Discriminatory Power of Superficial Vessel Density and Prelaminar Vascular Flow Index in Eyes With Glaucoma and Ocular Hypertension and Normal Eyes

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Purpose. We evaluate the ability of optical coherence tomography angiography parameters, such as the peripapillary vessel density of the superficial retina and prelaminar flow index of the optic disc (PLFI), to differentiate primary open-angle glaucoma (POAG) and ocular hypertension (OH) from normal eyes.

Methods. The vessel density, PLFI, mean deviation of the visual field, circumpapillary retinal nerve fiber layer thickness (cpNFLT), and global loss volume of the ganglion cell complex were evaluated in one eye of 105 subjects with POAG and OH and normal eyes. The discriminatory powers of these parameters were evaluated based on the area under the curve (AUC) of the receiver operation characteristic curve and multiple comparisons.

Results. The vessel density ($P < 0.001$) and PLFI/unit area (PLFI/UA; $P = 0.020$) in eyes with POAG were significantly less than in normal eyes. The vessel density in eyes with OH was significantly ($P = 0.018$) reduced, whereas the PLFI/UA, global loss volume and cpNFLT were unaffected. The AUCs of the vessel density to discriminate glaucoma and OH from normal eyes were 0.832 and 0.724, respectively, and were significantly better than the PLFI/UA, in which the AUCs were 0.662 ($P = 0.002$) and 0.569 ($P = 0.038$), respectively. The powers of the vessel density and PLFI/UA to discriminate POAG from normal eyes were inferior to the global loss volume ($P = 0.006$ and <0.0001) and cpNFLT ($P = 0.055$ and $P < 0.0001$, respectively).

Conclusions. The vessel density and PLFI/UA decreased significantly in glaucomatous eyes. The vessel density was more efficient than the PLFI/UA for differentiating glaucoma and OH from normal eyes.

Keywords: OCT angiography, vessel density, vascular flow index, glaucomatous optic neuropathy, autoregulation

Vascular insufficiency is a primary clinical feature of glaucomatous optic neuropathy.[1] Some reports have been published on low vascular flow index (mean decorrelation) and peripapillary vessel density in glaucomatous eyes.[2–4] In previous optical coherence tomography angiography (OCTA) studies, the areas under the curve (AUCs) of the vascular flow index and vessel density ranged from 0.82 to 0.892 and 0.80 to 0.94, respectively.[2–4] Therefore, the OCTA parameters of vascular insufficiency might be useful for differentiating glaucomatous from normal eyes.

In other reports, the peripapillary vessel densities decreased at the corresponding visual field defect in nonhighly myopic and highly myopic glaucomatous eyes.[5] Prelaminar blood perfusion also was reduced in glaucomatous eyes.[6] However, the statistical significance of the OCTA parameters has not been compared to structural parameters, such as the ganglion cell complex or circumpapillary retinal nerve fiber layer thickness (cpNFLT). In the current report, we evaluated the discriminatory power of parameters, such as the peripapillary vessel density of the superficial retina (VD), vascular flow index at the prelaminar region (PLFI), signal strength (SS), cpNFLT, and global loss volume (GLV) of the ganglion cell complex in primary open-angle glaucoma (POAG), ocular hypertension (OH), and normal eyes.

Methods

This was a prospective case-controlled study. From October 2014 to July 2016, one eye of 105 consecutive subjects (66 with POAG, 14 with OH, and 25 normal subjects) was enrolled. Primary open-angle glaucoma was diagnosed based on the gonioscopic open angle, signs of glaucomatous optic neuropathy using ophthalmoscopy and optical coherence tomography (OCT), glaucomatous visual field defects using the Humphrey Field Analyzer (750i; Carl Zeiss Meditec, Tokyo, Japan), and the absence of diseases that cause optic nerve atrophy. Subjects with OH had an open angle and a history of at least two repeated high IOP values exceeding 21 mm Hg but did not have glaucomatous visual field defects (normal glaucoma hemifield test and pattern standard deviation less than 95% by the Humphrey Field Analyzer) or glaucomatous optic neuropathy. Subjects with OH could use topical antiglaucoma eye drops. We

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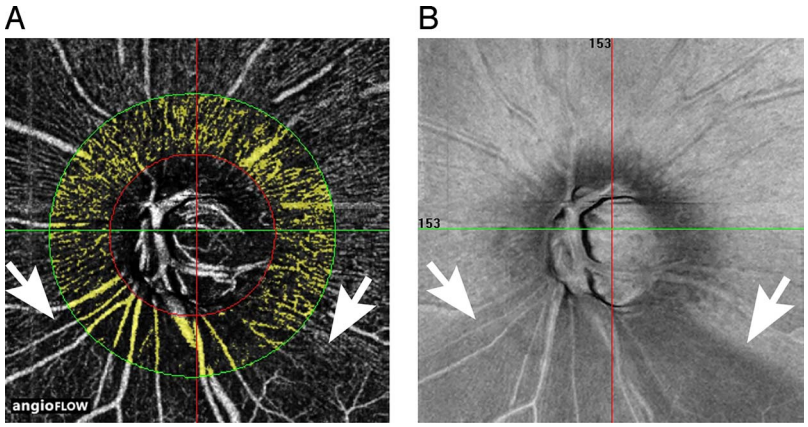

FIGURE 1. (**A**) The highlighted RPC of the superficial retina and (**B**) an en face image of the retinal nerve fiber layer defects (between *arrows*) in an eye with POAG. In this image, there is a defective RPC between the *arrows* and a corresponding retinal nerve fiber layer defect between the *arrows*. In this case, the tissue depth is between 0 and 80 μm, and the highlighted area is 700 μm from the disc margin (size, 4.5 × 4.5 mm).

did not include subjects with preperimetric glaucoma in whom signs of glaucomatous optic neuropathy were detected by OCT or assessment of retinal nerve fiber layer defects in eyes with no sign of glaucomatous visual field defects. Normal subjects had an IOP below 21 mm Hg and did not have glaucomatous optic neuropathy or visual field defects detected by the Humphrey Field Analyzer. Normal subjects were excluded who had a history of intraocular surgery, diabetes mellitus, high systemic hypertension exceeding 160/90 mm Hg, used alcohol within 6 hours, used cigarettes or coffee within 3 hours before examination, and used oral drugs that affect vessel diameter.

Patients with glaucoma or OH were outpatients who had not undergone a previous glaucoma surgery, retinal surgery, or photocoagulation, and were studied after successful medical control of the IOP between 10 and 21 mm Hg.

For each all enrolled patients, we recorded their age, spherical equivalent refractive error (SQRE), best-corrected visual acuity, gonioscopy, slit-lamp microscopy, Goldmann IOP, and mean deviation (MD) and pattern standard deviation of the Humphrey 24-2 visual field test.

Other exclusion criteria included eyes with cloudy media, epiretinal membrane, vascular diseases, iridocyclitis, congenital or acquired optic nerve atrophy, highly deformed optic disc, wide myopic peripapillary choroidal atrophy, refractive errors exceeding −9 and +4 diopters (D), amblyopia, POAG in the fellow eye, visual acuity less than 20/40, OCT angiographic poor-quality images (doubling of vessel image and artifact lines), and a SS less than 40. If both eyes were eligible, the right eye was selected for the study.

OCT Study

Enrolled eyes were studied using spectral-domain OCT equipped with angiographic function (AngioVue, RTVue XR OCT; Optovue, Fremont, CA, USA) with the 2014.2.0.90 software version. The details of the device have been reported previously.[7] In the current study, we evaluated 4.5 × 4.5-mm images. The parameters studied were the GLV of the ganglion cell complex, cpNFLT, SS, optic disc size using the RTVue algorithm, VD (0–80 μm from the internal limiting membrane) within 700 μm of the disc margin, vascular flow index in the cylindrical column between 50 and 250 μm from the surface of the disc that included the PLFI. The wavelength of the device was 840 nm and the vascular flow index reflects a mean decorrelation of the tissue included.[8] Figure 1 shows an example of highlighted superficial vessels and an en face image

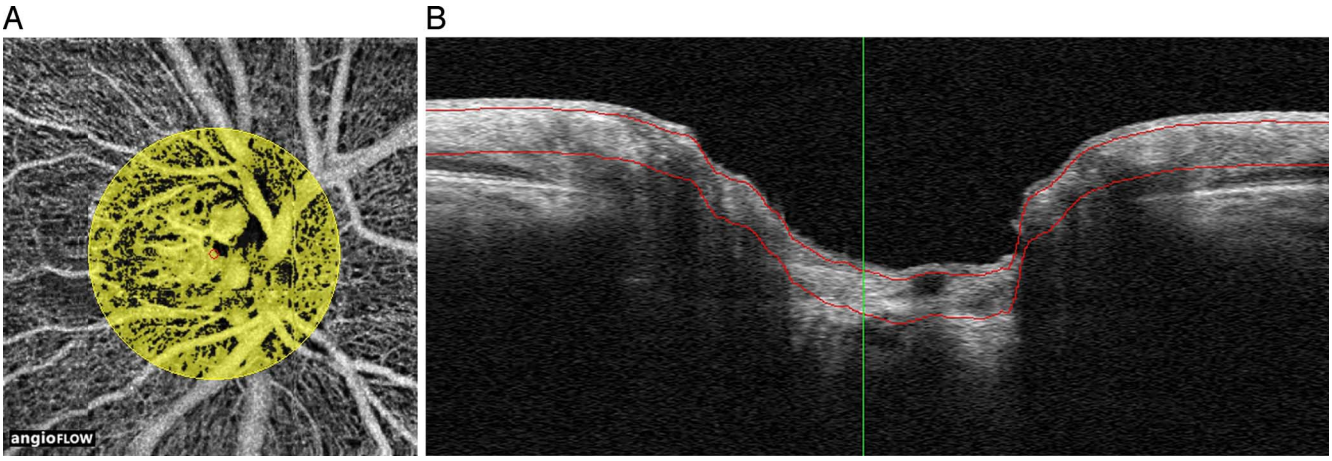

FIGURE 2. (**A**) An example of highlighted prelaminar vessels in a normal eye. The vascular flow index of the prelaminar area is calculated by measuring the mean decorrelation in the column between 50 and 250 μm deep within Elschnig's scleral ring. (**B**) In the sagittal section image of the same optic nerve head, a large part of the prelaminar region is included between the two *red lines* 50 and 250 μm from the disc surface (size, 3 × 3 mm).

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Table 1. Demographic Data From Normal, POAG, and OH Eyes and Multiple Comparisons Using the Tukey's Test

Eyes	Age	IOP, mm Hg	SQRE, D	SS	LogMAR	DA
Normal, $n = 25$	56.2 ± 13.5	16.5 ± 2.9	−1.23 ± 2.81	66.0 ± 10.2	−0.110 ± 0.121	2.01 ± 0.36
POAG, $n = 66$	60.4 ± 12.7	18.0 ± 4.1	−4.06 ± 4.08	61.1 ± 9.4	−0.074 ± 0.167	1.92 ± 0.54
OH, $n = 14$	52.0 ± 13.5	18.6 ± 2.7	−4.32 ± 3.73	61.9 ± 12.1	−0.119 ± 0.097	1.87 ± 0.29
P value						
Normal vs. POAG	0.258	0.285	0.005#	0.050#	0.852	0.850
Normal vs. OH	0.742	0.237	0.041#	0.377	0.969	0.722
POAG vs. OH	0.093	0.803	0.982	0.910	0.593	0.899

DA, disc area in mm^2.
# $P < 0.05$ is significant.

of the retinal nerve fiber layer defect. The definition of the GLV has been reported previously.[9]

In the current study, the VD was defined as the percentage of signal-positive pixels/area of interest. For image acquisition, the default signal intensity to cut off the noise was set at 4.1 log arbitrary units.[10] To study the patient's eye, the threshold for detecting the vessel density of the radial peripapillary capillary (RPC) was set at 0.05. The algorithm to detect the disc margin has been reported previously.[10] Considering the possible waning of the signal strength by individual variations in media opacity and tissue vascularity, the VD/SS and PLFI/unit area (PLFI/UA)/SS also were evaluated.

To study the PLFI, an experienced technician marked the disc margin and the vessels in the cylindrical column 50 to 250 μm from the surface (Fig. 2) and the mean decorrelation was measured. To avoid the effect of disc size, the PLFI was divided by the measured area (in square millimeters), and the PLFI/UA was used for analysis (Fig. 2).

The internal review board of Sensho-kai Eye Institute approved the study design. All subjects provided informed consent after they received an explanation of the nature and possible consequences of the study. The study adhered to the tenets of the Declaration of Helsinki.

Statistical Analyses

For statistical analysis, Bell Curve Excel Tokei 2016 (SSRI, Tokyo, Japan) was used to calculate the Pearson's correlation coefficient, receiver operating characteristic curve (ROC), multiple comparisons, and stepwise regression analysis with backward elimination.

Two independent skilled technicians studied the examinations of five normal subjects repeated five times to determine the intravisit data reproducibility.

Results

Table 1 shows the demographic data from the normal subjects and patients with POAG and OH, and the results of Tukey's multiple comparisons among the three cohorts. A value of $P < 0.05$ was considered significant. There were no significant differences among the three cohorts except for higher myopia in the POAG and OH eyes ($P = 0.005$ and $P = 0.041$, respectively) and low SS in the POAG ($P = 0.05$). Eleven of 14 eyes with OH and 42 of 66 eyes with POAG were treated with topical eye drops.

Concerning data reproducibility, the coefficients of variation of the intravisit examinations for the SS, VD, and PLFI were 4.3 ± 1.6%, 3.4 ± 3.1%, and 6.3 ± 2.7%, respectively.

In Table 2, the differences in the MD, GLV, VD, VD/SS, and cpNFLT between the normal and POAG cohorts were significant ($P < 0.001$). The difference in the PLFI/UA also was significant between normal and POAG eyes; however, the *P* value for this comparison was low ($P = 0.020$, Table 2).

When these parameters in OH were compared to those in normal eyes, two OCTA parameters, the VD ($P = 0.018$) and VD/SS ($P = 0.018$), were significantly less in OH eyes, whereas no significant differences were found for the MD, GLV, PLFI/UA, and cpNFLT (Table 2). This reduced VD and VD/SS in OH eyes led to nonsignificant difference in these two parameters between OH and POAG eyes ($P = 0.243$, and 0.061, respectively) despite presence of significant differences in MD, GLV, and cpNFLT ($P < 0.001$). Among the OCTA parameters PLFI/UA was more resistant to damage by intraocular hypertension and glaucomatous insult. Even though the difference in PLFI/UA between OH and normal eyes did not reach significance ($P = 0.582$), the VD and VD/SS were significantly lower in the OH and POAG eyes than those in normal eyes (Table 2).

Table 3 shows the Pearson's correlation coefficients and the *P* values from univariate linear regression among 10 parameters evaluated in the 25 normal eyes. In the normal eyes, the SS was associated significantly with the VD ($P = 0.001$), PLFI/UA ($P < 0.001$), and age ($P = 0.015$). Other positive associations were found between the GLV and cpNFLT ($P < 0.001$), VD and PLFI/UA ($P = 0.003$), and PLFI and disc area ($P = 0.0254$, Table 3).

Table 4 shows the Pearson's correlation coefficients and *P* values for univariate linear regression among 11 parameters in

Table 2. Comparison of Functional and Structural Parameters Among Three Cohorts by Multiple Comparisons Using the Tukey's Test

Eyes	MD	GLV	VD, %	VD/SS	PLFI/UA	cpNFLT, μm
Normal, $n = 25$	−0.04 ± 0.23	2.80 ± 3.31	49.8 ± 8.4	0.76 ± 0.11	0.72 ± 0.09	97.5 ± 8.0
POAG, $n = 66$	−7.50 ± 7.45	20.67 ± 9.56	38.2 ± 8.3	0.62 ± 0.08	0.66 ± 0.10	73.7 ± 12.8
OH, $n = 14$	−0.64 ± 1.09	3.30 ± 3.77	42.1 ± 9.1	0.68 ± 008	0.69 ± 0.11	95.5 ± 9.1
P value						
Normal vs. POAG	<0.001#	<0.001#	<0.001#	<0.001#	0.020#	<0.001#
Normal vs. OH	0.949	0.980	0.018#	0.018#	0.582	0.850
POAG vs. OH	<0.001#	<0.001#	0.243	0.061	0.533	<0.001#

# $P < 0.05$ is significant.

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Table 3. Pearson's Correlation Coefficient (*P* Value) Among 10 Parameters in Normal Eyes

Parameters	SS	SQRE	logMAR	GLV	cpNFLT	VD	PLFI	PLFI/UA	Age	DA
SS		−0.198 (0.358)	0.318 (0.131)	−0.081 (0.711)	0.075 (0.729)	0.626 (0.001)	0.087 (0.691)	0.805 (<0.001)	−0.486 (0.015)	−0.183 (0.397)
SQRE			0.351 (0.093)	0.2 (0.352)	−0.035 (0.874)	0.035 (0.873)	0.239 (0.264)	−0.348 (0.097)	0.387 (0.061)	0.438 (0.031)
LogMAR				0.312 (0.139)	−0.228 (0.288)	0.341 (0.104)	0.116 (0.592)	0.13 (0.548)	0.177 (0.412)	−0.094 (0.664)
GLV					−0.727 (<0.001)	−0.202 (0.347)	0.065 (0.766)	−0.2 (0.354)	0.251 (0.240)	0.195 (0.364)
cpNFLT				##		0.109 (0.617)	0.096 (0.659)	0.187 (0.386)	−0.317 (0.132)	0.095 (0.662)
VD	##						0.208 (0.333)	0.572 (0.003)	−0.074 (0.733)	−0.101 (0.642)
PLFI								0.232 (0.280)	0.328 (0.118)	0.453 (0.025)
PLFI/UA	##					##			−0.399 (0.053)	−0.336 (0.109)
Age	#									0.261 (0.240)
DA		#					#			

# $P < 0.05$, significant association by univariate linear regression. ## $P < 0.01$, significant association by univariate linear regression.

Table 4. Pearson's Correlation Coefficient (*P* Value) Among 11 Parameters in 80 POAG and OH Eyes (66 Eyes With POAG; 14 Eyes With OH)

Parameters	SS	SQRE	logMAR	MD	GLV	cpNFLT	VD	PLFI	PLFI/UA	Age	DA
SS		0.287 (0.010)	−0.196 (0.079)	0.229 (0.039)	−0.192 (0.088)	0.262 (0.018)	0.839 (<0.001)	0.245 (0.027)	0.686 (<0.001)	−0.048 (0.672)	−0.106 (0.348)
SQRE	#		0.051 (0.658)	0.065 (0.572)	−0.079 (0.492)	0.148 (0.192)	0.225 (0.046)	−0.004 (0.969)	−0.073 (0.523)	0.440 (<0.001)	0.048 (0.672)
LogMAR				−0.378 (<0.001)	0.217 (0.053)	−0.245 (0.028)	−0.319 (0.004)	−0.225 (0.044)	−0.417 (<0.001)	0.280 (0.011)	−0.028 (0.806)
MD	#		##		−0.759 (<0.001)	0.681 (<0.001)	0.346 (0.002)	0.036 (0.751)	0.355 (0.001)	−0.050 (0.657)	−0.164 (0.143)
GLV				##		−0.866 (<0.001)	−0.340 (0.002)	−0.047 (0.678)	−0.262 (0.019)	0.164 (0.147)	0.094 (0.409)
cpNFLT	#		#	##	##		0.377 (<0.001)	0.068 (0.547)	0.332 (0.003)	−0.170 (0.130)	−0.121 (0.282)
VD	##	#	##	##	##	##		0.117 (0.298)	0.659 (<0.001)	−0.185 (0.098)	−0.250 (0.024)
PLFI	#		#						0.575 (<0.001)	−0.089 (0.432)	0.869 (<0.001)
PLFI/UA	##		##	##	#	##	##	##		−0.190 (0.090)	0.112 (0.320)
Age		##	#								−0.002 (0.983)
DA							#	##			

# $P < 0.05$ by univariate linear regression in the 80 eyes. ## $P < 0.01$, by univariate linear regression in the 80 eyes.

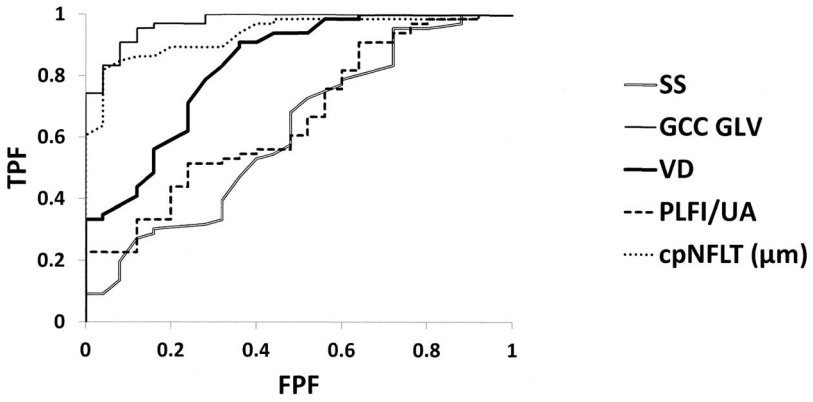

Figure 3. The ROC curve to discriminate POAG from normal eyes. The AUC is the largest for the GLV (0.974, $P < 0.001$) followed by the cpNFLT (0.936, $P < 0.001$), VD (0.832, $P < 0.001$), PLFI/UA (0.662, $P = 0.012$), and SS (0.619, $P = 0.085$). TPF, true positive fraction (sensitivity); FPF, false positive fraction (1 − specificity).

80 eyes with POAG ($n = 66$) and OH ($n = 14$). Vascular parameters, such as the VD and PLFI/UA, had significant univariate linear correlations with many parameters. Among them, the VD and PLFI/UA were correlated significantly with function parameters, such as the MD ($P = 0.002$ and $P = 0.001$, respectively) and structural parameters, such as the GLV ($P = 0.002$ and $P = 0.019$, respectively) and cpNFLT ($P < 0.001$ and $P = 0.003$). These findings suggested that the VD and PLFI/UA decreased in glaucomatous eyes and agreed with results of multiple comparisons (Table 2).

On the other hand, the SS decreased with advances in glaucomatous nerve loss and was associated with the SQRE, MD, cpNFLT, VD, PLFI, and PLFI/UA ($P = 0.010$, $P = 0.039$, $P = 0.018$, $P < 0.001$, $P = 0.027$, and $P < 0.001$, respectively). While, the refractive error was associated with the SS, VD, and age ($P = 0.010$, $P = 0.046$ and $P < 0.001$, respectively; Table 4).

When the cpNFLT was set as a dependent parameter and stepwise regression analysis with backward elimination was performed for eight independent parameters (SS, SQRE, logMAR, MD, GLV, VD, PLFI/UA, age), the GLV was associated most strongly ($P < 0.001$) with the cpNFLT. The acquired regression equation was cpNFLT = –0.093age + 0.435 SQRE – 1.10GLV + 14.78PLFI/UA + 94.37 (multiple coefficient of determination; $R^2 = 0.776$). The P values (and t values) for age, GLV, PLFI/UA, and SQRE and constant were $P = 0.199$ (–1.29), $P < 0.001$ (–13.95), $P = 0.079$ (1.78), $P = 0.067$ (1.86), and $P < 0.001$ (11.94), respectively.

When the powers of the SS, GLV, VD, PLFI/UA, and cpNFLT to discriminate POAG from normal eyes were studied by the ROC curves, there were significant differences among the five parameters. The AUC was largest for the GLV followed by the cpNFLT, VD, PLFI/UA, and SS. The AUCs and P values using the Sen and Delong test[11] were 0.974 ($P < 0.001$), 0.936 ($P < 0.001$), 0.832 ($P < 0.001$), 0.662 ($P = 0.012$), and 0.619 ($P = 0.085$), respectively (Fig. 3). The discriminatory power of the GLV was significantly stronger than that of the VD ($P = 0.006$), PLFI/UA ($P < 0.0001$), and SS ($P < 0.0001$), and marginally stronger than that of the cpNFLT ($P = 0.068$). The discriminatory power of the VD was stronger than that of the PLFI/UA ($P = 0.002$) and SS ($P < 0.0001$); however, it was less than that of the GLV ($P = 0.006$) and marginally less than cpNFLT ($P = 0.055$; Table 5).

When the powers of the five parameters to discriminate OH from normal eyes were studied, the AUC was the largest for the VD followed by the SS, PLFI/UA, cpNFLT, and GLV. The AUCs and P values were 0.724 ($P = 0.006$), 0.599 ($P = 0.388$), 0.569 ($P = 0.482$), 0.553 ($P = 0.613$), and 0.529 ($P = 0.784$), respectively (Fig. 4).

The VD was the only significant ($P = 0.006$) parameter that could discriminate OH from normal eyes (Fig. 4). The finding agreed with the multiple comparisons (Table 2), and its discriminatory power was significantly ($P = 0.038$) higher than that of the PLFI/UA (Table 6).

Table 5. Comparison of Discriminatory Power Among Five Parameters to Differentiate POAG From Normal Eyes by ROC Curves

Parameters	GLV	cpNFLT	VD	PLFI/UA	SS
GLV		$P = 0.068$	$P = 0.006^{\#}$	$P < 0.0001^{\#}$	$P < 0.0001^{\#}$
cpNFLT			$P = 0.055$	$P < 0.0001^{\#}$	$P < 0.0001^{\#}$
VD	#			$P = 0.002^{\#}$	$P < 0.0001^{\#}$
PLFI/UA	#	#	#		0.382
SS	#	#	#	#	

GLV has the strongest power to discriminate POAG from normal eyes followed by the cpNFLT, VD, PLFI/UA, and SS.

Significant difference.

Discussion

Jia et al.[10] and Wang et al.[2] previously reported reduced blood flow index in the entire optic disc and inferotemporal segment of the optic disc. The blood supply to the optic disc is divided into three parts; that is, the superficial layer in which the retinal arterial supply predominates, the prelaminar and laminar areas in which the short posterior ciliary artery supply predominates, and the retrolaminar area in which the vessels from the pia mater supply predominates.[12] Even though most vessels in front of the lamina cribrosa are autoregulated, disruptions in the autoregulation system, and fluctuations in the blood flow can occur in glaucomatous optic discs.[1] In experimental glaucoma, dysfunctions in autoregulatory mechanisms might lead to initial short-term increases and later decreases in the blood flow based on a laser speckle flow graphic study of the posterior optic nerve blood flow.[13] In this study, decreases in the VD in OH eyes occur in the superficial peripapillary retina and might reflect dysregulation of the blood flow in the superficial optic disc.

Large and small vessels are present in the optic disc, and small vessels might be susceptible to damage. Jia et al.[8] reported an attenuated blood flow index in the entire disc area and a "relatively unchanged" caliber of the large retinal vessels. In the OCTA studies, the interscan time is important to focus on the blood flow in the capillary level vessels.[14] The blood flow in the large vessels is fast compared to that in the capillary

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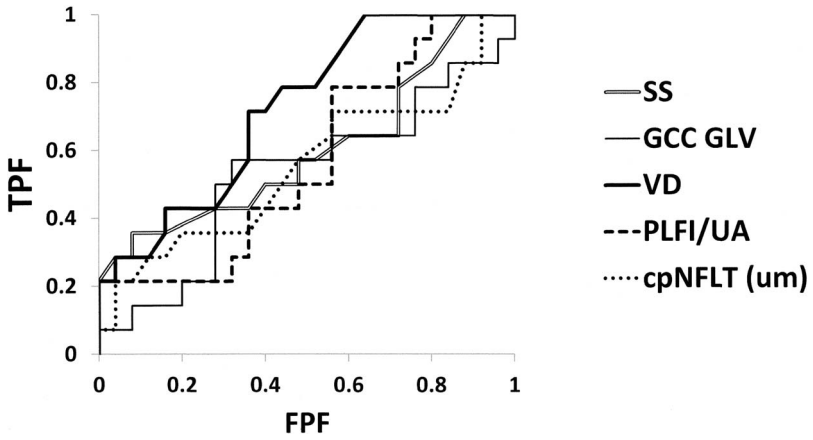

Figure 4. The ROC curve to discriminate OH from normal eyes. The AUC of the VD (0.724) is statistically significant ($P = 0.006$). However, the AUC of other parameters, such as the SS (0.599, $P = 0.388$), PLFI/UA (0.569, $P = 0.482$), cpNFLT (0.553, $P = 0.613$), and GLV (0.529, $P = 0.784$), are not significant. VD has significant power to discriminate OH from normal eyes; however, the other four parameters do not. This finding suggests damage to the RPC occurs in OH before structural changes in the GLV and cpNFLT.

level vessels, and the flow index of the large vessels will be saturated in the current OCTA system.[14] If the optimal interscan time to visualize small vessels was 1.5 to 3 ms, the interscan time of 5 ms in the AngioVue can lead to easy signal saturation from the large vessels.[13] If the signal from the large vessels is saturated and the caliber of the large vessels is relatively unchanged, the signals from the large vessels will not interfere with quantitative analysis of the flow index of the small vessels in the optic disc.

Bojikian et al.[6] focused on the prelaminar optic disc perfusion. To compensate for reduced tissue volume in glaucomatous eyes, they introduced the theory of "normalized flux (flux normalized by the vessel area)" and reported a significant association between the flux and MD and pattern standard deviation of the visual field, global average retinal nerve fiber layer thickness, cup-to-disc ratio, and rim area.[6] In glaucomatous eyes, the prelaminar tissue becomes atrophic and reduced in volume; thus, the total flow index might not reflect just local ischemia but local ischemia and loss of the neural tissue itself. To understand local ischemia, the flow index/unit volume is important. Therefore, we studied the flow index in the cylindrical column between 50 and 250 μm from the surface, which is sufficient to cover the prelaminar area (Fig. 2), and calculated the flow index/UA. The volume of the cylindrical column is a function of the measured area; thus, the PLFI/UA is proportional to the PLFI/unit volume. The thickness of the normal lamina cribrosa is 378 ± 117 μm,[15] and the current setting of the PLFI can include the anterior portion of the lamina cribrosa and might reflect vascular insufficiency of the small vessels in the prelaminar and laminar areas. When glaucomatous excavation of the disc progresses, increasingly more laminar tissue might be included in the 50 to 250-μm column. In this case, the difference between the blood flow in the lamina cribrosa and prelaminar area might be a concern. Geijer and Bill,[16] who used the microsphere technique in monkey eyes and studied blood flow at the lamina cribrosa and prelaminar area, found nearly equivalent blood flow/volume tissue in a perfusion pressure range of 60 to 100 mm Hg. Thus, differences in the vascular components of the laminar and prelaminar tissue might not be crucial to this study. Nevertheless, the effect of pathologic deformation of the laminar tissue on blood flow is unknown; this possibility must be elaborated in future studies.

Table 6. Comparison of Discriminatory Powers Among Five Parameters to Differentiate OH From Normal Eyes by ROC Curves

Parameters	GLV	cpNFLT	VD	PLFI/UA	SS
GLV		$P = 0.782$	$P = 0.126$	$P = 0.794$	$P = 0.641$
cpNFLT			$P = 0.174$	$P = 0.913$	$P = 0.752$
VD				$P = 0.038$[#]	$P = 0.055$
PLFI/UA			[#]		$P = 0.646$
SS					

The VD was stronger than the PLFI/UA to discriminate OH ($P = 0.038$).

[#] Significant difference.

In the current study, we confirmed that the PLFI/UA was significantly ($P = 0.020$) decreased in eyes with POAG (Table 2; AUC, 0.662). Therefore, it is plausible that vascular insufficiency of the small vessels occurs in the prelaminar region of the glaucomatous optic disc. This finding might agree with previous reports on preperimetric glaucoma using laser speckle flowgraphy[13,17] and the microsphere method.[13]

However, the discriminatory power of the PLFI/UA to differentiate eyes with POAG from normal eyes was weak compared to the VD (AUC 0.832, $P = 0.002$, Table 5). This finding suggested that radial peripapillary capillaries might be more susceptible to glaucomatous damage than deeper vessels in the optic disc. Akagi et al.[5] suggested that the superficial vessel density is a better indicator of nerve damage than the vasculature of the entire disc in high myopia. Recently, Chen et al.[18] reported that a superficial blood flux index is more susceptible to glaucomatous damage than vessel density. Even though vascular insufficiency might occur in the prelaminar area of glaucomatous eyes, a complex interaction among the ocular perfusion pressure, oxygen tension, vasoactive substances, extracellular matrix, and IOP might minimize ischemia and mask impaired blood flow in the prelaminar area of the glaucomatous optic disc.[1,19,20] In the current study, the reduced VD in eyes with POAG agreed with the results of previous reports.[3–5] However, it is not clear if this phenomenon reflects reduced RPC density alone or includes thinning of the major retinal arteries in eyes with POAG.[21,22]

Interestingly, the reduced VD was found in eyes with OH before any sign of GLV or cpNFLT damage, which was confirmed by multiple comparisons ($P = 0.018$; Table 2) and the ROC curve ($P = 0.006$; Fig. 4). In the ocular hypertension treatment study, the high IOP and splinter hemorrhage of the optic disc were risk factors for development of POAG.[23,24]

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Thus, dysregulation of blood flow had been suspected as a cause of glaucomatous optic neuropathy. The reduced VD before changes in the cpNFLT and GLV in eyes with OH might reflect disrupted autoregulation that was found in experimental nonhuman glaucoma eyes.[13,20] However, we must be careful that the question if reduced VD can lead to development of early nerve damage in eyes with OH or simply reflects vessel emaciation or diffuse RPC scattering is unanswered yet.

Another explanation for the reduced VD includes the effects of topical eye drops. In 11 of 14 eyes with OH, topical eye drops might have affected the superficial VD. Effects of drugs that can affect blood flow had been studied by laser speckle flowgraphy.[25]

In the current study, the IOPs of the patients were controlled to below 21 mm Hg, and impaired vascular flow at the prelaminar area might have been less significant compared to that associated with high IOP.

In a pilot study, Jia et al.[8] reported that a reduced blood flow index in the entire optic disc and cutoff line of 0.1515 (6% lower compared to the normal level) was useful to differentiate glaucomatous from normal eyes with 100% sensitivity and specificity. This sensitivity is better than the result in the current study in which the AUC was 0.662. This difference might be attributed to the small sample size in their study and different methodology in segmentation of the optic disc.

In OCTA, the VD and blood flow index are important parameters to estimate abnormalities in the blood flow; however, neither the VD nor flow index are the blood flow itself. In an experimental study, the association between OCT angiographic decorrelation and blood flow is not linear but follows a sigmoid pattern.[14] Another weak point of the OCTA includes poor ability to detect movement of erythrocytes on the z axis. So, the relationship of VD and blood flow index to three-dimensional blood flow is unknown at the present time. Other than this, media opacity hampers quantification and interindividual comparison of data. To address this problem, the VD and PLFI were divided by the SS and studied in Table 2, because the SS is an indicator of total reflectance intensity. However, this technique did not greatly affect the result (Table 2, data not shown for PLFI/UA/SS).

Regarding study limitations, other than the significantly correlated parameters, other parameters not included in this study might affect the PLFI/UA and VD. Blood pressure and perfusion pressure might affect the flow index or vessel density.[26] Jia et al.[8] reported that age did not affect the flow index; however, the association between the PLFI/UA and age in the current control eyes was marginally significant ($r = -0.399$, $P = 0.053$; Table 3). Peripapillary choroidal atrophy also might affect the blood flow index, because linear regression analysis between the refractive error and PLFI/UA was marginally significant ($r = -0.348$, $P = 0.097$; Table 3). Other conditions, such as diabetes mellitus, emotional stress, exercise, cardiac diseases, and other unknown parameters, might have unknown effects on the blood flow in the optic disc and await further elaboration.

Another limitation was software-dependent segmentation of the optic disc. Precise segmentation of the anterior and posterior boundaries of the lamina cribrosa might facilitate study of the blood flow or density in each part of pathologic optic nerves. Device and software improvements are desirable to improve our ophthalmologic knowledge.

In summary, the PLFI/UA and VD decreased in glaucomatous eyes. The discriminatory power of the PLFI/UA and VD to differentiate POAG from normal eyes was less than that of the structural parameters; that is, the GLV and cpNFLT. The VD decreased in OH eyes, whereas the PLFI/UA, GLV, and cpNFLT were unaffected.

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