH can be produced by conditions such as major surgery, pneumonia, subarachnoid haemorrhage, meningitis as well as drugs.
2. *Hypovolaemia*: For example, diarrhoea, vomiting, diuretics, renal tubular dysfunction, Addison's disease, etc.
3. *Hypervolaemia*: For example, cirrhosis, heart failure, cerebral contusion, nephrotic syndrome and myxoedema.

Hypernatraemia

Cushing's syndrome is associated with sodium retention. Steroid therapy and Conn's syndrome also cause salt and water retention, and hypernatraemia. Carbenoxolone causes pseudo-hyperaldosteronism with hypertension, hypernatraemia and hypokalaemia. Diabetes insipidus and the administration of hypertonic saline also cause hypernatraemia.

Hypokalaemia

Various causes of hypokalaemia include the following:

- Prolonged vomiting
- Diabetic ketoacidosis
- Conn's syndrome
- Bartter's syndrome
- Mineralocorticoid excess (hyperaldosteronism)
- Diuretics: bendroflumethiazide, furosemide
- Chronic diarrhoea
- Hormone secreting tumours of bronchus [adrenocorticotropic hormone (ACTH)]
- Familial hypokalaemic periodic paralysis (rarely)
- *Drugs causing hypokalaemia*: The following drugs can cause hypokalaemia:
  - Salbutamol for asthma, especially if administered in high doses in nebulisers for acute asthma
  - Vitamin B_{12} for the treatment of pernicious anaemia, particularly at the beginning of treatment
  - Carbenoxolone may cause hypokalaemic hypertension.

- Liquorice also causes hypokalaemia through inhibition of 11-beta-hydroxysteroid dehydrogenase.
- Bendroflumethiazide, a thiazide diuretic and furosemide promote potassium excretion.

Causes of hypokalaemia can be divided into the following:
- ❑ *Trans-cellular shift*: For example, alkalosis, administration of insulin, beta-agonists, ect.
- ❑ *Renal losses*: For example, diuresis, diabetic ketoacidosis after therapy, Conn's disease
- ❑ *Extra-renal losses*: For example, diarrhoea, nasogastric suction, laxative or enema abuse, vomiting, biliary drainage or entero-cutaneous fistulae.
- ❑ *Decreased intake*: For example, malnutrition, alcoholism.

Hyperkalaemia

Causes of hyperkalaemia include the following:
- ❑ Type IV renal tubular acidosis
- ❑ Hyperparathyroidism
- ❑ Hypoadrenalism
- ❑ CAH, Addison's disease, angiotensin-converting enzyme (ACE) inhibitors and rhabdomyolysis tend to cause hyperkalaemia.

When electrocardiogram abnormalities are present, treatment of hyperkalaemia is an emergency. The treatment for hyperkalaemia comprises of the following:
- ❑ Calcium chloride
- ❑ Sodium bicarbonate
- ❑ Dextrose/insulin
- ❑ Beta-agonists
- ❑ Loop diuretics
- ❑ Drugs to bind potassium in the gastrointestinal tract
- ❑ Dialysis
- ❑ Diagnosis and treatment of the underlying condition

Hypocalcaemia

Tetany occurs in association with low calcium or magnesium concentrations. Hypocalcaemia may be associated with convulsions, papilloedema, psychosis, muscle cramps, spasm and tetany. Raised intracranial pressure is associated with hypocalcaemia as is papilloedema, which may be related to an ischaemic optic neuropathy. Hypocalcaemia is also associated with a long QT interval.

Physiology of Respiratory System

Lung Volumes

Tidal volume (V_T): This is the volume of air that moves in and out of the lungs during each breath at rest. It is usually about 500 mL. Tidal volume signifies the normal depth of breathing.

Inspiratory reserve volume: The inspiratory reserve volume (IRV) can be defined as an additional volume of air that can be inspired forcefully after the end of normal inspiration. Its normal value is 3,300 mL.

Expiratory reserve volume: Expiratory reserve volume (ERV) can be defined as the additional volume of air that can be expired out forcefully after normal expiration. It measures about 1,000 mL.

Residual volume: Residual volume (RV) is the gas remaining in the lungs at the end of a forced expiration. Normally, lungs cannot be emptied completely even following forceful expiration. Some quantity of air always remains in the lungs after forced expiration. Men, on an average, have bigger thoracic cages than women. Therefore, residual volume is greater in men than in women. It is on an average around 1–1.5 litres. Residual air cannot be exhaled out; therefore, it cannot be measured using spirometry. It is measured indirectly by a dilution technique. Residual volume increases with age since the elastic recoil of the lungs decreases with aging. Significance of residual volume is as follows:
- ❑ It helps in aeration of the blood in between breathing and during expiration.
- ❑ It helps in maintenance of the contour of the lungs.
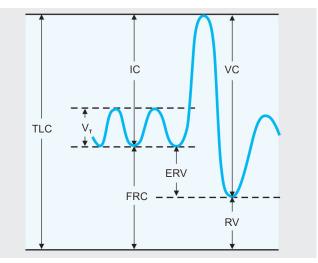
Lung Capacities

Static lung capacities are the combination of two or more lung volumes. Static lung capacities are of four types and are as follows **(Fig. 3.4)**.

Inspiratory Capacity

Inspiratory capacity (IC) is the maximum volume of air that is inspired after normal expiration. It includes tidal volume and IRV.

$$IC = TV + IRV = 500 + 3,300 = 3,800 \text{ mL}$$

FIG. 3.4: Lung capacities
Abbreviations: TLC, total lung capacity; VT, tidal volume; IC, inspiratory capacity; FRC, functional residual capacity; ERV, expiratory reserve volume; VC, vital capacity; RV, residual volume

Vital Capacity

Vital capacity (VC) is the maximum volume of air that can be expelled out forcefully after a deep (maximal) inspiration. VC includes IRV, tidal volume and expiratory reserve volume.

VC = IRV + TV + ERV = 3,300 + 500 + 1,000 = 4,800 mL

Functional Residual Capacity

Functional residual capacity (FRC) is the volume left in the lungs after the expiration of V_T. Therefore, FRC is a combination of residual volume with ERV.

FRC = ERV + RV = 1,000 + 1,200 = 2,200 mL

Total Lung Capacity

Total lung capacity (TLC) is the volume of air present in lungs after a deep (maximal) inspiration. It includes all the volumes.

TLC = IRV + TV + ERV + RV = 3,300 + 500 + 1,000 + 1,200
= 6,000 mL

Forced Vital Capacity

Forced vital capacity (FVC) is the volume of air that can be exhaled out forcefully and rapidly after a maximal or deep inspiration. It is a dynamic lung capacity. Normally, FVC is equal to VC. However, in some pulmonary diseases, FVC is reduced.

Forced Expiratory Volume

Forced expiratory volume (FEV) is the volume of air that can be expired forcefully in a given unit of time (after a deep inspiration). It is also called timed vital capacity or forced expiratory vital capacity (FEVC). It is a dynamic lung volume. Forced expiratory volume in a person with normal respiratory functions is described in **Table 3.6**.

The normal value of FEV1 in an adult patient is about 85% at the age of 20 years, falling to about 70% at the age of 60–70 years. In patients with restrictive diseases such as pulmonary fibrosis, lung volumes (particularly RV and FRC) are reduced. This can result in inadequate ventilation and

oxygenation, which may eventually result in respiratory failure.

Patient with restrictive disease may expire a volume greater than predicted percent of VC in the first second because the airways are not obstructed and vital capacity volume is reduced. Women have, on average, smaller vital capacities than males. Therefore, an adult female would be expected to expire a greater percent of VC in 1 second than a male of the same age. In presence of a moderately severe obstructive disease, the patient may take more than 5 seconds to complete expiration.

With normal lungs, the person should achieve a peak flow rate of at least 200 litres/minute. Most subjects, other than small elderly females, would have peak flow rates much higher than this.

Respiratory Diseases

Diseases of respiratory tract are classified into two types: (1) restrictive respiratory disease; (2) obstructive respiratory disease. These two types of respiratory diseases can be differentiated using pulmonary function tests. Two main components of the pulmonary function test are measured to make the diagnosis: the forced expiratory volume in the first second (FEV_1), which is the greatest volume of air that can be breathed out in the first second of a breath; and the forced vital capacity (FVC), which is the greatest volume of air that can be breathed out in a single large breath. Normally, FEV in the first second is 75–80% of the FVC.

Restrictive Respiratory Disease

Restrictive respiratory disease is the abnormal respiratory condition characterised by difficulty in inspiration. Expiration is not affected. Restrictive respiratory disease may be because of abnormality of lungs, thoracic cavity or/and nervous system. Some restrictive respiratory diseases include poliomyelitis, myasthenia gravis, flail chest, pleural effusion, etc.

In restrictive lung disease, both forced expiratory volume in one second (FEV_1) and forced vital capacity (FVC) are reduced. However, the decline in FVC is more than that of FEV1, resulting in a ratio of FEV_1/FVC which is usually higher than 80%.

Obstructive Respiratory Disease

Obstructive respiratory disease is the abnormal respiratory condition characterised by difficulty in expiration. Obstructive respiratory diseases include asthma, chronic bronchitis, emphysema, cystic fibrosis, epiglottitis, laryngotracheobronchitis, etc. Loss of pulmonary elastic tissue in "emphysema" reduces anatomical dead space because destruction of elastic fibres holding airways cause them to narrow down. Physiological dead space also increases as the walls between alveoli break down to form

TABLE 3.6	Forced expiratory volume in persons with normal respiratory functions	
Forced expiratory volume (FEV)	Definition	Normal values
FEV1	Volume of air expired forcefully in 1 second	83% of total vital capacity
FEV2	Volume of air expired forcefully in 2 seconds	94% of total vital capacity
FEV3	Volume of air expired forcefully in 3 seconds	100% of total vital capacity

large sacs. Residual volume is increased as airways close more readily than usual during expiration. Vital capacity decreases as the residual volume increases. The percentage of the vital capacity expired in one second (FEV1) also decreases in a typical obstructive airway disease. In obstructive lung disease, however, FEV1 is reduced while FVC remains stable, thereby resulting in a lower FEV1/FVC ratio. FEV_1/FVC ratio of less than 70% is usually diagnostic of the disease.

Respiratory Centre

Respiratory centre is responsible both for controlling the depth of respiration and its rhythmicity. The primary centres for the control of respiration are in the medulla and pons of the brainstem. Dorsal and ventral group of nuclei in the medulla interact with the pontine respiratory group and the medullary Bötzinger complex. Multiple connections between each of these centres provide stimulatory output to the intercostal muscles and the phrenic nerve. Respiratory neurons are of two types: inspiratory and expiratory. When the inspiratory neurons are stimulated at the respiratory centre, the expiratory neurons are inhibited and vice versa. The respiratory centre receives input from the higher voluntary centres. Therefore, factors such as pain and emotion may increase ventilation. The nucleus tractus solitarius (present in the upper part of the medulla oblongata) is a major integrating centre for the autonomic nervous system. It communicates with the medullary respiratory centres. However, in most healthy patients, ventilation is automatic and it is not necessary to be consciously aware of the need to breathe. Ventilation is stimulated by hypoxia, hypercapnia and acidosis and suppressed by the opposite.

During normal breathing at rest, inspiration is an active process brought about by the activity of diaphragm only. Diaphragm is driven by the activity of phrenic nerve (C3–C5). Increasing respiratory work requires activation of the intercostal muscles and accessory muscles of respiration to support ventilation. Intercostal muscles are innervated by T1–T11 nerves. In early inspiration, there is a fall in intrapulmonary pressure. This creates a pressure gradient between mouth and lungs. There is also a fall in the intrathoracic pressure. Pressure in the superior vena cava falls as intrathoracic pressure falls. Intra-abdominal pressure rises due to the descent of diaphragm. Dead space PO_2 rises as the oxygenated inspired air replaces deoxygenated alveolar air.

On the other hand, expiration is an entirely passive process. Reduction in the activity of phrenic nerve is followed by diaphragmatic relaxation before the beginning of next inspiration.

The most important input for the respiratory centre comes from the chemoreceptors: central chemoreceptors on the surface of upper medulla (but separate from the medullary respiratory centres) and the peripheral

TABLE 3.7	**Chemoreceptors**	
Characteristic feature	**Central chemoreceptors**	**Peripheral chemoreceptors**
Location	Medulla	Carotid body and the aortic arch
Activating stimuli	Reduced pH of cerebrospinal fluid (caused by increased $PaCO_2$)	Reduced PaO_2, increased $PaCO_2$ and reduced serum pH (regardless of the cause)
Overall response	Most of the response to pH	All the response to oxygen and metabolic acidosis

chemoreceptors around the aortic arch and the carotid body **(Table 3.7)**. Chemoreceptors help in achieving the homeostatic role of the respiratory centre by keeping the blood gases (oxygen and carbon dioxide) and pH constant. The aortic arch receptors are innervated by the vagus nerve, while the carotid body chemoreceptors are innervated by the glossopharyngeal nerve. The peripheral chemoreceptors are the only sites for the response to hypoxia and metabolic acidosis. The peripheral chemoreceptors are sensitive to changes in PCO_2, pH and PO_2. On the other hand, central chemoreceptors are probably only sensitive to the changes in pH. Any effect of the change in PCO_2 and PO_2 is mediated by the ensuing change in pH. Chemoreceptors can be stimulated by chemicals such as cyanide, nicotine, nikethamide and doxapram. Doxapram is the only respiratory stimulant used in clinical practice because it does not cause other stimulatory side effects (e.g. risk of convulsions). Doxapram is used as a central ventilatory stimulant in the treatment of apnoea associated with chronic carbon dioxide retention. Nikethamide is also a stimulant that affects the respiratory cycle.

The Hering-Bruer Reflex

Hering-Bruer reflex is a mechanism contributing to the cessation of inspiration and initiation of expiration. As the lung expands, slowly adapting stretch receptors in the small airways of the lungs are activated. Afferent signals are then conveyed to the nucleus tractus solitarius through the vagus nerve. The reflex output is a reduction in the activity of phrenic nerve.

Respiratory Minute Ventilation

As the name suggests, respiratory minute ventilation is the volume of air breathed (exhaled or inhaled) from a person's lungs per minute.

$$\text{Minute ventilation (V)} = V_T \times \text{frequency (f)}$$

Where V_T is the tidal volume and f is the frequency of respiration or the respiratory rate.

The importance of respiratory minute ventilation lies in lieu of its relationship with the blood carbon dioxide levels. Blood carbon dioxide levels vary inversely with the minute ventilation. Normal minute ventilation in adults is about 5–8 L/minute.

Dead Space

This can be defined as the volume of air in the airways which in unavailable for the gaseous exchange. It has two components:

1. *Anatomical dead space*: This is the total volume of air present in the conducting zone, i.e. the conducting bronchioles and above, where no gaseous exchange can occur. It measures about 150 mL.
2. *Alveolar dead space*: This is the volume of air present in the respiratory zone, i.e. respiratory bronchioles and below. It is zero in healthy people and increases in pathological conditions (e.g. pleural effusion where the alveoli fill with fluid) or in emphysema (where the alveoli rupture).

Lung Compliance and Elasticity

Pulmonary compliance can be defined as the ability of the lungs to expand and stretch, and is determined by pulmonary volume and elasticity. The compliance of lungs and chest wall is expressed as volume change per unit change in pressure. The normal value is about 0.1 litre/cm H_2O (1 litre/kPa).

Compliance = Change in volume/change in pressure

A high degree of compliance refers to the requirement of only a small amount of pressure to produce a large increase in the volume. Reduced lung compliance implies that a greater change in pressure is required for a given change in volume, e.g. atelectasis, oedema, fibrosis, loss of surfactant, etc. Pulmonary compliance affects ventilation because patients whose lungs have poor compliance must make greater effort to inhale in a normal volume of air. This is likely to result in dyspnoea on exertion. Elasticity is inversely proportional to the compliance.

Surface Tension and Pulmonary Surfactant

Pulmonary compliance also depends on the surface tension of the mucoid lining of the alveoli. Pulmonary surfactant decreases the surface tension, thereby increasing the pulmonary compliance and reducing the effort required to expand the lungs.

Two factors are responsible for the collapsing tendency of lungs: elastic property of lung tissues and the surface tension. Elastic tissues of lungs show constant recoiling tendency and try to collapse the lungs. Surface tension, on the other hand, is the tension exerted by the fluid secreted from alveolar epithelium on the surface of alveolar membrane.

Fortunately, there are some factors, which save the lungs from collapsing. These include the negative intrapleural pressure and the presence of surfactant, which helps in reducing surface tension and prevents the collapsing tendency produced by surface tension.

Surfactant is an amphiphilic fluid produced by the type II pneumocytes of the alveolar epithelium. It is a naturally occurring substance that predominantly comprises of lipids such as dipalmitoylphosphatidylcholine which are linked with a series of surfactant proteins (A, B, C and D) and organised into hydrophobic and hydrophilic complexes. Pulmonary surfactant is predominantly dipalmitoylphosphatidylcholine with lesser amounts of other phospholipids including phosphatidylglycerol, phosphatidylethanolamine and phosphatidylinositol. All are detectable within the amniotic fluid. Surfactant production begins in the foetus relatively late in pregnancy. Therefore, its levels may be reduced in cases of premature infants. Respiratory distress syndrome, which is a major cause of mortality and morbidity, commonly occurs due to the lack of surfactant and lung immaturity. Intramuscular injection of dexamethasone and betamethasone is associated with increased production of the surfactant and enhanced lung maturity. Therefore, its administration may be beneficial in cases where there is a risk of delivery before 34 weeks of gestation.

The surfactant helps in reducing the surface tension, which has the following consequences:

❑ Increased lung compliance requiring reduced effort for breathing
❑ The internal pressure required to maintain the alveolar inflation is reduced, thereby preventing the smaller alveoli from emptying into the larger ones.

Gaseous Exchange

Once the air is inhaled, gaseous exchange occurs through the passive diffusion of gases between the alveoli and the adjacent blood vessels, i.e. the pulmonary capillaries. This way, the deoxygenated blood from the body transported to the lungs gains oxygen and loses carbon dioxide. The oxygenated blood, which is depleted of carbon dioxide, is then carried back to the heart by the pulmonary veins. From here, it is transported to the rest of the body.

Transportation of Oxygen

Oxygen is transported from alveoli to the tissues by blood in two forms: as simple physical solution and in combination with haemoglobin. Oxygen dissolves in water of plasma and is transported in this physical form. However, the amount of oxygen transported in this way is very negligible, only about 3% of total oxygen in blood. Transportation of oxygen in combination with haemoglobin as oxyhaemoglobin, accounts for nearly 97% of oxygen. Combination of oxygen with haemoglobin occurs only as a physical combination, i.e. oxygenation and not oxidation because the iron atoms in haemoglobin remain in the ferrous form. Oxygen can be readily released from haemoglobin when it is required.

Oxygen carrying capacity of haemoglobin: Oxygen carrying capacity of haemoglobin is the amount of oxygen transported by 1 gram of haemoglobin. It is 1.34 mL/g.

Transport of Carbon Dioxide

Carbon dioxide is transported by the blood from cells to the alveoli. Carbon dioxide is transported in the blood in the four following ways:
1. As dissolved form (7%)
2. As carbonic acid (negligible)
3. As bicarbonate (63%)
4. As carbamino compounds (30%)

Combination of carbon dioxide with water results in the formation of carbonic acid inside the RBCs in presence of the enzyme carbonic anhydrase. Since carbonic anhydrase is present only in the RBCs, not in the plasma, formation of carbonic acid is 200–300 times more in RBCs than in plasma. Carbonic acid is highly unstable and nearly 99.9% dissociates to form bicarbonate and hydrogen ions. As the concentration of bicarbonate ions increases in the cells, more and more diffuses out through the cell membrane into the plasma **(Fig. 3.5)**.

Chloride shift or Hamburger's phenomenon: This is the exchange of chloride ions for the bicarbonate ions across the RBC membrane. Chloride shift occurs when carbon dioxide enters the blood from the tissues. As the negatively charged bicarbonate ions move out of RBCs into the plasma, negatively charged chloride ions move into the RBCs in order to maintain the electrolyte balance.

Reverse chloride shift: Reverse chloride shift is the process by which chloride ions are moved back into plasma from the RBC. The reverse chloride shift occurs in lungs. This process helps in elimination of carbon dioxide from the blood.

When blood reaches the alveoli, sodium bicarbonate in plasma dissociates into sodium and bicarbonate ions. Bicarbonate ion moves into the RBC. This makes chloride ion to move out of the RBC into the plasma, where it combines with sodium and forms sodium chloride. Bicarbonate ion inside the RBC combines with hydrogen
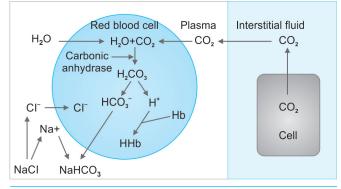
ion and forms carbonic acid, which dissociates into water and carbon dioxide. Carbon dioxide is then expelled out.

Nearly 30% of carbon dioxide is transported as carbamino compounds. In these cases, carbon dioxide combines with haemoglobin and plasma proteins. Carbaminohaemoglobin and carbamino proteins are together known as carbamino compounds.

Haemoglobin

Oxygen combines with the iron in haem part of haemoglobin. Haemoglobin is a tetrameric protein comprising of four sub-units. Each sub-unit comprises of a polypeptide chain and a haem group. Each molecule of haemoglobin contains four atoms of iron. Each iron atom combines with one molecule of oxygen. Iron of the haemoglobin is present in ferrous form. The four sub-units are linked to each other via non-covalent bonds. The four main types of sub-units, which can be present in haemoglobin, are alpha (α), beta (β), delta (δ) and gamma (γ). Different combinations of these different types of sub-units produce different types of haemoglobins **(Table 3.8)**. As described in this table, the most predominant haemoglobin type present in the early foetal life is foetal haemoglobin. Most adult type appears in the foetus around mid-gestation and disappears by four to see months of life. Foetal haemoglobin has a similar oxygen-carrying capacity as the adult haemoglobin. Foetal haemoglobin binds 2,3-diphosphoglycerate less ardently than does adult haemoglobin. It also has a higher affinity than the adult form for oxygen at low PO_2. This aids oxygen transfer in the placenta.

Oxygen-Haemoglobin Dissociation Curve

The oxygen dissociation curve (ODC) is a plot of oxygen saturation of haemoglobin against PO_2 (partial pressure of oxygen) at 37 degrees Celsius. This is a sigmoid-shaped (not hyperbolic) curve due to the increasing affinity of haemoglobin for successive oxygen molecules after binding to the first one **(Fig. 3.6)**. The middle range of the curve is therefore particularly important, because small changes in partial pressure may cause large changes in saturation. Certain factors may result in "shifts" in the curve. These are described next in the text.

Shift of Oxygen Dissociation Curve to the Left

A left shift reduces oxygen release to the tissues by increasing the affinity of haemoglobin to bind with oxygen. Factors which shift the ODC to the left include the following:
❏ Foetal haemoglobin, methaemoglobin and carboxyhae-moglobin all shift the curve to the left.
❏ The curve is shifted to the left in all the situations opposite to those causing a rightward shift

FIG. 3.5: Transport of carbon dioxide in form of bicarbonate

TABLE 3.8	Different types of haemoglobin		
Characteristic	**Haemoglobin A**	**Haemoglobin A2**	**Haemoglobin F**
Clinical significance	Main type of haemoglobin in adults	Has a minor role in normal adults, but may assume importance in individuals with β chain abnormalities	Main type of haemoglobin present in the foetus allowing exchange of gases between placenta and foetal circulation.
Proportion of total haemoglobin in an adult	98%	1%	1%
Proportion of total haemoglobin in a foetus	5–10%	0	90–95%
Types of globin chains	Two α chains; two β chains	Two α chains; two δ chains	Two α chains; two γ chains
Binding to 2,3-diphosphoglycerate	Binds 2,3-diphosphoglycerate to reduce oxygen affinity	Unable to bind with 2,3-diphosphoglycerate	Unable to bind with 2,3-diphosphoglycerate, so has a higher affinity for oxygen
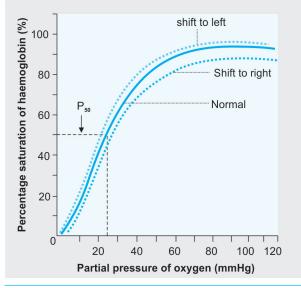
FIG. 3.6: Oxygen-haemoglobin dissociation curve

Shift of Oxygen Dissociation Curve to the Right

Factors which shift the ODC to the right reduce the affinity of haemoglobin for binding with oxygen, which favours the unloading of oxygen to the tissues. Some of these conditions include the following:

❑ Haemoglobin S shifts the curve to the right.
❑ Increased red cell concentration of 2,3 diphosphoglycerate (2,3 DPG)
❑ Anaemia and heart failure also improve unloading of oxygen.
❑ In actively metabolising tissues, oxygen needs to be released from haemoglobin therefore its affinity for oxygen must be reduced in these circumstances. These situations include high temperature, low oxygen and high pCO_2 levels (hypercapnia or Bohr effect). Bohr effect implies that the affinity of oxygen to bind with haemoglobin is inversely related both to the acidity and the concentration of carbon dioxide. All the factors, which shift the oxygen dissociation curve to right help in enhancing the Bohr effect.

Haldane's effect: Increased concentration of carbon dioxide will displace oxygen from haemoglobin and binding of oxygen to haemoglobin will displace carbon dioxide from the blood. This is known as the Haldane effect.

❑ The ODC is shifted to the right by acidosis (reduced pH) and hyperthermia, which favour the unloading of oxygen to the tissues.

Role of 2,3-DPG

2,3-diphosphoglycerate (2,3-DPG) is created in erythrocytes during glycolysis. 2,3-DPG cross-links the beta chains of haemoglobin and stabilises it in the deoxygenated state. Increased concentration of 2,3-diphosphoglycerate lowers the affinity of haemoglobin A for oxygen by binding to and stabilising deoxyhaemoglobin. High levels of 2,3-DPG therefore shift the curve to the right, while low levels of 2,3-DPG cause a leftward shift. The production of 2,3-DPG is likely to serve as an important adaptive mechanism by favouring the release of oxygen to the tissues. Consequently, its production increases in several conditions associated with diminished peripheral tissue O_2 availability, such as hypoxaemia, chronic lung disease, anaemia, congestive heart failure, etc. Hypoxia, e.g. due to lung disease or high altitude, also results in an adaptive rise in the concentrations of 2,3-DPG and an improvement in the quantity of oxygen delivered to the tissues.

Ventilation Perfusion Pressure

In normal lungs, alveolar ventilation (V) at rest is about 4 L/min; perfusion is about 5 L/min. Therefore, the normal ventilation/perfusion (V) ratio is 0.8. It exceeds 1.0 during maximal exercise because during maximal exercise, alveolar ventilation may rise to about 80 L/min, whereas alveolar perfusion rises to about 25 L/min. Also, ventilation and perfusion are not evenly distributed throughout the lungs because of the effects of gravity. Ventilation is higher at the base of the lungs than at the apex, because the tissues at the base of the lung are more compliant than those at the apex. Therefore, the lung bases receive nearly 2.5 times more air. Perfusion is also greater at the base of the lungs in comparison to the apices, with the bases receiving nearly six

times more blood than the apices. Therefore, the V/P ratio is higher at the apex than at the base of the lungs when a person is standing. Oxygen transfer across the alveoli can be explained by passive diffusion.

Pulse Oximetry

Pulse oximetry is a non-invasive method for assessing arterial oxygen saturation and heart rate.

Any cause of poor peripheral perfusion including external compression by a blood pressure cuff, causes unreliable readings. Though there may be mechanical causes for poor pulse oximetry readings, one must always look for a physiological cause first. For example, vasoconstriction is one of the first compensatory mechanisms employed by the body following blood loss. In these situations, it is often helpful to check pulse rate in comparison to the ECG.

Venous blood is 75% saturated and this normally corresponds to a PO_2 of 5.3 kPa (40 mmHg), whereas arterial blood is 97% saturated and has a PO_2 of 13.3 kPa (100 mmHg).

Effect on Respiration at High Altitude

At a high altitude, the following changes may take place in the respiratory physiology:
❑ At a high altitude where atmospheric pressure is halved, there is an increase in pulmonary ventilation due to stimulation of chemoreceptors by oxygen lack.
❑ Alveolar H_2O vapour pressure remains at the saturated pressure at body temperature.
❑ The fall in arterial PO_2 stimulates the carotid bodies to increase ventilation.
❑ Arterial pH increases because hyperventilation causes respiratory alkalosis.
❑ Cerebral blood flow decreases because the fall in PCO_2 causes cerebral vasoconstriction.

Disturbances in Respiration

Bronchial Asthma

Bronchial asthma is likely to be relieved by stimulation of beta adrenoceptors, which causes relaxation of bronchial smooth muscle. Drugs which stabilise mast cell membranes (e.g. chromoglycate drugs) also relieve asthma by reducing the release of bronchoconstrictor agents from mast cells. Glucocorticoids also prove useful by suppressing bronchoconstrictor and inflammatory mechanisms.

Chronic Respiratory Failure

A patient with chronic respiratory failure shows increased tolerance to high PCO_2 levels. Sensitivity to low PO_2 remains and is important for maintaining ventilation. The patients are likely to have high blood bicarbonate levels. HCO_3^-

levels rise to compensate for the raised PCO_2 in respiratory acidosis. Administration of 100% oxygen could arrest ventilation by removing hypoxic drive. Therefore, 24–28% O_2 must be administered in these cases.

Cyanosis

Cyanosis occurs when arterial blood contains more than 5 g/dL reduced haemoglobin; low haemoglobin values in anaemia make it difficult to reach this level.

Cough Reflex

Coughing is reflexly initiated by irritation of the trachea and bronchi. It is associated with contraction of the airways smooth muscle and increased velocity of flow.

It depends on expiratory muscles, particularly abdominal muscles for expulsion of air. Coughing differs from sneezing in that the glottis is initially closed. Thus, it is more explosive. Cough reflex is depressed during anaesthesia which may lead to retention of mucous secretions.

Physiology of Cardiovascular System

The overall function of the cardiovascular system is to provide the perfusion of the various body tissues, which helps in supplying nutrients and removal of waste products from the various body cells. The cardiovascular system operates as two circulations in series: systemic circulation and pulmonary circulation, united by the heart.

Blood flow Through the Heart

In an adult person, the right atrium receives blood from the systemic veins (the superior and inferior vena cavae) and transmits blood to the right ventricle via the tricuspid valve. The right ventricle then ejects blood into the pulmonary artery via pulmonary valves. After circulating through the lungs, oxygenated blood returns to the left atrium via pulmonary veins. From there, the blood circulates to the left ventricle via the mitral valve. Finally, left ventricle ejects oxygenated blood into the systemic circulation via the aortic valve. Circulation in an adult person is different from that in the foetus and has been described in details in Chapter 7.

Cardiac Cycle

Cardiac cycle is defined as the succession of coordinated events taking place in the heart during each beat. The series of events occurring during various phases of cardiac cycle are illustrated in **Figure 3.7**. Each heartbeat consists of two major phases called systole and diastole. During systole, heart contracts and pumps the blood through
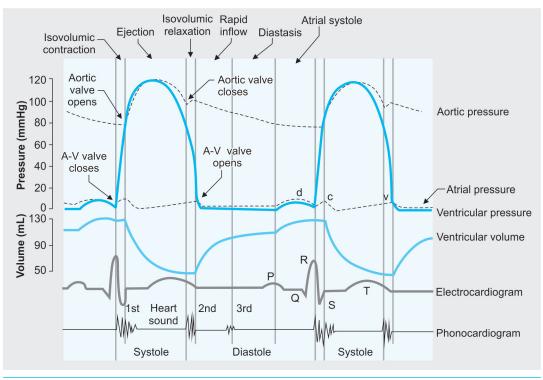
FIG. 3.7: Cardiac cycle

arteries. During diastole, heart relaxes and blood is filled in the heart. The atria contract together, with contraction of ventricles occurring 0.1–0.2 seconds later. All these changes are repeated during every heartbeat, in a cyclic manner.

At the beginning of cardiac contraction, the wave of depolarization is initiated in the atria from the SA (sinoatrial) node. This is followed by atrial contraction, which causes a rise in atrial pressure resulting in atrial systole. As blood is ejected from the atria into the ventricles, this results in an increase in the ventricular volume. Simultaneously, the flow of venous blood into the atria from the vena cava and pulmonary vein causes the atrial pressure to rise until it exceeds the ventricular pressure, causing the opening of atrioventricular valves and allowing the ventricles to fill.

The point at which the maximum ventricular volume has been reached is known as the end-diastolic volume. From the SA node, the wave of depolarization moves onto the atrioventricular node. Depolarization is briefly delayed at the atrioventricular node before spreading through the ventricular myocardium. As the wave of depolarization spreads over the ventricular myocardium, it results in ventricular systole. The onset of ventricular systole causes a rise in ventricular pressure. Therefore, ventricular pressure becomes higher than the atrial pressure. As a result, the atrioventricular valves are closed. The closure of atrioventricular valves (both the mitral valve on the left side and tricuspid valve on the right side) is heard as the first heart sound, "lub".

In relation to the ECG tracing, first heart sound corresponds with the QRS complex. It also corresponds with the C wave in the central venous pressure.

Closure of the atrioventricular valves is followed by the isovolumetric phase of contraction because during this phase of contraction, both atrioventricular valves, and aortic and pulmonary valves on left and right side respectively are closed and therefore the ventricular volume does not change. Soon with the further increase in the ventricular pressure, as the ventricular pressure exceeds the pressure in the aorta and the pulmonary artery, the aortic valves and the pulmonary artery valves open, resulting in the blood flow through the aorta and the pulmonary artery respectively. Concerning the left side of the heart, at the beginning of the ventricle systole, the mitral valve is open and the pressure in the left atrium is greater than that in the ventricle. As the pressure builds up in the left ventricle, the mitral valve closes.

At the end of the ventricular systole, the myocardium relaxes and begins to repolarize. With the reduction in ventricular pressure, the aortic and pulmonary valves close (heard as the second heart sound, "dub").

The second heart sound differs from the first heart sound in that it is about 20% shorter than the first sound. It is higher in frequency, i.e. about 50 Hz compared with 35 Hz for the first sound. Both the heart sounds can be occasionally split due to asynchronous valve closure. The closure of aortic and pulmonary valves is followed by the isovolumetric phase of relaxation because both the aortic valves and the atrioventricular valves are closed. At this point, the ventricular volume is at its lowest; this is the end systolic volume. With further ventricular relaxation, once the ventricular pressure falls below that of the atria, the atrioventricular valves open and the ventricular filling

begins once again. During the phase of atrial diastole, blood enters the left and right atrium respectively from the vena cava and pulmonary veins respectively.

In addition to the normal "lub-dub" heart sounds, a third heart sound is sometimes heard even in normal individuals. A pathological third heart sound may sometimes occur in the cases of mitral and tricuspid regurgitation, constrictive pericarditis, dilated left ventricle and acute myocardial infarction. This usually occurs due to rapid ventricular filling. A fourth heart sound, which is sometimes heard, is always pathological and occurs during atrial contractions, when a jet of blood hits an excessively stiff ventricle. Abnormally shift ventricle could be related to pathologies such as left ventricular hypertrophy, fibrotic left ventricle, hypertrophic cardiomyopathy, etc.

Stroke Volume

Stroke volume is calculated by subtracting end diastolic volume from the end systolic volume. The stroke volume is equal in both the ventricles and in an average-sized adult measures approximately 70 mL.

Electrocardiogram

Electrocardiography is the technique, which helps in evaluation of the electrical activities of heart. The spread of excitation through myocardium produces local electrical potentials. Electrocardiogram (ECG) is the record or graphical registration of electrical activities of the heart, which occur prior to the onset of mechanical activities.

Electrocardiographic grid refers to the markings (lines) on ECG paper. ECG paper has horizontal and vertical lines at regular intervals of 1 mm. Every 5th line (5 mm) is thickened. The standard paper speed for recording the ECG is 25 mm per second and 1 mm corresponds to 0.04 seconds. The standard calibration of voltage deflections is 0.1 mV which equals 1 mm. Different waves visualised on the ECG trace are summarised in **Table 3.9** and **Figure 3.8**.

Significance of ECG Changes

PR Interval

PR interval is between 0.1 and 0.2 seconds. PR interval of greater than 0.2 seconds implies delay in conduction from the atria to the ventricles. On the other hand, PR interval of less than 0.1 seconds implies abnormally rapid conduction from the atria to the ventricles. This could be related to the aberrant conduction systems as occurring in Wolff-Parkinson-White syndrome.

QT Interval

QT interval is between 0.3 and 0.4 seconds and is more dependent on the heart rate than is PR interval. It is increased in hypocalcaemia, hypokalaemia, rheumatic carditis and medication with quinidine. It is reduced by hypercalcaemia, hyperkalaemia and in cases of digoxin toxicity.

ECG Changes in Myocardial Infarction

The following changes may occur on ECG in case of myocardial infarction:

- ❑ The usual ECG changes following an acute myocardial infarction include ST segment elevation greater than 1 mm (convex upwards, concave downwards) developing within the first few hours.
- ❑ Tall peaked T waves may also be seen in the acute stages.
- ❑ Pathological Q waves (representing transmural infarction), a reduction in R wave height and deeply inverted T waves (in the leads facing the infarcted muscle) may develop over the next 72 hours. The absence of Q waves implies partial-thickness infarction.
- ❑ A sub-endocardial MI is associated with segment ST depression (not elevation) and T wave inversion in leads facing the infarction.
- ❑ The ST segment changes following a myocardial infarct usually resolve within days. The T wave changes often persist for weeks but may be permanent. The Q waves are usually, but not always permanent.
- ❑ It is possible to diagnose an acute MI in the presence of right bundle branch block and also in left bundle branch block when using non-standard ECG criteria.
- ❑ True posterior left ventricular infarction is characterised by tall R waves, ST depression and peaked upright (not inverted) T waves in leads V_1 and V_2.
- ❑ Right ventricular infarction does not produce a specific pattern in the standard 12 lead ECG, so the use of right-sided precordial leads (V_4R-V_6R) is required.

Atrial Fibrillation

The following changes are observed on ECG trace in case of atrial fibrillation:

- ❑ Atrial rate is higher than ventricular rate as some impulses are filtered out by the atrioventricular node. Respiratory sinus arrhythmia is not seen because sinus arrhythmia indicates normal sinus rhythm.
- ❑ P waves are absent. Small rapid waves indicate atrial fibrillation.
- ❑ The ventricular rate is irregular due to the irregularity of the impulses passing through the AV node. The QRS complexes are normal since the pattern of ventricular depolarization is normal.

Conduction System of the Heart

Specialised conducting tissue is present within the heart to ensure the orderly and synchronous contraction of the atria followed by that of the ventricles. The origin of the cardiac impulse starts at the sinoatrial node, which is located in the right atrium at the entry of the superior vena cava. From there, the impulse spreads through the smooth muscles to

TABLE 3.9	Different waves on ECG trace		
ECG wave	Phase of cardiac cycle	Time duration	Significance
P wave	Atrial depolarization		
PR interval	Depolarization across atrioventricular node (a rough approximation of atrioventricular conduction time). Atrial repolarisation occurs during this time.	Normal PR interval is 0.12–0.2 seconds	The PR interval is measured from the start of the P wave to the start of the QRS complex and serves as a rough approximation of atrioventricular conduction time.
QRS complex	Depolarization of ventricular myocardium	Normally less than 0.12 seconds	
T wave	Repolarization of ventricular myocardium		Changes in the shape of T wave may occur in different phases of myocardial infraction.
QT interval	It approximates to the ventricular refractory period	Normally 0.42 seconds	The QT interval is measured from the start of the QRS complex to the end of the T wave.
ST segment	Represents the time period between ventricular depolarisation and ventricular repolarisation		The ST segment is measured from the end of the QRS complex to the start of the T wave.
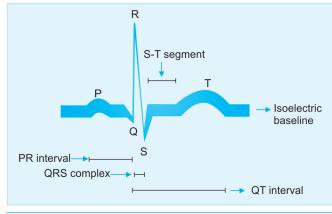
FIG. 3.8: Different waves in a normal ECG trace

the atrioventricular (AV) node situated in the right atrium above the artioventricular ring near the interatrial septum. There is no specialised conducting tissue between the SA node and the AV node. From the AV node, the cardiac impulse is then conveyed via the specialised tissues called Bundle of His, which help in conducting the impulses to the atrioventricular ring, to the septum where the bundle divides into left and right branches, innervating the left and right ventricle respectively. The right bundle is relatively narrow compared to the left bundle. The more distal fibres of the conducting system are known as the Purkinje system.

Purkinje fibres travel to the apex before proceeding to the base of the heart. They spread depolarization rapidly over the entire ventricular myocardium. They are responsible for the short duration of the QRS complex as well as its configuration. Damage to the Purkinje cells (as in bundle-branch block) changes the pattern of spread of ventricular depolarization, and hence the shape of the QRS complex. Purkinje tissue cells in the heart conduct impulses at the rate of around 4 metres/second, which is faster than that in some of the neurons. Small diameter nerve fibres conduct impulses at about 1 metre per second. Purkinje cells are larger than ventricular myocardial cells, which facilitates rapid conduction.

Mean Volume of Blood Flow

Mean volume of blood flow is the volume of blood, which flows into the region of circulatory system in a given unit of time. It is estimated with help of the following formula:

$$Q = V \times A$$

Where,

Q = Quantity of blood; V = Velocity of blood flow; A = Cross-sectional area of the blood vessel

Volume of blood flow is determined by the following five factors:

1. *Pressure gradient*: Volume of blood flowing through any blood vessel is directly proportional to the pressure gradient.
2. *Resistance to blood flow*: Volume of blood flow is inversely proportional to the resistance (or tension in the blood vessel against which the blood has to flow). Peripheral resistance implies the resistance offered to blood flow in the peripheral blood vessels. Peripheral resistance is inversely related to radius of the blood vessel, i.e. lesser the radius, more will be the resistance. Since the arterioles remain partially constricted all the time due to sympathetic tone, the resistance in arterioles is very high. Therefore, the arterioles are known as the resistant vessels. Arterioles play a major role in regulating arterial blood pressure and in regulating the local blood flow. They offer more resistance to flow than capillaries. Local flow varies directly with the fourth power of their radii. They have a smaller total cross-sectional area than do the capillaries, so blood flow velocity is higher in arterioles.
3. *Viscosity of blood*: Volume of blood flow is inversely proportional to the viscosity of blood. RBC count is the main factor, which determines the viscosity of the blood. Another factor determining viscosity is plasma protein, mainly albumin.
4. *Diameter of blood vessels*: Volume of blood flow is directly proportional to the diameter of the blood vessels.

5. *Velocity of blood flow*: Volume of blood flow is directly proportional to the velocity of blood flow.

Blood flow through a blood vessel can be of two types:
1. Streamline or laminar flow
2. Turbulent flow

Streamline flow: Streamline flow is also known as the silent flow because it does not produce any sound within the vessel. The velocity of blood flow is greater towards the centre of large blood vessels than at the periphery. In the circulation, it rises due to the compensatory increase in cardiac output. In capillaries, the velocity of blood flow is low because the capillary bed has a large total cross-sectional area. In veins, it is greater than in venules because the venous bed has a smaller total cross-sectional area than the venular bed. It can fall to zero in the ascending aorta during diastole.

Turbulent flow: Turbulent flow is chaotic. While all fluid moves in a single direction, flow is irregular and there are local currents or eddies. Turbulent flow is the noisy flow because the flowing blood in these cases produces a sound. The tendency for blood flow to be turbulent is directly proportional to vessel diameter, fluid density and velocity. The tendency is inversely proportional to viscosity. In anaemia, the increase in velocity and decrease in viscosity of blood in the hyperdynamic circulation promotes turbulence; bruits may be therefore heard over peripheral arteries.

Arterial Blood Pressure

Arterial blood pressure is defined as the lateral pressure exerted by the column of blood on wall of the arteries. Arterial blood pressure is directly proportional to the factors such as cardiac output, heart rate, peripheral resistance, blood volume, venous return, velocity of blood flow and viscosity of blood. Arterial blood pressure is indirectly proportional to factors such as elasticity and the diameter of blood vessel.

Hypertensive patients usually have no specific symptoms or signs. Abnormal signs may appear after a period of prolonged, severe hypertension. A fourth heart sound may be heard in the diastole preceding the first heart sound and is related to an increased atrial activity. Hypertension is often associated with left ventricular hypertrophy, resulting in a thrusting apex beat. A loud aortic second heart sound is also a classical finding in cases of hypertension.

Auscultatory Method

Auscultatory method is the most accurate way of measuring the arterial blood pressure. After determining the systolic pressure using the palpatory method, the pressure in the cuff is raised by about 20 mmHg above that level, so that the brachial artery is occluded due to compression. The chest piece of the stethoscope is then placed over the antecubital fossa and the arm cuff is slowly deflated. While doing so, series of sounds are heard through the stethoscope. These sounds are known as Korotkoff sounds. Korotkoff sounds are produced locally by the turbulence of blood being forced past the narrow segment of a partially occluded artery.

Korotkoff sounds have five phases:
1. *First phase*: This is associated with the appearance of tapping sounds. The sharp taps of phase 1 are generated as the peaks of systolic pressure force blood under the cuff.
2. *Second phase*: Following the clear tapping sound, a murmuring sound is heard.
3. *Third phase:* After the murmuring sound, a very clear and loud sound of the gong type is heard.
4. *Fourth phase*: Sudden muffling of heart sounds.
5. *Fifth phase*: Disappearance of the heart sounds. Disappearance of this sound is an indicator of the diastolic blood pressure.

Severe systemic hypertension may result in the following:
- An increase in the size of myocardial cells (hypertrophy) in the left ventricle
- Increased QRS voltage in certain leads due to left ventricular hypertrophy
- Increased coronary blood flow due to increased left ventricular work
- Pulmonary oedema due to left ventricular failure
- Impaired vision due to damage to retinal blood vessels

The Dinamap is a useful device for the continual estimation of patient's blood pressure but is less accurate than the gold standard device, mercury sphygmomanometer. The Dinamap is also unreliable in cases of arrhythmias/atrial fibrillation. It provides a more accurate reading of systolic than diastolic blood pressure. It relies upon oscillometric techniques to assess the BP. The advantage of the device is that it does not have to be positioned at the level of the heart as the cuff itself does the measurement; it also does not rely upon any specific expertise for use and is fully automated.

Blood Pressure Regulation

Circulating hormones such as adrenaline and angiotensin II [which is converted from angiotensin 1 and that from angiotensinogen via the renin-angiotensin conversion enzyme (ACE) conversion pathway] are potent vasoconstrictors, but they probably have little effect on controlling the local blood flow. In contrast, endothelium derived factors play an important role in controlling local blood flow. These substances are either produced or modified in the vascular endothelium, and include prostacyclin and nitric oxide, both of which are potent vasodilators. Bradykinin is known to be vasodilatory and hence produces a hypotensive effect. Serotonin also causes vasodilatation. As a result, selective serotonin reuptake inhibitor (SSRI) can cause hypotension.

Physiology of Urinary System

Microanatomy of Kidney

Nephron is defined as the structural and functional unit of kidney. Each kidney consists of 1 to 1.3 million nephrons and is on average 45–65 mm in length. Each nephron is formed by two parts **(Fig. 3.9)**: a blind end called renal corpuscle or Malpighian corpuscle and a tubular portion called renal tubule.

Renal corpuscle is formed by two portions: glomerulus and bowman capsule. Function of the renal corpuscle is the filtration of blood, which forms the first phase of urine formation. Blood is filtered at the glomerulus and the filtrate is subsequently modified by reabsorption or secretion in its passage through the nephron. Urine is formed as a result of the modifications occurring in the glomerular filtrate as it passes through the entire nephron, and finally leaves at the collecting duct.

Glomerulus is a tuft of capillaries enclosed by Bowman capsule. It consists of glomerular capillaries interposed between afferent arteriole on one end and efferent arteriole on the other end. Bowman capsule is a capsular structure, which encloses the glomerulus and is formed by two layers: inner visceral layer (covering the glomerular capillaries) and an outer parietal layer **(Fig. 3.10)**. Both the layers of Bowman capsule are composed of a single layer of flattened epithelial cells resting on a basement membrane. The glomerular filtrate has to cross two layers of cells, the capillary endothelium and the tubular epithelium separated by an amorphous basal lamina to pass from the blood vessels to the tubules. In case of renal diseases such as glomerulonephritis, this barrier is damaged.

Tubular portion of nephron is the continuation of Bowman capsule. It is made up of three parts: proximal convoluted tubule, loop of Henle and the distal convoluted tubule.

❑ *Proximal convoluted tubule*: This is the coiled portion of the tubule, which arises from the Bowman capsule and continues downwards as the descending limb of loop of Henle.

❑ *Loop of Henle*: This consists of the following structures: thin descending limb, hairpin bend and a thick ascending limb.

❑ *Distal convoluted tubule*: This is the continuation of thick ascending segment of the loop of Henle and it continues as the collecting duct. Distal convoluted tubule continues as the initial or arched collecting duct, which lies in the cortex. Seven to ten initial collecting ducts unite to form the straight collecting duct, which passes through medulla and eventually into the renal pelvis.

The loops of Henle differ between the tubules located in the cortex (cortical tubules form 85% of the total) and those located near the medulla (juxtamedullary tubules, 15% of the total). The juxtamedullary tubules have much longer loops of Henle.

Process of Urine Formation

The urine formation includes three processes: glomerular filtration, tubular reabsorption and tubular secretion.

Glomerular Filtration

When blood passes through glomerular capillaries, the plasma is filtered into the Bowman capsule. This process is called glomerular filtration. All the substances of plasma are filtered except the plasma proteins. The filtered fluid is called glomerular filtrate.

Filtrate from Bowman capsule passes through the tubular portion of the nephron. While passing through the tubule, the filtrate undergoes various changes both in quality and in quantity. Many wanted substances like glucose,
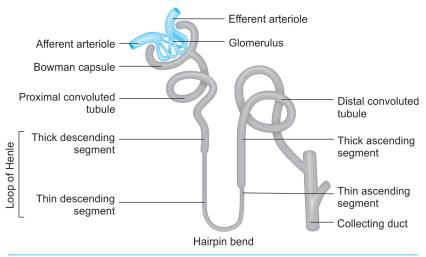

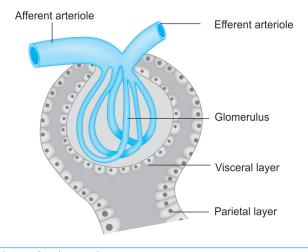
FIG. 3.9: Structure of a nephron

FIG. 3.10: Renal corpuscle

amino acids, water and electrolytes are reabsorbed from the tubules through the process of tubular reabsorption. Some unwanted substances are secreted into the tubule from peri-tubular blood vessels through a process is called tubular secretion or excretion.

In the urine of normal healthy people, specific gravity may range from 1.004 to 1.040. Osmolality may range from 100 to 1,000 mOsmol/litre. Colour is due to "urochrome", a pigment of uncertain origin.

Tubular Reabsorption

Tubular reabsorption is the process by which water and other substances are transported from renal tubules back to the blood. More than 99% of the absorbed water, electrolytes and other substances are reabsorbed by the tubular epithelial cells. The brush border of epithelial cells in proximal convoluted tubule increases the surface area and facilitates reabsorption. The reabsorbed substances move into the interstitial fluid of renal medulla. And, from there, the substances move into the blood in peritubular capillaries. Various hormones regulating the absorption of different substances are summarised in **Table 3.10**. Substances absorbed in the various region of the renal tubule are as follows:

Substances reabsorbed from proximal convoluted tubule: About 88% of the filtrate is reabsorbed in proximal convoluted tubule. Other substances reabsorbed from proximal convoluted tubule are glucose, amino acids, sodium, potassium, calcium, bicarbonates, chlorides, phosphates, urea, uric acid and water. Urea is the only molecule to be reabsorbed as well as secreted. Approximately 50% of it enters the glomerular filtrate and is excreted. Due to the absorption of both water and solutes, osmolality changes little in the proximal convoluted tubule.

Only a small amount of albumin is filtered normally at the glomerulus and this is reabsorbed by the tubules. It is a negatively charged molecule at the normal pH of blood. It is a first-class protein containing both essential and non-essential amino acids. Human plasma albumin contributes more to plasma colloid osmotic pressure than globulin. Its greater mass and lower molecular weight provides more osmotically active particles.

Substances reabsorbed from loop of Henle: Substances reabsorbed from loop of Henle are sodium and chloride. Water leaves the descending thin limb of the loop of Henle by diffusion. This is because of the high solute potential around the descending limb caused by active transport of sodium and chloride from the ascending limb.

Substances reabsorbed from distal convoluted tubule: Sodium, calcium, bicarbonate and water are reabsorbed from distal convoluted tubule. Acidification occurs mainly in the distal convoluted tubule.

About 99% of the glomerular filtrate (nearly 8 litres/hour) is reabsorbed by the renal tubules. Most or all of filtered amino acids, glucose and plasma proteins are absorbed. All the bicarbonate ions are also reabsorbed. These absorbed bicarbonate ions along with those HCO_3^- ions manufactured in the kidney compensate for the respiratory acidosis. Since the amount of chloride ions filtered at the glomerulus is nearly 20 times more than the potassium ions, absorption of chloride is more than that of potassium.

Absorption of glucose: Most or all of the glucose molecules are completely reabsorbed before the end of the proximal tubule. In the normally functioning kidneys, glucose is filtered and reabsorbed via secondary active transport. Normally, none of the glucose is secreted by the renal tubules. Most of the glucose molecules are reabsorbed by the apical region of the proximal convoluted tubules of the kidneys via Na/glucose transporters and also by glucose transporters (GLUTs). Therefore, concentration of glucose is less than that of plasma.

Under normal circumstances, all the filtered glucose is completely absorbed by the PCT and none of it appears in the urine. The renal threshold for glucose, which is the amount of plasma glucose that the kidneys are able to filter

TABLE 3.10	Hormones regulating tubular reabsorption
Hormone	**Action**
Aldosterone	Increases sodium reabsorption in ascending limb, distal convoluted tubule and collecting duct
Angiotensin II	Increases sodium reabsorption in proximal convoluted tubule, thick ascending limb, distal tubule and collecting duct
Antidiuretic hormone	Increases water reabsorption in distal convoluted tubule and collecting duct
Atrial natriuretic factor	Decreases sodium reabsorption
Brain natriuretic factor	Decreases sodium reabsorption
Parathormone	Increases reabsorption of calcium, magnesium and hydrogen Decreases phosphate reabsorption
Calcitonin	Decreases calcium reabsorption

without it being excreted in the urine, is about 200 mg/dL. This is known as the glucose tubular transport maximum (T_m). Once the filtered glucose levels rise above the T_m, glucose begins to appear in the urine **(Fig. 3.11)**.

Since filtration is directly proportional to the concentration, as the plasma glucose concentration rises above normal, rate of glucose filtration also increases linearly. Glucose should be completely reabsorbed from the urine in the kidneys, and if not, this generally demonstrates as diabetes mellitus. Transport maxima for glucose is constant and usually does not increase or decrease. On the other hand, clearance increases linearly. It remains at zero until the T_m is reached and then it rises linearly. Reabsorption increases and then levels off after T_m of glucose is reached. Excretion is therefore initially zero and then rises linearly. Glycosuria is usually suggestive of diabetes mellitus and requires further investigations.

Tubular Secretion
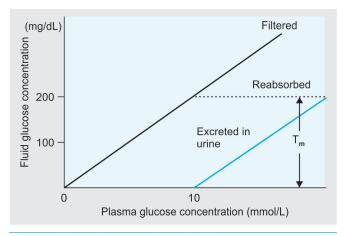
Substances secreted in different segments of renal tubules are as follows:
❑ Potassium is secreted actively by sodium-potassium pump in proximal and distal convoluted tubules and collecting ducts.
❑ Ammonia is secreted in the proximal convoluted tubule.
❑ Hydrogen ions are secreted in the proximal and distal convoluted tubules. Maximum hydrogen ion secretion occurs in proximal tubule.
❑ Urea is secreted in loop of Henle.

Renal Function Tests

Plasma Clearance

Plasma clearance is defined as the amount of plasma that is cleared off a substance in a given unit of time.

FIG. 3.11: Glucose absorption in the renal tubule
Abbreviation: T_m, transport maximum

Determination of clearance value for certain substances helps in assessing the following renal functions: glomerular filtration rate, renal plasma flow (RPF) and renal blood flow.

The following factors are required to determine the plasma clearance of a particular substance: volume of urine excreted; concentration of the substance in urine; and concentration of the substance in blood. Clearance is calculated using the following formula:

$$C = \frac{UV}{P}$$

Where, C = clearance; U = concentration of the substance in urine; V = volume of urine flow; P = concentration of the substance in plasma.

Substances such as creatinine or urea, which are excreted by the kidney and have a lower concentration in the renal vein than the artery are said to be cleared by the kidneys. Renal clearance of a substance is directly related to its urinary concentration (U) and to the rate of urine formation (V). It is inversely proportional to the concentration of the substance in the plasma. Clearance thus tends to fall at low urinary flow rates. It rises if the substance is normally reabsorbed by an active process.

Glomerular Filtration Rate

Glomerular filtration rate (GFR) is equivalent to the clearance of a substance which is completely filtered, but is neither reabsorbed from the renal tubule nor is secreted. The plasma constituent which most closely approaches this is creatinine. The glomerulus freely filters creatinine and 10–20% of creatinine is actively secreted by the proximal tubules. Creatinine clearance is therefore the measurement for the estimation of GFR. Normal GFR of both the kidneys is 120 mL/minute. Plasma concentration of creatinine increases after protein ingestion. Protein ingestion, therefore, increases the GFR.

Inulin clearance is also sometimes used for an accurate determination of GFR. However, the main disadvantage is that inulin has to be infused in order to maintain a steady plasma level. Clearance of radioactive B_{12} is also sometimes used. However, it cannot be used during pregnancy.

Measurement of GFR using creatinine clearance is easier than inulin clearance, because creatinine is already present in body fluids and its plasma concentration is steady throughout the day.

Renal Plasma Flow

Renal plasma flow of a substance is equivalent to the clearance of a substance, which is filtered and secreted but not reabsorbed. Such a substance is para-aminohippuric acid (PAH).

Physiology of Gastrointestinal System

Digestive System

Primary digestive organs are the organs where actual digestion takes place. These include the mouth, pharynx, oesophagus, stomach, small and large intestines. Pancreas is an accessory digestive organ, which aids primary digestive organs in the process of digestion.

Mouth/Saliva

Digestive juice present in the mouth is saliva, which is secreted by the salivary glands. In humans, the saliva is secreted by three pairs of major (large) salivary glands and some minor salivary glands. The major salivary glands include the parotid gland, submandibular gland and the sublingual glands. Saliva produced from different salivary glands have a different composition. Serous glands such as the parotids produce a watery juice; mucous glands such as the sublinguals produce a thick viscid juice.

Saliva is not necessary for digestion of food because other digestive tract enzymes can take over if salivary enzymes are absent. Saliva is also not required for swallowing of food. However, swallowing solids is difficult without saliva's moisturizing and lubricant effects. Saliva is required for normal speech. It is also required for antisepsis in the mouth and taste sensation. In the absence of saliva, the mouth becomes infected and ulceration occurs. For the proper sensation of taste, substances must go into solution before they can stimulate taste receptors.

Saliva contains an enzyme, salivary amylase which catalyses the hydrolysis of starch into sugars. The functions of salivary amylase (ptyalin) can be affected by enzymes from other digestive glands. Saliva is saturated with calcium ions; calcium salts are laid down as plaque on the teeth. Saliva is an important route of iodide excretion; its concentration in saliva is 20–100 times that in plasma. Saliva has a neutral pH; acidity in the mouth tends to dissolve the tooth enamel. Salivary enzymes are not effective at the low pH of gastric juice.

Stomach

Stomach is a hollow organ situated just below the diaphragm on the left side in the abdominal cavity and is composed of the four following parts:
1. Cardiac region
2. Fundus
3. Body or corpus
4. Pyloric region
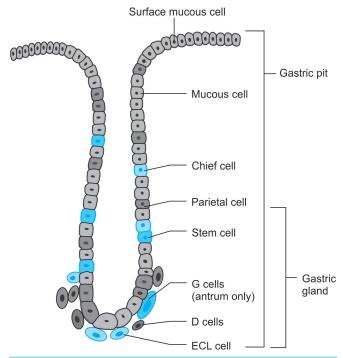
Several tubular structures called gastric glands open into the cavity of stomach. The secretory products of the cells in the gastric glands **(Fig. 3.12)** are elaborated in **Table 3.11**. Gastric juice is a mixture of secretions from different gastric glands **(Table 3.12)**. It is composed of enzymes such as pepsin, rennin, gastric lipase, gelatinase, urease, etc. and inorganic substances such as hydrochloric acid, sodium, calcium, potassium, bicarbonate, chloride, phosphate, etc. Organic substances such as mucus and intrinsic factor may also be present.

Pepsin is secreted in form of inactive pepsinogen by the chief cells of the stomach. Pepsinogen is converted into pepsin by hydrochloric acid. Pepsin is the active proteolytic form of pepsinogen present in the stomach. Optimum pH

TABLE 3.11	**Various secretory substances produced by the gastric glands**
Cell	**Secretory products**
Chief cells	Pepsinogen Rennin Lipase Gelatinase Urease
Parietal cells	Hydrochloric acid Intrinsic factor of Castle
Mucus neck cells	Mucin
G-cells	Gastrin
Enterochromaffin cells	Serotonin
Enterochromaffin-like cells	Histamine
D-cells (delta cells)	Somatostatin

FIG. 3.12: Cells of the gastric glands
Abbreviation: ECL, enterochromaffin-like cells

TABLE 3.12	Various enzymes in the gastric juice		
Enzyme	**Activator**	**Substrate which is digested**	**End products**
Pepsin	Hydrochloric acid	Proteins	Proteases, peptones and polypeptides
Gastric lipase	Acid medium	Triglycerides of butter	Fatty acids and glycerols
Gastric amylase	Acid medium	Starch	Dextrin and maltose (negligible action)
Gelatinase	Acid medium	Gelatin and collagen of meat	Peptides
Urease	Acid medium	Urea	Ammonia

for activation of pepsinogen is below 6. Pepsin converts proteins into proteases, peptones and polypeptides. Pepsin also causes curdling and digestion of milk (casein). Curdling of the ingested milk is also performed by rennin, a protease produced by the newborn ruminant animals.

Due to the production of hydrochloric acid (HCl) by the gastric parietal cells, the stomach pH varies between 2 and 3. Secretion of hydrochloric acid is an active process **(Fig. 3.13)**, which takes place in the canaliculi of parietal cells in gastric glands. Pentagastrin is a powerful pharmacological stimulant of mucosal cells for producing HCl.

Vagal stimulation also increases the secretion of acid and pepsinogen; this action is mediated by acetylcholine and gastrin-releasing peptide released from the vagal nerve endings. After vagotomy, food must enter the stomach to stimulate gastric secretions. Gastric juice does not digest the gastric mucosa because a mucus coat impregnated with bicarbonate protects it. A similar coat of mucus impregnated with bicarbonate also protects mucosal cells in the oesophagus. HCl in the gastric juice also reduces trivalent ferric iron to the divalent ferrous form in which it can be absorbed in the small intestine. Low gastic acidity imparted by HCl prevents bacterial growth. Therefore, the administration of histamine (H2) receptor antagonists, which blocks gastric acid secretion, is associated with a rise in the bacterial count. Factors influencing the secretion of hydrochloric acid are tabulated in **Table 3.13**. Functions of HCl can be summarised as follows:

❑ Activation of pepsinogen into pepsin
❑ Bacteriolytic action: Killing some of the bacteria entering the stomach along with the food substances
❑ Provision of acidic medium, necessary for the action of various enzymes.

Gastrin is a gastrointestinal hormone secreted by the G cells that are present in the pyloric glands of stomach. Small amount of gastrin is also secreted in mucosa of upper small intestine and duodenum. Gastrin is also secreted by islets of Langerhans in pancreas in foetuses and in adults with pancreatic gastrin-secreting tumours (gastrinomas). Gastrin has a variety of actions, but its principal physiological actions are as follows:

TABLE 3.13	Factors influencing the secretion of hydrochloric acid
Factors stimulating the secretion of hydrochloric acid	

- Gastrin
- Histamine
- Vagal stimulation

Factors inhibiting the secretion of hydrochloric acid

- Secretin
- Gastric inhibitory polypeptide
- Peptide YY
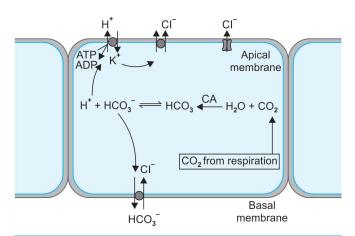
FIG. 3.13: Secretion of hydrochloric acid into the gastric lumen
Abbreviations: CA, carbonic anhydrase; ADP, adenosine diphosphate; ATP, adenosine triphosphate

❑ Stimulation of gastric acid and pepsin secretion
❑ Stimulation of the growth of the mucosa of the stomach and intestine.

Gastrin secreted in response to a meal increases tone of the cardiac (oesophageal-gastric) sphincter and so prevents regurgitation of gastric contents into the oesophagus during stomach contractions. Gastrin levels are increased in conditions associated with low acid production, for example, atrophic gastritis and therapy with proton pump inhibitor (PPI) due to loss of negative feedback. Gastrin levels only reach high enough levels following a protein meal to stimulate insulin secretion.

Pancreas

Pancreas is a dual organ having two functions, namely endocrine function and exocrine function. Endocrine function is concerned with the production of hormones. The exocrine function of the pancreas is concerned with the secretion of digestive juice called pancreatic juice. Pancreatic juice has a high bicarbonate content, about 110–150 mEq/L, against the plasma level of 24 mEq/L. High bicarbonate content in the pancreatic juice makes it highly alkaline, so that it protects the intestinal mucosa from acid chyme by neutralising it. Moreover, the bicarbonate ions provide the required pH (7–9) for the activation of pancreatic enzymes

present in the pancreatic juice. Pancreatic juice plays an important role in the digestion of proteins and lipids. It also has mild digestive action on carbohydrates. The proteolytic enzymes present in the pancreatic juice include trypsin, chymotrypsin, carboxypeptidase, elastase, nuclease, collagenase, etc. Various lipolytic enzymes present in the pancreatic juice include pancreatic lipase, cholesterol ester hydrolase, phospholipase A, phospholipase B, colipase and bile salt-activated lipase. Digestion and absorption of carbohydrates and proteins is described in **Figures 3.14** and **3.15** respectively.

In response to vagal stimulation, pancreatic secretion is scanty and rich in enzymes. In response to acid in the duodenum, the secretion is copious, bicarbonate rich and poor in enzymes; it buffers the acid secretions.

Hormones stimulating pancreatic secretion include secretin and cholecystokinin. Secretin stimulates the secretion of watery juice, which is rich in bicarbonate ions and high in volume, by acting on pancreatic ductules via cyclic AMP. Cholecystokinin, also known as cholecystokinin-pancreozymin stimulates the secretion of pancreatic juice that is rich in enzyme and low in volume, by acting on pancreatic acinar cells via inosine triphosphate. Cholecystokinin stimulates the gall bladder to contract. Besides its main action of stimulating the gall bladder to contract, cholecystokinin also decreases the rate of gastric emptying.

Small Intestine

Small intestine is composed of three parts: duodenum, jejunum and ileum. Important function of small intestine is absorption, though it may also play a small role in digestion. Maximum absorption of digested food products takes place in the small intestine. Vitamin B_{12} is absorbed mainly in the terminal ileum. Water is absorbed passively down the osmotic gradient set up by active absorption of sodium and glucose. Glucose absorption is dependent on sodium absorption. Sodium is required at the luminal surface for glucose to be absorbed by an active carrier-mediated process. Absorption of calcium occurs mainly in the duodenum.

Physiology of Liver

Liver is an important organ having dual functions, both secretory and excretory. Some of the functions performed by the liver are as follows:

❑ *Maintenance of the blood glucose levels*: When blood glucose levels fall, liver glycogen is broken down to form glucose; when blood glucose level increases, it is converted in the liver into glycogen.

❑ *Deamination of amino acids*: NH_4 ions are toxic and are converted into urea and excreted in the urine.

❑ *Synthesis of 25-hydroxycholecalciferol*: Cholecalciferol is produced in skin by the action of sunlight; the liver converts it to 25-hydroxycholecalciferol and the kidney completes its activation by further hydroxylation and forms 1,25-dihydroxycholecalciferol.

❑ *Manufacture of plasma proteins*: Liver manufactures most of the plasma proteins but lymphocytes manufacture immune globulins.
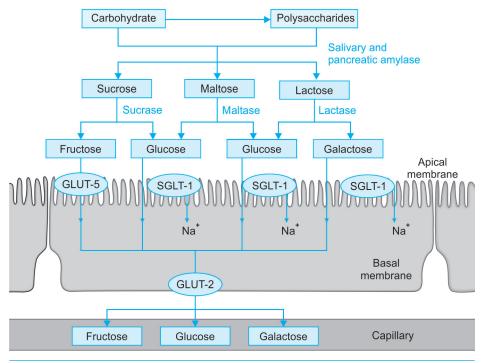

FIG. 3.14: Digestion and absorption of carbohydrates
Abbreviations: GLUT5, glucose transporter; GLUT 2, glucose transporter; SGLT, sodium-glucose linked transporter
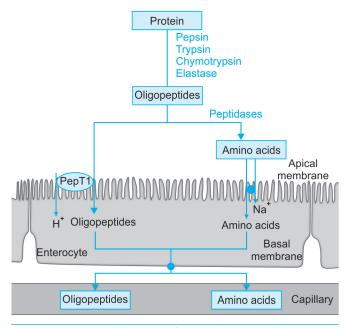
FIG. 3.15: Digestion and absorption of proteins
Abbreviation: PepT1, peptide transporter

❑ *Inactivation of steroid hormones*: The failure to inactivate oestrogens in men with liver failure can lead to breast enlargement.

Liver Function Tests

Tests of liver function include estimation of prothrombin time, serum albumin levels, bilirubin (direct and indirect) and estimation of the levels of hepatic enzymes: transaminases (AST, aspartate transaminase or SGOT and ALT, alanine transaminase or SGPT), alkaline phosphatase and γ-glutamyl transpeptidase. AST is not a specific hepatic enzyme like ALT, so it may also be raised in a range of pathologies, for example, MI, pulmonary embolism, hepatitis, etc. γ-glutamyl transpeptidase is a microsomal enzyme, usually raised in conditions such as alcohol excess, cirrhosis and hepatic metastatic disease. It may also be raised in cases of phenytoin therapy.

Bile

Bile is secreted by hepatocytes. The initial bile secreted by hepatocytes contains large quantity of bile salts, bile pigments, cholesterol, lecithin and fatty acids. Bile contains no digestive enzymes. From hepatocytes, bile is released into canaliculi, from where it passes through small ducts and hepatic ducts and reaches the common hepatic duct. From common hepatic duct, bile is diverted either directly into the intestine or into the gall bladder. Sodium, bicarbonate and water are added to bile when it passes through the ducts. Bile becomes more acidic during its storage in the gall bladder, which improves the solubility of bile solids. Production of bile is stimulated by increased vagal tone.

Bile is alkaline in nature, with a pH of around 8. Ninety five percent of bile is reabsorbed in the terminal ileum and undergoes the so-called enterohepatic circulation. Bile is essential for the absorption of fats and fat-soluble vitamins. Bile salts present in the bile assist in the emulsification and absorption of fat. They make cholesterol more water-soluble by forming cholesterol micelles.

Haemoglobin is broken down to bilirubin and excreted through bile. Iron is removed from haem during the formation of bilirubin. The bilirubin is conjugated by the hepatocytes before excretion.

Bile Salts

Bile salts are "choleretics", substances that stimulate bile secretion. They are synthesized from cholesterol in the liver. They are the only constituents of bile necessary for digestion. They have a characteristic molecular structure, which is partly water-soluble and partly fat-soluble. This property allows them to form micelles for fat transport, which are absorbed in the terminal ileum **(Fig. 3.16)**. Bile salts emulsify fat creating a greater surface area for lipase to act on. Absorption of dietary fat can only occur if emulsified into sufficiently small particles. Absorption of dietary fat involves uptake of fat by both the lymphatic and blood capillaries. The smaller fatty acids pass directly into blood; the larger ones are esterified, packaged into chylomicrons and taken into lymphatics.

Bilirubin

Bilirubin is a porphyrin pigment derived from haem caused by the body's clearance of aged red blood cells containing haemoglobin. Bilirubin is derived from biliverdin formed from haem. The reaction takes place with help of the enzyme haem oxygenase. The "blood-brain barrier" normally prevents bilirubin from entering brain tissue in an adult. However, in a newborn child, the blood-brain barrier may not be mature enough, thereby allowing excessive bilirubin to pass through resulting in the development of kernicterus. Bilirubin is sensitive to light; light converts bilirubin to lumirubin that is excreted more rapidly. Therefore, phototherapy may be used in the treatment of haemolytic jaundice in children.

Urobilinogen is a mixture of colourless compounds produced from the reduction of bilirubin. It is mainly excreted in the bile. Nearly half of urobilinogen is absorbed from the intestine and the remaining half is carried back to the liver in the enterohepatic circulation. Therefore, approximately 50% of the urobilinogen is reabsorbed and taken up via the portal veins to the liver. From there, it enters the circulation and is excreted by the kidneys. Rest of the urobilinogen reaches the intestines where it is directly reduced to a brown-coloured compound called stercobilin that is responsible for giving a characteristic colour to the faeces. Urobilinogen, which is excreted via the kidneys, is converted into the dark pigment, urobilin, on

TABLE 3.14	Characteristics of different types of jaundice		
Features	**Pre-hepatic (haemolytic) jaundice**	**Hepatic (hepato-cellular) jaundice**	**Post-hepatic (obstructive) jaundice**
Cause	Excess breakdown of RBCs	Liver damage	Obstruction of bile ducts
Urinary excretion of urobilinogen	Increases	Decreases	Decreases or is absent in cases of severe obstruction
Faecal excretion of stercobilinogen	Increases	Decreases (pale faeces)	Absent (clay-coloured faeces)
van den Bergh reaction	Indirect—positive	Biphasic	Direct—positive
Liver functions	Normal	Abnormal	Exaggerated abnormality
Blood picture	Anaemia, reticulocytosis, abnormal RBCs	Normal	Normal
Plasma albumin and globulin	Normal	Albumin: Decreases Globulin: Normal or increases A:G ratio: Decreases	Normal
Haemorrhagic tendency	Absent	Present due to lack of vitamin K	Present due to lack of vitamin K

Abbreviations: RBC, red blood cell; A:G ratio, albumin: globulin ratio
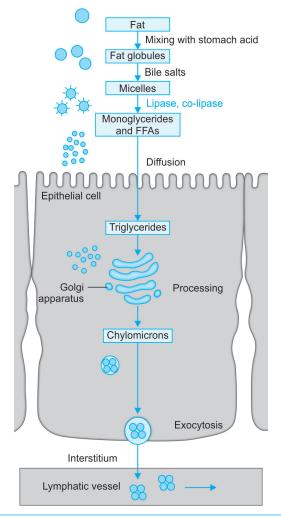
FIG. 3.16: Lipid digestion and absorption
Abbreviation: FFA, free fatty acid

exposure to air. Therefore, urine rich in urobilinogen (due to haemolysis) darkens on exposure to light.

Jaundice or icterus is the condition characterised by yellow colouration of the skin, mucous membrane and deeper tissues due to increased bilirubin level in blood. Jaundice is classified into three types:

1. Pre-hepatic or haemolytic jaundice
2. Hepatic or hepato-cellular jaundice
3. Post-hepatic or obstructive jaundice

The features of different types of jaundice are tabulated in **Table 3.14**.

Drugs such as halothane and isoniazid, which cause hepatic damage, may give rise to a picture resembling hepatic jaundice. On the other hand, drugs such as chlorpromazine and erythromycin estolate can give rise to a biochemical picture indistinguishable from extra-hepatic obstructive jaundice.

Portal Hypertension

In portal hypertension, the total vascular resistance of the hepatic sinusoids is increased. Portal blood flow through the liver is decreased as blood is diverted to alternative routes back to the great veins. This can result in the symptoms described next:

❏ *Ascites*: Raised hydrostatic pressure increases the rate of filtration from the visceral capillaries. This may cause the accumulation of the fluid in the peritoneal cavity to cause ascites.
❏ *Hepatic encephalopathy*: Failure of the liver to detoxicate toxic end products of protein metabolism in liver failure can result in hepatic encephalopathy.
❏ *Development of varices at the porto-caval anastomosis*: This includes oesophageal varices, gastric varices, colorectal varices, etc. A porto-caval shunt (anastomosis between portal vein and inferior vena cava) can decrease the tendency to bleed into the alimentary tract by diverting blood away from oesophageal varices. A porto-caval shunt, however, increases the risk of coma after bleeding into the alimentary tract.

Consequences of Removal of Various Organs of Gastrointestinal Tract

Gastrectomy: Removal of the stomach causes the ingested food to pass very rapidly to the small intestine. This can

result in unpleasant consequences due to fluid loss to the intestinal lumen and excessive insulin secretion. The syndrome is called the "dumping syndrome".

Pancreatectomy: Steatorrhoea may be seen following pancreatectomy because lack of lipase allows undigested fat to appear in the faeces.

Colectomy: Removal of the colon limits the ability of the alimentary tract to reabsorb water, resulting in the passage of watery stools.

Cholecystectomy: Removal of gall bladder causes loss of bile, resulting in poor digestion and impaired absorption of the meal. Hence, there may be abdominal discomfort, following a fatty meal.

Appendectomy: The appendix has no known function other than as a lymphoid organ and its immune function can be taken over by other lymphoid tissue in the intestines. Therefore, appendectomy is not associated with any digestive consequences.

Reproductive System

For information related to the endocrinology and embryology of the reproductive system, kindly refer to Chapters 11 and 7 respectively.

Physiology of Normal Pregnancy

Placenta

Placental Anatomy

Normal placental anatomy and development has been described in details in Chapter 7.

Function of the Placenta

The major role of placenta is to facilitate the exchange of various substances like gases (oxygen, carbon dioxide), nutrients and waste products between the mother and the foetus. In addition to its primary goal of facilitating transport between mother and foetus, the placenta is also a major endocrine organ. In almost all mammals, the placenta synthesizes and secretes the hormones—progestins and oestrogens, chorionic gonadotropins, relaxin and placental lactogens. Placental hormones have profound effects on both foetal and maternal physiology. The important functions performed by the placenta can be summarised as follows:

❑ Transportation
❑ Endocrine functions
❑ Immunological functions

Transport Function

The main mechanism of transport between the mother and the foetus is via diffusion across concentration or electrical gradient. Diffusion can be either free diffusion or via carrier proteins. Concentration gradient refers to the differences in concentration of various substances (gases, nutrients or waste products) between the mother and the baby's blood vessels. Most substances tend to move from places of higher concentration to places of lower concentration. The mechanism of transport across the placenta depends on the nature of the substance. The four major categories of substances that are transported across the placenta include gases (oxygen and carbon dioxide), waste substances, antibodies and nutrients.

Oxygen and carbon dioxide have very high permeability and are transferred by flow-limited passive diffusion across the placental barrier. The mean PO_2 in the mother is 50 mmHg and in the foetus is 30 mmHg. This pressure gradient facilitates the diffusion of O_2. On the other hand, though CO_2 crosses the placenta under a low pressure gradient, due to its extreme solubility in water, it crosses the placental barrier 20 times more rapidly than O_2. Another special property that allows efficient O_2 transport is that the foetal haemoglobin ($\alpha2\,\gamma2$) has a higher affinity for oxygen than a normal adult's red blood cells.

The waste products in baby's blood diffuse down the gradient into the mother's blood. The maternal antibodies, which pass through the placenta, provide passive immunity to the baby. Essential nutrients and minerals like iodine, sodium, calcium, etc. are also transferred. Glucose is transferred by facilitated diffusion and calcium by active carrier-mediated transport through the placental membrane. Glucose levels in foetal blood are usually 20–30% lower than in maternal blood, due to its rapid metabolisation by the foetus. Immunoglobulin IgG crosses by endocytosis and amino acids are actively transported by transporter proteins or a sodium-dependent carrier system.

Endocrine Function

One important function of the placenta is production and secretion of hormones. The main hormones produced by the placenta include oestrogen, progesterone, human placental lactogen (hPL), human chorionic gonadotropin (hCG) and relaxin. Leptin is another less significant placental hormone. Besides the various hormones, an enzyme, heat stable alkaline phosphatase and a placental protein, pregnancy-associated protein A (PAPPA) is also produced by the placenta.

Oestrogen: During pregnancy, oestrogen stimulates the growth of the uterus, enhances the blood flow between the uterus and the placenta, and causes breast enlargement in preparation for lactation. Oestrogen also stimulates uterine contractions at the time of delivery.

Progesterone: The main function of progesterone is to maintain the growth of uterine decidua. In addition, progesterone inhibits the movement of uterine walls during the first few weeks of pregnancy, following implantation in order to prevent the miscarriage of newly implanted embryo.

Beta-hCG: Human chorionic gonadotropin (hCG) is a glycoprotein with a half-life of 24 hours in comparison to luteinising hormone having a half-life of 2 hours (a 10-fold reduction). The molecular weight of hCG is approximately 30,000. hCG consists of two non-covalently linked sub-units called alpha (α) and beta (β). The α sub-unit is common to hCG, follicle-stimulating hormone (FSH), luteinising hormone (LH) and thyroid-stimulating hormone (TSH) and is encoded for by a single gene on chromosome 6. Hence, hCG has TSH-like activity. It is totally distinguishable from LH but its effects are similar. Intact hCG molecule can be produced virtually by all the normal human tissues. However, hCG produced at sites other than the placenta has little or no carbohydrate, and therefore it has a very short half-life and is rapidly cleared from the circulation. hCG can also be detected in the blood of normal men.

hCG is mainly secreted by the syncytiotrophoblast. Therefore, hCG will not be produced unless implantation has taken place and formation of placenta has occurred. Detection of hCG in maternal blood and urine 1 week after fertilisation forms the basis of urine pregnancy test. Implantation is believed to occur in most women 6–8 days after ovulation (or egg retrieval in cases of IVF). It is only following implantation that the placenta can be formed, which produces β-hCG. This means that the earliest a pregnancy test could become positive is day 6. Since the previous, older tests for hCG detected both alpha and beta chains, they could give a false positive result by detecting the high levels of LH at the time of ovulation. However, the newer tests available nowadays detect only the beta chains, which are unique to hCG, so LH levels cannot be detected. Therefore, these tests are more accurate. In the past, pregnancy tests were less sensitive and relied on high levels of hCG. So, pregnancy tests could be negative even after 12–14 weeks. Presently, the maximum sensitivity of a pregnancy test done on the first day of the missed period is approximately 90%. To avoid false negatives, women should give it a week from the 1st day.

The secretion of hCG peaks at 11–12 weeks' gestation (about 8 weeks post-ovulation). The maternal circulating hCG concentration is approximately 100 IU/L at the time of the expected but missed menses. The maximal hCG level is 100,000 IU/L, which is reached at 8–10 weeks' post-ovulation. hCG levels decrease to about 10,000–20,000 IU/L by 18–20 weeks. After that, the hCG levels almost remain stable at 10,000–20,000 IU/L until term.

Output of hCG doubles every 1.5 days in the early weeks and every 3 days at about 8 weeks. The level of hCG in the mother's blood usually rises until the end of second month and declines sharply by the fourth month. Conditions such as twin pregnancy and hydatidiform moles are associated with high hCG levels, which may also be responsible for their association with hyperemesis.

In the early phases of pregnancy (8–10 weeks), before the formation of placenta, the corpus luteum is responsible for the production of oestradiol and progesterone. The hCG rescues and maintains the function of corpus luteum through continuous production of progesterone. The hCG also promotes production of relaxin by the corpus luteum. The corpus luteum persists in early pregnancy for about 8 weeks when its functional role is taken over by the placenta. Persistence of the corpus luteum is believed to be due to hCG production from the trophoblast. This transition, occurring in the early pregnancy when the placenta takes over the role of progesterone production from the ageing corpus luteum, is known as the luteoplacental shift and occurs at around 7–8 weeks of gestation. Therefore, at around 7–8 weeks of gestation, placenta becomes the main source of progesterone production. The corpus luteum fails and eventually forms corpus albicans. Failure of the corpus luteum to maintain the progesterone levels prior to the shift may be associated with first-trimester miscarriage. Progesterone support is therefore required in cases of pregnancies via assisted reproductive techniques until the shift has occurred.

Human placental lactogen (hPL): Human placental lactogen is a single-chain polypeptide composed of 191 amino acids held together by two disulphide bonds. The half-life is very short (15 minutes); hence, it is used as an index of placental function. It has very similar structure to human growth hormone (hGH) but has only about 3% of its activity. The growth hormone-hPL gene family consists of five genes on chromosome 17. Two genes encode for hGH and three for hPL; however, only two of the hPL genes are active in the placenta, each producing the same hPL hormone.

The hPL has two major functions: Firstly, the hPL helps in regulating the mother's metabolism during pregnancy. Secondly, the hPL ensures that there is adequate amount of nutrient supply for the foetus by helping in movement of nutrients across the placenta. Blood levels of hPL are increased in the presence of hypoglycaemia and decreased in presence of hyperglycaemia. Since hPL has prolactin-like activity, it also stimulates breast development and preparation of milk along with oestrogen. However, its biological activity is less than that of prolactin.

The level of hPL in the maternal circulation is correlated with foetal and placental weight. In the second half of

pregnancy, hPL is a major force in the diabetogenic effects of pregnancy characterised by peripheral insulin resistance and hyperinsulinaemia. Its metabolic role is to mobilise lipids as free fatty acids. It induces insulin resistance and carbohydrate intolerance. It also stimulates the production of insulin-like growth factor-1 (IGF-1). During fasting states, hPL stimulates lipolysis, leading to an increase in circulating free fatty acids for utilization by the mother. Glucose and amino acids are conserved for the foetus. With sustained fasting, maternal ketone levels rise. These ketone bodies can be utilized by the foetus unlike the free fatty acids, which do not cross the placenta.

Relaxin: Relaxin is the hormone that is secreted by decidual cells of the placenta. Relaxin is found in pregnant humans but higher levels are present early in pregnancy than close to the time of birth. Relaxin promotes angiogenesis. In humans, it probably plays a more important role in the development of the interface between the uterus and the placenta than the role it plays during the birth process by relaxing the pelvic tissues and ligaments.

Leptin: This is the hormone produced by syncytiotrophoblast to regulate nutrient storage in the final stages of pregnancy

Immunological Function

The placenta plays an important role in maintaining foetal maternal immunological balance—passive immunity and suppression of the mother's immune response against the foetus (immunosuppression).

Passive immunity: Before birth, the baby's immune system is not fully developed yet, so the baby does not produce most of the antibodies by itself. Therefore, most of the antibodies needed by the baby are acquired from the mother directly. The membrane permeability of placenta enables the transport of antibodies from the mother to the baby; this is called passive immunity. The baby does not yet actively make its own antibodies but instead accepts the mother's antibodies passively. IgG immunoglobulins are the major type of antibodies that are able to cross the placental barrier.

Immunosuppression: Suppression of the mother's immune response against the foetus by the placenta enables the baby, which is a genetically foreign tissue to survive inside the mother's body.

Physiological Changes in Pregnancy

Normal physiological changes during pregnancy may be associated with clinical features that in a non-pregnant patient would point to organic disease. Some of these clinical features are summarised in **Table 3.15**.

TABLE 3.15	Non-pathological clinical features during pregnancy

- Polyuria
- Dyspepsia
- Decreased exercise tolerance
- Enlargement of the heart (with lateral displacement of the apex)
- A loud third heart sound
- An ejection systolic murmur (loudest at the left sternal edge)
- Fourth heart sound and a diastolic murmur (rarely)
- Left axis deviation with flattening or T-wave inversion in lead III on the electrocardiograph
- Reduction in the minimal alveolar concentration of volatile agents by 30–40% due to the sedative effect of progesterone
- Higher epidural pressures due to engorgement of the epidural veins.

Changes in the Genital Organs

Vagina

❑ *Chadwick's or Jacquemier's sign*: The vaginal walls show a bluish discolouration as the pelvic blood vessels become congested. This sign can be observed by 8–10 weeks of gestation.

❑ *Osiander's sign*: There is increased pulsation in the vagina felt through the lateral vaginal fornix at 8 weeks of gestation.

Uterus

❑ Enlargement of the uterus occurs due to hypertrophy and hyperplasia of the individual muscle fibres under the influence of hormones such as oestrogen and progestogens. The enlargement of the uterine muscles is due more to an increase in the size of the muscle cells rather than an increase in cell number.

❑ Uterine enlargement is more marked in the fundus. The uterine musculature during pregnancy is arranged in the form of three layers: (1) an outer hood-like layer arching over the fundus and extending into the various ligaments; (2) middle layer composed of dense network of muscle fibres perforated in all directions by the blood vessels and (3) an inner layer comprising of sphincter-like fibres around the orifices of fallopian tube and internal os of the cervix.

❑ Muscle fibres in the middle layer are arranged in an interlacing, "figure of 8" manner with blood vessels lying between these fibres.

As a result, when the uterine musculature contracts following the delivery of the foetus and placenta, the penetrating blood vessels are constricted, thereby preventing excessive blood loss. The occlusion of arteries during uterine contractions also diminishes placental perfusion, resulting in foetal hypoxia and/or foetal bradycardia.

❑ For the first few weeks of pregnancy, the uterus maintains its original pear shape but becomes almost spherical by

12 weeks of gestation. Thereafter, it increases more rapidly in length than in width becoming ovoid in shape. Until 12 weeks, the uterus remains a pelvic organ after which it can be palpated per abdominally.

❑ The uterus increases in weight from pre-pregnant 70 grams to approximately 1,100 grams at term.

❑ Due to uterine enlargement, the normal anteverted position is exaggerated up to 8 weeks. Since the enlarged uterus lies on the bladder making it incapable of filling, the frequency of micturition increases. However, after 8 weeks, the uterus more or less conforms to the axis of the inlet.

❑ *Hegar's sign*: At 6–8 weeks of gestation, the cervix is firm in contrast to the soft isthmus and fundus. Due to the marked softness of uterine isthmus, cervix and body of uterus may appear as separate organs. As a result, the isthmus of the uterus can be compressed between the fingers palpating vagina and abdomen, which is known as Hegar's sign.

❑ *Palmer's sign*: Regular rhythmic uterine contractions, which can be elicited during the bimanual examination, can be felt as early as 4–8 weeks of gestation.

❑ *Braxton Hicks contractions*: In the early months of pregnancy, uterus undergoes contractions known as Braxton Hicks contractions, which may be irregular, infrequent and painless without any effect on the cervical dilatation and effacement. Towards the last weeks of pregnancy, these contractions increase in intensity, thereby resulting in pain and discomfort for the patient and may occur after every 10–20 minutes, thereby assuming some form of rhythmicity. Eventually, these contractions merge with the contractions of labour.

❑ There is hypertrophy of the uterine isthmus to about three times its original size during the first trimester of pregnancy.

❑ After 12 weeks of pregnancy, the uterine isthmus unfolds from above downwards to get incorporated into the uterine cavity and also takes part in the formation of lower uterine segment.

❑ *Utero-placental blood flow*: Uterine blood flow increases gradually during pregnancy. There is an increase in the utero-placental blood flow ranging between 450–750 mL/minute near term. This increase is principally due to vasodilatation and can be nearly equal to 10–15% of the cardiac output. Vasodilatation can be related to prostaglandins such as prostacyclin. Blood flow can get interrupted during the uterine contractions.

❑ *Uterine soufflé*: This is a soft blowing sound synchronous with the maternal pulse. It is caused by rush of blood through the uterine arteries. On the other hand, foetal soufflé is a sharp whistling sound synchronous with the foetal pulse. It is caused by the rush of blood through the umbilical arteries.

Cervix

❑ There occurs hypertrophy and hyperplasia of the elastic and connective tissue fibres and increase in vascularity within the cervical stroma. This is likely to result in cervical softening (known as Godell's sign), which becomes evident by 6 weeks of pregnancy. Increased vascularity is likely to result in bluish discolouration beneath the squamous epithelium of portio vaginalis resulting in a positive Chadwick's sign.

❑ With the advancement of pregnancy, there is marked proliferation of endocervical mucosa with downward extension of the squamo-columnar junction. There is copious production of cervical secretions resulting in the formation of a thick mucus plug that seals the cervical canal. This mucus plug is rich in cytokines and immunoglobulins, and acts as an immunological barrier to protect the uterine contents against infection from the vagina.

❑ When the cervical mucus (secreted during pregnancy) is spread over the glass slide and dried, it shows a characteristic crystallization or beading pattern due to presence of progesterone.

Weight Gain and Metabolism

❑ *Maternal weight gain*: The average total weight gain during pregnancy is about 12.5 kilograms. The weight gain can be attributable to the following causes: foetus (3.4 kg); placenta (650 g); and amniotic fluid (800 g). The other major components of maternal weight gain during pregnancy involve the uterus (970 g); breasts (400 g); blood (1,250 g); extracellular extravascular fluid (1,700 g); and fat (3,500 g). Maternal weight gain averages 0.35 kg per week in early pregnancy, 0.45 kg per week in mid-pregnancy, and 0.35 kg per week in late pregnancy. Fat stores are laid down by the mother primarily in the first half of pregnancy and account for approximately 3 kilograms.

❑ *Basal metabolic rate*: Basal metabolic rate is increased by about 15% at term.

Changes in the Breast

Following changes are likely to take place in the breasts during pregnancy:

❑ The breast enlargement is due mainly to oestrogen and progesterone. Marked proliferation and hypertrophy of mammary ducts occurs under the effect of oestrogen and that of alveoli occurs under the effect of oestrogen and progesterone.

❑ Hypertrophy of the connective tissue stroma and increased vascularity results in the appearance of bluish veins under the breast skin. The axillary tail of breasts becomes enlarged and painful.

❏ The nipples become larger, erectile and pigmented. Hypertrophy of the sebaceous glands in the areola results in the formation of Montgomery's tubercles. Outer zone of irregular pigmented area appears around the areola in the second trimester, resulting in the formation of secondary areola. Sticky, thick, yellowish secretions can be squeezed from the breasts after about 12th week of pregnancy.

Haematological Changes during Pregnancy

The following haematological changes occur during pregnancy:

❏ Increase in the blood volume starting from 6th week of pregnancy. By 30–32 weeks, the blood volume may have increased by 40–50% above the non-pregnant level. The extracellular fluid increases by about 3 litres.

❏ There is an approximate 6–8 L increase in total body water (TBW) during pregnancy, resulting in the reduction of many of the electrolyte concentrations including urea, sodium/potassium and albumin.

❏ The increase in the blood volume is due to an increase in the plasma volume (by 50%) and RBC volume (by 20–30%). The disproportionate increase in the plasma and RBC volume is likely to cause haemodilution, resulting in a physiological haemodilution of pregnancy.

❏ Although more plasma than erythrocytes is added to the maternal circulation, the increase in erythrocyte volume averages to about 450 mL. Due to this, there is a slight decrease in the haemoglobin concentration, haematocrit and blood viscosity during pregnancy. There is a significant rise in the ESR due to haemodilution and a reduction in haemoglobin levels. Haematocrit falls due to the increased TBW early in pregnancy but stabilises and may increase towards the end of pregnancy.

❏ There is an increase in the concentration of total plasma proteins from a normal value of 180 grams in non-pregnant state to 230 grams at term. However, due to haemodilution, there is an actual decrease in the concentration of plasma proteins from 7 gm% to 6 gm%. Moreover, there is a reduction in A:G ratio from 1.7:1 to 1:1.

❏ There is neutrophil leucocytosis.

❏ Average haemoglobin concentration at term is 12.5 g/dL. A decline in haemoglobin value of less than 11 g/dL, especially late in pregnancy is abnormal and can be considered to be due to anaemia (most likely iron deficiency anaemia) rather than hypervolemia of pregnancy.

❏ Peripheral blood flow increases due to fall in peripheral vascular resistance.

Coagulation Parameters

Pregnancy is a hypercoagulable state. There is an increase in the fibrinogen levels from 200–400 mg% in the non-pregnant state to 300–600 mg% during pregnancy. There is also an increase in the activity of other clotting factors such as factors II, VII, VIII, IX and X. There is a slight decrease in the activity of other clotting factors such as factors XI and XIII. These changes in combination with venous stasis, lead to a marked tendency to clotting, which increases more than fivefold in pregnancy. Bed rest greatly increases the risk. Increased consumption of cigarettes during pregnancy is likely to be associated with an additional risk. Platelet count does not rise during pregnancy.

Physiological Changes during Anaemia in Pregnancy

❏ In cases with moderate to severe anaemia, the cardiac output rises to compensate for the blood's reduced O_2 carrying capacity.

❏ Bruits are common because increased flow velocity and decreased blood viscosity increase the likelihood of turbulent flow.

❏ Level of 2,3-DPG is increased, shifting the dissociation curve to the right so that blood gives up its oxygen more easily.

❏ Arterial PO_2 is normal; it is O_2 content that is reduced.

❏ Capacity to raise oxygen consumption during exercise is decreased due to the reduced capacity to deliver O_2 to the muscles.

Iron Metabolism during Pregnancy

There is an increased iron requirement during pregnancy amounting to about 1,000 mg, which may occur due to the following reasons:

❏ 270 mg of iron is actively transferred to foetus. Foetus utilises maternal iron for building up haemoglobin molecules.

❏ 170 mg is lost through various routes of excretion, primarily the gastrointestinal tract.

❏ Total amount of iron transferred to placenta and cord is 90 mg.

❏ 450–500 mg of iron is utilised due to expansion in the total volume of circulating maternal erythrocytes.

❏ Since pregnancy results in amenorrhea, this leads to a saving of nearly 240–300 mg of iron in the form of menstrual blood. Thus, total iron requirement during pregnancy is 600–700 mg (6–7 mg of daily requirement of elemental iron for about 100 days). Dietary iron along with that mobilised from the iron stores may be sometimes insufficient to meet average demands imposed by pregnancy.

Most units in the UK prescribe iron during pregnancy only if there is evidence of anaemia. A significant minority of women who enter pregnancy with depleted iron stores as a result of menstrual iron loss and inadequate diet would also be candidates for iron supplements. On the other hand, there are concerns that prescribing iron unnecessarily to women may lead to problems due to unphysiological levels of haemoglobin.

Changes in the Cardiovascular System during Pregnancy

Various cardiovascular changes occurring during normal pregnancy are summarised in **Table 3.16** and **Figure 3.17** and are described next in the text:

❏ There is a 30–50% increase in cardiac output. Normally, the cardiac output starts increasing by around 5th week and increases rapidly until the 34th week of gestation, following which it plateaus or continues to increase slightly. The increase in cardiac output is achieved by three factors:
  1. An increase in preload because of greater blood volume: Blood volume increases by 40–50% during normal pregnancy. The increase in plasma volume is greater than the increase in red blood cell mass, contributing to the fall in haemoglobin concentration (i.e. the "physiological anaemia in pregnancy"). The cause of underlying blood volume increase is related to an oestrogen-mediated stimulation of the renin-angiotensin system, which results in sodium and water retention.
  2. Reduced afterload due to reduction in systemic vascular resistance
  3. A rise in the maternal heart rate by 10–15 beats/minute

❏ Blood pressure and cardiac output are also reduced during epidural analgesia which may be administered during labour and delivery.

❏ Stroke volume increases during the first and second trimesters, but declines in the third trimester due to the compression of inferior vena cava by the uterus.

❏ Both plasma and interstitial colloid oncotic pressure decrease throughout pregnancy. There is an accompanying increase in the capillary hydrostatic pressure. Increase in capillary hydrostatic pressure or decrease in colloid oncotic pressure is likely to cause oedema.

❏ The systemic vascular resistance decreases by about 30% due to hormone-mediated vasodilation. The blood pressure decreases by 10% at 20 weeks' gestation. This results in an increased blood flow to various organs, particularly the uterus.

❏ A decline in systemic arterial pressure begins to occur during first trimester, reaches a nadir in mid-pregnancy and returns towards pre-gestational level before term. Blood pressure typically falls by about 10 mmHg below baseline by the end of the second trimester because of reduction in systemic vascular resistance and the addition of new blood vessels in the uterus and placenta. Systolic blood pressure does not change during pregnancy; diastolic blood pressure is reduced in the first two trimesters and returns to non-pregnant levels at term.

❏ Blood pressure is lower when the woman is lying down either in the supine position or on her side in comparison to the sitting position.

❏ Central venous pressures and pulmonary capillary wedge pressures remain stable.

❏ The gravid uterus pushes the diaphragm up, thereby displacing the heart forwards and towards the left.

ECG-Related Changes during Pregnancy

Due to the haemodynamic physiological changes during pregnancy, the ECG during pregnancy is slightly different from that during the non-pregnant state. The following changes can occur in the ECG:

❏ Left-axis deviation
❏ T-wave inversion or flattening in lead III
❏ Presence of Q waves in both the leads aVF and III.
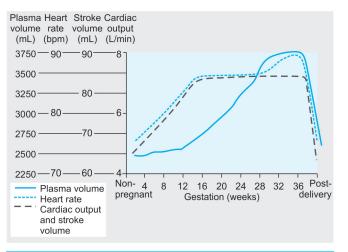
FIG. 3.17: Normal cardiovascular changes during pregnancy

TABLE 3.16	Cardiovascular changes during normal pregnancy		
Haemodynamic parameter	**Change during normal pregnancy**	**Change during labour and delivery**	**Change during postpartum**
Blood volume	Increases by 40–50%	Increases	Decreases (auto-diuresis)
Heart rate	Increases by 10–15 beats/min	Increases	Decreases
Cardiac output	Increases by 30–50% above the base line	Additional increase by 50%	Decreases
Blood pressure	Decreases by 10 mmHg	Increases	Decreases
Stroke volume	Increases during the first and second trimesters; decreases during the third trimester	Additional increase of 300–500 mL with each uterine contraction	Decreases
Systemic vascular resistance	Decreases	Increases	Decreases

Changes in the Respiratory System during Pregnancy

❏ There occurs a state of hyperventilation, resulting in an increase in tidal volume and respiratory minute volume by 40%. This occurs due to the effect of progesterone on respiratory centre and increase in the sensitivity of respiratory centre to CO_2.
❏ There is a decrease in the total lung capacity, residual volume, expiratory reserve volume (increases to about 200 mL), IRV, chest compliance and airway resistance.
❏ This hyperventilation causes changes in acid-base balance. There is fall in the arterial $PaCO_2$ from 38 to 32 mmHg and a rise in PaO_2 from 95 to 105 mmHg. These changes facilitate the transfer of CO_2 from foetus to the mother and O_2 from mother to the foetus. There is an overall rise in the pH and a base excess of 2 mEq/L. This results in respiratory alkalosis. Increased excretion of bicarbonates by the kidneys results in partial compensation.
❏ There occurs no change in the vital capacity, respiratory rate, lung compliance and the ratio FEV1/FVC.

Changes Occurring in the Renal System during Pregnancy

Glomerular Filtration Rate and Renal Plasma Flow

❏ There is an increase in GFR by 50% and RPF by 50–75% by 16 weeks of pregnancy and is maintained until 34 weeks. While the GFR remains elevated throughout pregnancy, RPF falls after 34 weeks of pregnancy. The increase in GFR can be up to 60% in late pregnancy.
❏ As a result, there is reduction in maternal plasma levels of creatinine, blood urea, uric acid, etc.
❏ There is failure of complete absorption of substances, such as glucose, uric acid, amino acid, etc., from the renal tubules, resulting in an increased excretion of proteins, amino acids and glucose.
❏ Serum creatinine levels decrease during normal gestation to greater than 0.8 mg/dL.
❏ There is decreased specific gravity; decreased plasma sodium, potassium and urea concentrations.
❏ The renal blood flow increases by 50% reaching a peak in the second trimester and then falls slightly in the third trimester.
❏ The kidneys normally increase in length during pregnancy.
❏ The renal pelvis, ureters and the urinary collecting system dilate considerably during pregnancy. This is thought to be mainly an effect of pregnancy hormones, particularly progesterone. Compression of the ureters at the pelvic brim from the expanding uterus could be an additional contributing factor. These changes do not reverse for many weeks after the puerperium, and if an intravenous pyelogram is required, it should not be arranged for at least three months after childbirth.

Acid-Base Balance

❏ There is decreased bicarbonate threshold, and progesterone stimulates the respiratory centre. Therefore, serum bicarbonate decreases by 4–5 mEq/L.
❏ The pH of venous blood and interstitial fluid is 7.35; intracellular pH averages 7.0. The main buffer system controlling blood pH is carbonic acid/sodium bicarbonate (in cells, potassium and magnesium bicarbonate).
❏ Maternal respiratory acidosis is transmitted to the foetus, as the excess carbon dioxide can cross the placenta. The placenta is not permeable to hydrogen ions; thus, metabolic acidosis has less effect on the foetus.

Plasma Osmolality

❏ Serum osmolality decreases by 10 mOsm/L during normal gestation. Increased placental metabolism of vasopressin may cause transient diabetes insipidus during pregnancy.

Changes Occurring in the Gastrointestinal Tract during Pregnancy

❏ There is increased congestion of the gums. Due to this, they may become spongy and thereby bleed to touch.
❏ There is reduced muscle tone and motility of the entire gastrointestinal tract under the effect of progesterone. This may be responsible for producing constipation.
❏ Relaxation of the cardiac sphincter may result in regurgitation of gastric acid into the oesophagus, thereby producing chemical esophagitis and heart burns. The women may become more susceptible to develop reflux.
❏ Gastric secretions are also reduced and the emptying time of the stomach is delayed. This results in a reduced risk for the development of peptic ulcer disease.
❏ The liver maintains a normal blood flow and size.
❏ Gall bladder motility and emptying rate are both reduced in pregnancy.

Changes in the Liver Function Tests during Pregnancy

❏ The levels of bilirubin largely remain unchanged during pregnancy.
❏ Due to pregnancy-related volume expansion, there is a fall in the levels of albumin, total protein and transaminases (alanine transaminase and aspartate transaminase).
❏ The alkaline phosphatase is frequently elevated but the remainder of the liver function tests should be more or less within the normal range. Levels of alkaline phosphatase increase throughout pregnancy, reaching a peak during the third trimester. This could be related

TABLE 3.17	Tests for liver function: values during the pregnant and non-pregnant state	
Liver function test	**Non-pregnant state**	**Pregnant state**
Alkaline phosphatase (IU/L)	30–130	32–418
Aspartate aminotransferase (IU/L)	7–40	10–30
Alanine aminotransferase (IU/L)	0–40	6–32
Albumin (g/L)	35–46	28–37
Total protein (g/L)	64–86	48–64
Bilirubin (μmol/L)	0–17	3–16

Source: Appendix 2: Normal laboratory values in pregnancy/non-pregnancy. In: Handbook of obstetric medicine, 4th edition. London: Informa Healthcare; 2010. pp. 273-4.

to placental production of alkaline phosphatase. Therefore, interpretation of the liver function tests during the pregnancy must be done after taking into account the above-mentioned normal physiological changes in the liver function tests **(Table 3.17)**.

Alkaline phosphatase is an enzyme present in the canalicular and sinusoidal membranes of the liver, as well as bone, intestine and placenta, and there is a large range of isoenzymes. Levels of alkaline phosphatase are also raised in cholestasis and may be elevated in bone disease, which is why levels of different isoenzymes may be required to differentiate the source of the elevation.

Carbohydrate Metabolism during Pregnancy

Insulin Resistance

❏ Pregnancy is a diabetogenic state, resulting in development of insulin resistance. In the first half of pregnancy, there is an increased sensitivity to insulin and therefore there is a tendency towards development of hypoglycaemia. On the other hand, the second half of pregnancy (especially after 24 weeks of gestation) is related with development of insulin resistance.

Causes of insulin resistance during pregnancy include:
❏ There occurs hyperplasia and hypertrophy of the beta cells of pancreas.
❏ Steroid hormones (especially corticosteroids, oestriol and progesterone), which are produced late in pregnancy, show an anti-insulin effect.
❏ Some insulin may be destroyed by placenta and kidneys.
❏ Production of hPL by placental tissue.

Changes Related with Carbohydrate Metabolism

❏ There is transfer of increased amount of glucose from the mother to the foetus throughout the pregnancy.
❏ These changes help in ensuring continuous supply of glucose to the foetus.
❏ There is mild fasting hypoglycaemia, postprandial hyperglycaemia and hyperinsulinaemia as well as greater suppression of glucagon.

❏ When fasting is prolonged in pregnant women, keto-naemia rapidly results.
❏ In the postprandial state, there is a switch from glucose to lipids as a source of principal fuel. Increasing levels of hPL with gestation is responsible for increased lipolysis and liberation of free fatty acids.

Changes in Thyroid Glands during Pregnancy

❏ There is an increased production of thyroid hormones by 40–100% in order to meet the maternal and foetal requirements.
❏ Moderate enlargement of thyroid glands is caused by glandular hyperplasia and increased vascularity.
❏ There is a marked and early increase in hepatic production of thyroid-binding globulins (TBG) and placental production of hCG. Levels of TBG start increasing from first trimester. They peak at 20 weeks of pregnancy and plateau during the remainder part of pregnancy. As a result of increased production of plasma protein binding molecules, such as TBG, concentration of total thyroxin (T4) is increased, but the levels of free T4 are typically normal.
❏ Levels of thyroid-releasing hormone (TRH) are not increased during normal pregnancy. However, this hormone can cross the placenta and stimulates the foetal pituitary to secrete thyrotropin.
❏ Serum levels of thyroid-stimulating hormone decrease slightly during the first trimester in response to elevations of hCG.
❏ Foetal thyroid function is totally dependent on maternal supply until 12 weeks of gestation when the thyroid forms, until which time the foetus does not produce the thyroid hormone. Tri-iodothyronine crosses the placenta and can cause foetal thyrotoxicosis.

Hormonal Changes during Pregnancy

❏ During pregnancy, there is an increase in the serum concentrations of the following:
  • Sex hormone-binding globulin (β-hCG)
  • Prolactin
  • Total thyroxin (as there is an increase in the binding proteins)
  • Oestrogens
  • Beta human chorionic gonadotropin (β-hCG)
  • Progestogens
❏ There is inhibition of the secretion of both LH and FSH caused due to the raised levels of oestrogen/prolactin.
❏ The pituitary gland increases the production of prolactin and adrenocorticotropin (ACTH) but the production of growth hormone is reduced.
❏ Plasma concentrations of cortisol, aldosterone, rennin and angiotensin rise.

Choose the Single Best Answer (SBA)

Q 1. Which of the following is not true regarding neutrophil granulocytes?
A. Are the most common leucocyte in normal blood
B. Contain proteolytic enzymes
C. Have a lifespan in the circulation of 3–4 weeks
D. Contain actin and myosin microfilaments
E. Are present in high concentration in pus

Q 2. Erythrocyte sedimentation rate (ESR) is increased in the following circumstances?
A. Following the infusion of high-molecular-weight dextran
B. In men compared with women
C. In polycythaemia rubra vera
D. Increase in the plasma fibrinogen concentrations
E. In young age

Q 3. Will the following factors lower the ESR?
A. Systemic lupus erythematosus
B. Female gender
C. Macrocytosis
D. Polycythaemia
E. Rheumatoid arthritis

Q 4. Prothrombin time is not increased in which of the following?
A. Aspirin therapy
B. Cirrhosis
C. Disseminated intravascular coagulation
D. Unfractionated heparin therapy
E. Warfarin therapy

Q 5. Normal clotting of blood requires
A. Inactivation of heparin
B. Inactivation of plasmin (fibrinolysin)
C. Calcium ions
D. An adequate intake of vitamin D
E. An adequate intake of vitamin C

Q 6. The conversion of fibrinogen to fibrin
A. Is affected by prothrombin
B. Involves the disruption of certain peptide linkages by a proteolytic enzyme
C. Is followed by reduced polymerisation of fibrin monomers
D. Is not inhibited by heparin
E. Is reversed by plasmin (fibrinolysin)

Q 7. A 15-year-old child is admitted to hospital with recent onset of widespread purpura (pin-head areas of haemorrhage into the skin). **What is the most likely abnormality to be revealed by the laboratory investigations in this case?**
A. Deficiency of vitamin K
B. Deficiency of factor VIII
C. Increased fibrinogen level
D. Platelet count 90 x 10^9 per litre
E. Deficiency of prothrombin

Q 8. A 50-year-old man is receiving anti-coagulant therapy (warfarin, a vitamin K antagonist) after heart valve replacement. He is admitted to hospital with haematuria (blood in the urine) and his INR (international normalised ratio, a measure of the prothrombin clotting time in relation to the normal time) is found to be 4.2. What is the most likely abnormality to be revealed by the laboratory investigations in this case?
A. Deficiency of vitamin K
B. Deficiency of factor VIII
C. Increased fibrinogen level
D. Platelet count 20 x 10^9 per litre
E. Deficiency of prothrombin

Q 9. A 90-year-old woman has blotchy purple areas about 5 cm diameter on her hands and arms. They are not uncomfortable and she has no health complaints. What is the most likely abnormality to be revealed by the laboratory investigations in this case?
A. Capillary abnormality
B. Deficiency of prothrombin
C. Increased fibrinogen level
D. Platelet count 20 × 10^9 per litre
E. Deficiency of vitamin K

Q 10. A 70-year-old man is operated on for aneurysm of aorta. Severe bleeding requires infusion of forty units of blood. His recovery is complicated by a bleeding tendency and he is found to have a very low level of fibrinogen. His treatment includes administration of heparin. What is the most likely abnormality to be revealed by the laboratory investigations in this case?
A. Deficiency of vitamin K
B. Deficiency of factor VIII
C. Increased fibrinogen level
D. Disseminated intravascular coagulation
E. Deficiency of prothrombin

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1. C   2. D   3. D   4. A   5. C   6. B   7. D   8. E   9. A   10. D

Q 11. **A 10-year-old child with no known medical problems has been admitted to hospital for persistent bleeding after tooth extraction. Haemostasis had been achieved initially after the extraction but subsequently prolonged oozing from the tooth socket began. What is the most likely abnormality to be revealed by the laboratory investigations in this case?**

A. Deficiency of factor VIII
B. Deficiency of vitamin K
C. Increased fibrinogen level
D. Disseminated intravascular coagulation
E. Deficiency of prothrombin

Q 12. **Which of the following substances occur in lower concentration in foetal blood in comparison to the maternal blood?**

A. Amino acids
B. Ca^{2+}
C. CO_2
D. Glucose
E. Urea

Q 13. **Which of the following is not true regarding prostaglandins?**

A. Are involved in the onset of labour
B. Are oxytocic
C. Have a role in causing dysmenorrhoea
D. Maintains the corpus luteum in early pregnancy
E. May be important in the development of menorrhagia

Q 14. **Which of the following is not true regarding bleeding from a small cut in the skin?**

A. Is normally diminished by local vascular spasm
B. Ceases within about five minutes in normal people
C. Is prolonged in severe factor VIII (anti-haemophilic globulin) deficiency
D. Is greater from warm skin than from cold skin
E. Is reduced if the affected limb is elevated

Q 15. **Which of the following is not correct regarding the monocytes?**

A. Originate from precursor cells in bone marrow
B. Can increase in number when their parent cells are stimulated by factors released from activated lymphocytes
C. Unlike granulocytes, do not migrate across capillary walls
D. Can transform into large multi-nucleated cells in certain chronic infections
E. Manufacture immunoglobulin M

Q 16. **Which of the following is not correct regarding the erythrocytes?**

A. Are responsible for the major part of blood viscosity
B. Contain the enzyme carbonic anhydrase
C. Metabolise glucose to produce CO_2 and H_2O
D. Swell to bursting point when suspended in 0.9 percent (150 mmol/litre) saline
E. The walls deform easily to squeeze through capillaries

Q 17. **Atypical lymphocytosis is not a feature of which of the following diseases?**

A. Cytomegalovirus (CMV) infection
B. Epstein-Barr virus (EBV) infection
C. Influenza B infection
D. Rubella infection
E. Toxoplasmosis

Q 18. **Which of the following is true regarding T lymphocytes?**

A. Are infected by Epstein-Barr virus in infectious mononucleosis
B. Are the primary host response in bacterial infection
C. Compose the majority of lymphocytes in plasma
D. T-cell lymphoma has a better prognosis in comparison to B cell lymphoma
E. Produce IgG

Q 19. **Red cell formation is increased in which of the following?**

A. In blood donors one week after a blood donation
B. In patients with haemolytic anaemia
C. By giving injections of erythropoietin to nephrectomised patients
D. All of the above
E. None of the above

Q 20. **Vitamin B_{12} deficiency may**

A. Result from disease of the terminal part of the ileum
B. Cause a reduction in the circulating platelet level
C. Cause pathological changes in the central nervous system
D. All of the above
E. None of the above

Q 21. **Which of the following statement regarding platelets is not true?**

A. In a normal person, 20% of the platelets are found in the spleen
B. Their life span in circulation is about 9–10 days
C. They are formed in the bone marrow from megakaryocytes
D. They contain adenosine diphosphate and serotonin
E. They produce prostacyclin

11. A   12. D   13. D   14. C   15. E   16. D   17. C   18. C   19. D   20. D   21. E

Q 22. Circulating red blood cells
- A. Are about 1 percent nucleated
- B. May show an intracellular network pattern if appropriately stained
- C. Are distributed evenly across the blood stream in large blood vessels
- D. Travel at slower velocity in venules than in capillaries
- E. Can easily pass through the capillaries without undergoing any deformation

Q 23. Which of the following statement is true regarding blood eosinophils?
- A. Have agranular cytoplasm
- B. Are about a quarter of all leucocytes
- C. Are relatively scarce in the mucosa of the respiratory, urinary and alimentary tracts
- D. Release cytokines
- E. Increase in number in viral infections

Q 24. Which of the following is not correct regarding lymph?
- A. Contains plasma proteins
- B. Lymph vessels are involved in the uptake and transport of absorbed fat
- C. Production increases during muscular activity
- D. Does not normally contain cells
- E. Flow is aided by contraction of adjacent skeletal muscles

Q 25. Which of the following is correct regarding the deficiency of factor VIII (anti-haemophilic globulin)?
- A. Increases the bleeding time
- B. Is due to an abnormal gene on the Y chromosome
- C. To 75 percent of its normal value results in excessive bleeding after tooth extraction
- D. Causes small (petechial) haemorrhages into the skin to cause purpura
- E. Affects the intrinsic pathway for blood coagulation

Q 26. Which of the following statement is true regarding the cell membranes in skeletal muscle?
- A. Are impermeable to fat-soluble substances
- B. Are more permeable to sodium than to potassium ions
- C. Becomes less permeable to glucose in the presence of insulin
- D. Become less permeable to potassium in the presence of insulin
- E. Show invaginations which connect to a system of intracellular tubules involved in excitation contraction coupling

Q 27. Which of the following is true about the smooth muscle cells?
- A. Controlled by the autonomic nervous system
- B. Do not contain actin and myosin
- C. Mitochondria are absent
- D. Present a striated appearance
- E. Do not cause spontaneous muscle contraction

Q 28. Which of the following is not true regarding the prothrombin time (PT)?
- A. Assesses the extrinsic pathway of the blood coagulation cascade
- B. Is increased with low-molecular-weight heparin
- C. Is increased with warfarin
- D. Is prolonged in patients with fat malabsorption
- E. May be restored to normal by the administration of vitamin K

Q 29. Pulmonary surfactant increases which of the following?
- A. The surface tension of the fluid lining alveolar walls
- B. Lung compliance
- C. In effectiveness as the lungs are inflated
- D. In amount when pulmonary blood flow is interrupted
- E. Breathing difficulties in the foetuses during the last month of pregnancy

Q 30. Which of the following is not true regarding the respiratory centre?
- A. Is in the medulla oblongata
- B. Sends impulses to inspiratory muscles during quiet breathing
- C. Sends impulses to expiratory muscles during quiet breathing
- D. Is involved in the swallowing reflex
- E. Is involved in the vomiting reflex

Q 31. Which of the following regarding the carotid bodies is not correct?
- A. Are stretch receptors in the walls of the internal carotid arteries
- B. Have the greatest flow rate/unit volume yet described in the body
- C. Are influenced more by blood PO_2 than by its oxygen content
- D. Generate more afferent impulses when blood H^+ ion concentration rises
- E. The aortic bodies are mainly responsible for the increased ventilation in hypoxia

Q 32. Which of the following statement is correct regarding carbon dioxide?
- A. Is carried as carboxyhaemoglobin on the haemoglobin molecule
- B. Uptake by the blood increases its oxygen-binding power
- C. Uptake by the blood leads to similar increases in H^+ and HCO_3^- ion concentrations
- D. Stimulates ventilation when breathed at a concentration of 20 percent
- E. Content is greater than oxygen content in arterial blood

22. B   23. D   24. D   25. E   26. E   27. A   28. B   29. B   30. C   31. A   32. E

Q 33. Bronchial smooth muscle contracts in response to
A. Bronchial mucosal irritation
B. Local beta-adrenoceptor stimulation
C. Circulating noradrenaline
D. All of the above
E. None of the above

Q 34. Which of the following is/are true concerning the oxygen dissociation curve (ODC)?
A. At 75% saturation the PO_2 is 40 mmHg (5.3 kPa)
B. Is hyperbolic in shape
C. Is shifted to the left by increase in 2,3-DPG
D. Is shifted to the right in methaemoglobinaemia
E. The Bohr effect shifts the curve to the left

Q 35. Oxyhaemoglobin dissociation curve shifted to the left in which of the following conditions?
A. Alkalosis
B. Hypoxia
C. Increased pCO_2
D. Increasing body temperature
E. Increasing concentration of 2,3-DPG

Q 36. Which of the following statement regarding the renal clearance of a substance is correct?
A. Is inversely related to its urinary concentration, U
B. Is indirectly related to the rate of urine formation, V
C. Is directly related to its plasma concentration, P
D. Is expressed in units of volume per unit time
E. Must fall in the presence of metabolic poisons

Q 37. Which of the following statement is correct regarding the fluid in the distal part of the proximal convoluted tubule?
A. Urea concentration is less than in Bowman's capsule
B. pH is less than 6 when the kidneys are excreting an acid urine
C. Glucose concentration is similar to that in plasma
D. Osmolality is about 25 percent that of glomerular filtrate
E. Bicarbonate concentration is lower than in plasma

Q 38. Which of the following is not correct regarding reabsorption by the renal tubules?
A. More water every hour than the entire plasma volume is reabsorbed
B. All filtered HCO_3^- in cases of respiratory acidosis
C. All filtered amino acids
D. All filtered plasma proteins
E. More K^+ than Cl^-

Q 39. As plasma glucose concentration rises above normal, glucose
A. Filtration decreases linearly
B. Transport maximum Tm increases linearly
C. Clearance increases linearly
D. Reabsorption increases and then levels off
E. Excretion increases and then decreases

Q 40. Hydrostatic pressure in renal glomerular capillaries
A. Is lower than pressure in efferent arterioles
B. Rises when afferent arterioles constrict
C. Is lower than in most capillaries at heart level
D. Falls by 10% when arterial pressure falls by 10%
E. Falls along the length of the capillary

Q 41. Which of the following is true concerning creatinine?
A. Has a plasma clearance rate equivalent to renal plasma flow
B. Is filtered out by the glomerulus
C. Is reabsorbed significantly by the proximal tubules
D. Plasma concentration does not change after protein ingestion
E. Plasma concentration increases during the first trimester of pregnancy

Q 42. Which of the following substances can be reabsorbed from the renal tubule?
A. Creatinine
B. Inulin
C. Mannitol
D. Sucrose
E. Urea

Q 43. In which of the regions of the nephron is the macula densa tissue situated?
A. Thick ascending limb
B. Thin descending limb
C. Distal convoluted tubule
D. Proximal straight tubule
E. Medullary collecting duct

Q 44. Which of the following regions of nephron does the water leave via diffusion?
A. Descending thin limb
B. Thick ascending limb
C. Proximal convoluted tubule
D. Distal convoluted tubule
E. Bowman's capsule

Q 45. This enzyme is present in the liver cell cytosol, brain and myocardium. A rise may occur in hepatitis and myocardial infarction (MI).
A. Alanine aminotransferase (ALT)
B. Alkaline phosphatase (ALP)
C. Aspartate aminotransferase (AST)
D. γ-Glutamyl transpeptidase
E. C-reactive protein (CRP)

Q 46. This is a microsomal enzyme. Its activity can be induced by phenytoin.
A. γ-Glutamyl transpeptidase
B. C-reactive protein (CRP)
C. Alanine aminotransferase (ALT)
D. Alkaline phosphatase (ALP)
E. None of the above

33. A   34. A   35. A   36. D   37. E   38. E   39. D   40. E   41. B   42. E   43. C   44. A   45. C   46. A

Q 47. **Which of the following is not correct concerning bile?**

A. 95% is reabsorbed in the terminal ileum
B. Has a pH of 8
C. Increases the absorption of vitamin K
D. Is concentrated under the influence of secretin
E. Production is inhibited after vagal stimulation

Q 48. **Glomerular filtration rate may be measured using**

A. Insulin
B. Para-aminohippuric acid
C. Glucagon
D. Glucose
E. Inulin

Q 49. **Which of the following statements regarding the cerebrospinal fluid is correct?**

A. Is actively secreted by the choroid plexus
B. Is the main source of the brain's nutrition
C. Has the same pH as arterial blood
D. Has a higher glucose concentration than has plasma
E. Has a higher calcium concentration than has plasma

Q 50. **Which of the following has a lower concentration in the cerebrospinal fluid (CSF) than plasma?**

A. Sodium
B. Potassium
C. Magnesium
D. Bicarbonate
E. None of the above

Q 51. **Which of the following is true regarding electrocardiogram (ECG) interpretation?**

A. 0.16 seconds represents a normal QRS duration
B. 50 mm per second is the standard paper speed
C. Standard calibration implies that 0.1 mV is equivalent to a deflection of 1 mm
D. The PR interval is measured from the start of the p wave to the end of the QRS complex
E. The QT interval is measured from the start of the QRS complex to the start of the T wave

Q 52. **Which of the following is not correct concerning the electrocardiogram (ECG) of an adult human?**

A. A PR interval of 0.3 seconds indicates impaired conduction
B. During the isoelectric phase between the S and T waves, the intracellular potential in ventricular muscle cells is positive with respect to the interstitial fluid
C. Normal QT interval is 0.5s
D. The Q wave coincides with repolarisation of the atria
E. The R wave coincides with depolarisation of the apex of the heart

Q 53. **Which of the following electrocardiogram (ECG) changes are associated with a suspected myocardial infarction?**

A. In a subendocardial MI, ST elevation and T wave inversion occurs in leads facing the infarcted area
B. Myocardial infarction cannot be diagnosed in the presence of right bundle branch block
C. Myocardial infarction causes "convex upwards" ST elevation
D. Right ventricular infarction can be diagnosed using a standard 12 lead ECG
E. True posterior left ventricular infarction is characterised by pathological Q waves, tall R waves and inverted T waves in V1 and V_2

Q 54. **Which of the following is true in the digestive system?**

A. Fructose is mainly absorbed by simple diffusion
B. Glucose transport into the cell depends upon the active transport of sodium ions
C. Lactase activity increases during childhood
D. One molecule of sucrose forms two molecules of glucose
E. Polysaccharides are broken down mainly in the stomach

Q 55. **Which of the following is true regarding gastrin?**

A. Is predominantly produced by G cells located in the pancreas
B. Levels are decreased in atrophic gastritis (pernicious anaemia)
C. Stimulates gastric acid secretion in response to hunger
D. Stimulates insulin secretion particularly after a carbohydrate meal
E. Stimulates the growth of cells in the gastric mucosa

Q 56. **Which of the following is not correct regarding fovea centralis?**

A. Lies where the visual axis impinges on the retina
B. Is not crossed by any major blood vessels
C. Is the thickest part of the retina
D. Has higher visual acuity than other parts of the retina
E. Lies on the temporal side of the optic disc

Q 57. **Which of the following is true regarding the reduction in the neutrophil granulocyte count?**

A. Is not due to the suppression of bone marrow activity
B. A consequence of tissue damage
C. Associated with painful throat ulcers
D. Associated with widespread purulent infections
E. Caused by high levels of circulating glucocorticoids

47. E   48. E   49. A   50. B   51. C   52. E   53. C   54. B   55. E   56. C   57. C

Q 58. **Which of the following statements regarding urea is not correct?**

A. It has a molar concentration similar to that of glucose in the normal blood

B. Concentration rises in tubular fluid as the glomerular filtrate passes down the nephron

C. Is actively secreted by the renal tubular cells into the tubular fluid

D. Concentration in blood may rise after a high protein meal

E. Causes an osmotic diuresis when its blood concentration is increased

Q 59. **Which of the following is true regarding voluntary micturition?**

A. Depends on the integrity of a lumbar spinal reflex arc

B. Is not possible after sensory denervation of the bladder

C. Involves stimulation of the detrusor muscle in the bladder by autonomic sympathetic nerves

D. Is normally accompanied by some reflux of bladder contents into the ureters

E. Is stimulated during ejaculation

Q 60. **Which of the following statement regarding aldosterone is correct?**

A. Is a steroid hormone secreted by the adrenal medulla

B. Production ceases following removal of the kidneys and their juxtaglomerular cells

C. Production increases in treatment with drugs which block angiotensin-converting enzyme

D. Secretion of aldosterone results in decreased potassium secretion by the nephron

E. Secretion of aldosterone results in a fall in urinary pH

Q 61. **Which of the following is true regarding the fate of potassium in the renal nephron?**

A. Is actively secreted in the distal convoluted tubule

B. Is reabsorbed in the proximal convoluted tubule

C. Excretion is determined largely by potassium intake

D. Blood levels tend to rise in patients with acute renal failure taking a normal diet

E. All of the above

Q 62. **Which of the following does not occur in chronic renal failure?**

A. Glomerular filtration rate may fall by 70 percent before the condition gives rise to symptoms

B. The specific gravity of the urine tends to be elevated, e.g. about 1.030

C. Blood PCO_2 tends to be low

D. Ionised calcium levels in the blood tend to fall

E. Anaemia is common

Q 63. **A 30-year-old woman with three children complains of wetting herself during coughing and sneezing. What could be the likely bladder abnormality?**

A. Atonic bladder with overflow

B. Stress incontinence

C. Chronic prostatic obstruction

D. Acute retention of urine

E. Automatic bladder

Q 64. **Which of the following is true regarding the ventricular filling?**

A. Depends mainly on atrial contraction

B. Begins during isometric ventricular relaxation

C. Gives rise to a third heart sound in some healthy people

D. Can occur only when atrial pressure is greater than atmospheric pressure

E. Is most rapid in the second half of diastole

Q 65. **Which of the following is not correct regarding the veins?**

A. Contain most of the blood volume

B. Have a sympathetic vasoconstrictor innervation

C. Receive nutrition from vasa vasorum arising from their lumen

D. Respond to distension by contraction of their smooth muscle

E. Undergo smooth muscle hypertrophy when exposed to high pressure through an arteriovenous fistula

Q 66. **Which of the following statement is correct regarding the heart?**

A. The left atrial wall is about three times thicker than the right atrial wall

B. Systolic contraction normally begins in the left atrium

C. Excitation spreads directly from atrial muscle cells to ventricular muscle cells

D. Atrial and ventricular muscle contracts simultaneously in systole

E. The contracting ventricles shorten from apex to base

Q 67. **Which of the following is observed while measuring blood pressure using the auscultatory method**

A. The sounds that are heard are generated in the heart

B. The cuff pressure at which the first sounds are heard indicate systolic pressure

C. The cuff pressure at which the loudest sounds are heard indicate diastolic pressure

D. Systolic pressure estimations tend to be lower than those made by the palpatory method

E. Narrow cuffs are required for larger arms

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58. C   59. B   60. E   61. E   62. B   63. B   64. C   65 C   66. E   67. B

Q 68. Sympathetic drive to the heart is increased in which of the following conditions?
- A. In exercise
- B. In excitement
- C. In hypotension
- D. None of the above
- E. All of the above

Q 69. Which of the following regarding sinoatrial node cells is correct?
- A. Found in both atria
- B. Innervated by the vagus
- C. Unable to generate impulses when completely denervated
- D. Connected to the AV node by fine bundles of Purkinje tissue
- E. Able to generate impulses because their membrane potential is unstable

Q 70. Increased sympathetic drive to the heart increases which of the following?
- A. Rate of diastolic depolarisation in sinoatrial node cells
- B. Coronary blood flow
- C. Rate of conduction in Purkinje tissue
- D. Slope of the Frank-Starling (work versus stretch) curve of the heart
- E. All of the above

Q 71. Which of the following statement is not correct regarding the velocity of blood flow?
- A. In capillaries is low because the capillary bed has a large total cross-sectional area
- B. In veins is greater than in venules
- C. Can fall to zero in the ascending aorta during diastole
- D. Is greater towards the centre of large blood vessels than at the periphery
- E. In the circulation falls as the haematocrit falls

Q 72. Which of the following occurs during the isometric ventricular contraction?
- A. The entry and exit valves of the ventricle are closed
- B. Pressure in the aorta rises
- C. Pressure in the atria falls
- D. Left coronary blood flow rises
- E. The rate of rise in pressure is greater in the right than in the left ventricle

Q 73. Which of the following is not true regarding the Purkinje tissue cells in the heart?
- A. Conduct impulses faster than some neurones
- B. Are larger than ventricular myocardial cells
- C. Lead to contraction of the base before the apex of the heart
- D. Are responsible for the short duration of the QRS complex
- E. Are responsible for the configuration of the QRS complex

Q 74. Arterioles offer more resistance to flow than other vessels due to which of the following reasons?
- A. Thicker muscular walls
- B. Richer sympathetic innervation
- C. Smaller internal diameters
- D. A smaller total cross-sectional area
- E. A greater pressure drop along their length

Q 75. Which of the following is not correct regarding the jugular venous pulse?
- A. Pulse is not visible in normal healthy people
- B. Pulse has greater amplitude in patients with tricuspid incompetence
- C. Pulse can vary widely in amplitude in patients with complete heart block
- D. Pressure is raised in patients with right ventricular failure
- E. Pressure is commonly raised in patients with mediastinal tumours

Q 76. Narrowing of the lumen of major arteries supplying the leg is associated with
- A. Pain in the calf during exercise which is relieved by rest
- B. Growth of collateral vessels
- C. Delayed healing of cuts in leg skin
- D. Reduced arterial pulse amplitude at the ankle
- E. All of the above

Q 77. Which of the following is not true regarding vasovagal fainting or syncope?
- A. Causes loss of consciousness
- B. Is associated with bradycardia
- C. Is associated with skeletal muscle vasodilation
- D. Is more likely to occur when standing than when lying down
- E. Is more likely to occur in a cold than in a hot environment

Q 78. Pulmonary embolism (blood clots impacting in lung blood vessels) usually decreases which of the following?
- A. Pulmonary vascular resistance
- B. Left atrial pressure
- C. Right atrial pressure
- D. Ventilation to perfusion ratios in the affected lung
- E. PO_2 in pulmonary venous blood

Q 79. The electrocardiogram shows
- A. Regular QRS complexes in complete heart block
- B. High voltage R waves over the right ventricle in right ventricular hypertrophy
- C. An irregular saw-tooth appearance in ventricular fibrillation
- D. All of the above
- E. None of the above

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68. E   69. E   70. E   71. E   72. A   73. C   74. E   75. A   76. E   77. E   78. B   79. D

Q 80. What is the main site of iron absorption?
- A. Stomach
- B. Rectum
- C. Upper small intestine
- D. Lower small intestine
- E. Colon

Q 81. Which of the following statement is correct regarding bile?
- A. Contains enzymes required for the digestion of fat
- B. Contains unconjugated bilirubin
- C. Salts make cholesterol more water-soluble
- D. Pigments contain iron
- E. Becomes more alkaline during storage in the gall bladder

Q 82. Secretion of saliva increases in all the following situations except
- A. Touch receptors in the mouth are stimulated
- B. The mouth is flushed with acid fluids with a pH of about 4
- C. A subject thinks of unappetising food
- D. Vomiting is imminent
- E. Their sympathetic nerve supply is stimulated

Q 83. Which of the following statement is correct regarding the stomach?
- A. pH rarely falls below 4.0
- B. Pepsinogen is converted to pepsin by hydrochloric acid
- C. Ferrous iron is reduced to ferric iron by hydrochloric acid
- D. Acid secretion is inhibited by pentagastrin
- E. There is a rise in the bacterial count after histamine (H1) receptor blockade

Q 84. What is the main site of vitamin B_{12} absorption?
- A. Stomach
- B. Upper small intestine
- C. Lower small intestine
- D. Colon
- E. Rectum

Q 85. Gastric juice deficiency is likely to result in which of the following abnormality?
- A. Pale stools
- B. Local infections
- C. Steatorrhoea
- D. Milk intolerance
- E. Anaemia

Q 86. Deficiency of saliva is likely to result in which of the following abnormality?
- A. Pale stools
- B. Local infections
- C. Steatorrhoea
- D. Milk intolerance
- E. Anaemia

Q 87. Which of the following biochemical change typically occurs during pregnancy?
- A. Increase in plasma prolactin concentration in the first trimester
- B. Increase in plasma urea concentration in the second trimester
- C. An increase in haematocrit in early pregnancy
- D. An average reduction in tidal volume of 100 mL
- E. Average 15 litres increase in total body water

Q 88. Which of the following regarding thyroid function is true during normal pregnancy?
- A. Foetal thyroid function is largely dependent upon the function of the maternal thyroid
- B. Plasma thyroid-binding globulin concentration increases
- C. Plasma total thyroxine concentration falls
- D. Plasma TSH concentration increases
- E. Tri-iodothyronine is not able to cross the placenta to the foetus

Q 89. The concentrations of which of the following increase during pregnancy?
- A. Albumin
- B. Sodium
- C. Fibrinogen
- D. All of the above
- E. None of the above

Q 90. Which of the following is true regarding aortocaval compression in a pregnant woman?
- A. A reduction in cardiac output may be due to compression of the superior vena cava
- B. A wedge should be placed under the left side
- C. Compression of the aorta may cause uterine hypo-perfusion
- D. Is greater when lying on the right side
- E. Uterine contractions reduce the cardiovascular effects of aortocaval compression

Q 91. Which of the following is not a normal finding in a healthy pregnant patient?
- A. A fourth heart sound
- B. A raised alkaline phosphatase
- C. Tall, peaked T waves in lead III
- D. Left axis deviation on the electrocardiograph (ECG)
- E. Thrombocytopenia

Q 92. Which of the following is correct regarding the diffusion of gases through the placental membrane?
- A. CO_2 crosses the placenta from foetus to mother because of a high concentration gradient
- B. CO_2 diffuses through the placental membrane 5 times more quickly than O_2
- C. The mean PO_2 in the foetus is 50 mmHg
- D. The mean PO_2 in the mother's blood is approximately 30 mmHg
- E. The only way the foetus can excrete CO_2 is through the placenta

80. C 81. C 82. C 83. B 84. C 85. E 86. B 87. A 88. B 89. C 90. C. 91. C 92. E

Q 93. Regarding the human placenta, which of the following is true?

A. Cytotrophoblast is in direct contact with maternal blood
B. Decidual cells are derived from myometrial stromal cells
C. Each cotyledon represents a primary stem villi
D. The anchoring villi are attached to the myometrium
E. The intervillous space communicates directly with branches of the uterine arteries

Q 94. In a normal pregnancy, which of the following is true regarding uterine blood flow?

A. Is about 50 mL/minute at term
B. Is increased during uterine contractions
C. Is reduced by prostacyclin
D. Represents about 10% of the cardiac output by the end of the first trimester
E. Within the choriodecidual space is maintained throughout the cardiac cycle

Q 95. Which of the following is not true regarding trophoblast?

A. Develops from the blastocyst
B. Enters the maternal circulation during normal pregnancy
C. Gives rise to the foetal blood vessels in the placenta
D. Is genetically identical to decidua
E. Replaces endothelium of pregnant spiral arterioles

Q 96. Which of the following is true regarding human chorionic gonadotropin?

A. Binds to luteinising hormone (LH) receptors
B. Has intrinsic anti-thyroid activity
C. Is a protein molecule
D. Is synthesised by the corpus luteum of pregnancy
E. Secretion peaks at 20 weeks of gestation

Q 97. Which of the following normally decreases during pregnancy?

A. Heart rate
B. Stroke volume
C. Systemic vascular resistance
D. All of the above
E. None of the above

Q 98. In the pregnant patient, which of the following does not occur?

A. Constipation is common
B. Epidural pressures are increased
C. Liver blood flow is increased
D. Lower oesophageal sphincter pressure is reduced
E. Minimum alveolar concentrations of volatile agents are reduced

Q 99. Which of the following is not true regarding foetal pulmonary surfactant?

A. Can be detected in amniotic fluid
B. Contains more than 10% lipid
C. Contains phosphatidylglycerol
D. Is more than 40% albumin
E. Is predominantly dipalmitol phosphatidylcholine

Q 100. Which of the following does not occur in normal pregnancy?

A. An erythrocyte sedimentation rate remains within the non-pregnant range
B. Cardiac output increases
C. Glomerular filtration rate increases by up to 50%
D. Iron supplementation is only required if there is an evidence of anaemia
E. Ureters and renal pelvis dilate, but return to normal within 2 weeks of delivery

Q 101. Which of the following is true regarding weight gain in pregnancy?

A. There is an average weight gain of 20 kilograms throughout the pregnancy
B. Basal metabolic rate is increased by about 15% at term
C. Fat stores cause a weight increase on average of approximately 6 kilograms
D. Intracellular fluid volume increases by approximately 3 litres at term
E. The majority of the increase in fat occurs in the third trimester of the pregnancy

Q 102. During pregnancy, there is an increase in which of the following?

A. Fibrinolytic activity
B. Anti-thrombin III concentration
C. The total lung capacity
D. Plasma osmolality
E. The proportion of B to T lymphocytes

Q 103. Which of the following is true regarding gastrin?

A. Its fasting plasma concentrations are abnormally high in most patients after vagotomy for duodenal ulcer
B. Vasoactive intestinal polypeptide (VIP) inhibits its secretion
C. Protein in the duodenum inhibits its secretion
D. Stimulates secretion of pepsinogen more strongly than secretion of hydrogen ions
E. Is elaborated in "G-cells" that are confined to the antral region of the stomach

93. E   94. E   95. D   96. A   97. C   98. C   99. D   100. A   101. B   102. E   103. B

Q 104. Which of the following is not correct regarding the foetal blood?

A. During the 6th week of embryonic life, extramedullary haematopoiesis begins mainly in the liver and spleen
B. Most haemoglobin in the foetus is HbF chains in place of the adult haemoglobins HbA and HbA2
C. Foetal red blood cells are smaller than maternal blood cells
D. Foetal red blood cells are more resistant than adult cells to osmotic lysis by alkali and/or acid
E. At birth, the mean capillary haemoglobin level is 18 g/dL

Q 105. Which of the following does not occur in normal pregnancy?

A. Blood volume increases by about 30%
B. Increased maternal metabolic rate is mainly caused by the foetus and placenta
C. Maternal metabolic rate increases by about 15%
D. Red cell mass increases by about 20%
E. The key stages of organogenesis occur between 10 and 12 weeks

Q 106. Which of the following is not correct regarding acid-base balance in the body?

A. The normal pH of arterial blood is 7.4
B. Magnesium bicarbonate is a buffer
C. The placenta is permeable to hydrogen ions
D. In metabolic acidosis, there is an excess of fixed acids
E. Respiratory acidosis is associated with an increased level of CO_2

Q 107. The anti-phospholipid syndrome is associated with which of the following?

A. Left ventricular thrombus
B. Myocardial infarction
C. Pulmonary hypertension
D. Venous thrombosis
E. All of the above

Q 108. Patients with moderate to severe anaemia have a reduced level of which of the following parameters?

A. Cardiac output
B. Incidence of vascular bruits
C. 2,3-diphosphoglycerate blood level
D. Arterial PO_2
E. Capacity to raise oxygen consumption in exercise

Q 109. A patient with long-standing indigestion has noticed increasing lack of energy and tiredness when walking uphill. On questioning, he has noticed that the bowel motions are unusually dark from time to time. Due to the indigestion, the patient takes a bland diet without much meat or vegetables. On performing various blood investigations, anaemia

was diagnosed. What could be the likely cause of anaemia in this case?

A. Iron deficiency anaemia
B. Pernicious anaemia
C. Microcytic anaemia
D. Macrocytic anaemia
E. Normocytic anaemia

Q 110. Which of the following statements regarding blood groups and blood products is correct?

A. Patients with blood group O are universal recipients of blood
B. Stored blood becomes progressively more acidotic and hyperkalaemic with time
C. Stored blood contains a normal amount of clotting factors
D. Stored whole blood does not contain dextrose, phosphate and citrate
E. The ABO system is inherited in an autosomal recessive pattern

Q 111. Which of the following statement is correct regarding plasma bilirubin?

A. Is a steroid pigment
B. Is converted to biliverdin in the liver
C. Does not normally cross cerebral capillary walls
D. Is freely filtered in the renal glomerulus
E. Is insensitive to light

Q 112. A person with group A blood has which of the following?

A. Has anti-A antibody in the plasma
B. May have the genotype AB
C. May have a parent with group O blood
D. May have children with group A or group O blood only
E. Whose partner is also A can only have children of groups A or AB

Q 113. Hyponatraemia is not a recognised complication of which of the following?

A. Carbenoxolone therapy
B. Cerebral contusion
C. Congestive heart failure
D. Hepato-cellular failure
E. Major surgery

Q 114. Which of the following normally has a higher concentration intracellularly than extracellularly?

A. Adenosine triphosphate
B. Chloride
C. Magnesium
D. Sodium
E. Total phosphate

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104. C   105. E   106. C   107. E   108. E   109. A   110. B   111. C   112. C   113. A   114. C

Q 115. **Which of the following is used for the treatment of hyperkalaemia?**
- A. Amiloride
- B. Atenolol
- C. Calcium
- D. Magnesium
- E. Sodium chloride

Q 116. **Which of the following clinical conditions does not prolong neuromuscular blockade?**
- A. Hypercalcaemia
- B. Hypermagnesaemia
- C. Hypokalaemia
- D. Metabolic alkalosis
- E. Respiratory acidosis

Q 117. **Raised level of calcium in the blood (hypercalcaemia) is associated with which of the following?**
- A. May occur when parathyroid activity decreases
- B. May occur when the plasma protein level falls
- C. May occur in chronic renal failure
- D. Causes increased excitability of nerve and muscle
- E. Increases the risk of stone formation in the urinary tract

Q 118. **Which of the following is not a cause of metabolic acidosis?**
- A. Starvation
- B. Thiazide diuretics
- C. Uraemia
- D. Diabetes mellitus
- E. None of the above

Q 119. **A 71-year-old woman has presented with recent onset of vomiting. She vomits up the content of every meal approximately 2 hours after eating. On examination, she has a palpable mass to the right of the midline in the epigastrium.**

What is the most likely electrolyte disturbance in this case?
- A. High potassium, high sodium, high bicarbonate
- B. Low chloride, low sodium
- C. Low potassium, low chloride, high bicarbonate
- D. Low potassium, low sodium, low chloride, low bicarbonate

Q 120. **A reduced arterial PCO_2 is not associated with which of the following?**
- A. Occurs in normal pregnancy
- B. Occurs at altitudes over 2,500 metres
- C. Decreases cerebral blood flow
- D. Leads to a more alkaline urine
- E. Reduces blood pH

Q 121. **Which of the following is correct regarding shock?**
- A. Is associated with bradycardia
- B. Endotoxic shock can cause kidney damage only by direct damage to renal epithelium
- C. Tissue hypoxia leads to metabolic acidosis
- D. Hypovolaemic shock follows haemorrhage of 5% or more of blood volume
- E. The metabolic rate is increased

Q 122. **In a human being, haemorrhage does not result in which of the following?**
- A. A fall in cardiac output
- B. Decreased blood flow to the skin
- C. Increased aldosterone secretion
- D. Splenic contraction
- E. Venous constriction

Q 123. **In a normal adult woman weighing 75 kg, which of the following is not true regarding the extracellular fluid (ECF)?**
- A. Contains plasma protein
- B. Forms a lesser proportion of the total body weight in an obese than in a lean woman
- C. Has a sodium concentration of 135–145 mmol/L
- D. Has a total volume of 12–15 litres
- E. Is isotonic throughout the body

Q 124. **Which of the following statement regarding osmotic balance is true?**
- A. 0.9% sodium chloride solution contains no free water and is thus restricted to the extracellular compartment
- B. After intravenous administration of crystalloids, the distribution of these fluids throughout the body depends only on its osmotic activity
- C. In patients with pathological capillary leakage, the oncotic pressure becomes increasingly important in determining fluid fluxes
- D. The intracellular fluid volume is insensitive to changes in the sodium concentration of the extracellular fluid
- E. The microvascular endothelium separating interstitial fluid from the intravascular compartment is freely permeable to water

Q 125. **Which of the following is true in a healthy, young, non-pregnant woman at rest?**
- A. 80% of the total body weight is water
- B. 75% of extracellular fluid is outside the blood vessels
- C. Plasma volume is about 5 litres
- D. The pH of the plasma is about 7.25
- E. The plasma osmolality is about 400 mosmol/litre

115. C   116. A   117. E   118. B   119. C   120. E   121. C   122. D   123. E   124. C   125. B

Q 126. **Which of the following drugs causes hypokalaemia?**
- A. Amiloride
- B. Carbenoxolone
- C. Digoxin
- D. Spironolactone
- E. All of the above

Q 127. **Which of the following statements regarding high blood potassium level (hyperkalaemia) is not correct?**
- A. Occurs in acute renal failure
- B. Follows severe crush injuries to the limbs
- C. May diminish cardiac performance and cause death
- D. Increases skeletal muscle strength
- E. May be reduced by intravenous infusion of insulin and glucose

Q 128. **Which of the following is not associated with potassium depletion?**
- A. Can be detected by analysis of a biopsied sample of muscle
- B. Can result from loss of gastrointestinal secretions
- C. Causes reduced activity of intestinal smooth muscle
- D. Improves pre-existing acidosis
- E. Increases T wave amplitude in the electrocardiogram

Q 129. **Which of the following is not true regarding thirst?**
- A. Produced by a rise in plasma tonicity
- B. Produced by stimulation of certain areas in the hypothalamus
- C. Produced by a fall in blood volume
- D. Associated with decreased secretion of ADH
- E. Relieved by water intake before the water has been absorbed from the gut

Q 130. **Which of the following is not true regarding the deposition of excessive tissue fluid (oedema) in the legs?**
- A. Be associated with a raised extracellular fluid volume
- B. Result from hepatic disease
- C. Result from blockage of pelvic lymphatics
- D. Increase local interstitial fluid pressure
- E. Result from a high arterial blood pressure in the absence of heart failure

Q 131. **Which of the following facts does not hold true regarding total body water?**
- A. Can be measured with an indicator dilution technique using deuterium oxide
- B. Is smaller on average in women than in men
- C. Rises following injection of posterior pituitary extracts
- D. Falls during starvation
- E. Is less than 80 percent in young adults

Q 132. **Which of the following substances is not associated with an increased capillary permeability in cases of acute inflammation?**
- A. Angiotensin
- B. Bradykinin
- C. Histamine
- D. Prostacycline
- E. Serotonin

Q 133. **Which of the following is not true regarding the normal menstrual cycle**
- A. Blood loss during menstruation averages around 30 mL
- B. The proliferative phase depends on oestrogen secretion
- C. Cervical mucus becomes more fluid around the time of ovulation
- D. Ovulation is followed by a surge in blood luteinising hormone level
- E. Basal body temperature is higher after ovulation

Q 134. **Which of the following is not true regarding the normal fertilization of the human ovum?**
- A. Occurs in the uterus
- B. Prevents further spermatozoa from entering the ovum
- C. Occurs 2–5 days after ovulation
- D. Occurs 5–7 days before implantation
- E. Leads to the secretion of human chorionic gonadotropin within 2 weeks

Q 135. **Which of the following is not true regarding the secretion of testosterone?**
- A. Depresses pituitary secretion of LH
- B. Causes the epiphyses of long bones to unite
- C. May lead to a negative nitrogen balance
- D. Causes the scalp hair to recede
- E. Stimulates growth of body hair

Q 136. **Which of the following is true regarding the human chorionic gonadotropic hormone?**
- A. It is a steroid
- B. Acts directly on the uterus to maintain the endometrium
- C. It is formed in the anterior pituitary
- D. Blood level rises steadily throughout pregnancy
- E. Can be detected in the urine as an early sign of pregnancy

Q 137. **Which of the following changes occur during pregnancy?**
- A. Uterine muscle enlarges due mainly to cell proliferation
- B. Uterus is quiescent until the onset of labour
- C. Breasts enlarge due mainly to the action of prolactin
- D. Haematocrit rises
- E. Basal metabolic rate rises by more than 15%

126. B   127. D   128. E   129. D   130. E   131. D   132. A   133. D   134. A   135. C   136. E   137. E

Q 138. **Females differ from males in which of the following aspects?**
- A. Pituitary gland secretes same gonadotropic hormones in both the sexes
- B. Hypothalamus shows different patterns of hormone secretion
- C. Gonads do not produce gametes until later in life
- D. Blood gonadotropin levels rise in later life in both the sexes
- E. Polymorphs show "drumsticks" of chromatin on their nuclei

Q 139. **Which of the following is true regarding foetal haemoglobin?**
- A. It is the only type identifiable in foetal blood
- B. Forms the bulk of total haemoglobin for the first year of life
- C. Has a higher oxygen-carrying capacity than adult haemoglobin
- D. Binds 2,3-DPG more avidly than does adult haemoglobin
- E. Has a higher affinity than the adult form for oxygen at low PO_2

Q 140. **Which of the following is not correct regarding the mammary glands?**
- A. Milk formation is stimulated by oestrogen and progesterone
- B. Milk formation can be depressed by hypothalamic activity
- C. Maintenance of lactation depends on suckling
- D. Lactation ceases if the anterior pituitary gland is destroyed
- E. Milk ejection ceases if the posterior pituitary gland is destroyed

Q 141. **Which of the following is true regarding the placenta?**
- A. Foetal and maternal blood mix freely in the sinusoids
- B. The PO_2 in sinusoidal blood is similar to that in maternal arterial blood
- C. The barrier to oxygen diffusion is much less than that in alveoli
- D. Foetal blood in umbilical veins has a PO_2 within 10 percent of that in maternal sinusoids
- E. Foetal blood becomes more than 50 percent saturated with oxygen

Q 142. **Which of the following is not true regarding the male post-pubertal state in comparison to the pre-pubertal state?**
- A. The gonads are responsive to gonadotrophic hormones
- B. There is a greater output of l7-ketosteroids in the urine

- C. Skeletal muscle is stronger per unit mass of tissue
- D. The circulating level of follicle-stimulating hormone is higher
- E. Hypothalamic output of gonadotropin-releasing factors is greater

Q 143. **Which of the following is not true regarding the oxygen content of the blood in the case of foetal circulation?**
- A. Femoral artery is less than that in the brachial artery
- B. Superior vena cava is higher than that in the inferior vena cava
- C. Left ventricle is higher than that in the right ventricle
- D. Pulmonary artery is higher than that in the pulmonary veins
- E. Cerebral arteries is lower than in the maternal cerebral arteries

Q 144. **Which of the following is true regarding the interstitial cells of the testis?**
- A. Contribute to the volume of seminal fluid
- B. Are the source of the hormone inhibin
- C. Are stimulated to secrete by luteinizing hormone (LH)
- D. Is not dependent on hypothalamic activity to function properly
- E. Are non-functional unless the testis descends from the abdomen to the scrotum

Q 145. **The size of the foetus at birth is likely to be smaller in which of the following?**
- A. Small than in large mothers
- B. Smoking than in non-smoking mothers
- C. Female than in male babies
- D. Firstborn than in subsequent babies
- E. All of the above

Q 146. **Which of the following is not true regarding testosterone secretion from the testis?**
- A. Increases at puberty because LH levels increase
- B. Is responsible for the growth of facial hair
- C. Has a negative feedback effect on FSH secretion by the anterior pituitary gland
- D. Peaks at the time of awakening
- E. Is responsible for interest in the opposite sex

Q 147. **Which of the following is true regarding the normal seminal ejaculate?**
- A. Has a volume of about 2–5 mL
- B. Contains fructose from the seminal vesicles
- C. Contains phosphate and bicarbonate buffers
- D. Contains prostaglandins
- E. All of the above

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138. E   139. E   140. A   141. E   142. A   143. B   144. C   145. E   146. C   147. E

Q 148. Which of the following is not true regarding the newborn baby in comparison to the adult?

A. Its ability to concentrate urine is poor
B. Its ability to dilute urine is also poor
C. Blood–brain barrier is more permeable to unconjugated bilirubin
D. Temperature regulation is more efficient because of brown fat
E. Blood has a greater affinity for oxygen at low oxygen pressures

Q 149. Which of the following is not true regarding the amniotic fluid?

A. Formed in early pregnancy by filtration from foetal skin capillaries
B. Formed in late pregnancy by filtration from the gut mucosa
C. Swallowed by the foetus
D. Similar in electrolyte composition to plasma
E. Inhaled and exhaled by the foetus

Q 150. Cessation of menstruation (secondary amenorrhoea) may occur due to which of the following?

A. Psychological stress and/or severe weight loss
B. Continuous administration of oestrogens
C. An adrenal tumour
D. Continuous administration of gonadotropin-releasing hormone (GnRH)
E. All the above

Q 151. Which of the following is not true regarding a normal foetus?

A. Gains more weight in the last ten weeks of gestation than in the first 30 weeks
B. Has a higher haemoglobin level at term than a normal adult
C. Stores sufficient iron in the liver to last a year after birth
D. Has a metabolic rate (per metre square) twice as that of an adult due to rapid growth
E. Passage of meconium before birth is a sign of distress

Q 152. Development of secondary sexual characteristics before the age of nine could be due to which of the following?

A. Due to abnormal secretion of adrenal cortical hormones
B. Associated with short stature
C. Due to a hypothalamic tumour
D. Present in a normal healthy child
E. All the above

Q 153. Which of the following statement regarding the foetus is not correct?

A. Blood in umbilical veins contains more amino acid than maternal blood in uterine veins
B. Aorta has a higher rate of blood flow than the distal pulmonary artery
C. Aortic blood pressure is lower than the pulmonary arterial pressure
D. Systemic resistance is lower than pulmonary resistance
E. Heart rate suggests foetal distress if it exceeds 100 beats/minute

Q 154. Lack of pulmonary surfactant is associated with which of the following?

A. Is unlikely in infants born after 30 weeks gestation
B. Can be diagnosed by examining the foetal amniotic fluid
C. Increases the effort required for expiration
D. Decreases the surface tension forces in the lungs
E. Leads to poor oxygenation of the blood before birth

Q 155. Which of the following statement regarding the neonatal development is correct?

A. Liver stores sufficient vitamin K for the first few months of life
B. Blood volume is closer to 750 than 250 mL
C. Blood glucose level does not fluctuates much in comparison to the foetal level
D. Gut usually lacks certain enzymes needed for digestion of milk
E. Peripheral vascular resistance is higher than that of the adult

Q 156. Which of the following is not correct regarding a newborn child?

A. Its haemoglobin level falls from around 170–200 g/litre to around 110 g/litre during the first year
B. There should be a delay in clamping the umbilical cord so that blood from the placenta can drain into the foetus
C. It should increase its weight by 10 percent at four months
D. Its brain can tolerate a lower blood glucose level than that of an adult
E. Its brain can tolerate a lower oxygen level than that of an adult

Q 157. Removal of the testes in the adult causes which of the following?

A. A rise in the pitch of the voice
B. Loss of the ability to copulate
C. Hot flushes, irritability and depression
D. A fall in the blood levels of LH and FSH
E. All the above

Q 158. Infertility usually occurs due to which of the following?

A. When the sperm count is reduced to 10 percent of normal
B. When posterior pituitary function is lost
C. When one uterine tube is blocked
D. Due to a reproductive disorder in the female partner
E. All of the above

148. D 149. B 150. E 151. C 152. E 153. E 154. B 155. E 156. C 157. C 158. A

Q 159. Women having their first child after the age of 35 are associated with an increased risk of which of the following?

A. Average blood loss than younger women
B. Incidence of ineffective uterine contractions during labour
C. Incidence of foetal abnormalities
D. Risk of spontaneous abortion
E. All of the above

Q 160. Infertility in the male can be explained by all the below-mentioned observations, except which of the following?

A. There are no motile sperms in semen 15 minutes after ejaculation
B. 50 percent of the sperms in the semen are abnormal
C. The sperm count is 10^8/mL
D. The sperm count is below 10–20 percent to cause infertility
E. There is widespread autonomic neuropathy

Q 161. Pregnant women with five or more previous deliveries have a greater risk of having which of the following disorders?

A. An unfavourable presentation of the baby in the pelvis
B. Complications due to rhesus incompatibility
C. Serious loss of blood after delivery
D. Involuntary urination while coughing (stress incontinence)
E. All the above

Q 162. Which of the following is not true regarding the secretion of androgens in the adult female?

A. Adrenal androgen secretion is normal
B. In large amounts can cause enlargement of the clitoris
C. Does not affect the voice
D. May lead to growth of facial hair
E. May result in amenorrhoea

Q 163. Foetal death is likely to result from serious impairment of which of the following?

A. Liver function
B. Alimentary tract function such as obstruction
C. Renal function
D. Cerebral function
E. Cardiac function

Q 164. Which of the following is not true regarding human chorionic gonadotropin?

A. Binds to luteinising hormone (LH) receptors
B. Has intrinsic anti-thyroid activity
C. Is a glycoprotein
D. Is synthesised by the by the outer layer of the blastocyst and placenta
E. Secretion peaks at 11-12 weeks gestation

Q 165. Which of the following statement is true regarding human chorionic gonadotropin?

A. Has a molecular weight of approximately 130,000
B. Has a similar molecular structure to LH secreted by the posterior pituitary
C. Stimulates the corpus luteum
D. Reaches a peak level at 16–20 weeks post-ovulation and conception
E. Is steroid in nature

Q 166. The corpus luteum of pregnancy does not produce which of the following?

A. 17 Alpha-hydroxyprogesterone
B. Human chorionic gonadotropin
C. Oestradiol
D. Progesterone
E. Relaxin

Q 167. Which of the following is not true concerning transfer of various substances across the placenta?

A. Aminoacids are actively transported
B. Calcium is transferred by passive diffusion
C. Glucose is transferred by facilitated diffusion
D. Immunoglobulin IgG is transferred by endocytosis
E. Oxygen is transferred by flow-limited passive diffusion

Q 168. Which of the following hormones is produced by the placenta?

A. Heat stable alkaline phosphatase (HSAP)
B. Human placental lactogen
C. Pregnancy-associated plasma protein A (PAPPA)
D. Schwangerschafts protein (SPI)
E. None of the above

Q 169. Which of the following statement is true regarding the pancreatic secretion?

A. In response to vagal stimulation is copious, rich in bicarbonate but poor in enzymes
B. In response to acid in the duodenum is scanty but rich in enzymes
C. In response to secretin secretion is low in bicarbonate
D. Contains enzymes that digest neutral fat to glycerol and fatty acids
E. Contains enzymes that convert disaccharides to monosaccharides

Q 170. The liver is the principal site for the synthesis of which of the following?

A. Synthesis of plasma albumin
B. Synthesis of plasma globulins
C. Synthesis of vitamin B12
D. Synthesis of vitamin C
E. None of the above

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159. E   160. C   161. E   162. C   163. E   164. B   165. C   166. B   167. B   168. B   169. D   170. A

Q 171. Which of the following statement is not true regarding the secretion of gastric juice?

A. Is secreted when the vagus nerves are stimulated
B. Is secreted in vagotomised animals when food is chewed but not swallowed
C. Inactivates the digestive enzymes secreted with saliva
D. Does not digest the gastric mucosa because it is protected by a coat of mucus impregnated with bicarbonate
E. Irritates the oesophageal mucosa if regurgitated from the stomach

Q 172. Which of the following is correct regarding the stomach?

A. Is responsible for absorbing about 10 percent of the ingested food
B. Contains mucosal cells containing low concentrations of carbonic anhydrase
C. Peristaltic contractions start from the pyloric region
D. Motility increases when fat enters the duodenum
E. Relaxes when food is ingested so that there is little rise in intra-gastric pressure

Q 173. The risk of developing gallstones increases in which of the following situations?

A. When cholesterol micelles are formed in the gall bladder
B. As the bile salt: cholesterol ratio increases
C. As the lecithin: cholesterol ratio increases
D. When supplementary bile salts are taken by mouth
E. In patients with haemolytic anaemia

Q 174. Which of the following is not a function of the cells of the liver?

A. Help to maintain the normal blood glucose level
B. Deaminate amino acids to form NH_4 ions
C. Synthesize 1,25-dihydroxycholecalciferol Vitamin D_3
D. Manufacture most of the plasma proteins
E. Inactivate steroid hormones manufactured in the gonads

Q 175. Which of the following statements regarding brown fat is correct?

A. Relatively more abundant in adults than in infants
B. Contains lesser amount of mitochondria than ordinary fat
C. Is less vascular than ordinary fat
D. Stimulated to generate more heat when its parasympathetic nerve supply is stimulated
E. Is more important than shivering in neonatal thermoregulation

Q 176. The passage of gastric contents to the duodenum does not cause which of the following?

A. Copious secretion of pancreatic juice rich in bicarbonate
B. Decreased gastric motility
C. Contraction of the gall bladder
D. Contraction of the sphincter of Oddi
E. Release of pancreozymin

Q 177. Which of the following is correct regarding the normally innervated stomach?

A. Is stimulated to secrete gastric juice when food is chewed, even if it is not swallowed
B. Cannot secrete HCl when its H1 histamine receptors are blocked
C. The denervated stomach cannot secrete gastric juice after a meal is ingested
D. Empties less quickly than the denervated stomach
E. Is stimulated to secrete gastric juice by the hormone secretin

Q 178. Which of the following statement regarding cholesterol is not correct?

A. Can be absorbed from the gut by intestinal lymphatics following its incorporation into chylomicrons
B. Can be synthesized in the liver
C. In the diet comes mainly from vegetable sources
D. Is eliminated from the body mainly by excretion in the bile
E. Is a precursor of adrenal cortical hormones

Q 179. Which of the following is not true regarding the impaired intestinal absorption of the various substances?

A. Iron occurs frequently following removal of most of the stomach
B. Iodide leads to an increase in size of the thyroid gland
C. Water occurs in infants who cannot digest lactose
D. Calcium may occur following removal of the terminal ileum
E. Bile salts may occur following removal of the terminal ileum

Q 180. Which of the following is not true regarding the estimation of metabolic rate?

A. It is equal to the total heat production
B. It is related to the calorific value of the food consumed in the previous 24 hours
C. It is based on the oxygen consumption provided the type of food being metabolized is known
D. It is based on the oxygen consumption and the respiratory quotient
E. Metabolic rate is proportional to carbon dioxide production and the respiratory quotient

Q 181. Which of the following is not correct regarding the symptoms of intestinal obstruction?

A. Constipation
B. Crampy pain due to intermittent vigorous peristalsis
C. Distension due to fluid and gas proximal to the obstruction
D. Hypotension
E. Vomiting which is more severe with low than with high bowel obstruction

171. B   172. E   173. E   174. C   175. E   176. D   177. A   178. C   179. D   180. B   181. E

Q 182. Auscultation of the heart can provide evidence of which of the following?

A. The direction of turbulent flow causing a murmur
B. Aortic stenosis, if there is a loud pre-systolic murmur in the aortic valve area
C. Mitral incompetence, if a diastolic murmur is heard in the axilla
D. Ventricular septal defect, if a loud diastolic murmur is heard
E. Mitral stenosis, if an early systolic murmur is heard

Q 183. Severe diarrhoea causes a decrease in all of the following except?

A. Body potassium
B. Body sodium
C. Extracellular fluid volume
D. Total peripheral resistance
E. Blood pH

Q 184. Lack of pancreatic juice in the duodenum may lead to which of the following?

A. The presence of undigested meat fibres in the stools
B. An increase in the fat content of the faeces
C. Faeces with a low specific gravity
D. A reduced prothrombin level in blood
E. All the above

Q 185. Which of the following is not true regarding portal hypertension?

A. The total vascular resistance of the hepatic sinusoids is increased
B. Portal blood flow through the liver is increased
C. The volume of fluid in the peritoneal cavity increases
D. A porto-caval shunt (anastomosis between portal vein and inferior vena cava) can decrease the tendency to bleed into the alimentary tract
E. A porto-caval shunt increases the risk of coma after bleeding into the alimentary tract

Q 186. Which of the following is not correct regarding absorption of glucose by intestinal mucosal cells?

A. Relies on a carrier mechanism in the cell membrane
B. Is blocked by the same agents that block renal reabsorption of glucose
C. Is impaired by blockade of active sodium transport in the cells
D. Involves the same carriers that are used for the absorption of galactose
E. Takes place mainly in the ileum

Q 187. Which of the following statement is not correct regarding vomiting?

A. Is caused by forced expiratory efforts by skeletal muscles in the presence of a closed glottis and pylorus, which are responsible for compressing the stomach
B. Is coordinated by a vomiting centre in the medulla oblongata
C. Of green fluid suggests that duodenal contents have regurgitated into the stomach
D. May be accompanied by a fall in arterial blood pressure
E. Is more marked in low intestinal obstruction than in high intestinal obstruction

Q 188. Which of the following is not correct regarding obesity?

A. If food energy intake exceeds energy expenditure, obesity develops regardless of the predominant food in the diet
B. Is associated with increased demands on pancreatic islet beta cells
C. Can be assessed by multiple measurements of skin fold thickness
D. Can be assessed by weighing the body in air and in water
E. Is not diagnosed until body weight is 40 percent above normal

Q 189. Which of the following is not true regarding muscle tone in the lower oesophagus?

A. Greater than tone in the middle oesophagus
B. A major factor in preventing heartburn
C. Increased in pregnancy
D. Increased by gastrin
E. Reduced by anticholinergic drugs

Q 190. Which of the following is correct regarding the neuromuscular junctions of skeletal muscles?

A. The motor end plate is the motor nerve terminal
B. Spontaneous (miniature) potentials may be recorded in the motor nerve terminal
C. Motor nerve terminals have vesicles containing acetylcholine
D. There is a low concentration of acetylcholinesterase
E. Transmission is facilitated by botulinum toxin

Q 191. Which of the following is true regarding the sensory receptors?

A. Stimulus energy is converted into local depolarization
B. The generator potential is graded and self-propagating
C. A generator potential can be produced by only one form of energy
D. The frequency of action potentials generated doubles when the strength of the stimulus doubles
E. Serving touch sensation, constant supra-threshold stimulation causes action potentials to be generated at a constant rate

Q 192. Which of the following statements regarding a somatic lower motor neurone is correct?

A. Innervates fewer fibres in an eye muscle than does one innervating a leg muscle
B. Conducts impulses at a speed similar to that in an autonomic postganglionic neurone
C. Is unmyelinated
D. Conducts impulses which cause relaxation in some skeletal muscles
E. Synapse with skeletal muscle but not with other neurones

182. A   183. D   184 E   185. B   186. E   187. E   188. E   189. C   190. C   191. A   192. A

Q 193. Which of the following is true regarding transmission of nerve impulses?

A. Can travel in one direction only in a nerve fibre
B. Can travel in one direction only across a synapse
C. Travel at the speed of an electric current
D. Do not correspond in duration to that of the nerve refractory period
E. Can be transmitted at higher frequencies in autonomic than in somatic nerves

Q 194. Which of the following is true regarding the contraction of skeletal muscles?

A. Contraction occurs when its pacemaker cells depolarize sufficiently to reach the threshold for firing
B. Calcium is taken up by the sarcotubular system when it contracts
C. Actin and myosin filaments lengthen during the contraction of skeletal muscles
D. The sarcomeres lengthen during contraction
E. Contraction strength is related to initial length of the muscle fibres

Q 195. Which of the following statements is not correct regarding saltatory conduction of the nerve fibres?

A. Occurs only in myelinated fibres
B. Does not depend on depolarization of the nerve membrane
C. Has a slower velocity in cold than in warm conditions
D. Transmits impulses with a velocity proportional to fibre diameter
E. None of the above

Q 196. Intracranial pressure does not rise in which of the following situation?

A. Cerebral venous pressure rises
B. Forced expiration is made against a closed glottis
C. There is a bout of coughing
D. Cerebral blood flow increases
E. Arterial PCO_2 falls below normal

Q 197. Changes in maternal physiology during pregnancy include which of the following?

A. Increased nitrogen retention
B. Mean arterial pressure of around 20 mmHg
C. Increased arterial PCO_2
D. Increased tone in the urinary tract
E. Increased renal threshold for glucose

Q 198. Increased intracranial pressure may cause which of the following?

A. Cranial enlargement in children
B. Squinting and loss of smell sensation in children
C. Cupping of the optic disc
D. Severe reduction in cerebral blood flow
E. All the above

Q 199. Pain receptors in the gut and urinary tract may not be stimulated by which of the following?

A. Cutting through their wall with a sharp scalpel
B. Distension
C. Inflammation of the wall
D. Acid fluid
E. Vigorous rhythmic contractions behind an obstruction

Q 200. Which of the following statement is not true regarding the pain receptors?

A. They are bare nerve endings
B. Stimulated by a rise in the local K^+ concentration
C. Quick to adapt to a constant stimulus
D. More easily stimulated in injured tissue
E. Damage to the gut wall by stimuli such as cutting or burning is painless

Q 201. Rapid eye movement (REM) sleep differs from non-REM sleep regarding which of the following statements?

A. The EEG shows waves of lower frequency
B. Muscle tone is higher
C. Heart rate and respiration are more regular
D. Secretion of growth hormone is increased
E. Dreaming is more common

Q 202. Muscle tone is increased by which of the following?

A. Curare-like drugs
B. Lower motor neurone lesions
C. Cerebellar lesions
D. Gamma efferent impulses to muscle spindles
E. None of the above

Q 203. Which of the following is not a feature of Parkinsonism?

A. Tremor which is more obvious when the patient is performing skilled movements
B. Muscle paralysis
C. Increased muscle tone throughout the range of passive movement
D. mask-like facies; there is poverty of facial movements
E. An unusual gait with small fast regular steps

Q 204. Which of the following is a feature of lower motor neuron disease?

A. Causes loss of voluntary movements but not of reflex movements
B. Is a later stage of upper motor neurone disease
C. Causes eventual hypertrophy of the muscles concerned
D. Does not affect ventilation of the lungs
E. Is associated with involuntary twitching of small fasciculi in the affected muscles

Q 205. Bulging of the optic disc into the vitreous humour (papilledema) is associated with which of the following?

A. A rise in intracranial pressure
B. Inflammation of the optic nerve (optic neuritis)
C. Interference with the venous drainage of the eye
D. All the above
E. None of the above

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193. B   194. E   195. B   196. E   197. D   198. E   199. A   200. C   201. E   202. E   203. B   204. E   205. D

Q 206. **Which of the following is true regarding hemiplegia following a right-sided cerebrovascular accident (stroke)?**
A. Left-sided muscle weakness is evident
B. Muscles in the left side of the body are unable to contract
C. Muscles which act on both sides of the body, such as respiratory muscles, are also not spared
D. Skilled movements are better preserved than unskilled movements
E. Speech movements are better preserved than swallowing movements

Q 207. **Which of the following is not true regarding cutaneous pain?**
A. Can be caused by excitation of specific pain receptors
B. Can be caused by excitation of receptors by chemicals released in injured tissue
C. Can be elicited more readily if the tissue has been injured recently
D. Receptors adapt to stimulation much slowly than the touch receptors
E. Transmission at spinal cord level is facilitated by opening of potassium channels in the post-synaptic membrane

Q 208. **Loss of pain sensation does not occur in which of the following situations?**
A. Feet may lead to skin ulceration
B. Knee may lead to joint damage
C. Ears and fingers usually precedes frostbite
D. Leg follows surgical division of the spinothalamic tracts in the spinal cord
E. Face follows surgical division of the facial nerve

Q 209. **Which of the following does not occur in response to severe pain?**
A. A fall in blood pressure due to a fall in vascular resistance in skeletal muscle
B. A fall in heart rate due to an increase in cardiac vagal tone
C. Vomiting through a reflex centre in the brainstem
D. Profuse sweating due to activation of sympathetic nerves
E. Suppression of cortisol secretion

Q 210. **Which of the following statements is not correct regarding endolymph?**
A. Is found within the membranous labyrinth
B. Has a potassium concentration close to that of intracellular fluid
C. Bathes the hair cells of the inner ear
D. Is electrically negative with respect to perilymph
E. Inertia is a factor in the stimulation of receptors in the semi-circular canals during rotatory acceleration

Q 211. **Which of the following is true regarding the olfactory cells?**
A. Are epithelial cells which synapse with olfactory nerves
B. Generate impulses when stimulated which are relayed in the thalamus
C. Are chemoreceptors
D. Show little adaptation
E. Are of little importance in appreciating the flavour of food

Q 212. **The cones in the retina differ from rods in that they are more**
A. Less numerous than rods
B. Concerned with colour vision
C. Less sensitive to light than rods
D. Concerned with high visual acuity
E. All the above

Q 213. **Which of the following statements regarding the olfactory system is not correct?**
A. Can differentiate between 2,000 and 4,000 different odours
B. Can detect differences in odour between isomers of the same substance
C. Can perceive the direction from which an odour comes.
D. Can detect small differences in the concentration of the substance responsible for the odour
E. Sensation of odour is better in young people than in old people

Q 214. **Which of the following is true regarding the tympanic membrane?**
A. Modifies the frequencies of sound waves impinging on the ear
B. Stops vibrating almost immediately after the sound stops
C. Bulges outwards when the pharyngotympanic tube is blocked
D. Transmits sound more effectively when the small muscles of the middle ear are contracted
E. Cannot transmit sound waves if it is perforated

Q 215. **The hair cells in the semi-circular canals are stimulated by which of the following?**
A. Movement of perilymph
B. Linear acceleration
C. Rotation at constant velocity
D. Gravity
E. Movement of endolymph relative to hair cells

206. A   207. E   208. E   209. E   210. D   211. C   212. E   213. D   214. B   215. E

Q 216. **An 80-year-old woman undergoes total abdominal hysterectomy. She has chronic obstructive airway disease. Post-operatively, she has a difficulty in extubation and requires a prolonged stay in the recovery unit. Which of the following is likely to be the most important factor for stimulating respiration?**
A. Decreased arterial pH
B. Reduced arterial pO_2
C. Reduced arterial pCO_2
D. Increased concentration of the H^+ ions in the cerebrospinal fluid
E. Increased pCO_2 of the CSF

Q 217. **Which of the following is true regarding light travelling from an object to the right of the visual axis?**
A. Impinges on the retina in the right eye to the right of the fovea
B. Impinges on the retina in the left eye to the left of the fovea
C. Generates impulses which travel in the right optic tract
D. Generates impulses which produce conscious sensation in the frontal lobe eye fields
E. Forms an erect image on the retina

Q 218. **Which of the following statements is true regarding the receptor cells serving taste?**
A. Are confined to the tongue
B. Are stimulated when chemicals diffuse through the overlying epithelium to reach them
C. For sweetness are more common at the tip than at the back of the tongue
D. Are primary sensory neurones
E. Are histologically different for the four primary taste modalities

Q 219. **Which of the following statements regarding an audiogram is not correct?**
A. It is a plot of hearing loss (or hearing ability) against sound frequency
B. Showing equal impairment of air and bone conduction suggests conductive deafness
C. Showing hearing loss at low frequencies for air conduction suggests ear drum damage
D. Showing loss at 8,000 Hz for air and bone conduction suggests basal cochlear damage
E. Showing hearing loss at the lower frequencies is common in elderly people

Q 220. **Which of the following is true in someone with short-sightedness (myopia)?**
A. The eye tends to be longer than average from lens to retina
B. A convex lens is required to correct the refractive error
C. Close vision is affected more than distance vision
D. The near-point is farther than normal from the eye
E. A circular object tends to appear oval

Q 221. **Which of the following is true regarding colour blindness?**
A. Results from inability to detect one of the three primary light colours, red, yellow and blue
B. Where red and green are indistinguishable is due to failure of red and green cone systems
C. In which no colours can be detected is due to failure of all the cones systems
D. Is more common in women than men
E. Is a disability linked to the Y-chromosome

Q 222. **Which of the following is not a cause of squinting (strabismus)?**
A. Poor vision in one eye in childhood
B. A refractive error in childhood
C. Central suppression of vision in one eye in childhood
D. Damage to the internal capsule
E. None of the above

Q 223. **Which of the following does not cause an impairment of the sense of smell?**
A. May occur in hydrocephalus
B. Is likely after thalamic damage
C. Can be caused by inflammation of the nasal mucosa
D. Is a recognized effect of frontal lobe tumour
E. None of the above

Q 224. **Which of the following is not correct regarding the involuntary oscillatory eye movements (nystagmus)?**
A. Can occur in normal healthy people
B. May result from the diseases of the semi-circular canal
C. Occur in cerebellar disease
D. Occur when cold fluid is run into one external ear canal
E. Do not affect acuity of vision

216. D   217. B   218. C   219. B   220. A   221. A   222. C   223. B   224. E

Biochemistry and Nutrition

Biochemistry

Structure and Function of Normal Cell

Introduction

Each cell **(Fig. 4.1)** is composed of a cell body (having a nucleus and the cytoplasm surrounding the nucleus), and a cell membrane covering the cell body.

Cell Membrane

Cell membrane is a protective sheath, a semipermeable membrane enveloping the cell body. It is also known as plasma membrane or plasmalemma. This membrane separates the fluid outside the cell called extracellular fluid (ECF) from the fluid inside the cell called intracellular fluid (ICF). The cell membrane mainly controls the intracellular electrolyte and biochemical environment
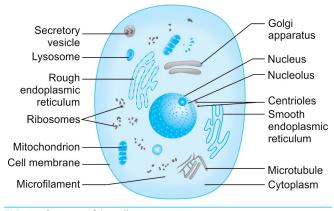
FIG. 4.1: Structure of the cell

by maintaining the active transport mechanisms. It also provides adhesion between individual cells and bears the individual's major human leucocyte antigens (HLA). In some cells, e.g. neutrophils, it helps in determining motility and phagocytosis. Thickness of the cell membrane varies from 75Å to 111Å.

Cytoplasm

Cytoplasm of the cell is the jelly-like material enclosed by a trilaminar cell membrane having a complex biochemical structure, composed of several proteins and lipids. Nearly 80% of the cytoplasm is formed by water. Cytoplasm also contains many organelles, each having individual structure and function.

Cytoplasm has two zones: (1) ectoplasm (peripheral part of cytoplasm, situated just beneath the cell membrane) and (2) endoplasm (inner part of cytoplasm, placed between the ectoplasm and the nucleus).

There are several cellular structures inserted in the cytoplasm known as the cytoplasmic organelles. Some organelles are bound by limiting membrane, whereas others do not have any limiting membrane **(Table 4.1)**, with each organelle having a definite structure and function **(Table 4.2)**.

Cytoplasmic Organelles with a Limiting Membrane

Endoplasmic Reticulum

Endoplasmic reticulum (ER) is a network of tubular and microsomal vesicular structures (cisternae) which are interconnected with one another **(Fig. 4.2)**. It is a membrane-bound organelle whose lumen contains a fluid medium called endoplasmic matrix. The ER forms a link between nucleus and cell membrane by forming a connection between the cell membrane and the nuclear

TABLE 4.1	Cytoplasmic organelles with and without a limiting membrane

Organelles with limiting membrane

- Nucleus
- Endoplasmic reticulum
- Mitochondria
- Golgi apparatus
- Lysosome
- Peroxisome
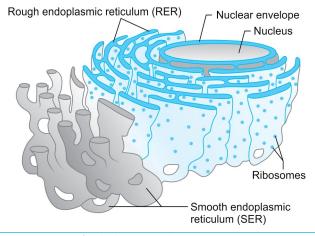- Secretory vesicles
- Centrosome and centrioles

Organelles without limiting membrane

- Ribosomes
- Cytoskeleton

TABLE 4.2	Functions of cytoplasmic organelles

Organelles	Functions
Rough endoplasmic reticulum	• Synthesis of proteins • Degradation of worn-out organelles
Smooth endoplasmic reticulum	• Synthesis of lipids and steroids • Role in cellular metabolism • Storage and metabolism of calcium • Catabolism and detoxification of toxic substances
Golgi apparatus	• Processing, packaging, labelling and delivery of proteins and lipids
Lysosomes	• Degradation of macromolecules • Degradation of worn-out organelles • Removal of excessive of secretory products • Secretion of perforin, granzymes, melanin and serotonin
Peroxisomes	• Breakdown of excess fatty acids • Detoxification of hydrogen peroxide and other metabolic products • Oxygen utilisation • Acceleration of gluconeogenesis • Degradation of purine to uric acid • Role in the formation of myelin • Role in the formation of bile acids
Centrosome	• Movement of chromosomes during cell division
Mitochondria	• Production of energy • Synthesis of ATP molecules • Initiation of apoptosis
Ribosomes	• Synthesis of proteins
Cytoskeleton	• Determination of the cell shape • Stability of cell shape • Cellular movements
Nucleus	• Control of all the cellular activities • RNA synthesis • Sending genetic instruction to cytoplasm for protein synthesis and formation of ribosomal subunits • Control of cell division • Storage of hereditary information in form of genes present in the DNA.

Abbreviations: ATP, adenosine triphosphate; DNA, deoxyribonucleic acid

FIG. 4.2: Structure of endoplasmic reticulum

membrane. ER is of two types, namely rough endoplasmic reticulum (RER) and smooth endoplasmic reticulum (SER) respectively based on the presence or absence of granular ribosomes on its outer surface. Both the types are interconnected and continuous with one another. The RER is mainly involved in the synthesis of proteins which are subsequently secreted outside the cell. RER also plays an important role in the degradation of worn-out cytoplasmic organelles like mitochondria.

Smooth endoplasmic reticulum, on the other hand, is responsible for synthesis of non-protein substances such as cholesterol and steroids. This type of ER is therefore abundant in cells, which are involved in the synthesis of lipids, phospholipids, lipoprotein substances, steroid hormones, sebum, etc. In most of the other cells, SER is less extensive than the RER. SER also plays a role in cellular metabolism. Outer surface of SER contains many enzymes which are involved in various metabolic processes of the cell. SER is also the major site of storage and metabolism of calcium. In skeletal muscle fibres, it releases calcium which is essential for initiating the contraction of muscles. SER is also concerned with catabolism and detoxification of toxic substances like some drugs and carcinogens.

Golgi Apparatus

Golgi apparatus (also known as Golgi body or Golgi complex) is a membrane-bound organelle, involved in the processing of proteins. It is present in all the cells except red blood cells. Each cell usually has one Golgi apparatus. However, some of the cells may have more than one Golgi apparatus. Each Golgi apparatus consists of 5–8 irregular sacs, vesicles or vacuoles and is situated near the nucleus. It has two ends or faces, i.e. cis face and trans face **(Fig. 4.3)**. The cis face is situated near the ER. Reticular vesicles from ER enter the Golgi apparatus through cis face. The trans face is situated near the cell membrane. The processed substances exit the Golgi apparatus through trans face. This way, it collects, modifies and transports secretions from the RER to the cell membrane.
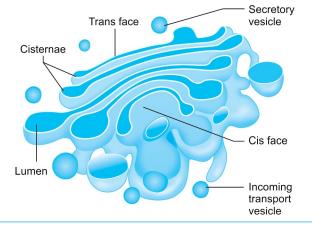
FIG. 4.3: Structure of Golgi complex

The Golgi apparatus is called "shipping department of the cell" because the major function of Golgi apparatus is processing, packing, labelling and delivery of proteins and other molecules like lipids to different parts of the cell.

Lysosomes

Lysosomes are the membrane-bound vesicular organelles containing proteolytic enzymes (acid hydrolases) which help in digesting unwanted endogenous and phagocytosed exogenous material. Important lysosomal enzymes include proteases, lipases, amylases, nucleases, etc. Due to their degradation activity, lysosomes are often known as the "garbage system" of the cell. Among the various cytoplasmic organelles, lysosomes have the thickest covering membrane, composed of a bilayered lipid material.

The lysosomes are present throughout the cytoplasm and are formed by Golgi apparatus. The enzymes synthesised in RER are processed and packed in the form of small vesicles in the Golgi apparatus. Then, these vesicles are pinched off from Golgi apparatus to form the lysosomes. There are two types of lysosomes: (1) primary and (2) secondary. Primary lysosomes are pinched off from Golgi apparatus. They are inactive in spite of having hydrolytic enzymes. Secondary lysosomes, on the other hand, are the active lysosomes which are formed by the fusion of a primary lysosome with phagosome or endosome. Phagosomes are membrane-bound bodies containing material ingested by phagocytosis. On the other hand, macromolecules taken inside the cell via pinocytosis or receptor-mediated endocytosis are called endosomes. The primary lysosome fuses with the phagosome or endosome to form the secondary lysosome. As a result of this fusion, pH in the secondary lysosome becomes acidic and the lysosomal enzymes are activated. The bacteria and the other macromolecules are digested and degraded by these enzymes. The secondary lysosome containing these degraded waste products then moves through cytoplasm and fuses with cell membrane, following which the waste products are eliminated by exocytosis. Lysosomes also help in degradation of worn-out organelles and removal of excess secretory products in the cells.

Recently, lysosomes have been discovered to have a secretory function, particularly in the cells of immune system. The conventional lysosomes are modified into secretory lysosomes upon combination with secretory granules (which contain the particular secretory product of the cell): e.g. lysosomes in the cytotoxic T lymphocytes and natural killer cells secrete perforin and granzymes, which help in destroying both viral-infected cells and tumour cells; secretory lysosomes of melanocytes secrete melanin; secretory lysosomes of mast cells secrete serotonin, etc.

Peroxisomes

Peroxisomes or microbodies are the membrane-bound vesicles, which are pinched off from ER and not from the Golgi apparatus, unlike the lysosomes. The main function of peroxisomes is the breakdown the fatty acids through beta (β)-oxidation. They also help in degrading the toxic substances such as hydrogen peroxide and other metabolic products in the cell by detoxification. Peroxisomes form the major site of oxygen utilisation in the cells; accelerate gluconeogenesis from fats; degrade purine to uric acid; participate in the formation of myelin and play a role in the formation of bile acids.

Centrosome and Centrioles

Centrosome is the membrane-bound cellular organelle situated almost in the centre of cell, close to nucleus. It consists of two hollow cylindrical structures called centrioles which are 0.3–0.7 μm in length and are made up of proteins. Centrioles are responsible for the movement of chromosomes during cell division. The centrioles replicate before mitosis and align the mitotic spindle.

Secretory Vesicles

Secretory vesicles are the cell organelles with limiting membrane, containing the secretory substances, present throughout the cytoplasm. These vesicles are formed in the ER and are processed and packed in Golgi apparatus. Secretory substances are released into the cytoplasm upon rupture of these vesicles.

Mitochondrion

Mitochondrion (plural = mitochondria) is a membrane-bound cytoplasmic organelle known as the "power house" or "power plant" of the cell because it produces the energy required for cellular functions. It is a rod-shaped or oval-shaped, elongated structure having a diameter of 0.5–1μ. It is covered by a bilayered membrane **(Fig. 4.4)**. The outer membrane is smooth and contains various enzymes such as acetyl-CoA synthetase and glycerophosphate acetyltransferase. The inner membrane is folded in the form of shelf-like inward septa called cristae and it covers the entire inner cavity of mitochondrion. Inner cavity of
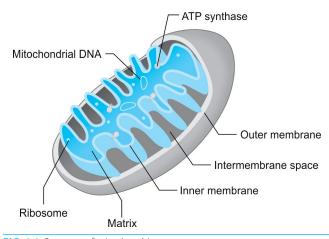
FIG. 4.4: Structure of mitochondrion
Abbreviations: DNA, deoxyribonucleic acid; ATP, adenosine triphosphate

mitochondrion is filled with matrix which contains many enzymes. Cristae contain many enzymes and other protein molecules which are involved in respiration and synthesis of adenosine triphosphate (ATP). The enzymes and other protein molecules in cristae collectively form the respiratory chain or electron transport system. The components of respiratory chain in mitochondrion are responsible for the synthesis of ATP by utilising the energy produced during the oxidation of digested food particles like proteins, carbohydrates and lipids by oxidative phosphorylation.

Mitochondrion moves freely in the cytoplasm of the cell. Mitochondrion is the only organelle in the cell other than nucleus, which has its own DNA and is therefore capable of replicating itself.

Nucleus

Nucleus is the most prominent and the largest cellular organelle having a diameter of 10–22 μ. It occupies nearly 10% of total volume of the cell. Presence of the nucleus is crucial for the cell division. Nucleus is present in all the body cells except the red blood cells. The cells with nucleus are called eukaryotes and those without nucleus are known as prokaryotes. Most of the body cells are uninucleated cells, i.e. having a single nucleus. However, some cells may also have multiple nuclei, i.e. multinucleated cells, e.g. skeletal muscles. The nucleus is usually located in the centre of the cell and is mostly spherical in shape. However, the shape and situation of nucleus may vary in some cells. Nucleus is covered by a membrane called nuclear membrane and contains many components, i.e. nucleoplasm, chromatin and the nucleolus.

Nuclear membrane is double layered and porous in nature, thereby allowing communication of the nucleoplasm with the cytoplasm. The outer layer of nuclear membrane is continuous with that of ER. The space between the two layers of nuclear membrane is continuous with the lumen of ER.

Chromatin is a thread-like material made up of large molecules of DNA, which are compactly packed with the help of a specialized basic protein called histone, thereby forming a DNA-histone complex. Just before cell division, the chromatin condenses to form chromosomes. Chromosome is the rod-shaped nuclear structure that contains a complete blueprint of all the hereditary characteristics of that species.

Nucleolus is a small, round granular structure present in the nucleus. Each nucleus contains one or more nucleoli. The nucleolus contains RNA and some proteins, which are similar to those found in ribosomes.

Nucleus is involved in controlling several important activities of the cell including metabolism, protein synthesis, cell growth and division. It is involved in the synthesis of RNA and formation of ribosomal subunits. Nuclear RNA is a precursor of cytoplasmic ribosomal RNA. It also sends genetic instruction to the cytoplasm for protein synthesis through messenger RNA (mRNA). Nucleus controls the division of cells through genes. The cell's hereditary information is stored in the genes and transformation of this information from one generation of the species to the next is also carried on by the nucleus.

Cytoplasmic Organelles without Limiting Membrane

Ribosomes

Ribosomes are the organelles without limiting membrane. These organelles are small round granular structures having a diameter of 15 nm. Ribosomes are made from complexes of RNAs (65%) and proteins (35%). Ribonucleic acid (RNA) present in ribosomes is called ribosomal RNA (rRNA).

Ribosomes are called "protein factories" because they are mainly concerned with synthesis of proteins required for intracellular metabolism. The ribosomes assemble amino acids to form specific proteins, essential for carrying out various cellular activities. In the cytoplasm, the two subunits of ribosomes are bound around the polymers of mRNA; proteins are then synthesised with the help of transfer RNA (tRNA). The number of ribosomes in a cell depends upon the activity of the cell. The existence of ribosomes is temporary. Following the synthesis of the polypeptide molecule, the two ribosomal subunits separate and may be then reused or broken up.

Ribosomes are of two types: (1) ribosomes that are attached to RER; and (2) free ribosomes that are distributed in the cytoplasm. Ribosomes attached to RER are involved in the synthesis of proteins such as the enzymatic proteins, hormonal proteins, lysosomal proteins and the proteins of the cell membrane. Free ribosomes are responsible for the synthesis of proteins in haemoglobin, peroxisome and mitochondria.

Cytoskeleton

Cytoskeleton is the cellular organelle, which helps in determining shape of the cell. It is present throughout the cytoplasm and gives support to the cell. It is a complex network of structures with varying sizes. It is also essential for determining the cellular movements and the response of the cell to external stimuli. Cytoskeleton consists of three major protein components: (1) Microtubule, (2) Intermediate filaments and (3) Microfilaments.

Microtubules

Microtubules are straight, hollow and tubular structures of the cytoskeleton, which are arranged in form of different bundles. These organelles do not have a limiting membrane. Each tubule has a diameter of 20–30 nm. Length of microtubule varies and it may be 1,000 times more than the thickness.

Structurally, the microtubules are formed by bundles of globular protein called "tubulin", which is composed of two subunits, namely α-subunit and β-subunit. Microtubules constitute the mitotic spindle filaments which separate the chromosomes during mitosis. Microtubules may facilitate intracytoplasmic transport and maintain cellular shape and strength. They are also responsible for the movement of centrioles and the complex cellular structures like cilia.

Intermediate Filaments

The intermediate filaments are formed by rope-like polymers, made up of fibrous proteins that form a network around the nucleus extending up to the periphery of the cell. Diameter of each filament is about 10 nm. Intermediate filaments are divided into five subclasses: (1) keratins (in epithelial cells); (2) glial filaments (in astrocytes); (3) neurofilaments (in nerve cells); (4) vimentin (in many types of cells) and (5) desmin (in muscle fibres). Intermediate filaments help in maintaining the shape of the cell. These filaments also connect the adjacent cells through desmosomes.

Microfilaments

Microfilaments are long and fine thread-like structures having a diameter of 4–12 nm having an indefinite length. These filaments are made up of non-tubular contractile proteins called actin and myosin. Actin is more abundant than myosin. Microfilaments are present throughout the cytoplasm.
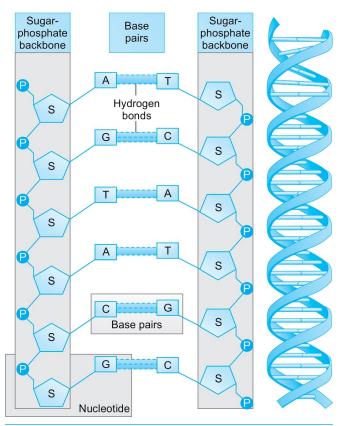
Carriers of Genetic Information

Deoxyribonucleic Acid

Deoxyribonucleic acid (DNA) is a nucleic acid which forms the chemical basis of hereditary characters through the formation of genes. It is present in the chromosomes and mitochondria of the cells. The DNA present in the nucleus is responsible for the formation of RNA which regulates the synthesis of proteins by ribosomes.

Structure of DNA

Deoxyribonucleic acid is a double-stranded complex nucleic acid having two anti-parallel strands, each of which is a helical polynucleotide chain with opposite polarity, i.e. one chain runs from 5' to 3' direction and other runs from 3' to 5' direction **(Fig. 4.5)**. The DNA molecule is coiled in form of a right-handed helical pattern with each turn having about 12 nucleotides. Each DNA molecule consists of several nucleotides each of which is composed of deoxyribose (sugar), phosphoric acid and four types of bases. The two chains of DNA are linked with help of sugar-phosphate backbone. DNA contains one of the following organic (nitrogenous) bases, purines: adenine (A) and guanine (G), and pyrimidines: thymine (T) and cytosine (C). The nucleic acids of the complementary strands form pairs, i.e. adenine pairs with thymine and cytosine pairs with guanine. Therefore, if one chain has the sequence 3'-AGGTCGCG-5', the other complementary chain would have the sequence 5'-TCCAGCGC-3'.

Deoxyribonucleic acid forms the component of chromosomes, which carries the hereditary information. The hereditary information that is encoded in DNA is called

FIG. 4.5: Structure of deoxyribonucleic acid (DNA)
Abbreviations: P, phosphate; S, sugar; A, adenine; T, thymine; C, cytosine; G, guanine

genome. Each DNA molecule is divided into discrete units called genes. Gene is a portion of DNA molecule containing the message or code for synthesis of a specific protein from amino acids. In the nucleotide of DNA, three of the successive base pairs are together called a triplet or a codon. Each codon contains information for a particular amino acid. There are 20 amino acids in all and each specific amino acid is coded by a specific codon. For example, the amino acid proline is coded by the triplet CCA. Each amino acid can be coded by more than one specific codon. However each codon represents only a specific amino acid.

DNA replication: DNA polymerase is an enzyme that assists in DNA replication. DNA polymerase initiates DNA replication by binding to a piece of single-stranded DNA.

This enzyme catalyses the polymerisation of deoxyribonucleotides alongside a DNA strand, which it "reads" and uses as a template. The newly polymerised molecule is complementary to the template strand and identical to the template's partner strand.

Methods of DNA Analysis

Polymerase chain reaction: Polymerase chain reaction (PCR) is a molecular biology technique **(Fig. 4.6)** used for amplifying a single copy or a few copies of a piece of DNA, thereby generating multiple copies of a particular DNA sequence. In this technique, the DNA is denatured by heating it to separate it into individual strands. Specific synthetic oligonucleotides (known as primers) are bound to the complementary sequences on the target DNA. This process is known as annealing. Primers or short DNA fragments containing sequences complementary to the target region of the DNA along with a DNA polymerase are used for selective and repetitive amplification of a particular DNA sequence. Use of the primer may sometimes prove to be a disadvantage because the primers used in the reaction must be complementary to the nucleic acid sequence surrounding the region to be amplified. Therefore, these sequences must be known.

DNA polymerase is then used for extending the oligonucleotide in the 5' to 3' direction, thus forming new DNA strands. This process is repeated several times to generate multiple copies of the original DNA. A heat stable DNA polymerase such as Taq polymerase (originally isolated from the bacterium *Thermus aquaticus*) is commonly used.

Polymerase chain reaction is a rapid technique, which produces a result in only a few hours. It is therefore extremely useful for rapid diagnosis of conditions such as tuberculosis where traditional method of culturing the bacteria can take several weeks. This technique allows scientists directly and exponentially to amplify small samples of DNA and even RNA through reverse transcriptase PCR. DNA is the standard template used in the PCR technique, but viral RNA sequence can also be amplified if the enzyme reverse transcriptase is used. The test may be used for the prenatal diagnosis of conditions such as cystic fibrosis.

In human immunodeficiency virus (HIV) and perhaps other viruses, sequence polymorphism may prevent binding of primers and result in failure of amplification. Primers which were developed for amplification of the predominant HIV clade B strain found in Europeans and Americans have proved unreliable for amplification of other HIV clades from Africa and Asia.

Restriction endonucleases: These are the enzymes, which recognise a short nucleotide sequence (restriction site) in a double-stranded DNA molecule. They cleave the DNA molecule whenever they encounter a restriction site (about 4-8 nucleotides). There are more than 100 restriction endonucleases each with a specific restriction site. Following the treatment with these enzymes, the DNA molecule is split into multiple fragments, which can be separated through gel electrophoresis.

Southern blot hybridisation: This method is named after its inventor, the British biologist, Edwin Southern. This technique involves identification of specific DNA fragments separated through electrophoresis. The individual fragments are detected through probe hybridisation.
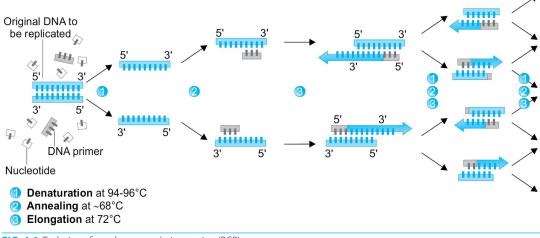

Denaturation at 94-96°C
Annealing at ~68°C
Elongation at 72°C

FIG. 4.6: Technique for polymerase chain reaction (PCR)
Abbreviation: DNA, deoxyribonucleic acid

Other blotting methods, e.g. Western blot, Northern blot, Northwestern blot, etc. enjoy similar principles, but use either RNA or proteins. Northern blotting is used for identifying specific RNA sequences in RNA-rich matter. Western blotting identifies specific proteins in samples using both gel electrophoresis and immunoblotting, whereby the antibodies mark the "target" proteins. Eastern blotting technique can be considered as an extension of the western blotting technique and helps in detecting post-translational protein modifications. Northwestern blotting is a fictional technique.

Restriction fragment length polymorphism (RFLP): This is a technique, which exploits variations in homologous DNA sequences. It refers to the difference between samples of homologous DNA molecules having different locations of restriction enzymes.

Polymorphism refers to changes in DNA base sequence. Polymorphisms occur only in the non-coding sequences and hence do not alter an individual's phenotype. These polymorphisms can be demonstrated by the use of restriction endonucleases and are then known as RFLPs. RFLP is represented by a non-coding DNA sequence present approximately one in every 100–200 base pairs in a non-coding DNA sequence. Diagnosis of a genetic disease is possible when the locus for the disease is included with a specific polymorphic DNA fragment. For example, the restriction endonuclease Hpal cleaves fragments of different sizes from the HbS and HbA gene. The fragment of HbA is 7.6 kb in size and can be easily distinguished from the HbS fragment which is 13 kb in size.

Ribonucleic Acid

Ribonucleic acid (RNA) is a single-stranded nucleic acid containing a long chain of nucleotide units, unlike the double-stranded DNA. While RNAs are typically single-stranded, in some viruses, RNA may be double stranded, e.g. rota viruses. Similar to DNA, the nucleotides in RNA are composed of sugar, phosphoric acid and four types of bases. However, unlike the DNA, the sugar is of ribose type in the RNA instead of deoxyribose. The nitrogenous bases present in RNA are similar to that of DNA, except that in RNA, uracil replaces the thymine of DNA and it has a structure similar to that of thymine. The formation of RNA is catalysed by RNA polymerase, which links together RNA nucleotides using a DNA template in a process called transcription. RNA is of three types. Each type of RNA plays a specific role in protein synthesis. The three types of RNA are as follows:

Messenger RNA: Messenger RNA carries the genetic code of the amino acid sequence for synthesis of protein from the DNA to the cytoplasm. Messenger RNA is translated into proteins by the joint action of transfer RNA and the ribosomes which are composed of numerous proteins and two major ribosomal RNA molecules.

Transfer RNA: Transfer RNA is responsible for decoding the genetic message present in mRNA.

Ribosomal RNA: Ribosomal RNA is present within the ribosome and forms a part of the structure of ribosome. It is responsible for the assembly of protein from amino acids in the ribosome.

Carbohydrate Metabolism

It is essential for every cell to maintain an adequate supply of energy. The energy obtained through metabolism of food stuffs (carbohydrates, fatty acids and at times amino acids) is stored in form of ATP molecules. The principal carbohydrate used in body metabolism is glucose. ATP is essential for bodies' several cellular processes such as protein synthesis and maintenance of ionic gradients across the plasma membrane. Carbohydrates are not essential to the diet, because the body can synthesize necessary carbohydrates from certain amino acids through the process of gluconeogenesis.

The metabolism of carbohydrates (sugars), fatty acids and amino acids begins with pathways that are specific for each energy source. The products from these pathways then feed into common pathways. Sugars, fatty acids and amino acids are eventually metabolised to produce acetate in form of acetyl coenzyme A, which serves as a central compound in metabolism. These acetyl coenzyme A molecules then enter the citric acid cycle where they are completely degraded resulting in the production of CO_2 and hydrogen in form of nicotinamide adenine dinucleotide (NADH). These NADH molecules then feed into the respiratory chain inside the mitochondria and the energy of NADH is used for driving oxidative phosphorylation which produces ATP from ADP. The reaction within the respiratory chain requires molecular oxygen. Thus, this is known as the respiratory chain.

Metabolism of Glucose

Glucose is phosphorylated in the cytoplasm of cells to glucose-6-phosphate. Glucose-6-phosphate can be metabolised by the anaerobic glycolysis (Embden-Meyerhof pathway), aerobic glycolysis (Kreb's cycle) or pentose shunt. In cases when energy intake exceeds energy expenditure, glucose-6-phosphate can be converted to acetyl co-enzyme A for production of fats. When carbohydrate supply is limited, the body uses fats for energy. Excess acetyl CoA may be produced, which condenses to form acetoacetyl-CoA, which helps in the production of the ketone bodies.

In anaerobic glycolysis, glucose-6-phosphate is converted to lactate. Conversion to lactate further provides energy in the form of ATP. This mechanism acts as a soul source of energy under anaerobic conditions, e.g. muscles during strenuous exercise. However, glycolysis through

the anaerobic pathway is relatively energy inefficient in comparison with the aerobic Kreb's pathway. In aerobic glycolysis, glucose-6-phosphate can be converted to pyruvate which then undergoes Kreb's cycle [tricarboxylic acid (TCA) cycle].

Since erythrocytes have no nucleus, they lack a Kreb's cycle and rely on glycolysis via anaerobic glycolysis and pentose pathways for energy production.

Pentose shunt is aerobic, i.e. oxygen is consumed and CO_2 is released. It is active in many tissues (e.g. fat and red cells) and leads to the reduction of NADP (nicotinamide adenine dinucleotide phosphate) to NADPH. Pentose phosphate pathway is a metabolic pathway which is parallel to glycolysis and is involved in the generation of NADPH and pentoses. It does not involve oxidation of glucose; its primary role is anabolic rather than catabolic. More details related to the pentose phosphate pathway are described later in the text.

Glycolysis (Embden-Meyerhof Pathway)

Glycolysis is the process whereby glucose is converted to either pyruvate (aerobic metabolism) or lactate (anaerobic metabolism). Under aerobic condition, pyruvic acid enters mitochondria and is completely oxidized to CO_2 and H_2O. On the other hand, under anaerobic conditions, pyruvate is converted to lactic acid. Glycolysis is an extramitochondrial pathway, occurring in the cytoplasm of cells and is carried out by a group of 11 enzymes. In this pathway, a molecule of glucose containing 6 carbon atoms is broken down into two molecules of pyruvic acid containing 3 carbon atoms each. The pathway for glycolysis is illustrated in **Figure 4.7**. Glycolysis of one molecule of glucose produces two molecules of ATP (four molecules are produced and two molecules are utilised, resulting in a net gain of two molecules of ATP per molecule of glucose undergoing glycolysis). At the same time, two molecules of NAD are reduced to NADH. All the reactions of glycolysis are reversible except for the reactions catalysed by hexokinase, phosphofructokinase and pyruvate kinase. Glycolysis gives rise to certain intermediate compounds which are important for other biochemical processes. For example, glyceraldehyde-3-phosphate is utilised for the biosynthesis of triglycerides and phospholipids; acetyl CoA is used for biosynthesis of fatty acid and cholesterol. In a nutshell, glycolysis is the degradative pathway whereby D-glucose is oxidised to pyruvate, which is further metabolised by either of the two routes: when the supply of oxygen is inadequate for complete oxidation, the pyruvate is reduced to lactate. On the other hand, when the supply of oxygen is adequate (aerobic conditions), the pyruvate is oxidatively decarboxylated to acetyl CoA, which enters the citric acid cycle, where it is oxidised to carbon dioxide and water.

Kreb's Cycle

Kreb's cycle, also called tricarboxylic acid (TCA) cycle or citric acid cycle, involves complete oxidation of the acetyl moiety. This cycle takes place in mitochondria and results in the production of CO_2 molecules and hydrogen atoms. CO_2 diffuses out of the cell and is exhaled by the lungs. The hydrogen atoms cause the production of NADH molecules from NAD^+. The NADH molecules enter the respiratory chain in the mitochondria and with the consumption of oxygen are converted into water. It operates only under aerobic conditions, where there is a supply of oxygen. This process also requires a supply of NAD^+ and FAD^+ which are regenerated when NADH and $FADH_2$ transfer their electrons to O_2 through the electron transport chain. This sequence of reactions is known collectively as oxidative phosphorylation.

Pyruvate molecules produced via glycolysis enter the mitochondria and get oxidatively decarboxylated to acetyl CoA, which represents the formation of a 2 carbon molecule from a 3 carbon molecule (with the loss of one CO_2 and the formation of one NADH molecule). This reaction is catalysed by a multienzyme complex called pyruvate dehydrogenase complex **(Fig. 4.8)**. Acetyl CoA then enters the Kreb's cycle **(Fig. 4.9)**. Kreb's cycle or the citric acid cycle forms the common pathway for the metabolism of carbohydrates, fats and proteins because it results in the complete oxidation of acetyl CoA to carbon dioxide and water. In the Kreb's cycle

FIG. 4.7: Pathway for glycolysis
Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; ATP, adenosine triphosphate; ADP, adenosine diphosphate

acetyl-CoA is then condensed with the anion of a 4-carbon acid, oxaloacetate, to form citrate which is a 6-carbon molecule. Citrate is subsequently converted into isocitrate,
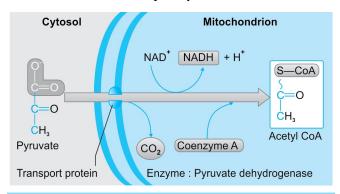

FIG. 4.8: Conversion of pyruvate to acetyl coenzyme A
Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; CO_2, carbon dioxide

α-ketoglutarate, succinyl-CoA, succinate, fumarate, malate and finally oxaloacetate. Alpha-ketoglutarate is the only 5 carbon molecule in the cycle. Thiamine is involved as a cofactor for numerous enzymes, and is essential in every cell for ATP production via the Kreb's cycle.

The rate-limiting step in the TCA cycle is the conversion of isocitrate to α-ketoglutarate. The enzyme involved is the citrate synthetase. The availability of acetyl CoA and oxaloacetate in plenty stimulates this enzyme while the presence of succinyl CoA inhibits this enzyme by competing with acetyl CoA.

Pyruvate can be channelled into TCA cycle as acetyl CoA or as oxaloacetate. This point, therefore serves as a switch point, which controls the main function of the cycle. If pyruvate is channelled to acetyl CoA then the cycle will generate mainly energy. On the other hand, if pyruvate is channelled into oxaloacetate, then the main function of the
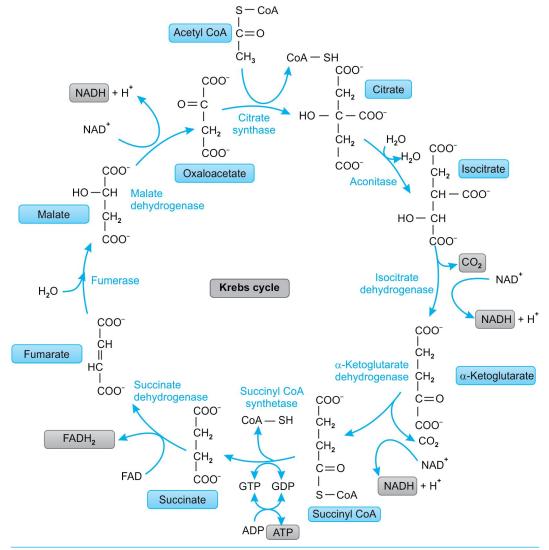
FIG. 4.9: Kreb's cycle
Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; $FADH_2$, flavin adenine dinucleotide (reduced); FAD, flavin adenine dinucleotide

cycle would be to produce carbon skeletons for the synthesis of amino acids or fats.

In cases of aerobic respiration, a single molecule of glucose can result in the formation of 36 molecules of ATP as a result of glycolysis, Kreb's cycle and oxidative phosphorylation via the electron transport chain. On the other hand, one molecule of glucose undergoing anaerobic respiration produces two molecules of ATP from glycolysis. This implies that aerobic respiration is much more energy efficient than the anaerobic respiration.

The citric acid cycle involves oxidation of several substrates including pyruvate and occurs under aerobic conditions. It is a major source of ATP and CO_2 and occurs in the mitochondrion in eukaryotic cells, and at the cell membrane in prokaryotic cells. Four molecules of NAD and one molecule of FAD are reduced during each TCA cycle to produce 4 molecules of NADH and one molecule of $FADH_2$ respectively. Each of the reduced nucleotide, NADH or $FADH_2$ may be oxidised in the mitochondrion via the electron transport chain to produce 3 or 2 molecules of ATP respectively. Therefore, each pyruvate molecule produces 14 molecules of ATP $[(3 \times 4) + (2 \times 1) = 14$ molecules] through oxidative phosphorylation via the electron transport chain. Also there is a gain of one molecule of ATP via the Kreb's cycle per se. The conversion of two molecules of pyruvate to acetyl coenzyme A also results in production of 2 molecules of NADH. The oxidative phosphorylation of each NADH molecule via the electron transport chain results in the production of 3 molecules of ATP per each molecule of NADH. At the same time one molecule of ATP is used for transporting each molecule of NADH from the cytosol to the mitochondrion. This results in a gain of 4 (6 – 2) molecules of ATP. Therefore there is a gain of 18 (14 + 4) molecules of ATP for each molecule of pyruvate, which undergoes oxidative phosphorylation. Since two pyruvate molecules are produced from each glucose molecule, there is a net production of 36 molecules of ATP. Also, the glycolytic pathway yields a net of 2 ATP molecules per glucose molecule. Therefore, the complete oxidation of glucose molecule was supposed to yield 38 molecules of ATP. However, according to the latest studies the total yield of energy during aerobic respiration is not 38 molecules of ATP, but 30–32 molecules per molecule of glucose oxidised because the energy produced as a result of oxidative phosphorylation may not be 3 and 2 for NADH and $FADH_2$ respectively, but 2.5 and 1.5 respectively.

On the other hand, one molecule of glucose yields only 2 ATP molecules during anaerobic metabolism. Therefore anaerobic respiration is associated with a much lower generation of energy.

Amphibolic Role of the Citric Acid Cycle

Citric acid cycle is primarily a catabolic process for the final oxidation of the carbohydrates, fats and proteins into CO_2 and H_2O. Simultaneously, this cycle also takes part in the various anabolic processes such as gluconeogenesis, fatty acid synthesis and amino acid synthesis by providing substrates, which are the normal intermediate products of this cycle. For example, the substrates, oxaloacetate and α-ketoglutarate are utilised for the synthesis of amino acids.

Thus, this cycle plays a dual or amphibolic role by exhibiting a vital role in both catabolism and anabolism.

Oxidative Phosphorylation

Mitochondria are the power house of the cell because the mitochondrial matrix serves as the site of citric acid cycle, the most important pathway for generation of energy in all aerobic organisms. Also, oxidative phosphorylation, the process by which the majority of energy in the cell is derived, occurs in the inner mitochondrial membrane. Oxidative phosphorylation can be defined as a series of redox reactions occurring along the proteins in the inner mitochondrial membrane, also known as the electron transport chain **(Fig. 4.10)**. Electrons are conducted through these proteins from electron donors to acceptors and are finally passed on to oxygen. These redox reactions release energy, which is ultimately collected to reform ATP.

Electron Transport System Pathway

The electron transport system consists of four large protein complexes and two small independent components known as ubiquinone (coenzyme Q_{10}) and cytochrome C. Electrons enter the electron transport system at the following two points:
1. Complex I (for electrons from NADH)
2. Complex II (for electrons from $FADH_2$).

These pathways meet at ubiquinone which is the start of the common electron transport system. The common pathway comprises of complex III, cytochrome C and complex IV. The final acceptor of electrons is molecular oxygen, which is ultimately reduced to form water. Protons are pumped from the matrix into the transmembrane space by complexes I, III and IV. The amount of chemical energy synthesised during the electron transport system per electron pair depends on where they join the chain:
❏ An electron pair from NADH passes through the complexes I, III and IV to produce 3 moles of ATP.
❏ From $FADH_2$, only 2 moles of ATP are synthesised because they join at complex II and bypass complex I.

Hexose Monophosphate Shunt Pathway

Though glycolysis is the principal pathway for the conversion of glucose into pyruvate in most tissues, there also exists an alternative pathway, the hexose monophosphate (HMP) shunt, which is also known as "Warburg-Dickens-Lipmann pathway", "pentose phosphate pathway", "phosphogluconate pathway", "direct oxidative pathway" or "reductive pathway". This is an alternative pathway to glycolysis and TCA cycle for the oxidation of glucose. About 10% of glucose enters this pathway every day. Tissues where this pathway is
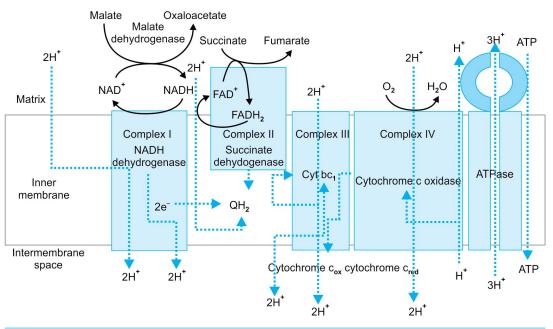
FIG. 4.10: Electron transport system
Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; FADH$_2$, flavin adenine dinucleotide (reduced); FAD, flavin adenine dinucleotide; ATP, adenosine triphosphate

more prominent are liver, adipose tissue, erythrocytes, lactating mammary gland, leucocytes, testes, adrenal cortex, etc. The enzymes of this pathway are found in the extra-mitochondrial cytoplasm.

Hexose monophosphate shunt is a unique multifunctional pathway, which starts with glucose 6-phosphate molecules. No ATP is directly utilised or produced in this pathway. Therefore, cells do not use the shunt pathway for energy production.

The reactions of the pathway can be divided into two phases: (1) oxidative and (2) non-oxidative **(Fig. 4.11)**.

Oxidative Phase

The oxidative phase comprises of the following steps:
- ❑ *Step 1*: Glucose 6-phosphate is oxidised by NADP-dependent Glucose 6-phosphate dehydrogenase, resulting in the formation of 6-phosphogluconolactone. NADPH is formed in this reaction and this serves as a rate limiting step.
- ❑ *Step 2*: 6-phosphogluconolactone is hydrolysed by the enzyme gluconolactone hydrolase to form 6-phospho-gluconate.
- ❑ *Step 3*: The next reaction involves the synthesis of NADPH. In this reaction, 6-phosphogluconate is decar-boxylated by the enzyme 6-phosphogluconate dehydro-genase to produce ribulose 5-phosphate.

Non-oxidative Phase

The non-oxidative phase comprises of the following steps:
- ❑ *Step 4*: The ribulose-5-phosphate is then isomerised to ribose-5-phosphate or epimerised to xylulose-5-phosphate.

- ❑ *Step 5*: This reaction is catalysed by the enzyme transketolase, which is a thiamine pyrophosphate (TPP) dependent enzyme. It transfers two-carbon unit from xylulose 5-phosphate to ribose 5-phosphate to form a 7-carbon sugar, sedoheptulose 7-phosphate and glyceraldehyde 3-phosphate respectively.
- ❑ *Step 6*: This reaction is catalysed by the enzyme transaldolase, which transfers a 3-carbon fragment from sedoheptulose 7-phosphate to glyceraldehyde 3-phosphate to give rise to fructose 6-phosphate and 4-carbon atom molecule erythrose 4-phosphate.
- ❑ *Step 7*: This reaction is catalysed by transketolase again. In this reaction, a 2-carbon unit is transferred from xylulose 5-phosphate to erythrose 4-phosphate to form fructose 6-phosphate and glyceraldehyde 3-phosphate respectively. Fructose 6-phosphate and glyceraldehyde 3-phosphate are further metabolised by glycolysis and TCA cycle.

Significance of Hexose Monophosphate Shunt Pathway

As previously described, this pathway generates NADPH, which is required in the reductive synthesis of fatty acids, triglycerides and steroids. NADPH is used in the synthesis of certain amino acids involving the enzyme glutamate dehydrogenase and is also used for scavenging the free radicals. This pathway also helps in the formation of pentose sugars (Ribose-5-phosphate), which are required for the synthesis of nucleotides and nucleic acids. This pathway is also important in plants, which synthesise glucose from CO_2 through the process of photosynthesis.

NADPH is used for maintaining the erythrocyte mem-brane integrity, prevention of methaemoglobinaemia,
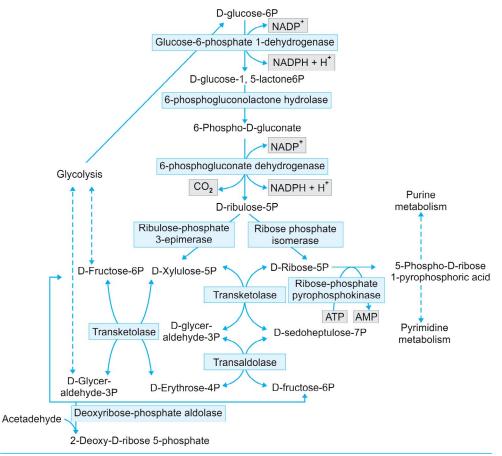
FIG. 4.11: Pentose phosphate pathway
Abbreviations: NADPH, nicotinamide adenine dinucleotide phosphate (reduced); NADP, nicotinamide adenine dinucleotide phosphate; ATP, adenosine triphosphate; AMP, adenosine monophosphate; CO_2, carbon dioxide

detoxification of many drugs and foreign substances, preservation of the transparency of lens of the eye, etc.

Glycogenesis

Glycogen is a highly branched homopolymer of α-D-glucose. It can be considered as the storage form of glucose in the animals. High glycogen content is mainly present in the muscles and liver of the humans. Glycogen in the liver helps in regulating the blood glucose levels. Glycogen stored in the muscles serves as a fuel at the time of strenuous exercise. In a molecule of glycogen, there may be up to 13 glucose residues (linked by α (1→4) linear glycosidic linkages. Branching may occur once after every 8–10 residues, with the branching linkages being α (1→6) glycosidic linkages **(Fig. 4.12)**.

The formation of glycogen from glucose is known as glycogenesis **(Fig. 4.13)**. Glycogenesis takes place in the cytosol and requires ATP and UTP, besides glucose.

Steps

Under the combined action of glycogen synthetase and branching enzyme, glucose units are added to the non-reducing ends of the pre-existing glycogen by α-(1,4) and α-(1,6) linkages to form glycogen.
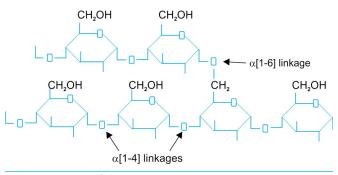
FIG. 4.12: Structure of glycogen

Glucose is phosphorylated to glucose-6-phosphate, by the enzyme hexokinase (in muscle) and glucokinase (in liver). Glucose-6-phosphate is then converted to glucose-1-phosphate, a reaction catalysed by the enzyme phosphoglucomutase. Glucose-1-phosphate reacts with uridine triphosphate (UTP) to form uridine diphosphate glucose (UDPG); this reaction is catalysed by the enzyme UDP-glucose pyrophosphorylase.

Now in the presence of the enzyme glycogen synthetase, C-1, carbon atom of glucose of UDPG forms a glycosidic linkage α-(1,4) with the C-4 atom of the pre-existing glycogen molecule. A small fragment of pre-existing glycogen must act as a "primer" to initiate glycogen synthesis. A specific
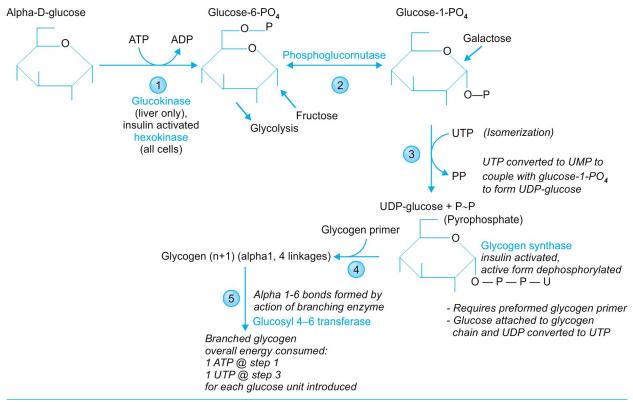
FIG. 4.13: Process of glycogenesis
Abbreviations: ATP, adenosine triphosphate; ADP, adenosine diphosphate; UTP, uridine triphosphate; UMP, uridine monophosphate; UDP, uridine diphosphate

protein "glycogenin" can accept glucose from UDP glucose. The hydroxyl group (OH) of the amino acid tyrosine of glycogenin is the site at which the initial glucose unit is attached. The enzyme glycogen initiator synthase transfers the first molecule of glucose to glycogenin. Then glycogenin itself takes up a few glucose residues to form a fragment of primer, which serves as an acceptor for the rest of the glucose molecules. Glycogen synthase is responsible for the formation of 1,4-glycosidic linkages. This enzyme transfers the glucose from UDP-glucose to the non-reducing end of glycogen to form α-1,4 linkages. Glycogen synthase catalyses the synthesis of a linear unbranched molecule with α-1,4 glycosidic linkages. However, glycogen is a branched tree-like structure. The formation of branches is brought about by the action of a branching enzyme, known as glucosyl-4-6 transferase. This enzyme transfers a small fragment of 5–8 glucose residues from the non-reducing end of glycogen chain (by breaking α-1,4 linkages) to another glucose residue where it is linked by α-1,6 bond. This leads to the formation of a new non-reducing end, besides the existing one. Branching is important because it helps increase the solubility of glycogen and provides a large number of non-reducing sugar terminals which are the sites of activity for glycogen phosphatase, the enzyme which helps in breaking up glycogen.

Glycogen is further elongated and branched, by the enzymes glycogen synthase and glucosyl 4-6 transferase.
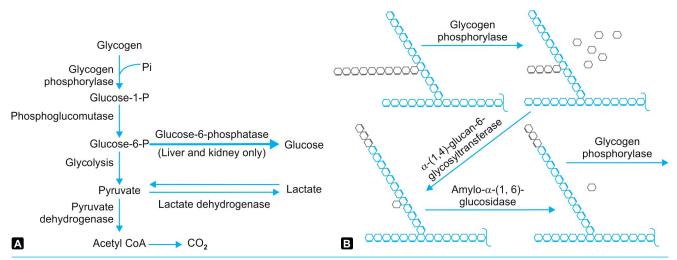
Regulation of Glycogenesis

Glycogenesis is controlled by both allosteric and hormonal regulation.

Allosteric regulation: In this process an effector molecule binds to protein at a site, other than the active site known as the allosteric site. This results in conformational change which can enhance activity (allosteric activation) or inhibit activity (allosteric inhibition). In the fed state, glycogen synthase, the target enzyme regulating glycogenesis, is allosterically activated by glucose 6-phosphate and ATP.

Hormonal regulation: Insulin stimulates the activity of glycogen synthase and therefore glycogenesis. Insulin is released by the pancreatic β cells when the blood glucose levels are high.

Glycogenolysis

Breakdown of stored glycogen to glucose is known as glycogenolysis and occurs in the cell cytoplasm **(Figs 4.14A and B)**. This pathway is usually triggered by low blood glucose levels and takes place in liver and muscle. Glycogen is gradually degraded between meals by glycogenolysis, releasing glucose to maintain blood glucose concentration. However total hepatic stores of glycogen are barely sufficient for maintaining blood glucose levels during a 12-hour fasting period. Therefore, there is a gradual

FIGS 4.14A AND B: Glycogenolysis (A) The process of glycogenolysis; (B) Action of various enzymes involved in glycogenolysis
Abbreviations: Pi, inorganic phosphate, CO_2, carbon dioxide

shift from glycogenolysis to de novo synthesis of glucose (gluconeogenesis).

Glycogenolysis in the liver is also activated when there is an increased demand for glucose either during the postabsorption state or in preparation for increased utilisation, i.e. during the periods of increased stress.

In liver, the end product of glycogen breakdown is glucose, whereas in muscles the end product is lactic acid. Glycogenolysis occurs due to the joint action of the enzyme phosphorylase [breaks only α-(1,4) linkages] and debranching enzymes [breaks only α-(1,6) linkages].

The breakdown of glycogen is initiated by the enzyme phosphorylase, which cleaves α-(1,4) glycosidic-linkages starting from non-reducing end of the glycogen molecule to give rise to glucose 1-phosphate after utilising one phosphate molecule. This process continues until four glucose residues remain on either side of the α-(1,6) branched point. Most glycogen breakdown occurs through this activity.

The branching links, the α-(1,6) glycosidic-linkages are then cleaved by glycogen-debranching enzyme which has both transglycolase and glucosidase activity. Transglycolase then transfers three glucose units from one side to another, leaving a single glucose residue at the branched point. The glycosidase activity of the debranching enzyme then helps in breaking the α-(1,6) glycosidic-linkage.

Glucose-1-phoshate produced by the enzyme phosphorylase is converted to glucose-6-phosphate by phosphoglucomutase. In the liver, glucose is released from glucose-6-phosphate by the enzyme glucose-6-phosphatase. The glucose molecules then exit through the glucose transporter type 2 (GLUT2) into the blood.

The breakdown of glycogen takes place in liver and muscle. Specific phosphorylases exist in the muscle and liver.

Liver phosphorylase: It exists in two forms: The active form, phosphorylase and the inactive form, the dephosphophosphorylase. Activation of the inactive form involves phosphorylation of the hydroxyl group of a serine residue by a specific kinase in the presence of ATP. Inactivation of the active form is catalysed by a specific phosphatase. Glucagon and adrenaline stimulate glycogenolysis.

Muscle phosphorylase: It exists in the following forms: the active form, phosphorylase a and the inactive form, phosphorylase b. Phosphorylase a is a tetramer containing 4 molecules of pyridoxal phosphate and is active only in the absence of 5-AMP. On the other hand, phosphorylase b is a dimer containing only 2 molecules of pyridoxal phosphate and is active only in the presence of 5-AMP.

Regulation of glycogenolysis: Glycogenolysis and glycogenesis are opposite pathways. Therefore, activation of glycogenolysis is co-ordinated with the inactivation of glycogenesis. Glycogenolysis is controlled by enzyme glycogen phosphorylase, which can be regulated by each of the following three mechanisms:
1. Allosteric regulation
2. Hormonal regulation
3. Influence of calcium.

Allosteric regulation: Glycogen breakdown inhibited by:
❏ High levels of glucose-6-phosphate
❏ ATP
❏ Free glucose in liver.

Glycogen breakdown is increased in the following cases:
❏ Low glucose concentration
❏ Low energy level.

Hormonal regulation: There are three hormonal activators of glycogenolysis: (1) glucagon, (2) adrenaline (epinephrine) and (3) cortisol. Low blood glucose fasting levels releases these hormones. These hormones stimulate intracellular pathway via increasing the levels of cAMP, which ultimately activates glucogen phosphorylase resulting in the breakdown of glycogen **(Fig. 4.15)**.
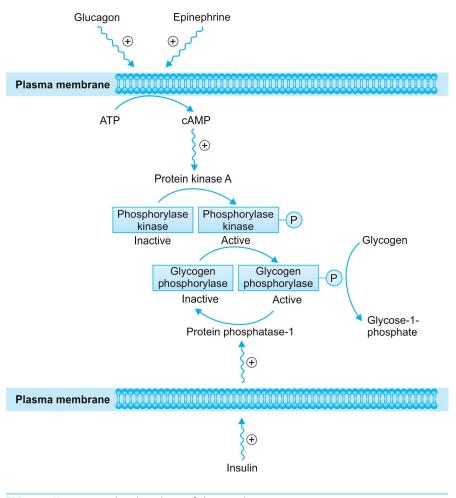
FIG. 4.15: Hormone-mediated regulation of glycogenolysis
Abbreviations: ATP, adenosine triphosphate; CAMP, cyclic adenosine monophosphate

Regulation by calcium ions: Calcium ions regulate glycogen breakdown in the muscles. Release of Ca^{2-} from the ER into cytosol of muscle cells causes muscle contraction resulting in an urgent requirement of ATP **(Fig. 4.16)**.

Glycogen Storage Diseases

Glycogen storage diseases result from defects in glycogen synthesis or breakdown. These are a group of inborn error of metabolic diseases in which there is an accumulation of abnormally large amount of glycogen in the tissues due to the deficiency or absence of enzymes involved in glycogen metabolism. These diseases arise as a result of a dysfunction of one of the enzymes in the pathway. Various types of glycogen storage diseases are described in the **Table 4.3** and **Figure 4.17**.

Cori Cycle

The Cori cycle is the body's way of recycling lactic acid. The cyclic process by which lactic acid is converted to glucose in the liver and eventually reappears as muscle glycogen is known as Cori cycle **(Fig. 4.18)**. During vigorous muscular activity, muscle glycogen is converted to lactic acid.

The lactic acid diffuses from the muscle into the blood stream and is transferred to the liver. In liver, lactic acid is converted to glucose by gluconeogenesis. Glucose formed in this way returns to the muscle via circulation. This cycle continues and is known as the Cori cycle.

Gluconeogenesis

Gluconeogenesis is the process by which glucose or glycogen is formed from non-carbohydrate substances such as glycogenic amino acids (alanine, glutamine, etc.), intermediates of TCA cycle, glycerol, pyruvate, lactate, etc. Gluconeogenesis is an important pathway for supplying glucose to various tissues when glucose is otherwise not available, especially at the times of prolonged fasting, starvation or strenuous exercises. The end product is the continuous supply of glucose for the brain and RBCs which rely on glucose as their energy source. Liver and kidneys are the major sites for gluconeogenesis. The tissues such as brain, adipose tissues or muscles are not gluconeogenic. Gluconeogenesis takes place when the energy requirements of the cell are at a minimal level and an energy source of ATP is available.

TABLE 4.3	Various types of glycogen storage diseases	
Type name of disease	Type of glycogen storage disease (GSD)	Enzyme which is deficient
Von Geirke's disease	GSD I (type Ia and Ib)	Glucose-6-phosphatase (type Ia is due to deficiency of the enzyme glucose-6-phosphatase, while type Ib is due to the deficiency of a specific translocase T1 which is related with the transport defect of glucose-6-phosphate into the microsomal compartment)
Pompe's disease	GSD II	α-(1,4) glucosidase
Forbe's or Cori's disease or limit dextrinosis	GSD III	Amylo-1, 6-glucosidase, i.e. debranching enzyme
Andersen's disease (amylopectinosis)	GSD IV	1,4 → 1,6 transglucosidase
McArdle's disease	GSD V	Muscle glycogen phosphorylase
Her's disease	GSD VI	Hepatic phosphorylase
Tauri's disease	GSD VII	Phosphofructokinase

FIG. 4.16: Calcium mediated regulation of glycogenolysis
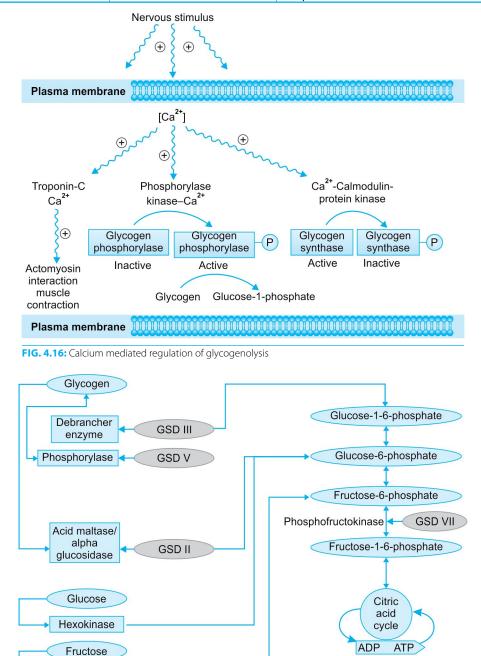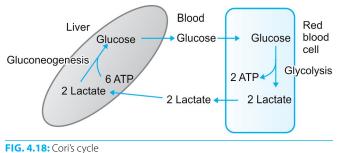
FIG. 4.17: Various types of glycogen storage diseases
Abbreviation: GSD, glycogen storage disease

FIG. 4.18: Cori's cycle
Abbreviation: ATP, adenosine triphosphate

Steps

The pathway for gluconeogenesis is described in **Figure 4.19**. Insulin blocks the synthesis of glucose whereas glucocorticoid hormones induce their de novo synthesis. Many reactions in this pathway are the reverse of glycolysis. However, some steps are irreversible. The pathway begins with the carboxylation of pyruvate catalysed by the enzyme pyruvate carboxylase resulting in the formation of oxaloacetate in the mitochondrion. The enzyme pyruvate carboxylase is stimulated by acetyl CoA and inhibited by ADP.

Oxaloacetate undergoes decarboxylation and then phosphorylation to form phosphoenolpyruvate with help of the enzyme phosphoenolpyruvate carboxykinase.

Next steps are the reversal of glycolysis, which eventually result in the formation of glucose-6-phosphate. Glucose-6-phosphate can be used in other metabolic pathways or dephosphorylated with the help of the enzyme glucose-6-phosphatase to form free glucose in the ER. This reaction results in a release of an inorganic phosphate molecule. The enzyme, glucose-6-phosphatase is stimulated by inorganic phosphate (Pi) and glucose.

While glucose-6-phosphate is locked inside the cells, free glucose can diffuse in and out of the cells. Glucose is transported into the cytoplasm with help of glucose transporters located on the membrane of ER (glucose transporter-1). For each molecule of glucose formed, there is a net consumption of 4 molecules of ATP and 2 molecules of GTP.

Other Fuels: Fructose and Galactose

Independent of the pathways of glycogenolysis and gluconeogenesis, glucose can also be produced in the body with the help of sugars such as fructose and galactose. Fructose is one of the three main dietary monosaccharides, along with glucose and galactose. In combination with glucose, it forms the disaccharide, sucrose. Galactose is a monosaccharide, which is derived mainly from lactose of the diet. Galactose is important for the formation of glycolipids and glycoprotein and for the formation of lactose during lactation.
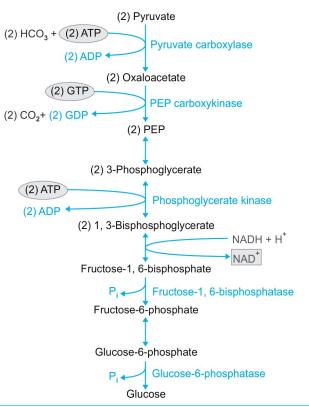

FIG. 4.19: Pathway of gluconeogenesis
Abbreviations: PEP, phosphoenolpyruvate; ATP, adenosine triphosphate; ADP, adenosine diphosphate; GTP, guanosine triphosphate; GDP, guanosine diphosphate; NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide

Fructose Metabolism

Fructose enters the glycolytic pathway through two routes:
1. *Liver*: Most of the fructose in the body is predominantly metabolised through this pathway **(Fig. 4.20)**. In the liver, fructose is converted to fructose 1-phosphate by the enzyme fructokinase. Fructose 1-phosphate is then split into dihydroxyacetone phosphate and glyceraldehyde by fructose 1-phosphate aldolase and both the molecules are then converted into glyceraldehyde 3-phosphate. Glyceraldehyde 3-phosphate is then used for replenishing liver glycogen stores or for the synthesis of triglycerides.
2. *Muscle and adipose tissues*: Fructose is converted to fructose 6-phosphate by hexokinase in the muscles and adipose tissues.

Galactose Metabolism

The main pathway of galactose metabolism is the conversion of galactose into glucose. The galactose derived from the milk sugar is readily converted into glucose in the liver. The body requires galactose because it is a constituent of glycolipids (cerebrosides), chondromucoids and mucoprotein. Galactose is also required for the lactose synthesis in the mammary gland with help of the enzyme lactose synthetase. Galactosaemia, disorder of galactose
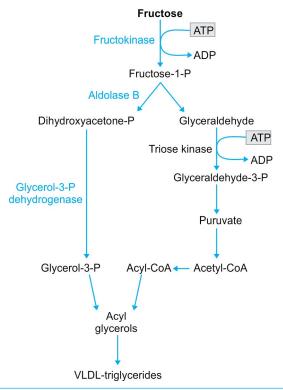
FIG. 4.20: Pathway of fructose metabolism
Abbreviations: ATP, adenosine triphosphate; ADP, adenosine diphosphate; VLDL, very low density lipoprotein
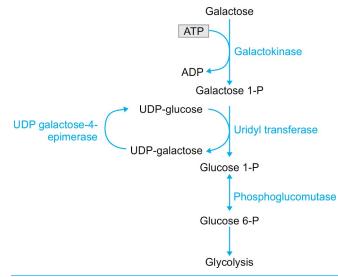

FIG. 4.21: Pathway of galactose metabolism
Abbreviations: ATP, adenosine triphosphate; ADP, adenosine diphosphate; UDP, uridine diphosphate

TABLE 4.4	Processes regulating the blood glucose levels
Processes which increase the blood glucose levels	Processes which reduce the blood glucose levels
• Absorption from the intestine • Breakdown of liver glycogen (glycogenolysis) • Gluconeogenesis	• Conversion to liver glycogen (hepatic glycogenesis) • Conversion to muscle glycogen (muscular glycogenesis) • Synthesis of fats (i.e. triglycerides) • Synthesis of glycoproteins such as nucleic acids (nucleoproteins), lactose, etc. • Loss of glucose in the urine.

metabolism, results from the deficiency of the enzyme galactose-I-phosphate-uridyl transferase. The metabolic pathway of galactose is described in **Figure 4.21**.

Steps

Phosphorylation of galactose by galactokinase results in the formation of galactose 1-phosphate.

Galactose 1-phosphate is then converted to glucose 1-phosphate via the enzyme galactose 1-phosphate uridyl transferase. Simultaneously in this reaction, UDP-glucose is converted into UDP-galactose. Conversion of UDP-galactose into UDP-glucose is a freely reversible reaction catalysed by UDP galactose-4-epimerase.

Glucose 1-phosphate is then isomerised into glucose-6-phosphate with help of the enzyme phosphoglucomutase.

Regulation of Blood Glucose

The concentration of glucose in the blood is the net result of two processes: (1) rate of glucose entrance into the bloodstream and (2) rate of glucose removal from the bloodstream **(Table 4.4)**. A balance between these two processes will help keep the blood sugar levels within normal limits.

Role of Liver in the Regulation of Blood Glucose Levels

Liver, being the centre of all metabolic activities plays a vital role in the regulation of blood glucose level. In liver there exists the developed mechanism for uptake of glucose from the blood, conversion of glucose to glycogen for storage (glycogenesis), release of glucose from glycogen (glycogenolysis) and de novo synthesis of glucose from non-carbohydrate precursors (gluconeogenesis). Glycogenesis in liver can occur from blood glucose or any substance capable of giving rise to pyruvate. Due to the presence of glucose-6-phosphatase, liver glycogen can contribute directly to blood sugar (gluconeogenesis).

Role of Extra-hepatic Tissues

Role of muscle: Muscle glycogen cannot directly cause an increase in the blood glucose levels due to the absence of the enzyme, glucose-6-phosphatase in the muscles. Glycogenolysis in muscle provides glucose to blood only through the formation of lactic acid, which is converted to glucose in the liver by Cori cycle.

Role of kidney: Kidney also exerts a regulatory effect on the blood glucose levels by reabsorbing glucose in the renal tubules. When the blood glucose levels rise above the renal threshold, excessive glucose may appear in the urine causing glycosuria.

Role of Hormones

Several hormones play an important role in maintaining the homeostatic mechanism for controlling the blood glucose levels. Out of the various hormones in the body, insulin is the only hypoglycaemic hormone (which results in a decline in the blood glucose levels) whereas others are hyperglycaemic hormones. These include various hormones such as glucagon, catecholamines (epinephrine, adrenaline), growth hormone, cortisol and to a lesser extent thyroxine, which are all anti-insulinic and recruit gluconeogenesis thereby causing an increase in the blood glucose levels. Growth hormone in particular increases hepatic gluconeogenesis and lipolysis. Blood sugar level is kept to normal due to insulin, by opposing the action of these hyperglycaemic hormones.

Insulin: Insulin in secreted by the pancreatic β cells and plays an important role in the regulation of blood glucose concentration. It is secreted into the blood in response to hyperglycaemia. Insulin helps reduce the blood glucose levels by increasing the transport of glucose across the cell membranes, increasing the glucose utilisation through the process of glycolysis, decreasing hepatic glycogenolysis and increasing glycogenesis.

Glucagon: Glucagon is secreted by the α-cells of the islets of Langerhans. Glucagon counter balances the action of insulin, which is secreted into the blood when the blood glucose level is high. Thus, glucagon secretion is stimulated by hypoglycaemia. Glucagon causes glycogenolysis by activating liver phosphorylase. Glucagon acts primarily on liver and does not affect glycogen breakdown in muscles. Glucagon also enhances gluconeogenesis from amino acids and lactate.

Epinephrine: Epinephrine stimulates glycogen breakdown in liver and muscle. The stimulation of glycogenolysis is due to its ability to activate phosphorylase. Epinephrine also inhibits muscle glycogen synthesis in liver and thus directs the production of increased blood glucose.

Adrenal cortex hormones: Adrenal cortex secretes gluco-corticoids, which lead to gluconeogenesis, which is the result of increased protein breakdown and stimulation of transaminase. It also inhibits glucose utilisation in extra-hepatic tissues.

Anterior pituitary hormones: Growth hormones and ACTH elevate the blood glucose levels. Growth hormones decrease glucose uptake by the tissues, whereas ACTH stimulates the secretion of hormones of the adrenal cortex.

Thyroid hormone: Thyroxine has a diabetogenic action. It increases blood glucose concentration by causing increased absorption of glucose from the intestines.

Metabolism of Fats
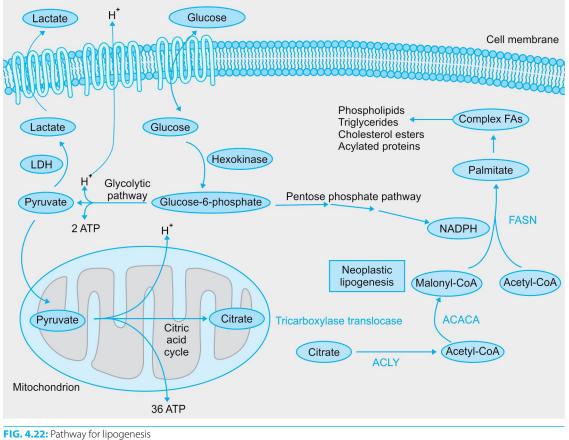
Lipogenesis

In the human body, nearly 90% of glucose is converted and stored in form of fat. Fatty acid synthesis takes place in the adipose tissues and to an extent in the liver. Fatty acids are derived from acetyl co-enzyme A (CoA). Several reductions of NADH, NADPH and FADH occur during this process **(Fig. 4.22)**. This pathway is reversible so that the fat can be metabolised at the times of energy requirement. Palmitate $[CH_3(CH_2)_{14}COOH]$ is the most common fatty acid in the body. It is also the first fatty acid to be synthesised in the body. The steps for fatty acid synthesis are as follows:

❏ *Formation of acetyl CoA and the citrate shuttle*: Fatty acids are synthesised in the cytoplasm from acetyl CoA molecules, which are produced from pyruvate through the action of the enzyme pyruvate dehydrogenase and through the mitochondrial β-oxidation of fatty acids. Since the synthesis of fatty acids occurs in the cytoplasm, the acetyl CoA molecules need to be transferred from the mitochondria to the cytoplasm with help of citrate shuttle. This comprises of the following steps:
   - Reaction of acetyl CoA with oxaloacetate to form citrate
   - Transportation of citrate to the cytosol with help of the enzyme tricarboxylase translocase
   - Conversion of citrate back to oxaloacetate and acetyl CoA in the cytosol with the help of an ATP-dependent enzyme ATP-citrate lyase.

One NADPH is generated for every molecule of acetyl CoA transported from the mitochondrion to the cytosol. This implies that 8 molecules of NADPH would be produced to generate one molecule of palmitate (16 carbon atom molecules).

❏ *Formation of malonyl CoA*: Some acetyl CoA molecules are carboxylated to form malonyl CoA in an irreversible two-step reaction, requiring bicarbonate and ATP. The reaction comprises of the following steps, both of which are catalysed by acetyl CoA carboxylase, a biotin-dependent enzyme:
   - Conversion of acetyl CoA to carboxybiotin by utilising one molecule of ATP
   - Transfer of the carboxyl group from biotin to the acetyl CoA to form malonyl CoA.

❏ *Formation of malonyltransferase by fatty acid synthetase using acetyl CoA or malonyl CoA as a substrate*: Fatty acid synthetase is a multienzyme protein, which synthesises palmitate from either acyl CoA or malonyl CoA. The enzyme fatty acid synthase is a multifunctional enzyme having three domains: (1) condensing unit (acetyl transferase, malonyltransferase and β-ketoacyl synthase); (2) reduction unit (β-ketoacyl reductase, dehydratase and enoyl reductase) and (3) the palmitate release unit (thioesterase).

FIG. 4.22: Pathway for lipogenesis
Abbreviations: LDH, lactate dehydrogenase; ACLY, ATP-citrate lyase; ACACA, acetyl CoA carboxylase; FASN, fatty acid synthase; NADPH, nicotinamide adenine dinucleotide phosphate (reduced); FA, fatty acid; ATP, adenosine triphosphate

❑ *The fatty acid elongation cycle*: This is the last stage of the fatty acid synthesis.
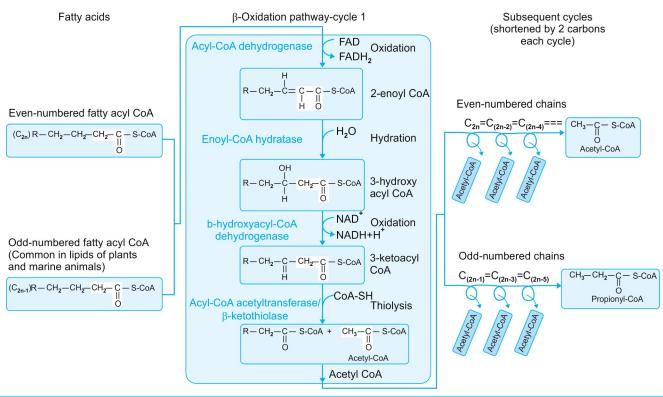
Fatty Acid β-Oxidation

Many tissues obtain energy through oxidation of fatty acids. Tissues such as heart derive only a limited energy from glucose and they mainly rely on the circulating free fatty acids as a source of energy. Some parts of kidneys also depend upon a source of fatty acid for the production of energy. Triacylglycerols (composed of three fatty acids and glycerol) are stored in the adipose tissues. The enzyme lipase becomes activated and cleaves three fatty acids from glycerol. The free fatty acids then travel to other tissues bound to albumin. The glycerol that is formed as a result of triglyceride hydrolysis does not travel to other tissues. Instead, it is either used for resynthesis of new triglycerides or alternatively, it is phosphorylated to 3-phosphoglycerate, component of the glycolytic pathway.

On reaching the cells where it is going to be utilised, the free fatty acids undergo a pathway called β-oxidation. Beta-oxidation can be defined as a process of four enzymic steps in which fatty acids are broken down in the mitochondrion to release acetyl CoA, NADH and $FADH_2$ **(Fig. 4.23)**. While acetyl CoA enters the citric acid cycle, NADH and $FADH_2$ act as cofactors in the electron transport chain. As a result of β-oxidation, the even-numbered fatty acyl CoA is reduced, hydrolysed, reduced again and finally hydrolysed to produce a molecule of acetyl CoA and a molecule of acyl CoA where the acyl group is two carbon atoms shorter than the original. The acetyl CoA molecule undergoes the TCA cycle, whereas the acyl CoA molecule undergoes the process again and again until the fatty acid molecule is completely degraded. Therefore, a molecule of palmitic acid having 16 carbon atoms is degraded into 8 molecules of acetyl CoA, which are further metabolised to produce the ATP molecules. Fatty acids with an odd number of carbons in the acyl chain are left at the end with propionyl-CoA (having 3 carbon atoms), which cannot enter another round of β-oxidation. The propionyl-CoA produced via this method is converted to succinyl-CoA, which then enters the Kreb's cycle.

In each cycle of fatty acid oxidation, fatty acyl is shortened by two carbon atoms, and $FADH_2$, NADH and acetyl CoA are generated. Each step in this four-step reaction is catalysed by an enzyme that is specific for the chain length of the acyl CoA, e.g. medium chain acyl CoA dehydrogenase oxidises fatty acyl molecules having 4–14 carbon atoms.

FIG. 4.23: Pathway for fatty acid β-oxidation

Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; FADH$_2$, flavin adenine dinucleotide (reduced); FAD, flavin adenine dinucleotide; H$_2$O, water

Steps

This process comprises of the following steps **(Fig. 4.23)**:

❏ *Oxidation of acyl CoA*: Acyl CoA is oxidised to 2-enoyl CoA with help of the enzyme acyl CoA dehydrogenase. In this process, FAD$^+$ is reduced to FADH$_2$.

❏ *Hydration of 2-enoyl CoA*: Hydration of 2-enoyl CoA to 3-hydroxy acyl CoA is catalysed by the enzyme enoyl CoA hydratase.

❏ *Oxidation of 3-hydroxyacyl CoA*: Oxidation of 3-hydroxy acyl CoA to 3-ketoacyl CoA is catalysed by the enzyme 3-hydroxy acyl CoA dehydrogenase. In this reaction NAD$^+$ is reduced to NADH.

❏ *Thiolytic cleavage of 3-keto acyl CoA*: Thiolytic cleavage of 3-keto acyl CoA to acetyl CoA is catalysed by the enzyme b-ketothiolase. This reaction requires the presence of reduced form of coenzyme A.

Beta-oxidation of Unsaturated Fatty Acids

Fatty acids can be of two types, saturated and unsaturated fatty acids. Saturated fatty acids have no carbon-carbon double bonds; the carbon atoms are saturated with the maximum number of hydrogen atoms. On the other hand the unsaturated fatty acids have one or more double bonds. Monounsaturated fatty acids have one carbon-carbon double bond, whereas the polyunsaturated fatty acid has more than one double bond.

Since the unsaturated fatty acid contains one or more double bonds, their oxidation requires two additional enzymes, an isomerase and reductase. Monounsaturated fatty acids, containing a single carbon-carbon double bond, require an isomerase to shift the position of the double bond so that it can act as a substrate for acyl CoA dehydrogenase. Polyunsaturated fatty acids have multiple double bonds and therefore require a reductase enzyme to generate a substrate, which can be acted on by enoyl CoA.

Regulation of Fatty Acid Metabolism

Fatty acid metabolism is predominantly regulated by the hormones, which control its activity in the following ways:

Glucagon and adrenaline: Hormones such as glucagon and adrenaline inactivate the enzyme acetyl CoA carboxylase in conditions where energy is required.

Insulin: Hormone such as insulin activates the enzyme acetyl CoA carboxylase in conditions where there is no requirement for energy.

When glucose levels are low during long periods between meals, the hormone glucagon secreted by the pancreas stimulates the activity of adipose lipase to release fatty acids from triglycerides. These free fatty acids can undergo β-oxidation to release energy. Epinephrine stimulates the same activity to release energy at the time

of fight or flight response. Since the β-oxidation of fatty acids occurs in the mitochondrial matrix, long-chain fatty acids must be actively transported from the cytoplasm into mitochondria by carnitine palmitoyltransferase or carnitine acyltransferase (a mitochondrial enzyme).

Comparison between Fatty Acid Oxidation and Synthesis

Lipogenesis and fatty acid β-oxidation are both tightly coordinated but distinct biochemical processes, with lipogenesis related to synthesis of lipids and β-oxidation to the breakdown of fats. Difference between fatty acid oxidation and fatty acid synthesis is described in **Table 4.5**.

Ketone Bodies

Acetyl CoA molecules produced as a result of β-oxidation can undergo a number of fates: in presence of adequate supply of oxaloacetate, acetyl CoA enters the citric acid cycle and combines with oxaloacetate to form citrate. However, in a fasting state, oxaloacetate molecules are diverted into the gluconeogenic pathway to facilitate the formation of glucose molecules. As a result, the acetyl CoA molecules enter another metabolic pathway to produce ketone bodies (water soluble molecules) as an alternative source of fuel to glucose during the periods of fasting. The ketones are water-soluble molecules produced from fatty acids in the liver. The two main ketone bodies in the body are acetoacetate and β-hydroxybutyrate.

The process of ketogenesis is described in **Figure 4.24.** The formation of ketone bodies occurs in the liver in three steps. The first step involves condensation of two molecules of acetyl CoA resulting in the formation of acetoacetyl CoA. This reaction is catalysed by the enzyme 3-ketothiolase. In the second step a condensation reaction occurs between acetoacetyl CoA, acetyl CoA and water molecules to form 3-hydroxy-3-methyl-glutaryl CoA (HMG CoA) and CoA. This reaction is catalysed by the enzyme HMG CoA synthase. In the final step, cleavage of HMG CoA occurs with help of the enzyme HMG CoA lyase to form acetyl CoA and acetoacetate.

Beta-hydroxybutyrate is formed by the reduction of acetoacetate in the mitochondria with the help of the enzyme β-hydroxybutyrate dehydrogenase. The equilibrium of this reaction is governed by the ratio of $NADH^+$ to NAD^+. In cases of predominance of NADH, β-hydroxybutyrate is preferentially formed. Acetoacetate can slowly decarboxylate to acetone, which results in a fruity breath in cases of diabetic or alcoholic ketosis.

Ketone bodies diffuse from liver into the circulation at the times of fasting or carbohydrate resistance. They play a vital role in cellular respiration as an alternative fuel source to glucose. Acetoacetate is preferentially used over glucose by the heart and renal cortex. The brain adapts to use acetoacetate during periods of starvation and in cases of diabetes mellitus. In cases of prolonged starvation, the

TABLE 4.5	Difference between fatty acid oxidation and fatty acid synthesis	
Characteristic	**Fatty acid oxidation**	**Fatty acid synthesis**
Intracellular location	Mitochondrion	Cytoplasm
Initial substrate	Fatty acyl CoA	Acetyl CoA or malonyl CoA
Coenzymes	NAD^+ and FAD^+	NADPH
Thioester linkage of intermediates	CoASH	Protein-SH (acyl carrier protein)
Bicarbonate dependence	No	Yes
Energy state favouring process	High ADP	High ATP
Citrate activation	No	Yes
Acyl CoA inhibition	No	Yes
Highest activity	Fasting or starvation	Fed

Abbreviations: NADPH, nicotinamide adenine dinucleotide phosphate (reduced); NAD, nicotinamide adenine dinucleotide; ATP, adenosine triphosphate; ADP, adenosine diphosphate; FAD, flavin adenine dinucleotide
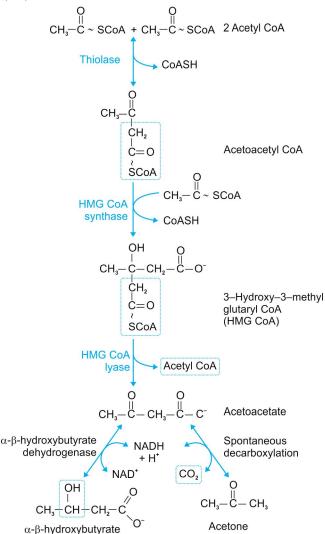

FIG. 4.24: The process of ketogenesis
Abbreviations: NADH, nicotinamide adenine dinucleotide (reduced); NAD, nicotinamide adenine dinucleotide; CO_2, carbon dioxide

brain derives nearly 75% of energy from acetoacetate. High acetoacetate levels in the blood inhibit lipolysis in the adipose tissues.

Prostaglandins

Prostaglandins are lipid autocoids derived from arachidonic acid. Arachidonic acid is liberated from phospholipids with the help of the enzyme phospholipase A_2. Each prostaglandin contains 20 carbon atoms including a 5-carbon ring. They are generated from arachidonate by the action of the enzyme cyclooxygenase and their biosynthesis is blocked by NSAIDs (nonsteroidal anti-inflammatory drugs). Synthesis of prostaglandins is described in **Figure 4.25**. Prostaglandins (e.g. prostacyclins, thromboxanes and leukotrienes) which function as locally acting hormones play a vital role in stimulating many of the body's processes such as inflammatory response, regulation of the blood flow to specific organs, modulation of ion transport across membranes, propagation of synaptic transmission, induction of sleep, etc.

Metabolism of Nucleotides

Nucleotide bases are classified into two: (1) purines and (2) pyrimidines, having a double-ring or a single-ring structure. Purines include adenine and guanine whereas pyrimidines include thymine, uracil and cytosine. Nucleotides such as adenine and cytosine are in both RNA and DNA. However, thymine occurs only in DNA and uracil in only RNA.

Purine Metabolism

Uric acid is a crystalline substance formed as a result of purine catabolism **(Fig. 4.26)**. Uric acid is insoluble, so it is excreted in form of its sodium salt, sodium urate. During pregnancy, the clearance of uric acid is increased, but this is balanced by increased by tubular reabsorption.

The concentrations decrease significantly by 8 weeks gestation and are maintained until about 24 weeks. Thereafter the concentrations increase so that by term they are greater than the prepregnancy values in the majority of patients.
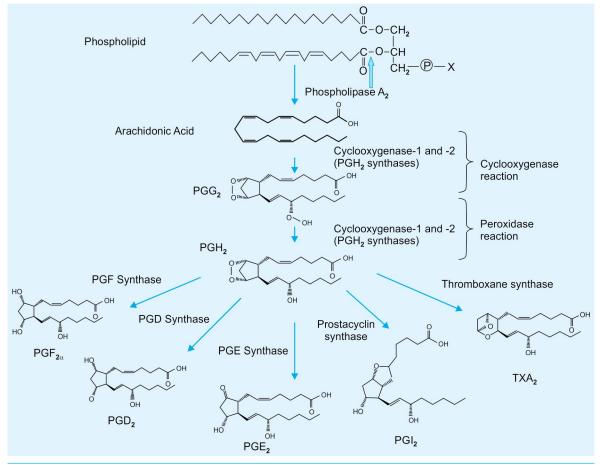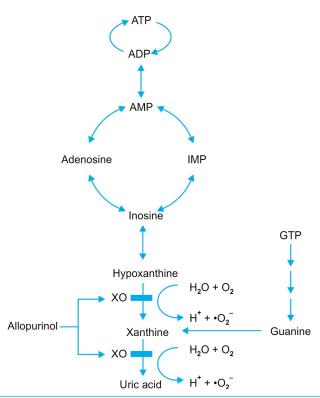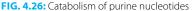

FIG. 4.25: Synthesis of prostaglandins
Abbreviations: PG, prostaglandin; TXA_2, thromboxane A_2

FIG. 4.26: Catabolism of purine nucleotides
Abbreviations: ATP, adenosine triphosphate; ADP, adenosine diphosphate; GTP, guanosine triphosphate; AMP, adenosine monophosphate; IMP, inosine monophosphate; XO, xanthine oxidase

FIG. 4.27: Basic structure of an amino acid

Urate is freely filtered at the glomerulus. An active anion exchange process in the early proximal convoluted tubule reabsorbs most of it. Reabsorption of uric acid occurs in the proximal convoluted tubule and then there is further secretion with the post-secretory reabsorption. It is excreted mainly in the urine, but some is excreted in the bile. It is not very soluble in body fluids. Of the total uric acid produced daily, the biliary and gastrointestinal tracts excrete 30% and the kidneys excrete 70%. Diuretics cause a shift of fluid from the intra- to the extravascular space and hence increase the serum concentrations of uric acid.

Normally the blood is almost completely saturated with uric acid. Therefore any slight impairment in purine catabolism is likely to result in hyperuricaemia. The excessive uric acid sometimes precipitates in form of crystals in the synovial fluid of joints resulting in a condition called gout. Allopurinol is a medication used for treatment of excessive uric acid in the blood and gout. It acts by blocking the enzyme xanthine oxidase.

Proteins, Peptides and Amino Acids

Proteins form the essential building blocks of the body. Each of the many cells in the body makes a unique set of proteins. Proteins are large biological macromolecules comprising of one or more long chains of amino acid residues. A linear chain of amino acid residue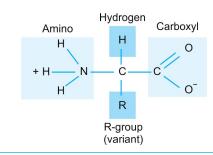s is known as polypeptide. Each protein molecule usually has one or more polypeptides. Short polypeptides containing less than 20–30 residues are known as peptides or oligopeptides. Peptide chains contain 2–30 amino acid residues, while polypeptide chains contain more than 30 to over 100 amino acid residues. Chains containing more than 100 amino acid residues are called proteins. Individual amino acids are binded together with the help of peptide bonds. Protein on average is composed of 16% nitrogen and 84% of carbon, hydrogen and sulphur combined.

There is a considerable variation in the types of proteins made by each cell. The proteins synthesised by a cell may play number of different roles. Some proteins may be intracellular and play a structural role. Other proteins may be secreted by a cell and used for supporting an extracellular structure, e.g. collagen. Another set of proteins may play an enzymic role. Most of the body's reactions: synthesis, degradation, energy production and storage are catalysed by enzymes. Some hormones and neurotransmitters are also composed of proteins.

Since all proteins are polymers of small nitrogen-containing molecules called amino acids, an adequate cellular supply of amino acids is vital for protein synthesis and cellular function. Amino acids are a large and diverse group of molecules, with each molecule containing an amino group ($-NH_2$), a carboxyl group ($-COOH$) and an organic side chain (the R group) which is unique to each amino acid **(Fig. 4.27)**. Most of the biologically important amino acids are the α-amino acids. In this type of amino acids, the amino group is attached to α carbon both the amine and carboxylic acid groups are attached to the first (alpha-carbon atom).

Amino acids not only serve as building blocks for proteins, they also serve as precursors for other important compounds such as nucleotides, porphyrins, neurotransmitters and hormones. Amino acids can be of two types: essential and non-essential. There are a total of 20 amino acids in the body **(Fig. 4.28)**, of which nine are the essential amino acids.

Essential amino acids: Essential amino acids are those, which cannot be synthesised de novo in the body. These need to be supplied in the diet. These nine essential amino acids, particularly important when considering nutrition, are listed in **Table 4.6**. Of these nine essential amino acids, methionine is particularly important.
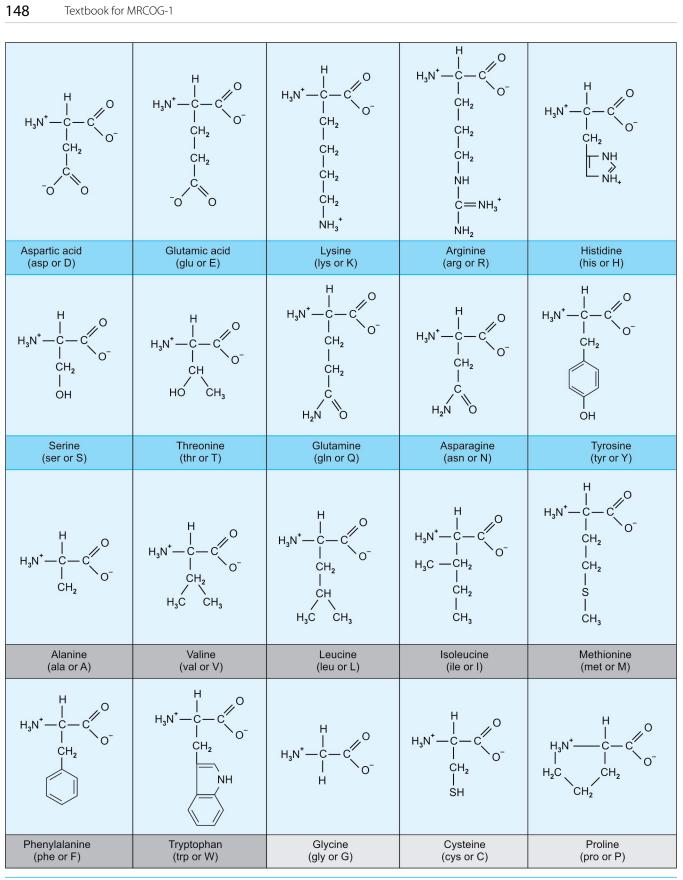
FIG. 4.28: Structure of the various amino acids in the body

TABLE 4.6	Various types of amino acids in the body
Essential amino acid	**Non-essential amino acid**
• Valine • Tryptophan • Lysine • Leucine • Isoleucine • Phenylalanine • Threonine • Methionine • Histidine	• Alanine • Aspartic acid • Asparagine • Arginine • Glutamine • Cysteine • Glutamic acid • Glycine • Proline • Serine • Tyrosine

Methionine is a neutral, genetically coded, sulphur-containing essential amino acid, which can cross the placenta and reach the foetus. It is particularly important because it supplies sulphur ions which help in maintaining skin tone, healthy hair, and strong nails. It is usually converted to homocysteine and is reabsorbed in the proximal convoluted tubule in the kidneys.

Non-essential amino acids: These amino acids can be synthesised de novo in the body. Therefore, these 11 amino acids do not require to be taken in the diet.

Structure of Proteins

Proteins must form a three-dimensional structure to function. This is described in terms of primary, secondary, tertiary and quaternary structures.

Primary structure: A protein's primary structure is the linear sequence of amino acids joined by peptide bonds. This bond is formed by linking the carbonyl group from one amino acid to amino group of another amino acid.

Secondary structure: Secondary structure results due to the local folding of the peptide chain as a result of hydrogen bonding between the carbonyl oxygen group of one peptide bond and the amide hydrogen of a neighbouring peptide bond. Secondary structures can be of two types: α helix (a right-handed coil of nearly 4–40 amino acid residues on the polypeptide chain) and the β-sheet (two segments of a polypeptide chain overlapping one another with rows of hydrogen bonds between them).

Tertiary structure: The three-dimensional, folded and biologically active structure of protein is known as the tertiary structure. It is determined and stabilised by the presence of the following: side chain functional groups, covalent disulphide bonds, hydrogen bonds, salt bridges, hydrophobic interactions, etc.

Quaternary structure: This implies complex assembly of two or more peptide chains which are held together by either covalent or non-covalent interactions, e.g. haemoglobin, a tetrameric protein containing two α and two β chains and a haem prosthetic group.

Purification and Analysis of Proteins

Various techniques are available for separation, analysis and purification of proteins. Various methods separate proteins based on their size. Some of these are as follows:

Gel permeation chromatography: This involves the use of a bed of resin beads containing pores of predetermined size. While some smaller proteins diffuse through these pores, the larger ones are excluded out.

Gel electrophoresis: In this process, an agarose or a polyacrylamide support is used and the protein solution is exposed to an electric field. The proteins move on the basis of their mass/charge ratio, in absence of a detergent. If a detergent is added, the proteins will move only on the basis of their size.

Ion exchange chromatography: This process involves the use of resins that contain negatively or positively charged groups on their surface. Proteins contain negatively charged carboxyl groups and positively charged amino groups. The proteins bind to the resin based on their unique pattern of charge and then can be eluted by changing either the pH or the salt strength of the buffers used. Different proteins can be separated based on their pattern of charge.

Affinity chromatography: In this method a chemical agent is coupled to a bed of support beads. The binding agent gets coupled with a high specificity to the protein which is to be purified. On passing the protein mixture through the bed, only the protein in question binds to the chemical agent. After washing off the other proteins, this protein can be eluted by changing the pH or salt concentration. This technique is quite powerful and complete purity can be achieved in a single step.

Protein Synthesis

Protein synthesis refers to the generation of new proteins by the biological cells. Gene expression is the process by which the information encoded in the gene is converted into functional gene product or an instructional document (RNA) that is used for protein synthesis. The protein synthesis occurs in the ribosomes, which are attached to RER. Protein synthesis involves two steps:
1. Transcription
2. Translation.

The sequence of amino acids in a protein is coded by the sequence of nucleotides in the mRNA molecule. The mRNA is a transcript of the DNA and therefore reflects the nucleotide sequence of the gene for that protein. Each amino acid is encoded by a triplicate sequence of nucleotides called a codon. There are 64 possible codons and each amino acid may be encoded by more than one codon.

Transcription of Genetic Code

The word transcription means copying. It indicates the copying of genetic code from DNA to RNA. Thus, the first stage in the protein synthesis is transcription of genetic code, which occurs within the nucleus. It involves the formation of mRNA and simultaneous copying or transfer of information from DNA to mRNA. The mRNA enters the cytoplasm from the nucleus and activates the ribosome resulting in protein synthesis. The formation of mRNA from DNA is facilitated by the enzyme RNA polymerase.

Translation of Genetic Code

Translation is the process by which protein synthesis occurs in the ribosome of the cell under the direction of genetic instruction carried by mRNA from DNA. In other words, it can be described as the process by which the mRNA is read by ribosome to produce a protein. This involves the role of other two types of RNA, namely tRNA and rRNA. Translation occurs in three phases: (1) initiation, (2) elongation and (3) termination.

The mRNA produced during transcription moves out of nucleus into the cytoplasm. Now, a group of ribosomes called polysome gets attached to mRNA. The sequence of codons in mRNA are exposed and recognised by the complementary sequence of base in tRNA. The complementary sequence of base is called anticodon. According to the sequence of bases in anticodon, different amino acids are transported from the cytoplasm into the ribosome by tRNA that acts as a carrier. With the help of rRNA, the protein molecules are assembled from amino acids. Ribosomes co-ordinate and catalyse the polymerisation of amino acids to form polypeptide chains. Ribosomes consist of two subunits: large and small, which contain ribosomal ribonucleic acid and the numerous proteins required for each phase of translation. Newly synthesised polypeptide chains then undergo post-translational modifications (e.g. chemical modification, folding, proteolytic cleavage, etc.) to produce the final functional protein.

Protein Metabolism

Most proteins are too large to be filtered by the glomerular membrane in the kidneys and to be excreted in the urine. Therefore the major mechanism for the removal of unwanted proteins is proteolysis, i.e. degradation of proteins into their respective amino acids. Proteolysis is also important for the utilisation of dietary proteins, which cannot be absorbed and utilised by the body until they are broken down into their respective amino acids. The amino acids released through the process of proteolysis can undergo one of the following fates **(Fig. 4.29)**:

❏ The essential amino acids may be used for the synthesis of proteins and other nitrogenous compounds
❏ They may be degraded further and used for energy metabolism.

Amino acids liberated as a result of proteolysis are small enough to be filtered by the kidney; however, the tubular cells reabsorb most. Human beings have no mechanism for storing amino acids, so excessive amino acids need to be degraded in the body. Degradation of amino acids involves removal of the amino group (deamination) to
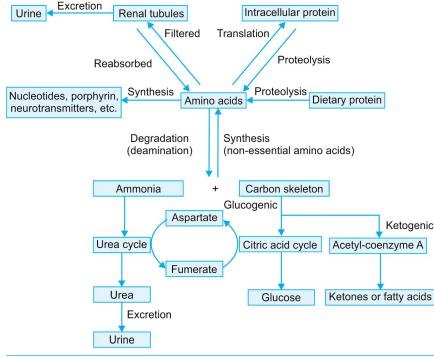

FIG. 4.29: Mechanism of protein turnover

produce ammonia and a carbon skeleton. The ammonia is removed through the urea cycle and excreted out through the urine. On the other hand, the carbon skeleton is used for providing a source of energy to the cell. The carbon skeletons of amino acids can be converted into one of the following intermediates **(Fig. 4.30)**:

❑ Pyruvate
❑ Oxaloacetate
❑ Fumarate
❑ Succinyl coenzyme A
❑ Alpha-ketoglutarate
❑ Acetyl coenzyme A
❑ Acetoacetyl coenzyme A.

Amino acids, which are degraded to acetyl CoA, or acetoacetyl CoA are called ketogenic amino acids because they are ultimately metabolised to ketones or fatty acids. The amino acids degraded to other intermediates are called glucogenic amino acids because they are potentially metabolised to glucose. Some amino acids may be degraded to more than one of the seven intermediates and therefore are both glucogenic and ketogenic. Different types of amino acids based on the fate of degradation of their carbon skeleton are tabulated in **Table 4.7**.

Urea Cycle

The steps of urea cycle are illustrated in **Figure 4.31**. The first step in the urea cycle is the generation of carbamoyl phosphate from ammonia and bicarbonate with the help of the enzyme carbamoyl-phosphate synthetase I. Carbamoyl phosphate then reacts with ornithine and aspartate to produce arginosuccinate, catalysed by the enzyme arginosuccate synthetase. Arginosuccinate is subsequently cleaved to arginine and fumarate with help of arginosuccinate lyase. Fumarate is recycled back to aspartate by the citric acid cycle. Arginine is hydrolysed to urea resulting in the regeneration of ornithine.

Cell Signalling and Second Messengers

Signal transduction pathways allow cells to respond to the environmental signals. Second messengers are molecules, which help in relaying signals from the receptors present on the surface of the cells to the target molecules inside the cells. They form a component of the cell signalling pathways and are responsible for greatly amplifying the signal strength. Through these pathways, a signal is amplified. Removal or degradation of second messengers helps in terminating the cellular response. There are four classes

TABLE 4.7	Types of amino acids based on the product of degradation	
Glucogenic amino acids	**Ketogenic amino acids**	**Amino acids which are both ketogenic and glucogenic**
• Arginine	• Leucine	• Alanine
• Asparagine	• Lysine	• Cysteine
• Aspartate	• Tryptophan	• Glycine
• Glutamate		• Isoleucine
• Glutamine		• Phenylalanine
• Histidine		• Serine
• Methionine		• Threonine
• Proline		• Tyrosine
• Valine		
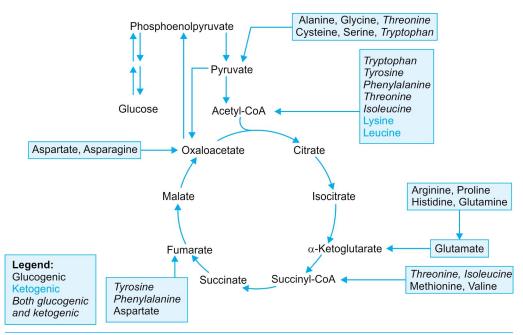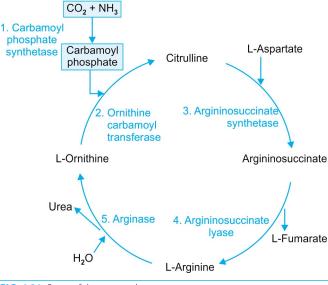
FIG. 4.30: Catabolism of proteinogenic amino acids

TABLE 4.8	Second messengers		
Second messenger	**Activation**	**Effects**	**Deactivation**
Cyclic AMP	Produced from ATP by adenylate cyclase	Activation of protein kinase A	Metabolised to 5'-AMP by cyclic phosphodiesterase
Inositol 1, 4, 5 triphosphate	Produced from PIP_2 by the action of phospholipase C	Opens calcium ion channels on cell membrane to increase intracellular calcium	Converted to derivatives
Diacylglycerol	Produced from PIP_2 by the action of phospholipase C	Activates protein kinase C	Hydrolysed to glycerol and fatty acids
Calcium	Enters cell through open calcium ion channels	Activates calmodulin and changes cell membrane potential	Sequestered into intracellular stores or pumped out of the cell

Abbreviations: ATP, adenosine triphosphate; AMP, adenosine monophosphate; PIP_2, phosphatidylinositol-2;

FIG. 4.31: Steps of the urea cycle
Abbreviations: CO_2, carbon dioxide; NH_3, ammonia

FIG. 4.32: Different classes of second messengers
Abbreviations: cAMP, cyclic adenosine monophosphate; cGMP, cyclic guanosine monophosphate; IP_3, inositol triphosphate; NO, nitric oxide; H_2S, hydrogen sulphide; CO, carbon monoxide

of second messengers: cyclic nucleotides (cyclic AMP, cyclic GMP); membrane lipid derivatives; Ca^{2+} ions, nitric oxide, inositol 1, 4, 5-triphosphate and diacylglycerol, etc. Second messengers essentially serve as chemical relays from the plasma membrane to the cytoplasm, thus carrying out intracellular signal transduction. Various second messengers are described in **Table 4.8** and **Figure 4.32**.

Cyclic AMP

This is a second messenger synthesised from ATP through the enzyme adenylyl cyclase. Binding of the hormone to its receptor activates a G-protein, which in turn activates adenylyl cyclase **(Fig. 4.33)**. This results in an appropriate response through either of the one or both the mechanisms:

❏ *Using protein kinase A*: A cAMP-dependent protein kinase which phosphorylates target proteins
❏ *Using a protein called CREB*: CREB (cAMP response element binding protein) and the resultant complex controls gene transcription.

Cyclic AMP is metabolised to 5'-AMP by phosphodiesterase, which helps in terminating its action. On the other hand, cAMP produces its action by binding to the specific
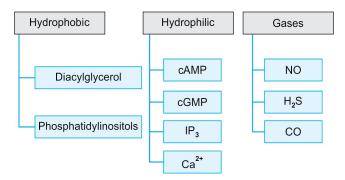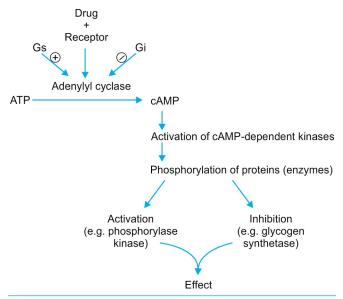

FIG. 4.33: Cyclic AMP second messenger system
Abbreviations: Gs, stimulatory G-protein; Gi, inhibitory G-protein

locations on the regulatory units of the protein kinase, causing them to dissociate from the tetramer, thus activating the catalytic units so they can perform their function. The cyclic AMP mechanism been shown to be an intracellular hormonal mediator for the following hormones:

❏ Glucagon
❏ Parathyroid hormone
❏ Secretin

- Thyroid-stimulating hormone (TSH)
- Vasopressin
- Adrenocorticotropin
- Luteinising hormone
- Follicle-stimulating hormone
- Catecholamines.

G Proteins

G proteins are cytoplasmic proteins intimately related to the cell surface receptor and are involved in the cell signalling processes occurring with hormone receptors. The G proteins may be stimulatory or inhibitory and notably are involved in the regulation of adenylate cyclase.

Cyclic GMP

Cyclic GMP (cyclic guanosine monophosphate) is synthesised from the nucleotide GTP through the enzyme guanylyl cyclase. Nitric oxide stimulates the synthesis of cyclic GMP. Many cells contain a cGMP-stimulated protein kinase that contains both catalytic and regulatory subunits. Some of the effects of cGMP are mediated through protein kinase G (PKG). cGMP serves as the second messenger for nitric oxide and the response of the rods of the retina to light.

Phosphatidylinositol-derived Second Messengers

Phosphatidylinositol (PI) is a negatively charged phospholipid and a minor component of eukaryotic cell membranes. The inositol can be phosphorylated to form the following molecules which can serve as second messengers:
- Phosphatidylinositol-4-phosphate (PIP)
- Phosphatidylinositol-4,5-bis phosphate (PIP_2)
- Phosphatidylinositol-3,4,5-trisphosphate (PIP_3)

Intracellular enzyme phospholipase C hydrolyses PIP2 which is found in the inner layer of the plasma membrane, resulting in the production of two products: Diacylglycerol (DAG) and inositol-1,4,5-trisphosphate (IP_3). Mechanism of action of phosphatidylinositol second messenger system is described in **Figure 4.34**.

Diacylglycerol

Diacylglycerol stimulates protein kinase C activity by greatly increasing the affinity of the enzyme for calcium ions. Protein kinase C phosphorylates specific serine and threonine residues in target proteins. Known target proteins include calmodulin, the glucose transporter, HMG-CoA reductase, cytochrome P450, etc.

Inositol Triphosphate

This soluble molecule diffuses through the cytosol and binds to receptors on the ER causing the release of calcium ions into the cytosol. The rise in intracellular calcium triggers the response by turning on the appropriate genes in the nucleus.
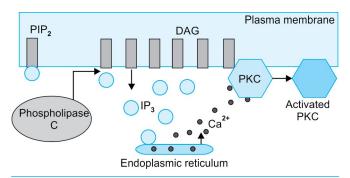

FIG. 4.34: Phosphatidylinositol second messenger system
Abbreviations: PIP, phosphatidylinositol-4,5-bis-phosphate; DAG, diacylglycerol; IP_3, inositol triphosphate; PKC, phosphofructokinase

This mechanism has been shown to be an intracellular hormonal mediator for the following:
- Peptide and protein hormones like vasopressin, TSH, etc.
- Neurotransmitters like gamma-aminobutyric acid (GABA).

Calcium Ions

Many cells respond to extracellular stimuli by altering their intracellular calcium concentration. Ca^{++} acts as a second messenger in two ways:
1. Binding to an effector molecule, such as an enzyme, thereby activating it
2. Binding to an intermediary cytosolic calcium-binding protein such as calmodulin.

The binding of Ca^{++} causes profound conformational changes in calmodulin that increase calmodulin's affinity for its effector molecules. "Calmodulin", when activated, causes contraction of smooth muscles.

Nitric Oxide

Nitric oxide (NO) is a free radical which is synthesised from arginine and oxygen by NO synthase in many tissues but particularly the endothelium.

Its half-life is in seconds and its effects are mediated through cGMP production, which functions as a second messenger. NO activates soluble guanylyl cyclase, which when activated produces another second messenger, cGMP. It produces vasodilatation through smooth muscle relaxation, this process being impaired in endothelial dysfunction. NO production is increased in normal and even more so in abnormal pregnancies.

Nutritional Physiology in Health and Disease

Biochemistry of Vitamins

Vitamins are organic dietary constituents necessary for life, health and growth that do not function by supplying the body with energy. Different types of vitamins include

A, B, C, D, E and K. These can be classified as water soluble (vitamins B and C) and fat soluble (vitamins A, D, E and K). All these vitamins have been briefly described next.

Vitamin A (Retinol)

Deficiency of vitamin A leads to night blindness and xerophthalmia. During pregnancy the requirement of vitamin A is approximately 1,000 g/day. Hypervitaminosis A is characterised by anorexia, headache, hepatosplenomegaly, patchy loss of hair and hyperostosis. It is stored in the liver.

Vitamin B Complex

Vitamin B complex group of vitamins include B_1 (thiamine), B_2 (riboflavin), B_3 (niacin or nicotinic acid), B_5 (pantothenic acid), B_6 (pyridoxine), B_7 (biotin), B_9 (folic acid) and B_{12} (various cobalamines commonly known as cyanocobalamin and methylcobalamin).

Deficiency of vitamin B_1 (thiamine) leads to impaired collagen formation. The body contains only 30 mg of vitamin B_1 and the average adult requirement of vitamin B_1 is 1–1.5 mg/day. As with other water-soluble vitamins, vitamin B crosses the placenta by active mechanisms, thereby resulting in higher concentrations in the foetus. Vitamin B_6 (pyridoxine) requirement in pregnancy is 2.5 mg/day and in non-pregnant adult, the requirement is 2 mg/day. Niacin is synthesised in the body from tryptophan.

Vitamin B_{12} is absorbed mainly in the lower ileum, aided by gastric intrinsic factor. It is essential for the metabolism of folic acid in the human. Its deficiency results in macrocytic anaemia. Animal foodstuffs are the main source of B_{12}. Vegetables alone are an inadequate source of vitamin B_{12}. Therefore, its deficiency is common amongst strict vegetarians.

Folic Acid

The normal Western diet contains approximately 500–700 g of folic acid per day, of which 10–100% may be impaired by cooking because it is not heat stable. Folic acid is rapidly absorbed from the upper part of small intestine. In stagnant loop syndrome, the ability of organisms to manufacture folic acid results in an elevated plasma concentration.

Tetrahydrofolic acid: The coenzyme tetrahydrofolic acid is derived in humans from the B-complex vitamin and folic acid. Tetrahydrofolic acid itself is synthesised in the cell from folic acid with the help of an enzyme, folic acid reductase. Methotrexate prevents the production of tetrahydrofolate from folic acid in all tissues.

Tetrahydrofolate is essential for both purine and pyrimidine biosynthesis. Coenzymes synthesised from tetrahydrofolic acid are required at two stages in the biosynthesis of purines (adenine and guanine) and at one stage in the synthesis of pyrimidines (thymine, cytosine and uracil). Tetrahydrofolate derived coenzymes are also required for one-carbon transfer reactions.

Malabsorption diseases such as tropical sprue and coeliac disease can both cause folate deficiency as a result of chronic diarrhoea and malabsorption.

Tropical sprue is characterised by features such as inflammation and flattening of the intestinal villi. The diagnosis of tropical sprue is usually confirmed on intestinal biopsy. The cause of the disease presently remains unknown. Possible causes include bacterial, viral, parasite and amoeba infection. This disease can be associated with symptoms such as chronic diarrhoea, steatorrhoea (foul-smelling, greasy stools), weight loss, abdominal cramps, etc. This may be ultimately responsible for causing malnutrition through multiple vitamin deficiencies and deficiency of minerals including calcium.

Another malabsorption syndrome, which may result in malnutrition and multiple vitamin deficiencies, includes coeliac disease or non-tropical sprue or gluten intolerance. Due to similar symptomatology, coeliac disease can be sometimes confused with tropical sprue. However, the pathophysiology of the two diseases is different. Coeliac disease is an autoimmune disease triggered by the presence of gluten in the diet, resulting in flattening and inflammation of the intestinal villi and mucosa. The diagnosis is confirmed by endoscopy and intestinal biopsy. Blood tests can also be performed showing presence of antibodies to gluten.

Vitamin C

Citrus food and leafy green vegetables are rich in vitamin C, while animal sources contain only traces. It is destroyed by heating. The eye and the adrenal glands contain large quantities of vitamin C. Vitamin C deficiency is associated with impaired wound healing. Excess vitamin C can lead to the formation of oxalate stones in the urinary tract.

Vitamin D

The dietary requirement of vitamin D is 10 mg/day. Its deficiency leads to rickets. Vitamin D is synthesised in the body in presence of sunlight. This process has been described in details in chapter 11. Similar to a classic steroid hormone, vitamin D acts by attaching to the cytosolic receptors. Vitamin D is bound to a transport protein in the circulation. Vitamin D is stored in the body fat and is absorbed from the small intestine. Being a fat soluble vitamin, it is poorly absorbed in cases of obstructive jaundice and also in cases of pancreatic disease where there is a deficiency of pancreatic lipase.

The active form of vitamin D, calcitriol $(1, 25\text{-}(OH)_2D_3)$ acts on the intestines, kidneys and bone to increase calcium and phosphate values in serum. On the other hand, $24,25(OH)_2D_3$ is a relatively inactive form of vitamin D. The main site of action is in the intestine, where calcitriol stimulates calcium and phosphate absorption. In the kidney, calcitriol promotes renal phosphate resorption in the proximal convoluted tubule and calcium

resorption in the distal convoluted tubule. Calcitriol also has antiproliferative effects and may cause differentiation in some tumour cell lines.

Vitamin E

Dietary requirement of vitamin E is 10 mg/day. It is present in most food stuffs. Its deficiency may cause intrauterine foetal death in animals. However, this has yet not been proven in humans. It potentiates the action of coumarin anticoagulants. Presently, there is no evidence that vitamin E increases virility, or plays any role in the treatment of infertility or recurrent abortion.

Vitamin K

Vitamin K is mainly found in the green leafy vegetables. It exists in two forms, K_1 and K_2. Both are derivatives of the cyclic structure naphthoquinone. Although vitamin K accumulates initially in the liver, its hepatic concentration soon declines rapidly. Vitamin K_1 is synthesised by the plants and is found in the highest amounts in green leafy vegetables. Vitamin K_1 can be converted into vitamin K_2 by the bacteria in the large intestine. In adults, no external supplements are necessary. The only exceptions are the pregnant patients on antiepileptic drugs who require vitamin K in the last month of pregnancy and the newborn babies whose colons have not yet been colonized by bacteria.

Disorders Related to Nutrition

Disorders related to the deficiency of various vitamins are tabulated in **Table 4.9**.

Energy Released by Different Food Products

Carbohydrates release 4.4 Kcal/g, proteins 4.2 Kcal/g and fats release 7 Kcal/g. Therefore, fats yield approximately 125% more energy than 1 g of carbohydrates when metabolised in the body. Protein, when metabolised in the body yield about 40% less energy because urea derived from protein is excreted in the urine and its free energy is

TABLE 4.9	Deficiency disorders
Nutrient substance	**Deficiency disorder**
Vitamin A	Night blindness, xerophthalmia
Thiamine	Beriberi
Cyanocobalamin (B_{12})	Pernicious anaemia and macrocytic anaemia
Niacin (B_3)	Pellagra
Ascorbic acid	Scurvy
Vitamin D	Rickets, osteomalacia

not released to the body. Carbohydrate, metabolised in the body, yields about the same energy as 1 g of protein. The free energy released by carbohydrate and protein in the body is therefore, similar. The energy released upon metabolism of various food products is usually stored by the body following a meal and later mobilised for metabolism on the basis of demand. Glycogen is the first resource, which is mobilised, followed by fats. Proteins are utilised for energy production only in cases of extreme starvation or shock.

The Specific Dynamic Action of Food

The specific dynamic action of food is the increase in metabolic rate that results from ingestion of food. It persists for about 6 hours. It is due mainly to the additional energy expended on processing absorbed material for detoxification, metabolism and storage. This results in about 30% of the energy value of ingested protein being unavailable for other purposes mainly because of the energy required to deaminate amino acids. It also results in about 4% of the energy value of ingested fat and 6% of carbohydrate being unavailable for other purposes.

Basal Metabolic Rate

Basal metabolic rate (BMR) can be described as the amount of energy expressed in calories that a person requires for the proper functioning of the body at rest. It is usually determined at rest after 12–14 hours following the last meal. BMR of an adult man can vary from 1,000 Kcal to 2,500 Kcal depending on the variation in fat-free mass.

Choose the Single Best Answer (SBA)

Q 1. **Which of the following is true regarding carbohydrate metabolism?**

A. The principal carbohydrate used in body metabolism is galactose

B. Glycolysis is the process of glycogen formation

C. The pentose shunt is active in all cells of the body except red blood cells (RBCs)

D. The tricarboxylic acid (TCA) cycle is the common pathway for the oxidation of dietary carbohydrates, fats and proteins to CO_2 and H_2O

E. Acetoacetic acid and beta-hydroxybutyric acid are types of fat

Q 2. **Which of the following is not true regarding the conversion of glucose to lactic acid?**

A. Is an irreversible process in skeletal muscles

B. Occurs in a single enzymatic reaction

C. Is inhibited by high cellular concentrations of ATP

D. Is the only pathway for the synthesis of ATP in the red blood cells

E. Occurs in skeletal muscle when the availability of oxygen is limited

Q 3. **Regarding the oxidation of pyruvate to carbon dioxide, which of the following is false?**

A. Can occur under anaerobic conditions

B. Involves intermediates that are also involved in amino acid catabolism

C. Is impaired in thiamine deficiency states

D. Is regulated by the concentration of acetyl CoA in the cell

E. Occurs exclusively in mitochondria

Q 4. **Which of the following statements is true regarding Kreb's cycle?**

A. Alpha-ketoglutarate is a five carbon molecule

B. Kreb's cycle can function under anaerobic conditions

C. Oxidative phosphorylation occurs within the cytoplasm

D. Only carbohydrates and fats are oxidised in Kreb's cycle

E. Pyruvate condenses with oxaloacetate to form citrate

Q 5. **Which of the following is not true concerning uric acid?**

A. It is excreted unchanged in the urine

B. Is formed from the breakdown of purines

C. Is reabsorbed in the proximal renal tubule

D. Serum concentrations are increased during thiazide diuretic therapy

E. Serum concentrations are raised during normal pregnancy

Q 6. **True regarding uric acid is:**

A. Is the end-product of pyrimidine metabolism in humans

B. Is excreted mainly in the bile

C. Is highly soluble in body fluids

D. The normal blood level is 4 mg/dL

E. Its plasma levels change significantly during pregnancy

Q 7. **Which of the following is an essential amino acid?**

A. Arginine

B. Methionine

C. Glycine

D. Tryptophan

E. Valine

Q 8. **Which of the following is true regarding folic acid?**

A. Is a fat-soluble vitamin

B. Requires gastric intrinsic factor for its absorption

C. Is necessary for nucleic acid synthesis

D. Is heat-stable

E. Is involved in the tricarboxylic acid (Kreb's) cycle

Q 9. **Which of the following is not true regarding the hormonal influence upon carbohydrate metabolism?**

A. Adrenal glucocorticoids enhance the effect of glucagon

B. Catecholamines increase blood glucose concentration

C. Growth hormone increases hepatic glucose output

D. Growth hormone inhibits mobilisation of free fatty acids from adipose tissue

E. Thyroid hormones increase blood glucose concentration

Q 10. **True statement regarding prostaglandins is:**

A. It is synthesised from cholesterol

B. Prostaglandins are small polypeptides

C. They are secreted by the pituitary gland

D. They are secreted by the prostate gland

E. They are rarely associated with gastrointestinal side effects

Q 11. **Which of the statement is correct regarding the various cell organelles?**

A. Lysosomes contain enzymes capable of digesting cells and cellular material

B. Ribosomes "Read" the mRNA and build proteins

C. Golgi apparatus is involved in the modification of lipids and proteins with storage of material prior to export out of the cell

D. All the above

E. None of the above

1. D	2. B	3. A	4. A	5. E	6. D	7. B	8. C	9. D	10. D	11. D

Q 12. Which of the following is true regarding glycogen?

A. It is a branched polymer of glucose
B. It is mainly stored in the muscles
C. It is broken down by glucose-6-phosphatase
D. Its synthesis is stimulated by adrenaline
E. Levels in the blood stream are highest in the morning

Q 13. Which of the following is not true regarding fat metabolism?

A. Fat cannot be metabolised anaerobically
B. Brain can utilise fat as a source of fuel
C. Oxidation of fatty acids occurs in the mitochondria
D. Beta-oxidation of fatty acids is controlled by the supply of substrates
E. Liver can convert fatty acids into ketone bodies

Q 14. Which of the following is not true regarding arachidonic acid?

A. It is a second messenger
B. It is a fatty acid
C. It is a precursor of thromboxane
D. It is inhibited by aspirin
E. It is converted into prostaglandins

Q 15. Which of the following is not true regarding the synthesis of steroids?

A. Progesterone is a precursor of pregnenolone
B. Cholesterol is the precursor of all other steroids
C. Corticosterone is converted to aldosterone
D. Pregnenolone is formed in the cellular mitochondria
E. Testosterone is a precursor of oestradiol

Q 16. Which of the following is not true regarding prostaglandins?

A. They are hydrophobic
B. They are synthesised from arachidonic acid
C. NSAIDs (non-steroidal anti-inflammatory drugs) act as their antagonists
D. Comprise of 18 carbon atoms
E. Bind to G-protein coupled receptors

Q 17. Which of the following enzymes act at the rate-limiting step of the glycolysis pathway?

A. Glucokinase
B. Hexokinase
C. Pyruvate dehydrogenase
D. Phosphofructokinase
E. Phosphoglucose isomerase

Q 18. Overall product of the glycolysis pathway is

A. Acetyl coenzyme A
B. Glucose molecules
C. Pyruvate molecules
D. 2 NADH + 2 ATP
E. 1 NADH + 1 ATP

Q 19. Net yield per glucose molecule undergoing glycolysis is:

A. One NADH molecule and one ATP molecule
B. Two NADH molecules and two ATP molecules
C. Two NADH molecules and one ATP molecule
D. One NADH molecule and two ATP molecules

Q 20. Which of the following is not an intermediate product of the citric acid cycle?

A. Alpha-ketoglutarate
B. Acetyl coenzyme A
C. Citrate
D. Oxaloacetate
E. Succinyl coenzyme A

Q 21. Which of the following laboratory technique is used for detecting DNA sequences?

A. Eastern blotting
B. Western blotting
C. Southern blotting
D. Western blotting
E. Northwestern blotting

Q 22. Which of the following best describes the function of low-density lipoproteins?

A. Transportation of cholesterol from the body's tissues to the liver
B. Transportation of cholesterol from the liver to the tissues around the body
C. Transportation of chylomicrons from liver to the tissues around the body
D. Transportation of triglycerides from the liver to other tissues around the body for storage
E. Transportation of triglycerides from the intestine to other tissues for storage

Q 23. Which of the following is correct regarding vitamin C?

A. Is found only in animal foodstuffs
B. Is not destroyed by heating
C. There are normally large stores in the pancreas
D. Unimpaired wound healing is one of the characteristic features of severe vitamin C deficiency
E. Excess vitamin C can lead to the formation of oxalate stones in the urinary tract

Q 24. Which of the following is not true regarding metabolism?

A. The metabolic rate is the amount of energy liberated per unit of time
B. Anabolism is defined as the formation of substances, which can store the energy
C. Basal metabolic rate (BMR) is defined as the metabolic rate determined at rest in a room at 12–14 hours after the last meal
D. The BMR of a man is about 500 kcal per day
E. The metabolic rate is increased after consumption of a meal that is rich in protein

12. A   13. B   14. D   15. A   16. D   17. D   18. C   19. B   20. B   21. C   22. B   23. E   24. D

Q 25. Which of the following is true regarding metabolism?
  A. Oxidation is the combination of a substance with oxygen, or loss of hydrogen or an electron
  B. Co-enzyme A is a high-energy compound which is formed from adenine, ribose, pantothenic acid and thioethanolamine
  C. A calorie is defined as the amount of heat energy needed to raise the temperature of 1 g of water by 1°, from 15°C to 16°C.
  D. All the above
  E. None of the above

Q 26. Which of the following is true regarding the ribo-nucleic acid?
  A. Contains deoxyribose
  B. It is made by RNA polymerases
  C. Uracil pairs with thymine
  D. It is always single-stranded
  E. Adenine pairs with guanine

Q 27. Which of the following statement regarding vitamin B is not correct?
  A. Vitamin B1 (thiamine) deficiency leads to impaired collagen formation
  B. Vitamin B1 (thiamine) stores in the body are adequate for up to 9 months
  C. Vitamin B2 (riboflavin) concentration is higher in the foetus than in the mother
  D. Vitamin B6 (pyridoxine) requirement in pregnancy is 2.5 mg/day
  E. Niacin is synthesised in the body from tryptophan

Q 28. Which of the following statement regarding vitamin E is correct?
  A. Is present in animal foodstuffs only
  B. Its deficiency may cause intrauterine foetal death
  C. It does not potentiate the action of coumarin anticoagulants
  D. Is used in the treatment of infertility
  E. Its dietary requirement is 10 mg per day

Q 29. Which of the following organelles have their own self-replicating DNA?
  A. Golgi body
  B. Lysosomes
  C. Mitochondria
  D. Nucleolus
  E. Rough endoplasmic reticulum

Q 30. Which of the following is not true regarding cyclic AMP?
  A. Activates protein kinase C
  B. Activates STAT 3
  C. Degraded by phosphodiesterase
  D. Produced from ATP
  E. Produced in response to glucagon

Q 31. The cyclic AMP mechanism has been shown to be an intracellular hormonal mediator for which of the following hormones?
  A. Glucagon
  B. Parathyroid hormone
  C. Secretin
  D. Vasopressin
  E. All the above

Q 32. Which of the following is not true concerning nitric oxide (NO)?
  A. Effects of nitric oxide are mediated through cGMP as the second messenger
  B. It has a long half-life
  C. Is synthesised in the endothelium
  D. It produces relaxation of the smooth muscles
  E. Its production is increased in normal pregnancy

Q 33. A 40-year-old man presented with a painful swelling in the big toe and is suspected to be suffering from gout. Which blood parameter must be measured to support its diagnosis?
  A. Calcium levels
  B. Serum creatinine
  C. Serum uric acid
  D. Blood urea
  E. Blood xanthine

Q 34. Which of the following is true regarding G proteins?
  A. They are involved in initiating hormone action
  B. G proteins are cytoplasmic proteins
  C. They are part of cell surface receptors
  D. All the above
  E. None of the above

Q 35. Which of the following is not an adhesion molecule?
  A. Cadherin
  B. Fibronectin
  C. Laminin
  D. Secretin
  E. Integrin

Q 36. Which of the following is not true regarding the basal metabolic rate?
  A. Basal metabolic rate falls with increasing age
  B. Increases with increasing percent of lean body mass
  C. Is greater in males than females
  D. Is related to serum leptin levels
  E. It is the single largest component of energy expenditure

Q 37. A 48-year-old woman has poorly controlled type 2 diabetes. What single test would help in determining long-term glycaemic control?
  A. Fructosamine
  B. Glucose
  C. Glycated haemoglobin
  D. Glycated albumin
  E. Two hours-oral glucose tolerance test

25. D   26. B   27. B   28. E   29. C   30. B   31. E   32. B   33. C   34. D   35. D   36. D   37. C

Q 38. Which of the following are tumour suppressors?

- A. *p53*
- B. *pRb*
- C. *BRCA1*
- D. All the above
- E. None of the above

Q 39. Which of the following is not true regarding phenyl-ketonuria?

- A. It is an autosomal recessive disorder
- B. It is associated with defect in tyrosine metabolism
- C. Screening for phenylketonuria is done using Guthrie's test in newborns
- D. Screening for phenylketonuria is done using Kleihauer-Betke test in newborns
- E. Treatment strategy involves life-long intake of diet low in phenylalanine

Q 40. Which of the following is true regarding folic acid?

- A. Bioavailability is impaired by cooking
- B. Blood level is reduced in stagnant loop syndrome
- C. Body stores are adequate for 3 years
- D. Is absorbed predominantly in the ileum
- E. It is effective treatment for alcohol-induced macro-cytosis

Q 41. Which of the following is not true regarding steroid-ogenesis?

- A. Cholesterol is the precursor of all the steroids
- B. Corticosterone is converted into aldosterone
- C. Progesterone is the precursor of pregnenolone
- D. Pregnenolone is formed in the cellular mitochondria
- E. Testosterone may be converted to either dihydrotes-tosterone or oestradiol

Q 42. Which of the following is not true regarding prosta-glandins?

- A. They are synthesised from arachidonic acid
- B. Are antagonised by nonsteroidal anti-inflammatory drugs
- C. All prostaglandins consist of 20 carbon atoms, arranged in form of 5 rings
- D. Prostaglandins are hydrophilic in nature
- E. They bind to G-protein coupled receptors

Q 43. An iron overload is not seen in which of the following conditions?

- A. Thalassaemia major
- B. Polycythaemia rubra vera
- C. Myelodysplasia
- D. Haemochromatosis
- E. None of the above

Q 44. Which of the following are true regarding protein metabolism?

- A. Proteins contain about 16% nitrogen
- B. Chains containing greater than 100 amino acid residues are called proteins
- C. Proteins yield 4 kilocalories per gram absorbed
- D. All the above
- E. None of the above

Q 45. Which of the following is a non-essential amino acid?

- A. Arginine
- B. Leucine
- C. Methionine
- D. Tryptophan
- E. Tyrosine

Q 46. Raised serum iron level seen in which of the following?

- A. Thalassaemia major
- B. Myelodysplasia
- C. Haemochromatosis
- D. All the above
- E. None of the above

Q 47. What is not correct regarding enzymes?

- A. Are proteins
- B. Heating usually results in a complete loss of enzyme activity
- C. A change in pH has no effect on the activity of an enzyme
- D. Are present in all cell organelles
- E. Organic solvents will usually destroy an enzyme's activity

Q 48. Which of the following is not true regarding methio-nine?

- A. Cannot be converted to cystine by the foetal liver
- B. Can cross the placenta
- C. Is a sulphur-containing amino acid
- D. Is an essential amino acid
- E. Is reabsorbed in the proximal convoluted tubule in the kidneys

Q 49. Which of the following is true concerning $1,25\text{-(OH)}_2\text{D}_3$ (vitamin D)?

- A. Facilitates calcium and phosphate reabsorption from bone
- B. Stimulates the absorption of calcium and phosphate from the gut
- C. Stimulates the excretion of calcium and phosphate into renal tubules
- D. Levels are low during lactation
- E. Is less active than $24,25\text{-(OH)}_2$ vitamin D

Q 50. Which of the following is true regarding tetrahydro-folic acid?

- A. Catalyses the conversion of glucose to glucose-6-phosphate
- B. Activity is not inhibited by methotrexate
- C. Is a coenzyme in amino acid synthesis
- D. Is a precursor of folic acid
- E. Is involved in purine synthesis

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38. D   39. D   40. A   41. C   42. D   43. B   44. D   45. E   46. D   47. C   48. A   49. B   50. E

Q 51. **Which of the following statement about vitamins is correct?**
  A. Vitamin K is water-soluble
  B. Vitamin D is normally absorbed in cases of obstructive jaundice
  C. Vitamin A is a water-soluble vitamin
  D. Vitamins supply the body with energy
  E. Vitamin D is bound to a transport protein in the circulation

Q 52. **Which of the following nutrients and the deficiency disorder related to it is correctly matched?**
  A. Ascorbic acid—night blindness
  B. Cyanocobalamin—microcytic anaemia
  C. Folates—sprue
  D. Niacin—beriberi
  E. Thiamine—pellagra

Q 53. **Which of the following is not true regarding 1,25(OH)$_2$ vitamin D?**
  A. 25-hydroxylation occurs in the kidney
  B. Release is stimulated by hypophosphataemia
  C. Promotes phosphate and calcium absorption from the gut

  D. Attaches to cytosolic receptors
  E. Concentrations are decreased in chronic renal failure

Q 54. **Which of the following is true concerning 1,25-(OH)$_2$D$_3$ (vitamin D)?**
  A. Facilitates calcium and phosphate reabsorption from bone
  B. Is more active than 24,25-(OH)$_2$ vitamin D
  C. Levels are low during lactation
  D. Reduces the absorption of calcium and phosphate from the gut
  E. Stimulates the excretion of calcium and phosphate into renal tubules

Q 55. **Which of the following is correct regarding vitamin B$_{12}$?**
  A. Is a fat-soluble vitamin
  B. Absorption takes place throughout the small intestine
  C. Is essential for the metabolism of folic acid in the human
  D. Deficiency leads to microcytic anaemia
  E. Deficiency is common in non-vegetarians

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51. E   52. C   53. A   54. B   55. C

CHAPTER 5

Pathology
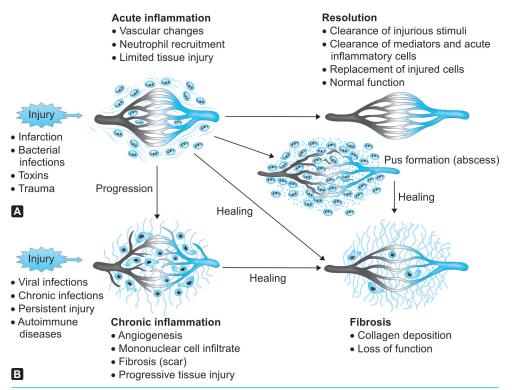
Inflammation

Inflammation is the local response of living mammalian tissues to injury from any agent, which could be microbial, immunological, chemical and physical agents such as trauma, radiation, heat and cold, etc. **(Figs 5.1A and B)**. Cardinal signs of inflammation are: redness, swelling, heat, pain and loss of function. Chemical mediators involved in production of an inflammatory response include globulin permeability factor, bradykinin, 5-hydroxytryptamine, plasma kinins, histamine, etc. Inflammation is of two types: acute and chronic. Inflammation is known as acute when it

occurs due to an early response by the body and is of short duration. Acute inflammatory response is characterised by two major events:
1. Vascular events and formation of an exudate
2. Cellular events and the release of mediators of acute inflammation.

Both these events have been discussed in details next in the text.

The inflammatory response is termed as chronic when it lasts for a longer duration of time and occurs after delay. Response by chronic inflammatory cells is the characteristic feature of chronic inflammation.

Acute inflammation
- Vascular changes
- Neutrophil recruitment
- Limited tissue injury

Resolution
- Clearance of injurious stimuli
- Clearance of mediators and acute inflammatory cells
- Replacement of injured cells
- Normal function

Injury
- Infarction
- Bacterial infections
- Toxins
- Trauma

Progression

A

Pus formation (abscess)

Healing

Healing

Injury
- Viral infections
- Chronic infections
- Persistent injury
- Autoimmune diseases

Healing

Chronic inflammation
- Angiogenesis
- Mononuclear cell infiltrate
- Fibrosis (scar)
- Progressive tissue injury

Fibrosis
- Collagen deposition
- Loss of function

B

FIGS 5.1A AND B: Inflammation in response to injury: (A) Acute inflammation; (B) Chronic inflammation

Acute Inflammation

Vascular Events

Alteration in the microvasculature (arterioles, capillaries and venules) is the earliest response to tissue injury. These alterations include: haemodynamic changes and changes in vascular permeability. The earliest vascular response is characterised by constriction of small vessels, rapidly followed by dilatation and a brief increase in the velocity of blood flow.

Progressive vasodilatation causes raised local hydrostatic pressure and transudation into the extracellular space. The crucial factor in formation of an inflammatory exudate is increased permeability of the vessel wall to plasma proteins. As a result, the exudate has the same composition as plasma. The exudate of acute inflammation contains more than 25 g/L protein. A transudate, on the other hand, has a low protein concentration (less than 25 g/L) and can result from a low serum protein concentration, high central venous pressure or portal hypertension. Formation of exudate, on the other hand, occurs in conditions such as tuberculous peritonitis, peritoneal malignancy, Budd-Chiari syndrome, pancreatic ascites, chylous ascites, Meigs' syndrome, etc.

The change in permeability of the vessel wall could be related to the widening of gaps between the endothelial cells. The exudate contains immunoglobulins and other antibacterial factors as well as fibrinogen, which may form the fibrin clot, thereby causing the exudate to clot upon standing. In acute inflammation, fibrin forms a union between severed tissues. It also forms a barrier against bacterial invasion, and aids phagocytosis.

Vasodilatation results in increased blood volume in microvascular bed of the inflamed area, which is responsible for redness and warmth at the site of acute inflammation. Increased transudation of fluid into the extracellular space is responsible for swelling at the local site of acute inflammation. Slowing or stasis of microcirculation follows which causes increased concentration of red cells, and thus, raised blood viscosity. Red cells also aggregate to form rouleaux. All these events result in the slowing of blood flow or sludging.

Stasis or slowing of blood flow is followed by leucocytic margination or peripheral orientation of neutrophils along the vascular endothelium. The leucocytes briefly adhere to the vascular endothelium, moving and migrating through the gaps between the endothelial cells into the extravascular space. This process is known as emigration.

Cellular Events

Cellular phase of inflammation consists of exudation of leucocytes through the endothelial gaps **(Fig. 5.2)**. Leucocyte exudation begins from change of normal axial blood flow to slowing and stasis. This is followed by margination, pavementing, rolling, adhesion, and finally
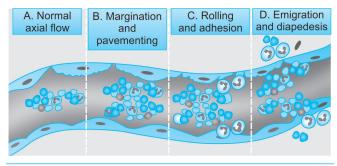
FIG. 5.2: Sequence of changes showing exudation of leucocytes in cases of acute inflammation

emigration of leucocytes into the extravascular space. The cells involved first are the neutrophil polymorphs. Later the blood monocytes also become phagocytic. In case of cutaneous wound healing, macrophages replace neutrophils within 48 to 96 hours.

This is followed by phagocytosis, which can be described as cellular eating. The process of engulfment of foreign particulate material, whether by polymorphs or the monocytes involves its initial recognition and opsonisation. While the mechanisms for phagocytosis are largely intra-cellular (oxidative and non-oxidative bactericidal), a few extracellular mechanisms can also occur. The energy requirements for phagocytosis are derived from aerobic glycolysis in the cells. Also, there occurs a huge increase in the hexose monophosphate shunt and NADPH in the neutrophil polymorphs.

Another early feature of acute inflammation is the release of histamine and heparin due to the degranulation of mast cells in the adjacent tissues. Additionally, many chemical substances may be involved in the process of acute inflammation. These are described in **Table 5.1** and

TABLE 5.1 Chemical mediators of inflammation
Cell-derived mediators
• Vasoactive amines:[1] Histamine, 5-hydroxytryptamine and neuropeptides
• Arachidonic acid metabolites (eicosanoids)[2]
– Metabolites via cyclooxygenase pathway: Prostaglandins, thromboxane A_2, prostacyclin and resolvins
– Metabolites via lipooxygenase pathway: 5-HETE acid, leukotrienes and lipoxins
• Lysosomal components (from PMNs, macrophages)
• Platelet activating factor
• Cytokines (IL-1, IL-6, IL-8, IL-12, IL-17, TNF-α, TNF-β, IFN-γ, chemokines)
• Free radicals (oxygen metabolites, nitric oxide)
Plasma protein-derived mediators (plasma proteases) products of:
• The kinin system[3]
• The clotting system
• The fibrinolytic system
• The complement system[4]

1. Vasoactive amines are responsible for producing early vascular response.
2. Prostaglandins participate in the later phases of inflammatory reaction; prostacyclin cause vasodilatation and increased capillary permeability; whereas leukotrienes contribute to smooth muscle contraction and chemotaxis.
3. Kinins (e.g. bradykinin, kallidin, etc.) cause smooth muscle contraction. Kinins such as tachykinins, substance P, etc. may be involved in local and general pain mechanisms.
4. Components of the complement system may act as chemotactic agents, attracting white blood cells to bacteria.
Abbreviations: HETE, hydroxyicosatetraenoic; PMN, polymorphonuclear neutrophil; IL, interleukin; TNF, tumour necrosis factor; IFN, interferon.
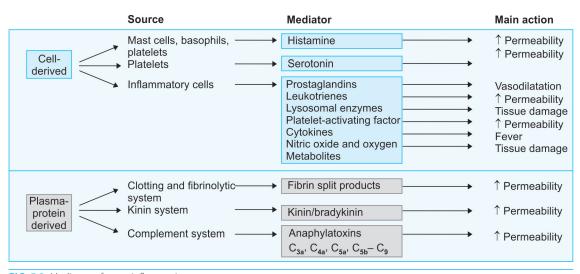
FIG. 5.3: Mediators of acute inflammation

Figure 5.3. Besides, liver also produces an increased amount of C-reactive protein (CRP) in response to the inflammation elsewhere in the body. The cells participating in the acute inflammatory process include circulating leucocytes, plasma cells and tissue macrophages.

The next series of events depend upon the cause and extent of inflammation (Fig. 5.4). A phase of demolition may occur which is characterised by the engulfment of fibrin, red cells, pus cells, bacteria, etc. by the macrophages. In case of minimal tissue destruction, complete resolution may occur. Resolution means the complete return to normal following acute inflammation. However in case of substantial tissue necrosis, the process of autolysis may result in pus (dead cells, debris, etc.) formation, which may be contained in a cavity to form an abscess. One of the essential features of an abscess is a lining pyogenic membrane consisting of necrotic tissue. Pyaemia is not an essential feature of abscess formation.

In cases of acute inflammation where the destruction is extensive or the bacteria persist in small numbers at the site of inflammation, the acute inflammatory process may develop into a chronic inflammatory process, e.g. osteomyelitis and pneumonia terminating into a lung abscess.

Chronic Inflammation

Chronic inflammation may result either following acute inflammation, may start anew after recurrent attacks of acute inflammation or may start de novo (infection with organisms of low pathogenicity, e.g. infection with *Mycobacterium tuberculosis*). A few general features of chronic inflammation are infiltration by mononuclear cells, tissue destruction and proliferation of blood vessels and fibroblasts. Other features of chronic infection may include evidence of the repair process, characterised by migration of capillaries and fibroblasts and formation of collagen. There may be endoarteritis obliterans, a condition
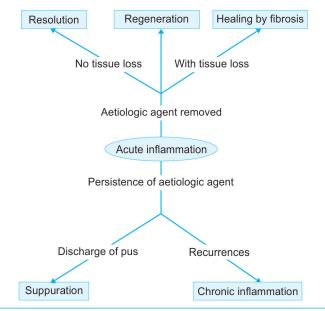
FIG. 5.4: Fate of acute inflammation

in which there is occlusion of small-sized arteries by intimal proliferation; accumulation of lymphocytes and plasma cells in the perivascular space; and excessive growth of the regenerating normal tissues.

A chronic inflammatory process may produce systemic features such as fever, anaemia, leucocytosis, raised erythrocyte sedimentation rate and development of secondary amyloidosis in the long-standing cases.

Amyloidosis is characterised by deposition of "amyloid material" (complex mucopolysaccharide containing globulins) in the connective tissue stroma and the walls of blood vessels of certain tissues and organs. The amyloid deposits lie around blood vessels. The liver is commonly involved and is enlarged, heavy, pale and firm. Renal failure may be the terminal manifestation of amyloidosis.

Pyrexial response is stimulated by cytokines such as tumour necrosis factor (TNF) and interleukins along with

prostaglandins. CRP is increased in febrile response but does not elicit response. Chronic inflammation is associated with increased levels of immunoglobulin G in the blood. In chronic inflammation the inflammatory and healing processes proceed side by side. Chronic inflammation is mainly of two types: non-specific and granulomatous type. In cases of non-specific chronic suppurative inflammation, abscesses may be formed. Example, in cases of chronic pyelonephritis, there is formation of multiple renal abscesses.

Granulomatous Type Chronic Inflammation

Formation of granuloma is a type IV granulomatous hypersensitivity reaction. It is a protective defense reaction by the host but eventually causes tissue destruction because of persistence of the poorly digestible antigen. Chronic inflammatory process in these cases may result in the formation of a solid tumour like mass or granulomas showing predominance of macrophages. A granuloma is composed of modified macrophages known as epithelioid cells in the centre, with some interspersed multinucleate giant cells, surrounded peripherally by lymphocytes (mainly T cells), and fibroblasts or collagen depending upon the age of granuloma. Examples include tuberculosis, syphilis, (the gumma), yaws, leprosy, actinomycosis, reaction to foreign bodies (talc granulomas), certain collagen diseases (e.g. Wegener's granulomatosis), etc. These granulomas may be sometimes associated with a central caseation, i.e. a solid central mass of necrotic debris. When macrophages encounter insoluble material, they may coalesce to form giant cells (e.g. Langhan's giant cells in cases of tuberculosis). Mechanism of granuloma formation is illustrated in the **Figure 5.5**. Differences between acute and chronic inflammation are summarised in **Table 5.2**.

Wound Healing

Healing is the body's response to injury in an attempt to restore normal structure and function. It comprises of two processes: regeneration and repair. Regeneration can be described as restoration to original tissue by proliferation of parenchymal cells. On the other hand, repair is healing which occurs due to the proliferation of connective tissue resulting in fibrosis and scarring. The process of repair involves initial inflammatory reaction by the body, clearance by proteolytic enzymes, followed by contraction, fibroplasia, angiogenesis and epidermal ingrowth.

Contraction occurs due to the shortening of the newly formed collagen fibres and due to the contraction of myofibroblasts. Fibroplasia is associated with budding of the adjacent capillaries, involving the canalisation of solid endothelial buds. This is followed by the migration of fibroblasts and macrophages into the wound cavity resulting in the formation of a vascular granulation tissue. There is synthesis of collagen and mucopolysaccharides
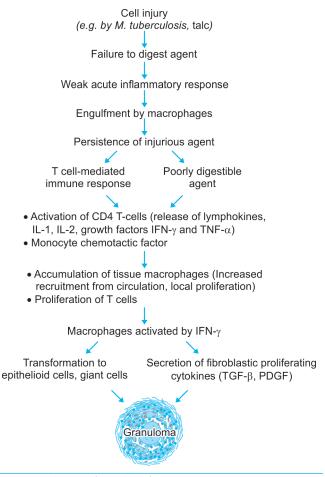
FIG. 5.5: Mechanism of granuloma formation
Abbreviations: IL-1, interleukin-1; IL-2, interleukin-2; INF-γ, interferon-gamma; TNF-α, tumor necrosis factor-alpha; TGF-β, transformation growth factor-beta; PDGF, platelet-derived growth factor

by the fibroblasts. This increasingly strengthens the scar tissues causing the scar to become relatively acellular and avascular. This is followed by the migration of healthy epidermal cells down the sides of the wound and across the granulation and fibrous tissues. New cells originate in the basal layer of epidermis near the wound margin. The epithelial ingrowth is limited to 1 cm from the wound margin. Wound healing can be accomplished in one of the following two ways **(Figs 5.6A and B)**:
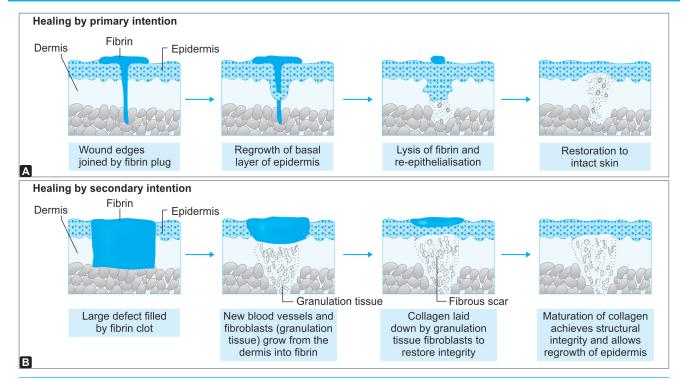1. Healing by first intention (primary union)
2. Healing by second intention (secondary union).

Difference between the two processes is highlighted in **Table 5.3**.

Healing by First Intention (Primary Union)

Healing by first intention can occur in a wound, which is clean and uninfected; created after surgical incision; and/or is associated with minimal loss of cells and tissue. It includes the cases where the edges of wound are approximated by surgical sutures. In cases where the wound edges are opposed, healing proceeds rapidly to closure and this is known as primary healing.

TABLE 5.2	Differences between acute and chronic inflammation	
Feature	**Acute inflammation**	**Chronic inflammation**
Definition	• Within short time • Lasts for short duration	• After delay • Lasts longer
Cardinal signs	**Invariably present**	**Generally indiscernible**
Pathogenesis	Acute inflammatory process is initiated as a result of vascular and cellular events: • *Vascular events*: Haemodynamic changes, increased vascular permeability • *Cellular events*: Exudation of leucocytes, phagocytosis (mediated via chemical mediators and regulators)	Chronic inflammatory process can occur as a result of the following: • Following acute inflammation • Recurrent attacks of acute inflammation • Chronic inflammation from beginning
Main inflammatory cells involved	• Neutrophils • Eosinophils • Lymphomononuclear cells (late) • Pus cells	• Lymphocytes • Plasma cells • Monocytes/macrophages (epithelioid cells in granulomas) • Giant cells (foreign body, Langhans')
Plasma exudation	**Present**	**May or may not be present**
Systemic effects	• Fever: High grade • Leucocytosis (neutrophilic, eosinophilic) • Lymphadenitis-lymphangitis • Septic shock (in severe acute infection)	• Fever: Mild • Leucocytosis (lymphocytic, monocytic) • Lymphadenitis-lymphangitis • Raised erythrocyte sedimentation rate • Anaemia • Amyloidosis (in long-term cases)
Main morphology	• Abscesses (suppuration) • Ulcers • Through blood (bacteraemia, septicaemia and pyaemia)	• Chronic nonspecific inflammation (infectious, others) • Granulomatous inflammation (tuberculosis, leprosy, sarcoidosis, syphilis, actinomycosis, Crohn's disease, etc.)
Fate	• Resolution • Healing (regeneration, fibrosis) • Chronicity	• Resolution • Healing (regeneration, fibrosis) • Dystrophic calcification
Common examples	Pyogenic abscess, cellulitis, bacterial pneumonia and pyaemia	Granulation tissue, granulomatous inflammation (tuberculosis, leprosy, etc.) and chronic osteomyelitis

FIGS 5.6A AND B: Mechanism of healing: (A) Healing by first intention; (B) Healing by second intention

TABLE 5.3	Comparison between the wound healing by primary union and the secondary union	
Characteristic	**Primary union**	**Secondary union**
Type of wound	Clean	Unclean
Presence of infection	Generally uninfected	May be infected
Wound margins	Surgical/clean	Irregular
Use of sutures for apposition of wound margins	Used	Not used
Healing	Scanty granulation tissue at the incised gap and along suture tracks	Exuberant granulation tissue to fill the gap
Outcome	Neat linear scar	Contracted irregular wound
Complications	Infrequent, epidermal inclusion cyst formation	Suppuration, may require debridement

Healing by Second Intention (Secondary Union)

Healing by second intention occurs in an open wound, having a large tissue defect, which at times may even be infected. These wounds may be associated with an extensive loss of cells and tissues. Such wounds are not approximated by surgical sutures but are left open.

The basic events occurring in healing by secondary union are similar to primary union but differ in the extent of tissue defect which needs to be bridged. Such wounds are usually associated with large tissue defects. Therefore, healing by secondary intention is a slower process due to the formation and contraction of granulation tissue resulting in a slow apposition of the opposing skin appendages. Healing not only takes place from margins inwards but also the base upwards. Healing by second intention often results in the formation of a large, at times, ugly scar. This is in contrast to the neat and clean scar formed due to rapid healing as a result of primary union. When the wound is infected it should heal by secondary intention. Attempting to heal the wound by primary measures would leave an underlying infection that would eventually lead to wound breakdown.

Complications of wound healing are infection, inclusion cyst formation, pigmentation, incisional hernia, hypertrophied scar and contracture.

The wound is strengthened by proliferation of fibroblasts and myofibroblasts, which get structural support from the extracellular matrix (ECM). ECM is comprised by collagen, adhesive glycoproteins, basement membrane, elastic tissue and proteoglycans. Tensile strength of the healing wound depends on the amount and arrangement of the collagen fibres. Content of collagen usually reaches its maximum on the day 80. However, the strength may not reach its maximum for many months. Elasticity of the skin depends upon the presence of elastin fibres in the dermis, which are arranged in parallel with the skin creases. Therefore, incisions perpendicular to these creases tend to gape and heal less well in comparison to the incisions, which are parallel to the creases.

Factors Influencing Wound Healing

Various local and systemic factors may influence wound healing. These have been described below in details.

Local Factors

❑ *Infection*: Infection is the most important factor acting locally, which delays the process of healing.
❑ *Vascularity*: Poor blood supply to wound is another important factor for delaying the healing process, e.g. injuries to the face and scalp heal quickly due to presence of rich blood supply. On the other hand, injury to leg with varicose ulcers having poor blood supply heals slowly.
❑ *Foreign bodies*: Presence of foreign bodies including sutures (especially catgut sutures) interferes with healing and causes intense inflammatory reaction and infection.
❑ *Movement*: Movement delays the process of wound healing and predisposes to stretching and keloid formation.
❑ *Exposure to ionising radiation*: This delays the formation of granulation tissue.
❑ *Exposure to ultraviolet light*: This usually facilitates the healing process.
❑ *Type, size and location of injury*: This helps in determining whether healing takes place by resolution or organisation.
❑ *Necrosis*: Presence of necrosis at the wound margins delays the repair process.
❑ *Lymph drainage*: Impairment of the lymph drainage causes oedema and delay in the repair process.
❑ *Amount of tissue separation in the wound*: A clean wound with neatly apposed edges would heal rapidly with the formation of minimal granulation tissue. On the other hand, a wound with separated edges would take longer because in these cases, healing process would occur via second intention.

Systemic Factors

❏ *Age*: Wound healing occurs rapidly in young people, whereas it occurs somewhat at a slow pace in aged and debilitated people due to poor blood supply to the injured area in the latter.

❏ *Nutrition*: Deficiency of constituents like proteins (especially due to the deficiency of sulphur containing amino acids such as methionine), vitamin C (scurvy or malabsorption), vitamin A and zinc delays the wound healing. Deficiency of vitamin C leads to failure of collagen formation by preventing the conversion of proline to hydroxyproline.

❏ *Systemic infection*: Presence of systemic infection usually delays wound healing.

❏ *Administration of glucocorticoids*: Corticosteroids have an anti-inflammatory effect. They can delay healing, especially if high doses are administered for prolonged periods.

❏ *Diabetic patients*: Uncontrolled diabetics are more prone to develop infections and hence delay in healing.

❏ *Haematologic abnormalities*: Presence of haematological abnormalities, such as defect in neutrophil functions (chemotaxis and phagocytosis), neutropaenia and bleeding disorders, slow the process of wound healing.

❏ *Temperature*: Wound healing usually occurs slowly at low temperatures.

Adaptive Disorders

Adaptive disorders are the adjustments which the cells make in response to stresses which may be for physiological requirements or pathological (in response to non-lethal pathologic injury). These could include the following as described next.

Atrophy

This can be described as a reduction of the number and size of parenchymal cells of an organ, which was once normal.

Hyperplasia

Hyperplasia is an increase in the number of parenchymal cells resulting in enlargement of the organ or tissue. Hyperplasia occurs due to the presence of a stimulating factor which causes increased recruitment of cells from G0 (resting) phase of the cell cycle to undergo mitosis. All body cells do not possess hyperplastic growth potential. Labile cells (e.g. epithelial cells of the skin and mucous membranes, cells of the bone marrow and lymph nodes) and stable cells (e.g. parenchymal cells of the liver, pancreas, kidney, adrenal and thyroid gland) can undergo hyperplasia. On the other hand, permanent cells (e.g. neurons, cardiac and skeletal muscle) have little or no capacity for regenerative hyperplastic growth.

Hyperplasia can occur due to physiological causes, e.g. hyperplasia of uterine smooth muscles during pregnancy and hyperplasia of breast tissue during puberty. In some cases, hyperplasia can occur due to pathological causes, e.g. hyperplasia of the adrenal cortex in cases of Cushing's syndrome; benign prostatic hypertrophy, and endometrial hyperplasia in response to unopposed oestrogens.

Hypertrophy

Hypertrophy can be defined as an increase in the size of parenchymal cells resulting in enlargement of the organ or tissue, without causing any change in the number of cells.

Metaplasia

Metaplasia is a pathological change, which refers to the reversible replacement of one type of differentiated cells with another type of mature differentiated cells. Metaplastic cells are normal cells without nuclear atypia and therefore do not act as precursors of malignancy. Metaplasia can be broadly of two types: epithelial (squamous and columnar) and mesenchymal (osseous and cartilaginous) **(Table 5.4)**. In squamous metaplasia, the normal tissue epithelium is converted into squamous epithelium, whereas in columnar metaplasia, the normal tissue epithelium is converted into columnar epithelium.

TABLE 5.4   Examples of epithelial metaplasia

Squamous metaplasia		
Tissue involved	**Normal epithelium type**	**Pathology**
Bronchus	Pseudostratified columnar ciliated epithelium	Chronic smokers
Uterine endocervix	Simple columnar epithelium	Prolapse of the uterus and in old age
Gallbladder	Ducts normally lined by simple columnar epithelium	Chronic cholecystitis with cholelithiasis
Prostate	Ducts normally lined by simple columnar epithelium	Chronic prostatitis and oestrogen therapy
Renal pelvis and urinary bladder	Lined by transitional epithelium	Chronic infection and stones
Columnar metaplasia		
Oesophagus	Normal squamous epithelium	Barrett's oesophagus
Bronchus	Pseudostratified ciliated columnar epithelium	Chronic bronchitis and bronchiectasis

Dysplasia, on the other hand, refers to the abnormal maturity of the epithelium. Atypical metaplasia with abnormal nuclear changes acts as a precursor of dysplasia and malignancy. Therefore, proliferative metaplasia without atypical mitotic activity should not be termed as dysplasia. Differences between metaplasia and dysplasia are tabulated in **Table 5.5**.

Dysplasia

Dysplasia is the process, which refers to an abnormal maturation of cells within the tissue. This process differs from metaplasia in the sense that normal differentiated cells are replaced by abnormal undifferentiated cells, unlike metaplasia in which one type of differentiated epithelial cells are replaced by another type of normal differentiated epithelial cells. Dysplasia is often indicative of an early neoplastic process and is characterised by presence of disordered cellular development (e.g. disorderly arrangement, loss of polarity, pleomorphism, increased nucleus: cytoplasmic ratio, increased mitotic activity, etc.).

Heteroplasia

Heteroplasia refers to abnormal growth of cells in the wrong location in absence of any stimulus.

Cellular Response to Injury

Fate of cell in response to injury is described in **Figure 5.7**.

Tissue Necrosis

Necrosis refers to the irreversible death of cells due to injury. Necrosis is defined as a localised area of death of tissue followed later by degradation of tissue by hydrolytic enzymes liberated from dead cells. It is invariably accompanied by inflammatory reaction. However, it is not an inevitable event in the cell cycle. Necrosis can be caused by various agents such as hypoxia, chemical and physical agents, microbial agents, immunological injury, etc. Based on aetiology and morphologic appearance, there are five types of necrosis: coagulative, liquefaction (colliquative), caseous, fat and fibrinoid necrosis. Caseous necrosis is characterised by the presence of cheese-like matter at the site of necrosis following infection with *Mycobacterium tuberculosis*.

The series of changes which can occur in the necrotic cells include cytoplasmic eosinophilia, pyknosis (shrinkage of cells), karyorrhexis (breakdown of the nucleus) and karyolysis (complete dissolution of the nucleus).

Apoptosis

Apoptosis is a form of "coordinated and internally programmed cell death" having significance in a variety of physiologic and pathologic conditions, e.g. tissue differentiation in embryogenesis and sloughing off of the endometrium during menstruation. Unlike necrosis, apoptosis is not accompanied by any inflammation and collateral tissue damage. A series of sequential changes are likely to occur in the cell and its organelles as a result of

| TABLE 5.5	Differences between metaplasia and dysplasia	
Characteristics	**Metaplasia**	**Dysplasia**
Definition	Change of one type of differentiated epithelial or mesenchymal cells to another type of adult epithelial or mesenchymal cells. It is characterised by mature cellular development	It is characterised by disordered cellular development (pleomorphism, nuclear hyperchromasia, mitosis, loss of polarity, etc.). It may be accompanied with hyperplasia or metaplasia.
Types	Epithelial (squamous, columnar) and mesenchymal (osseous, cartilaginous)	Epithelial only
Tissues affected	Most commonly affects bronchial mucosa, uterine endocervix; other mesenchymal tissues (cartilage, arteries, etc.)	Uterine cervix, bronchial mucosa
Natural history	Reversible on withdrawal of stimulus	May regress on removal of inciting stimulus, or may progress to higher grades of dysplasia or carcinoma in situ
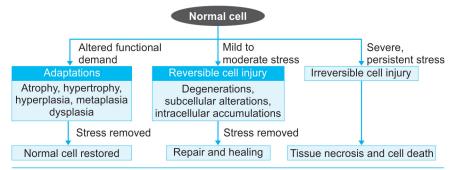
FIG. 5.7: Cellular responses to injury

apoptosis. These include cell shrinkage (pyknosis), nuclear shrinkage and condensation of chromatin, karyorrhexis, etc. Apoptosis may also be associated with the formation of cytoplasmic blebs called apoptotic bodies.

Tumourogenesis

Neoplasia

Neoplasia refers to abnormal proliferation of the cells. It is characterised by an abnormal, excessive, uncoordinated, autonomous and purposeless proliferation of cells even after cessation of stimulus for growth which caused it. Neoplastic changes can be either benign or malignant in nature. Neoplasia differs from hyperplasia in the sense that neoplasia is characterised by the loss of growth-regulatory mechanism due to change in genetic composition of the cell. On the other hand, hyperplasia persists so long as the stimulus is present.

Neoplastic changes are termed as "benign" when they are slow-growing and localised without causing much difficulty to the host. They are termed as "malignant" or "cancerous" when they proliferate rapidly, spread throughout the body, thereby eventually causing death of the host. Malignant tumours can be sarcomas or carcinomas. Malignant tumours of epithelial origin are called carcinomas, while malignant tumours of mesenchymal origin are termed as sarcomas. Besides these, some cancers are composed of highly undifferentiated cells and are referred to as undifferentiated malignant tumours. **Table 5.6** summarises the differences between benign and malignant tumours.

Epidemiology and Predisposing Factors for Cancer

The most common cancers encountered in the developed countries are lung, breast, prostate and colorectal cancers. On the other hand, the most common tumours in developing countries are liver, cervix, oral cavity and oesophagus. Several factors predispose to occurrence of cancers **(Table 5.7)**. These may include factors such as: familial and genetic factors, racial and geographic factors, environmental and cultural factors, and age and sex.

Symptoms

Malignant tumours are associated with more adverse ill-effects in comparison to the benign tumours and these may be local, or generalised and more widespread. Local effects of the tumour may be due to mechanical compression, obstruction, tissue destruction and infarction, ulceration and haemorrhage. Systemic effects, on the other hand, are in the form of cancer cachexia, fever, tumour lysis syndrome (a condition caused by extensive destruction of a large number of rapidly proliferating tumour cells), and paraneoplastic syndrome.

Paraneoplastic syndromes are a group of conditions developing in patients with advanced cancer. These are usually related with symptoms which can neither be explained by direct and distant spread of the tumour, nor by the usual hormone elaboration by the tissue of origin of the tumour. The various clinical syndromes included in this group are summarised in **Table 5.8**.

TABLE 5.6	Differences between benign and malignant tumours	
Characteristics	**Benign**	**Malignant**
Boundaries	Encapsulated or well-circumscribed	Poorly-circumscribed and irregular
Surrounding tissue	Often compressed	Usually invaded
Size	Usually small	Often larger
Secondary changes	Occur less commonly	Occur more commonly
Growth rate	Usually slow	Usually rapid
Local invasion	Often compresses the surrounding tissues without invading or infiltrating them	Usually infiltrates and invades the adjacent tissues
Metastasis	Absent	Frequently present
Prognosis	Good (there may be local complications)	Poor (death by local and metastatic complications)
Histopathological pattern	Usually resembles the tissue of origin closely	Often poor resemblance to tissue of origin
Features of cytological atypia (e.g. pleomorphism, anisonucleosis, hyperchromatism, chromosomal abnormalities, etc.)	Usually absent	Usually present
Nucleocytoplasmic ratio	Normal	Increased
Basal polarity	Retained	Often lost
Mitoses	May be present but are always typical mitoses	Mitotic figures increased and are generally atypical and abnormal
Function	Usually well maintained	May be retained, lost or becomes abnormal

TABLE 5.7   Likely predisposing factors for various cancers

Virus as the likely causative factor	
Types of cancer	**Causative virus**
Burkitt's lymphoma, Hodgkin's lymphoma, and nasopharyngeal carcinoma	Epstein-Barr virus (human herpes virus 4)
Cervical cancer	Human papillomavirus subtypes 16 and 18
Kaposi's carcinoma	Human herpes virus 8
Hepatocellular carcinoma	Hepatitis B and C; exposure to aflatoxin B1; alcoholism and haemochromatosis are other likely risk factors
Carcinogen as the likely causative factor	
Mesothelioma	Asbestos
Vaginal cell clear cancer	Exposure to diethylstilbestrol during foetal development
Leukaemia (acute myeloid leukaemia, chronic lymphocytic leukaemia)	Benzene
Bladder cancer	Aniline dye (also known as benzidine); β-naphthylamine (constituent of cigarette smoke)
Bowel cancer	Low fibre diet, high levels of processed meats, smoking, etc.
Underlying premalignant condition	
Oral cancer	Leucoplakia
Colorectal cancer	Ulcerative colitis
Squamous cell carcinoma	Actinic keratosis
Gastric adenocarcinoma	Atrophic gastritis
Cervical cancer	Cervical intraepithelial neoplasia
Oesophageal cancer	Barrett's oesophagus

TABLE 5.8   Paraneoplastic syndromes and their associated malignancies

	Paraneoplastic syndrome	**Associated malignancies**	**Mechanism**
Endocrine	Syndrome of inappropriate ADH secretion	Lung cancer and tumours of central nervous system	Inappropriate ADH production
	Cushing's syndrome	Small cell lung cancer, pancreas and neural tumours	Production of ACTH or ACTH-like substance
	Hypercalcaemia	Lung (squamous cell carcinoma), kidney, breast and adult T-cell leukaemia, lymphoma	Production of parathormone-like protein, vitamin D
	Hypoglycaemia	Pancreas (islet cell tumour), mesothelioma and fibrosarcoma	Insulin or insulin-like substance
	Carcinoid syndrome	Bronchial carcinoid tumour, carcinoma pancreas and stomach	Serotonin, bradykinin
	Polycythaemia	Kidney, liver and cerebellar haemangioma	Erythropoietin
Dermatological	Acanthosis nigricans	Uterine cancer	Immunologic
	Dermatomyositis	Breast cancer	Immunologic
	Seborrhoeic dermatitis	Bowel	Immunologic
	Exfoliative dermatitis	Lymphoma	Immunologic
Hematological	Polycythaemia	Renal cancer and hepatocellular cancer	Increased red cell production
	Lambert-Eaton myasthenic syndrome	Small cell lung cancer	Autoimmune disorder due to formation of auto-antibodies
	Thrombophlebitis (Trousseau's phenomenon)	Pancreas, lung and GIT	Hypercoagulability
	Nonbacterial thrombotic endocarditis	Advanced cancers	Hypercoagulability
	Disseminated intravascular coagulation (DIC)	AML and adenocarcinoma	Chronic thrombotic phenomena
	Anaemia	Thymoma	Unknown
Neuromuscular Syndromes	Myasthenia gravis	Thymoma	Immunologic
	Neuromuscular disorders	Lung (small cell carcinoma) and breast	Immunologic
Osseous, joint and soft tissues	Hypertrophic osteoarthropathy	Lung	Not known
	Clubbing of fingers	Lung	Not known
Gastrointestinal syndromes	Malabsorption	Lymphoma of small bowel	Hypoalbuminaemia
Renal syndromes	Nephrotic syndrome	Advanced cancers	Renal vein thrombosis, systemic amyloidosis
Amyloidosis	Primary	Multiple myeloma	Immunologic (AL protein)
	Secondary	Kidney, lymphoma and solid tumours	AA protein

Abbreviations: ADH, antidiuretic hormone; ACTH, adrenocorticotropic hormone; GIT, gastrointestinal tract; AML, acute myeloid leukaemia.

Tumour Markers

Tumour markers are biochemical assays of products elaborated by the tumour cells in blood or other body fluids. It is, therefore, pertinent to keep in mind that many of these products are produced by normal body cells too, and thus the biochemical estimation of the product in blood or other body fluids reflect the total substance and not that produced by the tumour cells alone. Specific markers for different types of cancers are summarised in **Table 5.9**.

Diseases of the Blood Vessels

Atherosclerosis

Atherosclerosis can be defined as the thickening and hardening of large and medium-sized muscular arteries, primarily due to involvement of tunica intima and is characterised by deposition of fibrofatty plaques or atheromas. It begins as patchy deposits of cholesterol and its esters in the intima at the orifices of the arterial branches **(Fig. 5.8)**. As a result, the adjacent arterial walls may become fibrosed, calcified and ulcerated. In large vessels, mechanical weakening may result in development of an aneurysm. Atherosclerosis is the most frequently occurring arterial disease and is the commonest cause for arterial thrombosis. Though any large and medium-sized artery may be involved in atherosclerosis, the most commonly affected are the aorta, the coronaries and the cerebral arterial systems. The major clinical syndromes resulting from ischaemia due to atherosclerosis are as follows:

❑ *Heart*: Angina and myocardial infarction or heart attacks
❑ *Brain*: Transient cerebral ischaemia and cerebral infarcts or strokes

❑ *Other sequelae*: These include abnormalities such as peripheral vascular disease, aneurysmal dilatation due to weakened arterial wall, chronic ischaemic heart disease, ischaemic encephalopathy and mesenteric arterial occlusion. Predisposing factor for atherosclerosis include the following:
- Hypertriglyceridaemia
- Hypercholesterolaemia
- Hyperlipidaemia (and high fat consumption)
- Hypertension
- Diabetes
- Smoking
- Obesity.

Besides atherosclerosis, obesity is also associated with a higher risk of diseases such as osteoarthritis, carcinoma of the breast, kidney and prostate, hypertension, diabetes
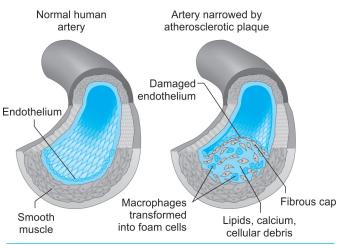
FIG. 5.8: Mechanism of atherosclerosis

TABLE 5.9	Specific markers for different types of cancers	
Class of marker	**Specific marker**	**Cancer**
Oncofoetal antigens	Alpha-foetoprotein (AFP)	Hepatocellular carcinoma and non-seminomatous germ cell tumours of testis
	Carcinoembryonic antigen (CEA)	Cancer of bowel, pancreas and breast
Enzymes	Prostate acid phosphatase (PAP)	Prostatic carcinoma
	Neuron-specific enolase (NSE)	Neuroblastoma and oat cell carcinoma lung
	Lactic dehydrogenase (LDH)	Lymphoma and Ewing's sarcoma
Hormones	Human chorionic gonadotropin (hCG)	Trophoblastic tumours and non-seminomatous germ cell tumours of testis
	Calcitonin	Medullary carcinoma thyroid
	Catecholamines and vanillylmandelic acid (VMA)	Neuroblastoma and pheochromocytoma
	Ectopic hormone production	Paraneoplastic syndromes
Cancer-associated proteins	CA-125	Ovarian cancer and primary peritoneal cancer
	CA 15-3	Breast cancer
	CA 19-9	Colon, pancreas and breast
	CD30	Hodgkin's disease and anaplastic large cell lymphoma
	CD25	Hairy cell leukaemia, adult T-cell leukaemia and lymphoma
	Monoclonal immunoglobulins	Multiple myeloma and other gammopathies
	Prostate specific antigen (PSA)	Prostate carcinoma

mellitus, and psychiatric conditions such as depression. It is relatively protective for osteoporosis.

The recent WHI (Woman's Health Initiative) study analysing the effect of postmenopausal hormonal therapy with cardiovascular risk revealed a small but significantly increased cardiovascular risk in postmenopausal women.

Berry Aneurysms

An aneurysm can be defined as a permanent abnormal dilatation of a blood vessel occurring due to congenital or acquired weakening or destruction of the vessel wall. Berry aneurysms result from abnormalities in the medial wall of the arteries. They are most often found in the circle of Willis. Berry aneurysms are not related to atheroma, and are dependent on genetic predisposition with physiological influences (for example, blood pressure). Berry aneurysms are strongly associated with polycystic renal disease. They however are not more commonly related to diabetes.

Thrombosis

Thrombosis is the process of formation of solid mass in circulation from the constituents of flowing blood; the mass itself is called a thrombus **(Fig. 5.9)**. Thrombogenesis involves interplay of the following events **(Figs 5.10A to C)**:

❑ Endothelial injury
❑ Platelets and their release reaction
❑ Activation of the coagulation system
❑ Alterations in the flow of blood
❑ Role of certain predisposing conditions and factors causing hypercoagulable states (or thrombophilia).

Endothelial injury exposes subendothelial matrix to circulating blood. This triggers the following steps involving platelet activation: adhesion, release and aggregation. Platelet release is associated with the release of granules (alpha granules and dense bodies). Concurrent activation of coagulation cascade generates fibrin strands and thrombin forming a tight meshwork called thrombus. For details related to coagulation cascade, kindly refer to Chapter 3.
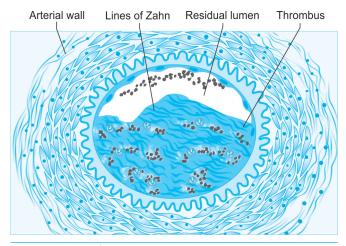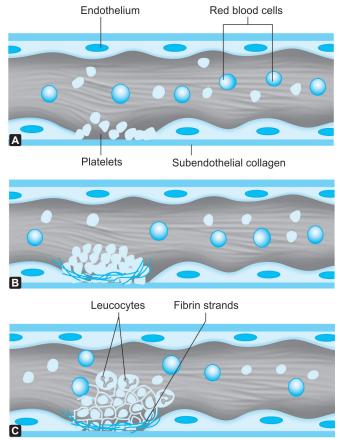
FIG. 5.9: Presence of thrombus in an artery

Thrombi may originate in the chambers of the heart, lumina of arteries, veins and microcirculation. The effects of thrombus depend upon its anatomic location of origin, speed of formation and nature of thrombi. Thrombi may assume importance by producing life-threatening harmful effects due to ischaemia (hypoxic injury due to blockage of blood flow) and by thromboembolism. Grossly, thrombi may be of various shapes, size, consistency and colour. Microscopically, all types of thrombi show lines of Zahn formed by alternate layers of light-staining aggregated platelets and dark-staining red cells. The possible fates of thrombi are as follows:

❑ *Resolution*: Dissolution of the thrombus due to activation of the fibrinolytic system. This may result in the complete restoration of blood flow in cases of small thrombi.
❑ *Organisation*: If the thrombus is not removed, it starts getting organised. The clot may become organised due to phagocytosis of fibrin and cell debris by phagocytic cells such as macrophages and neutrophils. There also occurs growth of fibroblasts and ingrowth of capillaries.
❑ *Propagation*: The thrombus may enlarge in size due to deposition of more and more constituents from the flowing blood. Eventually, the enlarged thrombus

FIGS 5.10A TO C: Process of thrombosis: (A) Endothelial injury; (B) Platelets aggregation and their release reaction; (C) Generation of fibrin strands and thrombus formation

may ultimately cause obstruction of some important vessel.

❏ *Deep vein thrombosis*: Thrombophlebitis and infection of a lodged thrombus in a deep vein of the leg can result in the development of deep vein thrombosis. This can occur as a result of extension of puerperal infection along the venous route and can be associated with pain swelling, tenderness and inflammation of the affected leg.

❏ *Thromboembolism*: The thrombus may sometimes get dislodged from the vessel wall. These may be released in part or completely in blood-stream in form of emboli and may produce adverse effects at the site they finally lodge into.

Tendency for increased thrombosis occurs in the following situations:

❏ Congestive cardiac failure
❏ Trauma
❏ Surgery
❏ Myeloproliferative disorders
❏ Oral contraceptives
❏ Other inherited causes such as:
  • Antithrombin III/protein C/protein S deficiency
  • Factor V Leiden thrombophilia
  • Dysplasminogenaemia
  • Dysfibrinogenaemia
  • Heparin cofactor II deficiency.

Embolus

A thrombus dislodged from its origin is known as an embolus. It may be carried via blood stream to some part of the circulatory system, where it may finally lodge to produce partial or complete obstruction, resulting in development of adverse effects due to hypoxia or anoxia.

Pulmonary Embolism

Pulmonary thromboembolism is common and fatal form of venous thromboembolism, most often originating from deep vein thrombosis of the lower legs. In these cases, the embolus originates from the deep veins in the legs (90% of the cases from iliofemoral veins) and lodges into the pulmonary veins.

A pulmonary embolus can present with symptoms such as dyspnoea, pleuritic chest pain (with an audible pleural rub), haemoptysis, cyanosis, pulmonary hypertension, right ventricular failure or cardiac arrest **(Fig. 5.11)**. They can sometimes also be asymptomatic. A pleural rub is not a diagnostic feature for confirming the diagnosis of pulmonary embolism because it may also be found on auscultation in cases of pneumonia.

Diagnosis

❏ *ECG*: The associated ECG changes in a case of pulmonary embolism are not diagnostic, but include signs of right
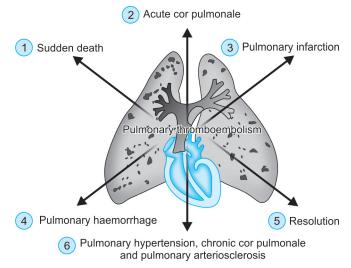
FIG. 5.11: Major consequences of pulmonary embolism

ventricular strain with right axis deviation, right bundle branch block and the S1, Q3, T3 pattern (S wave in lead I and Q wave and inverted T wave in lead III).

❏ *Arterial blood gas analysis*: The classical findings on arterial blood gas analysis show hypoxia, hypocarbia and increased alveolar-arterial oxygen gradient.

❏ *Ventilation-perfusion scan*: A positive ventilation-perfusion scan shows persistent ventilation in the affected region with absent perfusion. The presence of co-existing pulmonary pathology is likely to reduce the sensitivity of this investigation.

Treatment

Treatment of pulmonary embolism requires systemic anticoagulation (heparin followed by warfarin), and in selected cases only (central pulmonary embolism) surgical embolectomy may be performed.

Shock

Shock is a clinical syndrome of cardiovascular collapse and circulatory failure characterised by an acute reduction of effective circulating blood volume (hypotension) and an inadequate perfusion of cells and tissues (hypoperfusion). All forms of shock involve three main mechanisms **(Fig. 5.12)**.

1. Reduction in effective circulating blood volume
2. Impaired tissue oxygenation and
3. Release of proinflammatory mediators.

Shock can occur in three stages: initial reversible stage (compensated shock), progressive decompensated shock and finally the stage of irreversible decompensated shock. Hypoxic injury can occur in various body organs as a result of shock: brain (hypoxic encephalopathy), heart (haemorrhage and necrosis), lungs (acute respiratory distress syndrome), kidneys (tubular necrosis), adrenals (haemorrhage and necrosis), liver (focal necrosis), gut

Aetiology

Hypovolaemic shock
Haemorrhage Trauma Surgery Burns Dehydration

Septic (Toxaemic) shock
Gram-negative septicaemia Gram-positive septicaemia

Cardiogenic shock
Deficiency of emptying Deficiency of filling Outflow obstruction

Other types
Traumatic shock Neurogenic shock Hypoadrenal shock

Pathogenesis

↓ Effective circulating blood volume

↓ Venous return to heart

↓ Cardiac output

↓ Blood flow

↓ Supply of oxygen

Anoxia

Inflammatory mediators

Shock

FIG. 5.12: Aetiopathogenesis of circulatory shock

(haemorrhagic gastroenteropathy), etc. Clinically, shock is characterised by low blood pressure, low body temperature, feeble pulse, shallow respiration, pale face and cold clammy skin.

The mainstay of management for all forms of shock is the maintenance of optimal tissue perfusion and ventilation through the administration of oxygen and intravenous fluids. Further management usually is based on the alleviation of the likely underlying cause for shock, e.g. administration of intravenous antibiotics in cases of septic shock. There are three major forms of shock, namely hypovolaemic shock, cardiogenic shock and septic shock. These various types are described in details next in the text.

Hypovolaemic Shock

This form of shock results from inadequate circulatory blood volume due to various etiologic factors. There either may be loss of red cell mass and plasma due to haemorrhage, or loss of plasma volume alone, e.g. fluid loss due to burns, fluid loss due to gut infection such as cholera. The severity of clinical features related to hypovolaemic shock depend upon degree of blood volume lost. Depending on the amount of blood loss, haemorrhagic shock can be of the following types: compensated (≤1,000 mL); mild (1,000–1,500 mL); moderate (1,500–2,000 mL), and severe (>2,000 mL). The reduction in blood volume causes a fall in the central venous pressure, reduced venous inflow into the right atrium, a fall in arterial blood pressure and a generalised reduction in tissue perfusion. Tissue hypoxia may eventually result in the development of metabolic acidosis. Major clinical features in these cases are increased

heart rate (tachycardia), low blood pressure (hypotension), low urinary output (oliguria to anuria) and alteration in mental state, which can vary from agitated-to-confused-to-lethargic stage. The blood circulation to the brain, heart and adrenals is usually conserved unless the condition becomes very severe. The kidneys are also able to adapt to moderate decrease in blood volume due to reduced vascular resistance. A rise in platelets usually follows in an hour and there is a rise in white cells (usually the polymorphs) within 3–5 hours.

Cardiogenic Shock

In these cases, there is acute circulatory failure with sudden fall in cardiac output due to acute diseases of the heart. However, there is no actual reduction of blood volume, thereby resulting in normovolaemia in these cases.

Septic (Toxaemic) Shock

Septic shock is caused by **s**evere bacterial infections or septicaemia. It is commonly caused by the toxins of Gram-negative bacteria, e.g. *Escherichia coli, Aerobacter aerogenes, Bacillus proteus*, etc. The endotoxic shock results in these cases due to the release of lipopolysaccharide toxins from the cell wall. Septic shock may sometimes also result from Gram-positive bacteria (e.g. diphtheria and gas gangrene) resulting in exotoxic shock.
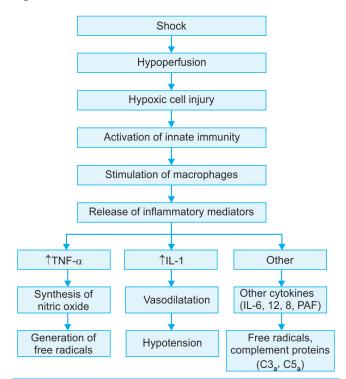
Binding of the endotoxin to the lipopolysaccharide binding protein causes stimulation of macrophages, causing the release of proinflammatory cytokines (especially TNF-α and interleukin-1). These cytokines lead to recruitment of neutrophils resulting in liberation of free radicals causing vascular injury **(Fig. 5.13)**. Moreover, there is activation of other inflammatory cascades which may have a profound effect in triggering septic shock. These cascades include complement pathway, mast cells, coagulation system, kinin system, etc. The net result of these mechanisms is vasodilatation and increased vascular permeability. Profound peripheral vasodilatation and pooling of blood causes hyperdynamic circulation in cases of septic shock. This is in contrast to the hypovolaemic circulation in cases of hypovolaemic shock. In advanced cases of septic shock, increased vascular permeability causes development of inflammatory oedema. Damage to the vessel wall may lead to the loss of circulating fluids and proteins into the tissues. The reduction in blood volume is associated with disturbances in the fluid and electrolyte and an increase in blood viscosity (sludging). All these events result in an impaired tissue perfusion and cell anoxia. Occurrence of disseminated intravascular coagulation (DIC) caused by the release of thrombogenic enzymes due to endothelial cell injury by toxins often aggravates the process. Reduced blood flow produces hypotension, inadequate perfusion of cells and tissues, finally leading to organ dysfunction.

Renal damage is likely to occur both due to the circulatory disturbances and damage to the renal epithelium. The main lesion occurring in the proximal convoluted tubules of the kidney is the reversible necrosis.

Pulmonary complications can commonly occur and sometimes even prove to be fatal. Damage to the pulmonary capillaries may result in pulmonary oedema. This may cause a rise in vascular resistance and impaired oxygenation of the pulmonary blood. The situation is further aggravated due to patchy atelectasis, opening of the arteriovenous shunts and pneumonitis (Staphylococcal and Klebsiella).

Skin Lesions

Characteristic skin lesions associated with specific pathologies are described in **Table 5.10**.

FIG. 5.13: Response to inflammatory mediators in septic shock
Abbreviations: TNF-α, tumour necrosis factor-alpha; IL, interleukin; PAF, platelet activating factor

Diseases of Lymph Nodes

Generalised Lymphadenopathy

Various causes for generalised lymphadenopathy include syphilis, HIV, lymphomas and Q fever. Toxoplasmosis is a commonly acquired infection, which is characterised by mild chronic febrile illness and a localised group of enlarged lymph nodes. Though congenital diseases and the immunocompromised states may also be associated with generalised lymphadenopathy, often there is also an accompanying splenomegaly. Generally, infection with Epstein-Barr virus or glandular fever causes cervical lymphadenopathy although generalised lymphadenopathy is also sometimes recognised.

Tumours of the Genital Tract

Most tumours in adults (both benign and malignant) are derived from the epithelium normally present at that site. Therefore, it is important to have an idea regarding the type of epithelium lining the various genital organs in a woman. **Table 5.11** tabulates the different types of epithelium lining the various organs of the genital tract.

Vulva

Vulvar cancer affects the vulva, an area of external female genitalia. The most common histological subtype of vulvar cancer is squamous cell cancer. Vulvar cancer usually occurs after menopause. The average age at diagnosis is 70 years. The main risk factor for developing vulvar cancer is the presence of precancerous/dysplastic changes and chronic inflammation associated with lichen sclerosus, etc. in the vulvar tissues. Preinvasive epithelial changes termed as vulvar intraepithelial neoplasia (VIN) have been recognised and are related to human papillomavirus (HPV) infection, similar to cervical intraepithelial neoplasia (CIN). This is associated with various features of cytological atypia such as pleomorphism, abnormal mitosis, high nuclear to

TABLE 5.10	Pathology of various skin lesions	
Skin lesion	**Description**	**Disease**
Erythema marginatum	Nonitchy, pale red, macular eruptions	Major criterion for the diagnosis of rheumatic fever but is also seen in acute glomerulonephritis and drug reactions
Erythema nodosum	Tender red swellings usually over the shins	Tuberculosis, sarcoidosis, leprosy, reaction to sulphonamides and inflammatory bowel disease
Erythema multiforme	Characterised by cutaneous "target" lesions and mucosal involvement	Orf, mycoplasma and herpes simplex
Erythema infectiosum	Associated with "slapped cheeks" appearance	Parvovirus B19
Erythema induratum	Nodular eruption usually over the lower legs	Cutaneous tuberculosis (also known as Bazin's disease)
Erythema chronicum migrans	An expanding annular lesion	Lyme's disease

Types of epithelium	Specific genital tract structure
Simple squamous epithelium	Blood and lymphatic vessel, and peritoneum
Simple cuboidal epithelium	Ovary and uterine endometrium in the infant
Simple columnar epithelium	Uterine endometrium and cervical canal
Simple ciliated columnar epithelium	Fallopian tube
Stratified squamous epithelium	Vaginal surface of cervix, vagina and vulva
Transitional epithelium	Ureter, bladder and urethra

TABLE 5.11 Types of epithelium lining the genital tract in a woman

cytoplasmic ratio, etc. The basement membrane, however, remains intact in cases of VIN.

The cancer usually presents as a thickened or an ulcerated area on the vulva. Microinvasive carcinoma of vulva can be described as lesions less than or equal to 2 cm with less than 1 mm of stromal invasion. When the degree of stromal invasion is greater than 1 mm, there is a high probability of lymph node metastasis. In 50% of cases, presentation is in the form of a lump or a mass along with a long-standing history of pruritus, which may be related to vulvar dystrophy. In 60% of the cases, the lesion is in labia majora; 20% of the cases in labia minora; 12% of the cases in the clitoris and 6% of the cases in the perineum. The vulvar cancer can spread by direct extension to the adjacent structures, such as vagina, urethra and anus, by lymphatic route to adjacent lymph nodes and via haematogenous route to distant organs such as lungs, liver and bone. Lymphatic metastasis occurs early in the disease and most commonly occurs to the inguinal group of lymph nodes. From here, the spread can occur to the femoral group of lymph nodes. Surgical staging of vulvar cancer consists of excision of the primary lesion and inguinofemoral lymph node evaluation.

Depending on the extent and type of the cancer, vulvectomy is performed. Lymphadenectomy may be also done depending upon the involvement of lymph nodes. For early stage cancers, such treatment is usually all that is required. However, for more advanced cancers, radiation therapy, along with cisplatin chemotherapy is usually required. Surgical procedures for the treatment of vulvar cancer include wide local excision, simple partial vulvectomy, radical partial vulvectomy, en block radical vulvectomy and radical complete vulvectomy. After the removal of the cancerous tissues, surgical reconstruction of the vulva and vagina may be performed.

Vagina

Vaginal neoplasms are usually rare and share many similarities with the cervical cancer. However, since the vagina is normally lined by non-keratinising squamous epithelium, malignancy if occurs is usually squamous cell carcinoma. It usually occurs in elderly women and presents as an ulcerative or fungating growth in the upper third. Lymphatic and local spread can commonly occur. The premalignant lesion in the vagina which can be described as a sequel to malignancy is known as VAIN (vaginal intraepithelial neoplasia). Radiation therapy forms mainstay of treatment. Radical surgery may sometimes be required in a selective group of patients.

Cervix

Cervical Intraepithelial Neoplasia

Cervical intraepithelial neoplasia (CIN) is a premalignant condition of the uterine cervix that arises from the area of metaplasia in the transformation zone at the squamocolumnar junction. CIN refers to squamous cell abnormalities. Diagnosis of cervical dysplasia/cervical intraepithelial neoplasia is mainly based on cytological screening (Papanicolaou test or Pap smear) of the population.

The normal ectocervix is covered with squamous epithelium, whereas the endocervix is covered with columnar type epithelium. The squamocolumnar junction represents the transformation zone of the cervix where columnar epithelium of endocervical canal meets the squamous epithelium of ectocervix. The reserve cells lying beneath the columnar epithelium at this junction, sometimes, transform into mature squamous cells through the process known as metaplasia. When this process of maturation becomes abnormal, it is termed as dysplasia. While dysplasia acts as a precursor of malignancy, metaplasia does not progress to invasive cancer.

The peak incidence of occurrence of dysplasias appears to be 10 years earlier than that of frank invasive cancer. Dysplasias can be graded as follows:
- *Mild dysplasia or CIN 1*: The undifferentiated cells are confined to the lower one-third of the epithelium. The cells are more differentiated towards the surface.
- *Moderate dysplasia or CIN 2*: Undifferentiated cells occupy the lower 50–75% of the epithelial thickness.
- *Severe dysplasia and carcinoma in situ or CIN 3*: In this grade of dysplasia, the entire thickness of epithelium is replaced by abnormal cells; there is no cornification and stratification is lost. The basement membrane, however, remains intact and there is no stromal infiltration.

Invasive carcinomas usually follow CIN 3. They initially spread locally, often presenting as a fungating or an ulcerative growth. The cancer spread later occurs via the lymphatic route.

Cervical Cancer

Cervical cancer develops from the cervix. Cervical cancer usually results from infection with the HPV transmitted

at the time of sexual intercourse. Due to the presence of various epithelial types in cervix, several malignant growths can occur in cervix, including squamous cell carcinoma, adenocarcinoma and sarcoma. Nevertheless, squamous cell carcinoma is the most common type of carcinoma affecting the transformation zone. Cancer of cervix usually is the end stage of the spectrum of disorders progressing from mild through moderate to severe dysplasia and cervical intraepithelial neoplasia grade 3 (CIN 3). This cancer may result in abnormal bleeding, such as irregular vaginal bleeding, postcoital bleeding, bleeding in between periods, etc. Papanicolaou (Pap) test is a screening test, which helps in detecting cervical abnormalities at an early stage. Regular testing with Pap smears and HPV vaccination can help prevent cervical cancer. Peak age for the development of cancer cervix is 60 years, with CIN developing around 20 years earlier. Some risk factors for cervical cancer are young age at the time of first sexual intercourse, having multiple sexual partners, history of smoking cigarettes and having disorders of immune system (e.g. AIDS).

Uterus
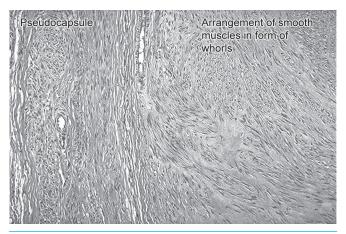
Endometrium

Endometrial cancer develops from the lining of the uterus, also known as the endometrium. It is the most common gynaecologic cancer and the fourth most common cancer amongst women. Endometrium is composed of numerous glands set within a background stroma. The structure of uterine endometrium varies throughout the menstrual cycle under the influence of hormones such as oestrogen and progesterone. In the first half of menstrual cycle (also known as the follicular phase), oestrogens cause proliferation of the endometrial endothelium. In the second half of menstrual cycle, secretory changes occur in the uterine endometrium under the influence of progesterone. As would be expected, the commonest malignancy occurring at this site would be endometrial adenocarcinoma. The probable precursor lesion for endometrial adenocarcinoma is endometrial hyperplasia, which may occur in the presence of high oestrogen states. The risk is greatest in cases of atypical endometrial hyperplasia, which is characterised by both architectural and cytological abnormalities. Approximately 1 in every 50 women is likely to get affected with the endometrial cancer. The most common symptom associated with endometrial cancer is abnormal uterine bleeding. Endometrial cancer usually affects women after menopause, commonly in the age group of 50–65 years.

Myometrium

The most common tumours arising from the myometrium are the benign tumours also known as leiomyomas or uterine fibroids. Uterine leiomyomas (uterine myomas, fibromyomas or fibroids) are well-circumscribed benign tumours developing from uterine myometrium, most commonly encountered amongst women of reproductive age group (30–44 years). A typical myoma is a pale, firm, rubbery, well-circumscribed mass distinct from neighbouring tissues and has a whorled appearance due to presence of interlacing fibres of myometrial muscle, surrounded by a connective tissue capsule **(Fig. 5.14)**. There are three types of fibroids **(Fig. 5.15)**. Of the different types of fibroids, the most common are intramural or interstitial fibroids (which are present within the uterine myometrium), followed by submucosal fibroids (which grow beneath the uterine endometrial lining) and subserosal fibroids (which grow beneath the uterine serosa).

Another lesion, which may be commonly present within the myometrium, is adenomyosis. It is characterised by the presence of nests or nodules of endometrium within the myometrial tissues (usually >2.5 mm beneath the basal endometrium). It is associated with myometrial hypertrophy, which may be either diffuse, or localised (adenomyoma).

FIG. 5.14: Histological appearance of a fibroid showing presence of interlacing smooth muscle fibres surrounded by varying amount of connective tissue
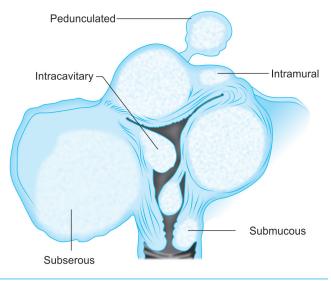
FIG. 5.15: Different types of fibroids

Ovary

In the United States, ovarian cancer is the second most common gynaecologic cancer. Ovarian cancer usually does not cause symptoms, until it is large or is in an advanced stage. Hence, cancer of the ovaries has the worst prognosis in comparison to any other type of gynaecologic cancer. As a result, it is the fifth most common cause of cancer deaths in women. This type of cancer develops most after in the elderly women aged 50–70 years. Some of the risk factors for ovarian cancer include old age, nulliparity, having the first child late in life, early menarche, late menopause and family history of cancer of the uterus, breast or large intestine. There are many types of ovarian cancer. Nearly 80% of the cancers are epithelial cell cancers, which begin from the surface epithelium of the ovaries. Ovarian carcinomas may differentiate along various pathways thereby resulting in the development of serous, mucinous or endometrioid adenocarcinomas.

Other types of ovarian cancers include the germ cell tumours or the stromal cell tumours. Benign epithelial tumours (cystadenomas) may also occur. Besides, a group of epithelial tumours of intermediate malignancy, also known as borderline tumours may sometimes occur. Sex cord stromal tumours represent neoplasms of specialised stromal cells such as granulosa cells, sertoli cells, theca cells, leydig cells or specialised fibroblasts. Germ cell tumours arise from totipotent germ cells. The majority of germ cell tumours are benign cystic teratomas, also known as dermoids. Dysgerminoma is another type of germ cell tumour of the ovary which is usually malignant in nature.

The ovarian cancer is one of the most aggressive types of cancers, which can spread directly to the surrounding tissues and through the lymphatic system to other parts of the pelvis and abdomen. It can also spread through the bloodstream to the distant body organs, mainly the liver and lungs. Many women with ovarian cancer may not have any symptoms, until the cancer is in an advanced stage. Moreover, if the symptoms do appear, they may be vague such as lower abdomen discomfort, indigestion, bloating, loss of appetite, backache, etc. Ovarian cancer rarely causes vaginal bleeding.

Gynaecological Abnormalities

Pathology of Miscarriage

A miscarriage (also known as spontaneous abortion) is any pregnancy, which undergoes spontaneous termination before reaching the period of viability (24 weeks). Different types of miscarriages are as follows:
- *Threatened abortion*: The process of abortion has started, but has not progressed towards completion.
- *Inevitable abortion*: The process of abortion has progressed to a stage from where continuation of pregnancy is impossible.
- *Incomplete abortion*: In this type of abortion, products of conception have not been fully expelled out of the uterine cavity and can be felt through the cervical os.
- *Complete abortion*: There is expulsion of products of conception en masse, following which there is subsistence of pain or bleeding. On per vaginal examination, the cervical os is closed and uterus is smaller than the period of amenorrhea.
- *Missed abortion*: The foetus is dead and gets retained inside the uterine cavity.

Aetiopathogenesis

Pathological examination of the aborted products of conception may help in identifying the underlying pathology for miscarriage. Pathological examination has demonstrated that a common mechanism for first trimester miscarriage is defective trophoblastic invasion of the decidual and uterine vasculature. Although the actual cause of the miscarriage is frequently unclear, the most common reasons include the following:
- *Genetic factors*: Chromosomal abnormalities are probably the most common underlying cause for first and early second trimester miscarriages. These include chromosomal anomalies such as trisomy, polyploidy, monosomy, structural chromosomal aberrations, etc.
- *Endocrine and metabolic disorders*: Endocrine disorders, such as luteal phase defects, thyroid anomalies, diabetes mellitus, etc.
- *Infections*: Acute viral infections, such as German measles, cytomegalovirus, variola, vaccinia, HIV, mycoplasma, etc. and bacterial infections such as ureaplasma, chlamydia, brucella, spirochaetes, etc. can also cause miscarriage. Ascending infection of the genital tract with either localised inflammation in the region of cervical os or chorioamnionitis is the most common cause for late second-trimester spontaneous miscarriage.
- *Anatomical abnormalities*: These include diseases and abnormalities of the internal genital organs, such as cervical incompetence, congenital malformations of the uterus (bicornuate or septate uterus), submucous fibroids and intrauterine adhesions (Asherman's syndrome).
- *Immunological disorders*: Autoimmune and alloimmune disorders including antiphospholipid syndrome are usually responsible for causing second trimester miscarriage.
- *Other factors*: These may include emotional factors or factors causing stress, certain drugs, caffeine, alcohol, tobacco, cocaine, etc. Maternal exposure to external agents such as drugs or radiation could be another cause for first-trimester miscarriage.
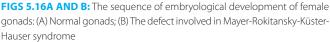
Pathology of Common Congenital Abnormalities

Müllerian Agenesis (Mayer-Rokitansky-Küster-Hauser Syndrome)

There is usually an absence or hypoplasia of the internal vagina and absence of Fallopian tubes and uterus in Mayer-Rokitansky-Küster-Hauser Syndrome (MRKH syndrome). This syndrome occurs due to defect in fusion of the Müllerian ducts resulting in absence of proximal one-third of vagina with or without the uterus **(Figs 5.16A and B)**. Since the ovaries are not Müllerian structures, they are normal. This may be the probable diagnosis in an individual with primary amenorrhea and no apparent vagina. This syndrome occurs in approximately 1 in 5,000 women.

The cause of this syndrome is unknown and is probably related to the mutations in the gene for anti-Müllerian hormones or the gene for anti-Müllerian hormone receptor. As a result, the affected women have normal chromosomal pattern (46XX). Other anomalies including the renal tract anomalies such as ectopic kidney, renal agenesis, horseshoe kidney and abnormal collecting ducts are frequently present. Extirpation of the Müllerian remnants, if any present, is not required unless they are causing some problems such as fibroid growth, hematometra, endometriosis, etc. Treatment of the condition usually involves progressive dilatation using Frank's dilators. Initially, the dilatation is begun in posterior direction and then after 2 weeks, it is changed to upward direction in the line of vaginal axis. This must be performed daily for 20 minutes to the point of modest discomfort. By utilising increasingly larger sized dilators, a functional vagina can be created within a period of several months. Operative treatment is used in the patients in whom the Frank's method is unacceptable or fails. It is important for the gynaecologist to provide adequate reassurance and support in these cases. Adequate counselling helps in avoiding problems with altered body image, which are likely to develop in these cases. Creating an artificial vagina either through the use of Frank's dilators or surgical procedure (McIndoe's vaginoplasty) at the time the patient plans to get married, helps in ensuring that she and her partner would be able to obtain adequate sexual enjoyment following their marriage. Having regular sexual intercourse helps in maintaining the patency of newly created artificial vaginal orifice. Though the patient remains infertile, she can lead an almost normal life. Genetic offsprings can be achieved by

FIGS 5.16A AND B: The sequence of embryological development of female gonads: (A) Normal gonads; (B) The defect involved in Mayer-Rokitansky-Küster-Hauser syndrome

collecting oocytes from genetic mother, fertilising them with sperms obtained from genetic father and their placement in a surrogate carrier.

Choose the Single Best Answer (SBA)

Q 1. Which of the following is not true regarding polymerase chain reaction?
A. DNA or RNA can be used as the template
B. Helps in diagnosis of infection
C. In diagnostic PCR, the exact sequence at both ends of the target region must be known
D. Polymorphisms in the viral genome may result in amplification failure
E. Takes several days to complete

Q 2. Chemical mediators concerned in production of an inflammatory response include which of the following?
A. Globulin permeability factor
B. Bradykinin
C. 5-Hydroxytryptamine
D. All the above
E. None of the above

Q 3. Which of the following is true regarding amyloidosis?
A. Is a type of coagulative necrosis
B. Granulation tissue is a feature of amyloidosis
C. The amyloid deposits lie around blood vessels
D. Renal failure may be the presenting complaint
E. Rarely affects the liver

Q 4. Which of the following is true regarding inflammation?
A. Following trauma, there may be initial vasodilatation followed by vasoconstriction
B. A cell-free plasmatic zone adjacent to the endothelium of venules is only seen in normal cells
C. The margination of white cells phenomenon is very characteristic of chronic inflammation
D. Pyaemia is an essential feature of abscess formation
E. Kinins cause relaxation of the smooth cells

Q 5. Which of the following statement regarding berry aneurysm is correct?
A. Are associated with diabetes mellitus
B. Are rarely associated with polycystic renal disease
C. Most often found in the circle of Willis
D. Result from abnormalities in the intimal wall of the arteries
E. Result from atheroma

Q 6. Which of the following is not a predisposing factor for atherosclerosis?
A. Cigarette smoking
B. Diabetes mellitus
C. Hormone replacement therapy
D. Hypertriglyceridemia
E. Systemic arterial hypertension

Q 7. Which of the following pathogens is commonly isolated from intra-abdominal pus?
A. *Actinomyces*
B. *Bacillus*
C. *Clostridia*
D. None of the above
E. All the above

Q 8. Which of the following tissues is not capable of cellular regeneration?
A. Bone marrow
B. Epidermis
C. Liver
D. Myocardium
E. skin

Q 9. Which of the following elicits a febrile response?
A. Corticosteroids
B. Interleukin
C. C reactive protein
D. All the above
E. None of the above

Q 10. Rigors are characteristic feature of which of the following?
A. Acute cholecystitis
B. Acute pancreatitis
C. Acute pyelonephritis
D. Hodgkin's disease
E. Ureteric calculi

Q 11. Wound healing by secondary intention takes place in which of the following circumstances?
A. When the wound becomes infected
B. When the wound does not break apart
C. When the wound edges are brought together
D. When there is irreparable skin loss
E. All the above

Q 12. Which of the following regarding the pathogenesis of thrombosis is correct?
A. Contact with subendothelial collagen causes platelet aggregation
B. Prostacyclin induces platelet aggregation
C. Thrombin inhibits platelet aggregation
D. All the above
E. None of the above

Q 13. Which of the following disease is not associated with HLA-B8?
A. Graves' disease
B. Insulin-dependent diabetes mellitus
C. Multiple sclerosis
D. Myasthenia gravis
E. Sjogren's syndrome

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1. E   2. D   3. C   4. B   5. C   6. C   7. C   8. D   9. B   10. C   11. A   12. A   13. C

Q 14. Which of the following is true regarding acute tubular necrosis?
A. Shock is a cause
B. The blood urea nitrogen/creatinine ratio is greater than 20
C. The urinary sodium is less than 20 mmol/L
D. Urine osmolality is greater than 500 milliosmoles/L
E. Urine specific gravity is greater than 1.010

Q 15. Which of the following is cause of generalised lymphadenopathy (LAP)?
A. HIV seroconversion illness
B. Q fever
C. Syphilis
D. *Toxoplasma gondii*
E. All the above

Q 16. Which obstetric complication has an increased prevalence in women with bicornuate uterus?
A. Breech presentation
B. Postpartum haemorrhage
C. Placenta accreta
D. Placenta praevia
E. Spontaneous miscarriage

Q 17. Within what timeframe from injury do macrophages replace neutrophils in case of cutaneous wound healing?
A. 2–3 hours
B. 6–12 hours
C. 18–24 hours
D. 48–96 hours
E. 8–10 days

Q 18. Which of the following tumour is hormone dependent?
A. Malignant melanoma
B. Adenocarcinoma of the prostate
C. Adenocarcinoma of the pancreas
D. None of the above
E. All the above

Q 19. Which of the following is true regarding sarcomas?
A. They are slow growing tumours
B. Include gastrointestinal stromal tumours
C. Rarely metastasise to lungs
D. Originates from embryonic ectoderm
E. Respond poorly to chemotherapy

Q 20. Which of the following statement is true regarding cell necrosis?
A. Caseous necrosis can occur in presence of *mycobacterium tuberculosis*
B. Is a natural sequelae of the cell cycle
C. Is not associated with the release of inflammatory mediators
D. Is reversible
E. Does not involve any changes in the nucleus

Q 21. Apoptosis is characterized by which of the following?
A. Cell swelling
B. Karyorrhexis
C. Release of inflammatory mediators
D. None of the above
E. All the above

Q 22. Which of the following is true regarding hyperplasia?
A. It can be reversible
B. It occurs in the adrenal cortex in the sufferers of Cushing's syndrome
C. It occurs in the uterus during pregnancy
D. All the above
E. None of the above

Q 23. Which of the following regarding cellular function is not correct?
A. Atrophy can be reversible
B. Dysplasia can also be reversible
C. Metaplasia is the conversion of fully differentiated cell type into another differentiated cell type
D. Hypertrophy is the increase in cell size
E. Neoplasia represents malignant change

Q 24. Which of the following regarding metaplasia is correct?
A. Is synonymous with heteroplasia
B. In the cervix, it describes change from columnar to transitional epithelium
C. It is the change of one differentiated cell type into an undifferentiated type
D. Can be reversible
E. Represents malignant change

Q 25. Which of the following term best describes the renal pathology of preeclampsia?
A. Atheromatous plaques
B. Glomerular hypertrophy
C. Glomerular capillary endotheliosis
D. Tubular vacuolization
E. Mesangial cell hypertrophy

Q 26. Which of the following statement is correct regarding Conn's syndrome?
A. Is caused by squamous cell carcinoma of adrenal glands
B. Is characterized by profound hypotension
C. Shows no response to spironolactone
D. Is associated with excessive production of aldosterone
E. Can cause hyperkalemia

Q 27. Which of the following vulval skin disorders is associated with the highest risk of malignancy?
A. Lichen planus
B. Lichen sclerosus
C. Psoriasis
D. Squamous cell metaplasia
E. Contact irritant dermatitis

14. A   15. E   16. A   17. D   18. B   19. B   20. A   21. B   22. D   23. E   24. D   25. C   26. D   27. B

Q 28. **Which of the following paraneoplastic syndromes is correctly paired with a recognized causal malignancy?**

A. Acanthosis nigricans and bowel cancer
B. Carcinoid and fibrosarcoma
C. Cushing's syndrome and small cell carcinoma
D. Dermatomyositis and renal cancer
E. Syndrome of inappropriate ADH secretion and testicular cancer

Q 29. **A 65-years-old woman presented to the GP with the complaints of abdominal distension, reduced appetite and shortness of breath. The GP, suspicious of malignancy, referred her to the oncology department of the local hospital and ordered CA-125 levels. CA-125 levels were found to be 820 U/mL. What is the likely diagnosis in this case?**

A. Colorectal cancer
B. Hepatocellular cancer
C. Gastric cancer
D. Primary peritoneal cancer
E. Pancreatic cancer

Q 30. **Which of the following statement is not correct regarding pheochromocytomas?**

A. High serum levels of metanephrine are diagnostic
B. They are corticosteroid producing tumours
C. Commonly develop in chromaffin cells of renal medulla
D. Surgical resection is the treatment of choice
E. May present with severe hypertension and palpitations

Q 31. **What is the genotype in a case of complete molar pregnancy?**

A. 45 XO
B. 46XX
C. 46 XY
D. 46XXX
E. 69XXY

Q 32. **Which of the following is not correct regarding choriocarcinoma?**

A. It is a malignant condition
B. Is more common in women above the age of 40 years
C. Can follow a normal pregnancy
D. Can commonly metastasise to the brain
E. Syncytiotrophoblasts are filled with eosinophilic material

Q 33. **Which of the following statement is correct regarding the congenital absence of the uterus?**

A. Has an incidence of 1:1,000 births
B. Has a chromosomal pattern of 45 XO
C. Hirsutism is commonly observed
D. Is also known as Mayer-Rokitansky-Küster-Hauser syndrome
E. The ovaries are commonly affected

Q 34. **Which of the following diseases is not associated with HLA-B8?**

A. Graves' disease
B. Ankylosing spondylitis
C. Insulin-dependent diabetes mellitus
D. Myasthenia gravis
E. Sjogren's syndrome

Q 35. **Which of the following is correct concerning shock?**

A. There is metabolic alkalosis
B. Hypoxia may not be present in some cases
C. Capillary permeability is reduced
D. Hypokalaemia occurs
E. There may be coagulopathy

Q 36. **In the presence of inflammation, which of the following is raised?**

A. Caeruloplasmin
B. Complement proteins
C. Ferritin
D. Fibrinogen
E. All the above

28. C   29. D   30. B   31. B   32. D   33. D   34. B   35. E   36. E.

Microbiology and Immunology

Bacteria

The bacteria can be classified into two major groups based on their ability to take up the Gram stain. The bacterium having peptidoglycan in its cell wall is able to take up Gram stain and is therefore considered as Gram positive. On the other hand, bacterium not capable of taking the Gram stain is classified as Gram negative. Besides Gram staining, bacteria can also be classified based on their appearance as cocci (spherical in shape) and bacilli (rod-shaped) **(Table 6.1)**.

Most of the bacterial organisms are aerobic, i.e. they require the presence of oxygen. Some of the bacterial organisms, however, may be anaerobic. These can be either obligate anaerobes or facultative anaerobes **(Table 6.2)**. Obligate anaerobes are organisms that live and thrive in the absence of oxygen; such organisms may die in the presence of oxygen. On the other hand, facultative anaerobes are those organisms which are able to alter their function depending on the presence or absence of oxygen.

TABLE 6.1	Common forms of bacteria			
Group	**Genus name**	**Species**	**Gram stain**	**Disease caused**
Cocci	*Streptococcus* (α-haemolytic)	*Streptococcus pneumoniae*	Positive	Lobar pneumonia
		S. milleri	Positive	Normal flora of the mouth, deep-seated abscesses and endocarditis
		S. faecalis/Enterococcus faecalis (enterococcus)	Positive	Normal flora of the bowel, urinary tract infection, opportunistic wound infection, endocarditis, bacteraemia and abdominal infections
		S. viridans	Positive	Dental caries; endocarditis
	Streptococcus (β-haemolytic)	*S. pyogenes* (Group A)	Positive	Upper respiratory tract infection, pharyngitis, pyoderma, erysipelas, cellulitis, necrotizing fasciitis, streptococcal toxic shock syndrome, bacteraemia, rheumatic fever, glomerulonephritis, wound infection, abscesses, bacteraemia/septicaemia, puerperal sepsis, necrotizing fasciitis, scarlet fever and septic arthritis
		S. agalactiae (Group B streptococci)	Positive	Normal flora of the vagina, neonatal bacteraemia/septicaemia, meningitis and chorioamnionitis, neonatal sepsis, meningitis, puerperal fever and pyogenic infection
	Peptostreptococcus	*P. anaerobius*	Positive	Anaerobic abscesses
	Staphylococcus	*S. aureus*	Positive	Wound infections, abscess, bacteraemia/septicaemia, osteomyelitis, TSS and food poisoning
		S. epidermidis	Positive	vascular cannula-associated infection
		S. saprophyticus	Positive	Urinary tract infections

Contd...

Contd...

Group	Genus name	Species	Gram stain	Disease caused
Cocci	Neisseria	N. gonorrhoeae	Negative	Gonorrhoea, pelvic inflammatory disease, arthritis, bacteraemia/septicaemia, infertility, neonatal ocular infection and suppurative urethritis (in males)
		N. meningitidis	Negative	Meningitis, meningoencephalitis, bacteraemia, pneumonia, arthritis and urethritis
	Moraxellae	Moraxella catarrhalis (Branhamella)	Negative	Respiratory flora, exacerbations of chronic bronchitis
	Veillonella	Veillonella spp.	Negative	Normal oropharyngeal flora
Rods	Mycobacteria	M. tuberculosis	Positive	Tuberculosis
	Listeria	L. monocytogenes	Positive	Maternal and neonatal listeriosis
	Lactobacillus	L. acidophilus	Positive	Normally occurs in the human and animal gastrointestinal tract and mouth
		L. casei	Positive	Normal vaginal flora
	Corynebacteria	C. diphtheriae		Diphtheria
		C. jeikeium		Skin flora, cannula/vascular associated bacteraemia/septicaemia
	Bacillus	B. anthracis	Positive	Anthrax
		B. cereus	Positive	Food poisoning with diarrhoea and vomiting
	Clostridium	C. perfringens	Positive	Gas gangrene
		C. tetani	Positive	Tetanus
		C. difficile	Positive	Pseudomembranous colitis
	Actinomycetes	Actinomyces israelli	Positive	Pelvic actinomycosis
	Nocardia	N. asteroides	Positive	Chronic infection in transplant patients
Rods	Enterobacteriaceae • Proteus • Escherichia • Klebsiella • Salmonella • Shigella	P. mirabilis E. coli K. pneumoniae S. typhi S. dysenteriae	Negative	Urinary tract infections Urinary tract infections, gastroenteritis Pneumonia (bronchopneumonia and bronchitis) Enteric fever Dysentry
	Pseudomonas	P. aeruginosa	Negative	Nosocomial urinary tract and respiratory tract infection, opportunistic wound infection, bacteraemia and septicaemia
	Brucella	B. abortus	Negative	Brucellosis or undulant fever
	Bacteroides	B. fragilis	Negative	Intra-abdominal infections, perirectal abscess, decubitus ulcers
	Gardnerella	G. vaginalis	Negative	Bacterial vaginosis
	Yersinia	Y. pestis	Negative	Plague
		Y. enterocolitica	Negative	Mesenteric adenitis
	Pasteurella	P. multocida	Negative	Animal bites
	Legionella	L. pneumophila	Negative	Atypical pneumonia
	Campylobacter	Campylobacter jejuni	Negative	Gastroenteritis
	Haemophilus	H. influenzae	Negative	Respiratory flora, exacerbations of chronic bronchitis
	Bordetella	B. pertussis	Negative	Pertussis/whooping cough
	Bartonellae	B. Henselae	Negative	Cat-scratch disease, bacillary angiomatosis
Spirochetes	Treponema Leptospira	T. pallidum L. interrogans	Negative Negative	Syphilis Leptospirosis
Vibrios (comma-shaped)	Vibrio	V. cholerae	Negative	Cholera

Abbreviation: TTS, toxic shock syndrome

TABLE 6.2	Anaerobic bacteria
Strictly anaerobic bacteria (Obligate anaerobes)	**Facultative Anaerobic Bacteria**
• Clostridia	• *Staphylococcus aureus*
• Bacteroides	• *Escherichia coli*
• Actinomyces	• Listeria

Streptococci

Streptococci are penicillin-sensitive Gram-positive cocci arranged in form of chains **(Fig. 6.1)**. A number of species within this genus cause major human diseases. The most important amongst them is *Streptococcus pyogenes* causing pyogenic infections, with a characteristic tendency to spread, as opposed to staphylococcal lesions, which are typically localised. In addition, it is also responsible for causing the non-suppurative lesions, acute rheumatic fever and glomerulonephritis.

Streptococcus pyogenes produces several exotoxins and enzymes, which contribute to its virulence. Besides these, the M protein (protein antigen present in its cell wall) also acts as a virulence factor by inhibiting phagocytosis. The various toxins produced by *S. pyogenes* include streptolysin O, streptolysin S and pyrogenic exotoxins (erythrogenic, dick and scarlatinal toxins). They also produce enzymes such as streptokinase (fibrinolysin), deoxyribonucleases (streptodornase DNase), hyaluronidase, proteinase, serum opacity factor and nicotinamide adenine dinucleotidase (NADase).

Streptococcal Infection

Acute diseases associated with *Streptococcus pyogenes* occur chiefly in the respiratory tract, bloodstream, or the skin. Two post-streptococcal sequelae (rheumatic fever following respiratory infection and glomerulonephritis following respiratory or skin infection) may occur in 1–3% of untreated infections.
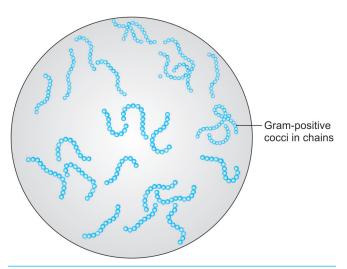
FIG. 6.1: Microscopic appearance of streptococci

Gram-positive cocci in chains

Respiratory Infections

These include infections such as sore throat, streptococcal pharyngitis, scarlet fever, suppurative complications (e.g. peritonsillar or retropharyngeal abscess, otitis media, mastoiditis, quinsy, Ludwig's angina (diffuse cellulitis of the floor of the mouth), suppurative adenitis, etc. Pharyngeal carriage rates vary with geographical location, season of the year and age group. Amongst school-aged children, rates of 15–20% have been reported; the carriage rate among adults is considerably lower.

Skin and Soft Tissue Infections

Streptococcus pyogenes causes a variety of suppurative infections of the skin, including infection of wounds or burns, with a predilection to produce lymphangitis and cellulitis. The two typical streptococcal skin infections are erysipelas (infection involving the superficial lymphatics) and impetigo. Other skin infections, which can occur, include necrotizing fasciitis (streptococcal gangrene), streptococcal toxic shock syndrome (TSS), etc.

Group A streptococci causing impetigo are frequently nephritogenic that leads to acute glomerulonephritis. The main causes leading to acute glomerulonephritis in children in the tropics are impetigo (pyoderma) and streptococcal infection of scabies lesions. Streptococcal TSS is another streptococcal soft tissue infection, typically involving the skin and deeper subcutaneous tissues, where the difference between infected and non-infected skin is not clear.

Classification of Streptococci

Classification of streptococci is described in **Figure 6.2**.

Group B β-haemolytic Streptococcus

Also known as *Streptococcus agalactiae*, this is a common commensal organism in the gastrointestinal tract and normal vaginal flora. As a result of its vaginal carriage, there is a risk of transmission of this organism to the foetus. This is associated with the potential to cause neonatal sepsis following the rupture of membranes.

Group B Streptococcal Infection in Pregnancy

Group B β-haemolytic Streptococcus (GBS) is the most common cause of major neonatal infection. But significant GBS infection only affects approximately 1:500 to 1:3,000 neonates. It is much more likely to affect premature babies and is particularly seen after premature rupture of membranes. It is often an evidence of maternal infection. GBS may also be associated with chorioamnionitis. Early onset disease with GBS in infants may manifest as pneumonia, septicaemia and meningitis.

Prophylactic administration of antibiotics to the mother results in the short-term elimination of the infection,

```
                    ┌─────────────────┐
                    │   Streptococci  │
                    └─────────────────┘
                             │
                      O₂ requirement
              ┌──────────────┴──────────────┐
     ┌────────────────────┐      ┌────────────────────┐
     │ Aerobes and         │      │ Obligate anaerobes  │
     │ facultative         │      │ peptostreptococci   │
     │ anaerobes           │      └────────────────────┘
     └────────────────────┘
              │
     ┌────────────┐
     │ Haemolysis │
     └────────────┘
       ┌──────────┴──────────────┐
┌────────────────────┐  ┌────────────────────┐
│ 1. Alpha (the       │  │ 3. Gamma (The       │
│ viridans group)     │  │ enterococcus group) │
│ classified into     │  │ classified into     │
│ species by          │  │ species by          │
│ physiological and   │  │ physiological and   │
│ biochemical         │  │ biochemical         │
│ properties          │  │ properties          │
└────────────────────┘  └────────────────────┘
              ┌──────────────┘
     ┌────────────────────┐
     │ 2. Beta            │
     │ (The haemolytic    │
     │ streptococci)      │
     └────────────────────┘
              │
     ┌────────────────────┐
     │ Serological group  │
     │ specific C         │
     │ carbohydrate       │
     │ antigen            │
     └────────────────────┘
              │
     ┌────────────────────┐
     │ 20 Lancefield      │
     │ groups             │
     │ (ABCDEFGHKLMNOPQRSTUV)│
     └────────────────────┘
              │
     ┌────────────────────┐
     │ Group A-streptococcus│
     │ pyogenes           │
     └────────────────────┘
              │
     ┌────────────────────┐
     │ Serological typing │
     │ (M protein)        │
     │ Griffith types     │
     │ (1,2,3, etc.)      │
     └────────────────────┘
```

FIG. 6.2: Classification of streptococci

but it returns rapidly. Also, there might be development of antibiotic resistant strains. So it is not considered worthwhile to screen every mother or to provide antenatal treatment. Moreover, it is difficult to get a positive culture result with GBS. Repeated culture swabs may be required to be taken, e.g. only 30% of cervical swabs will be positive in true carriers. Higher yields are obtained from rectal swabs. Routine screening of pregnant women for GBS is, therefore, currently not practiced in the UK.

There is growing evidence that the use of intrapartum antibiotics may prove to be effective. There are no fixed guidelines regarding the intrapartum use of antibiotics. The most commonly followed practice is the administration of antibiotics during labour to women who have had evidence of GBS in pregnancy, women with intrapartum pyrexia, women who have had prolonged rupture of membranes (> 24 hours), etc. Administration of antibiotics to women in premature labour is advocated by some. In the UK, if GBS is detected during the current pregnancy, the present recommendation by RCOG (Green Top guidelines, 2003) is administration of intrapartum antibiotic therapy in the dosage of 3 g IV penicillin G, followed by 1.5 g every 4 hourly in labour.

Group B Streptococcal Infection in the Newborn

Group B streptococcal infection in the newborn can cause symptomatic infection, which may be associated with 10–20% mortality. It can result in complications such as:

❏ Meningitis and neurological sequelae: The commonest aetiology for meningitis in the newborn babies is group B *Streptococci*, which may be acquired during or after delivery. The mortality is 5–15% in infants. Even those who survive may suffer from problems such as mental retardation, speech problems, visual impairment and neural deafness (rather than conductive deafness). Meningomyelocoele is a risk factor for the introduction of meningeal infection.

❏ Pneumothorax, persistent foetal circulation, and ARDS: a "ground glass" appearance is seen in 50% of cases with group B *Streptococcus* pneumonias, and patchy pneumonia is observed in approximately 30% cases. Apnoeic episodes are a frequent presentation of sepsis in the newborn.

❏ Arthritis and osteomyelitis.
  Conjunctivitis is rarely observed in these cases.

Vancomycin-resistant Enterococci

Enterococci were initially classified as enteric Gram-positive cocci and later classified as group D streptococci based on the Lancefield serological typing system. However, in the 1980s, enterococci were removed from the genus *Streptococcus* and placed in their own genus, *Enterococcus*. These bacteria have been recognised as an important cause of endocarditis and an important cause of nosocomial infection. Vancomycin-resistant enterococci (VRE) have emerged as a major therapeutic challenge due to their intrinsic resistance to most of the commonly used antibiotics. They cause clinical problems such as urinary tract infections (UTIs), bacteraemia, wound infections, neonatal infections, endocarditis, etc. These bacteria alter peptidoglycan precursors used for building the cell walls. Vancomycin binds to D-ala-D-ala but the resistant enterococci have D-ala-D-lac or D-ala terminating precursors. The inability to alter cell wall in VRE is due to the fact that the vancomycin-sensitive precursor genes have been turned off and the resistant ones only appear in the presence of vancomycin. They may also be found in healthy community volunteers not recently hospitalized (2% in the UK general practice). Community reservoir for this bacterium is present in meat, poultry and (?) cheese.

High-dose ampicillin is the treatment of choice only if the minimal inhibitory concentration of ampicillin is not too high. Anecdotal evidence exists for its use in *E. faecalis* endocarditis (20 g/day).

Staphylococcus Aureus

Staphylococci are Gram-positive cocci, non-motile bacteria that occur in grape-like clusters **(Fig. 6.3)**. It is a facultative
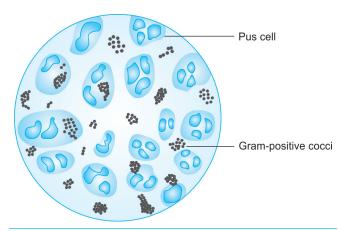
FIG. 6.3: *Staphylococcus* in a smear of pus

anaerobe, a non-spore-forming bacterium, which is mostly pathogenic. Species of staphylococci are classified by the coagulase test into two groups: the coagulase-positive (e.g. *S. aureus*) and coagulase-negative staphylococci (e.g. *S. epidermidis* and *S. saprophyticus*). The slide or tube coagulase test is performed to distinguish *S. aureus* from coagulase-negative species.

The strains of *S. aureus* usually exhibit following characteristics: (1) It produces a golden-yellow colony pigmentation and haemolysis on blood agar; (2) Coagulase positive; (3) Ferment mannite; (4) Liquefy gelatin; (5) Produce phosphatase; (6) Black colonies on potassium tellurite blood; (7) Produce thermostable nucleases which can be demonstrated by the ability of boiled cultures to degrade DNA in an agar diffusion test.

They are universally present and are the most common cause of localised suppurative lesions in human beings. Staphylococci may be typed, based on their susceptibility to bacteriophages. Staphylococci produce several toxins and enzymes which are responsible for its virulence. *S. aureus* produces a number of enzymes such as coagulase, catalase, hyaluronidase, fibrinolysin, lipases, nucleases, penicillinase and phosphatase. It also produces at least five cytolytic or membrane-damaging toxins [alpha, beta, delta, gamma and Panton-Valentine (P-V) leucocidin], an enterotoxin, toxic shock syndrome toxin-1 (TSST-1) and epidermolytic (exfoliative toxins).

Staphylococcal Infections

Cutaneous Infections

These include wound and burn infection, pustules (small cutaneous abscesses), furuncles or boils (large cutaneous abscesses), carbuncles, styes, impetigo and pemphigus neonatorum.

Deep Infections

These include: osteomyelitis, periostitis, tonsillitis, pharyngitis, sinusitis, bronchopneumonia, empyema, septicaemia, meningitis, endocarditis, breast abscess, renal abscess and abscesses in other organs.

Toxin-mediated Diseases

These include staphylococcal food poisoning, toxic shock syndrome (TSS) and exfoliative diseases [staphylococcal scalded skin syndrome (SSSS)]. TSS is a multisystem disease that primarily afflicts menstruating young women using tampons. The epidermolytic toxin produced by *S. aureus* is responsible for the staphylococcal scalded skin syndrome (SSSS), exfoliative skin disease in which the outer layer of epidermis gets separated from the underlying tissues.

Methicillin-resistant Staphylococcus Aureus

The ability of staphylococci to develop resistance to penicillin and other antibiotics enhances their importance as a human pathogen, especially in the hospital environment. Methicillin was the first compound developed to combat resistance due to penicillinase (beta lactamase) production by staphylococci. But strains of methicillin-resistant *Staphylococcus aureus* (MRSA) became common, which were resistant not merely to penicillin, but also to all other beta lactam antibiotics and many others. MRSA usually colonises wounds and venous access sites. Glycopeptides (vancomycin or teicoplanin) are the agents of choice in the treatment of systemic infection with MRSA.

Mycobacterium Tuberculosis

Mycobacteria are aerobic, rod-shaped, Gram-positive, non-motile, non-capsulated and non-sporing, slow growing bacteria. **Table 6.3** describes the classification of mycobacteria. It is highly resistant to drying. Mycobacteria

TABLE 6.3	Classification of mycobacteria

- Tubercle bacilli
  - Human—*M. tuberculosis*
  - Bovine—*M. bovis*
  - Murine—*M. microti*
  - Avium—*M. avium*
  - Cold blooded—*M. marinum*
- Lepra bacilli
  - Human—*M. leprae*
  - Murine—*M. lepraemurium*
- Mycobacteria causing skin ulcers
  - *M. ulcerans*
  - *M. balnei*
- Atypical mycobacteria
  - Photochromogens
  - Scotochromogens
  - Non-photochromogens
  - Rapid growers
- Johne's bacillus
  - *M. paratuberculosis*
- Saprophytic mycobacteria
  - *M. butyricum, M. phlei, M. stercoris.*

do not stain readily, but once stained with hot carbol fuchsin or other aryl methane dyes (Ziehl-Neelsen staining), they resist decolourization with dilute mineral acids (or alcohol). Mycobacteria are, therefore, known as acid-fast bacilli. The two most important species belonging to this genus are *Mycobacterium tuberculosis* and *Mycobacterium leprae,* the causative agents of tuberculosis (TB) and Hansen's disease (leprosy), respectively. The solid medium most widely employed for routine culture is Lowenstein-Jensen (LJ) medium without starch. This consists of coagulated hens' egg, mineral salt solution, asparagine and malachite green, glycerol or sodium pyruvate. On LJ media, *M. tuberculosis* forms dry, rough, raised, irregular colonies with a wrinkled surface, creamy white in colour, becoming yellowish or buff coloured on further incubation. The growth usually occurs within 4–6 weeks' time. They are tenacious and not easily emulsified. Mycobacteria protein (tuberculin) is responsible for development of delayed hypersensitivity in humans.

Infection produces both humoral and cell-mediated immunity. However, cell-mediated immunity is most important. Most commonly clinical tuberculosis represents delayed reactivation of the primary Ghon focus. *M. tuberculosis* causes primarily pulmonary tuberculosis. Tuberculosis of the genital tract is almost always a blood-borne infection from a focus elsewhere (usually in the lungs). The typical lesion of tuberculosis is a caseating granuloma known as the tubercle follicle. It usually begins with a primary focus in the lungs and may spread slowly through the lymphatics or rapidly through blood stream resulting in complications such as miliary tuberculosis, disseminated tuberculosis, tubercular meningitis, tuberculosis of the skin, tuberculosis of the middle ear and ocular structures, etc. Bacteriological diagnosis of tuberculosis can be established by direct microscopy, culture examination or by animal inoculation test. Sputum is the specimen of choice for diagnosis of pulmonary tuberculosis. Culture is the definite method to detect and identify *M. tuberculosis* and is sensitive and specific. Chemotherapy forms the mainstay of treatment of tuberculosis. Drug resistance to *M. tuberculosis* is developing due to mutation. The emergence of multidrug resistance-tuberculosis (MDR-TB) has become a very serious problem. The term multidrug resistance refers to resistance to rifampicin and isoniazid, with or without resistance to one or more other drugs.

Listeria Monocytogenes

It is a motile, Gram-positive bacillus, which is found in soft cheese and non-pasteurised milk. It is also found in salads and pre-cooked meat. The genus contains eight species but almost all cases of human listeriosis are caused by *L. monocytogenes.*

Pregnant women are nearly 20 times more likely than the non-pregnant to get this type of infection. Listeriosis is a common illness, which can occur throughout pregnancy and should be considered when the mother has a pyrexial flu-like illness. Many cases, however, may remain asymptomatic. It may particularly be associated with the signs of threatened premature labour. In addition to the risk from prematurity, there is an increased risk of intra-uterine and neonatal death.

It is a gut commensal, which is found in the intestinal tract of animals and humans. It resists freezing. It can proliferate at low temperature (can survive in temperatures of 4–60°C) or even in a refrigerator. Therefore, the pregnant woman must be advised not to eat food products stored at such temperatures, e.g. cooked meats, cooked-chilled meals, coleslaw, etc.

It produces haemolysins, therefore on blood agar, *L. monocytogenes* develops zones of slightly hazy β-haemolysis. The colonial appearances may be indistinguishable from those of group B streptococci, although listeriae are never pigmented. It can be diagnosed by blood culture. It is resistant to alkaline conditions. It has an ability to grow in salt in concentrations as high as 10%. Immunocompromised hosts are more susceptible to infection by *Listeria*.

Two types of listeriosis infection are described: "invasive" and "non-invasive". Most cases are "non-invasive" which fall in the category of "flu-like illness". Complete recovery usually occurs in these cases. "Invasive" listeriosis is totally different. It is a serious generalised infection affecting the immunocompromised, the pregnant and vulnerable groups like neonates and the elderly.

During pregnancy, this infection can cause miscarriage, stillbirth (up to 20% of cases), neonatal septicaemia and neonatal death. Infection is associated with an increased foetal mortality rate. Even if the baby is born alive, there is an increased risk of neonatal death with babies acquiring septicaemia and meningitis. In normal cases, meconium is rarely found in the liquor before 34 weeks. If found, it could be associated with the possibility of foetal infection producing enteritis. This is particularly associated with *Listeria*. It is treated with antibiotics such as ampicillin or co-trimoxazole or chloramphenicol.

Neisseria

Members of the genus *Neisseria* are aerobic, Gram-negative cocci typically arranged in pairs (diplococci) with adjacent sides flattened together (resembling coffee beans, e.g. *N. meningitidis*) **(Fig. 6.4)** or concave opposing edges (resembling kidney beans, e.g. *N. gonorrhoeae)* **(Fig. 6.5)**. All species are oxidase-positive, and most produce catalase. Important species of the genus *Neisseria* are *N. meningitidis* and *N. gonorrhoeae.* Both the species thrive in a moist environment having 5–10% level of carbon dioxide. While *N. gonorrhoeae* is always pathogenic, *N. meningitidis* may be found as a commensal inhabitant of the upper respiratory tract in approximately 10% population.

Neisseria meningitidis is transmitted via the droplet spread of respiratory secretions and may be associated with bacteraemia and meningitis. Rare manifestations include
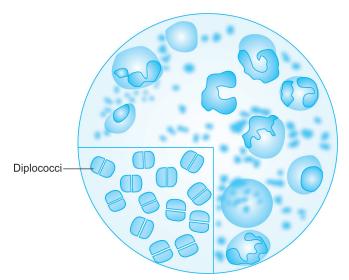
FIG. 6.4: *Neisseria meningitidis* in cerebrospinal fluid. Inset—enlarged view showing flat adjacent side of cocci
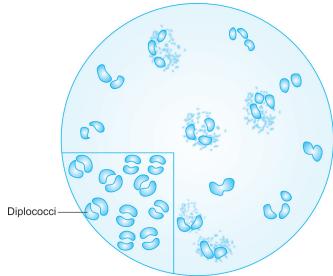
FIG. 6.5: *Neisseria gonorrhoeae* in urethral pus. Inset—enlarged view showing diplococci with adjacent surfaces concave

arthritis and osteomyelitis. Bacteriological diagnosis requires culturing of the cerebrospinal fluid obtained through lumbar puncture.

Gonococcus

Gonococci are Gram-negative diplococci seen within the cytoplasm of polymorphs. The outer surface of the bacteria is not covered with a true carbohydrate capsule, as is found in *N. meningitidis.* They grow well on chocolate agar and Thayer-Martin medium (chocolate agar containing vancomycin, colistin and nystatin) which inhibit most contaminants including non-pathogenic *Neisseria*. *N. gonorrhoeae* is a cause of the disease gonorrhoea.

Gonorrhoea is a venereal disease, acquired by sexual contact, having an incubation period of 2–8 days. Gonorrhoea is a disease essentially confined to the mucus-secreting epithelial cells of humans. Adhesion of gonococci to the urethra or other mucosal surfaces is the first step in infection. Gonococci possess pili on their surface, which facilitate adhesion of the cocci to mucosal surfaces and promote virulence by inhibiting phagocytosis. Stratified squamous epithelium (vagina) is relatively resistant to infection. Therefore, gonococcus infects the columnar epithelium (e.g. ectocervix). Hence a cervical swab is taken to investigate a case of suspected lower-genital tract gonorrhoea. Nevertheless, severe vulvovaginitis can still occur in prepubertal girls.

Gonococci can also attack mucous membranes of the genitourinary tract, eye, rectum, and throat, producing acute suppuration that may lead to tissue invasion. This is followed by chronic inflammation and fibrosis.

In the male, it may cause an acute suppurative urethritis and proctitis. The infection extends along the urethra to the prostate, seminal vesicles and epididymis. Although complications are rare; epididymitis, prostatitis and periurethral abscesses can occur. Chronic urethritis may lead to stricture formation. The infection may spread to the periurethral tissues, causing abscesses and multiple discharging sinuses (water-can perineum).

In women, endocervix is the primary site of infection and extends to the urethra and vagina, giving rise to mucopurulent discharge. Symptomatic patients commonly experience vaginal discharge, dysuria and abdominal pain. The infection may extend to skene's glands, bartholin's glands, endometrium and fallopian tubes. Peritoneal spread occasionally occurs and may produce a perihepatic inflammation *(*Fitz-Hugh-Curtis syndrome). Clinical disease is less severe in women, many of whom may carry gonococci in the cervix without developing any clinical symptoms. While asymptomatic carriage of gonococci commonly occurs in women, it is rare in men. Gonococci can also result in a non-venereal infection, ophthalmia neonatorum amongst the newborn. This may be associated with a severe purulent eye discharge and periorbital oedema within a few days of birth.

Currently, the Centers for Disease Control and Prevention (CDC) recommends that ceftriaxone, cefixime, ciprofloxacin, or ofloxacin may be used as the initial therapy for cases of uncomplicated gonorrhoea. Doxycycline or azithromycin should be added for infections complicated by dual infections with Chlamydia. Gonococci producing β-lactamase called penicillinase producing gonococci (PPNG) are plasmid-mediated. In β-lactamase-producing gonococci, treatment is with spectinomycin or cefoxitin. In case of resistance to these drugs, probenecid can also be administered.

Diplococci

Corynebacterium

Corynebacterium is Gram-positive bacilli with an irregular shape, tendency for clubbing at one or both ends, highly pleomorphic structure with Chinese letter or cuneiform arrangement **(Fig. 6.6)**. The granules in the cell are known as metachromatic granules, volutin granules or Babes-Ernst granules. Special stains, such as Albert's stain, have been devised for demonstrating the granules clearly. With Albert's stain, the granules stain bluish black and the protoplasm green.

The most important disease causing pathogen belonging to this genus is *C. diphtheriae,* which owes its pathogenicity to the production of a potent exotoxin active on a range of tissues, including heart muscle and peripheral nerves.

Two media are useful for culturing *Corynebacterium*: Löffler's serum slope and Tellurite blood agar. In the Löffler's serum slope, the diphtheria bacilli grow very rapidly and colonies can be seen in 6–8 hours. *C. diphtheriae* produce grey/black, shiny or dull black colonies on tellurite blood agar. Three different biotypes: gravis, intermedius and mitis are described.

Toxigenic strains of *C. diphtheriae* produce a very powerful exotoxin. Inhibition of protein synthesis is probably responsible for both the necrotic and neurotoxic effects of the toxin. Schick's test is used to distinguish susceptible individuals from those immune to diphtheria by injecting diphtheria toxins intradermally and observing for oedema and erythema (maximal in 2–4 days) in susceptible individuals.
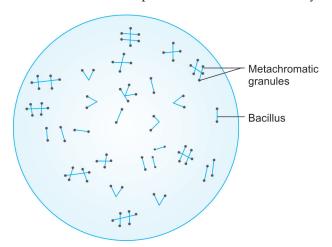
Diphtheria is primarily a paediatric disease having a worldwide distribution. Humans are the only known reservoir, with carriage in oropharynx or on skin surface. Spread occurs from person to person by exposure to respiratory droplets or skin contact. Diphtheria occurs in two forms (respiratory and cutaneous). In the respiratory form, a tough grey to white pseudomembrane may appear on the tonsils and then spread downwards into the larynx and trachea. Systemic effects involve the kidneys, heart and nervous system. It can result in complications such as asphyxia, acute circulatory failure, post-diphtheritic paralysis and septic complications, such as pneumonia and otitis media.

For protection against diphtheria, DPT vaccine and booster shots are administered. DPT is given at the age of 6 weeks, 10 weeks, 14 weeks and 16–24 months followed by booster dose of DT at the age of 5–6 years (school entry).

Diphtheroids are bacteria belonging to the genus *Corynebacteria* resembling *C. diphtheriae*, occurring as normal commensals in the throat, skin and other areas. These may be mistaken for diphtheria bacilli and hence are known as diphtheroids. They stain more uniformly than diphtheria bacilli, are arranged in V forms or palisades rather than Chinese letter arrangement and possess few or no metachromatic granules. The common diphtheroids are *C. pseudodiphtheriticum* and *C. xerosis*.

Clostridia

The genus *Clostridium* includes all obligatory anaerobic, Gram-positive bacilli capable of forming endospores. Clostridia are more commonly associated with skin and soft tissue infections, food poisoning and antibiotic-associated diarrhoea and colitis. Most *Clostridia* are normal commensals of human and animal gastrointestinal tracts, and are widely distributed in soil where, as spores, they may survive for years in adverse conditions. The shape and position of spores varies in different species and is useful in identification of clostridia. In the various species, the spore is placed centrally, subterminally, or terminally. Spores may be subterminal or central in *C. perfringens* and *C. botulinum;* drumstick appearance in *C. tetani* (terminal and spherical) **(Fig. 6.7)**, and oval and terminal in *C. difficile*.

FIG. 6.6: *Corynebacterium diphtheriae* showing metachromatic granules and Chinese letter arrangement
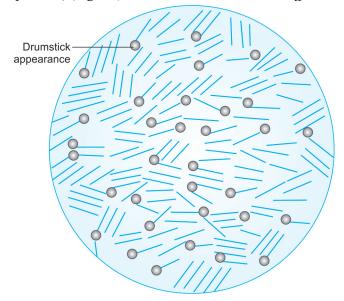
FIG. 6.7: Drum-stick appearance in case of *Clostridium tetani*

The genus contains bacteria responsible for causing the following major diseases of human beings: gas gangrene (*C. perfringens*), food poisoning (*C. botulinum*), tetanus (*C. tetani*) antibiotic associated diarrhoea (AAD) or pseudomembranous colitis (PMC) (*C. difficile*).

Clostridia grow on enriched media in the presence of reducing agent, or in an O_2 free gaseous atmosphere. Clostridia grow well on blood agar medium under anaerobic conditions. Liquid media like Robertson's cooked meat broth is also very useful for growing clostridia.

Clostridium Perfringens

Clostridium perfringens, also known as *C. welchii,* is a causative agent of gas gangrene and food poisoning. *C. perfringens* is a non-motile and capsulated bacillus. It is responsible for the formation of at least 12 distinct soluble substances or toxins. The four major toxins: alpha, beta, epsilon and iota are predominantly responsible for its pathogenicity. It produces lecithinase (phospholipase C), which cause severe tissue necrosis (gas gangrene), lymphocytosis and haemolysis.

It is responsible for producing soft tissue infections (e.g. cellulitis, suppurative myositis and myonecrosis), food poisoning, septicaemia, etc. It gives positive Nagler reaction (an opalescence in serum or egg-yolk media due to the production of phospholipase C) and is specifically neutralised by the antitoxin. Reverse CAMP test is also useful for identification of this bacteria. Systemic infections require surgical debridement and intensive antimicrobial therapy. Penicillin, metronidazole and an aminoglycoside may be given in combination. Alternatively, clindamycin plus an aminoglycoside or a broad-spectrum antibiotic such as meropenem or imipenem, may be considered. Hyperbaric oxygen therapy may also prove useful. Antiserum against the toxin is no longer used nowadays.

Clostridium Tetani

Clostridium tetani is the causative agent of tetanus, which is associated with tetanic contractions due to neuromuscular blockade by the tetanus toxin. Tetanic contractions and respiratory arrest are observed, but the level of consciousness typically remains unimpaired. *C. tetani* is motile, non-capsulated but non-invasive. Germination of tetanus spores occurs in damaged tissue where damage to the blood supply has reduced the supply of oxygen. It produces two distinct toxins: an oxygen-labile haemolysin (tetanolysin) and a neurotoxin (tetanospasmin). Tetanospasmin blocks release of neurotransmitters (i.e. gamma-aminobutyric acid, glycine) for inhibitory synapses, thus causing unregulated excitatory synaptic activity (spastic paralysis). Tetanolysin, on the other hand, may cause red blood cell lysis. Treatment comprises of tissue debridement (removal of the devitalised tissues), antibiotic therapy (long-acting penicillin injection),

passive immunisation with antitoxin globulin, and active immunisation with tetanus toxoid. Antitoxin should be promptly administered in all cases of suspected tetanus. It is ineffective when the toxin is already fixed in the central nervous system. Since the spores of *C. tetani* are so widely distributed, the only effective way to control tetanus is by prophylactic immunisation with tetanus toxoid. Prevention comprises of vaccination, consisting of three doses of tetanus toxoid followed by booster doses every 10 years.

Clostridium Botulinum

Botulism is a rare but serious illness caused by the organism known as *C. botulinum*. This organism is non-capsulated, motile with peritrichous flagella and produces spores which are oval, subterminal and bulging. It produces three types of illnes: (1) food-borne botulism, (2) wound botulism and (3) infant botulism. *C. botulinum* forms a powerful exotoxin which is responsible for the disease botulism. It can produce one of eight distinct botulinum toxins (A–G). Botulism toxin leads to a lethal form of food poisoning, "botulinism". It affects the cholinergic system, blocking the release of acetylcholine at the pre-synaptic level. This usually results in a symmetric descending paralysis, ending in death due to respiratory paralysis.

The botulinum toxin differs from a classic exotoxin in that it is not released during the life of the organism but appears in the medium only after death and autolysis of the organism. Botulinum toxin is one of the most potent toxins known.

Clostridium Difficile

Clostridium difficile is a proven cause of AAD, and PMC—a life-threatening condition. Community-acquired cases of AAD and PMC are sporadic. *C. difficile* is infectious and patients on antibiotics are at an increased risk of developing this infection.

The disease develops in people taking antibiotics because these agents alter the normal enteric flora, either permitting the overgrowth of these relatively resistant organisms or making the patient more susceptible to the exogenously acquired *C. difficile*. The disease occurs if the organisms proliferate in the colon and produce their toxins there.

The severity of disease varies widely from mild diarrhoea through varying degrees of inflammation of the large intestine to a fulminant PMC. The three broad-spectrum antibiotics, which are most commonly implicated in its causation include, clindamycin, ampicillin and the cephalosporins (third generation cephalosporins). Diagnosis is reached by detection of *C. difficile* toxin. Treatment is with oral metronidazole or vancomycin. Parenteral administration of the drugs may fail to eradicate the infection.

Chlamydia

Chlamydiae are non-motile, spherical, weakly Gram-negative, obligate intracellular bacteria. The most important specie in this genus is *C. trachomatis. Chlamydia trachomatis* has been divided into three biovars (biological variants) which cause trachoma, inclusion conjunctivitis (the so-called TRIC agents), and lymphogranuloma venereum (LGV) and genital infections respectively.

There are two morphologically distinct forms of chlamydiae: elementary body and reticulate body. The chlamydia growth cycle is initiated by the attachment of an infectious elementary body to the surface of a susceptible epithelial cell, followed by its endocytosis.

Infections of the genital tract caused by *C. trachomatis* are of two types and are sexually transmitted: first one caused by the oculogenital serotypes D through K collectively referred to as "genital chlamydiasis". The second type is LGV caused by serotypes L1, L2 and L3. LGV usually presents with swollen inguinal lymph nodes, which may have been preceded by one or more painless genital papules or ulcers.

In men, *C. trachomatis* causes nongonococcal urethritis (NGU), epididymitis, proctitis, conjunctivitis and Reiter's syndrome. Reiter's syndrome is a triad of recurrent conjunctivitis, polyarthritis and urethritis or cervicitis, associated with many infections but most commonly with *C. trachomatis.* NGU causes a discharge and/or burning on urination and is more often symptomatic in men than women. The discharge is often described as nonpurulent but ranges from barely apparent to watery or mucoid to mucopurulent. It is generally agreed that the discharge in a Chlamydia infection without a concomitant gonorrhoeal infection is less purulent than the discharge in gonorrhoea.

Most genital tract infections in women are asymptomatic (as many as 80%) but can nevertheless become symptomatic. In women, the site of infection is most often the cervix but from there it can ascend to the uterus, fallopian tubes and can also result in pelvic inflammatory disease. The clinical manifestations include acute urethral syndrome, bartholinitis, mucopurulent cervicitis, endometritis, salpingitis, pelvic inflammatory disease (PID), conjunctivitis, perihepatitis (Fitz-Hugh-Curtis syndrome) and Reiter's syndrome. Genital chlamydiasis may cause infertility, ectopic pregnancy, premature deliveries, perinatal morbidity and postpartum fever.

Treatment is with tetracycline, which should be given for at least 3 weeks. Laboratory diagnosis of chlamydial infections depends on direct detection of antigens, isolation of organism and serology for antibody detection. Frei's test, a skin test previously employed for diagnosis of LGV is now not in use due to the frequent occurrence of false positive reactions.

Mycoplasma

Mycoplasmas are the smallest prokaryotes capable of self-replication. It is Gram-negative, but is better stained by Giemsa stain. It is a strict aerobe composed of a trilaminar cell membrane comprising of sterols. Absence of cell wall and a cell membrane containing sterols makes it unique amongst bacteria. Due to lack of rigid cell wall, they are extremely pleomorphic. They were previously named as pleuropneumonia like organisms or PPLO. It can be grown on a cell free medium which comprises of a complex mixture of heart infusion, peptone, yeast extract, salts, glucose or arginine and horse serum (5–20%). This medium can be made solid by the addition of agar. On agar, colonies are typically biphasic that have a "fried egg" appearance. Mycoplasma infection is associated with the development of agglutinins to a non-haemolytic *Streptococcus* plus cold agglutinins to group "O" red cells.

They generally produce surface infections by adhering to the mucosa of the respiratory, gastrointestinal and genitourinary tracts. Mycoplasma can cause two types of diseases in humans: pneumonia and genital infections.

Infection with *M. pneumoniae* typically produces mild upper respiratory tract disease. More severe disease with lower respiratory tract symptoms occurs in less than 10% of patients. Tracheobronchitis can sometimes occur. Pneumonia (referred to as primary atypical pneumonia or walking pneumonia) can also develop. School-age children and young adults are especially susceptible to infection. Clinical disease is uncommon in very young children and older adults.

Mycoplasma hominis causes nongonococcal urethritis. It has also been found to be associated with postpartum sepsis, proctitis, acute salpingitis, pelvic inflammatory disease, cervicitis and vaginitis. It is transmitted by sexual contact.

Enterobacteriaceae

The family Enterobacteriaceae is the largest, most diverse collection of medically important Gram-negative enteric bacilli. They are aerobes or/and facultative anaerobes and grow readily on ordinary laboratory media, ferment glucose with the production of acid or acid and gas, and are either non-motile or motile with peritrichous flagella. They are catalase positive (except for *Shigella dysenteriae* type 1 which is catalase-negative), oxidase-negative, reduce nitrate to nitrites and are typically intestinal parasites of humans and animals.

The genus *Escherichia, Klebsiella* and *Enterobacter* are lactose fermenting, while the genus *Proteus, Shigella* and *Salmonella* are non-lactose fermenting. All lactose-fermenting enterobacteria are popularly known as "coliform bacilli". These organisms can be typically involved in infections such as appendicitis, diverticulitis, ischiorectal abscess, cholecystitis, peritonitis, postabortal sepsis and pelvic inflammatory disease.

Escherichia coli, an important species of the genus *Escherichia*, is a Gram-negative anaerobe producing both endotoxins and enterotoxins (enterotoxigenic *E. coli*).

It does not typically produce a malodorous infection as it is likely that other anaerobes are responsible for this, for example, *Bacteroides,* etc. Most strains have the potential to be pathogenic. *E. coli* has been identified as the most common agent associated with the UTI and gastroenteritis.

Escherichia coli 0157/H7: *Escherichia coli* 0157/H7 characteristically causes a haemorrhagic colitis with abdominal pain but little or no fever. Presently, there is no effective vaccination available against the organism. It is not a bowel commensal. An outbreak of 500 cases in the USA was described in 1993. This outbreak was associated with the consumption of hamburgers. There were over 50 cases of haemolytic uraemic syndrome and 4 fatalities. The source of another outbreak in Wishaw, Scotland in 1996 was a butcher's shop. There were over 500 cases and 18 fatalities.

Pathogens isolated from intra-abdominal pus: The most common aerobic bacteria in intra-abdominal pus in descending order are:

- ❑ *Escherichia coli*
- ❑ *Enterococci*
- ❑ *Proteus*
- ❑ *Klebsiella* spp.

The most common anaerobic bacteria in descending order are:

- ❑ *Bacteroides*
- ❑ *Clostridia*
- ❑ *Peptostreptococci*.

Spirochaetes

Spirochaetes are slender unicellular, motile, helical or spiral rods. Their motility is due to the presence of endoflagella. The members of genera *Treponema, Borrelia* and *Leptospira* are pathogenic to man. Various diseases which can be caused by spirochetes are summarised in the **Table 6.4**.

Treponema Pallidum

Treponema pallidum is a thin, coiled spirochete. It cannot be seen with Gram or Giemsa stains, but can be observed by darkfield or phase contrast microscopy. It is sensitive to water and drying. It crosses the placenta after 16 weeks' gestation. Direct fluorescent antibody test (DFA-TP) is a sensitive method for direct detection of treponemal antigens in the exudates for diagnosis of syphilis. Various diseases caused by subspecies of *T. pallidum* are listed in **Table 6.4**. Syphilis is a sexually transmitted disease found worldwide, which only affects humans. Syphilis is a disease of blood vessels and of the perivascular areas. The incubation period is 9–90 days. The natural course of syphilis can be divided into primary, secondary and tertiary stages based on the clinical manifestations.

The primary lesion in syphilis is the chancre. The chancre is a painless, relatively avascular, circumscribed, indurated, superficially ulcerated lesion. It is covered by a thick, glairy exudate very rich in bacteria. This is known as "hard chancre" to distinguish it from the non-indurated lesions of "soft sore" caused by *H. ducreyi*, and Hunterian chancre. The chancre of syphilis most frequently occurs on the external genitalia, but it may also occur on the cervix, perianal area, in the mouth or anal canal. The regional lymph nodes become swollen, discrete, rubbery and non-tender.

Secondary syphilis sets in 1–3 months after healing of the primary lesion. In this stage, patients typically experience a "flu-like" syndrome, lymphadenopathy, and a generalised mucocutaneous rash. Characteristic lesions of secondary syphilis are roseolar or papular skin rashes, mucous patches in the oropharynx and condylomata at the mucocutaneous junctions.

Two major types of serologic tests for syphilis exist: non-treponemal tests and treponemal tests. In non-treponemal tests or standard tests for syphilis (STS), cardiolipin or lipoidal antigen is used. Non-treponemal tests are

TABLE 6.4	Diseases caused by spirochetes		
Genus	**Species**	**Diseases**	**Transmission**
Treponema	*T. pallidum*	Syphilis	Sexual contact or congenital
	T. pallidum pertenue	Yaws	Traumatised skin comes in contact with an infected lesion
	T. pallidum carateum	Pinta	Traumatised skin comes in contact with an infected lesion
	T. pallidum endemicum	Endemic syphilis (Bejel)	Mouth to mouth through utensils
	T. pallidum pallidum	Venereal syphilis	Sexual contact
Borrelia	*B. recurrentis*	Epidemic relapsing fever Endemic relapsing fever	Body louse Soft-shelled tick
	B. vincentii	Vincent's angina (ulcerative gingivostomatitis or oropharyngitis)	*T vincentii* is a normal commensal of mouth, which can result in an opportunistic infection
	B. burgdorferi	Lyme disease	Tick bites (Ixodid ticks)
Leptospira	*L. interrogans* *L. biflexa*	Leptospirosis Do not cause any disease	Rats Saprophytic

flocculation tests and include venereal diseases research laboratory (VDRL) test and rapid plasma reagin (RPR).

On the other hand, treponemal tests are those in which treponemes are used as the antigen. These tests may use live *T. pallidum* strains (e.g. *T. pallidum* immobilisation test), killed *T. pallidum* (e.g. *T. pallidum* agglutination test, *T. pallidum* immune adherence test, and fluorescent treponema antibody test), or *T. pallidum* extracts as antigens (e.g. *T. pallidum* hemagglutination test and enzyme immunoassay). Penicillin is the drug of choice for treating infections with *T. pallidum*. So far there have been no reports of penicillin-resistance.

Borrelia

The important pathogenic borreliae of medical importance include *B. recurrentis, B. vincentii* and *B. burgdorferi. B. recurrentis* is the causative agent of relapsing fever while *B. vincentii* causes vincent's angina. Lyme disease is caused by *B. burgdorferi.* Lyme disease may be a progressive illness, and is divided into three stages:

Stage 1: In the first stage "localised infection" appears as a small red macule or papule at the site of bite (erythema migrans) following an incubation period of 3–30 days.

Stage 2: The second stage of "disseminated infection" develops with headache, fever, myalgia and lymphadenopathy. Some develop meningeal or cardiac involvement. This stage usually develops after weeks or months of the first stage.

Stage 3: This is the stage of "persistent infection" which sets in months or years later with chronic arthritis, polyneuropathy, encephalopathy and acrodermatitis.

Penicillins, the newer macrolides, cephalosporins and tetracyclines have all been used successfully in Lyme disease.

Leptospira

Leptospira stain poorly with aniline dyes (e.g. Gram stain) but stain well with silver impregnation methods (e.g. Levaditi's and Fontana's staining). They are obligate aerobes. It causes leptospirosis, which is the most common zoonotic bacterial disease throughout the world. Rodents are most important reservoirs and the disease is acquired through exposure to water contaminated by the urine of infected animals, typically rats. It causes mild virus-like syndrome (including symptoms such as fever, myalgia, headache, pneumonia and jaundice), systemic leptospirosis with aseptic meningitis, overwhelming disease (Weil's disease) with vascular collapse, thrombocytopenia, haemorrhage, and hepatic and renal dysfunction. It is treated with penicillins/doxycycline.

Actinomyces

Actinomycetes are traditionally considered to be transitional forms between bacteria and fungi. *Actinomyces* is an anaerobic or microaerophilic and non-acid-fast, Gram-positive, cast-forming, non-acid-fast, non-motile, non-spore-forming bacillus that is difficult to isolate and identify. Its filamentous growth and mycelia-like colonies have a striking resemblance to fungi. The mycelial forms break up into coccoid and bacillary forms. Most bacteria show true branching. The mycelial masses may be visible to the naked eye and are called sulphur granules, as they are often light yellow in colour. It is a soil organism, often found in decaying organic matter (for example: wet hay and straw).

The Actinomyces cause the disease known as actinomycosis, which is a chronic disease characterised by multiple abscesses and granulomata, tissue destruction, extensive fibrosis and the formation of sinuses. In humans, the disease occurs in five clinical forms: cervicofacial, thoracic, abdominal, pelvic and punch actinomycosis.

Actinomyces israelii, which causes human actinomycosis, is primarily a commensal microbe found in normal oral cavities, tonsillar crypts, dental plaques and in carious teeth. It is, however, associated with wound infections, abdominal and pelvic abscess formation and keratitis or canaliculitis. Surgical debridement supported by penicillin/cephalosporin is the treatment of choice.

Pseudomonas

It is a Gram-negative aerobic, motile bacillus. It grows well on ordinary media in the laboratory. Cetrimide agar is the selective medium for *P. aeruginosa. P. aeruginosa* produces two soluble pigments: pyocyanin (blue pigment) and pyoverdin (fluorescein).

Pseudomonas aeruginosa, the most important species, is an opportunistic pathogen which is responsible for causing the following community acquired infections: Otitis externa and varicose ulcers; corneal infections; industrial eye injuries, contact lens-acquired infection, etc. It can also cause hospital-acquired infections: localised lesions (infections of wounds and bedsores, eye infections and urinary infections following catheterisation), infection in burns, iatrogenic meningitis, post-tracheostomy pulmonary infection; septicaemia and endocarditis; skin lesions; infection of the nail bed; infantile diarrhoea and sepsis; osteomyelitis; infections of the gastrointestinal tract, CNS, and musculoskeletal system. It particularly causes infection in patients with severe burns, patients with cancer and patients who are immunosuppressed.

Pseudomonas aeruginosa is frequently resistant to many commonly used antibiotics. Although many strains are susceptible to gentamicin, tobramycin, colistin, and amikacin, resistant forms have developed. The combination

of gentamicin and carbenicillin is frequently used to treat severe Pseudomonas infections. *P. aeruginosa* is sensitive to quinolones, which inhibit DNA topoisomerase.

Campylobacter

Campylobacters are thin, curved Gram-negative bacilli. They are microaerobic and strongly oxidase positive. *Campylobacter jejuni* and *Campylobacter coli* have emerged as common human pathogens belonging to this genus. *C. jejuni* is associated with gastroenteritis, septicaemia, meningitis, spontaneous abortion, proctitis, Guillain-Barré syndrome. Attack rates are highest in young adults and children. It is transmitted to humans by milk or water infected by wild and domestic animals and poultry. Stool culture requires special conditions: temperature of 42°C, microaerobic atmosphere on blood agar with antimicrobials added. Infections are treated with ciprofloxacin and erythromycin, but most are self-limiting.

Principles of Infection Control and Outbreak Management

Infection and immunity involve interaction between the animal body (host) and the infecting microorganisms.

Infection

Sources of Infection

The most common sources of infection include the following: human beings (from a patient or carrier), animals (zoonotic diseases), insects, soil, water and food.

Modes of Transmission of Infection

The infection can be transmitted through one of the following routes: contact, inhalation, ingestion, inoculation, vertical transmission from the mother, congenital, iatrogenic and laboratory infections.

Zoonosis

The diseases and infections, which are transmissible to man from animals, are known as zoonotic disorders. Examples of some common zoonotic diseases are listed in **Table 6.5.**

Disease Causation

Hill devised criteria for assessing disease causation and proposed that the cause must precede the effect. Hill's criteria suggest that when assessing causation removing the factor of interest should reduce the risk of disease.

TABLE 6.5	Different types of zoonotic disorders
Type of infection	**Examples of the transmitted diseases**
Bacterial	• Anthrax (cattle and goats) • Brucellosis (sheep) • Q fever (cattle, sheep) • Leptospirosis (rats) • Bovine tuberculosis (cattle) • Bubonic plague • Salmonella food poisoning • Listeriosis (pets) • Toxocara (cats)
Viral	• Rabies • Yellow fever • Cowpox • Monkeypox
Protozoan	• Leishmaniasis • Toxoplasmosis (cats) • Trypanosomiasis • Babesiosis
Helminthic	• Echinococcosis • Taeniasis • Trichinellosis
Fungal	• *Microsporum canis* • *Trichophyton verrucosum*

Hill suggested that there should be a dose-response relationship, that is, higher levels of the causative factor should lead to more severe disease or more rapid disease onset. According to the Hill's criteria, the causative factor can be present in people both with and without the disease. Example, rheumatoid factor is found in people both with and without rheumatoid arthritis (RA). Also the causative agent may even be found in unaffected individuals. Again, autoantibody tests illustrate this principle as the rheumatoid factor can be found in unaffected patients.

Determinants of Virulence

Pathogenicity denotes the ability of a microbial species to cause a disease. The term virulence, on the other hand, denotes the ability of a strain of a species to produce disease. The various characters of a pathogen, which can determine its virulence, include factors such as transmissibility, adhesion, invasiveness, toxigenicity, enzymes, plasmids, bacteriophages, communicability, infecting dose, and the route of infection.

Toxins, both exotoxins and endotoxins (such as the cell wall of *Haemophilus influenzae*) act as important determinants of virulence. M protein on some bacteria helps to prevent phagocytosis. Pili on gonococcus allow them to adhere to mucosal surfaces. Beta-lactamase is an enzyme produced by some bacteria which hydrolyses penicillin but has no direct effect on host tissue. Therefore, they cannot be considered as the determinant of virulence. Presence of proteolytic enzymes also help in increasing virulence of an organism. These enzymes may be involved either in

direct or indirect destruction of an infected/colonised tissue and in dysregulation of many host defence pathways (e.g. effect of bacterial proteinases on fibrinolytic, kallikrein-kinin and complement cascades, as well as degradation of immunoglobulins, inactivation of endogenous proteinase inhibitors, and dysregulation of cytokine network system, etc.). Proteolytic enzymes are responsible for the virulence and activity of organisms such as *S. pyogenes*, *S. aureus*, *Escherichia coli* and *C. welchii* enabling the necrolytic effects on the skin, resulting in the development of cellulitis and gangrene.

Toxins

Toxigenesis, or the ability to produce toxins, is an underlying mechanism by which many bacterial pathogens produce disease. At a chemical level, there are two types of bacterial toxins: exotoxins and endotoxins. Difference between the two is listed in **Table 6.6**. Both Gram-negative and Gram-positive bacteria produce toxins. While the exotoxins are produced by both Gram-positive and Gram-negative bacteria, endotoxins are the constituents of the cell wall of Gram-negative bacteria. Bacterial protein toxins are the most powerful human poisons known and retain high activity at very high dilutions. Their effect may be neutralised by the antitoxin and this is used as a treatment strategy (toxoid).

Febrile Response: A Mediator of Infection

Fever is the most common response to infection. This is mediated via the endogenous as well as the exogenous pyrogens. Exogenous pyrogens include sources such as infective agent, immunological reactions or toxins.

Endogenous pyrogens consist of cytokines such as IL1 and 6, tumour necrosis factors-β and interferon-α. These are derived from monocytes, endothelial cells, B cells, glial cells and epithelial cells. Exogenous pyrogens cause a release of endogenous pyrogens, which act on the hypothalamus, thereby releasing prostaglandin E2. This acts on the temperature regulatory centre to reset it, resulting in heat conservation by the body and increased heat production.

Sterilisation and Disinfection

Sterilization is the process by which an article, surface, or medium is freed of all living microorganisms either in the vegetative or spore state.

Disinfection

This is the killing, inhibition, or removal of microorganisms that may cause disease.

Antisepsis

This is the prevention of sepsis or putrefaction either by killing microorganisms or by preventing their growth.

Performance of a Disinfectant

Performance of a disinfectant is influenced by the following factors:
- *pH of the medium*: pH plays an important role in the performance of a disinfectant. Many organisms have an optimum pH at which they work best. Deviation from this optimum value results in loss of activity.

TABLE 6.6	Bacterial toxins	
Feature	Exotoxin	Endotoxin
Bacteria responsible for production	Gram-negative and Gram-positive	Gram-negative
Release	Exotoxins are extracellular soluble proteins produced by the living bacteria. They are released by the bacteria into its surroundings	Endotoxins are lipopolysaccharides of the cell walls of Gram-negative bacteria and are released following cell death.
Examples	Tetanus toxins Cholera toxins	Lipopolysaccharides, which are seen in Gram-negative pathogenic bacteria, e.g. *Escherichia coli, Salmonella, Shigella, Pseudomonas*, etc.
Antigenicity	Susceptible to antibodies Destroyed by heating	Limited effect of antibodies
Amounts required	Lesser amounts are required in comparison to endotoxins	Larger amounts are required to produce effect in comparison to exotoxins
Conversion to toxoids	Can be converted to toxoids	Cannot be converted to toxoids
Effect of heating	Not heat stable	Heat stable (boiling for 30 minutes does not destabilise them)
Effects	—	Responsible for producing numerous effects of the infection (e.g. fever, disseminated intravascular coagulation, myolysis, etc.) by causing influx of the inflammatory compounds (e.g. tumour necrosis factors-α)

❏ *Temperature*: Warm liquid disinfectants are more effective than colder ones, because increasing the temperature of the liquid helps in decreasing the surface tension whilst also increasing the rate of chemical reactions at the same time.

❏ *Numbers of organisms*: Higher number of organisms lead to clump formation. Organisms inside the clump are shielded from the action of the disinfectant.

❏ *Concentration of disinfectant*: The concentration of disinfectant as well as the type of organisms is also important.

Methods of Sterilisation and Disinfection

Physical Agents

Dry heat: (A) Red heat, (B) Flaming, (C) Incineration, (D) Hot-air sterilisation, (E) Microwave ovens

Hot air oven is a method of choice for sterilisation of the following:

❏ *Glassware*: Such as tubes, flasks, measuring cylinders, all-glass syringes, glass petri dishes and glass pipettes.

❏ *Metal instruments*: Such as forceps, scissors and scalpels.

❏ *Non-aqueous materials*: This includes powders, oils and greases in sealed containers and swab sticks packed in test tubes.

Hot-air ovens have the benefit of not causing corrosion of non-stainless metals and not damaging fine cutting edges of delicate instruments.

Moist heat: (A) Pasteurization: For pasteurization of milk, the temperature employed is either 63°C for 30 minutes (holder method) or 72°C for 15–20 seconds (the flash method) followed by rapid cooling to 13°C or lower. This is able to kill all pathogenic bacteria and most non-pathogenic bacteria; (B) Boiling; (C) Steam under normal pressure.

Tyndallization: An exposure of steam at 100°C for 20 minutes on three successive days is called tyndallization or intermittent sterilisation.

Steam under pressure: This includes autoclaving, which is a very effective method of sterilisation. This is a process of sterilisation by saturated steam under high pressure, at about 15 psi of pressure (121°C). Flash autoclaving is no longer recommended or available for safety reasons; the preferred setting is 132°C (30 lb/in^2) held for 3 minutes.

Filtration

Various types of filters are now available: (1) Earthware filters, (2) Asbestos filters, (3) Sintered glass filters, (4) Membrane filters, (5) Syringe filters, (6) Vacuum and "in-line" filters, (7) Pressure filtration and (8) Air filters.

Radiation

Both ionising and non-ionising radiation can be used.

Chemical Agents

Although generally less reliable than heat, these chemicals are suitable for treating large surfaces and many heat-sensitive items. These include the following:

❏ *Surface-active agents*: These include (A) Cationic agents, (B) Anionic agents and (C) Ampholytic (amphoteric) agents.

❏ *Phenol derivatives*: Certain phenol derivatives like cresol, chlorhexidine, chloroxylenol and hexachlorophane are commonly used as antiseptics.

❏ *Alcohols*: Ethyl alcohol (ethanol) and isopropyl alcohol are the most frequently used.

❏ *Heavy metals*: Mercuric chloride and silver nitrate

❏ *Oxidising agents*
  • *Halogens*: Chlorine and iodine are the two most commonly used halogens for disinfection.
  • Hydrogen peroxide.
  • *Dyes*: Aniline and acridine are two groups of dyes which are used extensively as skin and wound antiseptic agents.
  • *Alkylating agents*: The lethal effects of aldehydes (formaldehyde and glutaraldehyde) and ethylene dioxide result from their alkylating action on proteins. Under optimal conditions of concentration, temperature and exposure time, ethylene oxide has a broad-spectrum cidal action. Ethylene oxide is toxic, irritant, mutagenic and carcinogenic; therefore its use requires extreme caution.

Specific Infections

Hospital-acquired Infections

Nosocomial or hospital-acquired infections are the infections which are acquired by an individual in the hospitals and other healthcare facilities. To be identified as a nosocomial infection, the patient must have been admitted in the hospital for reasons other than the infection. No signs of active or incubating infection must be present. Hospital-acquired infections may be caused by viral, bacterial and fungal pathogens. According to WHO (2002), the most common types of hospital-acquired infections are bloodstream infections, pneumonia, e.g. ventilator-associated pneumonia, UTI and surgical site infections. The sources of hospital-acquired infection may be exogenous or endogenous. Almost any microbe can cause a hospital-acquired infection. However, the organisms which are able to survive in the hospital environment for long periods and develop resistance to the commonly used antibiotics and disinfectants are particularly important in this respect.

Prevention

The following steps can be taken for the prevention of hospital-acquired infections:

❏ *Adequate sterilisation and disinfection*: There should be an adequate provision of sterile instruments, dressings, surgical gloves, facemasks, theatre clothing and fluids in the hospital.

❏ *Skin disinfection and antiseptics*: Person to person contact is a very common cause of infection spread in hospitals. Therefore, scrupulous hygiene when moving between patients in an intensive therapy unit is essential. Thorough hand washing after any procedure which involves nursing care or close contact with the patient is essential.

❏ *Preoperative surgical asepsis*: Procedures for preoperative disinfection of the patient's skin and for surgical scrubs are compulsory within the operating theatre.

❏ Rational antibiotic prophylaxis.

❏ *Isolation*: Isolation of the susceptible patients is essential for preventing the spread of specific infections to other patients and for the protection of susceptible or immunocompromised patients.

❏ *Monitoring*: Aerosols caused by air-conditioning units are notorious for spreading Gram-negative bacteria, for example, coliforms and *Legionella*. Therefore, monitoring of the physical performance of air-conditioning plants is essential.

❏ *Keeping the hospital equipment dry*: If an item of equipment is contaminated with bacteria, the number of bacteria will usually remain constant, or decline, if the item is dry. If the item is wet, some bacteria, e.g. *Pseudomonas*, may multiply.

❏ *Use of theatre air systems*: Theatre air systems generate a positive pressure compared to the surroundings. The positive pressure air is moved away from the patient and filtered, so that airborne infections are prevented from reaching the patient. The laminar flow used in orthopaedic theatre is the logical progression of this concept.

Food Poisoning

The term bacterial food poisoning is restricted to acute gastroenteritis due to the presence of bacteria (usually in large numbers) in the various food products. It is of three types **(Table 6.7)**.

1. *Infective type*: In this type, multiplication of bacteria occurs in vivo when infective doses of microorganisms are ingested with food. Incubation period is generally 8–24 hours. The typical example of this type of food poisoning is by *Salmonellae*.

2. *Toxic type*: In this type, the disease follows ingestion of food with preformed toxin. Incubation period is short (2–6 hours). Examples are staphylococcal food poisoning, poisoning by *Bacillus cereus* and *C. botulinum. S*taphylococcal food poisoning is caused by *S. aureus*, where this bacterium produces an enterotoxin. *Bacillus cereus* infection is usually caused by precooked rice. *B. cereus* produces two endotoxins: The emetic type (having a short incubation period of about 2 hours) and the diarrhoeal type (having an incubation period of 6–14 hours). Infection with *C. botulinum* is usually associated with canned food. It causes food poisoning by producing a potent exotoxin.

3. *Infective-toxic type*: In this type, bacteria release the toxin in the bowel. The incubation period is 6–12 hours. The typical example is *C. perfringens* food poisoning, where *C. welchii/perfringens* produces an enterotoxin. Food poisoning occurs in microepidemics after ingestion of infected meat or poultry.

Notifiable Diseases

Diseases notifiable to local authority officers in the UK as per the regulations of Health Protection Legislation (England) guidelines (2010) are as follows:

❏ Acute encephalitis
❏ Acute infectious hepatitis
❏ Acute meningitis
❏ Acute poliomyelitis
❏ Anthrax
❏ Botulism
❏ Brucellosis
❏ Cholera
❏ Diphtheria
❏ Enteric fever (typhoid or paratyphoid fever)
❏ Food poisoning
❏ Haemolytic uraemic syndrome
❏ Infectious bloody diarrhoea
❏ Invasive group A streptococcal disease
❏ Legionnaires' disease
❏ Leprosy
❏ Malaria
❏ Measles
❏ Meningococcal septicaemia
❏ Mumps
❏ Plague
❏ Rabies
❏ Rubella
❏ Severe acute respiratory syndrome
❏ Scarlet fever
❏ Smallpox
❏ Tetanus
❏ Tuberculosis
❏ Typhus

TABLE 6.7	Various types of food poisoning
Types of food poisoning	**Causative agent**
Infective type	• *Salmonella typhimurium* • *Campylobacter jejuni*
Toxic type	• *Staphylococcus aureus* • *Bacillus cereus* • *Clostridium botulinum*
Infective—toxic type	• *Clostridium perfringens*

- ❑ Viral haemorrhagic fever (VHF)
- ❑ Whooping cough
- ❑ Yellow fever.

Viruses

Viruses are the smallest known infective agents and are perhaps the simplest form of life known. They are obligate intracellular parasites containing only one type of nucleic acid (DNA or RNA) as their genome. They are not capable of growing in inanimate media and are resistant to antibiotics. Various types of viruses are classified in **Table 6.8**. Viruses of importance which can be associated with intrauterine infection are described in **Table 6.9**.

Herpes Group of Viruses

The Herpesviridae family of viruses is double-stranded (ds) DNA viruses that include herpes simplex, varicella zoster, cytomegalovirus (CMV) and Epstein-Barr virus (EBV). Herpes viruses are large, icosadeltahedral viruses surrounded by a lipid envelope containing peplomers. A unique property of herpesviruses is their ability to establish latent infections, lifelong persistent infections in their hosts, which can be periodically reactivated. The human herpes viruses have been classified in **Table 6.10**. Human herpesviruses include human herpesvirus 1 (HV-1) to human herpesvirus 8 (HV-8). Herpes virus usually causes lifelong infection.

Human Herpesvirus 1

HSV-1 causes superficial lesions of the face, mouth, pharynx and cornea including infections such as acute herpetic gingivostomatitis, acute herpetic pharyngotonsillitis, herpes labialis, herpes encephalitis, eczema herpeticum, and herpetic whitlow. The virus can remain latent in the trigeminal ganglion and dorsal root ganglion and may reactivate as cold sores.

Human Herpesvirus 2

HSV 2 is sexually transmitted and can cause infections such as genital herpes, neonatal infection, and aseptic meningitis. If encephalitis develops, mortality can be as high as 90%. Varicella zoster virus (VZV) causes chickenpox (varicella) and herpes zoster or shingles, two distinct clinical entities in humans.

Genital Herpes Infection

Genital herpes is one of the most common sexually transmitted diseases worldwide and is caused by HSV type 1 or 2.

TABLE 6.9	**Viruses which may cause intrauterine infections**	
Virus infection	**Birth defect**	**Foetal death**
Rubella	Yes	Yes
Cytomegalovirus	Yes	Yes
Varicella	Yes	Yes
Parvovirus B19	No	Yes
HIV1, HIV2	No	Yes
Hepatitis C	No	Unknown
Hepatitis E	No	Yes
Poliomyelitis	No	Yes
Coxsackie B virus	No	Yes
Japanese B encephalitis	Unknown	Yes
Lassa fever	No	Yes

TABLE 6.8	**Classification of viruses**		
RNA-containing viruses		**DNA-containing viruses**	
Name of virus	**Species**	**Name of virus**	**Species**
Picornavirus	• Enterovirus (Coxsackie, ECHO and polio) (ss) • Rhinoviruses (common cold) • Hepatitis A	Herpes viruses	• Herpes simplex I and II (ds), varicella zoster (ds) • Cytomegalovirus, (ds) • Infectious mononucleosis caused by the Epstein-Barr virus (ds) • HHV-6
Reoviruses (including rotaviruses): rotavirus (ds)	• Viruses causing respiratory tract infections and diarrhoea in children	Pox viruses	Small pox, vaccinia and molluscum contagiosum
Myxoviruses	• Influenza (ss)	Adenoviruses	Respiratory tract infections and keratoconjunctivitis
Paramyxoviruses	• Mumps, parainfluenza, measles and respiratory syncytial virus	Papovaviruses	Verrucae and human papilloma virus (ds)
Arboviruses	• Encephalitis, yellow fever, dengue and sandfly fever	Hepadnaviruses	Hepatitis B
Rhabdoviruses	• Rabies virus (ss)	Parvovirus	Parvovirus B19 (ss)
Retroviruses	• HIV (ss), HTLV 1, rota virus (ds)		
Rubivirus	• Rubella (ss)		
Flavivirus	• Japanese B virus (ss) • Hepatitis C		

Abbreviations: ss, single stranded; ds, double stranded; HHV, human herpes virus

TABLE 6.10	Classification of human herpesviruses			
Subfamily	**Virus**	**Primary target cell**	**Site of latency**	**Mode of spread**
A. *Alpha herpesvirinae*				
Human herpesvirus 1	Herpes simplex type 1	Mucoepithelial cells	Neuron	Close contact
Human herpesvirus 2	Herpes simples type 2	Mucoepithelial cells	Neuron	Close contact (sexually transmitted disease)
Human herpesvirus 3	Varicella-zoster virus	Mucoepithelial cells	Neuron	Respiratory and close contact
B. *Beta herpesvirinae*				
Human herpesvirus 5	Cytomegalovirus	Monocyte, lymphocyte and epithelial cells	Monocyte, lymphocyte and (?)	Close contact, transfusions, tissue transplant, and congenital
Human herpesvirus 6	Herpes lymphotropic virus	T cells and (?)	T cells and (?)	Respiratory and close contact (?)
Human herpesvirus 7	Human herpesvirus 7	T cells and (?)	T cells and (?)	(?)
C. *Gamma herpesvirinae*				
Human herpesvirus 4	Epstein-Barr virus	B cells and epithelial cells	B cell	Saliva (kissing disease)
Human herpesvirus 8	Kaposi's sarcoma-related virus	Lymphocyte and other cells	(?)	Close contact (sexual), saliva (?)

Transmission occurs during close contact with a person who is shedding the virus. Asymptomatic infection can rarely occur. Women, who acquire primary genital herpes during pregnancy, particularly in the third trimester, may transmit the infection to the baby at the time of delivery. However, genital herpes does not cause preterm delivery or spontaneous abortion.

Cytomegalovirus

Cytomegalovirus or HSV-5 is the causative agent of mononucleosis syndrome in immunocompetent hosts. CMV causes latent infection; hence reactivation may result in a severe form of disease in patients who are immunocompromised. CMV is transmitted orally and sexually, via blood transfusions, through tissue transplants, in utero, at birth, and by nursing. Infectious CMV may be shed in the bodily fluids of any previously infected person, and thus may be found in urine, saliva, blood, tears, semen and breast milk.

Cytomegalovirus can cause congenital CMV infection, acquired CMV infection and infections in immunocompromised and immunocompetent adult hosts. CMV generally causes subclinical infection. Presence of "Owl's-eye" inclusion body and basophilic intranuclear inclusion body on histopathology is the diagnostic feature of the cells infected by CMV. The enzyme-linked immunosorbent assay (or ELISA) is the most commonly available serologic test for measuring antibodies to CMV.

Congenital Cytomegalovirus Infection

Cytomegalovirus is transmitted across the placenta and is the most common congenital infection in the UK and developed part of the world. It may rarely cause lethal infection amongst the neonates. Nearly 5–10% of the congenitally infected babies may have symptoms apparent at birth. The cells of infected organs may show large intranuclear inclusions. CMV is a cause of non-immune hydrops due to haemolytic anaemia. Clinical features of congenital CMV infection include features such as microcephaly, sensorineural deafness, hepatosplenomegaly, chorioretinitis, evidence of intrauterine growth restriction (IUGR), jaundice, skin rashes, echogenic bowel, intracranial calcification, hearing loss, anaemia, prolonged elevation of bilirubin, and rarely inguinal hernia. Presence of intracranial calcification worsens the long-term prognosis of infected foetuses. Babies who are not symptomatic at birth may later show symptoms such as deafness (sensorineural), developmental delay, mental retardation and visual impairment. It is one of the main causes of childhood deafness. It ranks second to Down's syndrome as a cause of mental retardation.

The principal reservoir of infection is small newborn children. The biggest risk is to the baby which is "symptomatic" at birth after the intrauterine infection. Such babies can excrete the virus for months from their urinary and respiratory tracts. The infected neonate can excrete the virus for 5 years or more. As a result, nurseries are a common source of infection. If the baby is infected in-utero, and is born with the signs of infections, he/she is at a high risk of damage. However, if the baby is infected in-utero and is born healthy or with no signs of infections, he/she is at a much lower risk of damage.

Women seeking to avoid infection are advised to wash carefully after dealing with infected children. Sexual transmission of the virus can also occur, with infected men excreting the virus in semen for ages. Women should therefore avoid infected partners or ensure that they use condoms.

Maternal infection is asymptomatic in 90% of the cases and the features are non-specific. Maternal infection is not serious for the mother unless she is immunocompromised. Maternal infection usually produces a self-limiting illness with flu-like symptoms.

Primary maternal infection is immensely more dangerous than reinfection, which rarely occurs. Also, infection early in pregnancy is more likely to cause foetal damage than that occurring during the late pregnancy. Breast milk from mothers with primary infection in pregnancy usually contains the virus. However, the benefits of breastfeeding outweigh the risks to the baby. In addition, the mother is likely to transfer some protective immunoglobulin G (IgG), especially if she breast feeds.

The virus can be transmitted vertically from the mother: across the placenta, directly from the genital tract during delivery and in breast milk. Transmission through the breast milk is the most common source of infection of a neonate that was uninfected at birth.

Lab diagnosis: Foetal diagnosis is by amniocentesis. Viral DNA can be identified in amniotic fluid using polymerase chain reaction (PCR). The virus can also be sought in foetal blood obtained by cordocentesis. However, this is an invasive investigation and associated with a high risk of foetal loss. Ultrasound is used for assessing for an evidence of foetal damage. Some of the classical features indicative of CMV infection include enlargement of the ventricles, and periventricular leucomalacia and calcification. MRI has been used, but has not proved superior to ultrasound and is not recommended for routine use.

Routine maternal antenatal screening is not done. Screening for CMV infection during pregnancy is not recommended by the RCOG or the Department of Health. Assessment of the serum antibody levels help in knowing the stage of infection (recent or old) to some extent. Interpretation of IgG and IgM levels in maternal serum can, however, be difficult. IgM is the initial response to infection and is gradually replaced over a period of months by IgG. IgM is large antibody which is unable of crossing the placenta. Thus, in the early days of an infection, the baby gets no help from the maternal response as initially only IgM antibodies are formed which are unable to cross the placenta. Interpretation of anti-CMV antibodies in the serum is described in **Table 6.11**.

Epstein-Barr Virus

Epstein-Barr virus (EBV) causes diseases such as infectious mononucleosis (glandular fever) and has been causally associated with Burkitt's lymphoma, Hodgkin's disease and

TABLE 6.11	Interpretation of anti-cytomegalovirus antibodies in the serum
Antibody present	**Interpretation**
IgM only	Recent infection
IgG only	Old infection; presence of immunity
IgM + IgG	Recent infection with long-term immunity developing
Neither IgM or IgG	No immunity

nasopharyngeal carcinoma (common in Southeast Asia). EBV has also been associated with B-cell lymphomas in patients with acquired or congenital immunodeficiencies. EBV is a mitogen for B cells and immortalises B cells in tissue culture. EBV infects B lymphocytes and squamous epithelial cells of the oropharynx. The virus can transform B cells and epithelial cells to produce Burkitt's lymphoma (a subset of Hodgkin's lymphoma), nasopharyngeal carcinoma and oral hairy leucoplakia.

It is shed in pharyngeal secretions and transmission of EBV occurs via saliva, close oral contact (kissing disease), or sharing of items.

Infectious mononucleosis is associated with the production of IgM heterophile agglutinins in nearly 85–90% of patients during the acute phase of illness. These can be detected by the Paul-Bunnell test or a rapid slide agglutination test. Paul-Bunnell test aims at testing the presence of heterophile antibodies in the blood of infected patients and is based on the agglutination of sheep erythrocytes by the inactivated serum of patients with the disease. Monospot test, ELISA, Western blot, DNA probe, PCR and virus isolation are used for diagnosis nowadays.

Human Herpesvirus 6 to 8

Herpesvirus 6 and 7 cause infections in the childhood. They can cause erythematous febrile rashes also known as exanthema subitum. HV-6 infection has also been found to be associated with febrile convulsions. Human herpesvirus 8 can cause Kaposi's sarcoma.

Human Papillomavirus

Papovaviruses are double-stranded DNA viruses and can belong to two genera, *Papillomavirus* and *Polyomavirus*. Papillomaviruses are species-specific DNA viruses that infect the squamous epithelia and mucous membranes of vertebrates, including man and can cause several different kinds of warts in humans, such as cutaneous warts, genital warts, respiratory papillomatosis, oral papillomas and cancer.

Human papillomavirus (HPV) is the most common cause of sexually transmitted diseases world-wide. More than 100 types of HPV viruses are present. Types 6 and 11 are "low-risk" which has been found to be associated with benign genital warts and low grades of cervical intraepithelial neoplasia (CIN). Types 16, 18, 30, 33 and some others are "high-risk" as they cause CIN lesions of all grades and cervical cancer, both squamous carcinoma (more common) and adenocarcinoma. In the CIN lesions, the virus is free in the cells; in malignant cells, the viral DNA is integrated into various chromosomes. HPV has been detected in nearly 90% cases of cervical cancer. HPV transmission is by direct contact, usually sexual. Babies occasionally get laryngeal warts from transmission during passage through the birth canal. However, they do not get disseminated infection

from vertical transmission because the virus is unable to cross the placenta.

Two highly effective vaccines are available in the UK for protection against HPV. These two vaccines which have been approved for use in the UK in 2007 include Cervarix and Gardasil. Cervarix protects against HPV types 16 and 18. Gardasil not only provides protection against types 16 and 18, but also immunises against types 6 and 11. HPV types 6 and 11 are responsible for causing nearly 90% of genital wart infections, so Gardasil may be associated with an additional advantage. Both the vaccines are highly effective in reducing the risk of infection and of getting CIN. There is some evidence that those already infected with these virus types also respond to the vaccine. Questions have been raised about side-effects and safety of these vaccines, but the overall risk appears to be low. Though these vaccines do not contain a live virus, their safety in pregnancy and lactation remains unknown.

Since September 2008, all girls aged 12–13 years in the UK need to be vaccinated. A catch-up program for older girls would be started later. Boys are not to be immunised. Although type 16 HPV has been implicated in more than 50% of cases of penile cancer, the condition is rare. Therefore, presently it does not appear practical to immunise the whole male population for the prevention of penile cancer. In the near future, it is likely that the HPV screening would be incorporated into the cervical screening programme. An ongoing randomised trial since 2009, The "Artistic trial" aims at the evaluation of the role of HPV testing in primary cervical screening.

Hepatitis Viruses

At least six viruses, A through G (A, B, C, D, E and G) are hepatitis viruses. All the hepatitis viruses are RNA viruses, except the hepatitis B virus (HBV), which is a DNA virus belonging to the family Hepadnaviridae. Mode of transmission is enteric in hepatitis A and hepatitis E viruses while it is parenteral and sexual in both hepatitis B and hepatitis C viruses.

Hepatitis A

Hepatitis A virus (HAV) is an RNA virus, which spreads via faecal-oral route, having an incubation period of 15–40 days. Hepatitis A is generally a self-limiting infection and usually lasts for about 3–6 weeks. HAV can be demonstrated in the stools by immunoelectron microscopy. ELISA is the method of choice for detection of IgM and IgG antibodies in the serum. This infection is not associated with a carrier state. HAV can be prevented by use of vaccines containing formalin-inactivated HAV. Prophylaxis with hepatitis A immunoglobulin can be administered to the contacts within 2 weeks of exposure.

Hepatitis B

Hepatitis B virus is a double-walled spherical structure and measures 42 nm in diameter (Dane particle) **(Fig. 6.8)**. The outer surface or envelope of virus contains hepatitis B surface antigen (HBsAg). It encloses an inner icosahedral 27 nm nucleocapsid (core), which contains hepatitis B core antigen (HBcAg). Inside the core is the genome, a circular double-stranded DNA and a DNA polymerase. The incubation period of the virus varies between 60–160 days.

The virion envelope consists of three antigens: hepatitis B surface antigen (HBsAg), hepatitis B core antigen (HBcAg), and hepatitis B e-antigen (HBeAg).

❑ *Hepatitis B surface antigen*: Also known as the Australia antigen, hepatitis B surface antigen is present on the outer coat of the virus. It is always present during the acute phase of the infection and in the chronic carrier phase. The appearance of antibodies to HBsAg indicates recovery and subsequent immunity.

❑ *Hepatitis B core antigen*: The core antigen is present on the hepatocytes and is not detected in the serum. Presence of IgM antibodies to the core antigen indicates acute infection and presence of IgG antibodies indicates past exposure.

❑ *Hepatitis B e-antigen*: This antigen is an indicator of infection. This antigen is detected during the phase of acute infection and in the carriers having a high risk of transmission. Antibodies to HBeAg in the carriers is associated with a low risk of transmission.

Mode of Transmission

Hepatitis B virus is a blood borne virus and there are three important modes of transmission: parenteral transmission, perinatal transmission (vertical transmission from the mother to the child), and sexual transmission. It is usually transmitted through the inoculation of blood products (e.g. transfusion, sharing of contaminated needles by the drug addicts, etc.). Persons handling blood or those working in the haemodialysis units are at a special risk.
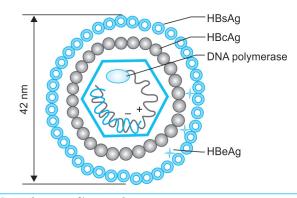
FIG. 6.8: Structure of hepatitis B virus

Laboratory Diagnosis

Clinical outcomes in case of acute hepatitis B infection are described in **Figure 6.9**. Although the majority of patients recover completely from hepatitis B, some patients may go on to develop hepatocellular carcinoma. Less than 50% of patients will progress to chronic liver disease.

Specific diagnosis of hepatitis B rests on serological demonstration of the viral markers and can be carried out by detection of HBsAg, anti-HBs, HBeAg, anti-HBe, IgM anti-HBc, IgG anti-HBc and HBV DNA in the serum. The sequence of appearance of viral markers in the blood is important. These can be detected by sensitive and specific tests like ELISA and RIA (radioimmune assay). HBV DNA is also an indicator of viral replication and infectivity. Molecular methods such as DNA hybridization and PCR are used for HBV. **Figure 6.10** shows hepatitis antigens, antibodies and DNA in a patient recovering from acute HBV infection.

Prophylaxis

Measures for the control of HBV infection include general prophylaxis and immunisation.

General prophylaxis: This consists of avoiding risky practices like promiscuous sex, injectable drug abuse and direct or indirect contact with blood, semen or other body fluids of patients and carriers.

Immunisation: Both passive and active methods of immunisation are available.

❏ *Passive immunisation*: Hyperimmune hepatitis B immune globulin (HBIG) administered IM in a dose of 300–500 IU soon after exposure to infection provides passive immunity. It may not prevent infection, but protects against illness and the carrier state. HBIG must be administered as soon as possible after an accident and preferably within 48 hours. A second dose is given 4 weeks later to those who do not respond to current vaccines. If the victim has not been previously vaccinated, a course of active immunisation is started along with the administration of HBIG. The two substances must be injected into different body sites.
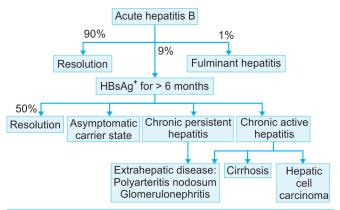
❏ *Active immunisation*: Active immunisation is more effective in comparison to passive immunisation. Older vaccines were made from the plasma of hepatitis B carriers, obtained by plasmapheresis. Vaccine currently used is a recombinant yeast hepatitis B vaccine prepared through genetic engineering and comprises of non-glycosylated HBsAg particles. Three doses given at 0, 1 and 6 months administered via IM injection into the deltoid muscle (in adults) or into the anterolateral aspect of the thigh (in infants) constitute the full course. Other hepatitis B vaccines which have also been successfully used include recombinant Chinese hamster ovary cell hepatitis vaccine, synthetic peptide vaccines and hybrid virus vaccine.

Hepatitis C Virus

The hepatitis C virus (HCV) is a single-stranded, enveloped RNA virus. It is structurally similar to the Flaviviruses, 30–38 nm in size, having a genome of 9379–9481 base pairs. Whilst both HBV and HCV are transmitted through blood and blood products, HBV is a DNA virus and hepatitis C is an RNA Flavivirus. HCV is mainly transmitted by IV drug use and blood products. Screening of blood donors and blood products has reduced the associated risks, so IV drug use is now the biggest risk factor. Therefore, individuals with HCV are at increased risk of other viral infections, such as HIV and hepatitis B. HCV (incubation period of 30–60 days) can cause acute HCV infection, chronic HCV infection, cirrhosis, hepatoma and other complications induced by hepatitis. It is asymptomatic in nearly 75% cases. It can establish a long-term carrier state in nearly 80% of infected individuals. The risk of sexual transmission from an infected to a non-infected partner is less than 5%. When coexisting HIV infection is present too, the risk of transmitting one or both viruses is much higher. Until recently, HCV was the most likely cause of post-transfusion hepatitis. It accounts for most cases of viral hepatitis previously designated as

FIG. 6.9: Clinical outcomes of acute hepatitis B infection
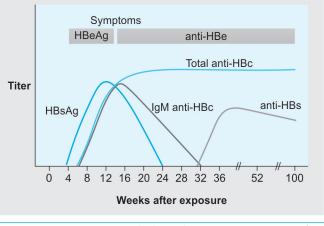
FIG. 6.10: Hepatitis antigens, antibodies and DNA in a patient recovering from acute hepatitis B virus infection

non-A, non-B viral hepatitis. Blood or blood products and as well as the organs of infected patients are the major sources of infection. Sexual and mother-to-baby transmission can sometimes occur. No prophylactic methods are available. For diagnosis, antibody to HCV antigen can be detected by ELISA. The HCV RNA can be amplified by reverse transcription-PCR.

Hepatitis C and Pregnancy

The main risk to the baby is infection with HCV. However, vertical transmission from mother to child is reckoned to occur in less than 10% of cases. The risk is highest for mothers with detectable virus in their blood. The woman whose partner has both HCV and HIV is at a much higher risk of infection. If a woman is both HCV and HIV positive, the risk of vertical transmission to her baby is much increased.

The long-term implications of perinatal infection are unknown. There is no evidence that pregnancy has an adverse effect on the course of the woman's infection. HCV positive women are more at risk of obstetric cholestasis. Interferon and the drug ribavirin have been shown to be effective in the non-pregnant women. Ribavirin is contraindicated in pregnancy.

Infection with HCV per se is not a contraindication for breastfeeding. However, many HCV infected mothers have coexisting other problems (e.g. HIV infection, cracked nipples, etc.), which disallow breastfeeding.

Hepatitis G Virus

This is a recently discovered *Flavivirus*, which is probably similar to hepatitis C. Clinical features of this virus are presently under evaluation. It probably has a predilection for chronic hepatitis disease.

Hepatitis D Virus

The hepatitis D virus (HDV) is an unusual, single-stranded, circular RNA virus and is unique in being an incomplete virus that requires hepadnavirus helper functions for propagation in hepatocytes. Transmission of HDV occurs parenterally. Spread is via co-infection or super-infection with hepatitis B. Double infection with hepatitis B and hepatitis D is particularly severe and carriers of both are at a risk of developing rapidly progressive cirrhosis.

Hepatitis E Virus (HEV)

Hepatitis E virus is the primary cause of enterically transmitted non-A, non-B hepatitis. It usually causes an acute, self-limiting disease similar to HAV. The infection with HEV can be particularly severe in pregnancy with a mortality rate of about 30%. Specific diagnostic tests for infection due to HEV include PCR to detect HEV RNA, and ELISA, which detects both IgG and IgM anti-HEV antibodies. General measures for preventing HEV infection include improving the standards of sanitation and chlorination of water. A vaccine may soon be available.

Rubella

Rubella virus, a single-stranded RNA virus, has been classified in the family Togaviridae as the only member of the genus Rubivirus. Rubella virus is a pleomorphic, roughly spherical particle, 50–70 nm in diameter. Rubella has an incubation period of 14–23 days. Acquired infection usually occurs through inhalation. Congenital infection, on the other hand, occurs via transplacental spread. Nearly 80–90% of the individuals become immune by the age of 15 years. About 10–20% of mothers are non-immune and therefore vulnerable to spread the virus to their foetus. Acquired infection is usually associated with a mild illness called rubella or german measles. This illness is characterised by mild exanthematous fever, transient macular rash (lasting for approximately 3 days) and lymphadenopathy. Primary infection with rubella offers lifelong immunity and therefore congenital malformations are not due to secondary infections. IgM antibodies develop within 2–3 weeks of an acute infection. Patients with acute rubella infection can transmit infection to others from around a week before to 4 days after the development of their rash.

Congenital Rubella

The virus may spread to the foetus through the bloodstream, causing death due to infection in early pregnancy, congenital malformations during the first trimester and more subtle damage in later infections. Congenital rubella infection is presumed to cause chromosomal breakages and inhibition of mitoses in infected embryonic cells. Foetal damage caused by maternal rubella is related to the stage of pregnancy. Maternal rubella is associated with low birth weight and prematurity. Transplacental infection if occurring within the first 4 months of pregnancy can be associated with a high incidence of congenital anomalies, heart lesions (e.g. patent ductus arteriosus, ventricular septal defects, peripheral pulmonary artery stenosis, etc.), cataracts/congenital glaucoma, sensorineural deafness, pigmentary retinopathy, purpura, splenomegaly, jaundice, microcephaly, mental retardation, meningoencephalitis, radiolucent bone disease, etc. These anomalies constitute the classical congenital rubella syndrome. Severe mental deficiency is uncommon.

Pregnancy during which primary rubella infection is contracted has a higher incidence of miscarriage. Thereafter rubella immunity is developed which protects subsequent pregnancies from this complication. Therefore, rubella infection in pregnancy is rarely associated with recurrent miscarriage.

Prophylaxis

The disease being so mild, prophylaxis is directed only towards its teratogenic hazard and so relevant only in women of childbearing age. An obvious method of protection is to acquire the infection before puberty.

This was achieved by "rubella parties", formerly practised in Australia, where adolescent girls voluntarily exposed themselves to known rubella cases.

Passive Prophylaxis

There is little evidence that administration of normal human immunoglobulins after contact reduces the risk of maternal rubella and foetal infection. However, this may help attenuate the severity of illness.

Active Prophylaxis

Several live attenuated rubella vaccines have been developed by serial passage of the virus in tissue culture. The vaccine currently in use is the RA 27/3 strain grown in human diploid cell culture. It is administered by a single subcutaneous injection in the dose of 0.5 mL. The vaccine is available as a single antigen or combined with measles and mumps components as a part of measles, mumps, rubella (MMR) vaccine. Vaccine-induced immunity persists in most vaccines for at least 14–years and probably is life-long.

The vaccine virus is apparently not teratogenic. The rubella vaccine contains a live virus with reduced ability to cause disease. Inadvertent administration of the vaccine to a pregnant woman may not therefore lead to congenital defects in the baby. Nevertheless, the vaccine must not be administered during pregnancy. Prophylaxis is relevant only in women of child-bearing age and is best carried out by immunisation with a live attenuated rubella vaccine. This vaccine is also administered to children at 15 months of age as such or in combination (MMR vaccine).

Parvovirus

Parvoviruses are the smallest amongst the various DNA viruses (about 20 nm). The only virus belonging to this group, which causes human infection, is parvovirus (B19). Human parvovirus B19 may cause respiratory infection with an erythematous maculopapular rash (erythema infectiosum, slapped cheek disease or Fifth disease), joint disease, aplastic crisis in children with chronic haemolytic anaemia (sickle cell disease), non-immune foetal hydrops following infection during pregnancy (second or third trimester of pregnancy) and persistent anaemia in immunodeficient individuals.

Human Immunodeficiency Virus

Acquired immune deficiency syndrome (AIDS) is caused by infection with human immunodeficiency virus (HIV). HIV shows two distinct antigenic types: HIV-1 and HIV-2. HIV is a retrovirus. HIV is a spherical enveloped virus measuring up to 120 nm in diameter and consists of two identical copies of single-stranded RNA genome. The most important enzyme present in the virus is reverse transcriptase, which helps in manufacturing DNA from single-stranded RNA. This DNA becomes incorporated in the host genome, whereby it codes the formation of new viruses.

Important structural components of the virus include the surface antigen gp120, the transmembrane antigen gp41, the matrix protein p17 and the capsid antigen p25. The genome of HIV contains the three structural genes as well as other non-structural and regulatory genes specific for the virus. The products of these genes, both structural and non-structural, act as antigens. Sera of infected persons contain antibodies to them. Detection of these antigens and antibodies is useful in the diagnosis and prognosis of HIV infections. There are three modes of transmission of HIV infection: Sexual contact, parenteral and perinatal transmission.

Human immunodeficiency virus shows trophism for CD4-expressing T-cells and macrophages and principally infects the CD4 lymphocytes. This causes damage to T helper (T4) lymphocytes. As a result, T4 cells are depleted in the humans and the T4:Tg (helper: suppressor) ratio is reversed. When CD4+ cells fall below 200 per mm^3, the titre of virus increases markedly resulting in an irreversible breakdown of immune defence mechanisms. This may result in the development of opportunistic infections and malignancies (e.g. Kaposi's sarcoma). The incubation period of the virus is 52 months. An incomplete virus is likely to have the same effect as the complete one.

Human immunodeficiency virus can cause acute infection, AIDS-related complex (ARC) and AIDS. AIDS is the end-stage disease of HIV infection associated with opportunistic infections, malignancies and neurologic diseases.

Laboratory diagnosis of HIV infection includes specific tests for HIV as well as tests for immunodeficiency. Specific tests include detection of P24 antigen (core antigen), isolation of the virus, detection of viral nucleic acid and antibody detection.

The p24 antigen is the earliest virus marker to appear in the blood. Viral isolation, detection of viral nucleic acid by PCR and p24 antigen detection are useful for diagnosis in window period. The detection of specific antibodies to HIV in the serum is the most commonly used method of serodiagnosis of patients with HIV and AIDS. There are two types of serological tests for anti-HIV antibodies: screening tests and supplementary tests. The screening test commonly includes ELISA, whereas the supplementary tests for confirmation of diagnosis include the western blot and indirect immunofluorescence assay. A safe and effective vaccine is yet not available against HIV. Antiretroviral treatment (ART) is presently the mainstay in HIV treatment.

HIV and Pregnancy

Foetal infection is associated with an increased risk of miscarriage, IUGR and stillbirth. Women with HIV are at

an increased risk of cervical disease. This is not only related to their life-style (promiscuous behaviour), but also to their impaired immunological state. In the western world, the risk of maternal transmission of HIV to the foetus is of the order of 15%. In the developing world, this risk is much greater, perhaps due to breastfeeding. It is now thought that the risk of HIV transmission in the UK can be reduced to less than 1% with the use of various strategies such as aggressive antiviral therapy, elective caesarean section and avoidance of breastfeeding. It is thought that breastfeeding carries some risk of transmitting the virus to the baby, so it should be avoided in a HIV positive woman.

Mother-to-child (M-T-C) transmission is "vertical" transmission. The transmission rates are high in cases where there is no intervention. With optimum intervention, the rate of transmission is less than 1%. In the UK, the rate of transmission is less than 2%. Majority of transmission (>80%) takes place after 36 weeks, mainly during labour and delivery. Less than 2% of transmissions occur during the first and second trimesters.

Risk factors for M-T-C transmission are tabulated in **Table 6.12**.

Currently, there is insufficient evidence for a plasma viral load threshold below which transmission never occurs. However, viral load titres <1,000 copies/mL is associated with a relatively lower risk.

Screening for HIV Infection

If screening has been refused in early pregnancy, screening should be offered again at a later stage in pregnancy. Some women, who are at particular risk of HIV infection and should be especially targeted for screening, include the following:

❑ Women who have arrived in the UK as refugees or asylum seekers from high prevalence countries
❑ Those with a history of injecting drug use
❑ Commercial sex workers.

TABLE 6.12	Risk factors for mother-to-child transmission

- Advanced maternal disease
- High maternal viral load
- Resistant strain of the virus
- Low maternal CD4 count
- Malnutrition, especially vitamin A deficiency
- Smoking and use of illicit drugs
- Maternal obesity
- Other sexually transmitted diseases, particularly ulcerative

Obstetric Risk Factors
- Prolonged duration of ruptured membranes
- Chorioamnionitis
- Preterm delivery: especially <34 weeks
- Vaginal delivery
- Use of scalp electrodes
- Episiotomy
- Perineal tears and lacerations
- Breastfeeding
- Prolonged labour
- Active genital ulcer disease

Consideration should be given in the third trimester for rescreening women who have continuing high risk of HIV acquisition. In certain situations, it is beneficial to test both the pregnant mother and her partner for HIV. If the pregnant mother is negative but her partner is positive, advice can be given about avoiding transmission of HIV during pregnancy.

Antenatal Period

Women diagnosed as HIV positive during pregnancy should be managed by a multidisciplinary team including an HIV physician, an obstetrician, a midwife, a paediatrician, as well as social support workers. Confidentiality regarding HIV status must be maintained at all times. The following steps need to be taken amongst the pregnant women:

❑ *Screening for genital infections*: All pregnant women who are HIV positive should be screened and treated for genital infections early in pregnancy. This should be repeated during the third trimester. Chlamydia, gonorrhoea, bacterial vaginosis and genital ulceration should especially be looked for. All these genital infections are associated with a greater risk of HIV disease. Additionally, bacterial vaginosis is also associated with preterm labour. Routine antenatal serological screening for hepatitis B and syphilis should also be carried out. Syphilis serology should be repeated in the third trimester. All HIV positive pregnant women should also be tested for HCV.

❑ *Invasive therapy*: Prophylactic therapy with HAART (highly active anti-retroviral therapy) should be considered before carrying out any invasive monitoring (amniocentesis, CVS, etc.).

❑ *Screening for foetal anomalies*: Screening for Down syndrome and foetal anomalies should be carried out according to local protocols. A detailed ultrasound scan for foetal anomalies should be carried out at 21 weeks gestation if antiretroviral therapy or prophylaxis for PCP (*Pneumocystis carinii* pneumonia) with folate antagonists [e.g. Septran (co-trimoxazole)] has been used during the first trimester.

❑ *Monitoring for drug toxicity*: Pregnant women with HIV should be monitored regularly for drug toxicity. Signs of toxicity due to the use of antiretroviral drugs include pre-eclampsia, liver dysfunction, lactic acidosis, glucose intolerance, diabetes, rashes, etc. Zidovudine is the antiretroviral for which the most extensive safety data is available regarding use in pregnancy.

❑ *Antiretroviral therapy during pregnancy*: Prescription of antiretroviral therapy may be required to reduce the risk of HIV transmission to the foetus. Women who do not need antiretrovirals to control disease need to be given antiretrovirals to prevent M-T-C transmission. The risk of HIV transmission needs to be balanced with the risk of the therapy related toxicities. Zidovudine monotherapy is an option for the woman with low levels of HIV viraemia and with a sensitive strain if she does not require HAART for her own health or who does not

wish to take HAART during pregnancy. Women with HIV who require antiretroviral therapy to control disease should be prescribed the same regimes as if they were not pregnant. However, certain combinations of drugs should be avoided during pregnancy. These include dual NRTI (non-reverse transcriptase inhibitors); combination of stavudine with didanosine (due to the risk of lactic acidosis), etc.

Women taking HAART are at risk of premature labour. Till date, there is no evidence of any increase in congenital malformations in humans with first trimester exposure to any antiretroviral therapy. However, there is inadequate data to exclude a teratogenic risk for most drugs. Mitochondrial depletion and haematological effects have been noted in infants exposed to antiretroviral therapy.

Labour and Delivery

- ❑ *Mode of delivery*: A planned caesarean section should be offered to all HIV positive women in the cases, where there is a detectable plasma viral load and/or the woman is not taking HAART at approximately 38 weeks of gestation. Planned caesarean section is also advised if there is coinfection with hepatitis B or C. Risks and benefits of caesarean section need to be considered in each individual case.
- ❑ The role of caesarean section in women with HIV taking HAART or those who have undetectable viral loads, presently remains unresolved. However, the latest British HIV Association guidelines recommend considering vaginal delivery as an option for these women.
- ❑ *Precautions during delivery*: Consideration should be given towards using IV zidovudine prophylaxis during delivery. In case zidovudine is being administered to a woman having a planned section, zidovudine infusion should be commenced 4 hours prior to the surgery and this should be continued until the cord is clamped. IV zidovudine prophylaxis is not usually indicated for mothers not on zidovudine treatment or for mothers on HAART with <50 HIV RNA copies/mL plasma.
- ❑ The cord should be clamped and the baby bathed as soon as possible after delivery.
- ❑ The use of foetal scalp electrodes and foetal blood sampling should be avoided in HIV infection.
- ❑ Artificial rupture of the membranes should preferably be avoided.
- ❑ Perioperative antibiotics should be given for all caesarean section deliveries and immediately if membranes rupture during the first stage of labour.
- ❑ Corticosteroids should be given for threatened preterm delivery.

Post-natal Care

- ❑ *Breastfeeding*: All women in the UK who are HIV positive should be advised not to breast feed because breastfeeding is likely to increase the overall rate of M-T-C HIV transmission.

- ❑ Zidovudine is usually administered orally for 4 weeks to all neonates of mothers with HIV. Alternative therapy may be prescribed if maternal therapy does not include zidovudine.
- ❑ Triple therapy as PEP (post-exposure prophylaxis) should be considered for infants born to untreated mothers or mothers with detectable viraemia despite combination therapy.

Mycology

Pathogenic fungal infections caused by the dimorphic fungi include blastomycosis, histoplasmosis, coccidioidomycosis and cryptococcosis. Of these various organisms, cryptococcosis and candidiasis are the most common systemic pathogenic organisms.

Cryptococcosis

Cryptococcosis is sub-acute or chronic infection caused by the capsulate yeast *Cryptococcus neoformans*. *C. neoformans* causes: Pulmonary cryptococcosis in immunocompromised hosts; CNS cryptococcosis; disseminated non-pulmonary non-CNS cryptococcosis. In unstained, wet preparations of CSF mixed with a drop of India ink, the capsule can be seen as a clear halo around the yeast cells.

Cryptococcal capsular polysaccharide antigen can be detected in CSF and blood by latex agglutination and ELISA test.

Candidiasis

Candidosis (candidiasis or moniliasis) is an infection of the skin, mucosa, and rarely of the internal organs, caused by a yeast-like fungus *Candida albicans*, and occasionally by other Candida species. Candida is Gram-positive yeast, which is a normal commensal organism present in the mouth, gut and vagina. Candidiasis is an opportunistic endogenous infection, the commonest predisposing factor being diabetes. It can cause superficial as well as systemic infections. Superficial candidial lesions include mucocutaneous lesions (oral thrush, vulvovaginitis, balanitis, conjunctivitis, keratitis, etc.), and skin and nail infections.

Vulvovaginal candidiasis is associated with a white, thick discharge, having no odour and a normal pH. Pruritus vulva is a cardinal feature. Women with vulvovaginal candidiasis frequently complain of pruritus, vaginal irritation, dysuria, vulvar and vaginal erythema and occasionally, vulvar burning, soreness, scaling and fissures of vulvar tissue.

Systemic candidiasis occurs when candida enters the bloodstream and the phagocytic host defences are inadequate to contain the growth and dissemination of the yeasts. Systemic infections include intestinal candidiasis, bronchopulmonary candidiasis, septicaemia, endocarditis, meningitis, kidney infections, UTIs, etc. Severe systemic infection can occur in cases of deficient cytotoxic immunity (e.g. administration of cytotoxic drugs).

Protozoal Infections

Various protozoal infections include malaria (protozoan belonging to the genus *Plasmodium*), amoebiasis (*Entamoeba histolytica*), toxoplasmosis (*Toxoplasma gondii*), trichomoniasis (Trichomonas vaginalis), leishmaniasis (protozoan belonging to the genus *Leishmania*), etc.

Toxoplasmosis

Toxoplasmosis is caused by *Toxoplasma gondii*, an obligate intracellular parasite belonging to the Apicomplexa family. The life cycle of the *T. gondii* parasite has three stages: cyst, oocyst, or tachyzoite. The organism is identified with the Giemsa stain. It is usually transmitted through ingestion of water, soil, vegetables, or anything contaminated with oocysts shed in the faeces of an infected animal (most often cats) but can also be acquired through consumption of raw or undercooked meat containing *T. gondii* tissue cysts. The protozoan can proliferate in the CNS forming cysts. In adults, it may cause a mild febrile illness. Infection in the foetus occurs when the mother acquires a primary infection during or shortly before pregnancy. This is particularly the case in the immunocompromised host and infection is a serious problem during pregnancy because foetal infection is associated with stillbirth, prematurity, IUGR, hepatitis, thrombocytopenia, neurological (hydrocephaly, microcephaly, etc.) and ophthalmological abnormalities (retinitis). If the foetus is not infected, acute infection in pregnancy can be treated with spiramycin. In case the foetus is infected, a combination of sulphadiazine and pyrimethamine is used.

Trichomoniasis

Protozoa *Trichomonas vaginalis* is a motile organism currently accounting for 10–25% of vaginal infections (trichomoniasis). The protozoan is pear-shaped, having 3–5 flagella at the anterior end and a single flagellum forming part of the undulating membrane at the posterior end. Trichomonads are usually transmitted sexually.

Classic manifestations of vaginal trichomoniasis include a purulent, frothy, yellow discharge with an abnormal odour, pruritus and dysuria. The typical discharge associated with this infection is profuse, thin, creamy or slightly green in colour, irritating and frothy. Trichomoniasis may commonly act as a vector for other STDs, including, the HIV.

Immunology

Organisation of the Immune System

The immune system is organised into several special tissues which are collectively termed as lymphoid or immune tissues. The lymphoid organs are classified into primary (central) and secondary (peripheral) lymphoid organs based on their function. The primary lymphoid organs include sites where lymphocytes mature and become antigenically committed. These include thymus and bone marrow. T lymphocytes mature within the thymus, and B lymphocytes arise and mature within the bone marrow of some vertebrates including humans.

Secondary primary lymphoid organs capture antigens and provide sites where lymphocytes become activated by interaction with antigens. Secondary lymphoid organs include spleen and the lymph nodes.

Thymus is a primary lymphoid organ that develops mainly from the endoderm of the third pharyngeal pouch. Some of the epithelial cells form the thymic (Hassall's) corpuscles; others form a network of epithelial reticular cells, which secrete thymic hormones that are linked to differentiation of T lymphocytes. The developing thymus descends in front of all the major structures of the neck to enter the anterior mediastinum. It rapidly becomes double in size after birth and stays the same in size throughout life. However, the lymphoid components reduce with age. Nevertheless, the secretion of thymic hormones and its effect on lymphocytes that migrate to it continue throughout life.

Immune Response

Immunity refers to the resistance exhibited by the host towards injury caused by microorganisms and their products. Immunity can be innate or natural and acquired. The immune response is the specific reactivity induced in a host by an antigenic stimulus and can be divided into two types: the humoral (antibody-mediated) and the cellular (cell-mediated) types.

- *Humoral immunity*: The humoral immunity results from the activation of naive lymphocytes (primary response) or memory lymphocytes (secondary response).
- *Cell-mediated immunity*: Cell-mediated immunity refers to the specific immune responses which involve functions mediated via T-lymphocytes. There is no involvement of antibodies in the cell-mediated immunity.

Primary cell-mediated immune response is produced by initial contact with a foreign antigen. Secondary cell-mediated immune response occurs if the same host is subsequently exposed to the same antigen. As a result, the secondary cell-mediated immune response is usually more pronounced and occurs more rapidly in comparison to the primary cell-mediated immune response.

Innate and Acquired Immunity

Innate Immunity

Innate or natural immunity is the resistance to infections, which an individual possesses by virtue of his genetic or

constitutional make up. Factors influencing the level of innate immunity are age, hormonal influences, gender, nutrition, and stress. Mechanisms of innate immunity include the following: mechanical barriers and surface secretions; presence of antibacterial substances in blood and tissues (e.g. complement system); microbial antagonisms; cellular factors in innate immunity (e.g. macrophages); inflammation, fever, acute phase proteins, etc.

Acquired Immunity

Acquired immunity refers to the resistance that an individual acquires during his/her lifetime. Acquired immunity can be obtained by natural or artificial means and actively or passively. Therefore, acquired immunity is of two types: (1) active immunity and (2) passive immunity. Comparison between active and the passive immunity is summarised in **Table 6.13**.

Active Immunity

Active immunity, also known as the adaptive immunity, is induced after contact with foreign antigens. This immunity involves the active functioning of the host's immune apparatus leading to the synthesis of antibodies and/or the production of immunologically active cells. Active immunity can be obtained by natural or artificial means. Natural active immunity results from either a clinical or an inapparent infection by a microbe. Artificial active immunity is the resistance induced by vaccines. Vaccines could be composed of any of the following: live, attenuated microorganisms; killed microorganisms; microbial extract; vaccine conjugates, and inactivated toxoids.

Passive Immunity

The immunity that is transferred to a recipient in a "readymade" form is known as passive immunity. Passive immunity can also be obtained through natural or artificial means. An example of the natural passive immunity is the passive transfer of immunity from mother to baby through the placenta. On the other hand, artificial passive immunity is the resistance passively transferred to a recipient by the administration of antibodies (e.g. pooled human gamma globulin, hyperimmune sera of animal or human origin and convalescent sera).

Cells and Humoral Elements of Acquired Immunity

The white blood cells called leucocytes are responsible for both non-specific and specific immunity in humans. All of the leucocytes originate from pluripotent stem cells in the foetal liver and in the bone marrow from where they migrate to other body sites. Here, they undergo further development and perform various functions.

Lymphocytes, a type of white blood cell have now been recognised as the major cellular elements responsible for immunological responses. They constitute 20–40% of the body's white blood cells. These lymphocytes continually circulate in the blood and lymph and are capable of migrating into the tissue spaces and lymphoid organs, thereby greatly integrating the immune system. Many mature lymphoid cells are long-lived, and persist as memory cells for many years. The lymphocytes can be broadly subdivided into three populations: B cells, T cells and natural killer (NK) cells. Difference between the B cells and T cells has been summarised in **Table 6.14**.

T Cells

T cells can be classified based on their surface markers, major histocompatibility complex (MHC) restriction, target cells and function. T cells are the predominant cell type in the paracortical regions of lymph nodes. These cells are capable of binding to the sheep erythrocyte, resulting in the formation of SRBC or the E rosettes, due to the presence of a CD2 receptor on the surface of T cells. The CD2 receptor also acts as a receptor for the measles virus. B cells on the other hand do not possess this.

TABLE 6.13	Comparison between active and passive immunity	
Characteristics	**Active immunity**	**Passive immunity**
Mode of production	Induced by infection or by immunogens	Readymade antibody transferred
Participation of the host's immune system	Produced actively by host's immune system	Received passively. There is no active host participation
Efficacy of the immunity provided	Durable effective protection	Transient, less effective
Time duration for immunity production	Immunity effective only after lag period, i.e. time required for generation of antibodies and immunocompetent cells.	Provision of immediate immunity
Presence of memory	Immunological memory present	No immunological memory
Effect of a subsequent dose	Subsequent dose is associated with a booster effect	Subsequent dose less effective
Occurrence of a negative phase	"Negative phase" may occur	No negative phase
Role in immunodeficient individuals	Not applicable in the immunodeficient individuals	Applicable in immunodeficient individuals

TABLE 6.14   **Comparison between the B cells and the T cells**

Property	T cell	B cell
Origin	Bone marrow	Bone marrow
Maturation	Thymus	Bursal equivalent: Bone marrow, Payer's patches
Cell location		
Peripheral blood	65–85%	15–25%
Lymph node	60–75%	30–35%
Spleen	25–45%	55–60%
Thoracic duct	80–90%	10–20%
Thymus	96%	Negligible
Thymus specific antigens	+	–
Presence of antigen-binding T cell receptor (TCR): showing resemblance to CD3-associated proteins	+	–
Surface immunoglobulins	–	+
Receptor for Fc piece of IgG	–	+
SRBC rosette (CD2; measles receptor)	+	–
EAC rosette (C3 receptor; CR2; EBV receptor)	–	+
Numerous microvilli, on surface	–	+
Blast transformation with:		
Anti-CD3	+	–
Anti-Ig	–	+
PHA	+	–
Concanavalin A	+	–
Endotoxins	–	+

Abbreviations: EAC, erythrocyte coated with antibody and complement; SRBC, sheep red blood cell; EBV, Epstein-Barr virus

Classification of T Cells

On the basis of surface markers: Classification of lymphocytes on the basis of surface markers makes use of two important characteristics: cluster of differentiation or cluster determinant (CD) and antigen recognition receptors.

The term CD refers to the family of surface glycoprotein antigens that can be recognised by specific antibodies produced against them. The binding of specific antibodies against CD identifies the cell displaying CD. Each class of leucocyte displays a diagnostic pattern of CDs. Over 200 CD markers have been identified so far. Some examples include the following:

❏ CD3 is expressed only by T cells
❏ CD19 is expressed only by B cells
❏ CD64 is expressed only by monocytes
❏ CD66 is expressed only by granulocytes
❏ CD68 is expressed only by macrophages.

Antigen recognition receptors include membrane-bound (surface) immunoglobulins (mIgs or sIgs) in B cells and T cell receptors (TCRs) in T cells. In contrast to CDs, which can serve as diagnostic feature for all leucocytes, antigen recognition receptors are limited to B and T lymphocytes only. Reaction of antigens with mIgs and TCRs activates B cells and T cells respectively, leading to proliferation and differentiation.

On the basis of function: On the basis of their function, T cells can be classified as regulatory T cells and effector T cells.

Regulatory T cells:
❏ *Helper/inducer cell (Th)*: "T helper" cells are CD4+ T cells. They are restricted in antigen recognition by self-MHC class II molecules. MHC class II restriction generally helps to stimulate and promote the growth of T cells and macrophages. They also help B cells to make antibodies in response to antigenic challenge and stimulate cell-mediated immunity. Based on the different profiles of cytokines produced, two subsets of helper cells have been identified: Th1 and Th2. They also mediate delayed type hypersensitivity by release of cytokines in response to antigenic stimulation.
❏ *Suppressor T cells (Ts cells)*: These have CD8 surface marker and MHC class I restriction. They can suppress the response of B cells and the T cells.

Effector cells:
❏ *Delayed type-hypersensitivity T cells (Td cells)*: They are involved in delayed hypersensitivity and cell-mediated immune response.
❏ *Cytotoxic T cells (Tc cells)*: They are also called CD8+ cells. They have CD8 surface marker and MHC class I restriction. They can kill and lyse target cells carrying new or foreign antigens, including tumour, allograft and virus infected cells.
❏ *Memory cells (Tm)*: Both CD4 and CD8 cells provide memory and an amnestic immune response.

The Major Histocompatibility Complex

Major histocompatibility complex (MHC) is a set of cell surface molecules, which play a major role in controlling the immune system. The genes of the MHC are located on the short arm of human chromosome 6. The MHC in humans is known as human leucocyte antigen (HLA) complex. In humans, the HLA complex of genes is located on short arm of chromosome 6 containing several genes that are critical to immune function. The genes encoding MHC proteins are classified into three groups or classes known as the class I, class II and class III molecules. HLA typing or tissue typing is usually performed for the following:

❏ Tissue transplantation
❏ Disputed paternity
❏ Anthropological studies
❏ An association between HLA types and diseases.

B Lymphocytes

B lymphocytes are produced in the bone marrow and require bone marrow cytokines and stromal cells for their maturation. B cells have surface bound immunoglobulin (unlike the T lymphocytes). They are the predominant cells in lymph node germinal follicles. Through binding of antigen to specific epitopes, the B cells synthesise antibodies, termed humoral immunity. B cells are capable of binding to erythrocytes coated with antibody and complement, forming EAC rosettes, due to the presence of a C3 receptor (CR2) on the B cell surface. This receptor (CR2) also acts as a receptor for the EBV. T cells, on the other hand, do not possess this.

Natural Killer Cells

Natural killer (NK) cells are a lymphocytic lineage discrete from T + B lymphocytes and are derived from large granular lymphocytes. The activity of NK cells is "natural" or "non-immune" as it does not require sensitisation by prior antigenic contact. These cells therefore form part of the innate immune setup. They are involved in providing defence against malignancy, viruses and probably bacteria and parasites. They are rarely found in thymus/lymph nodes—unlike T cells. NK cells are CD3 negative and CD16 and CD56 positive (the opposite is true of T cells). NK cells kill spontaneously, are stimulated by IL-2 or antibody coated cells through binding to their CD16 receptors. They also release interferon gamma (IFN-γ), granulocyte-macrophage colony stimulating factor and colony stimulating factor 1.

Phagocytic Cells

Phagocytic cells include the mononuclear macrophages (present in the blood and tissues) and the polymorphonuclear microphages.

Mononuclear Phagocytic System

The mononuclear phagocytic system consists of monocytes circulating in the blood as well as the macrophages present in the tissues. Both the types of cells are highly phagocytic. The blood macrophages (monocytes) are the largest of the lymphoid cells found in peripheral blood (12–15 μm). The tissue macrophages (histiocytes) are larger (15–20 μm). Monocytes leave the circulation and reach various tissues to become transformed into macrophages, having morphological and functional features characteristic of the tissues. Based on the final place of residence of the macrophages, they can be given different names, e.g. alveolar macrophages in the lung, histiocytes in connective tissues, Kupffer cells in the liver, mesangial cells in the kidney, microglial cells in the brain and osteoclasts in the bone. One of the most potent activators of macrophages is IFN-γ secreted by activated Th cells.

Opsonisation: Phagocytosis, though possible in a saline medium, is enhanced in the presence of fresh serum. This is due to presence of substances called opsonins. Opsonins include C3b, for which phagocytic cells possess a receptor. Immunoglobulins, especially the F_c portion of IgG1 and IgG3 are potent opsonising agents for the phagocytic cells possessing these receptors. Opsonisation is not MHC-restricted.

Polymorphonuclear Microphages

Microphages are the type of white blood cells, characterised by the presence of granules in their cytoplasm. They are therefore known as granulocytes. Due to the presence of an irregular-shaped nuclei, granulocytes are also called polymorphonuclear leucocytes. Three types of granulocytes exist: basophils, eosinophils and neutrophils. Neutrophils are actively phagocytic and form the predominant cell type in acute inflammation. The phagocytic property of neutrophils is non-specific, except for its augmentation by opsonins. The eosinophils have several granules containing a variety of hydrolytic enzymes which bring about extracellular killing of large parasites. Eosinophils possess phagocytic activity but only to a limited degree. They are found in large numbers in allergic inflammation, parasitic infections and around antigen-antibody complexes.

Basophil leucocytes are found in the blood and tissues (where they are known as the mast cells). Mast cells have been described later in the text.

Dendritic Cells

While macrophages are the major antigen presenting cells, another type of cell known as the dendritic cell also performs this function. Dendritic cells are derived from bone marrow and constitute a lineage different from the macrophages and T or B-lymphocytes. Four types of dendritic cells are known include Langerhans cells, interstitial dendritic cells, myeloid cells and lymphoid dendritic cells. They possess MHC class II antigens but not Fc or sheep RBC receptors or surface immunoglobulins.

Mast Cells

Mast cells are basophilic cells present in the connective and subcutaneous tissues, which are involved in inflammatory and immune responses. They contain storage granules that contain lytic enzymes (e.g. tryptase) and inflammatory mediators, (e.g. histamine, heparin, serotonin, leucotrienes, platelet aggregating factor, leucocyte chemotactic factor, hyaluronidase, etc.). Release of these mediators occur during mast cell degranulation, which can be triggered by events such as tissue injury, drugs, complement activation and foreign antigenic material. An anaphylactic reaction occurs when a previously sensitised individual is re-exposed to the antigen. It is an IgE-mediated immune response. Mastocytosis occurs when excess mast cells are present in the circulation or as tissue infiltrates.

Immunogenetics and Principles of Antigen Recognition

Antigens

Antigens are the substances that can stimulate an immune response by specifically binding with the effector molecules (antibodies) and effector cells (lymphocytes). Antigens are usually proteins or polypeptide molecules. Large carbohydrate molecules may also be antigenic. Antigens can be recognised by immune system cells even if previously not exposed to that antigen. The ability to recognise foreign antigens is innate and does not depend on previous exposure to them. Antigens, being proteins or carbohydrates, are not normally absorbed; they are digested in the gut. Antigens are taken up by antigen-presenting macrophages, which activate the immune system. Antigens can also act directly on receptors on lymphocyte membranes. Antigens can be of two types: complete or incomplete (haptens). Determinants of antigenicity are as follows: size, chemical nature, foreignness, susceptibility to tissue enzymes, antigenic specificity, species specificities, etc.

Super antigens are bacterial proteins which can interact with antigen-presenting cells (APCs) and T cells in non-specific manner. The antigens which provoke such a drastic immune response are termed as super antigens.

Immunoglobulins

An antibody or immunoglobulin (Ig) is a glycoprotein that is produced in response to an antigen, and can recognise and bind to the antigen that caused its production. They have an average molecular weight of approximately 100 kDa and are produced on the ribosomes of plasma cells. Since organogenesis starts occurring at 12 weeks, antibody production in the foetus does not develop until much later. Antibodies are absent from the blood in early foetal life. Immunological tolerance prevents the foetus from forming antibodies to its own proteins.

In humans, there are five classes of antibodies, namely IgA, IgD, IgE, IgG and IgM (Table 6.15). Immunoglobulins are secreted from B-lymphocytes (plasma cells) in response to a specific antigen. The response to the second antigenic exposure is greater in comparison to the first exposure since because the immune system has been sensitised by the first exposure.

Each antibody molecule consists of four polypeptide chains: two identical light chains and two identical heavy chains, which are linked by disulphide bonds (Fig. 6.11). Each heavy chain has an amino-terminal variable region followed by a constant region. The variable regions of one heavy chain and one light chain form an antigen binding site. Each immunoglobulin molecule comprises of two fragments: F_{AB} (antigen binding fragment) and F_c (crystalline fragment). F_{AB} is a highly variable region,

TABLE 6.15	Various types of antibodies and their properties	
Antibody	Function	Characteristic
IgA	Protection of the mucosal surfaces	Secreted in the breast milk, tears, saliva, etc.
IgD	Role is uncertain	Found in the serum
IgE	Activation of the mast cells	Involved in allergic response and anaphylaxis
IgG	Fixes complement; opsonising properties	Produced as a result of the secondary immune response; crosses placenta
IgM	Fixes complement; opsonising properties	Produced as a result of the primary immune response, i.e. it is a default antibody which is made first in the body
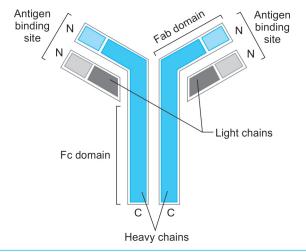
FIG. 6.11: Structure of an antibody

which determines the affinity/specificity of antibody for antigen. Within the amino-terminal variable domain of each heavy and light chain are three complementarity-determining regions (CDRs). These polypeptide regions contribute to the antigen-binding site of an antibody, which determines its specificity. Antigenic determinants on immunoglobulins are: (1) Isotypes; (2) Allotypes and (3) Idiotypes. F_c (fragment crystalline), on the other hand, is formed from constant domains of heavy chains, which define isotope of antibody (e.g. IgG, IgA, etc.). The isotypes determine the specific properties of various antibodies, for example, binding of complement, ability to cross placenta, and binding of mast cells and basophils, etc.

Each of the domains in the immunoglobulin molecule has a characteristic tertiary structure called the immunoglobulin fold. The majority of immunoglobulins produced in the humans are gamma-globulins, but there is some electrophoretic activity in the alpha and beta regions.

Abnormalities in immunoglobulins may be associated with diseases such as multiple myeloma, heavy chain disease C, cryoglobulinaemia, etc.

Immunoglobulin A

Immunoglobulin A is the major immunoglobulin present in the external secretions, e.g. intestinal fluids, saliva, bronchial secretions and colostrum. IgA antibody also provides antiviral activity. IgA plasma concentration is 200 mg/dL. IgA occurs in two forms: Serum IgA and secretory IgA.

Serum IgA occurs in the monomeric form, whereas secretory IgA occurs in a dimeric form **(Fig. 6.12)**.

Immunoglobulin E
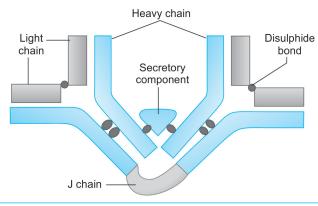
It is the rarest type of immunoglobulin. The main function of immunoglobulin E is the release of histamine from basophils and mast cells. It is involved in type I hypersensitivity mechanism.

Immunoglobulin M

Immunoglobulin M is the largest immunoglobulin. It is a pentamer comprising of five joined IgG units. In the circulation, IgM exists as a pentamer of five, four-chain units. The five identical IgM monomers are connected to each other by a polypeptide joining J chain **(Fig. 6.13)**. The main function of IgM is complement fixation. Immunoglobulin M (IgM) is cryoglobulin.

Immunoglobulin G

IgGs constitute approximately 75% of all immunoglobulins in a healthy individual. Concentration of IgG in plasma is 1,000 mg/dL. Out of the various immunoglobulins present in the human body, IgG is the only one which freely crosses the placental barrier. This is important in affording passive immunity to the neonate before its own immune system has matured. This is important as they provide immune protection for the newborn in the first few months of life. Babies do not start producing their own IgG until the maternal IgG has been catabolised at about 3–4 months of age.

Immunoglobulins G are produced by plasma cells. Immunoglobulin G is produced as the part of secondary immune response. Individuals vary widely in their production of IgG.

The IgG comprises of two antigen-binding sites and a site for the binding of complement. IgG chains are coded for by adjacent genes on the different chromosome. Kappa light chains are coded for on chromosome 12, lambda chains on chromosome 22, and heavy chains on chromosome 14.

Immunodeficiency

Immunodeficiency results from the failure of one or more components of the immune system, e.g. T cells, B cells, complement or phagocytes. For example, deficiencies in C2 and C4 components of the complement pathway can result in diseases such as paroxysmal nocturnal haemoglobinuria, and hereditary angioedema. Disorders of phagocytosis include diseases such as chronic granulomatous disease, myeloperoxidase deficiency (Chediak-Higashi syndrome), leucocyte adhesion deficiency, Job's syndrome, etc.

Disorders of immunodeficiency can be classified as primary or secondary immunodeficiencies. Primary immunodeficiency is a condition resulting from a genetic or developmental defect in the immune system. Primary immunodeficiency may affect either adaptive or innate immune functions. On the other hand, secondary immunodeficiency is the loss of immune function, resulting from exposure to various agents. It can result from malnutrition, use of immunosuppressive agents, infections (such as AIDS) and malignancies.

Autoimmunity

Autoimmunity is a condition in which structural or functional damage is produced by the action of immunologically

FIG. 6.12: Structure of secretory IgA
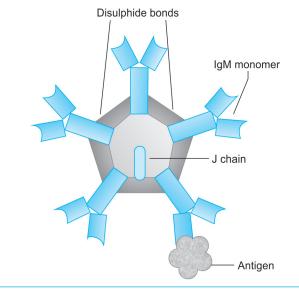
FIG. 6.13: Structure of IgM molecule composed of five identical monomeric units

competent cells or antibodies directed against the normal body components. A variety of mechanisms have been proposed for induction of autoimmunity, including release of sequestered antigens, molecular mimicry, inappropriate class II MHC expression on cells, and polyclonal B cell activation. Autoimmune diseases can be divided into organ-specific, or widespread and systemic diseases. Autoimmune diseases are usually treated with drugs that suppress the immune and/or inflammatory responses.

Immunological Problems in Pregnancy

Antiphospholipid Syndrome

Antiphospholipid syndrome (APS) is due to the presence of autoantibodies, which predispose to the development of serious disease, particularly vascular thrombosis. APS is characterised by the association of arterial and venous thrombosis with antibodies directed against phospholipids. APS affects phospholipid-based coagulation tests. Presence of anticardiolipin antibody (ACA) should principally be sought in these cases. APS is associated with characteristic features of recurrent foetal loss, impaired foetal growth, placental abruption, severe growth restriction and premature delivery, etc. This is a condition associated with the presence of auto-antibodies that predispose to the development of serious disease, particularly vascular thrombosis. Other manifestations of APS include renal vein thrombosis, Addison's disease, myocardial infarction, valvular lesions, transient ischaemic attacks (TIAs), stroke, epilepsy, leg ulcerations, deep vein thrombosis (DVT), pulmonary hypertension (due to thromboembolic disease), etc. Left ventricular thrombus is rarely reported.

Antiphospholipid syndrome may be primary or secondary. Primary APS implies that the antibodies are present without any underlying specific autoimmune disorder. Secondary APS, on the other hand, is associated with a recognised underlying autoimmune disorder, usually systemic lupus erythematosus (SLE). Therefore, there is often an overlap with SLE in the cases of APS. Some patients with SLE will thus have lupus anticoagulant (LA), which is often associated with thrombosis.

The two main antibodies present in cases of APS include ACA and LA. They prolong phospholipid-based coagulation tests such as the activated partial thromboplastin time and kaolin clotting time. Patients with LA or ACA, or both, have an increased risk of miscarriage, mostly occurring in the first trimester. So, screening for LA and ACA should be done in all patients with recurrent miscarriage.

The standard management in cases of APS comprises of low dose aspirin and heparin, which have been shown to improve the prognosis in a couple of trials. The combination of heparin and aspirin appears to reduce the incidence of late pregnancy complications.

Systemic Lupus Erythematosus and Pregnancy

In women, SLE has a prevalence of about 1 in 1,000. In women with SLE who conceive, 50% of babies will be born alive. The optimal situation is control of disease activity for at least 6 months before conception, without the use of cytotoxic agents. Worsening lupus nephritis during pregnancy may be associated with symptoms such as proteinuria, oedema, thrombocytopenia and raised blood pressure. This may resemble the symptoms of pre-eclampsia. Since it is difficult to differentiate between the two conditions, the patients are often treated for both conditions simultaneously. Patients with renal involvement due to SLE or a previous pregnancy with pre-eclampsia or intrauterine growth retardation, or both are advised to take aspirin 75 mg daily throughout pregnancy. At the same time, it should be considered that preterm babies whose mothers have taken aspirin are at an increased risk of intracranial haemorrhage.

Neonatal SLE usually results from passively acquired maternal anti-Ro antibodies. Cutaneous neonatal lupus is the most common form and appears as a self-limiting rash, usually on the face and scalp, which is exacerbated by sunlight. No treatment is required. High-dose corticosteroids, and also plasmapheresis is often administered to the mothers once the neonatal lupus is detected in the foetus. However, this is associated with little success.

Congenital heart block occurs due to maternal autoanti-bodies causing damage to the foetal cardiac conducting system. This is an extremely rare condition usually developing in utero from about 18 weeks' gestation and causing irreversible damage. Nearly 70% of the surviving neonates with heart block may require a pacemaker. High-dose corticosteroids, and also plasmapheresis, have been given to the mothers once the neonatal lupus is detected in the foetus. However, this is associated with little success.

Rheumatoid Arthritis

Pregnancy alleviates the symptoms of RA. Exacerbations of RA tend to occur in the puerperium. Treatment of RA with penicillamine during pregnancy may weaken foetal collagen. Therefore, use of penicillamine during pregnancy has been found to be associated with isolated case reports of cutis laxa, inguinal hernia and joint hypermobility. Use of azathioprine, on the other hand, has been found to be associated with intrauterine growth restriction.

Immunology of Transplantation

Transplantation can be defined as the transfer of cells, tissues, or organs from one site in an individual to another,

or between two individuals. There are four different basic types of transplants: autograft, isograft, allograft and xenograft.

❑ *Autograft*: Autograft is self-tissue transferred from one body site to another in the same individual.

❑ *Isograft*: Isograft is tissue transferred between genetically identical individuals (e.g. monozygotic twins).

❑ *Allograft*: Allograft is tissue transferred between genetically different members of the same species.

❑ *Xenograft*: Xenograft is tissue transferred between different species (e.g. the graft of a baboon heart into a human).

At times, the graft tissue may be rejected in the recipient's body. The immune response to tissue antigens encoded within the MHC is the strongest force in rejection. There are three major types of rejection reactions: hyperacute rejection, acute graft rejection and chronic rejection.

Graft-versus-host (GVH) Reaction

This is a complication that can occur after transplantation of stem cells or bone marrow. With graft-versus-host disease (GVHD), the newly transplanted donor cells attack the recipient's body. For graft-versus-host reaction to occur, three important components are required: the donor graft must contain immunocompetent T cells. The host must be immunocompromised so that the graft cannot be rejected. Also, the recipient should express antigens such as MHC proteins, which will be identified as foreign to the donor.

Hypersensitivity

Hypersensitivity is an exaggerated immune response that results in tissue damage and is manifested in the individual on the second or subsequent contact with an antigen. Hypersensitivity reactions are categorised according to the type of immune response and are traditionally known as types I, II, III, IV and V.

Type I Hypersensitivity Reaction

Type I hypersensitivity reaction, also known as immediate hypersensitivity, is mediated by IgE antibodies. In these cases, the associated allergen binds with the IgE antibodies present on the surface of mast cells. This causes activation of the mast cells, resulting in their degranulation and release of histamine into the circulation. Clinical manifestations of type I reactions include generalised or systemic anaphylaxis or localised anaphylactic (type I) reactions. This type of reaction is seen in the cases of atopy and anaphylaxis following exposure to allergens, e.g. peanuts.

Type II Hypersensitivity Reactions

Type II hypersensitivity reactions, or cytotoxic reactions are caused by antibodies (IgG or IgM) that can destroy normal cells by complement lysis or by antibody-dependent cellular cytotoxicity (ADCC). Transfusion reactions, pernicious anaemia, Goodpasture's disease and haemolytic disease of the newborn are type II hypersensitivity reactions. In cases of haemolytic disease of the newborn, maternal IgG crosses the placenta and attacks the foetal red blood cells.

Type III Hypersensitivity Reactions

Type III hypersensitivity reactions are mediated by small antigen-antibody complexes that activate complement and other inflammatory systems, attract neutrophils, and contribute to inflammation. Deposition of immune complexes near the site of antigen entry can induce an Arthus reaction, (localised reaction) and serum sickness. In Arthus reaction, there is localised vasculitis due to deposition of immune complexes, typically occurring after booster immunisation with toxoid vaccines such as tetanus toxoid. The small immune complexes are often deposited in small blood vessels in organs, where they cause inflammatory disease, e.g. glomerulonephritis in the kidney or arthritis in the joints. SLE is another example of a disease caused by type III hypersensitivity.

Type IV Hypersensitivity Reactions

These reactions involve delayed type of cell-mediated response to an antigen. T-cells play an important role in this reaction. Antigen activation of sensitised Th1 cells induces release of various cytokines that cause macrophages to accumulate and become activated. The net effect of the activation of macrophages is to release lytic enzymes that cause localised tissue damage. Two types of delayed hypersensitivity reactions are: tuberculin or Mantoux reaction and contact dermatitis.

The Complement System

The complement system comprises a group of approximately 20 soluble serum proteins, many of which exist in inactive forms. It is present in the blood of all normal individuals. It mediates the cell killing effects of innate immunity as well as acquired immunity. Complement cascade activation occurs via three pathways: the classical, alternative, or mannose-lectin pathways, each of which is initiated differently **(Fig. 6.14)**. The classical pathway is activated with the formation of soluble antigen-antibody complexes (immune complexes) or the binding of antibody (IgG or IgM) to antigen on a suitable target, such as a bacterial cell. On the other hand, activation of the alternative and lectin pathways is antibody-independent. The alternative pathway is activated by the action of microbial polysaccharides and aggregated IgA or IgG. The alternative pathway begins with the activation of complement protein C3. The mannose-lectin pathway is activated by lectins. In this pathway, carbohydrate residues on the surface of pathogens activate mannose-binding lectin (produced in the liver). This results
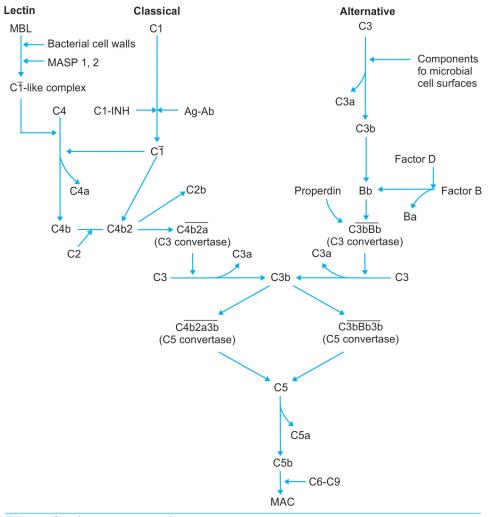
FIG. 6.14: Complement activation pathway
Abbreviations: MASP, mannose associated serine protease; MBL, mannose-binding lectin; MAC, membrane attack complex

in formation of a complex after combination with a protein called MASP (mannose associated serine protease). When the lectin binds to a pathogen containing mannose, the MASP protein complex converts the complement protein C3 to C3a and C3b.

The central component between all the three pathways is C3. C3Bb can become a C3 convertase, which amplifies the system, thus triggering more C3b formation. This is known as the amplification loop and causes amplification of both classical and alternative pathways.

Vaccines

As previously discussed vaccines are biological preparations, which help in proving artificial type of active immunity to the body against specific diseases. Different types of vaccines **(Table 6.16)** are as follows:
❑ Live-attenuated vaccines
❑ Inactivated vaccines
❑ Subunit vaccines
❑ Toxoid vaccines
❑ Conjugate vaccines
❑ DNA vaccines
❑ Recombinant vector vaccines.

Immunisation for Haemophilus Influenzae

Encapsulated strains of the organism *H. influenzae* can cause serious invasive disease, such as pneumonia, meningitis, acute epiglottitis, septic arthritis, cellulitis, etc. particularly in young children. This infection can be associated with sequelae such as deafness, convulsions and intellectual impairment. There are six typeable capsular serotypes (a–f), which are known to cause disease in children. Of these various serotypes, type b (Hib) is the most prevalent strain.

The vaccine in use in the UK is made from capsular polysaccharide that has been extracted from cultures of Hib bacteria. The polysaccharide is conjugated to a protein. In the UK, Hib vaccines are conjugated with either CRM197 (a non-toxic variant of diphtheria toxin) or tetanus toxoid. Conjugation increases the immunogenicity, especially in young children in whom the plain polysaccharide vaccines

TABLE 6.16	Different types of vaccines
Type of vaccine	**Example**
Live-attenuated vaccines Contain live, but attenuated organisms, which lack the ability to be pathogenic but are likely to initiate an immune response)	• MMR (Measles, mumps, rubella) • Chicken pox • Oral polio vaccine (Sabin) • Bacillus Calmette-Guérin (BCG) • Yellow fever (17 D vaccine) • Herpes zoster (shingles) vaccine • Intranasal influenza vaccine (Fluenz®) • Oral typhoid (not used in the UK)
Killed (inactivated) vaccines Considered generally safer than the live vaccines. Since immune response to killed vaccines is generally weaker than to the live vaccines, adjuvants such as aluminium hydroxide are added to the killed vaccines in order to precipitate an improved immune response.	• Hepatitis A • Pertussis • Influenza • Injectable polio vaccine (Salk vaccine) • Rabies virus • Cholera vaccine • Injectable typhoid vaccine (available in the UK)
Toxoid vaccines (contain a toxin or chemical made by the bacteria or virus, which confers immunity)	• Diphtheria vaccine • Tetanus vaccine
Biosynthetic/conjugate vaccines Contains manmade substances that are very similar to pieces of the virus or bacteria.	• The Hib (Haemophilus influenzae type B)
Subunit vaccines These use a specific antigen or purified protein in order to elicit appropriate immune response.	• Hepatitis B vaccine • Typhoid vaccine (Vi-antigen) • Hepatitis C vaccine (ongoing research)
DNA vaccines Genes for the microbe's antigens are introduced into the body which is taken up by some cells. The DNA then instructs those cells to make antigen molecules, thereby invoking the immune response.	• Vaccine against the West Nile Virus
Recombinant vector vaccines An attenuated virus or bacterium is used to introduce the microbial DNA into the body cells, which causes the formation of antigen molecule. This stimulates the body's immune mechanism. The virus or bacteria used as a carrier is known as the vector.	• There is an ongoing research for recombinant vector vaccines against diseases such as HIV, rabies and measles

Source: Immunisation Against Infectious Disease—"The Green Book". Chapter 16 Haemophilis influenzae type B (Hib) (August 2006) http://www.gpnotebook.com

are not immunogenic. The Hib vaccine is given as part of a combined product: diphtheria/tetanus/acellular *pertussis*/inactivated polio vaccine/*Haemophilus influenzae* type b (DTaP/IPV/Hib) vaccine. The vaccine is administered by deep subcutaneous or intramuscular injection. This vaccine is usually associated with an efficacy in the excess of 95%.

Primary immunisation is suggested for all the infants and children. DTaP/IPV/Hib is recommended for all children from 2 months up to 10 years of age. The prescribed amount consists of three doses of a Hib-containing product with an interval of 1 month between each dose. Although one dose of Hib vaccine is effective from 1 year of age, three doses of DTaP/IPV/Hib should be given to children who have either not been immunised or who have not completed a primary course. This also helps in providing complete protection against diphtheria, tetanus, pertussis and polio. If the primary course is interrupted it should be resumed but not repeated, allowing an interval of 1 month between the remaining doses.

Children of 1–10 years of age who have completed a primary course of diphtheria, tetanus, pertussis and polio, but have not received Hib-containing vaccines, should receive a single dose of Hib vaccine. A single reinforcing (booster) dose of Hib is recommended at 12 months for children who have received a complete primary course of three Hib-containing vaccines injections. This booster should be given 1 month before pneumococcal conjugate and MMR vaccines.

Adverse reactions include local swelling in 10%, but this is usually insufficient to contraindicate further doses. Contraindications to immunisation include acute illness (where by the vaccination must be delayed), and those who have had a severe local or general reaction with previous infection.

MMR Vaccination

The MMR vaccine is freeze-dried, and contains live attenuated measles, mumps and rubella viruses. The current MMR 2 (Merck) contains the Edmunston strain of measles, RA27/3 of rubella and Jeryl Lynn strain of mumps. It is administered intramuscularly into the upper arm or anterolateral aspect of thigh. Deep subcutaneous injections are used for patients with bleeding disorders to minimise the risk of bleeding.

Since October 1996, all children except those with valid contraindication have been advised to have two doses of MMR. These are given between 12 months and 15 months, with a second dose before school entry. Following the first dose, malaise, fever, and/or rash occur most commonly about a week after immunisation and lasts for about 2–3 days. Parotid swelling occurs in about 1% of children.

The previous Urabe strain of mumps virus was associated with mumps meningitis, but no cases of meningitis have been reported following the change to Jeryl Lynn vaccine.

Thrombocytopenia and arthropathy may also occur. Side effects of the second dose of MMR are considerably lower.

Absolute Contraindications

There are very few absolute contraindications to MMR vaccinations. These include the following:
❑ Children with allergies to neomycin or kanamycin (very rare)

❑ A severe reaction to previous MMR
❑ Untreated cancer or diseases of the immune system
❑ Children receiving immunosuppressive therapy or high dose steroids.

Relative Contraindications

Reasons to postpone vaccination include the following:
❑ An acute febrile illness
❑ Administration of another live vaccine within 3 weeks of proposed MMR vaccination, or
❑ Administration of immunoglobulins within 3 months of the proposed MMR vaccine
❑ *Allergy to egg*: Administration should be in a supervised, controlled environment (for example, day case in hospital).
❑ Should be avoided in pregnant women for 1 month (because of rubella)

Pregnancy and The Immune System

Changes in the Immune System

Levels of IgG decreases in pregnancy. There are reduced levels of T helper cells in normal pregnancy. IgG is the only immunoglobulin capable of crossing the placenta. Occasionally, maternal IgG may be harmful to the foetus, as in rhesus (Rh) isoimmunisation or maternal immune thrombocytopenic purpura. Maternal IgM does not cross the placenta in humans. IgD is increased in pregnancy.

ABO Incompatibility Disease

In ABO incompatibility, the mother usually has blood group O. The problem arises if the baby has blood groups A, B or AB. Naturally-occurring antibodies to blood groups A and B (predominantly IgG in nature) in the mother may cross the placenta and destroy the foetal red blood cells. ABO incompatibility usually produces mild disease, with jaundice rather than significant anaemia. Phototherapy may be required, but transfusion is rarely needed. It does not worsen from one pregnancy to the next.

Since A and B antigens are widely expressed in a variety of tissues besides red blood cells, only a small portion of antibodies crossing the placenta is available to bind to foetal RBCs. Additionally, foetal RBCs appear to have reduced surface expression of A or B antigen, resulting in fewer reactive sites. This is likely to result in a reduced incidence of significant haemolysis in affected neonates.

Rh Incompatibility Disease

Rh incompatibility disease **(Fig. 6.15)** can occur due to difference in Rh blood groups between the mother and foetus. Rh status is genetically transmitted and hence the development of the various antibodies is genetically

Destruction of fetal red cells resulting in anaemia, heart failure and death. Build-up of bilirubin resulting from red cell destruction leads to jaundice and brain damage in the baby

A couple (where father is Rh positive and mother is Rh negative) conceive a Rh-positive baby

Rh isoimmunisation is not a problem at the time of first pregnancy. At the time of delivery and rarely in the antenatal period Rh-positive antigens move into the maternal blood through placental circulation

These Rh-positive antigens stimulate the formation of anti-Rh antibodies in the maternal circulation

In the subsequent pregnancies with Rh-positive baby, these anti-Rh antibodies can reach the fetus via the placenta. These antibodies can destroy the Rh-positive RBCs, resulting in development of fetal complications like anaemia

Antibody

FIG. 6.15: Pathogenesis of Rh isoimmunisation

determined by simple Mendelian inheritance. Rh disease of the newborn as a result of Rh incompatibility is nowadays relatively rare. This is due to the use of anti-D immunoglobulins. Moreover, the women nowadays have fewer children. Having fewer children also helps to reduce the occurrence of the Rh disease because each pregnancy is likely to act as a potentially sensitising event. As a result, ABO incompatibility is nowadays more common cause of neonatal jaundice than Rh disease.

Rhesus incompatibility may develop when a woman with Rh negative blood marries a man with Rh positive blood and conceives a foetus with Rh positive blood group (who has inherited the Rh factor gene from the father). Rh positive foetal RBCs from the foetus leak across the placenta and enter the woman's circulation. Throughout the pregnancy, small amounts of foetal blood can enter the maternal circulation [fetomaternal haemorrhage (FMH)], with the greatest transfer occurring at the time of delivery or during the third trimester. This transfer stimulates maternal antibody production against the Rh factor, which is called isoimmunisation. The process of sensitisation has no adverse health effects for the mother. Rh incompatibility is not a factor in a first pregnancy, because fewer foetal blood cells reach the mother's bloodstream until delivery. The antibodies that form after delivery cannot affect the first child. However, if the mother is exposed to the Rh D antigens during subsequent pregnancies, the immune response is quicker and much greater. The anti-D antibodies produced by the mother can cross the placenta and bind to Rh D antigen on the surface of foetal RBCs, causing lysis of the foetal RBCs, resulting in development of haemolytic anaemia.

IgM is the initial response to an antigenic stimulus, with IgG replacing IgM in time. If the mother has IgM antibodies, this indicates recent infection. Presence of IgG suggests that the woman has gained some degree of immunity from earlier exposure or immunisation.

The Kleihauer Betke test is used to quantify foeto-maternal transfusion. It is particularly useful at the time of delivery, but should also be considered in other situations associated with high chances of foeto-maternal leak (e.g. significant trauma to the maternal abdomen. The basis of this test is that foetal haemoglobin is more resistant than the adult haemoglobin to acid and alkali. A film is made on a slide from the blood obtained from the mother. The cells are treated with an acid solution (acid elution) and then

FIGS 6.16A AND B: Coomb's test. (A) Indirect Coomb's test; (B) Direct Coomb's test

stained. This disrupts the maternal cells. The foetal cells retain their haemoglobin, thereby taking up the stain. The maternal cells do not take up the stain, thereby appearing as "ghost" cells. The ratio of foetal to maternal cells is used for calculating the amount of foetal blood, which has been transfused to the mother. While small amounts of feto-maternal leaks occur in most patients, huge transfusions of up to 50 mL may occur in about 2% patients. This has implications for the baby, which has lost a significant amount of its circulating volume. It also implies that the mother requires large and probably repeated doses of anti-D. Routine administration of anti-D during pregnancy is recommended and has now become a routine.

The Coomb's test uses antibodies that can help detect Rhesus antibodies.

The direct Coomb's test is used for detecting the maternal antibodies attached to the foetal cells. If such antibodies are present, the Coomb's test will show them. Indirect Coomb's test (ICT), on the other hand, aims at measuring the presence of antibodies, which are present unbound in the maternal serum. In the ICT, RhD-positive RBCs are incubated with maternal serum. Any anti-RhD antibody present in the serum will adhere to the RBCs. The RBCs are then washed and suspended in serum containing antihuman globulin (Coomb's serum). Red cells coated with maternal anti-RhD will be agglutinated by the antihuman globulin (positive ICT, **Fig. 6.16A**). The direct Coomb's test, on the other hand, aims at detecting the antibodies that are bound to the surface of RBCs **(Fig. 6.16B)**. This test is done after birth to detect the presence of maternal antibody on the neonatal RBCs. In the direct Coomb's test, the infant's RBCs are placed in Coomb's serum. If the cells are agglutinated, this indicates the presence of maternal antibody. Direct Coomb's test is positive in cases of haemolytic disease of the newborn. A measure of the amount of antibody can be obtained by running the test with different dilutions of maternal serum. This is the reason for expressing the amounts of antibody as ratios: 1/16, 1/32, etc. These reflect the dilution at which it can still be detected.

Direct Coomb's test is also positive in cases of cold antibody autoimmune haemolysis (e.g. atypical pneumonia due to mycoplasma, infectious mononucleosis, paroxysmal cold haemoglobinuria, etc.) and warm antibody autoimmune haemolysis (e.g. SLE). Direct Coomb's test is also positive in cases where methyldopa has been administered because methyldopa causes drug-induced, immune-mediated haemolysis. Usage of cephalosporins can also be associated with positive Coomb's test.

Choose the Single Best Answer (SBA)

Q 1. **Which of the following micro-organisms does not cause latent infection?**
- A. Cytomegalovirus (CMV)
- B. Chlamydia trachomatis
- C. Hepatitis A
- D. Mycobacterium tuberculosis
- E. Varicella zoster virus

Q 2. **Which of the following micro-organisms is responsible for causing chronic osteomyelitis after implant surgery?**
- A. *Streptococcus pyogenes*
- B. *Staphylococcus aureus*
- C. *Haemophilus influenzae*
- D. *Escherichia coli*

Q 3. **Which of the following micro-organisms is responsible for causing pseudomembranous colitis?**
- A. *Streptococcus pyogenes*
- B. *Clostridium difficile*
- C. *Haemophilus influenzae*
- D. *Escherichia coli*
- E. *Clostridium perfringens*

Q 4. **Which of the following is true regarding Neisseria?**
- A. *N. meningitidis* is a Gram-positive cocci
- B. Infection with *N. gonorrhoeae* may cause suppurative urethritis in males
- C. *N. gonorrhoeae* is a communal of the genital tract
- D. *N. gonorrhoeae* thrives in conditions with low levels of carbon dioxide
- E. Species cannot be cultured on chocolate agar

Q 5. **Which of the following micro-organisms is responsible for causing gas gangrene?**
- A. *Streptococcus pyogenes*
- B. *Clostridium difficile*
- C. *Haemophilus influenzae*
- D. *Escherichia coli*
- E. *Clostridium perfringens*

Q 6. **Which of the following diseases may not have animals as their source?**
- A. Brucellosis
- B. Cholera
- C. Listeriosis
- D. Tuberculosis
- E. Leptospirosis

1. C   2. B   3. B   4. B   5. E   6. B

Q 7. Which of the following is true regarding exotoxins?
A. Are derived from Gram-negative bacteria
B. Are more toxic than endotoxins
C. Are neutralised by their homologous antitoxin
D. Can be converted to a toxoid
E. All the above

Q 8. Which of the following statement is true regarding bacterial endotoxins?
A. Are components of the cell wall in Gram-negative bacteria
B. Are lipopolysaccharides
C. Induce fever
D. All the above
E. None of the above

Q 9. Which of the following is not true concerning endotoxins?
A. Activate components of the coagulation cascade
B. Are heat stable
C. Are produced by Gram negative bacteria
D. Can be converted to toxoid
E. Stimulate the production of tumour necrosis factor (TNF-α)

Q 10. Which of the following is not true regarding opsonisation?
A. Enhances phagocytosis
B. Is mediated by certain complement components
C. Does not involve immunoglobulins
D. May utilise fibronectin
E. Is not MHC restricted

Q 11. Which of the following is true regarding Mycobacterium tuberculosis (MBT)?
A. It does not form spores
B. It is a motile bacillus
C. Produces endotoxins
D. Provokes humoral immunity only
E. Is the only *Mycobacterium* which is acid-fast

Q 12. True statement regarding Streptococci is:
A. Are penicillin-resistant bacteria
B. Are oval in shape
C. Are arranged in a pair of two
D. Produce endotoxin
E. Does not produce hyaluronidase

Q 13. Which of the following is true regarding beta-haemolytic streptococci?
A. Cause localised infections
B. Are classified according to Lancefield groups, based on cell-wall carbohydrate antigen
C. Group A haemolytic streptococci are found in the throat in 25 percent of normal adults and children
D. They cause impetigo and furuncles
E. Group B streptococcus is the most common pathogenic organism

Q 14. True regarding *Staphylococcus aureus*:
A. Is arranged in chains
B. Is motile
C. Is a strict anaerobe
D. Is a spore-forming bacterium
E. Is a penicillin-resistant bacterium

Q 15. Which of the following is true regarding Listeria monocytogenes?
A. Is a non-motile bacterium
B. Is a Gram-positive coccus
C. Can be found in soft cheese and milk
D. Is sensitive to alkaline conditions
E. It gets destroyed by freezing

Q 16. Which of the following is true regarding gonococcus?
A. Is a Gram-positive diplococcus bacillus
B. Is treated with erythromycin
C. Can penetrate the stratified squamous epithelium
D. It is insensitive to cold
E. In the male it may cause an acute suppurative urethritis and proctitis

Q 17. Which of the following is true regarding Clostridium tetani?
A. Causes gas gangrene
B. Has a sub-terminal spore
C. Is an obligatory anaerobe
D. Produces an endotoxin
E. It is non-motile

Q 18. The germination of tetanus spores in a wound is inhibited by which of the following?
A. Injection of anti-toxin
B. Injection of toxoid
C. Tissue trauma
D. Reduced blood supply
E. Decreased oxygen supply

Q 19. Which of the following is true regarding *Clostridium difficile* infection causing diarrhoea?
A. Successfully treated with intravenous vancomycin
B. More likely to develop with benzylpenicillin than third generation cephalosporins
C. Transmissible from person to person
D. Diagnosed by the isolation of Clostridium difficile from stool cultures
E. More likely to be community acquired than hospital-acquired

Q 20. *Staphylococcus aureus* is not sensitive to which of the following?
A. Cefuroxime
B. Clindamycin
C. Flucloxacillin
D. Penicillin
E. Vancomycin

7. E   8. D   9. D   10. C   11. A   12. B   13. B   14. E   15. C   16. E   17. C   18. B   19. C   20. D

Q 21. Which of the following is not true regarding methi-cillin-resistant *Staphylococcus aureus* (MRSA)?
- A. It may cause nosocomial pneumonia
- B. It may be a cause of toxic shock syndrome
- C. It is an invasive organism when compared to methicillin sensitive strains of *Staphylococcus aureus*
- D. Mostly responds well to vancomycin therapy
- E. Resistance to methicillin is due to a plasmid

Q 22. Which of the following is true concerning *Actinomyces israelii*?
- A. *Actinomyces israelii* is aerobic
- B. Is treated with tetracycline
- C. Typically grows slowly
- D. Is a commensal in the vagina
- E. Is a *Rickettsia*

Q 23. Which of the following antibiotics is usually effective against *Pseudomonas aeruginosa*?
- A. Amoxicillin
- B. Carbenicillin
- C. Cephradine
- D. Trimethoprim
- E. Tetracycline

Q 24. Which of the following is not true concerning *Chlamydia trachomatis*?
- A. Causes lymphogranuloma venereum
- B. Causes non-specific urethritis
- C. Is a precipitant of Reiter's syndrome
- D. Treatment of choice is penicillin V
- E. Is gram negative bacteria

Q 25. Which of the following diseases are caused by Spirochaetes?
- A. Bilharzia
- B. Lymphogranuloma venereum
- C. Weil's disease
- D. Malaria
- E. Chancroid

Q 26. *Treponema pallidum* immobilisation (TPI) test is positive in which of the following disease?
- A. Chancroid
- B. Infectious mononucleosis
- C. Lyme disease (borreliosis)
- D. Malaria
- E. Yaws

Q 27. Which of the following statement is not true regarding *Treponema pallidum* (syphilis)?
- A. It only affects humans
- B. Its incubation period is 2–3 weeks
- C. May only be visualised by dark-ground illumination
- D. Is sensitive to water and drying
- E. Crosses the placenta after 16 weeks' gestation

Q 28. Which of the following statement regarding Chlamydia is true?
- A. Is motile.
- B. Is Gram-positive
- C. Is the most common sexually transmitted disease
- D. Causes chancre formation
- E. Causes chancroid formation

Q 29. Which of the following is not true regarding *Bacteroides*?
- A. Anaerobic
- B. Gram-positive
- C. Non-spore-forming
- D. Sensitive to metronidazole
- E. A cause of bacterial vaginosis

Q 30. Which of the following does not produce food poisoning?
- A. *Clostridium welchii*
- B. Streptococci
- C. Campylobacter
- D. *Clostridium botulinum*
- E. *Bacillus cereus*

Q 31. Regarding *Leptospira* spp, which of the following is not true?
- A. Are Gram negative bacilli
- B. Infection is associated with jaundice
- C. Infection may be acquired through skin contact
- D. Infection produces a positive Wasserman reaction
- E. Is transmitted in the urine of rats

Q 32. Which of the following is true regarding *Mycoplasma pneumoniae*?
- A. Predominantly causes infection in the elderly
- B. Can be grown on a cell-free medium
- C. Infection is associated with a polymorphonuclear leucocytosis
- D. White cell count is reduced
- E. Infection is associated with the development of agglutinins to a haemolytic *Streptococcus*

Q 33. Which of the following is not true regarding *Campylobacter jejuni*?
- A. Attack rates are highest in young adults and children
- B. Infections are treated with ciprofloxacin
- C. It is a recognised pathogen in domestic animals
- D. It is readily isolated in stool culture
- E. It causes colitis

Q 34. Which of the following are examples of DNA viruses?
- A. Enteroviruses
- B. Epstein-Barr virus
- C. Influenza virus
- D. All of the above
- E. None of the above

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21. C   22. C   23. B   24. D   25. C   26. E   27. B   28. C   29. B   30. B   31. A   32. B   33. D   34. B

Q 35. Which of the following hepatitis viruses is a RNA virus?
- A. EBV
- B. HBV
- C. HAV
- D. None of the above
- E. All of the above

Q 36. Which of the following diseases are caused by herpes simplex virus?
- A. Cervical warts
- B. Acute gingivostomatitis
- C. Shingles
- D. All the above
- E. None of the above

Q 37. Which of the following group of viruses does the herpes group not include?
- A. Cytomegalovirus
- B. Epstein-Barr virus
- C. Papilloma virus
- D. Herpes simplex
- E. Varicella-zoster virus

Q 38. Which of the following is true regarding the human papilloma virus?
- A. It is a small RNA virus
- B. It causes genital vesicles
- C. It readily crosses the placenta
- D. Effective vaccines now exist
- E. It has a weak association with carcinoma cervix

Q 39. Which of the following is not true regarding Cyto-megalovirus?
- A. Causes haemolytic anaemia in the neonate
- B. Is a cause of foetal cerebral calcification
- C. Is an adenovirus
- D. May be cultured readily in cell-free media
- E. May be transmitted in saliva

Q 40. Which of the following virus may exhibit oncogenic properties in humans?
- A. Enteroviruses
- B. Hepatitis B virus
- C. Papovavirus
- D. Rabies virus
- E. Rubella virus

Q 41. Epstein-Barr (EB) virus is not associated with which of the following?
- A. Burkitt's lymphoma
- B. Cervical neoplasia
- C. Nasopharyngeal carcinoma
- D. Pharyngitis
- E. Autoimmune haemolytic anaemia

Q. 42. Which of the following hepatitis viruses is RNA virus?
- A. HAV
- B. HCV
- C. HDV
- D. All the above
- E. None of the above

Q 43. Which of the following are the mediators of the febrile response?
- A. Prostaglandins
- B. Interferon
- C. Monocyte
- D. None of the above
- E. All of the above

Q 44. Proteolytic enzymes are released by which of the following organisms?
- A. *Clostridium perfringens*
- B. *Mycobacterium tuberculosis*
- C. *Neisseria meningitides*
- D. *Salmonella typhi*
- E. *Escherichia coli*

Q 45. Congenital abnormalities are not associated with which of the following maternal infections?
- A. Hepatitis B
- B. Group B streptococcus
- C. Parvovirus
- D. All the above
- E. None of the above

Q 46. Which of the following is a Gram-negative bacteria?
- A. *Lactobacillus acidophilus*
- B. *Campylobacter jejuni*
- C. *Clostridium difficile*
- D. *Listeria monocytogenes*
- E. *Staphylococcus aureus*

Q 47. Which of the following is true regarding immuno-globulins?
- A. IgMs can cross the placenta to the foetus
- B. Immunoglobulins are secreted from T-lymphocytes
- C. IgG constitutes approximately 25% of all immuno-globulins in a healthy individual
- D. The molecular structure of IgG is a Y shape
- E. Secretion of immunoglobulins does not require response of a specific antigen

Q 48. Which of the following is true regarding immuno-globulin E (IgE)?
- A. Present in normal serum in a concentration similar to that of IgG
- B. Attached to mast cells in the skin
- C. Involved in type II hypersensitivity mechanisms
- D. Transmitted across the normal placenta
- E. Found in low concentrations in the serum of patients with atopic eczema

35. C   36. B   37. C   38. D   39. C   40. B   41. B   42. D   43. E   44. A   45. D   46. B   47. D   48. B

Q 49. Which of the following is true regarding immuno-globulins?

A. Antiviral activity is provided mainly by IgA antibody
B. Immunoglobulins G are produced by T helper cells
C. All immunoglobulins are gamma-globulins
D. Immunoglobulin G (IgG) chains are coded for by adjacent genes on the same chromosome
E. Immunoglobulin A (IgA) is a cryoglobulin

Q 50. Which of the following statements regarding immunoglobulin G is correct?

A. Immunoglobulin G of foetal origin is secreted after birth
B. Immunoglobulin G is produced in similar amounts in all individuals exposed to the same stimulus
C. Immunoglobulin G crosses the placenta
D. Immunoglobulin G is produced in the primary immune response
E. Immunoglobulin formation requires complement activation

Q 51. Which of the following is true regarding the functioning of immune system during pregnancy?

A. Levels of IgG decreases in pregnancy
B. Helper T lymphocytes are increased in number in normal pregnancy
C. Maternal IgG has no harmful effect on the foetus
D. Foetal IgM is derived from maternal blood
E. IgD is decreased in pregnancy

Q 52. Which of the following is not true regarding the immune system?

A. Interleukin-1 is derived exclusively from monocytes
B. Helper T cells are essential in order for B cells to produce full activation and antibody formation
C. The genes of the major histocompatibility complex (MHC) are located on the short arm of human chromosome 6
D. Lymphokines are produced by lymphocytes
E. Most of the receptors on T cells are made up of alpha and beta polypeptides units

Q 53. Which of the following statements is true regarding the complement system?

A. The central component between the two pathways is C5
B. The complement system consists of a set of insoluble serum proteins
C. It mediates the cell killing effects of innate immunity as well as acquired immunity
D. The alternative pathway is activated by an antigen–antibody interaction
E. C3Bb can become a C3 convertase, which inhibits the system

Q 54. Live-attenuated vaccination is used against which of the following diseases?

A. Hepatitis B
B. Influenza
C. Pneumococcus
D. Tetanus
E. Tuberculosis

Q 55. Which of the following statements is true regarding Hib immunisation?

A. It contains the polysaccharide capsules of three strains of *Haemophilus influenzae*
B. Booster doses are recommended at 2 years of age in the UK
C. It is not protective against Hib periorbital cellulitis
D. It is protective in only about 75% of patients
E. Three doses should routinely be given at 2, 3 and 4 months

Q 56. Regarding MMR immunisation, which of the following statement is true?

A. The mumps component consists of homogenates of killed vaccine virus
B. The Jeryl Lynn strain of mumps has reduced the incidence of mumps encephalitis.
C. Egg allergy is an absolute contraindication
D. A second dose is recommended after starting school
E. A maculopapular rash may rarely occur within 10 days

Q 57. Which of the following is not correct regarding rubella?

A. Can be prevented by vaccination in over 80% of individuals
B. Is an indication for termination if it occurs in the first two months of pregnancy
C. Is more frequently associated with palpable splenomegaly than infectious mononucleosis
D. May be complicated by polyarthralgia
E. Typically has an incubation period of 7–10 days

Q 58. Which of the following is true regarding the Coombs' test?

A. The direct test detects maternal IgM on foetal cells
B. It is used in the investigation of thrombocytopenia
C. It is positive in the baby with jaundice due to spherocytosis
D. The direct test is used to detect antibodies in the maternal serum during pregnancy
E. The direct test uses anti-IgG serum

Q 59. Which of the following concerning the effectiveness of a vaccine is correct?

A. Is purely due to antibody response in tuberculosis
B. Is a purely cell-mediated response in streptococcal pneumonia
C. Is due to high levels of serum antibody in mucosal protection against polio
D. Is due to activation of cytotoxic cells in hepatitis
E. Can be measured by checking circulating IgG titres in rubella

49. A   50. C   51. A   52. A   53. C   54. E   55. E   56. B   57. E   58. E   59. E

Q. 60. Which of the following is not correct regarding attenuated vaccines?

A. The virulence has been artificially reduced
B. Adaptation to high temperatures is one of the two methods used to produce live attenuated vaccines
C. They make up the bulk of successful viral vaccines
D. The killed (inactivated) form of the polio vaccine is known as the Salk vaccine.
E. Rubella vaccine is a "live"-attenuated vaccine

Q 61. Which of the following is not true regarding mast cells?

A. An excess of circulating mast cells causes mastocytosis
B. Are basophilic cells involved in inflammatory and immune responses
C. Contain heparin
D. Degranulation releases lytic enzymes and inflammatory mediators from storage granules
E. Cross-linkage of surface IgA molecules by antigen may cause an anaphylactic reaction

Q 62. Which of the following is not true regarding hospital-acquired infections?

A. Contact between individuals is the most likely route of infection
B. Contaminated equipment is a higher infection risk if wet
C. Gram-negative organisms are usually spread by the aerosol route
D. Sterilisation may leave some bacterial spores intact
E. Theatre air systems generate a positive pressure relative to the surroundings.

Q 63. Which of the following is not true regarding sterilisation?

A. Ethylene oxide should only be used when heat sterilisation of an item is not possible
B. Flash autoclaving at 147°C and 40 lb/square inch is no longer the preferred method of sterilisation by steam
C. Sterilisation by ethylene oxide has a broad-spectrum static action against bacteria, spores and viruses
D. Hot air sterilisation is the preferred method to treat surgical instruments with fine cutting edges
E. Unwrapped instruments may be sterilised in theatre using a portable steam steriliser

Q 64. Which of the following factor affects the performance of a disinfectant?

A. Concentration of disinfectant
B. Number of organisms present
C. pH
D. None of the above
E. All the above

Q 65. Group B β-haemolytic streptococcus infection can cause which of the following:

A. Chorioamnionitis
B. Acute pharyngitis
C. Glomerulonephritis
D. Necrotising fasciitis
E. Toxic shock syndrome

Q 66. Which of the following is RNA-containing virus?

A. Varicella zoster
B. Smallpox
C. Papillomavirus
D. Hepatitis B
E. Mumps

Q 67. Which of the following is true regarding varicella zoster?

A. Distribution of lesions is myotomal
B. The use of antiviral agents helps prevent infection
C. It is an RNA virus
D. Maternal exposure at term infers minimal risk to the foetus
E. Varicella zoster immunoglobulin (VZV-IgG) should be given to exposed pregnant women who are antibody negative

Q 68. Which of the following statement is true regarding the use of various interventions for reducing the risk of infections at the surgical site?

A. There is no confirming evidence that the use of prophylactic antibiotics reduce the post-operative incidence of infection at the surgical site
B. Hair removal should be performed with clippers rather than a razor
C. Antibiotics should be given intravenously no more than 60 minutes prior to surgery
D. Blood sugar levels must be kept below 12 mmol/L in patients with diabetes
E. Use of prophylactic antibiotics reduces the risk of infection by *Clostridium difficile*

Q 69. Which of the following is the causative agent of Kaposi's sarcoma?

A. HIV
B. Human herpes virus 4
C. Human herpes virus 8
D. None of the above
E. All of the above

Q 70. A 25-year-old lady with 16 completed weeks of gestation has presented to the antenatal clinic with the complaints of a maculopapular rash and coryzal symptoms. Her serology reports are as follows:

Rubella IgG: positive; Rubella IgM: negative; Parvovirus B19 IgG: negative; Parvovirus B19 IgM: positive

What is the most likely diagnosis in this case?

A. Non-immunity to parvovirus B19
B. Non-immunity to rubella
C. Recent infection with rubella
D. Recent infection with parvovirus B19
E. None of the above

60. B   61. E   62. D   63. C   64. E   65. A   66. E   67. E   68. B   69. C   70. D

Q 71. Which of the following is true regarding the vancomycin-resistant enterococci?

A. Cause resistant infective diarrhoea
B. Produce an enzyme that inactivates vancomycin
C. May be found in healthy community volunteers not recently hospitalised
D. High-dose ampicillin is the treatment of choice
E. Are commonly vancomycin-dependent

Q 72. Which of the following is not true regarding *Escherichia coli*?

A. Characteristically produces a malodourous infection
B. Grows anaerobically
C. Is a Gram-negative rod
D. Most strains are pathogenic
E. Produces an enterotoxin

Q 73. Regarding *Escherichia coli* 0157/H7, which of the following is true?

A. Can be prevented from causing clinical illness by vaccination
B. Is a bowel commensal
C. Is an important cause of cholera-like illness
D. All the above
E. None of the above

Q 74. Which of the following is not true regarding an acute allergic reaction?

A. It may be triggered by acute complement activation
B. T-helper cells are involved
C. The gene for allergy is located on chromosome 12
D. There is an increase in bradykinins
E. There is an increase in the products of the 5-lipoxygenase pathway

Q 75. Which of the following is not true regarding *Toxoplasma gondii*?

A. It is a cause of congenital hydrocephalus
B. It is an obligate intracellular parasite of the Apicomplexa family
C. It is identified by Gram staining
D. Is transmitted by ingestion of raw meat
E. Proliferates in the central nervous system

Q 76. Which of the following predispose to microbial invasion?

A. Ciliary dyskinesia
B. Cystic fibrosis
C. Neutrophil deficiency
D. All the above
E. None of the above

Q 77. Which of the following is not a notifiable disease?

A. Rubella
B. Chicken pox
C. Food poisoning
D. Measles
E. Meningococcal meningitis

Q 78. Which of the following are not true regarding blood cultures?

A. Are best obtained when the patient is febrile or complains of chills
B. Are unreliable if the patient has already been commenced on antibiotics
C. Are usually negative
D. Grow and identify organisms within 24 hours
E. Require two culture (aerobic and anaerobic) specimens obtained from at least two different sites

Q 79. Which of the following is not a recognised virulence factors in bacteria?

A. Beta lactamases
B. Gonococcal pili
C. IgA-proteases
D. Streptococcal M protein
E. The capsular polysaccharides in *Haemophilus influenzae*

Q 80. Which of the following pairs of infections and complications is not correct?

A. Deafness: *Toxoplasma gondii*
B. Cardiac anomalies: Coxsackie B virus
C. Hydrocephalus: *Toxoplasma gondii*
D. Cataracts: Rubella
E. Encephalitis: herpes simplex type I

Q 81. Which of the following micro-organisms is generally sensitive to benzylpenicillin?

A. *Streptococcus viridans*
B. *Mycoplasma pneumoniae*
C. *Cryptococcus neoformans*
D. None of the above
E. All of the above

Q 82. Concerning cryptosporidiosis in HIV positive individuals, which of the following statement is true?

A. Can produce sclerosing cholangitis with inflammation and ulceration of intra and extra hepatic bile ducts
B. Produces liquid faeces in scanty amounts per day
C. Is effectively eradicated by spiramycin in the majority of cases
D. Is the commonest cause of diarrhoea in these patients
E. Presents with bloody diarrhoea

Q 83. Which of the following mechanisms of microbial resistance is not correctly ascribed?

A. *Enterococcus faecalis* by beta-lactamase production
B. Herpes simplex by mutations of viral thymidine kinase
C. *Staphylococcus aureus* by production of beta-lactamases
D. Pseudomonas aeruginosa by m*utation of specific binding proteins*
E. *Staphylococcus epidermidis* by slime production

71. C   72. A   73. E   74. C   75. C   76. D   77. B   78. D   79. A   80. A   81. A   82. A   83. D

Q 84. Which of the following is the possible means of diagnosis of congenital HIV infection in a neonate born to an infected mother?
A. Attempt detection of viral genome by polymerase chain reactions
B. Test for anti-p24 antibody in infant blood
C. Test for delayed hypersensitivity reactions
D. None of the above
E. All the above

Q 85. Which of the following antibodies is secreted in large amounts in the breast milk?
A. IgA
B. IgD
C. IgE
D. IgG
E. IgM

Q 86. Which of the following is true concerning natural killer (NK) cells?
A. Are a type of T lymphocyte
B. Are predominantly found in lymph nodes
C. Express cell surface CD-3
D. Kill antibody coated cells
E. Are unable to release tumour necrosis factor

Q 87. Which of the following is not true regarding CD4 + T cells?
A. Are cytotoxic for virally infected cells
B. Are restricted in antigen recognition by MHC class II molecules.
C. Release cytokines in response to antigen stimulation
D. Are a major target cell for infection with HIV
E. Are involved in delayed type hypersensitivity

Q 88. Which of the following is most important in the adaptive immune system?
A. Cytotoxic T cells
B. Helper T cells
C. Natural killer cells
D. Macrophages
E. Neutrophils

Q 89. Which of the following are not at a high risk of acquiring HIV infection?
A. Babies of HIV-positive fathers and HIV-negative mothers
B. Babies of HIV-positive mothers and HIV-negative fathers
C. Heterosexual IV drug addicts
D. Homosexual IV drug addicts
E. Homosexual males who are promiscuous

Q 90. Which of the following statement is true regarding B lymphocytes?
A. Characterised by rosette formation when mixed with sheep red cells
B. Normally the major type of circulating lymphocyte
C. The predominant cell type in the paracortical region of the lymph node
D. Produced in the lymph nodes
E. Associated with the development of humoral immunity

Q 91. Which of the following is not true regarding T lymphocytes?
A. Have an important role in contact dermatitis
B. Have an important role in secondary antibody response
C. Have surface bound immunoglobulin
D. Possess CD4 receptors
E. Require the thymus for differentiation

Q 92. Which of the following is/are true regarding antibodies?
A. Are formed in the foetus before 12 weeks of intrauterine life
B. Have an average molecular weight of around 10,000 daltons
C. Are produced at a greater rate after a first, than after a second, exposure to an antigen 6 weeks later
D. Are absent from the blood in early foetal life
E. They are produced by the T lymphocytes

Q 93. What is the common step to all complement activation pathways?
A. Activation of C1 to antibody-antigen complexes
B. Cleavage of C3 into C3a and C3b
C. Formation of mannose-binding lectin complex
D. Formation of the IgG antibody-antigen complex
E. Formation of IgM antibody-antigen complex

Q 94. Which of the following is true in patients with the acquired immune deficiency syndrome (AIDS)?
A. Neutrophils are more affected than lymphocytes
B. Total white cell count is a better indicator of progression than any subset of white cells
C. Host DNA is incorporated into the human immunodeficiency (HIV) virus
D. Occurrence in infancy results from inheritance rather than transmission of infection
E. There is an increased risk of malignant tumours

Q 95. Which of the following does not reliably inactivate HIV?
A. Chlorhexidine
B. Glutaraldehyde
C. Hypochlorites
D. The autoclave
E. The hot-air oven

Q 96. Which of the following is an example of type III hypersensitivity reaction?
A. Autoimmune haemolytic anaemia
B. Multiple sclerosis
C. Systemic lupus erythematosus
D. Goodpasture's disease
E. Contact dermatitis

84. A   85. A   86. D   87. A   88. B   89. A   90. E   91. C   92. D   93. B   94. E   95. A   96. C

Q 97. **Which of the following vaccines must not be administered to HIV positive patients?**

A. BCG
B. Havrix (hepatitis A vaccine)
C. Hib vaccine
D. Rabies virus
E. Cholera vaccine

Q 98. **In a 20-year-old pregnant woman, acute rubella infection is diagnosed at 20 weeks of gestation. What is the most likely foetal abnormality which is likely to occur as a result of this acute infection?**

A. Cerebral palsy
B. Sensorineural hearing loss
C. Microcephaly
D. Failure to thrive
E. Limb hypoplasia

Q 99. **Which of the following conditions should an aetiological factor satisfy before being considered to be causally related to a disease?**

A. Elimination of the factor decreases the risk of the disease
B. The factor is found in all cases with the disease
C. The factor is not found among persons without the disease
D. Exposure to the factor is not required for the development of the disease
E. The factor is found more frequently among the non-diseased than the diseased

Q 100. **Which of the following is true regarding human immunodeficiency virus?**

A. Decreases the risk of opportunistic infection
B. Induces a rise in CD4 lymphocytes, monocytes and antigen-presenting cells
C. Is a single stranded DNA retrovirus
D. Patients can be infective prior to seroconversion illness at about three months
E. The median survival with AIDS is greater than 10 years

Q 101. **Which of the following is not a clinical manifestation of HIV seroconversion?**

A. Aseptic meningitis
B. Dementia
C. Infectious mononucleosis-like disease
D. No symptoms
E. Lethargy

Q 102. **Which of the following is not true regarding *Pneumocystis carinii* pneumonia (PCP)?**

A. Can be successfully prevented with nebulised inhaled pentamidine
B. May be present despite a clear chest x ray
C. Is the commonest presenting feature in European AIDS patients
D. Occurs in non-HIV infected individuals

E. Is the commonest presenting feature in Ugandan AIDS patients

Q 103. **Which of the following is true concerning secondary lymphoid follicles?**

A. The cells of the follicle centre are exclusively B cells
B. Immunoblasts are numerous
C. A mantle zone surrounds the follicle centre
D. Centrocytes result from mitotic division of centroblasts
E. Antigen presenting cells are rarely present

Q 104. **Concerning a hepatitis E infection, which of the following is true?**

A. CT scan of the liver with contrast shows diagnostic appearances
B. It can be transmitted with hepatitis B
C. It is a recognised cause of chronic liver disease
D. The incidence of chronic liver disease is reduced by administration of alpha interferon
E. It does not result in a carrier state

Q 105. **Which of the following conditions is not associated with a positive Direct Coomb's test?**

A. Administration of cephalosporins
B. Administration of cyclosporin
C. Haemolytic disease of the newborn
D. *Mycoplasma pneumonia*
E. Administration of methyldopa

Q 106. **Which of the following is not true regarding listeria infection in pregnancy?**

A. *Listeria monocytogenes* can flourish at normal refrigerator temperatures
B. It is associated with premature labour
C. It occurs in every one pregnancy out of in 1,000 in the UK
D. It is associated with eating soft cheeses, cook-chilled meals and stored foods such as coleslaw
E. This infection may be suspected if meconium is present at gestations of <34 weeks.

Q 107. **Intrauterine infection may result in which of the following?**

A. Mental handicap
B. Prematurity
C. Growth failure
D. Cerebral palsy
E. All the above

Q 108. **Which of the following is correct regarding genital herpes infection?**

A. Can be responsible for pre-term delivery
B. Is always symptomatic
C. Is not caused by herpes simplex virus (HSV) type 2
D. It may be caused by herpes simplex virus type 1
E. May produce spontaneous abortion

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97. A   98. B   99. A   100. D   101. B   102. E   103. C   104. E   105. B   106. C   107. E   108. D

Q 109. **Which of the following is correct regarding hepatitis C in pregnancy?**

A. It is most often transmitted through sexual route

B. Coexisting HIV infection increases the risk

C. Breastfeeding should be discouraged to reduce vertical transmission

D. Vertical transmission occurs in 50%

E. Caesarean section is recommended to reduce vertical transmission

Q 110. **Which of the following is not a well-recognised clinical feature of rubella infection in the first trimester?**

A. Cataract

B. Hepatosplenomegaly

C. Large anterior fontanelle

D. Low birth weight

E. Purpura

Q 111. **Which of the following is not a recognised feature of the primary antiphospholipid syndrome (APS)?**

A. Arterial thrombosis

B. Leucopenia

C. Recurrent foetal loss

D. Ulcerations of the leg

E. Impaired foetal growth

Q 112. **Which of the following statement regarding the effect of systemic lupus erythematosus (SLE) on the neonate is correct?**

A. Neonatal SLE usually results from passively acquired maternal anti-Ro antigens

B. The most common foetal condition is congenital heart block

C. Congenital heart block occurs due to maternal autoantibodies causing reversible damage to the foetal cardiac conducting system

D. Two-thirds of surviving neonates with heart block will require a pacemaker

E. High-dose corticosteroids significantly improve foetal outcome

Q 113. **Which of the following is not true regarding HIV-infected woman?**

A. Are at an increased risk of cervical dysplasia

B. Foetal transmission is more likely to occur in women with recent infection

C. Should not breast feed

D. Have no significant increase in pregnancy complications

E. Have a 50% chance of transmitting the infection to the foetus in utero

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109. B   110. C   111. B   112. D   113. E

Embryology

Cell Division

There are two types of cell division, mitosis and meiosis.

Mitosis

The stages of mitosis are shown in **Figure 7.1**. In this type of cell division, the centromere splits into two parts so that the number of chromosomes in each daughter cell remains the same, i.e. 46 chromosomes. Mitosis occurs in all eukaryotic somatic cells. Mitosis has four main phases: prophase, metaphase, anaphase and telophase. The phase just prior to prophase is known as interphase. During this phase, the preparation for mitosis occurs. Interphase is not considered as the part of mitosis and comprises of three phases: G_1 phase (first gap), S phase (synthesis) and G_2 phase (second gap). Replication of chromosomes occurs in the S phase.

Prophase
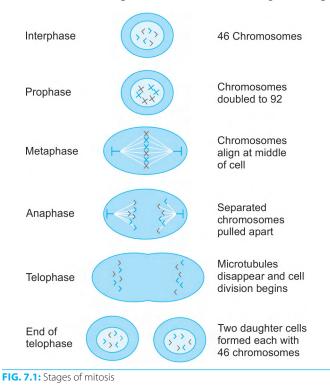
Genetic material is loosely arranged in form of thread like structure called chromatin inside the nucleus. In prophase, the chromatin gets condensed to form organized structure called chromosomes. Since the duplication of the genetic material had already occurred in the S phase, the chromosome, now therefore is made up of two sister chromatids bound together at centromere.

Metaphase

There is a structure called centrosome which is present in the nucleus and contains a pair of centrioles. This acts as the coordinating centre for the cells microtubules. In this phase, the two centrioles separate and move away from each other and begin to occupy the opposite poles of the cell. A number of microtubules are produced, which get organized between the centrioles, giving the appearance of a spindle. The chromosomes then start to occupy the equatorial plane of this mitotic spindle. In the meantime, the nuclear membrane breaks and the nucleoli disappear.

Anaphase

Two main events occur during this phase. During the first, the centromere splits into two halves so that the original chromatids form two independent daughter chromosomes. The second event which occurs is that the chromosomes of each pair moves away along the ray of the spindle, towards the centriole situated on either pole of the cell. Delayed movement of chromatids during mitosis or one homologous chromosome during meiosis can cause anaphase lag.

Interphase	46 Chromosomes
Prophase	Chromosomes doubled to 92
Metaphase	Chromosomes align at middle of cell
Anaphase	Separated chromosomes pulled apart
Telophase	Microtubules disappear and cell division begins
End of telophase	Two daughter cells formed each with 46 chromosomes

FIG. 7.1: Stages of mitosis

Anaphase lag is responsible for causing aneuploidy or dysjunction, leading to an unequal distribution of chromosomes between the daughter cells. The incidence of dysjunction is thought to increase with the increasing maternal age. Since the lagging chromosome does not get incorporated into the nucleus of one daughter cell, that cell is characterized by absence of one chromosome from the normal diploid complement. This is responsible for monosomy or development of cells with 45 chromosomes. Furthermore, due to anaphase lag, an extra chromosome can get incorporated into the nucleus of the other daughter cell, which is characterized by the presence of an extra chromosome in the normal diploid. This results in trisomy or development of cells with 47 chromosomes. On the other hand, division of the chromatids in an abnormal plane results in production of isochromosomes.

Telophase

The daughter nuclei are formed by the appearance of nuclear membrane. The chromosomes elongate, get uncoiled and form chromatin. The nucleoli appear again. The centriole also reduplicates, becoming two in number. Formation of two daughter cells containing a diploid number of chromosomes is thus complete.

Meiosis

Various stages of meiosis are described in **Figure 7.2**. This type of cell division is usually seen at the time of gametogenesis, i.e. formation of sperms in males and ova (eggs) in females. During these two phases, there are two successive nuclear divisions with only one round of DNA replication.

Meiosis occurs in two main phases: meiosis I and meiosis II. Meiosis I is a reductional division. No separation at the centromere takes place. As a result, the number of chromosomes going to each daughter cell is halved (haploid number). On the other hand, meiosis II is an equational division, similar to mitosis, where the sister chromatids are split at the centre, resulting in the production of four haploid cells. In prophase I of meiosis I chromosomal cross over occurs. This refers to the exchange of DNA between the homologous chromosomes and helps in producing new DNA combinations resulting in genetic variation.

To summarise, in one cycle of meiosis, one germ cell gives rise to four daughter cells where some exchange of genetic material has occurred. Each of the haploid cell formed, would contain a haploid number of chromosomes that is 22+X or 22+Y in males and 22+X in females. In female gamete, one primordial germ cell gives rise to four daughter cells each having 22+X chromosomes. However, only one of these four daughter cells would develop into a mature oocyte. The remaining three become the polar bodies, having very little amount of cytoplasm and these ultimately degenerate. On the other hand,

in males all the four daughter cells would develop into mature male gametes (spermatocytes). Of these, two will have the karyotype of 22+X and the other two have the karyotype of 22+Y. Abnormal separation of chromosomes in anaphase I of meiosis I or sister chromatids in anaphase II of meiosis II or anaphase of mitosis is termed as non-dysjunction. This usually occurs as a result of anaphase lag and can cause production of gametes with abnormal number of chromosomes. This may be responsible for producing chromosomal abnormalities such as Down syndrome, Turner's syndrome and Klinefelter's syndrome. In males, meiosis begins at the time of puberty during spermatogenesis. On the other hand, meiosis in the ovaries begins before birth even though it is actually completed just prior to ovulation.

Gametogenesis

Primordial germ cells are the precursors of the germ cells of gonads. These originate in the wall of the yolk sac. These germ cells start migrating towards the developing gonads by 4th week, where they reach by the end of 5th week. Initially, these cells begin to divide by mitosis so that their number increases and they contain a diploid number of chromosomes. Soon thereafter, these cells undergo meiotic

Interphase		46 Chromosomes
Prophase		Chromosomes doubled to 92
Metaphase 1		Chromosomes align at middle of cell
Anaphase 1		Separated chromosomes pulled apart
Telophase 1		Microtubules disappear and cell division begins
Interphase 2		Two daughter cells formed each with 46 chromosomes
Metaphase 2		Microtubules attach to centromeres
Anaphase 2		Chromosomes pulled apart to 23
Telophase 2		Microtubules disappear and cell division begins
Cytokinesis		4 cells formed each with 23 chromosomes

FIG. 7.2: Stages of meiosis

division, a process called as gametogenesis, in which the number of chromosomes gets halved (haploid number).

Spermatogenesis

The process in which spermatogonia get transformed into spermatozoon is known as spermatogenesis. The testes are responsible for producing sperms and male hormones, mainly testosterone. Spermatogenesis is primarily under the control of follicle-stimulating hormone (FSH) and the male hormone, testosterone. FSH is bound to Sertoli cells and stimulates the production of testicular fluid. The luteinising hormone (LH) also has a role in the regulation of spermatogenesis, and influences production of testosterone. The rate of testosterone synthesis and secretion is dependent on LH. In the testes, LH binds to receptors on Leydig cells and stimulates synthesis and secretion of testosterone. Besides producing testosterone, testes are also involved in the production of small amounts of other hormones such as oestrone and oestradiol. However, the major portions of these hormones, oestrone and oestradiol are derived by peripheral aromatisation of androstenedione and testosterone, respectively. Sertoli cells are equivalent to the granulosa cells of the ovary and respond primarily to FSH. On the other hand, Leydig cells have receptors for LH and therefore mainly respond to this hormone. In males, the site of synthesis of various hormones is described in **Table 7.1**. Testosterone is synthesised in the interstitial Leydig cells from where it diffuses into the seminiferous tubules and plays an important role in the facilitation of the process of spermatogenesis, which involves the production of sperms. The process of spermatogenesis takes place in the space between the Sertoli cells, with Leydig cells releasing testosterone to stimulate the process. Testosterone gets converted into dihydrotestosterone in the peripheral tissues with help of an enzyme 5-α reductase. Dihydrotestosterone is more potent than testosterone. In females, testosterone is mainly derived from the conversion of androstenedione. Increased testosterone levels reduce the secretion of sex hormone binding globulins and LH. Besides spermatogenesis, testosterone also induces the development of secondary sexual characteristics in males such as deepening of voice, growth of body hair, penile growth, etc. Androgens also stimulate the activity of sebaceous glands.

Initial process of spermatogenesis involves mitotic division, which is responsible for converting spermatogonia to primary spermatocytes. The spermatozoa then develop through a process of meiosis so that eventually diploid spermatocytes get converted into four haploid spermatids. At birth, the male germ cells are present in the sex cords of the testis in the form of large pale cells surrounded by supporting cells, also known as the Sertoli cells or the sustentacular cells. A little before puberty the sex cords acquire a lumen and get converted into seminiferous tubules. The spermatogonia, up to the stage of spermatids, are situated in the deep recesses of Sertoli cells which not only provide support to the germ cells during spermatogenesis, but also provide nourishment during their development.

Stages of Spermatogenesis

Stages of spermatogenesis are described in **Figure 7.3.** In this process, primary spermatocytes are formed from spermatogonium, which then divides into two secondary spermatocytes, which eventually give rise to four spermatids. The process of spermatogenesis comprises of the following steps:

1. The type "A" spermatogonia divide repeatedly by mitotic division to form a large number of daughter cells of the same type of spermatogonia. The last division of spermatogonia type "A" results in the formation of spermatogonia type "B". Spermatogonia type "A" constantly divide by mitosis providing an endless supply of stem cells, of which only some, termed as spermatogonia type "B" enlarge and eventually develop into primary spermatocytes.

TABLE 7.1	Site of synthesis of various hormones
Hormone	**Site of synthesis**
Testosterone	Testis (Leydig cells)
Oestradiol	Testis (Leydig cells)
17-alpha-hydroxyprogesterone	Adrenal gland
Prolactin	Anterior pituitary gland
Inhibin	Sertoli cells
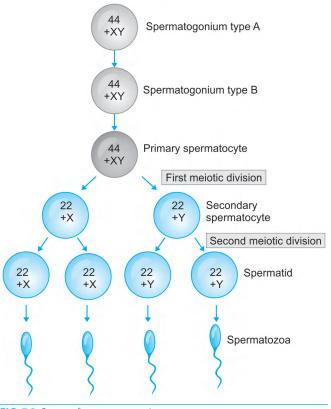
FIG. 7.3: Stages of spermatogenesis

2. Each primary spermatocyte divides by meiosis to form two secondary spermatocytes, having half the number of chromosomes, that is one cell with 22+X while the other with 22+Y chromosomes.
3. The secondary spermatocyte, which has 22+X or 22+Y chromosomes, divides by second meiotic division thereby producing the daughter cells with 22+X and 22+Y chromosomes. These daughter cells are the spermatids.
4. The spermatids transform into spermatozoa, by a process known as spermiogenesis. The time required for the completion of the entire process of development of spermatozoon from the spermatogonium is about 70–75 days. Nearly 200–300 million spermatozoa are discharged in each ejaculation. This large number of spermatozoa helps in ensuring fertilisation of the female gamete.

Sperms are formed in the seminiferous tubules only after puberty. After formation the mature spermatozoa enter the lumen of the seminiferous tubules. The contractions of the walls of the seminiferous tubules help in pushing the sperms towards the epididymis. The spermatozoa acquire the ability to become mobile here in the epididymis. Prior to ejaculation, sperms are stored in the vas deferens. Following ejaculation, the sperms retain their motility in the cervical canal and uterus for 5–7 days, but this does not mean that they have the power to fertilise. They retain their ability to fertilise only for approximately 24 hours after being implanted in the vagina. The spermatozoa are responsible for approximately 10% of the volume of the ejaculate. The vast majority of ejaculate is composed of secretions added from the various glands in the male reproductive tract—from the seminal vesicles, the prostate, the bulbourethral and urethral glands. The normal sperm count is 10^8/mL.

At about 5th week of gestation, the gonads are present in an indifferent stage in form of gonadal ridges. The differentiation of this gonad into testis or ovaries takes place at 6–9 weeks of gestation. The testes develop in the genital ridge after 7 weeks of gestation. Its development requires not only the presence of the SRY gene but also its interaction with other genes. The testes descend through the inguinal canal in the third trimester (probably under the influence of androgens). The development of indifferent gonad into testis is directed by TDF (testes determining factor) gene which is located in the SRY region (sex determining region) of Y chromosome. Peak synthesis of testosterone occurs between 15 to 18 weeks of gestation, following which there is a decline in its synthesis.

Testosterone

Testosterone is a steroid hormone, a C-19 derivative. It induces the development of secondary sexual characteristics in the male. These include deepening of the voice, body hair and penile growth. It is secreted by the interstitial calls of Leydig in testes in males and from the adrenal cortex in females. Systemic administration of testosterone inhibits spermatogenesis.

The secretion rate of dehydroepiandrosterone sulphate ($DHEA-SO_4$) is greater than that of testosterone. The secretion rate is 10 mg/day.

Oogenesis

The various stages of oogenesis have been described in **Figure 7.4**. The primordial germ cells, after arriving in the female gonad, differentiate into oogonia, resulting in the formation of several clusters of oogonia. It is assumed that each cluster of oogonia is formed by one primordial cell. These clusters get surrounded by flat epithelial cells which are derived from epithelial covering of the ovary. These represent the follicular cells. Starting from 6–8 weeks of gestation, rapid mitotic division occurs so that the number of oogonia reaches 6–7 million by 16–20 weeks (4–5 months). This represents the maximal oogonal content of the gonads. From this point onwards, germ cell content reduces irretrievably. By the 7th month, a large number of oogonia have degenerated except for those that are present near the surface of the ovary. At birth, the cortical content of germ cells is about 500,000 to 2 million as a result of pre-natal oocyte depletion. Also, the ovary at this time is about 1 cm in diameter and 250–350 grams in weight. During this time, the division into cortex and medulla has been achieved.
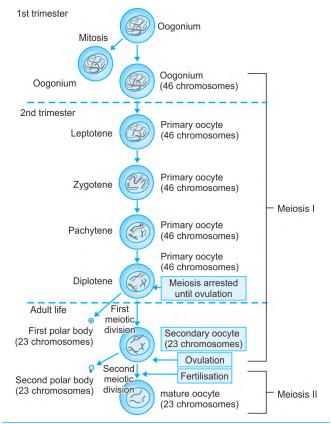
FIG. 7.4: Stages of oogenesis

At the onset of puberty, the germ cell mass has been further reduced to 300,000–500,000 units. Out of these, only about 500 would actually ovulate. The remaining oocytes would get depleted to the point of menopause. After attainment of puberty, each month about 15–20 oocytes would get selected to mature, out of which eventually only one would ovulate.

The germ cells give rise to oogonia around 9th week of foetal life. These enter the first meiotic division and are converted into oocytes. Progression of meiosis to the diplotene stage is accomplished throughout the pregnancy and completed by birth. In the last week before birth, all the primary oocytes complete the diplotene stage, but do not progress further. Instead, they get attested in the diplotene stage of prophase. During this phase, the chromatin network becomes lacy. The primary oocytes remain arrested at this stage and do not undergo the completion of first meiotic division till the age of puberty, when the completion of first meiotic division occurs at the time of ovulation. Second meiotic division starts, but gets arrested in the metaphase, which is completed only at the time of fertilisation.

Primordial follicle comprises of a primary oocyte arrested in the prophase of meiosis. It is enveloped by a layer of spindle-shaped pregranulosa cells, surrounded by a basement membrane. This unit is known as the primordial follicle, which gets converted into a primary follicle as the pregranulosa cells become cuboidal in nature, proliferate and get converted into the granulosa cells **(Fig. 7.5)**. In the primary follicle, granulosa cells multiply to form multiple layers and may acquire a diameter between 40 to 54 μm. The cells of the granulosa and the oocyte secrete a layer of glycoproteins on the surface of the oocyte which forms a tough covering, the zona pellucida around the oocyte. As the follicle grows, the cells of the theca folliculi get arranged into an inner layer of secretory cells called as theca interna and an outer fibrous layer—the theca externa which is derived from the ovary. With further growth, the primary follicle gets converted into pre-antral follicle and then into antral follicle. Prior to ovulation, there appear spaces filled with fluid amongst the granulosa cells and coalesce to form the cavity of the ovarian (Graafian) follicle. As a result of accumulation of this fluid (liquor folliculi), the oocyte, along with its covering of the follicular cells gets pushed eccentrically towards one side. The granulosa cells surrounding the oocyte form cumulus oophorus. The mature follicle may be 25 mm in diameter, which projects for about 15 mm on the surface of ovary.

Ovulation

Under the influence of the hormone, gonadotropin-releasing hormone (GnRH), produced by the hypothalamus, the anterior pituitary is stimulated to produce FSH and LH hormones. FSH is responsible for further development of primary follicles. Of these 15–20 primary follicles which start developing, only one follicle eventually develops near the surface of the ovary to the stage of full maturity. The other oocytes, which are not destined to ovulate, die and get converted into fibrous tissue, the corpus atreticum.

Ovulation takes place as the ovarian follicle ruptures and the discharged oocyte is carried into the peritoneal cavity via the uterine tube. Once the oocyte has been extruded out, the cells of the empty ovarian follicle get converted into the corpus luteum which produces progesterone for about 14 days, in absence of fertilisation and for 3–4 months if fertilisation has taken place, after which it eventually dies off. The oocyte moves from the ovary to the uterine tube and may get fertilised by the male gamete in the ampulla of the uterine tube. Even though many spermatozoa may
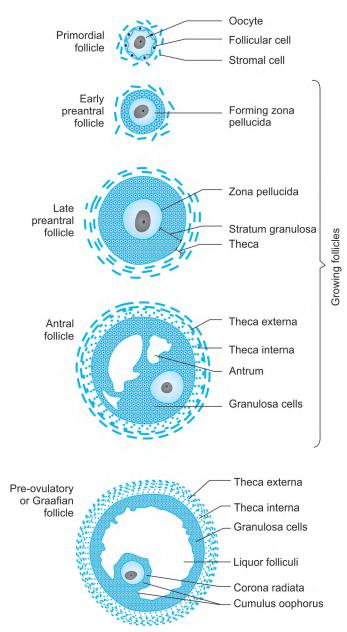
FIG. 7.5: Stages of development of ovarian follicles

approach the oocyte, only one spermatozoon is allowed to enter the oocyte. It passes the zona pellucida by capacitation and acrosome reaction. The process of fertilisation between two haploid gametes results in the formation of a diploid zygote, thereby restoring the number of chromosomes to that of the normal somatic cell. The ovum remains viable for up to 4 days after ovulation. There is a "window" of about 24 hours during which the ovum can be fertilised. On fertilisation the chromosomal configuration can be of two types, either 44 (XY), i.e. a male child or 44 (XX), i.e. a female child. The process of fertilisation and implantation has been summarised in **Figure 7.6**. Fertilisation of the sperm and egg occurs in the fimbrial/ampullary end of the fallopian tube and transport between this region and implantation in the endometrium takes between 5 to 7 days. The fertilised ovum implants, well past the 16-cell stage.

Development of Human Embryo

Cleavage Division and Formation of Morula

The zygote, a diploid cell with 46 chromosomes, formed as a result of fertilisation of mature egg with a sperm undergoes numerous cleavage divisions to produce cells known as blastomeres **(Fig. 7.7)**. At this stage, the zygote is present inside the fallopian tube and is surrounded by a thick zona pellucida. For 3 days as the blastomeres continue to divide, they produce a solid, mulberry-like ball of cells. This 16-celled ball is called morula. The morula enters the uterine cavity approximately 3 days after fertilisation, and floats around in the cavity for a few more days. During this time, fluid gradually accumulates between the morula's cells, transforming the morula into a blastocyst.

Formation of Blastocyst

As the fluid begins to pass through the zona pellucida and get accumulated between the cells, the morula looks like a cyst and becomes the blastocyst. When the blastocyst reaches 58-celled stage at about 4–5th day of fertilisation, it gets transformed into two types of cells: trophoblast cells and an inner cell mass, which forms the embryo proper **(Fig. 7.8)**. The cells of the inner cell mass get displaced to one side. This side of the blastocyst is known as embryonic pole and the opposite side as the abembryonic pole. The inner cell mass, (consisting of blastomeres) is destined to form the various tissues of the embryo. The trophoblast comprises of outer single layer of flattened cells, which later get converted into the future placenta. The cavity of the blastocyst is called the blastocoele.

The function of zona pellucida which is still surrounding the blastocyst is mainly protective. Its presence prevents implantation of the blastocyst in the uterine wall. As the fluid gets imbibed into the blastocyst and it increases in size, the tough layer of zona pellucida eventually ruptures. Once in the uterus, as the zona pellucida ruptures and gets casted off, the trophoblast comes in direct contact with the endometrium to begin the implantation. Implantation begins with the burrowing of the blastocyst into the endometrium. At this stage there are about 100–250 cells in the blastocyst. The blastocyst begins to implant at about 6–7 days after fertilisation. The most common site of implantation is upper posterior wall of the uterine cavity. The prerequisite for successful implantation requires an endometrium which has been primed with oestrogen and progesterone. The process of implantation involves the destruction of maternal tissue, following which, the
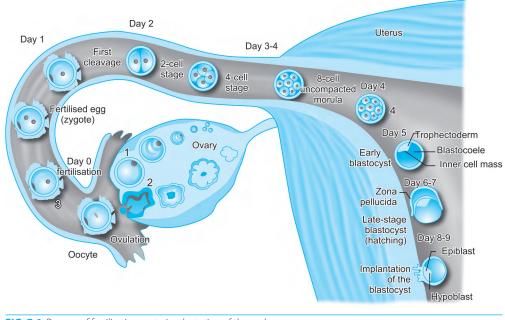
FIG. 7.6: Process of fertilisation up to implantation of the embryo
Key: 1. Maturation of ovarian follicle; 2. Ovulation and formation of corpus luteum; 3. Fertilisation in ampulla of uterine tube; 4. Formation of morula
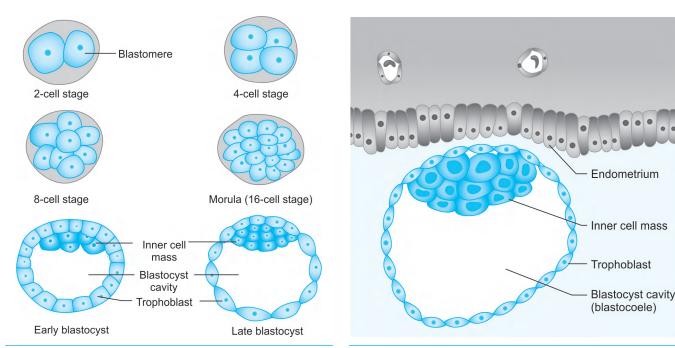
FIG. 7.7: Conversion of embryo from 2-celled stage to the late blastocyst stage

FIG. 7.8: Blastocyst at a later stage with inner cell mass shifted to one side

blastocyst sticks to the uterine wall, gradually eroding the uterine lining. The invasiveness of trophoblast helps in attachment of the blastocyst to the decidua and helps in deriving nutrition for the growth of the embryo. Moreover, once inside the uterine wall, the blastocyst invades the mother's blood vessels, and destroys the walls of these vessels. This process helps in establishing an uteroplacental blood flow. By 8th day post-fertilisation, the trophoblast gets differentiated into an outer multinucleated syncytium known as syncytiotrophoblast and an inner layer of cytotrophoblasts. In the multinucleated syncytium, there is absence of cell borders within the individual cells. As the trophoblastic cells invade deeper into the endometrium, by 10th day post-fertilisation, the blastocyst gets totally embedded within the endometrium **(Figs 7.9A and B)**. As the blastocyst implants into the uterine wall, simultaneously it also prepares its cells and surrounding endometrium to develop into a placenta. The blastocyst consists of two groups of cells: the inner cell mass, which become the embryo, and the trophoblast cells, which are crucial in producing the chorion (the embryonic portion of the placenta). As early as 7–8 days after fertilisation, the inner cell mass or the embryonic disc gets differentiated into a top layer, ectoderm (epiblast) and an underlying layer of endoderm (hypoblast). Small cells appear between the embryonic disc and trophoblast enclosing a space that later gets transformed into amniotic cavity. The ectoderm forms the floor of the amniotic cavity while the roof is formed by amniogenic cells. The endodermal germ layer produces additional cells which form a new cavity, known as the definitive yolk sac.
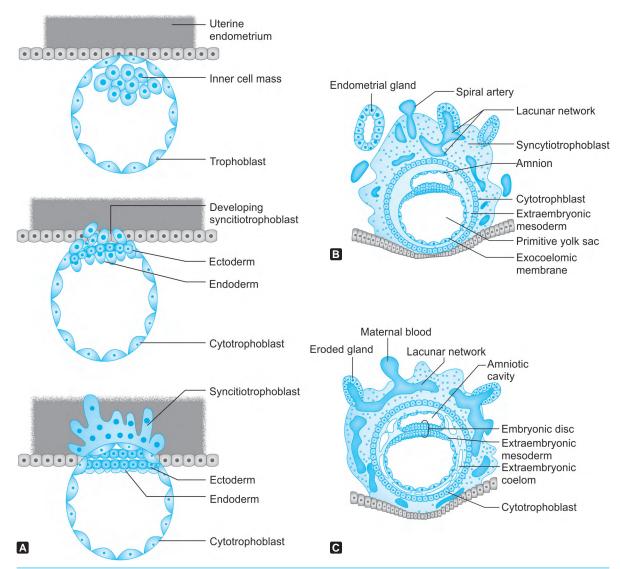
As the amniotic fluid accumulates in the amniotic cavity, it enlarges. Small embryonic mesenchymal cells appear as isolated cells within the cavity of blastocyst. They soon line the cavity of blastocyst. When the blastocyst is completely lined with mesoderm, it is termed as chorionic vesicle. This is surrounded by a membrane called chorion which is composed of trophoblasts and mesenchyme.
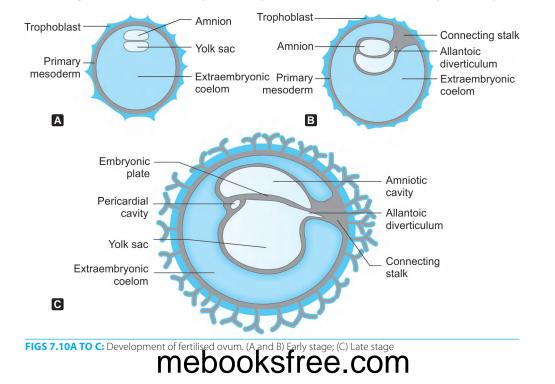
Soon numerous small cavities appear within the extraembryonic mesoderm. These cavities shortly become confluent and form the extraembryonic coelom **(Fig. 7.9C)**.

The extraembryonic coelom splits the extraembryonic mesoderm into two layers: The extraembryonic somato-pleuric mesoderm, lining the trophoblast and amnion, and the extraembryonic splanchnopleuric mesoderm, covering the yolk sac. The extraembryonic somatic mesoderm and the two layers of trophoblast constitute the chorion. The membrane called amnion is composed of amniogenic cells along with somatopleuric extraembryonic mesoderm. As the folding of the embryo takes place, amniotic cavity completely surrounds the embryo **(Figs 7.10A to C)**. With the development of extraembryonic coelom, the yolk sac becomes much smaller and is known as the secondary yolk sac. Around the amniotic cavity is the extraembryonic coelom. Outside the embryonic coelom is the chorion. As the foetus grows, there is enlargement of the amniotic cavity, resulting in progressive reduction in the size of extraembryonic coelom. Eventually the extraembryonic coelom completely disappears, causing the amnion to come in contact with chorion and fuse with it to form the chorioamniotic membrane.

The mesodermal cells, in which extraembryonic coelom has not extended, eventually condense to form the body stalk. This connects the embryo to the chorion, and is responsible for supplying the nutrients. Connecting stalk later forms the umbilical cord. As previously described, in

FIGS 7.9A TO C: (A) Stages of implantation of blastocyst; (B) Blastocyst at the time of implantation; (C) Blastocyst at 12th day after fertilisation

FIGS 7.10A TO C: Development of fertilised ovum. (A and B) Early stage; (C) Late stage

the early stages, the embryo acquires the form of a three layered disc. This disc may be known by various names such as the germ disc, the embryonic area, embryonic shield or embryonic disc. The three layers also called as germ layers, from outside inwards are: ectoderm (outer layer), mesoderm (middle layer) and endoderm (inner layer). These three layers of embryo are responsible for formation of different organ systems and tissues giving the embryo more "human-like" appearance **(Figs 7.11A to E)**. When the embryo becomes 7 or 8 weeks old, it is known as a "foetus".

The cells lining the secondary yolk sac which were flat earlier now become cuboidal. In a circular area near the margin of the disc the cuboidal cells of endoderm become columnar. This area is known as the prochordal plate. The prochordal plate decides the axis of the embryo, which can now be divided in two lateral halves, the right and the left.
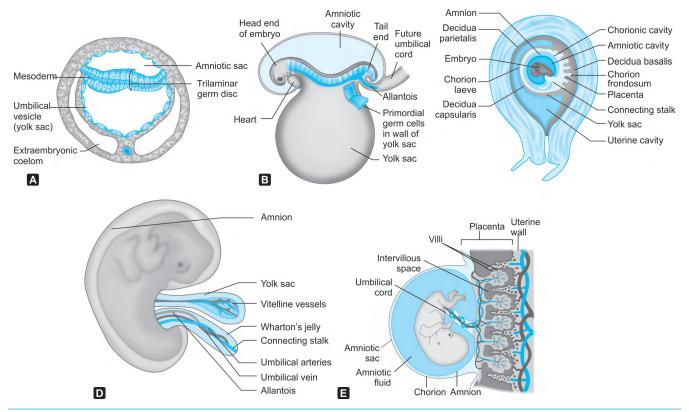
While the prochordal plate is being formed, some of the endodermal cells on the axis near the caudal end of the embryo start proliferating and form a linear elevation towards the amniotic cavity on the central axis of the embryo. This elevation is known as the primitive streak. In the region of the prochordal plate, the ectoderm and the endoderm are in contact since the mesoderm does not come here. This region is also known as the oral membrane or the buccopharyngeal membrane. The oral membrane is at the cephalic end of the embryo and is the future mouth. A similar area is formed at the caudal end where the ectoderm

and the endoderm are in contact. This area is the cloacal membrane and is the future anal and genital area.

Development of Human Placenta

The placenta is an organ with dual origin, developing both from the foetus and the mother. A part of placenta develops from foetal chorion, and the rest develops from maternal endometrium. The foetal component consists of chorionic plate and chorionic villi, whereas the maternal component consists of decidua basalis. A structure known as cytotrophoblastic shell attaches the maternal and foetal component to one another.

Decidua

On implantation of the blastocyst, the secretory phase of the endometrium undergoes decidual reaction, which comprises of the following changes: enlargement and vacuolation of the stromal cells and accumulation of glycogen and lipids within these cells. The first signs of the decidualisation reaction can be seen as early as day 23 (10 days after the peak of the LH surge) of the normal menstrual cycle. The progressive decidualisation of the endometrial stroma in the later part of the menstrual cycle prepares the uterine lining for the presence of the invasive trophoblasts, but at the same time closes the door to implantation.

FIGS 7.11A TO E: (A) Development of trilaminar germ disc; (B) Development of early stage embryo; (C) Uterus with embryo of 6 weeks size; (D) 6 weeks old embryo; (E) Development of foetus

Initially, the spiral arteries of the endometrium become prominent. Over the next few days, the stromal cells surrounding the spiral arteries become increasingly eosinophilic and become enlarged under the influence of progesterone. These cells are known as predecidual cells. This preparation of the endometrial lining in anticipation of pregnancy occurs during the secretory phase of every menstrual cycle. In the event that no pregnancy occurs, this decidual lining of the endometrium is shed off in form of menstrual cycle. During parturition, the decidua is shed off along with the placenta and membranes (amnion and chorion) which are sometimes also known as the "after birth". The decidual separation takes place at the level of stratum spongiosum and the entire thicknesses of the decidua parietalis along with the chorion laeve and amnion, which are fused with the decidua, are expelled. The placental separation causes rupture of uterine blood vessels, resulting in some bleeding at the time of child birth. Contraction of the myometrial muscles results in the closure of the ends of the ruptured blood vessels after the expulsion of the placenta. Following the delivery of the baby and placenta, a thin layer of stratum spongiosum is left behind in the uterus but it also gets degenerated and is soon cast off.

The endometrium of the uterus is in secretory phase of menstruation at the time of implantation. After implantation, the syncytiotrophoblast starts secreting the hormone human chorionic gonadotropin (hCG). Under the influence of this hormone, the secretory changes taking place in the endometrial lining are further intensified resulting in conversion of endometrial lining into specialised cells which are known as the decidua. This reaction is known as decidual reaction and endometrium is known as decidua. By the 10th day of implantation, the blastocyst completely penetrates below the surface of decidua. The part of decidua at the site of the foetal portion gets transformed into chorion frondosum (the foetal precursor of mature placenta), whereas the maternal part is known as decidua basalis. The decidua at this stage can be classified into three portions. The side lying in contact with the blastocyst at the site of implantation is the decidua basalis; the decidua lying over the surface of the implanted blastocyst is the decidua capsularis; the remainder of the decidua lining the inside of the uterus is the decidua vera **(Fig. 7.12)**. Initially, the decidua capsularis is separated from decidua parietalis by uterine cavity. With further enlargement of the uterine cavity, the decidua capsularis and decidua parietalis fuse with each other.

Chorionic Villi Formation

As previously mentioned, by the 8th day following fertilisation, the trophoblast gets divided into two layers: the syncytiotrophoblast and the cytotrophoblast. The syncytiotrophoblast erodes the maternal epithelium and enters the endometrium. Around 12th day after fertilisation, development of the chorionic villi takes place around the chorionic sac. The chorionic villi are the finger-like projections arising from chorion and serve as precursors of human placenta. Chorionic villi of the placenta primarily function to transfer oxygen and other important nutrients between mother and foetus. In the early pregnancy, the villi are distributed over the entire periphery of the chorionic membrane. The chorionic villi in contact with decidua basalis, proliferate to form chorion frondosum, the foetal component of the placenta. As the growth of embryonic and extraembryonic tissues continues, the blood supply of the chorion facing the endometrial cavity is restricted. Therefore, the villi in contact with the decidua capsularis cease to grow and degenerate. This portion of the chorion becomes the avascular and along with decidua parietalis is known as chorion laeve or the smooth walled chorion.

Stages of Chorionic Villi

As the blastocyst with its surrounding trophoblasts grows and expands into the decidua, the outer pole of the mass expands outwards towards the endometrial cavity. The opposite, inner most pole results in the formation of placenta comprising of villous trophoblasts and anchoring cytotrophoblasts. The chorionic villi are first formed from chorion, i.e. trophoblast and underlying extraembryonic mesoderm. The development of chorionic villi passes through three stages: primary villi, which develops into secondary villi and finally forms tertiary villi. When the villi are mature, this zone becomes a site of maternal-foetal exchange due to interaction between maternal-foetal blood vessels.

Primary Villi

With deeper invasion of blastocyst into the decidua, small cavities called lacunae appear inside syncytiotrophoblast. These lacunae fuse with each other, forming a complicated labyrinth which gets arranged radially around the chorion. The lacunae are separated from each other by columns of
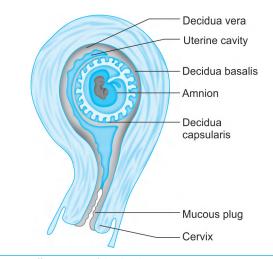
FIG. 7.12: Different parts of the decidua

syncytiotrophoblast. These columns are known as trabeculi. The lacunae communicate with each other around the trabeculi and are filled with blood. Soon there is proliferation of cytotrophoblastic cells which invade the trabeculi. This results in conversion of trabeculi into solid villi composed of cytotrophoblast core which is surrounded by syncytium. These villi are known as primary villi **(Fig. 7.13)**. Initially, these villi are located over the entire surface of blastocyst, but with the increasing period of gestation, they disappear from all areas except over the most deeply implanted portion, the site designed to form the placenta.

Secondary Villi

Mesenchymal cords derived from embryonic mesoderm invade the solid trophoblast columns resulting in the formation of secondary villi. The extraembryonic somatopleuric mesoderm lying deep to the cytotrophoblast invades the central part of each trabeculies. The trabeculies therefore now acquire three layers from outer to inner side and include: syncytiotrophoblast, cytotrophoblast and mesoderm. These are known as secondary villi **(Fig. 7.14)**.

Tertiary Villi

After the angiogenesis occurs in the mesenchymal core of secondary villi, the result is the formation of tertiary villi **(Fig. 7.15)**. These vessels are connected to foetal vascular system through chorionic and umbilical vessels. Maternal venous sinuses are tapped early in the implantation process, but until 14–15th day after fertilization, maternal blood does not enter the intervillous space. By 17th day, foetal blood vessels are functional and the placental circulation is established. The placental circulation is completed when the blood vessels of the embryo are connected with chorionic blood vessels.

Cytotrophoblastic Shell

With increasing gestation, the cells of the cytotrophoblast in the tertiary villi proliferate and pass through the syncytiotrophoblast at the tip of the villi resulting in the formation of a continuous layer of cytotrophoblasts on the surface of decidua which is known as the cytotrophoblastic
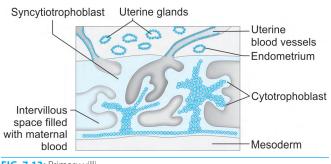
shell. This helps in fixing the chorionic villi to the decidua. Thus, the tertiary villi at one end are continuous with foetal component (chorion) and to the maternal component (decidua) at the other end. The villi develop numerous branches which also develop progressively into primary, secondary and tertiary villi.

As a result of these branches, the surface area for exchange between maternal and foetal blood increases tremendously. The intervillous space gets filled by maternal blood and the villi float in this blood.

Human Placenta

The human placenta is rounded and discoidal, flattened cake-like structure moulded within the endometrial lining of the uterus and weighs about 500 grams. The average diameter of the placenta is about 15–20 cm and is about 3 cm thick centrally tapering towards the edges. As the syncytiotrophoblast erodes endometrium of decidua basalis, the endometrial projections called septa projects into the intervillous space between the two villi. The area between two septa is known as lobes or cotyledons. On an average the placenta contains about 15–20 cotyledons. Each cotyledon contains 2–3 anchoring villi. Despite the small

FIG. 7.14: Secondary villi

FIG. 7.15: Tertiary villi
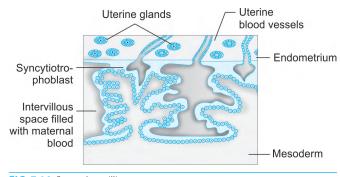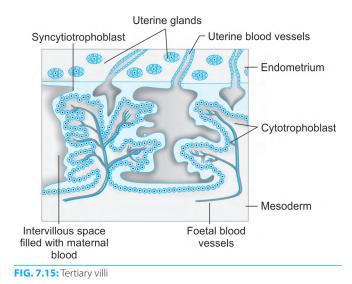
FIG. 7.13: Primary villi
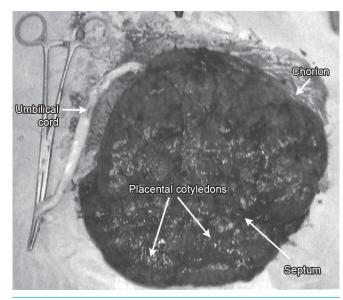
size of a placenta, the surface area available for maternal-foetal exchange is greatly large due to the presence of villi. Each primary chorionic villus divides at least five times, forming villous trees. This leads to formation of extremely large number of terminal villi, resulting in a large surface area, all of which is bathed in the uterine blood.

These placental cotyledons are visible on the maternal side of the placenta **(Fig. 7.16)**. On the other hand, the placenta shows a smooth surface from foetal side **(Fig. 7.17)** due to presence of smooth chorion. At the centre of the foetal surface the umbilical cord is attached. Umbilical cord serves as the connection between the mother and the foetus and develops from the connecting stalk. The connecting stalk is that part of extraembryonic mesoderm in which extraembryonic coelom does not develop. It is attached to the roof of amniotic cavity at one end and to trophoblast at the other. Gradually, the connecting stalk becomes narrower and its attachment moves towards the caudal end of the embryo. As the embryo folds develop, the attachment of the connecting stalk moves ventrally in the region of the umbilicus. With increasing period of gestation, as the placenta develops, the connecting stalk connects the embryo or foetus with the placenta. As the connecting stalk is converted into the umbilical cord, two umbilical arteries and one umbilical vein develop in the connecting stalk. The mesoderm of the connecting stalk later gets converted into a gelatinous substance called Whartons jelly. The umbilical cord varies in length but on an average is about 55 cm in length. The mature umbilical cord contains three large blood vessels: two umbilical arteries bringing foetal blood to the placenta, and a single umbilical vein returning blood to the baby's heart.

Blood present in the intervillous spaces comes through the maternal endometrial arteries and is drained by maternal endometrial veins. The chorionic villi contain foetal blood as they contain branches of umbilical vein and umbilical arteries. The maternal blood in the intervillous space is separated from the foetal blood in placental villi through a membrane known as placental membrane or placental barrier. The exchange of various substances like gases (oxygen and carbon dioxide), nutrients and waste products takes place across this membrane. The layers of placental membrane starting from the maternal side comprise of the following:
❑ Syncytiotrophoblast
❑ Cells of cytotrophoblast
❑ Basement membrane of cytotrophoblast
❑ Mesoderm
❑ Endothelium and basement membrane of the branches of umbilical blood vessels (foetal blood) in the villi.

Total area of the placental membrane is very large, which ensures effective exchange of substances across the mother and the foetus. The foetus receives oxygen from the mother through the umbilical vein and gets rid of carbon dioxide by passing it through umbilical arteries to the mother. Important nutrients, essential for the foetal growth and development, reach the foetus from the mother through this placental membrane, whereas the foetus eliminates its waste products by passing them through this membrane to the mother. At about 6 months of gestation, the thickness of placental membrane is about .025 mm. As the foetal growth continues and its nutritional demands increase, the placental membrane becomes thinner (.002 mm), thereby tremendously increasing its efficiency for transport. In addition to its primary goal of facilitating transport between mother and foetus, the placenta is also a major endocrine organ. In almost all mammals, the placenta synthesizes and secretes the hormones: progestins and oestrogens, hCG, relaxin, and human placental lactogen. Placenta also plays an important role in creation of foetal immunity.
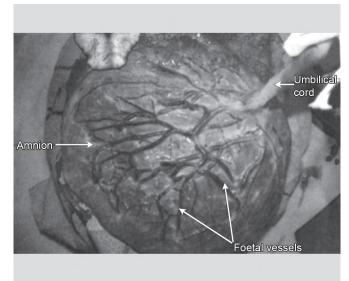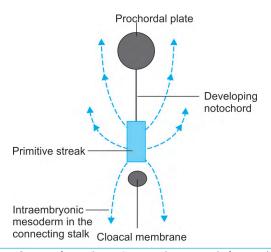
FIG. 7.16: Maternal side of the placenta
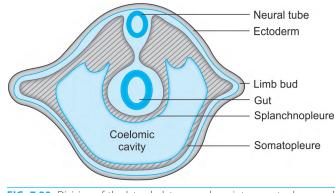
FIG. 7.17: Foetal surface of the placenta

Organogenesis

The key time for organogenesis is between 6 to 8 weeks, with the total process spanning 4–10 weeks. A linear raised area is formed over the superior surface of the embryonic disc towards the caudal end of the embryo due to linear proliferation of the ectodermal cells along the central axis of the embryo near the tail. This is known as primitive streak. As a result of the elongation of the embryonic disc, the primitive streak gets enlarged. The primitive streak gives rise to the intraembryonic mesoderm. The process of formation of the primitive streak and the intraembryonic mesoderm is known as gastrulation **(Fig. 7.18)**. The three germ layers: ectoderm, mesoderm and endoderm are initially present in the form of a circular disc-like structure. Soon there is a disproportionate growth of the ectoderm at the opposite poles so that the embryo prolongates into an oval structure with each end curving towards the yolk sac. One end forms the head fold and other the tail fold. The amniotic sac enlarges so that it completely surrounds the developing embryo and the yolk sac. A groove develops in the middle on the amniotic surface of the ectoderm. As the edges of the groove grow over, the groove gets converted into

a tube known as the neural tube. Neural tube gives rise to the nervous system as would be described later in the text.

Meanwhile the lateral growth of mesoderm occurs. The part nearest to the midline develops into the paraxial mesoderm, while the most lateral part becomes the lateral plate mesoderm. The mesoderm in between is known as intermediate cell mass. The three parts of intraembryonic mesoderm are illustrated in **Figure 7.19**. Simultaneously, the growth of endoderm first occurs laterally and then ventrally, gradually folding to form the gut tube. The lateral plate mesoderm divides to form somatopleure and splanchnopleure **(Fig. 7.20)**. While the somatopleure remains adjacent to the ectoderm, the splanchnopleure grows around the developing gut. The space between the somatopleure and splanchnopleure forms the coelomic cavity, which later forms the pleural and peritoneal cavities.

The paraxial and the intermediate cell mass develop into discrete masses of cells or somites along the length of the embryo. The somites related to paraxial mesoderm develop into the vertebrae, dura mater and muscles of the body wall. The intermediate cell mass develops in a ventral direction, forming the origins of the urogenital system. The limb buds also develop from the lateral plate mesoderm, pushing out a covering of ectoderm. Different body organs which are derived from various germ layers are enlisted in **Table 7.2**.

Epithelium is derived from all three germ layers, e.g. endoderm forms epithelial lining inside viscera; mesoderm

FIG. 7.18: Process of gastrulation. Arrows indicate spread of intraembryonic mesoderm from primitive streak

FIG. 7.20: Division of the lateral plate mesoderm into somatopleure and splanchnopleure
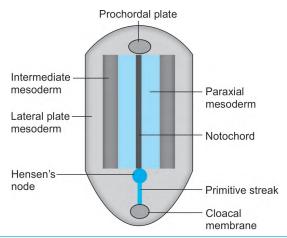
FIG. 7.19: Three columns of intraembryonic mesoderm

TABLE 7.2	Fate of germ layers
Germ layer	**Structures derived**
Endoderm	Epithelial lining of respiratory and gastrointestinal tract
Mesoderm	Cardiovascular system, reproductive/excretory organs, smooth and striated muscles, connective tissues, vessels, and skeleton
Ectoderm	Surface ectoderm—epidermis, and other external structures Neuroectoderm—central and peripheral nervous system, neural crest cells and derivatives

forms mesothelium lining outside of viscera, and ectoderm forms the skin epithelium.

The uterine endometrium is derived from the endoderm. The mammary gland is derived from the ectoderm as is the tongue epithelium and the pineal gland. Tongue muscle is derived from mesoderm. The mesoderm of the urogenital ridges gives rise to the ovarian stroma. The round ligament also arises from mesoderm.

Development of Genitourinary System

The sexual identity of individuals depends on their genetic, gonadal and phenotypic sex. Genetic or chromosomal sex is determined by the sex chromosomes, with XX karyotype being a genetic female and XY karyotype being a genetic male. Chromosomal sex is determined at the time of fertilisation and is dependent on the presence of Y chromosome. The main gene which is involved for directing the development of testes is the SRY gene, located on the Y chromosome. In the absence of Y chromosome, the bipotential gonad differentiates into an ovary about 2 weeks later than when testicular development begins in the male. Gonadal sex, which is determined by the genetic sex, is established next. Gonadal sex is dependent on the presence of gonads: testes in males and ovaries in females. Gonadal sex controls the development of both internal and external genitalia. Internal genitalia in males comprises of testes, epididymis and vas deferens, while in females it comprises of fallopian tubes, uterus and cervix. Phenotypic sex is determined by the appearance of external genitalia and secondary sexual characteristics, which develop at the time of puberty.

Development of Gonads

The development of gonads begins during the 5th week of gestation in the human embryos with the development of a protuberance known as the genital or gonadal ridge **(Fig. 7.21)**. The appearance of the primitive gonad is similar in both sexes until 42 days after fertilisation when differentiation of seminiferous tubules occurs. At this stage, the gonads are present in form of coelomic prominences overlying the mesonephros, which forms the gonadal ridges. Just below this ridge lies the mesonephric duct. Mesonephros and genital ridge are together termed as
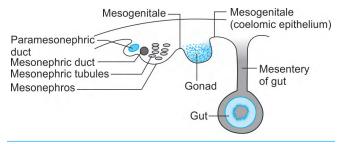
FIG. 7.21: Development of primitive gonads

the urogenital ridge. The differentiation of the primitive gonads along with a male or female line takes place at around 6–9 weeks of gestation. The primordial germ cells which eventually give rise to the ova or sperms in females and males respectively migrate from the wall of the yolk sac into the genital ridge between 4th to 6th weeks of gestation, simultaneously proliferating at the same time. As the cells of the genital ridge multiply and proliferate, its size increases and therefore the mesonephros gets displaced dorsally and laterally. Once the germ cells arrive in the nascent gonads, further differentiation into the male or female depends upon the sex of the gonadal somatic cells and the signals in the surrounding environment rather than the chromosomal sex of the germ cells themselves. The gonads do not obtain male or female status until the 7th week of development. The testis is formed in the lumbar region and thereafter descends to the scrotum by passing through the inguinal canal.

The ovary develops from the primitive cortex. Ovaries also develop in the lumbar region and they too descend, but their descent is arrested by the secondary attachment of gubernaculum to the uterus. At 10 weeks conception there is meiotic entry of oocytes in the medulla of the primitive gonad of the developing foetus. Gonocytes are diploid and only become haploid in the gonad via meiosis. Mitosis in oogonia is completed in the 7th month of foetal life.

Development of Internal Genitalia

The mesonephric ducts (Wolffian ducts) and paramesonephric ducts (Müllerian ducts) are two discreet duct systems, which are responsible for the development of internal genitalia in males and females respectively. These duct systems coexist in all embryos during the ambisexual period of development (i.e. up to 8 weeks of gestation). Thereafter, one duct system persists, giving rise to specialised ducts and glands and the other one regresses leaving behind the nonfunctional vestiges. Both the duct systems arise within the urogenital ridge during embryogenesis, running in length and terminating in the cloaca. The Wolffian duct or the mesonephric duct is the embryonic duct of the mesonephros. It connects the primitive kidney (Wolffian body or mesonephros) to the cloaca. The Müllerian tube, one on each side, runs close to the lateral side of the Wolffian duct caudally until it reaches a lower level where it turns medially, crosses in front of the Wolffian duct and joins its fellow from the opposite side. While forming the uterus Müllerian ducts fuse from below upwards. The epithelial covering of the ovary and the Müllerian duct are both formed from coelomic epithelium.

The Wolffian duct persists in the presence of a Y chromosome. Differentiation of the Wolffian duct system is stimulated by high local concentration of the hormone, testosterone. The Leydig cells of testis are responsible for producing testosterone and dihydrotestosterone. These

respectively determine the development of the Wolffian ducts into male internal genitalia, and the male external genitalia. The presence of testosterone in males facilitates the development of Wolffian duct into structures including rete testis, efferent ducts, epididymis, vas deferens, seminal vesicles and ductus deferens. In the male, the proximal part of the mesonephric duct becomes greatly elongated and convoluted to form the epididymis. Epididymis is a highly coiled structure about 6 metres in length. At the end of the epididymis is the vas deferens, which helps in storing the sperms. This ends into the ejaculatory duct, which joins the urethra. Simultaneous with the development of Wolffian duct system, the Müllerian duct system regresses in males. Müllerian inhibiting substance (MIS) is a non-steroidal substance, produced by the Sertoli cells in the testes, which causes regression of the paramesonephric duct. In the male, the paramesonephric ducts degenerate into the appendix testes and utriculus masculinus. The third (lower vertical) part of the paramesonephric duct remains rudimentary and gets incorporated in the prostate gland to form the prostatic utricle. Presence of MIS is required for the normal development of the Wolffian ducts along the lines of male phenotype. In its absence the Müllerian ducts contribute to the development of female phenotype. Embryological remnant of mesonephric duct in males is appendix of epididymis. Embryological remnants of the mesonephric tubules in the males include:

❑ The paradidymis
❑ The ductulus aberrans inferior
❑ The ductulus aberrans superior

On the other hand, in females, the Wolffian duct system regresses. The Müllerian duct system develops later and differentiates into the fallopian tubes, uterus and upper portion of the vagina **(Fig. 7.22)**. The uterus and upper
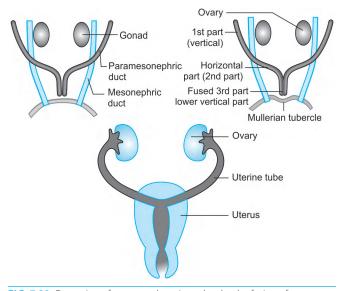
FIG. 7.22: Formation of uterus and uterine tubes by the fusion of paramesonephric ducts

one-third of the vagina develop from the paramesonephric ducts with the lower two-thirds of the vagina developing from the urogenital sinus. By the 5th month the vagina is usually completely canalised. In females, the remnants of the Wolffian duct can form the structures such as epoöphoron, paroöphoron, Skene's gland and Gartner's duct. The Gartner's duct runs medially through the broad ligament and down the side of the vagina, where cysts may form in it. It eventually gets incorporated into the wall of the cervix uteri. In both males and females, the Wolffian duct goes to form the trigone of the bladder, and the ureters.
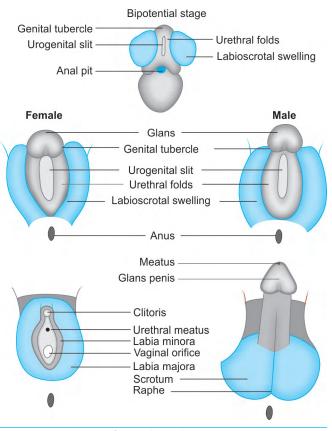
Development of the External Genitalia

External genitalia persists in the bipotential state until 9 weeks of gestation at which time it consists of a genital tubercle, urogenital sinus and lateral labioscrotal folds or swellings. The external genitalia are neutral primordia, which can develop into either male or female structures depending on the signals produced by the gonadal steroid hormones. The external genitalia can be recognised as male or female by the 16th week of foetal life by ultrasound examination. In females, the external genitalia are under the influence of oestrogen produced by the placenta and mother.

In males, the Leydig cells of foetal testis begin to secrete testosterone at 8–9 weeks of gestation. Under the influence of testosterone, the genital tubercle forms the penis, the edges of the urogenital sinus fuse to form the penile urethra and the labioscrotal folds fuse to form the scrotum. This process is complete by 12–14 weeks of gestation. Urogenital sinus also forms the prostate, bulbourethral glands, and urethra in males under the influence of androgens. Complete differentiation of the male external genitalia also depends upon the presence of the hormone, dihydrotestosterone, which is derived from testosterone with the help of the enzyme 5-α reductase.

In the females or males showing defects in androgen synthesis and action, the external genital primordia do not undergo masculinisation. In these cases, the genital tubercle develops into clitoris; the margins of the urogenital sinus remain separate and form the labia minora, whereas the labioscrotal folds form the labia majora and the urogenital sinus develops into the lower vagina and urethra **(Fig. 7.23)**. The urogenital sinus is endodermal because it is derived from the cloaca. It gives rise to caudal two-thirds of vagina in females; oestrogen is not necessary for this process. Female urethra is formed from the primitive urethra and the pelvic part of the urogenital sinus. Male urethra has three parts:

1. The prostatic part, as in females, is derived from the primitive urethra and the pelvic part of the urogenital sinus.
2. The membranous urethra is formed from the pelvic part of the urogenital sinus.
3. The penile urethra is derived from the phallic part of the urogenital sinus.

FIG. 7.23: Development of extrenal genitalia

FIGS 7.24A TO D: Formation of vagina: (A) Mesoderm of uterovaginal canal pressing on the posterior wall of the endodermal urogenital sinus forming Müllerian tubercle; (B) Sinovaginal bulbs formed by proliferation of the endoderm of urogenital sinus; (C) Solid vaginal plate formed by fusion of mesodermal uterovaginal canal and endodermal sinovaginal bulbs; (D) Vagina formed by canalisation of vagina plate (Part derived from mesoderm is blue; part derived from endoderm is grey)

FIG. 7.25: Parts of nephrogenic cord (pronephros, mesonephros and metanephros). The mesonephric duct gives out the ureteric bud and itself opens into the endodermal cloaca

The male urethra, up to the glans penis is endodermal while in the glans it is ectodermal. Prostate develops from the buds that arise from the caudal part of vesicourethral canal and the pelvic part of urogenital sinus.
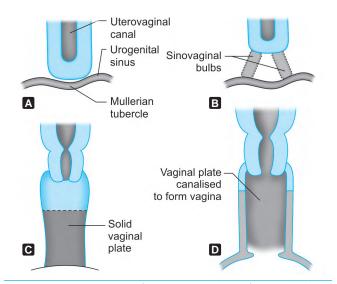
In females, abnormal androgen exposure between 9th to 14th week of gestation results in varying degree of masculinisation such as clitoral hypertrophy and labial fusion. In males the external genitalia does not undergo complete masculinisation if the androgen action is deficient during this period. This may result in the development of a small phallus, hypospadias and scrotal defects.

Development of Vagina
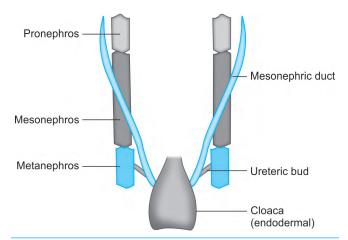
Vagina is derived from sinovaginal bulbs as well as from the vaginal plate **(Figs 7.24 A to D)**. It is partly mesodermal and partly endodermal in origin. The endodermal cells of the urogenital sinus proliferate to form two sinovaginal bulbs. A solid vaginal plate is formed by the fusion of mesodermal uterovaginal canal in the upper part and endodermal sinovaginal bulbs in the lower part. The central cells of the vaginal plate eventually break down to form the vaginal canal.

Development of the Urinary Tract

The kidneys, urinary tract and the majority of the reproductive organs, develop from the intermediate mesoderm between the somites and the lateral plate.

The kidney goes through three stages of development that recapitulate evolution of the kidney: pronephros, mesonephros and metanephros **(Fig. 7.25)**. In the majority of vertebrates, the pronephros becomes atrophic early in its embryonic life. The mesonephros becomes replaced by the metanephros. In adult humans, remnant mesonephric tissues are a source of cysts and tumours. The metanephros is responsible for the formation of the majority of the urogenital system in humans and is functional well before birth. Metanephros is formed by the mesoderm of the intermediate cell mass of the mesoderm also known as nephrogenic chord. Bowman's capsule and the glomerulus develop as part of the metanephros.

Ureter is formed by a diverticulum (ureteric bud) from the mesonephric duct. Ureter, at its upper end, divides to form the two major calices which also divide to form the minor calices. These structures form the collecting system of the kidney.

The urinary bladder is derived in part from the urogenital sinus, and in part from the ends of the mesonephric ducts. The trigone of the bladder and the posterior wall of the urethra are formed by the mesonephric ducts.

The kidneys attain their adult position during the 8th week of foetal life. The kidney which develops in the sacral region, subsequently ascends to its final place (thoracolumbar region). Urine formation begins at about the third month of foetal life, and continues increasing in volume till term. The mature foetus may well void 450 mL of urine daily into the amniotic cavity.

Pharyngeal Arches

The pharyngeal arches are any of the paired segmented ridges in each side of the throat of the early embryo. The pharyngeal arches give rise to several structures in the head and neck. The pharyngeal arches contribute to the formation of the head and neck and appear in the 4th and 5th week of development. Each arch has a mesodermal core lined by epithelium derived from endoderm. The outside is covered by ectoderm.

Formation of Pharyngeal Arches

With the formation of head and tail folds, the stomodeum (primitive mouth) lies caudal to the forebrain and cephalic to the pericardial cavity. The neck and the lower part of the face is formed between these two structures (forebrain cranially and pericardium caudally) by the formation of six pairs of visceral arches, which appear as proliferation of mesoderm in the region of hindbrain **(Fig. 7.26)**. These arches are known as pharyngeal or branchial arches. These prolongations of the mesoderm are covered on the outside by ectoderm and are lined by endoderm.

These arches grow ventrally to meet the similar arch on the opposite side. Between the arches, the ectoderm and the endoderm come almost in contact with each other having very little mesoderm between them. These depressions are called the pharyngeal clefts, or pouches (from the endodermal side) and branchial clefts or pouches (from the ectodermal side) **(Fig. 7.27)**.

Mesoderm of each of the pharyngeal arch gives rise to cartilage, branchial muscles and artery **(Fig. 7.28)**. Cartilage of each of these arches may remain as such or may ossify into bone or may disappear in some arches. The 5th arch is an abortive arch that disappears soon after its formation and no trace of its nervous or vascular elements are left.

Each branchial arch is supplied by a specific nerve which is derived from the cranial nerve. This nerve gives motor supply to the muscles of the arch as well as the sensory branches to the ectoderm and endoderm of the arch or its derivatives. The nerve that runs on the arch, that is immediately caudal to the cleft, is known as the post-trematic nerve of the arch. The nerve that runs on the caudal side of the arch is called as pre-trematic nerve. It must be noted that the pre-trematic nerve is present only in the first arch. Chorda tympani is the pre-trematic nerve of first arch. Different pharyngeal arches and their derivatives are described in **Table 7.3**.
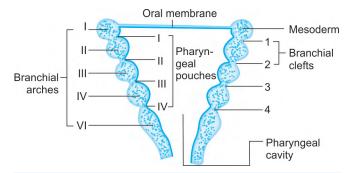
FIG. 7.27: Longitudinal section to show the formation of branchial arches
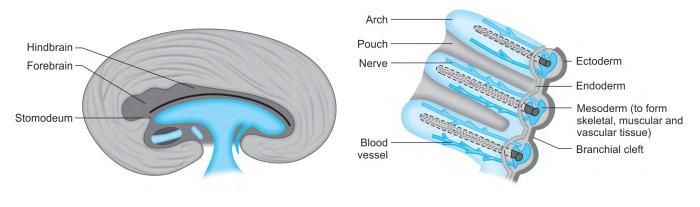
FIG. 7.26: Development of pharyngeal arches in the region of hindbrain

FIG. 7.28: Constituents of branchial arches

TABLE 7.3	Different types of arches and their derivatives	
Name of the arch	**Post-trematic nerve**	**Muscles/bones derived**
First pharyngeal arch/mandibular arch	Mandibular branch of trigeminal nerve (5th cranial nerve)	• Tensor tympani, Tensor palati, lateral and medial pterygoid • Masseter, temporalis, stylohyoid and anterior belly of digastric • Muscles of mastication • Mylohyoid • Anterior belly of digastric • Tensor palati and tympani muscles • The skeletal derivatives are: incus, malleus, part of the mandible, sphenomandibular ligament and zygomatic bone
Second pharyngeal arch/hyoid arch	Facial (7th cranial nerve)	• Muscles of facial expression (orbicularis oris and occuli, platysma, buccinator, frontalis) and muscles of auricle • Occipitofrontalis, platysma • Stapedius, stylohyoid ligament • Posterior belly of digastric • The skeletal derivative of second pharyngeal pouch: stapes
Third arch	Glossopharyngeal nerve	Stylopharyngeus
Fourth arch	Superior laryngeal nerve	Muscles of pharynx, soft palate, and cricothyroid
Fifth arch disappears		
Sixth arch	Recurrent laryngeal nerve	Muscles of larynx (except cricothyroid)

Derivative of Pharyngeal Pouches

First pouch: Derivatives of the first pouch are as follows:
❑ The ventral part of first pouch is obliterated because of the formation of tongue. The posterior third of the tongue is formed from the third pharyngeal arch.
❑ Its dorsal part along with the dorsal part of second pouch forms a diverticulum called the tubotympanic recess. The tubotympanic recess expands at its distal part to form the cavity of the middle ear while its proximal part forms the auditory tube (also known as the Eustachian tube), which connects the middle ear with the pharynx. External auditory meatus arises from the cleft of first pharyngeal arch.

Second pouch: The ventral part of the second pouch forms the tonsil. Thyroid gland also develops from the first and second pharyngeal pouches.

Third pouch: It gives rise to thymus and inferior parathyroid glands.

Fourth pouch: It gives rise to superior parathyroid glands. It may also contribute to the formation of some part of the lateral lobes of the thyroid gland.

(Third and fourth pharyngeal arches also form a complex structure known as "ultimobranchial body").

Fifth pouch: The 5th pharyngeal pouch is considered part of the fourth and is the last to develop. It gives rise to the ultimobranchial body which in the adult gives rise to the C cells of the thyroid which secretes calcitonin. Therefore for all practical purposes, pouch of fourth pharyngeal arch gives rise to superior parathyroid glands and C cells of the thyroid gland.

Development of Central Nervous System

Development of the central nervous system starts at the beginning of the 3rd week. The characteristic event occurring during the 3rd week is the appearance of primitive streak at the caudal end of the embryonic disc. The primitive streak has a primitive node at its cephalic end on the ectoderm. From the primitive node, a rod of cells grows out between the ectoderm and the endoderm known as the head process or the notochordal process. Changes occur in the notochordal process which convert this process into a plate and then again into a rod-like notochord.

Notochord is a supporting rod running dorsally through most of the length of animals belonging to the phylum chordate. The notochord consists of a solid rod of cells situated on the ventral aspect of the neural tube. It lies at first between the neural tube and the endoderm of the yolk-sac, but soon becomes separated from them by the mesoderm. While the notochord induces the formation of neural tube, it itself does not become the neural tube or crest. Most of the notochord disappears with the formation of the vertebrae but between the bodies of the vertebrae, the notochord remains in form of the nucleus pulposus of the intervertebral disc. The ectoderm lying on the dorsal (amniotic side) gets thickened to form the neural plate. This soon gets converted into neural groove by the formation of neural folds. The margins of the neural folds come to fuse with each other to form a neural canal having anterior and posterior neuropores which ultimately get closed **(Fig. 7.29)**. The anterior neuropore closes at the 18–20 somite stage on day 25. The posterior neuropore closes on day 27.
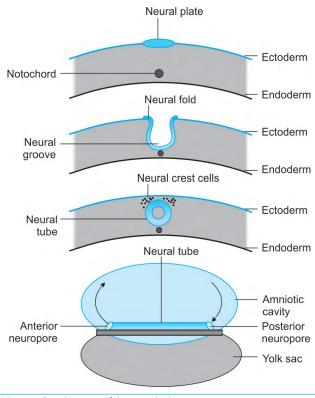
FIG. 7.29: Development of the neural tube

A dilatation converts the neural tube into a brain vesicle at the cephalic part while the caudal part of the tube forms the spinal cord. The brain vesicle further gets divided into three vesicles from the cephalic to the caudal side namely the prosencephalon (forebrain), the mesencephalon (mid brain) and the rhombencephalon (hindbrain).

Development of Foetal Heart

In the embryo, formation of the heart begins in the pharyngeal or the throat region. The first visible indication of the embryonic heart occurs in the undifferentiated mesoderm. The foetal heart arises from cardiogenic mesoderm and is formed by fusion of endocardial tubes. Pericardial area is formed in the embryonic disc and is situated cephalic to the prochordal plate (future oral membrane). It has also been shown that due to the formation of head fold the pericardium comes to lie caudal to the oral membrane and its surfaces get reversed.

The primitive atrium and sinus venosus lie outside the caudal end of the pericardial sac. Two endocardial tubes develop dorsal to the pericardial cavity. These tubes develop "in situ" and are the very early heart tubes. The term "in situ" implies that the structure develops at the place where it is found. These tubes have a cephalic (arterial) and a caudal (venous) end. The adjoining surfaces of these heart tubes start fusing with each other very soon. The fusion starts from the cephalic end and progresses caudally. At the caudal end, however, the fusion is not complete so that the heart now has one arterial end and two venous ends.
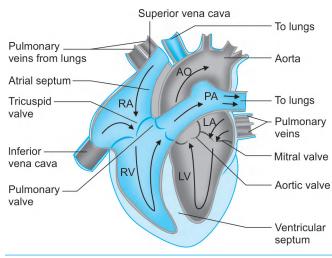
FIG. 7.30: Adult circulation
Abbreviations: RA, right atrium; LA, left atrium; RV, right ventricle; LV, left ventricle; AO, aorta; PA, pulmonary artery

At this very early stage, the heart tube starts arrhythmic contractions causing agitation of the blood inside. The blood just moves "to and fro". Blood begins to circulate through the primitive cardiovascular system and into chorionic villi at about 21 days. The heart starts to beat at day 22, but the circulation does not start until days 27 to 29. Cardiac pulsation is present by the 4th week after fertilisation.

The pulmonary circulation in the foetus is of lower volume than the systemic circulation.

The heart tube shows a few dilatations dividing it into the following parts from the cephalic to caudal end: (1) Bulbus cordis, (2) ventricular chamber and (3) sinoatrial chamber.
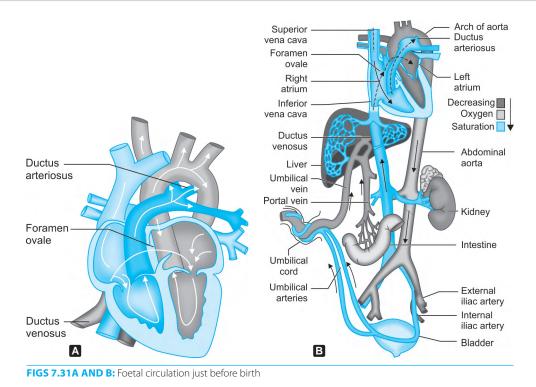
Foetal Circulation

Adult Circulation (Fig. 7.30)

In normal adults, the deoxygenated blood from upper and lower parts of the body enters the right atrium via superior vena cava and inferior vena cava (IVC), respectively. The blood from right atrium enters the right ventricle via atrioventricular valves. The deoxygenated blood from right ventricle flows through the pulmonary artery to the lungs where it gets oxygenated. This oxygenated blood from lungs moves to the left atrium via the pulmonary veins. The oxygenated blood from left atrium moves to the left ventricle and is then distributed to the whole body via aorta and its branches.

Foetal Circulation

Some of the anatomic differences between the foetal and adult circulations were described by Harvey in 1628. Foetal circulation is characterised by presence of three shunts: ductus venosus, foramen ovale and ductus arteriosus. These shunts permit the blood to bypass the liver and lungs, and

FIGS 7.31A AND B: Foetal circulation just before birth

shunt the most oxygenated blood from the right to the left side of the heart **(Figs 7.31A and B)**. These shunts disappear following the birth of the baby. In the foetal circulation, there are two umbilical arteries and one umbilical vein. The umbilical artery is a bilateral structure and is a branch of the anterior division of the internal iliac artery. It ascends out of the pelvis along the anterior abdominal wall and joins the umbilicus. In utero, the umbilical arteries carry deoxygenated blood from the foetus to the placenta. The inferior gluteal artery is another branch of the anterior division of the internal iliac artery, which supplies blood to the skin over the buttocks. Umbilical artery cannulation may therefore interfere with this supply, leading to the development of ischaemia in this area.

Also, in the foetus the lungs are not fully developed, therefore exchange between the oxygenated and deoxygenated blood does not take place in the lungs. Rather it takes place in the placenta. Therefore, the foetal circulation differs from adult circulation in the following ways: the deoxygenated blood from the hypogastric arteries, which are the direct continuations of the common iliac artery, moves into the two umbilical arteries. The umbilical arteries carry the deoxygenated blood from the foetus to the placenta. The deoxygenated blood in the foetus moves from the internal iliac arteries into two umbilical arteries that transport the deoxygenated blood back to the placenta. These arteries on reaching the placenta form numerous branches and enter the chorionic villi, where the exchange with oxygenated blood carried by maternal endometrial arteries takes place. The accompanying branches of umbilical veins in the chorionic villi, which carry the oxygenated blood, drain into umbilical vein, which carries the oxygenated blood to the foetus from placenta.

The foetus receives oxygenated blood from the mother through placenta in the form of umbilical veins. As this oxygenated blood bypasses a shunt called ductus venosus, some of the oxygenated blood goes to the liver, but most of it bypasses the liver and empties directly into the IVC. In the IVC, there is oxygenated blood from umbilical veins along with deoxygenated blood returning from the lower extremities, pelvis and kidneys. Ductus venosus is formed on the posteroinferior aspect of the liver by the union of the left umbilical vein and the left branch of the portal vein. The portal vein normally carries the deoxygenated blood. The ductus venosus provides a means for oxygenated umbilical vein (umbilical arterial blood has a lower PO_2) blood to bypass the sinusoids of the liver.

While the deoxygenated blood from the lower parts of the body is drained into the right atrium via the IVC, the deoxygenated blood from the upper parts of the body is drained via the superior vena cava into the right atrium. This less well-oxygenated blood from the right atrium enters the right ventricle and then enters the aorta via the ductus arteriosus distal to the left subclavian artery. The less well-oxygenated blood is therefore diverted to the trunk and lower body of the foetus.

The separation of the blood according to its oxygen content in the foetal heart and in the IVC is facilitated by the process called streaming, in which well-oxygenated blood travels along the medial side of IVC and less oxygenated along the lateral wall. The less oxygenated blood (lateral stream) is sent to the right ventricle, whereas the more oxygenated blood is shunted to the left atrium via foramen ovale and ultimately supplies the foetal heart and brain. Thus it is ensured that the better oxygenated blood from placenta supplies the more important organs (e.g. heart

and the brain). As the blood from the IVC enters the right atrium, a large proportion of it is shunted directly into the left atrium through an opening between the left and right atria called the foramen ovale. Thus the oxygenated blood returning from the placenta goes from the right atrium to the left atrium via the foramen ovale.

A small valve called the septum primum, located at the atrial septum, prevents blood from moving in the reverse direction. The oxygenated blood in the left atrium mixes with a small amount of deoxygenated blood returning from the lungs (by means of pulmonary veins), and then enters the left ventricle and ascending aorta. As the oxygenated blood from left atrium moves into the left ventricle and from there to the ascending aorta, the myocardium and brain are thereby supplied with the most oxygenated blood since the coronary and carotid arteries are the first to branch from the ascending aorta, before there is too much mixing with desaturated blood from other areas of the foetal heart.

Though in the normal adult heart, all the blood from right atrium moves into the right ventricle and from there through pulmonary arteries to the lungs, in the foetal heart, some, but not all blood flows from the right atrium to the right ventricle and then through the pulmonary artery to the lungs. Since the lungs of the foetus are not functioning to oxygenate, only a small amount of blood is required to go there for its adequate growth and development. There is high resistance in the pulmonary vessels that forces most of this blood to flow through the structure called ductus arteriosus into the descending aorta. The ductus arteriosus empties the oxygenated blood from the pulmonary artery into the aorta. Here it mixes with the blood from the proximal aorta to supply blood to the lower body.

Foetal Circulation outside the Uterine Cavity

As the foetus is delivered out of the uterine cavity and the umbilical cord is clamped, the placental circulation ceases and the baby's lungs become functional. Thus, following the birth of the baby, two major changes take place:

1. *Decreased resistance of pulmonary vascular system following expansion of lungs*: At the first breath, air fills the lungs and pulmonary vascular resistance falls. This reduces the pressure in the pulmonary artery and the right side of the heart. Blood flow to the lungs increases from 10% to 50% of cardiac output, and the increased pulmonary venous return raises left atrial pressure.
2. *Increased systemic vascular resistance*: Ligation of the umbilical cord increases systemic resistance. Increased systematic vascular resistance occurs due to breaking off from low resistance placental circulation. Therefore, there is an increase in the pressure on the left side of the heart. This causes blood to be shunted from left side of the heart to the right. Thus heart starts working in series. This has the following consequences:

- *Flap closure of foramen ovale*: The foramen ovale, which acts as a flap valve, closes due to increase in left atrial and decrease in right atrial pressure.
- *Functional closure of the ductus arteriosus*: The ductus arteriosus constricts due to exposure to increase PO_2, thereby preventing the blood from the aorta and pulmonary artery from mixing. Though the functional closure occurs within 28–48 hours of birth, permanent (anatomic) closure takes about 2–3 weeks for completion and results in the formation of a structure called ligamentum arteriosum.
- *Functional closure of ductus venosus*: The increase in systemic vascular resistance causes the ductus venosus to close. After birth, the blood within the umbilical vein clots and this extends to the ductus venosus.

The hypogastric arteries on atrophy become umbilical ligaments and the umbilical vein forms the ligamentum teres **(Table 7.4)**. The ductus venous closes and forms the ligamentum venosum.

TABLE 7.4	Structure in the foetus and the corresponding rudimentary structure at birth	
Structure in the foetus	**Rudimentary structure at birth**
Hypogastric arteries	Umbilical ligaments
Umbilical vein	Ligamentum teres
Ductus venosus	Ligamentum venosum
Ductus arteriosus	Ligamentum arteriosus

Formation of Foetal Blood Cells

Foetal haemoglobin concentration is high varying between 17 to 19 g/dL. Initially, nucleated red blood cells (RBCs) are made in the yolk sac from about the 3rd week of life. By 4th week, the endothelial cells make non-nucleated RBCs. Since the development of bones and the bone marrow occurs later during the process of foetal development, the liver functions as the main haematopoetic organ in the foetus. The liver begins production of blood cells at around 6 weeks of development. From 3rd month onwards, the bone marrow is the major source of blood cells. Spleen and lymphoid tissues have hardly any role in the process of haematopoiesis in a normal healthy foetus.

Foetal Blood

Glucose levels in foetal blood are usually 20–30% lower than in maternal blood, due to its rapid metabolism by the foetus. CO_2 levels in the maternal blood are usually around 40–45 mm Hg compared to 48 mm Hg in the foetus. Amino acids and Ca^{2+} ions are absorbed actively by the placenta. Therefore, their concentration is high in the foetal blood. Urea levels are only slightly higher in the foetus.

Choose the Single Best Answer (SBA)

Q 1. Which of the following is not true regarding mitosis?
A. Non-disjunction increases with decreasing maternal age
B. The chromosomes start occupying the equatorial plane of the mitotic spindle during metaphase
C. Lag during the anaphase can result in numerical chromosomal aberrations
D. Division of the chromatids in an abnormal plane result in production of isochromosomes
E. Two independent daughter chromosomes are formed during the anaphase

Q 2. Which of the following is true regarding meiosis?
A. Meiosis II is a reductional division
B. Exchange of paternal and maternal DNA takes place in meiosis II
C. Separation of whole chromosomes occur during meiosis II
D. Only one round of DNA replication occurs during two successive phases of meiosis
E. None of the above

Q 3. Which of the following statement regarding spermatogenesis is correct?
A. Secondary spermatocytes contain 23 chromosomes
B. The seminiferous tubules contain motile spermatozoa
C. The testes descend through the inguinal canal after birth
D. Normal testis development requires only the presence of the SRY gene
E. Sperms are stored in epididymis prior to ejaculation

Q 4. Which of the following is not true regarding spermatogenesis?
A. Dephosphorylation of adenosine triphosphate (ATP) is required as the source of energy
B. The process takes approximately 64 days for completion
C. FSH stimulates the process of spermatogenesis
D. Testosterone is required for the process of spermatogenesis to proceed
E. Each diploid spermatocyte gives rise to four spermatids

Q 5.Which of the following is true concerning the male androgen (testosterone)?
A. Is a polypeptide
B. Is not required for the development of the Wolffian duct in the male
C. Induces the development of male secondary sexual characteristics
D. Systemic administration of testosterone stimulates spermatogenesis
E. It is secreted by the adrenal medulla

Q 6. Which of the following is true regarding the development of the female genital tract and genitalia?
A. Sexual differentiation of the external genitalia is complete by 10 weeks
B. The clitoris is derived from the genital tubercle
C. The external genitalia only change under the influence of oestrogens produced by the placenta
D. The urogenital sinus forms the lower 1/3 of the vagina
E. The uterus and upper 2/3 of the vagina are derived from the paramesonephric ducts

Q 7. Which of the following is true concerning the Müllerian ducts?
A. Are derived from coelomic epithelium
B. Form the vas deferens
C. Form urogenital sinus in their lowest part
D. Fuse from above downwards
E. Grow medial to the Wolffian ducts

Q 8. Which of the following is true concerning the development of the genital system in a female?
A. The paramesonephric ducts develop from coelomic epithelium on the urogenital ridge
B. The sex of the embryo is determined at the 7th week of development
C. Male embryos have only the mesonephric duct whereas the female embryos have only the paramesonephric ducts
D. Mitosis in oogonia is not completed by the end of the first year of life
E. The ovary develops in the medulla of the primitive gonad

Q 9. Which of the following is true concerning gonadal development in a male?
A. Primary sex cells (gonocytes) have a haploid number of chromosomes
B. The histodifferentiation of the testis begins later than that of the ovary
C. The histological appearance of the primitive gonad is similar in both sexes until 70 days after fertilisation
D. The paramesonephric duct totally disappears in a male
E. The interstitial cells of Leydig are derived from mesenchyme

Q 10. Which of the following structures do not arise from the Wolffian ducts?
A. Epididymis
B. Epoöphoron
C. Gartner's duct
D. Paroöphoron
E. Round ligament

1. A   2. D   3. A   4. B   5. C   6. B   7. A   8. A   9. E   10. E

Q 11. **Which of the following statement is not correct regarding development of the genitourinary system?**

A. The fallopian tubes are derived from the Müllerian ducts

B. Double uterus may result from failure of fusion of the Müllerian ducts

C. The mesonephric and paramesonephric ducts coexist in all embryos up to 8 weeks of gestation

D. Müllerian inhibiting substance (MIS) is secreted by the Leydig cells of the testes

E. Development of the testes is determined by the presence of SRY gene

Q 12. **Which of the following statement is correct regarding the development of genitourinary system?**

A. Ova originate outside the developing gonad

B. Sertoli cells are derivatives of primordial germ cells

C. The external genitalia do not differentiate sexually before the 12th week of gestation

D. The Müllerian duct is an invagination of ectodermal tissue

E. Testosterone determines the development of male external genitalia

Q 13. **Which of the following statements regarding the development of the urinary system is not correct?**

A. The ureteric bud divides repeatedly to form successive generations of collecting tubules which in turn form the major calyces, minor calyces and finally the collecting tubules of the kidney

B. The kidneys attain their adult position during the 20th week of foetal life

C. Urine formation begins at about the 3rd month of foetal life, and continues increasing volume till term

D. The mature foetus may well void 450 mL of urine daily into the amniotic cavity

E. The urinary bladder is derived in part from the urogenital sinus, and in part from the ends of the mesonephric ducts

Q 14. **During the development of the urinary system which of the following statement is true?**

A. The ureteric bud gives rise to the collecting tubules of the metanephros

B. Initially the hilum of the metanephros faces dorsally

C. The metanephros becomes functional at birth

D. The mesonephros completely disappears

E. The trigone of the bladder is derived from the mesonephric ducts

Q 15. **Which of the following muscles develops from the second pharyngeal arch?**

A. Anterior belly of digastric

B. Posterior belly of digastric

C. Temporal muscle

D. Muscles of mastication

E. Mylohyoid

Q 16. **Which of the following statement is not correct regarding the fifth pharyngeal pouch?**

A. It gives rise to the C cells of the thyroid gland

B. It gives rise to the ultimobranchial body

C. C cells produce calcitonin in later life

D. May be considered part of the fourth pouch

E. It is the first to be developed

Q 17. **Which of the following is true concerning the foetal developing heart?**

A. It starts contracting at 3 weeks gestation

B. The dorsal mesocardium breaks down to form the transverse pericardial sinus

C. The sinus venosus is at its caudal end

D. All the above

E. None of the above

Q 18. **Which of the following is not true regarding the embryological development of the thyroid gland?**

A. Develops from the endoderm between the second and third pharyngeal pouches

B. Has C cells that are derived from the ultimobranchial body

C. It usually weighs about 25 g

D. Is at the level of the fifth to seventh cervical and first thoracic vertebrae

E. May have accessory nodules in the tongue

Q 19. **Which of the following is true concerning the notochord?**

A. It becomes the neural crest

B. It becomes the neural tube

C. It fuses temporarily with the endoblast of the yolk sac

D. It induces formation of the neural tube

E. It is a hollow structure as it forms

Q 20. **Regarding the development of the central nervous system, which of the following is correct?**

A. Development starts at the beginning of the 3rd week

B. The anterior neuropore closes at day 27

C. The anterior neuropore closes at the 10–12 somite stage

D. The neural plate is derived from endoderm

E. The posterior neuropore closes at day 29

Q 21. **Which of the following tissues is paired with the appropriate primary germ cell layer of origin?**

A. Endometrium—mesoderm

B. Pineal gland—ectoderm

C. Tongue epithelium—mesoderm

D. All the above

E. None of the above

---

11. D   12. A   13. B   14. A   15. B   16. E   17. D   18. A   19. D   20. A   21. B

Q 22. Which of the following is not correct regarding peritoneal ridges?
- A. The lateral umbilical ligaments are the obliterated umbilical arteries
- B. The lateral umbilical ligaments pass from the internal iliac arteries to the umbilicus
- C. The medial umbilical ligament is also called the urachus
- D. The urachus extends from the bladder to the umbilicus
- E. The urachus is the remains of the foetal allantois

Q 23. "A neonate is admitted to the hospital at 14 days of life with the complaints of failure to thrive, tachypnoea and difficulty in feeding. He is diagnosed as having a circulatory defect."

Administration of prostaglandin antagonists soon after birth can be used for therapeutic closure of which patent structure of foetal origin?
- A. Ductus venosus
- B. Foramen ovale
- C. Ductus arteriosus
- D. Fossa ovalis
- E. Ligamentum venosum

Q 24. At birth which of the following changes occur in the foetus?
- A. Pulmonary vascular resistance decreases
- B. The aortic pressure decreases
- C. The left ventricular pressure decreases
- D. The right atrial pressure increases
- E. The right ventricular pressure increases

Q 25. Which of the following is not true regarding the foetal circulatory system?
- A. Blood flows from the foetus to the placenta in the umbilical arteries
- B. The ductus arteriosus closes during labour
- C. Placental circulation starts at about 1 week after implantation
- D. The heart becomes a four-chamber organ at about 7 weeks' gestational age

- E. Reversed end diastolic flow in the umbilical artery is associated with fetal hypoxia

Q 26. In the foetal cardiovascular system, which of the following is not correct?
- A. Cardiac pulsation is present by the 30th day after fertilisation
- B. Oxygenated blood is transferred to the left atrium through the foramen ovale
- C. The ductus arteriosus closes during the last 4 weeks of pregnancy
- D. The heart arises from mesoderm
- E. The heart is formed by fusion of endocardial tubes

Q 27. Which of the following circulatory changes occur at the time of birth?
- A. A 20-fold increase in lung blood flow
- B. A rise in right atrial pressure
- C. Anatomical closure of the ductus arteriosus
- D. Flap closure of the foramen ovale
- E. Anatomical closure of the ductus venosus

Q 28. Which of the following statement regarding the foetal circulation is correct?
- A. The ductus arteriosus carries blood to the pulmonary arteries
- B. The ductus venosus empties into the inferior vena cava
- C. The foramen ovale connects the right and left ventricles
- D. The portal vein contains oxygenated blood
- E. There are two umbilical veins

Q 29. Which of the following is true regarding the ductus venosus?
- A. Carries blood with a higher PO_2 than umbilical arterial blood
- B. Gives rise to the ligamentum teres
- C. Is a shunt preventing blood from passing to the foetal lungs
- D. Is derived from the anterior cardinal vein
- E. Is part of the embryonic heart

22. C   23. C   24. A   25. B   26. C   27. D   28. B   29. A

CHAPTER **8**

Genetics

Deoxyribonucleic Acid

Deoxyribonucleic acid (DNA) is a nucleic acid containing the genetic information responsible for the development and functioning of all living organisms [the only exception being the ribonucleic acid (RNA) viruses]. DNA is double stranded in all mammalian cells, whereas RNA is single stranded. In some viruses, the DNA can be single stranded. The DNA segments carrying this genetic information are known as genes. The DNA from a single cell is nearly 2 metres long. The most basic structural unit of DNA is a nucleotide. Each nucleotide sub-unit comprises of a phosphate, deoxyribose sugar and one of the four nitrogenous bases. Nucleoside, on the other hand is a combination of a base with sugar DNA (deoxyribose).

Each DNA molecule comprises of two nucleotide strands intertwined with each other and running in opposite direction of one another (anti-parallel arrangement). One strand runs in 3'–5' direction, while the other strand runs in 5'–3' direction. In the backbone, joining the two strands are sugars and phosphate groups which are joined by ester bonds. The sugar in DNA is deoxyribose type, while in RNA it is ribose type. Attached to the sugar moiety are the molecules called bases. Both DNA and RNA are composed of four nitrogenous bases. However, the types of bases may be different. In DNA, the four nitrogenous bases are thymine, adenine, guanine and cytosine, abbreviated as T, A, G, and C, respectively. The nitrogenous bases in RNA are uracil (instead of thymine), and adenine, guanine and cytosine (same as that in DNA). They are abbreviated as U, A, G and C, respectively. Of these nitrogenous bases, adenine and thymine are purine bases whereas cytosine and guanine are pyrimidine bases. Adenine always pairs with either uracil in RNA or with thymine in DNA. On the other hand, cytosine always pairs with guanine. Purines are metabolised to xanthine through a reaction involving xanthine oxidase.

The bases lie stacked on each other 3.4 angstroms apart. Hydrogen bonds almost always form between an adenine base on one strand and a thymine on the other strand and between a cytosine base on one strand and a guanine base on the other. As a result the number of adenine and thymine bases as well as the number of guanine and cytosine bases in a given double helix is equal. The bond between adenine and thymine comprises of a double bond, whereas that between guanine and cytosine comprises of triple hydrogen bonds. The hydrogen bonds between the strands of the double helix are weak enough that they can be easily separated by enzymes known as helicases. Moreover, the two strands of the DNA helix denature and separate when heated to about 94° C for 5 minutes.

Long strands of DNA which are elaborately folded form the chromosomes. In eukaryotic cells, most of the DNA is stored inside the nucleus, while some may also be present in the organelles such as mitochondria. In the eukaryotic cells, the DNA material is packaged in form of chromatin. On the other hand, in prokaryotic cells, the DNA is present mainly in the cytoplasm. Prior to the cell division, the chromosomes are duplicated in a process called DNA replication. The sequence of the four nitrogenous bases in the DNA encodes for the genetic information. Each of the trinucleotide unit formed from these nitrogenous bases, is known as a codon and codes for a single amino acid. For example, a codon (GGU), formed by three bases, guanine-guanine and uracil, codes the amino acid glycine. While a codon is a sequence of three base pairs, a further sequence of opposite base pairs on tRNA represents the anticodon. The codons code for all amino acids but also code for extra or nonsense sequences which may encode termination sequences. These are known as the stop codons and include the sequences UAA, UGA and UAG. There are 20 amino acids in all and each specific amino acid is coded by a specific codon. However, there may be more than one specific codon for a particular amino acid. Abnormality of trinucleotide repeats sequences

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are seen in the disorders such as Huntington's chorea, fragile X syndrome and myotonic dystrophy.

Process of Protein Synthesis

The information present in the DNA sequence of various genes is first transcribed and then translated into a protein through the process of transcription and translation, both of which are described next **(Figs 8.1A and B)**.

Transcription

The process of reading of these codes is known as transcription. In the process of transcription, a complimentary copy of RNA is created from a DNA sequence with the help of the enzyme RNA polymerase. Regulatory elements of gene transcription include the following:

❏ Promoters: regions of DNA to which RNA polymerase bind and initiate transcription
❏ Enhancer sequences: modify activity of genes on the same chromosome
❏ Transacting proteins: modify genes on both pairs of homologous chromosomes.

Messenger RNA (mRNA) is transcripts of DNA, which are attached to transfer RNA and then translated in ribosomes to form protein. These then undergo considerable post-translational changes.

Translation

If the gene encodes for the protein, the process of transcription is followed by the process of translation. Translation is a process in which the information in the mRNA is used to direct the synthesis of polypeptides on the ribosomes. The ribosomes read the sequence of mRNA and translate it into a sequence of protein composed of several amino acids. The ribosome reads three nucleotides at a particular time. This sequence of three nucleotides is known as a codon and has been described previously in the text.

Gene

A gene is a length of DNA that carries information to make a single peptide chain. However, it is estimated that only a little percent of DNA comprising the whole human genome consists of genes encoding protein.

Exons are segments of DNA molecule containing information coding for a protein or peptide sequences. Introns, on the other hand, are intervening sequences of unknown function in mammalian genes.

Mitochondrial DNA

Mitochondrial DNA is the genetic material present in the mitochondria, an important organelle of eukaryotic cells. The mitochondria are present in the tail of the
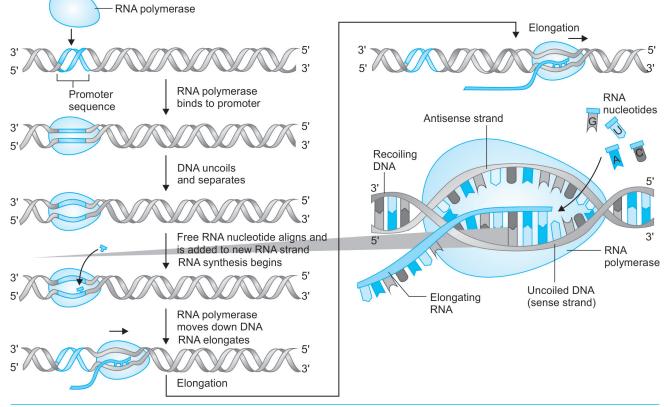
FIG. 8.1A: Process of protein synthesis: Transcription
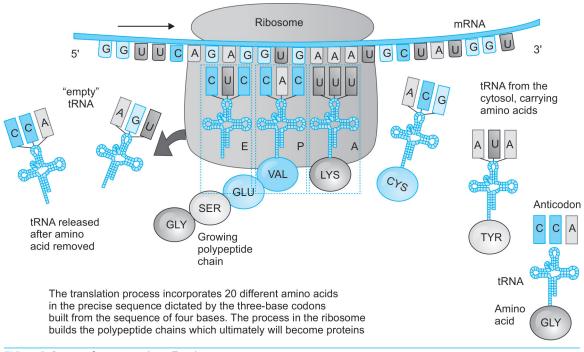
FIG. 8.1B: Process of protein synthesis: Translation
Abbreviations: GLY, glycine; SER, serine; GLU, glutamine; VAL, valine; LYS, lysine; CYS, cysteine; TYR, tyrosine

spermatozoa, but not transferred to the ovum and hence the mitochondrial DNA is only inherited from the mother. It is not inherited from the father. The mitochondrial DNA has their own genomes which code for certain genes, e.g. cytochrome oxidase enzymes. It contains 37 genes which are essential for normal mitochondrial function. They are expressed in neuronal tissue and other tissues rich in mitochondria. Therefore, their abnormalities result in the development of neurological syndromes, myopathies and cardiomyopathies. Some genetic conditions associated with mutations in the structure of mitochondrial DNA include, cytochrome c oxidase deficiency, Kearns-Sayre syndrome, Leber's hereditary optic neuropathy, Leigh syndrome, maternally inherited diabetes, deafness, etc. Mitochondrial inheritance is exclusively maternal, as none of the mitochondria from sperm survives fertilisation. Therefore, only daughters of an affected mother can transmit the trait related to the mitochondrial DNA.

Human Chromosomes

In the normal human there are 46 chromosomes, 22 pair of autosomes and 1 pair of sex chromosome. Each chromosome consists of two identical chromatids which are held together in the midline at the centromere **(Fig. 8.2)**. The short arm is known as p (after the word "petit" meaning "small") and the longer arm is labelled as q (simply because it is the next alphabet in the series after "p").

Q-banding is used to distinguish between chromosomes that are similar in size and shape. In this banding pattern, chromosomes are treated with quinacrine fluorescent
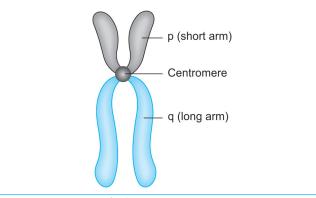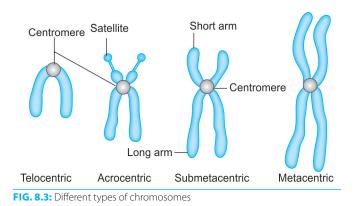
FIG. 8.2: Basic structure of a chromosome

stain, in order to identify specific chromosomal pattern and their structure. It is especially useful for distinguishing the Y chromosome. Banding patterns show genetic polymorphism and it is sometimes possible to trace foetal autosomes to a specific parent. Polymorphism can be defined as the occurrence of more than one morphological form in the population.

Depending upon the position of centromere and length of the two arms, the chromosomes can be classified as metacentric, submetacentric, acrocentric and telocentric **(Fig. 8.3)**. These chromosomes can be classified as follows:

Metacentric chromosomes: Metacentric chromosomes are those chromosomes which have a centrally placed centromere. In these cases the length of both the arms are equal.

FIG. 8.3: Different types of chromosomes

FIG. 8.4: Method of formation of isochromosomes

Submetacentric chromosomes: Submetacentric chromo-somes are those chromosomes where the centromere is slightly away from the centre. Therefore, the chromosome arm on one side is slightly longer than that on the other side.

Acrocentric chromosomes: In this type of chromosome the centromere is located closer to one end of chromosomal arm. Therefore, chromosomal arm on one side is very long and that on the other side is very short. On the side of the shorter chromatid, a small round structure called satellite, which is attached by a very thin thread may be observed. In the acrocentric chromosome, the p (short) arm may be sometimes so short that is hard to observe, but is still present. Acrocentric chromosomes seen in humans include chromosome numbers 13, 14, 15, 21 and 22. Acrocentric chromosomes are able to take part in Robertsonian translocations. This is a type of translocation in which the two chromosomes fuse at the centromere, giving rise to a translocation. Long arms of two acrocentric chromosomes fuse to form a single chromosome. The short arms also join to form a chromosome which is typically lost within a few cell divisions.

Telocentric chromosomes: Telocentric chromosomes are chromosomes in which the centromere is placed at the end of one arm. As a result, no segregation between the short and long arm is present. Telocentric chromosomes are not present in human cells.

Isochrome refers to an abnormal chromosome created as a result of mutation. In this chromosome, transverse rather than normal longitudinal splitting of replicating chromosomes occurs. As a result, two chromosomes having both identical arms are formed **(Fig. 8.4)**.

Human Sex Chromosomes

X Chromosome

Karyotype is the chromosomal composition of cells, including the total number of chromosomes and their structural appearance in the nucleus of an eukaryotic cell. Karyotype analysis is done after arresting the normally dividing cells in the metaphase stage of mitosis using a solution of colchicine. Each human cell has 22 pairs of autosomes, and one pair of sex chromosomes. This results in 46 chromosomes in all (44 + 2). The pair of sex chromosomes in females is XX, while that in males is XY. In human females, out of the two X chromosomes, only one is active, while the other is inactive and is present in form of a Barr body. Only one of the X chromosomes in the female undergoes inactivation randomly and this may be either the maternal or paternal X chromosome. Only one Barr body is present in normal female somatic cells, while no Barr body is present in normal male somatic cells. A single Barr body would be present in the individuals with Klinefelter's syndrome (47 XXY), even though these individuals are genotypically males. The individuals with Down syndrome have three No 21 chromosomes. The sex chromosomes are normal in these individuals. Thus the females would have one Barr body and the males would have none. Individuals with Turner's syndrome have XO karyotype. Thus they will have no Barr body. Individuals with testicular feminisation syndrome have a karyotype of XY. Therefore, there is no Barr body in these individuals. In rare cases of females with XXX karyotype, there are two Barr bodies. The Barr body is usually observed as a chromatin body in buccal smear. Sex chromatin is also found as a drumstick-shaped mass attached to one of the nuclear lobes in polymorphonuclear leucocytes in normal females.

The Lyon Hypothesis

Lyon hypothesis was described by Mary Lyon in 1962. The Lyon hypothesis relates to the fate of X chromosomes in the normal woman. It is no longer a hypothesis, but an accepted fact that one X chromosome is inactive in

every somatic cell. Germ cells are the exception, as both X chromosomes are required for manufacturing the egg. Therefore, Lyon's hypothesis does not relate to germ cells. Both the X chromosomes of maternal and paternal origin can be inactivated in each cell. According to the Lyon's hypothesis, only the terminal portion of the p arm of the X chromosome remains active. The process of inactivation occurs early in embryonic life, probably by day 16. The inactive chromosome is known as the Barr body and can be seen as a condensed dark mass of chromatin during interphase at the periphery of the nucleus. The Barr body is seen in up to 30% of cells on a buccal smear from a woman. It is also seen in neutrophils on a blood film in the form of "drumstick". The woman who is a carrier of an X-linked condition may show some of the features of the condition. In general, most of her somatic cells would suppress the abnormal X chromosome, but not all the cells may suppress the abnormal X chromosome. As a result, some of her cells will have the abnormal X chromosome as the active one, which may result in a few of the disease manifestations. In women with an extra X chromosome (XXX or triple X syndrome) only one X chromosome remains active and the remaining two become inactive, resulting in the presence of two Barr bodies. Presence of an extra X chromosome is usually associated with mental retardation.

Y Chromosome

Y chromosomes show the following features:

❑ Y chromosome contains the SRY gene on its short arm, which elicits the development of male gonad (testis). This can be considered as the gene of maleness, which specifies male gonads and male features. Genetic males having XY karyotype, but with a mutation or deletion of this SRY gene on the Y chromosome, will be phenotypically females despite the presence of most of the part of Y chromosome. Therefore, Y chromosome may be rarely detected in some phenotypic females showing the mutation of SRY gene. Moreover, the genetic females with XX karyotype, but also having a tiny piece of the Y chromosome with the SRY gene, will become male despite their female genotype.

❑ The Y chromosome is smaller than the X chromosome and is nearly half the size of X chromosome.

❑ No individuals having the karyotype YO have been identified even in case of aborted foetuses.

❑ Y chromosome demonstrates fluorescence with quinacrine.

❑ The normal Y chromosome is a submetacentric chromosome.

❑ Presence of hairy ears is associated with Y-linked inheritance pattern.

❑ Y chromosome similar to all other chromosomes has two arms **(Fig. 8.2)**, which are joined at the centromere. The long arm of Y chromosome has variable length.

Constitutional hirsutism just means that the women are just naturally hairy. Therefore the karyotype would be 46,XX. Individuals with testicular feminization syndrome are genetically male. Therefore, the karyotype is 46,XY. Individuals with Klinefelter's syndrome have a karyotype of 47,XXY. Due to the presence of Y chromosome, these individuals have a male gonad.

Sheehan's syndrome is where the pituitary is damaged by hypotension resulting from obstetric haemorrhage. Polycystic ovarian syndrome affects the ovary, therefore, it occurs in women having as karyotype of 46,XX. Hydatidiform mole is usually 46,XX. The genotype is typically 46,XX (diploid) due to subsequent mitosis of the fertilizing sperm. Therefore, the chromosomal material is all of paternal origin.

Individuals with MRKH syndrome or müllerian agenesis are genetically females. Thus, they have a 46,XX karyotype. Individuals with Down syndrome have trisomy 21 (i.e. three No 21 chromosomes), so they will have a karyotype of either 47,XX or 47,XY.

Principles of Inheritance

Autosomal Recessive Pattern of Inheritance

Autosomal recessive pattern of inheritance implies that an individual gets the disease, if he/she inherits two mutated genes, one from each parent. The disorders showing an autosomal recessive pattern of inheritance are listed in **Table 8.1**. Christmas disease and colour blindness show X-linked inheritance pattern. Neurofibromatosis shows an autosomal dominant inheritance pattern.

TABLE 8.1	Diseases showing autosomal recessive pattern of inheritance

- Alpha thalassaemia
- Beta thalassaemia
- Hurler's syndrome
- Sickle cell disease
- Adrenogenital syndrome
- Familial hypercholesterolaemia
- Mucopolysaccharidoses
- Phenylketonuria
- Tay-Sachs disease
- Cystic fibrosis
- Gaucher's Disease
- Wilson's disease
- Galactosaemia
- Homocystinuria
- Friedreich's Ataxia
- Hemochromatosis
- Familial Mediterranean Fever
- Alpha 1-antitrypsin deficiency
- 21-hydroxylase deficiency
- Galactosaemia
- Glycogen storage disease

Autosomal Dominant Pattern of Inheritance

The sex ratio for autosomal dominant conditions is usually 1:1. These conditions may only become apparent after 30 years of age. There is a 50% chance of the offspring of an affected individual acquiring the condition. Dominant inheritance means that if an individual has the abnormal gene, he/she gets the disease. As the person has two relevant genes, it implies that each of the person's progeny has a 50% chance of inheriting both the gene and the disease. The diseases showing an autosomal dominant pattern of inheritance are listed in **Table 8.2**. Thalassaemia major and galactosaemia show an autosomal recessive pattern of inheritance; whereas Vitamin D resistant rickets shows an X-linked inheritance pattern.

Co-dominant Inheritance

Co-dominant inheritance is one in which the two alleles are individually expressed. As a result, both the alleles of a gene pair in a heterozygote have full phenotypic expression. For example, the ABO blood group is co-dominantly inherited.

The ABO blood group system consists of three allelic genes: the A, B and O. There are six possible genotypes (OO,

AA, AO, BB, BO, AB), but the absence of a specific anti-O allows the serological recognition of only four phenotypes (O, A, B, AB). The universal donors blood is blood group O. The universal recipient is blood group AB. In the United Kingdom blood group O is the most common and AB the rarest. The frequency of the blood groups in UK is:

- ❑ O (46%)
- ❑ A (42%)
- ❑ B (9%)
- ❑ AB (3%).

The *A* and *B* genes control the synthesis of specific enzymes that are responsible for adding a carbohydrate to a glycoprotein or a glycolipid that has a terminal L-fructose (known as the H substance). The *O* gene is an amorph and does not transform the H substance. The A, B and H antigens are present in most body cells including white cells and platelets. In 80% of the population that have secretor genes, these antigens are also found in soluble form in body fluids and secretions (for example, saliva, sweat, plasma and semen).

X-linked Recessive Mode of Inheritance

X-linked recessive inheritance is a pattern of inheritance in which a mutant gene on X-chromosome causes the phenotype to be expressed in males because they have one X and one Y chromosome. On the other hand, the phenotype is expressed in only those females who are homozygous for the gene mutation. The diseases showing X-linked recessive pattern of inheritance are enumerated in **Table 8.3**. Adult polycystic kidney disease and Alzheimer's disease show an autosomal dominant inheritance pattern. Hurler's syndrome, on the other hand has an autosomal recessive mode of inheritance.

Polygenic Inheritance

There is no specific mode of inheritance associated with some diseases, e.g. diabetes mellitus. The inheritance is therefore considered polygenic—many genes contributing rather than one single gene defect.

Polygenic or multifactorial inheritance occurs in conditions such as

TABLE 8.2	Diseases showing autosomal dominant pattern of inheritance

- Achondroplasia
- Huntington's Chorea
- Familial hypercholesterolaemia (FH)
- Adult polycystic kidney disease (ADPKD)
- Retinoblastoma
- Neurofibromatosis
- Polyposis coli
- Multiple endocrine neoplasia (MEN) type 1
- Dystrophia Myotonica (variable penetrance)
- Spinocerebellar ataxia
- Familial adenomatous polyposis
- Hereditary non-polyposis colon cancer
- Familial breast and ovarian cancer
- Familial melanoma
- Basal cell nevus syndrome
- Factor V Leiden
- Hypertrophic cardiomyopathy
- Long QT syndrome
- Marfan's syndrome
- Primary Pulmonary hypertension
- Familial hypocalciuric hypercalcaemia
- Polycystic Kidney disease
- Neurohypophyseal diabetes insipidus
- Maturity onset diabetes of the Young
- Familial Parkinson's Disease
- Malignant hyperthermia
- Hypokalemic periodic paralysis
- Malignant hyperthermia
- Acute intermittent porphyria
- Alzheimer's disease (some cases)

TABLE 8.3	Diseases showing X-linked recessive inheritance pattern

- Both haemophilias A and B
- Glucose-6-phosphate dehydrogenase (G6PD) deficiency
- Colour blindness
- Duchenne's Muscular dystrophy
- Becker's Muscular Dystrophy
- Vitamin D resistant rickets
- Fragile X syndrome
- Kallmann's syndrome
- Adrenoleukodystrophy
- Nephrogenic diabetes insipidus (Autosomal recessive in some cases)

❏ Diabetes mellitus
❏ Rheumatoid arthritis
❏ Neural tube defects (NTDs)
❏ Cardiac defects, etc.

Variable Penetrance

Variable penetrance implies that a heritable trait is manifested with variable frequency by the individuals carrying the principal gene. This is seen in osteogenesis imperfecta, which is an autosomal dominant condition and is associated with varying penetrance. One parent, for instance, may just have blue sclera as a feature of the condition. Other may present with severe defects such as bone fractures or poor muscle tone.

Genetic Anticipation

The human genome is a dynamic apparatus, with the rearrangement of DNA sequences occurring as a normal mechanism which is susceptible to mutation. These tracts of DNA which show an increased susceptibility for mutation are known as unstable. This is responsible for the phenomenon of genetic anticipation. Genetic anticipation can be defined as the tendency of a genetic disorder to be expressed more severely or to become apparent at an earlier age as the disorder is passed on from one generation to the next. This phenomenon is usually observed with certain genetic disorders affecting the nervous system and include Huntington's disease, Myotonic dystrophy and fragile X syndrome. These disorders usually occur as a result of an unusual type of mutation known as a trinucleotide repeat expansion. A trinucleotide repeat is a sequence of three nucleotides, which may be repeated a number of times. DNA segments with abnormal number of these repeats are unstable and prone to development of aberrations at the time of cell division. As the gene is passed on from parent to child, number of these repeats is likely to get increased. This is known as trinucleotide repeat expansion. As a result of this expansion, the clinical features of these disorders become more severe with each successive generation as the cells divide within the body.

These trinucleotide repeat expansions can be classified into three types, with corresponding classes of phenotypes.

The first class is characterised by large expansions of a CGG trinucleotide (cytosine-guanine-guanine), leading to a fragile site in the chromosome. This site is associated with chromosome breakage under certain in vitro growth conditions. The prototype for this class is the fragile X syndrome (FRAXA), in which an expanded CGG repeat in the 5' untranslated region of the *FMR1* gene leads to under expression and a clinical phenotype of mental retardation, macro-orchidism, and other somatic changes in affected males.

The second class of disorder involves the relatively small expansion of an in-frame CAG (cytosine-adenine-guanine) repeat in the coding region of the respective genes, leading to a polyglutamine stretch in the resulting protein. Most of the disorders displaying this type of expansion show an autosomal dominant pattern of inheritance, e.g. Huntington disease.

The third class of disorder involving triplet repeat expansion is represented by the disorder myotonic dystrophy. In this case a CTG (cytosine-thymine-guanine) repeat in the 3'-{prime} untranslated region of the relevant gene is greatly expanded in affected individuals.

Anticipation results from the successive increases in repeat expansion and is observed to a lesser degree in the other classes of disorders. Facioscapulohumeral (FSH) muscular dystrophy (Landouzy-Dejerine disease) also shows the phenomenon of anticipation.

The genetic defect in autosomal dominant FSH muscular dystrophy is at the 4q35 locus.

Karyotypic Abnormalities

Karyotype abnormalities refer to the abnormalities related to the chromosomal composition and number. Karyotype of a normal female and a normal male is 46,XX and 46,XY, respectively. Non-dysjunction or the failure of replicated chromosomes to segregate during anaphase II of meiosis is responsible for many of the karyotype abnormalities resulting in an abnormality in the total number of chromosomes present in a cell (e.g. Down syndrome, Patau's syndrome, Turner's syndrome, Klinefelter's syndrome, etc.). Karytotypic disorders having an abnormal number of chromosomes (both autosomes and sex chromosomes) are known as aneuploidies. The majority of trisomies occur following a non-dysjunction event occur during the meiotic event. The most common trisomy encountered is trisomy 16. It is commonly found in the aborted foetuses because it is incompatible with life. The most common trisomy compatible with life is trisomy 21. Babies with trisomies 13 and 18 have considerably shortened life expectancy, with the majority not surviving infancy. Individuals having trisomy of X chromosome often remain undiagnosed and lead a near normal life. While loss of an autosome is not compatible with life, individuals with loss of a sex chromosome (e.g. Turner's syndrome) may survive; the chances of miscarriage are however increased in these cases.

Deletions, additions or translocations on the chromosomes resulting in structural aberrations are also responsible for producing karyotype abnormalities. Karyotype abnormalities are not as a result of single gene mutations (e.g., Marfan's syndrome, phenylketonuria, etc.) resulting in recessive or dominant genetic disorders. Acute lymphocytic anaemia and chronic myelocytic leukaemia are also associated with karyotype abnormalities because

the Philadelphia chromosome may be present in many of these cases of leukaemia. Philadelphia chromosome is the result of a reciprocal translocation between chromosome 9 and 22, and is labelled as t (9;22)(q34;q11). Cri du chat syndrome also results from karyotypic abnormality because it is associated with a deletion on the short arm of fifth chromosome. Some karyotype abnormalities associated with some common chromosomal disorders are listed in the **Table 8.4**.

Microdeletions

A microdeletion refers to the loss of a small subset of genes which are found adjacent to each other on a chromosome. Some of the syndromes occurring as a result of chromosomal microdeletions are listed in **Table 8.5**.

Genetic Carriers

The carrier of a genetic disease describes a person who is symptom-free or shows very mild manifestations of a medical disorder but is capable of passing the disease to their progeny. Genetically inherited autosomal diseases can be of two types: autosomal recessive and autosomal dominant. Autosomal dominant disorders would be manifested even if one autosome carries the defective gene. On the other hand, autosomal recessive disorders would be manifested only if both the autosomes carry the defective

gene. In case only one of the autosomes carries the defective gene, the person would be a carrier. Both males and females can act as carriers of autosomal recessive genetic diseases. Such carriers can pass the defective gene to their progeny through genetics. In case of X-linked recessive diseases, the female with one defective gene would be the disease carrier because there are two X chromosomes in females. Females with the defective genes on both the X chromosomes would only manifest the disease. However, males having even one defective gene would show the disease because they have only one X chromosome. Therefore, carrier state is present only in autosomal and X-liked recessive conditions and not in autosomal dominant conditions because in these cases presence of even one defective gene would result in a manifest disease. DNA studies can be used for detecting certain carrier states. In case of autosomal recessive disorders marriage between a normal unaffected male and an unaffected carrier female is likely to produce outcomes as depicted in **Figure 8.5**. In these cases, all the children would be unaffected, of which there is a 50% chance of being a carrier and 50% chance of being normal. **Figure 8.6** shows outcomes of marriage between an unaffected carrier mother and an unaffected carrier father. In these cases, there are 1 in 4 chances of giving birth to a normal child or an affected child. There are 2 in 4 chances of giving birth to unaffected carrier children.

In case of autosomal dominant disorders, marriage between a normal male and an affected female is likely to result in a progeny having 50% chances of showing

TABLE 8.4	Karyotype abnormalities in some common chromosomal disorders
Chromosomal disorder	**Karyotype abnormality**
Edward's syndrome	Trisomy 18
Patau's syndrome	Trisomy 13
Down syndrome	Trisomy 21
Turner's syndrome (gonadal dysgenesis)	XO
Klinefelter's syndrome	47,XXY
Testicular feminisation syndrome	46,XY
Cri du chat syndrome	Deletion on chromosome 5p

TABLE 8.5	Some common syndromes caused by chromo- somal microdeletions
Chromosomal disorder	**Chromosomal microdeletion**
Cri du chat syndrome	Deletion on the short arm of chromosome 5
Williams syndrome	Microdeletion affecting chromosome 7
Angelman syndrome	Microdeletion associated with the loss of maternally inherited chromosome 15
Prader-Willi syndrome	Microdeletion associated with the loss of paternally inherited chromosome 15
Smith-Magenis syndrome	Microdeletion affecting chromosome 17
Di-George syndrome, Shprintzen syndrome	Deletion of the proximal long arm of chromosome 22

FIG. 8.5: Pattern of inheritance of an autosomal recessive disorder when an unaffected carrier female marries a normal unaffected male
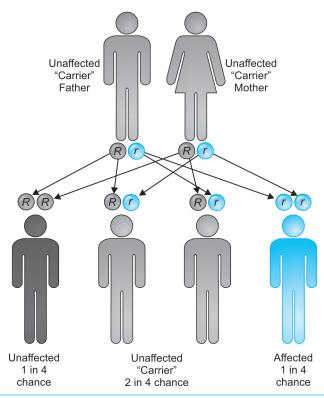
FIG. 8.6: Pattern of inheritance of an autosomal recessive disorder when an unaffected carrier female marries an unaffected carrier male

FIG. 8.7: Pattern of inheritance of an autosomal dominant disorder when an unaffected male marries an affected female

the manifest disease and 50% chances of being normal **(Fig. 8.7)**. In X-linked genetic diseases, the female carriers may show certain disease manifestations as a result of Lyon's hypothesis, which may result in the suppression of some but not all X chromosomes containing the abnormal gene. In case of X-linked recessive disorders, marriage between a carrier female and a normal male is likely to result in one in four chances of having one of the following: an affected male; a carrier female; an unaffected normal male and an unaffected normal female **(Fig. 8.8)**. Marriage between the blood relatives is associated with an approximately double the risk of getting inherited disorders (especially autosomal recessive disorders) and birth defects. In a consanguineous marriage, the risk of a developing a serious disease is double that for an unrelated mating. This risk is considerably reduced as the relationship between partners becomes more distant, so much so that second cousins have a risk equal to that of the background population.

Down Syndrome

Introduction

Down syndrome, also known as trisomy 21 is characterised by the presence of an entire or part of an extra chromosome 21. Trisomy 21 (three No. 21 chromosomes), resulting due to the presence of an extra chromosome No 21 is the most common cause of Down syndrome. This usually results

FIG. 8.8: Pattern of inheritance of an X-linked recessive disorder when an unaffected carrier female marries an unaffected normal male

due to non-dysjunction during meiosis. As a result, a gamete containing an extra copy of the chromosome 21 is produced. When this abnormal gamete combines with the normal gamete from the other parent, the embryo which is formed contains 47 chromosomes with three copies of chromosome 21. Another cause of Down syndrome could be mosaicism (46,XX, 47 XX+21) where some of the body cells are normal whereas others have trisomy 21. Robertsonian translocation can be another cause for the presence of an extra chromosome 21 material in the patient. In this case, the long arm of chromosome 21 gets attached to another chromosome, say chromosome 14. The incidence of Down syndrome has been estimated as 1 per 733 live births. However, this increases with the increasing maternal age (> 35 years).

Clinical Features

Some important clinical features of this syndrome are as follows:
❏ Some impairment in cognition and physical growth (which is usually lower than average)
❏ Moderate degree of mental retardation: average IQ of an individual with Down syndrome is 50, whereas that of a normal child is 100.
❏ The characteristic facial features in a patient with Down syndrome are as follows:
  • Broad head
  • Round face
  • Large protruding tongue (macroglossia)
  • Microgenia (abnormally small chin)
  • Almond shaped eyes (due to the presence of an epicanthic fold of the eyelid)
  • Up slanting palpebral fissures
  • Flat nasal bridge of the nose
  • Greyish-white spots on the iris (Brush field spots)
  • Shorter limbs resulting in a short stature
  • Hypotonia (or poor muscle tone)
  • Larger than normal space between the big and the second toe
  • Single transverse palmer crease
  • Single flexion furrow on the fifth finger
  • Large number of ulnar loop dermatoglyphs
  • Speech delay
  • Delay of fine motor skills

These individuals are an increased risk for the development of congenital heart defects, gastro-oesophageal reflux, recurrent ear infection, thyroid dysfunction and obstructive sleep apnoea.

Of the various congenital heart diseases, atrial septal defect is the most common type of defect which may be present in about 40% of the patients. The next most common type of defect is the ventricular septal defect. There is also an increased incidence of malignancies, particularly acute lymphoblastic anaemia in these patients. Moreover, these patients are also at an increased risk of other disorders such as thyroid disorders, Hirschsprung's disease, duodenal atresia, infertility, etc. Many patients may have strabismus and they are also at an increased risk of developing epilepsy.

Screening for Down Syndrome

Nuchal translucency refers to the most common of subcutaneous fluid at the foetal neck using ultrasound examination. Increased nuchal translucency is associated with numerous chromosomal abnormalities and structural defects such as cardiac defects, Down syndrome, Noonan's syndrome, Patau's syndrome, Turner's syndrome, etc. Nuchal translucency forms an important screening test for Down syndrome. Nuchal translucency can be used in combination with serum markers used in various combined tests such as triple test and quadruple test. Triple test involves the measurement of three blood parameters in the second trimester of pregnancy, namely alpha-fetoproteins, human chorionic gonadotropins and unconjugated oestriol levels. Alpha-fetoprotein is produced by foetal liver and yolk sac. Reduced levels of alpha-fetoprotein are observed in cases of Down syndrome, Edward's syndrome, etc. Low levels of alpha-fetoproteins, along with high levels of hCG are indicative of Down syndrome, while low levels of alpha-fetoproteins, along with low hCG levels are indicative of Edward's syndrome (trisomy 18). On the other hand, raised levels of alpha-fetoprotein are associated with neural tube defects such as spina bifida, anencephaly, etc. Quadruple test also involves the measurement of inhibin A levels along with that of the three other markers as done in cases of triple screening.

Ultrasound soft markers are commonly observed in a foetus with chromosomal anomalies. Choroid plexus cysts may be observed in foetuses with trisomy 18. Soft ultrasound markers indicative of Down syndrome include ecogenic bowel, mild renal pelvic dilatation, etc. Ecogenic bowel may also be observed in patients with cystic fibrosis.

Edward's Syndrome

Introduction

Edward's syndrome occurs due to trisomy of chromosome 18. Therefore, it is characterised by the presence of three copies of chromosome 18. It is the second most commonly occurring type of autosomal trisomy after Down syndrome. High incidence of this syndrome is associated with increasing maternal as well as increasing paternal age. Death usually occurs at 6 months of age due to presence of various congenital anomalies such as congenital heart malformations, e.g. ventricular septal defect, atrial septal defect, patent ductus arteriosus, omphalocele, oesophageal atresia, etc. Lung and urinary tract infections are also common amongst children suffering from Edward's syndrome.

Clinical Features

Edward's syndrome can be a cause of mental retardation and developmental delay amongst children. Some of the characteristic physical features which might be present in children suffering from Edward's syndrome are as follows:

❑ Microcephaly (small head) with a prominent occiput
❑ Low-set ears
❑ Micrognathia (abnormally small jaw)
❑ Cleft lip/palate
❑ Narrow palpebral fissures
❑ Upturned nose
❑ Ocular hypertelorism (widely spaced eyes)
❑ Ptosis
❑ Clenched hand with the index finger overlapping the middle finger or middle finger overlapping the ring finger
❑ Presence of 11 pair of ribs
❑ Rocker bottom feet (Dorsiflexion of big toes) or clubfoot
❑ Enlarged external genitalia
❑ Underdeveloped or absent thumbs or nails and/or absent radius
❑ Webbing of second or third toes
❑ Presence of choroid plexus cysts
❑ Presence of arthogryposis (multiple joint contractures) at birth
❑ Small breast bone or pelvis with limited movements of the hips

Patau's Syndrome

Introduction

Patau's syndrome is due to trisomy of chromosome 13. It is associated with motor and mental retardation, microcephaly, cleft lip/palate, polydactyly and cardiac defects.

Clinical Features

Various clinical features associated with Patau's syndrome are as follows:

❑ Polydactyly (presence of extra fingers or toes)
❑ Abnormalities in the eyes such as presence of colobomas in the iris, cataract, retinal detachment, microphthalmia, etc.
❑ Low-set ears
❑ Severe mental retardation
❑ Small head (microcephaly)
❑ Undescended testicles (cryptorchidism)
❑ Small lower jaw (micrognathia)
❑ Presence of a single umbilical artery
❑ Limb abnormalities
❑ Single palmer crease
❑ Hernias (umbilical or inguinal hernias)
❑ Clenched hands (with outer fingers on top of inner fingers)
❑ Holoprosencephaly
❑ Nearly 80% of the children die within first month of life

Klinefelter's Syndrome

Introduction

Klinefelter's syndrome is the most common genetic abnormality resulting in hypogonadism, infertility and or azoospermia in males. It is typically associated with low serum testosterone levels; elevated gonadotropin levels and thus hypergonadotrophic hypogonadism. It is a genetic abnormality characterised by the presence of an extra X chromosome resulting in the genetic constitution of 47,XXY. This usually occurs as a result of non-dysjunction during meiosis. Though the most common karyotype encountered in these individuals is 47,XXY, individuals with two extra X chromosomes (48,XXXY) and mosaics (46,XY; 47,XXY) have also been described.

Clinical Features

❑ The disease may affect nearly 1:600 males
❑ The IQ is less than that of siblings, but is not grossly impaired. It is not associated with severe mental retardation.
❑ It is characterised by immaturity, shyness and delayed acquisition of reading skills, etc.
❑ The testes are small and infertility occurs due to azoospermia.
❑ Gynaecomastia can occur.
❑ Hypogonadism can be treated with testosterone. Fertility treatment is possible with ICSI and then IVF even in the cases of azoospermia. Sperms in most men with Klinefelter's syndrome can be obtained via sperm retrieval, MESA (micro-epididymal sperm aspiration) or TESE (testicular sperm extraction).
❑ The gonads do not turn malignant in these cases. Therefore, there is no need to remove the rudimentary gonads
❑ Bone age may be delayed because the epiphyses fuse late. This usually occurs due to low testosterone levels and results in a tall stature
❑ The syndrome may be associated with an increased incidence of cancer of the breast and mediastinum, diabetes, varicose veins, and pulmonary disease

Testicular Feminisation Syndrome

Introduction

Testicular feminisation syndrome is also known as "androgen insensitivity syndrome". It is a genetic disorder resulting due to the abnormality of the testosterone receptors. It can be of two types: complete and incomplete. Numerous mutations of the androgen receptor gene can

result in this syndrome. The androgen receptor gene is located on the X chromosome (Xq 12). Therefore, complete androgen insensitivity syndrome follows an X-linked pattern of inheritance. As a result, a careful investigation for identification of other affected family members is essential.

The patients with this syndrome are male pseudohermaphrodites. This implies that the patient is genetically a male, i.e. has a male karyotype (46,XY). Since the karyotype is male, the patient has male gonads or testes. Pseudohermaphrodite implies that the external genitalia are opposite of the gonads. The individual is phenotypically a female. Since the tissues are insensitive to testosterone, the development of Wolffian ducts, which is induced by androgens, cannot proceed normally. On the other hand, the normal testes produce normal levels of anti müllerian hormone, which helps in suppressing the development of müllerian ducts. Therefore the uterus and fallopian tubes are typically absent. In the absence of the development of Wolffian ducts, the external genitalia develop along the female lines. Labia and clitoris may be slightly underdeveloped. The vagina may be blind and short or may be altogether absent. Therefore, the individual is phenotypically a female. Since the individual is genotypically a male, testes are present. However, they may be present at abnormal locations such as the abdomen or the inguinal canal. The gonads may show normal or an increased number of leydig cells with absent spermatogenesis. Malignant disease, usually a dysgerminoma, may develop in the abnormal testes, and it is therefore important to remove these after the development of secondary sexual characteristics and to administer oestrogen replacement therapy thereafter.

Due to unopposed action of oestrogens, breast development continues to be of female type and is enhanced. The overall body habitus is that of the female. In the individual with complete androgen insensitivity, serum testosterone concentrations are normal or only moderately increased, LH levels are increased and FSH levels are within the normal range. The increase in LH level is due to the resistance to the negative feedback effect of androgens at the level of pituitary gland. Oestrogen production (both oestrone and oestradiol) is increased by about 70% in comparison to that in the normal men. Although the serum testosterone levels are within the range of normal males, the production of dihydrotestosterone is decreased due to the absence of urogenital tissues, which are the primary sites of its production. The pubic and axillary hair is scant because testosterone is unable to exert its action.

Incomplete androgen insensitivity on the other hand is associated with a variety of disorders resulting from defects in the androgen action, which is usually less severe in comparison to those in patients with complete androgen insensitivity. In these syndromes, the Wolffian ducts are not completely absent but are present in an undeveloped pattern.

Clinical Features

Patients with complete androgen insensitivity syndrome usually present with the following:
- Primary amenorrhea
- Absent/scant pubic/axillary hair
- Short vagina
- Absent cervix and uterus
- Inguinal hernias or labial masses
- 46,XY karyotype
- Testosterone concentrations are in the normal male range but the receptor defect prevents the testosterone from exerting its effects
- Oestradiol concentrations are measurable and produced from the peripheral conversion of testosterone as well as by the gonads (through the action of aromatase).

The patients with androgen insensitivity need to be differentiated from those with müllerian agenesis on the basis of the features described in **Table 8.6**.

Treatment

Clinical treatment of patients with complete androgen insensitivity syndrome comprises of the following:
- Creation of a functional vagina through progressive vaginal dilatation and vaginoplasty
- Gonadectomy to prevent tumourigenesis in the cryptorchid testes.
- Oestrogen therapy may be started when gonadectomy is performed after puberty is complete or at the expected time of puberty if the gonads were removed prior to puberty. In the patients with complete androgen insensitivity syndrome, gonadectomy is best delayed until after the puberty is completed which is usually the age of 16–18 years. This is so because the pubertal development is likely to occur more smoothly in the face of endogenous hormone production. Also, the risk of development of tumours is quite low (about 5–10%), particularly before puberty.

TABLE 8.6	Differentiating between androgen insensitivity syndrome and müllerian agenesis	
Parameter	Androgen insensitivity syndrome	Müllerian agenesis
Pubic/axillary hair	Absent/scant pubic/axillary hair	Normal pubic/axillary hair
Testosterone levels	Within the range of normal males	Within the range of normal females
Karyotype	46,XY	46,XX

Müllerian Agenesis (Mayer-Rokitansky-Küster Hauser Syndrome)

Introduction

This may be the probable diagnosis in an individual with primary amenorrhoea and no apparent vagina. There is usually an absence or hypoplasia of the internal vagina and absence of fallopian tubes and uterus. The syndrome occurs due to defect in fusion of the müllerian ducts resulting in absence of proximal one-third of vagina with or without the uterus. Since the ovaries are not müllerian structures, they are normal.

The cause of this syndrome is unknown and is probably related to the mutations in the gene for anti-müllerian hormone or the gene for anti-müllerian hormone receptor.

Clinical Features

❏ Primary amenorrhoea and no apparent vagina
❏ Other anomalies including the renal tract anomalies such as ectopic kidney, renal agenesis, horse-shoe kidney and abnormal collecting ducts are frequently present. Extirpation of the müllerian remnants, if any present, is not required unless they are causing some problem such as fibroid growth, haematometra, endometriosis, etc.
❏ Pubic/axillary hair are normal

Treatment

Treatment of the condition usually involves progressive dilatation using Frank's dilators. Initially, the dilatation is begun in posterior direction and then after 2 weeks, it is changed to upwards direction in the line of vaginal axis. This must be performed daily for 20 minutes to the point of modest discomfort. By utilizing increasingly larger-sized dilators, a functional vagina can be created within a period of several months. Operative treatment is used in the patients in whom the Frank's method is unacceptable or fails. It is important for the gynaecologist to provide adequate reassurance and support in these cases. Adequate counselling helps in avoiding problems with altered body image, which are likely to develop in these cases. Creating an artificial vagina either through the use of Frank's dilators or surgical procedure (McIndoe's vaginoplasty) at the time the patient plans to get married, helps in ensuring that she and her partner would be able to obtain adequate sexual enjoyment following their marriage. Having regular sexual intercourse helps in maintaining the patency of newly created artificial vaginal orifice. Though the patient remains infertile, she can lead an almost normal life. Genetic offspring can be achieved by collecting oocytes from genetic mother, fertilising them with sperms obtained from genetic father and their placement in a surrogate carrier.

5-α Reductase Deficiency (5-ARD)

Introduction

In this anomaly, the main abnormality is the impaired conversion of testosterone to dihydrotestosterone (DHT), which prevents the virilisation of male genitalia during foetal development. The derivatives of Wolffian ducts (epididymides, vasa deferentia, seminal vesicles and the ejaculatory ducts) develop normally in response to the foetal testosterone levels. However, due to the deficiency of the enzyme 5-α reductase, which converts testosterone to dihydrotestosterone, the genital structures, which are derived from the urogenital sinus and genital tubercle (external genitalia, urethra and prostate), do not virilise normally. This is an autosomal recessive disorder characterised by a 46,XY karyotype.

Clinical Features

❏ Since DHT is required for the normal masculinisation of the external genitalia in utero, genetic males with 5-ARD are born with ambiguous genitalia (i.e., male pseudohermaphroditism). As a result, the external genitalia are predominantly female type at birth.
❏ The condition affects only genetic males (having a Y chromosome) because dihydrotestosterone has no known role in female development.
❏ The internal genitalia are male type, but empty into a shortened blind vagina. There may be a failed fusion of the labioscrotal folds and a urogenital sinus. There may be separate urethral or vaginal openings with or without clitoromegaly.
❏ The characteristic feature of this disorder is the virilisation of the affected individuals to varying degrees at the time of puberty.
❏ Most individuals are reared as females, although they may assume a male gender and behaviour at the time of puberty.
❏ Breast development is like that in normal males
❏ The features which the affected men may develop at the time of puberty include normal muscle mass, normal libido and deepening of voice. These features develop normally because they are derived from the action of testosterone.
❏ These individuals have reduced body hair, reduced temporal hairline recession and also reduced occurrence of acne because these features result primarily from the action of dihydrotestosterone.
❏ Serum hormone profile exhibits normal male serum testosterone concentration and an increased testosterone to dihydrotestosterone ratio.
❏ Testes may be present at abnormal locations such as inguinal canal, labia majora and scrotum and may exhibit impaired spermatogenesis.

❏ The clinical abnormalities of the disease range from individuals with normal male genital anatomy to underdeveloped male individuals with hypospadias to those with predominantly female external genitalia, most often with mild clitoromegaly. Since these patients have primary female characteristics, they are often raised as girls. At the time of puberty, these individuals often experience amenorrhoea and virilisation.

❏ In order to prevent virilisation later in life and development of tumours in cryptorchid testes, gonadectomy must be performed in these cases.

Treatment

Clitoromagaly can be surgically corrected. A functional vagina can be constructed with help of progressive vaginal dilatation. Vaginoplasty can also be performed in these cases. Oestrogens can be prescribed to induce and maintain feminine characteristics and can be started either at the time of puberty or immediately following gonadectomy in adults.

Turner's Syndrome

Introduction

This disorder results from the loss of an entire or a part of X-chromosome. Thus their karyotype is 45,XO. Nearly 50% of the patients may be mosaics (45,X, 46,XX). Only a part of the X chromosome may be missing in some patients. This may be associated with a number of structural abnormalities of X chromosome such as ring chromosome, isochromosome and terminal deletions. Most patients with Turner's syndrome experience the loss of X chromosome of paternal origin and retain the X chromosome of maternal origin. Since the Y chromosome is absent, this syndrome occurs only in the females. Although the uterus and vagina are present, there is complete amenorrhoea because no hormones are produced by the gonads. The ovaries are represented by streak gonads, which are characteristically composed of connective tissue with no follicles or only a few atretic follicles. In approximately one-third of the women, the ovaries can be imaged with the help of pelvic sonography. This syndrome occurs with a frequency of 1 in 2,500 live female births and can affect up to 3% of all conceptions. Approximately, 97–98% of the affected pregnancies land in a miscarriage and fail to reach term. At puberty there is no appearance of secondary sexual characteristics and the external genitalia and breasts remain infantile. Among these patients, the spontaneous conceptions are rare and if occur are associated with a high risk for sex chromosome aneuploidy and spontaneous abortions. There is a much higher incidence of hypothyroidism associated with Turner's syndrome although the cause for this is not completely known. It is supposed to be due to autoimmune causes.

Clinical Features

❏ Primary hypogonadism and presence of streak gonads

❏ *Primary amenorrhoea and delayed or absent pubertal development*: Though most patients with Turner's syndrome present with primary amenorrhoea and do not experience any pubertal development, some patients may develop normally and present with secondary amenorrhoea later in life. These patients may have experienced some amount of pubertal development.

❏ *Intelligence*: Intelligence usually remains unaffected. Normal intelligence is a typical feature of Turner's syndrome. Severe mental retardation may be present in a rare patient with small ring chromosome because the ring chromosome does not undergo X-inactivation. Attention deficit hyperactivity disorders and visual-spatial organization disorders are also commoner in patients with Turner's syndrome.

❏ *Cancer risk*: Though the overall risk of developing a cancer in a patient with Turner's syndrome is similar to that of general population, the incidence of CNS tumours, bladder cancer and endometrial cancer is increased. On the other hand, the incidence of breast cancer is decreased.

❏ *A characteristic phenotype*: A characteristic phenotype may be present, which includes the following features:
- Short stature: the pubertal growth spurt fails to occur and adult height is usually between 1.25 and 1.5 metres. The average height is approximately 1.42 metres. Short stature is the most commonly present abnormality, which is commonly present in virtually all the patients with this type of chromosomal abnormality.
- Female phenotype with webbed neck
- Wide carrying angle (cubitus valgus)
- Shield chest with widely spaced nipples
- A low occipital hairline
- A short fourth metacarpal and hypoplastic nails
- Low set ears may be present in up to 80% of cases
- It may be associated with cystic hygroma
- Infantile lymphedema: At birth there may be lymphoedema especially in the dorsum of the hand and foot.

❏ *Osteoporosis*: Osteoporosis is due to oestrogen deficiency. Osteoporosis is responsible for an increased incidence of bone fractures.

❏ *Congenital anomalies*: Turner's syndrome may be associated with some cardiac, renal or ocular abnormalities.
- *Cardiac anomalies*: The overall mortality rate is increased by 3-folds primarily due to the cardiovascular diseases. Some of the cardiac anomalies are as follows:
  – Aortic valve disease: This is the most common cardiac anomaly, which may occur in 20–30% cases and includes defects such as bicuspid aortic valve, aortic root dilatation, etc.

– Coarctation of the aorta: This is the most common serious cardiac anomaly, which may be found in up to 3–10% of the cases. Coarctation of aorta may be responsible for producing secondary hypertension and ejection systolic murmur.

– Other cardiac anomalies: Other associated cardiac anomalies, which may be sometimes present include, elongation of the transverse aortic arch, persistent left superior vena cava, anomalous pulmonary venous return and an aberrant right subclavian artery. Even young children with Turner's syndrome may have a prolonged QT interval.

- *Renal anomalies*: Approximately 30–50% patients with Turner's syndrome may have an associated renal anomaly. Horseshoe kidney is the most common anomaly amongst them.

- *Ocular anomalies*: Some individuals may have ocular abnormalities such as amblyopia, strabismus, ptosis, hypertelorism, epicanthus, red-green colour blindness etc.

- *Endocrine anomalies*: Endocrine abnormalities may include hypothyroidism and diabetes mellitus

- *Other anomalies*: Other abnormalities may include celiac disease, hearing loss and abnormality of liver function

❏ *Hypertension*: Hypertension is commoner in this condition; it may be idiopathic or secondary to some causes such as coarctation of aorta (described previously).

Treatment

Karyotype analysis is recommended for all girls with unexplained short stature, delayed puberty and other features suggestive of Turner's syndrome. Karyotype analysis must comprise of examination of at least 30 cells in order to detect significant mosaicism. Individuals having chromosomes of uncertain origin or those with any evidence of virilisation also must be evaluated using fluorescence in situ hybridisation (FISH) and Y-chromosome specific probes because those having all or part of a Y chromosome are at an increased risk of development of a gonadoblastoma. In these cases removal of the gonads may be required. Removal of gonads is usually not required unless the person is a mosaic having 46,XY karyotype.

Growth hormones must be prescribed to the patients as soon as the height falls below the fifth percentile for age. This usually occurs between the ages of 2 to 5 years. Early treatment with growth hormone can help in increasing the lean body mass, which helps the patient achieve a normal adult height. Oestrogen therapy initially started in the dosage of 0.25–0.5 mg micronised oestradiol and later increased to 2.0 mg of micronised oestradiol helps in achieving complete sexual maturity over a period of 2–3 years. Since oestrogen therapy reduces height velocity, its use is not recommended before the age of 13–14 years. Pregnancy may be possible in such patients given the option of oocyte donation. However, mortality risk during the pregnancy is increased mainly due to the complications of aortic dissection or rupture.

Noonan's Syndrome

Noonan's syndrome is an autosomal recessive disorder due to mutation of four genes which result in multiple organ defects. In Noonan's syndrome, infants are males but physical features resemble those found in Turner's syndrome.

Triple X Syndrome

Hypothetically, if a condition with one absent X chromosome is possible, a condition with three X chromosomes is also possible. These women are thought to have a karyotype of 47,XXX. However, this condition is usually not identified because most of these cases go undiagnosed. The main manifestation of this condition is that the woman's IQ may be lower than that of her siblings on the IQ scale by 10 points or more. There may be a tendency to immaturity. Though these women are fertile, there is an increased tendency for developing premature ovarian failure (POF). Goswami et al (2003) in their study found that nearly 3.8% of women with POF had triple X.

Chromosomal Abnormalities

Haemophilia A

Introduction

Haemophilia can be considered as a group of disorders characterised by abnormal coagulation. Haemophilia A is associated with the deficiency of factor VIII, while haemophilia B is associated with the deficiency of factor IX. Both these disorders are X-linked because they are more likely to occur in males in comparison to that in females. Haemophilia A is X-linked recessive disorder. Since women have two X chromosomes, they will manifest the disease only if they have the copy of abnormal genes on both the chromosomes. They will be a carrier in case they have the copy of abnormal gene on only one of the X chromosomes.

Since this is X-linked disorder, in order to suffer from the disease the father would be having a defective gene on the X chromosome. The carrier woman would be carrying a defective gene on one of the X chromosomes, the other X chromosome would be normal. The marriage between these two individuals is likely to result in 1 in 4 chances of

the following: giving birth to an affected daughter; giving birth to a carrier daughter; giving birth to a normal son and giving birth to an affected son.

It is possible for a woman to be affected with this disease if a carrier female marries an affected male. The people suffering from this disease are likely to require frequent blood transfusions. Blood transmitted diseases like hepatitis B or AIDS are likely to be screened nowadays. Therefore infections such as Hepatitis C or E, which can cause deranged liver function tests, are commoner. In this sickness due to the absence of clotting factors, the bleeding continues for a longer time. Levels of factor VIII related antigen is normal in these cases. Prenatal testing such as amniocentesis is available to the pregnant women who are the carriers of the disease.

Clinical Features

Even a minor injury can result in bleeding, which may last for days or weeks. In mild cases, which have 5–20% of the normal activity of factor VIII, bleeding may be controlled by DDAVP (desmopressin). Nearly 90% of the patients may present with bleeding due to minor trauma in the first year of their life.

Von Willibrand's Disease

This is another disease characterised by abnormalities in coagulation and occurs due to the deficiency of vWF (Von Willibrand factor), a protein required for promoting adhesion of platelets. The Patients may present with a variety of bleeding tendencies such as easy bruising, bleeding from gums or nose, menorrhagia, etc.

Duchenne Muscular Dystrophy

Both Duchenne muscular dystrophy and Becker muscular dystrophy are X-linked recessive disorders. The disorder is due to the mutation of dystrophin gene and can be diagnosed antenatally with the help of prenatal diagnostic tests.

Since this is an X-linked recessive disorder, the father suffering from the disease would have the defective gene on X chromosome, which he would pass to all his daughters. Therefore, all the daughters would be carriers. Affected father's Y chromosome would be normal, which he would pass to all his sons, who would all be normal. Carrier mothers would be having the defective gene on one of their X chromosomes. Thus, they would be transmitting the defective gene to 50% of their daughters and 50% of their sons.

The most commonly observed symptom is the weakness of the proximal muscles of the leg and pelvis along with the loss of muscle mass. The first clinical symptom of the disease may be delayed walking. There may be pseudohypertrophy of the calf muscles. The child may experience difficulty in walking or climbing up the stairs. The Becker's variety is much less severe in comparison to the Duchenne dystrophy and takes a slower course. The disease is characterised by progressive muscular wasting. The most common cause of death is respiratory failure occurring in early adult life.

McCune-Albright Syndrome

Introduction

McCune-Albright syndrome has been named after the two medical professionals, Donovan James McCune, and Fuller Albright who had independently described this illness in two separate reports published in 1936 and 1937, respectively. This is a rare disorder, with the prevalence ranging from 1 in 100,000 to 1 in 1,000,000.

This is not a hereditary condition. This syndrome occurs due to a spontaneous mutation of the GNAS gene (guanine nucleotide binding protein, alpha stimulating activity polypeptide 1) in early embryonic life. This gene is usually involved in the cell signalling mechanism. As a result of this mutation involving the alpha subunit of the stimulatory G protein, the enzyme adenyl cyclase is permanently activated. This causes activation of some intracellular signalling cascades in the absence of hormone stimulation.

A mosaic of cells, some having normal gene, while some others having the mutated gene are usually present in the affected individuals. This may lead to a wide variation in the phenotypic features. The severity of the disease condition depends on the number and location of cells containing the mutant gene. One side of the body can be more affected than the other. This may result in asymmetry of the head and face and different lengths of long bones on the two sides causing limping.

This syndrome is associated with a clinical triad of symptoms such as:

❑ *Precocious puberty*: Despite the low levels of gonado-tropins such as FSH and LH, the amount of oestrogen produced by the ovaries is quite high resulting in precocious puberty. Overactivity of other endocrine glands may also occur, resulting in endocrinopathies such as thyrotoxicosis (overactivity of thyroid gland), hyperparathyroidism (overactivity of parathyroids), Cushing's disease (overactivity of adrenal gland), pituitary gigantism (overactivity of growth hormone), hyperprolactinaemia (overactivity of prolactin), etc.
❑ *Café au lait spots*: These are usually present unilaterally and may have a ragged appearance, which is known as the "Coast of Maine" pattern.
❑ *Polyostotic fibrous dysplasia of bones*: Medullary tissue of the bones is replaced by fibrous tissues, resulting in an increased tendency for fractures and scoliosis.

There are no neurofibromas in this condition. Neurofi-bromatosis and café au lait spots are a characteristic feature

of Von Recklinghausen's disease or Neurofibromatosis type 1.

The disease may affect the kidneys resulting in "phosphate wasting". This can lead to rickets and osteomalacia. Bisphosphonates have a role in maintaining bone strength.

Glucose-6-Phosphate Dehydrogenase Deficiency

Introduction

This disease is associated with the deficiency of enzyme glucose-6-phosphate-dehydrogenase (G6PD). It is highly prevalent in individuals originating from most parts of Africa, Asia and from Southern Europe. It can rarely be found in other individuals. G6PD deficiency is the most common disease occurring due to the deficiency of an enzyme and shows X-linked inheritance. Due to the deficiency of this enzyme, G6PD, red cells become predisposed to haemolysis in response to certain chemical substances. Some of the chemicals that can trigger haemolysis include anti-malarial drugs such as primaquine, chloroquine and quinine; antibiotics like quinolones, e.g. nalidixic acid, and ciprofloxacin; mothballs containing naphthalene; consumption of Fava bean (also known as "Favism" because the bean consumption triggers haemolysis). The disease commonly affects the individuals residing in Africa, Mediterranean and Asia. There are more than 400 different mutations, which may be responsible for causing G6PD deficiency. This disease has become the most common cause of significant neonatal jaundice world-wide and usually occurs within the first 3 days of life. G6PD deficiency and Glucose-6-phosphatase deficiency are completely different conditions and the difference between the two is listed in **Table 8.7**.

Thalassaemia

Thalassaemia is a group of hereditary disorders characterised by the reduced synthesis of one or more of the globin polypeptides. There are two main types of globin chains, α and β. The gene for the synthesis of β globin is located on each of the two No 11 chromosomes. There are 2 pairs or 4 genes required for the synthesis of α chain; two each are located in each of the two chromosomes 16. The two chromosomes are inherited, one each from each of the parent. Due to this, the resulting adult haemoglobin (Hb A) is $\alpha_2 \beta_2$. Based on the fact whether the genetic defect lies in the transmission of α or β globin chain genes, thalassaemia can be classified as α or β thalassaemia respectively. Both α and β thalassaemia shows autosomal recessive inheritance. Each type of thalassaemia may occur as a homozygous or heterozygous defect. The homozygous state is termed as α or β thalassaemia major, whereas the heterozygous defect is termed as α or β thalassaemia minor or trait. In homozygous state, beta thalassaemia major causes transfusion dependent anaemia. In heterozygous state, beta thalassaemia trait

TABLE 8.7	Difference between glucose-6-phosphate dehydrogenase (G6PD) deficiency and glucose-6-phosphatase deficiency	
Parameter	G6PD deficiency	Glucose-6-phosphatase deficiency
Occurrence	Very common disorder	Rare disorder
Inheritance	X-linked recessive disorder	Autosomal recessive disorder
Pathogenesis	Haemolytic disorder in which haemolysis is triggered in response to certain chemicals	Glycogen storage disorder, also known as von Gierke's disease. Main defect is that glycogen cannot be converted to glucose
Clinical feature	Main presenting feature is severe haemolysis	Main presenting feature is profound hypoglycaemia

causes mild-moderate miocrocytic anaemia. Based on the type of gene deletions, α-thalassaemia can be classified into four types:
❑ Four α gene deletions: Hb Bart's hydrops fetalis
❑ Three α gene deletions: HbH disease
❑ Two α gene deletions: α thalassaemia trait
❑ One α gene deletion: α thalassaemia trait (carrier)

Pathophysiology

Beta thalassaemia genes are more prevalent in the following regions of the world: Mediterranean, African and south Eastern Asian population. The main defect in β thalassaemia is that the defect of β globin chains results in the accumulation of excessive α chains within the RBC's. Part of these excessive α chains is either removed by pairing with γ globin chain as hbF or after pairing with δ chains as hbA2. The remaining unpaired α chains precipitate within the cell in form of Heinz bodies. The precipitated α chains can cause damage to the red cell membrane. These red cells are damaged further and develop pitting upon the removal of precipitated aggregates. Thus these cells are irreversibly damaged and phagocytosed by the reticulo-endothelial cells of liver and spleen resulting in anaemia, hepotomegaly and excessive iron stores.

Clinical Features

Beta thalassaemia trait is usually asymptomatic and is identified during the routine blood evaluations. Thalassaemia major, on the other hand is detected during the first few months of life, when the patient's level of foetal haemoglobin decreases. Clinical features of β-thalassaemia are as follows:
❑ Appearance of anaemia in the first 4–6 months of life when switch from γ to β chain production occurs.
❑ There can be excessive RBC destruction, resulting in marked hepotoslenomegaly, extramedullary haematopoesis and iron overload.
❑ These can be marked erythroid hyperplasia resulting in bone expansion. This can cause typical thalassaemic facies and malocclusion of jaws due to expansion of maxillary bones, resulting in 'chipmunk' face.

❏ There may be characteristic changes on the X-ray of skull, resulting from the deformation of the skull bones. The skull may show hair-on-end appearance. This may be due to erythroid hyperplasia with intramedullary expansion and thinning of cortical bone. Bony changes may also be observed in other long bones, vertebrae and pelvis.

❏ Impaired immunity is associated with increased susceptibility to infection.

❏ Heart examination may reveal cardiac failure and arrhythmias due to severe anaemia and iron overload.

Lab Findings

❏ Blood film showing severe microcytic hypochromic red cell morphology, marked anisopoikilocytosis, basophilic stippling, nucleated red cells, presence of tear drop and target cells

❏ Low MCV and an elevated reticulocyte count is a feature of thalassaemia

Treatment

Patients with thalassaemia minor do not require any treatment. In cases of thalassaemia major, the aim is to maintain the haemoglobin level between 9–10 gm/dL through regular blood transfusions. Patients receiving blood transfusion may require iron chelation therapy with desferrioxamine.

Cystic Fibrosis

Introduction

Cystic fibrosis is an autosomal recessive condition, which is most prevalent in populations of Northern and Central European origin with an incidence as high as 1/625 live births. Incidence varies in other populations, being much less common among blacks and Asians.

This can be considered as the commonest hereditary disease in Caucasians, with the prevalence of carriers of cystic fibrosis in this population being approximately 1 in 25. Thus, the risk of both partners being carriers amongst Caucasians is about 1 in 625 (1 in 25 × 1 in 25).

Pregnant women affected with cystic fibrosis should undergo prenatal diagnostic tests to assess if their foetuses would be carrying the defective gene or not. In vitro fertilization and pre-implantation genetic diagnosis helps in the genetic analysis of embryo prior to its placement in the uterine cavity. Pre-implantation genetic diagnosis performed 3 days after fertilisation helps in the detection of abnormal cystic fibrosis genes. Embryos with two abnormal cystic fibrosis transmembrane conductance regulator (CFTR) genes are discarded and only those having at least one normal gene are implanted. Both tests such as amniocentesis and CVS will help in establishing the diagnosis of cystic fibrosis. A negative result on these tests cannot dismiss the diagnosis because they are only able to eliminate about 50 mutations or so. Thus, these tests are indicative of risk assessment.

Cystic fibrosis results from the mutation of CFTR gene. The CFTR gene encodes for a protein involved in water and chloride transport across cell membranes. This helps in regulating the components of various body fluids such as sweat, digestive juices and mucus. Mutations of this protein result in production of mucus, which has abnormal viscosity. Thick mucus is associated with reduced clearance, which causes tissue infection and damage. CFTR gene is found at q31.2 locus of chromosome 7. Though more than a 1000 mutations have been identified, the most common one is F508del (previously known as DF508 or ΔF508) – "del" or "D" or "Δ" stands for "deletion". F508del results in a 3 base-pair deletion, which causes the loss of a phenylalanine amino acid residue at position 508 on the protein. The gene in normal individuals is about 230,000 base pairs long and contains 1,480 amino acids. The normal gene product is a halide anion channel, which is important for creating sweat, digestive juices and mucus.

Universal neonatal screening has been established in the UK ever since the spring of 2007. Neonatal screening is done using heel prick for collecting blood sample. This is a biochemical test, which is based on measuring the levels of immunoreactive trypsinogen. Screening does not exclude carrier status.

Most individuals have two working copies of the CFTR gene, out of which only one copy is required to prevent cystic fibrosis. Cystic fibrosis develops in individuals who have no copy of functional CFTR gene. Development of cystic fibrosis requires that each parent passes on a mutated copy of the CFTR gene to their children. Since the disease has an autosomal recessive inheritance pattern, marriage between a woman with cystic fibrosis and normal man would result in all their children having carrier status.

Clinical Features

In the neonate the clinical features may include meconium ileus; respiratory infection; failure to thrive; malabsorption; steatorrhoea; rectal prolapse, etc. In children, the clinical features may include bronchiectasis; cor pulmonale; pancreatitis; diabetes; biliary cirrhosis; gall stones; portal hypertension; hypersplenism and heat exhaustion. In adults, the various clinical features include bronchiectasis; chronic pancreatitis; pulmonary hypertension; cor pulmonale; diabetes; malabsorption; gall stones; bile duct strictures and cirrhosis; duodenal ulcer; intestinal obstruction; azoospermia, etc.

The various clinical features of the disease can be described in details as follows:

Pulmonary Symptoms

Fibrosis and cyst formation in the lungs may result in the development of clinical symptoms such as:

❑ Difficulty in breathing and shortness of breath

❑ *Frequent infections of the lungs and/or coughing*: This can occur due to accumulation of thick, sticky mucus. The organisms commonly responsible for producing infection in cases of cystic fibrosis include *Staphyloccocus aureus, Haemophilus influenza*, and *Pseudomonas aeruginosa*. Lung colonisation with *Burkholderia cepacia* is associated with poor prognosis. Patients are more prone to develop allergic bronchopulmonary aspergillosis and infection with *mycobacterium avium* complex.

❑ *Infection of the sinuses*: There can be frequent infection of the sinuses, which may result in facial pain, fever, impaired nasal drainage and headache. Inflammation of the nasal tissues due to chronic sinus infection can result in the development of nasal polyps.

❑ *Other lung pathologies*: The various lung pathologies that can develop are pulmonary hypertension, heart failure, hypoxia, etc. The disease is often characterised by progressive lung disease and early death. Pulmonary hypertension and cor pulmonale may sometimes result. Development of pulmonary hypertension in a pregnant patient with cystic fibrosis can prove fatal. Infection of the lungs can commonly occur due to the clogging of the alveoli with mucus having abnormal viscosity.

Gastrointestinal Symptoms

❑ *Bowel obstruction*: Bowel obstruction can occur in childhood due to meconium ileus, which may occur in nearly 10% of affected neonates. This occurs when the meconium completely blocks the intestines due to the infant's failure of passing the meconium having abnormal consistency.

❑ *Rectal prolapse*: Rectal prolapse can occur due to increased faecal volume, malnutrition and increased intra-abdominal pressure.

❑ *Gastrointestinal abnormalities:* Malabsorption and diarrhoea can commonly occur. Malabsorption usually occurs due to difficult absorption of fat-soluble vitamins such as vitamin A, D, E and K. Lack of digestive enzymes may result in malabsorption. Resultant hypoproteinaemia may produce generalised oedema. The disease is also associated with a higher frequency of gastrointestinal pathologies such as heart-burn, intestinal blockage by intussusception and constipation.

❑ *Damage to the pancreas*: There can be development of cysts and fibrosis in the pancreatic tissue. Damage to the exocrine and endocrine part of pancreas can respectively result in pancreatitis and diabetes mellitus.

Reproductive Function

❑ *Fertility*: Most of males are infertile due to absence of the vas deferens. Azoospermia is commonly present in such patients. Though the problem is not yet remediable in most cases, there is the possibility of some becoming fathers. This is possible by use of ART techniques such as MESA (microsurgical epididymal sperm aspiration), followed by ICSI (intra-cytoplasmic sperm injection). Fertility in women is also reduced. Women may encounter fertility difficulties due to thickened cervical mucus, disruption of ovulation due to malnutrition, and/or amenorrhoea.

General Symptoms

❑ *Poor growth*: This typically manifests as the child's inability to gain weight or height at the same rate as their peers. Malabsorption leads to feeding problems and malnutrition is common. Growth failure can occur due to multifactorial causes such as chronic lung infections, poor absorption of nutrients through the GIT, malnutrition and increased metabolic demands due to chronic illness.

❑ *Clubbing*: Clubbing of fingers and toes can occur due to hypoxia as a result of chronic illness.

❑ The newborn baby may have salty tasting skin due to high chloride content of sweat.

❑ Presently, the life expectancy is about 51 years and is improving. It may be prolonged by heart and lung transplantation.

Treatment

Presently, there is no cure for the disease. Treatment basically involves the use of physiotherapy to clear the lungs of thickened secretions. Other drugs which can be used to provide supportive treatment include use of mucolytic drugs, antibiotics, digestive enzymes and supplements, etc. Such patients may also need dietary management along with the use of protein and vitamin supplementation (especially vitamins A, D and E).

Tay-Sachs Disease

Tay-Sachs disease is an autosomal recessive condition. It particularly affects Ashkenazi Jews, in whom the incidence is 1:3,600 pregnancies. It occurs in other populations, but the carrier frequency is about one tenth that of Ashkenazi Jews.

It is associated with progressive brain damage and symptoms such as blindness, convulsions etc., which may be fatal in early childhood.

Fragile X Syndrome

Introduction

Fragile X syndrome (also known as Martin-Bell syndrome or Escalante's syndrome) is an X-linked dominant condition, which mainly affects men. Despite of being a dominant condition, women are not usually affected because in women only one X chromosome remains active, which is usually the normal one (Lyon's Hypothesis). However, in some of the cells, X chromosome containing the mutation may also be expressed. As a result, approximately 30% of the female carriers of the mutations may show some of the disease symptoms such as intellectual impairment or mental retardation. Nevertheless, the women are less severely affected than men. Moreover, there is a risk of premature ovarian failure and early menopause, but only in women with the pre-mutation, not the full mutation. This risk of developing premature ovarian failure is about 16% in women with permutation in comparison with 1% risk in general population. An increased risk of premature ovarian function is observed if the pre-mutation was inherited from the father rather than the mother.

In men, this syndrome can result in mental retardation and autism. It can be considered as the most common inherited cause of mental handicap (ranking second after Down syndrome) as well as the most common known cause of autism. The degree of abnormality correlates with the size of the expansion.

Carriers of the pre-mutation are normal in early adulthood, but are at an increased risk of developing "Fragile X associated tremor/ataxia syndrome", (FXTAS), a neurodegenerative condition associated with dementia and ataxia, later in life. Women who are carriers are also at risk of FXTAS. However, their risk is lower than that of their male counterparts and usually do not have dementia. Men affected with FXTAS may develop symptoms like Parkinsonism or Alzheimer's disease such as dementia. It is now thought that some patients diagnosed with Parkinson's disease, Alzheimer's disease etc., who do not respond to the conventional treatment actually have FXTAS. Regular national screening program for this disease exists in Australia, but not in the UK. Screening in the UK is performed on an "ad hoc" basis by the local genetics laboratory. All potentially fertile girls in an affected family must be counselled.

The genetic abnormality is due to expansion (abnormal repetition) of a triplet of bases (CGG) in the FMR-1 gene (fragile X mental retardation gene 1) of X chromosome. This gene normally produces a protein, which is essential for the normal development of the brain. The CGG sequence or "triplet" is repeated many times within the gene. The gene has normally up to 50 repeats of the CGG triplet. The disorder can result from an incomplete mutation (pre-mutation) or complete mutation. The pre-mutation, which makes the female a carrier, has between 50 and 200 repeats

of the triplet. The full mutation, which gives rise to the full blown syndrome in the male, has greater than 200 repeats. The number of repeats is also known as the "expansion". In the female carriers, this expansion can enlarge when it is handed over to the offspring. The number of triplets can increase with subsequent generations, but only when transmitted from a mother. The increasing numbers of triplets in each generation only occurs in carriers, i.e. those with more than 50 repeats. Individuals who are not carriers, i.e. those having less than 50 repeats hand them on in a stable fashion from generation to generation. Furthermore, the increase in expansions only occurs when mothers hand it on to their sons. Fathers with a pre-mutation hand it on intact and always pass on the exact version to their daughters. A phenotypically normal woman may have a moderately large expansion, which further increases in her progeny, producing the full-blown illness in her sons. Thus an unaffected woman with pre-mutation can give birth to an affected son having full mutation.

Clinical Features

Males with the full mutation show the following clinical features:

❑ *Mental retardation*: Severe intellectual impairment, shyness, slowness in making social contact, tendency to avoid social contact, hyperactive and impulsive behaviour, speech or language delay, delay in crawling or walking and/or aggressiveness has also been described. Fragile X syndrome can be considered as the most common inherited cause of mental retardation and the second commonest genetic cause of mental retardation (first being the Down syndrome).

❑ *Autism*: it can be considered as the most common known cause of autism, with approximately 20% of men showing full-blown autism.

❑ *Facial features*: Characteristic facial features of the disease include:
  • Long face with coarse features
  • Large ears
  • Prominent lower jaw

❑ The body joints may be hyperextensible

❑ *Macro-orchidism*: Testicular enlargement following puberty may be observed in nearly 50% of cases.

Ambiguous External Genitalia

Ambiguous external genitalia is a defect where the genitals do not show an external appearance of either a male or a female.

Causes of ambiguous genitalia are as follows:
1. Masculinisation of female foetus:
  • Congenital adrenal hyperplasia
  • Maternal ingestion of androgenic or potentially androgenic drugs
  • Maternal masculinising tumour
2. Inadequate masculinisation of male foetus
3. True hermaphroditism.

The most common cause of ambiguous genitalia is congenital adrenal hyperplasia, of which 95% are deficient in 21-hydroxylase. Less common enzyme defects involve 11β hydroxylase, and 3-β hydroxysteroid dehydrogenase.

About two-thirds of those with 21-hydroxylase deficiency are salt losers, and present with a hypoadrenal crisis. Salt losers presenting in the newborn period with Addisonian crisis are more likely to be females. However, as they have abnormal genitalia they are easily spotted.

Transsexual Patients

Transsexual patients are those who have persistent discomfort with their assigned natal sex and its associated gender role. These patients are associated with strong and persistent cross-gender identification. There is an absence of any physical intersex condition. This condition is associated with clinically significant distress or impairment of social or occupational functioning. Such patients are usually treated with transsexual/sex change or gender reassignment surgery. This surgery involves the surgical conversion of a phenotypically normal male to a female or vice versa. This is usually followed by hormonal therapy and psychiatric assessment and counselling for these subjects.

Hermaphroditism

The chromosomal pattern is most commonly 46,XX, but some are mosaics or 46,XY.

Hermaphroditism is an extremely rare state in which there are both ovarian and testicular tissues: there may be a testis on one side and an ovary on the other; or ovotestes may be present; these are a histological mixture of ovary and testis. True hermaphrodites have an ovary on one side and a testis on the other and in such cases one or both of the gonads is almost invariably abnormal in structure and without function.

Pure Gonadal Dysgenesis

Gonadal dysgenesis is used to describe those situations in which primordial germ cells reach the ovary but are progressively destroyed so that few remain by the time of puberty. Chromosomal abnormalities usually underlie dysgenesis. Turner's syndrome and Turner's syndrome mosaic are common causes, but dysgenesis may also occur in females with normal karyotypes, that is, 46,XX. Patients with gonadal dysgenesis are hypo-oestrogenic with often elevated luteinising hormone/follicle-stimulating hormone ratio (LH/FSH), possess infantile secondary sexual characteristics, and are sterile. They may present with primary amenorrhoea.

Short Stature

Short stature is a well-known feature of Turner syndrome, achondroplasia and untreated hypothyroidism.

The presence of short stature in these children should alert clinicians to a co-existent endocrinopathy.

Obesity, thyrotoxicosis and nutrient excess are associated with tall stature.

Sweating

Sweats are seen in association with the following conditions:
❑ The menopause
❑ Phaeochromocytoma
❑ Acromegaly
❑ Thyrotoxicosis
❑ Hypoglycaemia (insulinoma).

Sweats are also a feature of:
❑ Tuberculosis (TB)
❑ Lymphomas and
❑ Brucella infection.

Choose the Single Best Answer (SBA)

Q 1. Which of the following statements regarding DNA is true?
A. Attached to the 2′ position of the sugar ring is one of four bases
B. Guanine-cytosine bonds consist of three hydrogen bonds
C. The two strands of DNA separate in vitro by heating it to 75°C
D. All codons have an identical function
E. There is a greater variety of amino acids than there are different codons

Q 2. Which of the following statement is true regarding human chromosomes?
A. Banding with quinicrine fluorescent stain can be used to identify X chromosome
B. Banding can be used to determine polymorphism in populations
C. Telocentric chromosomes have a centrally placed centromere
D. Terminal fragments called 'satellites' are present in the metacentric chromosomes
E. Telocentric chromosomes are present in human

1. B   2. B

Q 3. **The following are true regarding the Lyon's hypothesis**

  A. Lyon's hypothesis does not relate to germ cells

  B. The inactivation of X chromosome usually occurs during 16 weeks of life

  C. Barr body can be seen as a condensed mass during the prophase phase of mitosis

  D. Barr body may be observed in up to 80% of cells on a buccal smear from a woman

  E. Presence of an extra X chromosome is associated with intelligence above average

Q 4. **46 XX karyotype is associated with which of the condition?**

  A. Testicular feminisation syndrome

  B. Klinefelter's syndrome

  C. Muscular dystrophy

  D. Constitutional hirsutism

  E. None of the above

Q 5. **46 XY karyotype is associated with which of the following condition?**

  A. MRKH syndrome

  B. Testicular feminisation syndrome

  C. Klinefelter's syndrome

  D. All the above

  E. None of the above

Q 6. **Which of the following is not associated with a karyotype of 46 XY?**

  A. Hydatidiform mole

  B. Tay Sach's disease

  C. Muscular dystrophy

  D. All the above

  E. None of the above

Q 7. **Which of the following conditions is not associated with genetic anticipation?**

  A. Cystic fibrosis

  B. Fragile X syndrome

  C. Huntington's chorea

  D. Myotonic dystrophy

  E. None of the above

Q 8. **Increased numbers of chromosomes occur in which of the following condition?**

  A. Fragile X syndrome

  B. Down's syndrome

  C. Phenylketonuria

  D. Turners syndrome

  E. Cri du chat syndrome

Q 9. **Which of the following is not true regarding carriers of genetic disease?**

  A. Carrier state may be produced in an autosomal dominant

  B. Certain carrier states can be detected using DNA studies

  C. In case of cystic fibrosis if both the parents are carriers, the chances of having an affected child is 1 in 4

  D. In X-linked genetic diseases, the female carriers may show certain disease manifestations

  E. Marriage between the blood relatives is associated with an approximately double the risk of getting inherited disorders and birth defects

Q 10. **Which of the following is true regarding Down syndrome?**

  A. Shows X-linked pattern of inheritance

  B. There may be mild to moderate mental retardation

  C. Is characterised by the presence of muscle hypertonia

  D. Larger than normal space between the second and third toe

  E. The commonest type of congenital heart defect which may be present is coarctation of aorta

Q 11. **Which of the following is true regarding Edward's syndrome?**

  A. Is characterised by the presence of three copies of chromosome 13

  B. Has an autosomal recessive pattern of inheritance

  C. There may be absence of femur

  D. Increased space can be seen between the index and the middle finger

  E. There may be only 11 pair of ribs

Q 12. **Which of the following is true regarding Patau's Syndrome?**

  A. Is due to trisomy of chromosome 18

  B. May be characterised by the presence of colobomas in the iris

  C. There may be mild-to-moderate mental retardation

  D. The infant's head may be abnormally large

  E. Most individuals die within the first six months of life

Q 13. **Which of the following is true regarding Klinefelter's syndrome?**

  A. Is associated with hypogonadotropic hypogonadism

  B. It is inherited as an X-linked disorder

  C. Occurs due to non-dysjunction during mitosis

  D. Is associated with severe mental retardation

  E. It is associated with tall stature due to delayed fusion of the epiphysis

Q 14. **Which of the following is true regarding testicular feminisation syndrome?**

  A. Occurs due to the abnormality of the testosterone receptors

  B. Is associated with 47 XXY karyotype

  C. The individual is genotypically a female

  D. Has undetectable serum oestrogen concentrations

  E. Development of axillary/pubic hair is normal

Q 15. **Which of the following is true regarding müllerian agenesis?**

  A. The ovaries are absent

  B. The disease may be due to a genetic defect

  C. Abnormalities of gastrointestinal tract are frequently present

  D. Pubic/axillary hair are scanty or absent

  E. Use of Franks dilators is an invasive form of therapy

3. A   4. D   5. B   6. A   7. A   8. B   9. A   10. B   11. E   12. B   13. E   14. A   15. B

Q 16. Which of the following is true regarding 5-α reductase deficiency

A. The disease is characterized by an abnormality of testosterone receptors
B. Dehydrotestosterone is not produced
C. The external genitalia develop normally
D. Has an X-linked inheritance
E. The karytotype is XXY
F. Breast development is like that of normal females

Q 17. Which of the following is true regarding Turner's syndrome?

A. The individual is phenotypically a female
B. Is associated with congenital absence of the uterus
C. Is associated with mental retardation
D. Has an autosomal recessive pattern of inheritance
E. Is associated with oestrogen insensitivity

Q 18. Which of the following disorders have an autosomal recessive pattern of inheritance?

A. Christmas disease
B. Neurofibromatosis
C. Colour blindness
D. Tay Sachs disease
E. Achondroplasia

Q 19. Which of the following disorders have an autosomal dominant pattern of inheritance?

A. Thalassaemia major
B. Galactosaemia
C. Polyposis coli
D. Vitamin D resistant rickets
E. Hemochromatosis

Q 20. Which of the following disorders have an X-linked mode of inheritance?

A. Adult polycystic kidney disease
B. Hurler's syndrome
C. Fragile X syndrome
D. Alzheimer's disease
E. Achondroplasia

Q 21. Which of the following statement regarding haemophilia A is correct?

A. Haemophilia A is commoner in females
B. If father is suffering from the disease and mother is the carrier of the abnormal gene, there is a 1 in 4 chance that the offspring will be affected
C. In mild cases bleeding must be controlled by the infusion of factor VIII
D. The increased bleeding tendency usually manifests after 1 year of age
E. Levels of factor VIII related antigen is normal in these cases

Q 22. Which of the following statement is true regarding Duchenne muscular dystrophy?

A. It shows an autosomal recessive pattern of inheritance
B. Is characterised by the weakness of distal muscles of leg

C. The affected father passes the defective gene to all his sons
D. Carrier mothers will always produce affected sons
E. Carrier mothers will have 50% probability of producing carrier daughters

Q 23. Which of the following statement is correct regarding McCune Albright syndrome?

A. Has an autosomal dominant pattern of inheritance
B. Can result in ovarian failure
C. There may be presence of neurofibromatas
D. Is associated with polyostotic fibrous dysplasia
E. Levels of gonadotropins are markedly reduced

Q 24. Which of the following statement regarding glucose-6-phosphate dehydrogenase deficiency is correct?

A. Shows an autosomal recessive pattern of inheritance
B. Can be triggered by ciprofloxacin
C. Can cause profound hypoglycaemia
D. Is common amongst Ashkenazi Jews
E. None of the above

Q 25. Which of the following statement regarding thalassemias is correct?

A. Shows X-linked recessive inheritance
B. There is marked reticulocytosis
C. There may be undergrowth of maxillary regions of face
D. There may be persistence of HbF
E. None of the above

Q 26. Which of the following is true regarding cystic fibrosis?

A. It is more common amongst Caucasians
B. This disease is associated with X-linked recessive inheritance
C. The condition is due to a unique mutation
D. Pulmonary hypertension is a good prognostic feature for pregnant woman with cystic fibrosis
E. Presently neonatal screening for cystic fibrosis is not been done routinely in the UK

Q 27. Which of the following is true regarding the fragile X syndrome?

A. Regular national screening program for fragile X syndrome exists in the UK
B. There may be small, low set ears
C. May be associated with labial/ clitoral enlargement
D. Women are more severely affected than the males
E. Women with permutation are at an increased risk of premature ovarian failure

Q 28. A 27-year-old female developed insulin dependent diabetes mellitus. Her uncle and grandmother also had diabetes mellitus. What is the most likely mode of inheritance for her condition?

A. Autosomal co-dominant
B. Autosomal dominant
C. Autosomal recessive
D. Polygenic
E. Single gene defect

16. B   17. A   18. D   19. C   20. C   21. E   22. E   23. D   24. B   25. D   26. A   27. E   28. D

Q 29. A mother is concerned regarding her baby who has developed fractures which appear to occur with minimal trauma. He has blue sclera. What is the most likely mode of inheritance for the baby's condition?
- A. Autosomal co-dominant
- B. Autosomal dominant
- C. X linked recessive
- D. Polygenic
- E. Single gene defect

Q 30. Which of the following statements regarding multi-factorial inheritance is true?
- A. Blood groups are inherited in this manner
- B. Can be diagnosed by chromosome culture
- C. It is due to the effects of a large number of genes and the environment
- D. The recurrence risk in this type does not depend on the previous incidence of the same condition in the family.
- E. None of the above

Q 31. Which of the following is not true regarding ambiguous external genitalia at birth?
- A. Is associated with drug ingestion during pregnancy
- B. Is commonly due to congenital adrenal hyperplasia
- C. Occurs in true hermaphroditism
- D. Occurs in complete testicular feminisation syndrome
- E. There are normal male genitalia in 47,XXY-Klinefelter's syndrome

Q 32. Which of the following is characteristically true concerning transexual patients?
- A. Are of subnormal intelligence
- B. Are socially well-adjusted
- C. Have abnormal hormone profiles
- D. Have abnormal karyotype
- E. Have normal external genitalia

Q 33. Which of the following is true regarding hermaphroditism?
- A. Chromosomal sex is usually female (46,XX)
- B. End-organ resistance is a feature
- C. External sex is usually female
- D. Mosaics do not occur
- E. Either the primordial follicles or the seminiferous tubules are present

Q 34. Which of the following statement concerning patients with pure gonadal dysgenesis is not correct?
- A. Always have primary amenorrhoea
- B. Are frequently XO or XO/mosaic
- C. Have a uterus
- D. Have LH and FSH concentrations within the normal range
- E. Have poorly developed breasts

Q 35. Which of the following statement regarding genes is correct?
- A. Introns are the portions of a gene which code for protein
- B. Mitochondrial genes are inherited from the mother
- C. Most of the human genome encodes polypeptide
- D. The rate of DNA replication is directly under the control of enhancer sequences
- E. Transcription factors are mainly made of DNA

Q 36. Which of the following is true concerning adenine?
- A. Can be converted directly to a nucleotide by the action of phosphoribosyl-transferase enzymes
- B. Forms base pairs with thymine in RNA
- C. Is a pyrimidine base
- D. Is degraded by a pathway which involves the enzyme xanthine oxidase
- E. Is synthesised attached to ribose phosphate

Q 37. Which of the following technique is not directly used for used for identifying DNA?
- A. Denaturing gradient gel electrophoresis
- B. Northern blotting
- C. Polymerase chain reaction
- D. Southern blotting
- E. None of the above

Q 38. Which of the following is true concerning mitochondrial DNA?
- A. Is inherited from both parents
- B. Is not present in spermatozoa
- C. Have their own genome
- D. Are scantily expressed in the neuronal tissue
- E. All children of an affected mother can transmit the trait

Q. 39. Which of the following is/are true regarding the human chromosomes?
- A. Karyotype analysis is carried out after arresting the diving cells in anaphase stage of mitosis
- B. Only the X chromosome of maternal origin is active
- C. The number of Barr bodies visible at interphase in human somatic cells is always one less than the total number of X chromosomes
- D. Single Barr body is found in males with Down syndrome
- E. No Barr body is present in individuals with Klinefelter's syndrome

Q 40. Genetic counselling entails which of the following?
- A. Requires chromosome culture
- B. Always requires discussion of pre-natal screening
- C. Involves chorionic villus biopsy
- D. Involves a detailed family history
- E. Usually involves marriage guidance counselling

Q 41. A 38-years old primiparous woman with 12 weeks gestation undergoes a screening test for Down syndrome, which reveals an increased foetal risk for development of trisomy 21. Which of the following confirmatory test must be offered to her?
- A. Cell-free fetal DNA sampling
- B. Amniocentesis
- C. Chorionic villus sampling
- D. Nuchal translucency measurement
- E. Cordocentesis

29. B 30. C 31. D 32. E 33. A 34. D 35. B 36. D 37. D 38. C 39. C 40. D 41. C.

Biophysics

Radioactivity

Ionising Radiation

An atom is the smallest constituent of ordinary matter. Each atom is composed of a nucleus containing the positively charged protons and neutral neutrons of roughly equal mass orbited by one or smaller negatively charged electrons, circulating in up to 10,000 times the diameter of nucleus. Hydrogen (atomic weight 1) is composed of a proton in its nucleus and no neutron.

Ionising radiations is radiation carrying enough energy which is able to liberate electrons from atoms or molecules, thereby ionising them. Ionising radiation can be categorised based on the nature of the particles or the electromagnetic waves which create the ionising effect. Different types of ionising radiations **(Fig. 9.1)** are as follows:

Alpha particle: Alpha particle is identical to helium particles and has two protons and two neutrons. It has twice the charge and four times the mass of a proton. They are sometimes also denoted as He^{2+} (helium ion with +2 charge and two electrons missing). Alpha particles are highly ionising form of particle radiation. However, they have low penetration power when they are produced by alpha decay; they can be stopped by a few centimetres of air or by the skin.

Beta particles: Beta particle is a negatively charged particle known as electron, having high speed and high energy, which are emitted by certain radioactive nuclei.

Positron: A positron is an elementary particle of roughly equal size to an electron but is positively charged. Positrons are artificial sources of ionising radiations commonly used for PET scanning (positron emission tomography).

Gamma rays: Gamma rays are ionising radiations which are electromagnetic radiations having very high frequency because they consist of high-energy photons. These rays are produced during the process of γ-decay from the naturally occurring radioisotopes and are generated from the nucleus.

X-rays: X-rays are electromagnetic radiations, which are generated by the circulating electrons (either in orbitals outside of the nucleus or while being decelerated to produce the Bremsstrahlung effect (X-rays emitted by an electron when deflected by another charged particle, typically an atomic nucleus).

The energy is also transferred to the electrons in the material through which X-rays and γ rays pass. The amount of energy transferred is dependent on the frequency of the rays. This causes electrons to escape from their parent atom and cause ionisation. The X-rays and γ rays continue traveling, but its frequency is reduced. In case of X-rays, the velocity remains unchanged. Therefore, their tendency of interacting with one another largely remains unchanged.
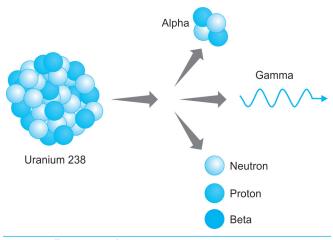
FIG. 9.1: Different types of radioactive particles

As a result, it does not have a well-defined range, rather dies away exponentially with distance.

Half value layer: Thickness of a material, which will reduce their ionization to half its initial value as a measure of the penetrating power of the X-rays or gamma rays.

Ionisation and Excitation
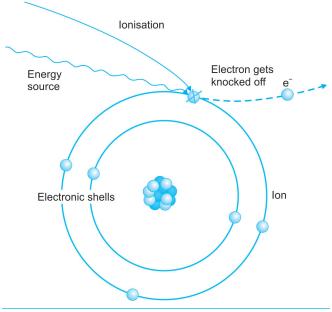
Ionisation can be considered as the most dominant marker of the activity of radiation. Radioactivity is usually measured in terms of ionization. The force exerted by ionisation causes the electron to be dragged out of the orbit. This force is usually longer and lasts for a longer duration of time. This results in the production of two new ions, positive and negative **(Fig. 9.2)**.

Excitations, on the other hand, are imperfect ionisations in which the energy is imparted to the outer electrons of the atom to put them into orbits of energy higher than their usual **(Fig. 9.3)**. However, energy is not sufficient for them to escape from the parent nucleus. Both the processes, excitation and ionisation, are likely to produce new chemical species within the cell, some of which may initiate further chains of damaging chemical reactions within the cell. Both these phenomena are responsible for producing biological effects of radiations.

The most important mode of cell destruction by ionising radiations is DNA disruption. Other processes, which may be responsible for cell destruction and injuries, may include damage to the cell membrane, osmotic changes, release of lysosomal contents, damage to mitochondria, etc.

Units of Radioactivity

The SI unit for the measurement of radioactivity is Becquerel (Bq) and is a measure of the amount of atoms which will disintegrate per second. One Bq is defined as one transformation (or decay or disintegration) per second.

1 Bq = 1 transformation per second

Previously, radioactivity was measured in terms of a unit known as Curie (Ci) and was originally defined as "the mass or quantity of radium emanation in equilibrium with the 1 gram of radium. According to the present definitions, 1 curie is defined as 3.7×10^{10} disintegrations per second so that

1 Curie = 3.7×10^{10} transformations per second

Another measure of radioactivity is half-life which defines the rate at which this disintegration occurs. This can be described as the time required for half the initial quantity of radioactive material to complete its transformation.

Children Exposed in Utero to X-Ray Irradiation

The risks to the foetuses are small following exposure to chest X-rays, but far greater with direct exposure following abdominal exposure.

Generally, there is an increased risk of childhood leukaemias and cancers (e.g. acute lymphoblastic leukaemia, cerebral gliomas, etc. There appears to be no evidence for IUGR, diabetes or mental retardation—although IQ on a population basis may be significantly lower).

Radiotherapy

Radiotherapy is a form of ionising radiation, which is commonly employed for the treatment of various malignant (e.g. carcinoma cervix and vulva) and non-malignant conditions. Radiotherapy is targeted at localized areas to avoid side effects related to generalised exposure. Radioactivity can result in several side effects which are summarized in **Table 9.1**.

Various sources of radiation in clinical practice are as follows:

Ionising Radiation

- ❑ X-ray
- ❑ Radiotherapy
- ❑ Positron emission tomography
- ❑ Barium studies (e.g. barium swallow, meal, enema, etc.)
- ❑ CT scan

FIG. 9.2: Ionisation
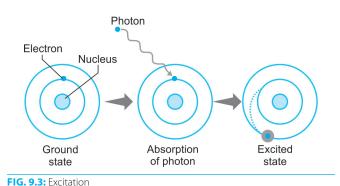
FIG. 9.3: Excitation

TABLE 9.1	Side-effects of radiotherapy
Acute	Late
Epithelial surface damage (e.g. mouth ulcerations)	Lymphoedema
Gastrointestinal damage, resulting in symptoms such as nausea, vomiting, abdominal pain, etc.	Hair loss
Cystitis	Further development of cancers
Fatigue	Tissue fibrosis
Oedema	Infertility

Non-ionising Radiation

❑ Electrocautery
❑ Laser
❑ MRI
❑ Ultrasound
❑ Microwaves
❑ Diathermy

The equivalence of radioactive exposure by various imaging modalities in terms of duration of exposure by the natural background radiation is compiled in **Table 9.2**.

Principles of Ultrasound

Introduction

Ultrasound waves are sound waves, having very high frequency (varying between 16–16,000 cycles per second), generated by the effect of electricity on a ceramic crystal. This is known as the piezoelectric effect **(Fig. 9.4)**. Medical ultrasound uses sound waves with frequency varying between 1 million and 10 million cycles per second. The ultrasound frequency most commonly used in obstetrics is 3.5 MHz. Ultrasound beam is produced as a series of pulses. Ultrasound waves are non-ionising and pulsatile in nature. Ultrasound is a longitudinal wave form, which can be focused, reflected and refracted.

Velocity = frequency × wavelength

The wavelength of the waves determines the axial resolution of the system, while the width of the waves determines their lateral resolution. The frequency of a sound wave is inversely related to its wavelength. Procedures for processing the ultrasound signals are described in **Table 9.3**.

Doppler ultrasound is used to determine the velocity of blood flow. Various types of Doppler ultrasounds which can be used include pulsed waves, continuous and colour.

As a sound beam passes through tissue, some of the energy of this sound wave is absorbed by the tissue. However for low intensities of ultrasound, the heat deposited is quickly dissipated and does not build up.

TABLE 9.2	Equivalent duration of natural background radiation of various imaging modalities
Imaging modality	Equivalent duration of natural background radiation
Chest X-ray	2–10 days
X-ray lumbar spine	6 months
Intravenous urogram	1 year
CT abdomen	3 years

TABLE 9.3	Different types of ultrasound signals
Type of ultrasound signal	Definition
A-mode (amplitude)	Echo is displayed in form of a deflection (spike) on oscilloscope as the ultrasound wave comes in contact with various tissues. Size of the wave spike is related to the strength of the echo. It is rarely used nowadays
B-mode (brightness)	Movement of the transducer yield a series of dots which build a two-dimensional image (brightness modulation)
Real-time	Transducer is moved automatically either mechanically or via electronic means to generate successive B-scans
M-mode (time-position mode)	Continuous displacement of B scan with respect to the time. This therefore demonstrates movement
Gray-scale ultrasound	Selective amplification of low-level echoes from the soft tissue
Doppler mode	Used for assessing movement and therefore can be used for looking at the blood flow
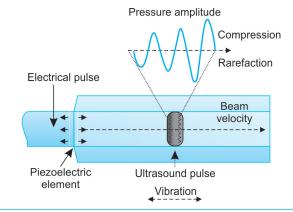
FIG. 9.4: Principle of ultrasound

Biological Effects of Ultrasound

There are two principal bio-effects of ultrasound:
❑ *Thermal*: Thermal effect is created through the impact of acoustic energy upon tissue.

❏ *Mechanical*: Mechanical bio-effects include cavitation and streaming associated with the violent agitation of particles within the medium.

- *Cavitation*: Cavitation is a recognized effect and is due to the generation, growth, vibration and possible collapse of microbubbles in the tissue. These microbubbles are generated by the ultrasound waves. These bubbles may "move" with the sound beam or some may oscillate so strongly that the bubbles collapse suddenly producing local effects.

Milestones of Pregnancy on Ultrasound

Using ultrasound, pregnancies can only be detected following implantation. Anomaly scanning is usually carried out in the second trimester of pregnancy. Most commonly this examination is now conducted around 20 weeks of gestational age. Foetal maturity cannot be reliably determined using ultrasound in the third trimester.

Foetal heart activity is usually visualized at 6–7 weeks of gestation using transabdominal sonography and a week earlier using transvaginal ultrasound. When the foetal heart activity is visualized, the crown-rump length is usually greater than 2 mm. However, in nearly 5–10% of embryos; no foetal heart activity may be visualized until the CRL becomes greater than 4mm. Absence of foetal heart activity in presence of CRL of ≥7 mm implies non-viable pregnancy. In these cases, a further scan is indicated after an interval of 1 week. CRL is about 1 mm at 5 weeks and 4 mm at 6 weeks of normal gestation.

Properties of Ultrasound

Resolution

Resolution of ultrasound increases with increased frequency. The higher the frequency, the less the depth of penetration; vaginal transducers can use higher frequencies than abdominal transducers, thereby providing better resolution. Medical ultrasound uses the range of 100–500 kHz.

Attenuation

Attenuation refers to the weakening of sound waves as it passes through the tissues because parts of it are reflected, scattered, absorbed, refracted or diffracted. Attenuation increases with increased frequency.

Sound has energy and on attenuation of that energy, heat is created.

Intensity

This describes the amount of energy passing through a certain cross-sectional area usually 1 cm^2. Intensity is measured in terms of watts per centimetre square (W/cm^2).

Refraction

Refraction is the bending of a wave beam when it crosses at an oblique angle the interface of two materials, through which the waves propagate at different velocities. In other words, refraction is the change in direction of the wave as a result of change in velocity.

Diffraction

Diffraction describes the bending of waves during its passage as a result of interaction with obstacles.

Impedance

Acoustic impedance can be described as the opposition to the passage of sound waves and is a function of density and elasticity. Impedance is the product of the density of a material and the speed of sound in that material. Impedance of a material describes how it resists being moved in response to a given sound wave. In case of soft tissues, impedance is proportional to the tissue density. When ultrasound encounters a boundary between tissues having different impedance, this mismatch of movements prevent a proportion of the sound energy from being transferred. The rest is reflected and produces echoes used in diagnostic ultrasound. Therefore, acoustic impedance determines beam reflection. Impedance is measured in $g/cm^2/sec$.

Velocity of Ultrasound

The velocity (V) can be defined as the speed in a given direction.

V = wavelength × frequency

It is directly proportional to frequency and upon the impedance of the medium through which the sound wave travels

Velocity = impedance/density of material through which the wave travels.

It is inversely proportional to the compressibility of the medium (a function of density). The greater the compressibility, slower is the velocity. Compressibility is affected by temperature, and hence this too will affect velocity of ultrasound. Ultrasound velocity in any given medium is constant.

Adverse Effects of Ultrasound

Though ultrasound is a relatively safe imaging modality, its usage requires monitoring to ensure that there are no adverse effects related to its use. The main concern related with the use of ultrasound is the occurrence of thermal effects due to the heating of the local tissues. Exposure to ultrasound therefore must be kept to minimum in order to reduce the potential impact of exposing the embryo or

the foetus to the thermal effects of ultrasound. Caution should be particularly applied when using the transvaginal ultrasound during early pregnancy. Safety protocols recommend the use of thermal indices, which serve as a measure of tissue heating effects; thermal index must be preferably less than 1 and the scanning time must be kept as short as possible.

Doppler Ultrasound

Doppler ultrasound has emerged as an important and widely accepted method of foetal monitoring.

Principles of Doppler

The ultrasound waves are targeted at a particular object (reflector), which they strike before getting reflected. If the reflector does not move, the frequency of the reflected wave is equal to that of transmitted wave **(Fig. 9.5)**. If the reflector moves towards the transceiver, the reflected frequency would be higher than that of the transmitted wave, while if the reflector moves away from the probe, the frequency of the reflected wave would be less than that of the transmitted wave. This frequency change is known as the Doppler shift and is proportional to the velocity of the reflector. The Doppler ultrasound probe is able to transmit ultrasound waves into the body, directed towards a particular blood vessel and receive their reflections from the body. The apparatus then measures the difference between the transmitted and received frequencies. The frequency difference (expressed in hertz) is proportional to the velocity of movement along the line that connects the wave transceiver and the moving reflector. In obstetric applications, the Doppler wave is produced by insonating the moving blood vessels with ultrasound waves. This principle has been named after its discoverer, an Austrian mathematician and physicist, Christian Andreas Doppler.
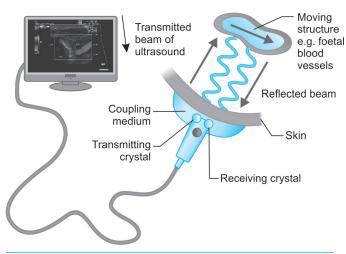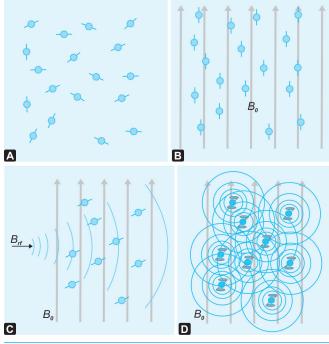
FIG. 9.5: Principle of Doppler ultrasound

Principles of MRI

Application of directional magnetic field, which causes change in the motion of the field of charged particles, forms the basis of MRI. When the human body is exposed to a strong magnetic field, the nuclei of the hydrogen atoms, which were previously randomly aligned, align themselves in the direction of magnetic field—"stand to attention" position **(Figs 9.6 A and B)**. A magnetic moment is thereby created. Next a radiofrequency pulse is applied perpendicular to the magnetic field. The nuclei of the hydrogen atoms are able to act as microscopic compass needles, due to which when they are submitted to pulses of radio waves, the energy content of the nuclei changes. Application of the radiofrequency pulse, therefore, causes the net magnetic moment of the nuclei to tilt away from the magnetic field **(Fig. 9.6C)**. On stopping the radiofrequency pulse, the nuclei return to equilibrium such that the net magnetic moment becomes parallel to the magnetic field **(Fig. 9.6 D)**. After the pulse, a resonance wave is emitted when the nuclei return to their previous state. The small differences in the oscillations of the nuclei are detected. By advanced computer processing, it is possible to build up a three-dimensional image that reflects the chemical structure of the tissue, including differences in the water content and in movements of the water molecules.
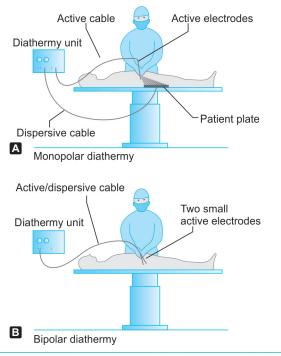
FIGS 9.6A TO D: Principles of magnetic resonance imaging; (A) Random alignment of hydrogen nuclei in the absence of a strong magnetic field; (B) On application of a strong magnetic field, the hydrogen nuclei align themselves in the direction of magnetic field; (C) Application of a radio-frequency pulse perpendicular to the direction of magnetic field, which causes net magnetic moment of the nuclei to tilt away from the magnetic field; (D) On stopping the radiofrequency pulse, the nuclei return to equilibrium

This results in a very detailed image of tissues and organs in the investigated area of the body. In this manner, pathological changes can be documented.

Surgical Diathermy

Diathermy/electrosurgery implies the use of electricity to generate heat in the tissues. It is used at the time of surgery to vaporise the tissues for cutting purposes or for coagulating the tissues to achieve haemostasis or for destroying the tissues. Diathermy (or Bovie) is a device used to pass an electric current through tissues which causes coagulation, cutting and tissue destruction by a heating effect. It uses alternating current with a frequency of 500 KHz–1 MHz. Cutting diathermy has an alternating sine-wave pattern, whereas coagulation diathermy uses current with a damped or pulsed sine wave pattern. The electricity has to be of high frequency, usually referred to as radio-frequency. One of the main advantages of the high frequency is that it is too fast to stimulate nerve fibres. This prevents spasm or paralysis of muscles. To avoid these unwanted effects, one needs to use frequencies greater than 100 kHz. Diathermy units use frequencies from 500 kHz (500,000 cycles per second) to 2 MHz (2 million cycles per second). Two types of diathermy are commonly used in gynaecological surgery: unipolar and bipolar **(Figs 9.7A and B)**. The current density is higher in bipolar (e.g. bipolar diathermy forceps) in comparison to unipolar or monopolar diathermy. Monopolar diathermy can be used to achieve cutting, whereas cutting is not possible with bipolar diathermy. Both the techniques can be, however, used to desiccate the tissues.

FIGS 9.7A AND B: Diathermy: (A) Monopolar diathermy; (B) Bipolar diathermy

Bipolar diathermy: Bipolar diathermy is a very safe form of diathermy. It is usually employed at the time of using forceps. The current only flows between the tips of the forceps, from the active electrode to the neutral electrode. So there is less risk of stray currents damaging tissues other that those, which are being aimed at. Diathermy employs use of continuous current at low voltage: 500–1,000 volts. The water in the cells is turned to steam, so the cells vaporise giving a cutting effect. The best effect will occur when the current flows through the smallest tissue volume. This can be produced by using a clean needle point. The low voltage (500–1000 volts) current flows continuously. Since the effect is relatively superficial and the tissues are vaporised, not coagulated, there is no great haemostasis. This is used in laparoscopic surgery for dividing adhesions, myomectomy etc. The total tissue damage is less, so smoke production is less.

Unipolar diathermy: This is the type of diathermy that is most often used in surgery, including open, minimally invasive, colposcopic and hysteroscopic. In these cases, electrons are driven via a circuit, from the surgeon's hand-held "active electrode" (connected to the diathermy machine) through the patient and leave the patient via the return electrode attached to the patient. Once the device is applied to the patient, the current will flow from the point of contact and spreads out as it passes through the patient, heading for the "return" pad, which is usually attached to the patient's thigh. From there it runs back to the diathermy machine, so completing the circuit. Tissues will be heated according to the amount of electric current running through them. The greatest current per cubic centimetre of tissue will be at the point of contact of the active electrode. The tissue in this area will usually be coagulated or vaporised. Away from the immediate point of contact of the active electrode, the current spreads out. So the amount of current passing through any cubic centimetre will be small and the temperature rise will be insufficient to cause tissue damage.

Precautions

Precautions to be taken while using diathermy include the following:
- The colon contains hydrogen and methane therefore use of diathermy on the colon is an explosive risk.
- Only the surgeon wielding the active electrode should activate the machine. The dial setting should be checked by the surgeon operating before the operation starts.
- Placing the active electrode in an insulating quiver prevents inadvertent burns.
- If diathermy performance is poor the plate and lead should be checked and replaced if necessary.

Complications of Diathermy

- *Risks of the surgical procedures*: Laparoscopy, hysteroscopy and open surgery have their own risks, to which

are added the risks of diathermy. The same problems with the use of diathermy can occur during laparoscopic and hysteroscopic surgery as with open surgery. The risks are greater with the latter. The medium used to distend the abdomen at laparoscopy must be non-combustible. At hysteroscopy, the medium used should be non-conductive if diathermy is to be used, so the electric current is not dissipated. Saline is conductive. Therefore, it is best avoided.

❑ *Operator error*: The main problem associated with diathermy is operator error. This could either involve treating the wrong tissue or allowing super-heated tissue after treatment to come into contact with other tissue, such as bowel, which may inflict thermal damage to the surrounding tissues. Precautions such as proper training/servicing and checking equipment can help prevent these errors. The following precautions may be required:
  • All staff requires proper training
  • The equipment used must be modern and up-to-date
  • The equipment must be regularly serviced and thoroughly checked before each use.

❑ *Thermal injury*: With bipolar diathermy, the risks of thermal injury are minimal. The current only flows between the tips of the instrument, so heat is only generated only at the tips. There is no risk of stray currents. There is a small risk of damage from heated tissue coming into contact with other tissues. Unipolar diathermy, on the other hand, is associated with considerable risk of inadvertent damage. Direct coupling is one of the major complications associated with unipolar diathermy and refers to the tissue damage caused by the electrode touching another nearby conducting instrument. In order to minimize the occurrence of side effects, strict adherence to the safety protocols must be followed. The whole of the probe is not in the field of view, so faulty insulation out of the field of view might cause unseen damage. Most damage is not seen or appreciated at the time. There is a need for careful assessment if recovery from surgery does not occur as per the norm. The thigh pad has to be applied over a big enough area to prevent burns.

Lasers

LASER is an acronym for Light Amplification of Simulated Emission of Radiation. A laser produces a highly directional beam of coherent (monochromatic) electromagnetic radiation. Photons of energy produced from energised atoms in the lasing medium are reflected back and forth many times between the mirrors, thereby amplifying their number. There is a production of perfectly parallel beams of light enabling extremely tight focusing **(Fig. 9.8)**. Due to concentration of energy in a small area, brief pulses
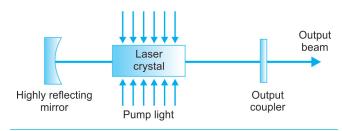
FIG. 9.8: Principle of laser

produce high local temperatures. These high temperatures are sufficient to vaporize tissues and also cause cauterising action, which seals the edges and reduces blood loss. Most commonly used laser in medical practice is Nd: YAG lasers (neodymium-doped yttrium aluminium garnet). This laser is commonly used for ablation of foetal vessels in cases of twin-twin transfusion syndrome. These are rod lasers pumped by a flash lamp having a broad spectrum of light. The flash causes excitation of electrons orbiting around the nucleus to move into orbits with higher energy. As these electrons drop back into their original position, they may emit a photon of light. The rod has polished squared off ends and is placed between mirrors with surfaces that are exactly parallel so that any light emitted within the rod is reflected back and forth.

Carbon dioxide lasers are also used for performing cone excisions from the cervix and for laser ablation of cervix. However, CO_2 lasers are unsuitable for operating through thicker tissues (e.g. ablation of endometriosis lesions). In these cases, Nd:YAG lasers are used because its wavelength is in the near infrared range.

The lasing medium determines the wavelength of electromagnetic radiation emitted. The lasing medium is most commonly gaseous but may be crystalline.

Classes of Lasers

Class 1: Class 1 is considered low risk. These are low power devices emitting radiation below the maximum permissible exposure (MPE).

Class 2: Class 2 is considered low risk. These are low power devices emitting visible radiation. Safety is normally afforded by natural aversion responses.

Class 3: Class 3 can be further classified as class 3a and 3b

Class 3a is considered low risk but may be a hazard if the beam is optically focused.

Class 3b is considered a medium risk. Direct viewing may be a risk.

Class 4: Class 4 is considered high risk. These are high powered devices with a potential fire hazard. Most medical lasers are in this class.

Safety Precautions at the Time of Using Lasers

All persons using a laser should be suitably trained and be aware of all safety precautions.

There should be control of personnel allowed to enter the area and the entrance should be marked with appropriate illuminated warning signs. Reflective surfaces should be avoided. However, matt black surfaces are not necessary. Eye protection must be appropriate to the type of laser being used. The ventilation should include an extraction system to vent the fumes produced. An LSO (laser safety officer) is appointed from the staff of each department using the laser and has custody of the laser key.

Choose the Single Best Answer (SBA)

Q 1. Children exposed in utero to X-ray irradiation are at an increased risk of which of the following?

A. Diabetes
B. Acute lymphoblastic leukaemia
C. Intra-uterine growth retardation (IUGR)
D. Mental retardation
E. None of the above

Q 2. Which of the following is true regarding diagnostic ultrasound?

A. Medical ultrasound uses the range of 1,000–5,000 kilohertz (kHz).
B. Is associated with a 1°C rise in body temperature after 15 minutes of scanning
C. It is ionising
D. It is pulsatile
E. High frequency ultrasound has greatest tissue penetration

Q 3. Which of the following is not true regarding ultrasound examination during pregnancy?

A. Can diagnose foetal ascites
B. Anomaly scanning is usually carried out in the second trimester of pregnancy
C. Cannot reliably establish foetal maturity at 34 weeks' gestation
D. Can diagnose a cleft lip
E. Is able to identify the fertilised ovum prior to implantation.

Q 4. In experimental conditions, which of the following biological effects can be produced on the tissues by the ultrasound waves?

A. Cavitation
B. Heat generation
C. Microstreaming
D. All the above
E. None of the above

Q 5. Which of the following statement regarding the properties of the ultrasound waves is not correct?

A. Impedance determines the proportion of sound energy reflected and transmitted at an interface
B. The size of a pulse generated in an A-scan is a measure of the intensity of the reflected ultrasonic echo
C. The sound travels poorly through air
D. The velocity is slower through a denser materials
E. The velocity is dependent on the temperature of the material through which it travels

Q 6. Which of the following statement is not correct regarding physics of ultrasound?

A. A thicker piezoelectric crystal has a longer wavelength
B. A thicker piezoelectric crystal has a lower resonance frequency
C. Acoustic impedance determines beam refraction
D. As the angle of incidence increases less sound is reflected
E. The acoustic impedance of a material is the product of its density and the velocity of sound within it

Q 7. Which of the following statement regarding the velocity of ultrasound is correct?

A. Is about 1540 m/s through the soft tissue
B. Is faster through air than water
C. Is faster through the soft tissue than the skull
D. Is proportional to the compressibility of the medium through which the sound travels
E. It is not dependent on the frequency

Q 8. Which of the following is true concerning a laser?

A. Is an acronym for Light Amplification of Stimulated Ejection of Radiation.
B. Produces multichromatic light
C. Requires a pair of mirrors at opposite ends of an optical cavity containing the lasing medium
D. The lasing medium can be gaseous or crystalline
E. The wavelength is determined by the stimulating current

1. B   2. D   3. E   4. D   5. D   6. C   7. A   8. C

Q 9. **Which of the following classes of lasers is considered to be of low risk?**
A. Class 3a
B. Class 3b
C. Class 4
D. None of the above
E. All the above

Q 10. **Which of the following is true concerning MRI?**
A. Blood vessels appear white on scanning
B. Has no recognised side effects on the foetus
C. It involves ionising radiation
D. The pregnant mother should be turned to her right side during scanning
E. Tissue with high hydrogen concentrations are difficult to distinguish

Q 11. **Which of the following is not true concerning surgical diathermy?**
A. Bipolar diathermy delivers ten times more power than unipolar
B. Coagulation diathermy current has a pulsed sine wave pattern
C. Cutting diathermy current has an alternating sine wave pattern
D. Diathermy current has a frequency of 0.5 - 1 Hz
E. The current density is higher in unipolar than in bipolar diathermy

Q 12. **At what crown-rump length would it be expected to observe the foetal heart beat using transvaginal sonography?**
A. Greater than 6 mm
B. Greater than 5 mm
C. Greater than 4 mm
D. Greater than 3 mm
E. Greater than 2 mm

Q 13. **Which of the following is not true concerning radiation physics?**
A. A neutron has almost the same mass as a proton
B. A positron has the same size as an electron
C. A proton has a positive charge
D. An electron has a greater mass than a proton
E. The hydrogen nucleus is a proton

Q 14. **Which of the following is true regarding hysterosalpingography?**
A. It should be performed in the luteal phase of the menstrual cycle
B. It requires the use of ultrasound
C. It may be used for investigating both primary and secondary subfertility
D. A pregnancy test may be required before the procedure
E. It is contraindicated in the individuals with the history of chlamydia

Q 15. **The standard chest X-ray is equivalent to what duration of natural background radiation?**
A. 7–10 days
B. 10 days
C. 2 months
D. 10 months
E. 2 years

Q 16. **Which of the following is true regarding electrosurgery?**
A. Bipolar diathermy can be used for cutting tissues
B. Monopolar diathermy necessitates the use of a return electrode
C. Diathermy uses low-frequency alternating current
D. Desiccation of the tissues is achieved only via bipolar diathermy and not by unipolar diathermy
E. Direct coupling is achieved by adhering to strict safety protocols

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9. A   10. B   11. E   12. E   13. D   14. C   15. A   16. B.

Epidemiology/Statistics

Epidemiology

Prevalence

The prevalence is the proportion of people in the entire population who are found to be with a disease at a certain point in time. Prevalence illustrates how often a situation occurs and is expressed as a proportion. It depends on the number of individuals who contract the disease in a particular time period.

Since the prevalence looks at the number of individuals with a disease at a given point in time, or within a defined time interval, if a patient has recovered from the illness in that duration then they would not be included in the prevalence rate. Prevalence is one measure, which can assess the health requirements of a community. The prevalence rate of a disease has the following features:

❏ It can be estimated from a cross-sectional study.
❏ It can be used to determine the health needs of a community.
❏ It is dependent on the duration of illness.
❏ It is dependent on the incidence of the disease.
❏ It measures all the current cases in the community.

Incidence

The incidence of the condition is the number of newly diagnosed cases of the condition over a specific time period. The incidence tells how common a situation is.

Evaluation of a Clinical Test

A clinical diagnostic test can normally be evaluated using four indices: sensitivity, specificity, positive predictive value and the negative predictive value. Sensitivity and specificity are most easily understood from a 2 by 2 table **(Table 10.1)** which gives the number of subjects showing positive and negative tests, and with and without disease in a cross-sectional random sample from a target population. It is only for such a sample that all four indices can be calculated from the same data.

Specificity

The specificity of a test is the probability of a negative test given the absence of the condition. Specificity is equal to true negatives/(true negatives + false positives)

Sensitivity

The sensitivity of a test is the probability of the test being positive in somebody with the condition. Sensitivity indicates the proportion of individuals with positive screening tests that actually have the disease. Sensitivity is equal to true positives/(true positives + false negatives).

TABLE 10.1	Simple formulas for calculating the sensitivity, specificity, positive predictive value and the negative predictive value		
	Disease present	**Disease absent**	
Test positive	a (true positive)	b (false negative)	a + b
Test negative	c (false negative)	d (true positive)	c + d
	a + c	b + d	a + b + c + d

a = true positives (Persons who have disease and are test positive on the screening test)
b = false positives (Persons who do not have disease and test positive on the screening test)
c = false negatives (Person with disease but with negative test)
d = true negatives (Persons who do not have disease and are test negative)
Sensitivity = a / a + c (Ability of the test to be correctly positive amongst those who are known to have the disease)
Specificity = d / b + d (Ability of the test to be correctly negative among those who are known to be without disease)
Positive predictive value = a / a + b (Ability of the test to correctly predict presence of the disease)
Negative predictive value = d / c + d (Ability of the test to correctly predict the absence of disease)

Positive Predictive Value

Positive predictive value can be defined as the probability of having a condition if the test is positive. It is the probability of an event in the active group divided by the probability of the event in the control group. It is calculated as follows:

Positive predictive value = true positive/true positive + false positive

Negative Predictive Value

Negative predictive value can be defined as the proportion of individuals with negative tests that are free of the condition. In other words, it is the probability that the person who is test negative is actually a true negative. It is calculated as follows:

Negative predictive value = true negative/true negative + false negative

Disease Causation

Disease is a dynamic process and denotes disharmony and deviation from the normal functioning of the various bodily functions. Besides the causative agent, there are many factors related to the man and environment, which may be responsible for disease causation. Criteria for assessing causation include:

- *Strength*: Strong associations are more likely to result in a disease in comparison to the weak ones.
- *Consistency*: Multiple studies finding the same thing are more likely to be causal.
- *Specificity*: If a variable is associated with a single outcome, and the outcome is associated with only a single possible cause, then the relationship is more likely to be causal.
- *Temporality*: Causes must definitely precede effects to suggest causation.
- *Biological gradient*: If an increased exposure is associated with an increased rate or severity of disease then causality is more likely.
- *Plausibility*: Hypotheses should sound reasonable (apparently valid).
- *Coherence*: Causal association is strengthened if epidemiological data fit in with pathology.
- *Experiment*: If the cause is removed and disease frequency declines, the likelihood of a causal link is strengthened.
- *Analogy*: If a similar association has been shown to be causal then the association under investigation is more likely to be causal.

Screening Program

The following criteria must be met before a screening program can be successfully implemented for disease detection and screening:

- The disease entity needs to be well characterised.
- Appropriate treatment/interventional strategies must be in place to provide a positive outcome due to early detection and for the test to be well validated and acceptable. It does not necessarily have to be painless.
- Natural history of the disease must be clearly defined.
- There should be an effective treatment available for the condition.
- There should be evidence that the screening test is effective at reducing morbidity or mortality.
- Benefits of the screening test should outweigh potential harm of diagnosing and treating an asymptomatic individual picked up through screening.

Variables

Variable can be defined as a value or a characteristic that can be changed. It is generally useful for the purpose of research studies. Two types of variables have been described: discrete variable and continuous variable.

Discrete variable: A discrete variable is a variable, which can take on only a certain number of values, i.e. they cannot have an infinite number of values. These variables do not change with time and circumstance, e.g. gender and ethnicity.

Continuous variable: Continuous variable are variables, which can take up an infinite number of values. These variables change with time and circumstances. Continuous variables include blood pressure, height, weight, plasma concentrations, etc.

Reliability

The reliability of a test is the ability of a test to produce the same result when repeated under identical conditions.

Relative Risk

The ratio of the risk of disease in subjects with a given characteristic compared with those without that characteristic.

Confounding Factor

It is a factor, which is associated with both the exposure and the outcome under scrutiny. Confounding factors are of major concern to the design of a study and are an important source of error. Where possible, study design attempts to suppress confounding factors as far as possible.

Percentile

It is a measurement used in statistics indicating the value below which a given percentage of observations in a group would fall. This results when the frequency is divided into 100. For example, 10th percentile is the value below which

10% of the population lies and above which 90% of the population lies.

Null Hypothesis

Null hypothesis is a hypothesis, which says that there is no difference between the two sets of data. Rejecting or disapproving the null hypothesis implies that the researcher must reach a conclusion that there exists a relationship between two phenomena (i.e. the potential treatment produces a measurable effect). For this, there should be clear set of definitions for both the outcome and exposure or treatment. The null hypothesis is rejected if the P value is less than the significance level, that is, P less than 0.05.

On the other hand, the alternative hypothesis is a one in which there is a difference between the two groups.

Confidence Interval

A confidence interval (CI) is a statistical range with a specified probability that a given parameter lies within the range. While interpreting a result from the sample, it is useful to express it within a range and enlist possible values it might have taken if other samples of the same size have been selected. This range of values is known as CI and is usually set as a percentage. Commonly used CI in the research studies is 95% CI because this corresponds to the P value of 0.05. CIs allow a true estimate of the breadth of a difference, reducing the likelihood of a type II error. The CIs decrease as the population sample increases.

Ninety five percent CIs can be used for both distributional and distribution-free data. A 95% CI points towards the range of values within which the researcher can be 95% confident that the true population parameter lies. CIs increase the accuracy when comparing means with another population by looking at the spread of differences. A wide CI indicates that the estimate is imprecise. If the 95% CI crosses zero (the value of no difference), it indicates that the treatment has no effect. The 95% CI may be calculated as the mean ± 1.96 times the standard error of the mean (SEM) for population greater than 30.

Probability

Probability (P) can be defined as the frequency with which an event would occur at random, given a total frequency of 1. A P value of 0.05 indicates that an event would occur randomly 5 out of 100 times or to put it simply, 1 out of 20 times. P value of 0.05 is traditionally considered as the cut-off point for statistical significance. P value of 0.05 or below is considered non-random or statistically significant. Unfortunately, statistical significance does not connect with clinical significance. For example, a doubling of risk from 0.0001% to 0.0002% may be completely insignificant, even though it is statistically highly significant.

Statistical significance is usually accepted at the P value less than 0.05. This implies that in less than 5% of occasions could such an observation be seen. A P value of 0.01 implies that the observation would be seen to occur by chance in 1 out of 100 occasions.

Power

Statistical power of the study is the ability of the study to detect a statistically significant difference between the two groups (i.e. if the difference really existed). Power of a study increases with the increasing sample size. This means that a large sample has a greater ability to detect a clinically important effect, if it exists.

Obstetrics Epidemiology

Perinatal Mortality

Perinatal mortality rate (PMR) is defined as the number of stillbirths and deaths in the first week of life per 1,000 total deliveries. It is calculated using the following formula:

$$PMR = \frac{Total\ mortalities}{Total\ mortalities + live\ births} \times 1,000$$

The rate is marginally higher in boys. PMR is lowest in mothers belonging to the age group 25–29 years and is higher amongst teenagers and women of advanced age (>35 years). Other risk factors for increased PMR include extremes of birth weight, nulliparity, obesity, race (highest risk in non-hispanic women), poor maternal health, smoking and low levels of maternal education. The stillbirth rate in the UK was 5.3 per 1,000 in 2006, statistically no better than the figure of 5.4 per 1,000 in 2000.

The WHO criteria for calculation of the perinatal mortality rate differ from those of the United Kingdom in the following aspect: PMR for WHO ranges from stillbirths at 22 weeks (WHO ICD 10 code suggests neonates greater than 500 g in weight) to neonates (less than 7 days old). On the other hand, in the UK, the legal definition of a stillbirth was altered to 24 weeks' gestation instead of 28 weeks or more in 1992. Therefore, in UK, stillbirth can be defined as a baby born after 24 weeks of life, showing no signs of life.

Stillbirth Rate

This can be defined as number of stillbirths per thousand total births (both live births and stillbirths).

Neonatal Deaths

Neonatal death can be defined as deaths (per 1,000 live births) occurring within 28 days of birth. Early neonatal deaths are deaths occurring within 7 days of birth (first week of life).

Infant Mortality Rate

Infant mortality rate (IMR) can be defined as number of infant death (deaths during first year of life) per 1,000 live births.

Statistics
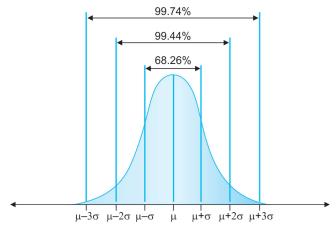
Distribution

The most commonly used type of distribution in medical statistics is the normal or Gaussian distribution. It forms the basis for many aspects of medical statistics. It is commonly used for large samples of observation (>20). Another type of distribution is the t distribution, which forms the basis for the Student's t-test.

Normal (Gaussian) Distribution

This is one of the most important and widely used methods for probability distribution in medical statistics. It generates a bell-shaped curve if it is plotted in terms of probability **(Fig. 10.1)**. 95% of the area under the curve lies between the points that fall 1.96 standard deviation on the either side of the mean value. In this case, 2.5% of individuals will be above and 2.5% below. 90% and 99% of the values respectively fall 1.65 and 2.58 standard deviations on the either side of the mean value. 68% of observations lie between 1 standard deviation on the either side of the mean value. If a characteristic is normally distributed in a population, the mean, mode and median are all equal and therefore they coincide. Data from a normal distribution are suitable for parametric tests without prior transformation.

Different tests for null hypothesis have been developed for the data that are hypothesised to follow normal distribution. Some of these tests are Z-test for normal distribution, t-test for student's t-distribution, and f-test for f-distribution. Pearson's chi-squared test can be used for data, which does not follow normal distribution. Chi-squared test is the standard method for comparing distributions, e.g. between the observed and expected

FIG. 10.1: Bell-shaped curve demonstrating normal/Gaussian distribution (μ is the mean and σ is the standard deviation)

frequency of a given event. Chi-squared test assesses proportions and does not depend upon normality. This is derived by squaring the normal distribution.

Student's t-test is a standard test for comparing the difference between the two means. On the other hand, f-test is the standard method for comparing the size of variance.

Parametric Tests and Non-parametric Tests

Parametric tests are ones where assumptions are made about the defining properties (parameters) of the population distribution from which one's data are drawn. On the other hand, non-parametric tests are ones where no such assumptions are made. Usually, the parametric tests are more powerful and sensitive than the non-parametric tests. Parametric tests are preferred over the non-parametric tests because fewer observations are required to provide evidence in favour of the hypothesis if it is true, e.g. student's t test is a typical parametric test for comparing the mean values of a continuous variable between two groups. In particular, t-tests and one-way analysis of variance (ANOVA) require that the data should have a normal/Gaussian distribution, and these tests are invalid if the data are non-parametric.

Example of a non-parametric test is Mann-Whitney U test, which ranks the observations in the order of size and compares the proportions, which fall above and below the median value for each of the groups in question. Wilcoxon signed rank test is another example of a non-parametric test. Best way of deciding whether to use parametric or non-parametric tests is by evaluating if the continuous data in the sample can be made to follow a normal distribution or not. Non-parametric tests are usually employed in cases with sample size less than 15 observations, unlikely to have a normal distribution. On the other hand, parametric tests are used in cases with more than 20 observations, with the data following a normal distribution.

Skewed Distribution

In case of normal distribution, the numbers of individuals above and below the mean will be equal; if they are not equal then the distribution is said to be skewed. For positively skewed data, the mean is larger than the median and the mean usually lies to the right of the mode.

On the other hand, for negatively skewed data, the median is larger than the mean. When summarising skewed data, it is better to quote the median and the interquartile range, whereas while summarising the normally distributed data, the parameters most commonly used include the mean and the standard deviation. In skewed data the geometric mean is the most appropriate measure.

Mean

The mean (denoted by the Greek letter mu, μ) is the average of the data. In case of normally distributed data (giving

a bell-shaped Gaussian curve), the mean is same as the median and mode. However, in case of data, which is not normally distributed the mean, mode and the median may be different values.

The arithmetic mean is calculated by using sum of the values [(a1+ ...+aN)/N]. On the other hand, geometric mean is defined as the nth root of the product of n numbers [n√(a1.a2.a3....aN)]. It indicates the central tendency for a set of numbers by using the product of their values. Hence, the geometric mean will always be less than the arithmetic mean. The arithmetic mean is preferred in normal statistics as it generally represents the average.

Median

The median is the middle observation value. Therefore, half the observations lie above this value and the other half lies below it. Sometimes, it may be more appropriate to quote the median rather than the mean because it is less sensitive to the large outlying values. Also, in case of skewed data, it is better to quote the median rather than the mean.

Mode

The mode is the observation that occurs with the greatest frequency. Mode refers to the most frequently encountered value and in a normally distributed data coincides with the mean and median values.

Range

Range denotes the total collection of values between the largest and the smallest observation. This indicates how widely varied the data is. It is calculated by subtracting the lowest value from the highest value.

Interquartile Range

This is similar to the median although the lower value (Q1) indicates that 25% of the observations lie below it and the upper value (Q3) indicates that 25% of the observations lie above it. Thus, the interquartile range represents the central 50% range of values (Q3 – Q1). For the calculation of interquartile range, firstly the data is arranged in an ascending or descending order. The list is then cut into four equal parts by making three cuts, Q1, Q2 and Q3 (where quartiles are the cut). As previously described, interquartile range = Q3 – Q1.

Variance

The variance can be defined as the spread of the observations. This is an indication of variability of the observations. To calculate the variance, each observation is subtracted from the mean, squared, added-up and divided by the number of observations minus one.

Correlation

This is the relationship between the two variables (between a dependent variable and one or more independent variables).

Linear Regression

This is the standard method for determining whether two variables are correlated or not. Linear regression can be of two types: simple and multiple linear regression. Correlation between one independent variable and one dependent variable is known as simple linear regression. Correlation between more than one independent variable and one dependent variable is known as multiple linear regression.

Coefficient of Correlation

This is indicative of the degree of correlation between the two variables, X and Y. The symbol denoting coefficient of correlation is r. Correlation coefficients vary between –1 and +1. A value of 1 is indicative of perfect correlation, whereas the value of 0 indicates no correlation at all.

Standard Deviation

Standard deviation (SD), represented by the Greek letter sigma (σ), is known as the root mean square deviation is the square root of the variance and is a measure of distribution of the data. The SD provides a good indication of distribution about the mean. It is a measure of observation variability and is greater than the SEM.

Standard Error of Mean

Standard error of mean can be defined as the standard deviation of the sampling distribution of a statistical measurement, most commonly mean. This can be defined as standard deviation divided by √n or √n—1.

$$\text{SEM} = \frac{\sigma}{\sqrt{n}} \text{ or } \frac{\sigma}{\sqrt{n-1}}$$

where SEM is standard error of mean
σ is standard deviation
n is the size (number of observations)

This gives an estimate of the limits to the mean of the total population from which the sample observations were drawn. In other words, SEM quantifies the accuracy with which the researcher knows about the true mean of the population. By definition, SEM is always smaller than the standard deviation. Also, SEM gets smaller as the samples become larger since mean of the large sample is likely to be closer to the true population than is the mean of a small sample. If testing to see if there is a difference between two population means (for example, t-test) then t = mean/SEM.

Error

Error is any random source of inaccuracy. For example, an error at the time of collecting variables is likely to lead to a less precise estimate. There are two kinds of statistical errors that can be made in significance testing.

Type I Error

Type I error occurs when the sample used in the experiment generates a significant result for the hypothesis, but this would not be significant if performed on the population. In other words, it is incorrect rejection of a true null hypothesis (a false positive). This implies that there is detection of an effect that is not present. The type I error rate increases as the number of comparisons increases, leading to spurious conclusions. The probability associated with type I error is the significance level.

Type II Error

Type II error occurs when the sample used in the experiment fails to generate a significant result for the hypothesis, but there would have been a significant result if the experiment is performed on the population. In other words, it is failure of rejection of a false null hypothesis (a false negative). This implies that there is failure of detection of an effect that is present.

Data Measurement

Methods for medical data measurement have been tabulated in **Table 10.2**. Measurements may be performed for either continuous or categorical data, which has been classified into single or two groups.

Odds Ratio

This can be defined as the ratio of the number of subjects classified in one category to the number of subjects classified in another category. Odds ratio cannot be used for

TABLE 10.2	Methodology for data measurement	
Continuous data	**Categorical data**
Single group • Normally distributed: Mean, standard deviation, 95% confidence interval • Non-normal: Median, range, interquartile range	Single group • Rate (risk) • 95% confidence interval
Two groups • Mean difference • 95% confidence interval	Two groups • Risk difference, 95% confidence interval • Relative risk, 95% confidence interval • Odds ratio, 95% confidence interval

time-dependent data unless all subjects have been followed for the same length of time.

Relative Risk

Relative risk is a measure of how much a particular risk factor (e.g. cigarette smoking) influences the risk of a specified outcome (e.g. lung cancer in this case), relative to the risk in the population as a whole.

Placebo Effect

Placebo effect is a beneficial effect occurring in a patient following a particular treatment that arises from the patient's expectations concerning the treatment rather than from the treatment itself. Studies reveal that this effect can be potent. Although one might think that placebo has no effect, use of placebo may be associated with a considerable psychological effect. Placebo studies are undertaken in patients with cancer, particularly to establish the palliative value of drugs or the effectiveness of a new treatment where none exists.

Bias

Bias refers to systematic inaccuracy either due to consistent over-recording or under-recording and is usually due to recorder bias, which may be subconscious. The two most common types of bias encountered in the epidemiological studies are the selection bias and the responder or the observer bias.

Selection Bias

It occurs when subjects that are chosen are not representative of the target population. This implies that the sample has not been collected at random from the population. Selection bias is an error in choosing the individuals or groups to take part in a study. Ideally, the subjects in a study should be very similar to one another and to the larger population from which they are drawn (for example, all individuals with the same disease or condition). If there are important differences, the results of the study may not be valid. Using randomised controlled trials is the best way for reducing the selection bias at the time of investigating the impact of treatment on the disease. The main advantage which randomisation offers is concealment, implying that when a decision is made to enter a subject into the trial, nobody knows what kind of treatment would be allocated to that patient. Example of selection bias: post-menopausal women from various health clubs are selected to participate in a study of hormone replacement therapy. In this case, there would be selection bias as the groups from the health clubs are very unlikely to be representative of the true population.

Observer/Responder Bias

Observer bias occurs when the investigator is aware of the disease status, treatment group or outcome of the study. In these cases, the ability of the investigator to interview the subject and collect or analyse data in an unbiased manner is compromised. Cognitive bias on the behalf of the investigators causes an unconscious influence on the participant. This bias may cause the researcher to interpret the results incorrectly because of the tendency to look for information, which conforms to their hypothesis, and overlook the information, which argues against it.

Also, the study subjects may respond differently to their levels of exposure if they have been classified as a subject with or without the disease under investigation.

Recall bias can also be challenging in cross-sectional and case-control studies. This can be reduced to minimum by keeping the hypotheses of the study undisclosed to the participating subjects. This, however, has become increasingly difficult in recent years because of the policy of informed consent, which involves provision of full information to the subjects. Whenever possible, both the observer and the study subject must be blinded to both the treatment and hypothesis of interest.

To reduce such bias various types of clinical trials are employed is clinical practice.

Clinical Trials

Results of all trials may be valid irrespective of whether they are double blind or not. However, inaccuracies can occur due to too small sample size. Again, bias may arise due to the withdrawal of selective patients and this may be positive or negative. Different types of clinical trials are described next.

Different Types of Clinical Trials

Double-Blind Trial

Double-blind study refers to a type of research study in which neither the study participants nor the person giving the treatment knows which treatment a particular subject is receiving. This helps in alleviating potential bias through randomisation of patients to the drug or placebo without either the doctor or the patient knowing which agent is being used. In this way, both the researchers and the study participants are "blind" to which subject is receiving what type of treatment during the study. This method helps researchers get more accurate results from their research. Double blinding allows researchers to "control" a study for the psychological effects that sometimes help people feel better, simply because they expect to feel better when they receive a medication. In other words, double blinding helps

researchers separate the "power of suggestion" from the real effects of a medication or therapy.

Placebo-controlled studies are most appropriately undertaken in a double blind fashion with both the observer and the patient blinded to treatment. Although one might think that placebo has no effect, in fact there may well be a huge placebo (psychological) effect. Placebo studies are undertaken in patients with cancer, particularly to establish the palliative value of drugs or the effectiveness of a new treatment where none exists. As a variation of this theme, patients can be randomised to receive either the new drug or an established therapy.

Example of a double-blinded trial is a study conducted amongst a group of 500 patients for assessing the effectiveness of a new anti-rheumatic agent. The drug company randomises the patients to receive either placebo or the active drug although neither the patients nor investigators know which treatment they are receiving.

Single-Blind Trial

This is a single-blind study where the patient does not know which arm of therapy they are receiving. However, the investigator does have this information. For example, a study is undertaken assessing the effects of a cholesterol-lowering agent on cardiovascular disease. Patients are randomised by the investigators to receive either the drug or the placebo. However, the patients are unaware regarding the kind of treatment they would receive.

Randomised Controlled Trials

These trials are used for investigating the effects of therapies and interventions in the course of a particular treatment. Subjects with a particular condition who meet the inclusion criteria for entry into the trial are randomly allocated to either receive treatment or some form of control (either no treatment or the current, "gold standard" treatment). A computer program is used for randomising the data in different participating centres. Various participating centres are also linked with telephone services to request the randomisation procedure. Wherever possible, double blinding should be performed, i.e. both the subjects and investigators must be blinded to the randomised allocation and the control group should be provided with some form of placebo. The outcome of interest is measured to evaluate if one group experiences any benefit over the others.

The main advantage of the randomised control trial is the concealment of the treatment allocation and thus minimisation of the selection bias and low chances of confounding. These studies are, however, time-consuming and expensive. Also, there can be an appreciable loss to follow-up if the infrastructure is not in place to ensure good data collection.

Different Types of Epidemiological Studies

Epidemiology is concerned with the investigation of health and illness across and within the population of people. Numerous methods of study design have been developed for the analysis. Various types of studies can be categorised into two major types: experimental and non-experimental studies. Non-experimental studies are mainly concerned with disease causation and progression, while the experimental studies are concerned with the disease treatment or prevention.

Case-Control Study

This is a study comparing the characteristics of subjects selected on the basis of their disease status. A case-control study compares two groups of people: those with the disease or condition under study (cases) and a very similar group of people who do not have the disease or condition (controls). While the cohort study can be visualised as a prospective study, following subjects forward through time, case-control study is a retrospective study. This study involves the recruitment of cases (subjects with the disease) and a group of control (subjects without the disease), both of which are looked back through time to compare their exposures to evaluate if any of the exposure appears to relate to the disease development. Case-control studies are relatively cheap in comparison with the cohort studies and can be used to investigate a number of different exposures simultaneously. The main difficulty associated with this type of study is the selection of the control group. Also, these studies can be often associated with considerable problems related to the selection bias and observer or recall bias. These studies differ from the cohort studies regarding the fact that cohort studies are not good at investigating rare exposures as a large number of subjects need to be recruited to obtain enough evidence of the exposure. Moreover, case-control studies cannot be used for making estimates of disease incidence and are not very helpful in investigating the series of events resulting in the disease diagnosis. Presence of confounding factors can also pose further problems.

Cross-Sectional Studies

In these studies, the data are collected from a sample of subjects at a given point of time and comparisons are made between the variables to investigate the extent of disease of interest or to assess which exposures may be linked with the disease. These studies represent a snapshot in time and therefore the prevalence is generally the main outcome measure. Moreover, no information is obtained on the disease incidence over time.

Open Study

Open study is a type of study in which both the researchers and the participants know which treatment is being administered. For example, a study compares the effect of low-molecular-weight heparin versus aspirin in the prevention of deep vein thrombosis (DVT) amongst post-operative gynaecological patients. Patients are randomised by the study co-ordinator to receive treatment as either a tablet or injection. This study assessing DVT post-operatively is an open study as patients and investigators will know which treatment they are receiving as it is either an injection or a tablet. If however, they wished to create a double blind study then patients could be randomised to receive either injection plus placebo tablet or aspirin plus placebo injection.

Cohort Studies

Cohort studies or longitudinal studies involve the follow-up of individuals. Subjects are recruited into cohort studies and followed-up over time to assess the incidence of a particular disease. In case of a disease that has already been diagnosed, disease progression can be assessed. A cohort study is prospective in the sense that the individuals who are exposed and non-exposed to a putative risk factor are followed-up over a defined period of time and the disease experience of the exposed group at the end of follow-up is compared with that of the non-exposed group. Cohort studies are more important than the cross-sectional studies because they provide far more information on the incidence of events. These studies also allow temporal assessments to be made regarding whether the exposure preceded the outcomes of interest or not.

A cohort study may also be historical (retrospective or non-concurrent). Cohort studies are, however, not useful at investigating rare studies. They are frequently used when the disease is common and the effects of various exposures are not well understood.

Meta-analysis

Analysis of data from published literature is termed as meta-analysis. Meta-analysis is a common way of assessing the effect of treatment or the potential risks of treatment by reviewing and assessing all the data published in the medical literature. Many of the guidelines are published through meta-analysis. For example, the study, which analyses the side effects reported in all published data relating to the use of COX-2 inhibitors, is a meta-analysis.

Choose the Single Best Answer (SBA)

Q 1. The incidence of a disease refers to which of the following?

A. The number of beds occupied in a designated population with the condition
B. The number of new cases emerging in a designated period and population
C. The period prevalence of an illness
D. The point prevalence of an illness
E. The readmission rate

Q 2. Based on the recommendations by the World Health Organisation, calculation of the perinatal mortality rate (PMR) should involve which of the following?

A. Be expressed as deaths per thousand live births
B. Includes all deaths occurring in the first month of life
C. Includes all foetuses and infants of gestational age of more than 20 weeks
D. Includes all foetuses and infants weighing 500 grams or more
E. Includes all foetuses and infants with a crown rump length of more than 35 cm

Q 3. Which of the following statement is correct if a characteristic is normally distributed in a population?

A. 5% of individuals will be more than two standard deviations from the mean
B. The mean will be greater than the median
C. The median will be less than the mean
D. The mode will be greater than the median
E. The numbers of individuals above and below the mean will be equal

Q 4. The National Screening Committee criteria for establishing a population-screening programme do not include which of the following?

A. Clearly defined natural history of the disease
B. Should be entirely painless
C. There should be an effective treatment available for the condition
D. There should be evidence that the screening test is effective at reducing morbidity or mortality
E. Benefits of the screening test should outweigh potential harm of diagnosing and treating an asymptomatic individual picked up through screening

Q 5. Which of the following describes the observation that occurs with the greatest frequency?

A. Correlation
B. Mean
C. Median
D. Mode
E. Variance

Q 6. Which of the following statement is correct regarding statistical distribution?

A. The mode is the value that occurs the most frequently
B. The median is that point on the scale of measurement above which lie exactly half the values and below which lie the other half
C. Of a normally distributed variable, the probability of attaining a value higher than two standard deviations above the mean is approximately 1 in 40 (p = approximately 0.025)
D. Having a normal distribution, approximately 95% of the values will lie within the range between (mean +2 standard deviations) and (mean -2 standard deviations)
E. All the above

Q 7. Which of the following is true regarding the standard deviation?

A. SD = SEM/(square root of n)
B. The SD equals the SEM in non-parametric tests
C. The SD is a measure of observation variability
D. The SEM determines the accuracy of measurement of the observations
E. The standard deviation (SD) is equal to the standard error of the mean (SEM)

Q 8. Which of the following is true if a characteristic is normally distributed in a population?

A. The median will be less than the mean
B. The mean will be of lesser value than the mode
C. The numbers of individuals above and below the mean will be equal
D. This implies that most of the population comprises of normal individuals
E. 20% of individuals will be more than two standard deviations from the mean

Q 9. For the data series: 2, 1, 6, 4, 2, which of the following statements is true?

A. The mean is 6
B. The mean is always identical to the median
C. The median is 3
D. The mode is 4
E. The standard deviation is 2

Q 10. Which of the following is true regarding the standard deviation of a group?

A. Is the square of the variance
B. Is a valid statistical parameter for observations that do not have a normal distribution
C. Is numerically equal to the standard error of the mean
D. Can be used for the calculation of Chi squared test
E. Is a measure of the scatter of observations about the mean

1. B   2. D   3. E   4. B   5. D   6. E   7. C   8. C   9. E   10. E

Q 11. **Which of the following statements related to statistics is not correct?**

A. In a normally distributed population, 95 percent of the values fall in the range of the mean plus or minus two standard deviation

B. Standard error and standard deviation are synonymous

C. The chi-squared test can be applied to non-parametric data

D. The statement "p is less than 0.01" means that there is less than 1 percent likelihood of an event having occurred by chance

E. Variance is equal to the square of the standard deviation

Q 12. **In a trial of a new therapeutic agent, the required sample size varies with which of the following?**

A. Experience of the investigator

B. Level of acceptance of failing to discover a true effect

C. Level of statistical significance required

D. Type of statistical test to be employed

E. None of the above

Q 13. **A report of a clinical trial of a new post-operative analgesic states, "In a comparison between the new drug and a placebo, a higher proportion of patients taking the new drug obtained relief from pain (p < 0.05)".**

In consequence, which of the following statements is correct?

A. The trial was well designed

B. Amongst 100 patients treated with the drug five would be expected to have a placebo response

C. The result may have occurred by chance alone in less than one in 20 occasions

D. The probable error of the observations is +/- 5%

E. The result should be regarded as not reaching conventional levels of statistical significance

Q 14. **A diagnostic test has a sensitivity of 90% and a specificity of 95%.**

Which of the following statements is likely to be true in this case?

A. 10% of positive tests are false positive

B. 5% of negative tests are false negative

C. Sensitivity is equal to true positives / (true positives + false negatives)

D. Sensitivity indicates the test will be negative in disease.

E. None of the above

Q 15. **In a clinical trial of a new drug treatment for inflammatory bowel disease, the following results are obtained:**

	Drug	Placebo
Improved	46 patients	34 patients
Not Improved	14 patients	26 patients

Which of the following statement is true concerning this data?

A. Data can be evaluated by computing a Chi-squared test

B. Data can be evaluated using a Student's t-test

C. If the statistical probability that the difference between drug and placebo is 0.1 then the drug can be introduced into clinical practice

D. Pearson's coefficient of linear correlation could be used to test significance

E. The superiority of the drug over placebo is so obvious that formal statistical testing is unnecessary

Q 16. **Which of the following is not true concerning the use of a placebo in a clinical trial?**

A. Is associated with no effects

B. Should be identical in appearance to the drug being studied

C. Placebo studies are undertaken in patients with cancer

D. Is pharmacologically inert

E. Is best administered by a person who is unaware of the drug's identity

Q 17. **A random selection of 1,200 adults agrees to participate in a study of the possible effects of drug X. They are followed prospectively for a period of five years to see if there is an association between the incidence of cataract and the use of drug X. What type of study is this?**

A. Case-control study

B. Cohort study

C. Cross-over study

D. Cross-sectional study

E. Randomised controlled clinical trial

Q 18. **Which of the following is true when evaluating a report of a clinical trial?**

A. Control and treatment groups must be equivalent in size

B. Even if randomisation is conducted properly, chance differences are inevitable

C. Adequate sample size commonly produces false positives and false negatives

D. Results are invalid if the trial is not of double-blind construction

E. Withdrawal of patients from a trial by the investigator does not cause any bias

Q 19. **In a double-blind placebo control clinical trial, which of the following statements is correct?**

A. Some of the patients are not treated

B. All the patients receive a placebo

C. The patients do not know which treatment they receive

D. Everybody receives both treatments

E. The clinician assessing the effects of the treatment knows which treatment the patient has been given

Q 20. **Which of the following statements concerning statistical tests is true?**

A. Wilcoxon's rank test needs equal sample sizes

B. Student's t-test is a non-parametric test

C. Correlation coefficients vary between −10 and +10

D. Sigma (σ) is the symbol denoting coefficient of correlation

E. $y = a + bx$ is a regression equation

11. B   12. C   13. C   14. C   15. A   16. A   17. B   18. B   19. C   20. E

Q 21. Which of the following is not true regarding the occurrence of errors in clinical trials?

A. Errors are more common when big samples are used
B. Type I error is more likely to occur when multiple t-tests are used
C. Type I error is wrongly rejecting the null hypothesis
D. Type II error is accepting the null hypothesis when it is invalid
E. Type II error is reduced by the use of a confidence interval

Q 22. In comparing confidence intervals with P values, which of the following statements is correct?

A. 95% confidence intervals are equivalent to a P value of 0.95
B. As the sample size increases, the confidence interval will increase
C. Confidence intervals refer to the target population
D. The confidence interval is approximately equal to the standard deviation
E. The P value measures statistical significance of result

Q 23. Which of the following is true regarding 95% confidence interval?

A. Can only be used in parametric data
B. It is a test of the null hypothesis
C. If zero difference lies within the 95% when comparing two groups to a treatment, it indicates the treatment is very effective
D. Is not useful when comparing data with another population
E. It is calculated at ±1.96 times the standard error of the mean

Q 24. Which of the following is standard methods of reporting the results?

A. Mean, standard deviation, and 95% confidence intervals
B. Median, range, and interquartile ranges
C. Odds ratio and 95% confidence interval
D. Rate and 95% confidence interval
E. Risk difference

Q 25. Which of the following is not a continuous variable?

A. Blood pressure
B. Gender
C. Haemoglobin concentration
D. Height
E. Plasma glucose concentration

Q 26. Which of the following is true in significance testing?

A. A Type I error is to reject the alternative hypothesis when it should be accepted
B. A Type II error is to accept the alternative hypothesis when it should be rejected

C. The probability associated with a Type I error is the significance level
D. The significance level is always set to 5%
E. The significance level is determined at the end of a significance test

Q 27. Which of the following statements regarding statistical evaluation is not correct?

A. The arithmetic mean is the measure to be preferred in data which are symmetrically distributed
B. The geometric mean is always less in value than the arithmetic mean
C. The median is also called the measure of central value
D. The standard deviation is a good indication of distribution about the mean
E. The value of the variable which occurs with the least frequency is the mode

Q 28. Which of the following statements regarding statistical terms is true?

A. In a positively skewed distribution, the mean always lies to the left of the mode
B. In distributions which are markedly skewed, the arithmetic mean is a more appropriate measure than the geometric mean
C. In non-parametric data, the mode is usually different in value from the mean
D. All the above
E. None of the above

Q 29. A cohort study suggests a statistical link between drinking a specific local herbal tea and the development of oesophageal cancer.

Which of the following would suggest that the link is causative?

A. An odds ratio of 8:1
B. The finding of similar results in a number of studies
C. The finding that increasing consumption is associated with an increased rate of disease
D. All the above
E. None of the above

Q 30. In statistics, which of the following statements is not correct?

A. The mode of a distribution is the most frequently occurring value
B. Wilcoxon's rank sum test is a non-parametric test
C. In any set of observations, half of the observations are greater than the median
D. The chi-square test compares the observed and expected frequencies of an event
E. Infant mortality rate is the number of infants dying during the first year per 10,000 live births.

21. A   22. C   23. E   24. C   25. B   26. C   27. E   28. C   29. D   30. E
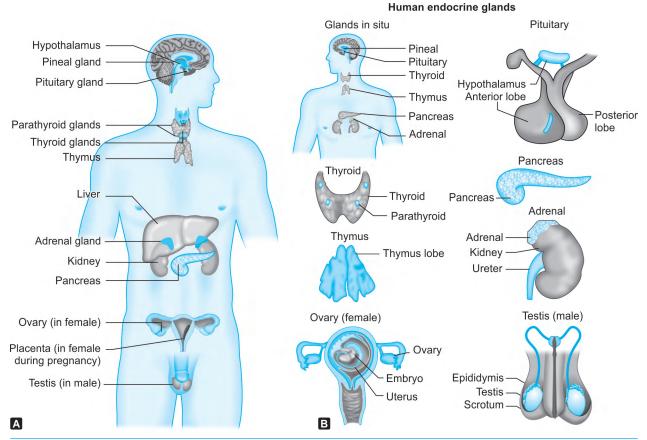
Endocrinology

General Endocrinology

Introduction

Endocrine glands are the ductless glands which synthesise and release the classical hormones into the blood circulation.

Since these are ductless, the hormones secreted by them are released directly into blood. The body's major endocrine glands are shown in **Figures 11.1A and B**. Various hormones secreted by the endocrine glands are listed in **Table 11.1**. Besides endocrine glands, hormones are also produced in the body from gonads **(Table 11.2)** and other organs **(Table 11.3)** such as thymus, kidneys, heart and placenta.

FIGS 11.1A AND B: Major endocrine glands in the body

TABLE 11.1	Various hormones produced in the body by the endocrine glands
Endocrine gland	**Hormones produced**
Hypothalamus	• Growth hormone-releasing hormone (GHRH) • Somatostatin (growth hormone-inhibiting hormone) • Thyrotropin-releasing hormone (TRH) • Corticotropin-releasing hormone (CRH) • Gonadotropin-releasing hormone (GnRH) • Dopamine (prolactin inhibiting hormone)
Anterior pituitary	• Growth hormone (GH) • Thyroid-stimulating hormone (TSH) • Adrenocorticotropic hormone (ACTH) • Follicle-stimulating hormone (FSH) • Luteinising hormone (LH) • Prolactin
Posterior pituitary	• Antidiuretic hormone (ADH) • Oxytocin
Thyroid gland	• Thyroxine (T4) • Triiodothyronine (T3) • Calcitonin
Parathyroid gland	• Parathormone
Pancreas – Islets of Langerhans	• Insulin • Glucagon • Somatostatin • Pancreatic polypeptide
Adrenal cortex	*Mineralocorticoids* • Aldosterone • 11-deoxycorticosterone *Glucocorticoids* • Cortisol • Corticosterone *Sex hormones* • Androgens • Oestrogen • Progesterone
Adrenal medulla	• Catecholamines • Adrenaline (Epinephrine) • Noradrenaline (Norepinephrine) • Dopamine

TABLE 11.2	Hormones produced by the gonads
Testis	• Testosterone • Dihydrotestosterone • Androstenedione
Ovary	• Oestrogen • Progesterone

Hormones Produced by Maternal-Foetal-Placental Unit

Placenta is not capable of independent steroidogenesis like the ovary. For steroidogenesis, it depends upon the precursors derived from the foetal and partly from the

TABLE 11.3	Hormones produced by other organs
Organ	**Hormone**
Thymus	• Thymosin • Thymine
Kidney	• Erythropoietin • Thrombopoietin • Renin • 1,25 dihydroxycholecalciferol (calcitriol) • Prostaglandins
Heart	• Atrial natriuretic peptide • Brain natriuretic peptide • C-type natriuretic peptide
Placenta	• Human chorionic gonadotropin (hCG) • Human chorionic somatomammotropin • Oestrogen • Progesterone
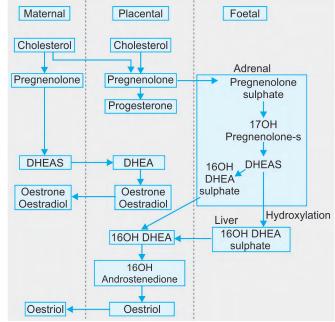
FIG. 11.2: Maternal-foetal-placental unit
Abbreviations: DHEA, dehydroepiandrosterone; DHEAS, dehydroepiandrosterone sulphate; OH, hydroxy

maternal sources. This concept is known as maternal-foetal-placental unit **(Fig. 11.2)**. Precursors from foetal origin are not required for progesterone synthesis as in oestrogen production. LDL cholesterol, derived from the mother, is used for progesterone synthesis (via pregnenolone).

Half-life

Half-life of a hormone is the time it takes for the initial concentration of a particular hormone, drug or any substance to fall by half. It is also defined as the time during which the activity or potency of a substance is decreased

to half of its initial value. Half-life is also called biological half-life. Half-life of a hormone denotes the elimination of that hormone from circulation.

Shorter half-life of a hormone allows more precise and continuous regulation, for example half-life of insulin is between 5 hours and 10 hours. This short half-life allows an accurate control of the blood glucose levels. Half-life of thyroxine is longer than that of adrenaline because moment to moment regulation of its level is less critical. Thyroxine is longer than that of triiodothyronine because it is more highly protein-bound which appears to prolong its life. Half-life of noradrenaline is longer than that of acetylcholine. Acetylcholine is broken down almost immediately by cholinesterase.

Diurnal Variation

Diurnal variation is characteristic of hormones such as cortisol, testosterone and melatonin and to a lesser extent growth hormone (GH) (pulses). During sleep there is a fall in the circulating level of cortisol, insulin and adrenaline, whereas that of antidiuretic hormone (ADH) and GH increases. Cortisol levels reduce because sleeping changes catabolic state into an anabolic state. Insulin secretion occurs mainly in association with meals, therefore it reduces during sleep. Adrenaline secretion is associated with stress, which is again reduced during sleeping. During sleep, ADH levels increase. This rises as plasma osmolality rises; water is lost but not replaced during sleep. GH levels increase, allowing growth and anabolic repair of tissue wear and tear.

Chemical Structure of Hormones

Peptide Hormones

Most hormones are peptide in nature and are composed of numerous amino acids. These hormones are secreted in form of a preprohormone, which is then first cleaved to form a prehormone, which is then cleaved to form the hormone itself. While some peptide hormones are secreted immediate, others are stored in form of secretory granules. Unlike the steroids, the vast majority of peptide hormones are lipophobic and interact with a cell surface receptor. These peptides have autocrine (acting on the cells which produced it), paracrine (acts on the neighbouring cells) as well as endocrine effects (acts on the cells at a distant site after being transported to that site via blood or the lymphatic system). These types of hormones can be divided into two groups:

1. *Short polypeptide chains and small proteins*: This group includes all the hormones secreted by the hypothalamus, posterior pituitary gland, most of the hormones of the anterior pituitary gland, thymus, digestive tract, and pancreas. It includes hormones such as ADH (nonapeptide), oxytocin (nonapeptide), GH (191 amino acids) and prolactin (198 amino acids), Adrenocorticotrophic hormone (ACTH), parathormone, calcitonin, insulin, glucagon, somato-statin, pancreatic polypeptide, hCG, human chorionic somatomammotrophin, etc.
2. *Glycoprotein hormones*: The glycoproteins include hormones such as thyroid-stimulating hormone (TSH), luteinising hormone (LH), and follicle-stimulating hormone (FSH) from the anterior pituitary gland as well as several hormones produced in other organs.

Steroid Hormones

Steroid hormones are synthesised from cholesterol and have the same basic 4-ring structure composed of 17 carbon atoms with different number of carbon atoms added in form of side chains. Three rings A, B and C are composed of 6 carbon atoms, whereas the last ring (D) is composed of 5-carbon atoms. The reproductive hormones, oestrogens and androgens, mineralocorticoids and glucocorticoids belong to the category of steroids. Oestrogens have an 18-carbon-based nucleus, whereas androgens and testosterone have a 19-carbon atom-based nucleus. Glucocorticoids, aldosterone and progesterone have 21 carbon atoms each.

The first step of steroid hormone formation from choles-terol occurs in the inner mitochondrial membrane. ACTH stimulates the conversion of cholesterol to pregnenolone, and also later biosynthetic steps (described later in the text). Ovaries produce oestrogen during the follicular phase and both oestrogen and progesterone in the luteal phase. Test-osterone, on the other hand, is produced by the interstitial cells of Leydig in the testis in response to the luteinising hormone. In some tissues testosterone is active as such, whereas in others it is converted into dihydrotestosterone by an enzyme 5-alpha-reductase. Both testosterone and dihydrotestosterone bind to a cytoplasmic receptor before passing into the cell nucleus to bind to the specific areas of the DNA to produce their effect. Steroid receptors are made up of two steroid binding units and one non-binding subunit.

Corticosteroids are degraded and conjugated with glucuronic acid in the liver, but are excreted by the kidneys. There is a delay of up to 2 days between the peak level of a steroid in blood and the peak levels of the conjugate in urine.

Amino Acid Hormones

Several hormones such as thyroid hormones, catecholamines and melatonin are composed of amino acids. These hormones are stored in form of granules. Hormones derived from the amino acid tyrosine include thyroxine (T4), triiodothyronine (T3), adrenaline (epinephrine), noradrenaline (norepinephrine) and dopamine. Melatonin, on the other hand, is derived from tryptophan.

Prostaglandins and Leukotrienes

These hormones are collectively known as the eicosanoids and are derived from arachidonic acid. Synthesis usually occurs in the cell wall.

Sex Hormone Binding Globulin

In circulation, some of the hormones are bound to the carrier proteins. However, only the free hormone or that fraction of the total hormone, which is unbound is active and is available to bind to the specific receptors to induce its effects.

All steroid hormones are bound to some extent to albumin. Some also have specific binding proteins in circulation. The majority of the principal sex steroids are bound to sex hormone binding globulin (SHBG). SHBG is a β-globulin that transports androgens [e.g. testosterone, dihydrotestosterone (DHT), androstenedione and dehydroepiandrosterone] and oestradiol in plasma. It is produced in the liver. The binding to these proteins is of much higher affinity than to albumin, but their concentrations in plasma are less. For example, SHBG binds testosterone with an affinity 50,000 times greater than that of albumin. However, none of steroids are metabolically active in the bound form. Levels of SHBG levels may be increased or decreased in the conditions enumerated in **Table 11.4**.

Serum levels of SHBG are increased by oestradiol and decreased by testosterone. Therefore, SHBG levels fall when testosterone production increases. This reduction in SHBG levels also leads to a reduction in total testosterone concentration.

Despite changes in the SHBG, free testosterone levels generally remain within the normal range because of feedback adjustment of gonadotropin secretion. This is so because increase in the binding proteins result in the reduction of the free fraction. This causes a compensatory increase in the levels of trophic hormones, thereby restoring the total amount of free hormone.

There is an increase in SHBG capacity with age. This change in SHBG concentration with age implies that a larger proportion of older men would have lower levels of free or bioavailable testosterone in comparison to the normal range for younger men.

On the other hand, increased oestrogen concentration increases SHBG synthesis in liver. Pregnancy leads to increased oestrogen concentration and thus increased SHBG synthesis **(Table 11.5)**.

Mechanism of Action of Hormones

In the body, hormones do not act directly on target cells. They firstly combine with receptor present on the target cells, resulting in the formation of a hormone-receptor complex. This hormone-receptor complex induces various changes or reactions in the target cells. Hormone receptors are the large proteins present in the target cells. Each receptor is specific for one single hormone, i.e. each receptor can combine with only one hormone. Thus, a hormone can act on a target cell, only if the target cell has the receptor for that particular hormone.

Situation of the Hormone Receptors

Hormone receptors may be situated either in cell membrane or cytoplasm or nucleus of the target cells **(Table 11.6 and Fig. 11.3)**.

Steroid hormone receptors: Receptors for steroid hormones are predominantly the nuclear receptors. They may be located in the cytosol and move to the nucleus upon activation or may remain in the nucleus waiting for the steroid hormone to enter and activate them. Each receptor is made up one steroid binding unit and two non-binding subunits. The binding unit has one binding site for steroid and one for DNA. The binding site for DNA attaches to short palindromic sequences of DNA upstream of the target gene. When the receptor molecule is not bound to the steroid, the

TABLE 11.4	Conditions resulting in the altered levels of sex hormone binding globulin (SHBG)
Reduced levels	**Increased levels**
• Obesity • Hypothyroidism • Exogenous androgen use • Nephrotic syndrome • Cushing's disease • Acromegaly • Abnormally high levels of growth hormones • Testosterone, corticoids, androgens, progestins, growth hormone, insulin and insulin-like growth factor (IGF-1) • Obesity	• Hepatic cirrhosis • Hyperthyroidism • Oestrogen use • Pregnancy • Increasing age

TABLE 11.5	Percentage of hormones bound to the sex hormone binding globulin (SHBG)		
Sex hormone	**Free (unbound) %**	**Albumin-bound %**	**SHBG-bound %**
Oestrogen	2	58	40
Testosterone	1	30	69
Dehydroepiandrosterone	4	88	8
Androstenedione	7	85	8
Dihydrotestosterone	1	71	28

TABLE 11.6	Location of the receptors for various hormones
Area of location	**Various hormones**
Cell membrane	Receptors of protein hormones and adrenal medullary hormones (catecholamines)
Nucleus	Receptors of steroid hormones
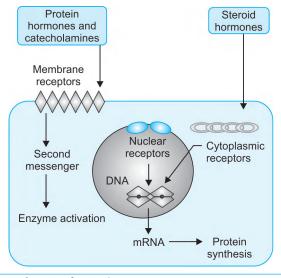
FIG. 11.3: Situation of various hormone receptors
Abbreviation: mRNA, messenger ribonucleic acid

FIG. 11.4: Seven-transmembrane receptor

DNA binding site is capped with a specific protein known as the heat shock protein-90 (HSP-90). Binding of the steroid with the receptor causes a conformational change, resulting in the displacement of HSP-90. Therefore, receptor molecules which are not attached to the steroid cannot bind with the DNA.

Protein hormone receptor: Protein hormones combine with a receptor, which crosses the cell membrane. Some receptors traverse the membrane once (e.g. GH, growth factors and prolactin). Others may traverse the membrane seven times (e.g. oxytocin and gonadotropins). The amino terminus of this seven-transmembrane receptor has the hormone binding domain and the carboxy terminus the G-protein transducer **(Fig. 11.4)**. These receptors couple to the G-proteins (guanine nucleotide-binding proteins, GPCR) following binding of the receptor with the hormone. This in turn activates adenyl cyclase and catalyses the production of a second messenger (cAMP). Excessive built-up of cAMP is prevented by presence of the enzyme, phosphodiesterase. Cyclic AMP activates protein kinases which cause phosphorylation and thereby activation of specific enzymes. The extracellular domain is specific to an individual hormone, even though hormones such as LH and hCG act through the same receptor.

Besides the receptors with seven transmembrane domain and those with a single transmembrane domain, the other groups of cell membrane receptors may include cytokine receptors (cytokines, GH, prolactin) and guanylyl cyclase-linked receptors (natriuretic peptides). Peptide hormones also affect the transcription of certain genes by activating the nuclear transcription factors (e.g. c-jun and c-fos) via kinases and phosphatases.

Hypothalamus

Hypothalamus is the centre of different endocrine, autonomic and homeostatic mechanisms which helps in maintaining the body's homeostasis and the reproductive activities. It produces the releasing hormones which control the activity of the pituitary gland which in turn regulates the activity of the reproductive axis, lactation, growth and secretion of the thyroid and the adrenal glands. The hypothalamus regulates the activity of the anterior pituitary through the release of factors which are carried by veins of the pituitary stalk. It secretes releasing hormones as a series of pulses, and the release is determined by the frequency rather than the amplitude of these pulses. Releasing hormones are polypeptides. The greater the secretion of endogenous releasing hormone, the lesser is the response to exogenous releasing factors. The various releasing hormones produced by the hypothalamus have been enumerated in **Table 11.1**.
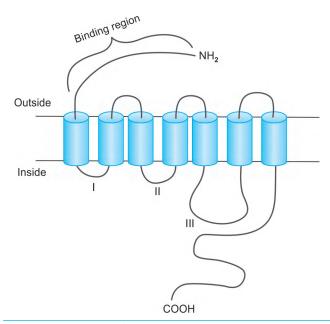
Anatomy

The hypothalamus stretches from the optic chiasma in front to the mammillary bodies behind. It forms part of the mid-brain. It is responsible for temperature regulation through its connection with the limbic system. Thalamus lies superior to the hypothalamus and is separated from it by the hypothalamic sulcus **(Fig. 11.5)**. Third ventricle lies medially while the pituitary stalk lies inferiorly. However, the boundaries of the anterior, posterior and the lateral side of the hypothalamus are not well defined. The median eminence is outside the blood-brain barrier.

Embryology

Thalamus and the hypothalamus develop from the lateral walls of the diencephalon. The cavity of diencephalon forms the third ventricle. The pituitary gland lies in close association with the hypothalamus and is composed of two parts: anterior pituitary or the adenohypophysis
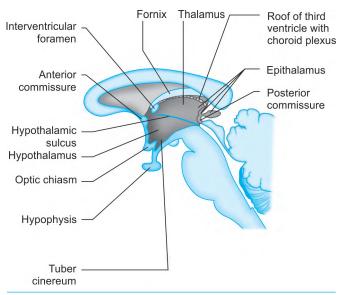
FIG. 11.5: Anatomy of the hypothalamus

FIG. 11.6: Diagrammatic representation of the relationship between pituitary gland and the hypothalamus

and posterior pituitary or the neurohypophysis. The neurohypophysis is in direct communication with the hypothalamus, while the adenohypophysis is connected to the hypothalamus via a rich vascular network called the portal system (Fig. 11.6). The portal system carries all the hypothalamic hormones, which regulate the functioning of anterior pituitary gland. The principal afferent and efferent neural pathways to and from the hypothalamus are unmyelinated.

Pituitary Gland

The pituitary gland lies within a bony depression in the sphenoid bone, the sella turcica (Fig. 11.7). Here it is covered by a layer of dura mater (the diaphragma sellae) through which the pituitary stalk passes. The diaphragma sellae bounds the gland superiorly and the cavernous sinuses laterally. The hypothalamus lies above the pituitary gland and is connected to the gland by the pituitary (or hypophyseal) stalk. The optic chiasma also lies superior to the pituitary and is above and towards the back of the diaphragma sellae. A growing pituitary tumour, rising upwards, presses on the lower anterior part of the chiasma and the medial sides of the optic nerves resulting in the development of bitemporal hemianopia. The pituitary gland lies posterior or superior to the sphenoidal sinus.

The pituitary gland is composed of an anterior lobe (adenohypophysis) and posterior lobe (neurohypophysis). Embryologically, the anterior part of pituitary (adeno-hypophysis) is derived from ectoderm and posterior (neurohypophysis) from neuroderm. Adenophysis is created from Rathke's pouch, an upward evagination of the ectoderm of the pharyngeal roof, while the neurohypophysis develops from a neuroectodermal down growth from the
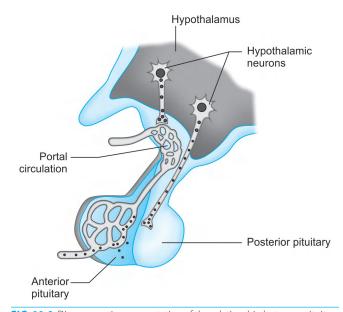
FIG. 11.7: Anatomy of the pituitary gland
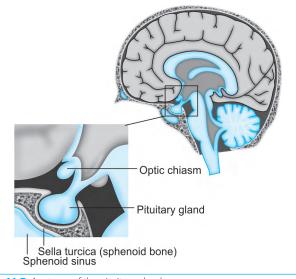
floor of the third ventricle. During pregnancy, the gland increases in weight by 30–50% due to an increase in the secretion of prolactin. The pituitary gland is composed of both the acidophil cells and the basophil cells. Acidophils secrete prolactin and GH. Basophilic cells secrete TSH, LH, FSH, ACTH and its precursor, pro-opiocortin.

It has a rich blood supply. It is supplied by the portal system originating from the hypothalamus (80%), and by a direct arterial supply. The direct blood supply is from the pituitary artery, which arises from the circle of Willis and drains into the inferior petrosal sinuses (IPS). Hence IPS sampling is performed in conditions such as Cushing's disease.

The anterior pituitary has basophil cells which secrete LH.

The anterior lobe secretes peptide hormones which are described in **Table 11.7**.

Glycoproteins, such as TSH, LH and the FSH, which are produced by the anterior pituitary, share a common alpha subunit with unique beta subunits. There is diurnal variation in the secretion of many hormones such as LH, ACTH and GH. The secretion of these hormones is controlled by releasing and inhibitory factors released by the hypothalamus. On the other hand, hormones such as oxytocin and vasopressin are secreted from the posterior lobe of the pituitary.

Hypopituitarism

A complete loss of pituitary function would be devastating and would not be compatible with life as it is required for the maintenance of normal metabolism and fluid balance. Hypopituitarism is typically caused by a non-functional pituitary tumour with a staged loss of hormones with first loss of GH, then LH/FSH, followed by ACTH and finally TSH.

Chromophobe Adenoma of the Pituitary

A chromophobe adenoma refers to the lack of uptake of staining within the pituitary tumour which means that the tumour is non-functioning. These tumours could be associated with hyperprolactinaemia due to stalk compression. Loss of body hair in this case could be related either to an associated hypogonadism or hypopituitarism.

Pineal Gland

Pineal gland lies in the roof of third ventricle. Its role in human beings is presently uncertain. It probably produces melatonin. While melanocyte-stimulating hormone (MSH) is produced in the intermediate lobe of the pituitary; melatonin is produced mainly in the pineal gland. Melatonin is synthesised in the body from serotonin (5-hydroxytryptamine). The necessary enzymes are in the pineal parenchymal cells. Unlike MSH, melatonin has no role in regulation of human skin pigmentation. Melatonin probably has a role in the regulation of circadian rhythm (body's biological clock) and puberty. Melatonin secretion has a pronounced circadian rhythm, low during the day and high by night. Melatonin secreted in relation to prevailing conditions of light/darkness may adjust pituitary hormonal rhythms appropriately. With increasing age, calcification of the pineal gland may occur, making it visible on the skull X-ray.

Growth Hormone

Growth hormone is a protein hormone with no carbohydrate residues. Human GH is composed of 191 amino acids with minor variations in structure across species. There is a structural similarity to prolactin. The genes for human GH are localised in the q22-24 region of chromosome 17 and are closely related to human placental lactogen (hPL) genes.

Growth hormone is an anabolic and lipolytic hormone. It promotes positive nitrogen and phosphorus balance. It stimulates the growth of all the body's organs, muscles and skeleton. It increases cell size and their mitotic activity. GH causes the recruitment of glucose from the liver and is thus anti-insulinic and therefore diabetogenic. It counteracts the effects of insulin on glucose and lipid metabolism, but shares protein anabolic properties with insulin. It increases the rate of protein synthesis by increasing the transport of amino acids into the cells and increasing the rate of transcription and translation. There is a decrease in the utilisation of proteins for energy. It also increases the rate of lipid mobilisation from the lipocytes to the cells. It is, however, associated with a reduced rate of glucose mobilisation. It therefore has a glucose sparing effect. It decreases glucose utilisation and causes reduced glucose transportation into the cells.

TABLE 11.7	Anterior pituitary hormones	
Hormone	**Hypothalamic control**	**Action**
Growth hormone (GH)	Secretion of GH is increased by GHRH (growth hormone-releasing hormone), whereas it is inhibited by somatostatin	It is responsible for soft tissue growth, catabolism and metabolic regulation
Prolactin (PRL)	PRL release is inhibited by Dopamine, whereas it is stimulated by TRH	It is responsible for lactogenesis and suppression of GnRH release
Adrenocorticotrophic hormone (ACTH)	Release of ACTH is under control of CRH (Corticotrophin releasing hormone)	It is responsible for the secretion of adrenal cortical hormone
Luteinising hormone (LH)/Follicle-stimulating hormone (FSH)	Release of LH and FSH is stimulated by GnRH (gonadotropin releasing hormone)	These hormones stimulate the release of sex steroids: oestrogen and progesterone by the ovary and testosterone by the testis. These hormones also promote the maturation of gametes
Thyroid-stimulating hormone (TSH)	Secretion of TSH is stimulated by TRH (thyrotropin-releasing hormone)	This hormone is responsible for stimulating the production of triiodothyronine (T3) and thyroxine (T4)

Secretion of GH during pregnancy is not increased; it may actually be reduced because of the increased level of hPL. Secretion is under hypothalamic control. Levels in the blood are similar in children and in adults. GH helps stimulate growth in children, but not in foetuses. GH level is normal in pygmies, but their small stature is due to the low levels of insulin growth factor.

GH secretion surges occur during sleep because sleep is a time of anabolic activity. It stimulates the liver to secrete somatomedins [insulin-like growth factors (IGFs)] which regulate the growth of bone and cartilage. These peptides mediate general stimulation of growth. Somatomedins also inhibit the pituitary secretion of GH by stimulating the release of somatostatin from the hypothalamus. Secretion of GH increases when the blood glucose level falls. This is the basis of a test for pituitary function. GH secretion is reduced by medroxyprogesterone, rapid eye movement sleep, glucose, free fatty acids and cortisol.

Pituitary gonadotropins, not GH, determine the time of onset of puberty. Therefore impaired GH secretion in children does not cause delayed puberty. GH in conjunction with priming of the anterior pituitary by sex hormones, is however, responsible for the growth spurt during puberty.

In children prior to the fusion of skeletal plates, the main action of GHs is to promote skeletal growth. Therefore, deficiency of GH in children is associated with dwarfism. Pituitary dwarfs are usually normally proportioned. Impaired secretion of the GH is also associated with pale, fine and soft skin. In addition, body hair is normally sparse. Impaired GH secretion has no detectable effect on organ size in adults.

The most effective form of therapy in these cases is replacement with only the human form of GH. It is used for the treatment of both adult and childhood GH deficiency. GH therapy is approved for the treatment of adult hypopituitarism and there is no evidence to suggest that it causes an increased risk in any malignancy. GH therapy produces an elevation of IGF-1 and therapy is monitored through measuring these concentrations. Treatment is, however, contraindicated in cases of active malignancy and proliferative retinopathy.

Dysfunction of the Growth Hormone

Deficiency of GH results in dwarfism in children and symptoms such as hypoglycaemia and micropenis in the neonates. In adults, deficiency of GH can result in symptoms such as weight loss, reduced body mass index, lethargy, poor bone density, impaired physical performance in adults, psychological problems, etc. Excessive secretion of GH, on the other hand, can result in gigantism in children and acromegaly in adults because in these cases closure of the epiphyseal plates has already occurred. Acromegaly is characterised by excessive growth of soft tissues (e.g. tongue, liver and heart) resulting in symptoms such as macroglossia, abnormal enlargement of an organ, splenomegaly, etc. There also may be an abnormal growth of the bones of hand, feet and jaw resulting in symptoms such as large hands and feet, prognathism, etc. Other symptoms may include intestinal polyposis, splenomegaly, hypercalciuria, palpable peripheral nerves, proximal myopathy, etc. It can also result in the development of diabetes mellitus and hypertension. Acromegaly is diagnosed on the basis of non-suppression of GH concentrations with the oral glucose tolerance test. GH secretion is suppressed by somatostatin analogues, for example, octreotide, which are used therefore in acromegaly.

Prolactin

Prolactin is a protein hormone produced by the acidophil cells of the anterior lobe of the pituitary gland. It mainly acts on the breast tissue producing milk. It stimulates growth of the milk ducts and glands during pregnancy and milk production once the baby is delivered. High levels of oestrogen, and, maybe, progesterone, during pregnancy prevent full-blown lactation before delivery.

PRL consists of 199 amino acids and is similar in structure to GH. It is also structurally related to hPL. Eighty-five percent of the amino acids in hPL are identical to that of human pituitary GH and human pituitary prolactin. Furthermore, hPL shares biologic properties with both GH and prolactin. Thus, it has primarily lactogenic activity but also exhibits some GH-like activity; therefore, it is also referred to as human chorionic growth hormone or human chorionic somatomammotropin (hCS).

Prolactin is a unique hormone in the sense that its production is not dependent on a releasing, stimulating or trophic hormone, as is the case for most the hormones of anterior pituitary gland. This makes PRL unique and distinct from the other anterior pituitary hormones. The molecular weight of PRL is 24,000 daltons and it has a half-life of 6 hours. Prolactin is the only hormone which is inhibited rather than stimulated by the hypothalamic factors (dopamine). Its release is pulsatile. There is circadian variation, with high levels in the first part of the night. Similar to the GH, its concentrations rise during sleep.

Prolactin helps in maintaining successful lactation. Other hormones involved in galactopoiesis include glucocorticoids and thyroid hormones. Normal prolactin levels are less than 500 mu/L. Levels less than 1,000 mu/L are usually of no great significance. Levels greater than 1,000 mu/L should be repeated and investigated if still elevated. High prolactin levels may cause anovulation and galactorrhoea. Increased prolactin levels could be due to any of the causes listed in **Table 11.8**.

Pituitary adenomas can expand in pregnancy and press on the optic chiasma.

Release of prolactin is stimulated by thyrotropin-releasing hormone (TRH). Other stimulants of prolactin

TABLE 11.8	Causes for increased level of prolactin

- *Dopamine antagonists*: Drugs like phenothiazines, methyldopa, chlorpromazine, haloperidol, metoclopramide, sulpiride, etc.
- Pituitary adenomas (microadenomas <10 mm; macroadenomas >10 mm
- Brain/pituitary tumours that destroy or compress the pituitary stalk
- Hypothyroidism
- Polycystic ovary syndrome
- Acromegaly
- Renal failure
- Fits

include insulin induced hypoglycaemia, dopamine antagonists (metoclopramide, domperidone, etc.), after fits and in renal failure. Secretion of prolactin is increased by sexual intercourse. Craniopharyngioma (as well as any pituitary tumour) may produce pituitary stalk compression and hence cause hyperprolactinaemia.

Hypothyroidism by causing increased secretion of TRH produces increased prolactin (PRL) concentrations.

A glycosylated form of prolactin is secreted by the endometrium in the luteal phase of the cycle. Concentrations normally rise dramatically during pregnancy. During pregnancy, it is secreted by the decidua in pregnancy. Prolactin is secreted in the middle trimester of pregnancy, and increases progressively towards term. Prolactin concentration is increased (surges) through mechanical effects such as suckling or breast stimulation.

The release of prolactin is inhibited by the hypothalamic secretion of dopamine. Thus dopamine agonists are used for the treatment of prolactinomas. Dopamine antagonists such as metoclopramide and haloperidol together with the new antipsychotics such as olanzapine produce hyperprolactinaemia. Other drugs, producing hyperprolactinaemia include monoamine oxidase inhibitors (MAOIs), methyldopa, reserpine, phenothiazines, butyrophenones and morphine. Oral contraceptives do not cause hyperprolactinaemia because they do not affect dopamine release.

Hyperprolactinaemia due to Prolactinoma

A chromophobe adenoma (non-functioning) may cause hyperprolactinaemia through stalk compression and hypogonadism due to associated hypopituitarism. A prolactinoma, on the other hand, is a prolactin producing tumour of the pituitary gland. A prolactinoma may present with the following clinical features:
- Oligomenorrhoea or amenorrhoea
- Bitemporal hemianopia due to chiasmal compression
- Reduced bone marrow density associated with long standing hypo-oestrogenism
- Hypopituitarism
- Galactorrhoea, spontaneous or expressible (60% of cases)
- Decreased libido in both sexes
- Decreased potency in men
- Subfertility

- Symptoms or signs of oestrogen or androgen deficiency—in the long-term osteoporosis may result, especially in women
- Delayed or arrested puberty in the peripubertal patient
- Additionally, headaches and/or visual field defects may be present if there is a large pituitary tumour. This occurs more commonly amongst men. However, this is rarely seen because most commonly the tumour is a microadenoma.

Hirsutism is not a feature of hyperprolactinaemia because hyperprolactinaemia causes hypogonadism and so does not produce hirsutism per se.

Prolactin levels above 1,000 mu/L are often due to adenomas. Larger tumours are usually accompanied by higher blood levels. Most tumours are less than 10 mm, known as microprolactinoma. They are rarely more than 10 mm, also known as macroprolactinoma. MRI is more sensitive to small microadenomas than CT. Generally the normal pituitary gland enlarges during pregnancy and a small but clinically non-significant enlargement is seen in microprolactinomas. Rarely, tumours enlarge during pregnancy to produce headaches and visual defects.

Microadenomas are associated with prolactin levels of 1,500–4,000 mu/L. Macroadenomas are associated with levels more than 5,000 mu/L and often more than 8,000 mu/L. Galactorrhoea occurs in about 30% of women with hyperprolactinaemia. Pituitary adenomas, which are usually prolactinomas, tend to enlarge in pregnancy. Even microadenomas (<1 cm diameter) can enlarge enough to cause visual loss.

Hyperprolactinaemia is associated with a positive progestogen challenge test. Prolactin decreases gonadotropin-releasing hormone (GnRH) pulsatility at the hypothalamic level and, to a lesser extent, blocks the action of LH on the ovary or testis, producing hypogonadism. Hyperprolactinaemia per se does not have an effect on other dynamic pituitary function tests such as cortisol and GH response to Insulin tolerance test.

Bromocriptine is a dopamine agonist commonly used for the treatment of hyperprolactinaemia. Surgery is nowadays not required even for the treatment of large pituitary tumours because these tumours can be effectively treated with dopamine agonist therapy, e.g. bromocriptine and cabergoline.

Hormones of Posterior Pituitary

The posterior pituitary is a neurosecretory gland, which releases two hormones, namely oxytocin and vasopressin or ADH. Both these hormones which are synthesised within the hypothalamus in the hypothalamic nuclei and released from the posterior pituitary are nonapeptides (a peptide molecule composed of nine amino acids). Release of vasopressin/ADH is controlled by the plasma osmolality. Secretion of ADH and oxytocin is inhibited by alcohol.

Since alcohol inhibits the secretion of oxytocin, it has been used in the past as a tocolytic agent for reducing uterine contractions during preterm labour.

Oxytocin

Oxytocin is synthesised in the paraventricular nucleus of the hypothalamus, which is then transported in secretory granules along axons to the posterior pituitary gland, from where it is released. It is involved in the control of smooth muscle contraction in the uterus and also for causing milk release from the lactating breast (the "milk ejection reflex"). The primary role of oxytocin is to eject milk from the lactating mammary gland in response to suckling. It achieves this by causing contraction of the myoepithelial cells that surround the alveoli of the mammary gland.

Foetal oxytocin plays a role in stimulating the uterus, especially towards the end of pregnancy. It is also released in the male during orgasm and by the foetus during labour. It is inactivated by oxytocinase, which is produced by the placenta. By lowering the threshold for depolarisation of uterine smooth muscle, oxytocin exerts a contracting effect on the gravid uterus. The sensitivity of the uterus to oxytocin increases as the pregnancy progresses. Oxytocin has approximately 0.5–1% of the antidiuretic activity of ADH when administered in high doses, introducing the possibility of water intoxication.

Clinical Uses

The principal clinical uses of synthetic oxytocin are to induce labour at term and to produce sustained uterine contractions following delivery, which is required for post-partum or post-termination haemostasis.

The uterus in early pregnancy is not very sensitive to oxytocin, but the latter may be used as a synergistic agent (with prostaglandin) in cases of therapeutic abortion.

Vasopressin

Vasopressin is a nonapeptide hormone, which is synthesised in the cell bodies of the supraoptic and paraventricular nuclei of the hypothalamus. It is then transported down their axons to the posterior lobe of the pituitary gland from where it is secreted. Plasma half-life of vasopressin is approximately 18 minutes. Similar to oxytocin, its secretion is inhibited by alcohol. Secretion of vasopressin is increased by angiotensin II.

Vasopressin increases the permeability of the distal tubules and collecting ducts of the kidney to water. Water reabsorption is therefore increased, and urine volume decreased. Vasopressin is the primary regulator of body water. It acts on the collecting ducts to increase total body water. This has the effect of reducing the plasma concentration (osmolality) and increasing blood volume. Vasopressin may increase glomerular filtration rate by selectively constricting the glomerular efferent arteriole.

Diabetes insipidus refers to a condition in which the individuals suffer from the deficiency of ADH. When a patient with diabetes insipidus is treated successfully with ADH, the urinary flow rate should fall by about 80%. Urinary output should be reduced to around 5 mL/minute. Urinary osmolality should rise to about 600–900 mosmol/L, 2–3 times normal plasma osmolality (300 mosmol/L). ADH does not interfere with salt regulation. Nevertheless, blood pressure should stabilise within the normal range due to greater stability of the body fluids.

Disorders of Thyroid Gland

Thyroid Gland

The thyroid gland is a small yellowish-brown gland in the neck at the level of C3 and C4. It is composed of two lobes connected to each other by an isthmus, which passes anterior to the trachea (Fig. 11.8). Thyroid gland develops as a thickening of the floor of the pharynx. The normal weight of the gland is approximately 30 g. The thyroid gland is composed of follicular cells (lining the follicles) and the parafollicular or the C cells (which produce the hormone calcitonin). The parathyroids lie posterior to the thyroid gland and the recurrent laryngeal nerves lie posteromedially (Fig. 11.9). Production of thyroid hormones by the thyroid gland is stimulated by the anterior pituitary hormone, TSH and secretion begins from approximately the 12th week of gestation. TSH acts on the thyroid gland to produce the thyroid hormones, thyroxine (T4) and tri-iodothyronine (T3) through iodination of tyrosine. The thyroid follicles synthesise T3 and T4, which is then stored within the colloid at the centre of the thyroid follicles. They are stored attached to thyroglobulin (a glycoprotein with the molecular weight 660,000 daltons) in the colloid and
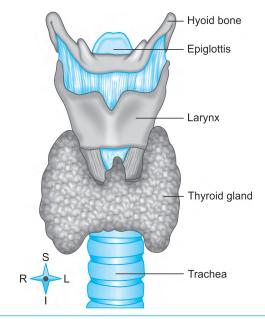
FIG. 11.8: Anatomical location of the thyroid gland
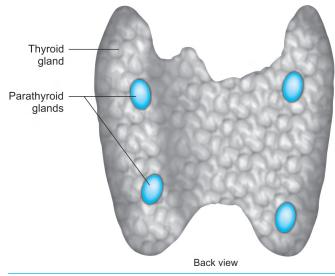
FIG. 11.9: Thyroid gland showing location of the parathyroids

FIG. 11.10: Synthesis of thyroid hormones
Abbreviations: MIT, mono-iodo-tyrosine; DIT, di-iodo-tyrosine; T_3, triiodothyronine; T_4, thyroxine

released through proteolysis into the blood where in, they are mainly transported bound to thyroid binding globulin (TBG). Seventy-five percent of T4 is bound to TBG.
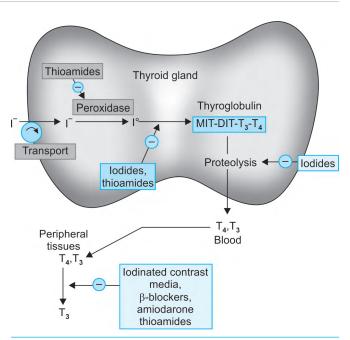
Thyroid Hormones

As previously mentioned, thyroid hormones include thyroxine (T4) and triiodothyronine (T3), which are glycoprotein in nature. Thyroxine is produced within the thyroid gland from the oxidation of four iodide molecules with two tyrosine residues under the influence of peroxidase and iodinase **(Fig. 11.10)**. Out of the two hormones, thyroxine and thyronine, thyronine is the most active hormone and is mainly produced by de-iodination of thyroxine. In the body, more T4 than T3 is produced, although T3 is more active at the thyroid hormone receptor. T4 is converted in some peripheral tissues (liver, kidney and muscle) to the more active T3 by 5'-monodeiodination; an alternative 3'-monodeiodination yields reverse T3 (rT3), which is largely inactive. T3 and T4 circulate in plasma almost entirely bound mainly to TBG, also to thyroid-binding prealbumin, and albumin. Only free hormone is available for tissue action, which acts by binding to the specific nuclear receptors within the cell.

Dyshormonogenesis

Dyshormonogenesis results from a deficiency or absence of one or more of the enzymes involved in thyroid hormone synthesis or secretion.

The most common enzyme abnormality is absent or insufficient thyroid peroxidase activity which results in failure of oxidation (organification) of iodide to iodine. With this type of defect, iodine will be trapped, but not organified. Deficiency of peroxidase will result in the deficiency of T3 and T4. Decreased levels of circulating thyroid hormone in such patients may cause an elevation of TSH levels. This may result in an enlarged thyroid gland in these patients.

There may be a high uptake of I-123 (at 4 hours) and Tc-pertechnetate. Thyroxine (T4) and free thyroxine index (FTI) are usually decreased. The perchlorate washout test will be positive in these patients.

Deficient peroxidase activity associated with a familial goitre and deafness or hearing loss is referred to as Pendred's syndrome

Changes in Thyroid Gland during Pregnancy

The thyroid gland like most other endocrine organs moderately enlarges during pregnancy. In the foetus, thyroxine (T4) can be detected in the serum before 18 weeks' gestation. During pregnancy, there is a marked increase in secreted levels of TBG. TBG levels are elevated by intake of oral oestrogen and increase in oestrogen levels related to pregnancy. TBG levels are depressed by glucocorticoids, androgens and the anti-oestrogen, e.g. danazol. The elevated levels of binding proteins result in an increase in total thyroxine and thyronine but free hormone concentrations remain unchanged. Although the total amount of T4 is increased in pregnancy, the mother remains euthyroid because the amount of free T4 remains the same. Free T4 (fT4) concentrations are usually low to normal in normal pregnancy due to dilutional effects. Therefore, it is best to regard the TSH concentration in order to assess the thyroid status of a pregnant woman. However, early pregnancy may be associated with slight rise in T4 associated with elevated human chorionic gonadotropin (hCG) levels which declines as pregnancy progresses. Daily iodine requirement is 120 micrograms for females and about 150 micrograms for males. During pregnancy iodine requirements increase by about 50%, i.e. 175 micrograms daily and about 200 micrograms while breast feeding.

Normal Functioning of Thyroid Glands

Thyroid hormones are required for maintaining the body's metabolic rate, normal growth and development of the brain and increased sympathetic neural activity. Thyroid hormones are essential for normal brain development and musculoskeletal growth of the foetus. In the absence of thyroid hormones, impaired development of the brain and musculoskeletal system may result in cretinism. Thyroid hormones are also responsible for maintaining the normal hypoxic and hypercapnic drives to the respiratory centre. At the metabolic level, T4 and T3 stimulate lipolysis, glycolysis, gluconeogenesis, absorption of glucose and improved metabolism of cortisol and insulin.

Hyperthyroidism

Hyperthyroidism is 8 times commoner than hypothyroidism. Thyroid hormones, when secreted in excess, may be associated with the following:
❑ Peripheral vasodilatation due to increased metabolism
❑ Increased frequency of defaecation.
❑ Increased energy expenditure required for a given workload. Thyroid hormones uncouple oxidation from phosphorylation so that more energy appears as heat
❑ Reduced duration of tendon reflexes.
❑ Increased heart rate when cardiac adrenergic and cholinergic receptors are blocked. This suggests a direct action on cells in the sinoatrial node.
❑ Negative nitrogen balance due to muscle wasting.
❑ Increased urinary excretion of calcium due to liberation of calcium from bone.
❑ Clinical picture consistent with excessive beta adrenoceptor stimulation. Beta adrenoceptor blocking drugs relieve such features, e.g. tachycardia.
❑ There is diminished heat tolerance. Heat intolerance is due to the increased heat production in these cases.
❑ Prominent eyeballs are characteristic of the exophthalmos due to hyperthyroidism.
❑ There may be increased tremors.
❑ Pretibial myxoedema is associated with Graves' disease and therefore usually with thyrotoxicosis, though Graves' disease may sometimes ultimately result in hypothyroidism.
❑ Most patients with sub-acute thyroiditis end up euthyroid, though there may be transient thyrotoxicosis at times.

Secretion of TSH is increased in the following situations:
❑ After partial removal of the thyroid gland due to a reduction in pituitary inhibition by circulating thyroxine.
❑ In infants born without a thyroid gland due to absence of the normal pituitary inhibition by circulating thyroxine.
❑ When the diet is deficient in iodine. Due to inadequate manufacture of thyroxine, pituitary inhibition is reduced.

In cases of starvation, TSH levels fall in order to conserve energy.

Hypothyroidism

Possible features of hypothyroidism include the following:
❑ A subnormal body core temperature: Due to the lowered metabolic rate.
❑ A tendency to fall asleep frequently: Due to the slowing of mental processes.
❑ Hair loss.
❑ Reduced sweating resulting in dry hands and feet.
❑ Increased time for tendon reflexes, e.g. hung up ankle jerk.
❑ Hypothyroidism may be associated with pericardial effusion and/or cerebellar ataxia.
❑ It is associated with an elevated CSF protein levels.
❑ Goitre is a frequent feature in both Hashimoto's thyroiditis and also iodine deficiency.
❑ Other presenting features include ataxia and neuronopathies particularly compression neuronopathies.
❑ Cognitive impairment, macrocytic anaemia and pleural effusions also feature.
❑ It may present with menstrual abnormalities, e.g. menorrhagia. Deficiency of the thyroid hormones may also lead to anovulation associated with increased levels of LH.
❑ Myxoedematous change may cause carpal tunnel syndrome in hypothyroidism.
❑ It may be associated with a macrocytic or microcytic anaemia.
❑ It may be caused by Hashimoto's thyroiditis.

Radioactive Iodine

Radioactive iodine (RAI) is an effective treatment for hyperthyroidism due to Graves' or toxic nodules. The most notable side effect of radioactive iodine therapy is hypothyroidism in 80% of those treated. It is also associated with an exacerbation of Graves' ophthalmopathy but this risk can be attenuated with the co-administration of steroids. This form of therapy is very safe but is absolutely contraindicated in pregnant females. Those that are breast feeding need to stop for approximately 8 weeks.

Understanding of Pancreas

The Pancreas

Pancreas is a retroperitoneal organ, having both endocrine and exocrine functions. It produces many hormones which are required for the normal metabolism of glucose, fat and proteins. The glandular acini are also responsible for the secretion of bicarbonate and enzymes, which may help in the process of digestion. Pancreas are made up of two types

of tissue, glandular acini and islets of Langerhans. The islets of Langerhans are composed up of three main cell types: α (alpha) cells; β (beta) cells; and the δ (delta) cells. Alpha cells produce glucagon; β cells produce insulin; and δ cells produce somatostatin.

Glucagon

Pancreatic glucagon is a polypeptide produced by the alpha cells of pancreas. This hormone is produced in response to hypoglycaemia and the fasting state. Output is inversely proportional to the blood glucose level. It normally prevents a serious fall in blood glucose levels. It has a short half-life of about 5–10 minutes, which allows glucagon levels in the blood to adjust rapidly to changes in blood glucose levels. It is a catabolic hormone. It is one of the main hormones signalling the hepatic breakdown of glycogen to glucose. It also mobilises fatty acids. It therefore helps antagonize the action of insulin and maintain a supply of energy during fasting. The increase in glucose causes a counteractive increase in insulin production.

Insulin

Insulin is a polypeptide hormone having a complex structure. The chemical structure of insulin contains two peptide chains. Minor differences occur in the insulin available in different mammalian species. However, these differences do not affect insulin action. It is ineffective when taken by the mouth because its peptide structure is broken down by digestive proteases in the gut. It is metabolised in the liver and undergoes renal excretion. Its half-life is roughly 4 minutes. Insulin acts by stimulating RNA synthesis in the nucleus. Insulin was first synthesised in the laboratory in 1964 by Katsoyannis. It can also be synthesised by bacteria using recombinant DNA technique. Insulin (and C-peptide which is inactive) is secreted by the beta cells within the islets of Langerhan as the active molecule following cleavage from preproinsulin, then proinsulin. The production of C peptide is therefore used as a marker for endogenous insulin synthesis. Insulin is secreted in two patterns: first is a basal/constitutive pattern, which is associated with continuous secretion of insulin regardless of the blood glucose levels. Secondly, there is a release of insulin which had been stored during the postprandial (fed) state. This response is related to high serum blood glucose and presence of carbohydrates in the small intestine.

Action of Insulin

Insulin promotes increased glucose uptake by the fat and muscles. It, however, does not increase its uptake into brain or liver either. Insulin is anabolic in nature and it predominantly inhibits glycogenesis in the liver though it also stimulates glycogenolysis. It also stimulates glucose

uptake in adipose tissues, cellular uptake of amino acids (protein synthesis) and synthesis of fatty acids in the liver. At the same time it inhibits gluconeogenesis, glycogenolysis, lipolysis and proteolysis. Insulin inhibits gluconeogenesis and promotes glycogen synthesis. Anti-insulinic hormones which increase the plasma blood glucose levels include GH, adrenaline, glucagon and corticosteroids (not mineralo-corticoids like aldosterone).

Insulin acts through a disulphide-bonded heterotetrameric cell surface receptor comprising of an extracellular alpha subunit coupled via disulphide bonds to a transmembrane and intracellular beta subunit. Signalling through the insulin receptor occurs through an intracellular tyrosine kinase domain resulting in the phosphorylation of receptor. Insulin interacts with cell surface receptors. Binding of insulin to its receptor results in receptor autophosphorylation on tyrosine residues and the tyrosine phosphorylation of insulin receptor substrates (IRS-1, IRS-2 and IRS-3) by the insulin receptor tyrosine kinase **(Fig. 11.11)**. Phosphorylated IRS-1 causes incorporation of the glucose transporter, GLUT-4 into the cellular membranes. This results in an increased uptake of glucose from the serum. Some tissues do not require insulin for efficient uptake of glucose. Important examples of such tissues are brain and the liver because these cells do not use GLUT4 (a hexose transporter for facilitated diffusion of glucose) for importing glucose, but rather, another transporter which is not insulin-dependent. Insulin is used for the treatment of hyperkalaemia because it "drives" K^+ back into cells. Insulin secretion is influenced by neurotransmitters interacting with islet cell receptors, particularly those that bind norepinephrine. Insulin secretion is stimulated by glucose, amino acids (arginine) and triglycerides. Pharmacologically its secretion is stimulated by sulphonylureas such as glibenclamide. Bendroflumethiazide may produce deterioration in insulin sensitivity and hence increase insulin secretion. Propranolol may inhibit insulin secretion. The gastrointestinal hormone, gastrin, which stimulates gastric acid secretion and proliferation of gastric mucosa, also stimulates insulin release.

Somatostatin

Somatostatin's effects are all inhibitory including that of insulin release.

Diabetes Mellitus

Diabetes mellitus is the most common metabolic disorder occurring due to the deficiency of insulin. It is characterised by hyperglycaemia, glycosuria, hyperlipidaemia, negative nitrogen balance and sometimes ketonaemia. Two major types of diabetes mellitus are type I insulin-dependent diabetes mellitus (IDDM)/juvenile onset diabetes mellitus
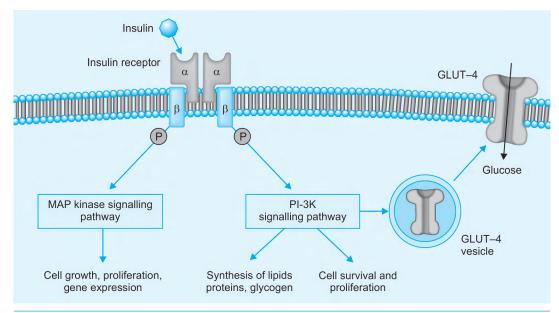
FIG. 11.11: Mechanism of action of insulin
Abbreviations: GLUT-4, glucose transporter type 4; PI-3K, phosphoinositide 3-kinase; MAP kinase, mitogen activated protein kinase

TABLE 11.9	Difference between type I and type II diabetes mellitus	
Characteristic feature	**Type I diabetes mellitus**	**Type II diabetes mellitus**
Pathological mechanism involved	There is a destruction of β cells, resulting in reduced levels of insulin in the body	Insulin in circulation is low, normal or even high. There is insulin resistance
Destruction of pancreatic β cells	There is β cell destruction in pancreatic islets	No loss or moderate reduction of in β cell mass
Presence of anti-β-cell antibody	Majority of cases of type I diabetes are autoimmune (type 1A)—antibodies that destroy β cells are detectable in blood, but some are idiopathic (type 1B)—no β cell antibody is found.	No anti-β-cell antibody is demonstrable. The histological appearance of the islets is of amyloid infiltration.
Genetic predisposition	Association with HLA DR3 and 4	Minimal association
Concordance rate amongst identical twins	Lower in type I diabetics (40%)	Higher rate in type II diabetics (80%)
Obesity as a risk factor	No association with obesity	More common amongst obese individuals
Effect of exercise	Exercise usually has no effect	Exercise reduces insulin requirements
Presentation	Acute	Presentation is insidious with the patient having the condition for many years before it is diagnosed
Predisposing factor	Minimal association with Western diet	Association with Western diet

and type II non-insulin-dependent diabetes mellitus (NIDDM)/maturity onset diabetes mellitus. The differences between the two types are summarised in **Table 11.9**.

Diagnosis

Glycosylated Haemoglobin

Glycosylated haemoglobin is haemoglobin to which glucose is bound. Glycosylated haemoglobin is formed through the non-enzymatic binding of a hexose sugar to the N-terminal amino acid of the beta-chain of haemoglobin. Glycosylated haemoglobin is found in the normal population but the percentage is lower than in diabetic patients. The normal level for glycosylated haemoglobin is less than 7%. Levels of glycosylated haemoglobin help in identifying plasma glucose concentrations over prolonged periods of time (approximately 12 weeks). Glycosylated serum proteins such as fructosamine are more accurate than glycosylated haemoglobin for the retrospective estimation of blood sugar levels. The risk of renal damage such as microalbuminuria increases when HbA1c exceeds 8%.

Complications of Diabetes

Diabetic Ketoacidosis

In diabetic ketoacidosis (DKA) the lack of insulin leads to a breakdown of fat and the production of ketone bodies. The ketone bodies produce an acidosis which leads to deep, rapid breathing (Kussmaul respiration). Both the ketones and glucose produce an osmotic diuresis causing severe

TABLE 11.10	Classification of diabetes ketoacidosis (DKA) based on the stages of severity			
Parameter	Normal	Mild DKA	Moderate DKA	Severe DKA
pH	7.35–7.45	7.25–7.30	7.0–7.25	<7.0
Serum bicarbonate level	>20 mmol/L	15–18 mmol/L	10–15 mmol/L	<10 mmol/L
State of consciousness	Alert	Alert	Mild drowsiness	Stupor or coma

dehydration. The ketones smell of acetone or nail polish. DKA is more common in type 1 diabetes. This condition infrequently occurs in patients with type II diabetes mellitus. The most common precipitating cause in these cases is infection; other precipitating causes include trauma, stroke, pancreatitis, stressful conditions and inadequate doses of insulin. Usually patients are unwell with infections and anorexia. Abdominal pain at onset is an unusual but recognised feature of diabetes ketoacidosis particularly in children. However it does not support a diagnosis of DKA. Serum bicarbonate of 10 mmol/L suggests metabolic acidosis. Typical cases of DKA are associated with hyperglycaemia (blood glucose levels greater than 30 mmol/L), presence of ketone bodies in the blood or urine analysis and acidosis. Normoglycaemic DKA can also occur in some cases and a glucose of less than 30 mmol/L does not rule out the diagnosis. DKA has been classified into three stages of severity by the American Diabetes Association as described in **Table 11.10**. DKA is typically associated with characteristics tabulated in **Table 11.11**. Usually ketones are found in the urine but occasionally are absent for reasons such as impaired clearance during DKA. Due to excessive renal losses, total body potassium is typically low even if plasma potassium is high.

However, it is well recognised that DKA may still be associated with normal or not particularly high glucose (for e.g. 15 mmol/L).

Hyperosmolar Non-ketotic Diabetic Coma

Hyperosmolar non-ketotic diabetic coma is a complication of diabetes, specifically type II diabetes, which is associated with increased blood glucose levels, increased plasma osmolarity and increased risk of coma and death. This condition is usually not associated with increased levels of ketone bodies, a characteristic feature of diabetic ketoacidosis (associated with type I diabetes) is commonest in the elderly. In these cases, there is absent to low ketonaemia on dipstick. Plasma osmolality is typically greater than 320 mOsm/kg. Profound dehydration up to an average of 9 L can occur in this condition. Serum pH is greater than 7.3. Bicarbonate levels are greater than 15 mEq/L. Hyperosmolar non-ketotic diabetic coma is typically associated with a blood sugar level greater than 30 mmol/L. It is not associated with acidosis. Therefore, it does not cause hyperventilation. This condition can predispose to thrombosis. Therefore, full anticoagulation forms part of the treatment.

TABLE 11.11	Characteristic features of diabetes ketoacidosis

- Increased plasma glucose levels
- Leucocytosis
- Reduced pH
- Reduced levels of bicarbonate
- Increased carbon dioxide partial pressure (pCO_2)
- Reduced oxygen partial pressure (pO_2)

Understanding of Sex Hormones

Gonadotropin-Releasing Hormone

Gonadotropin-releasing hormone is a decapeptide which passes via the portal veins from the hypothalamus to the anterior pituitary gland and controls the pulsatile release of FSH and LH. GnRH is derived by cleavage from a larger precursor called prepro-GnRH. It is a small peptide comprising 10 amino acids. It has a short half-life of 2–6 hours. The pulses of GnRH are directly under the influence of a dual catecholaminergic system. Norepinephrine (noradrenaline) is facilitatory, and dopamine inhibitory. It has both autocrine and paracrine functions throughout the body. Production of GnRH has been identified at other sites such as in the placenta, ovary and other regions of the brain. The cells that produce GnRH originate from the olfactory area of the brain. This is the reason why in Kallmann's syndrome there is an association between an absence of GnRH and a defect in sense of smell.

Secretion of GnRH secretion is pulsatile resulting in the pulsatile secretion of luteinising hormone/follicle-stimulating hormone (LH/FSH). GnRH begins to be secreted just before puberty in pulses and continues in this fashion for the rest of life. During childhood, minimal amount of GnRH is produced by the hypothalamus. However, by the time of late childhood, gradually the pulses of GnRH begin to be produced, initially only at night. These infrequent pulses of low amplitude and low frequency increase during the subsequent years and eventually occur both during the daytime hours as well as during the night. By the time of puberty, as the pulse frequency and amplitude of GnRH increases, the ovarian response is observed to occur. Sustained continuous administration of exogenous GnRH analogues has an initial flare-up effect (increased production of LH and FSH) followed by down-regulation associated with decreased production of LH and

FSH. GnRH agonists are administered by subcutaneous or intramuscular injections, as a nasal spray, as sustained release implants or as injections. Oral administration of GnRH agonists is not effective.

Testicular Hormones

The testis is responsible for secreting the following hormones:
❑ Testosterone
❑ Androstenedione
❑ Oestradiol
❑ Inhibin
❑ Small amount of progesterone.

In males, FSH causes generation of spermatozoa, where LH causes the secretion of testosterone from the testes. Testosterone is released by the Leydig cells of the testes in response to LH stimulation from the anterior pituitary gland. Testosterone is anabolic hormone which leads to skeletal muscle hypertrophy.

Increasing levels of testosterone, like oestradiol, produces a negative feedback effect at the hypothalamus/pituitary to inhibit the secretion of GnRH switching off the secretion of both LH and FSH. Similarly, the falling levels of testosterone stimulate the release of LH-releasing hormone (LHRH) in the hypothalamus leading to increased levels of LH, and subsequently, of testosterone.

Testosterone is metabolised to 17-ketosteroids in the liver, and after transformation to sulphate or glucuronide is excreted in the urine. Most testosterone circulates in the bloodstream bound either to SHBG or to albumin; only 2–3% is unbound and is functionally active.

In the female, testosterone is synthesised in small amounts, probably in the adrenals, but a weak androgen, androstenedione is formed as a step in the metabolism of progesterone.

Fructose and prostaglandins that nourish the spermatozoa are secreted by the seminal vesicles.

Androgens in Women

In women, androgens are synthesised both in the ovaries and the adrenal glands. The three principal androgens in normal women are DHT, testosterone and androstenedione. Of these three hormones, androstenedione is the least potent. However, this is converted to dihydrotestosterone in the follicle cells. In women, the secretion of androstenedione, a weak androgen, is relatively in equal amounts from the adrenals and ovaries, i.e. 50% is principally produced from the adrenal cortex and 50% from the stromal cells of ovary. Concentration of androstenedione and other androgens is typically raised in polycystic ovary syndrome (PCOS).

Nearly 25% of the body's circulating testosterone is formed directly from the ovaries. Of the remainder 75%, approximately 25% is directly derived from the adrenal glands and 50% through the peripheral conversion of androstenedione and to a much lesser extent of dehydroepiandrostenedione. In female, the androgens are responsible for the maintenance of pubic and the axillary hair and also the libido.

Hirsutism

Hirsutism can be defined as the presence of coarse, dark, terminal hair in a male pattern in a woman. For details related to hirsutism, kindly refer to Chapter 14. Testosterone is transported by SHBG. Decreased concentration of SHGB results in a rise in the levels of free testosterone. This may be a cause of hirsutism. Only free testosterone levels are available for biological activity. The hair follicles have androgen receptors to collect it from the blood. Inside the follicle, testosterone is converted to DHT with help of the enzyme 5-alpha-reductase. Dihydrotestosterone is much more powerful than testosterone and the active agent in the follicle.

Female Reproductive System

Hormones of Female Reproductive System

Follicle-stimulating Hormone

Follicle-stimulating hormone is produced by the chromophil cells of the anterior lobe of the pituitary gland. FSH, similar to TSH, hCG and LH are glycoproteins composed of two subunits, alpha and beta. The alpha chain is similar in the mentioned hormones, but the beta chain is unique in all hormones, conferring most of the specific functional properties of each hormone. In FSH, the alpha chain consists of 92 amino acids and two carbohydrate chains; the beta chain contains 111 amino acids and two carbohydrate chains. The molecular weight of FSH is 28,000 daltons. The receptors for FSH are similar to the LH receptors. These are the serpentine receptors coupled to adenyl cyclase.

Follicle-stimulating hormone is responsible for early development of the uterine endometrium and the ovarian follicle, whereas LH is responsible for final their maturity. FSH also stimulates the production of oestrogen from ovarian follicles. In the male, FSH is concerned with maintenance and growth of the germinal epithelium of the seminiferous tubules and with sperm production.

Menopause is associated with an increased secretion of FSH and LH from the pituitary in an attempt to stimulate ovulation. Elevated FSH is also seen with conditions such as primary ovarian failure, Turner's syndrome and Klinefelter's syndrome (testicular failure). Levels of LH and FSH may be normal in childhood and prepubertal girls, but are significantly elevated by 10-11 years in Turner syndrome and in women with gonadal dysgenesis. In the case of

women who are taking the combined oral contraceptive pill, the secretion of FSH is reduced due to inhibition of the pituitary by increased oestrogen concentrations.

Luteinising Hormone

Luteinising hormone, which is secreted from anterior pituitary, is responsible for ovulation and the initial formation of the corpus luteum. LH per se is not required for the maintenance of the corpus luteum, but beta-hCG produced from the fertilised ovum helps maintain this.

In the male, LH maintains the interstitial cells of the testes and stimulates them to secrete testosterone.

The highest pulse frequency of LH occurs during the late luteal phase, whereas the highest pulse amplitude occurs during the early luteal phase. The half-life of FSH is 170 minutes; that of LH is about 20 minutes. The circulating half-life of LH is mainly proportional to the amount of sialic acid present. The higher content of sialic acid in LH compared with FSH accounts for the more rapid clearance of LH from the circulation.

Oestrogens

Oestrogens are a mixture of steroids, the most important being 17 beta-oestradiol. The two other main oestrogens of importance are oestrone and oestriol, but are produced in smaller concentrations. The most active form of oestrogen, known as oestradiol, is normally metabolised to oestrone, and then to oestriol. Low oestriol levels suggest that the liver is not performing this metabolising function adequately. During pregnancy large quantities of oestriol are produced and the foetal adrenal and liver are involved in the production of oestriol in conjunction with the placenta.

The ovaries produce 17 beta-oestradiol from cholesterol. Oestradiol is secreted by the granulosa cells of the ovarian follicles and, after luteinisation, by the same cells in the corpus luteum. Some oestrogen is also produced by the adrenal gland from the aromatisation of androgens. The pathway of aromatisation of testosterone produces oestrogen, not the other way around. Like other steroids, oestradiol acts upon intracellular/nuclear receptors. Oestradiol does not require to be metabolised into a more potent substance for its physiological action. It is a steroid hormone and requires active internalisation and interaction with intracellular receptors to exert its effects. It does not require cyclic adenosine monophosphate (cAMP) for its action.

In pregnancy, the major oestrogen formed is oestriol, due to conversion of the foetal precursor 16-hydroxy-dehydroepiandrosterone (16OH-DHEA) by the placenta. As foetal 16OH-DHEA is the main substrate for the oestriol, urinary oestriol excretion of the mother can be monitored as an index of the state of the foetus. SHBG binds around 69% of oestradiol in the blood, 30% is bound to albumin, and 1% is free and biologically active. Oestrogens are excreted following conjugation in the liver with glucuronic acid, and to a lesser extent, with sulphuric acid and thereafter appear in the urine. About 70% of oestrogen is excreted in urine, and 30% in faeces. A rise in oestrogen concentration will suppress the release of GnRH. In turn the pituitary gland does not release FSH and LH. Inhibin, on the other hand, which inhibits the secretion of FSH, is produced by the corpus luteum while it is active.

Oestrogens stimulate the growth and activity of the mammary glands and the uterine endometrium during the follicular phase. Oestrogens are also responsible for the development of secondary sexual characteristics. Oestrogens cause the hypertrophy of the uterine tubes and lower genital tract which takes place at puberty, and the hypertrophy and increased vascularity during pregnancy. Oestrogens cause increased rhythmical contraction of the fallopian tube. Oestrogens cause proliferation of the vaginal stratified cells and causes deposition of glycogen in vaginal epithelium. This facilitates the formation of lactic acid by Doderlein's bacilli, thereby increasing resistance to infection. It increases the number of progesterone receptors in the endometrium. Oestrogens inhibit the release of prolactin by the anterior pituitary gland during pregnancy. It also helps inhibit lactation during puerperium. Oestradiol decreases blood and urine levels of calcium, but it increases the calcification of bone. Oestradiol also increase the level of clotting factors VII, VIII, IX and X.

Progesterone

Progesterone is mainly secreted in the second half of the cycle by the corpus luteum. It is a thermogenic hormone, which is probably responsible for the rise in basal body temperature at the time of ovulation. Progesterone is rapidly metabolised by the liver, and approximately 20% is excreted in the urine as sodium pregnanediol glucuronide. About 2% is free, 80% is bound to albumin, and 18% is bound to corticosteroid-binding globulin. Large doses of progesterone produce natriuresis. This probably occurs due to the blocking the action of aldosterone on the kidney by progesterone. Pregnanediol glucuronide is the major metabolite of secreted progesterone. The progesterone receptor is decreased by progestins at both the transcriptional and translational levels, and is induced by oestrogen at the transcriptional level. Progesterone levels are highest in the mid-luteal phase. The progesterone assay in the mid-luteal phase, about day 21 in the normal 28 day cycle, is the standard method of confirming ovulation in the patient seeking fertility. A concentration above 30 nmol/L is confirmatory of ovulation.

Progesterone matures the endometrium in preparation for implantation (secretory change) and supports the early pregnancy. During the luteal phase, progesterone blocks the positive feedback effect of oestradiol on LH. It also enhances the negative feedback effects of oestradiol. Thus injections

of oestradiol into women during luteal phase are not followed by an LH surge. During pregnancy, progesterone inhibits the stimulatory effect of oestrogen only until the last few weeks of pregnancy.

Progesterone is produced by corpus luteum until the midluteal phase. In case pregnancy occurs, corpus luteum persists and continues producing progesterone. Persistence of the corpus luteum is believed to be due to human chorionic gonadotropin (hCG) from the trophoblast. The corpus luteum persists in early pregnancy for about 8 weeks when its functional role is taken over by the placenta. In case the pregnancy does not occur, corpus luteum regresses and the progesterone concentrations fall until the next luteal phase.

Inhibin

Inhibin is a glycoprotein hormone produced and secreted by the granulosa and theca cells of the ovary. It inhibits the pulsatile secretion of FSH. Inhibin and relaxin are structurally dissimilar. Two types of inhibins have been described: A and B. Inhibin A is produced by the dominant follicle, the corpus luteum and by the placenta. It is thought to have some role in the control of the menstrual cycle, perhaps the LH surge. Inhibin B is produced by the early follicles and the emerging dominant one and is a hormone of the first half of the menstrual cycle. It is not found in the luteal phase or in pregnancy. It is thought to have a paracrine function in suppressing other follicles to allow emergence of the dominant one.

Normal Menstrual Cycle

The normal menstrual cycle can be divided up into three phases **(Fig. 11.12)**:
1. Follicular phase (proliferative)
2. Ovulation
3. Luteal phase (secretory phase).

Proliferative (Follicular) Phase

The proliferative phase begins from approximately 5th day of the menstrual cycles at the end of the menstrual periods. Normal menstrual periods last for about 5 days, starting from the day 28th which is the last day of each menstrual cycle. Normal menstrual periods involve the disintegration and sloughing of the functionalis layer of the endometrium. Interplay of various prostaglandins (e.g. prostaglandin F2-alpha and prostaglandin E2) is involved in the regulation of menstrual cycle. Prostaglandin F2-alpha causes myometrial contractions and vasoconstriction, whereas prostaglandin E2 causes vasodilatation and muscle relaxation. During the follicular phase GnRH is released from the hypothalamus in pulses every 90 minutes. By the 5th day of the menstrual cycle, the uterine endometrium shows proliferation of its stroma and glands and the glands start elongating. In the early proliferative phase, the cells lining the glands are cuboidal with definite limiting membranes and the stromal cells are thin and spindly. In a week's time (12th day), the time of late proliferative phase, the glands become very large and dilated. The blood vessels become more prominent and capillaries are dilated.

The cells of the theca interna of the follicle are the primary source of oestrogen. The early phase, follicular phase of the cycle corresponds with the development of follicles. The endometrium thickens throughout the cycle. Plasma oestrogen levels rise in the first half of the cycle. There is a rapid rise in oestrogen production, mainly oestradiol, between days 12 and 13 when levels peak.

Selection of the dominant follicle occurs by days 5–7 of the menstrual cycle. As oestrogen concentrations from the dominant follicle increase, FSH is inhibited centrally by negative feedback inhibition. This causes withdrawal of gonadotropin support towards the less developed follicles. The dominant follicle escapes the atretic consequences of falling FSH levels because it has a greater number of FSH receptors due to an increased mass of granulosa cells. Furthermore, the increased vascular development within the theca layer offers preferential delivery of FSH to the dominant follicle. By altering the gonadotropin secretion by its own production of oestrogen, the dominant follicle optimises its own environment to the detriment of other follicles.

When the dominant follicle reaches maturity at around day 12 of a 28 day cycle, the elevated level of oestradiol acts in a positive way on the hypothalamus which causes increased pulse frequency and amplitude of GnRH release at about 14th day of the cycle. This induces a surge in the output of LH and a concomitant smaller rise in the output of FSH. The following changes take place in the endometrium during the proliferative phase:

❑ The functional and the basal layers of endometrium become well-defined. The proliferation mainly occurs in the functional layer. The basal layer measures 1 mm in thickness, while the functional layer reaches a maximum thickness of about 3.5–5 mm by 14th day.

❑ The glands become elongated and slightly sinuous and the columnar epithelium lining them becomes taller. In the beginning, the glands are narrow and tubular, lined by low columnar epithelial cells. Mitosis becomes prominent and the areas of pseudostratification are observed.

❑ There is an increase in ciliated and microvillus cells in the endometrial glands.

❑ Endometrial stroma becomes oedematous with wide separation of the individual cells. The stroma gets infiltrated with numerous cells including macrophages, leukocytes, etc.

❑ In the initial phase, the spiral vessels are uncoiled and unbranched. However, soon the growth of the straight vessels occurs so that they start becoming more coiled and spiralled.
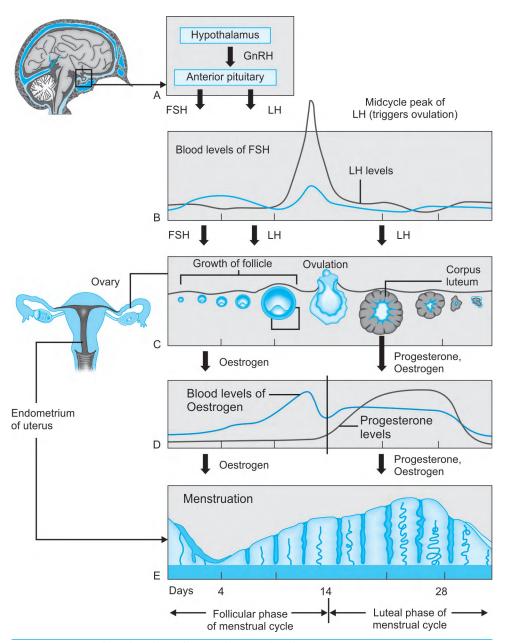
FIG. 11.12: Events taking place during the normal menstrual cycle
Abbreviations: GnRH, gonadotropin-releasing hormone; FSH, follicle-stimulating hormone; LH, luteinising hormone

Ovulation

The dominant follicle grows considerably in size throughout the proliferative phase and measures about 20 mm in size by the time of ovulation. Ovulation occurs approximately 34–36 hours after the start of the LH surge or 10–12 hours after the peak of the LH surge. Ovulation takes place on the 14th day of a 28 day cycle, or 14 days before the onset of the next menstrual period. At the time of ovulation, the dominant follicle reaches the surface of ovary and it it released due to the necrobiosis of the overlying tissues. The length of the luteal phase is always fixed to 14 days, while the length of the proliferative phase can be variable. The LH surge occurs only if there has been correct formation of dominant follicle

with appropriate secretion of oestrogen. At the time of ovulation, secondary oocyte is liberated from the follicle through rupture of the theca and is produced along with the first polar body as a result of the first meiotic division. Both of these cells are encased in a thick glycoprotein shell called the zona pellucida. Immediately following ovulation, the oocyte is also surrounded by a corona radiata. The oocyte also contains cortical granules. Mittelschmerz syndrome is the lower abdominal pain, occurring in the midcycle, experienced by nearly 1 in 4 women. This is thought to occur due to ovulation. Rupture of ovum is likely to cause the release of follicular fluid, resulting in peritoneal irritation.

Progesterone enhances the activity of proteolytic enzymes responsible, together with prostaglandins, for

digestion and rupture of the follicular wall. Ovum release takes about 2 minutes. On day 23 of a normal menstrual cycle, the appearance of subnuclear vacuolation on endometrial biopsy strongly indicates that ovulation has occurred. Ferning of the cervical mucus is another evidence that ovulation has occurred. Ovulation on average occurs 45 days after delivery in non-lactating women. Ovulation is unlikely to occur for up to 70 days in lactating women.

Secretory (Luteal) Phase

This phase of menstrual cycle following ovulation is termed as the luteal phase. During the luteal phase the pulse amplitude and pulse frequency decrease considerably and the pulsatile release of GnRH falls to every 120 minutes. Following ovulation, the ruptured ovarian follicle turns into the corpus luteum, and the granulosa cells and theca interna cells of the ruptured follicle form the corpus luteum. Corpus luteum mainly produces progesterone and oestrogen. The levels of progesterone rise after about a week following ovulation, peaking around the day 19 and then declining. Oestrogen concentrations fall after ovulation with a rise in progesterone concentration in preparation for implantation. Since progesterone is a thermogenic hormone, there occurs a rise in body temperature of between 0.5°C and 1°C at the time of ovulation. The temperature remains elevated throughout the luteal phase as a marker of progesterone activity, until a few days before the next period. Premenstrual mastalgia is a fairly reliable, but not definitive clinical evidence that ovulation has occurred. Following ovulation, the granulosa cells and the theca interna cells of the ruptured follicle form the corpus luteum.

The endometrial changes taking place in the secretory phase include the following:

❑ The most characteristic feature of this phase is development of subnuclear vacuolation in the glandular epithelial cells. In this, the glycogen filled vacuoles develop between the nuclei and the basement membrane (by the day 17–18). This is the first evidence that ovulation has taken place.
❑ The endometrium measures about 8–10 mm in the secretory phase. The secretory phase reaches its peak activity by the 22nd day of the cycle after which no growth occurs.
❑ The glands become crenated and tortuous to assume a characteristic corkscrew-shaped appearance. The corkscrew pattern of the glands becomes sawtoothed in the later part of the secretory phase.
❑ The stroma of the functional layer becomes oedematous further.
❑ The functional layer of the endometrium can be divided into two layers: (1) superficial or compact layer; (2) deep spongy layer.

❑ The spiral vessels become dense and deeply coiled.

If an embryo is not present or implantation does not occur, the corpus luteum undergoes degeneration over the last 7 days of the menstrual cycle and undergoes hyalinisation losing its fat and becoming pale in appearance and is then known as the corpus albicans. The overgrown secretory endometrium during this phase is unable to support itself and therefore it sloughs off at the end of the menstrual cycle (day 28), resulting in menstrual bleeding or periods. The average blood loss during each menstrual cycle is between 40 mL and 80 mL.

On the other hand if the pregnancy occurs, cells around the embryo secrete hCG, which rescues the corpus luteum, keeping it active and secreting progesterone. The corpus luteum (yellow body) is composed of the following:

❑ *Large cells*: Large centrally located granulosa lutein cells that are lipid rich giving the characteristic yellow colour
❑ *Small cells*: Theca lutein cells which are small peripherally located cells derived from the cells of the theca
❑ Pericytes/endothelial cells assist in angiogenesis to feed and transport the endocrinologically active tissue.

The small cells and the large cells of the corpus luteum are responsible for producing progesterone. The large cells are more active in steroidogenesis and are influenced by various autocrine/paracrine factors such as inhibin, relaxin, and oxytocin. The role of prolactin in the menstrual cycle remains to be clearly defined but its presence is thought to be imperative for normal follicular development.

After the sloughing of the uterine endometrium, prevention of clot formation is necessary to prevent scarring and obliteration of the endometrial cavity. For this, there appears to be an active fibrinolytic system in the endometrium in the late secretory phase mediated by plasmin. Therefore, there are increased fibrinolytic activators in the late secretory phase. Prostaglandins are produced in higher amounts in the late secretory phase/at menstruation leading to dysmenorrhoea and to help with vasoconstriction for reducing the blood loss at menstruation. Platelet activating factor (PAF) and endothelins are produced by the endometrium. Lower levels of the substances enumerated in **Table 11.12** have been reported in the menstrual fluid in comparison with the peripheral plasma:

TABLE 11.12	Substances present in reduced concentration in the menstrual fluid

• Activated prothrombin
• Antithrombin
• Antiplasmin
• Plasminogen
• Protein C and
• Factors V, VII, VIII and X

Puberty and Adolescence

Puberty

The sequence of pubertal maturation in females usually follows a predictable pattern. The various events being thelarche (appearance of breast tissues); pubarche (development of pubic hair) and menarche (onset of menses). Pubarche usually occurs as a result of adrenal androgen secretion (adrenarche). The reactivation of hypothalamic-pituitary-gonadal axis at the time of puberty can cause a rise in the levels of FSH and LH. This causes a gradual rise in oestradiol concentration which stimulates the breast development. Therefore, there is an increased secretion of androgen and oestradiol at the time of puberty in girls.

The first event of puberty is thought to be the nocturnal release of gonadotropin-releasing hormone from the hypothalamus. This stimulates the release of LH and FSH from the pituitary gland. This in turn, stimulates the synthesis of oestrogen and testosterone in the ovaries and testes of young girls and boys.

Puberty tends to begin from the age of 10 with the range for menarche between the ages of 11–15 years. The age of onset of puberty has been declining in the US over the past century by approximately 6–12 months. On an average, black American girls begin puberty between the ages of 8–9 years, while white American girls experience it by the age of 10 years. Early pubertal development is associated with a slightly reduced adult height and an increased risk for obesity in comparison to a late menarche. Mean age of occurrence of menarche is 12.9 years with a range of 11–15 years. In the beginning when the menstrual cycles first start, they may be anovular in nature with no follicular development due to irregular GnRH pulse frequency.

In general the four signs of puberty in most adolescent girls are an acceleration of growth, followed by breast budding (thelarche), and followed by the appearance of pubic hair (pubarche) and finally the onset of menses (menarche). This sequence of events occurs over a period of 1–6 years (average 4–5 years). In a substantial number of cases, the sequence of events may be reversed with pubarche preceding thelarche. In girls, the first sign of pubertal development can be considered as breast development. On the other hand testicular enlargement is the first sign of puberty in males and the growth spurt occurs at the final phase in males.

The menarche often begins with anovulatory cycles and traditionally is preceded by pubertal development including development of pubic hair, breast development and growth spurt. The peak height velocity in females occurs just before menarche. The peak height velocity is 6–11 cm/year in females but 7–13 cm/year in males. If peak height velocity is reached earlier than 8 years in females investigations

should be undertaken. Growth in Turner's syndrome is characterised by intrauterine growth retardation in infants and relatively normal rate of growth in the first few years of life. It then decelerates and the pubertal growth spurt does not occur.

Staging system for describing the physical changes of puberty were first described by Marshall and Tanner and is known as the Tanner staging system. There are five Tanner stages of breast and pubic hair development in girls, with stage 1 representing the prepubertal stage and stage 5 representing the adult development. The Tanner stages of breast development are described in **Table 11.13** and **Figure 11.13**, whereas Tanner stages for the development of pubic hair is described in **Table 11.14** and **Figure 11.14**.

In extremely premature babies born with retinopathy of prematurity, menarche has been observed to occur earlier, in comparison with their peers. Low body mass index (BMI) in adolescent women may result in delayed menarche as occurring in the cases of anorexia nervosa. Anorexia nervosa is an important cause of hypogonadotropic hypogonadism. In cases of anorexia nervosa, levels of LH and FSH are low and LH response to luteinising hormone-releasing hormone (LHRH) is impaired when weight loss is severe. Levels of cortisol and GH are elevated in these cases. Other electrolyte abnormalities which may occur in these cases include hypokalaemia, hypoalbuminaemia, anaemia, leukopenia, raised serum carotene levels, etc.

Adolescent Growth Spurt

There is approximately 17–18% gain in adult height during puberty. The growth spurt occurs 2 years earlier in girls in

TABLE 11.13	Tanner stages of breast development
Stage	**Characteristic features**
Stage 1	Prepubertal stage
Stage 2	Breast budding—enlargement and widening of the areolae
Stage 3	Breast enlargement beyond the areolae
Stage 4	Further enlargement of the breasts with the areolae and nipples forming the secondary mounds
Stage 5	Adult breast contour

TABLE 11.14	Tanner stages for development of pubic hair
Stage	**Characteristic features**
Stage 1	Prepubertal hair
Stage 2	Presexual hair—occurrence of long, relatively straight hair on labia majora
Stage 3	Sexual hair—the hair becomes curly, coarser and there is an outwards extension.
Stage 4	Mid-escutcheon—further extension of the hair to cover the labia
Stage 5	Female escutcheon—extension of hair over the medial thighs.
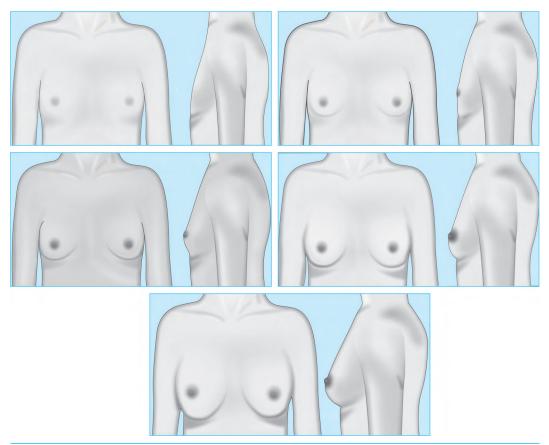
FIG. 11.13: Tanner stages of breast development

I	II	III
IV	V	
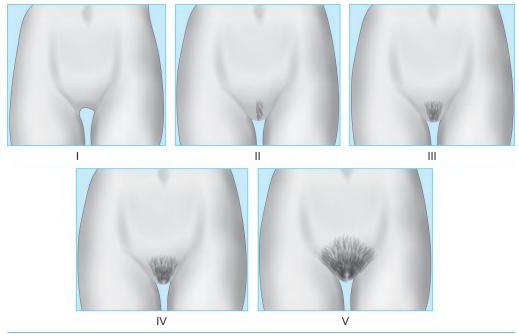
FIG. 11.14: Tanner stages of pubic hair development

comparison to the boys. The peak gain in height is reached approximately 6 months before menarche.

Accumulation of bone mass which occurs during puberty is critical for the development of peak bone mass in females. This serves as an important determinant for development of osteoporosis later in life. Peak of bone mass accumulation in women occurs about 9–12 months after peak velocity in height has been attained, usually at the time of menarche.

Pubertal growth is related to an increase in the circulating levels of GH and IGF-1. There is also a role of sex steroids in the stimulation of pubertal growth spurt. However, rising levels of sex steroids ultimately limit adult height by stimulating epiphyseal fusion.

Following menarche, the growth slows down and does not increase for more than 6 cm (2.4 inches). With the occurrence of menarche, the initial periods are anovulatory, irregular, and heavy and may persist for 12–18 months. Soon these cycles become ovulatory in nature with the maturation of oestrogen positive feedback mechanisms.

Changes in Vagina

During puberty, oestrogens cause proliferation of the vaginal stratified cells and increase their glycogen content. After puberty, during the reproductive phase, Doderlein's bacilli appear in the vagina and produce lactic acid by acting on the glycogen in the epithelial cells. This results in a vaginal secretion with an acidic pH of about 4. This leads to an increasing resistance to infection. Mature vaginal epithelium has no glands. In the sexually mature woman, the epithelial squamous cells form four distinct histologic zones:
1. *Basal cells*: This comprises of a single layer of cuboidal cells attached to the basement membrane. These cells are firmly attached to the basement membrane and do not exfoliate. Therefore, these cells are not present in vaginal smears. They are the least mature cells from which regeneration of the vaginal epithelium is maintained.
2. *Parabasal cells*: A zone of several rows of polyhedral cells lies superficial to the basal layer. These cells may be seen in Pap smears. When these cells exfoliate they lose their intercellular bridges and appear round or oval in the vaginal smears.
3. *Intermediate layer*: This layer comprises of several rows of slightly larger, flatter cells lying above the parabasal cellular layer. When these cells exfoliate, they appear larger and less rounded than the parabasal cells. When examined cytologically, the lower most parabasal cells stain bluish-green and those near the surface take up the eosinophilic stain.
4. *Superficial zone*: This comprises of several layers of large, flat cells with dark pyknotic nuclei. In Pap smears, these cells appear large and polyhedral having a clear transparent cytoplasm. The superficial squamous cells in this zone usually stain pink unless the pH of the vagina is abnormal. Under certain pathological conditions, these cells may lose their nuclei and become keratinised. In these cases, they take up an orange colour on Papanicolaou stain.

Leptin

Leptin is a recently-described hormone. It is a helical molecule comprising of 167 amino acids and is the product of *ob* gene in the white fat cells. It acts upon areas within the hypothalamus to suppress hunger. It is the member of tumour necrosis factor group of cytokines. Its function remains unclear, but it is probably the link between the body weight and menstruation. It is produced in fat. It is believed to influence the hypothalamus, affecting gonadotropin production. It is also likely to increase the production of insulin. Hyperinsulinaemia on its own is associated with numerous problems such as PCOS, obesity, elevated LH levels and amenorrhoea that can be difficult to treat. It is thought that leptin may play a part in the fact that puberty tends to occur earlier in girls who are heavier for their height. However, there is not a critical BMI below which sexual maturation fails to occur. In animal models a deficiency of leptin is associated with obesity and circulating concentrations correlate with BMI. Gender is an important factor determining plasma leptin, with women having markedly higher leptin concentrations than men for any given degree of fat mass.

Precocious Puberty

Precocious puberty is defined as pubertal development occurring more than 2.5 standard deviations earlier than the average age. If the average age of puberty were considered to be 10 years, the development of secondary sexual characteristics before 8 years in females would be defined as precocious puberty. Development of secondary sexual characteristics before 9 years in males is also considered precocious. Precocious puberty is characterised by development of both breasts and pubic hair in girls and by the development of pubic hair and testicular enlargement in boys (testicular enlargement can be defined as an increase in the volume of greater than 4 mL or a diameter of 2.5 cm).

Precocious puberty may be of two types: gonadotropin dependent precocious puberty due to central origin (true), or gonadotropin independent precocious puberty due to excessive sex steroid production (pseudo or false). The differences between the two are described in the **Table 11.15**.

McCune-Albright syndrome is a syndrome characterised by the presence of various abnormalities including polyostotic fibrous dysplasia of the skeletal system, patchy cutaneous pigmentation (in form of café-au-lait spots) and precocious pubertal development. The condition affects girls more often than boys. The endocrinologic

TABLE 11.15	Differences in between gonadotropin dependent and gonadotropin independent precocious puberty
Gonadotropin dependent precocious puberty	**Gonadotropin independent precocious puberty**
Also known as central precocious puberty or true precocious puberty	Also known as peripheral precocious puberty or pseudo-precocious puberty
Is characterised by early maturation and activation of the hypothalamic-pituitary-gonadal axis. However, the sequence of pubertal events is normal and proceeds at a normal pace	It is independent of GnRH and gonadotropins. It usually results from exposure to sex-steroid hormones, which may be derived from the gonads, adrenals, or the environment.
It is isosexual in nature, i.e. developing sexual characteristics are consistent with the child's gender	It could be either isosexual or contrasexual in nature, where sexual characteristics are inconsistent with the child's gender (e.g. virilisation in girls or feminisation in boys)
Levels of gonadotropins, particularly follicle-stimulating hormone (FSH) and luteinising hormone (LH) are increased.	Levels of gonadotropins are low.
Causes Idiopathic (70–80%) Central nervous system lesions: congenital such as hydrocephalus, acquired such as post-irradiation, infection or surgery, cysts, or tumours such as microscopic hamartomas, astrocytomas, ependymomas, pineal tumours, optic and hypothalamic gliomas) Hypothyroidism	*Causes* McCune-Albright syndrome; Ovarian tumours (granulosa tumour, malignant teratoma or Arrhenoblastoma of ovary); adrenal cortical lesions (tumour or hyperplasia); and testicular disorders (Leydig cell tumours) and administration of exogenous sex steroids

Abbreviation: GnRH, gonadotropin-releasing hormone

manifestations of the disease appear to reflect the autonomous hyperfunctioning of the peripheral target glands. The disorder is related to the somatic mutation of the gene encoding a sub-unit of the G-protein. This results in some cells bearing constitutively active adenylate cyclase. There also may be continuous stimulation of endocrine function resulting in abnormalities such as gigantism, Cushing syndrome, adrenal hyperplasia, thyrotoxicosis, etc. Early and repeated exposure to sex steroids can result in advanced bone age and ultimately reduced adult height. The diagnosis of McCune-Albright Syndrome must be considered in girls with recurrent functional ovarian cysts and episodic menses.

Investigations

Following the history and physical examination, the individuals who require further endocrine evaluation and imaging include the following:
- ❏ Children with advanced bone age
- ❏ Children with normal bone age accompanied by the development of both breasts and pubic hair.
- ❏ Children with normal bone age associated with accelerated growth and breast or pubic hair development.

The two types of precocious puberties can be differentiated from one another by measurement of GnRH-stimulated serum gonadotropin levels. Measurement of the stimulated serum LH concentration can be considered as the most useful diagnostic parameter. In case of gonadotropin-dependent precocious puberty as evidenced by elevated basal or stimulated serum LH levels, MRI head is indicated to exclude an intracranial lesion.

In case of gonadotropin-independent precocious puberty (as evidenced by normal basal and stimulated LH levels), other tests which need to be done include:

- ❏ Serum concentration of oestradiol, testosterone and hCG (for detection of functional ovarian cysts, tumours and functional adrenal tumours)
- ❏ Late afternoon cortisol levels (Cushing syndrome)
- ❏ DHEAS (premature adrenarche)
- ❏ 17-OHP (congenital adrenal hyperplasia).

Delayed Puberty

Delayed puberty can be defined as the failure to begin sexual maturation at an age which is 2.5 SD above the mean age of onset of puberty. In US the evaluation of delayed puberty is recommended as 13 years in girls and 14 years or older in boys who do not demonstrate any signs of sexual maturation. The first sign of sexual maturity can be considered as thelarche in girls and testicular enlargement in boys. However, if other secondary sexual characteristics have developed normally in a girl with primary amenorrhea, an expectant approach can be adopted until the age of 16 years, following which the girl must then be investigated.

The main cause of delayed puberty is hypogonadism, which could be either hypogonadotropic hypogonadism (inactive hypothalamic-pituitary axis) or hypergonadotropic hypogonadism (primary gonadal failure). Causes of delayed puberty are described below:

Causes of Delayed Puberty

Hypogonadotropic hypogonadism:
- ❏ Functional GnRH deficiency, reflecting a constitutional delay in the reactivation of hypothalamic-gonadal axis
- ❏ Normal variation, sometimes familial
- ❏ Suppressive effects of chronic stress due to illness, malnutrition or excessive exercise
- ❏ Systemic disease, for example, malnutrition, cystic fibrosis, renal failure, heart disease and malabsorption

- Genetic defects (Kallman's syndrome)
- Anatomic abnormalities (hypothalamic and pituitary tumours, (for e.g. craniopharyngioma)
- Thyroid deficiency (hypothyroidism)
- Hyperprolactinaemia
- Pituitary failure
- Androgen receptor defect, for example, testicular feminisation or partial defects
- Anorexia nervosa
- Emotional deprivation
- Excessive exercise.

Hypergonadotropic hypogonadism:
- Primary gonadal failure (primary testicular failure, e.g. Klinefelter's syndrome or primary ovarian failure, e.g. Turner's syndrome)
- Previous treatment of malignancy (gonadectomy, chemotherapy, gonadal irradiation, etc.).

Pathologies such as imperforate hymen and congenital absence of uterus do not cause delayed puberty. Initial evaluation in these patients comprises of history, physical examination and measurement of bone age.

Constitutional Growth Delay

Constitutional growth delay is a temporary delay in skeletal growth and height of a child with no other physical abnormality causing the delay. It is the most important cause of delayed puberty and short stature. In cases of constitutional delay of puberty, normal prepubertal growth nadir is prolonged and pulsatile GnRH secretion is slow to develop. Family history is often positive with the siblings or parents also giving a history of delayed puberty. Investigations demonstrate the bone age to be delayed. Bone age (X-ray of left hand and wrist) is delayed in cases of both constitutional delay and GnRH deficiency. If the bone age reaches 13 years in girls or 14 years in boys without the evidence of puberty, the patient is more likely to have GnRH deficiency rather than constitutional delay of puberty. Also, in cases of constitutional delay of puberty, laboratory evaluation reveals prepubertal testosterone levels and low or normal gonadotropin levels. In cases of constitutional delay of puberty, even though the puberty is delayed, it begins before the bone age of 13 years in girls or 14 years in boys. Height is often less than 5th percentile, but growth rate is normal for the skeletal age. Onset of adrenarche is delayed. Laboratory tests may resemble hypogonadism and it may be difficult to differentiate constitutional delay of puberty from idiopathic hypogonadotropic hypogonadism.

Differentiating between hypogonadotropic and hypergonadotropic hypogonadism can be achieved by measuring the levels of gonadotropins such as FSH, LH and female hormone oestradiol. Low or normal levels of gonadotropins is indicative of hypogonadotropic hypogonadism, whereas elevated levels of gonadotropins is indicative of hypergonadotropic hypogonadism. In case of hypogonadotropic hypogonadism, the following tests are indicated:

- *Serum prolactin levels*: In cases of raised prolactin levels, imaging by MRI is indicated to rule out a pituitary lactotropic adenoma, except in cases where hyperprolactinaemia can be attributed to be secondary to medications.
- *Measurement of serum TSH and free thyroxine levels*: Measurement of these can help in identifying the cases of primary and secondary hypothyroidism.
- *Measurement of serum DHEA-S concentration*: Measurement of serum DHEA-S levels is important to differentiate between constitutional growth delay and GnRH deficiency. Patients with congenital GnRH deficiency are likely to have normal adrenarche and therefore normal levels of DHEAS.
- *Other tests*: Other tests such as complete blood count, ESR, LFT, etc. These tests help in ruling out chronic illnesses.

In cases of hypergonadotropic hypogonadism, karyotype analysis must be performed.

Karyotype analysis: In girls with hypogonadotropic hypogonadism, a karyotype analysis should be done to rule out chromosomal abnormalities (especially Turner's syndrome and Klinefelter's syndrome).
- In a short female chromosome analysis should be performed to rule out Turner's syndrome.
- High serum testosterone indicates androgen insensitivity (male pseudohermaphroditism) which comprises of a normal male karyotype (46 XY) with an abnormality in the androgen receptor. This results in unresponsiveness to androgens, which prevents musculanisation of male genitalia. As a result, there is development of female external genitalia. As a result, the person is phenotypically a female with normal breasts, but absent pubic hair. There may be presence of intra-abdominal testes.

Treatment comprises of treating the underlying cause such as thyroid hormone therapy for hypothyroidism, therapy with dopamine agonists for hyperprolactinaemia, excision of craniopharyngioma, etc. Hormone therapy can be initiated in patients with congenital GnRH deficiency or constitutional delay of puberty over 12 years having no signs of sexual maturation and causing significant distress or anxiety.

Anovulation

Anovulation may occur in association with PCOS, anorexia/marked weight loss but is also associated with obesity. Hyperprolactinaemia through negative feedback on gonadotrophs in the pituitary produces hypogonadotropic hypogonadism and amenorrhoea.

Propranolol is not associated with anovulation.

Anovulatory cycles are typical of the early years of menstruation and are also common in the run up to the menopause. Anovulatory cycles are usually painless. They are common in polycystic ovary syndrome in which the endometrium is exposed to long term oestrogen. Without the maturing effect of progesterone hyperplasia is the result. This is a risk factor for endometrial cancer, but the anovulation protects against ovarian cancer. Women with anovulatory cycles do not need to be treated unless there is a problem.

Menopause

Menopause can be described as the cessation of normal menstruation. Menopause is defined as cessation of menses for a minimum of 6 months because of inadequate ovarian follicular development and declining oestrogen production.

Climacteric (perimenopause or the menopausal transition) is the phase of waning ovarian activity, which may begin 2–3 years before menopause and may continue 2–5 years after it. This can be regarded as the phase of transition between the active and inactive ovarian function. The period of menopausal transition varies from 2 years to 8 years. The average age of menopause in the United Kingdom is 51 years, with a large majority of women experiencing menopause between the ages of 45 and 55 years. The cessation of periods can occur suddenly or may be preceded by light and infrequent periods.

❑ The length of the menstrual cycle increases, beginning 2–8 years before the onset of menopause when women are in their forties because the cycles become anovular. The increased duration of the follicular phase is the major determinant of the length of menstrual cycle in these cases. Menstrual cycle changes prior to the menopause are marked by elevated levels of FSH and decline in the inhibin levels. Decrease in inhibin levels allows an increase in the FSH levels. Soon there is disappearance of ovarian follicles, resulting in the decline of oestrogen concentration. Declining concentrations of oestradiol, inhibin and progesterone help in relieving feedback inhibition on the pituitary gland, resulting in elevated concentrations of FSH and LH. Eventually there is a 10- to 20-fold increase in mean serum levels of FSH and a three-fold increase in LH levels. FSH levels are higher than LH levels because the half-life of LH is much shorter in comparison to that of FSH. A FSH level more than 30 on two occasions is usually regarded as diagnostic. Changes in the levels of various hormones at the time of menopause are depicted in **Figure 11.15**.

❑ The circulating oestradiol level after menopause decline substantially and measure about 10–20 pg/mL (40–70 pmol/L). Most of it is derived from peripheral conversion of oestrone. With the disappearance of follicles and declining oestrogen levels, the elevated gonadotropins
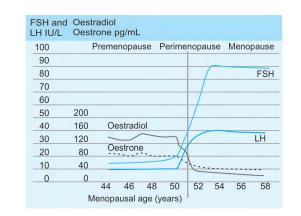
FIG. 11.15: Changes in the levels of various hormones at the time of menopause

drive the remaining stromal tissue in the ovary to produce testosterone. Thus, the postmenopausal ovary in most women secretes more testosterone than the premenopausal ovary. Oestrogen deficiency is not total as oestrogen is still produced from the adrenal and via aromatisation of androgens. Premenopausally, oestradiol is the oestrogen preferentially secreted. Postmenopausally, oestrone is the oestrogen with highest concentrations. Postmenopausally, there is also a significant drop in testosterone concentrations as this too is secreted by the ovary. There is a slight fall in prolactin concentrations after the menopause.

Menopausal Symptoms

Various effects related to menopause are summarised in **Table 11.16** and include the following:

❑ *Vaginal dryness*: Signs and symptoms of vaginal dryness include dryness, itching, burning, pain or light bleeding with sexual intercourse, increased urinary frequency or urgency.

❑ *Cessation of periods*: This could be a sudden cessation or gradual diminution in the amount of blood loss for each successive menstrual period, until the menstrual flow eventually ceases. Postmenopausal bleeding should be investigated by uterine curettage and cervical smear.

❑ *Hot flushes*: Hot flushes and sweating commonly occur as a result of vasomotor disturbances and may be present in nearly 85% of women. The flush coincides with a surge of LH and is preceded by a subjective prodromal awareness that a flush is beginning. It is often preceded by a headache. "Hot flushes" are more frequent and severe during the night, when a woman is often awakened from sleep. The flush persists for longer than 5 years in as many as 25–50% of women. Hot flush can be defined as an acute sensation of heat and skin changes, which may be associated with profuse perspiration. Skin changes may be in form of reddening of the skin over neck, chest and head accompanied by an increase in heart rate and a feeling of intense body heat.

TABLE 11.16	Various effects related to menopause		
Immediate effects		**Intermediate effects**	**Long-term effects**
• Vasomotor symptoms (hot flushes, sweating, palpitations) • Mood swings (depression, anxiety, irritability) • Sexual dysfunction (dyspareunia and reduced libido) • Urinary symptoms (dysuria, lower recurrent urinary tract infection, urgency, etc.) • Insomnia • Reduction of the rapid eye movement (REM) sleep time • Sexual dysfunction • Cognitive dysfunction (memory loss, poor concentration, tiredness, loss of motivation, etc.)		• *Genital atrophy*: thinning of the vaginal mucosa, loss of superficial keratinised cells, reduced secretions from the glands and an increase in the vaginal pH • *Reduction in collagen support and atrophy*: Skin changes, easy bruising, and an increased vulnerability to trauma and infection • *Urodynamic changes*: Stress incontinence, urgency and an increased frequency of urination • Pelvic organ prolapse	• *Osteoporosis*: It is due to oestrogen deficiency and affects the trabecular bone • Cardiovascular effects • Dementia

It is mediated by noradrenaline and serotonin. There is an increase in core body temperature and vasodilatation during hot flushes.

Treatments for Hot Flushes

Hormone replacement therapy (HRT) is the most effective treatment for hot flushes. Avoidance of alcohol, caffeine and spicy foods can help. Other drugs, which can help provide a relief from hot flushes, include the following:

❑ *Tibolone*: This is a unique drug and serves as a substitute for oestrogen-based HRT.

❑ *Testosterone therapy*: The testosterone therapy can be helpful for those with loss of libido or general fatigue.

❑ *Progesterone drugs*: Progesterone may be a helpful therapy for treatment of hot flushes. It can cause side effects such as weight gain and mood changes. Progesterone is not absorbed if administered orally, so it has to be given as creams or suppositories.

❑ *Selective serotonin reuptake inhibitors (SSRIs)*: These drugs (especially Prozac) are mainly used for depression and for mood changes such as premenstrual syndrome. There is some evidence that they are also helpful for the treatment of hot flushes.

- *Osteoporosis*: There is likely to be a reduction in bone mineral mass, resulting in osteopenia and/or osteoporosis, which may predispose to fracture development.
- *Mental symptoms*: Mental depression may occur due to disturbed sleep and inability to cope up with the body changes. There may also be irritability and loss of concentration. Pseudocyesis (fear of pregnancy) and cancer phobia may develop in some women.
- *Neurological symptoms*: These may include paresthesias (sensation of pins and needles).
- *Libido*: Although many women experience reduced libido, some women may also experience an increase in libido due to riddance of menstruation and fear of pregnancy.
- *Urinary symptoms*: These may include symptoms such as dysuria, stress and urge incontinence and recurrent vaginal infections. Genital symptoms, such as dryness of vagina, dyspareunia, genital prolapse and urinary and/or faecal incontinence may also occur.

- *Long-term effects*: In the long term, menopause is likely to result in complications such as arthritis, osteoporosis, fracture, cerebrovascular accidents, ischemic heart disease, myocardial infarction (MI), atherosclerosis, stroke, skin changes, Alzheimer's disease, etc.

Osteoporosis

Osteoporosis is defined as a disease characterised by reduced bone mass and micro-architectural deterioration of bone tissue, resulting in enhanced bone fragility and an increased fracture risk. Oestrogen appears to control the function of both osteoclasts and osteoblasts in bone, and this influences the rate of absorption and deposition of calcium. Remodelling of bone continues throughout life, but following menopause, due to oestrogen deprivation, the osteoclastic activity far exceeds the osteoblast's ability to lay down calcium. Low levels of natural oestrogen around and after menopause diminish the body's ability to absorb calcium and to metabolise vitamin D. This results in the thinning of trabecular bone and eventually osteoporosis. This is often associated with an increased risk of fractures of the hip and wrist. Compression fractures of the vertebrae can often occur resulting in development of a dowager hump. Recurrent compression fractures often result in back pain. Various risk factors for the occurrence of osteoporosis are enumerated in **Table 11.17**. Vitamin D treatment is associated with improvements in bone mineralisation.

Treatment for Osteoporosis

Hormone replacement therapy: HRT helps reduce the rate of osteoporosis.

Tibolone: It appears to be as effective as HRT for reducing the risk of osteoporosis.

Bisphosphonates: These drugs increase the amount of bone. They are used for treating osteoporosis or osteopenia.

Selective oestrogen receptor modulators (SERMS): These are relatively new drugs and are also known as the designer oestrogens. SERMS affect the way that oestrogen receptors work. These are synthetic molecules which have the ability

TABLE 11.17	Risk factors for the occurrence of osteoporosis

- Low body weight/Low BMI (body mass index)
- Medications such as corticosteroids or heparin
- Cigarette smoking
- Excess alcohol intake
- A sedentary lifestyle
- Family history of osteoporosis
- Hyperparathyroidism
- Vitamin D deficiency
- Immobility
- Early menopause/primary ovarian failure.
- Increasing age
- Steroid therapy
- Thyrotoxicosis
- Early menopause
- Haemochromatosis

to bind with the oestrogen receptors, thereby acting as oestrogen agonists or antagonists depending upon the target organ. The first SERM was Tamoxifen. It has been used for reducing the risk of recurrence in women who had breast cancer. It exerts its action by blocking the effects of oestrogen on breast tissue. Tamoxifen may cause endometrial hyperplasia due to its partial agonist-like action on the endometrial receptors.

The first SERM used for the treatment of menopausal symptoms was Raloxifene. It can be used instead of oestrogen to prevent osteoporosis. It is effective and does not harm the breast or endometrium. However, it is not effective for the treatment of hot flushes.

Alternative treatments: Some of the alternative strategies for the treatment of menopausal symptoms include strategies such as acupuncture, reflexology, nutritional supplements (such as calcium, vitamin D, vitamin C, vitamin E, etc.), homeopathy, soya products (phytoestrogens), etc. Phyto-estrogens have chemical structures similar to those of human oestrogen. There are three main types: isoflavones, lignans and coumestans.

Phytoestrogens seem to work like the SERMS. Herbal treatments, which have been approved by Commission E for treatment of menopausal symptoms include black cohosh, chasteberry, Ginkgo, Ginseng, lemon Balm, passion flower, St. John's Wort, valerian, etc. Though these therapies are commonly used, there is little evidence regarding its effectiveness. But it has almost no risk of being toxic.

Treatment for Menopausal Symptoms

Hormone Replacement Therapy

Hormone therapy is generally prescribed for treating troublesome menopausal symptoms, such as hot flushes or vaginal dryness. HRT refers to the intake of supplements of hormones such as oestrogen alone or oestrogen in combination with progesterone (progestin in its synthetic form). HRT is not prescribed to all menopausal women. The hormones must be consumed for the shortest period of time possible. Short-term use of hormones (<5 years) is usually not associated with an increased risk of complications. However, long-term oestrogen therapy should be encouraged only if the patient is having adequate relief of her symptoms and is often required over a year or two with gradual dose reduction and acclimatisation to the symptoms. Current advice suggests not using HRT for more than 10 years due to the increased risk of breast cancer.

Oestrogen therapy removes the principal vasomotor and atrophic symptoms of the climacteric. There is also a mood enhancing effect in most women. There is now good evidence that HRT prevents osteoporosis. Nevertheless, currently oestrogen therapy is not considered to be a first-line therapy for prevention of osteoporosis. Presently, bisphosphonates and/or raloxifene have been recommended as the first-line treatment for prevention of osteoporosis. Raloxifene is a SERM and is indicated for postmenopausal osteo-protection. However, it may worsen flushes. It is current practice to prescribe only natural oestrogens which are preparations containing oestrone (E1) or oestradiol (E2) or oestriol (E3) which can be normally and easily metabolised by the body.

Rather than the original belief that post-menopausal HRT reduces cardiovascular risk, various studies have demonstrated an increased risk of cardiovascular mortality and morbidity associated with HRT. Oestrogen therapy is also no longer indicated for the prevention of cardiovascular heart disease (CHD) in postmenopausal women. This recommendation initially made by the American Heart Association (AHA) in 2001 has been further strengthened by the results of the Women's Health Initiative (WHI) and Heart and Estrogen/Progestin Replacement (HERS)-II trials. The emerging evidence suggests that the risk of CHD events with HRT is majorly limited to older postmenopausal women, with younger postmenopausal women at very low risk for CHD-related events. In older women, HRT may be associated with an increased risk of CHD, venous thromboembolism and stroke. This increase is more in women having pre-existing risk factors for cardiovascular disease. The same risk does not occur in younger menopausal women. Based on the results of WHI study, postmenopausal hormone therapy should also not be prescribed after the age of 65 years for prevention of dementia.

Oestrogen therapy is not recommended for women with a personal history of breast cancer due to the increased risk of breast cancer. Neoplasia of the endometrium may be associated with unopposed oestrogen use. The risk increases with an increased duration of use. The risk increases by 3–6 times after 5 years of use and by 10 times after 10 years of use. Therefore, unopposed oestrogens are no longer used in patients with an intact uterus.

Selective Oestrogen Receptor Modulators

Selective oestrogen receptor modulators (e.g. raloxifene) may prevent osteoporosis, but have no beneficial effects on genitourinary or vasomotor symptoms. These agents show high affinity towards oestrogen receptors and have oestrogen agonist and antagonist properties. These drugs have selective actions on the specific target tissues. New agents are being developed which produce desirable actions without the unwanted side effects.

Raloxifene

This drug does not cause endometrial proliferation, but produces a favourable response in the bones and on lipid profile. While there has been no evidence of reduction in the evidence of wrist or hip fractures, it causes nearly 50% reduction in the incidence of vertebral fractures. The main side effect associated with the use of raloxifene is an increase in the incidence of venous thromboembolism. Raloxifene, therefore, can be considered as a treatment option for prevention of osteoporosis-related spinal fractures. This must be accompanied by periodic evaluation of bone density in the hips. If bone loss occurs, another treatment option can be considered.

Arzoxifene

It is an oestrogen agonist-antagonist similar to raloxifene. While it is an oestrogen agonist in bone and on lipids, it is an oestrogen antagonist in the endometrial and breast tissues.

Ospemifene

Ospemifene is another SERM, which is recently been used for the treatment of vaginal dryness. It is administered in the dosage of 60 mg/day orally for the treatment of vaginal and vulvar atrophy. It is a prescription medication that is similar to oestrogen, but is not oestrogen. In the vaginal tissue, it acts similarly to oestrogen. In the breast tissue, it acts as an oestrogen antagonist. The medication may cause hot flushes as a side effect. This medication may be associated with an increased risk for thromboembolism or endometrial cancer. Further research is required for evaluation of the risk of these complications.

Drugs in Development

There are many newly discovered SERMs (droloxifene, lasofoxifene, ormeloxifene, etc.), which are still under research stages. These drugs have the potential for prevention and treatment of osteoporosis.

Tibolone

This drug is being marketed as postmenopausal hormone therapy in many countries, including the European continent, but not in the US. It is available under the brand names of Livial, Livel and Liviella (in the dosages of 1.25–2.5 mg) and is used for prevention of osteoporosis. The standard dose of tibolone was 2.5 mg, but now with emerging evidence doses as low as 1.25 mg have also been shown to be effective. Tibolone has been shown to have a beneficial effect on treatment of hot flushes and vaginal dryness. The use of this drug has been found to be associated with an increase in libido and an improvement in sexual response. It also has a protective action on the bones. There is no evidence of an increase in adverse effects such as coronary artery disease, risk of venous thromboembolism or risk of breast cancer. It has also not been found to be associated with endometrial proliferation.

Principles of Parathyroids

Parathyroid Glands

Parathyroid glands are four small glands located on the posterior surface of the thyroid gland. Each lobe of the thyroid gland is associated with one superior and one inferior parathyroid gland. Each parathyroid gland comprises of two types of cells: densely packed chief cells, which produce parathyroid hormone (PTH) and the larger oxyphil cells whose function is largely unknown.

Serum Calcium Regulation

Maintenance of a stable concentration of serum calcium is important for sustaining the functioning of all excitable tissues in the body. Severe abnormalities in calcium concentration can cause cardiac arrhythmias due to the role of ca^{2+} ions in action potential. Therefore, it is important to maintain the serum calcium concentration within tight limits. The plasma level of calcium is controlled by the interaction of the gut and kidney responses to the PTH, calcitonin and vitamin D. PTH and vitamin D have been described in details next in the text. Calcitonin released by the C cells of the thyroid acts to reduce calcium and its release may be stimulated by alcohol. Calcitonin is released in response to a high serum concentration of calcium. It acts to lower the serum calcium levels by inhibiting the osteoclast-mediated resorption of the bone. It also has moderate effect on the kidneys, increasing the excretion of calcium and phosphate.

Parathyroid Hormone

Parathyroid hormone is an 84 amino acid-long-protein-chain produced by the parathyroid gland. The first 34 amino acid N-terminal fragment possesses biological activity. PTH binds to G-protein-coupled receptors (on target cell surfaces) and acts on several tissues to raise the plasma calcium levels, resulting in the following:

❏ Increased osteoclastic activity resulting in bone resorption and hence release of calcium

❏ Increased renal tubular resorption of calcium, whereas that of phosphate is reduced. In the kidney, PTH is a potent stimulus for the reabsorption of calcium in the loop of Henle.

❏ Increased renal expression of the enzyme 1-α hydroxylase, which synthesises vitamin 1,25 di (OH) vitD3. This induces synthesis of a calcium binding protein (calbindin-D) in the intestinal mucosa with resultant absorption of calcium.

Parathormone secretion is inversely proportional to blood calcium level. Parathyroid secretion is stimulated by low calcium levels and hypomagnesaemia. On the other hand, its secretion is inhibited in cases of hypercalcaemia. Hypercalcaemia may occur in association with the conditions enumerated in **Table 11.18**. Changes in serum calcium and phosphate levels in association with the various disorders of the bone are summarised in **Table 11.19**.

PTH secretion (secondary hyperparathyroidism) is increased in renal failure due to chronic vitamin D deficiency and hypocalcaemia. Tertiary hyperparathyroidism can ensue.

Role of Calcium Ion in Regulating 1,25-Dihydroxycholecalciferol Levels

Vitamin D (1,25-dihydroxycholecalciferol) is a steroid, predominantly synthesised in the skin from 7-dehydrocho-lesterol in response to the ultraviolet light. Primary food sources of vitamin D include dairy products, fish (salmon, sardines and mackerel), etc. In the UK, cows' milk is gener-ally not a good source of vitamin D because it is not fortified as it may be other countries.

Vitamin D, whether it is ingested or produced in the skin from sunlight, is not synthesised in its active form. Instead, it must undergo two hydroxylation reactions. Cholecalciferol (vitamin D_3) is formed from the ultraviolet rays in the skin from 7-dehydrocholesterol. First hydroxylation reaction occurs in the liver in presence of the enzyme 25-hydroxylase, resulting in the formation of 25-(OH)-D_3. Second hydroxylation reaction is that of α-hydroxylation. This is a rate-limiting limiting step which occurs in the renal tubules and is under the control of PTH. This results in the production of the active compound, 1,25 (OH)$_2$ D_3 **(Fig. 11.16)**. Concentrations of vitamin D are therefore reduced in cases of chronic renal failure. Though α-hydroxylation

TABLE 11.18	Causes of hypercalcaemia

• Hyperparathyroidism
• Vitaminosis D
• Sarcoid
• Addison's disease
• The milk alkali syndrome
• Thyrotoxicosis
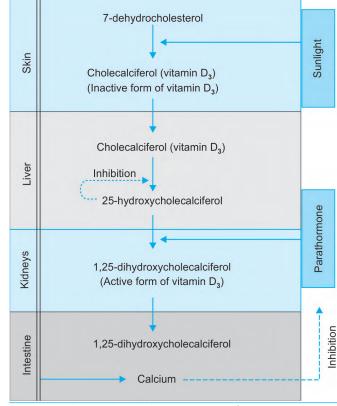
FIG. 11.16: Schematic diagram showing activation of vitamin D

TABLE 11.19	Disorders of the bone and their respective serum biochemistry				
Pathology	Serum calcium levels	Serum phosphate	Alkaline phosphatase	Parathyroid hormone	Comment
Osteoporosis	Normal	Normal	Normal	Normal	Decreased bone mass resulting in an increased risk of fractures
Osteopetrosis	Normal	Normal	Normal	Normal	Marble bone disease causing bones to harden due to which they dissolve and break
Osteomalacia (rickets)	Reduced	Reduced	Increased	Increased	Soft bones usually due to the deficiency of calcium or vitamin D
Osteitis fibrosa cystica	Increased	Reduced	Increased	Increased	Skeletal disorder due to hyperparathyroidism, resulting in bone pain, tenderness, bone fractures and skeletal deformities
Paget's disease of the bone	Normal	Normal	Increased	Normal	Abnormal excessive breakdown of bones followed by disorganised bone formation and remodelling, resulting in weakness, pain, misshapen bones, fractures and arthritis.

commonly occurs in the kidneys, it can also occur within granulomas, thereby resulting in association of sacroidosis with hypercalcaemia.

Vitamin D plays an important role in the metabolism of calcium. In cases of hypocalcaemia, vitamin D helps in increasing the blood calcium levels by increasing the gastrointestinal absorption of calcium. However, it does not cause the resorption of osteoid to release calcium. If the serum calcium levels are normal, vitamin D also aids in the deposition of hydroxyapatite on to the bone collagen. If however, there is hypocalcaemia, vitamin D does not prevent PTH-mediated bone resorption. Vitamin D helps mineralise the bones (provided that normal levels are present), but maintenance of Ca^{2+} by PTH is more important.

Increase in blood calcium levels inhibits the formation of 1,25-dihydroxycholecalciferol by directly suppressing the conversion of 25-hydroxycholecalciferol into 1, 25-dihydroxycholecalciferol. This helps in decreasing the gastrointestinal uptake of calcium and phosphorus.

If there is a primary deficiency of vitamin D rather than calcium in the body, the bones remain unmineralised. This may result in rickets in children or osteomalacia in adults, which differ due to the fusion of growth plates during adolescence. The bones contain adequate amounts of collagen, but this is not hardened by formation of osteoid. If the deficiency of vitamin D is so severe so as to cause hypocalcaemia, secondary hyperparathyroidism may result due to the negative feedback loop.

Hyperparathyroidism

Hypersecretion of PTH is called hyperparathyroidism and is associated with hypercalcaemia. Hyperparathyroidism is of three types: primary hyperparathyroidism, secondary hyperparathyroidism and tertiary hyperparathyroidism.

Primary Hyperparathyroidism

Primary hyperparathyroidism is due to the development of tumour in one or more parathyroid glands. Sometimes, tumour may develop in all the four glands. It is usually a consequence of a single adenoma but may be associated with hyperplasia and rarely carcinoma where PTH concentrations may be particularly high. Primary hyperparathyroidism is associated with hypercalcaemia and hypophosphataemia and there is usually hypercalciuria. Primary hyperparathyroidism does not proceed to tertiary but secondary may proceed to tertiary.

Secondary Hyperparathyroidism

Secondary hyperparathyroidism is due to the physiological compensatory hypertrophy of parathyroid glands, in response to hypocalcaemia which occurs due to the following pathological conditions:

- ❑ Chronic renal failure
- ❑ Vitamin D deficiency
- ❑ Rickets
- ❑ Impairment of 1-alpha-hydroxylation of vitamin D by the kidneys.

Tertiary Hyperparathyroidism

Tertiary hyperparathyroidism is due to hyperplasia of all the parathyroid glands that develops due to chronic secondary hyperparathyroidism.

Hypoparathyroidism

Reduced secretion of PTH is known as hypoparathyroidism. It is associated with hypocalcaemia by reducing the resorption of calcium from the bones. Hypocalcaemia causes neuromuscular hyperexcitability resulting in hypocalcaemic tetany. Tetany occurs when serum calcium levels fall below 6 mg/dL in comparison to the normal values of 9.4 mg/dL. Tetany can be treated with IV calcium gluconate. Sudden complete loss of parathyroid function may lead to skeletal muscle spasms. This is a main feature of tetany. This may be fatal if treatment is not given to raise the blood level of ionized calcium. Treatment can be in the form short-term by slow intravenous injection of calcium ions, e.g. calcium gluconate. This may be treated in the long term by regular doses of vitamin D, which acts by increasing intestinal calcium absorption.

Hypoparathyroidism occurring due to an abnormality of the PTH receptor is termed pseudohypoparathyroidism. Pseudohypoparathyroidism is associated with low serum calcium and high phosphate levels, which is similar to the biochemical abnormalities encountered in hypoparathyroidism. However, the levels of parathyroid hormone are elevated. Hence the term pseudo is used. This makes pseudohypothyroidism different from hypoparathyroidism. Pseudo-pseudohypoparathyroidism is an abnormality where there are typical physical features such as short fourth metacarpal (brachydactyly), but no biochemical abnormality is observed. Differences between these three conditions are summarised in **Table 11.20**.

In DiGeorge's syndrome, there are absent parathyroids and thymus associated with a chromosomal abnormality.

TABLE 11.20	Differences between hypoparathyroidism, pseudohypoparathyroidism, pseudo-pseudohypoparathyroidism					
Condition	**Appearance**	**PTH levels**	**Calcitriol**	**Calcium**	**Phosphates**
Hypoparathyroidism	Normal	Low	Low	Low	High
Pseudohypoparathyroidism	Normal	High	Low	Low	High
Pseudo-pseudohypoparathyroidism	Skeletal defects	Normal	Normal	Normal	Normal

Abbreviation: PTH, parathyroid hormone

Understanding of Adrenal Structure and Function

Adrenal Glands

There are two adrenal glands which are pyramidal in shape, yellowish-brown structures, overlying the superior pole of the kidney **(Fig. 11.17)**. Both the adrenal glands lie anterior to the diaphragm and the one on the left side is posterior to the pancreas. Both the glands are composed of an inner medulla and an outer cortex. Each adrenal gland consists of a cortical portion derived from the coelomic epithelium and a medullary portion originally composed of sympatho-chromaffin tissue. The foetal adrenal cortex represents 80% of the gland which undergoes rapid regression at the time of birth. Lymphatic drainage of the adrenal glands is to the lumbar nodes. The suprarenal vein returns the blood from the medullary venous plexus and on the right side opens into the inferior vena cava, on the left into the renal vein.

The adrenal cortex secretes steroid hormones, which are required for the following: control of body salt and water content, metabolism of carbohydrates and fats and normal sexual functions. Adrenal cortex comprises of three morphologically and functionally distinct regions called the zona glomerulosa (the outer), zona fasciculata (the intermediate) and zona reticularis (the inner). Zona glomerulosa is the outer most layer, which secretes the mineralocorticoid, aldosterone. Zona fasciculata is the intermediate layer and it produces glucocorticoids, most importantly cortisol. The ratio of secreted cortisol to corticosterone is approximately 1:17. Zona reticularis produces a proportion of the body's sex steroids (mainly androgens, which are converted peripherally into dehydro-epiandrosterone, oestrogens and testosterones). Pathway for the synthesis of steroid hormones is described in **Figure 11.18**. After hypophysectomy, the two zones, the intermediate and inner begin to atrophy because secretion from both these zones is under hypothalamic control. Due to the action of angiotensin II on the zona glomerulosa, this zone remains unchanged.

Adrenocorticotrophic Hormone

Adrenocorticotrophic hormone is a polypeptide with a molecular weight of 4,500 daltons. Protein hormones including ACTH are synthesised on ribosomes and stored by the Golgi apparatus in separate vesicles within the same cell. ACTH contains the sequence of alpha-melanocyte-stimulating hormone. The remainder of the molecule (amino acids 18–39) is the corticotrophin-like intermediate lobe peptide (CLIP).

Adrenocorticotrophic hormone can be expressed in numerous tissues besides the pituitary and include the placenta. ACTH concentrations increase during conditions such as stress, disease and during pregnancy.

Secretion of ACTH from the pituitary increases when the median eminence of the hypothalamus is stimulated resulting in the secretion of corticotropin-releasing hormone (CRH), the releasing hormone for ACTH. Production of ACTH or CRH regulates the levels of cortisol and the sex steroids produced by the zona reticularis. ACTH secretion increases when the cortisol blood levels fall. This negative feedback helps to maintain the blood cortisol level. It governs cortisol secretion due to which the cortisol
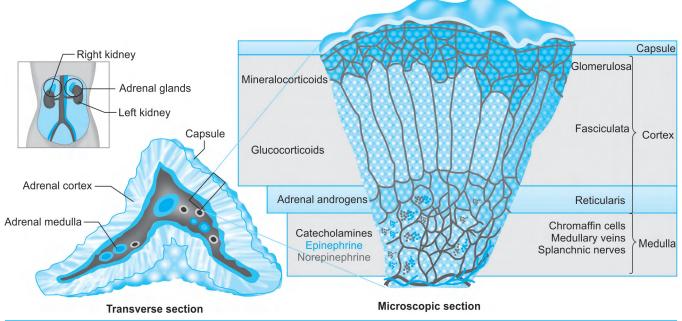
FIG. 11.17: Anatomy of the adrenal glands
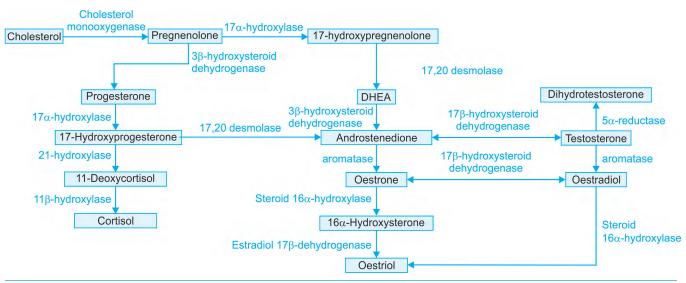
FIG. 11.18: Pathway for the synthesis of steroid hormones produced by the adrenal glands
Abbreviation: DHEA, dehydroepiandrosterone

is secreted maximally in the morning and concentrations are at a nadir during midnight. Through negative feedback, glucocorticoids (not mineralocorticoids—aldosterone) switch off ACTH production. ACTH secretion increases in bursts during the night as the normal hour of wakening approaches. This is part of the circadian rhythm which produces high cortisol levels in the morning. Most forms of stress including severe trauma increase ACTH output by their neural input to the median eminence of the hypothalamus where CRH is formed.

Aldosterone secretion, on the other hand, is mainly regulated by the renin/angiotensin system (secreted by the juxtaglomerular apparatus) and independently by the serum potassium concentrations and not by CRH or ACTH.

Adrenal Hormones

Mineralocorticoids

Aldosterone is the primary mineralocorticoid produced by the adrenal gland. Aldosterone secretion is regulated by four important factors:
1. Increase in potassium ion (K+) concentration in ECF
2. Decrease in sodium ion (Na+) concentration in ECF
3. Decrease in ECF volume (reduced circulating plasma volume, blood loss, hypovolaemia and hypotension)
4. Increased ACTH secretion.

Increase in the concentration of potassium ions is the most effective stimulant for aldosterone secretion. Secretion of aldosterone is entirely regulated by the renin-angiotensin system. On the other hand, secretion of glucocorticoids is exclusively controlled by ACTH and CRH. Though the main action of ACTH is on glucocorticoid secreting cells; it has a slight action on mineralocorticoid secreting cells as

well, thereby having a mild effect in increasing aldosterone secretion.

Aldosterone is the principal mineralocorticoid secreted by the adrenal cortex and acts on the distal convoluted tubules and the collecting ducts. Aldosterone acts via intracellular steroid receptors to increase sodium reabsorption. Aldosterone causes the renal reabsorption of sodium and water, with the loss of potassium into the urine. Its release is inhibited by increased sodium intake/hypertension. Aldosterone is released through Increased plasma volume reduces the secretion of aldosterone, whereas increased plasma osmolality increases ADH secretion. Regulation of the aldosterone secretion is described in **Figure 11.19**. Decrease in sodium ion concentration and ECF volume stimulates aldosterone secretion through renin-angiotensin mechanism. Renin secreted from juxtaglomerular apparatus of kidney acts on angiotensinogen in the plasma and converts it into angiotensin I. Release of renin is an essential component of the renin-angiotensin-aldosterone system (RAAS), which regulates the blood pressure and the blood volume.

The macula densa is a specialised tissue lining the wall of the cortical thick ascending limb at the transition to the distal convoluted tubules. Its role is to monitor blood flow into the capsule, and hence it is involved in blood pressure control. The cells of macula densa act by increasing the release of renin from the juxtaglomerular cells of the afferent and efferent arterioles (main storage sites for renin production) in response to reduced sodium concentration.

The angiotensin converting enzyme (ACE) secreted by the lungs then converts angiotensin I into angiotensin II, which is a vasoconstrictor and an important regulator of the extracellular volume as part of the renin-angiotensin system. ACE causes breakdown of the peptide, bradykinin, a potent vasodilator. Drugs inhibiting ACE (e.g. ramipril,
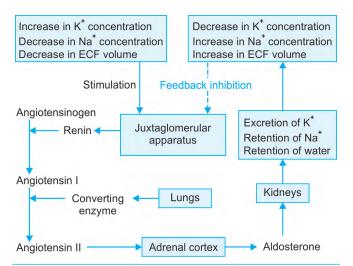
FIG. 11.19: Regulation of the aldosterone secretion

enalapril, etc.) prevent the formation of angiotensin II, thereby inducing a state of relative vasodilatation. Levels of ACE may be raised in numerous pathologies such as primary biliary cirrhosis, sarcoidosis, Gaucher's disease, etc.

Angiotensin II acts on the zona glomerulosa to secrete more aldosterone. Aldosterone in turn, increases the retention of sodium and water and excretion of potassium. This leads to increase in the sodium ion concentration and ECF volume. Eventually, the increased sodium ion concentration and the ECF volume inhibit the juxtaglomerular apparatus and stop the release of renin.

As the progesterone secretion increases in pregnancy, it stimulates the loss of sodium in the urine. This in turn, increases aldosterone production to conserve sodium. The conditions which may be associated with elevated levels of aldosterone are summarised in **Table 11.21**.

Glucocorticoids

Glucocorticoids act mainly on glucose metabolism. Glucocorticoids produced by the zona fasciculata of the adrenal gland include cortisol, corticosterone and cortisone. Glucocorticoids and mineralocorticoids have a similar structure and slightly overlapping functions. While the mineralocorticoids are only concerned with salt and water reabsorption and potassium excretion, glucocorticoids have other metabolic roles as well. Glucocorticoids can also weakly bind to the mineralocorticoid receptors, thereby having some effect on salt and water reabsorption. Some of the actions of cortisol include the following: anti-inflammation, sodium (and indirectly water) reabsorption, insulin antagonism (catabolism), foetal maturation, maintenance of arteriolar tone, reduced muscle mass and increased bone resorption.

Effect on carbohydrate metabolism: Glucocorticoids increase the blood glucose levels by promoting gluconeogenesis in liver from amino acids and by inhibiting the uptake and utilisation of glucose by peripheral cells.

TABLE 11.21	Conditions associated with an elevated levels of aldosterone

- Reduced circulating volumes
- Reduced sodium levels (this is usually rare because the ADH system normally corrects this rapidly by allowing water loss until Na+ levels return to normal).
- Increased potassium levels
- Increased production of angiotensin II (as part of renin-angiotensin-aldosterone system)
- Pregnancy
- Conn's syndrome
- Renal artery stenosis
- Diuretic therapy

Effect on protein metabolism: Glucocorticoids promote the catabolism of proteins, resulting in decrease in cellular proteins, increase in plasma level of amino acids and increased protein content in liver. In hypersecretion of glucocorticoids, there is excess catabolism of proteins, resulting in muscular wasting and negative nitrogen balance.

Effect on fat metabolism: Glucocorticoids cause mobilisation of fatty acids from adipose tissue, they help increase the concentration of fatty acids in blood and increase the utilisation of fat for energy. These hormones mobilise fats and make the fatty acids available for utilisation, by which energy is liberated. This results in the formation of a large amount of ketone bodies. This is known as the ketogenic effect of glucocorticoids. Hypersecretion of glucocorticoids causes an abnormal type of obesity by increasing the deposition of fat in certain areas such as abdomen, chest, face and buttocks.

Sex Steroids

As previously described, zona reticularis produces numerous sex steroids mainly androgens which are converted peripherally into dehydroepiandrosterone, androstenedione, oestrogens and testosterone. In adults, this represents a modest fraction of sex hormone production in comparison with that occurring in the ovaries or the testis. In men, secretion of sex steroids from adrenal glands is of little importance. In women, however, this remains as the major source of androgens and helps in maintaining libido and muscle mass.

Diseases due to the Abnormality of Adrenal Hormones

Congenital Adrenal Hyperplasia

Congenital adrenal hyperplasia (CAH) occurs due to 21-hydroxylase deficiency in over 90% of cases. 21-hydroxylase is one of the enzymes responsible for the conversion of 17-OHP (17-hydroxyprogesterone) to cortisol and aldosterone **(Fig. 11.18)**. Therefore, deficiency of 21-hydroxylase

is associated with the deficiency of both cortisol and aldosterone. At the same time, there are elevated levels of testosterone and 17OHP. Consequently, these metabolites are found in excess in CAH. Due to the deficiency of cortisol, the concentration of ACTH increases. Plasma renin activity may also be elevated. 11-Deoxycorticosterone may be increased in the cases of much rarer 11-hydroxylase deficiency.

Adrenal crisis may occur within a few days of birth. Salt losing adrenal crisis is a well-recognised feature of the classic variety of the disease, with severe hyponatraemia, hypotension and hypoglycaemia apparent from birth onwards. Early treatment (in vivo treatment with steroids for the mother) may prevent virilisation of the female foetus.

Addison's Disease

Addison's disease is a chronic disorder of the adrenal glands associated with the deficiency of both mineralocorticoids and glucocorticoids. This form of adrenal insufficiency must be differentiated from the deficiency of ACTH (resulting in secondary adrenal insufficiency) and deficiency of CRH (resulting in tertiary adrenal insufficiency). Addison's disease is often known as chronic primary adrenocortical insufficiency, which is different from acute primary adrenocortical insufficiency caused by Waterhouse Friderichsen's syndrome (acute adrenal failure caused due to the bleeding into the adrenal glands, most commonly caused by the meningococcus, *N. meningitides*). Despite of these various disorders, Addisonian crisis can occur in all forms of adrenal insufficiency.

Signs and symptoms develop in Addison's disease because of deficiency of both cortisol and aldosterone. Some common signs and symptoms of Addison's disease include pigmentation of skin and mucous membrane due to excess ACTH secretion, induced by cortisol deficiency. ACTH causes pigmentation due to its melanocyte-stimulating action. Other symptoms include muscular weakness, dehydration with loss of sodium, hyperkalaemia, hypotension, decreased cardiac output and decreased workload of the heart, hypoglycaemia, nausea, vomiting, diarrhoea, dehydration, loss of body weight, increased susceptibility to any type of infection, inability to withstand any stress, resulting in Addisonian crisis, etc. Addison's disease is associated with hyperkalaemia, not hypokalaemia.

Most common cause for Addison's disease is atrophy of adrenal cortex due to autoimmune diseases. Other causes of Addison's disease include destruction of glands due to tuberculosis, malignancy, etc. In nearly half of the patients with autoimmune Addison's disease, at least one other autoimmune disorder may be present. These may include disorders such as hypothyroidism, vitiligo, non-toxic goiter, premature menopause, Graves' disease, etc.

Acute Adrenal Insufficiency

Patients with acute adrenocortical insufficiency (Addisonian crisis) may present with symptoms such as weakness, nausea and vomiting, abdominal pain, hypotension, fever, etc. Biochemical findings associated with adreno-cortical insufficiency include electrolyte imbalance (e.g. hyponatraemia, hyperkalaemia, etc.), hypoglycaemia acidosis, etc.

Cushing Syndrome

Cushing syndrome occurs due to the hypersecretion of glucocorticoids, particularly cortisol. It may be either due to pituitary origin or adrenal origin. If it is due to pituitary origin (e.g. pituitary ACTH microadenoma), it is known as Cushing disease. If it is due to adrenal origin (e.g. cortisol secreting adrenal adenoma) or due to ectopic hormone production or iatrogenic glucocorticoid administration, it is called Cushing syndrome. Generally, these two terms are used interchangeably.

Characteristic feature of this disease include the following: there is disproportionate distribution of body fat, resulting in some abnormal features such as moon face, fat accumulation in the chest and abdomen, thin arms and legs in proportion to torso/trunk, buffalo hump due to fat deposit on the back of neck and shoulder, and pot belly. There may be the presence of purple-reddish striae on the abdomen. There occurs thinning of skin and subcutaneous tissues due to protein depletion caused by increased catabolism of proteins. There may be presence of acanthosis (characterised by darkened skin patches in certain areas such as axilla, neck and groin), pigmentation of skin (especially in ACTH dependent type), facial plethora, hirsutism, weakening of muscles because of protein depletion, bone absorption and osteoporosis, increased susceptibility to the occurrence of fractures, hyperglycaemia, glucosuria and adrenal diabetes, hypertension, immunosuppression resulting in an increased susceptibility for infection, poor wound healing, etc.

Cushing syndrome is classified into two types:
1. *ACTH-dependent Cushing syndrome*: This occurs due to hypersecretion of ACTH and the causes may include ectopic ACTH secretion (bronchial carcinoid or bronchogenic carcinoma).
2. *ACTH-independent Cushing syndrome*: In these cases, the secretion of ACTH is normal. The syndrome develops due to abnormal membrane receptors for some peptides like interleukin-1, gonadotropin-releasing hormone and gastric inhibitory polypeptide in the cells of zona fasciculata. The binding of these peptides to the abnormal receptors leads to an increased secretion of glucocorticoids, resulting in the development of Cushing syndrome. Iatrogenic Cushing syndrome developing due to the treatment with exogenous glucocorticoids or due to ectopic hormone production also belongs to this type.

Disorders of Adrenal Medulla

Catecholamines

Adrenaline and noradrenaline are the catecholamines secreted by the adrenal medulla. Adrenaline constitutes

some 80% of this secretion. Both these catecholamines increase the strength of myocardial contraction and cause vasoconstriction of blood vessels in the mucous membranes. Adrenaline has stronger beta agonist effects in comparison to noradrenaline. Adrenaline increases the heart rate when injected intravenously, whereas injection of noradrenaline causes reflex slowing of the heart. Catecholamine release is generated by the following situations:

❑ Stress (i.e. waking).
❑ Sympathetic stimulation (flight, fright, fight response).
❑ During hypoglycaemia (as response to stressful stimuli and counteraction of catecholamines in recruiting glucose).
❑ During illness (myocardial infarction, sepsis) or
❑ Hypotensive episode.
❑ Stimulation of the nerves to the adrenal gland.

Phaeochromocytoma

Phaeochromocytoma is caused by tumour of chromophil cells in adrenal medulla. It is a condition characterised by hypersecretion of catecholamines. Phaeochromocytoma occurs as part of MEN type II. Hypertension (also known as endocrine or secondary hypertension) is a typical symptom associated with phaeochromocytoma. Other features of phaeochromocytoma are enumerated in **Table 11.22**.

Palpitations associated with variable hypertension (postural hypotension) can also occur but are not common. Reflex bradycardia may be observed but classically tachycardia is seen. Though hyperglycaemia is a rare feature, hypoglycaemia is not observed. Hypokalaemia is also a rare occurrence.

Diagnosis: Presence of elevated levels of free metadrenaline/normetadrenaline in the urine is associated with a high sensitivity and specificity for diagnosing phaeochromocytoma. Though phaeochromocytoma can also be associated with an elevated urinary vanillylmandelic acid (VMA)

TABLE 11.22	Symptoms associated with phaeochromocytoma

- Hypertension (also known as endocrine or secondary hypertension)
- Paroxysmal panic attacks
- Fever, sweats
- Anxiety
- Tremor
- Diarrhoea
- Tachycardia
- Chest pain
- Headache
- Hyperglycaemia
- Metabolic disorders
- Nausea and vomiting
- Palpitation
- Weight loss
- Polyuria and glucosuria
- Sweating and flushing
- Weight loss
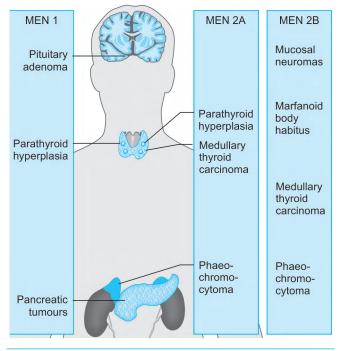
FIG. 11.20: Characteristic features of different types of MEN syndrome

levels, this can also occur in insulin dependent diabetic patients experiencing hypoglycaemia. It can also occur in association with foods such as vanilla and some drugs—beta blockers and ganglion blockers.

Miscellaneous Hormones

Multiple Endocrine Neoplasia

Multiple endocrine neoplasia (MEN) encompasses several distinct syndromes representing the characteristics related to the tumours of various endocrine glands, each having its own characteristic pattern. Some of these tumours may be malignant, while some may be benign. MEN syndromes usually show an autosomal dominant mode of inheritance. The characteristic features of various types of MEN syndrome are described in **Figure 11.20** and are also described next in the text.

MEN II syndrome: Also known as Wermer's syndrome, this syndrome is characterised by the presence of pancreatic tumours such as gastrinomas, insulinomas, VIPomas, glucagonomas, pituitary adenomas, angiofibromas, lipomas, parathyroid hyperplasia, etc.

MEN 2A syndrome: Also known as the Sipple syndrome; MEN 2A syndrome is associated with parathyroid hyperplasia, medullary thyroid carcinoma and phaeochromocytoma.

MEN 2B syndrome: This is associated with medullary thyroid carcinoma, phaeochromocytomas, marfanoid body habitus and mucosal neuromas.

Choose the Single Best Answer (SBA)

Q 1. Which of the following hormone acts on cartilage and liver to release IGF-1?

A. Growth hormone
B. Prolactin
C. Somatostatin
D. TSH
E. ACTH

Q 2. In males, which of the following hormone facilitates the generation of spermatozoa?

A. GnRH
B. Somatostatin
C. Dopamine
D. LH
E. FSH

Q 3. Which of the following statement regarding leptin is correct?

A. It is the toxin causing tissue necrosis in leprosy
B. It is a neurotransmitter produced by the hypothalamus leading to immature and reckless behaviour
C. It is a growth factor produced by the pubertal testicle with its main effect on Sertoli cells
D. It is a hormone produced by fat
E. It is implicated in obesity associated with polycystic ovary syndrome

Q 4. Which of the following is a recognised cause of hirsutism?

A. Anorexia nervosa
B. Cushing's syndrome
C. Haemochromatosis
D. Hypothyroidism
E. Sheehan's syndrome (postpartum pituitary necrosis)

Q 5. Which of the following hormones stimulate the gonadotrophs of the anterior pituitary?

A. Follicle stimulating hormone (FSH)
B. Gonadotropin releasing hormone (GnRH)
C. Inhibin
D. Luteinising hormone (LH)
E. Relaxin

Q 6. Which of the following is true regarding the pituitary gland?

A. Is located close to the floor of the third ventricle
B. Lies in the pituitary fossa which is located in the ethmoid bone
C. Has neural connections with the pineal body
D. Bounded laterally by the diaphragma sellae
E. Bounded superiorly by the cavernous sinus

Q 7. Which of the following is a recognised cause of loss of body hair?

A. Anorexia nervosa
B. Haemochromatosis
C. Hypothyroidism
D. Sheehan's syndrome (postpartum pituitary necrosis)
E. All the above

Q 8. Which of the following hormone is secreted by the anterior pituitary?

A. Antidiuretic hormone
B. Luteinising hormone
C. Oxytocin
D. Human placental lactogen
E. Gonadotropin releasing hormone

Q 9. Which of the following statement is correct regarding growth hormone is correct?

A. Is a glycoprotein hormone
B. Secretion is markedly increased during pregnancy
C. Is responsible for growth in children and in the foetus
D. Secretion is increased by hypoglycaemia
E. Pygmies have low levels of GH

Q 10. Growth hormone is responsible for which of the following metabolic processes?

A. Increased glycogen deposition
B. Decreased blood glucose concentration
C. Increased catabolism of protein and amino acids
D. All the above
E. None of the above

Q 11. Which of the following is not a recognised feature of acromegaly?

A. Intestinal polyposis
B. Splenomegaly
C. Hypocalciuria
D. Palpable peripheral nerves
E. Proximal myopathy

Q 12. Which of the following is true regarding prolactin?

A. Release is inhibited by metoclopramide
B. Release is inhibited by thyrotropin-releasing hormone
C. Release is increased by suckling
D. Has lower blood levels during normal sleeping hours
E. Is necessary for milk ejection

Q 13. Which of the following inhibits prolactin secretion?

A. Corticotropin-releasing factor
B. Dopamine
C. Gonadotropin releasing hormone (GnRH)
D. Growth hormone (GH)
E. Thyroid releasing hormone (TRH)

1. A   2. E   3. D   4. B   5. B   6. A   7. E   8. B   9. D   10. A   11. C   12. C   13. B

Q 14. Hyperprolactinaemia with hypogonadism is found in which of the following conditions?
A. Chromophobe adenoma of the pituitary
B. Addison's disease
C. Hyperthyroidism
D. Sheehan's syndrome
E. None of the above

Q 15. Which of the following is not a clinical feature of a prolactinoma in a 30-year-old female?
A. Amenorrhoea
B. Bitemporal hemianopia
C. Hirsutism
D. Reduced bone mineral density (BMD)
E. TSH deficiency

Q 16. Which of the following statement concerning prolactin-secreting pituitary tumours is correct?
A. A homonymous hemianopia is typically seen with a suprasellar extension
B. Are usually macroadenomas at presentation
C. They tend to cause higher prolactin levels than idiopathic hyperprolactinaemia
D. Suprasellar extension is an indication for immediate surgical intervention
E. Shrink in size during pregnancy

Q 17. Which of the following endocrine findings characteristically occur in hyperprolactinaemia?
A. Elevated FSH levels
B. Impaired cortisol response to the insulin tolerance test
C. Impaired short term pulsatility of LH secretion
D. All the above
E. None of the above

Q 18. Hyperprolactinaemia may be associated with which of the following?
A. Chlorpropamide therapy
B. LHRH analogue therapy
C. Chronic renal failure
D. Testosterone therapy
E. None of the above

Q 19. Secretion of the thyroid-stimulating hormone (TSH) is not increased in which of the following situations?
A. After partial removal of the thyroid gland
B. In infants born without a thyroid gland
C. In starvation
D. When the diet is deficient in iodine.
E. None of the above

Q 20. Possible consequences of hypothyroidism include which of the following?
A. A subnormal body core temperature
B. A tendency to fall asleep less frequently
C. Increased body hair (hirsutism)
D. Moist hands and feet
E. Prominent eyeballs

Q 21. Removal of the thyroid gland (without replacement therapy) leads to which of the following?
A. Reduced Blood TSH level
B. Reduced Blood cholesterol level
C. Increased Blood glucose level during an oral glucose tolerance test
D. Increased response time for tendon reflexes
E. Increased tremors of the fingers

Q 22. Which of the following occurs when secretory activity in the thyroid gland increases?
A. The gland takes up iodide from the blood at a slower rate
B. Its follicles enlarge and fill up with colloid
C. The follicular cells become more cuboidal
D. The follicular cells ingest colloid by endocytosis
E. The blood level of thyrotropin (TSH) increases

Q 23. Regarding hypothyroidism, which of the following statement is correct?
A. Carpal tunnel syndrome is caused by amyloid deposits within the flexor retinaculum
B. It may be associated with a macrocytic or microcytic anaemia
C. It is the end result of sub-acute thyroiditis
D. It is often associated with pretibial myxoedema
E. None of the above

Q 24. Which of the following is true regarding the thyroid hormones?
A. Does not require specific receptors in target organ cells
B. Consists of T4, T3 and TSH
C. Exerts its effect chiefly through T4 to which T3 is converted
D. Is not extensively bound to plasma proteins
E. Is stored in the thyroid gland on thyroglobulin

Q 25. Which of the following is true concerning thyroid hormones?
A. D-thyroxine is more active than L-thyroxine
B. Starvation causes plasma T3 to rise
C. Thyroid binding globulin (TBG) is increased in pregnancy
D. Triiodothyronine (T3) is converted in the tissues to thyroxine (T4)
E. T4 acts more rapidly than T3

Q 26. Which of the following is true regarding thyroid hormones?
A. Begins to be stored in the foetal thyroid at 8 weeks gestation
B. Daily iodine requirement doubles in pregnancy
C. Daily iodine requirement is about 120 micrograms per day
D. Free concentrations of thyroxine rises in normal pregnancy
E. None of the above

14. A   15. C   16. C   17. C   18. C   19. C   20. A   21. D   22. D   23. B   24. E   25. C   26. C

Q 27. Which of the following is true regarding thyroxine?
A. In blood, is mostly bound to thyroglobulin
B. Is inactivated by de-iodination to thyronine
C. It is a steroid hormone
D. It is composed of two tyrosine residues
E. Synthesis involves reduction of iodide

Q 28. With regards to radioactive iodine (RAI) therapy, which of the following is correct?
A. Breast feeding should be avoided for 8 weeks after administration
B. It is an effective therapy for thyrotoxicosis due to thyroiditis
C. It is contraindicated in patients with Graves' ophthalmopathy
D. It produces hyperthyroidism in the majority of patients treated
E. It should be avoided in women of child bearing age

Q 29. Which of the following hormone is secreted within the posterior lobe of the human pituitary gland?
A. Oxytocin
B. Thyroid-stimulating hormone
C. Luteinising hormone
D. Adrenocorticotropin
E. Prolactin.

Q 30. Which of the following is true regarding oxytocin?
A. Is synthesised in the supraoptic nucleus of the hypothalamus
B. Causes milk ejection
C. Is a large polypeptide
D. Is released directly into the circulation from its site of production
E. Relaxes the uterus

Q 31. Which of the following is true regarding oxytocin?
A. Causes relaxation of myoepithelial cells in mammary glands
B. Has 10% of the antidiuretic activity of ADH (antidiuretic hormone)
C. Is synthesised in the posterior pituitary gland
D. Lowers the threshold for depolarisation of the uterine smooth muscle
E. The sensitivity of the uterus to oxytocin decreases as pregnancy progresses

Q 32. Which of the following statement regarding vasopressin is not correct?
A. Is a nonapeptide hormone
B. Secretion is inhibited by alcohol
C. May elevate arterial blood pressure by direct action on the arteriolar smooth muscles
D. Secretion is increased by angiotensin II
E. Deficiency of vasopressin causes diabetes insipidus

Q 33. Type 1 insulin dependent diabetes mellitus is associated with which of the following?
A. About a 1:3 positive family history
B. Decreased islet cell antibodies with increasing time from diagnosis

C. An 80% concordance among identical twins
D. Low plasma glucagon levels
E. Insulin resistance

Q 34. Which of the following statement about testicular hormones is true?
A. Inhibin increases plasma follicle stimulating hormone levels
B. Testosterone in plasma is partly bound to albumin
C. Testosterone is excreted in urine as 17-ketosteroids
D. None of the above
E. All the above

Q 35. Which of the following is true concerning sex hormone binding globulin?
A. Has a lesser affinity than albumin for testosterone
B. Is the main binding protein for aldosterone
C. Is the main binding protein for progesterone
D. Levels are decreased during oestrogen therapy
E. Levels are increased in pregnancy

Q 36. Which of the following is true regarding androgens?
A. Are formed in the Leydig cells of the testis
B. Are produced in the ovary
C. Are secreted by the female adrenal cortex
D. All the above
E. None of the above

Q 37. Which of the following is true concerning androgens in normal pre-menopausal women?
A. 95% of circulating testosterone is derived from peripheral conversion of androstenedione
B. About 50% of circulating testosterone is bound to sex hormone binding globulin (SHBG)
C. DHEAS (dehydroepiandrosterone sulphate) is derived almost exclusively from the adrenal glands
D. Testosterone promotes the synthesis of sex hormone binding globulin
E. None of the above

Q 38. The human testis does not secrete which of the following hormones?
A. Testosterone
B. Androstenedione
C. Luteinising hormone
D. Oestradiol
E. Inhibin

Q 39. Which of the following is not true regarding hirsutism in females?
A. Is a presenting symptom of hyperprolactinaemia
B. Can be caused by phenytoin
C. May be due to congenital adrenal hyperplasia
D. When idiopathic, is associated with normal plasma testosterone levels
E. When ovarian in origin, is most commonly due to polycystic ovarian syndrome

27. D   28. A   29. A   30. B   31. D   32. C   33. B   34. B   35. E   36. D   37. C   38. C   39. A

Q 40. Which of the following statements regarding puberty is true?

A. Early pubertal development is associated with an overall increased height
B. The first sign of puberty in females is the development of breast tissues
C. First sign of puberty in males is the appearance of facial hair
D. The Ferriman-Gallwey scoring system may be used for describing various stages of breast development
E. Initial menstrual cycles are anovulatory in nature

Q 41. Which of the following changes do not occur in the vagina at the time of puberty?

A. A decrease in the vaginal pH
B. Colonisation by Doderlein's bacilli
C. Exfoliation of superficial cells with pyknotic nuclei
D. Glycogenation of the epithelium
E. The appearance of glands in the epithelium

Q 42. Which of the following is true concerning the adolescent growth spurt?

A. Has a peak velocity which is higher in girls than in boys
B. Has an onset after the age of 16 years in Turner's syndrome
C. In boys, commences before the testes begin to increase in size
D. In girls, has a peak velocity that coincides with menarche
E. Is considered to be abnormally early if in girls it occurs before 8 years

Q 43. True precocious puberty can be caused by which of the following?

A. Encephalitis
B. Fragile X syndrome
C. Congenital adrenal hyperplasia
D. McCune-Albright syndrome
E. Polycystic ovary syndrome

Q 44. Which of the following is a cause of delayed puberty?

A. An imperforate hymen
B. Congenital absence of the uterus
C. Klinefelter's syndrome
D. All the above
E. None of the above

Q 45. Which of the following patients with delayed puberty requires diagnostic evaluation?

A. A 15-year-old girl who has no features of pubertal development
B. A 13-year old boy who does not experience testicular enlargement
C. A 13-year-old girl with a 4 month history of irregular menstrual cycles and normal development of other sexual characteristics
D. A 15-year-old girl with primary amenorrhea who has normal secondary sexual characteristics.
E. None of the above

Q 46. Which of the following statement regarding delayed puberty is correct?

A. Is more common in obese children
B. In a short girl is an indication to perform chromosome analysis
C. Manifest by amenorrhoea in a 17-year-old girl and associated with an elevated serum testosterone level suggests an androgen resistance disorder
D. Presenting as immature genitalia and short stature in a 15-year-old boy indicates pathological gonadotropin deficiency
E. None of the above

Q 47. Which of the following is not produced by the endometrium during the normal menstrual cycle?

A. Coagulation factor VIII
B. Endothelins
C. Higher levels of prostaglandins in the secretory phase than in the proliferative phase
D. Increased fibrinolytic activators in the late secretory phase
E. Platelet activating factor (PAF)

Q 48. Which of the following is true regarding the normal menstrual cycle?

A. Basal vacuolation is the earliest histological sign of ovulation
B. Endometrial cystic hyperplasia is a sign of ovulation
C. The endometrium is completely shed in a normal menstrual cycle
D. The endometrium is supplied with blood by the spiral arteries
E. The endometrium regenerates from the superficial layers

Q 49. During the menstrual cycle, which of the following is true regarding ovulation?

A. Follows a mid-cycle FSH surge
B. Occurs 2 days after the peak of LH
C. Occurs 14 days before the onset of menstrual flow
D. Occurs after follicles have ripened in the ovary
E. Occurs immediately before the LH surge

Q 50. Which of the following plasma hormones peak at the middle of a normal menstrual cycle?

A. 17-alpha-hydroxyprogesterone
B. Follicle stimulation hormone
C. Inhibin
D. Oestradiol
E. Testosterone

Q 51. Which of the following is true regarding the menarche?

A. Is followed by the growth spurt
B. Is preceded by the onset of breast development
C. Occurs earlier in girls below normal weight
D. Occurs later in blind girls
E. Usually follows an ovulatory cycle

40. B   41. E   42. E   43. A   44. C   45. A   46. B   47. A   48. D   49. C   50. B   51. B

Q 52. Which of the following is correct regarding the follicle-stimulating hormone (FSH)?

A. FSH is a glycoprotein composed of two subunits, alpha and gamma
B. The alpha chain is composed of 12 amino acids
C. The receptors for FSH are serpentine receptors coupled to adenyl cyclase
D. Is produced by the posterior pituitary gland
E. It is responsible for the final maturation of the ovarian follicles, and oestrogen secretion from them

Q 53. Raised levels of follicle stimulating hormone are found in which of the following situations?

A. Gonadal dysgenesis
B. Postmenopausal females
C. Turner's syndrome
D. All the above
E. None of the above

Q 54. Which of the following is true regarding oestrogens?

A. Are aromatised to testosterone
B. Are mainly secreted by the ovary as oestrone
C. Small amounts are excreted in the urine
D. Are the dominant gonadal hormone at puberty
E. Cannot be detected in the blood of postmenopausal women

Q 55. Which of the following is not true regarding oestrogens?

A. Are responsible for the initial growth of the breast at puberty
B. Reduces FSH release from the pituitary
C. Inhibin reduces the secretion of FSH
D. Stimulate release of prolactin from the pituitary
E. Stimulate sebaceous gland activity

Q 56. Which of the following regarding oestrogen is correct?

A. Is responsible for the growth of pubic hair in the female at puberty
B. Causes an increase in blood and urine levels of calcium
C. Is inactive during pregnancy
D. Causes deposition of glycogen in vaginal epithelium
E. Reduces the number of progesterone receptors in the endometrium

Q 57. Oestrogens cause which of the following?

A. Hypertrophy of the uterus
B. Increased motility of the fallopian tube
C. Proliferation of vaginal epithelium
D. All of the above
E. None of the above

Q 58. Which of the following is true regarding inhibin?

A. Stimulates the release of follicle stimulating hormone
B. Is a steroid
C. Is produced by the ovarian follicle
D. Is released in pulses
E. Is structurally identical to relaxin

Q 59. In the menstrual cycle, which of the following is true regarding ovulation?

A. Follows a prolactin surge
B. May be inhibited by emotional disturbance
C. Is associated with maximal libido
D. Occurs when progesterone secretion is at its maximum
E. Will only occur as a reflex response to orgasm

Q 60. Which of the following is not a cause for anovulation?

A. Hyperprolactinaemia
B. Obesity
C. Polycystic ovarian syndrome
D. Propranolol
E. Weight loss

Q 61. Which of the following is true regarding the anovulatory cycles?

A. Are associated with dysmenorrhoea
B. Are a risk factor for ovarian cancer
C. May be due to polycystic ovary syndrome
D. Are a contraindication to the combined oral contraceptive
E. Are common after the menopause

Q 62. Which of the following stimulates androgen production from theca cells?

A. Follicle stimulating hormone (FSH)
B. Gonadotropin releasing hormone (GnRH)
C. Inhibin
D. Luteinising hormone (LH)
E. Relaxin

Q 63. Which of the following cell types are present in the human corpus luteum?

A. Endothelial cells
B. Fibroblasts
C. Pericytes
D. Macrophages
E. All the above

Q 64. Which of the following is not true concerning progesterone?

A. Falling levels of progesterone in the last days of the cycle trigger menstruation
B. Progesterone is mainly secreted by the corpus luteum
C. Progesterone reduces uterine sensitivity to prostaglandins
D. Trophoblastic HCG causes the corpus luteum to persist in the early weeks of pregnancy
E. Trophoblastic progesterone production takes over from the corpus luteum at about 8 weeks

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52. C   53. D   54. D   55. E   56. D   57. D   58. C   59. B   60. D   61. C   62. D   63. E   64. C

**Q 65. Which of the following is not true concerning lute-
inising hormone (LH)?**

A. Has a half-life in the circulation of approximately 12
hours

B. In the male stimulates testosterone production

C. Is released in pulses

D. Is required for normal corpus luteum survival

E. Plasma concentrations are increased in post-
menopausal women

**Q 66. Which of the following is true regarding the
menopause?**

A. The length of the menstrual cycle increases, beginning
2–8 years before the onset of menopause

B. The duration of the luteal phase is the major
determinant of cycle length

C. Menstrual cycle changes prior to the menopause are
marked by elevated follicle stimulating hormone
(FSH) and inhibin

D. Eventually there is a 10- to 20-fold increase in mean
serum levels of LH

E. Luteinising hormone (LH) levels are higher than FSH
because FSH is cleared from the blood so much
faster

**Q 67. Which of the following is not true regarding the
menopause?**

A. The postmenopausal ovary in most women secretes
more testosterone than the premenopausal ovary

B. The circulating oestradiol level after menopause is
approximately 10–20 pg/mL (40–70 pmol/L)

C. 'Hot flushes' are more frequent and severe during
the night

D. The flush persists for longer than 5 years in as many
as 25–50% of women

E. The flush coincides with a surge in follicle-stimulating
hormone (FSH)

**Q 68. Which of the following statements is not correct once
the menopause has occurred?**

A. Any vaginal bleeding should be investigated by
performing a D&C.

B. Carcinoma of the uterine body should be suspected
if there is postmenopausal bleeding.

C. There is a normalisation of LH/FSH 5 years after the
menopause

D. There is an increase in vaginal acidity

E. Treatment with oestrogen is often beneficial

**Q 69. Which of the following is a selective oestrogen
receptor modulator (SERM)?**

A. Tamoxifen

B. Tibolone

C. Cyproterone acetate

D. Metformin

E. None of the above

**Q 70. Which of the following factor does not predispose
to osteoporosis?**

A. A sedentary lifestyle

B. Corticosteroid treatment

C. Heparin therapy

D. Early menarche

E. Obesity

Q 71. Which of the following is true regarding tamoxifen?

A. Can cause osteoporosis

B. Is a pure antagonist of the oestrogen receptor

C. Is an antagonist of the progesterone receptor

D. Is associated with hypokalaemia

E. Increases the risk of endometrial carcinoma

Q 72. Which of the following is true regarding anastrozole?

A. Is associated with endometrial hyperplasia

B. Is contraindicated in patients with ischaemic heart
disease

C. It is a selective oestrogen receptor modulator

D. It is associated with improved outcomes in early
breast cancer compared with tamoxifen

E. It is associated with relative hypoadrenalism

**Q 73. Regarding parathyroid hormone, which of the
following statement is correct?**

A. Due to the high circulating serum levels of parathor-
mone, it can be detected by radio-immunoassay

B. It is composed of 184 amino acids

C. PTH stimulates activity of 1-α-hydroxylase in the liver

D. Production is increased by a drop in serum phosphate

E. Brachydactyly may be present in pseudohypopar-
athyroidism

**Q 74. Which of the following is correct regarding primary
hyperparathyroidism?**

A. Is associated with bone resorption by PTH to restore
depressed serum calcium levels to normal

B. Is associated with hypocalciuria due to elevated PTH
levels

C. Is usually caused by an adenoma of a single
parathyroid gland

D. Progresses to tertiary hyperparathyroidism with time

E. PTH is secreted in a pulsatile manner from the
posterior pituitary and acts through PTH receptors
on parathyroid cell membranes

**Q 75. Which of the following is not true regarding calcium
metabolism and its control?**

A. Calcitonin secretion may be stimulated by alcohol

B. Cholecalciferol is 25-hydroxylated in the liver

C. In plasma, calcium binding to protein is pH dependent

D. The average daily absorption of calcium from the
diet is 10 mmol

E. The major stimulant to parathyroid hormone secretion
is a fall in the plasma unionised calcium concentration

65. D   66. A   67. E   68. C   69. A   70. D   71. E   72. D   73. E   74. C   75. E

Q 76. **Which of the following is true regarding parathyroid hormone (PTH) secretion?**

A. Causes bone formation
B. Is influenced by plasma magnesium levels
C. Is not influenced by plasma potassium levels
D. Is increased in chronic renal disease
E. Raises plasma phosphate levels

Q 77. **Which of the following statements is true regarding aldosterone?**

A. Acts on specific cell surface receptors.
B. Is produced in the zona reticularis of the adrenal cortex
C. Secretion decreases when sodium intake is reduced
D. Secretion is increased following haematemesis
E. Secretion is increased in phaeochromocytoma

Q 78. **Which of the following concerning aldosterone is not correct?**

A. In the kidney, acts on the distal convoluted tubule
B. Is the principal mineralocorticoid secreted by the adrenal cortex
C. Production is increased in normal pregnancy
D. Secretion is entirely regulated by the renin-angiotensin system
E. Secretion is increased by a low potassium intake

Q 79. **Which of the following statement about the adrenal glands is not correct?**

A. Lymphatic drainage is to the lumbar nodes
B. The adrenal medulla is derived from mesoderm
C. The left adrenal gland lies behind the pancreas
D. The right adrenal vein drains directly into the inferior vena cava
E. The adrenal glands lie anterior to the diaphragm

Q 80. **Autoimmune Addison's disease is associated with which of the following?**

A. Premature ovarian failure
B. Hypothyroidism
C. Vitiligo
D. Sjögren's syndrome
E. All the above

Q 81. **Acute adrenal insufficiency causes which of the following?**

A. Alkalosis
B. Hyperglycaemia
C. Hypernatraemia
D. Hypertension
E. Hyperkalaemia

Q 82. **Myxoedema is not associated with which of the following features?**

A. Ataxic gait
B. Dementia
C. Diplopia
D. Fits
E. None of the above

Q 83. **Which of the following hormone is synthesised by the kidney?**

A. Aldosterone
B. Angiotensin I
C. Angiotensin II
D. Erythropoietin
E. 25 dihydroxycholecalciferol

Q 84. **Multiple endocrine neoplasia (MEN) type II does not consist of which of the following?**

A. Medullary carcinoma of the thyroid
B. Parathyroid hyperplasia
C. Phaeochromocytoma
D. Pituitary tumour
E. None of the above

Q 85. **De Quervain's (sub-acute) thyroiditis is not commonly associated with which of the following?**

A. Elevated ESR
B. Tender goiter
C. Dense fibrosis involving the thyroid and adjacent structures
D. Thyrotoxicosis
E. Transiently increased thyroid uptake of 99m-technetium

Q 86. **The action of noradrenaline released at sympathetic nerve endings is terminated by which of the following?**

A. Enzymatic decarboxylation
B. Enzymatic inactivation by catechol-O-methyl transferase
C. Its removal by the circulating blood
D. Oxidative deamination by monoamine oxidase
E. Re-uptake of noradrenaline by the axonal terminals

Q 87. **Glucocorticoid injections lead to an increase in which of the following?**

A. Lymph gland size
B. Fibroblastic activity
C. Anabolic activity in muscle
D. Membrane stability in mast cell and lysosomes
E. None of the above

Q 88. **Which of the following statement regarding cortisol is not correct?**

A. Is bound in the plasma to an alpha globulin
B. Is inactivated in the liver and excreted in the bile
C. Injections lead to a rise in arterial pressure
D. Inhibits release of ACTH from the anterior pituitary gland
E. Is released with a circadian variation so that cortisol blood levels peak in the morning

76. E   77. D   78. E   79. B   80. E   81. E   82. C   83. D   84. D   85. C   86. E   87. D   88. B

Q 89. Which of the following statement regarding the plasma level of adrenocorticotrophic hormone (ACTH) is not correct?
A. Is normally maximal around the time of awakening
B. Is regulated mainly by the hypothalamic circadian rhythm
C. Shows exaggerated circadian fluctuations with an adrenal tumour
D. Is raised in the presence of complete adrenal failure
E. Is reduced in patients on long-term high dosage glucocorticoids

Q 90. Which of the following statement regarding vitamin D is not correct?
A. Increases the intestinal absorption of calcium
B. Is essential for normal calcification of bones in childhood
C. Requires hepatic modification for activation
D. Cannot be synthesised in the body
E. Deficiency may result in hyperparathyroidism

Q 91. Secretion of parathormone is usually increased in which of the following situation?
A. In patients with chronic renal failure
B. In people taking excessive amounts of vitamin D
C. In patients with anterior pituitary tumours secreting excessive amounts of its hormones
D. When blood phosphate levels fall
E. When plasma protein levels fall

Q 92. The physiological action of oestradiol depends upon which of the following?
A. Active transport of the hormone into cells
B. Alteration of gene expression
C. Binding to an intracellular receptor
D. All of the above
E. None of the above

Q 93. Which of the following statement regarding insulin is correct?
A. Stimulates uptake of free fatty acids by adipose tissue
B. Secretion tends to lower the plasma potassium level by promoting its uptake by the cells
C. Facilitates entry of glucose into skeletal muscle
D. Facilitates entry of amino acids into skeletal muscle
E. All the above

Q 94. Severe uncontrolled diabetes mellitus does not lead to which of the following?
A. Increased H$^+$ ion concentration in body fluids
B. Increased Plasma K$^+$ concentration
C. Increased urinary specific gravity and osmolality
D. Increased blood volume
E. Reduced arterial PCO$_2$

Q 95. Which of the following increases the risk of tetany?
A. Sudden rises in plasma bicarbonate
B. Sudden rises in plasma magnesium
C. Removal of the anterior pituitary gland
D. The onset of respiratory failure
E. The onset of renal failure

Q 96. An adrenal medullary tumour (phaeochromocytoma) does not cause an increase in which of the following?
A. Systolic blood pressure which may be transient or constant
B. Tremor of the extended hand
C. Basal metabolic rate
D. Diastolic arterial pressure which does not respond to alpha adrenoceptor blocking drugs
E. Urinary catecholamines

Q 97. Which of the following statement is true concerning insulin?
A. Has equal biological activity to C-peptide
B. Has a half-life of eight minutes when given intravenously
C. Is secreted mainly as proinsulin
D. More than 80% is degraded by the liver and kidney
E. Release from the pancreatic beta cell is stimulated by biguanides

Q 98. Actions of insulin does not include which of the following?
A. Cellular uptake of amino acids
B. Cellular uptake of potassium
C. Entry of glucose into adipose tissue
D. Entry of glucose into neurons
E. None of the above

Q 99. Which of the following is true regarding insulin?
A. Interacts with the nuclear membrane
B. Causes an increased glucose-protein transport on the endoplasmic reticulum
C. Acts via a similar mechanism as steroid receptors
D. Can be detected in the lymph
E. Is synthesised in the alpha cells of islets of Langerhans

Q 100. Which of the following mechanism is involved in the mediation of insulin action?
A. Adenylate cyclase activation
B. Cell membrane receptor interaction
C. Hormone receptor DNA binding
D. All the above
E. None of the above

Q 101. Which of the following does not stimulate insulin secretion?
A. Arginine
B. Gastrin
C. Glucagon
D. Glibenclamide
E. Bendroflumethiazide

89. C 90. D 91. A 92. D 93. E 94. D 95. A 96. D 97. D 98. D 99. D 100. B 101. E

Q 102. Which of the following stimulates insulin secretion?
A. Noradrenaline (Norepinephrine)
B. Somatostatin
C. Propranolol
D. Bendroflumethiazide
E. Glibenclamide

Q 103. Which of the following regarding adrenocortico-trophic hormone is not correct?
A. Is a polypeptide hormone.
B. Has a molecular weight of 4,500 daltons
C. Is the main hormone controlling aldosterone secretion
D. Secretion is increased in congenital adrenal hyperplasia
E. Contains the sequence of alpha-melanocyte-stimulating hormone

Q 104. Which of the following steroid is produced by the ovary?
A. Cortisol
B. Dehydroepiandrosterone sulphate (DHEAS)
C. Oestriol
D. Testosterone
E. None of the above

Q 105. Adrenaline secretion from the adrenal glands does not increase which of the following?
A. Blood glucose level
B. Blood free fatty acid level
C. Blood flow to skeletal muscle
D. Blood flow to the splanchnic area
E. Release of renin in the kidneys

Q 106. Which of the following increases after the surgical removal of the pituitary gland?
A. Plasma osmolality
B. Menstrual frequency
C. Axillary hair
D. Sexual desire (libido)
E. Breast size

Q 107. Which of the following is not true regarding peptide hormones?
A. Act via receptors found within the cell nucleus
B. Are structurally dissimilar to steroid hormones
C. Are used in the treatment of prostate cancer
D. Can be synthesised through recombinant DNA techniques
E. Have paracrine effects

Q 108. Which of the following is true regarding gonadotro-pin releasing hormone?
A. Inhibits prolactin secretion
B. Is not present in the blood of postmenopausal women
C. Is not released in a pulsatile fashion
D. Is responsible for increasing the synthesis and release of LH
E. Is secreted by the anterior pituitary

Q 109. Which of the following statement is true regarding sex hormone binding globulin?
A. Is depressed by increasing plasma oestrogens
B. Is depressed in hyperthyroidism
C. Is increased by increasing levels of plasma androgens
D. Is increased in pregnancy
E. Is produced by the ovary

Q 110. Short stature is seen in adults who in childhood suffered from all the following except?
A. Chronic malnutrition
B. Castration
C. Premature puberty
D. Thyroid deficiency
E. Adrenal deficiency

Q 111. Which of the following is not likely to occur in the adrenal failure?
A. Fall in an extracellular fluid volume
B. Decrease in the total red cell mass
C. Decline in the sodium: potassium ratio in plasma
D. Fall in the arterial blood pressure
E. Increase in the blood urea levels

Q 112. Which of the following is a recognised feature of a chromophobe adenoma of the pituitary gland?
A. Erectile impotence
B. Glycosuria
C. Hyperprolactinaemia
D. Hypertension
E. None of the above

Q 113. Which of the following is a characteristic finding in anorexia nervosa?
A. A decrease in cortisol levels
B. An increase in LH levels
C. Impaired glucose tolerance
D. Raised androgen levels
E. Hyperkalaemia

Q 114. Which of the following is true regarding the poste-rior pituitary gland?
A. Function is inhibited by alcohol
B. Releases decapeptide hormones
C. Secretes IGF1
D. Synthesises somatomedins
E. None of the above

Q 115. Which of the following statement regarding thyroid gland is correct?
A. The amount of free (non-protein-bound) thyroxine (T4) in the plasma is four times less than the amount of free tri-iodothyronine (T3)
B. Thyroid hormone decreases the dissociation of oxygen from haemoglobin by increasing red cell 2,3-diphosphoglycerate (DPG)
C. Over-treatment of the pregnant mother with antithyroid drugs may result in cretinism
D. Reverse T3 is decreased in severe illness
E. Thyroid-stimulating hormone (TSH) levels are increased during normal pregnancy

102. E   103. C   104. D   105. D   106. A   107. A   108. D   109. D   110. B   111. D   112. C   113. C   114. A   115. C

Q 116. **Which of the following is not true regarding genetic deficiency of thyroid hormone production (dyshormonogenesis)?**

    A. It is associated with a diminished uptake of radioactive iodine

    B. Is best treated with iodine in mild cases

    C. Leads to the formation of a goiter

    D. May be associated with congenital nerve deafness

    E. May produce no signs or symptoms of thyroid deficiency

Q 117. **Which of the following is true concerning metformin?**

    A. Stimulates the release of insulin from the beta cells of the pancreas

    B. Therapy causes hypoglycaemic events

    C. Therapy is associated with lactic acidosis

    D. Therapy reduces prolactin concentrations in polycystic ovarian syndrome

    E. None of the above

Q 118. **Attacks of hypoglycaemia are not a recognised complication of which of the following?**

    A. Fructosaemia

    B. Galactosaemia

    C. Gaucher's disease

    D. Glucose-6-phosphate dehydrogenase deficiency

    E. Von Gierke's disease

Q 119. **Which of the following statement regarding diabetic ketoacidosis is not correct?**

    A. A normal plasma potassium level does not exclude significant potassium deficiency

    B. Amylase levels may be raised in the absence of pancreatitis

    C. Leucocytosis is common and does not confirm infection

    D. Plasma glucose may be low

    E. Urinary stick testing for ketones may be negative

Q 120. **Which of the following hormones is involved with the "rescue" of the corpus luteum?**

    A. Prolactin

    B. Chorionic gonadotropin (hCG)

    C. Oestradiol

    D. Oxytocin

    E. Progesterone

Q 121. **Which of the following is true regarding hypoparathyroidism?**

    A. May cause short stature, candidiasis and impaired nail and dental development in children

    B. May be a feature of Crohn's disease

    C. When due to an abnormality of the PTH receptor is termed pseudo-pseudohypoparathyroidism

    D. Biochemically is characterised by increased calcium, increased phosphate and normal alkaline phosphatase

    E. Positive Chvostek's sign can be treated with oral calcium

Q 122. **Pseudohypoparathyroidism is associated with which of the following?**

    A. Absent thymus

    B. DiGeorge's syndrome

    C. Hypothyroidism

    D. Low serum calcium, high serum phosphate, high PTH

    E. Low serum calcium, high serum phosphate, normal PTH

Q 123. **Which of the following is true regarding glycosylated haemoglobin?**

    A. Is absent in the plasma of people without diabetes mellitus

    B. Is increased in diabetic patient with concurrent sickle cell disease

    C. The levels of HbA1c are a good index of glucose-induced renal dysfunction in diabetics

    D. When measured as HbA1c in plasma gives more accurate retrospective estimates of blood sugar levels than other glycosylated products

    E. None of the above

Q 124. **Which of the following is true regarding hyperosmolar non-ketotic diabetic coma?**

    A. It is unusual in the elderly

    B. It is rarely associated with a blood sugar level greater than 30 mmol/L

    C. Causes hyperventilation

    D. May cause focal neurological signs

    E. It rarely predisposes to thrombosis

Q 125. **Which of the following is true concerning leptin?**

    A. Acts on the hypothalamus

    B. Concentrations are positively correlated with body mass index

    C. Concentrations are usually higher in females than males

    D. Deficiency is associated with obesity

    E. All the above

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116. B   117. C   118. D   119. D   120. B   121. A   122. D   123. C   124. D   125. E

Pharmacology

Clinical Pharmacy

Drug Interactions

Metabolism of drugs is likely to affect the duration and potency of the effect of various drugs. Drugs may be converted more polar metabolites, thereby facilitating their excretion. This is frequently catalysed by several enzymes. A large number of drugs are metabolised by hepatic phase I and II reactions.

Phase I reactions: These occur in the endoplasmic reticulum and involve formation of more polar metabolites of the original drug. These reactions may be catalysed by cytochrome P450 and include reactions such as oxidation, hydrolysis, reduction, cyclisation or decyclisation. These polar metabolites may be directly excreted in the urine or may be converted into simpler, less toxic molecules by phase II reactions.

Phase II reactions: These reactions occur in the cytoplasm and involve conjugation with molecules such as sulphates, glucuronides, glutathione, amino acid, etc. This results in the formation of less toxic and more easily excitable metabolites. The metabolism of various drugs can be affected by enzyme induction or inhibition. Drugs inhibiting and inducing hepatic microsomal enzymes are enumerated in **Table 12.1**.

Effect of various drugs on the effectiveness of the oral contraceptive pill (OCP): Hepatic enzyme inducing antiepileptic drugs lower OCP hormone levels by approximately 40%. These include carbamazepine, rifampicin and phenytoin. Also antibiotics like ampicillin, tetracyclines and metronidazole may reduce the efficacy of the OCP.

After taking tetracyclines for some time (as in acne) the bowel flora adapt and normal efficacy of combined oral contraceptive pill (COCP) is reinstated; however, acute courses will result in reduced efficacy.

TABLE 12.1	Drugs inhibiting and inducing hepatic microsomal enzymes	
Enzyme inducers	**Enzyme inhibitors**	
• Griseofulvin	• Metronidazole	
• Phenytoin	• Ciprofloxacin	
• Phenobarbitone	• Fluconazole	
• Rifampicin	• Erythromycin	
• Carbamazepine	• Ethanol (acute)	
• Ethanol (chronic)	• Cimetidine	
• Barbiturates	• Amiodarone	
	• Ketoconazole	

In contrast, valproic acid and gabapentin do not interfere with the effectiveness of OCPs. The pill is contraindicated if someone is on warfarin due to the increased risk of venous thrombosis but has no effect on warfarin.

Drug Combinations

The following drug combinations must be avoided to prevent development of adverse reactions:
❑ Combination of sildenafil and isosorbide mononitrate combined may cause a profound hypotension.
❑ Combination of statin therapy and fibrates are sometimes used in cases of hyperlipidaemia. However this combination may be associated with an increased risk of myositis.
❑ Metronidazole produces an antabuse/disulphiram effect associated with nausea/vomiting and flushing when used in combination with alcohol due to accumulation of metabolites.

Adverse Drug Reactions

The reporting of adverse drug reactions (ADRs) to the Medicines Control Agency is a voluntary process but all side effects noted with newer drugs denoted with a black triangle in the British National Formulary (BNF) should be reported.

Even ADRs with OTC therapies should be reported to ensure adequate monitoring and potentially ascertain whether the manufacturing of certain brands is appropriate.

The report should also contain drugs that the patient has taken (including OTCs) within the last 3 months to establish potential interactions.

Adrenergic Agents

Beta-sympathomimetic Drugs

Beta-sympathomimetic drugs in therapeutic doses cause a direct inotropic and chronotropic effect on the heart. There is little or no effect on the mean blood pressure because the increase in blood pressure resulting from increased heart rate and contractility is counteracted by the decrease in total peripheral resistance due to vasodilation in blood vessels perfusing skeletal muscle. Sympathomimetic amines include ephedrine, amphetamine and isoprenaline. Histamine, on the other hand, is a vasoactive amine.

Side effects: Arrhythmias can occur in large doses. Tachycardia is commonly associated with their use. Beta sympathomimetic drugs can have the following adverse effects, vasodilation, bronchial relaxation, uterine relaxation, intestinal and genitourinary wall relaxation, cardiac stimulation, renin release, glycogenolysis, gluconeogenesis, lipolysis, etc.

Clonidine

Clonidine is an alpha adrenergic receptor agonist (not antagonist), which acts centrally by stimulating presynaptic alpha-2 receptors, causing suppression of catecholamine release by a negative feedback mechanism.

It has been used as an antihypertensive drug, but it also has analgesic and sedative properties. When administered preoperatively it is associated with a reduction in the minimal alveolar concentration (MAC) of volatile anaesthetic agents.

Side effects: Side effects of clonidine include sedation, dry mouth, urinary retention and depression. It should be withdrawn slowly, as sudden withdrawal can cause a severe hypertensive crisis.

Beta Blockers

Propranolol

Propranolol is a non-selective beta-adrenoceptor antagonist (both beta 1 and 2) with no intrinsic sympathomimetic action (ISA). It has a large volume of distribution. Its elimination half-life is relatively short at 4 hours. It is used in the treatment of supraventricular tachycardias (SVTs), e.g. atrial fibrillation, as rate limitation. Through antagonism of the beta-1 receptors, propranolol has negatively chronotropic effects on the heart causing reduced myocardial oxygen consumption and hence is an effective treatment in angina. Propranolol unlike the more selective beta blockers nebivolol and metoprolol, is relatively unselective in its action on the beta-1 and -2 receptors on the heart and bronchi.

Side effects: It is lipid soluble and therefore readily crosses the blood brain barrier, leading to sedation, nightmares and depression. Its effect on the bronchial beta receptors is to induce bronchoconstriction.

Hypnotics, Sedatives and Anxiolytics

Some of the commonly used benzodiazepines are as follows:

Long-acting hypnotics: These include drugs such as nitrazepam, flurazepam, etc. They are commonly used for early morning insomnia, but may cause hangover.

Short-acting hypnotics: These include drugs such as oxazepam, temazepam and triazolam.

Benzodiazepines which act as anxiolytics include diazepam, chlordiazepoxide, lorazepam, etc.

Almost all benzodiazepines are able to cross the placenta. Therefore, they should be avoided in late pregnancy. Their effects can be potentiated by alcohol. Since they are addictive, their withdrawal following long-term use should be gradual. Short acting benzodiazepines are preferable during pregnancy.

Diazepam

Diazepam is a benzodiazepine and acts as a hypnotic, amnesic and anticonvulsant through agonism at the cerebral gamma-aminobutyric acid (GABA) receptors. It increases the chloride ion conductance of the neuronal membrane. It may cause respiratory depression and these effects can be antagonised by flumazenil.

Antipsychotic Drugs

These drugs are also known as neuroleptics. These drugs are useful in all types of functional psychosis, especially schizophrenia. They do not impair consciousness. They act by blocking dopamine receptors. Therefore, the two most important side-effects produced by these drugs are hyperprolactinaemia and extrapyramidal side effects (e.g. Parkinsonism, acute muscular dystonias, akathisia, malignant neuroleptic syndrome, tardive dyskinesia, etc.). They may also produce anticholinergic side effects such as dry mouth, constipation, blurring of vision, etc. These drugs are all capable of crossing the placenta. The classification of antipsychotic drugs is described in **Table 12.2**.

TABLE 12.2	Classification of antipsychotic drugs

- *Phenothiazines:* Chlorpromazine, triflupromazine, thioridazine, trifluoperazine, fluphenazine, etc.
- *Butyrophenones:* Haloperidol, trifluperidol, penfluridol
- *Thioxanthenes:* Flupenthixol
- *Other heterocyclics:* Pimozide, loxapine
- *Atypical antipsychotics:* Clozapine, aripiprazole, risperidone, ziprasidone, olanzapine, etc.

Antidepressant Drugs

❑ *Monoamine oxidase inhibitors (MAOI)*: These may include drugs such as phenelzine, iproniazid, etc. These drugs elevate the mood by inhibiting the enzyme MAO (Monoamine oxidase), which is a mitochondrial enzyme involved in the oxidative deamination of biogenic amines. These drugs greatly potentiate the pressor effects of tyramine present in certain food stuffs (e.g. cheese, broad bean pods, meat, yeast extract, etc.) resulting in hypertensive crisis, cerebrovascular accidents, etc. This reaction can be treated by intravenous (IV) injection of a rapidly-acting α blocker, e.g. phentolamine.

❑ *Reversible inhibitors of MAO-A (RIMAs)*: These drugs (e.g. moclobemide, clorgyline, etc.) have fewer side effects. Potentiation of pressor response to ingested amines is minor, and therefore, dietary restrictions are not required. It also lacks the anticholinergic, sedative, cognitive, psychomotor and cardiovascular adverse effects related to the typical tricyclic antidepressants.

❑ *Tricyclic antidepressants (TCAs)*: Some of the tricyclic antidepressants block the uptake of both noradrenaline and 5-hydroxytryptamine (e.g. imipramine, amitriptyline, trimipramine, doxepin, dothiepin, clomipramine) whereas others predominantly block the reuptake of noradrenaline (e.g. desipramine, nortriptyline, amoxapine, reboxetine, etc.). These drugs are effective in the treatment of moderate and severe degrees of depression. Tricyclic antidepressants prevent noradrenaline re-uptake by the nerve cells, so they do not potentiate the pressor effects of tyramine.

Side effects: The main side effects of tricyclic antidepressants are the anticholinergic side effects such as dry mouth, constipation, bad taste, epigastric distress, urinary retention, blurred vision, palpitation, etc. Tricyclic antidepressants can cause convulsions mainly in children. Sudden death due to arrhythmias and heart block may occasionally follow the use of tricyclic antidepressants, particularly amitriptyline. Tricyclic antidepressants have central and peripheral antimuscarinic actions due to which it can lead to urine retention and constipation. Anaesthetics given during imipramine therapy may increase the risk of arrhythmias and hypotension. Side effects such as sedation, mental confusion and weakness, can especially occur with amitriptyline. Amitriptyline can also cause galactorrhoea.

Tricyclic antidepressants may cause tachycardia and irritability in the neonate especially if taken in the third trimester (especially imipramine). These drugs have a relatively low safety margin. They are hazardous in overdose and fatalities are common. Due to the presence of numerous side effects with tricyclic antidepressants, drugs such as SSRIs, SNRIs and atypical antidepressants have presently become the first-line drugs for treatment of depression.

❑ *Selective serotonin reuptake inhibitors (SSRIs)*: These include drugs like fluoxetine, fluvoxamine, paroxetine, sertraline, citalopram, escitalopram and dapoxetine. These drugs have fewer side effects in comparison to the tricyclic antidepressants. The therapeutic efficacy of SSRIs is similar to that of tricyclic antidepressants, but without the cholinergic side effects. Use of fluoxetine in children is not recommended, as its safety and efficacy have not been established. Fluoxetine can also be a useful treatment for premature ejaculation.

❑ *Serotonin and noradrenaline reuptake inhibitors (SNRIs)*: These include drugs such as venlafaxine and duloxetine.

❑ *Atypical antidepressants*: These include drugs such as trazodone, mianserin, mirtazapine, bupropion, tianeptine, amineptine, etc.

Analgesics

The commonly used analgesic drugs are NSAIDs (non-steroidal anti-inflammatory drugs). These drugs inhibit renal prostaglandin synthesis and may result in sodium retention, reduced renal blood flow and renal failure. NSAIDs are particularly avoided during the third trimester due to the theoretical risk of premature closure of ductus arteriosus and unproven concerns of teratogenicity.

Narcotic analgesics are another class of commonly used class of analgesic drugs. Classification of narcotic analgesics is described in **Table 12.3**. All the centrally acting opioid drugs are capable of crossing the placenta. The foetal effects of the narcotic analgesics (e.g. respiratory depression, etc.) can be reversed by administration of naloxone. Pethidine is the most commonly used opioid. Following IV administration, it reaches the foetus within 2 minutes. Pethidine is used in labour to block the sympathetic response to pain. Respiratory depression can occur in the neonate if delivered between 1 to 3 hours after intramuscular (IM) administration of the drugs. Methadone is not used during pregnancy because it has a long elimination half-life of 23 hours in the foetus. Fentanyl is widely used in epidural

TABLE 12.3	Classification of narcotic (opioid) analgesic drugs

- *Natural opium alkaloids*: Morphine, codeine
- *Semisynthetic opiates*: Diacetylmorphine (Heroin), pholcodine, ethylmorphine, hydromorphone, oxymorphone, hydrocodone, etc.
- *Synthetic opioids*: Pethidine (Meperidine), fentanyl, methadone, dextropropoxyphene, tramadol.

block and spinal anaesthesia. Codeine is extensively used analgesic drug during pregnancy and is safe during pregnancy and lactation.

Side effects: The narcotic analgesic drugs are commonly associated with side effects such as nausea, vomiting, sedation, constipation, respiratory depression, blurring of vision due pupillary constriction, urinary urgency and retention, etc.

Morphine

Morphine is commonly used for severe pain. Morphine does not cause direct myocardial depression, although it may cause bradycardia. It can cause hypotension by decreasing the systemic vascular resistance which is due, in part, to histamine release. The histamine release may also cause bronchospasm. The production of antidiuretic hormone is also increased by morphine.

Infants of opiate-abusing mothers: Infants of opiate-abusing mothers are more likely to be small for gestational age. It is uncertain whether this is due to a direct effect of the drug or, more likely, due to compromised maternal nutrition associated with the chaotic lifestyle. Opiate withdrawal in the infants leads to an increased metabolic rate. The incidence of SIDS is significantly higher in infants of opiate abusers.

Nalorphine

Nalorphine is an opioid agonist-antagonist, which is equally potent with morphine as an analgesic but is not clinically useful due to a high incidence of dysphoria. It can displace opioid agonists from μ receptors, helping to reverse respiratory depression. However, it is not effective at reversing respiratory depression due to barbiturates/benzodiazepines.

Drugs Interfering with Glucose Metabolism

Drugs which can interfere with glucose metabolism, thereby resulting in hypoglycaemia or hyperglycaemia are listed in **Table 12.4**.

Oral Hypoglycaemic Agents

Oral hypoglycaemic drugs help in lowering blood glucose levels and are effective orally. These drugs include the sulphonylurea class of drugs (e.g. chlorpropamide or glibenclamide), meglitinide/phenylalanine analogues or the biguanide class of drugs (e.g. metformin).

Biguanides

Effects of biguanides are mostly mediated by improving insulin resistance. Metformin is a biguanide, which acts by improving insulin sensitivity through better hepatic and muscular glucose utilisation. Metformin reduces elevated blood glucose levels in diabetics and improves insulin sensitivity. Therefore, it does not cause hypoglycaemia. Biguanides only work in the presence of endogenous insulin, so there must be some residual islet cell function. Therapy with metformin also helps improve the rate of conception in cases of polycystic ovarian syndrome. PCOS is usually associated with increased insulin resistance. Metformin therapy may help improve the rate of conception, oligomenorrhoea and hirsutism.

Side effects: Though side effects with metformin are frequent, they are generally not serious. Some commonly occurring side effects include abdominal pain, anorexia, bloating, nausea, metallic taste, mild diarrhoea, tiredness, etc. which tend to subside with time. These agents may also cause SIADH. Metformin does not cause renal impairment but should be used with caution in patients with renal impairment because of risk of lactic acidosis. It should not be used in subjects whose creatinine clearance is above 130 mmol/L or heart failure.

Sulphonylureas

Sulphonylurea therapy, such as glibenclamide, etc. stimulates insulin secretion from the beta cells through the opening of potassium channels. Sulphonylureas do cross the placenta and may result in foetal beta-cell stimulation. Though the incidence of side effects with sulphonylureas is quite low, hypoglycaemia is the most common side effect associated with its use, which may be occasionally severe and rarely fatal. Tolbutamide carries lowest risk due to its low potency and short duration of action.

Chlorpropamide is a drug belonging to the sulphonylurea class of hypoglycaemic drugs used for treating type 2 diabetes. Chlorpropamide inhibits liver enzymes and stimulates antidiuretic hormone (ADH). It may rarely cause mild syndrome of inappropriate antidiuretic hormone.

Diuretics

Loop Diuretics

They are potent diuretics, also known as high ceiling diuretics and include bumetanide, furosemide (frusemide) and torsemide. They are more potent than thiazides. The loop diuretics inhibit reabsorption of sodium and water from the ascending limb of the loop of Henle, where they

TABLE 12.4	Drugs interfering with glucose metabolism
Drugs causing hyperglycaemia	**Drugs causing hypoglycaemia**
• Corticosteroids • Thiazide diuretics • Glucagon	• Ethanol • Aspirin • Gliclazide • Atenolol

inhibit Na$^+$-K$^+$-2Cl$^-$ cotransport. The side effects are similar to that of thiazide diuretics, but much more in severity. It can also cause hyperuricaemia.

Thiazide Diuretics

Metolazone and bendroflumethiazide are thiazide diuretics. These are medium efficacy diuretics with primary site of action in the cortical diluting segment or the early distal tubule. They act by inhibiting the Na$^+$-Cl$^-$ symport. It can cause hypokalaemia, hypochloremic alkalosis, hyperuricaemia and hyperglycaemia.

Potassium Sparing Diuretics

Potassium sparing diuretics are aldosterone antagonists (e.g. aldosterone) and renal epithelial Na$^+$ channel inhibitors (e.g. triamterene and amiloride), which indirectly conserve K$^+$ while inducing mild natriuresis. These drugs act on the distal convoluted tubule through various mechanisms, inhibiting the loss of potassium in exchange for sodium. These drugs can cause potassium retention and oestrogenic side effects. It can also be used in primary and secondary hyperaldosteronism.

Osmotic Diuretics

These include drugs such as urea, mannitol, etc. They act by expanding the blood volume. They may be useful in cases of cerebral oedema.

Drugs for Inducing and Inhibiting Vomiting

Antiemetic Agents

These are drugs used to prevent or suppress vomiting. Various drugs used as antiemetic agents are enumerated in **Table 12.5**.

Metoclopramide

Metoclopramide causes increased gastrointestinal motility, increases sphincter tone and also has central antiemetic actions mediated through dopaminergic receptors. It acts on central dopaminergic receptors, thereby decreasing gastric acid secretion and gastric fluid pH. Actions of metoclopramide are mediated through the antagonism of the dopaminergic receptors. Since metoclopramide has a central phenothiazine-like effect (sedative drug) with gastric emptying, it particularly used before anaesthesia. However, it may cause extrapyramidal side effects.

TABLE 12.5	Classification of antiemetic agents

1. *Anticholinergics*
   – Hyoscine
   – Dicyclomine
2. *H1 antihistaminics*
   – Promethazine
   – Diphenhydramine
   – Dimenhydrinate
   – Doxylamine
   – Meclozine (Meclizine)
   – Cinnarizine
3. *Neuroleptics (D2 blockers)*
   – Chlorpromazine
   – Perphenazine
   – Triflupromazine
   – Prochlorperazine
   – Haloperidol
   – Droperidol
4. *Prokinetic drugs*
   – Metoclopramide
   – Domperidone
   – Cisapride
5. *5-HT3 antagonists*
   – Ondansetron
   – Granisetron
6. *Adjuvant antiemetics*
   – Dexamethasone
   – Benzodiazepines
   – Chlorpropamide
   – Cyclizine.

Domperidone

Domperidone is a dopamine antagonist that does not easily cross the blood brain barrier and therefore is associated with reduced incidence of adverse effects such as dystonia and extra-pyramidal effects. It stimulates gastric peristalsis. It may be used to help/induce lactation.

It is used for the prevention of nausea and vomiting in Parkinson's patients treated with apomorphine. However, it needs to be administered at least 3 days before apomorphine is given.

Ondansetron

Ondansetron is a 5-hydroxytryptamine antagonist and is a particularly effective anti-emetic.

Emetic Drugs

These are drugs used to evoke vomiting. Treatment with these drugs is indicated to induce emesis in treatment of certain poisonings. Induction of emesis should only be considered if gastric lavage is inadvisable or refused. Some of the emetic drugs are described as follows:

❏ *Apomorphine*: It is a semisynthetic derivative of morphine; acts as a dopaminergic agonist on the chemoreceptor trigger zone (CTZ), the vomiting centre in the brain),

thereby inducing vomiting. It is injected via IM or subcutaneous (SC) routes.

❑ *Ipecacuanha*: The dried root of Cephaelis ipecacuanha contains emetine and is used in form of syrup ipecac for inducing vomiting. It is less dependable than parenteral apomorphine and takes 15 minutes or more to produce the effect. It acts by irritating gastric mucosa as well as through CTZ.

Contraindications

All emetics are contraindicated in the following situations:
❑ Corrosive (acid, alkali) poisoning
❑ Central nervous system (CNS) stimulant drug poisoning
❑ Kerosene (petroleum) poisoning
❑ Unconscious patient
❑ Morphine or phenothiazine poisoning

Drugs Acting on the Uterus

Tocolytic Agents

Tocolytic drugs, which inhibit the uterine contractions, include the following:
❑ Glyceryl trinitrate (GTN)
❑ Alcohol
❑ Magnesium sulphate
❑ Ritodrine
❑ Salbutamol
❑ Nifedipine, and
❑ Non-steroidal anti-inflammatory drugs (NSAIDs).

Progesterone in high concentrations also has some tocolytic activity and promotes the relaxant effects of more conventional tocolytics.

Oxytocin

Oxytocin is a nonapeptide, released by the posterior pituitary in the body. Therefore, it is regarded as an oligopeptide because it contains fewer than 10 amino acids. It is synthesised within the nerve cell bodies in supraoptic and paraventricular nuclei of hypothalamus and stored in the posterior pituitary. It is transported down the axon and stored in the nerve endings within the neurohypophysis. Stimuli such as coital activity, parturition, suckling, etc. help in the release of oxytocin molecule. Oxytocin has a similar chemical structure to antidiuretic hormone (ADH), and their physiological effects share similarity to some degree. The sensitivity of uterine muscle to oxytocin increases in late pregnancy. The synthetic form of oxytocin (syntocinon or pitocin) used as a drug is a decapeptide. Oxytocin can exert its effect on the uterus and breast as follows:

Uterus: Oxytocin, which has uterotonic action, helps in increasing the force and frequency of uterine contractions. In the full-term gravid uterus, oxytocin causes physiological uterine contractions, i.e. the contraction of upper uterine segment and retraction of the lower segment. With low doses, full relaxation occurs in between the uterine contractions. Basal tone increases only with high doses of oxytocin. Non-pregnant uterus and that during early pregnancy is rather resistant to oxytocin; sensitivity increases progressively in the third trimester, with a sharp increase occurring near term. The sensitivity quickly falls during the puerperium.

Breasts: In the breast tissues, oxytocin contracts the myoepi-thelial cells of mammary alveoli, thereby forcing the milk into the bigger milk sinusoids, resulting in the "milk ejection reflex" or the "let-down reflex". This reflex is initiated by suckling so that the ejected milk may be easily sucked by the infant.

Mode of Administration

Being a peptide molecule, oxytocin is inactive orally and is most commonly administered by IV route, rarely by intranasal/intrabuccal spray. Oxytocin can also be administered by IM/SC routes. However, these routes are not commonly used because the response through these routes may be erratic and the dose cannot be titrated to the response.

Indications

Postpartum Haemorrhage

Oxytocin (syntocinon) is an effective first-line treatment for PPH. Oxytocin 10–20 IU/500 mL of Ringer's lactate or normal saline may be administered by IV infusion for an immediate response, especially in hypertensive women in whom ergometrine is contraindicated. It is infused at a rate of 125 mL/hour (60 drops/minute) over 4 hours. As much as 500 mL can be infused over 10 minutes without complications. For a sustained effect, continuous infusion of oxytocin is usually preferred. In cases of circulatory collapse, 10 units may be administered intramyometrially. Oxytocin acts by forcefully contracting the uterine muscle, which compresses the blood vessels passing through its mesh work to arrest haemorrhage from the inner surface exposed by placental separation.

Active Management of the Third Stage of Labour

It is used for the active management of third stage of labour. Oxytocin is recommended as the first-line drug in active management of third stage of labour due to its short half-life and good intensity of action. Also, its action can be quickly terminated and it does not cause contraction of the lower segment.

Induction of Labour

It may be required to induce the labour in cases of post-maturity, pre-eclampsia, gestational diabetes, erythroblastosis, ruptured membranes or placental insufficiency. At term, the dose of syntocinon required to induce labour is 0.5–15 mU/min.

Oxytocin titration technique: For this purpose, oxytocin is administered via slow IV infusion wherein 5 IU of oxytocin is diluted in 500 mL of glucose or normal saline solution (10 mIU/mL). Infusion is started at a low rate (1–2 mIU/minute) and progressively accelerated at an interval of 20–30 minutes according to response (1–2 mIU/minute). When the optimal response is achieved, i.e. there are about three uterine contractions in 10 minutes, with each uterine contraction being sustained for about 45 seconds; the particular oxytocin concentration which was being used is continued. The oxytocin infusion is described in terms of milliunits/minute. The oxytocin infusion rate can either be manually regulated by counting the number of drops per minute or the other option is to use an oxytocin infusion pump, which automatically controls the infusion rate.

Continuous monitoring: While inducing the patient with oxytocin infusion, uterine contractions, foetal heart rate and any other complications must be closely monitored after every 5–10 minutes. The drug is discontinued when the uterine contractions become strong enough. The oxytocin dose can be increased in the range of 1–32 mIU/minute. Majority of patients respond to the dose of 16 mIU/minute or less. This dose rate can be attained by adding 2 IU of oxytocin to 500 mL of Ringer's lactate solution, with a drop rate of 60 drops/minute (where 15 drops = 1 mL). There is no upper limit to the permitted dose. If the uterus still remains inert at the oxytocin dosage of 100 mIU/minute, it may be wise to consider using prostaglandins for stimulation of the uterus.

Uterine Inertia

In cases where the uterine contractions are not strong enough and labour is not progressing satisfactorily, uterine contractions can be augmented by IV administration of oxytocin. Oxytocin, however, must not be used for accelerating the labour, which is progressing normally on its own. Oxytocin is the drug of choice for inducing and augmenting labour and is usually preferred over ergometrine/PGs for the following reasons:
❏ Intensity of oxytocin action can be controlled and be quickly terminated due to its short half-life and slow IV infusion.
❏ When used in low concentrations, oxytocin allows normal relaxation in between uterine contractions. Therefore, the foetal oxygenation is not compromised.
❏ Since the lower uterine segment is not contracted, foetal descent is not affected.
❏ Uterine contractions are consistently augmented.

Breast Engorgement

Oxytocin is effective in cases where the breast engorgement occurs in the woman due to inefficient milk ejection reflex. Oxytocin is administered by an intranasal spray few minutes before suckling. It does not increase milk production, rather just causes milk ejection.

Adverse Effects

❏ *Strong uterine contractions*: Injudicious use of oxytocin during labour can result in strong uterine contractions. This may force the foetal presenting part through incompletely dilated birth canal, resulting in harmful effects such as maternal and foetal soft tissue injury, rupture of uterus, foetal asphyxia and death.
❏ *Tachysystole/uterine hyperstimulation*: Injudicious use of oxytocin can result in continuous uterine contractions or strong uterine contractions. Tachysystole can be associated with a persistent pattern of more than five uterine contractions in 10 minutes, with each contraction lasting for 2 minutes (or more) or contractions of normal duration occurring within 1 minute of each other, there being no resting tone between contractions.
❏ *Maternal cardiovascular side effects*: Administration of oxytocin can cause certain side effects related to the cardiovascular system in the mother. These can include side effects such as increase in the heart rate, systemic venous return and cardiac output, cardiac arrhythmias, premature ventricular contractions, etc.
❏ *Water intoxication*: This occurs due to its antidiuretic hormone like action when administered in high doses (30–40 mIU/minute) along with IV fluids, especially in conditions such as pre-eclampsia and renal insufficiency. Water intoxication may manifest in the form of symptoms of hyponatraemia such as confusion, coma, convulsions, congestive cardiac failure and death.
❏ *Hypotension*: Bolus IV injection should be avoided in patients with PPH where patient is hypovolemic or in patients with heart disease because of the risk of development of hypotension. Occasionally, oxytocin may also produce anginal pain.
❏ *Foetal side effects*: It can result in foetal side effects such as bradycardia, neonatal jaundice, low APGAR score, etc.

Contraindications

Absolute Contraindications

The administration of oxytocin is contraindicated in the following situations:
❏ Grand multipara (risk of uterine rupture)
❏ Vaginal delivery is contraindicated (e.g. obstructed labour)
❏ Evidence of intrapartum foetal distress
❏ Pregnant women with underlying cardiac disease (to avoid the occurrence of fluid overload)
❏ Previous history of anaphylactic shock.

Relative Contraindications

❑ Previous uterine scar
❑ Vertex not fixed in the pelvis
❑ Unfavourable cervix
❑ Breech presentation
❑ Hydramnios
❑ Multiple pregnancy

Methergine

Ergotamine is an alkaloid isolated from a fungus *Claviceps purpurea*, which commonly occurs in cereals like rye, wheat, etc. It is known to act as a serotonin, dopaminergic and α-adrenergic agonist. Methergine is methylergometrine maleate, a semisynthetic derivative of ergometrine/methergine is available in the form of 1 mL ampoules. Though the true mechanism of action is not clearly understood, both methylergonovine (methergine) and ergometrine are oxytocics which cause generalised smooth muscle contraction. As a result, the upper and lower segments of the uterus contract tetanically and pass into a state of spasm without any relaxation in between. It produces a prolonged tonic uterine contraction with superimposed rapid clonic contractions. Ergometrine has a greater effect on the uterus at term than in early pregnancy. Ergometrine takes about 7 minutes to act on the uterus.

Indications

❑ Prophylaxis and treatment of severe atonic PPH
❑ Active management of third stage of labour
❑ Following LSCS/hysterectomy, to facilitate uterine contractions.

Route of Administration

It can be administered through IM/IV or oral routes. Ergometrine (ergonovine) is available in ampoules of 0.25 mg and 0.5 mg and tablets of 0.5 mg and 1 mg. Methergine is available in ampoules of 0.2 mg and tablets of 0.5 mg and 1 mg.

Dosage

Methergine is administered in the dose of 0.2 mg intramuscularly or intravenously stat for controlling atonic PPH as well as following the delivery of anterior shoulder in cases of normal vaginal delivery (active management of third stage of labour). A typical dose of methylergonovine, 0.2 mg administered intramuscularly, may be repeated as required at intervals of 2–4 hours. The dose of ergometrine is 0.25 mg IM which can be repeated every 5 minutes up to a maximum dose of 1.25 mg. The onset of action on the uterus after oral administration is 15 minutes, after IM injection 5 minutes and after intravenous injection

onset is almost immediate. It can be administered directly in the uterine muscle, if necessary. The total dose of ergometrine in 24 hours must not exceed 1,000 µg. Ergot alkaloids are sometimes used orally in a dose of 0.125 mg TDS for a maximum of 7 days to help in uterine involution in cases of secondary PPH. Ergometrine and syntocinon and commonly administered together. Syntometrine is a combination of ergometrine maleate 500 micrograms plus syntocinon 5 units in 1 mL.

Contraindications

Ergometrine should be used with caution in the following cases:
❑ *Suspected multifoetal pregnancy*: Administration of ergometrine with the delivery of the first baby can result in the entrapment of second baby due to tetanic contractions of the uterus.
❑ *Organic cardiac disease in the mother*: It may cause sudden sequestration of the uterine blood into the general circulation causing overloading of the right heart resulting in pulmonary oedema.
❑ *Severe pre-eclampsia and eclampsia*: Injection of ergometrine in these cases can result in a sudden rise in blood pressure.
❑ *Rh-negative mothers*: Injection of ergometrine can result in fetomaternal haemorrhage.

Side Effects

Since the stimulant action of ergometrine also involves the lower segment of the uterus along with the upper segment, this can occasionally result in the entrapment of the separated placenta. Its use can result in adverse effects such as vomiting, elevation of blood pressure and pain after birth requiring analgesia. It can inhibit lactation if higher doses are used for many days postpartum, as it inhibits prolactin release (dopaminergic action). Its prolonged use may lead to gangrene of the toes due to its vasoconstrictive effect. Rarely, it may cause chest pain due to acute coronary spasm.

Antimicrobial Drugs

Antimicrobial drugs are the drugs which are designed to inhibit/kill the infecting organism and to have no/minimal effect on the recipient. Classification of antibiotics based on the type of organisms (bacteria, virus, fungus, protozoa, helminths, etc.) against which they are primarily active is described in **Table 12.6**. The basis of selective microbial toxicity is the action of the drug on a component of the microbe (e.g. bacterial cell wall) or metabolic processes (e.g. folate synthesis) that is not found in the host. This could also be related to the high affinity of the antimicrobial drug for certain microbial biomolecules. Mode of action of various antimicrobial agents is summarised in **Table 12.7**.

TABLE 12.6	Classification of antibiotics based on the type of organisms against which they are primarily active
Type of organism	**Example of antibiotics**
Antibacterial agents	Penicillins, aminoglycosides, erythromycin, fluoroquinolones, etc.
Antifungal agents	Griseofulvin, amphotericin B, ketoconazole, etc.
Antiviral agents	Acyclovir, amantadine, zidovudine, etc.
Antiprotozoal agents	Chloroquine, pyrimethamine, metronidazole, diloxanide, etc.
Anthelmintic agents	Mebendazole, pyrantel, niclosamide, diethylcarbamazine, etc.

TABLE 12.7	Mechanism of action of various antibiotics
Mode of action	**Antibiotic**
Inhibition of cell wall synthesis	Penicillins, cephalosporins, cycloserine, vancomycin, bacitracin
Causing leakage from cell membranes	• Polypeptides—polymyxins, colistin and bacitracin • Polyenes—amphotericin b, nystatin and hamycin
Inhibition of protein synthesis	Tetracyclines, chloramphenicol, erythromycin, clindamycin and linezolid
Causing misreading of m-RNA code and effect on permeability	Aminoglycosides—streptomycin, gentamicin, etc.
Inhibition of DNA gyrase	Fluoroquinolones—ciprofloxacin and others
Interference with DNA function	Rifampin
Interference with DNA synthesis	Acyclovir, zidovudine
Interference with intermediary metabolism	Sulphonamides, sulphones, PAS, trimethoprim, pyrimethamine and metronidazole
Inhibition of nucleic acid synthesis	Metronidazole

Antibiotics

Antibiotics are the substances produced by microorganisms, which selectively suppress the growth of or kill other microorganisms at very low concentrations. Antibiotics can be classified as bacteriostatic or bactericidal based on the type of action they perform **(Table 12.8)**. Bacteriostatic antibiotic refers to a biological or chemical agent that stops bacteria from reproducing or inhibits their growth, while not necessarily killing them otherwise. On the other hand, bactericidal antibiotic refers to a biological or chemical agent which kills the bacteria.

Beta-Lactam Antibiotics

This is a group of antibiotics having a beta-lactam ring. The two major types of antibiotics belonging to this group are penicillins and cephalosporins. Monobactams and carbapenems are relatively newer drugs belonging to this group.

TABLE 12.8	Classification of antibiotics based on the type of action
Primarily bacteriostatic	**Primarily bactericidal**
• Sulphonamides • Erythromycin • Tetracyclines • Clindamycin • Chloramphenicol • Linezolid • Ethambutol	• Penicillins • Cephalosporins • Aminoglycosides • Vancomycin • Polypeptides • Ciprofloxacin • Rifampin • Metronidazole • Isoniazid • Cotrimoxazole • Pyrazinamide

Some primarily static drugs may become cidal at higher concentrations (as attained in the urinary tract), e.g. erythromycin, nitrofurantoin. On the other hand, some cidal drugs, e.g. cotrimoxazole, streptomycin, may only be static under certain circumstances.

Penicillins

Penicillin binds to specific penicillin-binding proteins (PBPs) in the cell wall, mainly of Gram-positive organisms. Penicillins are generally bactericidal and exert their effect by combining with and inhibiting the transpeptidase enzyme which cross-links the peptidoglycans in the cell wall. This weakens the cell wall and allows the cell to lyse under the influence of an osmotic gradient.

Penicillin resistance is usually due to production of altered PBPs or beta-lactamases which break the beta-lactam ring and inactivate the drug. Penicillin resistance is a common problem particularly in *Escherichia coli*, *Staphylococcus aureus* and *Enterobacter* which are mostly insensitive. *N. meningitidis* is generally sensitive and less than 20% resistance is found in *Pseudomonas* and *H. influenzae* species. However, the incidence continues to rise.

The beta-lactamase enzyme is often encoded in the bacterial plasmid. The plasmid is bacterial DNA (separate from the main chromosome) that can be translocated from bacterium to bacterium and it is for this reason that resistance may be also transferred.

Penicillin is mainly useful for group A *Streptococcus*, group B *Streptococcus*, meningococcal and pneumococcal infections, though anthrax is also generally sensitive. *Pneumococci* with modified PBPs are an increasing problem. Although the penicillins have similar spectra of action, their specific spectra may be changed by altering the beta lactam side chain. Different types of penicillins are described next.

Penicillin-G (Benzyl Penicillin)

Penicillin-G (PnG) is a narrow spectrum antibiotic; activity is limited primarily to Gram-positive bacteria, few Gram-negative ones and anaerobes. This is the most powerful penicillin and the drug of choice for sensitive organisms.

It is mainly administered via the parenteral route. It can be associated with adverse effects such as allergy, convulsions after massive bolus IV dosage, etc.

Phenoxymethyl Penicillin (Penicillin V)

It is usually administered orally. Though the spectrum of activity of penicillin V is similar to that of benzyl penicillin, its absorption is usually inadequate for serious infections. Therefore, it is used for milder infections, e.g. streptococcal pharyngitis, sinusitis, otitis media, less serious pneumococcal infections and trench mouth. It is used for the prophylaxis of rheumatic fever.

Ampicillin and Amoxicillin

Ampicillin and amoxicillin are broad spectrum antibiotics active against non-beta lactamase producing Gram-positive and Gram-negative organisms. They can be administered via oral or parenteral routes. They are inactivated by penicillinases and since up to 50% of *Escherichia coli* strains are now resistant, they would be an unsuitable choice. Co-amoxiclav consists of amoxicillin and the beta-lactamase inhibitor clavulanic acid. Thus, it is active against beta-lactamase producing bacteria that are resistant to amoxicillin, including strains of *Escherichia coli*. Amoxycillin is absorbed twice as well as ampicillin. It is excreted in the bile and urine.

Side effects: They do not have significant toxic effects on humans, but allergy is common. Rashes can occur in patients with infectious mononucleosis.

Cephalosporins

These are a group of semisynthetic antibiotics derived from "cephalosporin-C" obtained from a fungus *Cephalosporium*. They are chemically related to penicillins. Their spectrum of activity can be changed by the addition of different side chains at position 7 of beta-lactam ring. These antibiotics have been conventionally divided into four generations **(Table 12.9)**.

   All cephalosporins are bactericidal and have the same mechanism of action as penicillin, i.e. inhibition of bacterial cell wall synthesis. They have a broad spectrum of activity but are inactive against bacteroides, *Enterococcus faecalis*, *Pseudomonas*, etc. The main adverse effect related with cephalosporins is the risk of drug allergy. About 10% of the patients having penicillin allergies also react to cephalosporins. Though the incidence is low, resistance to these antibiotics has been developed by some organisms, even against the third generation compounds. The third generation compounds are highly active against Gram-negative Enterobacteriaceae; and few members inhibit *Pseudomonas* as well. However, they are less active on Gram-positive cocci and anaerobes. The distinctive feature of the

TABLE 12.9	Different generations of cephalosporins
First generation cephalosporins	
Parenteral • Cefazolin	*Oral* • Cephalexin • Cefadroxil
Second generation cephalosporins	
Parenteral • Cefuroxime • Cefoxitin	*Oral* • Cefaclor • Cefuroxime axetil • Cefprozil
Third generation cephalosporins	
Parenteral • Cefotaxime • Ceftizoxime • Ceftriaxone • Ceftazidime • Cefoperazone	*Oral* • Cefixime • Cefpodoxime proxetil • Cefdinir • Ceftibuten • Cefetamet pivoxil
Fourth generation cephalosporins	
Parenteral • Cefepime • Cefpirome	

fourth generation compound is non-susceptibility of this group of drugs to inducible chromosomal beta-lactamases as well as high potency against Enterobacteriaceae and spectrum of activity resembling the third generation compounds.

Sulphonamides

Sulphonamides (sulphamethoxazole and sulphadiazine) are primarily bacteriostatic against many Gram-positive and Gram-negative bacteria. Sulphonamides, being structural analogues of PABA (p-aminobenzoic acid), inhibit bacterial folate synthase. As a result, FA is not formed and a number of essential metabolic reactions suffer. It is mainly administered orally. Systemic use of sulphonamides alone is rare now except in cases of toxoplasmosis.

Side effects: Crystalluria or crystal formation in the kidneys and urine was related to the older, insoluble compounds, but is infrequent nowadays. Other side effects associated with sulphonamides include hypersensitivity reactions, hepatitis, erythema multiforme, etc. It also displaces bilirubin from albumin binding sites thereby aggravating neonatal jaundice. Kernicterus may be precipitated in the newborn, especially the premature ones, whose blood-brain barrier is more permeable.

Co-trimoxazole

The fixed dose combination of trimethoprim and sulphamethoxazole is called cotrimoxazole. Combination of sulphonamide and trimethoprim, with synergistic bactericidal effect acts via inhibition of folic acid synthesis.

It can be administered via oral or intravenous routes (not IM). Though cotrimoxazole is still used, its popularity in the treatment of systemic infections has declined. It is still sometimes used for the treatment of urinary tract infections, respiratory tract infections, bacterial diarrhoeas and dysentery, *Pneumocystis jiroveci* infection, chancroid, etc.

Side effects: All adverse effects seen with sulphonamides can be produced by cotrimoxazole. Nausea, vomiting, stomatitis, headache and rashes are the usual manifestations. Folate deficiency (megaloblastic anaemia) is infrequent and occurs only in patients with marginal folate levels. Blood dyscrasias occur rarely. Cotrimoxazole should not be given during pregnancy. There is theoretical teratogenic risk because trimethoprim is an antifolate. Neonatal haemolysis and methaemoglobinaemia can occur if it is administered near term.

Tetracycline

Tetracyclines are broad-spectrum antibiotics, which inhibit the growth of practically all microorganisms except fungi. However, their indiscriminate use has largely narrowed their field of usefulness. It is specifically indicated for brucellosis, chlamydial and rickettsial infections. It can be administered via oral or parenteral routes depending on the type used.

Side effects: They are likely to cause damage to the developing bones and teeth. Doxycycline can also result in photosensitivity. Fatty infiltration of liver and jaundice can occur occasionally with the use of tetracyclines. Oxytetracyclines are risky in pregnant women and can precipitate acute hepatic necrosis which can sometimes even be fatal. Renal damage can occur in presence of pre-existing renal disease. If administered from midpregnancy to 5th month of extrauterine life, it can affect the deciduous teeth, thereby resulting in their brownish discoloration. Ill-formed teeth which are susceptible to caries can also develop. Tetracycline if administered between 3 months and 6 years of age can affect the crown of permanent anterior dentition. If administered during late pregnancy or childhood, it can cause temporary suppression of the bone growth. The ultimate effect on stature is usually insignificant. However, deformity and reduction in height are a possibility with prolonged use.

Chloramphenicol

Chloramphenicol is primarily a bacteriostatic antibiotic, though high concentrations have been shown to exert cidal effect on some bacteria, e.g. *H. influenzae* and *N. meningitidis*. It is a broad-spectrum antibiotic, active against nearly the same range of organisms (Gram-positive and -negative cocci and bacilli, rickettsiae, mycoplasma) as tetracyclines. It can be administered via oral or parenteral routes. It is in little use nowadays except for in cases of typhoid and haemophilus meningitis.

Side effects: Bone marrow depression is the most important side effect of this drug. Of all drugs, chloramphenicol is the most important cause of aplastic anaemia, agranulocytosis, thrombocytopenia or pancytopenia. It can cause "grey baby syndrome" when administered to the infants, but does not harm the foetuses. This syndrome occurs when high doses (~100 mg/kg) were given prophylactically to neonates, especially the premature ones. The baby stops feeding, vomits, becomes hypotonic and hypothermic, abdomen gets distended, respiration becomes irregular; an ashen grey cyanosis develops in many, followed by cardiovascular collapse and death.

Erythromycin

Erythromycin is bacteriostatic at low but cidal (for certain bacteria only) at high concentrations. Cidal action depends on the organism concerned and its rate of multiplication. Sensitive Gram-positive bacteria accumulate erythromycin intracellularly by active transport which is responsible for their high susceptibility to this antibiotic. The spectrum of activity of erythromycin is similar to that of benzyl penicillin including *Haemophilus*. It is drug of choice for pertussis prophylaxis. The drug is usually administered orally and is excreted in the bile. The estolate salt of erythromycin is typically better absorbed orally. The estolate salt of erythromycin can be especially associated with cholestatic jaundice.

Nitrofurantoin

It is a broad spectrum, bacteriostatic antibiotic. However, it may be cidal at higher concentrations and in acidic urine. Its activity is enhanced at lower pH. It is widely used for the treatment of urinary tract infection. Previously many Gram-negative bacteria were susceptible, but due to development of resistance, activity is now restricted largely to *E. coli*. Resistance to nitrofurantoin does not develop during continued therapy. No cross resistance with any other antimicrobial agent is known. The main side effect which can occur with this drug is the development of pulmonary infiltration.

Lincosamides (Lincomycin and Clindamycin)

These potent lincosamide antibiotics have a mechanism of action and spectrum of activity which is similar to erythromycin. They are typically useful against *S. aureus* and bacteroides. They also exhibit partial cross resistance with erythromycin. Since lincosamides are particularly concentrated in bones, they are used for the treatment of osteomyelitis. They can be administered via oral or parenteral routes. The main adverse effect associated with this antibiotic is pseudomembranous colitis.

Quinolones

These are synthetic antimicrobials having a quinolone structure that are active primarily against Gram-negative bacteria, though the newer fluorinated compounds also inhibit Gram-positive ones. These are known as fluoroquinolones. The first generation fluoroquinolines includes drugs such as ciprofloxacin, ofloxacin, norfloxacin, and pefloxacin. Ciprofloxacin has a 6-fluoro substituent which confers enhanced antibacterial potency against both Gram-positive and Gram-negative organisms, including *Escherichia coli*. The second generation fluoroquinolones include drugs like levofloxacin, moxifloxacin, lomefloxacin, gemifloxacin, etc.

Side effects: Side effects include rashes, pseudomembranous colitis as it is a broad-spectrum antibiotic and rarely cholestasis. Convulsions are rarely associated with ciprofloxacin. It is unlikely to be associated with resistance.

Aminoglycosides

The aminoglycosides group includes gentamicin, neomycin, streptomycin, amikacin and tobramycin. All are bactericidal, and active against Gram-negative organisms. However, they are not active against anaerobes. All are used as sulphate salts, which are highly water soluble; solutions are stable for months. They ionise in solution; are not absorbed orally; distribute only extracellularly; and do not penetrate brain or CSF. All are excreted unchanged in urine by glomerular filtration.

The British National Formulary recommends that aminoglycosides, e.g. gentamicin should be used with penicillin or metronidazole, or both, to improve broad coverage. Specifically amikacin, gentamicin and tobramycin are active against *Pseudomonas*; streptomycin is effective and widely used against *Mycobacterium tuberculosis*. Streptomycin is mainly reserved for the treatment of tuberculosis. This drug must be avoided during pregnancy.

Excretion is via the kidney, and accumulation occurs in renal failure. Aminoglycosides should not be given with furosemide. If there is renal failure or high pre-dose serum levels, the dosing interval must be increased.

They are not absorbed from the gut; therefore, they cannot be administered orally. They are administered through parenteral route (usually via intravenous route). Tobramycin can be given by a nebuliser). Neomycin is too toxic for parenteral administration, but the ointment can be used for skin infection.

Side Effects: Aminoglycosides are nephrotoxic but this is a dose-related effect. Blood levels must be checked and caution taken with other nephrotoxic agents. Aminoglycosides are ototoxic (vestibular and auditory) particularly in the elderly. Since the excretion of the drug occurs via kidneys, the toxic effects of the drug are more likely to occur in patients with renal failure.

Side effects of the aminoglycosides include ototoxicity and renal toxicity (excretion is principally by the kidney) and drug concentrations need to be carefully monitored during treatment. Side effects are more common in elderly subjects due to the reduced volumes of distribution and reduced renal reserve. Auditory and renal function should be monitored regularly in the elderly. Tinnitus is usually the first manifestation of ototoxicity. Irreversible vestibular damage results from the ototoxicity. A rare side effect of aminoglycosides is ventilatory failure associated with impaired neuromuscular junction conduction and it should not be given to patients with myasthenia gravis.

Vancomycin

It is a glycopeptide antibiotic, which assumed special significance due to its efficacy against MRSA, *Strep. viridans, Enterococcus* and *Cl. difficile*. It also exerts bactericidal action on Gram-positive cocci, *Neisseria, Clostridia* and diphtheroids. In hospitals where it has now been extensively used, there has been an emergence of vancomycin-resistant *Staph. aureus* (VRSA) and vancomycin-resistant *Enterococcus* (VRE). These nosocomial bacteria are resistant to methicillin and most other antibiotics as well. It is excreted renally and liver impairment does not affect its elimination.

Side effects: Vancomycin is associated with high systemic toxicity. It can cause plasma concentration-dependent nerve deafness that may be permanent. It is also associated with dose-related kidney damage. Simultaneous administration of other ototoxic and nephrotoxic drugs like aminoglycosides must be therefore avoided. Vancomycin can cause histamine release by directly acting on the mast cells. Rapid IV injection can therefore cause a syndrome, known as the "Red man syndrome", which is associated with chills, fever, urticaria and intense flushing.

Antiamoebic Drug

Metronidazole

Metronidazole (flagyl) is effective for treating against infections with protozoa such as *Trichomonas vaginalis*, *Amoeba*, and *Giardia*. It also is effective against anaerobic bacterial infections. It is the treatment of choice for *Trichomonas* and bacterial vaginosis. Circulating concentrations may be affected by concomitant cimetidine administration. This is because its metabolism is inhibited by cimetidine, which is a hepatic enzyme inhibitor. It can be administered via oral or intravenous route. It can also be administered rectally.

Side effects: It can cause a "disulfiram-like" reaction when taken with alcohol. Therefore, advice should be given to patients to avoid alcohol when taking metronidazole due to a disulfiram reaction, including symptoms such

as sickness, abdominal pains and headache. Other side effects of metronidazole include peripheral neuropathy, discolouration of urine and a metallic taste in the mouth. Although no teratogenic potential has been demonstrated, its mutagenic potential warrants caution. Therefore, metronidazole is contraindicated during the first trimester of pregnancy. Its use in breastfeeding is controversial and lactation is withheld for 12–24 hours following a 2-gram dose.

Antiviral Drugs

Amantadine: Amantadine, which is an anti-Parkinsonian agent, is also used for the treatment of certain RNA viruses and influenza viruses. Amantadine has a deleterious effect on breastfeeding.

Interferon: Interferons are low molecular weight glycoprotein cytokines produced by host cells in response to viral infections. Alpha-interferon is produced by unstimulated T cells and monocytes. High levels of alpha-interferon are found in the amniotic fluid and in the placenta.

Beta-interferon is produced by stimulated epithelial cells in infected organs and by fibroblasts in tissue culture. Gamma-interferon is produced by sensitised T cells.

Zidovudine: It is a thymidine analogue. Following phosphorylation in the host cell, zidovudine triphosphate selectively inhibits viral reverse transcriptase in preference to cellular DNA polymerase. Zidovudine is used in the treatment of HIV. Zidovudine does not eliminate the HIV virus. It only reduces the HIV-related mortality rate and the incidence and severity of opportunistic infections. Anaemia is a common side effect of zidovudine**.**

Acyclovir: Acyclovir prevents DNA synthesis. It itself is inactive. However, it is metabolised by a herpes simplex specified enzyme (thymidine kinase) to acyclovir triphosphate, which prevents further DNA synthesis **(Fig. 12.1)**. Acyclovir triphosphate then competes with deoxyguanosine triphosphate for a position in the viral DNA. Acyclovir phosphate also competitively inhibits herpes virus DNA polymerase. Acyclovir helps treat
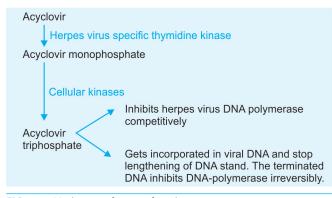
FIG. 12.1: Mechanism of action of acyclovir

herpes simplex encephalitis and decreases recurrence of genital herpes. It is administered intravenously for 10 days. It prevents herpetic neuralgia if administered at the onset of infection but is ineffective if given later (BNF 5.3). Ganciclovir, related to acyclovir, is effective against CMV.

Antifungal drugs

❑ *Antibiotics*
  • *Polyenes:* Amphotericin B (AMB), Nystatin, Hamycin
  • *Heterocyclic benzofuran*: Griseofulvin
❑ *Antimetabolite* Flucytosine (5-FC)
❑ *Azoles*
  • Imidazoles
    – *Topical*: Clotrimazole, econazole, miconazole, oxiconazole
    – *Systemic:* Ketoconazole
  • *Triazoles (systemic)*
    – Fluconazole, itraconazole, voriconazole, posacon-azole

Amphotericin B is active against a wide range of yeasts and fungi—*Candida albicans, Histoplasma capsulatum, Cryptococcus neoformans, Blastomyces dermatitidis, Coccidioides immitis, Torulopsis, Rhodotorula, Aspergillus, Sporothrix,* etc. The polyenes and imidazoles are topically used for vaginal infections.

Ketoconazole is the first orally effective broad-spectrum antifungal drug, useful in both dermatophytosis and deep mycosis. The most important adverse effect of ketoconazole is its hormonal effects. It decreases androgen production from testes, and displaces testosterone from protein binding sites. As a result it causes a reduction in androstenedione and testosterone levels and an increase in progesterone and 17-hydroxyprogesterone levels. If administered for a few weeks, this drug can result in gynaecomastia, loss of hair and libido, and oligozoospermia. Menstrual irregularities occur in some women due to suppression of oestradiol synthesis.

Anticancer Agents

Alkylating agents: These compounds produce highly reactive carbonium ion intermediates which transfer alkyl groups to cellular macromolecules by forming covalent bonds. Alkylating agents exert their cytotoxic effects through alkylation of DNA as well as many other cellular constituents. Alkylating agents cause structural damage to chromosomes at the time of replication during interphase. **Table 12.10** describes various types of alkylating agents.

Platinum derivatives: Platinum derivatives cause cross-linkage of complementary DNA strands, thus preventing replication. These include drugs like cisplatin and carboplatin. Cisplatin is very effective in the treatment of metastatic testicular and ovarian carcinoma. It is widely used in the treatment of many other solid tumours like

TABLE 12.10	Different types of alkylating agents
Agent	**Indication**
Mustine (Nitrogen mustard)	Used for the treatment of Hodgkin and non-Hodgkin lymphomas. It is highly reactive and local vesicant. Therefore, it can be given only by IV route; it is administered into the tubing of fast-running infusion. Mustine produces several acute effects like nausea, vomiting and haemodynamic changes. Extravasation during IV injection may cause sloughing.
Cyclophospha-mide	It is inactive as such. Transformation into active meta-bolites occurs in the liver. It produces few acute effects and is not locally damaging. It can cause alopecia and haemorrhagic cystitis. It is used for the treatment of non-Hodgkin's lymphomas and solid tumours.
Melphalan	Melphalan is very effective in multiple myeloma and has also been used in advanced ovarian cancers. Most important side effect of melphalan is bone marrow depression. Other complications include infections, diarrhoea and pancreatitis.
Busulphan	It is the drug of choice for chronic myeloid leukaemia. It has little adverse effect on lymphoid tissue and GIT. However, it can result in side effects such as hyper-uricaemia, pulmonary fibrosis, skin pigmentation and sterility.
Thiotepa	It is a highly toxic drug which is seldom used nowadays for the treatment of solid tumours.

TABLE 12.11	Uses of antibiotics for treatment of various cancers
Drug	**Uses**
Actinomycin D (Dactinomycin)	Wilms' tumour and childhood rhabdomyosarcoma
Daunorubicin (Rubidomycin)	Daunorubicin is used for the treatment to acute myeloid as well as lymphoblastic leukaemia
Doxorubicin	Treatment of acute myeloid and lymphoblastic leukaemia as well as solid tumours, such as breast, thyroid, ovary, bladder and lung cancers, sarcomas and neuroblastoma.
Epirubicin	Adjuvant therapy of breast carcinoma, and treatment of gastroesophageal, pancreatic, hepatic and bladder carcinoma
Mitoxantrone	Used for the treatment of acute myeloid leukaemia, advanced hormone refractory prostate cancer and occasionally in breast and hepatic carcinoma, and non-Hodgkin lymphoma.

Antimetabolites: These are analogues related to the normal components of DNA or of coenzymes involved in nucleic acid synthesis. They competitively inhibit utilisation of the normal substrate or get themselves incorporated forming dysfunctional macromolecules. These include folic acid antagonists, purine antagonists and pyrimidine antagonists. These are described briefly in **Table 12.12**.

Anticoagulants

These are drugs used for reducing the coagulability of blood. The two most commonly used anticoagulants are heparin and warfarin. These would be described in details next. The difference between the two is tabulated in **Table 12.13**.

Heparin

Heparin is a strongly acidic mucopolysaccharide (not protein), and as the name heparin implies it is derived from the liver. Heparin acts through antagonism of a number of factors involved in the coagulation cascade. It principally enhances antithrombin III activity, which in turn inhibits activated factors XII, XI, X, IX and thrombin. Heparin also activates lipoprotein lipase in plasma, lowering the plasma triglycerides and inhibiting platelet aggregation by fibrin.

Dosage

The dosage of heparin can be monitored based on the effect it has on the activated partial thromboplastin time (APPT or PTTK) which should be 1.5–2.0 times the normal or based on the measurement of the activity of factor Xa.

Side effects

Side effects of heparin therapy include haemorrhage, hypersensitivity reactions and osteoporosis (after prolonged administration). In high dosage heparin may have an antiplatelet effect, but it is also associated with heparin-induced thrombocytopenia (HIT). A side effect in up to 5% of patients is heparin-induced thrombocytopenia

malignancies of lung, bladder, oesophagus, stomach, liver, and head and neck. Cisplatin is a highly emetic drug. Antiemetics must be routinely administered before infusing this drug. The most important toxicity is renal impairment which is dependent on total dose administered. Other side effects include tinnitus, deafness, sensory neuropathy and hyperuricaemia. Carboplatin is a less reactive second generation platinum compound that is better tolerated and has a toxicity profile different from cisplatin, but mechanism of action and clinical utility are similar. Nephrotoxicity, ototoxicity and neurotoxicity are low in comparison to that of cisplatin. It is primarily indicated in ovarian carcinoma of epithelial origin, and has also been found to be effective in the treatment of squamous carcinoma of head and neck, small cell lung cancer, breast cancer and seminoma.

Spindle poisons: These include drugs like vincristine and vinblastine which arrest the cells in metaphase of mitosis after binding to tubulin. Besides the other side effects related with the use of chemotherapy drugs, these drugs may sometimes cause cranial nerve palsies.

Antibiotics: These include drugs like actinomycin D, daunorubicin, doxorubicin (adriamycin) and mitozantrone. These are the products obtained from microorganisms and have prominent antitumour activity. Practically all of them intercalate between DNA strands and interfere with its template function. Due to this, they also hamper the process of RNA replication/synthesis. The various cancers in which these antibiotics can be used are summarised in **Table 12.11**.

TABLE 12.12	Different types of antimetabolites used for treatment of various cancers
Drug	**Uses**
Folic acid antagonist (methotrexate)	Methotrexate acts by inhibiting dihydrofolate reductase, thereby blocking the conversion of dihydrofolic acid to tetrahydrofolic acid. It does not cross the blood brain barrier. It exerts major toxicity on bone marrow. Low doses given repeatedly cause megaloblastic anaemia, whereas high doses produce pancytopenia. Other common side effects may include, mucositis, diarrhoea, desquamation and bleeding in the GIT. It can have toxic and teratogenic effect on the embryonic mesenchyme. Toxicity of methotrexate is overcome by administering folinic acid (N5 formyl THFA, crtrovorum factor) or thymidine. Methotrexate is curative in cases of choriocarcinoma and is also highly effective in maintaining remission in children with acute leukaemias and Burkitt's lymphoma
Purine antagonists (mercaptopurine, azathioprine)	6-mercaptopurine is especially useful in treatment of childhood acute leukaemia, choriocarcinoma and some solid tumours as well. Azathioprine primarily suppresses cell mediated immunity and is used mainly for the treatment of autoimmune diseases (rheumatoid arthritis, ulcerative colitis) as well as in organ transplantation. The main toxic effect of mercaptopurine is bone marrow depression, which develops slowly. It produces a high incidence of reversible jaundice. Hyperuricaemia may also occur, and can be reduced by allopurinol.
Pyrimidine antagonists (fluorouracil and cytosine arabinoside)	5-Fluorouracil (5-FU) is a commonly used anticancer drug for treatment of many solid malignancies, especially of colon, rectum, stomach, pancreas, liver, urinary bladder, head and neck. Oral absorption of 5-FU is unreliable. It is primarily used by IV infusion. Major toxicity of 5-Fluorouracil includes bone marrow suppression and gastrointestinal side effects such as mucositis, diarrhoea, nausea and vomiting. Peripheral neuropathy (hand-foot syndrome) also occurs. Primary use of 5-fluorouracil is induction of remission in cases of acute myelogenous as well as lymphoblastic leukaemia in children and in adults. It is also used for treatment of blast crisis in chronic myelogenous leukaemia and non-Hodgkin's lymphoma.

(HIT). HIT is due to combination of heparin with heparin-binding protein in platelets (PF4). This antigenic complex induces the formation of IgG, which binds platelets causing clumping. If severe enough consumptive thrombocytopenia and thrombosis follow.

As heparin is so strongly acidic it binds avidly to proteins and so it poorly crosses the placenta and other membranes. Pregnant women requiring anticoagulation with warfarin are converted to heparin, as this has a much lower risk of teratogenicity.

TABLE 12.13	Difference between warfarin and heparin	
	Heparin	**Warfarin**
Chemistry	Mucopolysaccharide	Coumarin derivative
Source	Hog lung, pig intestine	Synthetic
Route of administration	Parenteral (IV, SC)	Oral
Onset of action	Immediate	Delayed (1–3 days)
Duration of action	4–6 hours	3–6 days
Activity	In vitro and in vivo	In vivo only
Mechanism	Blocks action of factor X and thrombin	Inhibits synthesis of clotting factors
Antagonist	Protamine sulphate	Vitamin K
Variability in response	Little	Marked
Laboratory control	aPTT/clotting time (desirable)	Prothrombin time/INR (essential)
Drug interactions	Few and not significant	Many and significant
Use	For initiation of therapy	For maintenance

Abbreviations: aPTT, activated partial thromboplastin time; INR, International normalized ratio; IV, intravenous; SC, subcutaneous

Warfarin

Warfarin and its congeners act as anticoagulants in vivo, not in vitro. This is because they act indirectly by interfering with the synthesis of vitamin K dependent clotting factors in the liver. They act as competitive antagonists and therefore cause lowering of the plasma levels of functional clotting factors in a dose dependent manner. They interfere with the regeneration of active hydroquinone form of vitamin K, which acts as a co-factor for the enzyme γ-glutamyl carboxylase. This enzyme is responsible for carrying out the final step of γ-carboxylation of the glutamate residues of prothrombin and factors VII, IX and X. This carboxylation is essential for the clotting factors to bind to bind with Ca^{2+} and to get bound to the phospholipid surface. This is essential for the coagulation sequence to proceed.

In case of first trimester exposure, it is likely to cause embryopathy in 5–10% of pregnancies. For details, refer to the section on teratogenesis. The embryopathy occurs due to the involvement of vitamin K in the post-translational modification of proteins enabling them to bind with calcium. In the second and third trimester, administration of warfarin is associated with recurrent brain microhaemorrhages, resulting in optic atrophy, dorsal midline dysplasia and mental retardation. Warfarin administration must be avoided after 36 weeks of pregnancy to prevent maternal and neonatal complications related to delivery.

Drug Interactions

Numerous drugs may interact with warfarin at pharmacokinetic or pharmacodynamic level, thereby enhancing or reducing its effect. These interactions are clinically important because it may sometimes even prove to be fatal

if bleeding occurs due to high effective dose of warfarin. The anti-coagulant effects of warfarin may be exacerbated through the inhibition of its metabolism by cytoP450. On the other hand, the anti-coagulant effect of warfarin may be reduced due to the induction of its metabolism by cytoP450. In this context, one must remember that rifampicin is a powerful enzyme inducer. Besides the changes in drug metabolism, there are many other mechanisms through which the anticoagulant action of warfarin can be enhanced or reduced. The various drugs which can result in enhanced or reduced effect of warfarin are described next:

Enhanced Anticoagulant Action

❏ *Broad-spectrum antibiotics*: Inhibit gut flora and reduce vitamin K production.
❏ *Newer cephalosporins (ceftriaxone, cefoperazone)*: Cause hypoprothrombinaemia by the same mechanism as warfarin—additive action.
❏ *Aspirin*: Inhibits platelet aggregation and causes gastrointestinal bleeding; this may be hazardous in anticoagulated patients. High doses of salicylates have synergistic hypoprothrombinemic action and also displace warfarin from protein binding site.
❏ *Long-acting sulphonamides, indomethacin, diazepam, phenytoin and probenecid*: Displace warfarin from plasma protein binding.
❏ *Chloramphenicol, erythromycin, celecoxib, cimetidine, allopurinol, amiodarone and metronidazole*: Inhibit warfarin metabolism.
❏ *Tolbutamide and phenytoin*: Inhibit warfarin metabolism and vice versa.
❏ *Liquid paraffin (habitual use)*: Reduces absorption of vitamin K.

Reduced Anticoagulant Action

❏ *Barbiturates (but not benzodiazepines), carbamazepine, rifampin and griseofulvin* induce the metabolism of oral anticoagulants. The dose of anticoagulant determined during therapy with these drugs would be higher: if the same is continued after withdrawing the inducer—marked hypoprothrombinemia can occur. This can result in fatal bleeding.
❏ *Oral contraceptives*: Increase blood levels of clotting factors.

Anti-hypertensive Agents

Different types of anti-hypertensive agents and their respective mechanisms of action are described in **Table 12.14**.

Angiotensin-converting-enzyme Inhibitors

Angiotensin-converting-enzyme (ACE) inhibitors include drugs such as lisinopril, trandolapril, ramipril, enalapril,

TABLE 12.14	Different types of antihypertensive agents
Drug	**Mechanism of Action**
Alpha-methyldopa	A centrally acting sympathomimetic agent
Propranolol/Atenolol	Non-selective beta-receptor antagonist
Losasartan, eprosartan and irbesartan	Blockade of the angiotensin II receptor
Prazosin and doxazosin	Specific alpha-1-receptor antagonist
Hydralazine hydrochloride	Directly acting vasodilator
Sodium nitroprusside	Vasodilatation
Carvedilol	Combined alpha- and beta-blocking effects
Phentolamine	Alpha-adrenoceptor blockade
Captopril	Angiotensin converting enzyme inhibition
Verapamil	Calcium antagonist

captopril, etc. These drugs prevent the conversion of angiotensin I to angiotensin II. These drugs are very effective in conjunction with diuretics. ACE inhibitors cause renal failure in patients with bilateral renal artery stenosis due to their inhibition of angiotensin II synthesis. This may cause a reduction in the rate of glomerular filtration or may even end it altogether. Therefore, they should be used with caution in patients who may have undiagnosed renovascular disease due to the risks of renal impairment.

There is good evidence that ACE inhibitors can cause intrauterine growth restriction (IUGR), oligohydramnios and neonatal anuria. Renal failure and death may occur when given in later pregnancy. Therefore, if possible, they should be stopped before conception and certainly by the 12th week of gestation. However, they can be safely used in the puerperium.

Alpha-methyldopa

Alpha-methyldopa is a central sympathomimetic agent, acting on the α-receptors in sympathetic nuclei within the medulla reducing sympathetic activity. It is the most tested antihypertensive drug for use in pregnancy. The action of methyldopa can be augmented with the help of calcium-channel blockers. It is unusual, however, to require more than one drug during pregnancy. An increasing need for medication to control severe hypertension must raise the suspicion not only of superimposed pre-eclampsia, but also of underlying renal problems. Long-term follow-up of children exposed in utero to alpha-methyldopa has demonstrated normal development up to the age of 8 years.

Dosage

The dose range is 1–3 gram daily, given in two to four divided doses.

Contraindications

It is contraindicated in the puerperium. Its adverse effects on the mother are the same as those in non-pregnant

individuals: for instance, somnolence or depression. These make it unsuitable for use in the puerperium.

Side Effects

Side effects of alpha-methyldopa include effects such as depression, nasal congestion, haemolytic anaemia, weight gain, oedema, pyrexia, visual disturbances, gastrointestinal disturbance, arthralgia, parkinsonism, nightmares, gynae-comastia, hyperprolactinaemia, galactorrhoea, hepatitis, etc.

Beta-adrenoceptor Blocking Drugs

These drugs act cause a reduction in blood pressure by causing a reduction in cardiac output, inhibition of renin release and peripheral effects on the adrenergic receptors. They are usually slow acting drugs. Beta-adrenoceptor blocking drugs cause intrauterine growth restriction when given from early pregnancy. This is most striking when treatment is started early in the second trimester. Therefore, beta-blockers are best contraindicated during pregnancy, except for short-term treatment in the third trimester. Its use is best avoided in case of asthmatics. It should be only used in cases with excessive catecholamine secretion.

Beta blockers such as oxprenolol, pindolol, acebutolol and celiprolol have intrinsic sympathomimetic activity, which reduces the incidence of side effects such as cold extremities (hands and feet) and bradycardia.

Calcium Antagonists

These drugs inhibit the entry of calcium into the cells and include drugs such as nifedipine, verapamil, etc. They may cause uterine relaxation and a throbbing headache.

Labetalol

Labetalol is a combined beta and alpha adrenergic receptor antagonist with a ratio of activity between 2:1 and 5:1 respectively. The alpha-blocking action causes arteriolar vasodilatation and lowers peripheral resistance, which is why labetalol is preferred antihypertensive drug in pregnancy. It is used for treating severe hypertension and pre-eclampsia. This drug helps in lowering BP smoothly but rapidly, without causing tachycardia. Labetalol has a half-life of approximately 4 hours (not 2) and is approximately 50% protein bound. It has been shown to cross the placental barrier, but not the blood brain barrier. Oral administration of the drug undergoes extensive first-pass metabolism. It is metabolised in the liver and excreted in the urine and faeces. It also helps in reducing bile secretion.

Indications

Labetalol is used for the following indications:
❑ It is a moderately potent hypotensive agent used for the treatment of pre-eclampsia (pregnancy-induced hypertension)
❑ Acute or chronic hypertension associated with pheo-chromocytoma
❑ Clonidine withdrawal
❑ Essential hypertension.

Route of Administration
❑ It can be administered via oral/intravenous route.
❑ Labetalol is available in the form of 100 mg, 200 mg and 300 mg tablets. Ampoules of labetalol are available containing 5 mg/mL of solution.

Dosage

Labetalol is given as a 20 mg intravenous bolus, followed by 40 mg after 10 minutes. If the first dose is not effective, then 80 mg is administered every 10 minutes. Maximum total dose of 220 mg can be administered. It can also be administered in the form of a continuous infusion—250 mg of labetalol in 250 mL of normal saline, administered at the rate of 20 mg/hour (20 mL/minute). The onset of action of intravenous dosage is within 5–10 minutes. Orally, labetalol is administered in the dose of 100 mg 8-hourly, which may be increased to 800 mg/day.

Contraindications

Labetalol is contraindicated in the following conditions:
❑ Asthma, airway obstructive disease
❑ Third-degree, second-degree heart block, or moderate-to-severe-first-degree heart block
❑ Congestive heart failure
❑ Bradycardia
❑ Hypotension
❑ Cardiogenic shock.

Side Effects

This drug can produce side effects like flushing, headache, nausea and vomiting. It is contraindicated in women with asthma and first-degree heart block. Therefore, its use must be avoided in women with asthma or congestive heart failure. Due to a lower incidence of side effects like maternal hypotension, the use of labetalol now supplants that of hydralazine. When administered orally to women with chronic hypertension, it seems to be as safe and effective as methyldopa, although neonatal hypoglycaemia can occur with higher doses. Severe hepatocellular damage has been reported after both short- and long-term use and the reduction of bile secretion may rarely lead to jaundice.

Antiepileptic Drugs

Pharmacokinetic properties of various antiepileptic drugs are altered during pregnancy. Therefore, therapeutic drug monitoring is beneficial during this time. Antiepileptic drugs such as phenytoin, primidone, phenobarbital, and carbamazepine and sodium valproate are all capable of crossing placenta. These drugs can cause major

abnormalities such as neural tube defects, orofacial and congenital heart defects, foetal hydantoin syndrome, etc. Foetal hydantoin syndrome has been described in details later in his chapter in the section on teratogenesis. Sodium valproate and carbamazepine may cause neural tube defects and spina bifida (always lumbar). Phenobarbital is usually safer than phenytoin. The risk of teratogenicity rises with the combination use of more than one drug. Carbamazepine, phenytoin and valproic acid are usually safe during pregnancy. Carbamazepine is an anticonvulsant which acts by blocking neuronal sodium channels reducing the sodium ion flux until it is insufficient to evoke an action potential. Succinimides (especially ethosuximide) are commonly used for the treatment of petit mal epilepsy during pregnancy. This drug has low or no teratogenic potential.

Altered pharmacokinetics during pregnancy may lead to the changes in drug levels, resulting in a fall in the concentration of free drug levels. Besides the teratogenic effects, the various antiepileptic drugs can be associated with a wide range of side effects which are described next in details.

Phenytoin

Phenytoin is a commonly prescribed anticonvulsant used for treating most types of seizure disorders, with the exception of absence seizures. It is also used for the treatment of status epilepticus.

If a woman is free of fits, there is usually no need to measure serial drug levels or adjust their dose. An exception is lamotrigine as levels of this drug invariably fall during pregnancy. In women having regular seizures, who are dependent on critical drug levels, regular monitoring of the blood levels is required. Dosage of the anticonvulsant drug can be changed based on the serum concentration of these drugs.

Side Effects

Most people who take phenytoin don't have too much trouble with side effects. That is reason for the common usage of this drug. Most people are able to tolerate these side effects without a change in the amount of phenytoin dosage. However, if the side effects persist, it is usually a sign indicative of high levels of phenytoin in the body. A change in the dosage or type of phenytoin in these cases may prove to be helpful. Various side effects which can occur with phenytoin include the following:

❏ *Effect on the facial features*: Gingival hypertrophy and tenderness as well as coarse facial features, hirsutism and acne may occur. Overgrowth of the gums (gingival hyperplasia) is more common in children than in adults. It can be reduced by vigorous brushing, daily flossing, and regular visits to the dentist, who may recommend additional treatments. If phenytoin is stopped, the gum problems won't get worse, and in some cases will go away within a few months.

❏ *Haematological side effects*: These include side effects such as megaloblastic anaemia, aplastic anaemia, thrombocytopaenia, etc.

❏ *Neurological side effects*: These include symptoms such as peripheral neuropathy, ataxia, reduced coordination, shaking of the hands, slowed thinking and movement, memory problems, slurred speech, poor concentration, etc. Rarely, this may be associated with symptoms such as nystagmus, dyskinesias, granulocytosis, etc.

❏ *Eye involvement*: Eye involvement may occur in cases of overdosage including blurred vision and nystagmus. Cataracts are not associated with phenytoin use.

❏ *Allergic reactions*: Approximately 1 in 10 people who take phenytoin have a red rash within the first few weeks of taking it.

❏ *Bones*: Long-term use of phenytoin also has been found to cause weakening of the bones. Bone disease is even more likely if a combination of seizure medicines is used. Precautions for reducing bone problems include regular daily exercise, regular intake of vitamin D supplements, and consumption of foods rich in calcium.

Sodium Valproate

Sodium valproate is used for the treatment of all forms of epilepsy including absences. Blood levels do not relate to anticonvulsant effectiveness.

Indications

Sodium valproate is indicated for all forms of epilepsy including absences.

Contraindications

❏ Active liver disease
❏ Family history of severe hepatic dysfunction
❏ Porphyria.

The liver function should be monitored before therapy and during the first 6 months. Liver dysfunction, including fatal hepatic failure has occurred especially in children under the age of 3 years and those with metabolic or degenerative disorders, organic brain disease or severe seizure disorders associated with mental restriction.

Raised liver enzymes are not uncommon, but are usually reversible. Patients or their carers should be told how to recognise signs of blood or liver disorders, and advised to seek immediate medical attention if symptoms develop.

Side Effects

There may be dose-related side effects and levels above the therapeutic index may be tolerated without symptoms. Hyperactivity and behavioural problems are listed as recognised side effects, however are very rarely seen. There

is a potential for prolonged bleeding as valproate can cause thrombocytopenia as well as inhibition of platelet aggregation. Liver dysfunction may also occur. There is a risk of teratogenic side effects with anticonvulsant therapy and there is a particular association of neural tube defects with the use of sodium valproate. Patients should be counselled and informed and the treatment changed (lamotrigine is an alternative). Other side effects include gastrointestinal upset, ataxia and tremor, hyperammonaemia, increased appetite and weight gain, transient hair loss, etc.

Drugs for Treating Galactorrhoea

Bromocriptine

Bromocriptine is a synthetic ergot derivative (2-bromo-α-ergocryptine), acting as potent agonist on D2, but as partial agonist or antagonist on D1 receptors. It is also a weak α-adrenergic blocker but not an oxytocic.

Most of the actions of this drug are based on its activity on D2 receptors. It decreases the release of prolactin from the pituitary gland by activating dopaminergic receptors on lactotrope cells. Therefore, it acts as a strong anti-galactopoietic drug. It reduces growth hormone levels in the majority of patients.

Indications

Bromocriptine is administered in the following conditions:
❑ Hyperprolactinaemia due to microprolactinomas causing galactorrhoea, amenorrhoea and infertility in women. Bromocriptine and cabergoline are the first-line drugs for most cases. Bromocriptine should be stopped when pregnancy occurs, though no teratogenic effects have been yet reported.
❑ Hyperprolactinomas causing gynaecomastia, impotence and sterility in men.
❑ *Induction of ovulation*: It can be used in combination with clomiphene citrate (CC) in patients with CC resistant anovulation.
❑ Suppression of lactation and breast engorgement in cases of neonatal death.
❑ *Non-gynaecological disorders*: Parkinsonism, type 2 diabetes mellitus, hepatic coma, acromegaly due to small pituitary tumours.

Dosage

Bromocriptine is administered orally should always be started at a low dose: 1.25 mg BD and then gradually increased till response occurs or until the side effects become limiting. Bromocriptine in the dosage of 2.5–10 mg/day proves to be useful in most cases.

Contraindications

Bromocriptine is contraindicated in patients having following disorders:

❑ Mental disturbances
❑ Coronary diseases
❑ Peripheral vascular diseases
❑ Cerebrovascular diseases
❑ Hypertension
❑ Chronic heart diseases.

Side Effects

The various side effects, which can occur with bromocriptine, are as follows:
❑ *Early*: These include nausea, vomiting, constipation, nasal stuffiness/blockage and conjunctival injection. Postural hypotension/syncope may occur at the initiation of therapy.
❑ *Late*: Behavioural alterations, mental confusion, hallucinations, abnormal movements, livedo reticularis. Bromocriptine is sometimes associated with reports of increased libido and hypersexuality.

Cabergoline

It is a newer D2 agonist; more potent; more D2 selective and is longer acting (half-life more than 60 hours) in comparison to bromocriptine. Cabergoline is a dopamine agonist used in Parkinson's disease but also in the treatment of hyperprolactinaemia-prolactinomas. Studies reveal that cabergoline is more effective and far better tolerated than bromocriptine in the treatment of prolactinomas.

Indications

Some patients not tolerating or not responding to bromocriptine have been successfully treated with cabergoline. It is preferred for treatment of hyperprolactinaemia and acromegaly.

Dosage

Cabergoline has a particularly long half-life unlike bromocriptine and is administered twice weekly. It is administered orally in the dosage of 0.25 mg twice weekly. If required, it can be increased after every 4–8 weeks up to a maximum of 1 mg twice weekly.

Contraindications

The drug is contraindicated in the following conditions:
❑ Patients with uncontrolled hypertension
❑ Known hypersensitivity to ergot derivatives.

Side Effects

Incidence of complications such as nausea and vomiting are also lower with cabergoline in comparison to bromocriptine. It can also result in hypotension. Although there is no evidence that therapy with dopamine agonists is harmful during pregnancy, as soon as patients find out that they are pregnant, the dopamine agonist is stopped. In women with prolactin secreting tumours, desiring pregnancy,

cabergoline is not licensed for use. However, bromocriptine may be sometimes used in conditions where benefits appear to exceed the risks.

Anaesthetic Drugs

Local Anaesthesia

Local anaesthetic agents block the smaller (less than 1 micrometre in diameter) unmyelinated C fibres first, then the myelinated B fibres (1–3 micrometres) and finally the larger A fibres (1–20 micrometres). Temperature and pain sensation are carried by the small C fibres, whereas proprioception is carried by A-alpha fibres. Therefore, temperature is blocked before proprioception. Also, unmyelinated fibres are theoretically blocked first than the myelinated fibres. Also, the postganglionic fibres are blocked before preganglionic fibres.

Local anaesthetic agents cause reverse blockage of nerve conduction. Although the action is local, side effects occur as a result of systemic absorption or accidental intravascular injection. Some of these side effects include convulsive excitation of CNS followed by depression and cardiovascular depression with hypotension and arrhythmias. All local anaesthetic agents are capable of crossing the placenta. Many local anaesthetics cause local vasodilatation and are often used in combination with adrenaline (1:80,000 to 1: 800,000).

Local anaesthesia combined with adrenaline should be avoided in patients taking tricyclic antidepressants. The total dose of adrenaline combined with local anaesthesia should not exceed 500 µg in a single operation. It is essential not to exceed a concentration of 1 in 200,000 (5 µg/mL). Characteristics of local anaesthetic agents are described in **Table 12.15**.

Lidocaine

Lidocaine (or lidocaine) is an amide type local anaesthetic, and is also classified as a Vaughan Williams class I antiarrhythmic drug (not class IV). It has no intrinsic vasoconstrictor activity and can cross the blood brain barrier. Lidocaine is associated with the inhibition of the conduction of neuronal impulses rather than inhibition of the impulse generation. Lidocaine has a stabilising effect on central neurones (sodium channel blockade) and has previously been used therapeutically to control seizure activity. It an effective local anaesthetic agent when used topically (e.g. urinary catheterisation).

It is a versatile local anaesthetic agent, which is good both for surface application as well as injection and is available in a variety of forms. When injected around a nerve it blocks conduction within 3 minutes. Vasodilatation occurs in the injected area. It is used for surface application, infiltration, nerve block, epidural, spinal and intravenous regional block anaesthesia.

TABLE 12.15	Characteristics of local anaesthetic agents
Name of the local anaesthetic	**Properties**
Lidocaine	Described separately in text
Bupivacaine	Bupivacaine is the drug most widely used in epidural anaesthesia (duration of 2–3 hours). It is usually used at strength of 0.25%, although higher concentrations may be used. It is highly protein-bound, thereby associated with limited transplacental exchange. Action starts slowly (within 30 minutes) but lasts for a longer duration of time (8 hours).
Mepivacaine	Does not cause vasodilatation and is therefore used in spinal anaesthesia. It does not cause vasodilatation.
Prilocaine	Similar to lignocaine
Benzocaine	Weak agent used for providing surface analgesia in mouth and anus
Ropivacaine	A newer bupivacaine congener, equally long-acting but less cardiotoxic. Continuous epidural ropivacaine is being used for relief of postoperative and labour pain. It can also be employed for nerve blocks.

Side Effects

Early central effects of lidocaine are depressant, i.e. drowsiness, mental clouding, dysphoria, altered taste and tinnitus. Overdose of this drug causes muscle twitching, convulsions, cardiac arrhythmias, fall in BP, coma and respiratory arrest. It is also associated with methaemoglobinaemia. Convulsions which are a side effect of systemic absorption of lignocaine can be controlled with thiopentone and adequate oxygenation.

General Anaesthesia

The most important procedure for general anaesthesia includes induction with an intravenous agent and maintenance with an inhalation via face mask, laryngeal mask or endotracheal tube. An opioid analgesic agent is also added. In pregnant women, endotracheal intubation is usually performed after 15–20 weeks of gestation. This helps in minimising the risk of aspiration. Use of muscle relaxants helps facilitate endotracheal intubation and intraoperative ventilation. At the end of surgery, muscle relaxation is reversed using a combination of neostigmine and glycopyrronium (or atropine). Premedication with drugs such as anticholinergic agents (e.g. atropine, hyoscine, etc.); opioid analgesics (e.g. morphine, pethidine, etc.); benzodiazepines (diazepam, temazepam, lorazepam, etc.); phenothiazines (e.g. promethazine); etc. are usually avoided in pregnancy. Antacid prophylaxis using sodium citrate and ranitidine can be administered during pregnancy.

Most general anaesthetic agents are lipid soluble and therefore can freely cross the placenta. Due to this the opioids are usually not administered to the mother at the time caesarean delivery before the delivery of the foetus.

On the other hand, most of the commonly used muscle relaxants, being highly ionised cannot cross the placenta. Classification of the general anaesthetic agents is described in **Table 12.16**.

Thiopentone: Thiopentone is an excellent and most commonly used induction agent. It is an ultrashort acting thiobarbiturate, highly soluble in water yielding a very alkaline solution, which must be prepared freshly before injection. It produces unconsciousness in 15–20 seconds. Extravasation of the solution or inadvertent intraarterial injection produces intense pain; necrosis and gangrene can occur. It has a narrow therapeutic margin, so cardiorespiratory depression may occur. Recovery occurs relatively slowly. Its use should be avoided in cases of porphyrias. Thiopentone is a poor analgesic. Painful procedures should not be carried out under its influence unless an opioid or N_2O has been administered.

Side effects: Shivering and delirium may occur at the time of recovery with this drug. Pain in the postoperative period is likely to induce restlessness. Therefore, adequate analgesia should be provided.

Halothane: It is a volatile liquid with sweet odour, non-irritant and non-inflammable. It is a potent anaesthetic—precise control of administered concentration is essential. Induction is slow and the speed of recovery is also increased. It is not a good analgesic or muscle relaxant, but it potentiates the action of competitive neuromuscular blockers. It is rarely used nowadays. It causes smooth muscle relaxation, thereby resulting in hypotension and uterine atony. Therefore, it can be used for the manipulation of foetus at the time of version and for the manual removal of the placenta. It can also result in an increased risk for postpartum haemorrhage. It sensitises the heart to catecholamines, causing arrhythmias.

Side effects: It can cause side effects such as cardiac arrhythmias (ventricular extrasystoles) and liver toxicity. It is a triggering agent for malignant hyperthermia. Repeated use can also result in liver damage (halothane hepatitis) and reduced urine formation.

Nitric oxide: It is an odourless gas with low potency. It is not used as a sole anaesthetic agent. It causes an increase in the activity of sympathetic nervous system. It is highly insoluble in blood and other tissues due to which rapid induction and emergence occurs during the time of anaesthesia. When NO is discontinued, it diffuses from the blood to the alveoli, thereby reducing the concentration of oxygen in the alveoli (diffusional hypoxia). Therefore, at the time of recovery from NO, 100% oxygen must be administered. A 50% concentration of NO is commonly used for providing analgesia in labour. It acts as an effective analgesic agent at doses which are too low to cause unconsciousness. Administration of NO is normally not associated with any adverse effects to the foetus or the embryo. However its use during delivery may result in neonatal depression.

TABLE 12.16	Classification of general anaesthetic agents

Inhalational
Gas
- Nitrous oxide

Volatile liquids
- Ether
- Halothane
- Isoflurane
- Desflurane
- Sevoflurane

Intravenous
Fast acting drugs
- Thiopentone sodium
- Methohexitone sodium
- Propofol
- Etomidate

Slower acting drugs
- Benzodiazepines
  - Diazepam
  - Lorazepam
  - Midazolam
- Dissociative anaesthesia
  - Ketamine
- Opioid analgesia
  - Fentanyl

Isoflurane: It is the most widely used volatile anaesthetic drug. It is a potent cardiac vasodilator and can cause hypotension.

Regional Anaesthesia

Epidural anaesthesia may lead to a higher rate of forceps delivery. Due to relaxation of the levator ani in these cases, it takes longer for the foetal head to descend and rotate after full dilatation. Epidural block is contraindicated in ante-partum haemorrhage due to the dual risk of hypotension, but is useful in the management of pre-eclampsia. Spinal anaesthesia may be complicated by maternal respiratory difficulties if the spinal block rises to too high a level. Headache is more common in spinal than epidural anaesthesia.

Neuromuscular Blocking Agents

These drugs are used as an adjunct to anaesthetics to provide muscular relaxation at the time of surgery. Skeletal muscle relaxants are drugs that act peripherally at neuromuscular junction/muscle fibre itself or centrally in the cerebrospinal axis to reduce muscle tone and/or cause paralysis. Neuromuscular blocking agents can be classified as follows:

❑ *Non-depolarising (competitive) blockers*
  1. *Long acting:* d-Tubocurarine, pancuronium, doxacurium, pipecuronium
  2. *Intermediate acting*: Vecuronium, atracurium, cisatracurium, rocuronium
  3. *Short acting*: Mivacurium

TABLE 12.17	Differences between non-depolarising and typical depolarising block	
Characteristics	Competitive (Non-depolarising block)	Depolarising (phase 1) block
Mechanism of action	Act by decreasing the frequency of channel opening that result in an action potential	Depolarising the membrane by opening sodium channels
Paralysis in man	Flaccid	Fasciculations → flaccid
Human neonates	More sensitive	Relatively resistant
Tetanic stimulation during partial block	Poorly sustained contractions	Well-sustained contractions
Neostigmine	Antagonises block	No effect
Post-tetanic potentiation	Present	Absent
Ether anaesthesia	Synergistic	No effect
Order of paralysis	Fingers, eyes → limbs → neck, face → trunk → respiratory	Neck, limbs → face, jaw, eyes, pharynx → trunk → respiratory
Effect of lowering temperature	Reduces block	Intensifies block

❏ *Depolarising blockers*
  1. Succinylcholine (SCh., Suxamethonium), decamethonium

Based on the mechanism of action, neuromuscular blocking agents can be of two types: depolarising (e.g. succinylcholine) and non-depolarising (e.g. pancuronium) blockers. Difference between the two is described in **Table 12.17**. The clinical conditions which are known for prolonging or potentiating the non-depolarising neuromuscular blockade include the following:
❏ Hypokalaemia
❏ Hypocalcaemia
❏ Hypermagnesaemia
❏ Metabolic alkalosis
❏ Respiratory acidosis
❏ Hypothermia.

Numerous drugs are also associated with prolonged neuromuscular blockade. These include the following:
❏ Antibiotics such as streptomycin, polymyxin and neomycin
❏ Cocaine, procaine and lidocaine
❏ Lithium due to its hypokalaemic effect.

Succinylcholine: Suxamethonium is an intravenous depolarising muscle relaxant which rapidly induces neuromuscular paralysis. Muscular fasciculations lasting several seconds may be seen prior to paralysis. Recovery is spontaneous following its metabolism by the enzyme plasma pseudocholinesterase, which is synthesised in the liver. Succinylcholine is the most commonly used muscle relaxant for passing tracheal tube. It induces rapid, complete and predictable paralysis with spontaneous recovery in approximately 5 minutes. Duration of depolarisation is prolonged due to its slow breakdown. It may be used for rapid sequence induction to allow for rapid intubation (e.g. caesarean section). No antagonist is available for this agent.

Pesticides and various drugs (echothiophate iodide) are known to inhibit cholinesterase activity and thus prolong the action of suxamethonium. Reduced levels of plasma cholinesterase are seen in liver disease, malnutrition and pregnancy, which increase its duration of action. A variety of genetically determined types of abnormal enzyme have been identified which also prolong the action of suxamethonium, e.g. prolonged paralysis may occur in patients with low or atypical plasma pseudocholinesterase. Neostigmine and other anticholinesterase drugs potentiate the neuromuscular block induced by suxamethonium and have no role as reversal agents.

Side effects: It has a tendency to cause side effects such as muscle fasciculations and soreness, changes in BP and HR, arrhythmias, postoperative myalgia, histamine release and K^+ efflux from muscles resulting in hyperkalaemia and its complications. It should, therefore, be avoided in patients with heart disease, trauma and burns. Suxamethonium is a potent trigger for both anaphylaxis and malignant hyperpyrexia. Malignant hyperthermia can occur due to the release of calcium from the sarcoplasmic reticulum of skeletal muscles. Malignant hyperthermia is treated with dantrolene which inhibits the release of calcium.

Drugs for Thyroid Abnormalities

Hypothyroidism may be caused by various drugs listed in **Table 12.18**.

Antithyroid Drugs

Antithyroid drugs or thioamides help in inhibiting thyroid synthesis. These drugs include propylthiouracil (PTU),

TABLE 12.18	Drugs causing hypothyroidism

• *Amiodarone*: a typical drug associated with both hypothyroidism and hyperthyroidism
• Lithium
• Cobalt
• Iodides
• Butazolidin
• Sulphonylureas
• Antithyroid drugs

carbimazole, methimazole, etc. These drugs typically inhibit the synthesis of thyroid hormones (T4) by preventing iodination of tyrosine residues. Thiomides cross the placenta: PTU less than carbimazole. Thioamides are used in thyrotoxicosis for variable periods from 6 months up to 2 years, but irrespective recurrence of thyrotoxicosis following withdrawal is of the order of 70%. In high doses, thioamides may cause foetal hypothyroidism and goitre if administered during pregnancy. Therefore, lowest possible dosage to maintain free thyroxine levels in the normal range must be used. These drugs are safe during breastfeeding, although maternal thyroid function tests should be checked if high doses are to be used.

Propylthiouracil

Propylthiouracil (PTU) inhibits the synthesis of thyroid hormones by blocking the incorporation of iodine into tyrosine. PTU also inhibits coupling of iodotyrosines and prevents the conversion of T4 to T3. PTU appears in breast milk in an insignificant amount.

Propylthiouracil crosses the placenta in high doses and may cause foetal goitre. Therefore, its lowest effective dose is used. Since PTU is more protein bound than carbimazole, lesser amounts of PTU is secreted in breast milk.

Carbimazole

Carbimazole is used in the treatment of thyrotoxicosis in pregnancy. It is not teratogenic and therefore can be used during pregnancy. It is normally secreted in the breast milk. However, it is not contraindicated during breastfeeding. Therefore, the lowest dose that may control the thyroid should be used. Patients who are on maintenance doses of carbimazole must not be switched to PTU during pregnancy.

Side effects: Gross teratogenesis has not been associated with carbimazole, although allegedly it has been found to be associated with aplasia cutis. It is suggested that this disorder may be more associated with thyrotoxicosis per se than the drugs. Bone marrow depression is a recognised side effect of carbimazole. Although agranulocytosis and neutropaenia are only rarely associated with carbimazole use, they constitute the most worrying side effect and practitioners need to be alert regarding this possibility. Other adverse events include pruritus, rash, cholestatic jaundice and alopecia. It is converted to methimazole, which can cause a serum-sickness-like adverse reaction including lymphadenopathy. Vasculitis is also rarely reported.

Teratogenesis

Teratogenesis can be defined as structural or functional dysgenesis/malformation of the foetal organs. It can present in form of congenital malformations with varying severity, intrauterine growth restriction, carcinogenesis, foetal death,

etc. Teratogenic influence is likely to be strongest in the period of early organogenesis. The most important body structures of the foetus are formed in the first 12 weeks after conception, usually 20–55 days after conception. Interference in this process results in a teratogenic effect.

Teratogenic influence is likely to be less strong if exposure to the drug is in the later period of foetal development. Though structural malformations are less likely to occur due to the exposure to the teratogen in later part of pregnancy, this can cause serious functional abnormalities, particularly of the neurobehavioural type.

Teratogenic Drugs in Pregnancy

❑ *Warfarin*: Warfarin is known to be teratogenic in the first trimester and can cause abnormalities such as intracerebral haemorrhage, nasal hypoplasia, stippling of the epiphyses, chondrodysplasia punctata and CNS abnormalities. Warfarin, unlike heparin has been shown to cause CNS damage in the foetus if given in the second and third trimesters. Warfarin is also not safe in the last 4 weeks of pregnancy because it crosses the placenta and causes haemorrhage.

❑ *Phenytoin*: Phenytoin can cause foetal hydantoin syndrome. Foetal hydantoin syndrome is a syndrome comprising of characteristic pattern of mental and physical birth defects, resulting from maternal use of the anticonvulsant drug, phenytoin during pregnancy. Some characteristic features of this syndrome may include features such as distinctive skull and facial features, growth deficiencies (prenatal and postnatal growth restriction), hypoplastic nails of the fingers and toes, and/or mild developmental delays. Other findings occasionally associated with this syndrome include cleft lip and palate, short nose with a broad flattened nasal bridge, microcephaly, hypertelorism, strabismus, low set or abnormally formed ears, and skeletal malformations particularly of the fingers or hands. Various craniofacial and digital abnormalities, together with more major anomalies (cardiac defects, cleft lip and palate), and neural tube defects have also been associated with maternal phenytoin ingestion during pregnancy. Folic acid supplements reduce the incidence of neural tube defects and should be given to patients taking phenytoin.

❑ *Diethylstilbestrol*: Causes carcinoma of the vagina and vaginal adenosis in young women.

❑ *Quinine*: Causes blindness and deafness by causing hypoplasia of optic nerve.

❑ *Thalidomide*: Causes phocomelia, which involves absence of the long bones of the upper and/or lower limbs, amongst other defects.

❑ *Lithium*: Lithium may cause cardiac abnormalities.

❑ *Chlorpropramide*: Chlorpropramide may cause neonatal hypoglycaemia.

❏ *Pseudoephedrine*: Gastroschisis is a recognised effect of pseudoephedrine.

❏ *Irradiation*: Leukaemia, carcinoma of thyroid gland.

❏ *Valproate*: Valproate is associated with neural tube defects (extra folate should be prescribed in these cases).

❏ *Lisinopril*: This drug is teratogenic.

❏ *Losartan*: Losartan is contraindicated in pregnancy.

❏ *Statins*: Statins also are associated with teratogenicity.

❏ *Maternal hyperthermia*: This can result in CNS abnormalities in the foetus.

❏ *Glucocorticoids*: This can result in cleft lip.

❏ *Cyproterone acetate and possibly other 19-norsteroids*: Androgenisation of the female foetus.

❏ *Rifampicin*: Rifampicin is an extremely powerful enzyme-inducer, so the COCP is contraindicated in those taking rifampicin. For such individuals an alternative method of contraception is usually preferred. Even if rifampicin is taken only for a few days, its effect on the COC should be assumed to last for at least a month.

Besides being a powerful enzyme inducer, rifampicin has been found to be associated with an increased incidence of neural tube defects and facial clefts in experimental animals. Despite the concerns, no teratogenic effect has been proved in the human. There also have been concerns regarding its potential to cause bleeding in terms of PPH and haemorrhagic disease of the newborn. Therefore, vitamin K should be prescribed in these cases.

❏ *Isotretinoin*: Treatment with isotretinoin is a recognised indication for termination of the pregnancy. Contraception is advised for 2 years after cessation of treatment. If consumed during pregnancy, it can result in anomalies such as hypoplastic ears, malformation of the facial bones, cardiac defects, hydrocephalus, thymic hypoplasia, etc.

❏ *Thiazide diuretics*: These have been shown to decrease placental perfusion.

Antibiotics to be Avoided during Pregnancy

Antibiotics are among the most common drugs used in pregnancy. None of the drugs prescribed in pregnancy can be guaranteed as 100% safe, but a few are significantly teratogenic. Some of the antibiotics which must be avoided during pregnancy include the following:

❏ *Aminoglycosides*: Aminoglycosides are known to be ototoxic. Aminoglycosides (streptomycin, gentamycin, tobramycin, etc.) may cause auditory or vestibular damage, especially when prescribed in the second/third trimester.

❏ *Co-trimoxazole*: It causes neonatal haemolysis and methaemoglobinaemia. It is also teratogenic in the first trimester.

❏ *Tetracycline*: Tetracycline causes dental discolouration (if taken in the second/third trimester) and maternal hepatotoxicity in large doses. Tetracycline is best avoided during pregnancy, but is not "unsafe" if prescribed in limited doses during the first trimester.

❏ *Chloramphenicol*: Chloramphenicol may cause "grey baby syndrome" usually in the third trimester. This syndrome occurs because the neonate cannot metabolise the drug and, therefore suffers serious toxicity which is often fatal. The term "grey" refers to the description of the baby's appearance.

❏ *Ciprofloxacin*: This is a quinolone drug linked to arthropathy in animal experiments. Therefore, its use is restricted in pregnancy. However, the reports suggest that its use may be safe.

❏ *Nitrofurantoin*: This drug should be avoided in late pregnancy because of its links to neonatal haemolysis.

❏ *Trimethoprim*: Since this is a folate antagonist, there is concern that it may be a teratogenic if taken in early pregnancy.

❏ *Metronidazole*: Metronidazole has not been shown to be teratogenic. In therapeutic doses is relatively safe during pregnancy. However, there is still reluctance to use it, particularly in the first trimester.

❏ *Erythromycin*: Erythromycin is safe during pregnancy.

Safe Drugs during Pregnancy

Methyldopa is used for treatment of hypertension during pregnancy. It is the only hypotensive that is safe in all stages of pregnancy. Paracetamol is safe throughout a normal pregnancy.

There is no evidence that ranitidine, metformin, aspirin or the OCPs are teratogenic. Although it was once believed that aspirin and the OCPs were teratogenic if consumed during pregnancy, studies indicate otherwise. Similarly, metformin is often used in cases of PCOS to induce fertility through reduction in insulin resistance. Antibiotics such as erythromycin and methylpenicillin are also largely safe during pregnancy.

Cocaine and Pregnancy

Cocaine abuse results in intrauterine growth restriction (IUGR) by causing vasoconstriction of the uterine vessels. This leads to reduced uterine blood flow, foetal hypoxia and increased foetal blood pressure. Foetal vasospasm can also occur, causing loss of a digit or cerebral infarction. There is an increased incidence of early pregnancy loss, plus placental abruption and preterm labour. Unlike opiates, cocaine is not usually associated with neonatal withdrawal.

Alcohol Consumption and Pregnancy

Prenatal alcohol exposure also increases the risk of pregnancy related maternal and foetal complications **(Table 12.19)**. Maternal complications include increased risk of preterm labour, miscarriage, etc. Foetal complications

TABLE 12.19	Maternal and foetal complications associated with alcohol consumption during pregnancy
Maternal effects	**Foetal effects**
Miscarriage Preterm labour	Foetal alcohol spectrum disorders Foetal alcohol syndrome Growth restriction Neurodevelopmental anomalies

include the risk for foetal alcohol spectrum disorders, foetal alcohol syndrome, intraventricular haemorrhage (IVH) and damage to white matter in preterm neonates, etc.

Foetal Alcohol Spectrum Disorders

The term foetal alcohol spectrum disorders (FASD) has been proposed for inclusion in the Diagnostic and Statistical manual of mental disorders (DSM-IV). Prenatal alcohol exposure can result in a whole spectrum of CNS sequel that persists throughout the life span and manifests in form of a spectrum (ranging from mild to severe) of structural anomalies, behavioural and neurocognitive disabilities, in the foetus. Children at the severe end of the spectrum have been defined as having the most serious form of the disorder, also termed as foetal alcohol syndrome (FAS). The Institute of Medicine (IOM, 1996) developed five diagnostic categories **(Table 12.20)** for classifying alcohol related disorders. The first two categories pertain to FAS itself. The other categories address various aspects of the spectrum of alcohol-related disorders.

Category 1

Foetal Alcohol Syndrome with Confirmed Maternal Alcohol Exposure

Historically FAS has been defined by growth deficiency, a pattern of facial anomalies, and presence of brain dysfunction **(Fig. 12.2)**. The diagnostic criteria for category 1 FADS are defined as follows:

Maternal alcohol exposure: This is defined as a pattern of excessive alcohol intake characterised by substantial, regular intake or a heavy episodic (i.e. binge) drinking. Binge-drinking (more than 5 units) is probably a major risk factor. This pattern of drinking may be associated with the signs of alcohol dependence. Binge-drinking also increases the risk of sexually transmitted disease and unplanned pregnancy. Low level alcohol consumption may carry no risk. But presently no safe level has been identified below which harm cannot occur. The RCOG advices low intake of alcohol comprising of one or two units of alcohol once or twice a week. On the other hand, the Department of Health (DOH) has advocated zero alcohol consumption during pregnancy in May 2007. NICE, however, advocates alcohol abstinence in the first trimester because of the risk of miscarriage.

Besides the adverse effects of alcohol per se, excessive alcohol intake also leads to reduced blood folate levels.

TABLE 12.20	IOM-recommended diagnostic criteria for foetal alcohol spectrum disorders

Category 1: FAS with confirmed maternal alcohol exposure
- Confirmed maternal alcohol exposure
- Characteristic pattern of facial anomalies, including short palpebral fissures, and abnormalities of the premaxillary zone (e.g. flat upper lip, flattened philtrum, flat midface)
- Growth retardation, such as low birth weight, lack of weight gain over time, disproportional low weight to height
- Neurodevelopmental abnormalities of the CNS, such as microcephaly at birth; structural brain abnormalities with age-appropriate neurological hard or soft signs (e.g. impaired fine motor skills, neurosensory hearing loss, poor tandem gait, poor eye-hand coordination)

Category 2: FAS without confirmed maternal alcohol exposure
- No exposure to alcohol, rest of the features are same as category 1

Category 3: Partial FAS with confirmed maternal alcohol exposure
- Confirmed maternal alcohol exposure
- Characteristic pattern of facial anomalies (all anomalies may not be present)
- Growth retardation
- Neurodevelopmental abnormalities of the CNS
- Complex pattern of behavioural or cognitive abnormalities inconsistent with developmental level and unexplained by genetic background or environmental conditions (e.g. learning difficulties; deficits in school performance; poor impulse control; problems in social perception; language deficits; poor capacity for abstraction; specific deficits in mathematical skills; and problems in memory, attention, or judgement)

Category 4: Alcohol-related birth defects (ARBDs)
- Confirmed maternal alcohol exposure
- One or more congenital defects, including malformations and dysplasias of the heart, bone, kidney, vision, or hearing systems

Category 5: Alcohol-related neurodevelopmental disorder (ARND)
- Confirmed maternal alcohol exposure
- CNS neurodevelopmental abnormalities as in Category 1 and/ or
- Complex pattern of behavioural or cognitive deficits as in Category 3

Source: Committee to Study Foetal Alcohol Syndrome: Division of biobehavioural sciences and mental disorders. Foetal alcohol syndrome: Diagnosis, epidemiology, prevention, and treatment. Stratton K, Howe C, Battaglia F (Eds). Institute of medicine. National academy press: Washington, D.C.; 1996.
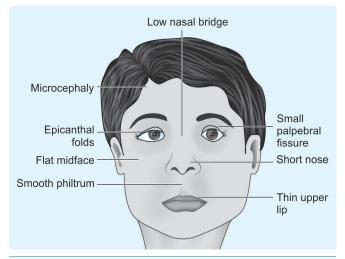
FIG. 12.2: Characteristic features of foetal alcohol syndrome

This could have implications for the developing foetus. Therefore, it is important to ensure that women take folic acid supplements in the dosage up to 5 mg daily.

There are blood tests which could be used to screen for alcohol abuse include gamma glutamyl transpeptidase (GGT) and carbohydrate-deficient-transferrin (CDT). The most commonly used screening tool for detecting alcohol abuse in antenatal clinics is T-ACE questionnaire, though at some places CAGE questionnaire is also used. Both the questionnaires are acronyms for a set of four questions which must be enquired from the patient for alcohol screening. These questionnaires, T-ACE and CAGE, are described respectively in **Tables 12.21 and 12.22**.

TABLE 12.21	T-ACE questionnaire for alcohol screening
1. How many drinks does it take to make you feel high? 0. less than or equal to 2 drinks 1. more than 2 drinks	**Tolerance**
2. Have people annoyed you by criticising your drinking? 0. No 1. Yes	**Annoyance**
3. Have you felt you ought to cut down on your drinking? 0. No 1. Yes	**Cut Down**
4. Have you ever had a drink first thing in the morning to steady your nerves or to get rid of a hangover? 0. No 1. Yes	**Eye Opener**

Total Score = _____

The T-ACE score has a range of 0–5. The value of each answer to the four questions is totalled to determine the final T-ACE score. A total score of 2 or greater is considered to be clinically significant.

Source: Robert JS. Finding the risk drinker in your clinical practice. In: Robinson G, Armstrong R (Eds). Alcohol and Child/Family Health: Proceedings of a Conference with Particular Reference to the Prevention of Alcohol-Related Birth Defects. Vancouver, BC.; 1988.

TABLE 12.22	CAGE questionnaire for alcohol screening
1. Have you ever felt you needed to cut down on your drinking? 0. No 1. Yes	**Cut**
2. Have people annoyed you by criticising your drinking? 0. No 1. Yes	**Annoyance**
3. Have you ever felt guilty about drinking? 0. No 1. Yes	**Guilty**
4. Have you ever had a drink first thing in the morning to steady your nerves or to get rid of a hangover? 0. No 1. Yes	**Eye Opener**

Total Score = _____

The CAGE score has a range of 0–5. The value of each answer to the four questions is totalled to determine the final CAGE score. A total score of 2 or greater is considered to be clinically significant.

Dysmorphia: Human congenital malformations are referred to as dysmorphic features or dysmorphia. The individual to be diagnosed with FAS must exhibit all three characteristic facial dysmorphic features: (1) smooth philtrum, (2) thin vermillion upper lip border, and (3) small palpebral fissures. Though the three above-mentioned dysmorphic features must definitely be present for the diagnosis of FAS, other additional dysmorphic features like microcephaly, short-nose, flattened nasal bridge, micrognathia, maxillary hypoplasia (with prognathism), presence of epicanthal folds **(Fig. 12.3)**, altered palmar fexional crease patterns (i.e. hockeystick crease), cardiac anomalies, joint disability, overlapping fingers, ear anomalies, haemangiomas, ptosis, hypoplastic nails, pectus deformities, cleft lip, micrognathia, protruding auricles, short or webbed neck, vertebra and rib anomalies, short metacarpal bones, meningomyelocele, hydrocephalus, hypoplastic labia majora, etc. may also be present. American Academy of Paediatrics has reported the rate of microcephaly to be more than 80% amongst the women consuming alcohol. After puberty, the characteristic facial features associated with FAS can become more difficult to detect.

FIG. 12.3: Epicanthal fold severity guide

Category 2

FAS without Confirmed Maternal Alcohol Exposure

If it is not known for sure that there was alcohol exposure during pregnancy even then a diagnosis of FAS can be made if the affected person appears to have all the signs of FAS.

Category 3

Partial FAS

Category 3 includes partial FAS with confirmed maternal alcohol exposure. In other words for diagnosis of this category, some, but not all, of the facial characteristics required for diagnosis of FAS must be present as well as there should be a confirmed evidence of maternal alcohol exposure. In addition, at least one of the three following indicators also must be present: Growth deficits normally characteristic of FAS, neurodevelopmental abnormalities, or behavioural and cognitive problems consistent with those observed in FAS. The cognitive abnormalities could be in the form of complex pattern of deficits in learning, school performance, impulse control, behavioural pattern, etc.

Growth restriction: Evidence of growth restriction is there if at least one of the following is present:
❑ Low-birth weight for gestational age
❑ Decelerating weight over time, which is not related to poor nutrition
❑ Disproportional low weight to height.

Neurodevelopmental and behavioural or cognitive abnormalities: Would be described later.

Category 4

Alcohol-related Birth Defects

Category 4 encompasses alcohol-related birth defects (ARBDs) and includes people who had been prenatally exposed to alcohol and were diagnosed with heart, bone, kidney, visual, or hearing defects. Although these anomalies may not be consistently observed, they are not uncommon either.

Category 5

Alcohol-related Neurodevelopmental Disorder (ARND)

Category 5 is defined when the individual shows behavioural and cognitive problems similar to those seen in FAS and partial FAS, but the facial features are normal. The various neurodevelopmental and cognitive abnormalities which can be present are described below:

Neurodevelopmental anomalies: Children exposed to alcohol in utero may suffer from serious cognitive effects and behavioural problems as well as alcohol-related changes in brain structure which can be identified by modern imaging techniques. Brain mapping based on MRI analysis suggests disproportionate reduction in white matter compared with grey matter in these individuals. Children with foetal alcohol syndrome have a much smaller brain size, with specific reductions in the size of the caudate nucleus, thinning or agenesis of the corpus callosum and reduced size of the hippocampus, and cerebellum.

Alcohol-related neurodevelopmental disorder may be related to other factors including, the timing of exposure, high levels of alcohol consumption, genetic factors affecting maternal or foetal metabolism or individual susceptibility, etc. Evidence of CNS neurodevelopmental abnormalities is believed to be there, if at least one of the following is present:
❑ Decreased cranial size at birth
❑ Structural brain abnormalities (e.g. microcephaly, partial or complete agenesis of the corpus callosum, cerebellar hypoplasia, etc.)
❑ Age appropriate neurological hard or soft signs such as impaired fine motor skills, neurosensory hearing loss, poor tandem gait, poor eye–hand coordination etc.

Cognitive and behavioural disturbances: Variety of behavioural and cognitive features have been proposed as indicators of brain dysfunction in FAS. Some of these include poor performance on tests of intelligence and educational achievement, impaired language development, poor impulse control, and problems with memory and judgement. At present, however, no consensus has been achieved as to which features are most appropriate for the diagnosis of FAS.

These features too are considered to be on a spectrum, ranging from near normal to severely impaired. They are also influenced by other factors such as parental intelligence, educational experience and various social (impoverished postnatal environment) and cultural influences. Furthermore, these cognitive and behavioural features are less specific to FAS than are the physical features.

Not only these features tend to change with time, they also tend to occur in association with a wide range of other childhood neurodevelopmental and psychiatric conditions, attention-deficit hyperactivity disorder, etc.

Foetal Behavioural Studies

Maternal alcohol consumption may also have an effect on spontaneous movements, startle reaction and habituation of the foetus at 18, 27 and 36 weeks of gestation.

Attention and Hyperactivity Problem

Attention problems are often noted for children with FAS, with many children receiving a diagnosis of attention-deficit hyperactivity disorder (ADHD).

Drugs Secreted in Breast Milk

Drugs which are secreted into the breast milk can be divided into three categories:

1. *Those drugs which are undetectable in the baby*: These include drugs which are bound to maternal proteins (e.g. warfarin) or those which are not absorbed from the baby's gut (e.g. aminoglycosides).
2. *Drugs which reach the baby in insignificant amounts*: Since these drugs reach the baby in insignificant amounts, they are usually not harmful to the baby. Some of the examples include non-narcotic analgesic drugs, penicillin and cephalosporin antibiotics and antihypertensive drugs.
3. *Drugs which reach the baby in sufficient amounts so as to cause harm*: These drugs are contraindicated during puerperium and include drugs such as laxatives, barbiturates, lithium, cytotoxics, immunosuppressive drugs, etc.

Drugs Contraindicated during Lactation

Drugs contraindicated during lactation are listed in **Table 12.23**. Some of these are described in details next:

Bromocriptine and cabergoline: Both bromocriptine and cabergoline cause suppression of lactation. However, once lactation has been established, bromocriptine is not very effective in suppressing it. The British National Formulary states that although licensed, bromocriptine and cabergoline should not be routinely used for suppressing lactation during the puerperium. Pain and breast engorgement can be relieved with analgesics and good support. If a drug has to be used, cabergoline is preferred over bromocriptine. It has some very serious side effects; fortunately these are rare. They include hypotension, hypertension, stroke, myocardial infarction and psychosis.

TABLE 12.23	Drugs likely to have deleterious effects during lactation

- Amantadine
- Amiodarone
- Antineoplastic drugs
- Atropine
- Barbiturates
- Benzodiazepines
- Bromide
- Carbimazole
- Chloramphenicol
- Ephedrine
- Ergotamine
- Iodide
- Lithium
- Phenindione
- Primidone
- Radioactive agents
- Streptomycin
- Sulphonamides

Sulphonamides: Sulphonamides may worsen neonatal jaundice and cause haemolysis in the babies with glucose-6-phosphatase deficiency.

Labetalol: Labetalol appears to cross to the baby in insufficient quantities to be able to cause problems of beta-blockade. Nevertheless, the baby should be monitored if the mother is taking labetalol.

Tetracycline: Tetracycline if taken in pregnancy may cause discoloration of the baby's teeth. It is thought that it is unlikely to do so via breast milk, but is best avoided to be safe.

Methyldopa: Methyldopa is contraindicated during puerperium to avoid exacerbating postpartum depression (depression being a side effect of the drug). However, the amount secreted in breast milk is too small to affect the baby.

Androgens: Androgens may cause masculinisation in the female infant or precocious development in the male infant. Therefore, they must not be used during puerperium.

Thiazide diuretics: Thiazide diuretics in large doses may reduce or inhibit milk production.

Drugs Which can be Used during Breastfeeding

Propranolol: Propranolol is secreted in breast milk and may be rarely associated bradycardia but generally has little impact. Therefore, it can be administered to breastfeeding mothers. Progesterone only contraceptive pills are also not contraindicated in breastfeeding mothers.

Warfarin: Warfarin is secreted in small amounts in breast milk and has little impact on foetal coagulation.

Ranitidine: Significant amounts are present in breast milk but it is not known to be harmful during breastfeeding.

Antihypertensive agents: Angiotensin-converting enzyme (ACE) inhibitors may be used safely during the puerperium. They can be used if they were used preconceptually or they can be used for the first time. There is also no contraindication to the use of beta-blockers during puerperium.

Although antihypertensive agents such as methyldopa, nifedipine or labetalol excreted into breast milk, no adverse effects on breast-fed infants exposed to these drugs have been reported. Therefore, these antihypertensive agents can be prescribed in puerperium.

Metronidazole: Metronidazole is secreted in significant amounts in breast milk. Manufacturer advises avoiding single large doses. In practice at normal doses it appears to be safe and is widely used.

Drugs in Gynaecology

Clomiphene Citrate

Clomiphene citrate is a non-steroidal triphenylethylene derivative, acting as selective oestrogen receptor modulator (SERM) having both oestrogen antagonist and agonist effects.

Mechanism of Action

Whether CC would act as an agonist or an antagonist depends upon the prevailing levels of endogenous oestrogens in the body. If the levels of endogenous oestrogens are too low, oestrogen agonist properties of the drug are manifested. Otherwise in presence of normal levels of endogenous oestrogens, antioestrogenic properties are exhibited. Clomiphene citrate is largely believed to exert its antioestrogen effect by competing with the oestrogen receptors at the level of hypothalamus, pituitary and ovaries. By blocking the oestrogen receptors within the hypothalamus, CC alleviates the negative feedback effect exerted by endogenous oestrogens. As a result, the gonadotropin-releasing hormone (GnRH) release gets normalised. Therefore, the secretion of FSH and LH is able to re-establish the normal process of ovulation and is capable of normalising follicular recruitment, selection and development. Therefore, this drug acts to indirectly induce ovulation.

Indications

❑ Women in whom the cause of infertility is anovulation or anovulatory disturbances
❑ Women with unexplained fertility problems
❑ Polycystic ovarian disease
❑ Anovulatory dysfunctional uterine bleeding
❑ Harvesting oocytes before an IVF cycle
❑ Oligospermia (in males).

Dosage

It is usually administered orally using the following protocols:

Standard protocol: The standard dose of CC is 50 mg PO once a day for 5 days, starting on the days 3–5 of the natural spontaneous menstrual cycle (follicular phase) or after progestin-induced bleeding. In case there is no response with 50 mg dosage, it can be increased up to a dosage of 250 mg/day. The dosage is increased in the increments of 50 mg every 5 days. The response to CC is monitored using pelvic ultrasonography starting on the day 12 of the menstrual cycle. The follicle should develop to a diameter of 23–24 mm

before a spontaneous LH surge occurs. The UK Committee on the Safety of Medicines (CSM) has recommended that clomiphene should not normally be used for longer than six cycles.

Extended protocol: Therapy with CC in the dosage varying between 50 mg and 250 mg/day is administered over 8–10 days. This protocol, however, can be associated with significant side effects and so it is not commonly used.

Combination protocols: These protocols are usually used in women who are resistant or refractory to standard treatment with CC. The following combinations can be offered to such women:
❑ CC + hCG (5,000–10,000 IU).
❑ CC + insulin sensitising agents (e.g. metformin, 1,000–2,000 mg/day in divided doses).
❑ CC + glucocorticoids (0.5 mg dexamethasone or 5 mg prednisolone daily at night).
❑ *Sequential CC with gonadotropins*: CC is administered in the dosage of 50–100 mg/day on days 5–9 of the cycle. This is immediately followed by low dose of FSH (e.g. 75 IU/day) on days 9–12 of the cycles.

Contraindications

Clomiphene citrate is contraindicated in the following conditions:
❑ Ovarian cysts/tumours
❑ Ovarian failure
❑ Hepatic disease/dysfunction
❑ Occurrence of visual disturbances
❑ Hormone dependent tumours
❑ Undiagnosed abnormal uterine bleeding
❑ Pregnancy
❑ Previous history of breast cancer (use is controversial).

Side Effects

The main risk for the women who are being prescribed CC is that this drug may be associated with the risk of multiple pregnancies (from about 1% to more than 5%). It may sometimes be associated with ovarian cyst formation and its rare, but extreme variant, ovarian hyperstimulation. Ovarian hyperstimulation, which is rare, can be a life-threatening complication with huge cysts, ascites and fluid balance disturbance.

Women undergoing treatment with CC should be offered ultrasound monitoring during at least the first cycle of treatment to ensure that they receive a dose that minimises the risk of multiple pregnancy. Another important adverse effect associated with the use of this drug is the thickening of the cervical mucus under the antioestrogenic effect of CC. This may create an iatrogenic cervical factor, which may be responsible for producing infertility in a patient who has otherwise ovulated. Other adverse effects, which may be associated with CC, include hot flushes (vasomotor

flushes), visual disturbances, scotomas, breast tenderness, nausea, vomiting, abdominal discomfort, depression, insomnia, headache, intermenstrual spotting, menorrhagia, endometriosis, convulsions, weight gain, rashes, dizziness, hair loss, dryness of the vagina, etc. There is no evidence that this drug is teratogenic. It is believed that absence of ovulation due to pregnancy, lactation, breastfeeding, the pill, etc. is protective with respect to ovarian cancer. Therefore, by inducing ovulation, clomiphene becomes a theoretical risk factor. There is a possible increased risk of ovarian cancer in patients treated for longer time durations. No increased risk of ovarian cancer was found by Jensen et al. in a Danish population based cohort study, published in the BMJ (2009), in a large series of women who had been treated with fertility drugs.

LHRH Analogues

Introduction

Luteinising hormone releasing hormone (LHRH) analogues are also known as GnRH analogues, GnRH being the superior term. This is a synthetic peptide molecule which interacts with the GnRH receptors to elicit the biological response by stimulating the release of pituitary hormones FSH and LH. In nature, LHRH, a decapeptide, (not a steroid) is produced by the hypothalamus and released in 90-minute pulses.

On the other hand, LHRH synthetic analogues are produced by substituting one or more of the peptides. The resulting drugs are more potent and longer-lasting than the natural hormone. While the physiological release of GnRH is in pulses, these agonists act continuously. Therefore, soon following the initial episode of stimulation, GnRH analogues produce a hypogonadotropic-hypogonadic state by inhibiting the secretion of gonadotropins by causing the downregulation of pituitary gland. The difference between the menopause like state produced by use of these drugs and the real menopause is that analogues produce their effect by inducing very low levels of FSH and LH, whereas the real menopause is associated with high levels of FSH and LH.

GnRH analogues do not have any effect on the hormones released by posterior pituitary, e.g. oxytocin and anti-diuretic hormone.

Mechanism of Action

After an initial episode of stimulation, by eventually causing the downregulation of pituitary gland, GnRH agonists produce a hypogonadotropic-hypogonadic state. They also act by inhibiting the mid-cycle FSH and LH surge and preventing steroidogenesis in the corpus luteum. Spermatogenesis or ovulation comes to an end and testosterone or oestradiol levels fall to the castration levels. Recovery usually occurs within 2 months of stopping treatment. Currently, goserelin and leuprolide acetate are the most commonly used GnRH agonists.

Indications

❑ *Endometriosis*: Efficacy of GnRH agonists for providing pain relief is similar to that of danazol. Treatment is usually restricted to monthly injections for 6 months.
❑ *Fibroids*: These drugs can be used for causing temporary shrinkage in the size of fibroids. However, these grow again when the treatment has been stopped. Therefore, they are not really an alternative to myomectomy. Nonetheless, the drugs may be useful for reducing the size of fibroids prior to myomectomy or hysterectomy, making the surgery simpler.
❑ *Oestrogen-dependent disorders*: These include conditions such as menorrhagia, adenomyosis or uterine fibroids. GnRH agonists are effective in these cases by producing a hypoestrogenic state.
❑ *Precocious puberty*: GnRH agonists are useful in treating precocious puberty in both boys and girls.
❑ *Ovarian stimulation protocols*: In these cases, GnRH agonists produce a hypoestrogenic state by causing the downregulation of hypothalamo-pituitary axis. FSH is then administered followed by hCG to trigger ovulation.

Route of Administration

Intramuscular/subcutaneous/intranasal/implants: Most of these drug preparations are expensive and are administered by depot injection or nasal spray. There are administered using specially designed microprocessor-driven pumps. Some GnRH agonist preparations are also available in the form of implants. The action of implants can last from 1 month to 12 months. Injectable preparations are formulated for daily, monthly or quarterly use.

Dosage

Some examples of GnRH analogues include leuprolide, buserelin, zoladex, synarel, etc. Leuprolide is used in the dosage of a single monthly 3.75 mg depot injection given intramuscularly. Goserelin, in a dosage of 3.6 mg, is administered subcutaneously every 28 days. The dose of goserelin is 3.6 mg SC q28d or 10.8 mg SC q12wk for 6 months. A nasal spray of nafarelin (synarel) is also available and is used in the dosage of 400 µg twice daily.

Contraindications

❑ Pregnancy/lactation
❑ Hypersensitivity to the constituents of a GnRH preparation.

Side Effects

Similar to danazol, use of GnRH agonists may result in hypoestrogenic side effects. Their use is specifically associated with the loss of trabecular bone density, which is restored by 2 years after cessation of therapy. Other

prominent adverse effects include hot flushes, mood swings, breakthrough bleeding, reduced libido, depression, vaginal dryness, etc.

There has been much recent research regarding whether the simultaneous use of add-back therapy would be helpful in preventing osteoporosis and other hypoestrogenic symptoms associated with the use of GnRH agonists. Various agents which can be used as the add-back therapy include hormone replacement therapy preparations (a combination of conjugated oestrogen 0.625 mg or 1.25 mg with norethindrone 5 mg/day), OCPs (containing ethinyl oestradiol and desogestrel), progestins (norethindrone 5–10 mg/day), tibolone maleate, bisphosphonates, etc. Use of add-back therapy does seem to reduce some of the hypoestrogenic side effects associated with the use of GnRH agonists, thereby allowing longer duration of therapy with these agents.

Hypersensitivity reactions also have been sometimes reported. Ovarian hyperstimulation, resulting in polycystic ovary, lower abdominal pain, ovarian bleeding and shock can also occur in women who are administered GnRH agonists.

Danazol

Danazol is a synthetic androgen is the derivative of ethinyl testosterone. It has been shown to be highly effective drug for relieving the symptoms of endometriosis by inhibiting pituitary gonadotropins (FSH and LH).

Mechanism of Action

By inhibiting the pituitary production of gonadotropins, danazol may result in the development of a relative hypoestrogenic state. Endometrial atrophy is the likely mechanism, which provides relief from pain due to endometriosis. Danazol acts by inhibiting the mid-cycle FSH and LH surges and preventing steroidogenesis in the corpus luteum.

Indications

It is a highly effective drug for treatment of endometriosis.

Dosage

Danazol therapy is administered orally and is started when the patient is menstruating, usually on the 1st day of the menses. The initial dosage should be 800 mg/day, given in two divided oral doses, but this dosage can be titrated down as long as amenorrhoea persists and pain symptoms are controlled. Patients with less severe symptoms may be given 200–400 mg/day, in two divided oral doses. Treatment is usually administered for 6 months in cases of endometriosis, but can be extended to 9 months in responsive patients with severe disease.

Contraindications

Contraindications for the use of danazol include the following:
❑ Pregnant and lactating women
❑ Porphyrias
❑ Epileptic seizures
❑ Breast cancer
❑ Impaired renal/hepatic/cardiac function
❑ Undiagnosed abnormal uterine bleeding.

Side Effects

The adverse effects caused by danazol are primarily related to oestrogen deficiency and the androgenic effects. Oestrogen deficiency can result in symptoms such as headache, flushing, sweating, atrophic vaginitis and breast atrophy. Androgenic effects associated with danazol include acne, oedema, hirsutism, deepening of the voice and weight gain.

Mifepristone

It is a synthetic, 19-norsteroid compound having potent antiprogestational and significant antiglucocorticoid, and antiandrogenic activities. Mifepristone binds better to progesterone receptors in comparison to progesterone.

Mechanism of Action

It acts as a progesterone receptor antagonist due to which it serves as an abortifacient in the early pregnancy. When administered during the follicular phase, its antiprogestin action results in decrease of the mid-cycle gonadotropin surge from pituitary gland. This causes slowing of follicular development and delay/failure of ovulation. If administered during the luteal phase, it prevents secretory changes by blocking the action of progesterone on the endometrium. Later in the cycle, it blocks progesterone support to the endometrium, liberating the release of prostaglandins (PGs) from it, thereby stimulating uterine contractions. Mifepristone also sensitises the myometrium to PGs and induces menstruation. If implantation has occurred, mifepristone blocks decidualisation, thereby causing dislodgement of the conceptus. If hCG production falls, secondary luteolysis occurs resulting in the reduction of endogenous progesterone secretion and cervical softening. All these effects eventually result in miscarriage.

Mifepristone also serves as a powerful antagonist agent at the glucocorticoid receptors.

Indications

❑ *Termination of pregnancy*: For termination of pregnancy up to 7 weeks of gestation, 600 mg of mifepristone can be administered in the form of a single oral dose. This single dose regime is successful in nearly 60–85% cases.

During the first 7–8 weeks of pregnancy, administration of mifepristone alone is likely to result in pregnancy failure in up to 85% of cases.

To improve the success rate, mifepristone can be followed up to 48 hours later by a single oral dose of 400 mg misoprostol. This combination regimen is associated with a success rate of greater than 90% and is the accepted non-surgical method of early first trimester abortion. This combination is likely to result in complete abortion in about 95% of cases, incomplete abortions in about 4% cases and ongoing pregnancy or missed abortion in about 1%.

❏ *Cervical ripening*: Mifepristone, in the dosage of 600 mg, administered 24–30 hours before attempting surgical abortion or induction of labour, helps in cervical softening and the facilitation of the procedure of surgical abortion.

❏ *Postcoital/emergency contraception*: Mifepristone in the dosage of 600 mg administered within 72 hours of intercourse interferes with implantation. This is not the method of choice for postcoital contraception, particularly when more effective methods exist.

❏ *Once-a-month contraceptive agent*: A single dose of 200 mg mifepristone administered 2 days after mid-cycle each month prevents conception on most occasions.

❏ *Induction of labour*: Mifepristone helps in blocking the relaxant action of progesterone on uterus, late in pregnancy, thereby promoting labour. It may also be administered in cases with intrauterine foetal death and to deliver abnormal foetuses.

❏ *Cushing's syndrome*: Mifepristone has palliative effect due to glucocorticoid receptor blocking property and may be used for inoperable cases of Cushing's syndrome.

❏ *Other uses*: Other proposed uses of mifepristone include endometriosis, uterine fibroids, certain breast cancers and meningioma.

Dosage

It is most commonly administered via the oral route. Dosage of mifepristone used for the termination of pregnancy has been described in the "indications".

Contraindications:
❏ Presence of IUCD
❏ Ectopic pregnancy
❏ Adrenal failure
❏ Haemorrhagic disorders
❏ Inherited porphyrias
❏ Intake of anticoagulants
❏ Long-term corticosteroid therapy.

Side Effects

Side effects such as nausea, vomiting, diarrhoea, weakness, dizziness, bleeding or cramping may commonly occur. Blood transfusion may be required in less than 1% of cases.

Rarely, there may be symptoms of a serious allergic reaction, including rashes/itching/swelling (especially of the face/tongue/throat), severe dizziness, dyspnoea, etc. Other side effects, such as infection and psychological upsets are uncommon and no more with medical than surgical abortion.

Cyproterone Acetate

Cyproterone acetate is a synthetic, steroidal antiandrogen drug, which also has additional progestogen-like properties. It is used in the treatment of acne and hirsutism in form of the OCP, dianette.

Mechanism of Action

Cyproterone inhibits LH release by its progestational activity. Lowering of serum testosterone occurs consequent to LH inhibition. This helps in supplementing the direct antiandrogenic action of cyproterone.

Indications:
❏ Precocious puberty in boys (prevention of pubertal changes)
❏ Inappropriate sexual behaviour in men (suppression of libido and androgenic anabolism)
❏ Acne and hirsutism in women (usually in combination with an oestrogen)
❏ *Combined oral contraceptive pills*: Due to its progestogenic effects, cyproterone acetate is a component of some COCPs (e.g. dianette and diane). These pills may be especially useful in women with severe acne and hirsutism desiring contraception.
❏ Metastatic prostate carcinoma (efficacy of this drug is inferior to other forms of androgen deprivation).

Dosage

This drug is administered orally in form of the following dosage schedule:
❏ In COCPs, cyproterone acetate (2 mg) is combined with 35 µg of ethinyl oestradiol. This is commercially available in form of dianette tablets. It is usually commenced on day 5 of the menstrual cycle and taken for 21 days followed by 7 days of pill-free interval. This combination of cyproterone acetate with ethinyl oestradiol is associated with an increased risk of thrombosis.
❏ For cases of hirsutism and hypersexuality, a dose of 25 mg BD is sufficient although dose up to 100 mg/day is permissible.
❏ For treatment of metastatic prostate cancer, dosage of up to 300 mg/day are sufficient.

Contraindications

The drug is to be prescribed with caution in the following conditions:

❑ History of allergy or sensitivity to any of the ingredients in the medicine

❑ Breastfeeding/pregnancy

❑ History of jaundice, gallstones, porphyria, systemic lupus erythematosus, hereditary angioedema, chorea

❑ Inflammatory bowel disease such as Crohn's disease or ulcerative colitis

❑ History of hypertension, renal problems, a family history of hypertriglyceridemia, thromboembolic problems (e.g. heart attack, angina, stroke, deep vein thrombosis, pulmonary embolism, etc.), chloasma, depression, liver tumours, migraine or severe headache

❑ Presence of risk factors for developing cervical cancer

❑ Sickle cell anaemia

❑ Vaginal bleeding having unknown cause

❑ Risk factors for developing breast cancer.

Side Effects

Use of this drug in men can be associated with complications such as decreased libido, erectile dysfunction, inhibition of gonadal function, reduced sexual drive and reduced sperm production and volume of semen. This may be associated with fertility problems. Less commonly, the use of this drug can result in complications such as a feeling of lack of interest or lack of energy, progestogenic side effects such as breast enlargement, and/or tenderness, weight changes, depressed mood, feeling of restlessness, hot flushes, etc., breathing difficulties, liver problems including jaundice, sweating, tiredness, etc. Rarely, it may result in complications such as benign breast lumps, galactorrhoea, hypersensitivity reactions, deranged liver function tests, increased risk of thrombosis, etc.

Other Drugs Used Commonly

Metformin: Metformin, an insulin sensitiser is a biguanide, which is used in the treatment of polycystic ovarian syndrome.

Duloxetine: This is a SNRI (serotonin and norepinephrine reuptake inhibitor) which is used in the treatment of incontinence.

Choose the Single Best Answer (SBA)

Q 1. Which of the following drugs does not cause renal impairment?
A. Cefuroxime
B. Diclofenac
C. Metformin
D. Ramipril
E. Simvastatin

Q 2. Which of the following is true regarding clonidine?
A. Is an alpha adrenergic receptor antagonist
B. Does not cause dryness of mouth
C. Reduces the minimal alveolar concentration of volatile anaesthetic agents
D. Stimulates the release of catecholamines
E. Sudden withdrawal is associated with hypotension

Q 3. Which of the following substances are sympathomimetic amines?
A. Ephedrine
B. Amphetamine
C. Isoprenaline
D. None of the above
E. All of the above

Q 4. Which of the following is not true regarding the drugs acting on the uterus?
A. Ergometrine is an oxytocic
B. Prostaglandin F2α may lead to an elevation of blood pressure

C. Oxytocin is a nonapeptide hormone
D. Mifepristone is an antiprogestogenic steroid
E. Ergometrine has a greater effect on the uterus at term than in early pregnancy

Q 5. Calcium antagonists are not used for treating which of the following diseases?
A. Angina
B. Hypertension
C. Pulmonary hypertension
D. Raynaud's phenomenon
E. Thyrotoxicosis

Q 6. Which of the following antibiotics is suitable for treating *Escherichia coli*?
A. Cefuroxime
B. Ciprofloxacin
C. Co-amoxiclav
D. All the above
E. None of the above

Q 7. Which of the following antibiotic acts on the bacterial walls?
A. Clindamycin
B. Polymyxin
C. Ceftazidime
D. Gentamicin
E. Metronidazole

1. C   2. C   3. E   4. B   5. E   6. D   7. C

Q 8. Which of the following statements is not correct regarding the antibiotics?
A. Metronidazole given by the rectal route is as effective as when given by the intravenous route
B. Streptomycin is nephrotoxic
C. Aminoglycosides are effective against anaerobes
D. Ampicillin may potentiate the anticoagulant effect of warfarin
E. Rifampicin use is not associated with an increased risk of neonatal bleeding in the third trimester

Q 9. Which of the following micro-organisms are sensitive to benzylpenicillin?
A. Bordetella pertussis
B. Cryptococcus neoformans
C. Mycoplasma pneumoniae
D. All the above
E. None of the above

Q 10. Resistance to penicillin is present in over 20% of isolates of which of the following bacterial species?
A. *Escherichia coli*
B. *Haemophilus influenzae*
C. *Neisseria meningitides*
D. Beta-haemolytic *Streptococci*
E. *Pseudomonas aeruginosa*

Q 11. Which of the following is true regarding penicillins?
A. Are bacteriostatic
B. Exert their actions by combining with a transpeptidase
C. Have a spectrum of action, which is independent of the beta lactam side chain
D. Have significant toxic effects on humans
E. Are not inactivated by the plasmid coded enzymes

Q 12. Which of the following is not true regarding co-trimoxazole?
A. Contains two different drugs
B. Displaces methotrexate from protein binding sites
C. Inhibits folic acid synthesis
D. Is bacteriostatic
E. Potentiates the action of warfarin

Q 13. Which of the following is a side effect of ciprofloxacin?
A. Convulsions
B. Discolouration of the urine
C. Nephrotoxicity
D. Ototoxicity
E. Pseudomembranous colitis

Q 14. Which of the following is true regarding aminoglycosides?
A. Act on the bacterial cell wall
B. Are active against Gram-negative bacteria
C. Are useful against anaerobes
D. Can be used in patients with renal failure
E. Can be administered orally

Q 15. Which of the following is not true concerning therapy with gentamicin?
A. Is associated with ototoxicity
B. Is administered via parenteral route
C. May produce ventilatory failure in sensitive patients
D. Requires monitoring of plasma concentrations
E. Undergoes hepatic excretion

Q 16. Which of the following is not true regarding the side effects of vancomycin?
A. Are liable to occur with chronic renal disease
B. Are more common in the elderly
C. Are not seen if the drug is given orally
D. Include irreversible vestibular damage
E. Tinnitus is another adverse effect which can occur

Q 17. Which of the following is not true regarding metronidazole?
A. Interferes with ethanol metabolism
B. Is effective against *Giardia lamblia*
C. Is effective when administered per rectum
D. Is usually effective against *Entamoeba histolytica*
E. Should not be administered intravenously

Q 18. Which of the following drug causes hypothyroidism?
A. Allopurinol
B. Doxycycline
C. Amiodarone
D. Probenecid
E. diazepam

Q 19. Which of the following is true concerning carbimazole?
A. Is not a prodrug
B. Is contraindicated in breast feeding and pregnant mothers
C. Is teratogenic
D. May cause lymphadenopathy
E. May cause irreversible agranulocytosis

Q 20. Which of the following is true regarding suxamethonium?
A. Echothiophate shortens its duration of action
B. Is a non-depolarising muscle relaxant
C. May be reversed using neostigmine
D. May trigger anaphylaxis
E. Has a shorter duration of action in pregnancy

Q 21. Which of the following statements is not true regarding low molecular weight heparin?
A. Exerts its anticoagulant effect by binding with antithrombin
B. Has fewer chains containing the unique pentasaccharide sequence (the binding site) than unfractionated heparin
C. Is excreted in the urine
D. Binds less to platelets, endothelium and von Willibrand factor
E. Inactivates thrombin more readily than unfractionated heparin

8. E   9. E   10. A   11. B   12. D   13. E   14. B   15. E   16. C   17. E   18. C   19. D   20. D   21. E

Q 22. Which of the following is true regarding heparin?
A. Enhances antithrombin III activity
B. Has no effect on platelet aggregation
C. Induces thrombocytopenia in 20% of patients
D. Is a strongly acidic protein
E. Readily crosses the placenta

Q 23. Which of the following does not have an anti-emetic action?
A. Chlorpropamide
B. Hyoscine hydrobromide
C. Morphine sulphate
D. Perphenazine
E. Promethazine hydrochloride

Q 24. Which of the following is a physiological effect of metoclopramide?
A. Acts on central dopaminergic receptors
B. Increases gastric acid secretion
C. Increases gastric fluid pH
D. Decreases gastroesophageal sphincter tone
E. Inhibits upper gastrointestinal motility

Q 25. Which of the following statements is true regarding domperidone?
A. Is not a recognised cause of galactorrhoea
B. Is less likely to produce acute dystonia than metoclopramide
C. Is typically associated with Parkinsonian-like adverse effects
D. Protects against drug-induced vomiting if given five minutes after apomorphine
E. Inhibits gastric peristalsis

Q 26. Ondansetron probably mediate its antiemetic effects by interacting with which of the following receptor systems?
A. Dopaminergic
B. GABA
C. Muscarinic/cholinergic
D. Nicotinic/cholinergic
E. Serotinergic

Q 27. Which of the following is not true concerning diazepam?
A. Effects may be antagonised by flumazenil
B. Has a hypnotic effect
C. Has an anticonvulsant effect
D. Has an antidepressant effect
E. Is a respiratory depressant

Q 28. Which of the following antihypertensives are ACE inhibitors?
A. Lisinopril
B. Losartan
C. Propranolol
D. All the above
E. None of the above

Q 29. Which of the following side effects can occur as a result of glucocorticoid therapy?
A. Hypertrichosis
B. Hypokalaemia
C. Lymphopenia
D. All the above
E. None of the above

Q 30. Which of the following statement is correct regarding the antiviral drugs?
A. Zidovudine eliminates the HIV virus
B. Inosine pranobex enhances the B-cell response to many viruses, including herpes and HIV
C. High levels of beta-interferon are found in the amniotic fluid and in the placenta
D. Acyclovir prevents DNA synthesis
E. Amantadine does not have an adverse effect on breastfeeding.

Q 31. Which of the following agents interfere with viral multiplication?
A. Amantadine
B. Ribavirin
C. Zidovudine
D. None of the above
E. All the above

Q 32. Which of the following are antiplatelet agents?
A. Heparin
B. Nitric oxide
C. Warfarin
D. None of the above
E. All of the above

Q 33. Which of the following is true concerning labetalol?
A. Causes bronchodilation
B. Decreases bile secretion
C. Has a half-life of 2 hours
D. Has only alpha blocking action
E. Is 70% protein bound

Q 34. Which of the following statement is correct regarding the antiviral drugs?
A. Zidovudine eliminates the HIV virus
B. Inosine pranobex enhances the B-cell response to many viruses, including herpes and HIV
C. High levels of beta-interferon are found in the amniotic fluid and in the placenta
D. Acyclovir prevents DNA synthesis
E. Amantadine does not have an adverse effect on breastfeeding

Q 35. Which of the following is true regarding aciclovir (acyclovir)?
A. Is effective against cytomegalovirus (CMV)
B. Acts via viral thymidine cycle
C. Stops herpetic neuralgia
D. All the above
E. None of the above

22. A 23. C 24. A 25. B 26. E 27. D 28. A 29. D 30. D 31. E 32. B 33. B 34. D 35. B

Q 36. **Which of the following is true regarding the anticancer agents?**
- A. Alkylating agents cause structural damage to chromosomes at the time of replication during interphase
- B. Vincristine binds to tubulin and causes metaphase arrest
- C. Androgens have a beneficial effect in certain mammary gland cancers
- D. All the above
- E. None of the above

Q 37. **Which of the following occurs more commonly in infants of opiate-abusing mothers?**
- A. Increased metabolic rate
- B. Intrauterine growth restriction
- C. Sudden infant death syndrome (SIDS)
- D. All the above
- E. None of the above

Q 38. **Which of the following is not true regarding cimetidine?**
- A. Has anti-androgenic actions
- B. Is inactive at neutral pH
- C. May reduce the metabolism of theophylline
- D. Reduces both vagal and gastrin induced acid secretion
- E. Reduces intracellular cyclic-AMP in the parietal cells

Q 39. **Which of the following statement is not true regarding morphine?**
- A. May cause bronchospasm
- B. Causes direct myocardial depression
- C. Has a higher affinity for the opioid receptor than diamorphine
- D. Increases the production of antidiuretic hormone (ADH)
- E. Is a phenanthrene

Q 40. **Nalorphine can antagonise the respiratory depression caused by which of the following drugs?**
- A. Diazepam
- B. Pentazocine
- C. Pethidine
- D. Thiopentone
- E. None of the above

Q 41. **Which of the following chemotherapies is/are alkylating agents?**
- A. Chlorambucil
- B. Melphalan
- C. Cisplatin
- D. None of the above
- E. All the above

Q 42. **Out of the following, which is not a tocolytic agent?**
- A. GTN
- B. Progesterone
- C. Propofol
- D. Salbutamol
- E. Nifedipine

Q 43. **Which of the following is not true regarding sodium valproate?**
- A. Is effective in treating petit mal
- B. Is the drug of choice in controlling epilepsy in pregnancy
- C. Levels above the therapeutic range may be tolerated without side effects
- D. May prolong the bleeding time
- E. Rarely causes hyperactivity in children

Q 44. **Which of the following are known complications of phenytoin therapy?**
- A. Hirsutism
- B. Dental caries
- C. Balding
- D. Microcytic anaemia
- E. None of the above

Q 45. **Which of the following is true regarding propranolol?**
- A. Crosses the blood brain barrier poorly
- B. Has a long elimination half-life
- C. Has a small volume of distribution
- D. Exacerbates bronchospasm
- E. Is a cardio-selective beta adrenoceptor antagonist

Q 46. **Use of beta sympathomimetic drugs can cause which of the following?**
- A. Bronchospasm
- B. Heart block
- C. Increase blood glucose concentration
- D. Increase in diastolic blood pressure
- E. Decrease in the frequency of uterine contractions

Q 47. **Which of the following is classed as loop diuretics?**
- A. Bumetanide
- B. Metolazone
- C. Spironolactone
- D. Triamterene
- E. None of the above

Q 48. **Which of the following are potassium-sparing diuretics?**
- A. Bendroflumethiazide
- B. Captopril
- C. Furosemide
- D. Triamterene
- E. None of the above

Q 49. **Which of the following drug in pharmacological doses have been shown to cause a rise in blood glucose?**
- A. Thiazide diuretics
- B. Ethanol
- C. Aspirin
- D. Gliclazide
- E. Atenolol

36. D   37. D   38. B   39. B   40. C   41. E   42. C   43. B   44. A   45. D   46. E   47. A   48. D   49. A

Q 50. **Which of the following are true regarding cabergo-line?**
- A. Has a half-life of 8 hours
- B. Is a dopamine antagonist
- C. Is an effective antiemetic
- D. Is not used during pregnancy
- E. May cause Parkinsonian side effects

Q 51. **Which of the following is not true regarding local anaesthetic drugs?**
- A. Local anaesthesia combined with adrenaline should be avoided in patients taking tricyclic antidepressants
- B. The total dose of adrenaline combined with local anaesthesia should not exceed 500 µg in a single operation
- C. Bupivacaine is highly protein-bound
- D. Convulsions which are a side effect of systemic absorption of lignocaine can be controlled with thiopentone and adequate oxygenation
- E. Mepivacaine, like other anaesthetics, causes vasodilatation

Q 52. **Which of the following is true concerning lidocaine?**
- A. Inhibits the conduction of neuronal impulses
- B. Inhibits the generation of neuronal impulse
- C. Is associated with haemolytic anaemia
- D. Is effective for the treatment of both supraventricular and ventricular tachycardia
- E. Is only effective via oral route

Q 53. **Prescription of the emetic drug, ipecacuanha must be indicated after accidental ingestion of which of the following?**
- A. 10 ferrous sulphate tablets
- B. 20 mL paraffin
- C. 50 mL turpentine
- D. None of the above
- E. All of the above

Q 54. **Which of the following statements is true concerning oral hypoglycaemic agents?**
- A. Chlorpropamide induces liver enzymes
- B. Sulphonylureas stimulate release of insulin
- C. Metformin inhibits glucose absorption from the gut
- D. All have mildly diuretic action
- E. Glibenclamide is excreted unchanged by the kidney

Q 55. **Which of the following is the side effects of alpha-methyldopa?**
- A. Gingival hypertrophy
- B. Nasal congestion
- C. Cataracts
- D. Hirsutism
- E. ECG abnormalities

Q 56. **Which of the following is true regarding sulphony-lurea therapy?**
- A. Enhances glucose stimulated insulin release from the pancreas
- B. Has hyponatraemia as a side effect
- C. Is useful in all type II diabetics
- D. May cause weight loss
- E. Stimulates peripheral glucose utilisation

Q 57. **Which of the following is not true regarding the drugs acting on the thyroid gland?**
- A. Propylthiouracil (PTU) blocks the incorporation of iodine into tyrosine
- B. Perchlorate prevents the uptake of iodide by the follicular cells in the thyroid gland
- C. Bone marrow depression is a recognised side effect of carbimazole
- D. PTU appears in breast milk in an insignificant amount
- E. Intravenous D-thyroxine is the treatment of choice in hypothyroid coma

Q 58. **Which of the following is true regarding the drugs acting on the thyroid gland?**
- A. Thyroid-stimulating hormone (TSH) is synthetised and released from the thyroid gland
- B. Carbimazole cause irreversible agranulocytosis
- C. Propylthiouracil crosses the placenta in high doses and may cause foetal goitre
- D. Anginal pain is not a recognised side effect of liothyronine (Tertroxin)
- E. Carbimazole is contraindicated in breastfeeding

Q 59. **Which of the following drugs do not cause thrombo-cytopenia?**
- A. Low molecular weight heparin
- B. Aspirin
- C. Isoniazid
- D. D-penicillamine
- E. Bendroflumethiazide

Q 60. **Angiotensin-converting enzyme inhibitors are not associated with which of the following?**
- A. Impair foetal renal function
- B. Are associated with polyhydramnios
- C. May cause foetal death in later pregnancy
- D. Should be stopped before conception
- E. Can be safely used in the puerperium

Q 61. **Which of the following statement is true regarding beta-adrenoceptor blocking drugs?**
- A. May improve asthma
- B. Can cause intrauterine growth restriction when given from early pregnancy
- C. Labetalol is a pure beta-blocker, and is the oral agent of choice in the United Kingdom for treating severe pre-eclamptic hypertension
- D. Labetalol has intrinsic sympathomimetic activity (ISA)
- E. Are absolutely contraindicated in diabetes mellitus

50. D   51. E   52. A   53. A   54. B   55. B   56. A   57. E   58. C   59. C   60. B   61. B

Q 62. Which of the following statement is true regarding sildenafil (viagra)?

A. Increases libido
B. Is rarely associated with flushing as a side effect
C. Is associated with nasal congestion as a recognised side effect
D. Is contraindicated in patients with ischaemic heart disease
E. Is of no use in patients who have developed impotence following prostate surgery

Q 63. Which of the following regarding carbimazole is not true?

A. Carbimazole is secreted in milk
B. Carbimazole is teratogenic and must be avoided in pregnancy
C. Relapse in thyrotoxicosis is very rare when carbimazole treatment is continued for 2 years
D. Skin rashes due to carbimazole are likely to recur if therapy is changed to propylthiouracil
E. Symptomatic hypocalcaemia following subtotal thyroidectomy is usually permanent

Q 64. Which of the following is not an adverse effect of carbimazole?

A. Agranulocytosis
B. Alopecia
C. Cholestatic jaundice
D. Gynaecomastia
E. All the above

Q 65. Which of the following statement regarding the antidepressant drugs is correct?

A. Tricyclic antidepressants may greatly potentiate the pressor effect of tyramine
B. Serotonin uptake inhibitors such as fluoxetine (Prozac) cause more side effects than the tricyclics
C. Tricyclic antidepressants may cause tachycardia and irritability in the neonate
D. Antidepressant drugs are effective in the treatment of mild depression
E. Tricyclic antidepressants do not cause convulsions

Q 66. Which of the following statement regarding antidepressant drugs is not correct?

A. Amitriptyline can cause sudden death
B. Fluoxetine (Prozac) is the only antidepressant that is safe to be used in children
C. Imipramine can lead to urine retention and constipation
D. Anaesthetics given during imipramine therapy may increase the risk of arrhythmias and hypotension
E. Amitriptyline can cause galactorrhoea

Q 67. Ventilation is increased due to stimulation of central receptors by which of the following drugs?

A. Doxapram
B. Hypoxia
C. Nikethamide
D. All the above
E. None of the above

Q 68. Which of the following is true regarding beta-sympathomimetic drugs in therapeutic doses?

A. Are contraindicated in thyrotoxicosis
B. Cause a decrease in cardiac output
C. Cause arrhythmias
D. Cause bradycardia
E. Lower blood pressure

Q 69. Which of the following drugs can cause broncho-constriction?

A. Aspirin
B. Atropine
C. Ritodrine
D. All the above
E. None of the above

Q 70. Which of the following statement regarding bromocriptine is not correct?

A. Suppresses lactation in the puerperium
B. May be a useful adjunct in acromegaly
C. May cause enlargement of the pituitary in a patient with a prolactinoma
D. Inhibits dopamine receptors
E. Is an ergot derivative

Q 71. Which of the following drug acts by blocking sodium channels?

A. Carbamazepine
B. Diazepam
C. Ethosuximide
D. All the above
E. None of the above

Q 72. Which of the following statement is correct regarding the drugs used during breastfeeding?

A. Ranitidine is safe to use as it is not secreted in breast milk
B. Methyldopa is the drug of choice to treat hypertension during the puerperium
C. Angiotensin-converting enzyme (ACE) inhibitors may be used safely during the puerperium
D. Thiazide diuretics have been shown to increase milk production
E. The combination of co-amoxiclav (Augmentin) and metronidazole (Flagyl) is recommended to treat puerperal infection in breastfeeding mothers

62. C   63. A   64. D   65. C   66. B   67. D   68. C   69. A   70. D   71. A   72. C

Q 73. Regarding drug therapy during pregnancy, which of the following statement is true?
- A. Folic acid supplements are not required for the patients taking phenytoin
- B. Heparin has been shown to cause central nervous system damage in the foetus if given in the second and third trimesters
- C. Methyldopa is contraindicated throughout
- D. Thiazide diuretics do not reduce placental perfusion
- E. Treatment with isotretinoin is a recognised indication for a termination

Q 74. Out of the following drugs, which one is not known to be teratogenic?
- A. Alcohol
- B. Methyldopa
- C. Warfarin
- D. Aminoglycosides
- E. Phenytoin

Q 75. Which of the following drugs is unsafe in the last 4 weeks of pregnancy?
- A. Paracetamol
- B. Co-trimoxazole
- C. Methylpenicillin
- D. Tetracycline
- E. None of the above

Q 76. Which of the following drug can be safely used during lactation?
- A. Amantadine
- B. Androgen
- C. Co-amoxiclav
- D. Captopril
- E. None of the above

Q 77. Maternal cocaine use in pregnancy is not associated with which of the following adverse effects?
- A. Foetal cerebral infarction
- B. Increased incidence of preterm labour
- C. Marked neonatal withdrawal symptoms
- D. Placental abruption
- E. Uterine vessel vasoconstriction

Q 78. Which of the following statements is true regarding foetal alcohol syndrome?
- A. It affects one foetus in 100
- B. The mothers at risk are usually identified antenatally
- C. It is associated with a 50% risk of mental restriction
- D. The affected foetus often shows macrosomia
- E. Facial malformations are rarely seen

Q 79. Which of the following drugs is contraindicated in the lactating woman?
- A. methyldopa
- B. Bromocriptine
- C. Heparin
- D. Insulin
- E. Warfarin

Q 80. Which of the following is true regarding LHRH analogues?
- A. Can be used to treat endometriosis
- B. Rarely cause side effects
- C. Can be administered orally
- D. Are inexpensive preparations
- E. Act principally at the uterine level

Q 81. Which of the following correct concerning clomiphene citrate is correct?
- A. It is usually started in a dosage of 100 mg daily
- B. It is an antiandrogen
- C. It is associated with visual disturbances
- D. It produces direct induction of ovulation
- E. It is usually started on day 7 of the cycle

Q 82. Which of the following statements regarding progesterone are not correct?
- A. Progesterone is mainly secreted by the corpus luteum
- B. Falling levels of progesterone in the last days of the cycle trigger menstruation
- C. Trophoblastic HCG causes the corpus luteum to persist in the early weeks of pregnancy
- D. Trophoblastic progesterone production takes over from the corpus luteum at about 8 weeks
- E. Progesterone increases uterine sensitivity to prostaglandins

Q 83. Which of the following is correct regarding mifepristone?
- A. Mifepristone has to be given parenterally
- B. Mifepristone during the luteal phase effectively stops pregnancy from occurring
- C. Mifepristone given in the first 12 weeks of pregnancy effectively induces abortion
- D. Medical abortion with Mifepristone carries a higher risk of infection that vacuum aspiration
- E. Mifepristone binds competitively to progesterone receptors and has greater affinity for them than has progesterone

Q 84. Which of the following is correct regarding cyproterone acetate?
- A. It is an agonist of the beta-oestrogen receptor
- B. Is associated with visual disturbance as a recognised side effect
- C. It is used in the treatment of acne
- D. Is usually commenced on the 1st day of the menstrual cycle
- E. It is an anti-oestrogenic preparation

Q 85. Heavy menstrual bleeding can be treated with which of the following?
- A. The combined oral contraceptive pill
- B. Tranexamic acid
- C. Medicated intrauterine devices
- D. All the above
- E. None of the above

73. E   74. B   75. B   76. C   77. C   78. C   79. B   80. A   81. C   82. E   83. E   84. C   85. D

Q 86. Which of the following statement regarding dysmenorrhoea is not correct?

A. Can be successfully treated with non-steroidal anti-inflammatory drugs
B. Does not occur in anovulatory cycles
C. Tends to be more severe with a retroverted uterus that is associated with a pelvic pathology
D. Can be caused by endometriosis
E. Can be caused by anterior vaginal wall prolapse

Q 87. Which of the following is true regarding GnRH?

A. It is distinct from LH-RH
B. It is produced in the posterior pituitary
C. It is a decapeptide
D. It exerts its main effect directly on the ovary
E. It is used for inducing ovulation in IVF programmes

Q 88. Which of the following may reduce the anticoagulant effect of warfarin?

A. Aspirin
B. Oral contraceptive pills
C. Ranitidine
D. Diazepam
E. Ciprofloxacin

Q 89. Which of the following are correct statements concerning the effect of local anaesthetics on nerve fibres?

A. Conduction in unmyelinated nerve fibres is blocked first
B. Nerve fibres with diameters less than one micrometre are blocked first
C. Preganglionic fibres are blocked before postganglionic fibres
D. Proprioception sensation is blocked before temperature sensation
E. Temperature sensation is lost at the same time as motor function

Q 90. Which of the following antibiotics are considered bactericidal?

A. Ethambutol
B. Gentamicin
C. Tetracycline
D. Erythromycin
E. Clindamycin

Q 91. Which of the following statement regarding opioid drugs is correct?

A. Cause pupillary dilation
B. Lead to a decrease in the release of vasopressin
C. Cannot cross the placenta
D. Codeine is more potent analgesic than dihydrocodeine
E. Morphine can cause hypotension associated with a reflex tachycardia

Q 92. What is the anticoagulant of choice in a pregnant woman with previous history of multiple pulmonary embolisms at 8 weeks of gestation?

A. Aspirin 500 mg
B. Heparin infusion
C. Low-molecular weight heparin
D. Warfarin
E. Aspirin 300 mg

Q 93. What is the most appropriate treatment option in an 8-week pregnant woman with vaginal candida infection unresponsive to clotrimazole cream?

A. Metronidazole 400 mg orally
B. Fluconazole 400 mg
C. Hydrocortisone cream 0.5%
D. Trimovate cream
E. Clotrimazole pessary

Q 94. In which of the following circumstances would the efficacy of the combined oral contraceptive pill (COCP) is reduced?

A. Oral omeprazole given for gastritis
B. Oral rifampicin given as acute prophylaxis for meningitis
C. Oral sodium valproate for epilepsy
D. Oral tetracycline given 2 months for treatment of acne
E. None of the above

Q 95. Which of the following statements about drug interactions is not correct?

A. Alcohol metabolism is impaired by metronidazole
B. Antacids decrease intestinal absorption of tetracyclines
C. The effects of bromocriptine are inhibited by chlorpromazine
D. The effects of warfarin are potentiated by combined oral contraceptive
E. None of the above

Q 96. Which of the following statements concerning the reporting of adverse drug reactions (ADR) in the United Kingdom is not correct?

A. A black triangle sign alongside a drug within the British National Formulary (BNF) requires that all ADRs should be reported with this drug
B. ADR reporting is compulsory
C. Reporting is required for ADRs associated with vaccinations
D. There is a requirement to report ADRs with over the counter (OTC) drugs as well
E. When submitting a yellow card concerning an ADR, all other drugs taken within the last 3 months should also be reported

86. E   87. C   88. B   89. B   90. B   91. D   92. C   93. E   94. D   95. D   96. B

Obstetrics

Early Pregnancy Care

The average duration of human pregnancy is 280 days from the first day of the last menstrual period (LMP) until delivery.

Pregnancy Dating

The expected date of delivery (EDD) is most commonly calculated using Naegele's rule.

Naegele's rule: Using Naegele's rule, the EDD is calculated by adding 9 calendar months and 7 days to the first day of the LMP (28-day cycle). For in vitro fertilisation pregnancies, date of LMP is 14 days prior to the date of embryo transfer.

Ultrasonographic dating: This is most accurate from 7 weeks to 11 weeks of pregnancy. Measurement of the sac size between 6 weeks and 8 weeks is a good indicator of the gestational age. Very early foetal measurements can be made using transvaginal ultrasound. While measurement of the crown-rump length is used in the first trimester, second trimester measurements use the bi-parietal diameter. Clinical examination is very poor at assessing the period of gestation.

Early Pregnancy Loss

A stillbirth can be defined as foetal death after 24 completed weeks of pregnancy. A miscarriage is the loss of the products of conception before the foetus attains viability (i.e. before 24 completed weeks of gestation). An abortion is the premature expulsion of the products of conception, either embryo or non-viable foetus from the uterus. Measurement of serum β-hCG levels and transvaginal ultrasound have revolutionised the management of early

pregnancy complications. Most intrauterine pregnancies will be visible on transvaginal scan (TVS) once the β-hCG levels are greater than 1,000 IU/L. TVS is highly accurate in assessing whether or not the pregnancy is viable. hCG titres are also helpful because in normal cases β-hCG levels double after every few days.

Miscarriage at 10 Weeks

Most miscarriages occurring in the first trimester (>60%) are due to genetic causes. This could be related to major chromosomal abnormalities such as triploidy (having a full extra half set of chromosomes resulting in a total number of 69 chromosomes). In cases of non-viable pregnancy, the growth of placental structures may continue, leading to the development of the sac. However, there is no development of the embryo, resulting in an "empty sac" phenomenon on the ultrasound scan. If the pregnancy is of correct size in accordance with the period of gestation, and foetal heart activity can be seen, the risk of miscarriage is less than 5%.

If a woman has two early miscarriages, both will most probably be of chromosomal origin. However, the abnormalities may be different and unrelated to each other, and the risk of recurrence is likely to be small. In case of recurrent miscarriages, one of the partners may have a balanced translocation.

In case of missed miscarriages, the foetus becomes dead and is retained inside the uterine cavity. The pregnancy becomes non-viable but remains intact. There is no bleeding and the internal os is closed. Some patients may miscarry within a couple of weeks, but most would require medical or surgical intervention.

Threatened abortion is a type of abortion where the process of abortion has begun but has yet not progressed to a stage from where the recovery would be impossible.

In case of threatened abortion, despite the occurrence of bleeding before 20 weeks of gestation, the cervical os is

closed. The bleeding may be with or without pain. In the majority of cases, the uterus will be of normal size and the hCG levels normal as most of the pregnancies will carry on normally. There is no evidence that any form of hormonal treatment appears to be beneficial.

Investigations

Appropriate maternal investigations following a term stillbirth include the following:

❏ *Glycosylated haemoglobin*: Maternal glycosylated haemoglobin levels are required for the detection of diabetes mellitus (DM), because the postprandial blood glucose values may be within normal limits. This is related to the fact that glycaemic control may have returned to within normal limits post-partum.
❏ *Kleihauer blood test*: Kleihauer blood test is an acid-stained film of maternal blood. For details related to this test, kindly refer to Chapter 6. This test helps in establishing the presence of foetal red blood cells and in quantitating the volume of foetomaternal transfusion.
❏ *Platelet count*: An increasing incidence of consumptive coagulopathy develops with time following foetal demise. Therefore, the platelet count should be checked in a woman with a retained dead foetus in utero.
❏ *Blood pressure measurement*: Hypertensive diseases with various aetiologies increase the perinatal mortality rate.
❏ *Antinuclear antibody estimation*: Systemic lupus erythematosus is associated with an increased pregnancy loss in all trimesters.

Treatment

Numerous treatment options for threatened and recurrent miscarriage have been postulated and tried.

Previously stilbestrol was used, which was associated with numerous uterine and cervical abnormalities in the progeny and an increased risk of vaginal adenocarcinoma in the female offspring. Use of progestogens (based on testosterone) may produce virilisation in the female foetus. Bed rest also does not prove to be useful. Even with a non-viable pregnancy, evacuation is not mandatory. If there are no grounds for immediate evacuation, such as heavy bleeding, patients are now given the following options:

❏ Expectant/watchful management
❏ Medical (mifepristone followed by misoprostol or cervagem (gemeprost) management
❏ Surgical management.

Though surgical evacuation is associated with immediate results, it is also associated with the risks of anaesthesia, infection, perforation, etc.

Patients undergoing medical and expectant management must be advised to visit the doctor immediately in the event of having an episode of bleeding or severe pain necessitating emergency admission.

Anti-D is now thought unnecessary if the period of gestation is under 12 weeks, unless there has been instrumentation of the uterus. It is believed that beneath 12 weeks of gestation, the possibility of foetomaternal transfusion is minimal.

Medical Termination of Pregnancy

As per the Abortion Act (1967) in the UK, the abortions can only be carried out in a hospital or a specialist licensed clinic. Medical termination of pregnancy for social reasons has an upper gestational age limit of 24 weeks. However, in the presence of significant foetal abnormality, no such upper limit exists. Termination of pregnancy requires the signature of two independent medical practitioners. Induction of uterine contractions using prostaglandins is a safe and successful method for medical termination of pregnancy. Prostaglandin can be administered via the vaginal, extra-amniotic, intra-amniotic and intravenous routes. Hysterotomy is rarely used to achieve termination of pregnancy.

Gestational Trophoblastic Diseases

Complete Hydatidiform Mole

Hydatidiform mole (H. mole) belongs to a spectrum of disease known as gestational trophoblastic disease (GTD), resulting from overproduction of the chorionic tissue, which is normally supposed to develop into the placenta. It can be considered as an abnormal pregnancy in which placental villi become oedematous (hydropic) and start proliferating, resulting in the development of a cystic, grape-like structures filled with watery fluid. There are two types of benign (potentially pre-malignant) form of GTD: complete H. mole (CHM) and partial H. mole (PHM). Comparison between a complete mole and partial mole has been illustrated in **Table 13.1**.

Aetiology

The World Health Organization (WHO) classification of GTD is described in **Table 13.2**.

The incidence of GTD in the UK is 1 in 714 total live births, 1 in 387 for Asian women, and 1 in 752 for non-Asian women. Age is the biggest risk factor for the population at large. A ten-fold increase in incidence of H. mole is observed in women more than 40 years and a 1.3-fold increase in the incidence of H. mole is observed in girls less than 16 years of age. GTD is believed to be about twice as common amongst Asian women compared to Caucasians. Another important risk factor is the history of occurrence of molar gestation in the previous pregnancy. A woman who has had a mole has a risk of recurrence of 1:80. There is some evidence that the risk of a molar pregnancy is increased if conception

TABLE 13.1	Comparison between complete and partial mole	
Parameter under consideration	Complete mole	Partial mole
Cytogenetic studies	Karyotype 46XX	Triploid karyotype 69XXY
Aetiology	Duplication of the haploid sperm following fertilisation of an "empty" ovum or dispermic fertilisation of an "empty" ovum	These contain two sets of paternal haploid genes and one set of maternal haploid genes. They usually occur following dispermic fertilisation of an ovum.
Histopathological analysis	There is no evidence of foetal tissue.	There may be an evidence of foetal tissue or red blood vessels.
Invasive potential and propensity for malignant transformation	Persistent trophoblastic disease following uterine evacuation may develop in about 15% cases with a complete mole.	Persistent trophoblastic disease may develop in <5% of cases of partial mole formation.

TABLE 13.2	The WHO classification of gestational trophoblastic disease

Potentially pre-malignant conditions

- Complete hydatidiform mole
- Partial hydatidiform mole

Malignant conditions

- Choriocarcinoma
- Invasive mole
- Placental site trophoblastic tumour

TABLE 13.3	Causes where the uterine size is larger than the dates	
Abnormal pathology	Characteristics	
Uterine fibroids/leiomyomas	Uterine abnormalities	
Polyhydramnios	Overproduction of liquor/foetal abnormalities	
Foetal macrosomia	Increased size of various body organs	
Multiple births	Presence of more than one foetus	
Diabetes	Could be associated with macrosomia	

occurs while on the pill, but this still remains debatable. An increased risk of complete mole is associated with inadequate intake of carotene or animal fats. Nulliparity has been described as a risk factor by some studies. Blood group serves as another risk factor. Some studies have suggested links to dietary or genetic factors.

Clinical Presentation

Initially, the symptoms may be suggestive of early pregnancy; however, the uterus is often larger than the period of gestation. The foetal movements and heart tones are usually absent. The uterus may appear doughy in consistency due to lack of foetal parts and amniotic fluid. External ballottement is absent.

There may be history suggestive of vaginal bleeding or passage of grape-like tissue early in pregnancy. There may be excessive nausea and vomiting. Hyperemesis may commonly occur. There may be symptoms suggestive of hyperthyroidism. H. mole may be associated with early appearance of pre-eclampsia.

Complete moles are mostly present with bleeding in the first trimester and are diagnosed on scan. In previous times, complete moles usually presented late with features like exaggerated pregnancy symptoms: hyperemesis, uterus large for dates, passage of vesicles, ovarian cysts, hyperthyroidism, early-onset pre-eclampsia, etc. Nowadays, early diagnosis tends to make these features uncommon. Previously, it was thought that the ratio of a partial:complete

mole is 3:1. It is now considered to be less than 3:1. The requirement for chemotherapy following complete and partial moles is respectively 15% and 0.5%.

General Physical Examination

Signs suggestive of pre-eclampsia, hyperthyroidism and/or early pregnancy may be observed. Extreme pallor may be sometimes seen. The patient's pallor may be disproportionate to the amount of blood loss due to concealed haemorrhage.

Per Abdominal Examination

On the abdominal examination, the uterine size is usually abnormal in relation to the period of gestation. In most of the cases of CHM, the uterine size may be larger than the period of gestation. Other causes where the uterine size is larger than the dates are listed in **Table 13.3**. On the other hand, in cases of PHM, the uterine size may be smaller in relation to the period of gestation. The uterus may appear doughy in consistency due to lack of foetal parts and amniotic fluid. Foetal movements and foetal heart sounds are absent. Foetal parts are usually not palpable. External ballottement is absent.

Vaginal Examination

There may be some vaginal bleeding or passage of grape-like vesicles. Internal ballottement cannot be elicited due to lack of foetus. Unilateral or bilateral enlargement of the ovaries in the form of theca lutein cysts may be palpable.

Investigations

❏ Complete blood count, blood grouping and cross-matching

❏ β-*hCG levels*: β-hCG levels in both serum and urine are raised. In cases of complete mole, β-hCG levels may be more than 100,000 mIU/mL. β-hCG assay is fundamental for the follow-up because the level correlates well with the amount of residual tumour. In addition to the urinary hCG monitoring, a blood specimen for beta-hCG is usually requested after 6 weeks or so. There is some evidence that elevated values of β-hCG indicate a higher risk of progression to choriocarcinoma.

❏ *Ultrasound of the pelvis*: In case of a complete mole, the following features are observed:
  • Features of missed miscarriage or anembryonic pregnancy (blighted ovum) may be observed on ultrasound examination
  • Characteristic vesicular pattern, also known as "snowstorm appearance" may be present due to generalised swelling of the chorionic villi and presence of multiple, small cystic spaces
  • There may be presence of an enlarged uterine endometrial cavity containing homogeneously hyperechoic endometrial mass with innumerable anechoic cysts **(Fig. 13.1)**
  • Ultrasound may also show the presence of theca lutein cysts in the ovaries.

❏ *Histopathological examination*: Pathologic evaluation may demonstrate swollen chorionic villi having a grape-like appearance, along with presence of hyperplastic trophoblastic tissue.

Obstetric Management

The two main treatment options in case of H. mole are suction evacuation and hysterectomy.
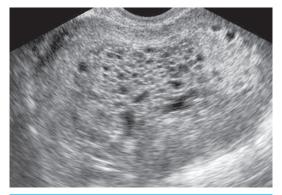
FIG. 13.1: Transvaginal sonogram of a second trimester complete hydatidiform mole (transverse section)

Suction Evacuation

Due to the lack of foetal parts, a suction catheter, up to a maximum size of 12 mm, is usually sufficient to evacuate all complete molar pregnancies. In order to ensure that complete sustained remission has been achieved, serial assays of serum and urine β-hCG levels should be carried out on 2 weekly basis until three negative levels are obtained. Post evacuation, contraceptive measures should be instituted and the patient advised to avoid pregnancy until hCG values have remained normal for 6 months.

Evacuation of the retained products of conception with sponge holders and curettes should be avoided because of the theoretical risk of increasing trophoblastic embolisation, and risk of heavy bleeding. Medical termination and cervical "ripening" are to be avoided. The Royal College of Obstetricians and Gynaecologists (RCOG) advises that syntocinon infusions should not be used until the uterus has been evacuated and that prostaglandins should only be used if syntocinon has failed. The RCOG advises avoiding mifepristone as it increases the sensitivity of the uterus to prostaglandins. Hysterectomy is rarely required. Chemotherapy following evacuation may be required in cases of persistent disease as evidenced by rising or plateauing of β-hCG levels.

Hysterectomy with Mole In Situ

Hysterectomy may serve as an option in elderly multiparous women (age >40 years) who do not wish to become pregnant in the future; women with H. mole desiring sterilisation; those with severe infection or uncontrolled bleeding; and patients with non-metastatic persistent disease who have completed childbearing or are not concerned about preserving their fertility.

The main complication associated with H. mole is the risk of development of persistent gestational trophoblastic neoplasia (GTN).

Follow-up

The length of follow-up will vary from case to case, particularly depending on whether it was a complete or partial mole. However, it will usually be for 6–12 months for a partial mole and up to 24 months for a complete mole. An hCG level after each subsequent pregnancy is recommended irrespective of the period at which it ends. The risk of recurrence of a mole is only about 1 in 55. An early scan may be required.

Partial Mole

Partial H. mole is another form of benign disease belonging to the spectrum of GTD. Difference between the complete and partial mole has already been described in **Table 13.1**.

Clinical Presentation

The clinical features are same as that with CHM, described previously.

Investigations

❑ *Ultrasound examination*: There may be presence of a large placenta, cystic spaces within the placenta, an empty gestational sac or sac containing amorphous echoes or growth-retarded foetus. There is an increase in the ratio of transverse to anterior-posterior dimension of the gestational sac to a value greater than 1.5.
❑ *Histopathological examination*: Although complete mole does not contain any foetal tissue, non-viable foetal tissue may sometimes be present in PHM.

Management

Management in cases of PHM is same as that described with CHM.

Gestational Trophoblastic Neoplasia

Gestational trophoblastic neoplasia or the persistent disease represents a spectrum of malignant diseases associated with the spectrum of GTDs. These include:
❑ Invasive mole
❑ Choriocarcinoma
❑ Placental site trophoblastic tumour (PSTT)
❑ Epithelioid trophoblastic tumour.

If the β-hCG level does not normalise within 10 weeks, the disease is classified as persistent. If metastasis is detected on various investigations (chest X-ray, CT, MRI, etc.), the disease is classified as metastatic. If no metastasis is detected, the disease is classified as non-metastatic. The lungs are the most common site for metastasis in case of malignant GTD.

Persistent GTD can occur after molar pregnancy, normal pregnancy and abortion, ectopic pregnancy, miscarriage and termination of pregnancy. Choriocarcinoma usually follows a complete mole.

Clinical Presentation

The metastatic disease can spread through the blood stream to lungs (80%), vagina (30%), pelvis (20%), brain (10%) and liver (10%). Metastasis to the lungs may result in symptoms like dyspnoea, cough, haemoptysis, chest pain, etc.

Investigations

❑ *Investigations to detect the metastatic disease*: Besides the investigations which must be done with CHM, in the cases of GTN, investigations, such as chest X-ray and CT scan of brain, chest, abdomen and pelvis, need to be done in order to detect the metastatic disease. On chest X-ray, the lungs may show presence of distinct nodules or cannon ball appearance.
❑ β-*hCG titres*: Women who have the malignant form of GTD may show β-hCG titres, which either plateau or rise and remain elevated beyond 8 weeks.

Management

In most of the cases, non-metastatic disease can be treated with a single chemotherapeutic drug, either methotrexate (more commonly used) or dactinomycin (used in cases of resistance to methotrexate). If single drug chemotherapy is ineffective, hysterectomy or multidrug chemotherapy can be tried.

In case of metastatic disease, the system adopted by the WHO and Federation International of Gynaecologists and Obstetricians (FIGO) for classifying gestational trophoblastic tumours (GTTs) and treatment protocols is shown in **Table 13.4**. According to this scoring system, low-risk group has a score of 0–6; the moderate-risk group has a score between 5 and 7; and the high-risk group will have a score of 7 or higher.

Low-risk metastatic disease is treated with single- or multiple-drug chemotherapy (intramuscular methotrexate or a combination of intravenous dactinomycin and etoposide). Moderate-risk metastatic disease is usually treated with multiagent chemotherapy. Women with

TABLE 13.4	The classification system by the WHO and FIGO for classifying gestational trophoblastic tumours and treatment protocols			
Risk factor	**0**	**1**	**2**	**4**
Age (years)	<40	>40	—	—
Antecedent pregnancy	Mole	Abortion	Term	—
Interval (end of antecedent pregnancy to chemotherapy in months)	<4	4–6	7–13	>13
Human chorionic gonadotropin (IU/L)	$<10^3$	$10^3–10^4$	$10^4–10^5$	$>10^5$
Number of metastasis	0	1–4	5–8	>8
Site of metastasis	Lung	Spleen, kidney	Gastrointestinal tract	Brain, liver
Largest tumour mass	—	3–5 cm	>5 cm	—
Previous chemotherapy	—	—	Single drug	>2 drugs

high-risk GTT usually require combination chemotherapy along with selective use of surgery and radiotherapy. The standard multi-agent chemotherapy regimen in high-risk group is EMA/CO in which the drugs, like etoposide, dactinomycin and methotrexate, are alternated at weekly intervals with vincristine and cyclophosphamide.

Most women remain fertile after chemotherapy. Ovulation usually returns by 6 months. Combination chemotherapy, unlike the monotherapy with methotrexate increases the risk of other cancers later in life, particularly myeloid leukaemia, breast cancer and bowel cancer.

Persistent Gestational Trophoblastic Disease and Choriocarcinoma

About 50% of choriocarcinomas occur after molar pregnancy and approximately 25% follow normal pregnancy. The rest may occur after miscarriage, ectopic or no recognised pregnancy.

The risk of choriocarcinoma after a mole is enormously greater than after a normal pregnancy, by a factor of approximately 1,000. According to Luesley and Baker, the risk of choriocarcinoma is 3% after a complete mole, i.e. 15 of every 100 complete moles will go on to develop persistent GTD. But, only 3 of the 15 would develop into choriocarcinoma without treatment.

In the UK, most cases of moles are rigorously followed up. Persistent GTD and the development of choriocarcinoma will be detected by the following investigations as described in **Table 13.5**. Features suggestive of worse prognosis of choriocarcinoma are listed in **Table 13.6.**

TABLE 13.5	Investigations suggestive of persistent gestational trophoblastic disease

- β-hCG levels greater than 20,000 IU/L even 4 weeks after the uterus was emptied
- β-hCG levels increasing over two samples
- Proven choriocarcinoma on histopathological studies
- hCG level still elevated and hitting a plateau over 3 samples
- Heavy bleeding and elevated hCG levels
- Evidence suggestive of metastases

TABLE 13.6	Features suggestive of poor prognosis of choriocarcinoma

- High hCG levels (reflecting high residual tumour loads)
- Large or multiple secondaries, especially in brain, liver and bowel
- Long interval between the diagnosis and the antecedent pregnancy
- Failure of preliminary drug therapy
- Advanced maternal age
- Blood groups B and AB
- Type of antecedent pregnancy (choriocarcinoma following a normal pregnancy has a worse prognosis than that following a mole)

Advice Related to Contraception

Women with molar pregnancy should be advised to avoid pregnancy until hCG levels have been normal for 6 months following evacuation of a molar pregnancy and for 1 year following chemotherapy for GTN. There has been some controversy regarding the use of oral contraceptive pills (OCPs) as a method of contraception following evacuation, until the serum β-hCG levels have returned to normal. Some studies suggest that the use of combined OCPs may increase the risk of malignancy in women whose β-hCG titres remain high, whereas other studies have shown no risk. The RCOG advices that the combined pill should not be used until one month after the hCG levels have returned to normal. Most centres from the UK recommend that these women must not take the pills until their hormone concentrations have returned to normal. If the woman had started taking OCPs before the diagnosis of GTD had been made, she should be counselled regarding a small, but an increased risk of developing GTN in case she chooses to remain on oral contraception. The small potential risk of using emergency hormonal contraception (comprising of progesterone), in women with raised β-hCG levels, is outweighed by the potential risk of pregnancy to the woman. Emergency contraception is considered acceptable as the benefits in preventing pregnancy outweigh the potential risks. An intrauterine contraceptive device (IUCD) is inadvisable as it may cause bleeding which may be confused with the presence of persistent disease. Also, insertion of IUCD before the normalisation of hCG levels may be associated with the risk of uterine perforation. Surgical methods or barrier contraception prove useful in these cases. Also, presently there is no evidence regarding any adverse effect of single-agent progestogens on GTN. RCOG advises that other hormonal methods (e.g. the progesterone only pill, Mirena, Depot, etc.) can be used irrespective of the β-hCG levels. Since pregnancy should definitely be avoided in these cases, RCOG advises the use of barrier contraception (condoms with spermicidals).

Hormone Replacement Therapy

Hormone replacement therapy may be used safely once hCG levels have returned to normal.

Management of Twin Pregnancy with Molar Gestation

Twin pregnancy, where one foetus is normal and other one is molar gestation mole, can be allowed to continue. These pregnancies tend to be complicated by miscarriage, pre-eclampsia, venous thromboembolism, bleeding, and increased risk of persisting GTD. The chance of getting a healthy baby varies between 25% and 40%.

Invasive Mole

Invasive mole follows a complete mole. It is a histologically benign condition resulting due to the invasion of abnormal trophoblasts into the myometrium. It may also develop due to embolisation of molar tissue through pelvic venous plexus. On histological examination, it looks similar to a complete mole. However, the tumour invades the myometrium, resulting in bleeding and abdominal pain. The invasive mole can invade as far as the bladder and rectum and may even cause intra-peritoneal bleeding. It may produce secondary deposits in the lungs. It may clear spontaneously but may require treatment with hysterectomy. Sometimes, chemotherapy is needed. Nowadays, with modern hCG monitoring and accurate scanning, invasive mole rarely occurs.

Placental Site Trophoblastic Tumour (PSTT)

It is a rare manifestation of GTT, which develops at placental implantation site. These tumours usually originate from the intermediate trophoblastic cells. They may present with a wide spectrum of clinical behaviour, ranging from a self-limited state to persistent disease to a highly aggressive metastatic neoplasm, showing metastases to the lung, liver, peritoneal cavity, brain, etc. Due to the lack of syncytiotrophoblastic tissue, levels of serum hCG are only modestly elevated in PSTTs. It is a malignant, slow-growing tumour, which may present years after the antecedent pregnancy. It may be associated with raised levels of placental alkaline phosphatase. The disease is relatively resistant to chemotherapy but responds well to surgery (hysterectomy) if the disease is localised.

Ectopic Pregnancy

Ectopic means "out of place". In an ectopic pregnancy, the fertilised ovum gets implanted outside the uterus because of which the pregnancy occurs outside the uterine cavity **(Fig. 13.2)**. Most commonly, i.e. in nearly 95% of cases, the fertilised ovum gets implanted inside the fallopian tube. Other extrauterine locations where an ectopic pregnancy can get implanted include the ovary, abdomen or the cervix. Since none of these locations has been equipped by nature to support a growing pregnancy, with continuing growth of the foetus, the gestational sac and the organ containing it may burst open. This can result in severe bleeding, sometimes even endangering the woman's life. Ectopic pregnancy is a relatively rare condition presenting typically with severe lower abdominal pain plus/minus vaginal bleeding. It typically occurs 6 weeks after the last period.

Aetiology

The major cause of ectopic pregnancy is acute salpingitis, accounting for nearly 50% of cases. In nearly 40% of cases, the cause remains unknown. The risk factors for ectopic pregnancy are as follows:
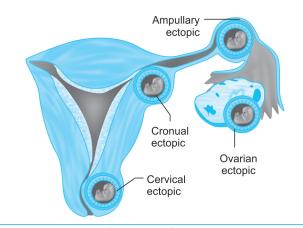
FIG. 13.2: Common sites of occurrence of ectopic pregnancy

- ❑ Prior history of an ectopic pregnancy
- ❑ *Pelvic infections*: History of pelvic infections, such as pelvic inflammatory disease, STD, etc.
- ❑ Salpingitis and tuberculosis are important causes for ectopic pregnancy.
- ❑ Prior surgeries to the fallopian tubes, including tubal reconstructive surgery, tubectomy, etc.
- ❑ Endometriosis and pelvic scar tissue (pelvic adhesions)
- ❑ Congenital abnormalities of tubes
- ❑ Smoking is a risk factor in about one-third of ectopic pregnancies.
- ❑ Patients belonging to the age group of 35–44 years
- ❑ Use of ovulation induction drugs and other assisted reproductive techniques
- ❑ Use of progestin-only pills or progesterone-releasing intrauterine device
- ❑ Intrauterine contraceptive devices
- ❑ Salpingitis isthmica nodosa
- ❑ History of in utero exposure to diethylstilbestrol.

Symptoms

Initial symptoms are very much similar to those of a normal early pregnancy, e.g. missed periods, breast tenderness, nausea, vomiting or frequent urination.

The typical triad on history for ectopic pregnancy includes bleeding, abdominal pain and a positive urine pregnancy test. These symptoms typically occur 6–8 weeks after the last normal menstrual period. Acute blood loss in some cases may result in the development of dizziness or fainting and hypotension.

Many women with ectopic pregnancy may remain asymptomatic.

Clinical Examination

General Physical Examination

- ❑ Normal signs of early pregnancy (e.g. uterine softening)
- ❑ Evidence of haemodynamic instability (hypotension, collapse, signs and symptoms of shock).

Abdominal Examination

❑ Abdominal pain and tenderness
❑ Signs of peritoneal irritation (abdominal rigidity, guarding, etc.) are indicative of ruptured ectopic pregnancy.

Pelvic Examination

❑ Vaginal bleeding may be observed on per speculum examination.
❑ Uterine or cervical motion tenderness on vaginal examination may suggest peritoneal inflammation.
❑ The uterus may be slightly enlarged and soft.
❑ An adnexal mass may be palpated (with or without tenderness).

Investigations

❑ *Blood (ABO and Rh type)*: Rh-negative patients must be injected with 50 µg of anti-D immune globulins to prevent the occurrence of haemolytic disease of the newborn.
❑ *Urine or serum β-hCG levels*: Determination of the levels of β-hCG in the urine or serum helps in establishing the diagnosis of pregnancy. According to the ACOG recommendations (2008), an increase in serum β-hCG levels of less than 53% in 48 hours confirms an abnormal pregnancy. Ectopic pregnancy is suspected if transabdominal sonography does not show an intrauterine gestational sac and the patient's β-hCG level is greater than 6,500 mIU per mL (6,500 IU per L) or if TVS does not show an intrauterine gestational sac and the patient's β-hCG level is 1,500 mIU per mL (1,500 IU per L) or greater. A low rate of increase in hCG levels or a very low level of hCG after the early weeks is most probably due to a failed pregnancy or ectopic pregnancy.
❑ *Imaging studies*: Ultrasound examination in case of an ectopic pregnancy may show a thick, bright echogenic, ring-like structure, which is located outside the uterus, having a gestational sac containing an obvious foetal pole, yolk sac or both. This usually appears as an intact, well-defined tubal ring (Doughnut or Bagel sign). There may be an empty uterus or presence of a centrally placed pseudo-gestational sac. Cystic or solid adnexal or tubal masses **(Fig. 13.3)** and severe adnexal tenderness with probe palpation are also suggestive of ectopic pregnancy. In case of a ruptured ectopic pregnancy, the ultrasonographic findings include presence of free fluid or clotted blood in the cul-de-sac or in the intra-peritoneal gutters.

Various treatment options in case of an ectopic pregnancy are shown in **Figure 13.4**.

FIG. 13.3: Ultrasound showing an ectopic pregnancy of the left tube

Pregnancy of Unknown Location

"Pregnancy of unknown location" can be defined as a situation where the pregnancy test is positive, but there are no signs of intrauterine or extrauterine pregnancy on the TVS. By the time normal pregnancy is producing β-hCG levels greater than 6,500 IU/L, an intrauterine pregnancy can be seen on abdominal scan in almost all cases. Transvaginal scanning is much more sensitive in detecting intrauterine pregnancy and will almost always show a pregnancy with β-hCG levels greater than 1,500 IU/L. Hence, β-hCG levels greater than 1,500 IU/L, with no evidence of an intrauterine pregnancy on a transvaginal ultrasound scan, suggests that it could be present elsewhere. This is called as the "pregnancy of unknown location". The protocol for the management of pregnancy of unknown location usually comprises of serial scans, β-hCG assays and measurements of progesterone levels. Progesterone levels less than 20 nmol/L usually indicate a failed pregnancy, miscarriage or an ectopic pregnancy. Expectant management plan appears to be successful in most of the cases.

Heterotopic Pregnancy

Heterotopic pregnancy is pregnancy in more than one site. It is a rare complication of pregnancy where both intrauterine and extrauterine pregnancies (ectopic pregnancy) co-exist simultaneously. Previously, the natural incidence of heterotopic pregnancy was quite low, about 1:10,000–30,000. However, after IVF, the incidence of heterotopic pregnancy has greatly increased to about 1:100. The ongoing intrauterine pregnancy results in normal β-hCG levels, and the ultrasound scan shows an intrauterine sac and eventually a viable intrauterine pregnancy. Co-existing ectopic pregnancy may produce an adnexal mass and free fluid in the pouch of Douglas.
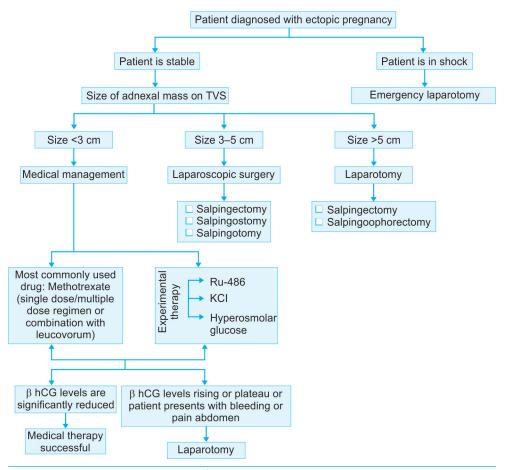
FIG. 13.4: Various treatment options in case of an ectopic pregnancy
Abbreviations: hCG, human chorionic gonadotropin; TVS, transvaginal sonography

Medical Disorders During Pregnancy

Iron Deficiency Anaemia

The WHO defines anaemia as presence of haemoglobin of less than 11 g/dL and haematocrit of less than 0.33 g/dL. Centre of Disease Control (CDC, 1990) has defined anaemia as haemoglobin levels below 11 g/dL in the pregnant woman in first and third trimester and less than 10.5 g/dL in second trimester. Based on the findings of the peripheral smear and the results of various blood indices, anaemia can be classified into three types as shown in **Table 13.7**. Various causes of anaemia during pregnancy include physiological changes during pregnancy; anaemia due to iron deficiency; microcytic anaemia which is resistant to iron due to an underlying asymptomatic bacteriuria, thalassemia, multiple gestation, etc.

Out of the various blood indices used, mean corpuscular volume and mean corpuscular haemoglobin concentration are the two most sensitive indices of iron deficiency. The earliest haematological response to treatment is reticulocytosis. The WHO recommends universal iron supplementation comprising of 60 mg elemental iron and 400 μg of folic acid once or twice daily for 6 months in pregnancy, in countries with prevalence of anaemia less than 40% and an additional 3 months post-partum in countries where prevalence is greater than 40%.

Aetiology

Various causes of iron deficiency anaemia are listed in **Table 13.8**.

The pregnancy imposes a large increase in iron requirements. There is an increased iron requirement during pregnancy amounting to about 1,000 mg. Iron requirements in singleton pregnancy increase from about 2.5 mg per day in the first trimester to about 6.5 mg per day in the third trimester.

The maternal red cell mass rises by about 20% and the baby requires an iron load as well. Iron requirements increase by a factor of two to three times during pregnancy. The average daily requirement of iron is approximately 4 mg.

Clinical Symptoms

With mild anaemia, the woman may present with vague complaints of ill health, fatigue and diminished capability

TABLE 13.7   Classification of anaemia based on the blood values

Type	Lab value	Causes
Macrocytic normochromic anaemia	Increased MCV, normal MCHC (MCV >100 fL; MCHC-34)	Vitamin B12 deficiency; folate deficiency anaemia
Microcytic hypochromic anaemia	Low MCHC; low MCV (MCV <80 fL; MCHC < 30)	Thalassemia; iron deficiency anaemia; anaemia of chronic disease (rare cases)
Normocytic normochromic anaemia	Normal MCHC; normal MCV	Physiological anaemia of pregnancy, anaemia due to chronic disease, anaemia due to acute haemorrhage; aplastic anaemia; haemolytic anaemia

Abbreviations: MCV, mean corpuscular volume; MCHC, mean corpuscular haemoglobin concentration

TABLE 13.8   Causes of iron deficiency anaemia

Nutritional causes
Iron deficiency anaemia (60%)
- Dimorphic anaemia both due to deficiency of iron and folic acid

Haemolytic anaemia
- Haemoglobinopathies

Anaemia due to blood loss
- Acute: Antepartum haemorrhage, post-partum haemorrhage
- Chronic: Hookworm infestation, bleeding piles, malarial infestation

to perform hard labour, loss of appetite, digestive upset, breathlessness, palpitation, dyspnoea on exertion, easy fatigability, fainting, light-headedness, tinnitus, exhaustion, nocturnal leg cramps, headache, paraesthesias and numbness in the extremities, oral and nasopharyngeal symptoms, pica, hair loss, etc.

General Physical Examination

❑ *Pallor*: There may be pallor in lower palpebral conjunctiva, pale nails, pale palmar surface of hands, pale tongue, lips, nail beds, etc.
❑ *Epithelial changes*: The epithelial tissues of nails, tongue, mouth, hypopharynx and stomach are affected resulting in development of nail changes (thinning, flattening and finally development of concave "spoon-shaped nails" or koilonychias), glossitis, angular stomatitis and atrophic gastritis, etc.
❑ *Pedal oedema*: In severe anaemic cases, there may be pedal oedema.
❑ *Abdominal examination*: Splenomegaly may occur with severe, persistent, untreated iron deficiency anaemia.

Investigations

❑ *Haemoglobin and haematocrit*: Haemoglobin is less than 11 g/dL and haematocrit is less than 0.33 g/dL.
❑ *Blood cellular indices*: Abnormalities in various blood indices with iron deficiency anaemia are described in **Table 13.9**. Iron-deficiency anaemia is hypochromic and microcytic.
❑ *Peripheral smear in iron deficiency anaemia*: Peripheral smear of blood shows microcytic and hypochromic cells. There may be anisocytosis (abnormal size of cells) in the

TABLE 13.9   Abnormalities in blood indices with iron deficiency anaemia

Blood index	Normal value	Value in iron deficiency anaemia
MCH	26.7–33.7 pg/cell (average 30.6 pg/cell)	<26.7 pg/cell
MCHC	32–36 g%	<30 g%
MCV	83–97 fL (average 90 fL)	<76 fL
Haemoglobin	12.1–14.1 g/dL	<11 g/dL in 1st and 3rd trimesters and less than 10.5 g/dL in 2nd trimester
Haematocrit	36.1–44.3%	<36.1%
Red cell count	$3.9–5.0 \times 10^6$ cells/µL (average 4.42×10^6 cells/µL)	$<3.9 \times 10^6$ cells/µL or normal
Red cell distribution index	31–36%	<31%

Abbreviations: MCV, mean corpuscular volume; MCHC, mean corpuscular haemoglobin concentration; MCH, mean corpuscular haemoglobin

TABLE 13.10   Changes in serum iron studies with iron deficiency anaemia

Blood parameter	Normal value	Value in iron deficiency anaemia
Serum transferrin levels	200–360 mg/dL	>360 mg/dL
Serum iron concentration	60–175 µg/dL	<60 µg/dL
Transferrin saturation	25–60%	<25%
Ferritin levels	50–145 ng/mL	<20 ng/mL
Serum protoporphyrin	30–70 µg/dL	>70 µg/dL

form of microcytosis and/or poikilocytosis (abnormal shape of cells) in the form of pencil cells and target cells. There may be presence of ring or pessary cells with central hypochromia.
❑ *Osmotic fragility*: RBC osmotic fragility is slightly reduced.
❑ *Serum iron studies*: Changes in the various serum iron parameters are described in **Table 13.10**. Serum ferritin levels are estimated to be the best gauge of total body iron stores. A ferritin level less than 12 µg/L is diagnostic of iron deficiency.

❑ *Stool examination*: This helps in excluding out parasitic infestation as a cause of anaemia.
❑ *Urine routine/microscopy*: Urine routine/microscopy helps in detecting the presence of pus cells/occult blood or schistosomes.
❑ *Haemoglobin electrophoresis*: Haemoglobin electrophoresis and measurement of haemoglobin A2 and foetal haemoglobin are useful in establishing either beta-thalassemia or haemoglobin C or D as the aetiology of microcytic anaemia.
❑ *Bone marrow examination*: A bone marrow aspirate stained for iron (Perls stain) can be diagnostic of iron deficiency.

Management in the Antenatal Period

Dietary changes: Eating a healthy and a well-balanced diet during pregnancy helps in maintaining the iron stores.

Iron supplements: Prescription of oral iron can be considered as the most common form of treatment. If the period of gestation is less than 30 completed weeks of gestation, oral iron supplements (containing 200–300 mg of iron salt with 500 μg of folic acid) must be prescribed in divided doses. Treatment with ferrous sulphate (200 mg tds) in iron deficiency anaemia should result in a rise in haemoglobin levels of 1 g/dL/week after two weeks. The main problems associated with the use of oral iron supplements are occurrence of side effects including anorexia, diarrhoea, epigastric discomfort, nausea, etc.

Previously, it used to be the norm for all pregnant women to be given iron supplements. Currently, it is believed that iron supplements improve the blood picture, but it is not associated with any evidence of clinical benefit. Therefore, there is no requirement of universal iron prescription to all pregnant women. The WHO considers an Hb value of 11 g/dL as the starting point for treatment, but this is not universally agreed. In the UK, haemoglobin level less than 10.5 g/dL are regarded as abnormal and can be considered as an indication for therapy.

Since ferrous salts are better absorbed than ferric, they are more commonly used. Vitamin C increases absorption. Almost all preparations of iron are associated with gastrointestinal side effects, which are mainly dose-related. There are some slow-release preparations that seem to reduce unpleasant symptoms. These iron preparations work by releasing the active substances further down in the bowel. However, most iron is absorbed in the upper duodenum. So, these preparations are much less efficient at delivering iron to the patient.

Sometimes, parenteral iron therapy (by intramuscular or intravenous routes) is started in cases where there is intolerance to oral form of iron; when iron deficiency is not correctable with oral treatment; there is non-compliance on part of the patient; the patient is unable to absorb iron orally or the patient is near term. The two most commonly used parenteral iron preparations include iron sorbitol citric acid complex (Jectofer) and iron dextran (Imferon). IM and IV preparations can be used, and they help in rapidly reloading the depleted iron stores.

Intramuscular iron is effective and not associated with a significant level of side effects or allergic reactions. It has to be administered by deep intramuscular injection using a Z-technique because it is believed that if the injected iron leaks back into the subcutaneous tissues, it may result in much pain and tissue staining. In this technique, the subcutaneous tissue is pulled in one direction and the needle is pierced through the displaced tissue into the underlying muscle.

When the needle is removed, the subcutaneous tissue is released.

Blood transfusion: This may be required when there is not enough time to achieve a reasonable haemoglobin level before delivery; for example, patient presents with severe anaemia beyond 36 weeks; there is acute blood loss or associated infections and anaemia is refractory to iron therapy.

Complications due to Anaemia

Maternal
❑ *Throughout the pregnancy*: These include high-maternal mortality rate, cerebral anoxia, cardiac failure, increased susceptibility to develop infection, abortions, preterm labour, etc.
❑ *During antenatal period*: Poor weight gain, preterm labour, PIH, placenta praevia, accidental haemorrhage, eclampsia, premature rupture of membranes (PROM), etc.
❑ *During intra-natal period*: Dysfunctional labour, intranatal haemorrhage, shock, anaesthesia risk, cardiac failure, etc.
❑ *During post-natal period*: Post-natal sepsis, sub-involution, embolism.

Foetal
❑ Pre-term birth, low-birth-weight and intrauterine growth restriction (IUGR) babies
❑ Foetal distress and neonatal distress requiring prolonged resuscitation
❑ Impaired neurological and mental development
❑ Tendency of the infants to develop conditions, such as iron deficiency anaemia, failure to thrive, poor intellectual development, delayed milestones and other morbidities later in life.

Megaloblastic Anaemia

Megaloblastic anaemia used to be common previously. However, it is no longer common nowadays, due to improved diet and folate supplementation. At the present

time, it is almost always due to folate deficiency, rarely due to that of B_{12}. Risk factors for the development of megaloblastic anaemia are listed in **Table 13.11**.

Folate deficiency during pregnancy can cause serious neural tube defects and other developmental anomalies. Folate supplementation, 1 month before conception and then throughout the first trimester, helps in the prevention of neural tube defects.

Sickle Cell Disorders in Pregnancy

Sickle cell disease shows an autosomal recessive mode of inheritance. It results from an amino acid substitution of glutamine for valine on the beta globin chain of the haemoglobin molecule. Highest incidence of sickle cell disorders is observed in the African and West Indian populations. Conditions such as lowered oxygen tension, acidosis, infection and dehydration may precipitate a crisis. Sickle cell disorders are associated with an increased incidence of hypertension during pregnancy. Therefore, women with sickle cell disease need to be regularly screened for hypertension/pre-eclampsia, urinary tract infection and reduced foetal growth. Pre-pregnancy or first trimester screening is recommended in these cases so that the couple can be advised on the possible risk of a serious haemoglobin defect in their offspring and subsequently counselled about prenatal diagnostic options.

Intrauterine Growth Restriction

Intrauterine growth restriction refers to low-birth-weight infants whose birth weight is below the 10th percentile of the average for the particular gestational age. Even if the infant's birth weight is less than 10th percentile, he/she may not be pathologically growth restricted. These infants are termed as small for gestational age and have simply failed to achieve a specific weight or biometric size in accordance with the gestational age. However, if the growth restriction is due to some pathological process (either intrinsic or extrinsic), it is known as IUGR. IUGR can be due to several maternal, foetal or placental causes as described next.

Maternal: Maternal causes are as follows:
❏ Constitutionally small mothers or low maternal weight
❏ Excessive alcohol intake
❏ Strenuous physical exercise
❏ Poor socio-economic conditions (IUGR is inversely related to social class)
❏ Maternal anaemia, especially sickle cell anaemia
❏ Tobacco smoking, drug abuse during pregnancy (though smoking increases the risk for IUGR, it reduces the risk for hypertensive diseases)
❏ Chronic placental insufficiency due to pre-eclampsia, chronic hypertension, renal disease, connective tissue disorders, gestational diabetes, etc.

TABLE 13.11	Risk factors for the development of megaloblastic anaemia

• High parity
• Low income
• Multiple pregnancies
• Epilepsy
• Patients with malabsorption

❏ Maternal hypoxia (e.g. pulmonary diseases, cyanotic congenital heart disease, etc.)
❏ Endocrine disorders (e.g. diabetic nephropathy, hyperthyroidism, Addison's disease).

Foetal: Various foetal causes are as follows:
❏ Multiple pregnancy
❏ Congenital malformations (e.g. congenital heart disease, renal agenesis, etc.)
❏ Chromosomal abnormalities (e.g. trisomy 13, 16, 18, 21, etc.)
❏ Chronic intrauterine infection, e.g. congenital syphilis, TORCH, viral, bacterial, protozoal and spirochetal infections.

Placental: Placental abnormalities including chorioangioma, circumvallate placenta, marginal or velamentous cord insertion, placenta praevia, placental abruption, etc. may also be responsible.

Epilepsy and Pregnancy

All drugs used for treating epilepsy during pregnancy should be regarded as potential teratogens in clinical practice. The main risks associated with these drugs are neural tube defects, facial clefts and cardiac abnormalities. The risk of anencephaly is not particularly increased. Sodium valproate and carbamazepine are particularly associated with neural tube defects, with incidences up to 4% and 1% respectively. There is also the risk of foetal anti-convulsant syndrome, comprising one or lesser abnormalities such as abnormal teeth, low-set ears, wide nose, nail hypoplasia etc. There are now also concerns about intellectual and educational impairment of the foetus. The baby may be at an increased risk of developing epilepsy. The overall risk of developing epilepsy is about 0.5–1% but is up to 5% in the children of mothers with epilepsy. If a sibling has epilepsy, the risk goes up to 10%. If both parents have epilepsy, the risk is up to 20%.

Some anti-epileptic drugs impair vitamin K production, increasing the risk of maternal and neonatal bleeding. Vitamin K administration to the baby is recommended. Seizures carry significant risk to the mother. There is no evidence that a single seizure is harmful to the baby. However, a single episode of status epilepticus can be harmful.

The greater the number of drugs used to treat the mother, the greater the foetal risk. Also, the higher the dosage of the drugs, the greater the risk to the foetus. Newer drugs, e.g. gabapentin, levetiracetam and tiagabine may be safer, but the data in humans is limited. Lamotrigine appears to carry similar risks to the older drugs. Cessation of drug therapy in a seizure-free patient should be in consultation with a neurologist, after appropriate counselling related to the risks of further seizures, with an interval of at least 6 months before conception. Folic acid in the dosage of 5 mg daily should be prescribed, though its efficacy remains unproven.

There is no "safest drug" to be used during pregnancy, but it is hoped that the newer drugs, gabapentin, levetiracetam and tiagabine, may prove safer. Detailed foetal screening for abnormality is necessary. Caesarean section is not required because of the epilepsy per se, unless there is status epilepticus. There is increased drug turnover and some recommend that drug levels be monitored regularly, perhaps monthly.

Others take the view that if the woman is fit-free and the levels are not elevated, less frequent monitoring is satisfactory, e.g. once each trimester. There is debate regarding the mode of management in individuals with drug levels below the normal range. If the patient is fit-free, the best policy would be not to interfere with drug dosages. However, in cases where the patient has been experiencing fits, the drug dosage must be adjusted to bring the level within the therapeutic range.

She should be told that her drug levels will be monitored during the pregnancy and that the dosage may need to be increased. The dosage would then be gradually returned to normal after the pregnancy. Breastfeeding is acceptable with almost all of the drugs, so she should be encouraged to go for it.

Many of the anti-epileptic drugs induce cytochrome P450 enzymes that reduce vitamin K levels in mother and baby. There is a small risk of neonatal bleeding. Most advise that the mother be given vitamin K (10 mg daily) in the last month of pregnancy. This may also result in the reduced effects of drug therapy on hormonal contraception. Effective contraception should be provided. Contraception is an issue, particularly hormonal contraception, which is rendered less effective by anti-epileptic drugs. The progesterone only pill is also affected.

Patients should be taught about airway maintenance and nursing in the "recovery position". They should ensure that family, friends and work colleagues are appropriately trained.

Regarding the patient who has had no fits for years, the general advice is that she should be advised to continue with drugs if further fits are likely. If she does stop, she has to be informed of the risks of a further seizures and its consequences. If a fit were to occur, it is most likely that it will be in the first 6 months, so the usual advice is to wait for 6 months before conceiving.

Cardiac Disease During Pregnancy

Mitral stenosis is a frequent complication (90%) of rheumatic valvular heart disorder during pregnancy. The New York Heart Association (NYHA) classification of heart disease is based on the physical abilities of the mother and is divided into four classes. The NYHA classification system has been described in details in Chapter 14. Nearly 90% cases belong to the milder categories (class 1 and 2). On the other hand, cases belonging to the serious categories (class 3 and 4) account for only 10% of heart disease in pregnancy. However, these cases are responsible for nearly 85% of cardiac-caused deaths.

In case of the mother with congenital heart disease during pregnancy, the foetus is also at an increased risk of developing congenital heart disease. The foetus has a greater risk of congenital heart disease when the abnormality is present on the maternal side rather than the paternal one. There is also an increased incidence of prematurity and IUGR.

Diabetes in Pregnancy

Approximately 2–5% of the total pregnancies may be affected by diabetes. Amongst the pregnancies complicated by diabetes, nearly 65% cases involve gestational diabetes, whereas 35% cases are associated with pre-existing diabetes. Diabetes in pregnancy is associated with an increased rate of miscarriage, congenital abnormalities, perinatal mortality, perinatal morbidity and maternal complications during pregnancy. Women who develop diabetes during pregnancy are said to have gestational diabetes. Gestational diabetes is defined by the WHO as "carbohydrate intolerance resulting in hyperglycaemia of variable severity with the onset or first recognition during pregnancy." Gestational diabetes now includes both gestational impaired glucose tolerance and gestational diabetes mellitus (GDM). Some women with gestational diabetes may remain diabetic after delivery of the foetus, while others may return back to normal. Women with insulin-dependent DM have significantly lower levels of maternal α-feto-protein and unconjugated oestriol in comparison to the women without diabetes. This information is therefore required to prevent an excessively high rate of false positive test results on performing triple test amongst diabetic women. Early diagnosis and treatment of gestational diabetes is especially important because good glycaemic control from before conception helps in significantly reducing almost all of the complications associated with gestational diabetes.

Management

Modern diabetic management involves obtaining strict control of the blood sugar levels before and during the pregnancy, at the same time avoiding the occurrence of hypoglycaemic episodes.

Pre-pregnancy Advice

All diabetic women of reproductive age must be educated about the benefits of good glycaemic control starting before conception, the requirement for pre-pregnancy counselling, and the requirement for effective contraception to prevent unplanned pregnancy. The National Institute for Health and Care Excellence (NICE) guidelines (March 2008) recommend that women with diabetes who plan pregnancy should adhere to the following advice:

❏ They should be provided with a structured education programme
❏ They should have individualised targets for glucose control
❏ The aim must be to keep their HbA1c levels below 6.1%, because this will help reduce the incidence of congenital abnormalities. They should be told to avoid pregnancy if the HbA1c is greater than 10%.

Screening for Diabetes

Universal screening for gestational diabetes is not recommended. Glucose challenge test is a screening test for gestational diabetes, in which plasma blood glucose levels are measured 1 hour after giving 50 g glucose load to the woman, irrespective of the time of the day or last meal. It is not necessary for the woman to follow a special diet before test or to be in the fasting stage. The timing for this test depends on the woman's likely risk of developing gestational diabetes during her pregnancy. This test need not be routinely performed in women at low risk for diabetes, must be performed at 24–28 weeks in women with average risk of diabetes and as soon as possible in women at high risk for diabetes. A value of 140 mg/dL or higher indicates high risk for development of gestational diabetes. An abnormal result on glucose challenge test must be followed by a 100 g oral glucose tolerance test. While a 100 g, 3-hour GTT is a standard in the US, in the UK, a 75 g, 2-hour GTT is preferred. However, if the glucose challenge test shows plasma glucose values larger than 200 mg/dL, there is no need for glucose tolerance test.

Seventy-five grams oral glucose tolerance test (OGTT) is performed in the morning after the patient has had at least three days of unrestricted diet comprising of greater than 150 g of carbohydrates. Firstly, a fasting blood sample is taken, following which the patient is advised to drink 75 g of anhydrous glucose in 150–300 mL of water over the course of 5 minutes. The second blood sample is taken 2 hours following the glucose load. If two or more of these values on GTT are abnormal, the patient has gestational diabetes **(Table 13.12)**. The patient cannot be diagnosed as being a gestational diabetic, if only one value is abnormal. However, she is at an increased risk for developing complications, such as macrosomia (18%) and pre-eclampsia (7.9%). Even patients who show no abnormal values in their 3-hour GTT have risks of 6.6% and 3.3% for development of macrosomia

TABLE 13.12	World Health Organization criteria for 75 g OGTT			
	Whole blood venous	Whole blood capillary	Plasma venous	Plasma capillary
Fasting	≥6.1 mmol/L	≥6.1 mmol/L	≥7.0 mmol/L (126 mg/dL)	≥7.0 mmol/L
2 hours	≥6.7 mmol/L	≥6.7 mmol/L	≥7.8 mmol/L (140 mg/dL)	≥8.9 mmol/L

Abbreviation: OGTT, oral glucose tolerance test

and pre-eclampsia respectively. This shows that even small alteration in maternal carbohydrate metabolism may have a significant impact on the foetus.

In high-risk cases, the glucose testing must be performed as soon as possible. Some of these cases include:

❏ Body mass index (BMI) >30 kg/m^2
❏ History of delivery of previous baby having a weight greater than 4.5 kg
❏ History of gestational diabetes
❏ First-degree relative with diabetes
❏ Strong family history of type II DM
❏ Ethnicity (origin in India, Pakistan, Bangladesh, Caribbean and Middle East).

Antenatal Care

Insulin requirements rise with increasing period of gestation because pregnancy is a diabetogenic state associated with the development of insulin resistance. The following physiological changes are responsible for the diabetogenic state of pregnancy:

❏ Increased production of steroid hormones (especially corticosteroids, oestriol and progesterone) and human placental lactogen that are produced late in pregnancy show an anti-insulin effect.
❏ Some insulin may be destroyed by the placenta and kidneys.

Under normal circumstances, women are able to make more insulin for meeting the increased requirements. However, women who already have impaired glucose tolerance or insulin resistance may be unable to do so and may develop gestational diabetes. One must also remember that the insulin requirements fall rapidly after delivery. Therefore, all efforts must be taken to prevent the occurrence of hypoglycaemia following delivery.

Antenatal care in patients with GDM should preferably be hospital-based, involving a multidisciplinary team approach comprising of an endocrinologist, obstetrician and the paediatrician. The blood glucose levels need to be assessed every 1–2 weeks throughout pregnancy. HbA1c should be measured monthly and kept below 6.1. In case HbA1c levels are greater than 10, the pregnancy outcomes are likely to be poor. The rates of congenital abnormalities in

these cases are likely to be particularly high. The two main aims for management of diabetic patient during pregnancy are as follows:

1. Maintenance of blood glucose levels
2. Regular foetal monitoring.

During pregnancy, fasting blood sugar levels should lie between 3.5 mmol/litre and 5.9 mmol/litre and 1-hour postprandial levels be less than 7.8 mmol/litre. Above these levels, there is an increase in the rate of all the complications, particularly perinatal mortality rates and sudden foetal death in utero. On the other hand, with exceptionally tight control, e.g. blood sugars less than 5.6 mmol/L, hypoglycaemia tends to occur, outweighing the benefits.

Women also require good glycaemic control during labour and delivery along with an early initiation of feeding of the baby to prevent neonatal hypoglycaemia. Women must be counselled regarding the increased risk of the baby being admitted to the neonatal unit, and of the risks of the baby developing diabetes/obesity as an adult.

❏ *Requirement for hypoglycaemic therapy*: The clinician needs to substitute oral hypoglycaemic drugs with insulin. In case of previously diabetic women, oral diabetes medication needs to be changed to insulin. In case of women with gestational diabetes, initial control of blood glucose levels must be through diet and nutritional advice. If these options do not work, insulin may be advised.

The insulin regimen needs to be individualised and the patient must be advised to check their blood glucose levels at least four times a day. The women must be encouraged to monitor their blood glucose regularly and to adjust their insulin dosage in order to maintain their blood glucose levels within the normal (non-diabetic) range. The aim should be to maintain her HbA1c levels below 6.1%. HbA1c should not be used routinely for assessing glycaemic control in the second and third trimesters of pregnancy.

Diabetic ketoacidosis is a serious complication that can cause foetal death at any stage. Urine of all diabetic women should be tested for ketones, especially if their blood glucose levels are high, if vomiting occurs or if they are unwell.

❏ *Retinal assessment during pregnancy*: Pregnant women with pre-existing diabetes should be offered retinal assessment by digital imaging with mydriasis using tropicamide.

This can be offered following their first antenatal clinic appointment and again at 28 weeks if the first assessment is normal. If at any stage diabetic retinopathy is present, an additional retinal assessment should be performed at 16–20 weeks. Women, in whom pre-proliferative diabetic retinopathy has been diagnosed during pregnancy, should have ophthalmological follow-up for at least 6 months following the birth of the baby. Diabetic retinopathy should not be considered a contraindication to vaginal birth.

❏ *Renal assessment during pregnancy:* Diabetic nephropathy is a progressive disease that can be divided into the following stages:
- Micro-albuminuria (incipient nephropathy), which can be defined as albumin:creatinine ratio of ≥3.5 mg/mmol or albumin concentration of ≥20 mg/litre.
- Macro-albuminuria or proteinuria (overt nephropathy), which is defined as albumin:creatinine ratio of ≥30 mg/mmol or albumin concentration of ≥200 mg/litre as a result of widespread glomerular sclerosis
- End-stage renal disease, which is associated with decreasing creatinine clearance, increasing serum creatinine levels and uraemia.

Women with diabetic nephropathy are at increased risk of adverse pregnancy outcomes, in particular IUGR, chronic hypertension, pre-eclampsia and pre-term birth. If renal assessment has not been undertaken in the preceding 12 months in women with pre-existing diabetes, it should be arranged at the time of first contact in pregnancy. If serum creatinine is abnormal (≥120 mmol/litre) or if total protein excretion exceeds 2 g/day, referral to a nephrologist should be considered. Estimated glomerular filtration rate as used during the pre-conceptional period should not be used during pregnancy.

❏ *Diabetes education and information*: The obstetrician needs to discuss information regarding the effect of diabetes on pregnancy; importance of blood glucose control at the time of pregnancy; changes in the hypoglycaemic therapy during and after birth; complications related to use of insulin therapy (e.g. hypoglycaemia); advice regarding timing, mode of delivery and its management; management of the baby after birth; and advice related to early parenting (including breastfeeding and initial care of the baby); contraception and follow-up.

❏ *Calorie restriction*: Women with gestational diabetes whose pre-pregnancy BMI was between 25.9–29.9 kg/m² should be advised to restrict their calorie intake to 25 kcal/kg/day or less; women with the normal body weight (BMI between 18.5–24.9 kg/m²) must consume approximately 30 Kcal/kg/day. Calorie consumption must be approximately 35–40 Kcal/kg/day for women who are underweight (BMI < 18.5); 20 Kcal/kg/day per day for women with moderate and severe obesity (BMI between 30.0–49.9) and 12 Kcal/kg/day for morbidly obese women (BMI ≥ 50).

This total calorie intake must be distributed in form of multiple small evenly spaced meals and snacks throughout the day, e.g. three small meals in morning, afternoon and night and three snacks in mid-morning, mid-afternoon and a bedtime snack. The bedtime snack is particularly important as it helps in avoiding overnight hypoglycaemia

and ketosis. Of the total calorie intake, 40–50% must come from carbohydrates; 30–40% from fats (two-thirds of which should be unsaturated fats and remaining one-third should be saturated fats); and 15–20% must come from proteins.

❏ *Hypoglycaemic therapy in antenatal period*: Most cases of gestational diabetes will respond to changes in diet and exercise. Hypoglycaemic therapy should be considered for women with gestational diabetes if diet and exercise fail to maintain blood glucose targets during a period of 1–2 weeks. Previously, the use of oral hypoglycaemic agents was not recommended during pregnancy due to the safety concerns to the foetus and the risk of development of foetal hypoglycaemia. This is so as most of the oral hypoglycaemic agents are capable of crossing placenta. However, since long, there is a requirement of safe and highly effective oral agent for the treatment of gestational diabetes. Several small prospective and retrospective studies have demonstrated the effectiveness and probable safety of the oral hypoglycaemic sulfonylurea (glyburide) in the treatment of gestational diabetes. Treatment with glyburide can be considered as a practical alternative for women who are either unable to take insulin or are non-responsive to it. Metformin, a biguanide compound (glucophage) may be considered as another option for women with gestational diabetes. The objective of the treatment is to maintain the fasting capillary glucose values under 95 mg/dL and 1 or 2-hour postprandial values under 140 mg/dL and 120 mg/dL, respectively.

❏ *Insulin therapy*: Insulin therapy may include regular insulin, or rapid-acting insulin analogues (aspart and lispro). Presently, there is insufficient evidence regarding the use of long-acting insulin analogues (insulin glargine, protamine zinc insulin, etc.) during pregnancy. Therefore, isophane insulin (intermediate-acting insulin) is used during pregnancy. The National Service Framework for Diabetes (NSF for diabetes, 2002) recommends that women with type 2 diabetes who require treatment with oral hypoglycaemic agents and are planning to become pregnant or are already pregnant should be transferred to insulin therapy because of the theoretical risk associated with these drugs crossing the placenta. If the patient has already been taking drugs such as angiotensin-converting enzyme (ACE) inhibitors, ARBs (angiotensin-II receptor blockers) and statins, these should be stopped. Folic acid, 5 mg daily should be prescribed.

Thyroid Disorders During Pregnancy

Thyrotoxicosis occurs in about 1 pregnancy in 500. Hypothyroidism occurs in about 1 pregnancy in 100. Both carry risks for the foetus and neonate. Thyrotoxicosis carries bigger risk to the mother. The major changes related in thyroid function during normal pregnancy are increase in serum thyroxine-binding globulin (TBG) concentrations; stimulation of the thyrotropin receptor by chorionic gonadotropin; TBG excess resulting in an increase in the concentration of both serum total thyroxine (T4) and triiodothyronine (T3), whereas free serum T4 and T3 concentrations remain within normal range. Moreover, the levels of thyroid-stimulating hormone (TSH) do not change during pregnancy. Maternal thyroxine is transferred to the foetus throughout pregnancy. This hormone is important for normal foetal brain development, especially before the development of foetal thyroid glands.

Aetiology

❏ *Hyperthyroidism*: The commonest cause is Grave's disease, which accounts for about 95% cases of thyrotoxicosis. All such patients should be stabilised before pregnancy. It is an autoimmune condition, in which antibodies are produced to the TSH receptors in the thyroid cells. These antibodies stimulate the receptors resulting in an increased thyroid activity. The condition is characterised by exophthalmos, evidence of thyroid overactivity and "peau d'orange" appearance of the skin in which the skin is thickened and the pores become prominent.

The condition often improves as the pregnancy proceeds because pregnancy is a state of relative immunosuppression. However, the condition reverts back in the puerperium. Diagnosis is based on assay of TSH and T4. T4 is raised due to stimulation of the thyroid by autoantibodies. As a consequence of thyroid stimulation, there occurs a fall in TSH levels.

❏ *Hypothyroidism*: Hypothyroidism affects about 1% of pregnancies. Hashimoto's disease is the commonest cause of hypothyroidism followed by treated Graves' disease. Hashimoto's disease is an auto-immune condition, featuring the presence of microsomal autoantibodies. An important clinical feature is goitre. Hypothyroidism can also result from surgery, radioactive iodine, drugs, etc. Drugs which can result in hypothyroidism include carbimazole and thiouracil, radioactive iodine, amiodarone, lithium, etc.

De Quervain's thyroiditis is an inflammatory thyroiditis, which can result in hypothyroidism. It is thought to be of viral origin and is the commonest cause of a painful thyroid. There may be initial thyrotoxicosis, then temporary hypothyroidism.

With adequate replacement using thyroxine, the outcomes for the mother and baby are good. With severe disease, pregnancy is unlikely. The foetus is unable to produce its own thyroxine before 12 weeks. So, there are concerns about embryonic brain development in presence of inadequate maternal thyroxine levels. Worldwide, the biggest problem related to hypothyroidism is congenital cretinism due to iodine deficiency. In these cases, the

baby is born with hypothyroidism and goitre. In the UK, all newborn babies must be screened for hypothyroidism as part of the Guthrie test. This is a heel prick bloodspot test.

Post-partum thyroiditis is a condition that may develop in approximately 10% of the individuals during the post-partum period. It usually presents with hyperthyroidism. This is followed by hypothyroidism that may last for some months. Goitre may be present in approximately 50% cases. These women are predisposed to hypothyroidism in later life. Affected women may present with postnatal depression. All women with postnatal depression should be screened for thyroid function.

Clinical Presentation

❏ *Hypothyroidism*: The various symptoms include dry skin with yellowing, especially around eyes, hair loss, weakness, tiredness, fatigue, hoarseness, constipation, sleep disturbance, depression, cold intolerance, muscle cramps, weight gain, oedema, dry skin, prolonged relaxation phase of deep tendon reflexes and/or a pathologically enlarged thyroid gland or goitre (in cases of endemic iodine deficiency or Hashimoto's thyroiditis).

❏ *Hyperthyroidism*: The various symptoms include palpitations, nervousness, irritability, breathlessness, tachycardia, tremors in hands, heat intolerance, insomnia, increased bowel movements, light or absent menstrual periods, weight loss, muscle weakness, warm moist skin, hair loss, nervousness, etc. There is an association of thyrotoxicosis with hyperemesis gravidarum (HG). Patients with excessive vomiting in early pregnancy should have their thyroid function checked. Hydatidiform mole is also particularly associated with thyroid overactivity.

Investigations

❏ *Hyperthyroidism*: The diagnosis of hyperthyroidism in pregnant women should be based primarily on serum TSH value less than 0.01 mU/litre and a high serum free T4 value.

❏ *Hypothyroidism*: Hypothyroidism is characterised by low serum T4 levels and raised TSH levels.

Management

❏ *Hyperthyroidism*: Thioamides [propylthiouracil (PTU), methimazole and carbimazole] are recommended for the treatment of moderate to severe hyperthyroidism complicating pregnancy. Beta-blockers may be given to ameliorate the symptoms of moderate to severe hyperthyroidism in pregnant women.
  • Carbimazole and thiouracil are the drugs commonly used, carbimazole in the dosage of 15–40 mg daily, and thiouracil in the dosage of 150–400 mg daily.

The initial dose is usually gradually reduced after a month to six weeks to maintenance levels of carbimazole, 5–15 mg daily, and thiouracil, 50–150 mg daily. If treatment is being started in pregnancy, thiouracil is preferred. Though the evidence is limited, carbimazole is linked to a small risk of teratogenicity. Rare cases of embryopathy, including aplasia cutis, have been reported with use of methimazole (carbimazole) during pregnancy. No such cases have been reported with PTU use during pregnancy. Therefore, PTU may be more appropriate for patients with Graves' disease who are in their first trimester of pregnancy. However, thiouracil has been linked with a small risk of liver diseases. It is advised that the patient be closely monitored for signs of liver problems, especially in the first 6 months of treatment.

Carbimazole is also linked to agranulocytosis or bone marrow suppression. Therefore, clinician needs to remain vigilant in patients on carbimazole who develop signs of infection and the need to stop treatment if there is neutropenia. Both drugs can cause skin rashes in up to 5% individuals. Both drugs cross the placenta and reach the baby, thereby causing hypothyroidism. Therefore, the baby needs to be closely observed for the signs of hypothyroidism.

  • *Breastfeeding*: Carbimazole and thiouracil are excreted in small amounts in breast milk. Usually, they are of no consequence and breastfeeding is not contraindicated. However, the baby needs to be observed, particularly if the mother requires high drug doses or breastfeeding is prolonged.

  • Ablation with radioiodine is absolutely contraindicated during pregnancy due to the possibility of the ablation of foetal thyroid tissue as well, which is usually present by 10–12 weeks of gestation.

❏ *Hypothyroidism*: Replacement therapy with levothyroxine is administered in the dosage of 1–2 μg/kg/day (approximately 100 μg/day) in cases of hypothyroidism.
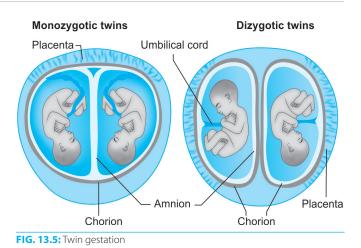
Complications

❏ *Hyperthyroidism*: This includes complications such as spontaneous abortion or miscarriage, premature labour, low birthweight, stillbirth, pre-eclampsia, heart failure, thyroid storm (rarely), foetal or neonatal hyperthyroidism or hypothyroidism, with or without a goitre, non-immune hydrops and foetal demise. The biggest risk to the mother is the rare "thyroid storm". This can cause heart failure and death in up to 30% cases.

❏ *Hypothyroidism*: This includes complications such as pre-eclampsia, gestational hypertension, placental abruption, non-reassuring foetal heart rate tracing, preterm delivery, low birthweight, increased rate of caesarean section, neuropsychological and cognitive

impairment, post-partum haemorrhage (PPH), and overall increased rate of perinatal morbidity and mortality.

Serum TSH is the most reliable indicator of genuine hypothyroidism. Overt hypothyroidism complicating pregnancy is unusual because hypothyroidism may be associated with anovulation, as well as a high rate of first trimester spontaneous abortion.

Conditions Specific to Pregnancy

Obstetric Cholestasis

There is no agreed definition for obstetric cholestasis (OC). It can be generally defined as presence of pruritus and abnormal liver function tests (LFT) that resolve during the puerperium. The diagnosis of OC is made once other causes of pruritus and abnormal LFTs have been excluded out. Intense pruritus often occurs on the palms and soles. Usually, there is no rash, unless one inflicted by scratching. If there is a rash, a dermatological opinion must be taken. Elevated bile acids is a parameter widely used for the diagnosis of OC in the UK. However, it is not mandatory for the diagnosis. Araucanian women have the world's highest incidence of OC (approximately 5%). It shows a wide geographic variation, being commonest in Chile, Scandinavia and SE Asia. OC probably results due to an oestrogenic effect. The incidence is highest in third trimester, because oestrogen levels peak during this time. There is a risk of recurrence on oestrogen-containing contraception.

Obstetric cholestasis is likely to cause premature delivery and late gestation foetal death in utero. Presently, there is no way of identifying the "at-risk baby" and no proven effective treatment is available. According to RCOG, there is no evidence to support early delivery and it thinks that it is reasonable to prescribe vitamin K, 10 mg daily, from the time of diagnosis. It has further advised that "the timing and risks of delivery should be discussed on an individual basis".

Ursodeoxycholic acid and dexamethasone possibly have a beneficial effect. However, there is insufficient research to support their routine use.

Multiple Pregnancy

Development of two or more embryos simultaneously in a pregnant uterus is termed as multifoetal gestation. Development of two foetuses (whether through monozygotic or dizygotic fertilisation) simultaneously is known as twin gestation **(Fig. 13.5)**; development of three foetuses simultaneously as triplets; four foetuses as quadruplets; five foetuses as quintuplets and so on.

FIG. 13.5: Twin gestation

Dizygotic twins are more common than the monozygotic twins. Difference between monozygotic and monozygotic twins is described in **Table 13.13**. Monozygotic twin cannot be reliably distinguished from dizygotic twins by the naked-eye examination of the foetal membranes and placentae. Monozygotic twins can be monochorionic and/or monoamniotic. Dizygotic twins are dichorionic and diamniotic.

Aetiology

The risk factors, which are most likely to result in multi-foetal gestation, include the following: increased maternal age and parity; previous history of multifoetal gestation; family history of multifoetal gestation (especially on maternal side); conception following a long period of infertility; pregnancy attained through the use of assisted reproductive technology (in vitro fertilization or use of clomiphene citrate); racial origin (multifoetal gestation is more common amongst the women of West African ancestry); history of using progestational agents or combined oral contraceptives, which may cause reduction in the tubal mobility; previous history of multifoetal gestation, etc.

Complications

Maternal Complications during the Antenatal Period

❑ Spontaneous abortion
❑ *Anaemia*: Due to increased iron requirement by two foetuses, early appearance of anaemia is a common complication.
❑ *Fatty liver of pregnancy*: It is rare complication that occurs more often in multifoetal than in singleton pregnancies.
❑ Hyperemesis gravidarum
❑ Polyhydramnios
❑ Pre-eclampsia
❑ Placenta praevia (due to increased placental size, the chances of placental encroachment upon the lower segment of the uterus greatly increases)

TABLE 13.13	Difference between monozygotic and dizygotic twins	
Parameter	Monozygotic twins (identical twins)	Dizygotic twins (non-identical or fraternal twins)
Aetiology	Division of a fertilised ovum into two	Fertilisation of two or more ova by sperms
Sex	Same	Can be different
Placenta	Single	Each foetus has a separate placenta
Communication between foetal vessels	Present	Absent
Genetic features (DNA fingerprinting)	Same	Different
Blood group	Same	Different
Skin grafting	Acceptance by the other twin	Rejection by the twin
Intervening membrane between the two foetuses	Composed of three layers: a fused chorion in the middle surrounded by amnion on two sides	Composed of four layers: two chorions in the middle surrounded by amnion on two sides
Foetal growth and congenital malformations	More common	Less common
Incidence	Comprises of one-third of total cases of twins	Comprises of two-third of total cases of twins
Frequency	The frequency of monozygotic twin births is relatively constant worldwide and is approximately one set per 250 live births	Variable
Influence of various factors	Though the occurrence of monozygotic twins is largely independent of factors such as race, heredity, age and parity, there is now increasing evidence that assisted reproductive technology increases the incidence of zygotic splitting	The incidence of dizygotic twinning is greatly influenced by factors such as race, heredity, maternal age, parity, nutrition and fertility treatment

❑ Antepartum haemorrhage
❑ Pre-term labour (minus 2 weeks from term for each extra child)
❑ Varicosities, dependent oedema.

Labour

❑ Foetal malpresentation
❑ Vasa praevia
❑ Cord prolapse
❑ Premature separation of placenta, resulting in abruption placenta
❑ Cord entanglement
❑ Post-partum haemorrhage
❑ Dysfunctional uterine contractions
❑ Increased operative interference.

Puerperium

❑ Sub-involution
❑ Infection
❑ Failure of lactation
❑ Miscarriage
❑ *Prematurity*: The most frequent neonatal complications of pre-term birth are hypothermia, respiratory difficulties, intracranial bleeding, hypoglycaemia, necrotising enterocolitis, infections and retinopathy of prematurity, low-birthweight babies, etc.
❑ *Congenital anomalies*: These can especially occur with the monozygotic twin pregnancies.
❑ Intrauterine death
❑ Intrauterine growth restriction

TABLE 13.14	Degrees of polyhydramnios
Grading	Criteria
Mild	Largest vertical pocket of liquor measures 8–11 cm
Moderate	Largest vertical pocket of liquor measures 12–15 cm
Severe	Largest vertical pocket of liquor measures >16 cm

❑ *Foetal complications specific to twin gestation*: These may include complications such as discordant growth, twin-to-twin transfusion, acardiac twin or twin reversed arterial perfusion syndrome, conjoined twins, etc.

Hydramnios

Polyhydramnios or hydramnios is defined as presence of amniotic fluid volume of 2,000 mL or greater at term. In late pregnancy, liquor is mainly formed from foetal urine. There is also a significant contribution of fluid from the foetal lungs. Equilibrium is affected due to the swallowing of liquor by the baby. Hydramnios results due to disruption of this equilibrium resulting in increased production of liquor or due to reduced swallowing. The commonest type of hydramnios is of mild to moderate severity and has a chronic onset. Most cases are idiopathic. Acute hydramnios is associated with twin-twin transfusion in monochorionic pregnancy. Severe hydramnios is uncommon, forming nearly 5% of the total cases. These cases are associated with a high incidence of underlying pathology. The varying degrees of polyhydramnios **(Table 13.14)** are based on the measurement of the largest vertical pocket of liquor.

Aetiology

In about two-thirds of cases, the cause of polyhydramnios is unknown. Polyhydramnios is more likely to occur due to the causes listed in **Table 13.15**.

TABLE 13.15	Aetiology of hydramnios

Maternal causes

Multiple pregnancy
• Monochorionic twin pregnancy is often accompanied by excess liquor
• Twin-twin transfusion

Medical causes
• Poorly-controlled diabetes (maternal hyperglycaemia leads to foetal hyperglycaemia and increased foetal urine production).
• Maternal/foetal infection
– Parvovirus infection
– Toxoplasmosis
– Cytomegalovirus
– Syphilis

Foetal causes

Increased urine production
• Heart failure
• Congenital heart anomaly
• Arrhythmia, congenital heart block
• "Shunts" as in sacro-coccygeal tumour
• Severe foetal anaemia:
– Rhesus disease
– Foetomaternal bleeding
– Parvovirus infection
– Alpha-thalassemia
• Assorted foetal renal conditions:
– Finnish congenital nephrosis
– Bartter's syndrome

Decreased swallowing of liquor
• Central causes: Anencephaly (no brain, no swallowing)
• Mechanical problems:
– Upper gut atresia (oesophageal or duodenal)
– Pressure effects (head and neck tumours, masses in the chest)
  - Cystic adenomatoid malformation
  - Diaphragmatic hernia
  - Bronchial cysts
– Masses in the abdomen
– High airway obstruction syndrome: Congenital stenosis of the pharynx, larynx or their occlusion due to pressure from external masses
• Neurological problems:
– Myotonic dystrophy
– Arthrogryposis (presence of multiple joint contractures at birth)

Neural tube and skin defects:
• Neural tube defects
• Abdominal wall defects:
– Exomphalos
– Gastroschisis

Chromosomal problems: Trisomy 13, 18 and 21

Placental causes
• Placental tumours (e.g. chorioangioma, which is a vascular malformation of the placenta)
– These are rare and presumably just leak fluid.

Clinical Presentations

Women with minor polyhydramnios experience few symptoms. Those who are more severely affected may experience the following symptoms:
❑ Difficulty in breathing
❑ Presence of large varicosities in the legs and/or vulva
❑ Presence of new haemorrhoids or the worsening of those present previously.

Abdominal Examination

❑ Abdomen is markedly enlarged, along with fullness of flanks.
❑ The skin of the abdominal wall appears to be tense, shiny and may show appearance of large striae.
❑ Clinically, the patients have a fundal height greater than the period of amenorrhea.
❑ Foetal heart sounds may appear muffled as if coming from a distance.
❑ A fluid thrill may be commonly present.
❑ It may be difficult to palpate the uterus or the foetal presenting parts due to presence of excessive fluid.

Investigations

Most important investigation, which helps in establishing the diagnosis of hydramnios, is ultrasound examination. Nowadays, the ultrasound diagnosis of hydramnios is based on the calculation of "amniotic fluid index" (AFI) or evaluation of the "deepest vertical pool" (DVP). AFI correlates best with liquor volume. For the AFI, the deepest pool of liquor in each quadrant is measured. The values are added together to give the AFI. The largest of these four measurements is the DVP. A DVP of 8 cm or more is diagnostic. Hydramnios is defined as an AFI greater than 95th centile for that particular gestational period. Once the diagnosis of hydramnios has been made on ultrasound, other investigations must be directed towards detection of the underlying cause. These include the following:
❑ A GTT must be done to exclude diabetes.
❑ A detailed ultrasound scan is essential to look for evidence of abnormality, both structural abnormalities and pointers of aneuploidy.
❑ Amniocentesis or cordocentesis may be needed to exclude aneuploidy.
❑ Maternal blood should be taken for infection screen.
❑ Cordocentesis may be needed to confirm foetal infection.
❑ Maternal blood should be taken for Rhesus and Kell antibodies.
❑ Middle cerebral artery (MCA) velocimetry has been accepted as the gold standard for diagnosis and monitoring of foetal anaemia.

Management

Mild degree of hydramnios usually resolves on its own without any treatment. No active management may be

required in patients with asymptomatic hydramnios. In symptomatic patients, management should be in consultation with a multidisciplinary team, including an anaesthetist, paediatrician, etc. In a case of acute or severe hydramnios, the patient must be referred to the foetal medicine expert.

Maternal Treatment

❑ Usually no treatment is required for maternal symptoms.
❑ Simple analgesics may prove useful.
❑ Bed rest can be advised, but the clinician must remain vigilant regarding the development of deep vein thrombosis.
❑ *Treatment of the underlying cause*: Intrauterine surgery has been done to deal with severely stenotic foetal aortic or pulmonary valves. Significant foetal arrhythmia may cause heart failure. In these cases, drugs, which can correct or slow arrhythmias, can be used for treatment. Digoxin can be used to improve bradycardia and to correct heart failure.
❑ *Symptomatic treatment*: In cases of severe and acute hydramnios, the symptoms may demand the following treatment options:
  • *Indomethacin*: Administration of indomethacin helps to reduce foetal urine production. Though indomethacin has been known to be effective for many years, there are concerns that this may have long-term adverse effects on renal function. There are also concerns about its ability to cause premature closure of the ductus arteriosus.
  • *Serial drainage of amniotic fluid/amnio-reduction*: Amnio-reduction involves the drainage of various volumes of liquor. The process has to be repeated several times as the liquor is rapidly replaced.
  • Early delivery.

Foetal Treatment

❑ Correction of foetal anaemia via cordocentesis
❑ Administration of steroids for foetal lung maturation
❑ Antibiotics for treatment of infection like syphilis
❑ Surgical and medical treatments associated with foetal medicine.

Complications

Maternal

❑ Respiratory compromise
❑ Antepartum and post-partum haemorrhage
❑ Abnormal foetal presentations
❑ Uterine dysfunction, gestational diabetes
❑ Increased incidence of operative intervention
❑ Increased risk of premature delivery and PROM
❑ Increased risk of placental abruption and stillbirth
❑ Placental abruption if liquor is released too quickly

❑ Premature rupture of the membranes and premature labour.

Foetal

❑ Increased perinatal mortality and morbidity, mainly due to:
  • Increased rates of PPROM and prematurity
  • Increased foetal abnormality rate
❑ Abnormal lie
❑ Cord prolapse
❑ Macrosomia and shoulder dystocia, especially in presence of maternal diabetes.

Pregnancy-Induced Hypertension

According to the working group report on high blood pressure in pregnancy (2000), pre-eclampsia can be considered as a potentially serious disorder, which is characterised by high blood pressure (>140/90 mmHg) and proteinuria (>300 mg/dL or >1+ on the dipstick). It usually develops after the 20th week of pregnancy and goes away after the delivery (usually within the 12th post-partum week).

Aetiology

The exact pathophysiology of pre-eclampsia is not yet understood. The most likely causes for pre-eclampsia and their underlying mechanisms are as follows:
❑ Inadequate trophoblastic invasion
❑ Maternal inflammatory response resulting in vascular endothelial dysfunction
❑ *Hereditary factors*: The exact genetic defect or pre-eclampsia gene has yet not been identified
❑ *Immunological factors*: Reduced production of blocking antibodies to various placental antigenic sites is associated with an increased risk
❑ Endothelial dysfunction and vasospasm due to increased sensitivity to the action of angiotensin II (vasopressor) and due to the imbalance in production of various prostaglandins.

Clinical Presentation

❑ *Increased blood pressure*: Presence of an increased BP (>140/90 mmHg) for the first time during pregnancy, after 20 weeks of gestation.
❑ *Proteinuria*: Proteinuria is defined as significant if the excretion of proteins exceeds 300 mg per 24 hours or there is persistent presence of the protein (30 mg/dL or 1+ dipstick) in a random urine sample in the absence of any evidence of urinary tract infection.
❑ *Oedema*: Oedema of hands and face can commonly occur amongst women with pre-eclampsia. Since oedema is a universal finding in pregnancy, it is not

considered as a criterion for diagnosing pre-eclampsia. Oedema is often present but is not a diagnostic feature of pre-eclampsia. However, pathological or severe oedema in association with other signs helps with diagnosis.

❏ *Other symptoms*: Indicators of severe pre-eclampsia during pregnancy are headache, visual problems, epigastric or right upper quadrant abdominal pain, oliguria (urine volume <500 mL/24 hours), hyper-reflexia, clonus, convulsions, etc.

❏ *Shortness of breath or dyspnoea*: This could be reflective of pulmonary oedema or acute respiratory distress syndrome.

❏ *Reduced foetal movements*: This is especially observed in association with IUGR and oligohydramnios.

❏ *Weight gain*: Weight gain of more than 2 pounds per week or 6 pounds in a month can be considered as significant.

❏ *Abdominal examination*: There may be evidence of placental insufficiency in the form of oligohydramnios and/or IUGR.

❏ *Placental abruption*: Hypertension in pregnancy is associated with an increased incidence of placental abruption and associated bleeding.

Investigations

❏ *Haematocrit and CBC*: The decrease in blood volume in pre-eclampsia can lead to a rise in maternal haemoglobin concentration resulting in an increased haematocrit.

❏ *Platelet count*: Platelet count of less than $150–400 \times 10^9$/litre could be indicative of the haemolysis, elevated liver enzymes and low platelet count (HELLP) syndrome.

❏ *Kidney function tests*: In severe pre-eclampsia, raised serum creatinine and uric acid levels are associated with a worsening outcome both for the mother and the baby.

❏ *Liver function test*: An aspartate aminotransferase (AST) level of above 75 IU/L can be considered as significant.

❏ *Ophthalmoscopic examination*: The abnormalities include presence of retinal oedema, constrictions of the retinal arterioles and alteration of normal ratio of vein:arteriolar diameter. Grading of hypertensive retinopathy is described in **Table 13.16**.

❏ *Foetal growth restriction*: Patients with pre-eclampsia, medically treated or not, are usually screened by serial ultrasound measurements to detect foetal growth restriction. Poor placental perfusion and diminished foetal renal perfusion can result in reduced liquor

TABLE 13.16	Grading of hypertensive retinopathy
Grade	**Features**
Grade 1	Arterial narrowing
Grade 2	Arteriovenous nipping
Grade 3	Haemorrhages and exudates
Grade 4	All of the above plus papilloedema

production in these cases. This may be evident in form of oligohydramnios on ultrasound examination.

Obstetric Management

Many cases of mild to moderate hypertension can be successfully managed on an outpatient basis. However, hospital admission is required in severe cases.

❏ *Anti-hypertensive medication*: The aim of treatment must be to maintain diastolic BP between 95 mmHg and 105 mmHg.

• Drug therapy: The most commonly used first-line drugs include hydralazine (for severe hypertension), alpha-methyldopa, labetalol and nifedipine. Use of diuretics, ACE inhibitors and angiotensin II receptor blockers should be preferably avoided during pregnancy. Nifedipine has been used in cases of severe hypertension. However, concern remains about the possibility of muscle relaxation causing uterine haemorrhage. Hydralazine has long been used in the acute situation and has a good safety record. Methyldopa has been used since long and is not associated with significant risk. It is probably the hypotensive drug of first choice. Side effects of alpha-methyldopa include depression, nasal congestion, oedema, pyrexia, haemolytic anaemia, weight gain, gastrointestinal disturbance, arthralgia, parkinsonism, nightmares, gynaecomastia, galactorrhoea, hepatitis, etc. Beta-blockers, such as atenolol, are also commonly used anti-hypertensive agents. Diuretics, which were popularly used some decades ago as anti-hypertensive agents are no longer preferred for use in pregnant women because they were shown to reduce maternal plasma volume. They might have an adverse effect on foetal weight and may increase perinatal mortality. Labetalol has been widely used and is believed to be acceptably safe.

❏ *Magnesium sulphate*: Magnesium sulphate should be considered for women with pre-eclampsia, especially in whom there is concern about risk of eclampsia. Magnesium sulphate has been very popular in the USA for managing severe pre-eclampsia and eclampsia since the publication of the Magpie trial. It has become a standard in most of the UK hospitals' protocols for management of eclampsia and severe pre-eclampsia. A total dose of 14 g is administered in form of loading (4 g IV) and maintenance dose (5 g in each buttock by deep IM injection).

❏ *Conservative therapy*: Initial therapy for mild-to-moderate pre-eclampsia is bed rest until foetal maturation becomes adequate. Prescription of the sedatives or tranquilizers is not required. Low-dose aspirin in the dose of 60 mg daily can be prescribed.

❏ *Maternal monitoring*: In severe cases, blood pressure should be checked at every 15 minutes in the beginning,

until the woman has stabilised and then after every 30 minutes in the initial phase of assessment. This can be later increased to 4-hourly intervals and then twice daily. Urine should be carefully monitored for proteinuria twice daily. In the presence of significant proteinuria, a 24-hour estimation of urine proteins may be done. Maternal weight must be measured every day. Tests, like platelet count, kidney function tests and tests of liver function must be done at the time of admission and then twice weekly.

❑ *Foetal surveillance*: Some of the tests for foetal surveillance include daily foetal movement count; weekly measurement of fundal height and abdominal girth in order to detect IUGR; weekly non-stress test; ultrasound examination for evaluating the period of gestation; biophysical profile and/or serial assessment of liquor after every 2 weeks and Doppler ultrasound at every 3–4 weekly intervals. The frequency of the tests can be increased depending upon the severity of the condition.

❑ *Timing of delivery*: The only way to cure pre-eclampsia is to deliver the baby. The pregnancy should not be allowed to continue beyond EDD. In cases of severe pre-eclampsia, when the pregnancy is more than 32 weeks of gestation, delivery is the treatment of choice. Prophylactic steroids should be given to induce foetal lung maturity in case the period of gestation is less than 32 weeks.

❑ *Mode of delivery*: Severe pre-eclampsia per se is not an indication for caesarean section. If the cervix is ripe, labour can be induced by using intravenous oxytocin and artificial rupture of membranes (ARM). In case of presence of unfavourable cervix or other complications (e.g. breech presentation, foetal distress, etc.), a caesarean section may be required.

❑ *Management in the postpartum period*: Ergometrine, given intravenously or intramuscularly post-partum in combination with oxytocin, should be withheld. Use of ergometrine must be avoided during third stage of labour because it may worsen an elevated BP. Therefore, oxytocin alone is preferred. Anti-hypertensive drugs should be given if the BP exceeds 150 mmHg systolic or 100 mmHg diastolic in the first 4 days of the puerperium.

Complications

Some of the maternal and foetal complications related to pre-eclampsia are enumerated in **Table 13.17**.

Eclampsia

Eclampsia can be defined as the onset of tonic-clonic convulsions in a pregnant patient with pre-eclampsia, usually occurring in the third trimester of pregnancy, intra-partum period or more than 48 hours post-partum.

TABLE 13.17	Complications related to pre-eclampsia
Maternal	**Foetal**
• HELLP syndrome (**h**aemolysis, **e**levated **l**iver enzymes and **l**ow **p**latelet count) • Abruption placenta • Cerebral haemorrhage • Sepsis/shock • Eclampsia • Pre-eclampsia in subsequent pregnancies • Impaired renal/liver function tests • Pulmonary oedema • Maternal death	• Oligohydramnios • Intrauterine death • Premature delivery (before 37 weeks of gestation) • Intrauterine growth restriction • Intrauterine asphyxia and acidosis • Pre-eclampsia in subsequent pregnancies • Infant death

It accounts for approximately 50,000 maternal deaths worldwide annually and is relatively uncommon in United Kingdom.

Eclampsia is most common during the antepartum period, with most cases presenting in the third trimester of pregnancy. However, 20–25% of cases occur during the post-partum period—usually within the first 48 hours following delivery. Although seizures may occur as long as 3 weeks post-partum, the majority of cases (98%) occur on the first post-partum day. Eclampsia is thought to be related to cerebral vasospasm, which can cause ischaemia, disruption of the blood brain barrier and cerebral oedema.

Symptoms

❑ *Premonitory stage*: In this stage, there is unconsciousness; twitching of the muscles of face, tongue and limbs; and rolling and fixation of eyeballs.

❑ *Tonic stage*: There is tonic spasm of the body muscles.

❑ *Clonic stage*: There is alternate contraction and relaxation of the skeletal muscles. Twitching starts from the face onto the extremities and soon involves the whole body.

❑ *Coma*: This may be present for a brief period or may persist for a longer time.

Obstetric Management

❑ Immediate care involves maintenance of airway, oxygenation, maintenance of circulation, and prevention of trauma or injury to the patient. An IV line must be secured and the patient must be given IV Ringer's lactate or 0.9% normal saline solution. The patient should be shifted to the eclampsia room. Injury to the patient can be prevented by placing her on a railed bed and using a tongue blade to prevent her from biting her tongue.

❑ Monitoring of vitals including pulse, blood pressure, respiratory rate and oxygen saturation needs to be done at every 15 minutes. Parameters, such as knee jerks, fluid intake and urine output need to be monitored at every half-hourly interval.

❏ Treatment of choice for convulsions is the administration of magnesium sulphate. In order to prevent the occurrence of eclampsia, women with severe pre-eclampsia (BP >160/110 along with proteinuria) should be given magnesium sulphate as a prophylactic measure.
❏ Once the patient has stabilised, an obstetric examination must be performed, and foetal status must be evaluated and plan to deliver the patient as soon as possible must be made.
❏ Continued foetal monitoring is required until the baby is delivered.
❏ Intra-partum management: Strict blood pressure monitoring must be continued throughout labour. Eclampsia per se is not an indication for caesarean delivery. In case the cervix is not favourable, labour can be induced using vaginal prostaglandins and oxytocin infusion. Second stage of labour should be cut short.
❏ Following delivery, close monitoring should be continued for a minimum of 24 hours.

Complications

Maternal

❏ Injuries due to fall from bed, tongue bite, etc.
❏ *Pulmonary complications*: These include pulmonary oedema, pneumonia, adult respiratory distress syndrome, embolism, etc.
❏ Hyperpyrexia
❏ Cardiac: Acute left ventricular failure
❏ Renal failure
❏ Hepatic necrosis
❏ *Cerebral complications*: Cerebral anoxia, cerebral oedema, cerebral dysrhythmia, cerebral haemorrhage, neurological deficits, etc.
❏ *Visual complications*: These could be due to retinal detachment or occipital lobe ischaemia.
❏ *Haematological complications*: These include thrombo-cytopenia, disseminated intravascular coagulation, etc.
❏ *Post-partum complications*: These include shock, sepsis and psychosis.

Foetal

❏ These are same as those described with pre-eclampsia.

Hyperemesis Gravidarum

There is no universally agreed definition for HG. It occurs in up to 1% of pregnancies. The most commonly used explanation of HG is occurrence of severe vomiting causing loss of greater than 5% of body weight. Persistent vomiting related to hyperemesis may be associated with one of the following features: loss of weight, ketosis, dehydration, disturbed electrolytes: low potassium and sodium, raised liver transaminases, low blood urea level, etc. The presence

of ketones in the urine tends to confirm the diagnosis. It occurs in up to 1% of pregnancies.

Hyperemesis gravidarum is usually related to increased β-hCG levels. Onset is in the first trimester. Most cases of HG resolve with rehydration and intake of small meals at regular intervals. IV rehydration should preferably be with saline or Hartmann's solution, not dextrose solution. Fluids containing carbohydrates must not be administered.

Complications are rare but may include maternal death, deep vein thrombosis, pulmonary embolism, IUGR, stillbirth, hypokalaemia, hyponatraemia, Mallory-Weiss oesophageal tears, Wernicke's encephalopathy due to thiamine (vitamin B_1) deficiency, Korsakoff's psychosis, etc. NICE recommends the use of ginger and acupuncture. If drugs are to be used, NICE recommends anti-histamines, e.g. cyclizine. Second-line drugs that are believed to be effective and safe include chlorpromazine (phenothiazine) and metoclopramide (a dopamine antagonist). If indigestion is a problem, cimetidine, omeprazole and ranitidine can be used.

Risk factors for hyperemesis are as follows:
❏ Multiple and molar pregnancies
❏ Hyperthyroidism
❏ HG in a previous pregnancy
❏ Positive family history of HG
❏ History of motion sickness and nausea on the pill
❏ Down syndrome (DS) pregnancies carry a higher risk (This could be related to high hCG levels). Pregnancies with DS behave as if they were less mature in relation to biochemical screening and AFP and hCG levels.

The investigations, which must be done in these cases, include ultrasound scan (to rule out molar pregnancy); mid-stream urine analysis for ruling out urinary tract infection; urine analysis for ketones and specific gravity; thyroid function tests, etc.

Simple antacids sometimes help if indigestion and heartburn are the problems. If indigestion is a problem, cimetidine, omeprazole and ranitidine can be used. They are effective and are believed to be safe.

Screening for Foetal Anomalies

Pre-implantation Genetic Diagnosis

Pre-implantation genetic screening is increasingly used in embryos produced by IVF prior to implantation. It is particularly used for the detection of X-linked disorders, single gene defects or chromosomal abnormalities. The conditions screened for include, cystic fibrosis, Tay-Sachs disease, haemophilia, fragile X syndrome, etc. It is primarily used in women above 35 years of age, when semen has been obtained using intracytoplasmic sperm injection (ICSI) and when there are particular genetic disorders.

Pre-implantation genetic diagnosis (PIGD) is used to diagnose abnormalities in a fertilised egg before it is implanted in the mother's uterus.

One or two cells are removed from the surface of the blastocyst and the removed cells are subjected to a molecular analysis—polymerase chain reaction (PCR), fluorescent in situ hybridisation, etc. Therefore, this technique cannot be performed in natural conceptions. This process can prevent the transmission of sex-linked diseases by eliminating all male embryos. This technique is currently available to couples whose offspring are at a high risk (25–50%) for a specific genetic condition due to one or both parents being carriers or affected by the disease. The limiting factor, however, is that few cells (usually only one to two) are available for diagnosis unlike that following amniocentesis or chorionic villus sampling.

Screening for Down Syndrome

All pregnant women must be offered screening for DS. DS screening should be discussed at their first encounter with a health professional. Women should be aware that screening is voluntary. Women should have access to good information about the subject. Informed consent is mandatory before undertaking screening for DS. There are three basic ways to screen:

1. First trimester biochemical screening involving assay of pregnancy-associated plasma protein A (PAPP-A) and hCG
2. Second trimester biochemical screening involving the assay of maternal serum alpha-fetoprotein (MSAFP), hCG, oestriol and inhibin.
3. Ultrasound for measuring nuchal translucency (NT) is another test for screening, which is not good enough to be used on its own.

Previously, biochemical screening in the second trimester had been the mainstay of screening for DS. It can be performed from 15 weeks to 20 weeks, but the late results are a major weakness. According to the Department of Health (DOH) guidelines, first trimester-combined screening has now become the standard test for screening since 2010. This test includes a combination of NT and first trimester biochemistry (PAPP-A and hCG). This is best done between 11 weeks and 13 weeks, but 14 weeks is the absolute upper limit. Second trimester biochemical screening (triple test or quadruple test) is performed in women who were unable to have first trimester-combined screening (e.g. late booking) or in cases where the clinician is unable to determine the NT because of high BMI or awkward foetal position. Triple test involves with assay of AFP, free β-hCG and oestriol. Some prefer a double test, using only AFP and free β-hCG. Quadruple test includes all the parameters as required for the triple test along with the measurement of serum inhibin levels.

Biochemical testing in the second trimester is mainly to screen for Down syndrome. AFP was the first marker used to improve the age-related risk. The risk of DS is increased by the degree to which the AFP level lies below the norm. In Down syndrome, the hCG and inhibin are high and the others low. However, since the raised levels of MSAFP are associated with an increased risk for neural tube defects, triple test also helps in diagnosing the neural tube defects. One must remember that the pregnancy with raised MSAFP but a normal scan is also a high-risk pregnancy because such pregnancies are associated with an increased risk of IUGR and pregnancy loss. Biochemical screening also gives a risk for Edward's syndrome, trisomy 18. In this condition, all three main markers AFP, free β-hCG and oestriol levels are low.

Groups at an increased risk of Down syndrome include older mothers, women who previously gave birth to a baby with DS, parents with a balanced translocation, etc. Age-related risk for Down syndrome is elaborated in **Table 13.18**. Most commonly associated translocation is these cases are t (14; 21). In these cases, there is one normal 21 chromosome; the second chromosome 21 is a vestigial chromosome with little or no chromosomal material. There is a combined 14 and 21 chromosome, with the chromosome 21 attached onto the chromosome 14. The other chromosome 14 is a normal chromosome. So, the genetic information is "balanced" and the individual is normal.

Levels of Alpha-fetoprotein

As previously mentioned, increased levels of alpha-fetoprotein could be associated with an increased risk of neural tube defects (NTDs). An affected parent or an affected sibling is associated with approximately 5% risk, whereas the risk in presence of two affected siblings increases to about 10%. Recurrence risk is also higher for women who have previously given birth to a child with a neural tube defect (2–3%). If there are two previously affected children, the risk is approximately 6%. The prevalence of NTD is estimated to be around 1 in 1,000 births, but there is much racial and geographic variation. For example, prevalence is higher in Wales in comparison to the rest of England.

Background levels of MSAFP are higher in Afro-Caribbean women. However, on the contrary, they have a lower incidence of NTDs than Caucasians. Laboratory reports must take this into account. Therefore, once the levels are corrected for ethnicity, Afro-Caribbean women have a reduced risk of NTD. The converse is true of women with IDDM and obesity. They have a significantly increased risk of NTD, but lower overall AFP levels.

TABLE 13.18	Age-related risk for Down syndrome
Maternal age (in years)	**Risk of Down syndrome**
20	1 in 1,500
35	1 in 350
40	1 in 100
45	1 in 40

Neural tube defects are a group of defects characterised by presence of an opening in the spinal cord or brain starting from an early period of human development, e.g. spina bifida, anencephaly, encephalocoele, etc. These lesions may occur anywhere along the spine but are more common in the lumbar and sacral regions. Spina bifida is a congenital defect of the spine resulting from the failure of closure of the neural tube at three to four gestational weeks. In cases where the meninges protrude through this defect, it is called a meningocoele, and if neural tissue is involved, it is known as a myelomeningocoele.

The ultrasound diagnosis of spina bifida is based on the evaluation of the foetal spine, which is usually visualised clearly by 16 weeks of gestation in the majority. In presence of neural tube defects, ultrasound examination may show the "lemon sign", in which the foetal frontal bones are distorted. It may also show the "banana sign" in which the shape of the cerebellum is altered.

Polyhydramnios may also be associated with neural tube defects. Children with spina bifida are at risk of having a neuropathic bladder and hence urinary incontinence.

Folic acid taken peri-conceptually is likely to reduce the incidence of neural tube defects by approximately 70%. There is evidence that it also reduces the risk of other congenital abnormalities. The recommended dose for low-risk women is 400 micrograms daily. It should be started pre-conception and continued until at least 12 weeks post-conception. High-risk individuals should be prescribed 5 mg of folic acid daily. Folic acid is thought at least reduce the risk by 50%, but does not eliminate it completely.

Fortification of flour with folic acid has been introduced to good effect in a number of countries. Fortification has been recommended to the DOH by its advisory committees. It led to a greater reduction in the incidence of neural tube defects as well as other congenital abnormalities, e.g. cardiovascular abnormalities, urinary tract obstructive defects, limb defects, congenital pyloric stenosis, etc. Folic acid fortification of flour has been advocated by many authorities including the RCOG in 1997 and Committee on Medical Aspects for Food and Nutrition Policy (COMA), in the UK in 2000. COMA recommended fortification of flour in the dosage of 240 microgram/100 g.

Increased levels of alpha-fetoprotein are also associated with malignancies such as ovarian and testicular malignancies, hepatoma, pregnancy and choriocarcinoma. Occasionally, AFP may be raised in association with breast, pancreatic and secondary hepatic deposits, but levels are usually only slightly elevated.

Routine Ultrasound Scan at 18–20+6 Weeks

A routine ultrasound examination for assessment of congenital anomalies is recommended at 18–20+6 weeks of gestation. The National Screening Committee has recommended six basic measurements that should be taken during this scan. The National Screening Committee has identified 11 foetal anomalies, which must be looked for in this scan. Presence of choroid plexus cysts no longer requires any response. Echogenic bowel, however, requires a response.

Echogenic Bowel

Echogenic bowel is evident in the form of "bright bowel" on ultrasound scan at 20 weeks. Presence of echogenic bowel is an uncommon finding, approximately 1 case in 200 pregnancies. Most of the babies will be normal. However, some may present with abnormalities such as aneuploidy, especially trisomy 21, cystic fibrosis, placental insufficiency and foetal infection, particularly cytomegalovirus. Some cases of echogenic bowel may be related to the swallowing of blood by the baby. VACTERL is another rare condition (1 in 6,000 births) which may be associated with echogenic bowel. In these cases, baby has at least three of the following: **V**ertebral defects, **A**nal atresia, **C**ardiac abnormality, **T**racheo-**E**sophageal fistula, **R**enal abnormalities and **L**imb defects. The major risk associated with this condition is IUGR, so close monitoring of growth and foetal well-being is required. Its diagnosis involves comparing the echogenicity of the bowel with adjacent bone, such as the iliac crest. This method of comparison forms the basis of most modern definitions. Meconium peritonitis has to be excluded as the main differential diagnosis.

Anaesthesia in Pregnancy

Epidural Anaesthesia in Labour

Epidural analgesia has presently become a commonly employed technique for providing pain relief during labour. Epidural analgesia should be administered only once the diagnosis of labour has been established and the patient requests for pain relief. It should be provided by practitioners only in settings where facilities for resuscitation are immediately available. This technique involves injection of a local anaesthetic agent into the epidural space (between the dura mater and the ligamentum flavum) in the space between the vertebra L3 and L4 **(Fig. 13.6)**. An indwelling catheter is usually kept in place for repeat injections or continuous infusion.

Indications

Indications for administration of epidural anaesthesia are as follows:
- Provision of pain relief during first and second stages of labour
- Facilitation of patient cooperation during labour and delivery
- Provision of anaesthesia for episiotomy, forceps delivery or extension for caesarean delivery.
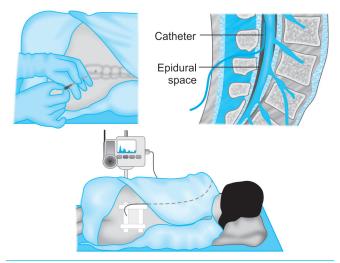
FIG. 13.6: Administration of epidural analgesia

Method of Administration

For the administration of epidural analgesia, under all aseptic precautions, the epidural needle is inserted in the epidural space between the vertebra L3 and L4. Epidural space can be identified by loss of resistance at the time of needle insertion. The epidural space extends downwards from the foramen magnum to the sacral hiatus (at S2) and is triangular in cross-section. The pressure in the epidural space is sub-atmospheric (negative), due to transmission of the sub-atmospheric intrathoracic pressure through the intervertebral foramina. Negative pressure within the space is greatest in the upper and middle thoracic regions and lowest in the lumbar and sacral regions, as distance from the thorax increases. The epidural space contains fat, epidural veins (Batson's plexus), small arteries, lymphatics and spinal nerve roots.

Complications

❏ *Hypotension*: Epidural anaesthesia tends to cause a drop in blood pressure due to pooling of blood in the periphery. The reduced venous return caused by reduced vascular tone and peripheral pooling results in hypotension. These patients are also at an increased risk of developing supine hypotension syndrome. Therefore, administration of fluid pre-load prior to the procedure helps reduce the incidence of hypotension.

    Added to the hypotension related to the procedure is the aortocaval compression, which causes supine hypotensive syndrome in a normal pregnancy. It occurs due to compression of the inferior vena cava by the gravid uterus resulting in a decrease in cardiac output and uterine hypoperfusion. It is relieved by lying on the left side, left tilting of the table or placing a wedge under the right side. Lying supine and presence of uterine contractions make the syndrome worse.

❏ *Increased incidence of malpositions*: Epidural anaesthesia abolishes the tone of the pelvic floor, which is the source of rotation of the foetal head. So, there is an increased incidence of occipito-posterior and transverse positions. So long as mother and baby are well and progress is being made, the second stage should be allowed to take its own time and the mother should not be encouraged to push until the head is on the perineum. Epidurals are associated with an increased rate of forceps deliveries.

❏ *Risk of bleeding*: The administration of epidural is contraindicated in the presence of a coagulopathy because of the risk of bleeding from the venous sinuses and haematoma formation. A combined spinal epidural is helpful for elective deliveries, providing both immediate and postnatal analgesia. Concern about neurological damage from bleeding at the epidural site makes pre-existing coagulation a problem and low platelet count a contraindication for epidural anaesthesia. Most anaesthetists go ahead with platelet count greater than 80,000/μL. Significant bleeding may occur with counts less than 50,000/μL.

    High concentrations of bupivacaine injected via epidural route may cause an increase in the rate of instrumental delivery but not an increase in the rate of caesarean sections. Tinnitus may occur when bupivacaine is given intravascularly or when the plasma levels of bupivacaine reach toxic levels. A total spinal or high spinal block occurs when a large volume of bupivacaine is injected into the subarachnoid space and is a rare complication of an epidural. Epidural bupivacaine does not decrease uterine contractility. Also, epidural anaesthesia is rarely associated with complications such as Mendelson's syndrome and aspiration of stomach contents, which are frequently encountered with general anaesthesia.

Adverse effect on the first stage of labour:

❏ There is some confusion about the possible adverse effect of epidural anaesthesia on the first stage of labour. This arises from the usual policy of not setting one up until labour is definitely established. This is to prevent a woman, who has not become established in labour and subsequently stops having contractions, from having an unnecessary epidural. The general rule is that epidural anaesthesia generally does not affect progress in the first stage of labour. However, the second stage may be prolonged because the epidural abolishes the urge to push. When epidural anaesthesia was first introduced, the rates of instrumental delivery were quite high because of the old rules about the permissible length of the second stage. Presently, the policies that have been implemented allow the second stage to be carried on so long as mother and baby are well and progress is being made. Therefore, the rates of instrumental delivery have considerably reduced.

Nowadays, caesarean deliveries are also increasingly being performed under regional anaesthesia (e.g. epidural and spinal). However, this can be associated with some complications as follows:

❑ *Delayed respiratory depression with hydrophilic opioids*: The addition of opioids to local anaesthetic solutions used in regional anaesthesia is associated with delayed respiratory depression, and this is more likely to occur with hydrophilic opioids than with lipophilic opioids.

❑ *Aspiration of gastric contents*: The risk of aspiration of gastric contents is reduced under regional anaesthesia but it can still occur, especially with a high block or total spinal.

❑ *Venous air embolism*: The incidence of a venous air embolism (VAE) during lower-segment caesarean section under regional anaesthesia is about 25% (using Doppler ultrasound and echocardiography). Thrombus and amniotic fluid emboli have also been reported.

❑ *Evidence of myocardial ischaemia on the electrocardiograph*: The incidence of electrocardiograph (ECG) ischaemic changes is believed to be due to the increase in myocardial work and oxygen demand that occurs secondary to the hypotension induced by the sympathetic blockade.

❑ *Postural headache*: A postural headache usually suggests that there is a cerebrospinal fluid leak close to the level of insertion of the regional block. This may be an indication for an epidural blood patch in order to seal the puncture.

Pethidine Analgesia in Labour

Intravenous pethidine administered via a patient-controlled analgesia pump provides better analgesia compared with nurse-controlled analgesia. However, this results in using almost double the amount of drug. Pethidine causes foetal side effects that include:

❑ Loss of beat-to-beat variability
❑ Depression of the APGAR scores
❑ Respiratory depression.

Pruritus usually occurs due to opiates and not due to pethidine. The analgesic effects of pethidine becomes apparent after 10–15 minutes of administration. Pethidine is 40–60% protein bound.

Entonox

Entonox is a gaseous mixture of nitrous oxide and oxygen and has been used since the 1960s for provision of analgesia during labour. It is twice as effective as pethidine at providing labour analgesia, but inhalation should begin as soon as the uterine contraction is felt, because it takes 45 seconds before the maximum analgesic effect is achieved.

Low-dose isoflurane and sevoflurane have also been administered in addition to entonox, which has demonstrated an increased analgesic efficacy over entonox alone. Combining the analgesic effects of entonox with other analgesics agents provides superior analgesia in comparison to using entonox alone.

Normal Labour and Delivery

Normal labour can be defined as spontaneous onset, low-risk start of labour and remaining so throughout the process of delivery. The infant is born spontaneously in the vertex position between 37 and 42 completed weeks of pregnancy. The mechanisms, which trigger the onset of labour, are still unclear.

Labour comprises of a series of events taking place in the genital organs, which help to expel the foetus and other products of conception outside the uterine cavity into the outer world. It can be defined as the onset of painful uterine contractions accompanied by any one of the following: rupture of membranes; bloody show; cervical dilatation and/or effacement. Braxton-Hicks contractions may occur from 20 weeks and do not necessarily signify the onset of labour.

Labour normally comprises of four stages. Various stages of labour are described in **Table 13.19** and **Figure 13.7**. Artificial rupture of the membranes and syntocinon are often used to augment labour, but by definition, neither is a part of normal labour. Syntocin may be used to augment contractions in the first stage, or contract the uterus in the third stage. Foetal heart rate may be monitored using a cardiotocograph, and a heart rate of 120–160 per minute is normal, as are accelerations after a uterine contraction.

Delay in the Second Stage of Labour

Delay in the second stage of labour may be caused by the following:

❑ The lack of expulsive sensation in the presence of an adequately working epidural can cause the second stage to be prolonged.

❑ Maternal exhaustion, which can be multifactorial, is a common cause of delay in the second stage.

High Foetal Head

Causes of high/floating foetal head are as follows:

❑ *Placenta praevia*: Placenta, which significantly encroaches into the lower uterine segment, prevents engagement of the foetal head.

❑ *Fibroid in the lower uterine segment*: Any "tumour", which obstructs the lower segment, can prevent descent of the foetal head.

❑ *Pre-term infant*: The pre-term infant would not be expected to have engaged in the pelvis and therefore the dating of the pregnancy should be checked. The widespread use of early ultrasound has assisted in the more accurate assessment of pregnancy gestation.

TABLE 13.19	Various stages of labor			
Stages of labour	Description	Characteristics	Duration in primigravida (in minutes)	Duration in multigravida (in minutes)
Stage I	Starts from the onset of true labour pains and ends with the complete dilatation of the cervix	Can be divided into: _Latent phase:_ Slow and gradual cervical effacement and dilatation (up to 3 cm) _Active phase:_ Active cervical dilatation (3 cm to 10 cm) and foetal descent. The active phase should be associated with cervical dilatation at a rate of at least 1 cm per hour. It comprises of: – Acceleration phase – Phase of maximum slope – Deceleration phase	—	—
Stage II	Starts from full dilatation of cervix and ends with expulsion of the foetus from birth canal	—	50	20
Stage III	It begins after expulsion of the foetus and is associated with expulsion of placenta and membranes	—	15	15
Stage IV	Stage of observation which lasts for at least 1 hour after the expulsion of afterbirths	—	60	60
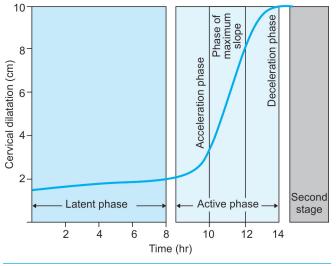
FIG. 13.7: Sages of labour

❑ _African race_: African races commonly have a pelvic inlet with a higher angle of inclination than Caucasian women, and therefore the head may fail to engage before the onset of labour.

In a significant number of women, descent of the foetal head into the pelvis occurs during labour and eventually results in a successful vaginal delivery, and therefore routine caesarean section cannot be justified in all cases of free-floating foetal head in the pelvis. The clinician must, however, keep the possibility of cephalopelvic disproportion in mind in these cases.

Partogram

Normal labour should be plotted graphically on a partograph **(Fig. 13.8)**. The partogram is the best means yet devised for charting labour. Besides plotting the parameters for assessing the progress of labour such as cervical dilatation and descent of the foetal presenting part at regular intervals, many other parameters are plotted on the partogram including maternal pulse, BP and temperature, foetal heart rate, station of the presenting part, strength and frequency of contractions, drugs and fluids administered, colour of liquor, interventions such as ARM, scalp pH measurement, etc.

The partograph is divided into a latent phase and an active phase. The latent phase ends while the active phase begins when the cervix is 3 cm dilated. Cervical dilation and descent of the presenting part are plotted in relation to an alert line and an action line. Alert line starts at the end of latent phase and ends with the full dilation of the cervix (10 cm) within 7 hours (at the rate of 1 cm/hour). The action line is drawn 4 hours to the right of the alert line. Labour is considered abnormal when the cervicograph crosses the alert line and falls on zone II, and intervention is required when it crosses the action line and falls in the zone III. Once slow progress has been identified on the partogram, the clinician must make efforts to exclude obstructed labour, which could be due to causes such as brow presentation, mentoposterior face presentation or an excessively big baby or too small pelvis. Obstructed labour can lead to uterine rupture, particularly in the multiparous woman or one with a scarred uterus. Various causes for abnormal progress of labour are as follows:
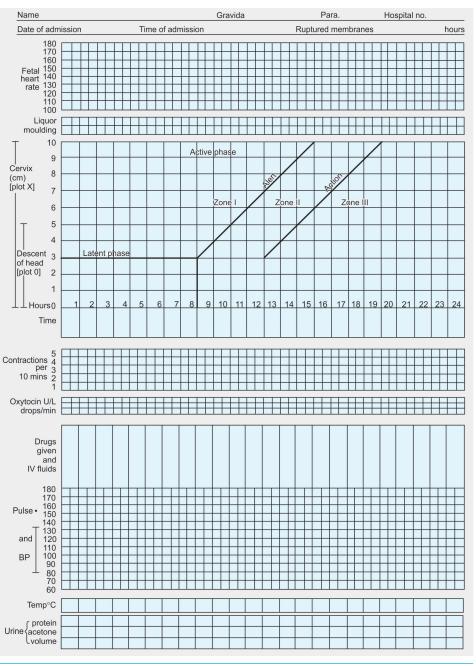
FIG. 13.8: Partograph
Source: World Health Organization (2008)

Abnormalities in expulsive forces: These include:
- Hypotonic uterine dysfunction (uterine inertia)
- Hypertonic uterine dysfunction
- Poor maternal expulsive efforts (related to maternal fatigue or epidural analgesic use).

Foetal abnormalities: These include:
- Abnormalities in foetal size (e.g. foetal macrosomia, with foetal weight >4,000 g)
- Abnormalities in foetal presentation (e.g. brow, shoulder, face, etc.)
- Abnormalities in foetal position (e.g. occiput posterior, occiput transverse, etc.)
- Abnormalities in foetal attitude (extension, asynclitism, etc.)
- Foetal congenital abnormalities (anencephaly, foetal ascites, foetal tumours, etc.).

Pelvic abnormalities: These include cephalopelvic disproportion and cervical dystocia.

Elective Episiotomy

Previously, it was believed that all primigravida patients and all those who previously had episiotomy should have elective episiotomy. Nowadays, it is left for the midwife to

gauge if episiotomy will be of benefit and inflict less trauma than a tear. It may be used to speed up the delivery if there is an evidence of foetal distress. Episiotomy is a routine with breech delivery and forceps, though not with ventouse delivery.

Foetal Distress

Foetal "hypoxia" or "acidosis" are better terms to describe the foetal condition rather than the term "foetal distress".

Indicators of Foetal Distress

Cardiotocograph (CTG) Trace

Indicators of foetal distress are usually evident in form of abnormalities of the foetal heart rate on the CTG trace. These abnormalities can include tachycardia, bradycardia, reduced baseline variability, absence of accelerations, presence of decelerations (especially the late decelerations) and sinusoidal pattern. Electronic monitoring of the foetal heart rate is not a very accurate way for assessing the foetal well-being. Rarely, a compromised baby could have a normal CTG. All doctors and midwives working on labour wards should have 6 monthly training in CTG interpretation. CTGs are usually categorised as normal (reassuring), suspicious (non-reassuring) or abnormal (pathological). Features of various types of CTG trace are described in **Table 13.20**. Normal trace, however, does not guarantee the baby is completely fine, and "suspicious" and "abnormal" trace does not always imply foetal hypoxia.

Meconium Staining of the Liquor

Meconium staining of the liquor is another indicator of foetal distress, which must not be taken lightly because meconium may be inhaled by the baby, resulting in the development of meconium aspiration syndrome. Meconium staining of the liquor may be a problem and could be associated with underlying foetal compromise.

The baby swallows liquor through most of the pregnancy. The residues from this and normal desquamation from the bowel comprise the meconium. The dark black-green colour of meconium comes from bile pigments. Normal meconium is odour-free. Presence of meconium at gestation less than 34 weeks could be associated with *Listeria* infection.

In cystic fibrosis, the meconium is thicker and collects in the ileum. This can sometimes be seen on the 20-week scan as bright echoes—"echogenic" or "bright" bowel. It may present as meconium ileus, characterised by failure to pass meconium, abdominal distension and vomiting. The normal neonate usually passes meconium within a matter of hours and failure to do so within 24 hours implies that the clinician must check the baby to ensure that there is no underlying problem.

Foetal Scalp pH

Presently, there is no perfect method for assessing the baby's condition. Electronic monitoring tends to increase caesarean section rates. The use of scalp pH measurement goes some way to counterbalance this.

The lower limit of normal is usually taken as 7.2 and levels below this are normally grounds for immediate delivery. Levels greater than 7.25 provide reasonable reassurance that the baby is still in good condition. pH measurements can also be done in breech presentation; the blood can be obtained from the buttock.

Management

In case of foetal distress, the following steps must be taken:
- Stopping the syntocinon infusion
- Administration of oxygen to the mother
- Turning the mother on her side to reduce compression of the inferior vena cava
- Tocolytic agent such as ritodrine can be administered to reduce intrauterine pressure and the strength and frequency of uterine contractions.
- Amnio-infusion in cases of oligohydramnios
- Assessment of the scalp pH to assess the baby's condition and possibly intervention by way of caesarean section or instrumental delivery.

Breech Presentation

Breech presentation is a type of abnormal presentation where the foetus lies longitudinally with the buttocks

TABLE 13.20	Features of various types of CTG trace		
Characteristic	**Normal CTG trace**	**Non-reassuring or suspicious trace**	**Abnormal or pathological trace**
Baseline rate	A normal baseline rate (110 to 160 beats per minute)	The baseline is a bit up or a bit down—100 to 109 or 161 to 180 beats per minute	The baseline rate is way off - <100 or >180 beats per minute
Baseline variability	Baseline variability >5 beats/minute	The variability is <5 beats per minute for >40 but <90 minutes	Baseline variability <5 beats per minute for > 90 minutes
Accelerations	At least 2 accelerations per 20 minutes	Early or variable decelerations	a sinusoidal pattern for >10 minutes
Decelerations	No decelerations	Not more than one prolonged deceleration and that should last less than 3 minutes	Abnormal variable decelerations, late decelerations or a deceleration lasting >3 minutes

Abbreviation: CTG, cardiotocograph

presenting in the lower pole of the uterus. The different types of breech presentation include complete breech, footling breech and frank breech. Prematurity is a major risk factor for breech presentation. The incidence of breech presentation declines rapidly from 25% at 28 weeks to 2% or 3% at term. Uterine malformation, such as a bicornuate uterus, is often associated with persistent abnormal lies, such as transverse lie and breech presentation. Hydramnios is also associated with an increased incidence of breech presentation.

Elective caesarean section for breech presentation is now pretty much universal after the publication of the results of "term breech trial" (2000), which demonstrated greater safety for the baby. There is still debate about the best means for delivering the premature breech baby, but most clinicians opt for caesarean section. External cephalic version is also being commonly used nowadays with the hope for reducing the number of breech presentations at term and consequent caesarean sections.

External Cephalic Version

External cephalic version (ECV) can be defined as a procedure in which the clinician externally rotates the foetus from a breech presentation into a cephalic presentation (**Figs 13.9A to D**). External version is associated with the complications listed in **Table 13.21**.

The procedure is usually performed in a special ECV clinic. It is preferable to wait until term (37 completed weeks of gestation) before external version is attempted because of an increased success rate and avoidance of pre-term delivery if complications arise. Tocolytics may be used to relax the uterus. Anti-D needs to be given if the mother is Rh negative. It is normal to do CTGs before and after the procedure. If the first CTG trace is abnormal, the clinician must not proceed with the procedure.

Cord Prolapse

Cord prolapse has been defined by RCOG as descent of the umbilical cord through the cervix alongside the presenting part (occult presentation) or past it (overt presentation) in the presence of ruptured membranes. Breech presentation, prematurity and ECV can be considered as independent risk factors for cord prolapse.
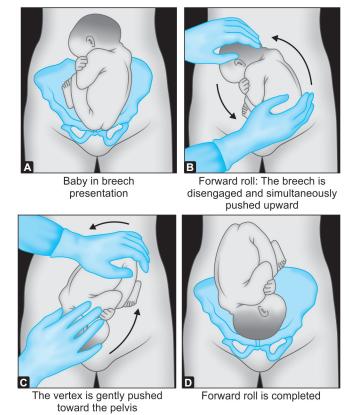
Diagnosis

This is by digital or speculum examination. Speculum examination may be more appropriate at early gestations to reduce the risk of infection. Cord presentation may also be diagnosed on ultrasound examination.

Management

The following steps should be taken for the prevention of cord prolapse:
❏ If there is a significant risk of cord prolapse, e.g. in cases of abnormal or unstable lie, the RCOG's Greentop

FIGS 13.9A TO D: Procedure of external cephalic version

TABLE 13.21	Complications of external cephalic version

- Failure of the procedure
- Antepartum haemorrhage (abruption)
- Onset of premature labour
- Foetomaternal transfusion (resulting in the development of Rh isoimmunisation)
- Foetal distress (leading to an emergency caesarean delivery)
- Foetal bradycardia
- Foetal death

guidelines recommend that the patient's admission should be discussed from 37^{+6} weeks.
❏ Artificial ROM should be avoided whenever possible if the presenting part has yet not engaged or is mobile. In cases where ROM becomes necessary even in such circumstances, this should be performed in an operation theatre with facilities available for an immediate caesarean birth. Suspicion of cord prolapse should be considered if the foetal heart rate or pattern changes significantly following membrane rupture.

Definitive management in cases of cord prolapse comprises of immediate delivery. In cases where vaginal delivery is possible, forceps can be applied in cases of cephalic presentation if the head has engaged. In case of breech presentation, breech extraction can be done. In case of transverse lie, internal version followed by breech extraction must be performed. Caesarean delivery, however,

is preferred in these cases. A caesarean section is the recommended mode of delivery in cases of cord prolapse when vaginal delivery is not imminent. A caesarean section should ideally be performed within 30 minutes or less (from the point of diagnosis to the delivery of the baby).

In cases of cord prolapse where immediate vaginal delivery is not possible, assistance should be called immediately; venous access should be obtained, consent taken and immediate preparations be made for an urgent caesarean delivery. The following steps can be followed until facilities for caesarean section are made available:

❑ Replacing the cord above the presenting part is not recommended due to lack of evidence about its benefit. It is believed that handling the cord increases the risk of vascular spasm, so this should be minimised.
❑ To prevent vasospasm, there should be minimal handling of loops of cord lying outside the vagina, which can be covered with surgical packs soaked in warm saline.
❑ To prevent cord compression, it is recommended that the presenting part be elevated either manually or by filling the urinary bladder with normal saline.
❑ Cord compression can be further reduced by advising the mother to adopt knee-chest position or head-down tilt (preferably in left lateral position).
❑ If there are foetal heart rate abnormalities despite these manoeuvres, tocolytics can be used.

Caesarean Section

Caesarean section is a surgical procedure commonly used in the obstetric practice. In this procedure, the foetal delivery is attained through an incision made over the abdomen and uterus, after 28 weeks of pregnancy. Nowadays, a lower-segment caesarean section is commonly performed.

Previously, a classical caesarean section was commonly performed. In this surgery, a longitudinal incision was administered in the upper uterine segment. A mid-line abdominal incision was commonly given in these cases. Nowadays, a Pfannenstiel incision is commonly used and is much less prone to dehiscence than a mid-line wound.

The main disadvantage of classical caesarean delivery is that the uterine scar tends to rupture in subsequent pregnancies. This may occur without warning and before the onset of labour. Due to this, elective caesarean at 37–38 weeks is a standard for patients with previous classical caesarean section.

Even though lower-segment caesarean delivery has become a norm in the present times, classical operation is still done occasionally. This may be done in cases where the lower segment may not be easily accessible due to fibroids or may be covered in enormous varicosities due to an anterior placenta praevia or in presence of carcinoma cervix. Classical caesarean delivery may be typically done in these cases where the patient is also planning tubal ligation. Classical caesarean delivery may also be done in cases where there may be hesitation in dissecting the bladder off

the lower uterine segment, e.g. if the patient had suffered a vesicovaginal fistula after a previous caesarean section.

Post-Partum Complications

Deep Vein Thrombosis

Extension of puerperal infection along venous route can result in thrombosis and thrombophlebitis of the affected vein. Thrombosis usually originates as an aggregation of platelets and fibrin on the valves in the veins of the lower extremities (especially calf veins). The thrombus can either breakoff and embolise to other veins or cause total occlusion of the veins.

Aetiology

Factors that predispose to the formation of thrombus include endothelial injury, blood stasis and hypercoagulability of blood. Pregnancy is a naturally occurring thrombogenic state associated with an increased concentration of factors VIII, IX and X. Some risk factors for deep vein thrombosis (DVT) are described in **Table 13.22**.

Clinical Presentation

The clinical problem is that the presentation is not at all clear-cut and may be negative in up to 50% of cases. Clinical presentation in cases of DVT may comprise of the following signs and symptoms:

❑ Pain in the calf muscles, oedema of legs
❑ Rise in the skin temperature
❑ Difference in the circumference between the affected and the normal leg may be more than 2 cm
❑ Homan's sign: Homan's sign is positive (Homan's sign is inaccurate and eliciting it may increase the risk of embolisation).

Investigations

❑ *Doppler ultrasound*: This helps in detecting changes in the velocity of blood flow in the femoral veins.

TABLE 13.22	Risk factors for deep vein thrombosis
• Long duration of labour	
• Dehydration	
• Failed instrumental delivery	
• Previous history of VTE	
• Air travel	
• Family history of VTE	
• Surgery	
• Immobility	
• Ovarian hyperstimulation syndrome	
• Factor V leiden mutation	

Abbreviation: VTE, venous thromboembolism

❑ *Duplex Doppler ultrasound*: This is highly sensitive and specific for detection of femoral DVT. Though venograms are the gold standard, ultrasound being an easier investigation is more commonly used.
❑ CT or MRI
❑ *I^{125} fibrinogen scanning*: This is not recommended for diagnosis of DVT in pregnancy due to the risk of radiation exposure to the foetus.

Management

Management in these cases comprises of the following steps:
❑ Bed rest with foot elevation above the level of heart
❑ Analgesics can be used to provide pain relief
❑ Antimicrobial therapy must be started
❑ Anticoagulants such as heparin, low-molecular-weight heparin and oral anticoagulants such as warfarin can also be used
❑ Knee-length or thigh-length graduated elastic compression stockings help in reducing the risk of thrombosis. Early ambulation also helps in reducing the risk
❑ Vena cava filters can be used in the cases where anticoagulant therapy is contraindicated.

Pulmonary Embolus

This condition can be characterised by partial or complete blockage of pulmonary vessels resulting in acute respiratory and/or hemodynamic compromise. Acute respiratory consequences of pulmonary embolism include increased alveolar dead space, hypoxaemia and hyperventilation. Pulmonary embolism can be either acute (embolus is situated centrally within the vascular lumen and is causing its occlusion) or chronic (embolus is eccentric and contiguous with the vessel wall, thereby reducing the arterial diameter by more than 50%). Clinical features are proportional to the size of embolus. Immediate full anticoagulation is mandatory for all patients suspected to have DVT or pulmonary embolism. Pulmonary embolism can be considered as the most important cause for maternal death in developed nations, only after sudden cardiac arrest. Death usually occurs due to shock and vagal inhibition.

Aetiology

Venous thromboembolism is more common in obese patients due to poor mobility and venostasis. Surgeries on the hips and pelvis and trauma to the lower limbs also increase the risk of thromboembolism. A pulmonary infarct may develop in the territory of a pulmonary embolism and will appear as a wedge-shaped area of consolidation on the chest radiograph. Surgical termination of pregnancy is a short procedure and does not increase the risk of thromboembolism.

Clinical Symptoms

❑ Sudden collapse with acute chest pain and air hunger
❑ Tachypnoea, dyspnoea, haemoptysis
❑ Pleuritic chest pain, cough, tachycardia
❑ Temperature of greater than 37°C.

Investigations

All the investigations such as echocardiography (trans-oesophageal), helical MRI, pulmonary angiography, spiral CT, ventilation/perfusion scintigraphy may be used to diagnose pulmonary embolus. In the case of the risk of foetal radiation exposure with spiral CT and angiography, abdominal shields need to be used.

Management

❑ *Patient resuscitation*: This comprises of cardiac massage and oxygen therapy.
❑ *Heparin therapy*: Heparin is usually administered in a bolus dose of 5,000 IU, followed by 40,000 IU/day to maintain a clotting time over 12 minutes in the first 48 hours. Thereafter, heparin levels are regulated to maintain APTT of twice the normal. Warfarin may be started on day one, but it often takes several days to achieve adequate anticoagulation levels (INR 2.0–3.0). Contraindications to systemic anticoagulation for a pulmonary embolus include recent major haemorrhagic trauma, recent central nervous system haemorrhage or infarct, an active gastrointestinal haemorrhage, etc.
❑ Maintenance of blood pressure using dopamine or adrenaline
❑ Thrombolytic therapy using streptokinase may be administered
❑ Tachycardia can be counteracted using digitalis
❑ *Surgical treatment*: The insertion of inferior vena cava filters (for example, Greenfield filter), thrombolysis and surgical embolectomy may be necessary.

Post-partum Haemorrhage

According to the World Health Organization, PPH can be defined as excessive blood loss per vaginum (>500 mL in case of normal vaginal delivery or >1,000 mL following a caesarean section) from the time period extending within 24 hours of delivery and lasting until the end of the puerperium. The ACOG has defined PPH as a decrease in haematocrit by 10% or requirement of blood transfusion 24 hours after the delivery.

The WHO has classified PPH into two: (1) primary PPH and (2) secondary PPH. Primary (PPH) can be defined as blood loss, estimated to be greater than 500 mL, occurring from the genital tract, within 24 hours of delivery. Secondary PPH, on the other hand, can be defined as abnormal bleeding from the genital tract, occurring 24 hours after

delivery until 6 weeks post-partum. Infection is a common cause.

Endocrinology of Lactation

Established lactation provides about 800 mL of milk daily. Breast milk has higher energy content, more lactose and fat, less proteins and low sodium content in comparison to the cows' milk. Its composition varies from feed to feed and even within a feed, with more fat being produced late in the feed. Milk secretion requires prolactin, which is produced in response to suckling. Lactation fails in cases of Sheehan's syndrome as the damaged pituitary cannot produce prolactin. Bromocriptine inhibits the release of prolactin from the pituitary and is therefore useful for the suppression of lactation.

"Let down" reflex, the release of milk, is stimulated by oxytocin, which is released from the posterior pituitary by various stimuli, such as suckling and stimuli such as the baby's cry. Following delivery, secretion of colostrum, a deep, lemon-yellow liquid occurs from the breasts. It contains high amounts of antibodies that help in protecting the newborn. Colostrum contains high quantity of immunoglobulin A, which is part of its protective effect against infection, particularly that of the bowel. Breastfed babies are also at a reduced risk of atopy. Colostrum is secreted for approximately the first two days post-partum; the change to milk occurs on the third and fourth day. Some commonly occurring pathologies in the breast tissue during the puerperal period are described next:

Breast engorgement: Women who do not breastfeed may develop breast engorgement. This is associated with breast pain and milk leakage, which may peak at 3–5 days post-delivery. Treatment comprises of using a well-fitting brassiere, ice packs and oral analgesics for 12–24 hours. Lactation can also be successfully suppressed by demand feeding or manual emptying of the engorged breasts. Bromocriptine to suppress milk production is no longer used.

Mastitis: Mastitis can be defined as the parenchymatous infection of the mammary glands. Mastitis is associated with milk stasis, nipple trauma and poor nursing technique. Pathogenic bacteria most commonly involved are of the staphylococcal type.

Structural Changes in the Newborn

Normal Neonate

The mean red cell volume in a normal neonate is greater than 100 fl. The platelet count is in the normal adult range. The white cell count may be 25×10^9/L.

Presence of ballotable kidneys usually indicates the presence of polycystic disease, but may sometimes be even present in normal neonates. Spleen tips may also be sometimes identified in normal newborns. The femoral epiphysis may develop and ossify later.

Single palmar creases appear normally in approximately 1 out of 30 babies. It may be sometimes associated with Down syndrome, though in these cases other physical abnormalities would also be manifested. The finding of bilateral single palmar creases in an otherwise normal-appearing baby does not always require a chromosome analysis. Oedema of the feet and hands is a very rare clinical finding and prompts an investigation for Turner's syndrome. In a normal baby, occipital-frontal head circumference should be greater than chest circumference. In a normal term baby at birth the prepuce is not retractile, but 50% of boys have a retractile prepuce by the age of ten years and over 90% following puberty. The various reflexes, which may be present in a newborn baby at birth include:

❑ The stepping reflex
❑ The Moro or startle reflex
❑ Walking or stepping
❑ Tonic neck reflex
❑ The palmar and plantar grasp.

APGAR Score

The APGAR score was invented by Virginia Apgar and is described in **Table 13.23**. This scoring system is named after the acronym APGAR, which summarises the five criteria used in this system: **A**ppearance, **P**ulse, **G**rimace, **A**ctivity and **R**espiration. This is a simple scoring system, which describes the baby's health immediately after delivery. It is performed at 1, 5 and sometimes 10 minutes after delivery. Score between 7 and 10 indicates that the newborn is in good health. Score lower than 7 indicates that the baby requires some kind of medical attention. Resuscitation in some form is required in approximately one-third of babies.

Recognition of the need for resuscitation should be prompt (in the first minute) if there is no regular respiration, if the heart rate is below 100 beats/minute or if the APGAR score remains below 7. Ventilatory rates should be normally set at 20–30 per minute.

TABLE 13.23	APGAR score		
Feature	**Score = 0**	**Score = 1**	**Score = 2**
Appearance	White	Blue	Pink
Pulse	0	<100	100 or more
Grimace/reflex irritability	None	Grimace	Cry
Activity/Muscle tone	Limp	Some flexion of the extremities	Active movement/good flexion
Respiration	Absent	Gasping/weak cry	Regular/good cry

Intubation should be performed if respiration is absent or is poor and does not improve with stimulation by two minutes or the heart rate is less than 80 beats/minute. Meconium seen in the posterior pharynx and larynx is another indication for intubation. Gentle suction should be applied to the endotracheal tube until no further meconium is obtained. The tube must be changed in case it becomes blocked during the process. Naloxone cannot be given safely to all infants. Care should be taken in the infant of a mother who is a known opiate addict. Use of naloxone in these cases may precipitate acute withdrawal in the neonate.

There has been a lot of interest in the possible link between the APGAR score and the risk of cerebral palsy or other neurological damage. It used to be thought that cerebral palsy was principally due to damage sustained during delivery. Nowadays, things are more complex and cerebral palsy is thought to be the result of developmental problems in early pregnancy.

Congenital Anomalies

Cleft Lip and Palate

Most cases of cleft lip and palate are due to multifactorial inheritance. They have been described in association with more than 100 rare genetic defects and are a feature of trisomy 13. The risk of recurrence varies with the severity of the problem. It is about 1:20 in the child with bilateral facial and palate clefts and 1:50 for the isolated facial cleft. The incidence of clefts is increased by a factor of ten for the babies of women taking anti-epileptic drugs. The results of surgery in these cases are quite good. The case must be managed by a multidisciplinary team comprising of specialised paediatricians, surgeons, orthodontists, etc. Cleft lips are usually repaired in the early months of life, with the palate being dealt with at about a year.

Exomphalos

Exomphalos is an uncommon condition, occurring in approximately 1:3,000 pregnancies. If it occurs as an isolated defect in the baby then it is treatable and more than 90% of the babies will survive and do well. Exomphalos can occur in association with chromosomal abnormalities, usually T13 or T18. If exomphalos is identified then amniocentesis should be offered as the risk of T18 is approximately 22% and that of T13 is approximately 5%. It is important to appreciate that the counselling of the mother is different in this situation in comparison to the counselling in cases where there is a risk of T21. T18 and T13 are lethal abnormalities; so, the mother does not face the prospect of having to care for a handicapped child for life.

Congenital Hip Dislocation

Congenital hip dislocation (CHD) is a condition with multi-factorial inheritance and is characterised by limitation of abduction and an evidence of easy dislocation. Its incidence is cited as 1 per 500 births and is higher in breech presentation by a factor of 10 in comparison with vertex presentations. The incidence is higher amongst ethnic groups (e.g. North Americans) who swaddle their infants with the hips extended. On the other hand, its incidence is lower in those ethnic groups (e.g. Africans) who cart the baby on the mother's hip or back, so that the baby's hips are abducted. Girls are six times more likely to be affected than boys. Ortolani's test is used for screening.

The prognosis is best if the condition is diagnosed early, but clinical screening is not 100% effective in detecting all cases in the neonatal period. X-rays examination does not prove to be useful in cases less than 3 years of age. Ultrasound examination helps in defining the shape of the acetabulum and overall hip anatomy. Some clinicians have advocated its routine use for all babies, but this has not been generally adopted.

Congenital Heart Defects

Congenital heart anomalies can be considered as the commonest congenital abnormalities. The incidence is about 8 per 1,000 births. One affected sibling is associated with a 2% risk, two affected siblings with a 10% risk and an affected mother with about a 10% risk. As these are the most common birth defects, hospitals are increasingly incorporating a cardiac scan into the routine anomaly scan at 18–20 weeks. However, a detailed cardiac scan for the "at-risk" patient is best done at 20+ weeks, often as late as 24 weeks. If the defect is serious enough to cause cardiac failure, the baby will develop hydrops. By definition, this will be "non-immune".

Jaundice in the Newborn

Neonatal jaundice (NJ) is common though usually transient condition. It is of no great significance to the baby's welfare and development. However, it can sometimes result in the development of kernicterus, with devastating, life-long brain damage. Jaundice can develop in about 60% of babies at term and about 80% of premature babies.

Jaundice with early onset usually implies haemolysis in the neonate. Jaundice occurring after the first couple of days is most often physiological. The breakdown of haemoglobin releases bilirubin. It is conjugated by the hepatic enzyme glucuronyl transferase. The more immature the baby, the less able is its liver to capture and process bilirubin. This results in high levels of unconjugated bilirubin. Sick and dehydrated babies are also not able to deal well with the processing of bilirubin. Neonatal infection further increases the risk of NJ.

In its free form, unconjugated bilirubin may be relatively insoluble in water, but it is fat-soluble. It can cross lipid-containing membranes such as the blood-brain barrier and be deposited in the basal ganglia of the brain, resulting in kernicterus. Kernicterus is characterised by choreoathetoid

movements and cerebral palsy. It can also be deposited in the hippocampus and cranial nerve nuclei, particularly the cochlear and oculomotor nuclei. Hearing loss, most often high-frequency, is very common. Abnormal eye movements also occur. Mental retardation may be a feature, but is not the dominant one.

Bilirubin is not synthesised from cholesterol (like the steroids) but is rather a breakdown product of haem metabolism. It is toxic to the tissues and therefore it is transported in the blood bound to albumin. Bilirubin level greater than 340 mmol/L is a cause for concern in a mature baby. Lower levels can be associated with kernicterus in premature and sick babies. Bilirubin levels less than 340 mmol/L are unlikely to cause kernicterus in a mature baby. Some causes for NJ are described below:

❑ *Causes related to increased haemolysis*: Spherocytosis and elliptocytosis, in which RBCs may be easily haemolysed.
❑ *An excessive load of blood*: Excessive load of blood may result in formation of excessive amounts of bilirubin, which the neonate may be unable to deal with. This may occur in cases of cephalo-haematoma (which is more common with the ventouse), bruising from traumatic delivery or in haemorrhagic disease of the newborn, swallowed maternal blood, etc.
❑ *Drugs*: A number of drugs such as sulphonamides, diazepam, etc. increase the risk for NJ. Sulphonamides compete with bilirubin for binding sites on albumin.

Levels of conjugated bilirubin >25 mmol/L point to major hepatic abnormality, especially biliary atresia. Jaundice not clearing within a week could point towards serious problems such as biliary atresia and hypothyroidism. Biliary atresia must be operated upon in the first 6 weeks via Kasai procedure. Else, the surgery may prove ineffective and the child may require a liver transplant.

Sickle cell disease will cause anaemia as the child replaces HbF with HbS and sickling crisis begins to occur.

However, the amount of haemolysis in a neonate would not be more than normal.

Management

The clinician must look for a cause (e.g. urinary tract infection) and initiate appropriate treatment. General measures include maintenance of adequate hydration and adequate food intake.

Phototherapy: This is the most commonly used form of therapy in these cases. The light used is often referred to as the ultraviolet light, but belongs to the blue spectrum of normal light. It converts bilirubin into biliverdin, a harmless, soluble metabolite. Phototherapy can overheat the baby. On the other hand, a naked baby is at risk of excessive cooling, so its temperature needs to be monitored at the time of phototherapy. The eyes must be covered due to an increased risk of retinal damage.

Exchange transfusion: In severe cases, exchange transfusion may be required. Rapidly rising bilirubin levels (>8.5 mmol/L per hour) are also a cause for concern. Additionally, any baby showing clinical features suggestive of kernicterus should also have aggressive treatment. The excessive bilirubin may be accompanied by significant anaemia, particularly if there is a pathological cause for the haemolysis. This, too, could be another cause requiring exchange transfusion. In this procedure, twice the baby's blood volume is exchanged (80 mL/kg in the mature baby).

The exchange can be done by taking off 10–15 mL of baby's blood and replacing it with 10–15 mL of donor blood. The process must be repeated until the required volume has been exchanged. The donor blood is usually Rh Negative, blood group O, and the blood is irradiated to prevent graft-host reactions. IV immunoglobulin may also be administered to reduce the requirement for exchange transfusion.

Choose the Single Best Answer (SBA)

Q 1. Which of the following is not correct regarding the detection of β-hCG levels during early pregnancy?
  A. levels which rise less than 50% in 48 hours at 6 weeks may indicate ectopic pregnancy
  B. levels > 8,000 i.u./l with no scan evidence of an intrauterine pregnancy strongly suggest ectopic pregnancy
  C. levels < 1,000 i.u./l at 8 weeks suggest ectopic pregnancy or pregnancy failure
  D. levels are above normal in hydatidiform mole
  E. None of the above

Q 2. Which of the following statement is not correct regarding miscarriage?
  A. If recurrent, it can be associated with parental chromosomal translocation.
  B. If recurrent, it can be associated with sickle cell trait.
  C. The clinical diagnosis of incomplete miscarriage is assisted by digital examination of the cervical canal.
  D. Cervical incompetence causes recurrent second-trimester miscarriage.
  E. Missed miscarriage should be suspected if the uterine size is smaller than that expected for gestational age.

1. D   **2. B**

Q 3. Which of the following statement is correct regarding the early pregnancy loss?

A. Is usually due to hormone deficiency.

B. Is due to an abnormal karyotype in approximately 5% of cases.

C. Intrauterine pregnancy cannot co-exist with tubal pregnancy.

D. An intrauterine pregnancy should be visible on transvaginal scan if the HCG is > 1,000 i.u./l.

E. Always needs evacuation.

Q 4. Which of the following is true regarding threatened miscarriage at eight weeks of gestation?

A. The prognosis can be determined in most cases by ultrasound.

B. Can be effectively treated with depot progestogens

C. Always needs evacuation if non-viable

D. Anti-D immunoglobulin should be administered if the mother is Rhesus negative.

E. May co-exist with ectopic pregnancy

Q 5. Which of the following is true regarding haemoglobin values of less 10g/dL during pregnancy?

A. Is a recognised side-effect of anti-convulsant therapy

B. Is associated with urinary tract infection

C. Is a complication of multiple pregnancy

D. Increases the risk of post-partum haemorrhage

E. All the above

Q 6. Which of the following is not true concerning thalassaemia in pregnancy?

A. Thalassaemia minor may be suspected on a blood film.

B. Thalassaemia is particularly concentrated in a broad band encompassing the Mediterranean and Middle East.

C. The carrier rate in the UK is approximately 1 in 10,000.

D. A woman with α-thalassaemia minor can be reassured that the baby will be healthy.

E. Presence of thalassaemia trait has no association with the occurrence of preeclampsia.

Q 7. Which of the following is true concerning sickle cell disorders in pregnancy?

A. Sickle cell disorders are most common in women of Asian origin.

B. A sickle cell crisis can be precipitated in conditions of heightened oxygen tension.

C. Sickle cell disorders are associated with an increased incidence of hypertension during pregnancy.

D. Sickle cell disease results from a variant on the alpha globin chain.

E. Partner screening is recommended during the second trimester

Q 8. Which of the following is not true regarding intrauterine growth restriction?

A. It is associated with premature labour

B. It may be associated with a low socio-economic status

C. These babies are at an increased risk of developing respiratory distress syndrome

D. It may be associated with raised serum AFP at 16 weeks followed by normal scan at18 weeks

E. There may be neonatal hypoglycaemia in the babies born with IUGR

Q 9. Which of the following is not correct concerning maternal cardiac disease in pregnancy?

A. A classification system exists to determine the mortality risk.

B. Involvement of the aorta in Marfan's syndrome increases the mortality.

C. The foetus has an increased risk of congenital heart disease.

D. Mitral stenosis is an infrequent complication following rheumatic heart disease.

E. Women with primary pulmonary hypertension should be advised against pregnancy.

Q 10. Which of the following is true regarding thyrotoxicosis in pregnancy?

A. It is usually due to a solitary adenoma

B. The major maternal risk is congestive cardiac failure

C. Beta-blocking drugs are contra-indicated

D. May be treated with radioactive iodine as the drug does not cross the placenta

E. Usually occurs as new disease as a result of HCG stimulation of the thyroid

Q 11. Which of the following is not true regarding thyroid disease in pregnancy?

A. Hyperthyroidism may be associated with IUGR

B. Thiouracil may cause severe liver disease

C. Congenital hypothyroidism is not routinely screened for in the UK

D. Thyrotoxicosis is associated with increased hepatic metabolism of "the Pill"

E. raised T_3 and T_4 levels are found in hyperemesis and molar pregnancy

Q 12. Which of the following statement is correct regarding obstetric cholestasis (OC)?

A. OC usually presents with jaundice.

B. Ursodeoxycholic acid is recommended to reduce foetal risk.

C. Dexamethasone is recommended to reduce foetal and maternal risk.

D. The symptoms of OC can recur if an affected woman takes the pill

E. The risk of recurrence in a subsequent pregnancy is a rare event

3. D   4. E   5. E   6. D   7. C   8. A   9. D   10. B   11. C   12. D

Q 13. **Which of the following is true regarding the monozygotic twins?**

A. Are more common than dizygotic twins
B. Are commonly familial
C. May be reliably distinguished from dizygotic twins by the naked-eye examination of the foetal membranes and placentae
D. Have a higher incidence of placenta praevia than singleton pregnancies
E. Are associated with oligohydramnios.

Q 14. **Which of the following complication is associated with multiple births?**

A. Increased incidence of congenital abnormalities
B. Increased incidence of growth retardation
C. Increased incidence of postpartum haemorrhage
D. Increased incidence of preterm labour
E. All the above

Q 15. **Which of the following is true regarding hypertension in pregnancy?**

A. It is of little significance unless accompanied by proteinuria
B. It causes foetal growth restriction in more than half of affected women
C. It is not associated with an increased incidence of bleeding from placental praevia
D. It should be assessed by admission to hospital
E. It is a contraindication to the use of intramuscular ergometrine.

Q 16. **Which of the following statement is not correct regarding preeclampsia?**

A. The perinatal mortality is raised
B. Epigastric pain may indicate impending eclampsia
C. There are lowered serum urate levels
D. There may be no foetus
E. The liquor volume may be diminished.

Q 17. **Which of the following is required for the diagnosis of pre-eclampsia?**

A. High urate levels
B. Low plasma magnesium levels
C. More than 24 weeks gestation
D. Oedema
E. Proteinuria

Q 18. **Which of the following is not a side effect of alpha-methyldopa?**

A. Depression
B. Nasal congestion
C. Oedema
D. Pyrexia
E. Visual disturbances

Q 19. **Which of the following features is compatible with hypertension?**

A. A fourth heart sound
B. A soft aortic second heart sound
C. Retinal haemorrhages and soft exudates, indicating a grade 2 hypertensive retinopathy
D. A tapping apex beat
E. A third heart sound

Q 20. **Which of the following is not true concerning the blood pressure regulation?**

A. Adrenaline acts primarily upon the vasomotor centre
B. Angiotensinogen is inactive without modification
C. Bradykinin decreases blood pressure
D. Prostacyclin lowers blood pressure
E. Serotonin is vasodilatory

Q 21. **Which of the following is true regarding the non-invasive blood pressure monitoring (Dinamap)?**

A. Is accurate in patients in atrial fibrillation
B. Is more accurate than a mercury sphygmomanometer
C. Provides a more accurate systolic than diastolic reading
D. Relies on Doppler ultrasonic measurements
E. The device needs to be positioned at the same level as the patient's heart

Q 22. **Which of the following drugs are not accepted to be safe for treating pregnancy hypertension?**

A. Nifedipine
B. Hydralazine
C. Magnesium sulphate
D. Methyl dopa
E. Diuretics

Q 23. **Second trimester bleeding may be due to which of the following cause?**

A. Missed abortion
B. Premature labour
C. Erythroblastosis fetalis
D. Threatened abortion
E. Monilial infection

Q 24. **Which of the following is not true regarding placental abruption?**

A. May have no associated vaginal bleeding
B. Is an indication for delivery
C. Has a higher incidence with maternal cocaine abuse
D. May be identified using ultrasound to demonstrate retroplacental clot
E. The diagnosis of concealed abruption can be easily confused with that of acute appendicitis.

Q 25. **Which of the following is true regarding ABO incompatibility between mother and the foetus?**

A. May affect the first pregnancy.
B. Worsens with successive pregnancies
C. Usually causes significant anaemia of the foetus at birth.
D. Often requires exchange transfusion.
E. Is caused by the Rh(D) antigen.

13. D   14. E   15. E   16. C   17. E   18. E   19. A   20. A   21. C   22. E   23. D   24. B   25. A

Q 26. Which of the following is not true regarding ABO blood group incompatibility between mother and foetus?

- A. It is associated with a strongly positive direct Coombs' test
- B. Its severity does not vary between the first and subsequent pregnancies
- C. It is usually detected in the antenatal period
- D. Manifests itself on the first or second day of life
- E. Affects only the first pregnancy

Q 27. Which of the following is not true regarding pre-implantation genetic diagnosis?

- A. Is used for selection of embryo gender
- B. Is used to detect embryos homozygous for cystic fibrosis
- C. Is used to detect sex-linked disorders
- D. Is used to screen embryos produced from semen obtained by ICSI
- E. Is used to screen embryos when mothers above 35 years of age undertake IVF

Q 28. Which of the following is true regarding the Kleihauer test?

- A. It may be used to confirm the presence of Rhesus antibodies
- B. It should be performed routinely at 28 and 36 weeks in the woman who is rhesus negative
- C. It is no longer required after delivery in the Rhesus negative woman
- D. It is based on the relative resistance of foetal haemoglobin to denaturation using an acid solution
- E. It is no longer required after delivery in the Rhesus negative woman

Q 29. Which of the following is not true regarding pre-implantation genetic diagnosis?

- A. Disorders caused by a single gene defect can be detected
- B. It should be used to exclude Down syndrome in a couple undergoing IVF using a donor ovum from a 23-year-old, into a 46-year-old recipient
- C. Foetal sex can be determined
- D. HLA status can be determined
- E. It is usually not performed in natural conceptions

Q 30. Which of the following is not correct regarding a raised MSAFP level at 16 weeks of gestation?

- A. May be due to incorrect assessment of gestation.
- B. May be due to gastroschisis
- C. Is more likely with multiple pregnancy
- D. Is more likely if the foetus has T21 (trisomy 21)
- E. Is more likely after threatened miscarriage.

Q 31. Which of the following is true regarding a 37 year-old-woman at 16 weeks' gestation?

- A. Has a risk of Down syndrome of 1:100
- B. Should be advised to have screening for Down syndrome
- C. Should be advised to have amniocentesis, not the 'triple' test
- D. Is at increased risk of Edward's syndrome
- E. Is at increased risk of having a baby with a neural tube defect

Q 32. Which of the following statements concerning eclampsia is not correct?

- A. Disseminated intravascular coagulation is an associated hazard
- B. It is the commonest cause of maternal death in the United Kingdom
- C. It occurs following delivery in about 20–25% of cases
- D. Placental abruption is a recognised association
- E. The maternal mortality rate is highest when it occurs in the postpartum period

Q 33. Regarding 'HELLP' syndrome which of the following statement is not true?

- A. It is related to preeclampsia
- B. It may occur in non-pregnant patients
- C. Upper abdominal quadrant pain is a characteristic feature
- D. Patients may have low platelets
- E. The 'H' stands for haemolysis

Q 34. Which of the following is correct regarding hyperemesis gravidarum?

- A. Occurs in 5% of pregnancies
- B. Is associated with missed miscarriage
- C. Is associated with raised maternal alpha-feto protein levels
- D. Is associated with raised hCG levels
- E. Is commonly due to pyelonephritis

Q 35. Indicators of foetal distress include which of the following?

- A. Maternal acidosis
- B. Foetal heart rate of 100 beats per minute
- C. Absence of accelerations
- D. Maternal pyrexia
- E. Tangential foetal heart rate pattern

Q 36. Which of the following statement is correct regarding the foetal scalp pH?

- A. A result of 7.28 is normal
- B. Accurately determines the condition of the baby
- C. Carries no risk
- D. Cannot be performed in cases of breech presentation
- E. Increases the Caesarean section rate in patients having continuous electronic foetal heart rate monitoring

26. E   27. A   28. D   29. B   30. D   31. D   32. B   33. B   34. D   35. C   36. A

Q 37. **Which of the following statement regarding meconium is correct?**

A. Normal meconium is dark green and malodorous
B. It is normally passed by the neonate within the first 24 hours
C. Meconium is completely normal in babies with cystic fibrosis
D. It is most commonly found in liquor in the premature baby
E. Aspiration of meconium is harmless

Q 38. **Which of the following is true regarding epidural anaesthesia in labour?**

A. May cause hypertension
B. Is contra-indicated in the patient who has had LSCS
C. Is contra-indicated in twin pregnancy
D. Can be administered in patients with coagulopathy
E. Administration requires written consent

Q 39. **Which of the following is not true regarding the epidural space?**

A. Commences at the foramen magnum
B. Contains Batson's plexus
C. Ends at the level of S2 in the adult
D. Negative pressure within the space is greatest in the lumbar and sacral regions
E. Is triangular in cross-section

Q 40. **Which of the following is not true concerning pethidine analgesia in labour?**

A. Analgesic effect takes 10–15 minutes to become apparent
B. Causes an elevation of the APGAR scores
C. Causes loss of foetal cardiac beat-to-beat variability
D. Is 50% protein bound
E. Patient-controlled analgesia provides better pain relief than nurse-controlled analgesia

Q 41. **The bladder is at risk of damage at the time of which of the following surgery?**

A. Repair of enterocoele
B. Classical caesarean section
C. Lower segment caesarean section
D. Uterine myomectomy
E. Laparoscopy

Q 42. **Which of the following is true regarding classical caesarean section?**

A. Is Caesarean section performed through a mid-line abdominal incision
B. Is never done nowadays
C. Carries an increased risk of dehiscence of the abdominal wound
D. Is a ground for elective repeat caesarean section
E. Is a ground for emergency tubal ligation at the time of the operation

Q 43. **Which of the following is not true regarding urinary tract infection in pregnancy?**

A. It is associated with preterm labour
B. It is commonly due to staphylococci
C. Acute pyelonephritis is associated with intrauterine growth retardation
D. There is no known, statistically proven association with foetal lie
E. It may present with vomiting.

Q 44. **Which of the following is a recognised complication of external cephalic version?**

A. Positive Kleihauer test
B. Fetal bradycardia
C. Transient maternal hypertension
D. Premature rupture of the membranes
E. All the above

Q 45. **Which of the following is not correct regarding rotational delivery?**

A. May be preceded by a labour during which back pain is a prominent feature
B. Can be achieved using a silastic ventouse cup
C. Can be safely attempted when none of the foetal head is palpable per abdomen
D. Can correct a deep transverse arrest
E. Should be attempted with a foetal pH of 7.12.

Q 46. **Which of the following is not true regarding the ventouse method of delivery?**

A. May employ a metal cup
B. Has increased in popularity with electronic pumps
C. Can be used safely in the absence of criteria necessary for a forceps delivery
D. Requires the patient to be in the lithotomy position
E. May be performed in conjunction with a pudendal block.

Q 47. **Which of the following is true regarding the management of a pulmonary embolus?**

A. An inferior vena cava filter (for example, Greenfield filter) may be required
B. Anticoagulation should initially be with warfarin
C. Low molecular weight heparins should be monitored using the activated partial thromboplastin time (APTT)
D. The aim of warfarin therapy is an International Normalised Ratio (INR) of 3 to 4
E. Presence of a subarachnoid haemorrhage is not a contra-indication to anticoagulation

Q 48. **Which of the following may be associated with pulmonary embolism?**

A. Femoral fracture
B. Obese BMI greater than 35
C. Pelvic surgery
D. Pulmonary infarct
E. All the above

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37. B   38. E   39. D   40. B   41. C   42. D   43. B   44. E   45. E   46. C   47. A   48. E

Q 49. Which of the following is true regarding thrombo-embolic disease?

A. The risk in a multiple gestation is similar to that in a singleton pregnancy.
B. Heparin should not be given in the first trimester.
C. Warfarin decreases the likelihood of foetal haemorrhage.
D. In pregnancy heparin therapy can cause maternal osteoporosis.
E. The risk is increased in women who are blood group O.

Q 50. Which of the following steps must be taken at the time of massive post-partum haemorrhage?

A. An anaesthetist is essential to assist in the management of the patient
B. Initial cross-matching of three units of blood is sufficient
C. Bimanual uterine compression has no role in these cases
D. Uncross-matched O rhesus-positive blood may be given in an emergency
E. None of the above

Q 51. Which of the following is true regarding secondary post-partum haemorrhage?

A. It is abnormal bleeding that occurs 12 hours post-partum
B. It may be due to infection
C. It cannot be controlled by uterine contracting agents
D. Occurs following 5% of births
E. Can usually be diagnosed by ultrasound examination of the pelvic organs.

Q 52. Which of the following is true regarding breastfeeding?

A. Human milk contains more protein than cow milk.
B. Human milk has a higher energy content and more fat than cow milk
C. Breast feeding does not protect against infection in the baby.
D. Breast milk is of uniform composition throughout a feed.
E. Lactation is effectively stimulated by bromocriptine.

Q 53. Which of the following is true regarding the termination of pregnancy?

A. Is illegal after 20 weeks' gestation
B. After 16 weeks is most safely achieved by hysterotomy
C. Requires the signature of two gynaecologists
D. Can be achieved by the intra-muscular administration of prostaglandins.
E. Complications include infertility in about 2% of cases.

Q 54. Which of the following factor does not predispose to ectopic pregnancy?

A. Endometriosis
B. Intrauterine contraceptive device
C. Ovarian fibroids
D. Pelvic inflammatory disease
E. Progesterone only contraceptive pill

Q 55. Which of the following is true regarding gestational trophoblastic disease (GTD)?

A. GTD is associated with 47XXY
B. Follow up is by serial assay of AFP
C. Secondary spread is particularly via the uterine lymphatics
D. The chromosome content is mainly paternal
E. Choriocarcinoma follows in 10% of cases of molar pregnancy

Q 56. Which of the following statement regarding neonatal intraventricular haemorrhage is not correct?

A. Is associated with prematurity
B. Occurs in about 50% of very low birth weight babies
C. Usually occurs in the early days of life
D. Is diagnosed by lumbar puncture
E. May lead to hydrocephalus

Q 57. Hydramnios is not associated with which of the following conditions?

A. Twin-twin transfusion syndrome
B. Diabetes
C. Potter's syndrome
D. Hydrops fetalis
E. Oesophageal atresia

Q 58. Which of the following is true regarding the raised alphafetoprotein (AFP) levels at 16 weeks gestation?

A. Is an indication for amniocentesis
B. Is caused by gastroschisis
C. Accurate dating of pregnancy is not required for its assessment
D. May be due to Down syndrome
E. Should be confirmed by a repeat blood test

Q 59. Which of the following is not true regarding the routine ultrasound scan at 18– 20^{+6} weeks?

A. The National Screening Committee has recommended 6 basic measurements that should be taken
B. The National Screening Committee has identified 11 foetal anomalies to be looked for
C. Screening for placenta praevia is at the top of the list
D. Choroid plexus cysts no longer need any response
E. Presence of echogenic bowel requires a response

49. D   50. A   51. B   52. B   53. C   54. C   55. D   56. D   57. C   58. B   59. C

Q 60. Presence of echogenic bowel is not associated with which of the following?
A. Down syndrome
B. Turner's syndrome
C. Cystic fibrosis
D. Bleeding in pregnancy
E. Viral infection

Q 61. Which of the following malignancy would be expected to be associated with an elevated alpha fetoprotein (AFP) concentration of 300 mU/L?
A. Carcinoid syndrome
B. Cirrhosis of the liver
C. Colonic carcinoma with hepatic metastases
D. Hepatoma
E. All the above

Q 62. Epidural bupivacaine administered during labour may cause which of the following?
A. An increased rate of caesarean delivery
B. Decreased uterine contractility
C. Pruritus
D. Tinnitus
E. Total spinal block

Q 63. Which of the following is true concerning Entonox for labour analgesia?
A. Has been used since the 1990s
B. Is less effective than pethidine
C. It should be inhaled as the pains start
D. Low dose sevoflurane may be used to augment its analgesic effect
E. Should not be used with other forms of analgesia

Q 64. Which of the following statement regarding induction of labour is correct?
A. Can be achieved by amniotomy
B. Is easiest when the cervix is in a posterior position
C. Could be achieved by an ergometrine infusion
D. Is indicated with an uncomplicated dichorionic twin pregnancy of greater than 36 weeks' gestation
E. Cannot be achieved by intravenous prostaglandin infusion.

Q 65. Which of the following statement regarding labour is not correct?
A. A foetal heart rate of 140 per minute is normal
B. An acceleration in foetal heart rate after a uterine contraction is normal
C. Braxton-Hicks contractions signify the onset of the first stage of labour
D. Syntocinon may be given during the first and third stages of labour
E. The placenta is delivered during the third stage

Q 66. Which of the following is true regarding fistulae due to obstructed labour?
A. Are best repaired immediately after delivery
B. Are commonly uretero-vaginal
C. Are repaired by a sling operation
D. Cause continuous urinary incontinence
E. Are usually repaired by an abdominal approach

Q 67. Foetal well-being in the third trimester can be usefully assessed by which of the following parameters?
A. Serial assessment of symphyseal fundal height
B. Ultrasound measurement of crown–rump length
C. Measurement of serum alpha-fetoprotein levels
D. Measurement of serum oestradiol levels.
E. None of the above

Q 68. Which of the following is not true regarding a high foetal head at term in a primipara?
A. Can be caused by placenta praevia
B. Can be caused by a lower-segment uterine fibroid
C. Is associated with incorrect pregnancy dating
D. Is an indication for a caesarean section
E. Has a higher incidence in patients of African origin.

Q 69. Which of the following statement is not correct regarding the first stage of labour?
A. The latent phase may last for more than four hours
B. The active phase should be associated with cervical dilatation at a rate of at least 1 cm. per hour
C. The active phase starts when the cervix is effaced and 3 cm dilated
D. Is best charted using a partogram
E. Epidural anaesthesia has an adverse effect on the rate of progress in the first stage of labour

Q 70. Which of the following statement is not correct regarding external cephalic version?
A. Anti-D should be given to rhesus negative mothers.
B. Is best done at 32 weeks
C. Reduces the incidence of breech presentation at term
D. Is more successful if a beta-sympathomimetic is used
E. May cause foetal bradycardia

Q 71. Which of the following is a recognised complication of a lower segment caesarean section performed under regional anaesthesia?
A. Aspiration of gastric contents
B. Delayed respiratory depression with hydrophilic opioids
C. Evidence of myocardial ischaemia on the electrocardiograph
D. Venous air embolism
E. All the above

60. B   61. D   62. D   63. D   64. A   65. C   66. D   67. A   68. D   69. E   70. B   71. E

Q 72. **Which of the following regarding deep vein thrombosis is not correct?**

A. Caesarean section is particularly associated with DVT and pulmonary embolism
B. Is more likely in association with pregnancy
C. Is more common in women with the factor V Leiden mutation
D. Is best diagnosed by Homan's sign
E. Is more common on the left than the right

Q 73. **Which of the following is not true regarding a newborn term infant?**

A. Anaemia may be caused by a cephalhaematoma
B. Ballotable kidneys may not always be abnormal
C. Oedema of the feet and hands suggest Turner's syndrome
D. The Apgar score at 5 minutes is more predictive of later neurodevelopment prognosis than Apgar score at 1 minute
E. The findings of bilateral single palmar creases in an otherwise normal-appearing baby demands a chromosome analysis

Q 74. **In the normal neonate, which of the following is correct?**

A. Nucleated red cells are rarely seen in the peripheral blood
B. The mean cell volume is greater than 100 fl
C. Haemoglobin A is the predominant haemoglobin
D. The platelet count is more than the normal adult range
E. The white cell count may be less than 25 x 10^9/l

Q 75. **Which of the following is true regarding a baby with birthweight > 4.5 kg?**

A. Is almost always due to poorly controlled diabetes.
B. Is not associated with an increased risk of shoulder dystocia.
C. Is a contra-indication to vaginal delivery of a baby presenting by the breech.
D. Another large baby is unlikely in a subsequent pregnancy.
E. Is normally due to post-maturity.

Q 76. **Which of the following statement regarding exomphalos is correct?**

A. Occurs in 1 out of 150 pregnancies
B. Is usually fatal
C. Is a marker for Down syndrome
D. Is a marker for trisomy 18
E. Is associated with renal agenesis

Q 77. **Which of the following statement is correct regarding haemolytic disease of the newborn?**

A. Affects mainly babies of Rh-positive mothers.
B. Occurs mainly in babies who lack D agglutinogen.
C. Causes jaundice which clears rapidly after birth.
D. Can be treated by transfusing the affected baby with Rh-positive blood.
E. Can be prevented by injecting the mother with anti-D agglutinins just after delivery.

Q 78. **Which of the following is not a feature of cleft lip and palate?**

A. Occur in approximately 1 per 1,000 births
B. May be associated with genetic or chromosomal abnormalities
C. Is a feature of trisomy 18
D. the risk of recurrence is about 1:50
E. is increased in the offspring of women taking drugs for epilepsy

Q 79. **Which of the following does not occur in a neonate?**

A. The bowel is sterile at birth.
B. The respiratory rate is in the region of 25–35 per minute.
C. The ductus arteriosus closes functionally within an hour of birth.
D. Compared with the adult, the neonate has higher blood levels of vitamin K.
E. Compared with the adult, the neonate has reduced blood levels of clotting factor X.

Q 80. **Which of the following is true about bilirubin?**

A. Conjugates iron
B. Reduces absorption of fat from the gut
C. Is a steroid
D. Is bound to albumin in the circulation
E. Is conjugated to glycerine

Q 81. **Which of the following does not occur in uncomplicated haemolytic jaundice?**

A. High levels of urobilinogen
B. High levels of unconjugated serum bilirubin
C. High serum alkaline phosphatase
D. Reticulocytosis
E. Urobilinuria

Q 82. **Which of the following is correct regarding the neonatal jaundice?**

A. Early onset usually indicates a haemolytic process
B. May be due to sickle cell disease
C. Is more common after forceps delivery compared with the ventouse
D. Mild cases may benefit from ultraviolet light
E. may be due to glucose 6 phosphatase deficiency

72. D   73. E   74. B   75. C   76. D   77. E   78. E   79. D   80. D   81. C   82. A

Gynaecology

Surgical Skills

Pre-operative Preparation

Pre-anaesthetic Medications

The drugs used for pre-anaesthetic medication prior to gynaecological surgery commonly include the following:

❑ An anxiolytic, e.g. benzodiazepines such as midazolam and lorazepam (to allay anxiety the night before surgery)

❑ *Antiemetic (to reduce nausea and vomiting)*: Ondansetron is an effective antiemetic agent. Metoclopramide is used preoperatively to stimulate gastrointestinal emptying. However, it is rather ineffective as a preoperative antiemetic in standard doses (10 mg) due to short duration of action

❑ H2 blockers (to reduce the pH and volume of gastric secretions)

❑ Atropine, an anticholinergic, is preferentially used to dry up secretions.

❑ Analgesic drugs such as non-steroidal anti-inflammatory drugs (NSAIDs) and opioids, etc. can also be used.

Preparation of the Skin

❑ The area around the proposed incision site must be washed with antiseptic soap solution (e.g. savlon and/or betadine solution). Antiseptic skin cleansing before surgery is thought to reduce the risk of post-operative wound infections. The antiseptic solution must be applied at least three times over the incision site, using a high-level disinfected sponge-holding forceps and cotton or gauze swab. The surgeon must begin at the proposed incision site and move outwards in a circular motion away from the incision site. After reaching the edge of the sterile field, the previous swab must be discarded and new swab must be used. At the end, the inner aspects of thighs and umbilicus must be swabbed. The surgeon must keep his/her arms and elbows high and surgical gown away from the surgical field. The woman must be draped using sterile drapes immediately after the area of surgery has been adequately prepared, in order to avoid contamination. If the drape has a window, it should be placed directly over the incision site

❑ Chlorhexidine is a commonly used skin cleanser and antiseptic. It can also be used for bladder irrigation and checking the catheter patency. Pure chlorhexidine is a clear solution, which has a pink dye added to enable the surgeon to see which areas of the skin have been painted. Chlorexidine is a commonly used antiseptic solution because of its rapid action, broad-spectrum activity and longer duration of action in comparison to the povidone-iodine solution. Ethyl alcohol with povidone-iodine is a skin disinfectant which must be used with caution in pregnancy, breast-feeding, broken skin, and renal impairment. Alcoholic based solutions give better disinfection when rubbed on until the skin is dry. Alcohol may, however, ignite if used in the presence of diathermy. Cetrimide is used when a detergent-effect is required. Glutaraldehyde is used in the treatment of warts and calluses.

Surgery in High-risk Patients

Patients who are HIV or hepatitis B positive are at high risk. Stating that a patient is high risk allows the surgeon to modify his surgical technique to avoid sharp injuries. High-risk patients do not pose a risk of infection to subsequent patients on the operating list. Therefore, placing them last in the operative list is discriminatory and a wrong practice.

In high-risk patients, the patient's skin is cleaned in the normal way. Eye protection must always be worn when operating on a high-risk patient. Double pair of gloves are usually worn by the surgeon while operating such patients. Wearing two pairs of the surgeon's usual size gloves may constrict the blood flow to the fingers and reduce dexterity. Therefore, surgeons usually prefer to wear larger sized glove on the outside.

Cardiac Disorders in Surgical Patients

The mortality for peri-operative myocardial infarction is approximately 40%. Therefore, elective surgery is undertaken after 6 months and urgent surgery at 4–6 weeks following myocardial infarction, with invasive monitoring and a pre-operative stress test. The greatest risk is to those undergoing thoracic, abdominal or vascular surgery (especially aortic surgery).

Myocardial infarction, and more commonly, fluid overload may precipitate heart failure in the surgical patient. The risk of peri-operative heart failure is approximately 17% if there is a pre-operative cardiovascular history.

Heart failure may be classified into four groups according to the New York Heart Association (NYHA) classification system **(Table 14.1)**, to allow prediction of operative mortality **(Table 14.2)**:

TABLE 14.1	New York Heart Association functional classification of heart failure
Class I	Patients with cardiac disease, but without resulting limitation of physical activity. Ordinary physical activity does not cause fatigue, palpitations, dyspnoea or anginal pain.
Class II	Patients with cardiac disease resulting in slight limitation of physical activity. They are comfortable at rest. Ordinary physical activity results in fatigue, palpitations, dyspnoea or anginal pain.
Class III	Patients with cardiac disease resulting in marked limitation of physical activity. They are comfortable at rest. Less than ordinary physical activity results in fatigue, palpitations, dyspnoea or anginal pain.
Class IV	Patients with cardiac disease resulting in an inability to carry on any physical activity without discomfort. Symptoms of cardiac insufficiency may even be present at rest. If any physical activity is undertaken, discomfort is increased.

TABLE 14.2	Prediction of mortality rates based on the patient's NYHA class	
NYHA class	**Mortality rate**	
Class I	4%	
Class II	11%	
Class III	27%	
Class IV	67%	

Abbreviation: NYHA, New York Heart Association

Approximately 30% of the patients who have already had a myocardial infarct may develop a re-infarction if operated within 3 months. Creatine kinase is a MB isoenzyme that peaks at 24–48 hours following myocardial infarction but currently troponins are used as the specific markers of myocardial injury.

Dehydration

Dehydration is a common finding in both pre-operative and post-operative patient and requires correction before undertaking surgery. Excessive loss of body fluids during surgery may result in the development of dehydration that causes dryness of mucous membranes and loss of skin turgor. This requires correction in the immediate post-operative period. In cases of dehydration, the patient may well display features of shock with hypotension, tachycardia and hypothermia. Mucosal membranes would appear dry. There may be high serum sodium levels, with decreased urine output and increased urine sodium and osmolality.

Post-operative Changes

Immediate Effects of Surgery in Normal Person

Hypertension 1 Hour Following Laparotomy

Hypertension can be frequently encountered amongst the operated patient in the immediate post-operative period. This could be related to the following causes:

- ❑ *Inadequate analgesic effect*: The commonest cause of post-operative hypertension is the inadequately controlled pain.
- ❑ *Hypercapnia*: Use of general anaesthetic or opioids for pain relief may result in respiratory depression. Hypercapnia due to alveolar hypoventilation can cause hypertension.
- ❑ *Urinary retention*: A full bladder due to urinary retention or a blocked catheter can also cause agitation and pain.
- ❑ *Diffusion hypoxia*: Hypertension can accompany diffusion hypoxia or any cause of hypoxia. Diffusion hypoxia is a feature of an anaesthetic involving nitrous oxide. To avoid diffusion hypoxia, supplementary oxygen should be administered to patients for about 20 minutes in the recovery period as the residual nitrous oxide diffuses into the alveoli.

Malignant Hyperpyrexia

Malignant hyperpyrexia is an inherited disease showing autosomal dominant mode of inheritance. It is a life-threatening condition that usually presents early during an anaesthetic exposure but sometimes can be delayed for several hours. It can affect the contraction of skeletal muscles and metabolism. It follows exposure to triggering agents, particularly volatile anaesthetic agents and suxamethonium. It is associated with features such

as cardiovascular instability, cyanosis, hypercapnia, hyperkalaemia and hyperthermia.

Metabolic Changes

The following metabolic changes can occur in response to surgery in a normal person:
❑ The stress response and increased cortisol levels result in a relative hyperglycaemia, along with tachycardia
❑ Fluid retention and potassium loss
❑ Rise in the metabolic rate.

Post-operative Wound Infection

In the majority of cases, the cause of pelvic and abdominal wound infection is the bacteria found amongst the endogenous microflora of the lower genital tract. Risk factors which predispose for development of wound infection are as follows:
❑ *Poor haemostasis*: Poor haemostasis results in haematoma, which can readily become infected, and cause a wound breakdown.
❑ *Diabetes*: Patients with diabetes are more likely to develop a wound infection due to impaired white cell function.
❑ *Rough handling of tissues*: Rough handling of tissue is more likely to result in tissue necrosis predisposing to wound infection.
❑ *Tissue hypoxia*: Tissue hypoxia increases the risk of poor wound healing and subsequent infection.

Incorporation of synthetic material does not increase the risk of wound infection but if they become infected, they may form a chronically discharging wound.

Steps, which can be taken to reduce the incidence of post-operative infection, are as follows:
❑ Prophylactic antibiotics 30 minutes prior to the incision or at the time of induction of anaesthesia
❑ Use of meticulous surgical techniques at the time of surgery
❑ Ensuring adequate haemostasis at the time of surgery. Drains must not be used as an alternative against good haemostasis.
❑ Judicious use of cautery at the time of surgery
❑ Gentle handling of the tissues
❑ Sterilisation of the operation theatre and high-level disinfection of the instruments.

Surgical Wound Infections

Surgical wounds differ from non-surgical wounds in that they always have a source of infection, which can be drained surgically. Tissue necrosis results from trauma or through a pathophysiological process. Inflammation leads to the events visible at the surface. Once the source of infection has been drained, antibiotics are usually unnecessary unless the surrounding tissues are infected. It is important to have a good idea of what organism is likely to be responsible for a particular infection so that one can tailor the antibiotic therapy accordingly. For example, chronic osteomyelitis may be caused by many organisms such as *Salmonella* spp. and *Mycobacterium,* but *Staphylococcus aureus* is the commonest organism responsible for this infection.

Staphylococci, which are aerobic, facultative anaerobic, Gram-positive cocci, are the commonest organisms to infect the surgical wound, because they are common skin commensals. Methicillin-resistant *Staphylococcus aureus* (MRSA) wound infection is hospital-acquired and the risk of acquisition can be minimised by observing basic precautions such as handwashing before wound inspection. Surgical wounds become infected with opportunistic organisms because of the relatively immunocompromised state of the post-operative patient resulting in reduced inhibition of microbial growth.

Necrotising fasciitis is a deep-seated aggressive infection of subcutaneous tissue and skin. It is commonly caused by group A *Streptococci* and there is no evidence to suggest it is commoner in carriers of MRSA.

Intra-abdominal Pus

The commonest aerobic bacteria isolated from the intra-abdominal pus in descending order are as follows:
❑ *Escherichia coli*
❑ *Enterococci*
❑ *Proteus*
❑ *Klebsiella* spp.

The commonest anaerobic bacteria in descending order are:
❑ *Bacteroides*
❑ *Clostridia*
❑ *Peptostreptococci.*

Actinomyces and *Bacillus* anaerobes are commonly found in breast abscesses.

Defective Wound Healing

Defective wound healing can result in the following:
❑ *Superficial wound disruption*: This usually results from a superficial wound infection.
❑ *Incisional hernias*: Hernias result when the deeper tissue layers (especially the rectus muscle) give way but the skin remains intact.
❑ *Cicatrisation*: This is associated with extensive scar formation and occurs when wounds have broken down or left to close by secondary intention.
❑ *Wound dehiscence*: Dehiscence of the wound occurs when both the deep layers and the skin breaks down.

Hypertrophic scarring and keloid formation are not the results of defective wound healing. Hypertrophic scarring occurs in individuals predisposed to the condition.

Keloid Scars

Keloid scars are characterised by smooth hard nodules caused by excessive collagen production. Keloid scarring is much commoner in people of Afro-Caribbean origin. They also tend to affect young adults a lot more. Keloid scarring may occur spontaneously but are associated with skin trauma, infection and surgery. There is no evidence to suggest that keloid scarring is associated with steroid therapy. However, if keloid scarring is treated with surgical removal then it must be followed by steroid injection or superficial radiotherapy or it may make the problem worse. There is no evidence to suggest that keloid scarring is associated with wound healing by secondary intention. However, there is some evidence to suggest that primary wound closure is a risk factor.

Local anaesthetics are not associated with keloid scarring. They can be used in surgical removal of the scar. Other methods of treatment include triamcinolone injection and compression with silica gels.

Metabolic Responses to Trauma

There occur increased levels of the following: adrenocorticotropic hormone, anti-diuretic hormone (ADH), glucagon and growth hormone. Increased production of growth hormone and glucagon contributes to the hyperglycaemia seen following surgery and trauma. Hypovolaemia results in increased ADH production and an increase in urine osmolality.

The metabolic response to trauma is mediated by both endocrine and paracrine factors. In the neurohumoral response, the pituitary-adrenal axis and the sympathetic nervous system are important. Production of locally released cytokines (for example, tumour necrosis factor-alpha, interleukins, etc.) is important for modulating the response.

Immediately after a large haemorrhage, there is a precipitant fall in pulse pressure with narrowing of blood vessels. Increased secretion of ADH results in thirst. Thirst also occurs due to the stimulation of the thirst receptors. Anaerobic glycolysis occurs due to reduced perfusion with increased predisposition to lactic acidosis. Coronary vasodilatation would be expected and chemoreceptors would therefore be stimulated.

Post-operative Nutrition

Total Parenteral Nutrition

The side effects of total parenteral nutrition (TPN) are numerous and include catheter-related sepsis and metabolic abnormalities resulting from the administered nutrients. It is often given centrally to reduce the risk of thrombophlebitis. TPN is hyperosmolar. It typically contains about 250 g glucose and 14–16 g of L-amino acids.

TPN is associated with metabolic disturbances in about 5% patients. Some of the commonly occurring metabolic abnormalities are:

- *Fatty acid deficiency*: Deficiency of fatty acids may develop during prolonged TPN. However, administration of 3% of the total caloric input as linoleic acid helps in preventing or correcting this deficiency.
- *Metabolic acidosis*: Hyperchloraemic metabolic acidosis may occur because of the liberation of hydrochloric acid during the metabolism of amino acids in the TPN.
- *Hypercarbia*: Hypercarbia occurs from the increased production of carbon dioxide resulting from the metabolism of large amounts of glucose. A requirement for ventilatory support or weaning difficulties may subsequently occur.
- *Hyperglycaemia*: Hyperglycaemia is a potential problem until endogenous insulin production increases, requiring frequent glucose monitoring.
- *Hypovolaemia*: Hypovolaemia due to an osmotic diuresis and a non-ketotic hyperosmolar hyperglycaemic coma are both potential complications of TPN, which may necessitate the addition of insulin to the TPN solutions.
- *Hypoglycaemia*: Accidental or sudden discontinuation of the TPN infusion may also cause hypoglycaemia because the pancreatic insulin response may persist despite discontinuing the TPN, resulting in a high plasma insulin concentration. Consequently, intravenous glucose administration may be required, or alternatively TPN may be gradually discontinued over 60–90 minutes.

Contraception

The goal of family planning is to enable couples and individuals to freely choose how many children to have and when to have them. This can best be done if the obstetrician provides them with a full range of safe and effective contraceptive methods and gives them sufficient information to ensure that they are able to make informed choices. Various contraceptive methods are based on three general strategies: prevention of ovulation; prevention of fertilisation or prevention of implantation. Various methods for contraception are described in **Table 14.3**.

The World Health Organization (WHO) eligibility criteria for the use of various contraceptive methods have been described in **Table 14.4**.

Barrier Contraception

Introduction

These methods are moderately effective, but one of the commonly used methods of contraception. These

TABLE 14.3	Various methods of contraception

Temporary Methods
- Natural regulation of fertility
- Barrier methods
- Hormonal contraception:
  - Combined hormonal contraception:
    - Combined oral contraceptive pills:
      a. Monophasic pills (each tablet containing a fixed amount of oestrogen and progestogen)
      b. Biphasic pills (each tablet containing a fixed amount of oestrogen, while the amount of progestogen increases in the luteal phase of the cycle)
      c. Triphasic pills (the amount of oestrogen may be fixed or variable, while the amount of progestogen increasing over three equally divided phases of the cycle)
  - Progestogen only contraception:
    - Progestin only pill
    - Injections (Depo-provera)
    - Implants (Norplant I and II)
    - Patches
    - Vaginal rings
- Intrauterine contraceptive devices
- Emergency (post-coital) contraception

Permanent Methods
- Sterilisation
  - Female sterilisation
  - Vasectomy

TABLE 14.4	World Health Organization eligibility criteria for the use of various contraceptive methods
Category	**Description**
1	A condition where there is no restriction for the use of the contraceptive method
2	A condition where the advantages of using the method generally outweigh the theoretical or proven risks
3	A condition where the theoretical or proven risks usually outweigh the advantages of using the method
4	A condition that represents an unacceptable health risk if the contraceptive method is used

methods aim at creating a type of barrier, which prevents the sperm from meeting the ovum. Barrier contraceptives are associated with a failure rate of 9–30 per 100 women years of use. Some of the commonly used barrier methods of contraception include male condom, female condom, diaphragm, cervical cap, vaginal sponge and spermicides.

Male Condom

A male condom is a thin sheath made of latex or other materials. The sheath is drawn or rolled onto the penis after the erection has occurred. The man puts the condom on his erected penis, while the condom holds the semen. After having sexual intercourse, the man must carefully take off the condom so that it does not leak. Each condom can be used only once. The use of condoms reduces the risk of venereal infection and is particularly important in controlling the spread of HIV infection. Condoms also provide limited protection against HPV that can cause genital warts, thereby lowering the risk for development of cervical dysplasia and cancer. Failure rates of 10–15 per 100 woman years are quoted for the condom. Occasional failures occur because of the following reasons:
- The sheath is defective.
- It is not worn in the earlier phases of coitus.
- It slips from the penis after ejaculation.

The Family Planning Association (FPA, 2014), UK now advises that spermicides should not be used with latex condoms and that spermicides do not offer additional contraceptive efficacy when used in conjunction with latex condoms. The main disadvantages associated with the use of condoms are:
- Condoms can interrupt with sexual activity, thereby interfering with sexual pleasure.
- Condoms may sometimes tear or leak and can cause an allergic reaction.

Female Condom

This comprises of strong, soft, transparent polyurethane sheath, which is inserted in the vagina before sexual intercourse. It is approximately 15 cm in length and 7 cm in diameter. It has two flexible rings, the inner ring and an outer one. The inner ring at the closed end of the condom eases insertion into the vagina, covering the cervix and holding the condom in place.

The outer ring, which is larger than the inner one, stays outside the vagina and covers part of the perineum and labia during intercourse. Female condom is available under the brand names of Reality®, Femidom®, Dominique®, etc. Female condoms may be expensive or limited in their availability and may be difficult to insert.

Diaphragm

A diaphragm is a shallow rubber dome with a firm flexible rim. It is available in different sizes ranging from 50 mm to 105 mm. It is often used in combination with contraceptive jelly, spermicide, etc. It is immediately effective and a reversible method of contraception, which can be inserted up to 6 hours before intercourse. Failure rates as high as 4–18 per 100 woman years have been quoted for the diaphragm use. Due to a high failure rate, diaphragms are more effective in the older multiparous woman, in whom the fertility is declining rather than nulliparous women. It does not prevent HIV infection. A diaphragm may, however, reduce the risk of cervical cancer.

It is usually recommended that the diaphragm must be left in situ for 6 hours or so after intercourse. It should be used with a spermicide. Spermicides are effective for about 3 hours. If the diaphragm is left in anticipation of further

intercourse some hours later, further spermicide should be inserted if the interval is 3 hours or more. It is advised that the device be removed after 24 hours because of the possible risk of infection or toxic shock. Once removed, it should be washed with soap and water and stored dry. The diaphragm should be clean, but complete sterility is not required. It is important to advise refitting of the diaphragm after each child is born and when there has been a significant weight change in the user.

The diaphragm is not an appropriate method if the man or woman has allergy to rubber, latex or spermicide, or if the woman has frequent urinary tract or bladder infections and/or anatomical abnormalities. It may be difficult to insert and remove diaphragm, and it may sometimes cause vaginal irritation.

Cervical Cap

A cervical cap is a soft, deep rubber cup with a firm, round rim that fits snugly over the cervix. The cap provides effective contraception for 48 hours. Use of cervical cap may be associated with complications such as toxic shock syndrome, unpleasant odour, discomfort and awareness of the cap during coitus and accidental dislodgment.

Spermicides

Two basic components of spermicides include active spermicidal agents such as surfactants (Nonoxynol-9, Octoxynol-9, Menfegol) and the base (carrier) agent such as foams, jellies, creams, foaming tablet, melting suppositories, aerosols, soluble films or vaginal suppositories. The woman must be instructed to insert the recommended dose of the spermicide deep into vagina to cover the cervix completely, just before sexual intercourse. A second dose of spermicide may be required if more than 1 hour passes before she has sexual intercourse. An additional application of spermicides is needed for each additional act of intercourse.

Spermicides may cause irritation in the vagina or on the penis, or an allergic reaction. They cause interruption of sexual activity. They do not provide protection against STDs.

Spermicides do not require a prescription; may be discontinued at any time and are safe. While using spermicides, douching should not be allowed for at least 6 hours after coitus.

Progestogen-only Contraceptive Methods

Progestogen-only contraceptive methods are available in various formulations:
❏ Progestogen-only pill (POP) or minipill
❏ Sub-dermal contraceptive implants (Norplant I, II and Implanon)
❏ Progestogen-only injectables (POI), e.g. depot medroxy-progesterone acetate (DMPA)
❏ Intrauterine system (Mirena and progestasert).

Progestogen-only Pill

The POPs may contain 350 μg of norethisterone or 75 μg of norgestrel or 30 μg of levonorgestrel.

Mechanism of Action

Continuous oral progestogens act primarily by causing increased viscosity of cervical mucus and endometrial changes. Ovulation is inhibited in about 60% of cycles although it does not occur in 100% of cycles without combined hormonal contraception. Progestogens also induce a premature secretory change in the endometrium, thereby making it unfavourable for implantation. The progesterone-only pill decreases tubal motility, hence its association with tubal ectopic pregnancy.

Prescription

Progesterone increases low-density lipoproteins and decreases high-density lipoproteins. Progestogen-only methods have no deleterious effect on blood pressure. There is also no association between the progesterone-only pill and thromboembolic disease. Therefore, the prescription of POPs is indicated in the following cases: hypertension, superficial thrombophlebitis, history of thromboembolism, biliary tract disease, thyroid disease, epilepsy, diabetes without vascular disease, etc. The progesterone-only pill can be safely prescribed immediately following delivery. Therefore, the POP is also recommended for women who are lactating and in whom the oestrogen component of the combined pills would interfere with lactation. Minipills are recommended over COCPs in women who are breastfeeding because they do not affect milk production.

Progestogen-only pills must be started within 5–7 days of menstruation. Unlike the COCPs, these pills must be taken on a continuous basis without any breaks between packets. These must be consumed in accordance with a strict time schedule everyday (within 3 hours vs 12 hours for COCPs). A backup method should be used for 2 days if a woman is more than 3 hours late in taking a dose. Backup contraception should be considered during the first month when the woman first starts taking minipills and then at mid-cycle every month thereafter (the time when ovulation is likely to occur).

Complications

First-generation (norethindrone, norethindrone acetate and ethynodiol diacetate) and second-generation progestins (levonorgestrel) can result in side effects such as depression/mood changes, amenorrhoea, adverse lipoprotein and carbohydrate changes, weight gain, acne, oily skin, hirsutism, etc.

The third-generation progestins (desogestrel, gestodene and norgestimate) have minimal impact on blood glucose levels, plasma insulin concentrations and the lipid profile. They have also been shown to resolve or reduce acne and

hirsutism. Furthermore, they do not adversely affect weight or blood pressure.

Some of the common complications associated with the use of POPs are as follows:

❑ Irregular bleeding, spotting, breakthrough bleeding, amenorrhoea, etc.
❑ Depression, headache, migraine
❑ Weight gain and ectopic pregnancy
❑ Mastalgia (breast tenderness)
❑ Mood swings
❑ Abdominal cramps.

Unlike the COCPs, minipills are not associated with an increased risk of complications such as deep vein thrombosis or heart disease. POPs, however, have lower effectiveness than COCPs (pregnancy rate of 2–3 per 100 women years of use)

Injectable Progestogens

These comprise of delivering certain hormonal drugs in form of deep intramuscular injections into the muscles of the arms or buttocks. The POIs contain only the hormone, progestogen. Two main types of POIs are DMPA and norethisterone enanthate (NETEN). The mechanism of action of injectable progestogens is similar to that of POPs as described previously.

Prescription

The medicine must be injected into the thigh, buttocks or deltoid muscle four times a year (every 11–13 weeks). It provides pregnancy protection starting a week after the first injection. The injection site should not be massaged afterwards, because this may accelerate absorption of the drug. Since DMPA is an aqueous suspension, a DMPA vial must be shaken vigorously, before it is loaded into the syringe, to re-suspend any active ingredient in the bottom of the vial.

Complications

❑ *Menstrual irregularities*: Disruptions of the menstrual cycle including amenorrhoea, prolonged menses, spotting between periods, and heavy or prolonged bleeding
❑ *Other side effects*: These include adverse effects such as weight gain, headache, dizziness and low bone mass.

Injectable contraceptives provide women with safe, highly effective and reversible contraceptive protection, with the failure rate being 0.1–0.4%. They overcome the inconvenience of daily compliance required with POPs or COCP. This is also a suitable method for women in whom oestrogens present health risks, for example, they have no deleterious effect on blood pressure or blood coagulation. POIs can be used by breastfeeding women at 6 weeks postpartum without adverse effects on nursing infants because progestogens do not inhibit lactation. Fertility is not impaired after discontinuation of DMPA or NETEN, although its return may be delayed. However, they do not provide protection against HIV and STDs.

Depo-Provera®

Depo-provera contains 150 mg medroxyprogesterone in an aqueous microcrystalline suspension, which is administered at every 12 weekly intervals. It may produce amenorrhoea in about 50% of women in long term.

Similar to the copper intrauterine contraceptive devices, they particularly reduce the number of intra-uterine pregnancies, but have little or no effect on ectopic pregnancy. Therefore, if a patient does become pregnant, the ratio of ectopic pregnancies to those of intra-uterine pregnancies increases.

Therefore, ectopic pregnancy must be excluded as early as possible in women who conceive despite these methods of contraception.

Intrauterine Contraceptives

Intrauterine devices (IUDs) are flexible plastic devices made up of polyethylene, which are inserted inside the uterine cavity for the purpose of contraception. Each device has a nylon thread, which protrudes through the cervical canal into the vagina, where it can be felt by the patient or the doctor. Initially, biologically inert devices such as lippes loop and saf-Tcoil were introduced, which have now been withdrawn from the market. Newer devices are medicated and contain substances such as copper, progestogens, etc. Copper carrying devices include copper T 200, copper 7, multiload, copper 250, copper T 380, copper T 220 and nova T. Their effective life varies from 3 to 5 years. Inert devices can be left in place until the menopause, but copper devices need renewal after every 3–5 years, depending on the type, due to the gradual absorption of copper. Copper IUDs produce local concentrations of copper salts that provide some protection against bacterial contamination. Pelvic infection with *Actinomyces* organisms is most likely with a plastic device that has been in situ for some years.

Intrauterine contraceptive devices containing progestogen include progestasert, levonova and Mirena. Mirena is a type of progestogen containing IUD, having 52 mg of levonorgestrel, which is released at the rate of 20 μg/day. The effects of Mirena last for about 5 years. It is sometimes also known as levonorgestrel-intrauterine system (LNG-IUS). The IUD is radio-opaque and can be located by X-ray or ultrasound. Hysteroscopy and MRI are not required, the latter is not indicated due to the metallic content of the IUD and the generation of heat associated with moving magnetic fields.

The possible mechanisms of action of IUD are as follows:
❑ Copper IUD acts as a foreign body in the uterine cavity, which makes migration of spermatozoa difficult.

❏ Increased release of prostaglandins provokes uterine contractility. This causes the fertilised egg to be rapidly propelled along the Fallopian tube so that it reaches the uterine cavity before the development of chorionic villi and thus is unable to implant.
❏ Leucocytic infiltration of the endometrium.
❏ Presence of copper results in certain enzymatic and metabolic changes in the endometrial tissues, which may inhibit implantation of the fertilised ovum.

Complications

❏ *Difficulties at the time of insertion*: Immediate difficulties at the time of insertion include vasovagal attack, difficulty in insertion of the copper device and presence of uterine cramps.
❏ *Bleeding*: Irregular menstrual bleeding, spotting, menorrhagia, etc. are the commonest side effects of IUDs in the first month after insertion. Use of NSAIDs or tranexamic acid may be helpful. Increased menstrual loss may be caused by increased fibrinolytic activity, which occurs round the IUD. The progestogen intrauterine system (IUS), however, reduces menstrual flow and often dysmenorrhoea.
❏ *Pain or dysmenorrhoea*: Pain may be a physiological response to the presence of the device, but the possibility of infection, malposition of the device (including perforation) and pregnancy should be excluded. The LNG-IUS has been associated with a reduction in menstrual pain.
❏ *Systemic hormonal side effects*: These may be typically associated with the LNG-IUS and include side effects such as depression, acne, headache and breast tenderness.
❏ *Functional ovarian cysts*: They may occur in up to 30% of LNG-IUS users and usually resolve spontaneously
❏ *Uterine perforation*: Uterine perforation is a rare, but serious complication of IUD insertion, occurring at a rate of 0.6–1.6 per 1,000 insertions. This may occur either at the time of insertion or at a later stage due to the embedment of the device into the myometrium and its subsequent migration into the intra-abdominal cavity.
❏ *Infection*: The major risk of infection is in the first 3 weeks after insertion. Hence, careful attention must be paid to asepsis. Infection at the time of insertion can result in the development of pelvic inflammatory disease (PID) in the long run. To prevent the occurrence of vaginal infection, IUD users should continue to use condoms for protection against STDs. Actinomycosis infection also occurs commonly.
❏ *Expulsion*: Expulsion of the IUD is most common in the first year of use (2–10% of users). If the IUD strings are not seen in the cervical os, the device may have been expelled or may have perforated the uterine wall. If the IUD thread is located within the cervical canal, it can be retrieved using Spencer Wells forceps or similar instrument. There are also specifically designed devices to retrieve the threads from higher up. Most IUDs can be removed in the outpatients' clinic. Alternative contraception should be arranged, if appropriate, until the IUD has been found. If the IUD strings cannot be found, ultrasound is the preferred method to identify the location of the IUD.

If the device is not identified within the uterus or the pelvis, a plain X-ray of the abdomen should be performed to determine whether the device has perforated the uterine wall. If the IUD is in utero, it can be left where it is and a periodic scan will reassure the patient about its intrauterine location. If the IUD is extra-uterine, "the Faculty of Family Planning" advises its surgical retrieval (either via laparoscopy or laparotomy).

If there is the possibility of pregnancy, then beta-hCG levels should be arranged. If the patient has become pregnant, the IUD may have been drawn up within the expanding uterus and the IUD tail may no longer be visible.
❏ *Ectopic pregnancy*: IUDs per se do not increase the risk of ectopic pregnancy. While the use of IUDs is associated with a reduced rate of intrauterine pregnancy, the rate of ectopic pregnancy is not decreased. Hence, there is a relative increase in the risk of ectopic pregnancy following IUD insertion. In women who conceive with an IUD in place, the diagnosis of ectopic pregnancy should be excluded.
❏ *Pregnancy with IUD in situ*: If an IUD is left in place, there is a slight risk of intrauterine infection, preterm labour and antepartum haemorrhage. However, most pregnancies remain uncomplicated and the device is delivered with the placenta.

Intrauterine contraceptive devices form a highly effective method of contraception with the pregnancy rate being 0.1–1 per 100 women years, with the exact range varying from one device to the other. Though IUD is commonly inserted in multiparous women, nulliparity is not a contraindication for IUD use. It can be successfully used in carefully selected nulliparous women. The copper IUD may decrease the risk of endometrial cancer.

Combined Oral Contraceptives

The combined hormonal contraception contains both oestrogen and progestogen. It is commonly available as combined oral contraceptive pills (COCPs). Three types of COCPs formulations are available: monophasic pills, biphasic pills and triphasic pills. Most COCPs are contained in a compact package of 21 active pills and 7 inactive pills. However, some 21-day packages may not contain any inactive pills. COCPs act through following mechanisms:
❏ Prevention of ovulation
❏ Thickening of the cervical mucus so that sperms cannot pass through

❏ Changing the environment of the uterus and Fallopian tubes to prevent fertilisation and/or implantation.

Complications

Minor side effects: These include clinical features such as irregular/breakthrough bleeding, breast tenderness, nausea, weight gain and mood changes. Breakthrough bleeding is related to the ratio of oestrogen to progestin in a pill formulation.

Major risks: These include side effects such as venous thromboembolism, myocardial infarction, stroke, gall-bladder disease, breast cancer, cervical cancer, etc. Oral contraceptive pills generally are not prescribed to smokers over 35 years of age. Strong smoking cessation assistance should be provided to women who wish to use oral contraceptive pills. Progesterone-only contraception is particularly appropriate for women having an absolute contraindication for oestrogen, for example, history of thromboembolism, smokers (who refuse to give it up), and diabetic patients. The patient should be counselled to report any adverse effects related to the use of COCPs, which can be remembered with the mnemonic ACHES:

❏ A—Abdominal pain (severe)
❏ C—Chest pain (severe), cough, shortness of breath or sharp pain upon breathing
❏ H—Headache (severe), dizziness, weakness or numbness (especially one-sided)
❏ E—Eye problems (complete loss of or blurring of vision)
❏ S—Severe leg pain (calf or thigh).

The use of COCPs provides a protective effect against the development of ovarian and endometrial cancer and probably even colorectal cancer. However, it may be associated with an increased risk of breast cancer, cervical carcinoma, and hepatoma. There is no evidence that the COCPs cause teratogenic effects if taken inadvertently during pregnancy. Use of COCPs is a highly effective method of reversible contraception, with the failure rate being approximately 0.1 per 100 women years of use. COCPs do not provide any protection against STD or HIV infection. Normal menstrual cycles are likely to occur in 99% of the women within 6 months of stopping the pills. However, return of fertility may be slightly late due to delayed return of ovulation.

Post-coital/Emergency Contraception

Emergency contraception (EC) also known as "post-coital contraception" or the "morning-after pill" is a method of contraception, which is used after intercourse and before the potential time of implantation. EC provides women with a safe means of preventing pregnancy following unprotected sexual intercourse or potential contraceptive failure. EC is a backup method for occasional use and should not be used as a regular method of birth control. There are two methods of EC:

1. *Hormonal methods:* This involves the use of emergency contraceptive pills. The two hormonal preparations which can be used are:
   • One containing only the progestin levonorgestrel: The regimen consists of two doses of 750 µg levonorgestrel taken orally 12 hours apart.
   • The other containing a combination of ethinyl estradiol and levonorgestrel (Yuzpe method): This method comprises of the oral administration of two doses of 100 µg ethinyl estradiol and 500 µg levonorgestrel taken 12 hours apart.
   • *Ulipristal*: Ulipristal, a selective progesterone receptor modulator was licensed for EC in 2009. It seems to be at least as effective as levonorgestrel and can be used for up to 120 hours after unprotected sexual intercourse.
2. *Non-hormonal method*: This comprises of post-coital insertion of a copper containing IUD. The copper IUD is almost 100% effective if inserted before implantation, which occurs about 5 days after ovulation.

Indications

Indications for the use of EC are as follows:
❏ Unwanted pregnancy
❏ Failure to use a contraceptive method
❏ Condom breakage or leakage
❏ Dislodgement of a diaphragm or a cervical cap
❏ Two or more missed birth control pills
❏ Depo-provera injection is late by 1 week or more
❏ Sexual assault when the woman is not using reliable contraception.

Management

A pelvic examination is not a prerequisite for providing EC. There should be no history of recent PID and vaginal or cervical infection.

Hormonal EC should be considered for any woman wishing to avoid pregnancy who presents within 5 days of unprotected or inadequately protected sexual intercourse. Although they have generally been used only up to 72 hours after intercourse, both hormonal methods of EC are effective when taken between 72 hours to 120 hours after unprotected intercourse. A post-coital IUD insertion can be considered up to 7 days after unprotected intercourse. IUDs containing at least 380 mm² of copper have the lowest failure rates and should be the first-line choice, particularly if the woman intends to continue the IUD as a method for long-term contraception.

The levonorgestrel EC regimen is more effective and causes fewer side effects than the Yuzpe regimen. One double dose of levonorgestrel EC (1.5 mg) is as effective as the regular two-dose levonorgestrel regimen (0.75 mg each dose), with no difference in the incidence of side effects. The British Medical Journal (BMJ, 2003) has recommended

a single 1.5 mg dose of LNG. It is available as "Levonelle One Step" and "Levonelle 1500" (The 0.75 mg tablets were "Levonelle 2"). The legal status of LNG as emergency contraception in the UK was changed from a prescription-only medicine to a pharmacy medicine in January 2001. It can now be bought from chemists without a prescription. A barrier method such as the condom can be used for the remainder of the current menstrual cycle, and a regular contraceptive method can be initiated at the beginning of the next cycle if the woman desires.

Complications

❑ The common side effects of hormonal EC are gastro-intestinal and mainly include nausea, vomiting, dizziness and fatigue. Antiemetics such as meclizine can be used for controlling these side effects.
❑ Less common side effects of hormonal methods include headache, bloating, abdominal cramps, and spotting or bleeding.
❑ Possible complications of post-coital IUD insertion include pelvic pain, abnormal bleeding, pelvic infection, perforation and expulsion.

Emergency contraception prevents pregnancy and does not interrupt the previously established pregnancy. ECs are not a good option for providing long-term contraception. ECs do not protect against STDs. On the contrary, approximately 5% of women seeking emergency contraception have a STD, especially *Chlamydia*. ECs do not increase the risk of ectopic pregnancy, nor do they affect future fertility. Presence of pregnancy (either confirmed or suspected) is a contraindication for the use of EC because it would not be effective in these cases.

Gynaecological Abnormalities

Amenorrhoea

Amenorrhoea or absence of menstrual periods can be of two types: primary (woman has never experienced menstrual cycles) and secondary (woman had experienced menstrual bleeding previously before experiencing cessation for at least 6 months). Causes of primary and secondary amenorrhoea are respectively tabulated in **Tables 14.5** and **14.6** respectively. Though abnormalities such as polycystic ovary syndrome, anorexia nervosa and craniopharyngioma may present with both primary and secondary amenorrhoea, they usually present with secondary amenorrhoea. Primary amenorrhoea can be defined as follows:

TABLE 14.5	Causes of primary amenorrhoea

- Genitourinary malformation, e.g. imperforate hymen, transverse vaginal septum, absent vagina with or without a functioning uterus
- Müllerian agenesis (Mayer Rokitansky Küster-Hauser syndrome)
- Androgen insensitivity: XY female or testicular feminisation. There is absence of secondary sexual characteristics
- Resistant ovary syndrome
- Hypothalamic dysfunction, e.g. chronic illness, anorexia, weight loss, stress
- Gonadotropin deficiency, e.g. Kallmann's syndrome
- Tumours of the hypothalamus or pituitary gland
- Hypopituitarism
- Hyperprolactinaemia
- Gonadal failure, e.g. ovarian dysgenesis/agenesis, premature ovarian failure
- Hypothyroidism

TABLE 14.6	Causes of secondary amenorrhoea

- Anorexia nervosa
- Congenital hypothyroidism
- Asherman's syndrome
- Virilising ovarian tumours
- Following surgical procedures of cervix (e.g. cervical stenosis)
- Use of LH-RH analogues
- Thyrotoxicosis
- Hyperprolactinaemia
- Congenital hypothyroidism

❑ Absence of menses by age of 14 years with the absence of growth or development of secondary sexual characteristics or
❑ Absence of menses by the age of 16 years with normal development of secondary sexual characteristics.

Secondary amenorrhoea is defined as cessation of menstruation for more than 6 months, but not attributable to pregnancy or the menopause. Investigations should begin after 6 months. Lactational amenorrhoea must also be excluded.

Cryptomenorrhoea

Cryptomenorrhoea means "hidden menstruation". This implies that though menstruation occurs, the menstrual blood remains concealed. Cryptomenorrhoea can occur with conditions such as imperforate hymen; cervical stenosis after cone biopsy or Manchester repair or rarely due to the congenital absence of the vagina. Crytomenorrhoea can present as both primary and secondary amenorrhoea. The usual presentation is a pubertal girl with primary amenorrhoea, recurrent abdominal pain and an imperforate hymen. The accumulation of menstrual blood within the vagina and the uterus results in the development of haematocolpos and haematometra respectively. This may produce a palpable abdominal mass, which can also be observed on ultrasound examination. A large vaginal mass

is likely to cause urethral obstruction resulting in urinary retention. Since the retained blood is usually sterile, development of infection usually does not occur.

Infertility

Infertility is defined as the inability to conceive even after trying with unprotected intercourse for a period of 1 year for couples in which the woman is under 35 years and 6 months of trying for couples in which the woman is over 35 years of age. In nearly 30% of cases, the cause can be attributed to the male partner.

Male Infertility

Introduction

Nearly 30% cases of infertility are due to the male factor. **Table 14.7** lists the causes of male infertility.

Treatment

❑ *Lifestyle modification*: The patient must be advised to discontinue smoking, stop consumption of excessive alcohol and/or intake of drugs, such as bodybuilding steroids and illicit drugs, wear loose fitting underwear and cool clothes and avoid high temperature baths like saunas, etc. Coital frequency should be increased in order to improve the chances of conception.

❑ *Medical therapy*: Some of these include clomiphene citrate, tamoxifen, gonadotropins, antibiotics (for

TABLE 14.7	Causes of male infertility

- *Idiopathic:* No obvious cause of infertility can be found.
- Varicocoele
- *Infections* (may be suspected clinically on rectal examination with tenderness of the prostate or seminal vesicles)
  - *STDs:* Chlamydia, gonorrhoea and syphilis
  - *Acute systemic infections:* Smallpox, mumps, other viral infections, etc.
  - *Chronic systemic infections:* TB, leprosy, filariasis, prostatitis, renal, hepatic diseases, diabetic neuropathy, etc.
- *Undescended testes* (cryptorchidism)
- *Genetic and endocrine disorders:* Klinefelter's syndrome, androgen insensitivity syndrome, disorders of pituitary and adrenal glands, adrenal hyperplasia, etc.
- *Substance abuse* (excessive intake of alcohol and/or drugs)
- *Testicular factors:* Torsion, undescended testes, damage to the testis due to exercise or heat, tumours (seminoma), hydrocele, etc.
- *Long-term use of drugs:* Anti-hypertensive drugs such as reserpine, methyldopa, guanethidine, cimetidine, spironolactone, propranolol, corticosteroids, anabolic steroids, antipsychotics and certain anti-cancer drugs
- *Environmental factors:* Exposure to chemicals, such as lead, nickel, mercury, anaesthetic agents, pesticides, tobacco smoking, excessive alcohol intake, etc.
- *Previous surgery:* Inguinal, scrotal, retroperitoneal, bladder neck, vasectomy, hernia repair, etc.
- *Sexual dysfunctions*: Ejaculatory disturbances, impotence, etc.

treatment of infection), steroids, etc. Vitamin E can help counter oxidative stress, which is associated with sperm DNA damage. A hormone-antioxidant combination may improve sperm count and motility. Phosphodiesterase Type 5 inhibitors, e.g. sildenafil, can be used in the patients with ejaculatory sexual dysfunction. Drug therapy has limited benefit, apart from bromocriptine, used for treating raised prolactin levels. Nonetheless, gonadotropins, clomiphene, androgens, etc. have all been used.

❑ *Caverject*: Caverject (intracavernosal alprostadil) is an effective therapy for impotence and is generally used following failure of oral phosphodiesterase inhibitors.

❑ *Squeeze technique*: The squeeze technique where the penile shaft is firmly squeezed during intercourse can help in the treatment of premature ejaculation. Fluoxetine [a selective serotonin reuptake inhibitor (SSRI)] amongst others such as clomipramine can be a useful treatment for premature ejaculation.

❑ *Surgical therapy*: Surgery may be employed for treatment of conditions, such as duct obstruction, varicocoeles, undescended testes, etc. Modern microsurgical techniques can also prove to be useful for procedures such as vasectomy reversal and tubal re-canalisation.

❑ *Assisted reproductive techniques*: This includes procedures, such as sperm washing/capacitation, intrauterine insemination, gamete intra-Fallopian transfer, in vitro fertilisation (IVF) and micromanipulation [intra-cytoplasmic sperm injection (ICSI)]. ICSI involves injection of sperm directly into an ovum, followed by IVF. It is a successful technique even with very low counts. This process cannot work if there are no sperms. After successful vasectomy, one would get azoospermia, but sperm could still be retrieved from the testicle and used for ICSI. Even precursors of sperm have been used successfully. ICSI has been found to result in successful pregnancies in men with Klinefelter's syndrome.

❑ *Incurable cases*: In cases where none of the treatment modalities seem to work, the only options may be donor insemination or adoption.

Diagnosis

General Physical Examination

These include examination of the patient for development of male secondary sex characteristics, gynaecomastia or hirsutism. The complete physical examination also includes a digital rectal examination.

Examination of Male External Genitalia

❑ *Scrotum*: This must be evaluated for the presence of congenital abnormalities, such as hypospadias, cryptorchidism, absence of vas deferens (unilateral or bilateral), etc.

❑ *Testis*: This involves assessment of testicular size, presence of tenderness on palpation of testicles and presence of any associated mass, such as an inguinal hernia or varicocoele (bag of worms appearance).

Investigations

The main test used for investigation of male infertility is semen analysis.

Semen Analysis

The primary values that are evaluated at the time of semen analysis include the volume of the ejaculate, sperm motility, total sperm concentration, sperm morphology, motility and viability.

After 3 days of abstinence, a specimen of semen should be collected in a suitably sized sterile plastic container by masturbation and examined generally within 2 hours. A normal semen analysis usually shows the values described in **Table 14.8**. Some commonly encountered abnormalities associated with abnormal sperm count are described in **Table 14.9**.

Female Infertility

Various causes of female infertility are illustrated in **Figure 14.1** and **Table 14.10**.

Impaired fertility may be associated with the following conditions:
❑ Previous ectopic pregnancy
❑ Previous pelvic infection
❑ Cystic fibrosis (in a male)
❑ Reversal of vasectomy (in a male)

TABLE 14.8	Normal parameters for semen analysis (World Health Organization, 4th Edition, 1999)
Parameter	**Normal reference value**
Volume	2–5 mL
Liquefaction	Complete in 30 minutes
Sperm density	>20 million spermatozoa per mL or more
Total sperm number	40 million spermatozoa per ejaculate or more
Motility	50%, forward progression; 50% or more motile (grades a* and b**) or 25% or more with progressive motility (grade a) within 60 minutes of ejaculation
Sperm morphology	>50% normal (WHO, 1987, 2nd Edition) >30% normal (WHO, 1992, 3rd Edition) >14% normal (WHO, 1999, 4th Edition)
White blood cells	<1 million/mL
Immunobead test	<20% spermatozoa with adherent particles
SpermMar test	<10% spermatozoa with adherent particles

* Grade a: Rapid progressive motility (sperm moving swiftly, usually in a straight line)
** Grade b: Slow or sluggish progressive motility (sperms may be less linear in their progression)

Polycystic Ovarian Syndrome

The condition, polycystic ovarian syndrome, also known as PCOS, is a relatively common endocrine disorder amongst women of reproductive age group. It is characterised by the presence of many minute cysts in the ovaries and excessive production of androgens. According to the American Society of Reproductive Medicine (ASRM) and the European Society of Human Reproduction and Embryology (ESHRE) joint consensus meeting in November 2003, the diagnosis

TABLE 14.9	Abnormalities associated with abnormal sperm count in the semen
Semen abnormality	**Definition**
Oligozoospermia	Less than 20 million spermatozoa per mL
Azoospermia	No spermatozoa in the semen
Teratozoospermia	Excess of abnormally formed spermatozoa

TABLE 14.10	Causes of female infertility

Cervical factor infertility
• Abnormalities of the mucus-sperm interaction
• Narrowing of the cervical canal due to cervical stenosis
Uterine factor infertility
• Total absence of the uterus and vagina (Rokitansky-Küster-Hauser syndrome)
• DES-induced uterine malformations
• Asherman's syndrome, endometritis (due to tuberculosis)
• Leiomyomas
Ovarian factor infertility
• Polycystic ovarian syndrome
Tubal factors
• PID associated with gonorrhoeal and chlamydial infection
Peritoneal factors
• Infection
• Adhesions and adnexal masses
• Endometriosis

Abbreviations: DES, diethylstilbestrol; PID, pelvic inflammatory disease
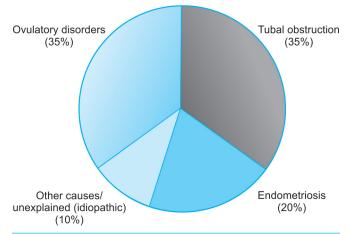
FIG. 14.1: Female causes of infertility

of PCOS should be made, when two of the following three criteria are met:
1. Infrequent or absent ovulation
2. Clinical or biochemical features of hyperandrogenism, such as excessive hair growth, acne, raised LH and raised androgen levels
3. Morphologically, there is bilateral ovarian enlargement, thickened ovarian capsule, multiple follicular cysts (usually ranging between 2 mm to 8 mm in diameter) and an increased amount of stroma.

Clinical Features

Polycystic ovarian syndrome is characterised by the following features:
- Hirsutism
- Oligomenorrhoea (usually with normal oestrogen concentrations)
- Infrequent or absent ovulation
- Obesity
- The risk of endometrial hyperplasia and carcinoma may be increased.
- Miscarriage and infertility.

Diagnosis

Diagnosis of PCOS is established by ultrasound examination and serum hormonal assay.
- *Blood hormone levels*: Various endocrinological abnormalities encountered in cases of PCOS are enumerated in **Table 14.11**. FSH levels are low or normal and LH levels are often raised, resulting in a raised LH/FSH ratio. The levels of androgens and testosterone may also be raised.
- *Ultrasound examination*: Features of polycystic ovarian morphology on ultrasound scan are as follows **(Fig. 14.2)**:
  - Greater than 12 follicles measuring between 2 mm and 9 mm in diameter, located peripherally, resulting in a pearl necklace appearance
  - Increased echogenicity of ovarian stroma and/or ovarian volume greater than 10 mL.

Progesterone levels in excess of 30 nmol/litre indicate ovulation. If there is evidence of ovulation, there is no point in prescribing clomiphene, unless ovulation and menstruation are very infrequent.

TABLE 14.11	Endocrinological abnormalities in cases of polycystic ovarian syndrome

- Increased oestrone levels
- low SHBG
- Increased free testosterone levels
- Increased LH and decreased FSH levels. An LH:FSH ratio greater than 2.5 or 3 is used to diagnose polycystic ovary syndrome
- Androgen levels: Increased production from ovaries and adrenals
- Hyperinsulinaemia
- Hyperprolactinaemia (low to moderate levels <2,500 mU/L)

Abbreviations: SHBG, sex hormone-binding globulin; LH, luteinising hormone; FSH, follicle-stimulating hormone

Medical Treatment

- *Lifestyle changes*: Exercise to maintain a normal body mass index
- *Ovulation induction drugs*: The treatment of choice in patients with PCOS is ovulation induction with clomiphene citrate, which is associated with nearly 70% rate of ovulation after the first treatment cycle.
- *Metformin*: Metformin has now become the first line of management in cases of clomiphene citrate-resistant women with PCOS.

Surgical Management

Laparoscopic ovarian drilling (LOD) is sometimes used for women with PCOS, who do not respond to first-line treatment options, such as weight loss and use of medicines. In LOD, different techniques, such as electro-cauterisation, laser, electro-coagulation, biopsy, etc. are used for destroying ovarian follicles.

Ovarian Hyperstimulation Syndrome

Ovarian hyperstimulation syndrome is an iatrogenic condition that occurs in patients undergoing ovulation induction with clomiphene or human menopausal gonadotropin (hMG) or controlled ovarian hyperstimulation for assisted reproductive technologies. The pathophysiology of the disease is not well understood but is associated with massive extravascular accumulation of fluid. This causes severe depletion of the intravascular volume resulting in dehydration, haemoconcentration, and electrolyte imbalance (i.e. hyponatraemia, hyperkalaemia, etc.). Ovaries become dramatically enlarged by the presence of cystic follicles, ascites of varying degrees follows, and in the most severe cases, pleural effusion, hypovolaemia with an increased tendency to thrombosis and emboli, and even
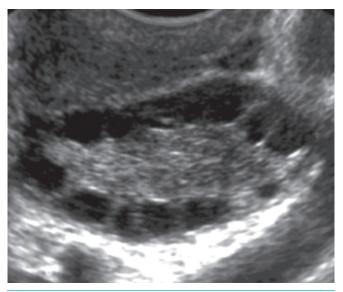
FIG. 14.2: Ultrasound features of polycystic ovarian morphology

renal failure and death can occur. Ovarian hyperstimulation syndrome can be classified as mild, moderate or severe. Although this condition is idiosyncratic, it is more common in presence of the following conditions:
- When there are many follicles (say more than 15)
- When the plasma oestrogen level has exceeded 2,500 pg/mL on the day of hCG administration
- Where pregnancy has occurred.

It is thus sensible for the hMG regime to be used only in cases where there are appropriate facilities for monitoring and for taking care of the complications related to ovarian hyperstimulation syndrome.

Asherman's Syndrome

Asherman's syndrome is characterised by development of intrauterine adhesions, occurring in women who have had endometrial trauma associated with vigorous curettage, especially following abortion or delivery. The incidence becomes even more pronounced if a pre-existing or post-operational infection occurs. These adhesions may cause amenorrhoea, repeated miscarriages, infertility and ectopic pregnancy. Diagnosis of Asherman's syndrome can be reached by doing tests like hysteroscopy and transvaginal ultrasound examination. Treatment involves hysteroscopic surgery to cut and remove the adhesions or scar tissue. After the removal of scar tissue, the uterine cavity must be kept open. Over the years, many surgical adjuncts have been tested in an attempt to prevent the reformation of adhesions. Some of these surgical adjuncts include IUDs (inert), intrauterine Foley's catheters, anti-adhesion barriers, amnion grafts, hormonal therapy, antibiotic therapy, etc. Post-operative evaluation for reformation of adhesions in form of hysterosalpingography (HSG) or hysteroscopy should be considered mandatory. Following the treatment of Asherman's syndrome, the rate of fertility restoration is high, but not 100%.

Fallopian Tube Occlusion

Pelvic inflammatory disease (PID) is the most important cause of Fallopian tube obstruction. PID is typically associated with gonorrhoeal and chlamydial infection. Chlamydial infection, which may be asymptomatic, can cause considerable tubal damage. It is more common than gonorrhoea as the infection responsible for causing Fallopian tube occlusion. However, the mechanism by which infection ascends through the cervical canal and reaches the Fallopian tubes is still unknown.

Formation of peritoneal adhesions secondary to PID can compromise the motility of the Fallopian tubes. Furthermore, obstruction of the distal end of the Fallopian tubes results in accumulation of the normally secreted tubal fluid, creating distention of the tube. This subsequently causes damage to the epithelial cilia and may result in development of hydrosalpinx. A hydrosalpinx may be seen

TABLE 14.12 Causes of dyspareunia
- Superficial vulvovaginitis (especially infection by *Trichomonas* or *Candida*)
- Vaginal cysts
- Infection of Bartholin's gland
- Post-menopausal shrinkage
- Thick hymen (rarely)
- Deep retroverted uterus with prolapsed ovaries (the "ovarian entrapment" syndrome)
- Chronic pelvic infection
- Endometriosis/adenomyosis
- Pelvic tumours including ectopic pregnancy

ultrasonographically, but occlusion of the tube and fimbrial end clubbing cannot be diagnosed by this means.

Tubal occlusion is surprisingly uncommon even in the presence of moderately severe pelvic endometriosis. On the other hand, appendicitis can result in considerable tubal damage, both from the local pelvic inflammatory reaction and the associated surgery.

Test for Tubal Patency

The injection of a radio-opaque aqueous solution through the cervix under radiographic control is a useful investigation for assessment of tubal patency and uterine shape. Laparoscopic examination of the uterus and Fallopian tubes has presently become the method of choice for investigating tubal patency. The procedure is generally combined with injection of a dilute solution of methylene blue or indigo carmine dye through a tightly fitting cannula placed in the cervical canal. If the tubes are patent, they fill with dye that then can be seen spilling from the distal ends.

Dyspareunia
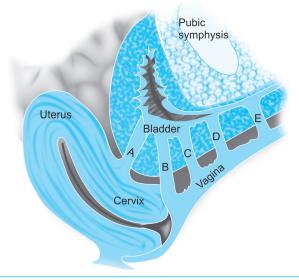
Dyspareunia can be defined as difficult or painful sexual intercourse. As a result, dyspareunia is often related with sexual dysfunction and infertility. Some causes of dyspareunia are summarised in **Table 14.12**.

Genital Tract Fistulae

Urogenital fistulas (UGFs) can be defined as abnormal communication tracts (lined with epithelium) between the genital tract and the urinary tract or the alimentary tract or both. UGFs can be classified as follows **(Fig. 14.3)**:
- Urethrovaginal
- Vesical fistula [vesicovaginal fistula (VVF) or vesico-cervical]
- Ureterovaginal
- Rectovaginal.

Vesicovaginal fistula is an abnormal fistulous tract extending between the bladder and the vagina that allows the continuous involuntary discharge of urine into the vaginal vault.

FIG. 14.3: Types of genitourinary fistulas

Abbreviations: A, uterovesical fistula; B, cervicovesical fistula; C, midvaginal VVF; D, VVF involving the bladder neck; E, urethrovaginal fistula

Causes

❑ *Obstetric injury*: Most rectovaginal fistulae are the result of unrepaired third degree lacerations of the perineum and posterior vaginal wall, or repairs that have broken down, so that an opening is left from the rectum into the vagina. Obstructed labour is an important cause for development of VVF in developing countries. In cases of obstructed labour, disproportion between the size of the pelvic opening and the foetal head results in prolonged labour, causing compression of the bladder base between the foetal head and the pubic bone, leading to avascular necrosis of the bladder. This commonly results in development of a VVF. The diagnosis of VVF is generally obvious from the history of urinary incontinence and a constant leakage of urine from the vagina. The fistula can be visualised through the direct inspection of the anterior vaginal wall, using a Sim's speculum.

❑ *Surgery:* The bladder may be opened during the course of obstetric and gynaecological operations, especially during caesarean section and abdominal or vaginal hysterectomy.

❑ *Complication of radiotherapy*: They are most likely to occur during the treatment of advanced growths, especially if the radiation dose is excessive.

❑ *Neoplastic growths*: Carcinoma of the cervix may invade the bladder and eventually cause a fistula. Advanced carcinoma of rectum or vagina may give rise to colovaginal fistulae, which may result from rupture of a pericolic abscess into the posterior vaginal fornix; the abscess is usually secondary to acute diverticulitis.

Fistula Repair

Fresh injuries may be repaired immediately. However, in cases where the fistula is noticed some days after the injury, the timing of the surgery is important. Several weeks may be required to allow any urinary tract infection (UTI) or local inflammation to be eliminated. Most vesicovaginal fistulae can be closed by surgery via the vaginal route. The principle of the surgery is to separate the bladder mucosa from the vaginal skin. The mucosa is then carefully closed in one or two layers, without tension, using polyglycolic acid sutures.

Pelvic Prolapse

Uterine prolapse can be described as a descent or herniation of the uterus into or beyond the vagina. Weakness of the anterior compartment results in cystocoele and urethrocoele, whereas that of the middle compartment in the descent of uterine vault or uterine prolapse and enterocoele. The weakness of the posterior compartment results in rectocoele.

Symptoms

Symptoms produced as a result of pelvic prolapse include the following:

❑ *Local discomfort*: There may be vaginal discomfort, dragging and the sensation of "something coming down" the vagina, sensation of lump in the vagina, a feeling of pelvic insecurity and low backache. Sensation of "something coming down the vagina" results due to the bulging of prolapsed part into the vagina and eventually protrusion through the vaginal opening.

❑ *Backache*: Backache due to prolapse is worse on standing and is relieved when the patient lies down. In the majority of women with prolapse, backache may be due to some other cause.

❑ *Urinary symptoms*: Urinary symptoms, such as difficulty in passing urine and recurrent UTIs, may be associated with cystocoele and cystourethrocoele.

❑ *Bowel symptoms*: A patient with a rectocoele may have difficulty passing stool; manual manipulation may be required for complete defecation.

❑ *Ulceration and bleeding*: Blood stained vaginal discharge may be present in the cases of procidentia and decubitus ulcerations.

❑ *Dyspareunia.*

The sensation of prolapse is increased on coughing, standing or exertion and is relieved by lying down. Sometimes, the apparent uterine prolapse may be due to a significant elongation of the cervix.

Supports of the Uterus

The uterus and vagina are held in the pelvis by the cardinal and uterosacral ligaments and by the pelvic floor
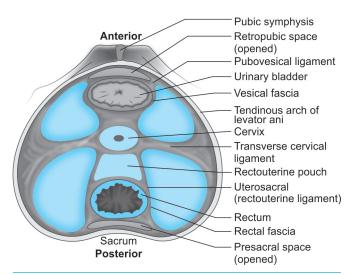
FIG. 14.4: Ligaments of the pelvic floor

FIG. 14.5: Muscles of the pelvic floor

TABLE 14.13	Different levels of support for vaginal tissue
Different levels of support of vagina	**Support elements**
Level I (for proximal one-third of vagina)	Cardinal and the uterosacral ligaments
Level II (for middle one-third of vagina)	Paravaginal fascia
Level III (for distal one-third of vagina and the introitus)	Levator ani and perineal muscles

musculature, mainly the levator ani muscles. Different levels of support for vaginal tissue are described in **Table 14.13**.

Important ligaments supporting the uterus are described in **Figure 14.4**. The most important ligaments supporting the uterus are the transverse cervical or cardinal ligaments that attach the cervix and vaginal vault to the sidewalls of the pelvis. Cardinal ligaments fan out laterally from the vaginal vault and attach to the anterior border of the greater sciatic foramen and ischial spines and the parietal fascia of the obturator internus and piriformis muscles. The cardinal ligaments contain the uterine arteries and provide attachment of uterus to the pelvic side walls.

Other important ligaments, which help in supporting the uterus are the uterosacral ligaments, which pass upwards and backwards from the cervix and vaginal vault to blend with the fascia covering the front of the second and third sacral segments.

The round ligaments help to keep the uterus anteverted, but have little or no supporting function. Similarly, the broad ligament is not a ligament but a peritoneal fold, and it does not support the uterus.

Muscles of the Pelvic Floor: Levator Ani

The levator ani muscle, the most important muscle of the pelvic floor **(Fig. 14.5)**, consists of a pair of broad, flat muscles, the fibres of which pass medially, downwards and inwards. Together with its fellow on the opposite side, the two muscles constitute the pelvic diaphragm.
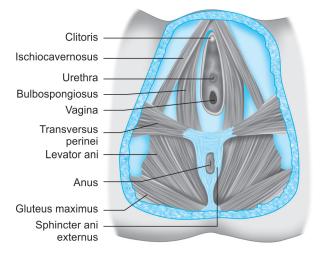
Each levator ani muscle consists of three main divisions: (1) pubococcygeus, (2) iliococcygeus and (3) ischiococcygeus. The levator ani muscle creates a hammock-like structure, by extending from the left tendinous arch to the right tendinous arch. The muscle has openings, through which the vagina, rectum and urethra traverse. Contraction of the levator muscles tends to pull the rectum and vagina inwards towards the pubic symphysis. This causes narrowing and kinking of both vagina and rectum. The origin of levator ani muscles is fixed on the anterior end, because the muscle arises anteriorly either from the bone or from the fascia, which is attached to the bone. As a result, the anterior attachment of the muscle largely remains immobile. On the other hand, the levator ani muscles posteriorly get inserted into the anococcygeal raphe or into the coccyx, both of which are movable. Thus, the contraction of levator ani muscles tends to pull the posterior attachment towards the pubic symphysis. The main nerve supply of the levator ani muscles comes from the third and fourth sacral nerves. The uterus does not rest on the levator muscles but is held in place at a higher level by the ligaments and connective tissues of the pelvic fascia.

The periurethral levator ani is under voluntary control and actively contracts during abdominal straining. As soon as the bladder fills to its functional capacity, a signal from within the detrusor muscle receptors is sent to higher cortical centres in the brain to initiate the emptying phase. Normal voiding occurs when urethral muscles relax before the detrusor muscle contracts. The voiding process begins with the inhibition of both sympathetic relaxation of the detrusor muscle and of sympathetic contraction of the proximal urethral sphincter. This is followed by the inhibition of the pudendal and sacral efferent nerves, resulting in the relaxation of the external urethral sphincter and levator ani muscles. Finally, parasympathetic stimulation via the interaction of released acetylcholine and cholinergic receptors causes the detrusor muscle to contract, thereby emptying bladder contents.

Rectal distention stimulates relaxation of internal anal sphincter and the sampling reflex. If defecation is to be delayed, voluntary contraction of the external anal sphincter and levator ani muscles occurs. Accommodation refers to the relaxation of the rectal ampulla after an initial increase in pressure. At the appropriate time for defecation or when rectal pressure is high, the levator ani muscle, puborectalis muscle and external anal sphincter relax.

Relaxation of the pelvic floor, along with a squatting position, straightens the anorectal angle. An increase in abdominal pressure along with colonic and rectal contractions allows expulsion of a faecal bolus.

Perineal Body

The perineal body, a pyramid-shaped fibro-muscular structure lying at the centre of perineum (midpoint between the vagina and the anus), assumes importance in providing support to the pelvic organs as it provides attachment to the following eight muscles of the pelvic floor: superficial and deep transverse perineal muscles; levator ani muscles of both the sides; bulbocavernosus anteriorly and the external anal sphincter posteriorly **(Fig. 14.6)**. It lies in front of the anal canal, and behind the posterior border of the perineal membrane.

Management

The only definitive cure for prolapse is surgery. Indications for various surgeries performed for uterine prolapse are described in **Table 14.14**. In patients unfit for surgery, non-surgical management approaches are sometimes used. Non-surgical management must be primarily used in cases with mild degree of uterovaginal prolapse with no or minimal symptoms. The current mainstays of non-surgical management of patients with uterine prolapse consist of expectant management including the pelvic floor exercises (Kegel exercises) and pessaries.

Ring Pessary

Pessaries are a non-surgical method for supporting the uterine and vaginal structures.Various indications for using a pessary are described in **Table 14.15**.

Manchester Repair

Manchester repair is performed in those cases where removal of the uterus is not required. Indications of Manchester operation are described as follows:
❑ Childbearing function is not required.
❑ Malignancy of the endometrium has been ruled out by performing a dilatation and curettage.
❑ Absence of UTI
❑ Presence of a small cystocoele with only first- or second-degree prolapse
❑ Absence of an enterocoele
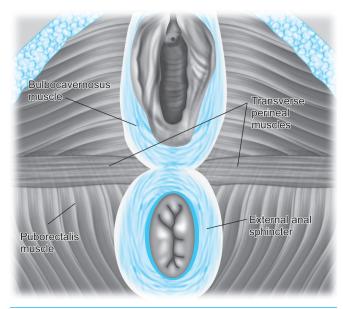
FIG. 14.6: Attachments of perineal body

TABLE 14.14	Indications for various surgeries performed for uterine prolapse

Hysterectomy
• Removal of a non-functioning organ in postmenopausal women
• Concomitant uterine or cervical pathology (e.g. large fibroid uterus, endometrial carcinoma, etc.)
• Patient desires removal of the uterus

Anterior Colporrhaphy
• Presence of cystocoele, urethrocoele or a cystourethrocoele
• Repair of anterior defects

Posterior Colpoperineorrhaphy
• Presence of a rectocoele
• Repair of posterior defects

Manchester Operation
• Childbearing function is not required.
• Malignancy of the endometrium has been ruled out.
• Absence of UTI
• Presence of a small cystocoele with only a first- or second-degree prolapse
• Absence of an enterocoele
• Symptoms of prolapse are largely due to cervical elongation.
• Patient requires preservation of menstrual function.

Le Fort Colpocleisis
• No sexual activity at present or no plans for sexual activity in future
• Patient is medically fragile.

TABLE 14.15	Current indications for using a pessary

• A young woman planning a pregnancy in future
• During early pregnancy, immediately after delivery and during lactation
• Temporary use while clearing infection and decubitus ulcer prior to the actual surgery
• Women unfit for surgery (patient is unfit for an anaesthetic)
• Women who do not desire surgery
• The patient refuses surgery or whilst awaiting surgery.

❏ Symptoms of prolapse are largely due to cervical elongation. Patient requires preservation of the menstrual function.

This surgery involves amputation of the cervix after exposing, clamping and cutting the Mackenrodt's (cardinal) ligaments. These are shortened and sutured back onto the anterior surface of cervical stump. As the cardinal ligaments run from the cervix to the sidewalls of the pelvis, this process helps lift up the uterus. Anterior colporrhaphy is almost always done as well, with posterior colporrhaphy and repair of enterocoele if present.

Patients should not become pregnant after this surgery because it carries a significant risk of premature labour and prolapse recurrence. Caesarean section needs to be considered in these cases to reduce the risk of prolapse recurrence, though pregnancy itself increases this.

Incontinence

Urinary incontinence can be defined as an involuntary loss of urine, which is a social or hygienic problem and can be demonstrated with objective means. There are two main types of urinary incontinence: stress incontinence and urge incontinence.

Stress Urinary Incontinence

Stress urinary incontinence (SUI) can be defined as involuntary leakage of urine during conditions causing an increase in intra-abdominal pressure (exertion, sneezing, coughing or exercise) which causes the intra-vesical pressure to rise higher than that which the urethral closure mechanisms can withstand (in the absence of detrusor contractions). Urine loss is instantaneous and is often described as a "squirt" of urine. SUI is often associated with other pelvic relaxation problems, for example, cystocoele, rectocoele and uterine prolapse.

Bladder Urodynamic Studies

Urodynamic studies are a method for assessing the pressure-flow relationship between the bladder and the urethra. This helps in defining functional status of the lower urinary tract, which ultimately helps in correctly diagnosing the type of urinary incontinence based on the pathophysiology. Urodynamic investigation of bladder function involves an investigation of bladder movements and tensions during different levels of filling, and includes measurement of bladder activity (cystometry) and urethral flow (uroflowmetry). The cystometry is a fundamental test of bladder function and measures changes in bladder pressure with changes in bladder volume. Normal urodynamic findings in an adult female are described in **Table 14.16**.

Management

Surgery forms the mainstay of treatment for cases of stress incontinence. Various procedures for stress incontinence share the common goal of stabilising the bladder neck and proximal urethra. Various procedures available for urinary incontinence are retropubic bladder neck suspension procedures or colposuspension; transvaginal urethropexies/needle suspension procedures/Pereyra's procedure; and sub-urethral sling procedures and peri-urethral injections.

Retropubic Bladder Neck Suspension Procedures or Colposuspension

All these procedures are performed through lower abdominal (transverse supra-pubic) incision and involve the attachment of peri-urethral and peri-vesical endopelvic fascia to some other supporting structure in the anterior pelvis **(Table 14.17** and **Fig. 14.7)**. Nowadays, some of these procedures are performed through laparoscopic and robotic surgery.

Colposuspension mainly deals with stress incontinence and this operation is superior to vaginal surgery in the form of anterior colporrhaphy, except in cases with significant degrees of prolapse. Exactly how this restores continence is a matter of debate, but the operation is effective in about 80%. One theory is that the bladder neck is lifted above the pelvic floor. In this location, the bladder and proximal urethra are subject to the same external pressures, e.g. from coughing sneezing, etc. So long as the "urethral closing pressure", from the intrinsic tone of its musculature and the effect of surrounding tissues, is higher than the pressure inside the

TABLE 14.16	Normal urodynamic findings in the adult female
Parameter	**Normal value**
Voiding pressure	45–70 cm H_2O
Residual urine	50 mL
First sensation of bladder filling	150–200 mL
Maximum voiding pressure	70 cm H_2O (some people consider 60–70 cm H_2O as borderline for obstruction)
Bladder capacity	400–600 mL
Intra-vesical pressure rise during early filling	Less than 10 cm H_2O
Maximum urine flow rate	60 mL per second
Voiding volume	250 mL
Residual volume	Less than 10 mL

TABLE 14.17	Various supporting structures in different types of retropubic procedures
Name of surgical procedure	**Supporting structure in the anterior pelvis**
Paravaginal procedure	Arcus tendineus
Modified Marshall-Marchetti-Krantz procedure	Back of pubic symphysis
Burch colposuspension	Iliopectineal ligament (Cooper's ligament)
Turner-Warwick vaginal obturator shelf procedure	Fascia over obturator internus
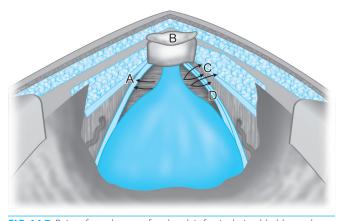
FIG. 14.7: Point of attachment of endopelvic fascia during bladder neck suspension procedures
Abbreviations: A, arcus tendinous; B, periosteum of the pubic symphysis; C, iliopectineal ligament (Cooper's ligament); D, obturator internus fascia

bladder, continence will exist, regardless of various types of stress (e.g. coughing, etc.). If the bladder neck is below the pelvic floor, coughing pressures are not transmitted through the pelvic floor to the urethra, but continue to affect the bladder. This can increase intra-vesical pressure above the "urethral closing pressure" and may result in incontinence.

It is important to counsel patients that a small percentage can develop detrusor instability or voiding difficulty after the operation. Also, the surgery does not resolve uterine descent or posterior wall prolapse, with the latter often being more pronounced after the operation. Frequency, nocturia, and urgency usually indicate bladder dysfunction, which is treated with a combination of behavioural and medical treatments.

Most patients who used to have colposuspension are now having transvaginal tape (TVT), transobturator tape (TOT) or equivalent. In TVT, a tape is inserted underneath the urethra with the ends being passed up each side of the urethra behind the pubic bone. It thus forms a "U" shape and provides support to the bladder neck and proximal urethra during increased intra-abdominal pressure. It seems to be roughly as effective and durable as colposuspension, with similar incidences of bladder instability and urinary retention. It is an easy and quick procedure and the patient can be discharged home earlier, usually within 24 hours. TVT was the first of such procedures on the market, but is now being challenged by alternatives like TOT, which is a technically easier surgery.

Transvaginal Urethropexies/Needle Suspension Procedures/Pereyra's Procedure

This involves passage of sutures between the vagina and anterior abdominal wall using an especially designed long needle carrier, which is inserted through the vaginal incision made at the level of bladder neck. The other end of the suture passes through a small abdominal incision which is made transversely just above the pubic bone

and is carried down to the rectus fascia. Anterior repair elevates the bladder neck from below and, whilst there is a recurrence rate, is useful in the elderly or the physically frail. Bladder neck suspension elevates the bladder from above and below. It also elongates the urethra preventing pressure transmission to the posterior urethra.

Sub-urethral Sling Procedures and Peri-urethral Injections

These methods are used for stress incontinence resulting from intrinsic sphincteric damage or weakness. Peri-urethral injections increase urethral resistance and are particularly useful in the old patients or those who have failed surgery previously. Both these methods work by compressing the urethral lumen at the level of bladder neck to compensate for a faulty urethral closure mechanism. Various materials have been used for making slings such as synthetic materials, cadaveric donor fascia, endogenous rectus fascia, fascia lata, etc. Sling operations can be performed using a combined vaginal and abdominal approach and involve mid-urethral placement of mesh.

Peri-urethral injections are performed under local anaesthesia and involve administration of various types of materials around the peri-urethral tissues to facilitate their coaptation under conditions of increased intra-abdominal pressure. Various bulking agents have been used including collagen; carbon-coated zirconium; ethylene vinyl alcohol; polydimethylsiloxane; polytetrafluoroethylene and glutaraldehyde cross-linked bovine collagen (contigen).

Urge Incontinence

Urge urinary incontinence can be defined as involuntary leakage of urine accompanied by or immediately preceded by urgency. The corresponding urodynamic term is detrusor overactivity, which is evident in the form of involuntary detrusor contractions at the time of filling cystometry. Urge incontinence is caused by uninhibited contractions of the detrusor muscle. Urge incontinence is worse in the night because of bladder filling. Generally, urine volume is less than 2 litres daily.

Management of Detrusor Overactivity

Management options for urge incontinence are enumerated in **Table 14.18** and are described next in details.

TABLE 14.18	Treatment of urge incontinence
Treatment option	**Kind of treatment**
First-line treatment	Behavioural therapies such as bladder training and bladder drill help in establishing or re-establishing cortical control over a hyperactive micturition reflex
Second-line treatment	Medical treatment: Anti-cholinergic drugs oxybutynin (Ditropan) or imipramine (Tofranil)
Third-line treatment	Surgical procedures (rarely used)

Medical Treatment

Pharmacological management options for urge incontinence include the following:

- *Anticholinergic agents*: Propantheline bromide is an anticholinergic agent which is commonly prescribed in the dosage of 15–30 mg every 4–6 hours.
- *Tricyclic antidepressants*: They possess both central and peripheral anti-cholinergic effect as well as alpha-adrenergic agonist effect and central sedative effect. The resultant clinical effect is bladder muscle relaxation and increased urethral sphincter tone.
- *Musculotropic relaxants*: The main smooth muscle relaxant used in these cases is oxybutynin in the dosage of 5 mg, 2–4 times per day.
- *Behaviour modification*: Behavioural interventions help in establishing or re-establishing cortical control over a hyperactive micturition reflex.
- *Intermittent catheterisation*: This type of management is most appropriate for patients with detrusor hyper-reflexia and functional obstruction.
- *Vaginal prosthetic devices*: A disposable vaginal device made of polyurethane has been found to be moderately effective in patients with detrusor overactivity.

Surgical Treatment

Surgical therapy should be considered only in severe and refractory cases of urge incontinence and include bladder augmentation procedures, denervation procedures, urinary diversion, sacral neuromodulation, etc.

Hirsutism

Hirsutism is defined as the presence of coarse, dark, terminal hair in a male pattern in a woman. The commonest areas, where increased hair growth is apparent are upper lips, chin, side burns, upper abdomen, back, breasts, inner thighs, chest and linea alba of abdomen. Hirsutism only affects women. Excessive hair in cases of hirsutism is "terminal" hair, coarse and pigmented as opposed to "vellus" hair, the fine hair that covers much of the body. Hirsutism can be scored using the modified Ferriman-Gallwey system. A score of greater than 8 is considered as diagnostic. Various causes of hirsutism are summarised in **Table 14.19**. Hirsutism is more likely to have a serious underlying cause if it is severe, of sudden onset or pre-pubertal, associated with signs of virilisation or associated with amenorrhoea.

Hirsutism is in contrast to virilisation, which reflects very high levels of androgens and manifests in form of features such as deepening of the voice; enlargement of the clitoris; male pattern hair loss, e.g. temporal recession; breast atrophy; increased muscle mass, etc. Women with virilism will have hirsutism, but vice versa is usually not true.

Another term, which can be confused with hirsutism, is hypertrichosis that refers to excess growth of fine hair in males and females.

Investigations

Investigation of a case of hirsutism involves taking a detailed history and performing a complete clinical examination. This involves elicitation of the risk factors for significant disease as enlisted in **Table 14.20**.

Investigations

Management of hirsutism is described in **Figure 14.8**. The following investigations must be ordered:

- *An ultrasound scan of the pelvis*: This helps in diagnosis of conditions such as polycystic ovary syndrome, ovarian tumours, etc.

TABLE 14.19 Causes of hirsutism

- *Polycystic ovary syndrome* (up to 80% of all cases)
- *Idiopathic hirsutism* (up to 15% of all cases)
- *Other causes:*
  - Conditions associated with raised androgen levels:
    - HAIR-AN syndrome [hyperandrogenism (HA), insulin resistance (IR), and acanthosis nigricans (AN)]
    - Late-onset congenital adrenal hyperplasia
    - Androgen-secreting tumours (adrenal and ovarian)
    - Cushing's syndrome
    - Hypothyroidism (increased concentration of SHBG, resulting in high levels of free testosterone)
  - Conditions associated with normal androgen levels:
    - Acromegaly
    - Drugs (Androgenic agents: danazol, 19-nortestosterone derived progestogens e.g. "Primolut-N", norethisterone, anabolic steroids, etc. and non-androgenic agents: methyldopa, metoclopramide, phenothiazines, phenytoin, valproate, etc.)

Abbreviation: SHBG, sex hormone binding globulin

TABLE 14.20 Risk factors for significant hirsutism to be elicited at the time of taking history and performing clinical examination

- Recent onset
- Rapid progression
- Severe hirsutism
- Signs of acromegaly or Cushing's disease
- Signs of virilisation
  - Deepening of the voice
  - Enlargement of the clitoris
  - Reduction in breast volume
  - Hair loss
  - Acne
- Family history of hirsutism
- Ethnicity (hair distribution is associated with huge racial variation)
- Drugs
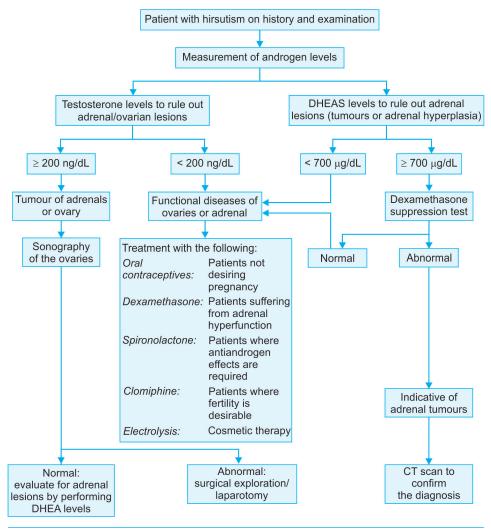
FIG 14.8: Management of hirsutism

Abbreviations: DHEA, dehydroepiandrosterone sulphate; CT, computed tomography

❑ A basic hormone profile:
- Total and free testosterone (total testosterone levels >5 mmol/litre indicate possible adrenal disease)
- 17-hydroxyprogesterone (OHP) levels (this is raised in cases of congenital adrenal hyperplasia)
- Other investigations might be added on an individual basis (e.g. tests for acromegaly, Cushing's disease, etc.)

Treatment

❑ Counselling or psychotherapy
❑ *Weight loss*: This is one of the most effective approaches, which is particularly useful for women having obesity and PCOS. It increases SHBG, thereby reducing the level of androgens. It can help restore ovulatory cycles and fertility in the absence of any other treatment.
❑ *Physical treatment*: These include several methods such as shaving, depilatory creams, waxing, bleaching, electrolysis, thermolysis, laser treatment, etc. The laser frequencies suitable for destroying the hair follicle are those that melanin absorbs. It works best for dark hair.
❑ *Drug treatment*: This can include hormonal and non-hormonal treatment.

Hormonal Treatment

❑ *Oral contraceptive*: All of the combined oral contraceptives will have an effect by reducing LH production. They also increase SHBG, thereby reducing the levels of free testosterone. The treatment may take months to produce significant improvement.
❑ *Dianette*: This is an oral contraceptive with 35 μg of ethinyl oestradiol and 2 mg of cyproterone acetate. For further details related to dianette and cyroterone acetate, kindly refer to Chapter 12.
❑ *Yasmin* ®: This is the trade name of an oral contraceptive pill comprising 30 μg ethinyl oestradiol and 3 mg drospirenone. Drospirenone is a derivative of spirono-lactone and has similar anti-androgenic properties.

It binds to testosterone receptors, thereby producing anti-androgenic effects.

❏ *Progestogenic agents*: Some progestogens have androgenic effects, e.g. norethisterone. So, preparations with non-androgenic progestogens should be chosen, e.g. medroxyprogesterone. However, medroxyprogesterone (Depo-Provera) has to be administered for months to be able to produce effective results. Depo-Provera has also been linked to the loss of bone marrow density. Therefore, its use is contraindicated for adolescents whose bones are still maturing.

❏ *GnRH agonists*: Initial administration of GnRH agonists stimulates the release of FSH and LH. This is followed by the eventual downregulation of pituitary gland, thereby producing a hypogonadotropic-hypogonadic state. These drugs are effective in hirsutism because of the "downregulation" of FSH and LH production. The limiting factors are its high cost, and risks of long-term treatment (e.g. loss of bone mass, etc.). However, they can be used with "add back" therapy, e.g. the oral contraceptives or just oestrogen. For further details related to GnRH agonists, kindly refer to Chapter 12.

Non-hormonal Drugs

❏ *Metformin*: Insulin increases androgen production and reduces SHBG. Metformin reduces insulin levels and counteracts this. Therefore, this is particularly useful in women with PCOS.

❏ *Spironolactone*: This is an aldosterone antagonist having a beneficial effect on hirsutism.

❏ *Finasteride*: 5α-reductase is an enzyme, which converts testosterone into its more active metabolite dihydrotestosterone. The enzyme 5α-reductase has two forms, type 1 and type 2, which are present in different amounts in different tissues. Finasteride is a synthetic chemical that inhibits (type 2) 5α-reductase.

❏ *Flutamide*: This is an anti-androgen that binds to androgen receptors. There are some concerns about its hepatotoxicity.

❏ *Eflornithine*: Polyamines are critical components of hair. A key enzyme involved in their synthesis is ornithine decarboxylase. Eflornithine inhibits ornithine decarboxylase. It is applied as a cream and is effective in reducing hair growth. It may cause side effects such as skin irritation and acne in a small number of patients.

Ovarian Masses

Dermoid Cysts of the Ovary

Dermoid cysts of the ovary are also known as benign cystic teratomas. They are the commonest ovarian tumour in pregnancy. Dermoids are bilateral in 10–20% of cases. These mostly occur in women of reproductive age; so they form the likeliest diagnosis in pregnancy. Dermoids are relatively heavy; so they are particularly likely to undergo torsion. It is estimated that about 1% of these tumours may undergo malignant degeneration in the elderly.

Uterine Retroversion

Normal uterine position is that of anteversion and anteflexion, i.e. the uterine body is bent forward at the utero-cervical junction over the bladder **(Fig. 14.9A)**. Retroversion is a type of uterine displacement in which the uterine body is displaced backwards at the utero-cervical junction **(Fig. 14.9B)**. Retroversion could be either fixed or mobile. Mobile retroversion, which is uncomplicated by pelvic disease is of little clinical significance. Fixed retroversion could be related to conditions such as PID (salpingo-oophoritis), pelvic tumours, chocolate cysts of the ovary and pelvic endometriosis. Diagnosis is mainly established on the basis of findings of pelvic examination. On bimanual examination, a mass is felt in the pouch of Douglas. Since this mass moves with the cervix, it can be considered a part of the uterus. Uterus may be tender to touch. Retroversion is recognised on bimanual pelvic examination when the cervix is found to be directed forwards. With the examining fingers in the posterior vaginal fornix, the body of the uterus can be felt.

If pregnancy occurs in case of retroverted uterus, the uterus nearly always rises up into the abdomen in the normal way at about the 12th week and after delivery, it resumes its retroverted position. The uterus is sometimes discovered to be retroverted in the puerperium or at a
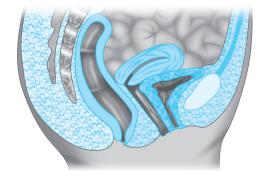
FIG. 14.9A: Uterus in the position of anteversion and anteflexion
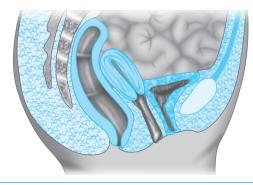
FIG. 14.9B: Retroverted uterus: long axis of the uterus is directed backward

postnatal examination. Most of these cases are merely instances of pre-existing retroversion in which the uterus has returned to its usual position.

Management

In asymptomatic cases of mobile retroversion, no treatment is required. Insertion of a pessary may be required in symptomatic cases, where the uterus is bimanually replaced and a Hodge pessary is inserted inside to keep the uterus in an anteverted position. The Hodge pessary was designed for insertion into the vagina in such a way as to maintain a uterus in the anteverted position once the retroversion had been manually corrected. It is usually retained for 3 months in position and then removed.

Surgical Treatment

Surgical treatment may be required in the cases of fixed retroversion and comprises of the following options:

❑ *Modified Gillam's ventrosuspension*: This is the most commonly used surgical option in which the round ligaments are anchored to the anterior rectus sheath
❑ Plication of the round ligaments
❑ *Baldy-Webster's operation*: This surgery involves shortening of the round ligaments. Round ligaments are anchored to the posterior surface of the uterus by passing them through the anterior and posterior leaves of the broad ligament.

Fibromyomata

Uterine leiomyomas (uterine myomas, fibromyomas or fibroids) are well-circumscribed benign tumours developing from uterine myometrium, most commonly encountered amongst women of reproductive age group (30–44 years). There are three types of fibroids: intramural or interstitial fibroids (which are present within the uterine myometrium), sub-mucosal fibroids (which grow beneath the uterine endometrial lining) and sub-serosal fibroids (which grow beneath the uterine serosa). Of these various types of fibroids, the commonest are the intramural fibroids. Most fibroids are asymptomatic, but the symptoms which they can commonly cause, include bleeding, pressure symptoms (e.g. urinary symptoms, low backache, rectal tenesmus and constipation), anaemia (due to excessive bleeding) and less commonly pain. The pattern of bleeding is usually excessive or prolonged menses (menorrhagia). Other causes of abnormal uterine bleeding must be ruled out by endometrial sampling in these cases.

Pain is a rare symptom, which may occur as a result of degeneration or torsion of the fibromyomata. Severe cramping may be due to uterine contractions as the uterus attempts to pass a sub-mucosal tumour out from the uterine cavity. Fibroids are usually not responsible for causing infertility. Infertility might ensue from distortion of the uterus and damage to the tubes or from consequent infection. Although sub-serosal and interstitial fibromyomatas are commonly associated with pregnancy, sub-mucosal tumours may rarely present with some problems due to sub-fertility.

Management

Various treatment options for a patient with uterine fibroids are described in **Figure 14.10**. Women with asymptomatic uterine fibroids do not require any treatment. Small asymptomatic tumours, however, should be re-examined regularly so that treatment can be immediately administered if the tumour increases in size or symptoms arise. Presently, the main modality of curative treatment in a patient with leiomyoma is surgery and acts as a definitive cure.

Options for surgical treatment include abdominal myomectomy, vaginal myomectomy, endoscopic myomectomy, abdominal hysterectomy and vaginal hysterectomy. Myomectomy of the fibroids should not be undertaken in pregnancy due to the risk of haemorrhage. The exception to this may be presence of symptomatic sub-serous fibroids on a pedicle less than 5 cm thick. If the woman has completed her family and does not wish to preserve her uterus, hysterectomy can be done. Myomectomy is an option for women who desire future pregnancy or wish to preserve their uterus. Various indications for surgical treatment are listed in **Table 14.21**.

Myomectomy

Surgical removal of myomas from the uterine cavity is termed as myomectomy. Although myomectomy allows preservation of the uterus, present evidence indicates a higher risk of blood loss and greater operative time with myomectomy in comparison to hysterectomy. Numerous techniques are used nowadays for performing myomectomy. These include the following: performing a myomectomy through an abdominal incision, vaginal incision, with the help of a laparoscope or a hysteroscope. Though abdominal myomectomy is nowadays uncommonly performed, removal of fibroids, especially hysteroscopically and laparoscopically, has become more popular in recent years. Nowadays, myomectomy is mainly useful for the pedunculated fibroids. A pedunculated sub-mucosal fibroid can be removed through hysteroscopic myomectomy. The

TABLE 14.21	Indications for surgical treatment of fibroids

- Heavy or prolonged bleeding
- Large tumours (14-week pregnancy), even if these are not causing symptoms
- Possible malignant change (for example, growth of the tumour after menopause)
- Retention of urine (acute retention of urine rarely occurs)
- Tumours which obstruct labour
- Tumours which have undergone torsion
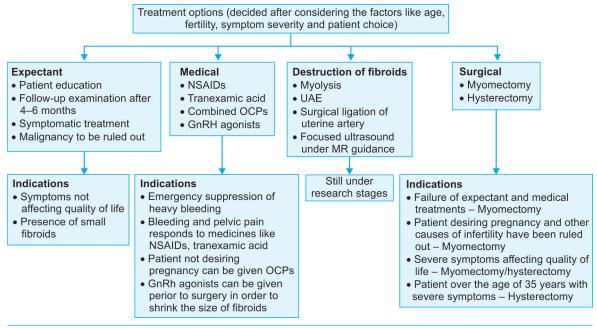
FIG 14.10: Treatment options for a patient diagnosed with fibroid uterus
Abbreviations: NSAIDs, non-steroidal anti-inflammatory drugs; GnRH, gonadotropin releasing hormone; OCPs, oral contraceptive pills; UAE, uterine artery embolisation, MR, magnetic resonance

pedunculated sub-serous type of fibroid, which bulges out from the uterus into the peritoneal cavity can be removed via laparoscopic myomectomy. Indeed, there is always a risk with myomectomy that the bleeding may be as severe as to require a hysterectomy.

Hysterectomy would be more likely to provide relief from symptoms such as bleeding, pain and pressure symptoms. There is not a lot of evidence that myomectomy achieves a lot of symptomatic improvement, especially with regard to infertility and menorrhagia. However, caesarean section is not always necessary after myomectomy. The risk of scar dehiscence at the time of caesarean delivery is greatest if the whole thickness of the myometrium has been involved and the surgery was particularly extensive.

Uterine Artery Embolisation

Uterine artery embolisation (UAE) is a relatively new, novel technique for treatment of uterine fibroids, which was first performed by Ravina, a French gynaecologist in 1995. UAE is a non-hysterectomy surgical technique, which helps in reducing the size of the uterine fibroids by shrinking them, without actually removing them. The procedure involves injection of an embolising agent [gelatin microspheres (trisacryl gelatin) or polyvinyl alcohol] via a cannula inserted through the femoral artery into the internal iliac and the uterine vessels, which helps in blocking both the uterine arteries, thereby cutting off the blood supply to the fibroid. Embolisation is mainly performed to provide symptomatic relief and is effective in a reasonable proportion of cases. However, the procedure is not without hazard because the iliac and uterine vessels have to be cannulated, and there may be infection and bleeding after the procedure. There was concern about pregnancy after the procedure, but several successful pregnancies have been reported to occur.

Pregnancy in Presence of Fibroids

In case of pregnancy with fibroids, the patient should be allowed to go to term, and, if there appears to be a chance that the fibromyoma may be pulled up, labour may be allowed to begin. However, if it becomes evident that normal delivery is impossible, caesarean section should be performed. It is occasionally possible to perform myomectomy at the same time as caesarean section, but it is usually safer to leave this until later because of the risk of haemorrhage. In the case of a patient who is unlikely to become pregnant again, caesarean hysterectomy may be the best treatment.

Degenerative Changes in the Fibromyomatas

Certain degenerative changes can occur in a fibroid, which can cause an interference with capsular circulation. As a result of circulatory disturbances, the tumour becomes painful, tender, softened and enlarged. Some such degenerative changes taking place in the fibroids are described below:

❏ *Atrophy*: Shrinkage of the fibroid can occur as a result of reduced blood supply to the fibroid, usually following menopause.
❏ *Hyaline degeneration*: This is the commonest type of degeneration in which the fibrous tissue cells are

replaced by a homogeneous substance that stains pink with eosin. The bundles of muscle fibres become isolated and die off causing large areas of the tumour to become structureless. Eventually, the liquefaction of hyaline material occurs, leaving behind ragged cavities filled with colourless or bloodstained fluid.

❑ *Calcification*: This type of degeneration may initially occur with the presence of fatty deposits within the leiomyomas. At a later stage in this process, there is deposition of phosphates and carbonates of calcium along the course of blood vessels. Calcification usually begins at the periphery of the fibroid and can be identified with the help of radiography. At a later stage, there may be widespread deposition of calcium throughout the tumour resulting in "womb stone" appearance or a peripheral distribution resulting in an "egg shell" appearance.

❑ Myxomatous/cystic degeneration

❑ *Red/carneous degeneration*: This type of degeneration of uterine fibroid usually develops during pregnancy. It may be associated with constitutional symptoms like malaise, nausea, vomiting, fever and severe abdominal pain. The myoma may become soft and necrotic in the centre and is diffusely stained red or salmon pink in colour. Though the pathogenesis of the condition is not yet clear, it is believed that the purple-red colour of the myoma is probably due to the thrombosis of blood vessels supplying the tumour. The myoma may also develop a peculiar fishy odour due to infection by the coliform organisms. The fibroid outgrows its blood supply and central haemorrhagic breakdown occurs.

Although the patient may develop mild leucocytosis and a raised erythrocyte sedimentation rate (ESR), the condition is essentially an aseptic one. It needs to be differentiated from other conditions including appendicitis, twisted ovarian cyst, accidental haemorrhage, etc. Good history taking, clinical examination and ultrasound examination usually helps in establishing the correct diagnosis. On ultrasound examination, the tumour shows a mixed echo-dense and echo-lucent appearance. Red degeneration occurring during pregnancy must be managed conservatively. The patient must be advised bed rest and prescribed analgesics to relieve the pain. The acute symptoms subside gradually within the course of 3–10 days and pregnancy then proceeds uneventfully.

The major hazard associated with red degeneration is the risk of unnecessary caesarean section (with possible prematurity) due to a mistaken diagnosis of placental abruption. The key of establishing the correct diagnosis is taking a good history and conducting a proper clinical examination. In "red degeneration", there is no bleeding; the area of pain and tenderness is localised to the fibroid; the rest of the uterus is soft; the foetal heart rate is normal. On the other hand, in cases of abruption, bleeding is usually present. There may be an evidence of shock. The uterus is "woody" hard and tender all over; foetal parts are difficult to feel and localise; and foetal heart activity cannot be usually detected.

❑ *Sarcomatous change*: Occurrence of malignant changes in a leiomyoma is an extremely rare occurrence.

Primary Dysmenorrhoea

Dysmenorrhoea has been defined by the ACOG as a gynaecological medical condition characterised by presence of pain during the menstrual phase. The first thing is to reassure the patient and her mother that there is no serious pathology. Dysmenorrhoea can be of two types: primary (spasmodic or the 1st day pain) and secondary (congestive type). Dysmenorrhoea is labelled as primary in the absence of underlying medical disease/pathology. Secondary dysmenorrhoea on the other hand, is associated with an underlying medical disease/pathology. An ultrasound scan is useful for this purpose. The mainstay for treatment of primary dysmenorrhoea is the NSAIDs with or without the oral contraceptive pills. Cervical dilatation used to be popular, but it was ineffective and was associated with the risk of cervical incompetence in subsequent pregnancies. Division of the utero-sacral ligaments has also been tried but has largely been abandoned in the present times. Use of GnRH analogues as the second-line option may prove to be effective in cases not responding to the first-line therapy. However, the GnRH analogues, through abolition of ovarian function may produce bone loss, so they do not serve as a very practical option.

Pre-menstrual Syndrome
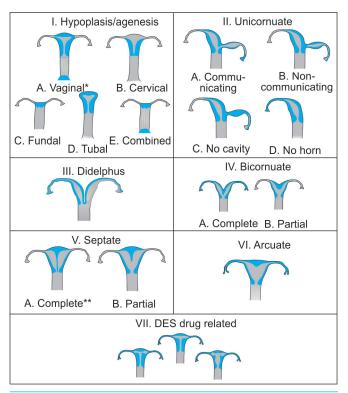
Pre-menstrual syndrome (PMS) or pre-menstrual tension includes a combination of physical, psychological and emotional symptoms, which the women experience for a few days (usually 7–10 days) preceding menstruation. Some of the symptoms, which are commonly observed, include the following: abdominal bloating, breast tenderness, headache, sleeplessness, fatigue, emotional liability and emotional outbursts, mood swings, depression, irritability, lassitude, insomnia, fluid retention, increase in appetite, craving for sweet foods, intestinal distention, colonic spasm, spasmodic dysmenorrhoea, etc.

Uterine Malformations

Congenital uterine anomalies may arise from malformations at any step of the Müllerian developmental process. The classification of Müllerian abnormalities as proposed by the American Society for Reproductive Medicine is described in **Figure 14.11** and **Table 14.22**.

Bicornuate Uterus

Bicornuate uterus occurs due to abnormality of the fusion process in the upper parts of Müllerian ducts. As a result,

FIG. 14.11: Classification of the uterine anomalies
* Uterus may be normal or take a variety of abnormal forms; ** May have two distinct cervices
Source: The American Fertility Society classifications of adnexal adhesions, distal tubal occlusion, tubal occlusion secondary to tubal ligation, tubal pregnancies, Müllerian anomalies and intrauterine adhesions. Fertil Steril. 1988;49(6):944-55.

TABLE 14.22	American Society for Reproductive Medicine classification of congenital uterine anomalies	
Classification	**Clinical finding**	**Description**
I	Segmental or complete Müllerian agenesis or hypoplasia	• Vaginal • Cervical • Fundal • Tubal • Combined
II	Unicornuate uterus with or without a rudimentary horn	• With a communicating rudimentary horn • With a non-communicating rudimentary horn
III	Didelphys uterus	Characterised by complete or partial duplication of the vagina, cervix and uterus
IV	Complete or partial bicornuate uterus	• Complete • Partial
V	Complete or partial septate uterus	• Complete • Partial
VI	Arcuate uterus	A small septate indentation is present at the fundus
VII	Diethylstilbestrol-related abnormalities	Presence of a T-shaped uterine cavity with or without dilated horns

there is a single cervical canal in the lower part, but the upper part is bifurcated, having two horns. Significant uterine malformation is said to occur in less than 1% of women, but in more than 10% of those with recurrent miscarriage. It is still a matter of debate whether uterine anomalies have any role in the pathogenesis of recurrent miscarriage. Nevertheless, it is not thought to be the major cause. Surgery can be done but would only be considered in extreme cases such as recurrent second trimester miscarriage.

The genital and urinary tracts develop hand in hand; abnormality in one is associated with an increased risk of malformation in the other. Therefore, bicornuate uterus may be associated with simultaneous presence of urinary tract abnormalities. Bicornuate uterus is often associated with premature labour, and a persistent abnormal lie, usually breech, but possibly transverse.

Uterus Didelphys

Incomplete fusion of the Müllerian or paramesonephric ducts results in the commonest types of uterine malformation. Complete failure of the fusion of Müllerian ducts results in uterus didelphys, which is an extremely rare condition, characterised by the following:
❑ Double vagina
❑ Double cervix
❑ Entirely double uterus, that is, two single-horned uteruses

Endometriosis

Endometriosis is characterised by occurrence of endometrial stroma and glands outside the uterus in the pelvic cavity, including all the reproductive organs as well as on the bladder, bowel, intestines, colon, appendix and rectum **(Fig. 14.12)**. The ectopic endometrial tissues, both the glands and the stroma, are capable of responding to cyclical hormonal stimulation and have the tendency to invade the normal surrounding tissues. The pathogenesis of endometriosis is yet not clear. Some likely mechanisms for its pathogenesis are as follows:
❑ *Retrograde menstruation*: Retrograde menstrual flux can be considered as an essential element in the pathogenesis of endometriosis.
❑ Theory of coelomic metaplasia: Peritoneal epithelium can get "transformed" into endometrial tissue under the influence of some unknown stimulus.
❑ Metastatic theory of lymphatic and vascular spread: Metastatic deposition of endometrial tissues at ectopic sites can occur via lymphatic and vascular routes.
❑ Immunological defects and genetic factors.

Classically, endometriosis causes dysmenorrhoea, particularly pain that starts days before the onset of bleeding. Cramping pain that starts with the bleeding is typical of spasmodic dysmenorrhoea. Deep dyspareunia is also a typical feature of endometriosis. Other features can be chocolate cysts of the ovaries (due to collection of old blood/clotted blood) and fixed retroversion of the uterus. Rarely, bowel symptoms may be present.
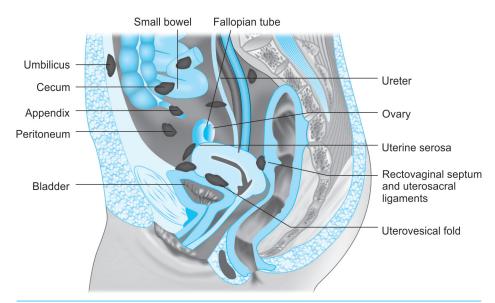
FIG. 14.12: Common sites of endometriotic lesions

TABLE 14.23	Diagnostic features of the commonest causes of vaginitis		
Basis of diagnosis	**Bacterial vaginosis**	**Vulvovaginal candidiasis**	**Trichomoniasis**
Signs and symptoms	Thin, greyish to off-white coloured discharge; unpleasant "fishy" odour especially increasing after sexual intercourse. The discharge is usually homogeneous and adheres to vaginal walls.	Thick, white (curd-like) discharge, with no odour	Copious, malodorous, yellow-green (or discoloured) discharge, pruritus, and vaginal irritation, dysuria, or asymptomatic in many cases
Physical examination	Normal appearance of vaginal tissues; greyish white-coloured discharge may be adherent to the vaginal walls.	Vulvar and vaginal erythema, oedema and fissures; thick white discharge that adheres to the vaginal walls	Vulvar and vaginal oedema and erythema, "strawberry" cervix in up to 25% of affected women; frothy purulent discharge
Vaginal pH	Elevated (> 4.5)	Normal	Elevated (> 4.5)
Microscopic examination of wet-mount and KOH preparations of the vaginal discharge	"Clue cells" (vaginal epithelial cells coated with coccobacilli), few lactobacilli, occasional motile, curved rods, belonging to preparations of *Mobiluncus* species	Pseudo-hyphae, mycelial tangles or budding yeast cells	Motile trichomonads, many polymorphonuclear cells
"Whiff" test (Normal = no odour)	Positive	Negative	Can be positive
Additional tests	Amsel's criteria is positive in nearly 90% of affected women with bacterial vaginosis	KOH microscopy, gram stain, culture	*DNA probe tests*: Sensitivity of 90% and specificity of 99.8% *Culture*: Sensitivity of 98% and specificity of 100%.

Vaginal Discharge

Diagnostic features of the commonest causes of vaginitis are described in **Table 14.23**.

Bacterial Vaginosis

Bacterial vaginosis (BV) is one of the most important causes of vulvovaginitis. Initially, this was termed as bacterial vaginitis. However, later it was discovered that the condition was primarily caused due to the alteration of normal vaginal flora, rather than due to any specific infection. Due to the absence of inflammation, the term "vaginosis" rather than "vaginitis" is now being preferred. The normal vaginal epithelium contains numerous bacteria called *L. acidophilus*. These bacteria release hydrogen peroxide, which is toxic to other aerobic and anaerobic bacteria. BV typically is associated with a reduction in

the number of the normal hydrogen peroxide-producing *Lactobacilli* in the vagina. The resultant change in pH allows proliferation of organisms that are normally suppressed such as *Haemophilus vaginalis, Gardnerella mobiluncus, Mycoplasma hominis, Gardnerella vaginalis, Peptostreptococcus* species, etc. These organisms may produce metabolic byproducts, such as amines, that further increase the vaginal pH and cause exfoliation of vaginal epithelial cells. These amines are also responsible for the characteristic malodorous discharge in BV. Bacterial vaginosis is not dangerous, but it can cause disturbing symptoms. Certain factors have been identified that increase the chances of developing BV. These include multiple or new sexual partners, vaginal douching and cigarette smoking. However, the role of sexual activity in the development of the condition is not fully understood and BV can still develop in women who have not had sexual intercourse. Risk factors include Afro-Caribbean race, use of douches, smoking, and presence of an IUD. It is not regarded as sexually transmitted and can occur in those who are not sexually active. It is linked to an increased risk of preterm premature rupture of membranes (PPROM) and preterm labour. Diagnosis can be clinical or using laboratory tests. Amsel's diagnostic criteria for BV are as follows:

❑ Thin, homogeneous discharge
❑ Positive "Whiff test"
❑ Presence of "clue cells" on microscopic examination
❑ Vaginal pH greater than 4.5

Treatment

❑ *Metronidazole*: A 7-day course of oral metronidazole, 400 mg TDS or vaginal metronidazole gel (metrogel) is an effective treatment.
❑ *Tinidazole*: Tinidazole is an antibiotic that appears to have fewer side effects than metronidazole and is also effective in treating BV.
❑ *Ornidazole*: Ornidazole, 500 mg vaginal tablet daily for 7 days, is another effective option.
❑ *Ampicillin*: Ampicillin 500 mg TDS or cephalosporin 500 mg BID for 7 days is also effective.
❑ *Tetracyclines*: Tetracycline 500 mg, four times a day or doxycycline 100 mg twice daily for 7 days may also be used.
❑ *Lincosamides*: Vaginal clindamycin cream 2% (cleocin) or oral clindamycin 300 mg daily for 7 days is also effective.
❑ *Clindamycin*: The treatment of choice in pregnancy for BV is clindamycin, which has been shown to reduce the risk of pre-term delivery. Metronidazole is also an option.

Monilial Infection

Monilial infection is most commonly caused by the organisms belonging to the genus *Candida* (Monilia). *Candida albicans* is the infecting organism in more than 80% of women with "thrush", with other forms of *Candida* causing the rest. It is a fungus and mycelia may be seen on microscopy. It is not a sexually transmitted disease (STD). Maturity onset diabetes may present with monilial infection, often of the vulva and vagina. But, the juvenile variety will usually present with the classical clinical features such as polyuria, polydypsia, etc. It is said that it can be found in the vagina in more than 25% of asymptomatic women. The drug of choice for treatment of candidal infections is fluconazole. Fluconazole (Diflucan) is available to the public in a 150 mg single-dose preparation. It is not known to be a teratogen but is avoided in pregnancy, as its safety has not yet been established.

The clinician would only remove an IUD if there was a significant risk of ascending infection or signs of it, e.g. pelvic tenderness. The common infections, due to *Candida* and *Trichomonas*, are not grounds. *Actinomyces* is a rare cause of ascending infection. It is not uncommonly reported on routine cervical smears in asymptomatic patients. There is an increased risk of ascending infection in this situation, which has led to advice that the IUD be removed.

Trichomonas vaginalis is a flagellate protozoan and is transmitted by sexual intercourse.

Sexually Transmitted Infections

Various causes of genital ulcers are listed in **Table 14.24**.

Gonorrhoea

Gonorrhoea is a STD, which is derived from the Greek words gonos (seed) and rhoia (flow) implying "flow of seeds" and is caused by the bacterium *Neisseria gonorrhoeae*. The bacterium causing gonorrhoea is a Gram-negative intracellular diplococcus. It is detected on cervical and urethral swabs. The disease is characterised by adhesion of the gonococci to the surface of urethra or other mucosal surfaces. Gonorrhoea spreads through contact with the penis, vagina, mouth or anus. Gonorrhoea can also spread from mother to baby at the time of delivery.

Symptoms

❑ The commonest clinical presentation of the disease in men is acute urethritis resulting in dysuria and a purulent penile discharge. The majority of gonococcal-infected men develop urethritis, dysuria and urethral discharge.

TABLE 14.24	Causes for genital ulcers
Painless genital ulcers	**Painful genital ulcers**
• Circinate balanitis • Granuloma inguinale • Primary syphilis	• Herpes simplex/zoster and genital herpes • Behçet's disease • Reiter's syndrome • Chancroid • Gonococcal disease

❑ Symptomatic women commonly experience vaginal discharge, dysuria and abdominal pain.
❑ The infection, if untreated, may extend to Bartholin's glands, endometrium and Fallopian tubes. The gonococci can typically ascend to the fallopian tubes at the time of menstruation or after instrumentation (for MTP) giving rise to acute salpingitis.
❑ Systemic manifestations, peri-hepatitis and septicaemia can all be caused by gonococcal infection.

Investigations

The swabs are usually taken from the endocervix and urethra. In some circumstances, throat and rectal swabs should be considered. The bacterium is difficult to grow and successful culture requires rapid transport to the laboratory and special culture media (e.g. chocolate agar). The investigations performed on these samples include:
❑ Culture and sensitivity
❑ DNA probes.

Medical Management

Treatment comprises of using the following antibiotics: Ceftriaxone 125 mg IM, cefixime 400 mg, ciprofloxacin 500 mg PO or ofloxacin 400 mg PO. This is followed by:
Doxycycline 100 mg BID × 7 days or azithromycin 1 g PO (single dose).

Complications

The infection may extend along the urethra to the prostate, seminal vesicles and epididymis, resulting in complications such as epididymitis, prostatitis, peri-urethral abscesses and chronic urethritis. The infection may spread to the peri-urethral tissues, resulting in formation of abscesses and multiple discharging sinuses (watercan perineum). Acute salpingitis may be followed by PID. This may be associated with a high probability of sterility if not treated adequately. Peritoneal spread occasionally occurs and may produce a peri-hepatic inflammation, resulting in Fitz-Hugh-Curtis syndrome. Gonococcal infection can result in neonatal eye infection and can cause substantial damage if not treated promptly. In America, the neonate is given prophylactic antibiotic eye drops to prevent such infection. Gonococcal ophthalmia neonatorum can lead to severe conjunctivitis, keratitis and blindness if not promptly treated.

Fitz-Hugh-Curtis Syndrome

It is related to peri-hepatic infection with formation of fine adhesions known as the "violin string" secondary to pelvic infection. The clinical presentation is similar to that of cholecystitis. Previously, the commonest infecting organism was the gonococcus, but nowadays, the commonest organism is *Chlamydia*. Most patients will have the following clinical features:
❑ Right upper quadrant pain
❑ Evidence of chlamydial infection

❑ *Evidence of inflammation*: Raised ESR, C-reactive protein and white cell count.

Apparently, it is also a common finding in pelvic tuberculosis. As it is secondary to pelvic infection, it is often associated with impaired fertility. Treatment is with appropriate antibiotic therapy. A similar phenomenon can also occur around the appendix.

Bartholin's Abscess

The Bartholin's glands are situated at the level of the introitus within the labia majora. The cause for development of cysts and abscesses in the Bartholin's glands largely remains unknown; it could be related to the blockage of ducts, but the explanation of this remains obscure. The infection may be due to the gonococcus. Treatment is by "marsupialisation", in which the cyst is effectively de-roofed to allow drainage.

Genital Warts

Genital warts are caused by different strains of human papilloma virus, especially HPV types 6, 11, 16 and 18.

Additional cervical screening for detecting genital warts is not justified. The incidence of genital warts is increasing at present. The mean incubation period is about 3 months. However, it is quite variable and may range from 3 weeks to 8 months. There is debate about the role of Caesarean section for patients with active herpes, but not for patients with warts.

Podophyllin in spirit is the standard treatment in cases of genital warts, but can be toxic in pregnancy; so, it is not to be used. If podophyllin cannot be used or fails to produce positive results, the warts can be frozen or treated with diathermy.

Genital Herpes Infection

Genital herpes is one of the commonest sexually transmitted diseases worldwide and is a viral infection caused by the herpes simplex virus (most commonly HSV-II), which is transmitted through sexual contact. Genital herpes spreads only by direct person-to-person contact. The virus enters through the mucous membrane of the genital tract via microscopic tears. From there, the virus travels to the nerve roots near the spinal cord and settles down permanently.

Symptoms

The primary infection may be associated with constitutional symptoms like fever, malaise, vulval paraesthesia, itching or tingling sensation on the vulva and vagina followed by redness of skin. Eventually, there is formation of blisters and vesicles on the vulva, vagina, cervix, perianal area or inner thigh, which ultimately develop into shallow and painful ulcers within a period of 2–6 weeks. They are frequently accompanied by itching and mucoid vaginal discharge. Swollen and tender lymph nodes may occur in the groin region.

Investigations

Diagnosis is usually based on clinical examination. Genital herpes is suspected when multiple painful blisters are present on the external genitalia. The various investigations which can be performed are as follows:

- *Cytological tests*: The blister fluid may be sent to the laboratory for culturing the virus. However, it is associated with a high false negative rate of nearly 50%.
- *Immunological tests*: These tests are specific for HSV-I or HSV-II and may be able to demonstrate that a person has been infected at some point in time with the virus.
- *Other diagnostic tests*: These include tests such as polymerase chain reaction and rapid fluorescent antibody screening tests.
- *Biopsy*: The Tzanck smear is a rapid, fairly sensitive and inexpensive method for diagnosing HSV infection. Smears are preferably prepared from the base of the lesions and stained with 1% aqueous solution of toluidine blue "O" for 15 seconds. Positive smear is indicated by the presence of multi-nucleated giant cells with faceted nuclei and homogeneously stained "ground glass" chromatin (Tzanck cells).

Medical Management

There is still no curative medicine available for genital herpes, and the antiviral drugs only help in reducing the severity of symptoms and duration of outbreaks.

- Oral antiviral medications, such as acyclovir, (Zovirax), famciclovir (Famvir) or valacyclovir (Valtrex), which prevent the multiplication of the virus, are commonly used. For the treatment of primary outbreaks, oral acyclovir is prescribed in the dosage of 200 mg five times a day for 5 days.
- Local application of acyclovir provides local relief and accelerates the process of healing.
- In severe cases, acyclovir can be administered intravenously in the dosage of 5 mg/kg body weight every 8 hourly for 5 days.
- The couple is advised to abstain from intercourse starting right from time of experiencing prodromal symptoms until total re-epithelialisation of the lesions occurs.
- Pregnant women with active herpetic lesion must be preferably delivered by caesarean section.

Herpes can be spread from one part of the body to another during an outbreak. Thorough handwashing is a must during outbreaks in order to prevent the spread of infection. Couples who want to minimise the risk of transmission should always use condoms if a partner is infected. Such couples must be instructed to avoid all kinds of sexual activity, including kissing, during an outbreak of herpes.

Urinary Tract Infections

Escherichia coli accounts for 80% of UTIs. *Enterobacter* and *Klebsiella* spp. tend to be hospital-acquired infections. *Pseudomonas* and *Candida* spp. are opportunistic infections, which affect immunosuppressed patients. *Proteus* spp. are often associated with urinary calculi. *Chlamydia* rarely causes UTIs but is associated with PID.

Asymptomatic Bacteruria

Asymptomatic bacteruria (ASB) occurs in about 5% of women, irrespective of whether they are pregnant or not. The infecting organism is usually a coliform. A significant bacterial count is 100,000 organisms per mL, i.e. 10^5. Many hospitals screen for the infection via microscopy and culture. However, nowadays, use of dipsticks for screening is increasingly becoming popular. Some research evidence suggests that dipsticks are less effective than formal microscopy and culture for initial screening purposes. Microscopy and culture is still useful for the patient admitted later in pregnancy with UTI, premature labour, etc. The importance of ASB in pregnancy is related to its predisposition for ascending infection. This is probably due to urinary stasis from dilatation of ureters due to progesterone. Another factor could be related to the compression of ureters at the pelvic brim by the gravid uterus. Dextrorotation of the uterus may cause more compression on the right ureter and a tendency for the condition to occur more commonly on the right side. ASB has been linked with an increased risk for preterm delivery; this may be due to the increased risk of pyelonephritis. Treatment involves nursing measures to lower the temperature, rehydration and antibiotic therapy.

Non-gonococcal Urethritis

Mostly non-gonococcal urethritis (NGU) is due to *Chlamydia*, and more rarely due to *Mycoplasma*, *Ureaplasma*, *Trichomonas* or meningococcal disease. These may occur together.

Non-gonococcal urethritis is also part of Reiter's syndrome (reactive arthritis, conjunctivitis, urethritis). This can be caused by gonococcus and *Campylobacter*.

Complications of NGU: Salpingitis, peri-hepatitis, conjunctivitis, sterility.

Treatment of NGU: Doxycycline or erythromycin.

Pelvic Abscess

Pelvic abscess is the commonest variety of intra-peritoneal abscess. Pus can track down the peritoneal cavity to form a pelvic abscess. The abscess can irritate the bladder causing urinary frequency/nocturia. If an abscess drains spontaneously via the rectum, it rarely requires any further treatment. Ultrasound imaging requires a full urinary bladder to identify the pelvic organs and any abscesses present.

Choose the Single Best Answer (SBA)

Q 1. **Which of the following statement regarding endometriosis is correct?**
A. Does not usually affect the ovaries
B. Effective drug therapy significantly improves potential fertility
C. May be a cause of fixed retroversion of the uterus
D. Classically causes superficial dyspareunia
E. tubal damage is common and occurs early in the progression of the disease

Q 2. **Which of the following is true regarding bacterial vaginosis?**
A. Is associated with a purulent, green discharge
B. Is associated with pre-term labour
C. Is associated with intense vaginitis
D. Is associated with a cheesy smell
E. Is best treated with tetracycline

Q 3. **Which of the following is true regarding Fitz-Hugh-Curtis syndrome?**
A. Presents with the features of appendicitis
B. Is frequently due to tuberculosis
C. Is associated with sub-fertility
D. Is best treated by laparotomy
E. Is due to allergy to violin strings

Q 4. **Which of the following is not true regarding syphilis infection?**
A. Syphilitic chancre is a firm painless nodule
B. Chancre usually appears on the genital region 2–4 weeks after infection.
C. The Wasserman reaction is a non-specific test.
D. Aortitis is a feature of secondary syphilis.
E. Is caused by a spirochaete.

Q 5. **Which of the following is true regarding Bartholin's abscess?**
A. Is located on the cervix uteri
B. May be associated with gonococcal infection.
C. Results from poor repair of tears sustained in childbirth.
D. Is normally treated by excision of the gland.
E. Is usually bilateral.

Q 6. **Which of the following is true concerning genital herpes infection?**
A. Can be responsible for pre-term delivery
B. Is always symptomatic
C. Tender inguinal lymphadenopathy frequently occurs
D. First attack is milder than subsequent attacks
E. Has an incubation period of 2 months

Q 7. **Which of the following organisms is not a common cause of urinary tract infection?**
A. *Candida*
B. *Chlamydia*
C. *Enterobacter*
D. *Klebsiella*
E. *Proteus*

Q 8. **Which of the following is true regarding asymptomatic bacteriuria (ASB)?**
A. Should be investigated postpartum by IVP
B. Ascending infection occurs in about 30% of pregnant women with ASB
C. A significant count is more than 10^6 organisms per mL of urine
D. Is usually due to chlamydia
E. Screening should be by microscopy and culture of a "clean catch" urine specimen, not dipsticks

Q 9. **In cases of non-gonococcal urethritis (NGU), which of the following statement is correct?**
A. Association with septic arthritis is common
B. *Chlamydia trachomatis* is the commonest organism
C. Chronic conjunctivitis is not a recognised sequel
D. Cystitis is typical
E. It is usually treated with Septrin

Q 10. **Which of the following is not correct concerning pelvic abscess?**
A. Is a common cause of intra-abdominal abscess
B. Can be associated with a perforated peptic ulcer
C. Can only be identified on CT if there is a full bladder
D. Commonly present with nocturia
E. Frequently drain spontaneously into the rectum

Q 11. **Which of the following statement is true regarding monilial infection?**
A. Is due to a flagellate organism.
B. Responds to metronidazole.
C. Is a common presenting feature in juvenile diabetes.
D. The organisms are commonly present in asymptomatic women.
E. Fluconazole is of proven safety in pregnancy.

Q 12. **Which of the following is not true concerning bacterial vaginosis?**
A. Can be diagnosed by the finding of clue cells on microscopy
B. Cannot be detected by Gram staining
C. Causes a rise in the pH of vaginal secretions
D. Is often asymptomatic
E. Should be treated in pregnancy with clindamycin to help prevent late miscarriage and pre-term birth

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1. C   2. B   3. C   4. D   5. B   6. C   7. B   8. B   9. B   10. C   11. D   12. B

Q 13. Which of the following is not true regarding genital warts?

A. They are always sexually transmitted

B. Additional cervical screening is not justified, provided that the woman has had a screening test within the previous 3–5 years

C. Podophyllin paint cannot be used with safety during pregnancy

D. Condom usage with regular sex partners has not been shown to affect the treatment outcome

E. All treatments have significant failure and relapse rates

Q 14. Gonococcus can be found in which of the following tissues?

A. Anus

B. Endocervix

C. Endometrium

D. Epididymis

E. All the above

Q 15. Which of the following viruses cannot be sexually transmitted?

A. Echovirus

B. Hepatitis B

C. Herpes simplex virus

D. Papovavirus

E. None of the above

Q 16. Which of the following is true regarding bicornuate uterus?

A. Is the commonest cause of unstable lie

B. Should be treated surgically if pregnancy has resulted in premature delivery

C. Is associated with premature labour

D. Is a proven cause of recurrent miscarriage

E. is associated with placental abruption

Q 17. A 16-year-old girl has primary dysmenorrhoea. Which is the most suitable treatment in this case?

A. D&C

B. Paracervical block

C. Non-steroidal anti-inflammatory drugs

D. GnRH analogue

E. Laser ablation of the endometrium

Q 18. Which of the following procedures does not help correct retroversion of the uterus?

A. Hodge pessary

B. A laparoscopic procedure

C. A sling operation

D. Pelvic floor repair

E. Gilliam's operation

Q 19. Which of the following is true concerning intermenstrual bleeding (IMB)?

A. IMB occurs in about 10% of normal menstrual cycles.

B. Laparoscopy should be included as part of the investigation.

C. A luteal phase progesterone is essential.

D. IMB may be associated with ovulation.

E. IMB is a feature of cervical intra-epithelial neoplasia.

Q 20. Which of the following is not true regarding submucous uterine fibroids?

A. Can become infected

B. Frequently cause infertility

C. May become polypoidal

D. May protrude through the cervix

E. Often present with menorrhagia

Q 21. Which of the following is true regarding uterine fibroids?

A. Undergo malignant degeneration in 5% of cases

B. If present in pregnancy, myomectomy should be performed at 14 weeks to prevent "red degeneration" later in pregnancy

C. Can be effectively treated with an LH-RH analogue

D. Commonly co-exist with endometriosis

E. Are a common cause of acute retention of urine

Q 22. Which of the following symptoms is characteristically associated with uterine fibromyomata?

A. Abdominal pain

B. Dysmenorrhoea

C. Dyspareunia

D. Menorrhagia

E. Vaginal discharge

Q 23. Which of the following is true regarding myomectomy?

A. Is an underused alternative to hysterectomy

B. Is useful in the management of infertility

C. Is useful in managing menorrhagia

D. Makes Caesarean section obligatory in subsequent pregnancy

E. None of the above

Q 24. Which of the following is true regarding colposuspension for treatment of uterine retroversion?

A. Effectively treats dyspareunia

B. Effectively treats frequency of micuturition

C. Is an appropriate treatment for enterocoele

D. Is associated with a need to treat rectocoele

E. Is usually performed vaginally

Q 25. Which of the following statement is not correct regarding the mobile retroversion of the uterus?

A. It is suspected when the cervix points anteriorly on speculum examination

B. Is usually asymptomatic

C. Occurs at 20 weeks gestation

D. May occur in 20% of women

E. Occurs in the puerperium

13. A   14. E   15. A   16. C   17. C   18. D   19. D   20. B   21. D   22. D   23. E   24. D   25. C

Q 26. Which of the following changes does not commonly occur in uterine leiomyomata?

A. Atrophy
B. Calcification
C. Hyaline degeneration
D. Squamous metaplasia
E. Sarcomatous change

Q 27. Which of the following is not correct regarding the masses of ovarian origin?

A. Include benign teratomas
B. Those of germ cell origin may secrete hormones
C. Are always malignant in the presence of ascites
D. May be confused with developmental abnormalities of the renal tract
E. Careful surgical staging is essential to determine the appropriate subsequent management

Q 28. Which of the following is true regarding central parenteral nutrition?

A. Does not cause derangement of liver function tests
B. Is a hypo-osmolar solution
C. Is not associated with any metabolic disturbance
D. Typically contains about 250 g glucose
E. Typically contains 14–16 g nitrogen as D-amino acids

Q 29. Which of the following is true regarding hirsutism?

A. Is synonymous with virilisation
B. Is pathological in most cases
C. Is a side effect of cyproterone
D. Androgens in women are produced solely in the adrenal
E. May be due to an ovarian tumour

Q 30. Dyspareunia may result from which of the following?

A. Adenomyosis
B. The climacteric
C. Superficial vulvovaginitis
D. All the above
E. None of the above

Q 31. Which of the following is suggestive of genuine stress incontinence (GSI)?

A. Constant wetness
B. Prolapse
C. Dysuria
D. Haematuria
E. Passage of large amounts of urine

Q 32. Incontinence of urine in the female is investigated by which of the following tests?

A. Cystometry
B. Intravenous urography
C. Urodynamic investigations
D. All the above
E. None of the above

Q 33. Which of the following is true regarding urge incontinence in the female?

A. Is improved greatly by an anterior repair procedure
B. Is improved by bladder drill and re-education
C. Is worse during the night than at day
D. Results in the daily passage of larger volumes of urine than normal
E. Urodynamic studies in upper motor neuronal diseases (for example, multiple sclerosis) show an increased bladder capacity

Q 34. Which of the following is acceptable surgical operation for the treatment of stress incontinence?

A. Anterior colporrhaphy
B. Colposuspension
C. Marshall-Marchetti-Kranz operation
D. All the above
E. None of the above

Q 35. Which of the following is true regarding colposuspension?

A. Is normally done as a vaginal procedure
B. Is effective in relieving urge incontinence.
C. Is effective in relieving dyspareunia
D. Is associated with an increased incidence of posterior vaginal wall prolapse
E. cannot be done via the laparoscope

Q 36. Treatment with a ring pessary for vaginal prolapse is generally indicated in which of the following patients?

A. As the primary therapy when the patient is over 75-years-old who is unfit for surgery
B. Who refuse operation
C. Wishing to become pregnant
D. Patient awaiting surgery
E. All the above

Q 37. Which of the following is true regarding male infertility?

A. Is the primary cause in 10% of couples who fail to conceive
B. Always occurs after vasectomy
C. May be due to genital tract infection
D. Normal semen analysis involves a count > 20 million per ejaculate
E. Azoospermia can be treated with ICSI.

Q 38. Which of the following is true regarding a normal semen specimen?

A. Contain 10 million white blood cells per ml
B. Have a sperm count of more than 20 million/mL
C. Have a volume of more than 5 mL
D. Have at least 80% motility
E. Liquefy within 3 minutes

Q 39. Which of the following does not cause secondary amenorrhoea?

A. Asherman's syndrome
B. Virilising ovarian tumours
C. Endometriosis
D. Hyperprolactinaemia
E. Congenital hypothroidism

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26. D   27. C   28. D   29. E   30. D   31. B   32. D   33. B   34. D   35. D   36. E   37. C   38. B   39. C

Q 40. A 30-year-old woman recently had amenorrhoea and galactorrhoea. Which of the following investigation is not required?

A. Prolactin
B. Oestradiol
C. Luteinising hormone (LH) and follicle stimulating hormone (FSH)
D. Pregnancy test
E. X-ray skull

Q 41. Which of the following statements concerning the hyperstimulation syndrome associated with the use of ovulation inducing agents is correct?

A. It occurs with clomiphene
B. It may be avoided by withholding the mid cycle injection of HCG in the presence of high oestrogen levels
C. Patients with polycystic ovaries before treatment are more likely to develop the syndrome
D. The incidence can be reduced by ultrasonic examination
E. All the above

Q 42. Which of the following regarding crytomenorrhoea is correct?

A. Is associated with the failure of development of the mesonephric system
B. May be associated with Turner's syndrome
C. There is failure of development of secondary sexual characteristics
D. May be associated with acute retention of urine
E. May have a karyotype 47 XXX

Q 43. Which of the following concerning post-coital contraception is correct?

A. The progesterone-only pill can be used.
B. Oral methods should be administered within 24 hours.
C. It is available only through prescription of the GP.
D. The intrauterine device has a role.
E. Follow-up of individuals who have used post-coital contraception is not worthwhile.

Q 44. Use of the combined oral contraceptive pills is associated with an increased risk of which of the following?

A. Breast carcinoma
B. Endometrial carcinoma
C. Ovarian carcinoma
D. None of the above
E. All the above

Q 45. Which of the following is a typical adverse effect of combined oral contraceptive preparations?

A. Breast tenderness
B. Hyperprolactinaemia
C. Loss of libido
D. All the above
E. None of the above

Q 46. A 39-year-old smoker with two healthy children asks for contraceptive advice. In these circumstances, which of the following contraceptive advice must not be given?

A. A progesterone-only oral contraceptive
B. An IUD
C. Barrier methods
D. Sterilisation
E. Combined oral contraceptive pills

Q 47. Which of the following statement regarding sterilisation is not correct?

A. In men has a failure rate of about 1 in 300 cases
B. Prevents pregnancy in a similar order of magnitude to the Mirena levonorgestrel intrauterine system
C. When it fails, it can be associated with an ectopic pregnancy
D. In women cause no change in the volume of menstrual bleeding loss
E. In women can be successfully reversed if clips were used for the original operation in approximately 80% of cases.

Q 48. Which of the following statement is true concerning copper-containing IUDs?

A. Do not cause menorrhagia
B. Have a higher incidence of actinomycosis colonisation than plastic devices
C. Have not been implicated as a cause of fatal infection in pregnancies
D. Cause a relative increase in ectopic pregnancies
E. Should be changed every year

Q 49. Which of the following methods is used to locate a lost IUD?

A. Ultrasound
B. MRI
C. Hysteroscopy
D. Colpotomy
E. None of the above

Q 50. Which of the following statement is not correct regarding the progesterone-only pill?

A. Is a recognised cause of secondary amenorrhoea
B. Can be safely given to lactating women and before recommencing menstruation
C. Should not be prescribed to an individual with a history of a deep venous thrombosis
D. Acts mainly by thickening the cervical mucus
E. Decreases tubal motility.

Q 51. Progestogen-only pills work as contraceptive agent by which of the following mechanism?

A. Acting as a spermicide
B. Altering the cervical mucus
C. Producing endometrial hyperplasia
D. Reducing libido
E. Suppressing ovulation in all cases

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40. E   41. E   42. D   43. D   44. A   45. A   46. E   47. E   48. D   49. A   50. C   51. B

Q 52. Which of the following is true regarding progestogen-only contraception?
A. Are contraindicated in mild hypertension
B. Cannot be taken during the period of lactation
C. Causes HDL levels to rise
D. Has a lesser effect on hepatic function than the combined oral contraceptive pill
E. Produces headaches more commonly than the combined oral contraceptive

Q 53. Which of the following is true regarding injectable progestogens used for contraceptive purposes?
A. Carry a risk of venous thrombosis
B. Cause amenorrhoea in approximately 30% of patients
C. Cause hypertension
D. Can cause irregular vaginal bleeding
E. They have an effect on blood coagulation

Q 54. Which of the following instruction is appropriate when advising on the use of the diaphragm?
A. Always use a spermicide.
B. Sterilise the diaphragm prior to insertion.
C. The diaphragm cannot be used at the same time as the sheath.
D. Refitting the diaphragm is not required after childbirth.
E. The diaphragm must be immediately removed following intercourse

Q 55. Regarding condoms, which of the following statement is correct?
A. Has the highest failure rate when used alone
B. Is the most effective contraceptive measure
C. Should be put on just before ejaculation
D. Can reduce the transmission of sexually transmitted diseases
E. Should be used in conjunction with spermicides

Q 56. Which of the following is a recognised risk associated with the use of combined oral contraceptive (COC) pills?
A. Increased incidence of endometrial carcinoma
B. Pelvic inflammatory disease
C. Benign ovarian cysts
D. Hypertension
E. Increased risk of ovarian carcinoma.

Q 57. Which of the following statement regarding contraception is correct?
A. There is an increased incidence of ovarian cancer in the users of the combined oral contraceptive pill.
B. Conversion of cholesterol to pregnenolone is the rate-limiting step in the production of sex steroid hormones.
C. The combined oral contraceptive pill contains between 10 to 100 μg of ethinyl oestradiol.
D. Progesterones have a major role in the treatment of threatened abortion.
E. Contraceptive implants (e.g. Implanon) provide up to 5 years of continuous contraceptive efficacy.

Q 58. Which of the following statement regarding contraceptive use is correct?
A. Oral testosterone is effective in the treatment of male hypogonadism.
B. Ovulation is inhibited by medroxyprogesterone acetate.
C. Intra-uterine device insertion is less effective than the hormonal methods of emergency contraception.
D. The oral contraceptive pill is free from central nervous system side effects.
E. Combined oral contraceptives increase the risk of ovarian and endometrial cancers.

Q 59. Which of the following statement regarding cyproterone acetate is correct?
A. Is an agonist of the beta oestrogen receptor
B. Is associated with visual disturbance as a recognised side effect
C. Is used in the treatment of acne hirsutism
D. Its administration has no correlation with the day of menstrual cycle
E. Has androgenic properties

Q 60. Which of the following statement regarding the use of combined oral contraception is correct?
A. The effectiveness of the combined oral contraceptive pill is increased by rifampicin.
B. Combined oral contraceptives increase the incidence of premenstrual tension.
C. Spironolactone, an aldosterone antagonist which is used as a diuretic, is also an antiandrogen.
D. Previous venous thrombosis is a relative contra-indication to the use of combined oral contraceptives.
E. Cyproterone acetate can cause hirsutism.

Q 62. Which of the following statement is true regarding the various types of hormonal contraception?
A. Mifepristone is progestogenic.
B. Cyproterone acetate leads to increased levels of cortisol in the blood.
C. Thyroid-binding globulin plasma concentration is decreased in women using the combined oral contraceptive pill.
D. Regular menstrual cycles and fertility return to normal within 6 months of the last progestogen injection (Depo-Provera R).
E. Progestogens are not contra-indicated in patients with porphyria.

Q 62. Which of the following statement is not true regarding the surgical wound infections?
A. Anaerobic organisms exert their lethal effects by producing endo- and exotoxins
B. MRSA wound infection is usually the result of wound contamination by hospital staff
C. Necrotising fasciitis is commoner in carriers of MRSA
D. *Staphylococcus aureus* is the commonest organism to infect the surgical wound
E. With opportunistic organisms, they are the result of a patient's increased immune defence

52. D   53. D   54. A   55. D   56. D   57. B   58. B   59. C   60. C   61. D   62. D.

Q 63. **Which of the following is true regarding Asherman's syndrome?**

A. Is associated with obstetric complications including post partum haemorrhage
B. Is characterised by menorrhagia
C. May be treated by forceps
D. May be treated by a Foley catheter
E. None of the above

Q 64. **Which of the following statement is not true regarding Fallopian tube occlusion?**

A. may be caused by chlamydial infection
B. Tubal occlusion is surprisingly uncommon even in the presence of moderately severe pelvic endometriosis
C. When caused by infection most commonly ascends from the lower genital tract
D. May follow appendicitis
E. Can be assessed using transvaginal ultrasound.

Q 65. **Which of the following drugs used in conventional doses would be an effective pre-operative antiemetic agent?**

A. Atropine
B. Lorazepam
C. Metoclopramide
D. Midazolam
E. Ondansetron

Q 66. **Which of the following is true regarding high-risk patients?**

A. Higk-risk patients must be placed at the end of an operating list to avoid infecting any subsequent patients
B. The hepatitis B virus can be transmitted by splash contamination of the conjunctiva
C. The skin is cleaned at least four times with chlorhexidine
D. When double gloving it is usual to wear the inner glove a half-size larger than the surgeon's usual size
E. None of the above

Q 67. **Which of the following is not true regarding skin preparation before surgery?**

A. Alcohol pooled in the umbilicus must be removed
B. Application of alcoholic solutions chlorhexidine gluconate or povidone-iodine gives better disinfection
C. Chorhexidine and cetrimide (Savlon) is advised when disinfecting the vagina and perineum
D. Chlorhexidine gluconate 4% w/v is a clear solution
E. Povidone-iodine has a broader spectrum and persists longer than chlorhexidine gluconate

Q 68. **With regard to cardiac disorders in surgical patients, which of the following is true?**

A. 60% of patients will re-infarct if operated on within 3 months of a myocardial infarction

B. Creatine phosphokinase (CPK) is a myocardial specific isoenzyme
C. Heart failure can be classified into four groups according to the NYHA classification system
D. Peri-operative myocardial infarction has a mortality of approximately 10%.
E. Risk of perioperative heart failure is approximately 47% if there is a pre-operative cardiovascular history

Q 69. **Hypertension 1 hour following a laparotomy can be due to all the below except which of the following?**

A. Hypocapnia
B. Hypoxia
C. Inadequate analgesia
D. Malignant hyperpyrexia
E. Urinary retention

Q 70. **Following a major surgery in a normal person, which of the following is observed?**

A. Decreased heart rate
B. Decreased metabolic rate
C. Fall in blood glucose concentration
D. Fluid retention
E. Potassium retention

Q 71. **Which of the following is not a risk factor for post-operative wound infection?**

A. Chronic obstructive airways disease
B. Diabetes mellitus
C. Haematoma formation
D. Incorporation of a synthetic mesh
E. None of the above

Q 72. **Which of the following is a predisposing factor for the development of keloid scars?**

A. Patients of Afro-Caribbean origin with dark complexion
B. Secondary wound closure
C. Steroid therapy
D. Use of local bupivacaine
E. Triamcinolone injection

Q 73. **Does failure of wound healing in a surgical wound does not result in which of the following?**

A. Cicatrisation
B. Hypertrophic scarring
C. Incisional hernia
D. Superficial wound disruption
E. Wound dehiscence

Q 74. **Which of the following is caused by the total parenteral nutrition?**

A. Fatty acid deficiency
B. Hypercarbia
C. Hyperglycaemia
D. Hypoglycaemia
E. All the above

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63. D   64. E   65. E   66. B   67. E   68. C   69. A   70. D   71. D   72. A   73. B   74. E.

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