Therapeutic Penetrating Keratoplasty Using Full-Thickness Gamma-Irradiated Sterile Cornea Tissue (VisionGraft®)

Gustavo A. Corrales, M.D.1
Sina J. Sabet M.D., F.A.C.S.2
Bhaskar Kallakury, M.D.3
Eileen Rusnock4
Esen K. Akpek, M.D.5

Keywords: acellular cornea, sterile cornea, VisionGraft®, infectious keratitis, penetrating keratoplasty, corneal transplant

Abstract

Purpose: To report the ultrastructural and biological features of a gamma-irradiated sterile cornea based on its successful use in a therapeutic penetrating keratoplasty for a case of severe fungal keratitis.

Methods: A commercially available acellular, sterile cornea was examined by electron microscopy and immunohistochemical staining after in-vivo explantation in a 50 year-old diabetic male who presented with a culture positive Aspergillus keratitis that did not respond to maximal medical therapy. A penetrating keratoplasty was performed for tectonic and therapeutic purposes using a commercially available acellular, sterile cornea (VisionGraft®). Five months later, a second penetrating keratoplasty with fresh tissue was performed. This is the first known ultrastructural examination of the VisionGraft® with electron microscopy after in-vivo explantation.

Results: Examination of the acellular cornea showed no infiltration of leukocytes or antigen-presenting cells. There was also no interstitial edema. The graft was partially covered with epithelium.

Conclusion: The commercially available acellular, sterile cornea can be considered in cases where an emergency penetrating keratoplasty must be performed for tectonic or therapeutic purposes. Its advantages over fresh corneal tissue appear to be: availability, sterility, long shelf-life at room temperature, and a decreased risk of allosensitization that minimizes immune rejection of a future corneal graft for visual rehabilitation.

CASE REPORT

A diabetic 50 year-old Hispanic male was referred for a non-healing corneal ulcer. His problem had started with a mild trauma to the eye with vegetative matter while mowing the lawn about a month prior of being referred to us. What started as a foreign body sensation after the trauma, rapidly progressed to worsening pain, increasing inflammation,
and decreased vision. He had no history of contact lens wear or previous eye problems. On presentation his visual acuity was hand-motions. Slit-lamp examination of the left eye demonstrated a central mid-stromal ring infiltrate, with an overlying epithelial defect measuring 1.5 mm in diameter as well as several satellite infiltrates. The left eye was promptly cultured and the patient was started on fortified vancomycin (25mg/ml), tobramycin (15mg/ml) and voriconazole 1% every hour due to the high suspicion of a fungal or polymicrobial infection. The initial corneal culture was negative for any microorganisms. However, a subsequent culture grew Aspergillus flavus. Despite aggressive medical therapy, including oral itraconazole, hourly topical voriconazole 1%, and topical fortified antibiotics, the patient’s condition deteriorated over the next few weeks. He developed a hemorrhagic hypopyon, iris neovascularization, and corneal thinning with impending perforation or microperforation. The hypopyon increased in size and became organized, raising the suspicion of intraocular invasion (figure 1). An emergency surgical approach was selected and the use of a sterile acellular cornea versus a fresh donor cornea was discussed with the patient. Owing to sterility and lack of antigenicity it was recommended that the transplantation be performed using VisionGraft®. The patient provided an informed consent. A therapeutic penetrating keratoplasty was performed using an 8.75 mm VisionGraft® cornea, oversizing the graft by 0.25mm. The donor was sutured in place with interrupted 10-0 nylon sutures. Aqueous fluid sample was sent for culture and an intracameral injection of 0.2 ml of 100 mcg/100 microliter voriconazole was given at the end of the procedure. In the immediate postoperative period, the patient developed a hyphema due to intraoperative bleeding from the iris. The intraocular pressure increased to 40 mmHg. Despite maximal medical therapy the patient had to be taken back to the operating room for an anterior chamber wash-out. Following the anterior chamber wash-out the intraocular pressure remained normal for the rest of the follow-up. The topical antifungals were continued for three weeks after the keratoplasty. The VisionGraft® integrated very well with the recipient bed. The graft-host junction healed well, with normal epithelialization. However, after about two weeks a central epithelial defect that slowly gained the appearance of a sterile neurotrophic defect was noted. This central defect persisted despite aggressive use of lubricants and a bandage contact lens (figure 2). Although the graft remained clear, a dense, white cataract developed and necessitated a triple procedure with cataract extraction, intraocular lens implantation, and a full thickness penetrating corneal transplantation using fresh donor tissue for visual rehabilitation.

Results: Light microscopy of sections stained with hematoxylin and eosin showed corneal tissue with complete absence of the endothelium. The epithelium was present in the periphery of some sections, but largely absent over the rest (figure 3). The stroma was predominantly devoid of keratocytes. However, in the periphery of the graft, a few stromal keratocytes were visible with irregular nuclei. The lamellae of the stroma appeared regular, but lacked normal artifactitious clefting throughout most of the sections. Bowman’s and Descemet’s membranes appeared normal, but no endothelial cells were present. Keratocytes were largely absent, except for some irregular cells in the periphery of the graft. No inflammatory cells were present. Immunohistochemical staining with neurofilament for axonal nerve fibers, and S-100 for Schwann cells, were both negative. Electron microscopy showed the maintenance of the typical hexagonal arrangement of the corneal stromal collagen fibers arranged in lamellae. There was evidence of cross-linking

Figure 1

Figure 2

Figure 3

Figure 4

Figure 5

Figure 6

Figure 7
trauma emergent penetrating keratoplasties are performed to remove the offending infection (therapeutic) or restore the integrity of the eye (tectonic).15

Grafts following emergency keratoplasty are more likely to fail and suffer immune rejections compared to non-emergency keratoplasties. Maier et al found that the failure risk for emergency keratoplasties is equivalent to scheduled high risk penetrating keratoplasties.23 The clear graft survival at four years was 67.9% for emergency keratoplasties, 70.2% for high-risk keratoplasties and 86.9% for non-high risk keratoplasties. Another retrospective study by Ang et al.24 reviewed the outcomes of tectonic keratoplasties, finding that the Kaplan-Meier probability for survival at 10 years was 44.2% for penetrating keratoplasties performed for tectonic purposes. Active corneal inflammation and recipient graft sizes ≥9mm were significant risk factors for graft failure.

As in high risk keratoplasties, several approaches have been suggested to improve the outcome of emergency keratoplasties, including the use of systemic immunosuppression15,25 and— if possible— waiting until the inflammation has subsided before performing the corneal transplant.15,26 Systemic immunosuppression is usually not effective in preventing an allograft rejection in these grafts.21 Additionally well-known side effects make them less than ideal for some patients.

We herein introduce a new approach to emergency keratoplasties that has the potential to improve outcome of future grafts performed for vision rehabilitation following the initial emergency situation. The rationale for using a sterile acellular cornea in emergency situations with active inflammation is that in these cases a penetrating keratoplasty is performed for therapeutic reasons, even though it is well-known that these grafts have a high risk of rejection and failure if fresh tissue is used. These patients may need to undergo a second or third corneal transplant, which in turn have a progressively increased risk of failure.

Glycerin-preserved corneas can be used in emergency procedures. However, they are not sterile, may not be clear initially, and are difficult to handle given their thick and rubbery texture, making them difficult to handle. Preservation in a -78°C freezer may be necessary to obtain transparent and pliable glycerol-preserved corneas.21 VisionGraft® sterile cornea is a sterile gamma-irradiated human donor corneal tissue that can be confirmed in future studies with HLA typing of these keratocytes and comparing them to the host and donor haplotypes. Peripheral epithelial growth was also present, although quite modest. This can perhaps be explained by the lack of axonal regeneration to provide neurotrophic support. It is not clear if the VisionGraft® can support axonal regeneration, or whether the absence of such fibers in this case was simply due to insufficient time for regeneration and host factors, like diabetes. Previous cases reporting the use of VisionGraft® tissue have not had difficulty sustaining healthy epithelium.

In conclusion, VisionGraft® sterile cornea should be considered in lieu of fresh donor corneas, cryo-preserved, or glycerin-preserved tissues for emergency tectonic full-thickness keratoplasty because of its availability, sterility, ease of handling, and lack of immunogenicity.
References


Corresponding Author:
Gustavo Corrales, MD
Tel: 703-532-0728
gcorrales@novavcs.com

TBI/Tissue Banks International
TBI Corporate
800-858-2020
www.tbionline.org