AMD National Knowledge Week – June 2007

What is Autofluorescence (AF) Imaging?

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Autofluorescence imaging (AF), using a confocal Laser Scanning Ophthalmoscope, is a recent technique that has been developed to allow 'in vivo' imaging of the RPE of the retina.[1] It assesses the accumulation of the lipid aggregate lipofuscin, with age, which is believed to be toxic to cells and may play a role in the pathogenesis of macular degeneration.[2,3]

Substantial research now suggests that lipofuscin is derived from residual bodies that result from incompletely digested photoreceptor outer segments that are phagocytosed by the RPE.[3,4] The levels of lipofuscin increase with age and in some disease states.

AF imaging has now become an important diagnostic tool in various retinal diseases but has yet to establish a role in AMD diagnosis.[5,6]

How does AF imaging work?

It was Delori who discovered that excitation of lipofuscin with short wavelength light resulted in emission of light, with a maximum wavelength of 620-630 nm, which could be detected both in vitro and in vivo.[2,7]. With appropriate barrier filters, a confocal scanning laser ophthalmoscope (SLO) could be used to record fundus AF within the RPE.

To image fundus AF, three machine types have been described, the Heidelberg Retina Angiograph (HRA), the Rodenstock cSLO and the Zeiss prototype SM 30-4024.[8] The field of view is set to between 30˚ and 40˚ depending on the machine used and in all, the confocal detection unit uses a pinhole aperture to suppress light originating from outside the focal plane. This enhanced the image contrast. To produce an image, an argon blue laser (488nm) is used for excitation and emitted light is detected above a barrier filter which is inserted in front of the detector. The cut off wavelength of the filter varies from 500 to 521nm, depending on the type of machine.[1] Several images are then taken, aligned and an average image calculated using image analysis software .[1,9] The brightest images have been obtained with the HRA and Zeiss prototype.[8] Media opacities limit the quality of the image in all machines.

Interpretation of the image

Fundus autofluorescence generally appears grey and the intensity increases (i.e becomes more white) with age. The highest intensity is at the posterior pole (between 5˚and 15˚ from the fovea), it then dips at the fovea and reduces again toward the periphery.[10] The optic disc and retinal blood vessels are shown as black structures and remain consistent regardless of age (see Figure 1). This indicates that the image is not caused by reflected light as the disc is the most reflectant feature of the fundus.[11] The distribution of lipofuscin generally matches that of rods and reflects the pattern of age- related loss of rod photoreceptors.[10] Areas of melanolipofuscin in pigment clumps produce increased AF and appear white. In disease states, further increased AF (seen as white or light grey areas) is believed to represent lipofuscin-laden RPE cells which are a marker for areas at increased risk of atrophy.[6,9] As atrophy occurs, the autofluorescence becomes dark and represents removal of atrophic RPE cells.

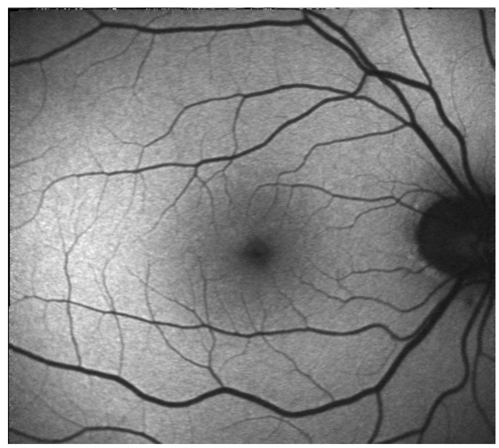

Figure 1:

Colour fundus photograph (right) and autofluorescence image (left) of normal fundus Courtesy of Miss S Mann's MD thesis collection

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