Corneal Wound Healing Using the EpiCorneal Tissue Model

Objectives

To investigate corneal wound healing using the EpiCorneal in vitro human 3D tissue model.

Methods

Tissues were equilibrated for 1 hour and abrasion wounds were introduced by: (a) gently scraping the tissue surface with a P100 pipette tip (scratch wound), or (b) gently applying a 2mm biopsy punch and carefully removing the cut-out tissue fragment with an aid of gentle vacuum application (biopsy punch wound). Chemical wounds were introduced by applying 0.5 µl of 1M NaOH for 15 min to the tissue surface. Wounded tissues were incubated in the Maintenance medium (COR-100-MM, MatTek) with or without Erlotinib (10 µM) for up to 5 days.

Figure 1: Histological cross-sections of H&E stained cross-section of EpiCorneal tissue model (A). The EpiCorneal tissue structure closely parallels that of the human corneal epithelium.

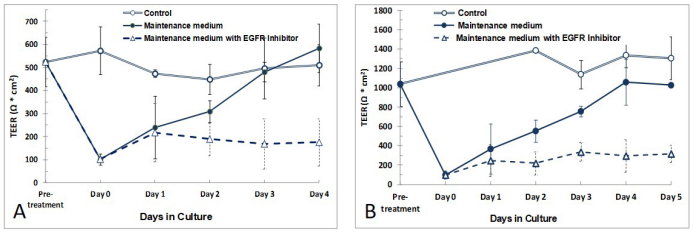

Figure 2: Recovery of the barrier properties of EpiCorneal tissues as assessed by transepithelial electrical resistance (TEER) after a: (A) scratch wound and (B) biopsy punch wound. Tissues were incubated in the maintenance medium (COR-100-MM) alone or in the presence of EGFR Inhibitor (Erlotinib, 10 µM).

Results

Incubation of the wounded tissues in the growth factor-containing maintenance medium (COR-100-MM) resulted in complete tissue recovery in 3-4 days. The presence of 10 µM Erlotinib, EGFR inhibitor, inhibited wound healing of the EpiCorneal tissues. No or little expression of Cyclin D1 is observed right after tissue abrasion; upregulation of Cyclin D1 was observed 24 hr post-abrasion.

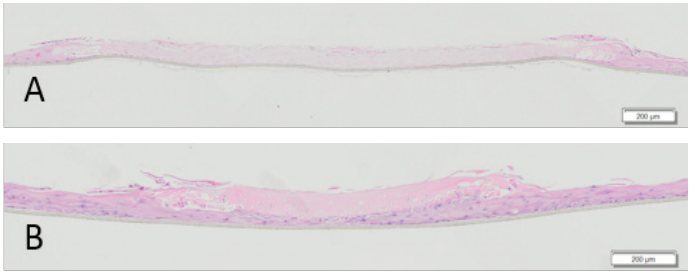

Figure 3: Cross-sections of EpiCorneal tissues in maintenance medium: (A) 30 min after and (B) 96 h after application of 0.5 µl of1N NaOH; H&E staining.

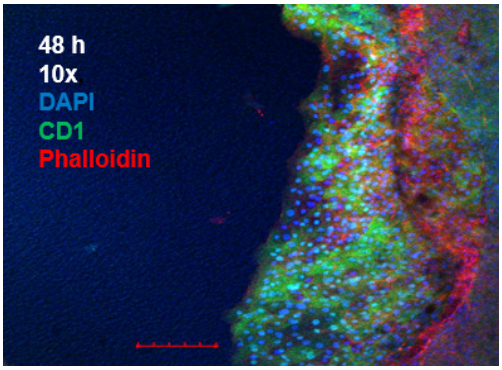

Figure 4: Immunofluorescent analysis. Cyclin D1 (CD1, green) expression in the leading edge of corneal epithelial cells during wound healing.

Conclusion

The EpiCorneal tissue model structurally and functionally reproduces corneal wound healing and will provide a comprehensive means to study and develop ophthalmic pharmaceuticals.