Microkeratome Cut Time as a Variable in Endothelial Keratoplasty Graft Thickness

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ABSTRACT

Purpose: To evaluate the effect of the time used to pass the microkeratome through the cornea during the production of a DSAEK graft on the thickness of the posterior lamellar corneal graft.

Methods: Twelve human corneoscleral discs were used to evaluate the effect of microkeratome (Moria CB, Antony, France) cut time on post-sectioned corneal lamellar thicknesses. Sectioning was completed using a microkeratome equipped with three microkeratome head sizes (250, 300, and 350 µm) utilizing two different cut times (1 or 6 seconds). Eye bank excised corneas with scleral rims were mounted on a Moria artificial anterior chamber and the intrachamber pressure was increased to >90 mmHg. The Moria DSAEK microkeratome was used to prepare the posterior lamellar graft tissue. Pachymetry was performed before and after construction of the graft tissue.

Results: All corneas sectioned over a 1 second period produced thicker residual stromal beds compared with 6 second sectioning times. The difference between the mean residual stromal bed thickness for all head sizes was 37 ± 50 µm thicker than intended for 1 second cuts and 41 ± 32 µm thinner than intended for 6 second cuts (P = 0.01). The longer pass always resulted in a thicker corneal flap. The mean total difference in corneal flap thickness between a 1 second cut time and a 6 second cut time for all 3 microkeratome head (250, 300 and 350 µm) sizes was 77.7 µm.

Conclusions: This pilot study provides proof of principle that microkeratome cut time (transit time) is an important variable, which affects graft thickness, in endothelial keratoplasty graft preparation.

Key Words: Descemet-stripping automated endothelial keratoplasty (DSAEK), endothelial keratoplasty, microkeratome cut time, pachymetry

The surgical management for endothelial failure has traditionally required penetrating keratoplasty (PK). PK entails prolonged postoperative care and is complicated by astigmatism, higher-order aberrations, and intensive suture management. Alternatively, endothelial keratoplasty (EK) selectively replaces damaged corneal endothelium allowing faster visual recovery, shorter postoperative management time, and possibly decreased risk of graft rejection.

EK was first introduced by Melles et al1 who described manual dissection and replacement of the posterior lamellae. Terry and Ousley2 refined the procedure by changing the instruments of trephination and named the procedure deep lamellar endothelial keratoplasty (DLEK). Further refinements included a stripping method for removal of the recipient’s endothelium and Descemet membrane3 as well as automated microkeratome preparation of the donor tissue4,5 which became known as Descemet-stripping automated endothelial keratoplasty (DSAEK). DSAEK enhanced postoperative visual outcomes by providing smoother recipient and donor tissue surfaces compared with earlier EK techniques.

Pre-cut donor tissue for DSAEK is now commonly supplied by eye banks. Processing posterior lamellar tissue in the eye bank eyes involves a mechanical microkeratome and artificial anterior chamber system for graft preparation. Although pre-cut tissue improves the efficiency of the DSAEK procedure, the predictability of donor tissue thickness has been limited.6,7 Two important variables purported to influence donor cornea and lamellar thicknesses in microkeratome sectioning of the cornea include

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Acknowledgments: The authors thank David Folden and Colin Brown for their work on this project and Rachel Folden for data analysis and manuscript preparation and Ann Holleschau for statistical guidance.

Funding/Support: Supported by a Grant from the Minnesota Lions Club and Research to Prevent Blindness.

International Journal of Eye Banking • vol. 5 no. 3 • December 2017 © 2017 Eye Bank Association of America. All rights reserved
intrachamber pressure\textsuperscript{8,9} and microkeratome cut time, also called microkeratome transit time.\textsuperscript{10,11} The purpose of this pilot study is to provide proof of principle and preliminary data on microkeratome cut time and its influence on post-sectioned corneal lamellar thickness. We provide data utilizing two different cut times (1 and 6 seconds) and three microkeratome head sizes (250, 300, and 350 µm) while maintaining a constant intrachamber pressure.

**MATERIALS AND METHODS**

**Grouping of the Corneas**

Twelve human corneoscleral discs not suitable for transplantation were processed and stored in Optisol GS (Bausch & Lomb Surgical, Irvine, CA) at 2 to 6° C and obtained from the Minnesota Lions Eye Bank, St. Paul, MN. All corneoscleral discs were obtained < 4 weeks postmortem.

The corneas were subdivided into two groups of six corneas each according to microkeratome cut time. The two groups were designated Group A (1 second) and Group B (6 seconds). The second group was also subdivided into 3 groups of two corneas each according to microkeratome head size (250, 300, and 350 µm) and microkeratome cut time (1 and 6 seconds). The subgroups were designated subgroup A1 (250 µm and 1s), subgroup B1 (250 µm and 6s), subgroup A2 (300 µm and 1s), subgroup B2 (300 µm and 6s), subgroup A3 (350 µm and 1s), and subgroup B3 (350 µm and 6s).

**Tissue Preparation Technique and Pachymetry**

All tissue preparation was performed by a single cornea trained surgeon (SK) to maintain consistency. Corneoscleral discs were mounted with non-toothed forceps, endothelial side down on the Moria ALTK microkeratome system (Moria/Microtek, Doylestown, PA) and manually locked into position. The epithelium was removed. Intrachamber pressure was maintained at >90 mmHg through an intravenous (IV) infusion of balanced saline solution (BSS), which was hung 123 cm above the work station from an IV line pole. After 90 seconds, the anterior chamber pressure was measured using a Barraquer tonometer. Central corneal thickness (CCT) was measured using an ultrasonic pachymeter (Sonogauge, Cleveland, OH), taking the average of three readings for each central corneal measurement.

The corneoscleral discs were placed in the artificial anterior chamber as indicated above. Ultrasonic pachymetry measurements were performed on the corneoscleral discs prior to sectioning and after intrachamber pressure was established (90 seconds). Corneal sectioning was completed using a Moria CB microkeratome (Moria, Antony, France), equipped with a 250, 300, and 350 µm head. Movement of the microkeratome was performed with the surgeon’s right hand in a right-to-left pass. Each head was used to cut an anterior corneal flap on two different corneoscleral discs over a 1 second (n = 6) or 6 second (n = 6) time interval. The microkeratome transit time was measured using a timer and a verbal countdown. Following the keratectomy, the central thickness of the residual posterior lamellar bed was measured. Anterior corneal flap thickness was determined by subtracting the ultrasonic pachymeter derived residual lamellar bed thickness measurement from the total corneal thickness.

**Statistical Analysis**

Statistical analysis of the mean difference between the residual stromal bed thickness compared with the intended stromal bed thickness for all head sizes was performed using the paired t test (Instat, GraphPad Software, San Diego, CA).

**RESULTS**

All procedures and testing was performed without incident or complication. Pachymetry was performed centrally on all corneal tissue and each value was determined by averaging three measurements.

**Pachymetry and Microkeratome Head Size**

Central corneal thickness measurements, as relate to microkeratome head size, are summarized in Table 1. The mean corneal flap thickness for the 250 µm head size was 200 ± 81 µm cut over 1s and 300 ± 59 µm cut over 6 s. The residual stromal bed thickness for the 250 µm head size was 406 ± 101 µm and 324 ± 133 µm cut over 1 s and 6 s respectively. The difference in residual stromal bed thickness compared with the intended thickness was 51 ± 81 µm and -50 ± 59 µm for 1 s and 6 s cuts respectively.

<table>
<thead>
<tr>
<th>Microkeratome Head size (µm)</th>
<th>Mean CCT Before Sectioning</th>
<th>Mean Stromal Bed Thickness After Sectioning</th>
<th>Intended Stromal Bed Thickness</th>
<th>Difference Between Actual versus Intended Stromal Bed Thickness</th>
<th>Mean Cornal Flap Thickness After Sectioning</th>
<th>Difference Between Actual versus Intended Cornal Flap Thickness</th>
</tr>
</thead>
<tbody>
<tr>
<td>250</td>
<td>605 ± 19 / 624 ± 74</td>
<td>406 ± 101 / 324 ± 133</td>
<td>355 ± 19 / 374 ± 74</td>
<td>51 ± 81 / 50 ± 59</td>
<td>200 ± 81 / 300 ± 59</td>
<td>-51 ± 81 / 50 ± 59</td>
</tr>
<tr>
<td>300</td>
<td>617 ± 61 / 577 ± 37</td>
<td>317 ± 51 / 249 ± 6</td>
<td>317 ± 61 / 277 ± 37</td>
<td>-6 ± 10 / -27 ± 31</td>
<td>306 ± 10 / 327 ± 31</td>
<td>6 ± 10 / 27 ± 31</td>
</tr>
<tr>
<td>350</td>
<td>639 ± 95 / 588 ± 63</td>
<td>355 ± 93 / 152 ± 60</td>
<td>289 ± 95 / 238 ± 43</td>
<td>66 ± 3 / 46 ± 3</td>
<td>284 ± 3 / 396 ± 3</td>
<td>66 ± 3 / 46 ± 3</td>
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</table>
The mean corneal flap thickness for the 300 µm head size was 306 ± 10 µm cut over 1 s and 327 ± 31 µm cut over 6 s. The residual stromal bed thickness for the 300 µm head size was 311 ± 51 µm and 249 ± 6 µm cut over 1 s and 6 s respectively. The difference in residual stromal bed thickness compared with the intended thickness was -6 ± 10 µm and -27 ± 31 µm for 1 s and 6 s cuts respectively.

The mean corneal flap thickness for the 350 µm head size was 284 ± 3 µm cut over 1 s and 396 ± 3 µm cut over 6 s. The residual stromal bed thickness for the 350 µm head size was 355 ± 93 µm and 192 ± 60 µm cut over 1 s and 6 s respectively. The difference in residual stromal bed thickness compared with the intended thickness was 66 ± 3 µm and -46 ± 3 µm for 1 s and 6 s cuts respectively.

The mean total difference in corneal flap thickness between a 1 second cut time and a 6 second cut time for all 3 microkeratome head (250, 300 and 350 µm) sizes was 77.7 µm, which is demonstrated by a total spread of 77 µm in figure 1.

**Figure 1**: Mean Difference Between Measured and Intended Corneal Flap Thickness as a Function of Cut Time

DISCUSSION

EK is an important surgical technique in the management of endothelial failure; DSAEK involves less intensive postoperative management and faster visual recovery compared with PK and earlier EK techniques. Although pre-cut eye bank tissue has improved DSAEK efficiency, the graft thickness of microkeratome-prepared graft tissue has shown variability. Consistency of graft tissue thickness and high endothelial cell counts in DSAEK is important to the surgeon. Variability in donor tissue can make the procedure technically more difficult and alter outcomes. Thinner than expected grafts can be difficult to control, while thicker than expected grafts can result in excessive trauma during graft insertion. Mishandling of the graft in either situation can corneal endothelial cell loss that results in reduced postoperative endothelial cell counts. This paper provides data on microkeratome cut time and resultant post-sectioned corneal lamellar thickness.

Our study consistently found that the slower, 6 second cut time resulted in an increased corneal flap thickness, compared to the shorter 1 second cut time. Thus, it reasonably follows that the residual stromal bed was thinner for the 6 second cut time, compared with 1 second transit time for each microkeratome head. A statistically significant difference was found in the mean stromal bed thickness comparing 1 second (37 ± 50 µm thicker than intended) and 6 second (41 ± 32 µm thinner than intended) microkeratome cut times (P = 0.01), with a mean difference of 78 µm. Behrens et al described a cut rate of 1 mm / s using a 135 µm microkeratome head size with a constant pressure of 65 mmHg in a series of pig eyes. They found the mean central corneal flap thickness to be 135 µm (SD, 35 µm). A lower intrachamber pressure may explain why a relatively thicker stromal bed was attained with a slower microkeratome cut time compared to the present study. They also found flaps to be thinner at the beginning of the pass and attributed this to an initial faster cut rate although this was not substantiated. Other studies have described variability in corneal lamellar thickness after sectioning with an automated microkeratome and artificial anterior chamber system although no information regarding the speed of the pass was given.

Intrachamber pressure has also been shown to be important in donor cornea and lamellar thicknesses in microkeratome sectioning of the cornea in an artificial anterior chamber system. Thinner donor cornea and posterior graft thicknesses have been observed with higher intrachamber pressures.

Variables affecting microkeratome-assisted flap thickness in LASIK include the microkeratome model, the surgeon specific differences in their procedure, and mean preoperative pachymetry. Thinner corneas are associated with thinner flaps and thicker corneas are associated with thicker flaps. These variables may also play a role in the preparation of DSAEK donor tissue. Alternatively, femtosecond laser-assisted flap creation in LASIK produces a narrower range of flap thicknesses and smaller standard deviations compared with microkeratome-assisted flaps. The use of microkeratome prepared endothelial keratoplasty tissue is still the most common tissue preparation method for DSAEK. How-
ever, recent studies have used femtosecond laser technology protocols for donor tissue preparation in EK.  

In conclusion, consistent DSAEK graft thickness aids the surgeon in producing optimal surgical results. This study provides preliminary data and proof of principle that microkeratome cut time (transit time) is an important variable in endothelial keratoplasty graft thickness, although larger studies are warranted to verify these findings.

REFERENCES