Published in final edited form as:

Cornea. 2010 May ; 29(5): 523–527. doi:10.1097/ICO.0b013e3181c11e5d.

Effect of incision width on graft survival and endothelial cell loss after DSAEK

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Abstract

Purpose—To assess the effect of incision width (5.0 and 3.2 mm) on graft survival and endothelial cell loss six months and one year after Descemet stripping automated endothelial keratoplasty (DSAEK).

Methods—One hundred sixty-seven subjects with endothelial decompensation from a moderate-risk condition (principally Fuchs' dystrophy or pseudophakic corneal edema) underwent DSAEK by two experienced surgeons. The donor was folded over and inserted with single point fixation forceps. This retrospective analysis assessed graft survival, complications, and endothelial cell loss, which was calculated from baseline donor and 6-month and 1-year postoperative central endothelial images evaluated by an independent specular microscopy reading center.

Results—No primary graft failures occurred in either group. One-year graft survival rates were comparable (98% vs. 97%) in the 5.0- and 3.2-mm groups, respectively ($P=1.0$). Complications included graft dislocation, graft rejection episodes, and elevated intraocular pressure and occurred at similar rates in both groups ($P \geq 0.28$). Pupillary block glaucoma did not occur in either group. Mean baseline donor endothelial cell density did not differ: 2782 cells/mm² in the 5.0-mm ($n=64$) and 2784 cells/mm² in the 3.2-mm ($n=103$) groups. Percent endothelial cell loss was $27 \pm 20\%$ ($n=55$) vs. $40 \pm 22\%$ ($n=71$; 6 months) and $31 \pm 19\%$ ($n=45$) vs. $44 \pm 22\%$ ($n=62$; 12 months) in the 5.0-mm and 3.2-mm incision groups, respectively (both $P < 0.001$).

Conclusions—One year after DSAEK, overall graft success was comparable for the two groups; however, the 5.0-mm incision width resulted in substantially lower endothelial cell loss at 6 and 12 months.

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Financial Disclosure:

Drs. Price have received travel grants from Moria (Antony, France).

Keywords

Descemet stripping automated endothelial keratoplasty; Descemet stripping endothelial keratoplasty; posterior lamellar keratoplasty; penetrating keratoplasty; DSAEK; DSEK; PLK; PKP; endothelial cell loss; graft survival

Descemet stripping automated endothelial keratoplasty (DSAEK) has become a preferred surgical treatment for corneal endothelial decompensation because it provides rapid visual recovery, uses a smaller wound size, minimizes induced astigmatism and, most importantly, better maintains globe integrity than penetrating keratoplasty (PKP).¹ However, since DSAEK is a relatively new procedure, little is known about the operative parameters that influence long-term graft survival and endothelial cell loss with DSAEK and how these parameters of performance compare with PKP.

We recently conducted a prospective study of DSAEK to assess graft survival and endothelial cell loss in comparison with a subset of patients from the Cornea Donor Study (CDS) and its ancillary study, the Specular Microscopy Ancillary Study (SMAS),^{2, 3} employing the same donor and recipient inclusion/exclusion criteria and follow-up examination procedures, and utilizing the same specular microscopy reading center for determination of the central endothelial cell density.^{3,4} The 1-year endothelial cell loss in the DSAEK eyes significantly exceeded that in SMAS PKP eyes (38% vs. 20%, $P < 0.001$).

Two sites participated in this study, with the principal variation in surgical technique being the incision width used for graft insertion (3.2 mm vs. 5.0 mm). A previous *ex vivo* study on donor eyes examining different methods of corneal graft delivery suggested that incision width has a significant effect on endothelial cell loss.⁴ This study from the same DSAEK data set⁵ examined whether the difference in incision width had a significant effect on 1-year graft survival and endothelial cell loss.

Methods

In this retrospective, interventional study, subjects were treated with DSAEK at Gorovoy Eye Specialists (Fort Myers, FL) or Price Vision Group (Indianapolis, IN) between June 2006 and September 2007. The University Hospitals Case Medical Center (Cleveland, OH) institutional review board approved the study. Each subject provided written informed consent to participate.

As in the CDS, eligible subjects had conditions of moderate risk for graft failure from endothelial decompensation (principally Fuchs' dystrophy or pseudophakic corneal edema).^{2, 3} Patients undergoing a regrant, or with two or more quadrants of stromal neovascularization, uncontrolled uveitis, uncontrolled glaucoma, previous placement of a glaucoma shunt, or fellow eye visual acuity $< 20/200$ were excluded. Only one eye per patient was included in the study. Eligible donor corneas were from 10–75 years old, with an eye bank-measured central endothelial cell density (ECD) of 2300–3300 cells/mm², death to surgery time of ≤ 5 days, and death to preservation time of ≤ 12 hours, if the body was refrigerated or eyes were iced, and ≤ 8 hours, if not. These donor eligibility criteria were the same as those used in the SMAS.^{2,3,6}

The Case Vision Research Coordinating Center (VRCC) developed data collection forms that paralleled those used in the CDS and the SMAS, provided them to participating eye banks and clinical sites, collected and analyzed the data, and audited both clinical sites during and at conclusion of the follow up period to verify all data.^{2, 3, 7}

Surgical technique

Two experienced surgeons (FWP and MG) performed the DSAEK procedures, as previously described.^{8, 9} If a cataract was present, a phacoemulsification procedure was performed before DSAEK; this had the added benefit of creating more space in the anterior chamber to safely position the graft. Patients with anterior chamber intraocular lenses were not included in this study.

The donor cornea was dissected with a microkeratome and associated artificial anterior chamber (model CB, Moria USA, Doylestown, PA). The artificial anterior chamber was pressurized with air when the 3.2-mm incision was used and with balanced salt solution when the 5.0-mm incision was used. The donor cornea was transferred to a punching system, and cut with a trephine, with a diameter of 8.25 to 9.0 mm. Surgery was performed on the recipient with topical anesthesia and monitored intravenous sedation. A 3.2-mm clear corneal temporal incision at one site or a 5-mm clear corneal or scleral tunnel temporal incision at the other site was made, and Descemet's membrane and endothelium were stripped from within the planned graft area. The donor cornea was folded into a "taco" configuration, with a drop of cohesive sodium hyaluronate viscoelastic on the inside to protect the endothelium, and inserted into the anterior chamber using single-point fixation forceps (both surgeons used Goosey and Ogawa forceps, Moria, Antony, France). The graft was unfolded and air was injected into the anterior chamber posterior to the graft. Three or four small, equally spaced incisions were placed anteriorly in the mid-peripheral recipient cornea down to the graft interface to help drain any entrapped fluid.

When the 3.2-mm incision was used, the anterior chamber was completely filled with air for one hour followed by partial air removal at the slit lamp before discharging the patient. When the 5-mm incision size was used, the anterior chamber was completely filled with air for eight minutes, most of the air was removed and replaced with balanced salt solution, a drop of homatropine 5% was instilled to prevent pupillary block, tobramycin/dexamethasone ointment was placed in the eye, and the patient was discharged after remaining face up for thirty minutes in the recovery room to allow the remaining small air bubble to press the graft against the recipient cornea.

Specular microscopy and ECD determination

Participating eye banks that provided donor corneas eligible for the study included Central Florida Lions Eye and Tissue Bank (Tampa, FL), Indiana Lions Eye and Tissue Transplant Bank (Indianapolis, IN), North Carolina Eye Bank (Winston-Salem, NC), Heartland Lions Eye Bank (St. Louis, MO), and SightLife (Seattle, WA); each had previously participated in the SMAS, and were familiar with image capture and transmission procedures.^{3, 7-10} As in the SMAS, after their specular microscopes had been again calibrated by the Case Specular Microscopy Reading Center (SMRC), the eye banks electronically submitted a single image of the central endothelium of each study donor cornea to the SMRC for ECD determination.^{3, 7-10} After SMRC calibration of the clinical specular microscope (Konan Medical Corp, Torrance, CA) or confocal microscope (Nidek Inc., Fremont, CA) at each site, three postoperative images of the central corneal graft endothelium were captured at the 6-month (range 5 to 7 months) and 12-month (range 9 to 15 months) study exams and transmitted to the SMRC for analysis. SMRC analysis procedures for donor and postoperative images were as previously described.^{3, 7-10}

Statistical Methods

Endothelial cell loss was calculated for each subject by subtracting postoperative ECD from baseline ECD, dividing by baseline ECD, and multiplying by 100. As in the SMAS, if the donor image could not be analyzed by the SMRC or was unavailable, the eye-bank-

determined ECD was utilized. All postoperative ECD data was derived from SMRC-determined ECD analyzed images. For normally distributed variables, descriptive statistics were reported as mean $\pm$ standard deviation and compared using a two-sample Student t-test. Otherwise, they were reported as median (25th and 75th quartile) and compared using the Wilcoxon rank-sum test, or they were expressed as a proportion and compared using Chi-square analysis or Fisher exact test, as appropriate. All reported P values were 2 sided, and P-values < 0.05 were considered significant. All statistical analyses were performed with SAS software (version 9.1, SAS Institute, Cary, NC).

Results

Sample size

A total of 175 subjects were originally enrolled in the study comparing a prospective series of DSAEK cases to a subgroup of the SMAS.⁵ For the purpose of this study, eight cases were excluded: 1) one eye was the fellow eye of a previously enrolled study subject; 2) one subject received a DSAEK to treat a previously failed DSAEK; and 3) in 6 eyes, the graft was inserted with a funnel glide. Thus, 167 eyes (167 subjects) were available for this retrospective analysis of folded graft insertion with forceps through different incision widths. Of the 167 DSAEK eyes, 134 (80%) were examined and found to have clear grafts at 1 year, 19 (11%) had clear grafts at 6 months and returned to their referring doctors, 6 (4%) were lost to follow up before the 6-month exam, 3 (1.8%) died, 1 (0.59%) withdrew, and 4 (2.4%) experienced a graft failure within the first 15 months. Of the 134 DSAEK subjects with clear grafts at 12 months, 107 (80%) had analyzable specular images, 4 (3%) had clear grafts but the 12-month postoperative image was not analyzable, and the remaining 23 (17%) had clear grafts but no image was obtained.

Sixty-four eyes were treated with a 5.0-mm incision width and 103 eyes with a 3.2-mm incision width. Comparable percentages of eyes in each group had analyzable 12-month specular images obtained (70% vs. 60%, respectively, $P=0.072$).

Donor and recipient characteristics

Donor characteristics are summarized in Table 1. The mean baseline ECD was comparable for the two sites ($p=0.97$). The 3.2-mm incision group had a mean death to preservation time 1.3 hours longer ($P=0.0034$), and a mean death to surgery time 0.6 days shorter ($P=0.0056$), and the mean donor age was 5 years older ($P=0.036$). The mean graft diameter was comparable for the two sites (8.9 vs. 8.8 mm, $P=0.30$).

Recipient characteristics are summarized in Table 2. The two sites had comparable proportions of males and females ($P=0.82$), comparable preoperative diagnoses ($p=0.47$), and comparable postoperative lens status ($P=0.45$). In the 5-mm incision group, the mean recipient age was 5 years younger ($P=0.021$), the racial distribution was slightly more diverse ($P=0.045$), and about one third of the subjects were phakic before undergoing DSAEK ($P<0.001$).

Graft success

The regrant rate was comparable for the two sites ($P=1.0$). None of the grafts experienced primary failure. Within the 5-mm incision group, one eye (1.6%) was regrafted to treat visually significant wrinkles in the graft. Within the 3.2-mm incision group, three eyes (2.9%) were regrafted, one to treat visually significant wrinkles in the graft, one because of unsatisfactory vision that was subsequently determined to be related to stromal haze in the recipient cornea, and one to treat graft decompensation caused by immunologic rejection.

Complications

The rate of postoperative complications was comparable at the two sites. Among the treated eyes in the 5-mm incision and 3.2-mm incision groups respectively, intraocular pressure spikes exceeding 25 mmHg occurred in 20% vs. 14% ($P=0.28$), immunologic graft rejection episodes occurred in 8% vs. 4% ($P=0.31$), and graft dislocation and re-bubbling occurred in 3% vs. 8% ($P=0.32$). No eyes in either group experienced pupillary block glaucoma.

Endothelial Cell Loss

Compared with the 3.2-mm incision width cohort, the 5.0-mm incision width cohort experienced significantly lower endothelial cell loss at 6 months ($27\pm 20\%$ [range 23% gain to 67% loss] vs. $40\pm 22\%$ [range 1 to 81% loss]) and at 12 months ($31\pm 19\%$ [range 25% gain to 70% loss] vs. $44\pm 22\%$ [range 4% gain to 80% loss], both $P<0.001$) (Table 3). Endothelial cell loss did not increase by a statistically significant amount between 6- and 12-months in either cohort ($P=0.35$ and $P=0.25$, respectively).

Discussion

The key finding in this study was that compared with insertion through a 3.2-mm wide incision, DSAEK graft insertion through a 5.0-mm wide incision resulted in significantly lower 6- and 12-month endothelial cell loss. Identifying such a simple technique variation that can reduce endothelial cell loss after the first year of this procedure is particularly important because endothelial decompensation is a leading cause of graft failure after standard PKP, and endothelial cell loss during this period is significantly higher with DSAEK.^{2, 5, 11} These 6- and 12-month results confirm for the first time in a clinical study of DSAEK predictions of significantly lower cell loss with a 5.0-mm incision width, compared with a 3.0-mm width using single-point fixation forceps, based on a laboratory model by Terry et al.⁴ Folding the graft through a smaller incision width likely causes more tissue compression and associated endothelial cell damage. Our findings are consistent with observations of increased 1-year cell loss with a smaller incision width (5-mm vs. 9-mm) for posterior lamellar keratoplasty and deep lamellar endothelial keratoplasty procedures.^{12, 13} Despite the significant difference in endothelial cell loss, 1-year graft survival was comparable for the two incision widths. Importantly, no primary graft failures occurred in either group. However, additional follow up is required to evaluate long-term graft survival rates.

The results of this study are specifically applicable to cases in which the graft is folded approximately in half, then grasped and inserted with single-point fixation forceps. Other methods of graft insertion include the suture pull-through method or pulling the graft into the anterior chamber through or over a glide.^{14–16} Recently, several different injector designs have been introduced to curl the graft and protect it from compressive forces as it goes through the incision; however, there are no comparative studies examining endothelial cell loss with these new injector designs, which are a more expensive option as single-use instruments.

This study had several limitations. The ideal study design would be a prospective trial randomizing on the basis of wound size and all other donor, recipient and operative parameters the same. In this study the DSAEK group was enrolled prospectively for a comparative study with a subset of PKP patients in the SMAS of the CDS, sharing similar donor and recipient criteria without regard to specifying all operative parameters in the DSAEK procedure for the two surgeons.⁴ Any operative differences and their impact on graft survival and endothelial cell loss were not known at the outset of the study. This data set thus afforded the opportunity to perform a retrospective analysis of the major difference

between the two surgical techniques, wound size. However, the investigators were not masked to wound size, eyes were not randomized on this basis, and a number of minor donor, recipient, and operative differences between the two groups that could have confounded the results arose. These differences included in the 5.0 mm incision group a shorter mean donor death to preservation time by 1.3 hours, mean donor death to use time 0.6 days longer, mean donor and recipient ages 5 years younger, use of balanced salt solution rather than air to pressurize the artificial anterior chamber used for donor dissection, a shorter complete air fill (8 vs. 60 minutes), and 25% of the DSAEK cases combined with cataract extraction and IOL implantation, whereas no triple procedures were performed in the 3.2-mm incision group.

However, previous studies suggest that these minor differences were unlikely to have had a significant effect on the outcomes. A study of 263 DSEK eyes showed that donor death to preservation time, donor death to use time, and recipient age were not significantly associated with 6-month endothelial cell loss.¹⁷ Although donor age was weakly correlated with 6-month cell loss in that earlier study, donor age and ECD were confounded.¹⁷ That earlier study also showed that clear corneal incisions were associated with lower cell loss than scleral tunnel incisions. In the current study, the 5.0-mm incision group included some scleral tunnel incisions, whereas the 3.2-mm incision group did not. So differences in incision construction would not account for the lower cell loss seen in the 5.0-mm incision group.¹⁷ To our knowledge, no studies have compared the duration of full air fill with endothelial cell loss. Finally, endothelial cell loss has been shown to be comparable for eyes undergoing triple vs. single procedures.¹⁸ Most importantly, in the current study baseline donor ECD was virtually identical for the two sites and recipient diagnosis was the same, an important risk factor for graft failure noted in the CDS.⁶

Despite these limitations, given the significant and greater than 10% difference in endothelial cell loss between the groups at both 6- and 12-month postoperative time points, we feel confident that the use of a 5.0-mm incision width is associated with significantly lower 6- and 12-month endothelial cell loss than use of a smaller 3.2-mm incision width for forceps insertion of folded DSAEK grafts. Despite this difference in cell loss, the early one-year graft survival rates were comparable and thus additional follow up is needed to determine whether the larger incision has an advantage over the smaller incision in terms of long term graft survival.

Acknowledgments

The authors wish to gratefully acknowledge Megin Murray, Marie Norell, Andrea Wojtowicz, and Dr. Matt Albright in data collection and entry at the Case Vision Research Coordinating Center, Kelly Fairchild and Clorissa Quillin from Cornea Research Foundation of America, and Becky Barr and Bob Lehet from Gorovoy Eye Specialists. Essential support for this study was provided by Jake Requard of Vision Share, Tim Fischer of the Indiana Lions Eye and Tissue Transplant Bank, Jason Woody and Patrick Gore of the Lions Eye Institute for Transplant & Research, Central Florida Lions Eye Tissue Bank (Tampa, FL), Mark Soper of the North Carolina Eye Bank (Winston-Salem, NC), Tony Bavuso of Heartland Lions Eye Bank (St. Louis, MO), and Monty Montoya and Bernie Iliakis of SightLife (Seattle, WA). Finally, we appreciate the advice on study design and support and provision of data from the SMAS and the CDS from Robin Gal, M.S.P.H., C.C.R.A. and Roy Beck, M.D., Ph.D. of the Jaeb Center for Health Research in Tampa, Florida.

Supported by the National Eye Institute, Bethesda, Maryland (EY015145, EY12728), Eye Bank Association of America, Washington, DC; Vision Share, Apex, NC; Cornea Research Foundation of America, Indianapolis, IN; Research to Prevent Blindness, New York, NY; and Ohio Lions Eye Research Foundation, Grove City, OH.

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Table 1

Donor characteristics for Descemet's stripping automated endothelial keratoplasty (DSAEK) performed with two incision widths.

	5.0-mm incision (N=64) Mean (SD)	3.2-mm incision (N=103) Mean (SD)	P= value
Death to Preservation (hours)	6.5 (2.5)	7.8 (2.9)	0.0034
Death to Surgery (days)	4.1 (1.3)	3.5 (1.2)	0.0056
Endothelial Cell Density (cells/mm²)	2782 (320)	2784 (308)	0.97
Age (years)	51 (17)	56 (16)	0.036

Table 2

Recipient Characteristics for Descemet's stripping automated endothelial keratoplasty (DSAEK) performed with two incision widths

	5.0-mm incision (N=64)	3.2-mm incision (N=103)	P= value
Age in years (mean± SD)	69±12	74±10	0.021
Gender, number (%)			0.82
Male	26 (41)	40 (39)	
Female	38 (59)	63 (61)	
Race, number (%)			0.045
White	60 (94)	103(100)	
African-American or Hispanic	4 (6.3)	0	
Diagnosis, number (%)			Fuchs'/Others 0.47
Fuchs' Dystrophy	56 (88)	86 (84)	
Pseudophakic/Aphakic Corneal Edema	7 (11)	15 (15)	
Other endothelial failure	1 (1.6)	2 (1.9)	
Pre-Op Status			< 0.001
Phakic	20	0	
Posterior chamber intraocular lens	44	103	
Post-Op Status			0.45
Phakic	4	0	
Posterior chamber intraocular lens	60	103	

Table 3

Endothelial cell density and cell loss for Descemet stripping automated endothelial keratoplasty (DSAEK) performed with 2 incision widths

	5.0-mm incision Mean (SD)	Number of Eyes	3.2-mm incision Mean (SD)	Number of Eyes	P-value
Endothelial cell density (cells/mm²)					
Baseline	2780 (294)	64	2780 (311)	103	0.97
6-Month	2050 (578)	55	1650 (618)	71	<0.001
12-Month	1940 (570)	45	1580 (635)	62	<0.001
Endothelial cell loss (%)			Mean (SD) [maximum loss] [minimum loss]		
6-Month	27 (20) [-67%][+23%]	55	40 (22) [-81%][-1%]	71	<0.001
12-Month	31 (19) [-70%][+25%]	45	44 (22) [-80%][+4%]	62	<0.001