Microkeratome Versus Femtosecond Laser Predissection of Corneal Grafts for Anterior and Posterior Lamellar Keratoplasty

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Purpose: To compare 2 different techniques for predissection of human anterior and posterior lamellar corneal grafts for eye bank storage.

Methods: A mechanical microkeratome (group 1, N = 5) and a femtosecond laser (group 2, N = 5) were used to dissect intended 350-μm-deep lamellar planes in deepithelialized donor corneas mounted on an artificial anterior chamber. These corneas were replaced in Optisol GS at 4°C postoperatively and examined 2 days later to simulate a clinical scenario. Ultrasonic pachymetry of corneal lamellar sections was measured before and after separation of the lamellar grafts. Group 1 sections were separated by the mechanical microkeratome, whereas group 2 sections were manually separated 2 days after laser dissection. Endothelial cell viability was evaluated in posterior grafts.

Results: Total corneal thicknesses immediately before dissection were 559 ± 61 (group 1) and 578 ± 79 μm (group 2; P = 0.46). Immediate postdissection anterior and posterior graft thicknesses were 361 ± 60 and 203 ± 74 μm (group 1), respectively. Achieved anterior and posterior graft thicknesses 2 days later were 282 ± 34 and 413 ± 35 μm (group 1) and 324 ± 112 and 397 ± 51 μm (group 2), respectively. Percentage of devitalized endothelial cells were 3.4% ± 1.6% (group 1) and 1.6% ± 1.2% (group 2; P = 0.35).

Conclusions: Centralized predissection by both techniques, cold storage, and shipping by air mail results in viable grafts without significant endothelial cell loss 2 days later.

Key Words: lamellar, cornea, transplant, anterior, posterior, endothelium, microkeratome, femtosecond, laser

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brought to room temperature, the anterior and posterior grafts were separated, and each section was remeasured by ultrasound pachymetry while mounted on the artificial anterior chamber, bathing both surfaces in Optisol.

Endothelial cell damage after graft harvest was assessed by staining with trypan blue 0.25% (Sigma-Aldrich, St. Louis, MO) and alizarin red S 0.2% (Sigma-Aldrich). In our experimental design, we chose to not separate the laser-cut sections until day 2 (>48 hours later), to reduce the exposure time of the stromal surfaces to media to limit induced swelling. However, the laser device did not show any significant difference from the steel blade instrument in terms of accuracy and reproducibility after the 2-day postdissection storage period. Although the laser cut is known to have a finer precision and accuracy than the mechanical microkeratome, after 2 days of postdissection swelling in Optisol media, the difference disappeared.

On closer examination of the observed changes in lamellar thickness of the mechanical microkeratome–cut sections on day 2, there appeared to be significant thinning of the anterior lamellar sections ($P = 0.043$). In contrast, there was also significant swelling of the posterior sections ($P = 0.043$), despite reapposition of the anterior and posterior sections while in storage. However, these findings are in agreement with previous reports of human corneal swelling in Optisol GS corneal storage media. Namely, it has previously been reported that removal of the epithelium before storage results in increased stromal hydration. Also, it is known that, although hydration of the stroma increases during swelling through the posterior surface, it paradoxically decreases during swelling through the anterior surface, in agreement with our own experimental findings.

Because we did not separate the laser-cut lamellar sections immediately after the procedure, we could not compare the swelling at day 2 to that at day 0. However, average total corneal thickness did increase similarly by the end of the study period, using both instruments (mechanical microkeratome [group 1], 559 to 695 μm; laser [group 2], 578 to 721 μm).

Although it seems that the femtosecond laser offers results comparable to those of its mechanical microkeratome counterpart, there still exist concerns that will need to be addressed before its clinical implementation for lamellar keratoplasty. First, despite centralization of the unit for eye

**TABLE 1.** Total and Lamellar Corneal Thicknesses on Days 0 and 2 After Microkeratome Dissection (Mean ± SD, μm)

<table>
<thead>
<tr>
<th>Microkeratome</th>
<th>Day 0</th>
<th>Day 2</th>
<th>$P$ (Day 0 vs. 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mechanical (N = 5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>559 ± 61</td>
<td>282 ± 44</td>
<td>0.043</td>
</tr>
<tr>
<td>Anterior</td>
<td>361 ± 68</td>
<td>282 ± 44</td>
<td>0.043</td>
</tr>
<tr>
<td>Posterior</td>
<td>203 ± 74</td>
<td>413 ± 35</td>
<td>0.043</td>
</tr>
<tr>
<td>Laser (N = 5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>578 ± 79</td>
<td>324 ± 112</td>
<td></td>
</tr>
<tr>
<td>Anterior</td>
<td>324 ± 112</td>
<td>397 ± 51</td>
<td></td>
</tr>
<tr>
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**DISCUSSION**

Here we describe techniques for predissection of donor tissue for anterior and posterior lamellar keratoplasty in which cornescleral sections are mounted onto an artificial anterior chamber and stromal dissection is accomplished using either a mechanical microkeratome or femtosecond laser. In this study, after 2-day storage in cold Optisol, the endothelial cell loss associated with the predissections seemed low and comparable to that previously described immediately after dissection. In our experimental design, we chose to not separate the laser-cut sections until day 2 (>48 hours later), to reduce the exposure time of the stromal surfaces to media to limit induced swelling. However, the laser device did not show any significant difference from the steel blade instrument in terms of accuracy and reproducibility after the 2-day postdissection storage period. Although the laser cut is known to have a finer precision and accuracy than the mechanical microkeratome, after 2 days of postdissection swelling in Optisol media, the difference disappeared.

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Although it seems that the femtosecond laser offers results comparable to those of its mechanical microkeratome counterpart, there still exist concerns that will need to be addressed before its clinical implementation for lamellar keratoplasty. First, despite centralization of the unit for eye
banking, it is still significantly more costly to both purchase and maintain than the classic microkeratome. Second, it has been shown recently that, in current form, the Intralase femtosecond laser produces posterior grafts that contain imperfect ridges in the periphery of the produced sections. However, these same authors state that new hardware and software are currently in development to overcome this problem.

Despite these concerns, however, we are optimistic about eye bank implementation of the femtosecond laser. The posterior corneal sections obtained here were achieved using close to the deepest settings available with both instruments. In some cases, the thickness achieved after 2 days of cold storage may be more than is desired. Using hardware modifications currently in development, the femtosecond laser may be able to cut much thinner posterior grafts more accurately and reproducibly than its mechanical microkeratome counterpart.

Using modern microkeratome technology, we propose that, at the eye bank level, one donor cornea may be divided into at least 2 transplantable lamellar grafts. Here we showed that centralized predissection, cold storage, and shipping by airmail results in viable grafts without significant endothelial cell loss 2 days later.

REFERENCES