Depth-resolved vascular profile features for artery-vein classification in OCT and OCT angiography of human retina

Tobiloba Adejumo\textsuperscript{a,c}, Tae-Hoon Kim\textsuperscript{a,c}, David Le\textsuperscript{b}, Taeyoon Son\textsuperscript{b}, Guangying Ma\textsuperscript{b}, Xincheng Yao\textsuperscript{a,b,*}

\textsuperscript{a}Department of Bioengineering, University of Illinois at Chicago, Chicago, IL 60607; \textsuperscript{b}Department of Ophthalmology and Visual Sciences, University of Illinois at Chicago, Chicago, IL 60612; \textsuperscript{c}These authors contributed equally to this work; \*xcy@uic.edu Tel: 1 312 413-2016; Fax: 1 312 996-4644

ABSTRACT

The purpose of this study is to use optical coherence tomography (OCT) to characterize the reflectance profiles of retinal blood vessels and to use these features for artery-vein classification in OCT angiography (OCTA). The retinal arteries and veins show unique features in the depth-resolved OCT. Both the upper and lower side of the retinal arteries have hyper-reflective boundaries. However, retinal veins reveal only hyper-reflective boundary at the upper side. In both small and large arteries, relatively uniform lumen intensity was observed. On the other hand, the vein lumen intensity was dependent on the vessel size; the bottom half of the lumen of small veins show a hyper-reflective zone while the bottom half of the lumen of big veins a hypo-reflective zone.

Keywords: Retina; Artery-vein classification; Optical coherence tomography; Optical coherence tomography angiography;

1. INTRODUCTION

The retinal neurovascular system is commonly targeted by systemic and retinal diseases. Reduced retinal blood flow velocities in the short posterior ciliary artery might indicate the presence of glaucoma [1]. Diabetic retinopathy (DR) is characterized by retinal artery narrowing [2] and venous beading [3, 4]. Furthermore, Alzheimer’s patients are hallmark by the presence of retinal venous narrowing and blood flow decrease [5], while retinal venous abnormalities is commonly associated with Parkinson’s patients [6]. Differential artery-vein (AV) analysis offers enhanced sensitivity for disease detection, prediction and staging since different diseases impact the retinal arteries and veins in different ways. A prerequisite for differential AV analysis is AV classification. OCT angiography (OCTA) is a novel optical coherence tomography (OCT) technique that allows for noninvasive label-free imaging of the retinal vasculature with capillary level resolution. OCTA has been widely used in laboratory research and clinical care for a variety of diseases [7]. However, clinical OCTA does not inherently allow AV classification. The purpose of this study is to show that OCT axial reflectance profiles or cross-sectional imaging features of blood vessels, may be used to distinguish arteries and veins in OCT and OCTA of the human retina.

2. METHODS

2.1 Data acquisition

This study was approved by the Institutional Review Board of the University of Illinois at Chicago and is in pursuance with the ethical standards stated in the Declaration of Helsinki. Five healthy young subjects with no history of ocular or systemic diseases, ranging in age from 24 to 39 years, were recruited from the Lions of Illinois Eye Research Institute of the University of Illinois. The mean refractive error of subjects was found to be \(-2 \pm 0.86\) diopters (range: \(-3.25\) to \(-1.75\) diopters) using the ARK-900 autorefractor system (Nidek, San Jose, CA, USA). Informed consent was provided by each subject before participation in the study. AngioVue SD-OCT angiography system (Optovue, Fremont, CA, USA) with a 70-KHz A-scan rate was used to acquire OCT and OCTA images. The axial and lateral resolution was 5 \(\mu\)m and 15 \(\mu\)m, respectively. OCT/OCTA imaging included a 6 \(\times\) 6-mm field of view area centered on the fovea, and ONH with 400 A-scans \(\times\) 400 B-scans. For OCTA volume reconstruction, split-spectrum amplitude-decorrelation angiography (SSADA) algorithm was used, and the repeated 2 B-scans were averaged for OCT volume construction. OCT/OCTA volumetric images were exported from Optovue graphical user interface (GUI) and further reconstructed using custom-developed...
MATLAB R2021a (Mathworks, Natick, MA, USA) functions. Color fundus images were captured using Pictor Plus (Volk Optical, Mentor, OH, USA), a nonmydriatic retinal camera with a field-of-view (FOV) up to 45 degrees, with frame resolution of 1536 x 1152 pixels. The color-fundus images were exported from Pictor Plus and used to verify the AV classification in OCT/OCTA. In Figure 1B, the OCT volumes were resliced by tracing individual blood vessels to obtain the cross-sectional vessel images. To generate a single representative vessel image with enhanced contrast, several resliced B-scans at the center of the blood vessel were averaged as illustrated in Figure 1C and Figure 1D. The cross-sectional vessel image in Figure 1D was further flattened, followed by a vertical rescaling for quantitative analysis.

![Figure 1](image)

**Figure 1.** Cross-sectional vessel image preparation. (A) Representative en face OCT. (B) Illustration of the vessel tracing process from the ONH to obtain a cross-sectional vessel image. The image is an enlarged view of the gray box in (A). The green line in (B) illustrates the maximum width from the center of the vessel used to obtain the B-scans in (C). (C) Stack of B-scans resliced from the vessel tracking in (B). AIP: average intensity projection. (D) Representative cross-sectional vessel image. Scale bars: 500 μm.

3. **RESULTS**

3.1 Establishing ground truth for AV analysis

For quantitative profile feature analysis, the ground truth information was created to distinguish cross-sectional vessel images into artery and vein. Color fundus photography and en face OCT/OCTA were used to determine ground truth of arteries and veins by manually examining established criteria for AV classification in retinal images (Figure 2) [8, 9]. In Figure 2A, the light reflex of the interior parts of the vessels was apparent in arteries. Due to the difference in oxygen level in arteries and veins, arteries also revealed a brighter color than veins (Figure 2B). Furthermore, artery and vein crossing made a roughly 90-degree angle. We also established that arteries did not cross arteries, and veins did not cross veins, while arteries crossed veins and vice versa (Figure 2C).
Figure 2. Establishing ground truths for AV classification. (A) Representative fundus image. (B) Enlarged illustration of the white window region in (A). The white circle showing nearly 90-degree angle crossing between AV. In addition, the artery is brighter than the vein. (C) Representative en face OCTA image. White circles show AV cross each other. The image is an enlarged view of the OCTA inset in (A). A: artery; V: vein.

3.1 Reflectance profiles and blow flow patterns in retinal arteries and veins

The vessel walls of arteries reveal continuous hyper-reflective boundaries. As illustrated in Figure 3A, the hyper-reflective boundary intensity gradually decreases in subsequent branches but remain distinct. Normalized vascular intensity maps were created from the individual branch vessels of all subjects. In the arterial profile, a sharp increase in lower vessel boundary intensity was consistently observed. However, in the veins, a continuous reduction in lower vessel boundary intensity with some pixels before the lower boundary was observed, as illustrated in Figure 3C and 3D. The AV intensity plots in Figure 3E and 3F show relatively homogenous intensity distribution in arteries, hypo-reflectivity in the bottom half of the venous lumen in large veins, and hyper-reflectivity in the bottom half of the venous lumen in small veins.
3.1 AV classification in OCT and OCTA images

The source nodes for each of the vessels at the ONH region was identified and the corresponding reflectance profile was obtained as shown in Figure 4A. Blood vessel tracking algorithm from the source nodes, which employs curvature angle information was used to classify the entirety of the OCT vasculature into artery or vein [10]. To generate the OCTA vessel map, the outer retina was removed to clean small capillary mesh that could not be resolved as artery or vein. The en face OCT and OCTA images were created by the three-dimensional projection of OCT B-scans and OCTA B-scans. Because the OCTA is naturally based on the OCT speckle variance processing to improve the visibility of blood vessels, image registration is not required during superimposition. However, the OCTA vessel map shows additional vascular details compared to OCT vessel map. Based on the relationship between the OCT and OCTA, the OCT can be directly used to guide the AV classification and vessel tracking in OCTA.
4. DISCUSSION

In summary, we demonstrated the feasibility of using OCT reflectance profiles in retinal blood vessels to distinguish arteries and veins and validated the cross-sectional OCT profile analysis guided AV classification in OCTA. Cross-sectional OCT vessel images were obtained from the ONH and macular regions. Individual vessel images were first classified into artery and vein based on the established imaging features that appeared on fundus photograph and en face OCT/OCTA images. In arteries, normalized reflectance profiles revealed hyper-reflective wall boundaries, whereas veins had a layered intensity (Figure 3).

In the arterial walls, hyper-reflective boundaries were apparent. The distinct components of blood vessel walls have been characterized in previous histologic research. Endothelial cells and mural cells are the two types of cells that make up the walls of blood vessels. Endothelial cells cover the inner wall lining. Mural cells are used to line the outer walls. In small vessels, this is referred to as pericytes, whereas in larger vessels, it is referred to as vascular smooth muscle cells (SMCs) [11].

The well-developed tunica media of the retinal arteries, which is bordered by layers of mural cells, distinguishes them from other organ arteries. As arteries branch into arterioles, the number of SMCs abates [12]. In contrast, the venous wall is thin, with only a single layer of endothelial cells and few SMCs [13]. Hyper-reflective boundary signals in arterial radial B-scans were also seen in previous studies of rat retinal vessels [14]. Thus, the arterial wall morphology would be responsible for the hyper-reflective boundary signals in arteries.

In addition, three unique luminal intensity distributions were observed in different vessel sizes. A distinct hyper-reflective zone was observed in the bottom half of the lumen of small veins. This hyper-reflective zone might be due to venous laminar flow [15]. Because of the laminar flow features, axial mixing of blood cells leaving the deep capillary plexus (DCP) would be limited; as a result, most erythrocytes would follow the lumen's basal blood stream. However, when multiple branches of small venules merge to form a large venous branch, the laminar flow stream can be potentially disrupted at the merging point. In these larger veins, a hypo-reflective zone was observed in the bottom half of the lumen. We speculate that the large blood volume may be a primary cause of OCT signal attenuation. As a result, bottom half of
the lumen may appear dark due to light attenuation (Figure 3). In small and large arteries, we consistently observed relative homogeneous intensity distribution with a hypo-reflective zone at the center of the lumen. The observed hypo-reflective zone was mainly caused by shear-induced erythrocytes orientation in concert with the vessel boundaries which causes low backscattering of light [16].

In conclusion, we demonstrated the feasibility of using OCT reflectance profiles in retinal blood vessels to differentiate arteries and veins and validated the cross-sectional OCT profile analysis guided AV classification in OCTA.

ACKNOWLEDGEMENTS

This research was supported in part by National Institutes of Health (NIH) (R01 EY023522, R01 EY029673, R01 EY030101, R01 EY030842, P30 EY001792); Richard and Loan Hill endowment; unrestricted grant from Research to Prevent Blindness.

REFERENCES

[1] O. Zeitz, P. Galambos, L. Wagenfeld et al., “Glaucoma progression is associated with decreased blood flow velocities in the short posterior ciliary artery,” British Journal of Ophthalmology, 90(10), 1245-1248 (2006).
[2] L. Pedersen, P. Jeppesen, S. T. Knudsen et al., “Improvement of mild retinopathy in type 2 diabetic patients correlates with narrowing of retinal arterioles. A prospective observational study,” Graefe's Archive for Clinical and Experimental Ophthalmology, 252(10), 1561-1567 (2014).
[3] R. A. Fonseca, and M. A. Dantas, “Retinal venous beading associated with recurrent branch vein occlusion,” Canadian journal of ophthalmology, 37(3), 182-183 (2002).
[4] B. Piguet, M. Gross-Jendroska, F. G. Holz et al., “Inherited venous beading,” Eye, 8(1), 84-88 (1994).
[5] G. T. Feke, B. T. Hyman, R. A. Stern et al., “Retinal blood flow in mild cognitive impairment and Alzheimer's disease,” Alzheimer's & Dementia: Diagnosis, Assessment & Disease Monitoring, 1(2), 144-151 (2015).
[6] R. Kromer, C. Buhmann, U. Hidding et al., “Evaluation of retinal vessel morphology in patients with Parkinson's disease using optical coherence tomography,” PLoS One, 11(8), e0161136 (2016).
[7] N. K. Waheed, T. De Carlo, A. Chin et al., “OCT angiography in retinal diagnosis and treatment,” Retinal Phys, 12, 26-42 (2015).
[8] C. Kondermann, D. Kondermann, and M. Yan, "Blood vessel classification into arteries and veins in retinal images." 6512, 651247.
[9] J. Motte, F. Alten, C. Ewering et al., “Vessel labeling in combined confocal scanning laser ophthalmoscopy and optical coherence tomography images: criteria for blood vessel discrimination,” PLoS One, 9(9), e102034 (2014).
[10] M. Alam, T. Son, D. Toslak et al., “Combining ODR and blood vessel tracking for artery–vein classification and analysis in color fundus images,” Translational vision science & technology, 7(2), 23-23 (2018).
[11] T. Y. Chui, T. J. Gast, and S. A. Burns, “Imaging of vascular wall fine structure in the human retina using adaptive optics scanning laser ophthalmoscopy,” Investigative ophthalmology & visual science, 54(10), 7115-7124 (2013).
[12] M. J. Hogan, and L. Feeney, “The ultrastructure of the retinal blood vessels: I. The large vessels,” Journal of ultrastructure research, 9(1-2), 10-28 (1963).
[13] D.-Y. Yu, E.-N. Su, S. J. Cringle et al., “Local modulation of retinal vein tone,” Investigative ophthalmology & visual science, 57(2), 412-419 (2016).
[14] T.-H. Kim, D. Le, T. Son et al., “Vascular morphology and blood flow signatures for differential artery-vein analysis in optical coherence tomography of the retina,” Biomedical Optics Express, 12(1), 367-379 (2021).
[15] E. Helps, and D. McDonald, “Observations on laminar flow in veins,” The Journal of physiology, 124(3), 631 (1954).
[16] P. Cimalla, J. Walther, M. Mittasch et al., “Shear flow-induced optical inhomogeneity of blood assessed in vivo and in vitro by spectral domain optical coherence tomography in the 1.3 μm wavelength range,” Journal of biomedical optics, 16(11), 116020 (2011).