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What should I note when assessing the retinal vasculature?

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Introduction

The retinal vasculature is unique in that, unlike any other systemic vascular bed, it can be assessed directly and non-invasively *in vivo.* Optometrists routinely evaluate the state of the retina and its microvasculature as part of any normal ophthalmic assessment and if considered carefully, such an evaluation has the potential to provide a unique insight into general vascular health. Advances in imaging technology have enhanced our ability to evaluate and document changes in the retinal vasculature over time and the recent introduction of new objective retinal image analysis techniques has the potential to significantly improve the accuracy and usability of quantifiable retinal vessel parameters such as calibre, arteriovenous ratio (AV ratio) and vessel tortuosity. This article will aim to outline the relevant anatomy and physiology of the retinal vasculature and go on to summarise the current evidence linking alterations in the retinal microvasculature to the occurrence of ocular and systemic vascular disease, with reference to the recent developments in imaging technology that are aiding our ability to make these links now and into the future.

Anatomy and physiology of the retinal vasculature

The retina has the highest oxygen consumption per volume in the body (Yu and Cringle, 2001). To maintain normal retinal function a stable and sufficient blood supply, that can meet the metabolic demands of the retinal tissues whilst not impeding on the precise optics and transparency of the system, is required. To this effect the retina has evolved to have a dual blood supply, with the inner retinal layers (extending from the nerve fibre layer to the inner section of inner nuclear layer) being supplied by the central retinal artery (CRA) and the outer retina layers (extending from outer section of inner nuclear layer to the retinal pigment epithelium) being supplied from below by the choriocapillaries, via the short posterior ciliary arteries (Zhang, 1994). Maintenance of the outer retinal layers and photoreceptors in particular is vital for normal retinal function and as such 65-85% of the total blood supply to the retina comes via the choriocapillaries, with the remaining 20-30% being distributed across the inner retinal layers via the CRA (Lutty et al., 2012).

The CRA, on entering the eye via the optic nerve, divides into 4 main arterial branches, visible on ophthalmoscopy as the superior and inferior, nasal and temporal arcades. These 4 main branches lie within the nerve fibre and ganglion cell layers of the retina and branch down further into arterioles and ultimately into capillaries as they spread across the retina. These retinal capillaries form a complex network that is distributed throughout the inner retinal layers and is commonly divided into three main sections, namely the radial peripapillary capillaries (RPCs), the inner (superficial) capillary bed and the outer (deep) capillary bed (Zhang, 1994). Whilst not directly visible themselves, the localisation of these capillary beds has relevance with regard to the different forms of retinal haemorrhage that can be visualised on fundus examination and their associated causes, as detailed in table 1. The RPCs lie within the inner part of the nerve fibre layer and run along the paths of the major CRA branches. The inner capillaries on the other hand lie in the nerve fibre and ganglion cell layer, underlying the RPCs and form a complex superficial capillary inner plexus. Finally, the outer capillaries lie deeper within the inner plexiform layer and inner nuclear layer and run to the border of the outer plexiform layer (Zhang, 1994). This complex retinal capillary network extends throughout the length of the retina, with the only exception being in the central macula region where a capillary free zone exists parafoveally (Zhang, 1994). Drainage of the retinal circulation is achieved via the central retinal vein (CRV), which travels centrally within the optic nerve, alongside the CRA, before exiting the nerve and ultimately draining into either the superior ophthalmic vein, other intraorbital venous branches or directly into the cavernous sinus (Zhang, 1994). Drainage of the choroidal circulation and hence the outer portion of the retina is via the vorticose veins.

Anatomically, the retinal vascular system can be described as having a traditional end-arterial hierarchy, whereby the main central retinal artery branches into arterioles, which divide to form capillaries, which are then drained by venules that ultimately feed into veins. On leaving the optic disc, normal retinal arteries have a luminal diameter of around 160μm (Lee et al., 2007), with a thin endothelial basement membrane and thick wall of smooth muscle. In contrast, normal retinal veins have a much wider luminal diameter of around 230μm (Lee et al., 2007) at this same point, with only a thin layer of smooth muscle. The retinal capillaries are much smaller, with a diameter of between 3.5 to 6μm and are composed of a single continuous layer of endothelial cells which are surrounded by pericytes in a 1:1 ratio (Lutty et al., 2012).

It is at the level of the retinal capillaries that the exchange of oxygen and nutrients and the removal of waste products to and from the surrounding retinal tissue, occurs. In order to ensure the integrity of the retinal tissue is maintained, tight junctions exist between the

retinal capillary endothelial cells, forming an inner blood-retinal barrier (BRB) similar to the blood-brain barrier of the central nervous system. This inner BRB, in conjunction with the RPE which forms an outer BRB, is an important feature of the retinal vasculature as it helps to prevent the passage of all but essential metabolites from the bloodstream into the retinal tissues. Indeed, a breakdown of the BRB, for example under conditions of hypoxia or ischaemia, is the originating factor in the development of a number of classic signs of retinal pathology commonly observed clinically, as will be discussed later in this article.

Finally, within the architecture of the retinal vasculature many arteriovenous crossings exist. At the majority of these crossing points, the retinal artery is situated anterior to the vein and both vessels are enclosed by a common basement membrane. The close approximation of these vessels at these crossing points makes these locations vulnerable sites for vascular occlusion, for example as a result of occlusion of the underlying vein by an overlying arteriosclerotic retinal artery (Lutty et al., 2012).

From a physiological viewpoint, retinal circulation is characterised by a low level of flow and high level of oxygen extraction (Delaey and van de Voorde, 2000). It is autoregulated, allowing vessel diameter and hence blood supply to be altered according to changes in metabolic demand, blood pressure, oxygen and carbon dioxide levels, however, unlike many other systemic vascular beds it receives no autonomic innervation (Delaey and van de Voorde, 2000). This is in comparison to the choroidal circulation which is characterised by very high flow and low oxygen extraction and has the greatest density of autonomic innervations in the body, but little autoregulatory capacity. These two vascular supply systems work together to maintain normal retinal function.

Why is it important to assess the retinal vasculature?

Both the physiological process of ageing and the pathological effects of ocular and/or systemic disease can cause alterations in the anatomy and physiology of the retinal vasculature. Being able to recognise these alterations and understand their implications is becoming of increasing importance to eye care professionals, as is the need to consider systemic as well as ocular health when examining a patient's retina.

Recent evidence has highlighted the potential role that retinal vascular assessment could play in evaluating an individual's cardiovascular risk (Seshadri et al., 2014, Ikram et al., 2013), as well as in determining the presence of systemic vascular diseases, such as coronary heart disease (Wong et al., 2006), chronic kidney disease (Yau et al., 2011), stroke

(Ikram et al., 2006a) and Alzheimer's disease (Cheung et al., 2014), alongside the more traditional diabetes mellitus (Hiller et al., 1988) and arterial hypertension (Dimmitt et al., 1989). Such insights have largely arisen as a result of both the introduction of new high-resolution imaging technologies and an increase in awareness of the anatomical and physiological similarities that the microvasculature of the retina shares with other systemic vascular beds, in particular the cerebral and coronary circulations (Patton et al., 2005). Indeed, the anatomical positioning of the cerebral and coronary microcirculations makes them notoriously difficult to assess non-invasively, however it has been proposed that targeted non-invasive assessment of the more easily accessible retinal microvasculature could be used to provide a window into cerebral and/or coronary vascular health (Mroczkowska et al., 2014, Liew et al., 2008b). Whilst this has exciting potential and promising links have been identified, the task moving forward is to determine which parameters could be most useful in this regard and how they can be best evaluated, imaged and monitored in clinical practice.

How can the retinal vasculature be assessed?

Traditionally, in accordance with the guidance for professional practice set out by the College of Optometrists (College of Optometrists., 2014), the retinal vasculature has been routinely assessed by means of either direct ophthalmoscopy or slit lamp indirect biomicroscopy, through either dilated or undilated pupils, with judgements on factors such as retinal vessel calibre, AV ratio, vessel integrity, vessel tortuosity and the presence of retinopathy lesions being made subjectively by the examiner and recorded on the patient's record. Whilst these techniques remain a main stay of optometric practice, rapid advances in imaging technology over recent years have seen imaging of the retina become an increasingly established component of the clinical care and management of patients, particularly in those presenting with retinal and/or systemic disease (Patton et al., 2006). Fundus photography and optical coherence tomography (OCT), for example, are now widely used in the diagnosis and monitoring of ocular disease in both the primary and secondary care settings and are becoming much more familiar features of everyday optometric practice. Fundus photography is a mainstay in the population-based screening of diabetic retinopathy (DR) and the use of OCT has become increasingly widespread in the diagnosis and evaluation of both age-related macular degeneration (AMD) and glaucoma. Furthermore, in the hospital setting, retinal vascular integrity is commonly evaluated using the gold standard imaging technique of fundus fluorescein angiography (FFA) and this technique has proven particularly useful in the assessment and characterisation of conditions such as neovascular AMD and ischaemic central retinal vein occlusion.

Alongside considering the structure of the retina, the importance and relevance of evaluating the function of the retinal vessels, with regard to determining cardiovascular risk and evaluating systemic vascular health, has also been increasingly recognised over recent years (Lim et al., 2013, Mroczkowska et al., 2013, Seshadri et al., 2014). Assessing how retinal vessels respond to a flicker light stimulus for example, using a device called the dynamic retinal vessel analyser (DVA), can infer information about the autoregulation capacity and functioning of the retinal vascular endothelium (Garhofer et al., 2010). Autoregulation and microvascular endothelial function is thought to be altered at the earliest stages of a vascular disease process, often before symptoms or alterations in the macrovasculature are clinically detectable. This makes functional evaluation of the retinal microvessels an attractive proposition. The use of DVA as an imaging and evaluation technique is still primarily restricted to the research setting however and is yet to fully translate into clinical practice.

So what should be looked for when assessing the retinal vasculature?

When conducting any evaluation of the retina, be it through direct ophthalmoscopy, slit lamp indirect biomicroscopy or fundus photography, the examiner needs to be mindful of signs of both physiological and/or pathological retinal vascular changes. These signs can be broadly split into two main groups, those that fall under the heading of 'classic retinopathy' and those that relate more specifically to the structure and appearance of the retinal vessels and their architecture, both are important to look out for.

Classic retinopathy lesions and their implications

Classic retinopathy lesions include microaneurysms, haemorrhages, hard exudates, cotton wool spots and neovascularisation, along with vessel nipping, focal vessel narrowing, arteriolar wall opacities and the presence of emboli. These signs primarily develop in response to the presence of known or recognisable systemic disease such as hypertension or diabetes mellitus and the classic stages and associated features of these two retinopathies should be familiar to the optometrist.

The presence of any classic retinopathy lesion is an indicator that structural and/or functional alterations have occurred at one of the anatomical sites of the retina or within one of retinal capillary beds. Retinal haemorrhages, for example, occur when the endothelial cells which line the walls of the retinal microvasculature are damaged, meaning the BRB is compromised and the constituents of the vessel leak out. The most common trigger of such

damage is hypoxia, caused by an insufficient or inconsistent blood supply to the affected region; however inflammation and other pathological processes have also been implicated (Bek, 2013). Alongside red blood cells, blood plasma also leaves the damaged vessels, causing localised oedema. The blood plasma is laden with proteins and lipids, the latter of which can accumulate to form hard exudates. Occlusion of the pre-capillary arterioles can create an ischaemic environment, which triggers vessel calibre changes and potentially a neovascular response (Wong et al., 2001).

Although most commonly linked to the presence of recognisable systemic disease, retinopathy lesions can also develop in isolation, with a number of studies having demonstrated that such lesions can be present in around 2-14% of the non-diabetic adult population aged over 40 (Klein, 1992). The occurrence of such isolated lesions in this population presents a diagnostic challenge, as they could reflect damage to the retinal vasculature from normal ageing or more likely, from other forms of undiagnosed cardiovascular or systemic disease.

The concept that a link may exist between the presence of retinopathy and the presence of systemic disease has been around for some time, being first introduced back in 1988, when it was proposed that the development of retinopathy lesions could be indicative of the presence of a more generalised microangiography, affecting not only the eyes of the individual, but also other systemic vascular beds (Hiller et al., 1988). Over recent years, and following the introduction of retinal photography, along with more standardised techniques for assessing and categorising retinopathy lesions (Patton et al., 2006), a large body of evidence has accumulated linking the presence of retinal haemorrhages, microaneurysms and cotton wool spots, to an increased risk of subclinical and clinical stroke (Wong et al., 2001, Liew et al., 2008b) and cardiovascular mortality (Wong et al., 2001, Liew et al., 2008b). The diagnostic importance of these signs should therefore not be undervalued and careful documentation; imaging and onward referral to the general practitioner for investigation of the underlying cause of such findings is warranted.

Changes in the architecture of the retinal vasculature

Alongside detecting classic retinopathy lesions, an assessment of the architecture of the retinal vasculature is also judged to be an essential element of any retinal examination. Such architectural alterations most commonly refer to changes in retinal artery and/or vein calibre, which is traditionally reflected in the recording of the arteriovenous (AV) ratio, or as alterations in the degree of retinal vessel tortuosity. Retinal vascular architectural changes such as these can be very subtle to detect with the naked eye and have traditionally only

been able to be judged subjectively, which has historically limited their use and diagnostic ability (Heitmar et al., 2014). Recent advances in imaging and analysis software, along with the introduction of more standardised semi-automated measurement techniques however, has the potential to make the evaluation of these parameters much more accessible. Furthermore, in the advent of such new technologies, additional architectural parameters, such as fractal dimension and retinal vascular branching angle, have the potential to also be considered alongside the more traditional measurements and subsequently the relevance of global changes in the geometry of the retinal vasculature to systemic vascular health have the potential to be evaluated quantitatively for the first time (Kalitzeos et al., 2013, Liew et al., 2008a).

In current optometric practice, the awareness and use of semi-automated parameters is still not widespread, with most practitioners still resorting to subjective judgements of AV ratio and vessel tortuosity. An increasing amount of attention however is now being paid to evaluating and measuring retinal vessel structure and geometry quantifiably and in a standardised manner, as the potential insight that these parameters can give is being increasingly realised (Patton et al., 2006, Liew et al., 2008b). Indeed, alterations in the small microvessels in particular are thought to develop at the earliest stages of a disease process making the microvasculature a good screening target and furthermore retinal vessel calibre changes and functional abnormalities of the retinal micro-vessels have been suggested to be good indicators of an increased risk for future damage (Ikram et al., 2013). This is as opposed to the presence of retinopathy lesions, which are considered a relatively late indication of target organ damage (Wong et al., 2001). So what semi-automated options are there and are eye care professionals really missing important vascular indicators by solely relying on subjective assessment of the retina?

Subjective (quantitative) versus objective (qualitative) evaluation of the retinal vasculature

Subjective analysis of the morphology and progression of retinal vascular signs, although still a mainstay of optometric practice, has been shown to suffer from a number of drawbacks, including low sensitivity and specificity, poor inter-observer agreement and high exposure to observer bias (Patton et al., 2006, Liew et al., 2008b). This is particularly the case when it comes to trying to judge subtle changes in vessel calibre, arteriovenous (AV) ratio and retinal vessel tortuosity (Dimmitt et al., 1989, Heitmar et al., 2014). One of the main factors that has contributed to these drawbacks is the lack of standardisation that has existed with regard to measurement location and lack of awareness of what is considered to

represent a normal/healthy retinal vascular structure. This is particularly the case for subjective AV ratio assessment where, depending on the source used, evaluation of the ratio of the retinal artery and veins can be recommended to be taken after the 1st bifurcation (Grosvenor, 1982), after the 2nd bifurcation, or at a certain disc diameter distance away from the optic nerve head (Elliott, 2014, Bass, 2009). Additionally, what is considered to be a 'normal' AV ratio can vary from 2/3 or 3/4 depending on which resources are used and there are inconsistencies over whether the measurement should be recorded as a ratio or as a percentage (Elliott, 2014, Grosvenor, 1982, Bass, 2009). These issues are all further exacerbated by the wide degree of variation that is known to exist in the retinal vascular branching patterns between healthy individuals (Stokoe and Turner, 1966) which makes the process of selecting vessels of a comparable order of division and hence subjective analysis of the retinal architecture inherently difficult (Stokoe and Turner, 1966, Wong et al., 2001).

Semi-automated, quantitative, methods of measuring retinal vascular changes have the potential to provide a much more objective and reliable assessment of the retinal vasculature than can be obtained subjectively and to also allow vascular changes to be better monitored over time. Whilst they are receiving great interest in a research setting however, their use has not yet become that widespread in optometric practice. As our understanding of what the semi-automated parameters are telling us, along with accessibility to suitable software grows however, it is envisaged that their usage in a clinical setting will increase. In the same regard, fully automated techniques that allow the identification and detection of classic retinopathy lesions from fundus photographs in, for example, diabetic patients are also being explored and as technology progresses the integration of this type of quantitative measure into a practice setting may also increase (Patton et al., 2006).

Measurement of the retinal architecture using semi-automated techniques

The most basic requirement for the objective or quantitative assessment of the retinal vessel architecture using a semi-automated analysis software package is the obtainment of a fundus photograph, which ideally, is centred on the disc and monochromatic (red-free). This set-up ensures vessel visibility and contrast is at a maximum for measurement (figure 1). Some imaging devices, depending on the manufacturer, have such semi-automated measurement packages incorporated into their software; however there are also a number of independent software packages available that can operate with any fundus image, independent of the camera system used to acquire the image.

One of the prime benefits of semi-automated analysis techniques is that they have the potential to allow a more reliable and repeatable analysis of the calibre of the retinal vessels

and hence AV ratio, than is possible by subjective means. Some of these analysis techniques also allow the semi-automated measurement of retinal vessel tortuosity, retinal branching angle and fractal dimension. Semi-automated software packages that are currently available for license include Visualis Vesselmap2 software (Imedos Systems, Germany)(IMEDOS, 2009), SIVA (Singapore 'I' Vessel Analysis, Singapore Eye Research Institute, Singapore) (SingaporeEyeResearchInstitute, 2011), IVAN (Vasculo-matic ala Nicola version 1.1, Department of Ophthalmology and Visual Science, University of Wisconsin-Madison, Madison, WI)(Shah et al., 2009) and Vampire (Vessel assessment and measurement platform for images of the Retina)(Perez-Rovira et al., 2011), amongst others. Additionally, the free software Image J can also be used to obtain semi-automated retinal vessel measurements.

1. Semi-automated measurement of retinal vessel calibre

The first attempts at semi-automated assessment of retinal vessel calibre were made in the 1960s and 1970s, shortly after the introduction of retinal photography (Patton et al., 2006). These so called micrometric measurements were taken from enlarged projected images using callipers and used to generate a quantitative assessment of AV ratio (Patton et al., 2006). Since then, imaging technologies and software options have advanced and semi-automated methods of vessel analysis have become significantly more standardised and sophisticated.

For the majority of the software packages now available, once a fundus image is acquired, a concentric annulus is superimposed on to the image. This concentric annulus is situated half a disc diameter (DD) from the outer boundary of the ONH and is half a DD wide (figure 2). The examiner is then simply required to select the relevant arteries and veins from within this concentric annulus (figure 3). All calculations are then computed and generated for the user automatically. It has been shown that by measuring the diameter of the vessels within this concentric region it ensures that the arteriolar vessels which are more likely to exhibit changes are being used for measurement purposes as opposed to the bigger arteries and it allows a much greater standardisation in measurement than has been achievable previously. The retinal vessel calibre measurements generated by these semi-automated techniques are denoted as the Central Retinal Artery Equivalent (CRAE) and Central Retinal Vein Equivalent (CRVE), which are then used to derive the arteriovenous ratio (AVR).

This measurement technique and the parameters derived from it all stems from the work of Parr and Spears, who in 1974, derived a formula based on this concentric annulus approach for the calculation of central retinal artery calibre from selection of all retinal arteries within

the annulus, termed the 'central retinal artery equivalent' (CRAE) (Parr and Spears, 1974). This work was expanded on by Hubbard and colleagues in 1992 (Hubbard et al., 1999), who derived a similar formula for the calculation of vein diameter, termed the central retinal vein equivalent (CRVE) and then used this in conjunction with the CRAE to determine the semi-automated AVR in a less time-consuming manner than had previously been possible (Patton et al., 2006). The formulae obtain the CRAE and CRVE outputs by first combining pairs of the narrower and wider vessel branch diameter measurements taken from within the concentric circle and converting them into estimates of their larger trunk diameters. Pairs of the trunk diameters are then combined in a similar manner until all arterioles and venules are built up and summarised into a single central retinal artery (CRAE) and vein (CRVE) equivalent parameter (Hubbard et al., 1999). More extensive details of the iterations of these formulas can be found elsewhere (Hubbard et al., 1999, Parr and Spears, 1974, Knudtson et al., 2003, Heitmar et al., 2015)

The Atherosclerosis and Risks in Communities (ARIC) study was the first to utilise this objective approach to measure AV ratio semi-automatically in large patient cohorts, and found semi-automated AVR to be a good measure of generalised arteriolar attenuation (Hubbard et al., 1999). Later, in 2003, Knudtson and colleagues went on to publish a revised formula for CRAE and CRVE calculation, based on selection of the 6 largest arterioles and venules from within the concentric measurement ring as opposed to using all of the acceptable vessels as had been done previously (Knudtson et al., 2003). This revised formula has been shown to be in agreement with the previous calculation proposed by Parr-Hubbard (Heitmar et al., 2015), but has the added advantage of being independent of the units of scale and of the number of vessels measured (Patton et al., 2006). Recently Heitmar et al (2015) demonstrated that the most important factor to assure agreement in output between the two formulas with regard to AVR calculation is to ensure that the number of selected arterioles and venules were kept equal for each, be that at the traditional 6 or at 5 or another alternative number. This is likely to become especially relevant as the use of these techniques in the clinical setting increases. The revised formula proposed by Knudtson et al (2003) has been adopted by some of the newer semi-automated programs and it currently the most widely used formula for calculating retinal vessel calibre.

2. Retinal Tortuosity

Alongside AVR, retinal vessel tortuosity is a common parameter recorded during assessment of the retinal vasculature and increased retinal vessel tortuosity has been linked to the presence of a number of systemic vascular pathologies, including more unusually, Alzheimer's disease (Cheung et al., 2014), retinopathy of prematurity (Shah et al., 2009) and

chronic anaemia (Incorvaia et al., 2003). In the same manner as AV ratio however, subjective evaluation of retinal tortuosity by optometrists suffers from a number of drawbacks related to the high risk of observer bias and low specificity and sensitivity. As such a quantitative and repeatable means of determining retinal tortuosity has also been desired.

The first quantitative assessment of retinal vessel tortuosity was described by Lotmar et al in 1979. This method was then expanded upon slightly by Bracher (1982) and the generation of tortuosity indices has evolved further from there (Kalitzeos et al., 2013). Whilst most quantitative measurements of vessel tortuosity involve the subdivision of the tortuous vessel into a series of single arcs through the manual selection of points on a fundus photograph, a standardised method of obtaining this measure is yet to be clearly outlined. Instead, a large variety of different tortuosity indices are currently in existence (Kalitzeos et al., 2013). This has meant that, although it is provided with some semi-automated software packages, the use of quantitative retinal tortuosity measurements has struggled to move out of the research setting to so far but has the potential to do so more in the future.

3. Fractal dimension and retinal vessel branching angle

Some semi-automated analysis software packages allow the measurement of fractal dimension and retinal vessel branching angle. Fractal dimension represents a 'global' measure that summarises the whole branching pattern of the retinal vascular tree (Liew et al., 2008a). Retinal vessel branching angle on the other hand can be defined as the first angle subtended between two daughter vessels at each vascular bifurcation (Cheung et al., 2014). Fractal dimension has been shown to be a relatively sensitive early indicator of vascular changes in diabetic retinopathy (Avakian et al., 2002), systemic hypertension and systemic cardiovascular disease (Liew et al., 2008a) by some studies, but not by others (Kunicki et al., 2009). Again, this is a parameter that, whilst offered by some semi-automated software packages, has not fully crossed the path from research into the clinical setting to date. Indeed, there are natural limitations that exist when trying to apply fractal analysis to a branching biological structure and further insight into its clinical relevance is still required before its full potential can be realised.

So what is the value of assessing the retinal vasculature?

The retina offers an ideal location for the non-invasive evaluation of vascular structure and function. The increase in use and availability of imaging technologies has significantly enhanced the ability of the optometrist to document and monitor both classic retinopathy and vascular architectural changes over time and such changes have been shown to have the

potential to infer valuable information regarding not only ocular but also systemic vascular health.

Optometrists are ideally placed to routinely document and monitor retinal vascular parameters over time due to their captive patient base and the regularity of routine eye examinations. The traditionally subjective nature of measuring and recording retinal vascular changes has, to date however, somewhat limited the usability of retinal vessel parameters as potential biomarkers for cardiovascular risk and systemic vascular health. With the emerging role of semi-automated methods of measuring architectural changes however, there is an increasing potential that the appearance and progression of vascular architectural changes in particular, could be more precisely measured and monitored over time. Indeed, determining the validity, clinical relevance and usability of semi-automated measurements of the retinal architecture is currently a research area of intense interest and associations between altered retinal vessel calibre and the presence of ocular and systemic diseases are being continually explored, with a view to determining whether such measurements have the potential to act as biomarkers for cardiovascular risk in certain patient subgroups (Ikram et al., 2013, Wong et al., 2001).

What does the evidence suggest to date?

There are a wide range of studies that have looked into the associations between the semi-automated architectural parameters, CRAE, CRVE and AVR and the presence of both clinical and subclinical cardiovascular disease states. Interestingly, whilst AVR is the traditionally recommended measurement parameter, it is CRAE and CRVE individually that have emerged as the more sensitive biomarkers of cardiovascular disease, as they have been recognised to provide more clinically relevant information than can be offered by considering AVR in isolation. Indeed, AVR, due to its nature, is an ambiguous measure as both an alteration in the diameter of the artery and an alteration in the diameter of the vein could cause an equivalent alteration in AVR. For example the same change in AVR would occur if either CRAE was decreased or if CRVE were increased, but by just considering the AVR alone, the information about the specific direction of the change cannot be obtained. The requirement for this additional insight has become of particular relevance in recent years as imaging and semi-automated analysis methods have become more accessible, Indeed, changes in CRAE and changes in CRVE, when considered individually have been shown to link to very different disease mechanisms.

With regard to retinal artery diameter, it is widely accepted that systemic hypertension leads to attenuation of arteries throughout the body and indeed in line with this, a decrease or

narrowing in CRAE, beyond that which would be expected through normal ageing (Ikram et al., 2013), has been strongly linked to the presence and degree of systemic hypertension (Hubbard et al., 1999). More interestingly, a number of longitudinal cohort studies have additionally demonstrated that a decrease in CRAE may actually precede the development of systemic hypertension and may therefore signal an increased risk for the subsequent development of hypertension and could act as a biomarker of risk in certain individuals (Chew et al., 2012). As well as systemic hypertension, a decrease in CRAE has also been linked to a higher risk of developing chronic kidney disease (Yau et al., 2011) and to a 2-fold increase in the risk of developing coronary heart disease in those with hypertension (Wong et al., 2006). The renal and coronary microvasculature beds share anatomical and physiological associations with that of the retina and therefore these findings reinforce the potential role that semi-automated retinal microvascular imaging could play as a window into general systemic vascular health.

Interestingly, alongside CRAE, CRVE is also emerging as an important indicator of systemic vascular health, with an increase in CRVE having been linked to the presence of cardiovascular risk factors such as cigarette smoking, obesity, raised BMI, dyslipidaemia (Sun et al., 2009) and other subclinical signs such as the presence of both inflammatory markers and markers of vascular endothelial dysfunction (Sun et al., 2009). In turn, increased CRVE has been linked to the presence and development of cerebral small vessel disease (Ikram et al., 2006b), vascular dementia (de Jong et al., 2011), diabetes (Ikram et al., 2006c), coronary heart disease (Wong et al., 2006) and stroke (Ikram et al., 2006a). It is possible therefore, that recording an increase in CRVE could have the potential to signal a high risk for future cardiovascular disease development in certain sub-groups of patients.

Aside from systemic disease and cardiovascular risk, alterations in semi-automated retinal vascular calibre parameters have also been linked to the development and progression of ocular disease. For example, larger CRVE has been linked to increased incidence and accelerated progression of diabetic retinopathy in type 1 diabetics (Klein et al., 2004). Furthermore wider CRAE has also been independently associated with the development of age-related macular degeneration (AMD) (Jeganathan et al., 2008) and a narrowing of CRAE and CRVE has been linked to RNFL loss and long term risk of primary open angle glaucoma development (POAG) (Ikram et al., 2013). The strength and nature of these associations is however still relatively uncertain as these findings have not been replicated by all studies and more research is required (Ikram et al., 2013), what these associations do demonstrate however is the potential insight that could be gained by the more widespread

use and quantifiable evaluation of retinal architectural changes by semi-automated means in the future.

What does the future hold?

Imaging technology is constantly developing and is highly likely to become even more sophisticated in future years, with increased image resolutions and enhanced image processing power offering the potential for an even greater insight to be gained into structural and functional changes of the retinal vasculature than is currently possible (Patton et al., 2006). Accessibility to imaging technology within the optometric community is continually growing and as awareness and usability of objective and/or semi-automated image analysis methods increases, the diagnostic capacity of the optometrist, with regard to determining the presence and future risk of both ocular and systemic disease also has the potential to expand, especially as our understanding of the pathogenesis of these conditions improves.

The exciting introduction of new imaging modalities, such as longer wavelength and swept source OCT and en-face imaging, is opening up the potential for very subtle changes in not only the retinal but also the choroidal vasculature to be detected and evaluated (Adhi and Duker, 2013). Furthermore, the ongoing development of Doppler OCT and the introduction of retinal oximetry is offering the potential for functional changes in the retinal vasculature to be considered in closer conjunction with structural changes (Adhi and Duker, 2013, Patton et al., 2006). As the evidence base, awareness and commercial availability of these technologies improve it is likely that their usability will start to move out of the research setting and into the clinical setting more in the near future.

Summary

Evaluation of the retinal vasculature is an essential and routine part of any optometric examination (College of Optometrists, 2014). Both the presence of classic retinopathy lesions and/or the presence of subtle changes in the retinal architecture can infer valuable information about not only ocular, but also systemic vascular health. The retina offers an ideal location for non-invasive evaluation of the microvasculature and due to the anatomical and physiological similarities that are known to exist between different microvascular beds, there is a real potential for retinal evaluation to provide a valuable window into systemic vascular health. Promising associations between retinal vessel calibres and cardiovascular risk have been identified, however to date, reliable evaluation and monitoring of such subtle

retinal architectural changes has been limited by the subjective nature by which such parameters have been assessed. The use of imaging technology is now relatively widespread in routine optometric practice and optometrists, with their captive and regularly attending patient base, are ideally positioned to routinely document and monitor retinal vascular parameters over time. Semi-automated measurement techniques are now commercially available and optometrists should familiarise themselves with these techniques and the parameters that they can generate. The possibility that subtle architectural changes could act as biomarkers for cardiovascular risk is an exciting one and as awareness of the available imaging and analysis methods continue to improve the optometrist's ability to evaluate and understand the clinical relevance of retinal vascular changes should progress significantly from now and into the future.

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