T lymphocytes of the normal human cornea

O F SCHEIFFARTH, F H STEFANI, N GABRIEL, AND O-E LUND

From the University Eye Hospital, Munich, Federal Republic of Germany

SUMMARY Lymphocytes in the periphery of the normal human cornea are identified as being only T lymphocytes by immunohistochemical methods. OKT-4 positive cells (T-helper/inducer lymphocytes) and OKT-8 positive cells (T-suppressor/cytotoxic lymphocytes) are found in similar numbers in most of the corneas examined. OKT-4 positive cells in the cornea present a risk of transferring HTLV-III (HIV) by corneal grafting.

Lymphocytes are present in the epithelium and the stroma of the normal human cornea even in the absence of any pathological process.¹ They occur sporadically in the periphery and in low numbers in more central parts of the cornea.

Lymphocytes can be specified into their subtypes by means of monoclonal antibodies against different epitopes of glycoprotein nature on the cells. Meanwhile an increasing number of different clones and antibodies is produced reacting with the same subtype of leucocytes. Recently the Committee on Human Leucocyte Differentiation Antigens proposed a new nomenclature system using the so-called cluster designation (CD) to characterise the cells and their antigens.² For instance, T-helper/inducer cells are characterised by the cluster CD4, corresponding to a variety of antibodies like T4, Leu3a, or 91D6. The cytotoxic/suppressor subset of T lymphocytes in this nomenclature is characterised by the cluster designation CD8 corresponding to a number of monoclonal antibodies like Leu2a, TB, M236, etc.³ Some of these antibodies are produced commercially, such as OKT-4 (T-helper/inducer cells), OKT-8 (suppressor/cytotoxic T cells), and OKT-3, OKT-11 (all T cells, T cells forming E-rosettes.)⁴ Even B lymphocytes can be identified by an antibody like OKB-7.⁵

These antibodies became an important tool for analysing immune mechanisms under normal and pathological conditions. One of the most recent instances is the acquired immune deficiency syndrome (AIDS), a disease caused by the human T-lymphotropic virus type III (HTLV-III, HIV), which injures the T-helper/inducer cells and finally leads to opportunistic infections and neoplasms. Meanwhile it seems to be proved that the CD4 antigen is an essential and specific component of the receptor for the HTLV-III.⁶⁻¹⁰

Material and methods

We examined 20 normal corneas which had been obtained as fresh donor material for keratoplasty. A specimen of the corneal periphery was excised and cryostat sections cut at −20°C. The sections were allowed to dry at room temperature and were fixed in acetone for 10 minutes.

Histochemical staining of lymphocytes was performed by immunoperoxidase technique: (1) pretreatment of tissue with normal serum; (2) incubation with the primary mouse monoclonal antibody; (3) incubation with antimouse immunoglobulin (linking reagent); (4) incubation with peroxidase labelled mouse IgG; (5) addition of the substrate reagent (amino-ethylcarbazole and hydrogenperoxide). After each step the specimens were washed several times with phosphate buffered saline.

As primary antibody we used the OKB-7 antibody as reagent for B lymphocytes and OKT-11 for T cells in general. T-helper/inducer cells were identified by the OKT-4 antibody and the T-suppressor/cytotoxic subset by the OKT-8 antibody.

Results

Specimens of the periphery of 20 different corneas were examined, and lymphocytes were identified by immunohistochemical staining in 16 samples. Using the OKB-7 antibody we could detect no B lymphocytes in the cornea. Positive controls for the binding of B lymphocytes to the OKB-7 antibody were performed with fresh tissue obtained from tonsil-
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Lymphocytes are present as T cells in the normal human cornea even in absence of any inflammatory process. While they are quite common in the periphery of the cornea, they also occur sporadically in the central part of the cornea. In most corneas T4-positive and T8-positive cells occur in similar numbers. An exact quantitative determination needs large numbers of sections from different parts of the cornea, which were not obtained in this study.

These T lymphocytes probably migrate into the cornea from the limbal vessels. Their exact function in the defence mechanism of the cornea is unknown.

The finding of T4-positive cells in the normal human cornea is of great interest in connection with the possible transfer of the acquired immune deficiency syndrome (AIDS) by a keratoplasty. Since the CD4 (T4) antigen is an essential component of the receptor for the AIDS retrovirus, the question arises whether lymphocytes occurring in normal corneas could possibly transmit HTLV-III in corneal grafting, though no transmission of HTLV-III by corneal grafting has yet been reported. HTLV-III has been demonstrated in tears of patients suffering from AIDS. This has led to proposals from the Centers for Disease Control. No advice for corneal grafting was recommended at that time, possibly because no documented case of AIDS transmission by corneal grafting has yet occurred.

The Eye Bank Association of America uses two screening techniques to detect infected donor corneas: (1) identification of high-risk individuals, and (2) seropositivity detected by the enzyme linked immunosorbent assay (ELISA) test. Pepose et al. have discussed the safety of these screening methods. Meanwhile HTLV-III has also been demonstrated in corneal epithelial cells of a donor with a positive serum antibody to HTLV-III. The virus was not demonstrated in keratocytes or endothelium of the cornea. Along with these findings corneal grafting bears a so far unknown potential risk of transplanting the infection. We also wish to stress O’Day’s point: ‘the risk-benefit ratio needs to be considered carefully’.

**References**


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O F Scheiffarth, F H Stefani, N Gabriel and O E Lund

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