

## Measuring macular pigment levels: An in-practice procedure?

The macula lutea derives part of its name from its colour. 'Lutea' is the characteristic yellow colouration the macula exhibits due to the presence of macular pigment. Macular pigment is an oleaginous substance, found in the axons of the photoreceptors in the macular area. It is concentrated mainly within the foveola and its levels drop off to a minimum by about 6 degrees of eccentricity. It is comprised of lutein and zeaxanthin, which are isomers of xanthophyll, a commonly occurring carotenoid. Carotenoids are pigments that occur naturally such as carotene which is found in carrots. The ratio of lutein to zeaxanthin varies across the fovea. At the foveola, zeaxanthin predominates, but its levels reduce more quickly than that of lutein, allowing this carotenoid to prevail in the perifoveal area.

There is increasing evidence to suggest that macular pigment helps protect against age-related macular disease. The terms age-related maculopathy (ARM) and age-related macular degeneration (AMD), were defined in 1995 by the International ARM Epidemiological Study Group and a uniform grading system put forward.<sup>1</sup> ARM is the term used to encompass all signs of age-related changes at the macula. It is characterised by any of the following: soft drusen and areas of hypo/hyperpigmentation associated with drusen. In advanced stages of ARM the term AMD is used. This covers both wet and dry types. Choroidal neovascularisation, subretinal haemorrhage and retinal pigment epithelium detachment are the defining criteria for wet AMD while dry AMD refers to geographic atrophy.

It is thought that the part that macular pigment plays in the eye is twofold. It was proposed by Khachik, et al, in 1997 that the retinal carotenoids have antioxidant properties.<sup>2</sup> It would seem that they act as free radical scavengers, preventing these by-products of cellular metabolism from reacting harmfully with cell constituents, causing a decrease in the cell's function or even cell death. The second role that macular pigment plays is as a filter. This has a potential visual acuity benefit as chromatic aberrations are reduced by the selective absorption of light at

the blue end of the visible spectrum. More importantly, this absorption may protect the macula from the damaging effects of shorter wavelength light (these effects include the photodynamic production of free radicals).

Macular pigment cannot be manufactured by the body. In humans, as in all primates, its origin is entirely alimentary. Sommerburg, et al, aimed to determine which foodstuffs might be useful as dietary supplements<sup>3</sup>. It is interesting to note that the dark green leafy vegetables, such as Brussels sprouts, spinach, broccoli and lettuce are a good source of lutein but contain very little, if any, zeaxanthin.

It would appear that naturally yellow/orange coloured foodstuffs (with the exception of pumpkins and carrots) contain more zeaxanthin than their green counterparts. Landrum et al and Hammond et al have demonstrated that alterations to diet can have an effect on the levels of macular pigment.<sup>4,5</sup>

The Macular Pigment Screener has been developed at UMIST to measure the macular pigment optical density. Tinsley Ophthalmic Instruments are supporting the project and the research was funded by The Department of Health. The aim of the project is to determine whether patients in the early stages of ARM have reduced levels of macular pigment. If this is established, then this would be further evidence that low macular pigment optical density can be considered as a risk factor for ARM.

### How the instrument works

The instrument is relatively compact (maximum dimensions 51cm deep x 44cm wide x 42cm high) and portable, weighing about 5 kg. The viewing distance is 20cm and the use of an appropriate near vision correction is advised. The rectangular viewing window is 8.5cm x 7.5 cm in size and a forehead rest aids observer comfort. Its body has been developed from that of a Henson Pro Field Screener. The observer should be dark adapted for two minutes prior to the screening being carried out and it would seem sensible to use this time to explain the test to him/her. The instrument is used monocularly and the fellow eye should be occluded throughout

the process. The view the observer has of the targets is a so-called 'free-view' as opposed to a Maxwellian view. A Maxwellian view, using an external Maxwellian lens to image the source in the centre of the observer's pupil, has an advantage in that any variation in pupil size does not affect the readings. However, alignment and calibration problems arise with this method and in a clinical setting a free-viewing system is easier and more comfortable for observers to cope with.

The instrument works on the principle of heterochromatic flicker photometry. This principle was developed in the early 1900s by Frederick Ives.

Two differently-coloured stimuli are presented to the observer. One of these stimuli will be a reference one and the other a variable one. The reference and the variable test stimuli need to occupy exactly the same location and be exactly the same shape and size. These stimuli are then repeatedly presented one at a time. If the rate of alternation is fairly low, as when each colour is presented twice within each second, the observer will be able to detect both colours and the change between them will be distinct. If the speed of alternation of the stimuli is increased, the two original colours will initially be augmented by a third before starting to fuse to a single colour. This colour will be the colour of the additive mixture of the two component stimuli and it will appear to pulse. This pulsation is the perceived brightness flicker and will disappear at a high frequency of alternation, around 35-40 Hz unless the difference in radiance between the reference stimuli and the test stimuli is great. With slightly lower frequencies, in the range of 15-20 Hz, this perception of flicker can be removed by adjusting the relative intensities of the two stimuli. At the point at which the observer reports zero or minimum flicker the two stimuli are said to have equal luminance. In reality there are two settings which will produce this threshold flicker. The first setting will be when the variable stimulus is slightly greater than the reference and the second will be when the reference stimulus is slightly greater than the variable one. During macular pigment screening, both

values are obtained and the mean of the two is taken.

The variable stimulus used is a blue light with an output spectrum that peaks at 470 nm. This is close to the spectral optical density peak of the macular pigment. The reference stimulus used in this instrument is a green light with a wavelength of 540 nm. Macular pigment does not absorb this wavelength. The stimuli are presented on a matt grey screen, which has an overall background luminance of 23 cd/m<sup>2</sup>. The light sources for the stimuli are light-emitting diodes and the background light is provided by incandescent bulbs. The background luminance and that of the stimuli have been chosen to avoid rod stimulation. In 1866, Max Schultze concluded that a retina without a yellow spot would see more blue light than one with such a spot.<sup>14</sup> The Macular Pigment Screener incorporates this idea in measuring the density of an individual's macular pigment. It is assumed that if the macular pigment is absorbing blue light, then more blue light will be needed to match the luminance of the two stimuli at the foveola (greatest density of macular pigment) than in the parafoveal area (minimal density of macular pigment). It is also assumed that flicker sensitivity of the fovea and the parafovea are the same. A frequency of 25Hz is used as it is above the critical flicker frequency of the rods and the short-wavelength S cones which are distributed more randomly than the medium-wavelength M-cones and the long-wavelength L cones across the area under consideration.

Two fixation points are therefore used, one centrally and a second at a point 5 degrees nasally. This results in two measurements being taken, one centrally and one at a point 5 degrees nasal to the fovea. While the observer is fixating the second point he/she may find that the flickering target seems to disappear. This occurrence is known as the Troxler phenomenon and may also be picked up during use of an Amsler grid. The observer should be encouraged to blink or look away before recommencing the task.

Five sets of measurements are taken for each fixation target and the mean of these measurements is used.

The optical density of the macular pigment can then be calculated from the ratio of the central measurement to that of the parafoveal indicating how much of the blue light is absorbed by the central fovea compared to that of the parafoveal

area.

The average figure of the absorbency unit of macular pigment is 0.2-0.3.

Koh has shown that the repeatability of test results using this instrument is good with a correlation of 0.96615. This would suggest that if a difference in density is found between one measurement and the next, eg after three months of dietary supplements, this could be thought to be a genuine change rather than an artefact.

#### Clinical Implications

This instrument may well find a place alongside other 'screening' equipment such as field screeners and non-contact tonometers. Its portability means that it may be shared between clinics or brought in to monitor the macular pigment levels of specific individuals. The demands made of the observer are not great and are easily learnt, although it must be remembered that, as with all psychophysical testing, some limitations apply. The observer must be perceptive, have reasonably unimpaired vision and understand what is required of him/her.

The author found the process of determining the position of minimal flicker for herself (acting as both observer and operator) and performing the necessary calculations a little onerous. However she could see that by having the encouragement of an experienced operator, the process would become a more straightforward one for the observer. The practice version of this screener will perform the calculations automatically and give an indication of whether the results are normal or abnormal. This will relieve the operator of the need to gather multiple data and calculate individuals' macular pigment optical density. The author feels that these modifications will mean that the instrument would become an attractive one to use in practice, taking approximately 5-7 minutes or so to use for each patient.

As the UK population is an ageing one, more and more of us will experience the debilitating conditions of old age. AMD is the leading cause of blindness in the elderly in the Western world and is on the increase. In 1996, Evans and Wormald found that blind registrations due to AMD had increased in the order of 30-40 per cent in the years between 1950 and 1990.<sup>16</sup> Owen et al estimated that in the UK there are currently 214,000 individuals with a visual impairment caused by AMD and predicted that

this number would rise to 239,000 by the year 2111.<sup>17</sup> As the numbers increase, so does the burden on health and social care provision for the elderly in the UK. It would therefore make sense to look at ways to reduce the incidence of ARM and AMD.

By using the Macular Pigment Screener, perhaps initially in those thought to be at greater risk, an opportunity may then arise for optometrists to be involved in giving advice aimed at preventing the onset of ARM. This advice may range from recommending a balanced healthy diet and eye protection from strong sunlight, to actually prescribing nutritional supplements once a definite link between levels of macular pigment and incidence of ARM has been acknowledged. The role of the optometrist may also be developed to allow monitoring of those with lower than normal levels of macular pigment who are taking steps to increase their retinal levels of lutein and zeaxanthin.

The use of this instrument in practice could therefore have a beneficial effect for individuals, aiming to reduce the incidence or severity of AMD, and a wider impact in reducing the health and social care costs within the community.

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#### Further Reading

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UMIST Times piece. Spinach is good for your eyes.  
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