ELSEVIER

LABORATORY INVESTIGATIONS

Lectin Cytochemistry of the Rabbit Conjunctiva and Lacrimal Sac

Seiji Maeda, Makoto Ishikawa, Tohru Abe and Shozo Sakuragi

Department of Ophthalmology, Akita University School of Medicine, Akita, Japan

Abstract: Four different types of lectin were applied to the rabbit conjunctiva and the lacrimal sac to examine the distribution of glycoconjugates. The conjunctival epithelium is comprised of goblet cells and nongoblet cells. The mucus granules of the goblet cells were stained with soybean agglutinin (SBA), and the cell body of the nongoblet cells was labeled with concanavalin A (Con A). The glycocalyx of the apical surfaces of the goblet cells and of the nongoblet cells was labeled with *Maculura pomifera* agglutinin. The lacrimal sac mucosa is comprised of superficial light and dark cells, and basal cells. The cell bodies of the light cells were stained with SBA. The glycocalyx of the apical surfaces of the light and dark cells was characteristically labeled with Con A. These results suggest that the composition of glycoconjugates is markedly different between the conjunctiva and the lacrimal sac, especially in the cell surface glycocalyx. **Jpn J Ophthalmol 1998;42:443–449** © 1998 Japanese Ophthalmological Society

Key Words: Conjunctiva, glycocalyx, lacrimal sac, lectin-binding properties, rabbit, ultrastructural pre-embedding labeling.

Introduction

Tears enter the lacrimal sac through the lacrimal punctum and the canaliculus after moistening the epithelial surface of the cornea and the conjunctiva.[1] Although the conjunctiva is connected to the lacrimal sac via the canaliculus, there is a marked difference in the cell composition and function of these membranous structures. The conjunctival epithelium is characterized by goblet cells, which secrete mucins into the innermost layer of the laminar preocular tear film.[2,3] The stroma (substantia propria) of the conjunctiva is highly vascularized, with well-developed lymphoid tissues, which function as a defense system against infectious microbes.[4] Although the lacrimal sac mucosa has a number of goblet cells, its main function is the excretion of tears into the nasal cavity. The human lacrimal sac also contains foci of ciliated epithelium, which facilitate the transportation of tears.[1,5]

The term *glycocalyx* is often used to describe the carbohydrate-rich layer on the apical surfaces of cells. This layer can be visualized by a variety of stains, such as ruthenium red, as well as by labeled lectins.[6] It is known that cell surface glycoconjugates play an important role in cell identification, cell-to-cell recognition, and defense mechanisms against microbes.[7] Although there are a few histochemical studies of the conjunctival glycocalyx,[8–10] an analysis of the glycocalyx of the lacrimal sac has not been reported.

Lectins have been used as probes for the characterization of glycoconjugates in various tissues,[11,12] In the present study, four different types of biotinylated lectins were applied to the rabbit conjunctiva and the lacrimal sac to reveal differences in the composition of the glycoconjugates. Lectin-binding sites were observed by light and electron microscopy.

Material and Methods

Fifteen male New Zealand white rabbits weighing approximately 3 kg each were used this study.

Received: January 23, 1998

Address correspondence and reprint requests to: Shozo SAKURAGI, Department of Ophthalmology, Akita University School of Medicine, Hondo 1 - 1 - 1, Akita 010, Japan

Jpn J Ophthalmol 42, 443–449 (1998)

Published by Elsevier Science Inc.

0021-5155/98/$19.00
PII S0021-5155(98)00051-3

Jpn J Ophthalmol
Vol 42: 443–449, 1998

Dacryocystography

Dacryocystography was undertaken on several rabbits under ketamine hydrochloride (10 mg/kg) anesthesia. Contrast medium (lipiodol ultra fluid) was injected into the lacrimal drainage system through the lacrimal punctum.

Histological Procedures

Rabbits were sacrificed by an overdose of ketamine. The palpebral conjunctiva in the vicinity of the lacrimal punctum and the lacrimal sac were removed *en bloc* and dissected in chilled saline. Following immersion fixation in 3.5% formaldehyde solution for 1 hour, the conjunctiva and lacrimal sac were embedded in paraffin. All sections were cut at 10 μm and stained with hematoxylin-eosin for light microscopic observation. A portion of each specimen was fixed with 2.5% glutaraldehyde (in 0.1 mol/L sodium cacodylate) for 2 hours. After postfixation with 1.0% OsO_4 (in 0.1 mol/L sodium cacodylate) for 2 hours, they were dehydrated with an ethanol series and embedded in Epon for electron microscopic observation.

Lectin Cytochemistry

Reagents. Biotinylated soybean (SBA), *Ulex europaeus* I (UEA), concanavalin A (Con A) and *Maculura pomifera* (MPA) agglutinins (Vector Laboratories, Burlingame, CA, USA) were commercially obtained; haptenic sugars, N-acetyl-D-galactosamine, L-fucose, α-methyl-D-mannoside, and D-galactose were used against SBA, UEA, Con A, and MPA, respectively.

Staining with biotinylated lectins. Following immersion fixation in 4% paraformaldehyde (in 0.1 mol/L sodium cacodylate) for 30 minutes, a part of the specimen was embedded in OCT compound to be sectioned at 10 μm on a cryostat at −20°C. The cryosections were incubated with 25 μg/mL biotinylated lectins, and then with ABC complex consisting of avidin and biotinylated horseradish peroxidase (Vector). After rinsing several times, the sections were developed with diaminobenzidine solution (Sigma Chemical, St. Louis, MO, USA). The specificity of lectin binding was verified by competitive inhibition of haptenic sugars at 200 mmol/L.

Ultrastructural pre-embedding labeling. The specimens, fixed with 4% paraformaldehyde (in 0.1 mol/L sodium cacodylate), was incubated with 100 μg/mL biotinylated lectins, and then with the ABC complex. After development, specimens were post-fixed with 1.0% OsO_4 and embedded in Epon. Ultrathin sections were cut with a diamond knife on an MT-II ultramicrotome (Du Pont, Newtown, CT, USA), and examined by electron microscopy. Specificity of lectin binding was verified by competitive inhibition of haptenic sugars at 200 mmol/L.

Results

Topological Anatomy of Lacrimal Drainage System

The excretory portion of the rabbit lacrimal system consisted of the single lacrimal punctum, the canaliculus, the lacrimal sac, and the nasolacrimal duct. The punctum lay slightly anterior to the nictitating membrane and opened in a slit-like manner with a length of 1.0 mm. During dacryocystography, the contrast dye filled the lacrimal sac and nasolacrimal duct (Figure 1a). The lacrimal sac gradually decreased in diameter and connected to the nasolacrimal duct. The mucous membrane within the sac was sometimes arranged into membranous folds.

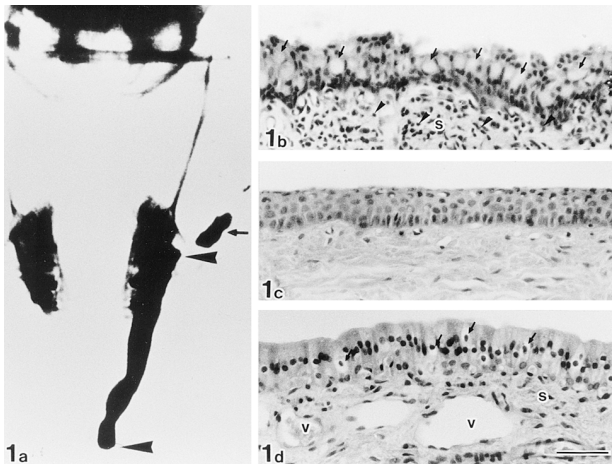

Figure 1. (**a**) Postero-anterior view of left dacryocystogram showing free flow of contrast medium into rabbit nose. Proximal end of lacrimal sac and distal end of nasolacrimal duct are indicated by arrowheads. Note retention of some contrast medium in conjunctival sac (arrow). (**b**) Light micrograph of hematoxylin-eosin (H-E) stained paraffin section of conjunctiva. Conjunctiva is covered by nonkeratinizing cuboidal epithelium intermixed with goblet cells (arrows). There is lymphoid cell infiltration (arrowheads) in substantial propria (S). (**c**) Light micrograph of H-E stained paraffin section of lacrimal canaliculus mucosa. Mucosa is covered by squamous stratified epithelium. (**d**) Light micrograph of H-E stained paraffin section of lacrimal sac mucosa. Lacrimal sac is lined with nonkeratinized columnar epithelium, and contains a number of light cells with clear cell bodies (arrows). S: substantia propria, V: vasculature × 150. Bar = 100 μm.

Structure of Lacrimal Drainage System

Conjunctiva. The epithelium of the conjunctiva was three to four cells thick. The goblet cells were distributed among the epithelial cells (Figure 1b). The external surface of the conjunctiva mainly comprised the apical surfaces of the goblet cells and the nongoblet epithelial cells. Both types of cells had short microvilli on the apical surfaces, and they were loosely interconnected by interdigitating desmosomes (Figure 2). There was a small number of light cells in the superficial layer of the epithelial cells. The basal cells were cuboidal and rested on a basement membrane.

The substantia propria comprised well-developed connective tissue, which was filled with a large number of mononuclear cells.

Lacrimal canaliculus. The epithelial lining of the canaliculus had a nonkeratinizing stratified appearance of squamous cells (Figure 1c). There seemed to be no goblet cells or mucus secreting cells in the epithelium.

Lacrimal sac. The lining of the epithelium had a two-layered columnar appearance (Figure 1d). In the superficial layer, the light epithelial cells with pale cytoplasm and a few mitochondria were intermixed with the dark epithelial cells, which were filled with a large number of mitochondria, and with well-developed rough endoplasmic reticula and the Golgi apparatus. Both types of epithelial cells had microvilli on the luminal surface of the lacrimal sac. There seemed to be no ciliated epithelium. The basal cells were cuboidal and rested on a basement membrane (Figure 3).

The substantia propria was composed of richly vascularized connective tissue.

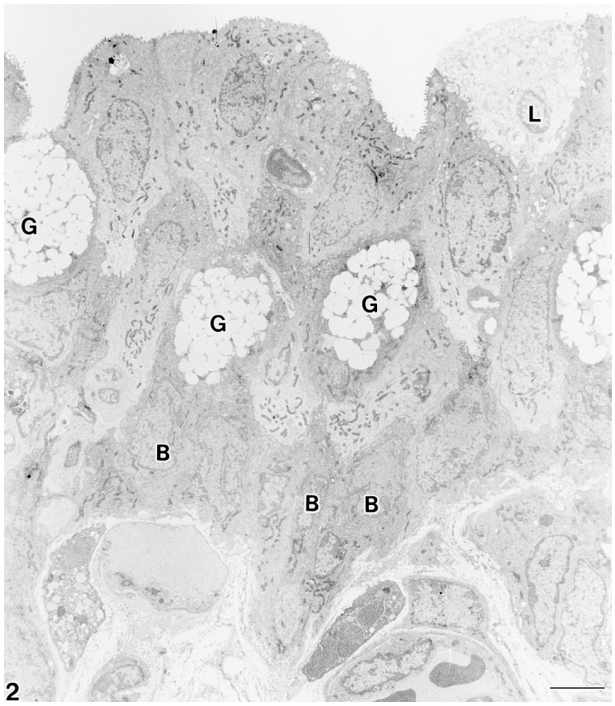

Figure 2. Electron micrograph of conjunctiva. Epithelial layer is three to four cells thick. Epithelial cells are polygonal and tightly packed with interdigitated desmosomes. Mucus-forming cells, goblet cells (G), are intermixed with epithelial cells. Apical surfaces of epithelial cells and goblet cells have short microvilli. L: Light-epithelial cell, B: Basal cell. × 3000. Bar = 5 μm.

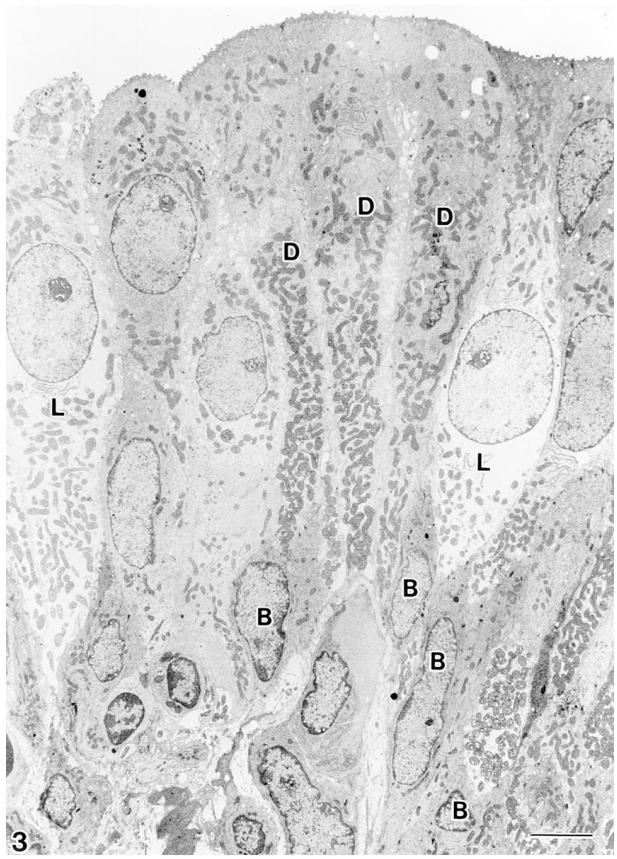

Figure 3. Electron micrograph of lacrimal sac mucosa. Epithelial layer shows two-layered arrangement. Superficial layer is comprised of dark (D) and light (L) epithelial cells. Former have many mitochondria and well-developed endoplasmic reticulum and Golgi apparatus. Latter have pale cytoplasm, and resemble holocrine-secreting cells. Apical surfaces of both types of cells have short microvilli. Basal cells (B) reside on basement membrane. × 3000. Bar = 5 μm.

Jpn J Ophthalmol
Vol 42: 443–449, 1998

Lectin Staining

Soybean agglutinin. Soybean agglutinin was strongly bound to the goblet cells and weakly to the nongoblet cells in the conjunctival epithelium. Soybean agglutinin was also bound to the vasculature in the substantia propria (Figure 4a), and weakly to the light cells in the lacrimal sac epithelium. There seemed to be no positive reactions in the substantia propria of the sac (Figure 4b).

By ultrastructural pre-embedding labeling, there seemed to be vestigial labeling on the apical surfaces of the conjunctiva and the lacrimal sac (data not shown).

Ulex europaeus. *Ulex europaeus* was bound to the external surface of the conjunctiva and to the luminal surface of the lacrimal sac mucosa (Figures 4c and 4d). Positive reactions were not found in other parts of the conjunctiva and lacrimal sac. The vasculature in the substantia propria of these tissues also did not show reactions.

Ultrastructural pre-embedding labeling revealed DAB products deposited on the apical surfaces of the goblet and nongoblet cells in the conjunctival epithelium (Figure 5a), and also on the corresponding surfaces of the light and dark epithelial cells in the lacrimal sac mucosa (Figures 5b and 6b).

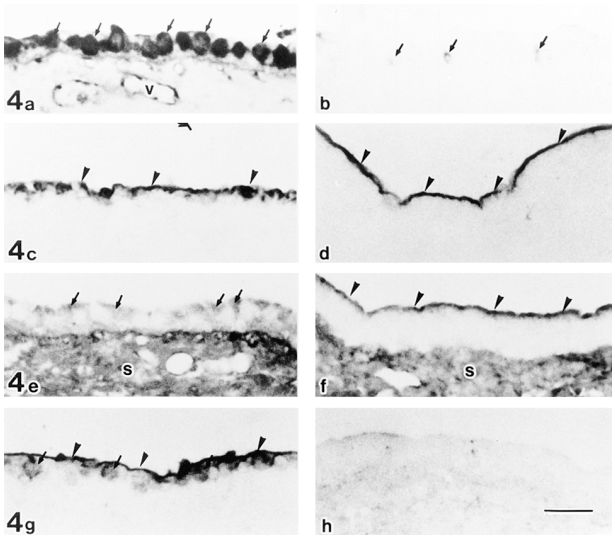

Figure 4. Light micrographs of soybean agglutinin (SBA)-stained frozen sections of conjunctiva (**a**) and lacrimal sac (**b**). Conjunctival goblet cells (arrows) and vasculature (V) in substantia propria are strongly stained with SBA. In contrast, light epithelial cells (arrows) in lacrimal sac are weakly stained with SBA. Light micrographs of *Ulex europaeus* (UEA)-stained frozen sections of conjunctiva (**c**) and lacrimal sac (**d**). External surface of conjunctival epithelium (arrowheads) and luminal surface of lacrimal sac (arrowheads) are positively stained with UEA. Light micrographs of Concanavalin A (Con A)-stained frozen sections of conjunctiva (**e**) and lacrimal sac (**f**). Cell body of nongoblet cells (arrows) and substantia propria (S) in conjunctiva are stained with Con A. By contrast, luminal surface (arrowheads) and substantia propria (S) of lacrimal sac are stained with Con A. Light micrographs of *Maculura pomifera* (MPA)-stained frozen sections of conjunctiva (**g**) and lacrimal sac (**h**). Conjunctival goblet cells (arrows) are weakly stained with MPA. External surface of conjunctiva is stained with MPA (arrowheads). By contrast, lacrimal sac is negatively stained with MPA. × 120. Bar = 100 μm.

Concanavalin A. Concanavalin A was bound to the nongoblet epithelial cells and the substantia propria of the conjunctiva. The goblet cells did not show positive reactions (Figure 4e). In the lacrimal sac, the luminal surface of the epithelium and the substantia propria were stained with Con A (Figure 4f).

By electron microscopy, the apical surfaces of the conjunctival cells showed only vestigial labeling (Figure 5c). In contrast, the apical surfaces of the light and dark lacrimal epithelial cells were positively stained with Con A. However, some groups of dark epithelial cells showed negative labeling. (Figure 5d).

Maculura pomifera. Maculura pomifera was bound to the external surface as well as to the goblet cells of the conjunctiva. There was no positive reaction in the substantia propria (Figure 4g). The lacrimal sac showed negative reactions over all regions. (Figure 4h).

By electron microscopy, DBA products could be seen deposited on the apical surfaces of the goblet and nongoblet cells in the conjunctiva. However, some groups of dark epithelial cells showed negative labeling (Figures 5e and 6a). The corresponding surfaces of light and dark cells in the lacrimal sac epithelium showed no positive reactions (Figure 5f).

In the specimens incubated with a mixture of biotinylated lectin and a specific haptenic sugar, reaction for lectin was significantly reduced over all regions (Figure 6c). Observations of lectin binding in the conjunctiva and the lacrimal sac are summarized for each epithelial cell category and their cell surfaces in Tables 1 and 2.

We did not find any differences in the lectin binding pattern between the proximal and distal parts of the lacrimal sac.

Discussion

The nasolacrimal duct and the lacrimal sac develop from a linear thickening of ectoderm in the floor of the nasolacrimal groove. The thickening

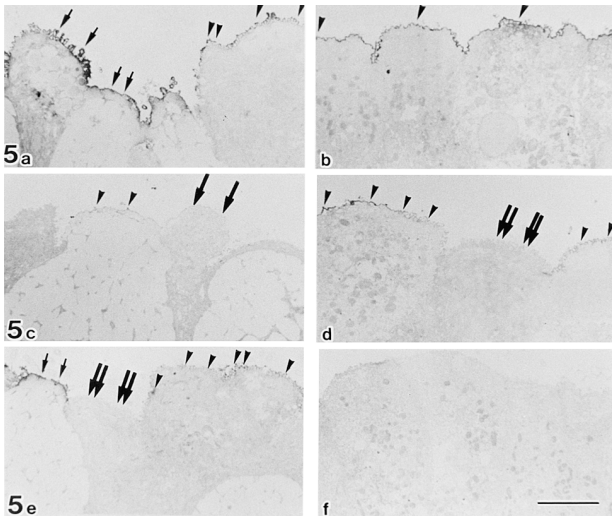

Figure 5. Electron micrographs of conjunctiva (**a**) and lacrimal sac (**b**) after labeling with *Ulex europaeus* (UEA). (**a**) Apical surfaces of nongoblet epithelial cells (arrowheads) and goblet cells (arrows) show positive reaction to UEA binding. (**b**) Corresponding surfaces of lacrimal epithelial cells (arrowheads) are also stained with UEA. Electron micrographs of conjunctiva (**c**) and lacrimal sac (**d**) after labeling with concanavalin A. (**c**) There appears to be vestigial labeling on apical surfaces of nongoblet (arrows) and goblet cells (arrowheads). (**d**) HRP-reactive products deposited on apical surface of lacrimal epithelial cell (arrowheads). The corresponding surfaces of some dark epithelial cells are without labeling (double arrows). Electron micrographs of conjunctiva (**e**) and lacrimal sac (**f**) after labeling with MPA. (**e**) HRP-reactive products are deposited on apical surfaces of goblet (arrows) and nongoblet cells (arrowheads). Corresponding surface of some epithelial cells is not labeled (double arrows). (**f**) There seems to be negative reaction on apical surfaces of epithelial cells. ×3000. Bar = 5 μm.

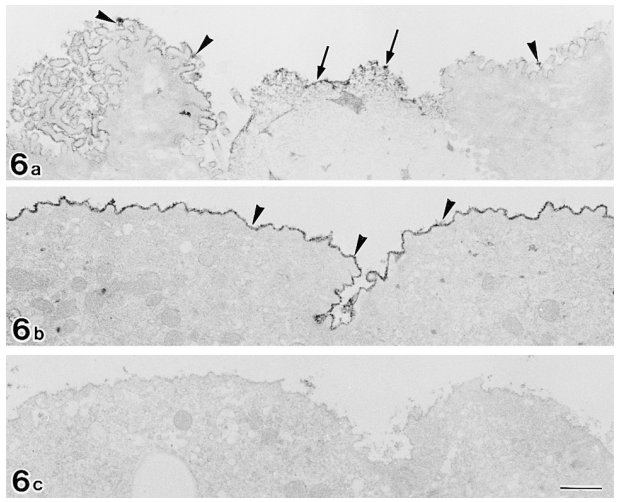

Figure 6. (**a**) High magnification of apical surface of conjunctiva after labeling with *Maculura pomifera* (MPA). HRP-reactive products are deposited on apical surface of goblet (arrows) and nongoblet cells (arrowheads). (**b**) High magnification of apical surface of lacrimal sac after labeling with *Ulex europeaus* (UEA). HRP-reactive products are deposited on apical surface of lacrimal epithelial cell (arrowheads). (**c**) Electron micrograph of apical surface of lacrimal sac after labeling with UEA and its haptenic sugar. There seems to be negative reaction on apical surface of epithelial cell. ×10000, Bar = 1 μm.

gives rise to an epithelial cord, and its cranial end expands to form the lacrimal sac. Then, the lacrimal sac merges with the conjunctiva, which is derived from surface ectoderm via the lacrimal canaliculus.[13] Although the conjunctiva and the lacrimal sac are of ectodermal origin and their epithelia are connected, the present study reveals that the composition of cell surface glycocalyx is markedly different from the conjunctival epithelia and the lacrimal sac mucosa in the rabbit.

In the present study, UEA, which has an affinity for fucose,[14] was specifically bound to the glycocalyx of the conjunctiva and the lacrimal sac. Therefore, there are common UEA-binding glycoconjugate residues in the conjunctiva and the lacrimal sac. However, the glycocalyx showed a different labeling pattern than the other lectins. Concanavalin A, which has an affinity for mannose,[12] was specifically bound to the glycocalyx of the lacrimal sac, but showed no binding to that of the conjunctiva. By contrast, MPA, which has an affinity for galactose,[11] specifically labeled the glycocalyx of the conjunctiva, but did not bind to that of the lacrimal sac. These results indicate that the glycocalyx of the conjunctiva is characterized by galactose, and that of the lacrimal sac by mannose. As cell surface carbohydrates are thought to play an important role in the defense mechanism against infectious microbes,[7] the difference in glycocalyx composition of the conjunctiva and the lacrimal sac may indicate that the absorption process of the microbes or the molecular mechanisms of inflammation may be different in these structures. There have been few reports concerning the changes in the constituent glycoconjugates of the conjunctiva and the sac during the process of inflammation. Our next goal is to reveal the changes in lectin-binding patterns during conjunctivitis and dacryocystitis.

It is of interest that there are Con A-negative and Con A-positive dark epithelial cells in the lacrimal sac mucosa, as well as MPA-positive and MPA-negative dark epithelial cells in the conjunctival epithelium.

Jpn J Ophthalmol
Vol 42: 443–449, 1998

Table 1. Lectin Binding to Rabbit Conjunctival Epithelium

	Goblet Cell Body	Nongoblet Cell Body	External Surface of Goblet Cell/ Nongoblet Cell	
SBA	+++	+	−	−
UEA	−	−	++++	+++
Con A	−	+	−	−
MPA	++	−	+++	+++,−

Con A: concanavalin A agglutinin; MPA: *Maculura pomifera* agglutinin. SBA: soybean agglutinin; UEA: *Ulex europaeus* agglutinin.

Intensity of lectin labeling was qualitatively evaluated by visual judgment as negative (−), weak (+), moderate (++), or intense reaction (+++). Reaction of cell body was examined by light microscopy, and that of external surface, by electron microscopy.

Table 2. Lectin-binding to rabbit lacrimal sac epithelium

	Light cell body	Dark cell body	External surface	
			Light cell	Dark cell
SBA	++	−	−	−
UEA	−	−	+++	+++
Con A	−	−	+++	+++, −
MPA	−	−	−	−

SBA, soybean agglutinin; UEA, *Ulex europaeus* agglutinin; Con A, concanavalin A agglutinin; MPA, *maclura pomifera* agglutinin.

Intensity of lectin labeling was qualitatively evaluated by visual judgment as negative (−), weak (+), moderate (++), or intense reaction (+++). Reaction of cell body was examined by light microscopy, and that of external surface, by electron microscopy.

These findings indicate that there are at least two types of dark cells in these structures. There is a possibility that the presence of Con A- or MPA-binding glycoconjugate residues may represent different stages of secretory activity and cell maturity in the dark cells of the lacrimal sac and the conjunctiva, respectively.

In the present study, the mucus granules in the goblet cells were positively labeled with SBA and MPA. The positivity to SBA and MPA indicates the presence of terminal N-acetyl-galactosamine[15] and of galactose residues on the mucin macromolecule, respectively. Although MPA is also bound to the glycocalyx of the conjunctival cells, in addition to the goblet cell mucus, SBA shows negative reaction to the cell surface glycocalyx. The carbohydrate is attached mainly to intrinsic plasma membrane molecules.[6] However, the glycocalyx can also contain both glycoproteins and proteoglycans that have been secreted and then absorbed through the cell surface.[16] Therefore, the negativity of the external surface of the conjunctiva to SBA seems to be inconsistent with other findings. It is possible that some kind of inhibiting factor might influence the SBA binding to the glycocalyx on the apical surface of the conjunctiva. In the human conjunctiva, peanut agglutinin (PNA) shows similar binding patterns to SBA; positive binding to the goblet cell mucus and negative binding to the external surface of the conjunctiva.[10] Pretreatment of cryosections of human conjunctiva with neuraminidase caused increased staining of the external surface of the epithelium with PNA.[10,17] Neuraminidase digestion can be attributed to the removal of terminal sialic acid residues, which inhibit PNA binding to galactose-galactosamine residues.[12] In our preliminary labeling, wheat germ agglutinin,[8] which has an affinity for sialic acids, showed positive reaction on the external surface of the conjunctiva. Therefore, terminal sialic acid residues might inhibit the binding of SBA to N-acetyl-galactosamine residue on the cell surface of the conjunctival epithelium.

In the lacrimal sac, it appears that SBA specifically labels the cell body of the light epithelial cells. The light epithelial cell is characterized by pale cytoplasm and fewer mitochondria than the dark cell. The appearance of the light cells might indicate that SBA-positive substances are reserved in the cytoplasm like a holocrine-type secreting cell. Although the physiological meaning of the SBA-positive substance in the cell body remains to be established, the specific SBA labeling of the light cell may imply that the light cell has a markedly different glycoconjugate metabolism as compared with the dark cell.

The lacrimal sac is connected to the nasal cavity via the nasolacrimal duct. The present study focused on the difference in the lectin-binding pattern between the conjunctiva and the lacrimal sac. Whether there is a resemblance of the lacrimal sac mucosa to the nasal mucosa has been a continuing biological question. A number of lectin cytochemical studies have been reported for the murine nasal mucosa.[18,19] Based on those findings, the nasal mucosa is characterized by the presence of sialic acid. In the present study, SBA, UEA, Con A, or MPA did not show specific affinity to sialic acid. A study using sialic acid-specific lectins in the rabbit lacrimal and nasal mucosa might provide new data on this question.

References

1. de Toledo AR, Chandler JW, Buffman FV. Lacrimal system. Dry eye states and other conditions. In: Fodos SM, Yanoff M, eds. Textbook of ophthalmology. London: Mosby, 1994:145–6.
2. Dilly PN. On the nature and the role of the subsurface vesicles in the outer epithelial cells of the conjunctiva. Br J Ophthalmol 1985;69:477–81.

3. Versura P, Maltarello C, Caramazza R, Laschi R. Mucus alternation and eye dryness. Acta Ophthalmol 1989;67:455–64.
4. Chandlar JW, Gillette TE. Immunologic defense mechanisms of the ocular surface. Ophthalmology 1983;90:585–91.
5. Rodnot M. Die Flimmershaare des Traenensackepithels (Rasterelektronenmikroskopishe Untersuchungen). Klin Monatsbl Augenheilkd 1977;170:428–32.
6. Hirano H, Parkhause B, Nicolson GL, Lennox ES, Singer SJ. Distribution of saccharide residues on membrane fragments from a myeloma-cell homogenates: Its implication for membrane biogenesis. Proc Natl Acad Sci USA 1972;69:2945–9.
7. Ishikawa N, Horii Y, Nawa Y. Immune-mediated alteration of the terminal sugars of goblet cell mucins in the small intestine of *Nippostrongylus brasiliensis*-infected rats. Immunology 1993;78:303–7.
8. Kawano K, Uehara F, Ohba N. Lectin-cytochemical study on epithelial mucus glycoprotein of conjunctiva and pterygium. Exp Eye Res 1988;47:43–51.
9. Latkovie S. Ultrastructural localization of lectin-binding sites on the surface of the guinea pig conjunctival epithelium. Graefe's Arch Clin Exp Ophthalmol 1991;229:153–6.
10. Wells PA, DeSiena-Shaw C, Rice B, Foster CS. Detection of ocular mucus in normal human conjunctiva and conjunctiva from patients with cicatricial pemphigoid using lectin probes and histochemical techniques. Exp Eye Res 1988;46:485–97.
11. Goldstein IJ, Hayes CE. The lectins; carbohydrate-binding proteins of plants and animals. Adv Carbohydr Chem Biochem 1977;35:150–89.
12. Lis H, Sela B-A, Sachs L, Sharon N. Specific inhibition by N-acetyl-D-galactosamine of the interaction between soybean agglutinin and animal cell surfaces. Biochem Biophys Acta 1970;211:582–5.
13. Mann IC. The development of the human eye. 3rd ed. New York: Grune & Stratton, 1964.
14. Holthofer H, Virtanen I, Kariniemi A-L, Hormia M, Linder E, Miettinen A. *Ulex europaeus* I lectin as a marker for vascular endothelium in human tissues. Lab Invest 1982;47:60–6.
15. Lis H, Sharon N. Lectins as molecules and tools. Ann Rev Biochem 1986;55:35–67.
16. Olden K, Bernard BA, Humphries MJ, et al. Function of glycoprotein glycans. Trends Biochem Sci 1985;10:78–82.
17. Lotan R, Skutelsky E, Dannon D, Sharon N. The purification, composition, and specificity of the anti-T lectin from peanut (*Arachis hypogaea*). J Biol Chem 1975;250:8518–23.
18. Ueno K, Hanamura Y, Ohyama M. Differences in terminal carbohydrate structures of sialomucin in the murine nasal cavity. Eur Arch Otorhinolaryngol 1994;251:119–22.
19. Lundh B, Brockstedt U, Kristensson K. Lectin-binding pattern of neuroepithelial and respiratory epithelial cells in the mouse nasal cavity. Histochem J 1989;21:33–43.