A
tofluorescence photography of the eye provides important diagnostic information about diseases that cause accumulation of fluorophores in the retinal pigment epithelium. Central serous chorioretinopathy, vitelliform macular dystrophy, acute exudative polymorphous vitelliform maculopathy, choroidal tumors, and vitreomacular traction syndrome are among the diseases that cause accumulation of fluorophores.

ADVANTAGES OF AUTOFLUORESCENCE IMAGING

Figures 1-4 show examples of what can be seen with fundus autofluorescence compared with color fundus photography. There are many diseases that can affect the retinal pigment epithelium (RPE), and in every case autofluorescence can be a beneficial diagnostic aid. Whether looking at geographic atrophy where the RPE dies and atrophy of the overlying retina and underlying choriocapillaris occurs, or an inherited disease, such as retinitis pigmentosa, which causes injury or death of the RPE, the best way to image the RPE is with autofluorescence.

Other diseases, such as vitelliform macular dystrophy, cause an accumulation of highly autofluorescent material under the retina. A comprehensive theory about these diseases was developed through the use of optical coherence tomography and autofluorescence imaging, identifying a separation of the retina from the underlying RPE by subretinal fluid. The RPE ordinarily clears spent photoreceptor outer segments through a process of phagocytosis. If the retina is separated from the underlying retinal pigment epithelium, the outer segments are not cleared properly and accumulate.

The RPE plays an important role in supporting retinal function by recycling visual pigment, absorbing excess light and maintaining proper fluid and electrolyte levels in the subretinal space, and phagocytosing spent outer segments of the photoreceptors, among other things. By recording autofluorescence information, we can gain critical knowledge about the health of the retina and RPE.

AN ACCESSIBLE IMAGING METHOD

Autofluorescence imaging is most commonly obtained with scanning laser ophthalmoscopic (SLO) systems, but...
they can also be effectively accomplished with fundus photography if the camera is outfitted with special filters to offset interference from the crystalline lens. Crystalline lens autofluorescence causes fluorescein angiograms to look washed out in the eyes with nuclear sclerosis. The autofluorescence of the lens adds to the fluorescence coming from the fundus to produce an image with low contrast. To produce useful autofluorescence images, we must either reject or bypass the fluorescence of the lens.

A new generation of filters developed by Richard F. Spaide, MD, and marketed exclusively with the IMAGEnet (Topcon, Paramus, NJ) fundus photography systems, are designed to foster improved visualization of fundus autofluorescence. SLO systems are cost-prohibitive for some practices, particularly in comparison with fundus cameras, which are relatively affordable and pervasive. The ability to achieve good contrast using a fundus camera to perform autofluorescence imaging will make this diagnostic option more readily available.

**MATCHED INTERFERENCE FILTERS**

The new excitation and barrier filters, termed “matched interference filters” have been optimized to allow better visualization of fundus autofluorescence. The excitation filter was selected to mimic the function of a green monochromatic filter; the green light is absorbed by blood and enables improved contrast. The fluorophores in the eye, particularly lipofuscin, have the potential to absorb excitation light in a broad range from 300 nm to greater than 700 nm; however, many ranges of these wavelengths occupy regions used for other types of ocular imaging. One design goal was to replace the green filter of a typical fundus camera with the excitation filter.
for fundus autofluorescence. For the excitation filter to appear green, the bandpass region had to occupy the region at about 550 nm. The wavelengths of the excitation filter were selected to be 535 nm to 585 nm, which is not within the absorption curve of fluorescein, so that autofluorescence photography can be performed on a patient who has had fluorescein angiography. The excitation wavelengths also are not absorbed by macular pigments. The barrier filter is designed to allow passage of wavelengths starting at about 615 nm, and was selected to have a bandpass region that was 100 nm wide.

By moving the location of the wavelengths for the excitation and barrier filters and improving their optical transmission characteristics, the new filter sets are 20 times more efficient than the previous generation. They require 40% less exposure and produce a more vivid, detailed image of the fluorophores. Image contrast is significantly improved, while reducing noise in the images. Prior to the availability of these new filters, a photographer would have to set the illumination at 300 watt-seconds, which is the maximum, and set the gain at 22 to 36, which is also the maximum. With the new filters, 100 or 150 watt-seconds and a gain of 12 are used. The more gain, the more noise—analogous to turning up a radio to hear a weak radio station.

**ADVANTAGES OVER SLO**

The matched interference filters do not stimulate fluorescein or indocyanine green, so autofluorescence photographs can be taken before or after angiography; SLO devices do not have this capability. There are several differences between the images taken with an SLO and a fundus camera. With SLO imaging, noise from the laser contributes to image noise. Because of the noise and the weak autofluorescence signal, 15 images are taken, and typically the best nine are averaged. Also, the wavelengths used in SLO imaging are absorbed by nuclear sclerosis and by macular pigment. Areas of decreased autofluorescence can appear in the central macula from macular pigment and not necessarily from macular disease.

**SUMMARY**

By imaging the autofluorescence, we can not only obtain anatomic information from the lipofuscin that is in the retinal pigment epithelial cells, but we can also make inferences about functional aspects of these cells.

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