RESEARCH ARTICLE Indian Journal of Animal Research, Volume 57 Issue 1: 29-34 (January 2023)

Histomorphological, Histochemical and Histoenzymic Studies on the Dermis of Skin of Goat (*Capra hircus*)

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ABSTRACT

Background: Skin is a versatile organ, which is extremely important for protection, perception, water regulation and wound healing. The skin of goat is considered extremely durable used by the tannery Industry to make rugs and carpet binding.

Methods: The present study was conducted on the skin of goats of either sex to study the age wise changes in the histomorphology, histochemistry and histoenzymology of dermis. The skin samples were collected from Nagpur Muncipal Corporation Slaughter house immediately after natural death and subsequently processed and stained for histological, histochemical and histoenzymic studies. The total of 220 samples, 20 each from different body regions namely dorsal neck, lateral neck, ventral neck, dorsal thorax, lateral thorax, ventral thorax, dorsal abdomen, lateral abdomen, ventral abdomen, lateral thigh and medial thigh were collected. The animals were divided into two groups, *viz*. six to twelve month of age (Gr I) and above one year age (Gr II).

Result: It was observed that the dermis was composed of collagen, elastic, reticular fibers, cells of connective tissue, blood vessels, nerves, hair follicles, sweat glands, sebaceous glands and arrector pili muscles in all body regions in Gr I as well as in Gr II. The dermis was divided into thin superficial papillary layer and a thick deep reticular layer. The dermal papillae were the upward projections of superficial papillary layer between epidermal pegs. In reticular layer, fibroblasts were larger, while in papillary layer, fibroblasts were thin long, compressed and spindle shaped. The collagen fibers were more in number as compared to other fibers in dermis and they were densely arranged in papillary layer and parallel with the skin surface below the epidermis. The elastic fibers were present in between the collagen fibers around the blood vessels and hair follicles. The arteriovenous shunt was found surrounded by capsule of connective tissue consisting of smooth muscle cells. The more number of hair follicles at deep part of papillary layer and few follicles were present in reticular layer. The hair follicle composed of outer root sheath, inner root sheath, cortex and medulla. The depth of primary hair follicle increased with the advancement of age. Moderate PAS positive activity was noted in the papillary layer, while it was weak to moderate in reticular layer. The reticular layer of the dermis showed moderate Sudan black B activity for the presence of lipid than the papillary layer. The mild acid phosphatase activity was seen in the papillary and reticular layer of dermis, while moderate acid phosphatase activity was noticed in the blood vessels. A mild alkaline phosphatase activity was present in dermis in Gr I and Gr II.

Conclusion: The present study would form a baseline data on the histomorphology, histochemistry and histoenzymology of skin of goat which would be helpful in future research prospective.

Key words: Dermis, Goat, Histochemistry, Histoenzymology, Histomorphology.

INTRODUCTION

The skin is the largest organ in mammalian body and constitutes 8.5 per cent of live body weight (Mainde *et al*., 2019a). The skin of goat is often used for making gloves and boots. It is a versatile organ, which is extremely important for protection, perception, water regulation and wound healing (Razvi, 2015, Mainde *et al*., 2018 and Mainde *et al*., 2019b). The skin of goat is considered extremely durable used by the tannery Industry to make rugs and carpet binding. Skin is thicker on the dorsal and extensor surfaces than on the ventral and flexor surfaces. It is thicker in the male (Mainde *et al*., 2018). The capacity of the skin to move and be stretched depends on its own thickness, the number of its folds, its intrinsic elasticity and firmness of fixation by the tela subcutanea and age of individual. The thickness of skin varies on the basis of species, breed, age, sex and body region in domestic animals. The literature on the histomorphology, histochemistry and histoenzymology of skin of goat is scanty and therefore, the present study was undertaken to develop a baseline data for future research perspectives.

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How to cite this article: Mainde, U.P., Banubakode, S.B., Nandeshwar, N.C., Charjan, R.Y., Sathapathy, S. and Salankar, A.M. (2023). Histomorphological, Histochemical and Histoenzymic Studies on the Dermis of Skin of Goat (*Capra hircus*). Indian Journal of Animal Research. 57(1): 29-34. doi: 10.18805/IJAR.B-4243.

Submitted: 07-07-2020 **Accepted:** 10-11-2020 **Online:** 14-04-2020

MATERIALS AND METHODS

The skin samples of goats were collected from healthy animals irrespective of sex from Slaughter house of Nagpur

Municipal Corporation and also from animals after natural death. The total of 220 samples, 20 each from different body regions namely dorsal neck, lateral neck, ventral neck, dorsal thorax, lateral thorax, ventral thorax, dorsal abdomen, lateral abdomen, ventral abdomen, lateral thigh and medial thigh were collected. The animals were divided into two groups, *viz*. six to twelve month of age (Gr I) and above one year age (Gr II). The samples of skin were collected in ice box and brought to the laboratory. The skin was washed and cleaned by removing hairs using shaving blade and cut into pieces of 4-6 mm size. Samples were fixed in 10 per cent Neutral buffered formalin, Bouin's fluid and Zenker's fixative for histological and histochemical studies. Tissue blocks of paraffin were prepared and sections were cut at 4-5 µ thickness with the help of rotary microtome. Both transverse and vertical sections were taken from each body region. Sections were mounted on clean albuminized glass slide dried on a hot plate at 45 - 50°C for three hours. The sections prepared were stained with Haematoxylin and Eosin for study of normal histological structure as per the method of Singh and Sulochana (1996). Differential staining for demonstration of connective tissue was performed. Histochemical and histoenzymic staining for demonstration of polysaccharides, lipids, Alkaline and Acid Phosphatase activity were also performed on paraffin sections.

RESULTS AND DISCUSSION

Histology

The dermis comprised the chief layer of skin. The dermis was composed of collagen, elastic, reticular fibers, cells of connective tissue, blood vessels, nerves, hair follicles, sweat glands, sebaceous glands and arrector pili muscles in all body regions in Gr I as well as in Gr II. These observations of the present study are in accordance with the findings reported by Dellmann and Eurell (1998) in domestic animals, Mugale (2001) in cattle, Mandage (2003) in sheep and Kapadnis (2004) in goat.

During the present study, the dermis was divided into thin superficial papillary and a thick deep reticular layers (Fig 1) which was similar to the findings of Sathapathy *et al*. (2017) in Zebra. The connective tissue fibers of papillary layer blended with reticular layer. The sweat glands and connective tissue fibers were sparsely distributed in between papillary and reticular layer in Gr I and Gr II. There was no clear cut demarcation between these two layers. The papillary layer was present below the epidermis. The ground substance, connective tissue cells and reticular fibers were more in papillary layer than in reticular layer. This observation of present study corroborates with the findings noted by Gayen (1989) in buffalo and goat, Bhayani and Vyas (1991) in cattle, Wakuri (1995) in cattle, Sharma (1996) in yak, Samuelson (2007) in domestic animals and Razvi (2015) in goat. However, Mobini (2015) in sheep reported that dermis was divided into thick superficial papillary layer and thin deep reticular layer. The dermal papillae were the upward projections *of* superficial papillary layer between epidermal pegs. The dermal papillae were well developed in Gr II as compared to Gr I. In the ventral neck region dermal papillae were less developed. This finding was in agreement with Sar and Calhoun (1966) in American goat skin. The blood capillaries were more in dermal papillae than remaining portion of dermis in different body region. Similar observations were made by Akers and Denbow (2008) and Samuelson (2007) in domestic animals.

In reticular layer, fibroblasts were larger, while in papillary layer, fibroblasts were thin long, compressed and spindle shaped. This variation in shape of fibroblast could be due to seasonal variation and associated with normal physiological activity. The reticular fibers were thin and loosely present in papillary layer. The reticular fibers were present in between the collagen fibers. The reticular fibers were more at epidermo-dermal junction, around the blood vessels, sweat gland, hair follicle, sebaceous gland and in between myofibers of arrector pili muscle. The reticular fibers were more in papillary layer than reticular layer (Fig 5). The reticular fibers were present below the epidermis, around the sweat gland and sebaceous gland and provided support to the skin components.

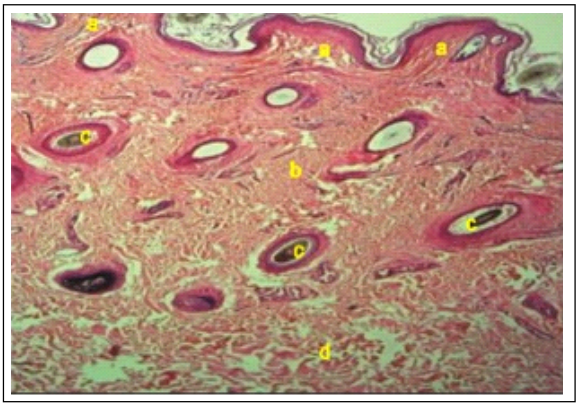


Fig 1: Photomicrograph of skin (Gr II) of goat in dorsal neck region showing. a) Dermal papillae, b) Papillary layer, c) Hair follicle, d) Reticular layer (H&E, × 50).



Fig 2: Photomicrograph of skin (Gr I) of goat in dorsal thorax region showing sweat gland associated with primary hair follicle. a) Outer root sheath, b) Inner root sheath, c) Cortex, d) Medulla, e) Connective tissue (H&E, × 400).

In the present study, the collagen fibers were more in number as compared to other fibers in dermis. It was observed that collagen fibers were densely arranged in papillary layer and parallel with the skin surface below the epidermis (Fig 3). The cross and longitudinal section of skin showed longitudinal as well as concentric arrangement of collagen fibers around sweat gland, sebaceous gland and hair follicle. The collagen fiber bundles were irregularly present in the reticular layer. This type of arrangement was present in all the body region of goat skin in Gr I as well as in Gr II, which was in accordance with that reported by Kapandis (2005). The collagen fibers were loosely arranged in Gr I as compared to Gr II. The dense layers of collagen fibers were present below the epidermis for strong attachment between epidermis and dermis. The few collagen fibers were perpendicular to it. The arrangement of collagen fiber bundles indicated the direction of extensibility of skin.

The elastic fibers were present in between the collagen fibers around the blood vessels and hair follicles (Fig 4). The sweat and sebaceous gland were found covered by thin elastic fibers and interposed between myofibrils of arrector pili muscle. The elastic fibers were more in the papillary layer than reticular layer. This observation was in agreement with the findings reported by Mugale (2001) in skin of Deoni cattle and Gayen (1989) in buffalo. In contrary with the findings of the present study, Meyer (1981) reported that elastic fibers were more in reticular layer than papillary layer in pig. This variation may be attributed to the variation in species.

During the present study, elastic fibers were more in Gr I as compared to Gr II. The distribution of elastic fibers was more in neck and thorax region as compared to abdomen. This finding of presence of more elastic fibers in skin of young animals may be correlated with more physical activity during young age than that of adult age.

The elastic fibers were very thin and present below the layer of epidermis in papillary layer. These fibers were more in the middle to lower part of papillary layer. The age and region did not show any significant difference in number of elastic fibers. In agreement with the findings of the present study, Bhayani and Vyas (1991) reported similar observations in Gir cattle.

The presence of elastic fibers around the sweat gland, sebaceous gland and hair follicles might be correlated to provide elasticity for the enlargement of hair follicle during its growth and for distension of the glands caused by accumulation of their secretory contents. The elastic fibers at the end of arrector pili muscle might be necessary for the relaxation of muscle for contraction.

During the present study, it was observed that large blood vessels were located at the junction of dermis and hypodermis. The blood vessels were parallel to skin surface. The arterioles and veins were observed in dermis. The small blood capillaries were present in papillary layer and formed network below the epidermis in all body region in Gr I and Gr II. This rich vascular bed observed at the junction of dermis and hypodermis might be necessary to contribute for the effective thermoregulation as well as for acting as source for supply of nutrients to the cutaneous glands for their secretory activity. The arteriovenous shunt was found surrounded by capsule of connective tissue consisting of smooth muscle cells. The glomus bodies were oval structures present in all skin regions. The thin collagen fibers were present in between the vessels of glomus body.

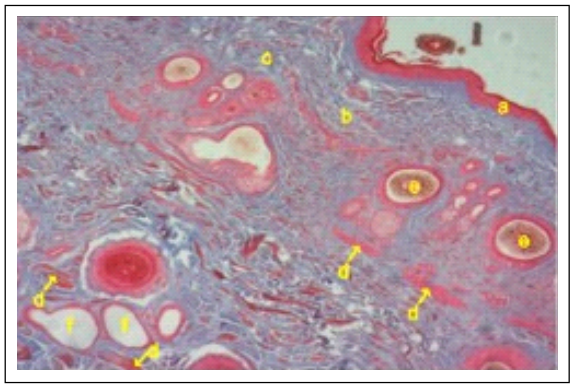


Fig 3: Photomicrograph of skin (Gr I) of goat in lateral neck region showing. a) Epidermis, b) Papillary layer, c) Collagen fibers, d) Arrector pili muscle, e) Hair follicle, f) Sweat gland (Masson's trichrome stain, × 100).

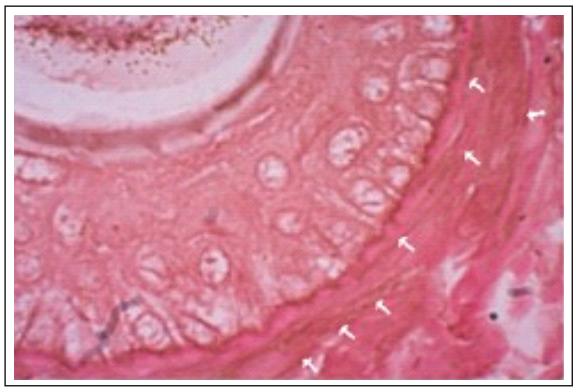


Fig 4: Photomicrograph of skin (Gr II) of goat in lateral neck region arrow showing elastic fibers in connective tissue sheath of hair follicle (Verhoeff's stain, × 1000).

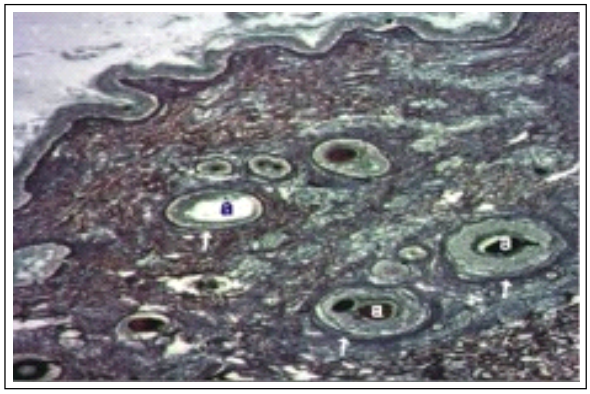


Fig 5: Photomicrograph of skin (Gr I) of goat in ventral thorax region arrow showing reticular fibers in papillary and reticular layers (Gomori's silver stain, × 100).

The more number of glomus bodies were present in the dorsal part of neck, thorax and abdomen as compared to lateral and ventral region. The presence of more number of glomus bodies in the above region may be correlated to its direct exposure to environment. The small blood vessels were present in the connective tissue sheath of hair follicle.

The glomus body has to shunt blood away from the skin surface, when exposed to cold temperature, thus preventing heat loss and allowing maximum heat flow to the skin in warm weather to allow heat to dissipate. The glomus body has high sympathetic tone and potentiation that leads to near complete vasoconstriction.

In the present study, all the body regions, showed that arrector pili muscle were associated with hair follicles, sweat gland and sebaceous gland. This muscle was found arranged obliquely in dermis. The bundles of smooth muscles were more in lower part than superficial part of papillary layer and it was also present in reticular layer. In between the arrector pili muscles, collagen fiber bundles were present. The presence of arrector pili muscle in between the connective tissue sheath of hair follicle might be useful in hair erection due to its movement. These observations of the present study are in accordance with that reported by Banks (1993) in domestic animals. During the present work, it was noted that the arrector pili muscle was more developed in dorsal region as compared to ventral region and it was thicker in Gr I as compared to Gr II. These observations are in accordance with the findings reported by Sar and Calhoun (1966) in American goat skin and Wakuri (1995) in horse skin. The presence of arrector pili muscle in the dorsal region of the body observed during present study might be correlated with higher integumentation activity present in the dorsal region.

The hair follicles were present in dermis in Gr I and Gr II in all body regions during present study. The more number of hair follicles were present at deep part of papillary layer and few follicles were present in reticular layer. The primary hair follicles were associated with arrector pili muscle, sweat (Debbarma *et al*., 2017 and Debbarma *et al*., 2018) and sebaceous gland. Many hair follicles were associated with two or three sebaceous gland. These observations of the present study are in accordance with the reports made by Zade (2007) in goat and sheep skin, Nagaraju (2012) in deer, cattle and goat, Mandage (2003) in sheep and More (2007) in deer. The duct of the sweat and sebaceous glands were found to open into hair follicle. The density of hair follicle was more in Gr II as compared to Gr I in neck, thorax, abdomen and thigh region.

The hair follicle was found to be composed of outer root sheath, inner root sheath, cortex and medulla (Fig 2). Hair were keratinized structure, which grew out of the epidermis and sunken into dermis called hair follicle, which were seen slanted. In dermis, mostly primary follicles were found. The cavity of the follicle contained single hair, which had three part root, body and shaft. The root is enlarged at its base called as bulb. The body was the major part of the hair and shaft lied above the epidermis outside the hair follicle. This observation of the present study is in accordance with the reports made by More (2007) in deer and Zade (2007) in goat.

Hair follicles were found surrounded by connective tissue sheath. The connective tissue fibers were arranged in an inner circular and outer longitudinal layer. The connective tissue sheath was continuous above with the papillary layer and below with the base of follicle and dermal papillae. Few melanocytes with melanin granules were observed in hair follicle. These observations were in accordance with the reports made by Bhayani and Vyas (1991) in cattle and Gaykee (2008) in pig.

Histochemistry

During the present study, the moderate PAS positive activity was noted in the papillary layer, while it was weak to moderate in reticular layer (Fig 6). The arrector pili muscle showed intense PAS positive activity, while the tunics of blood vessels in papillary layer showed moderate activity. This moderate to intense PAS positive activity in the dermis of the skin is probably indicative of higher dermal integumentary dynamism. The cells of external root sheath, cuticle of cortex and medulla above the hair bulb showed intense PAS positive activity, where as internal sheath and dermal papillae showed negative PAS activity. This absence of PAS activity is indicative of reduced carbohydrate metabolism. However, Razvi (2016) in goat reported presence of mild PAS activity in the inner root sheath, which is in contrast with the findings of present study.

The fibroblast cells of connective tissue sheath of the hair follicle showed presence of glycogen granules. Intense PAS positive activity was noticed in the outer sheath of the hair follicle. The upper third part of hair follicle showed weak PAS positive activity in both the groups *i.e*. Gr I and Gr II, whereas the cells of cuticle of cortex and the large medulla cells above the hair bulb exhibited intense PAS positive activity for the presence of glycogen. It was also noted that the large medulla cells towards the epidermis were shrinken and showed keratogenous changes. This observation might be indicative of the fact that growth of hair occurs in phased

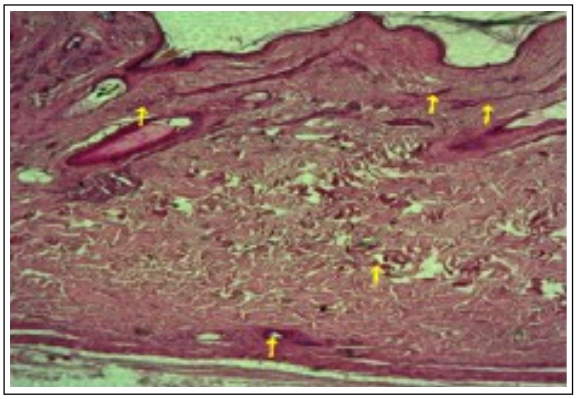


Fig 6: Photomicrograph of skin (Gr II) of goat in dorsal thorax region arrow showin PAS positive activity in papillary layer and reticular layer (PAS, × 40).

manner. These observations of the present study are supported by Ahmad (2010) in sheep.

The reticular layer of the dermis showed moderate Sudan black B activity for the presence of lipid than the papillary layer. The cuticular area of the hair follicle showed moderate Sudan black B activity. During the present study, the Sudan black B activity was found to decrease from dorsal to ventral region in both the age groups. The intense Sudan black B activity in the reticular layer might be correlated to its close proximity with the blood vessels. Similarly, decrease in the intensity of Sudan black B activity in the hair follicle from dorsal to ventral region might be due to reduction in the number of hair follicle from dorsal to ventral region due to its reduced exposure to sunlight. The moderate to intense Sudan black B activity was present in capsule and peripheral acinar cells of sebaceous gland and mild Sudan black B activity was present in secretory cells of sweat gland. This finding coincides with the observation reported by Wakuri (1995) in horse, Ahmad (2010) in sheep and Razvi (2013) in goat.

Histoemzymology

The mild acid phosphatase activity was seen in the papillary and reticular layer of dermis, while moderate acid phosphatase activity was noticed in the blood vessels. The fibroblast present in the connective tissue sheath of hair follicle showed moderate acid phosphatase activity.

The cortical part of hair follicle showed moderate alkaline phosphatase activity in both the age groups. A mild alkaline phosphatase activity was present in dermis in Gr I and Gr II. The blood vessels in the papillary layer of dermis showed mild to moderate alkaline phosphatase activity. This finding of the present study coincides with the observation reported by Baba (1990) in goat. The presence of alkaline phosphatase activity in the blood vessels of papillary layer at the epidermodermal junction is indicative of increased metabolic activity required for mitotic activity at the stratum basale.

CONCLUSION

The dermis was composed of collagen, elastic, reticular fibers, cells of connective tissue, blood vessels, nerves, hair follicles, sweat glands, sebaceous glands and arrector pili muscles in all body regions in Gr I as well as in Gr II. The collagen fibers were more in number as compared to other fibers in dermis and they were densely arranged in papillary layer and parallel with the skin surface below the epidermis. The elastic fibers were present in between the collagen fibers around the blood vessels and hair follicles. The hair follicle composed of outer root sheath, inner root sheath, cortex and medulla. The cells of external root sheath, cuticle of cortex and medulla above the hair bulb showed intense PAS positive activity. The Sudan black B activity in the dermis decreased from dorsal to ventral region of the body. The fibroblast present in the connective tissue sheath of hair follicle showed moderate acid phosphatase activity. The blood vessels in the papillary layer of dermis showed mild to moderate alkaline phosphatase activity. The present study would form a baseline data on the histomorphology, histochemistry and histoenzymology of dermis of skin of goat which would be helpful in future research prospective.

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