We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,300

Open access books available

172,000

International authors and editors

190M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

Lipoproteins and Apolipoproteins of the Ageing Eye

Edward Loane

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/45985>

1. Introduction

In this chapter, we outline the structure of the retina and the aetiopathogenesis of the major age-related eye disease: age-related macular degeneration (AMD). We then discuss the role that lipoproteins and apolipoproteins play in the ageing eye and in the development of AMD.

2. The macula and retina

The macula is the central part of the retina, the neurosensory portion of the eye, and it is responsible for detailed central and colour vision due to its high concentration of cone photoreceptors. Anatomically, the macula is centred on the foveola, and has a ganglion cell layer of more than one cell in thickness. The macula has a diameter of approximately 5.5 mm. The macula is characterised by a yellowish colour (hence the term *macula lutea*, which is Latin for 'yellow spot'), attributable to the presence of macular pigment (MP).[1] The concentration of MP peaks at the centre of the macula, where the appearance of the 'yellow spot' may be clearly evident on clinical examination or fundus photography [Figure 1]. MP is optically undetectable outside the macula.[2] Within the layer structure of the retina, the highest concentration of MP is seen in the receptor axon layer and the inner plexiform layer.[1]

The retina consists of a neurosensory portion comprised of nine individual layers, and an external retinal pigment epithelium (RPE). The RPE plays an important physiological role in the maintenance of neurosensory retinal health, through functions including Vitamin A metabolism, phagocytosis of photoreceptor outer segments, maintenance of the outer blood-retina barrier, heat exchange, and the active transport of substances in and out of the RPE.[3] The blood supply of the retina is derived from the inner retinal vasculature and the outer choriocapillaris. Non-pathological changes that occur in the RPE with age include an

increase in cellular pleomorphism and a decrease in cell number, with migration of peripheral RPE cells towards the macula, reduced melanin composition, and an accumulation of the age-pigment lipofuscin.[4;5] These changes may lead to a reduction in the metabolic activity of the RPE, with subsequent apoptosis, which pre-dates pathological change.[5;6] The RPE is separated from the choriocapillaris by Bruch's membrane (BrM). BrM is a semipermeable filtration barrier, comprised of five individual layers.[7;8] Disruption of BrM may result in alteration of its filtration properties, impacting on the function of the RPE and the neurosensory retina.[9] Changes that occur in BrM with age include an increase in its overall thickness, with a reconfiguration of associated lipids and proteins and the accumulation of debris.[10;11] When this debris accumulates between BrM and the RPE, it is referred to as a basal laminar deposit (BlamD) and is not specifically pathological in nature.[12] However, when deposits accumulate within the inner collagenous layer of BrM, they are referred to as basal linear deposits (BlinDs) and are a histopathological hallmark of AMD.[13] These deposits (BlamDs and BlinDs) contain a wide range of constituents including collagen, inflammatory proteins and lipoproteins. When sufficient debris accumulates in BlinDs, they are visible clinically as drusen.[14;15]

3. Age-related macular degeneration

Age-related macular degeneration (AMD) is the leading cause of blindness in people over 50 years of age in the developed world, and it results in loss of central and colour vision if not treated, or if not amenable to treatment.[16-18] The loss of central vision impacts greatly on the individual, as their ability to perform simple daily tasks, such as reading, watching television, driving and recognizing people's faces becomes increasingly difficult. Thus, their quality of life and their ability to lead an independent life diminish significantly as the disease progresses. The peripheral retina is not affected in individuals with AMD, regardless of stage, such that, in the absence of other ocular pathology, peripheral (navigational) vision remains unchanged.

It is currently estimated that late AMD affects 513,000 people in the United Kingdom (2.4% of those over the age of 50), and that this number will increase to 679,000 by the year 2020.[19] Prevalence data from the United States in 2004 estimated that more than 1.75 million individuals were affected by the disease, with this latter figure expected to rise to almost 3 million by the year 2020.[20] The prevalence of this condition is likely to increase dramatically in the future, as a result of increasing life-expectancy and the resultant increasing senescence of society.[21] Data from the National Eye Institute in the United States in 2004 indicated that the prevalence of advanced AMD in people over 40 years of age was 1.47%, rising to 15% in white females aged over 80 years. Beyond its impact on the individual sufferer,[22] the predicted increase in longevity (Figure 2), coupled with the predicted growth in world population (Figure 3) will significantly increase the socio-economic burden that AMD places on countries and their health-care systems.[23-26]

Figure 1. Colour fundus photograph showing the macula, surrounding the fovea, which is centred on the foveola (not marked, but evident as the 'yellow spot') of a left eye.

Figure 2. Male and female life expectancy 1950-2050.
* Figures from 1950 and 2001: Irish Department of Health and Children data;
Projected figures for 2028 and 2050: USA data.

World Population 1950-2050

Figure 3. World population 1950-2050 (predicted).

4. Classification of AMD

In 1995, the International Age-Related Maculopathy Epidemiological Study Group clarified the definition and core grading system used to detect and define AMD.[27] This was done to homogenize the systems used to identify and classify this disease in all future clinical and epidemiological studies. This current classification system defines AMD primarily on the basis of morphological changes, without reference to visual acuity.

AMD is defined as a disorder of the macular area, most often clinically apparent after 50 years of age, and characterised by any of the following findings, which are not patently due to another disorder:

1. Soft drusen $\geq 63 \mu\text{m}$ in diameter. Drusen are whitish-yellow spots that lie external to the neurosensory retina or the RPE (Figure 4). Drusen may be soft and confluent, soft distinct, or soft indistinct. Hard drusen do not, of themselves, characterize AMD.
2. Hyperpigmentation in the outer retina or choroid associated with drusen.
3. Hypopigmentation of the RPE, most often more sharply demarcated than drusen, without any visible choroidal vessels associated with drusen.

Figure 4. Macular soft drusen of a left eye.

These age-related pathological changes, which are associated with progressive accumulation of debris under the retina, predispose to the late stage of AMD.[28;29] Late AMD is classified as either geographic atrophy (atrophic AMD) or neovascular AMD (choroidal neovascularisation, also referred to as 'exudative AMD' or 'disciform AMD').

Geographic atrophy (GA) is characterised by the following, which is not patently due to another disorder:

1. Any sharply delineated area of hypopigmentation, or depigmentation, or apparent absence of the RPE, in which the choroidal vasculature is more visible than in the surrounding area. The area of atrophy must be $\geq 175 \mu\text{m}$ in diameter (Figure 5).

Neovascular AMD is characterised by any of the following, which are not patently due to another disorder:

1. RPE detachment(s), which may be associated with neurosensory retinal detachment.
2. Subretinal or sub-RPE neovascularisation.
3. Epiretinal, intraretinal, subretinal, or sub-RPE glial tissue or fibrin-like deposits.
4. Subretinal haemorrhage (Figure 6).
5. Hard exudates (lipids) within the macular area, related to any of the above, in the absence of other retinal vascular disease.

Rarely, neovascular AMD may develop in an area of GA. If this happens, the affected eye is re-classified as having neovascular AMD.

Figure 5. Geographic atrophy, affecting the entire macula of a right eye.

Figure 6. Neovascular AMD, showing sub-retinal haemorrhage in a left eye.

5. Pathogenesis of AMD

AMD has a multi-factorial pathogenesis.[30;31] Therefore, the development of AMD is dependent on a complex interaction between an individual's genetic composition (genotype) and lifestyle (or environmental) factors. This interaction is complex and incompletely understood; however, certain factors have been well established as representing risk for this condition, whereas others are known as putative risk factors, according to our current understanding of this disease. The well-established risk factors for the development of AMD are: increasing age, a positive family history of AMD (including specific genotypes), and tobacco smoking.[30;32;33] Therefore, tobacco smoking is the only proven environmental/lifestyle risk factor for this disease.[34;35] Putative risk factors include: obesity,[36;37] hypertension,[38] light iris colour,[39] cumulative sunlight exposure,[40] and a diet low in anti-oxidant fruits and vegetables,[41] particularly those

containing the hydroxy-carotenoids: lutein and zeaxanthin.[42] Although the pathogenesis of AMD remains incompletely understood, there is a growing consensus that one or more of the following processes contribute to this condition: inflammation; oxidative stress; cumulative blue light damage; RPE cell and BrM dysfunction; reduced foveolar choroidal circulation.

6. Macular pigment

Macular pigment (MP) is composed of the hydroxy-carotenoids lutein (L), zeaxanthin (Z), and *meso*-zeaxanthin (*meso*-Z). L and Z are of dietary origin and are not synthesized *de novo* in humans, whereas *meso*-Z is not found in a conventional western diet, but is understood to be primarily formed in the retina following conversion from L.[43;44] Interestingly, it has been shown that L is the dominant carotenoid in the diet,[45] whereas Z/*meso*-Z have been shown to be the dominant carotenoids at the central macula.[46;47] MP is found in highest concentration at the central macula, where it functions as a powerful antioxidant and acts as a filter of actinic short wavelength blue light, thus limiting (photo-)oxidative damage to retinal cells.[48] These properties of MP are believed to be the mechanism whereby it may protect against the development, and/or progression, of AMD.

Although MP is entirely of dietary origin, it is also subject to heritability, as reported in 2005 by Liew *et al.* in a classic twin study.[49] In that study of 76 monozygotic and 74 dizygotic female twin pairs, they estimated that heritability accounted for between 67% and 85% of an individual's MP level. However, to date a direct significant association between MP levels and the major risk genes for AMD has not been shown.[50]

MP can be measured *in vivo* by non-invasive psychophysical means, resulting in an MP optical density measurement.[51;52]

7. Lipoproteins

Circulating lipoproteins consist of a complex of triglycerides, phospholipids and cholesterol, and one or more specific proteins, referred to as apolipoproteins. The association of lipoproteins with high affinity receptors on cell surfaces regulates lipid metabolism and transport in the body.[53] Lipoproteins are classified into the following six groups: chylomicrons; chylomicron remnants; very low density lipoproteins (VLDL); intermediate density lipoproteins (IDL); low density lipoproteins (LDL); high density lipoproteins (HDL).[53]

Chylomicrons are synthesised by the intestine and deliver dietary triglycerides to muscle and adipose tissue, and dietary cholesterol to the liver. Lipoprotein lipase, located at capillary endothelial cell surfaces, hydrolyses the triglyceride core of the chylomicron, thus liberating fatty acids and glycerol, which are used as energy sources by various cells, or are taken up by adipocytes and stored as triglycerides. Chylomicron remnants, which are rich in cholesterol, result from chylomicron metabolism, and are rapidly cleared by the liver.[53]

Subsequently, the liver synthesises a second class of triglyceride-rich lipoprotein, referred to as VLDL, which, upon secretion, functions as a transporter of lipids and cholesterol. In the bloodstream, VLDL undergoes progressive removal of triglycerides from its core by lipoprotein lipase, in a similar way to chylomicrons. The VLDL particles thus become increasingly smaller, leading to the formation of IDL, and LDL. LDL are the final metabolic products of VLDL and are responsible for most of the cholesterol transport in serum.[53]

HDL are the smallest lipoproteins, arising from several sources including the intestine and liver. HDL are involved in a process known as 'reverse cholesterol transport', whereby HDL acquire cholesterol from cells and deliver it to the liver.[53] This is a particularly important mechanism in humans, as the quantities of cholesterol transported out of the gut and liver far exceed the quantities converted to steroid hormones, or those lost through the skin in sebum. Thus, unless the requirement for cell membrane repair or synthesis is high, excess cholesterol must be returned to the liver for excretion.[54]

8. Association of carotenoids with plasma lipoproteins

The majority of plasma carotenoids are transported on LDL, with 55% of total carotenoids associated with this lipoprotein, whereas HDL is associated with 33%, and VLDL is associated with 10-19%, of the total carotenoids.[55] However, in the case of the hydroxycarotenoids, L and Z, some studies have reported that they are relatively equally distributed between LDL and HDL molecules, but other studies have reported that HDL is the preferential carrier of the MP carotenoids in plasma.[56;57]

MP is inversely related to percentage body fat.[58] Interestingly, Viroonudomphol *et al.* have demonstrated lower levels of HDL in overweight and obese subjects, consistent with the possibility that a relative lack of HDL may impair transport and/or retinal capture of the carotenoids.[59] Furthermore, Seddon and co-workers have demonstrated a significantly increased risk of AMD in association with obesity.[33] These findings have prompted the suggestion that an individual's lipoprotein, and apolipoprotein, profile may influence the transport and delivery of these carotenoids to the retina, with a consequential impact on MP.

A recent study, designed to investigate the respective relationships between lipoprotein profile, MP optical density and serum concentrations of L and Z, was conducted in 302 healthy adult subjects.[60] This study found that there was a statistically significant inverse association between serum triglyceride concentration and MP optical density, and an inverse association between serum triglyceride concentration and serum L concentration in subjects with a positive family history of AMD. There have been no previous reports on the association between serum triglyceride concentration and either MP optical density or serum concentrations of L and/or Z. Elevated serum triglyceride concentration is an element of an undesirable lipoprotein profile and represents risk for cardiovascular disease.[61;62] Since there is an inverse association between serum triglyceride concentration and serum HDL concentration,[62] one could expect an inverse association between serum triglyceride concentration and serum L, since HDL appears to be the most important lipoprotein

involved in the transport of L in serum. This expected inverse association was observed in subjects with a positive family history of AMD. In this study sample there was a positive and significant association between serum HDL concentration (and serum cholesterol concentration) and serum L and Z concentrations. Of note, there was no significant association observed between MP optical density and either serum cholesterol concentration or serum HDL concentration. There was also no association between serum LDL concentration and MP optical density (or serum concentrations of its constituent carotenoids). These findings suggest that a desirable lipoprotein profile (higher serum HDL, lower serum LDL and lower serum triglyceride concentrations) is associated with greater serum L concentration. However, the impact of lipoprotein profile on the capture and/or stabilization of these carotenoids at the macula, where they comprise MP, is less clear from this data.

In this study, the lipoprotein particle-concentration of L and/or Z in serum was not directly measured, nor were lipoprotein subspecies measured, as performed by Goulinet *et al.*[57] In their study, they fractionated HDL and LDL subspecies on the basis of their hydrated density by gradient ultracentrifugation, and they found that serum L and Z (combined) were relatively equally distributed between HDL and LDL; but more importantly, they found that there was a progressive decrease in the concentration of these carotenoids with increasing density (and decreasing lipoprotein particle size) from light to dense LDL. They also found that the majority of macular carotenoid transport by LDL was accounted for by the most abundant subspecies, LDL3 (intermediate LDL) and LDL4 (dense LDL). This is highly relevant to the transport of L and Z in serum, as LDL3 and LDL4, despite being the most abundant subspecies of LDL in that study, had reduced particle-concentrations of these carotenoids compared to less dense LDL subspecies, making them more vulnerable to oxidation.[63] LDL is the primary component of total cholesterol,[62] and has previously been reported in various studies to transport between 22-44% of L and Z in serum.[55;57;64-66] Of note, it has been shown that there is no significant difference in the transport of L and Z by lipoproteins between subjects with and without AMD.[65]

The findings of Goulinet *et al* in relation to HDL were similar to that of LDL, in that there was a progressive and marked decrease in HDL particle concentration of L and Z, with maximal carotenoid concentration evident in the lightest, largest HDL subspecies (HDL2-1), and minimal concentration in the densest HDL. Certainly, the findings of Goulinet *et al* with respect to HDL, in concert with our findings, are consistent with the view that HDL plays an important role in the transport of L and Z in human serum, and are provocative given that AMD and cardiovascular disease share certain antecedents.[32;57;60;67-70] Furthermore, and again consistent with a shared pathogenesis between AMD and cardiovascular disease, the finding of an inverse association between serum triglyceride concentration and MP optical density (and between serum triglyceride concentration and serum L concentration) in subjects with a positive family history of AMD, is noteworthy.[60] Since AMD has been shown to be associated with low serum concentrations of L,[71] and given that risk factors for AMD are associated with a relative lack of MP,[31] our observations are yet another example of how AMD and cardiovascular disease share risk factors.[32;60-62;67-70]

In 2007, Connor *et al* reported on the role that HDL plays in the transport of L and Z in serum in a study involving WHAM chicks.[64] WHAM chickens have a recessive sex-linked mutation in the *ABCA1* transporter gene that results in very low circulating HDL concentration, with normal, or increased, concentrations of other plasma lipoproteins, particularly LDL. The analogous mutation in humans results in Tangier disease, which is characterized by a similar deficiency in circulating HDL concentration.[72] In their study, involving 24 WHAM chicks and 24 control chicks, Connor *et al* found that one-day old WHAM chicks had only 9% of the L concentration in plasma when compared with control chicks, and only 6% of the retinal concentration of controls (the corresponding concentrations of Z were 6% and 9%, respectively). Following a high-L diet for 28 days, there was a significant increase in the plasma and retinal concentrations of L in WHAM chicks and controls, but the increases were still greatly inferior in the WHAM chicks when compared with control chicks and, furthermore, still did not reach the concentrations observed in the one-day old control chicks. The observations of Connor *et al* suggest an important role for HDL in the transport of L and Z in serum and/or their incorporation into the retina, and are consistent with our findings.[60;64]

Interestingly, although all subjects in our study were healthy volunteers with no evidence of ocular pathology, it is notable that, on average, subjects with a positive family history of AMD had a higher serum concentration of L than subjects with a negative family history of AMD, yet MP optical density levels in both groups were comparable, as were serum concentrations of HDL.[60] As was shown in this study, and as has previously been documented,[73] serum concentrations of L and Z generally correlate positively with MP optical density. Therefore, it is plausible to suggest that in the subjects in this study with a positive family history of AMD, the delivery to, and/or uptake by, the retina of the macular carotenoids is defective when compared to subjects without such a family history.[60] Indeed, although MP optical density levels were comparable between subjects with and without a family history of AMD, subjects with a positive family history of this disease also had higher serum L concentrations. This is consistent with the observations of Nolan *et al*, where a relative lack of MP was seen in association with a positive family history of AMD in 828 healthy subjects, but where dietary and serum concentrations of L and Z were comparable for subjects with and without a family history of this condition, suggesting defective retinal capture of circulating L and/or Z in persons who are genetically predisposed to AMD.[31] Mechanisms governing the retinal capture and/or stabilization of L and/or Z may be subject to influence by HDL subspecies profile, by affecting receptor-mediated uptake of these carotenoids from serum. Indeed, apolipoprotein profile is probably a determinant of retinal uptake of the macular carotenoids from serum, reflected in our recently reported finding that individuals with at least one Apo ϵ 4 allele exhibit significantly higher MP optical density than individuals without this protective allele, despite statistically comparable serum concentrations of L and Z.[74] Interestingly, the lack of an association between MP optical density and either serum cholesterol concentration or serum HDL concentration in our study would suggest that our observations are more likely due to impaired uptake and/or stabilization of circulating L and/or Z by the macula than

due to any impact the HDL subspecies profile may have on the transport of the macular carotenoids in serum.

Another recent study has shown somewhat conflicting evidence regarding the association between circulating lipoprotein levels and MP levels in serum and in the macula.[75] These differences may be attributable to differences in the methods used to measure serum lipoproteins, although it should be noted that this study also found a positive association between serum L and serum HDL levels, underscoring the importance of HDL as a transporter of L in serum. However, it should be emphasised that a notable paucity of data still remains regarding the mechanism(s) whereby L and Z accumulate in the liver, are repackaged into lipoproteins, and transported via the circulatory system to specific target tissues such as the retina.

9. Apolipoproteins

Plasma lipoproteins include one or more protein constituents, known as apolipoproteins. Apolipoproteins have been classified into several subgroups, including apolipoprotein A (ApoA), apolipoprotein B (ApoB), apolipoprotein C (ApoC), and apolipoprotein E (ApoE). These subgroups are themselves further sub-classified, for example: ApoA-I, ApoA-II etc. Each lipoprotein class is associated with certain apolipoproteins, for example: chylomicrons and VLDL are associated with ApoB; chylomicrons, VLDL and HDL are associated with ApoE.[76] The primary role of apolipoproteins is the transport and redistribution of lipids amongst various tissues in the body. Specific apolipoproteins are recognised by cell surface receptors, and this facilitates the high affinity binding required for delivery to target tissues. Certain apolipoproteins also act as cofactors of enzymes involved in lipoprotein metabolic pathways, including those of lipoprotein lipase and lecithin-cholesterol acyl transferase (LCAT), which catalyse the formation of cholesterol esters. Another role of specific apolipoproteins is the maintenance of the structure of lipoproteins, by stabilizing their micellar structure, and by providing a hydrophilic surface in association with phospholipids.[53] The function of apolipoproteins has provoked interest in their possible role in a range of degenerative conditions. In particular, several investigators have suggested an association between ApoE and various diseases, including Alzheimer's disease, atherosclerosis and AMD.[77-80]

Abalain *et al.* investigated the association between AMD and serum levels of lipoproteins and lipoparticles.[78] They found that there was no difference in serum ApoA-I and ApoB levels between AMD patients and controls. However, they found that serum ApoE levels were higher, and that serum ApoC-III levels were lower, in AMD patients compared with controls. The higher level of serum ApoE in AMD patients is consistent with the findings of Boerwinkle and Utermann, who found that the Apo ϵ 4 allele is associated with lower serum ApoE levels, and that the Apo ϵ 2 allele is associated with higher serum levels of ApoE.[79] ApoC-III interferes with lipoprotein metabolism and, when associated with ApoB as a lipoparticle, it has been shown to be involved in atherogenesis.[80] Abalain *et al.* found no difference in the levels of this particular lipoparticle between AMD patients and

controls.[78] The evidence to date suggests that, of the apolipoproteins, ApoE has the strongest association with AMD.

10. Apolipoprotein E

ApoE is a structural component of plasma chylomicrons, VLDL, and a subclass of HDL. It is a 299 amino-acid protein, and is synthesised in a large number of tissues including the spleen, kidneys, lungs, adrenal glands, liver, brain and retinal Müller cells.[81] ApoE is polymorphic, with three common isoforms: E2, E3 and E4, which are coded for by three separate alleles: Apo ϵ 2, Apo ϵ 3 and Apo ϵ 4. These alleles are differentiated on the basis of cysteine-arginine residue interchanges at sites 112 and 158 in the amino acid sequence.[82] As a result of this polymorphism, six common phenotypes exist: three homozygous phenotypes (ϵ 3 ϵ 3, ϵ 2 ϵ 2, ϵ 4 ϵ 4) and three heterozygous phenotypes (ϵ 2 ϵ 3, ϵ 2 ϵ 4, ϵ 3 ϵ 4). ApoE is crucial to many processes, including: cholesterol transport and metabolism; receptor-mediated uptake of specific lipoproteins; heparin binding; formation of cholesteryl-ester-rich particles; lipolytic processing of type III β -VLDL; inhibition of mitogenic stimulation of lymphocytes; transport of lipids within the brain.[53]

ApoE is an important regulator of cholesterol metabolism because of its affinity for ApoE-specific receptors in the liver, and its affinity for LDL receptors in the liver and other peripheral tissues requiring cholesterol.[53] ApoE-specific receptors are present on the membranes of hepatic parenchymal cells, and have a high binding affinity for chylomicron remnants, IDL and a sub-class of HDL. ApoE also regulates the activity of several lipid-metabolising enzymes, including lipoprotein lipase, and LCAT.

ApoE is found in greatest concentrations in the liver. However, it is also the predominant apolipoprotein in the brain, and is responsible for lipid transport and cholesterol regulation within the central nervous system (CNS). ApoE is a major component of plasma and cerebrospinal fluid, and plays a fundamental role after CNS injury, where it appears to regulate the transport of cholesterol and phospholipids during the early and intermediate phases of the reinnervation process.[83;84]

ApoE polymorphisms result in differences in the metabolism of ApoE-containing lipoprotein particles.[85] For example, it is possible that certain ApoE polymorphisms affect their ability to interact with lipoprotein lipase in the conversion of VLDL to LDL.[86] Indeed, ApoE polymorphism influences plasma lipid levels both in sedentary states and in their response to exercise, and it is therefore believed to be related to risk for coronary artery disease. In general, carriers of the Apo ϵ 4 allele have higher levels of total cholesterol and LDL-cholesterol than those with the Apo ϵ 3 allele. ApoE polymorphism also appears to play a role in the responsiveness of blood lipids to dietary and lipid-lowering drug interventions. Thus, the ApoE gene-environmental interactions contribute to population variance in blood lipid-lipoprotein levels.[87]

ApoE receptors also play an important role in lipoprotein metabolism. The primary physiological role of ApoE is to facilitate the binding of lipoproteins to LDL receptors,

thereby regulating the uptake of cholesterol required by the cell. For instance, large amounts of lipids are released from degenerating cell membranes after nerve cell loss, thus stimulating astrocytes to synthesise ApoE, which binds these excess lipids and distributes them appropriately for reuse in cell membrane biosynthesis.[88] This observation prompted Klaver *et al.* to speculate that a high degree of ApoE biosynthesis is required to support the high rate of photoreceptor renewal at the macula.[88] Indeed, it has been demonstrated that mice which were fed a high-fat diet, or which were deficient in ApoE, exhibit an increase in the thickness of BrM, which is seen in association with ageing and with AMD.[89]

Ishida *et al.* identified the presence of ApoE and lipids at the inner aspect of the RPE, and proposed that both compounds may be secreted by the RPE.[90] The role of ApoE in reverse cholesterol transport prompted the authors to suggest that this apolipoprotein may also facilitate the efflux of lipids from the RPE into the adjacent BrM, and they proposed a possible pathway for RPE cell-secreted lipids to cross BrM, where partially digested or undigested photoreceptor outer segments are secreted across the basal surface in association with ApoE. Subsequent binding with HDL at BrM may then facilitate desorption of the lipid particles into the circulation.[90]

In the retina ApoE is synthesised in Müller cells and in the RPE, and the presence of ApoE has been demonstrated in drusen.[81;91;92] It has been suggested, therefore, that age and/or disease-related disruption of normal ApoE function may result in the accumulation of lipoproteins at the interface between the RPE and BrM, consistent with observations that lipid deposits in drusen are largely composed of cholesteryl esters and unsaturated fatty acids.

These findings are consistent with the view that ApoE plays an important physiological role in the maintenance of macular health, and that an impaired ApoE system may affect the functional integrity of BrM. Furthermore, there is a biologically plausible rationale whereby the ApoE profile might influence the transport, capture, and stabilization of key compounds, such as L and Z, at the macula.

11. Lipoproteins, apolipoproteins and the retina

As noted previously, the ageing retina features changes in the RPE and BrM, which include changes in the lipoprotein and apolipoprotein composition of both structures. These changes may progress to the disease state of AMD. In recent times, evidence accrued from light microscopy, ultrastructural studies, lipid histochemistry, isolated lipoprotein assays, and gene expression analysis had led to the identification of many of the constituents that deposit in the RPE and BrM with age and AMD.[93] One of the universal changes that occurs with age is the development of BlamDs between the RPE and BrM.[11;12] This process may progress to the development of a 'lipid wall', mainly composed of neutral lipid deposits, decreasing the permeability of BrM and hindering metabolic activity between the RPE and BrM, preceding pathological changes associated with AMD.[10;93;94] When these deposits accumulate within the inner collagenous layer of BrM, they are referred to as basal linear deposits (BlinDs) and are a histopathological hallmark of AMD, which, when sufficiently large, can be recognised clinically as drusen.[13-15;95]

Much of the debris that accumulates in BrM in the form of BlinDs is composed of lipoproteins and lipoprotein particles.[14] It has been found that almost 60% of the total cholesterol within these lipoproteins is esterified cholesterol.[96] Furthermore, the esterified cholesterol within BrM was enriched between 16 and 40-fold compared to plasma. If these extracellular lipid deposits had been derived from plasma, more than 90% of the phospholipid would be phosphatidylcholine, whereas in actual fact, these lipoproteins are comprised of less than 50% phosphatidylcholine.[96] Indeed, the composition of drusen, which are essentially large BlinDs, has been shown to include esterified and unesterified cholesterol, and multiple apolipoproteins, including apolipoproteins B, A-I, C-I, C-II, and E, appearing with frequencies ranging from 100% (ApoE) to approximately 60% (A-I).[88;91;97;98] Interestingly, ApoC-III, although abundant in plasma, is present in fewer drusen (16.6%) than ApoC-I (93.1%), which is not present in plasma in large quantities, indicating either a specific retention of plasma-derived apolipoproteins within drusen, or an intraocular source for these apolipoproteins.[93] It is now understood that the majority of lipoproteins in BrM have undergone intracellular processing within the RPE prior to secretion as neutral lipids, mainly esterified cholesterol.[99;100] The RPE origin has been definitively shown by two groups using metabolic labelling and immunoprecipitation in rat-derived and human-derived RPE cell lines that were shown to secrete full-length ApoB.[101;102] This evidence is further strengthened by the finding of microsomal triglyceride transfer protein within native human RPE, indicating that the RPE is capable of secreting lipoprotein particles.[102] The pattern of lipid deposition in BrM with age, in which debris appears firstly in the elastic layer and then fills in towards the RPE, is also consistent with this lipid being primarily of RPE origin.[103]

The hydrophobic nature of the age-related thickening of BrM has been implicated in the aetiopathogenesis of AMD. In the case of Apo E, it is noteworthy that ApoE4 presents a positive charge relative to both ApoE2 and ApoE3. ApoE4 possesses arginine at residue 112 of the amino acid sequence, whereas ApoE3 possesses cysteine at this position, and in the case of ApoE2, the most frequent variant has cysteine instead of the normally occurring arginine at residue 158. Thus, ApoE3 presents a neutral charge, and ApoE2 a negative charge, relative to ApoE4.[53] Souied *et al.* suggested that this difference in charges between the ApoE isoforms may also contribute to differences in the clearance of debris through BrM.[104]

It appears that Müller cells are the most prominent biosynthetic sources of ApoE in the neural retina, and RPE cells are the most prominent sources in the RPE/choroid.[91] However, it remains unclear whether the concentration of ApoE in the cytoplasm of some RPE cells, especially those in close proximity to drusen, is the result of biosynthesis or selective accumulation. It has been shown that, in both the central and peripheral nervous systems, ApoE expression by astrocytes is up-regulated in response to neuronal injury and neuro-degenerative disease.[84;105;106] Indeed, there is evidence for ApoE up-regulation by Müller cells in degenerating human retina, where increased ApoE immuno-reactivity is found in the sub-retinal space of detached retinas[107] and in the Müller cells of retinas affected by glaucoma or AMD.[108] Furthermore, the relatively high levels of ApoE mRNA detected in the retina, especially in the eyes of older donors and in an individual with

documented AMD, support the view that up-regulation by retinal glia may be responsible for the observed increase in ApoE expression.[91]

12. Apo ϵ 4 allele status and AMD

ApoE gene status is believed to be a determinant of AMD risk.[88;104;109-111] The *ApoE* gene has three separate alleles: Apo ϵ 2, Apo ϵ 3 and Apo ϵ 4, resulting in six common phenotypes: three homozygous (ϵ 3 ϵ 3, ϵ 2 ϵ 2, ϵ 4 ϵ 4) and three heterozygous (ϵ 2 ϵ 3, ϵ 2 ϵ 4, ϵ 3 ϵ 4) phenotypes. The ϵ 4 allele has been found to be associated with a reduced risk of AMD, whereas the ϵ 2 allele has been associated with an increased risk of developing this disease.[88;104;109-113]

Due to the lack of cysteine residues at positions 112 and 158, preventing the formation of disulphide bridges with ApoA-II or other peptide components, the Apo ϵ 4 allele has an inability to form dimers. It has been suggested that this inability of the Apo ϵ 4 allele to form dimers, when compared with the Apo ϵ 2 and Apo ϵ 3 alleles, favours easier transport of lipids through BrM because of the smaller sized lipid particles, thus protecting against a loss of permeability of BrM.[104]

In the same way, it is possible that the neurosensory retina and the RPE respond to conditions of high oxidative injury by up-regulation of ApoE synthesis and/or accumulation, with implications for selective capture and stabilisation of L and Z in the retina.[91] It has been demonstrated that there is selective binding of certain receptors within the CNS to HDL particles enriched with ApoE, and that there is a lack of binding of these receptors to HDL particles deficient in ApoE.[114] Should this selectivity of the uptake mechanism be dependent on the ApoE polymorphism of the transporting lipoproteins, and given that the Apo ϵ 4 allele is putatively protective for AMD, it is tempting to hypothesise that retinal capture of L and Z may be related to apolipoprotein profile. In other words, the apolipoprotein composition as well as the lipoprotein profile, may play an important role in the transport and delivery of L and Z, and their subsequent accumulation and stabilisation within the retina.[115] Therefore, it is possible that the putative protective effect of the Apo ϵ 4 allele against AMD is attributable, at least in part, to the role its phenotypic expression (ApoE4) plays in the transport and delivery of the macular carotenoids to the retina, and to their stabilisation within the retina. Furthermore, recent research has shown an association between possession of at least one Apo ϵ 4 allele and higher levels of MP across the macula, which is consistent with the view that apolipoprotein profile influences the transport and/or retinal capture of the macular carotenoids.[74]

13. Conclusion

In conclusion, the role that lipoproteins and apolipoproteins play in the ageing eye and in the aetiopathogenesis of AMD is complex and, as yet, incompletely understood. Lipoproteins and apolipoproteins play an important role in the delivery of potentially protective nutrients from the digestive tract to the eye. The local ocular metabolic activity,

centred on the RPE and BrM, involves an exchange of nutrients from the choroidal circulatory system via BrM to the RPE and retina, with a reverse process whereby waste products are removed from the retina by the RPE through BrM in association with locally produced lipoproteins and apolipoproteins (particularly ApoB and ApoE). Unfortunately, over time it appears that these lipoproteins and apolipoproteins can accumulate between the RPE and BrM, and within BrM, leading to degradation in the metabolic efficiency between these two structures and the choroidal circulation. This deposition has been described as a 'lipid wall' and precedes the development of AMD.[93;94] Methods to detect and arrest or delay this process before it becomes clinically apparent and visually consequential to the patient have yet to be developed. Recent advances in our understanding of the lipoprotein and apolipoprotein molecular biology of the ageing and AMD-affected eye will help to direct future treatment strategies.[100]

Author details

Edward Loane

Department of Ophthalmology, Mater Misericordiae University Hospital, Dublin, Ireland

14. References

- [1] Snodderly DM, Brown PK, Delori FC, Auran JD. The Macular Pigment .1. Absorbance Spectra, Localization, and Discrimination from Other Yellow Pigments in Primate Retinas. *Investigative Ophthalmology & Visual Science* 1984;25(6):660-73.
- [2] Werner JS, Donnelly SK, Kliegl R. Aging and human macular pigment density. Appended with translations from the work of Max Schultze and Ewald Hering. *Vision Research* 1987;27:275-68.
- [3] Bok D. The retinal pigment epithelium: a versatile partner in vision. *J Cell Sci Suppl* 1993;17:189-95.:189-95.
- [4] Boulton M, yhaw-Barker P. The role of the retinal pigment epithelium: topographical variation and ageing changes. *Eye (Lond)* 2001 Jun;15(Pt 3):384-9.
- [5] Del Priore LV, Kuo YH, Tezel TH. Age-related changes in human RPE cell density and apoptosis proportion in situ. *Invest Ophthalmol Vis Sci* 2002 Oct;43(10):3312-8.
- [6] Dunaief JL, Dentchev T, Ying GS, Milam AH. The role of apoptosis in age-related macular degeneration. *Arch Ophthalmol* 2002 Nov;120(11):1435-42.
- [7] American Academy of Ophthalmology. Basic and Clinical Science Course, Section 2: Fundamentals and Principles of Ophthalmology. 2011.
- [8] Snell RS, Lemp MA. *Clinical Anatomy of the Eye*. Second ed. Wiley-Blackwell; 1998.
- [9] Marshall J. The ageing retina: physiology or pathology. *Eye (Lond)* 1987;1(Pt 2):282-95.
- [10] Zarbin MA. Current concepts in the pathogenesis of age-related macular degeneration. *Arch Ophthalmol* 2004;122(4):598-614.
- [11] Pauleikhoff D, Harper CA, Marshall J, Bird AC. Aging changes in Bruch's membrane. A histochemical and morphologic study. *Ophthalmology* 1990 Feb;97(2):171-8.

- [12] van der Schaft TL, de Bruijn WC, Mooy CM, de Jong PT. Basal laminar deposit in the aging peripheral human retina. *Graefes Arch Clin Exp Ophthalmol* 1993 Aug;231(8):470-5.
- [13] Curcio CA, Millican CL. Basal linear deposit and large drusen are specific for early age-related maculopathy. *Arch Ophthalmol* 1999 Mar;117(3):329-39.
- [14] Curcio CA, Presley JB, Millican CL, Medeiros NE. Basal deposits and drusen in eyes with age-related maculopathy: evidence for solid lipid particles. *Exp Eye Res* 2005 Jun;80(6):761-75.
- [15] Lommatzsch A, Hermans P, Muller KD, Bornfeld N, Bird AC, Pauleikhoff D. Are low inflammatory reactions involved in exudative age-related macular degeneration? Morphological and immunohistochemical analysis of AMD associated with basal deposits. *Graefes Arch Clin Exp Ophthalmol* 2008 Jun;246(6):803-10.
- [16] Bressler NM. Age-related macular degeneration is the leading cause of blindness. *JAMA* 2004 Apr 21;291(15):1900-1.
- [17] Congdon NG, Friedman DS, Lietman T. Important causes of visual impairment in the world today. *JAMA* 2003 Oct 15;290(15):2057-60.
- [18] Klein R, Wang Q, Klein BEK, Moss SE, Meuer SM. The Relationship of Age-Related Maculopathy, Cataract, and Glaucoma to Visual-Acuity. *Investigative Ophthalmology & Visual Science* 1995;36(1):182-91.
- [19] Owen CG, Jarrar Z, Wormald R, Cook DG, Fletcher AE, Rudnicka AR. The estimated prevalence and incidence of late stage age related macular degeneration in the UK. *Br J Ophthalmol* 2012 Feb 13.
- [20] Friedman DS, O'Colmain BJ, Munoz B, Tomany SC, McCarty C, de Jong PT, et al. Prevalence of age-related macular degeneration in the United States. *Arch Ophthalmol* 2004 Apr;122(4):564-72.
- [21] van Leeuwen R, Klaver CC, Vingerling JR, Hofman A, de Jong PT. Epidemiology of age-related maculopathy: a review. *Eur J Epidemiol* 2003;18(9):845-54.
- [22] Augustin A, Sahel JA, Bandello F, Dardennes R, Maurel F, Negrini C, et al. Anxiety and depression prevalence rates in age-related macular degeneration. *Invest Ophthalmol Vis Sci* 2007 Apr;48(4):1498-503.
- [23] Gupta OP, Brown GC, Brown MM. Age-related macular degeneration: the costs to society and the patient. *Curr Opin Ophthalmol* 2007 May;18(3):201-5.
- [24] Owen CG, Fletcher AE, Donoghue M, Rudnicka AR. How big is the burden of visual loss caused by age related macular degeneration in the United Kingdom? *Br J Ophthalmol* 2003 Mar 1;87(3):312-7.
- [25] Bandello F, Lafuma A, Berdeaux G. Public health impact of neovascular age-related macular degeneration treatments extrapolated from visual acuity. *Invest Ophthalmol Vis Sci* 2007 Jan;48(1):96-103.
- [26] Cruess AF, Zlateva G, Xu X, Soubrane G, Pauleikhoff D, Lotery A, et al. Economic burden of bilateral neovascular age-related macular degeneration: multi-country observational study. *Pharmacoeconomics* 2008;26(1):57-73.
- [27] Bird AC, Bressler NM, Bressler SB, Chisholm IH, Coscas G, Davis DM, et al. An international classification and grading system for age-related maculopathy and age-

- related macular degeneration. The International ARM Epidemiological Study Group. *Survey of Ophthalmology* 1995;39(5):367-74.
- [28] Gass JD. Pathogenesis of disciform detachment of the neuroepithelium. *Am J Ophthalmol* 1967 Mar;63(3):Suppl-139.
- [29] Sarks SH. Council Lecture. Drusen and their relationship to senile macular degeneration. *Aust J Ophthalmol* 1980 May;8(2):117-30.
- [30] Tomany SC, Wang HJ, van Leeuwen R, Klein R, Mitchell P, Vingerling JR, et al. Risk factors for incident age-related macular degeneration - Pooled findings from 3 continents. *Ophthalmology* 2004;111(7):1280-7.
- [31] Nolan JM, Stack J, O'Donovan O, Loane E, Beatty S. Risk factors for age-related maculopathy are associated with a relative lack of macular pigment. *Exp Eye Res* 2007 Jan;84(1):61-74.
- [32] Delcourt C, Michel F, Colvez A, Lacroux A, Delage M, Vernet MH, et al. Associations of cardiovascular disease and its risk factors with age-related macular degeneration: the POLA study. *Ophthalmic Epidemiology* 2001 Sep;8(4):237-49.
- [33] Seddon JM, Cote J, Davis N, Rosner B. Progression of age-related macular degeneration: associated with body mass index, waist circumference, and waist-hip ratio. *Arch Ophthalmol* 2003;121:785-92.
- [34] Tomany SC, Cruickshanks KJ, Klein R, Klein BEK, Knudtson MD. Sunlight and the 10-year incidence of age-related maculopathy - The Beaver Dam eye study. *Arch Ophthalmol* 2004;122(5):750-7.
- [35] SanGiovanni JP, Chew EY, Clemons TE, Ferris FL, III, Gensler G, Lindblad AS, et al. The relationship of dietary carotenoid and vitamin A, E, and C intake with age-related macular degeneration in a case-control study: AREDS Report No. 22. *Arch Ophthalmol* 2007 Sep;125(9):1225-32.
- [36] Hammond BR, Johnson MA. The Age-related Eye Disease Study (AREDS). *Nutrition Reviews* 2002;60(9):283-8.
- [37] Klein R, Klein BEK, Franke T. The Relationship of Cardiovascular-Disease and Its Risk-Factors to Age-Related Maculopathy - the Beaver Dam Eye Study. *Ophthalmology* 1993;100(3):406-14.
- [38] Hyman L, Schachat AP, He QM, Leske MC. Hypertension, cardiovascular disease, and age-related macular degeneration. *Arch Ophthalmol* 2000;118(3):351-8.
- [39] Tomany SC, Klein R, Klein BEK. The relationship between iris color, hair color, and skin sun sensitivity and the 10-year incidence of age-related maculopathy - The beaver dam eye study. *Ophthalmology* 2003;110(8):1526-33.
- [40] Klein R, Tomany SC, Cruickshanks KJ, Klein BEK. Sunlight and the 10-year incidence of age-related maculopathy. The Beaver Dam Eye Study. *Arch Ophthalmol* 2004 May;122(5):750-7.
- [41] Delcourt C, Carriere I, Delage M, Barberger-Gateau P, Schalch W. Plasma lutein and zeaxanthin and other carotenoids as modifiable risk factors for age-related maculopathy and cataract: the POLA Study. *Invest Ophthalmol Vis Sci* 2006 Jun;47(6):2329-35.

- [42] Sommerburg O, Keunen JEE, Bird AC, van Kuijk FJGM. Fruits and vegetables that are sources for lutein and zeaxanthin: the macular pigment in human eyes. *Br J Ophthalmol* 1998;82(8):907-10.
- [43] Bone RA, Landrum JT, Hime GW, Cains A, Zamor J. Stereochemistry of the Human Macular Carotenoids. *Investigative Ophthalmology & Visual Science* 1993;34(6):2033-40.
- [44] Johnson EJ, Neuringer M, Russell RM, Schalch W, Snodderly DM. Nutritional manipulation of primate retinas, III: effects of lutein or zeaxanthin supplementation on adipose tissue and retina of xanthophyll-free monkeys. *Investigative Ophthalmology Visual Science* 2005 Feb 1;46(2):692-702.
- [45] Bialostosky K, Wright JD, Kennedy-Stephenson J, McDowell M, Johnson CL. Dietary intake of macronutrients, micronutrients, and other dietary constituents: United States 1988-94. *Vital Health Stat* 11 2002 Jul;(245):1-158.
- [46] Bone RA, Landrum JT, Fernandez L, Tarsis SL. Analysis of the macular pigment by HPLC - Retinal distribution and age study. *Investigative Ophthalmology & Visual Science* 1988;29(6):843-9.
- [47] Snodderly DM, Handelman GJ, Adler AJ. Distribution of individual macular pigment carotenoids in central retina of macaque and squirrel monkeys. *Investigative Ophthalmology & Visual Science* 1991;32(2):268-79.
- [48] Snodderly DM. Evidence for Protection Against Age-Related Macular Degeneration by Carotenoids and Antioxidant Vitamins. *Am J Clin Nutr* 1995;62(6):S1448-S1461.
- [49] Liew SHM, Gilbert C, Spector TD, Mellerio J, Marshall J, van Kuijk FJGM, et al. Heritability of Macular Pigment: a Twin Study. *Investigative Ophthalmology & Visual Science* 2005;46(12):4430-6.
- [50] Loane E, Nolan JM, McKay GJ, Beatty S. The association between macular pigment optical density and CFH, ARMS2, C2/BF, and C3 genotype. *Exp Eye Res* 2011 Nov;93(5):592-8.
- [51] Loane E, Stack J, Beatty S, Nolan JM. Measurement of macular pigment optical density using two different heterochromatic flicker photometers. *Curr Eye Res* 2007 Jun;32(6):555-64.
- [52] Wooten BR, Hammond BR, Land RI, Snodderly DM. A practical method for measuring macular pigment optical density. *Investigative Ophthalmology & Visual Science* 1999;40(11):2481-9.
- [53] Mahley RW, Innerarity TL, Rall SC, Jr., Weisgraber KH. Plasma lipoproteins: apolipoprotein structure and function. *J Lipid Res* 1984 Dec 1;25(12):1277-94.
- [54] Durrington PN. Lipoproteins and their metabolism. In: Durrington PN, editor. *Hyperlipidaemia: Diagnosis and Management*. Butterworth-Heinemann Ltd; 1989.
- [55] Clevidence BA, Bieri JG. Association of carotenoids with human plasma lipoproteins. *Methods in Enzymology* 1993;214:33-46.
- [56] Erdman JW, Jr., Bieri TL, Gugger ET. Absorption and transport of carotenoids. *Ann N Y Acad Sci* 1993 Dec 31;691:76-85.
- [57] Goulinet S, Chapman MJ. Plasma LDL and HDL subspecies are heterogenous in particle content of tocopherols oxygenated and hydrocarbon carotenoids - Relevance to

- oxidative resistance and atherogenesis. *Arteriosclerosis Thrombosis and Vascular Biology* 1997;17(4):786-96.
- [58] Nolan J, O'Donovan O, Kavanagh H, Stack J, Harrison M, Muldoon A, et al. Macular pigment and percentage of body fat. *Investigative Ophthalmology Visual Science* 2004 Nov 1;45(11):3940-50.
- [59] Viroonudomphol D, Pongpaew P, Tungtrongchitr R, Changbumrung S, Tungtrongchitr A, Phonrat B, et al. The relationships between anthropometric measurements, serum vitamin A and E concentrations and lipid profiles in overweight and obese subjects. *Asia Pacific Journal of Clinical Nutrition* 2003;12(1):73-9.
- [60] Loane E, Nolan JM, Beatty S. The respective relationships between lipoprotein profile, macular pigment optical density, and serum concentrations of lutein and zeaxanthin. *Invest Ophthalmol Vis Sci* 2010 Nov;51(11):5897-905.
- [61] Hokanson JE, Austin MA. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996 Apr;3(2):213-9.
- [62] Morrison A, Hokanson JE. The independent relationship between triglycerides and coronary heart disease. *Vasc Health Risk Manag* 2009;5(1):89-95.
- [63] DeJager S, Bruckert E, Chapman MJ. Dense low density lipoprotein subspecies with diminished oxidative resistance predominate in combined hyperlipidemia. *J Lipid Res* 1993 Feb 1;34(2):295-308.
- [64] Connor WE, Duell PB, Kean R, Wang Y. The Prime Role of HDL to Transport Lutein into the Retina: Evidence from HDL-Deficient WHAM Chicks Having a Mutant ABCA1 Transporter. *Investigative Ophthalmology & Visual Science* 2007 Sep 1;48(9):4226-31.
- [65] Wang W, Connor SL, Johnson EJ, Klein ML, Hughes S, Connor WE. Effect of dietary lutein and zeaxanthin on plasma carotenoids and their transport in lipoproteins in age-related macular degeneration. *Am J Clin Nutr* 2007 Mar;85(3):762-9.
- [66] Cardinault N, Abalain JH, Sairafi B, Coudray C, Grolier P, Rambeau M, et al. Lycopene but not lutein nor zeaxanthin decreases in serum and lipoproteins in age-related macular degeneration patients. *Clin Chim Acta* 2005 Jul 1;357(1):34-42.
- [67] Klein R, Klein BEK, Tomany SC, Cruickshanks KJ. The association of cardiovascular disease with the long-term incidence of age-related maculopathy - The Beaver Dam Eye Study. *Ophthalmology* 2003;110(4):636-43.
- [68] Rizzo M, Berneis K. Low-density lipoprotein size and cardiovascular risk assessment. *QJM* 2006 Jan;99(1):1-14.
- [69] Snow KK, Seddon JM. Do age-related macular degeneration and cardiovascular disease share common antecedents? *Ophthalmic Epidemiology* 1999;6:125-43.
- [70] Klein R, Deng Y, Klein BE, Hyman L, Seddon J, Frank RN, et al. Cardiovascular disease, its risk factors and treatment, and age-related macular degeneration: Women's Health Initiative Sight Exam ancillary study. *Am J Ophthalmol* 2007 Mar;143(3):473-83.
- [71] Gale CR, Hall NF, Phillips DIW, Martyn CN. Lutein and zeaxanthin status and risk of age-related macular degeneration. *Investigative Ophthalmology & Visual Science* 2003;44(6):2461-5.

- [72] Brooks-Wilson A, Marcil M, Clee SM, Zhang LH, Roomp K, van DM, et al. Mutations in ABC1 in Tangier disease and familial high-density lipoprotein deficiency. *Nat Genet* 1999 Aug;22(4):336-45.
- [73] Beatty S, Nolan J, Kavanagh H, O'Donovan O. Macular pigment optical density and its relationship with serum and dietary levels of lutein and zeaxanthin. *Archives of Biochemistry and Biophysics* 2004;430(1):70-6.
- [74] Loane E, McKay GJ, Nolan JM, Beatty S. Apolipoprotein E genotype is associated with macular pigment optical density. *Invest Ophthalmol Vis Sci* 2010 May;51(5):2636-43.
- [75] Renzi LM, Hammond BR, Jr., Dengler M, Roberts R. The relation between serum lipids and lutein and zeaxanthin in the serum and retina: results from cross-sectional, case-control and case study designs. *Lipids Health Dis* 2012 Feb 29;11:33.:33.
- [76] Mahley RW, Innerarity TL. Lipoprotein receptors and cholesterol homeostasis. *Biochim Biophys Acta* 1983 May 24;737(2):197-222.
- [77] Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, et al. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. *Science* 1993 Aug 13;261(5123):921-3.
- [78] Abalain JH, Carre JL, Leglise D, Robinet A, Legall F, Meskar A, et al. Is age-related macular degeneration associated with serum lipoprotein and lipoparticle levels? *Clinica Chimica Acta* 2002 Dec;326(1-2):97-104.
- [79] Boerwinkle E, Utermann G. Simultaneous effects of the apolipoprotein E polymorphism on apolipoprotein E, apolipoprotein B, and cholesterol metabolism. *Am J Hum Genet* 1988 Jan;42(1):104-12.
- [80] Parra HJ, Arveiler D, Evans AE, Cambou JP, Amouyel P, Bingham A, et al. A case-control study of lipoprotein particles in two populations at contrasting risk for coronary heart disease. The ECTIM Study. *Arterioscler Thromb* 1992 Jun;12(6):701-7.
- [81] Shanmugaratnam J, Berg E, Kimerer L, Johnson RJ, Amaratunga A, Schreiber BM, et al. Retinal muller glia secrete apolipoproteins E and J which are efficiently assembled into lipoprotein particles. *Brain Res Mol Brain Res* 1997 Oct 15;50(1-2):113-20.
- [82] Utermann G, Langenbeck U, Beisiegel U, Weber W. Genetics of the apolipoprotein E system in man. *Am J Hum Genet* 1980 May;32(3):339-47.
- [83] Boyles JK, Zoellner CD, Anderson LJ, Kosik LM, Pitas RE, Weisgraber KH, et al. A role for apolipoprotein E, apolipoprotein A-I, and low density lipoprotein receptors in cholesterol transport during regeneration and remyelination of the rat sciatic nerve. *J Clin Invest* 1989 Mar;83(3):1015-31.
- [84] Poirier J. Apolipoprotein E in animal models of CNS injury and in Alzheimer's disease. *Trends Neurosci* 1994 Dec;17(12):525-30.
- [85] Gregg RE, Zech LA, Schaefer EJ, Stark D, Wilson D, Brewer HB, Jr. Abnormal in vivo metabolism of apolipoprotein E4 in humans. *J Clin Invest* 1986 Sep;78(3):815-21.
- [86] Ehnholm C, Mahley RW, Chappell DA, Weisgraber KH, Ludwig E, Witztum JL. Role of apolipoprotein E in the lipolytic conversion of β -very low density lipoproteins to low density lipoproteins in type III hyperlipoproteinemia. *PNAS* 1984 Sep 1;81(17):5566-70.

- [87] Leon AS, Togashi K, Rankinen T, Despres JP, Rao DC, Skinner JS, et al. Association of apolipoprotein E polymorphism with blood lipids and maximal oxygen uptake in the sedentary state and after exercise training in the HERITAGE family study. *Metabolism* 2004 Jan;53(1):108-16.
- [88] Klaver CC, Kliffen M, van Duijn CM, Hofman A, Cruts M, Grobbee DE, et al. Genetic association of apolipoprotein E with age-related macular degeneration. *Am J Hum Genet* 1998 Jul;63(1):200-6.
- [89] Ong JM, Zorapapel NC, Rich KA, Wagstaff RE, Lambert RW, Rosenberg SE, et al. Effects of cholesterol and apolipoprotein E on retinal abnormalities in apoE-deficient mice. *Investigative Ophthalmology & Visual Science* 2001 Jul 1;42(8):1891-900.
- [90] Ishida BY, Bailey KR, Duncan KG, Chalkley RJ, Burlingame AL, Kane JP, et al. Regulated expression of apolipoprotein E by human retinal pigment epithelial cells. *J Lipid Res* 2004 Feb 1;45(2):263-71.
- [91] Anderson DH, Ozaki S, Nealon M, Neitz J, Mullins RF, Hageman GS, et al. Local cellular sources of apolipoprotein E in the human retina and retinal pigmented epithelium: implications for the process of drusen formation. *American Journal of Ophthalmology* 2001 Jun;131(6):767-81.
- [92] Dentchev T, Milam AH, Lee VM, Trojanowski JQ, Dunaief JL. Amyloid-beta is found in drusen from some age-related macular degeneration retinas, but not in drusen from normal retinas. *Mol Vis* 2003 May 14;9:184-90.
- [93] Curcio CA, Johnson M, Huang JD, Rudolf M. Apolipoprotein B-containing lipoproteins in retinal aging and age-related macular degeneration. *J Lipid Res* 2010 Mar;51(3):451-67.
- [94] Ruberti JW, Curcio CA, Millican CL, Menco BP, Huang JD, Johnson M. Quick-freeze/deep-etch visualization of age-related lipid accumulation in Bruch's membrane. *Invest Ophthalmol Vis Sci* 2003 Apr;44(4):1753-9.
- [95] Sarks S, Cherepanoff S, Killingsworth M, Sarks J. Relationship of Basal laminar deposit and membranous debris to the clinical presentation of early age-related macular degeneration. *Invest Ophthalmol Vis Sci* 2007 Mar;48(3):968-77.
- [96] Curcio CA, Millican CL, Bailey T, Kruth HS. Accumulation of cholesterol with age in human Bruch's membrane. *Invest Ophthalmol Vis Sci* 2001 Jan;42(1):265-74.
- [97] Malek G, Li CM, Guidry C, Medeiros NE, Curcio CA. Apolipoprotein B in Cholesterol-Containing Drusen and Basal Deposits of Human Eyes with Age-Related Maculopathy. *Am J Pathol* 2003 Feb 1;162(2):413-25.
- [98] Li CM, Clark ME, Chimento MF, Curcio CA. Apolipoprotein localization in isolated drusen and retinal apolipoprotein gene expression. *Invest Ophthalmol Vis Sci* 2006 Jul;47(7):3119-28.
- [99] Ebrahimi KB, Handa JT. Lipids, lipoproteins, and age-related macular degeneration. *J Lipids* 2011;2011:802059. Epub;2011 Jul 28.:802059.
- [100] Curcio CA, Johnson M, Rudolf M, Huang JD. The oil spill in ageing Bruch membrane. *Br J Ophthalmol* 2011 Dec;95(12):1638-45.

- [101] Wu T, Fujihara M, Tian J, Jovanovic M, Grayson C, Cano M, et al. Apolipoprotein B100 secretion by cultured ARPE-19 cells is modulated by alteration of cholesterol levels. *J Neurochem* 2010 Sep;114(6):1734-44.
- [102] Li CM, Presley JB, Zhang X, Dashti N, Chung BH, Medeiros NE, et al. Retina expresses microsomal triglyceride transfer protein: implications for age-related maculopathy. *J Lipid Res* 2005 Apr;46(4):628-40.
- [103] Huang JD, Presley JB, Chimento MF, Curcio CA, Johnson M. Age-related changes in human macular Bruch's membrane as seen by quick-freeze/deep-etch. *Exp Eye Res* 2007 Aug;85(2):202-18.
- [104] Souied EH, Benlian P, Amouyel P, Feingold J, Lagarde JP, Munnich A, et al. The epsilon4 allele of the apolipoprotein E gene as a potential protective factor for exudative age-related macular degeneration. *Am J Ophthalmol* 1998 Mar;125(3):353-9.
- [105] Mouchel Y, Lefrancois T, Fages C, Tardy M. Apolipoprotein E gene expression in astrocytes: developmental pattern and regulation. *Neuroreport* 1995 Dec 29;7(1):205-8.
- [106] Snipes GJ, McGuire CB, Norden JJ, Freeman JA. Nerve injury stimulates the secretion of apolipoprotein E by nonneuronal cells. *PNAS* 1986 Feb 15;83(4):1130-4.
- [107] Schneeberger SA, Iwahashi CK, Hjelmeland LM, Davis PA, Morse LS. Apolipoprotein E in the subretinal fluid of rhegmatogenous and exudative retinal detachments. *Retina* 1997;17(1):38-43.
- [108] Kuhrt H, Hartig W, Grimm D, Faude F, Kasper M, Reichenbach A. Changes in CD44 and ApoE immunoreactivities due to retinal pathology of man and rat. *J Hirnforsch* 1997;38(2):223-9.
- [109] Baird PN, Guida E, Chu DT, Vu HTV, Guymer RH. The ϵ 2 and ϵ 4 alleles of the apolipoprotein gene are associated with age-related macular degeneration. *Investigative Ophthalmology & Visual Science* 2004 May 1;45(5):1311-5.
- [110] Simonelli F, Margaglione M, Testa F, Cappucci G, Manitto MP, Brancato R, et al. Apolipoprotein E polymorphisms in age-related macular degeneration in an Italian population. *Ophthalmic Res* 2001 Nov;33(6):325-8.
- [111] Zarepari S, Reddick AC, Branham KEH, Moore KB, Jessup L, Thoms S, et al. Association of apolipoprotein E alleles with susceptibility to age-related macular degeneration in a large cohort from a single center. *Investigative Ophthalmology & Visual Science* 2004;45(5):1306-10.
- [112] Bojanowski CM, Shen D, Chew EY, Ning B, Csaky KG, Green WR, et al. An apolipoprotein E variant may protect against age-related macular degeneration through cytokine regulation. *Environ Mol Mutagen* 2006 Oct;47(8):594-602.
- [113] Fritsche LG, Freitag-Wolf S, Bettecken T, Meitinger T, Keilhauer CN, Krawczak M, et al. Age-related macular degeneration and functional promoter and coding variants of the apolipoprotein E gene. *Hum Mutat* 2008 Dec 18.
- [114] Stewart JE, Skinner ER, Best PV. Receptor binding of an apolipoprotein E-rich subfraction of high density lipoprotein to rat and human brain membranes. *The International Journal of Biochemistry & Cell Biology* 1998 Mar 1;30(3):407-15.

- [115] Schneider WJ, Kovanen PT, Brown MS, Goldstein JL, Utermann G, Weber W, et al. Familial dysbetalipoproteinemia. Abnormal binding of mutant apoprotein E to low density lipoprotein receptors of human fibroblasts and membranes from liver and adrenal of rats, rabbits, and cows. *J Clin Invest* 1981 Oct;68(4):1075-85.

IntechOpen

IntechOpen