ABSTRACT

Purpose: Lectins play an important role in many biological processes. The aim of this work was to analyse mainly the expression of endogenic lectins, such as galectins and plant lectin, e.g. *Dolichos biflorus* agglutinin (DBA), and their glycoligands in the tear fluid, human corneal and conjunctival epithelium in physiological and disease conditions. Further, we studied the human natural antibody against Galα1,3Gal–R, which is mainly responsible for hyperacute rejection of xenografts transplants. We tried to investigate its localization in human corneal epithelium, lacrimal gland and tears.

Material and Methods: Human tissue (lacrimal gland, tear fluid, conjunctiva, cornea, epidermis, keratinocyte and cultured corneal epithelium), as well as porcine tissue (cornea, liver and epidermis) were examined. Endogenous galectins (galectins–1, -3 and -7) were detected using immunohistochemistry methods. Binding sites for galectins, as well as binding sites for plant lectin *Dolichos biflorus* agglutinin, were localized by lectin histochemistry. Reverse lectin histochemistry was used for the study of binding reactivity of endogenous lectins using labelled (neo)glycoligands. Employing biotinylated natural human IgG anti α–galactosides, as well as anti β–galactosides, we detected reactive epitopes in human cornea, lacrimal gland, tear fluid, skin, muscle capillaries and in porcine cornea, skin, vein and liver. The expression of galectin–1 and -3, laktoferrin and α , β galactosides in tear fluid was confirmed by using western blot.

Results: Galectin–1 was markedly present in tear fluid, corneal and limbal epithelium, and was absent in conjunctival epithelium. Galectin–3 was found in tears from patients with ocular surface disorders, in normal conjunctival and corneal epithelium, but not in the lacrimal gland. Inflammatory leucocytes and goblet cells found in galectin–3-containing tear fluid also expressed galectin–3. Galectin–3-binding sites were detected on the surface of conjunctival and corneal epithelium colocalizing with desmoglein. All cell layers of the corneal epithelium were positive for galectin–7. The binding of *Dolichos biflorus* agglutinin was typical for postmitotic early differentiated epithelial cells. Concerning cellular reactivity, the porcine corneal epithelium was negative for $Gal\alpha 1,3Gal$ structures, which are known to be abundantly expressed on cells of non–primate grafts, consequently causing an immunological barrier between humans or other Old World primates and non–primate mammals.

Conclusions: The monitoring of the presence of galectin–3 and its binding sites prompts the elucidation of the functional role of galectin in the eye under both normal and pathological condition. The results show potential participation of galectin–3 in mediation of intercellular contacts of corneal epithelium, namely in suprabasal cells. The specific binding of *Dolichos biflorus* agglutinin for postmitotic early differentiated epithelial cells lends strong support for using glycohistochemical methods in the study of differentiation of cells of the squamous epithelium. The absence of $Gal\alpha 1,3Gal$ structures in the porcine corneal epithelium raise the question whether it might be possible to use porcine cornea and the epithelial cell layer in clinical medicine as viewed from the perspective of α Gal.