tvst

Artificial Intelligence

Characterization of the Retinal Microvasculature and FAZ Changes in Ischemic Stroke and Its Different Types

Hongyu Duan[1,3], Jianyang Xie[2], Yifan Zhou[3], Hui Zhang[4], Yiyun Liu[3], Chuhao Tang[3], Yitian Zhao[2], and Hong Qi[1,3]

[1] Institute of Medical Technology, Peking University Health Science Center, Beijing, China
[2] Cixi Institute of BioMedical Engineering, Ningbo Institute of Materials Technology and Engineering, Chinese Academy of Sciences, Ningbo, China
[3] Department of Ophthalmology, Peking University Third Hospital, Beijing Key Laboratory of Restoration of Damaged Ocular Nerve, Beijing, China
[4] Department of Neurology, Peking University Third Hospital, Beijing, China

Correspondence: Hong Qi, Department of Ophthalmology, Peking University Third Hospital, Beijing Key Laboratory of Restoration of Damaged Ocular Nerve, 49 North Garden Rd, Haidian District, Beijing 100191, China. e-mail: doctorqihong@163.com
Yitian Zhao, Cixi Institute of BioMedical Engineering, Ningbo Institute of Materials Technology and Engineering, Chinese Academy of Sciences, No. 1219, Zhongguan West Road, Zhenhai District, Ningbo 315200, China. e-mail: yitian.zhao@nimte.ac.cn

Received: March 17, 2022
Accepted: September 16, 2022
Published: October 14, 2022

Keywords: ischemic stroke; optical coherence tomography angiography; retinal microvasculature; foveal avascular zone

Citation: Duan H, Xie J, Zhou Y, Zhang H, Liu Y, Tang C, Zhao Y, Qi H. Characterization of the retinal microvasculature and FAZ changes in ischemic stroke and its different types. Transl Vis Sci Technol. 2022;11(10):21, https://doi.org/10.1167/tvst.11.10.21

Purpose: This study aimed to assess morphological changes in the retinal microvasculature and foveal avascular zone (FAZ) in patients with ischemic stroke and its different subtypes.

Methods: Thirty-three patients with ischemic stroke (14 with nonlacunar infarction and 19 with lacunar infarction) and 27 control participants were enrolled in this study. Based on optical coherence tomography angiography (OCTA), three vascular parameters, including vascular area density, vascular fractal dimension (VFD), and vascular orientation distribution (VOD), and four FAZ-related parameters, including FAZ area, FAZ axis ratio (FAR), FAZ circularity (FC), and FAZ roundness, in the superficial capillary plexus (SCP) and deep capillary plexus (DCP) were extracted and analyzed.

Results: Logistic regression results showed that worse best-corrected visual acuity (odds ratio [OR], 0.21), higher FAR (OR, 2.77) and lower FC (OR, 0.36) of the DCP were associated with ischemic stroke. Furthermore, lower VOD of the SCP was significantly associated with lacunar infarction compared with nonlacunar infarction.

Conclusions: Our study shows that the FAR and FC of the DCP may be potential biomarkers of ischemic stroke. Moreover, we demonstrated that OCT showed specific damage patterns in retinal microvascular and macular morphology in different subtypes of ischemic stroke.

Translational Relevance: This work lays the foundation for the pathophysiological characteristics of cerebrovascular diseases assisted by retinal imaging and artificial intelligence.

Introduction

Stroke is a leading cause of mortality and disability worldwide, and the financial burden of treatment and poststroke care is high.[1] In 2013, it accounted for almost 6.5 million deaths and 25.7 million stroke survivors. Ischemic stroke accounts for approximately 71% of all strokes.[2] There is extensive evidence that microvascular damage, reflecting small vessel disease, is a major cause of ischemic stroke.[3] However, existing neuroimaging techniques may not allow direct visualization of minor changes in vivo owing to resolution.[4]

Lacunar infarctions are responsible for approximately 25% of ischemic strokes, which are defined as small infarction lesions (3–15 mm diameter) in the deep perforating arterial territory. Although lacunar infarction has been an recognized stroke subtype for more than 50 years, its causes and whether it differs from cortical stroke are still being debated.[5,6] Currently, the prevention and treatment of this common ischemic stroke subtype is still unsatisfactory. Therefore, study of microvasculature abnormalities in other vascular beds (e.g., the retina, where small vessels can be directly observed) is beneficial to understand the pathophysiological mechanisms of lacunar infarction.[7]

Retinal vessels are similar to brain vessels in embryonic origin, anatomic features, and physiological characteristics and hence provide a new window for exploring the pathophysiology of ischemic stroke in vivo.[8] Previous studies based on fundus photography have revealed an association between retinal vessel damage (traditional retinopathy and retinal vascular diameter changes) and cerebrovascular disease.[9] Nonetheless, fundus photography only detects retinal microvasculature changes confined to arterioles and venules (100–300 µm in diameter), but cannot resolve the finer retinal capillaries (5–6 µm in diameter), which are more representative of the entire microvascular network. In addition, the low resolution and limited imaging depth of fundus photography limit the imaging of the retinal deep layer.

Optical coherence tomography angiography (OCTA) is a novel, noninvasive, contrast-free, and high-resolution imaging technique that allows real-time quantitative evaluation of the retinal vasculature and structure of the foveal avascular zone (FAZ). OCTA can detect a range of changes (decreased in capillary and perfusion density) before apparent damage appears on retinal photographs.[10] In addition, OCTA technology combined with artificial intelligence contributes to automating the detection and quantification of ophthalmic diseases, particularly diabetic retinopathy.[11,12] Artificial intelligence techniques, such as deep learning, are more accurate and efficient in detecting subclinical changes than operators. In recent years, OCTA has been used to assess retinal microvascular dysfunction in many cardiovascular and cerebrovascular diseases and neurodegenerative diseases, including acute coronary syndrome,[13] cerebral small vessel disease, and Alzheimer's disease.[14,15] Nonetheless, almost all these previous studies used built-in software, which provided limited information on the retina. In contrast, a deep learning algorithm based on manual labeling can retain more blood vessels topology information, which is conducive to the exploration of new clinical indicators.

Currently, the association between ischemic stroke and abnormal findings on OCTA has not yet been explored based on machine learning. In the present study, we used OCTA combined with machine learning to quantitatively assess the retinal microvasculature of the superficial capillary plexus (SCP) and deep capillary plexus (DCP), with the aim of detecting the geometrical changes of the retinal microvasculature and FAZ in patients with ischemic stroke and its different subtypes.

Materials and Methods

Study Participants

This cross-sectional study was conducted in the ophthalmology department of Peking University Third Hospital from January to September 2021. Patients diagnosed with ischemic stroke using magnetic resonance imaging were scheduled for OCTA examination (Fig. 1). The diagnosis of ischemic stroke was obtained from previous diagnostic reports or electronic medical records, according to the diagnostic criteria of the Trial of Org 10172 in Acute Stroke Treatment.[16] The patients were divided into two major

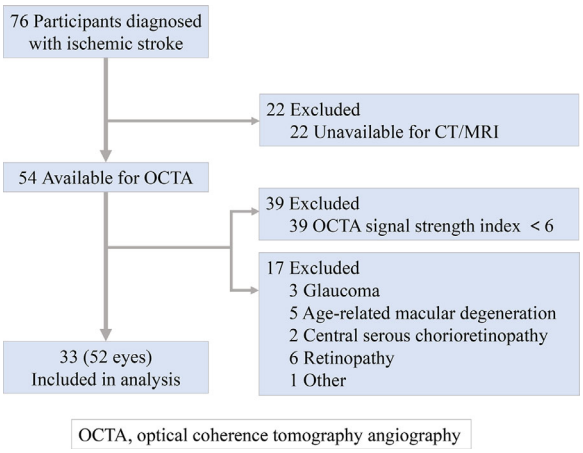

Figure 1. Flow diagram of the inclusion and exclusion process. CT, computed tomography; MRI, magnetic resonance imaging.

groups: (1) lacunar infarction and (2) other types of ischemic stroke (nonlacunar infarction). This study was approved by the Research Ethics Committee of Peking University Third Hospital (registration number: M2021538) and was performed in accordance with the Declaration of Helsinki. Informed consent was obtained from all the participants before the study.

Exclusion criteria were (1) other neurological disorders affecting retinal microvasculature, such as Alzheimer's disease and Parkinson's disease; (2) retinal diseases or diseases affecting vessel structure in fundus photography, including glaucoma, age-related macular degeneration, central serous chorioretinopathy, hypertensive or diabetic retinopathy, and other ocular pathologies; (3) eye diseases that significantly affect best-corrected visual acuity (BCVA), such as cataract and keratopathy; (4) poor cooperation in ophthalmic examinations; (5) high refractive error (spherical equivalent $>+6.00$ D or <-6.00 D); and (6) low-quality OCTA images (signal strength index of $<6/10$). The inclusion criterion for healthy controls was no history of ischemic stroke or other neurological or ocular diseases.

All participants underwent the following ophthalmic examinations: BCVA, slit-lamp biomicroscopy, fundus photography, and OCTA.

OCTA Imaging and Processing

OCTA images were obtained after pupillary dilation using a spectral domain OCT system (AngioVue, RTVue XR Avanti spectral domain OCT, Optovue, Fremont, CA). The instrument takes 304 × 304 volumetric A-scans with 70,000 A-scans per second, and the split-spectrum amplitude-decorrelation angiography algorithm was used to detect flow. OCTA parameters were analyzed using 3 × 3 images centered on the fovea. Images with low quality, artifacts, and local signal loss were excluded. The angiography scan was automatically segmented into SCP and DCP using the built-in software split-spectrum amplitude-decorrelation angiography. The SCP was defined as the internal limiting membrane to 10 µm above the inner plexiform layer. The DCP was 10 µm above the inner plexiform layer to 10 µm below the outer plexiform layer. Both eyes of each individual were examined and analyzed unless the image quality of one eye failed.

OCTA Metrics From a Machine Learning–Based Method

All of the parameters were calculated based on the segmentation result of vasculature and FAZ. In our method, these segmentation maps were obtained by using a machine learning–based method. The specific methods are described in detail in our previous studies. The OCTA-Net[17] was applied on the vascular segmentation in our method, and the FAZ was segmented by applying the lever set–based method.[18]

The following three retinal microvascular parameters were quantitatively extracted and analyzed (Fig. 2).

Vascular Parameters

Vascular Area Density (VAD). The total length in millimeters of perfused retinal microvasculature per unit area in square millimeters in the region of the analyzed area.

Vascular Fractal Dimension (VFD). A well-known parameter that indicates the degree of geometric complexity of the vasculature.

Vascular Orientation Distribution (VOD). This calculates the direction of each vascular pixel to indicate the trend of blood flow. The VOD is an innovative metric that was proposed for the first time.

The VOD is represented as the direction histogram in polar coordinates, where the direction of each vascular pixel is calculated using the hessian matrix. In this study, the multiscale hessian algorithm[19] was applied to detect vascular directions with various widths. As shown in Figure 3, the direction of each vascular pixel is represented as an angle from 0° to 180°. To satisfy the angle range in polar coordinates, we doubled the angle of each pixel. A polar plot of orientation distribution was then generated to show the number of pixels at each angle from 0° to 360°. This orientation distribution curve exhibits a unique pattern in the vasculature map of the OCTA image. Quantitative measures of the vessel orientation pattern can be achieved by analyzing the polar plot region encompassed by the orientation distribution curve. According to the observation that the orientation pattern for the OCTA vasculature depicts a roughly elliptical shape, the ellipse fitting method proposed in a previous study[20] was used to estimate the general equation of the ellipse, and then the major and minor axes were calculated with the fitting result. Finally, the VOD is represented as the ratio of the lengths of the major and minor axes. The VOD is a representation of vessel anisotropy; a larger VOD indicates that the flow of the vasculature tends to be consistent and that there are fewer curves on it, and vice versa.

Second, we used the FAZ area and three different FAZ metrics (axial ratio, circularity, and roundness) to characterize the size and shape of the FAZ (Fig. 2).

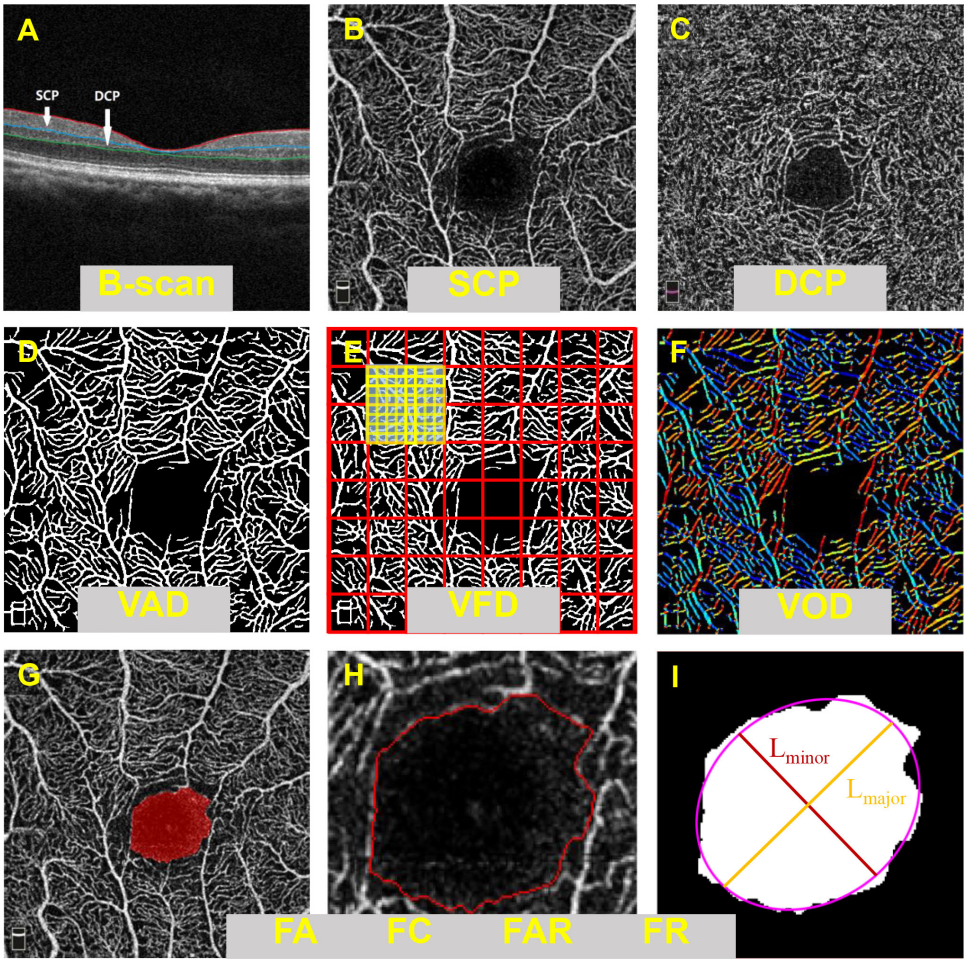

Figure 2. Vascular and FAZ-related parameters used in the quantitative measurements. (A) B-scan of the sample OCT volume with illustrated retinal layers. (B) and (C) show 3 × 3 mm^2 en face angiograms of the SCP and DCP, respectively. (D) shows the automated segmented vessel map of (B). VAD can be derived by calculating the ratio of the vasculature in (D). (E) and (F) show vascular fractal dimension (VFD) and VOD, respectively. (G) Detected FAZ area (FA). (H) The FP. (I) The major axis (*orange line*) and minor axis (*red line*) of the fitted ellipse of the FAZ. FC = 4π*FA/FP^2; FAR = L_{major}/L_{minor}; *FAZ Roundness (FR)* and *Vascular Fractal Dimension (VFD)* was given in method. FR = 4π*FA/L_{major}^2; L_{major} = length of major axis; L_{minor} = length of minor axis.

FAZ-Related Parameters

FAZ Area (FA). Total number of pixels in the FAZ region.

FAZ Axis Ratio (FAR). The ratio between the major and minor axes of the fitted ellipse from the FAZ boundary. The axis ratio of a perfect circular FAZ is equal to 1. A higher FAR indicates an elongated FAZ with greater eccentricity.

FAZ Circularity (FC). The ratio of the perimeter of the FAZ to the perimeter of a circle with equal area, reflecting the degree of roundness of the FAZ.

FAZ Roundness. Similar to FC; however, it is less sensitive to irregular borders along the perimeter of the FAZ.

Statistical Analyses

Statistical analysis was performed using software (SPSS, 23.0; SPSS Inc., Chicago, IL). Quantitative data are expressed as mean ± standard deviation. Qualitative data are expressed as medians (interquartile range). An unpaired Student *t* test or rank-sum test was used when data were normally or non-normally distributed, respectively. The χ^2 test was used for qualitative data. Ischemic stroke and control (or lacunar infarction and nonlacunar infarction in subtypes analyses) was taken as an independent variable, and baseline and OCTA parameters between the two groups were analyzed using univariate regression. The baseline characteristics with statistically significant differences were entered into a multifactorial regression model.

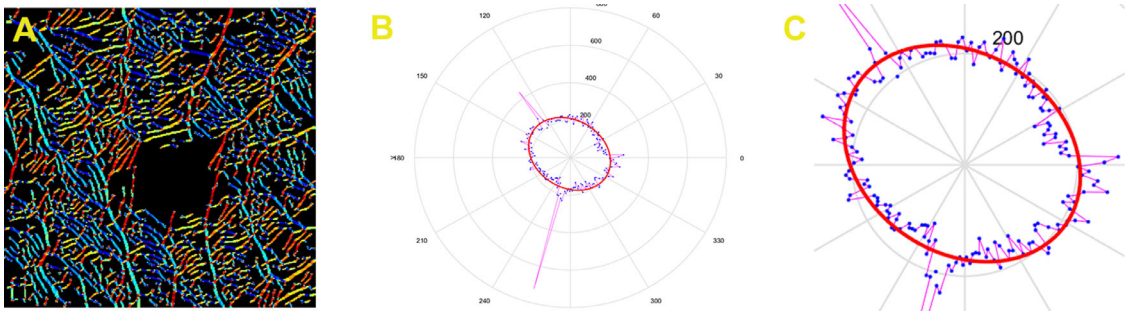

Figure 3. The VOD. (A) A color-encoded map describing the VOD of the OCTA image, where similar colors indicate the same direction of blood flow. (B) Polar plot showing the number of vascular pixels in (A) from 0° to 360° based on their directions. (C) illustrates the fitted ellipse of the orientation distribution points and shows the major axis (*blue line*) and minor axis (*green line*). L_{major} = length of major axis; L_{minor} = length of minor axis; VOD = L_{major}/L_{minor}.

The results of the regression analysis were expressed as odds ratios (ORs), 95% confidence intervals (CIs), and *P* values. Statistical significance was set at a *P* value of less than .05. To make the sum of *P* value for multiple testing to be 0.05, the *P* value of two logistic regressions is corrected to 0.025.

Results

Study Population

A total of 33 participants with ischemic stroke and 27 healthy controls were enrolled. The demographic and clinical information of the two groups are shown in Table 1. There were significant differences between the two groups in terms of age ($P < 0.001$), diabetes ($P = 0.005$), and BCVA ($P < 0.001$). No significant differences ($P > 0.05$) were observed in sex, duration of stroke, hypertension, or signal strength index.

Logistic Regression Analysis

Univariate logistic regression analysis (Table 2) revealed that older individuals (OR, 2.91) was associated with ischemic stroke, whereas participants with a worse BCVA (OR, 0.31) was associated with ischemic stroke. Regarding OCTA parameters in the SCP, participants with higher FAR (OR, 2.25) and lower FR (OR, 0.54) were associated with ischemic stroke. For DCP parameters, a lower VAD (OR, 0.59), higher FAR (OR, 2.34), and lower FC (OR, 0.51) were associated with ischemic stroke. Finally, two factors that were statistically significant in baseline characteristics (age and BCVA) and four OCTA parameters (FAR of SCP, VAD, FAR, and FC of DCP) were included in the multivariate logistic regression model.

Multivariate logistic regression analysis (Table 2) showed that worse BCVA (OR, 0.21), higher FAR (OR, 2.77) of DCP, and lower FC (OR, 0.36) of DCP were associated independently with ischemic stroke. The remaining parameters were not statistically significant.

Table 1. Demographic and Clinical Information of Participants

Characteristics*	Ischemic Stroke	Control	*P* Value†
Subjects/eyes, *N/n*	33/52	27/43	–
Female/male, *N/n*	16/17	13/14	0.979§
Age, years	71.35 (8.28)	65.63 (3.74)	<0.001†,‡
Duration of stroke, years	6.50 (7.27)	–	–
Hypertension, *n*	15	8	0.210§
Diabetes, *n*	10	0	0.005†,§
BCVA, decimal	0.83 (0.15)	0.96 (0.11)	<0.001†,§
SSI	7.02 (0.98)	7.50 (1.26)	0.082‡

SSI, signal strength index.

*Continuous variables are displayed as mean (standard deviation) according to their distributions; categorical data are displayed as numbers.

§χ^2 test.

‡Unpaired Student *t*-test.

†*P* value: ischemic stroke vs. control group statistically significant, $P < 0.05$.

Table 2. Logistic Regression Analysis of Ischemic Stroke and OCTA Parameters

Characteristics	Univariate Model, OR (95% CI)	*P* Value	Multivariate Model, OR[†] (95% CI)	*P* Value
Female/male, *N/n*	0.92 (0.41–2.08)	0.848		NI
Age, years	2.91 (1.60–5.30)	0.001[*]	2.16 (0.91–5.11)	0.081
Hypertension, *n*	1.78 (0.77–4.11)	0.180		NI
Diabetes, *n*	N/A			NI
BCVA	0.31 (0.17–0.56)	<0.001[*]	0.21 (0.08–0.60)	0.003[*]
SSI	0.63 (0.37–1.07)	0.085		NI
SCP				
VAD	0.96 (0.64–1.44)	0.829		NI
VFD	0.95 (0.63–1.42)	0.790		NI
VOD	0.90 (0.59–1.35)	0.595		NI
FA	0.79 (0.52–1.20)	0.269		NI
FAR	2.25 (1.33–3.81)	0.003[*]	1.82 (0.82–4.02)	0.140
FC	0.71 (0.47–1.09)	0.116		NI
FR	0.54 (0.34–0.86)	0.009[*]		NI
DCP				
VAD	0.59 (0.36–0.96)	0.034[*]	0.65 (0.30–1.41)	0.273
VFD	0.61 (0.37–1.00)	0.050		NI
VOD	1.07 (0.70–1.65)	0.749		NI
FA	0.89 (0.58–1.36)	0.584		NI
FAR	2.34 (1.32–4.14)	0.003[*]	2.77 (1.30–5.91)	0.008[*]
FC	0.51 (0.29–0.90)	0.019[*]	0.36 (0.15–0.86)	0.022[*]
FR	1.31 (0.55–3.10)	0.546		NI

CI, confidence interval; FA, FAZ area; FR, FAZ roundness; NI, not included; OR, odds ratio; VFD, vascular fractal dimension.
[*] $P < 0.025$ (adjustment after multitesting), binary logistic regression analysis.
[†] Adjusted for age, BCVA, FAR of SCP, VOD of DCP, FAR of DCP, and FC of DCP.

Study Population of Two Ischemic Stroke Subtypes

The study enrolled 14 patients (24 eyes) with nonlacunar stroke and 19 patients (28 eyes) with lacunar stroke. Baseline characteristics of the study population are shown in Table 3. No significant differences in age, sex, duration of stroke, hypertension, diabetes, hyperlipidemia, BCVA, or signal strength index were found between the two groups.

Table 3. Clinical Characteristics of Patients With Two Subtypes of Ischemic Stroke

Characteristics[*]	Lacunar Infarction	Nonlacunar Infarction	*P* Value
Subjects/eyes, *N/n*	19/28	14/24	–
Female/male, *N/n*	10/9	7/7	0.881[†]
Age, years	72.07 (8.84)	70.50 (7.70)	0.501[‡]
Duration of stroke, years	5.00 (10.00)	3.00 (3.00)	0.068[§]
Hypertension, n	9	6	0.797[†]
Diabetes, n	5	5	0.704[†]
Hyperlipidemia, n	13	10	1.000[†]
BCVA	0.84	0.82	0.563[‡]
SSI	7.00 (1.10)	7.04 (0.88)	0.897[‡]

SSI, signal strength index.
[*] Continuous variables are displayed as mean (standard deviation) according to their distributions; categorical data are displayed as numbers.
[†] χ^2 test.
[‡] Unpaired Student *t* test.
[§] Kruskal–Wallis test.

Table 4. Logistic Regression Analysis of Two Subtypes of Ischemic Stroke and OCTA Parameters

Characteristics	Univariate Model, OR (95% CI)	*P* Value	Multivariate Model, OR† (95% CI)	*P* Value
Female/male, *N*/*n*	1.33 (0.45–3.99)	0.607		NI
Age, years	1.22 (0.70–2.13)	0.493	1.74 (0.64–2.15)	0.602
Duration of stroke, years	1.31 (0.70–2.47)	0.400		NI
Hypertension, n	1.02 (0.34–3.06)	0.966		NI
Diabetes, n	0.47 (0.14–1.52)	0.205		NI
Hyperlipidemia, n	1.21 (0.40-3.65)	0.730		NI
BCVA	0.85 (0.49–1.47)	0.555	0.91 (0.49–1.70)	0.749
SSI	1.04 (0.60–1.80)	0.895		NI
SCP				
VAD	0.88 (0.51–1.53)	0.656		NI
VFD	0.92 (0.53–1.60)	0.771		NI
VOD	0.41 (0.20–0.84)	0.015	0.41 (0.20–0.85)	0.017*
FA	0.71 (0.40–1.26)	0.240		NI
FAR	1.15 (0.66–2.02)	0.617		NI
FC	1.15 (0.66–2.00)	0.618		NI
FR	0.97 (0.56–1.68)	0.907		NI
DCP				NI
VAD	0.72 (0.39–1.34)	0.305		NI
VFD	0.72 (0.38–1.35)	0.302		NI
VOD	1.54 (0.82–2.90)	0.183		NI
FA	0.77 (0.43–1.40)	0.396		NI
FAR	1.29 (0.70–2.37)	0.409		NI
FC	0.53 (0.24–1.14)	0.103		NI
FR	1.75 (0.18–17.34)	0.634		NI

OR, odds ratio; CI, confidence interval; FA, FAZ area; FR, FAZ roundness; NI, not included; SSI, signal strength index; VFD, vascular fractal dimension.

*$P < 0.025$ (adjustment after multitesting), binary logistic regression analysis.

†Adjusted for age, BCVA, and VOD of SCP.

Logistic Regression Analysis of Two Ischemic Stroke Subtypes and OCTA Parameters

Univariate logistic regression analysis (Table 4) revealed lower VOD of SCP (OR, 0.41) were associated with lacunar infarction. Finally, two factors that were clinically meaningful in baseline characteristics (age and BCVA) and one OCTA parameter (VOD of SCP) were included in the multivariate logistic regression model.

The multivariate logistic regression analysis (Table 4) showed that a lower VOD of SCP (OR, 0.41) was independently associated with lacunar infarction. The remaining parameters were not statistically significant.

Discussion

In this study, we assessed retinal microvascular and FAZ changes in patients with ischemic stroke and its subtypes, namely, lacunar and nonlacunar stroke. We found that a worse BCVA, higher FAR of DCP, and lower FC of DCP were associated with ischemic stroke. Moreover, a lower VOD in the SCP was significantly associated with patients with a lacunar infarction than patients with a nonlacunar infarction. Our study adds to the knowledge of the retinal microvasculature and FAZ in ischemic stroke and its subtypes.

We are not aware of any studies focusing on the shape-based metrics of the retinal microvasculature and FAZ in patients with ischemic stroke using OCTA. In terms of differences in FAZ morphology, our study demonstrated that a higher degree of FAZ irregularity (higher FAR and lower FC) was associated with ischemic stroke.

The FAR, introduced by Krawitz et al. (2017),[21] is a parameter used to quantify the irregularity of the FAZ shape. Researchers have revealed that shape may be more appropriate for characterizing the FAZ because FAZ size is highly heterogeneous in normal

populations.[22] Few studies have focused on the alterations of FAR in diseases. Our study showed that an increased FAR was associated with ischemic stroke, suggesting that the FAZ becomes more elongated and irregular. We hypothesized that FAZ deformation could better reflect the pathophysiological alterations in the retinal microvasculature in patients with ischemic stroke.

FC is a sensitive indicator of papillary loss and macular ischemia.[23] When the shape becomes less round or less smooth, the FC is close to zero.[24] Previous studies have shown that a diminished FC was associated with vascular dropout and disease progression in vascular maculopathy.[25,26] According to the neurovascular unit theory of the pathogenesis of stroke, ischemic stroke is associated with functional impairment in the neurovascular unit,[27] which is also present in the retina, and the neurovascular unit is damaged in patients with cerebrovascular diseases.[28] These similar pathological processes may explain the mechanism of more severe FAZ damage (a lower FC) in patients with ischemic stroke.

The FAZ area has been used as a common indicator of microvascular changes in diabetes because of its simplicity and ease of measurement.[23] In addition, some scholars have investigated changes in the FAZ area in neurodegenerative diseases and cerebral small vessel diseases, such as Alzheimer's disease, mild cognitive impairment, and cerebral autosomal-dominant arteriopathy with subcortical infarcts and leukoencephalopathy.[14,29,30] A previous study by Hao et al.[31] found an increased FAZ area in patients with cerebral infarction. Inconsistent with this study, we observed that the FAZ area was not associated with ischemic stroke. It is possible that the two studies included populations with different age and sex ratios, which are factors that have been reported to affect FAZ area.[32,33]

The most interesting finding in this study was that the VOD were higher in patients with nonlacunar infarction than in those with lacunar infarction. We proposed a VOD parameter for the first time to evaluate the direction of retinal blood flow. The mechanisms underlying a higher VOD in nonlacunar stroke are unclear. A previous study identified that the mean flow velocities of cerebral arteries in patients with lacunar infarction were significantly lower than those in patients with nonlacunar infarction. Different hemodynamic alterations might be associated with the pathophysiology of lacunar infarction compared with other subtypes of ischemic stroke.[34] Based on the similarity between the retinal and cerebral vasculature, we hypothesized that the changes in retinal blood flow direction in patients with lacunar infarction were less complex than those in patients with nonlacunar infarction. Our data provide evidence that lacunar stroke is associated with retinal microvascular changes that might reflect pathological processes similar to those in the cerebrovasculature. Conventional hypotheses suggest that lacunar infarction is caused by cerebral small arteriolar abnormality, whereas cortical cerebral infarction, the main type of nonlacunar infarction in our study, is mostly due to cardiac or aortic embolism.[7] However, little differences in risk factors between lacunar and nonlacunar infarcts might not support a distinct arterial pathological process underlying lacunar infarction.[35] Currently, the pathogenesis of both has been unclear. Considering that OCTA can provide surrogate markers of microvascular alteration owing to intercurrent hypertension, endothelial dysfunction, inflammation, arteriosclerosis, and atheroma, it is promising to explore the distinction between different subtypes of ischemic stroke by retinal biopsy.

We noted that only one previous study focused on retinal structural and microvascular changes in two acute ischemic stroke subtypes: large artery atherosclerosis and small vessel occlusion.[4] They found that the superior peripapillary retinal nerve fiber layer thickness in large artery atherosclerosis was significantly different from that in small vessel occlusion. Although the types of ischemic stroke included in their study were not consistent with ours, they all shed light on the use of OCTA to explore retinal microcirculatory alterations in the different subtypes of ischemic stroke.

In terms of FAZ-related parameters, our study showed that the alterations in FAR and FC were more apparent in the DCP than SCP between patients with ischemic stroke and control participants. A comparison of these findings with those of other studies confirmed that the DCP is more sensitive to ischemia and hypoxia. Previous reports have also shown that capillary damage in the deep retina is more obvious than in the SCP in patients with cerebral infarction.[31] In addition, another study revealed that the DCP vessel density in some regions of the deep retina was lower in patients with large artery atherosclerosis than in the controls.[4] There are several possible explanations for this finding. The DCP is located at the junction of the inner and outer plexiform layers, where the oxygen content is low. In addition, the DCP consists of capillaries with fine diameters and is susceptible to harmful factors such as ischemia and hypoxia.[36]

Previous studies have identified retinal vessel changes in ischemic stroke using fundus photography. For example, Seidelmann et al.[37] demonstrated that narrower retinal arterioles and wider retinal

venules were associated with the long-term risk of ischemic stroke. Another study found that a decreased retinal fractal dimension and increased retinal vessel tortuosity were associated with ischemic stroke.[38] These specific parameters based on fundus photography are similarly defined as those from OCTA; however, the association or difference between the retinal parameters found in these two instruments is unclear. Therefore, the integration of parameters from fundus photography and OCTA may help us to better explore retinal vascular changes in ischemic stroke. In fact, preliminary studies from Arnould et al.[39] have led to useful exploration in estimating the cardiovascular risk score based on quantitative retinal vascular parameters from fundus photography and OCTA.

In this study, we applied OCTA-Net, a novel vessel segmentation network to detect thick and thin vessels separately, and Level Sets macro for automated measurements of retinal microvasculature and FAZ in ischemic stroke, respectively. This is a beneficial attempt to explore the pathophysiology of disease with the aid of machine learning methods. Currently, artificial intelligence shows advantages in the direction of OCTA signal generation, OCTA image enhancement, artifact removal, feature segmentation, and diagnosis. For example, artificial intelligence-assisted OCTA image analysis can remove artifacts that are difficult to remove with manual algorithms and can quantify the complex signals generated by many pathological conditions.[12] At the same time, the need to optimize computational techniques and the extraction of more appropriate parameters and the lack of a database with quality annotated data are some challenges or deficits to overcome.[40] In the future, the use of OCTA combined with artificial intelligence may help to understand the pathological mechanisms underlying cerebrovascular ischemic diseases and can be complementary to existing neuroimaging tools.

The advantage of the present study is that, for the first time, OCTA image analysis is combined with machine learning to explore the characteristic changes of the retinal microvasculature and FAZ in ischemic stroke and its different types. Moreover, taking advantage of the ophthalmic enrollment of patients, we excluded ocular diseases that could affect retinal vascular changes, which are easily overlooked in other studies. Nonetheless, this study has various limitations. First, the OCTA 3 × 3 mm high resolution is accurate within a small range, limiting our observation of the peripheral retina in patients with ischemic stroke. Combining multimodel imaging or wide-angle OCTA imaging instruments may be a good choice. Second, considering the asymmetry between the eyes of the same individual, we cannot ignore the statistical problems of including both eyes in the analysis, because of the nonindependence of both eyes.[41] Finally, we detected retinal microvascular changes using OCTA several years after the diagnosis of ischemic stroke. Intercurrent vascular events (hypertension, stroke, endothelial dysfunction and inflammation) may be associated with these abnormal vascular parameters. The cross-sectional design of this study did not account for the temporal linkage of these associations. Future prospective studies with larger sample sizes are needed to determine whether OCTA could provide additional value for exploring the pathological changes of existing ischemic stroke disease.

Conclusions

Overall, our study showed that increased FAR of the DCP and decreased FC of the DCP were associated with ischemic stroke. We also showed that decreased VOD of the SCP is associated with lacunar infarction compared with nonlacunar infarction. These findings indicate that OCTA is a potential and useful tool for studying retinal microvascular changes in different types of ischemic stroke.

Acknowledgments

Supported by the National Natural Science Foundation of China (82171022, 81974128) and The Capital Health Research and Development of Special (2020-2-4097).

Disclosure: **H. Duan**, None; **J. Xie**, None; **Y. Zhou**, None; **H. Zhang**, None; **Y. Liu**, None; **C. Tang**, None; **Y. Zhao**, None; **H. Qi**, None

References

1. CO Johnson, Nguyen M, Roth GA, et al. Global, regional, and national burden of stroke, 1990–2016: A systematic analysis for the Global Burden of Disease Study 2016. *Lancet Neurol*. 2019;18:439–458.
2. Feigin VL, Norrving B, Mensah GA. Global burden of stroke. *CircRes*. 2017;120:439–448.
3. Wardlaw JM, Smith C, Dichgans M. Small vessel disease: Mechanisms and clinical implications. *Lancet Neurol*. 2019;18:684–696.
4. Zhang Y, Shi C, Chen Y, et al. Retinal structural and microvascular alterations in different

acute ischemic stroke subtypes. *J Ophthalmol*. 2020;2020:8850309.
5. Futrell N. Lacunar infarction: Embolism is the key. *Stroke*. 2004;35:1778–1779.
6. Norrving B. Lacunar infarction: Embolism is the key: Against. *Stroke*. 2004;35:1779–1780.
7. Wardlaw JM. What causes lacunar stroke? *J Neurol Neurosurg Psychiatry*. 2005;76:617–619.
8. Cheung CY, Ikram MK, Chen C, Wong TY. Imaging retina to study dementia and stroke. *Prog Retin Eye Res*. 2017;57:89–107.
9. Rim TH, Teo AWJ, Yang HHS, Cheung CY, Wong TY. Retinal vascular signs and cerebrovascular diseases. *J Neuroophthalmol*. 2020;40:44–59.
10. Mohammed MA, Lolah MM, Doheim MF, AbouSamra A. Functional assessment of early retinal changes in diabetic patients without clinical retinopathy using multifocal electroretinogram. *BMC Ophthalmol*. 2020;20:411.
11. Kwan CC, Fawzi AA. Imaging and biomarkers in diabetic macular edema and diabetic retinopathy. *Curr Diabetes Rep*. 2019;19:95.
12. Hormel TT, Hwang TS, Bailey ST, Wilson DJ, Huang DV, Jia YL. Artificial intelligence in OCT angiography. *Prog Retin Eye Res*. 2021;85:100965.
13. Arnould L, Guenancia C, Azemar A, et al. The EYE-MI pilot study: A prospective acute coronary syndrome cohort evaluated with retinal optical coherence tomography angiography. *Invest Ophthalmol Vis Sci*. 2018;59:4299–4306.
14. Zhang JF, Wiseman S, Valdes-Hernandez MC, et al. The application of optical coherence tomography angiography in cerebral small vessel disease, ischemic stroke, and dementia: A systematic review. *Front Neurol*. 2020;11:1009.
15. Zhang X, Xiao H, Liu C, et al. Optical coherence tomography angiography reveals distinct retinal structural and microvascular abnormalities in cerebrovascular disease. *Front Neurosci*. 2020;14:588515.
16. Adams HP, Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischemic stroke - definitions for use in a multicenter clinical-trial. *Stroke*. 1993;24:35–41.
17. Ma Y, Hao H, Xie J, et al. ROSE: A retinal OCT-angiography vessel segmentation dataset and new model. *IEEE Trans Med Imaging*. 2021;40:928–939.
18. Lin A, Fang D, Li C, Cheung C, Chen H. Improved automated foveal avascular zone measurement in cirrus optical coherence tomography angiography using the level sets macro. *Transl Vis Sci Technol*. 2020;9:20.
19. Jerman T, Pernus F, Likar B, Spiclin Z. Enhancement of vascular structures in 3D and 2D angiographic images. *IEEE Trans Med Imaging*. 2016;35:2107–2118.
20. Szpak ZL, Chojnacki W, van den Hengel A. Guaranteed ellipse fitting with the Sampson distance. *12th European Conference on Computer Vision (ECCV)*. Florence, Italy; 2012:87–100.
21. Krawitz BD, Mo S, Geyman LS, et al. Acircularity index and axis ratio of the foveal avascular zone in diabetic eyes and healthy controls measured by optical coherence tomography angiography. *Vis Res*. 2017;139:177–186.
22. Shiihara H, Terasaki H, Sonoda S, et al. Objective evaluation of size and shape of superficial foveal avascular zone in normal subjects by optical coherence tomography angiography. *Sci Rep*. 2018;8:10143.
23. Sun Z, Yang D, Tang Z, Ng DS, Cheung CY. Optical coherence tomography angiography in diabetic retinopathy: An updated review. *Eye (Lond)*. 2021;35:149–161.
24. Olson E. Particle shape factors and their use in image analysis–part 1: Theory. *Journal of GxP Compliance*. 2011;15:85–96.
25. Conrath J, Giorgi R, Raccah D, Ridings B. Foveal avascular zone in diabetic retinopathy: Quantitative vs qualitative assessment. *Eye (London, England)*. 2005;19:322–326.
26. Mo S, Krawitz B, Efstathiadis E, et al. Imaging foveal microvasculature: Optical coherence tomography angiography versus adaptive optics scanning light ophthalmoscope fluorescein angiography. *Invest Ophthalmol Vis Sci*. 2016;57:OCT130–OCT140.
27. Steliga A, Kowianski P, Czuba E, Waskow M, Morys J, Lietzau G. Neurovascular unit as a source of ischemic stroke biomarkers-limitations of experimental studies and perspectives for clinical application. *Transl Stroke Res*. 2020;11:553–579.
28. Peng QS, Hu YJ, Huang MQ, et al. Retinal neurovascular impairment in patients with essential hypertension: An optical coherence tomography angiography study. *Invest Ophthalmol Vis Sci*. 2020;61:42.
29. Bulut M, Kurtulus F, Gozkaya O, et al. Evaluation of optical coherence tomography angiographic findings in Alzheimer's type dementia. *Br J Ophthalmol*. 2018;102:233–237.
30. Nelis P, Kleffner I, Burg MC, et al. OCT-Angiography reveals reduced vessel density in the deep retinal plexus of CADASIL patients. *Sci Rep*. 2018;8:8148.

31. Kwapong WR, Yan Y, Hao Z, Wu B. Reduced superficial capillary density in cerebral infarction is inversely correlated with the NIHSS score. *Front Aging Neurosci*. 2021;13:626334.
32. Yu J, Jiang CH, Wang XL, et al. Macular perfusion in healthy Chinese: An optical coherence tomography angiogram study. *Invest Ophthalmol Vis Sci*. 2015;56:3212–3217.
33. Iafe NA, Phasukkijwatana N, Chen XJ, Sarraf D. Retinal capillary density and foveal avascular zone area are age-dependent: Quantitative analysis using optical coherence tomography angiography. *Invest Ophthalmol Vis Sci*. 2016;57:5780–5787.
34. Kim JT, Lee SH, Hur N, Jeong SK. Blood flow velocities of cerebral arteries in lacunar infarction and other ischemic strokes. *J Neurol Sci*. 2011;308:57–61.
35. Jackson C, Sudlow C. Are lacunar strokes really different? A systematic review of differences in risk factor profiles between lacunar and nonlacunar infarcts. *Stroke*. 2005;36:891–901.
36. Cao Y, Yan J, Zhan Z, Liang Y, Han Z. Macula structure and microvascular changes in recent small subcortical infarct patients. *Front Neurol*. 2020;11:615252.
37. Seidelmann SB, Claggett B, Bravo PE, et al. Retinal vessel calibers in predicting long-term cardiovascular outcomes the Atherosclerosis Risk in Communities study. *Circulation*. 2016;134:1328–1338.
38. Ong YT, De Silva DA, Cheung CY, et al. Microvascular structure and network in the retina of patients with ischemic stroke. *Stroke*. 2013;44:2121–2127.
39. Arnould L, Guenancia C, Bourredjem A, et al. Prediction of cardiovascular parameters with supervised machine learning from Singapore "I" vessel assessment and OCT-angiography: A pilot study. *Transl Vis Sci Technol*. 2021;10:20.
40. Sarmento RM, Vasconcelos FFX, Filho PPR, Wu W, de Albuquerque VHC. Automatic neuroimage processing and analysis in stroke—a systematic review. *IEEE Rev Biomed Eng*. 2020;13:130–155.
41. Cameron JR, Megaw RD, Tatham AJ, et al. Lateral thinking - interocular symmetry and asymmetry in neurovascular patterning, in health and disease. *Prog Retinal Eye Res*. 2017;59:131–157.