Conbercept improves macular microcirculation and retinal blood supply in the treatment of nonischemic branch retinal vein occlusion macular edema

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Abstract
Purpose: To investigate the effect of conbercept on macular microvascular system and retinal blood supply in the treatment of nonischemic branch retinal vein occlusion macular edema (BRVO-ME).
Methods: Patients were divided into three groups: group A (containing 12 nonischemic BRVO-ME eyes), group B (containing contralateral 12 healthy eyes), and group C (containing 30 cataract eyes to obtain normal aqueous humor cytokine levels). Group A received monthly intravitreal injections of conbercept for 3 months. General data and best-corrected visual acuity (BCVA) were compared among the three groups. Optical coherence tomography angiography (OCTA) results (including central macular thickness [CMT], retinal vascular density and perfusion, and foveal avascular zone [FAZ]) at baseline were compared among groups A and B. Aqueous humor cytokine levels (including VEGF, IL-8, PDGF-AA, TNF-α, and ANGPTL-4) at baseline were compared between groups A and C. Moreover, BCVA, OCTA results, and aqueous humor cytokine levels of group A before and after conbercept treatment were compared.
Result: At baseline, group A had a significantly worse BCVA, lower retinal vascular density and perfusion, and numerically larger CMT and FAZ area comparing to the group B, and had a higher aqueous cytokine level (IL-8, VEGF, and ANGPTL-4) comparing to the group C (all ps < 0.05). After the injection of conbercept, group A presented a better BCVA (at initial diagnosis vs. after three conbercept injections: 1.16 ± 0.51 vs. 0.81 ± 0.30, logMAR, p < 0.05), higher retinal vascular density (11.56 ± 4.73 vs. 15.88 ± 2.31, mm⁻¹, p < 0.05) and perfusion (0.28 ± 0.12 vs. 0.39 ± 0.06, mm², p < 0.05), smaller CMT (504.92 ± 184.11 vs. 219.83 ± 46.63, mm², p < 0.05), as well as a lower levels of VEGF (before first injection vs. before third injection: 113.84 [70.81, 235.4] vs. 3.94 [3.56, 8.07], pg/ml, p < 0.05) and ANGPTL-4 (45,761 [7327.5, 81,402.5] vs. 25,015.5 [6690, 43,396], pg/ml, p < 0.05). However, the average FAZ area of group A expanded (at initial diagnosis vs. after three conbercept injections: 0.41 ± 0.14 vs. 0.62 ± 0.36, mm², p < 0.05).
1 | INTRODUCTION

Retinal vein occlusion (RVO) is a common retinal vascular disease with a high incidence, among which nonischemic branch retinal vein occlusion (BRVO) is more common in clinical situation.¹ The main reason of visual impairment in patients with BRVO is secondary macular edema (ME).² Significantly elevated intraocular vascular endothelial growth factor A (VEGF-A) level was found in patients with severe RVO-ME, leading to capillary leakage by disrupting tight binding proteins between vascular endothelial cells, which provides a clear pathological basis for anti-VEGF therapy.¹

By far, anti-VEGF therapy mainly includes two types of drugs: monoclonal antibodies (bevacizumab/Avastin, ranibizumab/Lucentis) and fusion proteins (afibercept, conbercept). The two types of drugs have been applied to the treatment of RVO-ME generally and have been proved capable of improving the patients’ vision.³,⁴ As China’s first self-developed ophthalmic anti-VEGF drug, conbercept is a novel soluble VEGF-capture-receptor fusion protein (VEGF-TRAP), which is formed by fusion of the second domain of VEGF receptor 1, the third and fourth domains of VEGF receptor 2, and the FC fragment of 100% humanized IgG. It has been reported that conbercept can mainly block VEGF-A, as well as VEGF-B and PLGF in a lesser degree.⁵ And the affinity of conbercept for VEGF-A is found to be much higher than Avastin and ranibizumab. Therefore, the clinical application of such high-affinity, multitarget fusion protein may be more advantageous than monoclonal antibody drugs.⁶ However, it has been reported that anti-VEGF therapy of Avastin has some side effects, such as impairing macular microcirculation and retinal blood supply and facilitating retinal vascular occlusion in RVO eyes.⁷ It is not entirely clear whether conbercept could cause these side effects.

In recent years, optical coherence tomography angiography (OCTA) has been widely used in the detection of macular blood flow state and macular microvascular changes in a noninvasive and rapid way.⁸ It has been shown that OCTA can be used to measure central macular thickness (CMT), retinal vascular density and perfusion, as well as foveal avascular zone (FAZ) area to effectively monitor and evaluate RVO-ME.⁹ Moreover, studies have shown that cytokines such as VEGF, interleukin 8 (IL-8), tumor necrosis factor α (TNF-α), platelet derived growth factor AA (PDGF-AA), and angiopoietin-like protein 4 (ANGPTL-4) are abnormally high in the aqueous humor of patients with RVO-ME, indicating that these cytokines are closely correlated with retinal ischemia. However, few studies have combined OCTA and aqueous humor cytokine levels to assess retinal blood supply.

The purpose of this study was to determine whether conbercept treatment of nonischemic BRVO-ME can induce retinal ischemia by combining OCTA quantitative parameters with aqueous humor cytokine levels and try to elucidate the underlying molecular mechanism, which is of great practical significance for clinical guidance of nonischemic BRVO-ME treatment and vision saving.

2 | MATERIALS AND METHODS

2.1 | Study design

The study was approved by the Ethics Committee of the First Affiliated Hospital of Hainan Medical University (No.: 2019/040) and adhered to the principles of the Declaration of Helsinki. Twelve patients with primary unilateral nonischemic BRVO-ME who were treated at our hospital between January 2019 and January 2020 were included in this clinical study. Group A included the 12 affected eyes of the nonischemic BRVO-ME patients, while group B contained the contralateral 12 healthy eyes served as self-control representing the normal standard of OCTA parameters. In addition, 30 cataract patients (30 eyes) without other eye diseases were randomly selected as group C and performed aqueous sample extraction during cataract surgery, so as to obtain the level of related cytokines in the normal population.

2.2 | Patients

The inclusion criteria were designed as follows: monocular onset patients who had clinical features of nonischemic BRVO-ME based on acknowledged diagnostic procedure references and having received no treatment measures with best-corrected visual acuity (BCVA, converted to the logarithmic expression of the minimum resolution angle [logMAR]) <0.5, fluorescein fundus angiography (FFA) showing dilated and tortuous branch retinal veins with strong fluorescein leakage, and retinal capillary nonperfusion area less than five optic disc areas, OCT showing increased CMT (≥250μm) and cyst-like macular lutea on initial diagnosis and indications of anti-VEGF therapy. Patients were not considered for this study if they had the following characteristics: patients with another ocular disorder or
ocular surgery history, including diabetic retinopathy, uveitis, glaucoma, retinal detachment, and ocular trauma; patients with serious systemic disorder, such as cardiovascular disease, diabetes, and immune system disease; patients who had undergone retinal laser photocoagulation or anti-VEGF therapy; or patients unable to cooperate with the ophthalmological examinations.

2.3 Patient assignment

Group A received three routine intravitreal injections of conbercept after diagnosis of nonischemic BRVO-ME (0, 1, 2 months after diagnosis; conbercept, Chengdu Kanghong Biological Technology Co., Ltd.; 0.5 mg/0.05 ml each time) and underwent a comprehensive ophthalmic examination at four certain time points (at initial diagnosis, after first, second, third intravitreal conbercept injection), including BCVA, slit-lamp examination (SL-3G, Topcon), fundus photography, FFA (Topcon, TRC-50DX), and OCTA (Carl Zeiss, HD-OCT 5000). Furthermore, aqueous samples of group A were obtained when conbercept injection was performed to detect related cytokine levels (VEGF, IL-8, PDGF-AA, TNF-α, ANGPTL-4) by Luminex (Luminex X-200, Thermo Fisher Scientific). Group B underwent ophthalmic examinations without aqueous humor extraction. Group C only recorded the ophthalmic examination results at initial diagnosis and underwent aqueous humor extraction during cataract operation.

2.4 OCTA and image analysis

In this study, the same examiner used CIRRUS HD-OCT5000 (Carl Zeiss) to examine the fundus of all subjects in a quiet and dark environment. After the pupil was fully dilated, OCTA was performed. First, 512 × 218 scan was performed, followed by 6 × 6 mm scan using angiography mode with macular fovea as the center. Clear images with high signal index were saved.

The thickness of macular fovea from inner limiting membrane (ILM) to retinal pigment epithelium (RPE) was obtained from 512 × 218 scanning images, which is regarded as the CMT. Moreover, OCTA angiography mode was used to automatically analyze and quantify the FAZ area and superficial retinal vascular density and perfusion on 6 × 6 mm images (by automated segmentation selecting area between ILM and external boundary of ganglion cell layer). We showed an example of the application of OCTA technology (see Appendix S1).

2.5 Aqueous humor extraction and intravitreal injection

These procedures are performed in the operating room by the same ophthalmologist with a sharp 30-gauge needle. After surface anesthesia, the needle was inserted 1 mm inside the limbal of the temporal cornea into the anterior chamber to extract 0.1 ml aqueous humor sample, which was immediately stored in a −80°C environment. Then the needle was inserted into the vitreous cavity through the pars plana (4 mm posterior from the limbus) and 0.05 ml of solution containing 0.5 mg conbercept was injected.

2.6 Measurement of aqueous humor cytokines

The levels of VEGF, IL-8, TNF-α, PDGF-AA, and ANGPTL-4 in the aqueous humor sample were detected by Luminex (Luminex X-200, Thermo Fisher Scientific) following a given protocol.

2.7 Statistical analyses

All statistical analyses were carried out by using SPSS statistical software (SPSS, 25.0). First, the normality tests (Shapiro–Wilk method) were carried out for quantitative data. If the data conformed to the normal distribution, mean ± SD was used for data presentation. One-way ANOVA was applied to the comparison among the three groups, and LSD test was used for pairwise comparison afterward. Paired t test was used for the comparison among groups A and B. If the data did not conform to the normal distribution, M (P25, P75) was used for data presentation and Wilcoxon rank sum test was used for the comparison between groups. Qualitative data were represented by n (%), and comparison between groups was performed by chi-square test. Repeated measurement data acquired at different time points were compared using ANOVA for repeated measurement. Uncalibrated F bounds were used for data that met the “spherically symmetric” hypothesis, while Greenhouse–Geisser method was used for those that did not meet to correct the F boundary value. LSD test was used for pair comparison afterward. Generalized estimation equation (GEE) was used for repeated measurement data that did not conform to ANOVA for repeated measurement, and Bonferroni method was used to calibrate the significance level in paired comparison afterward. A p value of <0.05 was considered statistically significant.

3 RESULTS

3.1 Comparison of the general clinical data of study subjects in each group

There were no statistically significant differences in age or sex proportion among these three groups (p > 0.05), while BCVA were greater in group A than in groups B and C with a statistically significant difference (p < 0.05) (see Table 1).

3.2 Comparison of retinal blood flow parameters among groups A and B

Vascular density and blood perfusion were lower in group A than in group B (p < 0.05), while CMT and FAZ area of group A was larger.
than group B, and the differences were all statistically significant (all $p < 0.05$) (see Table 2).

### 3.3 | Comparison of BCVA and retinal blood flow parameters before and after conbercept treatment in group A

BCVA and CMT were greater in group A before the treatment than after the first, second, and third treatment with a statistically significant difference ($p < 0.05$). However, retinal vascular density, blood perfusion, and FAZ were lower in group A before the treatment than after the first, second, and third treatment with a statistically significant difference ($p < 0.05$) (see Table 3).

### 3.4 | Measurement of aqueous humor cytokines

1. Baseline levels of IL-8, VEGF, and ANGPTL-4 were higher in group A than in group C with a statistically significant difference ($p < 0.05$), while there was no statistically significant difference as for TNF-α and PDGF-AA levels ($p > 0.05$) (see Table 4).

2. VEGF and ANGPTL-4 levels were lower after conbercept treatment than before in group A, and the differences were statistically significant ($p < 0.05$). There was no statistically significant difference as for TNF-α, IL-8, or PDGF-AA levels ($p > 0.05$) (see Table 5 and Figure 1).

### TABLE 1 General clinical data of the three groups ($n$, mean±SD)

| Group | $n$ | Male (n) | Female (n) | BCVA (logMAR) |
|-------|----|---------|-----------|---------------|
| A     | 12 | 7       | 5         | 1.16±0.51     |
| B     | 12 | 7       | 5         | 0.22±0.44*    |
| C     | 30 | 15      | 15        | 0.02±0.04*    |

| $F_{x}^{2}$ | 0.004* | 0.372b | 57.199a |
| $p$        | 0.996  | 0.830  | <0.001  |

Abbreviation: BCVA; best-corrected visual acuity.

* $F$ value of one-way ANOVA.

b Chi-square value.

* Statistically significant difference compared to group A ($p < 0.05$).

### 4 | DISCUSSION

Vascular endothelial growth factor A was proved as a major mediator for macular edema secondary to RVO. Anti-VEGF therapy has become the first-line option for RVO-ME,\textsuperscript{1,10,11} As one of the mainstream ocular anti-VEGF agents, conbercept has been proved to achieve a very satisfactory therapeutic effect in the treatment of BRVO-ME.\textsuperscript{12,13}

In our study, 12 patients (12 eyes) with nonischemic BRVO-ME had lower mean BCVA, retinal vascular density, and blood perfusion, and larger CMT and FAZ area, compared with age- and sex-matched normal control eyes. These patients with BRVO-ME then received monthly intravitreal injection of conbercept for 3 months. The results showed that BCVA, retinal vascular density, and blood perfusion increased while CMT decreased significantly after conbercept injection indicating that macular edema was relieved, which confirmed the exact efficacy of conbercept in the treatment of non-ischemic BRVO-ME. These results are consistent with most previous studies.\textsuperscript{12,14–16}

With the widespread use of anti-VEGF agents, it has become a hot issue of clinical concern whether anti-VEGF therapy would induce or aggravate macular ischemia. As one of the indicators reflecting macular ischemia, the discussion on the changes of FAZ area before and after anti-VEGF therapy for RVO remains controversial so far. Winegarner et al. reported that there was no significant difference in FAZ area of RVO patients 12 months after anti-VEGF treatment, while other studies found that FAZ area expand after anti-VEGF drug injection in the treatment of macular edema.\textsuperscript{17–19} It should be noted that RVO itself can lead to FAZ enlargement when the vascular arch in macular area is affected, and FAZ can be further expanded as the disease progresses. In fact, many studies have shown that anti-VEGF therapy could help alleviate macular edema while aggravate macular ischemia simultaneously because of its antagonistic effect on VEGF. Vance et al. reported that intravitreal injection of ranibizumab in the treatment of RVO may cause decrease of retinal vascular diameter, resulting in retinal hemorrhage and infarction of the nerve fiber layer.\textsuperscript{20} Meanwhile, a case report of refractory diabetic macular edema treated with bevacizumab showed by FFA that FAZ gradually expanding from 0.69 mm to 1.26 mm and CMT decreased from the original 564 microns to 262 microns, while the vision of the patient dropped from 20/80 to 20/200. It suggested that intravitreal injection of bevacizumab might affect macular microvascular structure.

### TABLE 2 Comparison of retinal blood flow parameters among groups A and B ($n$, mean±SD)

| Group | $n$ | CMT ($\mu$m) | Vascular density (mm$^{-1}$) | Blood perfusion (mm$^3$) | FAZ (mm$^2$) |
|-------|----|-------------|-----------------------------|--------------------------|-------------|
| A     | 12 | 504.92 ± 184.11 | 11.56 ± 4.73 | 0.28 ± 0.12 | 0.41 ± 0.14 |
| B     | 12 | 236.67 ± 7.15* | 17.07 ± 1.83* | 0.40 ± 0.05* | 0.24 ± 0.06* |
| $t$   | 5.042* | 3.362* | 3.196* | 3.699* |
| $p$   | <0.001 | 0.006 | 0.009 | 0.004 |

Abbreviations: CMT, central macular thickness; FAZ, foveal avascular zone.

* Paired t test.

* Statistically significant difference compared to group A ($p < 0.05$).
improved from 0.5 to 0.1, and FAZ area gradually expanded from 0.48 to 0.65 mm. CMT decreased from 623 to 173.

BRVO was also treated with 3 times of conbercept injection. Her injection, this CMT decreased from 643 to 249 (see Appendix S1) with left eye BRVO. After 3 times of conbercept injection, clonal antibodies. In our study, there was a 79-year-old male patient (see Appendix S1) with left eye BRVO. After 3 times of conbercept injection, his CMT decreased from 643 to 249 μm, logMAR vision improved from 0.5 to 0.1, and FAZ area gradually expanded from 0.36 to 0.57 mm². Another 53-year-old female patient with right eye BRVO was also treated with 3 times of conbercept injection. Her CMT decreased from 623 to 173 μm, logMAR vision improved from 0.5 to 0.4, and FAZ area gradually expanded from 0.48 to 0.65 mm². Our results showed that despite the expansion of FAZ area, the patients' vision and CMT recovered after the treatment as well as retinal vascular density and blood perfusion. Thus, we believe it was the progression of RVO rather than conbercept injection that caused the vascular network to collapse finally resulting in FAZ expansion. That is, the treatment of nonischemic BRVO-ME with conbercept did not aggravate macular ischemia, but significantly improved the prognosis of patients.

In order to further demonstrate the effect of conbercept on macular ischemia, we detected the level of several kinds of relevant cytokines in the aqueous humor of BRVO-ME patients. Jung et al. reported that the expression of VEGF, IL-8, PDGF-AA, and TNF-α was significantly higher in aqueous humor of patients with ischemic RVO than nonischemic RVO, indicating that these cytokines are closely associated with retinal ischemia. Another study showed that hypoxia-induced factor (HIF-1) and ANGPTL-4 were highly expressed in hypoxic Muller cells, and HIF-1 played an important role in ischemic retinopathy by upregulating the expression of ANGPTL-4. Recent studies further demonstrated that the expression of ANGPTL-4 and VEGF in the aqueous humor of patients.

| Time points       | CMT (mm²)     | Vascular density (mm⁻¹) | Blood perfusion (mm²) | FAZ (mm²) | BCVA (logMAR) |
|-------------------|---------------|-------------------------|-----------------------|----------|---------------|
| At initial diagnosis | 504.92 ± 184.11 | 11.56 ± 4.73            | 0.28 ± 0.12           | 0.41 ± 0.14 | 1.16 ± 0.51 |
| After first injection | 252.58 ± 48.97* | 15.38 ± 2.57*           | 0.36 ± 0.08*          | 0.57 ± 0.21* | 0.87 ± 0.33* |
| After second injection | 254.25 ± 37.33* | 15.73 ± 2.10*           | 0.37 ± 0.05*          | 0.59 ± 0.35* | 0.86 ± 0.40* |
| After third injection | 219.83 ± 46.63* | 15.88 ± 2.31*           | 0.39 ± 0.06*          | 0.62 ± 0.36* | 0.81 ± 0.30* |

*p < 0.05.

TABLE 3 Comparison of BCVA and retinal blood flow parameters before and after conbercept treatment in group A (mean ± SD)

| Time points       | CMT (mm²)     | Vascular density (mm⁻¹) | Blood perfusion (mm²) | FAZ (mm²) | BCVA (logMAR) |
|-------------------|---------------|-------------------------|-----------------------|----------|---------------|
| Before first injection | 1.1 (1.03, 1.36) | 55.53 (15.63, 103.88)  | 113.84 (62.52, 345)   | 20.33 (16.74, 28.83) | 41.65 (10,898.25, 77,037.75) |
| Before second injection | 1.03 (1.03, 1.03) | 6.53 (3.98, 12.27)*    | 38.24 (23.59, 51.96)* | 17.13 (11.62, 19.86) | 2355 (1360.25, 2929)* |
| Before third injection | 1.03 (1.03, 1.22) | 43.36 ± 34.84          | 3.56 (3.56, 6.37)*    | 18.13 ± 5.55     | 29,874.5 (6261.75, 54,009)* |

*p < 0.05.

TABLE 4 Comparison of baseline aqueous cytokine level between groups A and C (M [P25, P75]) (pg/ml)

|                  | TNF-α         | IL-8          | VEGF          | PDGF-AA      | ANGPTL-4     |
|------------------|---------------|---------------|---------------|--------------|--------------|
| Group A          | 1.1 (1.03, 1.36) | 55.53 (15.63, 103.88) | 113.84 (62.52, 345) | 20.33 (16.74, 28.83) | 41.65 (10,898.25, 77,037.75) |
| Group C          | 1.03 (1.03, 1.03) | 6.53 (3.98, 12.27)* | 38.24 (23.59, 51.96)* | 17.13 (11.62, 19.86) | 2355 (1360.25, 2929)* |

*p < 0.05.

TABLE 5 Comparison of aqueous cytokine level before and after conbercept treatment in group A (mean ± SD, M [P25, P75]) (pg/ml)

|                  | TNF-α         | IL-8          | VEGF          | PDGF-AA      | ANGPTL-4     |
|------------------|---------------|---------------|---------------|--------------|--------------|
| Before first injection | 1.27 (1.03, 1.36) | 63.11 ± 51.66 | 113.84 (70.81, 235.4) | 23.03 ± 8.53 | 45,761 (7327.5, 81,402.5) |
| Before second injection | 1.03 (1.03, 1.36) | 46.61 ± 36.97 | 3.56 (3.56, 6.37)* | 18.13 ± 5.55 | 29,874.5 (6261.75, 54,009)* |
| Before third injection | 1.03 (1.03, 1.22) | 43.36 ± 34.84 | 3.94 (3.56, 8.07)* | 17.16 ± 4.48 | 25,015.5 (6690, 43,396)* |

*p < 0.05.

system and induce macular ischemia. With more binding sites, higher affinity, and even longer half-life comparing with monoclonal antibody anti-VEGF drugs, it is natural to suspect whether conbercept may induce more severe retinal vascular events than monoclonal antibodies. In our study, there was a 79-year-old male patient (see Appendix S1) with left eye BRVO. After 3 times of conbercept injection, his CMT decreased from 643 to 249 μm, logMAR vision improved from 0.5 to 0.1, and FAZ area gradually expanded from 0.36 to 0.57 mm². Another 53-year-old female patient with right eye BRVO was also treated with 3 times of conbercept injection. Her CMT decreased from 623 to 173 μm, logMAR vision improved from 0.5 to 0.4, and FAZ area gradually expanded from 0.48 to 0.65 mm². Our results showed that despite the expansion of FAZ area, the patients' vision and CMT recovered after the treatment as well as retinal vascular density and blood perfusion. Thus, we believe it was the progression of RVO rather than conbercept injection that caused
with severe nonproliferative and proliferative diabetic retinopathy and patients with BRVO-ME was significantly higher than that of the normal control group, suggesting that ANGPTL-4 can be used as a sensitive biomarker to predict retinal ischemia.\textsuperscript{24,25} Other studies found that ANGPTL-4 and VEGF had a synergistic effect to destroy the stability of retinal vascular barrier and ANGPTL-4 might induce vascular permeability independent of VEGFR2.\textsuperscript{26} Our results showed that the levels of IL-8, VEGF, and ANGPTL-4 in the aqueous humor of nonischemic BRVO-ME patients were higher than those in the healthy control group, which provided a theoretical basis for us to predict the retinal blood supply status by detecting the levels of VEGF and ANGPTL-4 in the aqueous humor of patients. Meanwhile, we found that the levels of VEGF and ANGPTL-4 in aqueous humor declined obviously after conbercept injection, which together with the results of BCVA and OCTA above further confirmed that conbercept could improve the blood supply in macular microvascular system rather than induce macular ischemia. In fact, the levels of VEGF and ANGPTL-4 both remained at a low level between the two injections (see Figure 1), indicating that the clinical medication interval can be analyzed by detecting the level of the cytokines, so as to guide the clinical medication. However, we noticed that there was no significant difference in the levels of IL-8, TNF-\(\alpha\), and PDGF-AA in pathogenetic or therapeutic process of BRVO-ME. This may be because all the cases in this study are elderly patients whose incidence of BRVO was mostly closely related to vascular sclerosis, while might have little significant correlation with inflammatory factors.

The main strength of this study is to demonstrate that intravitreal injection of conbercept improves macular microcirculation and increases retinal blood supply in the treatment of nonischemic BRVO-ME by combining visual acuity, OCTA parameters, and aqueous humor cytokine assay results. To our knowledge, there is no study that has combined the results of these examinations. Therefore, our study provides an innovative and applicable method to detect macular microcirculation and retinal blood supply in patients with nonischemic BRVO-ME. However, there were some deficiencies in this study. First, due to the COVID-19 pandemic and poor patient compliance, some patients failed to return to the clinic on time, which resulted in a relatively small sample size and a short observation time in this study. Second, since the clinical samples were all from our hospital and mainly from the elderly, our results may not be generalizable to the general population. Third, we only included patients with nonischemic BRVO-ME, so it remains to be determined whether the results could apply to the patients with ischemic BRVO-ME or central retinal vein occlusion. Therefore, it is necessary to conduct further studies with large sample size and long follow-up time to verify the conclusion.

**AUTHOR CONTRIBUTIONS**

All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Yikeng Huang, Minli Linghu, Weiwen Hu, and Xionggao Huang. The first draft of the manuscript was written by Yikeng Huang, Minli Linghu, and Weiwen Hu. All authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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**CONFLICT OF INTEREST**

All authors report no conflicts of interest. The authors alone are responsible for the content and writing of this article.

**DATA AVAILABILITY STATEMENT**

Data that support the findings of this study are available from the corresponding author, Xionggao Huang, upon reasonable request.

**INFORMED CONSENT**

Informed consent was obtained from all individual participants included in the study.
CONSENT TO PUBLISH
All authors declared their consent to the publication of the manuscript.

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SUPPORTING INFORMATION
Additional supporting information can be found online in the Supporting Information section at the end of this article.

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