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Ultra-widefield Imaging of the Peripheral Retinal Vasculature in Normal Subjects

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Purpose: To establish the extent of the peripheral retinal vasculature in normal eyes using ultra-widefield (UWF) fluorescein angiography.

Design: Prospective, observational study.

Participants: Fifty-nine eyes of 31 normal subjects, stratified by age, with no evidence of ocular disease in either eye by history and ophthalmoscopic examination.

Methods: Ultra-widefield fluorescein angiographic images were captured centrally and with peripheral steering using the Optos 200Tx (Optos, Dunfermline, United Kingdom). Images obtained at different gaze angles were montaged and corrected for peripheral distortion using a stereographic projection method to provide a single image for grading of the peripheral edge of the visible vasculature. The border of the vascularized retina was expressed as a radial surface distance from the center of the optic disc. The vascularized area was calculated based on this mean peripheral border position for each quadrant.

Main Outcome Measures: Mean distance (mm) from the center of optic disc to the peripheral vascular border.

Results: In normal eyes, the mean radial surface distance from the center of the optic disc to the peripheral edge of the visible vasculature was 20.3±1.4 mm and the mean area of normal perfused retina was 977.0 mm^2. There was no significant difference between right and left eyes or between male and female participants. However, the distance to the periphery differed depending on the quadrant, with temporal (22.5±0.9 mm) being larger than inferior (20.4±1.7 mm) being larger than superior (19.2±1.5 mm) being larger than nasal (17.4±0.9 mm; $P < 0.001$) for all interquadrant comparisons. Interestingly, the distances to the perfused vascular border were significantly shorter in older individuals (≥60 years) than in younger subjects.

Conclusions: Ultra-widefield fluorescein angiography is an important tool for studying the extent of peripheral retinal vasculature. With the increasing use of UWF imaging to evaluate and manage patients with retinal vascular disease, the normative data from this study may provide a useful reference when assessing the pathologic significance of findings in the setting of disease. *Ophthalmology 2016;■:1–7 © 2016 by the American Academy of Ophthalmology.*

The introduction of ultra-widefield (UWF) imaging systems has had a significant impact on the diagnosis and management of various retinal disorders. The UWF imaging provides up to a 200° view of the retina in a single capture and allows detection of peripheral pathologic features that can be missed on the 7 standard fields of the Early Treatment Diabetic Retinopathy Study.[1–6] Identification of peripheral retinal nonperfusion is thought to be of importance for the management of eyes with retinal vascular diseases such as retinal vein occlusion and diabetic retinopathy.[6–11] Several studies have demonstrated that the extent of retinal nonperfusion was associated with the severity of macular edema and with its resolution after treatment.[7–9] These studies also suggest that retinal nonperfusion is related to the upregulation of vascular endothelial growth factor caused by hypoxia.[8,9]

The single, centered image used by previous investigators captures a maximum of 80% of the fundus, but includes an inevitable nonlinear distortion caused by the elliptical mirror used in the Optos UWF system (Optos, Dunfermline, United Kingdom).[1–11] In addition, these previous studies were unable to determine the precise size of the nonperfused area, because there was no solution at that time to resolve the inherent peripheral distortion present in large-field fundus images. They quantified the severity or extent of nonperfusion by expressing the number of pixels within an area of nonperfusion as a percentage of the number of pixels seen within the total visible retina (ischemic index).[7–11] However, the total visible retina also can include the physiologic peripheral nonperfused area near the ora serrata just beyond the normal vascular terminus. Ideally, to depict the extent of pathologic nonperfusion most accurately, this physiologic nonperfusion area should be excluded from the total visible retina when computing the ischemic index. This is particularly important because the total visible retina may vary from case to case depending on limitations of image acquisition.

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An understanding of the normal peripheral vasculature is particularly important, given the recent increase in multicenter, randomized clinical trials such as Diabetic Retinopathy Clinical Research Network protocol AA (designed to assess the impact of peripheral lesions on diabetic retinopathy severity and progression) and the Study of Comparative Treatments for Retinal Vein Occlusion 2, which aims to evaluate the impact of treatment on peripheral nonperfusion.

Recent advances in UWF imaging hardware and software have made accurate quantification of the normal perfused retina now possible. Stereographic projection software, now available in commercial UWF devices, allows images obtained at different gaze angles to be montaged and corrected for peripheral distortion. Using this software, researchers can calculate the anatomically correct areas of nonperfusion in metric units using spherical trigonometry, rather than expressing the area as a percentage.[12] Our previous study showed that this quantification methodology can generate accurate retinal measurements in a human eye using a retinal prosthesis as an in vivo reference standard.[13]

We sought to develop a database of the extent of the peripheral retinal vasculature in actual anatomic units (millimeters) in normal eyes using UWF fluorescein angiography (FA). We also evaluated the change of the extent of perfused retina, with stratification controlled for age.

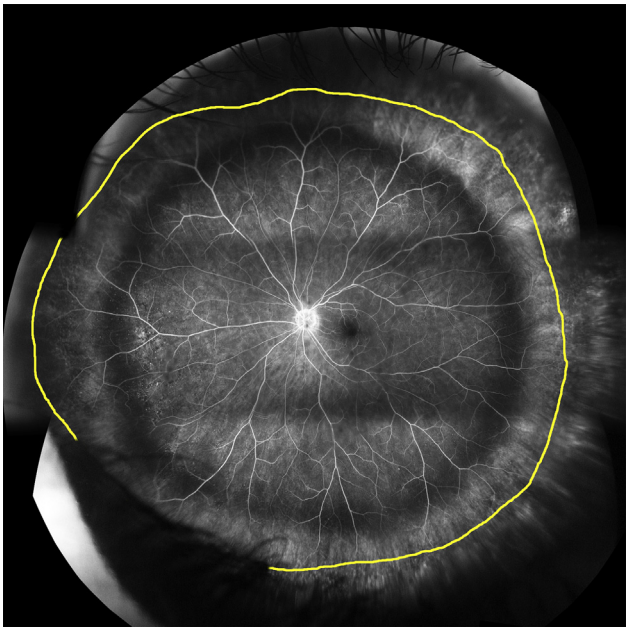

Figure 1. Ultra-widefield fluorescein angiogram image after stereographic projection and montage of central, superior-steered, and inferior-steered images. The extent of peripheral retinal vasculature has been defined by the grader by demarcating (yellow line) the peripheral extent of the blood vessel arborization (the junction between the vascularized and nonvascularized retina).

Methods

Study Population

This prospective observational study was conducted at Medical Center Ophthalmology Associates, San Antonio, Texas, and was approved by its institutional review board. This study adhered to the tenets of the Declaration of Helsinki. Written informed consent was obtained from all subjects before imaging.

The normal subjects were recruited for standardized UWF FA using an Optos 200Tx device with age stratification by decade. All subjects underwent detailed clinical examinations including autorefraction, visual acuity, intraocular pressure measurement, slit-lamp examination, ophthalmoscopy, and optical coherence tomography. Subjects were eligible for inclusion if they were older than 20 years with no known retinal or systemic diseases. The main exclusion criteria included the following: age younger than 20 or older than 80 years; contraindication to dilation; presence of retinal or optic nerve disease, including glaucoma; past history of vitreoretinal surgery; any ocular condition that would interfere with good-quality image acquisition, such as corneal opacities, cataract, or dense vitreous hemorrhage; any medical condition that might interfere with the subject's compliance with study procedures, such as ataxia or nystagmus, that could affect the subject's ability to maintain steady head or eye positioning; a history of diabetes, high blood pressure, or vascular diseases (cardiovascular, peripheral vascular, or cerebrovascular); or the use of vasodilators. Pregnant women or those who might be pregnant also were excluded from participation.

Widefield Image Acquisition and Quantification

Subject eyes were dilated with tropicamide 1% and phenylephrine 2.5%, and UWF pseudocolor images were captured, centered on the fovea, and steered peripherally (nasally, temporally, superiorly, inferiorly). After intravenous administration of fluorescein dye, UWF FA images were obtained during the early (45 seconds), middle (2 minutes and 30 seconds), and late (5 minutes) phases of the angiography. At the 3 time points, in addition to a central image centered on the macula, the FA images were steered superiorly, inferiorly, temporally, and nasally to allow clear visualization of the peripheral edge of the visible vasculature.

Uncorrected raw images were exported from the device and sent to the Doheny Image Reading Center, Doheny Eye Institute, Los Angeles, California. All images for each subject were transformed to stereographic projection images using proprietary prototype software available from the manufacturer. This software is now available in the commercial device or product. This projection technique was accomplished by ray tracing every pixel through a combined optical model of the Optos 200Tx and a Navarro UWF model eye with an axial length of 24 mm.[14] This optical model represented the projection used by the Optos 200Tx scanning laser ophthalmoscopy platform to create the 2-dimensional optomap. The software also allowed the grader to register the 4 steered images to the on-axis image automatically to create a montage of all images (by adding each image one by one). Image registration between a pair of images first extracted their vasculature and subsequently applied rotational affine translation with cross-correlation (i.e., an algorithm slightly rotated the peripheral images to align vasculature). Finally, segments were blended together to create a contiguous montage. Because angiographic images were obtained at 3 time points (early, middle, and late), a separate montage was created for each time point. All montage images then were graded independently by a trained reading center–certified ophthalmologist (M.S.) who was masked to the patient's clinical data, including age and gender. Using ImageJ version 1.49b (ImageJ version 1.49b; US National Institutes of Health, Bethesda, MD) the graders manually outlined the peripheral extent of the blood vessel arborization (the junction between the vascularized and nonvascularized retina; Fig 1). Because the small-vessel detail is expected to be best at the early time point, the graders generally chose the early montage for assessments, zoomed in to the peripheral retina, and panned for 360°