Abstracts

Purpose Human corneas preserved in bioreactor (BR) are characterized by not only a better endothelial viability, but also a more differentiated and stratified epithelium compared to corneas preserved in organoculture. By using corneal preservation in BR, we aimed to analyze the respective contribution of corneal (C), limbal (L), and conjunctival (Conj) epithelia in corneal epithelial regeneration.

Methods Five pairs of corneas from body donation to Science were used with a death-to-collection time <20 hours. A 3- to 5-mm-wide conjunctival flap was kept intact. Five patterns were set up by complete mechanical removal of 1, 2, or 3 epithelia (-): C-L+Conj+, C-L-Conj+, C-L+Conj-, C+L- Conj- (control) (n=2 for each pattern). The L epithelia was destroyed by scraping and thermocoagulation. Corneas were then kept in BR (21mmHg, 2.5μl/min of Corneamax Eurobio, 31°C) for 3 weeks to allow epithelial regeneration. The epithelium was then analyzed using immunofluorescence (IF) on flat mounted cornea by targeting CK12 (corneal epithelium) and CK15 (limbal epithelium). Cell nuclei were counterstained with DAPI. Corneal transparency was quantified using a transpameter.

Results No epithelium was reconstituted in the C-L-Conj- control group. In the other 4 models including the C-L-Conj+ group, the cornea was transparent and covered by a pluristratified corneal epithelium, characterized by CK12 expression.

Conclusion In this BR model, conjunctival epithelial cells alone allowed the regeneration of a typical corneal epithelium whereas corneal epithelium was able to migrate to the limbus and conjunctiva. We hypothesize that all 3 ocular surface epithelia contain stem cells or progenitors able to migrate throughout the cornea and restore the corneal epithelium independently of each other. The main difference between our ex vivo model and in vivo situation is the absence of neovascularization. This suggests that the main cause of limbic insufficiency is due to the loss of the anti-angiogenic barrier rather than the loss of limbic stem cells.

Today, split cornea technique is an established procedure and is mostly used for two recipients by combining deep anterior lamellar keratoplasty (DALK) and Descemet membrane endothelial keratoplasty (DMEK) surgeries. However, for some surgical interventions including block excision with tectonic corneoscleral grafting, split cornea procedure is not planned regularly up to now. In the run-up for this procedure, normally a donor cornea with a bigger scleral ring is gained. Nonetheless, the preparation of the tectonic graft for covering the corneoscleral defect after block excision results in a rest donor cornea transplant which is normally too small for further regular size penetrating keratoplasties (PKs) or combined DALK/DMEK surgeries. However, using a modified donor transplant trephination technique, a corneoscleral transplant for regular size keratoplasties can be gained, also after preparation of a tectonic graft for block excision. Herein, we describe shortly this novel donor preparation technique, the differences compared to the standard procedure, possible applications, and the advantages and disadvantages for the first time.