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Posterior Vitreous Detachment and the Posterior Hyaloid Membrane

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Purpose: Despite posterior vitreous detachment being a common ocular event affecting most individuals in an aging population, there is little consensus regarding its precise anatomic definition. We investigated the morphologic appearance and molecular composition of the posterior hyaloid membrane to determine whether the structure clinically observed enveloping the posterior vitreous surface after posterior vitreous detachment is a true basement membrane and to postulate its origin. Understanding the relationship between the vitreous (in both its attached and detached state) and the internal limiting membrane of the retina is essential to understanding the cause of rhegmatogenous retinal detachment and vitreoretinal interface disorders, as well as potential future prophylactic and treatment strategies.

Design: Clinicohistologic correlation study.

Participants: Thirty-six human donor globes.

Methods: Vitreous bodies identified to have posterior vitreous detachment were examined with phase-contrast microscopy and confocal microscopy after immunohistochemically staining for collagen IV basement membrane markers, in addition to extracellular proteins that characterize the vitreoretinal junction (fibronectin, laminin) and vitreous gel (opticin) markers. The posterior retina similarly was stained to evaluate the internal limiting membrane. Findings were correlated to the clinical appearance of the posterior hyaloid membrane observed during slit-lamp biomicroscopy after posterior vitreous detachment and compared with previously published studies.

Main Outcome Measures: Morphologic appearance and molecular composition of the posterior hyaloid membrane.

Results: Phase-contrast microscopy consistently identified a creased and distinct glassy membranous sheet enveloping the posterior vitreous surface, correlating closely with the posterior hyaloid membrane observed during slit-lamp biomicroscopy in patients with posterior vitreous detachment. Immunofluorescent confocal micrographs demonstrated the enveloping membranous structure identified on phase-contrast microscopy to show positive stain results for type IV collagen. Immunofluorescence of the residual intact internal limiting membrane on the retinal surface also showed positive stain results for type IV collagen.

Conclusions: The results of this study provide immunohistochemical evidence that the posterior hyaloid membrane is a true basement membrane enveloping the posterior hyaloid surface. Because this membranous structure is observed only after posterior vitreous detachment, the results of this study indicate that it forms part of the internal limiting membrane when the vitreous is in its attached state. *Ophthalmology 2017;■:1–10 © 2017 by the American Academy of Ophthalmology. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).*

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The vitreous humor is the largest anatomic structure in the human eye; it is a specialized transparent connective tissue that fills the posterior segment of the eye, occupying more than three quarters of the total ocular volume. In common with all connective tissues, which are separated from their adjacent epithelium, mesothelium, or endothelium by true basement membranes composed of type IV collagen,[1] the vitreous is separated from its adjacent neuroepithelium (retina) by the internal limiting membrane composed of type IV collagen.[2]

Uniquely in humans, the vitreous undergoes a progressive morphologic remodelling with an increase in fluid-filled lacunae (synchysis)[3] and an increase in collagenous condensations (syneresis)[4] as part of the aging process. In youth, collagen fibrils of the posterior vitreous cortex are firmly adherent to the internal limiting membrane of the retina by proteoglycans, including laminin and fibronectin.[5–8] Both vitreous syneresis and posterior vitreous detachment are thought to be influenced by age and refractive error, but are separate pathologic entities and frequently occur independently of the other.[3,9,10] Posterior vitreous detachment may be an asymptomatic finding in many patients; however, in those patients who are symptomatic, separation of the vitreous from the retina is

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associated with an acute onset of flashes and floaters. Flashes refer to the pathognomonic features of temporal photopsia, described as a momentary arc of white or golden light in the lateral field of vision, whereas floaters refer to the subjective perception of the shadows cast onto the retina by vitreal opacities.[11,12] Despite being a common ocular event affecting most individuals in an aging population,[13–15] there is still recognized difficulty in making an accurate diagnosis in all cases and little agreement regarding the precise anatomic definition of posterior vitreous detachment.

Imaging-based diagnosis of posterior vitreous detachment traditionally has relied on dynamic B-scan ultrasonography; with an experienced operator, it reliably correlates to clinical biomicroscopy and intraoperative evidence of posterior vitreous detachment.[16] More recently, OCT has been introduced in an attempt to objectify and standardize the diagnosis of posterior vitreous detachment. Although initial correlations to clinical and intraoperative posterior vitreous detachment have been limited,[16,17] developments in increased-resolution spectral-domain OCT, wide-field scanning patterns, dynamic examination protocols, and combination imaging with scanning laser ophthalmoscopy promise to improve the potential of this imaging method to diagnose posterior vitreous detachment reliably in the future.[18,19]

The generally accepted consensus regarding the definition and diagnosis of posterior vitreous detachment is that there is a separation of the condensed outer layers of type II collagen fibrils of the vitreous, known as the posterior vitreous cortex, from the internal limiting membrane of the retina.[20–24] The term *posterior hyaloid face* is used commonly to describe the outermost cortical layer of the vitreous. Clinical diagnosis of a suspected posterior vitreous detachment is based on the clinical history of new-onset flashes and floaters, with or without the identification of epipapillary glial tissue torn form the optic nerve head (Weiss ring) noted in the posterior vitreous cortex on slit-lamp biomicroscopy.[25–27]

An alternative, more stringent clinical interpretation defines posterior vitreous detachment as the separation of the vitreous and its enveloping posterior hyaloid membrane from the surface of the retina.[12,28,29] Proponents of this concept advocate the term *posterior hyaloid membrane* to describe the membranous structure visualized on slit-lamp biomicroscopy encasing the detached vitreous. Clinical diagnosis of a suspected posterior vitreous detachment is based on clinical history, in addition to the identification of a continuous, discrete, highly creased, and refractile membranous sheet observed by slit-lamp dynamic vitreous biomicroscopy with a wide illumination observation angle (Fig 1; Video 1, available at www.aaojournal.org). It follows that notwithstanding any associated syneresis within the gel itself, this structure must form part of the vitreoretinal interface and internal limiting membrane immediately before vitreous detachment.

Previous studies examining patients with and without posterior vitreous detachment antemortem and correlating the findings with the postmortem histologic investigation of the vitreous in the same patients demonstrated that cadaveric posterior vitreous detachment is consistent with, and a true representation of, antemortem posterior vitreous detachment.[14,30] This study compared the histopathologic and immunohistochemical profiles of the posterior hyaloid membrane in donated human globes with posterior vitreous detachment.

Methods

Study Design

The posterior surface of the vitreous body in donated human globes identified to have posterior vitreous detachment was interrogated with phase-contrast microscopy and subsequently histochemically stained for basement membrane (collagen IV), vitreoretinal junction (fibronectin, laminin), and vitreous gel (opticin) markers to facilitate immunofluorescent confocal microscopy analysis. The posterior retina similarly was stained with collagen IV immunofluorescence to evaluate the status of the remaining internal limiting membrane. The resultant micrographs were compared with the clinical appearance of the posterior hyaloid membrane observed during slit-lamp biomicroscopy examination of patients with posterior vitreous detachment.

Materials

Under a material transfer agreement, the Corneal Transplant Service Eye Bank, University of Bristol, supplied human globe tissue after harvesting an anterior segment 18-mm corneoscleral button for corneal transplantation surgery. The globe tissue, which comprised all ocular structures internal to and including the scleral coat posteriorly and the iris anteriorly, was fixed in 4% paraformaldehyde at 4°C and transported by cold chain courier. Upon receipt, the globe tissue was rinsed repeatedly in cold phosphate-buffered saline, cryoprotected in a 30% sucrose and phosphate-buffered saline solution, frozen in acetone chilled in liquid nitrogen, and stored at −80°C before being thawed on ice for dissection at room temperature.

Dissection Protocol

To evaluate posterior vitreous detachment status, a modification to the suspended-in-air examination, originally described by Foos,[31] was used. A circumferential full-thickness scleral coat incision into the suprachoroidal space was placed around the optic nerve stalk posteriorly. Anteriorly, the iris was removed circumferentially at its root. Radial full-thickness scleral incisions (connecting the anterior scleral edge to the posterior peripapillary incision) facilitated removal of the scleral coat from the remaining eviscerated vitreous body, covered by retina and choroid, and attached posteriorly to the optic nerve stalk and surrounding rim of scleral tissue.

The modification of Foos' air suspension technique assessed the posterior vitreous detachment status before and after removal of the choroid. Upon removal of the choroid, the retina of specimens with a posterior vitreous detachment would tear at their firm anterior attachment to the vitreous base under the weight of the vitreous body, leaving the denuded posterior surface of the vitreous body exposed. In distinct contrast, those globes without posterior vitreous detachment were found to have retina adherent to the entire posterior vitreous surface that was possible to remove only with piecemeal dissection.

Posterior-to-anterior 18-mm trephination through the vitreous body in specimens with posterior vitreous detachment allowed removal of the anterior annulus of retinal tissue attached to the vitreous base and peripheral anterior vitreous gel. This yielded a