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the Ophthalmologist

In the Eye of the Storm

The new role for ophthalmologists in tackling chronic diseases

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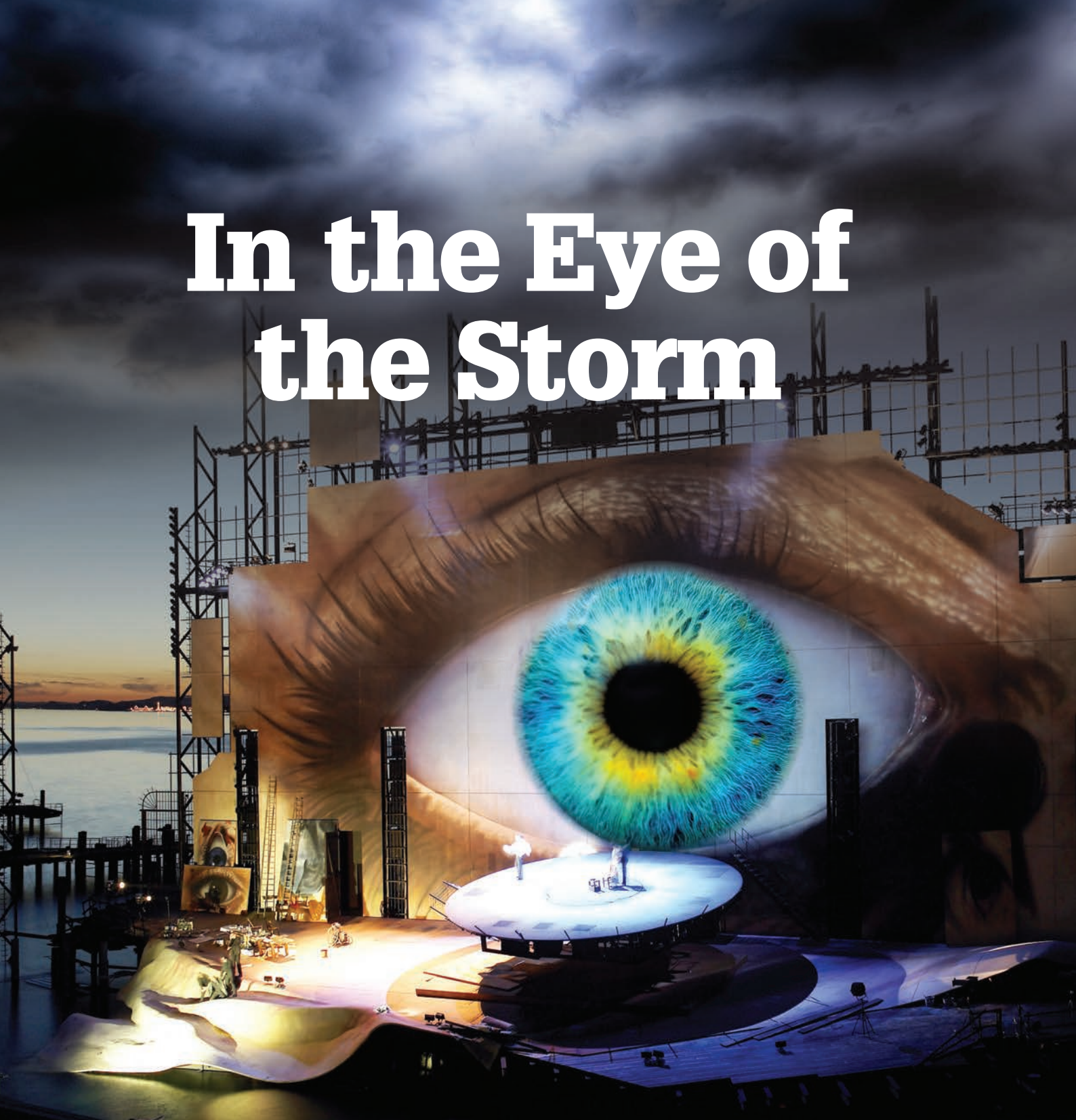


The global obesity pandemic is causing a chronic cardiovascular and diabetic disease crisis. Advances in retinal microvascular imaging offer hope in tackling these problems… but are ophthalmologists ready for a prominent role in the fight against systemic disease?

By Mark Zacharria

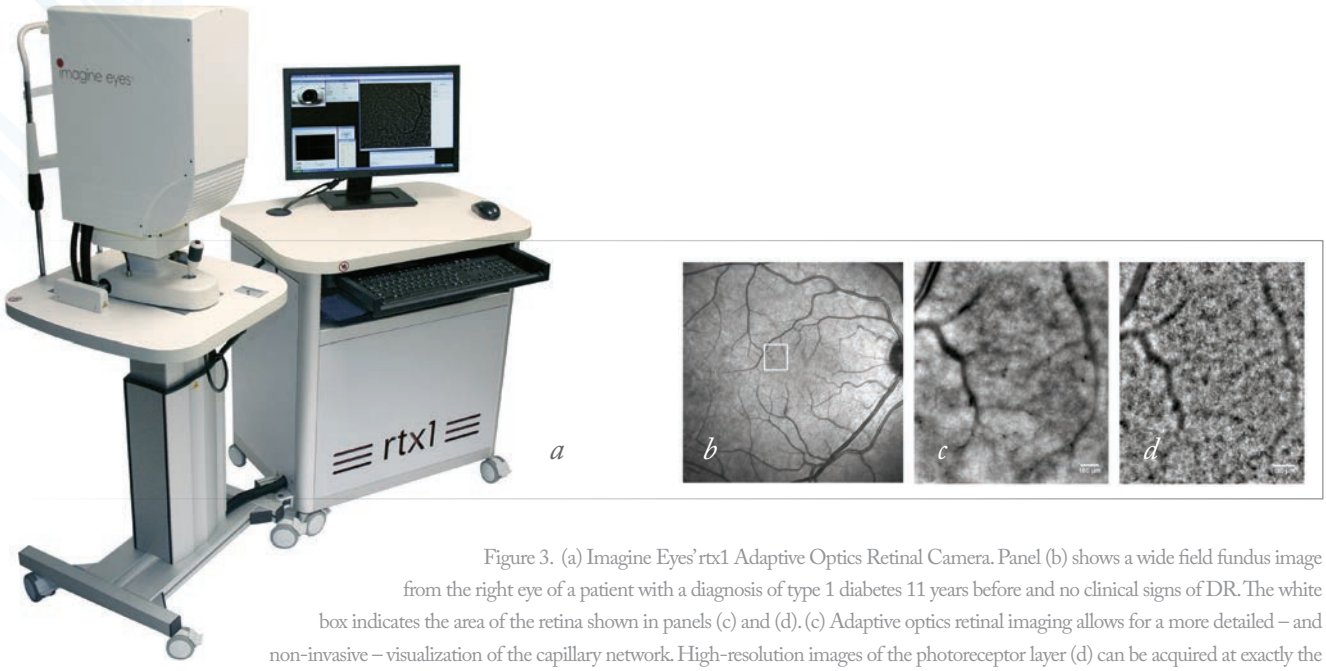


Figure 3. (a) Imagine Eyes' rtx1 Adaptive Optics Retinal Camera. Panel (b) shows a wide field fundus image from the right eye of a patient with a diagnosis of type 1 diabetes 11 years before and no clinical signs of DR. The white box indicates the area of the retina shown in panels (c) and (d). (c) Adaptive optics retinal imaging allows for a more detailed – and non-invasive – visualization of the capillary network. High-resolution images of the photoreceptor layer (d) can be acquired at exactly the same location of the overlying structures of the inner retina with AO ophthalmoscopy. (c) and (d) Scale bars represent 100 µm.

Adaptive optics imaging in non-prolific diabetic retinopathy

A conversation with Marco Lombardo

Can use describe your study?

We recruited eight patients with a diagnosis of type 1 diabetes and NPDR with no macular edema. Eight age-matched healthy subjects were recruited as controls. Patients and controls were submitted both to AO retinal imaging and conventional imaging using SLO and color fundus retinography (CFR). Using AO, the focal plane was adjusted to acquire images of the retinal capillaries of the inner nuclear layer in order to maximize the sharpness of vascular images. The capillary network of the inner nuclear layer was imaged 210-230 µm anteriorly to the photoreceptor layer. We used a semi-automated procedure to measure the retinal capillary lumen caliber in two regions of interest located close to the border of the foveal avascular zone (FAZ).

What device did you use?

We used was an rtx1 AO fundus camera commercialized from Imagine Eyes (see Figure 3a). The device uses IR reflectance to provide 4° x 4° images at a resolution of 250 lppmm, which literature reports as ±2 µm in transverse resolution that can be acquired at different depths depending on the structures of interest.

Did AO reveal information that the other techniques did not?

Retinal capillaries were not resolved by SLO or CFR in any eye. In AO images, the retinal capillaries appeared as faint vessel segments intersecting each other and forming a network of arterioles and venules both in NPDR and control eyes (see Figure 3b-d). The average lumen of retinal capillaries was statistically significantly narrower in NPDR eyes (6.27 ± 1.63 µm) than controls (7.31 ± 1.59 µm; (7)). On average, the retinal capillary lumen was 15 percent narrower in NPDR than in control eyes. Microaneurysms could equally be noninvasively observed in NPDR cases.

How might this information be used in diabetes care?

The detection of pre-clinical abnormalities of retinal microcirculation may represent the real advantage of AO retinal imaging in the management of patients with diabetes. The capability to resolve retinal capillaries has been shown for all existing AO ophthalmic imaging modalities, namely AO-flood, AO-SLO and AO-OCT. Fluorescein angiography (FA) has been implemented in an AO-SLO, providing further in-depth investigation of the capillary network.

The combined longitudinal assessment of the capillary density, capillary lumen caliber and the FAZ area by AO retinal imaging might provide valuable information on DR onset and progression. AO-SLO and AO-OCT can also characterize the

blood flow in retinal capillaries, and a significant reduction in the capillary blood velocity in patients with diabetes has been shown as one of the earliest changes in DR.

AO retinal imaging promises early detection of DR and monitoring of the progression of the disease with micrometric resolution. Moreover, AO can be used to evaluate the efficacy of new treatment options at a level that was heretofore unavailable.

What needs to be accomplished for AO to become a part of everyday clinical practice?

Several factors must be resolved, including the development of easy-to-use software interfaces, fast image processing approaches and reliable analysis software. Improvements in AO-SLO have already enabled it to obtain images of retinal capillaries with incredible resolution – comparable to a histological section.

While there are certainly challenges to the clinical application of AO retinal imaging, the rapid growth in the past few years suggests these will soon be overcome. A number of multi-disciplinary collaborations between clinical and non-clinical researchers have been initiated to resolve the specific needs of clinical AO imaging.

Marco Lombardo is Senior Researcher at the Clinical Trial Research Center of the IRCCS Fondazione G.B. Bietti in Rome, Italy. He is responsible for the study protocol on AO imaging in patients and other projects related to the application of innovative biotechnologies to ophthalmology, including nanotechnology and regenerative medicine. These projects are a collaboration with CNR-IPCF, under the supervision of Giuseppe Lombardo.

High-resolution AO-SLO fluorescein angiography

A conversation with Richard Rosen

What are the key differences between conventional FA and AO FA?

AO-SLO FA permits enhanced resolution, allowing finely detailed imaging of multiple layers of capillaries, microaneurysms, microvascular anomalies, capillary dropout and microleakage.

What device do you use?

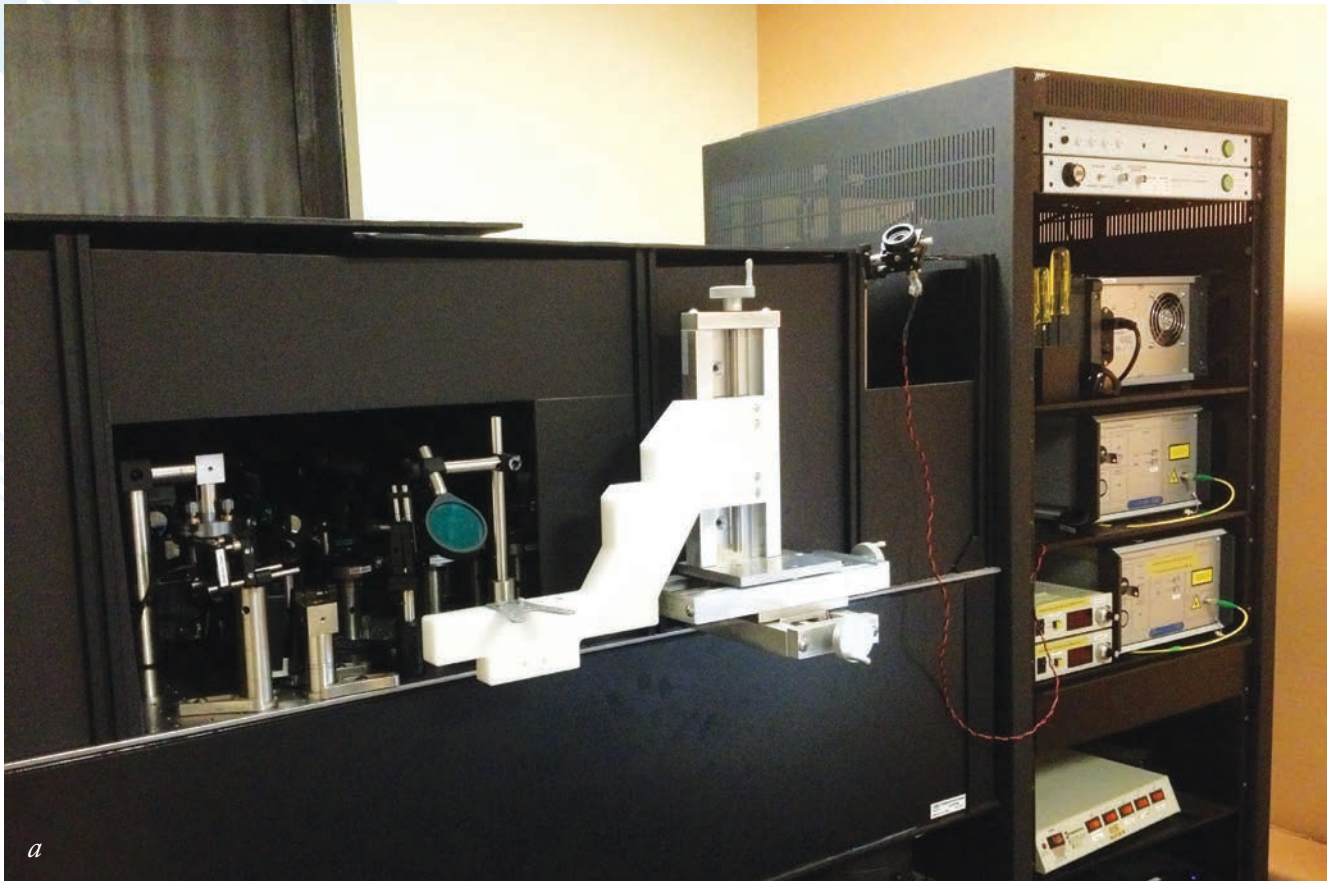
The AO-SLO used in our lab is a replica of the one developed by Dubra and Sulai at the Medical College of Wisconsin (see Figure 4a), with the visible channel modified for fluorescein angiography (FA) imaging (1). During AO-SLO imaging, simultaneous reflectance (790 nm) and fluorescence (488 nm) image sequences are acquired and registered.

Does the system have any particular strengths?

While conventional FA can identify vessels based on their fluorescence alone, we are able to simultaneously image both the intraluminal space with AO-SLO FA (visible on the 488nm channel; see Figure 4b) and the vessel wall with AO-SLO reflectance (visible on the 790 nm channel). This allows us to delve into the delicate relationship between wall changes and luminal infiltrates. Differentiating between perfused and non-perfused vessels is also possible by comparison of AO-SLO FA images (functional perfusion map) and AOSLO reflectance images (structural).

How did you work around any potential glitches?

The ability to gain early-phase information and monitor transit time of conventional FA is sacrificed in AO-SLO FA due to the more time-consuming technique of successive acquisition of individual small fields of 1.75°, which are then tiled together into larger montages offline. To accommodate the extended imaging sessions, oral fluorescein was chosen to provide a more consistent signal for a longer time than with an intravenous bolus. This is easier to administer and improves the safety and comfort of the procedure, when compared with intravenous administration.



How will your research improve diagnosis and management of vascular disease?

Improved resolution of fine capillary structures along with the ability to look at the photoreceptor mosaic and nerve fiber layer will lead to a better understanding of the anatomic basis of retinal disease. We hope that, with this advance in resolution to image the microvasculature, we will discover more accurate explanations for the pathogenesis of vasculopathy and visual malfunction. We hope this will lead to more targeted approaches to treatment and prevention of progressive deterioration.

The ability to study microvascular disease in vivo will allow us to study microscopic changes dynamically over extended periods of time, giving an advantage over conventional FA and traditional histology. Following these changes in patients undergoing various treatment regimens will help us to better understand some of the underlying processes taking place

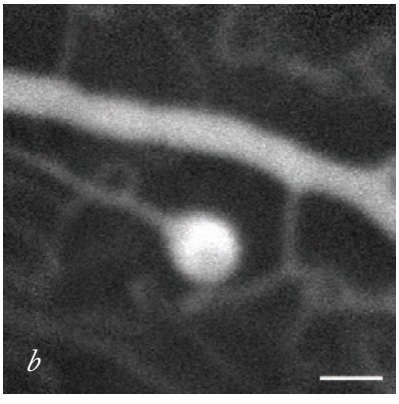


Figure 4 (a) AOSLO system built by the Advanced Ocular Imaging Program at the Medical College of Wisconsin and installed at the New York Eye and Ear Infirmary. (b) AOSLO FA image from NY Eye & Ear acquired at ~8° superior on retina from a 48 year old female with NPDR. Scale bar = 50 µm.

within the macroscopic picture of disease progression and clinical response to specific therapies.

We have already witnessed the impact of anti-VEGF therapy on capillary reperfusion and microaneurysm resolution and are in the process of studying the impact of laser photocoagulation and the fine structure of response of the retinal tissue. With this ability to study capillary dynamics clinically, we plan to investigate the changes that manifest as glaucoma, progressive vascular retinopathies, toxic maculopathies and episodic inflammatory diseases.

Studying vascular remodeling after branch artery and vein occlusions may help us to better understand stroke recovery whereas studying the microvascular events which accompany capillary dropout and partial revascularization may help us understand similar processes in renal and cardiac disease. Understanding the various paths to progression of diabetic retinopathy may lead to more rational approaches to management.

When will AO become part of everyday clinical practice, and what needs to be accomplished for that to happen?

AO will be ready for real-time once image processing improves, the speed of the study shortens the test to a length congruous with clinical encounters, and the cost and design of hardware allow the construction of a compact, semi-automatic instrument that can deliver an answer in a few minutes. Currently, the AO mirrors are hand-made and very expensive, which drives the price of the systems to levels that are not commercially sustainable. Due to current technical limitations of AO-SLO FA, we are only able to visualize 15° radius around fovea (1.75° at a time) whereas wide-field conventional FA can image as much as 180° in a single take. Clinical implementation will also demand an application, which depends on this level of imaging for monitoring, the so-called "killer app" – the way OCT is a surrogate VEGF meter and is critical for managing macular edema.

Richard Rosen leads a team at the New York Eye and Ear Infirmary that includes Michael Dubow, Alexander Pinhas, Nishit Shah, Toco Chui, Alexander Gan, and Moataz Razeen. The group receives funding from Marrus Family Foundation, Bendheim-Lowenstein Family Foundation, Wise Family Foundation, Chairman's Research Fund of the NYEEI and NIH.