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Research Article

HISTOMORPHOLOGICAL AND HISTOCHEMICAL STUDIES OF THE MEIBOMIAN AND CILIARY GLANDS OF GOAT (*CAPRA HIRCUS*)

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ABSTRACT

The present study was conducted on six healthy young goats of local mixed breed of either sex to study the histomorphology and histochemistry of the Meibomian and ciliary glands of goat. Meibomian glands of goat were located in the tarsal plate of both upper and lower eyelids. The tarsal glands were numerous and better developed in the upper than in the lower eyelid. Tarsal glands were lobulated holocrine gland and consisted of solid glandular mass surrounding a central excretory duct. The glandular acini were compound tubuloalveolar, sebaceous and encapsulated by tarsal plate. The glandular acini were separated from each other by connective tissue especially collagen and elastic fibers along with blood vessels of different dimensions. The ciliary glands (glands of Moll) were modified apocrine sweat glands located on the margin of the eyelid, placed next to the base of the eyelashes. The histological studies revealed that the glands of Moll were small, simple tubular, unencapsulated and serous gland. The secretary unit were lined by cuboidal to columnar cells and showed an irregular narrow lumen. Layers of collagen and muscle fibers of different density were demonstrated between the apocrine units. Histochemical studies of both Meibomian and ciliary glands showed the absence of neutral mucopolysaccharides, weakly acidic sulphated mucosubstances, sialomucins and hyaluronic acid.

Keywords: Ciliary gland, Goat, Histology, Histochemistry, Meibomian gland

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The Meibomian gland, also known as tarsal gland, is a holocrine type of exocrine gland responsible for the supply of meibum, an oily substance that spreading the tear on the cornea and prevents evaporation of the eye's tear film (Al-Rikabi, 2015). This oily layer forms coating outside the tear film that keeps tears from drying up too quickly. Dysfunctional Meibomian glands often cause dry eyes, one of the most common eye conditions. The ciliary glands, also known as glands of Moll, have apocrine secretion and leave their secretions inside the eyelash follicles. The ciliary and tarsal glands may play an essential role in the moistening and the protection of eyelid and ocular surface (Yasui *et al.*, 2006). Dermis part of the palpebral conjunctiva contains organized lymphoid follicles and these were termed as conjunctiva-associated lymphoid tissue (CALT). CALT seems to play an important role in the protection of ocular surface by initiating and regulating immune responses (Steven and Gebert, 2009). Although the research has been carried out on the histology and histochemistry of the Meibomian and ciliary glands in cattle (Ahmed, 2012), Philippine water buffalo (Maala *et al.*, 2009), dromedary camel (Al-Ramadan, 2015), calf (Yuksel *et al.*, 2005) and pig (Yasui *et al.*, 2006). However, there is a paucity of literature on the light microscopic details of the Meibomian and ciliary glands in goat. Keeping in view the pathophysiological significance of these glands, the present study was undertaken to elucidate the histological and histochemical findings of Meibomian and ciliary glands in goat.

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MATERIALS AND METHODS

The present study was conducted on 06 healthy young goats of either sex, of local mixed breed. The heads were procured from local slaughter house immediately after decapitation and the tissues were collected from the upper and lower eyelids. The tissues were fixed in a 10% neutral buffered formalin solution for 48 hours, subjected to routine tissue processing for light microscopic examination and embedded in paraffin blocks. The paraffin sections (5-6 µ thick) were made through the upper and lower eyelids of left as well as right eyes and stained with routine Harris' hematoxylin and eosin stain for general histomorphological examination, Gomori's stain for reticular fibres, Weigert's method for elastic fibres (Luna, 1968) and Crossman's trichrome stain for collagen fibres (Crossman, 1937). For histochemical demonstration of mucopolysaccharides Periodic Acid Schiff-Alcian blue method, Alcian blue (pH 2.5) method, Colloidal iron method and Mayer's mucicarmine method (Luna, 1968) were used.

RESULTS AND DISCUSSION

Anatomically upper and lower eyelids comprised of anterior and posterior surface. The anterior surface of the eyelids is made of thin and delicate skin covered with short hairs as observed earlier in cattle (Ahmed, 2012). The skin was covered by a keratinizing stratified squamous epithelium (Fig. 1). These results were in concordance with the findings

in local Iraqi breed goats (Al-Rikabi, 2015). Dense irregular connective tissue especially collagen and elastic fibers along with blood vessels of different dimensions were found directly beneath the epidermis. Long and thick eyelashes arranged in rows grew from the anterior palpebral margins. Sebaceous glands (Zeis glands) and ciliary (Moll) glands, which are modified sweat glands located deep in the follicle. The muscle layer was situated deep in the dermis and consisted of bundles of the orbicularis oculi muscle (Fig. 1 & 2). Under the muscle layer was a layer of connective tissue with blood vessels and nerve fibres. Similar findings were reported in local Iraqi breed goats (Al-Rikabi, 2015). Transition of epithelium from non-keratinized stratified squamous epithelium to pseudo-stratified columnar epithelium with goblet cells was observed on the posterior surface of the eyelids/ palpebral conjunctiva. The numerous goblet cells were identified in the palpebral conjunctiva zone of the lower eyelid as compared to upper eyelid as observed by Kleckowska-Nawrot *et al.* (2016) in red kangaroo and Voigt *et al.* (2012) in chinchillas. However, the palpebral conjunctiva of dromedary camel was lined by stratified squamous to stratified columnar with goblet cells except over the lymphoid follicles (Al-Ramadan, 2015). Whereas, stratified squamous epithelium was observed at the palpebral edges in horse (Bourges-Abella *et al.*, 2007). Variation in the palpebral epithelium was reported in other domestic animal species (Samuelson, 2007).

The conjunctiva-associated lymphoid tissue (CALT) in the form of organized lymphoid follicles was also observed towards palpebral conjunctiva of both upper and lower eyelids (Fig. 5 & 6). Similar findings were reported in Angora goats, Japanese monkeys and domestic animals (Cain and Philips, 2008), chickens (Fix and Arp, 1991) and chinchillas (Voigt *et al.*, 2012). However, CALT was in the form of several lymphoid follicles on the palpebral surface of the eyelids of dromedary camel (Al-Ramadan, 2015). Whereas in red kangaroo, CALT was observed as organized lymphoid follicles only in the lower eyelid (Kleckowska-Nawrot *et al.*, 2016). The goblet cells towards the palpebral conjunctiva showed presence of strong reaction for Alcian blue indicating the presence of sialomucins and hyaluronic acids towards the Alcian blue stain (pH 2.5) (Fig. 6). The colloidal iron and Mayer' mucicarmine showed positive activity in goblet cells for mucopolysaccharides. Under the epithelium, there was loose connective tissue which contained several blood vessels. Similar findings were reported in cattle (Ahmed, 2012), dromedary camel (Al-Ramadan, 2015) and guinea pig (Gasser *et al.*, 2011).

Meibomian glands of goat were located in the tarsal plate of both upper and lower eyelids. The tarsal glands were numerous and better developed in the upper than in the lower eyelid as reported earlier by Maala *et al.* (2009) in Philippine water buffalo, Al-Rikabi (2015) in dromedary camel and Ahmed (2012) in cattle. However, tarsal glands were found to be less prominent in the upper eyelid in the porcine (Yasui *et al.*, 2006). The long axis of gland was perpendicular to the eyelid margin (Fig. 1). In transverse section, the appearance of gland was spherical to oval and was arranged in a parallel row between skin, connective tissue and muscle layer on outer surface and palpebral conjunctiva on inner side (Fig. 2). These findings were in fully agreement with Al-Rikabi (2015) in local Iraqi breed goats, Raymond-Letron *et al.* (2012) in dog and Al-Ramadan (2015) in dromedary camel. Tarsal glands were lobulated holocrine gland consisted of solid glandular mass surrounding a central excretory duct (Figs. 2 & 3). The glandular acini were compound tubuloalveolar, sebaceous gland as reported earlier by Maala *et al.* (2009) in Philippine water buffalo. The acini were separated from each other by connective tissue especially collagen (Fig. 4) and elastic fibers along with blood vessels of different dimensions. These results were in concordance with the findings in dromedary camel (Al-Ramadan, 2015). The glandular acini were comprised of polyhedral (meibocytes) cells. The centrally placed cells were vacuolated with lightly stained nucleus (Fig. 3). Few cells were devoid of nuclei. These findings were in fully agreement with those reported in dromedary camel (Al-Ramadan, 2015).

A periodic acid-Schiff (PAS) negative reaction was observed in the secretory units of the gland indicating the absence of neutral mucopolysaccharides as described earlier in Philippine water buffalo (Maala *et al.*, 2009) and chinchillas (Voigt *et al.*, 2012). Secretory units and cells showed absence of weakly acidic sulphated muco substances, sialomucins and hyaluronic acid at pH 2.5. Similar findings were reported in Philippine water buffalo (Maala *et al.*, 2009) and guinea pig (Gasser *et al.*, 2011). PAS-Ab technique revealed negative reaction for both neutral and acid mucopolysaccharides in the secretory units (Fig. 5). The epithelium of the ducts also showed negative reaction for both PAS and AB methods (Fig. 6). Mayer's mucicarmine and colloidal iron showed negative reaction which indicated the absence of acid mucopolysaccharides in the secretory cells (Fig. 7). However, PAS positive reaction was observed in both glands and in the apical portion of cytoplasm of ducts in dromedary camel (Al-Ramadan, 2015).

The ciliary glands (glands of Moll) were modified apocrine sweat glands located on the margin of the eyelid, placed next to the base of the eyelashes (Fig. 8). Similar findings were also reported in cattle (Ahmed, 2012). Sebaceous (Zeis) glands were also observed with the hair

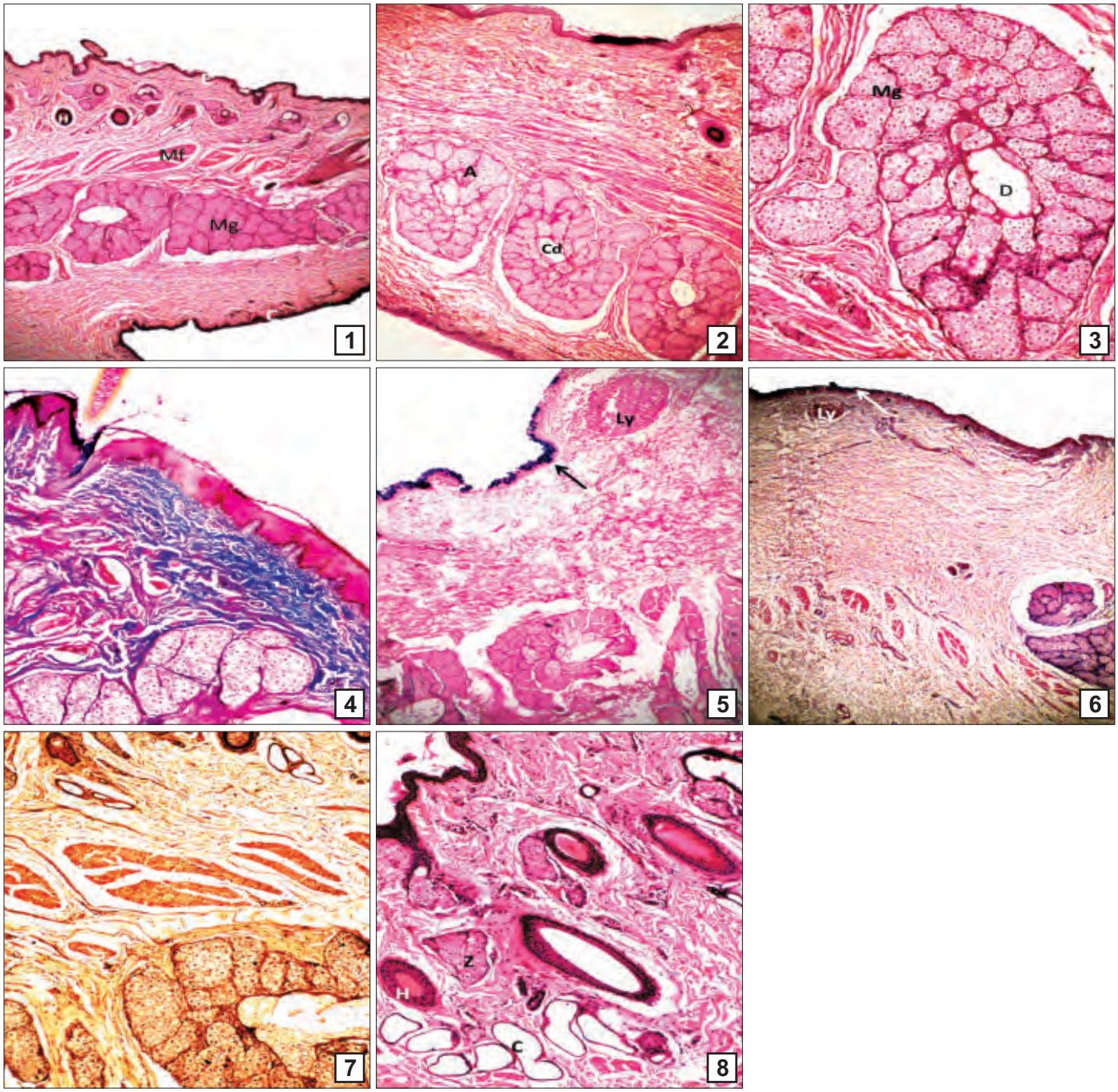

Fig. 1-8. (1) Photomicrograph of eyelid of goat showing various histological layers (E = epidermis; H = hair follicles; Z = zeis glands; Cg = ciliary/Moll glands, Mf = muscle fibres of orbicularis oculi; Mg = Meibomian/tarsal glands; C= conjunctiva). H & E × 40; **(2)** Photomicrograph of eyelid of goat showing the linear arrays of Meibomian glands (Mg) arranged in grape-like clusters attached to a central stalk (Cd). H & E × 100; **(3)** Higher magnification showing large sized sebaceous (Meibomian) glands of goat eyelid with alveoli (A) emptying into long central duct (Cd). H & E × 200; **(4)** Photomicrograph of eyelid of goat showing the connective tissue mainly made up of collagen fibres (blue colour). Crossman Trichrome × 100; **(5)** Photomicrograph of eyelid of goat showing a negative PAS-AB reaction for both neutral and acid mucopolysaccharides in the secretory units; however the goblet cells (arrow) showed the presence of acid mucopolysaccharides. Lymphatic nodules (Ly) were also seen in the subconjunctival tissue towards the palpebral conjunctiva. PAS-AB × 100; **(6)** Photomicrograph of eyelid of goat showing a negative reaction for sailomucins, hyaluronic acid and mucopolysacharides in the secretory units; however the goblet cells (arrow) showed the presence of these substances. Lymphatic nodules (Ly) were also seen in the subconjunctival tissue towards the palpebral conjunctiva. AB × 100; **(7)** Photomicrograph of eyelid of goat showing a negative reaction for muco substances in the secretory units. Mayer's Mucicarmine × 100; **(8)** Photomicrograph of eyelid of goat showing the modified apocrine (ciliary) glands (Cg), hair follicles (H) and Zeis glands (Z). The secretory portion of ciliary glands showed large irregular lumen. H & E × 100

follicles. The histological studies revealed that the glands of Moll were simple coiled tubular gland as observed in Philippine water buffalo (Maala *et al.*, 2009). The secretory unit were lined by simple cuboidal to columnar cells and showed an irregular narrow lumen (Fig. 8). The cells had round, centrally placed nuclei with cytoplasm containing large eosinophilic granules. Similar findings were reported in pig (Yasui *et al.*, 2006). Layers of collagen and muscle

fibers of different density were demonstrated around the apocrine unit. PAS-Alcian blue method showed negative activity for both acidic and neutral mucopolysaccharides in the glandular acini. The acini also showed negative reaction for glycogen by McManus' method as observed earlier by Ahmed (2012) in cattle. Mayer's mucicarmine and colloidal iron also showed negative activity for acidic mucopolysaccharides in the acini. The acini presented the absence of weakly acidic sulphated muco-substances, hyaluronic acid and sailomucins as described earlier in Philippine water buffalo (Maala *et al.*, 2009). Yasui *et al.* (2006) in pig, observed weakly positive reaction intensities in the secretory epithelium and luminal secretion.

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