Limbal transplantation is now widely accepted as the treatment of advanced limbal stem cell deficiency. Herein, we describe a technique for harvesting thin limbal grafts from cadaveric corneoscleral rims and a sutureless method to secure the grafts to the recipient eye using fibrin glue. We report the results of fibrin glue–assisted keratolimbal allograft in 19 eyes of 16 patients, with the outcome measures being ocular surface stability, visual acuity, and postoperative complications. The results indicate that limbal allograft transplantation can be performed safely and successfully using only fibrin glue to secure the grafts. This can potentially improve surgical efficiency and patient comfort postoperatively.

Arch Ophtalmol. 2011;129(2):218-222

A retrospective review was conducted on 19 eyes (16 patients) with limbal stem cell deficiency that underwent fibrin glue–assisted KLAL between January 21, 2006, and June 26, 2009. Institutional review board approval was obtained. The causes of limbal stem cell deficiency included congenital aniridia (14 eyes), chemical injuries (2 eyes), atopic keratoconjunctivitis (1 eye), radiation induced (1 eye), and idiopathic (1 eye). Three of the aniridic pa-

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of the anterior edge of the adhesive strip of viscoelastic (Viscoat; Alcon Laboratories, Mansfield, Massachusetts) was spread on a sterile plastic platform. After securing the tissue to the surface, superficial lamellar dissection is carried forward onto the cornea, extending at least 1 mm anterior to the limbus, using a crescent blade.

Figure 1. Donor tissue preparation for keratolimbal allograft. A, A thin strip of viscoelastic is placed near the edge of the N-butyl-2-cyanoacrylate adhesive (Indermil Tissue Adhesive; Tyco Healthcare Group LP, Mansfield, Massachusetts) on a sterile plastic platform. B, After securing the tissue to the surface, superficial lamellar dissection is carried forward onto the cornea, extending at least 1 mm anterior to the limbus, using a crescent blade.

Figure 2. Keratolimbal allograft surgery in a recipient eye. A, Three 180° limbal donor grafts (asterisks) are placed in close proximity to one another to cover the entire bed (the graft at the upper left was slightly trimmed to fit). B, The fibrin glue is injected beneath the donor tissue to attach the limbal donor grafts to the corneoscleral bed.

RECIPIENT EYE

The recipient eye was typically anesthetized with a peribulbar or retrobulbar injection—occasionally general anesthesia was used. A 360° peritomy was performed with undermining and dissection of underlying adhesions. This allowed the conjunctiva to fall back and create space for the grafts. Superficial keratectomy was also performed to remove the abnormal epithelium and fibrovascular pannus using a No. 64 Beaver blade (Becton, Dickinson, and Co, Franklin Lakes, New Jersey). Next, three 180° donor limbal grafts were placed in close proximity to one another to cover the entire bed (Figure 2A). Tissue fibrin glue (Baxter AG, Vienna, Austria) was injected beneath the donor tissue. This was done using 2 separate syringes, with fibrinogen injected first beneath the grafts followed by thrombin applied in dropwise fashion as well as injected under the grafts. The excess glue was immediately milked from under the grafts using closed forceps (Figure 2B). After 2 to 3 minutes when the grafts appeared to be attached, any loose conjunctival tissue and excess fibrin glue were trimmed from the donor grafts. The host conjunctiva was approximated with the graft and sometimes pulled over on top of the donor conjunctiva (video 2). At the conclusion, a subconjunctival injection of 0.5 mL of dexamethasone disodium phosphate (10 mg/mL) and 0.5 mL of cefazolin (100 mg/mL) was given, and a neomycin sulfate, polymyxin B sulfate, and dexamethasone ointment was placed in the eye. The eye was patched and a shield was placed over the eye until the following day.

In the first 3 eyes, given the uncertainty with the fibrin glue, 1 or 2 sutures were used just to hold the grafts together and minimize the possibility of a gap forming between the grafts. No sutures were used in the remaining 16 eyes.

POSTOPERATIVE MANAGEMENT

Immediately postoperatively, all patients received topical prednisolone acetate, 1%, 4 times a day; gatifloxacin (Zymar) 4 times a day, and frequent

DOCTOR TISSUE PREPARATION

For patients with total limbal stem cell deficiency, typically 2 donor corneas under 50 years of age and less than 5 days old were obtained from the Illinois Eye Bank. The tissue was specifically requested to have large scleral rims (>3 mm) with as much conjunctiva as possible. The central corneas from both donor corneoscleral buttons were removed using a 7.5-mm trephine. Each rim was cut in half to produce a total of four 180° crescents. A thin layer of tissue N-butyl-2-cyanoacrylate adhesive (Indermil Tissue Adhesive; Tyco Healthcare Group LP, Mansfield, Massachusetts) was spread on a sterile plastic platform, followed by application of a thin strip of viscoelastic (Viscoat; Alcon Laboratories, Inc, Fort Worth, Texas) near the anterior edge of the adhesive (Figure 1A). One of the donor corneoscleral crescents was placed with the epithelial side up on the adhesive and allowed to attach firmly onto the platform—the viscoelastic helps to prevent the glue from tracking up the anterior and lateral edges of the tissue. Starting from its free edge, the conjunctiva was lifted up and dissected off the sclera by cutting the underlying attachments using Wescott scissors (Acme United Corp, Fairfield, Connecticut). This dissection was carried forward up to the limbus, after which it was continued forward in the cornea in a lamellar fashion using a crescent blade and keeping at a depth of approximately one-third to one-fourth (Figure 1B). The same procedure was performed on 2 additional donor corneoscleral crescents all from the same donor (video 1, http://www.archophthalmol.com).

POSTOPERATIVE MANAGEMENT

Immediately postoperatively, all patients received topical prednisolone acetate, 1%, 4 times a day; gatifloxacin (Zymar) 4 times a day, and frequent...
lubrication with nonpreserved artificial tears. Once the corneal epithelial defect had healed, the antibiotics were stopped and the patients began treatment with cyclosporine (Restasis), 0.05%, 2 to 3 times a day. All patients additionally received systemic immunosuppression consisting of oral prednisone, 1 mg/kg/d, tacrolimus, 4 mg twice daily (or cyclosporine, 3 mg/ kg/d, in 2 cases), and mycophenolate mofetil, 500 to 1000 mg twice daily (or azathioprine, 100 mg/d, in 2 cases) starting in the evening of the day of surgery or sooner depending on the level of inflammation. The oral prednisone was typically tapered off by 3 to 4 months, while the other 2 agents were continued for a minimum of 18 to 24 months.

The primary outcome measures were ocular surface stability, visual acuity, and postoperative complications. Ocular surface stability was defined as the absence of conjunctivalization, no recurrent epithelial defects, and no corneal neovascularization. Failures were defined as the presence of abnormally high fluorescein permeability with diffuse late-staining epithelium, recurrence of conjunctivalization, increased neovascularization, and persistent or recurrent epithelial defects.

### RESULTS

A total of 19 procedures were performed in 16 patients (6 males, 10 females) with the mean age of 44.1 years (range, 12-81 years). The procedures included KLAL (15 cases), sectoral (only 180°) KLAL (3 cases), and combined KLAL and conjunctival-limbal autograft (1 case). The mean (SD) follow-up was 17.3 (7.2) months (range, 7-31 months). The mean (SD) epithelial healing time was 8.4 (4.0) days (range, 5-22 days), excluding 1 patient with a history of squamous cell carcinoma and extensive eyelid tissue loss whose epithelial defect never healed and was ultimately considered a failure at 2 months after surgery. The mean best-corrected visual acuity improved from 20/900 (range, 20/200-20/3200) preoperatively to 20/300 (range, 20/80-20/1600) postoperatively (P < .001), with 15 cases (78.9%) showing an improvement of 2 or more Snellen lines.

At last follow-up, 15 of the 19 eyes had maintained a stable ocular surface. There were a total of 4 limbal transplantation failures, 3 of which were due to rejection in younger aniridic patients (aged 19, 25, and 41 years). The other failure was due to inadequate eyelid closure in a patient with a history of extensive eyelid resection and radiation-induced limbal stem cell deficiency. The mean time of failure was 7.5 months (range, 2-12 months), and all failures occurred in the first year after surgery. Cumulative survival of the ocular surface transplantation (ocular surface stability) was 76.9%.

There were no intraoperative or immediate postoperative complications. Of note, there were no graft detachments or major displacements. Two patients developed a conjunctival inclusion cyst near the posterior edge of the graft that diminished over time. Preoperatively, 11 patients had a history of glaucoma, of whom 6 had previously undergone surgery (5 tube shunts, 1 diode laser). Postoperatively, the intraocular pressure increased in 7 patients, all of whom had preexisting glaucoma. In all cases, the pressure was controlled medically.

### COMMENT

Limbal stem cell transplantation is currently the main surgical treatment for visually disabling limbal stem cell deficiency. Keratolimbal allograft is typically considered in patients with bilateral disease who are also candidates for systemic immunosuppression. The surgical procedure for KLAL consists of 2 parts, the donor tissue dissection and the recipient eye surgery. Technically, preparing the donor tissue by lamellar dissection is one of the challenging aspects of the surgical procedure. A number of different techniques have been described to facilitate the donor tissue dissection. One of the earlier techniques was to dissect the limbal tissue from a whole globe.4 Dissecting limbal grafts from a corneoscleral tissue that is not stabilized often requires an assistant to hold the tissue.5 One strategy to stabilize the tissue is to use an artificial anterior chamber and dissect the tissue manually or by a microkera-tome.6 Mannis et al7 described using a standard silicone orbital sizing sphere and three 25-gauge needles to fix the corneoscleral button. Adlave and Wong8 used cyanoacrylate glue to secure a corneoscleral rim to a disposable acrylic sphere. Meisler et al9 presented a device to secure corneoscleral buttons using suction. More recently, Lim et al10 have described a technique similar to ours using cyanoacrylate glue to secure 180° corneoscleral crescents to a flat surface.

The dissection technique described here may offer some potential advantages. First, when harvesting the donor tissue, only conjunctiva and some Tenon capsule is taken and typically there is minimal scleral tissue in the graft. As a result, the transplanted tissue is much thinner compared with tissue from other techniques described to date. A thin graft that is not elevated above the cornea and sclera is desirable in part because it avoids having a large step off, which can occasionally lead to dellen or epithelial healing problems. Likewise, the donor sclera is often not visible after transplantation as with many other techniques, and over the long term, the conjunctival edge blends nicely with the surrounding conjunctiva (Figure 3). Finally, a thin graft can be easily secured to the recipient cornea and sclera using only fibrin glue.

Fibrin glue is a tissue adhesive based on the 2 biological components of the coagulation cascade, fibrinogen and thrombin. On contact with each other, thrombin converts fibrinogen to fibrin, which is rapidly polymerized. This leads to hemostasis and tissue adhesion. The use of fibrin glue in ophthalmology dates back more than 20 years.12 It is used commonly in conjunctival surgery to secure a conjunctival autograft or amniotic membrane graft after pterygium excision. Likewise, it has been used for other ocular surface reconstructive procedures, particularly amniotic membrane transplantation.13-17 The advantages of using fibrin glue in ocular surgery are well known and include a decrease in operative time and postoperative pain.18-20 Postoperatively, not having sutures is also advantageous because sutures can induce inflammation and occasionally provide a nidus for infection or neovascularization.
To our knowledge, there is only 1 published report on the use of fibrin glue for limbal stem cell transplantation; however, that study involved conjunctival limbal autografts mainly after pterygium excision and included only a few cases of total limbal stem cell deficiency. While the results in this study involved KLAL, we have used fibrin glue successfully to secure conjunctival limbal autografts for unilateral cases of limbal stem cell deficiency (H.K.P., Jeffrey D. Welder, BA, N.N., and A.R.D., unpublished data, August 10, 2006). Actually, one of the patients in our series underwent combined KLAL and conjunctival-limbal autograft for extensive limbal and conjunctival deficiency.

Because the components of fibrin glue are often derived from pooled human plasma, there is a theoretical risk of human disease transmission. However, to our knowledge, there have not been any documented cases of disease transmission despite extensive use of these products in various surgical fields. Thus, the risk appears to be minimal. It should also be mentioned that the use of fibrin glue for ocular surface procedures is considered an off-label use of this product.

A feature of our technique, which is modeled after the technique described by Croasdale et al, is the use of three 180° donor tissues. This provides more stem cells for the recipient eye compared with a standard 360° technique. The extra graft also provides more tissue to cover the ocular surface in patients with extensive conjunctival deficiency. It results in a larger corneal surface untouched by the limbal graft and may provide improved vision or a better surface for contact lens wear. In this technique, it is crucial to place the three 180° donor tissues immediately adjacent to one another to avoid any gaps between tissues, which could allow conjunctival invasion onto the surface. This overcomes the disadvantage of using only two 180° grafts like Lim et al, which can leave gap areas between the donor tissues.

Technically, the procedure described here may be considered a conjunctival-limbal allograft given the generous amount of conjunctiva included in the graft. However, because the transplanted conjunctiva is not always viable and the procedure is used primarily to reconstruct the limbus, we have chosen to keep the name KLAL, in part because it is the more familiar term in the literature.

An important technical consideration in this procedure is the posi-
tion of the donor and host conjunctival edges. It is recommended that the host and donor conjunctiva be well approximated to each other or to have the host conjunctiva slightly pulled over the donor conjunctiva. This avoids the possibility of host conjunctival growth under the grafts while also preventing the formation of inclusion cysts. Two patients in this study developed an inclusion cyst, both of which diminished with- out intervention.

Other than the use of fibrin glue intraoperatively, our postoperative management of patients is the same as standard KLAL. In particular, the patients must receive systemic immunosuppression to prevent immune rejection of the allograft tissue. Currently, we use the Cincinnati protocol, with a regimen consisting of systemic steroids, tacrolimus, and mycophenolate. Comanagement with an organ transplant team with experience in the use of immuno suppressive medications is highly recommended for this purpose.

Postoperatively, 78.9% of cases showed a significant improvement of visual acuity from baseline, which is comparable to the rates of 30% to 67% reported by previous studies. It is important to note that the final visual acuities in many patients may be limited by optic nerve deficits.

Graft survival rates for KLAL have been reported to be 33% to 84%. In our study, the graft survival rate was 76.9% after follow-up of 31 months (Figure 4). Long-term studies are still needed to determine the ultimate fate of the transplanted limbal grafts.

In summary, a technique for dissecting limbal allografts and securing them to the recipient limbus using fibrin glue has been described. The early results appear comparable to previous reports. The use of fibrin glue in KLAL can potentially enhance surgical efficiency and improve patient comfort postoperatively.

Submitted for Publication: January 10, 2010; final revision received April 9, 2010; accepted April 13, 2010.

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Financial Disclosure: None reported.


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