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Original Article

Sinsky hook assisted roll preparation (SHARP): A modified technique for Descemet membrane endothelial keratoplasty donor preparation

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Abstract

Purpose: To describe a simple technique of sinsky hook assisted roll preparation (SHARP) for Descemet membrane endothelial keratoplasty (DMEK) donor preparation.
Methods: This experimental study was conducted at National Eye Bank, India with 40 optical grade human donor corneoscleral tissues found not suitable for surgery. 25 tissues were initially used to standardize the technique and remaining 15 for establishing the final technique. Donor corneal tissues were initially placed on a sterile Teflon block partially filled with tissue culture media. Initially, a partial thickness trephination was done followed by sinsky assisted 360° separation of the Descemet membrane (DM) from the underlying stroma (2 mm from the edge). The separation was further extended by 3–4 mm from the edge for 4–5 clock hours followed by bimanual peeling of the DM. This was followed by central 8 mm trephination. The primary outcome measures were a complete success (8 mm roll without peripheral edge tears) and partial success (8 mm roll with peripheral edge tears).
Results: DMEK roll was successfully peeled in 86.6% tissues (n = 13/15). Complete success was obtained in 66.6% tissues while partial success was obtained in 20% tissues. The median age of donor tissue was 45 years. The donor age of tissues, from which DMEK roll could not be obtained (2/12) was 15 days and 18 years.
Conclusion: SHARP is a simple technique of DMEK that does not require any sophisticated instruments.

Keywords: DMEK, Endothelial keratoplasty, Keratoplasty

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Introduction

Introduced by Melles in 2006, Descemet membrane endothelial keratoplasty (DMEK) has gained increasing popularity and interest as a method for posterior lamellar transplantation.[1]

In this procedure, the recipient's diseased Descemet-endothelium is replaced with the donor's healthy Descemet–endothelium complex. The major advantages of DMEK over other methods of endothelial keratoplasty is early visual rehabilitation with better visual outcomes and a low risk of graft rejection.[2–6] The other less discussed but often the most important advantage from the perspective of developing countries is its cost-effectiveness. It does not require sophisticated instruments like microkeratome.

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Despite these advantages, DMEK is not a widely practiced surgery. The significant limitations are difficult graft preparation, increased surgical manipulation and higher rates of early postoperative graft detachment. Besides, a steep learning curve leading to wastage of good quality donor tissues is a major concern in developing countries which may be a reason for poor acceptability of this surgical technique among corneal surgeons.

Several techniques that have been described in the past require some specialized instrument such as Muraine punch, Barron vacuum block, artificial anterior chamber, curvilinear forceps with half-moon shaped non toothed anterior segment, Y-hook instrument, etc.[4–6] In this experimental study, we describe a simple technique of *sinsky hook assisted roll preparation (SHARP)* for the preparation of DMEK graft.

Methods

Total of 40 optical grade donor corneal tissues were obtained from the National Eye Bank of which 25 were used to standardize our technique. Remaining 15 tissues [ten in McCarey-Kaufman medium (MK) media and five in Cornisol] were used for establishing the final technique. Any tissue preserved in MK media for >48 h and in Cornisol for >7 days were excluded. Also, tissues with DM folds involving the center of the cornea were excluded.

In the first 25 tissues, different size of trephines (9 mm, 9.5 mm, 10 mm, and 11 mm) were used to find the most appropriate size that would give us the best results. It was observed that with the trephine size of >9.5 mm, it was difficult to initiate the plane of dissection between the Descemet and posterior stroma. Dense adhesion was noted which often resulted in peripheral tears of DM roll. So we inferred that 9–9.5 mm is the most appropriate size for initial trephination. Several observations were made in the standardization which has been elaborated in the discussion section.

The major steps of the procedure have been depicted in Figs. 1 and 2. The corneoscleral rim is first placed on a Teflon block partially filled with tissue preservation media. An initial partial thickness trephination is done with a 9.5 mm manual trephine (Madhu trephines, India). At this step, it is essential not to apply undue force in order to avoid a full thickness punching. A useful sign for adequate depth of trephination is a ring formation, observed within the inner edge of trephine during this step. A broad ring is seen in the case of deep punching of the tissue while a narrow ring suggests a superficial trephination [Fig. 3A]. Alternatively a guarded trephine can also be used as it would be both precise and safe. However, in our experience, manual trephine also works well, especially if the ring sign is appreciated carefully. The tissue is then stained with Trypan blue 0.06% for 3 min, followed by a gentle wash with tissue fluid. A 360° separation of the Descemet-endothelium complex from the posterior stroma was obtained using a sinsky hook [Fig. 3B]. The separation plane extended 2 mm inside the edge of the partial thickness trephination. The angulation of sinsky hook with reference to the tissue plane was kept at around 30–45° for best results [Fig. 3C]. Inside out slicing movements were made with the sinsky hook for separating the Descemet endothelium complex. While making this slicing movement, it is essential to apply pressure only at stroma rather than the Descemet membrane (DM). In case, there is difficulty in separating the DM roll from the underlying stroma, the stroma can be held with limbs or plain forceps at the site of partial thickness trephination and pulled outwards while proceeding with the sinsky assisted dissection of the Descemet endothelium complex. This step makes the underlying tissue taut, and the edge of DM roll more prominent leading to ease in tissue dissection at the appropriate plane. Also, depressing the

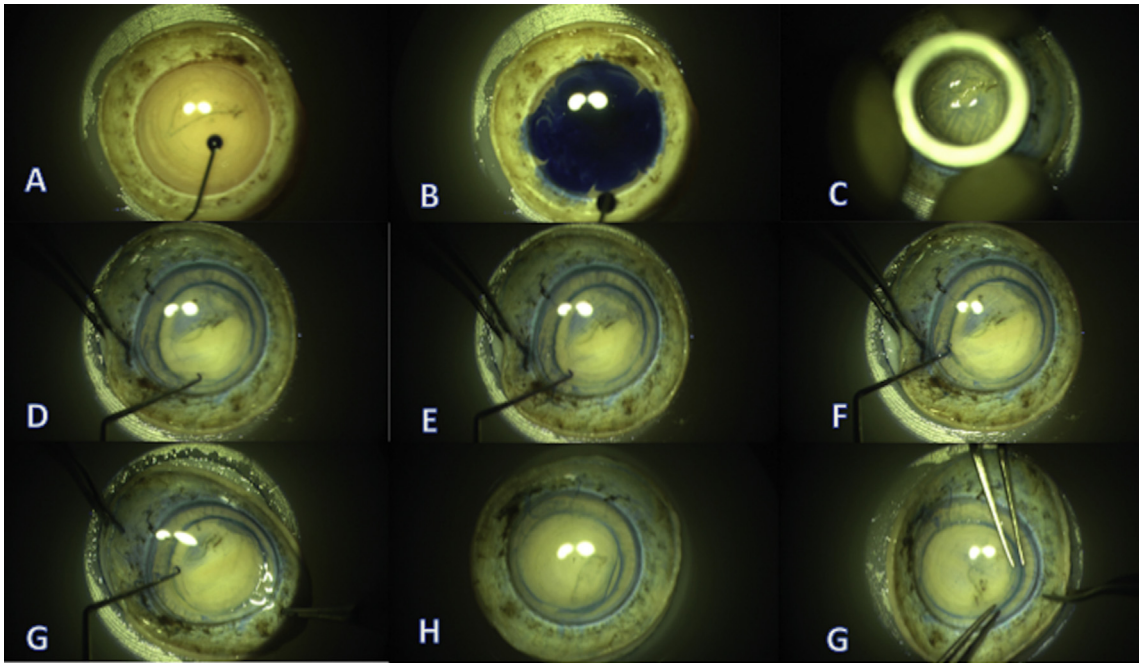

Fig. 1. *A*- Donor corneal tissue on Teflon block; *B*- Staining of Descemet Membrane with 0.06% Trypan blue; *C*- Partial trephination of donor tissue from the endothelial side using 9.5 mm trephine; *D/E/F/G*- inside-out movement of Sinsky hook to separate the Descemet membrane from an underlying stroma; *H*- extending the plane of separation to about 3–5 mm towards centre; *I*- Bimanual technique of peeling.

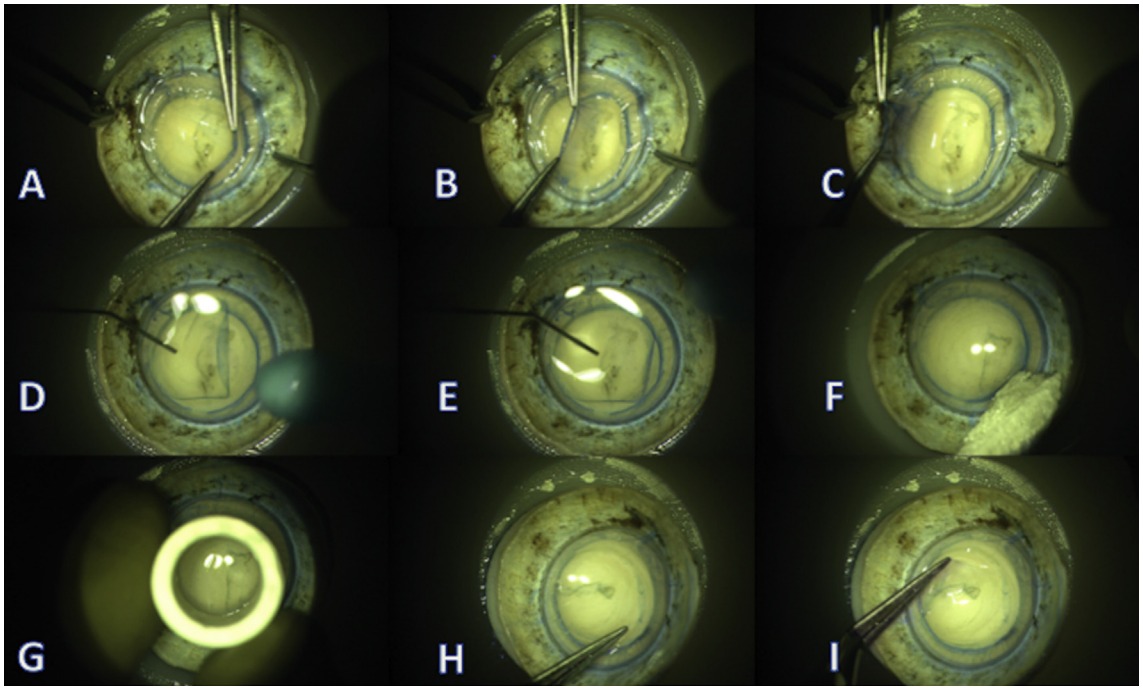

Fig. 2. *A/B/C*- the bimanual technique of membrane peeling; *D/E/F*- repositioning of freed Descemet roll; *G*- trephination of the Descemet roll using 8 mm trephine; *H/I*-complete peeling of the Descemet roll.

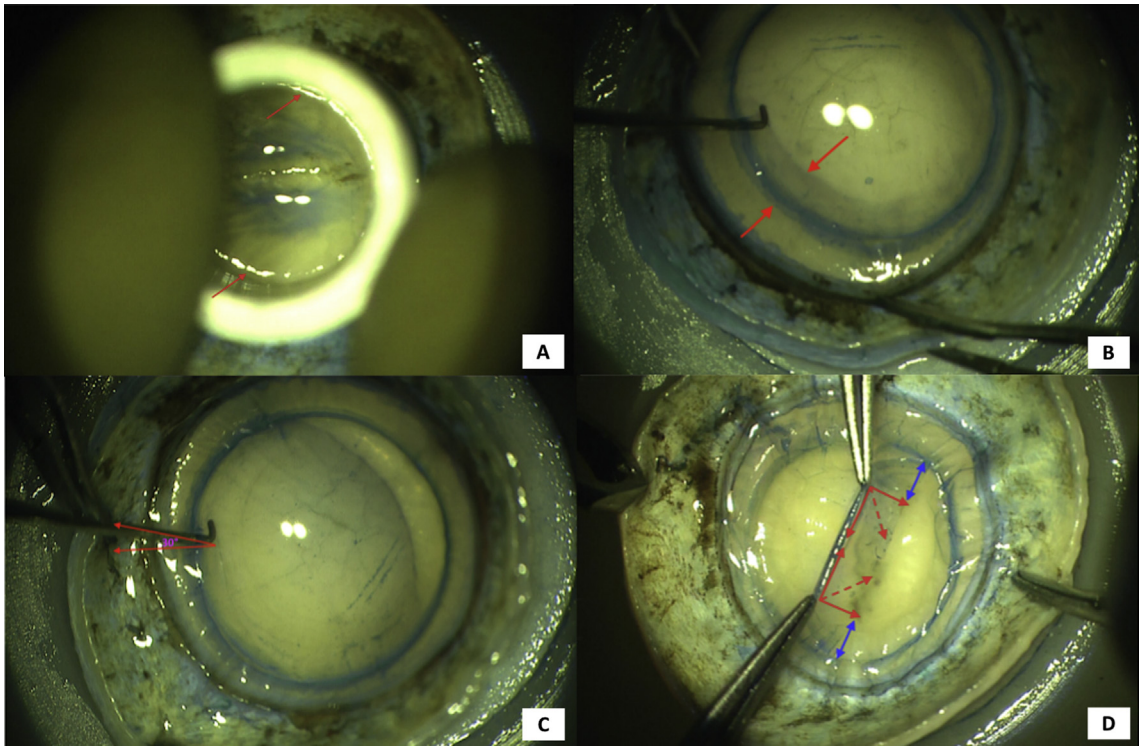

Fig. 3. (A). Ring sign shown by red arrows, giving indirect guidance about the depth of trephination. (B). 2 mm of a peripheral frill of Descemet membrane separated using Sinsky hook as shown by red arrows. (C). Direction of pushing force of Sinsky hook indicated by red arrows. (D). Vectors showing different forces, in red arrows, acting at the Descemet stroma junction during bimanual peeling. Blue arrows suggest the traction free zone.

peripheral tissue (beyond the edge of partial thickness trephination) makes the edge of DM roll more prominent leading to ease in tissue dissection. After obtaining a 360° frill, the separation plane is further extended 3–4 mm from the edge to the extent of around 4–5 clock hours. This site is now placed diagonally opposite to the surgeon. The assistant supports the Teflon block and holds the tissue firmly with a toothed forceps. At every step, it is essential to have good

assistance for holding the corneoscleral button in position. However, the same could be done, without any assistants support, using a suction teflon block for graft preparation. Bimanual peeling of the Descemet endothelium complex is initiated from the same site by holding the edge of the frill with two McPherson forceps 2–3 clock hours apart [Fig. 3D]. Alternately a suture tying forceps can also be used. The tissue is then gently lifted and pulled towards the surgeon leaving it attached for around 1–2 mm at the opposite end. The DM roll is then reposited back. The tissue is now trephined with an 8 mm trephine. At this step, it is essential to note that if there are any peripheral micro tears or ripped off area, then the placement of trephine should be such that these areas are avoided as far as possible in the final graft. However, if there are no peripheral tears, then a well-centered trephination should be attempted. It is important to keep the tissue wet throughout the procedure by intermittent use of donor preservation media. The marking of the DM roll can be performed by any of the currently available techniques of DMEK preparation.[4–9]

Results

DMEK roll was successfully peeled in 86.6% of tissues (n = 13/15). Complete success was obtained in 66.6% of tissues. While partial success was obtained in 20% of tissues. In two cases, there was complete failure to peel the DM roll. In one of these case, the DM got ripped right across the center of the graft. In another case, multiple points of adhesion between the DM and stroma was observed, and hence DM roll could not be prepared. The median age of the donor tissue was 45 years. The donor age of the tissues, from which DMEK roll could not be obtained (n = 2/15) was 15 days and 18 years. The three tissues which had peripheral edge tear/rip off, the extent of the defect was less than 1 × 1 mm after final trephination with an 8 mm trephine. The details of the donor tissues have been described in Table 1.

Ten tissues were preserved in McCarey-Kaufman (MK) medium while five were in Cornisol. The comparison of various parameters between the two groups has been described in Table 2. The median donor age was lower in the cornisol group. The donor endothelial density and the death enucleation time (DET) were comparable between the two groups. There was no difference in the success rate between the two groups (p = 1.0). Regression analysis was attempted to know the impact of individual factors such as age, sex, DET and preservation media on the success rate of donor tissue preparation, however, due to the relatively small number of tissues in the failure group, it could not be done.

Discussion

DSAEK and DMEK are the most commonly performed techniques of EK. Low risk of rejection and cost of microkeratome has often led the surgeons to prefer DSEK (Descemet stripping endothelial keratoplasty) over DSAEK with relatively inferior outcomes in terms of visual function.[9] DMEK can be extremely useful in this circumstance. However, the primary deterrent for the wide acceptance of this technique is a steep learning curve and fear of wastage of good quality donor corneal tissue. The fear is justified as there is a wide demand-supply gap of donor corneal tissues in these countries.

We describe a technique that can be easily mastered and performed with the use of routinely used keratoplasty instruments without the need for any specialized or expensive instruments. The initial attempts for standardization of the technique provided us some valuable observations. Some of our observations reinforce the earlier findings by Kruse et al.[5,10], Tenkman et al.[6] and Schlotzer-Schrehardt et al.[11].

Firstly, the adhesion between Descemet and stroma is more towards the periphery which increases the chances of a rip off of DM in the periphery. Thus, the use of a trephine size of 9–9.5 mm may reduce the chances of DM tear while peeling it off.

Secondly, while peeling the DM, we realized that the chances of the tear are high at the edges of trephination. When a 360° edge of DM was made free with the assistance of a sinsky hook, the instance of DM tear was almost negligible. Thus creating a 360° frill of free DM to the extent of 1–2 mm [Fig. 3B] is an essential step in achieving a successful DM roll.

Thirdly, while creating the frill with sinsky hook, it is essential to apply a pushing-down force at the DM-stroma junction at an angle of 30–45° [Fig. 3C]. The potential space between

Table 1. Details of the Donor tissues used for the Sinsky Hook Assisted Roll Preparation Technique.

Sl no	Age in years	Preservation Media	Pre-op Specular count	DET in hours	Outcome
1	55	MK	NA	13	Complete success*
2	54	MK	NA	7	Complete success
3	45	MK	1669	3	Partial success**
4	18	Cornisol	2506	12	Complete success
5	18	Cornisol	2435	6	Partial success
6	65	MK	NA	5	Complete success
7	46	MK	NA	4	Complete success
8	32	Cornisol	2257	6	Complete success
9	37	Cornisol	2033	12	Complete success
10	37	MK	1828	1	Partial success
11	40	MK	2024	8	Complete success
12	60	MK	1742	1	Complete success
13	64	MK	NA	5	Complete success
14	64	MK	1975	3	Failure
15	15 days	Cornisol	NA	6	Failure

MK- Mc Carey Kaufman; DET: Death enucleation time.
* Complete success- DMEK scroll with no peripheral edge tears.
** Partial success (8 mm DMEK scroll with peripheral edge tears/ripped of area).

Table 2. Comparison of different donor corneal parameters preserved in two types of media used for donor preservation.

Parameters		MK Media (Median, Range)	Cornisol (Median, Range)	P value
Age (years)		54.5 (37–65)	25 (18–37)	0.005
DET (h)		4.5 (1–13)	9 (6–12)	0.62
ECD (cells/mm^2)		2142 (2024–2669)	2346 (2033–2506)	0.08
Outcome (Number of cases)	Complete success	7	3	1.0
	Partial success	2	1	
	Failure	1	1	

MK, McCarey-Kaufman; DET, Death enucleation time; ECD, endothelial cell density.

DM and stroma, which is understandable due to their different source of development during embryogenesis, has been successfully exploited for the big bubble deep anterior lamellar keratoplasty technique.[11,12] Thus, when a pushing force was applied at 30–45° to the stroma, near DM stroma junction, the obvious plane of separation was between DM and stroma. However, at this step, it is important to remember not to apply direct force to the DM as it would lead to DM tear.

Additionally, it is better to re-stain the DM after partial trephination so that the edge of trephination becomes clearly visible, thereby facilitating the step of frill formation and edge lifting. It is essential to do an initial trephination that is superficial only, as deep trephination would lead to difficulty in initiating the process of DM separation. The ''ring sign'' as described in the result section is extremely helpful in this regard. A thin ring suggests shallow while a broad ring suggests a deeper plane of trephination [Fig. 3A].

After a 360° frill separation, the DM roll was separated for 3–4 mm towards the center for an extent of 4–5 clock hours. During standardization, we had attempted to peel it bimanually from the initial 1 mm frill only, but it was much easier to peel after the DM was separated for about 3–4 mm towards the center. This may be due to a longer arc of force, hence more controlled, with 3–4 mm frill when compared to a shorter arc of force with a 1 mm frill. The application of several principles of physics is helpful in better understanding of this technique.[13] We strongly recommend bimanual peeling instead of a single-handed peeling as in a single-handed peeling the force of traction appears at multiple points, while in bimanual peeling the force of traction appears at four points.

Additionally, in bimanual peeling, the force is applied over a wider area resulting in less traction at each point (pressure = force/area, assuming that equal force is used). Also, it is important to note that the horizontal vector component of the force neutralizes the traction exerted by each other in bimanual peeling (shown by the red arrows in Fig. 3D). Lastly, since the peripheral frill has been freed using sinsky (as highlighted by the blue arrows in Fig. 3D), the traction points are primarily located within the central 6 mm zone, and it has been proven that the adhesive forces are minimal between DM and stroma within this area of the cornea.[5,6] All the factors discussed above would reduce the risk of DM tears during DMEK if appropriately followed.

We recommend the beginners to go for peeling at a slow speed. As per Newton's rule F = ma where f stands for force, m for mass, and a for acceleration. Assuming that the mass of DM remains constant, the force (or in other words the traction at DM stroma junction) is directly proportional to the acceleration (which is the speed of peeling in this scenario). Thus the surgeon must always remember ''Go steady, Go slow''. Any sudden jerky movement or too fast peeling can lead to excessive traction at the stroma-DM junction with consequent DM tear and hence must be avoided.

Keeping a safe margin of around 1.5 mm (initial trephine 9.5 mm, final trephination 8 mm) allows for the exclusion of any torn or ripped off areas of DM at the time of the last trephination.

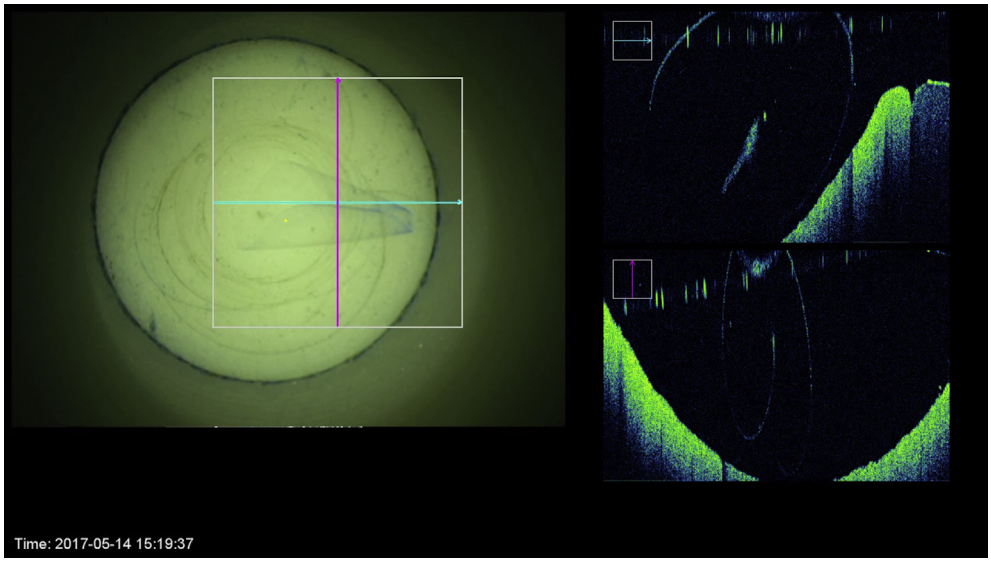

Fig. 4. Descemet Membrane Roll as seen in intra-operative OCT.

Lastly, we attempted to evaluate if there is any advantage of using intraoperative optical coherence tomography while DMEK roll preparation. We found that it does not have any significant advantage during DM roll peeling. However, it can delineate the DM-stroma junction which may be useful in some situations. Also, it can provide an idea about the type of DM roll prepared (Fig. 4) that may be helpful in the unfolding of donor tissue intra-operatively.

The success rate of our technique is relatively less compared to other studies reporting success rate as high as 99%.[6] This is mainly due to the inclusion of young donors. If young donors are excluded then our success rate would be nearly 100%. Most experts recommend a donor age of 55 years or above for successful DMEK roll preparation.[5] During standardization of our technique, we realized that though it is not impossible to peel a young donor, the primary difficulty with young donors is a tight DM roll. A tight roll will be extremely difficult, at least for the beginners, to unroll inside the eye. It is better for the beginners to select a donor corneal tissue of age between 55 and 70 years initially.

To conclude, the SHARP technique for DMEK donor preparation is a simple, easy to learn and a cost-effective technique that can be performed with the help of a few commonly used keratoplasty instruments. It combines the different lessons that have been learned over the decades by different researchers. Till the submission of this manuscript, we have used this technique in two cases of PBK with final BCVA of 20/30 and 20/40 at six months with a clear cornea, and endothelial cell loss of 28% and 30% respectively. Although we found our technique extremely useful, we would like to advise the readers to practice this in experimental conditions first and then to use it for their patients.

Performing electron microscopy on the peeled DM could have provided us with useful information, but it could not be performed due to lack of facilities for the same. Besides, we could not evaluate the endothelial cell loss since it was an experimental study and DMEK rolls were not used in any patients of endothelial dysfunction. It may be argued that the usefulness of this technique can be verified only after its use in human subjects, however, we believe that the DMEK roll preparation is the most challenging step of DMEK surgery and our work describes this in a simple technique with the help of common but often ignored concepts of physics.

Conflict of interest

None of the authors has conflict of interests to declare.

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