




Association of crossing capillaries in the finger nailfold with diabetic retinopathy in type 2 diabetes mellitus

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Keywords

Capillaries, Diabetic retinopathy, Nailfold capillaroscopy

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ABSTRACT

Aims/Introduction: Crossing capillaries in the finger nailfold might potentially be a novel diabetic retinopathy (DR) biomarker that could be assessed non-invasively in the clinical setting. However, the association between crossing capillaries and DR is controversial. This study aimed to investigate the association between the percentage of crossing capillaries in the finger nailfold and DR in patients with type 2 diabetes mellitus.

Materials and Methods: This cross-sectional study enrolled 108 type 2 diabetes mellitus patients (aged 40–75 years) who visited the outpatient diabetic clinic at Osaka University Hospital, Osaka, Japan, between May and October 2019. Capillary morphology was assessed using nailfold capillaroscopy based on the simple capillaroscopic definitions of the European League Against Rheumatism Study Group. Details of DR and other laboratory data were obtained from medical records. The association between the tertile of the percentage of the crossing capillary and DR was analyzed using multivariable logistic regression.

Results: After adjusting for age, sex, diabetes duration, glycated hemoglobin, systolic blood pressure, body mass index, and use of renin–angiotensin system inhibitor and anti-hyperlipidemic medication, the percentage of crossing capillaries was significantly associated with DR (multivariable-adjusted odds ratios for increasing tertiles of the percentage of crossing capillary: 1 [reference], 2.05 [95% confidence interval 0.53–7.94], and 4.33 [95% confidence interval 1.16–16.21]; *P*-trend = 0.028).

Conclusions: A higher percentage of crossing capillaries in the nailfold was associated with a higher risk of DR, independent of traditional risk and inhibiting factors, in patients with type 2 diabetes mellitus.

INTRODUCTION

Diabetic retinopathy (DR) is a major microvascular complication of diabetes and remains the leading cause of preventable blindness worldwide^{1–4}. Early diagnosis through annual screening for DR, and early, appropriate treatment can reduce the risk of blindness^{5,6}. However, given the increasing incidence and prevalence of type 2 diabetes mellitus, it is crucial to identify patients who are at a higher risk of developing DR based on risk factors and biomarkers, and confer a priority for the

access to ophthalmological services⁷. The major risk factors for the onset of DR (e.g., duration of diabetes, poor glycemic control, high systolic blood pressure [SBP] and high body mass index [BMI]) are well-established; however, these traditional risk factors are inadequate to predict the risk of DR^{4,7,8}. Therefore, novel clinical factors that are associated with DR should be identified for the prediction of the risk for onset of DR.

A previous cross-sectional study showed that among the abnormal capillary features, the presence of crossing capillaries in the finger nailfold was associated with an increased risk for DR⁹. As the microvascular morphology of the finger nailfold

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can be examined easily, non-invasively and safely¹⁰, crossing capillaries might potentially be a novel DR biomarker that could be used in the clinical setting. However, a recent cross-sectional study showed no significant association between crossing capillaries and the presence of DR¹¹. It should be noted, however, that these two studies evaluated the presence of crossing capillaries using semiquantitative capillary assessments, which could have resulted in low reliability^{9,11}. Furthermore, these previous studies were not adequately controlled for confounding factors that could affect the abnormal morphology of nailfold capillaries and DR^{9,11}.

Therefore, the present study aimed to assess the association between crossing capillaries of the finger nailfold and DR in patients with type 2 diabetes mellitus while controlling for confounding factors and by using a quantitative capillary assessment.

METHODS

Study design and population

The present cross-sectional study evaluated outpatients with type 2 diabetes mellitus aged 40–75 years between May and October 2019 at the Department of Metabolic Medicine, Osaka University Hospital, Osaka, Japan. The exclusion criteria included history of Raynaud phenomenon, collagen tissue disease, glaucoma, uveitis, dementia, cerebrovascular disease, coronary artery disease, dialysis, blindness, cancer treatment, smoking, removal of fingernail cuticle, use of nail polish at 1 month before the test and pregnancy. Of the 126 patients referred to the study by their physicians, we further excluded patients who lacked data on DR ($n = 15$) and metabolic profile ($n = 3$). Finally, 108 patients were evaluated.

The study adhered to the principles of the Declaration of Helsinki and its later amendments, and the protocol was approved by the institutional review board of Osaka University (approval no. 18546). All participants provided written informed consent.

Measurement of finger nailfold capillaroscopic findings

A GOKO Bscan-Z microscope with approximately $\times 390$ magnification (GOKO Imaging Devices Co., Kanagawa, Japan) was used to examine the finger nailfold capillaries (Figure 1a). The NTSC-USB 2.0 Converter was used to transmit images of the capillaroscopy to a computer, and PowerDirector 8 (CyberLink Corp., New Taipei City, Taiwan) was used to store the images.

The participants were seated in a temperature-controlled (23–26°C) room for 15–20 min to acclimatize and relax before the test¹². Three pictures per finger were taken in the middle of the nailfold finger, and eight fingers were examined, excluding the thumbs, for each participant¹⁰. To maximize the amount of light and improve image quality, a drop of vegetable oil was applied to the nailfold of each finger before the observation¹³. The procedure was carried out by a trained operator who was masked to the presence or absence of DR. The approximate size of the capillaroscopic image was 0.5×0.7 mm.² Fingers

with physical injuries or without skin transparency were excluded from the analysis¹³.

Two masked raters analyzed capillaries in the distal row of the nailfold. A regular capillary is shaped similar to a hairpin or an upside-down letter “U”, albeit with a thinner arterial arm, an upper part and a venous arm¹³. The nailfold capillary morphology was classified into crossing (the limbs cross once or twice and the capillary head is convex) or other (Figure 1b) types according to the simple capillaroscopic definitions of the European League Against Rheumatism Study Group^{14,15}. This definition, which has high reliability, was developed to standardize and simplify the definitions used to describe the morphology of a single capillary¹⁴. In the present study, the interrater reliability of simple capillaroscopic definitions was excellent (Cohen’s kappa 0.84). The percentage of crossing capillaries was calculated by dividing the number of crossing capillaries by the number of assessed capillaries (Figure 1c).

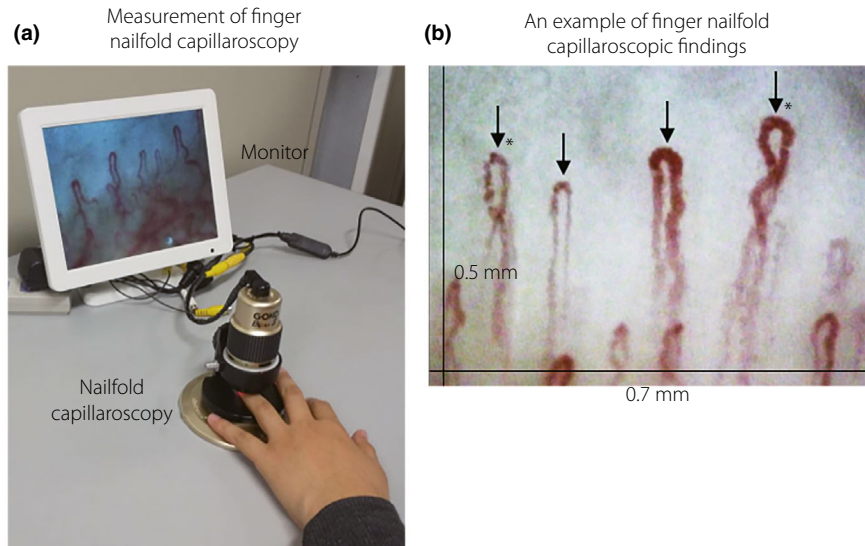
DR

The presence and severity of DR were determined by reviewing the patient’s medical records. DR was diagnosed and classified into no retinopathy, simple DR, pre-proliferative DR or proliferative DR, according to the Davis classification¹⁶.

Patient’ demographics and clinical characteristics

Anthropometric measurements (weight, height, waist circumference) and blood pressure (BP) were recorded by the medical staff. The BMI was calculated as weight (kg) divided by squared height (m^2). Waist circumference was measured at the umbilical level at the end of the exhalation phase with the patient standing upright. With the patient in the sitting position, BP was measured twice in the same arm by using a calibrated electronic sphygmomanometer (HEM-8713; Omron Corporation, Kyoto, Japan), and the mean of two measurements was calculated.

Information on age, sex, duration of diabetes, antihyperglycemic treatment, medication use (antihypertensive medication, renin–angiotensin system [RAS] inhibitor, antilipidemic medication), the most recent glycated hemoglobin (HbA1c) value, estimated glomerular filtration rate, total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglycerides (TGs) and urine findings were collected from the medical records. Blood sampling was usually carried out under fasting conditions. LDL-C was calculated using the Friedewald equation¹⁷. If the TGs exceeded 4.5 mmol/L (400 mg/dL), or if data were missing for total cholesterol, HDL-C or TGs, we used the LDL-C value measured using the direct method. The estimated glomerular filtration rate was calculated by using the formula specified by the Japanese Society of Nephrology¹⁸. Nephropathy was classified into five stages based on the 2014 Classification of Diabetic Nephropathy in Japan, as follows: (i) stage 1, pre-nephropathy: urinary albumin-to-creatinine ratio < 30 mg/g creatinine; (ii) stage 2, incipient nephropathy: $30 \leq$ urinary albumin-to-



(c) An example of the calculation of the percentage of crossing capillary of the finger nailfold in a patient

	Number of assessed nailfold capillaries [†]	Number of crossing capillaries [‡]	Percentage of crossing capillaries [§]
Patient 1	90	60	66.7%

Figure 1 | Assessment of the finger nailfold capillary. (a) A representative image of the measurement of finger nailfold capillaroscopy. (b) An example of finger nailfold capillaroscopic findings. Arrows indicate the nailfold capillaries assessed in the distal row. The asterisk (*) indicates crossing capillary (the limbs cross once or twice and the capillary head is convex). (c) An example of the calculation of the percentage of crossing capillary of the finger nailfold in a patient. [†]Number of assessed nailfold capillaries: sum of the number of capillaries assessed in the distal row of the nailfold in eight fingers. [‡]Number of crossing capillaries: sum of the number of the crossing capillaries in the distal row of the nailfold in eight fingers. [§]Percentage of crossing capillaries per patient: the number of crossing capillaries divided by the number of assessed nailfold capillaries.

creatinine ratio < 300 mg/g creatinine; (iii) stage 3, overt nephropathy: urinary albumin-to-creatinine ratio ≥ 300 mg/g creatinine or urinary protein-to-creatinine ratio ≥ 0.5 g/g creatinine or positive proteinuria by dipstick analysis examination; (iv) stage 4, kidney failure: estimated glomerular filtration rate < 30 mL/min/1.73²; and (v) stage 5, dialysis therapy¹⁹. Patients with stage 5 nephropathy were excluded from this study.

Metabolic syndrome was defined based on the Japanese Society of Internal Medicine criteria²⁰, and was diagnosed when a patient had an increased waist circumference (≥ 85 or ≥ 90 cm for male or female, respectively) plus at least two of three additional components: (i) impaired glucose metabolism (fasting blood glucose ≥ 6.1 mmol/L [110 mg/dL] and/or HbA1c ≥ 42 mmol/mol [6.0%] or medication use for diabetes); (ii) elevated BP (SBP ≥ 130 mmHg and/or diastolic BP ≥ 85 mmHg or antihypertensive drug treatment); and (iii) elevated TGs (≥ 1.7 mmol/L [150 mg/dL] or drug treatment for elevated TGs) and/or reduced HDL-C (<1.0 mmol/L [40 mg/dL] or drug treatment for reduced HDL-C). Hypertension was defined as SBP ≥ 140 mmHg, diastolic BP ≥ 90 mmHg and/or use of antihypertensive drugs²¹. Dyslipidemia was defined as LDL-C ≥ 3.6 mmol/L (140 mg/dL), HDL-C <1.0 mmol/L (40 mg/

dL), TGs ≥ 1.7 mmol/L (150 mg/dL) or use of antilipidemic medications²².

Regular exercise was defined as exercising two or more times per week for at least 30 min each time for at least 6 months. Regular alcohol consumption was defined as drinking any amount of alcohol one or more times per week.

Statistical analysis

Patients were classified into tertiles of the percentage of crossing capillaries. Tertile 1 indicated the tertile with the lowest percentage of crossing capillaries. Differences between patient characteristics according to the tertiles of the percentage of crossing capillaries were analyzed using the Cochran–Armitage and Jonckheere–Terpstra tests for trend for proportions and continuous variables, respectively.

The presence or absence of DR according to the tertiles of the percentage of crossing capillaries was analyzed using the Cochran–Armitage test for trend. The association between tertiles of the percentage of crossing capillaries and the presence of DR was evaluated using multivariable logistic regression models (response variable: 1 = presence of retinopathy and 0 = absence of retinopathy), using the lowest category

(tertile 1) as the reference group. We adjusted for potential confounding factors by traditional risk and inhibiting factors, including age, sex, duration of diabetes, HbA1c, SBP, BMI, use of RAS inhibitors and use of antilipidemic medication. For linear trend analyses, the categorical variable tertile of the percentage of crossing capillaries was used in the regression models.

All *P*-values were two-tailed, and *P* < 0.05 was considered statistically significant. All analyses were carried out using IBM SPSS Statistics, version 24.0 (IBM SPSS, Armonk, NY, USA).

RESULTS

In total, 31 (28.7%) of the 108 patients were diagnosed with DR. In this study population, DR was present in 28.7% of the 108 patients. The mean (\pm standard deviation) number of nailfold capillaries assessed per patient was 93.7 (\pm 25.8). Among them, the mean (\pm standard deviation) percentage of crossing capillaries was 59.6% (\pm 11.7%).

Table 1 shows the clinicodemographic characteristics of the overall population and according to the tertiles of the percentage of crossing capillaries. The proportion of patients who used RAS inhibitors and antilipidemic medication tended to increase as the percentage of crossing capillaries increased (*P* for trend < 0.100).

As shown in Table 2, 22.2, 27.8 and 36.1% of the patients in the first, second and third tertiles of crossing capillaries had DR, respectively (*P* for trend = 0.195). The multivariable-adjusted odds ratios (ORs) and 95% confidence interval (95% CI) for DR according to the tertile of the percentage of crossing capillaries are shown in Table 2. After adjustment for potential confounding factors and comparison with patients with the lowest percentage of crossing capillaries (reference category), the multivariable-adjusted ORs for DR according to the tertiles of the percentage of crossing capillaries were 2.05 (95% CI 0.53–7.94) and 4.33 (95% CI 1.16–16.21) in patients with the middle and highest percentages of crossing capillaries, respectively. There were significant linear increases in the ORs for DR (*P* for trend = 0.028). In the multivariable analysis (model 2), age (adjusted OR 0.92, 95% CI 0.86–0.996) and duration of diabetes (adjusted OR 1.12, 95% CI 1.04–1.20) were associated with DR. Furthermore, when data on the crossing capillary were added to a model that included the known risk and inhibiting factors, the Nagelkerke *R*² increased from 0.29 to 0.35. In an additional analysis, the results were not substantially altered when we excluded patients without screening for DR within 2 years of the study (Table S1).

The proportion of DR severity according to the tertile of crossing capillaries is shown in Table 3. The proportion of those with pre-proliferative or proliferative retinopathy tended to increase as the percentage of crossing capillaries increased. The proportion of diabetic nephropathy severity according to the tertile of crossing capillary is shown in Table 4. The proportion of those with overt nephropathy and kidney failure tended to increase as the percentage of crossing capillaries increased. However, these analyses were hampered by power loss.

DISCUSSION

Although crossing capillaries in the finger nailfold might potentially be a novel non-invasive DR marker, the association between crossing capillaries and DR is yet to be clarified. In this study, a higher percentage of crossing capillaries in the finger nailfold was positively associated with DR in patients with type 2 diabetes mellitus. This association is independent of traditional risk and inhibiting factors for DR, including age, sex, duration of diabetes, HbA1c, SBP, BMI, and the use of RAS inhibitor and antilipidemic medication.

Several cross-sectional studies have reported that crossing capillaries were the most frequently observed features among abnormal finger nailfold morphologies in type 2 diabetes mellitus patients^{11,23–25}. However, the association between crossing capillaries and DR is controversial due to uncontrolled confounding factors and the lack of semiquantitative assessment of nailfold capillaries^{9,11}. A previous study defined the presence of crossing capillaries based on whether two or more crossing capillaries existed per 1-mm length⁹. Another study defined the presence of crossing capillaries by determining whether >50% of the assessed capillaries were evaluated as crossing capillaries¹¹. Compared with previous studies, the present study includes more reliable data because of the use of a standardized quantitative assessment of nailfold capillaries. Furthermore, to our best knowledge, the present study is the first to report an association between the percentage of crossing capillaries of the finger nailfold and DR, after adjusting for potential confounding factors.

The etiological mechanism underlying crossing capillaries is unclear, although the findings of the present study imply the likelihood of a common pathological pathway that produces both the finger crossing capillaries and DR in the diabetic milieu. Previous studies have shown that early morphological changes of the retinal capillaries in DR are associated with the loss of pericytes in the vascular bed of the retina, both *in vivo* and *in vitro*^{26,27}. Pericytes are present in all microvasculature, and support and stabilize the structural microvasculature^{26,28}. Pericyte loss is believed to be an early hallmark of diabetes-associated microvascular disease, including retinopathy and nephropathy²⁶. It is also observed in the dermal capillary networks of patients with diabetes²⁹. In addition, a study of Alzheimer's disease, wherein pericyte loss is observed³⁰, showed that a greater severity of crossing capillaries of the finger nailfold was associated with a 10.6-fold increased risk of Alzheimer's disease dementia after adjustment for confounding factors³¹. Thus, pericyte loss in the nailfold capillary might lead to instability of the capillary, resulting in an increased number of finger crossing capillaries.

The present study found that crossing capillaries are associated with DR, independent of the traditional risk and inhibiting factors for DR. Furthermore, when data on the crossing capillary were added to a model that included the known risk and inhibiting factors, the *R*² of the model increased by 6% from

Table 1 | Clinicodemographic patient characteristics

Characteristics	All (<i>n</i> = 108)	Tertiles of the crossing capillaries (%)			<i>P</i> for trend
		<56.9 (<i>n</i> = 36)	56.9–63.2 (<i>n</i> = 36)	≥63.2 (<i>n</i> = 36)	
Crossing capillary (%)	59.6 ± 11.7	47.4 ± 8.9	59.7 ± 1.9	71.9 ± 5.7	
Age (years)	63.9 ± 8.7	62.2 ± 9.5	65.6 ± 8.7	64.0 ± 7.5	0.549
Male (%)	54.6	50.0	63.9	50.0	>0.999
Smoking, former (%)	36.1	36.1	33.3	38.9	0.807
Regular alcohol consumption (%)	25.9	25.0	27.8	25.0	>0.999
Regular exercise, absence (%)	49.1	61.1	38.9	47.2	0.241
Duration of diabetes (years)	14.9 ± 9.0	14.9 ± 10.4	16.3 ± 9.1	13.5 ± 7.4	0.756
Antihyperglycemic medication use (%)	92.6	94.4	91.7	91.7	0.654
Oral medication (%)	87.0	86.1	88.9	86.1	>0.999
GLP-1 analogs (%)	9.3	5.6	11.1	11.1	0.418
Insulin (%)	18.5	19.4	16.7	19.4	>0.999
HbA1c (mmol/mol)	55.4 ± 10.0	54.3 ± 7.8	57.1 ± 10.6	54.8 ± 11.4	0.826
HbA1c (%)	7.2 ± 0.9	7.1 ± 0.7	7.4 ± 1.0	7.2 ± 1.0	0.826
eGFR (mL/min/1.73 m ²)	68.2 ± 21.6	71.4 ± 20.7	66.5 ± 23.2	66.6 ± 21.1	0.582
HDL cholesterol (mmol/L)	1.5 ± 0.4	1.5 ± 0.4	1.5 ± 0.4	1.6 ± 0.3	0.323
LDL cholesterol (mmol/L)	2.8 ± 0.	3.0 ± 0.7	2.7 ± 0.6	2.8 ± 0.7	0.240
Triglycerides (mmol/L)	1.3 (0.8, 1.9)	1.4 (0.9, 1.8)	1.4 (0.9, 2.0)	1.0 (0.8, 1.5)	0.218
SBP (mmHg)	133.1 ± 17.0	133.1 ± 15.0	134.6 ± 18.4	131.6 ± 17.7	0.491
DBP (mmHg)	80.8 ± 10.4	81.6 ± 11.6	82.2 ± 10.2	78.5 ± 9.3	0.202
Waist circumference (cm)	91.7 ± 12.1	90.2 ± 13.7	91.8 ± 12.4	93.1 ± 10.0	0.160
Body mass index (kg/m ²)	25.6 ± 4.6	25.1 ± 5.4	25.7 ± 4.6	25.9 ± 3.7	0.256
Metabolic syndrome (%)	60.2	58.3	55.6	66.7	0.472
Hypertension (%)	72.2	77.8	72.2	66.7	0.295
Dyslipidemia (%)	79.6	75.0	80.6	83.3	0.382
Antihypertensive medication use (%)	59.3	58.3	58.3	61.1	0.811
Renin–angiotensin system inhibitor (%)	46.3	33.3	50.0	55.6	0.060
Lipid-modifying medication use (%)	62.0	50.0	66.7	69.4	0.091

Continuous data were analyzed using the Jonckheere–Terpstra test for trend and presented as mean ± standard deviation or median (25th percentile, 75th percentile), as appropriate. Categorical data were analyzed using the Cochran–Armitage test for trend and presented as the percentage. The percentage of crossing capillaries was calculated by dividing the number of crossing capillaries by the number of assessed nailfold capillaries. Metabolic syndrome was defined by the criteria of the Japanese Society of Internal Medicine. Hypertension was defined as systolic blood pressure (SBP) ≥140 mmHg, diastolic blood pressure (DBP) ≥ 90 mmHg, and/or use of antihypertensive drugs. Dyslipidemia was defined as low-density lipoprotein (LDL) cholesterol ≥3.6 mmol/L (140 mg/dL), high-density lipoprotein (HDL) cholesterol <1.0 mmol/L (40 mg/dL), triglycerides ≥1.7 mmol/L (150 mg/dL) or use of antilipidemic medications. eGFR, estimated glomerular filtration rate; GLP, glucagon-like peptide; HbA1c, glycated hemoglobin; SD, standard deviation.

0.29 to 0.35. A previous clinical study showed that traditional clinical risk factors for DR explained only a fraction of the variation in the risk of DR. For example, the Diabetes Control and Complications Trial showed that HbA1c values and duration of diabetes explained no more than 11% of the risk of DR^{32,33}. The present findings show that crossing capillaries in the finger nailfold might be a novel imaging biomarker for DR that could be related to residual risk. In addition, crossing capillaries of the finger nailfold might contribute to the prediction and early management of the risk of DR when combined with known risk factors.

Despite the small number of patients with diabetic nephropathy assessed in the present study, the proportion of those with overt nephropathy and kidney failure tended to increase along with a higher percentage of crossing capillaries. This shows that

those with a higher percentage of crossing capillaries might have a higher risk of diabetic nephropathy.

Nailfold capillaroscopy is a portable, easy and non-invasive tool for the assessment of the microvascular structure, which can be made available in a clinical setting³⁴. In addition, patients can confirm their microvascular damage visually through a monitor. Furthermore, the increased percentage of crossing capillaries might have potential for use as an indicator for the promotion of diabetes self-care and adherence to DR screening.

The present study had some limitations. First, because of its cross-sectional design, we could not establish the time course of the associations between finger nailfold crossing capillaries and DR. Second, we defined the presence of DR based on a review of medical records. However, the results were not substantially

Table 2 | Odds ratio for diabetic retinopathy according to tertiles of the percentage of crossing capillaries in patients with type 2 diabetes

Variables	Patients with DR, % (case/n)	Odds ratio (95% confidence interval)	
		Model 1	Model 2
Crossing capillary			
Tertile 1: <56.9 (%)	22.2 (8/36)	Reference	Reference
Tertile 2: 56.9–63.2 (%)	27.8 (10/36)	1.45 (0.48–4.35)	2.05 (0.53–7.94)
Tertile 3: ≥63.2 (%)	36.1 (13/36)	2.01 (0.71–5.74)	4.33 (1.16–16.21)
Age (per 1 year)			0.92 (0.86–0.996)
Male (vs female)			0.40 (0.14–1.16)
Duration of diabetes (per 1 year)			1.12 (1.04–1.20)
HbA1c (per 1 unit)			1.37 (0.76–2.48)
Body mass index (per 1 unit)			0.86 (0.73–1.003)
Systolic blood pressure (per 1 unit)			1.03 (0.998–1.06)
Renin–angiotensin system inhibitor use (vs none)			1.75 (0.59–5.22)
Lipid-modifying medication use (vs none)			0.41 (0.14–1.25)

Model 1: adjusted for age and sex. Model 2: adjusted for age, sex, duration of diabetes, glycated hemoglobin (HbA1c), body mass index, systolic blood pressure, use of renin–angiotensin system inhibitor and use of antilipidemic drugs. Response variable: 1 = presence of diabetic retinopathy (DR), 0 = absence of DR. HbA1c, glycated hemoglobin.

Table 3 | Proportion of diabetic retinopathy severity according to the tertile of crossing capillaries

Stage of diabetic retinopathy	Tertile of crossing capillaries (%)			P-value
	<56.9 (n = 35)	56.9–63.2 (n = 36)	≥63.2 (n = 36)	
No diabetic retinopathy (%)	80.0	72.2	63.9	0.585
Simple retinopathy (%)	11.4	16.7	16.7	
Pre-proliferative retinopathy or proliferative retinopathy (%)	8.6	11.1	19.4	

The stage of diabetic retinopathy was analyzed using the χ^2 -test and presented as the percentage. Diabetic retinopathy was evaluated based on the Davis classification. Available in 107 patients (excluded a patient with no information about the stage of diabetic retinopathy).

Table 4 | Proportion of diabetic nephropathy severity according to the tertiles of crossing capillaries

Stage of diabetic nephropathy	Tertile of the crossing capillaries (%)			P-value
	<57.3 (n = 30)	57.3–65.3 (n = 29)	≥65.3 (n = 30)	
Stage 1 (pre-nephropathy)	70.0	65.5	70.0	0.082
Stage 2 (incipient nephropathy)	23.3	27.6	6.7	
Stage 3 (overt nephropathy) or stage 4 (kidney failure)	6.7	6.9	23.3	

Diabetic nephropathy severity was analyzed using the χ^2 -test and presented as the percentage. The stage of diabetic nephropathy was assessed based on the 2014 Classification of Diabetic Nephropathy. Available in 89 patients (excluded patients with other renal diseases or without the urine examination within 1 year).

altered when we excluded patients without screening for DR within 2 years before the study. Third, we could not adequately assess the association between finger nailfold crossing capillaries and DR severity because of the small number of patients with retinopathy. Finally, our sample was limited to the Asian population, which might limit extrapolation and generalization of these findings to other regions or ethnic groups.

In conclusion, a higher percentage of crossing capillaries in the finger nailfold was significantly associated with an increased risk of DR in patients with type 2 diabetes mellitus after adjusting for confounding factors. Prospective studies are required to confirm whether an increased number of crossing capillaries in the finger nailfold can predict the risk of DR onset in patients with type 2 diabetes mellitus.

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DISCLOSURE

The authors declare no conflict of interest.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1 | Odds ratio for diabetic retinopathy according to tertiles of the percentage of crossing capillary in patients with type 2 diabetes mellitus except for those without screening for diabetic retinopathy within 2 years ($n = 85$).