Chapter 3

Amino acids, Peptides and Proteins

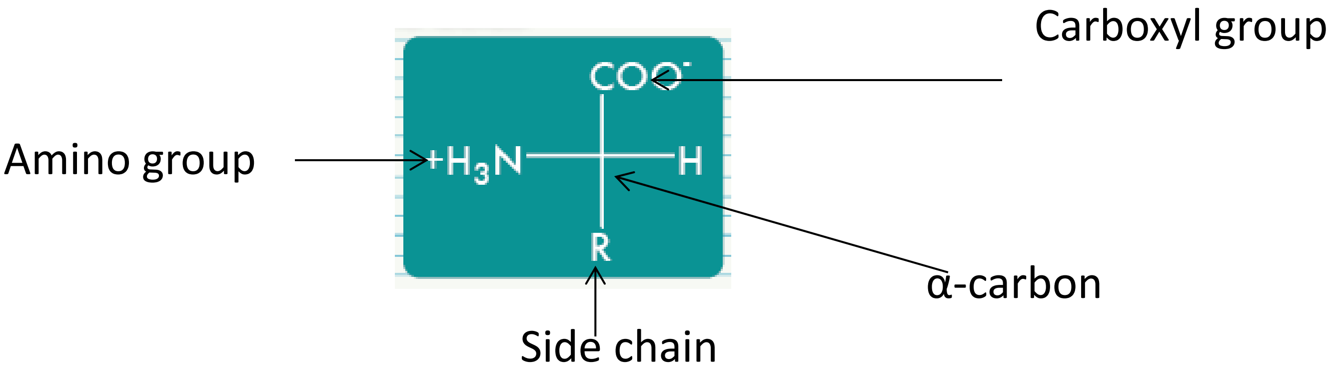
α-Amino Acids

❖ **Structure**

➢ α-Amino acids are organic molecules which consists three groups:

- ✓ Carboxylic (-COOH) group
- ✓ Amino ($-NH_2$) group and
- ✓ Side-chain (R) (specific to each amino acid)

attached to an α-carbon



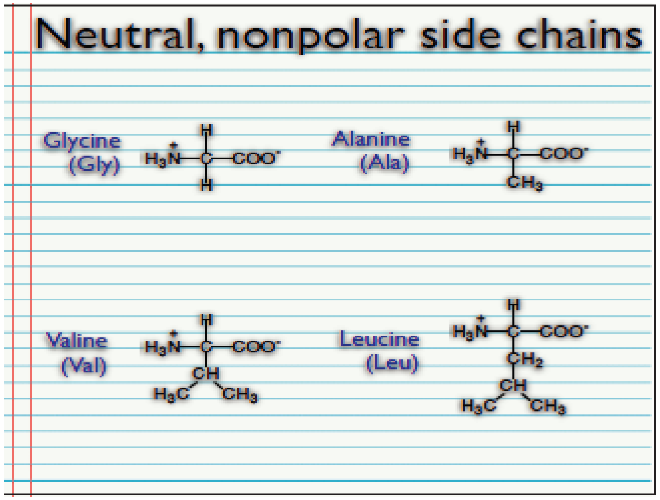
➢ 22 α-amino acids constitute all naturally occurring peptides and proteins (two of these are very recently discovered -Identify which?)

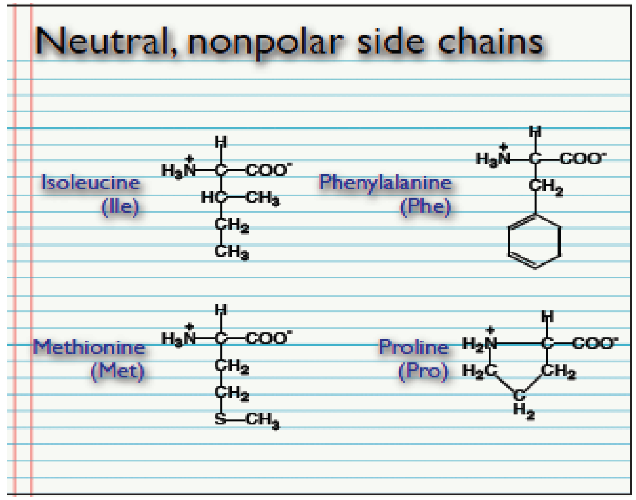
➢ Amino acids can be represented by a text name, a three-letter or one-letter symbolic representations

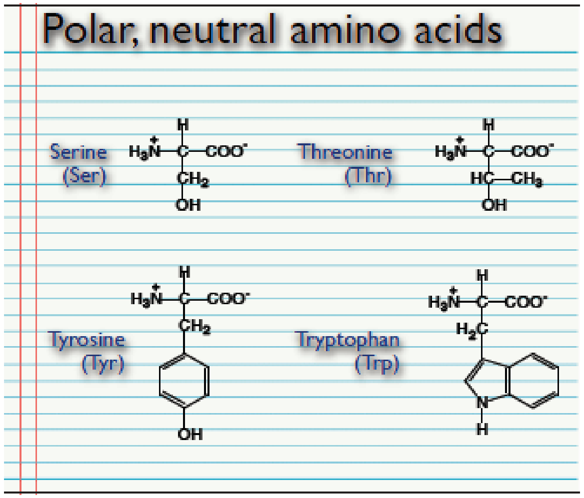
Classification of α-Amino Acids

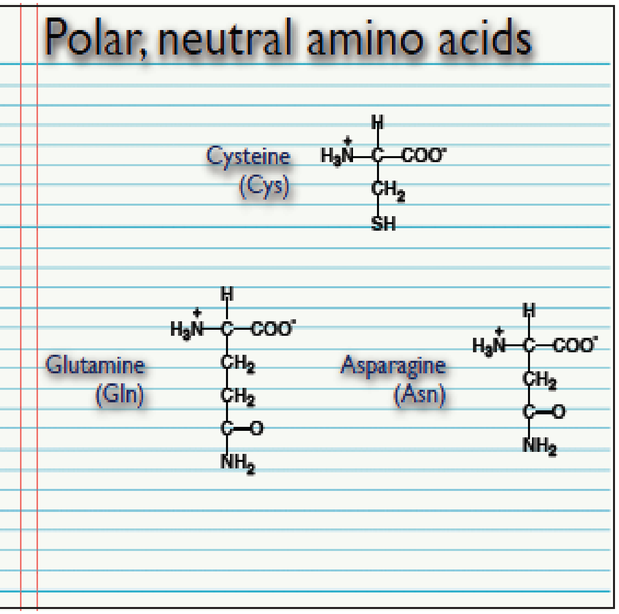
❑Amino acids can be categorized on various bases

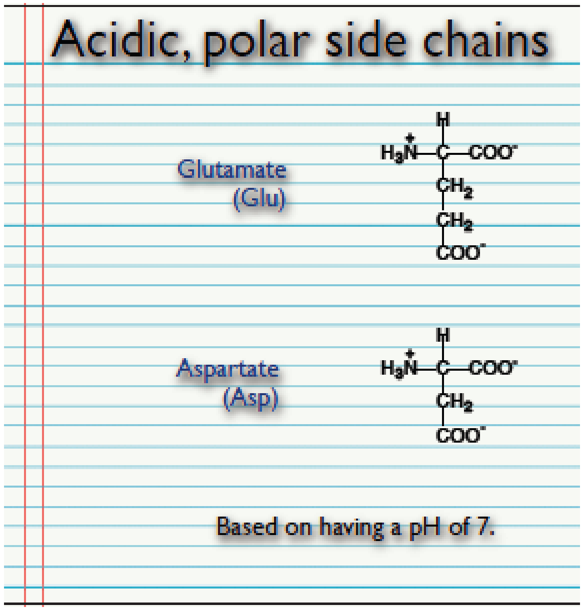
i) <u>Based on nature of their side chains</u>

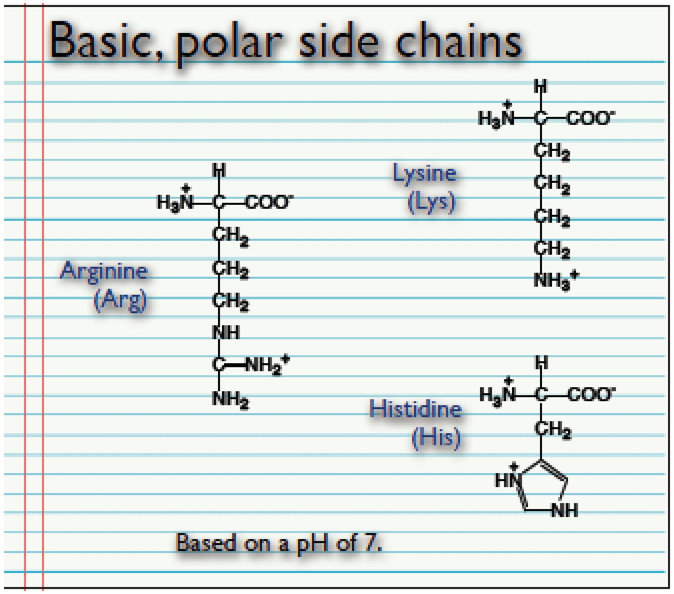












Classification of α-Amino Acids

ii) Polarity of side chain

- **Hydrophobic amino acids**: include all aa's with non-polar side chains
- **Hydrophilic amino acids**: include all polar, neutral chains , negatively charged, positively charged

iii) Based on their biosynthesis

- **Essential amino acids**
  - -aa's not synthesized in the human body (or synthesized at a rate insufficient to meet body's need)
  - - usually obtained from dietary sources (e.g Dairy products, mushroom, grains, vegetable etc.)

  e.g valine, isoleucine, leucine, lysine, Arginine*, histidine*, methionine, threonine, phenylalanine, tryptophan

- **Non-essential amino acids**
  - -Are synthesized in the body in suitable amount

  e.g glycine, Alanine, aspartate, glutamate, asparagine, glutamine, cysteine, serine, tyrosine, proline

iv) Based on their metabolic fates

- **Ketogenic amino acids-** those to end up with metabolite used for synthesis of ketone bodies
- **Glucogenic amino acids-** those to end up with metabolite used for synthesis of glucose

Properties of α-Amino Acids

Physical properties

- All amino acids are crystalline solids with high melting points.
  - ✓ With melting and decomposition tend to occur with in the 200 - 300°C range.

- Amino acids are generally soluble in water and insoluble in non-polar organic solvents such as hydrocarbons (Due to presence of the zwitterions)
  - ✓ The extent of water solubility depends on the size and nature of the "R" group.

- With exception of glycine all α-amino acids are optically active
  - ✓ With variable effect on plane polarized light

    e.g: Dextrorotatory :-(+)alanine, (+)valine

    Levorotatory :- (-)cysteine, (-)tyrosine
  - ✓ But all are with L- configuration

Properties of α-Amino Acids

Ionization of α-amino acids

- ❖ Amino acids contain weakly acidic groups (-COOH, $-NH_2$, -R) capable to ionize in aqueous medium

- ❖ The pKa of each group varies with their proton donor/acceptor properties
  - ➢ The **α-COOH group**: pK1 $\simeq$ 2-3

    -Hence tend to donate its proton at pH ≥ pK1

  - ➢ The **α- NH_2 group**: pK2 $\simeq$9-10

    -Hence unshared e-pairs on **N** can accept proton until pH ≥ pK2

  - ➢ The side chains (R):- pK3 $\simeq$ 3.9 – (~13)

    -Hence can donate its proton at pH ≥ pK3/R (for Ser, Thr, Asp, Glu, Cys, Tyr)

    -Hence can accept proton until pH ≥ pK3/R (for Arg, Lys, His)

Properties of α-Amino Acids

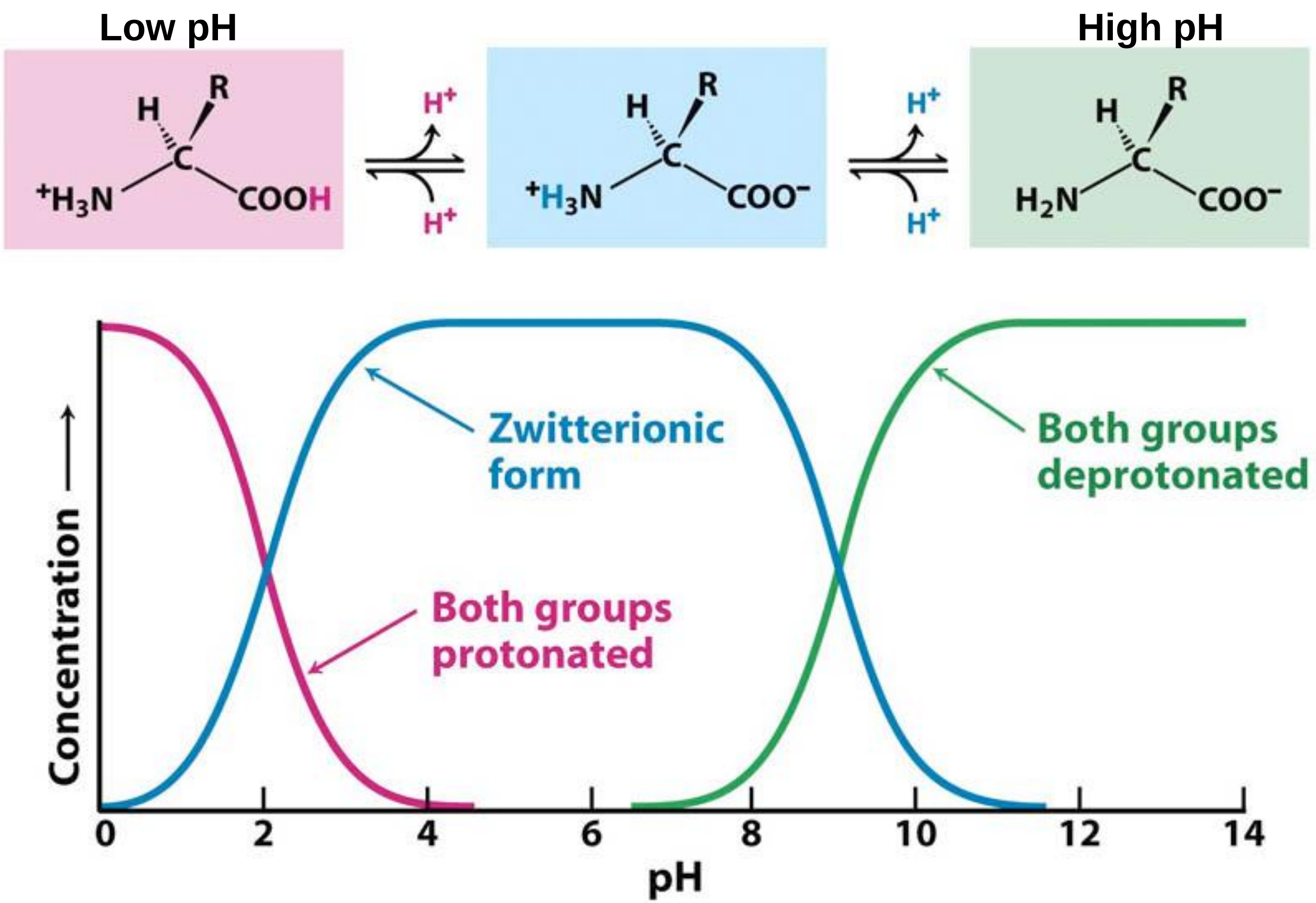
Dissociation constants of α-amino acids

Name	Symbol	R	pK_1, α-COOH	pK_2, α-NH_2	pK_R, R-group
Neutral/non-polar(Aliphatic /alkane side chain) Amino acids					
Glycine	Gly - G	H	2.4	9.8	
Alanine	Ala- A		2.4	9.9	
Valine	Val - V		2.2	9.7	
Leucine	Leu - L		2.3	9.7	
Isoleucine	Ile - I		2.3	9.8	
Polar Uncharged Amino Acids					
Serine	Ser - S		2.2	9.2	~13
Threonine	Thr - T		2.1	9.1	~13
Sulfur containing amino acids					
Cysteine	Cys - C		1.9	10.8	8.3
Methionine	Met-M		2.1	9.3	
Acidic Amino Acids and their Amides					
Aspartic Acid	Asp - D		2.0	9.9	3.9
Asparagine	Asn - N		2.1	8.8	
Glutamic Acid	Glu - E		2.1	9.5	4.1
Glutamine	Gln - Q		2.2	9.1	
Basic Amino Acids					
Arginine	Arg - R		1.8	9.0	12.5
Lysine	- K		2.2	9.2	10.8
Histidine	His - H		1.8	9.2	6.0
Aromatic Amino Acids					
Phenylalanine	Phe - F		2.2	9.2	
Tyrosine	Tyr - Y		2.2	9.1	10.1
Tryptophan	Trp-W		2.4	9.4	
Imino Acids					
Proline	Pro - P		2.0	10.6	

Properties of α-Amino Acids

Ionization cont..

❖ Because of presence of acidic/basic groups present in amino acids exist in +/- form at neutral pH (called **zwitterion** form)



Properties of α-Amino Acids

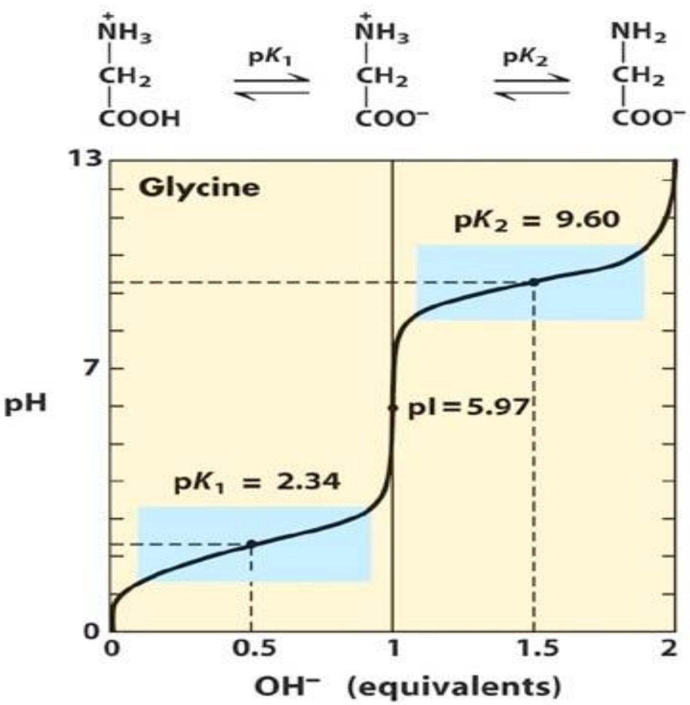
Titration curve and the isoelectric point (PI) of amino acids

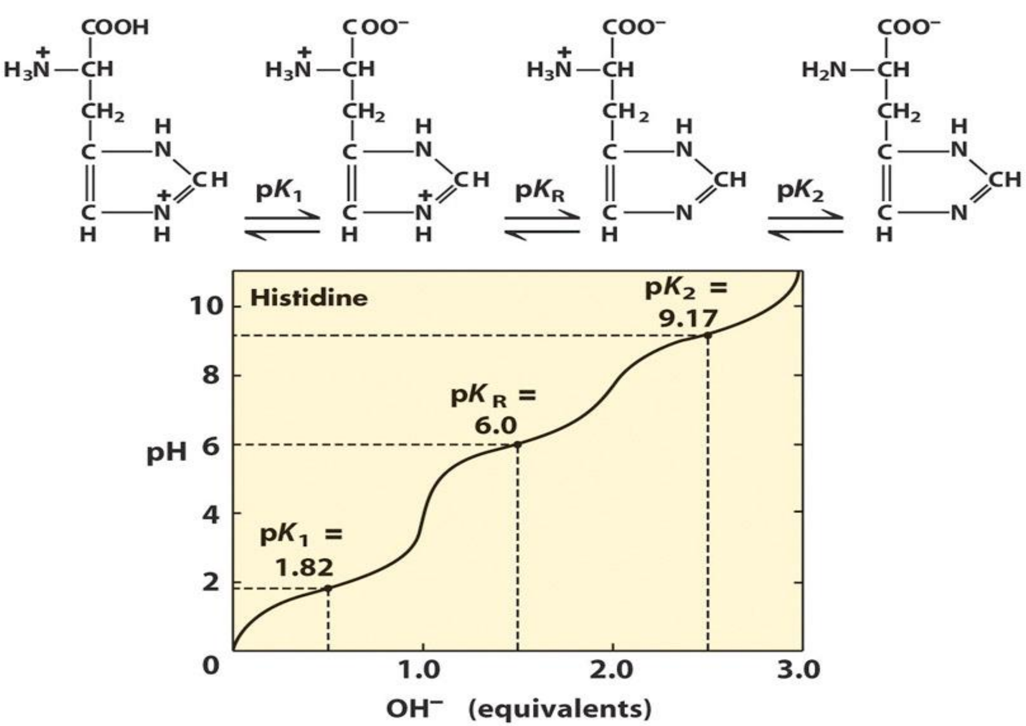
- ❖ Amino acids have two or more titrable groups
- ❖ Accordingly upon titration they
  - ✓acquire different charge at d/t pH
  - ✓give different curve patterns

❖ **Isoelectric point**(PI):Distinct pH at which net charge of an amino acid/protein becomes zero

- ➢ For simple aa's:- **$pI = (pK_1 + pK_2)/2$**
- ➢ For aa's with additional acidic /basic groups:- **$pI = (pK_X + pK_Y)/2$**

where X & Y are pKa values around net zero charge

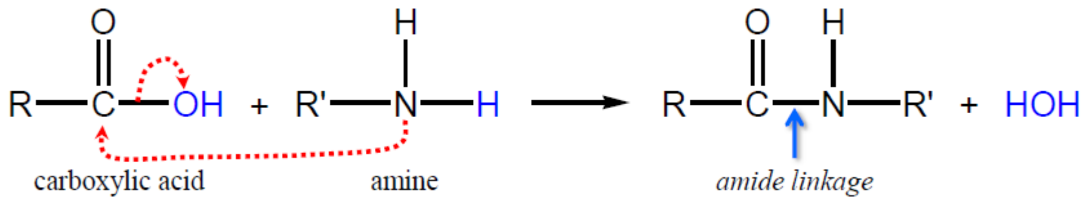


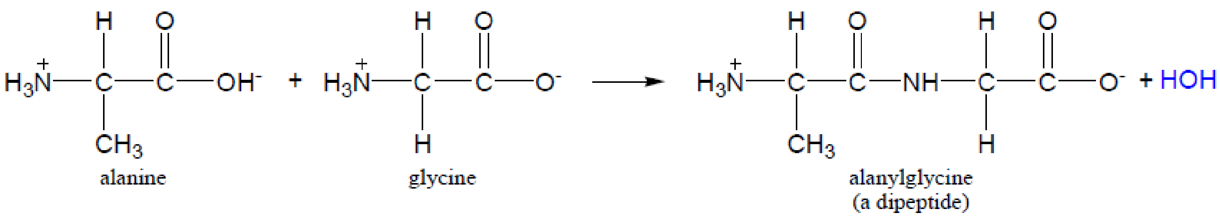


Properties of α-Amino Acids

Linkage

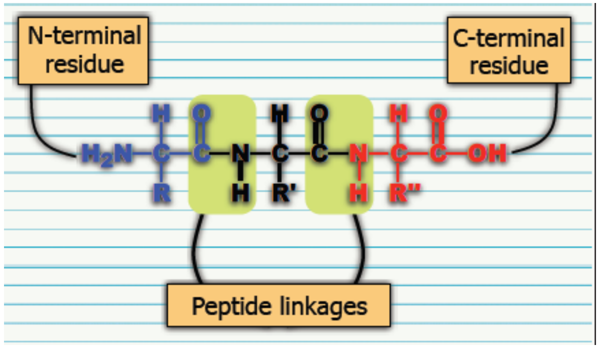
❖ The amino and carboxylic groups of amino acids can be joined thorough **condensation reaction** to form a peptide/amide bond





❖ The condensation product will have two ends : **N-terminal** & **C-terminal ends**

free α-amino group



free α-carboxyl group

Peptides

- ❖ Peptides are oligomeric molecules made by Joining 2-50 amino acids.
  - ➢ by condensation reaction b/n amino and carboxylic groups of each aa's

- ❖ Typical peptides includes
  - ➢ **Peptide hormones**:- bradykinins, gastrins, oxytocins etc.
  - ➢ **Neuropeptides**:- endorphins, vasopressin, atrial-natriuretic peptide etc.
  - ➢ **Peptide antibiotics**:- tyrothricinm,bacitracin,gramicidin,valinomicin etc.
  - ➢ **Toxins**:-palutoxins, agatoxins, curtatoxins etc.
  - ➢ **Regulation peptides**:-anserine, carnosine, etc.

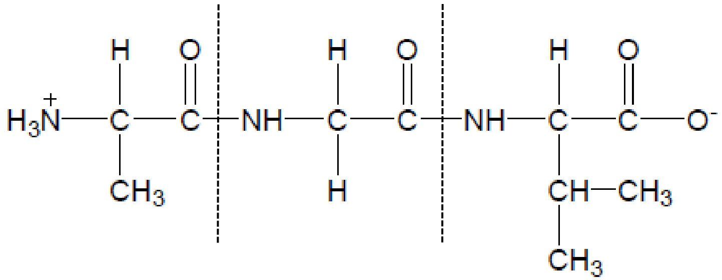
Peptides

Naming of Peptides/proteins

- ❖ Peptides are named by starting at the N-terminal end and listing the amino acid residues from left to right.

- ❖ **Three ways** can be used

Example : for a tripeptide



- ➢ Complete textual name :- **alanylglycylvaline**
- ➢ Three-letter abbreviations (letters should be separated by dashes):- Ala-Gly-Val
- ➢ One-letter abbreviations :-AGV

Proteins

- ❖ Proteins are polymeric molecules made by Joining more than 50 amino acids.
  - ➢ by condensation reaction b/n amino and carboxylic groups of each aa's

Size of proteins

- ❖ Proteins are very large polymers of amino acids with molecular weights that vary from 6000 amu to several million amu.

Protein	Molecular Weight (amu)	Number of Amino Acid Residues
Insulin	6,000	51
Cytochrome c	16,000	104
Growth hormone	49,000	191
Rhodopsin	38,900	348
Hemoglobin	65,000	574
Hexokinase	96,000	730
Gamma globulin	176,000	1320
Myosin	800,000	6100

- ❖ Proteins are too large to pass through cell membranes, and are contained within the cells where they were formed unless the cell is damaged by disease or trauma.
  - ➢ Persistent large amounts of protein in the urine are indicative of damaged kidney cells.
  - ➢ Heart attacks can also be confirmed by the presence of certain proteins in the blood that are normally confined to cells in heart tissue.

Proteins

Classification of proteins

i) By solubility

- On the bases of solubility in a range of solvents proteins can be classified as follows
  - **Albumins**-Soluble in water and salt solutions
  - **Globulins**:-Insoluble/sparingly soluble in water but soluble in salt solutions
  - **Prolamines**- Soluble in 70-80% EtOH but insoluble in water and absolute EtOH
  - **Histones**- are water- and dilute acid-**soluble** basic proteins
  - **Glutelins** :-Are plant proteins insoluble in water and absolute alcohol but soluble in dilute alkalies and acids.
  - **Protamines** are basic proteins soluble in water and are not coagulated by heat
  - **Albuminoids (scleroproteins)**- are characterized by great stability and insolubility in water and salt solutions and resistant to proteolytic enzymes.

Proteins

<u>Classification of proteins</u>

ii) By Structural Shape

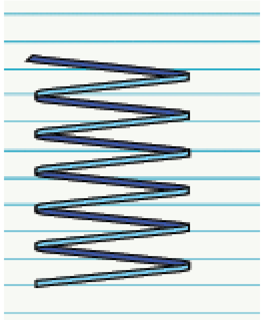
❖ On the bases of their structural shapes proteins can be classified as follows

➢ **Fibrous proteins :-**are made up of long rod-shaped or string like molecules that can intertwine with one another and form strong fibers.

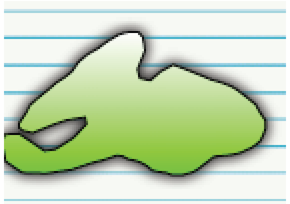
–insoluble in water

–major components of connective tissue, elastic tissue, hair, and skin

e.g. Actin, Collagen, Elastin, Fibronectin, Keratin, Myosin, Tubulin



➢ **Globular proteins**:- are more spherical in shape



–dissolve in water or form stable suspensions.

–not found in structural tissue but are transport proteins, or proteins that may be moved easily through the body by the circularity system

e.g. Albumins, globulin, Fibrin, Hemoglobin, Myoglobin, Thrombin, Transferrin

Proteins

Classification of proteins

iii) By composition

- Proteins can also be classified as:-
  - **Simple proteins**:- contain only amino acid residues
  - **Conjugated proteins**:- contain organic or inorganic components called prosthetic groups
    - ✓ **Nucleoproteins** :- contain nucleic acids
    - ✓ **Lipoproteins**:- contain lipids (e.g fibrin in blood, serum lipoproteins)
    - ✓ **Glycoproteins** :- contain carbohydrates (e.g gamma globulin in blood, mucinin saliva)
    - ✓ **Phosphoproteins** :- contain phosphate groups (e.g casein in milk)
    - ✓ **Hemoproteins** :- contain heme(e.g hemoglobin, myoglobin, cytochromes)
    - ✓ **Metalloproteins** :- contain metal ions such as iron (in feritin, hemoglobin) or zinc (in alcohol dehydrogenase)

Proteins

Classification of proteins

iv) By functions

- A typical human cell contains 9000 different proteins; the human body contains about 100,000 different proteins.
- Proteins perform crucial roles in all biological processes
  - **Catalytic function:-** Nearly all reactions in living organisms are catalyzed by proteins functioning as *enzymes. Without these catalysts, biological reactions would proceed much more slowly.*
  - **Structural function:-** In animals structural materials other than inorganic components of the skeleton are proteins, such as *collagen (mechanical strength of skin and bone) and keratin (hair, skin, fingernails).*
  - **Storage function:-** Some proteins provide a way to store small molecules or ions, e.g., ovalbumin(used by embryos developing in bird eggs), casein (a milk protein) and gliadin(wheat seeds), and ferritin(a liver protein which complexes with iron ions)
  - **Protective function:-** Antibodies are proteins that protect the body from disease by combining with and destroying viruses, bacteria, and other foreign substances. Another protective function is *blood clotting, carried out by thrombin and fibrinogen*.
  - **Regulatory function:-** Body processes regulated by proteins include growth (growth hormone) and thyroid functions (thyrotropin).
  - **Nerve impulse transmission:-** Some proteins act as receptors for small molecules that transmit impulses across the synapses that separate nerve cells. (Rhodopsinin in vision)
  - **Movement function:-**The proteins actin and myosin are important in muscle activity, regulating the contraction of muscle fibers
  - **Transport function:-** Some proteins bind small molecules or ions and transport them through the body.
    - –Serum albumin -blood protein that carries fatty acids between fat (adipose) tissue and other organs.
    - –Hemoglobin :-carries oxygen from the longs to other body tissues.
    - –Transferrin :- carries of iron in blood plasma

Proteins

Levels of Protein Structure

- Many protein molecules consist of a chain of amino acids twisted and folded into a complex three-dimensional structure

- The complex 3D structures of proteins
  - impart unique features to proteins
    - ✓ that allow them to function in diverse ways

- There are four levels of organization in proteins structure
  - Primary
  - Secondary
  - Tertiary and
  - Quaternary

Proteins

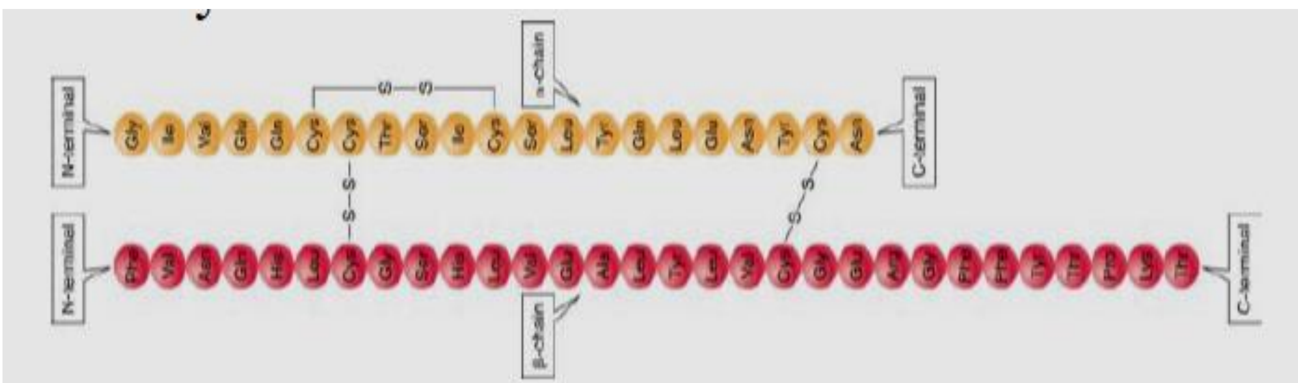
Levels of Protein Structure

i) **Primary structure**

- ❖ The primary structure of a protein is the linear sequence of the side chains that are connected to the protein backbone

- ❖ Each protein has a unique sequence of amino acid residues that cause it to fold into a distinctive shape that allows the protein to function properly.

e.g primary structure of human insulin



Proteins

Levels of Protein Structure

ii) **Secondary structure**

- ❖ Hydrogen bonding causes protein chains to fold and align to produce orderly patterns called **secondary structures.**
- ❖ Two distinct types of protein secondary structures are

➢ **α-helix**

- ✓ Involve single protein chain twisted to resemble a coiled helical spring
- ✓ Proteins are held in this shape by hydrogen bonding interactions between amide groups, with the side chains extending outward from the coil.
- ✓Every amide hydrogen and carbonyl oxygen is involved in a hydrogen bond.

➢ **β-Pleated sheets**

- ✓ Created between adjacent sheets of protein held together by hydrogen bonding
- ✓Every amide hydrogen and carbonyl oxygen is involved in a hydrogen bond.

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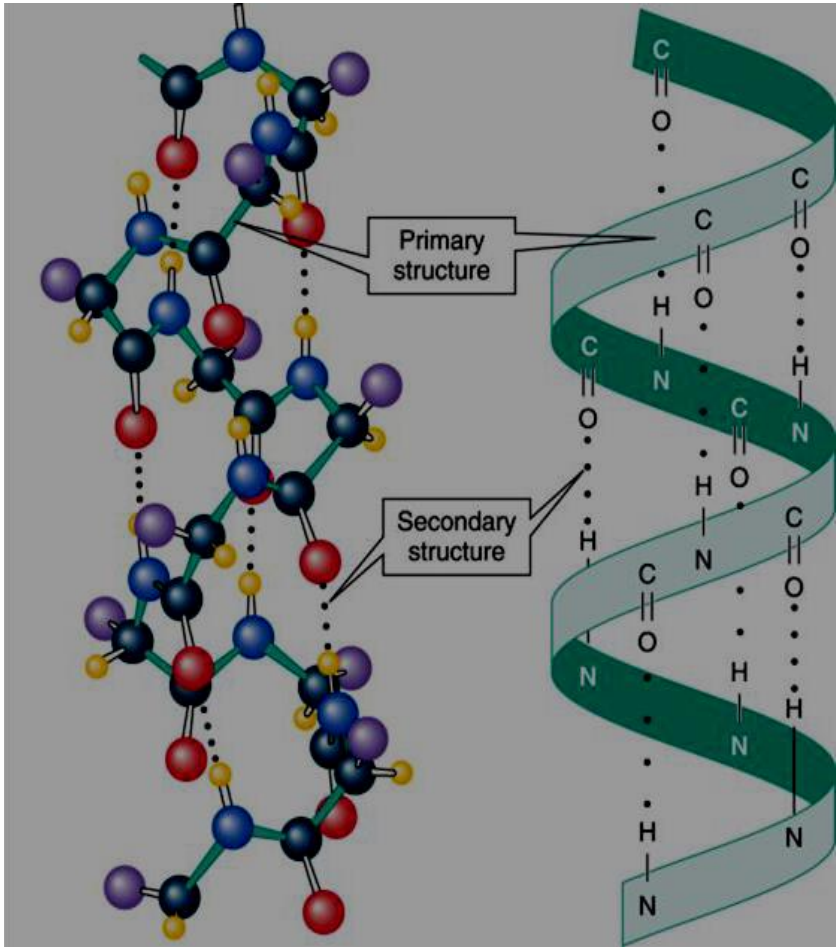
Proteins

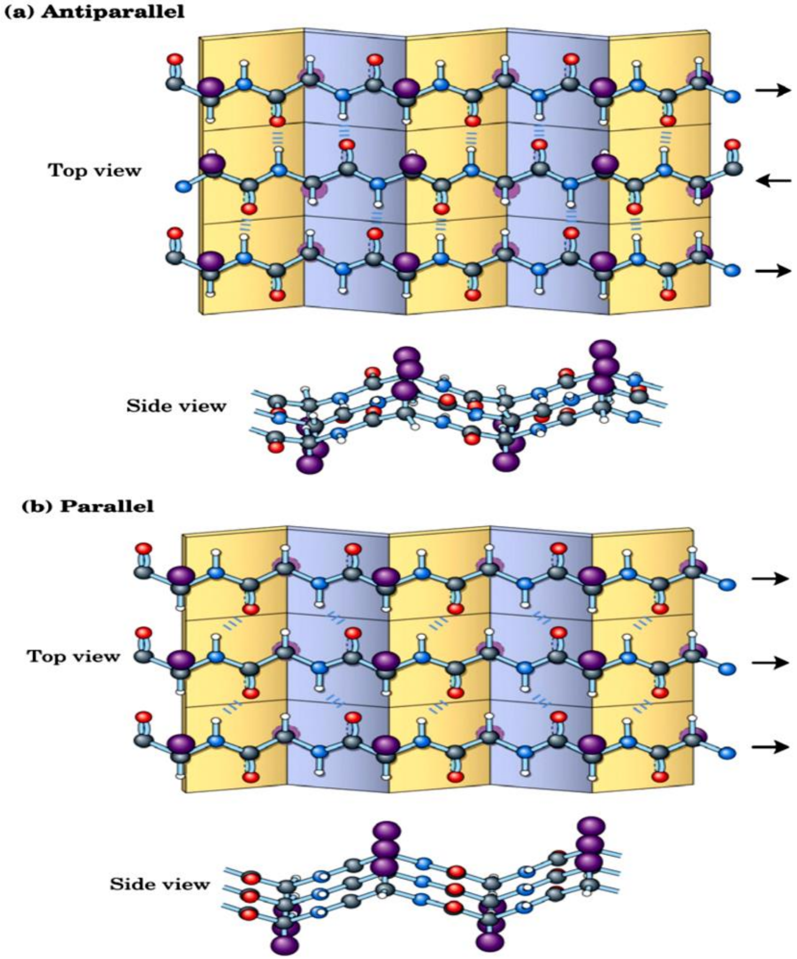
Levels of Protein Structure

ii) Secondary structure cont…

α-helix



β-Pleated sheets



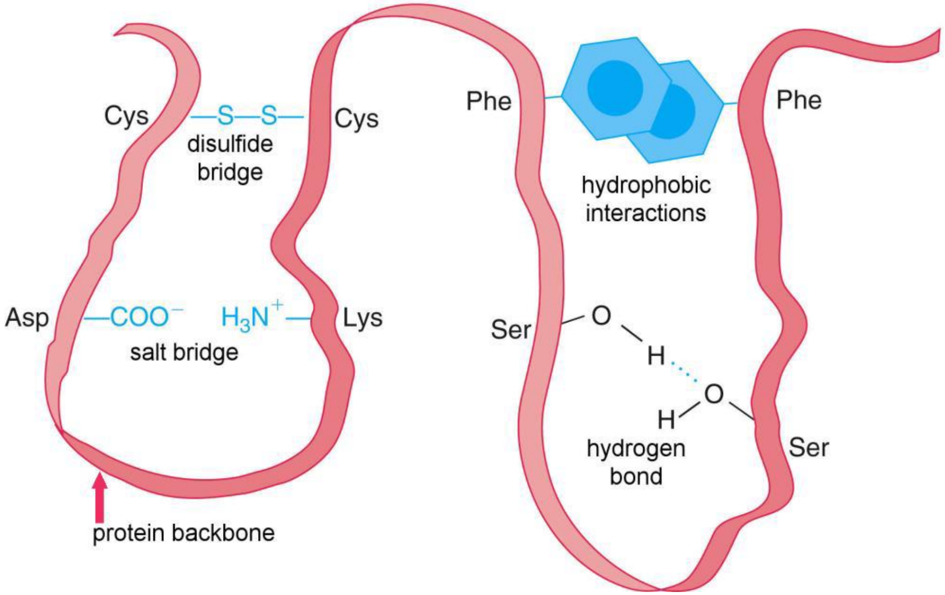
Proteins

Levels of Protein Structure

iii) **Tertiary structure**

- Refers to the bending and folding of the protein into a **specific three-dimensional shape**.

- These structures result from four types of interactions between aa side chains
  - **Disulfide bridges**:- b/n two cysteine residues
  - **Salt bridges** :- result from the interactions of the ionized side chains of acidic amino acids(-COO-) and the side chains of basic amino acids (—NH3+).
  - **Hydrogen bonds:-** can form between a variety of side chains
  - **Hydrophobic interactions:-** result from the attraction of nonpolar groups

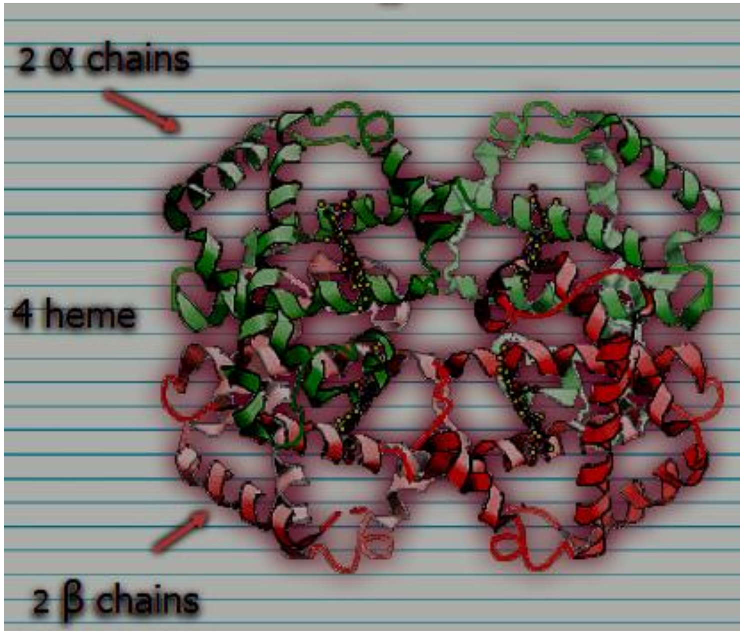


Proteins

Levels of Protein Structure

iv) **Quaternary structure**

- When two or more **polypeptide chains (subunits)** are held together by disulfide bridges, salt bridges, hydrogen bond, or hydrophobic interactions
    - a larger protein complex called **quaternary structure** will be formed
- Hemoglobin is a complex protein made of four subunits
    - Two identical alpha chains containing 141 AA's and
    - Two identical beta chains containing 146 AA's.
    - Each subunit contains a *heme group located in crevices near the exterior of the molecule.*



Proteins

Protein *Hydrolysis*

- Amide bonds of proteins can be hydrolyzed (broken down) under **different** conditions

  - into smaller peptides, or all the way to amino acids under
    - ✓ **Acidic or basic conditions or**
    - ✓ **Enzymatically**

- The hydrolysis product depends on the hydrolysis time, temperature, and pH

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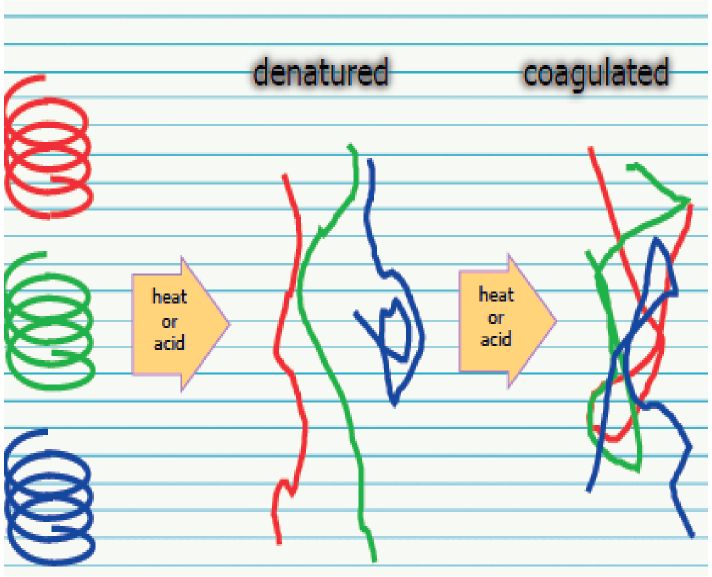
$$\text{protein} + H_2O \xrightarrow{H^+ \text{ or } OH^-} \text{smaller peptides} \xrightarrow{H^+ \text{ or } OH^-} \text{amino acids}$$

Proteins

Protein *denaturation*

- ❖ Proteins are maintained in their native state (their natural 3D conformation) by stable secondary and tertiary structures, and by aggregation of subunits into quaternary structures.

- ❖ These native and stable structures can be randomized and disorganized by different conditions
  - ➢ Such as extreme temprature , pH or others
    - ✓ This phenomenon is called **denaturation**



Substance or condition	Effect on Proteins
Heat and ultraviolent light	Disrupt hydrogen bonds and ionic attractions by making molecules vibrate too violently; produce coagulation, as in cooking an egg
Organic solvents (ethanol and others miscible with water)	Disrupt hydrogen bonds in proteins and probably form new ones with the proteins
Strong acids or bases	Disrupt hydrogen bonds and ionic attractions; prolonged exposure results in hydrolysis of protein
Detergents	Disrupt hydrogen bonds, hydrophobic interactions, and ionic attractions.
Heavy-metal ions (Hg^{2+}, Ag^{+}, and Pb^{2+})	Form bonds to thiol groups and precipitate proteins as insoluble heavy-metal salts