Pathology of vitreoretinal junction biopsies

Paul Hiscott

Abstract

Advances in ophthalmic microsurgical techniques have provided surgical options in a variety of disorders that involve the interface between the retina and the vitreous. These conditions include epiretinal membrane, macular hole, macular oedema, vitreomacular traction syndrome and haemorrhage beneath the vitreous surface of the retina (as, for example, may be seen in Valsalva retinopathy and Terson's syndrome). This review aims to examine some of the histological features of the specimens that may present to the pathologist from this region of the eye and explore how the surgical interventions themselves, or surgical adjuncts, may alter the pathology of the specimens.

Keywords epiretinal membrane; macular hole; macular oedema; surgical dyes; vitreomacular traction syndrome; vitreous substitutes

Introduction

Almost 10 years ago, in *Current Diagnostic Pathology* (the forrunner to this journal), I presented an article "Vitreous biopsy pathology: new kid on the block".[1] The article outlined how the then relatively recent advances in microsurgery of the vitreous had enabled surgeons to obtain biopsy material at early stages of vitreous diseases. As a result, understanding of the pathobiology of the vitreous had been enhanced. That article concluded by stating: "We can expect further challenges for the pathologist as novel vitreous replacements are introduced."

Over the last decade, new vitreous replacement agents (or "tamponade agents": instilled by surgeons after vitrectomy to stop retinal detachment) have indeed been introduced. Moreover, instrumentation has again advanced and other techniques, such as the use of dyes intra-operatively in order to improve surgical visualization of vitreoretinal structures, have also become established. These developments permit improved surgical intervention in conditions that were previously difficult to treat, such as macular hole and fluid accumulations at the vitreoretinal interface. They have also led to refinements in surgery for local proliferative disorders. Thus more accurate dissection of the layers at the vitreoretinal junction ensures complete removal of abnormal proliferative tissue and hence a reduced risk of re-proliferation. As a result of these changes, pathologists who serve vitreoretinal surgeons are likely to receive a range of specimens from this part of the eye. The surgeon is not looking for a diagnosis with most of these specimens, since in the majority the aetiology is already known. Rather, the surgeon may ask the pathologist for other data such as information concerning the surgical dissection or cleavage plane, whether there is evidence of cellular proliferation within the retina (see Section on reactive proliferations below) or if the retina appears to have been damaged during surgery. Such data may allow the surgeon to provide a prognosis with regard to visual outcome after surgery as well as yield feedback for the surgeon (for example, in relation to surgical techniques or adjuncts).

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Anatomy of the vitreoretinal junction (Figure 1)

The vitreous is the "core" of the eye, representing up to 80% of the bulk of the organ. It is a rudimentary mass of transparent connective tissue that contains predominantly type II collagen fibrils and hyaluronic acid in an aqueous gel. Near the retina, the collagen fibres tend to condense and form a poorly-demarcated zone known as the vitreous cortex (Figure 1). Apart from the retina, the vitreous cortex is related to the optic nerve head (posteriorly) and the ciliary body, zonules and lens (anteriorly). The few cells that the vitreous contains are found in the cortex. Although these are sometimes called hyalocytes, they probably represent cells derived from circulating monocytes. They tend to be concentrated in front of the optic nerve head (or optic disc) and in the vitreous base (which straddles the anterior limit of the retina or "ora serrata").

Most vitreous fibres run parallel to the retinal surface so that the union between posterior retina and vitreous is delicate. A few vitreous fibres do pass into the innermost layer of the retina. In the vitreous base area, however, bundles of vitreous cortical fibres extend into crypts within the most peripheral part of the retina.

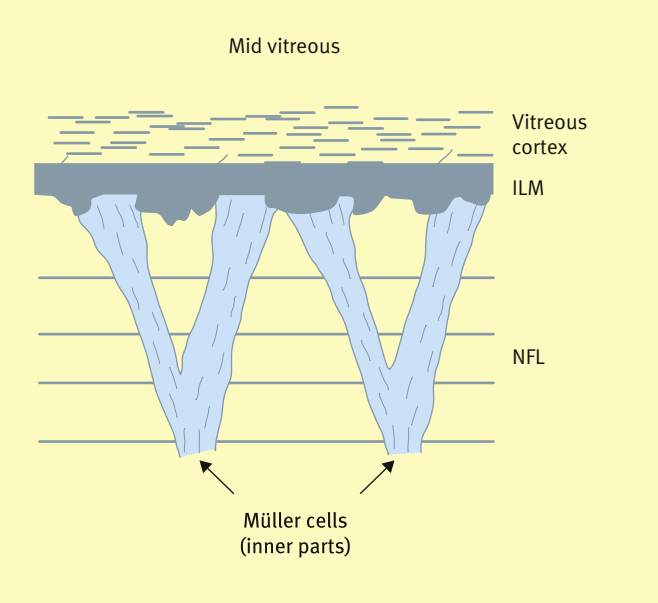

Figure 1 Diagrammatic representation of the anatomy of the vitreoretinal junction. ILM = internal limiting membrane of the retina; NFL = nerve fibre layer. [NB. There is a gap between the ILM and the Müller cell footplates that appears as an electron-lucent "sublaminar space" of around 400 Å on transmission electron microscopy].

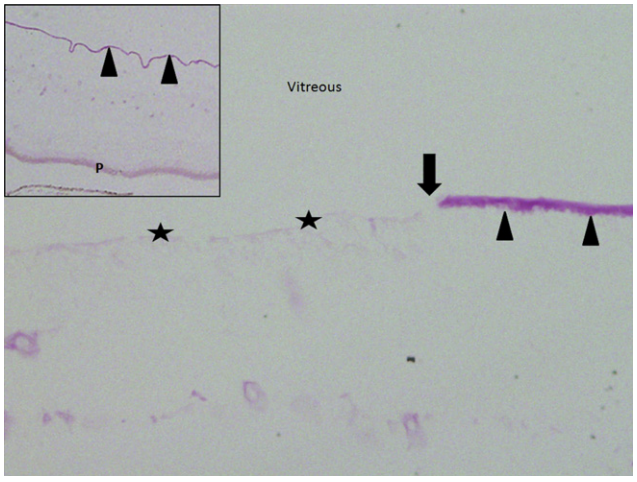

Figure 2 Diastase-PAS stain of the retina (no counterstain). *Inset*: normal retina showing the PAS-positive ILM (*arrowheads*) — the photoreceptor outer segments are marked (*P*) for orientation purposes. Main panel also depicts ILM (*arrowheads*) and the irregular or undulating configuration of the retinal side of the ILM can be seen. There is an abrupt discontinuity in the ILM (*arrow*) at the site of previous surgery 12 months previous to enucleation of the eye. Note that the ILM has not regenerated in the 1-year period since this surgery, so that the vitreoretinal junction remains deficient in ILM (*stars*).

There has been some confusion with regard to the terms used to describe structures at the vitreoretinal junction. Adjoining the vitreous cortex is the homogenous lamellar structure that represents the innermost part of the retina. It was thought that this structure was the basement membrane of Müller cells (modified astrocytes that provide the sustentacular architecture of the retina) and it was called the inner limiting lamina of the retina.[2] The combination of this lamina with the "overlying" vitreous cortex and the "underlying" meshwork of Müller cell inner processes with their footplate-like extensions was termed the internal limiting membrane of the retina. However, more recent evidence has emerged that the lamina, though containing basement membrane materials, is largely derived from proteins secreted from the ciliary body and lens during ocular development (at least in avian species).[3] Further evidence for this concept has emerged from observations that the lamina does not regenerate significantly after surgical removal in man (Figure 2). Perhaps in part in reaction to these observations, the term internal limiting membrane (ILM) of the retina is now applied to the lamina itself (i.e. without including the vitreous cortex and Müller cell processes) by many vitreoretinal surgeons. Therefore, I shall use "ILM" for this structure. In histological preparations, this structure is intensely PAS-positive while in transmission electron microscopy the ILM appears as an electron-dense layer (Figures 2 and 3).

Whereas the vitreous surface of the ILM is relatively smooth, the close proximity of the Müller cell footplates imparts an irregular or undulating configuration to the retinal side of the ILM (Figures 1–3): a feature that can help orientate biopsies from the vitreoretinal interface. Despite their close proximity, a natural cleavage plane can appear between ILM and Müller cells (sub-laminar space: see below). Occasionally, non-Müller glial cells (accessory glia or fibrous astrocytes) can be found close to the ILM.[2]

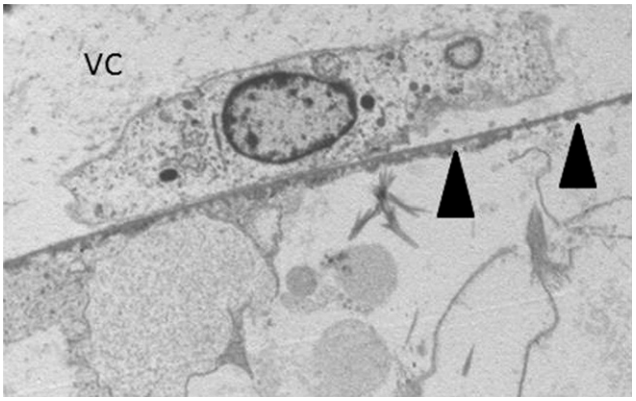

Figure 3 Transmission electron micrograph of the vitreoretinal interface. The vitreous cortical (*VC*) fibres are visible and the ILM appears as an electron-dense layer: again the irregular or undulating configuration of the retinal side of the ILM can be seen (*arrowheads*). Note the presence of a single cell on the vitreous ("epiretinal") surface of the ILM.

The Müller cells themselves pass through the neuroretina to the external limiting membrane, embracing the other layers of the sensory retina as they go. These include the nerve fibre layer, next to the Müller cell footplates, and the ganglion cell layer next to the nerve fibre layer.

Topographical and chronological variations in the vitreoretinal junction

Both spatial and temporal variations occur at the vitreoretinal junction. The main spatial variation relates to the thickness of the ILM. In the peripheral retina, the ILM is relatively thin and it also contains the discontinuities that permit vitreous cortical fibres to enter the retinal crypts. The ILM becomes thicker towards the back of the eye so that in the posterior pole it reaches up to 3 μm thick. However, over the fovea and retinal vascular arcades the ILM is again relatively thin (around 0.1 μm thick).

With age, the vitreous undergoes a degenerative process ("syneresis") that results in the gradual liquefaction of the gel. Cavities of fluid ("syneretic cavities") form. At the same time there is a weakening of vitreoretinal adhesion. Often, this combination allows fluid vitreous to enter the potential space between vitreous cortex and ILM so that the vitreous cortex becomes detached from the retinal surface ("posterior vitreous detachment" or PVD). The vitreous cortex remains attached to retina in the vitreous base. It may also remain attached, at least for a while, to parts of the posterior retina such as the macula or optic disc rim. In these latter situations, the PVD is said to be "partial" rather than "complete" (complete is when the entire vitreous cortex is detached up to the vitreous base).

The vitreous attachment around the optic nerve head may be more tenacious than elsewhere in the posterior pole of the eye, perhaps because glial cells mixed with cortical vitreous (which here represent the remnants of the posterior of Cloquet canal) serve to anchor the vitreous in this region. When a PVD commences, traction may be placed on the optic disc margins (sometimes causing the vitreopapillary traction syndrome where the optic nerve head appears oedematous). Usually, however, the adhesion at the margin gives way with part of the fibroglial material remaining attached to the back of the detached vitreous.