Peripapillary Retinal Nerve Fiber Layer and Microvasculature in Prolonged Type 2 Diabetes Patients Without Clinical Diabetic Retinopathy

Min-Woo Lee,[1] Woo-Hyuk Lee,[2] Cheon-Kuk Ryu,[2] Yong-Min Lee,[1] Young-Hoon Lee,[1] and Jung-Yeul Kim[2]

[1]Department of Ophthalmology, Konyang University College of Medicine, Daejeon, Republic of Korea
[2]Department of Ophthalmology, Chungnam National University College of Medicine, Daejeon, Republic of Korea

Correspondence: Jung-Yeul Kim, Department of Ophthalmology, Chungnam National University Hospital, #640 Daesa-dong, Jung-gu, Daejeon, 301-721, Korea; kimjy@cnu.ac.kr.

MWL and WHL contributed equally to this work as first authors.

Purpose. The purpose of this study to identify the effects of prolonged type 2 diabetes (T2DM) on the peripapillary retinal nerve fiber layer (pRNFL) and peripapillary microvasculature in patients with prolonged T2DM without clinical diabetic retinopathy (DR).

Methods. Subjects were divided into 3 groups: controls (control group; 153 eyes), patients with T2DM < 10 years (DM group 1; 136 eyes), and patients with T2DM ≥ 10 years (DM group 2; 74 eyes). The pRNFL thickness and peripapillary superficial vessel density (VD) were compared. Linear regression analyses were performed to identify factors associated with peripapillary VD in patients with T2DM.

Results. The mean pRNFL thicknesses of the control group, DM group 1, and DM group 2 were 96.0 ± 7.9, 94.5 ± 0.9, and 92.2 ± 8.2 μm, respectively ($P < 0.001$). The VDs were 18.24 ± 0.62, 17.60 ± 1.47, and 17.15 ± 1.38 mm^{-1} in the control group, DM group 1, and DM group 2, respectively ($P < 0.001$). In multivariate linear regression analyses, visual acuity ($B = -2.460$, $P = 0.001$), axial length ($B = -0.169$, $P = 0.008$), T2DM duration ($B = -0.056$, $P < 0.001$), and pRNFL ($B = 0.024$, $P = 0.001$) were significant factors affecting the peripapillary VD in patients with T2DM.

Conclusions. Patients with T2DM without clinical DR showed thinner pRNFL and lower peripapillary VD and perfusion density (PD) compared with normal controls, and such damage was more severe in patients with T2DM ≥ 10 years. Additionally, peripapillary VD was significantly associated with best-corrected visual acuity (BCVA), axial length, T2DM duration, and pRNFL thickness in patients with T2DM.

Keywords: diabetes, optical coherence tomography angiography, retinal nerve fiber layer, vessel density

Type 2 diabetes (T2DM) is a complex metabolic disease, and its macrovascular and microvascular complications account for more than 2 million deaths every year.[1] Additionally, its prevalence has been increasing during recent decades from 108 million in 1980 to 422 million in 2014, and is expected to increase to 629 million by 2045.[1–3] This increased prevalence has led to an increase in the prevalence of diabetic retinopathy (DR).[4] DR is a common microvascular complication of T2DM and is the leading cause of blindness in the working population in the world.[5] Recently, many studies focused on retinal damage in patients with T2DM before emerging clinical DR to find out more efficient preventive strategies based on a better understanding of the pathogenesis of the early stages of the disease. Lim et al.[6] found that the rates of reduction of peripapillary retinal nerve fiber layer (pRNFL) in the patients with T2DM without DR was faster than those of the control group. Simo et al.[7] reported that T2DM caused retinal neurodegeneration, which coalesces with progressive disruption of the retinal neurovascular unit before clinical DR, thus contributing to the early stages of microvascular disease.

With the development of optical coherence tomography angiography (OCTA), noninvasive imaging modality allowing microvascular visualization of the retina as well as quantitative measurements of perfusion, many studies reported the microvasculature in patients with T2DM without DR using OCTA. Zeng et al.[8] reported that parafoveal and perifoveal vessel density (VD) decreased in patients with T2DM without DR in comparison to the control group. Dimitrova et al.[9] found that superficial and deep retinal VDs in the parafovea of patients with T2DM without DR are both decreased compared with healthy subjects. Studies for microvasculature of the peripapillary area also have been reported. Shin et al.[10] reported that the T2DM without DR groups showed lower peripapillary VD and perfusion density (PD) in superficial capillary plexus compared with the control group. Vujosevic et al.[11] reported that there were early changes in the peripapillary vessel morphology and VD of the radial peripapillary plexus in patients with T2DM without DR that correlate to pRNFL thinning. However, studies on prolonged T2DM, which is more than 10 years, on peripapillary microvasculature in patients without DR are insufficient.

The purpose of this study is to identify the effect of prolonged T2DM on peripapillary microvasculature and find factors associated with the peripapillary microvasculature in patients with prolonged T2DM without clinical DR.

METHODS

Patients

This retrospective cross-sectional study adhered to the tenets of the Declaration of Helsinki and was approved by the Institutional Review Board of Chungnam National University Hospital, Daejeon, Republic of Korea (2020-05-069). We reviewed the medical records of patients with diabetes who visited the retina clinic of Chungnam National University Hospital for checkups for DR from March 2017 to April 2020. The requirement for obtaining informed consent was waived due to the retrospective nature of the study. We recorded best-corrected visual acuity (BCVA), intraocular pressure, spherical equivalent, and axial length. Subjects were divided into 3 groups: controls (control group), patients with T2DM < 10 years (DM group 1), and patients with T2DM ≥ 10 years (DM group 2). The exclusion criteria consisted of a history of systemic disease other than diabetes and hypertension, glaucoma, optic nerve disorder, intraocular pressure ≥ 21 mm Hg, axial length ≥ 26.0 mm, or any other optic nerve or retinal dysfunction. We also excluded patients with clinical evidence of DR, such as retinal hemorrhage or microaneurysm. If both eyes met the inclusion criteria, one eye was randomly selected.

Spectral-Domain Optical Coherence Tomography Measurements

Spectral-domain optical coherence tomography (SD-OCT) examination was performed using a Cirrus HD-OCT (Carl Zeiss Meditec, Dublin, CA, USA). The pRNFL measurements were made using an optic disc cube scan. The optic nerve head was brought to the center of the simultaneously scanned image, and a 200 × 200 pixel resolution axial scan was made over an approximate area of 6 × 6 mm that included the optic nerve head and its surroundings. The average and four quadrant sector (superior, temporal, inferior, and nasal) thicknesses were analyzed. We excluded images with a signal strength < 7, obvious decentration, or segmentation errors.

OCTA Measurements

OCTA examination was performed using a Cirrus HD-OCT 5000 with AngioPlex software (Carl Zeiss Meditec), with a wavelength of 840 nm taking 68,000 A-scans per second. The volumetric scans were processed using optical microangiography (OMAG) algorithms to generate the flow images. The OMAG algorithm analyzes differences in both phase and intensity information from repeated B-scan to quantify motion contrast. We obtained an optic disc centered scan using 6 × 6 mm pattern mode, which contains an approximate area of 6 × 6 mm size over the optic nerve head, and all scans were analyzed using en face OCTA images generated automatically by the OMAG algorithm used in AngioPlex software. The VD (total length of perfused vasculature per unit) and PD (total area of perfused vasculature per unit) of the superficial capillary plexus, which spanned from the internal limiting membrane to the inner plexiform layer,

was measured automatically by the software. The software quantified VD and PD via the Early Treatment of Diabetic Retinopathy Study subfields (Fig. 1). We analyzed the peripapillary VD and PD in superficial capillary plexus of the quadrants of the inner and outer rings, and 6 mm full area. Any images with fixation loss, segmentation errors, motion artifacts, or signal strength < 8 were excluded.

Statistical Analysis

All statistical analyses were performed using SPSS statistical software (version 18.0; IBM Corp., Armonk, NY, USA). Demographic characteristics and ocular parameters were compared using 1-way ANOVA with the post hoc Bonferroni correction and the χ^2 test. Analysis of covariance was used to compare the pRNFL thicknesses and OCTA parameters among groups after adjusting for age and BCVA. Univariate and multivariate linear regression analyses were performed to identify factors associated with peripapillary VD in patients with T2DM; we analyzed the VD of the outer circle to exclude the optic disc area.

RESULTS

Demographics

A total of 363 eyes were enrolled: 153 in the control group, 136 in DM group 1, and 74 in DM group 2. The mean age of each group was 59.8 ± 9.0, 58.4 ± 10.6, and 61.9 ± 7.7 years, respectively ($P = 0.060$; Table 1). The mean BCVA was −0.017 ± 0.059, 0.008 ± 0.082, and 0.026 ± 0.099 ($P < 0.001$; control group versus DM group 1, $P = 0.021$; control group versus DM group 2, $P < 0.001$; and DM group 1 versus DM group 2, $P = 0.237$). Sex, laterality, spherical equivalent, intraocular pressure, and axial length were not significantly different among the groups. The HbA1c levels of DM group 1 and DM group 2 was 6.87 ± 0.97 and 7.03 ± 0.94%, respectively ($P = 0.269$).

pRNFL of Each Group

The mean pRNFL thicknesses of the control group, DM group 1, and DM group 2 were 96.0 ± 7.9, 94.5 ± 0.9, and 92.2 ± 8.2 μm, respectively, and they were significantly different after adjusting BCVA ($P < 0.001$; Table 2, Fig. 2). In the post hoc analyses, there were significant differences between the control group and DM group 2 ($P = 0.037$), and between DM group 1 and DM group 2 ($P = 0.049$). In sectoral thickness, temporal and inferior sectors showed significant differences among groups ($P = 0.001$ and $P < 0.001$, respectively), and the superior sector showed a similar trend but did not show a significant difference ($P = 0.060$).

Peripapillary VD and PD in Each Group

The VDs of the full area were 18.24 ± 0.62, 17.60 ± 1.47, and 17.15 ± 1.38 mm^{-1} in the control group, DM group 1, and DM group 2, respectively, and they were significantly different after adjusting BCVA ($P < 0.001$; Table 3). The VDs of the inner and outer circles also showed significant differences among groups ($P < 0.001$ and $P = 0.001$, respectively). In the post hoc analyses, all VDs were significantly different except the VD of the inner circle in DM group 1 versus DM group 2 ($P = 0.288$).
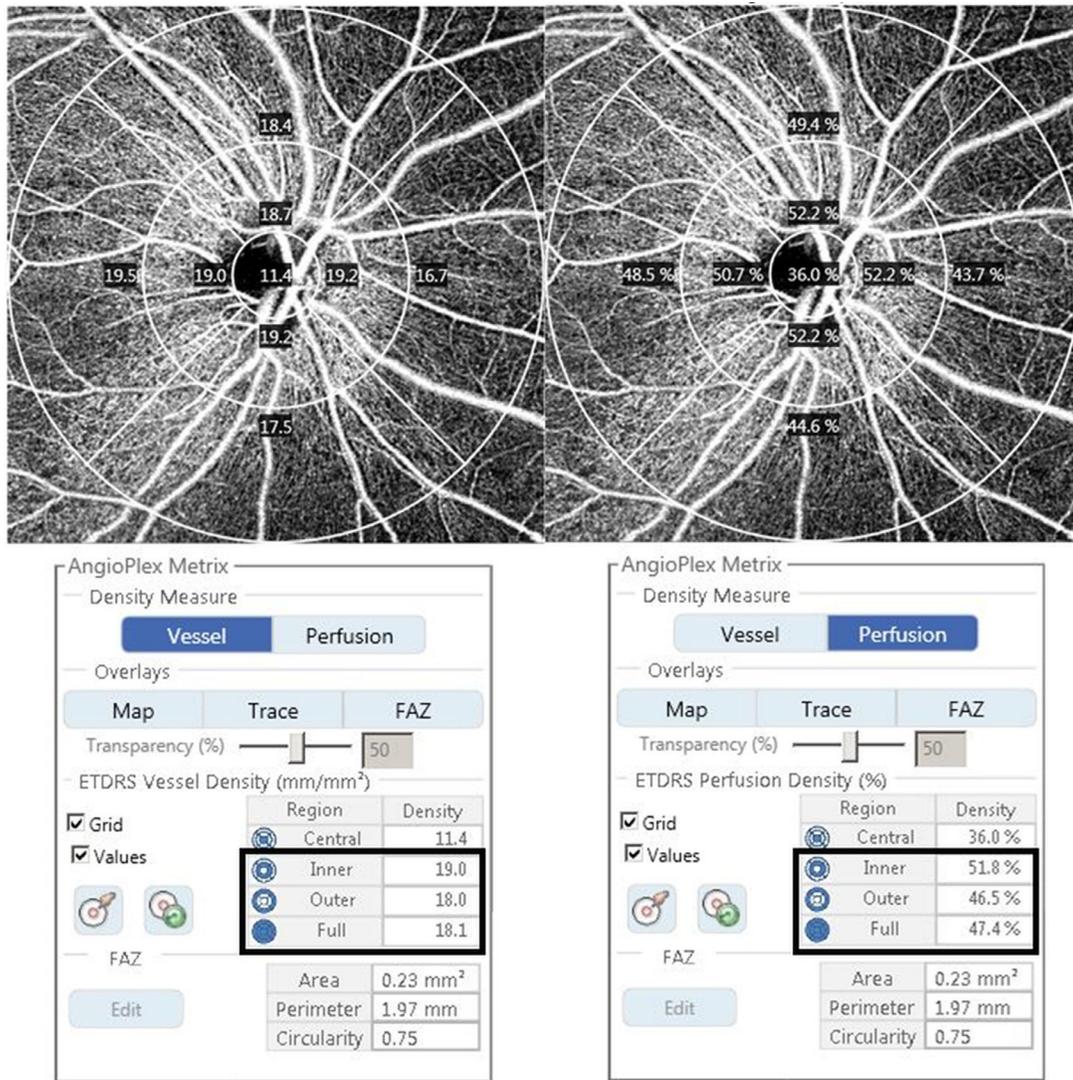
FIGURE 1. A 6 × 6 mm optical coherence tomography angiography image centered on the optic disc. The en face image of the superficial capillary plexus overlaid with the Early Treatment of Diabetic Retinopathy Study grid. The diameters of the 3 concentric circles are 1, 3, and 6 mm. The *black boxes* show the automatic quantitative measurements for an average vessel density and perfusion density of the inner ring, outer ring, and full area.

The PDs of the full area of each group were 0.466 ± 0.015, 0.448 ± 0.043, and 0.430 ± 0.071, respectively ($P < 0.001$). The PDs of the inner and outer circles were also significantly different among groups (both $P < 0.001$). In the post hoc analyses, all PDs were significantly different except the PD of the inner circle in DM group 1 versus DM group 2 ($P = 0.635$).

Factors Associated With Peripapillary VD in Patients With T2DM

In univariate analyses, age (B = −0.022, $P = 0.027$), sex (B = 0.454, $P = 0.022$), BCVA (B = −2.810, $P = 0.012$), axial length (B = −0.251, $P = 0.024$), T2DM duration (B = −0.056, $P < 0.001$), ganglion cell-inner plexiform layer (GC-IPL) thickness (B = 0.043, $P < 0.001$), and pRNFL thickness (B = 0.047, $P < 0.001$) were significant factors affecting the peripapillary VD of outer circle in patients with

T2DM (Table 4). In multivariate analyses, BCVA (B = −2.460, $P = 0.001$), axial length (B = −0.169, $P = 0.008$), diabetes mellitus (DM) duration (B = −0.051, $P < 0.001$), and pRNFL thickness (B = 0.024, $P = 0.001$) showed significant results (Fig. 3).

Discussion

Many studies reported the retinal damages by T2DM before clinical DR. Zeng et al.[8] reported that delayed implicit time and decreased amplitude in electroretinogram (ERG) were found in the T2DM without DR group in comparison to the control group. Loss of dark adaptation and contrast sensitivity, color vision disturbance, and abnormal microperimetry were also reported in patients with T2DM without DR in previous studies.[12–14] These functional alterations would be related to diabetic retinal neurodegeneration, which includes reactive gliosis, diminished retinal neuronal function, and neural-cell apoptosis resulting in

TABLE 1. Demographics and Clinical Characteristics

	Normal Controls ($n = 153$)	DM Group 1 ($n = 136$)	DM Group 2 ($n = 74$)	P Value
Age, mean ± SD, y	59.8 ± 9.0	58.4 ± 10.6	61.9 ± 7.7	0.060
Sex (male, %)	66 (43.1)	65 (47.8)	37 (50.0)	0.564
Laterality (right, %)	75 (49.0)	69 (50.7)	36 (48.6)	0.943
BCVA, mean ± SD, logMAR	−0.017 ± 0.059	0.008 ± 0.082	0.026 ± 0.099	**<0.001**
SE, mean ± SD, diopters	−0.22 ± 1.72	−0.49 ± 1.71	−0.10 ± 1.99	0.248
IOP, mean ± SD, mm Hg	14.6 ± 2.8	15.8 ± 2.8	15.1 ± 2.9	0.060
Axial length, mean ± SD, mm	23.7 ± 1.0	23.9 ± 0.9	23.6 ± 1.0	0.051
HTN (n, %)	41 (26.8)	46 (33.8)	21 (28.4)	0.320
HTN duration, mean ± SD, y	8.90 ± 5.26	7.43 ± 3.54	9.95 ± 5.55	0.091
DM duration, mean ± SD, y	0	3.74 ± 2.52	14.65 ± 4.92	**<0.001**
HbA1C, mean ± SD, %	N/A	6.87 ± 0.97	7.03 ± 0.94	0.269
Disc parameters, mean ± SD				
Rim area, mm^2	1.33 ± 0.23	1.26 ± 0.24	1.23 ± 0.23	0.294
Disc area, mm^2	1.85 ± 0.34	1.94 ± 0.40	1.92 ± 0.40	0.657
Cup-disc ratio	0.49 ± 0.16	0.54 ± 0.16	0.55 ± 0.18	0.363
CMT, mean ± SD, μm	252.2 ± 18.0	254.0 ± 30.6	254.9 ± 26.4	0.352
GC-IPL thickness, mean ± SD, μm	83.8 ± 5.7	82.6 ± 8.8	79.4 ± 6.9	**<0.001**

SD, standard deviation; BCVA, best-corrected visual acuity; SE, spherical equivalent; IOP, intraocular pressure; HTN, hypertension; DM, diabetes mellitus; CMT, central macular thickness; GC-IPL, ganglion cell-inner plexiform layer.

DM group 1 = patients with type 2 diabetes < 10 years, and DM group 2 = patients with type 2 diabetes ≥ 10 years.

Values in boldface ($P < 0.05$) are statistically significant.

TABLE 2. Peripapillary Retinal Nerve Fiber Layer Thickness in Each Group

	Control	DM Group 1	DM Group 2	ANCOVA P Value[*]
Mean	96.0 ± 7.9	94.5 ± 9.9	92.2 ± 8.2	**<0.001**
Sector				
Superior	120.8 ± 20.2	116.1 ± 21.0	114.7 ± 17.6	0.060
Temporal	72.4 ± 10.5	71.5 ± 14.1	67.3 ± 11.4	**0.001**
Inferior	123.3 ± 15.1	120.8 ± 19.6	118.8 ± 22.9	**<0.001**
Nasal	68.4 ± 9.1	68.4 ± 11.8	71.3 ± 16.2	0.207

[*] ANCOVA analyses by adjusting for best-corrected visual acuity.

DM group 1 = patients with type 2 diabetes < 10 years, and DM group 2 = patients with type 2 diabetes ≥ 10 years.

Values in boldface ($P < 0.05$) are statistically significant. All values are expressed as the mean ± standard deviation (μm).

a reduced thickness of inner retinal layers and the nerve fiber layer.[7,15,16] Furthermore, diabetic retinal neurodegeneration could cause the progressive dysfunction of neurovascular coupling, which would affect retinal microcirculation and microvasculature, although longitudinal studies are needed to confirm this.[7] In this study, we identified that prolonged T2DM could cause more severe damages on pRNFL and peripapillary microvasculature even under relatively well glycemic control. Additionally, BCVA, axial length, T2DM duration, and pRNFL thickness were significantly associated with peripapillary VD in patients with T2DM ≥ 10 years.

Lim et al.[6] reported that the estimated mean pRNFL loss was −0.92 μm/year in the non-DR group, which was 2.9-fold ($P = 0.003$) greater than that of the control group (−0.35 μm/year). Sohn et al.[17] also reported the progressive loss of inner retina, including RNFL and GC-IPL in patients with DM and no to minimal DR. Our study showed consistent results with previous studies that DM groups had thinner pRNFL than the control group. Additionally, DM
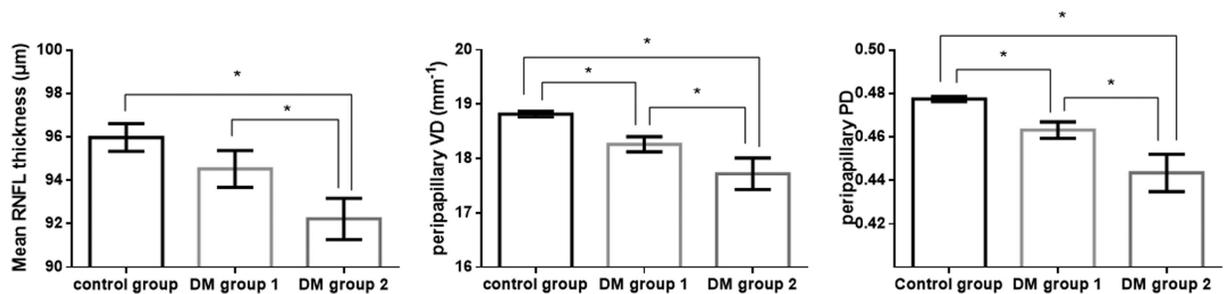
FIGURE 2. Bar graphs with standard errors of the mean retinal nerve fiber layer (RNFL) thickness, peripapillary vessel density (VD), and peripapillary perfusion density (VD) of each group. *Statistically significant difference. DM group 1, patients with type 2 diabetes < 10 years. DM group 2, patients with type 2 diabetes ≥ 10 years.

TABLE 3. Superficial Peripapillary Vessel Density and Perfusion Density in Each Group Using Optical Coherence Tomography Angiography

	Control	DM Group 1	DM Group 2	ANCOVA *P* Value[*]
Vessel density				
Full area	18.24 ± 0.62	17.60 ± 1.47	17.15 ± 1.38	**<0.001**
Outer circle	18.82 ± 0.65	18.26 ± 1.62	17.72 ± 2.48	**<0.001**
Inner circle	17.72 ± 1.19	17.21 ± 1.86	16.84 ± 2.13	**0.001**
Perfusion density				
Full area	0.466 ± 0.015	0.448 ± 0.043	0.430 ± 0.071	**<0.001**
Outer circle	0.478 ± 0.015	0.463 ± 0.044	0.443 ± 0.074	**<0.001**
Inner circle	0.465 ± 0.032	0.452 ± 0.050	0.446 ± 0.044	**<0.001**

[*] ANCOVA analyses by adjusting for best-correct visual acuity.
DM group 1 = patients with type 2 diabetes < 10 years, and DM group 2 = patients with type 2 diabetes ≥ 10 years.
Values in boldface ($P < 0.05$) are statistically significant. All values are expressed as the mean ± standard deviation.

TABLE 4. Univariate and Multivariate Linear Regression Analyses Determining the Factors Associated With Peripapillary Vessel Density in Patients With Type 2 Diabetes

	Univariate		Multivariate	
	B (95% CI)	*P* Values	B (95% CI)	*P* Values
Age, y	−0.022 (−0.041 to 0.002)	**0.027**	−0.008 (−0.020 to 0.004)	0.180
Sex	0.454 (0.067–0.841)	**0.022**	0.118 (−0.111 to 0.348)	0.312
Laterality	−1.050 (−2.182 to 0.082)	0.068		
BCVA	−2.810 (−5.002 to −0.318)	**0.012**	−2.460 (−3.900 to −1.020)	**0.001**
IOP	−0.014 (−0.082 to 0.055)	0.694		
Axial length	−0.251 (−0.470 to 0.033)	**0.024**	−0.169 (−0.295 to 0.044)	**0.008**
DM duration	−0.056 (−0.086 to 0.026)	**<0.001**	−0.051 (−0.070 to 0.031)	**<0.001**
HbA1c	0.006 (−0.712 to 0.724)	0.988		
HTN	0.030 (−0.008 to 0.068)	0.118		
HTN duration	0.067 (−0.042 to 0.211)	0.094		
CMT	−0.003 (−0.010 to 0.003)	0.331		
pRNFL	0.047 (0.027–0.067)	**<0.001**	0.024 (0.010–0.039)	**0.001**
GC-IPL	0.043 (0.020–0.066)	**<0.001**	0.015 (−0.003 to 0.033)	0.099

BCVA, best-corrected visual acuity; SE, spherical equivalent; IOP, intraocular pressure; DM, diabetes mellitus; HTN, hypertension; CMT, central macular thickness; pRNFL, peripapillary retinal nerve fiber layer; GC-IPL, ganglion cell-inner plexiform layer.
Values in boldface ($P < 0.05$) are statistically significant.

group 2 showed thinner pRNFL than DM group 1 after adjusting BCVA, although HbA1c was not significantly different between the 2 groups. Once retinal neurodegeneration started, neuronal apoptosis and glial dysfunction seem to persist even under well glycemic control, resulting in severe pRNFL damage in patients with prolonged T2DM. Meanwhile, glial dysfunction, retinal ganglion cell loss by cellular apoptosis, and impaired microcirculation are also the mechanism of optic neuropathy in glaucoma, which results in

pRNFL thinning and visual field loss.[18,19] These common mechanisms would be helpful to explain the relationship between the two diseases that T2DM is one of the risk factors for glaucoma and the duration of diabetes was associated with a significantly increased risk of glaucoma.[20]

Shin et al.[10] reported that the no DR and NPDR groups showed a significantly lower peripapillary VD and PD compared with the control group. Vujosevic et al.[11] found that there was a significant decrease in peripapil-
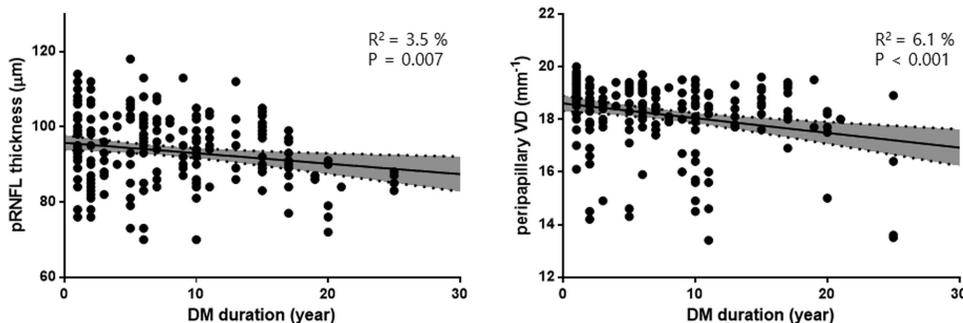
FIGURE 3. Scatterplots and linear regression analyses between the duration of type 2 diabetes mellitus (DM) and peripapillary retinal nerve fiber layer (pRNFL) thickness in patients with DM.

lary VD, number of branches, and total branches length in no DR groups versus control groups. Our study also showed decreased peripapillary VD and PD in patients with T2DM, and DM group 2 had significantly lower VD and PD than DM group 1, which is consistent with the trend of pRNFL thickness. These results were similar to our previous study that patients with prolonged T2DM exhibited thinner macular GC-IPL and more severe impairment of macular microvasculature.[21] Neuronal death and glial dysfunction by diabetic retinal neurodegeneration can cause a breakdown of the blood-retinal barrier, vasoregression, and impairment of neurovascular coupling, which may result in early microvascular impairment.[16] As the damages caused by such diabetic retinal neurodegeneration accumulated, the patients with prolonged T2DM would show more severely damaged microvasculature similar to pRNFL thinning. Therefore, physicians should not assume that the microvasculature is under a stable state in patients with T2DM without clinical DR and should monitor its status constantly.

Previous studies reported a significant correlation between BCVA and macular VD in patients with T2DM with or without DR.[22,23] Superficial peripapillary VD was also significantly associated with BCVA in patients with T2DM in our study. Augstsburger et al.[24] reported that BCVA was significantly correlated with radial peripapillary complex VD in patients with nonarteritic anterior ischemic optic neuropathy. Mammo et al.[25] identified that there was a significant correlation between radial peripapillary complex density and visual field index value in patients with glaucoma or glaucoma suspect. The impairment of peripapillary microvasculature by various mechanisms seemed to be associated with decreased visual function. In prolonged T2DM, the blood supply of the outer retina, which is directly related to visual function, could be more affected by the inner retina because of the impairment of choroidal circulation by a failure of the autoregulatory mechanism.[26] Additionally, compromise of the outer blood-retinal barrier could result in photoreceptor apoptosis and increased inflammation in the outer retina.[27,28] Therefore, damages on superficial microvasculature may affect the outer retinal layer, which could result in decreased visual function.

The axial length was one of the significant factors associated with superficial peripapillary VD in patients with T2DM. He et al.[29] reported that radial peripapillary complex VD showed a significantly negative correlation with axial length. They speculated that the elongation of the eyeball could cause thinning of the retina, and the thinning of the retinal tissues might cause reduced oxygen demand, leading to a decrease in blood circulation. Additionally, previous studies reported that high myopia showed a greater reduction in the inner retina, including pRNFL and GC-IPL.[30,31] This may be associated with the acceleration of diabetic retinal neurodegeneration, which could result in severe impairment of peripapillary microvasculature. However, this result associated with axial length would contain a limitation that measured and analyzed area of the retina could be different in each subject according to the axial length. Sampson et al.[32] reported that errors in OCTA parameters, including VD and the foveal avascular zone area, could arise in the absence of image size correction. Savini et al.[33] also identified that comparing the RNFL thickness measurements with the normative database could be misleading for short and long eyes because of axial length-induced ocular magnification. Although we did not include the eyes with axial length ≥ 26.0 mm to reduce such confound, there are still possibil-

ities that not all images had a scan size of 6×6 mm even in the normal range of axial length.[34,35] Therefore, further studies, including high myopia with analyses considered the retinal magnification effect by image size correction, are needed to confirm this relationship more precisely.

Vujosevic et al.[11] reported that there was a significant correlation between OCTA parameters, such as VD or PD and pRNFL thickness in the peripapillary region in patients with T2DM without DR. The pRNFL thickness affected peripapillary VD significantly in our study, which is consistent with the previous study. Additionally, a correlation between pRNFL and peripapillary VD was reported in various diseases.[24,25] Mase et al.[36] identified this correlation in healthy subjects, suggesting that the radial peripapillary complex is the most important structure in maintaining pRNFL integrity. Therefore, although the exact relationship requires further study, these two factors would reflect the damages of each other. The peripapillary VD could be useful for the evaluation of retinal ganglion cell damage in patients with T2DM especially with an anomalous optic disc, such as myopic disc, which is hard to evaluate the exact pRNFL thickness.

In patients with T2DM, the HbA1c level was not a significant factor affecting the peripapillary VD. Sohn et al.[17] reported that the HbA1c level did not influence the thickness of the RNFL or GC-IPL in their longitudinal study. As retinal ganglion cell and microvasculature are closely related by neurovascular unit, diabetic retinal neurodegeneration and retinal microangiopathy may show a similar trend for the HbA1c level. However, the longitudinal studies are needed to identify the relationship between the glycemic control and impairment of peripapillary microvasculature more exactly. Because only glycemic control is not enough for such retinal damage, studies for neuroprotective agents have been reported recently.[37,38] The European Consortium for the Early Treatment of Diabetic Retinopathy (EUROCONDOR) reported that somatostatin and brimonidine were effective in preventing the increase of the mean implicit time in multifocal ERG in patients with diabetic retinal neurodegeneration.[37] They also reported that there were arteriolar widening and venular dilation in patients with mild DR treated with brimonidine and somatostatin.[38] However, such agents did not show any effects in preventing or arresting the microvascular disease, and they also have not been proven to have effects for inner retinal damage by diabetic retinal neurodegeneration. Although brimonidine and somatostatin would have effects for neuronal dysfunction to some extent, more studies for the prevention or arrest of diabetic retinal neurodegeneration and microangiopathy are needed.

Our study had several limitations. First, this study was a retrospective cross-sectional design without longitudinal data, which would be hard to confirm the temporal relationship. Second, the number of cases in DM group 2 was relatively small compared with other groups because of the strict inclusion criteria. Third, we could not afford various information about visual functions, such as contrast sensitivity, color vision test, or microperimetry. Fourth, we analyzed peripapillary VD, including large vessels, as we used the VD parameters automatically provided by the software, which did not differentiate between capillaries and large vessels. Fifth, we could not analyze the intermediate or deep capillary plexus because we analyzed the OCTA values provided by the Angioplex software automatically, which only provided values for superficial capillary plexus. Although analysis of the superficial capillary plexus is more

accurate than that of the intermediate or deep capillary plexus because of projection artifacts, it would be very meaningful to analyze each capillary plexus depending on the DM duration in the future study. The strengths of our study were that we included OCTA images with signal strength $\geq$ 8 for accurate analyses. Additionally, few studies have reported the impact of prolonged T2DM on pRNFL and peripapillary microvasculature with a relatively large number of cases.

In conclusion, patients with T2DM without clinical DR showed thinner pRNFL and lower peripapillary VD and PD compared with normal controls, and such damages were more severe in patients with T2DM $\geq$ 10 years. Additionally, the peripapillary VD was significantly associated with BCVA, axial length, T2DM duration, and pRNFL thickness in patients with T2DM. Physicians should know that diabetic retinal neurodegeneration and peripapillary microvascular impairment could persist over time even under well glycemic control, and further studies for prevention or arrest of such damages are needed.

Acknowledgments

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