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Optical Coherence Tomographic Angiography in Type 2 Diabetes and Diabetic Retinopathy

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IMPORTANCE Optical coherence tomographic angiography (OCT-A) is able to visualize retinal microvasculature without the need for injection of fluorescein contrast dye. Nevertheless, it is only able to capture a limited view of macula and does not show leakage.

OBJECTIVES To evaluate the retinal microvasculature using OCT-A in patients with type 2 diabetes as well as the association of OCT-A characteristics with diabetic retinopathy (DR) and systemic risk factors.

DESIGN, SETTING, AND PARTICIPANTS A prospective, observational study was conducted from January 1 to June 30, 2016, at medical retina clinics at the Singapore National Eye Center among 50 patients with type 2 diabetes with and without DR (n = 100 eyes). We examined the retinal microvasculature with swept-source OCT-A and a semiautomated software to measure the capillary density index (CDI) and fractal dimension (FD) at the superficial vascular plexus (SVP) and deep retinal vascular plexus (DVP). We collected data on histories of patients' glycated hemoglobin A_{1c}, hypertension, hyperlipidemia, smoking, and renal impairment.

MAIN OUTCOMES AND MEASURES The CDI and FD at the SVP and DVP for each severity level of DR and the association of systemic risk factors vs the CDI and FD.

RESULTS The mean (SD) glycated hemoglobin A_{1c} of the 50 patients (26 men and 24 women; 35 Chinese; mean [SD] age, 59.5 [8.9] years) was 7.9% (1.7%). The mean (SD) CDI at the SVP decreased from 0.358 (0.017) in patients with no DR to 0.338 (0.012) in patients with proliferative DR ($P < .001$) and at the DVP decreased in patients with no DR from 0.361 (0.019) to 0.345 (0.020) in patients with proliferative DR ($P = .04$). The mean (SD) FD at the SVP increased from 1.53 (0.05) in patients with no DR to 1.60 (0.05) in patients with proliferative DR ($P < .01$) and at the DVP increased from 1.55 (0.06) in patients with no DR to 1.61 (0.05) in patients with proliferative DR ($P = .02$). For systemic risk factors, hyperlipidemia (odds ratio [OR], 9.82; 95% CI, 6.92-11.23; $P < .001$), smoking (OR, 10.90; 95% CI, 8.23-12.34; $P < .001$), and renal impairment (OR, 3.72; 95% CI, 1.80-4.81; $P = .05$) were associated with reduced CDI, while increased glycated hemoglobin A_{1c} (≥8%) (OR, 8.77; 95% CI, 5.23-10.81; $P < .01$) and renal impairment (OR, 10.30; 95% CI, 8.21-11.91; $P < .001$) were associated with increased FD.

CONCLUSIONS AND RELEVANCE Optical coherence tomographic angiography is a novel imaging modality to quantify the retinal capillary microvasculature in patients with diabetes. It can be potentially used in interventional trials to study the effect of systemic risk factors on the microvasculature that was previously not accessible in a noninvasive manner. The relevance of these findings relative to visual acuity, however, remains largely unknown at this time.

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For the past 50 years, the retinal microvascular structure and its function, such as flow, have been measured using fundus fluorescein angiography. Since the description of this technique by Novotny and Alvis,[1] fundus fluorescein angiography has been the criterion standard diagnostic tool to evaluate types 1 and 2 diabetes and diabetic retinopathy (DR).[2] Fundus fluorescein angiography provides detailed phenotyping of early microvascular changes in the retina, allowing, for example, the differentiation of red spots seen in the fundus into either microaneurysms or dot hemorrhages, as well as the early detection of ischemia. However, fundus fluorescein angiography has 3 major limitations. First, it is an invasive test that involves administration of an intravenous dye that can potentially result in systemic adverse effects such as anaphylaxis, cardiac and respiratory distress, and renal disorder.[3] Second, fundus fluorescein angiography is time consuming and not readily available in nonhospital settings. This fact has, therefore, affected the ability to study early structural and functional changes in the retinal microvasculature in larger-scale clinical studies and trials. Finally, anatomically, the retinal vasculature has 2 layers: the superficial and deep capillary plexus, and there is a capillary-free zone at the fovea. The complex branching pattern of each plexus is considered to be engineered to maintain smooth laminar flow, and change in vascular diameter, tortuosity, and closure of the capillary bed profoundly affects oxygen supply to the inner retina. Information obtained by fundus fluorescein angiography is limited, however, and does not provide depth-resolved images of the 2 layers.

The technology of optical coherence tomographic angiography (OCT-A) is a major advance in ophthalmology and offers the opportunity to noninvasively visualize different retinal capillary layers without the need for injection of fluorescein sodium dye.[4-8] Optical coherence tomographic angiography allows clinicians for the first time to visualize precise 3-dimensional microarchitecture of the vessels such as the perifoveal superficial and deep capillary network, previously only visible with histologic examination. Moreover, it also provides information about changes in blood flow in patients with diabetes that can be observed at an earlier stage. Optical coherence tomographic angiography, however, is not able to show leakage and there is still incomplete understanding of artifacts, especially at the deep plexus. Since the availability of OCT-A, there have been several retrospective studies that have reported the changes of retinal microvasculature in patients with DR.[9-11] Most of these studies have been small and none have evaluated the effect of systemic metabolic and vascular risk factors on the retinal microvascular density and branching complexity measured via OCT-A.

The purpose of this study was to describe retinal microvascular changes measured via OCT-A in patients with diabetes and different severity of DR, and to evaluate the effect of systemic metabolic and vascular risk factors on retinal capillary density and morphologic characteristics.

Key Points

Question What is the effect of the severity of diabetic retinopathy and systemic vascular risk factors on the structural changes at the retinal capillary level?

Findings In this observational cohort study, structural changes in the retinal microvasculature were associated with severity of diabetic retinopathy and systemic metabolic and vascular risk factors in patients with type 2 diabetes.

Meaning These data suggest that optical coherence tomographic angiography potentially can be used in larger epidemiologic and clinical studies, including interventional trials, to directly visualize the previously inaccessible microvasculature in a noninvasive manner.

Methods

Study Design

This was a prospective cohort study of 100 eyes in 50 patients with type 2 diabetes who attended the DR screening clinic in the Singapore National Eye Center, Singapore, Singapore, from January 1 to June 30, 2016. We measured capillary density index (CDI) and fractal dimension (FD) using a swept-source OCT-A machine (Topcon Corp) to examine the degree of perifoveal ischemic changes and capillary branching complexity, respectively, at the superficial vascular plexus (SVP) and deep vascular plexus (DVP) layers.

This study was approved by the SingHealth Centralized Institutional Review Board, Singapore (protocol No. R1277/83/2015), and was conducted in accordance with the Declaration of Helsinki.[12] All participants provided signed informed consent.

All treatment-naive patients with diabetes were included in the study if there was no media opacity affecting photography or OCT capture. The exclusion criteria included pregnancy, any previous retinal or macular diseases, and glaucoma. All patients underwent a comprehensive ophthalmic examination, including assessment of best-corrected visual acuity using logarithm of the minimal angle of resolution (logMAR), slit-lamp biomicroscopy, color fundus photographs, swept-source OCT-A (Topcon Corp) and fundus fluorescein angiography (TRC-50X/IMAGEnet 2000; Topcon Corp).

Severity of DR was categorized into none, mild nonproliferative DR (NPDR), moderate NPDR, severe NPDR, and proliferative DR (PDR), based on the International Clinical Diabetic Retinopathy Severity Scales.[13] Similarly, the presence of diabetic macular edema (DME) was classified based on the same scale.

Clinical information including history of glycated hemoglobin A_{1c} (HbA_{1c}), hypertension, hyperlipidemia, smoking, and renal impairment was recorded using a standardized clinic record form. Two sets of systolic and diastolic blood pressure readings were taken 5 minutes apart, with a third reading taken if the 2 differed by more than 10 mm Hg systolic or more than 5 mm Hg diastolic. The mean of the 2 closest readings were used for analysis. In our study, hypertension was defined as a systolic blood pressure of more than 140 mm Hg, a diastolic blood pressure of more than 90 mm Hg, or a self-reported history of hypertension. Hyperlipidemia was defined as either a total cholesterol level of 240 mg/dL or higher (to convert cholesterol to millimoles per liter, multiply by 0.0259) or the use of lipid-lowering drugs. Chronic kidney disease was defined as an estimated glomerular filtration rate of less than 60 mL/min/1.73 m^2,

using the National Kidney Foundation Kidney Disease Outcomes Quality Initiative Working Group definition.[14]

OCT-A Measurements and Segmentation

The swept-source OCT-A images were all processed using the angiography ratio analysis method. A volumetric OCT scan was acquired on a 3 × 3-mm field of view in 4 seconds of total OCT scan time. Each B-scan position was scanned 4 times. The en face OCT image was segmented with an inner boundary at 3 µm beneath the internal limiting membrane and the outer boundary was set at 15 µm beneath the inner plexiform layer to obtain images of the SVP. The en face image was segmented with an inner boundary at 15 µm beneath the inner plexiform layer and the outer boundary was set at 70 µm beneath the inner plexiform layer to obtain images of the DVP. For images with segmentation errors, we would manually correct them by choosing the appropriate layers for analysis.

As characteristics that describe vascular density and morphologic features, we used CDI and FD. A custom semiautomated algorithm was developed and used to quantify CDI. For CDI, all OCT-A images were analyzed using public domain software (ImageJ; National Institutes of Health).[15] A circle with a radius of 1.5 mm centered at the subfoveal region was made and further divided into the superonasal, superotemporal, inferonasal, and inferotemporal quadrants. Using the Niblack thresholding technique, all images were binarized and converted to 8 bits with a mean pixel value and SD of all points. Subsequently, the luminal area was highlighted within the circle with the brightness set to 0 and 254. The luminal area in the individual quadrant was merged with the corresponding threshold area and measured using the Roi manager tool of ImageJ. The CDI of each quadrant was defined as the ratio of the luminal area to the total area of that quadrant. The global CDI was the mean CDI value within the 1.5-mm-radius circle centering on the subfoveal region. We computed the CDI of the SVP and DVP for all the 3 × 3-mm scans.

For the retinal capillary branching complexity, we used Image J to convert the image to 8 bits and processed the images using Frangi vesselness.[15] Once the images were binarized, we measured the complexity of the vessel using the fractal box count method.[16]

Statistical Analysis

Statistical analysis was performed using JMP software, version 9.0 (SAS Institute). We used *t* tests to compare the CDI and FD between 2 DR groups (no DR, mild NPDR, and moderate NPDR vs severe NPDR and PDR). When the data from both eyes of the same patients were used, 2-tailed *P* values were calculated using the mixed model approach regarding the eye as a unit of analysis. $P < .05$ was considered significant. For correlation analysis, we used the CDI and FD from the left eye of the patients and evaluated the association of systemic vascular risk factors (HbA_{1c}, hypertension, hyperlipidemia, smoking, and renal impairment) with OCT-A characteristics, using the lowest CDI quartile and highest FD quartile with nominal regression analysis.

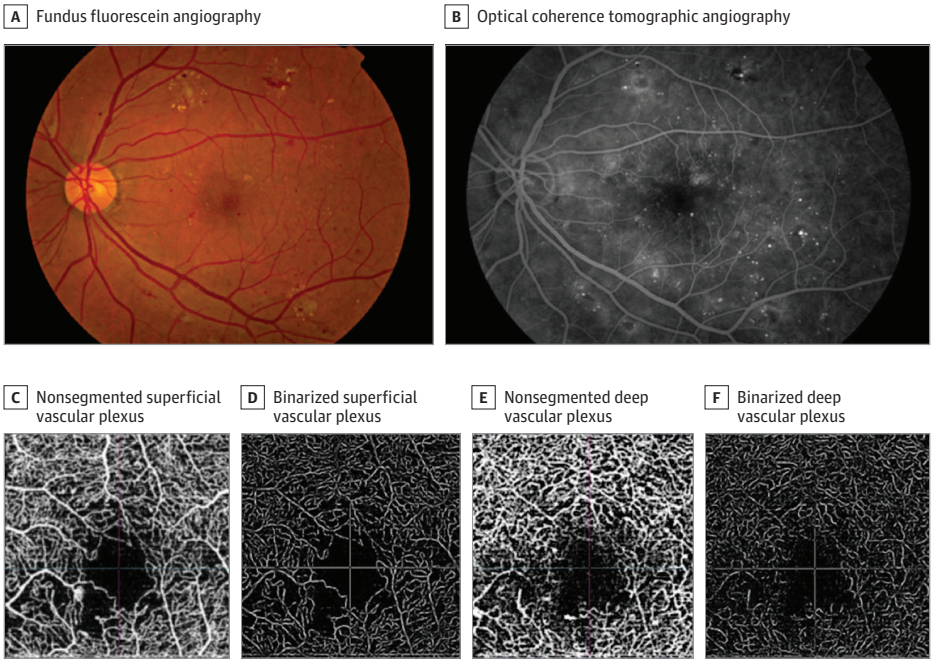
Results

The mean (SD) age of the 50 patients was 59.5 (8.9) years and the mean (SD) HbA_{1c} was 7.9% (1.7%) (in SI units, this is the proportion of total hemoglobin). More than half the patients were male (26 [52%]) and Chinese (35 [70%]). For DR severity level, 19 eyes had no DR, while 17, 21, 22, and 21 eyes had mild NPDR, moderate NPDR, severe NPDR, and PDR, respectively. Of those, 16 eyes had DME. Two eyes (2%) were excluded from the final study group owing to poor OCT-A image quality. **Figure 1** shows the color photograph of the retina of a patient with severe NPDR with corresponding fundus fluorescein angiography and OCT-A at the SVP and DVP layers. The mean (SD) visual acuity was 0.387 (0.271) logMAR, 0.473 (0.316) logMAR, 0.276 (0.256) logMAR, 0.264 (0.133) logMAR, and 0.252 (0.179) logMAR for no DR, mild NPDR, moderate NPDR, severe NPDR, and PDR, respectively.

As shown in **Table 1**, the mean (SD) global CDI at the SVP for no DR, mild NPDR, moderate NPDR, severe NPDR, and PDR were 0.358 (0.017), 0.354 (0.090), 0.346 (0.013), 0.342 (0.019), and 0.338 (0.012), respectively. A similar trend was also observed at the DVP, with mean (SD) global CDI values of 0.361 (0.019), 0.353 (0.021), 0.352 (0.018), 0.351 (0.025), and 0.345 (0.020), respectively, for no DR, mild NPDR, moderate NPDR, severe NPDR, and PDR (Table 1). For DME, the mean (SD) CDI at the superonasal, superotemporal, inferonasal, and inferotemporal quadrants of the SVP were 0.339 (0.018), 0.339 (0.018), 0.354 (0.025), and 0.343 (0.024), respectively, whereas at the DVP, the mean (SD) CDI were 0.349 (0.024), 0.347 (0.025), 0.366 (0.037), 0.362 and (0.043), respectively. The global CDI was lower in patients with DME than those without DME at both the SVP (0.344 vs 0.347; $P = .16$) and the DVP (0.349 vs 0.357; $P = .12$). **Table 2** shows that the FD of the SVP and DVP correlates positively with worsening DR severity levels. For the SVP, the mean (SD) FD increased from 1.53 (0.05) in patients with no DR to 1.60 (0.05) in those with PDR ($P < .01$), whereas for the DVP, it increased from 1.55 (0.06) in patients with no DR to 1.61 (0.05) in those with PDR ($P = .02$). The changes of CDI and FD correlated between the SVP and DVP layers. The Pearson correlations for global CDI and FD between the SVP and DVP were 0.55 ($P < .001$) and 0.88 ($P < .001$), respectively. Similarly, the sectoral CDI at the superotemporal, superonasal, inferonasal, and inferotemporal quadrants was 0.27, 0.46, 0.67, and 0.65, respectively ($P < .001$). **Figure 2** shows the nonsegmented and binarized OCT-A images of patients with increasing severity of DR.

The lower quartile, median, and upper quartile for CDI were 0.33, 0.35, and 0.36 and and for FD were 1.54, 1.58, and 1.62, respectively. When controlled for age, sex, race/ethnicity, and severity of DR, low CDI (lower quartile) was significantly associated with hyperlipidemia (odds ratio, [OR], 9.82; 95% CI, 6.92-11.23; $P < .001$), smoking (OR, 10.90; 95% CI, 8.23-12.34; $P < .001$), and renal impairment (OR, 3.72; 95% CI, 1.80-4.81; $P = .05$). High FD was associated with an increased HbA_{1c} of 8% or more (OR, 8.77; 95% CI, 5.23-10.81; $P < .01$) and renal impairment (OR, 10.30; 95% CI, 8.21-11.91; $P < .001$) (**Table 3**).

Figure 1. Retina of a Patient With Severe Nonproliferative Diabetic Retinopathy



A, Corresponding fundus fluorescein angiography. B, Optical coherence tomographic angiography. C, Nonsegmented superficial vascular plexus. D, Binarized superficial vascular plexus. E, Nonsegmented deep vascular plexus. F, Binarized deep vascular plexus.

Table 1. Individual Quadrant and Global Capillary Density Index at the Superficial and Deep Vascular Plexuses

Severity of DR	Global Capillary Density, Mean (SD)				
	Superotemporal Quadrant	Superonasal Quadrant	Inferotemporal Quadrant	Inferonasal Quadrant	Total
Superficial vascular plexus					
No DR	0.353 (0.021)	0.354 (0.021)	0.364 (0.030)	0.359 (0.018)	0.358 (0.017)
Mild NPDR	0.364 (0.021)	0.357 (0.017)	0.351 (0.031)	0.343 (0.030)	0.354 (0.090)
Moderate NPDR	0.347 (0.020)	0.343 (0.020)	0.351 (0.026)	0.342 (0.021)	0.346 (0.013)
Severe NPDR	0.347 (0.029)	0.341 (0.021)	0.336 (0.025)	0.343 (0.026)	0.342 (0.019)
PDR	0.336 (0.022)	0.339 (0.021)	0.336 (0.029)	0.339 (0.023)	0.338 (0.012)
P value	.02	<.01	<.01	.07	<.001
P value (adjusted)[a]	.02	<.01	<.01	.05	<.001
Deep vascular plexus					
No DR	0.359 (0.021)	0.353 (0.024)	0.367 (0.036)	0.366 (0.022)	0.361 (0.019)
Mild NPDR	0.361 (0.029)	0.361 (0.025)	0.343 (0.037)	0.344 (0.036)	0.353 (0.021)
Moderate NPDR	0.355 (0.024)	0.342 (0.025)	0.361 (0.026)	0.349 (0.029)	0.352 (0.018)
Severe NPDR	0.355 (0.039)	0.348 (0.030)	0.343 (0.022)	0.358 (0.038)	0.351 (0.025)
PDR	0.349 (0.025)	0.334 (0.029)	0.339 (0.039)	0.358 (0.040)	0.345 (0.020)
P value	.15	.04	<.01	.68	.04
P value (adjusted)[a]	.41	.09	.04	.08	.15

Abbreviations: DR, diabetic retinopathy; NPDR, nonproliferative diabetic retinopathy; PDR, proliferative diabetic retinopathy.

[a] Adjusted for age, sex, and a history of glycated hemoglobin A_{1c}, hyperlipidemia, renal impairment, and smoking.

Discussion

In this prospective study, we describe the application of OCT-A to study a cohort of patients with diabetes and DR and examine a variety of measures with metabolic and systemic vascular risk factors. We document functional and structural changes in the retinal microvasculature associated with severity of DR and risk factors. Our study suggests that OCT-A is a new, time-efficient, noninvasive tool to quantify the retinal capillary microvasculature to study diabetes, its microvascular and macrovascular complications, and potentially its outcomes.

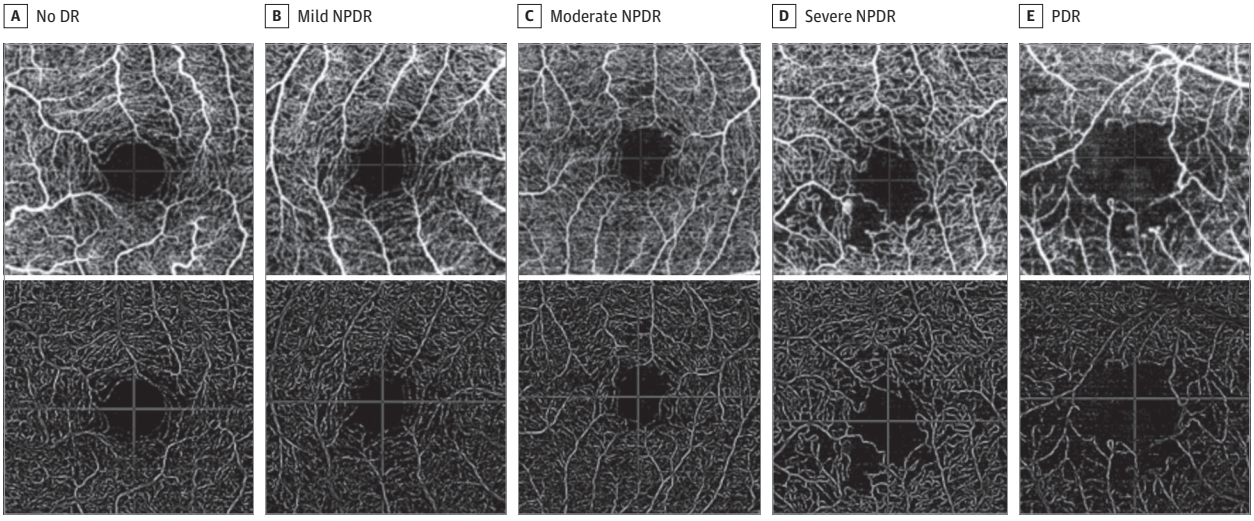
This series evaluates the use of OCT-A with automated software (angiography ratio analysis) and associations with systemic risk factors and the retinal microvasculature. We report several significant findings that are important in advancing our understanding of the structural changes at the retinal capillary level with increasing severity of DR and systemic vascular risk factors. First, this study showed that the quantitative changes in vascular density and complexities are useful

Table 2. Fractal Dimension at the Superficial Vascular Plexus and Deep Vascular Plexus

	Fractal Dimension, Mean (SD)					
Location	No DR	Mild NPDR	Moderate NPDR	Severe NPDR	PDR	*P* Value
Superficial vascular plexus	1.53 (0.05)	1.58 (0.05)	1.57 (0.07)	1.57 (0.06)	1.60 (0.05)	<.01
Deep vascular plexus	1.55 (0.06)	1.60 (0.06)	1.60 (0.06)	1.59 (0.07)	1.61 (0.05)	.02

Abbreviations: DR, diabetic retinopathy; NPDR, nonproliferative diabetic retinopathy; PDR, proliferative diabetic retinopathy.

Figure 2. Nonsegmented and Corresponding Binarized Optical Coherence Tomographic Angiography Images



A, No diabetic retinopathy (DR) (capillary density index [CDI], 0.358; fractal dimension [FD], 1.56). B, Mild nonproliferative diabetic retinopathy (NPDR) (CDI, 0.351; FD, 1.57). C, Moderate NPDR (CDI, 0.342; FD, 1.59). D, Severe NPDR (CDI, 0.340; FD, 1.60). E, Proliferative diabetic retinopathy (PDR) (CDI, 0.335; FD, 1.61). For all images, the top row is the nonsegmented optical coherence tomographic angiography image and the bottom row is the corresponding binarized optical coherence tomographic angiography image.

to assess for DR severity level. We showed that the CDI decreased with worsening severity of DR, while FD was positively correlated with increasing DR severity levels. Second, this study confirmed previous reports of a positive correlation between the retinal capillary FD with an increasing DR severity level, consistent with what had been published previously on larger retinal vessels.[17] Third, the changes of CDI and FD correlated between the SVP and DVP layers, suggesting that the retinal capillary ischemic changes and branching complexity of the retinal capillaries are diffuse processes in DR and systemic vascular diseases. Finally, more important, we showed that a range of systemic metabolic and vascular risk factors including hyperlipidemia, smoking, and renal impairment were associated with reduced CDI, while increased HbA_{1c} (≥8%) and renal impairment were associated with increased FD, suggesting that systemic risk factors could lead to retinal microvascular structural changes before clinical changes.

Previous studies had shown that both increased foveal avascular zone and decreased vessel density were associated with worsening DR severity.[18,19] Our results showed decreasing CDI with worsening DR severity, which is congruent with another study that used different algorithm to calculate vessel density on spectral domain (SD)-OCT-A.[9] The difference between each DR severity level is small, which could be owing to the limited 3 × 3-mm scanning width. The range of CDI in our study was between 0.3 to 0.4 for both SVP and DVP with higher CDI detected at deeper layers. This range of values was similar to the those reported in Kim et al.[9] There were slight differences in terms of the exact value owing to the patients' demographics (Asians vs Caucasians), use of a different machine (swept source- vs SD-OCT-A) and algorithm (split-spectrum amplitude-decorrelation angiography [SSADA] vs angiography ratio analysis). Future research could potentially compare the use of a different machine and algorithm with larger scanning width to allow better understanding on the overall status of the retinal capillary network in patients with diabetes.

Retinal vascular characteristics, including tortuosity, branching angle, FD, and ratio of length to diameter have all been linked to diabetes.[20-22] Although there is some debate,[9] accumulating evidence suggests an increasing trend in retinal capillary FD in patients with more severe DR. Given that FD had been shown to reflect arteriovenous differentiation after hypoxic insult during embryonic development of the retinal vasculature,[23] it may reflect the changes in the capillary network in response to hypoxic stimuli to increase blood flow in patients with more severe DR. Our study is the first to our knowledge to explore the FD association between the smaller capillary vessels and the larger retinal vessels. Further studies are warranted to evaluate the efficacy of retinal capillary

Table 3. Multivariate Analysis of Risk Factors for Low Capillary Density Index (Lower Quartile) and High Fractal Dimension (Upper Quartile)

	Low Capillary Density Index[b]		High Fractal Dimension[c]	
Systemic Vascular Risk Factor[a]	Odds Ratio (95% CI)	*P* Value[d]	Odds Ratio (95% CI)	*P* Value[d]
Glycated hemoglobin A_{1c} ≥8%	1.86 (1.10-3.58)	.95	8.77 (5.23-10.81)	<.01
Hypertension	9.60 (7.80-13.5)	.95	0.53 (0.15-0.92)	.47
Hyperlipidemia	9.82 (6.92-11.23)	<.001	0.36 (0.11-0.56)	.56
Smoking	10.90 (8.23-12.34)	<.001	0.18 (0.05-0.92)	.67
Renal impairment	3.72 (1.80-4.81)	.05	10.30 (8.21-11.91)	<.001

[a] All characteristics adjusted for age, sex, race/ethnicity, and severity level of diabetic retinopathy.

[b] Lower quartile of capillary density index value based on our study patients.

[c] Upper quartile of fractal dimension value based on our study patients.

[d] *P* value adjusted for age, sex, glycated hemoglobin HbA_{1c}, hyperlipidemia, renal impairment, and smoking.

FD to be a novel biomarker in prognosticating the incidence and progression of microvascular and macrovascular complications associated with diabetes.

Our study also showed that renal impairment was associated with a reduced CDI and high FD. These findings were consistent with those of a study conducted on larger retinal vessels,[24,25] suggesting that the damage of the retinal microvasculature may be associated with increased risk of chronic kidney disease. In our study, hyperlipidemia was also associated with reduced CDI, corroborating the experimental finding that hypercholesterolemia attenuates angiogenesis.[26] Hyperlipidemia had always been shown to be a risk factor for chronic kidney disease in patients with diabetes.[27-29] However, many studies had not been able to show any associations between hyperlipidemia and larger retinal vascular changes in patients with DR.[30,31]

In the multivariate analyses, low CDI was associated with smoking, which was previously shown to be an independent risk factor to decrease retinal blood flow in patients with diabetes.[32] Our study showed that smoking was associated with reduced CDI in patients with DR, suggesting ischemic changes at the level of the retinal microvasculature. The effect of smoking on retinal microcirculation has not been well studied previously owing to the difficulties of imaging the microvasculature.[33] Our study offered an insight to understand the consequence of smoking on the ischemic effect on the retinal capillary structure and density. We postulated that the reduction in retinal CDI secondary to smoking could lead to decreased retinal blood flow and oxygen reactivity, but this finding will need to be confirmed in future research in a larger pool of patients.

Strengths and Limitations

The strength of the study includes the prospective nature of patients' recruitment that allows future data collection on prognosticating incidence and progression of DR and other vascular complications. In addition, our study consists of a fair distribution of patients at all severity levels of DR, with sufficiently powered difference between the groups. We also reported the use of OCT-A to evaluate the effect of systemic vascular risk factors including hypertension, hyperlipidemia, smoking, and renal impairment on the retinal microvasculature in our cohort of patients with diabetes. Our study, however, has several limitations. First, the cross-sectional nature of the study limits our ability to draw definite conclusions in assessing the temporal patterns and long-term implications of the changes in the retinal microvasculature on the incidence and progression for DR, DME, and other systemic vascular complications. Larger numbers of patients with OCT-A should be evaluated in a prospective cohort before these findings can be fully validated. Despite the presence of the algorithm to reduce motion artifacts, some of the OCT-A images still had motion artifacts owing to patients' eye movement. We also observed a few projection artifacts, a well-described technical limitation,[34] at the deep levels of the OCT-A images. The few images with artifacts were excluded from analysis to avoid erroneous results. Given that this novel technology is still in its infancy, further enhancement and refinement of the software is essential to improve its reproducibility and usability for more systemic vascular diseases in the future.

Conclusions

We document structural changes in the retinal microvasculature associated with severity of DR and systemic metabolic and vascular risk factors in patients with diabetes. Optical coherence tomographic angiography is a new noninvasive tool to quantify the retinal capillary microvasculature to study diabetes and its complications, including complications not related to the eye. Optical coherence tomographic angiography has the potential to be used in larger epidemiologic and clinical studies, including interventional trials, to directly visualize the microvasculature that was previously not accessible in a noninvasive manner.

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Acquisition, analysis, or interpretation of data: Ting, Tan, Yanagi, Sie, C. W. Wong, Yeo, Lee, Cheung.
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Study supervision: Tan, Yeo, T. Y. Wong.

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