

Therapeutic approaches with intravitreal injections in geographic atrophy secondary to age-related macular degeneration: current drugs and potential molecules

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ABSTRACT

The present review focuses on recent clinical trials that analyze the efficacy of intravitreal therapeutic agents for the treatment of dry age-related macular degeneration (AMD), such as neuroprotective drugs, and complement inhibitors, also called immunomodulatory or anti-inflammatory. A systematic literature search was performed to identify randomized controlled trials published prior to January 2019. Patients affected by dry AMD treated with intravitreal therapeutic agents were included. The changes in the correct visual acuity and the reduction in geographic atrophy progression were evaluated. Several new drugs have shown some promising results, including those targeting the complement cascade and agents called neuroprotective. The action potential of the two groups of drugs is to block the complement cascade model for immunomodulating agents, and prevent the degeneration and apoptosis of ganglion cells for the neuroprotectors, respectively. To the best of knowledge, and after extensive studies on the matter, there are still many investigations to be carried out on dry AMD in collaboration between researchers. They will have to identify truly effective molecules, understand the practical potential of pluripotent stem cells, and refine gene therapies. Only in-depth clinical trials will be able to allow the most appropriate and personalized treatments for each dry AMD patient.

Keywords: age-related macular degeneration, anti-inflammatory agents, dry AMD, geographic atrophy, intravitreal injection, complement inhibitors, neuroprotective agents, non-exudative AMD.

1. INTRODUCTION

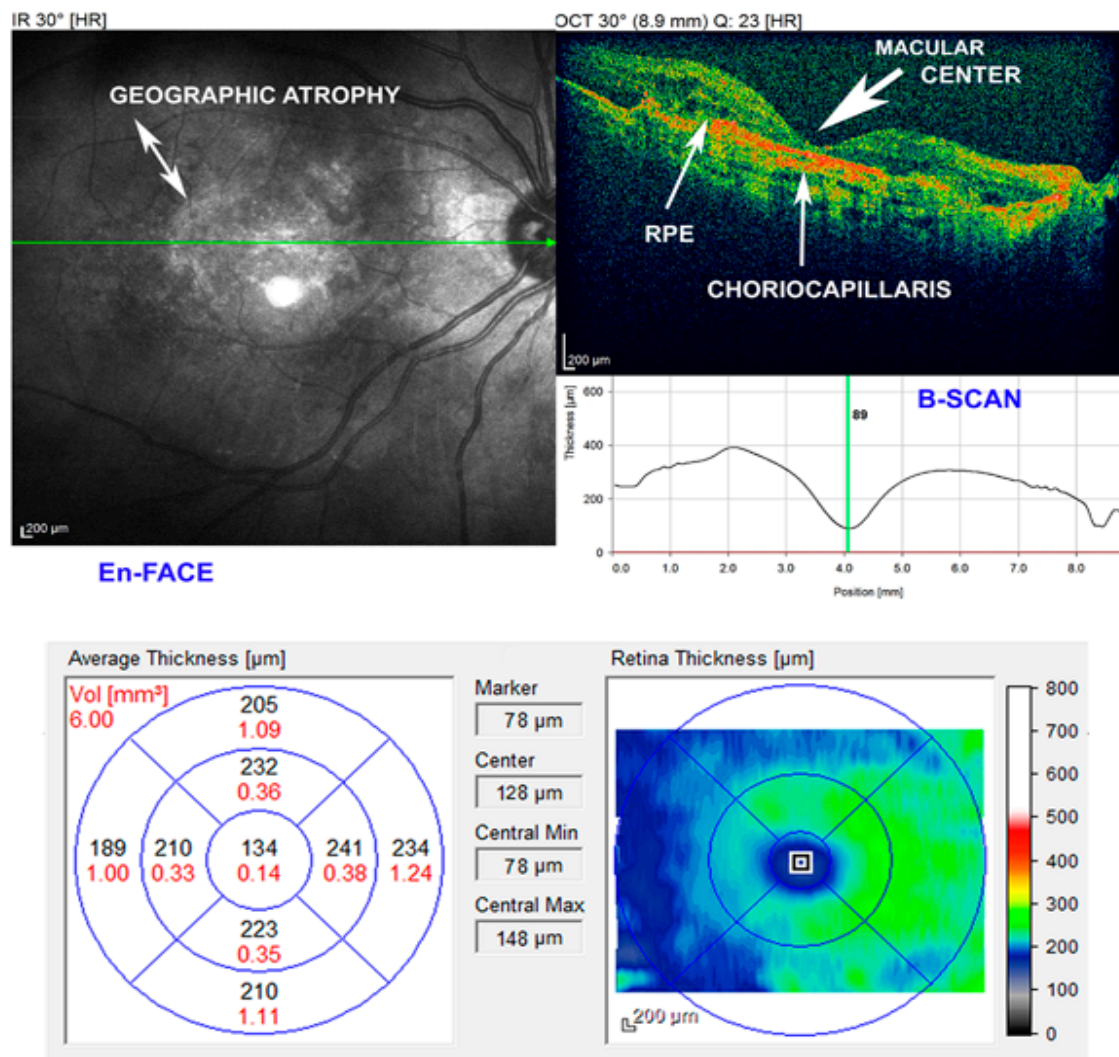
Age-related macular degeneration (AMD) is the leading cause of irreversible blindness in the elderly population, and it is defined as a chronic, multifactorial, and progressive central retinal

disease. The prevalence of AMD is approximately 8.69% in the worldwide population and 12.3% in Europe [1]. There were 8.4 million AMD patients with moderate to severe vision impairment in 2015 and, probably, there will be an increase to about 196 million in 2020 [1]. Macular pigmentation changes in the chorioretinal layers and the drusen are present in early stage of AMD, while, the advanced stages are characterized by dry and/or neovascular forms [1,2]. Choroidal neovascularization (CNV) in AMD, often accompanied by serum-hemorrhagic retinal detachment, eventually leads to the degeneration of photoreceptors [2,3]. Likewise, in dry AMD or non-exudative AMD, and then subsequently in geographic atrophy (GA), there are areas of progressive atrophy and thinning mainly involving the layers of the retinal pigment epithelium (RPE) and underlying choriocapillaris; these modifications are a prelude to the degeneration of photoreceptors leading to irreversible loss of visual function. [2-4] (Figure 1). The main risk factors for GA are ageing, family history, cigarette smoking, cardiovascular risk factors, previous cataract surgery, etc. [3]. Thus, the characteristics of GA with progressive and irreversible loss of retinal cells inevitably is responsible for around 20% of cases of legal blindness [1-4]. No effective treatment for GA is currently available, unlike neovascular AMD in which anti-angiogenic treatment are effective in improving visual acuity [4]. Inflammation, complement activation, oxidative stress, blood flow regulation, and reduced neuroprotection are the main pathways implicated in the progression of GA [3,4]. Nowadays, new therapies are under investigation, but unfortunately, no valid drugs are currently available for dry AMD. The only preventive option is the Age-Related Eye Disease Studies (AREDS) formulation, which reduces the risk of AMD progression [5]. The etiology of AMD is believed to be the resultant of a combination of oxidative stress, chronic inflammation, predisposing genetic and environmental factors. Excessive exposure to light, intensive oxygen metabolism, polyunsaturated fatty acids, and the presence of photosensitizers increase the production of retinal reactive oxygen species (ROS) resulting in

oxidative stress that can cause the induction of programmed necrosis in RPE cells and chronic inflammation [6]. Polymorphism in a number of genes, including members of the complement pathway, have been associated with AMD indicating the involvement of inflammation, lipid metabolism, angiogenesis, and RPE dysfunction in the disease [7].

The present review focuses on recent, or still ongoing, clinical trials evaluating the efficacy of intravitreal therapies in dry AMD, such as neuroprotective drugs, complement inhibitors, immunomodulatory or anti-inflammatory agents.

Figure 1. Imaging of geographic atrophy (GA). En-face and B-scan spectral-domain optical coherence tomography (SD-OCT). Decreased macular thickness (center marker 89/78 μm), retinal pigment epithelial (RPE) irregularities, and of the underlying choriocapillaris. The atrophic area shows hyperreflective clumps at different levels, segmented plaques of the outer band and elevations with variable reflectivity. GA is a form of advanced dry age macular degeneration (AMD). An eye may have uni-or multi-focal atrophic lesions, which when summed determine the central total lesion area. Scale bars = 500 μm .



2. METHODS

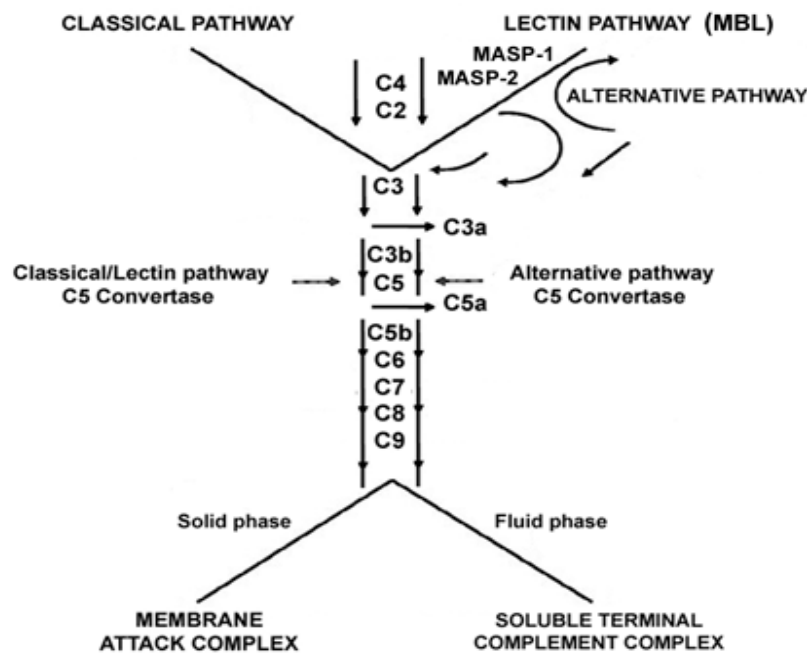
Our systematic literature search was conducted in PubMed, Embase, Cochrane Library, and Web to identify randomized controlled trials published prior to January 2019. The search keywords were: “age-related macular degeneration”, “atrophic”, “dry”, “current drugs”, “geographic atrophy”, “non-exudative”, “intravitreal injections”, and “potential molecules”.

The patients dry AMD treated with intravitreal therapeutic agents were included. The changes in the correct visual acuity (BCVA) and the reduction in GA progression were evaluated. We also studied reviews, comments, and disquisitions on the pathology, a current hot topic for international literature and the scientific world.

3. CURRENT DEVELOPMENTS IN INTRAVITREAL THERAPY

The therapies target different aspects of GA, including inflammatory pathways, oxidative stress and RPE degeneration, products of the visual cycle, restoration of choroidal perfusion, use of growth factors (GFs), modification of cellular DNA through genetic therapy, and replenishing RPE cells with stem cell-derived RPE cells [6-9]. For example, it has been hypothesized that anti-inflammatory agents could be represent a therapeutic option [8]. In fact, chronic inflammation is reported to play an important role in early AMD pathogenesis. The deposition of intracellular drusen in the RPE layer, containing cell debris and proteins such as complement components, triggers the inflammatory response [7,8]. The disease progresses, consequently, with sustained inflammatory response, further drusen accumulation and oxidative stress resulting in damage and eventually cell death. Immunohistochemical examination of drusen shows many proinflammatory proteins including apolipoprotein E, acute phase proteins, coagulation proteins, β -amyloid ($A\beta$), and complement activation components during the development of the disease [7-9]. The complement cascades in its various forms, classical, alternative and mannose-binding lectin, converge on a final single pathway via the cleavage of complement factor C3 into C3a and C3b that leads to phagocytosis, inflammation, formation of a membrane attack complex (MAC) and ultimately to cell death [9] (Scheme 1).

Scheme 1. Diagram outlining the complement pathways. Three pathways of complement activation: classical, lectin, and alternative. MBL: mannose-binding lectin; MASP: MBL-associated serine proteases, MASP-1, and MASP-2.

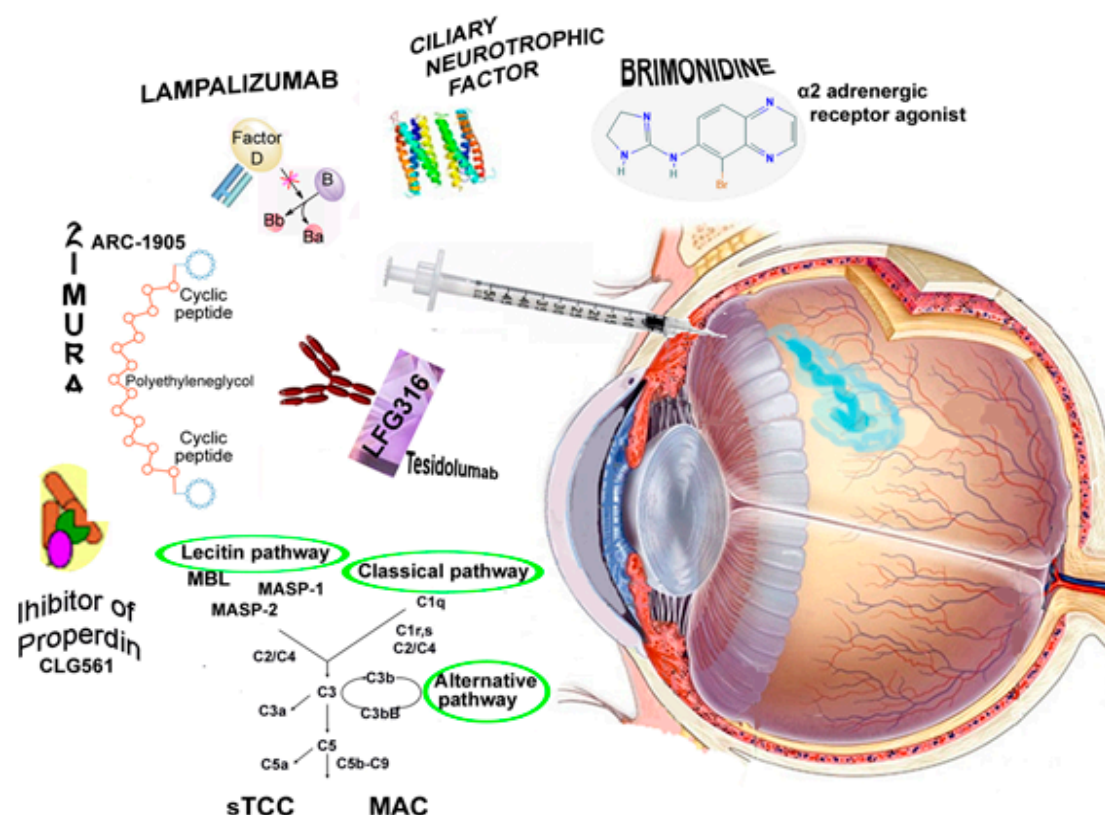


The alternative pathway has been implicated in the pathogenesis of the disease, and, in particular, complement components 2, 3, and 7 have been shown to be associated with AMD. The formation of MAC consequent to the activation of the complement pathway causes cell lysis and chemokine release, thus, causing the recruitment of inflammatory cells and the increase of vascular permeability [9]. Hence, the inhibition of the overactive complement pathway represents a viable therapeutic approach to arrest the progression of GA. Moreover, there is an interest in the role of immune dysfunction, such as inappropriate complement cascade activation, in the etiology of AMD. The knowledge of the major role that complement cascade activation plays in the disease has led to several therapeutic options [9-12].

In summary, we know that numerous proteins and polypeptides have therapeutic effects in the treatment of degenerative diseases, but localized treatment of retinal diseases is complicated by the blood-retinal barrier that hinders the penetration of several molecules from the circulatory blood system to the neurosensory retina [3,4]. Therefore, the intravitreal injection (IVT), without systemic exposure, could be the simplest and most useful route to allow an

adequate chorioretinal restoration in order to avoid, as far as possible, the final negative prognosis of the disease in GA [3,4]. With these premises, the present review will be focused on neuroprotective, and immunomodulatory, or anti-inflammatory agents that have been used for IVT in patients with GA (Figure 2). The summary of clinical trials for the IVTs of AMD patients with GA are in Table 1.

Figure 2. The intravitreal injection (IVT) is a procedure to place a medication directly into the vitreous cavity. IVTs are used to administer medications in various retinal conditions. Representation of molecules, neuroprotective, immunomodulatory or anti-inflammatory agents, that have been used for IVT in patients with geographic atrophy (GA). MBL: mannose-binding lectin; MASP: MBL-associated serine proteases, MASP-1, and MASP-2; sTCC: soluble terminal complement complex; MAC: membrane attack complex.



TARGET	DRUG	ACTION OF THE DRUG	STUDIES	AUTHORS
NEUROPROTECTORS	Brimonidine Tartrate α2 adrenergic receptor agonist	Prevents RGCs death via the non-amyloidogenic Aβ-pathway	NCT00658619 (2011) Ph1 NCT02087085 (2019) Ph2 Beacon <i>Allergan Inc.</i>	Nizari S, et al. 2016; Doozandeh A, et al. 2016
	Ciliary neurotrophic factor Encapsulated cell technology (ECT) and NT-501 implant	Member of the IL-6 family of neurotrophic cytokines, prevents photoreceptors degeneration	NCT00063765 (2006) Ph1 NCT00447954 (2009) Ph2 NCT00447993 (2009) Ph2 NCT00447980 (2010) Ph2 <i>Neurotech Pharmaceuticals</i>	Lambert PD, et al. 2001; Thanos CG, et al. 2004; Zhang K, et al. 2011; Kaufer K, et al. 2012
IMMUNE MODULATING – ANTIINFLAMMATORY or COMPLEMENT INHIBITORS	Lampalizumab Humanized monoclonal antibody	Inhibits complement factor D (CFD)–mediated activation and amplification of the alternative complement pathway	<i>NCT01229215 (2013) Ph2 Mahalo</i> NCT02247479 (2018) Ph3 Chroma NCT02247531 (2018) Ph3 Spectri <i>Genentech/Roche</i>	Do DV, et al. 2014; Yaspan BL, et al. 2017; Holz FG, et al. 2018
	Zimura ARC-1905. Single strand nucleic acid aptamer	Inhibits the cleavage of C5 and prevents the formation of the membrane attack complex (MAC)	NCT00950638 (2012) Ph1 NCT03362190 (2018) Ph2/3 NCT02686658 (2018) Ph2 <i>Ophthotech/Archemix</i>	Drolet DW, et al. 2016; Hariri, et al. 2015; Wei Y, et al. 2018; Sun H, et al. 2015
	APL-2 POT-4/AL-78898A Synthetic cyclic peptide conjugated to polyethylene glycol polymer. It is a modified version of POT-4 designed to have longer half-life	Binds to C3 blocking all three pathways of complement activation	NCT000473928 (2017) Ph1 NCT02503332 (2018) Ph 2 Filly NCT03525613 (2022) Ph3 Oaks NCT03525600 (2022) Ph3 Derby <i>Apellis Pharmaceuticals Inc.</i>	Kassa E, et al. 2019
	POT-4 AL-78898A. Cyclic peptide	Inhibits complement pathways and prevent MAC formation	NCT00473928 (2010) Ph1 <i>Alcon Inc.</i>	Kaushal S, et al. 2009; Singer M, et al. 2014
	CLG561 Inhibitor of properdin	Stabilizes the alternative pathway C3 and C5 convertases	NCT01835015 (2016) Ph1 NCT02515942 (2018) Ph2 <i>Novartis and Alcon Inc.</i>	Kassa E, et al. 2019. Ricklin D, et al. 2016
	LFG316 Tesidolumab. Human IgG1	Inhibits the complement system	NCT01255462 (2011) Ph1 NCT02515942 (2018) Ph2 NCT01527500 (2018) Ph2 <i>Novartis Pharmaceuticals</i>	Sagar A, et al. 2017 Kassa E, et al. 2019 Ricklin D, et al. 2016
SUPPRESSORS of INFLAMMATION	Iluvien Fluocinolone acetonide Corticosteroid	Vasoconstriction, release of inflammatory mediators, mitotic activity, suppression of membrane permeability, and immune response	NCT00695318 (2013) Ph2 <i>Alimera Sciences</i>	Taskintuna I, et al. 2016

Authors in References section. RGCs: retina ganglion cells; Ph: phase of the clinical trial; IL-6: interleukin-6; C: complement; IgG1: immunoglobulin G1.

4. POTENTIAL THERAPEUTIC MOLECULES IN DRY AMD

4.1. NEUROPROTECTIVE AGENTS DESIGNED TO PREVENT RETINAL GANGLION CELL APOPTOSIS

4.1.1. Brimonidine

Brimonidine is an α_2 adrenergic (α_2A) receptor agonist that has been used for its pressure lowering effects in the treatment of glaucoma for several years. Its mechanism in lowering the intraocular pressure involves both the decrease of aqueous humor production and increased uveoscleral outflow [13]. In short, brimonidine has been demonstrated to be neuroprotective via the modulation of $A\beta$ toxicity. This pathway is strongly implicated in neurodegenerative conditions such as Alzheimer's disease and in glaucoma-related models. $A\beta$ is the major constituent of senile plaque and may, therefore, play a key role in the stress-response to induce cellular apoptosis, even though to this day a comprehensive mechanism is not fully understood [13].

Nizari et al. proposed a model of α_2A agonists' neuroprotective effect against $A\beta$ toxicity [13]. $A\beta$ is associated with abnormal processing of amyloid precursor protein (APP). APP can be processed into $A\beta$ or soluble $APP\alpha$ (sAPP α) through the amyloidogenic and non-amyloidogenic pathways, respectively. Therefore, α_2A receptor agonists can negatively affect the amyloidogenic pathway, preventing cell death, partly by modulating excitotoxicity caused by glutamate. Moreover, α_2A receptor agonists can also affect APP processing via the extracellular matrix, by modulating matrix metalloproteinase-9 (MMP-9) and laminin through laminin-binding protein (LBP), preventing further toxic interactions with $A\beta$ and by increasing processing of APP into sAPP α , promoting the non-amyloidogenic pathway. Furthermore, α_2A receptor agonists can also increase levels of phosphorylated arabinose-inducible BAD promoter (P-Bad), and therefore promote cell survival and neuroprotection [13,14]. The initial results of a study (Clinical trials.gov: NCT00658619) involving 119

participants demonstrated that, in comparison to the sham arm, administration of low (200 µg) and high (400 µg) dose inserts of 22 gauge sustained-release brimonidine at baseline and at month 6 resulted in a statistically significant reduction in the size of GA for both low (by 18%) and high (by 27%) dose implants. No significant adverse effects have been reported for either inserts [14]. A larger phase 2 trial of 310 patients is currently underway (Beacon, NCT02087085). It is a double-blind, sham-control study with its primary outcome being the changes in size of GA lesion from baseline to 24 months [14]. Participants were randomized to receive either 400 µg sustained release solid brimonidine tartrate implant administered intravitreally on day one then every 3 months through month 21 or sham via needleless applicator. This clinical trial expected to end in 2019.

4.1.2. Ciliary neurotrophic factor

Ciliary neurotrophic factor (CNTF) is a neurotrophic factor member of the interleukin-6 (IL-6) family of neuropoietic cytokines. It influences the survival and differentiation of cells in the nervous system, including retinal cells, although the function of CNTF is not fully understood [15].

Its activities are mediated through a heterotrimeric complex formed by a specific α subunit CNTF receptor (CNTFR α) and two β subunits, leukemia inhibitory factor receptor (LIFR β) and IL-6 signal transducer (gp130). CNTF is able to delay the loss of cells during retinal degeneration by protecting the photoreceptors in 12 animal models [15].

Delivery of CNTF to the retina is a major challenge: encapsulated cell technology (ECT) and the NT-501 implant were developed in some studies [16]. The implant was developed to deliver the drug directly to the retina over a sustained period. The availability of the protein is assured by a stable and long-term secretion that also allows a continuous exposure of the target site. Preliminary results involving the NT-501 ECT implant appear promising,

demonstrating sustained, safe and efficacious delivery of the protein therapy of up to several years in the eye. ECT is a system consisting of human cell lines that is genetically engineered to endogenously express a selected therapeutic protein at a regulated delivery rate and released in situ [16-20]. Many studies on animal models have demonstrated the possibility of using the CNTF as an approach to reduce photoreceptor cell loss. Between February and October 2007, 51 patients with GA were enrolled and randomly assigned to phase 1 and 2 of the studies to evaluate the effect on retinal structure and function (NCT00063765, NCT00447954, NCT00447980, NCT00447993) [17,21]. Patients were randomly assigned to receive a high-dose implant, a low-dose implant or sham surgery in one eye at 2:1:1 ratio. All patients completed the 12-mo endpoint, and no patients dropped out of the study. There was a statistically significant difference in total macular volume in the study eye compared with baseline in the high- or low-dose CNTF groups but not in the sham group [17,21]. The high- and low-dose CNTF groups had a significant increase in total macular volume compared to baseline at all-time points (months 4, 6, and 12) ($p < 0.001$). The high-dose group had a significantly better outcome than the low-dose group at all-time points ($p < 0.05$). These results were associated with an increased width of the outer layer complex, as seen on cross-sectional evaluation of high-resolution line scans. The GA area varied slightly among the 3 groups at baseline but the difference was not statistically significant. The three treatment groups did not show a statistically significant difference in their rate of progression in the GA area. Although no statistically significant improvement in visual acuity was observed across all treatment groups at 12 mo after implant, the CNTF implant appeared to preserve vision. The treatment resulted in a dose-dependent increase in retinal thickness as early as 4 mo after implant and this increase was maintained through 12 mo ($p < 0.001$). The anatomical change was associated with visual function stabilization and the high-dose treated eyes maintained stable vision regardless of baseline BCVA [17,21].

4.2. IMMUNE MODULATING OR ANTI-INFLAMMATORY AGENTS

4.2.1. Lampalizumab

Lampalizumab, previously referred to as anti-complement factor D antibody, anti-factor D, FCFD4514S, RG7417, RO 5490249, or TNX-234, is an antigen-binding fragment (Fab) of a humanized monoclonal antibody (mAb) directed against complement factor D (CFD).

Lampalizumab selectively inhibits CFD-mediated activation and amplification of the alternative complement pathway, but it does not affect initiation of the classical or mannose-binding lectin pathways of the complement system [22]. The Mahalo phase 2 clinical trial (NCT01229215) investigated the safety, tolerability, pharmacokinetics, and evidence of activity of lampalizumab, including targeted genetic analyses, in patients with GA secondary to AMD [23]. This trial enrolled 120 patients, and it demonstrated an acceptable safety profile during the 18-month treatment period. Monthly lampalizumab treatment demonstrated a 20% reduction in lesion area progression versus sham control. A more substantial benefit from the monthly treatment (44% reduction in GA area progression compared to control) was observed in a subgroup of complement factor I (CFI) risk-allele carriers (57% of the patients analyzed were CFI risk-allele carriers). The Mahalo study, published in 2013, showed a potential effect of the treatment in patients with GA and supported therapeutic targeting of the alternative complement pathway for treating AMD pathogenesis [23]. Between August 2014, and October 2016, 906 Chroma (GX29176; NCT02247479) participants and 975 Spectri (GX29185; NCT02247531) participants were randomized to receive sham every 4 weeks (153 Chroma participants; 161 Spectri participants), lampalizumab every 4 weeks (298 Chroma participants; 330 Spectri participants), sham every 6 weeks (152 Chroma participants; 160 Spectri participants), or lampalizumab every 6 weeks (303 Chroma participants; 324 Spectri participants) [24]. No significant differences in the primary end-point of mean change from baseline in GA lesion area at week 48 were identified among the

eyes receiving 10-mg lampalizumab IVTs either every 4 weeks or every 6 weeks vs. sham. These phase 3 trials showed that lampalizumab was ineffective as a treatment of GA secondary to AMD [24].

4.2.2. Zimura

Zimura (ARC-1905) is a polyethylene glycol (PEG), oligonucleotide, chemically synthesized single strand nucleic acid aptamer that targets and inhibits complement factor C5. Inhibition of C5 in the complement cascade prevents the formation of key terminal fragments (C5a and C5b-9). C5b-9 is involved in the formation of the MAC, which causes cellular death through the disruption of the cell membrane [25-27]. Phase 1 trial for dry AMD (NCT00950638), started in 2009 and was completed in 2012. It evaluated the safety and tolerability of intravitreal Zimura injections. Forty-seven participants, 50 years or older, with GA secondary to dry AMD in both eyes were recruited. The study completed with no results posted. The phase 2a trial (NCT03362190) evaluated the safety profile of Zimura administered intravitreally in combination with 0.5 mg of Lucentis, in 65 wet AMD patients who had not previously been administered an anti-vascular endothelial GF (VEGF) drug [28]. A considerably higher percentage of patients receiving the Lucentis–Zimura combination showed improved visual acuity compared with controls of patients receiving Lucentis monotherapy. Later, on October 2018, Ophthotech completed patient enrolment (estimated one hundred-twenty) in its phase 2b clinical trial to evaluate the safety and efficacy of Zimura compared to sham in subjects with autosomal recessive Stargardt disease 1. The company has decided to modify its ongoing phase 2/3 clinical trial of Zimura monotherapy in 200 participants with GA secondary to dry AMD (NCT02686658). The trial has been adjusted to accelerate the deadline by reducing the number of patients, shortening the time for attaining

the primary efficacy endpoint and thereby reducing the cost to complete the study. Estimated primary completion date is November 2019 [28].

4.2.3. POT-4 and APL-2

POT-4 is a cyclic peptide comprising 13 amino acids derived from compstatin, that irreversibly binds to C3 and prevents its proteolytic activation to C3a and C3b; thus, inhibiting the complement pathways and prevent MAC formation. It forms a gel when injected into the vitreous. A phase I clinical trial (NCT00473928) in wet AMD patients was completed in 2010 without safety concerns [29,30]. APL-2 is a modified version of POT-4 designed to have a longer half-life. APL-2 (POT-4/AL-78898°) is a synthetic cyclic peptide conjugated to a PEG polymer that binds specifically to C3, effectively blocking all three pathways of complement activation: classical, lectin, and alternative [31]. It has undergone phase 1 and 2 trials (NCT00473928, NCT02503332), and it showed no safety concerns. The Filly trial is a 246-patient phase 2 multicenter, randomized, single-masked, sham-controlled clinical trial, in patients with GA conducted at over 40 clinical sites, located in the United States, Australia and New Zealand. APL-2 was administered as an IVT monthly or every other month for 12 months, followed by six months of monitoring without active treatment for a total follow up period of 18 months [31]. Apellis Pharmaceuticals reported that APL-2 met its primary endpoint of reducing the growth rate of the GA lesion compared to sham after 12 months of treatment. APL-2 administered monthly via IVT showed a 29% reduction in the rate of GA lesion growth compared to sham after 12 months of treatment. With every other month administration of APL-2, a 20% reduction was observed. After the 12-month period, subjects were followed for a further six months without treatment. During this period of non-treatment, the GA lesions in the previously treated groups grew at a rate similar to sham. Subjects previously treated with monthly APL-2 showed only a 12% reduction over the

six month period compared to sham, while those previously treated with every other month APL-2 showed a 9% reduction compared to sham. Two AMD phase 3 clinical trials, Oaks and Derby (NCT03525613-NCT03525600) are being initiated for the development of APL-2 in the treatment of GA secondary to advanced AMD in 600 participants each. A multi-center, randomized, double-masked, sham controlled study to compare the efficacy and safety of intravitreal APL-2 therapy with sham injections the subject population will consist of subjects with GA secondary to AMD: patients' recruitment is still ongoing and the estimated study completion date is December 2022 [31].

4.2.4. CLG561

CLG561 is an inhibitor of properdin. It acts to stabilize the alternative pathway C3 and C5 convertases by extending the half-lives of the C3 and C5 converting enzymes; based on human genetics as well as pathophysiological features of AMD that implicate complement activation [31]. A phase 1 study (NCT01835015) has ended in 2016; it has evaluated the safety, tolerability, and serum pharmacokinetics of CLG561 in subjects with AMD. A phase 2 study (NCT02515942) started on 2015 enrolling 114 participants, to evaluate the safety and efficacy of 12 (every 28 days) IVTs of CLG561 as a monotherapy and in combination with LFG316 as compared to sham in subjects with GA [31,32]. The study was completed, but the reporting date is on August 2020.

4.2.5. LFG316

LFG316, or Tesidolumab, is a fully human IgG1 targeting complement factor C5 and inhibits the complement system activation [31-33]. A clinical phase 1 dose-escalation and safety study with single IVTs of 0.15–5mg LFG316 were performed in patients with GA or choroidal neovascularization due to AMD (NCT01255462). No adverse effects have been published and

the drug was well-tolerated. A phase 2 study (NCT02515942) started on 2015 enrolling 114 participants, to evaluate the safety and efficacy of 12 (every 28 days) IVTs of CLG561 in combination with LFG316 and CLG561 as a monotherapy and compared to sham in subjects with GA [33,34]. The studies were completed, but the reporting date is on August 2020. In a clinical trial phase 2 testing low doses of LFG316 (NCT01527500), 158 participants with GA were treated. Study was divided into 2 parts: part A evaluated safety & efficacy of multiple 5 mg/50 μ L doses of IVT LFG316 against sham every 28 days for 505 days and part B evaluated the safety and pharmacokinetics of a single IVT dose of 10 mg/100 μ L of LFG316. At the completion of the trial, LFG-316 was found to have an acceptable safety profile, but it was not effective in reducing GA lesion growth rate or improving visual acuity.

4.2.6. Alternative medicine: Iluvien

Iluvien is a sustained-release formulation of fluocinolone acetonide. It is a corticosteroid, a synthetic hydrocortisone derivative [35]. The fluorine substitution at position 9 in the steroid nucleus greatly enhances its activity. It is only approved for the treatment of diabetic macular edema (DME). Iluvien could slow the progression of GA. A total of 40 patients affected bilaterally by GA were recruited in a phase 2 study (NCT00695318). The study was completed, but the results are not yet available [35].

5. DISCUSSION

Nowadays, there are still no treatments that slow progression of dry AMD. Some clinical trials are ongoing, with the goal of finding a viable solution to prevent and/or treat the pathology. Several new drugs have shown some promising results including those targeting the complement cascade and the so-called neuroprotective agents [13-36]. For example, lampalizumab, an antibody directed against complement factor D, could be used for its potential therapeutic effect, but it was disproved by Chroma and Spectry during phase 3

clinical trials [22-24]. At this moment, APL-2, that binds the C3 protein blocking all three pathways of complement activation, may be the most promising molecule [29-32]. The clinical trials Oaks and Derby, enrolling 600 patients each and ongoing on 58 different locations, will be completed in 2022 [29-31]. Similarly, we are awaiting the results on brimonidine, a neuroprotective agent, which will be published in the 2019 [13,14]. No statistically significant improvement in visual acuity was observed across treatment groups after implant of CNTF with NT-501 ECT, a system consisting of human cell lines that is genetically engineered to endogenously express a selected therapeutic protein in situ [16-21]. It is possible to speculate that the aforementioned molecules might be more effective if injected in the initial phases of the macular degeneration, especially because they act as neuroprotectors or immunomodulating/anti-inflammatory agents. We know that the mechanism of action of the neuroprotectors is to prevent degeneration and apoptosis of ganglion cells whilst the mechanism of action for the immunomodulating agents is to block the complement cascade model. An association and complementary pharmacological use may be suggested given the different action mechanism of the two classes of molecules. The main issue the researchers are called to resolve, is when to start the treatment of dry AMD patients. We should be sure not to start too early to avoid aggressive therapies, nor too late for the high risk of losing most of the healthy tissue. Moreover, during more advanced stages of the pathology, new genetic therapies could be able to block the processes that create a biomolecular malfunction inducing the death of retinal cells. In the late stages, after the development of GA and the cell death in the macular area, only new therapies using staminal cells might be able to regenerate lost tissues. On the other hand, we hypothesize that could be important to distinguish different stages of the dry AMD to regulate the effectiveness of the guidelines in the differently affected patients [37,38]. The retinal morphological changes might be used to identify patients early in the course of AMD development who might still be

at a reversible phase of the disease and therefore amenable to intervention. Minimum sizes to define atrophic areas vary; the commonly used Wisconsin Grading System includes lesions $\geq 175 \mu\text{m}$ in diameter. The definitions of non-exudative AMD in the international statistical classification of diseases are in Table 2 [38-43]. A recent consensus of retina international experts has highlighted how multimodal imaging is strongly recommended to measure and classify the different forms of neovascular- and non neovascular-AMD [44]. So, color fundus photography, confocal fundus autofluorescence, confocal near-infrared reflectance, and high-resolution optical coherence tomography volume scans should be acquired at regular intervals throughout the clinical studies [44]. The use of the different therapies depending on the stage might avoid a consequent severe visual loss in old age.

In light of these considerations, we would like to recall our research in which subjects affected by GA have been treated with eye drops based on nerve GF (NGF) [45], or with autologous stem cell transplantation [46-48]. These cells, derived from adipocytes obtained from the abdominal tissue and pretreated platelets, are inserted into a scleral pocket obtained in the affected eye associated with an orbital adipose peduncle to allow the survival of the autograft. It has been ascertained, through the Regen Lab SA of tissue engineering in Switzerland (swissbiotech.org - Swiss Biotech Association), that the autograft produces numerous neurotrophic and angiotrophic GFs [46-48]. For example: VEGF, basic fibroblast GF (bFGF), pigment-epithelium-derived factor (PEDF), macrophage colony-stimulating factor (M-CSF), granulocyte-macrophage colony-stimulating factor (GM-CSF), placental GF (PIGF), platelet-derived GF (PDGF), platelet-derived angiogenesis factor (PDAF), epidermal GF (EGF), insulin-like GF-1 (IGF-1), transforming GF β (TGF- β), hepatocyte GF (HGF), thrombospondin (TSP), IL, and adiponectin [45-47,49]. Through the scleral pocket, where the autograft is housed, GFs can pass through the thin choroidal barrier and reach the retinal

level, determining a beneficial impact in terms of visual acuity and retinal sensitivity, evaluated with microperimetry and ocular electrophysiology [45-48].

Table 2. Definitions of dry age-related macular degeneration (AMD) or non-exudative AMD in the international classification of diseases (ICD) and clinical modification (CM) according to World Health Organization (WHO).

<i>World Health Organization (WHO). ICD-10 2016 [38].</i>	<i>World Health Organization (WHO). ICD-11 Beta Draft (Mortality and Morbidity Statistics); 2017 [40].</i>	<i>American Academy of Ophthalmology (AAO). ICD10-CM: subspecialty ICD-10 decision trees and guides; 2016 [41].</i>
<i>Centers for Disease Control (CDC). International Classification of Diseases, Clinical Modification (ICD-10-CM): 2016 [39].</i>		<i>Centers for Disease Control (CDC). International Classification of Diseases, Tenth Revision, Clinical Modification (ICD-10-CM): 2017 [42].</i>
Nonexudative AMD Any AMD with no choroidal neovascularization, including early/intermediate AMD and geographic atrophy (GA).	Early AMD Consists of a combination of multiple small drusen, few intermediate drusen (63 and 124 μ m in diameter), or retinal pigment epithelial (RPE) abnormalities	Early AMD Combination of multiple small drusen ($\leq 63 \mu$ m), few intermediate drusen (> 63 and $\leq 124 \mu$ m), or RPE abnormalities
	Intermediate AMD Consists of extensive intermediate drusen, at least 1 large druse (125 μ m in diameter), or GA not involving the center of the fovea	Intermediate AMD Extensive intermediate drusen (> 63 and $\leq 124 \mu$ m) or at least 1 large druse ($> 125 \mu$ m)
	Advanced Dry AMD Characterized by 1 of the following (in the absence of other causes): GA of the RPE and choriocapillaris involving the center of the fovea	Advanced Atrophic AMD Without subfoveal involvement GA not involving the center of the fovea
		Advanced Atrophic AMD With subfoveal involvement GA involving the center of the fovea

6. CONCLUSIONS

To the best of our knowledge, and after extensive studies on the matter, we are aware of the fact that there are still many researches that need to be developed on dry AMD in collaboration between researchers. They will have to identify truly effective molecules, understand the practical potential of pluripotent stem cells, and refine gene therapies. In conclusion, we are sure that only in-depth clinical trials will be able to allow the most appropriate and personalized treatments for each individual dry AMD patient.

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