# IMMUNOMODULATION OF CORNEAL EPITHELIAL CELLS FOLLOWING ELECTROPORATION WITH MRNA ENCODING IL-10 AND FASL

*ZAKARIA N., TASSIGNON M.J., COOLS N., VAN TENDELOO V., BERNAMAN Z.*

**Affiliation Institution:** University Hospital Antwerp
**Promotors of the Project:** TASSIGNON M.J., M.D., PhD, Dr. COOLS N., M.D., VAN TENDELOO V., M.D., PhD., BERNAMAN Z., M.D., PhD

## AIM OF THE PROJECT

The aim of this project is to transfect corneal epithelial cells with mRNA encoding IL-10 and FasL in order to evoke down modulation of allogenic T cell response in an *ex vivo* model.

## DEVELOPMENT OF THE PROJECT

Limbal stem cell deficiency (LSCD) can be caused by a variety of different conditions including Steven Johnson's Syndrome, Ocular Cicatricial Pemphigoid, anirida, chemical and thermal burns as well as severe bacterial keratitis. LSCD results in conjunctivalization of the cornea with vascular encroachment which can lead to a decrease in visual acuity and result in considerable pain and discomfort for the patient. As a new treatment strategy, we started a clinical trial for cultivated limbal stem cell transplantation for the treatment of LSCD, the results of which have varied. Autologous transplants fared well, although the allogenic cultivated limbal transplants showed signs of graft rejection despite strong systemic immunosuppression. This could be explained by the fact that the corneas in these cases are vascularized and hence lack the immune privilege of healthy corneas (1).

This leads us to investigate the possibility of immunomodulation of the limbal epithelial cells in culture prior to transplantation, by electroporation. This is a technique whereby we can induce a significant increase in the electrical conductivity and permeability of the cell plasma membrane by an externally applied electrical field. As a consequence, this allows the introduction of mRNA or a piece of coding DNA into the cell, that can potentially alter its immunogenicity.
Interleukin 10 (IL-10) is capable of inhibiting synthesis of pro-inflammatory cytokines like IFN-γ, IL-2, IL-3, TNFγ and GM-CSF made by cells such as macrophages and Type 1 T helper cells (2). IL-10 also displays potent abilities to suppress the antigen presenting capacity of antigen presenting cells which in this case would be the Corneal Epithelial Cells (CECs). Cells in immune privileged areas such as the cornea or testes express Fas ligand and induce the apoptosis of infiltrating lymphocytes (3). It is one of many mechanisms the body employs in the establishment and maintenance of immune privilege. Therefore we postulate that by introducing mRNA encoding IL-10 and FasL into the CECs, we will be able to cause an up regulation of expression of these molecules, resulting in a reduced allogenic T cell response.

## REFERENCES


(1) CURSIEFEN C., NIEDERKORN J.Y., KAPLAN H.J. – Immune Privilege and Angiogenic Privilege of the Cornea. Immune Response and the Eye. Chem Immunol Allergy. Basel, Karger, 2007, vol 92: 50-57

(2) CASSATELLA M., MEDA S., BONORA S., CESKA M., CONSTANTIN G. – Interleukin 10 (II.-10) Inhibits the Release ofProinflammatory Cytokines from Human Polymorphonuclear Leukocytes. Evidence for an Autocrine Role of Tumor Necrosis Factor and II,-10 in Mediating the Production of II.-8 Triggered by Lipopolysaccharide, 1993, 17:2207-11.

(3) GRIFFITH S., BRUNNER T., FLETCHER S.M., GREEND D.R., FERUGUSON T.A. – Fas Ligand-Induced Apoptosis as a Mechanism of Immune Privilege; Science 1995; 270 (5239): 1189. November 1995) Science 270 (5239), 1189.