GROWTH FACTOR SECRETION IN HUMAN KERATOZYTE CULTURES FOLLOWING PHOTODYNAMIC INACTIVATION (PDI)

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Purpose: Photodynamic inactivation (PDI) may be an alternative treatment option of infectious keratitis, with increasing resistance of microorganisms to antibiotics. In previous studies we determined viability, apoptosis, proliferation, CD34 and alpha-smooth actin expression of keratocytes following PDI. The purpose of our present study was to determine the secretion of different growth factors in human keratocytes following PDI, in vitro.

Methods: Primary human keratocytes were isolated by digestion in collagenase A (1 mg/ml) from human corneal buttons, and cultured in DMEM/Ham’s F12 medium supplemented with 10% FCS. Five and twenty-four hours after PDI (100 nM chlorine e6, illumination 13 minutes at 670 nm), the release of growth factors was determined using enzyme-linked immunosorbent assay (ELISA).

Results: Five hours following PDI, KGF (0.42 to 0.24 pg/µg Protein, p=0.02) and HGF (0.43 to 0.25 pg/µg Protein, p=0.01) secretion decreased, FGFb expression (3.47 to 5.27 pg/µg Protein, p= 0.0001) increased significantly compared to controls. At this time point TGFβ1 was not detectable and VEGF secretion was not significantly different from control cultures. Twenty-four hours after illumination expression of none of the growth factors had changed significantly compared to controls. Treatment only with illumination or Ce6 did not show changes in the expression of growth factors at any time point.

Conclusions: Five hours after PDI, KGF and HGF expression decreases and FGFb expression increases in keratocyte cell cultures. However, 24 hours after treatment growth factor secretion seems to be normalized. The altered secretion of KGF, HGF and FGFb may play a role in activation of keratocytes and wound healing response after PDI in the cornea.