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Comparative effectiveness of three treatment strategies for submacular hemorrhage secondary to age-related macular degeneration

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Abstract

We aimed to compare the effectiveness of three treatment strategies for extensive submacular hemorrhage (SMH) secondary to age-related macular degeneration (AMD): intravitreal anti-vascular endothelial growth factor (VEGF) monotherapy, anti-VEGF combined with sulfur hexafluoride (SF_6) gas tamponade, and triple therapy consisting of anti-VEGF, SF_6 gas, and intravitreal tissue plasminogen activator (tPA).

This multicenter retrospective study included 61 eyes of 61 patients treated at three institutions. Patients received anti-VEGF monotherapy (n = 19), anti-VEGF plus SF_6 gas tamponade (n = 20), or triple therapy (n = 22). Inclusion criteria were logMAR best-corrected visual acuity (BCVA) ≥ 0.40, SMH ≥ 2 disc diameters, and treatment within 14 days of onset. The primary outcome was longitudinal BCVA change over 12 months, analyzed using a linear mixed-effects model adjusted for baseline BCVA.

A significant overall treatment effect was observed, with triple therapy demonstrating consistently superior BCVA. Unadjusted 12-month BCVA also differed significantly among groups. Treatment modality and baseline ellipsoid zone integrity were independent predictors of 12-month BCVA. Hematoma displacement rates were 100%, 35.0%, and 5.3%, and re-hemorrhage rates were 18.2%, 55.0%, and 42.1% in the triple, combination, and monotherapy groups, respectively.

Triple therapy provided superior and more predictable visual and anatomical outcomes, supporting early intervention.

Introduction

Submacular hemorrhage (SMH) is a vision-threatening complication of neovascular age-related macular degeneration (AMD), occurring in approximately 10–20% of affected individuals [1, 2]. The accumulation of blood between the retinal pigment epithelium (RPE) and photoreceptors can lead to rapid and potentially irreversible visual loss through multiple mechanisms, including mechanical separation of photoreceptors from the RPE, iron-mediated toxicity from hemoglobin degradation products, and tractional damage caused by fibrin clot contraction [3, 4]. Without timely and appropriate intervention, SMH frequently results in severe and permanent visual impairment, with final visual acuity often worse than 20/200 [5, 6].

Several therapeutic strategies have been proposed for SMH management, including observation, intravitreal anti-vascular endothelial growth factor (VEGF) therapy, pneumatic displacement with intravitreal gas injection, intravitreal tissue plasminogen activator (tPA) to facilitate clot lysis, and pars plana vitrectomy with subretinal tPA administration [7–14]. Among minimally invasive approaches, the combination of intravitreal anti-VEGF agents, gas tamponade buoyant force and tPA has emerged as a promising outpatient treatment option that may achieve effective hematoma displacement without the need for vitrectomy [13, 14].

The rationale for combining these three modalities is multifactorial. Anti-VEGF agents target the underlying choroidal neovascularization and reduce the risk of recurrent hemorrhage [15]. Gas tamponade provides a buoyant force to facilitate the displacement of the liquefied hematoma away from the fovea [16]. tPA accelerates the conversion of plasminogen to plasmin, promoting fibrin clot degradation and enhancing hematoma liquefaction and displacement [17, 18]. Despite these theoretical advantages, direct comparative studies evaluating anti-VEGF monotherapy, anti-VEGF combined with gas tamponade, and triple therapy with tPA remain limited.

The efficacy of gas tamponade without exogenous tPA has been inconsistent across studies [19–21]. This variability may be explained by the temporal dynamics of endogenous fibrinolysis. Following vascular injury and clot formation, endogenous tPA is gradually released from damaged tissues and vascular endothelium, facilitating physiological clot dissolution over several days [22, 23]. However, this process is relatively slow and time-dependent. Endogenous tPA activity typically peaks approximately 1 week after hemorrhage onset, after which progressive clot organization and fibroblast infiltration reduce the likelihood of successful displacement [22, 24]. Consequently, the effectiveness of gas tamponade alone depends on precise timing within a narrow therapeutic window, leading to unpredictable clinical outcomes.

In contrast, exogenous tPA administration allows for active control of the fibrinolytic process. By directly supplying tPA, the conversion of intra-hematoma plasminogen to plasmin can be reliably induced regardless of the timing of endogenous tPA release, provided that plasminogen remains present within the clot—typically within 14 days of hemorrhage onset [22, 25]. Therefore, this active approach is expected to achieve more consistent hematoma liquefaction and displacement, particularly in patients presenting beyond the optimal 1-week window for endogenous fibrinolysis.

We aimed to compare the effectiveness of three treatment strategies—anti-VEGF monotherapy, anti-VEGF combined with gas tamponade, and triple therapy with anti-VEGF, gas tamponade, and intravitreal tPA—in patients with extensive SMH secondary to AMD. We hypothesized that triple therapy would result in superior visual outcomes and higher hematoma displacement rates than the other strategies, and these benefits would be observed even in patients with poor baseline visual acuity. To our knowledge, this study represents the first direct comparison of all three treatment approaches within a single multicenter patient cohort.

Materials and Methods

Study design and setting

This multicenter, retrospective, comparative study was conducted at three tertiary referral centers in Japan: Kansai Medical University Hospital, Kansai Medical University Medical Center, and Kobe University Hospital. The study protocol adhered to the tenets of the Declaration of Helsinki and was approved by the institutional review boards of all the participating institutions (approval number

#2025003). Given the retrospective design and use of de-identified medical records, the requirement for written informed consent was waived by the ethics committee.

Patient selection

Medical records of all patients diagnosed with SMH secondary to AMD and treated between April 1, 2010, and March 31, 2024, were reviewed. Inclusion criteria were: (1) SMH secondary to neovascular AMD confirmed by fundus examination and optical coherence tomography (OCT); (2) logarithm of the minimum angle of resolution (logMAR) best-corrected visual acuity (BCVA) ≥ 0.40 at presentation, indicating moderate or worse vision loss; (3) SMH size ≥ 2 optic disc diameters, representing extensive hemorrhage; (4) treatment initiated within 14 days of hemorrhage onset; and (5) minimum follow-up of 12 months with complete data availability. Exclusion criteria were: (1) SMH resulting from causes other than AMD, such as trauma or retinal arterial macroaneurysm; (2) previous vitrectomy; (3) concurrent retinal detachment requiring surgical intervention; (4) significant media opacity preventing adequate fundus examination or OCT imaging; (5) history of submacular surgery; and (6) coexisting ocular conditions independently affecting visual acuity (e.g., advanced glaucoma or severe diabetic retinopathy).

Treatment groups and protocols

Patients were retrospectively assigned to one of three treatment groups based on the intervention received. Group A (anti-VEGF monotherapy, n = 19): Patients received an intravitreal anti-VEGF injection alone, without gas tamponade or tPA. The anti-VEGF agents administered were ranibizumab 0.5 mg (n = 2), aflibercept 2.0 mg (n = 13), or faricimab 6.0 mg (n = 4). The choice of agent was determined by the treating physician based on availability and clinical judgment. Group B (combined gas tamponade, n = 20): Patients received an intravitreal anti-VEGF injection combined with intravitreal gas injection (0.4 mL of sulfur hexafluoride [SF_6]), without tPA. Following the injection, patients were instructed to maintain a prone position for 7 days to facilitate gas-associated hematoma displacement. Group C (triple therapy, n = 22): Patients received a combined intravitreal injection of an anti-VEGF agent and tPA (40,000 IU of GRTPA®, Tanabe Pharma Co., Osaka, Japan), followed by intravitreal SF_6 gas injection (0.4 mL) on the next day. The injection sequence began with anti-VEGF, followed by tPA. Patients maintained a supine position for 1 hour to 1 day after the initial injection to allow clot lysis. After gas injection on the following day, patients maintained prone positioning for 7 days to promote hematoma displacement.

All intravitreal injections were performed under sterile conditions using standard preparations and topical anesthesia. Patients were evaluated at baseline, 1 week, and 1, 3, 6, and 12 months using comprehensive ophthalmologic assessments, including BCVA measurement, slit-lamp biomicroscopy, indirect ophthalmoscopy, and spectral-domain OCT.

Outcome measures

Primary outcome: The primary outcome was the longitudinal change in BCVA over 12 months, measured using a standard Landolt C chart and converted to logMAR units for statistical analysis.

Secondary outcomes:

1.**Unadjusted 12-month BCVA:**BCVA at 12 months post-treatment, measured with a standard Landolt C chart and converted to logMAR units. Group comparisons were performed without adjustment for baseline values.

2. **Hematoma displacement rates:** Complete displacement of the submacular hematoma away from the foveal center, evaluated on fundus examination and OCT at 1 month post-treatment by two independent masked graders.

3. **Re-hemorrhage rates:** Incidence of new or recurrent submacular bleeding during the 12-month follow-up period, documented by fundus photography and OCT.

4. **Anatomical outcomes:** Central retinal thickness (CRT), pigment epithelial detachment (PED) height, and central choroidal thickness (CCT) measured by OCT at 1 and 12 months.

5. **Ellipsoid zone (EZ) and external limiting membrane (ELM) integrity:** Assessed by OCT at baseline and 12 months as indicators of photoreceptor structural integrity.

6. **Prognostic factors for 12-month BCVA:**Variables associated with BCVA at 12 months were analyzed using multivariate regression models.

Data collection

The following data were extracted from medical records: patient demographics (age and sex); baseline ocular characteristics, including BCVA, intraocular pressure, and lens status; SMH characteristics (size in disc diameters and thickness on OCT); time from hemorrhage onset to treatment; OCT parameters (CRT, PED height, CCT, EZ, and ELM integrity); systemic factors, such as anticoagulant or antiplatelet medication use; treatment details; and all outcome measures at each follow-up visit.

All OCT examinations were performed at the three institutions using spectral-domain OCT systems. CRT was measured on horizontal B-scan cross-line images centered on the fovea and defined as the vertical distance from the internal limiting membrane to the RPE at the foveal center. The EZ and ELM integrity were assessed on the same B-scan images, with disruption defined as any discontinuity or absence of the corresponding hyperreflective bands within the central 1-mm diameter zone. Two independent masked graders evaluated outer retinal integrity at all time points, and any discrepancies were adjudicated by a senior grader.

Statistical analysis

All statistical analyses were performed using JMP software version 17 (SAS Inc., Cary, NC, USA). Continuous variables are presented as mean ± standard deviation for normally distributed data or median with interquartile range (IQR) for non-normally distributed data. Normality was assessed using the Shapiro–Wilk test. Categorical variables are expressed as frequencies and percentages.

Baseline characteristics were compared among the three groups using the Kruskal–Wallis test for continuous variables and the chi-square or Fisher's exact test for categorical variables, as appropriate.

For the primary analysis of longitudinal changes in BCVA, a linear mixed-effects model was employed to account for repeated measures, with baseline BCVA included as a covariate. Fixed effects included treatment group, time, and the treatment-by-time interaction, and a random intercept was incorporated for each patient to account for within-subject correlation. Post-hoc pairwise comparisons at 12 months were performed using Tukey's honest significant difference (HSD) test.

Hematoma displacement and re-hemorrhage rates were compared using the chi-square test.

To identify independent predictors of 12-month BCVA, multivariate linear regression analysis was conducted, including treatment group, baseline BCVA, time to treatment, hemorrhage size, hemorrhage thickness, and baseline EZ integrity.

Statistical significance was set at $P < 0.05$. Appropriate adjustments for multiple comparisons, including Bonferroni correction or Tukey's HSD, were applied where indicated.

Results

Patient characteristics

A total of 61 eyes from 61 patients met the inclusion criteria and were included in the analysis. Group A comprised 19 patients (anti-VEGF monotherapy); Group B included 20 patients (combined SF_6 gas tamponade); and Group C included 22 patients (triple therapy). Patient demographics and baseline characteristics are summarized in Table 1.

The mean age was similar across groups, ranging from 74.7 to 77.2 years ($P = 0.209$). Male predominance was observed in all groups (54.6–73.7%), with no significant differences among groups ($P = 0.388$). Baseline logMAR BCVA was 0.86 ± 0.34 in Group A, 1.02 ± 0.39 in Group B, and 1.06 ± 0.34 in Group C, with no statistically significant difference ($P = 0.128$).

Other baseline characteristics—including cataract presence (50.0–63.6%, $P = 0.670$), anticoagulant use (31.6–45.5%, $P = 0.630$), SMH size ($P = 0.108$), SMH thickness ($P = 0.420$), preoperative CRT ($P = 0.770$), PED height ($P = 0.455$), CCT ($P = 0.102$), EZ integrity ($P = 0.282$), and ELM integrity ($P = 0.398$)—were all comparable among the three groups.

However, a significant difference was observed in the interval from hemorrhage onset to treatment initiation. Group A had the longest delay (10.2 ± 4.2 days), Group B had the shortest (3.4 ± 3.4 days), and Group C was intermediate (5.5 ± 4.3 days) ($P < 0.001$).

Visual acuity outcomes

Baseline-adjusted visual acuity: A linear mixed-effects model adjusted for baseline BCVA demonstrated a significant overall treatment effect over 12 months ($P = 0.009$) and a significant effect of time ($P < 0.001$), with no significant treatment-by-time interaction ($P = 0.510$), indicating consistent treatment effects throughout the follow-up period (Fig. 1).

The estimated baseline-adjusted BCVA at 12 months (least-squares means ± standard error) was:

Group A: 1.01 ± 0.13

Group B: 0.93 ± 0.13

Group C: 0.45 ± 0.12

Post-hoc pairwise comparisons using Tukey's HSD test revealed:

Group C vs. Group A: Difference = 0.52 logMAR (95% CI: 0.10–0.94), $P = 0.011$

Group C vs. Group B: Difference = 0.40 logMAR (95% CI: 0.00–0.80), $P = 0.049$

Group A vs. Group B: Difference = –0.12 logMAR (95% CI: –0.54–0.31), $P = 0.784$

These findings indicate that triple therapy (Group C) was associated with consistently superior visual acuity over the 12 months than that of anti-VEGF monotherapy and SF_6 gas tamponade alone, with between-group differences of approximately 0.4–0.5 logMAR units.

Unadjusted visual acuity: At 12 months, unadjusted logMAR BCVA was 0.95 ± 0.50 in Group A, 0.96 ± 0.74 in Group B, and 0.49 ± 0.28 in Group C (Fig. 2). Group C demonstrated significantly better visual outcomes than those of both Groups A and B ($P = 0.011$).

Hematoma displacement rates

Complete hematoma displacement at 1 month was achieved in 1 of 19 eyes (5.3%) in Group A, 7 of 20 eyes (35.0%) in Group B, and all 22 eyes (100%) in Group C.

The differences among groups were highly significant ($P < 0.001$, chi-square test) (Fig. 3**a**).

Re-hemorrhage rates

During the 12-month follow-up period, re-hemorrhage occurred in 8 of 19 eyes (42.1%) in Group A, 11 of 20 eyes (55.0%) in Group B, and 4 of 22 eyes (18.2%) in Group C.

The differences among groups were statistically significant (P = 0.037, chi-square test) (Fig. 3**b**), with Group C demonstrating significantly lower re-hemorrhage rates than those of the other treatment groups.

Anatomical outcomes

At 12 months, there were no significant differences among the three groups in CRT (P = 0.114), PED height (P = 0.316), or CCT (P = 0.334).

These findings suggest that while triple therapy achieved superior functional outcomes and hematoma displacement, the final anatomical parameters were largely comparable across groups, likely reflecting the common endpoint of macular neovascularization (MNV) stabilization with anti-VEGF therapy.

Outer retinal structure

EZ and ELM integrity were assessed at baseline and 12 months. At baseline, EZ and ELM disruption was observed in 89.5% and 47.4% of eyes in Group A, 80.0% and 60.0% in Group B, and 95.5% and 68.2% in Group C (P = 0.282 and P = 0.398, respectively). At 12 months, disruption rates were 94.7% and 63.2% in Group A, 85.0% and 60.0% in Group B, and 72.7% and 45.5% in Group C, respectively (P = 0.142 and P = 0.469). Although Group C showed a trend toward less progression of outer retinal disruption, these differences did not reach statistical significance.

Prognostic factors for 12-month BCVA

Multivariate linear regression analysis was performed to identify independent predictors of 12-month BCVA (Table 2a). Significant predictors included:

1. Treatment modality (Type test, P = 0.001), with triple therapy associated with superior visual outcomes.

2. Baseline EZ integrity (P = 0.015), with an intact EZ predicting better visual acuity.

Non-significant factors included baseline visual acuity (P = 0.094), age (P = 0.746), hemorrhage size (P = 0.215), hemorrhage thickness (P = 0.335), and time to treatment (P = 0.930). The persistence of treatment modality as a significant predictor, even after adjusting for baseline EZ integrity and other covariates, highlights the independent therapeutic benefits of triple therapy in patients with extensive SMH secondary to AMD.

Treatment modality and baseline EZ integrity were identified as significant predictors of 12-month BCVA. The overall effect of treatment modality—a multilevel categorical variable—was statistically significant in

the multivariate model (Type III test, P = 0.001). Baseline EZ integrity was also independently associated with 12-month BCVA (P = 0.015), with an intact EZ predicting better visual outcomes.

Because the treatment modality consisted of multiple categories, statistical significance was first evaluated using a Type III test within the multivariate model. Upon confirmation of a significant overall treatment effect, pairwise comparisons were conducted using Tukey's HSD test based on the estimated least-squares means to appropriately account for multiple comparisons. These post hoc analyses demonstrated that triple therapy was associated with significantly better 12-month BCVA than that of the other treatment modalities (Table 2b).

In contrast, baseline visual acuity (P = 0.094), age (P = 0.746), SMH size (P = 0.215), SMH thickness (P = 0.335), and time to treatment (P = 0.930) were not significant predictors of 12-month BCVA. When hematoma displacement was compared between patients treated within 7 days of symptom onset and those treated between 8 and 14 days, Group C achieved a 100% displacement rate in both time intervals. Although hematoma displacement was observed in a subset of patients in Group B, the displacement rate remained significantly lower than that in Group C (Fig. 4). Representative clinical courses from each treatment group, including pre- and post-treatment fundus photographs and OCT images, are presented in Fig. 5.

Discussion

This multicenter retrospective study provided the first direct comparison of three treatment strategies for extensive SMH secondary to AMD: anti-VEGF monotherapy, combined gas tamponade, and triple therapy incorporating anti-VEGF, gas tamponade, and tPA. Our results demonstrate that triple therapy was associated with consistently better visual acuity outcomes over 12 months than those of the other treatment groups ($P < 0.05$). Furthermore, triple therapy achieved a 100.0% hematoma displacement rate (22/22 eyes), which was approximately 20-fold higher than that observed with anti-VEGF monotherapy (5.3%) and nearly 3-fold higher than that achieved with combined gas tamponade (35.0%). These findings support triple therapy as a highly effective treatment strategy for extensive SMH, particularly in patients presenting with poor baseline visual acuity. Previous studies have generally compared only two treatment modalities or evaluated a single approach [7–14]. By directly comparing three commonly employed conservative strategies, our study provides clinically relevant, evidence-based guidance to inform treatment selection. Notably, triple therapy demonstrated superior outcomes even among patients with poor baseline visual acuity (logMAR BCVA ≥ 0.40). All patients in our cohort presented with moderate or severe visual impairment, representing a clinically challenging population. The significant visual improvement observed with triple therapy in this subgroup suggests that meaningful functional recovery is achievable even in cases of advanced visual loss. This is particularly relevant for clinical decision-making, as patients with poor baseline vision are often considered to have limited potential for visual improvement.

The benefit observed in patients with poor baseline vision likely reflects the ability of triple therapy to achieve rapid and complete hematoma displacement, thereby minimizing the duration of photoreceptor exposure to toxic blood components. As demonstrated by our analysis of baseline EZ integrity (P = 0.015), photoreceptor structural integrity is a critical determinant of visual outcomes. Therefore, prompt and effective hematoma displacement through triple therapy may help preserve photoreceptor structure and function, even in eyes presenting with severely reduced visual acuity.

A key contribution of our study is the clarification of why gas tamponade alone often yields inconsistent results, whereas triple therapy with tPA produces more reliable outcomes. This difference can be explained by the temporal dynamics of clot formation, fibrinolysis, and organization, as described in physiological studies of hemostasis and fibrinolysis [22–25]. Following vascular rupture, the hemostatic process proceeds through well-defined stages. In the immediate phase (0–60 minutes), vasoconstriction, platelet plug formation, and fibrin clot development occur rapidly. Clot retraction is typically completed within 20–60 minutes, resulting in the formation of a dense fibrin meshwork that is relatively resistant to mechanical displacement [22, 23]. During the early fibrinolytic phase (1–3 days), damaged tissue and vascular endothelial cells gradually release endogenous tPA, which converts plasminogen trapped within the clot to plasmin. Plasmin then degrades the fibrin meshwork, promoting clot liquefaction. Endogenous fibrinolytic activity generally peaks 1–3 days after hemorrhage [22, 24, 25]. However, this window is temporally limited. In the subsequent organization phase (1–2 weeks), fibroblasts infiltrate the clot, produce collagen, and progressively transform the hematoma into fibrous tissue. As the organization advances, the clots become increasingly resistant to both enzymatic degradation and mechanical displacement [22, 24]. In the late phase (beyond 14 days), complete fibrotic organization occurs, with plasminogen largely consumed or inactivated, rendering hematoma displacement extremely difficult regardless of the treatment modality employed [22, 25].

The effectiveness of gas tamponade without exogenous tPA depends on the balance between fibrin, which provides structural integrity to the clot, and plasmin, which mediates fibrin degradation at the time of treatment. If gas injection is performed during the optimal window—when endogenous fibrinolytic activity is sufficiently active—the clot may be partially liquefied, thereby facilitating mechanical displacement. However, when treatment is administered too early (while the clot remains densely organized) or too late (after fibrotic organization has begun), displacement is likely to be incomplete or unsuccessful. This strong temporal dependence may explain the variable outcomes reported in the literature for gas tamponade alone [19–21], as well as the modest displacement rate of 35.0% observed in Group B. In this context, the success of gas tamponade without tPA is highly contingent on the timing of presentation relative to the endogenous fibrinolytic phase. Early or delayed intervention may substantially reduce treatment efficacy, resulting in inconsistent and less predictable clinical outcomes.

In contrast, the active administration of exogenous tPA (as performed in Group C) appears to overcome the intrinsic limitations of endogenous fibrinolysis. Exogenous tPA can reliably induce clot lysis independent of the timing of endogenous tPA release, provided that plasminogen remains present within the hematoma—a condition generally maintained within the first 14 days after hemorrhage [22, 25]. By

extending the effective therapeutic window, exogenous tPA reduces the reliance on the precise timing of intervention. Moreover, the administered tPA dose can be adjusted to ensure adequate plasmin generation, thereby promoting complete fibrin degradation and overcoming the slow and variable kinetics of endogenous tPA production [17, 18]. Through active modulation of the fibrinolytic process, triple therapy achieves consistent hematoma liquefaction and displacement, as reflected by the 100.0% displacement rate (22/22 eyes) observed in our cohort—nearly 3-fold higher than the 35.0% rate achieved with gas tamponade alone. This near-universal displacement suggests that exogenous tPA effectively mitigates the timing-dependent variability inherent to endogenous fibrinolysis. Furthermore, rapid clot dissolution shortens the duration of photoreceptor exposure to iron, hemosiderin, and other cytotoxic blood degradation products, which have been shown to induce irreversible retinal damage within 3–14 days [3, 4, 26].

This theoretical framework, supported by the fundamental principles of hemostasis and fibrinolysis described in Guyton and Hall's Textbook of Medical Physiology [22], provides a mechanistic rationale for the superior outcomes observed with triple therapy in our study. The 100.0% hematoma displacement rate achieved with triple therapy, compared with only 35.0% with gas tamponade alone, offers compelling clinical support for this physiological model.

Our findings have important implications for clinical practice. Triple therapy should be considered a first-line treatment for extensive SMH (≥ 2 disc diameters) in patients with moderate or worse visual impairment, particularly when treatment can be initiated within 14 days of hemorrhage onset. The superior visual outcomes, higher displacement rates, and lower re-hemorrhage rates observed with triple therapy support its use even in patients with poor baseline visual acuity. For patients presenting within the first week after hemorrhage—when endogenous fibrinolysis may still be active—combined gas tamponade without tPA could be considered as an alternative approach. However, given the inherent variability of endogenous fibrinolytic activity and the modest displacement rates observed in our cohort (35.0%), triple therapy with exogenous tPA remains the more reliable strategy, even during this early phase. In contrast, anti-VEGF monotherapy alone appears insufficient for the management of extensive SMH, achieving a displacement rate of only 5.3% and providing no clear visual advantage over combined gas tamponade. This approach may be more appropriately reserved for smaller hemorrhages (< 2 disc diameters) or for cases in which gas injection is contraindicated. The observation that time to treatment was not a significant predictor of visual outcome in the multivariate analysis (P = 0.463) suggests that triple therapy may remain effective even when administered beyond the optimal 1-week window, provided treatment is initiated within 14 days of hemorrhage onset. This finding is clinically relevant, as many patients present after a delay due to late symptom recognition or referral-related factors.

Our results align with and extend previous studies evaluating tPA-based strategies for SMH management. Haupert et al. [9] reported that pneumatic displacement combined with intravitreal tPA resulted in superior visual outcomes than those observed with observation alone. Similarly, Olivier et al. [27] demonstrated that pars plana vitrectomy with subretinal tPA injection can achieve favorable anatomical and functional outcomes; however, this approach necessitates invasive surgical intervention.

In contrast, our study demonstrates that a conservative, outpatient-based triple therapy regimen can achieve comparable or potentially superior outcomes without the need for vitrectomy. Although recent meta-analyses have suggested potential advantages of tPA-containing regimens [12, 28], robust direct comparative data remain limited. By directly comparing three commonly employed conservative treatment strategies, our study provides robust comparative evidence supporting the superiority of triple therapy over alternative non-surgical approaches.

This study has some limitations. First, its retrospective design and nonrandomized treatment allocation introduce the potential for selection bias. Notably, there was a significant difference in time to treatment among the groups ($P < 0.001$). However, multivariate analysis showed that the time to treatment was not an independent predictor of visual outcome ($P = 0.463$), suggesting that the treatment modality itself, rather than timing alone, was the principal determinant of the observed differences in outcomes. Second, the sample size was relatively modest ($n = 61$), limiting statistical power for subgroup analyses. Larger prospective randomized controlled trials are required to validate our findings and determine the optimal dosing strategies for both tPA and gas tamponade. Third, the follow-up period was restricted to 12 months. Long-term studies are necessary to assess whether the visual benefits of triple therapy are sustained beyond 1 year and to evaluate the incidence of late complications. Finally, the study population consisted exclusively of Japanese patients, which may limit the generalizability of the findings to other ethnic groups. Variations in choroidal thickness, AMD phenotypes, and genetic background across populations may influence treatment response and clinical outcomes.

In conclusion, this multicenter retrospective study demonstrated that triple therapy—combining anti-VEGF therapy, gas tamponade, and intravitreal tPA—achieved significantly superior visual outcomes and hematoma displacement rates than those of anti-VEGF monotherapy or combined gas tamponade alone in patients with extensive SMH secondary to AMD. At 12 months, baseline-adjusted visual acuity with triple therapy showed a clinically meaningful improvement of approximately 0.5 logMAR compared with the other treatment strategies (0.95–0.96 logMAR). The remarkable 100.0% hematoma displacement rate with triple therapy (22/22 eyes)—20-fold higher than that with anti-VEGF monotherapy (5.3%) and nearly 3-fold higher than that with gas tamponade alone (35.0%)—highlights the exceptional effectiveness and predictability achieved by actively controlling the fibrinolytic process with exogenous tPA. These outcomes are grounded in the temporal dynamics of clot lysis and organization. Gas tamponade alone depends on endogenous fibrinolysis, which is constrained to a narrow 1-week window after hemorrhage. In contrast, exogenous tPA provides reliable clot liquefaction regardless of endogenous tPA timing, provided treatment is initiated within 14 days while plasminogen remains present in the hematoma. Notably, triple therapy demonstrated efficacy even in patients with poor baseline visual acuity, suggesting that prompt and effective hematoma displacement preserves photoreceptor structure and function, even in eyes with initially severe vision loss.

Overall, these findings support triple therapy as the preferred first-line treatment for extensive SMH secondary to AMD, particularly in patients with moderate or worse vision loss presenting within 14 days of hemorrhage onset. By combining anti-VEGF to target choroidal neovascularization, tPA to ensure

reliable clot lysis, and gas tamponade to facilitate hematoma displacement, this approach maximizes visual recovery while minimizing the risk of re-hemorrhage.

Abbreviations

BCVA
best-corrected visual acuity
logMAR
logarithm of the minimum angle of resolution.

Declarations

Author contributions

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References

1. Scupola, A., Coscas, G., Soubrane, G. & Balestrazzi, E. Natural history of macular subretinal hemorrhage in age-related macular degeneration. *Ophthalmologica***213**, 97-102 (1999).
2. Avery, R. L., Fekrat, S., Hawkins, B. S. & Bressler, N. M. Natural history of subfoveal subretinal hemorrhage in age-related macular degeneration. *Retina***16**, 183-189 (1996).
3. Glatt, H. & Machemer, R. Experimental subretinal hemorrhage in rabbits. *Am. J. Ophthalmol.***94**, 762-773 (1982).
4. Toth, C. A., Morse, L. S., Hjelmeland, L. M. & Landers, M. B. 3rd. Fibrin directs early retinal damage after experimental subretinal hemorrhage. *Arch. Ophthalmol.***109**, 723-729 (1991).
5. Bennett, S. R., Folk, J. C., Blodi, C. F. & Klugman, M. Factors prognostic of visual outcome in patients with subretinal hemorrhage. *Am. J. Ophthalmol.***109**, 33-37 (1990).
6. Confalonieri, F. et al. Outcomes in the treatment of subretinal macular hemorrhage secondary to age-related macular degeneration: a systematic review. *J. Clin. Med.***13**, 367 (2024).
7. Hillenkamp, J., Surguch, V., Framme, C., Gabel, V. P. & Sachs, H. G. Management of submacular hemorrhage with intravitreal versus subretinal injection of recombinant tissue plasminogen activator. *Graefes Arch. Clin. Exp. Ophthalmol.***248**, 5-11 (2010).
8. Kamei, M. et al. Surgical removal of submacular hemorrhage using tissue plasminogen activator and perfluorocarbon liquid. *Am. J. Ophthalmol.***121**, 267-275 (1996).
9. Haupert, C. L. et al. Pars plana vitrectomy, subretinal injection of tissue plasminogen activator, and fluid-gas exchange for displacement of thick submacular hemorrhage in age-related macular degeneration. *Am. J. Ophthalmol.***131**, 208-215 (2001).
10. Chen, C. Y. et al. Management of submacular hemorrhage with intravitreal injection of tissue plasminogen activator and expansile gas. *Retina***27**, 321-328 (2007).
11. Stopa, M., Lincoff, A. & Lincoff, H. Analysis of forces acting upon submacular hemorrhage in pneumatic displacement. *Retina***27**, 370-374 (2007).
12. Stanescu-Segall, D., Balta, F. & Jackson, T. L. Submacular hemorrhage in neovascular age-related macular degeneration: A synthesis of the literature. *Surv. Ophthalmol.***61**, 18-32 (2016).
13. Guthoff, R., Guthoff, T., Meigen, T. & Goebel, W. Intravitreous injection of bevacizumab, tissue plasminogen activator, and gas in the treatment of submacular hemorrhage in age-related macular degeneration. *Retina***31**, 36-40 (2011).
14. Treumer, F., Klatt, C., Roider, J. & Hillenkamp, J. Subretinal coapplication of recombinant tissue plasminogen activator and bevacizumab for neovascular age-related macular degeneration with submacular haemorrhage. *Br. J. Ophthalmol.***94**, 48-53 (2010).

15. Brown, D. M. et al. Ranibizumab versus verteporfin for neovascular age-related macular degeneration. *N. Engl. J. Med.***355**, 1432-1444 (2006).
16. Ohji, M., Saito, Y., Hayashi, A., Lewis, J. M. & Tano, Y. Pneumatic displacement of subretinal hemorrhage without tissue plasminogen activator. *Arch. Ophthalmol.***116**, 1326-1332 (1998).
17. Collen, D. On the regulation and control of fibrinolysis. Edward Kowalski Memorial Lecture. *Thromb. Haemost.***43**, 77-89 (1980).
18. Rijken, D. C. & Lijnen, H. R. New insights into the molecular mechanisms of the fibrinolytic system. *J. Thromb. Haemost.***7**, 4-13 (2009).
19. Hattenbach, L. O., Klais, C., Koch, F. H. & Gümbel, H. O. Intravitreous injection of tissue plasminogen activator and gas in the treatment of submacular hemorrhage under various conditions. *Ophthalmology***108**, 1485-1492 (2001).
20. Murphy, G. S. P. et al. Tissue plasminogen activator or perfluoropropane for submacular hemorrhage in age-related macular degeneration: a factorial randomized clinical trial. *JAMA Ophthalmol.***142**, 1157-1164 (2024).
21. Ota, H., Takeuchi, J., Nonogaki, R., Tamura, K. & Kominami, T. Pneumatic displacement and anti-VEGF therapy for submacular hemorrhage in neovascular age-related macular degeneration: A retrospective study. *J. Clin. Med.***14**, 3154 (2025).
22. Guyton, A. C. & Hall, J. E. Hemostasis and blood coagulation in *Guyton and Hall textbook of medical physiology*. 13th ed. 475-486 (Elsevier, 2016).
23. Cesarman-Maus, G. & Hajjar, K. A. Molecular mechanisms of fibrinolysis. *Br. J. Haematol.***129**, 307-321 (2005).
24. Wiman, B. & Collen, D. Molecular mechanism of physiological fibrinolysis. *Nature***272**, 549-550 (1978).
25. Chandler, W. L. et al. Clearance of tissue plasminogen activator (TPA) and TPA/plasminogen activator inhibitor type 1 (PAI-1) complex: relationship to elevated TPA antigen in patients with high PAI-1 activity levels. *Circulation***96**, 761-768 (1997).
26. Hochman, M. A., Seery, C. M. & Zarbin, M. A. Pathophysiology and management of subretinal hemorrhage. *Surv. Ophthalmol.***42**, 195-213 (1997).
27. Olivier, S., Chow, D. R., Packo, K. H., MacCumber, M. W. & Awh, C. C. Subretinal recombinant tissue plasminogen activator injection and pneumatic displacement of thick submacular hemorrhage in age-related macular degeneration. *Ophthalmology***111**, 1201-1208 (2004).
28. de Jong, J. H. et al. INTRAVITREAL versus SUBRETINAL ADMINISTRATION OF RECOMBINANT TISSUE PLASMINOGEN ACTIVATOR COMBINED with GAS FOR ACUTE SUBMACULAR HEMORRHAGES DUE, TO AGE-RELATED MACULAR DEGENERATION: an exploratory prospective study. *Retina***36**, 914-925 (2016).

Tables

Table 1. Baseline characteristics of patients with submacular hemorrhage secondary to age-related macular degeneration

	Group A	Group B	Group C	P-value
Number of patients	19	20	22	
Mean age, years	74.7 ± 8.0	77.0 ± 8.4	77.2 ± 10.8	0.209*
Sex, male (%)	14 (73.7%)	14 (70.0%)	12 (54.6%)	0.388**
Lens status, phakic (%)	12 (63.6%)	10 (50.0%)	14 (63.6%)	0.670**
Systemic anticoagulants (%)	6 (31.6%)	7 (35.0%)	10 (45.5%)	0.630**
Duration of SMH, days	10.2 ± 4.2	3.4 ± 3.4	5.5 ± 4.3	<0.001*
SMH size, disc diameters	5.8 ± 2.6	7.0 ± 2.8	5.6 ± 3.0	0.108*
Baseline logMAR BCVA	0.85 ± 0.34	1.02 ± 0.39	1.06 ± 0.34	0.128*
SMH thickness, μm	402.1 ± 225.4	560.8 ± 383.2	536.4 ± 305.2	0.420*
Baseline CRT, μm	595.9 ± 322.6	704.4 ± 423.0	603.5 ± 305.5	0.770*
Baseline PED height, μm	109.1 ± 126.7	204.6 ± 300.3	244.4 ± 286.5	0.455*
Baseline CCT, μm	263.4 ± 110.7	193.2 ± 73.7	192.7 ± 100.5	0.102*
EZ disruption (%)	2 (10.5%)	4 (20.0%)	1 (4.6%)	0.282**
ELM disruption (%)	10 (52.6%)	8 (40.0%)	7 (31.8%)	0.398**

Continuous variables are presented as mean ± standard deviation and were compared using the Kruskal–Wallis test (*). Categorical variables are presented as numbers (percentages) and were compared using the chi-square test (**). A P-value < 0.05 was considered statistically significant.

Abbreviations: SMH, submacular hemorrhage; logMAR, logarithm of the minimum angle of resolution; BCVA, best-corrected visual acuity; CRT, central retinal thickness; PED, pigment epithelial detachment; CCT, central choroidal thickness; EZ, ellipsoid zone; ELM, external limiting membrane.

Table 2a. Multivariable linear regression analysis for BCVA at 12 months

Variable	β (Estimate)	SE	95% CI	P-value
Intercept	0.083	0.253	−0.425 to 0.592	0.744
Treatment group: Group B†	0.111	0.106	−0.103 to 0.324	0.304
Treatment group: Group A†	0.251	0.113	0.025 to 0.477	0.030
Time to treatment (days)	−0.013	0.018	−0.049 to 0.022	0.463
SMH size (per 1 DD)	0.041	0.026	−0.011 to 0.092	0.120
Baseline BCVA (logMAR)	0.300	0.188	−0.076 to 0.677	0.115
SMH thickness	0.00010	0.00023	−0.00037 to 0.00057	0.679
Baseline EZ disruption	−0.269	0.107	−0.483 to −0.055	0.015

Dependent variable: BCVA at 12 months (logMAR)

Model fit: $R^2 = 0.347$, Adjusted $R^2 = 0.261$, RMSE = 0.492, $n = 61$

Overall model: F = 4.03, P = 0.001

†Reference category: Group C (triple therapy)

The overall effect of the treatment group was assessed using a Type III test within the multivariable model (P = 0.001).

Data are presented as regression coefficients (β) with SE and 95% CI. All variables were entered simultaneously in the multivariate linear regression model.

Abbreviations: BCVA, best-corrected visual acuity; logMAR, logarithm of the minimum angle of resolution; SE, standard error; CI, confidence interval; SMH, submacular hemorrhage; DD, disc diameter; EZ, ellipsoid zone.

Table 2b. Least-squares mean BCVA at 12 months and Tukey's HSD post hoc comparisons

Treatment group	LS mean BCVA (logMAR)	SE
Group A	0.845	0.150
Group B	0.704	0.145
Group C	0.232	0.144

Pairwise comparisons (Tukey's HSD)				
Comparison	**Difference**	**SE**	**95% CI**	**P-value**
Group A − Group C	0.613	0.175	0.190 to 1.036	**0.003**
Group B − Group C	0.472	0.164	0.078 to 0.867	**0.015**
Group A − Group B	0.140	0.199	−0.340 to 0.620	0.762

α = 0.05, Tukey's HSD.

Least-squares means were estimated from the multivariable linear model. Comparisons with $P < 0.05$ indicate statistically significant differences between groups. Groups that do not share the same letter are significantly different according to Tukey's HSD.

Abbreviations: BCVA, best-corrected visual acuity; logMAR, lagarithm of the minimum angle of resolution; SE, standard error; CI, confidence interval; LS, least-squares.

Figures

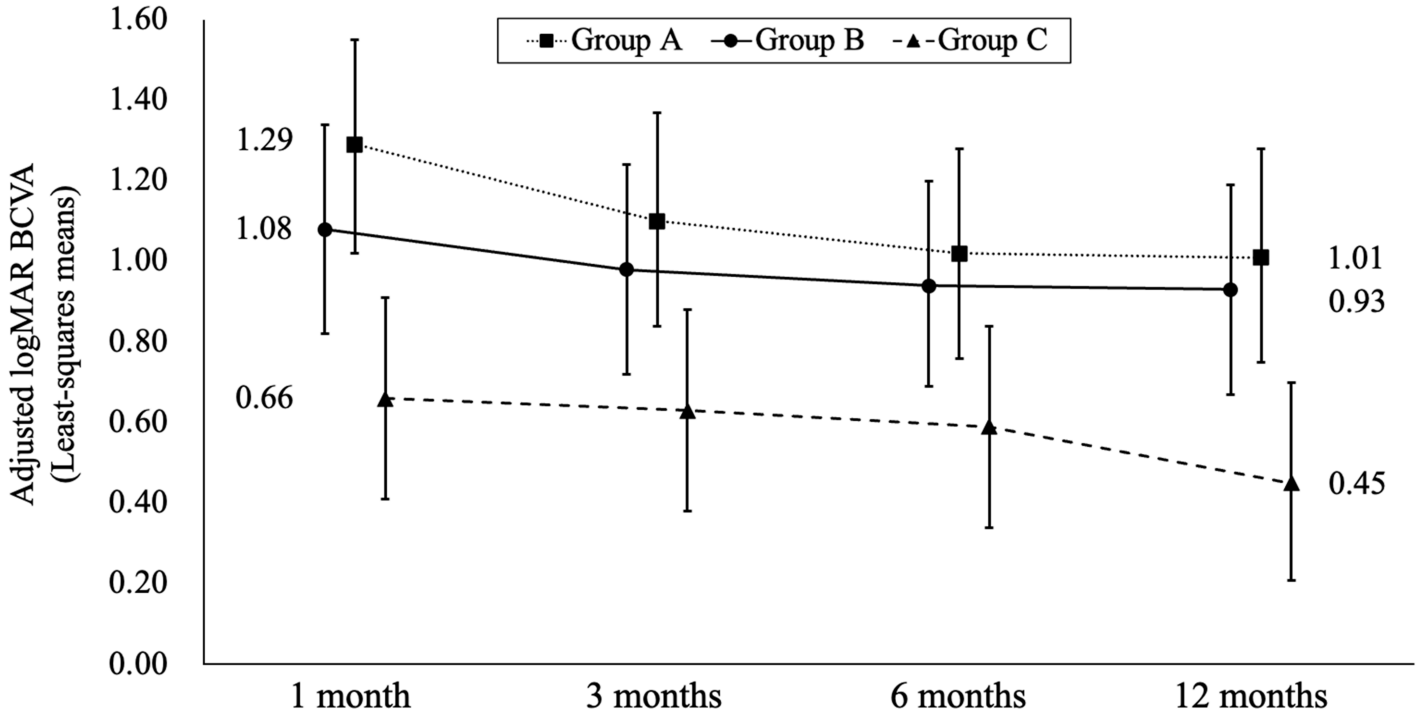


Figure 1

Baseline-adjusted visual acuity over 12 months

Longitudinal changes in baseline-adjusted logMAR BCVA were estimated using a linear mixed-effects model. Error bars represent 95% confidence intervals. Final adjusted values at 12 months were 1.01 for Group A, 0.93 for Group B, and 0.45 for Group C. The model showed a significant effect of treatment ($P = 0.009$) and time ($P < 0.001$), whereas the treatment-by-time interaction was not significant ($P = 0.510$).

Abbreviations: BCVA, best-corrected visual acuity; logMAR, logarithm of the minimum angle of resolution.

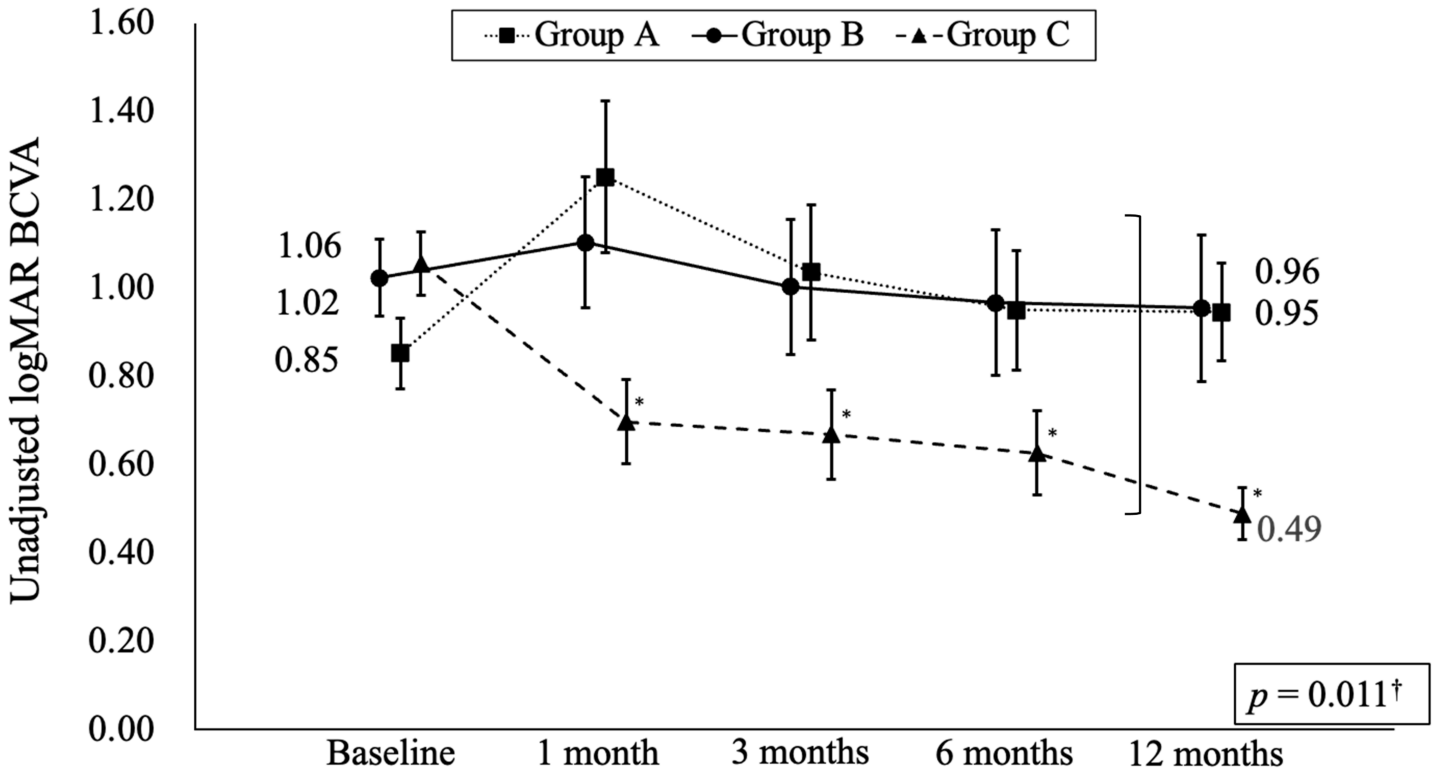


Figure 2

Unadjusted visual acuity over 12 months

Unadjusted logMAR BCVA changed from 0.85 to 0.95 in Group A, from 1.02 to 0.96 in Group B, and from 1.06 to 0.49 in Group C over 12 months. Error bars represent the standard error. Within-group changes were analyzed using the Friedman test (Group A, P = 0.319; Group B, P = 0.305; Group C, P < 0.001). Since only Group C showed a significant overall effect, post hoc comparisons were performed using the Wilcoxon signed-rank test with Holm correction (1 month, P = 0.001; 3 months, P = 0.006; 6 months, P = 0.001; 12 months, P < 0.001). Between-group differences at 12 months were analyzed using the Kruskal–Wallis test (P = 0.011). Asterisks (*) indicate significant differences compared with baseline, and daggers (†) indicate significant between-group differences at 12 months.

Abbreviations: BCVA, best-corrected visual acuity; logMAR, logarithm of the minimum angle of resolution.

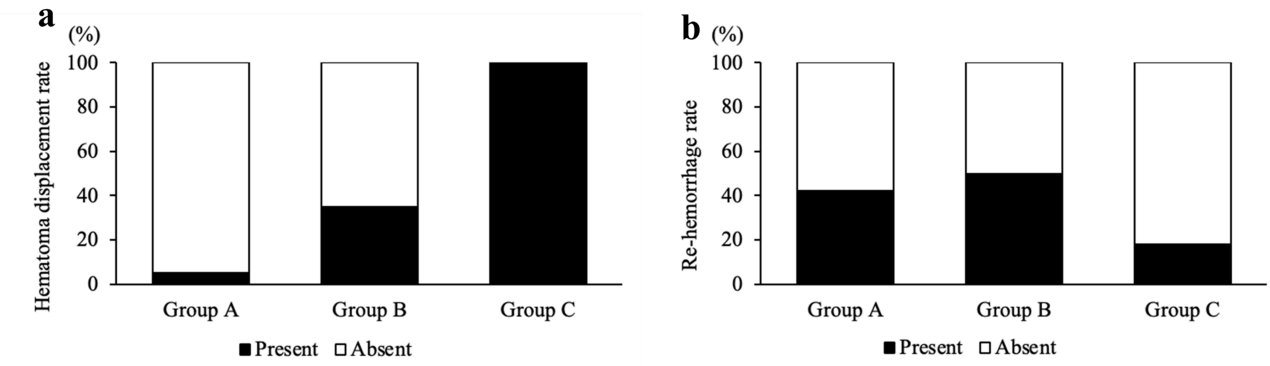


Figure 3

Hematoma displacement and re-hemorrhage rates

(**a**) Complete hematoma displacement rates: Group C, 100%; Group B, 35.0%; Group A, 5.3%.

(**b**) Bar graph comparing re-hemorrhage rates: Group C, 18.2%; Group B, 55.0%; Group A, 42.1%.

Overall group differences were analyzed using Pearson's chi-square test ($P < 0.001$ for displacement; $P = 0.037$ for re-hemorrhage).

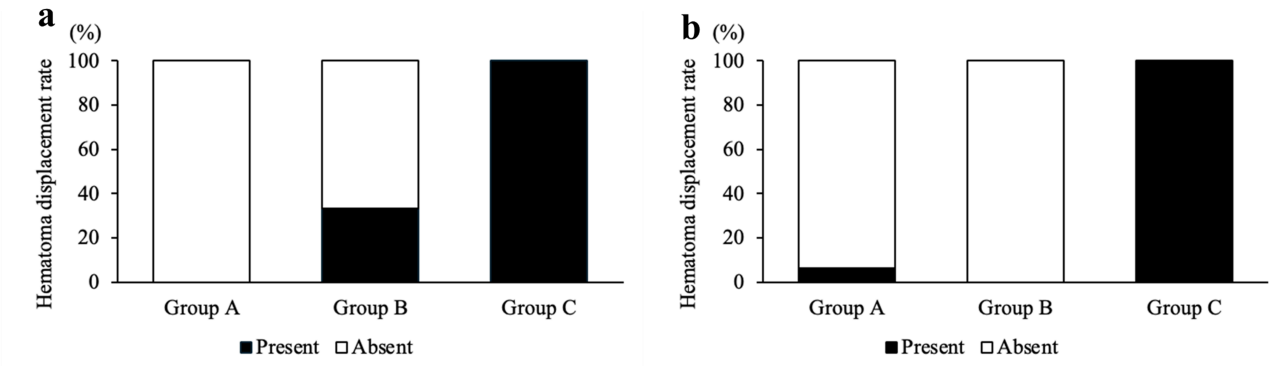


Figure 4

Hematoma displacement rates stratified by time to treatment

(**a**) Complete hematoma displacement in the 0–7-day subgroup: Group A, 0/3 (0%); Group B, 6/18 (33.3%); Group C, 18/18 (100%).

(**b**) Complete hematoma displacement in the 8–14-day subgroup: Group A, 1/16 (6.3%); Group B, 0/2 (0%); Group C, 4/4 (100%).

Pairwise comparisons were performed using Fisher's exact test. In the 0–7-day subgroup, displacement rates were significantly lower in Groups A and B than in Group C (both P< 0.001). In the 8–14-day subgroup, displacement rates were significantly lower in Group A than in Group C (P = 0.002), whereas the difference between Groups B and C did not reach significance after Bonferroni correction (P = 0.022; adjusted significance threshold, P < 0.017).



Figure 5

Representative cases from each treatment group

(**a**) Group A: intravitreal anti-VEGF monotherapy; (**b**) Group B: anti-VEGF combined with SF_6 gas tamponade; and (**c**) Group C: triple therapy consisting of anti-VEGF, SF_6 gas tamponade, and intravitreal tPA.

For each group, color fundus photographs (upper panels) and corresponding OCT images (lower panels) are shown at baseline, 1 month, and 12 months.

At 1 month, Group C demonstrated complete displacement of the submacular hemorrhage from the foveal center, with a significant reduction in residual blood on fundus photography and OCT.In contrast, Groups A and B showed only partial hematoma displacement at the same time points.

By 12 months, the submacular hemorrhage was largely absorbed in all groups. Disruption of the EZ was evident in Groups A and B, whereas partial preservation or restoration of EZ integrity was observed in Group C.

Abbreviations: OCT, optical coherence tomography; EZ, ellipsoid zone; SF_6, sulfur hexafluoride; tPA, tissue plasminogen activator; anti-VEGF, anti-vascular endothelial growth factor.