

# Autologous translocation of the choroid and retina pigment epithelial cells (RPE) in age-related macular degeneration: Monitoring the viability of choroid and RPE patch with indocyanine green angiography(ICGA) and fundus autofluorescence(FAF)

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## ABSTRACT

**Purpose:** To investigate the functional and anatomical results of autologous retinal pigment epithelial(RPE) cells and choroidal translocation after removal of the subfoveal choroidal neovascular membrane(CNVM) in patients with exudative age-related macular degeneration(AMD). To monitor the viability of choroidal patch with indocyanine green angiography(ICGA) and fundus autofluorescence(FAF)

**Methods:** This study was conducted as a retrospective, interventional case series, and evaluation of 8 patients ;4 patients had large (> 1 disk diameter) subfoveal choroidal membranes, 3 with massive subretinal hemorrhage and 1 case with suprachoroidal hemorrhage(SCH) + rhegmatogenous retinal detachment(RRD). After removal of the CNVM, the autologous full-thickness patch of the RPE, bruch's membrane, choriocapillaris, and choroid was excised from the midperiphery and placed under the macula. At the 1 st month, 3rd month, 6th month and final examination, color fundus pictures and optical coherence tomography (OCT) were performed by preferred fixation of the OCT-light. Visual test with the early treatment of diabetic retinopathy study(ETDRS), OCT imaging with fixation, scanning with laser ophthalmoscopy autofluorescence, and ICGA were performed to evaluate the viability of choroidal patch at each visits.

**Results:** This study was carried out in 8 patients with a mean follow-up of  $14.12 \pm 8.16$  (range 7-30 months) months. The mean age was  $73 \pm 7.17$ (range, 60-80 years) years. Pre-operative visual acuity ranged from hand motion (HM) (20/2000) to light perception (LP)(20/20000). Post-operative vision ranged from HM (20/2000) to 0.15(20/125). In 6 patients, autofluorescence was reflected in FAF imaging and lipofuscin activity was evaluated as viable. Post-operative subretinal hemorrhage was encountered in 1 (12.5%) patient and it also resolved spontaneously. There was a statistically significant increase in visual acuity at the postoperative final visit compared to baseline. ( $p = 0.027$ )

**Conclusions:** After removal of the CNVM, translocation of a full-thickness patch with the autologous peripheral RPE and choroid can be performed at the macula, resulted in survival and functional graft for 6 months and moreover, hereby viability of the choroid and RPE patch were monitored by imaging methods such as FAF and ICGA.

## 1. Introduction

Age-related macular degeneration (AMD) is one of the leading cause of permanent blindness in developed countries at the age of 60 and after. [1] The most common reason of severe vision loss due to AMD are choroidal ischemia and neovascularization which develops based on perfusion damage.[2]

Today, treatment of exudative AMD is usually based on thermal laser, photodynamic therapy, and frequently preferred, the intravitreal anti-vascular endothelial growth factor (VEGF) therapy. [3–8] Using these treatment modalities, the underlying cause of the disease cannot be completely eliminated but the course of the disease is slowed down.

Surgical option is a way of completely eliminating choroidal neovascularization (CNV). However, completely removal of the choroidal

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neovascular membrane (CNVM) from the eye, disrupts the functionality of the retinal pigment epithelium (RPE), bruch's membrane and choriocapillaris, compromising anatomic and functional success.[9–14]

In various studies, the viability of the graft after RPE and choroidal patch translocation was evaluated by indocyanine green angiography (ICGA) and fundus autofluorescence (FAF).[15]

Indocyanine green (ICG) is a water-soluble and tricarbo-cyanine dye of 775-dalton molecular weight, which binds to 98% protein after intravenous injection as well widely used photodiagnosis methods to show the pathologies in the choroidal tissue. [16]

Non-invasive imaging of autofluorescence of lipofuscin and other fluorophores formed under the neural retina in RPE using without sodium fluorescein by confocal scanning laser ophthalmoscope (CLO) or modified fundus cameras. The device uses the argon blue laser with a wavelength of 488 nm as the excitation light, and the barrier filter allows the passage of wavelengths of 500 nm and above. The task of FAF is to give information about RPE and photoreceptor viability by demonstrating lipofuscin activity in retinal pigment epithelial tissue. [17–30]

This study supports that only a healthy RPE, bruch's membrane, and choriocapillaris can recover macular photoreceptors and central visual function. Therefore, we preferred macular translocation surgery with a full thickness of healthy RPE cells from the mid-peripheral retina and aimed to demonstrate the viability of the graft which can be monitored by ICGA and FAF.

## 2. Materials and methods

Retrospective analysis of 8 patients performed to the autologous choroid and RPE translocation from October 2015 to March 2017 was done. This study was approved by the Institutional Review Board of Beyoğlu Education and Research Hospital and all patients received written approval in accordance with the ethical standards set out in the Helsinki Declaration. This report relates to patients with follow-up periods longer than 6 months. Patients with subfoveal CNVM greater than 1 disc diameter with or without more than 50% submacular hemorrhage in fundus fluorescein angiography (FFA) were eligible for RPE translocation.

*This surgery was performed in these eyes;*

- 1) Visual loss due to active or progressive CNVM unresponsive to treatment despite at least three anti-VEGF injections and once photodynamic therapy (mean 9.5 injections, individual aflibercept, ranibizumab, bevacizumab or injections of three agents together)
- 2) RPE tear
- 3) Large submacular hemorrhage
- 4) Subfoveal fibrosis without atrophy of the outer retinal layers on spectral domain optical coherence tomography (SD-OCT) images and we have decided to include eyes with foveal external limiting membrane(ELM) that can only be detected in SD-OCT.

*Exclusion criteria for this study were;*

- 1) RPE–choroid translocation for diseases other than AMD and rhegmatogenous retinal detachment (RRD) with suprachoroidal hemorrhage (SCH)
- 2) Patients younger than 60 years
- 3) Follow-up of less than 6 months

Informed consent was obtained from participants after the risks and benefits of the surgery were fully explained.

Pre-operative examinations included complete ophthalmologic examinations which involve best corrected visual acuity(BCVA) were taken according to the early treatment of diabetic retinopathy study (ETDRS). Even more ophthalmologic examinations including dilated funduscopy, FFA, FAF and ICGA were performed.

Post-operative visits were performed at 1 st day, 1 st week, 1 st, 3rd and 6th month and at the final visit. The best-corrected ETDRS vision test and complete ophthalmic examinations were performed at each visit.

At the 1 st month, 3rd month, 6th month and final examination, color fundus pictures and optical coherence tomography (OCT) were performed by preferred fixation of the OCT-light. And at the same time, the viability of the graft was monitored by ICGA and FAF. Patients in the study were tested with a CLO (Heidelberg Retina Angiography [HRA; Engineering GmbH, Dossenheim, Germany) for FAF. The selected patients underwent FFA or ICGA to exclude the re-formation of the CNVM.

### 2.1. Surgical procedure

After that stimulation of the posterior vitreous detachment and a complete vitrectomy was performed. The CNVM was removed from the subretinal area through para-macular retinotomy with the subretinal forcers. At inferotemporal position of mid-peripheral retina, the full-thickness RPE / choroid was diathermyzed as a rectangular shape, and after removal of the diathermyzed patch, the vitreous scissors were used to cut a full thickness RPE / choroidal patch of approximately 1.5 to 2 mm diameter. With the use of a cannula, re-positioning of the patch under the macula was achieved through the present para-macular retinotomy.

We surrounded the region of the mid-peripheral retinotomy with laser photocoagulation and used the silicone oil(SiO) tamponade. (Fig. 1) In a second procedure, SiO was removed after about 3–6 months. Surgeries were performed by 23 and 25-gauge Constellation Vision System (Alcon Laboratories, Inc., Fort Worth, Texas, USA)for all participants.

### 2.2. Statistical analysis

Snellen visual acuity was converted to logarithm of the minimal angle of resolution (logMAR) for statistical analyses. Data were expressed as mean and range. Wilcoxon signed rank test was used for the comparisons of BCVA.  $P < 0.05$  was accepted to statistically significant.

## 3. Results

The mean age was  $73 \pm 7.17$  (range, 60–80 years) years and 4 of the patients were male, 4 of the patients were female. The mean follow-up time was  $14.12 \pm 8.16$  (range 7–30 months) months. The distributions of diagnoses were;

- 1) 4 patients with CNVM
- 2) AMD with massive subretinal hemorrhage in 3 cases
- 3) 1 case with suprachoroidal hemorrhage (SCH) + rhegmatogenous retinal detachment (RRD)

The clinical and demographic data of the patients are summarized in Table 1.

Although inferior proliferative vitreoretinopathy (PVR) was seen in 4 patients, the retina was re-attached with a second surgery. Subretinal hemorrhage developed on the postoperative 3rd day in a patient but spontaneously resolved during follow-up periods. Re-pars plana vitrectomy (PPV) was performed in one patient (Patient 3) due to inferior PVR at the postoperative 6th month. However total retinal detachment developed after 4 months, therefore, PPV has performed again and SiO was placed as tamponade. Fig. 2 shows the patient's FFA, FAF, color fundus photo and OCT images. Table 2 shows information about the complications. Fig. 3 shows the monitored of the RPE / choroid graft with ICGA images. There was a statistically significant increase in visual acuity in the postoperative 1 st and final visit compared to baseline,

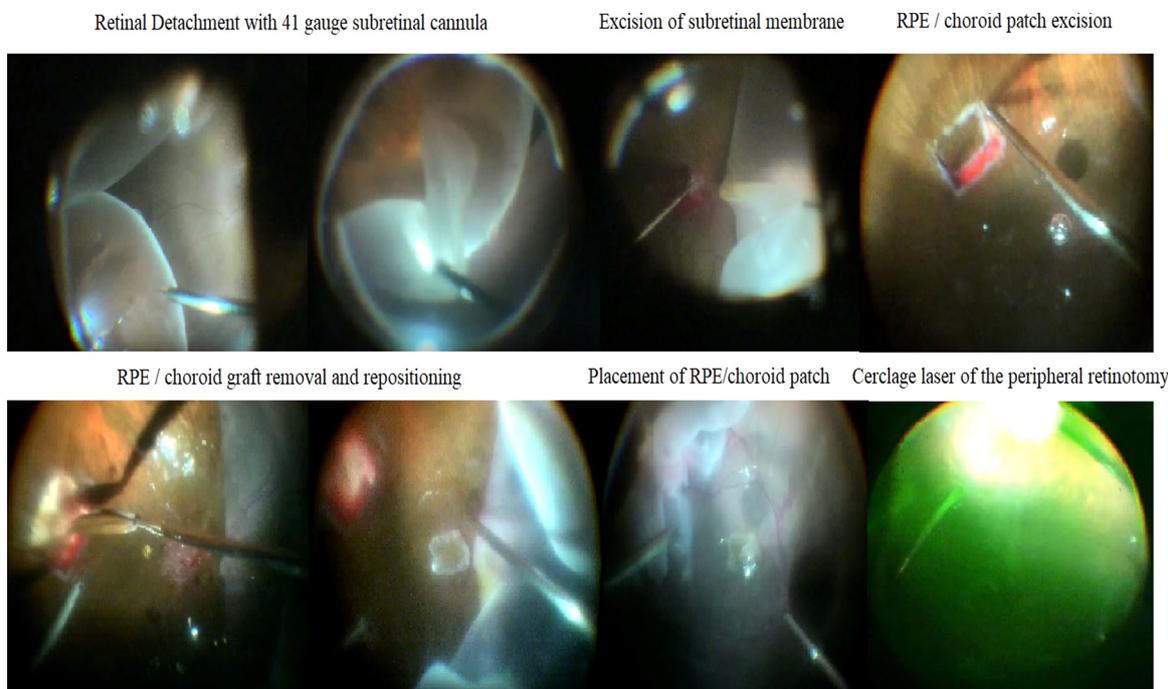


Fig. 1. Surgical procedure and stages are shown in figure.

respectively. ( $p = 0.034$  and  $p = 0.027$ )

Details of BCVA changes are shown in Table 3.

4. Discussion

A healthy RPE and choroidal patch in the submacular region of patients with AMD can lead to improvements of visual acuity, central fixation, and retina of the pigment epithelium. In FFA and ICGA, the presence of a choroid and choriocapillaris in the graft were evaluated in favor of perfusion. [15–21]

Surgical removal of CNVM in AMD patients always causes damage to the RPE in collaboration with bruch's membrane, the choriocapillaris complex, and subsequently can cause visual impairment. [18,22–24]

The regenerations or proliferations of RPE cells of the damaged area are ineffective or very small if present. [25,26] Therefore, macular rotation surgery is a promising approach to creating a viable lower surface tissue [27].

In a study conducted by Heussen et al., reported on the 1-year follow-up of 30 patients after translocation of a free graft of RPE and choroid. All participants included in the study had loss of vision due to AMD: 1 eye with a classic membrane, 1 eye with a mixed membrane, 1 eye with retinal angiomatous proliferation II lesion, 7 eyes with occult membranes, 13 eyes with pigment epithelial detachment, 2 eyes with

rupture of the RPE and 5 eyes with massive subretinal bleeding. They chose patients with unresponsive to therapy with large subfoveal lesions or poorly circumscribed lesions with no improvement. There was no change in BCVA between 6 and 12 months, but a deterioration of BCVA observed mostly within the first 3 months after surgery and was associated with postoperative complications. [28]

However, there is a risk of late CNV formation due to the peripheral growth of the graft resulting from the excision edges of the CNV. It can also be monitored with FFA and ICGA under favorable circumstances. [13]

In their series at 8 patients (26%), PVR due to vitreoretinal traction was observed. In the post-operative period, they experienced subretinal hemorrhages in 15 (50%) patients. This was the close observation as in the submacular surgery trial (SST) and the blood remained in the subretinal or sub-RPE space in 154 eyes (93%), and in 42 (25%) eyes with > 4 disc areas were found to presence of blood. [29]

In a recent study of Jan C. van Meurs et al., they investigated the efficacy of translocated autologous RPE cells and choroid after the removal of a subfoveal CNVM in patients with exudative AMD. 6 patients with subfoveal CNVM greater than 1 disc diameter were followed up for 7 to 13 months, and 5 patients had a subretinal hemorrhage. Baseline visual acuity ranged from 20/400 to 20/200. Fixation was seen at the patch region in 4 patients. Post-operative vision ranged from 20/200 to

Table 1

The clinical and demographic data of the patients.

| Patient | Age | Follow-up | Diagnosis | Preop.BCVA | Postop. 1 month BCVA | Postop. 3 months BCVA | Postop. 6 months BCVA | Final BCVA  | FAF | FFA/ICG |
|---------|-----|-----------|-----------|------------|----------------------|-----------------------|-----------------------|-------------|-----|---------|
| 1       | 80  | 15        | CNVM      | HM         | HM                   | HM                    | HM                    | HM          | (-) | (-)     |
| 2       | 75  | 14        | SRNVM     | HM         | 1 meter CF           | 1 meter CF            | 1 meter CF            | 2 meters CF | (+) | (+)     |
| 3       | 73  | 9         | CNVM      | HM         | 1 meter CF           | HM                    | HM                    | 1 meter CF  | (+) | (-)     |
| 4       | 60  | 6         | SRNVM     | HM         | 0.05                 | 0.05                  | 0.05                  | 0.05        | (+) | (+)     |
| 5       | 70  | 8         | CNVM      | HM         | 1 meter CF           | 1 meter CF            | HM                    | 1 meter CF  | (+) | (-)     |
| 6       | 69  | 6         | SRNVM     | HM         | 1 meter CF           | HM                    | HM                    | HM          | (-) | (-)     |
| 7       | 79  | 7         | CNVM      | HM         | HM                   | HM                    | 1 meter CF            | 0.1         | (+) | (+)     |
| 8       | 78  | 8         | SCH + RRD | LP+        | LP+                  | HM                    | 0.15                  | 0.15        | (+) | (+)     |

CNVM, Choroidal neovascular membrane; SRNVM, Subretinal neovascular membrane; SCH + RRD, Suprachoroidal hemorrhage + Rhegmatogenous retinal detachment; BCVA, Best corrected visual acuity; LP+, Light perception; CF, counting finger; HM, Hand motion; FAF, Fundus autofluorescence; FFA, Fundus fluorescein angiography; ICG, Indocyanine green angiography.

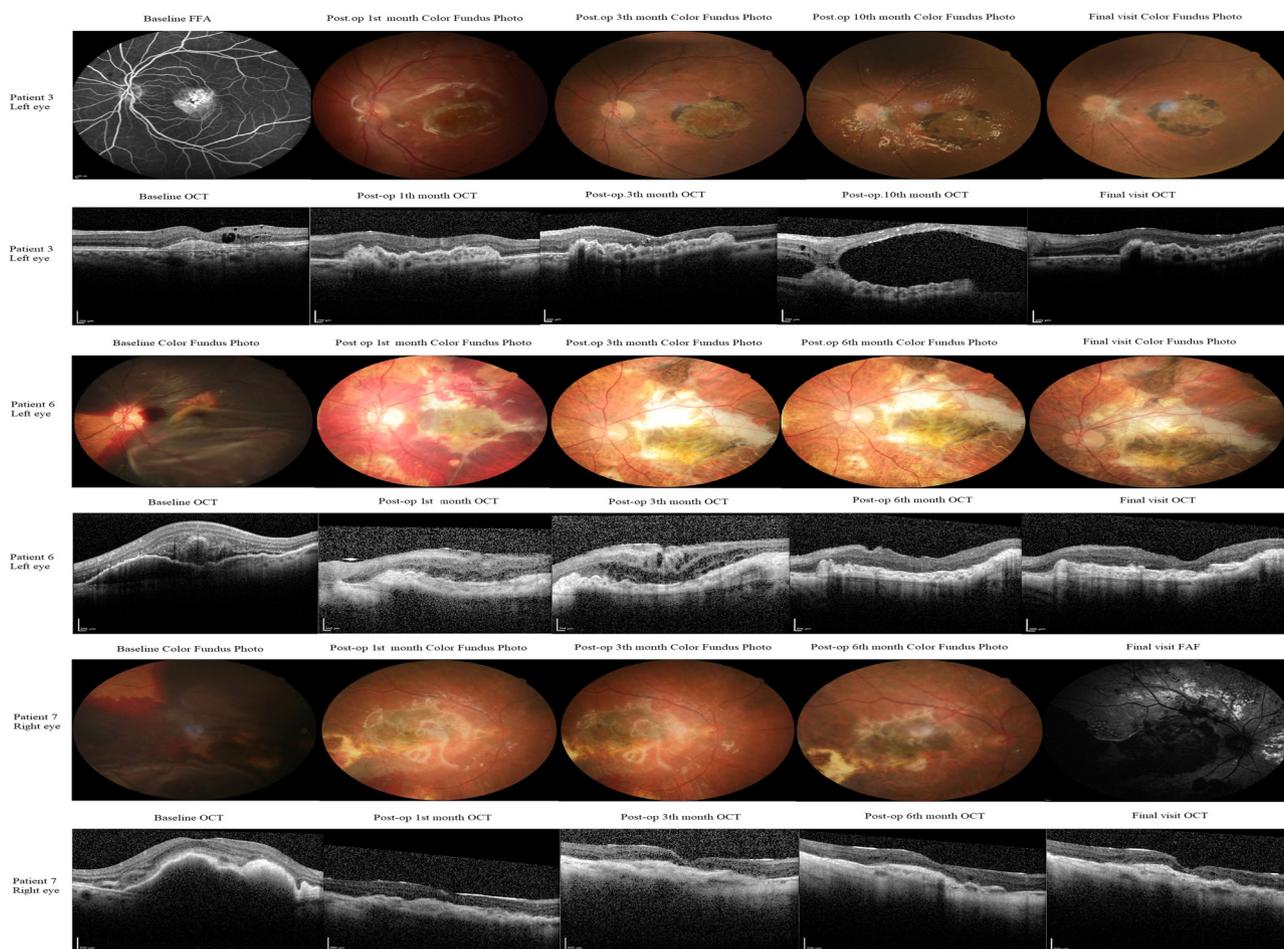


Fig. 2. Fundus images during follow-up periods.

Table 2  
Details about the complications.

| Patient | Complications                        | Last examination                   |
|---------|--------------------------------------|------------------------------------|
| 1       | none                                 | Re-attached                        |
| 2       | Inferior PVR(localized)              | Re-attached                        |
| 3       | 1-Inferior PVR(PPV) 2-Total RD (PPV) | Re-attached                        |
| 4       | none                                 | Re-attached                        |
| 5       | Inferior PVR(localized)              | Re-attached                        |
| 6       | subretinal hemorrhage post-op. 3     | spontaneous resolution-Re-attached |
| 7       | none                                 | Re-attached                        |
| 8       | Inferior PVR(localized)              | Re-attached                        |

PVR, Proliferative vitreoretinopathy; RD, Retinal detachment; PPV, pars plana vitrectomy.

20/64, and 3 patients had a 2-line increase. 4 patients had a normal RPE auto-fluorescence was seen on the patch and they used FFA and ICGA to monitor the formation of new re-growth of CNVM as well as to observe the viability of the graft. [15]

In a study by Saskia et al., they investigated the long-term results of autologous RPE and choroid translocation with a retinotomy for exudative AMD. A total of 81 patients participated in the study and the mean follow-up time was 38 months. BCVA increased from 1.30 logMAR (20/400) to 0.90 logMAR (20/160) one year after surgery. Severe complications such as submacular hemorrhage (n = 8, 10%), macular hole (n = 6, 7%) and PVR (n = 3, 4%) were observed in patients. [30]

A high incidence of sight-threatening complications due to a tilted

image and PVR at successful cases were the inconveniences of this technique. [14]

In our series at 4 patients (50%), PVR with inferior vitreoretinal traction was observed. PPV was performed in one patient (Patient 3) due to inferior PVR at the postoperative 6th month. However total retinal detachment developed after 4 months, therefore, PPV has performed again and SiO was placed as tamponade. In one patient(patient 5), inferior localized PVR was present at the 3rd month, membrane peeling + SiO exchange was performed. The vision increased during the first few months, followed by rapidly worsening fibrosis and serous macular detachment due to increased accumulation of submacular fluid towards the 70th day eventually BCVA decreased. In an other patient (patient 6) had bleeding around the graft on OCT at postoperative 4th day and increasing intraretinal fluid was observed at the end of the 2nd month.

19% of PVR has been reported after full macular translocation surgery. [13] The reason for our high rate of PVR was wound-healing response of the choroidectomy close to the vitreous base. A more precise shaving of the vitreous base and complete laser cerclage around the flap removal zone could have been reduced the PVR ratio.

Post-operative subretinal hemorrhage was encountered in 1 (12.5%) patient and it also resolved spontaneously. Although post-operative subretinal hemorrhage was related to an increased likelihood of sub-retinal fibrosis in our study, no correlation with CNV recurrence.

Autofluorescence was reflected in 6 patients and, consequently, the presence of lipofuscin activity was evaluated as viability of the graft. In terms of ICGA and FAF, ICGA was positive in 4 patients and matched with FAF, but choroidal perfusion could not be observed in ICGA despite lipofuscin activity seen in 2 patients at FAF. Fig. 3 shows FFA, ICG,

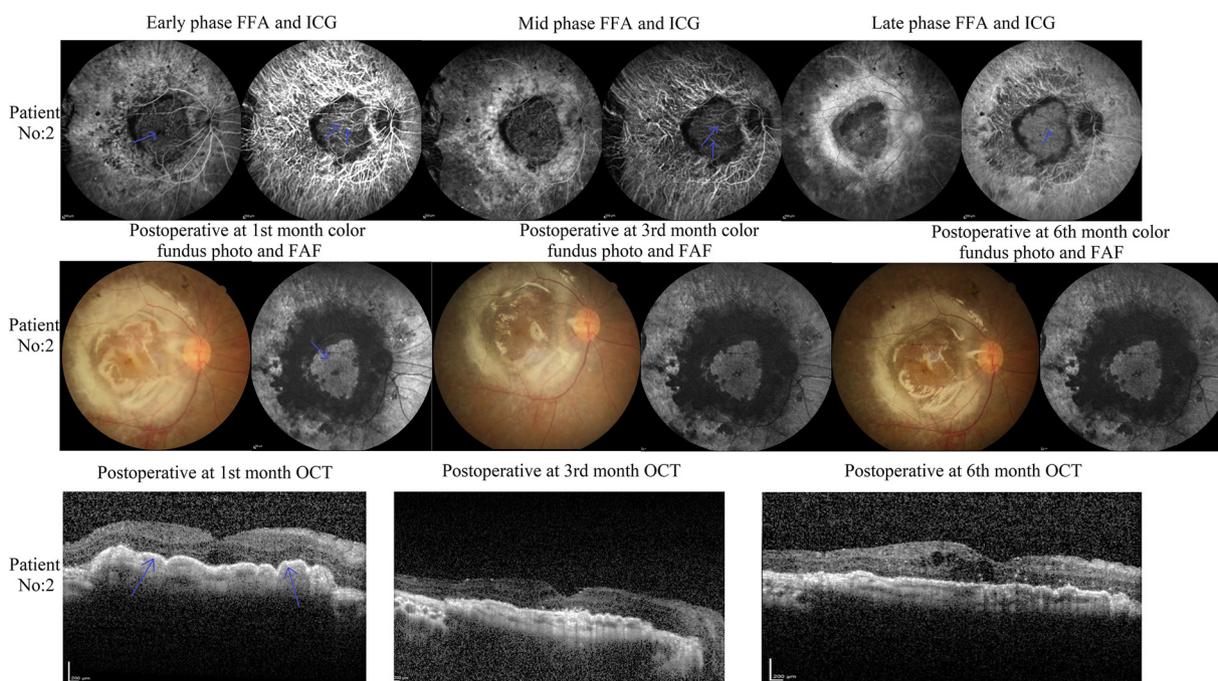


Fig. 3. Blue arrows show choroidal perfused vessels at ICGA, lipofuscin activity at FAF and choroidal patch at OCT images belonging to patient 2, respectively.

Table 3

Visual acuity changes during follow-up periods.

| Follow-up periods             | Mean ± SD-BCVA (log MAR) | Descriptions                           | p value |
|-------------------------------|--------------------------|--|---------|
| Baseline BCVA                 | 2.46 ± 1.04              |  |         |
| Post-operative 1st month BCVA | 2.16 ± 1.19              | Baseline-Post-operative 1st month BCVA | 0.034*  |
| Post-operative 3rd month BCVA | 1.89 ± 0.30              |  |         |
| Post-operative 6th month BCVA | 1.73 ± 0.46              |  |         |
| Final visit BCVA              | 1.52 ± 0.46              | Baseline-Final visit BCVA              | 0.027*  |

Wilcoxon Signed Ranks Test, \*p < 0.05.

BCVA, Best corrected visual acuity; SD, Standard deviation; log MAR; Logarithm of the Minimum Angle of Resolution.

BCVA values were calculated by converting log MAR into the all patients.

FAF, OCT images and color fundus photographs of patient number 2 according to postoperative follow-up periods. We attributed this to ongoing choroidal ischemia without affecting RPE and photoreceptors. This study had several limitations including retrospective nature, small sample size, single-centered, short follow-up time and lack of microperimetry for evaluating fixation. As there was no control group in our study, the results were more prone to bias and confounding.

In order to encouraging vitreoretinal surgeons about this surgery, the most important advantage of our study was that it could be performed using any vitrectomy methods such as a 23 or 25 gauge PPV system.

In conclusions, the BCVA was significantly increased in patients with AMD after RPE and choroidal translocation, and it was observed that long-term visual gain was possible and FAF, ICGA, and OCT have an important role in monitoring the viability of the graft after translocation of the choroid and RPE patch.

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**Informed consent**

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

**Declaration of Competing Interest**

Author Bugra Karasu declares that he has no conflict of interest. Author Gürkan Erdoğan declares that he has no conflict of interest.

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