Review Article

# Lamellar keratoplasty techniques

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Access this article online
Website: www.ijo.in
DOI: 10.4103/ijo.IJO_95_18
Quick Response Code:

Lamellar keratoplasty (LK) has revolutionized corneal graft surgery in several ways. Deep anterior LK (DALK) has eliminated risk of failure due to endothelial rejection. Endothelial keratoplasty (EK) has almost eliminated induced astigmatism and the "weak" graft–host junction as seen with penetrating keratoplasty (PK) and also reduced the risk of endothelial rejection. LK provided new insights into posterior corneal anatomy that led to better understanding and performance of DALK and to the development of another EK procedure, namely pre-Descemet's EK (PDEK). Surgical procedures for LK were further refined based on the improved understanding and are able to deliver better surgical outcomes in terms of structural integrity and long-term patient satisfaction, reducing the need of further surgeries and minimizing patient discomfort. In most specialist centers, anterior lamellar techniques like DALK and EK techniques like Descemet's stripping EK (DSEK) and Descemet's membrane EK (DMEK) have replaced the full-thickness PK where possible. The introduction of microkeratome, femtosecond laser, and PDEK clamp have made LK techniques easier and more predictable and have led to the innovation of another LK procedure, namely Bowman membrane transplant (BMT). In this article, we discuss the evolution of different surgical techniques, their principles, main outcomes, and limitations. To date, experience with BMT is limited, but DALK has become the gold standard for anterior LK. The EK procedures too have undergone a rapid transition from DSEK to DMEK and PDEK emerging as a viable option. Ultrathin-DSEK may still have a role in modern EK.

**Key words:** Deep anterior Lamellar keratoplasty, Descemet's membrane endothelial keratoplasty, Descemet's stripping endothelial keratoplasty, Lamellar keratoplasty, pre-Descemet's endothelial keratoplasty

In 2005, the ophthalmic community celebrated one hundred years of penetrating keratoplasty (PK). Starting with humble beginnings in 1905 the technique of PK and its outcomes improved in parallel with improvements in our understanding of wound healing, immunology, the advent of steroids, suture materials, microscopes, micro-instrumentation, and eye-banking techniques.[1] Despite the progress, some fundamental problems persist and defy acceptable solutions. Immune-mediated rejection is a major issue, with 34%–68%[2,3] of grafts being lost depending on the risk category of the host eye. The graft–host junction remains forever weak, dehiscing with trivial trauma even decades after the original surgery. Corneal warpage, related to scarring at the graft–host junction or sutures, often induces astigmatism that adversely affects visual outcome, despite a clear graft. In those patients where the above are avoided, attainment of the full visual potential, which is often excellent, takes a long time, up to 12–18 months.[2-5]

The potential of lamellar corneal grafting in isolated stromal pathology for addressing some of the abovementioned problems has long been recognized. However, lack of appropriate instrumentation and technique led to PK taking precedence. Recent advances in understanding of corneal microanatomy and microsurgery have allowed us to revisit lamellar surgery for both stromal and endothelial pathology, with considerable success; and at the same time address most of the major issues related to PK. Once the principles and concepts were understood, an exponential increase in techniques and instrumentation has occurred such that lamellar transplants have become the preferred choice for several indications. When the recipient healthy endothelium and Descemet's membrane (DM) is retained, and only the stroma is replaced, the risk of failure due the endothelial rejection is completely avoided. Conversely, when only the recipient endothelium is replaced, the integrity of the globe is preserved and induced astigmatism too is more or less completely avoided. Visual recovery is rapid. However, a small, but much-reduced risk of endothelial rejection remains.

This review deals with the principles and concepts behind lamellar keratoplasty (LK), the benefits and limitations of the different types of LK, the advantages offered by different surgical techniques, and the visual outcomes reported and expected. It will help budding corneal surgeons in understanding the principles of lamellar corneal surgery, make decisions on which option to pursue for a given patient and what outcomes to expect. It will also inform surgeons on the

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Manuscript received: 18.01.18; Revision accepted: 26.04.18



**Cite this article as:** Singh NP, Said DG, Dua HS. Lamellar keratoplasty techniques. Indian J Ophthalmol 2018;66:1239-50.

different steps and pitfalls associated with lamellar corneal graft surgery.

## History

The history of these surgeries is important because, "…if you don't learn from what happened in the past, you are doomed to repeat it." In fact, the concept of LK is older than PK. The evolution of corneal transplantation can be traced to early mythology. Greek physician Galen (130-200 AD) proposed a form of superficial lamellar keratectomy to treat the opaque cornea.[1,4] In 1818, Franz Reisinger coined the term "Keratoplasty." In 1886, Von Hippel performed a lamellar transplant where a full-thickness rabbit cornea was grafted on the lamellar bed of a human cornea.[1,4] It took a while after Eduard Zirm's first successful PK, to recognize the importance of within-species allograft. Development in the field of immunology led to the recognition of the phenomenon of graft rejection.[5] Increasing availability of antibiotics, corticosteroids, the introduction of the operating microscope and nylon sutures by Ethicon and the latest, femtosecond laser-assisted corneal incisions led to success and worldwide acceptance of this technique.[6-8]

In 1914, Anton Elschnig completed the first anterior LK (ALK), in a patient with interstitial keratitis.[9] Subsequently, in 1956, Charles Tillet performed the first endothelial keratoplasty (EK) in a patient with chronic corneal edema.[9] The popularity of ALK reduced over 1960s and 1970s. This was mainly due to its poor visual outcome resulting from irregular scattering of light at the recipient-donor interface.[10] In 1965, while performing full-thickness graft, Brown incidentally found that it was possible to leave only DM after performing a deep lamellar dissection.[11] Special surgical skill was required to attain a smooth interface at a deeper corneal plane.[12] Archila was the first to inject air into the stroma to reach a deep stromal plane.[13] Sugita and Kondo tried hydrodelamination to expose the DM.[14] Tsubota then tried layer by layer manual dissection also called the "divide and conquer" technique.[15] Melles devised a technique for intrastromal dissection in a closed fashion.[16] A viscoelastic bubble was tried by Manche.[17] The breakthrough was in 2002 by Anwar who invented the "big bubble technique."[18] This allowed a smooth interface resulting in better visual outcome in patients. Microkeratome- and femtosecond-assisted approaches are gaining popularity.

The surgical technique of posterior lamellar surgery was initially attempted through a superficial flap approach. After designing his first ophthalmological instrument, the Barraquer keratome with pneumatic fixation, Jose Barraquer introduced an anterior approach to EK.[19] In this procedure, after manually creating a partial-thickness hinged corneal flap (LASIK-like), the posterior cornea consisting of stroma and DM was trephined. The donor graft was then sutured in place, and the flap was replaced and sutured again. The most important limitations of this procedure were irregular astigmatism and vascular ingrowth. Most importantly, he speculated that simply filling the anterior chamber (AC) with air may help keep the graft in place.

In 1998, Gerrit Melles described a procedure called posterior LK (PLK), where he dissected the posterior stroma and DM through a 9-mm sclerocorneal incision.[20] After trephining the posterior stroma with a flat trephine in the AC, a donor button with posterior stroma and DM was inserted and held in place with an air bubble. Mark Terry adopted a similar procedure using a specialized intrastromal trephine to remove 100 microns of posterior stroma and named it deep lamellar EK (DLEK).[21] In 2002, Melles group published regarding folding the donor tissue into half and inserting through a 5-mm incision.[22,23] In 2004, Melles group also developed the technique of "Descemetorhexis" to score off the diseased DM.[24] The donor tissue grafted had some amount of posterior stroma which did not affect the visual outcomes.[25] This technique was then called as Descemet's stripping EK (DSEK). However, this led to a dramatic increase in the number of graft dislocations.[26] If the donor tissue preparation involved a microkeratome, then the technique was called Descemet's stripping automated EK (DSAEK).

In 2006, Melles group developed a new technique called DMEK, where the donor DM was stripped and injected into the host AC after Descemetorhexis through a 3.5-mm clear corneal incision.[27,28] The membrane was unscrolled using air and fluid. DMEK lenticule preparation by "SubHys" technique was also described, where liquid culture medium is used to create a cleavage plane between DM and the stroma.[29]

Meanwhile, Vajpayee and group from India described a "hitch-suture" technique to unfold the donor lenticule in DSAEK to minimize endothelial damage.[30] A "taco fold" technique was also described.[31] The Busin glide was invented to insert the tissue roll into the AC with correct orientation.[32] In 2008, Donald Tan invented a glide technique to insert donor lenticule without folding.[33]

The latest innovation in lamellar surgeries is pre-Descemet's EK (PDEK). In 2013, after Dua and group demonstrated the presence of Dua's layer and its surgical implications in cornea, the first case series of PDEK was officially reported by Agarwal *et al.* and Dua *et al*.[34,35] Other surgeons had previously, inadvertently created PDEK tissue but had reported the procedure as DMEK.[36] Recently in February 2017, Dua and group[37] introduced the PDEK clamp to assist in pneumodissection for obtaining PDEK tissue.

## Introduction to Lamellar Surgeries

By default, PK is the gold standard against which the lamellar procedures are compared. The major comparative parameters are the visual function and graft survival. Graft survival includes vulnerability to trauma, rejection episodes, and late failure due to decline in endothelial count.

LK can be broadly divided into two categories, replacement of epithelium and stroma (anterior lamellar) and replacement of DM (posterior lamellar or EK). ALK can be (a) Bowmans membrane transplant, (b) superficial ALK (SALK), or (c) deep ALK (DALK). PLK can be (a) DSEK, (b) DMEK, or (c) PDEK. Most of the above can be performed manually or with the assistance of a microkeratome or using the femtolaser to make lamellar cuts at desired planes. LK creates tissue planes that are not just the vertical apposition of the graft edge to the host rim as in PK but also the interface between the graft and host bed in the coronal plane. Different types of donor-recipient interfaces are recognized. In DMEK, it can be donor DM to host PDL or donor DM to host DM when host DM is not removed. In DSEK, it can be donor stroma to host PDL or donor stroma to host DM

when it is not removed. In PDEK, it is always donor PDL to host PDL. In DALK, it can be donor PDL to host DM or donor PDL to host PDL or donor DM to host PDL when donor DM is not removed. When manual dissection is required, and the PDL is not exposed, the interface is donor PDL to host stroma.

## Bowman's Layer Transplant

The anterior-most compact collagen of the corneal stroma is made of the Bowman's layer or zone. This is believed to give biomechanical strength and shape to the anterior cornea. In progressive corneal ectasias, such as seen in advanced keratoconus, the Bowman's layer thins and disrupts, weakening the cornea, and exacerbating the tendency for further ectasia.

The advocates of Bowman's layer transplant argue that replacing the Bowman's layer will add strength to the anterior cornea, restore shape (induce flattening) and arrest progression. It will thus allow visual rehabilitation with contact lenses by improving fit and retention of the lens, which in turn would delay or perhaps avoid the need for DALK/PK. The innovator of Bowman's layer transplantation had initially reported the use of this technique in treating subepithelial scarring after photorefractive keratectomy.[38] The technique does not carry any risk of allograft rejection because no biological material is transplanted.

After removing the epithelium, air is injected in the donor cornea beneath the Bowman's layer to separate the anchoring fibrils to form the "Bowman's roll," which is immersed in 70% ethanol to remove remnant epithelial cells. Manual dissection or femtosecond laser is used to create a stromal pocket in recipient cornea at around 60 μ depth, and the "roll" is placed in it using a glide and then unrolled and covered with a scleral contact lens. The roll is stained with trypan blue for better visualization.

This procedure leads to decrease in keratometry values, improved corneal thickness, and better tolerance of contact lens.[39] The visual acuity is also significantly improved.[40] However, in a few cases, an intrastromal cavity has been documented during the initial postoperative period.[39,40] In a recent study with a 5-year follow-up of 20 eyes in 17 patients, it was demonstrated that the decrease in Kmean and Kmax values seen at 1 month postoperative remained stable at the 5-year follow-up time point. Best-corrected visual acuity (BCVA) improved up to 1 year and then remained stable for 5 years.[41]

## Superficial Anterior Lamellar Keratoplasty

SALK is the replacement of the anterior corneal stroma. Very anterior corneal opacities are amenable to phototherapeutic keratectomy where the stroma with the pathology and surrounding normal stroma is ablated and the surface allowed to heal by cell migration from the limbus. Slightly deeper opacities can be managed by superficial anterior keratectomy, wherein more stroma than what is ablated by PTK is removed but not replaced, and healing occurs as with PTK. Stromal pathologies located in the anterior third of the stroma (e.g., Reis-Buckler) and surface irregularity, astigmatism, and stromal thinning (keratoconus) can be managed by SALK.[42]

In SALK, the diseased stroma is removed and replaced by healthy stroma of similar thickness and an intact epithelium; however, replacement of epithelium is not necessary. In corneal ectasia (e.g., Keratoconus), no stromal tissue is removed before replacement with donor stromal lenticule. Dissection depth is usually up to 160 μ or approximately less than one-third of the corneal thickness. In case of recurrence of the same dystrophy in the graft, replacement of the superficial graft can be easily done.

With superficial opacities, a microkeratome or femtolaser flap, of a thickness corresponding to the level of the opacities, is made and removed (free cap). Generally speaking, in eyes with anterior stromal scars, the microkeratome is preferred over femtolaser flap creation as penetration of laser light through scars can be limited and variable.[43] In case of depressed superficial scars, this technique is not suitable because the microkeratome cut follows the surface profile and leads to the same defect in the stromal bed. A similar flap is cut in the donor eye and punched to fit the diameter of the recipient bed. The donor tissue is either glued or sutured in place.[42,44]

Where the cornea is thin and ectatic (e.g., keratoconus) a recipient flap is made as described above. A stromal lenticule is prepared from a donor eye-bank eye and placed in the bed of the recipient cornea, and the flap is repositioned-stromal sandwich technique.[45] An additional step to perform photorefractive keratectomy on the donor stroma lenticule to address induced myopia, followed by repositioning of the recipient flap, has also been described. Alternatively, residual or induced refractive error is corrected at a second stage, around 6 weeks later by transepithelial photorefractive keratectomy.[45]

These grafts clear almost immediately, but irregular astigmatism can limit the visual improvement. Theoretically, ALK minimizes the risk of keratectasia in keratoconus patients and ectasias after refractive surgery as it augments corneal thickness.[44,45] Anterior corneal opacities and stromal dystrophies where the pathology extends up to mid stroma can also benefit from ALK. Here, it improves vision and alleviates symptoms of recurrent corneal erosions as seen with granular and lattice dystrophies. The technique can also be performed in previous penetrating grafts where anterior recurrence of the dystrophy has occurred.[46]

## Deep Anterior Lamellar Keratoplasty

Several stromal pathologies such as granular and lattice dystrophy, stromal scars, and corneal ectasias (e.g. Keratoconus) result in visual impairment due to an obstruction to the passage of light or distortion of focus. The DM is often normal in these cases. It, therefore, makes sense to remove only the stroma and replace it with a full-thickness stromal button (often with donor epithelium) devoid of DM and endothelium, taken from a donor eye. This avoids the risk of graft failure due to endothelial rejection. The challenge lies in its complete removal, leaving behind only the host DM or DM with the PDL (Dua's layer). The button needs to be sutured in place, which introduces the major issues of post-DALK astigmatism and a prolonged recovery period requiring adjustment of astigmatism with selective suture removal. The bursting pressure of PDL is reported to be between 500 mm and 700 mm of Hg[47] and phacoemulsification with implant has been successfully carried out under the PDL.[48] Hence, it is likely that when the PDL is retained the eye has greater strength and does not rupture easily following trivial trauma, unlike PK. How much strength is conferred by retention of host DM only is currently unknown.[47,48]

The surgical technique aims to induce a cleavage in the PDL or DM plane and the anterior stroma. When this plane is successfully accessed, the host and recipient beds are very smooth with precise apposition of the donor button and host bed [Fig. 1]. The regularity of the interface determines the visual outcome. With Anwar's "big bubble (BB) technique," this difficult but important step has become increasingly possible.

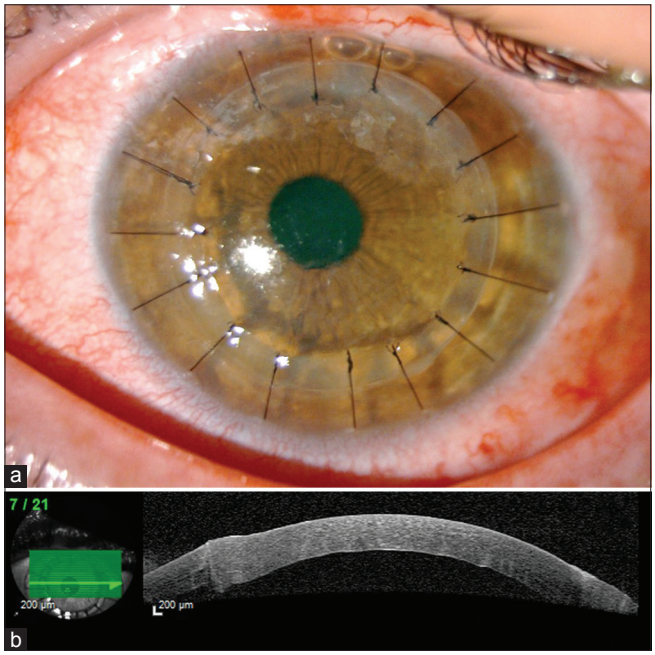


**Figure 1:** Deep anterior lamellar keratoplasty with a Type-1 big bubble. (a) Postoperative day 1 clear graft diffuse slit-lamp view. (b) Optical coherence tomography showing normal graft thickness centrally and thickening at the graft–host junction as also occurs with penetrating keratoplasty. The pre-Descemet's layer is closely applied to the donor stroma

In the BB procedure, air is injected into the recipient stroma through a trephine cut of desired diameter (usually 7.5–8 mm). Injected air follows a defined and reproducible path[49] to reach the PDL plane, to produce a Type-1 BB. This is the most common type, occurring in approximately 80% of patients. At times, air accesses the plane between PDL and DM by passing through tiny fenestrations along the corneal periphery, central to the attachment of the DM. The BB so produced is the Type-2 BB, where only the DM (without PDL) is exposed. Often a Type-1 and Type-2 BB (mixed BB) occur simultaneously with the former usually being complete and the Type-2 BB appearing as a smaller bubble. A Type-2 or mixed bubble occurs in approximately 12% of cases.[50] When a mixed BB occurs, a tiny puncture with a needle, through a drop of viscoelastic placed on the PDL (anterior wall of the Type-1 component), allows the Type-2 bubble to deflate. Air injected into the AC at the end of the operation helps to tamponade the DM to the PDL.

Pneumodissection does not always yield a BB, in which case a layer by layer dissection of the anterior stroma needs to be done till the PDL plane is reached. This invariably leaves behind more stroma,[51] but the interface is usually compatible with improved visual acuity. In some patients, it is possible to identify a deeper small bubble that represents air trapped between the deep stroma and/or the PDL or DM. When this is noted, the small bubble wall is punctured to access the plane of cleavage and manually dissect up to the trephine edge, manually.[52] A visco-bubble technique where intrastromal injection of viscoelastic can be used to achieve a deeper stromal plane has been described by Guell *et al*. However, it has been recently shown that unlike with air injection, the plane of insertion of the needle tip is crucial, because with mid stroma injections of viscoelastic, an intrastromal bubble, mimicking a Type-1 BB, often occurs.[53]

Inadvertent punctures or tears in the PDL in a Type-1 BB are compatible with successful DALK. The dissection depth is very important. It has been observed that DALK patients with

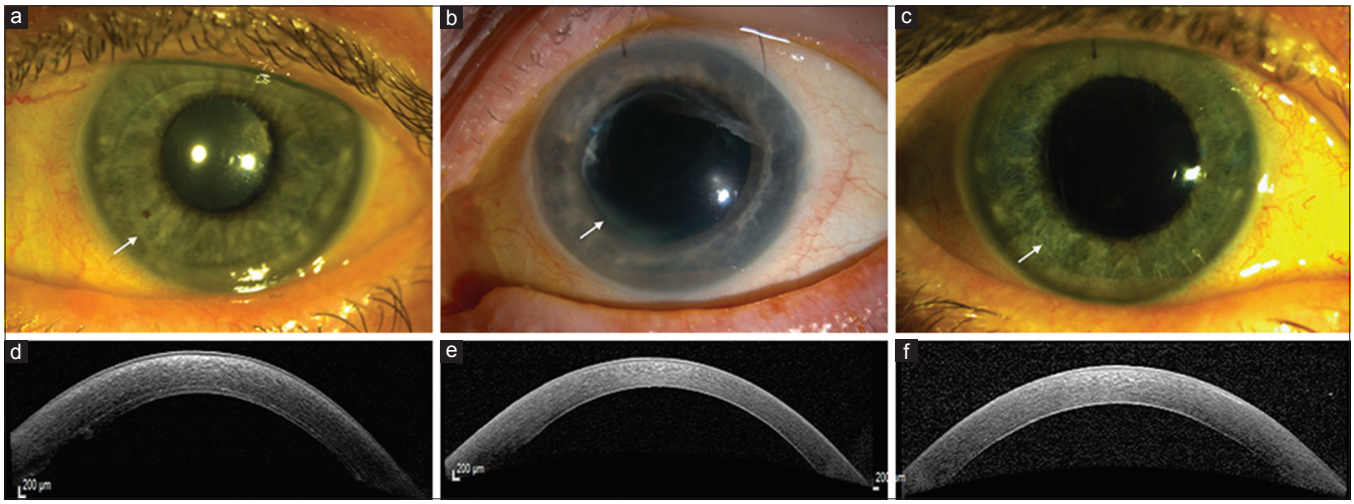


**Figure 2:** Endothelial keratoplasty: Postoperative slit-lamp and optical coherence tomography images of patients who had undergone Descemet's stripping endothelial keratoplasty (a and b), Descemet membrane endothelial keratoplasty (c and d), and pre-Descemet's endothelial keratoplasty (e and f). The edges of the grafts are visible (arrows). The Descemet's stripping endothelial keratoplasty graft is the thickest on optical coherence tomography. Descemet membrane endothelial keratoplasty and pre-Descemet's endothelial keratoplasty graft show similar outcomes in terms of graft thickness and visual outcome

**Table 1: Complications of deep anterior lamellar keratoplasty**

| Complications of DALK | Comment |
|---|---|
| Failure to separate DM/PDL by air or viscoelastic | May require repeated attempts or eventually manual dissection |
| Intrastromal bubble with viscoelastic technique mimicking Type-1 big bubble | Needle or cannula tip should be in deep stroma. Intraoperative OCT is helpful in recognizing this |
| Perforation or bursting of DM/PDL | PDL perforations tend not to extend and DALK can be completed. DM perforation (type 2 bubble) can enlarge and requires conversion to PK |
| Double anterior chamber | Can be due to perforation of PDL/DM and requires anterior chamber air tamponade. Also can be due to Mixed bubbles, air from the type 2 component can be released by making a tiny perforation in the overlying PDL |
| Urrets-Zavalia syndrome | Always be wary of postoperative pressure rise. Check intraocular pressure an hour after surgery if air is retained in the AC. Dilate the pupil. Release air from anterior chamber, at the slit lamp |
| Interface wrinkling | Mismatch in surface area of recipient DM and donor stromal button can cause wrinkling. Oversizing the donor button by 0.25-0.5 can reduce or prevents this |
| Early suture loosening | Occurs with shallow sutures. 80%-90% suture depth should be aimed for |
| Interface vascularisation | Is a sign of stromal rejection and can cause interface scarring. Aggressive treatment with topical steroids and fine-needle diathermy of the trunk at limbus can be considered |
| Graft rejection | Epithelial rejection does occur but the risk of this triggering endothelial rejection, like in PK, is not an issue. Stromal rejection is usually associated with vascularization. Both can be treated with increased use of topical steroids, cyclosporine drops, or oral steroids |

DM: Descemet's membrane, PDL: Pre-Descemet's layer (Dua's layer), DALK: Deep anterior lamellar keratoplasty, OCT: Optical coherence tomography, PK: Penetrating keratoplasty, AC: Anterior chamber

a residual stromal bed of less than 20 μ achieved a visual acuity comparable to PK.[54]

Femtosecond laser in DALK (the Intrabubble technique): Dissection of the stroma and precise channel formation for needle placement at predecided depth of 50 μ and 30° angle for BB formation can be achieved using femtosecond technology.[55,56] Matching donor and recipient edge using zigzag cuts allow precise tissue apposition.[57] The possibility of elimination of suture-related complications has also been tried through sutureless DALK.[58]

No significant difference has been noted in the refractive outcomes of DALK as compared to PK. Theoretically, sutures can be removed earlier in DALK, thus reducing the risk of other suture-related complications such as infection, irritation, and astigmatism. Most surgeons tend to take superficial passes while suturing with an intention to avoid perforating the DM. Hence, suture loosening and ocular surface inflammation are common.

Preserving the host's healthy endothelium and thus, virtually eliminating the chances of rejection related graft failure is the main aim of ALK. The endothelial loss after DALK is biphasic. There is an early phase of rapid loss followed by a late phase of slow decline.[59] DALK is, however, better than PK when it comes to endothelial cell counts. If there is an intraoperative perforation during the procedure, it leads to 25% reduction in endothelial cell counts.[60] DALK also lets the eye be more secure than after PK. This is applicable to the eyes where DALK was completed with a Type-1 bubble, as the Dua's layer, with the bursting pressure is 500 mmHg, is retained.[47]

Complications [Table 1] like double AC can result from break-in DM through which aqueous can access the plane between DM and DL (Type-2 BB) or between DL and posterior stroma (Type-1 BB). DM tears tend to extend and the edges or the tear, roll. Breaks or tears in the DL + DM tissue (Type-1 BB) do not extend, and as the DM remains attached to the DL, management of the tear can be achieved by injection of air in the AC. Dramatic bursting of a Type-2 BB can occur intraoperatively. In DALK regardless of the type of BB, it is important to keep the eye pressure low by repeated evacuation of aqueous through the paracentesis site.

Pupillary block glaucoma is a relatively common complication when the AC is filled with air as part of any lamellar procedure though not commonly required in DALK. This can result in a fixed dilated pupil (Urrets-Zavalia syndrome).[61] This can be prevented by preoperative inferior iridotomy, wide dilation of the pupil, reducing the volume of air before the patient is taken off the operating table, checking intraocular pressure (IOP) within an hour postoperative and releasing air from the AC if IOP is high (burping the bubble). Intravenous acetazolamide and/or mannitol can help if the pressure is not too high. Interface infection is another serious but rare complication. Interface debris in the form of fibers and particles from surgical swabs is more common and annoying.

Due to the steep learning curve of the procedure, the decision to continue with DALK after an intraoperative perforation or to convert to PK must be made after weighing the risk-benefit ratio to the patient. In conditions such as keratoconus and stromal dystrophies, where PK has repeatedly shown a good outcome, it is acceptable to convert. However, in patients with vascularized corneas and those with ocular surface disease in whom the chances of rejection are high, DALK serves as a boon and significantly reduces the chances of endothelial rejection [Table 1].

## Endothelial Keratoplasty

Endothelial dysfunction syndromes such as Fuchs endothelial dystrophy, posterior polymorphous dystrophy, iridocorneal

endothelial syndrome, failed PK, pseudophakic, or aphakic bullous keratopathy can lead to stromal and epithelial edema, which causes visual deterioration. In these conditions where only the corneal endothelium is damaged, and there is no associated stromal scarring, replacement of the DM only is possible. The major benefit of endothelial transplantation is that it maintains the structural integrity of the eye, is astigmatically neutral, and has a reduced risk of rejection.

Early attempts at EK, in the form of DLEK and PLK, were relatively crude and traumatic to the host cornea and produced a rough interface.[22] Modern EK procedures have undergone rapid evolution with refinements in technique and in our understanding of posterior corneal anatomy. Three procedures are in vogue, namely DSEK, which when performed with the help of automated trephine for donor tissue preparation is termed DSAEK; DMEK and PDEK. Although the latter two are more recent than DSEK and provide comparatively better visual outcomes, DSEK is still practiced in many centers due to the technical difficulties and steep learning curve of the other EK procedures. All EK procedures are inherently associated with endothelial loss related to preparation, insertion, and attachment of the donor EK tissue. Up to 50% EC loss has been reported at 5-year postoperative.[62,63] Hence, it is important that donors with good endothelial cell counts, at least 2300 cells/mm$^2$[64] are selected and donors of 70 years of age or older are usually avoided.

## Descemet's Stripping Endothelial Keratoplasty

Although there is a learning curve in transitioning from PK to DSEK many surgeons worldwide have taken to DSEK rapidly.[65]

Donor tissue preparation remains a critical step in DSEK and that is the main reason why most surgeons prefer to complete donor tissue preparation before shifting the patient in the operation theater. Of late, precut donor tissues have become available through many eye banks.

Methods to prepare the donor tissue include manual dissection, use of microkeratome (DSAEK) or femtosecond laser. Donor tissues with high endothelial cell counts of 2500–3000 cells/mm$^2$ and from young donors are preferred. A large diameter scleral rim is required to mount the donor tissue on the artificial chamber. In microkeratome-assisted technique, decreased scleral rigidity from very young donors may lead to irregular dissections. Donor lenticule diameter should be at least 3 mm less than the recipient cornea diameter to prevent closure of the AC angle. The meniscus shape of the donor tissue where it is thinner in the center and thicker in the periphery leads to approximately 1.5 diopters of hyperopic shift postoperatively. The graft is also thicker than with other EK procedures [Fig. 2a and b].

A smooth stromal bed of the donor tissue can be obtained using a microkeratome or a femtosecond laser, better than the manual dissection technique. This leads to better and improved visual acuity. The microkeratome gives a smoother surface, but inflammation is less with femtolaser.[66,67] Eccentric cuts or nonuniform donor tissue thickness increases the chances of primary graft failure and tissue dislocations.

The storage media of the tissue remains an area of controversy. It has been postulated that dextran and chondroitin sulfate in the solution lubricates and increases chances of graft dislocations while glutathione and glucose in balanced salt solutions (BSSs) increases endothelial function leading to better adherence of the donor tissue to the recipient stroma.[68]

Creation of the entry wound: For insertion of the donor tissue into the AC, the entry wound size has seen a rapid decline from 9 mm initially, to 5 mm or even 3 mm, with correspondingly enhanced wound security and reduction in induced astigmatism.[22,69] Reduction in size of incision necessarily involves folding of the donor lenticule either in two unequal halves (taco technique with forceps) or its rolling into a loose roll as with the Busin glide or other inserting devices. The entry tunnel can be either corneal or sclera-corneal.

Descemetorhexis: It is usually carried out with the AC filled with BSS by continuous irrigation. It can also be carried out with viscoelastic filling the AC, as is often the case in phaco-DSEK, but requires thorough removal before insertion of the graft. Filling the AC with air, either during or after Descemetorhexis provides the best visualization of the DM and helps to easily identify any remnants that may be attached to the graft bed. The diameter of the rhexis should preferably be larger than that of the graft lenticule.[70] The area of DM to be removed is marked (on the epithelial surface) and scored with a reverse Sinsksy hook. The membrane edge is gently lifted off with a "stripper" and the DM is peeled off by grasping the lifted edge and pulling or by stroking with a stripper.

Insertion of the graft lenticule: After descemetorhexis, insertion of the donor tissue in the AC and its accurate positioning is a major challenge. It should be ensured that the AC maintainer is switched off during this step to ensure that the graft does not shoot out of the wound.

Maneuvers used in folding of the tissue, its unfolding after insertion and positioning are associated with stromal folds and loss of endothelial cells. Donor endothelial cell loss of up to 34%–51%[62,63] in the first 6 months has been reported with the folding technique. With the Sheets glide, it was only 13.5%.[71] Subsequently, there is only 1% cell loss over the following 6 months. Primary graft failure rates vary from 6% to 45%[72,73] using the folding technique and are as low as 2% using the TAN EndoGlide™ (Angiotech, Vancouver, Canada) technique. Furthermore, graft inversion chances are significantly reduced if the Sheets glide is used.

Opening the folded graft in the correct orientation is crucial. To ensure these most surgeons use an "S" or "F" mark with ink (in DSAEK), on the stromal surface. The letters should be read in their correct orientation after unfolding the graft. With manual techniques, two identifiable marks (e.g. one dot and two dots) need to be placed at 90° to each other to assess correct orientation. A combination of stroking and irrigation maneuvers is used to unfold the graft and position it centrally. Finally, an air bubble is used to approximate the donor to the posterior corneal surface. A complete air fill without unduly raised pressure is desirable. Intracameral air itself is associated with 10%–20% loss of endothelial cells.[74-77] Air in the AC is maintained at 100% for at least 10 min, after which some air is burped to prevent pupillary block. An inferior iridectomy performed intraoperatively or before surgery (with YAG laser) is desirable.

Like with DALK and air in the AC, it is important to assess the IOP within an hour after surgery and raised IOP controlled by burping the bubble. The patient is advised to lie flat on the back for a few hours, and the pressure checked again. Venting incisions can be made to drain the interface fluid and accelerate the adherence. Intraoperative anterior segment OCT can help confirm the opposition of the graft to host bed.

Small amounts of retained DM on the graft can interfere with graft attachment. Aphakic or pseudophakic eyes without a posterior capsule or even eyes with glaucoma drainage devices can be difficult candidates for air retention, but strict supine position or use of gases such as $SF_6$ can help.

Donor detachment and dislocation of 14%–23% are major complications requiring graft repositioning and rebubbling.[78,79] However, DSEK gives tectonically stronger globes as compared to PK.[80]

If the attached graft fails to clear even after 2 weeks, it is known as primary graft failure, usually caused due to unhealthy donor epithelium or traumatic operative technique. It has been observed that the incidence of primary graft failure is lower for automated dissection than the manual technique.[26] In addition, it is lower with more experienced surgeons.[79] If the graft remains unattached, which may be due to retained DM, viscoelastic or interface fluid; it is termed as secondary graft failure. Graft survival rates of 60%–94% have been reported in long-term studies.[81-83] Five-year graft survival rates after DSEK have been reported to be superior as compared to PK.[84] However, primary graft failure of up to 5% after DSEK is reported.[81] During the first month, donor endothelial cell loss is higher as compared to that after PK.[81] This may be due to handling and apposition techniques employed. There is an early plateau phase seen at 6 months in EK procedures. The endothelial cell loss after DSEK is 53% at 5 years which then continues to reduce at 1% every year and that of PK is 69%–75% at 5 years,[72,85,86] which becomes statistically same for both at 10 years.

Endothelial graft rejection rates of 10% have been reported.[78] Graft folds or wrinkles in the pupillary area can lead to reduced visual outcome, which may be an indication for a regraft. Interface scarring/haze and irregularity is another cause of postoperative reduced vision. Hence, it is very important to preoperatively rule out stromal scarring as it may persist postoperatively. If intraocular lens implantation is planned in the same sitting, the hyperopic shift should be taken into account when ascertaining the power of the lens to be implanted.

Most patients have a speedier visual recovery as compared to PK. Visual outcomes in terms of BCVA is much better than that of PK because of reduced astigmatism due to the absence of sutures.[87] There is some evidence, which supports PK to give clearer vision than DSEK, which could be due to the absence of an interface.[88,89] The final refractive correction in PK is not known till all the sutures are out. DSEK induces a hyperopic shift of 1–2 diopters, which can impact on vision.[90,91]

The use of microkeratome in DSAEK procedure is beneficial in terms of decreased astigmatism, rapid visual recovery, and reduced rejection rate.[92] In a post-DSAEK cornea, the posterior surface of the cornea is not parallel to the anterior surface of the DSAEK donor tissue. This leads to increase in the higher order aberrations of up to 25%.[93]

Femtosecond laser has also been used for graft preparation in DSAEK termed F-DSAEK. Along with the irregularity during tissue cutting, F-DSAEK causes laser-induced roughness due to deep ablation. It has been postulated that if the anterior surface of the cornea is depressed, as during f-DSAEK, persistent wrinkles form on the posterior surface of the donor cornea transplant tissue.[92] There is also an element of interface scatter due to corneal collagen denaturation.[94] This leads to the worse visual outcome as compared to DSAEK. F-DSAEK still needs further technical enhancement before it gains acceptance.

The problem of graft thickness was theoretically solved with the introduction of ultrathin (UT) DSAEK grafts, which are thinner than 130 μ.[95] These grafts give a spherical equivalent similar to DSAEK with visual outcome comparable to DMEK.[96,97] In addition, the complication rate and ease of procedure are better with UT-DSAEK than with DMEK.[95] However, this remains an area of debate and surgeon preference.

## Descemet's Membrane Endothelial Keratoplasty

DMEK obviates the major disadvantage of DSEK, which is the addition of additional stroma and consequent hyperopic shift [Fig. 2c and d]. DMEK tissue is made of DM only. Vision obtained from DMEK is usually 6/9 to 6/6 while that from DSEK is around 6/12.[98,99] The indications for this procedure are the similar to DSEK.

The donor tissue preparation techniques range from manual dissection[100] to the submerged cornea using backgrounds away technique,[101] where the dissection is mainly done after submerging the donor cornea under Optisol GS or BSS, enabling better visualization and easy handling of the tissue while dissecting. Pneumodissection has also been attempted (Type-2 BB), but this can result in the formation of a Type-1 BB and provide PDEK tissue (see below). Initially, before the knowledge of the PDL, surgeons described this technique for obtaining DMEK tissue but were, in many instances, transplanting PDEK tissue.[36,102] As the DM is relatively firmly attached to the Dua's layer in young donors' cornea, it has always been the tendency to harvest DMEK tissue from eyes above 50 years of age only.

The donor tissue is usually 8 to 8.5 mm in diameter. The next step is to transfer the tissue into the eye using a device that carries the tissue with minimal damage to the endothelium. DMEK tissue always scrolls with the endothelial cell layer on the outside. Several devices are in vogue ranging from conventional lens inserters to glass pipettes and tubes made for this purpose. The "no-touch" technique for implantation and apposition of the DMEK graft is widely used.[103] The tissue is stained with trypan blue dye (for 2–3 min), and the scroll is aspirated into a glass tube from its wide end and injected into the eye through the narrow nozzle end by switching the syringe attachment. DM scrolls can be very tight (cigar shaped), double rolls or loose folds.[104]

During injection, active irrigation of BSS into the AC should be stopped, and the eye should be soft. Positive pressure can expel the scroll from the wound or even from a paracentesis opening. Once in the AC, it is advisable to stitch the entry wound to keep the chamber closed during manipulations to unscroll and attach the DM graft. Different strategies using

mechanical tapping, fluidics, and air have been described to unscroll the tissue. The less manipulation required, the better it is for retaining healthy endothelial cell counts. The general principle is to orientate the scroll such that when it opens, it does so with the endothelial surface toward the iris.

After orienting the scroll, the AC is made shallow and the cornea above the scroll is tapped with a blunt instrument. The forces the scroll to open and stay opened. When the scroll is centered before opening, the graft remains centered when open. Half open or asymmetrically open scrolls can be opened by use of air or gentle irrigation. Once open and centered, an air bubble is injected between the tissue and iris to elevate the graft and oppose it to the posterior surface of the cornea from which the host diseased DM has been stripped off.

Slightly decentered grafts are acceptable as the gap between the host DM and the graft is usually repopulated with migrating endothelial cells. However, peripheral infolding of the graft should be avoided as it can lead to the formation of double AC and graft dislocations postoperatively. Such infolding can be flattened out with a "bubble-bumping maneuver" where gentle taps on the outer surface of the cornea over the fold create an aqueous flow which leads to their disappearance. Finally, filling the AC completely with air for 20 min helps in graft fixation.

The uptake of DMEK by corneal surgeons has not been as rapid as DSEK because of a steep learning curve and difficulties in handling the very thin and friable DM.[105,106] DMEK has higher rates of primary graft failure and donor tissue wastage than DSEK.[101] The technique needs to be simplified. Modified tissue preparation techniques where some amount of peripheral stroma is retained for structural support of the friable tissue have been tried.[107,108] This technique, however, is associated with 5%–30% tissue wastage.[109] A "SubHyS" technique of graft preparation has been recently described where liquid is injected into the corneal stroma to separate the DM from the PDL.[29]

Endothelial cell loss is shown to be 26% in the first month which rose to 39% at 5-year postsurgery, which is significantly better than DSEK and PK which are 53% and 70%, respectively.[110] Primary graft failure rates have been reported to be 6%–8%.[101,111] Rate of postoperative graft detachment is reported to be greater in DMEK than DSEK.[112]

DMEK has better visual outcomes in terms of contrast visual acuity and aberrations than DSEK.[101] 36%–79% of DMEK patients achieve logMAR 0.8 or better, as compared to 23%–47% of DSEK patients.[113] The general consensus is that DM-to-stroma interface in DMEK is better than stroma-to-stroma interface in DSEK for optical clarity.[98,99] Furthermore, the thin DM graft has almost no hyperopic shift postoperative.

The only issue of concern, besides the steep learning curve, is that donor age for DMEK tissue should be more than 50 years to facilitate easy removal of the DM scroll.[114] Theoretically, increased donor age reduces the endothelial cell viability of the graft in the host eye.[115]

## Pre-Descemet's Endothelial Keratoplasty

The paper reporting the presence of the PDL also describes its use, together with the DM in EK.[34] Based on this method of harvesting donor tissue for EK, a technique was devised to transplant the tissue in patients as an alternative to DSEK and DMEK[35] and was termed PDEK. It was noted that the PDEK tissue scrolled less and was easier to handle and unscroll in the eye compared to DMEK tissue. The donor endothelial graft in DSEK has 100–150 μ of posterior stroma, while the PDEK graft is much thinner as it consists of DM and PDL, the PDL being 10–13.6μ thick[34] [Fig. 2a-f]. Visual outcomes are similar for both PDEK and DMEK grafts. PDEK is still in its early stages and more experience, with time, will help establish its position in EK.

The donor tissue preparation technique of PDEK involves intrastromal injection of air using a 30-gauge needle. The needle tip, bevel facing endothelium, is inserted through the scleral rim of the sclerocorneal disc and advanced to mid-peripheral cornea. Air is injected until a Type-1 BB forms.[35,49] Air injected in the stroma moves in the coronal plane of the sclera-corneal disc as a radial "spoke" to reach the limbus. On reaching the limbus, the air moves circumferentially like a band in clockwise and anticlockwise directions to complete the circle. This air then moves centripetally to fill the stroma and collects as tiny bubbles under DM, which then coalesce to form the Type-1 BB.[49]

When the BB has expanded to approximately 7–8 mm, air injection is stopped and the needle withdrawn. A PDEK clamp can be used to facilitate bubble formation as it prevents the escape of air from the periphery of the PDL and almost eliminates risk of a Type-2 BB forming.[37] The bubble can be deflated by advancing the needle tip into the cavity of the bubble and aspirating the air, and the donor tissue then trephined with a 6 mm–7.5 mm trephine depending on the size of the bubble. Alternatively, without deflating the bubble, an incision is made at the junction of the bubble wall with the corneal stroma of sufficient size to allow entry of one blade of a Micro-Vannas scissors, with which the bubble wall is cut along the circumference of the bubble.

The graft scrolls with the endothelial side out. It is transferred to the AC after stripping off the host diseased endothelium. Graft insertion can be done with a Busin glide and forceps or with an injector. The graft is unfolded and centered like a DMEK graft by tapping and irrigation. The PDEK requires much less manipulation as compared to the DMEK tissue as it scrolls less than a DMEK graft.[116] An added advantage of PDEK is that, unlike DMEK, the donor tissue can be harvested from very young donors, with associated higher endothelial cell counts. A Type-1 BB has been obtained in donor tissue as young as 3 weeks[117] and PDEK has been performed with donor tissue from a 1-year old.[118]

Endothelial cell loss during donor tissue preparation is slightly less in PDEK than DMEK.[119] As the diameter of the PDEK graft is smaller than the DMEK graft, fewer endothelial cells are transplanted. However, it is postulated that lesser manipulations required during unrolling may compensate for it. As it is a relatively new procedure, graft failure rates and long-term outcomes are yet to be published.

## Conclusion

Lamellar corneal transplantation offers huge advantages and addresses the major risks associated with PK. Endothelial

rejection related graft failure is eliminated in DALK, and the eye is left stronger than after a PK on account of the retention of the PDL. Graft–host junction weakness and induced astigmatism are almost totally eliminated in EK as the incision size is very small and need for sutures is minimal or not at all. The most obvious limitation is that these surgeries are of no use in cases of full-thickness corneal pathologies or scars, which have to be treated with PK. Although there is around 6%–9% endothelial cell loss at the cut edge after PK,[120] surgical manipulation of the DM is minimal as compared to EK where the endothelial cell loss of up to 56% has been noted with 3-mm incisions.[121] Although LK is here to stay, PK will always remain for a "rainy day."

### Financial support and sponsorship

Nil.

### Conflicts of interest

Professor Harminder S Dua is inventor of the PDEK clamp but does not receive any royalties for this. He is also consultant for Dompe, Santen, Croma, Thea and speaker for Allergan, Visufarma and Bausch and Lomb.

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