An update on RPE cell senescence as a key contributor to age-related macular degeneration: support from current and experimental treatments

Michael R. Kozlowski [1]

[1] *Arizona College of Optometry, Midwestern University, Glendale, AZ, USA*

ABSTRACT

Background: Age-related macular degeneration (AMD) is a major cause of vision loss. Its prevalence has increased over the past decade. This increase is partly due to the scarcity of preventive and therapeutic interventions for this disorder, except when it is in its advanced neovascular form. Discovery of effective treatments for AMD is complicated by the multifactorial pathology of the disorder. Thus, it is difficult to determine which potential disease mechanism to target in order to achieve the greatest clinical benefit.

Hypothesis: Over a decade ago, it was hypothesized that many of the pathologies observed in AMD stem from retinal pigment epithelial (RPE) cell senescence. This provided a potentially key mechanistic target. Supporting this hypothesis, many of the agents that were in development or clinical use for AMD at that time influenced RPE cell senescence, although they were not utilized for this specific effect. The present article re-evaluates this hypothesis by exploring the logical prediction that if RPE cell senescence is a key contributor to AMD, then inhibition of RPE cell senescence is important in the treatment of AMD. If this hypothesis holds true, the inhibition or reversal of RPE cell senescence or its effects should be a common characteristic of new treatments for AMD.

Conclusions: Over the past decade, many agents have been investigated for the treatment of AMD. Although a few were designed to address cell senescence, the majority targeted other potential pathological mechanisms. In support of our original hypothesis, we now present evidence that many of the newer agents investigated for the treatment of AMD, even those that were not meant to reduce cell senescence or its effects, have this function as part of their activity profiles. Further experimental studies or clinical trials exploring the safety and efficacy of inhibiting RPE cell senescence or reversing its effects are needed to pave the way for improved AMD treatment.

KEYWORDS

age-related macular degeneration, retinal pigment epithelium, cell senescence, VEGF, vascular endothelial growth factor, dry AMD, exudative AMD, wet macular degeneration, geographic atrophy, macular degeneration

Correspondence: Michael R. Kozlowski, Midwestern University, Arizona College of Optometry, 19555 North 59th Avenue, Glendale, Arizona 85308. Email: mkozlo@midwestern.edu. ORCID iD: https://orcid.org/0000-0003-1213-7015

INTRODUCTION

Currently, the worldwide incidence and progression rate of age-related macular degeneration (AMD) are approximately 2% and 6%, respectively [1]. AMD treatments have had variable success rates over the past decade. Fortunately, the prevalence of blindness due to AMD decreased between 2010 and 2020 [2, 3]. However, the prevalence of moderate-to-severe vision impairment due to AMD increased during the same period [2, 3]. The increase in vision impairment reflects the limited development of effective treatments for most forms of the disorder, particularly the early stages (Figure 1A, B), whereas the decrease in blindness largely reflects the growing use of new drugs to treat the neovascular stage of AMD (Figure 1C, D) [2, 3].

In fact, only three of the clinical stages of AMD have approved or accepted treatments [4-9]. One stage is *advanced neovascular AMD*, which can be treated using several available medications, including vascular endothelial growth factor (VEGF) inhibitors, and other treatments are under development [4-8, 10-12]. Another is *advanced AMD with geographic atrophy (GA)*, for which a new drug has been approved [13]. Lastly, a dietary supplement with antioxidant vitamins, carotenoids, and zinc can decrease the risk of progression from intermediate to advanced AMD by approximately 30% [14, 15].

Cell senescence was first described by Hayflick and Moorhead in 1961 as the loss of replicative capacity in cell cultures that have undergone a large number of subcultivations [16]. The trigger for this phenomenon was discovered to be a critical level of telomere shortening after many cell divisions due to the end-replication problem [17]. Many additional triggers for cell senescence were subsequently identified, including genomic damage, aberrant oncogenic signaling, mitochondrial dysfunction, disrupted nicotinamide adenine dinucleotide metabolism, hyperglycemia, and oxidative stress [16, 18]. Oxidative stress may be a particularly important driver of senescence in tissues that produce high levels of reactive oxidative species, such as the retina [19-21].

Although the phenotypic changes that occur in cell senescence may differ somewhat based on cell type, triggering mechanism, and the cellular environment, some features are nearly ubiquitous and have been used as markers for senescence [16, 18]. These features include decreased cell replication, increased levels of the cyclin-dependent kinase inhibitors p16 and p21, adoption of a pro-inflammatory secretory profile called the senescence-associated secretory phenotype (SASP), adoption of an enlarged and flattened morphology, and the presence of lysosomal dysfunction. Lysosomal dysfunction, often measured as an increase in senescence-associated beta-galactosidase activity (SABG), is a standard indicator of cell senescence [16, 22].
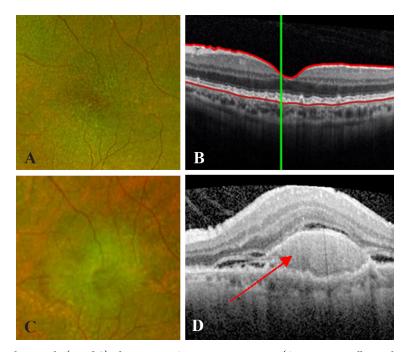

Figure 1. Fundus photographs (A and C) taken using an Optos Daytona™ camera (Optos, Inc., Marlborough, MA) and macular optical coherence tomography (OCT) images (B and D) (Heidelberg Spectralis HRA + OCT; Heidelberg Engineering Inc., Franklin, MA) in a patient with intermediate age-related macular degeneration (AMD) (A and B) that later converted to advanced, wet AMD (C and D). In intermediate AMD, drusen appeared as light dots on the fundus photograph (A) and as elevations of the retinal pigment epithelial cell layer (white layer above the lower red line) in the OCT image (B). In advanced AMD (C and D), a choroidal neovascular membrane (D, red arrow) can be seen. Lines and arrows have been added to the OCT images for illustrative purposes

Another characteristic of senescent cells is their persistence in the tissues in which they occur. This durability appears to be achieved through the inhibition of apoptotic processes that normally operate to eliminate compromised cells [23, 24]. Given their altered morphological and functional characteristics, senescent cells within a tissue can negatively impact tissue function [16, 18, 24]. Furthermore, the chronic release of pro-inflammatory mediators by these cells can damage or kill adjacent non-senescent cells [16, 18, 24].

AMD is closely associated with dysfunction of the retinal pigment epithelial (RPE) cell layer [25-27]. For example, the loss of pigment uniformity observed early in the disorder is partially due to pigmentary changes in the RPE cells [26-28]. In addition, the drusen that occurs in AMD contains material from the photoreceptor outer segments, indicating improper processing by RPE cells [26-29]. Furthermore, a complete loss of RPE cells occurs over large retinal areas in the advanced GA stage of AMD [25, 26, 28]. Changes in RPE function are also linked to the other advanced form, neovascular AMD (nAMD), in which choroidal blood vessels enter the retina, causing severe structural damage and significant vision impairment [26]. The growth of these vessels into the retina is supported by VEGF and requires a breach of the choroidal-retinal barrier [11, 26, 30]. Since RPE cells participate in both VEGF production and maintenance of the choroidal-retinal barrier, their dysfunction is implicated in the development of neovascularization [11, 26, 30, 31]. Many possible causes have been proposed for the RPE cell dysfunction seen in AMD, including abnormal complement activity, inflammation, mitochondrial damage, oxidative stress, and RPE cell senescence [4, 6, 7, 27, 32, 33].

Cell senescence is thought to contribute to the pathologies of several age-related disorders, including Alzheimer disease, cardiovascular disease, diabetes, frailty, multiple sclerosis, Parkinson disease, and pulmonary fibrosis [16, 18, 24, 34-39]. The contribution of cell senescence to age-related pathology is supported by the demonstration that senolytic agents, which eliminate senescent cells from tissues, can improve health and function in aging humans and animal models of age-related human disorders [16, 18, 37, 38]. Since the late 1990s, several investigators have suggested that RPE cell senescence plays a role in the pathology of AMD [33, 40-43].

HYPOTHESIS

Over a decade ago, senescence of RPE cells was hypothesized as a key contributor to AMD pathology [33]. Three arguments were made in support of this hypothesis. First, the changes in cell physiology that accompany senescence are consistent with pathological changes seen in AMD [33]. Second, there are significant parallels between AMD risk factors and causes of cell senescence [33]. Third, many of the clinical and experimental treatments for AMD known at that time were able to reduce RPE cell senescence or its effects [33].

The present article re-evaluates this hypothesis by exploring the logical prediction that if RPE cell senescence is a key contributor to AMD, then inhibition of RPE cell senescence is important in the treatment of AMD. If this hypothesis holds true, the inhibition or reversal of RPE cell senescence or its effects should be a common characteristic of new treatments for AMD.

EVALUATION OF THE HYPOTHESIS

To evaluate the prediction that the inhibition of RPE cell senescence is important for the treatment of AMD, the actions of a group of agents that have been discovered and, in some cases, developed for treating AMD are presented below. These agents were generally introduced over the past decade. The influence of these substances on cellular senescence is also discussed.

Metformin

In recent years, four separate retrospective studies found a significant decrease in the incidence of AMD in patients using metformin to treat diabetes and other metabolic disorders [44-47]. Two other studies did not observe this association; however, this may have been due to the study designs, as one used a short dosing duration for metformin [48], and the other included some patients younger than those in which AMD normally occurs [49]. Based on these data, metformin is currently undergoing phase 2 clinical trials for GA [50].

Considerable evidence suggests that cell senescence plays a role in the pathology of type 2 diabetes [38, 51-58]. Senescence produces cellular changes that can impair the functioning of tissues in which the senescent cells reside, as discussed above. Cell senescence appears to be associated with decreased insulin release by the pancreas, adipose tissue dysfunction, and insulin resistance [51-58]. Moreover, the removal of senescent cells reduces diabetic pathology [38].

Although different classes of antidiabetic drugs target diverse physiological mechanisms underlying diabetes, a remarkable number, including biguanides, gliflozins, sulfonylureas, and thiazolidinediones, can also reduce cell

senescence [59-69]. Given the potential involvement of senescence in type 2 diabetes, the ability of antidiabetic medications to reduce cell senescence has been suggested to play a role in their therapeutic efficacies [38, 51-58]. We recently found that metformin inhibits senescence in RPE cells exposed to oxidative stress, as measured by reduced p21 levels and decreased SABG staining [70]. Thus, the ability of metformin to reduce AMD incidence may be at least partly due to its ability to reduce RPE cell senescence [71, 72].

Brimonidine
Brimonidine tartrate, delivered through a biodegradable intravitreal implant, reduces the rate of lesion growth in patients with GA [4, 73]. Brimonidine is an alpha-2 adrenergic receptor agonist used to treat glaucoma and has neuroprotective effects in several models of ocular injury [74]. Part of this protective activity may be mediated by increased levels of fibroblast growth factor [74], a peptide that reduces senescence in several cell types, although its effects on RPE cell senescence have not been reported [74-76]. Thus, the anti-GA effect of brimonidine may be indirect, at least partly, due to reduction in cell senescence.

Complement pathway inhibitors
Complement pathway activation has been implicated in the pathology of AMD, and certain polymorphisms that decrease the activity of complement factor H, a negative regulator of the complement pathway, are genetic risk factors for AMD [33, 77]. Several agents that inhibit the complement pathway have been explored for treatment of AMD [4-7]. One such drug, pegcetacoplan (Syfovre™, a C3 inhibitor), was recently approved by the FDA for treatment of GA [13]. Others that are still in, or have been in, clinical trials include APC-1905 (anti-C5 aptamer), eculizumab (anti-C5 monoclonal antibody [mAb]), lampalizumab (anti-CFD mAb), LGF316 (anti-C5 antibody), and avacincaptad pegol (Zimura™, a C3 inhibitor) [4-7].

The complement pathway may be activated in AMD by the materials present in drusen, which are themselves linked to RPE cell senescence [33, 77]. Alternatively, the presence of apoptotic or necrotic cells may lead to pathway activation [77]. Although senescent RPE cells are resistant to apoptosis [23, 24], they may cause the death of surrounding cells by releasing immunomodulators [16, 18, 24]. Thus, while not directly inhibiting RPE cell senescence, complement pathway inhibitors may act in part by reducing its effects.

Senolytic agents such as UBX1325
If senescent cells within a tissue produce pathological effects, then removal of the cells or their influence should alleviate these effects. Senolytic agents selectively clear senescent cells from tissues [23, 78]. Senolytics generally counteract the resistance of senescent cells to apoptosis [23, 78]. Two senolytic agents used to evaluate the contribution of cell senescence to AMD are Nutlin-3a and UBX1325 [79-81]. Nutlin-3a antagonizes the mouse double minute 2 protein, which blocks the action of the proapoptotic tumor suppressor p53 [82]. UBX1325 is an inhibitor of Bcl-xL, a pro-survival member of the BCL-2 family of proteins that regulates commitment to apoptosis [83].

The effects of Nutlin-3a have been examined in animal models of three clinical stages of AMD: early-to-intermediate, GA, and nAMD [80, 81]. In each case, Nutlin-3a decreased the number of senescent RPE cells, as well as the signs of AMD. Furthermore, Nutlin-3a increased visual function under each condition, as assessed using electroretinography. Bhisitkul et al. demonstrated that a single intravitreal injection of UBX1325 improved visual function in patients with nAMD for at least 12 weeks [79]. The effectiveness of senolytic compounds in AMD is consistent with a role of RPE cell senescence in the disorder.

Sirolimus (rapamycin)
In a recent pilot study in patients with refractory nAMD, sirolimus was significantly more effective than anti-VEGF treatment in reducing retinal pathology [84]. Sirolimus is a senomorphic agent, in that it suppresses the effects of senescent cells on the surrounding normal cells, whereas senolytics eliminate the cells [23, 78]. Sirolimus reduces the SASP, and thereby, its damaging effects [85, 86]. The senomorphic effects of sirolimus extend to RPE cells [87, 88].

One component of the SASP is increased secretion of interleukin-6 (IL-6). This phenomenon occurs in many cell types, including RPE cells [89-92]. Increased IL-6 levels have been linked to AMD; elevated IL-6 levels were found in the serum of patients with GA or nAMD as well as in the eyes of patients with nAMD [91, 93-95]. In addition, systemic IL-6 levels are correlated with the progression of AMD [93, 96]. At least a fraction of the IL-6 expressed in the retina originates from RPE cells [97]. Increased IL-6 levels are thought to promote AMD via pro-inflammatory activity [93, 94, 96]. Sirolimus decreases this effect by reducing the SASP [85, 86].

Risuteganib

Integrins are a family of cell adhesion receptors. They exist as heterodimers composed of one of 18 alpha subunits combined with one of eight beta subunits [98]. In addition to facilitating attachment to the extracellular matrix, integrins have signaling activities that regulate complex cellular processes, including adhesion, migration, proliferation, and survival [99]. Certain functions of integrins, such as increasing inflammation and responsiveness to oxidative stress, originally suggested a role of integrin inhibitors in treating AMD [100]. The integrin inhibitor risuteganib (Luminate™) was recently reported in a phase 2 clinical study to significantly improve visual acuity in patients with intermediate-stage AMD [100]. This efficacy profile is different from that of the Age Related Eye Disease Study Research Group (AREDS) dietary supplements, which decrease the odds of progression from intermediate to advanced AMD but do not improve visual acuity [101].

Integrin signaling is partly mediated by integrin-linked kinase (ILK) [102]. In rat kidney cells, an elevation in ILK level occurs with cell aging and is correlated with markers of cell senescence, including increased SABG activity and reduced proliferative capacity [103]. In cardiac fibroblasts from young rats, ILK overexpression produces several signs of cell senescence, including increased cell size, reduced proliferative capacity, elevated p21 levels, and increased SABG activity. Conversely, in cardiac fibroblasts from older rats, ILK knockdown prevents these senescence-associated changes [104]. In mouse renal cells, oxidative stress, an inducer of cell senescence, increases ILK expression and activity, as well as p16 levels and SABG activity. Ectopic expression of ILK, even in the absence of oxidative stress, increases p16 levels. Conversely, ILK downregulation prevents the elevation of p16 levels caused by oxidative stress [105].

Integrin interacts with ILK through the cytoplasmic domains of β1 and β3 subunits [106]. In rat kidney cells, aging increases the association of integrin β1 with ILK [103]. Risuteganib inhibits the activity of two integrin heterodimers that contain the β1 or β3 subunits (α5β1, αVβ3) [100]. The reduction in ILK activity resulting from this inhibition is expected to decrease senescence in RPE cells, as in other cells [104]. Thus, reduced RPE senescence may contribute to the efficacy of risuteganib in treating dry AMD.

AKST4290

The C-C motif chemokine receptor 3 (CCR3) antagonist, AKST4290, improved best-corrected visual acuity in a small trial involving patients with newly diagnosed nAMD [107]. This effect was attributed to inhibition of the stimulatory effect of CCR3 receptor activation on angiogenesis [107, 108]. Interestingly, one of the natural ligands of CCR3, eotaxin-1, is associated with several aging disorders, although its direct effect on cell senescence has not yet been reported [109]. However, another CCR3 antagonist (CCR3A) reportedly inhibits senescence in periodontal cells [110]. If the anti-senescence effects of CCR3 antagonists extend to RPE cells, this may contribute to the anti-AMD activity of AKST4290.

VEGF inhibitors

Drugs that inhibit VEGF or lower the levels of VEGF signaling are quite effective in treating nAMD [111]. Over the past ten years, many new agents and technologies in this class have been introduced [7, 8, 12]. The most common pathology of nAMD is extension of choroidal blood vessels into the retina [25, 26, 111]. This vessel growth is thought to be caused by increased VEGF levels [111]. In support of this idea, elevated VEGF levels were found in the intraocular compartments of patients with AMD compared to those of age-matched controls [112, 113].

Choroidal neovascularization requires not only the growth of new choroidal blood vessels but also their penetration through the boundary between the choroid and retina, which is formed by Bruch's membrane and the RPE layer. The importance of this breach is highlighted by the finding that artificial breaks in Bruch's membrane and the RPE layer are themselves sufficient to produce choroidal neovascularization in animal models of nAMD [114-116]. In addition to supporting blood vessel growth, VEGF decreases the trans-epithelial electrical resistance (TEER) of RPE layers *in vitro*, which is a sign of reduced integrity in the RPE layer [117]. Thus, VEGF inhibitors may also prevent choroidal neovascularization by maintaining the barrier between the choroid and retina.

The elevation in VEGF levels that stimulates the growth of new choroidal blood vessels in AMD may be a result of RPE cell senescence. Several studies have demonstrated that RPE cell senescence increases with aging *in situ* and that RPE cells of aged donors have increased VEGF expression compared with those of younger donors [118]. In addition, primary RPE cells with oxidative stress-induced senescence show increased expression of VEGF [119]. As an increase in VEGF levels is also observed in several other types of senescent human cells, it may be among the sequelae of cell senescence [120, 121].

A weakened choroid-RPE barrier, which permits the entry of choroidal blood vessels into the retina, may also be caused by RPE cell senescence. We found that the reduction in TEER caused by VEGF is greater in RPE layers formed by cells approaching senescence [122]. This suggests that the integrity of RPE layers containing senescent cells may be more easily disrupted by VEGF. Thus, although VEGF inhibitors do not directly influence RPE cell senescence, their reduction in VEGF signaling would inhibit many effects of senescence on the retina.

The AREDS-2 nutritional supplement
A nutritional supplement cocktail containing beta-carotene, vitamins C and E, and zinc, developed by AREDS, reduces the progression of AMD from intermediate to advanced stages by approximately 30% [101]. The potential for this decrease to be mediated by a reduction in RPE cell senescence was discussed in a previous article [33]. Beta-carotene, vitamins C and E, and zinc all have antioxidant activity and could prevent RPE cell senescence by reducing oxidative stress, an inducer of cell senescence [123]. In addition, zinc can increase telomerase activity, thereby decreasing telomere shortening, another inducer of cell senescence [124].

A new version of this nutritional supplement, called AREDS2, has been developed [15]. This new formulation was based on a large study examining the effect of several changes to the original AREDS formulation, including the addition of macular pigments (lutein and zeaxanthin) and omega-3 fatty acids (docosahexaenoic acid and eicosapentaenoic acid) [15]. These additions, individually and in combination, did not improve the efficacy of the original formulation. At first glance, this seems unsupportive of the importance of anti-senescence activity in treating AMD because both omega-3 fatty acids and macular pigments have antioxidant effects that can protect against senescence, and omega-3 fatty acids can activate telomerase [33, 125, 126]. However, because the constituents of the original AREDS formulation already have high concentrations of both antioxidant and telomerase-activating agents, adding other constituents with the same activities may not produce a noticeable change in efficacy [33].

RPE cell senescence as a key contributor to AMD
Designating RPE cell senescence as a "key contributor" to AMD [33] implies that many of the pathological processes that occur in AMD are rooted in RPE cell senescence. One pillar of support for the original hypothesis was that many of the effective agents against AMD, at that time, had the potential to reduce RPE cell senescence. In the past ten years, several new agents have been discovered and investigated, and in some cases, have been clinically developed for the treatment of AMD [4-9]. Based on the original hypothesis, it seems logical that many of these newer anti-AMD agents would also exhibit anti-senescence activity. The present review of the most significant of these agents seems to support this prediction (Table 1).

Perhaps the most direct indication of the importance of inhibiting RPE senescence in the treatment of AMD comes from the experimental and clinical effectiveness of senolytic and senomorphic agents [79-81]. The senolytic agent Nutlin-3a was effective against many stages of AMD in animal models and eliminated senescent RPE cells *in vivo* [79-81]. The senolytic compound UBX1325 was active against nAMD in a phase 2 clinical trial [79-81], and the senomorphic agent sirolimus was also active against nAMD in a clinical trial [84]. For these compounds, a reduction in the impact of senescent cells is the primarily known element of their activity profiles.

Table 1. Agents with activity against AMD and their effects on cell senescence (see **EVALUATION OF THE HYPOTHESIS**)

Drug or drug class	Reduction in cell senescence	Reduction in the effects of cell senescence	Activity demonstrated in RPE cells
Metformin [44-47, 50, 70-72]	✓		✓
Brimonidine [4, 73-76]	✓		
Complement pathway inhibitors [4-7, 13]	✓	✓	
Senolytics [79-81]		✓	
Sirolimus [23, 78, 84-88]		✓	✓
Risuteganib [100, 104]	✓		
AKST4290 [107, 108, 110]	✓		
VEGF inhibitors [122]		✓	
AREDS nutritional supplement [123, 124]	✓		

Abbreviations: AMD, age-related macular degeneration; RPE, retinal pigment epithelium; Brimonidine, an alpha-2 adrenergic receptor agonist; Risuteganib, the integrin inhibitor; AKST4290, the C-C motif chemokine receptor 3 antagonist; VEGF, vascular endothelial growth factor; AREDS, age-related eye disease study research group. Note: The "✓" tick symbol indicates a reported drug action.

Circumstantial support for the hypothesis that inhibition of cell senescence is a key to the treatment of AMD comes from the sheer number of agents effective against AMD that also have anti-senescence effects, several of which have been reviewed in this article. Perhaps metformin currently has the highest profile [70-72]. Several studies have reported that patients receiving metformin have a reduced incidence of AMD [44-47]. Metformin is known to have anti-senescence activity, and this activity has been proposed to contribute to its antidiabetic effects [62, 65]. Recently, the anti-senescence activity of metformin was shown to extend to RPE cells [70].

Although the hypothesis that RPE cell senescence is a key contributor to AMD has not been disproven by an additional decade of research, it has not yet achieved ultimate validation, which will occur only when anti-senescence activity is correlated with anti-AMD activity. Although the realm of senolytic and senomorphic compounds is relatively new [23, 78], early clinical results may be the first step in this validation. However, many hurdles remain in developing a therapy for AMD based on the inhibition of RPE cell senescence or its effects. The most significant is that the hypothesis may be incorrect. However, the data presented in the current report, as well as those from an earlier study [33], do not appear to suggest this. Another hurdle is to inhibit RPE cell senescence with sufficient effectiveness and selectivity to produce a clinically safe and useful effect. To overcome this hurdle, additional research may be needed to gain a better understanding of cell senescence as it occurs in RPE cells, and how to prevent it. An additional obstacle is that cell senescence may be a trigger for AMD but not a sustaining factor. If so, inhibiting cell senescence may not be valuable in treating the disorder once it is established, but it may still have prophylactic use.

Nonetheless, the potential value of senescence-based therapies in treating AMD justifies overcoming the hurdles faced during their development. For example, if RPE cell senescence is foundational or contributes to many other pathological processes in AMD, then inhibitory agents may be effective against multiple stages of the disorder, as observed with the senolytic compound Nutlin-3a in animal models [82]. It would be particularly valuable to treat the mild and moderate stages of AMD, the incidences of which are increasing [2, 3]. In addition, agents that inhibit or delay cell senescence may be prophylactic against AMD [33], as appears to be the case for metformin [44- 47]. Finally, as a corollary benefit, the successful treatment of AMD through a senescence-based approach may encourage the pursuit of anti-senescence treatments for other aging disorders.

CONCLUSIONS

The idea that the pathology of AMD involves RPE cell senescence was first proposed more than two decades ago. Over a decade ago, it was hypothesized that RPE cell senescence was, in fact, a key contributor to this pathology. Over the past decade, many agents have been investigated for the treatment of AMD. Although a few were designed to address cell senescence, the majority targeted other potential pathological mechanisms. In support of our original hypothesis, we now present evidence that many of the newer agents investigated for the treatment of AMD, even those that were not meant to reduce cell senescence or its effects, have this function as part of their activity profiles. Further experimental studies or clinical trials exploring the safety and efficacy of inhibiting RPE cell senescence or reversing its effects are needed to pave the way for improved AMD treatment.

ETHICAL DECLARATIONS

Ethical approval: Not required.
Conflict of interests: None

FUNDING

ACKNOWLEDGMENTS

REFERENCES

1. Wang Y, Zhong Y, Zhang L, Wu Q, Tham Y, Rim TH, et al. Global Incidence, Progression, and Risk Factors of Age-Related Macular Degeneration and Projection of Disease Statistics in 30 Years: A Modeling Study. Gerontology. 2022;68(7):721-735. doi: 10.1159/000518822 pmid: 34569526

2. GBD 2019 Blindness and Vision Impairment Collaborators; Vision Loss Expert Group of the Global Burden of Disease Study. Causes of blindness and vision impairment in 2020 and trends over 30 years, and prevalence of avoidable blindness in relation to VISION 2020: the Right to Sight: an analysis for the Global Burden of Disease Study. Lancet Glob Health. 2021;9(2):e144-e160. doi: 10.1016/S2214-109X(20)30489-7. Erratum in: Lancet Glob Health. 2021;9(4):e408. pmid: 33275949

3. Furtado JM, Jonas J, Peto T, Steinmetz JD, Briant PS, Wong TY, et al. Global vision loss due to age-related macular degeneration. Investigative Ophthalmology & Visual Science. 2021;62(8):3504. Link

4. Cabral de Guimaraes TA, Daich Varela M, Georgiou M, Michaelides M. Treatments for dry age-related macular degeneration: therapeutic avenues, clinical trials and future directions. Br J Ophthalmol. 2022;106(3):297-304. doi: 10.1136/bjophthalmol-2020-318452 pmid: 33741584

5. Taskintuna I, Elsayed ME, Schatz P. Update on Clinical Trials in Dry Age-related Macular Degeneration. Middle East Afr J Ophthalmol. 2016;23(1):13-26. doi: 10.4103/0974-9233.173134 pmid: 26957835

6. Al-Zamil WM, Yassin SA. Recent developments in age-related macular degeneration: a review. Clin Interv Aging. 2017;12:1313-1330. doi: 10.2147/CIA.S143508 pmid: 28860733

7. Hadziahmetovic M, Malek G. Age-Related Macular Degeneration Revisited: From Pathology and Cellular Stress to Potential Therapies. Front Cell Dev Biol. 2021;8:612812. doi: 10.3389/fcell.2020.612812 pmid: 33569380

8. Flores R, Carneiro Â, Vieira M, Tenreiro S, Seabra MC. Age-Related Macular Degeneration: Pathophysiology, Management, and Future Perspectives. Ophthalmologica. 2021;244(6):495-511. doi: 10.1159/000517520 pmid: 34130290

9. Samanta A, Aziz AA, Jhingan M, Singh SR, Khanani AM, Chhablani J. Emerging Therapies in Neovascular Age-Related Macular Degeneration in 2020. Asia Pac J Ophthalmol (Phila). 2020;9(3):250-259. doi: 10.1097/APO.0000000000000291 pmid: 32511123

10. Ciulla TA, Rosenfeld PJ. Antivascular endothelial growth factor therapy for neovascular age-related macular degeneration. Curr Opin Ophthalmol. 2009;20(3):158-65. doi: 10.1097/ICU.0b013e32832d25b3 pmid: 19417570

11. Wallsh JO, Gallemore RP. Anti-VEGF-Resistant Retinal Diseases: A Review of the Latest Treatment Options. Cells. 2021;10(5):1049. doi: 10.3390/cells10051049 pmid: 33946803

12. Kaiser SM, Arepalli S, Ehlers JP. Current and Future Anti-VEGF Agents for Neovascular Age-Related Macular Degeneration. J Exp Pharmacol. 2021;13:905-912. doi: 10.2147/JEP.S259298 pmid: 34616189

13. Pavluk L, Kaya M (2023). 'FDA Approves SYFOVRE™ (pegcetacoplan injection) as the First and Only Treatment for Geographic Atrophy (GA), a Leading Cause of Blindness'. Available at: https://investors.apellis.com/news-releases/news-release-details/fda-approves-syfovretm-pegcetacoplan-injection-first-and-only (Accessed: April 09, 2023)

14. Age-Related Eye Disease Study Research Group. A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E, beta carotene, and zinc for age-related macular degeneration and vision loss: AREDS report no. 8. Arch Ophthalmol. 2001;119(10):1417-36. doi: 10.1001/archopht.119.10.1417. Erratum in: Arch Ophthalmol. 2008;126(9):1251. pmid: 11594942

15. Age-Related Eye Disease Study 2 Research Group. Lutein + zeaxanthin and omega-3 fatty acids for age-related macular degeneration: the Age-Related Eye Disease Study 2 (AREDS2) randomized clinical trial. JAMA. 2013;309(19):2005-15. doi: 10.1001/jama.2013.4997. Erratum in: JAMA. 2013;310(2):208. pmid: 23644932

16. Wiley CD, Campisi J. The metabolic roots of senescence: mechanisms and opportunities for intervention. Nat Metab. 2021;3(10):1290-1301. doi: 10.1038/s42255-021-00483-8 pmid: 34663974

17. Shay JW, Wright WE. Telomeres and telomerase: three decades of progress. Nat Rev Genet. 2019;20(5):299-309. doi: 10.1038/s41576-019-0099-1 pmid: 30760854

18. Di Micco R, Krizhanovsky V, Baker D, d'Adda di Fagagna F. Cellular senescence in ageing: from mechanisms to therapeutic opportunities. Nat Rev Mol Cell Biol. 2021;22(2):75-95. doi: 10.1038/s41580-020-00314-w pmid: 33328614

19. Ruan Y, Jiang S, Musayeva A, Gericke A. Oxidative Stress and Vascular Dysfunction in the Retina: Therapeutic Strategies. Antioxidants (Basel). 2020;9(8):761. doi: 10.3390/antiox9080761 pmid: 32824523

20. B Domènech E, Marfany G. The Relevance of Oxidative Stress in the Pathogenesis and Therapy of Retinal Dystrophies. Antioxidants (Basel). 2020;9(4):347. doi: 10.3390/antiox9040347 pmid: 32340220

21. Ozawa Y. Oxidative stress in the light-exposed retina and its implication in age-related macular degeneration. Redox Biol. 2020;37:101779. doi: 10.1016/j.redox.2020.101779 pmid: 33172789

22. Lee BY, Han JA, Im JS, Morrone A, Johung K, Goodwin EC, et al. Senescence-associated beta-galactosidase is lysosomal beta-galactosidase. Aging Cell. 2006;5(2):187-95. doi: 10.1111/j.1474-9726.2006.00199.x pmid: 16626397

23. Lee KS, Lin S, Copland DA, Dick AD, Liu J. Cellular senescence in the aging retina and developments of senotherapies for age-related macular degeneration. J Neuroinflammation. 2021;18(1):32. doi: 10.1186/s12974-021-02088-0 pmid: 33482879

24. Burton DG. Cellular senescence, ageing and disease. Age (Dordr). 2009;31(1):1-9. doi: 10.1007/s11357-008-9075-y pmid: 19234764

25. Bhutto I, Lutty G. Understanding age-related macular degeneration (AMD): relationships between the photoreceptor/retinal pigment epithelium/Bruch's membrane/choriocapillaris complex. Mol Aspects Med. 2012;33(4):295-317. doi: 10.1016/j.mam.2012.04.005 pmid: 22542780

26. Spaide R (2006). 'Etiology of late-age-related macular disease'. In Alfaro III DV, Liggett PE, Mieler WF, Quirox-Mercado H, Jager RD, Tano Y (Ed.). Age-related macular degeneration: A comprehensive textbook (pp. 23-39). Philadelphia: Lippincot, Williams and Wilkins. ISBN 13: 978-0-7817-3899-6. Link

27. Somasundaran S, Constable IJ, Mellough CB, Carvalho LS. Retinal pigment epithelium and age-related macular degeneration: A review of major disease mechanisms. Clin Exp Ophthalmol. 2020;48(8):1043-1056. doi: 10.1111/ceo.13834 pmid: 32710488

28. Thompson W (2006). 'Classification of age-related macular disease'. In Alfaro III DV, Liggett PE, Mieler WF, Quirox-Mercado H, Jager RD, Tano Y (Ed.). Age-related macular degeneration: A comprehensive textbook (pp. 44-52). Philadelphia: Lippincot, Williams and Wilkins. ISBN 13: 978-0-7817-3899-6. Link

29. Edwards AO, Ritter R 3rd, Abel KJ, Manning A, Panhuysen C, Farrer LA. Complement factor H polymorphism and age-related macular degeneration. Science. 2005;308(5720):421-4. doi: 10.1126/science.1110189 pmid: 15761121

30. O'Leary F, Campbell M. The blood-retina barrier in health and disease. FEBS J. 2023;290(4):878-891. doi: 10.1111/febs.16330 pmid: 34923749

31. Ford KM, Saint-Geniez M, Walshe T, Zahr A, D'Amore PA. Expression and role of VEGF in the adult retinal pigment epithelium. Invest Ophthalmol Vis Sci. 2011;52(13):9478-87. doi: 10.1167/iovs.11-8353 pmid: 22058334

32. Datta S, Cano M, Ebrahimi K, Wang L, Handa JT. The impact of oxidative stress and inflammation on RPE degeneration in non-neovascular AMD. Prog Retin Eye Res. 2017;60:201-218. doi: 10.1016/j.preteyeres.2017.03.002 pmid: 28336424

33. Kozlowski MR. RPE cell senescence: a key contributor to age-related macular degeneration. Med Hypotheses. 2012;78(4):505-10. doi: 10.1016/j.mehy.2012.01.018 pmid: 22296808

34. Minamino T, Komuro I. Vascular cell senescence: contribution to atherosclerosis. Circ Res. 2007;100(1):15-26. doi: 10.1161/01.RES.0000256837.40544.4a pmid: 17204661

35. Muñoz-Espín D, Serrano M. Cellular senescence: from physiology to pathology. Nat Rev Mol Cell Biol. 2014;15(7):482-96. doi: 10.1038/nrm3823 pmid: 24954210

36. Rodier F, Campisi J. Four faces of cellular senescence. J Cell Biol. 2011;192(4):547-56. doi: 10.1083/jcb.201009094 pmid: 21321098

37. Justice JN, Nambiar AM, Tchkonia T, LeBrasseur NK, Pascual R, Hashmi SK, et al. Senolytics in idiopathic pulmonary fibrosis: Results from a first-in-human, open-label, pilot study. EBioMedicine. 2019;40:554-563. doi: 10.1016/j.ebiom.2018.12.052 pmid: 30616998

38. Salaami O, Kuo CL, Drake MT, Kuchel GA, Kirkland JL, Pignolo RJ. Antidiabetic Effects of the Senolytic Agent Dasatinib. Mayo Clin Proc. 2021;96(12):3021-3029. doi: 10.1016/j.mayocp.2021.06.025 pmid: 34772496

39. Kritsilis M, V Rizou S, Koutsoudaki PN, Evangelou K, Gorgoulis VG, Papadopoulos D. Ageing, Cellular Senescence and Neurodegenerative Disease. Int J Mol Sci. 2018;19(10):2937. doi: 10.3390/ijms19102937 pmid: 30261683

40. Blasiak J, Piechota M, Pawlowska E, Szatkowska M, Sikora E, Kaarniranta K. Cellular Senescence in Age-Related Macular Degeneration: Can Autophagy and DNA Damage Response Play a Role? Oxid Med Cell Longev. 2017;2017:5293258. doi: 10.1155/2017/5293258 pmid: 29225722

41. Sreekumar PG, Hinton DR, Kannan R. The Emerging Role of Senescence in Ocular Disease. Oxid Med Cell Longev. 2020;2020:2583601. doi: 10.1155/2020/2583601 pmid: 32215170

42. Liang FQ, Godley BF. Oxidative stress-induced mitochondrial DNA damage in human retinal pigment epithelial cells: a possible mechanism for RPE aging and age-related macular degeneration. Exp Eye Res. 2003;76(4):397-403. doi: 10.1016/s0014-4835(03)00023-x pmid: 12634104

43. Majji AB, Cao J, Chang KY, Hayashi A, Aggarwal S, Grebe RR, et al. Age-related retinal pigment epithelium and Bruch's membrane degeneration in senescence-accelerated mouse. Invest Ophthalmol Vis Sci. 2000;41(12):3936-42. pmid: 11053297

44. Blitzer AL, Ham SA, Colby KA, Skondra D. Association of Metformin Use With Age-Related Macular Degeneration: A Case-Control Study. JAMA Ophthalmol. 2021;139(3):302-309. doi: 10.1001/jamaophthalmol.2020.6331 pmid: 33475696

45. Chen YY, Shen YC, Lai YJ, Wang CY, Lin KH, Feng SC, et al. Association between Metformin and a Lower Risk of Age-Related Macular Degeneration in Patients with Type 2 Diabetes. J Ophthalmol. 2019;2019:1649156. doi: 10.1155/2019/1649156 pmid: 31781371

46. Brown EE, Ball JD, Chen Z, Khurshid GS, Prosperi M, Ash JD. The Common Antidiabetic Drug Metformin Reduces Odds of Developing Age-Related Macular Degeneration. Invest Ophthalmol Vis Sci. 2019;60(5):1470-1477. doi: 10.1167/iovs.18-26422 pmid: 30973575

47. Stewart JM, Lamy R, Wu F, Keenan JD. Relationship between Oral Metformin Use and Age-Related Macular Degeneration. Ophthalmol Retina. 2020;4(11):1118-1119. doi: 10.1016/j.oret.2020.06.003 pmid: 32525055

48. Lee H, Jeon HL, Park SJ, Shin JY. Effect of Statins, Metformin, Angiotensin-Converting Enzyme Inhibitors, and Angiotensin II Receptor Blockers on Age-Related Macular Degeneration. Yonsei Med J. 2019;60(7):679-686. doi: 10.3349/ymj.2019.60.7.679 pmid: 31250582

49. Eton EA, Wubben TJ, Besirli CG, Hua P, McGeehan B, VanderBeek BL. Association of metformin and development of dry age-related macular degeneration in a U.S. insurance claims database. Eur J Ophthalmol. 2022;32(1):417-423. doi: 10.1177/1120672121997288 pmid: 33607930

50. Stewart JM (2019). Metformin for the Minimization of Geographic Atrophy Progression in Patients With AMD (METforMIN). ClinicalTrials.gov Identifier: NCT02684578. Available at: https://clinicaltrials.gov/ct2/show/NCT02684578 (Accessed: November 20, 2022)

51. Ahmed SM, Elshenawy SE, Sedky S, Elmehrath AO, El-Badri N. Pancreatic β-Cell senescence: mechanisms and association with diabetes. EMJ. 2021;6(1):59-72. doi: 10.33590/emj/20-00128

52. Aguayo-Mazzucato C, Andle J, Lee TB Jr, Midha A, Talemal L, Chipashvili V, et al. Acceleration of β Cell Aging Determines Diabetes and Senolysis Improves Disease Outcomes. Cell Metab. 2019;30(1):129-142.e4. doi: 10.1016/j.cmet.2019.05.006 pmid: 31155496

53. Berlanga-Acosta JA, Guillén-Nieto GE, Rodríguez-Rodríguez N, Mendoza-Mari Y, Bringas-Vega ML, Berlanga-Saez JO, et al. Cellular Senescence as the Pathogenic Hub of Diabetes-Related Wound Chronicity. Front Endocrinol (Lausanne). 2020;11:573032. doi: 10.3389/fendo.2020.573032 pmid: 33042026

54. Helman A, Avrahami D, Klochendler A, Glaser B, Kaestner KH, Ben-Porath I, et al. Effects of ageing and senescence on pancreatic β-cell function. Diabetes Obes Metab. 2016;18 Suppl 1:58-62. doi: 10.1111/dom.12719 pmid: 27615132

55. Lee JH, Lee J. Endoplasmic Reticulum (ER) Stress and Its Role in Pancreatic β-Cell Dysfunction and Senescence in Type 2 Diabetes. Int J Mol Sci. 2022;23(9):4843. doi: 10.3390/ijms23094843 pmid: 35563231

56. Murakami T, Inagaki N, Kondoh H. Cellular Senescence in Diabetes Mellitus: Distinct Senotherapeutic Strategies for Adipose Tissue and Pancreatic β Cells. Front Endocrinol (Lausanne). 2022;13:869414. doi: 10.3389/fendo.2022.869414 pmid: 35432205

57. Palmer AK, Tchkonia T, LeBrasseur NK, Chini EN, Xu M, Kirkland JL. Cellular Senescence in Type 2 Diabetes: A Therapeutic Opportunity. Diabetes. 2015;64(7):2289-98. doi: 10.2337/db14-1820 pmid: 26106186

58. Palmer AK, Tchkonia T, Kirkland JL. Senolytics: Potential for Alleviating Diabetes and Its Complications. Endocrinology. 2021;162(8):bqab058. doi: 10.1210/endocr/bqab058 pmid: 33705532

59. Anisimov VN, Semenchenko AV, Yashin AI. Insulin and longevity: antidiabetic biguanides as geroprotectors. Biogerontology. 2003;4(5):297-307. doi: 10.1023/a:1026299318315 pmid: 14618027

60. Cao F, Wu K, Zhu YZ, Bao ZW. Roles and Mechanisms of Dipeptidyl Peptidase 4 Inhibitors in Vascular Aging. Front Endocrinol (Lausanne). 2021;12:731273. doi: 10.3389/fendo.2021.731273 pmid: 34489872

61. Madonna R, Doria V, Minnucci I, Pucci A, Pierdomenico DS, De Caterina R. Empagliflozin reduces the senescence of cardiac stromal cells and improves cardiac function in a murine model of diabetes. J Cell Mol Med. 2020;24(21):12331-12340. doi: 10.1111/jcmm.15699 pmid: 32940423

62. Fang J, Yang J, Wu X, Zhang G, Li T, Wang X, et al. Metformin alleviates human cellular aging by upregulating the endoplasmic reticulum glutathione peroxidase 7. Aging Cell. 2018;17(4):e12765. doi: 10.1111/acel.12765 pmid: 29659168

63. Moiseeva O, Deschênes-Simard X, St-Germain E, Igelmann S, Huot G, Cadar AE, et al. Metformin inhibits the senescence-associated secretory phenotype by interfering with IKK/NF-κB activation. Aging Cell. 2013;12(3):489-98. doi: 10.1111/acel.12075 pmid: 23521863

64. Zhang C, Chen M, Zhou N, Qi Y. Metformin Prevents H_2O_2-Induced Senescence in Human Lens Epithelial B3 Cells. Med Sci Monit Basic Res. 2020;26:e923391. doi: 10.12659/MSMBR.923391 pmid: 32336745

65. Chen D, Xia D, Pan Z, Xu D, Zhou Y, Wu Y, et al. Metformin protects against apoptosis and senescence in nucleus pulposus cells and ameliorates disc degeneration in vivo. Cell Death Dis. 2016;7(10):e2441. doi: 10.1038/cddis.2016.334 pmid: 27787519

66. Algire C, Moiseeva O, Deschênes-Simard X, Amrein L, Petruccelli L, Birman E, et al. Metformin reduces endogenous reactive oxygen species and associated DNA damage. Cancer Prev Res (Phila). 2012;5(4):536-43. doi: 10.1158/1940-6207.CAPR-11-0536 pmid: 22262811

67. Karnewar S, Neeli PK, Panuganti D, Kotagiri S, Mallappa S, Jain N, et al. Metformin regulates mitochondrial biogenesis and senescence through AMPK mediated H3K79 methylation: Relevance in age-associated vascular dysfunction. Biochim Biophys Acta Mol Basis Dis. 2018;1864(4 Pt A):1115-1128. doi: 10.1016/j.bbadis.2018.01.018 pmid: 29366775

68. Mao Z, Liu W, Huang Y, Sun T, Bao K, Feng J, et al. Anti-aging effects of chlorpropamide depend on mitochondrial complex-II and the production of mitochondrial reactive oxygen species. Acta Pharm Sin B. 2022;12(2):665-677. doi: 10.1016/j.apsb.2021.08.007 pmid: 35256938

69. Chen L, Bi B, Zeng J, Zhou Y, Yang P, Guo Y, et al. Rosiglitazone ameliorates senescence-like phenotypes in a cellular photoaging model. J Dermatol Sci. 2015;77(3):173-81. doi: 10.1016/j.jdermsci.2015.01.007 pmid: 25703056

70. Kozlowski MR, Grossman B, Kneeland A, Zemski S, Crandell A, Kozlowski RE. Inhibition of Retinal Pigment Epithelial Cell Senescence by Metformin: Implications for the Treatment of Macular Degeneration. Japanese Journal of Ophthalmology and Research. 2020;2(1):1-5. doi:10.31546/2732-4516.1005

71. Le Pelletier L, Mantecon M, Gorwood J, Auclair M, Foresti R, Motterlini R, et al. Metformin alleviates stress-induced cellular senescence of aging human adipose stromal cells and the ensuing adipocyte dysfunction. Elife. 2021;10:e62635. doi: 10.7554/eLife.62635 pmid: 34544550

72. Rena G, Hardie DG, Pearson ER. The mechanisms of action of metformin. Diabetologia. 2017;60(9):1577-1585. doi: 10.1007/s00125-017-4342-z pmid: 28776086

73. Kuppermann BD, Patel SS, Boyer DS, Augustin AJ, Freeman WR, Kerr KJ, et al; Brimo DDS Gen 1 Study Group. Phase 2 study of the safety and efficacy of brimonidine drug delivery system (brimo dds) generation 1 in patients with geographic atrophy secondary to age-related macular degeneration. Retina. 2021;41(1):144-155. doi: 10.1097/IAE.0000000000002789 pmid: 32134802

74. Saylor M, McLoon LK, Harrison AR, Lee MS. Experimental and clinical evidence for brimonidine as an optic nerve and retinal neuroprotective agent: an evidence-based review. Arch Ophthalmol. 2009;127(4):402-6. doi: 10.1001/archophthalmol.2009.9 pmid: 19365015

75. Ito T, Sawada R, Fujiwara Y, Seyama Y, Tsuchiya T. FGF-2 suppresses cellular senescence of human mesenchymal stem cells by down-regulation of TGF-beta2. Biochem Biophys Res Commun. 2007;359(1):108-14. doi: 10.1016/j.bbrc.2007.05.067 pmid: 17532297

76. Farooq M, Khan AW, Kim MS, Choi S. The Role of Fibroblast Growth Factor (FGF) Signaling in Tissue Repair and Regeneration. Cells. 2021;10(11):3242. doi: 10.3390/cells10113242 pmid: 34831463

77. Martin M, Blom AM. Complement in removal of the dead - balancing inflammation. Immunol Rev. 2016;274(1):218-232. doi: 10.1111/imr.12462 pmid: 27782329

78. Romashkan S, Chang H, Hadley EC. National Institute on Aging Workshop: repurposing drugs or dietary supplements for their senolytic or senomorphic effects: considerations for clinical trials. The Journals of Gerontology: Series A. 2021;76(6):1144-52. doi: 10.1093/gerona/glab028

79. Bhisitkul R, Klier S, Tsuruda P, Xie B, Masaki L, Bautista J, et al. UBX1325, A Novel Senolytic Treatment for Patients with Advanced DME or wet AMD: 24-Week Results of a Phase 1 Study. Investigative Ophthalmology & Visual Science. 2022;63(7):4287. Link

80. Chung H, Kim C. Nutlin-3a for age-related macular degeneration. Aging (Albany NY). 2022;14(14):5614-5616. doi: 10.18632/aging.204187 pmid: 35849498

81. Chae JB, Jang H, Son C, Park CW, Choi H, Jin S, et al. Targeting senescent retinal pigment epithelial cells facilitates retinal regeneration in mouse models of age-related macular degeneration. Geroscience. 2021;43(6):2809-2833. doi: 10.1007/s11357-021-00457-4. Erratum in: Geroscience. 2022;44(3):1885. pmid: 34601706

82. Aubrey BJ, Kelly GL, Janic A, Herold MJ, Strasser A. How does p53 induce apoptosis and how does this relate to p53-mediated tumour suppression? Cell Death Differ. 2018;25(1):104-113. doi: 10.1038/cdd.2017.169 pmid: 29149101

83. Czabotar PE, Lessene G, Strasser A, Adams JM. Control of apoptosis by the BCL-2 protein family: implications for physiology and therapy. Nat Rev Mol Cell Biol. 2014;15(1):49-63. doi: 10.1038/nrm3722 pmid: 24355989

84. Minturn RJ, Bracha P, Klein MJ, Chhablani J, Harless AM, Maturi RK. Intravitreal sirolimus for persistent, exudative age-related macular degeneration: a Pilot Study. Int J Retina Vitreous. 2021;7(1):11. doi: 10.1186/s40942-021-00281-0 pmid: 33593448

85. Laberge RM, Sun Y, Orjalo AV, Patil CK, Freund A, Zhou L, et al. MTOR regulates the pro-tumorigenic senescence-associated secretory phenotype by promoting IL1A translation. Nat Cell Biol. 2015;17(8):1049-61. doi: 10.1038/ncb3195. Erratum in: Nat Cell Biol. 2021;23(5):564-565. pmid: 26147250

86. Ekshyyan O, Khandelwal AR, Rong X, Moore-Medlin T, Ma X, Alexander JS, et al. Rapamycin targets Interleukin 6 (IL-6) expression and suppresses endothelial cell invasion stimulated by tumor cells. Am J Transl Res. 2016;8(11):4822-4830. pmid: 27904683

87. Weichhart T. mTOR as Regulator of Lifespan, Aging, and Cellular Senescence: A Mini-Review. Gerontology. 2018;64(2):127-134. doi: 10.1159/000484629 pmid: 29190625

88. Chen Y, Wang J, Cai J, Sternberg P. Altered mTOR signaling in senescent retinal pigment epithelium. Invest Ophthalmol Vis Sci. 2010;51(10):5314-9. doi: 10.1167/iovs.10-5280 pmid: 20445122

89. Marazita MC, Dugour A, Marquioni-Ramella MD, Figueroa JM, Suburo AM. Oxidative stress-induced premature senescence dysregulates VEGF and CFH expression in retinal pigment epithelial cells: Implications for Age-related Macular Degeneration. Redox Biol. 2016;7:78-87. doi: 10.1016/j.redox.2015.11.011 pmid: 26654980

90. Lazzarini R, Nicolai M, Pirani V, Mariotti C, Di Primio R. Effects of senescent secretory phenotype acquisition on human retinal pigment epithelial stem cells. Aging (Albany NY). 2018;10(11):3173-3184. doi: 10.18632/aging.101624 pmid: 30444724

91. Chalam KV, Grover S, Sambhav K, Balaiya S, Murthy RK. Aqueous interleukin-6 levels are superior to vascular endothelial growth factor in predicting therapeutic response to bevacizumab in age-related macular degeneration. J Ophthalmol. 2014;2014:502174. doi: 10.1155/2014/502174 pmid: 25110587

92. Kumari R, Jat P. Mechanisms of Cellular Senescence: Cell Cycle Arrest and Senescence Associated Secretory Phenotype. Front Cell Dev Biol. 2021;9:645593. doi: 10.3389/fcell.2021.645593 pmid: 33855023

93. Krogh Nielsen M, Subhi Y, Molbech CR, Falk MK, Nissen MH, Sørensen TL. Systemic Levels of Interleukin-6 Correlate With Progression Rate of Geographic Atrophy Secondary to Age-Related Macular Degeneration. Invest Ophthalmol Vis Sci. 2019;60(1):202-208. doi: 10.1167/iovs.18-25878 pmid: 30644965

94. Yildirim Z, Ucgun NI, Yildirim F, Sepici-Dincel A. Choroidal neovascular membrane in age-related macular degeneration is associated with increased interleukin-6. International Journal of Gerontology. 2012;6(2):101-4. doi: 10.1016/j.ijge.2012.01.018

95. Nahavandipour A, Krogh Nielsen M, Sørensen TL, Subhi Y. Systemic levels of interleukin-6 in patients with age-related macular degeneration: a systematic review and meta-analysis. Acta Ophthalmol. 2020;98(5):434-444. doi: 10.1111/aos.14402 pmid: 32180348

96. Seddon JM, George S, Rosner B, Rifai N. Progression of age-related macular degeneration: prospective assessment of C-reactive protein, interleukin 6, and other cardiovascular biomarkers. Arch Ophthalmol. 2005;123(6):774-82. doi: 10.1001/archopht.123.6.774 pmid: 15955978

97. Sato K, Takeda A, Hasegawa E, Jo YJ, Arima M, Oshima Y, et al. Interleukin-6 plays a crucial role in the development of subretinal fibrosis in a mouse model. Immunol Med. 2018;41(1):23-29. doi: 10.1080/09114300.2018.1451609 pmid: 30938258

98. Barczyk M, Carracedo S, Gullberg D. Integrins. Cell Tissue Res. 2010;339(1):269-80. doi: 10.1007/s00441-009-0834-6 pmid: 19693543

99. Pan L, Zhao Y, Yuan Z, Qin G. Research advances on structure and biological functions of integrins. Springerplus. 2016;5(1):1094. doi: 10.1186/s40064-016-2502-0 pmid: 27468395

100. Shaw LT, Mackin A, Shah R, Jain S, Jain P, Nayak R, et al. Risuteganib-a novel integrin inhibitor for the treatment of non-exudative (dry) age-related macular degeneration and diabetic macular edema. Expert Opin Investig Drugs. 2020;29(6):547-554. doi: 10.1080/13543784.2020.1763953 pmid: 32349559

101. Age-Related Eye Disease Study Research Group. A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E and beta carotene for age-related cataract and vision loss: AREDS report no. 9. Arch Ophthalmol. 2001;119(10):1439-52. doi: 10.1001/archopht.119.10.1439. Erratum in: Arch Ophthalmol. 2008;126(9):1251. pmid: 11594943

102. Widmaier M, Rognoni E, Radovanac K, Azimifar SB, Fässler R. Integrin-linked kinase at a glance. J Cell Sci. 2012;125(Pt 8):1839-43. doi: 10.1242/jcs.093864 pmid: 22637643

103. Li Z, Chen X, Xie Y, Shi S, Feng Z, Fu B, et al. Expression and significance of integrin-linked kinase in cultured cells, normal tissue, and diseased tissue of aging rat kidneys. J Gerontol A Biol Sci Med Sci. 2004;59(10):984-96. doi: 10.1093/gerona/59.10.b984 pmid: 15528771

104. Chen X, Li Z, Feng Z, Wang J, Ouyang C, Liu W, et al. Integrin-linked kinase induces both senescence-associated alterations and extracellular fibronectin assembly in aging cardiac fibroblasts. J Gerontol A Biol Sci Med Sci. 2006;61(12):1232-45. doi: 10.1093/gerona/61.12.1232 pmid: 17234816

105. Troyano-Suárez N, del Nogal-Avila M, Mora I, Sosa P, López-Ongil S, Rodriguez-Puyol D, et al. Glucose Oxidase Induces Cellular Senescence in Immortal Renal Cells through ILK by Downregulating Klotho Gene Expression. Oxid Med Cell Longev. 2015;2015:416738. doi: 10.1155/2015/416738. Erratum in: Oxid Med Cell Longev. 2016;2016:8392708. pmid: 26583057

106. Olmos G, López-Ongil S, Ruiz Torres MP. Integrin-linked kinase: A new actor in the ageing process? Exp Gerontol. 2017;100:87-90. doi: 10.1016/j.exger.2017.10.026 pmid: 29101014

107. Stewart MW, Garg S, Newman EM, Jeffords E, Konopińska J, Jackson S, et al. Safety and Therapeutic Effects of Orally Administered Akst4290 in Newly Diagnosed Neovascular Age-Related Macular Degeneration. Retina. 2022;42(6):1038-1046. doi: 10.1097/IAE.0000000000003446 pmid: 35537111

108. Takeda A, Baffi JZ, Kleinman ME, Cho WG, Nozaki M, Yamada K, et al. CCR3 is a target for age-related macular degeneration diagnosis and therapy. Nature. 2009;460(7252):225-30. doi: 10.1038/nature08151 pmid: 19525930

109. Hoefer J, Luger M, Dal-Pont C, Culig Z, Schennach H, Jochberger S. The "Aging Factor" Eotaxin-1 (CCL11) Is Detectable in Transfusion Blood Products and Increases with the Donor's Age. Front Aging Neurosci. 2017;9:402. doi: 10.3389/fnagi.2017.00402 pmid: 29249965

110. Zayed M, Iohara K, Watanabe H, Nakashima M. CCR3 antagonist protects against induced cellular senescence and promotes rejuvenation in periodontal ligament cells for stimulating pulp regeneration in the aged dog. Sci Rep. 2020;10(1):8631. doi: 10.1038/s41598-020-65301-9 pmid: 32451381

111. Bae K, Noh SR, Kang SW, Kim ES, Yu SY. Angiographic Subtypes of Neovascular Age-related Macular Degeneration in Korean: A New Diagnostic Challenge. Sci Rep. 2019;9(1):9701. doi: 10.1038/s41598-019-46235-3 pmid: 31273295

112. Sharma K, Sharma NK, Singh R, Anand A. Exploring the role of VEGF in Indian Age related macular degeneration. Ann Neurosci. 2015;22(4):232-7. doi: 10.5214/ans.0972.7531.220408 pmid: 26526736

113. Shahidatul-Adha M, Zunaina E, Aini-Amalina MN. Evaluation of vascular endothelial growth factor (VEGF) level in the tears and serum of age-related macular degeneration patients. Sci Rep. 2022;12(1):4423. doi: 10.1038/s41598-022-08492-7 pmid: 35292705

114. Edwards AO, Malek G. Molecular genetics of AMD and current animal models. Angiogenesis. 2007;10(2):119-32. doi: 10.1007/s10456-007-9064-2 pmid: 17372852

115. Pennesi ME, Neuringer M, Courtney RJ. Animal models of age related macular degeneration. Mol Aspects Med. 2012;33(4):487-509. doi: 10.1016/j.mam.2012.06.003 pmid: 22705444

116. Zeiss CJ. Animals as models of age-related macular degeneration: an imperfect measure of the truth. Vet Pathol. 2010;47(3):396-413. doi: 10.1177/0300985809359598 pmid: 20382825

117. Ablonczy Z, Crosson CE. VEGF modulation of retinal pigment epithelium resistance. Exp Eye Res. 2007;85(6):762-71. doi: 10.1016/j.exer.2007.08.010 pmid: 17915218

118. Wang X, Ohji M. Vascular endothelial growth factor and its inhibitor in age-related macular degeneration. Taiwan Journal of Ophthalmology. 2013;3(4):128-33. doi: 10.1016/j.tjo.2013.09.002

119. Cao S, Walker GB, Wang X, Cui JZ, Matsubara JA. Altered cytokine profiles of human retinal pigment epithelium: oxidant injury and replicative senescence. Mol Vis. 2013;19:718-28 pmid: 23559866

120. Coppé JP, Kauser K, Campisi J, Beauséjour CM. Secretion of vascular endothelial growth factor by primary human fibroblasts at senescence. J Biol Chem. 2006;281(40):29568-74. doi: 10.1074/jbc.M603307200 pmid: 16880208

121. Mikuła-Pietrasik J, Sosińska P, Naumowicz E, Maksin K, Piotrowska H, Woźniak A, et al. Senescent peritoneal mesothelium induces a pro-angiogenic phenotype in ovarian cancer cells in vitro and in a mouse xenograft model in vivo. Clin Exp Metastasis. 2016;33(1):15-27. doi: 10.1007/s10585-015-9753-y pmid: 26433963

122. Kozlowski MR. Senescent retinal pigment epithelial cells are more sensitive to vascular endothelial growth factor: implications for "wet" age-related macular degeneration. J Ocul Pharmacol Ther. 2015;31(2):87-92. doi: 10.1089/jop.2014.0071 pmid: 25453983

123. Ren JL, Pan JS, Lu YP, Sun P, Han J. Inflammatory signaling and cellular senescence. Cell Signal. 2009;21(3):378-83. doi: 10.1016/j.cellsig.2008.10.011 pmid: 18992324

124. Nemoto K, Kondo Y, Himeno S, Suzuki Y, Hara S, Akimoto M, et al. Modulation of telomerase activity by zinc in human prostatic and renal cancer cells. Biochem Pharmacol. 2000;59(4):401-5. doi: 10.1016/s0006-2952(99)00334-2 pmid: 10644048

125. Farzaneh-Far R, Lin J, Epel ES, Harris WS, Blackburn EH, Whooley MA. Association of marine omega-3 fatty acid levels with telomeric aging in patients with coronary heart disease. JAMA. 2010;303(3):250-7. doi: 10.1001/jama.2009.2008 pmid: 20085953

126. Johra FT, Bepari AK, Bristy AT, Reza HM. A Mechanistic Review of β-Carotene, Lutein, and Zeaxanthin in Eye Health and Disease. Antioxidants (Basel). 2020;9(11):1046. doi: 10.3390/antiox9111046 pmid: 33114699