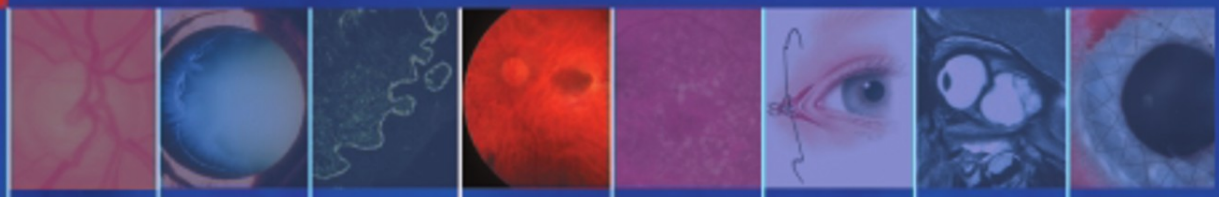
ESSENTIALS IN OPHTHALMOLOGY

G. K. KRIEGLSTEIN · R. N. WEINREB

Series Editors



Glaucoma | Cataract and Refractive Surgery | Uveitis and Immunological Disorders | **Vitreo-retinal Surgery** | Medical Retina | Oculoplastics and Orbit | Paediatric Ophthalmology, Neuro-ophthalmology, Genetics | Cornea and External Eye Disease

Vitreo-retinal Surgery

Edited by

B. KIRCHHOF

D. WONG

Springer

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Vitreo-retinal Surgery

With 79 Figures, Mostly in Colour,
and 16 Tables

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ISBN 3-540-20044-4
Springer Berlin Heidelberg New York

ISSN 1612-3212

Library of Congress Control Number: 2004105920

Springer is a part of Springer Science + Business Media

springeronline.com

Printed in Germany

Cover picture "Cataract and Refractive Surgery" from Kampik A, Grehn F (eds) Augenärztliche Therapie. Georg Thieme Verlag Stuttgart, with permission.

Editor: Marion Philipp, Heidelberg
Desk editor: Martina Himberger, Heidelberg
Production: ProEdit GmbH, Elke Beul-Göhringer, Heidelberg
Cover design: Erich Kirchner, Heidelberg
Typesetting and reproduction of the figures: AM-productions GmbH, Wiesloch

Printed on acid-free paper
24/3150beu-göh 5 4 3 2 1 0

Foreword

Essentials in Ophthalmology is a new review series covering all of ophthalmology categorized in eight subspecialties. It will be published quarterly; thus each subspecialty will be reviewed biannually.

Given the multiplicity of medical publications already available, why is a new series needed? Consider that the half-life of medical knowledge is estimated to be around 5 years. Moreover, it can be as long as 8 years between the description of a medical innovation in a peer-reviewed scientific journal and publication in a medical textbook. A series that narrows this time span between journal and textbook would provide a more rapid and efficient transfer of medical knowledge into clinical practice, and enhance care of our patients.

For the series, each subspecialty volume comprises 10–20 chapters selected by two distinguished editors and written by internationally renowned specialists. The selection of these contributions is based more on recent and noteworthy advances in the subspecialty than on systematic completeness. Each article is structured in a standardized format and length, with citations for additional reading and an appropriate number of illustrations to enhance important points. Since every subspecialty volume is issued in a recurring sequence during the 2-year cycle, the reader has the opportunity to focus on the progress in a particular subspecialty or to be updated on the whole field. The clinical relevance of all material presented will be well established, so application to clinical practice can be made with confidence.

This new series will earn space on the bookshelves of those ophthalmologists who seek to maintain the timeliness and relevance of their clinical practice.

G. K. Krieglstein
R. N. Weinreb
Series Editors

Preface

This first issue of *Vitreoretinal Surgery,* in the series *Essentials in Ophthalmology,* has been written to update our knowledge on the large body of experimental research performed to date on the most urgent problems of vitreoretinal disease. Priority is given to the most important problems in terms of patient numbers – retinal degeneration, retinal oedema, and proliferative vitreoretinopathy.

Proliferative vitreoretinopathy (PVR) is the leading cause of blindness in retinal detachment (RD). Recent progress in surgical techniques, sophisticated surgical tools and new vitreous tamponades has reduced the number of enucleations, at least in Europe, but ultimately the risk of PVR has not been reduced, which is reported to be between 5% and 10% for idiopathic PVR, and between 10% and 45% for ocular trauma (the incidence being higher in cases of perforating and blunt injuries and lower with intraocular foreign bodies). Functional outcome of surgery for PVR is often disappointing despite attached retina. Carl Sheridan (Liverpool) reports on the cellular mechanisms of PVR. Adjunct pharmacotherapy reduces the number of reoperations in eyes with established PVR. Improvement of functional outcome requires that high-risk eyes are identified and selected, and that adjunct pharmacotherapy is applied prior to the establishment of PVR. The chapter by Chee Kon (London) elaborates the criteria for detecting eyes with increased risk of PVR, justifying a prophylactic dose of cytostatic drugs, and that by Martin Snead (Cambridge) portrays the "giant retina tear" as an example of a high-risk PVR situation. David Charteris (London) describes the pharmacological progress made in preventing PVR in eyes at risk, and David Wong (Liverpool) elaborates the rationale for heavier than water long-term vitreous substitutes in the prevention and treatment of PVR.

Retinal degeneration is common as a complication of age-related retinal pigment epithelial cell insufficiency (age-related macular degeneration), as a consequence of the inflammatory diabetic metabolism (diabetic macular oedema), and as a result of inherent outer retinal genetic disease (retinitis pigmentosa). Peter Walter (Aachen) reports on the latest progress on epiretinal implants for retinitis pigmentosa as research project results are turned into a commercially available medical device. The chapter by Antonia Joussen (Cologne) explains medical aids for macular oedema of different origins according to pathogenesis. Jan van Meurs (Rotterdam) reports the first experience with translocation of autologous whole grafts of choroids and retinal pigment epithelium under the macula, and Johann Roider (Kiel) questions the rationale of transpupillary thermotherapy in age-related macular degeneration.

Other chapters in the volume are by Silvia Bopp (Bremen), who discusses the latest surgical techniques for modulating macular oedema due to epiretinal membranes, and Tom Williamson (London), who describes the diagnostic and therapeutic value of vitrectomy in uveitis.

We hope that this review of the latest research in the field of vitreoretinal surgery will be of interest to practicing ophthalmologists and researchers alike.

Bernd Kirchhof
David Wong

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Retinal Implants

Peter Walter

1

Core Messages

- Retinal implants may restore vision in retinitis pigmentosa (RP) related blindness
- Retinal implants provide local electrical stimulation by an electrode array
- Electrode arrays can be placed onto or underneath the retina
- Animal experiments have shown promising results
- Clinical trials are in preparation or have just started

1.1 Introduction

Although considerable progress has been made in treating vitreoretinal disorders with modern surgical or pharmacological approaches, there are still some untreatable conditions which lead to blindness. One of the major causes of untreatable blindness is the manifestation of progressive retinal degeneration as in retinitis pigmentosa (RP) and RP-like dystrophies. It is estimated that worldwide 1.5 million individuals are affected by RP. It is further estimated that in Germany 15,000 subjects are legally blind as a result of RP [30].

A number of treatments have been tried, including immunostimulation [48], vitamin supplementation [6], oxygen therapy [41], scleral resection, and combinations of these techniques and others [5, 34]. All these approaches have failed to show a benefit for patients in terms of improvement of visual acuity or visual field. Because RP is caused by mutations in genes coding for key enzymes in the primary visual processes, gene therapy has been suggested as a therapeutic option. It has been shown that it is possible to transfer copies of these genes or growth factor encoding genes into retinal photoreceptors using different viral vectors [1, 3, 21, 31, 42]. These approaches have shown promising results but are also possibly associated with severe systemic complications [29]. Retinitis pigmentosa is caused by a variety of mutations so that the substitution of a single gene may not be effective in a large number of cases. Moreover, gene therapy may be useful in preventing the disease from progressing but may be less useful in very advanced cases of atrophy of the outer retinal layers.

In the late 1960s it was suggested that visual perception in blind subjects could be induced by electric stimulation of different levels of the visual system beyond the photoreceptors. Brindley and his group implanted early cortical stimulators with which he obtained visual sensations in subjects blind from RP [7, 8]. The visual cortex is the primary target in Dobelle's system, which is based on a small camera and an ultrasound detector. Information from both sensor systems is used in a visual processor, and stimulation pulses are provided to an electrode array positioned on the dura at the occipital cortex. A few patients were implanted with this system, and according to information from the company visual perception was achieved, allowing a blind subject to walk in unknown terrain [15, 16].

Considerable improvements in cortical prostheses have now been achieved by fabrication of new electrode arrays [33, 47]. Although the cor-

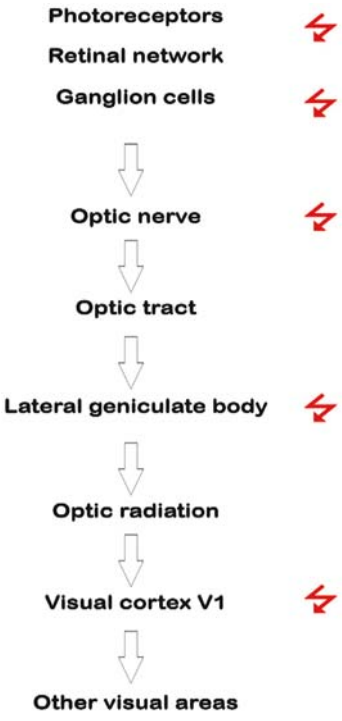

Fig. 1.1. Flow chart of the sensory input to the visual system and currently discussed targets for electrical stimulation as visual prostheses. *From top to bottom* subretinal retinal implant, epiretinal retinal implant, optic nerve cuff electrode, LGB stimulator, cortical prosthesis

tical prosthesis approach has been followed for some years, a retinal stimulator may be more useful with respect to topography (Fig. 1.1). Because many interneurons are involved in the processing of information between retinal photoreceptors and the visual cortex, a stimulation paradigm for realistic visual perception must be much more complex when the target for stimulation is central in the cortex when compared to peripheral stimulation in the retina. Cuff electrodes have been used for the stimulation of peripheral nerves [12]. A variation of such an electrode has also been considered for use as a stimulating electrode for the optic nerve. Experiments have already been performed in blind subjects. In these experiments localized phosphenes were elicited [14, 44]. Because the optic nerve fibers are very densely packed, a topographic correlation between the stimulation electrodes and visual perception was difficult to obtain. Several research groups have therefore decided to work on retinal stimulation.

Because in RP the degenerative process starts in the photoreceptors, the ideal approach would be to simply replace the abnormal photoreceptors by technical elements such as very small photodiodes, which can transform light energy into electrical power which can then be used to stimulate naturally the postsynaptic bipolar and horizontal cells in the retina. This idea was followed by Alan Chow and co-workers in the USA [9, 35] and by Eberhart Zrenner and his group in Germany [50]. The approach was published as the subretinal approach to a retinal prosthesis. Another concept for retinal implants comprises the fixation of a microelectrode array onto the retinal surface. In this epiretinal approach energy for the implant is provided by inductive or optoelectronic pathways. This method was described by Eugene deJuan and co-workers at Wilmers Eye Institute in Baltimore, by Joe Rizzo and his group at the MIT in Boston, and by Rolf Eckmiller and the German EPI-RET consortium, and was published as the epiretinal approach towards a retinal prosthesis [17, 26, 36].

1.2 Approaches for Retinal Implants

Electrical stimulation of the retina has been widely used in animal experiments to study the physiology of the retina with single electrodes [22, 24, 43]. Wolf and Dawson published experiments on therapeutic approaches using direct or indirect electrical retinal stimulation [13, 49]. Although these results were published 30 years ago, a device for application in blind humans was not fabricated. To obtain visual perception an array of very small microelectrodes has to be fabricated supplying stimulation pulses at various locations simultaneously and independently depending on the picture required. Therefore flexible and very thin microelectrode arrays for stimulation have been discussed which should be placed onto or underneath the retina (Fig. 1.2).

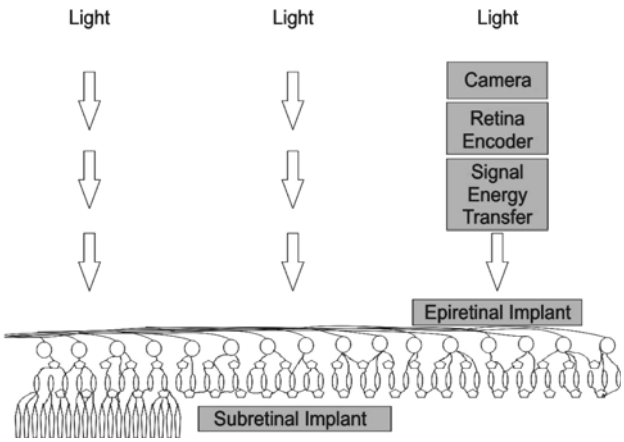

Fig. 1.2. Principal approaches for a retinal prosthesis. *Left* normal flow of light through the retina; *centre and left* receptor degeneration; *centre* subretinal approach: light activates microphotodiodes in the subretinal space inducing stimulation of postsynaptic neurons; *right* epiretinal approach: light is captured by a camera outside the eye. The signal is processed by simulation of receptive field properties. Energy and signal transfer is mediated by inductive or optoelectronic transponder systems

1.3 The Subretinal Approach

The idea of A. Chow and E. Zrenner was to use devices which could transform light energy into electrical energy and to insert a large number of these elements underneath the retina in the subretinal space. The basic concept of the subretinal approach was to replace the degenerated photoreceptors with technical elements with a similar function, i. e. the transformation of light to electrical energy. The charming advantage of this approach is that the topography of the incoming signal is more or less the original one. The postsynaptic cells in the retina, bipolar cells and horizontal cells are supplied with a local electrical input from the respective technical elements. No signal processing system is needed for this approach. The US and German groups working on the subretinal approach fabricated microphotodiode arrays as small discs or flexible foils which were inserted in the subretinal space. Animal experiments showed that the implantation of subretinal devices is possible and safe either through a transvitreal approach (ab interno) after vitrectomy and retinotomy or through a transchoroidal route (ab externo). The materials which were used for subretinal implants were clinically well tolerated; however, the material itself showed signs of oxidative or enzymatic damage of the metal layers. Therefore an adequate coating of subretinal devices is indispensable [40, 11, 28]. Chow had already implanted his device in seven humans suffering from end-stage RP. He described that the surgery was well tolerated by all patients and that no complication occurred. The patients reported visual perceptions. At present, it is still not clear whether this effect is a response to specific action of the device or if this effect is an unspecific response to the surgery itself. It could be speculated that vitrectomy and the opening of the subretinal space for insertion of the implant may lead to a release of neurotrophic factors, which could explain the results [10]. Another question arises from calculations demonstrating that the energy released by currently available microphotodiodes is not enough to generate electric power in a range sufficient to stimulate retinal neurons. Therefore, German researchers working on the subretinal prosthesis are developing a secondary system for amplification of the incoming signal to provide enough energy for neuronal stimulation. Figure 1.3 shows a prototype of an active subretinal implant.

Summary for the Clinician

- **In the subretinal approach the retinal stimulator is placed underneath the retina**
- **The stimulator consists of thousands of miniaturized photodiodes which transform light into electric power**
- **Postsynaptic cells in the retina are the target for stimulation**
- **Image processing equipment or a camera is not necessary**

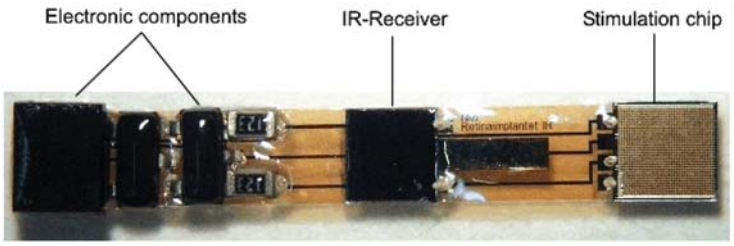

Fig. 1.3. Prototype of an active subretinal implant with a receiver for IR energy (*centre*), electronic components (*left*) and the subretinal stimulator (*right*)

1.4 The Epiretinal Approach

Electrical stimulation of the inner surface of the retina has been followed as a possible approach to retinal prostheses. The target cells for epiretinal electrical stimulation are the ganglion cells. Due to the natural processing of the visual input in the healthy retina by interneurons, ganglion cells do not efficiently respond to pulses directly derived from the visual input. Therefore the visual input in this approach has to be processed outside the retina in a neural network simulating functional properties of receptive fields with respect to contrast, colour, orientation, velocity, and other parameters. As a result of this signal, processing ganglion cells can be efficiently stimulated by series of short current pulses. In contrast to the subretinal approach, in the epiretinal approach the scene needs to be captured by a small camera and processed before the ganglion cells can be stimulated.

Cameras can be fabricated as CMOS systems characterized by low energy consumption, small size and efficient on-chip signal processing [27]. This camera module has to be integrated into the frame of normal spectacles. The output from the camera is further processed by the retina encoder simulating properties of the target cells. Because prior to implantation it is not known which electrode is in good contact with which ganglion cell, the signal processing should be adjustable depending on the visual sensations of the patient. The retina encoder calculates the pulse sequence and the parameters of each stimulation pulse at each electrode depending on the camera input. The number of pulses, amplitude and duration of each phase of each pulse have to be determined and then transmitted to the implant [17–19]. Not only the signal is transmitted to the implant but also the necessary energy to drive the electric circuits of the device. Currently an electromagnetic inductive coupling with a primary and a secondary coil is being fabricated. However, an optoelectronic solution which may provide higher data rates is also under construction. Figure 1.4 shows the general concept of an epiretinal prosthesis.

Summary for the Clinician

- **In the epiretinal approach the retinal stimulator is placed onto the retinal surface**
- **Ganglion cells are the target of epiretinal stimulation**
- **The image needs to be captured by an extraocular camera**
- **The camera signal is further processed by a retinal encoder simulating normal retinal signal processing based on receptive field properties**
- **The information on the pattern of electrode activity is transmitted via radiofrequency into the eye**
- **Energy to drive the implant is transmitted via radiofrequency into the eye**

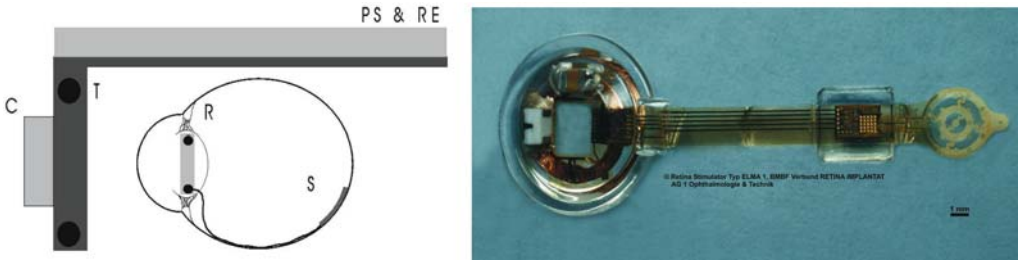

Fig. 1.4. *Left* General concept of an epiretinal prosthesis. The camera *C* captures visual data, the retina encoder *RE* processes the data according to the functional properties of receptive fields of the assumed target ganglion cells (*PS* power supply). The transponder system *T* transmits data and energy using inductive coupling into the implant consisting of the receiver *R* unit and the stimulator *S*, which is fixated onto the retinal surface. *Right* prototype of an epiretinal prosthesis (EPI-RET research group) with the receiver embedded into a silicone disc (*left part*), a polyimide-based microcable, and the microcontact array with 25 active electrodes independently driven by a stimulation microchip just left of the array

1.5 Experimental Studies

Early studies showed that in the cat cortical activation can be recorded as a result of stimulating indwelling electrodes [13]. Although in this study it was shown that cortical activation was achieved over a considerable follow-up of 6 months, this effort did not end up with a retinal prosthesis available for use in humans. In research initiatives in the USA and Germany, experimental data have been collected which show that retinal implants can restore visual perception in blind humans suffering from RP. The first studies concentrated on the biocompatibility of implanted materials and on surgical feasibility.

1.5.1 Biocompatibility of Implanted Materials

In a series of in vitro experiments, neural cells and connective tissue cells as well as retinal cell cultures were exposed to material specimens of implant and encapsulation components. The cells were also exposed to basic substances for electronic components such as silicon and gold [23]. It could be shown that some silicones were toxic to these cells whereas others were not. Polydimethylsiloxane (PDMS), which is also known as a standard material for the fabrication of intraocular lenses, proved to be non-toxic in these studies. Electrically inactive components of the devices were implanted into the eye of pigmented rabbits (Fig. 1.5). It was shown that the components were well tolerated even 6 months after implantation [2].

1.5.2 Surgical Feasibility

In rabbits and pigs it was demonstrated that subretinal implants can be inserted either after vitrectomy through a retinotomy in the subretinal space or by an external approach through an incision through the choroid (Fig. 1.6). Complications such as retinal detachments or choroidal bleeding were rarely reported. The retina overlying the implant could be preserved if the implant was very thin and perforated. With the subretinal position of the implant a specific fixation procedure was not necessary. The implant showed a stable position over a period of more than 1 year [40].

Chow's group did not report any complications occurring during or after the implantation of their subretinal device in patients.

The implantation of epiretinal devices is more complex because a fixation procedure has to be followed as well as a procedure for implantation of the receiver part of the implant. Walter and co-workers as well as Majji and deJuan's group independently applied tack fixation to stabilize the microelectrode array onto the reti-

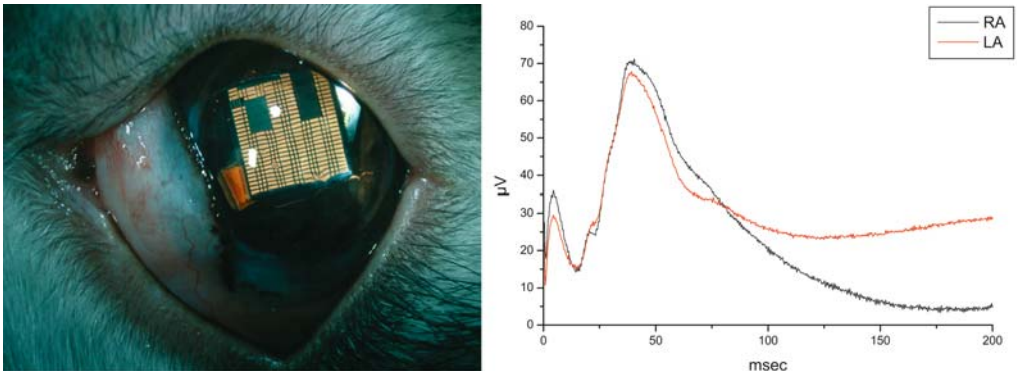

Fig. 1.5. *Left* Electrical passive silicon structure encapsulated in PDMS 6 months after implantation in the capsular bag of a rabbit. *Right* Same rabbit, 6 months after implantation. Electroretinogram: *red* left eye (control); *black* right eye (study eye after implantation)

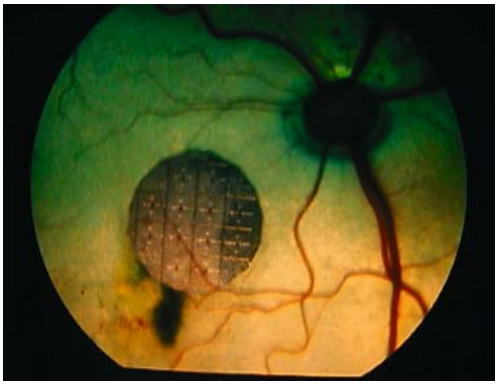

Fig. 1.6. Subretinal implant 12 months after transvitreal implantation in the subretinal space of a pig

nal surface. They found that with the use of retinal tacks (Fig. 1.7) a stable position of the implant could be achieved in over 6 months and that the complication rate was low [32, 46]. The complete removal of the vitreous in the rabbit is much more complex than in man. The adherence of the vitreous to the retina is stronger and therefore in our series we performed a two-step approach. In the first step a core vitrectomy with endolaser of the prospective fixation area was performed. Two weeks later in a second vitrectomy vitreous remnants were removed and the implant was inserted and tack fixated.

Alternatives to this approach use enzyme assisted vitrectomy. Plasmin or tPA was used in these procedures to separate the posterior vitreous from the retinal surface. It is mandatory in the epiretinal approach that the stimulating electrodes are placed as close as possible to the ganglion cell layer. Tissue or any material in between the stimulator and the target cells will reduce the effectivity of stimulation. Under such circumstances the resistance will increase and therefore much more energy is necessary for an effective stimulation. In terms of long-term biocompatibility the currents for stimulation should be as small as possible. Currently it is unknown if in chronic experiments tissue will grow in between the stimulator and the retinal surface. It could be expected that if the primary contact between the retina and the stimulator is very good, the ingrowth of fibrous tissue will be limited. Further experiments are necessary to demonstrate the behaviour of the interface between the implant and the retinal surface.

In a number of experiments it was demonstrated that tack fixated stimulators could also be explanted [4]. Explantation of microelectrode arrays could be less traumatic when biological fixation procedures are available. The explantation of tack fixated microelectrode arrays is comparable to trauma surgery and is characterized by the use of perfluorcarbon liquids, endolaser and silicone oil. Improvements in the design of retinal tacks are therefore desirable.

In 2003 the German EPI-RET consortium fabricated the first functional wireless epiretinal

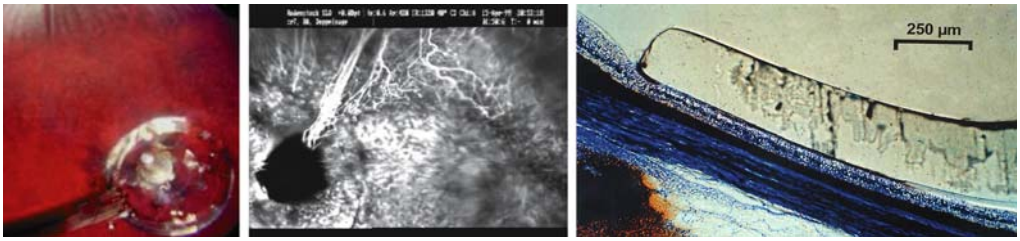

Fig. 1.7. *Left* Fundus photograph of a tack fixated microelectrode array onto the retinal surface of a rabbit 6 months after implantation; *centre* angiogram 6 months after implantation showing no neovascular elements only a slight deviation of a vessel towards the tack; *right* histology after tack fixation of an epiretinal contact array on the retinal surface. Grinding technique

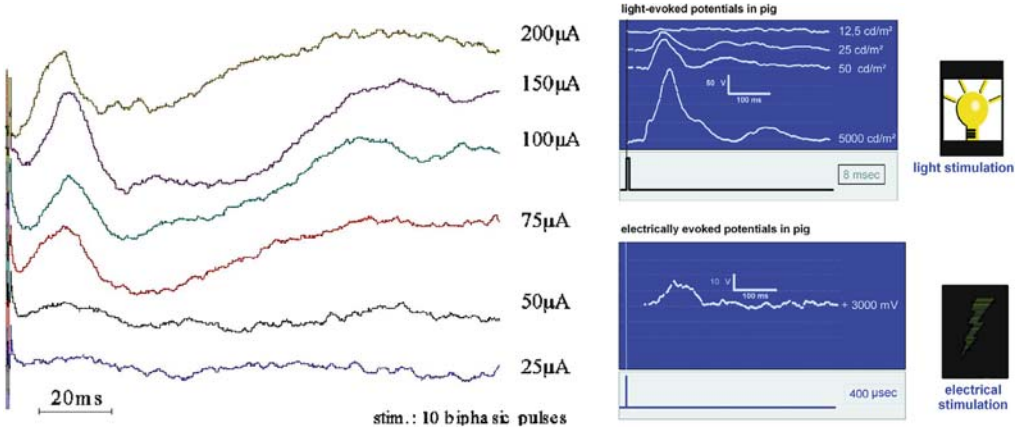

Fig. 1.8. *Left* Electrically evoked cortical potentials in the rabbit after pulse train stimulation of the inner retinal surface with repetitive short biphasic current pulses; *right* light evoked (*top*) and electrically evoked cortical potentials in the pig after subretinal electrical stimulation

prosthesis with 25 electrodes and a transponder system for data and energy. After developing procedures for the implantation of such complex prostheses in rabbits the system was implanted in cats and pigs to study cortical activation. The lens was removed with a standard phacoemulsification procedure. The vitreous was removed with vitrectomy, and the eye was filled with perfluordecalin. The posterior capsula was opened and the implant inserted through a corneal incision. The receiver was placed in the capsular bag, the stimulator was positioned on the decalin surface after pushing it through the posterior opening of the capsula and with removal of decalin it was placed on the retinal surface. Then with the use of a retinal tack it was fixed onto the retinal surface in or close to the area centralis.

1.5.3 Studies on Cortical Activation

Functional studies on cortical activation have been performed in rabbits, cats, pigs, but also in humans (Fig. 1.8). In early approaches acute experiments were performed in rabbits in which the cortical activation was demonstrated by recordings of evoked potentials (EPs) as a result of retinal stimulation achieved by electrode arrays directly connected to a stimulator device. Thresholds for the detection of EPs after epiretinal stimulation were found at 30 µA with repeated biphasic pulse trains of ten pulses (1 ms for each pulse) [45]. For subretinal stimulation, Zrenner and his group and Chow and co-work-

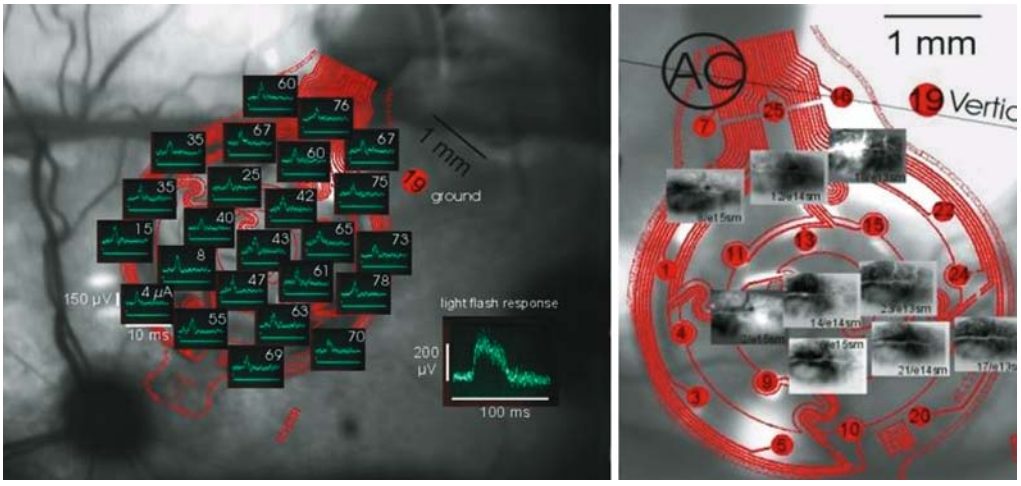

Fig. 1.9. *Left* Optic tract field potentials of the cat after local electrical stimulation of the inner surface of the retina; *right* corresponding optical imaging data representing intrinsic evoked activity of the primary visual cortex where *dark areas* represent an increase in metabolic activity

ers found evoked potentials similar to EPs evoked by light [9, 40].

In the cat cortical activation experiments were performed by Eckhorn and co-workers and by Eysel and his group. Eckhorn analysed field potentials recorded with multiple electrodes directly from the visual cortex whereas Eysel detected cortical activation by optical imaging indicating local oxygen consumption (Fig. 1.9).

With both experimental setups it could be demonstrated that either with subretinal electrodes or with epiretinal electrodes a specific local activation of the visual cortex could be achieved. It could be shown that moving the retinal stimulation electrodes resulted in an activation of a different cortical area. These experiments helped to estimate the range of potential visual acuity which could be achieved by the different approaches. From the data obtained with these experiments a potential visual acuity of about 20/200 was estimated [20, 38, 39].

Summary for the Clinician

- **Biocompatibility studies showed no serious adverse reactions**
- **Surgical procedures for subretinal and epiretinal implantations have been developed**
- **The implantations were performed successfully in rabbits, cats, and pigs**
- **Cortical activation has been demonstrated after epiretinal and after subretinal stimulation in rabbits, cats, and pigs using evoked potential techniques, optical imaging of intrinsic metabolic signals of the visual cortex, and recordings of local cortical field potentials**
- **First prototypes were implanted in humans suffering from RP. The implants were well tolerated and localized visual percepts were elicited**

1.6 Clinical Studies

In the USA acute experiments in humans were performed to demonstrate whether blind subjects may have visual perception after electrical stimulation. In these experiments performed by Humayun and by Rizzo it was shown that visual percepts can be elicited with direct stimulation of the retinal surface. Rizzo reported on perception with charge densities below 1 mC/cm² [37] and in the Humayun series thresholds for charge densities were between 0.16 and 56 mC/cm² [26]. Several factors were identified

influencing the efficacy of stimulation. The most important factors were electrode size and the timing of the stimulation pulses.

Currently studies are performed with chronic epiretinal implants using the cochlea implant approach for signal and energy transfer. In that approach an electrode array is positioned onto the retinal surface using retinal tacks. This array is connected to a cable entering the eye through the sclera with a receiver for signal and energy positioned subcutaneously [25]. The authors reported that in both subjects who were implanted visual perception could be obtained, that surgery was performed without any complications and that the implant was well tolerated. Experiments with subretinal implants were performed by Chow and his group. They found in patients with retinitis pigmentosa that subretinal implants survived for more than 2 years and that patients reported an increase in vision [10].

1.7 Outlook and Perspectives

The last 10 years of visual prosthesis research has demonstrated that the interdisciplinary approach of combining engineering and medical know-how has an important contribution to make in a field where no treatment is available for blind patients. Several research groups have developed prototypes for retinal implants which in the future will be evaluated in terms of safety and efficacy. With the first generation of these implants it can be expected that visual perception will be elicited in blind subjects. The quality of vision or visual acuity or visual field properties will depend on a number of factors of the individual patient and not only of the technical implant. However, even if the first implants will restore only minor visual functions such as light detection or identification of large objects, further improvements in technology will make a visual acuity of 20/200 realistic. A large number of open questions remain, the most important being the long-term stability and function of such a device, the biological behaviour of the interface between the electrodes and the target tissue, and the target tissue itself. At present it is not known how in the retina such a coupling between a technical device and the retina itself will function over many years. These questions can only be answered in human clinical trials in the future.

1.8 Summary

Retinal implants have been designed and fabricated to restore vision in blind RP patients. These implants work by activating neural retinal cells not affected by the degenerative process with an electrode array placed onto (epiretinal approach) or underneath (subretinal approach) the retina. Surgical procedures have been developed for both types of visual prosthesis. Animal experiments have shown that local activation of the visual cortex can be achieved with stimulation currents and charge transfers which are in a biocompatible range. Initial clinical trials in blind human subjects have shown that visual percepts can be achieved. It is hoped that with this technical approach ambulatory vision can be restored in otherwise not treatable degenerative diseases of the retina.

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Therapeutic Approaches to Macular Oedema

2

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Core Messages

- Breakdown of the blood-retinal barrier occurs as a consequence of a variety of conditions such as metabolic alterations, ischaemia, hydrostatic and mechanical forces, or inflammation
- Laser treatment of macular oedema is controversial for diffuse oedema and is not indicated in ischaemic forms
- Surgical internal limiting membrane (ILM) peeling is thought to lower the tractional forces and enhance the diffusion of substances from the retina into the vitreous cavity
- Intravitreal triamcinolone is used successfully to reduce macular oedema despite ongoing discussions about formulation and dosage. However, controlled prospective clinical trials are required to investigate its efficacy in restoring or maintaining visual function
- The search for a specific pharmacological treatment is ongoing on the basis of new findings regarding the involvement of cytokines and growth factors in the formation of macular oedema. Vascular endothelial growth factor (VEGF) inhibitors are currently being investigated in clinical studies

2.1 Introduction

As laser treatment is not applicable to all forms of macular oedema and frequently remains unsatisfactory, there is a significant demand for more efficient options to reduce macular oedema, either pharmacologically and/or surgically. This chapter reviews current knowledge on the pathogenesis of macula oedema, and discusses the rationale and efficacy of surgical approaches to macular oedema.

2.2 Macular Oedema as a Result of Various Disease Mechanisms

2.2.1 Causes of Macular Oedema

Macular oedema is a common phenomenon in various diseases where fluid accumulates in between the retinal cells. Both the focal and diffuse as well as the cystic form are characterized by extracellular accumulation of fluid, specifically in Henle's layer and the inner nuclear layer of the retina. The compartmentalization of the accumulated fluid is likely to be due in part to the relative barrier properties of the inner and outer plexiform layers. The fluid originates from the intravascular compartment.

The classic pattern of cystoid macular oedema with a petalloid appearance originating from the fluorescein leakage from perifoveal capillaries may be seen in cases of advanced oedema of various origins. This includes postsurgical cystoid macular oedema as well as cystoid oedema

Table 2.1. Causes of macular oedema in relation to the underlying disorders

Disease group	Disorder	Pathogenesis
Metabolic alterations	Diabetes	• Abnormal glucose metabolism • Aldose reductase
	Retinitis pigmentosa	• CME: leakage at the level of RPE
	Inherited CME (aut. dom.)	• Müller cell disease: leakage from perifoveolar capillaries
Ischaemia	• Vein occulsion • Diabetic retinopathy	• Inner blood-retinal barrier (retinal capillary hypoperfusion)
	• Severe hypertensive retinopathy • HELLP syndrome • Vasculitis, collagenosis	• Outer blood-retinal barrier (ischaemic hypoperfusion of the choroid: serous detachment)
Hydrostatic forces	Retinal vascular occlusions • Venous occulsion • Arterial hypertension • Low IOP	• Increased intravascular pressure • Failure of the BRB
Mechanical forces	Vitreous traction on the macula	• Epiretinal membranes with tangential traction • Vitreomacular traction syndrome
Inflammation	Intermediate uveitis	• Mediated by prostaglandins • CME is indication for treatment
	Postoperative CME	• Perivascular leucocytic infiltrates
	Diabetic macular oedema	• Diabetic leucostasis mediates vascular leakage by endothelial cell apoptosis
	Choroidal inflammatory diseases	• Vogt-Koyanagi-Harada syndrome • Birdshot retinochoroidopathy
Pharmacotoxic effects	e.g. • Adrenaline (in aphakia) • Betaxolol • Latanoprost	• Mostly via prostaglandins

associated with one of the following conditions: diabetes, vascular occlusion, hypertensive retinopathy, epiretinal membranes, intraocular tumours (e.g. melanoma, choroidal haemangioma), intraocular inflammation (e.g. pars planitis), macroaneurysm, retinitis pigmentosa, choroidal neovascularization and radiation retinopathy.

Given the heterogeneous aetiology of macular oedema, its effective treatment depends upon a better understanding of its pathogenesis. In general, formation of macular oedema is related to metabolic changes, ischaemia, hydrostatic forces, and inflammatory and toxic mechanisms that influence the formation of macular oedema to various degrees in the different conditions (Table 2.1).

Metabolic alterations have a causal role in diabetic maculopathy, but also in inherited diseases such as the autosomal dominant form of macular oedema or macula oedema in retinitis pigmentosa. Furthermore, ischaemia of the inner or outer blood-retinal barrier leads to macular oedema. Decreased perfusion of the retinal capillaries is seen, e.g. in vein occlusion and diabetic retinopathy, whereas ischaemia plus decreased perfusion of the choroid with associated serous retinal detachment occurs in severe hypertensive retinopathy, in eclampsia or in rheumatoid disorders. Following retinal vascular occlusion the intravascular pressure increases and leads to dysfunction of the blood-retinal barrier. Similarly, hydrostatic forces are effec-

tive in arterial hypertension or in eyes with low intraocular pressure and may cause fluid accumulation in the macula. Mechanical traction such as in epiretinal membranes or in vitreomacular traction syndrome promotes macular oedema by physical forces.

The importance of inflammation for macular oedema is discussed in more detail below. Inflammation apparently plays a role in intermediate uveitis, postoperative cystoid macular oedema (Irvine-Gass syndrome), diabetic macular oedema and various forms of choroidal inflammatory diseases including Vogt-Koyanagi-Harada syndrome and birdshot retinochoroidopathy. All prostaglandin-like pharmacological agents, even if applied topically, can induce macular oedema via a cytokine response similar to inflammatory conditions.

The current therapy for macular oedema targets conditions where mechanical traction, hydrostatic force or inflammation play a pathogenetic role in the formation of macular oedema. Unfortunately, even the currently available surgical and pharmacological treatments have suboptimal results in many cases. Therefore, there is an obvious need for the development of a more effective and targeted treatment that can be satisfied only by a better understanding of the pathophysiology of macular oedema formation, which differs according to the underlying disease.

2.2.2 Molecular and Cellular Alterations Leading to Macular Oedema

The breakdown of the blood-retinal barrier seems to be the most important mechanism in explaining the extravasation of fluid although similary changes to the retinal blood flow may play a role [5]. The blood-retinal barrier consists of the retinal pigment epithelium layer (outer blood-retinal barrier) and the vascular endothelium (inner blood-retinal barrier) that prohibit the passage of macromolecules and circulating cells from the vascular compartment to the extracellular and therefore intraretinal space.

In general, an increase in passive permeability through the endothelium can occur via three general mechanisms (Fig. 2.1):

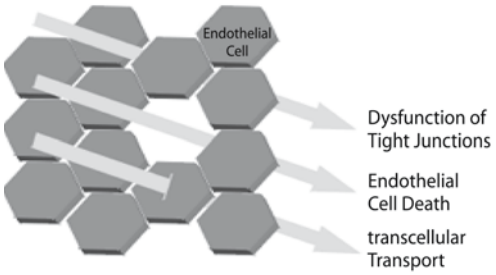

Fig. 2.1. Three mechanisms of vascular leakage

- Dysfunction of the intercellular junctions
- Increased transcellular transport
- Increased endothelial cell destruction

The initial site of damage that results in the increased vascular permeability has so far been controversial. Impairment of the perivascular supporting cells such as pericytes and glial cells might play a role, however, the endothelial cell dysfunction and injury seem more likely to be the first pathogenetic step towards the breakdown of the blood-retinal barrier early in the course of the disease. In order to dissect the molecular and pathophysiologic mechanisms that lead to the accumulation of fluid in the macular area, we have chosen diabetic macular oedema as a model.

2.2.2.1 Cell-to-Cell Junctions and Vascular Permeability

Fluid homeostasis and endothelial permeability are mostly regulated by intercellular junctions in the non-diseased retina. Intercellular junctions are complex structures formed by the assembly of a transmembranous and cytoplasmic/cytoskeletal protein component. At least four different types of endothelial junction have been described: tight junctions, gap junctions, adherence junctions and syndesmosis. Tight junctions are the most apical component of the intercellular cleft (Fig. 2.2).

Although the molecular structure of tight junctions generally appears to be similar in all barrier systems, there are some differences between epithelial and endothelial tight junctions,

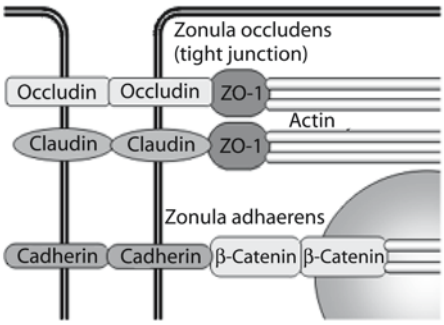

Fig. 2.2. Tight junctions in endothelial cells

and between tight junctions of peripheral and retinal endothelial cells [118]. In contrast to tight junctions in epithelial systems, structural and functional characteristics of tight junctions in endothelial cells respond promptly to ambient factors. It is likely that inflammatory agents increase permeability by binding to specific receptors that transduce intercellular signals, which in turn cause cytoskeletal reorganization widening of the interendothelial clefts. Endothelial junctions also regulate leucocyte extravasation. Once leukocytes have adhered to the endothelium, a coordinated opening of interendothelial cell junctions occurs.

2.2.2.2 Cellular Interaction and Vascular Permeability

Leukocytic infiltration of the retinal tissue characterizes many inflammatory diseases such as diabetes, pars planitis, and choroidal inflammatory diseases. In diabetes, activated leukocytes adhere to the retinal vascular endothelium [94, 117]. Increased leukostasis is one of the first histologic changes in diabetic retinopathy and occurs prior to any apparent clinical pathology.

Adherent leukocytes play a crucial role in diabetic retinopathy by directly inducing endothelial cell death in the capillaries [71], causing vascular obstruction and vascular leakage. Endothelial cell death precedes the formation of acellular capillaries [117]. With time, however, acellular capillaries prevail and become widespread. Although the mechanism of this destructive process remains elusive, it is clear that the interaction between the altered leukocytes and the endothelial cells and the subsequent endothelial damage represents a crucial pathogenic step [71, 76, 94].

2.2.2.3 Growth Factors and Vascular Permeability

The disruption of endothelial integrity leads to retinal ischaemia and vascular endothelial growth factor (VEGF)-mediated iris and retinal neovascularization [74, 94, 96]. VEGF is 50,000 times more potent than histamine in causing vascular permeability [10, 21, 22, 36, 77, 120]. Previous work has shown that retinal VEGF levels correlate with diabetic blood-retinal barrier breakdown in rodents [31] and humans [4]. Flt-1(1–3Ig)F_c, a soluble VEGF receptor, reverses early diabetic blood-retinal barrier breakdown and diabetic leukostasis in a dose-dependent manner [72]. Early blood-retinal barrier breakdown localizes, in part, to retinal venules and capillaries of the superficial inner retinal circulation [109, 71] and can be sufficiently reduced by VEGF inhibition. Although VEGF is only one of the cytokines involved in the pathogenesis of the vascular leakage, it is likely to be one of the most effective therapeutic targets.

2.2.2.4 Endothelial Cell Death and Vascular Permeability

Blood-retinal barrier breakdown is at least in part due to endothelial cell damage and apoptosis. The proapoptotic molecule Fas-ligand (FasL) induces apoptosis in cells that carry its receptor Fas (CD 95) [19]. There is evidence that FasL is expressed on vascular endothelium, where it functions to inhibit leukocyte extravasation. The expression of FasL on vascular endothelial cells might thus prevent detrimental inflammation by inducing apoptosis in leukocytes as they attempt to enter the vessel. In fact, during inflammation and ensuing tumor necrosis factor alpha (TNF-α) release, the retinal endothelium upregulates several adhesion molecules [131] that mediate the adherence of the leukocytes, but also downregulate FasL, thus allowing the leukocyte survival and migration to active sites

of inflammation and infection. In experimental diabetic retinopathy, inhibition of Fas-mediated apoptotic cell death reduces vascular leakage [75]. The cumulative endothelial cell death during the course of diabetes plays a causal role in the pathogenesis of the diabetic vascular leakage and maculopathy.

2.2.2.5 Extracellular Matrix Alterations and Vascular Permeability

Degradation of the extracellular matrix affects endothelial cell function at many levels, causing endothelial cell lability, which is required for cellular invasion and proliferation, or influencing the cellular resistance and therefore the vascular permeability. The degradation and modulation of the extracellular matrix is performed by matrix metalloproteinases, a family of zinc-binding, calcium-dependent enzymes [27, 88]. Elevation of MMP-9 and MMP-2 expression has been shown in diabetic neovascular membranes [25, 116], although a direct effect of glucose on MMP-9 expression in vascular endothelial cells could not be shown [53]. It is likely that MMPs participate at various stages during the course of the blood-retinal barrier dysfunction and breakdown. Their actions include early changes in the endothelial cell resistance with influence on the intercellular junction formation and function [35]. Further, they actively participate in the endothelial and pericyte cell death [9] that occurs late in the course of the disease.

2.2.2.6 Transcellular Transport and Vascular Permeability

In addition to all the above, an important factor that is involved in the regulation of fluid homeostasis is the active cellular transport of nutrients and fluid via pinocytosis. Despite the fact that pinocytic transport is critically involved in the transepithelial fluid exchange, its role in the pathogenesis of increased vascular leakage in diabetes is just emerging [2, 40]. Still, the molecular factors that are involved in the pinocytic fluid transport, how they are influenced from disease stages and how they contribute to the increased vascular permeability are unclear.

It is currently known that one of the factors involved in the regulation of pinocytic transport is VEGF, which increases vascular permeability not only by disrupting the intercellular tight junctions between the retinal endothelial cells but also by inducing the formation of fenestrations and vesiculovacuolar organelles. The role of VEGF in the disruption of the pinocytic transport that is translated into increased vascular permeability in disease states is still controversial [61]. Whereas in highly permeable blood vessels the number of pinocytotic vesicles at the endothelial luminal membrane transporting plasma IgG is significantly increased, no fenestrations or vesicles have been found in the endothelial cells of the VEGF affected eyes when examined by electron microscopy.

Knowledge of the basic mechanisms involved in vascular leakage is essential for the development of an effective clinical treatment. With the growing understanding of the pathophysiology of the macular oedema, the therapeutic thinking is likely to change from a merely symptomatic treatment (either surgical or medical) to a treatment that targets specifically the causal factors involved in its formation (e.g. cytokine or growth factor inhibition).

Summary for the Clinician

- **Macular oedema is a common phenomenon in different diseases resulting from either metabolic alterations, ischaemia, hydrostatic and mechanical forces, inflammation, pharmacotoxic effects, or a combination of these**
- **Blood-retinal barrier breakdown may occur to a variable extent via dysfunction of intercellular junction, increased transcellular transport, or increased endothelial cell destruction**
- **Among the growth factors involved, vascular endothelial growth factor plays a dominant role as a mediator of vascular leakage**
- **Inflammatory phenomena are causally linked to vascular cell death and leakage**

2.3 Treatment of Macular Oedema

In an effort to reduce macular oedema, at least with some rationale, different approaches have been used and found effective in certain conditions. Laser coagulation, pharmacological approaches, and surgical measures are most frequently used.

2.3.1 Laser Treatment

Laser therapy is well established in diabetic macular oedema as well as macular oedema secondary to retinal vein occlusion. While in cases related to vessel obstruction the treatment aims to prevent the growth factor release from the ischaemic peripheral areas that could alter vascular permeability, in diabetic macular oedema a different approach is followed. The 3-year risk of massive visual loss from macular oedema without focal laser treatment is about 30 %, compared to 15 % after focal laser treatment [29, 30]. Interestingly, scatter (panretinal) laser coagulation was not effective, but may even be deleterious.

Prophylactic treatment of a non-significant macular oedema is not advantageous over no treatment. Prophylactic laser coagulation is therefore not justified [3, 29, 62]. Laser coagulation of diabetic macular oedema should only be considered when the oedema is clinically significant (CSME). CSME as defined by ETDRS includes any one of the following lesions:

- Retinal thickening at or within 500 µm from the centre of the macula
- Hard exudates at or within 500 µm from the centre of the macula, or there is thickening of the adjacent retina
- An area or areas of retinal thickening at least one disc area in size, at least part of which is within one disc diameter of the centre of the macula

Focal laser coagulation reduces hypoxic areas and directly occludes leaky microaneurysms. The rationale for grid laser treatment in diffuse macular oedema is not yet well established. Grid laser treatment is believed to enhance proliferation of retinal pigment epithelial and endothelial cells, leading to a repair of the blood-retinal barrier [130]. Another theory suggests a reduction of oxygen consumption of the whole tissue due to the destruction of photoreceptors [14, 133]. Still others believe that laser induced necrosis stimulates the release of factors which stabilize the blood-retinal barrier. Moreover, the grid laser may have its effect by thinning the retina, bringing retinal vessels closer to the choroidal vessels, and permitting the retinal vessels to constrict by autoregulation, thereby decreasing retinal blood flow and consequently decreasing oedema formation [136].

Currently there is no confirmed evidence that grid laser treatment improves diffuse diabetic macular oedema (Table 2.2). Olk and coworkers demonstrated in a randomized study of 303 eyes that 3 years after grid laser treatment 14.5 % of the treated eyes improved by 2 lines or more, 60.9 % did not change significantly and 24.6 % deteriorated by more than 2 lines. The results, however, were not compared with a non-treated control group [100, 101]. Data from other studies showed similar results. The comparability of the different studies is limited due to the different criteria of inclusion, exclusion, monitoring and treatment. Despite the lack of functional improvement (visual acuity) there is a reduction of retinal thickness (anatomical oedema) after grid laser treatment as shown in several studies [82, 101].

Grid laser photocoagulation has also been described as a method of treatment in pilot studies on postsurgical cystoid macular oedema. However, confirmation through randomized controlled studies is still lacking [57, 106].

In the macular oedema caused by retinal vein occlusion, treatment with a grid laser is generally considered beneficial when the perfused macular oedema causes visual impairment to the level of 20/40 or worse or shows no signs of spontaneous improvement [12].

Both in diabetic macular oedema and in macular oedema secondary to branch vein occlusion, central laser surgery is not recommended in eyes with ischaemic maculopathy ([29], Table 2.3).

Table 2.2. Published studies investigating the efficacy of grid laser treatment in diabetic diffuse macular oedema [worse ≥ deterioration >2 lines; better ≥2 lines better (given as percentages)]

Studies		Patients	Eyes treated	Eyes control	Observation period	V/A after grid laser			Untreated			
						Better	Un-changed	Worse	Better	Un changed	Worse	
ETDRS	Prosp. randomized	No separate evaluation for focal and diffuse macular oedema										
Olk	Prosp. randomized		42	37	24 months	45.2	45.2	9.5	8.1	48.5	43.2	Sign
Lee	Retrospective		302		36 months	14.5	60.9	9.5				
Ladas	Prosp. randomized		26	23	24 months	15.4	65.4	19.2	0	56.5	43.5	p=0.049
			24	22	36 months	8.3	54.2	37.5	0	40.9	59.1	ns
Wiznia	Prosp. randomized		35	35	17 months	22.9	0.6	17.1	5.7	54.3	0.4	ns
McNaught	Prosp. randomized		37	35	6 months (distant and near V/A)	72	14	14	17	66	17	p<0.001
			18	17	24 months (only near V/A)	61	11	28	18	47	35	ns
Tachi	Retrospective	32	41		12 months	12	39	49				
McDonald	Retrospective		89	0	3–33 months	17	77	6				
Bailey	Prospective	33			9 months			28.9				

Table 2.3. Grading of the foval avascular zone (in fluorescein angiograms). [Early Treatment Diabetic Retinopathy Study Research Group (1991) Classification of Diabetic Retinopathy from Fluorescein Angiography. ETDRS Report No. 19; Ophthalmology 98:807–822]

Grade 0	<300 µm
Grade 1	=300 µm
Grade 2	>300 µm and <500 µm
Grade 3	>or =500 µm
Grade 4	Cannot grade

For other causes of macular oedema, especially inflammation- or mechanical traction-related, laser treatment is not recommended. Grid laser treatment of postsurgical macular oedema has not been investigated in the setting of a controlled clinical trial, and so there is no information available regarding its efficacy or safety.

Although definite clinical data are lacking, the use of grid macular laser treatment in the elderly population has additional drawbacks as it may induce or accelerate RPE atrophy in the macular region.

In non-ischaemic vascular occlusions, "re-routing" from the retinal to the choroidal vasculature has been postulated [34, 37, 89] through laser photocoagulation. However, since the first report in 1995, re-routing strategies have not been confirmed.

2.3.2 Medical Treatment

Medical treatment of macular oedema is best established in postsurgical and predominantly inflammatory oedema, e.g. in uveitis. The majority of therapeutic strategies inhibit the release of inflammatory mediators and therefore target the pathogenetic factors responsible for the altered vascular permeability. The remaining treatments are symptomatic and include carbonic anhydrase inhibitors and methods that increase blood flow and oxygenation (e.g. hyperbaric oxygenation, diuresis and dialysis [15, 79, 80, 85, 93, 107, 125, 128]).

The medical treatment consists of therapeutic agents that are collectively categorized into three groups: corticosteroids, cyclooxygenase inhibitors and carboanhydrase inhibitors. Application modalities based on the current clinical treatment are listed in Table 2.4.

Table 2.4. Therapeutic agents in the medical treatment of macular oedema

Corticosteroids	Topical	Prednisolone actetate 1%	Four times daily
		Prednisolone sodium acetate 1%	
		Dexamethasone 0.1%	
	Peribulbar	Triamcinolone	20 mg (0.5 ml) every 3–6 weeks
		Methylprednisolone (Depo-Medrol)	20 mg every 3–6 weeks
	Oral	Prednisolone	1–1/5 mg/kg daily
	Intravitreal	Triamcinolone	4 (up to 25) mg (can be repeated)
Cyclooxygenase inhibitors	Topical	Diclofenac sodium 0.1% (Voltaren)	Four times daily
		Flubiprofen sodium 0.03% (Ocufen)	Four times daily
		Ketorolac tromethamine 0.5% (Acular)	Four times daily
Carbonic anhydrase inhibitors	Oral	Acetazolamide (Diamox)	500 mg dialy

2.3.2.1 Carbonic Anydrase Inhibitors

The rationale of carbonic anhydrase inhibitors as a therapeutic agent in the treatment of macular oedema is to improve the ability of the retinal pigment epithelial cells (RPE cells) to pump fluid out of the retina [85]. Currently, there are no available randomized studies that confirm a beneficial effect of carbonic anhydrase inhibitors in the treatment of macular oedema. Non-randomized observations demonstrated improved visual function in patients with postsurgical macular oedema, e.g. after cataract surgery or buckling procedures [24, 132]. The effect lasts only as long as the patient takes the drug (on-off effect, tachyphylaxis) [132]. The favourable reports that were described at first regarding the application of carbonic anhydrase inhibition in patients with macular oedema secondary to retinitis pigmentosa are not supported by long-term observations. With the continuous use of methazolamide, a rebound phenomenon is observed [38, 39].

In contrast to the tenacious clinical habit, the use of carbonic anhydrase inhibition for macular oedema is not based upon scientific evidence to date.

2.3.2.2 Non-steroidal Anti-inflammatory Drugs (NSAIDs)

As cyclooxygenase inhibitors (NSAIDs) block the synthesis and release of prostaglandins, non-steroidal drugs have been investigated in the prophylaxis and therapy of postsurgical cystoid macular oedema. It has been shown that topical administration achieves better ocular penetration than systemic administration [41].

In the prophylaxis of postsurgical cystoid macular oedema, oral indomethacin and piroxicam are of limited value. Although they reduce the angiographic leakage, they fail to improve visual acuity compared to untreated groups [90, 91, 92]. Even in the best cases the effect is a short-term one. A long-term follow-up study [92] demonstrated that patients treated prophylactically by topical indomethacin had improvement in visual acuity early after surgery; however, this effect did not last [140]. Similarly, the use of topical diclofenac [110] and keratolac [43] has been shown to decrease the incidence of angiographic cystoid macular oedema. A randomized, double-blind clinical trial demonstrated that flubiprofen-treated patients enjoyed an improvement in their visual acuity (20/40 or better) sooner than vehicle-treated patients. However, no significant difference between the groups could be found after day 121 of treatment [122]. The widespread usage of corticosteroids in the postoperative period and the possibility of synergistic effects [41] is a confounding factor that complicates the interpretation of the majority of the clinical trials. In clinically significant macular oedema, neither indomethacin [139] nor topical fenoprophen treatment [18] managed to show a statistically significant improvement in visual acuity. Two double-blinded, prospective randomized studies suggested that topical keratolac tromethamine 0.5% is effective for the treatment of chronic postsurgical macular oedema with visual impairment [42, 44]. However, the value of this study is limited by the short-term follow-up and the lack of comparison with topical corticosteroids. The effect of local treatment with NSAIDs on inflammatory cystoid macular oedema such as in uveitis has not been evaluated on a large scale [56, 112].

Systemic treatment with NSAIDs has been considered controversial for a long time. Rojas et al. [112] showed that systemic treatment with NSAIDs (orally) has an effect similar to that of local treatment with steroid periocular injections, when applied at least for several months.

There is no evidence for an effect of NSAIDs in macular oedema following vein occlusion.

In conclusion, the optimal dose of NSAIDs is not known. Oral NSAIDs penetrate poorly into the eye [1] and topical NSAIDs penetrate to varying degrees. It is clear that NSAIDS target the inflammatory mediators that are responsible for the oedema formation and although they may not be an optimal stand-alone treatment they can be used as steroid-sparing agents.

There may be several explanations as to why NSAIDs cannot improve vision. Chronic oedema, inflammation and ischaemia induce permanent structural alterations and damage to

the fovea. Müller cell degeneration, ischaemic damage, and RPE changes may contribute to the long-term visual loss. These limitations also apply to other treatment modalities.

2.3.2.3 Corticosteroids

Steroids are currently regaining attention from the growing use of intravitreal triamcinolone. Corticosteroids block the release of arachidonic acid from cell membranes and thus reduce the synthesis of prostaglandins. Furthermore, they inhibit the migration of leukocytes and the release of proinflammatory mediators such as TNF-α and VEGF. Steroids specifically stabilize endothelial tight junctions and increase their numbers [8, 113]. As discussed previously, this is especially important in the development of macular oedema.

Routes of administrations are manifold, including topical, periocular, oral, and intravenous routes. Subtenon injections of corticosteroids are widely used in patients with asymmetric or unilateral uveitis [141]. The advantages of the periocular injections are: high concentrations of corticosteroids in the posterior eye, and reduction of the adverse effects compared to systemic administration. Intraocular levels of corticosteroids are identical between subtenon and retrobulbar administration [126]. For oral administration, the initial high dose (1–1.5 mg/kg) is subsequently decreased according to clinical effect [26, 45, 112].

If macular oedema is a complication of vascular dysfunction, such as in diabetes, vein occlusion, or RPE dysfunction (retinitis pigmentosa), then the anti-oedematous effect of corticosteroids is uncertain. Recent publications suggest that the intravitreal application of triamcinolone seems to be a promising therapeutic method for macular oedema that fails to respond to conventional treatment [65, 86]. Martidis et al. [86] published a prospective, non-comparative, interventional case series to determine if intravitreal injection of triamcinolone acetonide is safe and effective in treating diabetic macular oedema unresponsive to prior laser photocoagulation [87]. Sixteen eyes with clinically significant diabetic macular oedema (CSME) that failed to respond to at least two previous sessions of laser photocoagulation were included in the study. The response of the laser treatment was measured by clinical examination and optical coherence tomography (OCT) at least 6 months after initial laser therapy. Eyes with a residual central macular thickness of more than 300 µm (normal, 200 µm) and visual loss from baseline were offered intravitreal injection of 4 mg triamcinolone acetonide. In this study, the mean improvement in visual acuity measured 2.4, 2.4, and 1.3 Snellen lines at the 1-, 3-, and 6-month follow-up intervals, respectively. The central macular thickness as measured by OCT decreased by 55 %, 57.5 %, and 38 %, respectively, over these same intervals from an initial pretreatment mean of 540.3 µm (±96.3 µm). Intraocular pressure exceeded 21 mmHg in five, three, and one eye(s), respectively, during these intervals. One eye exhibited cataract progression at 6 months. No other complications were noted over a mean follow-up of 6.2 months. Re-injection was performed in three of eight eyes after 6 months because of recurrence of macular oedema.

Similar pilot studies were performed in patients with uveitis, central vein occlusion, and cystoid macular oedema after cataract surgery [6, 54, 65, 142]. In most published reports, complications do not appear to be prohibitive; however, all reports demonstrate a limited number of selected cases.

Further randomized studies are therefore warranted to assess long-term efficacy and need for retreatment.

Reviewing the published data on intravitreal injections of triamcinolone acetonide, the therapeutic window seems very wide. The dose range of intravitreally injected triamcinolone acetonide varies from 2 mg [6] to 4 mg [87, 142] and even 25 mg in a single report [66]. Interestingly, reaccumulation of fluid in cystoid spaces occurs between 6 weeks and 3 months after injection, and this does not seem to be dose dependent. Repeated injections at intervals ranging from 10 weeks [67–70] to more than 6 months [6] show a variable treatment response.

There are currently no data on the pharmacokinetic profile of intravitreal triamcinolone,

which might be altered from a previous vitrectomy. Physiological intravitreal cortisol levels are reported to be 5.1 ng/ml, and vitreous levels after peribulbar injections are in the range of 13 mg/ml. The effective dose of the triamcinolone acetonide is further influenced by the mandatory washes of the widely used stabilizing agent benzylethanol during the preparation of the injection, so that even if standardized they alter the remaining amounts of the drug in the solution. Additionally, an inhibitory effect of the stabilizing agent on the drug cannot be excluded.

Jaffe and co-workers [63, 64] constructed a fluocinolone acetonide drug delivery device that releases fluocinolone acetonide in a linear manner over an extended period. A clinical phase III study by Bausch & Lomb investigated the efficacy of 0.5 mg (slow release) fluocinolone acetonide in 80 patients with diffuse diabetic macular oedema. Patients receiving the implant showed a statistically significant regression of retinal thickness after 6 months in comparison to the control group. Furthermore, 80 % of the eyes in the treatment group demonstrated a stable or improved visual acuity compared to only 50 % of the eyes in the control group.

Complications of intravitreal triamcinolone acetonide delivery systems comprise retinal detachment, vitreous haemorrhage, increased intraocular pressure, cataract formation, and pseudohypopyon. Elevation of the intraocular pressure after triamcinolone acetonide of more than 5 mmHg has been reported in up to 30 % of eyes [137]. It is therefore prudent that patients are asked about a history of a previous steroid response. The incidence of culture-positive endophthalmitis following intravitreal triamcinolone amounts to 0.87 % in a large, multicentre, retrospective case series [97]. It occurs rapidly (median 7.5 days) and can result in severe loss of vision and the eye. The risk of endophthalmitis is considerably higher compared to other intravitreally injected drugs. Engstrom and Holland reported the rate of endophthalmitis following intravitreal ganciclovir injection as 0.29 % (4 cases in 1,372 injections) [32]. The greater risk of endophthalmitis following intravitreal triamcinolone injection may be partly due to a small-size bias or an increased susceptibility to infections in diabetic individuals. Roth and co-workers reported about seven patients who developed a clinical picture simulating endophthalmitis following intravitreal triamcinolone injection [114]. Extensive signs of inflammation developed 1–2 days after injection, at an earlier time point than in bacterial endophthalmitis. Vitreous taps were sterile and inflammation resolved spontaneously with recovery to pre-injection visual acuity or better. This inflammatory response might be a response to the stabilizing additive benzylethanol (see above).

In any case, it is recommended to follow a sterile protocol for intravitreal injections of triamcinolone acetonide (see Table 2.5).

Table 2.5. Recommendations when performing intravitreal triamcinolone acetonide injections (modified after Moshfegi et al. 2003 [97])

- Strict adherence to a sterile technique with the use of a lid speculum
- If the patient is immunocompromised, consideration should be given to delaying the procedure
- Any external ocular inflammation/infection should be controlled prior to administering intravitreal triamcinolone acetonide injections
- Intravitreal triamcinolone acetonide injections should only be performed in the presence of a filtering bleb after careful consideration of the risk of endophthalmitis vs. the possibility of visual improvement
- Patients should be seen by an ophthalmologist at least once in the immediate postoperative period (1–7 days) after intravitreal triam cinolone acetonide injections
- Patients should be instructed to return to their ophthalmologist immediately at the first sign of any visual disturbance or pain
- Prompt intervention should be instituted, including obtaining a vitreous specimen for culture and administering intravitreal antibiotics

2.3.2.4 Antiangiogenic Treatment

VEGF is one of the most potent angiogenic growth factors to date. It is secreted by a variety of normal and cancer cells, and acts as an endothelial cell mitogen, and permeability factor, For this reason it was originally named vascular permeability factor (VPF). The induction of VEGF during the course of diabetes and several other inflammatory diseases is mediated by hypoxia, advanced glycation end products, reactive oxygen intermediates and growth factors such as TNF-α and insulin-like growth factor (IGF-I) [72, 108].

VEGF increases vascular permeability through specific binding to receptors on vascular endothelial cells that induce the disruption of the endothelial tight junctions with subsequent leakage of vascular fluid (see above: [7, 16]).

Intravitreous VEGF levels are elevated in diabetic maculopathy and retinopathy, and in hypoxia-mediated neovascularization (e.g. central vein occlusion). Recently aqueous levels of VEGF and IL-6 were found elevated in eyes with macular oedema following cataract surgery (Fanatsu 2003). There was a statistically significant correlation between the level of VEGF and severity of disease.

VEGF antagonists might be effective in diabetic macular oedema through inhibition of vascular leakage. There are currently two products undergoing phase III clinical trials in the treatment of neovascular age-related macular degeneration: the anti-VEGF-pegylated aptamer ("Macugen", Eye Tech Pharmaceuticals, Pfizer) and an anti-VEGF humanized neutralizing antibody ("Lucentis", Genentech, Novartis).

Both drugs need to be injected intravitreally every 6 weeks. Although severe side effects have not been reported so far, one would expect some risk of endophthalmitis similar to the intravitreal injection of steroids (see above).

The results of phase I studies have been promising: The investigators noted significant improvements in vision (three or more lines) in 25% of patients who received "Macugen" alone and in up to 60% of the patients who received Macugen and photodynamic therapy (PDT).

Similarly, positive preliminary data from a Phase Ib/II randomized, single-agent study with the investigational anti-VEGF product, rhuFab V2 (ranibizumab, "Lucentis"), for patients with the wet form of age-related macular degeneration (AMD) have been reported. Of the 53 patients treated with rhuFab V2, 50 patients (94%) had stable or improved vision, of whom 14 patients (26%) improved 15 letters or more on the ETDRS chart, and 36 patients (68%) had stable vision at day 98. The most common side effect of treatment with rhuFab V2 injection was mild, transient, reversible inflammation.

Currently there are ongoing studies in the USA and Europe to establish the safest and most efficacious dose of the VEGF Aptamer "Macugen" in patients with clinically significant macular oedema (CSME) involving the centre of the macula given intravitreally (0.3, 1.0 or 3.0 mg/eye) as compared to sham injections every 6 weeks for 12–30 weeks. The primary endpoint will be the retinal thickness measured by OCT. Secondary endpoints will include the mean best-corrected visual acuity, the need for focal/grid laser at 12 weeks or later, area of retinal thickening measured by photography and a fluorescein angiographic assessment of macular capillary leakage and cystoid spaces. Similar studies will be performed for rhuFab and sol-flt, a soluble receptor of VEGF.

Summary for the Clinician

- **Currently there is no confirmed evidence that grid laser treatment improves diffuse (diabetic) macular oedema**
- **In contrast to the tenacious clinical habit the use of carbonic anhydrase inhibition for macular oedema is not based upon scientific evidence**
- **Intravitreal triamcinolone acetonide is gaining attention for treatment of macular oedema not only as an additive during surgery or in the treatment of persistent macular oedema, but also as the primary treatment in diffuse macular oedema. Randomized multicentre studies are ongoing**
- **Clinical studies using VEGF inhibitors are currently performed with two specific molecules; however, phase III data are not yet available**

2.3.3 Surgical Approaches

The observation that patients with a diffuse diabetic macula oedema have a reduced incidence of posterior vitreous detachment has generated the idea that posterior vitreous detachment during vitrectomy could be used as a therapeutic approach in patients without macula oedema [98, 99]. Hikichi and co-workers report a 55% resolvement of diabetic macular oedema following posterior vitreous detachment. In contrast only a 25% resolvement of diabetic macular oedema was observed in the patients with attached hyaloid [60]. Peeling of the inner limiting membrane of the retina ensures complete release of tractional forces, removes a potential diffusional barrier, and inhibits reproliferation of fibrous astrocytes [51]. It is, however, still a matter of investigation whether such mechanical interventions effectively resolve macular oedema in the long run.

2.3.3.1 Pars Plana Vitrectomy with Posterior Vitreous Detachment

It has been demonstrated that a surgically induced posterior vitreous detachment in patients with a diffuse diabetic macular oedema leads to a reduction of macular oedema with a subsequent increase in visual acuity [51, 58, 84].

In the study by Pendergast and co-workers, 27 (49.1%) of 55 eyes demonstrated an improvement in best-corrected visual acuity of 2 or more lines. Fifty-two (94.5%) of the 55 vitrectomized eyes showed an improvement in clinically significant macular oedema, and in 45 eyes (81.8%) the macular oedema resolved completely during a mean period of 4.5 months (range 1–13 months). Eyes with macular ischaemia and preoperative best-corrected visual acuity of 20/200 or less tended to respond less favourably to vitrectomy than eyes lacking these characteristics. All eyes had at least 6 months of follow-up after surgery, with a mean follow-up of 23.2 months [105].

Similarly favourable results were found in macular oedema from central retinal vein occlusion, chronic uveitis, and postsurgical cystoid macular oedema [28, 78, 104, 119, 123, 135].

Although the exact mechanism of the observed beneficial effect is not known, removal of inflammatory mediators and better access of locally applied steroids are postulated mechanisms of actions of vitrectomy. Following favourable results of non-randomized studies in aphakic eyes [33, 48], a multicentre, randomized, prospective clinical trial [49, 50] demonstrated a visual benefit of vitrectomy in aphakic chronic macular oedema with vitreous incarceration in the wound. The authors recommend delaying vitrectomy until the cystoid macular oedema persists for at least 2–3 months [50]. Dugel and co-workers [28] report 11 eyes with persistent macular oedema unresponsive to steroids, seven of which improved by four lines or more after surgery.

2.3.3.2 Vitrectomy and Peeling of the Inner-Limiting Membrane

The rationale for vitrectomy (removal of the hyaloid) plus peeling of the internal limiting membrane is the postulated improvement of fluid diffusion from the retina to the vitreous cavity. Vitrectomy including removal of the internal limiting membrane (ILM) aids the resolution of diffuse diabetic macular oedema and improvement of visual acuity and prevents epiretinal membrane formation [51]. Advantage of the ILM peeling over the vitrectomy alone is the complete release of tractional forces and inhibition of reproliferation of fibrous astrocytes, which seems to be prudent in the eyes of patients with diabetes and advanced vitreoretinal interface disease of the macula.

Looking at the histological appearance of a cystic macular oedema as demonstrated schematically in Fig. 2.3, large bullous oedema can potentially be worsened after ILM peeling and proceed to a pseudo-macular hole. In this respect careful examination and the use of optical coherence tomography might help to exclude patients with large bullae.

Our own data suggest that ILM peeling is ineffective to resolve the macular oedema in central retinal vein occlusion (CRVO) and prolifer-

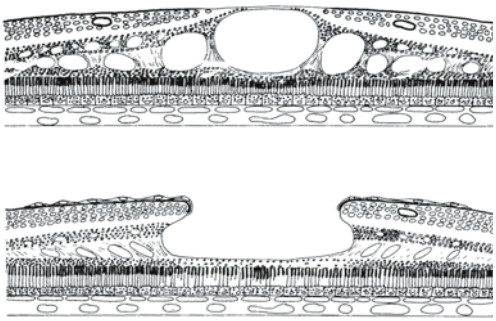

Fig. 2.3. Cystic macular oedema: pseudo-hole

ative diabetic retinopathy. Improvements were apparent only in non-proliferative diabetic retinopathy: A retrospective review analysed a series of 23 eyes from 23 patients with persistent macular oedema after pars plana vitrectomy (PPV) with indocyanine green (ICG) assisted peeling of the ILM. The main diagnoses were uveitis (anterior, intermedia, posterior and panuveitis) (n=9), CRVO (n=4), diabetic retinopathy (DR) (n=5), vitreoretinal traction syndrome (n=2), and Irvine-Gass syndrome (n=3). Visual acuity improved in 9 out of 23 patients after 3 months and in 6 out of 21 and in 7 out of 21 patients after 6 months. This improvement was predominantly seen in patients with uveitis (5/9) or diabetic maculopathy (3/5); one patient with Irvine-Gass syndrome showed a significant reduction, whereas one with vitreoretinal traction showed an improvement in visual acuity. The group of CRVOs showed no significant change during the follow-up. The use of endotamponade did not influence the outcome. Patients with uveitis and non-proliferative diabetic maculopathy demonstrated a transient benefit. The lack of long-term improvement is in accordance with the hypothesis that ILM peeling cannot not interfere with the mechanism of macular oedema [110a].

Long-term data on the effectiveness of ILM peeling for resolution of macular oedema are not available. Additional randomized studies have to be performed that should include data on reading performance and retinal thickness to better correlate anatomical alterations to clinically relevant functional parameters.

2.3.3.3. Pars Plana Vitrectomy with Sheathotomy and Radial Optic Neurotomy

Recently sheathotomy, surgical separation of the arteriovenous adventitial sheath, has been proposed for cystoid macular oedema (CME) in branch vein occlusion. First reported by Osterloh and Charles [103], sheathotomy is performed preferably on first and second order arterioles. Several reports demonstrate that sheathotomy is feasible [46, 83, 102, 121]. However, the literature must be interpreted with extreme caution due to the large variation of the natural course of vein occlusions and the lack of a control group.

Radial optic neurotomy, a stab incision to the papilla down to the lamina cribrosa, is an attempt at a decompression of the central retinal vein after central retinal vein occlusion [52]. The improvement might be attributed to the optic nerve decompression or the vitrectomy alone. Possibly, the resolution of retinal oedema is accelerated by inducing chorioretinal shunts that drain retinal circulation to the choroids [115]. Complications include retinal detachment and visual field defects. As for the sheathotomy, randomized trials are needed to evaluate the potential long-term benefit of radial optic neurotomy in vein occlusion.

2.3.4 Modification of Systemic Blood Flow

Hyperbaric oxygen is thought to alter the blood flow via vasoconstriction, and to facilitate the reformation of damaged junctional complexes in the vessel wall. Thus, hyperbaric oxygen works predominantly at the level of the inner blood-retinal barrier. Intermittent hyperbaric oxygen for 21 days showed an improvement in visual acuity in patients with chronic CME after cataract extraction [107]. The improvement in visual acuity, however, does not correlate with a reduction of macular oedema. It is possible that hyperbaric oxygen alters macular ischaemia or affects the anterior segment of the eye. In uveitis-associated CME, hyperbaric oxygen had no significant effect [80, 93].

Other rheological treatments such as plasma membrane filtration demonstrated good effects in initial studies; however, they have not entered large-scale prospective studies [134].

Summary for the Clinician

- **Vitrectomy without ILM peeling might be beneficial in patients with a taut posterior hyaloid lacking a posterior vitreous detachment**
- **ILM peeling is thought to act through the removal of a diffusion barrier and the prevention of epiretinal membrane formation**
- **Sheathotomy (surgical separation of the arteriovenous adventitial sheath) is a potential option for recent branch vein occlusion. As with radial optic neurotomy, the treatment needs to be evaluated in randomized studies**
- **Hyperbaric oxygenation and plasma membrane filtration might be indicated in selected cases**

2.4 Discussion: Open Questions and Technical Aspects

Most of the presented data for the surgical methods as well as pharmacological treatments represent small case series. In order to further evaluate the discussed treatment approaches, randomized prospective studies in a large population are needed. However, not only the endpoint criteria and the measurement approaches should be evaluated, but also the best time point for treatment or the question of whether to treat ischaemic forms of macular oedema and how.

Many studies presenting positive treatment results use the anatomical reduction of macular oedema as their endpoint. It is well known that the anatomical endpoint (decrease in retinal thickness, or a "dry macula" on angiography) in many cases differs considerably from the functional endpoint (visual acuity and reading ability). Grid laser has been shown to be efficacious in reducing vascular leakage; however, it does not improve visual acuity (see Table 2.2). Evaluation of trials with different protocols remains difficult. Thus, to evaluate the efficacy of different treatment approaches a prospective randomized design is necessary, which should emphasize functional rather than anatomical endpoints. Reading ability is an excellent measure of macular function. The measurement should include reading acuity, as well as the maximum reading speed. For this purpose, the standardized Radner Reading Charts provide clinically reliable and reproducible results for individuals with normal eyesight and for patients with visual impairment. These reading test systems, which consider the current international standards for visual acuity measurements (EN ISO 8596, NAS-NRC) and the psychophysical requirements for controlling optical item interactions, can provide reliable measures for clinical and scientific analyses of reading performance [111, 124].

Retinal thickness measurement with OCT is an established method for quantitative assessment of macular oedema. Retinal thickness is determined as micrometres of maximal thickness within 500 µm around the fovea. The normal thickness values for patients without oedema are as follows: central part of central subfield 155 µm; central subfield as a whole 180 µm; inner superior, nasal and inferior subfields 260 µm; inner temporal subfield 250 µm; outer superior, inferior, and temporal subfields 230 µm; outer nasal subfield 250 µm. A desired effect for any treatment is decreasing retina thickness to within normal (175–200 µm). A reduction of 75–150 µm from baseline may be shown to be clinically significant. It is important to consider the high variability in fixation in patients with macular oedema. For quantitative comparison of retinal thickness in repeated measurements during treatment, care should be taken that the system software chooses the point of measurement independent of the patient's fixation.

The OCT is currently unable to distinguish the ILM from the outer retinal layers. However, it might be useful to exclude any kind of retinal traction. As discussed above, a large oedematous cyst might be clinically difficult to distinguish from a full thickness macular hole associated with a macular pucker. OCT can easily answer these questions; however, it does not answer the question of ischaemia, which in most

cases determines whether to treat or not. The almost only indication for fluorescein angiography in diabetic patients and patients with macula oedema is to exclude macular ischaemia (Table 2.3).

Ischaemic maculopathy remains untreatable in the area of laser coagulation. A foveal avascular zone of more than 500 µm should be considered ischaemic and thus on the current recommendations cannot be treated. However, there is still hope that ischaemic maculopathy can be treated pharmacologically. For ischaemic ophthalmopathy this has been demonstrated in a preliminary report using intravitreal triamcinolone, which was demonstrated to reduce iris neovascularization and increase visual function [69]. In contrast it is important to note that antiangiogenic drugs such as VEGF antagonists potentially increase ischaemia and have to be carefully investigated in this respect.

Early intervention in macular oedema is undoubtedly advantageous, as the risk of ultrastructural alterations induced by a persistent macular oedema increases with time. It is well known that with time the central avascular zone and the areas of ischaemia are likely to increase. Late treatment risks transition to untreatable ischaemic forms of macular oedema. To date, however, most surgical approaches will only be considered for persistent macular oedema unresponsive to laser treatment or pharmacological approaches. As a general rule surgical and medical treatment could be considered for eyes with a BCVA 20/50 +3 (68 ETDRS letters) and 20/320 (25 ETDRS letters).

The different treatment approaches are likely to affect the clinical course of macular oedema at variable time points and for different time periods. While the effect of intravitreal steroids is known to deteriorate with time, similar fluctuations are likely for other treatments. Especially for the surgical options long-term data are currently unavailable. Thus, any treatment option should be evaluated for the duration of the anticipated beneficial effect and beyond. As macular oedema requiring treatment appears to be mostly chronic, a follow-up of only 6–8 weeks is inefficient.

2.5 Summary: Clinical Treatment Dependent on the Origin of the Macular Oedema

Macular oedema is generated by different mechanisms, requiring specific interventions. In general, a combination of hydrostatic forces, mechanical forces, inflammation and pharmacotoxic effects is involved and requires modulation of treatment according to which factor is prominent in the condition (Table 2.1). Most treatment strategies focus on the release of either vitreous traction or inflammation.

Hypertensive macular oedema or *macula oedema in pregnancy* relate to hydrostatic forces and can resolve if the hypertensive stimulus is diminished upon the reduction of the blood pressure.

More difficult remains the treatment for *macula oedema secondary to vascular occlusion*. CRVO leads to a poor visual outcome in most cases, especially in the ischaemic type or in eyes with persistent macular oedema. The Central Vein Occlusion Study Group reported a visual acuity of 20/100 or less after 3 years in 58 % of patients and an improvement by two or more lines in only 20 % of cases. Treatment with laser photocoagulation or isovolemic haemodilution has no significant impact on the visual outcome of eyes with CRVO and macular oedema [20]. In contrast, the Branch Vein Occlusion Study demonstrated some benefit of grid laser coagulation with respect to an increase in visual acuity and reduction of retinal oedema. Macular ischaemia, however, remains the limiting factor [11, 13].

As the restoration of the vessel patency similar to the thrombectomy of peripheral vessels usually cannot be achieved, recent strategies have focussed on (see above for details):

- "Rerouting" from the retinal to the choroidal circulation (iatrogenic anastomoses by focal laser coagulation with high energy)
- Pars plana vitectomy with sheathotomy or radial optic neurotomy respectively
- Hyperbaric oxygenation (vasoconstrictive effect of O_2)
- Metabolic alterations

Diabetic macular disease is considered a structural alteration of the macula in any of the following situations:

- Collection of intraretinal fluid in the macula with or without exudates (lipids) and with or without cystoid changes
- Non-perfusion of parafoveal capillaries with or without intraretinal fluid
- Traction in the macular by fibrous tissue proliferation that is dragging the retinal tissue, causing surface wrinkling or detachment of the macula
- Intraretinal or preretinal haemorrhage in the macula
- Lamellar or full-thickness retinal hole formation
- Combination of the above

Treatment by laser coagulation is limited to focal oedema, but is controversial in diffuse oedema and has proven to be ineffective in ischaemic diabetic maculopathy. Currently, pharmacological approaches, which inhibit growth factor activity (anti-VEGF therapies) as well as anti-inflammatory strategies, are being established. Furthermore, clinical trials are ongoing to assess the surgical reduction of the diffusion barrier by ILM peeling in comparison to triamcinolone acetonide.

Choroidal neovascularization leads to macular oedema by inducing a chronic neurosensory detachment. Choroidal neovessels are leaky as they lack tight junctions between their endothelial cells. The treatment of the associated macular oedema depends on the effective treatment of the underlying cause, e. g. in age-related macular degeneration (AMD) or presumed ocular histoplasmosis syndrome (POHS).

A similarly limited prognosis is reported for persistent *macular oedema in patients with uveitis*. In 21–52 % of patients with uveitis there is a clinically significant macular oedema with visual impairment [23, 55, 112]. Long-term examination demonstrates a persistent reduction in visual acuity in 74 % of patients despite anti-inflammatory treatment with topical NSAIDs, steroids and systemic anti-inflammatory and immunosuppressive agents. Similarly, topical and systemic treatment with CAI (carbonic anhydrase inhibitors) failed to reduce the macular oedema [138]. Carbonic anhydrase inhibitors were shown to reduce macular oedema anatomically but failed to improve visual acuity. Pars plana vitrectomy was repeatedly reported to have a beneficial effect on the course of uveitis and associated cystoid macula oedema [123]. As expected, the majority of patients with vitreous opacities improved their initial visual acuity, but the cystoid macula oedema was also noted to resolve in 32–59 % of cases [17, 28, 29, 127, 129, 135]. However, randomized prospective trials are needed to define the role and indications of vitrectomy in altering the course of inflammatory cystoid macular oedema [59].

In *retinitis pigmentosa* and *dominant cystoid macular oedema* leakage is predominantly found at the level of the RPE and from parafoval capillaries (inner- and outer blood-retinal barrier). In these patients carbonic anhydrase inhibitors might act via an increase in active fluid transport through the RPE. Experimental treatment with octreotid (somatostatin) is currently under investigation.

2.6 Conclusions

The current treatment of macular oedema remains unsatisfactory. The dissection of the molecular mechanisms responsible for the formation of macular oedema will lead to the identification of specific therapeutic targets, and the successful application of this knowledge will result in the generation of more effective treatment modalities.

Acknowledgements. The assistance of Vassiliki Poulaki, MD, PhD, Boston, with this chapter is gratefully acknowledged.

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Is There Room for Improvement in Pucker Surgery?

3

Silvia Bopp[1]

Core Messages

- Epimacular membrane formation is an age-dependent more or less vision-threatening finding. Increased life expectancy and patient demand for better vision have led to increasing numbers of patients seeking therapy
- Surgery in less advanced cases using conventional peeling techniques can be frustrating, for no recognizable membrane edges are found for grasping by forceps and traction relief may remain incomplete
- The impressive improvements in macular hole surgery (MHS) have influenced epiretinal membrane (ERM) surgery and have led to a renewal of its therapeutic approach
- New pathoanatomical insights and technical refinements have enabled surgery for ERM at an early stage with a reasonable risk/benefit ratio
- Intended internal limiting membrane (ILM) peeling during surgery for ERM seems to be advantageous for preventing recurrences and presumably enhancing postoperative visual improvement
- A battery of visualization aids is available that allow delicate, thorough, and controlled peeling manoeuvres. Their safety profile, however, remains to be investigated
- Functional limitations must be accepted in eyes with poor initial visual acuity (VA) and long-standing disease as a result of pre-existing irreversible macular tissue alterations

3.1 Introduction

3.1.1 Definition – Clinical Features – Nomenclature

Epiretinal membranes (ERMs) are non-vascularized tissue sheets that grow on the retinal surface and cause more or less traction on the underlying neuroretina. Depending on the extent of membrane proliferation and contraction, distortion of the inner retina or the whole retinal layers, or even traction retinal detachment may occur.

Accordingly, ophthalmoscopic features vary considerably. The spectrum of changes includes (Fig. 3.1):

- A translucent glinting light reflex over the macular area
- Striae or crinkling of the inner retina and vascular tortuosity
- A visible opaque tissue sheet covering the retinal surface with full-thickness folds or a localized traction detachment

Contraction of an ERM surrounding the fovea may result in a macular pseudohole that may be misinterpreted as a full-thickness macular hole. Additional findings, such as macular oedema, spotty retinal haemorrhages, and cotton wool spots reflect secondary tissue alterations due to traction-induced vessel and nerve fibre impairment. Rarely, pigmentation within the membranes or subretinal pigmentary changes may be present.

The author has no financial or proprietary interest in any material or method mentioned.

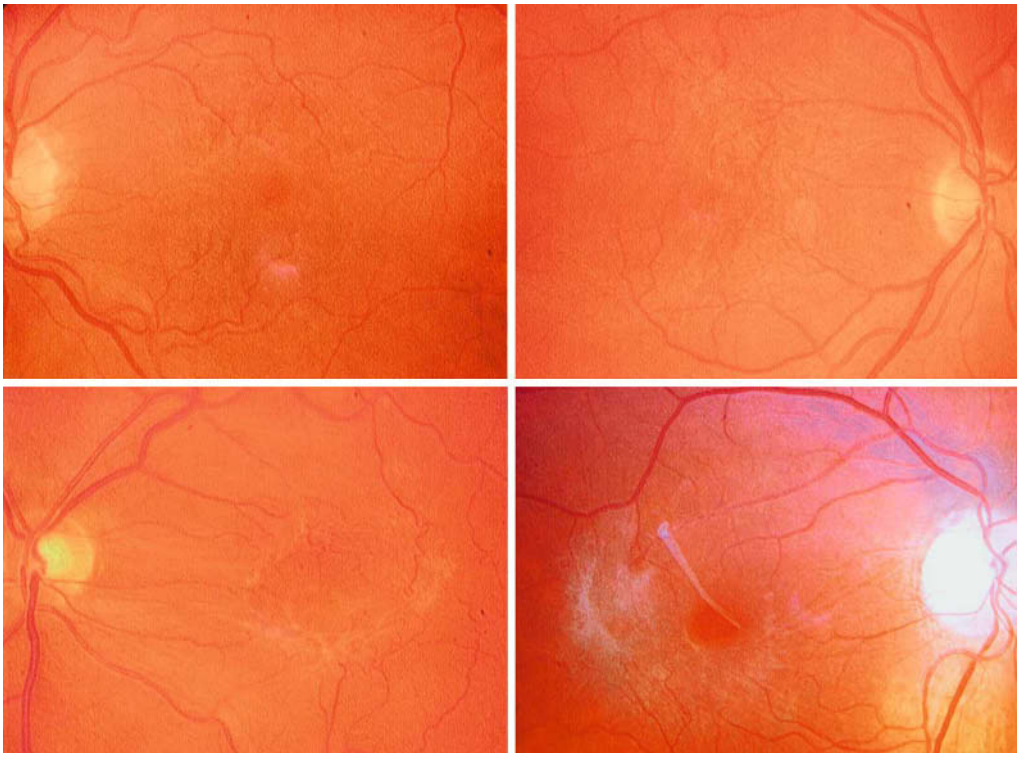

Fig. 3.1. Spectrum of clinical ERMs: asymptomatic, cellophane-type with pseudohole, VA=1.0 (*top left*). Diffuse, cellophane-type membrane, VA=0.5 (*top right*). Advanced cellophane-type with gliotic condensations and significant traction, VA=0.2 (*lower left*). Pucker type with pseudohole, minor retinal traction, VA=0.7 (*lower right*)

The heterogeneous clinical appearance of ERM is expressed by numerous clinical terms, which are used to describe the different ophthalmoscopic pictures. The most commonly used terms include:

- Cellophane maculopathy
- Surface wrinkling retinopathy
- Pre-/epiretinal gliosis
- Macular pucker

However, no standard nomenclature is in use so far. The term "ERM" is preferred here, as it includes all variants of epiretinal proliferation. To describe the severity of ERM formation, I will use the descriptions: "cellophane type" and "pucker type".

3.1.2 Classification

ERMs in the macular area most often occur as a primary or idiopathic disorder, but may also develop secondarily after laser- or cryocoagulation of retinal breaks or after retinal detachment surgery. Furthermore, membrane formation may be present as an associated phenomenon with numerous other vitreoretinal diseases (vascular, inflammatory, trauma, tumor). This article will focus on primary idiopathic and secondary ERMs after surgery for retinal detachment (RD).

3.1.3 Clinical Symptoms

Visual symptoms range from no functional impairment to severe visual deterioration. They are somewhat related to the morphologic appearance, but no strong correlation exists. Loss of visual acuity is one functional parameter, but the presence or absence of metamorphopsia is of equal importance. There are patients that present with good central vision, but suffer severely from disturbing metamorphopsia and vice versa. This has to be taken into account if surgical treatment is considered. Micropsia, macropsia and diplopia are less frequently reported, but also reflect macular tissue alterations due to traction and oedema of the macula.

3.1.4 Natural History

Idiopathic ERMs are a relatively frequent finding and increase with age. Previous reports on autopsy eyes and on large office patient series have shown that the prevalence in patients older than 50 years increases from approximately 2% up to 20% in the 8th decade, with bilaterality in up to 30% [70, 63, 58].

Many ERMs remain asymptomatic in a significant number of patients and do not show progressive growth. Vision is not significantly affected or is mildly decreased. Once diagnosed, ERMs remain stable in 38.8%, progress in 28.6% and regress somewhat in another 25.7% [22]. Within this period, 13.5% of patients develop ERM in their second eye, which indicates a symmetric trend for this disease. It is noteworthy to mention that about 10% of patients who initially present with cellophane reflex only, progressing to a pucker-like fibrosis as defined by the presence of retinal folds and opaque membranous material. Visual loss of one line or more will occur in almost half of cases, but 85% will have vision better than 0.3.

Clinically significant secondary ERM after otherwise successful retinal or vitreous surgery for retinal detachment is observed in 4–8% [45, 87, 28]. It is our impression, however, that the true incidence is much higher. Symptoms may be missed because of limited visual recovery in eyes with initial macular-off retinal detachment. Surgeons may miss mild forms of ERM, because they give priority to the fact that the retina is reattached. Our observations are supported by a prospective study [41] and an autopsy study [91]. ERMs after surgery for rhegmatogenous retinal detachment are observed in 46% and 76%, respectively. The majority of ERM formation is mild and does not cause severe gliosis. Different from idiopathic ERM, severe postdetachment pucker occurs early after surgery within a few weeks and is commonly associated with significant macular oedema [7].

Summary for the Clinician

- **Epiretinal membranes (ERMs) may occur as a primary idiopathic disorder or secondarily after treatment of retinal tears or after surgery for retinal detachment as well as in association with numerous other ocular disorders. Idiopathic and postdetachment ERMs are addressed in this paper**
- **Clinical presentation varies from cellophane-type to pucker-type gliosis**
- **Visual symptoms range from no functional impairment to severe visual deterioration**
- **Idiopathic ERM formation usually shows slow progression or may remain stable with minor visual impairment**
- **In ERMs after surgery for retinal detachment, however, an early and progressive membrane growth is more common**

3.2 Patholomorphology

3.2.1 Morphologic Findings

In the pre-vitrectomy era, autopsy eyes or eyes enucleated due to blinding or tumour diseases served as the material for histologic and ultrastructural investigations. Fundamental investigations were performed in the 1970s [3, 11, 20, 70, 92]. These studied the cellular composition of various ERMs extensively, but also focused on

the vitreoretinal interface and considered the significance of an attached or detached vitreous.

After vitrectomy techniques began to evolve in the early 1980s, most specimens were obtained during surgery, and the findings verified by Foos and others [56, 79]. Immunohistochemical techniques have provided further information in terms of the identification of the cellular elements and extracellular components of ERM.

Idiopathic ERMs are mainly composed of glial cells of retinal origin (Müller cells, fibrous astrocytes) and newly formed collagen. Surgically excised membranes also show fragments of basement membrane that indicate partial ILM removal during membrane dissection. ERM and ILM material removed during macular hole surgery (MHS) show similar morphologic features.

Secondary ERMs show a mixed cellular composition. In addition to glial cells, retinal pigment epithelial (RPE) cells and macrophages as well as fibroblasts and myofibroblasts are present [11]. By means of their ability to migrate and proliferate under pathologic conditions, glial and RPE cells undergo a process of transformation and dedifferentiation. They change their appearance and function, which makes it difficult to identify the original cell type. The pathogenesis of ERM formation therefore is not fully understood. Morphologic similarities to preretinal membranes in proliferative vitreoretinopathy (PVR) indicate a similar pathogenetic process. The presence of typical macular pucker in eyes after coagulation therapy or after retinal detachment surgery is regarded as a limited PVR reaction.

3.2.2 Role of the Vitreous

Physiologic aging of the corpus vitreum (liquefaction) leads to weakening of the adhesion between the vitreous cortex and the ILM and results in a posterior vitreous detachment (PVD). In true PVD, a clean and smooth surface without vitreous remnants occurs [21]. Some eyes, however, develop an anomalous PVD that is characterized by the presence of more or fewer remnants of the vitreous cortex left on the inner retinal surface. In these cases, splitting of the vitreous cortex (vitreoschisis) mimics true PVD. This phenomenon is well known in highly myopic and diabetic eyes, but may also occur in emmetropic eyes with idiopathic ERM [73, 74].

Careful observation during surgery for idiopathic cellophane-type ERM reveals that a delicate sheet of presumed vitreous cortex frequently covers the central retina and the ERM, although the eye shows PVD. Eyes with advanced primary or secondary macular pucker usually do not reveal obvious vitreous cortex remnants. This leads to the hypothesis that residual vitreous cortex may serve as a scaffold for ERM formation on the inner retinal surface, at least in early stages of idiopathic membrane formation.

3.2.3 New Imaging Techniques

Surgery for macular holes (MHS) was first introduced by Kelly and Wendel [38] and the impressive advances in surgical techniques seen over the past 10 years have given us new insights with regard to vitreoretinal interface changes. In addition, new diagnostic facilities, such as optical coherence tomography (OCT), have contributed to a better understanding of the delicate structural changes in both early MH stages and early idiopathic ERM formation. In addition, OCT offers the possibility of monitoring postoperative tissue restitution and correlating the cross-sectional images and retinal thickness with functional results [52, 85] (Figs. 3.2, 3.3).

The frequent association between cellophane membranes and MH indicates a possible common pathogenetic pathway. Traction on the fovea plays a critical role. In MH cases without visible cellophane reflex, this may be induced by a prolonged or incomplete PVD process or by subclinical alterations of the ILM. In cases with associated cellophane changes, delicate ERMs cause tangential traction, which ultimately leads to a lamellar or full thickness MH. If traction forces by epiretinal membranes are not centred on the centre of the macula, but spread more diffusely, gliosis without hole formation predominates the clinical picture.

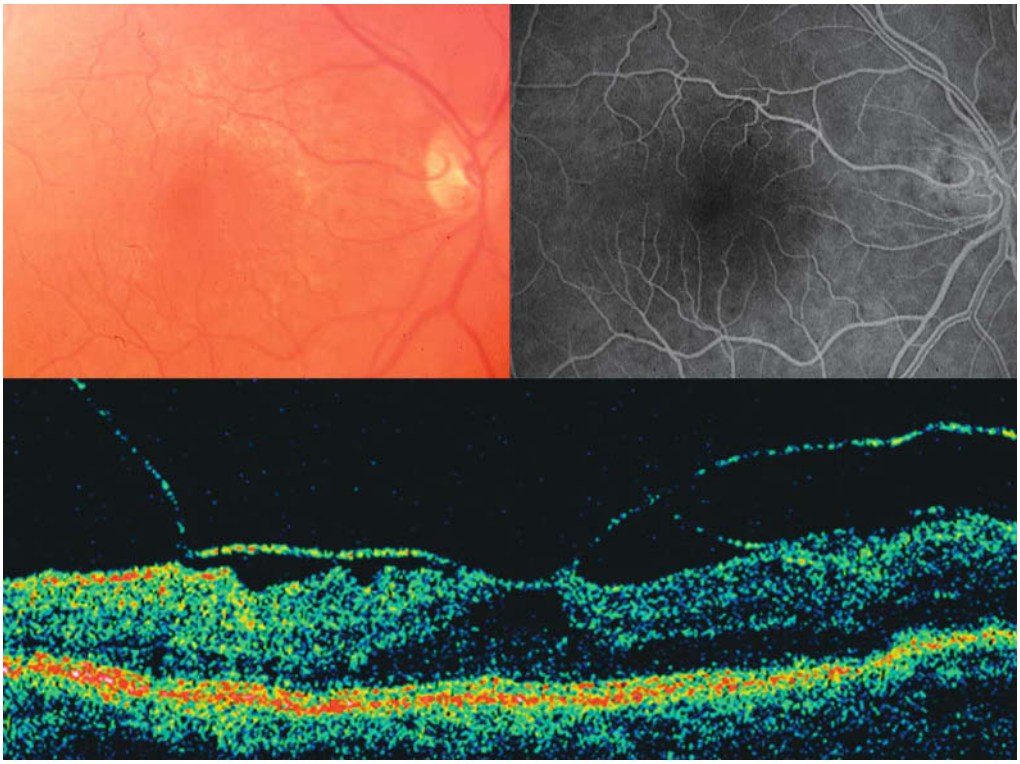

Fig. 3.2. Correlation between clinical, angiographic and OCT findings in an eye with cellophane-type ERM: diffuse glinting reflex, normal appearing FAG, membranous structures with traction on the foveola and adjacent inner retina. VA=0.5

Thus, MH without evidence of ERM and macular pucker possibly present two sides of the same disease, which can be termed traction maculopathy or (vitreo)macular traction syndrome. I would hypothesize that the condition of the vitreous and activity of cellular proliferation modulate the disease. Clinically the process ends in MH formation or macular pucker, which may show various intermediate stages.

Summary for the Clinician

- **Major pathomorphologic findings of excised ERMs include glial cells, newly formed collagen and ILM fragments**
- **In postdetachment ERMs additional cellular elements may be found, such as RPE cells, macrophages and myofibroblasts. The cellular composition indicates these membranes present a limited PVR process**
- **Incomplete vitreous separation (PVD) leaving cortical remnants on the retinal surface may act as a scaffold for cellular proliferation and is important in early ERM formation**
- **Idiopathic ERMs and MHs share some common features as revealed by ophthalmoscopy and OCT, and possibly a common pathogenetic pathway exists. The particular vitreofoveal relationship, tractional components and the activity of cellular proliferation determine the clinical picture**

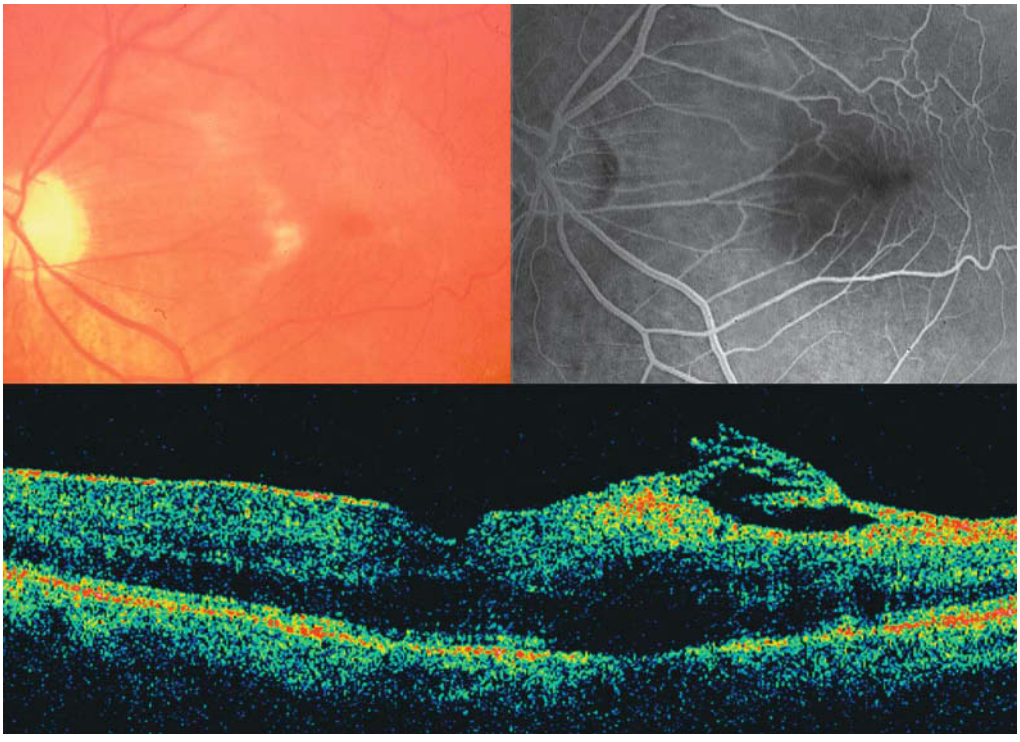

Fig. 3.3. Correlation between clinical, angiographic and OCT findings in an eye with pucker-type ERM: cellophane reflex, macular pseudohole, and cotton-wool-like lesions nasally; FAG reveals distorted perimacular vessels; OCT shows thickening of the parafoveal tissue and hyporeflective areas corresponding to the nasal portion of the ERM. VA=0.1

3.3 Conventional ERM Peeling

3.3.1 History

Surgery for macular pucker was one of the first indications in the beginning of the vitrectomy era and was introduced by Machemer in the early 1970s. Removal of vitreous opacities, haemorrhages, and vitreal traction membranes were further indications. Throughout the 1980s, removal of proliferative tissue in PVR and PDR became more common as well as treatment for epimacular membranes alone. This was the beginning of what we now call "macular surgery". Meanwhile this term stands for a large indication group for vitreous surgery.

After he had developed the basic pars plana vitrectomy technique in 1972, R. Machemer reported his first ERM dissections [48, 49]. He directed a bent needle to the edge of the membrane and mobilized the tissue from the central retina. The first microforceps was developed by S. Charles in the late 1970s, not only to peel but also to remove the tissue in one step. During the 1980s, various modifications were created to facilitate peeling in different clinical situations (membranes at the posterior pole and in the periphery; with retina on or off, fibrocellular and fibrovascular membranes).

In the 1990s, MHS led to significant improvement in vitreous surgery instrumentation. Refinement of the tips or diamond-dusted coating enhanced the ability to grasp more delicate tissue, to prevent tissue slipping out of the forceps, and to avoid damage of the underlying inner retinal layers. In 1999, K. Eckhardt and the

DORC company developed an end-gripping forceps specifically designed to peel delicate membranes and remove the ILM. The so-called "Eckhardt forceps" has become a standard instrument for this purpose. Another recently designed forceps by S. Charles is a bent forceps ("conformal"), the curvature of which parallels the retinal surface in order to avoid grasping the retina while approaching the membrane.

Viewing systems have also improved during the past decade. Direct (contact lenses) and indirect viewing systems (BIOM) with special lenses designed for macular surgery provide a high magnification, resolution, and enhanced three-dimensional view. More powerful light sources are contributing to the visualization of delicate pathologic structures.

Recently, dye-assisted vitreous surgery was introduced to facilitate ILM peeling [37, 10]. Selective staining of ERM (trypan blue) or the ILM (indocyanine green, ICG) enables the surgeon to target precisely the pathologic structures and remove the ERM or ILM more safely, quickly, and completely. Despite its widespread use, the application of ICG as an intraoperative visualization aid is under debate. Some reports indicate potential toxicity, and studies proving its safeness are still lacking. Most surgeons who have used ICG regularly for the past few years have noticed no toxic side effects and aver that the advantages at least outweigh the possible adverse effects.

Summing up, the progress in techniques has given us the ability to remove not only opaque epiretinal gliotic membranes but also delicate transparent tissue, without threatening the fragile macular structures. As a result of better control of manipulation and less risk of iatrogenic damage, surgery tends to be performed at an earlier stage and with better initial visual acuity.

3.3.2 Indications for ERM Removal and Functional Outcome

In general, ERM removal is considered for selected cases only, e.g. eyes with substantial visual loss (0.3 or worse) and disturbing metamorphopsia [56, 51]. Ophthalmologic criteria are either pucker-like epiretinal tissue (dense, opaque, gliotic) or cellophane-like (transparent) tissue, which causes retinal folds and marked vascular tortuosity. Major arguments for a cautious approach to ERM are the relatively benign nature of idiopathic ERM regarding visual impairment on the one hand and the well-known potential vision-threatening complications of vitreous surgery on the other. Retinal breaks and consecutive retinal detachment occur in about 5–8 %, ERM recurrence in 4–8 %, nuclear sclerosis and significant cataract in up to 63 %, and endophthalmitis rarely (<1 %) [57, 68, 13, 14].

Furthermore, functional recovery usually remains limited. Although visual improvement (2 lines) is observed in 65–95 % of cases, a high vision level (0.5) is achieved in 20–35 % only. These results are valid for both idiopathic and secondary ERM [57, 13, 14, 67, 68]. Several reasons were identified that explain incomplete functional improvement: persistent or enhanced postoperative cystoid macular oedema, remnants of membranous tissue and suspected irreversible tissue damage to the macula due to a long-standing disease. Preoperative factors that allow a prognostic estimation of the functional outcome are: poor initial visual acuity, pre-existing leakage, or cystoid macular oedema and blurred vision of long duration.

Well-defined indications for surgery are still lacking and largely depend on the surgeon's individual preferences. Since idiopathic ERMs with their associated membrane contraction usually progress slowly and may maintain a stable morphologic appearance and function, surgeons advise patients to wait while the disease process increasingly advances. Secondary macular pucker after retinal detachment occurs much more rapidly (within weeks) and therefore is usually treated earlier. The true time window for a successful outcome, however, is not known.

3.3.3 Technique for ERM Removal

A conventional, three-port pars plana approach is usually employed. In advanced stages of idiopathic ERM and in cases of postcoagulation or postdetachment macular pucker, a posterior vitreous detachment is present in the majority of cases. After complete vitrectomy, the membrane is inspected for visible edges. These rims can be engaged with a forceps directly. If a preexisting edge is not present, a bent-tip needle or a pick, applied to the margin of the epiretinal tissue, helps to induce a circumscribed flap. After grasping the preretinal tissue with a forceps, careful and gentle dissection is started from the peripheral portion to the epicentre of the membrane. Directing the tip tangentially to the retinal surface and engaging the membrane from different directions helps avoid fragmentation of the tissue sheet and inadvertent tearing of the retina. Unusually firm adhesions to the retina may necessitate amputation of the tissue with horizontal scissors or a vitreous cutter. If inspection of the peripheral retina surgery shows no iatrogenic damage, surgery is finished by sclerotomy closure.

During the peeling process, we observe ERMs with varying characteristics: Some can be peeled off easily and completely in one piece leaving a smooth inner retinal surface. Others show a firm adherence to the retina, and removal may risk causing damage to the superficial retina. Multiple attempts to engage the tissue may be necessary. Since identification of membranous material and distorted retina can be difficult at times, the surgeon must memorize the areas in which the ERM was removed and those where peeling is still necessary. Otherwise, he is likely to damage the retina by repeated attempts to peel where no membrane is left, or he may leave remnants of epiretinal tissue with incomplete traction relief and the risk of recurrent membrane formation. Another variant of ERM shows a soft tissue consistency and resembles so-called immature PVR membranes. They fray during peeling, which leaves the impression of incomplete separation. A repeat approach often does not succeed in a clean cleavage between retina and membrane, particularly in areas with pre-existing retinal whitening (associated cotton wool spots and retinal oedema). For these cases, presumed membrane parts should be removed using horizontal scissors, but otherwise left in situ. Usually the retina will flatten and smoothen postoperatively.

Although peeling manoeuvres aim for maximal ERM removal and traction relief, some postoperatively visible glinting reflex, superficial wrinkling, or vessel distortion hints at residual epiretinal tissue or possibly persistent intraretinal contraction. Furthermore, recurrent ERMs can be observed in up to 21% [62].

3.3.4 Surgical Variants

Cataract formation is the most frequent complication after vitreous surgery, and its incidence correlates with the age of the patient and the complexity of the surgical procedure, especially the use of an intravitreal tamponade [53, 36]. In surgery for idiopathic ERM, progressive nuclear sclerosis is reported in 50–75% [14]. Some surgeons believe that core vitrectomy is more likely to protect the lens than complete vitrectomy [51]. For these reasons, most surgeons also avoid an intraocular tamponade (air, short-acting gas), unless intraoperative complications, such as retinal breaks or retinal detachment, require additional manoeuvres. Nevertheless, postvitrectomy cataract formation remains frequent and is largely unpredictable.

Some surgeons favour the use of an encircling band in addition to pars plana vitrectomy in order to prevent postoperative retinal detachment. However, no studies have verified a protective effect.

Summary for the Clinician

- **Standard three-port vitrectomy with thorough peeling manoeuvres is the gold standard for ERM removal**
- **Visible edges, preretinal cellophane or opaque structures are landmarks for the surgeon to approach the membrane**
- **As the consistency of the epiretinal tissue and its adherence to the retina vary consid-**

erably, peeling manoeuvres may turn out to be easy or difficult and possibly remain incomplete

- **However, discrimination of epiretinal tissue components and oedematous distorted retina is crucial to avoid unnecessary retinal damage**
- **Recurrent ERM formation is observed in up to 20 % after conventional membrane peeling**
- **Whether alternative techniques, such as performing a limited vitrectomy (core vitrectomy) only or using a small incision approach (transconjunctival 25-gauge systems), are as safe and efficient as the conventional approach remains to be proven. Similarly, the usefulness of an encircling band to prevent postvitrectomy detachments is unclear and remains the surgeon's individual decision**

3.4 Advanced ERM Peeling

3.4.1 The Vitreoretinal Interface in Eyes with ERM

In ERM surgery, multiple layers must be sequentially peeled off at times in order to obtain a clean retinal surface [51]. In addition, it has been believed that PVD was almost always present [79] and that surgical separation of the ERM occurred at the level of the ILM [78]. Thus, the presence of multiple layers was attributed to the ERM itself. The current understanding of the vitreoretinal interface and its pathologic alterations allows a more precise correlation of anatomic and intraoperative findings. In ERM surgery, we have to face remnants of the posterior vitreous cortex, newly formed epiretinal tissue, and the ILM.

The Vitreous. Idiopathic ERM is present in eyes with and without posterior vitreous detachment (PVD). Biomicroscopy and B scans have turned out to be unreliable in establishing the true incidence of PVD. Intraoperative findings allow a more precise diagnosis. Biomicroscopic examination and ultrasound findings report a PVD in up to 100 % [2, 92] and similar rates were found during surgery [79]. In our experience, complete PVD in idiopathic ERM is much less frequent (see Sect. 3.5.1). This may be explained by the fact that surgery in earlier times was restricted to advanced pucker-like membranes, but today is performed in much earlier stages. Thus, a PVD is no precondition for ERM formation and growth.

The ERM and ILM. Vitreomacular traction syndrome, lamellar and full-thickness macular hole, and ERM share the common feature of vitreoretinal interface abnormalities: MHS has shown the significance of an incomplete vitreous separation that causes vitreomacular traction and moreover the presence of delicate cellophane membranes that result in a tangential traction component. In addition, histological findings of ILM material removed during MHS show some glial proliferation overlying the ILM [16, 55, 44]. Similarly, fragments of ILM are a regular finding in surgically excised ERM and show similar morphologic features [87, 79]. Inadvertent ILM removal during ERM surgery, furthermore, has no noticeable adverse effects [47]. Thus, in the face of the surgical problems mentioned above, performing a thorough and complete removal of epiretinal tissue together with the ILM seems a logical concept. Prevention of recurrent membranes and possible extension of surgery to early cases with very delicate and transparent membranes may be achieved by this approach.

3.4.2 ERM Removal with ILM Peeling

Surgically removed ERM specimens often demonstrate various degrees of attached ILM. Peeling of both the epiretinal tissue plus the adjacent ILM should facilitate complete removal of the newly formed tissue according to the hypothesis that "if the ILM is removed there cannot be any proliferative tissue at all". Interestingly, there is clinical evidence that the ILM itself may reveal traction on the retina. During surgery for cellophane maculopathy, we fre-

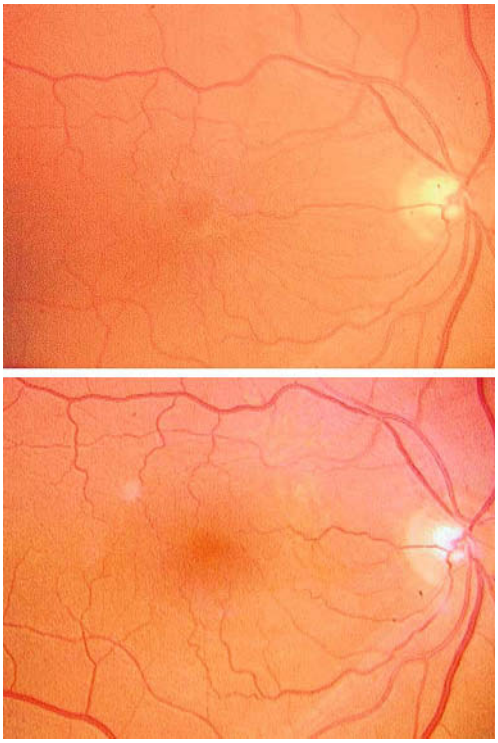

Fig. 3.4. Case example with cellophane-type ERM before and after surgery using advanced peeling techniques. Note the smooth epimacular surface and margins of the ILM-peeled zone

quently notice persistent intraretinal traction folds after having removed the epiretinal tissue. Only after peeling of the ILM do those fine striae disappear and the retinal surface smoothen (Fig. 3.4). Although there are no morphologic characteristics known that allow differentiation between normal and pathologically altered ILM, surgical experience of ILM removal for various macular diseases shows that consistency, thickness, and retinal adhesion of the ILM vary significantly. Thus, ILM peeling in pucker surgery will eliminate any continued contraction on the macula, or, in other words, since we assume that the ILM serves as a scaffold for epiretinal cellular proliferation, persistent contraction will be eliminated and possible recurrent membranes prevented.

3.4.3 Identification of Specific Tissue Sheets and Meticulous Peeling Manoeuvres

The basic surgical technique is described above. From the anatomic point of view there may be three different tissue layers present: remnants of the posterior vitreous cortex, epiretinal proliferative tissue, and the ILM. Although all three layers have a more or less transparent appearance, each type of tissue has certain characteristics that allow identification. Complete and controlled membrane removal is only possible if the surgeon is aware of which tissue she/he is holding in his forceps.

Posterior vitreous cortex remnants typically present as a sheet of fragile elastic tissue that is thickest in the centre of the retina and thins towards the vascular arcade and midperiphery. During peeling, it will form delicate strands and separate from the retina without evidence of retinal traction. It shows no distinct borders and vanishes towards the periphery.

Characteristics of ERM removal are described above. The semitransparent or opaque nature becomes obvious once the membrane is mobilized and separated. Usually the membrane shows varying thicknesses (thickest in the centre and thinner towards the peripheral edges) and irregular borders. Its consistency ranges from soft and fragile to fibrous and condensed. The membrane usually separates at the plane of the ILM, although in exceptional cases the membrane and ILM may come off together (see below).

With the advent of intentional ILM removal [60, 8, 16] came scepticism about whether it was indeed the ILM that was being removed. The ILM appearance is unique, however, and can be reliably distinguished from ERM by certain features. Furthermore, numerous morphologic studies on excised specimens confirmed the correct intraoperative diagnosis. It is now generally accepted that tissue with the following characteristics represents ILM: it is highly reflective, looks absolutely transparent, appears homogeneous, and shows no distinct borders. Peeled ILM curls up much like the lens capsule after capsulorrhexis. Occasionally, some super-

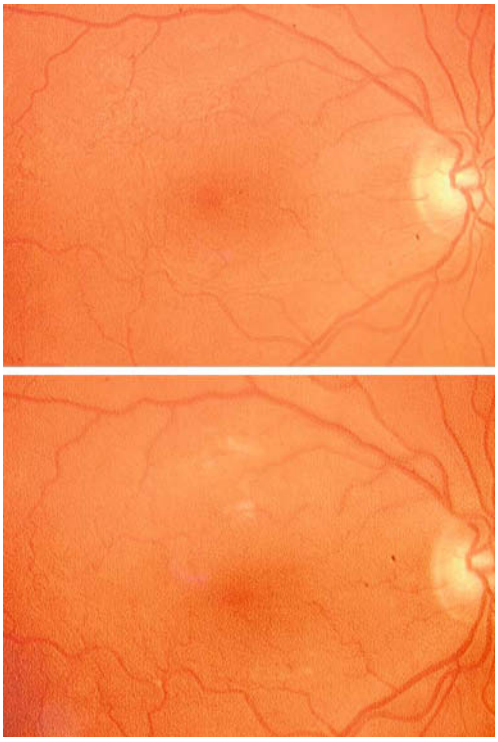

Fig. 3.5. Case example with cellophane-type ERM before and 1 day after vitreous surgery. ILM peeling was performed without ICG. Two whitish dots above the fovea indicate a superficial trauma to the nerve fibre layer by the forceps used to grasp the delicate transparent tissue

ficial petechial haemorrhages occur after ILM peeling that vanish within 1–2 days. ILM-denuded retinal areas lack their normal shiny reflex, rather showing a whitish dull surface reflex. This phenomenon will disappear within a few days, indicating some regeneration of the Müller cell basement membrane [59, 61].

Eyes designed for ERM surgery ideally present with these three types of membranous tissue. In reality, however, we may face different situations, such as: eyes lacking the posterior vitreous layer, the posterior vitreous sheet may be removed together with the ERM, or the ERM plus ILM come off as a single sheet. ICG-guided peeling has clearly shown, however, that only parts of the adjacent ILM will be removed during ERM peeling and significant remnants stay behind. Thus, a second peeling procedure must be performed to remove the remaining ILM in the desired area. The surgeon must remember the areas he already peeled and directly continue to remove the remaining parts.

There is no doubt that atraumatic and complete ILM removal is extremely difficult without extensive surgical experience. Even in the hands of well-trained surgeons, iatrogenic damage by the instrumentation happens. Inadvertent pinches to the nerve fibre layer, retinal oedema and focal haemorrhages and contusional RPE lesions may occur (Fig. 3.5). Retinal defects, however, fortunately are an extremely rare complication. In addition, control of the completeness of ILM peeling in ERM cases is far more demanding in comparison to eyes with macular holes. Intraoperative visualization using supravital dyes (indocyanine green, trypan blue) will allow much more easy, efficient and controlled manoeuvres. Beyond these obvious practical advantages, the use of visualization aids has gained a high popularity. However, from the scientific point of view and for safety reasons, their application is still under debate (see Sect. 3.6).

Summary for the Clinician

- **Careful ERM peeling often reveals multiple tissue layers**
- **The current understanding of pathologic vitreoretinal interface changes in ERMs allows a correlation between anatomic and intraoperative findings**
- **During surgery, vitreous cortex remnants, epiretinal proliferative tissue and ILM material can be distinguished by their specific characteristics**
- **Intended ILM peeling is advantageous with respect to favourable visual outcomes and prevention of recurrent membranes**
- **The use of visualization aids (ICG, trypan blue, triamcinolone) as an intraoperative tool allows an easier, more atraumatic and complete tissue removal**

3.4.4 Alternative Surgical Techniques

Numerous observation systems are available for vitreous surgery. Most surgeons use indirect viewing systems (BIOM, EIBOS) in combination with high-resolution contact or non-contact lenses (so-called macular lenses). Light sources are either a handheld fibre optic or a sclerotomy-fixed fibre optic.

Some surgeons, particularly in France, favour the slit-lamp system for vitreous and retinal surgery [6]. Originally used for retinal detachment surgery, this technique has proven advantageous for macular surgery as well. A slit-lamp attached to the microscope plus a plano-concave contact lens (type Kilp lens) allow a high magnification of the central fundus and excellent viewing of the vitreoretinal interface. Illumination of the macular area by a small, slightly angled light beam causes reflections at the vitreoretinal interface which increase visibility of shiny and gliosed surface alterations. Surgeons routinely using a slit-lamp microscope for ERM surgery do not feel it necessary to use dyes, because the different membranous tissues (posterior hyaloid remnants, epiretinal gliosis and ILM) can be distinguished by their specific appearance and reflectivity [15].

So-called minimal invasive surgery reflects a new trend in vitreoretinal surgery, particularly for macular cases. According to the slogan: "maximize the efficiency by saving operating time and inducing less surgically induced trauma", a variety of techniques are under investigation.

Transconjunctival sutureless vitrectomy techniques using 25-gauge instrumentation as developed by de Juan, involve a reduction in diameter by one-third in comparison to standard 20-gauge instruments and have proven to be effective for macular holes, ERM, and mild diabetic retinopathy [23]. The technique has its limitations with respect to a complete vitrectomy and is time-consuming because of the small lumen of the vitrectomy probe. Most surgeons using the 25-gauge system limit vitreous removal to a central core vitrectomy.

For selected macular pucker cases, membrane peeling without vitrectomy was suggested [72]. A bent needle is introduced transconjunctivally and guided through the vitreous cavity to the membrane. Similar to the technique first described by Machemer, the ERM is separated from the retinal surface, but left somewhere in the vitreous cavity. The goal of therapy is to relieve foveal traction and avoid postoperative lens changes. Basically, this procedure can be performed at the slit lamp or in the operating room. In my experience, there are only very rare situations in which one could consider this technique: cases which present with a typical opaque pucker membrane and very distinct edges (chance to remove the membrane in a single piece) and young patients (to exclude the risk of cataract formation). The procedure appears somehow like a step backward, if we take into account the current knowledge and treatment concepts in vitreoretinal interface disorders.

3.5 Studies on Idiopathic ERM Using Advanced Peeling Techniques

3.5.1 Surgery for Minimal Variants of ERM (mERM) by the Author

The refinements of instrumentation and advancement of peeling techniques initiated by MHS raise the question of whether surgery for ERM is safe and effective at an early stage of the disease. The author investigated the functional outcome after surgery for delicate ERM as defined by the presence of a cellophane reflex with or without fine surface wrinkling of the inner retinal layers [5]. We called them minimal variants of ERM (mERM). Many patients remain asymptomatic with such findings. Some cases, however, suffer from visual deterioration and disturbing metamorphopsia. Surgery on these eyes using conventional techniques is often frustrating: Difficulty in engaging the delicate membranes can result in incomplete traction relief, involving a high risk of producing iatrogenic retinal lesions, and leading to unsatisfac-

tory functional and anatomic results. The efficacy and risk/benefit ratio of combined ERM and ILM removal on eyes with mERM was investigated.

3.5.1.1 Patients and Methods

A total of 153 eyes underwent surgery for mERM between March 1999 and February 2001. Patients were elected for surgery if they complained about visual deterioration or had binocular confusion (orientation in space and reading disturbance). A standard three-port vitrectomy approach was performed. The peeling manoeuvre included all membranous material plus the ILM. Intraoperative ICG staining was introduced in January 2001, and 15 out of 153 cases had dye-assisted peeling manoeuvres. All primarily phakic patients over 50 years of age had concomitant lens surgery with phako/IOL. A total of 88% had a combined procedure (phako/IOL/vitrectomy). Follow-up ranged from 3 to 21.4 months (mean 7 months). Main outcome measures were far distance and near vision (decimal lines), perception of metamorphopsia (Amsler chart), and patient satisfaction (subjective score: satisfied – uncertain – dissatisfied). Pre- and postoperative functional results were categorized into three groups that reflected vision levels for practical life, such as reading, car driving, artistic and other vision demanding activities (e.g. hunting, golf): group A ≤ 0.3 (= marked visual deterioration, no reading ability, 32.3%), group B 0.4–0.6 (= moderate visual impairment, reading difficulties; 51.5%) and group C ≤ 0.7 (subjective blurred vision and disturbing metamorphopsia, 16.1%).

3.5.1.2 Results

Preoperative visual acuity (VA) ranged from 0.1 to 1.0; two-thirds (67.6%) of patients presented with an initial VA of 0.4 and better. Intraoperatively, complete vitreous attachment necessitating surgical vitreous delamination was present in 32.3%. In most eyes, two layers of membranous material were identified: firstly, a soft elastic one that was consistent with posterior cortex remnants, and, secondly, the ILM. Only after removal of the latter did the macular area regain a smooth surface and the irregular reflex or fine folds disappear. The excised ILM usually had its typical transparent homogeneous appearance. In several eyes, parts of the ILM showed a slight opacity and thickening, suggesting that the ILM was overgrown with some ERM on its vitreal

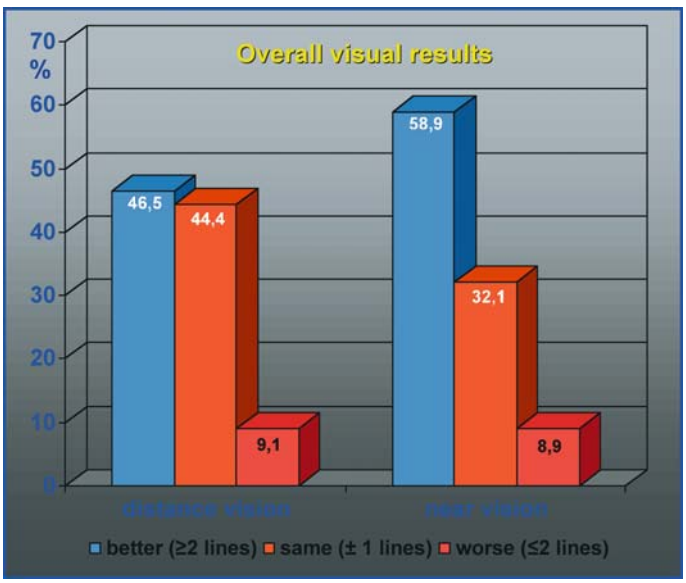

Fig. 3.6. Overall visual results (distance and near vision) after surgery for mERM

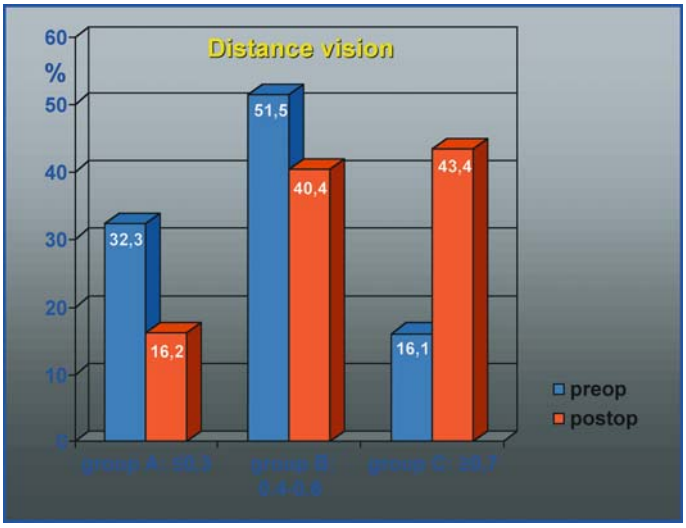

Fig. 3.7. Pre- and postoperative percentage of eyes with severe (group A), moderate (group B) and minor visual impairment (group C). With respect to distance vision, a significant shift towards group B and C indicates improved function for daily practical life

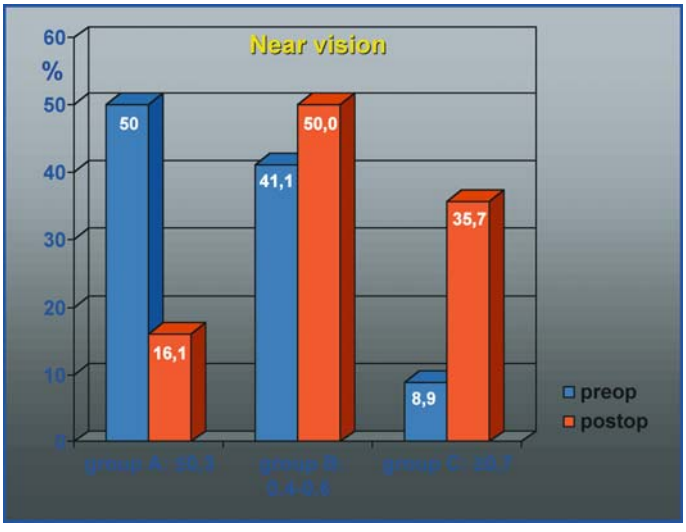

Fig. 3.8. Pre- and postoperative percentage of eyes with severe (group A), moderate (group B) and minor visual impairment (group C). Improvement of near vision is comparable to distance vision. About 85% of patients have reading ability

side. Postoperative results are depicted in detail in Figs. 3.6–3.10. In total, 90% of patients experienced significant reduction or complete disappearance of metamorphopsia (Fig. 3.9). Visual results varied more widely: 46.5% improved (2 lines), 44.4% remained stable (±1 line), and 9.1% decreased (2 lines) (Fig. 3.6). A final VA of 0.4 and better was achieved in 83.8%, equal to or better than 0.7 in 43.4% of cases. Retinal detachment occurred in 3% and was treated by pneumatic retinopexy. One case progressed to PVR. Virtually all phakic eyes that had no lens surgery during vitrectomy developed nuclear sclerosis and had subsequent cataract surgery within 6–12 months after vitrectomy. The major causes of functionally unsatisfactory results were persistent (9%) and newly postoperative new macular oedema (5%) as revealed by fluorescein angiography or a long history of blurred vision. Analysis of the patient's subjective evaluation showed: 71.5% were satisfied, 20.9% were uncertain and 7.7% were dissatisfied with the outcome.

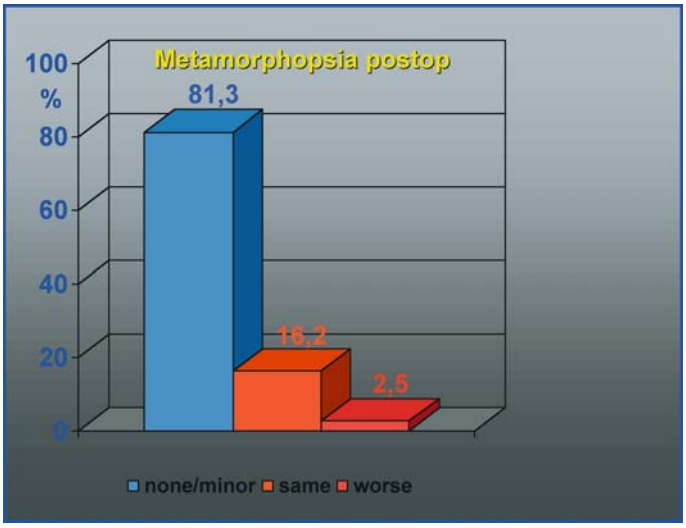

Fig. 3.9. Postoperative analysis of metamorphopsia, which was a motive to seek therapy. Complete absence or significant diminution was achieved in more than 80 %

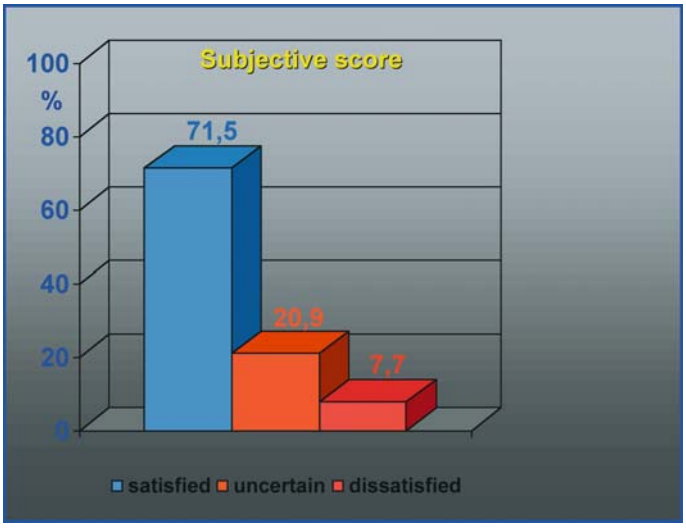

Fig. 3.10. Analysis of patient's satisfaction. Results strongly correlate with postoperative perception of metamorphopsia. The absence of distorted images was greatly appreciated, even if no significant vision improvement was achieved

3.5.1.3 Discussion

Previous reports on surgery for ERM address the more advanced stages of the disease or include all variants from cellophane-type to pucker-type membranes. We focused on surgery for minimal variants of ERM, i.e. ophthalmosopically transparent tissue. Accordingly, 67.6 % of eyes presented with an initial VA of 0.4 or better and 16.1 % even higher. Macular oedema was not evident by ophthalmoscopy, but fluorescein angiography revealed some macular leakage or cystoid oedema in one-third of the eyes.

Analysis of pre- and postoperative vision among the three groups showed a shift from group A towards group C (Figs. 3.7, 3.8). In other words, during the postoperative course, the percentage of eyes with initial poor or moderate VA decreased (group A, B) and that with good VA

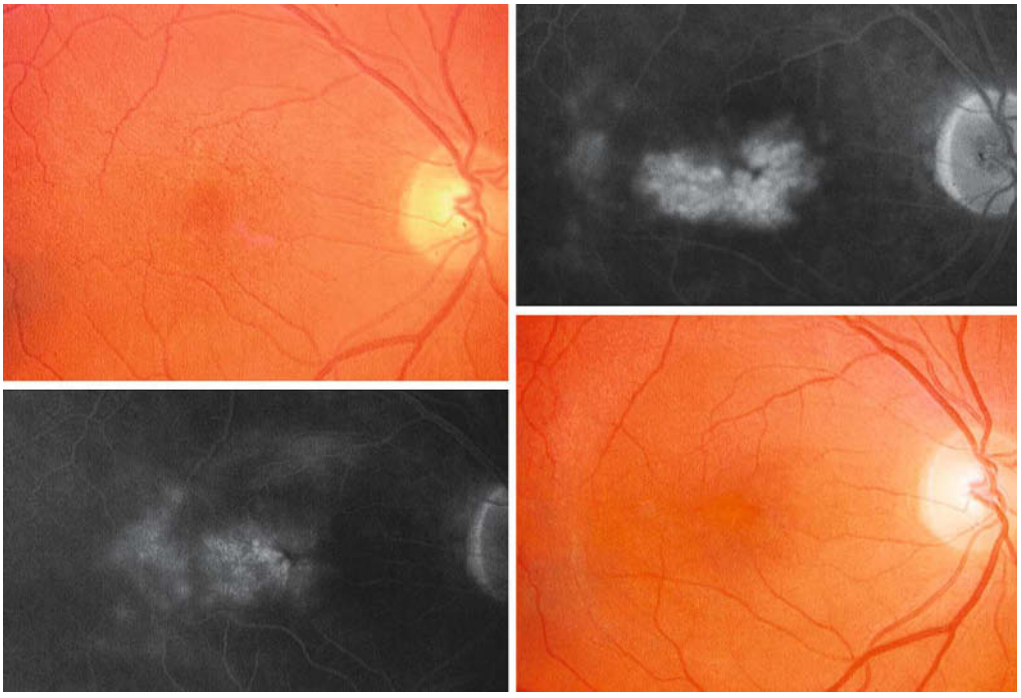

Fig. 3.11. Case example with mERM before and after membrane/ILM peeling. Preoperative ophthalmoscopy (*top left*) and FAG (*bottom left*) revealed a cellophane-type membrane with a subclinical cystoid macular oedema, VA=0.3. Postoperatively, cystoid macular oedema initially increased (*top right*), and vision failed to improve. Acetazolamide was given and macular oedema slowly resolved. Final VA was 0.6 six months postoperatively (*bottom right*)

increased significantly (group C). The major shift took place from group B to C. Far distance and near vision behaved similarly, but near vision remained at a slightly lower level. Analysis of individual cases showed that patients with moderate visual loss (group B) for the most part profit from surgery, whereas those with low initial vision (group A) often remained at a low level. For eyes presenting with good initial VA (group C), further improvement is limited. Their clinical course is difficult to assess in the diagram. Their visual results did not change significantly; five out of six eyes remained at the same vision level, one case showing a decrease from 1.0 to 0.6. He did not complain, as metamorphopsia was no longer present.

We have learned that vision alone is an insufficient criterion for ERM patients, as metamorphopsia negatively affects the quality of vision and is a major motive for seeking therapy. Symptoms diminished or vanished in the vast majority of cases. This was greatly appreciated by the patients even if vision did not change significantly.

Less than 10 % of cases lost some vision. An equal number of patients had no improvement of disturbing metamorphopsia. Two major reasons were found: Firstly, postoperative macular oedema (persistent or new) was identified by routine fluorescein angiography (FAG) (Fig. 3.11). Very slow, spontaneous resolution was observed, over months. Medical therapy with acetazolamide was given. Treatment seemed to be effective, with the macular oedema resolving within a few months. Secondly, another subgroup of patients failed to improve and complained of persistent metamorphopsia despite a normal appearing macula and regular FAG findings. They reported long-standing symptoms and had poor initial VA. Delays in

treatment obviously lead to irreversible macular tissue damage and prohibit functional recovery after surgical repair.

Vision threatening complications were rare: No posterior breaks were noted. Five eyes developed postoperative rhegmatogenous retinal detachment, which was successfully repaired in four eyes by pneumatic retinopexy. One case developed PVR and had subsequent silicone oil surgery. Initial VA was 0.5 and dropped to 0.1. No recurrent ERM was observed during the entire follow-up.

3.5.1.4 Conclusions

Minimal variants of ERM (mERM) can cause significant subjective symptoms and show a variable clinical course. There are no clinical features that predict at which vision level the disease will stop. For patients who suffer from visual impairment and disturbing metamorphopsia, surgical treatment with advanced peeling techniques should be considered independently from visual acuity. Delay in treatment negatively affects the functional outcome after surgery. Since vision threatening complications are rare, early indication for surgery is justified. However, there is a rate of 10 % postoperative macular oedema that may lead to a mild functional decrease or a prolonged postoperative course. This is of particular importance for patients with good initial VA.

We suggest the following criteria for recommending surgery for patients with idiopathic mERM: recent onset or recent progression of symptoms and corresponding subjective complaints. In our experience, functional recovery takes a relatively long time, and final VA does not occur earlier than a few months after surgery. Patients should be informed about a possible enduring postoperative course. From the time when we introduced intravitreal application of 4 mg triamcinolone acetonide as a routine at the end of surgery (January 2002), the incidence of postoperative macular oedema decreased significantly and visual recovery occurred much earlier.

Summary for the Clinician

- **Disturbing metamorphopsia, visual deterioration, and binocular disturbance (reading, orientation in space) are major ERM-related symptoms and motivate patients to seek therapy**
- **Function, as a surgical indication, has gained in relevance as a result of improvements in vitreous surgery. ERM removal can be considered a relatively safe procedure today**
- **Visual acuity alone is no criterion for/against surgery in a patient with ERM. The patient's demands regarding his vision in daily life and expectations regarding the postoperative result should be taken into account**
- **Patients with moderate visual loss, recent onset of symptoms, or progression are the best candidates for ERM surgery**
- **Functional outcome in patients with poor initial VA or long-standing disease is dissatisfying**
- **Postoperative persistent or new macular oedema has an important impact on the postoperative course and visual outcome. Intraoperative application of 4 mg triamcinolone has proven efficient in preventing postoperative macular oedema and enables early visual rehabilitation**

3.5.2 Other Studies

Since surgical specimens of ERM often demonstrate varying degrees of ILM, the question arises as to whether intentional and complete ILM removal has an impact on the functional and anatomic outcome. So far, the pathophysiological response of the retina and Müller cells after ILM removal is not known. Histologic findings in an eye after a postdetachment pucker peeling revealed a very thin renewed ILM with few glial cells in the area previously peeled [59]. Clinical findings after ILM peeling show a sharp delineation at the margin of the ILM peeling zone and give the impression that the ILM-denuded retinal surface is somewhat thinner. Sivalingam

et al. [77] performed a clinicohistopathological study and found that functional results after ERM surgery were worse when the specimen showed large segments of ILM.

In MHS, there is a general consensus that ERM should be removed if present. The necessity of routine ILM peeling, however, is still under debate. The fact is that anatomic success rates using this manoeuvre have never been that good and have approached almost 100 %. Several clinical studies on intentional ILM removal during MHS did not reveal any adverse effect on vision. Brooks [9] reported even better functional results in comparison to conventionally treated eyes.

Similarly, a recently published clinical report on ERM surgery with and without ILM peeling demonstrated no adverse effect of ILM removal [62]. Visual acuity improved or was unchanged in all patients with ERM plus ILM peeling in comparison to 79 % in eyes with ERM removal only. Furthermore, 21 % of eyes in the latter group showed evidence of persistent traction at the inner retinal surface or some recurrent ERM. None of the eyes in the ILM peeling group had these symptoms. The major outcomes of this study – favourable functional results and no recurrences – are consistent with the results of our study.

As a synopsis, there are obvious analogies between ERM and MH with respect to ILM peeling. Both entities share common pathologic features with respect to the vitreoretinal interface, such as varying degree of ERM formation and subtle ILM alterations. Benefits include: firstly, ILM peeling does not negatively affect postoperative vision; secondly, we achieved maximal traction relief and obviously removed the scaffold for reproliferation. As a result, we observed virtually no recurrences after ERM surgery and no reopenings after MHS.

3.6 Visualization Aids in Pucker Surgery: Terrific Surgical Tools or Harmful Agents?

ILM peeling plays an increasingly important role in vitreoretinal interface disorders. Using this technique, primary anatomic success rates in MHS approach 100 % [9], control of macular oedema can be achieved in various diseases, and a favourable outcome after pucker surgery has been described [62]. So far, there is no evidence that ILM peeling itself causes a deleterious effect on retinal function. On the other hand, the technique of ILM peeling remains a challenge for the vitreoretinal surgeon.

The introduction of intravitreal dyes was a breakthrough as a method of dealing with this problem. ICG was used initially, followed by trypan blue. The new manoeuvres, which allow a better visualization of vitreoretinal interface structures, were immediately taken up by the surgeons. There is a consensus that dye-enhanced vitreous surgery allows faster, more complete, precise and atraumatic removal of the ILM. Furthermore, these techniques demonstrate vitreoretinal interface relationships not previously verifiable in terms of clinical examination. Insecurity and hesitation arose, however, when first reports on potential toxicity of ICG were published. In fact, ICG safety studies are lacking. Despite the tension between the obvious practical advances and the possible vision-threatening side effects, the subject has been discussed dispassionately in the current literature and during meetings.

3.6.1 Indocyanine Green

Peeling of delicate membranes and ILM peeling include an intrinsic risk of damaging the retina and RPE, particularly with lack of experience or aggressive handling of the translucent structures. Nerve fibre injuries, retinal haemorrhages and oedema, retinal holes, and RPE damage may occur, which negatively affect visual outcome. This is of particular significance during the learning curve of this technique. Intravitreal ICG applica-

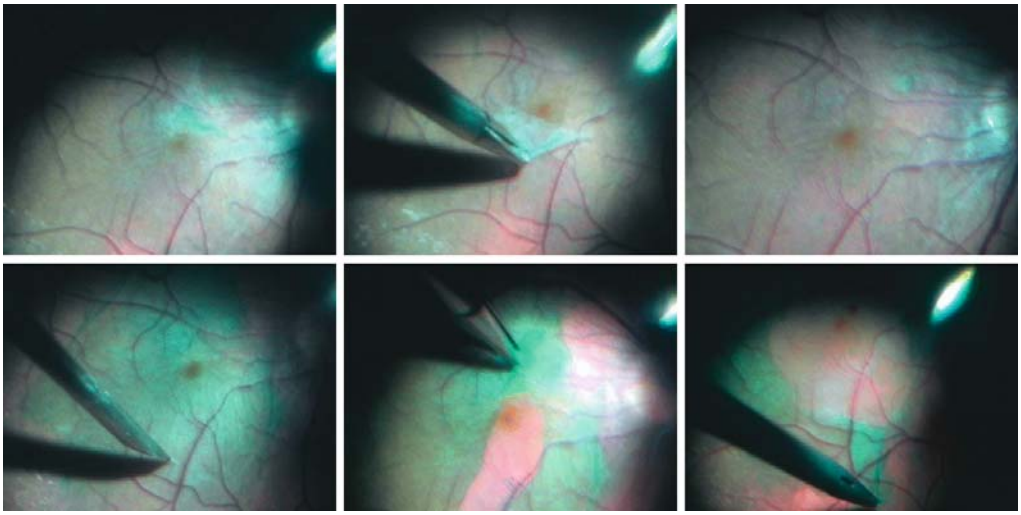

Fig. 3.12. ERM surgery with advanced peeling techniques and use of ICG: After vitrectomy and vitreous delamination, ICG was applied to the posterior pole. A weak, patchy staining pattern only is visible (*top left*). A large transparent ERM is removed (*top centre*). Delicate folds in the macula persist (*top right*). Another ICG injection is performed and a homogeneous staining of the inner retinal surface achieved (*bottom left*), which allows atraumatic and complete ILM removal (*bottom centre, right*)

tion for the purpose of visualizing the vitreoretinal interface was a breakthrough and allows easy and safe peeling manoeuvres (Fig. 3.12).

ICG is a frequently used dye with a long history of safety after intravenous administration, e.g. for ICG angiography. It binds quickly to proteins, particularly to serum albumin. Leakage in the retina and beneath the RPE has never been shown to cause any tissue damage. Intact cell membranes do not allow the dye to enter the cells. Furthermore, ICG has been introduced in cataract surgery to stain the lens capsule and to allow a controlled capsulorrhexis in difficult cases. In anterior segment surgery no toxic effect has been demonstrated, neither in vitro nor in vivo [34, 35].

ICG dye for vitreous surgery was described first in 1999/2000 [39, 37, 10]. At first, this technique was proposed for ILM peeling in MHS. The idea was taken up quickly and enthusiastically by retinal surgeons and has become routine in most operating rooms. Meanwhile so-called ICG-guided vitreous surgery has expanded to other indication groups, such as macular pucker, proliferative vitreoretinopathy, and macular oedema of various origins. It has been proven to stain vitreous collagen and the ILM selectively, but not epiretinal proliferative tissue [25, 10] (Fig. 3.12). Recently, however, a degree of uncertainty and scepticism arose when certain clinical studies and experimental data hinted at the potential toxicity and other side effects of intraviteally applied ICG.

Experimental studies showed possible RPE cytotoxicity, which may be especially relevant in MHS, where the RPE is directly exposed to the dye [76, 82, 33, 43]. Morphologic findings after experimental intravitreal injections into non-vitrectomized rat eyes [17] and ultrastructural findings of surgically excised ILM material [26, 27] suggest potential retinal toxicity. Clinical data on possible ICG-related side effects showed unusually frequent RPE changes after MHS [18]. Furthermore, less favourable functional outcomes after both MHS and pucker surgery were observed in ICG-exposed eyes compared to those without using the adjuvant [29, 30]. Other studies did not confirm a negative impact on visual results after macular pucker and MHS [81, 42, 69].

The current scientific information on ICG toxicity is difficult to evaluate. There are two major problems: (1) there is no agreement as to

how to apply the dye and what kind of ICG preparation should be used. Thus, its application is in fact experimental. Concentration of ICG solution varies from 0.05% to 0.5%. Various amounts of aqueous solvent and subsequent dilution either with BSS or glucose are used. Some surgeons irrigate the solution over the retinal surface in a BSS-filled eye; others inject the dye after fluid/air exchange. Since ICG binds to the retinal surface very quickly, most surgeons immediately rinse the eye and remove the surplus. Others suggest a prolonged contact time up to 2 min in order to increase the staining intensity. (2) Adverse effects indicating retinal or RPE toxicity were found in experimental settings. Prolonged exposure times of ICG solution to the retinal surface (>5 min) and high concentrations (0.5 mg/cc and more) were necessary to achieve pathologic alterations. Hyposomolarity of conventionally prepared ICG (ICG powder dissolved in aqueous and diluted with BSS) has shown in vitro toxicity to the RPE, suggesting that isotonic infracyanine should be used [82].

Such experimental conditions are hardly comparable to the clinical situation. This is why the majority of surgeons suggest that ICG is safe for clinical use. Large randomized comparative series with a standard surgical technique and specific protocol of ICG preparation are warranted. We worked on various ICG solutions to achieve a pH-neutral, iso-osmolar 0.1% solution. We inject only as much as is needed and immediately wash out the dye. Careful observation has shown no anatomic or functional side effects. This is in agreement with other comparative clinical studies [42, 69].

Some other new ICG-related phenomena were observed after intraocular injection for pucker or MHS: Infrared scanning laser ophthalmoscopy revealed that retention of the dye was demonstrable over months. No negative effect on retinal function was observed, but the long-term clinical significance remains unclear and should be carefully looked at [90, 1, 50, 86]. Photocoagulation with an infrared (810-nm diode) laser showed an enhanced uptake of laser energy in stained areas, resulting in more intense and superficial laser burns [4]. Similarly, potential light toxicity dependent on the spectral output of various light sources utilized for vitrectomy must be considered. An overlap between the absorption of ICG and the emission curve of the fibreoptic light (600–800 nm) may result in a photodynamic effect [31, 27].

Summing up the current toxicity controversy: The scientific data on ICG-assisted vitreous surgery is inconsistent. Nonetheless, we can draw some preliminary conclusions from the aforementioned studies. ICG is not an inactive/inert substance, but has potential side effects. Toxicity and less clearly defined adverse effects related to the intravitreal application of ICG have been seen to correlate with high concentrations, use of hypo-osmolar preparations, long exposure time to the retina, direct contact to the RPE layer, and prolonged endoillumination by endo-optics. Negative effects may accumulate and actually cause retinal/RPE damage in some eyes [32].

Unquestionably, the technique of ICG-assisted vitreous surgery facilitates controlled peeling of ERM and the ILM and makes proper and atraumatic manoeuvres easier, safer, and faster, leading to better anatomic results. The staining pattern allows differentiation between vitreous cortex and ILM (positive staining) and epiretinal gliosis (negative staining). Since ERMs are characterized by a negative ICG staining pattern, a repeated dye application after epiretinal membrane peeling may be useful to outline residual proliferative tissue and the ILM. We prefer this method because we intend to remove both tissue structures, the ERM and the ILM during pucker surgery. Doubtlessly, the peeling manoeuvre is more complete in ERM surgery as well as in MHS.

Furthermore, ICG-assisted vitrectomy provides new information about clinically applicable vitreoretinal relationships. The clinical advantages are striking and surgeons will not do without it once they have performed ICG-guided vitreous surgery. However, the window of safety is not exactly known. It is urgent to work on the critical question of ICG toxicity. Further investigations must result in a standardized protocol for ICG preparation and recommendations for its application. According to our clinical experiences, the benefits will certainly outweigh the risks.

3.6.2 Trypan Blue

As ICG was introduced for capsular staining in complicated cataract surgery, trypan blue was concomitantly suggested for the same purpose [54]. Trypan blue 0.3% has been used for many years to examine endothelial cell viability of organ-cultured corneas prior to corneal transplantation without evidence of toxicity. Sufficient visualization of the lens capsule during capsulorrhexis in eyes without a red reflex is achieved using a 0.1% solution. Meanwhile, 0.06% trypan blue is commercially available for this purpose.

As with ICG, the option to use trypan blue for dye-enhanced vitreous surgery came into being. Veckeneer et al. [89] investigated trypan blue in vitro and found a dose-dependent staining of ERM. Furthermore he performed toxicity studies in rabbit eyes in a gas-compression vitrectomy model. At 4 weeks after intravitreal injection, he found no morphologic or functional alterations using 0.06% trypan blue. However, after having given 0.2%, damage to the outer retina was observed. Stalmans [83] did not detect any sign of toxicity in human RPE cultures up to 0.3%. Based on the current scientific data, 0.15% trypan blue solution is now commercially available for vitreous surgery in Europe.

The application method for trypan blue during vitrectomy is relatively uniform: a sufficient staining intensity is achieved by applying a few drops of the dye onto the macula or any area with ERM in a temporarily air-filled eye. Some surgeons wait for 1 min; others aspirate the surplus immediately before an air-BSS exchange is done. If trypan blue is used in a fluid-filled eye, the dark colour of the dye inhibits visualization of the intraocular structures and the increased dilution gives a less favourable surface staining.

Clinical experience has shown that staining of ERM is far more intense than staining of the ILM or vitreous remnants. Furthermore, the colouring intensity was found to be variable in different clinical situations, e.g. trypan blue staining in fresh, immature membranes was less intense in comparison to mature membranes [19]. Thus, the staining pattern of trypan blue is different from ICG and less specific. The latter has shown a strong affinity to vitreous gel and the ILM, but spares proliferative tissue. Nonetheless, there is general consensus that improved visualization and delineation of membranous structures is achieved, which enables the surgeon to perform more complete tissue removal and minimizes the risk of inadvertent damage to the retina.

Clinical studies on trypan-blue-assisted peeling manoeuvres include macular pucker, PVR and MHS [19, 64, 65, 44]. No apparent side effects were observed. Histologic and immunohistologic findings on specimens obtained intraoperatively did not differ from those removed without the aid of dye [44]. Although trypan blue seems to be safe in the aforementioned clinical settings, further studies on its safety are required.

3.6.3 Double Staining Technique

Trypan blue and ICG may have complementary staining properties: ICG binds more selectively to the ILM, whereas trypan blue shows a high affinity to mature ERM. Based on these experiences, Stalmans reported on a double-staining technique using both vital dyes [84]. Intraoperatively, epiretinal tissue and the ILM were clearly distinguishable. This clinical observation was confirmed by histologic examination of the excised tissue. Whether the double staining technique is a tool to achieve optimal anatomic results and presumably improve functional recovery needs to be investigated by additional studies.

3.6.4 Triamcinolone Acetonide

Within the past 3–4 years, intraocular injection of triamcinolone acetonide (TCAC) is increasingly being used for various ocular diseases. Apart from its therapeutic anti-inflammatory and anti-exudative effects, intraoperative application of the crystalline particles has been useful to visualize the transparent vitreous and to obtain a more complete vitreous separation and

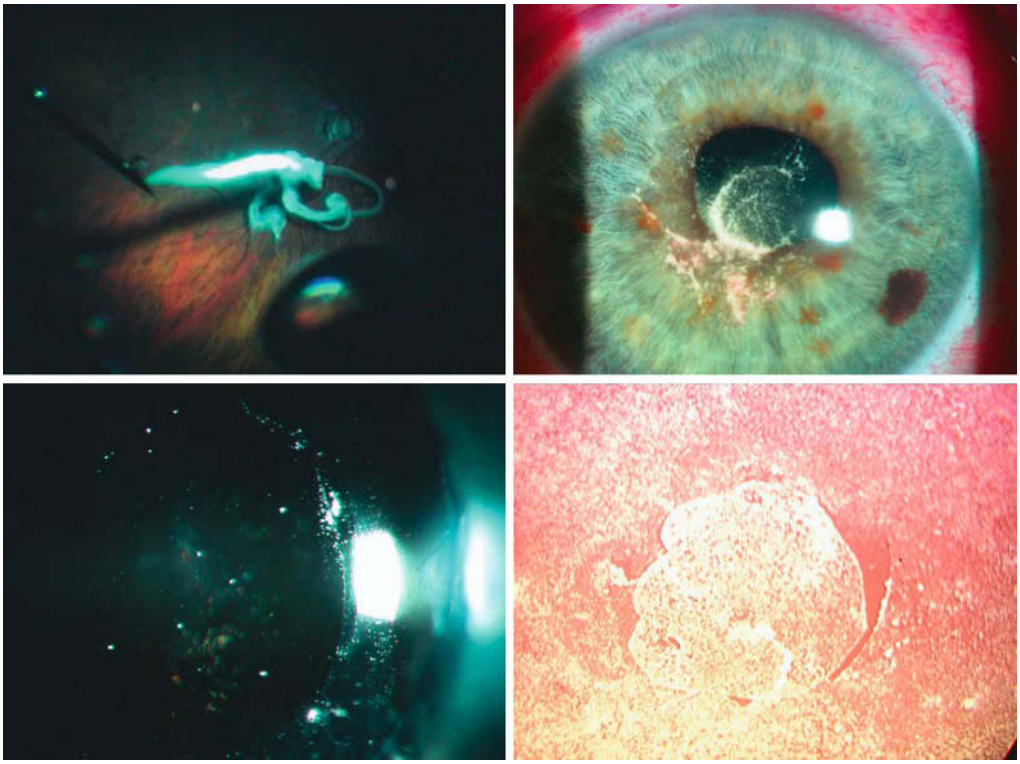

Fig. 3.13. Triamcinolone acetonide as an adjunct to vitreous surgery: Injection at the end of the procedure (*top left*). The cortisone crystals may enter the anterior segment, particularly if combined lens/vitreous surgery is performed (*top right*). Slit-lamp examination on day 1 after surgery usually shows minimal surgical inflammatory reaction (*bottom left*) and fundus examination reveals particles on the retinal surface dependent on the patient's position. Note that the ILM peeled area is outlined (*bottom right*) by accumulated crystals

removal [66]. This has proven a highly valuable tool in eyes with pathologic vitreoretinal adherence (high myopic and diabetic eyes). Recently, TCAC-assisted vitrectomy was applied to pucker, MH and PVR cases also in order to visualize the posterior hyaloid, cortical remnants, and to outline ERM and peeled areas [71, 24, 40]. Adverse effects were not observed. On the contrary, there was evidence that postoperative breakdown of the blood-ocular barrier was significantly lower in comparison to vitrectomy without TCAC [71] (Fig. 3.13). Postoperative inflammatory response may be reduced by residual TCAC left in the eye. TCAC is a useful adjunct during vitrectomy for selected cases; however, its supplementary anti-inflammatory effect remains to be established.

Summary for the Clinician

- **Complete traction relief during pucker surgery is believed to be a precondition for good functional results. Intended ILM peeling is a tool to achieve this goal, but remains a technical challenge to the surgeon**
- **Visualization aids (ICG, trypan blue, triamcinolone) enable the surgeon to perform a more complete tissue removal and minimize the risk of inadvertent retinal damage**
- **From the practical point of view, dye-enhanced membrane peeling facilitates controlled and meticulous peeling manoeuvres on a delicate transparent**

structure. Extensive surgical experience and perfect optic conditions can hardly compensate for improved visualization and tissue identification by their specific staining patterns

- **From the scientific point of view, intravitreal application of dyes, particularly ICG, is still under debate, because the window of safety is not proven**

3.7 New Indications for ERM and ILM Peeling

3.7.1 ILM Peeling in PVR Surgery

ERM formation is pathognomonic for PVR detachment and frequently involves the macular area. Recurrent membranes occur in up to 30 % and remain the major cause of failed PVR surgery. Thorough and complete membrane removal is crucial to reattach the retina and to reduce the incidence of recurrent PVR. On the assumption that complete membrane removal will be achieved using ILM peeling techniques, and that this technique results in maximal traction relief, it seems a logical approach to consider ILM peeling in PVR cases.

Based on these considerations, Lucke first reported his clinical experiences [46]. ICG was used to distinguish between ILM and preretinal membranes. We observed the following intraoperative peculiarities: sand dunes, indicating intraretinal shrinkage and stiffening that disappear after posterior ILM removal. Enhanced relaxation results in a smoother and easier reattachment. The ICG staining pattern in the peripheral retina allows the identification of areas in which epiretinal membranes are present or not. Restaining can outline residual membrane edges that necessitate further peeling. In posterior PVR with starfolds outside the vascular arcades, one may be able to remove the ILM adjacent proliferative tissue. Initially, distorted retinal tissue flattens, while afterwards there is no visible contraction.

In addition, several postoperative findings were noted: No eye revealed an epimacular gliosis, which otherwise is a frequent finding (10 %)

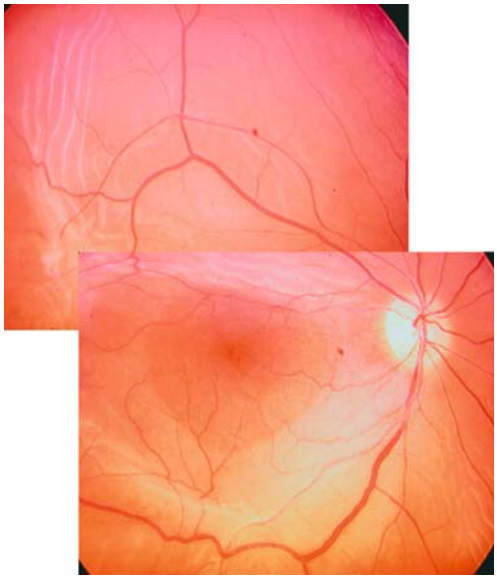

Fig. 3.14. PVR C2 after vitrectomy for rhegmatogenous RD. Prior surgery had been performed with ILM peeling. Note the macula appears normal and is attached. VA was 1.0. The remaining retina is completely detached

after conventional PVR surgery. Recurrent PVR developed in a few cases. This was caused by new membrane formation in areas previously not peeled. In other words, no recurrent membranes were seen in those areas of ILM delamination, neither centrally nor peripherally (Fig. 3.14). Some eyes with recent-onset, but advanced, PVR with macula involvement regained surprisingly good visual acuity (0.3–0.5). Our 2-year experience using this technique shows that ICG-assisted PVR surgery facilitates more thorough membrane removal and effectively prevents recurrent membrane formation. Most importantly, improved anatomic restoration of the central retina obviously results in improved functional recovery.

Trypan blue is another substance used for dye-assisted PVR surgery. The staining pattern of trypan blue applied to the vitreoretinal interface differs from the ICG staining pattern. A pilot study on ten eyes by Feron et al. [19] stated that trypan blue created a sufficient contrast between stained ERM and a non-stained retinal surface to facilitate precise and safe ERM peel-

ing. However, staining intensity was found to be variable, particularly in early PVR with immature membranes. This pilot data was supported by several others, who have recently presented their clinical findings at international vitreoretinal meetings.

3.7.2 ILM Peeling During Vitrectomy for Rhegmatogenous Retinal Detachment

Cellophane membranes and macular pucker after surgery for retinal detachment (RD) is a common finding. The reported incidence varies considerably, depending on the method of investigation: Clinically obvious pucker formation occurs in 4–8%, whereas careful ophthalmoscopy reveals about 46% and the histology of eyeballs shows ERM formation in 76% [41, 91]. The functional significance of minor ERM in eyes with initially macular-off detachments is difficult to estimate. Such eyes rarely regain full vision. However, metamorphopsia is a relatively reliable symptom in ERM-related pathologies.

Currently, there is an exciting debate about the most advantageous treatment for RD: buckle or primary vitrectomy? Although the question remains to be answered, there is a clear trend towards vitrectomy not only for complicated, but also for so-called simple, retinal RD.

In our institution, all RD patients over 50 years and all pseudophakic eyes are treated by vitreous surgery. ILM peeling as an additional manoeuvre was introduced in 2002. A comparative analysis (consecutive series, single surgeon) was carried out to compare the results without (group A) and with ILM peeling (group B). Baseline data (age, sex, preoperative vision) were similar in both groups with the exception of a shorter follow-up and a higher rate of simultaneous cataract surgery/IOL implantation in the ILM-peeling group (group B).

Major findings are as follows (group A vs. group B): follow-up was 21 months and 6 months. Primary reattachment was achieved in 90% and 93%, final attachment in 100% each. Postoperative PVR occurred in 6.7% and 7.7%. Median visual acuity improved from 0.08 to 0.39 and from 0.05 to 0.4. At the end of follow-up, 93% of eyes were pseudophakic in both groups.

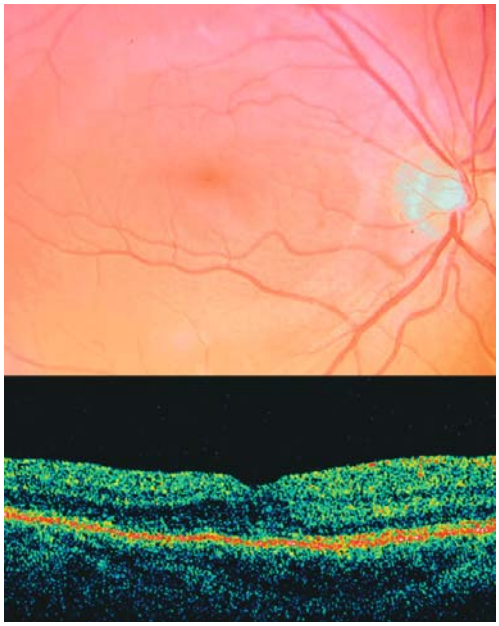

Fig. 3.15. Fundus and OCT picture of an eye 6 weeks after vitrectomy for rhegmatogenous macula-off RD. VA has improved from LP to 0.8. The fundus appears normal with no sign of macular oedema or ERM formation. OCT findings show minor tissue irregularities within the foveola pit, but otherwise a normal profile

Significant differences in both groups are found by looking more closely at the data. Taking into account the considerably shorter follow-up in group B, visual recovery obviously occurs much faster in ILM-peeled eyes. Furthermore, clinically significant macular alterations were found in 20% of group A (macular oedema, ERM), but only 5% of group B (macular oedema only). Some particular findings are depicted in Figs. 3.14 and 3.15.

The main conclusions from this pilot study are: ILM peeling during vitreous surgery for RD may lead to fast visual recovery, reduce the incidence of postdetachment CME and prevent ERM formation. However, PVR cannot be avoided and seems unaffected by additional ILM peeling. Since visual recovery after macular-off RD takes many months, long-term fol-

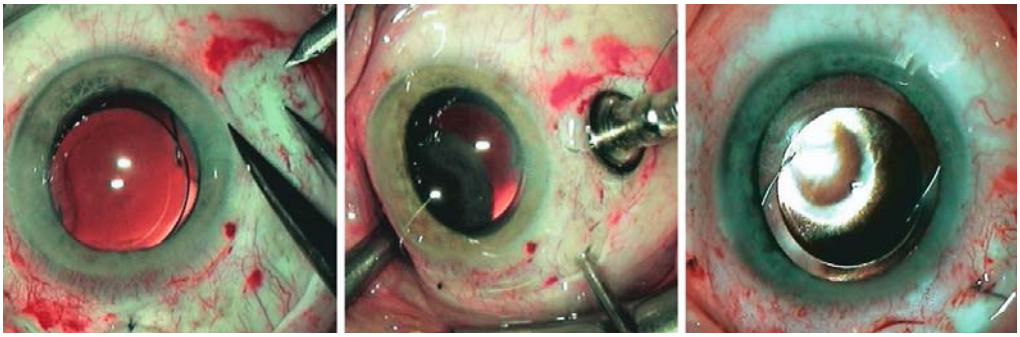

Fig. 3.16. Scenes from a combined lens surgery/vitrectomy procedure: After the phako/IOL part, surgery is continued in the posterior segment. Optic media are absolutely clear. The eye is stable even in the case of deep indentation. Corneal transparency and IOL position are maintained until the end of surgery with air tamponade

low-up is necessary to assess the potential of ILM peeling for the anatomic restoration and visual recovery. It is important to mention that primary macula-on RD cases will also profit from fewer secondary macular complications, such as oedema and ERM.

Summary for the Clinician

- **Epimacular ERM formation is a common finding in postdetachment eyes, and ubiquitous epiretinal growth is pathognomonic for PVR**
- **Preliminary results on removal of epimacular cellophane membranes and the ILM during surgery for rhegmatogenous retinal detachment indicate an improved functional outcome and the prevention of postoperative puckers**
- **Intended ILM peeling during surgery for PVR results in an enhanced traction relief in comparison to ERM removal only and shows no membrane recurrences in those areas in which the ILM has been removed previously**
- **Whether improved anatomic macular findings observed after ILM peeling in RD and PVR cases correlate with less cystoid macular oedema and better visual results remains to be investigated**

3.8 Surgery for ERM and Cataract

The vast majority of patients designed for ERM removal are 50 years and older. Subsequent cataracts will occur in up to 100 % [53, 12]. In our experience, final and permanent VA is achieved 3–6 months after pucker surgery. By the time the retina recovers and vision improves, cataract development begins and contributes to substantial visual loss again. In primary phakic eyes, stable postoperative vision is usually not attained until cataract surgery is performed. For these reasons we strongly recommend simultaneous lens surgery combined with vitrectomy for ERM cases.

Combined phaco/in-the-bag-IOL/vitrectomy is a routine procedure in our institution. Within the past 10 years, more than 3,500 such procedures have been carried out. An institutional analysis for selected indications (diabetic vitrectomies, pucker surgery, retinal detachment) has shown that outcomes are com- parable to those for sequential surgery. This was confirmed by some small case series [75, 80]. With regard to surgery for mERM, we found that visual results were similar in phakic eyes treated with combined phaco-/vitrectomy and primary pseudophakic eyes that had vitrectomy alone. Since postoperative macular oedema occurred in both groups equally, this could not be attributed to the phaco/IOL part of surgery.

A combined approach offers several further advantages: Firstly, cataract surgery using small incision techniques and foldable IOLs is easy and almost non-traumatic to the anterior segment (Fig. 3.16). Cataract surgery in vitrectomized eyes with an advanced nuclear sclerosis, however, is unquestionably more challenging and includes a higher risk of capsular complications. Secondly, lens surgery preceding the vitrectomy part provides an excellent view of the fundus. In my opinion, visualization of the macular structures is even better than in phakic eyes with clear lenses. Thirdly, from the patient's viewpoint, ocular morbidity and visual rehabilitation are considerably reduced. Simultaneous surgery offers the possibility that the initial problem the patient presents with, which is the ERM, can be solved with a single surgical procedure. From the patient's viewpoint, the experience of visual deterioration again as a result of lens opacification, waiting for a second operation to be done and a second phase of postoperative care – these unpleasant time-consuming events can be avoided by a primary combined surgical approach.

Summary for the Clinician

- **Cataract is the most frequent cause of visual deterioration after ERM surgery. Patients profit from stable and permanent visual improvement only once cataract surgery is performed**
- **Combined small-incision phaco- with foldable lenses and vitrectomy maximizes surgical visualization of the posterior segment and avoids the need for patients to undergo a second procedure for cataract extraction soon after vitreoretinal surgery**
- **We recommend vitrectomy in conjunction with phaco/IOL in all eyes with cataractous lenses and all patients aged over 50 years, even if they show a clear lens**

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Is There Sufficient Evidence to Support Transpupillary Thermotherapy for Age-Related Macular Degeneration?

4

Johann Roider

Core Messages

- Based on current data there is little evidence for the superiority of transpupillary thermotherapy in age-related macular degeneration (AMD) over conventional laser treatment
- It cannot be ruled out that most authors describe the natural course of occult choroidal neovascularisation (CNV) or complications of "overtreatment"
- Transpupillary thermotherapy may be effective in the future, when a heat-specific sensibility of choroidal neovascular tissue has been shown and temperature control of the tissue during irradiation can be achieved by online feedback mechanisms

4.1 Introduction

Age-related macular degeneration (AMD) is the leading course of blindness in the Western world. Choroidal neovascularisation (CNV) is the reason for a rapid loss in visual acuity in most cases. Established therapeutic options are laser treatment, surgery and PDT. In most cases therapeutic options exist only for classic choroidal neovascularisations. However, more than 80 % of CNVs are occult choroidal neovascularisations. Conventional continuous wave (CW) laser photocoagulation is mostly used for extrafoveal choroidal neovascularisations. A complete destruction of all retinal layers is the price for a cessation of the growth of the choroidal neovascularisation. Transpupillary thermotherapy (TTT) is a new treatment which was first published by Reichel et al. in 1999 [16]. Sixteen eyes with occult CNV were treated by transpupillary thermotherapy. The follow-up period varied between 6 and 24 months. Three out of 16 eyes were classified as better, 9/16 as equal and 4/16 as worse [16].

4.2 Mechanism of Conventional Continuous Wave Photocoagulation and Transpupillary Thermotherapy in AMD

Using CW laser photocoagulation of choroidal neovascular membranes, the area of CNV is irradiated by a laser beam of typically between 200 and 500 µm. In most cases a green laser (514 nm or 532 nm) is used. Pulse durations are typically between 200 and 500 ms. Laser power is adjusted to produce a chalk-white lesion. A laser power of 500–1,000 mW is usual. With these laser parameters the CNV is confluently treated spot by spot.

These laser parameters correspond to a power density of 200–400 W/cm². If the laser beam is focused on the CNV, laser energy is absorbed by the melanin granules, mainly located within the retinal pigment epithelium (RPE) and additionally located in clusters within the choroid. Laser energy is transformed to heat and all surrounding tissues are thermally destroyed. The goal of the laser treatment is thermal necrosis of the choroidal neovascular complex. Based on the power settings, the temperature within the tissue can be calculated relatively accurately [7,

18]. With power settings of, e.g. 200 W/cm², peak temperatures of 100–150 °C occur within the tissue, especially within the CNV [6, 18].

Summary for the Clinician

- **Conventional CW laser photocoagulation is mostly used for extrafoveal choroidal neovascularisations**
- **The goal of such laser treatment is thermal necrosis of the choroidal neovascular complex**

In transpupillary thermotherapy (TTT) of AMD, an infrared (810 nm) diode laser is used. Exposure time is significantly longer, typically 60 s. In TTT the whole area is treated by a single large laser port, e.g. of 2 mm diameter. Laser energy should be chosen so that no blanching or only a slight greying of the retina is seen during treatment. A power of 500 mW is usual for such a spot diameter. These energy settings correspond to a power density of about 12.5 W/cm². Due to the linearity of the heat conduction equation

$$\frac{\partial T}{\partial t} - \alpha \nabla T(r,t) = \frac{S(r,t)}{\rho c}$$

the peak temperature is related linearly to the power density [8, 18]. Therefore peak temperature elevations of about 5–10 °C can be estimated in the tissue where laser energy is being absorbed. Temperature elevations of 5–10 °C do not regularly produce a thermal coagulation.

4.3 Clinical Results of Transpupillary Thermotherapy

Initial results were published by Reichel and coworkers in 1999 [16]. Reichel treated 16 eyes with occult CNV by transpupillary thermotherapy. The follow-up period varied between 6 and 24 months. Three out of 16 eyes were classified as better, 9/16 as equal and 4/16 as worse. Preoperative visual acuity was below 0.125 in 11/16 and 0.25 in 4/16. Newson et al. published similar results in 2001 from 44 eyes [15]. They treated both classic (n=12) and occult CNV (n=32). With a mean follow-up period of 6 months about 75 % were occluded. In the follow-up period a variety of studies have been published. Table 4.1 summarises the main results from various papers.

Hardly any of the papers report prospective or controlled studies. No studies are randomised, nearly all being retrospective analyses. Nearly all papers report a follow-up period of 6–12 months. A common finding by these studies is that visual acuity is stable in about 60 % and worse in about 25 % after 6 months. After 1 year the situation has become worse. Only 30–40 % of eyes are only stable in visual acuity.

Despite the initial findings of the harmless nature of this treatment, a variety of complications are described in the literature. Algevre described complications in 46 out of 113 eyes [2]. The most common findings were progressive

Table 4.1. Visual acuity after treatment of choroidal neovascularisations by transpupillary thermotherapy (*n.i.* no information)

	Year of publication	Eyes	Follow-up period (months)	Equal (<3 lines)	Worse (3 lines)
Reichel et al. [16]	1999	16	6–24	56 %	25 %
Rougier et al. [20]	2002	45	n.i.	>60 %	n.i.
Kim et al. [11]	2002	58	6	72 %	n.i.
Sanders et al. [21]	2002	78	12	n.i.	43.3 %
Algevre et al. [2]	2003	113	12	41 %	51 %
Haas et al. [10]	2003	14	18	11 %	55 %
Atarashi et al. [4]	2004	19	8.8	64 %	18 %

fibrosis in 18 eyes and atrophy of the RPE in 13 eyes [2]. Chorioretinal atropy was also described by Lanzetta [12] and Auer [1]. Benner has described marked whitening and closure of the perifoveal capillaries in two cases [15]. Yamaji described a closure of the retinal capillaries in 48 % of 48 patients treated [24]. Even occlusion of choroidal vessels was described. Dosimetry is repeatedly described as a problem in daily routine. Blanching of the retina or choroidal bleeding can occasionally be found.

4.4 Discussion

Transpupillary thermotherapy (TTT) is a novel therapy. However, its value in clinical practice is unclear. Despite the lack of controlled studies it is widely used. The application in daily routine is based on small studies, which are all short-term, non-randomised and in most cases retrospective studies. Figure 4.1 shows the spontaneous course of classic and occult CNV over a period up to 5 years. These data are based on the TAP and MPS studies, all well-controlled, prospective and randomised studies (e.g. [22], [13]). After 6 months about 60 % of the patients were stable within two lines. After 1 year about 44 % were stable. These data are not much different from the data which are presented by the publications on transpupillary thermotherapy.

Some publications describe significant damage to the retina after TTT as mentioned above. The interaction mechanism of transpupillary thermotherapy is under debate. Based on the physical parameters a uniform heating of all tissues around the CNV can be assumed. Figure 4.2 shows the axial temperature profile from milliseconds up to seconds. It can be assumed that over a duration of 60 s, as used in transpupillary thermotherapy, after only a few seconds all the tissues around the choroidal CNV are uniformly heated and held at a fixed temperature for 60 s. This means that no temperature-specific selectivity, or only a marginal small specific tissue effect, exists. There is no specific spatial selectivity between the choroidal neovascular tissue and the adjacent neural retina and choroid. Based on the physical parameters, temperature elevations of about 5–10 °C seem to be reasonable [14, 18]. The temperature elevations are in accordance with current temperature models of the retina, which are also validated by animal models [6]. Temperature elevations of 5–10 °C are on the border of thermal denaturation of biological tissue, as can be derived by Arrhenius' law [3].

Such threshold effects can produce tissue damage or no damage. Threshold effects are those which produce a biological response in 50 %. In considering of these physical facts, the clinical findings as described in the literature can be explained. In many cases no biological tissue effects occur, which leads to a spontaneous course of CNV after treatment. The 'success rates' of TTT of 60 % after 6 months (stabilisation of visual acuity) are in accordance with the natural course. In other cases a tissue effect occurs. This leads to thermal tissue necrosis of all tissues of the retina. The findings of chorioretinal atropy, closure of the retinal capillaries, occlusion of choroidal vessels or blanching of the retina can easily be explained by thermal damage, as a result of the uniform heat conduction to surrounding tissues.

Recently the production of heat shock proteins has been claimed as being the basis for the effectiveness of transpupillary thermotherapy [14]. Heat shock proteins (e.g. HSP70) are not retina-specific proteins. They are produced everywhere and have been well investigated in all fields of medicine, e.g. in lesions after burns of the skin. HSPs are produced if the cell is elevated to temperatures of up to 42 °C for a short period. The role of heat shock proteins is to protect from cell damage. During temperature elevations HSP70 proteins facilitate the unfolding process of proteins within the endosplasmic reticula. During irradiation of the retina of rabbits, HSP70 proteins have been demonstrated within the choroid and retina with laser settings of 6.8–10.5 W/cm² [19]. In rabbits lesions of 10.5 W/cm² always produce retinal damage. These findings again are in accordance with the theory of transpupillary thermotherapy as threshold treatment but do not support a TTT-specific effect. Based on current knowledge no specific production of heat shock

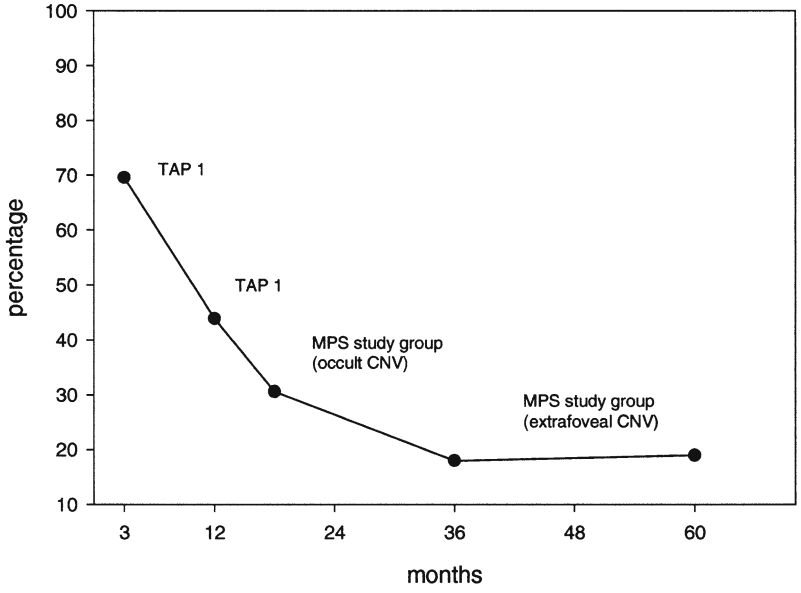

Fig. 4.1. Percentage of patients with stable visual acuity (VA <3 lines) with classic or occult CNV. No treatment has been performed (natural course). Data are based on the TAP and MPS studies. After 1 year even in classic CNV 45% of patients still have the same visual acuity

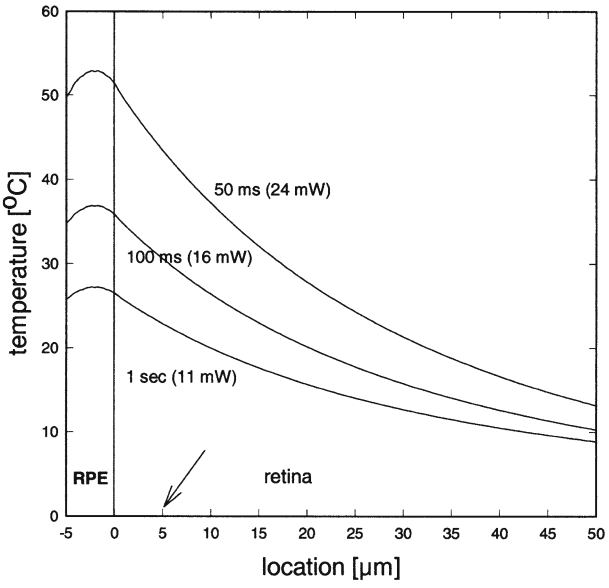

Fig. 4.2. Calculated temperature distribution within the RPE and the neural retina after application of laser pulses of 50 ms, 100 ms, and 1 s. Laser power has been chosen to produce the same thermal effect (slight grey lesion). With increasing irradiation time spatial selectivity is being lost

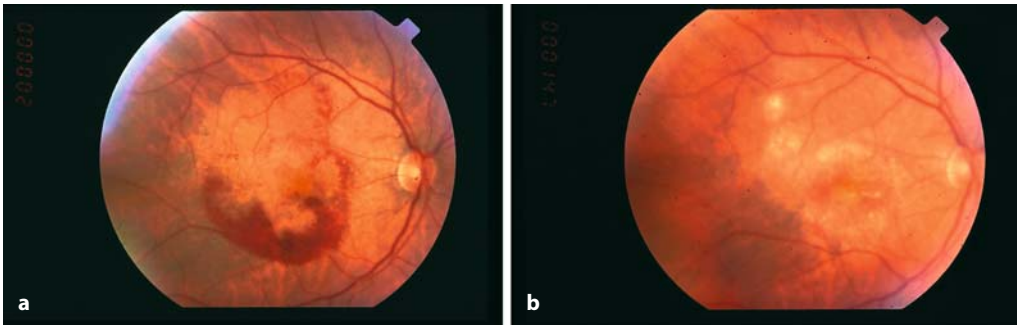

Fig. 4.3. Preoperative fundus image of an occult CNV (**a**). The retina has been lifted off and the CNV has been irradiated by a long diode laser pulse. The retina remained unaffected during laser treatment. One month later (**b**) the patient has developed chorioretinal atrophy

proteins is known in choroidal neovascular tissue.

If transpupillary thermotherapy is a threshold treatment, the target tissue may be the RPE and the choroid. The melanin granules within the RPE and choroid have a high content of melanin granules. At 810 nm more than 60 % of laser energy is absorbed within the RPE and choroid. It is reasonable to assume that the RPE and choroid will always be affected. If an effect on choroidal neovascular tissue occurs, complete damage of the RPE and choroid will always lead to consecutive damage of the retina, even if the neural retina is initially undamaged. This has been demonstrated by artificial lifting off the neural retina by vitrectomy and consecutive irradiation of the neovascular choroidal complex by a 1-s-long pulse of laser light of 810 nm [17]. The retina itself is transparent for 810 nm. As a result, in all cases RPE atrophy has been concomitant with a decrease in visual acuity (see Fig. 4.3).

Summary for the Clinician

- **The information on TTT is based on retrospective analysis**
- **The most common findings are progressive fibrosis and atrophy of the RPE**
- **There is no specific spatial selectivity between the choroidal neovascular tissue and the adjacent neural retina and choroid**
- **The success rates of TTT of 60 % after 6 months are in accordance with the natural course of occult CNV in AMD**
- **If an effect of TTT on the choroidal neovascular tissue occurs, complete damage of the RPE and choroid will always lead to consecutive damage of the retina, even if the neural retina is initially intact**

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Retinal Pigment Epithelium and Choroid Translocation in Patients with Exudative Age-Related Macular Degeneration

5

Jan C. van Meurs

Core Messages

- There is no evidence based treatment for patients with age-related macular degeneration with a predominantly occult choroidal subfoveal membrane, with or without submacular blood
- Existing evidence based treatments, such as laser and photodynamic therapy, may decrease visual loss, but fail to improve vision
- With "simple" choroidal membrane removal we also remove the subfoveal retinal pigment epithelium (RPE), which is necessary for functioning of the overlying macula
- Macular rotation surgery may result in improved vision at the risk of sight-threatening surgical complications
- The ideal reconstitution for a monolayer of differentiated RPE cells is at present best achieved either by macular rotation or by autologous RPE and choroid translocation
- Future developments for RPE reconstitution may include using an artificial membrane as substratum to be repopulated by RPE improved by gene transfer or stem cells

5.1 Introduction

5.1.1 Epidemiology

In the industrialized countries, age-related macular degeneration (ARMD), the end stage of age-related maculopathy (ARM), is the principal cause of irreversible legal blindness in elderly persons [20, 21]. This end-stage disease occurs in two forms atrophic and exudative; the exudative form leads more quickly to a deeper and larger scotoma and is moreover twice as common as the atrophic form. Bilateral involvement may develop in 40 % of patients over a period of 5 years. Based on large population based studies in the USA, Australia and the Netherlands, a reasonable overall estimate of the prevalence of end-stage macular degeneration is about 1 % in persons aged 65–74 years, increasing to 5 % in persons aged 75–84 years and 13 % in persons 85 years or older [21, 42].

With 1.82×10^6 persons aged 65–74 years, 0.77×10^6 persons aged 75–84 years and 0.23×10^6 persons aged over 85 years in the Netherlands, a country with 16.19×10^6 inhabitants in 2003, it is clear that particularly exudative macular degeneration causes considerable human suffering and loss of quality of life in a large number (approximately 66,000) of persons.

To assess treatment capacity requirements, it may be more realistic to study incidence figures as they represent the more acute patients that may still have a stage of disease amenable to treatment.

Extrapolating the incidence rates found in the Rotterdam Study to the general population

in the Netherlands (16.19×10^6), we can estimate the 5-year incidence risk of neovascular ARMD to be 7,287 persons aged 65–74 years and 15,480 persons aged 75 years and older. In round figures this represents 4,550 new patients every year, with bilateral involvement in 1,820 patients.

Summary for the Clinician

- **Exudative macular degeneration causes severe visual loss in 7.5 % of the population older than 75 years in the industrialized countries. In the Netherlands, a country with 16 million inhabitants, every year 4,550 persons will develop exudative ARMD, of whom 1,820 persons have bilateral involvement**

5.1.2 Pathology

Although the aetiology and pathogenesis of ARMD are not yet fully understood, the resulting pathology is well defined [5]. In the exudative form, choroidal neovascular ingrowth occurs under the retinal pigment epithelium (RPE) and through the RPE under the retina, causing a haemorrhagic RPE and retinal detachment and eventually a fibrovascular scar with subsequent dysfunction of the overlying neurosensory retina (fovea, macula). In the atrophic form a gradual loss of submacular RPE cells finally leads to macular dysfunction.

Summary for the Clinician

- **In exudative age related macular degeneration, choroidal neovascularization not only invades the subretinal space, but also grows under the RPE**

5.2 Treatment Approaches to Exudative Age-Related Macular Degeneration

5.2.1 Non-surgical Interventions

5.2.1.1 Argon Laser

Argon laser photocoagulation was shown to be better than no laser in randomized controlled trials for extra- and juxtafoveal neovascularizations [6, 25]. Unfortunately, even with the combined use of fluorescein angiography and indocyanine green angiography, at best only 15 % of all patients presenting with submacular neovascularization may be eligible for such treatment [16]. Within 2 years recurrent neovascular membranes occurred in 50 % of the patients, typically towards the fovea.

For some subfoveal lesions a treatment benefit was demonstrated after 2 years, but the immediate central scotoma following laser treatment has meant that this treatment is not widely applied [29].

5.2.1.2 Photodynamic Therapy

While photodynamic laser therapy in randomized controlled trials has been shown to be better than no treatment in patients with predominantly classic and some with occult neovascular membranes, it has only limited vision loss without restoring vision and often requires multiple retreatments [10–12]. Transpupillary thermotherapy [4] and radiation therapy [7] suffer from the same drawbacks as the abovementioned modalities. With these treatment limitations, researchers are developing alternative treatments, including medical treatment and surgery:

5.2.1.3 Antiangiogenesis

A randomized, placebo-controlled trial on the use of subcutaneous octreotide, which principally affects vascular leakage but also angiogenesis, just failed to show a significant 1-year treatment benefit (Seerp Baarsma, verbal communication, January 2004). Phase II trials of peribulbar anecortave acetate, an antiangiogenic steroid, showed promising results, although the lack of a dose-response effect remained puzzling. Phase I and II studies of intravitreal injections of an antivascular endothelial growth factor (VEGF) antibody (Arvo Abstracts 2003, 9720) or an anti-VEGF aptamer report some improvement of vision in some patients, particularly in combination with photodynamic therapy (PDT) (Eyetech Study 2003). In these studies, the number of retreatments and the duration of treatments required, as well as the risk of recurrent disease, are uncertain as yet.

Summary for the Clinician

- **Thermal laser and PDT have been proven to decrease visual loss in a subset of patients with exudative ARMD. Current phase III trials with biologicals claim promising results, and even an improvement in vision**

5.2.2 Surgery

5.2.2.1 Membrane Removal

In some young patients with a submacular choroidal membrane secondary to, the presumed histoplasmosis syndrome, subfoveal choroidal neovascularization grows through a focal extrafoveal break in Bruch's membrane. In such patients, surgical removal of the membrane may spare the subfoveal RPE and may result in a preserved foveal function [35]. In ARMD patients, however, the neovascular tissue growth is under the RPE, as well as under the retina. Therefore, simple surgical removal of neovascular membranes in patients with ARMD almost invariably leads to damage of the subfoveal RPE, as well as the Bruch's mem-brane/choriocapillaris complex, and does not restore visual function [23, 36, 37]. Spontaneous RPE cell repopulation of the damaged area is ineffective or too late, if present at all [31]. Moreover, in 40 % of patients recurrent membranes were detected within 2 years after membrane removal. Despite these undesired effects, the resulting scotoma may be less disturbing than an untreated progressive exudative choroidal membrane. Therefore, simple membrane removal is currently being studied in a controlled manner in the USA in the Submacular Surgery Trial (SST).

Summary for the Clinician

- **Simple membrane removal damages the subfoveal RPE layer, which limits the potential to preserve foveal function. Nevertheless, a controlled trial is underway in the USA (Submacular Surgery Trial)**

5.2.3 Membrane Removal with the Reconstitution of the Underlying RPE

The spectacular functional restoration achieved in some patients with exudative age-related macular detachment after macular rotation has proved the potential for creating a fresh undersurface of functioning RPE cells [15]. However, a tilted image in successful cases, complex and time-consuming surgery and a high percentage of vision threatening complications because of proliferative vitreoretinopathy have remained drawbacks of this technique.

Other cornerstones in the concept of restoring the RPE underlayer of the macula are:

- Functioning RPE cells were shown to be essential for the preservation of Bruch's membrane and the survival of the choriocapillary in rabbits [22].
- Blaauwgeers et al. [9] showed that human RPE cells secreted VEGF on their basal side and that the facing choriocapillary had VEGF receptors.
- Subretinal RPE injection was capable of postponing photoreceptor death in RCS rats [26].

Table 5.1. Overview of studies using a cell suspension or cell sheets, allograft or autograft, and RPE or IPE

Type of graft	Publication	No. of patients
Allograft of a cell suspension	Valtink et al. [40]	20
Allograft of an RPE patch	Peyman et al. [30], Algvere et al. [3], del Priore et al. [14]	1, 8, 12
Autograft of a cell suspension	Binder et al. [8], van Meurs et al. [44]	60, 8
Autograft of an RPE patch	Peyman et al. [30], Stanga et al. [32], van Meurs et al. [43], Holz et al. [19]	1, 8, 18, 2
Autograft of a cell suspension	Thumann et al. [38], Lappas et al. [24]	8, 12
Autograft of an IPE patch	Navea [28]	5

Table 5.2. Advantages and drawbacks of the different RPE transplantation approaches

Type of graft	Advantages	Disadvantages
Cell suspension	The ease and elegance of an injection requiring only a small retinotomy	1. The lack of demonstrable presence or function of the cells 2. If cells are to be rejuvenated or improved in culture first, sterility demands are crucial 3. The possibility of reflux into the vitreous cavity, possibly increasing the risk for proliferative vitreoretinopathy
Cell sheet	RPE cells adhere to a substratum and may be in their native differentiated monolayer	1. Difficult to find the right artificial underlayer that allows handling of the sheet, adherence of RPE and no interference with a possible RPE/choroid cross-talk 2. Introduction of the sheet and its correct positioning (RPE up; not rolled over) requires a larger retinectomy and is surgically challenging 3. If the sheet has been doctored in the laboratory, sterility demands are paramount
Allograft	Cadaver eyes or RPE cultures could be used. The supply of donor tissue would be more abundant	Although the anterior chamber has an immune privilege and the same may hold true for the subretinal space, immune rejection was thought to play a role in the lack of function and reaction around the graft in the studies by Alvere, Del Priore and Engelmann
Autograft	No immune reaction because of non-self, although a tissue response in which the immune system is involved may be generated by the disease process and the subsequent surgery [27]	1. RPE cells have the same age and possibly the same pathology 2. Material is relatively restricted in amount
RPE or IPE	RPE is more likely than IPE to take over submacular RPE functions	IPE is easier to obtain through a iridectomy

Consequently, several different surgical approaches to recreating a functioning underlayer of the macula have been tried. We can subdivide these approaches into: autografts versus allografts, loose cells in suspension versus cell sheets or patches; and RPE versus iris pigment epithelium (IPE) cells (Table 5.1).

The advantages and disadvantages of the different transplant approaches are listed in Table 5.2.

5.2.3.1 Autograft Versus Isograft

Fibrosis with oedema and persistent dye leakage on fluorescein angiography was observed in patients with a fetal RPE patch [1–3], HLA-typed RPE cell suspension [41] or cadaver patch [14], which was thought to result from an immune rejection. Therefore, autologous tissue would be preferable. Immune involvement and inflammation may nevertheless occur because of the surgical trauma, as not only self- and non-self, but also damaged, tissue may trigger an immune response (the danger model) [27]. However, it makes sense to reduce both factors by using autologous tissue and trying to minimize surgical manipulation.

5.2.3.2 Iris Pigment Epithelium Versus RPE

Using iris pigment epithelium has the advantage of being a relatively easy way of harvesting by performing a surgical peripheral iridectomy. Iris pigment epithelium, however, may not have all the functions required of RPE.

5.2.3.3 Cell Suspension Versus a Cell Sheet

A considerable metamorphosis is required of transplanted RPE cells in suspension to reconstitute an RPE layer in patients after choroidal membrane extraction. After being scraped off their native Bruch's membrane or culture substratum, the cells are expected to adhere to a damaged Bruch's membrane, to survive and redifferentiate into a functional monolayer. In vitro studies show RPE cells adhere poorly to damaged Bruch's membrane [13, 33, 34, 39, 46, 47]; RPE cells from patients with exudative ARMD, moreover, may even have less ability to proliferate than RPE cells from patients without ARMD [45].

RPE cells on some substrata, on the contrary, are already adherent and differentiated; the delivery of a sheet is more problematic, however, than a cell suspension through a small-bore cannula.

Summary for the Clinician

- **RPE cell reconstitution is necessary to maintain macular function. At present, autologous RPE and IPE cell suspensions fail to do so; autologous sheets of RPE cells may show more sustained function than homologous ones**

5.3 Translocation of a Full-Thickness Patch from the Midperiphery

5.3.1 Rationale

With the current lack of a demonstrable presence or function of autologous RPE suspension transplants in patients, we decided to pursue the use of a sheet of autologous RPE on its own substratum. Peyman reported a patient on whom a full-thickness flap with a pedicle was used. The follow-up was 6 months and stabilization of a vision of 20/400 was reported [30]. Aylward, in eight patients, used a full-thickness patch cut out from a location adjacent to the removed subfoveal membrane. In four patients some function on microperimetry could be shown over the patch; preoperative vision was too low to assess postoperative vision properly [32]. Fibrosis of the patch, however, developed in the 2nd year of follow-up in most patients (verbal communication, May 2003).

In Aylward's patients the grafted paramacular choriocapillary appeared sclerotic and damaged by the surgery and we speculated that it was therefore less likely to be successfully revascularized. We thought we could improve on Aylward's technique by harvesting a relatively

healthy midperipheral full-thickness RPE and choroid patch with the advantage of easy accessibility to cut out the patch and a direct control of bleeding from the donor site.

Summary for the Clinician

- **Our method of choice is to translocate a midperipheral full-thickness autologous RPE and choroid graft to the macula after membrane removal**

5.3.2 Patients and Methods

5.3.2.1 Inclusion

Patients with a subfoveal choroidal neovascular membrane that was more than 50% occult on fluorescein angiography (FAG) and larger than one disk diameter, with or without submacular blood, were eligible for RPE translocation. This study was approved by the Institutional Review Board of the Rotterdam Eye Hospital and written informed consent was obtained from all patients, in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki. The present report concerns those patients who have had a follow-up of 12 months or longer.

Preoperative examination included general and ophthalmologic history taking, and an ophthalmologic examination, including best corrected ETDRS vision, dilated funduscopy and fluorescein angiography or indocyanine green angiography.

Postoperative visits were scheduled at 1, 3 and 6 weeks, and at 3, 6, 9, 12, 18 and 14 months. The censoring date was 1 October 2003. During each visit best corrected ETDRS vision testing and a comprehensive examination were performed. At 6 and 12 months, fundus pictures were taken and preferred fixation on the fixation light of the optical coherence tomograph (OCT) was monitored on the OCT fixation screen. Patients with a follow-up of 6 months or longer were tested (some twice) with a confocal scanning laser ophthalmoscope (HRA, Heidelberg Retina Angiograph, Engineering GmbH, Dossenheim, Germany) for autofluorescence (AF). An argon blue laser (488 nm) was used for excitation; emitted light was detected above 500 nm (barrier filter). To amplify the autofluorescence signal, several images were aligned, and a mean image could be calculated after detection and correction of eye movements by using image analysis software [17, 18].

In selected patients we performed fundus perimetry with the Nidek MP-2. In selected patients fluorescein or indocyanine green angiography was performed to exclude the regrowth of a neovascular choroidal membrane.

Summary for the Clinician

- **The results of 18 patients are reported with predominantly occult subfoveal membranes with a disk diameter (DD) of 1–3 and submacular blood; the follow-up was 1–2 years**
- **Surgical improvements, however, are based on the experience of all patients treated so far (n=38)**
- **Functional outcome was measured with ETDRS vision testing. In selected patients fluorescein and indocyanine green angiography, fundus autofluorescence and OCT were performed**

5.3.2.2 Surgery

After the induction of a posterior vitreous detachment, a complete vitrectomy was performed. The choroidal membrane was removed through a paramacular retinotomy from the subretinal space with Thomas subretinal forceps (Fig. 5.1). After circular heavy diathermia in the midperiphery at the 12 o'clock position and removal of the retina within the diathermia marks, we used vitreous scissors to cut a full-thickness patch of RPE/choroid of approximately 1.5×2 mm (Fig. 5.2). We then loaded the cut-out patch on an aspirating spatula (Fig. 5.3) and repositioned the patch under the macula through the existing paramacular retinotomy (Fig. 5.4). We surrounded the midperipheral retinotomy site with laser coagulation and left a silicone oil tamponade. In a second procedure, approximately 3 months later, we removed the silicone oil, performed a lensectomy and inserted an intraocular lens (IOL).

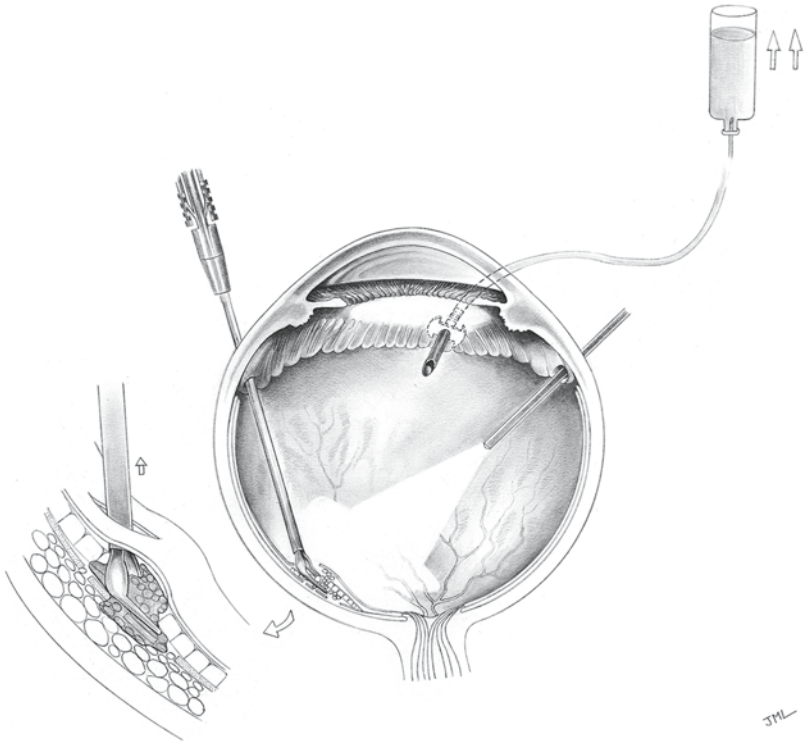

Fig. 5.1. Removal of the choroidal membrane with subretinal forceps

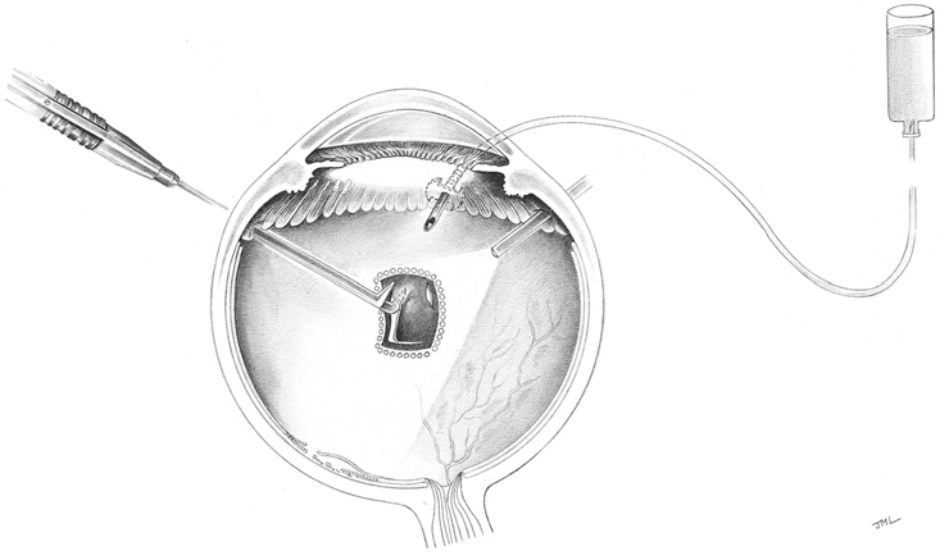

Fig. 5.2. A full-thickness patch of RPE, choriocapillary and choroid is cut out after removal of the overlying retina

Fig. 5.3. Loading of the RPE graft on an aspirating spatula

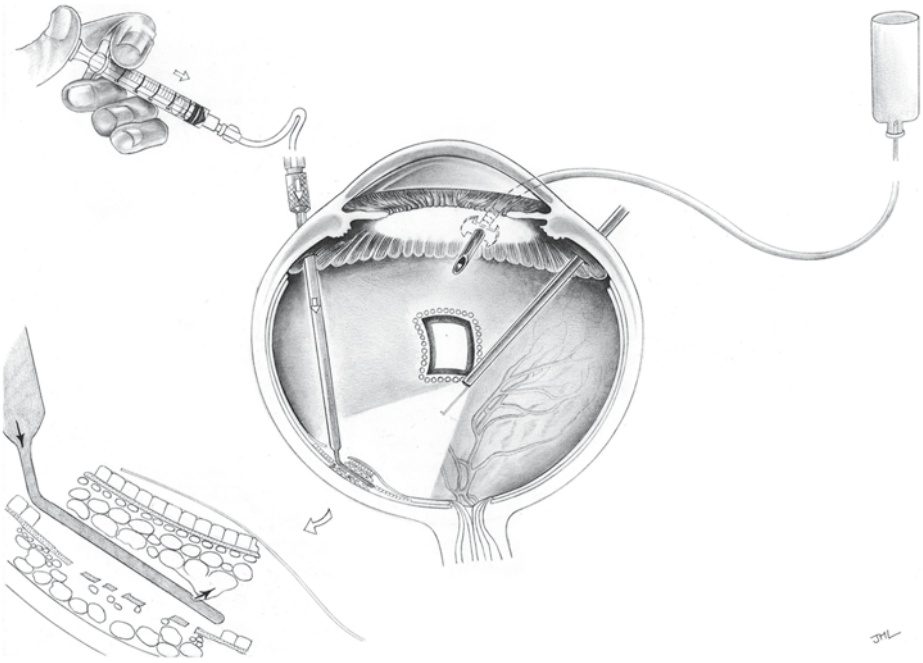

Fig. 5.4. Insertion and release of the graft under the fovea

Table 5.3. Follow-up results of patients

Patient	m/f	Preoperative vision	Postoperative vision			Remarks
			1 year	1.5 years	2 years	
1S	f	20/400	20/80	20/125	20/100	
2K	f	20/200	20/63	20/40	20/40	
3vL	f	20/200	20/200	20/400	20/400	
4vS	f	CF	20/200	20/200	20/200	
5vdP	m	20/200	10/160	20/125	20/160	
6V	m	20/200	20/80	20/80		
7B	m	20/400	CF	CF		Recurrence
8S	m	20/160	20/160			Deceased
9B	f	20/200	20/40	20/50		
10T	m	20/200	20/160	20/400		Recurrence
11T	m	20/160	CF	CF		Recurrence
12K	f	20/160	20/100	20/80		
13A	f	20/160	20/200			
14K	f	20/200	20/200			
15F	f	20/400	20/160			
16dK	f	20/200	20/160			PVR
17dV	f	20/160	20/40			
18V	m	20/160	20/125			PVR

5.3.3 Results

Thirty-seven patients have been included in the study from 12 October 2001, but only the first 18 have been followed up for more than 12 months. The present report covers the results of these 18 patients; surgical techniques, however, have evolved during the entire study period and notes on surgical technique include surgical experience up to the present time.

The preoperative duration of visual loss in the operated eye ranged from 2 weeks to 4 months. Visual acuity ranged from 20/400 to 20/160. End-stage macular degeneration was present in the fellow eye in 12 patients (Table 5.3).

On fluorescein angiography, 17 patients had a mixed or occult subfoveal neovascularization; in one patient (patient 1) no angiogram was performed because of a thick submacular haemorrhage. On angiography the size of the neovascular membranes varied from one to three disk diameters. Subretinal blood was present in 14 patients, extending to the vascular arcade in 4. Eight patients used aspirin and did not stop its use prior to the surgery. Five patients discontinued their use of coumarin anticoagulants 1 week before surgery.

5.3.3.1 Peroperative Course

Despite their advanced age, in only two patients did we not have to actively induce a posterior hyaloid detachment. To prevent bleeding when removing the choroidal membrane we raised the intraocular pressure to 120–140 mmHg, and slowly decreased the pressure afterwards. At the first sign of bleeding we raised the bottle again (as advised by Matthew Thomas, MD). Subretinal blood was best flushed away with a subretinal cannula.

The area of damaged RPE resulting from membrane and haemorrhage removal was approximately three to five disk diameters in each

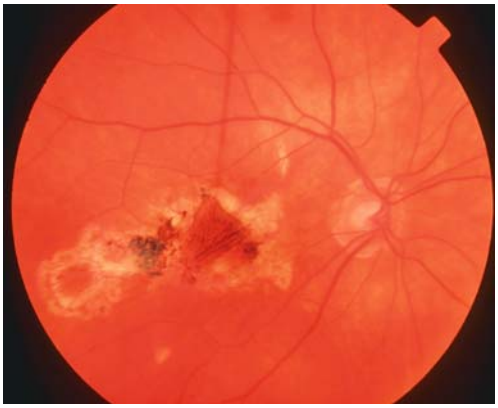

Fig. 5.5. Patient 1, two years postoperatively, vision 20/80, fixation over the patch. The velvety RPE patch is smaller than the area denuded of RPE by the surgical removal of the choroidal membrane

patient and included the area under the fovea in each patient. The RPE patch was smaller than the damaged RPE/Bruchs membrane/choriocapillary area in all patients (Fig. 5.5).

Instruments

Instruments were designed and manufactured in close collaboration with Ger Vijfvinkel, of the Dutch Ophthalmic Research Center (DORC), Zuidland, the Netherlands.

5.3.3.2 Finding a Cleavage Plane Between Sclera and Choroid

When we try to remove the patch, remnants of connecting tissue between choroid and sclera may jeopardize a clean release. Before cutting out the patch, we now separate the patch from the sclera by introducing and sweeping a long spatula under the patch.

Preparation of the Graft

Once all four sides of the rectangular 1.5–2×2–3 mm graft and the collagenous connection of the choroid to the sclera have been cut with scissors, the graft has the tendency to roll up into a half cylinder with the RPE on the convex side, usually with the half cylinder limbus parallel. This occurs in balanced salt solution (BSS) and the free floating patch may be subsequently difficult to position on the spatula. Moreover, the infusion bottle should be really low to minimize turbulence and to prevent the patch from disappearing through a sclerotomy when changing instruments. A great help is the use of extra ceiling illumination to be able to work with two hands, for example, so that the patch can be held when changing instruments.

Preparation of the patch under perfluorocarbon (PFCL) has proven to be best for visualization and keeping the patch flat on the spatula, while it allows us to keep the bottle raised, thereby decreasing the risk of bleeding from choroidal or retinal vessels. Most recently, however, we have stopped "working under PFCL", because we could not exclude that a thin film of PFCL would remain adherent to the RPE and might interfere with subsequent RPE-photoreceptor interaction.

Positioning of the Graft Under the Fovea

To allow the insertion of the patch through the retinotomy, however, one has to aspirate the PFCL. It helps to lift the foveal edge of the retinotomy with the PFCL-aspirating cannula to allow an easy insertion. Positioning of the graft under the fovea with a common spatula is difficult. Horizontal forceps, even when designed not to close entirely, proved to be unsuitable because the patch could not be released since it remained adherent to the forceps. The best instrument to hold and release the patch turned out to be a cannulated spatula with one opening, with an assistant applying aspiration to hold the patch or reflux to release the patch. A single opening appeared better than more openings; since once occlusion is lost over one opening, release can no longer be effected by refluxing.

When releasing the patch by refluxing the spatula (at present manually by the assisting person, but foot control would more ideal), we simultaneously cover the macula with PFCL, to decrease the chance that the patch will move out again on withdrawal of the spatula.

Tamponade

Silicone oil has been used to enable a better examination of the patients in the early postoperative period. A gas tamponade would be possible too.

Summary for the Clinician

- Do not forget to induce a posterior vitreous detachment (PVD)
- PVD is rarely present in patients with exudative ARMD
- A very high bottle and patience is needed to prevent bleeding from the choroid after membrane extraction
- After PFCL removal it helps to lift an edge of the retinotomy to make the insertion of the patch easier
- Reinjection of some PFCL over the macula aids a proper release of the patch from the cannulated spatula

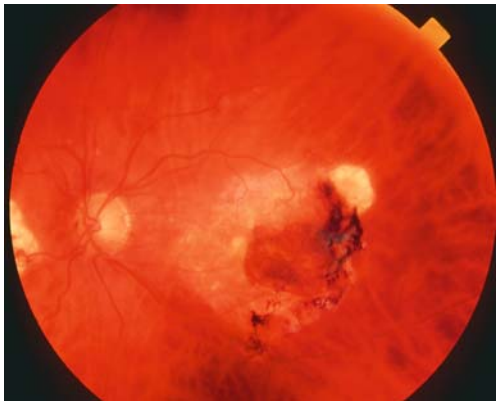

Fig. 5.6. Patient 2: 1.5 years after surgery, vision 20/64

5.3.3.3 Postoperative Course

Follow-up was 12–24 months (Table 5.3). The patch was biomicroscopically flat in 16 patients and had a brown, furry appearance in 14 (Fig. 5.5). In patients 3 and 4 part of the patch appeared to be folded double. Only patient 5 showed a fine fibrotic line over the patch.

Vision at the last recorded visit ranged from counting fingers to 20/40 (Table 5.3). A two-line or greater improvement in ETDRS visual acuity occurred in eight patients. Vision decreased two lines or more in three patients, all three with recurrent choroidal neovascularization. Vision remained within two lines in seven patients, including two with proliferative vitreoretinopathy (PVR) (Table 5.3).

Preferred fixation on the OCT monitor was over the patch in 12 patients, as well as fixation on a fixation rod (Fig. 5.5). OCT images were not easy to read; the retina could be better evaluated than the RPE and choroid. The retina over the patch remained thicker than normal in most patients; there appeared to be a correlation between a thinner retina and a better function.

Confocal scanning laser ophthalmoscopy (SLO) showed almost normal autofluorescence over the patch in six out of seven tested patients up to 2 years (patient 2, Figs. 5.6, 5.7) and 1.5 years (patient 5), respectively, postoperatively. In the two patients with a partly folded patch, autofluorescence was less, but not absent.

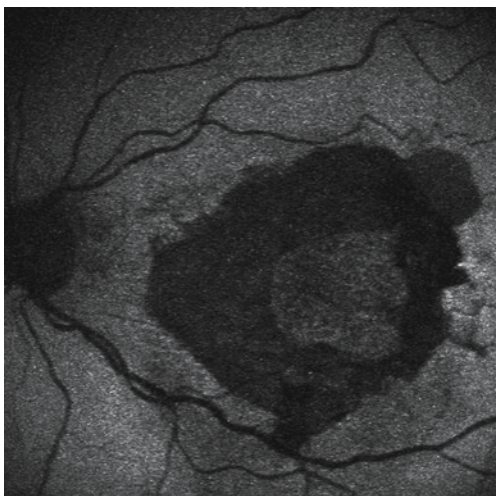

Fig. 5.7. Patient 2: almost normal fundus autofluorescence 2.0 years after surgery

Indocyanine green angiography showed perfused choroid in or under the patch in eight of nine examined patients (from 6–16 months postoperatively) (Fig. 5.8 a, b). Fluorescein angiography was performed postoperatively in six patients, revealing background filling in the patch comparable to the rest of the fundus, suggesting the presence of perfused choroid and choriocapillary in or under the graft.

In three patients recurrent or persistent choroidal neovascular membranes were detected; despite laser treatment and closure of the

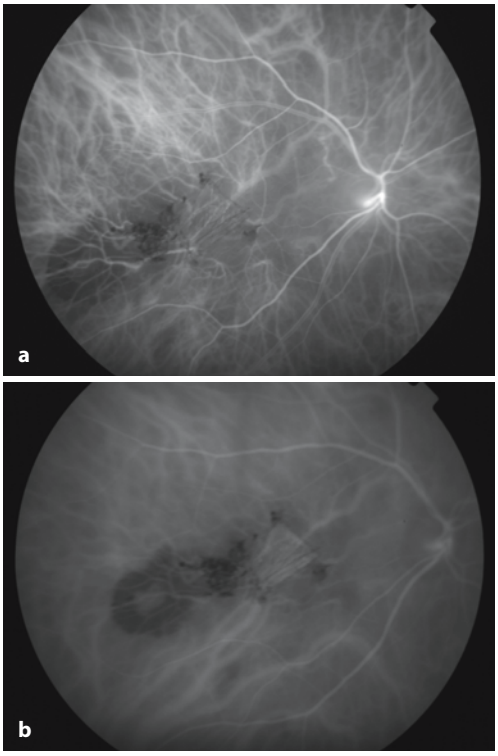

Fig. 5.8 a,b. Patient 1: early (**a**) and late (**b**) phase ICG angiography demonstrated perfusion of the choroid under the RPE patch

membranes, vision dropped to finger counting in all three patients.

Retina detachment due to PVR developed in patients 16 and 18. The retina over the RPE patch remained adherent, however. Revitrectomy, membrane peeling and silicone oil tamponade were performed.

Silicone oil was removed in all 19 patients, typically 3–4 months after the first procedure.

When removing silicone oil some degree of retinal puckering/cellophane maculopathy was present in 12 patients. We removed the internal limiting membrane (ILM) in these patients, with the help of indocyanine green staining.

Summary for the Clinician

- **ETDRS vision improved 2 lines in over 25 % of patients**
- **Angiography shows reperfusion in or under the patch in 9 of 10 patients**
- **Fundus autofluorescence was present in 7 of 8 patients**
- **Fixation on the OCT monitor was present in 13 of 18 patients**

5.3.3.4 Comment

For the following indications – a subretinal haemorrhage extending to the equator (three patients) where we would not have been able to remove the clot through a small parafoveal retinotomy; and an end-stage glaucoma (one patient) where we did not wish to increase the intraocular pressure during surgery – we have used another technique, derived from macular rotation surgery and the technique used for the transvitreal removal of choroidal melanoma (Kirchhof, verbal communication).

A temporal retinal detachment was created by the subretinal infusion of BSS through a 41-gauge needle, followed by a retinotomy at the ora serrata in the temporal 13 clock hours with subsequent folding over of the retina over the disk to expose the temporal subretinal space. While working under PFCL (membrane removal and preparation of the patch as well as the repositioning of the graft in the macular area), a better control of choroidal bleeding from the site of membrane removal as well as obtaining a correctly sized patch was possible. For some surgeons, this approach may be more controlled and standardized than the paramacular technique. Specific care, however, had to be taken to prevent the oversized temporal retina from being incarcerated in the nasal retinotomy. An advantage was certainly that it was no longer necessary to create a paramacular retinotomy, which was likely to be an important factor in the development of the frequent macular puckering in the patients with the paramacular technique. Drawbacks, however, were the longer surgery time and the at least theoretically increased risk for proliferative vitreoretinopathy. Moreover, due to the short follow-up of the four

patients treated with the flap-over approach, visual results are as yet uncertain.

In our study, one-fourth of patients reached a visual acuity of 20/80 or better after a follow-up of 1 year or longer, a level of visual acuity not to be expected in such patients. We were unable to identify patient characteristics that would predict a better outcome, because our series was a pilot study with an evolving surgical technique and numerous confounding factors besides patient selection. Because the RPE patch appeared to be revascularized, viable fixation and function on the fundus perimetry were over the patch in the majority of the patients and there was a sustained two-line improvement in several patients with a follow-up of almost up to 2 years, our approach may be good way to proceed.

Whereas laser treatment and pharmacological treatment have been studied or are being studied in prospective controlled trials, all surgical approaches discussed in this chapter (certainly including the discussed patch technique) have been uncontrolled single-centre pilot studies, without robust outcome measurements and varying follow-up. Therefore, data on visual results are not easily comparable to the data from controlled studies.

Fortunately, simple membrane extraction is currently under study in a multicentre, controlled study in the USA (the Submacular Surgery Trial). The MARAN study, however, which is a multicentre study in Europe on macular rotation, has experienced difficulty in recruiting patients.

Nevertheless, the above-described surgical method combines several desirable objectives: functioning, differentiated RPE cells on their native substrate were transplanted with relatively simple technology in a one-step 1-h surgical procedure, which was applicable to patients with a wide range of membranes (occult, very large), with or without subretinal blood and widespread RPE disease. Although this surgery may only be an intermediate stage before more sophisticated upgraded cultivated RPE cells on a suitable artificial substratum are available, its concept and the surgical technique required may be useful in the future. If surgery is to hold any place at all beside the use of newer pharmacological biologicals, the patch technique may remain of interest.

Summary for the Clinician

- **We report a surgical pilot study, in patients with subfoveal membranes of 1–3 DD, most with blood. There was no control group**
- **The results are promising, because 25 % of patients have a vision of 20/80 or better after 1 year**
- **The surgical technique discussed above was evolving; a technique based on macular rotation may be preferable in selected patients or may be preferred by other surgeons**
- **At present, an autologous full-thickness graft of RPE and choroid best combines the ideal characteristics of a monolayer of differentiated RPE on a suitable substratum**
- **Future developments for RPE reconstitution include using an artificial membrane as substratum to be repopulated by RPE improved in culture by gene transfer or stem cells**

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Giant Retinal Tear

Martin Snead

Core Messages

- Only a minority of giant retinal tears occur secondary to trauma. Retinal detachment due to blunt trauma is more commonly associated with a disinsertion at the ora (dialysis) rather than true giant tear
- Giant tears are frequently associated with abnormal gel and inherited vitreo-retinopathies and the posterior hyaloid membrane is separated anteriorly to the ora allowing the posterior flap to develop independent mobility
- An inquiry should be made into any associated history of mid-line cleft, hearing loss, arthropathy or family history of retinal detachment
- Stickler syndrome patients have a high risk of retinal detachment and prophylactic 360-degree contiguous retinopexy reduces the risk of retinal detachment secondary to giant retinal tear from 70% to 7%

6.1 Introduction

The term "giant retinal tear" (GRT) should be restricted to very large tears at the ora serrata associated with an abnormally anterior separation of the posterior hyaloid membrane. Under the influence of gravity and freed from the mechanical restraints of a more limited gel separation, they are often characterised by an independent mobility of their posterior flap and occasionally if this is excessive, by radial posterior extensions at their apices.

Although popular in public perception only a minority of giant retinal tears occur secondary to trauma and even with successful repair in these circumstances the visual prognosis may be limited by associated collateral ocular damage [1]. Retinal perforation or incarceration from penetrating trauma rarely causes acute rhegmatogenous retinal detachment, but the associated corneoscleral wound provides access for extrinsic fibroblasts so that the more common sequel is a pathologically anterior separation of the posterior hyloid membrane (PHM) and late giant tear complicated by combined tractional and rhegmatogenous components [1].

Retinal detachment due to blunt trauma is more commonly associated with a disinsertion at the ora serrata rather than true giant tear. The sudden anteroposterior compression is associated with a corresponding coronal expansion and retinal avulsion injury characterised by an accompanying festoon of non-pigmented pars plana epithelium. There is a greater preponderance for superior quadrant involvement in contrast to the usual lower temporal quadrant involvement in non-traumatic dialysis [3, 6]. Although the disinsertion may exceed 90 degrees of the circumference and superficially resemble a giant retinal tear, the vitreous gel characteristically remains attached to the posterior flap so that independent mobility is not a feature (Fig. 6.1a). They respond well to conventional scleral buckling techniques. Subretinal fluid recruitment is slow so that unless the ora serrata is routinely inspected after blunt trau-

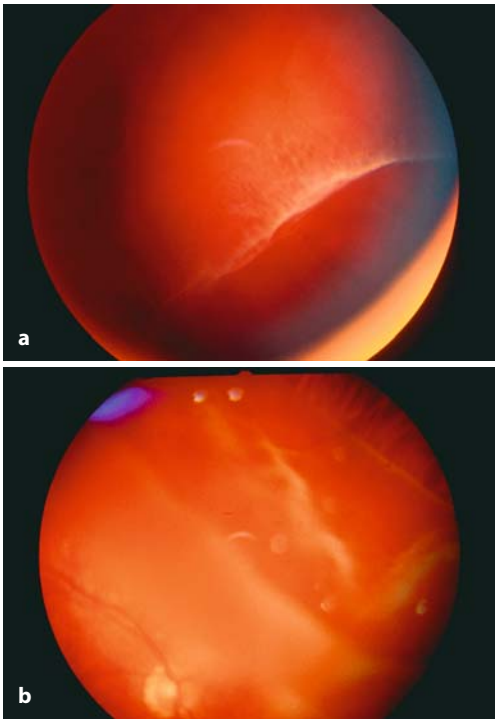

Fig. 6.1 a, b. Differential diagnosis. **a** Retinal dialysis. The posterior hyaloid membrane is attached. Note the characteristic bridges spanning the retraction and the cystic change in the border of the dialysis which is continuous with the frill at the ora serrata. **b** Giant retinal tear. The posterior hyaloid membrane is separated anteriorly to the ora allowing the posterior flap to develop in independent mobility

ma, the diagnosis may be delayed by several weeks until macular involvement ensues [6]. Further features distinguishing dialysis from giant retinal tear are the absence of radial extensions, which may occur at the apices of giant tears, and the normal compact healthy vitreous gel architecture in patients with retinal dialysis. Giant tears are frequently associated with abnormal gel and inherited vitreoretinopathies (see below) and the posterior hyaloid membrane is separated anteriorly to the ora allowing the posterior flap to develop independent mobility (Fig. 6.1b). Adults with giant tears tend to present acutely with visual loss, but young children frequently present late either as a result of second eye involvement or having been identified through screening or incidental examination. In these tragic instances, the visual prognosis is frequently poor even with successful repair because of the associated advanced PVR.

6.2 Genetics of Giant Retinal Tear

Stickler syndrome is the commonest inherited cause of giant retinal tear [23]. It forms part of the spectrum of type II/XI collagenopathies, which also includes the more severe Kniest dysplasia (MIM 156550), spondyloepiphyseal dysplasia congenita (SEDC, MIM183900) and spondyloepimetaphyseal dysplasia (Strudwick type, MIM 184250) [4] and all have a similarly high risk of GRT formation [23] (Table 6.1).

In contrast to the more severe disproportionate stature syndromes that result mainly from dominant negative mutations, the majority of patients with Stickler syndrome have premature termination mutations in the gene for type II collagen. This results in haploinsufficiency and they exhibit a characteristic vitreoretinal phenotype [13, 15, 20] (Fig. 6.2a). Pedigrees with a different vitreous phenotype (Fig. 6.2b) have mutations in the gene COL11A1 encoding type XI collagen and are now known as type 2 Stickler (STL2) syndrome [9, 12, 14, 22]. Other families exhibit neither of these two vitreoretinal phenotypes or linkage to known loci so there is further locus heterogeneity to be resolved. Exon 2 of the COL2A1 gene is principally expressed in the eye and spliced out of cartilage so that mutations occurring in this exon result in a predominantly ocular form of Stickler syndrome [13]. It is important to remember to consider the diagnosis in:

1. Neonates with Pierre-Robin sequence or midline cleft
2. Infants with spondyloepiphyseal dysplasia associated with myopia or deafness
3. Patients with a family history of rhegmatogenous retinal detachment
4. Sporadic cases of retinal detachment associated with joint hypermobility, midline clefting, or deafness

Table 6.1. Summary of fibrillar collagenopathies associated with GRT

Disorder	MIM	Gene	Map locus	Phenotype
Stickler syndrome type1	STL1 #108300	COL2A1	12q13.11-q13.2	Membranous vitreous anomaly, skeletal and aural features
Stickler syndrome type2	STL2 #604841	COL11A1	1p21	Beaded vitreous anomaly, skeletal and aural features
Stickler syndrome type3	STL3 #184840	COL11A2	6p21.3	Arthropathy and deafness. No ocular involvement
Stickler syndrome type4	STL4	*Unknown*	*Unknown*	Arthropathy, deafness, midline cleft
"Ocular only" Stickler syndrome	STL1 #108300	COL2A1	12q13.11-q13.2	Membranous vitreous anomaly, minimal systemic involvement
Dominant RRD	Not assigned	COL2A1	12q13.11-q13.2	Disorganised vitreous gel, dominant RD, no systemic features
Kniest dysplasia	#156550	COL2A1	12q13.11-q13.2	Shortened trunk and limbs, congenital megalophthalmos, flattened nasal bridge, fingers long and knobbly, deafness
Spondyloepiphyseal dysplasia congenita (SEDC)	#183900	COL2A1	12q13.11-q13.2	Barrel-shaped chest, lumbar lordosis. Disproportionate limb shortening affecting mainly the proximal limbs with hands and feet appearing relatively normal. Myopia, retinal detachment and giant retinal tear. Deafness
Spondyloepimetaphyseal dysplasia (Strudwick type) (SEMD)	#184250	COL2A1	12q13.11-q13.2	Severe dwarfism, pectus carinatum and scoliosis. Cleft palate. Disproportionately short limbs and delayed epiphyseal maturation are present at birth

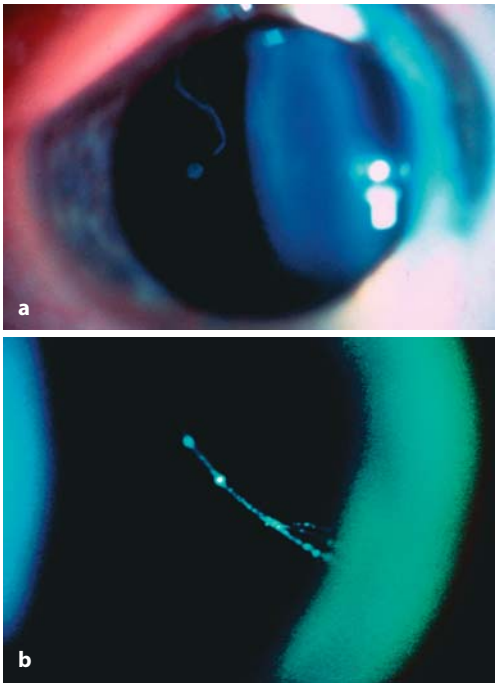

Fig. 6.2 a, b. Vitreous phenotypes. **a** Type 1 Stickler syndrome (membranous phenotype). **b** Type 2 Stickler syndrome (beaded phenotype)

Although well recognised, the association of giant tear with Marfan syndrome, a dominantly inherited disorder of fibrillin production with a prevalence of approximately one in 20,000 [11], is less common. The fibrillins are high molecular weight extracellular glycoproteins, and mutations in the fibrillin gene on chromosome 15 (FBN1) cause both Marfan syndrome and dominant ectopia lentis [2, 7, 8]. Mutations in a second fibrillin gene on chromosome 5 are responsible for congenital contractural arachnodactyly [25]. Recent work has confirmed fibrillin to be widespread in lens capsule, iris, ciliary body and sclera [26].

Approximately 75% of detachments in Marfan syndrome occur below 20 years of age [10]. Although there is a significant association with myopia, the myopia is characteristically developmental, in contrast to the congenital nonprogressive myopia more usually found in type 1 Stickler syndrome [17, 19, 21]. In Marfan syndrome the pupils characteristically dilate poorly because of associated smooth muscle structural iris abnormality [26] and when combined with lens subluxation and weak scleral architecture the repair of retinal detachment in Marfan syndrome patients can provide a formidable surgical challenge.

Summary for the Clinician

- **Stickler syndrome is the commonest inherited cause of giant retinal tear**
- **It is important to remember to consider the diagnosis of giant retinal tear in neonates with Pierre-Robin sequence or midline cleft, infants with spondyloepiphyseal dysplasia associated with myopia or deafness, patients with a family history of rhegmatogenous retinal detachment, and sporadic cases of retinal detachment associated with joint hypermobility, midline clefting, or deafness**

6.3 Preoperative Assessment

Preoperative assessment of the patient should include attention to systemic features in addition to the ocular examination. In particular, an enquiry should be made into any associated history of mid-line cleft, hearing loss, arthropathy or family history of retinal detachment, which may not be volunteered without a direct request for the information. Many patients will be myopic, but a significant minority are not and the term "cryptomyopia" has been suggested to encompass the patients without significant refractive error, but megalophthalmos associated with giant tear. Preoperative biometry of both eyes can be instructive and helpful, particularly if lensectomy is subsequently required.

Since the opportunity to assess the extent and associated features will be available at examination under anaesthesia, preoperative examination will focus particularly on related anterior segment abnormalities, the status of the vitreous (especially for the various congenital anomalies), the mobility and integrity of the posterior hyaloid membrane and the fundus and posterior hyaloid membrane examination of the fellow eye. If the PHM has separated with-

out complication, then prophylaxis will be unnecessary for the fellow eye. Alternatively, if the posterior hyaloid membrane remains attached then serious consideration to prophylaxis will be required.

6.3.1 Vitreous Examination

Vitreous examination should commence with examination of the gel quality, its structural architecture, the status of the posterior hyaloid membrane and any associated congenital anomaly. The examination is a useful first step in the differential diagnosis of the more common varieties of rhegmatogenous retinal detachment, including giant tear (Table 6.2). The gel phenotype is also a useful diagnostic indicator of any underlying genetic predisposition. The gel is assessed by slit-lamp specular biomicroscopy with a narrow slit beam illumination set at a wide angle from the axis of observation. The mobility and PHM status are assessed dynamically by asking the patient to look down and then straight ahead, which results in the gel being flung superiorly so that it can be observed as it slowly settles under gravity to its resting position. In patients with GRT the PHM separates very anteriorly and can usually be observed coming back to rest in the anterior third of the vitreous cavity. Occasionally, a condensing lens may be required if the membrane is shortened and settles more posteriorly, and this is also useful for inspecting the integrity of the PHM and the size of any associated defects which may influence the likelihood of secondary macular pucker.

6.3.2 Retinal Examination

Retinal examination includes both slit-lamp and indirect ophthalmoscopic assessment. Important factors which might influence the surgical approach include the extent and (meridional) position of the tear, the presence of associated radial extensions, retinal immobility and PVR, macular involvement and possible secondary macular hole. In younger patients, much of this assessment will be made with the patient under general anaesthesia and a portable hand held slit lamp should be available in theatre.

Table 6.2. The "Cambridge Guide" to the features associated with the seven most common varieties of primary retinal break in rhegmatogenous retinal detachment (*PHM* posterior hyaloid membrane, *Macular hole* refers to retinal detachment secondary to macular hole as distinct from isolated "idiopathic" macular hole)

Break type	PHM status	Vitreous architecture	Sex	Typical age group (years)	Refractive error	Fellow eye involvement/ pathology
Atraumatic dialysis	On	Normal	M>F	8–20	Emmetropia/ hypermetropia	5–15%
Giant retinal tear	Off	Congenital anomaly	M=F	5–50	Moderate/ high myopia	Variable up to 80%
Horseshoe tear	Off	Usually syneretic	M=F	45–65	Moderate/ high myopia	10%
Round retinal hole	On	Usually normal	F>M	20–40	Moderate myopia	55%
Macular hole	Off	Syneretic	M=F	45–65	High myopia	Unusual
Reticular schisis	On	Normal	M>F	70+	Hypermetropia	80%
X-linked retinoschisis	On	Normal, may have haemorrhage	M	10–20	Emmetropia	100%

6.4 Surgical Preparation

6.4.1 Examination Under Anaesthesia (EUA)

Surgery may be carried out with the patient under general or local anaesthesia. An anaesthetist should be present throughout and the author's preference is for general anaesthesia to facilitate a careful examination of the fellow eye when considering prophylaxis (see below). The initial examination also allows an assessment of the extent of the tear, the mobility of the posterior flap and the degree of any associated scleromalacia. It is especially important to carry out a 360-degree examination of the retinal periphery to locate any associated satellite horseshoe tears which are common and may be small and otherwise easily overlooked. Recurrent detachment and a failed primary repair will result if these are not identified and treated at the primary repair. The EUA also provides an opportunity to plan the surgical approach and in particular the position of entry sites for the deployment of instruments. As with any complex surgical procedure, careful planning and anticipation (and avoidance) of likely difficulties or variations from the norm will simplify successful completion of the surgical goal. Careful choice of entry sites will improve access and thereby make subsequent manoeuvres easier, avoiding the need for unnecessary lensectomy and reducing the likelihood of associated complications. Unlike horseshoe tears, giant tears occur at the junction of the retina and pars plana so that instruments or infusion cannulas introduced during vitrectomy will enter *through* the tear rather than anterior to it. For this reason if an infusion is required, it is helpful for this to be sited away from the tear as this helps to avoid further instability of the tear during surgery caused by the infusion currents. In contrast, access to the entire extent of the GRT, including both apices, can be assisted by deploying instruments at the mid-point of the tear. In the majority of cases, scleral buckles are not required during giant tear repair and in some instances can be an impediment to access over the entire extent of the tear during later stages of the surgical procedure. However, if there is substantial PVR and retinal shortening or associated inferior radial extensions to the GRT, a scleral buckle may occasionally be required as an adjunct to assist in break closure. Application of a scleral buckle, adjustment of buckle height and scleral suturing are generally more controlled with a closed globe, prior to commencement of vitrectomy.

Summary for the Clinician

- **In patients with GRT the PHM separates very anteriorly and can usually be observed coming back to rest in the anterior third of the vitreous cavity**
- **Unlike horseshoe tears, giant tears occur at the junction of the retina and pars plana so that instruments or infusion cannulas introduced during vitrectomy will enter through the tear rather than anterior to it**
- **If there is substantial PVR and retinal shortening or associated inferior radial extensions to the GRT, a scleral buckle may occasionally be required as an adjunct to assist in break closure**

6.4.1.1 Key Points to Assess at EUA

1. Assess the extent of tear and decide how this might influence choice of the entry site positions
2. Assess the quality of sclera in all four quadrants with regard to scleromalacia and suitability for scleral buckle (if required)
3. Assess the need for adjunctive scleral buckle
4. Assess the number and position of satellite tears remote from GRT
5. Assess the most appropriate patient head position with regard to flap position and subsequent fluid/perfluorocarbon liquid (PFCL)/oil exchange
6. Assess retinal mobility and extent of associated PVR

6.4.2 Vitrectomy

In most instances, the gravitational influence on the posterior flap will mean that in the supine position the posterior edge of the tear will have collapsed towards the posterior pole of the eye. Giant tear rotating tables have been superseded by the use of perfluorcarbon liquids to unroll the posterior flap and maintain its stability during tamponade exchange. Alternatively, "venturi" manipulation of the intraocular currents during direct fluid/silicone exchange can unroll and stabilise the flap avoiding the need for PFCL exchange altogether. If PFCLs are to be used, then a complete vitrectomy is essential to facilitate their introduction and it is also important to remember to remove any gel which may have prolapsed into the subretinal space through the GRT. Provided the entry points have been chosen with care to facilitate access to the entire extent of the GRT it is rarely if ever necessary to carry out a lensectomy. The detached pars plana should be excised during this part of the procedure, which will assist during the later internal drainage and also reduces the risk of a persistent proliferative scaffold and subsequent tractional complications. In those cases complicated by PVR, attention should also be paid to removal of any surface membrane or distortion of the posterior flap to assist in smooth reattachment during PFCL exchange. If any associated shortening is not relieved, there is a significant risk of PFCL and/or silicone being forced into the subretinal space.

6.4.3 Management of the Posterior Flap

After vitrectomy completion and removal of any surface membrane, the repositioning of the posterior flap can be addressed. The advent of perfluorocarbon liquids has simplified this manoeuvre enormously. Because of the surface tension differences there is a greater likelihood of posterior slippage with PFCL/air/gas exchange and for this reason the author's preference is to carry out a direct PFCL/oil exchange

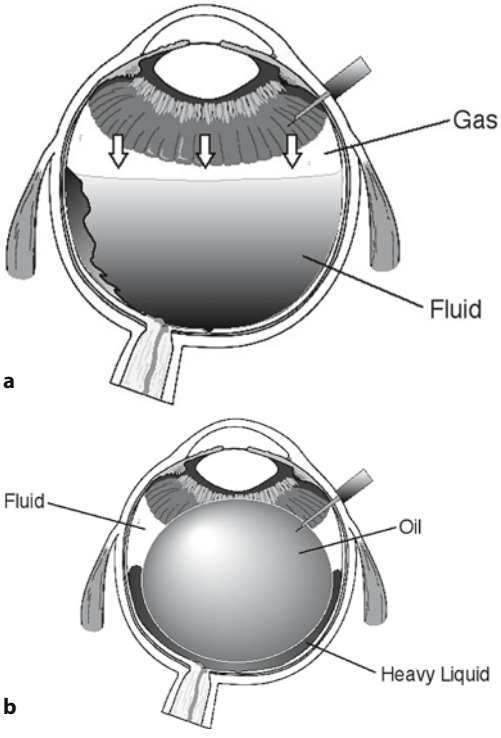

Fig. 6.3 a, b. Tamponade exchange. Fluid/gas/silicone exchange (**a**) is more likely to induce posterior slippage and is more time consuming than direct PFCL/oil exchange (**b**)

[27] (Fig. 6.3). This is also simpler and quicker since it avoids the interchange of air, gas and oil and it is important to remember that the PFCL is only being used as a peroperative tool to unfold the flap and maintain its position whilst the permanent tamponade is introduced. It is unnecessary and can be counterproductive to exchange the entire posterior segment for heavy liquid since this increases the likelihood of PFCL being forced into the subretinal space.

6.4.4 Retinopexy

Having successfully repositioned the giant tear, there can be a tendency to consider the major operative challenge to be complete. However, it should be remembered that application of retinopexy is the most important step in the en-

tire surgical programme, since this will result in success (or failure) of a permanent long-term adhesion. Several aspects to the retinopexy should be considered.

6.4.4.1 Timing

In most instances, retinopexy will be applied at the time of the primary repair. However, rarely the posterior flap can remain unstable and in these instances it can be beneficial to posture the patient postoperatively and apply retinopexy once the position of the posterior flap has stabilised. This is preferable to the uncertainty of where to position the retinopexy and the need for subsequent or repeated top-up treatment. In those exceptional cases where a scleral buckle has been necessary, then retinopexy may need to be applied to that part of the tear before the buckle is sutured into position.

6.4.4.2 Modality

Laser or monitored transscleral cryotherapy is generally employed. Monitored cryotherapy under indirect control is usually preferable because of the anterior position of GRT formation. It also provides a broader ribbon of treatment as a safeguard in the event of any minor postoperative slippage or retraction in the early postoperative phase and has the advantage over laser of "lighting" up small satellite tears which are so often found in association with the main pathology and easily overlooked. Conversely, in cases with posterior extensions or where a retinectomy has been necessary, endolaser or endocryotherapy provides much easier access.

6.4.4.3 Delivery

Whichever modality (or combination) is chosen, extreme care should be taken to ensure continuity of treatment without any gaps and particularly without re-treating any area already covered.

6.4.4.4 Prophylaxis for the Fellow Eye

In contrast to most other blinding retinal disorders, blindness through retinal detachment is in most cases potentially avoidable if a rationale for the prediction and prevention of retinal detachment could be developed. In the past this goal has been frustrated by a lack of understanding of the factors influencing retinal detachment even in high-risk groups.

Factors traditionally associated with retinal detachment include refractive error, a positive family history, visible lattice retinopathy and fellow eye involvement, but the nature of these associations is poorly understood. The prevalence of myopia varies enormously and even in Stickler syndrome up to 20 % of patients may exhibit no significant refractive error. Many patients with GRT exhibit none of the accepted risk features such as equatorial lattice retinopathy and in those that do GRT formation occurs anteriorly to this so that unsurprisingly retinopexy to areas of "visible" pathology is ineffective in preventing subsequent GRT (Fig. 6.4).

Unlike horseshoe tear detachments where the site of initiation can be very difficult to predict with accuracy, the surgeon has the advantage of a reasonably accurate idea of where to apply prophylaxis for GRT. The rationale for offering prophylaxis in such high-risk cases is to prevent progression of GRT to detachment by applying treatment at the postoral retina and the predicted site of giant tear initiation. The application of this retinopexy is every bit as demanding as repair of a GRT itself with accurate monitoring of every single application in a planned fashion without gaps and extreme care to avoid retreating any areas (Figs. 6.5, 6.6).

Both Type 1 and Type 2 Stickler syndrome patients have a high risk of retinal detachment and prophylactic 360-degree contiguous retinopexy reduces the risk of retinal detachment secondary to giant retinal tear from 70 % to less than 7 % [24].

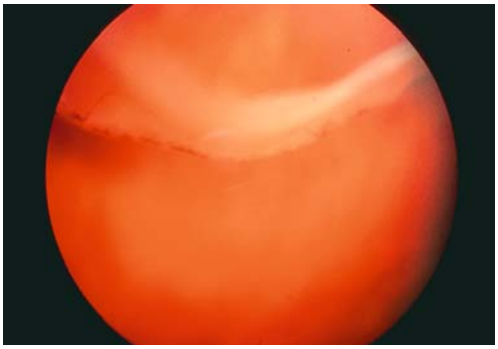

Fig. 6.4. Unsuccessful prophylaxis. Laser applied too posteriorly to prevent giant tear progression. Only eye, type 1 Stickler syndrome, fellow eye blind following detachment

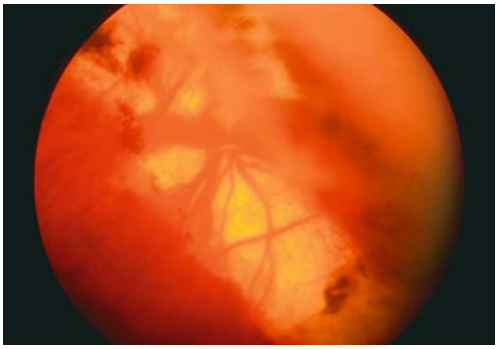

Fig. 6.6. Effective prophylaxis. This giant tear occurred 2 years after prophylaxis. Retinal detachment did not ensue

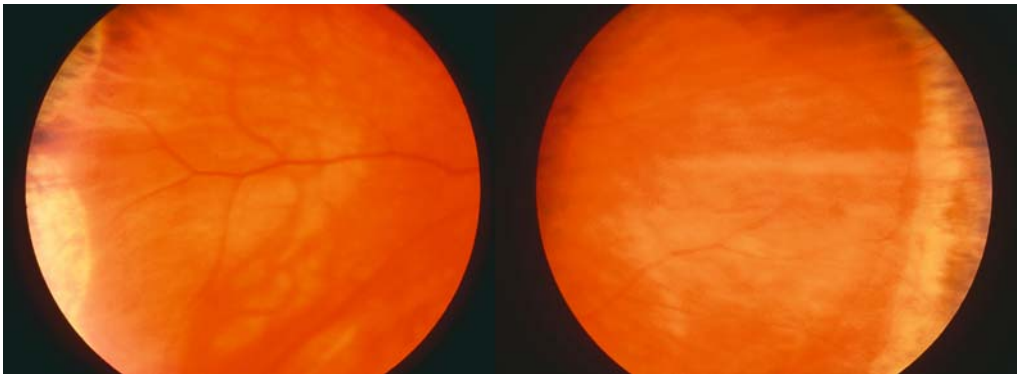

Fig. 6.5. Prophylactic 360-degree cryotherapy. Applied in contiguous "ribbon" to the postoral retina to prevent giant tear progression from pars plana

6.5 Postoperative Care

With extensive tears, the tendency for slippage of the posterior flap will persist during the early postoperative period and appropriate posturing of the patient to keep the tear stable is important whilst the retinopexy matures to full strength. Appropriate face down posturing will usually result in pronounced lid swelling during the early postoperative phase as the oedema settles under gravity to the lids and subcutaneous tissues. Systemic non-steroidal anti-inflammatory medication is highly effective for postoperative analgesia in addition to topical treatment with a steroid and mydriatic of choice.

6.6 Complications

6.6.1 Haemorrhage

Even in the absence of refractive myopia, many eyes with giant tear will have megalophthalmos with associated scleromalacia. Choroidal haemorrhage may occur due to fracture of vortex vessels traversing a thin scleral passage with little structural support, if the ocular pressure/volume relationships are not maintained at a constant level throughout the procedure. Patients with giant tear tend to have fragile scleral architecture and surgical manoeuvres should be gen-

tle and unhurried throughout the procedure. Although rarely necessary for giant tears, if a scleral buckle is required it should be applied with care to avoid inadvertent perforation, incarceration and haemorrhage.

6.6.2 Lens

The association between zonule abnormalities, cataract and giant tear is well recognised and not restricted purely to those conditions usually associated with ectopia. Even during otherwise routine surgery, silicone or gas may present into the anterior chamber, which may pose problems with fundus visualisation preoperatively or pressure problems postoperatively [5], and the surgeon should be prepared to anticipate this potential difficulty. All phakic patients with giant tear will develop subsequent accelerated cataract (typically at 15–18 months post giant tear) [18] and appreciation and anticipation of problems during cataract surgery will help to avoid an unnecessarily poor visual outcome to an otherwise successful retinal repair. When planning phacoemulsification for cataract following a previous giant tear repair the surgeon should take into account the following factors:

1. The large globe with thin sclera and a vitrectomised eye will be much more unstable than for routine cataract surgery. The anterior chamber depth may fluctuate considerably during surgery and this can be uncomfortable for a patient under local anaesthesia.
2. The possible associated zonule abnormalities compromising capsular bag support. If cataract surgery is being combined with oil removal, silicone may present to the anterior chamber if the pressure is not maintained during surgery. There may also be additional difficulties during phacoemulsification necessitating a capsular tension ring to stabilise the position of the posterior chamber implant.
3. Higher incidence of cystoid macular oedema. This can be reduced by pre- and postoperative cover with systemic non-steroidal anti-inflammatory cover.

6.6.3 Recurrence

The majority of adult patients will present acutely and the success rates with primary repair are excellent. Recurrent detachment is uncommon and when it occurs is usually either due to a new satellite tear or occasionally progressive shortening and reopening of a localised area of the original break. In either instance, the use of primary silicone tamponade will usually limit progression of the recurrent detachment, which can frequently be repaired with either a local explant or retinectomy prior to planned subsequent oil removal.

In those cases presenting with advanced PVR at presentation (typically children, adults with penetrating trauma or late presentation) the risk of redetachment is significantly higher following oil removal and the decision to proceed with oil removal will be balanced on an individual basis relating to the perceived stability of the retina, visual prognosis, the vision in the fellow eye and patient preference.

6.7 Follow-up and Two-Stage Surgery

Silicone removal can be planned as a second stage in the visual rehabilitation of the patient, once the retinopexy has matured to full strength and the retina is stable. Theoretically this could be as early as a month, but in practical terms some patients choose to defer further major surgical intervention a little longer. In many instances this has practical advantages as any pre-existing myopia is reduced by the silicone tamponade so that the patients can usually return to work in the interim and silicone removal can be timed to combine with cataract surgery, thereby avoiding a third subsequent surgical intervention.

Summary for the Clinician

- **There is likelihood of posterior slippage with PFCL/air/gas exchange. It is recommended to carry out a direct PFCL/oil exchange. Monitored cryotherapy under indirect control is usually preferable because of the anterior position of GRT formation. Retinopexy restricted purely to areas of visible pathology is ineffective in preventing subsequent GRT. The association between zonule abnormalities, cataract and giant tear is well recognised and not restricted purely to those conditions usually associated with ectopia. Even during otherwise routine surgery, silicone or gas may present into the anterior chamber, which may pose problems with fundus visualisation preoperatively or pressure problems postoperatively. Silicone removal can be planned as a second stage in the visual rehabilitation of the patient, once the retinopexy has matured to full strength and the retina is stable**

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Retinal Pigment Epithelium Differentiation and Dedifferentiation

7

Carl Sheridan, Paul Hiscott, Ian Grierson

Core Messages

- In their normal adult location, retinal pigment epithelial (RPE) cells are highly differentiated cells that undertake a variety of specific tasks crucial to the well-being of the photoreceptors. However, following retinal insults ranging from inflammation to ageing, these cells may switch from their differentiated phenotype and begin to look and act like macrophages or fibroblasts. This change, or dedifferentiation, is dependent on a variety of factors including growth factors and extracellular matrix with which the RPE cells are in contact. The challenge for the future is to find ways of reversing the process of RPE dedifferentiation, a challenge that is of particular relevance to RPE cell transplantation and to the management of retinal diseases such as PVR

7.1 Introduction

The retinal pigment epithelium (RPE) is a multifunctional cell that maintains the photoreceptors in optimal condition [7]. We have described the RPE cell in its crucial homeostatic role as being the nursemaid to the photoreceptors [22]. Protected and cosseted by the RPE cells, which provide a whole range of housekeeping roles that include storage of metabolites, nutrient transport, barrier functions, free radical defence and phagocytosis of spent outer segment materials, the photoreceptors can survive a lifetime [7, 22] (see Fig. 7.1).

The normal RPE forms a mosaic of static, hexagonal, polarized cells beneath the neural retina (Figs. 7.1, 7.2). A consequence of being highly differentiated is the need for RPE cells to maintain themselves outside the cell cycle. The precise nature of the cell cycle checks and controls that are necessary remains obscure but, unlike photoreceptors that are terminally differentiated, the RPE cells do retain the ability to divide. In the healthy eye, however, the turnover of RPE cells is remarkably low. On the other hand, in tissue culture conditions [10, 29] and pathologies such as PVR [30, 33] and PDR [34], the RPE cells undergo dedifferentiation and in these "adverse" environments they become migratory and very actively proliferating cells.

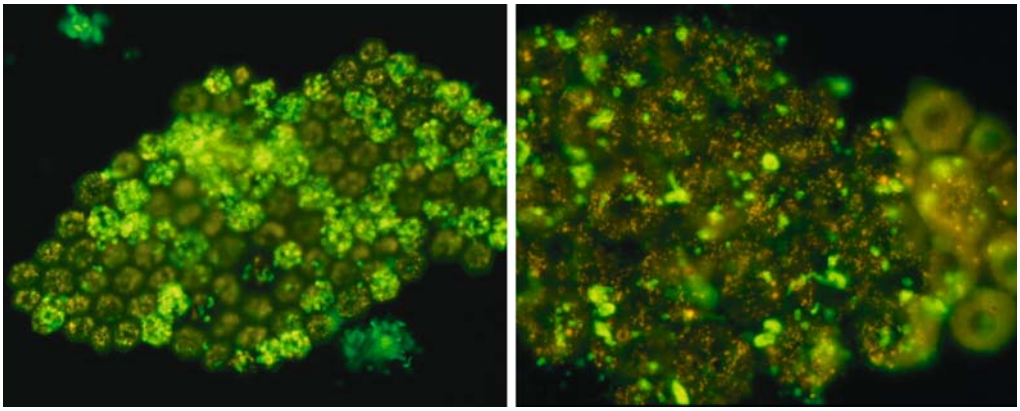

Fig. 7.1. Isolated clumps of RPE where phagocytosed rod outer segments fluoresce green

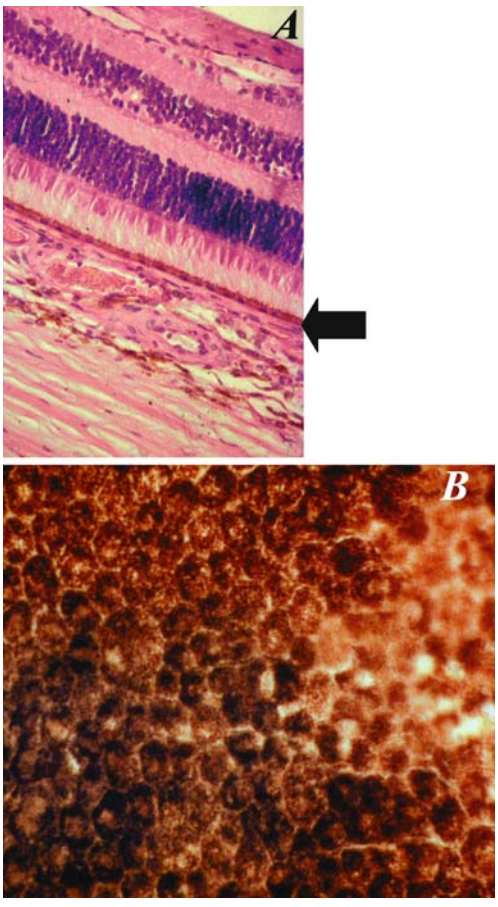

Fig. 7.2. **A** The normal RPE in histological sections is seen as a pigmented monolayer beneath the retinal photoreceptors. **B** In flat sections the RPE mosaic is appreciated

7.2 Dedifferentiated RPE Cells

The transition from differentiated to dedifferentiated cells has a dramatic functional and phenotypic effect on the RPE cells that are involved in this process. At one time the changes in the RPE were considered to be metaplasic [24], the cells forming fibroblasts and macrophages. However, the RPE cells do not become fibroblasts or macrophages per se [22, 31], but merely look and act like them with characteristics common to both (see next section). Alternative descriptive terminologies to "dedifferentiation" are "transdifferentiation" and epithelial to mesenchyme transition. All three emphasize the magnitude of the change in the RPE that clinically turns the RPE cell from being a friend to a foe of the retina [22].

RPE cells going into their "repair mode" complicate about 10 % of simple detachments and this figure has remained surprisingly constant over the years. Why RPE cells dedifferentiate is not always predictable but broadly relates to such factors as the duration and extent of the detachment or injury, the severity and persistence of inflammation, the presence of appropriate cytokines and growth factors, etc. Under the detachment the RPE cells retract their surface microvilli, become rounded and are variable in size (Fig. 7.3). At this stage they will incorporate cell cycle markers. They start to detach from their basement membrane and can form sheets of cells under the retina usually called subreti-

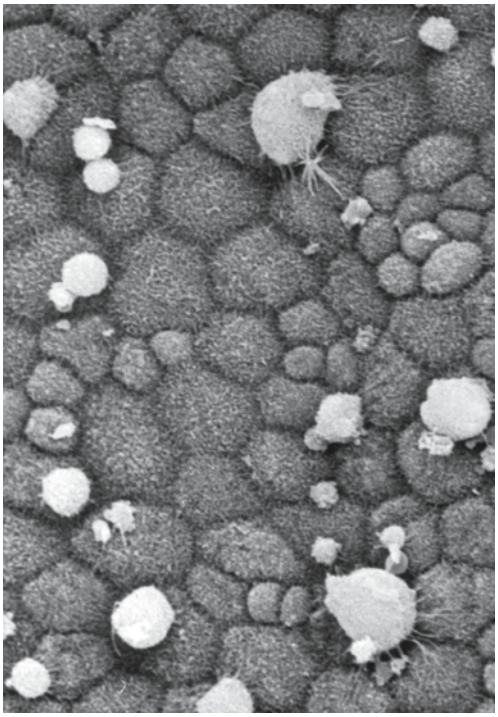

Fig. 7.3. A scanning electron micrograph of RPE under a retinal detachment. The microvilli are retracted and the cells have a rounded appearance

nal membranes. However, the more clinically relevant event is the movement of mobile dedifferentiated RPE cells through the retina or through holes in the retina to form, along with retinal glia and true fibroblasts, scar tissue sheets that are better known as epiretinal membranes. The epiretinal membranes, dominated by dedifferentiated RPE cells in either a macrophagic or fibroblastic phenotype, contract and distort the retina, compounding the visual defect and producing PVR. Contractile compound epiretinal membranes also develop in severe diabetic eye disease. Unlike PVR membranes, these PDR membranes are usually vascularized but, when a retinal hole is present, they also usually are replete with fibroblastic and macrophagic dedifferentiated RPE cells [34].

Aspects of the RPE dedifferentiation process seen in the proliferative retinopathies in vivo are reproduced when RPE cells grow in tissue culture conditions. Many of the proliferating cells are mobile and fibroblastic [29], whereas non-dividing or poorly dividing cells retain an epithelioid shape [10]. Although the fibroblastic phenotype of the RPE abounds in tissue culture [21, 24], the macrophagic form that is so common in pathological conditions is rarely observed in RPE cultures.

If the RPE monolayer is only partly dissociated by enzyme treatment, then clumps of RPE can be cultured (Fig. 7.4 A) and the clumps are a useful model to observe and modulate the dedifferentiation process in vitro (Fig. 7.4 B). The cells at the centre of the clump remain differentiated, static and non-proliferating, whereas the cells at the periphery of the clump dedifferentiate quickest and rapidly undergo the epithelial to mesenchymal transition. As a result after a few days in culture conditions you can follow a clump from centre to periphery and undergo a longitudinal journey down the path of dedifferentiation (Fig. 7.4 B).

If differentiation agents are introduced into the culture medium, then RPE dedifferentiation can be slowed down. We have experience with retinoids, sodium butyrate and mushroom lectin and all three are effective retarding agents. On the other hand, growth factors like FGF, TGFbeta and particularly HGF [12] accelerate either the proliferation or the fibroblastic transition or both. The use of cultured RPE cells for transplant to treat macular and retinal degenerations is of considerable research and even practical clinical interest. However, a full understanding of the biology of RPE differentiation and its controls is essential if transplant and even patching procedures are ever going to progress beyond being just a nice idea.

Summary for the Clinician

- **RPE cells do not become fibroblasts or macrophages per se but merely look and act like them. Under the detachment the RPE cells retract their surface microvilli, become rounded and are variable in size.**
  The epiretinal membranes, dominated by dedifferentiated RPE cells in either a macrophagic or fibroblastic phenotype, contract and distort the retina compounding the visual defect and producing PVR.
  Aspects of the RPE dedifferentiation process seen in the proliferative

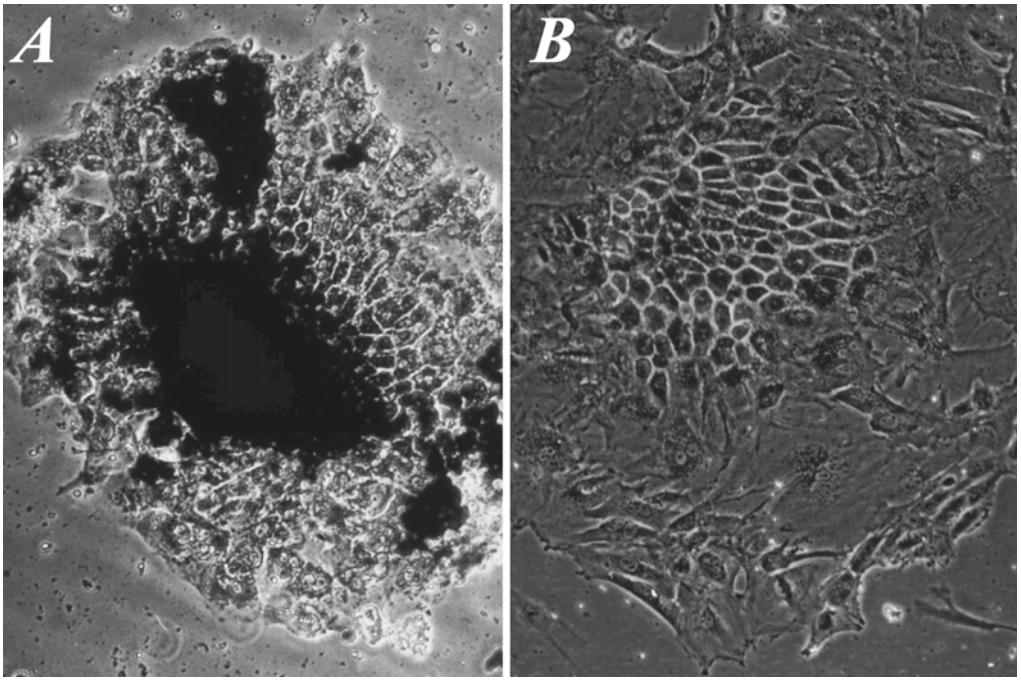

Fig. 7.4. **A** After a few days in tissue culture a clump of the RPE mosaic starts to become depigmented and dedifferentiated at the periphery of the clump. **B** After a few weeks depigmentation is seen throughout the clump. At the periphery RPE cells have elongated and flattened, and many have a fibroblastic or macrophage phenotype

retinopathies in vivo are reproduced when RPE cells grow in tissue culture conditions. If differentiation agents are introduced into the culture medium, then RPE dedifferentiation can be slowed down. We have experience with retinoids, sodium butyrate and mushroom lectin and all three are effective retarding agents

7.3 Detection of Dedifferentiated RPE Cells in Tissues: Variations in Behaviour-Related Proteins Fit Well with Proposed Differences in Activities Between RPE Phenotypes

Perhaps not surprisingly, the expression pattern of various surface and intracellular proteins changes when the RPE cell dedifferentiates. These changes presumably reflect altered RPE cell behaviour. Thus a number of proteins associated with the activities of differentiated RPE cell, such as the cellular retinol- and retinaldehyde-binding proteins that are involved in RPE cell-photoreceptor interactions, are lost in RPE cell dedifferentiation [1]. Conversely proteins linked to activities that RPE cells undertake following dedifferentiation, including a variety of cytoskeletal proteins that are associated with cell migration and tissue contraction, are acquired or increased in the dedifferentiated cells [1, 15, 24, 50, 57]. Although changes in specific proteins can help us to determine the activities of a dedifferentiated cell, the attendant switch in cell appearance makes it notoriously difficult to determine the *origin* of the cell in a pathological tissue such as a PVR epiretinal membrane. This difficulty is especially the case for RPE cells, which not only can adopt the morphology of macrophages or fibroblasts but also tend to "drop" their pigment while dedifferentiating. Moreover, the problem is compounded because other cells in such pathological tissues (including retinal glia and haematogenous – or mo-

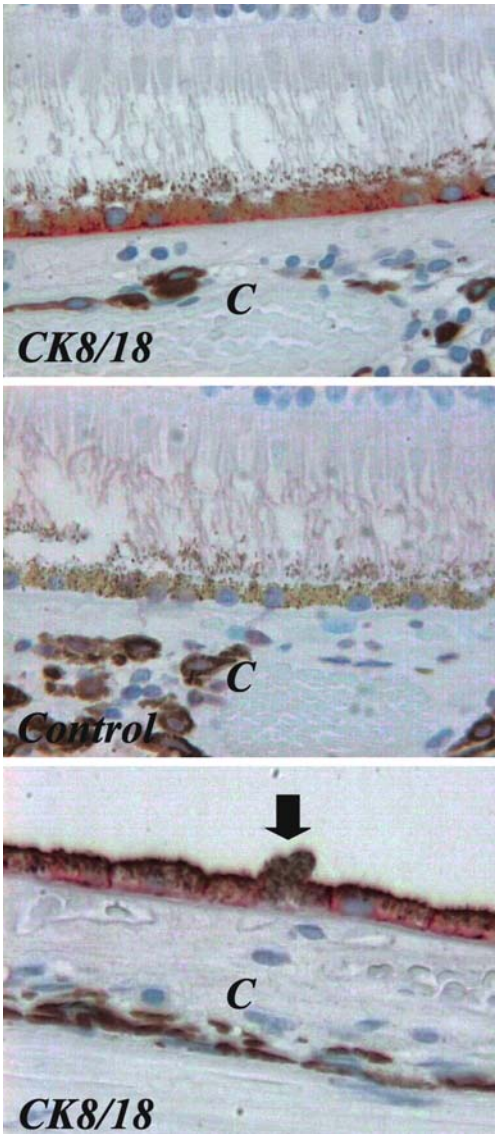

Fig. 7.5. Sections through the chorioretinal interface stained by an immunohistochemical method to reveal the presence of cytokeratin subtypes (stained with a red chromogen, haematoxylin counterstain). The choroid is marked for orientation purposes (*C*). *Top* Differentiated RPE cells express cytokeratins 8 and 18; *middle* control section in which an inappropriate antibody has replaced the cytokeratin antibody; *bottom* beneath a retinal detachment, some RPE cells exhibit a rounded (epithelioid) shape and appear to contain fewer, or even lack completely, cytokeratins 8 and 18

nonuclear phagocyte system – macrophages) tend to take up the pigment. Nonetheless, we need to be able to identify the origins and activities of dedifferentiated cells that are involved in retinal diseases if we are to understand the pathobiology of, and develop logical therapeutic strategies for, these conditions. Fortunately, dedifferentiated RPE cells retain some groups of proteins that are expressed by differentiated cells and that are relatively specific to RPE cells in the context of retinal pathology. Thus these proteins can be used to detect (or "mark") morphologically altered RPE cells in the pathological tissues of a variety of retinal diseases.

One of the best characterized "markers" of dedifferentiated RPE cells is the family of cytoskeletal (intermediate filament) proteins known as cytokeratins [29, 30, 32, 50]. There are over 20 members of this family. Fully differentiated RPE cells express a cytokeratin pattern similar to that seen in simple epithelia in which cytokeratins 8 and 18 are prominent (Fig. 7.5) [44, 50]. Although these two proteins may be downregulated in the epithelioid phenotype (Fig. 7.5), fibroblastic RPE cells are often found to contain them (Fig. 7.6). It is uncertain whether this observation reflects a direct transition from differentiated to fibroblastic cell with retention of the two cytokeratins or a re-emergence of cytokeratins 8 and 18 as epithelioid cells become fibroblastic. Indeed, it is not clear which of these "routes" (direct or via epithelioid) from differentiated to fibroblastic cell occurs in pathological tissues although the latter route is observed in tissue culture (see above) and both fibroblastic and epithelioid cells can be seen in pathological tissue [35]. Since other fibroblastic RPE cells lack cytokeratins 8 and 18 even in the same tissue (Fig. 7.6), it is possible that both routes are in operation.

One cytokeratin that we have not observed in the differentiated RPE monolayer in situ but that is quite abundant in dedifferentiated cells, particularly fibroblastic ones, is cytokeratin 7 (Figs. 7.6–7.8). Our observations fit well with the results of investigations on RPE overlying choroidal melanomas in situ (see review by Kivela and Uusitalo [44]) and with a more recent study that employed proteome analysis in cultured RPE cells [1]. Taken together, our findings and these two studies provide strong evi-

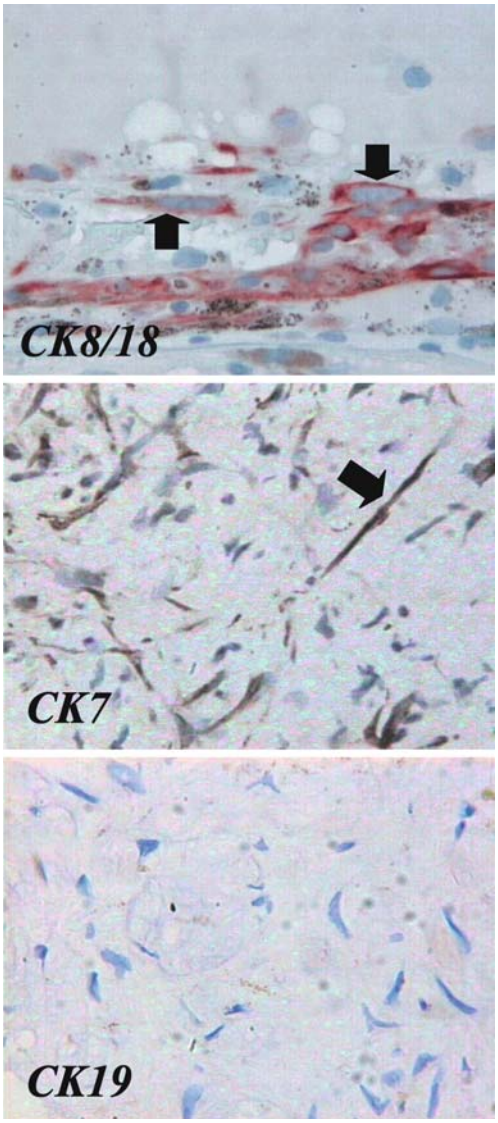

Fig. 7.6. Sections showing fibroblastic dedifferentiated RPE cells in subretinal membranes (SRM) stained by an immunohistochemical method to reveal the presence of cytokeratin subtypes: stained with a red chromogen (*top*) and a brown chromogen (*middle* and *bottom*); haematoxylin counterstain. In the SRM shown at the *top,* several of the fibroblastic RPE cells have retained cytokeratins 8 and 18 (*arrows*). In the SRM depicted in the *middle* and *bottom,* most of the fibroblastic cells contain cytokeratin 7 (*middle*), including extremely spindle-shaped cells (*arrow*). In this SRM, the fibroblastic RPE cells lack cytokeratin 19 (shown at *bottom*) and also are devoid of cytokeratins 8 and 18 (not shown)

dence that dedifferentiated RPE cells may or may not retain cytokeratins 8 and 18, but that cytokeratin 7 can be a major supplementary cytokeratin in dedifferentiated cells.

Cytokeratin 7 is not the only cytokeratin to be upregulated in dedifferentiated RPE cells. Cytokeratin 19 also appears in dedifferentiated cells although in our experience it is less abundant than cytokeratin 7 in pathological tissues [50, 57] (Figs. 7.6–7.8). Nevertheless, this member of the cytokeratin family is interesting because there is evidence that cytokeratin 19 is expressed in migratory RPE cells. This raises the possibility that cytokeratins are not only markers of origin for RPE cells but also may provide information about the activities of the cells. Indeed we have found that an epitope of cytokeratin 18, that is recognized by an antibody called RGE53 and that may be an alternatively phosphorylated type of cytokeratin 18 [44, 50, 57], might too be linked to cell migration. In fact, RGE53 staining appears to be more widespread than cytokeratin 19 in pathological tissues (unpublished observations). By comparing the distribution of these two cytokeratins with extracellular matrix, cell surface receptor or cytoplasmic molecules, it may be possible to obtain information about the mechanisms involved in dedifferentiated RPE cell migration at the same time as confirming the origin of the cell [35, 59].

Cell migration is important in the development of pathological tissues because it may be involved in contraction of reparative material and also is to some extent thought to be responsible for cellular recruitment to the tissue. One RPE phenotype that is considered to be highly motile, and which may be a "vehicle" type for getting to tissues, is the macrophagic one. We have observed macrophage-like CK19-containing cells that also contain CK7 but lack cytokeratin 8 and 18 (Figs. 7.7, 7.8) in tissues. Interestingly, these cells also express the protein CD68 (Figs. 7.7, 7.8). CD68 is a surface and cytoplasmic protein that is highly expressed by bone-marrow-derived macrophages. Although its function is unclear, CD68 may play a role in binding and internalization of ligands and/or lysosomal activities in these cells and it is not found in tertiary differentiated RPE cell. However, it has been shown that cultured RPE cells may synthesize CD68 and the protein

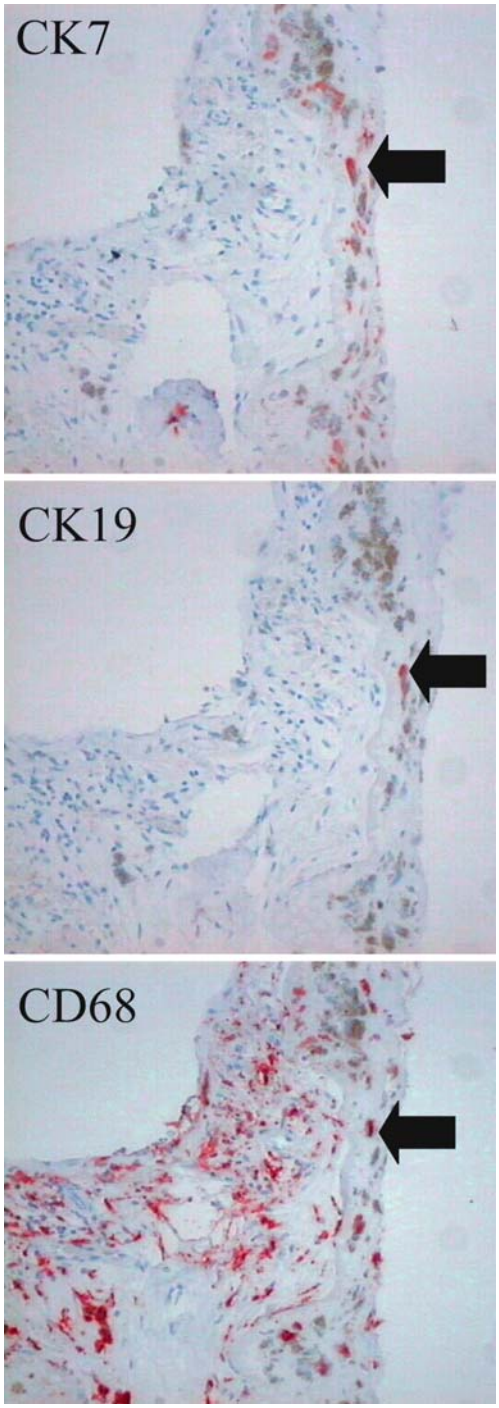

Fig. 7.7. Sections through a choroidal neovascular membrane containing dedifferentiated RPE cells stained by an immunohistochemical method (red chromogen, haematoxylin counterstain) for cytokeratins 7 and 19, and for the macrophage marker CD68 pg respectively. Cytokeratin-7-containing RPE cells are more abundant than those containing cytokeratin 19. One cell is seen to label with both cytokeratins as well as CD68 pg (*arrow*)

has also been found in RPE cells adjacent to laser damage [20, 46]. Therefore, CD68 expression can be used to separate macrophagic RPE cells from other RPE cell phenotypes and such expression correlates well with the hypothesis that these cells at least in part behave (as well as look) like "proper" macrophages.

We have found pigmented cells which contain CD68 but lack CK 8, 18, 7 and 19 (Figs. 7.7, 7.8). Since these cells also lack the leucocyte common antigen CD45, which is present on most leucocytes and is coexpressed with CD68 on bone-marrow-derived macrophages [3, 39] (Fig. 7.8), the origin of these cells is uncertain although they most probably represent macrophagic RPE cells that contain other cytokeratin family members. We have attempted to clarify the origin of these cells with other function-related proteins including those involved in melanogenesis such as tyrosinase. Although it is unclear whether adult RPE cells synthesize melanin, there is evidence that they contain tyrosinase in several species and that adult human RPE cells in tissue culture (where they tend to be dedifferentiated) can synthesize melanin in certain conditions (reviewed by Boulton 1998) [11]. We have attempted to use tyrosinase as a marker for dedifferentiated RPE cell generally, and specifically for macrophage-like RPE cell. We have been able to show that in retinal detachment some, but not all, macrophagic RPE cells contain both CD68 and tyrosinase (Figs. 7.8, 7.9). Thus, by investigating different cytokeratin subtypes and/or tyrosinase in conjunction with CD68, we can demonstrate that at least some pigmented macrophagic cells in pathological tissues are indeed of RPE origin and that during dedifferentiation these cells have acquired parts of the functional "machinery" that bone-marrow-derived macrophages possess.

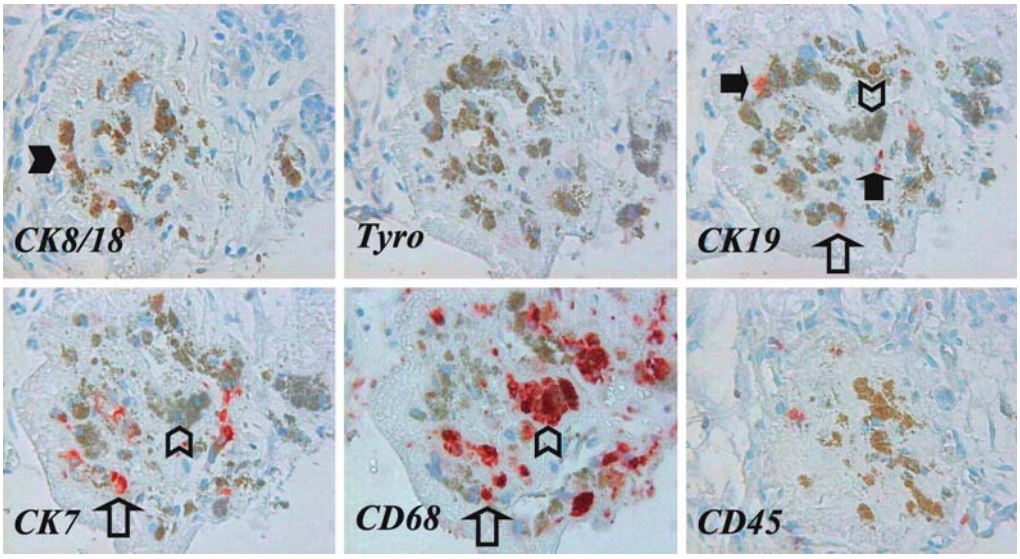

Fig. 7.8. A cluster of dedifferentiated RPE cells stained by an immunohistochemical method (red chromogen, haematoxylin counterstain). Only one or two cells contain simple epithelial-type cytokeratins (*closed arrowhead* CK8 and 18) and all lack tyrosinase (*Tyro*). Several cells contain cytokeratin 19 (*closed arrows* CK19), but these cells mostly lack cytokeratin 7 (*CK7*). Cytokeratin 7 seems to be the most abundantly expressed of the cytokeratins investigated in this tissue (*CK7*). A group of macrophage-like pigmented cells that labels with CD68 pg lacks cytokeratins 7 and 19 (*open arrowheads* CK19, CK7 and CD68) although at least one other cell appears to label for CK19, CK7 and CD68 (*open arrows*). The macrophages lack CD45 although other CD45 positive cells are present (CD45)

Summary for the Clinician

- **One of the best characterized "markers" of dedifferentiated RPE cells is the family of cytoskeletal (intermediate filament) proteins known as cytokeratins. Dedifferentiated RPE cells may or may not retain cytokeratins 8 and 18, but cytokeratin 7 can be a major supplementary cytokeratin in dedifferentiated cells. CD68 expression can be used to separate macrophagic RPE cells from other RPE cell phenotypes. In general, the identification of various cytoplasmic and cell surface proteins can help detect dedifferentiated RPE cells in retinal pathology. Furthermore, these investigations are beginning to yield information concerning the activities of these cells in the tissues. Therefore, we are exploiting this approach to examine other key proteins involved in RPE differentiation, including junctional proteins such as the connexins and the cadherins, growth factors and extracellular matrix (ECM) elements**

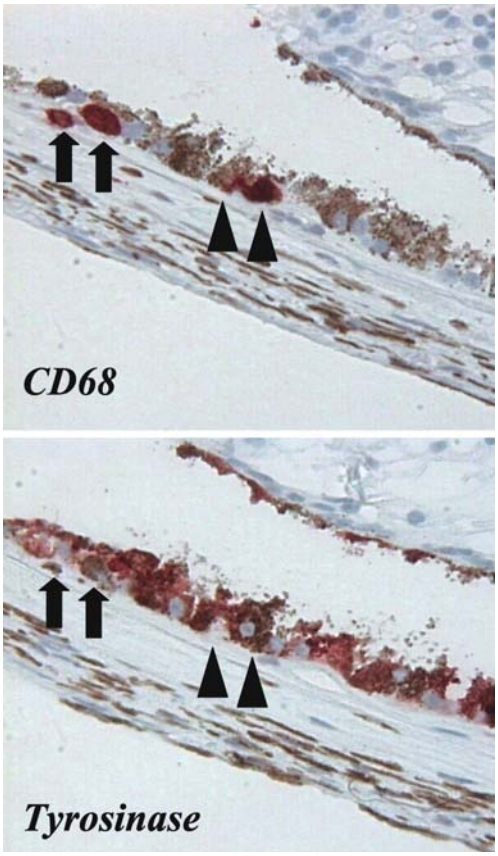

Fig. 7.9. Sections through the choroid and RPE stained by an immunohistochemical method for the macrophage marker CD68 pg and for tyrosinase (stained with a red chromogen, haematoxylin counterstain). Two CD68 positive cells stain for tyrosinase (*arrowheads*) and two do not (*arrows*)

7.4 Experimental Studies of RPE Cell Differentiation and Dedifferentiation

Many growth factors and ECM proteins have been implicated in the differentiation and dedifferentiation of RPE cells in diseases such as those seen in proliferative retinopathies. This work has been mainly based on experimental data obtained from either studies of in vitro cultured RPE cells or histopathological studies of surgically removed human tissue, or from using suitable animal models. Studies such as these have identified the RPE not only as an important source of mediators (e.g. growth factors; GF) involved in tissue maintenance and homeostasis in the differentiated monolayer but also involved in the protracted wound repair processes seen in diseases such as PVR. This encom- passes growth factor and ECM production along with cellular migration, proliferation and tissue contraction. Moreover, the RPE cell has revealed itself to be a central cell type acting in and regulating these processes.

7.4.1 Differentiation

Although in vitro cultures of human RPE cells have been used extensively to elucidate cellular functions, a note of care when interpreting results from different studies is required. It is essential to realize that tissue culture conditions are likely to provide an environment for RPE cells to adopt a dedifferentiated morphology. This can result in their ability to express functions completely different from the highly differentiated non-dividing RPE cells within the monolayer in the normal eye [28, 66]. However, by using specific measures, RPE cells in vitro can be made to differentiate into a phenotype resembling RPE in vivo, displaying polarity, a columnar morphology, the presence of gap and tight junctions as well as the presence of microvilli at their apical surface [8, 54, 66]. Unfortunately, many publications fail to describe the state of differentiation of the RPE cells under investigation, often rendering the findings of these studies of limited value. RPE cells may be cultured on filter systems, allowing evaluation of polarized GF secretion, or in co-culture systems with choriocapillaris (CC) endothelial cells or neuroretinal explants [8]. Exposing the cells to substances relevant to the pathogenesis of specific diseases, e.g. for AMD, rod outer segments (ROS) or advanced glycation end products (AGE), provides ways to investigate particular mechanisms involved in a specific disease process such as lipofuscin formation and oxidative stress in AMD [4]. There are obviously a plethora of growth factors, ECM proteins and cell-cell interactions involved in both the differ-

entiation and dedifferentiation of RPE and as such each topic will be discussed briefly, using specific examples where appropriate to highlight the depth and complexity of these interactions.

7.4.1.1 Cell-Growth Factor Interactions

The differentiated RPE cells serve many functions including storage of essential reserves and metabolites for the photoreceptors; they act as a barrier between the leaky choroidal vessels and the neural retina, they scavenge free radicals and also they actively transport materials to the photoreceptors. Being pigmented, they absorb stray light and help to create a clear image. Collectively the differentiated RPE cell performs functions essential for photoreceptor survival and secretes factors important for homeostasis of the outer neuroretina. Due to the polarized nature of the RPE cells, they can secrete components in a directional fashion. For example, matrix metalloproteinase-2 (MMP-2), tissue inhibitor of metalloproteinase-1 (TIMP-1) and hyaluronan are synthesized by the RPE and are secreted preferentially from the apical surface towards the photoreceptors [55], while TIMP-3, fibroblast growth factor 5 (FGF-5), interleukin-6 (IL-6) and interleukin-8 (IL-8) are secreted from the basolateral surface towards the underlying choroid [16, 19, 38]. The polarization of RPE cells is a dynamic event that is under constant regulation by signalling molecules coming from the choroid and the retina or extracellular stimuli. Extracellular matrix components from RPE modulate the polarity of secretion of angiogenic factors, such as vascular endothelial growth factor, from the RPE [8]. Thus the polarity of the RPE plays a role in maintaining a balance of secreted factors essential for the functional RPE. Indeed the complexity of RPE function is readily demonstrated when we investigate how the monolayer maintains a healthy underlying choroid with pro-angiogenic factors while maintaining anti-angiogenic properties to prevent abhorrent vessel growth.

Clinical and experimental observations show that in the normal adult eye, destruction of the RPE by laser invariably leads to atrophy of the CC and photoreceptors, which is preceded by loss of fenestrations of the CC endothelium. Numerous studies have illustrated that one of the functions of the RPE may be to lend trophic support to the retina by production of neurotrophic GF such as nerve growth factor (NGF), brain-derived growth factor (BDNF) [40] and pigment epithelium-derived factor (PEDF) [40, 41]. BDNF may also promote differentiation of the RPE itself in an autocrine manner [25]. PEDF, a 50-kDa secreted glycoprotein, is an important neurotrophic factor and has been shown to contribute to survival and differentiation of photoreceptor and neuronal cells of the retina and central nervous system [5, 14]. Both in vivo and in vitro, PEDF is mainly produced by highly differentiated RPE cells [53]. In addition to its neurotrophic properties, PEDF is a potent inhibitor of angiogenesis. PEDF may contribute to maintaining the angiogenic balance in ocular tissues as it is responsible for excluding vessels from invading the retina, vitreous and cornea [9] and as it is downregulated in human and experimental conditions with ocular neovascularization, such as proliferative retinopathy and exudative AMD [53]. These biological activities are of great importance for the development and maintenance of normal physiological functions in the eye.

In addition to these angiogenesis inhibitors, the RPE has been convincingly shown to produce the pro-angiogenic VEGF-A, both in vivo and in vitro. In vitro, VEGF production is considerably higher in differentiated than in undifferentiated RPE cells, a finding that supports a physiological role of VEGF-A [8]. In vitro, VEGF secretion by highly differentiated RPE cells was shown to be directed preferentially towards the basal side of the cells [8], whilst in vivo in the adult human choroid, the VEGF receptors 1, 2 and 3 are expressed by the CC endothelium on its side facing the RPE cells [8]. As VEGF can act as a survival factor for endothelial cells [4], and as it induces a fenestrated permeable endothelial phenotype [18], these findings indicate that VEGF-A is involved in the physiological maintenance function of the resting RPE on the choroid [8]. Indeed, in line with this view, it was shown that in the absence of an RPE layer, the choroid does not develop and it has been sug-

gested that VEGF is one of the factors involved in choroidal development driven by the embryonic RPE, a paracrine relationship that may persist throughout life to maintain the CC [67]. However, the expression of other members of the VEGF family indicates there are several factors also likely to have a role in this function. Indeed, constitutive expression of placenta growth factor (PlGF) in mouse choroid [56] has recently been demonstrated as has VEGFR-3, a ligand for VEGF-C and -D (but not for VEGF-A), in the human CC in vivo [8].

7.4.1.2 Cell-ECM Interactions

Although for the context of this chapter the cell-ECM interactions and cell-growth factor interplay have been divided into separate sections for convenience, there is considerable overlap between these groupings. For instance, not only does the substrate on which a cell resides influence its response to a specific factor but also growth factors themselves can often be found to be resident ECM proteins. Interestingly, biochemical and immunochemical studies have identified PEDF as a extracellular protein localized in the interphotoreceptor matrix (IPM), vitreous and aqueous of several mammalian species [6].

In the normal eye, the direct cell-ECM interaction with the RPE monolayer is at the apices of the RPE closely applied to the photoreceptor outer segments and IPM whilst the basal surfaces of the RPE adhere to Bruch's membrane.

Bruch's membrane is a basement membrane complex located between the retinal pigment epithelium (RPE) and the choroid, and it is a pentalaminar structure with a central elastin layer bordered on either side by a collagenous zone. There is the basal lamina of the RPE at the innermost collagenous layer of Bruch's membrane and a second basal lamina, produced by the endothelial cells of the choriocapillaris, which is juxtaposed to the outer collagenous layer. Although much more remains to be learned, the basic ultrastructural organization and biochemical composition of Bruch's membrane are similar to basement membrane complexes in other tissues, such as the choroid plexus, kidney glomerulus, and airway alveoli, where an epithelial-endothelial juxtaposition occurs. All ionic exchange and metabolic traffic from the neural retina and RPE to the choroidal capillaries, and vice versa, must traverse Bruch's membrane, thus leaving the neural retina vulnerable to any disruptions of those processes. It is well documented that Bruch's membrane undergoes a number of changes throughout life that include increased thickening, protein cross-linking and reduced permeability to nutrients as well as increased amounts of lipid deposition and the accumulation of basal laminar deposits and drusen (for more detailed reviews see later in this book).

The composition of Bruch's membrane appears to alter with age, but at any time of life the membrane contains a variety of components. Constituents which have been reported in Bruch's membrane include proteoglycans such as heparan and chondroitin sulphate, collagens such as types I and III to VI collagen, and glycoproteins such as laminin and fibronectin (reviewed by Campochiaro et al. 1986; Marshall et al. 1993 [13, 48]). However, the distribution of the individual components varies between layers of the membrane. Thus not surprisingly types I, III and V collagen are found chiefly in the inner and outer collagenous zones [47] whilst collagen type IV and laminin are found in the basement membranes of the RPE and of the choriocapillaris. Although there appears to be more immunoreactivity for both proteins in the CC, it may be that this difference reflects variation in the availability of antigenic sites between the two basement membranes.

Marshall and colleagues [47] found focal deposition of laminin in the inner collagenous laminar immediately adjacent to the RPE basement membrane; these authors suggested that the laminin patches may represent anchoring plaques in a rudimentary basement membrane complex. However, there is still a gap in our knowledge as to the exact composition of Bruch's membrane and all receptors are receptors involved in RPE-Bruch's interactions. This is despite the explosion of new information over the last decade or so regarding composition of basement membrane components elsewhere in the body and the number of potential receptors.

If we take laminin as an example of a protein known to be present in Bruch's membrane, it is not in fact a single protein but a complex of multi-domain proteins built up from different modules. The diversity of the molecules is made greater by the occurrence of homologous but distinct α, β, and γ chains which combine into different trimeric isoforms and can be compounded further with different variants due to alternative splicing and proteolytic processing. At least 12 known forms of laminin trimer show a restricted expression in basement membranes and although at least one member of the α-, β-, and γ-chain families appears to be present in every basement membrane studied, this does not preclude the presence of other laminins [64].

The presence of laminin in potential adhesion plaques in the inner part of Bruch's membrane is consistent with the expression of cell surface receptors known as β_1 integrins for which laminin isoforms are ligands [17] on normal RPE [2]. Since integrins (heterodimeric receptors composed of an α and a β subunit) are major cell surface receptors which mediate adhesion, it may be that interactions between β_1 integrins on the RPE basal aspect and matrix components such as laminin and type IV collagen in Bruch's membrane play an important role in RPE adhesion to the membrane. Consistent with this theory, recent in vitro studies demonstrate that blocking β_1 integrins with appropriate antibodies reduces the RPE attachment rate to Bruch's membrane [37].

Plaque-like adhesions would not only provide attachment for RPE to Bruch's membrane but also could permit indirect contact between RPE and components "deeper" in Bruch's membrane (e.g. in the inner collagenous zone). These components would in turn be in a position to signal to the cells. However, the integrity of Bruch's membrane and of the RPE cells themselves is also a key component in this interaction. This has been clearly demonstrated in the work investigating RPE cell transplantation. Cellular adhesion is key for the survival of any epithelia and the RPE cell requires adhesion to a suitable substrate if it is to avoid undergoing death by anoikis (apoptosis induced by detachment of a cell from a substrate) or apoptosis. Indeed work by Tezel and Del Priore [61] showed that RPE cells die within 24 h if attachment to Bruch's membrane has not occurred. Furthermore, even in successful RPE transplants into laboratory animals onto a normal Bruch's membrane or onto in situ RPE cells, there are considerable numbers of transplanted cells, which do not adhere and subsequently die.

Adhesion to Bruch's membrane is therefore a crucial step in the success of the transplant procedure. However, the condition of Bruch's membrane, which affects RPE adhesion (see above), will be difficult to predict and varies with each patient. That deterioration of Bruch's membrane is problematic is supported by the work of Del Priore and Tezel [62], who have shown that RPE cells have markedly different settlement rates on the different layers and components of Bruch's membrane. Furthermore, much less adhesion occurs to the deeper levels of Bruch's membrane than to the RPE basal lamina. Furthermore, aged cells appear to be much less adept at repopulating Bruch's compared to fetal cells. In addition, Shirigami et al. [60] have shown that embryonic RPE cells are unable to differentiate on severely damaged Bruch's membrane. On the other hand, cultured RPE harvested from older donor eyes can attach and grow well, and reach confluence in culture, on substrates such as bovine ECM coated culture dishes. Work by Tsukahara and colleagues [63] has shown that freshly isolated autologous cells do not adhere as well to or survive on surfaces similar to that found in patients following removal of CNVs. Few if any cells were seen to survive on the submacular basement membrane or on the inner collagenous layer. This implies that attachment followed by proliferation would not repair such a defect. Indeed it has recently been shown that RPE basement membrane supports RPE resurfacing of localized defects but the deeper portion of the inner collagenous layer impedes such repopulation [65]. However, these fundamental differences show that cells can be manipulated in culture prior to transplantation into the eye and it is perhaps this point that bodes well for the future of transplantation.

Summary for the Clinician

- **The polarity of the RPE plays a role in maintaining a balance of secreted factors essential for the functional RPE. One of the functions of the RPE may be to lend trophic support to the retina by production of neurotrophic GF such as nerve growth factor (NGF), brain-derived growth factor (BDNF) and pigment epithelium-derived factor (PEDF). In vitro, VEGF secretion by highly differentiated RPE cells was shown to be directed preferentially towards the basal side of the cells**
- **In the normal eye, the direct cell-ECM interaction with the RPE monolayer is at the apices of the RPE closely applied to the photoreceptor outer segments and IPM whilst the basal surfaces of the RPE adhere to Bruch's membrane. It may be that interactions between β_1 integrins on the RPE basal aspect and matrix components such as laminin and type IV collagen in Bruch's membrane play an important role in RPE adhesion to the membrane. Adhesion to Bruch's membrane is a crucial step in the success of an eventual transplant procedure. Cells can be manipulated in culture prior to transplantation into the eye and it is perhaps this point that bodes well for the future of transplantation**

7.4.2 Dedifferentiation

As outlined in the above sections the RPE cell normally is a differentiated cell, and as a consequence it is maintained out of the cell cycle of division, but unlike its neighbour the photoreceptor cell the differentiation is not terminal. In numerous retinal pathologies including inflammation and vascular disease, RPE cells change from being differentiated, stationary and non-dividing cells into undifferentiated, migratory and proliferating ones. This loss of differentiation is associated with a change in appearance to one more characteristic of a fibroblast or macrophage (Fig. 7.10). Proliferating RPE cells in culture have the characteristics of the undifferentiated RPE cells that are seen in retinal pathologies. Therefore, they are of considerable use to us for studying in simple models the role of RPE in scar formation.

7.4.2.1 Cell-Growth Factor Interactions

There have been a number of growth factors and enzymes that have been implicated in RPE dedifferentiation in various retinopathies such as PVR. These include hepatocyte growth factor/scatter factor (HGF), platelet derived growth factor (PDGF-B), tumour necrosis factor (TNF-α), TGF-β and connective tissue growth factor (CTGF), MMP-2 and MMP-9. In the context of PVR these factors will be discussed in greater depth elsewhere in this book (see Chap. 9 by D. Charteris). However, a brief discussion of the cellular effects of HGF on RPE as an example of growth factor-RPE cell interaction is outlined below.

HGF is a glycoprotein that causes epithelial cells to become fibroblastic and mediates epithelial-mesenchymal interactions in many tissues. It has autocrine-paracrine activity in RPE cells and is a major regulator of RPE barrier function. It does not take much of a leap to think that the HGF/SF and c-Met system could play a key role in the RPE loss of communication, shape change and mobilization so important to the early pathogenesis of PVR. It turns out, not too surprisingly, that HGF/SF may well be a key player. Human RPE cells do not form particularly good colonies so scattering cannot easily be appreciated in culture. On the other hand, these cells do undergo an impressive elongated shape change to a more fibroblastic appearance in the presence of HGF/SF [12]. In addition, HGF/SF is a weak mitogen but an effective motogen for cultured human RPE cells [23, 45]. Since migratory RPE cells are a key factor in retinal pathology, HGF/SF may play an important role in a variety of retinal diseases including PVR membrane formation. Indeed, we have found positive immunohistochemical localization of the HGF/SF receptor c-Met to cells in PVR epiretinal membranes [12]. Subsequent work in our laboratory has established a co-distribution of c-Met and cytokeratin (epithelial marker) in at least some

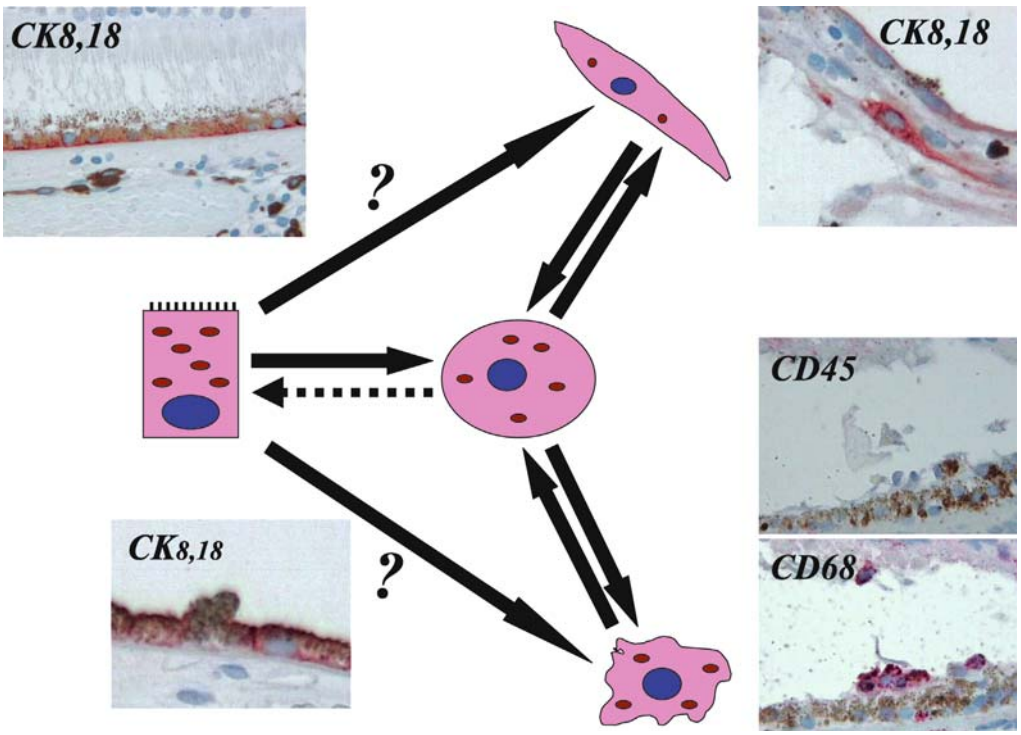

Fig. 7.10. Schematic diagram illustrating the phenotypic changes that the RPE cell can undergo. Photomicrographs illustrate known immunochemical markers for each phenotype (see text). The factor(s) that drive these phenotypic changes are not entirely known, whilst the challenge in RPE transplants is whether the dedifferentiated cell can be differentiated fully (*dotted arrow*), (?) = possible additional dedifferentiation route

of the epiretinal membranes. Further, work by others [45] found that three out of three PVR membranes contained cells positively immunostained for the c-Met receptor whereas only one out of three idiopathic epiretinal membranes was positive. That glial cells rather than RPE cells predominate in many idiopathic membranes is the probable explanation for the lack of positive cells seen by the authors.

Based on current thinking it would be unlikely for any of the major constituent cells in idiopathic epiretinal membranes (glia, fibroblasts and inflammatory cells) to carry the c-Met receptor. It does appear likely that the vitreous in PVR patients contains more HGF/SF than the vitreous of patients with pathologies associated with little proliferative and migratory behaviour such as macular holes. However, the values for individual patients can be extremely variable. Values published by Nishimura et al. [52] ranged between 1.10 and 7.38 ng/ml whereas our own figures gave a range of 3 ng/ml going up to 250 ng/ml [12]. Such variation is intriguing but by no means exceptional for studies of cytokine distribution in vitreoretinal diseases. However, at least some of the HGF/SF is active and mature because PVR vitreous can induce scattering of colonies of MDCK cells [12].

Limited evidence is available in the literature on c-met receptor distribution in the neural retina [45]. RPE cells also are producers of HGF/SF. He and co-workers [27] consider, on the basis of positive RT-PCR and ELISA assays on serum starved RPE in culture, that the cells produce message for HGF/SF and secrete the peptide into the culture medium. These authors think that RPE cells are one of the cell types, uncommon in the body as a whole but strangely prevalent in the eye, that express both the receptor and the growth factor (perhaps indicating

an autocrine loop). Alternatively, others [45] have failed to identify HGF/SF production by RPE in culture. As a result of these findings it is suggested that HGF/SF reaches RPE by a paracrine route. It remains to be established which observation is correct.

7.4.2.2 Cell-ECM Interaction

Cell-matrix interactions are also key to the pathogenesis of retinopathies such as PVR, and PVR membranes often contain numerous dedifferentiated RPE cells along with a number of extracellular proteins. The extracellular proteins include the adhesive proteins like collagen, laminin and fibronectin as discussed above; however, several matricellular proteins that have potential counteradhesive functions have also been localized. Two such matricellular proteins, thrombospondin 1 and osteonectin (or SPARC: secreted protein acidic and rich in cysteine), tend to be co-distributed with the dedifferentiated RPE cells in PVR membranes [59]. Their ability to modulate focal cell adhesions implies that thrombospondin 1 and SPARC may reduce RPE cell-matrix adhesion and so permit the key RPE cellular activities characterized in dedifferentiation such as migration, shape change and proliferation [36]. If we are to obtain a functioning monolayer on Bruch's membrane by transplantation, establishment and maintenance of differentiated cells is imperative and the effect of the plethora of factors that can cause RPE cells to dedifferentiate including ECM proteins, growth factors such as HGF see also Chap. 9 by D. Charteris) as well as even the vitreous itself may need to be neutralized.

7.4.2.3 In Vitro Collagen Gel Model of RPE Dedifferentiation

Following the discovery of a number of factors associated with RPE dedifferentiated in vivo, we and others have further examined their role in more detail using an in vitro model of RPE dedifferentiation. We have adapted an in vitro model [49] of cell-matrix interactions (i.e. the collagen matrix system) for this purpose (Fig. 7.11).

The mechanisms by which RPE cells interact with collagen matrices are beginning to be understood and we have recently published data relating some of the different mechanisms involved [58]. It is apparent that these mechanisms may depend on cell surface receptors such as integrins, cell-ECM interaction via glycoproteins (such as TSP1 and SPARC [26, 36,], lectins [42, 43] or enzymatic intervention, i.e. matrix metalloproteinases or MMPs) [58].

The model has been utilized to examine cell-cell, cell-ECM and cell-GF interaction. We have shown that TSP1 and SPARC are present during collagen contraction, but neutralising these glycoproteins with monoclonal antibodies has not prevented RPE cells from dedifferentiating and contracting the surrounding collagen matrix. However, antibodies directed against cell surface receptor integrins $\alpha 2$ and $\beta 1$ subunits have significantly prevented RPE-mediated collagen matrix contraction [58].

We have also shown there is a role for MMPs during RPE cell mediated contraction of collagen matrices [58]. Using various techniques (immunohistochemistry, ELISA, zymography), we were able to show expression of a number of MMPs during the contraction assay. Furthermore, the evaluation of a broad-spectrum MMP inhibitor, a hydroxamic derivative known as Galardin-MPI, which is known to inhibit the activity of all MMPs, has helped to determine that MMP production within the collagen gels was essential rather than incidental. We have shown that Galardin-MPI has no effect on cell viability, adhesion or proliferation and as such its anti-contractile effects appear to be via inhibition of MMPs [58]. What was of particular interest, however, was that even when RPE cells are seeded on the same substrate (collagen type I) in otherwise identical conditions, they can still adopt different morphologies depending on whether the cells are seeded within or on the substrate [21, 49, 58].

Although many of the proteolytic enzymes produced by RPE cells in situ probably are apically secreted towards the interphotoreceptor matrix (which occupies the subretinal space between the photoreceptors and is composed of large glycoproteins and proteoglycans: see [51]

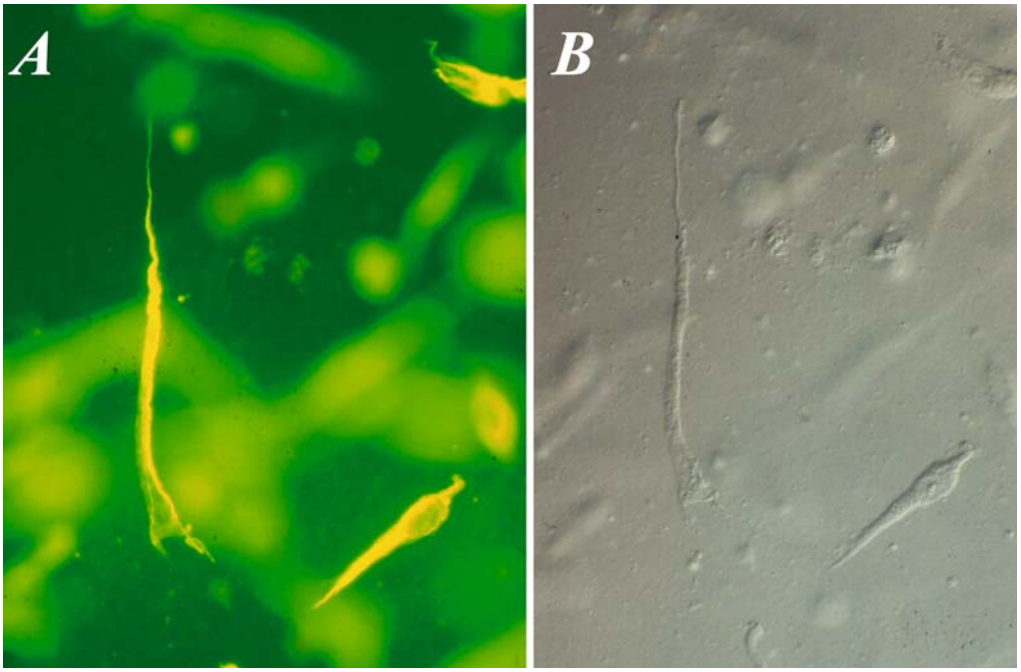

Fig. 7.11 A, B. Photomicrograph of RPE cells within a collagen matrix. The cells are immunochemically positive for cytokeratin 18 (**A**) and adopt a fibroblast spindle shaped morphology. **B** Corresponding DIC micrograph of **A**

for review), it is possible that such enzymes also would allow the cells to modify their underlying matrix after synthesis and deposition. It is known that RPE cells in culture are capable of producing a range of enzymes which lyse a variety of matrix proteins [58], including matrix metalloproteinases. There is now evidence that RPE cells in situ also make some of these enzymes as well as tissue inhibitors of the enzymes [55]. Perhaps the balance between matrix metalloproteinases and their tissue inhibitors supports the strong adhesion that the normal RPE has for components of Bruch's membrane. Loss of this balance might play an important role in the early stages of PVR and other retinal pathologies.

Summary for the Clinician

- **Proliferating RPE cells in culture have the characteristics of the undifferentiated RPE cells that are seen in retinal pathologies. Therefore, they are of considerable use to us for studying in simple models the role of RPE in scar formation. RPE cells do undergo an impressive elongated shape change to a more fibroblastic appearance in the presence of HGF/SF. We have found positive immunohistochemical localization of the HGF/SF receptor c-Met to cells in PVR epiretinal membranes. Two matricellular proteins, thrombospondin 1 and osteonectin (or SPARC: secreted protein acidic and rich in cysteine), tend to be co-distributed with the dedifferentiated RPE cells in PVR membranes. They permit the key RPE cellular activities characterized in dedifferentiation such as migration, shape change and proliferation**

Acknowledgements. This work was supported by the Dunhill Medical Trust, Fight for Sight, the R&D Support Fund of the Royal Liverpool and Broadgreen University Hospitals and St. Paul's Foundation for the Prevention of Blindness and the Guide Dogs for the Blind Association.

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Risk Factors in Proliferative Vitreoretinopathy

8

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Core Messages

- Clinical and biological risk factor analysis has a major impact on the management of proliferative vitreoretinopathy (PVR)
- Understanding risk factors helps us improve surgical techniques and devise new drug therapies
- Prediction of the probability of PVR formation can categorize patients into high and low risk
- Risk factors used for prediction include PVR grade, crystalline lens status, uveitis, size of retinal detachment, vitreous haemorrhage and the previous use of cryotherapy
- Adjuvant drug therapy can be targeted at patients who are at high risk of developing PVR

8.1 Introduction

Modern vitreoretinal surgery has greatly improved the success rate of retinal reattachment surgery. Proliferative vitreoretinopathy (PVR) is still the most important cause of failure of surgery. The understanding of risk factors involved in the development of PVR can have a major impact on the management of retinal detachment surgery, for example if cryotherapy is known to be a risk factor, the alternative technique of laser retinopexy should be considered.

Our increasing knowledge of the biological risk factors and pathological processes can contribute to the development of new drug treatments targeted at specific pathological pathways.

Clinical and biological risk factors can be used to predict the probability of PVR formation in a particular patient. Using this information, patients at high risk of developing PVR can be targeted for adjuvant drug treatment.

8.2 Clinical Risk

8.2.1 Preoperative Risk Factors

8.2.1.1 Vitreous Status

The state of the vitreous and its association with the development of PVR remains controversial. Vitreous haemorrhage, incomplete posterior vitreous detachment and vitreous syneresis have been suggested to be risk factors.

Vitreous haemorrhage in retinal reattachment surgery may occur either preoperatively following development of retinal breaks, intraoperatively associated with surgical manipulations or more rarely postoperatively usually as a result of residual vitreous traction to retinal vessels with or without formation of recurrent retinal breaks. Although the association between vitreous haemorrhage and PVR has been reported in several studies [7, 28, 56, 60], it is debatable whether the former constitutes an independent risk factor [19].

Oguchi was the first to postulate that blood elements could induce "connective tissue prolif-

eration" on the retinal surface [51]. Subsequent animal studies have shown that intravitreal injection of whole blood or red blood cells stimulates the formation of glial epiretinal membranes [20, 46]. The investigators suggested that injection of autologous red blood cells into the vitreous induces a phagocytic response. Haemoglobin particles released from lysed erythrocytes are removed by intravitreal or extravitreal macrophages. This low grade chronic inflammatory response results in focal dissolution of the inner limiting membrane (ILM), allowing Müller cell processes and astroglial cells to migrate through the breaks in the ILM and proliferate on the retinal surface [46]. In the vitreous cavity these cells de-differentiate into non-specific fibroblasts and lay down newly formed collagen [17].

In addition, serum components including fibronectin and platelet-derived growth factor (PDGF), along with other factors released by increased retinal pigment epithelial cell proliferation and migration, result in the development of PVR [46].

Clinical studies investigating the role of vitreous haemorrhage in PVR formation have been inconclusive. Grizzard et al. in a prospective study of retinal detachments treated with scleral buckling reported that vitreous haemorrhage was a significant risk factor for poor anatomical surgical outcome [28]. Similarly, Tolentino in a series of 54 cases observed that vitreous haemorrhage was a common precipitating factor for development of PVR [56]. Subsequent studies have also demonstrated that the incidence of preoperative vitreous haemorrhage was significantly higher in eyes with evidence of PVR either at the initial presentation or following retinal reattachment surgery [7, 60]. However, Kon and Ducuesne in two prospective studies of 140 and 48 eyes respectively showed that preoperative vitreous haemorrhage is not an independent risk factor for the development of postoperative PVR in rhegmatogenous retinal detachment [19, 37].

The inconsistent results above are predominantly attributable to the different study designs, inclusion criteria and diversity in surgical techniques used. The wealth of evidence suggests that an association exists between vitreous haemorrhage and PVR; however, whether vitreous haemorrhage is an independent risk factor remains to be fully evaluated in the future.

The presence of incomplete posterior vitreous detachment (PVD) in patients with retinal detachment has also been implicated as a risk factor in the development of both pre- and postoperative PVR [7, 12]. Capeans et al., in a prospective, controlled study of 102 eyes with retinal detachment, reported that the prevalence of partial PVD in eyes complicated by PVR was 87 % as opposed to 35 % in the control eyes with no evidence of PVR [12]. In addition, over a mean follow-up of approximately 6 months the incidence of recurrent retinal detachment due to postoperative PVR was 33 % and 4.9 % in eyes with or without incomplete PVD respectively. It seems that in eyes with partial PVD, the posterior hyaloid remains attached to the retina by means of either early proliferative tissue or some previous abnormal vitreoretinal adherence. The predisposition to PVR is thought to be associated with the regulatory role of the vitreous on retinal pigment epithelial (RPE) cell behaviour when they come into contact [12].

Proliferative vitreoretinopathy in the presence of partial PVD can occur with or without syneresis of the vitreous gel. Tolentino, in his series of eyes with massive preretinal retraction, reported that in all cases the fundamental finding was extensive degeneration and shrinkage of the posterior vitreous cortex, which remained adherent to the detached retina [56]. Conversely, other studies show that incomplete PVD with no collapse of the vitreous gel may be a determining factor in the development of severe PVR [7].

8.2.1.2 Preoperative PVR

A number of studies have identified the presence of preoperative PVR (Fig. 8.1) as a major risk factor for the development of postoperative PVR [7, 18, 25, 37, 50]. Girard et al. in a large retrospective study of 1,020 patients with no or up to Grade C1 PVR at initial presentation reported that preoperative PVR Grade A and to a lesser extent Grade C were significant predictive variables for postoperative PVR [25]. Moreover, Bonnet et al. in two

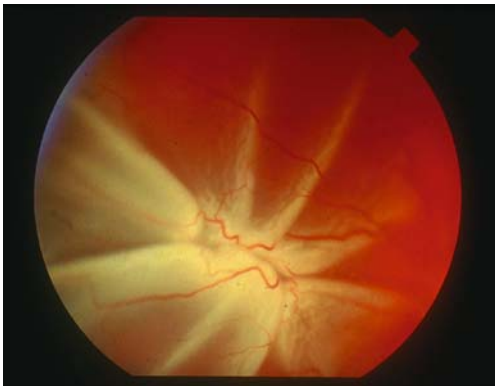

Fig. 8.1. Preoperative PVR as a risk factor

studies, demonstrated the clinical correlation between preoperative PVR Grade B or C and the development of postoperative PVR [6, 7]. More recently, Kon et al. in a prospective study of 140 patients who underwent primary pars plana vitrectomy for retinal detachment found that preoperative PVR Grade C involving more than 1 clock hour of the retina constitutes a significant risk factor for postoperative PVR [37]. However, in this study PVRs of Grade A or B were not evaluated as risk factors as a large number of these patients did not require vitrectomy.

Although the strong association between pre- and postoperative PVR is widely accepted, it remains controversial which stage of preoperative PVR poses the greatest risk. It has been suggested that Grade A and B represent an immature form of PVR with a definite potential for progression, whereas Grade C may represent a spontaneously arrested, non-evolutive form of the disease. However, other studies have confirmed that the presence of clinically significant primary PVR (greater than stage C1) is an independent risk factor for developing postoperative PVR [37, 58].

In general, preoperative PVR of any grade is associated with variable breakdown of the blood-ocular barrier and consequent generation and upregulation of chemical mediators associated with inflammation and wound healing. Despite intraoperative attempts to remove these epiretinal membranes, their growth is likely to continue to be stimulated, often resulting in postoperative PVR [18, 37, 50].

8.2.1.3 Aphakia

Chignell et al. were the first to associate aphakia with PVR, reporting that 34% of patients who developed postoperative PVR which resulted in failure of retinal reattachment surgery were aphakic [15]. Kon et al. in a prospective study classified the crystalline lens status into phakia and aphakia [37]. Pseudophakia with breached posterior capsule (e.g. following laser capsulotomy) was classified as aphakia while pseudophakia with intact capsule was classified as phakia. The authors identified aphakia to be an independent risk factor for PVR in patients undergoing primary vitrectomy for retinal detachment [37]. Other studies, however, did not find aphakia to be a risk factor [6].

Interestingly, in aphakic patients, other coexisting clinical factors have been found to be associated with higher risk of developing PVR and include choroidal detachment, duration of retinal detachment longer than 3 months, retinal breaks larger than three disc diameters and cataract surgery complicated by vitreous loss [48, 49].

Although the pathological mechanism by which aphakia could be related to the development of PVR remains unclear, functional or mechanical breakdown of the blood-ocular barrier may play a major role. It has been reported that the disruption to the blood-retinal barrier is more significant after intracapsular as opposed to extracapsular cataract extraction [47]. It seems that the intact posterior lens capsule provides a physical barrier moderating transmission of inflammatory cytokines from the anterior chamber into the vitreous cavity. The disruption of the blood-retinal barrier associated with aphakia may permit serum factors to invade the vitreous, resulting in induction of the PVR process.

8.2.1.4 Retinal Breaks

The type of retinal break not only may affect the clinical features of a rhegmatogenous retinal detachment but also influences its prognosis [44]. Proliferative vitreoretinopathy is more

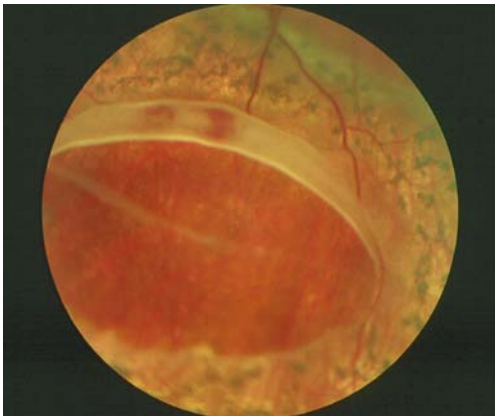

Fig. 8.2. Giant retinal tears with large exposed area of RPE carry a significant risk of developing PVR

likely to complicate retinal detachments caused by horseshoe- or crescent-shaped tears with evidence of vitreous traction. Conversely, PVR is not typically associated with retinal dialysis, round holes or macular holes [6, 44, 60]. The size and number of retinal breaks is also a significant determinant for the development of PVR. Giant retinal tears or large breaks with the exposed area of RPE more than three disc diameters have been found to carry a significant risk [6, 18, 25] (Fig. 8.2). Machemer suggested that the high incidence of PVR following surgery for giant retinal tears might be related to the large surface area of bare RPE exposed to the vitreous [43]. Those RPE cells could collapse the vitreous, induce preretinal membrane formation and exert tractional forces. Equally, in patients with unsealed retinal breaks after retinal reattachment surgery, viable RPE cells are exposed to the vitreous contributing to the development of PVR [18, 60].

Since the total surface area of bare RPE in retinal breaks appears to be a highly significant parameter, it has been postulated that early destruction of these cells with the aid of laser photocoagulation may reduce the risk of PVR. However, to date, there have been no clinical studies conducted to support this hypothesis.

8.2.1.5 Choroidal Detachment

Preoperative choroidal detachment in patients with rhegmatogenous retinal detachment is a rare occurrence and is usually associated with myopia and ocular hypotony. However, choroidal detachment appears to be significantly more common in patients who develop PVR. Cowley reported that choroidal detachment may be an independent risk factor for PVR since it is associated with the breakdown of blood-retinal barrier with subsequent stimulation of membrane formation and tractional retinal detachment [18].

Similarly, retinal detachment surgery which is complicated by postoperative choroidal detachment may also be associated with a higher risk of PVR. In a large retrospective study of 1,020 retinal detachments with no or mild preoperative PVR, Girard et al. identified that postoperative choroidal detachment is a significant predictive factor for postoperative PVR [25].

Although it is very difficult to alter the effect of preoperative choroidal detachment on the development of PVR, it has been suggested the use of steroids and vitreoretinal microsurgery may minimize the risk of postoperative PVR associated with choroidal detachment [2].

8.2.1.6 Duration and Size of Retinal Detachment

The duration of retinal detachment has also been reported to be a risk factor for the development of PVR [50, 60]. Proliferative vitreoretinopathy typically does not develop immediately following a stimulus. In eyes with longstanding retinal detachment the RPE cells are exposed to the vitreous body over a prolonged period. This prolonged exposure allows sufficient time for the RPE cells to migrate into the vitreous, transform and proliferate, leading to formation of contractile membranes.

A number of studies have reported a correlation between the size of retinal detachment and the severity of PVR. Girard et al. identified retinal detachment involving more than two quadrants to be a significant risk factor [25]. It has been suggested that extensive retinal detach-

ment may be associated with greater breakdown of the blood-ocular barrier which increases the risk of PVR. Moreover, the rupture of the blood-ocular barrier is further exacerbated by the requirement of extensive surgery that may be employed for the management of large retinal detachments [50].

8.2.1.7 Intraocular Inflammation

The development of contractile PVR membranes can be likened to the healing phase of the inflammatory response. There is much clinical, histological and experimental evidence that inflammatory cells including macrophages and lymphocytes play an important role in its pathogenesis.

Experimental models of PVR in which intravitreal injection of autologous cells has been performed have shown that death of the injected cells stimulates an inflammatory response that plays a part in subsequent cellular events. Breakdown of the blood-retinal barrier secondary to inflammation allows serum proteins to diffuse into the retina and vitreous cavity. Some of these, including platelet-derived growth factor and fibronectin, are involved in the development of PVR by their regulatory effects on RPE and glial cells [24].

Clinical studies have confirmed the above experimental results, identifying the presence of intraocular inflammation at initial examination of patients with retinal detachment as a significant risk factor for the development of PVR [18, 25].

8.2.1.8 Trauma

PVR is a common complication following mechanical ocular injuries. It occurs in approximately 4–19 % of cases and its incidence varies depending on the type and nature of the trauma [13]. Eyes with a history of perforating injury have the highest frequency of PVR followed by globe rupture, penetrating injury, intraocular foreign body and ocular contusion. The high incidence of PVR in these eyes may be due to the displacement, proliferation and migration of RPE cells during the initial ocular trauma resulting in membrane formation.

The strongest predictor of PVR in eyes which have suffered mechanical injury is the presence of vitreous haemorrhage. Moreover, the amount of blood seems to be an important determinant for the development of traction retinal detachment. Other independent risk factors of PVR following ocular trauma include extensive, posteriorly located wounds and persistent intraocular inflammation [13].

Despite surgical advances in the management of open globe injuries, PVR remains a devastating complication resulting in poor visual outcome. Eyes which develop PVR after trauma are more likely to suffer visual loss compared with those that do not.

8.2.2 Intraoperative Risk Factors

8.2.2.1 Cryopexy

Transcleral cryotherapy is a widely accepted technique of retinopexy in retinal reattachment surgery. It induces an inflammatory reaction, which results in development of strong adhesive forces and chorioretinal scarring between the retina and RPE layer.

Although there is vast clinical experience to support the safety and efficacy of cryotherapy, several clinical and laboratory studies have shown that cryotherapy may be implicated in the development of postoperative PVR [10, 12, 18, 26]. Cowley et al., in a retrospective review of 607 eyes with retinal detachment, the majority treated with scleral buckling, found that cryotherapy is an independent risk factor associated with PVR [18]. The stimulating effect of cryotherapy for postoperative PVR appears to be more prominent in eyes with evidence of preoperative PVR or large retinal breaks as opposed to cases where retinal detachment is associated with atrophic holes in lattice, oral dialyses, macular holes, or horseshoe tears with mobile posterior edges [8, 12].

Animal and human experimental models have shown that following application of cry-

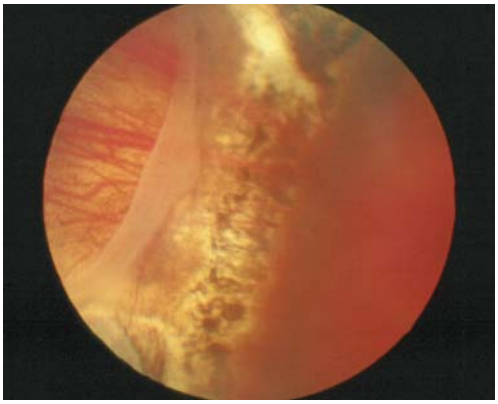

Fig. 8.3. In high risk cases of PVR, including eyes with preoperative PVR or eyes requiring extensive areas of chorioretinal adhesion, laser retinopexy should be considered

opexy an alteration in cellular adhesion of RPE cells to Bruch's membrane causes release of RPE cell clumps into the vitreous. These clumps contain viable RPE cells, indicating that the area of altered cellular adherence may be more extensive than the area of cell death; hence viable cells around the margin of an area of cryogenic necrosis are released [10, 26, 42]. Interestingly, diathermy does not release viable RPE cells probably because its coagulative effect on proteins is associated with more cell death and less disruption of cell adherence [10].

In addition to dispersion of RPE cells into the vitreous, both cryopexy and laser photocoagulation induce an inflammatory reaction which results in breakdown of blood-ocular barriers. This allows the release and upregulation into the vitreous of serum components including fibrin and fibronectin that enhance the ability of RPE cells to migrate into the vitreous cavity and accelerate a wound healing response leading to formation of PVR membranes [1, 10, 26, 31].

Despite its association with postoperative PVR, cryotherapy remains a valuable tool in vitreoretinal surgery and its cautious use should be encouraged. However, in high risk cases of PVR, including eyes with preoperative PVR or eyes requiring extensive areas of chorioretinal adhesion, alternative methods of retinopexy such as laser should be considered when appropriate (Fig. 8.3).

8.2.2.2 Internal Tamponade Agents

Air, gas and silicone oil are the mainstays of internal tamponade in vitreoretinal surgery. Although silicone oil has been widely used in complex vitreoretinal cases including retinal detachment complicated by PVR, experimental studies have shown that intravitreal silicone oil injection may stimulate membrane formation and subsequent tractional retinal detachment [39]. It has been suggested that silicone-filled vitreous induces increased mitogenic activity in RPE cells compared to gas- or fluid-filled eyes. Silicone oil may increase proliferative activity by acting either as a direct mitogen or by releasing more or different growth factors. Another possible mechanism is that it may concentrate active factors in a smaller aqueous volume between the silicone oil and the retinal surface [39].

Although some studies have reported low rates of PVR with intravitreal air, others have identified air tamponade as a risk factor for PVR [25]. In a retrospective multivariate analysis, Girard et al. identified the use of air tamponade to be associated with increased risk of PVR compared with other tamponading agents. PVR developed in 24% and 13% of eyes that had undergone intravitreal air injection with or without vitrectomy respectively. The authors postulated that especially in vitrectomized eyes, the short-lived tamponade effect provided by air is not sufficient to ensure retinal reattachment, leading potentially to PVR [25].

8.2.2.3 Surgical Technique

Pars plana vitrectomy is gaining increasing popularity in retinal reattachment surgery. Although initially reserved for complex vitreoretinal cases, many surgeons consider primary vitrectomy as the treatment modality of choice for retinal detachment repair. However, it has been suggested that the use of vitrectomy in the management of retinal detachment may predispose to postoperative PVR. Cowley reported that vitrectomy was the strongest risk factor for PVR independent of the indication for which it was performed [18].

Table 8.1. Clinical risk factors: the pathological mechanisms in the development of PVR

Clinical risk factors	Breakdown of blood-ocular barrier/serum components into the vitreous	Dispersion of RPE cells
Vitreous haemorrhage	+	–
Preoperative PVR	+	–
Aphakia	+	–
Large retinal breaks	–	+
Choroidal detachment	+	–
Long duration of retinal detachment	–	+
Extensive retinal detachment	+	–
Intraocular inflammation	+	–
Trauma	+	+
Silicone oil tamponade	+	–
Air tamponade	+	+
Cryopexy	+	+
Pars plana vitrectomy	+	–

Other experimental animal studies have also shown that mechanical stimulation, irrigation solution, fluctuation of the intraocular pressure and intraocular illumination may be responsible for the breakdown of both blood-aqueous and blood-retinal barrier that occurs during pars plana vitrectomy [55]. The breakdown of the blood-ocular barriers may stimulate the cellular processes implicated in PVR. In addition, removal of vitreous gel with or without induction of posterior vitreous detachment is often associated with the occurrence of iatrogenic retinal breaks. This results in release of viable RPE cells into the vitreous both during the development of the break and following retinopexy.

Although reports suggest that pars plana vitrectomy may be associated with PVR, it still remains an effective and irreplaceable technique in vitreoretinal surgery. Judicious use of vitrectomy in cases that cannot be managed by cryobuckling procedures is probably advisable in order to minimize the risk of postoperative PVR.

Pneumatic retinopexy has also been implicated in the development of PVR. During the procedure the expanding gas bubble injected into the vitreous causes the subretinal fluid to be squeezed through the open break. This results in disruption of the blood-ocular barrier, increase in vitreous flare, and migration and proliferation of RPE cells into the vitreous cavity potentially inducing PVR [29].

Summary for the Clinician

- **Knowledge of the numerous clinical risk factors (Table 8.1) implicated in the development of PVR has changed our management strategy for retinal reattachment surgery, for example the use of laser instead of cryotherapy**
- **Important clinical risk factors to consider include preoperative PVR, aphakia, preoperative inflammation, size of detachment, vitreous haemorrhage and the use of cryotherapy**
- **Clinical risk factors are also used to help us identify high risk patients so that they may be targeted for possible adjuvant therapy**

8.3 Biological Risk Factors

Proliferative vitreoretinopathy may develop as a response to a break in the integrity of the internal limiting membrane of the retina. It is a complex process which involves a series of biochemical and cellular interactions similar to those of the wound healing response with inflammation, migration, and proliferation of a variety of cells combined with secretion and remodelling of the extracellular matrix. Inflammatory cells such as monocytes, macrophages, neutrophils and lymphocytes play a critical role in these processes, providing cytokines, growth factors and important proteolytic enzymes which interact and mediate in the development of PVR.

8.3.1 Vitreous Protein

Total vitreous protein represents the sum of all detectable proteinaceous components in the vitreous. Although it is not specific for individual biological factors, it provides information on the state of inflammation, breakdown of the blood-ocular barrier and the severity of wound healing. Many studies have evaluated the association between vitreous protein levels and PVR in patients with retinal detachment. Connor et al. reported a fivefold increase of vitreous protein in patients with preoperative PVR as opposed to those without PVR, while Kauffman et al. found a threefold increase [16, 33]. Other studies have reported that for each milligram increase in the protein level the odds of developing PVR are increased by 1.10 times [37].

Overall, the level of vitreous protein in eyes complicated by retinal detachment appears to be a significant predictor for the development of PVR. Methods of measuring vitreous protein are advancing and the time taken to do so is shortening. It is now feasible to carry out intraoperative measurement of vitreous protein concentration, which can provide valuable information regarding the potential risk of PVR; decisions can therefore be taken on whether aggressive pharmacological treatment modalities should be employed.

8.3.2 Cytokines

Cytokines are polypeptides acting as chemical communicators between cells by binding to specific receptors on the surface of target cells. Moreover they regulate cellular migration, production of extracellular matrix and contraction. Most cytokines are growth and/or differentiation factors and although their exact role in the pathogenesis of PVR has not been fully elucidated, many studies have indicated that they act in opposition or synergistically on target cells to induce the various orderly sequences of events in wound healing. The important cytokines that have been implicated in the development of PVR are described below.

8.3.2.1 Transforming Growth Factor-β (TGF-β)

Transforming growth factor-β is a family of dimeric polypeptide cytokines which are found in large quantities in platelets and are secreted by a variety of cells including activated lymphocytes, macrophages, fibroblasts and RPE cells. Although there are three main isoforms of TGF-β (types 1–3), Pfeiffer et al. concluded in their study on monkey eyes that type 2 is the predominant isoform in the retina and vitreous [53].

TGF-β is an accelerator of wound healing and appears to be important in fibrotic processes such as PVR. Its effects in vivo are dependent on many factors including the type of the target cell and the presence of other growth factors. TGF-β upregulates the synthesis of fibronectin, increases the deposition of collagen and fibronectin and has chemotactic properties for monocytes and fibroblasts. In addition, it enhances the degree of collagen contraction by both fibroblasts and RPE cells [52].

Immunohistochemical studies by Baudouin et al. have detected TGF-β1 in cells from the vitreous and subretinal fluid of patients with PVR [5]. Other studies have reported increased vitreous levels of TGF-β2 in patients with retinal detachment complicated by primary or postoperative PVR compared with those who did not develop PVR [52]. TGF-β2 levels in the vitreous

increase with increasing severity of PVR, suggesting that inhibition of TGF-β2 activity may have the potential to control the severity of the PVR disease process.

8.3.2.2 Platelet-Derived Growth Factor

This monomeric polypeptide is released from platelets during thrombus formation but is also produced by other cells including monocytes, endothelial cells and fibroblasts [54]. Platelet-derived growth factor (PDGF) is a wound healing cytokine and acts as a chemoattractant as well as a potent mitogen for neutrophils, monocytes, fibroblasts and glial cells. Moreover, it stimulates the production and contraction of extracellular matrix.

Several studies have suggested an important role for PDGF in PVR. Campochiaro et al. have shown that PDGF is a potent stimulator of RPE migration when compared with other growth factors, while cultured RPE cells produce PDGF-like proteins which are capable of stimulating fibroblast chemotaxis and proliferation [11]. Using immunostaining techniques, PDGF receptors have been detected on the RPE cells within PVR membranes, unlike normal RPE cells, which have no evidence of these receptors. In addition, experimental animal models have shown that intravitreal injection of PDGF and fibronectin results in development of PVR and tractional retinal detachment [59].

8.3.2.3 Hepatocyte Growth Factor

Hepatocyte growth factor (HGF), also known as scatter factor, is a multipotential cytokine that can produce a range of responses in target cells. It is a heparin-binding glycoprotein, originally derived from platelets, which predominantly acts on epithelial cells, where it has morphogenic and mitogenic activities [9]. HGF interacts with a variety of extracellular matrix molecules. Pro-inflammatory cytokines including tumour necrosis factor (TNF) and interferon-gamma upregulate expression of HGF, while HGF itself upregulates expression of TGF-β.

Injury to the retina induces an inflammatory response that increases HGF expression inducing migration of multilayered groups of RPE cells. Immunohistochemical studies have shown that HGF induces RPE cells to acquire an elongated shape with a more fibroblastic appearance typical of cell morphology found in PVR [27, 30].

Increased expression of HGF has been reported in human PVR membranes and in the vitreous of PVR patients, and there is a tendency towards higher HGF levels with increasing severity of PVR [27, 30].

8.3.2.4 Interleukins

The interaction between immune and inflammatory cells is mediated by proteins, termed interleukins (IL), that are able to promote cell growth, differentiation, and functional activation. Among the different types of interleukins, it is only IL-1β, IL-6 and IL-8 that appear to be involved in the development of PVR.

Interleukin-1β belongs to a family of two functionally related proteins (the other being IL-1α) that are pro-inflammatory molecules. Their production occurs soon after injury or microbial invasion. IL-1β is also a stimulant for other cytokines including IL-6 and has been shown to be chemotactic to human RPE cells, which participate in the PVR process [34]. Previous studies have reported increased vitreous levels of both protein and mRNA encoding IL-1β in patients with PVR, indicating local production of the cytokine by vitreous cells and the significant contribution of the invading cells in PVR pathogenesis [22].

Interleukin-6 participates in the wound healing process by stimulating the proliferation of fibroblasts and glial cells and the synthesis of collagen. Several studies have reported elevated levels of IL-6 in vitreous specimens of patients with PVR [22, 36]. In addition, IL-6 mRNA-positive cells have been detected in PVR membranes, providing further evidence of their role in the development of PVR [41].

Interleukin-8 is a chemotatic factor and activator of neutrophils and lymphocytes. It has also been recognized as a potent agent causing vas-

cular permeability and angiogenesis [23]. IL-8 alone or in combination with other cytokines appears to play a role in the pathogenesis of PVR by promoting fibrocellular proliferation [23].

8.3.2.5 Fibroblast Growth Factors

Fibroblast growth factors (FGFs) exist in two closely related forms, acidic-FGF (a-FGF) and basic-FGF (b-FGF). They are mitogenic for fibroblasts, endothelial and RPE cells and have an influence on the synthesis and distribution of extracellular matrix. The physiological role of FGF in the retina includes neovascularization of ocular tissue and neuronal wound healing [45].

Malecaze et al., using immunohistochemical techniques, detected the presence of a-FGF in PVR membranes. Other studies have reported increased levels of both a-FGF and b-FGF in cells derived from vitreous samples and subretinal fluid of patients with PVR [14, 38].

8.3.2.6 Tumour Necrosis Factor Alpha

Tumour necrosis factor alpha (TNF-α) is a cytokine whose presence has been implicated in the development of PVR. It is known to be chemotactic for monocytes and fibroblasts, and it acts synergistically with interferon gamma and interleukin-1 in the induction of intercellular adhesion molecule-1 by RPE cells. TNF-α is often derived from activated macrophages although in PVR membranes, RPE and glial cells may also be a source. Generally TNF-α is a major regulator of RPE activation responses, including cell attachment, spreading, chemotaxis, and migration, which may contribute to the immunochemical process of PVR. Both mRNA and its proteins are widely expressed in the vitreous of patients with PVR and in PVR membranes [21, 32].

8.3.2.7 Matrix Metalloproteinases

The migration, proliferation, differentiation, adhesion and other biological functions of cells are influenced to a large extent by the surrounding extracellular matrix. The matrix metalloproteinases (MMPs) are a family of enzymes that degrade and remodel the extracellular matrix, playing a central role in wound healing. These molecules are produced by a variety of cells involved in the wound healing process, whilst their activities are regulated by the tissue inhibitors of matrix metalloproteinases (TIMMPs).

Immunohistochemical studies have detected MMP-2, MMP-3 and TIMP-1 in epiretinal and subretinal membranes of PVR but not in normal retina, indicating that these enzymes may play an important role during the development of PVR following rhegmatogenous retinal detachment [57]. Other prospective studies have demonstrated a significant association between vitreous levels of MMP-2 and MMP-9 with the development of postoperative PVR, suggesting that MMPs may be important predictors of PVR [35].

8.3.2.8 Adhesion Molecules

Cellular mechanisms of adhesion and migration are involved in the pathogenesis of PVR. Both macrophages and lymphocytes are present in fibrocellular membranes surgically removed from patients with PVR. The recruitment of leucocytes to sites of inflammation requires their adhesion to vascular endothelium and their subsequent migration into surrounding tissue. This complex process is mediated through multiple adhesion molecules present on the surface of leucocytes and endothelial cells.

Two important groups of adhesion molecules implicated in the development of PVR are the intercellular adhesion molecule-1 (ICAM-1) and the vascular cellular adhesion molecule-1 (VCAM-1). Soluble forms of these molecules have been found to be increased in the vitreous of patients with retinal detachment complicated by PVR [4]. Limb et al. reported that soluble ICAM-1 was significantly higher in vitreous samples from eyes with retinal detachment and at high risk of developing PVR as opposed to those from eyes with retinal detachment at low risk of developing this complication [40].

Overall high vitreous levels of ICAM-1 and VCAM-1 indicate a high degree of inflammation and a high risk of occurrence of PVR.

Summary for the Clinician

- PVR is a complex pathological process comprising inflammatory and immune responses as part of the wound healing event
- Cytokines, growth factors and enzymes provide communication between participating cells and regulate the wound healing response
- Increased levels of cytokines and enzymes that have been investigated and thought to be important in the formation of PVR include TGF, PDGF, HGF, interleukins, FGF, TNF, and matrix metalloproteinases
- Total vitreous protein represents the sum of all proteinaceous components present in the vitreous, including cytokines. Total vitreous protein is easy to measure intraoperatively and although non-specific, may be used to indicate the general level of cytokine activity and predict the likelihood of PVR formation
- In the future, better and quicker methods of detecting individual vitreous cytokines may improve our predictive ability

8.4 Prediction of Risk for PVR

Clinical and biological risk factors have consistently been reported in the literature to be predictive for the development of PVR. Recent research has focused on the prevention of PVR by the introduction of adjuvant agents such as steroids, heparin, 5-fluorouracil and daunorubicin [2]. These drugs although effective in reducing the risk of PVR may potentially be toxic to ocular tissues. Hence, identification of eyes at greatest risk of developing PVR and targeting treatment to these cases would increase the benefit:risk ratio of these treatments.

Recent studies have analysed several risk factors and developed formulas which predict the probability of an individual developing PVR [3, 37]. Asaria et al. constructed a risk predicting formula and applied it to 219 patients undergoing vitrectomy for retinal detachment [3].

The equation used to calculate the risk of postoperative PVR was:

PVR score:
= 2.88×(Grade C PVR)+1.85×(grade B PVR)
+2.92×(aphakia)
+ 1.77×(anterior uveitis)+1.23×(quadrants of detachment)
+ 0.83x(vitreous haemorrhage)+23×(previous cryotherapy)

One or zero is added to the equation if the risk factor is present or absent respectively. For quadrants detached, a value of 1–4 should be added. If the PVR score is greater than 6.33, then the patient is at high risk.

The efficacy of this formula in predicting PVR was found to be good. Only 9.2% of patients in the low risk group as opposed to 28% in the high risk group developed postoperative PVR. However, further studies are required to confirm these results as well as to calculate the risk of PVR following buckling procedures. The identification of high risk patients using readily accessible clinical tools such as the formula described above is of vital importance as this group of subjects can be targeted for aggressive treatment, which may moderate the risk of postoperative PVR.

Summary for the Clinician

- A useful tool for the clinician to identify high risk patients for adjuvant therapy:
  PVR score:
  = 2.88×(Grade C PVR)+1.85×(grade B PVR)
  +2.92×(aphakia)
  + 1.77×(anterior uveitis)+1.23×(quadrants of detachment)
  + 0.83x(vitreous haemorrhage)+23×(previous cryotherapy)

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Prevention of Proliferative Vitreoretinopathy

David G. Charteris

Core Messages

- Proliferative vitreoretinopathy (PVR) remains an important cause of surgical failure and visual loss following retinal detachment
- Analysis of the pathobiology of PVR provides the basis to design strategies for preventing PVR development or recurrence
- Retinal detachment cases at high risk of PVR can be identified on the basis of their clinical features
- Selection of appropriate surgical techniques for retinal detachment repair can reduce the incidence of PVR and improve surgical outcomes
- A peroperative infusion of 5-fluorouracil and low-molecular-weight heparin reduces PVR development in high-risk retinal detachments
- Peroperative daunomycin infusion reduces reoperations in established PVR

9.1 Introduction

9.1.1 Proliferative Vitreoretinopathy

Proliferative vitreoretinopathy (PVR) is a process of cellular proliferation, extracellular matrix deposition and membrane formation and contraction which occurs as a complication of retinal detachment [14, 44]. The accepted standardised descriptions and grading of PVR refer to the clinical condition which follows rhegmatogenous retinal detachment [40, 53]; however, variants of PVR also occur after ocular trauma and other intraocular pathologies.

9.1.2 Incidence/Clinical Relevance

Recent reports on success rates in primary rhegmatogenous retinal detachment document the incidence of PVR at between 5.1 % and 11.7 % [8, 20, 25, 27, 30, 31, 51]. Where there is a final failure of retinal reattachment surgery this is due to PVR in over 75 % of cases [17, 46, 53].

In other vitreoretinal conditions the incidence of PVR is higher, for example following giant retinal tears PVR has been reported to occur in between 16 % and 41 % of eyes [12, 13, 35, 37, 56]. In eyes sustaining penetrating ocular trauma PVR has been documented in between 10 % and 45 % of cases [10, 18, 23, 26, 29, 34, 41–43, 50, 52, 62], the incidence being higher in perforating (through and through) and blunt injuries and lower in cases of intraocular foreign body [10]. PVR has also recently been shown to be an

important complication of retinal translocation surgery for age-related macular degeneration occurring in between 10 % and 18 % of eyes [21, 24, 45].

Success rates for vitreoretinal surgery on eyes which have developed PVR have improved markedly as vitrectomy techniques have evolved. Lewis, Aaberg and Abrams have reported final posterior reattachment rates of 90 % for initial PVR surgery and 86 % for repeat surgery [38, 39]. In the Silicone Study final posterior reattachment was achieved in 77 % of eyes managed with silicone oil and 79 % of those managed with C3F8 gas [54, 55]. Despite these high anatomical success rates visual results are frequently poor after PVR surgery. In the Lewis, Aaberg and Abrams series [38, 39], of the eyes with full posterior reattachment, after initial PVR surgery 19 % and after repeat surgery 11 % attained 20/100 vision. In the Silicone Study, overall 25 % attained 10/100 vision [54, 55]. Similarly binocular visual outcome is often unsatisfactory – it has been reported that following PVR surgery over 50 % of patients describe their visual comfort as medium to bad [1].

It is, however, notable that where a patient has undergone surgery for PVR the fellow eye may also have poor vision. Schwartz and Kreiger [49] reported vision-threatening pathology in over 50 % of fellow eyes (75 % of these had retinal tears or detachment). Of the fellow eyes 47 % had final vision of 20/50 or worse and half of these had visual acuity of 20/250 or less.

Given the continued incidence of PVR in rhegmatogenous retinal detachment, its higher incidence in other vitreoretinal conditions, the unsatisfactory visual results of surgery (despite reasonably good anatomical success rates) and the uncertain status of the fellow eye, there is a clear need for adjunctive strategies to prevent the initial development and the recurrence of PVR. Such strategies involve both surgical considerations and potentially the use of pharmacological or biologically active treatments.

9.2 PVR Pathobiology

Extensive laboratory research over the last 30 years has provided data on the pathobiology of PVR. A review of this data provides a basis for the rational design of adjunctive therapeutic strategies for the prevention and management of PVR.

9.2.1 PVR Evolution [14, 44]

Rhegmatogenous retinal detachment is conventionally viewed as the starting point for PVR development. A retinal tear, in addition to allowing egress of vitreous cavity fluid into the subretinal space, results in the dispersion of a variable quantity of retinal pigment epithelial (RPE) cells into the vitreous cavity, where they can play a role (with other cell types) in epiretinal membrane formation. The blood-retinal barrier (BRB) breakdown which follows retinal detachment appears to have a central role in the dispersion of cells and growth factors which promote the further evolution of PVR. Situations where there is a greater degree of BRB breakdown such as extensive vitreous haemorrhage and pre-existing uveitis have a higher incidence of PVR. Subsequent cellular proliferation and extracellular matrix elaboration results in the formation of fibrocellular epiretinal and subretinal membranes. Cellular proliferation in the vitreous base is an additional factor causing vitreoretinal traction. These contractile membranes produce the characteristic clinical features of established PVR [40, 53].

9.2.2 Cellular Involvement

Analysis of PVR epiretinal and subretinal membranes together with animal models has generated data on the cell types found in PVR [14, 44]. These are generally considered to be of four types: RPE cells, glial cells, inflammatory cells and cells of fibroblastic morphology.

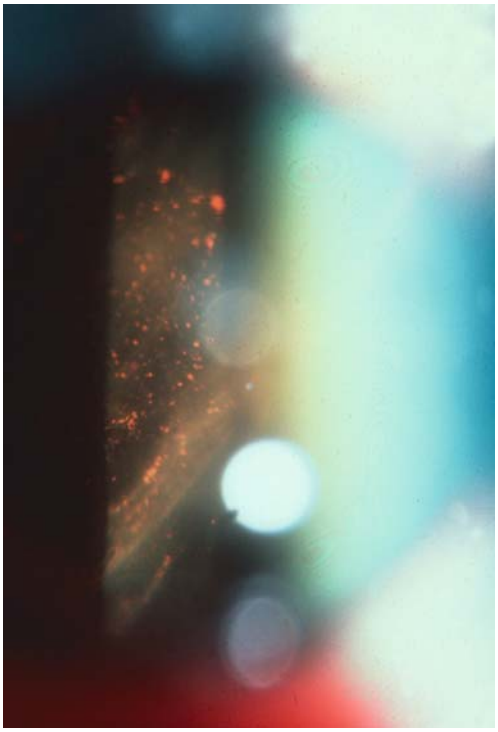

Fig. 9.1. RPE cell clumps in the anterior vitreous of a patient with retinal detachment and early PVR

RPE cells are released into the subretinal space and vitreous cavity (Fig. 9.1) at the time of a retinal tear and retinal detachment and are a consistent finding in PVR membranes. It is notable that both clinical [28] and experimental [9] studies have demonstrated that trans-scleral cryotherapy applied to eyes with retinal detachment enhances the dispersion of viable RPE cells into the vitreous cavity.

Glial cells have been shown to be present in PVR membranes and may be derived from Muller's cells, astrocytes, microglia or perivascular glia within the retina. Recent work has demonstrated glial cells extending through the inner limiting membrane from the retina to overlying epiretinal membranes [16], suggesting that they may impede separation of membrane from retina during surgical peeling.

Macrophages and T lymphocytes have been identified in PVR membranes [14] and, although relatively small in number, they may play an important role in membrane development and contraction through growth factor production. Cells of fibroblastic morphology are found in most studies of PVR membranes and may be derived from RPE cells, vascular epithelial cells, glia or hyalocytes. Such fibroblastic cells may contain myofibrils and could be responsible for the contraction seen in PVR membranes.

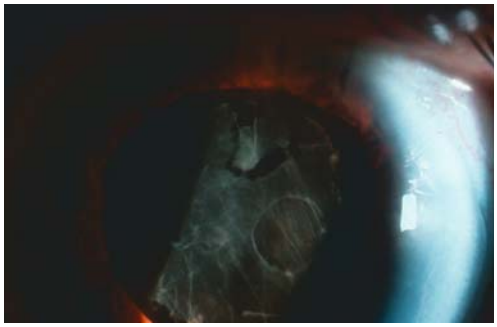

Fig. 9.2. Fibrin sheet on the anterior face of silicone oil endotamponade in an aphakic eye which has undergone surgery for PVR

9.2.3 Fibrin

Eyes which have developed PVR have a greater tendency to develop marked intraocular fibrin formation after vitrectomy surgery [32, 33] (Fig. 9.2). It has been proposed that the formation of fibrin in the early phase of blood-retinal barrier breakdown following retinal detachment or after vitreoretinal surgery could potentially form a scaffold for the subsequent development of the complex fibrocellular membranes seen in PVR. This is supported by the experimental observation that fibrin contact causes RPE cells to de-differentiate and migrate into a fibrin clot to form sheets of fibrocyte-like cells [57].

9.2.4 Extracellular Matrix

PVR periretinal membranes have been demonstrated to contain various extracellular membrane components [14]. Collagen types I, II, III and IV have been shown to be present (RPE or

glial cells may be the source of these) as have the cell attachment protein fibronectin and the basal lamina proteins heparan sulphate and laminin.

Turnover and remodelling of extracellular matrix is regulated by a group of proteolytic enzymes known as matrix metalloproteinases (MMPs) and their natural inhibitors: tissue inhibitors of metalloproteinases (TIMPs). MMPs 1, 2, 3 and 9 and TIMPs 1, 2 and 3 have been demonstrated to be present in PVR membranes [47] and have the potential to play an important role in membrane formation.

9.2.5 Growth Factors

Growth factors/cytokines can mediate the processes of cellular activation and contractile membrane formation outlined above. Various growth factors have been shown to be present in PVR epiretinal membranes and in the vitreous of eyes with PVR [11, 14, 15, 22], and experimental work has demonstrated that these growth factors have in vitro activity in RPE chemotaxis and proliferation and glial cell chemotaxis [14]. Some of these growth factors, for example platelet derived growth factor (PDGF), fibroblast growth factors and transforming growth factor β, are known to have fibrogenic activity and are likely to be involved in upregulating the fibrocellular scarring response seen in PVR. Blocking the action of these growth factors has the potential to decrease membrane formation in PVR.

9.2.6 Targets of Pharmacological Adjuncts

The processes of cellular activation, proliferation and contractile membrane formation outlined above and the cells, growth factors and extracellular matrix involved in these are potential targets for pharmacological adjuncts to the surgical treatment of PVR. Adjunctive strategies are outlined in Table 9.1.

Summary for the Clinician

- **Recent reports on success rates in primary rhegmatogenous retinal detachment document the incidence of PVR at between 5.1 % and 11.7 %. In other vitreoretinal conditions the incidence of PVR is higher. Despite high anatomical success rates visual results are frequently poor after PVR surgery. Situations where there is a greater degree of BRB breakdown such as extensive vitreous haemorrhage and pre-existing uveitis have a higher incidence of PVR. In PVR there are generally considered to be four types of cells: RPE cells, glial cells, inflammatory cells and cells of fibroblastic morphology. Eyes which have developed PVR have a greater tendency to develop marked intraocular fibrin formation after vitrectomy surgery. PVR periretinal membranes have been demonstrated to contain various extracellular membrane components and various growth factors.**

Table 9.1. Targets for adjunctive treatment

Pathological process	Strategy
Blood-retinal barrier breakdown	Anti-inflammatory treatment
Cellular activation	Antiproliferatives Growth factor manipulation
Cellular proliferation	Antiproliferatives: (a) cell-specific, (b) non-specific
Fibrin formation	Decrease production/increase breakdown
Extracellular matrix formation	Inhibition of cellular activation MMP/TIMP manipulation
Membrane contraction	Contraction inhibition

9.3 Surgical Considerations

There is an ongoing debate as to whether PVR arises solely as a consequence of inadequate primary retinal detachment surgery and subsequent redetachment or whether it may occur de novo in a predisposed eye with otherwise adequate primary surgery. Regardless of the answer to this issue (and it would appear likely that both theories have relevance in PVR pathogenesis), there are steps in the perioperative management of individual patients that surgeons can take to minimise the risk of PVR development.

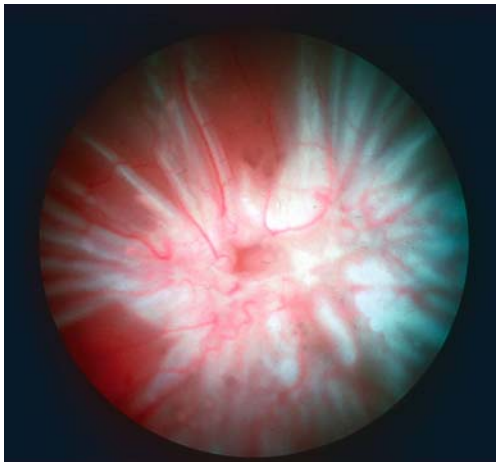

Fig. 9.3. Advanced PVR (closed funnel retinal detachment)

9.3.1 Preoperative Management

9.3.1.1 Patient Selection

Recognition of early PVR is of central importance to the management of primary retinal detachment since regression analyses have demonstrated preoperative PVR to be an important determinant of subsequent postoperative PVR development [3, 7, 19, 36]. Both surgical technique and the use of adjunctive medication may alter considerably if PVR is present before surgery. Whilst the features of more advanced PVR (star folds, irregular full-thickness folds, circumferential contraction [40], Fig. 9.3) are easily recognised, the features of less advanced, grades A or B, PVR such as vitreous haze and pigment clumps (Fig. 9.1), wrinkling and stiffness of the retina and rolled edges of retinal breaks may be more subtle. It is important that clinicians are alert to the presence of these signs since assessing the longevity of a retinal detachment from the history alone may be misleading and certain patients may be unaware of, or may minimise, the duration of their symptoms.

Preoperative assessment of the risk of PVR development plays an important role both in selection of surgical technique and in the use of adjunctive medications. The use of a formula derived from regression analysis has been shown to have predictive value for PVR development [3, 36]. This formula states:

Risk of PVR developing =
2.88×(preoperative grade C PVR)+1.85×(preoverative grade B PVR)
+2.92×(Aphakia or pseudophakia without intact posterior capsule)
+1.77×(Anterior uveitis)+1.23×(Quadrants of detachment)
+0.83×(vitreous haemorrhage)+1.23×(previous cryotherapy)

If a risk factor is present, 1 is added to the equation; for quadrants of detachment 1–4 is added. Using a discriminant rule a total risk factor score of greater than 6.33 defines a retinal detachment as high risk and less than 6.33 as low risk. High-risk cases have been shown to have a postoperative incidence of PVR of 28 % and low-risk cases of 9 % [3].

Clinical experience suggests that other features of retinal detachments and/or coexisting pathology may also be important risk factors for PVR. Vitreous haemorrhage, particularly if dense and fundus obscuring, appears to increase the risk considerably [48]. This may be due to increased presence of fibrogenic growth factors in the vitreous cavity. Eyes with active

uveitis and choroidal detachment (both of which have increased BRB breakdown) are generally considered to be at higher risk, as are eyes with prior trauma which may have both increased BRB breakdown and vitreous haemorrhage. Children and young adults, possibly due to a more aggressive wound healing response, also appear to be at high risk of postoperative PVR development. The above presenting features should be carefully assessed when determining the best surgical management strategy for retinal detachment cases.

A further preoperative consideration is the judgement of a patient's ability to comply with postoperative posturing instructions. Children and patients with coexisting disabling systemic disease may find prolonged posturing extremely difficult. This may determine the choice of tamponading agent used – silicone oil in general will require less stringent posturing regimes.

9.3.1.2 Preoperative Treatment

In certain situations medical treatment before vitreoretinal surgery is indicated. In selected cases this may involve deferring the planned surgical intervention. In eyes with active uveitis it is recommended that the inflammation is controlled prior to surgery. Frequent topical steroid drops are given together with systemic steroids (optimally prednisolone 40 mg/day for 2 weeks in adults) before surgery.

Based on preoperative findings, for example marked blood-retinal barrier breakdown, choroidal detachment or post-trauma uveitis, clinicians may decide to treat selected eyes medically with steroid or other anti-inflammatory medication prior to surgery. Given the known involvement of inflammatory cells in PVR development (see Sect. 2.2), minimizing preoperative inflammation is an important step in PVR prevention.

9.3.2 Peroperative Surgical Management

9.3.2.1 Retinal Detachment Without PVR

It is clear that the most important consideration in preventing PVR is successfully reattaching the retina at the first surgical intervention. There are, however, other factors which may be of importance in minimising postoperative proliferation. Cryotherapy is known to release viable RPE cells into the vitreous cavity [9, 28] (see Sect. 2.2) and it would therefore appear justified to minimise the amount of cryotherapy performed and to use laser retinopexy where possible, particularly where extensive retinopexy is necessary.

When vitrectomy is performed as part of the initial retinal detachment repair, the choice of tamponading agent can influence primary success. Children and some adults will find it difficult to follow posturing instructions and silicone oil is a more suitable agent where individuals are unable to posture and/or prolonged tamponade is required.

9.3.2.2 Retinal Detachment at High Risk of PVR/with Early PVR

Adequate primary surgical repair of retinal detachments at high risk or with early PVR is critical to the ultimate outcome of such eyes. Knowledge of the pathogenesis of PVR and the documented clinical risk factors helps define the surgical techniques necessary. Vitrectomy will often be used to manage these cases. It removes vitreous gel containing blood and activated RPE cells and reduces the potential scaffold for subsequent contractile membrane formation. It is important in this situation to clear vitreous blood adequately (a known risk factor) and adequately trim the vitreous base. When the surgeon is concerned that there is residual vitreous base traction or that there is a high risk of postoperative basal gel contraction and secondary traction, then an encircling buckle can be employed to counteract this. The

tendency to use encircling buckles and the threshold of perceived residual vitreous traction varies between individual surgeons.

9.3.2.3 Established PVR

In eyes with recurrent retinal detachment and established PVR, in addition to reattaching the retina, the surgical aims include the prevention or reduction of re-proliferation. This necessitates adequate vitreous clearance with particular attention to the basal vitreous which can form the focus for anterior PVR. To achieve adequate anterior vitreous dissection it may be necessary to perform a lensectomy (or phakoemulsification) in selected eyes. It should be noted, however, that the binocular visual results in eyes with PVR which are aphakic following surgery are often very poor [1] and secondary lens implantation may be required.

The results of the Silicone Study [54, 55] demonstrated silicone oil and C3F8 gas to be equally effective and better than SF6 gas in the management of established PVR. While individual surgeons may favour either oil or gas tamponade, the lower level of complications seen with modern highly purified silicone oil makes it a satisfactory tamponade agent for PVR cases.

In more advanced PVR membrane peeling may be extremely difficult particularly of PVR membranes anterior to the equator where there can be continuity of glial cells between retina and membrane [16]. Subsequently there may be residual anterior traction. Scleral buckling is often inadequate in counteracting such traction. In this situation retinotomy and anterior retinectomy are necessary to relieve anterior traction and also to "de-bulk" residual PVR tissue which might provide a nidus for reproliferation.

9.3.3 Postoperative Care

Prevention of re-proliferation and membrane recurrence involves careful attention to the short-term anatomical results of surgery and the management of any postoperative complications. The importance of positioning in providing adequate intraocular tamponade should be emphasised to the patient. Any evidence of early re-detachment should be dealt with promptly. This can involve additional laser retinopexy, altering positioning instructions or augmenting intraocular tamponading agents. In certain situations, where the initial surgery appears to have failed in reattaching the retina, early reoperation is necessary.

Postoperatively eyes with PVR have a greater tendency for inflammation (and fibrin formation) and this in turn may increase the likelihood of re-proliferation. Adequate anti-inflammatory treatment, both topical and if necessary systemic, should be administered to prevent postoperative inflammation in eyes which have undergone surgery for PVR.

Summary for the Clinician

- **Preoperative PVR is an important determinant of subsequent postoperative PVR development. Preoperative features of less advanced, grades A or B, such as vitreous haze and pigment clumps, wrinkling and stiffness of the retina and rolled edges of retinal breaks, are less easily recognised. High-risk cases have been shown to have a postoperative incidence of PVR of 28 % and low-risk cases of 9 %. Vitreous haemorrhage, particularly if dense and fundus obscuring, appears to increase the risk considerably. Children and young adults also appear to be at high risk of postoperative PVR. In eyes with active uveitis it is recommended that the inflammation is controlled prior to surgery. The most important consideration in preventing PVR is successfully reattaching the retina at the first surgical intervention. The tendency to use encircling buckles and the threshold of perceived residual vitreous traction vary between individual surgeons. While individual surgeons may favour either oil or gas tamponade the lower level of complications seen with modern highly purified silicone oil makes it a satisfactory tamponade agent for PVR cases. Retinotomy and anterior retinectomy may be necessary to relieve anterior traction and also to "de-bulk" residual PVR tissue which might provide a nidus for reproliferation.**

9.4 Adjunctive Treatment

The use of various adjunctive agents to prevent PVR and the recurrence of PVR have been investigated in both laboratory and clinical studies [14].

9.4.1 Initial Clinical Studies

Initial non-randomised clinical studies on the antiproliferative agent 5-fluorouracil (5FU) demonstrated that this agent appeared to be non-toxic in human eyes undergoing vitrectomy [5, 6] although these studies were too limited to demonstrate any therapeutic effect. The antiproliferative daunomycin was also shown to be non-toxic in non-randomised clinical studies [58, 59] on PVR and post-trauma PVR. The lack of toxicity of these agents and the perceived surgical success rates encouraged the development of further randomised clinical investigations.

9.4.2 Randomised Controlled Clinical Trials

The effect of postoperative irradiation in preventing recurrent PVR in eyes with established PVR was investigated in a small-scale randomised controlled trial [4]. A total dose of 3,000 cGy of irradiation was given in divided doses. No benefit in outcome was seen although the number of patients involved in the trial was small.

A prospective, randomised pilot study investigated the potential of combined heparin and dexamethasone in vitrectomy infusion fluid to reduce reproliferation in established PVR [61]. The treatment group had a higher anatomical success rate and a lower incidence of reproliferation; however, the number of patients involved in the study was relatively small and the result did not reach statistical significance. Larger scale randomised controlled trials have been conducted on the intraoperative use of daunomycin and a combination of 5FU and low-molecular weight heparin (LMWH).

9.4.3 Daunomycin

The efficacy of the antiproliferative agent daunomycin in established PVR has been investigated in a prospective, randomised, controlled clinical trial [60]. Patients with PVR grade C2 or greater undergoing vitrectomy and silicone oil exchange were randomised to treatment with or without a 10-min intraoperative infusion of daunomycin (7.5 µg/ml). The primary outcome measure, retinal attachment without additional vitreoretinal surgery at 6 months, showed a trend towards a benefit in the treatment group which marginally failed to reach significance (P=0.07). The trial did, however, demonstrate a statistically significant reduction in the number of vitreoretinal reoperations within 1 year (P=0.005), thus demonstrating that PVR was amenable to pharmacological treatment.

9.4.4 5-Fluorouracil/Low-Molecular Weight Heparin

The antiproliferative agent 5FU is known to be active against RPE cells and has been shown to reduce experimental tractional retinal detachment [14]. Low-molecular-weight heparin (LMWH) reduces post-vitrectomy fibrin, inhibits RPE proliferation, binds fibrogenic growth factors and reduces experimental tractional retinal detachment. Theoretically LMWH has less potential for haematological side effects than heparin. The combination of these adjunctive agents has been investigated in a randomised controlled clinical trial of patients undergoing vitrectomy surgery for retinal detachment who were determined to be at high risk of postoperative PVR [2]. Patients were selected as high risk on the basis of a previous prospective analysis of known risk factors for PVR [3, 36]. The adjunctive medications were infused for 1 h during vitrectomy at concentrations of 200 µg/ml (5FU) and 5 IU/ml (LMWH) in Hartmann's solution. The primary outcome measure, development of postoperative PVR, was significantly improved in the treatment

group (12.6% vs. 26.4%, P=0.02). Secondary outcome measures did not show significant differences between treatment and control groups and there were no differences in complication rates. On the basis of this study this treatment regime can be considered for use in patients at high risk of PVR. A further prospective clinical trial using the same adjunctive combination on eyes with established PVR did not show a treatment benefit (D.G. Charteris, American Academy of Ophthalmology, 2001).

Summary for the Clinician

- **The use of adjunctive medications should be considered in cases of established PVR or retinal detachments at high risk of PVR. High-risk cases can be identified on the basis of a formula derived from analysis of known risk factors [3, 36]. Alternatively surgeons may use their individual clinical experience to select cases they consider to be at high risk, for example traumatic retinal detachments or giant retinal tears. On the basis of the randomised clinical trial described above, the combination of 5FU and LMWH, given as a peroperative infusion, can reduce the incidence of postoperative PVR in high-risk cases. In established PVR a 10-min intraoperative infusion of daunomycin (7.5 μg/ml) may be used on the basis of the evidence that it can reduce the incidence of reoperations.**

9.5 Future Directions

Advances in preventing PVR in the future will require further clinical analysis of risk factors to determine which cases are suitable for adjunctive medication. Additionally randomised clinical trials are necessary to study the effectiveness of the range of potential adjunctive medications available in various clinical situations including high-risk retinal detachments and ocular trauma. Further development of adjunctive medications aimed at producing more targeted treatments (potentially by gene transfer) and more sustained delivery offer the potential for more effective PVR prevention.

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The Tamponade Effect

10

David Wong, Rachel Williams

Core Messages

- The tamponade effect involves three phases in contact with one another: namely, the retina, the aqueous and the tamponade agent
- The interfacial tension acts to minimise the surface area of a bubble of tamponade agent, which tends not to go through a retinal break in the detached retina; to do so would require an increase in surface area and therefore higher surface energy. In this way tamponade agents act as a "plug"
- When the retina is attached, a tamponade agent needs to exclude the access of aqueous from the break
- Specific gravity is the chief determinant of the shape of a bubble of tamponade in the eye. Bubbles that approximate a spherical cap in shape are more efficient at excluding aqueous from retinal breaks
- It is difficult if not impossible to achieve a total tamponade effect. This is not due to lack of tamponade efficiency, but rather to the fact that the eye is a near spherical cavity
- There will always be a slight underfill which leaves a large area of retina not supported
- If we believe bubbles that float are more efficient at supporting pathology in the upper part of the fundus, then it is logical to think that tamponade agents that sink are better at treating the lower retina
- Debate is ongoing as to whether it is desirable to exclude aqueous from the retina or indeed whether this is the cause of retinal toxicity
- We contend that agents with low viscosity and high specific gravity are not suitable as long-term tamponade agents. Solutions of semifluorinated alkanes and silicone oil can have a high viscosity and specific gravity just greater than 1 and may prove to be the long-awaited heavier-than-water tamponade

10.1 Introduction

This chapter concentrates on the physical properties of liquid agents used in the eye and explains how they produce the tamponade effect. We illustrate certain principles using air and silicone oil; we also cover semi-fluorinated alkanes especially in the context of designing new and heavier-than-water tamponade agents.

The successful repair of rhegmatogenous retinal detachments depends crucially on the identification and closure of retinal breaks. There are only two manoeuvres which lead directly to break closure:

- Scleral buckling (sometimes referred to as external tamponade)
- Insertion of gas or liquid agents in the vitreous cavity (internal tamponade)

The two manoeuvres are sometimes used in combination; the D-ACE surgical sequence is used for the treatment of bullous superior retinal detachments [11]. After external drainage of subretinal fluid, air is injected and it temporari-

ly approximates the retinal tear to the underlying retinal pigment epithelium; this facilitates accurate localisation of the retinal tears and minimises cryotherapy. The air in this operation is acting as an intraoperative tool as well as an internal tamponade. As traction is relieved by the scleral buckling, postoperative posturing is not needed.

In all other situations, the tamponade agent needs to be in contact with the retinal breaks in order for there to be a "tamponade effect". What exactly is the tamponade effect and how do we envisage tamponade agents "closing" retinal breaks? We may ask: do tamponade agents

1. Act as plugs?
2. Act by excluding access of aqueous to retinal break?
3. Act by pushing against the retina?
4. Act by totally filling the vitreous cavity thereby obliterating the space into which the retina can be detached?

To answer these questions, it is necessary to understand some of the physical properties of tamponade agents.

10.2 Interfacial Energies

When two immiscible fluids come into contact with one another an interface is formed. One of the immiscible fluids could for example be air and the other water. Figure 10.1 illustrates a droplet of water at the tip of a pipette. A molecule "I" at the centre of the droplet is attracted in all directions to every other molecule inside the droplet, whereas a molecule "S" at the surface has a net attraction inwards. Surface tension is the van de Waal's attractive force between the molecules on the surface of the droplet. This force will tend to minimise the area of the surface.

Surface tension generally refers to energy between a liquid and air. A more general term is "interfacial energy" and this can be applied to situations when two or more phases are in contact with each other.

The interfacial energy of a system defines how the materials in contact will interact with

Fig. 10.1. A droplet of water at the end of a pipette. *I* represents a molecule inside the water droplet and *S* a molecule on the surface

each other so as to reduce the overall energy at the interface. For example, if a water droplet is placed on a hydrophobic material in air (Fig. 10.2), the water droplet remains rounded because it is more energetically favourable for the air to contact the hydrophobic material than the water. In contrast, if a water droplet is placed on a hydrophilic material in air the droplet spreads because it is more energetically favourable for the water to contact the surface than the air (Fig. 10.2). The amount of spreading of the water droplet in these conditions will de-

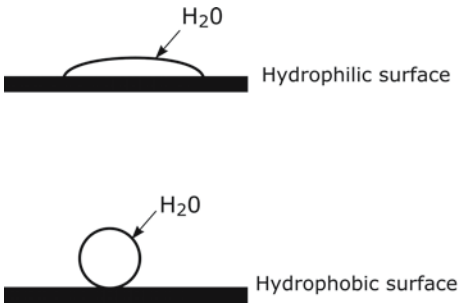


Fig. 10.2. A droplet of water on a hydrophilic and on a hydrophobic surface

pend on the interfacial energies of the three phases, in this case the material, the air and the water. This property is very important in determining the behaviour of tamponade agents in the vitreous cavity. In this situation the three phases to consider are the retina, any remaining aqueous and the tamponade agent. The retina, as with most tissue, is highly hydrated and therefore hydrophilic; any remaining aqueous will therefore spread on the retina.

When a tamponade agent comes into contact with a retinal break in a detached retina, the agent tends not to go through the break. The interfacial energy acts to minimise the surface area. For the tamponade agent to go through the retinal break, it needs to increase in surface area, which requires higher energy. In this sense, a tamponade agent acts as a "stopper" and literally "plugs" a retinal hole. If subretinal fluid is evacuated at the same time as the tamponade is injected, the retina will become reapposed to the underlying retinal pigment epithelium.

Care, however, must be exercised when carrying out this fluid exchange procedure in the presence of a detached retina stiffened by epiretinal membrane. The interfacial energy can be overcome by the force of injection. For example, if silicone or air is injected as subretinal fluid is evacuated, there is a pressure gradient across the retina. Instead of the retina being reapposed, the silicone oil can pass through the retinal break and gain access to the subretinal space. This then becomes an awkward complication to handle [24]. Some surgeons therefore prefer to use an air/fluid exchange. Air has a higher interfacial energy against water than silicone and is therefore less likely to go through a retinal break in detached retina. Once that a retina is reapposed, there is no longer a pressure gradient across the retinal break and silicone oil can be injected with no fear of it getting into the subretinal space.

When the retina is reattached, the role of the tamponade agent is to maintain this apposition. To achieve this, all that is required is for the tamponade agent to exclude aqueous from the retinal break. Because of their hydrophobicity, all tamponade agents tend to be rounded rather than spread on the hydrophilic retina. If a bubble is rounded, it will not be very efficient at excluding aqueous gaining access to the retinal break unless:

1. The patient postures to position the bubble against the retinal break or breaks
2. The bubble totally fills the eye

How rounded a bubble is inside the eye depends slightly on the interfacial energies of the bubble used; the shape of a tamponade bubble is determined principally by its specific gravity.

Summary for the Clinician

- **Surface energy acts to minimise the surface area for a given volume. A bubble of tamponade tends not to go through a retinal break because to do this will increase its surface area. In this sense, the tamponade "plugs" the hole**

10.3 Specific Gravity

The term "specific gravity" is used to describe the weight or density of a liquid compared to an equal volume of fresh water at 4 °C. Thus water has a specific gravity of 1 and a material with a specific gravity less than 1 will float on water and one with a specific gravity greater than 1 will sink below the water.

The shape of very small bubbles of tamponade agent in aqueous will be controlled primarily by the interfacial energies of the system and will therefore be rounded. As the volume of the tamponade bubble increases the influence of buoyancy forces will become apparent.

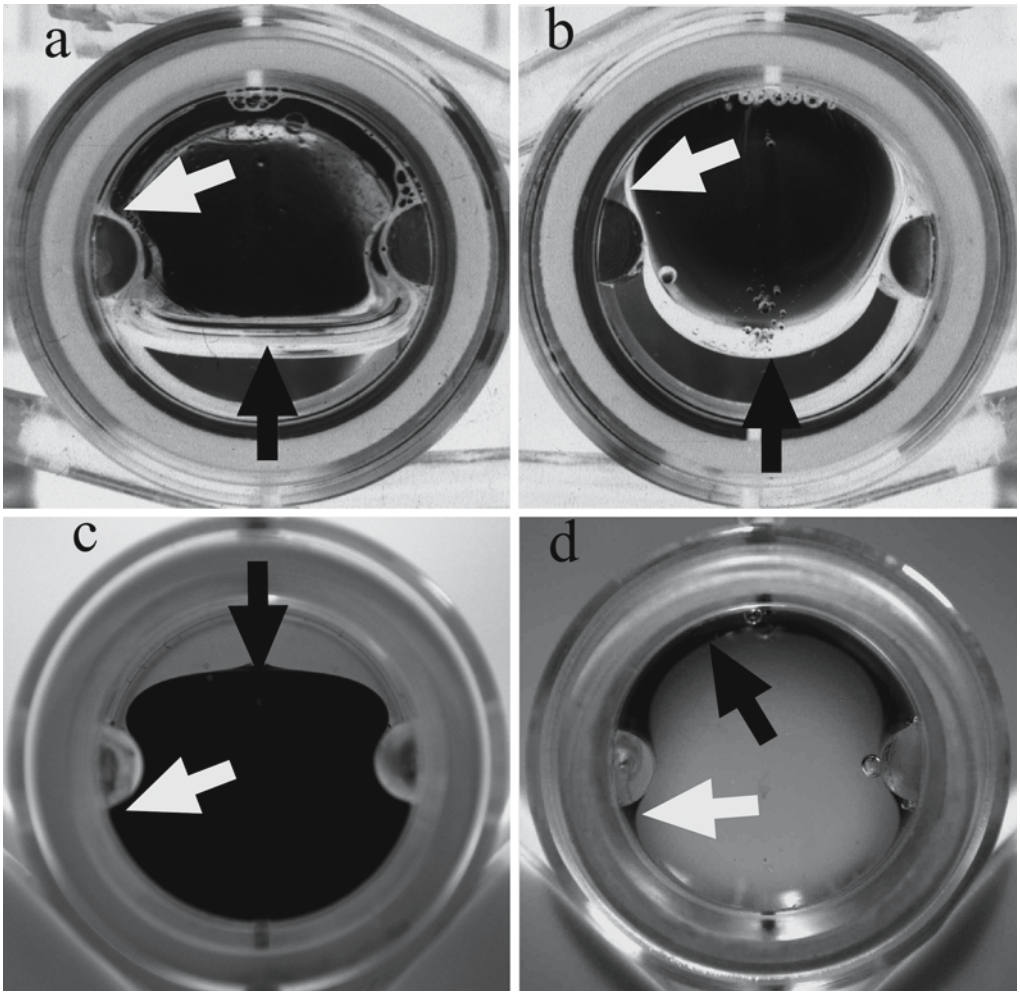


Fig. 10.3 a–d. Model eye chamber with two indents at the 3 and 9 o'clock positions. Chamber **a** is filled with air, **b** with silicone oil, **c** with perfluorohexyloctane and **d** with perfluorohexyloctane-silicone oil solution (Densiron-68). The *white arrow* points to the recess between the indent and the chamber wall. Air and perfluorohexyloctane fit into the recess, whereas silicone oil and Densiron do not. The *black arrow* points to the top surface of the bubbles. Air and perfluorohexyloctane have a flattened top surface whereas silicone oil and Densiron have a rounded top surface

Archimedes' principle states that the buoyant force on a submerged object is equal to the weight of the fluid displaced by the object. So for a bubble inside an eye, the weight of fluid can be calculated knowing the volume of the bubble and the specific gravity of aqueous.

The buoyancy force, F_B, is given as:

$$F_B = \rho_{\text{aqueous}} \times g \times V_{\text{bubble}}$$

where

- ϱ_{aqueous} = density of aqueous
- g = gravitational constant
- V_{bubble} = volume of the bubble

This force is always upwards. The downward force, F_D, on the bubble is the weight of the bubble itself, which is given as:

$$F_D = \rho_{\text{bubble}} \times g \times V_{\text{bubble}}$$

where

- ϱ_{bubble} = density of the tamponade agent.

The net force, F_N, acting on the bubble is therefore:

Buoyancy force – weight of the bubble

$F_N = F_B - F_D$

$= (\rho_{aqueous} - \rho_{bubble}) \times g \times V_{bubble}$

Let us consider an eye with a 2-ml-size bubble of **air. The difference in the density of air and aqueous** is large. Air at sea level weighs 0.0013 g/ml^3 whereas water weighs 1 g/ml^3. The net force is:

$F_N = (1 - 0.0013)\text{g/ml}^3 \times 980\ \text{cm/s}^2 \times 2\ \text{ml} = 1960$ dynes

This force is acting on the whole bubble and every molecule inside wants to float upwards. It is therefore not surprising that the bubble takes on almost a spherical cap shape.

Contrast this to a 2-ml silicone oil bubble. The net force is:

$F_N = (1 - 0.97)\text{g/ml}^3 \times 980\ \text{cm/s}^2 \times 2\ \text{ml} = 58$ dynes

This net force is still positive and acting upwards, but is small compared to that acting on the air bubble. The shape of the bubble is therefore mainly determined by the interfacial energies and is rounded.

The net force acting on a tamponade bubble is directly proportional to the size of the bubble and the difference in specific gravities between the tamponade agent and aqueous. So for small bubbles, the net force is small also. Clinically, one notices for example that when 0.3 ml of C_3F_8 gas bubble is injected for pneumatic retinopexy [14], the bubble is initially rounded. It is only after 12 h or more when the bubble has expanded in size that the bubble takes on the flat bottom shape.

The differences in specific gravity may also determine how well a tamponade agent fits into the nooks and crannies. In a model eye study, we have shown that the very light (air) and heavier tamponade agents (perfluorohexyloctane) fit much better into tight spaces than does silicone oil with a specific gravity close to that of water [36] (Fig. 10.3).

Summary for the Clinician

- **The specific gravity determines the shape of a bubble. Air is light and the buoyancy force makes a bubble of air inside an eye approximate a "spherical cap"**

10.4 Tamponade Efficiency

Because of the hydrophobicity of tamponade agents, all bubbles inside the eye are slightly rounded in shape. Bubbles of air have a flat bottom surface; in geometry, the term "spherical cap" is used to describe such a three-dimensional shape. The specific gravities of all other commonly used gases are very close to that of air. For example, perfluoropropane (C_3F_8) is only 1.01 times heavier than air. The buoyancy force for all gaseous tamponade is high. All gas bubbles approximate the spherical cap shape.

If a bubble is truly a spherical cap in the geometrical sense, then all the volume is used towards making contact with the retinal surface. Although gas bubbles in aqueous approximate to being perfectly spherical cap shaped, they are not perfectly so. The bottom is slightly rounded. Figure 10.4 is a nuclear magnetic resonance image of a bubble of inside an ex vivo pig's eye. The aqueous is highlighted by contrast and appears as white; the bubble is black. It can be seen that the edge of the bubble is slightly rounded to form a meniscus. A small amount of volume is therefore wasted and provides no tamponade effect. Silicone bubbles are much more rounded and so more of the volume is wasted in forming the rounded meniscus.

The volume of the spherical cap is given by the formula (Fig. 10.5):

$V_{cap} = 1/3 \times \pi h^2 \times (3R - h)$

where

- h is the height of the spherical cap
- R is the radius of the sphere

The arc of contact, θ, is the angle subtended at the centre of the sphere and $h = R - R \cos \theta / 2$

We can then plot the arc of contact against the volume of a theoretical tamponade agent which has the shape of a spherical cap; R for example is taken as 11 mm, half the diameter of a normal-sized eye. This plot shows a sigmoid curve in three parts (Fig. 10.6 a). The initial part of the curve is almost vertical such that a small volume gives rise to a large arc of contact. For example, a volume of 0.24 ml would give a 90° arc (or 3 clock hours) of contact. This is the part

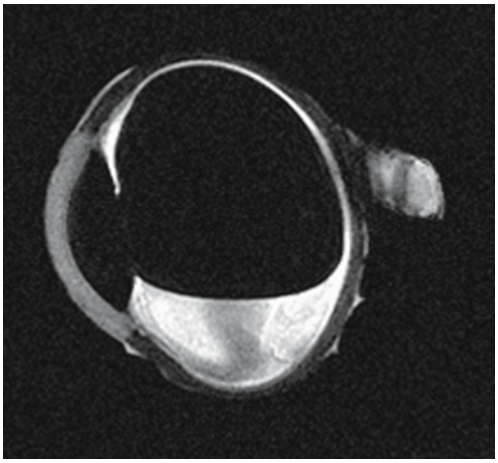


Fig. 10.4. Nuclear magnetic image of a bubble of air inside a vitrectomised ex vivo pig's eye

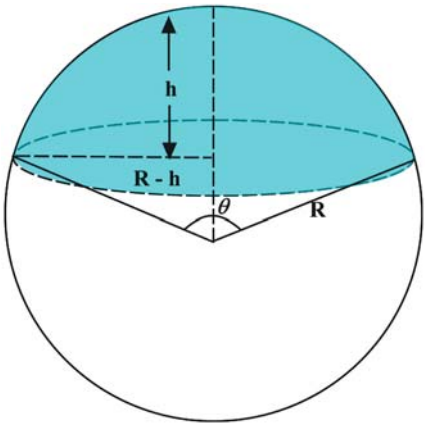
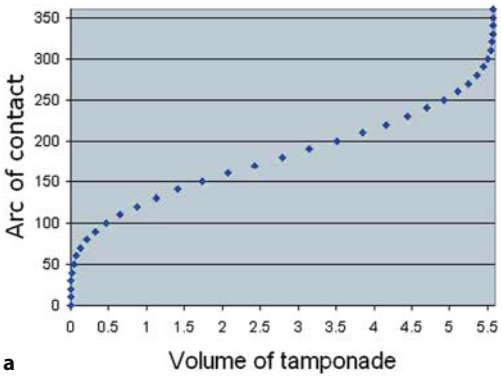


Fig. 10.5. Geometry of the spherical cap



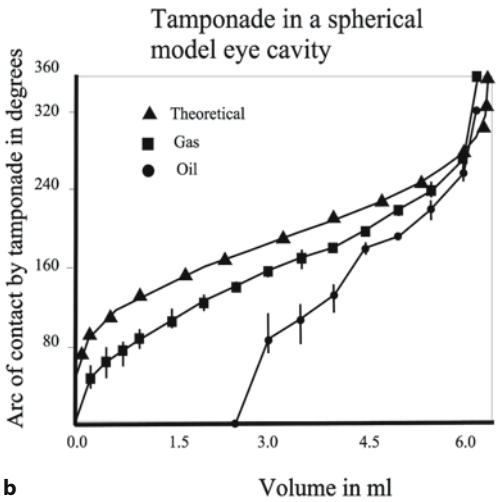


Fig. 10.6. **a** A plot of the arc of contact versus the volume of the spherical cap; **b** measurements from a spherical eye model chamber

of the curve that is used in pneumatic retinopexy where a small bubble can give a large arc of contact. In reality, small gas bubbles are rounded and not spherical caps and as such are less efficient (see above). The next part of the plot is almost linear. The greater the volume, the larger the arc of contact. The last part of the curve rises exponentially. A slight underfill of 0.24 ml would leave the lower 3 clock hours of the sphere not in contact with the tamponade agent.

A spherical model eye chamber was made and the arcs' of contact of air and silicone bubbles were measured and the results are shown in Fig. 10.6b [9]. The plots for air and oil were both shifted to the right of the plot for the spherical cap. If the spherical cap is considered as the ideal tamponade agent, then both air and oil are less than ideal and of the two, air is more efficient [9]. For silicone oil, there is virtually no tamponade effect until the chamber is about half filled; at 90 % fill by volume, the arc of contact is about 180 degrees [9]. Two important points should be appreciated:

1. The spherical cap is the most efficient shape for a tamponade bubble. All tamponade agents are hydrophobic (relative to water);

therefore, a meniscus will be formed. Even agents that have specific gravities far from water such as gas or perfluorocarbon liquids (PFCLs) form bubble shapes that are less efficient than the spherical cap.

2. It is difficult, if not impossible, to achieve a total tamponade effect. The problem is not that tamponade agents are not efficient; even the most efficient theoretical agent, i.e. the spherical cap, has an exponential curve plot such that a slight underfill leaves a large area of retina unsupported.

The fact that it is difficult to achieve a total tamponade effect is unappreciated by many. Previous attempts have been made to achieve simultaneous superior and inferior tamponades using silicone and fluorosilicone oils resulting in the discovery of a third compartment [1]. More recent attempts at double filling using semifluorinated alkanes and silicone oil resulted in bubbles with an egg shape, offering very poor tamponade superiorly and to the sides [13].

There may be other factors which contribute to underfilling at the time of surgery. The choroid can be congested due to periods of hypotony peroperatively. Postoperatively, deturgescence of the choroid will create a greater capacity in the eye and an effective underfill. Similarly, it is difficult to achieve a complete vitrectomy. Even if an air/fluid exchange was carried out prior to silicone oil injection, there would still be a frill of gel at the base. Postoperatively, this residual vitreous gel will be compressed and lose its water content, once again leading to an underfill. In fact, compression of the vitreous by a tamponade is well known. Gas-mediated compression vitrectomy [31] was first introduced in the 1980s as an efficient alternative to mechanical vitrectomy in rabbits' eyes.

Summary for the Clinician

- **The most efficient tamponade agents assume the shape of a spherical cap**
- **It is difficult, if not impossible, to achieve a total tamponade effect**

10.5 Viscosity

Viscosity is a measure of the strength of adhesion within the material and is important in terms of tamponade agents in that a low viscosity material is easier to handle surgically and place within the vitreous cavity, but a high viscosity material is require to reduce the dispersion of the material once in the eye. Dispersion refers to the break-up of a bubble into droplets. Dispersion is caused by the shear forces at the interface of the tamponade agent and the aqueous or retina during eye movement. The multiple small bubbles, of course, have a larger surface area than a single bubble. In controlled situations, small bubbles will coalescence to form larger bubbles in order to achieve a lower surface energy. In the presence of surfactants, or cellular debris, the interface of the small droplets may be stabilised and the dispersed droplets then form an emulsion.

Emulsification is detrimental to the behaviour of the tamponade agent:

- Emulsification results in dispersed droplets of tamponade agent in aqueous. As such, it has no ability to plug or to exclude access of water to retinal breaks. Therefore it has no tamponade effect whatsoever.
- Emulsification leads to droplets of different sizes which may be small enough to activate macrophages and cause inflammation in the eye.

Summary for the Clinician

- **The higher the viscosity, the greater is the shearing force needed to disperse a tamponade agent into small droplets**

10.6 Nature of Toxicity

Toxicity is the prime concern in the development of any new tamponade agents. The literature, however, is often contradictory and in the case of silicone oil consensus has not been reached until perhaps the last 10 years or so, and only after the results of large clinical series [18] including prospective randomised controlled trials were published [19, 27]. Previous reports emphasised several different aspects of toxicity, namely histological changes, inflammatory changes, electrodiagnostic tests, proliferative response and changes observed in cell culture.

The interpretation of the histological changes is often difficult; most experiments are carried out in rabbits' eyes, which may not be a good model to extrapolate to human eyes; it is unclear whether the agents tested were purified and whether they contain contaminants including low molecular components and cyclical forms [21, 32]; agents were left in situ for varying lengths of time.

We thought it would be interesting to review the literature grouping agents according to their physical properties:

Agents that are close to the specific gravity of water: Silicone-fluorosilicone oil copolymer had a specific gravity of 1.16 g/cm^3, and a viscosity of 180 mPa produced changes in the rabbit eye of thinning and loss of outer plexiform layer in the inferior fundus. It is interesting to note that in the same experiment conventional silicone oil with specific gravity 0.97 g/cm^3 and a viscosity of 1,000–5,000 mPa produced similar changes in the superior fundus.

Agents with specific gravity much higher than water: These agents include perfluorophenanthrene [30] (2.03 g/cm^3) and fluorosilicone oil (1.29 g/cm^3) [6]. When heavier tamponade agents were tested, thinning and loss of plexiform layer were observed, but additionally migration of the photoreceptor nuclei towards the retinal pigment epithelium and even disorganisation of the photoreceptors were found.

Agents with low viscosities: Virtually all agents with a viscosity of less than 200 mPa caused an inflammatory reaction and a macrophagic response. These include perfluorophenanthrene [30], perfluoro-octane [7], perfluorodecalin [33], silicone-fluorosilicone copolymer [5] and perfluorohexyloctane [15].

It is tempting to explain all the observed changes on the basis of the physical properties alone. It is likely that a macrophage response will occur even against biologically inert substances if they are dispersed to a sufficiently small size. Polytetrafluoroethylene (or Teflon) is known to be biologically compatible and result in minimal inflammatory reactions as a bulk material. Particles of polytetrafluoroethylene and also silicone (of around 180 µm in size) were used to treat post-prostatectomy incontinence [4, 16]. When injected in the periurethral tissues they caused inflammation and fibrosis that resulted in an increased outflow resistance. It has already been pointed out that tamponade agents with low viscosity require a much lower shearing force for dispersion. Therefore, we think that it is illogical to consider any low viscosity agents (including all the PFCL liquids and the semifluorinated alkanes) for long-term tamponade.

The reverse, however, may not apply. Agents with high viscosity are not automatically safe. There are notable exceptions including the oligomers of semifluorinated alkanes [25]; despite a high viscosity of 1,750 mPa, OL62HV produced white deposits on the surface of the retina which consisted of cystic cells and amorphous material. The cause of the white deposits is uncertain although it is likely to be related to inflammation as similar changes occur with perfluorohexyloctane and to a much lesser extent with silicone oil [25].

It is probably true that the more gross histological changes tend to occur with the heavier agents. Again, perfluorohexyloctane seemed to be an exception, even though its specific gravity is relatively high at 1.35 g/cm^3; only early histological changes were observed in rabbits at 9 weeks. Many have argued that the histological changes are due to the weight of the heavy liquids. The additional pressure even from agents of the highest specific gravity, namely perfluorophenanthrene, is of the order of 1–2 mmHg (for a normal-sized

eye)*, which seemed insignificant when compared to the range of normal intraocular pressure and its diurnal variation. Instead, we have argued that the trophic changes might also be caused by the occlusion of water from the retinal surface. Winter et al. pointed out that a fine film of water is necessary for the function of the Müller cell potassium siphoning pump [37]. The thickness of the film of aqueous immediately in contact with the retina may be critical; they measured the thickness of the film of aqueous between the tamponade agents and the retina using optical computerised tomography in ex vivo eyes filled with perfluorodecalin and perfluorophenanthrene. They postulated that the pathological changes observed were those caused by excitotoxicity from potassium ion accumulation. We believe this explanation is more plausible whereas the mechanical pressing of the liquids on the retina is probably not of a significant magnitude. Exclusion of aqueous from the surface of the retina can account for pathological changes in the upper retina with silicone oil, whereas the negligible buoyancy pressure cannot do so convincingly.

Gas tamponade agents are efficient at excluding access of aqueous from the retinal break. One might ask why excitotoxicity does not occur. In all likelihood, the gas inside the eye will be saturated with moisture. There is likely to be a film of water on the surface of the retina at all times; visual field loss during macular hole surgery had been attributable to drying of the retina as a result of continuous air infusion and the use of moist air was advocated [22].

* Added pressure from any liquid column can be derived from a buoyancy formula similar to that given above: $P=h\times(\rho_{aqueous}-\rho_{bubble})\times g$
where
- h is the height of the column, the diameter of the eye
- $\varrho_{aqueous}$ is the specific gravity of water
- ϱ_{bubble} is the specific gravity of the heavy liquid
- g is the gravitational constant

For perfluorophenanthrene, assuming the diameter of the eye to be 2.2 cm, the added pressure is: 2.2 cm (1–2.03)×980 cm/s²=−2221 dynes/cm² or 1.66 mmHg (downwards).

Summary for the Clinician

- **Low-viscosity tamponade agents have a greater tendency to disperse and cause inflammatory reaction**
- **Tamponade agents with high specific gravity may be too efficient at excluding water from the retinal surface, thereby causing excitotoxicity and may account for histological changes**
- **Even for liquids with high specific gravity, the mechanical pressure is small (of the order 1–2 mmHg)**

10.7 Semifluorinated Alkanes

More recently semifluorinated alkanes (SFAs) have been developed as tamponade agents [20]. They are transparent liquids that are immiscible with water. In terms of their interfacial energies they are amphiphilic in that they have both a hydrocarbon end, which is highly hydrophobic, and a fluorocarbon end, which is less hydrophobic. Measured against water their interfacial energies are similar but slightly lower than that of PFCLs. They have a lower specific gravity than the PFCLs at between 1.2 and 1.7 g/cm³ and thus, although they will sink, the buoyancy forces will be less and therefore the bubbles will be more rounded than the same volume of PFCLs [39]. These materials have low viscosities of around 2.5 mPa, making them capable of injection and removal through small-bore instruments but susceptible to dispersion.

Due to their lower specific gravity compared to other PFCLs, it was thought that SFAs could be used as long-term tamponade agents; this was considered, theoretically, to carry a lower risk of retinal damage. Initial experimental studies in rabbits have shown that when left intraocularly for periods up to 3 months, perfluorohexyloctane caused no identifiable ERG abnormalities [41]. Similarly, no major retinal abnormalities were detected by light- and electron-microscopy studies. Dispersion of F_6H_8 was observed, however, early on after injection (between the 1st and the 15th postoperative days). In humans the use of SFAs as postopera-

tive intraocular tamponade agents has been reported [17, 29] to be tolerated without obvious signs of damage to the retina or optic disk; however, dispersion of the SFA was an early and frequent complication. Further studies have shown that this dispersion can induce dense macrophagic infiltration as well as epiretinal and retrolental membranes [15]. This inflammatory reaction limits the clinical usefulness of this agent [10, 26, 34].

Summary for the Clinician

- **Perfluorohexyloctane can cause inflammatory reaction which ultimately limits its clinical usefulness**

The amphiphilic properties of the SFAs makes them soluble in hydrocarbons and silicone oil. It is possible to take advantage of this property and mix SFAs with silicone oil. This is an attempt to make a tamponade agent that has the advantageous properties of each material and overcomes the disadvantages of each.

10.8 Combining Tamponade Agents

There are two distinct ways in which tamponade agents are used in combination, namely double filling of agents and solutions of agents.

10.8.1 Double Filling of Tamponade Agents

As discussed earlier, the SFA is soluble in silicone oil; however, this solubility is limited. The solubility is dependent on the viscosity of silicone oil and on the molecular weight of the SFA. The higher the viscosity of silicone oil the greater the difficulty of dissolving SFA into it. Equally, SFAs with higher molecular weights are less soluble in silicone. In all cases, there is a solubility gap; this means that the two substances are not soluble in each other in all proportions. At body or room temperature, there is a limit to the amount of SFA that can dissolve in silicone oil.

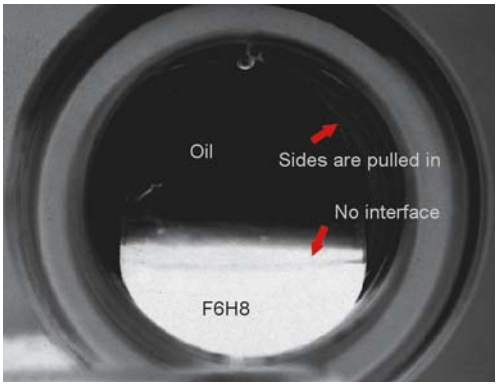


Fig. 10.7. Photograph of an egg-shaped tamponade formed by doubling filling with silicone oil and perfluorohexyloctane

Double filling was originally conceived to give simultaneous support to the upper and lower retina. At room temperature in an aqueous environment it is possible to make contact between a bubble of SFA and a bubble of silicone oil. Owing to the hydrophobicity of both materials, it is energetically favourable for the two tamponade agents to make contact and exclude water from the interface. This has the effect of producing a single bubble with two layers. The lower layer is the SFA and the top layer is silicone oil. In between there will be a layer in which some solubility of the SFA in silicone oil has occurred. The behaviour of this single bubble is different from either that of pure SFA or pure silicone oil. The higher specific gravity of the SFA and its attachment to the silicone oil tends to pull the double bubble below any remaining aqueous. However, the low specific gravity of the silicone oil will also tend to pull the SFA bubble upwards. Thus the overall shape of the double bubble is different from either pure fluid. We have shown that there is a tendency for the double bubble to be egg shaped (Fig. 10.7); thus it may have little tamponade effect at the sides and at the top.

The actual shape of the double bubble will depend on the ratio of SFA to silicone oil. In our model eye chamber, we found that if the amount of silicone oil was small that the bubbles would not unite [13]. When we used the model eye

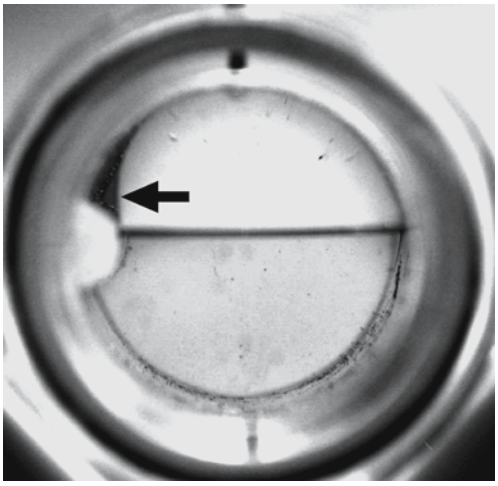


Fig. 10.8. Double filling in a chamber with a single indent at the side. Note the perfluorohexyloctane provides good contact below, but the silicone oil does not. The *white arrow* points to the recess between the indent and chamber wall

chamber with an indent (to mimic the scleral buckle or encircling band), we found that as the proportion of silicone oil was increased, progressively less lateral tamponading was observed (Fig. 10.8). One advantage of the silicone oil upper layer of the double bubble was that the increased viscosity of the silicone oil tended to produce a viscous cap on the SFA. Thus when agitated there was a reduction in the dispersion of the SFA.

Summary for the Clinician

- **Doubling filling was originally conceived to provide a simultaneous tamponade to the upper and lower fundus. The double bubble is egg shaped and effectively only gives good contact to the inferior retina**
- **A silicone oil cap may reduce the tendency of the perfluorohexyloctane to disperse**

10.8.2 SFA and Silicone Oil Solutions

The complete solubility of SFA in silicone oil can be produced in the laboratory within limits using high heat and sonication. It is possible to produce limits of different viscosity and specific gravities. In the past, we tested some of these materials with a range of properties as shown in Table 10.1. These have the advantage of being heavier than water but with a lower buoyancy force in comparison with SFA or PFCL alone. They also have a significantly increased viscosity in comparison with pure SFA and PFCL, making them potentially less susceptible to emulsification. Thus these mixtures have the potential to overcome two of the disadvantages of pure SFA and silicone oil. That is they will gently tamponade the inferior retina owing to the fact that their specific gravity is slightly higher than that of water and they have a viscosity in a useful range. Evaluation of the behaviour of these materials in a model eye chamber is shown in Fig. 10.9. Quantification of the bubbles was eval-

Table 10.1. Physical characteristics of agents tested in the eye model chamber

	Specific gravity (g/cm^3)	Interface tension at 25 °C against water (mN/m)	Viscosity (mPa)
Perfluoro-octane	1.73	55.0	1.76
F6H8	1.35	49.1	2.5
Silicone oil	0.97	35.4	5,000
F6H8-SiO solution A	1.01	56.25	3,167
F6H8-SiO solution B	1.03	45.43	1,948
F6H8-SiO solution C	1.06	40.82	1,387

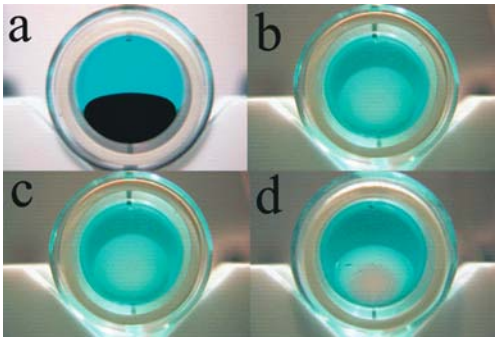


Fig. 10.9 a–d. Chamber filled with four different tamponade agents. **a** Perfluorohexyloctane (F6H8) (stained with Sudan black for better visualisation); **b** F6H8-silicone oil solution of specific gravity 1.01; **c** F6H8-silicone oil solution of specific gravity 1.03; **d** F6H8-silicone oil solution of specific gravity 1.06. Note **b** is the roundest and tallest bubble; pure F6H8 has the flattest and lowest profile

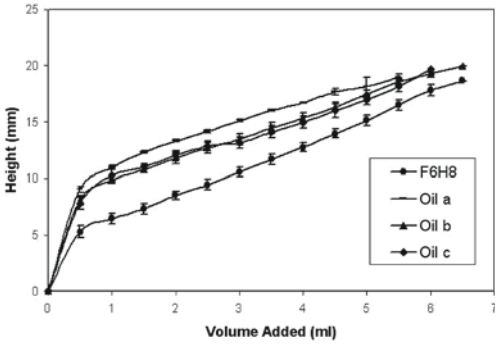


Fig. 10.10. A plot of the height of the bubble against incremental volumes of tamponade agents

uated by measuring the height of the bubbles and the results are presented in Fig. 10.10. This shows that the change in specific gravity from 1.01 to 1.03 does not cause a significant difference in the height of the bubble, but by increasing the specific gravity to 1.06 there is a significant decrease in the height for a specific volume of tamponade agent and therefore a greater tamponade effect [35]. The interfacial energies cause the SFA/silicone oil mixture to behave in a similar manner to pure silicone oil, that is they will make poor contact with the retina and they will not fit well into recesses formed by scleral explants (Fig. 10.3). However, it is expected that they will be as effective as silicone oil in the inferior section of the cavity. These data warrant further clinical evaluation.

10.9 Epilogue

So far, we have argued that the tamponade effect depends on fluids being immiscible with aqueous, thus forming an interface. It is interfacial energy which prevents tamponade agents going through retinal breaks in the detached retina. Once attached, a tamponade agent needs only to exclude access of aqueous to the retinal break. The most efficient tamponade bubble approximates the shape of a spherical cap. Most crucially, it is important to appreciate that a 100% fill of the vitreous cavity is probably impossible and a slight underfill gives rise to large arcs of the retina with no tamponade effect. This can be partly overcome by the patients adopting head-down posture. It is unrealistic to expect one or more than one tamponade agent to achieve a total tamponade effect, simultaneously supporting all parts of the retinal surface.

It is specific gravity that principally determines the shape of the bubble. Bubbles that are either very light or heavy better approximate the spherical cap. The buoyancy pressure by air or silicone oil and the downward pressure created by any agent are small, of the order of 1 or 2 mmHg. It does not seem convincing that forces of such small magnitude can give rise to trophic changes histologically. The pressure from heavy liquid is smaller than the diurnal fluctuation of intraocular pressure. Bubbles with high specific gravities may be too efficient at excluding aqueous from the retinal surface and this fact has been postulated as the cause of the retinal toxicity findings [37].

The availability of SFA and silicone oil solutions allows us to choose viscosity and specific gravity independently (within the limits of solubility). From our understanding of the tamponade effect we arrive at a crossroads. We desire liquids that are heavy so the bubbles would approximate better the spherical cap in shape. At the same time, we are concerned that such

heavy liquids would be too efficient at excluding aqueous from the retinal surface and therefore interfere with the normal ionic exchange [37].

The question comes back to why do we need new tamponade agents? After all, both gases and silicone oil work fairly well. Although not without complications, gas and silicone are used so frequently that we have become accustomed to using them and have learnt how to handle their complications. In the absence of PVR, silicone oil works well when 360° retinotomies are carried out [8, 23, 40]. Similarly, it has been shown that vitrectomy and gas tamponade can be effective for treating retinal detachments arising from inferior retina without scleral buckling.

Nonetheless, if we believe that the tamponade effect depends on contact between the tamponade agent and the retina, it is logical also to believe that tamponade agents that float are more efficient at treating pathology in the superior fundus; it is also realistic to expect that tamponade agents that sink would be more efficient at treating inferior pathology. Tamponade agents that are heavier than water may be particularly useful in cases of PVR as this has a propensity for the inferior retina [28]. If a retina becomes redetached after conventional surgery with gas or oil, the likelihood is that the redetachment would arise from retinal breaks situated inferiorly and be associated with epiretinal membranes.

In future, it may be acceptable to consider strategically staging the repair of the complicated retinal detachment, treating successively retinal pathology in the upper half then the lower half of the retina (or vica versa) using the most effective tamponade agent for each operation, protecting the macula from becoming detached at all times.

10.9.1 Serendipity

For long-term tamponade we have had available silicone oil [3] for the last 40 years and long-acting gases [2] for the last 20 years. It may be serendipity that gases form near spherical caps inside the eye but allow sufficient moisture to the retinal surface and avoid trophic changes. Equally, it may be chance that silicone oil is just light enough to form rounded tamponade bubbles in the eye, allowing sufficient aqueous around the bubble to prevent a toxic effect.

Others have argued that it might be desirable to design tamponade agents that make better contact with the retina, excluding aqueous milieux which contain proliferative cytokines and growth factors from the surface of the retina [17].

Now we have available to us SFAs and silicone oil solutions which behave very much like silicone oil. We have hopefully achieved our own serendipity by design. The initial clinical experience is certainly favourable [12, 38]. The questions are asked: will an SFA and oil solution be as efficient as silicone for treating PVR and more efficient than silicone oil for inferior pathology? An international multicentre randomised clinical trial is under way to find the answers.

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Vitreous Surgery in Uveitis and Allied Disorders

11

Tom H. Williamson

Core Messages

- In the Western population the commonest presentations of uveitis are likely to be intermediate uveitis and juvenile chronic arthritis
- Visual recovery is often limited because of the presence of optic atrophy or retinal damage particularly from cystoid macular oedema (CMO)
- Retinal vasculitis can produce ischaemia and a neovascular response associated with vitreous haemorrhage
- Rhegmatogenous retinal detachment (RRD) may occur from posterior vitreous detachment (PVD) formation and can be dealt with by vitreosurgery whilst being aware of the possibility of exacerbation of the uveitis
- In patients with intermediate uveitis, cystoid macular oedema may account for between 40% and 60% of eyes with poor vision
- Ultimately phthisis bulbi from hypotony is the most severe end point of these inflammatory conditions
- Uveitic syndromes may occasionally be difficult to discriminate from other causes of posterior infiltration such as infection and neoplasm
- Ocular lymphoma should be considered in a patient with steroid resistant posterior uveitis
- Highly active antiretroviral therapy (HAART) has changed the clinical picture of cytomegalovirus retinitis and reduced the number of presentations
- Fungal endophthalmitis (predominantly candida) is usually seen in patients with intravascular long lines, e.g. in intensive care units, or in patients with a history of intravenous drug abuse

11.1 Introduction

Inflammatory, infiltrative or infectious disorders of the posterior segment of the eye may require vitreous surgery to treat or diagnose. This chapter has been designed to examine the commoner presentations and to provide a guide to the most appropriate management options.

11.2 Inflammation

11.2.1 Non-infectious Uveitis of the Posterior Segment

- Presentations are varied
- Visual loss is common from cystoid macular oedema
- Hypotony is a catastrophic outcome

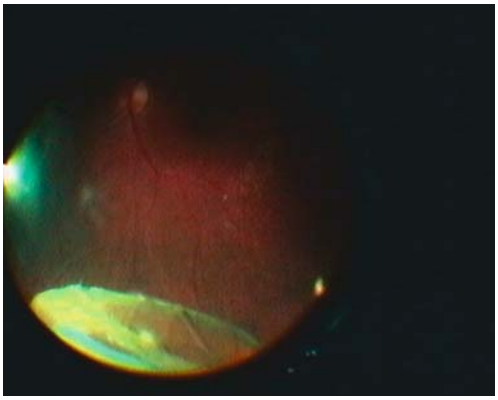

Fig. 11.1. Inferior snow banking in a patient with intermediate uveitis

The variety of possible presentations of uveitis of the posterior segment makes it difficult to generalise on the surgical approach [32]. The conditions that the surgeon may encounter, depending on the racial mix and geographical location, include:

- Intermediate uveitis
- Uveitis of juvenile chronic arthritis
- Sarcoidosis
- Idiopathic vasculitis including Eales disease
- Behçet's disease
- Bird shot chorioretinopathy
- Vogt-Koyanaga-Harada syndrome
- Sympathetic uveitis

In the Western population the commonest conditions are likely to be intermediate uveitis (Fig. 11.1) and juvenile chronic arthritis. Although often controllable with systemic therapy, those patients with more severe disease may require vitreoretinal intervention for the following reasons:

- Vitreous opacification
- Rhegmatogenous retinal detachment (RRD)
- Tractional retinal detachment (TRD)
- Exudative retinal detachment
- Cystoid macular oedema (CMO)
- Hypotony
- Diagnostic confirmation

11.2.2 Vitreous Opacification

The vitreous may become opaque because of the presence of cellular deposits, proteinaceous infiltration and degeneration of the gel structure. Intermediate uveitis is known to be complicated by vitreous haemorrhage, which can be treated successfully by vitrectomy [39]. Removal of the gel may restore vision. Many of these patients are young and have attached posterior hyaloid membrane (PHM), which will require removal but can be difficult to detach in the presence of vitreoretinal adhesions. Postoperatively further inflammation can ensue requiring systemic immunosuppressive cover. Visual recovery is often limited because of the presence of optic atrophy or retinal damage particularly from CMO [45]. Some surgeons claim that removal of the gel reduces the ability of the eye to hold inflammatory mediators and thereby reduces recurrence of inflammation in the long term. Evidence for this remains uncertain. Others argue that improvement following surgery is a result of the removal of vitreous opacity rather than any influence on the inflammatory process [32]. Reduction in medical treatment after surgery has been blamed for a rebound of inflammation 3–6 months later.

Retinal vasculitis can produce ischaemia and a neovascular response associated with vitreous haemorrhage. Pars plana vitrectomy (PPV) can relieve traction to prevent recurrent haemorrhage and clear the visual axis, but unlike diabetic retinopathy panretinal photocoagulation is not universally required.

11.2.3 Retinal Detachment

The inflammatory process can cause shrinkage of the gel, which in the presence of vitreoretinal adhesion may produce either TRD or RRD [22, 32]. In TRD from vitreous shrinkage without neovascularisation or preretinal fibrosis, PPV and peeling of the PHM will suffice. When TRD is associated with neovascularisation or fibrosis, delamination and dissection of the membranes is required (Fig. 11.2). A vitreoschisis as

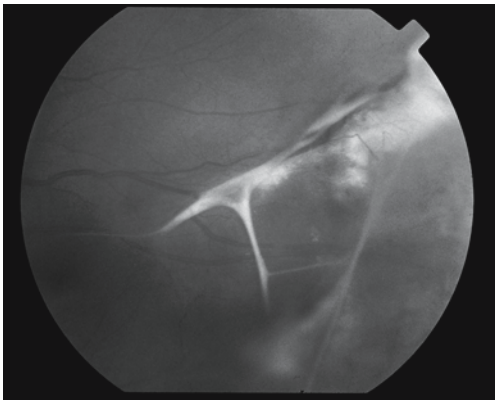

Fig. 11.2. A patient with epiretinal membranes associated with tractional retinal detachment and subretinal exudation from idiopathic vasculitis

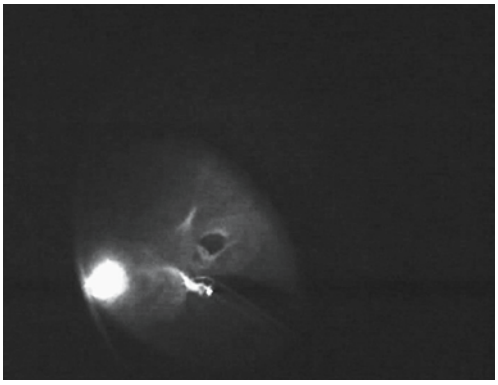

Fig. 11.3. Cutting away vitreous traction from a retinal tear caused by vitreous shrinkage in a patient with Behçet's disease

seen in diabetic retinopathy may be present and must be recognised to allow appropriate dissection under the plane of the PHM to aid delamination. Unfortunately TRDs are often associated with severe subretinal exudation and visual recovery is often poor. RRD may occur from posterior vitreous detachment (PVD) formation and can be dealt with by conventional means whilst being aware of the possibility of exacerbation of the uveitis (Fig. 11.3). Exudative retinal detachment may be encountered and diagnosed by shifting fluid and the absence of retinal tears, traction and epiretinal fibrosis (despite a long-standing duration of retinal detachment). Beware of the patient with uveal effusion syndrome. If immunosuppressive therapy does not reattach the retina, PPV and retinotomy to drain the subretinal fluid (SRF) may help. During the surgery the exudative nature of the retinal detachment can be confirmed by inserting heavy liquids onto the posterior retina. This will displace SRF anteriorly where it is trapped (because there is no retinal hole to allow drainage) and forms a tight ring bulla which overhangs the heavy liquid. Removal of the heavy liquid reveals a return of the retinal detachment to its previous configuration confirming no loss of SRF. Perform a small peripheral retinotomy to drain SRF, fill with long-acting gas or silicone oil, and laser the retinotomy. If the uveitis is then controlled, return of the retinal detachment is unlikely.

11.2.4 Cystoid Macular Oedema

In patients with intermediate uveitis, cystoid macular oedema may account for between 40 % and 60 % of eyes with poor vision [16]. Steroid injections into the vitreous cavity can reverse CMO in uveitis; however, the chronic nature of these conditions causes a return of the CMO after the steroid has been cleared from the eye [3]. Slow release steroid implants or injections may overcome this difficulty but as yet have not been extensively investigated. The vitreous is more often attached in patients with CMO. PPV has been performed to try to relieve traction on the macula to resolve CMO [45]. Separating the response to vitrectomy from the natural history of the condition and from the effects of concomitant therapies is difficult because randomised studies have not been done.

11.2.5 Hypotony

Ultimately phthisis bulbi from hypotony is the most severe end point for these inflammatory conditions. It causes a catastrophic visual loss and even a cosmetically unacceptable eye. Vitrectomy has been used to try and relieve traction on the ciliary body in hypotony (Fig. 11.4).

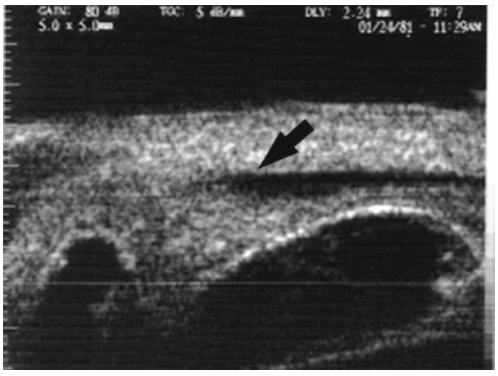

Fig. 11.4. A ciliary body detachment (*arrow*) on ultrasound biomicroscopy in a patient with hypotony from severe intermediate uveitis. The patient has silicone oil in situ

Inspection of the ciliary body with dissection of any tractional membranes has been performed only in a few patients and is as yet of uncertain worth especially as often the ciliary processes are atrophic and may be non-functional. Insertion of hyaluronic acid to provide a temporary intraocular pressure (IOP) rise has been employed. Silicone oil can be used for a more prolonged effect and to prevent severe shrinkage of the size of the eye if hypotony persists [35]. Long-term results of these interventions are unknown.

11.2.6 Diagnostic Confirmation

Uveitic syndromes may occasionally be difficult to discriminate from other causes of posterior infiltration such as infection and neoplasm. Laboratory processing of a vitreous sample may confirm the diagnosis especially if polyclonal white cells are seen on cytology without evidence of infection (Table 11.1). A significant number of patients with uveitis do not have a definitive diagnosis; only 66 % of cases of ante-

Table 11.1. Cytology of the vitreous in infectious and non-infectious uveitis

Non-infectious uveitis	Findings
Lymphoma	Atypical lymphocytes
Leukaemia	Atypical lymphoid cells
Metastatic tumour	Tumour cells
Melanoma	Tumour cells with melanin
Inflammatory uveitis	Inflammatory cells (plasma cells, lymphocytes, polymorphonuclear leucocytes, monocytes
Lens induced uveitis	Lens material, inflammatory multinucleate cells or phacolytic cells
Epithelial downgrowth	Fibroblasts
Amyloidosis	Acellular globules
Juvenile xanthogranuloma	Histiocytes, Touton giant cells
Infectious Uveitis	
Bacteria	Bacteria, neutrophils
Mycobacteria	Acid-fast bacilli
Fungal	Yeast, hyphae, mononuclear cells
Toxoplasmosis	Tachyzoites
Toxocariasis	Eosinophilia, plasma cells, second-stage larvae
Acute retinal necrosis	Inflammatory cells
Viral infections	Mononuclear cells

rior uveitis are associated with characteristic clinical and laboratory abnormalities, increasing to 85% in posterior uveitis with some baseline blood tests and a diagnostic vitrectomy can be obtained. Laboratory examination of the vitreous is particularly indicated when unusual or non-characteristic presentations occur.

Biopsy of the vitreous by needle aspiration may be effective in postoperative endophthalmitis where the vitreous is liquefied by the infection but may not be appropriate in non-infectious uveitis. Use of a vitreous cutter is recommended because the increased vitreoretinal adhesion in uveitic patients may make the risk of retinal detachment or tear higher [45]. Also many of these patients are young and likely to have non-syneretic vitreous gel, increasing the likelihood of a "dry tap" with a needle. In some cases vitrectomy is the ideal; in addition to providing a vitreous sample, vitrectomy may allow visualisation of the fundus allowing characteristic features of the disease process to be recognised and hence revealing a supplementary diagnosis.

Summary for the Clinician

- **Presentations are varied. Visual loss is common from cystoid macular oedema. Hypotony is a catastrophic outcome. The inflammatory process can cause shrinkage of the gel which in the presence of vitreoretinal adhesion may produce either TRD or RRD. Some surgeons claim that removal of the gel reduces the ability of the eye to hold inflammatory mediators and thereby reduces recurrence of inflammation in the long term. Steroid injections into the vitreous cavity can reverse CMO. Insertion of hyaluronic acid into hypotonous eyes to provide a temporary IOP rise has been employed. In some cases vitrectomy is the ideal; in addition to providing a vitreous sample, vitrectomy may allow visualisation of the fundus allowing characteristic features of the disease process to be recognised and hence revealing a supplementary diagnosis.**

11.3 Infiltration

11.3.1 Ocular Lymphoma

Lymphoma in the eye presents in elderly, usually female patients and often bilaterally. Ocular lymphoma should be considered in a patient with steroid resistant posterior uveitis. The clinical features, however, can be vague and varied with intravitreal white cells in a quiet eye, subretinal infiltration and occasional haemorrhagic retinal necrosis [2]. Pseudohypopyon can occur. Fifty percent of cases present because of ocular symptoms or signs, the rest because of CNS involvement (20% of CNS lymphoma will affect the eye). Usually a diffuse large cell B-cell lymphoma is implicated. Patients with AIDs may develop ocular lymphoma in which Epstein-Barr virus may be causal [33].

11.3.1.1 Diagnosis

- The lymphoma cells are fragile.
- Immunotyping is required.

A vitreous biopsy should be taken but requires rapid processing of the sample because the lymphoma cells are fragile and barely viable. Often cytology fails to identify the cells and differentiation from inflammation is difficult. Immunotyping to identify monoclonal cell lines is useful to overcome the latter problem [14]. Other options that have been employed include retinal biopsy or aspiration of subretinal infiltrates [12]. Investigation of systemic or intracerebral lymphoma is advised. Biopsy for neoplasia accounts for 14% of vitreous biopsies with 72% of these having ocular lymphoma [46].

11.3.1.2 Treatment

Surgery can be performed either for diagnosis or for restoration of vision because the vitreous cells are reducing vitreal clarity. PPV can be used to clear the visual axis and is usually uneventful. Chorioretinal biopsy leaves the sur-

geon with the problem of what to do with the biopsy site. Often silicone oil insertion is required to maintain a flat retina. Chorioretinal biopsy should therefore be reserved for those eyes with poorer visual potential. Intraocular methotrexate can be inserted in resistant cases. Otherwise, low dose radiotherapy is very effective in reducing infiltration in these eyes [29] and systemic chemotherapy may be considered [2].

11.3.1.3 Prognosis

- Survival is relatively short because of central nervous system involvement.

The prognosis for visual recovery is good. However, the patients have a shortened life expectancy due to the development of intracerebral lymphoma resulting in a poor duration of survival for these patients of median 3 years [6].

Summary for the Clinician

- **Ocular lymphoma should be considered in a patient with steroid resistant posterior uveitis. Biopsy for neoplasia accounts for 14 % of vitreous biopsies with 72 % of these having ocular lymphoma. Patients have a shortened life expectancy due to the development of intracerebral lymphoma resulting in a poor duration of survival for these patients of median 3 years**

11.4 Infections

11.4.1 Cytomegalovirus Retinitis

- Highly active antiretroviral therapy (HAART) has changed the clinical picture and reduced the number of presentations.
- Patients may now lose vision from immune recovery uveitis.

11.4.1.1 Presentation

Cytomegalovirus (CMV) infects the retina in immunocompromised patients. Overwhelmingly these patients suffer from AIDs. Classically in AIDs the patient has a severe reduction in their CD4 count to less than 50 cells/μl. Others requiring systemic immunosuppression such as Wegener's granulomatosis or rheumatoid arthritis occasionally present.

Prior to the availability of HAART, 40 % of AIDs patients developed CMV retinitis. Retinal detachment was a common complication of the retinitis (50 % at 1 year after development of retinitis), usually slow in onset because of the presence of a formed and attached vitreous gel in these young patients, and was bilateral in 70 %. This was linked to early mortality at approximately 6 months [15].

With the introduction of HAART, control of viral load is much improved and consequently CD4 counts are more often preserved. This has led to a massive reduction in the numbers of new cases of retinitis which now may only occur when there is failure or resistance to HAART [34]. Since HAART, patients with CMV retinitis have shown an 81 % improvement in mortality [26] and a 60 % reduction in retinal detachment [25], but most will develop a condition called immune recovery uveitis which can reduce vision. This is characterised by posterior segment inflammation which can cause secondary complications such as CMO, vitreomacular traction [10], vitreous haemorrhage from retinal neovascularisation [49], and even activation of previously quiescent infections of the choroid such as mycobacteria [50].

11.4.1.2 Diagnosis

The retinal appearance is usually typical in the at risk patient with a necrotising, haemorrhagic retinitis with a sharp demarcation between healthy and affected retina. However, biopsy is required to allow targeted therapy. An intravitreal biopsy of 0.2 ml is adequate for the detection of viral DNA on polymerase chain reaction (PCR) for CMV (Table 11.2).

Table 11.2. Causes of vitritis detectable by PCR

1. Herpes simples virus 1 and 2 (HSV 1 and 2)
2. Varicella zoster virus (VZV)
3. Cytomegalovirus (CMV)
4. Epstein-Barr virus (EBV)
5. *Borrelia burgdorferi*
6. *Toxoplasma gondii*
7. *Mycobacterium tuberculosis*
8. *Propionibacterium acnes*
9. Whipple's disease

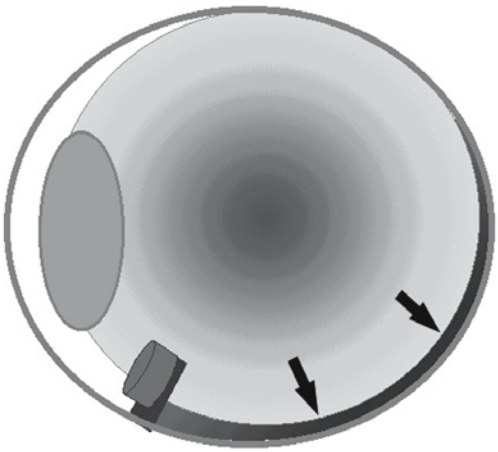

Fig. 11.5. The ganciclovir from an implant is concentrated in a thin aqueous layer (*arrows*) when silicone oil is in situ in the vitreous cavity

11.4.1.3 Surgery

- Retinal detachment occurs less often since the availability of HAART and can be treated without permanent silicone oil insertion.

Treatment of retinitis involves intravitreal antiviral often in the form of a slow release ganciclovir implant. This contains 4.5 mg of ganciclovir, is inserted into the pars plana at 4 mm from the corneoscleral limbus and may last up to 1 year. The implant provides local drug delivery, bypassing the blood ocular barrier with low dosage whilst minimising systemic side effects. In approximately 12%, problems are encountered such as extrusion, vitreous haemorrhage, or CMO [28]. Endophthalmitis occurs in 0.4% [43]. Implants can be used in eyes with silicone oil insertion although the reduced aqueous layer means increased concentrations of the drug [30] (Fig. 11.5). Increasingly, the control of the viral load is most important to the control of the retinitis by allowing cessation of anti-CMV therapy as the CD4 count recovers [49].

The clinical picture of retinal detachment has changed because of the use of HAART. Previously patients required PPV with silicone oil insertion. Removal was not performed because of the likelihood of progression of the CMV infection and because the patient's shortened life span restricted the development of oil induced complications [4, 15]. Silicone oil has been used with and without inferior external buckle with similar success rates [21]. Immune recovery means that retinitis is no longer progressive and the life span of patients is very much prolonged; therefore surgery may be more appropriate with gas tamponade [9] or with silicone oil with later removal of the oil [42].

Attempts to restrict RRD formation or progression with prophylactic laser therapy around areas of retinitis has limited success [13] because the retinitis or retinal detachment extends through the laser barrier.

11.4.1.4 Prognosis

If retinal detachment occurs, the chance of visual recovery is better when the retina can be fixed with one operation but good vision is only possible in approximately 50% [4] although this may have improved with HAART.

Summary for the Clinician

- **Cytomegalovirus (CMV) infects the retina in immunocompromised patients. Classically in AIDs the patient has a severe reduction in their CD4 count to less than 50 cells/μl. HAART has changed the clinical picture and reduced the number of presentations. Patients may now lose vision from immune recovery uveitis**

11.4.2 Acute Retinal Necrosis

- Be aware of the risk of second eye involvement and encephalitis.
- Retinal detachments are common.

Viral infections of the retina cause a mixed arteritic and infiltrative retinitis. The causative viruses are commonly of the herpes simplex family. Herpes simplex is commoner in young patients either type 1 or 2 [27, 40]. These patients may have a history of cold sores.

Herpes zoster is commoner in the elderly [20] and can be associated with herpes zoster ophthalmicus and chicken pox. Epstein-Barr virus can rarely be implicated. The patients are generally immunocompetent, but herpes zoster has been implicated in both ARN [48] and progressive outer retinal necrosis in AIDs patients [37]. There is a significant risk of bilateral disease with fellow-eye involvement even years later and a long-term risk of encephalitis.

The retina has the appearance of peripheral haemorrhage and infiltration, which spreads posteriorly to involve the macula, but the presentation has variable severity. The retina may become moth eaten and retinal detachment is common, up to 50 %. In severe presentations exudative retinal detachment can occur [17]. Patients have been described with giant retinal tears [44], retinal neovascularisation [47] and peripheral retinal pigment epithelial tears [19].

11.4.2.1 Diagnosis

The clinical pattern can be useful in diagnosis, but vitreous biopsy is required to determine the diagnosis and to identify the infective agent. A vitreous sample of 0.2 ml is usually sufficient to identify the virus on PCR with a high yield.

11.4.2.2 Treatment

Systemic antiviral therapy such as acyclovir is given over a period of months to try to prevent involvement of the second eye and encephalitis. Intravitreal antiviral such as foscarnet can be inserted during biopsy.

Management of retinal detachment requires PPV, gas or silicone oil, laser and buckle depending on the situation [7]. Insertion of silicone oil is often necessary because a causative single break is frequently difficult to identify, large areas of the retina are thinned and damaged, and because proliferative retinopathy is common. Retinal attachment after multiple procedures is common (90 %) but visual recovery is poor.

11.4.2.3 Prognosis

The prognosis for vision is poor in the affected eye; therefore systemic therapy is essential to prevent involvement of the other eye.

Summary for the Clinician

- **The causative viruses are commonly of the herpes simplex family. Herpes simplex, either type 1 or 2, is commoner in young patients and varicella zoster commoner in the elderly. A vitreous sample of 0.2 ml is usually sufficient to identify the virus on PCR with a high yield. Systemic therapy is essential to prevent involvement of the other eye**

11.4.3 Fungal Endophthalmitis

- Should have a good prognosis if picked up early enough.
- The patient groups involved can cause delay in diagnosis.

Fungal endophthalmitis (predominantly candida) is usually seen in patients with intravascular long lines [23], e.g. in intensive care units, or in patients with a history of intravenous drug abuse [1]. The presentation is a slowly progressive endophthalmitis, sometimes bilateral, commencing with preretinal puffball infiltration (Fig. 11.6). The condition is often seen after routine examination of the fundus in an asymptomatic patient [1]. An intravenous line may have

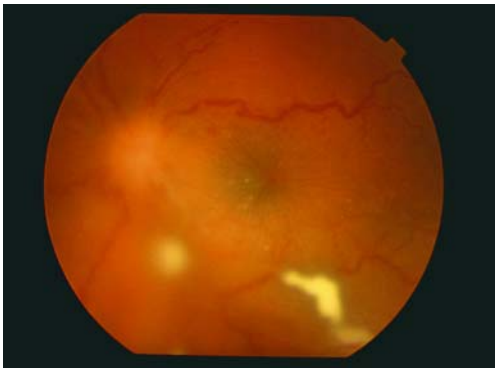

Fig. 11.6. An example of candida endophthalmitis requiring PPV and systemic antifungal therapy is shown

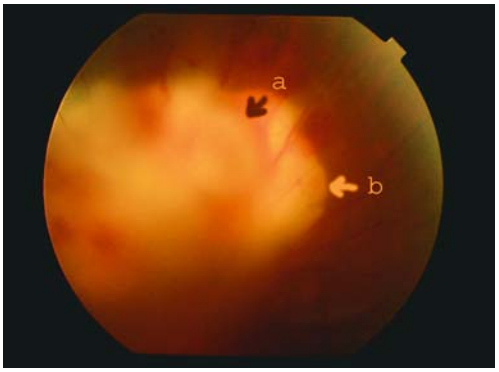

Fig. 11.7. A large focus of aspergillus in the vitreous overlying the macula (*a*), with infiltration of the choroid and retina (*b*)

been used on only one occasion. In heroin abuse the patient presents with reduction of vision after using acidic agents such as lemon juice (infected with candida) to dissolve brown heroin. Infections have been reported after gynaecological procedures [11] and postpartum [8]. Premature infants may also develop the infection. Contaminated infusion fluid in cataract extraction caused one surgical outbreak and the infection can be introduced during penetrating injury [38].

The infection progresses to a more severe vitreal infiltration with a string of pearl puffballs often with balls of white cells on the retina if the vitreous is detached. There may one or more foci of infiltration in the retina at the posterior pole. If untreated, epiretinal membranes may form resulting in macular pucker [31]. Retinal detachment can occur, and phthisis bulbi results.

11.4.3.1 Diagnosis

Often the clinical picture is so obvious that microbiological confirmation is not required. Screening by fundoscopy of intensive care patients with candidaemia can detect ocular involvement in a few percent [41]. Pathologically the hyphae reside in the puffballs [36]. A vitreous biopsy may fail to identify the fungus because the hyphae are scanty in the vitreous. PPV with microbiological processing of the washings in the vitrectomy cassette usually yields the diagnosis, but PCR has also been advocated [24]. The usual agent found is *Candida albicans* and rarely others occur such as *Candida krusei*. Fifteen percent of cases involve *Aspergillus* (Fig. 11.7), whilst *Fusarium* is rare [18].

11.4.3.2 Treatment

The mainstay of treatment is systemic antifungal therapy, e.g. fluconazole and flucytosine. This will easily deal with early infection without the need for surgery and should be commenced immediately. More advanced infection with significant intravitreal infiltration requires PPV, which because of the poor viability of the fungus in the eye will remove the local infection [5]. This can be performed usually on the next available operating list assuming lists every 2–3 days. Intravitreal amphotericin B may be inserted during PPV.

11.4.4 Vitrectomy

The surgeon should perform a dry vitreous biopsy at commencement of the surgery with the vitreous cutter. Many of these patients are young and therefore have an attached posterior hyaloid membrane (PHM). After core vitrectomy the PHM should be separated from the reti-

na. Any large focus of infiltration on the retina will usually detach with the PHM without causing undue traction on the retina. Residual white cells on the retinal surface can be aspirated. Secondary complications such as RRD or epiretinal membrane (ERM) can be dealt with by conventional methods.

11.4.4.1 Prognosis

The visual recovery depends on the severity of the infection and the location of any chorioretinal foci. In general the visual recovery is good if the infection is dealt with early. Late presentation or diagnosis is the main reason for poor visual outcome. The recovery of vision tends to be worse for *Aspergillus* than with *Candida*.

Summary for the Clinician

- **Fungal endophthalmitis (predominantly *Candida*) is usually seen in patients with intravascular long lines or intravenous drug abuse. The usual agent found is *Candida albicans* and rarely others occur such as *Candida krusei*. Fifteen percent of cases involve *Aspergillus; Fusarium* is rare. The recovery of vision tends to be worse for *Aspergillus* than with *Candida***

11.4.5 Other Infections

There are other less common presentations such as toxoplasmosis, which is associated with retinal detachments in approximately 6% of cases, and *Toxocara canis* may be the cause of tractional retinal detachments in childhood. Tuberculosis produces a vasculitis similar to idiopathic vasculitis and Eales disease and can result in retinal detachment despite response to systemic therapy.

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