OPEN

Imaging the Choroid: From Indocyanine Green Angiography to Optical Coherence Tomography Angiography

Alessandro Invernizzi, MD†, Marco Pellegrini, MD*, Elisa Cornish, PhD, FRANZCO†, Kelvin Yi Chong Teo, MMED(OPHTH)†‡§, Matteo Cereda, MD*, and Jay Chabblani, MD¶*

Abstract: The choroid is the vascular structure nourishing the retinal pigment epithelium and the outer retina and it plays a key role in the homeostasis of the eye both under physiological and pathological conditions. In the last 20 years we have moved from "guessing" what was happening beyond the retinal pigment epithelium to actually visualize structural and functional changes of the choroid in vivo noninvasively. In this review we describe the state of the art of choroidal imaging, focusing on the multiple techniques available in the clinical and research setting including indocyanine green angiography, labeled-cells angiographies, optical coherence tomography (OCT), enhanced depth imaging, swept source OCT, and OCT angiography. In the first section of the article, we describe their main applications and the basic principles to interpret the imaging results. Increasing evidence suggests that the choroid is much more involved than we used to think in many pathological conditions from uveitis to intraocular tumors, from vascular diseases to age-related macular degeneration. All clinicians should hence know which is the most appropriate imaging investigation to explore the choroid in the disease they are dealing with and how to interpret the results. For this reason the second section of this review summarizes the best imaging approach and the most common findings visible on choroidal imaging in different diseases of the eye.

Key Words: angiography, choroid, imaging, indocyanine green, optical coherence tomography

(*Asia Pac J Ophthalmol (Phila)* 2020;9:335–348)

The choroid is the vascular structure nourishing the retinal pigment epithelium (RPE) and the outer retina. It plays a key role in the homeostasis of the eye both under physiological and pathological conditions since supplies delivery to the retina and immune response regulation depend on choroidal proper functioning.[1] As a result, investigating choroidal structural and functional variations has always been of great interest for clinicians and researchers.

A resolution close to that of histology is needed to appreciate choroidal fine structures and functional changes. At the same time, the choroid is located between the Bruch membrane (BM) and the sclera; thus, one needs to cross several media and anatomical layers (eg, the retina, the RPE, and so on) to visualize it. This particularly unfortunate combination of factors has limited our ability to investigate the choroid for decades.[2]

In the last 10 years, a series of advances in technology has impressively improved the imaging techniques to assess the posterior segment of the eye. Nowadays new imaging modalities and improved traditional methods allow a good visualization of the choroid in vivo, enhancing the insights on the pathophysiology of many ocular conditions and changing our management of several ophthalmic diseases.[2,3]

In this review, we describe the state of the art of choroidal imaging, focusing on the multiple techniques available in the clinical and research setting, their main applications, and the basic principles to interpret the imaging results. In addition, we summarize the best imaging approach and the common most findings visible on choroidal imaging in different diseases of the eye.

CHOROIDAL ANATOMY AND PHYSIOLOGY

The choroid is a highly pigmented, vascular, loose connective tissue found between the sclera and the retina.[1] It extends from the optic nerve head margins to the ciliary body and is thickest at the posterior pole.[4] Together with the ciliary body and the iris, the choroid forms the uveal tract.

Histologically, the choroid is generally divided into 5 layers. The BM (lamina vitrea), the choriocapillaris layer, Sattler's layer of medium sized vessels, Haller's layer of large vessels and the suprachoroid, from inside out.[5] The choriocapillaris is a highly anastomosed network of capillaries adjacent to BM and is about 10 μm thick at the macula. These capillaries arise from the arterioles of Sattler's layer. The suprachoroid is a transitional zone between choroid and the sclera and contains large lacunae which empty into veins (Fine and Yanoff).[5]

BM is a fibroelastic layer found between the fenestrated endothelium of the choriocapillaris and the basal lamina of the RPE. It provides a tensile strength to the choroid but is also an important component of the blood retinal barrier with its charge-selective restrictions to the passage of ions and solutes.[6]

From the *Eye Clinic, Department of Biomedical and Clinical Science "Luigi Sacco," Luigi Sacco Hospital, University of Milan, Milan, Italy; †The University of Sydney, Save Sight Institute, Discipline of Ophthalmology, Sydney Medical School, Sydney, New South Wales, Australia; ‡Singapore National Eye Center, Singapore; §Singapore Eye Research Institute, Singapore; and ¶Department of Ophthalmology, University of Pittsburgh Medical Center, Pittsburgh, PA, USA.

Submitted May 2, 2020; accepted June 4, 2020.

The authors report no conflicts interest.

Correspondence: Alessandro Invernizzi, MD, Eye Clinic, Department of Biomedical and Clinical Science, L. Sacco Hospital University of Milan, via G.B. Grassi 74, 20157 Milano, Italy. E-mail: alessandro.invernizzi@gmail.com.

ISSN: 2162-0989

DOI: 10.1097/APO.0000000000000307

Branches of the ophthalmic artery supply the uvea which does not pass through the optic nerve in contrast to the retinal artery. The choroid is innervated by long and short ciliary nerves from the nasociliary branch of the Ophthalmic nerve (V1), with the sympathetic fibers to the dilator pupillae also passing through. A perivascular plexus made of both divisions of the autonomic nervous system has terminals throughout the stroma, terminating on nonvascular smooth muscle, intrinsic choroidal neurons of the suprachoroid and choroid but do not extend into the choriocapillaris. There are also primary afferent sensory fibers that project to the trigeminal ganglion; their immunohistochemical characteristics suggest that they play a role in mechanosensory changes of the choroid.[7]

Most of the retina's oxygen is consumed by the photoreceptors and is provided by the choroid. To obtain a high transport of oxygen a steep gradient of oxygen tension is required, which is maintained by the high blood flow in the choroid, the highest of any tissue in the body per unit tissue weight.[6] The choroid does have other roles, in particular in thermoregulation of the retina via heat dissipation and modulation of intraocular pressure via vasomotor control of blood flow.

LABELLED IMAGING TECHNIQUES

Indocyanine Green Angiography

Indocyanine green (ICG) angiography was first described in ophthalmology to explore the choroidal circulation in 1973,[8] but it became widely used in the early 1990s.[9] Today, coupled with newer imaging technology, ICG is an established technique for the identification of many retinal and choroidal diseases.

Indocyanine green is a water-soluble tricarbocyanine dye with a molecular weight of 775. Two properties of ICG are important in the clinical utility of visualizing the choroidal circulation. First, in serum, ICG absorbs at 790 to 805 nm and has a peak emission 835 nm. These spectral properties are ideal as they can penetrate the RPE, macular xanthophyll, and other ocular pigments.[10] Second, ICG is a highly protein-bound (98%) dye which has limited diffusion through the small fenestrations of the choriocapillaris.[11]

Nowadays ICG angiography (ICGA) is performed using scanning laser ophthalmoscopes that can produce high-definition images.[12] Current ultrawide field systems also have ICGA filters incorporated and provide ICGA images of up to 200 degree field of view. (Fig. 1)

For a standard ICGA, 25 mg of ICG dye diluted to the manufacturers recommendation is administered intravenously as a bolus. The procedure is generally well tolerated with minimal side effects.[13] Severe reactions such as hypotensive shock and anaphylactic shock are very rare but have been reported.[14] ICG is contraindicated in patients with iodine or shellfish allergies, liver disease, and end-stage renal disease. The dye is rapidly eliminated by the liver and there is minimal uptake in peripheral tissues.

ICGA can be divided into early, middle, and late phases. In the early phase (0 to ~1 minutes), both medium and large choroidal vessels are well visualized beneath the hyper-fluorescent retinal vasculature. In the middle phase (6–15 minutes), the choroidal veins become less distinct with diffused choroidal hyper-fluorescence. Lesions that demonstrate abnormal hyperfluorescence on ICGA are best detected here. In the late phases (beyond 15–30 minutes), all details of normal retinal and choroidal vessels are lost. The choroidal vessels are seen as hypofluorescent channels, retinal vessels are no longer visible, and the optic nerve head is dark.[13]

Labeled Cells and Choroidal Blood Flow

Conventional angiographies which bind fluorescent dyes to plasma proteins do not provide detailed blood flow dynamics of blood cells. Several experimental techniques have been used to study these phenomenas more closely in the retina and choroid.[15] Much of this work is restricted to animal models and not performed in human.

Common technique involves the use of ICG and sodium fluorescein to label erythrocytes and leukocytes. Most flow dynamic studies provide information regarding the characteristic and velocity of blood flow within the retina circulation with few describing the choroidal blood flow.[16]

Matusda et al and Takasu et al described the blood flow dynamics in the choroid with ICG-labeled leucocytes. They found that blood flow in the choriocapillaris was significantly slower than posterior choroidal blood flow which was also found to be faster than retina flow. The movement of the labeled cells also seemed to reflect the anatomy of choroidal vasculature where feeding arteries and draining veins were at right angles to the choriocapillaris.[17,18]

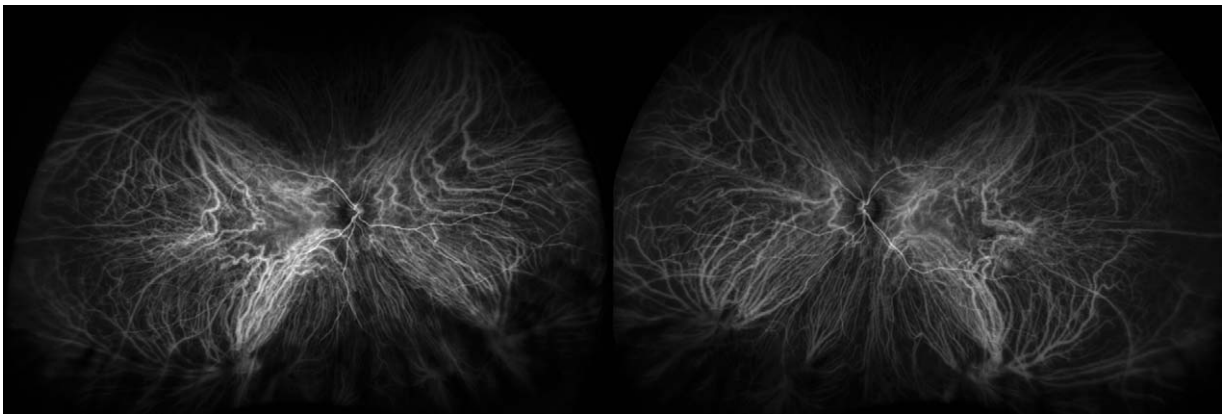

FIGURE 1. Bilateral wide field indocyanine green angiography. Current ultrawide field systems provide indocyanine green angiography images of up to 200 degree field of view.

NONINVASIVE IMAGING TECHNIQUES

Ophthalmic Ultrasound

Ophthalmic ultrasonography (USG) is a noninvasive diagnostic technique providing real-time qualitative and quantitative assessment of the eye and the orbit and where images are obtained by collecting the acoustic signal reflected from different tissues.

Since its introduction in ophthalmology in 1956, USG has been widely used for the study and documentation of several ocular conditions.[19] In the last few years, the availability of probes characterized by higher frequency such as the 20 MHz has further improved USG technique, allowing for a higher resolution if compared with the previous 10-MHz transducers and providing sufficient penetration for a detailed assessment of the retina and deeper tissues.

Ocular USG is routinely performed using different techniques including A-mode, B-mode, and ultrasound biomicroscopy and has to be considered a dynamic exam. In detail, ultrasound biomicroscopy relies on shorter wavelengths (ranging from 50 to 100 MHz) and provides better resolution of the anterior structures of the eye with poor depth of penetration and no possibility to visualize posterior anatomical structures. In the study of the retina and, in particular, the choroid, standardized A-scan and B-scan are often used simultaneously offering complementary information. Standardized A-scan offers a linear representation of the echo amplitude and offers the possibility to perform a precise echostructure assessment and quantification; its use is currently mainly limited to the assessment of intraocular masses.[20,21] Differently, B-scan mode offers a 2-dimensional cross-sectional image of the globe and an immediate view of the pathology; the exam is typically conducted using a systematic scheme throughout radial and longitudinal scans driven by the orientation of the transducer tip and may be performed directly on the anesthetized cornea or the eyelids using coupling medium like methylcellulose. Measurements in B-scan are also possible, but it should be kept in mind that choroidal cleavage from the retina and the sclera may not be possible in this modality, thus leading to some inaccuracy; for this reason, previous reports have documented higher feasibility of enhanced depth imaging optical coherence tomography (EDI-OCT) in documenting choroidal thickness (CT)[22] and for the follow-up of small lesions.[23]

Doppler USG may be performed as a further tool to study blood flow within normal or pathological tissues but, so far, its application is limited.

Currently, USG still plays a mandatory role in the evaluation of the globe and posterior structures in case of ocular traumas, in opaque media such as corneal leukomas, dense cataracts, and vitreous opacities, in the assessment of advanced diabetic retinopathy (DR) or retinal detachments and, particularly, in the diagnosis and follow-up of intraocular tumors.

Optical Coherence Tomography, Enhanced Depth Imaging, and Swept Source

Since its introduction in research settings and even more in the clinical practice, optical coherence tomography (OCT) has revolutionized the way we understand the physiology of the eye and how we manage ocular diseases. OCT is able to collect, in a noninvasive way, images with a resolution close to that of histology in vivo that provide clinicians and researchers with an enormous amount of information.[24]

The most common OCT technology available at the moment is the spectral domain OCT (SD-OCT). This technique investigates the target tissue with an exploring beam of light, usually in the spectrum of the near infrared. The signal returning to the device is analyzed using the Fourier equation and bi-dimensional (b-scan) or 3-dimensional images (Volume) are created according to the tissue reflectivity. As a result, structures with a high reflectivity will appear bright on SD-OCT images, whereas tissues that are easily crossed by the OCT signal will be dark.[25]

SD-OCT signal gets weaker as it deepens into the retina as the penetration is affected by each element the beam finds on its way. Furthermore, the RPE is highly reflective and it blocks most of the OCT light. In a normal eye, SD-OCT can hence provide good-quality images of the vitreous, the retina and the RPE but the visualization of the choroid is limited to its innermost layers (Fig. 2A).[25]

In 2009, a new imaging technique called enhanced depth imaging OCT (EDI-OCT) was described. This modality was still based on spectral domain technology but provided good-quality images of the deeper layers of the investigated structure using a different analysis of the SD-OCT signal.[26] Unfortunately, a drawback of EDI-OCT imaging is that details of the inner retina and the vitreous are lost to enhance the visualization of deeper structures (Fig. 2B).[27]

The introduction of EDI-OCT has been a real game changer in choroidal imaging as high-resolution images of the choroidal structure became easily collectable noninvasively both for research and clinical applications. EDI-OCT allows to measure CT and to appreciate its changes overtime in response to physiological stimuli (eg, circadian variations) and to pharmacological treatment. EDI images can also be used to visualize the structural features of the choroid, to distinguish its internal vascular layers and to detect and investigate choroidal lesions when present.[27]

Unfortunately, EDI-OCT has some limitations in imaging particularly thick choroids or highly pigmented eyes due to the fact that it is still based on the spectral domain technology. In fact, the choroid itself absorbs/deflects a great portion of OCT signal due to its internal structure, particularly if it is rich in melanin.[28]

Recently, a new technology called swept source OCT (SS-OCT) has become available in the clinical setting. SS-OCT devices employ a longer and broader wavelength compared with SD-OCTs and are characterized by a lower axial sensitivity decay.[24] These features make them able to overcome most of the EDI-OCT limitations, allowing a better penetration of the signal and good visualization of both the superficial and the deep structures at the same time in most conditions (Fig. 2C).[29]

OCT Angiography

OCT angiography (OCTA) is the latest iteration of noninvasive imaging and combines the structural information provided by OCT scans with blood flow detection, without the need to inject any dye.[30] OCTA is based on the repetition of multiple OCT scans on the same area of tissue and the identification of differences existing between consecutive scans. Any element of the tissue that changes its position from one scan to the next is considered to be moving and consequently coded as "flow." In contrast to OCTs that are usually interpreted by looking at b-scans, OCTA images are easier to be read by combining b-scans with a coronal reconstruction of the imaged area parallel to the surface of the retina (c-scan).[31]

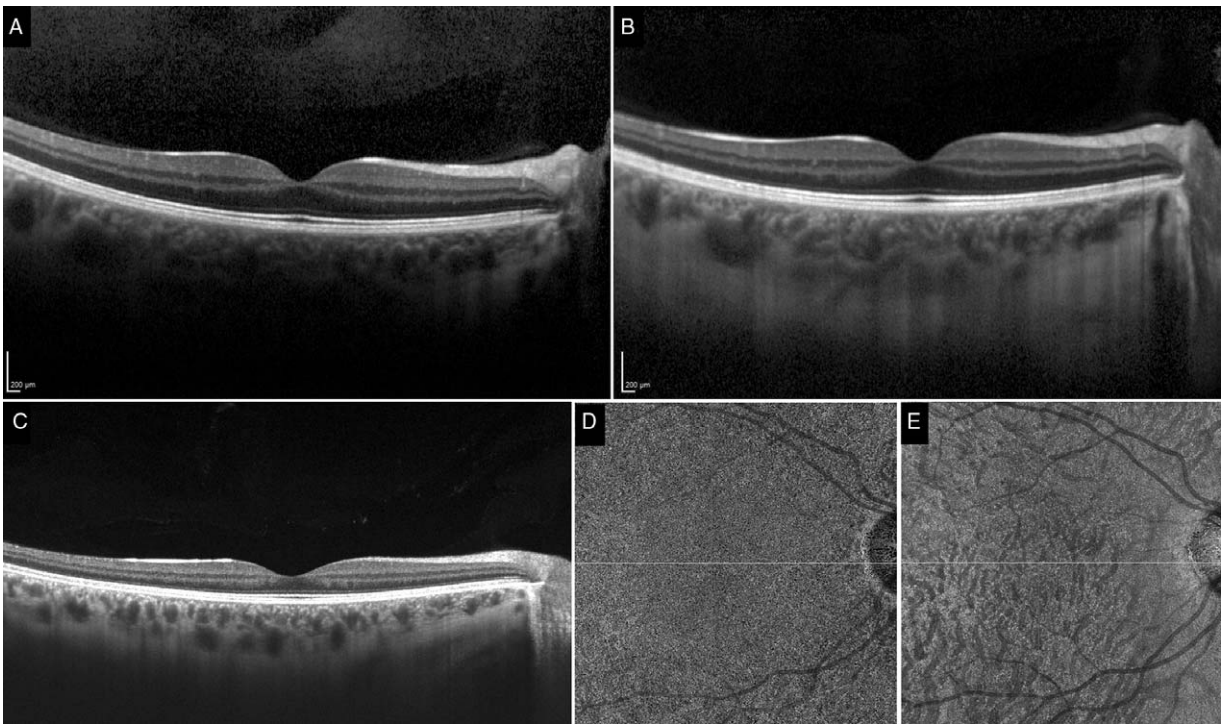

FIGURE 2. Optical coherence tomography (OCT) and OCT angiography (OCTA). (A) Spectral domain OCT of a normal eye encompassing the fovea. Note the visibility of the premacular bursa and the progressive loss of signal underneath the retinal pigment epithelium. (B) The same eye imaged with enhanced depth imaging (EDI) technology. Details of the vitreous are lost but the choroid is well-visualized. (C) Swept source technology allows to bypass spectral domain and EDI-OCT limitations allowing a good visualization of the vitreous, the retina, and the choroid at the same time. Nowadays, OCTA based on swept source technology allows to visualize the blood flow within the choriocapillaris (D). Moving deeper the flow within larger choroidal vessels is visible (E).

When OCTA was firstly introduced in the clinical practice its applications were limited to the study of the retinal circulation and the detection of choroidal neovascularization (CNV). The choroid was hard to be imaged for 2 reasons: first OCTA devices were based on SD-OCT technology thus the signal strength was too low to allow good-quality images beyond the RPE; secondly the presence of different capillary plexa in the retina created many projection artifacts on the underlaying tissues.[31,32]

With time, software-based correction of projection artifacts has been developed, devices with a higher acquisition speed and better resolution improved the quality of the scans, and the first analysis of the choroid with OCTA were attempted. Recently, OCTA devices based on swept source technology allow a good visualization of the choroidal blood flow in a noninvasive way.[31]

In a normal eye, the choroidal structure and flow can be easily visualized with SS-OCTA. A clear demarcation of the choriocapillaris from the underlying larger vessels of the choroid is usually performed by the automatic segmentation software provided with the device (Fig. 2D and E).[33] The combined analysis of b-scans and c-scan images allows investigation of structure and flow of the choroid at the same time.

First OCTA devices could image only very limited portions of the retina and choroid at a time and were not able to perform the scans outside the posterior pole. In the last few years, this technology has improved to the point that we are now able to image areas as large as 12×12 mm in a single acquisition and to merge multiple scans to cover a wider field. It is very likely that in the near future we will be able to collect images of the entire fundus and to explore the choroid up to the ora with noninvasive OCTA imaging.[30]

Advanced OCT Imaging Analysis: En-face OCT and Choroidal Vascularity Index

En-face OCT provides coronal or en-face view of choroid with transverse confocal scanning using volumetric cross-sectional OCT scans over the macular area (Fig. 3A). With the help of en-face imaging and increased penetration, high-quality images of the choroid with high axial resolution of 5.9 microns can be obtained. With advanced technique, posterior ciliary artery entry sites also can be visualized.[34]

Using en-face imaging, Alasil et al reported branching vascular pattern and focal or diffuse large choroidal vascular dilation in polypoidal choroidal vasculopathy.[35] Similarly, Ferrera et al described focal and diffuse pattern of vascular dilatation in patients of chronic central serous chorioretinopathy (CSCR) using enface SS-OCT imaging.[36] Wong et al reported a newer approach to evaluate choroidal vessels by using binarized en-face images (Fig. 3B). This technique provides binarized images of en-face choroidal scans at every 5 microns along with choroidal vascularity index of each slab.[37] Challenges with en-face imaging include shadowing from anterior structures, poor resolution, segmentation errors, and need for flattening to assess a particular layer. However, en-face imaging with increasing resolution can help noninvasively understand choroidal disorders especially diseases in the spectrum of the pachychoroid.

Choroidal vascularity index (luminal area/total choroidal area, CVI) can be calculated by binarizing cross-sectional single scan and for the whole volume (Fig. 3C–E). This index has been evaluated in healthy controls and in various diseases.[38,39] With advanced computational analysis, topographic choroidal

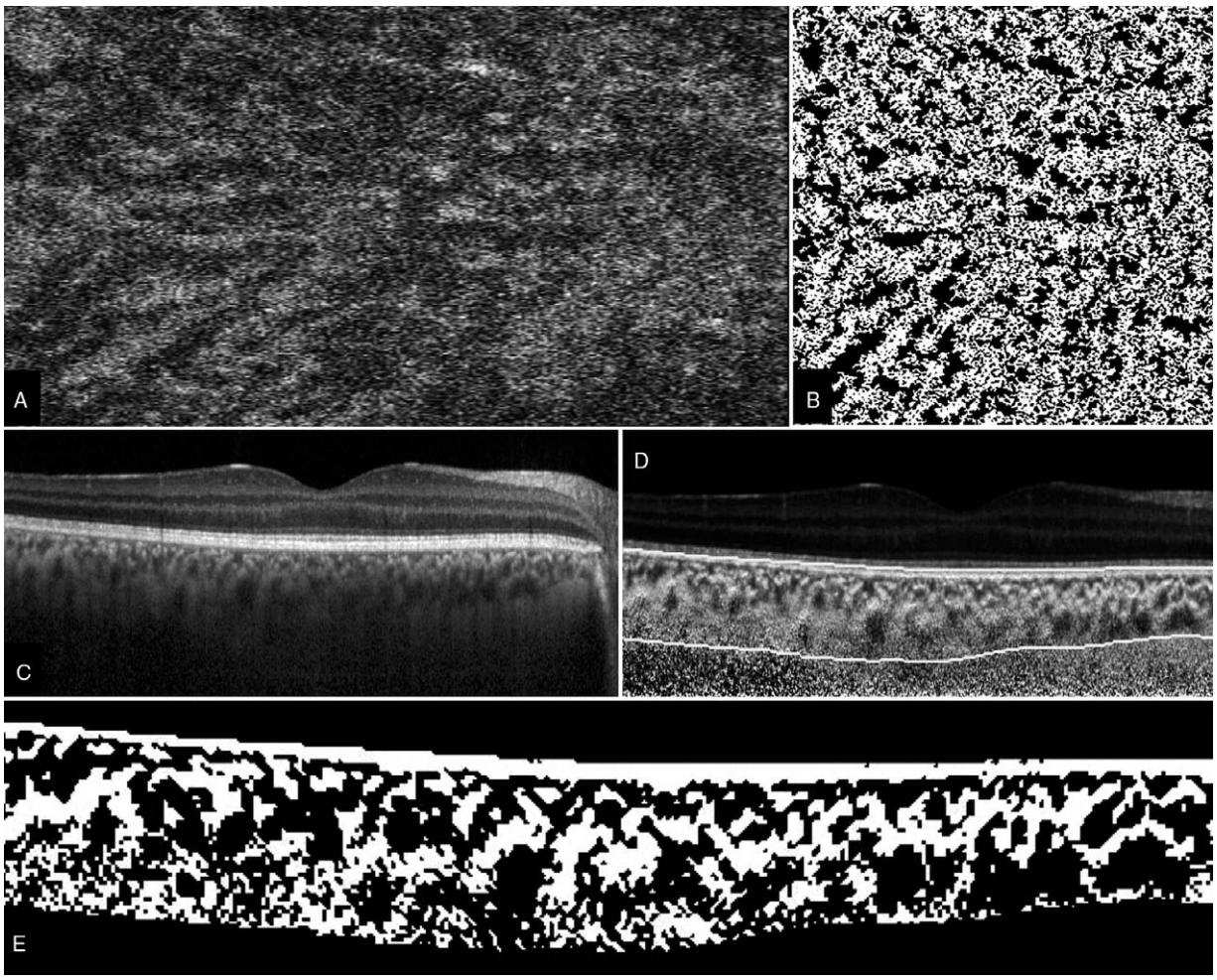

FIGURE 3. Advanced optical coherence tomography analysis. En-face optical coherence tomography allows to visualize the structure of the choroid on a coronal plan parallel to the retinal surface (A). Binarizing en-face images allows to appreciate the structure and calculate the extent of vessels lumen and stroma (B). On a standard enhanced depth imaging image (C) it is hard to appreciate the choroidal vascularity index (CDI = lumen/stroma). Such measurement is obtained by segmenting the choroid (D) and binarizing the image (E).

vascularity has also been reported to evaluate sectoral changes in the macular area. CVI appears to be more reproducible and reliable parameter than CT. CVI is proposed as imaging biomarker for disease diagnosis, disease activity, predictor of outcome, and recurrence.

Evaluation of individual layers of the choroid seems to be important especially with growing evidence for pachychroidal diseases. Thinning of inner choroid and enlargement of outer large choroidal vessels on OCT helps make the correct diagnosis. Automated segmentation of Haller vessel layers has been attempted for single scan.[40] However, differentiation of choriocapillaris from medium choroidal vessels still seems to be challenging. A recent report showed newer topographic assessment of CVI over the macular area using ETDRS map.[41] Sectoral analysis of CVI may further explain focal changes responsible for choroidal pathology. CVI analysis for wide field OCT scans has been reported and found to have large topographic variation with maximum CVI nasally.[42]

Choroidal assessment depends upon the visualization of choroidal borders and therefore further improvement in OCT techniques will help in making choroidal imaging biomarkers as integral part of clinical practice.

IMAGING THE CHOROID IN OCULAR DISEASES

Imaging the Choroid in Vascular Diseases

Many systemic diseases can cause alterations to the vasculature of the eye.

Choroidal evaluation of these conditions is important as the choroidal vasculature supplies the outer retina and altered choroidal blood flow may result in decreased photoreceptor functions. In addition, retinal vascular disorders may modulate the expression of vascular endothelial growth factors that can mediate changes in the choroidal vasculature.

The choroid is generally thick in vascular diseases that generate stasis at the level of the retinal vasculature. In central retinal vein occlusion subfoveal choroidal thickness (CT) is significantly thicker compared with fellow eyes.[43] In branch retinal vein occlusion eyes, this association was not found. However, CT was found to be significantly thicker in eyes with macular edema (ME) with subretinal fluid (SRF) but not in eyes with ME alone or no ME.[44] The investigators hypothesized that an increase in vascular endothelial growth factor (VEGF) caused by widespread ischemia increased choroidal hyperpermeability resulting in choroidal thickening and SRF.[45,46] (Fig. 4A and B)

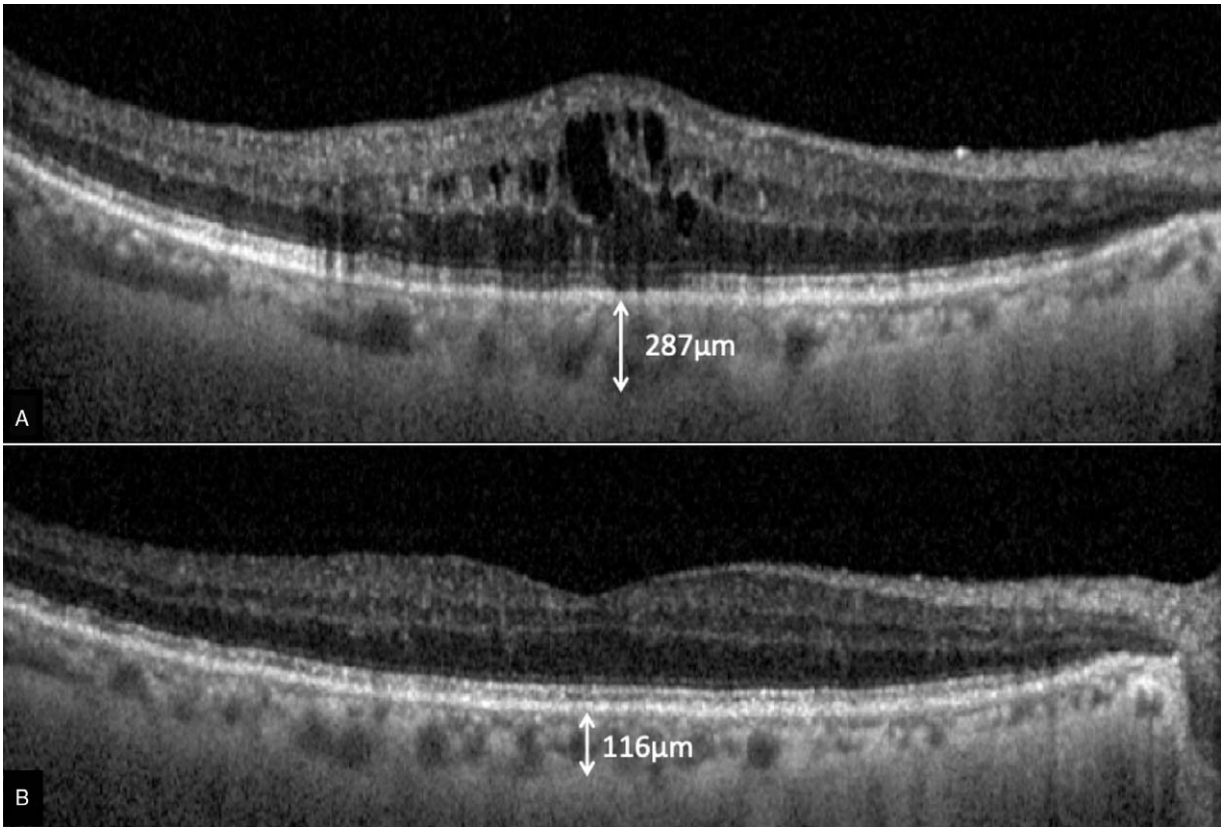

FIGURE 4. Choroidal changes in an eye with central retinal vein occlusion before and after treatment. Enhanced depth imaging optical coherence tomography (EDI-OCT) in an eye with central retinal vein occlusion demonstrates the presence of SRF and macular edema (A). After treatment with antivascular endothelial growth factor intravitreal injections the subretinal and intraretinal fluid disappear and the choroid gets much thinner (B).

Interestingly treatment with VEGF inhibitors and intravitreal dexamethasone implant resulted in thinning of the choroid.[46,47]

Choroidal thickening is also observed in hypertensive chorioretinopathy. Findings on EDI-OCT include marked choroidal thickening in eyes with hypertensive chorioretinopathy with SRF. These choroidal changes are noted to resolve on the normalization of blood pressure. It is postulated that the vasoconstriction of the choroidal vessels may result in the breakdown on the blood retina barrier and choroidal hyper permeability leading to choroidal thickening and accumulation of SRF.[48]

There are conflicting reports regarding the choroidal characteristics of eyes with diabetic retinopathy (DR). Several reports reported thin choroid in DR, with this change being more pronounced in proliferative DR and diabetic ME (DME).[49,50] Opposing studies report a thickening of the choroid in eyes with DME.[51] Recently, OCTA studies have reported microvasculature abnormalities and areas of flow impairment at the level of the choriocapillaris in all stages of DR.[52] The clinical relevance of these new findings is still to be determined.

Pan retinal photocoagulation appears to result in a reduction in CT.[53,54] The suggested mechanism for this appears to be a reduction in choroidal hyperpermeability or atrophy of choroidal vessels.[55] As reported for vein occlusions, intravitreal anti-VEGFs and steroids result in significant thinning of CT.[56,57] These findings lend evidence to the suggested VEGF driven mechanism of CT thickening in DR and DME but require further research to ascertain.

There is a paucity of data regarding choroidal changes in eyes with retinal artery occlusions and ocular ischemic syndrome. In addition, shadowing from the higher internal reflectivity of the retina from ischemic changes makes it difficult to differentiate the chorioscleral interface. Nonetheless, thinning of the choroid is noted as a result of the disruption of blood flow in the choroidal vasculature. This is similar in cases of ocular ischemic syndrome.[58]

Imaging the Choroid in Retinal Dystrophies

Inherited retinal disease (IRD) primarily affects the outer retinal layers and results in loss of photoreceptors and RPE. Eyes with IRD may appear normal at ophthalmoscopic examination and functional studies are often required to help diagnose. However, imaging techniques including angiography, fundus autofluorescence (FAF), and OCT can help sort out retinal degradation and dystrophy types.[59]

In a patient with Stargardt macular dystrophy (STMD) fundus fluorescein angiography (FFA) demonstrates a “dark" or “silent" choroid from blockage of early choroidal fluorescence by the lipofuscin deposition within the RPE.[60] STMD also has a particular appearance on ICG. In fact, in late phases of the angiogram, the atrophic areas turn dark (dark atrophy) (Fig. 5).[61] This allows differentiation of patients affected by STMD from those with atrophy due to other causes. The dark atrophy is explained by the extensive damage to the choriocapillaris in STMD eyes, which is also confirmed by OCTA.[62]

Some retinal dystrophies demonstrate increased autofluorescence in areas of excess lipofuscin buildup such as RP and typically have an FAF pattern encircling the macula.[63] Classically, RP is defined as degeneration primarily of rod photoreceptors followed by degeneration of the cone photoreceptors, but many studies show RPE loss and also changes at the level of

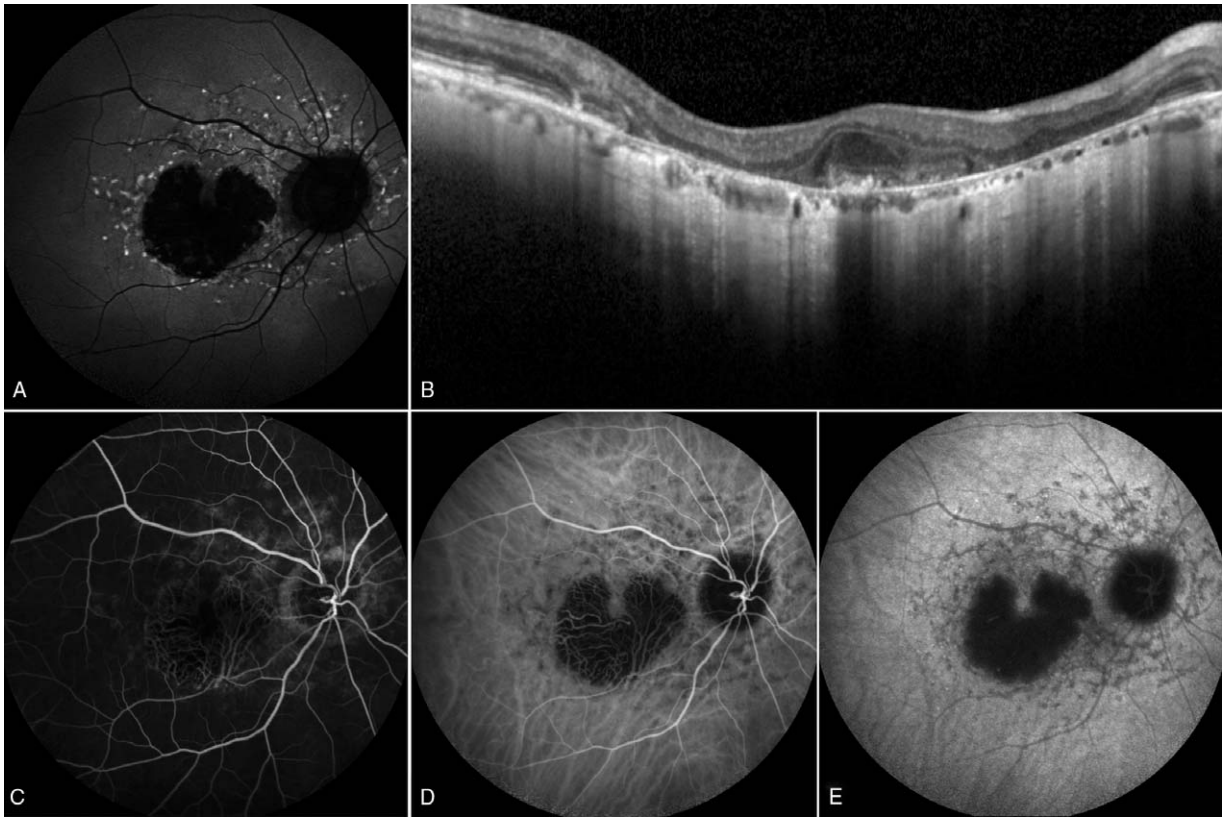

FIGURE 5. Multimodal imaging in a patient with Stargardt Macular Dystrophy (STMD). Fundus autofluorescence (A) demonstrates an area of atrophy with decreased autofluorescence at the posterior pole surrounded by the typical hyperfluorescent flecks. Enhanced depth imaging optical coherence tomography (B) allows to visualize the atrophy of the retinal pigment epithelium and the outer retina. Only the large vessels of the choroid are well represented, whereas the choriocapillaris is almost invisible. Early phases of fluorescein angiography (C) allows to appreciate the so-called ''Dark Choroid.'' Large choroidal vessels are seen during the early phases of indocyanine green angiography (ICGA) in the atrophic central area (D). During the later phases of the ICGA (E) the same area becomes dark (dark atrophy) due to the lack of choriocapillaris.

the choriocapillaris and the choroid. Imaging can help in detecting and monitoring these vascular alterations; in particular, OCTA is able to visualize a reduction in the choriocapillaris density and the presence of many flow voids in patients with RP.[64] The deeper layers of the choroid have also been shown to be affected with decrease of the CVI on OCT compared with healthy eyes.[65]

Differently from RP, some retinal dystrophies have decreased FAF due to the loss of lipofuscin with end-stage chorioretinal atrophy.[66] Choroideremia is a typical example of the latter and is the most common X-linked hereditary choroidal dystrophy characterized by a progressive degeneration of the outer retina and choriocapillaris imaged with OCT and FAF.[67]

Although genetic testing is becoming more readily available, multimodal imaging in combination with clinical evaluation and patient's symptoms provides a useful framework for directing specific genetic investigations. IRDs typically present with outer retinal changes; however, the choriocapillaris and the choroid are more affected than we used to think and should be investigated to monitor the progression of these diseases.

Imaging the Choroid in Age-Related Macular Degeneration

Early choriocapillaris loss has been noted on histopathology in early age-related macular degeneration (AMD) compared with age-matched controls.[68] Such changes are more obvious under drusen and this choriocapillaris loss has been reported to be associated with disease progression. Seddon et al reported choriocapillaris loss in all stages of AMD with this loss being higher in eyes with geographic atrophy (up to 39.0%) and choroidal neovascularization (up to 38.2%).[69] In neovascular AMD, abnormal choroidal vessels were noted above or below the RPE with associated decreased choroidal vascularity especially choriocapillaris loss.

Although FFA shows window defects over the areas of RPE atrophy and FFA does help in defining CNV activity, it does not provide clear information about the choroid. By contrast, ICGA plays an important role in differentiating various types of choroidal neovascular membranes, stages of choroidal vascularization, and unique entities such as type 3 and polypoidal lesions (PCV).

From diagnosis to follow-up and prognosticating outcome, OCT plays an important role in every stage of AMD management. EDI-OCT demonstrates progressive subfoveal choroidal thinning of the choroid in all stages of AMD.[70] CT has also been evaluated in eyes with different types of drusen and is found to be the thinnest in eyes with reticular drusen which have the highest risk of progression to advanced AMD and neovascular conversion.[71,72] Thicker choroid on EDI-OCT suggests PCV which can often require a different treatment approach.[73]

Choroidal vascularity index has been shown to be reduced in AMD eyes compared with age-matched control.[74] CT and CVI can also be used as biomarkers for disease activity as they both increase during the active stage of the neovascular AMD (Fig. 6 A and D).[75]

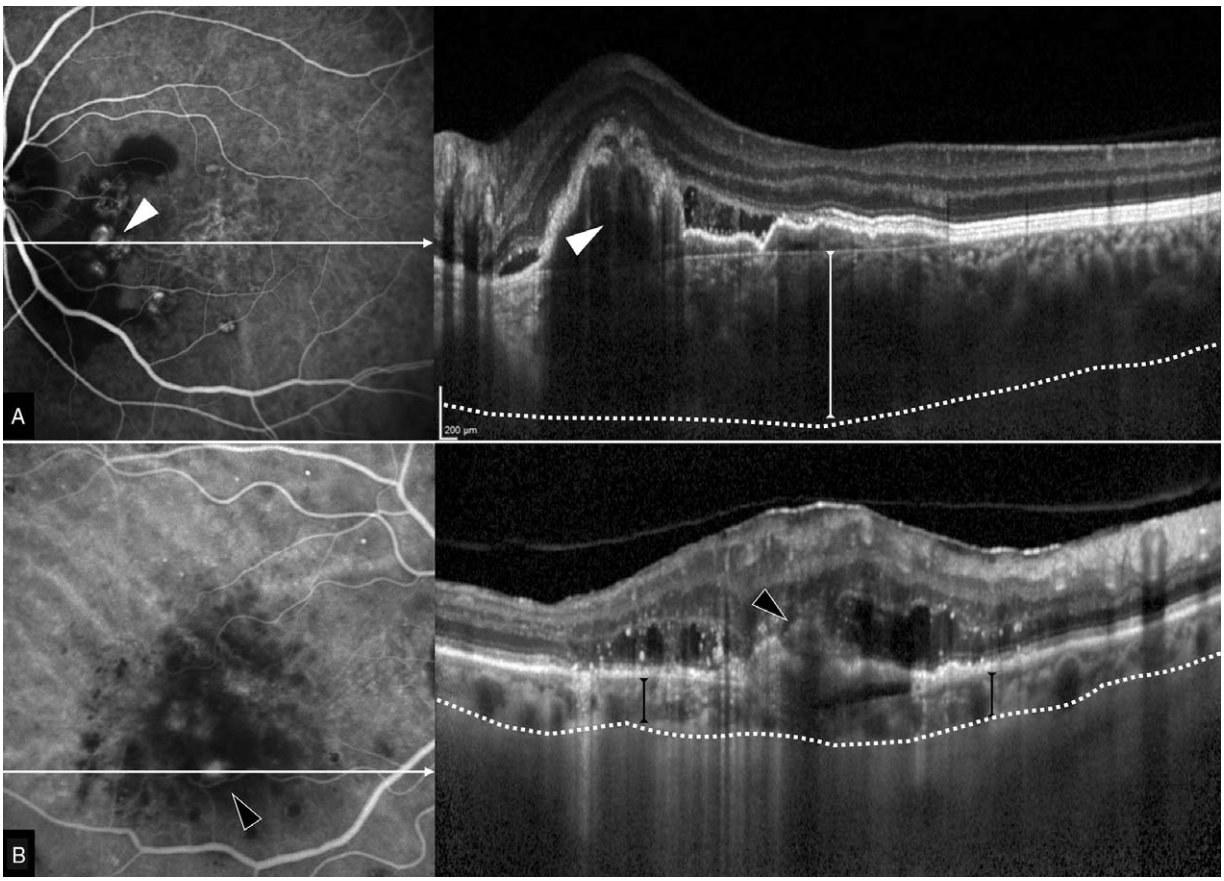

FIGURE 6. Choroidal features in age related macular degeneration. (A) Combined indocyanine green angiography (ICGA) and enhanced depth imaging optical coherence tomography (EDI-OCT) in a patient with pachychoroid and polypoidal lesions (white arrowheads). The choroid is very thick and enlarged vessels are visible deep in the choroid. (B) Combined ICGA and EDI-OCT in a patient with a type 3 choroidal neovascularization (retinal angiomatous proliferation). The lesion (black arrowhead) shows the typical leakage on late ICGA and the intraretinal fluid on OCT. In contrast with the previous case, the choroid is very thin, a common feature of eyes with type 3 lesions.

Using en-face OCT, Baek et al showed presence of pachyvessel in 46% of typical AMD, which is still lower than typical PCV and CSCR. However, the pattern of pachyvessels was focal in eyes with neovascular AMD in contrast to diffusively thick-choroid in eyes with PCV and CSCR.[76] Similarly, mean choroidal vascular density was higher in CSCR and thick-choroid PCV (52%–55%) compared with AMD (45%–47%).

OCTA also assists with the understanding of choroidal vascularity changes in AMD. There is choriocapillaris loss in early stages of AMD, with further loss associated with progression in AMD stages.[77] Borrelli et al showed increased choriocapillaris (CC) impairment underneath drusen and surrounding region.[78] CC loss correlates with the enlargement rate of geographic atrophy.[79] Further into the predictive power of OCTA in dry AMD, Nassisi et al reported that significant CC flow impairment under the regions of intact RPE may suggest enlargement of drusen or new drusen formation.[80]

Imaging the Choroid in Uveitis

The vascular tissue plays a key role in any inflammatory process occurring within our body. During acute inflammation, vessels dilate to increase the blood flow toward the inflamed area and capillary permeability increases to promote the diffusion of inflammatory cells and proteins across the vascular wall to the interstitial compartment. If inflammation resolves within a short amount of time, things return to normal; however, if the process lasts longer, chronic changes and tissue damage can occur.

Being the vascular tunic of the eye, the choroid undergoes all the above-mentioned changes anytime inflammation occurs within the eye regardless of the primary site of inflammation.[6] In fact, choroidal thickening and blood flow perturbations have been described in all kind of uveitis from anterior to posterior.[81] In most uveitis, the choroid is secondarily involved as its vessels respond to the inflammatory stimulus,[82] but in some entities it represents the primary site of inflammation[83] and pathognomonic lesions or patterns of change can be imaged.[84]

In anterior uveitis, intermediate uveitis, and retinitis or retinal vasculitis, the choroid is not the primary site of inflammation. Still its thickness increases when active inflammation is present and decreases when the eye is quiet. EDI-OCT or SS-OCT is useful imaging technique to monitor CT overtime in these conditions and can drive the clinician in the assessment of treatment efficacy (Fig. 7A and B).[82]

Some uveitis conditions can directly target the choroid as the primary site of inflammation. In these cases, more comprehensive imaging is required to characterize the lesions and to differentiate between the various diseases.

ICGA should always be performed in case of posterior uveitis to identify choroidal involvement.[85] Hypofluorescent choroidal lesions becoming more evident in the late phases of the angiogram denotes choriocapillaris hypoperfusion typically seen in diseases

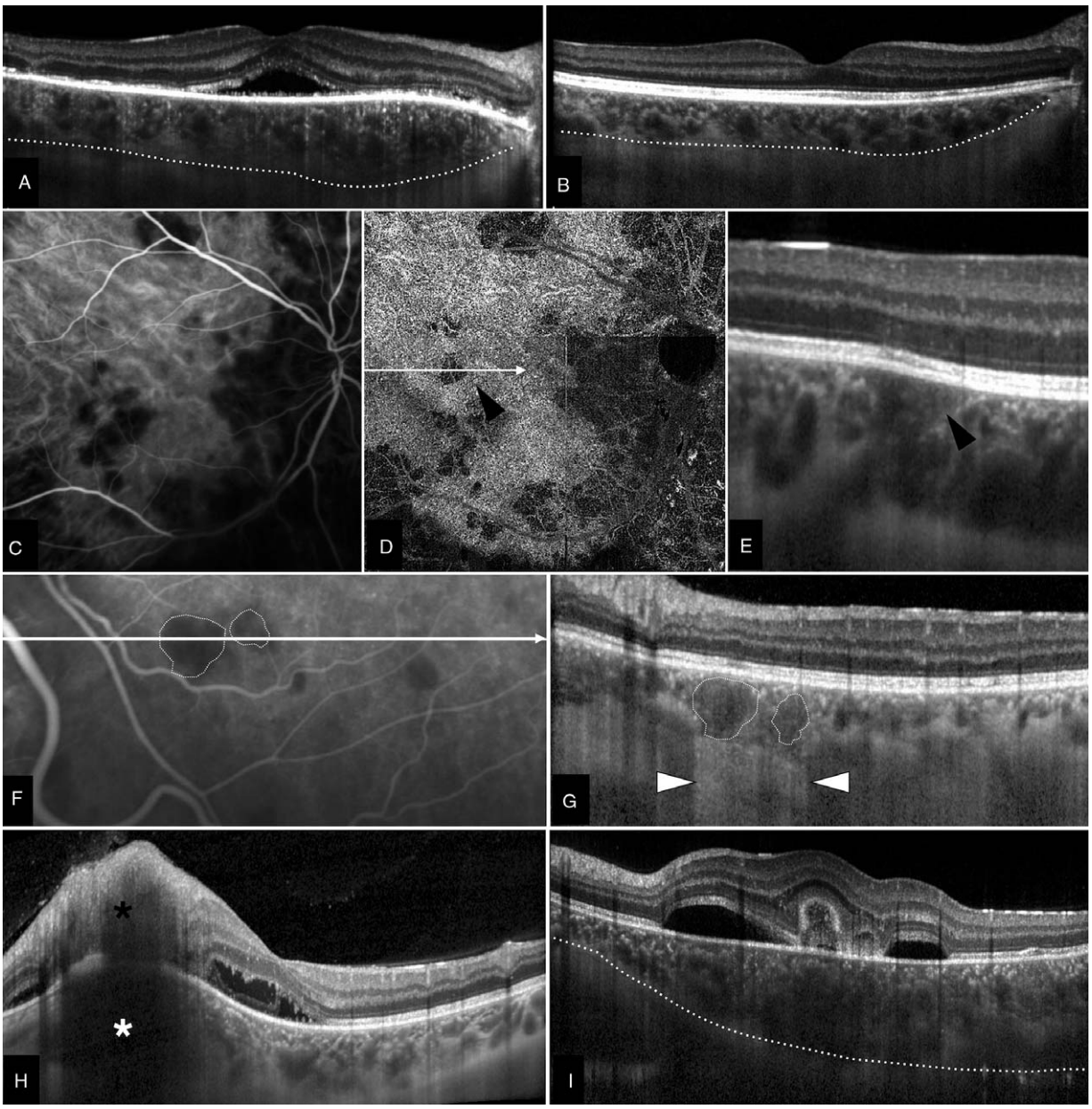

FIGURE 7. Multimodal imaging of the choroid in uveitis. Monitoring choroidal thickness with enhanced depth imaging optical coherence tomography (EDI-OCT) or swept source OCT is of extreme importance in inflammatory diseases of the eye as the choroid gets thicker when the eye is inflamed (A) and thins in response to treatment (B). In case of choriocapillaris hypoperfusion (acute posterior multifocal placoid pigmented epithelitis, serpiginous choroidis etc.) indocyanine green angiography ICGA (C) demonstrates hypofluorescent more visible in the late phases of the examination. OCT angiography (D) perfectly overlaps with ICGA demonstrating absence of flow in the same areas. The affected regions (black arrowhead) show thickened and darker choriocapillaris on EDI-OCT scans (E). Choroidal granulomas (encircled by white dotted line) appear as hypofluorescent areas less visible during the late phases of the angiogram on ICGA (F). The same formations look homogeneously hyperreflective on EDI-OCT (G) with a distinctive increased transmission effect underneath (white arrowheads). A focal choroidal thickening (white asterisk) under an area of retinitis (black asterisk) on EDI-OCT (H) is a suggestive sign for toxoplasmic chorioretinitis. On the contrary, a diffuse choroidal thickening is typical of Vogt-Koyanagi-Harada disease (I).

such as acute posterior multifocal placoid pigment epitheliopathy and serpiginous choroiditis (Fig. 7C).[86] By contrast, hypofluorescent choroidal lesions that fade or turning isofluorescent in the late phases of the investigation usually correspond to choroidal granulomas due to sarcoidosis, tuberculosis, or Vogt-Koyanagi-Harada disease (Fig. 7F).[87]

Structural images of the choroid obtained with EDI-OCT and SS-OCT allow to visualize specific patterns of thickening or focal lesions in diseases targeting the choroid. In case of acute posterior multifocal placoid pigment epitheliopathy and serpiginous choroiditis, the choriocapillaris gets thicker and hyporeflective in the hypoperfused areas (Fig. 7E).[86] A massive and diffuse thickening of the entire choroid is typical of Vogt-Koyanagi-Harada disease (Fig. 7I).[88] By contrast, a focal thickening with loss of the typical sponge-like architecture of the choroid is seen in toxoplasmic chorioretinitis (Fig. 7H).[89] Finally, choroidal granulomas can be seen on EDI-OCT and SS-OCT scans as hyporeflective round-shaped areas characterized by an increased transmission effect on the underlying structures (Fig. 7G).[87]

Advanced imaging analysis has also been reported as useful in uveitis. CVI usually increases in inflamed eyes as the vessels dilate and the ratio between the lumen and the stroma increases.[90] Enlargement of specific choroidal layers has also been described in specific uveitis diseases.[91] The clinical impact of these advanced images is still to be determined.

OCTA of the choroid is a relatively new imaging technique, so there is less experience and literature on it.[33] Choriocapillaris hypoperfusion is easily seen with OCTA (Fig. 7D) and can be followed overtime, whereas large choroidal granulomas can compress the surrounding vasculature and appear as dark areas on OCTA.[33] Finally, OCTA is very useful in distinguishing inflammatory CNV that can complicate certain types of uveitis from inflammatory lesions without new vessels.[92]

To conclude, a multimodal imaging approach is always recommended in patients with posterior uveitis to investigate possible choroidal involvement and to aid in diagnosis. In all uveitis, including those primarily affecting the anterior segment,

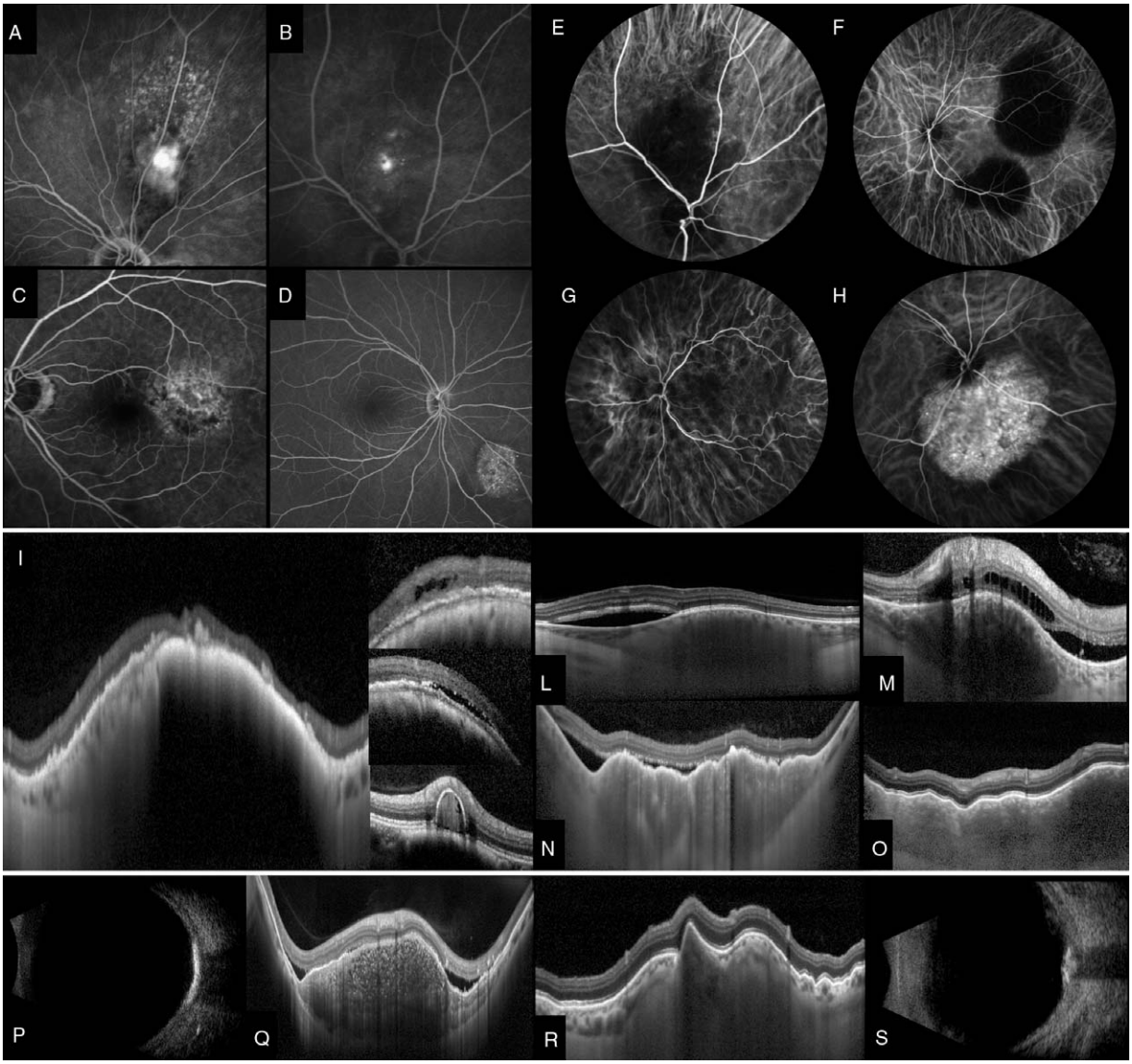

FIGURE 8. Multimodal imaging of the choroid in ocular onchology. (A–D) Fluorescein angiography (FA) respectively in a case of choroidal nevus (A), choroidal melanoma (B), circumscribed choroidal hemangioma (C), and metastasis originating from a lung adenocarcinoma (D). FA does not provide useful insights in the management of these patients with similar staining or leakage patterns in the late frames. Only in case A, FA was used to confirm a secondary choroidal neovascularization (CNV) in a nevus with SRF detected at optical coherence tomography (OCT) study. (E–H) Indocyanine green angiography (ICGA) study in a choroidal melanoma (E), multiple choroidal metastasis (F), choroidal lymphoma (G), and a circumscribed choroidal hemangioma (H). As reported in the dedicated paragraph, ICGA does not represent a feasible tool for differentiating a nevus from a melanoma or a metastasis; similarly, ICGA in choroidal lymphoma may resemble a granulomatous uveitis. Ideally, ICGA imaging should be rather used to prove or confirm the diagnosis of a circumscribed choroidal hemangioma and its hyperfluorescence should be considered pathognomonic also in absence of the well-known ''late wash-out'' phenomenon. OCT study in a choroidal nevus showing a typical hyper-reflective band with posterior shadowing in the pigmented component of the lesion and significant outer retinal remodeling and atrophy as a sign of previous subretinal exudation (I). OCT in nevi may also show intraretinal fluid, subretinal exudation, or a pigment epithelium detachment (PED). Choroidal melanoma (L) and hemangioma (M) also typically show a dome shape configuration with the first one showing inner choroidal structures compression and the latter a global expansion of the choroid and choriocapillaris assocated to intraretinal schisis and subretinal fluid (SRF). (N, O) Undulated patterns in a choroidal metastasis (N) and choroidal lymphoma (O). In calcified lesions appearing similar at ultrasound (US) (P, S) examination, OCT may also allow a precise distinction between a benign choroidal tumor (choroidal osteoma) (Q) and a scleral lesion (sclerochoroidal calcification) (R).

CT should be monitored using EDI-OCT or SS-OCT as this marker may be the first to indicate a response to treatment or a disease recurrence.[93,83] Specific lesions such as choriocapillaris hypoperfusion or choroidal granulomas can also be investigated and monitored over time with ICG, EDI-OCT/SS-OCT, and OCTA. To conclude, a multimodal imaging approach is always recommended in patients with posterior uveitis to investigate possible choroidal involvement and to aid in diagnosis. In all uveitis, including those primarily affecting the anterior segment, CT should be monitored using EDI-OCT or SS-OCT as this marker may be the first to indicate a response to treatment or a disease recurrence.[93,83] Specific lesions such as choriocapillaris hypoperfusion or choroidal granulomas can also be investigated and monitored over time with ICG, EDI-OCT/SS-OCT, and OCTA.

Imaging the Choroid in Ocular Oncology

Despite major improvements in the field of fundus imaging, the diagnosis and management of choroidal tumors remain mainly clinical and performed by means of indirect ophthalmoscopy and ocular ultrasound in the majority of cases. Ophthalmoscopy and fundus photography, in particular, provide instrumental information such as color, location, shape, multifocality, possible presence of associated features (orange pigment, secondary retinal detachment, bleeding, chronic changes), whereas ultrasonography should be considered mandatory offering qualitative information and providing reliable measurements. Nevertheless, especially for small choroidal tumors, ultrasound interpretation may be challenging and possibly misleading and a multiimaging approach should be performed in doubtful, atypical cases to facilitate the referral to ocular oncology practices, avoiding delays in diagnosis and treatments.

In regard to the application of dye-based imaging techniques, neither FFA nor ICGA currently represent the ideal tools for diagnosing choroidal tumors. Fluorescein angiography, in particular, allows for an optimal assessment of the retinal circulation but does not provide reliable images of the choroid (Fig. 8A–D). ICGA represents the ideal tool to study subretinal alterations but to date no diagnostic reliable patterns have been described to differentiate small melanocytic tumors and the majority of choroidal neoplasms show variable degrees of early and late hypofluorescence (Fig. 8 E-H). Studies have focused on ICGA feasibility in evidencing tumor intrinsic microvascular patterns but intralesional vascularity has never been considered a reliable diagnostic element.[94–97] For these reasons, ICGA should be considered when suspecting a circumscribed choroidal hemangioma (showing a pathognomonic rapid and lobular filling with early hyperfluorescence and late wash-out) (Fig. 8H),[98] for better defining tumor margins, for the assessment of uveal pseudomelanomas and for a baseline imaging to be used in monitoring treatment results and side effects.

Among noninvasive imaging techniques, FAF and EDI-OCT provide new insights in the study of choroidal tumors.[99] FAF can be useful for identifying lipofuscin at the apex of a melanocytic tumor, to document either chronic RPE atrophy or outer retinal layers remodeling and to see gravitational patterns. In contrast, OCT by means of EDI technique and swept source technology is nowadays an instrumental tool for analyzing choroidal tumors since it offers the opportunity to recognize typical patterns. Dome-shaped configuration has been described for both melanocytic tumors and circumscribed choroidal hemangioma (Fig. 8 I–M); melanocytic lesions also have overt choriocapillaris compression and sometimes SRF[23] (Fig. 8 I, L), whereas choroidal hemangiomas is also characterized by diffuse expansion of medium/large size choroidal vessels with no choriocapillaris compression (Fig. 8M). Undulated patterns (Fig. 8N,O) are typical of choroidal metastases (lumpy-bumpy appearance) and choroidal lymphoma (sea-sick pattern), whereas in choroidal osteoma (Fig. 8Q) a typical transparency with multilayer appearance, possible intrinsic spongy pattern,[100] bone lamellae are usually observed despite the typical hyperreflectivity casting a posterior shadow at USG (Fig. 8P) .

Despite its increasing use in ophthalmology, OCTA has not yet played a noticeable role in ocular oncology at least from a diagnostic standpoint. With the diffusion of swept source devices that are able to guarantee higher tissue penetration and increased scan window depth (currently up to 6 mm), OCTA is primarily used to determine the presence of secondary CNV in small melanocytic lesions with SRF.[101] However, its application in the study of radiation maculopathy has been validated and more recently, the use of "extended field imaging" OCTA techniques[102] has guaranteed wider fields of examination and a simultaneous, rapid assessment of macular and post-equatorial changes.

Despite the diagnosis remaining mainly clinical, a multiimaging approach is warranted in the majority of patients. FA and ICGA should be performed to document the tumor size at baseline, but they do not play a real diagnostic role with few exceptions. EDI-OCT and SS-OCT are currently essential tools for documenting some of the above-mentioned patterns and highlighting minor changes in shape or size that may be not visible at USG, whereas OCTA application still needs validation.

CONCLUSIONS

In the last 20 years we have moved from "guessing" what was happening beyond the RPE to actually visualize structural and functional changes of the choroid in vivo noninvasively. Imaging is on an exponential growth and technological development, continuously providing clinicians and researchers with new information to understand the role of the choroid in many ocular conditions and better manage chorioretinal diseases. This review summarized the state-of-the-art in choroidal imaging and it is hoped that provided readers with good basic principles to interpret different imaging techniques and understand choroidal findings in the most common diseases affecting the posterior segment of the eye.

REFERENCES

1. Nickla DL, Wallman J. The multifunctional choroid. *Prog Retin Eye Res*. 2010;29:144–168.
2. Borrelli E, Sarraf D, Freund KB, Sadda SR. OCT angiography and evaluation of the choroid and choroidal vascular disorders. *Prog Retin Eye Res*. 2018;67:30–55.
3. Ferrara D, Waheed NK, Duker JS. Investigating the choriocapillaris and choroidal vasculature with new optical coherence tomography technologies. *Prog Retin Eye Res*. 2016;52:130–155.
4. Rasheed MA, Singh SR, Invernizzi A, et al. Wide-field choroidal thickness profile in healthy eyes. *Sci Rep*. 2018;8:17166.

5. Hogan MJ, Alvarado JA, Weddell JE. Histology of the Human Eye; an Atlas and Textbook. Philadelphia: Saunders; 1971.
6. Levin LA, Adler FH. Adler's Physiology of the Eye. Edinburgh: Saunders/Elsevier; 2011.
7. May CA, Neuhuber W, Lütjen-Drecoll E. Immunohistochemical classification and functional morphology of human choroidal ganglion cells. *Invest Ophthalmol Vis Sci*. 2004;45:361–367.
8. Flower RW, Hochheimer BF. A clinical technique and apparatus for simultaneous angiography of the separate retinal and choroidal circulations. *Invest Ophthalmol*. 1973;12:248–261.
9. Yannuzzi LA, Slakter JS, Sorenson JA, Guyer DR, Orlock DA. Digital indocyanine green videoangiography and choroidal neovascularization. *Retina*. 1992;12:191–223.
10. Destro M, Puliafito CA. Indocyanine green videoangiography of choroidal neovascularization. *Ophthalmology*. 1989;96:846–853.
11. Flower RW. Choroidal angiography today and tomorrow. *Retina*. 1992;12:189–190.
12. Scheider A, Schroedel C. High resolution indocyanine green angiography with a scanning laser ophthalmoscope. *Am J Ophthalmol*. 1989;108:458–459.
13. Yannuzzi LA. Indocyanine green angiography: a perspective on use in the clinical setting. *Am J Ophthalmol*. 2011;151:745–751.
14. Hope-Ross M, Yannuzzi LA, Gragoudas ES, et al. Adverse reactions due to indocyanine green. *Ophthalmology*. 1994;101:529–533.
15. Beem E, Segal MS. Evaluation of stability and sensitivity of cell fluorescent labels when used for cell migration. *J Fluoresc*. 2013;23:975–987.
16. Kornfield TE, Newman EA. Measurement of retinal blood flow using fluorescently labeled red blood cells. *eNeuro*. 2015;0005-15.2015.
17. Matsuda N, Ogura Y, Nishiwaki H, et al. Visualization of leukocyte dynamics in the choroid with indocyanine green. *Invest Ophthalmol Vis Sci*. 1996;37:2228–2233.
18. Takasu I, Shiraga F, Okanouchi T, Tsuchida Y, Ohtsuki H. Evaluation of leukocyte dynamics in choroidal circulation with indocyanine green-stained leukocytes. *Invest Ophthalmol Vis Sci*. 2000;41:2844–2848.
19. Mundt GH, Jr., Hughes WF, Jr. Ultrasonics in ocular diagnosis. *Am J Ophthalmol*. 1956;41:488–498.
20. Pavlin CJ, Harasiewicz K, Sherar MD, Foster FS. Clinical use of ultrasound biomicroscopy. *Ophthalmology*. 1991;98:287–295.
21. Foster FS, Pavlin CJ, Harasiewicz KA, Christopher DA, Turnbull DH. Advances in ultrasound biomicroscopy. *Ultrasound Med Biol*. 2000;26:1–27.
22. Singh R, Invernizzi A, Agarwal A, Kumari N, Gupta A. Enhanced depth imaging spectral domain optical coherence tomography versus ultrasonography B-Scan for measuring retinochoroidal thickness in normal eyes. *Retina*. 2015;35:250–256.
23. Shah SU, Kaliki S, Shields CL, Ferenczy SR, Harmon SA, Shields JA. Enhanced depth imaging optical coherence tomography of choroidal nevus in 104 cases. *Ophthalmology*. 2012;119:1066–1072.
24. de Boer JF, Leitgeb R, Wojtkowski M. Twenty-five years of optical coherence tomography: the paradigm shift in sensitivity and speed provided by Fourier domain OCT [Invited]. *Biomed Opt Express*. 2017;8:3248–3280.
25. Drexler W, Fujimoto JG. State-of-the-art retinal optical coherence tomography. *Prog Retin Eye Res*. 2008;27:45–88.
26. Margolis R, Spaide RF. A pilot study of enhanced depth imaging optical coherence tomography of the choroid in normal eyes. *Am J Ophthalmol*. 2009;147:811–815.
27. Mrejen S, Spaide RF. Optical coherence tomography: imaging of the choroid and beyond. *Surv Ophthalmol*. 2013;58:387–429.
28. Invernizzi A, Giani A, Cigada M, Staurenghi G. Retrobulbar structure visualization with enhanced depth imaging optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2013;54:2678–2684.
29. Spaide RF. Visualization of the posterior vitreous with dynamic focusing and windowed averaging swept source optical coherence tomography. *Am J Ophthalmol*. 2014;158:1267–1274.
30. Kashani AH, Chen CL, Gahm JK, et al. Optical coherence tomography angiography: a comprehensive review of current methods and clinical applications. *Prog Retin Eye Res*. 2017;60:66–100.
31. Spaide RF, Fujimoto JG, Waheed NK, Sadda SR, Staurenghi G. Optical coherence tomography angiography. *Prog Retin Eye Res*. 2018;64:1–55.
32. Spaide RF, Fujimoto JG, Waheed NK. Image artifacts in optical coherence tomography angiography. *Retina*. 2015;35:2163–2180.
33. Pichi F, Sarraf D, Arepalli S, et al. The application of optical coherence tomography angiography in uveitis and inflammatory eye diseases. *Prog Retin Eye Res*. 2017;59:178–201.
34. Motaghiannezam R, Schwartz DM, Fraser SE. In vivo human choroidal vascular pattern visualization using high-speed swept-source optical coherence tomography at 1060 nm. *Invest Ophthalmol Vis Sci*. 2012;53:2337–2348.
35. Alasil T, Ferrara D, Adhi M, et al. En face imaging of the choroid in polypoidal choroidal vasculopathy using swept-source optical coherence tomography. *Am J Ophthalmol*. 2015;159:634–643.
36. Ferrara D, Mohler KJ, Waheed N, et al. En face enhanced-depth swept-source optical coherence tomography features of chronic central serous chorioretinopathy. *Ophthalmology*. 2014;121:719–726.
37. Wong RL, Singh SR, Rasheed MA, et al. En-face choroidal vascularity in central serous chorioretinopathy. *Eur J Ophthalmol*. 2020. doi:10.1177/1120672120908719.
38. Velaga SB, Nittala MG, Vupparaboina KK, et al. Choroidal vascularity index and choroidal thickness in eyes with reticular pseudodrusen. *Retina*. 2020;40:612–617.
39. Agrawal R, Chhablani J, Tan KA, Shah S, Sarvaiya C, Banker A. Choroidal vascularity index in central serous chorioretinopathy. *Retina*. 2016;36:1646–1651.
40. Uppugunduri SR, Rasheed MA, Richhariya A, Jana S, Chhablani J, Vupparaboina KK. Automated quantification of Haller's layer in choroid using swept-source optical coherence tomography. *PLoS One*. 2018;13:e0193324.
41. Goud A, Singh SR, Sahoo NK, et al. New Insights on Choroidal Vascularity: A Comprehensive Topographic Approach. *Invest Ophthalmol Vis Sci*. 2019;60:3563–3569.
42. Singh SR, Invernizzi A, Rasheed MA, et al. Wide-field choroidal vascularity in healthy eyes. *Am J Ophthalmol*. 2018;193:100–105.
43. Tsuiki E, Suzuma K, Ueki R, Maekawa Y, Kitaoka T. Enhanced depth imaging optical coherence tomography of the choroid in central retinal vein occlusion. *Am J Ophthalmol*. 2013;156:543–547.
44. Shin YU, Lee MJ, Lee BR. Choroidal maps in different types of macular edema in branch retinal vein occlusion using swept-source optical coherence tomography. *Am J Ophthalmol*. 2015;160:328–334.

45. Gallego-Pinazo R, Dolz-Marco R, Pardo-Lopez D, et al. Ranibizumab for serous macular detachment in branch retinal vein occlusions. *Graefes Arch Clin Exp Ophthalmol*. 2013;251:9–14.
46. Kim KH, Lee DH, Lee JJ, Park SW, Byon IS, Lee JE. Regional choroidal thickness changes in branch retinal vein occlusion with macular edema. *Ophthalmologica*. 2015;234:109–118.
47. Lee EK, Han JM, Hyon JY, Yu HG. Changes in choroidal thickness after intravitreal dexamethasone implant injection in retinal vein occlusion. *Br J Ophthalmol*. 2015;99:1543–1549.
48. Ugarte M, Horgan S, Rassam S, Leong T, Kon CH. Hypertensive choroidopathy: recognizing clinically significant end-organ damage. *Acta Ophthalmol*. 2008;86:227–228.
49. Adhi M, Brewer E, Waheed NK, Duker JS. Analysis of morphological features and vascular layers of choroid in diabetic retinopathy using spectral-domain optical coherence tomography. *JAMA Ophthalmol*. 2013;131:1267–1274.
50. Vujosevic S, Martini F, Cavarzeran F, Pilotto E, Midena E. Macular and peripapillary choroidal thickness in diabetic patients. *Retina*. 2012;32:1781–1790.
51. Kase S, Endo H, Yokoi M, et al. Choroidal thickness in diabetic retinopathy in relation to long-term systemic treatments for diabetes mellitus. *Eur J Ophthalmol*. 2016;26:158–162.
52. Choi W, Waheed NK, Moult EM, et al. Ultrahigh speed swept source optical coherence tomography angiography of retinal and choriocapillaris alterations in diabetic patients with and without retinopathy. *Retina*. 2017;37:11–21.
53. Kim JT, Lee DH, Joe SG, Kim JG, Yoon YH. Changes in choroidal thickness in relation to the severity of retinopathy and macular edema in type 2 diabetic patients. *Invest Ophthalmol Vis Sci*. 2013;54:3378–3384.
54. Okamoto M, Matsuura T, Ogata N. Effects of panretinal photocoagulation on choroidal thickness and choroidal blood flow in patients with severe nonproliferative diabetic retinopathy. *Retina*. 2016;36:805–811.
55. Zhang Z, Meng X, Wu Z, et al. Changes in choroidal thickness after panretinal photocoagulation for diabetic retinopathy: a 12-week longitudinal study. *Invest Ophthalmol Vis Sci*. 2015;56:2631–2638.
56. Ornek N, Ornek K, Ogurel T, Timur IE. Effect of intravitreal bevacizumab on macular and peripapillary choroidal thickness in injected and fellow eyes of patients with diabetic macular oedema. *Acta Ophthalmol*. 2015;93:e684–e686.
57. Sonoda S, Sakamoto T, Yamashita T, et al. Effect of intravitreal triamcinolone acetonide or bevacizumab on choroidal thickness in eyes with diabetic macular edema. *Invest Ophthalmol Vis Sci*. 2014;55:3979–3985.
58. Ahn SJ, Woo SJ, Park KH, Jung C, Hong JH, Han MK. Retinal and choroidal changes and visual outcome in central retinal artery occlusion: an optical coherence tomography study. *Am J Ophthalmol*. 2015;159:667–676.
59. Chiang JP, Trzupek K. The current status of molecular diagnosis of inherited retinal dystrophies. *Curr Opin Ophthalmol*. 2015;26:346–351.
60. Lambertus S, van Huet RA, Bax NM, et al. Early-onset stargardt disease: phenotypic and genotypic characteristics. *Ophthalmology*. 2015;122:335–344.
61. Giani A, Pellegrini M, Carini E, Peroglio Deiro A, Bottoni F, Staurenghi G. The dark atrophy with indocyanine green angiography in Stargardt disease. *Invest Ophthalmol Vis Sci*. 2012;53:3999–4004.
62. Pellegrini M, Acquistapace A, Oldani M, et al. Dark atrophy: an optical coherence tomography angiography study. *Ophthalmology*. 2016;123:1879–1886.
63. Robson AG, Tufail A, Fitzke F, et al. Serial imaging and structure-function correlates of high-density rings of fundus autofluorescence in retinitis pigmentosa. *Retina*. 2011;31:1670–1679.
64. Guduru A, Al-Sheikh M, Gupta A, Ali H, Jalali S, Chhablani J. Quantitative assessment of the choriocapillaris in patients with retinitis pigmentosa and in healthy individuals using OCT angiography. *Ophthalmic Surg Lasers Imaging Retina*. 2018;49:e122–e128.
65. Tan R, Agrawal R, Taduru S, Gupta A, Vupparaboina K, Chhablani J. Choroidal vascularity index in retinitis pigmentosa: an OCT study. *Ophthalmic Surg Lasers Imaging Retina*. 2018;49:191–197.
66. Schmitz-Valckenberg S, Holz FG, Bird AC, Spaide RF. Fundus autofluorescence imaging: review and perspectives. *Retina*. 2008;28:385–409.
67. Foote KG, Roorda A, Duncan JL. Multimodal imaging in choroideremia. *Adv Exp Med Biol*. 2019;1185:139–143.
68. Lutty GA, McLeod DS, Bhutto IA, Edwards MM, Seddon JM. Choriocapillaris dropout in early age-related macular degeneration. *Exp Eye Res*. 2020;192:107939.
69. Seddon JM, McLeod DS, Bhutto IA, et al. Histopathological insights into choroidal vascular loss in clinically documented cases of age-related macular degeneration. *JAMA Ophthalmol*. 2016;134:1272–1280.
70. Lee JY, Lee DH, Lee JY, Yoon YH. Correlation between subfoveal choroidal thickness and the severity or progression of nonexudative age-related macular degeneration. *Invest Ophthalmol Vis Sci*. 2013;54:7812–7818.
71. Lee J, Kim M, Lee CS, et al. Drusen subtypes and choroidal characteristics in asian eyes with typical neovascular age-related macular degeneration. *Retina*. 2020;40:490–498.
72. Spaide RF. Disease expression in nonexudative age-related macular degeneration varies with choroidal thickness. *Retina*. 2018;38:708–716.
73. Lai K, Zhou L, Zhong X, et al. Morphological difference of choroidal vasculature between polypoidal choroidal vasculopathy and neovascular AMD on OCT: from the perspective of pachychoroid. *Ophthalmic Surg Lasers Imaging Retina*. 2018;49:e114–e121.
74. Koh LHL, Agrawal R, Khandelwal N, Sai Charan L, Chhablani J. Choroidal vascular changes in age-related macular degeneration. *Acta Ophthalmol*. 2017;95:e597–e601.
75. Invernizzi A, Benatti E, Cozzi M, et al. Choroidal structural changes correlate with neovascular activity in neovascular age related macular degeneration. *Invest Ophthalmol Vis Sci*. 2018;59:3836–3841.
76. Baek J, Lee JH, Jung BJ, et al. Morphologic features of large choroidal vessel layer: age-related macular degeneration, polypoidal choroidal vasculopathy, and central serous chorioretinopathy. *Graefes Arch Clin Exp Ophthalmol*. 2018;256:2309–2317.
77. Braun PX, Mehta N, Gendelman I, et al. Global analysis of macular choriocapillaris perfusion in dry age-related macular degeneration using swept-source optical coherence tomography angiography. *Invest Ophthalmol Vis Sci*. 2019;60:4985–4990.
78. Borrelli E, Shi Y, Uji A, et al. Topographic analysis of the choriocapillaris in intermediate age-related macular degeneration. *Am J Ophthalmol*. 2018;196:34–43.
79. Thulliez M, Zhang Q, Shi Y, et al. Correlations between choriocapillaris flow deficits around geographic atrophy and enlargement rates based on swept-source OCT imaging. *Ophthalmol Retina*. 2019;3:478–488.
80. Nassisi M, Tepelus T, Nittala MG, Sadda SR. Choriocapillaris flow impairment predicts the development and enlargement of drusen. *Graefes Arch Clin Exp Ophthalmol*. 2019;257:2079–2085.

81. Kim M, Choi SY, Park YH. Analysis of choroidal and central foveal thicknesses in acute anterior uveitis by enhanced-depth imaging optical coherence tomography. *BMC Ophthalmol*. 2017;17:225.

82. Gabriel M, Kruger R, Shams-Mafi F, et al. Mapping retinal and choroidal thickness in unilateral nongranulomatous acute anterior uveitis using three-dimensional 1060-nm optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2017;58:4778–4783.

83. O'Keefe GA, Rao NA. Vogt-Koyanagi-Harada disease. *Surv Ophthalmol*. 2017;62:1–25.

84. Invernizzi A, Cozzi M, Staurenghi G. Optical coherence tomography and optical coherence tomography angiography in uveitis: a review. *Clin Exp Ophthalmol*. 2019;47:357–31.

85. Herbort CP, Mantovani A, Papadia M. Use of indocyanine green angiography in uveitis. *Int Ophthalmol Clin*. 2012;52:13–31.

86. Invernizzi A, Agarwal A, Cozzi M, Viola F, Nguyen QD, Staurenghi G. Enhanced depth imaging optical coherence tomography features in areas of choriocapillaris hypoperfusion. *Retina*. 2016;36:2013–2021.

87. Invernizzi A, Mapelli C, Viola F, et al. Choroidal granulomas visualized by enhanced depth imaging optical coherence tomography. *Retina*. 2015;35:525–531.

88. Nakayama M, Keino H, Okada AA, et al. Enhanced depth imaging optical coherence tomography of the choroid in Vogt-Koyanagi-Harada disease. *Retina*. 2012;32:2061–2069.

89. Invernizzi A, Agarwal AK, Ravera V, et al. Comparing optical coherence tomography findings in different aetiologies of infectious necrotising retinitis. *Br J Ophthalmol*. 2018;102:433–437.

90. Agrawal R, Salman M, Tan KA, et al. Choroidal Vascularity Index (CVI)—a novel optical coherence tomography parameter for monitoring patients with panuveitis? *PLoS One*. 2016;11:e0146344.

91. Mehta H, Sim DA, Keane PA, et al. Structural changes of the choroid in sarcoid- and tuberculosis-related granulomatous uveitis. *Eye (Lond)*. 2015;29:1060–1068.

92. Agarwal A, Invernizzi A, Singh RB, et al. An update on inflammatory choroidal neovascularization: epidemiology, multimodal imaging, and management. *J Ophthalmic Inflamm Infect*. 2018;8:13.

93. Pichi F, Invernizzi A, Tucker WR, Munk MR. Optical coherence tomography diagnostic signs in posterior uveitis. *Prog Retin Eye Res*. 2020;75:100797.

94. Mueller AJ, Freeman WR, Folberg R, et al. Evaluation of microvascularization pattern visibility in human choroidal melanomas: comparison of confocal fluorescein with indocyanine green angiography. *Graefes Arch Clin Exp Ophthalmol*. 1999;237:448–456.

95. Mueller AJ, Bartsch DU, Folberg R, et al. Imaging the microvasculature of choroidal melanomas with confocal indocyanine green scanning laser ophthalmoscopy. *Arch Ophthalmol*. 1998;116:31–39.

96. Schaller UC, Mueller AJ, Bartsch DU, Freeman WR, Kampik A. Choroidal melanoma microcirculation with confocal indocyanine green angiography before and 1 year after radiation brachytherapy. *Retina*. 2000;20:627–632.

97. Rummelt V, Folberg R, Rummelt C, et al. Microcirculation architecture of melanocytic nevi and malignant melanomas of the ciliary body and choroid. A comparative histopathologic and ultrastructural study. *Ophthalmology*. 1994;101:718–727.

98. Shields CL, Shields JA, De Potter P. Patterns of indocyanine green videoangiography of choroidal tumours. *Br J Ophthalmol*. 1995;79:237–245.

99. Shields CL, Pellegrini M, Ferenczy SR, Shields JA. Enhanced depth imaging optical coherence tomography of intraocular tumors: from placid to seasick to rock and rolling topography–the 2013 Francesco Orzalesi Lecture. *Retina*. 2014;34:1495–1512.

100. Pellegrini M, Invernizzi A, Giani A, Staurenghi G. Enhanced depth imaging optical coherence tomography features of choroidal osteoma. *Retina*. 2014;34:958–963.

101. Pellegrini M, Corvi F, Say EAT, Shields CL, Staurenghi G. Optical coherence tomography angiography features of choroidal neovascularization associated with choroidal nevus. *Retina*. 2018;38:1338–1346.

102. Hirano T, Kakihara S, Toriyama Y, Nittala MG, Murata T, Sadda S. Wide-field en face swept-source optical coherence tomography angiography using extended field imaging in diabetic retinopathy. *Br J Ophthalmol*. 2018;102:1199–1203.