# Overview of Anti-VEGF Treatment Regimens and New Agents in Age-Related Macular Degeneration

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**ABSTRACT**

Age-related macular degeneration (AMD), which can result in central vision loss, is among the leading causes of blindness worldwide. The beginning of the anti-vascular endothelial growth factor (anti-VEGF) era has revolutionized the treatment of AMD. The efficacy of anti-VEGFs (bevacizumab, ranibizumab and aflibercept) has been proven in important clinical studies such as ANCHOR, MARINA and VIEW, leading to the widespread use of these agents in the treatment of AMD. This review focuses on important clinical studies using 3 anti-VEGF agents and their link with real-world evidence including data point specific to our country. Also, the efficacy of anti-VEGF agents and real-life treatment regimens will be reviewed. New treatments under development in the search for the ideal agent for AMD treatment will also be discussed.

**Keywords:** Age-related macular degeneration, Bevacizumab, Ranibizumab and aflibercept.

## INTRODUCTION

Until about ten years ago, it was accepted that the congenital retinal disorders are primary cause of retinal disease-related blindness. However, by advances in medicine and progressively increasing survival rate, the World Health Organization (WHO) has proposed that age-related macular degeneration (AMD) is now leading cause of retinal disease-related blindness.[1] The AMD is a progressive, degenerative disorder of retina characterized by loss of central vision that occurs more frequently by advancing age.

In developed countries, the AMD affects more than 200,000 new patients and accounts for approximately 90% of severe loss of vision.[2] The AMD incidence is increased by prolonged life expectancy and the prevalence has been reported at 1.2-1.7%.[3] Epidemiological studies estimate that the risk for AMD-related progression loss of vision is increased with every 10 years after 50 years. It is estimated that number of patients with AMD will reach 288 million in 2040, which is 196 million today.[4] The AMD is associated with several risk factors including advanced age, genetics, Caucasian race, gender, smoking, systemic hypertension, hyperlipidemia and mutations in immunomodulatory proteins. The YMD is primarily classified in two subgroups including dry form of AMD that accounts for 80-90% of cases and wet form of AMD that accounts for 10-15 of cases but 90% of legal blindness.[4]

Given the progressively increasing numbers, comprehensive studies have been conducted to identify clinical and pathological characteristic, diagnostic tools for early diagnosis and therapeutic modalities for effective treatment in AMD in prior decades.

The AMD pathogenesis hasn't been fully elucidated and several hypotheses have been proposed. One hypothesis advocates that impaired choroidal circulation occurs initially in wet AMD; thus, angiogenic agents, mainly vascular endothelial growth factor (VGEF), are released from retinal pigment epithelium (RPE) cells in response to ischemia and vascular angiogenesis that causes active neovascularization and hyperpermeability play role in the development of wet AMD.[2, 5] Choroidal neovascular membrane (CNV) develops.[5] Although physiological

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angiogenesis is essential for human growth, development and repair, pathogenic angiogenesis leads tumoral growth and tissue damage as a result of tissue hypoxia or inflammation.[2]

In previous studies, it was shown that many factors such as VGEF, placental growth factor (PIGF), insulin-like growth factor (IGF), fibroblast growth factors (FGFs), platelet-derived growth factor (PDGF), transforming growth factor-ß (TGF-ß) and pigment epithelium-derived factor (PEDF) as well as interleukins and angiopeptides play role in any stage of AMD.[6] Although the AMD isn't considered as an inflammatory disease, it has been reported that several immunological alterations, particularly in complement system, are involved in the progression of AMD.[7]

Based on these findings, the AMD is a multifactorial disease in which genetic and environmental factors are involved. Definitive cure is not available. Based on findings that angiogenic agents, mainly VGEF, play key role in the pathogenesis of wet AMD, treatment with agents blocking VGEF has been introduced with promising results. Today, VGEF inhibition is mainstay of wet AMD.[8-10]

The efficacy and safety of the most important agents, namely bevacizumab, ranibizumab and aflibercept, for intravitreal anti-VEGF treatment have been demonstrated in may clinical trials and these agent remain to be choice of treatment in AMD.[8-10]

Regular follow-up and treatment sessions are warranted to maintain efficacy achieved by these agents, raising some problems regarding cost, administration and compliance. In recent years, different treatment protocols have been introduced to minimize such problems and novel protocols are being defined. Although these agents as golden standard have led significant improvements in the AMD treatment when compared previous treatment methods, they primarily suppress the disease and do not offer definitive treatment.[11] Thus, more effective agents are being investigated to further improve visual acuity although anti-VGEF agents remain to be important in the treatment of AMD. In the treatment of AMD, the goal is to achieve best possible visual outcomes while minimizing treatment burden. In this review, we will discuss anti-VGEF agents, treatment protocols and novel agents in the seek for an optimal agent..

**Anti-VEGF agents**

The development of anti-VEGFs has led a change in paradigm which makes these agents standard care in the management of ocular disorders. Bevacizumab (Altuzan®; Roche) is a humanized, recombinant monoclonal antibody that produces inhibition by binding all isoforms of VEGF-A. It is first anti-VGEF developed and approved in the first-line treatment of metastatic colorectal cancer by US Food and Drug Administration (FDA) in 2004.[12] Initially, systemic intravenous bevacizumab was used as off-label agent in the treatment of AMD. However, due to systemic adverse effects in systemic route, ophthalmologists began to use via intravitreal route and achieved good outcomes while eliminating systemic adverse effects. The bevacizuamb is continued to be used as a off-label intravitreal agent in the treatment of retinal vascular disorders.[8] Pegaptanib Na (Macugen® 0.3 mg, Pfizer) is a pegylated aptomer that blocks VEGF-165 isomer. The pegaptanib is the first ophthalmic anti-VGEF agent approved in the treatment of neovascular AMD by FDA in 2004 based on VISION studies.[13] However, its use has become limited over time.

Ranibizumab (Lucentis®; Novartis) is the next anti-VGF inhibitor and smaller molecule than bevacizumab in order to enlace retinal penetration. Its efficacy was shown in the ANCHOR and MARINA studies. The ranibizumab is a humanized, recombinant monoclonal antibody fragment produces inhibition by binding to all isoforms of VGEF-A and approved in the treatment of neovascular AMD by FDA in 2006.[14, 15] Its affinity to VGEF is fairly high.

Aflibercept (EYLEA® 2mg/0.05 ml, Regeneron Pharmaceuticals, New York, USA), the latest anti-VGEF agent, is a humanized, recombinant fusion protein that binds to all isoforms of VGEF-A, PDGF, PIGF and VEGF-B with higher affinity than ranibizumab and bevacizumab and show prolonged effect. It was approved by FDA in 2011. Aflibercept binds VEGF-A with more prolonged and higher affinity and pharmacokinetic features required for VGEF binding shortens dosing frequency.[2, 16] The AMD is the primary indication for which these agents were developed and anti-VGEF use is markedly increased in developing countries based on the promising results. The Anti-VEGFs exert a significant efficacy in the treatment of angiogenic disorders of eye. However, many practical problems have been emerged in the use of these molecules despite dramatic advances in anti-VGEF treatment in last two decades. These include need for frequent injections, higher costs, incomplete response or unresponsiveness in some patients, which promoted development of treatment protocols to maximize visual gain while minimizing costs.

Here, we aimed to present an update about efficacy of 3 anti-VGEF agents, namely bevacizumab, ranibizumab and aflibercept, widely used in the AMD. We presented available evidence from phase 3 studies, real-world studies and studies from Turkey. The aim was to provide

an overview together with clinical perspective to select accurate anti-VGEF agent in the treatment of AMD. We also reviewed novel anti-VGEF agents under investigation to address shortcomings of available agents. There is no effective, definitive treatment method eliminating pathology in AMD which has an increasing incidence and impairs vision severely. Current treatments aim to protect visual acuity. The seek for novel treatments are ongoing.

**Anti-VGEFs in AMD treatment: pivotal studies**

This section aims to demonstrate perspectives of major studies about use and approval of bevacizumab, ranibizumab and aflibercept in the treatment of AMD. Two major phase 3 studies, CATT and IVAN, were conducted to prove efficacy of bevacizumab in the treatment of AMD.[8, 17] Both studies compared visual acuity (VA) by monthly and pro re nata (PRN) bevacizumab and ranibizumab regiments at the end of year 1 and 2. Table 1 summarizes results of these studies. In both studies, bevacizumab showed almost similar efficacy with ranibizumab throughout 24 months.

Two major phase 3 studies, ANCHOR and MARINA, demonstrated efficacy of ranibizumab in VA improvement at year 2.[14, 15] Fixed doses were used in both studies and 2 different doses of ranibizumab (0.3 mg and 0.5 mg) were compared with sham group in patients with AMD. Table 2 summarizes results of these studies. Based on encouraging results of ANCHOR and MARINA studies, researchers focused on reducing treatment burden of monthly injections.[16]

The MARINA and ANCHOR studies demonstrated monthly ranibizumab is highly effective in treatment of AMD; however, sustainability of monthly treatment is not feasible. Thus, recent studies investigated protocols minimal dose, number of visit, effective treatment planning and recurrence prevention that may provide visual gain as much as monthly treatment. Such studies are denoted as individualized treatment protocol. Individualized treatment protocols can be classified in different categories. Available treatment regimens are termed as monthly fixed dose, quarterly therapy, PRN and TREX (treat and extend). The PIER and EXCITE studies, evaluated efficacy of ranibizumab with 3 monthly loading dose, followed by repeated injections every 3 months in AMD. Both studies demonstrated lower success rate when compared to VA stabilization seen achieved by monthly treatment in the ANCHOR and MARINA studies.[18, 19]

In the PIER study, three monthly intravitreal ranibizumab injections followed by repeated monthly intravitreal injections (0.3 mg and 0.5 mg) were performed and ranibizumab groups were compared with sham group. Despite significantly higher visual acuity gain when compared to sham group, the visual acuity gain following loading dose could not be protected at the end of treatment, returning to baseline levels.[18] In the EXCITE study, the

**Table 1:** *Summary of major studies using BVZ in AMD patients.*

| Study | Number of patients | Study type | Frequency of administration | Mean change in letters within one year |
|---|---|---|---|---|
| CATT | 1208 (1 year) | BVZ (1.25 mg) | monthly | 7.3±0.8 vs.. 7.2±0.7 |
| | | RBZ (0.5 mg ) | PRN | 6.1±0.7 vs. 6.4±0.6 |
| IVAN | Number of patients | Study type | Frequency of administration | Mean change in letters within two years |
| | 628 (2 years) | BVZ (1.25 mg) | monthly | −1·37 (p = 0.26) |
| | | RBZ (0.5 mg) | PRN | −1·63 (p = 0.16) |

BVZ: Bevacizumab, RBZ: Ranibizumab; CATT: Comparison of AMD Treatments Trials; IVAN: Inhibition of VEGF in Age-related choroidal Neovascularization; PRN: Pro re nata;

**Table 2:** *Summary of major studies using RBZ in AMD patients.*

| Study | Number of patients | Study type | Frequency of administration | <15 letters vision loss (%) |
|---|---|---|---|---|
| ANCHOR | 423 (2 years) | RBZ (0.3 mg / 0.5mg) vs. Sham group, vertoporfin group | monthly | 94.3%%96.4% vs. 64.3% |
| MARINA | 716 ( 2 years) | RBZ (0.3 mg / 0.5mg) vs. Sham group, | monthly | 94.5%/ 94.6% vs. 62.2% (p <0.001) |

ANCHOR: Anti-VEGF Antibody for the Treatment of Predominantly Classic Choroidal Neovascularization in Age-Related Macular Degeneration; MARINA: Minimally Classic/Occult Trial of the Anti-VEGF Antibody Ranibizumab in the Treatment of Neovascular Age-Related Macular Degeneration; BVZ: Bevacizumab, RBZ: Ranibizumab

patients received quarterly injections and those received monthly injections were compared and visual acuity gain was found to be higher in monthly dose regimen when compared to quarterly dose regimen at the end of month 12.[19]

The efficacy of aflibercept was shown in two important studies, VIEW 1 and VIEW 2. In the studies, it was found that aflibercept given with 3 loading doses followed by bimonthly injections achieved results comparable to monthly ranibizumab injections.[20] However, number of injections was smaller in aflibercept group than ranibizumab group (5 less injection in average). The subgroup analysis of VIEW 2 study including only Asian population showed results which were comparable to those in remaining study population and proven efficacy of aflibercept in Asians.[21] Ina study on 200 eyes, Inoue et al. showed similar efficacy even with 3 loading doses of aflibercept and ranibizumab followed by PRN regimen.[22]

The PRN regimen was investigated in the SUSTAIN and HARBOUR studies. In the PRN regimen, treatment is given as needed. The patients are scheduled for monthly control visits and injection is given if there are findings of activation while injection was delayed if there is no activation. It is a reactive model focusing on lesion activation. In the PRN regimen, treatment is planned with guidance of optical coherence tomography (OCT).[22] The repeated treatment was considered when there was ≥100 µm increase in central retinal thickness on OCT, ≥5 letter loss with macular fluid on OCT, de novo classical CNV area, de novo macular hemorrhage and permanent fluid finding (≥1 month) on OCT. Periodic follow-up is recommended if the activation findings are lacking. In the SUSTAIN study, repeated injections were performed, if needed, after 3 monthly loading doses and no repeated injection was required in 20% of cases.[23] In the HARBOR study comparing PRN regimen and fixed monthly doses, it was shown that optimal results were achieved by PRN ranibizumab (0.5 mg) regimen in majority of patients.[24] It was observed that PRN regimen was not as effective as fixed monthly doses although it improved visual gain when compared to baseline.[25] Nevertheless, it is important in the context of pioneering for individualized treatment and therapeutic efficacy.

Although PRN regimen reduces number of injections in the treatment by all agents, monthly control visits are still required to determine disease recurrence timely. The workload leads a serious burden for both clinicians and patients. This promotes seek for a novel treatment regimen. In TREX, a proactive regimen, monthly treatments are given to all patients until lesion becomes inactive and control examinations are performed under OCT Guidance. If there is no activation in control visits, treatment is given and control interval is prolonged by 2 weeks. In all control visits, treatment is necessarily given even if lesion is inactive and control interval is prolonged by 2 weeks as maximum interval being 3 months. If there is recurrence in control visit, treatment is given and control interval is shortened by 2 weeks. In this treatment paradigm, the aim is to decrease number of control visits and injections. An individualized profile for recurrence time is created by determining safe period where lesion remain dry and treatment is maintained based on the profile, offering individualized treatment. It was first established by Bailey Freund (February, 2016) and presented in literature by Spaide. It was subsequently revised by Gupta.[26]

In the TREC-YBMD study comparing TREX regimen and fixed monthly treatment regimen, anatomic and functional outcomes comparable to fixed monthly treatment were reported with less treatment burden.[10] Currently, the TREX has become the most commonly preferred treatment regimen.[10, 27] For TREX regimen, the major advantages are similar efficacy with less injections, reduction of novel, vision-threatening activations, and treatment planning by identifying an individualized recurrence time profile via definition of safe period where lesion remains dry. Given that the treatment is administered in each visit regardless of activation finding, the disadvantages include increased risk for atrophy, burden of unnecessary injection, uncertainty regarding time for completion of treatment and lack of control visits during prolonged treatment intervals. "Treat-and extend" and "observe and plan" are also attempted regimes allowing individualized treatment.

The introduction of OCT and OCT-angiography in ophthalmology practice due to rapid advances in imaging systems in last decade have incontrovertible effects on emerging of substantial number of regimes. The rapid and reproducible imaging provided by two techniques allowed more effective planning in diagnosis, follow-up and treatment of AMD. The fact that OCT and OCT-angiography have patient-specific features in each treatment demonstrated individualized treatments are contemporary approaches. However, the efforts to identify optimal agent and treatment are ongoing.

**An overview to anti-VGEF agents and treatment regimes for treatment of AMD in Turkey**

As similar to world, the AMD prevalence and number of patients treated have been increasing due to increase in elder population. Three anti-VGEF agents, ranibizumab, aflibercept and, by regulations in national healthcare system in prior year, bevacizumab, are widely used in the treatment of AMD in Turkey. The agent preference

and treatment regimes are determined in accordance to clinical literature and treatment options required are provided to patients as possible. However, the numbers of control visits and injections could not reach to those in randomized, controlled studies as it is the case worldwide. The real-world studies have demonstrated that it is not possible to follow strict follow-up and treatment criteria required in randomized, controlled studies.[17, 28-31] Given the economic burden associated with monthly injections and challenges in the clinical practice, different treatment regimes, mainly PRN, have been attempted worldwide. Many studies showed that frequency of control visits and injections were decreased with PRN regime. Many single-center and national, multi-center studies were conducted to assess efficacy of PRN regime.[29, 31]

As a first national, broad-based AMD study in Turkey, a retrospective, multicenter, intervention, non-comparative real-world study was conducted at 9 tertiary centers from İstanbul and Kocaeli.[32] In the study, data from 883 eyes from 783 AMD patients underwent intravitreal anti-VGEF treatment with PRN regime between January, 2013 and December, 2015 were analyzed. In the study, some patients initially received 3 monthly loading dose of an anti-VGEF agent; followed by PRN regime (3 plus PRN) while remaining patients received single dose of an anti-VGEF agent; followed by PRN regime (1 plus PRN). The first report was published in 2018, including real-world outcomes of anti-VGEF treatment in AMD patients by focusing on number of control visits and injections. It was found that mean numbers of control visits and injections were 6.9 and 4.1 respectively. In most real-world studies, mean number of injections was found as 3-4 while mean number of control visits was found to range from 6 to 12, which are below those warranted for optimal PRN treatment.[29, 31]

In conclusion, as proven by single-center and multicenter studies from different countries, it is challenging to follow strict control and re-treatment criteria required by PRN regime in AMD patients.[29, 31] The above-mentioned study was the first multicenter study from Turkey, which indicated this fact, and data obtained were consistent with world data. In 2020, the same Work Group published a manuscript that compared anatomic and visual changes of two different anti-VGEF agents used with 1 plus PRN and 3 plus PRN regimes. Although both regimes resulted in comparable anatomic outcomes, 3 plus PRN arm revealed positive effect of 3 loading doses on visual gain independent from anti-VGEF agent used.[33]

Currently, the TREX has become most commonly preferred treatment regime worldwide.[10-27] It is also used in many clinics in Turkey and individualized treatment becomes increasingly important over time.

Again, a novel treatment regime has introduced into literature by a study from Turkey and it is apparent that this regime will lead major gains. Karaçorlu et al. from İstanbul Retina Institute (İstanbul, Turkey) defined a novel, risk-driven treatment protocol individualized according to morphological characteristics of CNV lesion and vision of contralateral eye.[34] In their publication, authors reported, by less injection, visual outcomes comparable with those in other accepted treatment regimes. In details, authors retrospectively reviewed 210 eyes from 184 patients treatment-naive patients who were treated using risk-driven, algorithm-assisted protocol based on classification individualize according risk for vision loss and followed at least 2 years. The risk-driven, algorithm-assisted approach assigned patients into 3 groups according to risk assessment based on morphological characteristics of the lesion and visual acuity of contralateral eye(treatment-naive): low-, moderate- and high-risk. The lesion risk definition was made when determining 3 groups, which was classified into two groups. Authors defined low-risk lesion as active classical or occult choroidal neovasculopathy lesion with lesion ≤1 disc area. The lesion was defined as high-risk if neovasculupathy lesion was >1 disc area and there was one of the followings: polypoidal choroidal vasculopathy and retinal angiomatous proliferation. Based on visual acuity of contralateral eyes (≥20/63 or <20/63), 3 patient groups were defined and different treatment protocol was planned for each group.

The low-risk group included cases with low-risk lesion and good visual acuity (≥20/63) at contralateral eye and received "short-term monthly injections". The moderate-risk group included cases with low-risk lesion and poor visual acuity at contralateral eye or high-risk lesions and good visual acuity and received "short-term TREX" protocol. The high-risk group included cases with high-risk lesion and poor visual acuity at contralateral eye and received "extended TREX" protocol.[34]

The patients presented to less control visit and received less injections according to protocol, resulting in reduction in treatment burden. The SUSTAIN study confirmed that 20% of patients did not require re-treatment after 3 monthly injections months and only 33% required one or two additional injections during 12-months follow-up.[23] If these patients were treated with standard monthly or TREX regimes, they would manifest with unnecessary control visits and injections. The study by Karaçorlu et al. demonstrated value of individualization in anti-VGEF treatment since identification of patient population of

20-33% with good treatment response can be translated as it is possible to minimize overtreatment. In particular, loading dose alone may be sufficient in some small lesions without need for treatment in subsequent years. Risk-driven approaches discriminated the limited number of patients by defining as low-risk. The patients without need for frequent injections can continue to receive short-term monthly injections; however, the TREX regime seems more appropriate if recurrence is experienced within a few months. To achieve long-term success, authors proposed a protocol driven by baseline lesion characteristics and risk for loss of vision, in which treatment and follow-up intervals are determined according to treatment response, Authors also recommended to discontinue treatment when anatomic stability is achieved in order to reduce treatment burden and atrophic potential. It is thought that the algorithm offering a clear roadmap for treatment and follow-up will be valuable to reduce CNV recurrence-related complications by improving treatment compliance of the patient.

There are valuable studies from Turkey; however, we only presented recent studies in details.

**Outcomes of clinical trials with real-world data**

The anti-VGEF treatments developed for age-related macular degeneration are used in the clinical practice. Visual data from different clinical trials may not be observed in real-world data. Table 3 summarizes real-world

**Table 3:** *Summary of real-world data for 3 anti-VEGF agents used in AMD management.*

| Anti -VEGF agent | Number of patients and Study duration | VA changes after study (ETDRS) | Result |
|---|---|---|---|
| BVZ | 128, 4 year | Initial VA: 0.66±0.07<br>Improvement at year 1 :0.48±0.05 (p=0.012)<br>End of study, Year 3: Worsening 0.69±0.07 (p=0.6) | Initial VA gain but not persisted |
| RBZ | 20 real-world data in 54 eyes, Meta-analysis, 1 year | Initial VA: 48.8 to 61.6.<br>Mean change in VA: −2.0 to +5.5 letters<br>≥15 letters improvement in 11 study 19±7.5 (mean),12<br>Mean percent of patients with ≤15 letters loss in 12 studies:89±6.5% | Real-world data are more deficient than clinical studies |
| | LUMINOUS , 6241 patients, 5 years | VA gain at year 1 year (letter) ≤3 (n = 537) = +1.6<br>3–6 (n = 1924) = + 3.3<br>>6 injections (n = 918) = + 3.7<br>≥10 injection (n = 224) +5.7 | VA gain depends on number of injections |
| | UK Neovascular AMD Study, Loading dose plus PRN, follow-up up to 7 years | Initial VA= 55.1 letters, VA on month 6=61.4 letters, VA at year 7= -2.6 letters compared to baseline, at the end of year 7, 40% of patients lost ≥ 70 letters | VA outcomes are better than those in clinical studies; correlated with number of injections. Same efficacy could not be protected at year 7 |
| | HELIOS, 267 patients, loading dose plus PRN | Initial ETDRS score= 56.3±14.3 letters<br>ETDRS on month 2.5 = 61.7±14.9<br>ETDRS on month 6= 60.8±15.7<br>ETDRS on month 12 = 58.5±17.8<br>ETDRS on month 24= 53.3±19. | VA outcomes persistently improved on months 3-6 (p<0.0001) but not persisted on month 24. |
| Aflibercept | 109 eyes, 2 years<br>Treat and extend regime | Mean initial VA: 55.9 ± 15 letters<br>Year 1: 61.3 ± 16.9 letters (VA gain, 5.4 letters)<br>Year 2: 61 ± 17.1 letters (VA gain 5.1 ± 14.9 letters) | VA gain persisted at year 1 and 2 by treat and extend regime. |
| | 136 eyes, 2 years, Treat and extend regime | ≥70 letters improvement compared to baseline: 40%–58% (P < .001), On month 24, 98%of eyes ≤35 letters , 10% of eyes, change (P = 0.547) | If number of injections required is fulfilled at year 1, similar results with clinical trial can be achieved but results may vary. |

BVZ: Bevacizumab, RBZ: Ranibizumab; VA: visual acuity; ETDRS: Early Treatment Diabetes Diabetic Retinopathy Study score; HELIOS: Health Economics with Lucentis in Observational Setting

data with 3-dose anti-VGEF treatment used in AMD. The visual gain is comparable among studies comparing real-world data from patients received 3 different anti-VGEF agents. Compared to clinical trials using varying doses of 3 different anti-VGEF agents, studies using "treat and extend" regimes showed more successful results.

In two real-world studies, efficacy was compared between ranibizumab and aflibercept.[35, 36] In both studies, it was aimed to compare visual acuity outcomes and injection frequency with controlled, clinical trials. Table 4 summarizes outcomes.

Both studies confirmed that visual acuity gain and efficacy were comparable for ranibizumab and aflibercept at year 1 and that number of injections for aflibercept was similar to those for ranibizumab at year 1 despite potential advantage of injection every 8 weeks against monthly injections.[35]

In Denmark, a study was initiated during a period where clinicians shifted AMD treatment from ranibizumab to aflibercept in order to demonstrate injection frequency. It was proposed that the shift to aflibercept will reduce injection frequency; however, it was shown that number of injections was increased after shifting aflibercept based on real-world data. The study rule out the hypothesis that aflibercept has longer duration of action and requires less injections when compared to ranibizumab.[37]

The Intelligent Research in Sight (IRIS) study compared 3 anti-VGEF agents regarding efficacy and injection frequency during one-year follow-up. All three agents were prescribed as monotherapy (bevacizumab n = 6723; ranibizumab n = 2749; aflibercept n = 4387). The outcomes showed that all three agents were comparable regarding VA gain (≥3 order improvement) when given over a year. The injection frequency was lowest recorded with bevacizumab (5.9 for bevacizumab, 6.2 for aflibercept and 6..4 for ranibizumab).[38]

All studies conducted confirmed the efficacy of three anti-VGEF agents in the treatment of AMD. However, "treat and extend" regime seems as a good protocol since controlled, monthly injections are feasible based on real-world data. The "treat and extend" regime provides good efficacy as similar to controlled, experimental conditions.

**Agents at horizon and novel anti-VGEF agents**

Based on evidence, approximately 20% of patients receiving repeated doses of anti-VGEF agents experience worsening in vision in both clinical trials and real-world studies. The major challenges include high cost, frequent injections, anxiety about intravitreal intervention, discomfort and time constraints. Such challenges have promoted more advanced, novel treatments.[39] The novel molecules acting on different parts of VGEF-based mechanisms are discussed below.

DARPin (designed ankyrin repeat protein): Abicipar, a DARPin) is synthesized to specifically block all forms VGEF-A with higher potency, smaller molecule size (34 kDa), longer half-life (2 weeks) and low systemic exposure. In a phase 2 study, the Abicipar showed higher efficacy than ranibizumab with lesser dose frequency. Based on these results, 2 phase 3 trials (CEDAR and SEQUOAI) are ongoing.[40]

Brolucizumab: It is a humanized, single-chain antibody fragment with lowest molecular weight among anti-VGEF, which inhibits all VEGF-A isoforms. It is available as 0.5 mL intravitreal injection. Brolicuzamb is in a form eligible for preparation of a solution with high molar concentration that facilitate ocular penetration with low systemic exposure. At high molar concentration, brolucizumab is equivalent to 12 folds of aflibercept dose and 22 folds of ranibizumab dose.[39] The brolucizumab was investigated in phase 2 and 3 trials including patients with wet type AMD and compared with aflibercept. The phase 3 studies, HAWK and HARRIER trials, are large studies including 1800 patients from 400 centers. The two phase 3 studies with similar design compared brolucizumab and aflibercept regarding visual and anatomic outcomes in AMD. The patients were randomly assigned to receive 3 mg (only HAWK) and 6 mg brolucizumab or 2 mg aflibercept. After loading doses 3 monthly injections, brolucizumab was given at every 12 weeks and dose interval was adjusted as 8 weeks if disease activity was present. At the end of week

**Table 4:** *Summary of real-world data in RBZ vs. aflibercept.*

| Study | Number of patients | VA at year 1 | Injection frequency at year 1 |
|---|---|---|---|
| Lotery et al. [33] | RBZ n = 3350 Aflibercept n =4300 | VA letter score change -0.30 -0.19; p = 0.81 | 6.7 vs. 7.7 |
| Gilles[34] | RBZ n = 197 Aflibercept n =197 | VA gain compared to baseline +3.7 (p = 0.001) +4.26 (P < 0.001) | 8.1 vs. 8.0 |

48, visual acuity gain was similar to aflibercept arm in both brolicizuamb arms. Of the eyes in 6 mg brolucizumab arm, more than 50% (56% in HAWK trial and %51 in HARRIER trial) received treatment at every 12 weeks throughout 48-weeks study period. Anatomic outcomes were found to be better in brolucizumab arm. The published outcomes for week 96 were supportive for those at week 48.[40] In brolucizumab, overall safety was found to be comparable with aflibercept.[40] The brolucizumab is beyond available features of anti-VGEFs regarding structure, function and adverse effects. However, further studies are needed. MERLIN study including AMD cases with persistent retinal fluid will provide further understanding.

Conbercept: It is a recombinant anti-VGEF fusion protein similar to aflibercept which was approved for wet type AMD in China. It blocks all forms of VGEF-A as well as VGEF-C and PIGF.[41,42]

OPT-302: The OPT-302 targets VEGF-C and VGEF-D blockade; thus, it may play role in anti-VGEF treatment. Favorable results were revealed by phase 1 study comparing OPT-302 with ranibizumab. Future trials are awaited.[39]

Small interfering RNA (siRNA): Small RNA molecules involves 2 RNA sequences. It induces short-term inhibition of protein encoding genes, preventing VGEF production. Bevasiranib, first siRNA for ocular use, was assessed in the treatment of neovascular AMD in a phase 3 study. However, trial was prematurely stopped due to lack of efficiency.[43]

Sphingosine-1-phosphate (S1P) antibody: It is an antibody developed against S1P. The S1P is produced by RPE in retina. It play role in neovascular AMD-related pathological angiogenesis, vascular permeability, inflammatory responses and fibrosis.[43]

**Anti-VGEF and anti-PDGF combination**

**Anti- VEGF and anti- PDGF combination therapy**

Pericytes and endothelial cells have common basal membrane. Thus, pericytes do not only encase endothelial cells but they also transmit VGEF-A and growth factors via paracrine signaling. The receptors of neovascular endothelial cells may be inaccessible to anti-VGEFs due to protection provided by pericytes, resulting reduced efficacy of and resistance to anti-VGEF agents. The PDGF play role in pericyte uptake, maturation and survival; thus, PDGF inhibition will enhance endothelial cell-anti-VEGF contact and anti-VGEF efficacy PDGF may also have role in subretinal fibrosis. The PDGF blockade may decrease blindness secondary to AMD.[39]

Pegpleranib: Pegleranib is a 32-mer pegylated DNA aptamer that prevents interaction of PDGF with receptors over pericytes. In a phase 2 study, pegpleranib plus ranibizumab showed positive results in AMD patients; however, the positive effects were not seen in phase 3 study.

Rinucimab: Rinucumab is anti-PDGF receptor-beta antibody. In a phase 2 study, rinucumab used with aflibercept did not provide additional benefit when compared to aflibercept alone.

DE-120: A third anti-PDGF inhibits both VGEF and PDGF and is under investigation in phase 2 trials.

X-82: It is an oral anti-PDGF and VEGF-A inhibitor and a phase 2 study comparing its efficacy with aflibercept.[39]

Anti- VEGF plus Tie-2 receptor modulator

Likewise VEGF, Tie-2 tyrosine kinase receptor is also released from endothelial cells. The Tie-2 receptor activation results in limited vascular permeability. Angiopoietin-1 (Ang-1) is an activator for Tie-2 receptor while angiopoietin-2 (Ang-2) is a competitive antagonist of Ang-1. Since it is thought that elevated Ang-2 level leads an increase in vascular leakage, agents inhibiting Ang-2 have been developed; thus, more Ang-1 will activate Tie-receptor and reduce vascular permeability.

Nesvacumab: It is aAng-2 inhibitory which showed promising results in improving visual outcomes in combination with aflibercept.

RG7716: It is a bi-specific cross-linked monoclonal antibody which has affinity to both VGEF-A and Ang-2. A phase 2 study showed favorable results and a phase 2 study is considered.[39]

VE-PTP: Another enzyme, namely vascular endothelial protein tyrosine phosphatase (VE-PTP) plays role in the Tie-2 receptor activation. Accordingly, a novel agent can be developed that prevents Tie-2 receptor inactivation and stabilizes vascularity and limits leakage.[39]

**Topical therapies**

The topical use of Anti-VEGFs were assessed. Table 5 presents outcomes as well as development process. Based on promising results, topical route may be considered as an alternative solution for ocular angiogenic disorder in the near future.

ARP-1536: ARP-1536 is another novel monoclonal antibody that is investigated in preclinical studies and inhibits VE-PTP via alternative route which results in

| **Table 5:** *Topical formulations in advance.* | | |
|---|---|---|
| Topical agent | Definition | Development stage |
| Pazopanib | TKI of VEGF-A/PDGF | Phase 2b results - comparable with ranibizumab. |
| PAN-90806 | TKI of VEGF-A/PDGF | Phase 1/2 -comparable efficacy was detected for currently available anti-VGEFs in patients with neovascular AMD |
| Squalamine lactate | Prevents down-signaling of multiple angiogenic factors | Combination with ranibizumab provided positive results |
| Regorafenib | A multi-kinase inhibitor acting on intracellular signaling of VEGF-A and PDGF receptors | No efficacy in Phase 2a DREAM study<br>No ongoing trial. |
| TKI: Tyrosine kinase inhibitor; VEGF: Vascular endothelial growth factor; PDGF: Platelet-derived growth factor | | |

increased Tie-2 receptor activation by VE-PTP binding to cell surface.[39]

### Extended drug formulations

Frequent intravitreal injections are concern for management of intraocular conditions and risk for eyes. Thus, studies focuses on novel sustained-release formulations.[39, 44] Biodegradable implants, biodegradable microspheres, encapsulated cells and gene therapy are among newly developed approaches.[44]

Gene therapy may be a novel treatment modality by cellular transcription via viral vectors. They have potential to produce VEGF-A continuously by a single injection.[39] The RetinosStat is a system that increases endogenous endostatin and angiostatin using lentiviral technology. Both factors inhibit ocular neovascularization in preclinical studies.[45]

Table 6 summarizes other agents. The favorable visual outcomes in available anti-VGEF treatments have resulted development of new advances that will address concerns about available treatment in the management of AMD.

### Immunosuppression

Sirolimus (rapamycin) is an immunosuppressant agent used after organ transplantation. There are studies indicating that the agent used in resistant uveitis may also be used in neovascular AMD.[46]

### Radiation therapy

Radiation was previously tried in the management of AMD, however, it could not reach widespread use due to practical challenges.[47] In addition, radiation therapies allowing treatment at safer and lower radiation doses such as epimacular brachytherapy and stereotactic radiotherapy

| **Table 6:** *Sustained-release implant formulations.* | | |
|---|---|---|
| Topical agent | Definition | Development stage |
| Ranibizumab Port Distribution system [10,43] | Non-biodegradable sustained drug delivery implant | • In phase 1 studies, visual improvement was achieved in AMD patients<br>• There is an ongoing phase 2 trial (LADDER) in AMD patients |
| GB-102 (Sunitinib)[10] | Multi-target TKI - inhibits both VGEF-A and PDGF<br>Designed as a injectable depot twice yearly.<br>Encapsulated agent from polymer nano-particles with slow solubility | More effective than aflibercept in preclinical studies. |
| NT-503 Encapsulated Cell Therapy (ECT) implant [10] | A novel VEGF-A receptor fusion protein.<br>An intravitreal implant with sustained release for at least 2 years via ECT | Prematurely discontinued in phase 2 study |
| Hydrogel anti-VEGF depot [10] | Hydrogel has a tight mesh form that is consisted of anti-VGEF-A with slow solubility and allows sustained drug release | No ongoing phase 1 study. |
| Durasert[10] | A degradable implant providing TKIs that can inhibit both VEGF-A and PDGF. | Preclinical phase |
| ENV1305[10] | Nano-particle technology is used for better control of drug release | Preclinical studies are ongoing |
| TKI: Tyrosine kinase inhibitor; VEGF: Vascular endothelial growth factor; PDGF: platelet-derived growth factor; AMD: Age-related diabetic macular edema; DME: Diabetic Macular Edema | | |

provided good outcomes.[48,49] In a study, it was reported that visual outcomes were similar to intravitreal ranibizumab but anatomic outcomes were poorer in radiotherapy group.[50] Thus, radiotherapy is not recommended as initial treatment in cases with wet type AMD.

## CONCLUSION

AMD is an one of the major causes of vision loss worldwide. Recent advances in pharmacotherapy and increased use of anti-VGEF agents have led benefit for patients. Anti-VGEFs are current standard of treatment in AMD patients. Bevacizumab, ranibizumab and aflibercept are most commonly used anti-VGEF agents and have well-known efficacy and tolerability. However, long-term (>2 years) safety and tolerability remain to be unclear. Real-world data indicate that regular treatment with monthly or bimonthly injections is failed in these agents and lead higher costs. Another important point is that some patients experience progression in vision los despite anti-VGEF treatment. This is one of the main reasons underlying development of numerous new and more effective therapeutic agents. There are ongoing clinical trials on novel agents. These studies have potential to lead better visual outcomes in patients with AMD.

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