

# Pull-Through Insertion Technique for Descemet Stripping Automated Endothelial Keratoplasty (DSAEK): Graft Survival and Endothelial Cell Loss after 1 Year in 100 Eyes

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## Abstract

**Aim:** To assess graft survival and endothelial cell loss after DSAEK using a pull-through insertion technique with incision width of 3.2mm or 4.1mm.

**Methods:** The medical records of 100 consecutive eyes that had undergone DSAEK at the Turin University Ophthalmology Institute between November 2007 and April 2010 were reviewed. Surgery was performed by a single surgeon and included: descemetorexis, insertion of a 8.00-9.00mm microkeratome-prepared lamella using a Busin glide, and air injection in the anterior chamber. The lamellas were inserted through a 3.2mm incision in the first 46 eyes, and through a 4.1mm incision in the subsequent 54 eyes. Graft survival, endothelial-cell loss, visual recovery and complications were assessed one year after surgery.

**Results:** DSAEK surgery was performed in 87 patients with mean age  $72 \pm 12$  years. Cornea guttata was the most common preoperative diagnosis (58%). One year after surgery, graft survival was 95%: 91% and 98%, respectively, in eyes with 3.2mm and 4.1mm incision, the difference not being statistically significant ( $p=0.11$ ). Mean endothelial-cell loss was  $37 \pm 12\%$ . Endothelial-cell loss was significantly higher in eyes in which donor tissue had been inserted through a 3.2mm corneal incision ( $39 \pm 15\%$  versus  $32 \pm 10\%$ ;  $p<0.001$ ). In eyes without comorbidities, CDVA was  $\geq 0.5$  in 91% of cases, and  $\geq 0.8$  in 52%. The most common complication was posterior lamella detachment, which occurred in 14 eyes (14%); incidence decreased progressively: 6% in the more recent 50 interventions.

**Conclusion:** After DSAEK with pull-through insertion technique, one-year graft survival rates were high. Graft survival was comparable in eyes in which donor tissue was inserted through 3.2mm and 4.1mm incisions. However, the wider incision lead to lower postoperative endothelial-cell loss.

**Keywords:** DSAEK; Pull-through insertion technique; Corneal incision width; Graft survival; Endothelial-cell loss

In recent years, Descemet-stripping automated endothelial keratoplasty (DSAEK) has become a preferred surgical technique to treat corneal endothelial disorders [1,2]. The procedure enables the endothelium and Descemet membrane to be selectively replaced with a microkeratome-prepared posterior lamella, consisting of endothelium, Descemet membrane, and a thin layer of posterior stroma. The chief advantages of DSAEK, over the traditional technique of penetrating keratoplasty, are the possibility to perform surgery through a small incision, the preservation of innervation and of the biomechanical strength of the cornea, moderate changes of refraction, and faster visual recovery [2-5].

However, endothelial-cell loss after DSAEK is higher than after PK [5]. The technique and incision-width used to insert the donor tissue are considered important factors influencing postoperative endothelial-cell loss [3,4,6,7]. This study assessed graft survival and endothelial-cell loss, after DSAEK using a pull-through insertion technique, with incision width of 3.2 or 4.1 mm.

## Patients and Methods

After approval had been obtained from the institutional review board, the medical records of the first 100 consecutive eyes that had undergone DSAEK at the Ophthalmology Institute of Turin University, between November 2007 and April 2010, were retrospectively reviewed. Cases were included if patient age was above 18 years, and if they had undergone primary DSAEK surgery. For each case, the following medical records were recorded: patient age and gender, preoperative diagnosis, presence of comorbidities, date and type of DSAEK surgery, intraoperative and postoperative complications, additional surgical procedures after DSAEK, preoperative and postoperative Snellen

visual acuity, graft transparency, and endothelial-cell loss one year after surgery.

The primary outcome of the study was graft survival one year after DSAEK. Graft survival was defined as the preservation of corneal transparency. Eyes with opaque corneas or that had undergone repeated corneal transplantation (whether DSAEK or PK) were considered graft failures. The secondary outcome was endothelial-cell loss one year after DSAEK.

## Surgical technique

All procedures were done by the same surgeon (UdS) using a standardized procedure. Donor corneas were received from the Cornea Bank of Turin, where they had been selected following the European Eye Bank Association guidelines [8] and preserved in organ culture medium. At the beginning of the procedure, donor corneas were dissected using the artificial AC and the CBM-ALTK microkeratome (Moria, Anthony, France) equipped with a 300 micron head. The posterior

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lamella was then transferred to a Hanna punch block (Moria, Antony, France) and cut with a trephine of 8.00-9.00 mm diameter. Surgery was performed on the recipient using peribulbar anesthesia. Initially, a 20 gauge anterior chamber maintainer was introduced through a limbal paracentesis. The Descemet membrane and endothelium were stripped off using a reverse Sinsky hook and a stripper (Moria, Antony, France) through a 2 mm corneal temporal incision. A drop of cohesive sodium hyaluronate viscoelastic was placed onto the endothelial side of the donor tissue, which was then introduced using a Busin glide (Moria, Antony, France) through a nasal clear-cornea incision (Figure 1). The incision width was 3.2 mm for the 46 eyes operated between November 2007 and February 2009, and 4.1 mm for the 54 eyes operated between March 2009 and April 2010. An inferior peripheral iridectomy was created. The temporal and nasal corneal incisions were sutured with interrupted 10.0 nylon sutures. An air bubble was injected through the limbal paracentesis, to fill the AC and press donor tissue against the recipient cornea for 10 minutes. Approximately 30% of the air bubble was then removed and replaced with a balanced salt solution. In case of simultaneous cataract or IOL implantation/exchange surgery, this was performed through the temporal incision before DSAEK. At the end of the procedure, the eye received 1 drop each of homatropine 5%, tobramycin 0.3%, and dexamethasone 0.1% solutions. The eye was patched and patients were instructed to lie face up for 2 hours in the recovery room, to allow the remaining air bubble to push the donor tissue against the recipient cornea.

### Postoperative management

After surgery, patients received topical tobramycin 0.3%, four times per day for 1 week, and topical dexamethasone 0.1%, five times per day, which was gradually tapered off over a 12-month period (reduced after one month to three times daily until the sixth month, then once a day). Postoperative examinations were scheduled 1, 7, 30, 90, 180 and 365 days after surgery. Supplementary examinations were arranged according to the needs of individual cases. If posterior lamella detachment occurred, the patient was taken to a minor operating room, where the donor tissue was repositioned under sterile conditions, injecting another air bubble into the AC through a limbal paracentesis.

### Endothelial cell measurement

Baseline endothelial-cell density (ECD) was measured by an Eye

Bank certified technician who used the Laborlux microscope equipped with a 10 X metallographic lens and the 519966 reticule (Leica, Wetzlar, Germany). Postoperative ECD was measured in our clinic by an expert examiner (FD) using the Konan CC7000 non-contact specular microscope (Konan Medical Corp). The instrument's calibration was checked on the photographed endothelial area employing manufacturer's data. Endothelial analyses were always performed in the center of the image, excluding peripheral cells with low-contrast boundaries. Using the Konan Center Method, the center of each cell in a contiguous group of cells was manually marked, after which ECD was detected automatically.

Endothelial-cell loss was calculated for each subject by subtracting postoperative ECD measured one year after DSAEK from baseline ECD, dividing this difference by baseline ECD, and multiplying by 100.

### Statistical analysis

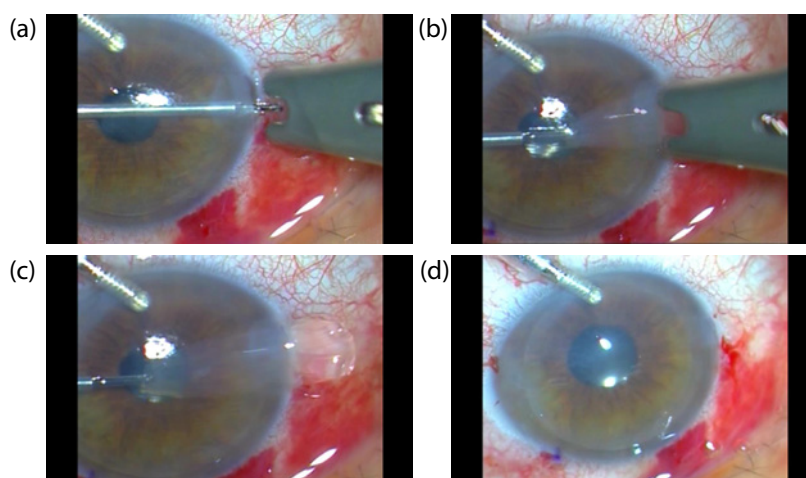
The student's t test was used to compare normally distributed variables. The  $\chi^2$  test was used to compare survival and endothelial-cell loss rates. All reported p values were two-sided, and p values < 0.05 were considered significant. SPSS for Windows, version 17.0 (SPSS, Inc, Chicago, IL), was used for the statistical analysis.

### Results

The study included 100 eyes of 87 patients (61 women and 26 men) who had undergone primary DSAEK surgery. Patient mean age at the time of surgery was  $72 \pm 12$  years (range 33-88). Surgery was performed in one eye in 74 patients, and in both eyes in 13 patients. Preoperative diagnoses are listed in Table 1.

Forty-eight (48%) eyes had comorbidities that influenced visual recovery: macular/retinal diseases (31 eyes), glaucoma (16 eyes), amblyopia (3 eyes), uveitis (2 eyes), ischemic optic neuropathy (1 eye) and one patient had Alzheimer's disease.

DSAEK surgery was performed alone in 67 eyes and combined with other interventions in 33 eyes. Combined surgical procedures comprised phacoemulsification and posterior chamber IOL implantation in 27 eyes, IOL exchange in 4 eyes, and secondary IOL implantation in 2 eyes.



**Figure1:** Pull-through insertion technique using the Busin glide (Moria, Anthony, France). The Busin glide tip was positioned next to the corneal nasal incision and the edge of the donor tissue was grasped using forceps introduced through the temporal incision (Figure 1a). The donor tissue was pulled into the anterior chamber, in this case through a 3.2 mm incision (Figure 1b-c). The donor tissue in the anterior chamber after insertion (Figure 1d).

Preoperative diagnosis	Number of eyes
Fuchs dystrophy	58
Bollous keratopathy	28
- post cataract extraction	(21)
- post glaucoma surgery	(4)
- post pars plana vitrectomy	(3)
PK failure	9
Irido-corneal endothelial syndrome	3
Posterior polymorphous dystrophy	2

**Table 1:** Preoperative diagnosis.

	35 eyes with 3.2 mm incision mean values $\pm$ SD (range)	47 eyes with 4.1 mm incision mean values $\pm$ SD (range)
<b>Baseline ECD (cell/mm<sup>2</sup>)</b>	2678 $\pm$ 155 (3214-2506)	2705 $\pm$ 162 (3012-2530)
<b>ECD one year after DSAEK (cell/mm<sup>2</sup>)</b>	1635 $\pm$ 404 (1993-690)	1844 $\pm$ 312 (2203-852)
<b>Endothelial cell loss (%)</b>	39 $\pm$ 15% (23 - 75%)	32 $\pm$ 10% (28 - 65%)

**Table 2:** Endothelial cell density (ECD) before (baseline) and one year after DSAEK using the pull-through insertion technique with a 3.2 mm and 4.1 mm corneal incisions.

Type of complication	Number of eyes
Posterior lamella detachment	14
Late endothelial decompensation	3
Pupillary block glaucoma	2
Endothelial rejection	2
Deposits in the interface	2
Folds in the interface	1
Herpetic keratitis	1
Cataract	1
Cistoid macular edema	1
Ischemic optic neuropathy	1

**Table 3:** Postoperative complications.

Intraoperative complications comprised posterior capsule rupture during phacoemulsification in 1 eye, incomplete removal of Descemet membrane in 1 eye, and graft decentration in 1 eye.

## Graft survival

All eyes were examined at 1, 7 and 30 days, 97 eyes at 3 months, 91 eyes at 6 months, and 94 eyes at 12 months. Graft survival rate one year after surgery was 95% (89/94). Graft survival rates were 91% (41/45) in eyes with corneal incisions of 3.2 mm, and 98% (48/49) in eyes with incisions of 4.1 mm. Although the survival rate was slightly higher in eyes with 4.1 mm incisions, the difference was not statistically significant ( $p = 0.11$ ). Graft failure was noted in 5 eyes. These cases included 3 eyes with late endothelial decompensation, 1 eye with visually significant folds in the interface, and 1 eye with persistent lamella detachment. The causes of late endothelial decompensation were primary endothelial failure (1 eye), severe endothelial rejection (1 eye), and traumatic endothelial detachment (1 eye).

## Endothelial-cell loss

The mean baseline ECD of donor corneas was  $2.703 \pm 164$  cell/mm<sup>2</sup> (range 2412-3327 cell/mm<sup>2</sup>). Endothelial-cell loss one year after surgery was determined in 82 eyes; in 18 eyes it could not be determined, because the postoperative examination was not available (7 eyes), the quality of endothelial images was poor (6 eyes) or because

primary DSAEK had failed (5 eyes). Endothelial-cell loss one year after surgery was  $37 \pm 12\%$  (range 23-75%); ECD decreased on average from  $2691 \pm 157$  cell/mm<sup>2</sup> (range 3214-2506) to  $1680 \pm 389$  cell/mm<sup>2</sup> (range 2203-690).

Donor age (respectively  $73.8 \pm 10$  vs.  $72.1 \pm 11$  years;  $p=0.27$ ) and baseline ECD ( $p = 0.18$ ; Table 2) of donor corneas inserted through 3.2 and through 4.1 mm incisions were not statistically different. One year after surgery, endothelial-cell loss was significantly higher in eyes in which donor tissue had been inserted through a 3.2 mm corneal incision ( $p < 0.001$ ). Mean endothelial-cell loss was  $39\% \pm 15\%$  in eyes with 3.2 mm incision, and  $32\% \pm 10\%$  in eyes with 4.1 mm incision. In the former group, endothelial-cell loss was higher (34% for 3.2 mm versus 27% for 4.1 mm incision) even when eyes that had undergone rebubbling for graft detachment were excluded from the analysis.

## Visual acuity (Snellen)

One year after surgery, both UDVA and CDVA had significantly increased ( $p < 0.01$ ) in comparison with preoperative values. Mean UDVA improved from  $0.13 \pm 0.04$  to  $0.33 \pm 0.11$ , and CDVA from  $0.25 \pm 0.25$  to  $0.56 \pm 0.32$ . In eyes without comorbidities, CDVA was  $\geq 0.5$  in 91% of cases, and  $\geq 0.8$  in 52% of cases.

## Postoperative complications and additional surgical procedures

The postoperative complications are listed in Table 3. The most common was posterior lamella detachment (14%); the incidence of this complication decreased progressively: in the last 50 interventions it occurred in 3 eyes (6%). This complication was detected within 48 hours of surgery in all eyes, except in one case when it occurred due to a blunt trauma 2 months after surgery. In 12 eyes, the posterior lamella was repositioned by injecting an air bubble through a limbal paracentesis, but in 2 cases the detachment resolved spontaneously.

Other additional surgical procedures included repeated DSAEK in 4 eyes with failed graft, air removal in 2 eyes with pupillary block glaucoma, and phacoemulsification in 1 eye.

## Discussion

DSAEK surgery has been shown to have many advantages compared to PK [1,2]. However, recent studies have reported that endothelial-cell loss is higher after DSAEK than after PK, and that it can reach mean values above 50% one year after surgery [9,10]. The surgical trauma during insertion of the donor tissue into the anterior chamber is considered one of the most important causes of endothelial-cell loss [5,7]. For this reason, different techniques have recently been proposed to optimize donor tissue insertion and reduce endothelial trauma.

The current study assessed graft survival rate and endothelial-cell loss in 100 consecutive eyes that had undergone DSAEK surgery using a pull-through insertion technique with Busin glide [11]. The graft survival rate one year after DSAEK was 95%, which is satisfactory, and comparable with survival rates reported in other studies [5,12-16].

Among the cases of DSAEK failures (5 eyes), just one eye showed primary endothelial-graft failure. Graft failures included 2 eyes that had undergone repeated DSAEK because of folds at the interface and persistent graft detachment. In the first case, surgery was repeated to reduce glare and improve visual acuity, which was limited to 0.4. In the second case, graft detachment persisted after repeated air injection (2 injections). In this eye, with irido-corneal-endothelial syndrome and previous trabeculectomy, a third air injection might have produced further endothelial trauma to the donor tissue, and thus it was decided to perform another graft.



Mean endothelial-cell loss one year after surgery was 37%. Endothelial-cell loss was significantly higher in eyes that had undergone DSAEK through a corneal incision of width 3.2 mm ( $39\% \pm 15\%$  with 3.2 mm incision;  $32\% \pm 10\%$  with 4.1 mm incision). This difference is probably due to the fact that trauma to the endothelium is more severe when donor tissue is introduced into the anterior chamber through a narrower incision. During tissue insertion, the tip of the Busin glide was not allowed to enter the anterior chamber, to avoid iris prolapse and anterior chamber shallowing. The Busin glide tip was positioned at the edge of the nasal incision; the tissue was then grasped using forceps and pulled into the anterior chamber. When the donor tissue passed through the lips of the wound, compression and thus endothelial damage, were more accentuated if the incision was shorter. Similar findings have been reported in laboratory [6] and clinical studies [16] using the forceps insertion technique. In a recent comparative study, Price et al. [16] found that endothelial-cell loss one year after DSAEK was 44% using a 3.2 mm incision, and 31% using a 5 mm incision.

However, prospective and randomized studies will be necessary to confirm these results, as several factors could have biased the results of this retrospective study. Donor death-to-preservation time and donor death-to-use time [17] are unlikely to have a significant effect on postoperative endothelial-cell loss, and differences of these parameters in corneas introduced through a 3.2 mm versus 4.1 mm were not evaluated in this study. Another potential bias was the effect of the surgeon's learning curve on procedure results. The 3.2 mm incision was used in the surgeon's first cases of this technique, when his experience of DSAEK surgery was still limited. In these earlier cases, the graft detachment rate was higher. This complication occurred in 9 of 46 eyes with a 3.2 mm incision, but only in 5 of 54 eyes with a 4.1 mm incision. Graft dislocation and repeated air injection might lead to additional endothelial trauma [18]. However, when endothelial loss was analyzed in eyes that had not previously suffered graft detachment, it was still higher in eyes with 3.2 mm incision (34% versus 27%).

After DSAEK with pull-through insertion technique, one-year graft survival rates were high. Graft survival was comparable in eyes in which donor tissue was inserted through 3.2mm and 4.1mm incision. However, the wider incision lead to lower postoperative endothelial-cell loss.

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