Ultrasound biomicroscopy and optical coherence tomography imaging of filtering blebs after deep sclerectomy with new collagen implant

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PURPOSE. *To identify the clinical and anatomic characteristics of filtering blebs after glaucoma surgery with a new biodegradable collagen implant, Ologen, using ultrasound biomicroscopy (UBM) and Visante anterior segment optical coherence tomography (OCT).*

METHODS. *The authors conducted a prospective interventional case series in 15 eyes with open-angle glaucoma. The authors performed limbal-based deep sclerectomy with Ologen implantation in the scleral bed. UBM, Visante anterior segment OCT, and a complete ophthalmic examination were performed at each follow-up visit, at 1 day, and 1, 4, and 12 weeks postoperatively.*

RESULTS. *Intraocular pressure (IOP) was significantly reduced (p<0.001) from a mean preoperative value of 24.2±6.8 mmHg (n=2.82 glaucoma medications) to a mean postoperative value of 8.1±1.2 (n=0), 8.5±1.3 (n=0), 11.7±3.2 (n=0), and 14.2±3.9 mmHg (n=0.33) at 1 day, and 1, 4, and 12 weeks, respectively. Lower IOP correlated with bleb height and low trabeculocorneal membrane thickness (r=0.79, p<0.01, r=0.91, p<0.001) based on UBM examination. Lower IOP correlated with thin bleb wall, large subconjunctival fluid spaces, and low bleb tissue reflectivity (r=0.81, p<0.01, p<0.001, and p<0.001) based on OCT examination. No postoperative complications were reported.*

CONCLUSIONS. *UBM and OCT examinations are useful methods to evaluate outflow mechanisms after glaucoma surgery. Deep sclerectomy with Ologen implantation seems to be an effective and well-tolerated method to reduce IOP. (Eur J Ophthalmol 2009; 19: 223-30)*

KEY WORDS. *Ologen, Ultrasound biomicroscopy, Optical coherence tomography, Filtering blebs, Deep sclerectomy*

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INTRODUCTION

Standard trabeculectomy, the main surgical treatment for glaucoma, requires full thickness perforation of the sclera (1), which leads to opening of the anterior chamber and decompression of the eye. Thus, complications of trabeculectomy, such as hypotony, hypotony maculopathy, shallow flat anterior chamber, choroidal detachment, choroidal hemorrhage, endophthalmitis, and bleb leaks, are commonly observed (2). To minimize the risk of overfiltration and to avoid these serious complications, several investigators have proposed nonperforating glaucoma surgery. Deep sclerectomy, which allows for filtration through a natural trabeculo-descemetic membrane acting as an aqueous humor outflow resistance site, was first described in 1989 (3). The safety and efficacy of deep sclerectomy has been demonstrated in many clinical studies (4, 5).

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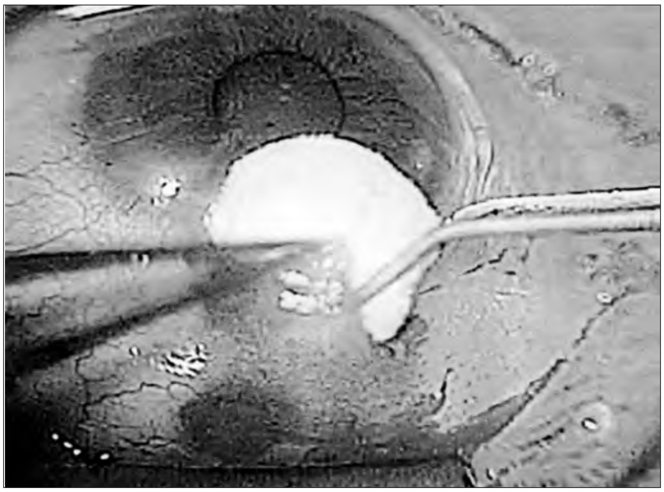

Fig. 1 - *Ologen implantation under conjunctiva.*

As with the trabeculectomy procedure, the success of deep sclerectomy strongly depends on the extent of the healing processes at the filtering site. The growth of fibrous tissue under the conjunctiva or in the filtration fistula is a natural part of the healing process of a cut or incision and this increases outflow resistance, leading to reduced aqueous flow through the filtration fistula. Deep sclerectomy is less sucessful in the eyes of patients who tend to produce larger amounts of fibrous tissue, such as young patients, keloid formers, patients with uveitic glaucoma, patients with repeat ocular surgeries, and African American patients (5, 6). Antifibrotic agents, such as 5-fluorouracil or mitomycin-C, given as a single application during surgery or as subconjunctival injections, may be used to minimize scar formation in high-risk patients (7), but the use of antifibrotic agents increases the risk of complications, especially wound leaks, late bleb breakdown, and corneal epithelial defects (8).

The purpose of Ologen, a new three-dimensional biodegradable collagen-glycosaminoglycan copolymer implant, is to produce a success rate after deep sclerectomy or trabeculectomy with Ologen equivalent to that after deep sclerectomy or trabeculectomy with antifibrotic agents, such as 5-fluorouracil or mitomycin-C. The purpose of this device is also to decrease the risk of complications compared to trabeculectomy or deep sclerectomy with antifibrotic agents. Placement of this device into the subconjunctival space is aimed at inducing random fibroblast growth, leading to normal wound healing (9). A previous study in rabbit eyes demonstrated that this porous matrix reduces the proliferation of fibroblasts and myofibroblasts, and the deposition of dense linear collagen, resulting in the regeneration of a nearly physiologic subconjunctival space (10). The collagen porous structure promotes the random growth of myofibroblasts and creates a new physiologic environment between the anterior chamber and subconjunctival space, thereby improving regenerating tissue-remodeling and reducing scar formation (10, 11).

In this study, we used Visante anterior segment optical coherence tomography (Visante OCT) and ultrasound biomicroscopy (UBM) to study the effect of Ologen implantation in deep sclerectomy, particularly to assess bleb morphology. The clinical efficacy and tolerability were also evaluated. To our knowledge, this is the first clinical study to evaluate this new biodegradable implant and to correlate the morphologic findings with the postoperative intraocular pressure (IOP) results (9-11).

METHODS

Ologen implant

The Ologen implant (OculusGen Biomedical Inc., Taipei, Taiwan) is indicated for use in glaucoma surgeries. Ologen comprises more than 90% collagen and less than 10% glycosaminoglycan (chondroitin-6-sulfate), which are both biodegradable. Ologen has a cylindrical shape (2.0±0.3 mm height × 6.0±0.5 mm diameter) with a 95% porous space. The pore sizes of the collagen matrix range from 20 µm to 200 µm (average 140±20 µm). After implanting this device into the subconjunctival space, the porous structure guides fibroblasts to proliferate randomly during the wound healing process, and therefore reduces scar formation (9). The structure is intended to reduce intraocular pressure by inducing a non-scarring physiologic regeneration of the extracellular matrix (10, 11). After implantation, the device completely degrades within 30 to 90 days. The Ologen implant has obtained European Conformity (CE) certification (Fig. 1).

Patients and procedures

We performed a prospective interventional case series on patients aged 18 years or older with medically or laser uncontrolled open-angle glaucoma requiring filtering surgery. Medically or laser uncontrolled open-angle glaucoma were defined as uncontrolled intraocular pressure (>21 mmHg) despite maximal tolerable medical treatment or previous argon laser/selective laser trabeculoplasty, with progressive visual field loss or cup/disc ratio progression. Maximum medical treatment was defined as all the drugs the patient was able to tolerate. Patients were included if they did not have laser trabeculoplasty or eye surgery within the past 6 months. Exclusion criteria were known hypersensitivity to porcine collagen, allergy reaction after collagen allergy test, and pregnancy. The study followed the tenets of the treaty of Helsinki and all patients provided both oral and written informed consent. All patients underwent a standardized nonpenetrating deep sclerectomy performed by one surgeon (P.D.). A traction suture was placed on the limbal cornea, and the conjunctiva and Tenon capsule were opened in the upper fornix. A superficial limbal-based scleral flap measuring 4 mm x 4 mm was created. A second triangular limbal-based deeper scleral flap was created and then removed. Schlemm's canal was then unroofed and significant aqueous humor filtration could be observed through the remaining trabeculo-descemetic membrane. The Ologen implant was placed in the scleral bed without sutures. No scleral sutures were used. The conjunctiva was then closed with a running 8/0 vicryl suture. Postoperatively, patients were treated topically with tobramycin and dexamethasone (Tobradex, Alcon Inc., Ft. Worth, TX) four times a day for 1 month.

All patients underwent a standard ophthalmic examination including best-corrected visual acuity, slit lamp examination, Goldman applanation tonometry, and binocular fundus examination before surgery and on day 1, and 1, 4, and 12 weeks postoperatively. UBM and OCT examination were performed on day 1, and 1, 4, and 12 weeks postoperatively.

Imaging procedure

The Zeiss Visante OCT Model 1000 (Carl Zeiss Meditec Inc, Dublin, CA) was used with the corneal acquisition protocol. The upper lid was lifted by the operator to maximize bleb exposure. Several radial and transverse sections were assessed. The parameters analyzed in the study were bleb wall thickness (distance between the conjunctiva to the top of the subconjunctival fluid space), bleb wall reflectivity (classified as low, moderate, or high), subconjunctival fluid space (classified as none, single small, multiple small, or large), suprascleral fluid space (classified as none, single small, multiple small, or large), mean deep bleb tissue reflectivity (classified as low, moderate, or high), visibility of the route under the scleral flap (classified as yes or no), and visibility of the sclera beneath the filtering zone (classified as none, poor, moderate, or good).

The Ultrasound Biomicroscope Model 840 (Humphrey Inc., San Leandro, CA) was used. Patients were placed in supine position, and a lid speculum and gel examination were used. Several radial and transverse scans were obtained with the 50-MHz probe. The following factors were assessed: presence of subconjunctival filtering bleb (classified as yes or no), filtering bleb reflectivity, maximal bleb height (peak distance between the most superior point of bleb reflectivity and the sclera), presence and dimensions of an intrascleral cavity (maximal height, maximal radial and transverse dimensions, volume), visibility of the route under the sceral flap (classified as yes or no), thickness of the residual trabeculocorneal membrane, and presence of a hyporeflective suprachoroidal space (classified as yes or no). The imaging parameters selected for the present study were established in earlier studies.

Statistical methods

The linear relationships between IOP and imaging parameters were measured with the Pearson Product Moment Correlation. The statistical significance of r was tested using a *t* test. Student *t* test was used to compare means and percentages. Chi-square tests were used for the analysis of dichotomous variable. SPSS statistical software version 9.0 was used for data analysis. The relationship between two variables was considered statistically significant when the p value was less than 0.05. The correlation between IOP and imaging parameters was evaluated at week 12. The criteria for complete success were IOP less than 14 mmHg at week 12 without additional medical, laser, or surgical therapies. Any requirement for Nd:YAG goniopuncture was considered to constitute partial success. Any requirement for additional medical treatment, laser trabeculoplasty, or filtering surgery was considered to constitute failure.

RESULTS

Clinical characteristics

We examined 15 eyes of 15 patients (8 men, 7 women). The ages ranged from 58 to 77 years, with an overall mean age of 68.7±7.2 years. Thirteen patients were Caucasian and two were of African origin. All patients had primary open-angle glaucoma with characteristic visual field and optic nerve head findings. Five patients had undergone previous intraocular surgeries; four patients had undergone cataract surgeries 2 to 11 years prior to the present study and one patient had undergone combined surgery (phacoemulsification and deep sclerectomy) 3 years prior to the present study. The mean number of previous glaucoma topical therapies was 2.82±0.84.

Surgical procedure, IOP decrease, and safety

No perforations of the trabecular membrane occurred during any of the surgeries. Further, there were no incidents associated with the use of Ologen. The implant at first seems rather large, but the volume instantly diminishes when the implant comes in contact with biologic fluids. As soon as the implant is in contact with the scleral bed, it melts and takes a gelatinous consistency, decreases in volume, and the conjunctiva can be sutured without difficulty.

IOP significantly reduced ($p<0.001$) from a mean preoperative value of 24.2±6.8 mmHg (n=2.82 glaucoma medications) to a mean postoperative value of 8.1±1.2 (n=0), 8.5±1.3 (n=0), 11.7±3.2 (n=0), and 14.2±3.9 mmHg (n=0.33) at 1 day, and 1, 4, and 12 weeks, respectively. No postsurgical complications (hypotony <5 mmHg, hyphema, bleb leaks, bleb infection) or allergic reactions were observed with this new device. In four patients, one of whom had undergone previous deep sclerotomy and one of whom was of African origin, the IOP decrease was considered to be insufficient to control the disease, and the patients required additional medical therapies (beta-blocker in one patient, and beta-blockers and carbonic anhydrase inhibitors in two patients) or laser therapy (Nd:YAG laser goniopuncture in one patient 3 months after surgery). The complete success rate was therefore 73.3% (11/15 patients) at week 12 and the partial success rate was 80% (12/15 patients) at week 12.

Imaging parameters

The UBM measurements are presented in Table I. Bleb height and trabeculocorneal membrane fineness correlated with low IOP ($r=0.79$, $p<0.01$; $r=0.91$, $p<0.001$) based on the UBM examination. Patients with a visible filtering bleb presence and hyporeflective suprachoroidal space had a lower IOP than those without a visible filtering bleb ($p<0.001$) and suprachoroidal space ($p<0.01$). Patients with highly reflective blebs had a higher IOP than those who had blebs with low reflectivity ($p<0.01$). Scleral route visibility and intrascleral cavity volume did not correlate significantly with IOP ($p>0.10$). Gradual resorption of the implant was observed; the complete resorption time was 1 month in 12 patients and 3 months in 3 patients (Fig. 2). The OCT measurements are presented in Table II. A thin

TABLE I - UBM MORPHOLOGIES OF THE 15 EYES

	Day 1	Week 1	Week 4	Week 12
IOP (mmHg)	8.1 ± 1.2	8.5 ± 1.3	11.7 ± 3.2	14.2 ± 3.9
UBM:				
- filtering bleb presence	15 yes, 0 no	15 yes, 0 no	15 yes, 0 no	12 yes, 3 no
- high or low reflexive bleb	14 low, 1 heigh	13 low, 2 heigh	12 low, 3 heigh	12 low, 3 heigh
- bleb height	6.2 ± 2.4 mm	6.6 ± 2.6 mm	4.1 ± 2.0 mm	0.8 ± 0.4 mm
- intrascleral cavity (height, radial and transverse dimensions, volume)	4.2 ± 1.3 mm; 13.2 ± 2.9 mm; 10.6 ± 2.5 mm; 587.7 mm^3	6.5 ± 2.6 mm; 15.4 ± 3.7 mm; 12.4 ± 1.8 mm; 1241.2 mm^3	2.7 ± 1.1 mm; 11.8 ± 2.5 mm; 8.5 ± 2.5 mm; 270.8 mm^3	0.5 ± 0.1 mm; 2.1 ± 0.6 mm; 1.3 ± 0.3 mm; 1.4 mm^3
- sceral route visibility	15 yes, 0 no	15 yes, 0 no	13 yes, 2 no	12 yes, 3 no
- trabeculocomeal membrane thickness	14 ± 2.4 μm	15 ± 2.6 μm	13 ± 2.2 μm	15 ± 2.8 μm
- hyporeflexive suprachoroidal space presence	12 yes, 3 no	12 yes, 3 no	5 yes, 10 no	3 yes, 12 no

IOP = intraocular pressure; UBM = ultrasound biomicroscopy.

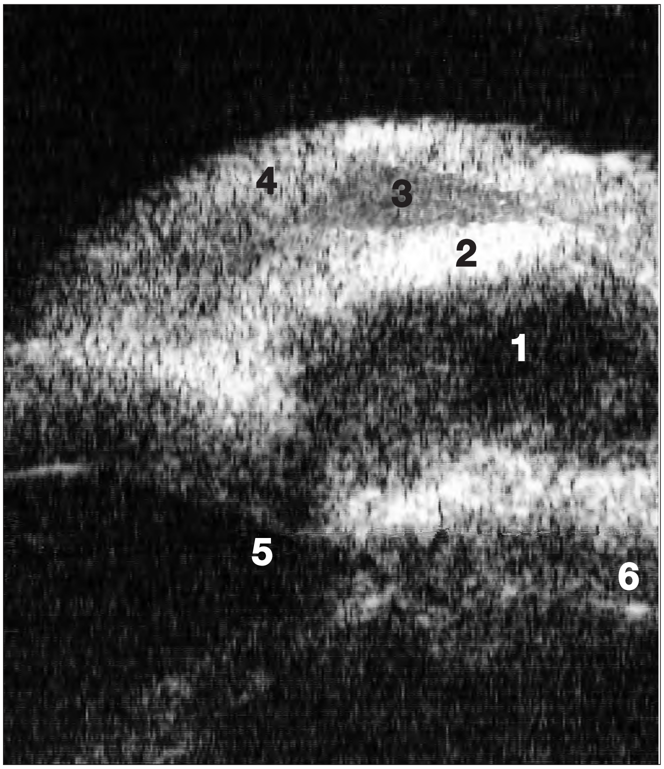

Fig. 2 - *Ultrasound biomicroscopy imaging of functional filtering bleb: **1** Ologen implant, **2** scleral flap, **3** low reflexive bleb, **4** bleb wall, **5** trabeculo-corneal membrane, and **6** suprachoroidal hyporeflexive space.*

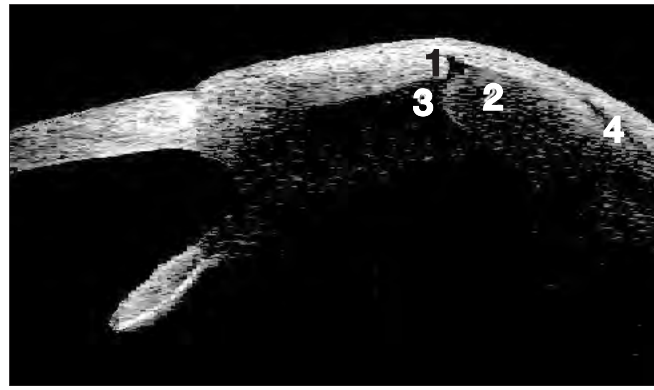

Fig. 3 - *Optical coherence tomography imaging of functionnal filtering bleb: **1** scleral flap, **2** Ologen implant, **3** scleral route, and **4** subconjunctival fluid space.*

bleb wall correlated with a low IOP (r=0.81; p<0.01). Large subconjunctival fluid spaces and low bleb tissue reflectivity were associated with a lower IOP (p<0.001). In contrast, a single small suprascleral fluid space, high deep bleb tissue reflectivity, and good visibility of the sclera beneath the filtering zone were significantly associated with higher IOP (p<0.01, p<0.01, and p<0.05). Poor visibility of the route under the scleral flap did not significantly correlate with IOP (p>0.10). Because of its low depth of penetration, OCT could not be used to assess implant resorption (Fig. 3).

TABLE II - OCT MORPHOLOGIES OF THE 15 EYES

	Day 1	Week 1	Week 4	Week 12
IOP (mmHg)	8.1 ± 1.2	8.5±1.3	11.7±3.2	14.2± 3.9
OCT:				
- bleb wall thickness	68.4 ± 33.9 µm	88.3 ± 21.8 µm	100.2 ± 26.2 µm	104.2 ± 30.8 µm
- mean reflectivity bleb wall	15/15 low	14/15 low, 1/15 moderate	12/15 low, 3/15 moderate	11/15 low, 3/15 moderate, 1/15 strong
- suprascleral fluid space (none, single small, multiple small, large)	5/15 large, 3/15 multiple small, 4/15 single small, 3/15 none	5/15 large, 3/15 multiple small, 4/15 single small, 3/15 none	3/15 large, 3/15 multiple small, 5/15 single small, 4/15 none	3/15 large, 2/15 multiple small, 6/15 single small, 4/15 none
- mean reflectivity deep bleb tissue	15/15 low	14/15 low, 1/15 moderate	13/15 low, 2/15 strong	12/15 low, 3/15 strong
- scleral route visibility	12 yes, 3 no	12 yes, 3 no	11 yes, 4 no	6 yes, 9 no
- subconjunctival fluid space (none, single small, multiple small, large)	13/15 large, 2/15 multiple small	13/15 large, 1/15 multiple small, 1/15 single small	11/15 large, 1/15 multiple small, 3/15 single small	9/15 large, 2/15 multiple small, 4/15 single small
- sclera visibility	13/15 no, 2/15 poor	12/15 no, 3/15 poor	11/15 no, 4/15 poor	6/15 no, 2/15 poor, 7/15 good

IOP = intraocular pressure; OCT = optical coherence tomography.

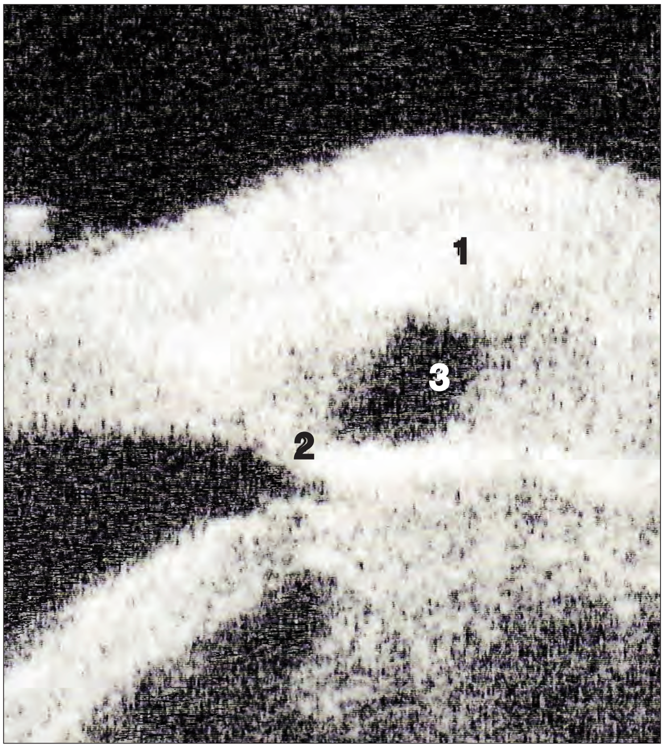

Fig. 4 - *Ultrasound biomicroscopy imaging of encapsuled bleb:* ***1*** *high reflexive bleb,* ***2*** *trabeculocorneal membrane, and* ***3*** *Ologen implant.*

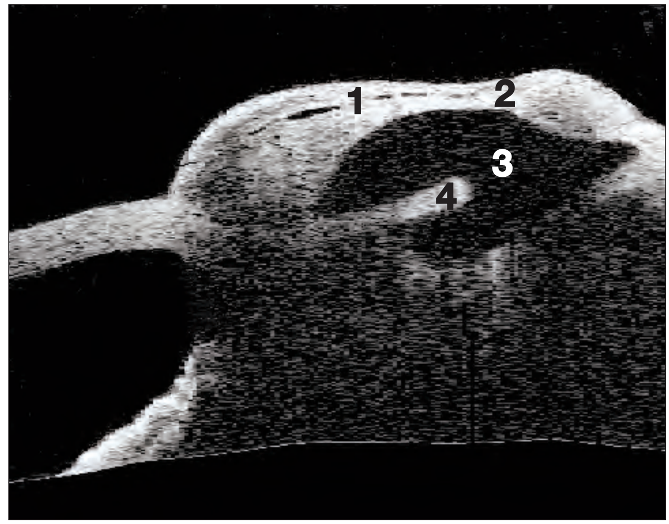

Fig. 5 - *Optical coherence tomography imaging of cystic bleb 12 weeks after surgery:* ***1*** *thick and high reflexive bleb wall,* ***2*** *subconjunctival fluid space,* ***3*** *large suprascleral fluid space, and* ***4*** *scleral flap.*

DISCUSSION

Successful glaucoma surgery strongly depends on the extent of the healing process. Bleb morphology is an indicator of the healing process and the surgical success. Intrableb morphology cannot be evaluated under slit-lamp biomicroscopy. UBM and OCT allow for detailed anatomic assessment of the bleb morphology. Several investigators have established that UBM and OCT can be used to identify morphologic changes in blebs related to wound healing, and to identify parameters for the functional prognosis of the filter (12-21). In a previous study, Kazakova et al reported that a hyporeflective suprachoroidal space significantly correlated with a lower IOP based on UBM examination after deep sclerectomy (12). In a similar study, Roters et al demonstrated that filtering blebs with low reflectivity, a visible route under the scleral flap, and a thick remaining trabeculocorneal membrane were associated with a long-term IOP reduction based on UBM examination after viscocanalostomy (14). In another study with OCT examination, Savini et al reported a significant association between numerous subconjunctival hyporeflective fluid-filled spaces, diffuse hyporeflective tissue, and good IOP control after glaucoma surgeries, including deep sclerectomy (19).

Ologen is a new biodegradable collagen implant designed to reduce scar formation by guiding fibroblasts to randomly proliferate through the matrix pores (9-11). The purpose of this implant is to act as an alternative adjunct to 5-fluorouracil and mitomycin-C with fewer side effects. This implant is new and no human data regarding the antihealing properties of Ologen have been published. In the present study, we used OCT and UBM examination to identify the anatomic characteristics of filtering blebs after deep sclerectomy with this new device. In particular, we assessed the parameters related to bleb healing and surgical outcome discussed in previous studies. After 12 weeks, in 11 of 15 patients an IOP of less than 14 mmHg was achieved and maintained without medication. Similar to previously reported findings (12-21), we frequently observed high filtering blebs, a thin trabeculocorneal membrane, a hyporeflective suprachoroidal space, and blebs with low reflectivity based on UBM examination, and thin bleb walls, large subconjunctival fluid spaces, and low bleb tissue reflectivity based on OCT examination. Some of these parameters have been identified after trabeculectomy in previous studies, but not after deep sclerectomy

(17-21). In contrast, the IOP reduction was considered to be insufficient for control of the disease in four patients who required additional medical or laser therapies. In those patients, we found highly reflective blebs with UBM examination and a single small subconjunctival fluid space, high deep bleb tissue reflectivity, and poor visibility of the route under the scleral flap with OCT examination. Some of these parameters were associated with poor IOP control after deep sclerotomy or trabeculectomy in previous studies (12-20). Clinically, two of these four patients seemed to have encapsulated blebs, with a morphology similar to the nonfunctioning encapsulated blebs related to scar formation after trabeculectomy described by Van Buskirk et al (22) (Fig. 4). One patient had a flattened bleb with no elevation and Nd:YAG goniopuncture was successfully performed. In this case, the implant prevented the formation of fibrous adherences between the scleral bed and scleral flap, and probably facilitated goniopuncture success. Another patient had a cystic, avascular, and thin-walled filtering bleb (Fig. 5).
The present study has some limitations. Because of the noncomparative design of the study, the efficacy of this new device must be accepted with caution. Some multicenter, randomized clinical trials to evaluate the efficacy of glaucoma surgeries with this implant versus classical glaucoma surgeries without the implant are in progress. The qualities of this device are its ease of use and the enhanced possibility of minimizing scar formation without an antimitotic, thus avoiding associated complications, such as postsurgical healing defects, chronic bleb leakage, bleb infection, and endophthalmitis. In some cases, the use of the Ologen implant seemed less efficacious than the classical antimitotic. Of the 15 patients, 4 required additional medical therapies to control IOP. Further studies could compare the use of Ologen to the use of antimitotic, or could assess the use of Ologen combined with 5-fluorouracil and mitomycin-C in high risk-patients (keloid formers, those with uveitic glaucoma, those with repeat ocular surgeries, and patients of African descent).
In conclusion, the results of our study indicate that UBM and OCT examinations are useful methods in clinical practice to evaluate outflow mechanisms and intrableb morphology after glaucoma surgery. Successful filtering blebs show characteristic imaging parameters. Despite the above-mentioned limitations, deep sclerectomy with Ologen implantation seems to be an effective and well-tolerated method to lower IOP.

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