

MICROANATOMY OF SEBACEOUS GLANDS IN DEER, GOAT AND SHEEP

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ABSTRACT

Histology, histochemistry and ultra-structure of sebaceous glands in deer, goat and sheep were conducted using skin samples collected from spotted deer brought for post mortem from Thrissur zoo and forest department; and from goat and sheep freshly slaughtered in Meat Technology Unit, Mannuthy. Sebaceous glands appeared as large, lobulated, sac-like structures associated with the hair follicles. They were of simple branched alveolar type, arranged in a rosette fashion around the bristles. All these were embedded in the dermis and not extended into the subcutaneous tissue. They were most abundant in dorsal neck region among the different areas under study. The secretions of sebaceous glands exhibited positive reaction as green fluorescence to Fluorescein iso-thiocyanate (FITC) conjugated lectin from *Ulexeuropaeus* (UEA). The intensity of reaction was more in sheep among the three species studied. Sebaceous glands and ducts exhibited diffused positive reaction for lipids. A diffused positive reaction to the acid phosphatase was also noticed in sebaceous glands. Transmission and scanning electron

microscopy revealed the branched nature of sebaceous glands.

Keywords: Deer, goat, sheep, micro-anatomy, sebaceous gland

INTRODUCTION

Mammals have an array of different skin glands. These include the sebaceous sweat, wax producing and mammary glands. Sebaceous glands are simple branched alveolar glands generally associated with hairs. In certain places of the body, they occur independent of hair too, viz., glans penis, prepuce, labia vulvae, anus, external ear canal and tarsal glands of eyelids. Sebaceous glands secrete sebum into the hair follicle. The sebum is an oily substance resulting from the destruction of the epithelial cells, hence holocrine. It keeps the hair supple and prevents the growth of bacteria. Sebaceous glands are absent in the foot pads, hoofs, claws, horns and teats. Even though a few studies reveal the structural capabilities of sebaceous glands in large ruminants (Miranda and Jenkinson, 2015; Smith and Ahmed, 1976 and Taha, 1988), the present study was carried out to elucidate the comparative histological,

histochemical and ultrastructural characteristics of sebaceous gland in deer, goat and sheep.

MATERIALS AND METHODS

Histology, histochemistry and ultrastructure of sebaceous gland in deer, goat and sheep were explored using skin samples collected from spotted deer brought for post mortem at College of Veterinary and Animal Sciences, Mannuthy, from Thrissur zoo and forest department; and of goat and sheep freshly slaughtered in Meat Technology Unit, Mannuthy. Samples of one cm³ were collected for histological and histochemical studies from 27 regions of skin, viz., muzzle, infraorbital, horn glands, dorsal face, lateral face, ventral face, pinna ear, dorsal neck, lateral neck, ventral neck, dorsal abdomen, lateral abdomen, ventral abdomen, dorsal forelimb, palmar, dorsal hindlimb, plantar, interdigital forelimb, interdigital hindlimb, foot pad forelimb, foot pad hindlimb, inguinal, preputial, scrotal (from male), dorsal thorax, perineum and dorsal nasal. Specimens for histological purpose were fixed in 10 per cent neutral buffered formalin (10% NBF) for 48 hours. The fixed specimens were washed, dehydrated and embedded in high melting paraffin (MP 58-60°C). Serial sections of 5µm thickness were made and stained histologically using haematoxylin and eosin for routine studies, Gomori's one step trichrome method for collagen and muscle fibres and Ayoub Shalkar method for Keratin and pre-keratin (Luna, 1968). Histochemical observations were recorded after staining with Oil Red 'O' in propylene glycol method for lipids, Gomori's alkaline phosphatase cobalt method and Gomori's method for acid phosphatase (Singh and

Sulochana, 1996). Digital images were stored in Leica DM 2000 LED microscope. Lectin histochemistry was done using Fluorescein iso-thiocyanate (FITC) conjugated lectin from *Ulexeuropaeus* 1 (West *et al.*, 2012) and examined under Fluorescence (Leica DM 2000 LED) microscope with green filter. Samples of one mm³ size were fixed in 2.5% gluteraldehyde in 0.1 M phosphate buffer (pH 7.2) for 24 hrs at 4°C and processed for Scanning Electron Microscopy (SEM - Model: JOEL-JSM 5600) and Transmission Electron Microscopy (TEM - Model: Hitachi, H-7500 from JAPAN) as per the standard procedures (Bozzola and Russell, 1998) at Ruska labs, College of Veterinary Science, Hyderabad.

RESULTS AND DISCUSSION

Sebaceous glands appeared as large, lobulated, sac-like structures associated with the hair follicles in all three species studied. They were of simple branched alveolar type as revealed by scanning electron microscopy (Fig. 1) and they were arranged in a rosette fashion around bristles. The lobes appeared as round, oval, triangular, quadrilateral or elongated ones. All these were embedded in the dermis and did not extend into the subcutaneous tissue. The secretory units consisted of a solid mass of epidermal cells, enclosed by a connective tissue sheath that blended with surrounding connective tissue of the dermis. At the periphery of the glandular mass, a single layer of low cuboidal cells with round nucleus rested on the basal lamina (Fig. 2 & 3). As they moved inward, they became polygonal or spheroidal and accumulated numerous lipid droplets. The nuclei gradually shrunk and disappeared. The smaller peripheral cells contained

only a few small fat droplets or none at all. The inner cells of secretory unit were larger. The cytoplasm of the most central ones and those in the lumen of the duct almost converted into fat and their nuclei were disintegrated. Necrotic changes were observed in these cells as they are pushed further towards the centre of the gland. In the middle zone, the cells were in intermediate stages. Thus the secretion of the sebaceous glands in the present study indicated a holocrine nature; as against the observations of Miranda and Jenkinson (2015) in cattle, who found that the mode of secretion of sebaceous glands in cattle unlikely to be holocrine, since the glandular mitotic activity was not altered by single or repeated stimulation, and the number of cells necessary to produce the sebum by a holocrine mechanism greatly exceeded the estimated level of cell production.

A diffused positive reaction to the acid phosphatase was noticed in sebaceous glands in the species under study, probably indicating the moderate lysosomal activity in the necrotic cell stages of the holocrine gland.

The alveoli of the sebaceous glands opened into a short duct. These ducts opened through pilosebaceous canal into the upper portion of the hair follicle (Fig. 4 & 5). The glands were most abundant in dorsal neck region among the different areas under study. Most of them opened by only one duct. It was lined by stratified squamous epithelium and was continuous with the outer root sheath of the hair follicle.

In the present study, the secretions of sebaceous glands (Fig. 6) exhibited positive reaction as green fluorescence

to Fluorescein iso-thiocyanate (FITC) conjugated lectin from *Ulexeuropaeus* (UEA). The intensity of reaction was more in sheep among the three species studied probably owing to the chemical nature of the secretion in the species and also to the stages of cell division, since the pattern of lectin binding to routinely processed sections of normal skin is related to cellular maturation.

Sebaceous glands and ducts in all three species studied exhibited diffused positive reaction for Oil red O indicating the presence of lipids in the secretion, in concurrence with reports by Smith and Ahmed (1976) in cattle, who detected the presence of a higher proportion of phospholipid and unesterified fatty acid and a lower proportion of triglyceride in the sebaceous glands.

Transmission electron microscopy clearly exhibited the lipid granules in the sebaceous ducts in the present study (Fig. 7) which is in accordance with the findings of Taha (1988) in camel, he revealed that cells at different stages of development and maturation are distinct in the sebaceous gland of the camel, the cells those are programmed to produce lipid secretion are probably the ones which loose contact with the basal lamina. Various forms of smooth endoplasmic reticulum are also reported in the sebaceous cells, viz., grid lattice, membranous whorls and parallel cisterns; all of them are associated with lipid droplets which suggest that the smooth endoplasmic reticulum may be involved in lipid synthesis.

SUMMARY

Sebaceous glands appeared as large, lobulated, sac-like structures associated with the hair follicles with similar structure in all three species studied. The secretion of the sebaceous glands indicated a holocrine nature. A diffused positive reaction to the acid phosphatase was noticed in sebaceous glands in all the species under study. The glands were most abundant in dorsal neck region among the different areas studied. Secretions of sebaceous glands exhibited positive reaction to FITC conjugated lectin from UEA. The intensity of reaction was more in sheep among the three species studied probably owing to the chemical nature of the secretion in the species and also to the stages of cell division. Sebaceous glands and ducts in all three species studied exhibited diffused positive reaction for Oil red O indicating the presence of lipids in the secretion.

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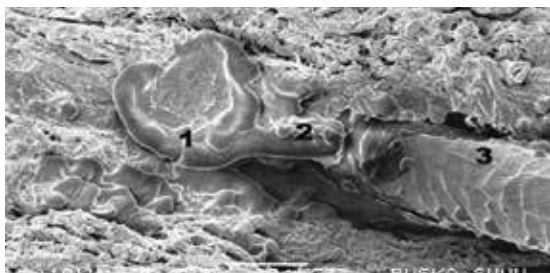


Fig. 1. Dorsal abdomen. Six years-old female deer. SEM x 850, 1. Branched tubule of sebaceous gland 2. Duct 3. Hair

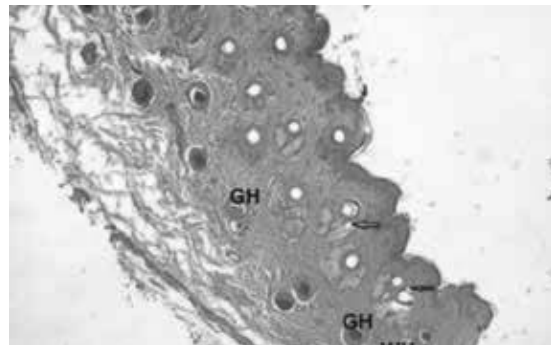


Fig. 2. Two days-old female goat. Base of horn. H & E x 40 GH - Guard Hair; WH (& Arrows) - Wool Hair

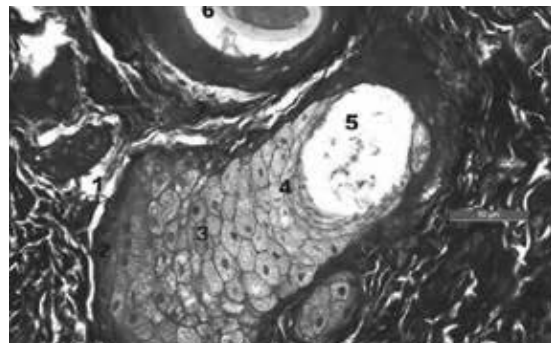


Fig. 3. Infraorbital region. Adult female sheep. Gomori's one-step Trichrome x 400 1. Basal lamina 2. Peripheral cells 3. Intermediate cells 4. Central cells 5. Lumen 6. Hair

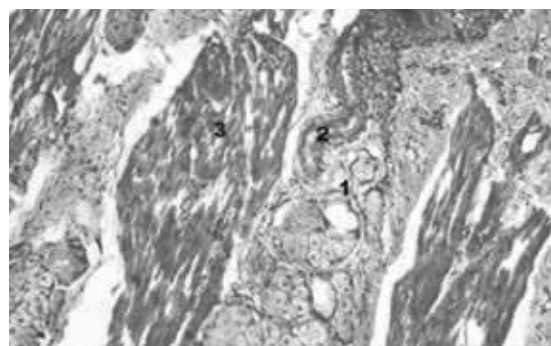


Fig. 4. Foetal deer Interdigital Region Hind Limb. Ayoub Shalkar method for Keratin and pre-keratin x 200 1. Sebaceous gland 2. Duct of sebaceous gland 3. Inter-follicular muscle

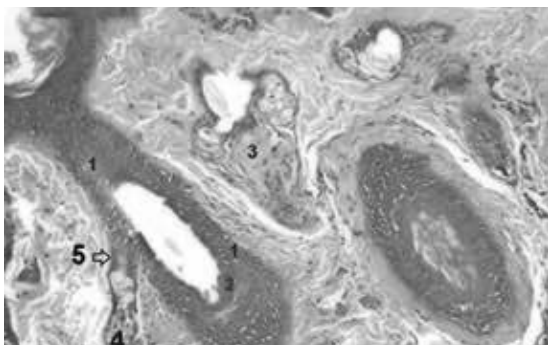


Fig. 5. Interdigital Region. Adult female goat. H & E x 100

1. Outer root sheath 2. Inner root sheath
3. Telogen germinative units 4. Sebaceous gland
5. Duct of sebaceous gland

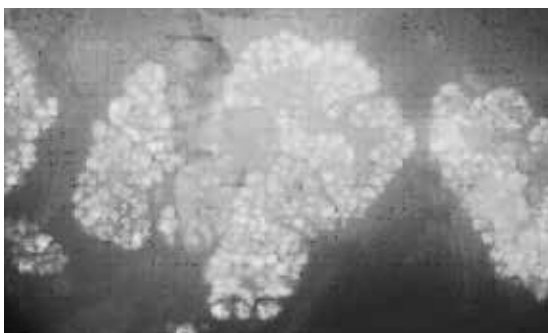


Fig 6. Sebaceous gland of adult sheep exhibiting positive response of green fluorescence to FITC-conjugated lectin from *Ulex europaeus* (UEA) x 100

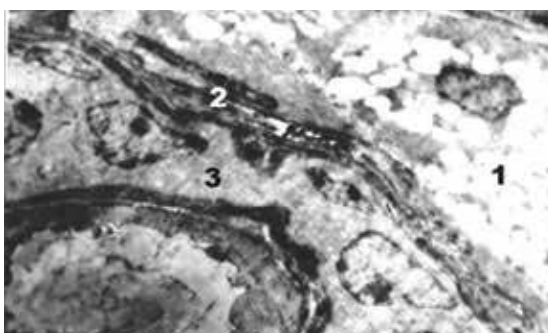


Fig. 7. Sebaceous gland with duct in the pinna of ear in six years-old female deer. TEM x 3860

1. Lipid granules 2. Basal Lamina with flat lining cells of duct 3. Gland

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