UDC: 617.713:576.32/36

Di Zazzo A.

# SIMPLE LIMBAL EPITHELIAL TRANSPLANTATION (SLET): REVIEW OF INDICATIONS, TECHNIQUE, MECHANISM, OUTCOMES AND IMPACT (LITERATURE REVIEW)

*Cornea and Refractive Surgery Services, Complex Operative Unit in Ophthalmology, University Hospital Campus Bio Medico of Rome, Italy*

**Key words:** *simple limbal epithelial transplantation, cultivated limbal epithelial transplantation, conjunctival limbal autograft, limbal stem cell deficiency, chemical burn*

***Introduction***

The cornea is covered by a very delicate and thin layer of stratified squamous epithelium, which is continuously renewed throughout life by stem cells present at the limbus [1]. Limbal stem cell deficiency (LSCD) is a clinical condition, characterized by progressive vascularization, conjunctivalization and scarring of the corneal surface due to irreversible traumatic or inflammatory damage to these limbal stem cells (LSC). In severe and chronic cases LSCD can lead to visual impairment and even blindness [2]. Fortunately, this potentially blinding disease can be treated by transplanting healthy limbal tissue containing the stem cells from a normal donor eye, a process called limbal stem cell transplantation (LSCT). Depending on the source of the donor tissue, LSCT can either be autologous (from the unaffected fellow eye of the same person) or allogeneic (from another person).

Several different surgical techniques of LSCT have emerged with time. The conventional approach, first described by Kenyon and Tseng in 1989 for autologous transplants is popularly called conjunctival-limbal autografting (CLAU) [3]. In this technique 3 to 6 clock hours of limbal tissue is harvested from the healthy eye and directly transplanted to the affected eye. However, this puts the healthy eye at some risk of developing iatrogenic LSCD. To avoid this risk, Pellegrini et al in 1997 described cultivated limbal epithelial transplantation (CLET) in which tiny amounts of limbal epithelial cells obtained from the donor eye could be expanded as a sheet in the laboratory [4]. Unfortunately, cell expansion necessitates a clinical grade laboratory with regulatory approval, which is extremely expensive to build and maintain. In 2012 Sangwan et al described a new technique of LSCT called simple limbal epithelial transplantation (SLET), which combined the advantages of CLAU and CLET while avoiding the limitations of both approaches [5]. Since then, SLET has become a popular technique of LSCT, particularly in the developing world. In this review we will enumerate further the indications, surgical technique, mechanism of action, outcomes and impact of SLET in patients with blinding LSCD.

***Proposed indications***

Primarily, SLET is indicated in cases of unilateral LSCD where the donor limbal tissue can be harvested from the healthy fellow eye of the same patient, without the risk of immunological rejection and requirement for systemic immunosuppression. One the most common causes of unilateral LSCD is chemical burns [2] and hence, this condition forms the major indication for autologous SLET. All the large-scale studies on SLET have been on autologous procedures and on patients with LSCD secondary to chemical burns [6-8]. However, SLET has also been described in eyes with LSCD secondary to ocular surface squamous neoplasia excision (OSSN) and multiple surgical interventions [9-11]. Other conditions that have been successfully treated with SLET include both primary and recurrent pterygia [12,13] and failed prior LSCT [14,15].

Allogeneic SLET for bilateral LSCD has also been reported in few case reports [16,17]. The first was in a patient with bilateral chemical burns [16] and the second case was in a patient with bilateral dry eye [17]. In both cases, cadaveric allogeneic donor tissue was used. It is generally understood that patients with bilateral LSCD undergoing allogeneic SLET will need long-term systemic immunosuppression for graft survival [16], although long-term outcomes or systemic immunosuppression protocols in such cases have not been reported yet. Cadaveric allogeneic SLET has also been used in the treatment of severe acute chemical burns to achieve faster epithelialization of the ocular surface [18].

***Pre-operative considerations***

Like any organ or tissue transplantation procedure, the main factor determining the outcome of SLET is the health of the donor limbus. Therefore, a careful preoperative inspection of the donor site to ensure that

it is viable is critical. Typically, the superior limbus is preferred as the limbal palisades are more in number at this location. In the affected eye, the following are absolute contraindications for SLET: (i) dry surface (defined as repeated Schirmer's I score with anesthesia of less than 15mm or presence of corneal or bulbar conjunctival keratinization), (ii) blind eye with no visual potential; (iii) disorganized anterior segment (adherent leucoma) and (iv) presence of uncorrected adnexal pathologies like lagophthalmos, ectropion, entropion, trichiasis and dacryocystitis. Since SLET is an epithelial regenerative procedure, it does not correct corneal stromal opacification. Thus, cases with severe stromal opacification (leucoma) will require additional corneal stromal replacement with an anterior lamellar or penetrating keratoplasty (PK). An anterior segment optical coherence tomography (AS-OCT) of the diseased eye is extremely useful in this context. It not only reveals the underlying stromal thickness (therefore alerting for extremely thin areas likely to perforate during dissection of the conjunctivalized pannus), the infra-red photograph of the cornea also shows the degree of opacification of the underlying stroma. The ideal cases for autologous SLET are those with no history of any trauma, inflammation or surgery in the donor eye; with the following characteristics in the affected eye: (i) wet ocular surface without unaddressed adnexal pathologies; (ii) no severe symblepharon that is reaching up to the cornea and (iii) clear to translucent underlying corneal stroma. Those cases which have severe symblepharon will also require additional conjunctival autografts, either during or after SLET. It is recommended that cases with severe stromal opacification or disorganized anterior segment, which would also need a corneal grafting undergo conjunctival limbal grafting and not SLET.

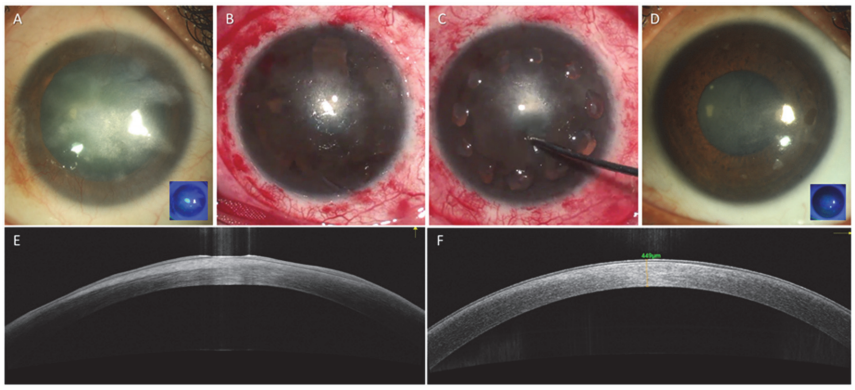
***Surgical technique***

Anesthesia: For children, general anesthesia is mandatory. For adults the limbal biopsy can be harvested from the donor eye under topical anesthesia, but beginners may find using peribulbar or sub-tenons anesthesia preferable. The affected eye in adults is always given a peribulbar block.

Preoperative vasoconstriction: It is recommended to use 2-3 applications of brimonidine tartrate 0,15% and phenylephrine 5% eye drops alternatingly 5-10 mins before shifting the patient to the operating room. This significantly reduces intra-operative bleeding in both the donor and recipient eye.

Donor eye: The donor limbus should not be marked directly with a skin-marking pen, as the alcohol in the ink can damage the delicate limbal stem cells. "One-clock hour" or roughly 3,5-4 mm graft piece should be measured with a caliper and marking should be done slightly behind the limbus on the conjunctiva. A conjunctival bleb is created with fluid just behind the selected area of the biopsy and a limbus based conjunctival flap is lifted until the insertion of the Tenons capsule at the limbus. This area is lined by blood vessels and marks the posterior boundary of the limbus. Dissection with a no.15 blade held as flat as possible is then carried forward in the same plane until the grey clear cornea is visible. The flap is reposed, and the conjunctival part is excised off. The limbal tissue is excised using a pair of Vannas or Wescott scissors. The tissue is preserved wet in balanced salt solution (BSS).

Recipient eye: If symblepharon is present which prevents insertion of the speculum, it should be excised first flush to the cornea. A peritomy is then performed all around about 2-3 mm from the corneal margin. Dissection is then carried forward using a pair of Vannas scissors to release the conjunctivalized pannus covering the cornea from the limbus 360 degrees before proceeding centripetally. The pannus in then removed from the corneal surface using a combination of sharp and blunt dissection. It is not necessary to remove stromal scar tissue till a clear plane is reached. The surrounding conjunctiva is then recessed by using tenotomy scissors. The human amniotic membrane (hAM - basement membrane side up) is then placed and secured over the recipient cornea with the help of fibrin sealant. It is ensured that the hAM is tucked under the conjunctival edge in all quadrants. The hAM is smoothened out over the cornea using a blunt spatula to ensure that there are no folds. The limbal tissue is then removed from the BSS and cut approximately into 6-10 pieces with the help of Vannas scissors. These pieces are placed (epithelial side up) in the mid-periphery of the cornea in a concentric pattern over the hAM (Figure 1). The correct orientation of the small pieces can be identified from the pigmentation and/or smooth surface of the epithelial side and white fibrous strands on the stromal side. Care is taken to ensure that the pieces of limbal tissue are not placed over the pupillary area or on the limbus. A drop of fibrin sealant is placed over each piece to ensure that they adhere to the AM. After waiting for at least one minute for the fibrin glue to polymerise over the limbal pieces, a soft bandage contact lens (BCL) is placed. Care is taken to cut away the excess glue that may be sticking to the speculum as it is removed and not to pull at the strands which may dislodge the film of glue on the surface that is holding the pieces in place.




**(A)** Pre-operative appearance showing superficial corneal vascularization and scarring with central epithelial defect (inset: fluorescein stained photograph);
**(B)** Intra-operative image after removal of the fibrotic conjunctivalized pannus from the corneal surface;
**(C)** Intra-operative image showing the circumferential arrange of limbal transplant pieces being secured to the human amniotic membrane graft with fibrin glue;
**(D)** At 6-months post-operatively the corneal surface is stable, avascular and epithelized (inset: fluorescein stained photograph);
**(E)** Pre-operative anterior-segment optical coherence tomography (AS-OCT) image showing highly-reflective irregular conjunctival epithelium with superficial stromal haze and central epithelial defect;
**(F)** Post-operative AS-OCT image showing transparent regular low-reflective corneal epithelium with reduction in stromal haze

**Figure 1. Typical post-operative outcome after autologous simple limbal epithelial transplantation (SLET) for unilateral chemical burn induced limbal stem cell deficiency (LSCD)**

***Post-operative regimen***

Post-operatively topical prednisolone acetate 1% drops are started 6 times a day for 1 week and then tapered every week over the next 6 weeks in both the recipient and the donor eyes while topical moxifloxacin 0,5% is started 4 times/day in both eyes and is continued until the epithelial defect heals. The BCL is removed at the 1-week post-operative visit. The cornea is stained with fluorescein and if complete epithelial healing has not occurred, then a BCL is replaced. It is also imperative to check the epithelial healing at the donor site simultaneously. The progress of the recipient eye is monitored until the epithelium completely heals. In very young children, a suture tarsorrhaphy can be performed and kept in place for a couple of weeks to prevent the loss of the BCL or transplants in the immediate postoperative period because of the risk that the children may inadvertently rub their own eyes.

***Clinical outcomes***

The results of SLET in different indications have been enlisted in Table 1. Only one study compared CLAU and SLET in 10 eyes each, and found both surgical techniques to be equally effective in achieving a stable epithelized ocular surface and for regression of corneal vascularization [19]. The 3 major studies from different groups reported the outcomes in a total of 223 eyes [6-8]. The primary indication was LSCD secondary to chemical burns in 213 (96%) of 223 eyes. At a mean follow-up period of 1,2 years, 173 (77,6%) eyes had a clinically successful outcome with a stable epithelized and avascular corneal surface, while 153 (68,6%) had a two-line improvement in visual acuity.

The outcomes of sequential secondary surgeries such as PK and deep anterior lamellar keratoplasty (DALK) after SLET have also been reported [20, 21]. In seven eyes of 7 patients who underwent PK 9,5 ± 11,9 months after SLET, 6 eyes maintained a clear graft 15,1 ± 5,4 months after PK [20]. Another report of 11 eyes of children who underwent DALK following SLET for unilateral severe chemical injury with LSCD with follow-up of 13 ± 4,6 months following DALK reported anatomical success in 8 (72%) eyes and visual acuity improvement in 6 (54%) eyes [21].

***Mechanism of action***

Mittal et al studied four eyes that had undergone SLET and were serially imaged using fluorescein staining to elegantly demonstrate that epithelial cells grew centrifugally from each explant on the surface and merged to form a sheet of epithelium [22]. In all four eyes, complete ocular surface epithelialization occurred within 14 days. The explants were not visible after two months in three eyes after complete epithelialization was achieved. They also observed that all the explants placed on the surface were not active and thus there was considerable variation in explant activity. However, the corneal surface around the inactive explants were epithelialized by epithelium arising from the surrounding explants. This study clearly demonstrated that hAM acts as a substrate for secure attachment for the epithelial cells and supports their proliferation and migration in eyes where SLET has

been performed. Amescua et al used ultra-high-resolution optical coherence tomography in an eye where SLET was performed and showed the persistence of hAM while the epithelial cells grew and proliferated over it [9]. In another series, excised corneal buttons from eyes undergoing PK after SLET were examined and it was confirmed with both histopathological examination and immunohisto-chemistry, that not only was the newly regenerated epithelium of corneal phenotype (CK3+, CK12+, CK19-, MUC5AC-) but also that there was focal retention of stem cells (ABCG2+, ΔP63α+) in the basal epithelium layer of the newly regenerated epithelium [6]. This study confirmed that these cells were derived from the explants that were placed on the cornea during SLET. On histopathological examination the presence of a thick periodic-acid Schiff (PAS) positive membrane was noted below the newly regenerated epithelium. This also confirmed the persistence of hAM after SLET.

***Possible complications***

No studies have reported serious adverse outcomes in the donor eye. Localized, non-progressive focal LSCD were reported in two cases of at the donor site after SLET, which did not affect visual acuity [7, 23]. Pyogenic granuloma was also reported in 2 donor eyes at the site of limbal tissue excision [6]. The most common complication in the recipient eye after SLET was focal recurrence of LSCD which has been reported in 18% to 31% eyes [6-8]. Most clinical failures after SLET occurred in the first six months after surgery [7, 23]. Early complications included hemorrhage under the hAM which usually resolves without any consequence [6]. Early loss of the SLET transplants and detachment of the hAM also can lead to failure [6, 24]. Pre-existing symblepharon, if not addressed at the time of surgery with a conjunctival autograft, was noted to be a risk factor for recurrent conjunctivalisation and failure of the primary procedure [6-8]. Penetrating keratoplasty done simultaneously with SLET was also a risk factor for early failure [6-8]. Indications such as acid injury was noted to have a higher rate of failure than SLET performed for other forms of chemical burns such as alkali injury [6, 8]. Other rare complications that have been reported are sterile keratitis, microbial keratitis, persistent epithelial defect which could lead to thinning and perforation if not addressed in time, and recurrence of corneal neovascularization.

***Conclusions***

The clinical efficacy of SLET is now well established in large studies with adequately long follow-up [6-8]. The persistence of LSC on the corneal surface after SLET has also been demonstrated [6] and it's mechanism of corneal epithelial healing is well understood [22]. Although no randomized controlled trial has so far been published comparing head-to-head the outcomes of SLET, CLAU or CLET, the advantages of SLET over the other techniques are obvious. In resource limited settings, the option of CLET is largely theoretical and the choice for the corneal surgeon is essentially between CLAU and CLET. Since SLET can be achieved within one-clock-hour shaped graft piece of limbus instead of CLAU, which does the same purpose within three to six-hours graft, so the obvious choice is straight forward. However, it is important to understand that SLET alone is not effective in cases with severe symblepharon, which have both limbal and conjunctival deficiency and therefore need both LSCT and conjunctival grafting. It is in these cases that SLET and CLAU (modified as mini-CLAU) can perhaps be combined [25]. Another exceptional scenario where CLAU may hold an advantage over SLET is cases of complex reconstruction requiring conjunctival, limbal and corneal grafting. In these cases, the corneal graft remains at high-risk of immunological rejection and may need being replaced in the future, therefore placing the limbal graft in its anatomical location beyond the cornea as in CLAU may be advantageous over SLET, where the limbal transplants will be lost if the corneal graft is replaced.

The advent of SLET has made life easier for corneal surgeons, particularly those in the developing world dealing with a huge burden of unilateral LSCD due to chemical burns [2]. The technique has a relatively short learning curve and surgeons otherwise inexperienced in ocular surface surgery are quickly able to replicate the same results as experienced ones [6]. This easy replicability has also allowed SLET to spread to other specialties like oculoplastics, where it has been successfully adapted to both treating and preventing LSCD after extensive OSSN excision by surgeons who were otherwise naive to LSCT [10, 11]. This reliability and replicability of SLET is also demonstrated in the consistent outcomes reported across large studies by different surgeons in different geographies [6-8].

One can imagine SLET as in vivo CLET, where the cell expansion takes place on the surface of the eye instead of a petri-dish using the natural environment, growth factors and tears as tissue-culture reagents. The central dogma of CLET that one-clock hour graft of limbal tissue is enough to resurface the entire cornea is reaffirmed by SLET. No wonder that those who developed SLET had spent close to a decade performing CLET themselves [26], before they chanced upon the idea. However, the two paradigms that SLET challenges are the fate of LSC when placed on the cornea instead of the limbal niche and the pattern of corneal epithelial wound healing. As the individual pieces in SLET have no conjunctival component, the epithelial cells grow from all sides leading to rapid corneal epithelialization unlike circumferential followed by centripetal healing as seen physiologically in corneal abrasions and that after CLAU [27,28]. What SLET has also taught us is how little we yet understand of the ocular surface physiology in general and LSC in particular close to three decades after the discovery of these unique stem cell at the limbus.

Table 1

**Summary of published studies on outcomes of simple limbal epithelial transplantation (SLET) for the treatment of various ocular surface pathologies**

| Most common indication (number of eyes)‡ | Author | Year | Country | Study design | Number of eyes | Intervention | Amount of limbal tissue harvested | Eyes ratio with completely epithelized, stable, avascular corneal surface (number of eyes) | Eyes ratio with 2-line improvement in BCVA (number of eyes) | Eyes ratio which underwent simultaneous/ subsequent LK/PK | Mean follow-up in years (range) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Chemical burn (5) | Sangwan et al. | 2012 | India | R | 6 | SLET | 2×2 mm / 1-clock-hour | 100 (6/6) | 100 (6/6) | None | 0.8 (0.6-1) |
| Chemical burn (2) | Amescua et al. | 2014 | U.S.A | R | 4 | SLET | 2×2 mm / 1-clock-hour | 100 (4/4) | NA | None | 0.6 (0.5-0.75) |
| Simultaneously after primary pterygium excision | Hernadez-Bogantes et al. | 2015 | Mexico | R | 9 | Mini-SLET | 2×2 mm from ipsilateral eye | 100 (9/9) | NA | NA | 0.67 |
| Chemical burn (125) | Basu et al. | 2016 | India | P | 125 | SLET | 2×2 mm / 1-clock hour | 76 (95/125) | 75 (94/125) | 8 (10/125) | 1.5 (1-4) |
| Chemical burn (62) | Vazirani et al. | 2016 | Multi-national | R | 68 | SLET | 1-2-clock hours | 84 (57/68) | 65 (44/68) | 7 (5/68) | 1 (0.5-4.9) |
| Chemical burn (4) | Quieroz et al. | 2016 | Brazil | R | 4 | SLET | 4×2 mm | 50 (2/4) | 24 (1/4) | None | 0.5 |
| Chemical burn (26) | Gupta et al. | 2018 | India | P | 30 | SLET | 1-2-clock hours | 70 (21/30) | 50 (15/30) | 10 (3/30) | 1.1 (0.5-3.4) |
| Simultaneously after OSSN excision | Kaliki et al. | 2017 | India | R | 8 | SLET | 1-clock-hour from ipsilateral or contralateral eye | 100 (8/8) | NA | NA | 1 |
| Simultaneously after recurrent pterygium excision | Mednick et al. | 2018 | Canada | R | 4 | SLET | 4×2 mm | 100 (4/4) | NA | NA | 1.2 (0.7-2.5) |
| R = randomized,<br>P = prospective cases series | | | | | | | U = unclear,<br>NA = not available | | | | |

# REFERENCES

1. Kinoshita S., Kiorpes T.C., Friend J. et al. Limbal epithelium in ocular surface wound healing // Invest. Ophthalmol. Vis. Sci., 1982, v.23, p.73-80.
2. Vazirani J., Nair D., Shanbhag S. et al. Limbal stem cell deficiency-demography and underlying causes // Am. J. Ophthalmol., 2018, v.88, p.99-103.
3. Kenyon K.R., Tseng S.C. Limbal autograft transplantation for ocular surface disorders // Ophthalmology, 1989, v.96, p.709-722.
4. Pellegrini G., Traverso C.E., Franzi A.T. et al. Long-term restoration of damaged corneal surfaces with autologous cultivated corneal epithelium // Lancet, 1997, v.349(9057), p.990-993.
5. Sangwan V.S., Basu S., MacNeil S. et al. Balasubramanian D. Simple limbal epithelial transplantation (SLET): a novel surgical technique for the treatment of unilateral limbal stem cell deficiency // Br. J. Ophthalmol., 2012, v.96, p.931-934.
6. Basu S., Sureka S.P., Shanbhag S.S. Simple Limbal Epithelial Transplantation: Long-term clinical outcomes in 125 cases of unilateral chronic ocular surface burns // Ophthalmology, 2016, v.123, p.1000-1010.
7. Vazirani J., Ali M.H., Sharma N. et al. Autologous simple limbal epithelial transplantation for unilateral limbal stem cell deficiency: multicentre results // Br. J. Ophthalmol., 2016, 100, p.1416-1420.
8. Gupta N., Joshi J., Farooqui J.H. et al. Results of simple limbal epithelial transplantation in unilateral ocular surface burn // Indian J. Ophthalmol., 2018, v.66, p.45-52.
9. Amescua G., Atallah M., Nikpoor N. et al. Modified simple limbal epithelial transplantation using cryopreserved amniotic membrane for unilateral limbal stem cell deficiency // Am. J. Ophthalmol., 2014, v.158, p.469-475.
10. Kaliki S., Mohammad F.A., Tahiliani P. et al. Concomitant simple limbal epithelial transplantation after surgical excision of ocular surface squamous neoplasia // Am. J. Ophthalmol., 2017, v.174, p.68-75.
11. Mittal V., Narang P., Menon V. et al. Primary simple limbal epithelial transplantation along with excisional biopsy in the management of extensive ocular surface squamous neoplasia // Cornea, 2016, v.35, p.1650-1652.
12. Hernandez-Bogantes E., Amescua G., Navas A. et al. Minor ipsilateral simple limbal epithelial transplantation (mini-SLET) for pterygium treatment // Br.J. Ophthalmol., 2015, v.99, p.1598-600.
13. Mednick Z., Boutin T., Einan-Lifshitz A. et al. Simple limbal epithelial transplantation for recurrent pterygium: A case series // Am. J. Ophthalmol. Case Rep., 2018, v.12, p.5-8.
14. Bhalekar S., Basu S., Lal I. et al. Successful autologous simple limbal epithelial transplantation (SLET) in previously failed pediatric limbal transplantation for ocular surface burns // BMJ Case Rep., 2013, doi: 10.1136/bcr-2013-009888.
15. Basu S., Mohan S., Bhalekar S. et al. Simple limbal epithelial transplantation (SLET) in failed cultivated limbal epithelial transplantation (CLET) for unilateral chronic ocular burns // Br. Ophthalmol., 2018, v.102, p.1640-1645.
16. Bhalekar S., Basu S., Sangwan V.S. Successful management of immunological rejection following allogeneic simple limbal epithelial transplantation (SLET) for bilateral ocular burns // BMJ case reports, 2013, doi: 10.1136/bcr-2013-009051.
17. Arya S.K., Bhatti A., Raj A. et al. Simple Limbal Epithelial Transplantation in acid injury and severe dry eye // J. Clin. Diagn. Res., 2016, v.10(6), Nd06-7.
18. Iyer G., Srinivasan B., Agarwal S. et al. Outcome of allo simple limbal epithelial transplantation (alloSLET) in the early stage of ocular chemical injury // Br. J. Ophthalmol., 2017, v.101, p.828-833.
19. Arora R., Dokania P., Manudhane A. et al. Preliminary results from the comparison of simple limbal epithelial transplantation with conjunctival limbal autologous transplantation in severe unilateral chronic ocular burns // Indian J. Ophthalmol., 2017, v.65, p.35-40.
20. Gupta N., Farooqui J.H., Patel N. et al. Early results of penetrating keratoplasty in patients with unilateral chemical injury after simple limbal epithelial transplantation // Cornea, 2018, v.37, p.1249-1254.
21. Singh D., Vanathi M., Gupta C. et al. Outcomes of deep anterior lamellar keratoplasty following autologous simple limbal epithelial transplant in pediatric unilateral severe chemical injury // Indian J. Ophthalmol., 2017, v.65, p.217-222.

22. Mittal V., Jain R., Mittal R. Ocular Surface Epithelialization Pattern After Simple Limbal Epithelial Transplantation: An In Vivo Observational Study // Cornea, 2015, v.34, p.1227-1232.
23. Mittal V., Jain R., Mittal R., Vashist U et al. Successful management of severe unilateral chemical burns in children using simple limbal epithelial transplantation (SLET) // Br. J. Ophthalmol., 2016, v.100, p.1102-1108.
24. Queiroz A.G., Barbosa M.M., Santos M.S. et al. Assessment of surgical outcomes of limbal transplantation using simple limbal epithelial transplantation technique in patients with total unilateral limbal deficiency // Arq. Bras. Oftalmol., 2016, v.79, p.116-118.
25. Kheirkhah A., Raju V.K., Tseng S.C. Minimal conjunctival limbal autograft for total limbal stem cell deficiency // Cornea, 2008, v.27, p.730–733.
26. Sangwan V.S., Basu S., Vemuganti G.K. et al. Clinical outcomes of xeno-free autologous cultivated limbal epithelial transplantation: a 10-year study // Br. J. Ophthalmol., 2011,v.95(11), p.1525-1529.
27. Dua H.S., Forrester J.V. Clinical patterns of corneal epithelial wound healing // Am. J. Ophthalmol., 1987, v.104(5), p.481-489.
28. Dua H.S., Azuara-Blanco A. Autologous limbal transplantation in patients with unilateral corneal stem cell deficiency // Br. J. Ophthalmol., 2000, v.84(3), p.273-278.

Di Zazzo A.

# SADƏ EPİTELİAL LİMBAL TRANSPLANTASİYA (SELT): GÖSTƏRİŞLƏR, CƏRRAHİ TEXNİKA, MEXANİZM, KLİNİK NƏTİCƏLƏR VƏ TƏSİRİ (ƏDƏBİYYAT İCMALI)

*Refraksiya və Buynuz Qişa Cərrahiyyəsi xidmətləri, Oftalmologiyada Kompleks Əməliyyat Bölməsi, Kampus Bio Medico Universitetinin Klinikası, Roma, İtaliya*

**Açar sözlər:** *sadə epitelial limbal transplantasiya, kultivasiya edilən limbal epitelial transplantasiya, konyunktival limbal autotransplantat, limbal kök hüceyrələrinin çatışmazlığı, kimyəvi yanıq*

## XÜLASƏ

Sadə epitelial limbal transplantasiya (SELT) metodikasının klinik effektivliyi hal-hazrda adekvat uzunmüddətli müşahidə müddəti ilə aparılan böyük tədqiqatlarda əsaslı qaydada sübuta yetirilmişdir [6-8]. Həmçinin, SELT aparılmasından sonra buynuz qişanın üzərində limbal kök hüceyrələrinin (LKH) persistensiyası göstərilmişdir [6], və bu zaman buynuz qişanın epitel qatının sağalması mexanizmi ətraflı tədqiq edilmişdir [22]. SELT, KLAT (konyunktival limbal autotransplantat) yaxud kultivasiyalı limbal epitelial transplantasiya (KLET) metodikalarının bilavasitə nəticələrinin müqayisəsi üzrə heç bir randomizasiya olunmuş, nəzarət edilən tədqiqatın indiyə kimi dərc edilməməsinə baxmayaraq, SELT üsulunun digərlərdən üstünlükləri bəllidir. Məhdud vasitələr şəraitində KLET üsulunun seçimi daha çox nəzəri hesab olunur, və buynuz qişa üzərində çalışan cərrahın seçimi əsasən KLAT və SELT arasında aparılır. SELT zamanı limbal toxumanın parçasının alınması saat əqrəblərinin bir radələrində mümkün olduğu halda, KLAT zamanı limbin üç – altı saat zonasında mümkünlüyünü nəzərə alaraq, seçim asandır. Buna baxmayaraq, anlamaq lazımdır ki, həm limbal, həm də konyunktival hüceyrələrinin çatışmazlığı ilə müşayiət olunan ağır simblefaron hallarında təkcə SELT üsulu effektiv deyil, nəticə etibarilə həm limbal kök hüceyrələrinin transplantasiyası (LKHT) metodikasına, həm də konyunktivanın transplantasiyasına ehtiyac yaranır. Məhz bu hallarda SELT və KLAT (mini-KLAT kimi modifikasiya olunmuş) üsulları kombinasiya oluna bilərlər [25]. KLAT üsulunun, SELT metodikası ilə müqayisədə, üstünlüyü ola biləcək digər istisna hallarına konyunktivanın, limbin və buynuz qişanın köçürülməsini tələb edən kompleks rekonstruksiya hadisələri aiddir. Bu zaman buynuz qişanın transplantatı immunoloji ayırma yüksək riskinə məruz qalır, və ehtimal olunur ki, gələcəkdə onun dəyişdirilməsinə zərurət yarana bilər, bu səbəbdən, KLAT metodikasında olduğu

kimi, limbal transplantatın onun anatomik yerində, yəni buynuz qişanın arxasında yerləşdirilməsi, SELT ilə müqayisədə, daha əlverişli ola bilər, çünki SLET zamanı buynuz qişanın transplantatının dəyişdirilməsi hallarında limbal transplantatlar itirilmiş olacaq.

SELT metodikasının meydana çıxması ilə buynuz qişa üzərində çalışan cərrahların, xüsusilə inkişaf edən ölkələrdə kimyəvi yanıqlar səbəbindən birtərəfli limbal kök hüceyrələrinin çatışmazlığı (LKHÇ) yükünü daşıyan cərrahların fəaliyyəti xeyli asanlaşmışdır [2]. Əməliyyatın icra edilməsi texnikasına yiyələnmə prosesi nisbətən qısa zaman çərçivəsində mümkündür və gözün ön hissəsində əməliyyatlar zamanı təcrübəsiz olan cərrahlar tez bir zamanda təcrübəli cərrahlarla eyni nəticələri əldə edə bilərlər [6]. Metodikaya yiyələnmə prosesinin asan olması SELT üsulunun okuloplastik cərrahlar kimi digər ixtisaslara da yayılmasına imkan yaratmışdır, bu zaman cərrahlar həmin metodikanı həm müalicə ünün, həm də gözün ön hissəsinin skvamoz neoplaziyasının (GÖHSN) götürülməsindən sonra LKHÇ qarşısının alınması üçün uğurla adaptasiya etmişdirlər, əks halda LKHT məruz qalardı [10,11]. SELT üsulunun bu cür etibarlılığı və yiyələnmə qabiliyyəti həmçinin müxtəlif coqrafi bölgələrdən olan cərrahlar tərəfindən aparılan böyük tədqiqatların sabit nəticələrində də göstərilmişdir [6-8].

SELT üsulunu in vivo KLET üsulu kimi qiymətləndirmək olar, və bu zaman hüceyrələrin bölünməsi toxuma becərilməsi üçün reagent kimi istifadə edilən təbii mühitin, böyümə amillərinin və göz yaşının iştirakı ilə Petri kasası əvəzinə gözün səthində baş verir. SELT üsulu KLET metodikasının əsas doqmasını təsdiq edir ki, limb toxumasının saat əqrəbinin bir istiqamətindəki parçası buynuz qişasının səthinin bütövlükdə bərpa olunması üçün kifayət edir. Təəccüblü deyil ki, SELT üsulunu işləyib hazırlayan cərrahlar həmin fikirə gəlmək üçün KLET metodikasının icra edilməsinə təxminən on il sərf etmişdirlər [26]. Buna baxmayaraq, SELT üsuluna meydan oxuyan iki paradiqmalar mövcuddur – limbal çuxura buynuz qişanın üzərinə yerləşdirilən LKH aqibəti və buynuz qişanın epitel qatının zədələnmələrinin sağalma modeli. SELT üsulunda ayrı-ayrı hissələrin konyunktival komponenti olmadığından epitelial hüceyrələr hər tərəfdən bölünür, bu isə buynuz qişanın sürətli epitelizasiyası ilə nəticələnir, buynuz qişanın sıyrıntıları zamanı fizioloji olaraq və KLAT üsulundan sonra müşahidə olunan periferik, sonra isə mərkəzəqaçan şəkildə sağalma ilə müqayisədə [27,28]. SELT həmçinin bizə sübut etmişdir ki, limbdə bu cür unikal kök hüceyrələrinin kəşf edilməsindən sonra otuz ilə yaxın keçməsinə baxmayaraq, ümumilikdə gözün ön hissəsinin fiziologiyası və xüsusilə LKH haqqında məlumatımız olduqca azdır.

Ди Заццо А.

# ПРОСТАЯ ЭПИТЕЛИАЛЬНАЯ ЛИМБАЛЬНАЯ ТРАНСПЛАНТАЦИЯ (ПЭЛТ): ПОКАЗАНИЯ, ХИРУРГИЧЕСКАЯ ТЕХНИКА, МЕХАНИЗМ, КЛИНИЧЕСКИЕ РЕЗУЛЬТАТЫ И ВОЗДЕЙСТВИ (ЛИТЕРАТУРНЫЙ ОБЗОР)

*Службы Рефракционной Хирургии и Хирургии Роговицы, Комплексное Оперативное отделение в Офтальмологии, Университетская клиника Кампус Био Медико, Рим, Италия*

**Ключевые слова:** *простая эпителиальная лимбальная трансплантация, культивируемая лимбальная эпителиальная трансплантация, конъюнктивальный лимбальный аутотрансплантат, дефицит лимбальных стволовых клеток, химический ожог*

## РЕЗЮМЕ

Клиническая эффективность ПЭЛТ в настоящее время основательно подтверждена крупными исследованиями с адекватным длительным периодом наблюдения [6-8]. Было продемонстрировано персистирование лимбальных стволовых клеток (ЛСК) на поверхности роговицы после ПЭЛТ [6]. При этом хорошо был изучен механизм заживления эпителия роговицы [22], несмотря на то, что до сих пор не было опубликовано ни одного рандомизированного контролируемого исследования, сравнивающего непосредственные результаты ПЭЛТ, конъюнктивального лимбального аутотрансплантата (КЛАТ) или культивируемой лимбальной эпителиальной трансплантации (КЛЭТ), хотя преимущества ПЭЛТ перед другими методами очевидны. В условиях ограниченных ресурсов выбор КЛЭТ является в значительной степени теоретическим, а выбор для хирурга, работающего с роговицей, в основном лежит между КЛАТ и ПЭЛТ. Так как

при ПЭЛТ можно получить лимбальную ткань в зоне одного часа, что при КЛАТ возможно в зоне лимба от трех до шести часов, то выбор хирурга прост. Тем не менее, важно понимать, что один только ПЭЛТ не эффективен в случаях с тяжелым симблефароном, при котором наблюдается дефицит как лимбальных, так и конъюнктивальных клеток, следовательно, требуется как LSCT трансплантация лимбальных стволовых клеток (ТЛСК), так и трансплантация конъюнктивы. Именно в этих случаях ПЭЛТ и КЛАТ (модифицированный как мини-КЛАТ) могут комбинироваться [25]. Другой исключительный сценарий, в котором КЛАТ может иметь преимущество перед ПЭЛТ – это случаи комплексной реконструкции, требующие пересадки конъюнктивы, лимба и роговицы. В этих случаях роговичный трансплантат остается подверженным высокому риску иммунологического отторжения и, возможно, в будущем потребуется его замена, поэтому размещение лимбального трансплантата в его анатомическом месте за роговицей, как в КЛАТ, может быть более выгодным по сравнению со ПЭЛТТ, где лимбальные трансплантаты будут утеряны, если роговичный трансплантат будет заменен.

Появление ПЭЛТ облегчило жизнь хирургам, работающим с роговицей, особенно тем, кто в развивающихся странах испытывает огромное бремя одностороннего дефицит лимбальных стволовых клеток (ДЛСК) из-за химических ожогов [2]. Обучение технике проведения операции возможно за относительно короткий период, и хирурги, не имеющие опыта в хирургии переднего отрещзка глаза, могут быстро получить те же результаты, что и опытные [6]. Эта легкая обучаемость позволила также методике ПЭЛТ распространиться на другие специальности, такие как окулопластические хирурги, где она была успешно адаптирована хирургами как для лечения, так и для предотвращения ДЛСК после обширного удаления сквамозной неоплазии переднего отрезка глаза (СНПОГ), которая в противном случае подлежала бы ТЛСК [10,11]. Такая надежность и обучаемость ПЭЛТ также продемонстрирована в стабильных результатах, о которых сообщают в больших исследованиях разные хирурги в различных географических регионах [6-8].

Можно представить ПЭЛТ как in vivo КЛЭТ, где размножение клеток происходит на поверхности глаза вместо чашки Петри с использованием естественной среды, факторов роста и слез в качестве реагентов для тканевой культуры. ПЭЛТ подтверждает центральную догму КЭЛТ о том, что отростка лимбальной ткани в зоне одного часа достаточно для восстановления поверхности всей роговицы. Неудивительно, что те, кто разработал ПЭЛТ, потратили около десяти лет, выполняя КЛЭТ сами [26], прежде чем они столкнулись с этой идеей. Тем не менее, две парадигмы, которые бросают вызов ПЭЛТ: необходимость помещения ЛСК вместо лимбальной ниши на роговицу и модель заживления раны эпителия роговицы. Поскольку отдельные части в ПЭЛТ не имеют конъюнктивального компонента, эпителиальные клетки растут со всех сторон, что приводит к быстрой эпителизации роговицы, в отличие от периферического, с последующим центростремительным заживлением, что наблюдается физиологически при ссадинах роговицы и после КЛАТ [27,28]. ПЭЛТ также научил нас тому, как мало мы еще понимаем в физиологии переднего отрезка глаза в целом и ЛСК, в частности, спустя почти три десятилетия после открытия этих уникальных стволовых клеток в лимбе.

## Corresponding author:

*Antonio Di Zazzo, MD*
*Cornea and Refractive Surgery Services, Complex Operative Unit in Ophthalmology, University Hospital Campus Bio Medico of Rome*
*E-mail: a.dizazzo@unicampus.it;*
*Telephone: +06225418893*
*Fax: +0622541456*