



A New Method of Subretinal Injection of Tissue Plasminogen Activator and Air in Patients With Submacular Hemorrhage

Subretinal macular hemorrhage (SRMH) is an important cause of irreversible visual loss¹ because it causes permanent functional and anatomical damage to photoreceptors, mainly as a result of 1) barrier effect (separation of photoreceptors from the retinal pigmented epithelium by the blood), 2) tractional changes (clot contraction causing sharing of the photoreceptors),² and 3) toxic damage (hemosiderin from hemoglobin is toxic to the photoreceptors).³ In fact, Litts et al⁴ demonstrated histologically that cones overlying an SRMH become degenerated, with translocated mitochondria.

Recent and small SRMH may be treated with intravitreal injections of perfluoropropane (C₃F₈) associated with or without tissue plasminogen activator (tPA) followed by patient positioning in an attempt to move the blood away from the fovea, to avoid the loss of central visual acuity.¹ However, if the SRMH is massive, thick, subfoveal, and predominantly inferior, it should be displaced from the macular area using a more direct and invasive procedure. After pars plana vitrectomy (PPV), the most frequently used technique in such cases is subretinal injections of tPA, followed by fluid–air exchange and intravitreal injection of non-expandable gas.⁵ According to a recent study, submacular injection of filtered air may be a helpful adjuvant to tPA injection.⁶ Finally, in SRMH associated with choroidal neovascularization (CNV) caused by age-related macular

degeneration (AMD), intravitreal injection of vascular endothelial growth factor antagonist (Avastin, Roche, Basel, Switzerland; Lucentis, Roche, Basel, Switzerland; or Eylea, Bayer, Leverkusen, Germany) should be added at the end of the surgery to ensure regression.⁷

The technique of choice for subretinal tPA or air injection for treating SRMH usually requires an assistant physician to push the insulin syringe plunger while the surgeon holds the syringe body caring for the cannula to be steady at the subretinal space. However, significant shaking both by the surgeon and the assistant may occur during the procedure, possibly leading to complications, such as hemorrhage, and if the plunger is pushed too fast, macular hole can develop. To reduce the risk of such complications the surgeon can connect the subretinal cannula to a specific flexible tubing; nevertheless, it continues to be assistant dependent and associated with an uncontrolled infusion pressure.

Thus, we present a safe and affordable method of delivering tPA and filtered air into the subretinal space using an insulin syringe with a 41-gauge cannula coupled to the viscous fluid control unit of a standard vitrectomy system. Using such a technique, a slower and semiautomated controlled mechanism was obtained, with marked stability during the procedure, which is the primary advantage of this technique.

Methods

Following approval by an institutional review board, this retrospective cohort study was performed between January 2013 and October 2015, according to the principles of the Declaration of Helsinki. All study participants gave their informed written consent. Patients with overt massive foveal SRMH and symptoms of central scotoma for up to 10 days were eligible. Patients with retinal macroaneurysms, blood dyscrasia, anticoagulant drug therapy, and AMD with a history of poor visual acuity associated with macular scar or atrophy on complementary tests were excluded.

The use of adjuvants, such as subretinal air or intraocular gas at the end of the procedure, was at the

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discretion of the surgeon. Preoperative and postoperative evaluations, included visual acuity measurement (Snellen chart), biomicroscopy, applanation tonometry, funduscopy and fundus photography.

Surgical Technique

A 1-mL insulin syringe was cut with a scalpel at the 0.5-mL mark. The plunger was also cut, leaving it 1 cm longer than the syringe body (Figure 1A, and see **Video, Supplemental Digital Content 1**, <http://links.lww.com/IAE/A584>). Then, 0.04 mL of tPA (0.125 mg/0.1 mL) was aspirated with a 23-gauge needle, which was subsequently replaced by a nonextendable 41-gauge cannula. The cut syringe was placed into the 10-mL syringe of the viscous fluid control unit of a vitrectomy device (Constellation; Alcon Laboratories, Inc, Fort Worth, TX), fastening it in a circular hole cut in the rubber tip of the plunger of the 10-mL syringe (Figure 1C and see **Video, Supplemental Digital Content 1**, <http://links.lww.com/IAE/A584>). After PPV, the retina was penetrated with the 41-gauge cannula at three optic disk diameters from the fovea (dislocated inferiorly and temporally) over the hemorrhagic retinal detachment. Pressing the foot-switch of the vitrectomy system gently and progressively, a constant 15 mmHg pressure was exerted on the plunger of the insulin syringe, pushing it forward and delivering 0.4 mL of tPA (0.125 mg/0.1 mL) (see **Video, Supplemental Digital Content 1**, which

instructs how to prepare the insulin syringe with tPA and to place it into the 10-mL syringe of the viscous fluid control unit of a vitrectomy device; <http://links.lww.com/IAE/A584>). At this point, a small bullous retinal detachment could be observed.

In 3 of our 10 patients, an additional 0.3 mL of air was injected into the same subretinal space 1 minute later. The other 7 patients were submitted to complete fluid–gas exchange and 12% C₃F₈ infusion. Patients with CNV-AMD-related SRMH received an intra-vitreous injection of anti-vascular endothelial growth factor. After the procedure, all patients were instructed to remain lying down at an angle of 60° for 3 days. The procedure was considered successful when SRMH displacement was confirmed by fundus photography and optical coherence tomography (OCT).

Results

Ten patients (6 men) were enrolled in the study. The average age was 64.7 years (range, 31–90 years). In 7 patients (70%), SRMH was caused by CNV-AMD and trauma in 3 (30%) (Table 1). None of the patients had baseline vision better than 20/200. Best-corrected visual acuity (BCVA) was hand motion in 40%, counting fingers in 40%, and 20/400 in 20%. The average duration of SRMH was 4.9 days (range, 2–10 days). In three patients (30%), the macular subretinal space was injected with air during the surgery. The remainder

Fig. 1. A. *Rubber tip of the plunger of the 10-mL syringe with a hole in the center made manually by a scalpel; **insulin syringe plunger cut 1 cm longer from its body; ***1-mL insulin syringe manually cut at the 0.5-mL mark and a non-extendable 41-gauge cannula coupled at its tip. **B.** Insulin syringe prepared with tPA fastened with a rubber tip of the plunger of the 10-mL syringe and nonextendable 41-gauge cannula. **C.** Prepared insulin syringe placed inside the 10-mL syringe of the viscous fluid control unit of a vitrectomy device and ready to be used.

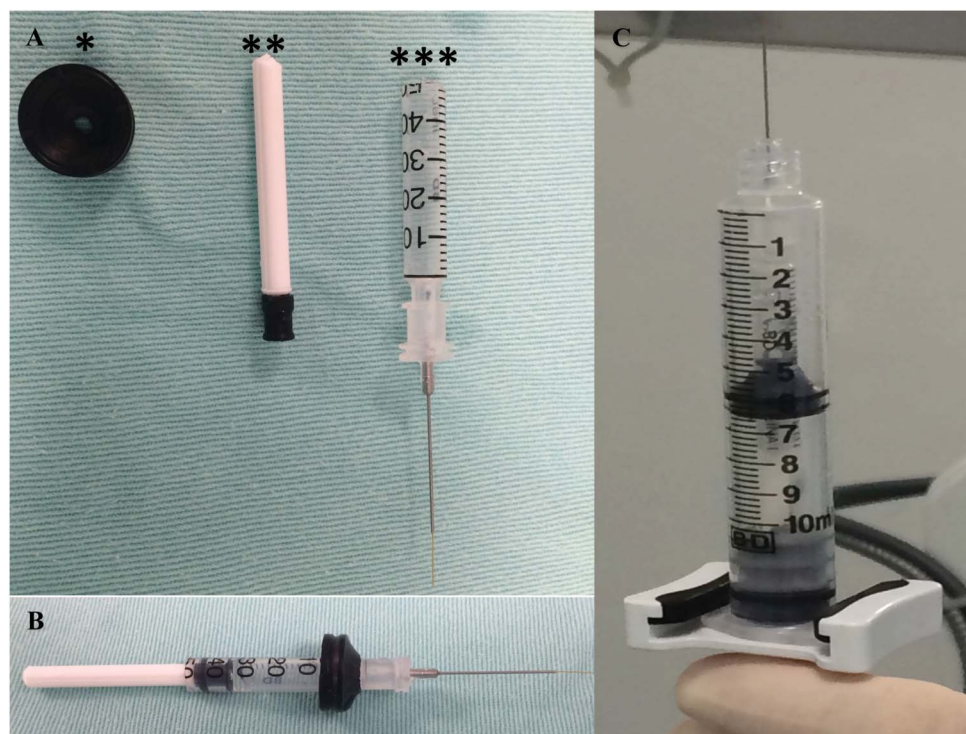


Table 1. Summary of Patient Data

Case	Sex	Age, year	Diagnosis	Duration SRMH, days	SR Air	Visual Acuity	
						Preoperative	Postoperative
1	F	80	AMD	3	Yes	CF 50 cm	20/60
5	M	90	AMD	10	Yes	20/400	20/30
3	F	80	AMD	7	No	HM	CF 1.5 m
4	M	88	AMD	7	No	HM	CF 1.5 m
5	M	32	Trauma	7	No	CF 50 cm	CF 1 m
6	M	63	AMD	5	No	HM	20/400
7	F	74	AMD	4	No	CF 1 m	20/400
8	F	72	AMD	2	No	CF 1 m	CF 2 m
9	M	31	Trauma	2	No	HM	20/400
10	M	37	Trauma	2	No	20/400	20/200

CF, counting fingers; cm, centimeter; HM, hand motion; m, meters; SR, subretinal.

(70%) received a subretinal injection of tPA and an intravitreal infusion of C₃F₈ after the fluid–air exchange.

In all cases, the injection of tPA into the subretinal space was performed safely, at the recommended speed and volume. All patients presented postoperative SRMH displacement and anatomical improvement but still with poor visual results because of causative disease.

Case Reports

Case 1

An 80-year-old white woman with a BCVA of 20/40 in the right eye underwent anti-vascular endothelial growth factor therapy for

CNV-AMD. Three months after the last injection, she presented with a central scotoma. On fundus examination, an SRMH was observed affecting the entire posterior pole, including the fovea (Figure 2A). At the time, BCVA was counting fingers at 50 cm. Preoperative spectral-domain OCT revealed a large and thick SRMH and loss of foveal depression (Figure 2B). After 3 days of SRMH, the patient was submitted to PPV with subretinal injections of tPA (0.125 mg/mL) and air (0.3 mL). One month after the surgery, BCVA had improved to 20/60, and SRMH had almost totally resolved (Figure 2C) and was barely visible on OCT, with persistence only of cystoid macular edema, indicating CNV-AMD activity (Figure 2D). The patient was prescribed pro re nata treatment with intravitreal injections of bevacizumab.

Case 2

A 90-year-old male patient was referred with acute vision loss in the left eye for 2 days. Vision in the right eye had been poor for 10

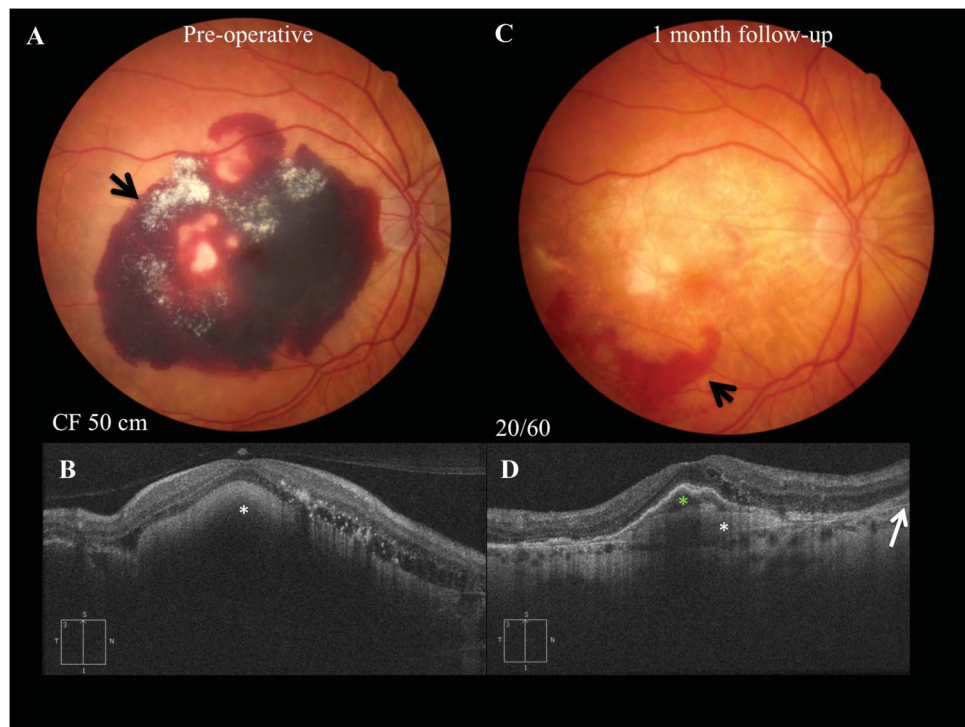


Fig. 2. Case 1. **A.** Preoperative retinography showing subretinal hemorrhage (black arrow). **B.** Preoperative OCT showing large SRMH (white asterisk). **C.** Follow-up retinography at 1 month showing multiple drusen and displacement of blood in the lower part of the macula (dark arrow). **D.** At 1-month follow-up, OCT showing residual subretinal hemorrhage (white arrow), retinal pigment epithelium detachment (green asterisk), and neovascular membrane (white asterisk).

years because of a disciform scar from CNV-AMD. On presentation, visual acuity was hand motion in the right eye and 20/400 in the left eye. Fundus examination of the right eye showed a disciform scar affecting the entire macular region and drusen in the midperiphery. An SRMH was observed inferiorly in the macular region of the left eye (Figure 3A), affecting the fovea on OCT (Figure 3B). After 10 days with symptoms, the patient was submitted to PPV, injection of tPA and air in the subretinal space, and intravitreal injection of bevacizumab. Thirteen days after the procedure, BCVA was 20/80, despite signs of CNV activity on OCT. The patient was treated with three monthly intravitreal injections of bevacizumab. After 3 months, BCVA was 20/30 in the left eye, with a small CNV-AMD-related subretinal hemorrhage at 6 o'clock under the main inferior temporal vascular arcades (Figure 3C) and no signs of foveal CNV activity or SRMH on foveal OCT (Figure 3D).

Discussion

Nowadays, massive SRMH is generally managed with PPV, including the injection of tPA and/or air into the subretinal space (with or without intraocular gas infusion), to reduce the chances of permanent central blindness.^{5,8,9}

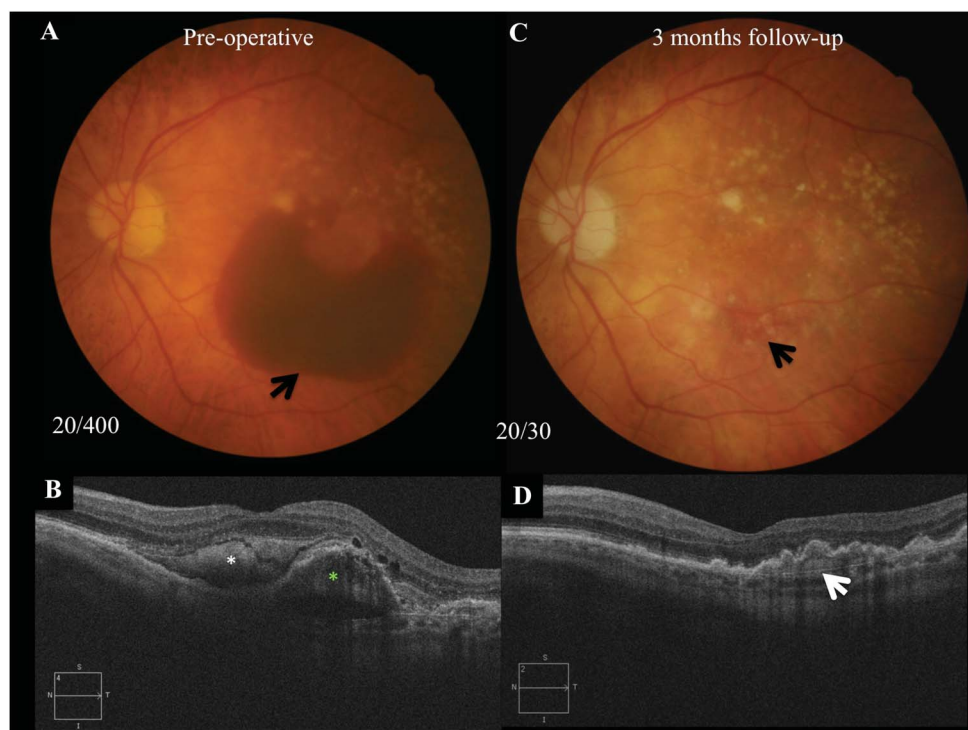
However, between the 2 methods that are used to manually inject subretinal tPA or air, significant shaking may occur while the surgeon holds the body of the insulin syringe with the 41-gauge cannula in the subretinal space and the assistant physician pushes the plunger and also a high pressure stream flow of liquid or air could generate a macular hole even if the

cannula is connected to the syringe by a flexible tube. In the current study, we designed a simple adaptation to the procedure that leads to some automatization and greater safety using the viscous fluid control unit of the vitrectomy system. Designed for silicone oil injection only, the system uses a 10-mL syringe, making it difficult to exactly inject the usual small amount of tPA (0.4 mL) and air (0.3 mL) into the macular subretinal space. To bypass this difficulty, an insulin syringe was fitted into the 10-mL syringe.

The technique was used in 10 patients with SRMH caused by CNV-AMD ($n = 7$) or blunt trauma ($n = 3$). In all cases, the adapted injection system was effective, enabling subretinal air and tPA to be delivered in the submacular space without complications and displacing the blood from the macula successfully, as described in other studies.^{5,7,10} Visual acuity presented a small average improvement in most CNV-AMD patients but with a considerable line gain in Case 1 and Case 2 (Table 1), matching findings from other studies.^{5,7} Visual acuity improvement in blunt trauma patients was slight ($n = 2$) or absent ($n = 1$). Regardless of the etiology, the poor visual acuity outcome may in part be caused by previous macular damage from underlying disease and the toxic effects of blood in the subretinal space.

Although significant complications during manual subretinal injection of tPA are rarely reported, it may occur particularly in cases with smaller volumes and shallow subretinal hemorrhages. For this reason, this

Fig. 3. Case 2. **A.** Preoperative retinography showing a large subretinal hemorrhage at the macula (black arrow) and drusen above. **B.** Preoperative OCT with subretinal hemorrhage at the fovea (white asterisk) associated with adjacent hemorrhagic retinal pigment epithelium detachment (green asterisk). Note intraretinal cysts (a secondary sign of CNV activity). **C.** Follow-up retinography at 3 months showing completely resolved subretinal hemorrhage at the fovea with a small residual area of blood below (black arrow). Note the multiple soft drusen. **D.** Horizontal foveal OCT showing double-layer sign with CNV inside the large and shallow retinal pigment epithelium detachment (white arrow).



method would be helpful in cases that will receive subretinal stem cells or genetic therapy. In our experience, manual injection of tPA may lead to complications, such as increase of the retinotomy at the time of retinal piercing, hemorrhage from retinal vessels, and/or withdrawal of the 41-gauge cannula tip from the subretinal space before the substance is injected. Moreover, in our opinion, the greatest shortcoming of the manual technique is the extreme difficulty of injecting air slowly into the subretinal space and the risk of macular hole formation. Therefore, the introduction of the aforementioned technique has greatly facilitated the procedure in our hands.

Analysis of the results in our study was limited by its retrospective nature, the fact that it was performed by a single surgeon, the relatively small sample, and the absence of control group using the manual injection technique. Despite these, the ease of performing, the simplicity of its execution, and low cost involved indicate us that the use of an insulin syringe coupled to the viscous fluid control unit of a vitrectomy device is a safe and affordable method for automatically injecting tPA and air into the subretinal space of patients submitted to treatment for massive SRMH. Further studies are welcome to validate our findings.

Key words: age-related macular degeneration, submacular hemorrhage, subretinal injection of tPA, optical coherence tomography, vitreoretinal surgery, macular surgery, pars plana vitrectomy.

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