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RESEARCH ARTICLE

Long term results of three anti-vascular endothelial growth factor agents in pachychoroid neovasculopathy

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ABSTRACT

Purpose: To assess morphological changes and visual results in eyes with pachychoroid neovasculopathy (PNV) that underwent different intravitreal anti-vascular endothelial growth factor (VEGF) agents.

Materials and methods: This is a retrospective, observational, comparative study that included 76 PNV eyes in 76 patients that were allocated to three groups according to the monotherapy injection procedure, as follows: the intravitreal bevacizumab (IVB) group, intravitreal ranibizumab (IVR) group, and intravitreal aflibercept (IVA) group. Central macular thickness (CMT), best-corrected visual acuity (BCVA), and subfoveal choroidal thickness (SFCT) were measured at baseline, after treatment 1st month, 3rd month, 6th month, and 12th month, and at the final post-treatment examination.

Results: Mean age of the patients was 57.31 ± 5.91 years (range: 34–67 years). The mean duration of follow-up was 31.50 ± 12.91 months (range: 13–60 months). The IVB group included 30 eyes, the IVR group included 22 eyes, and the IVA group included 24 eyes. There weren't any significant differences in BCVA changes between the groups at any post-baseline measurement time point. Although CMT did not change significantly in the IVB group from baseline to the final follow-up visit (baseline: 376.33 ± 86.31 μm; final visit: 340.80 ± 122.70 μm) ($p = 0.172$), CMT did change significantly in the IVA group (baseline: 383.41 ± 131.83 μm; final visit: 297.33 ± 103.81 μm) ($p = 0.029$) and IVR group (baseline: 379.18 ± 97.93 μm; final visit: 335.72 ± 111.45 μm) ($p = 0.041$). SFCT decreased significantly in the IVR and IVA groups ($p = 0.015$ and $p < 0.001$, respectively). The mean number of injections was 12.06 ± 4.72 (range: 6–20) in the IVB group, 11.81 ± 3.31(range: 7–17) in the IVR group, and 7.16 ± 3.15 (range: 4–13) in the IVA group ($p = 0.004$).

Conclusion: All three anti-VEGFs were effective in terms of visual results in patients with PNV. Patients treated with IVA required fewer injections than those treated with IVB or IVR. Furthermore, IVR and IVA treatment significantly decreased SFCT, whereas IVB did not.

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Pachychoroid neovasculopathy; chronic central serous chorioretinopathy; subfoveal choroidal thickness; anti-vascular endothelial growth factor agents

Introduction

Pachychoroidopathy is characterized by enlargement of the outer choroidal vessels (pachyvessels) that is a result of a decrement of choriocapillaris thickness and an increment of choroidal thickness[1]. As central serous chorioretinopathy (CSC) is a pachychoroid spectrum disorder, choroidal neovascularization (CNV) caused by CSC can be defined as pachychoroid neovasculopathy (PNV)[2].

Pang and Freund[1] were the first to suggest the diagnosis of PNV in eyes with type 1 neovascularization (NV) and choroidal thickening, without typical age-related macular degeneration (AMD) or degenerative alterations. More recently, it was reported that type 1 NV can develop due to chronic CSC (cCSC), which is a typical pachychoroid spectrum disease, and that it can mimic neovascular AMD (nAMD)[3,4]. Research suggests that pachychoroid disease falls within the spectrum of CSC and polypoidal choroidal vasculopathy (PCV)[5].

The pathophysiology of PNV has been mistakenly thought of as nAMD and has been considered as type 1 NV of the enlarged choroidal vessels; hence, it can be predicted that patients with PNV might benefit from anti-VEGF therapies. Furthermore, several studies reported that intravitreal anti-VEGF agents effectively treat PNV[5,6]. Following the publication of the MINERVA study's results of the intravitreal anti-VEGF agent ranibizumab in 2018, it became the first-line on-label treatment for CNV caused by cCSC[7].

The current study's goal was to compare the results of three anti-VEGF agents [intravitreal bevacizumab (IVB), intravitreal ranibizumab (IVR), and intravitreal aflibercept (IVA)] administered as monotherapy for PNV.

Materials and methods

The medical records of 76 PNV eyes in 76 patients that were treated with intravitreal injections at a tertiary university

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hospital between October 2014 and March 2020 were retrospectively reviewed. The study protocol was approved by the local ethics committee and was conducted in accordance with the tenets of the Declaration of Helsinki. All the patients provided written informed consent.

Patients were diagnosed with PNV if all the undermentioned criteria were disclosed.

1. PNV was the type 1 NV form, consisting of fluid (sub-RPE fluid and whole retinal layers), fibrin, haemorrhage, and exudative changes, as well as pachyvessels exceeding choroidal thickness in the healthy eye[8].
2. The following CSC or PNV properties: An increase in choroidal vascular permeability based on indocyanine green angiography (ICGA); RPE abnormality not associated with CNV lesions; enlarged outer choroidal vessels on optical coherence tomography (OCT); an increase in choroidal thickness due to type 1 NV or having been previously diagnosed with CSC, even if subfoveal choroidal thickness (SFCT) was $<270\,\mu m$[9,10].
3. Spectral-domain optical coherence tomography (SD-OCT) findings of shallow irregular retinal pigment epithelium detachment (PED) in the CNV region and optical coherence tomography angiography (OCTA) (Topcon DRI-OCT Triton Plus, Topcon Corporation, Tokyo, Japan) findings indicative of CNV from the outer capillary plexus to the choriocapillaris[9] (shallow irregular PED was referred to as a double layer sign, primarily covering the branching vascular networks that feed polypoid lesions in PCV[11]).
4. All patients included in the study were emmetropic and were homogeneously distributed in terms of age[12]. Furthermore, the presence of drusen was excluded from PNV diagnostic criteria, because pachychoroid-related drusen (pachy drusen) can be observed in pachychoroid spectrum disorders[13,14].

The diagnosis of PNV was confirmed *via* fundus examination, fundus photography, SD-OCT (Spectral Domain OCT, Cirrus Zeiss, and Heidelberg Spectralis, Heidelberg Engineering), fundus fluorescein angiography (FFA), and ICGA (Heidelberg Spectralis HRA + OCT, Heidelberg Engineering). Patients with recalcitrant subretinal fluid (SRF) were followed-up more intensively if required.

Inclusion criteria

Patients diagnosed with PNV that did not receive treatment (anti-VEGFs and/or photodynamic therapy [PDT]) during the previous 6 months (washout period) were included in the study. All patients with CNV, and a documented history and/or clinical characteristics of PNV based on FFA and SD-OCT were included. All the included PNV patients used only one anti-VEGF agent (no switching).

Exclusion criteria

Patients with systemic disorders that can exacerbate macular dysfunction, including diabetes and glaucoma, those that did not regularly present for follow-up, patients with a history of retinal disease (notably retinal vascular disorders), those with AMD, those with refractive errors ≤ -3 dioptres (D) or $\geq +3$ D, patients with CNV secondary to other diseases, except for cCSC, patients with a history of ocular surgery, except phacoemulsification, and those with a history of argon laser treatment were excluded from the study. Moreover, patients have media opacities that impaired fundus imaging throughout the follow-up period was excluded.

Examinations

Ocular examinations and OCT measurements were performed before intravitreal injections (baseline), after post-injection months 1, 3, 6, and 12, and at the final follow-up visit. BCVA was taken with a Snellen chart and was converted to a logarithm of the minimum angle of resolution (log MAR) for statistical analysis. Goldmann applanation tonometry was used to assess intraocular pressure (IOP). Slit lamp microscopy was used to examine the anterior and posterior segments of each eye using a 90 dioptre non-contact lens.

Treatment protocol

The patients were classified into three groups according to monotherapy injection treatment, as follows: IVB group, IVR group, and IVA group. When choosing monotherapy, the physician and the patient's preferences were considered. First, all patients received three consecutive injections (loading dose), followed by monthly follow-up; when OCT showed that central macular thickness (CMT) increased and/or BCVA decreased, a single dose of IVR (0.5 mg/0.05 ml), IVA (2 mg/0.05 ml), or IVB (1.25 mg/0.05 ml) was administered as a monotherapy based on the Pro Re Nata (PRN) protocol[15]. Eyes were treated until a complete anatomical response was achieved after the disappearance of hyper reflective spots noted *via* SD-OCT or the inactivity on FFA. Underlying cCSC activation was considered in cases of recalcitrant SRF despite repeated anti-VEGF injections. This situation was evaluated in favour of CNV inactivation and anti-VEGF injections were discontinued.

Inadequate treatment response criteria

- Inadequate response to ≥ 3 anti-VEGF injections, despite monthly dosing.
- Evidence of prior disease activity (CMT decreased $<50\,\mu m$, BCVA decreased >5 letters, intraretinal or subretinal fluid increased based on OCT) was observed in the PNV eye following anti-VEGF was initiated.
- Exacerbation of disease activity during the course of follow-up, such as increased specific CNV activation caused by PNV, including the presence of intraretinal or subretinal fluid based on OCT.
- When SRF persisted to a lesser degree for 4 weeks following the last anti-VEGF injection, it was considered inactivation of CNV or an inadequate response; these two

conditions were differentiated based on FFA, ICGA, and OCTA.

Inactivation of CNV and cCSC activation criteria

- SRF that persists for 4 weeks after the last anti-VEGF injection.
- Administration of >3 injections of anti-VEGF.
- No increase or unchanged BCVA (≥5 letters) following initiation of anti-VEGF.
- Unchanged or increased SRF density after the last anti-VEGF injection, excluding the diagnosis of CNV.
- CNV inactivation is based on regression of vascularization *via* FFA, ICGA, and OCTA.

Eyes unresponsive to anti-VEGF treatment or that developed tachyphylaxis other than the above criteria were excluded from the study. SD-OCT was performed during every follow-up visit. There were no criteria for repeat FFA, which was not performed at every visit and was left to physician preference. Subgroup analysis was performed for eyes with recalcitrant SRF.

Criteria for repeating anti-VEGF injections

- Eyes with persistent subretinal haemorrhage received anti-VEGF monotherapy during follow-up visits.
- No improvement or deterioration of BCVA (≥5 letters) in addition to CNV findings after anti-VEGF therapy.
- No decrease in CMT ≥50 μm based on SD-OCT.
- Additional doses were injected in patients with inadequate treatment response and change in SRF was evaluated 4 weeks after injection. In the case of no change in SRF anti-VEGF administration was terminated and considered CNV inactivation.

All procedures were performed under sterile conditions. Intravitreal anti-VEGFs were injected using a 30 gauge needle at a distance of 4 mm from the temporal limbus in phakic eyes and 3.5 mm from the temporal limbus in pseudophakic eyes. Full ophthalmic examination was performed during each follow-up visit, and BCVA, CMT, SFCT, and central macular volume (CMV) were analyzed using SD-OCT and enhanced depth imaging optical coherence tomography (EDI-OCT). All examinations were performed between 09.00 and 14.00 in consideration of the fact that choroidal thickness varies during the day.

Subfoveal choroidal thickness was measured as the vertical distance between the base of the subfoveal RPE (hyperreflective line of Bruch's membrane) and the hyperreflective margin of the choroidoscleral junction, using fovea-centered SD-OCT images (including either 19 or 31 horizontal lines [6 × 6-mm areas]).

Inter-rater and intra-rater agreement

Two raters accurately evaluated 10% of eight SD-OCT images to correctly determine inter-rater agreement. To calculate rater dependability the same eight images were segmented by a classifier 1 week later. Internal and inter-rater reliability for image binarization was assessed using the exact agreement model of the intra-class correlation coefficient (ICC) (95% CI). In addition, Bland-Altman analysis was performed to determine the mean difference between binarization image measurements[16]. Furthermore, random scans showing distinct choroidal thicknesses were reviewed by both raters to ensure agreement amongst evaluators. After an agreement was compromised by evaluators, all measurements were combined by one author.

Statistical analysis

Data were analyzed using IBM SPSS Statistics for Windows v.22.0 (IBM Corp., Armonk, NY, USA). Descriptive statistics are shown as mean ± *SD* and range. The student's *t*-test or the Wilcoxon signed-rank test was used to identify significant differences in BCVA, CMT, and SFCT between baseline and various follow-up time points. Baseline and post-injection BCVA, CMT, and SFCT were compared using one-way ANOVA or the Kruskal–Wallis test. The Kolmogorov-Smirnov test was used to determine if the normality of data distribution between groups was homogeneous. The chi-square test was utilized to analyze independent quantitative data between groups. The Mann–Whitney *U* test was used for group comparison of quantitative data. The distribution of non-homogeneous data at baseline, at months 1, 3, 6, and 12, and the final follow-up visit was compared *via* MANOVA or the Friedman test. Bivariate correlations were analyzed using Spearman's rank correlation test. The homogeneity of the variance of recalcitrant SRF was calculated using the Levene test. The Bonferroni test was used for *post-hoc* correction. Pairwise comparison between groups in the presence of recalcitrant SRF percentages was performed. The level of statistical significance was set at $p < 0.05$.

Results

The medical records of 91 eyes with PNV were reviewed; however, 15 eyes were excluded because of poor OCT image quality ($n = 5$), lack of visualization of the choroscleral interface ($n = 2$), and anti-VEGF switching ($n = 8$). Intra-examiner ICC for SFCT was 0.996 (95% CI: 0.989–0.999) and inter-examiner ICC was 0.989 (95% CI: 0.982–0.998). The IVB group included 30 eyes, the IVR group included 22 eyes, and the IVA group included 24 eyes.

In terms of both systemic and topical medical treatment of cCSC before the study, 19 eyes were treated with carbonic anhydrase inhibitors (systemic), and brinzolamide and nepafenac (topical), and 10 eyes received full-dose PDT (IVB group: $n = 3$; IVR group 2: $n = 3$; IVA group: $n = 4$) ($p = 0.894$), and seven eyes half-dose PDT (IVB group: $n = 3$; IVR group: $n = 2$; IVA group: $n = 2$ ($p = 0.959$). In all, two eyes were treatment-naive before the study because there was only PED, without the involvement of the central macula or SRF accumulation. Before the study, none of the patients received anti-VEGF treatment.

The mean age of the patients was 57.31 ± 5.91 years (range: 34–67 years). In total, 40 (52%) of the patients were

male and 36 (48%) were female. The study included 40 right eyes (52%) and 36 left eyes (48%). The mean duration of follow-up was 31.50 ± 12.91 months (range: 13–60 months). There were not any significant differences in age ($p = 0.828$), gender ($p = 0.799$), duration of follow-up ($p = 0.375$), baseline BCVA ($p = 0.256$), CMT ($p = 0.517$), CMV ($p = 0.262$), IOP ($p = 0.154$), and SCFT ($p = 0.763$), or presence of SRF at baseline between the three groups ($p = 0.649$). Table 1 summarizes the clinical and demographic characteristics of the study cohort. The study results are presented in Table 2. The change in IOP from baseline to the follow-up period in the IVR and IVA groups was significant ($p < 0.05$) (Table 2).

In the IVB group, there was a significant increase in BCVA at the end of month 1, as compared to baseline ($p = 0.033$). Although CMT and CMV decreased at the end of month 1, it subsequently increased during the remainder of the follow-up period, but not significantly ($p > 0.05$). There was a significant decrease in CMT between baseline, and month 1 and month 3 ($p = 0.001$ and $p = 0.015$, respectively). There wasn't a significant decrease in SFCT during the follow-up period ($p = 0.148$).

In the IVR group, there was a significant increase in BCVA at month 1, as compared to baseline ($p = 0.011$). CMT decreased significantly throughout the follow-up period ($p = 0.041$). Additionally, there was a significant difference in CMV from baseline to the final follow-up visit ($p = 0.038$). SFCT changed significantly throughout all follow-up periods ($p = 0.015$).

In the IVA group, there was a significant increase in BCVA from baseline to month 1 and month 3 ($p = 0.011$ and $p = 0.023$, respectively). CMT decreased significantly throughout the follow-up period and to a greater degree than in the IVB and IVR groups ($p = 0.029$). There was also a significant decrease in SFCT ($p < 0.001$).

When we compared three groups based on follow-up periods, no statistical change was found in CMT, CMV, SFCT, and IOP values ($p > 0.05$). In terms of BCVA, the inter-group analysis showed that the IVA group had significantly better visual acuity beginning at 3 months than the IVB and IVR groups (month 3: $p = 0.048$; month 6: $p = 0.039$; month 12: $p = 0.028$; final follow-up visit: $p = 0.032$). There was a positive correlation between baseline and final BCVA ($r = 0.473$, $p = 0.003$). Increased SFCT thickness had a negative effect on baseline BCVA ($r = -0.464$, $p = 0.003$).

There was a significant difference in the presence of recalcitrant SRF between the three groups ($p = 0.027$). Recalcitrant SRF (expressed as the presence/absence ratio and presence percentage [%]) was 20/10 and 66% in the IVB group, 14/8 and 63% in the IVR group, and 6/18 and 25% in the IVA group. The presence percentage of recalcitrant SRF was significantly lower in the IVA group than in the IVB and IVR groups ($p = 0.031$ and $p = 0.037$, respectively); however, there wasn't a significant difference in the presence percentage of recalcitrant SRF between the IVR and IVB groups ($p = 0.991$).

Figures 1 and 2 show FFA, ICGA, and SD-OCT images of representative patients in the IVR and IVA groups. En-face and structural OCT with flux signal images of a membrane that developed as a result of CNV are shown in Figure 3. Changes in BCVA in all three groups during follow-up are shown in Figure 4. Table 3 shows intra-group comparisons of CMT, BCVA, and SFCT measurements between baseline and each follow-up visit.

There was a significant difference in the number of injections between the three groups ($p = 0.004$). In the IVA group, the number of injections was significantly lower than in the IVB and IVR groups ($p = 0.004$ and $p = 0.003$, respectively). Moreover, there wasn't a significant difference in the number of injections between the IVB and IVR groups ($p = 0.878$). The number of injections and differences in SFCT in and between the three groups during the follow-up period are shown in Figure 5.

In total, 40 eyes had recalcitrant SRF (40/76, 52%). Significant changes in BCVA were not observed in the eyes with recalcitrant SRF (0.83 ± 0.89 at baseline *vs.* 0.98 ± 0.98 at the final follow-up visit) ($p = 0.374$). SRF was not observed in any of the patients with a history of PDT (both full and half dose) before the study. There was a positive correlation between recalcitrant SRF and choroidal thickening ($p < 0.001$, $r = 0.867$) and younger age ($p = 0.023$, $r = 0.681$). Dry macula was achieved at a higher rate in the IVA group than in the IVB and IVR groups ($p = 0.027$). In terms of CNV type, all 76 eyes had type 1 NV according to baseline OCT; PCV was present in eight of the eyes based on ICGA and OCTA (IVB group: $n = 4$; IVR group: $n = 2$; IVA group: $n = 2$). Subretinal haemorrhage was not observed in any of the patients at baseline and during follow-up periods. In addition, there were no complications that required surgical intervention, such as pars plana vitrectomy during follow-up periods.

Discussion

The present study shows that IVA is better for achieving SRF absorption in eyes with PNV during long-term follow-up than IVB and IVR. Although BCVA results did not differ significantly between the three groups following three loading doses, an evident increase in visual acuity was observed in the IVA group at month 3. Also concluded in complete SRF absorption in 75% of eyes in the IVA group.

Peiretti et al.[17] compared the efficacy of intravitreal anti-VEGF agents alone or combined with PDT for the treatment of CNV caused by cCSC. The use of PDT and anti-VEGF agents individually or in combination exhibited similar clinical efficacy in the treatment of CNV caused by cCSC. Furthermore, a recent report by Radke et al.[18] reported that intravitreal ziv-aflibercept (IVZ) used to treat CNV secondary to naive cCSC resulted in positive anatomical and visual outcomes; however, these studies did not provide sufficient data on anti-VEGF treatment of PNV.

Bevacizumab and ranibizumab are monoclonal antibodies that target all isomeric forms of the VEGF-A family. Both drugs were proven to be quite potent in the treatment of CNV by many clinical trials. Aflibercept has the capacity to function as a VEGF-A factor antagonist as well as to bind to other growth factors. In contrast to bevacizumab and ranibizumab, aflibercept does not have the structure of an antibody; it consists of a recombinant soluble decoy receptor fusion protein[19].

Table 1. The clinical and demographic data of the patients.

Clinical characteristics	Group 1 Bevacizumab	Group 2 Ranibizumab	Group 3 Afflibercept	*p*-Values
Eyes	30	22	24	
Gender	$14^{f}16^{m}$	$11^{f}11^{m}$	$11^{f}13^{m}$	0.799
Age (mean ± *SD*)	57 ± 5	57.72 ± 4.14	57.33 ± 8.37	0.828
Side	$18^{r}12^{l}$	$10^{r}12^{l}$	$12^{r}12^{l}$	0.831
Follow up (months) (mean ± *SD*)	31.13 ± 11.69	33.81 ± 11.60	32.58 ± 15.77	0.375
Follow up (months) (range)	13–59	14–60	13–60	
Number of injections (mean ± *SD*)	12.06 ± 4.72	11.81 ± 3.31	7.16 ± 3.15	*0.004**
Number of injections (range)	6–20	7–17	4–13	
Presence of recalcitrant SRF (presence/absence)	(20/10)	(14/8)	(6/18)	*0.027**

SRF: subretinal fluid; SD: standard deviation; ^{f}: female; ^{m}: male; ^{r}: right; ^{l}: left.
*Kruskal Wallis test; $p<0.05$.

Table 2. The results of the study.

Groups		Baseline	1st month	3rd month	6th month	12th month	Final visit	*p*-Values*
Group 1 Bevacizumab	BCVA	0.82 ± 0.40	0.63 ± 0.39	0.72 ± 0.46	0.81 ± 0.49	1.05 ± 0.74	1.17 ± 0.68	<0.001*
	CMT	376.33 ± 86.31	274.33 ± 46.80	298.46 ± 115.97	327.26 ± 142.49	339.53 ± 160.96	340.80 ± 122.70	0.172
	CMV	8.91 ± 1.31	8.12 ± 0.48	8.42 ± 0.98	8.75 ± 1.21	8.59 ± 1.11	8.31 ± 1.23	0.043*
	SCFT	288.81 ± 57.30	280.00 ± 49.61	275.18 ± 43.13	286.54 ± 52.59	268.63 ± 49.02	263.36 ± 59.96	0.148
	IOP	15.66 ± 1.91	15.60 ± 1.63	15.46 ± 1.99	15.86 ± 1.88	16.06 ± 1.66	16.80 ± 1.74	0.059
Group 2 Ranibizumab	BCVA	0.83 ± 0.30	0.76 ± 0.38	0.75 ± 0.36	0.80 ± 0.59	0.90 ± 0.59	1.07 ± 0.58	0.026*
	CMT	379.18 ± 97.93	280.45 ± 84.08	290.54 ± 75.23	330 ± 124.56	293.72 ± 70.31	335.72 ± 111.45	0.041*
	CMV	8.73 ± 0.70	8.20 ± 0.63	8.43 ± 0.83	8.36 ± 0.69	8.22 ± 0.65	8.21 ± 0.83	0.038*
	SCFT	296.83 ± 35.58	274.75 ± 41.05	267.25 ± 52.81	266.25 ± 52.55	270.91 ± 39.51	257.25 ± 67.14	0.015*
	IOP	14.5 ± 1.78	14.83 ± 2.28	15 ± 1.59	15.33 ± 2.53	16 ± 2.08	17 ± 2.55	<0.001*
Group 3 Aflibercept	BCVA	0.85 ± 0.46	0.63 ± 0.50	0.66 ± 0.56	0.73 ± 0.52	0.85 ± 0.58	0.97 ± 0.55	<0.001*
	CMT	383.41 ± 131.83	328.33 ± 108.81	282 ± 85.47	300.66 ± 112.99	307.16 ± 97.67	297.33 ± 103.81	0.029*
	CMV	9.40 ± 1.73	8.82 ± 1.23	8.52 ± 0.97	8.64 ± 1.18	8.66 ± 1.35	8.50 ± 1.17	0.428
	SCFT	291.33 ± 43.43	269.26 ± 51.74	253.33 ± 67.51	246.06 ± 67.13	245.00 ± 57.94	230.46 ± 83.06	<0.001*
	IOP	14.83 ± 1.26	14.33 ± 1.96	15.16 ± 1.40	15.83 ± 1.52	16.83 ± 2.36	16.16 ± 2.12	0.001*

*Friedman test.

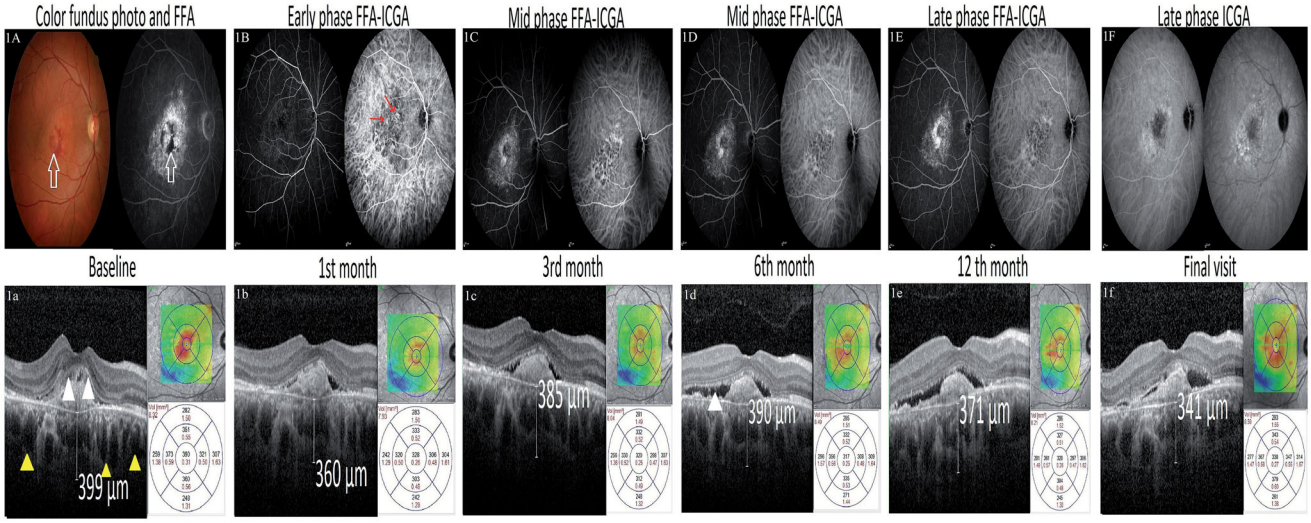


Figure 1. Colour fundus photographs, fundus fluorescein angiography (FFA), indocyanine green angiography (ICGA), central macular thickness (CMT), central macular volume (CMV), and subfoveal choroidal thickness (SFCT) images of a patient treated with IVR. The arrows in (A) indicate a haemorrhage caused by choroidal neovascularization (CNV) and FFA shows blockage related to the haemorrhage. The hyperreflective region seen in (a) is subretinal exudation due to CNV. Early-phase ICGA shows dilated choroidal vessels (red arrowheads) in (B). Optical coherence tomography (OCT) at baseline shows type 1 NV with pachy vessels (yellow arrowheads) in (a). Serous detachment associated with central serous chorioretinopathy (CSCR) activation is shown in (d).

Recent studies show that VEGF may not be the primary mediator in PNV, as it is in nAMD; therefore, the efficacy of such other growth factors as placental growth factor (PlGF) in the pathogenesis of CNV has become more important[20,21]. As such, aflibercept might cause choroidal thinning following CNV inactivation, because thinning of the choroidal stroma can develop after the decrease of leakage into the choroid from a CNV lesion. Invernizzi et al.[22] noted that choroidal thickness and the choroidal vascularity index are strongly associated with CNV activity in eyes with nAMD. CNV resolution with aflibercept might be more effective than other anti-VEGF agents at thinning of the choroid; therefore, it can prevent the progression of the disease more effectively and rapidly than IVR or IVB. In addition, choroidal thinning might hypothetically be caused by the efficacy of aflibercept's anti-VEGF action in the choroid.

Due to aflibercept's larger molecular structure and more robust binding to VEGF-A than those of ranibizumab and

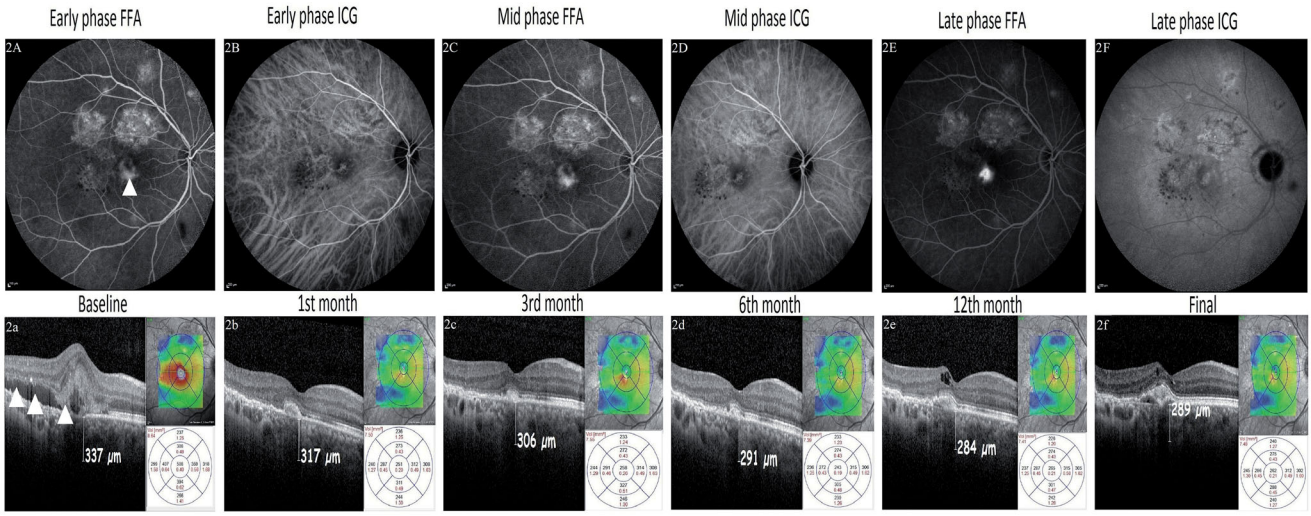


Figure 2. FFA, ICGA, CMT, CMV, and SFCT images of patients treated with IVA, according to follow-up period. Arrows in (a) indicate hyperreflective dots and exudations with subretinal fluid due to CNV. The arrow in the FFA in (A) corresponds to the region of CNV.

bevacizumab, it has an effective half-life action intravitreally, providing a longer duration of potential biological activity, despite the limited intravitreal duration of action, up to 14 days like as ranibizumab in animal model studies and also it has lower systemic biodistribution in animal models[23]. The present findings are consistent with those that show aflibercept is more effective than ranibizumab for treating nAMD patients[24,25]; however, there hasn't been a sufficient number of studies on long-term outcomes in patients with PNV for reaching a clear conclusion concerning which treatment is most effective. The present findings show that dry macula was more common and gains in visual acuity were greater in the IVA group than in the IVB and IVR groups during long-term follow-up.

Jung et al.[5] studied 31 PNV eyes treated with IVR and 23 PNV eyes treated with IVA; the drugs were injected monthly for 3 months as a loading dose. IVR was switched to IVA and IVA was switched to PDT in the 3rd month if incomplete SRF absorption was observed. The complete SRF absorption rate was higher (82.6%) in the IVA group than in the IVR group (51.6%) at 3 months ($p = 0.018$). Although the decrease in SFCT was greater in the IVA group (-35 *vs.* $-9\,\mu m$) than in the IVR group ($p = 0.013$), there wasn't a significant difference in the improvement of visual acuity or CMT ($p > 0.05$). In 13 of 15 eyes (86.7%) complete SRF absorption was achieved after switching from IVR to IVA[5]. In the present study, anti-VEGF switching was not performed in any of the eyes and the SRF absorption rate in the IVA group was 75%.

Some patients that present with type 1 NV can have clinical and imaging findings that are more compatible with a diagnosis of cCSC than a diagnosis of AMD. Such patients are generally younger males with a thick choroid togetherness with type 1 NV and with a high frequency of PCV[3]. Schworm et al.[26] recently reported that unresponsiveness to IVR can be overcome by switching to IVA. PNV eyes with persistent SRF detected 4 weeks after the last IVR injection and that had an inadequate response despite $\geq$3 IVR injections were switched to monthly IVA injections for 3 months. Complete absorption of SRF was not achieved in any of the eyes (0%) during IVR treatment, whereas complete SRF resolution was observed in eight eyes (57.1%) after switching to three injections of IVA. A significant change in SRF ($p = 0.0009$) and SFCT ($p = 0.044$) was observed only in the IVA group.

Romdhane et al.[27] reported that complete SRF resorption was noted in 50% of PNV eyes treated with intravitreal anti-VEGFs. The researchers suggested that a history of treatment for PNV had a positive effect on the current treatment response and that non-responsiveness to anti-VEGFs might be more common in treatment-naive PNV eyes. The positive response criteria for anti-VEGF therapy were associated with female gender, higher CMT measurement at the initial period, presence of final SRF, and a larger CNV flow area measured by OCTA before therapy[28]. Ateriogenesis in PNV eyes is the most commonly indicated reason for resistance to anti-VEGFs and/or PDT[29]. The present study included patients treated for PNV that had a mean of 10.44 ± 4.41 anti-VEGF injections during their follow-up.

In recent studies, a significant reduction in SFCT was reported after anti-VEGF therapies[30–32]. Schworm et al. have reported a significant reduction in SFCT in 22 eyes with PNV after 2 years of anti-VEGF therapy (first three loading doses and post-PRN regimen). And they introduced the definition of vanishing pachychoroid for the first time and argued that the reduction in long-term SFCT may not be a diagnostic criterion[30]. Similarly, Perez et al. also conducted anti-VEGF treatment according to the PRN regimen during the 12-month follow-up period in their study of 18 eyes with PNV and observed a significant reduction in SFCT. They claimed that this reduction could be evident up to the resolution of PNV activity as a result of the decrease in choroidal vascular permeability[31]. Nagai et al. administered IVA or IVR for 24 months according to the PRN protocol to 100 eyes with PCV. And the presence of baseline pachyvessels in the choroid (vertical choroidal vessel diameter $\geq 180\,\mu m$) and pachychoroid (central choroidal thickness $\geq 220\,\mu m$) were indicated as risk factors for residual exudative changes after the induction phase; in addition, they also reported that pachyvessels in the choroid were a risk factor for relapse in

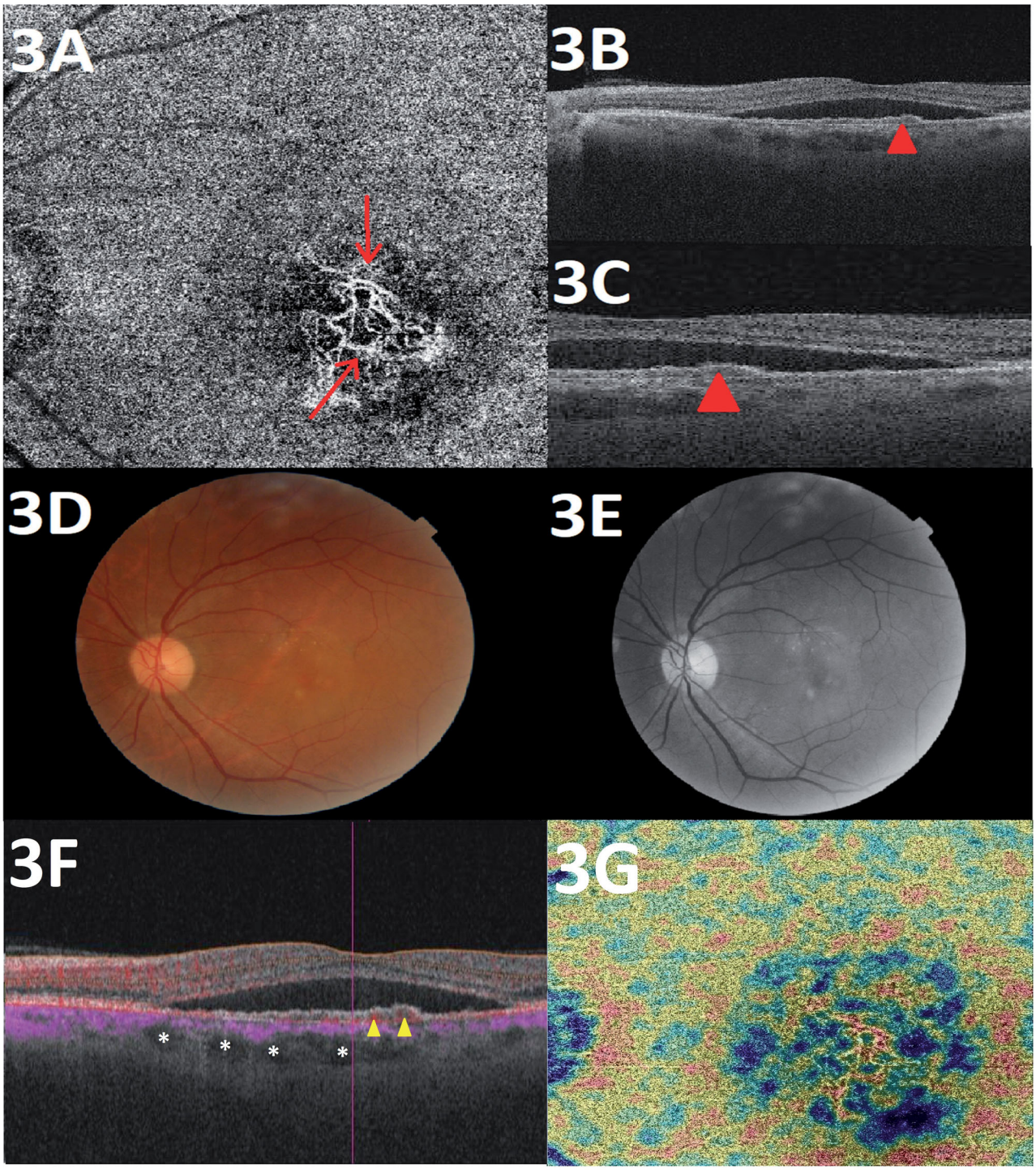
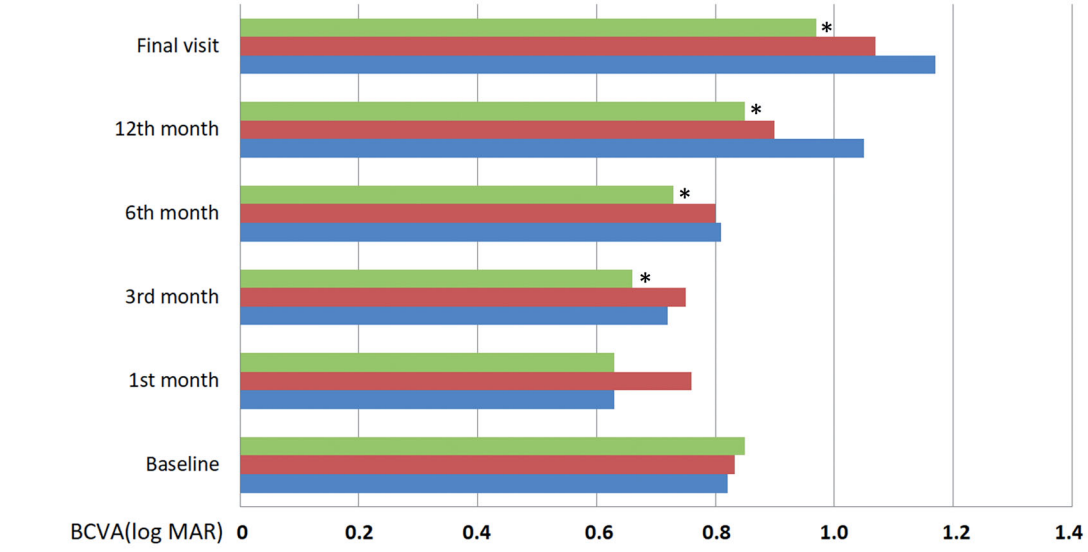


Figure 3. Optical coherence tomography angiography (OCTA) image of a membrane that developed as a result of choroidal neovascularization. In (A), the red arrows indicate the membrane imaged *via* OCTA. In (B,C), the red arrows indicate flat irregular retinal pigment epithelial detachment (PED) (double layer sign), as seen in an OCT image of the same patient. (F) Shows a flux signal in a structural OCT at baseline and type 1 NV with pachy vessels (white asterisks). Yellow arrows show flat irregular PED in the structural OCT. A density map is shown in (G).

PCV with or without pachychoroids[32]. In another study conducted by Shimizu et al., they divided 115 eyes with PCV into two groups pachychoroid and non-pachychoroid, and anti-VEGF monotherapy was implemented for 5 years. And they reported that anti-VEGF monotherapy had similar efficacy in eyes with PCV which had pachycoroid and non-pachychoroid phenotype[33]. However, a different study showed that an SFCT cut-off value of 267.5 μm could be used to predict PCV diagnosis and that PCV is more refractory to anti-VEGF treatment[34]. In the present study, only 54 (71%) eyes had a baseline SFCT >267.5 μm based on EDI-OCT and 48 (63%) eyes had SFCT <267.5 μm at the final follow-up visit. As there was a progressive decrease in SFCT during the present study's follow-up period, it is reasonable to estimate that SFCT could be >267.5 μm at the initial visit of newly diagnosed cCSC in 22 patients (29%) who had <267.5 μm. PCV was noted in eight eyes with recalcitrant SRF and SFCT >267.5 μm in the present study. Additional research is



	Baseline	1st month	3rd month	6th month	12th month	Final visit
Group 1 (bevacizumab)	0.82±0.40	0.63±0.39	0.72±0.46	0.81±0.49	1.05±0.74	1.17±0.68
Group 2 (ranibizumab)	0.83±0.30	0.76±0.38	0.75±0.36	0.80±0.59	0.90±0.59	1.07±0.58
Group 3 (aflibercept)	0.85±0.46	0.63±0.50	0.66±0.56	0.73±0.52	0.85±0.58	0.97±0.55
p value	0.256	0.538	0.048*	0.039*	0.028*	0.032*

Figure 4. Changes in visual acuity, according group and follow-up period. *p*-Values are for group comparisons at each follow-up visit.
*$p<0.05$.

Table 3. Intra-group comparison of CMT, BCVA, and SFCT between baseline and each follow-up visit.

Measurements Mean ± *SD*	Bevacizumab Group 1	Ranibizumab Group 2	Aflibercept Group 3
CMT (μm)			
Baseline	**376.33 ± 86.31**	**379.18 ± 97.93**	**383.41 ± 131.83**
Follow-up (λ)			
Month 1	274.33 ± 46.80*	280.45 ± 84.08*	328.33 ± 108.81*
Month 3	298.46 ± 115.97†	290.54 ± 75.23†	282 ± 85.47†
Month 6	327.26 ± 142.49	330 ± 124.56	300.66 ± 112.99∈
Month 12	339.53 ± 160.96δ	293.72 ± 70.31δ	307.16 ± 97.67δ
Final visit	340.80 ± 122.70	335.72 ± 111.45	297.33 ± 103.81β
p-Value	*0.001, †0.015, δ0.030	*<0.001, †0.007, δ0.003	*0.005, †<0.001, ∈0.001, δ0.002, β<0.001
BCVA (logMAR)			
Baseline	**0.82 ± 0.40**	**0.83 ± 0.30**	**0.85 ± 0.46**
Follow-up (λ)			
Month 1	0.63 ± 0.39*	0.76 ± 0.38*	0.63 ± 0.50*
Month 3	0.72 ± 0.46†	0.75 ± 0.36†	0.66 ± 0.56†
Month 6	0.81 ± 0.49	0.80 ± 0.59	0.73 ± 0.52
Month 12	1.05 ± 0.74	0.90 ± 0.59	0.85 ± 0.58
Final visit	1.17 ± 0.68	1.07 ± 0.58	0.97 ± 0.55
p-Value	*0.033, †0.03	*0.011, †0.009	*0.011, †0.023
SFCT (μm)			
Baseline	**288.81 ± 57.30**	**296.83 ± 35.58**	**291.33 ± 43.43**
Follow-up (λ)			
Month 1	280.00 ± 49.61*	274.75 ± 41.05	269.26 ± 51.74*
Month 3	275.18 ± 43.13†	267.25 ± 52.81†	253.33 ± 67.51†
Month 6	286.54 ± 52.59	266.25 ± 52.55	246.06 ± 67.13∈
Month 12	268.63 ± 49.02δ	270.91 ± 39.51δ	245.00 ± 57.94δ
Final visit	263.36 ± 59.96β	257.25 ± 67.14β	230.46 ± 83.06β
p-Value	*<0.001, †0.028, δ0.001, β0.003	†0.015, δ<0.001, β<0.001	*<0.001, †0.002, ∈<0.001, δ0.002, β0.001

p-Values from the intra-group comparison between each visit and baseline visit (λ).
Continuous and normally distributed data was assessed by unpaired Student's *t*-test, and non-normally distributed data with Mann–Whitney *U* test.

needed to identify the prognostic significance of anti-VEGF in the treatment response of a cut-off value of 267.5 μm in PNV disease.

Brandl et al.[35] observed that SFCT initially increased in patients with cCSC, and then gradually decreased over the course of the next 3 months and returned to normal after 1 year. Although the mean annual decrease in SFCT is reported to be 4.1–9.33 μm, the mean annual decrease in the present study was 25.63 μm[36]. Furthermore, after 1 year of follow-up in the present study SFCT decreased the mean by 25.91 μm in the IVB group, 16.18 μm in the IVR group, and 32.33 μm in the IVA group. In contrast to earlier studies, the PNV eyes were

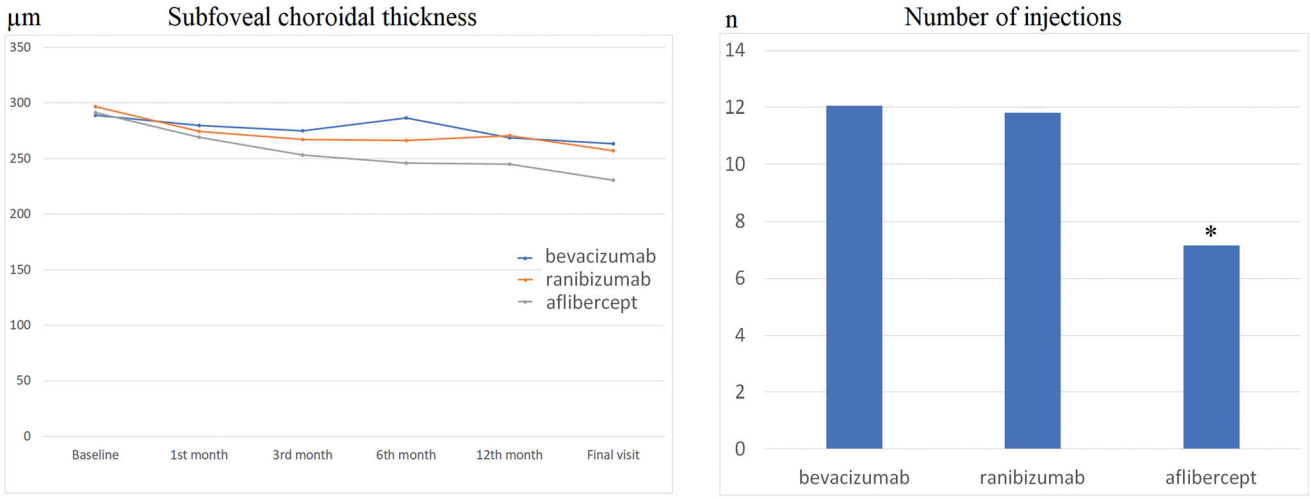


Figure 5. Comparison of the number of injections and changes in SFCT between the three groups during follow-up.
*$p<0.05$.

treated at the start of the present study and the observed decrease in SFCT was greater than that previously reported.

Variability in the present study results might be associated with such factors as pharmacokinetic and pharmacodynamic variations in treatment response, and the disease process. The strengths of the present study are comparability for different anti-VEGF agents, long-term follow-up, homogeneous distribution of SFCT measurements, and the fact that diurnal variation was taken into consideration. The present study does have several limitations, including a small sample, a retrospective design, and manual measurements, despite high intra- and inter-examiner ICC. We could not interpret drug advantages and interactions more clearly as a result of the inclusion of no anti-VEGF switched eyes in the study.

In the present study, PNV eyes were treated with three anti-VEGF agents, but the findings make it difficult to conclude which treatment protocol is superior, although IVA treatment might be the best option for maintaining anatomical and visual gains during long-term follow-up. Additional larger-scale prospective studies with longer follow-up periods are required to clarify the role of anti-VEGF agents in the treatment of PNV.

Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent

Informed consent was obtained before every surgical procedure from all individual participants included in the study.

Disclosure statement

Author Bugra Karasu declares that he has no conflict of interest. Author Yusuf Berk Akbas declares that he has no conflict of interest. Author Mert Kaskal declares that he has no conflict of interest. Author Aslan Aykut declares that he has no conflict of interest. Author Ali Rıza Cenk Celebi declares that he has no conflict of interest.

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References

1. Pang CE, Freund KB. Pachychoroid neovasculopathy. Retina 2015; 35:1–9.
2. Gallego-Pinazo R, Dolz-Marco R, Gómez-Ulla F, et al. Pachychoroid diseases of the macula. Medical hypothesis. Med Hypothesis Discov Innov Ophthalmol 2014;3:111–115.
3. Fung AT, Yannuzzi LA, Freund KB. Type 1 (sub-retinal pigment epithelial) neovascularization in central serous chorioretinopathy masquerading as neovascular age-related macular degeneration. Retina 2012;32:1829–1837.
4. Yannuzzi LA, Freund KB, Goldbaum M, et al. Polypoidal choroidal vasculopathy masquerading as central serous chorioretinopathy. Ophthalmology 2000;107:767–777.
5. Jung BJ, Kim JY, Lee JH, et al. Intravitreal aflibercept and ranibizumab for pachychoroid neovasculopathy. Sci Rep 2019;9:2055.
6. Cho HJ, Jung SH, Cho S, et al. Intravitreal anti-vascular endothelial growth factor treatment for pachychoroid neovasculopathy. J Ocul Pharmacol Ther 2019;35:174–181.
7. Lai TYY, Staurenghi G, Lanzetta P, et al. Efficacy and safety of ranibizumab for the treatment of choroidal neovascularization due to uncommon cause: twelve-month results of the MINERVA study. Retina 2018;38:1464–1477.
8. Dansingani KK, Balaratnasingam C, Naysan J, et al. En face imaging of pachychoroid spectrum disorders with swept-source optical coherence tomography. Retina 2016;36:499–516.
9. Dansingani KK, Balaratnasingam C, Klufas MA, et al. Optical coherence tomography angiography of shallow irregular pigment epithelial detachments in pachychoroid spectrum disease. Am J Ophthalmol 2015;160:1243–1254.e2.
10. Matsumoto H, Hiroe T, Morimoto M, et al. Efficacy of treat-and-extend regimen with aflibercept for pachychoroid neovasculopathy and type 1 neovascular age-related macular degeneration. Jpn J Ophthalmol 2018;62:144–150.

11. Sato T, Kishi S, Watanabe G, et al. Tomographic features of branching vascular networks in polypoidal choroidal vasculopathy. Retina 2007;27:589–594.
12. Ikuno Y, Kawaguchi K, Nouchi T, et al. Choroidal thickness in healthy Japanese subjects. Invest Ophthalmol Vis Sci 2010;51: 2173–2176.
13. Spaide RF. Disease expression in nonexudative age-related macular degeneration varies with choroidal thickness. Retina 2018;38: 708–716.
14. Matsumoto H, Mukai R, Morimoto M, et al. Clinical characteristics of pachydrusen in central serous chorioretinopathy. Graefes Arch Clin Exp Ophthalmol 2019;257:1127–1132.
15. Lalwani GA, Rosenfeld PJ, Fung AE, et al. A variable-dosing regimen with intravitreal ranibizumab for neovascular age-related macular degeneration: year 2 of the PrONTO study. Am J Ophthalmol 2009;148:43–58.e1.
16. Bland JM, Altman DG. Agreed statistics: measurement method comparison. Anesthesiology 2012;116:182–185.
17. Peiretti E, Caminiti G, Serra R, et al. Anti-vascular endothelial growth factor therapy versus photodynamic therapy in the treatment of choroidal neovascularization secondary to central serous chorioretinopathy. Retina 2018;38:1526–1532.
18. Radke N, Kalamkar C, Mukherjee A, et al. Intravitreal Ziv-aflibercept in treatment of naive chronic central serous chorioretinopathy related choroidal neovascular membrane. Case Rep Ophthalmol Med 2017;2017:5036248.
19. Cheng C-K, Chan T-Y. Rapid response to intravitreal aflibercept in neovascular age-related macular degeneration after development of tachyphylaxis to bevacizumab and ranibizumab. Taiwan J Ophthalmol 2014;4:40–44.
20. Papadopoulos Z. Aflibercept: a review of its effect on the treatment of exudative age-related macular degeneration. Eur J Ophthalmol 2019;29:368–378.
21. Terao N, Koizumi H, Kojima K, et al. Distinct aqueous humour cytokine profiles of patients with pachychoroid neovasculopathy and neovascular age-related macular degeneration. Sci Rep 2018;8: 10520.
22. Invernizzi A, Benatti E, Cozzi M, et al. Choroidal structural changes correlate with neovascular activity in neovascular age related macular degeneration. Invest Ophthalmol Vis Sci 2018;59: 3836–3841.
23. Christoforidis JB, Briley K, Binzel K, et al. Systemic biodistribution and intravitreal pharmacokinetic properties of bevacizumab, ranibizumab, and aflibercept in a nonhuman primate model. Invest Ophthalmol Vis Sci 2017;58:5636–5645.
24. Koizumi H, Kano M, Yamamoto A, et al. Subfoveal choroidal thickness during aflibercept therapy for neovascular age-related macular degeneration: twelve-month results. Ophthalmology 2016;123: 617–624.
25. Yamazaki T, Koizumi H, Yamagishi T, et al. Subfoveal choroidal thickness after ranibizumab therapy for neovascular age-related macular degeneration: 12-month results. Ophthalmology 2012;119: 1621–1627.
26. Schworm B, Luft N, Keidel LF, et al. Ranibizumab non-response in pachychoroid neovasculopathy: effects of switching to aflibercept. Sci Rep 2020;10:8439.
27. Romdhane K, Zola M, Matet A, et al. Predictors of treatment response to intravitreal anti-vascular endothelial growth factor (anti-VEGF) therapy for choroidal neovascularization secondary to chronic central serous chorioretinopathy. Br J Ophthalmol 2020; 104:910–916.
28. Spaide RF. Optical coherence tomography angiography signs of vascular abnormalization with antiangiogenic therapy for choroidal neovascularization. Am J Ophthalmol 2015;160:6–16.
29. Siedlecki J, Schworm B, Priglinger SG. The pachychoroid disease spectrum-and the need for a uniform classification system. Ophthalmol Retina 2019;3:1013–1015.
30. Schworm B, Luft N, Keidel LF, et al. Vanishing pachy-choroid in pachychoroid neovasculopathy under long-term anti-vascular endothelial growth factor therapy. BMC Ophthalmol 2021;21:269.
31. Padrón-Pérez N, Arias L, Rubio M, et al. Changes in choroidal thickness after intravitreal injection of anti-vascular endothelial growth factor in pachychoroid neovasculopathy. Invest Ophthalmol Vis Sci 2018;59:1119–1124.
32. Nagai N, Suzuki M, Minami S, et al. Dynamic changes in choroidal conditions during anti-vascular endothelial growth factor therapy in polypoidal choroidal vasculopathy. Sci Rep 2019;9:11389.
33. Shimizu Y, Miyata M, Ooto S, et al. Pachychoroid-phenotype effects on 5-year visual outcomes of anti-VEGF monotherapy in polypoidal choroidal vasculopathy. Acta Ophthalmol. 2022 Jun;100(4): e943–e949.
34. Chang Y-C, Cheng C-K. Difference between pachychoroid and non-pachychoroid polypoidal choroidal vasculopathy and their response to anti-vascular endothelial growth factor therapy. Retina 2019;1:1.
35. Brandl C, Helbig H, Gamulescu MA. Choroidal thickness measurements during central serous chorioretinopathy treatment. Int Ophthalmol 2014;34:7–13.
36. Wei WB, Xu L, Jonas JB, et al. Subfoveal choroidal thickness: the Beijing eye study. Ophthalmology 2013;120:175–180.