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Research Article

LIGHT AND SCANNING ELECTRON MICROSCOPIC STUDIES ON LINGUAL TONSIL OF GOAT

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ABSTRACT

Lingual tonsils of 15 young goats were studied by light and scanning electron microscopy. Their outer surface lined by stratified squamous non-keratinized epithelium showed reticular epithelium associated with lymphoid tissue in the crypt areas. The epithelium comprised of strata basale, spinosum and superficiale presented varying numbers of cell layers. The strata were indiscernible in the reticular areas where these were infiltrated by different lymphoid cells. Small amount of lymphoid tissue in the form of dense aggregates and few lymphoid follicles was localized irregularly in the superficial lamina propria mucosae. Mucus glandular acini present in deeper part were PAS positive for acidic and neutral mucopolysaccharides. The lipids were sparsely distributed in the epithelium and the lamina propria mucosae. Scanning electron microscopy revealed elevated surface with flat squamous cells having microplicae which were continuous with those of adjacent cells.

Key words: Stratified squamous epithelium, lingual tonsil, goat

The tonsils of head region are sites of first contact of the immune system with ingested or inhaled antigens and may play role in mucosal immune protection (Gebert *et al.*, 1995). The lingual tonsil formed by the collection of tonsillar or crypto-lymphatic units at the base of the tongue (Nair and Rossinsky, 1984) has been termed follicular glands (Balgdrusen) with structural similarity to tonsil and other lymphoid tissue (Kolliker, 1855).The lingual tonsil has been studied extensively in the horse (Kumar and Timoney, 2005 a, b). The present study has been envisaged to explore histomorphological and surface structure of the lingual tonsil in goat.

MATERIALS AND METHODS

The heads of 15 young male goats of about 6-9 months age were procured from local slaughter house immediately after decapitation and lingual tonsils were collected. The tissues for histomorphology were collected from 5 heads, fixed in 10 per cent neutral buffered formalin and processed for routine paraffin technique. Paraffin sections (5–6 μ) were stained with routine Harris' hematoxylin and eosin stain, Weigert's method for elastic fibres, Gomori's method for reticulum, Bielschowsky's method for axis cylinder and dendrites, Ayoub-Shklar's method for keratin and pre-keratin (Luna, 1968), Crossman's trichrome stain for collagen fibres (Crossman, 1937).

The paraffin sections were also used to demonstrate mucopolysaccharides and proteins. The sections were stained by McManus' method for glycogen (PAS), alcian blue method for mucosubstances (pH 2.5), PAS-alcian blue method for mucosubstances (pH 2.5), diastase digestion method (Luna, 1968), Best's carmine method for glycogen (Drury and Wallington, 1967), mercury bromphenol blue method, performic acid alcian blue method (Pearse, 1968). In addition, frozen sections of 10-12 μ made from another 5 animals were preserved at -20°C in the cryostat and stained by oil-red-o in propylene glycol method, Sudan black-B method and Nile blue sulphate method for lipids (Pearse, 1968).

Fresh tissues from 5 goat heads collected for scanning electron microscopy (SEM) were fixed in 2 per cent glutaraldehyde solution for

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6-8 h after thorough washing in chilled 0.1 M phosphate buffer (pH 7.4). The tissues were again washed twice with 0.1 M phosphate buffer and rest of the procedure was carried out at EM-Laboratory, All India Institute of Medical Sciences, New Delhi. The tissues were critically point dried, fixed on stubs and sputter coated before viewing in the scanning electron microscope (Leo-435VP, Japan).

RESULTS AND DISCUSSION

Histology: Outer surface of lingual tonsil was lined by stratified squamous non-keratinized epithelium (Fig. 1) as reported in the horse (Kumar and Timoney, 2005 a). The free epithelial surface was irregular whereas deepest part presented papillae or interpapillary pegs which extended in the superficial part of the lamina propria mucosae (Fig. 1). The epithelium was comprised of strata basale, spinosum and superficiale layers as reported in the horse (Kumar and Timoney, 2005 a). The stratum basale was constituted by single layer of columnar shaped cells having oval to rod shaped nuclei with darkly stained dense chromatin material masking the presence of nucleoli. These cells possessed granular eosinophilic cytoplasm. Stratum spinosum had varying number (8-12) of cell layers. Its deepest nuclei resembled to those of stratum basale whereas superficially placed round to oval nuclei were less basophilic, larger and oriented parallel to longitudinal axis of the epithelium. The fine granular cytoplasm of these cells was eosinophilic with slight basophilic tinge, and presented spine like arrangement. A few nuclei with regressive changes were also interspersed in between other nuclei. Outermost stratum superficiale of varying thickness had small, round, oval or elongated basophilic nuclei. The nuclear density gradually reduced toward outer surface. The cytoplasm was more eosinophilic, finely granular and uniform.

The surface epithelium was modified into reticular epithelium (Fig. 1) toward small crypts. The latter were only few and were localized superficially. The crypts and associated reticular epithelium were extensive and extended into deeper part of lamina propria mucosae in the lingual tonsil of the horse (Kumar and Timoney, 2005 a). The reticular epithelium lacking interpapillary pegs, had fewer cell layers with indistinct strata and was associated with lymphoid tissue. Its stratum basale was heavily infiltrated by lymphoid cells. At places only a single layer of epithelial cells infiltrated by lymphoid cells was observed. This arrangement has been termed as lymphoepithelial symbiosis because of its lymphoid appearance and absence of classic epithelial features (Fioretti, 1957). Characteristic of reticular epithelium was the coexistence of epithelial and non-epithelial cells and, sometimes there was predominance of the latter as reported in the horse (Kumar and Timoney, 2005 a). The non-epithelial components included a varying number of lymphocytes, plasma cells, and macrophages.

The lamina propria mucosae had dense irregular connective tissue which extended in the spaces between interpapillary pegs. The fine reticular fibres formed regular basement membrane and in addition formed meshwork which along with elastic fibers extended toward the interpapillary pegs. The deeper part was constituted by connective tissue cells and fibres, lymphoid tissue, clumps of mucus acini and fatty tissue. A few nerve bundles present in the superficial part showed their increased concentration in deeper part. The lymphoid tissue was distributed in dense aggregates (Fig. 1) and a few lymphoid follicles of different dimensions which generally possessed a peripheral darkly stained corona. These follicles were separated from each other by a dense meshwork of reticular fibres and a few collagen, elastic and nerve fibres. The collagen and reticular fibres also surrounded the lobules of glandular acini, outer wall of the blood vessels and interglandular spaces. The concentration of longitudinally oriented elastic fibres was more in deeper part. The glandular acini were purely mucus (Fig. 2). Their pyramidal cells possessed elongated rod shaped and strongly basophilic nuclei. The less eosinophilic cytoplasm was vacuolated due to wash out of the mucus (Fig. 2) but it accentuated toward intercellular junctions. The myoepithelial cells observed in interacinar spaces had elongated nuclei. The intraglandular and

interglandular ducts had simple to stratified cuboidal epithelium, generally of two cell layers thickness (Fig. 2). These ducts opened on the epithelial surface. The deeply placed lobules of glandular acini were separated from each other by connective tissue and striated muscles. The latter were oriented in longitudinal, vertical and oblique planes. The bundles of these muscle fibres were separated from each other by similarly oriented collagen fibres and a few reticular fibres. The deepest part of the lamina propria mucosae showed fatty tissue, blood vessels and striated muscles cut in different planes.

Histochemistry: The stratified squamous epithelium and most of the lamina propria mucosae showed negative PAS reaction by McManus' method. The PAS activity localized in the mucus glandular acini was strong toward their basal parts. A reduced PAS activity after diastase treatment showed presence of mucopolysaccharides other than glycogen. In addition, acidic and neutral mucopolysaccharides were also observed. Alcian blue method exhibited a moderate to strong reaction. Inter and intraglandular ducts were devoid of PAS positive reaction like those of the horse (Kumar and Timoney, 2005 a). Sudan black-B and oil-red-o methods demonstrated very less amount of lipids in the epithelium. Superficial part of the lamina propria mucosae showed small-localized areas for lipid activity whereas their concentration increased around the glandular acini. Neutral lipids in the form of very large droplets were observed in deep lamina propria mucosae adjacent to the striated muscles. A negligible reaction for proteins that too inconsistently, could be demonstrated in the lamina propria mucosae.

Scanning electron microscopy: SEM of the lingual tonsil presented a round elevated surface with flat cells of varying shapes. These cells were attached with each other by distinct junctions which, could be delineated at higher magnification. Some of these cells appeared in the form of small leaf like scales were exfoliated due to desquamation of epithelial cells (Fig.3). These squamous cells presented membranous projections in the form of microplicae of different patterns resembling to finger prints of humans as reported in the horse (Kumar and Timoney, 2005 a). Some

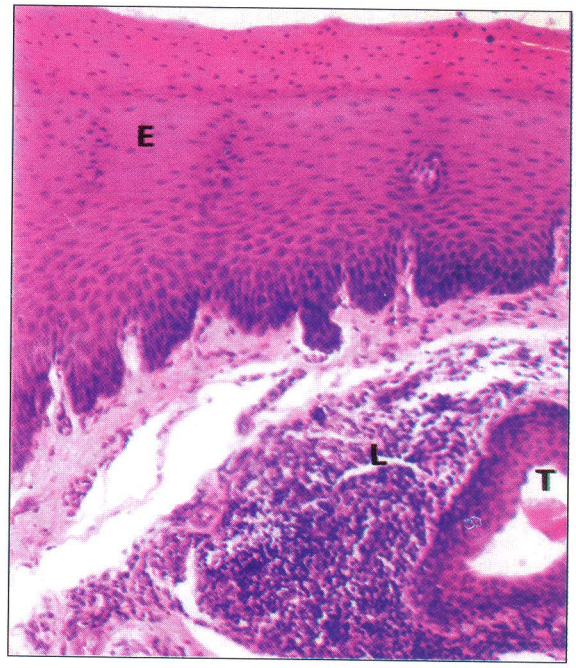

Fig.1. Photomicrograph of lingual tonsil showing stratified squamous epithelium (E) toward outer surface and reticular epithelium (T) associated with lymphoid tissue (L) toward crypt. (H. & E. x 50)

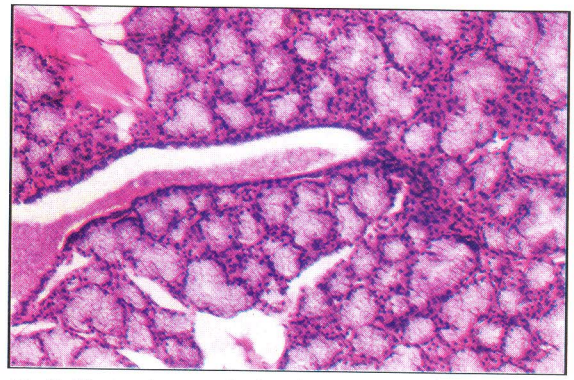

Fig.2. Photomicrograph showing presence of mucus acini in the deeper part of lamina propria mucosae. Note stratified cuboidal epithelium of glandular duct. (H. & E. x 50)

Fig.3. Scanning electron micrograph of lingual tonsil showing round elevated and adjacent irregular surface. (x 171).

of the patterns were close channel like whereas others had an open arrangement. The microplicae of one cell were continuous with those of adjacent cells and at places of desquamation, these were continuous with those of deeper ones. The junctions between adjacent cells were distinct due to dense uniform arrangement of microplicae.

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