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REVIEW PAPER

Aflibercept administration in neovascular age-related macular degeneration refractory to previous anti-vascular endothelial growth factor drugs: a critical review and new possible approaches to move forward

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Abstract
Purpose The recent introduction of anti-VEGF drugs has widely changed the prognosis of exudative age-related macular degeneration (AMD), even if a variable percentage of patients showed an insufficient response. Aflibercept is a new anti-VEGF drug approved by FDA for the treatment of exudative AMD with a wider binding capacity than either bevacizumab or ranibizumab. Therefore, the purposes were as follows: (i) to report anatomical and functional outcomes of switching from bevacizumab/ranibizumab to aflibercept previously described in the scientific literature, (ii) to hypothesize the possible pathophysiological mechanisms of the resistance and tachyphylaxis to anti-VEGF drugs, and (iii) to suggest possible clinical actions to increase the chances of success for such difficult cases.
Methods We reviewed the available scientific literature in Medline, Cochrane database, Current Contents, PubMed, and cross-referencing from identified articles, regarding the treatment of exudative AMD patients refractory to bevacizumab and/or ranibizumab and switched to aflibercept monotherapy. We included in this review all the cases in which the diagnosis of refractory or resistant exudative AMD was properly made, and the results of at least one aflibercept injection were described.
Findings We reported the outcomes of 21 papers for a total of 1066 eyes affected by exudative AMD resistant to previous anti-VEGF drug injections and switched to aflibercept. Enrolled reports were divided into two groups: 5 prospective reports and 16 retrospective reports. All the reported papers conclude their analysis, stating that switching from bevacizumab/ranibizumab to aflibercept injections can improve outcomes successfully in refractory neovascular AMD patients.
Implications Analysis of the papers reported in this review demonstrates that switching from bevacizumab/ranibizumab to aflibercept injections can improve outcomes successfully in refractory neovascular AMD patients. The mechanism for these effects is not yet completely understood.

Keywords Aflibercept · Resistant wet AMD · Recurrent exudative AMD · Intravitreal injections · Anti-VEGF drug

Introduction

Age-related macular degeneration (AMD) is the most common disease causing irreversible visual loss in patients of the industrialized countries over the age of 50 years [1]. The neovascular (wet or exudative) form is the most devastating configuration of AMD. Its main characteristic is the neovascularization process into sub-retinal and/or sub-retinal pigment epithelium (sub-RPE) spaces with consequent exudation and bleeding, followed by the formation of a scar in the macular region with loss of the central vision [2]. The emblem of the neovascular form is the choroidal neovascularization (CNV), and the vascular endothelial growth factor (VEGF) plays a key role in its pathogenesis.

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Two landmark clinical trials, using intravitreal injections of an anti-VEGF-A antibody fragment ranibizumab in eyes with new-onset neovascular AMD, showed that over 90 % of treated eyes lost fewer than 15 Early Treatment Diabetic Retinopathy Study (ETDRS) letters after 2 years of monthly treatment, and the visual acuity could be improved in 33.3–40.0 % of treated eyes [3, 4]. Moreover, a recent randomized, multicenter, 24-month clinical trial revealed that bevacizumab, an anti-VEGF-A monoclonal antibody, was not inferior if compared to ranibizumab for the treatment of wet AMD [5].

However, despite the good results obtained, the CATT study showed that optical coherence tomography (OCT)-detectable macular fluid was present in 53.2 and 51.5 % of patients with monthly ranibizumab therapy after 1 and 2 years of therapy, respectively [5, 6]. Less frequent injections, pro re nata (PRN) protocol or bevacizumab use resulted in even greater percentages of patients with persistent retinal fluid [5]. This persistent macular fluid may be responsible for the ongoing visual impairment [5].

Aflibercept (or VEGF Trap-Eye) is a new anti-VEGF drug approved by FDA on November 18, 2011 [7]. It represents a recombinant decoy fusion protein with the two extracellular binding domains of VEGF receptors 1 (domain 2) and 2 (domain 3), neutralizing the biological activity of the pro-angiogenic factors VEGF-A, VEGF-B and placental growth factor (PlGF), which are present in the retina [8]. The presence of the extracellular regions of both VEGF receptors enables aflibercept to have a wider binding capacity than either bevacizumab or ranibizumab. Moreover, it has been hypothesized that this pharmacological characteristic could allow a more persistent, long-lasting VEGF blockade [8–11].

The VEGF Trap-Eye: Investigation of efficacy and safety in wet AMD (VIEW) 1 and VIEW 2 studies demonstrated that aflibercept, given every 2 months after a loading phase of 3 monthly intravitreal injections, was not inferior to monthly ranibizumab in maintaining visual function in patients with treatment-naïve neovascular AMD over 1-year period [7]. These results were maintained over the 96 weeks of the study [12].

However, information on long-term functional outcomes of anti-VEGF therapy in neovascular AMD is few and scattered because they are limited by the decrease in patient population in follow-up. A 7-year follow-up study on ranibizumab showed a mean vision loss $\geq$8 ETDRS letters [13]. Moreover, 34 % of patients had a loss of at least 3 lines in vision in the final visit [13]. The authors concluded that the vision loss in long-term follow-up is probably multifactorial [13]. Indeed, this vision loss could be due to pathological processes and pharmacological determinants such as the natural progression of the underlying non-neovascular AMD, too few injections of anti-VEGF drugs despite a still active disease, and, above all, the loss of treatment effectiveness over time [14]. Several studies have shown that a reduced anatomical response can be found for both ranibizumab and bevacizumab in neovascular AMD over time [15, 16]. Currently, such long follow-up has not been reported for aflibercept.

Different terms have been used to describe this clinical finding, ranging from tolerance or tachyphylaxis to resistance or not (insufficient or low) responsiveness [14, 17]. In general, in case of chronic administration of medications the decrease in pharmacological effect is a major concern that can limit the long-term efficacy. This phenomenon has been termed *tachyphylaxis*, referring to a progressive decrease in therapeutic response after repetitive administrations of a pharmacologically active substance [18]. On the other hand, *innate resistance* may be defined as cases that do not respond well functionally and/or morphologically since the first administration of the drug [19].

To overcome both *innate resistance* and *tachyphylaxis*, several treatment strategies have been proposed, such as switching to other anti-VEGF therapies [20] or increasing the concentration of the drug [21, 22] or the frequency of the injections [9]. Indeed, neovascular AMD patients refractory to standard ranibizumab doses (0.5 mg/0.05 ml), who were treated with higher dosage (ranibizumab 2.0 mg/0.05 ml), showed both anatomical and functional response [21, 22].

Gasperini and colleagues reported a significant decrease in sub-retinal fluid in the majority of tachyphylactic patients after switching treatments from either ranibizumab or bevacizumab to bevacizumab or ranibizumab, respectively [20]. The study showed that 81 % of the patients responded favorably after changing intravitreal injection medication, even if functional success was limited [20].

In this review, we specifically examine published articles reporting the influence of switching therapy from bevacizumab and/or ranibizumab to aflibercept in patients affected by resistant neovascular AMD. We hypothesized possible mechanisms responsible for the onset of *tachyphylaxis* or the presence of *innate resistance* to current available therapies. Moreover, treatment options to manage neovascular AMD patients refractory to "old" anti-VEGF drugs are proposed.

Methods

Search strategy and selection criteria

Data were identified by searches in Medline, Cochrane database, Current Contents, PubMed, and cross-referencing from identified articles; some articles were identified

through searches of the extensive files of the authors. Search terms for the online research were as follows: "intravitreal aflibercept and age-related macular degeneration," "VEGF Trap-Eye," "VEGF Trap-Eye and age-related macular degeneration," "aflibercept," "aflibercept and macular diseases," "Eylea," "Vascular Endothelial Growth Factor Trap-Eye," "insufficient responders to anti-VEGF," "not responders to anti-VEGF," "recurrent wet AMD," "resistant wet AMD," English language papers were reviewed.

All cases of "naïve AMD patients" were excluded. We reviewed the available scientific literature regarding the treatment of exudative AMD patients refractory to bevacizumab and/or ranibizumab and switched to aflibercept monotherapy. We included in this review all the cases in which the diagnosis of refractory or resistant exudative AMD was properly made, and the results of at least one aflibercept injection were described. Published literature has been reviewed until February 17, 2015.

Cases of polypoidal choroidal vasculopathy (PCV) were excluded from the analysis [23–27]. PCV is characterized by polyp-like lesions at the end of choroidal vessels. PCV is a common type of exudative AMD in the Asian population [28, 29]. Although typical AMD and PCV have been classified in the same category of exudative AMD [30], some of the PCV cases have the unique characteristic of choroidal vascular hyperpermeability [31, 32]. Moreover, the efficacy of intravitreal ranibizumab to treat PCV has been described to be lower than that for typical AMD [33–35]. In particular, PCV cases with choroidal hyperpermeability seem to be not related to VEGF, and thus, they may not respond favorably to anti-VEGF monotherapy [36].

Ho et al. reported the results of 96 eyes, one of which was a PCV, but they did not specify the outcome of this single eye, and consequently, it was not possible to separate this case from the overall analysis [37]. They also included 5 eyes (5.2 %) previously treated with verteporfin photodynamic therapy (V-PDT) and 1 eye with pegaptanib sodium injections [37]. Also, Kumar et al. [38] included in their analysis 5 eyes (14.71 %) previously treated with V-PDT, while Grewal et al. [39] included only 1 patient previously treated with one PDT. Finally, Yonekawa et al. [40] analyzed 6 eyes (5.88 %) previously treated with PDT, 1 eye (0.98 %) with thermal laser, and 2 eyes (1.96 %) with pegaptanib sodium. However, given the low percentages and the inability to extrapolate the individual results from the overall analysis, we decided to include these clinical studies in the analysis of the current review. Moreover, Kawashima et al. [41] reported the outcomes of both resistant neovascular AMD and PCV patients after switching the therapy from ranibizumab to aflibercept; however, they divided the analysis into two groups and reported outcomes from each group, so it was possible to extrapolate the outcomes from only neovascular AMD patients, which have been added in this review.

Also, the work of Fujii et al. [42] was not considered for the analysis because all the 3 cases of refractory nAMD reported in the article (one was a PCV) were complicated by retinal epithelial tear. Hanh reported a case in a vitrectomized eye, and it was excluded for this reason [43].

In the work of Rusu et al. [44], the authors focused their attention only on the modifications of intraocular pressure after switching the therapy, so it was not possible to include this article in our review because of the lack of clinical outcomes.

Broadhead et al. [45] analyzed the response of pigment epithelial detachment (PED) to intravitreal aflibercept among patients with treatment-resistant neovascular AMD in the same sample of the work of Chang et al., and it was excluded from our analysis. Finally, the case report of de Oliveira et al. [46] was excluded because they used ziv-aflibercept (Zaltrap; Sanofi-Aventis, Paris, France), which is different from aflibercept, because it is hyperosmolar (1000 mOsm/L) relative to the vitreous, while aflibercept is iso-osmolar.

Most of the reports were retrospective studies [37, 38, 40, 47–59], whereas only five were prospective trials [39, 41, 60–62].

Results

Using strict a priori criteria for this review, we identified a total of 21 papers. We selected two different groups: the first group included prospective works [39, 41, 60–62] and the second group included retrospective works [37, 38, 40, 47–59]. The results of our review are summarized in Tables 1, 2, 3, 4, 5, 6, 7, and 8.

Prospective reports

Demographics (Table 1)

A review of the literature revealed 5 prospective reports for a total of 157 eyes affected by exudative AMD previously treated with anti-VEGF intravitreal injections and switched to aflibercept intravitreal treatment (mean age 78.13 years; 65 males, 66 females and 26 gender unknown) [39, 41, 60–62]. Laterality was reported only in one paper (23 right eyes and 26 left eyes) [61], while in the other works it was not reported [39, 41, 60, 62]. Only Chang et al. [61] reported the lens status (32 phakic eyes and 17 pseudophakic eyes). Follow-up duration ranged from a minimum of 6 months [41, 60–62] to a maximum of 12 months [39] after the first aflibercept injection. Inclusion and exclusion criteria, as well as the definition of refractory or

Table 1 Prospective clinical trials: demographics

Reference	No. of patients/ N° eyes	Age (years) (range or ±SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status
Kawashima [41]	15/15	76.9 ± 8.0	13/2	N/A	Neovascular AMD refractory to previous ranibizumab injections	Neovascular AMD refractory to previous ranibizumab injections for longer than 6 months and showed recurrent or residual exudative changes after the last 3 injections	PDT performed within 6 months of the conversion; follow-up less than 6 months after the conversion	N/A	6 months	N/A
Wycoff [60]	46/46	77.8 (range 55–95)	22/24	N/A	Neovascular AMD refractory to previous anti-VEGF drugs	The presence of edema; minimum follow-up of 24 months before starting treatment with aflibercept injections (only patients who completed the 2 years, prospective SAVE trial in which recalcitrant wet AMD eyes were treated with 2.0 mg ranibizumab were eligible); 28-day minimum "washout" before enrollment	Significant sub-retinal fibrosis or geographic atrophy involving the fovea; any history of vitrectomy surgery, previous treatment PDT, any previous radiation treatment, any previous intravitreal drug delivery aside from ranibizumab or bevacizumab; sub-retinal hemorrhage involving the central fovea > 1 disk area (2.54 mm^2), or previous retinal pigment epithelial tear	Intraretinal fluid 17/37 %, sub-retinal fluid 21/46 %, sub-RPE fluid 18/39 %	6 months	N/A
Chang [61]	49/49	77.8 ± 7.5	21/28	23/26	Neovascular AMD refractory to previous anti-VEGF drugs	Al least 4 anti-VEGF injections in the last 6 months before the enrollment and persistent intraretinal and/or sub-retinal fluid on OCT during this period; BCVA between 35 and 90 ETDRS letters (equivalent to 6/60 to 60/60 Snellen acuity)	PCV; uncontrolled IOP (> 25 mmHg); current vitreous hemorrhage or inflammation; prior vitrectomy or AMD-related surgery or any intraocular surgery within 2 months of study commencement; anti-VEGF therapy within the previous 30 days; PDT within the previous 90 days or more than 6 prior PDT; intravitreal TA within the previous 180 days; significant sub-retinal fibrosis or geographic atrophy; significant corneal or lenticular opacities; myocardial infarction, transient ischemic attack or cerebrovascular accident within the previous 90 days; current pregnancy or lactation	N/A	6 months	32 phakic (31 with cataract); 17 pseudophakic

Table 1 continued

Reference	No. of patients/ N° eyes	Age (years) (range or ±SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status
Singh [62]	26/26	78 (range 69–90)	N/A	N/A	Active neovascular AMD that had an initial response on OCT defined as a decrease in retinal edema and/or sub-retinal fluid to anti-VEGF injections followed by recurrent increase in fluid on OCT (defined as intraretinal, cystoid, sub-retinal fluid, or worsening PED)	Active sub-foveal CNV secondary to exudative AMD confirmed by FA; BCVA between 25 and 80 ETDRS letters; at least one prior injection of 1.25 mg bevacizumab or 0.5 mg ranibizumab within 3 months of enrollment; initial response on OCT followed by recurrent increase in fluid on OCT	Any prior concomitant therapy with another investigational agent to treat neovascular AMD in the study eye; history of vitrectomy, trabeculectomy, surgery for retinal detachment, or any intraocular or periocular surgery in the study eye (within 3 months of day 1); prior treatment with PDT in the study eye; previous investigational treatments for AMD,; history of sub-foveal laser photocoagulation, uncontrolled glaucoma or uveitis; prior systemic anti-VEGF within 3 months of day 1; history of vascular diseases affecting the retina within the study eye.	N/A	6 months	N/A
Grewal [39]	21/21	80.7 ± 4.5	9/12	N/A	Recalcitrant exudative AMD defined	Recalcitrant exudative AMD defined as CNV secondary to AMD with OCT documentation of sub-retinal and/or intraretinal fluid and/or sub-RPE fluid with adjacent to sub-retinal/intraretinal fluid following >6 months of monthly anti-VEGF treatment.	Idiopathic PCV and CSR; anti-VEGF therapy <28 days prior, PDT <3 months prior or >4 prior PDT treatments; significant sub-foveal fibrosis (>50 % of lesion); prior TA (<6 months), intraocular surgery (<2 months); history of vitrectomy; active intraocular inflammation; vitreous hemorrhage; sub-retinal hemorrhage involving >1 disk area of central foveal; previous RPE tear; or BCVA < 20/400	N/A	12 months	N/A

[a] Fassnacht-Riederle et al., Ho et al. and Kumar et al. defined sex on the number of enrolled eyes and not on the number of the enrolled patients

SD standard deviation, *M/F* male/female, *R/L* right eye/left eye, *N/A* not available, *Anti-VEGF* anti-vascular endothelial growth factor, *AMD* age-related macular degeneration, *PDT* photodynamic therapy, *Sub-RPE* sub-retinal pigment epithelial, *OCT* optical coherence tomography, *BCVA* best-corrected visual acuity, *ETDRS* early treatment diabetic retinopathy study, *PCV* polypoidal choroidal vasculopathy, *IOP* intraocular pressure, *TA* triamcinolone acetonide, *PED* pigment epithelial detachment, *CNV* choroidal neovascularization, *FA* fluorescein angiography, *CSR* central serous retinopathy, *RAP* retinal angiomatous proliferation

Table 2 Retrospective clinical trials: demographics

Reference	N° patients/ N° eyes	Age (years) (range or ± SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status (Phakic/ pseudophakic)
Michalewsky [59]	23/23	72	6/17	N/A	Neovascular AMD with persistent intraretinal or sub-retinal fluid	Neovascular AMD with persistent intraretinal and/or sub-retinal fluid despite at least 6 previous intravitreal injections of bevacizumab	N/A	13 eyes were type 1 CNV (3 with PED); 8 eyes were type 2 lesion; 2 eyes were type 3 lesion. nine eyes with persistent intraretinal fluid; 8 eyes with persistent sub-retinal fluid; 3 eyes with both intraretinal and sub-retinal fluid; and 3 eyes with PED	1 month	N/A
Eadie [58]	63/67	79.9	20/43	N/A	Neovascular AMD with persistent exudation on OCT	A minimum of 3 injections of ranibizumab or bevacizumab; evidence of persistent exudation (any retinal thickening with signs of active exudation such as cystic spaces, sub-retinal fluid, or serous PED)	Retinal thickening due to sub-retinal fibrosis with no signs of activity	N/A	N/A	N/A
Thorell [57]	65/73	76.2 ± 8.7	22/43	N/A	Neovascular AMD with persistent or recurrent intraretinal or sub-retinal fluid	At least 12 months of previous treatment with bevacizumab and/or ranibizumab due to persistent or recurrent intraretinal or sub-retinal macular fluid	Follow-up visits performed outside the clinic; incomplete follow-up; concomitant retinal pathology that could interfere with interpretation of outcomes such as history of vitreoretinal surgery or laser	PED in 70 eyes: 38 predominantly serous and 32 predominantly fibrovascular	6 months	N/A
Griffin [56]	47/47	80.5 ± 8.02 (59–98)	20/27	27/20	Neovascular AMD resistant to at least 3 previous injections with either ranibizumab or bevacizumab	Age > 55 years; a minimum of 3 intravitreal injections of bevacizumab and/or ranibizumab; eyes considered treatment resistant (excluding partial responders); baseline visit that was recorded immediately prior to the first aflibercept injection	Dry OCT images at any time during the three injections prior to conversion to aflibercept; elapsed time between prior treatment and the switch to aflibercept exceeded 63 days; interruption of aflibercept therapy before the first three injections	N/A	3 months	N/A

Table 2 continued

Reference	N° patients/ N° eyes	Age (years) (range or ± SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status (Phakic/ pseudophakic)
Arcinue [55]	58/63	Median 81 (range 76–87)	24/34	31/32	Neovascular AMD resistant (multiple recurrences or persistent exudation) to bevacizumab or ranibizumab monotherapy	Neovascular AMD defined as resistant to previous anti-VEGF therapy. Resistance was defined as having multiple recurrences (minimum of 2 recurrences after the eyes have been completely dry following a series of at least 3 monthly injections per treatment cycle) or persistence of exudation (poor response to monthly ranibizumab or bevacizumab for at least 5 months) as evident on clinical examination and on imaging studies (leakage on FA, or fibrovascular PED with IRF or SRF on OCT) while on monthly anti-VEGF monotherapy. Recurrence was diagnosed as new or increased IRF or SRF with or without vision changes or symptoms.	Poor compliance; previous aflibercept injections elsewhere prior to the first aflibercept injection at the institute of the authors; other retinal conditions other than typical AMD (macular hole, vitreomacular traction, epiretinal membrane, retinal detachment, PCV, pseudovitelliform macular dystrophy, peripapillary CNV)	IRF 38/60.3 % SRF 30/47.6 % IRF + SRF 5/7.9 %	12 months	19/44
Gharbiya [54]	30/31	70.1 ± 8.1 (range 60–86)	9/21	N/A	Neovascular AMD characterized by at least 6 monthly anti-VEGF injections in the last 6 months	Neovascular AMD with persistent intraretinal or sub-retinal fluid with or without PED; at least six consecutive monthly injections with ranibizumab before aflibercept initiation: interval between the last bevacizumab or ranibizumab injection and the first aflibercept greater than 4 weeks and smaller than 6 weeks; at least six months of follow-up on a monthly basis	Prior treatment with PDT; diagnosis of RAP or idiopathic PCV; any ocular disease that could affect BCVA; history of intraocular surgery except for uncomplicated phacoemulsification performed within the preceding 6 months; any systemic condition contraindicating the use of intravitreal anti-VEGF agents.	IRF 27/87 %, SRF 19/61 %, any retinal fluid 31/100 %, hollow PED 8/30 %, solid PED 7/22 %, mixed PED 19/70 %	6 months	N/A

Table 2 continued

Reference	N° patients/ N° eyes	Age (years) (range or ± SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status (Phakic/ pseudophakic)
Messenger [53]	109/109	80.3 (range 59–96)	48/61	N/A	Neovascular AMD treated with at least 12 months of prior anti-VEGF injections	Neovascular AMD treated with at least 12 months of prior anti-VEGF therapy with ranibizumab or bevacizumab (cases were only included if they had received 1 year of anti-VEGF therapy prior to conversion with a "baseline study visit" defined as the visit nearest to 12 months prior to conversion and an allowable range of 10–14 months prior to conversion); BCVA at conversion $\geq$ 20/400; 12 months of follow-up after first aflibercept injection without switching therapy (final "study visit" defined as the visit nearest to 12 months following conversion and an allowable range 10–14 months)	N/A	Intraretinal fluid 56/51.6 %, sub-retinal fluid 57/52.7 %, any retinal fluid 95/87.1 %	12 months	N/A
Fassnacht-Riederle [51]	88/96	78.9	43/53[a]	N/A	Only exudative AMD showing insufficient anatomical response to previous anti-VEGF drugs, defined as any persisting or increasing sub-retinal or intraretinal fluid observed in OCT	At least 3 intravitreal 0.5 mg ranibizumab or 1.25 mg bevacizumab over a period of no more than 4 months prior to switching to aflibercept	None	N/A	3 months	N/A
Hall [52]	30/30	80.4 ± 1.45	8/22	18/12	Exudative AMD previous treated with anti-VEGF drugs (both refractory and responders)	At least 2 previous anti-VEGF injections, either 1.25 mg bevacizumab and/or 0.5 mg ranibizumab; no maximum limit for BCVA; at least 6 months of follow-up after the first aflibercept injection.	Non-central edema	N/A	12 months	N/A

Table 2 continued

Reference	N° patients/ N° eyes	Age (years) (range or ± SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status (Phakic/ pseudophakic)
Bakall [48]	31/36	79 (range 60–88)	13/18	22/14	Exudative AMD with recurrent or resistant intraretinal and/or sub-retinal fluid to previous monthly injections of bevacizumab or ranibizumab	Age >60 years; initial response (decrease in retinal edema and sub-retinal fluid) followed by a recurrent or persistent sub-retinal fluid or retinal edema on OCT. The presence of intraretinal or sub-retinal fluid for a minimum of 3 months prior to the first aflibercept. This fluid had to be refractory to at least 3 continued monthly injections within 3 months prior the first aflibercept injection. At least 3 initial 2 mg aflibercept injections every 4–6 weeks followed by a visit within 4–8 weeks after the third aflibercept injection.	Measurements for the eyes with a change in treatment regimen or eyes that were lost to follow-up after the third injection, but before the 6-month visit, were not included in data collection for the 6-month visit.	N/A	6 months	N/A
Heussen [49]	65/71	77 (range 43–95)	24/41	N/A	Exudative AMD with recurrent or resistant intraretinal and/or sub-retinal fluid to previous anti-VEGF drug injections	N/A	PCV and RAP	N/A	4 months	N/A
Cho [50]	28/28	80.68 (range 62–95)	14/14	15/13	Exudative AMD with recalcitrant or persistent intraretinal and/or sub-retinal fluid to previous anti-VEGF drug injections (1 eye was a RAP)	Persistent intraretinal or sub-retinal fluid 28–35 days after a minimum of 6 anti-VEGF drug injections prior to switching to aflibercept; last injection of ranibizumab and/or bevacizumab within 28–35 days of switching to aflibercept; follow-up OCT and examination 28–35 days after switching to aflibercept	OCT and/or FA suggesting for outer retinal tubulation without intraretinal or sub-retinal fluid, PED without intraretinal or sub-retinal fluid, or cystic degeneration; dry OCT at any time during the 3 months before switching to aflibercept (allowing inclusion of previously responsive or tachyphylactic eyes); less than 6 months of follow-up after first aflibercept injection	N/A	6 months	N/A

Table 2 continued

Reference	N° patients/ N° eyes	Age (years) (range or ± SD)	Sex (M/F)[a]	Laterality (R/L)	Pathology	Inclusion criteria	Exclusion criteria	Characteristics of edema before aflibercept injections n°/ %	Follow-up	Lens status (Phakic/ pseudophakic)
Ho [37]	85/96	79 (range 62–91)	34/51[a]	N/A	Persistent, recurrent, or worsening exudative AMD on examination or OCT; 1 PCV	At least 2 aflibercept injections and 4 ± 1 months of follow-up; not mandatory active exudation at baseline	Follow-up intervals outside of the specified time frame; reversion to another anti-VEGF agent	Exudative fluid and/or hemorrhage 82/85 %. intraretinal fluid 31/32 %, sub-retinal fluid 49/51 %, PED 73/76 %, sub-macular hemorrhage 18/19 %	4 ± 1 months	N/A
Kumar [38]	33/34	79 ± 8 (range 72–84)	10/24[a]	N/A	Exudative AMD with persistent foveal intraretinal and/or sub-retinal fluid despite previous ranibizumab injections	At least 3 monthly intravitreal injections of ranibizumab in the last 3 months; interval between the each ranibizumab and between the last ranibizumab injection and the first aflibercept less than 42 days	Not sub-foveal fluid; other treatment other than ranibizumab injections in the last 3 months before the first aflibercept injection	N/A	6 months	N/A
Yonekawa [40]	94/102	79.6 (range 57–93)	36/58	49/53	Exudative AMD with refractory (68 eyes) or recurrent (34 eyes) exudation despite previous anti-VEGF drug injections	At least previous 3 anti-VEGF drug injections	Concomitant visual significant ocular pathology (such as vitreous hemorrhage), insufficient clinical records, fewer than 3 previous anti-VEGF drug injections, lack of follow-up after conversion to aflibercept	N/A	18.4 weeks	N/A
Patel [47]	3/3	56.3 (range 49–65)	0/3	1/2	Exudative AMD with persistent foveal intraretinal and/or sub-retinal fluid despite previous ranibizumab injections	N/A	N/A	N/A	3 months	N/A

[a] Fassnacht-Riederle et al., Ho et al. and Kumar et al. defined sex on the number of enrolled eyes and not on the number of the enrolled patients

SD standard deviation, *M/F* male/female, *R/L* right eye/left eye, *N/A* not available, *AMD* age-related macular degeneration, *Anti-VEGF* anti-vascular endothelial growth factor, *PDT* photodynamic therapy, *Sub-RPE* sub-retinal pigment epithelial, *OCT* optical coherence tomography, *BCVA* best-corrected visual acuity, *ETDRS* early treatment diabetic retinopathy study, *PCV* polypoidal choroidal vasculopathy, *IOP* intraocular pressure, *TA* triamcinolone acetonide, *PED* pigment epithelial detachment, *CNV* choroidal neovascularization, *FA* fluorescein angiography, *CSR* central serous retinopathy, *RAP* retinal angiomatous proliferation

Table 3 Prospective clinical trials: previous anti-VEGF drugs and aflibercept treatment

Reference	Previous anti-VEGF drug/s	Other previous treatments	Protocol treatment of previous anti-VEGF drug/s	No. of injections of previous anti-VEGF drug/s	No. of eyes treated with only 1.25 mg/ml bevacizumab/mean no. of injections	No. of eyes treated with only 0.05 mg/ml ranibizumab/mean no. of injections
Kawashima [41]	Ranibizumab 0.5 mg	None	Loading phase of 3 monthly ranibizumab injections followed by PRN protocol	8.1 ± 3.1	0	15/N/A
Wycoff [60]	Ranibizumab 0.5 mg; ranibizumab 2.0 mg	None	For the inclusion in the SAVE study, at least 9 injections of 0.5 mg of ranibizumab or 1.25 mg of bevacizumab within 12 months before the enrollment. For the SAVE study, a loading phase of 3 monthly ranibizumab 2.0 mg followed by PRN reinjections for 24 months	42 (range 19–67)	0	46/42 [21 ranibizumab 0.5 mg; 21 ranibizumab 2.0 mg (range 13–24)]
Chang [61]	Ranibizumab 0.5 mg	PDT [1 patient at least 4 ranibizumab in the last 6 months plus bevacizumab and PDT more than 6 months before switching]	Patients followed for at least 24 months from the first injection and underwent to at least 4 ranibizumab injections in the last 6 months before the first aflibercept	34.9 ± 16.1	0	41/N/A
Singh [62]	Ranibizumab 0.5 mg bevacizumab 1.25 mg	None	Monthly PRN treatment	9.62 ± 6.58 (range 3–23)	7/N/A	17/N/A
Grewal [39]	ranibizumab 0.5 mg bevacizumab 1.25 mg	PDT [only one patient]	Monthly till signs of exudation (treat until dry)	29.8 ± 17.1 (range 6–70, 11.33 per year)	5/27.4 (range 6-43)	4/20.4 (range 6–37)

Reference	No. of eyes treated with both anti-VEGF drugs/mean no. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Kawashima [41]	0	N/A	N/A	Loading phase of 3 monthly aflibercept injections followed by bimonthly injections	N/A	Monthly and bimonthly
Wycoff [60]	0	N/A	33.3 days (median 28 days; range 28–68 days)	Loading phase of 3 monthly injections (baseline, month 1 and month 2), one mandatory dose at month 4 and PRN doses at months 3 and 5 (presence of intraretinal and/or sub-retinal fluid on OCT or decrease in BCVA > 5 ETDRS letters)	5.6	Monthly
Chang [61]	8/N/A [7 at least 4 ranibizumab plus at least 1 bevacizumab in the last 6 months; 1 at least 4 ranibizumab plus 1 bevacizumab more than 6 months before switching]	Last 6 months: 5.0 ± 0.7	At least 30 days	Loading phase of 3 monthly injections (baseline, month 1 and month 2) plus mandatory injection at weeks 16 and 24	5.0	Monthly for the loading phase, bimonthly for the other two injections

Table 3 continued

Reference	No. of eyes treated with both anti-VEGF drugs/mean no. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Singh [62]	2/N/A	N/A	N/A	Loading phase of 3 monthly injections (baseline, month 1 and month 2) plus mandatory bimonthly injections	5	From monthly to bimonthly
Grewal [39]	12/33.8 (range 7–70)	N/A	N/A	Loading phase of 3 monthly injections (baseline, month 1 and month 2) plus bimonthly injections in case of resolution of the edema or monthly in case of edema	10.2 ± 1.2	From monthly to bimonthly

Anti-VEGF anti-vascular endothelial growth factor, *PRN* pro re nata, *N/A* not available, *OCT* optical coherence tomography, *BCVA* best-corrected visual acuity, *ETDRS* early treatment diabetic retinopathy study, *PDT* photodynamic therapy

recurrent exudative AMD, differed from each paper as reported in Table 1.

Previous anti-VEGF drugs and aflibercept treatment (Table 3)

Chang et al. [61] and Kawashima et al. [41] enrolled only patients previously treated with ranibizumab 0.5 mg/ml monotherapy, and Wycoff et al. [60] included patients previously treated with ranibizumab 2.0 mg from the SAVE Study, whereas Singh et al. [62] and Grewal et al. [39] enrolled patients previously treated with ranibizumab 0.5 mg/ml and/or bevacizumab 1.25 mg/ml.

The mean number of previous anti-VEGF injections was 29.55 for patients, even if great differences about the number of injections were reported not only among different works, but also within the same study (i.e., Grewal reported a range from 6 to 74 [39]). Interestingly, 22 eyes have been previously underwent to at least one switch from ranibizumab to bevacizumab or vice versa, and 46 eyes were previously switched from ranibizumab 0.5 mg/ml to ranibizumab 2.0 mg/ml [60] while all the other patients received only monotherapy with ranibizumab 0.5 mg/ml (77 eyes) or bevacizumab (12 eyes). The mean number of anti-VEGF drug injections in the last 6 months prior of the switching to aflibercept has been reported only in the work of Chang et al. (mean of 5.0 ± 0.7 injections) [61]. The mean time interval between the last anti-VEGF injection and the first aflibercept injection was reported only by Wycoff et al. [60] and Chang et al. [61] (mean of 33 days in the first case and at least 30 days in the second one).

The mean number of aflibercept injections was reported in all papers except for Kawashima et al. (mean number 5.96, range 5.0–10.2) [39, 60–62]. Aflibercept injection protocols provided in all cases: a loading phase of 3 monthly injections [39, 41, 60–62], followed by bimonthly injections [41, 62], one mandatory dose at month 4 and PRN doses at months 3 and 5 [60], mandatory injection at weeks 16 and 24 [61], bimonthly injections in case of resolution of the edema or monthly in case of edema [39].

Anatomical outcomes (Table 5)

Mean baseline central macular thickness (CMT) was 335.34 μm, even if great differences were reported not only among different works (range of mean values 202.1 μm [41] to 448.4 μm [61]), but also within the same study (range 193–637 μm) [39]. After the first aflibercept injection, CMT has been reported only in one work [60] with a significant reduction ($P < 0.05$). Data were available for three works after a 3-month follow-up [39, 60, 61]: Mean reduction in CMT was statistically significant in all works ($P < 0.05$) [60, 61] except for Grewal and co-

Table 4 Retrospective clinical trials: previous anti-VEGF drugs and aflibercept treatment

Reference	Previous anti-VEGF drug/s	Other previous treatments	Protocol treatment of previous anti-VEGF drug/s	No. of injections of previous anti-VEGF drug/s	No. of eyes treated with only 1.25 mg/ml bevacizumab/mean no. of injections	No. of eyes treated with only 0.05 mg/ml ranibizumab/mean no. of injections
Michalewsky [59]	Bevacizumab 1.25 mg	N/A	N/A	N/A	23/10 (6–20)	N/A
Eadie [58]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	% pts with PDT several years prior to the transition to aflibercept: 1 pt with triamcinolone	Treat and extend protocol or every 4 weeks	(3–38)	N/A	N/A
Thorell [57]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg, pegaptanib sodium	None	Treat and extend strategy, starting from every 4-week treatment	30.7 ± 15.5 during 44.9 ± 23.3 months of follow-up	15/N/A	47/N/A
Griffin [56]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg,	N/A	N/A	11.3 ± 5.96 (3–27)	15/10 ± 5.29 (3–22)	14/11.14 ± 7.14 (3–26)
Arcinue [55]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	Monthly anti-VEGF monotherapy until the eye become completely dry, after which 1–2 bonus injections before going to an observation period	Median 13 (range 7–22)	N/A/median 13 (range 7–21)	N/A/median 15 (range 3–8)
Gharbiya [54]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	Loading dose of 3 monthly anti-VEGFs (bevacizumab or ranibizumab) injections followed by PRN protocol	34.4 ± 11.9 (15–50)	0/0	20/N/A
Messenger [53]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	At least 12 months of prior anti-VEGF therapy with ranibizumab or bevacizumab	21.4 (range 4–60)	51/6.96 in the last 12 months	40/7.28 in the last 12 months
Fassnacht-Riederle [51]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	At least 3 anti-VEGF drug intravitreal injections (ranibizumab 0.5 mg or bevacizumab 1.25 mg) in the last 4 months before switching	26.9 (24.8 for ranibizumab and 9.2 for bevacizumab)	4/N/A	64/N/A
Hall [52]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	At least 2 previous anti-VEGF drug injections	14.9 ± 2.01 (range 2–53)	18/12.4 ± 2.18	2/19 ± 6
Bakall [48]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	Loading phase of 3 monthly anti-VEGF drug injections (baseline, month 1 and month 2), followed by PRN retreatment	25.6 (range 6–74; 10.6 for ranibizumab and 15.0 for bevacizumab)	4/38.25	1/10
Heussen [49]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	Loading phase of 3 monthly anti-VEGF drug injections (baseline, month 1 and month 2), followed by other 3 monthly injections if not responders or, sometimes, only 1 injection	9 (range 3–43; 3.25 per year)	0	67/N/A
Cho [50]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	None	At least 6 anti-VEGF drug injections (no specific about protocol has been mentioned; only "regular treatment")	20.2 ± 7.6 (range 7–37)	N/A/N/A	N/A/N/A

Table 4 continued

Reference	Previous anti-VEGF drug/s	Other previous treatments	Protocol treatment of previous anti-VEGF drug/s	No. of injections of previous anti-VEGF drug/s	No. of eyes treated with only 1.25 mg/ml bevacizumab/mean no. of injections	No. of eyes treated with only 0.05 mg/ml ranibizumab/mean no. of injections
Ho [37]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	PDT (5 eyes) and pegaptanib (1 eye)	As a general rule, anti-VEGF drug injection every 4–5 weeks as long as presence of exudation	17 (range 1–60)	30/14 (range 1–53)	43/19 (range 1–19)
Kumar [38]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg	PDT (5 eyes; mean 0.4 ± 1.1; range 0–1)	At least 3 ranibizumab injections just before the switching with an interval not exceeding 42 days. No information about intravitreal protocol before the last 3 ranibizumab injections has been mentioned.	28.6 ± 20.1 (range 11–43) [bevacizumab 1.8 ± 2.8, range 0–3; ranibizumab 26.5 ± 18.4, range 10–44]	N/A/N/A	N/A/N/A
Yonekawa [40]	Ranibizumab 0.5 mg, bevacizumab 1.25 mg, bevacizumab 2.00 mg, and/or ranibizumab 0.75 mg or 1.00 mg (38 eyes)	PDT (6 eyes); thermal laser (1 eye); pegaptanib (2 eyes)	PRN and/or treat and extend regimen (mean time of treatment every 5.9 weeks)	20.4 (median 18; range 3–65)	26 (24 refractory and 2 recurrent)/N/A	48 (22 refractory and 26 recurrent)/N/A
Patel [47]	Ranibizumab 0.5 mg, ranibizumab 1.0 mg, ranibizumab 1.5 mg, bevacizumab 1.25 mg	None	PRN	9.3 (range 7–11)	0/0	0/0

Reference	No. of eyes treated with both anti-VEGF drugs/mean No. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Michalewsky [59]	N/A	N/A	N/A	N/A	1	N/A
Eadie [58]	N/A	N/A	5.88 weeks (4–10)	Treat and extend protocol or every 4 weeks	5.53 (2–11)	6.47 weeks (4–8)
Thorell [57]	11/N/A	5.1 ± 1.3 in the last 6 months; 9.8 ± 2.4 in the last 12 months	The same required by the strategy treatment with previous anti-VEGF	Treat and extend strategy, starting from every 4-week treatment	4.5 ± 1.0 (reduction in 0.6 ± 1.1 inj compared with previous anti-VEGF drugs; $P < 0.001$)	Treat and extend strategy, starting from every 4-week treatment
Griffin [56]	18/12.5 ± 5.57 (8–27)	N/A	42.9 ± 1.9 days (27–63)	Monthly injections	3	4 weeks

Table 4 continued

Reference	No. of eyes treated with both anti-VEGF drugs/mean No. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Arcinue [55]	N/A/Median 13 (range 7–22)	N/A	Median 6 weeks (range 4–18)	Every 8-week aflibercept injections, without the 3 initial monthly injections. In case of persistent fluid despite strict every 8-week injections (after a median of 4), regimen was shifted to every 4 weeks	The median number of every 8 weeks was 4 (range 4–6). Twenty-one eyes were switched to every 4-week aflibercept injections	Every 8 weeks and/or every 4 weeks
Gharbiya [54]	10/N/A	6 in the last 6 months	4.9 ± 0.8 weeks (4–6)	Loading dose of three monthly aflibercept injections and retreatment according to any of the following criteria: (1) visual acuity loss of at least five letters with OCT evidence of fluid in the macula; (2) persistent or recurrent intraretinal or sub-retinal fluid on OCT; (3) new sub-retinal hemorrhage from the CNV	4.5 ± 1.3 (3–6)	5.8 ± 1.7 (4–8)
Messenger [53]	18/N/A	Last 12 months: 7.4 (range 1–16). 29 patients ≥10 injections in the last 12 months	N/A	N/A	7.2 (range 1–12)	N/A
Fassnacht-Riederle [51]	28/N/A	N/A/N/A	N/A	Loading phase of 3 monthly injections (baseline, month 1 and month 2)	3.0	Monthly
Hall [52]	10/19.3	N/A/N/A	N/A	In the presence of sub-retinal fluid on OCT at the time of the switch: loading phase of 3 monthly injections (baseline, month 1 and month 2), then treat and extend regimen. In absence of sub-retinal fluid on OCT at the time of the switching: treat and extend regimen	6.27 ± 0.37 (range 4–11) [6-month follow-up: 30 patients received 4.50 ± 0.11 injections. 9-month follow-up: 26 patients received 6.00 ± 0.23. 12-month follow-up: 22 patients received 7.17 ± 0.38]	From monthly to bimonthly

Table 4 continued

Reference	No. of eyes treated with both anti-VEGF drugs/mean No. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Bakall [48]	31/24.48	Last 6 months: 5.2 (range 4–6)	4 weeks	At least 3 injections every 4–6 weeks, followed by PRN retreatment until month 6	5.72 [in 9 eyes treatment was stopped prior to 6 months: in 8 patients after the fifth injection and in 1 patient after the fourth injection]	Every 4–6 weeks for the loading phase and at the discretion of the retina specialist for the PRN phase
Heussen [49]	4/N/A	N/A/N/A	77 days (range 20–422 days, median 37)	PRN treatment without loading phase	2.73 (range 1–4) [5 eyes received 1 injection; 21 eyes received 2 injection; 33 eyes received 3 injections; 12 eyes received 4 injections]	4 weeks
Cho [50]	N/A/N/A	N/A/N/A	28–35 days	Initially monthly loading phase of 3 injections (baseline, month 1 and, month 2), then every 6–8 weeks for 21 eyes (75 %)	4.4 (range 3–6)	4–8 weeks
Ho [37]	23/13 bevacizumab (range 1–32) and 8 ranibizumab (range 1–36)	N/A/N/A	49 days (range 18–355)	Loading phase of 3 monthly injections (baseline, month 1, and month 2) plus fourth injection 1–2 months later	2.6 (range 2–4)	4 weeks
Kumar [38]	16/N/A	N/A/N/A	34.4 ± 5.0 days (range 32–37)	Loading phase of 3 monthly injections (baseline, month 1 and month 2) followed by PRN treatment	5.3 ± 0.6 (range 5–6)	35 days
Yonekawa [40]	28 (22 refractory and 6 recurrent)/ N/A	N/A/N/A	7.0 weeks (5.7 weeks for the refractory group and 9.5 weeks for the recurrent group)	PRN and/or treat and extend regimen	3.8 (median 3.5; range 1–8)	7.3 weeks (5.2–6.1 weeks for refractory group; 7.2–9.6 for recurrent group)

Table 4 continued

Reference	No. of eyes treated with both anti-VEGF drugs/mean No. of injections	Mean no. of injections in the last 6 or 12 months before the first aflibercept injection	Time interval between the last anti-VEGF drug and the first aflibercept injection	Protocol treatment with aflibercept injections	Mean no. of aflibercept injections	Interval between aflibercept injections
Patel [47]	*First patient*: 2 bevacizumab, 6 ranibizumab, 2 ranibizumab 1.0 mg. *Second patient*: 6 ranibizumab, 2 bevacizumab, 3 ranibizumab 1.5 mg. *Third patient*: 3 bevacizumab, 4 ranibizumab	N/A/N/A	N/A	Loading phase of 3 monthly injections (baseline, month 1 and month 2)	3.0	4 weeks

Anti-VEGF anti-vascular endothelial growth factor, *PRN* pro re nata, *N/A* not available, *OCT* optical coherence tomography, *BCVA* best-corrected visual acuity, *ETDRS* early treatment diabetic retinopathy study, *PDT* photodynamic therapy

workers ($P > 0.05$) [39]. At the 6-month follow-up, data were reported for all works [39, 41, 60–62]. Mean reduction in CMT was statistically significant in all works ($P < 0.05$) [39, 41, 60–62]. Total mean change was −52.47 μm (range from −25.24 μm [39] to −89.5 μm [61]). Only Grewal et al. [39] reported data from the 12-month follow-up with a significant reduction in the CMT [292.71 ± 91.35 μm ($P = 0.038$), range 193–627 μm].

The maximum height of PED was analyzed only in two studies [39, 41]. Total mean baseline value was 227.87 μm (range from 167 μm [41] to 288.73 μm [39]); mean total change at the 6-month follow-up was −60.52 μm (range from −72.9 μm [41] to −43.13 μm [39]) with statistical significance reported in all works ($P < 0.05$). Data of other parameters and follow-up visits are reported in Table 5.

Functional outcomes (Table 7)

In three studies, best-corrected visual acuity (BCVA) was measured with ETDRS charts at 4 meters [60–62]: mean baseline value was 64.83 ETDRS letters. After 6 months, mean change was +4.14 ETDRS letters. Only Wycoff et al. [60] did not report any significant change at 6-month follow-up after the switching to aflibercept ($P > 0.05$), whereas Chang et al. [61] and Singh et al. [62] reported a significant change at the last follow-up visit ($P < 0.001$). Sub-analyses showed great differences in individual response (range, from −10 letters to >+15 letters [60–62]).

In the other two articles, BCVA has been reported as Logarithm of Minimum Angle of Resolution (LogMAR) [39, 41]. These authors showed not significant change in BCVA at the 6-month follow-up ($P > 0.05$) [39, 41].

Adverse events (Table 7)

Reported ocular adverse events were rare and were reported only in two works [60, 61]. Wycoff et al. [60] reported cataract progression in 3 patients (7 %); progression of geographic atrophy in 3 patients (7 %); atrial fibrillation in one patient (2 %); finally one patient (2 %) died related to complications of acute onset leukemia; while Chang et al. [61] extensive sub-macular hemorrhage in 1 patient (2 %); worsening of a previous sub-macular hemorrhage after first injection in one patient (2 %); progression of cataract in one patient (2 %); acute myocardial infarction in one patient (2 %); deep vein thrombosis in one patient (2 %); atrial fibrillation in 2 patients (4 %); syncope in 1 patient (2 %); spondylolisthesis in 1 patient (2 %), gastritis in 1 patient (2 %); pneumonia in 1 patient (2 %).

Table 5 Prospective clinical trials: anatomical outcomes after aflibercept injections

Reference	Baseline		After the first injection	After the second injection	After the third injection		
	Mean baseline CMT (μm)	Mean baseline PED MH (μm)	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid/number of eyes with dry macula on OCT
Kawashima [41]	202.1 ± 113.7	167.0 ± 150.3	N/A	N/A	N/A	N/A	N/A
Wycoff [60]	347 (range 188–565)	N/A	323.4/−5 % (range −384 to +32) ($P < 0.05$)	N/A	309/−38 ± 12 ($P < 0.05$)	N/A	Decrease >10 % in 9 patients (19.6 %); increase >10 % in no patient. Complete resolution in 17 patients (32 %)
Chang [61]	448.4 ± 141.2	N/A	N/A	N/A	321.4/−127 ± 134 ($P < 0.001$)	N/A	22 eyes (45 %) were fluid-free
Singh [62]	304.08 ± 75.44 (range 210–505)	N/A	N/A	N/A	N/A	N/A	N/A
Grewal [39]	329.38 ± 102.67 (range 193–637)	PED MH (15 pts): 288.73 ± 175.91 (range 110–635)	N/A	N/A	332.14 ± 101.70 (range 202–658)	N/A	N/A

Reference	After the fourth injection	Six-month follow-up				Nine-month follow-up	Twelve-month follow-up
	Mean CMT (μm)/mean change (μm) compared to baseline	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid (n of eyes)	No. of eyes with complete resolution of intraretinal and/or sub-retinal fluid	Mean CMT (μm)	Mean CMT (μm)
Kawashima [41]	N/A	131.2 Reduction in 70.9 ± 77.4 ($P = 0.003$)	94.1 Reduction in 72.9 ± 83.0 ($P < 0.05$)	N/A	7 eyes (46.7 %)	N/A	N/A
Wycoff [60]	N/A	319.7/−6 % (range −381 to +59; $P = 0.018$)	N/A	Decrease >10 % in 7 patients (15.2 %); increase >10 % in 2 (4.3 %)	10 out of 46 patients (21.7 %)	N/A	N/A
Chang [61]	N/A	358.9 ($P < 0.001$)	N/A	Reduction > 100 μm in 33 % of eyes (16); reduction >150 μm in 20 % of eyes (10). Stabilization of fluid (±100 μm) in 63 % of patients (31 eyes). Increase >150 μm in 2 eyes (4 %)	14 out of 49 patients (29 %)	N/A	N/A

Table 5 continued

Reference	After the fourth injection	Six-month follow-up				Nine-month follow-up	Twelve-month follow-up
	Mean CMT (μm)/mean change (μm) compared to baseline	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid (n of eyes)	No. of eyes with complete resolution of intraretinal and/or sub-retinal fluid	Mean CMT (μm)	Mean CMT (μm)
Singh [62]	N/A	265.5/−38.6 ($P < 0.001$)	N/A	Anatomical improvement: sub-retinal fluid in 50 %; intraretinal fluid in 34.6 %, and PED in 19.2 % of eyes. Anatomical stabilization: sub-retinal fluid in 34.6 %, intraretinal fluid in 53.9 %, and PED in 76.9 % of eyes. Anatomical worsening: sub-retinal fluid in 15.4 %, intraretinal fluid in 11.5 %, and PED in 3.9 % of eyes	N/A	N/A	N/A
Grewal [39]	N/A	304.14 ± 83.48 (range 206–609) ($P < 0.05$)	240.60 ± 135.81 (range 68–510)	N/A	N/A	N/A	292.71 ± 91.35 ($P = 0.038$) (range 193–627). PED MH: 248.27 ± 146.22 (range 90–506) ($P = 0.002$)

CMT central macular thickness, *PED* pigment epithelial detachment, *MH* maximal height, *OCT* optical coherence tomography, *N/A* not available, *Sub-RPE* sub-retinal pigment epithelial

Table 6 Retrospective clinical trials: anatomical outcomes after aflibercept injections

Reference	Baseline		After the first injection	After the second injection	After the third injection		
	Mean baseline CMT (μm)	Mean baseline PED MH (μm)	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/ mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid/number of eyes with dry macula on OCT
Michalewsky [59]	521	N/A	446 ($P = 0.02$) Complete resolution of either intraretinal or sub-retinal fluid in 6 eyes	N/A	N/A	N/A	N/A
Eadie [58]	228.6	425.1 (from the ILM to Bruch's membrane at the center point)	N/A	N/A	N/A	N/A	N/A
Thorell [57]	257.6 ± 49.6	N/A	N/A	N/A	N/A	N/A	N/A
Griffin [56]	370.57 (281–429)	N/A	N/A	N/A	295.70 (232–335.5) ($P < 0.001$)	N/A	7 patients showed persistent IRF; 27 patients showed persistent SRF; 9 patients showed persistent multiple layer fluid
Arcinue [55]	355 (325–427)	N/A	N/A	N/A	N/A	N/A	N/A
Gharbiya [54]	449 ± 179	262 ± 134	N/A	N/A	224 ± 131 ($P < 0.001$)	181 ± 118 ($P < 0.001$)	Intraretinal fluid 8/26 %, sub-retinal fluid 8/26 %, any retinal fluid 23/74 %
Messenger [53]	325 ± 90.6 Intraretinal fluid: 52.7 % of eyes. Any retinal fluid: 87.1 % of eyes	N/A	N/A	N/A	N/A	N/A	N/A
Fassnacht-Riederle [51]	337	241	N/A	N/A	298/−39 ($P < 0.001$)	195/−46 ($P < 0.001$)	N/A
Hall [52]	261 ± 10.9	N/A	238 ± 12.4/−23 ($P = 0.021$)	N/A	245 ± 10.6/−16 ($P = 0.102$)	N/A	N/A
Bakall [48]	358 (range 181–579). Manual measure: 410 (range 174–1027)	153	N/A	N/A	309 [range 181–579; (−49 μm; $P < 0.001$)]. Manual measure: 346 [range 155–784; (−65 μm; $P < 0.001$)]	127/−26 ($P = 0.015$)	Decrease in fluid: 18/36 (50 %). Stabilization of fluid: 15/36 (41.7 %). Increase in fluid: 3/36 (8.3 %)

Table 6 continued

Reference	Baseline		After the first injection	After the second injection	After the third injection		
	Mean baseline CMT (μm)	Mean baseline PED MH (μm)	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/ mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid/number of eyes with dry macula on OCT
Heussen [49]	382.7 ± 100 (range 232–628)	N/A	347.6 ± 124 (range 198–772, $P = 0.021$)	283.9 ± 58 ($P = 0.034$)	260.8 ± 30 ($P = 0.006$)	N/A	N/A
Cho [50]	295	N/A	272/−23 ($P < 0.001$). Decrease in fluid: 19 eyes with recalcitrant fluid (86 %); 7 eyes with persistent fluid (86 %); 6 eyes with sub-RPE (50 %). Complete resolution of fluid: 5 eyes (18 %)	N/A	N/A	N/A	N/A
Ho [37]	276 (range 130–559)	N/A	N/A	N/A	N/A	N/A	N/A
Kumar [38]	416 ± 217 (range 263–487)	260 ± 162 (range 129–368)	N/A	N/A	351 ± 172 (range 224–445; $P < 0.001$)	228 ± 140 (range 114–340; $P = 0\ 001$)	N/A
Yonekawa [40]	305.07 ± 80.65. [Refractory 311.57 ± 77.83; recurrent 288.83 ± 86.86]	N/A	271.05 ± 68.89 ($P < 0.001$) [Refractory 283.33 ± 66.97 ($P < 0.001$); recurrent 250.83 ± 69.82, ($P < 0.001$)]	N/A	N/A	N/A	N/A
Patel [47]	N/A	N/A	N/A	N/A	N/A	Complete reabsorption of the PED in all the cases	N/A

Reference	After the fourth injection	Six-month follow-up				Nine-month follow-up	Twelve-month follow-up
	Mean CMT (μm)/mean change (μm) compared to baseline	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/ mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid (no. of eyes)	No. of eyes with complete resolution of intraretinal and/ or sub-retinal fluid	Mean CMT (μm)	Mean CMT (μm)
Michalewsky [59]	N/A	N/A	N/A	N/A	N/A	N/A	N/A

Table 6 continued

Reference	After the fourth injection	Six-month follow-up				Nine-month follow-up	Twelve-month follow-up
	Mean CMT (μm)/mean change (μm) compared to baseline	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/ mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid (no. of eyes)	No. of eyes with complete resolution of intraretinal and/ or sub-retinal fluid	Mean CMT (μm)	Mean CMT (μm)
Eadie [58]	N/A	176.9 ($P = 0.001$)	324.7 ($P = 0.012$) (from the ILM to Bruch's membrane at the center point)	24 eyes (34 %) complete resolution of exudation; 17 eyes (25 %) clear improvement without complete resolution; 23 eyes (34 %) no improvement; 4 eyes (6 %) worsening of exudation	N/A	N/A	N/A
Thorell [57]	N/A	239.0 ± 49.9 ($P < 0.001$)	N/A	29 eyes with predominantly serous PED and 41 eyes with predominantly fibrovascular PED	N/A	N/A	N/A
Griffin [56]	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Arcinue [55]	N/A	269 (range 227–325) ($P < 0.001$)	N/A	N/A	38/60.3 %	N/A	248 (range 201–279) ($P < 0.001$) [38 completely dry/60.3 %]
Gharbiya [54]	N/A	269 ± 145 ($P < 0.001$)	183 ± 100 ($P < 0.001$)	Intraretinal fluid 12/39 %, sub-retinal fluid 8/26 %	23/74 %	N/A	N/A
Messenger [53]	N/A	295 ± 77.1/−29 ($P = 0.0001$)	N/A	Intraretinal fluid: 37.3 % of eyes ($P = 0.0014$). Any retinal fluid: 57.8 % of eyes ($P < 0.0001$).	N/A	N/A	299 ± 83.0/−25 ($P = 0.0047$)
Fassnacht-Riederle [51]	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Hall [52]	N/A	245 ± 10.4/−16 ($P = 0.099$)	N/A	N/A	N/A	242 ± 9.66/−20 ($P = 0.015$)	237 ± 10.2/−27 ($P = 0.012$)
Bakall [48]	N/A	298 [range 168–561; (−49 μm60; $P < 0.001$)]. Manual measure: 296 [range 151–528; (−87 μm; $P < 0.001$)]	101/−13 ($P = 0.14$)	Decrease in fluid: 14/27 (51.9 %). Stabilization of fluid: 10/27 (37 %). Increase in fluid: 3/27 (11.1 %)	6 out of 36 enrolled eyes (20 %)	N/A	N/A
Heussen [49]	272.0 ± 49 ($P = 0.012$)	N/A	N/A	N/A	N/A	N/A	N/A

Table 6 continued

Reference	After the fourth injection	Six-month follow-up				Nine-month follow-up	Twelve-month follow-up
	Mean CMT (μm)/mean change (μm) compared to baseline	Mean CMT (μm)/mean change (μm) compared to baseline	Mean PED MH (μm)/ mean change (μm) compared to baseline	Modification of the intraretinal and/or sub-retinal fluid (no. of eyes)	No. of eyes with complete resolution of intraretinal and/ or sub-retinal fluid	Mean CMT (μm)	Mean CMT (μm)
Cho [50]	N/A	274/−21 ($P = 0.008$)	Decrease in maximum height of PED in 7 eyes (47 %); stabilization in 8 eyes (53 %); increase in no patient	Eyes with recalcitrant fluid: decrease in fluid in 22 eyes (64 %); stabilization of fluid in 5 eyes (23 %); and increase in fluid in 3 eyes (14 %). Eyes with persistent fluid: decrease in fluid in 5 eyes (63 %); stabilization of fluid in 2 eyes (25 %); and increase in fluid in 1 eye (13 %)	7 out of 28 eyes (25 %)	N/A	N/A
Ho [37]	258/−18 (range −242 to 198; $P = 0.06$)	N/A	N/A	At month 4: decrease in fluid in 40 eyes (49 %); stabilization of fluid in 26 eyes (32 %); increase in fluid in 12 eyes (14 %)	4 out of 82 eyes (5 %)	N/A	N/A
Kumar [38]	N/A	348 ± 171 (range 235–419; $P < 0.001$)	215 ± 142 (range 111–305; $P < 0$ 001)	N/A	N/A	N/A	N/A
Yonekawa [40]	N/A	276.20 ± 69.82 ($P < 0.001$) [Refractory 283.01 ± 68.73 ($P < 0.001$); recurrent 260.97 ± 70.00, ($P < 0.001$)]	N/A	N/A	N/A	N/A	N/A
Patel [47]	N/A	N/A	N/A	N/A	N/A	N/A	N/A

CMT central macular thickness, *PED* pigment epithelial detachment, *MH* maximal height, *OCT* optical coherence tomography, *N/A* not available, *Sub-RPE* sub-retinal pigment epithelial

Table 7 Prospective clinical trials: functional outcomes and adverse events after aflibercept injections

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up
Kawashima [41]	N/A	0.41 ± 0.37 logMAR	N/A	N/A	N/A	N/A
Wycoff [60]	N/A	74.2 ETDRS letters (range 41–91) [Snellen equivalent 20/32; range 20/100–20/16]	N/A	N/A	N/A	N/A
Chang [61]	N/A	60.5 ± 16.2 ETDRS letters (range 41–91)	N/A	N/A	N/A	N/A
Singh [62]	N/A	56.42 ± 17.04 ETDRS letters (range 24–80)	N/A	N/A	N/A	N/A
Grewal [39]	N/A	0.42 ± 0.28 logMAR (20/52 Snellen; range 0.10–0.90 logMAR)	N/A	N/A	N/A	N/A

Reference	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Kawashima [41]	N/A	N/A	0.01 ± 0.09 ($P = 0.699$)	N/A	N/A
Wycoff [60]	N/A	74.4 ETDRS letters (range 41–91)	+0.2 ETDRS letters (range -10 + 13; $P = 0.71$). [improvement ≥5 ETDRS letters in 4 eyes (9 %); improvement ≥10 ETDRS letters in 1 eye (2 %); decrease ≥ 5 ETDRS letters in 4 eyes (9 %); and decrease ≥10 ETDRS letters in 1 eye (2 %)]	N/A	Cataract progression in 3 patients (7 %); progression of geographic atrophy in 3 patients (7 %); no systemic arterial thromboembolic events; atrial fibrillation in one patient (2 %); one patient (2 %) died related to complications of acute onset leukemia.
Chang [61]	N/A	67.4 ETDRS letters	+6.9 ± 8.12 ($P < 0.001$) [increase ≥5 ETDRS letters in 27 eyes (55 %); increase ≥10 ETDRS letters in 13 eyes (26 %); increase ≥15 ETDRS letters in 5 eyes (10 %); stabilization (± 5 ETDRS letters in 22 patients (43 %); decrease ≥5 ETDRS letters in 1 eye (2 %)	N/A	Extensive sub-macular hemorrhage in 1 patient (2 %); worsening of a previous sub-macular hemorrhage after first injection in one patient (2 %); progression of cataract in one patient (2 %); Irvine-Gass syndrome in non-study eye after cataract extraction and IOL implantation in one patient (2 %); acute myocardial infarction in one patient (2 %); deep vein thrombosis in one patient (2 %); atrial fibrillation in 2 patients (4 %); syncope in 1 patient (2 %); spondylolisthesis in 1 patient (2 %), gastritis in 1 patient (2 %); pneumonia in 1 patient (2 %)
Singh [62]	N/A	62.3 ETDRS letters	Change: +5.9 ETDRS letters ($P < 0.001$). No patients lost > 15 ETDRS letters. 4 patients (15.39 %) lost between 1 and 10 ETDRS letters. 11 patients (42.31 %) gained between 0 and 5 ETDRS letters. 7 patients (26.92 %) gained between 10 and 15 ETDRS letters. 4 patients (15.39 %) gained ≥ 15 ETDRS letters	N/A	No serious side events

Table 7 continued

Reference	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Grewal [39]	N/A	0.42 ± 0.28 (0–0.83)	N/A	0.40 ± 0.28 logMAR (20/50 snellen; range 0–1, $P = 0.5$)	N/A

BCVA best-corrected visual acuity, *Anti-VEGF* anti-vascular endothelial growth factor, *N/A* not available, *ETDRS* early treatment diabetic retinopathy study, *P* *P* value, *IOL* intraocular lens, *PED* pigment epithelial detachment, *RPE* retinal pigment epithelium, *IOP* intraocular pressure, *logMAR* logarithm of minimum angle of resolution, *SRH* sub-retinal hemorrhage, *SD* standard deviation

Retrospective reports

Demographics (Table 2)

A review of the literature revealed a total 16 retrospective reports for a total of 909 eyes affected by exudative AMD previously treated with anti-VEGF intravitreal injections and switched to aflibercept intravitreal treatment (mean age 78.66 years; 331 males and 530 females) [37, 38, 40, 47–59]. Laterality was reported only in six papers (136 right eyes and 126 left eyes) [40, 47, 48, 50, 52, 55], while in the other works it was not reported [37, 38, 49, 51, 53, 54, 56–59]. Only Arcinue et al. [55] reported the lens status (19 phakic eyes and 44 pseudophakic eyes). Follow-up duration ranged from a minimum of 1 month [59] to a maximum of 12 months [52, 53, 55] after the first aflibercept injection. Inclusion and exclusion criteria, as well as the definition of refractory or recurrent exudative AMD, differed from each papers, as reported in Table 2.

Previous anti-VEGF drugs and aflibercept treatment (Table 4)

All the authors of the included papers enrolled patients previously treated with ranibizumab 0.5 mg/ml and/or bevacizumab 1.25 mg/ml [37, 38, 48–58], except for Michalewsky et al. (only bevacizumab 1.25 mg/ml) [59], Yonekawa et al. [40] (ranibizumab 0.5 mg/ml, ranibizumab 0.75 mg/ml, ranibizumab 1.00 mg/ml, bevacizumab 1.25 mg/ml, bevacizumab 2.00 mg/ml) and Patel et al. [47] (ranibizumab 0.5 mg/ml, ranibizumab 1.00 mg/ml, ranibizumab 1.50 mg/ml, bevacizumab 1.25 mg/ml).

The mean number of previous anti-VEGF injections was reported in all papers [37, 38, 40, 47–54, 56, 57] except in 3 works [55, 58, 59]: a mean of 21.22 anti-VEGF injections were administered before the switch to aflibercept treatment. Also in this group, great differences about the number of injections were reported not only among different works, but also within the same study (i.e., Bakall et al. [48] reported a range of 6–74). 161 on 694 eyes (23.2 %) have been previously underwent to at least one switch from ranibizumab to bevacizumab or vice versa, while all the other patients received only monotherapy with ranibizumab 0.5 mg/ml (174 eyes) or bevacizumab (359 eyes). Data were not available from 4 works [38, 50, 55, 58]. The mean number of anti-VEGF drug injections in the last 6 months prior of the switching to aflibercept has been reported only in 3 works (mean of 5.33 injections) [48, 54, 57] and in the last 12 months only in 2 works (mean of 8.36 injections) [53, 57]. The switch from anti-VEGF injections and aflibercept injections was greatly variable among different works, but also within the same report there was not uniformity (i.e., range 20–422 days [49]).

Table 8 Retrospective clinical trials: functional outcomes and adverse events after aflibercept injections

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Michalewski [59]	N/A	0.53 logMAR (0.39 Snellen lines)	0.42 logMAR (0.49 Snellen lines)($P = 0.03$ for logMAR; $P = 0.01$ for Snellen lines). Improvement >5 ETDRS letters in 7 eyes; Stable in 18 eyes	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Eadie [58]	N/A	0.494 logMAR, Snellen equivalent 20/62	N/A	N/A	N/A	N/A	N/A	0.505 logMAR, Snellen equivalent 20/64 ($P = 0.84$)	N/A	N/A	N/A
Thorell [57]	68.0 ± 12.2 ETDRS letters 6 months before the first aflibercept injection	69.0 ± 10.9 ETDRS letters ($P < 0.05$)	N/A	N/A	N/A	N/A	N/A	69.5 ± 11.3 ETDRS letters ($P < 0.05$)	N/A	N/A	N/A
Griffin [56]	N/A	0.56 logMAR (0.29–0.99)	N/A	N/A	0.53 logMAR ($P = 0.301$) (0.24–0.71)	N/A	N/A	N/A	N/A	N/A	N/A
Arcinue [55]	N/A	0.40 logMAR (range 0.30–0.70)	N/A	N/A	N/A	N/A	N/A	0.40 logMAR (range 0.20–0.70); mean variation −0.05 logMAR (+2.5 ETDRS letters) ($P = 0.2559$)	Increase >5 ETDRS letters in 24 eyes (36.5 %); stabilization in 24 eyes (38.1 %); decrease >5 ETDRS letters in 15 eyes (23.8 %)	0.40 logMAR (range 0.20–0.90); mean variation 0.04 logMAR (−2 ETDRS letters) ($P = 0.1081$). Increase >5 ETDRS letters in 16 eyes (25.4 %); stabilization in 17 eyes (27 %); decrease >5 ETDRS letters in 28 eyes (44.4 %)	No significant ocular adverse events

Table 8 continued

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Gharbiya [54]	N/A	42.5 ± 12.5 ETDRS letters (0.29 logMAR)	N/A	N/A	42.3 ± 10.5 ETDRS letters (0.26 logMAR) ($P > 0.05$)	N/A	N/A	42.8 ± 10.0 ETDRS letters (0.24 logMAR) ($P > 0.05$)	Increase > 5 ETDRS letters in 8 eyes (26 %); stabilization in 19 eyes (61 %); decrease > 4 ETDRS letters in 4 eyes (13 %)	N/A	None
Messenger [53]	One year prior to the conversion to aflibercept: 0.51 logMAR	0.50 logMAR	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0.51 ($P = 0.89$). 14 eyes (12.8 %) gained ≥10 ETDRS letters. 15 eyes (13.8 %) lost ≥10 ETDRS letters	N/A
Fassnacht-Riederle [51]	65.1 ETDRS letters	61.6 ETDRS letters	N/A	N/A	63.5 ETDRS letters	+1.9 ETDRS letters ($P = 0.061$) [decrease >15 ETDRS letters in 4 patients]	N/A	N/A	N/A	N/A	No severe ocular or systemic adverse events. Despite a high percentage of eyes with PEDs, no new tear of the RPE was seen. None of the eyes showed a non-spontaneously resolving intraocular inflammation. IOP showed a mild but significant decrease from 15.1 mmHg to 13.7 mmHg ($P < 0.001$)

Table 8 continued

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Hall [52]	N/A	0.506 ± 0.054 logMAR [20/64]	0.504 ± 0.055 logMAR [20/64] ($P = 0.903$)	N/A	0.458 ± 0.061 logMAR [20/57] ($P = 0.112$)	N/A	N/A	0.413 ± 0.071 logMAR [20/52] ($P = 0.036$)	Increase >15 ETDRS letters in 4 eyes (13 %); stabilization in 25 eyes (84 %); decrease >15 ETDRS letters in 1 eye (3 %)	0.521 ± 0.076 logMAR [20/66] ($P = 0.836$) [Increase >15 ETDRS letters in 2 eyes (9 %); stabilization in 16 eyes (73 %); decrease >15 ETDRS letters in 4 eyes (18 %)]	No significant sight-threatening complications. Two patients developed increasing in IOP > 25 mmHg and IOP-lowering agents were administered and after adequate pressure control by a glaucoma specialist, anti-VEGF treatment resumed. No other drug- or injection-related adverse events except for mild sub-conjunctival hemorrhage
Bakall [48]	0.36 logMAR	0.45 logMAR ($P = 0.07$)	N/A	N/A	0.52 logMAR ($P = 0.052$)	[Increase >5 ETDRS letters in 8 eyes (22 %); stabilization in 14 eyes (39 %); decrease >5 ETDRS letters in 14 eyes (39 %)]	N/A	0.50 logMAR ($P = 0.36$)	[Increase >15 ETDRS letters in 2 eyes (7.4 %); increase ≥5 and <15 ETDRS letters in 6 eyes (22.2 %); stabilization in 9 eyes (33.4 %); decrease >5 ETDRS letters in 10 eyes (37 %)]	N/A	Endophthalmitis in 1 eye with coagulase-negative Streptococcus (the infected eye underwent to vitrectomy and antibiotic injections, with the vision subsequently returning to 20/40, same as prior to the onset of aflibercept injections
Heussen [49]	0.54 logMAR	0.67 ± 0.46 logMAR	0.65 ± 0.48 logMAR ($P = 0.426$)	0.60 ± 0.43 logMAR ($P = 0.677$)	0.43 ± 0.20 logMAR ($P = 0.074$)	N/A	0.25 ± 0.47 logMAR ($P = 0.079$)	N/A	N/A	N/A	No local or systemic side events

Table 8 continued

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Cho [50]	N/A	0.52 logMAR (20/67 Snellen)	0.52–0.54 logMAR (20/67–20/69 Snellen; $P = 0.64$)	N/A	N/A	N/A	N/A	0.57 logMAR (20/76 Snellen; $P = 0.49$)	N/A	N/A	No eye developed significant ocular safety events such as endophthalmitis, vitreous hemorrhage, retinal tear, retinal detachment or sustained elevations in IOP
Ho [37]	N/A	20/50 logMAR [20/40 in the only bevacizumab group; 20/50 in the only ranibizumab group and 20/60 in bevacizumab plus ranibizumab group]	N/A	N/A	N/A	N/A	N/A	N/A	Mean gain 0.02 logMAR (range -0.46–0.70; $P = 0.14$). Only bevacizumab group: change of −0.03 logMAR (range −0.38–0.12, $P = 0.27$). Only ranibizumab group: Change of −0.004 (range −0.46–0.47, $P = 0.8$). Bevacizumab plus ranibizumab group: change of +0.13 (range −0.06–0.20, $P = 0.003$)	N/A	No ocular adverse events, including endophthalmitis, retinal detachment, RPE tears, massive sub-macular hemorrhage, uveitis, or sustained IOP. Four eyes showed focal areas of SRH during the study, of which 1 worsened, 1 remained unchanged and 2 resolved by the end of the study period. No systemic complications
Kumar [38]	N/A	0.57 ± 0.36 logMAR (range 0.30–1.0) [20/75 Snellen]	N/A	N/A	0.52 ± 0.34 logMAR (range 0.30–0.70) [20/66 Snellen] ($P = 0.24$)	N/A	N/A	0.47 ± 0.32 logMAR (range 0.30–0.60) [20/60 Snellen] ($P = 0.004$)	N/A	N/A	No significant ocular safety events such as endophthalmitis, acute sterile inflammation, retinal tears, retinal detachment, vitreous hemorrhage or sustained elevation in IOP

Table 8 continued

Reference	BCVA before any anti-VEGF drugs	BCVA immediately before the first aflibercept injection	BCVA after the first aflibercept injection	BCVA after the second aflibercept injection	BCVA after the third aflibercept injection or at 3-month follow-up	Modification of BCVA after the third aflibercept or at 3-month follow-up	BCVA after the fourth aflibercept injection or at 4-month follow-up	BCVA after the sixth aflibercept injection or at 6-month follow-up	Modification of BCVA after the sixth aflibercept or at 6-month follow-up	BCVA at 12-month follow-up	Adverse events
Yonekawa [40]	N/A	0.42 logMAR (SD 0.30) [Refractory: 0.44 logMAR (SD 0.33); Recurrent: 0.38 logMAR (SD 0.33)]	0.44 logMAR (SD 0.36), $P = 0.723$; [Refractory: 0.46 logMAR (SD 0.41) $P = 0.897$; Recurrent: 0.39 logMAR (SD 0.25), $P = 0.778$]	N/A	N/A	N/A	N/A	0.38 logMAR (SD 0.27) $P = 0.253$; [Refractory: 0.38 logMAR (SD 0.28) $P = 0.215$; Recurrent: 0.38 logMAR (SD 0.25), $P = 0.811$]	N/A	N/A	No case of endophthalmitis; no systemic complications or death
Patel [47]	*First patient*: N/A *Second patient*: 20/25 *Third patient*: N/A	*First patient*: 20/70. *Second patient*: 20/30. *Third patient*: 20/40	N/A	*Third patient*: 20/25	*First patient*: 20/25. *Second patient*: 20/20	N/A	N/A	N/A	N/A	N/A	Not reported by the authors

BCVA best-corrected visual acuity, *Anti-VEGF* anti-vascular endothelial growth factor, *N/A* not available, *ETDRS* early treatment diabetic retinopathy study, *P P* value, *IOL* intraocular lens, *PED* pigment epithelial detachment, *RPE* retinal pigment epithelium, *IOP* intraocular pressure, *logMAR* logarithm of minimum angle of resolution, *SRH* sub-retinal hemorrhage, *SD* standard deviation

The mean number of aflibercept injections was reported in all papers except for Arcinue et al. (mean number 4.27, range of mean values was 1.0–6.27). Aflibercept injections protocols was greatly different among the included papers, as reported in Table 4.

Anatomical outcomes (Table 6)

Mean baseline Central Macular Thickness (CMT) was 326.15 μm, even if great differences were reported not only among different works (range of mean values 228.6 μm [58] to 521 μm [59]), but also within the same study (i.e., Bakall et al. [48] reported a range of baseline CMT from 181 to 579 μm). After the first aflibercept injection, CMT has been reported only in five works [40, 49, 50, 52, 59] with a significant reduction in all cases ($P < 0.05$). Total mean reduction in these five works was 35.51 μm (range, from −23 μm [50, 52] to −75 μm [59]). Data were available for seven works after a 3-months follow-up [38, 48, 49, 51, 52, 54, 56]. Mean reduction in CMT was statistically significant in all works ($P < 0.05$) except for Hall and co-workers ($P = 0.102$) [52]. Total mean change was −79.27 μm (range, from −16 μm [52] to −121.9 μm [49]). At the 6-month follow-up, data were reported for ten works [38, 40, 48, 50, 52–55, 57, 58]. Mean reduction in CMT was statistically significant in all works ($P < 0.05$) except for Hall and colleagues ($P = 0.099$) [52]. Total mean change was −48.13 μm (range, from −16 μm [52] to −180 μm [54]). Finally, three works reported the 12-months follow-up data [52, 53, 55]. Mean reduction in CMT was statistically significant in all the reports ($P < 0.05$), with a mean reduction in −93.11 μm.

The maximum height of PED was analyzed only in five studies [38, 48, 51, 54, 58].Total mean baseline value was 280.64 μm (range, from 153 μm [48] to 425.1 μm [58]); mean total change at the 3-month follow-up was reported in four works [38, 48, 51, 54] and mean reduction was −45.43 μm (range, from −26 μm [48] to −101 μm [54]) with statistical significance reported in all works ($P < 0.05$). At the 6-month follow-up data were available for four works [38, 48, 54, 58] that showed a mean change of −80.17 μm from baseline and statistical power in all the works. Data of other parameters and follow-up visits are reported in Table 6.

Functional outcomes (Table 8)

In three studies, best-corrected visual acuity (BCVA) was measured with ETDRS charts at 4 meters [51, 54, 57]: All the authors did not report any significant changes at 3 [51, 54] and 6 months [54, 57] after the switching to aflibercept ($P > 0.05$).

In the most part of the enrolled articles, BCVA has been reported as logarithm of minimum angle of resolution (LogMAR) [38, 40, 48–50, 52, 53, 55–59]. Michalewsky and co-authors reported a significant increase in BCVA 1 month after the first aflibercept injection ($P = 0.03$) [59]; Hall and co-workers and Kumar and colleagues found a significant improvement in BCVA at the 6-month follow-up ($P < 0.05$) [38, 52], whereas in the other works there was not a significant change at the last follow-up visit [40, 48–50, 53, 55, 56, 58]. Also in these works, a great individual variability has been found, with gaining of >15 letters or loosing >15 letters. Finally, Ho et al. [37] and Patel et al. [47] measured BCVA in 20/*x*, without any significant change after the treatment with aflibercept ($P > 0.05$).

Adverse events (Table 8)

Reported ocular adverse events were rare and included one case of endophthalmitis (subsequently resolved by vitrectomy with restoration of visual acuity) [48], onset of focal areas of sub-macular hemorrhage in 4 eyes [37], and increasing of intraocular pressure (IOP) >25 mmHg in 2 patients [52].

No systemic adverse events were observed during the study period in all the enrolled papers [37, 38, 40, 47–59].

Discussion

Eyes with recalcitrant exudative AMD represent a substantial clinical burden. It is unknown why some eyes with neovascular AMD dry up anatomically with fewer intravitreal injections of anti-VEGF drugs, whereas up to half have OCT findings of disease activity even with continuous monthly therapy [5]. However, funduscopy presentation of neovascular membrane represents only the final result of a complex cascade of events that can be modulated at different steps; therefore, they may require different treatments from patient to patient [63]. The early identification of these cases is essential in order to maintain the costs of these therapies sustainable for the national health systems.

The authors of the enrolled papers have addressed the problem switching to a new anti-VEGF now available for the treatment of neovascular AMD. Indeed, VEGF-A is the only target of bevacizumab and ranibizumab, whereas aflibercept binds all the isoforms of VEGF-A and VEGF-B, as well as PlGF [8]. Furthermore, aflibercept has a different molecular structure than ranibizumab and bevacizumab [64] with a significantly higher binding affinity for VEGF than either bevacizumab or ranibizumab (about 100 times greater) [11, 65]. Recent mathematical simulations predicted that a single intravitreal injection of aflibercept

2.0 mg would last between 48 and 83 days (compared with 30 days of ranibizumab 0.5 mg) and thus should be efficacious in neutralizing VEGF longer and more effectively [9, 11].

The aims of this review are as follows: (i) to report anatomical and functional outcomes of switching from bevacizumab/ranibizumab to aflibercept previously described in the scientific literature, (ii) to hypothesize the possible pathophysiological mechanisms of the resistance and tachyphylaxis to anti-VEGF drugs, and (iii) to suggest possible clinical actions increasing the chances of success for such difficult cases.

We reported the outcomes of 21 papers for a total of 1066 eyes affected by exudative AMD resistant to previous anti-VEGF drug injections and switched to aflibercept injections. Enrolled reports were divided into two groups: 5 prospective reports and 16 retrospective reports. Outcomes were not easy to decipher because of the different criteria used in these studies, although the detailed analysis of these data provided really interesting insights.

Of note, about a quarter of the eyes included in this analysis [68 out of 157 (43.31 %) and 161 out of 694 (23.19 %) eyes in the prospective and retrospective groups, respectively] were subjected to a previous switch from bevacizumab to ranibizumab and/or vice versa, potentially reducing the effects of switching to aflibercept. However, this may be not the case because, as previously described, bevacizumab and ranibizumab have the same target (i.e., VEGF-A) [64], whereas aflibercept binds all the isoforms of VEGF-A and VEGF-B, as well as PlGF, with a different molecular structure and significantly higher binding affinity for VEGF than either bevacizumab or ranibizumab (about 100 times greater) [11, 64, 65]. These dissimilarities might produce a better success rate than the simple switch bevacizumab/ranibizumab or vice versa. Finally, a single intravitreal injection of aflibercept is longer lasting than ranibizumab and thus should be efficacious in neutralizing VEGF longer and more effectively [9, 11].

Also, the "washout period" between the last anti-VEGF drug injection and the first aflibercept injection represents an important factor that could influence the final results of switching to aflibercept (Tables 3, 4). However, inconclusive data can be extrapolated from the included papers because not all the authors reported these data and great variability was present. In our hypothesis, if aflibercept has been administered too early, its effect could be limited by the previous anti-VEGF drug injection, competing for the same target. As reported by mathematical model of Stewart and co-workers, the pharmacological effect of ranibizumab 0.5 mg lasts about 30 days; hence, the switch to aflibercept should be performed from 4 to 5 weeks after the last anti-VEGF drug treatment [9].

Most of the studies reported in this review used a loading phase of three monthly aflibercept (Tables 3, 4), while subsequent injections were administered with different protocols and with different intervals of time (range from 4 to 8 weeks) (Tables 3, 4). Chang et al. [61] and Ho et al. [37] demonstrated that when the injections were administered every 8 weeks, the effects of aflibercept may not last for this duration as suggested by the fluctuation pattern of CRT. However, this pattern does not seem to impact significantly on vision [61]. The CRT was reversible with reinjection of aflibercept, even if it is currently not known whether fluctuating sub-retinal or intraretinal fluid, or both, is associated with long-term visual impairment [7].

Kumar et al. [38] described that in patients with persistent fluid, despite the previous treatments with other anti-VEGF drugs, the continuation of the therapy with aflibercept beyond three injections could be needed to achieve an improvement in visual acuity. Indeed, these eyes responded differently if compared to treatment-näive eyes evaluated in the clinical trials where a significant anatomical and visual improvement occurred after three injections [7]. Only Hall et al. and Grewal et al. reported data at 12-month follow-up, showing a significant anatomical response ($P = 0.012$ and $P = 0.002$, respectively) (Tables 5, 6), but not a functional one ($P = 0.836$ and $P = 0.5$, respectively) (Tables 7, 8) [39, 52]. In all the other papers, follow-up lasts maximum 6 months after the first aflibercept injection (Tables 3, 4).

The great variability in number of previous anti-VEGF drug injections could affect the results. Indeed, we hypothesize that few anti-VEGF drug injections could mean a sub-optimal treatment, whereas an excessive number of injections without obtaining a complete fluid reabsorption could represent an inadequate treatment for a long period, leading to the maturation of the blood vessels within the neovascular lesions that becomes less dependent of the VEGF action. This great variability that we can observe in the enrolled papers is the same of the daily practice, and it opens an important question: Which is the "perfect number" to define the non-responsiveness to anti-VEGF drug injections and to take into consideration other therapeutic options? In all the included papers, the mean numbers of previously anti-VEGF drug injections were really big (29.55 injections/patients and 21.22 injections/patients in the prospective and retrospective groups, respectively). This aspect could have influenced the final outcomes. Monthly intravitreal ranibizumab injections showed maximum responsiveness after the third injection in the ANCHOR study [4], whereas in the EXCITE study the maximum responsiveness was achieved after the sixth injection [66], followed by CMT and BCVA stabilization. Therefore, the "perfect number" could be smaller than that

reported in the papers included in this review and should be in the range between 3 and 6 injections of the same anti-VEGF drug: If there is not a complete fluid reabsorption or reduction in CMT of at least 100 μm from baseline, treatment is not effective and a different therapeutic approach should be taken into consideration.

The most plausible pathophysiological hypothesis for initial resistance could be related to the different levels of vitreous VEGF-A in each patient. This hypothesis is supported by studies that have found higher VEGF vitreous concentrations in patients with neovascular AMD compared with healthy controls [67] and higher VEGF vitreous concentrations being associated with a worse prognosis [68]. Indeed, anti-VEGF drugs work by blocking the VEGF-A protein. In the presence of higher vitreous VEGF-A concentrations, the standard dose of the commercially available anti-VEGF drug could not completely neutralize the vascular growth factor [63]. Increasing the injected drug dose appears as a valid alternative, but extends biological activity by only half-life time [69]. In the SAVE study, neovascular AMD patients refractory to commercially available anti-VEGF drugs (ranibizumab 0.5 mg/0.05 ml) were switched to higher intravitreal concentrations of the same drug (ranibizumab 2.0 mg/0.05 ml) [21]. Despite the good results obtained, this clinical trial is burdened by an important limitation: The enrolled patients were probably treated for months with an inadequate dose of ranibizumab. Therefore, the functional recovery was limited (+3.3 ETDRS letters at 3 months), and retinal fluid was still present in 70 % (45/64) of patients at the end of 2 years [21]. Interestingly, also the LAST trial has investigated the effects of ranibizumab 2.0 mg/0.05 ml in refractory exudative AMD, with significant improvement in BCVA ($P < 0.001$), even though the interpretation of these data is limited by the small number of enrolled patients ($n = 9$) [22]. However, it should be noticed that in naïve exudative AMD patients the HARBOR study failed to demonstrate any clinical advantage of 2.0 mg ranibizumab dose over the 0.5 mg ranibizumab dose [70].

No large studies have been conducted to specifically evaluate the increased frequency of treatment to more than the standard, monthly treatment regimens. Using pharmacokinetic modeling, Stewart et al. [71] evaluated whether dosing bevacizumab or ranibizumab every 2 weeks could be beneficial. Their results suggested that the increased trough binding activity achieved with the 2 weekly dosing could explain the improved results noted in some of their patients with persistent fluid. However, biweekly ranibizumab is not approved by Food and Drug Administration (FDA) and by European Medicines Agency (EMEA); moreover, this treatment regimen has significant cost implications and higher ocular risks. In addition, any regimen requiring appointments and potential treatment every 2 weeks may be difficult to follow by the elderly AMD patients, potentially affecting compliance and hence outcomes.

Another explanation for the *innate resistance* to anti-VEGF drugs could be represented by early up-regulation of pro-angiogenic factors other than VEGF-A, such as VEGF-B, PlGF, tumor necrosis factor α (TNFα), and/or down-regulation of anti-angiogenic factors, such as soluble vascular endothelial growth factor receptor-2 (VEGFR-2) or thrombospondins [63].

Currently, the mechanism of *tachyphylaxis* is unclear. It has been estimated that nearly 2 % of the patients may develop *tachyphylaxis* during the anti-VEGF therapy [15]. The lower pharmacological response over time was noticed to develop regardless the initial treatment with ranibizumab or bevacizumab. Some authors observed that in some patients a decreased clinical response occurred just after two anti-VEGF drug injections, whereas other patients did not show *tachyphylaxis* until they underwent ten or eleven injections [20]. In the article by Forooghian and colleagues, the median time to develop *tachyphylaxis* was 100 weeks, and the median number of intravitreal bevacizumab injections prior to establish *tachyphylaxis* was eight [72]. Additionally, Schaal and co-workers described that nearly three injections were required before the efficacy decreased to 50 % of the initial OCT response [16]. Although no conclusive data are currently available on the modulation of pro- and anti-angiogenic factors in the vitreous after anti-VEGF drug injections, the clinical drug resistance could be reasonably determined by up-regulation of pro-angiogenic factors other than VEGF-A, such as VEGF-B [73] and PlGF [74] and/or down-regulation of anti-angiogenic factors such as thrombospondins. Indeed, continuous VEGF blockade up-regulates the production of pro-angiogenic factors and overwhelms the effects of anti-VEGF agents [17, 75–77]. Finally, a recent work by Leveziel and colleagues seems to suggest that immunization against ranibizumab could be observed and may influence the clinical response [78]. Indeed, after intravitreal injection, a systematic immune response to VEGF inhibitors in patients' serum as well as a local immune response from a compromised blood-ocular barrier may contribute to the formation of measurable, neutralizing antibodies to bevacizumab and ranibizumab. Such a response also may account for the occurrence of sterile uveitis after repeated injections with anti-VEGF agents [79, 80]. Therefore, the switch to aflibercept, a drug with a wider range of targets (i.e., VEGF-A, VEGF-B, and PlGF) associated with the fact that no immune response could be present for the new drug, theoretically may lead to sustained benefits in eyes refractory to bevacizumab, ranibizumab or both.

Finally, pharmacogenetic aspects may have an important role in the identification of non-responders and in the

management of neovascular AMD patients. In particular, the certain identification of a clear association between genotypes or haplotypes of genes involved in the therapeutic response of neovascular AMD could be a prerequisite for any further study to establish effective anti-VEGF treatment regimens and dosing outsets [81, 82]. As an example, Kitchens and colleagues, based on the results of optical coherence tomography, described that patients who carried the *LOC387715 A69S TT* genotype were significantly more likely to be classified as a non-responder compared to those with the GG and GT genotypes ($P = 0.00071$) [83], whereas Korean patients harboring *VEGF* rs699947 AA genotype had an increased chance of good response to ranibizumab compared with other genotypes ($P = 0.0071$) [84]. Moreover, the same group described, for visual outcome measures, that *VEGFA* rs3025039 CC and CT genotypes were significantly associated with the lack of visual improvement after month 24 from the beginning of ranibizumab treatment compared to patients carrying TT genotype [85].

Thus, based on the growing pharmacogenetic data, it may be conceivable to personalize the anti-angiogenic therapy based on the genetic background, starting the administration of the proper anti-VEGF drug, rather than waiting for the treatment failure before choosing a different therapeutic approach. However, randomized multicenter pharmacogenetic studies on ranibizumab, bevacizumab, and aflibercept should be required before to answer to this important issue.

Conclusion

In conclusion, analysis of the papers reported in this review demonstrates that switching from bevacizumab/ranibizumab to aflibercept injections can improve outcomes successfully in refractory neovascular AMD patients. The mechanism for these effects is not yet completely understood.

However, based on these data and premises, standard criteria for the early identification of the subset of non-responder patients are urgently needed. It would be possible to switch to a different therapeutic approach, such as aflibercept injections, increasing the chances to preserve visual acuity. Indeed, the quality of scar tissue is another key aspect for the final prognosis: if a proper and earlier treatment is administered, a smaller scar in the retinal tissue will be found, resulting in a better functional recovery. In fact, most of the reports of this review highlighted a significant anatomical response after switching to aflibercept, but not a significant functional improvement. This could be the results of an inadequate treatment administered for months or years, which led to a macular scar formation, reducing margins of visual acuity improvement even in case of complete reabsorption of the intraretinal and/or sub-retinal fluid.

Larger studies and longer follow-up are needed to determine whether these anatomical gains and VA findings can be sustained.

Compliance with ethical standards

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