Inner Retinal Optic Neuropathy: Vitreomacular Surgery–Associated Disruption of the Inner Retina

Billy X. Pan,[1,2] Kenneth M. Yee,[1,2] Fred N. Ross-Cisneros,[2] Alfredo A. Sadun,[2] and Jerry Sebag[1,2]

[1]VMR Institute for Vitreous Macula Retina, Huntington Beach, California, United States
[2]Doheny Eye Institute, Los Angeles, California, United States

Correspondence: J. Sebag, VMR Institute for Vitreous Macula Retina, 7677 Center Avenue, Huntington Beach, CA 92647, USA; jsebag@VMRinstitute.com.

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Purpose. Macular pucker (MP) and macular hole (MH) are vitreomaculopathies treated by vitrectomy and membrane peel. The complication of postoperative central scotoma can be associated with significant reduction in visual acuity (VA). We seek to determine whether retinal nerve fiber layer (RNFL) disruption is the pathophysiologic basis of this defect. Mitigating clinical circumstances also were sought.

Methods. Eleven eyes from 10 pseudophakic patients who had undergone vitrectomy with peeling for either MH or MP were studied with clinical measures, including optical coherence tomography (OCT). Membrane specimens were evaluated by immunohistochemistry for neurofilament, a marker for the inner retina. Ten eyes from 10 pseudophakic patients who underwent repeat surgery for persistent or recurrent pathology were evaluated to determine the relationship between the timing of reoperation and clinical outcome.

Results. Cases with a postoperative central scotoma ($N = 4$) had worse VA (~20/600) compared to those without ($N = 7$, ~20/30, $P = 0.01$). Eyes with a central scotoma had significantly reduced RNFL thickness in the temporal quadrant (53.67 vs. 72.33 μm, $P = 0.05$) by OCT. A central scotoma was associated with more disruption of the inner retina on immunohistochemistry ($P = 0.03$). In patients with persistent or recurrent pathology, waiting six months before reoperation resulted in better functional outcomes ($P = 0.03$).

Conclusions. Central scotomata and poor VA were associated with disruption of the RNFL during membrane peeling. Affected patients have RNFL thinning and signs of optic neuropathy, for which we propose the term inner retinal optic neuropathy (IRON). In patients requiring reoperation, waiting six months between surgeries may reduce the risk of IRON.

Keywords: inner retinal optic neuropathy, IRON, central scotoma, inner limiting membrane, neurofilament protein, immunohistochemistry, macular pucker, macular hole, vitrectomy, chromodissection, membrane peel

The pathogenesis of macular hole (MH) and macular pucker (MP) is not fully understood, although the unifying concept of anomalous posterior vitreous detachment recently has been proposed as the initiating event in both vitreomaculopathies.[1,2] Liquefaction of the gel vitreous without sufficient dehiscence at the vitreoretinal interface can result in a split within the posterior vitreous cortex, known as vitreoschisis.[3,4] Depending on the presence or absence of vitreopapillary adhesion,[5] the resultant tangential tractional force either will pull outward (centrifugal tangential traction) producing a MH, or contract inward (centripetal tangential traction) causing MP.[1,3–7]

Vitrectomy with peeling of a pathologic premacular membrane (PMM), with or without chromodissection,[8,9] is highly effective,[10–13] although there is some debate regarding whether or not the inner limiting membrane (ILM) also should be removed.[13–17] While surgery is safe and effective, complications remain a reality, ranging from cataract progression and retinal tears,[18,19] to relatively rare events, such as endophthalmitis and retinal detachment.[20,21] Peripheral visual field defects have been reported with increasing frequency,[22–31] and while many theories have been proposed to explain these phenomena, a definitive pathogenic mechanism has yet to be determined.

At times, dense central scotomata also may occur following surgery, resulting in significant impact on visual acuity (VA). An extensive literature search revealed only one case series in which the investigators concluded (based on clinical findings alone) that damage to the inner retina during membrane removal was the likely cause of central scotoma.[32] The present study was undertaken to identify the cause of central vision loss by immunohistologic evaluation of tissue removed at vitrectomy surgery to test the hypothesis that poor vision associated with a central scotoma after vitrectomy with membrane peel is due to disruption of the underlying retinal nerve fiber layer (RNFL).

Other complications may occur with surgical treatment for MH and MP, including treatment failure, and recurrent vitreomaculopathy that may transpire weeks, months, or even years later.[14,33–37] While a repeat membrane peel operation often will yield an anatomically successful result, the functional outcome from these reoperated cases is not always favorable.[38–41] A possible explanation for this is that there was an inadequate amount of time to allow for adequate wound

healing and tissue recovery after the first operation. Thus, the present study also tested the hypothesis that successful reoperation, specifically with respect to visual outcome, occurs more frequently if performed more than 6 months after the initial operation.

Materials and Methods

Study Design

Approval from the affiliated Institutional Review Board was obtained and all tenets of the Declaration of Helsinki were followed. Informed consent was obtained from the subjects after explanation of the nature and possible consequences of the study. A thorough review of patient records at the VMR Institute for Vitreous Macula Retina (Huntington Beach, CA, USA) was performed for the period between 2006 and 2012. Selection criteria for the histology portion of the study included patients with tissue specimens obtained at surgery for either MH or MP. Selection criteria for the repeat operation portion of the study included patients who had two surgeries for a PMM causing either MH or MP. Exclusion criteria were comorbid retinal pathologies, including exudative age-related macular degeneration and diabetic retinopathy. Only pseudophakic eyes were included in the study to avoid possible confounding effects of cataracts on postvitrectomy vision.

All diagnoses were confirmed by cross-sectional imaging of the retina using combined optical coherence tomography (OCT) with scanning laser ophthalmoscopy (OCT-SLO; OPTOS, Marlborough, MA, USA). Postoperative evaluation consisted of Snellen VA testing, slit-lamp biomicroscopy, OCT-SLO imaging to confirm resolution of the initial pathology and determine RNFL thickness. A select group of patients (those with optic atrophy noted postoperatively) were referred for neuro-ophthalmology consultation, which included further evaluation for afferent pupillary defect (APD), dyschromatopsia, brightness sense, and Humphrey visual field (HVF) testing (Humphrey Field Analyzer; Carl Zeiss Meditec, Inc., Dublin, CA, USA).

Patient Population (Histology)

We studied 11 eyes from 10 patients (one had a second operation for a recurrent MP). Patients were categorized into two groups based on postoperative VA: those with 20/60 or better were allocated to the "Good VA" control group, while those that were 20/200 or worse were allocated to the "Poor VA" group. In the poor ($\leq$20/200) postoperative VA group, there were four eyes (two MPs, one lamellar hole, one full-thickness MH [Gass stage II; International Vitreomacular Traction Study (IVTS)[42] classification, small full-thickness MH with broad vitreomacular traction]) from four patients (two men, two women) with an average age of 73 years (range, 62–86) at the time of surgery. In the "Good VA" control ($\geq$20/60) group there were seven eyes (five MPs, two full-thickness MHs [Gass stage III; IVTS classification, medium-sized full-thickness MH with broad vitreomacular traction]) from seven patients (three men, four women) with an average age of 75 years (range, 60–86) at the time of surgery.

All operations were performed within six months of symptom onset, and patients were followed for an average of 14 months (range, 10–23) in the poor VA group and 12 months (range, 4–23) in good VA controls. There was no statistically significant differences in age, sex distribution, or follow-up duration between the groups. Four patients underwent repeat surgery for recurrent disease. Cases 3, 5, and 6 had recurrent MP at 36, 8, and 5 months, respectively, after their initial operation. Notably, Case 6 is the same patient and same eye as Case 4. One patient (Case 11) had lamellar hole formation five months after surgery for a MP. Table 1 provides a detailed summary of patient clinical data.

Patient Population (Repeat Operation)

Reoperation was performed in cases where the MH reopened after dissipation of the intravitreal gas bubble ($N = 2$) and in cases when the MP either never resolved or recurred ($N = 8$). Ten eyes from 10 patients met these criteria and were categorized into two groups based on the time interval between surgeries. The group with $\geq$6 months between surgeries included four eyes from four patients (three male, one female) with an average age of 77 years (range, 58–91). The group with <6 months between surgeries included six eyes from six patients (three male, three female) with an average age of 70 years (range, 60–78). Patients were followed for an average of 9 months (range, 4–12) in the <6-month group and 7 months (range, 4–10) in the $\geq$6-month group. There was no statistical difference in age, sex distribution, or follow-up duration between the groups. Table 2 provides a detailed summary of the re-operated patient clinical data.

Surgery

Surgery consisted of a three-port, 25-gauge pars plana central and posterior vitrectomy. In primary (nonrecurrent) MP cases, either a pick or forceps were used to establish a surgical plane without the assistance of tissue staining. The ILM was not removed in primary MP cases. Indocyanine green (ICG) was not used in these cases. Chromodissection[8] was performed in all cases of MH and in recurrent MP using doubly-diluted (final concentration = 1.25 mg/mL) ICG (PULSION Medical Systems, Germany). A flexible, diamond-embedded "membrane scraper" (Synergetics, O'Fallon, MO, USA) was used to initiate a surgical plane. A macular membrane peel was performed from inferotemporal vascular arcade to superotemporal arcade and from the temporal margin of the optic disc to the temporal macula with forceps in all cases (Fig. 1). The MH surgeries concluded with an air–fluid exchange (AFX) performed at the nasal optic disc border with vitreous substitute using 16% C_3F_8 gas. Patients were instructed to maintain prone positioning until the bubble was gone, usually 2 to 3 weeks with liberalization toward the end of that time period.

Tissue Processing and Immunohistochemistry (IHC)

Immediately after surgical removal, dissected membranes were placed into 10% neutral buffered formalin for initial fixation. Tissues remained in fixative for at least 2 months (mean, 9.8; range, 2–19) before further processing. All tissues (11) were processed for and embedded into paraffin. The tissue blocks were oriented perpendicular to the cutting face of each paraffin block. Cross-sections were cut serially at 5 μm through the entire sample with a retracting microtome. Four tissue sections were placed on individual electrostatically charged glass microscope slides, resulting in 20 μm of tissue per slide (average number of slides = 55; range, 34–100). Each slide was labeled with a consecutive numerical value that allowed for easy identification of the start, middle, and end of a particular membrane. Tissues were sampled at approximately every 20th slide. This method allowed for representative serial analysis of the entire membrane. Human spinal cord samples were used as a positive control.

Deparaffinization and rehydration were performed using xylene, descending grades of ethanol, and distilled water baths.

Table 1. Patient Clinical Characteristics, Surgical Details, and Histopathologic Findings

Case No.	Age	Sex	Eye	Diagnosis	Repeat Operation?	ICG Use	Initiation Tool	Initiating Location*	Preop BCVA	Postop BCVA	Follow-Up, mo	IHC for NF
1	60	M	L	MP	No	No	Forceps	Inferotemporal	20/50	20/20	5	Negative
2	75	F	R	MP	No	No	MVR Blade	Nasal	20/80	20/20	23	Negative
3	77	M	L	MP	Yes	Yes	DDMS	Inferior	20/40	20/30	8	Negative
4†	86	M	L	MP	No	No	Forceps	Superior	20/50	20/40	4	Negative
5	73	F	L	MP	Yes	Yes	DDMS	Inferior	20/80	20/60	19	Negative
6†	86	M	L	MP	Yes	Yes	DDMS	Inferior	20/40	20/200	13	Positive
7	72	M	L	MP	No	No	Pick	Superotemporal	20/50	CF 2′	10	Negative
8	78	F	R	MH - Stage 3‡	No	Yes	DDMS	Inferotemporal	20/80	20/40	8	Negative
9	73	F	R	MH - Stage 3‡	No	Yes	DDMS	Inferior	20/100	20/40	13	Negative
10	62	F	R	MH - Stage 2‡	No	Yes	DDMS	Inferior	20/60	CF 4′	23	Positive
11	73	F	R	LH	Yes	Yes	DDMS	Not specified	20/140	CF 1′	11	Positive

F, female; M, male; R, right eye; L, left eye; LH, lamellar hole; MVR, micro-vitreoretinal; DDMS, diamond dusted membrane scraper; BCVA, best corrected visual acuity; CF, count fingers; NF, neurofilament.

* Site of surgical plane initiation with respect to the macula.

† Cases 4 and 6 are from the same patient who underwent reoperation due to a recurrent MP.

‡ Air–fluid exchange performed at the nasal optic disc.

Table 2. Patient Clinical Characteristics, Reoperation

Case No.	Age at First Sx	Sex	Eye	First Op Dx	Preop BCVA	Postop BCVA	Months Between Sx	Second Op Dx	Preop BCVA	Postop BCVA	Follow-Up, mo
A	59	M	L	MH	20/50	20/25	3	MH	20/100	20/50	15
B	86	M	L	MP	20/40	20/40	5	MP	20/40	20/200	6
C	73	F	R	MP	20/100	20/140	5	MH	20/140	CF 1′	11
D	91	M	R	MP	20/40	20/60	5	MH	20/60	HM	4
E	72	F	L	MP	N/A	N/A	8	MP	20/80	20/60	9
F	60	F	L	MP	20/80	20/40	11	MH	20/400	20/30	6
G	78	F	R	MH	20/80	20/40	13	MH	20/60	20/50	5
H	65	M	L	MP	20/60	20/60	22	MH	20/100	20/60	4
I	74	M	L	MP	N/A	N/A	42	MP	20/40	20/25	8
J	73	M	R	MP	N/A	20/140	69	MH	20/140	20/140	3

Op, operation; Dx, diagnosis; Sx, surgery; HM, hand motion; N/A, not available.

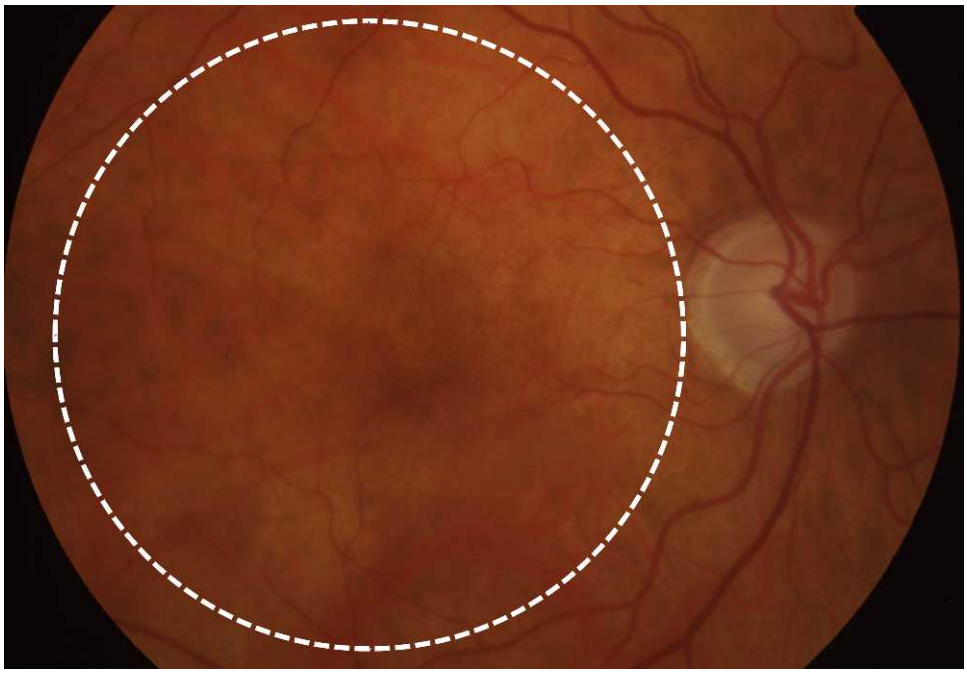

Figure 1. Color fundus photo of the right eye from a patient without any ocular pathology. The circular area enclosed by the *dashed white line* indicates the approximate extent of a membrane peel surgery.

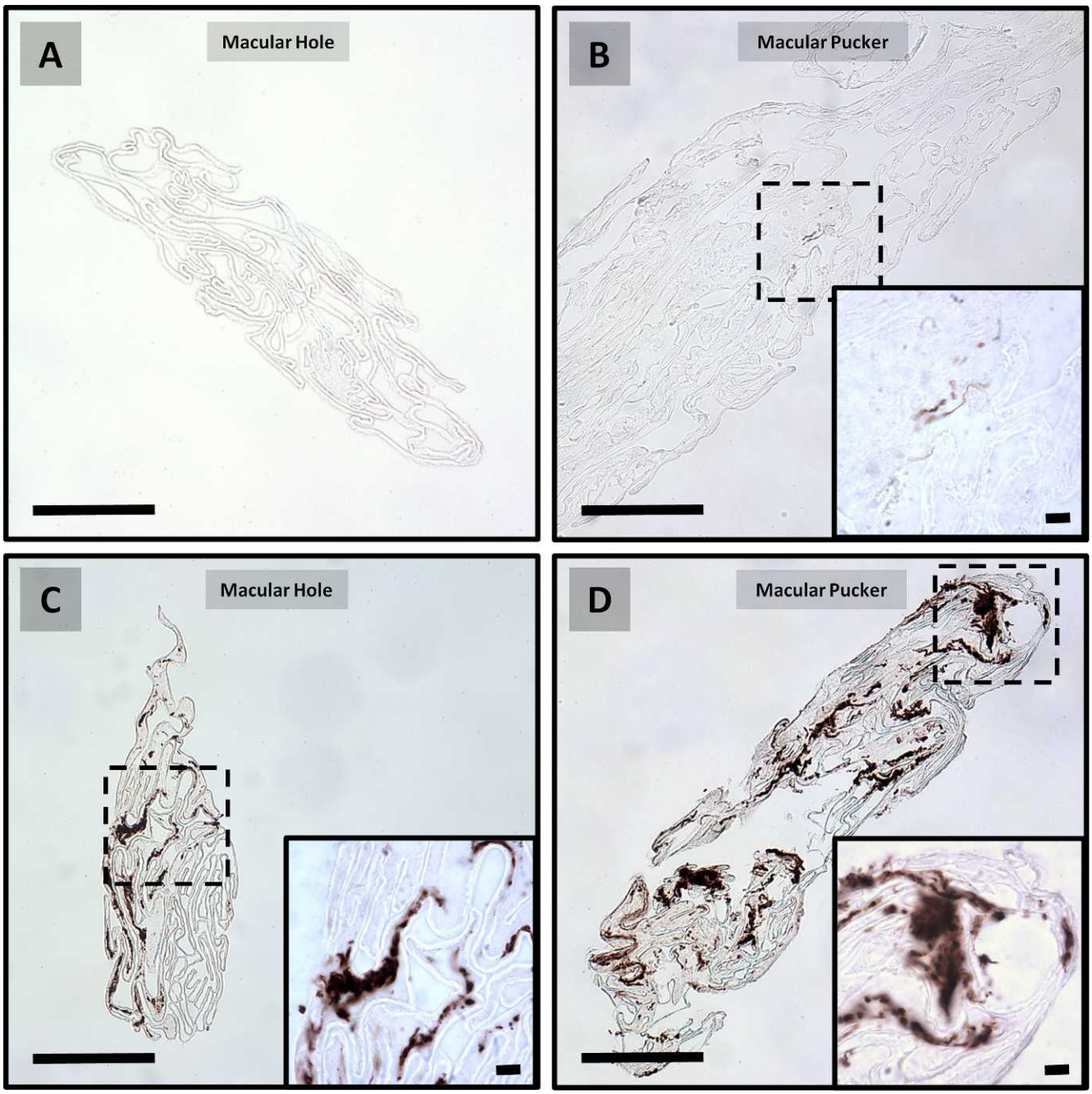

FIGURE 2. Bright field light microscopic images of membrane peel surgery specimens after immunohistochemistry using anti-neurofilament antibody. The DAB chromogen turned areas of positive antigen activity *brown*. No counterstain was used in the specimens to enhance contrast. An *inset* at a higher magnification is included in images where positive neurofilament is noted. *Main image large scale bars*: 50 μm. *Inset image small scale bars*: 5 μm. (**A**) An MH specimen revealing no evidence of positive staining. (**B**) An MP specimen revealing minimal positivity for neurofilament, (**C**, **D**) An MH and MP specimen, respectively, revealing diffuse evidence of neurofilament present on the membrane.

Antigen retrieval was performed using ×1 sodium citrate buffer solution while heating under pressure in a microwave oven. After antigen retrieval, the tissues were washed in ×1 wash buffer (0.05 mol/L Tris/HCl, 0.15 mol/L NaCl, 0.05% Tween 20, pH 7.6). To reduce background staining for endogenous peroxidase, 3% hydrogen peroxide was used for 10 minutes. Rabbit anti-human primary antibody for neurofilament protein was diluted to the manufacturer's recommendation of 1:100 using an antibody diluent with background reducer and subsequently incubated on the tissue sections for one hour. Horseradish peroxidase conjugated to a goat anti-rabbit secondary antibody then was applied to the sections and allowed to incubate for one-half hour. The brown enzyme product then was developed using 3,3′-diaminobenzidine (DAB) as the chromogen for five minutes. No counterstaining was performed to maximize contrast of the immunostain if present. The sections then were dehydrated with ascending grades of alcohol, cleared in xylene, and cover-slipped using permanent mounting media. All immunohistochemical reagents used were from a single manufacturer (Dako North America, Inc., Carpinteria, CA, USA).

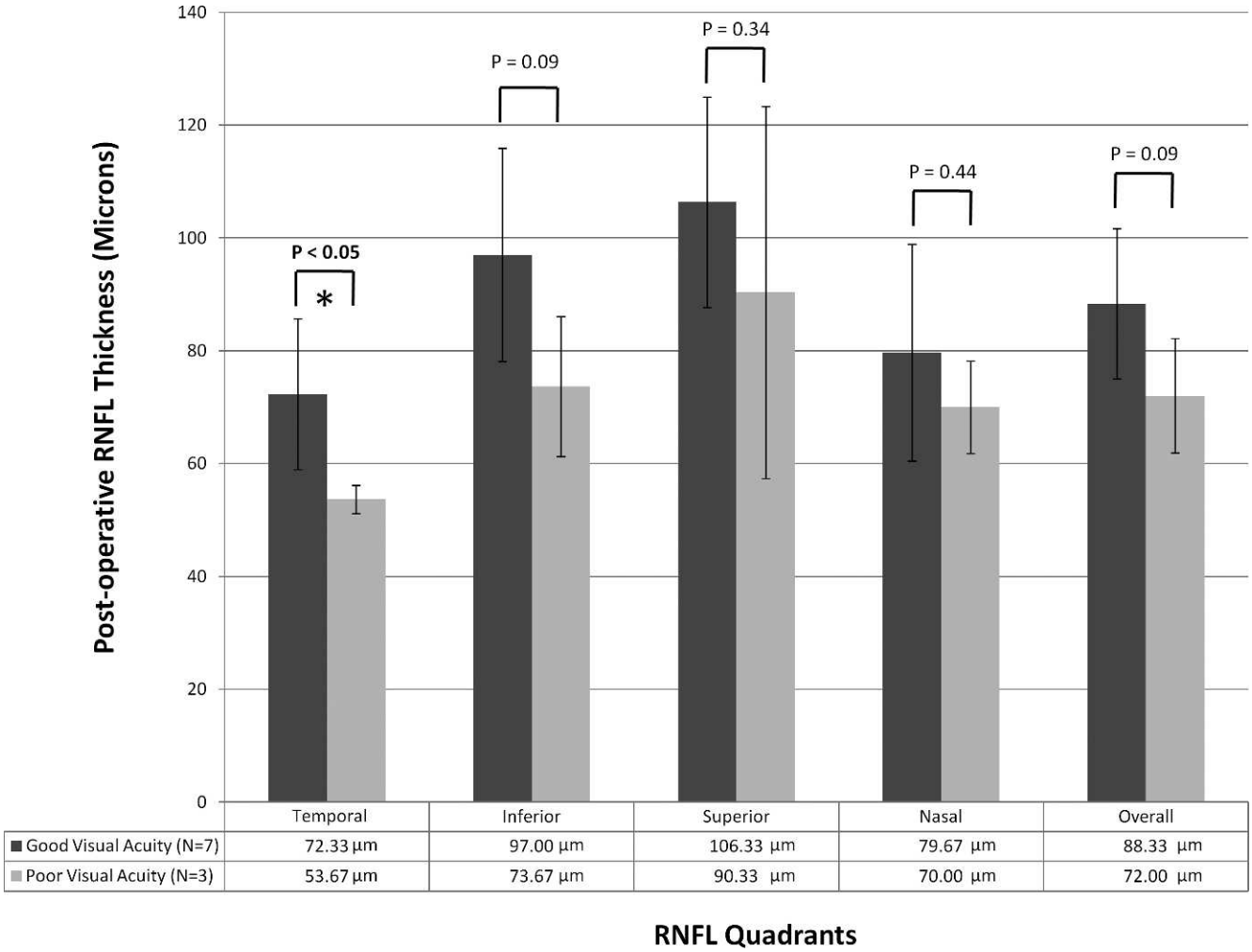

FIGURE 3. Bar graphs comparing postoperative RNFL thickness as measured in microns (*y*-axis) in eyes with good versus poor postoperative visual acuity. The *x*-axis displays the comparisons in different quadrants, as well as an aggregate RNFL thickness. *Error bars* represent standard deviation. One patient in the poor postoperative visual acuity category was lost to follow-up (Case 10).

Immunostained tissue sections were viewed with a Zeiss Axioskop brightfield light microscope (Carl Zeiss Microscopy, LLC., Thornwood, NY, USA) and images were captured on a Spot II digital camera (Diagnostic Instruments, Inc., Sterling Heights, MI, USA). Images were analyzed and categorized based on a dichotomous classification system. Neurofilament staining was considered "negative" if the majority of sections had no evidence (Fig. 2A), or only weak (Fig. 2B) staining. If the majority of sections from a particular specimen were noted to have strong staining (Figs. 2C, 2D), it was characterized as "positive."

RESULTS

Clinical Parameters

Patients with good postoperative VA had an average preoperative decimal VA = 0.32 ± 0.11 (~20/60), which improved to an average postoperative decimal VA = 0.64 ± 0.26 (~20/30, P = 0.02). Patients with poor postoperative VA had an average preoperative decimal VA = 0.34 ± 0.15 (~20/60), and an average postoperative decimal VA = 0.03 ± 0.05 (~20/600, P = 0.07). Table 1 provides a listing of the individual VA measurements.

The RNFL thickness measurements were obtained postoperatively in nine patients (Fig. 3); one patient in the poor VA group was lost to follow-up (Case 10). In the good VA group, the average thickness and standard deviation of the temporal, inferior, superior, and nasal quadrants were 72.33 ± 13.37, 97.00 ± 18.87, 106.33 ± 18.65, and 79.67 ± 19.23 µm, respectively, with an overall peripapillary thickness of 88.33 ± 13.34 µm. In the poor VA group, the measurements were 53.67 ± 2.52, 73.67 ± 12.42, 90.33 ± 32.96, and 70.00 ± 8.19 µm, respectively, with an overall peripapillary thickness of 72.00 ± 10.15 µm. There was a statistically significant difference in the temporal quadrant thickness between groups (P = 0.05). However, no such difference was detected in the inferior, superior, nasal, or overall thickness measurements.

Neuro-ophthalmology consultation was sought for all patients noted to have postoperative optic atrophy (Cases 5, 6, 7, 10, 11). Case 11 refused further evaluation. In case 5, the affected eye had only mild dyschromatopsia with 6/8 color plates, and a subjective reduction of brightness sense to 75% of normal. Cases 6, 7, and 10 all had 0/8 color plates in the affected eye, and brightness sense was reduced to 50%, 25%, and 10% of normal, respectively. A Humphrey 30-2 Stimulus III visual field test was obtained in each instance, revealing a predominantly superonasal defect sparing fixation in Case 5, a superonasal defect involving fixation in Case 6, a central

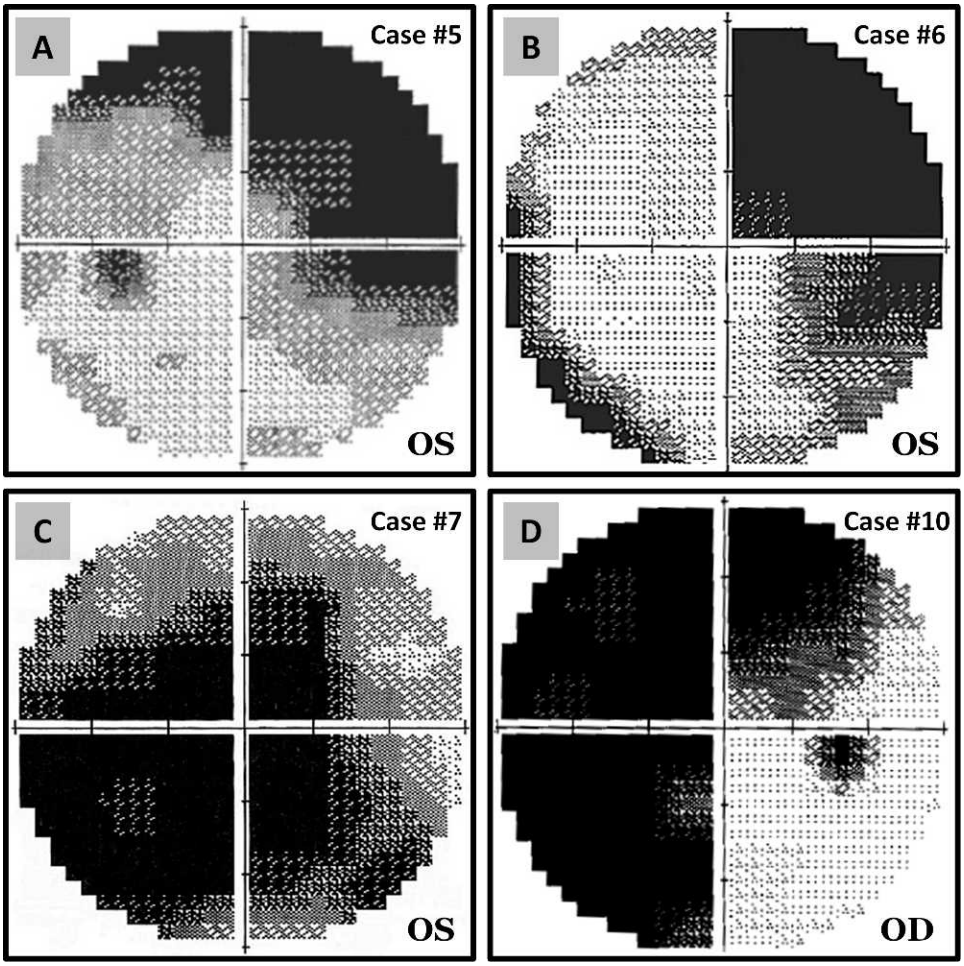

FIGURE 4. Humphrey Visual Field 30-2 Stimulus III testing results. (**A**) The left eye from Case 5 who had a good postoperative visual acuity (20/60). Note the sparing of central fixation, and a scotoma localized predominantly in the superonasal quadrant. (**B**) The left eye from Case 6 who had a poor postoperative visual acuity (20/200). The scotoma again is predominantly superonasal; however, fixation is affected in this patient. (**C**) The left eye from patient 7 who had a poor postoperative visual acuity (count fingers at 2 feet). There is a large central scotoma with diffuse depression, especially severe in the inferotemporal quadrant. (**D**) The right eye from Case 10 who had a poor postoperative visual acuity (count fingers at 4 feet). The scotoma is predominantly nasal, with some crossover temporally in the superior aspect. OS, left eye; OD, right eye.

scotoma with generalized depression in Case 7, and a nasal hemi-field loss involving fixation in Case 10 (Fig. 4).

Histology

In the primary MP cases (1, 2, 4, 7 in Table 1) there was no evidence of ILM and neurofilament staining was negative. In the good VA group, none (0 of 7) of the specimens had positive neurofilament staining (strong staining in the majority of processed specimens). In the poor VA group, three of four (75%) specimens had positive neurofilament staining, and, indeed, had strong neurofilament presence in all processed sections. Case 7 did not have evidence of neurofilament staining. The difference in positive neurofilament staining between the good (0/7) and poor (3/4) VA groups was significant ($P = 0.03$).

Repeat Operation

Reoperation was performed for failed surgery or recurrent pathology in 10 eyes. The duration of time between operations ranged from 3 to 69 months. In the <6-month group, three of four (75%) patients had worsening of VA after reoperation and one of four (25%) remained stable or improved. In the ≥6-month group, 6 of 6 patients (100%) either remained stable or improved ($P = 0.03$). In the <6-month group, patients had an average (± SD) preoperative decimal VA = 0.29 ± 0.16, which decreased to an average postoperative decimal VA = 0.13 ± 0.19. In the ≥6-month group, patients had an average preoperative decimal VA = 0.24 ± 0.15, which improved to an average postoperative decimal VA = 0.45 ± 0.24. There was a significant difference in VA between the two groups postoperatively ($P = 0.03$), though there was no difference in preoperative VA between the two groups ($P = 0.63$). Table 2 provides a listing of the individual VA measurements.

The RNFL thickness measurements were obtained following repeat surgery in nine patients (Fig. 5); one patient in the ≥6-month group was lost to follow-up (Case J). In the <6-month group, the average thickness and standard deviation of the temporal, inferior, superior, and nasal quadrants were 53.75 ± 8.42, 80.50 ± 10.38, 86.75 ± 27.20, and 74.50 ± 8.06 μm, respectively, with an overall peripapillary thickness of 73.75 ± 7.41 μm. In the ≥6-month group, the measurements were 72.60 ± 13.26, 87.80 ± 19.15, 103.60 ± 7.02, and 85.20 ± 24.69 μm, respectively, with an overall peripapillary thickness of 87.00 ± 14.95 μm. There was a significant difference in the temporal quadrant between groups ($P = 0.04$). However, no such difference was detected in the inferior, superior, nasal, or

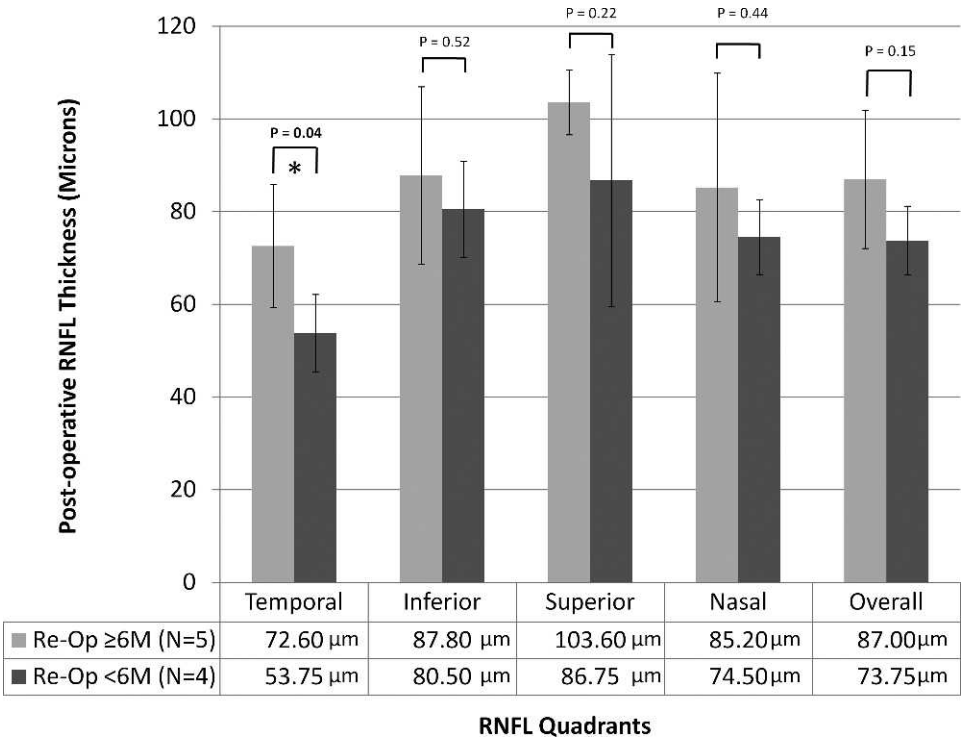

Figure 5. Bar graphs comparing postoperative RNFL thickness as measured in microns (*y*-axis) in eyes with that required reoperation for recurrent or persistent pathology. The *x*-axis displays the comparisons in different quadrants, as well as an aggregate RNFL thickness. *Error bars* represent standard deviation. One patient in the poor postoperative visual acuity category was lost to follow-up (Case J).

overall thickness measurements. The RNFL measurements were taken an average of 4.8 months after the repeat surgery in the <6-month group (range, 4–6), and an average of 5.4 months in the ≥6-month group (range, 3–7).

Immunohistochemistry was performed in a subset of patients (6 of 10) who had tissue specimens available after the repeat surgery. In the <6-month group, four of four cases (100%) had evidence of strong neurofilament staining in all processed sections, compared to none (zero of two) in the ≥6-month group.

Discussion

Most previous studies on the pathogenesis of visual field defects following MH and MP surgery drew conclusions that were based solely on clinical findings without histological evidence.[22,25,26,30,32] Among these were OCT studies that identified a reduction in the ganglion cell layer (GCL) following vitrectomy with ICG chromodissection for MHs.[43] Interestingly, another study found no difference in GCL thickness between chromodissection with ICG or Brilliant Blue G.[44] In cases where ultrastructural analysis was performed, no firm conclusions could be reached due to an inability to adequately identify the nature of cellular debris found on excised membranes.[45–47] The present study used IHC to determine that in cases with poor clinical outcomes, the heretofore unidentified elements apposed to the posterior aspect of surgically excised membranes are elements of the RNFL. The IHC targeting neurofilament revealed positive amounts of staining in three of four (75%) cases with poor visual outcomes (postoperative VA $\leq$ 20/200), compared to zero of seven patients with good (postoperative VA $\geq$ 20/60) outcomes ($P = 0.02$, Table 1). Neurofilament staining, being a marker for axonal elements,[48,49] confirms the suspicion that a mechanical disruption of the RNFL is the most likely explanation for the visual defects in these patients.

On clinical evaluation, five patients (four of four with poor visual outcomes, one of seven with a good visual outcome) were noted to have evidence of new onset optic neuropathy. Four of five patients underwent neuro-ophthalmology consultation, which substantiated the finding by the presence of all three signs of optic nerve dysfunction: afferent pupillary defect (APD), reduction in brightness sense, and dyschromatopsia. One patient (Case 11) chose not to pursue further evaluation. In light of the histopathologic evidence of RNFL damage associated with optic nerve dysfunction, we propose the term "inner retinal optic neuropathy" (IRON) for the iatrogenic vision abnormalities that can occur after vitreomacular surgery with membrane peeling. This study did not compare cases of membrane peeling with and without chromodissection, although it would seem that the present methodologies would be appropriate to undertake such a comparison. Further, it should be noted that cases in which a premacular membrane (also called epiretinal membrane [ERM]) alone is dissected, there is little of risk of IRON, unless the ILM is inadvertently peeled along with the premacular membrane. On the other hand, the risk of IRON is a consideration when the ILM is peeled either in a primary or secondary surgical intervention.

Visual field testing revealed a predominantly nasal field defect in three of four patients (Cases 5, 6, 10) and a central scotoma with generalized depression (Case 7) in the fourth (Fig. 4). The involvement of central fixation in Cases 6, 7, and 10 explains their poor postoperative VA, while the sparing of the central field in Case 5 explains the relatively good VA. Based on the trajectories of nerve fiber bundles, one would expect a nasal field loss to be associated with a thinning of the superior and/or inferior RNFL. On the other hand, a loss of central fixation would involve the papillomacular bundle (PMB), which is located at the temporal aspect of the optic

disc.[50] Indeed, from the present study population, a significant thinning of the PMB was evident in the poor VA group (Fig. 3). Loss of the inferior fibers in the poor VA group also is suggested, but a relatively small sample size makes it difficult to detect this smaller difference. Similar cases of RNFL thinning associated with visual field defects after MH surgery have been reported by Yamashita et al.[49] and Iriyama et al.[51,52] The visual field defects seen in affected patients were likely central field defects due to a reduction in their VA, a function of primary gaze. The HVF available in a subset of the patient population supports this theory (Fig. 4). Visual field loss following vitrectomy with membrane peeling has been described previously[53] and theorized to result from dehydration injury to the inner retina.[54] Strategies to mitigate this with humidification have been proposed.[55]

While membrane peel surgery has great success rates in treating MH and MP, there are times when the initial pathology does not resolve after one surgery,[33,34,56] and other times when the pathology recurs months to years later.[14,15,35–37] In these situations, a second membrane peel surgery often is warranted. Since repeat surgeries are not commonplace, the available literature is rather limited. However, the studies that do exist seem to reveal that the final VA after repeat surgery is not as good as after a single surgery.[38–41] Results from the present study suggested that the time interval between a second membrane peel surgery can have significant effects on postoperative outcome. The present study found that reoperating too soon (<6 months) after an initial surgery can lead to much worse results (postoperative decimal VA of 0.13 ± 0.19). On the other hand, waiting ≥6 months before reoperation is associated with excellent functional outcomes (postoperative decimal VA of 0.45 ± 0.24, $P = 0.03$). Furthermore, it is noteworthy that immunostaining for neurofilament protein revealed significant staining exclusively in the patients who underwent repeat surgery < 6 months after the first procedure. One potential explanation for the difference in visual acuity outcomes between the ≥6- and the <6-month groups is that the former may experience less RNFL damage due to protection afforded by a reformed Müller cell layer. Nakamura et al.[57] studied a chimpanzee model of membrane peel surgery, in which they enucleated the eye postoperatively and examined the retinal interface at progressive time points. In their study, the reformation of a Müller footplate border over the denuded RNFL first occurred at approximately the 6-month mark. Furthermore, it should be noted that in the present study there was no correlation between visual outcomes and time between surgeries (Pearson correlation coefficient, $R = 0.185$; $P = 0.61$). This would suggest that the visual outcomes did not continue to improve the longer the duration between the two surgeries, and that a maximum period of time to wait can be established. It would appear that based on this animal model and our present clinical findings, 6 months is the critical period of time. Another explanation, however, is that less aggressive recurrence of pathology made it unnecessary for the surgeon to intervene as early, and, therefore, preoperative factors (before reoperation) resulted in the better visual acuity outcomes.

In conclusion, the present findings support the hypothesis that iatrogenic disruption of the RNFL during membrane peel surgery for MH or MP can be associated with a poor functional outcome. In addition to nasal and central visual field defects, affected patients exhibit evidence of an optic neuropathy, including optic disc pallor, reduced brightness sense, dyschromatopsia, and an APD. In light of these findings, the term "inner retinal optic neuropathy" (IRON) is proposed to brand this entity. To avoid IRON, care should be taken to avoid overly aggressive peeling of the ILM, especially in the papillomacular bundle. In cases of requisite repeat membrane peel surgery, the risk of IRON is heightened if the inner retinal surface has not had adequate time to heal and regenerate a Müller cell basement membrane (ILM) barrier. To reduce the risk of reoperation-associated damage, a minimum interval of six months might be advisable between consecutive membrane peel operations. While the results of this relatively small study should be considered highly suggestive, future studies with larger numbers are needed to confirm these findings before such policies are enacted.

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