International Journal of Engineering, Science and Mathematics
Vol. 10 Issue 3, March 2021
ISSN: 2320-0294 Impact Factor: 6.765
Journal Homepage: http://www.ijmra.us, Email: editorijmie@gmail.com
Double-Blind Peer Reviewed Refereed Open Access International Journal - Included in the International Serial Directories Indexed & Listed at: Ulrich's Periodicals Directory ©, U.S.A., Open J-Gage as well as in Cabell's Directories of Publishing Opportunities, U.S.A

HISTOGENESIS AND AGE RELATED CHANGES IN SKIN RETICULAR FIBERS OF FETAL, NEONATAL & ADULT GOAT (*CAPRA HIRCUS*)

Tripathi M[1*], Sarkhel S[2*], Pouranik M[3*], Siddiqua A[4*] & Gupta R[5*]

***Mata Gujri Mahila Mahavidyalaya Auto. Jabalpur.**

Abstract

Reticular fibres are actually type III collagen, an un-banded form of collagen that is produced at the time of development of supporting tissues. Histological study was conducted on 10 goat fetuses ranging from CR length 20 cm to 38 cm, 10 neonates and 10 adult goats (*Capra hircus*) assembled in separate groups to investigate reticulin origin and growth. Reticular fibres originated in the fetus group stage with CR length 23 and above and form an orderly network with the growing age. These fibres are meshwork of very fine hair like fibrils and their intensity varies from sparse to dense in various parts of skin. The distribution is rather restricted; they are mainly found in the papillary layer underlying the epithelium specifically in dermal epidermal junction and surrounding the follicles & glands. The fibres are consistently found highly intense in the neck and dorsal region, gradually decreasing in thigh, flank and ventral region respectively.

Key words: Fetus, Neonate, Dermis, Reticulin, Papillary & Reticular layer.

Introduction

The development of skin and its associated structures has been explored in general in domestic animals, chick and man (Shumway,1949; Bloom and Fawcett,1982 and Arey, 1974). The epidermis of a goat skin covers approximately 1 to 2.6 % of the total thickness of the skin. Its outermost layers comprised of keratin, which strengthens the skin and melanin pigments found in the basal layer of the epidermis, responsible for skin color. Here the stratum corneum which is the first epidermal layer of skin being the primary barrier provides protection against the environment.

The skin dermal layer is 10 to 40 times thicker than epidermis and provides a kind of scaffold for strength and support. At the junction with the epidermis, its surface bristles with fibrous, vascular and nervous projections – the dermal papillae. The dermis of skin is usually described as having two structurally distinctive layers: the papillary layer and the reticular layer (Montagna, 1962). The papillary layer underlies the epidermis and is

composed of open networks of fine fibres. Below this the reticular layer, composed of densely intertwining coarse fibres, forms the main dermal anchoring. It also contains nerves, blood vessels and various glands in its connective tissue matrix that provide sensory receptors, deliver nutrients, and maintain the structural foundation of the skin.

Weinstein and Boucek (1960) and Montagna (1956) reported in the connective tissue in the dermis human skin composed of the fibrous proteins, collagen, elastin and reticulin. Collagen is predominant about 77 %, elastin 4 % and reticulin 0.4 %. In which 90% of the collagen in the body is of type I followed by type II and III and the reason for the abundance of Type I collagen is due to its wide prevalence in almost all connective tissues (Cheah, 1985).

Reticular fibres are specific types of very thin and delicately woven strands of type III collagen, these strands build a highly ordered fibrillar network and provide skeletal supporting networks in the skin. They are more resistant to boiling, acids, and alkalies than collagenous fibres, and very sensitive to their reactions to Gomori's stain. By the Gomori's method, certain membranes that were formerly considered to be homogeneous have been demonstrated to consist of a feltwork of argyrophilic fibres, e.g. the basement membrane of the epidermis and the glands, the membranelles (sheaths) of smooth muscle fibres, the sarcolemma of striated muscle, and the fat cell membrane. The gelatinous fluid ground substance which fills the intercellular spaces in embryonic connective tissue remains as a mucoid, binds the fibrils into fibres in the later growth period.

These fibres were confined to the basal lamina of the epidermis, around the hair follicles , sebaceous glands and sweat glands. Their distribution varied from sparse to dense in various parts of skin . The name of these fibres is derived from their presence in reticular connective tissue, and from the fact that they branch and anastomose, that character is not found with other fibres. They are usually very fine hairy fibres form no bundles, but lattice-like networks.

Reticulin forms an extensive supportive network in certain regions of skin and consistently found highly intense in the neck and dorsal region, gradually decreasing in thigh, flank and ventral region respectively. In light microscopy, these fibres are not visible in initial prenatal stages under 20 CR length.The reticulin fibres are incorporated in the finer structure of many other organs. They are joined by the collagenous fibres, with which they may be continuous (Mahesh, 2014). The arrangement of these fibres are important for understanding the functional role of the fibres in the skin.

Material and Methods

Pre and postnatal histogenesis histomorphological studies of skin reticulin fibres were conducted in foetal goats measuring from 20 cm to 45 cm crown-rump (CR) length, neonatal (birth to 3 months of age) and in adult indigenous goats. They are arranged in groups - Group I – 10 fetuses (CR length from 20 cm to 38 cm), Group II - 10 Neonates and Group III - 10 adult indigenous goats.

Female genitalia of healthy indigenous, nondescript goats irrespective of breed, age and nutritional status were procured from the slaughterhouse, City Corporation, Jabalpur. The material was immediately transported from the abattoir to the Department of Zoology, College of Mata Gujri Mahila Mahavidyalaya, Jabalpur in an icebox to avoid postmortem changes. The female genitalia was cleared off the fat and fascial attachments and examined for gravid and non gravid horns of the uterus.

Approach to exteriorize the conceptus was made by caudo rostral incision on the mid-dorsal surface of the gravid horn by working carefully in between the opposed surfaces of chorion allantois and amnion. The fetuses were fixed harvested, examined for their normal appearance. They were fixed in 10 percent buffered formalin and were taken out after 48 hours of fixation, blotted to dry and the following measurement of Crown-Rump (CR) length- Distance from the crown (median frontal eminence) of the head to the rump (base of the coccyx) of the fetuses was recorded with the help of vernier caliper, meter tape and thread.

The skin samples ranging approximately from 4 mm to 8 mm in diameter from different regions of the body of foetal, neonatal and adult goat-neck (dorsolateral part), dorsal region (paramedian part of the thoraco-lumbar junction), thigh (midlateral), ventral region (paramedian part of the epigastric region) and flank (midflank) were taken with the help of razor blade, scissors and forceps. The tissue pieces were properly labeled for group, animal-number and region of the body. The respective samples/tissue pieces were immediately fixed in 10 percent buffered formalin and processed further as per standard techniques (Lillie and Fullmer, 1976; Drury and Wallington, 1980). Reticulin staining has been done by Gomori's (1937) reticulin method.

Result And Discussion

Group I fetuses (Prenatal stage - CR length 20 cm to 38 cm)

Reticular fibres started originating at 23cm CR length as fine hair like fibrous structures and gradually creating meshwork of very fine, dark fibrils growing incessant over thin sheets of collagen fibres. Initially these fibres appeared in a form of fine hair like fibres in dermal-epithelial junctions and around hair follicles (Figure 1.A, 1.B, 1.D and 1.E). Because of its argyrophilic nature they stained black with silver stain. They outlined as delicate network of fine fibrils underlined the epithelial layer adjoining the dermal papilla, started giving coverage to the hair follicles and with the further growth appeared around other skin glands such as sebaceous and sweat glands in late fetuses of group I CR length 30cm (Figure 1.C). Similar observation was given in the results of Ushiki (2002) in human skin. Razvi *et al.* (2015) reported in histological studies of skin in gaddi sheep foetus, reticulin origin in the second month of age before collagen origin differs with the present investigation. Similarly Kumar *et al.* (2017) workred on *Capra hircus* and observed the subepithelial connective tissue was cellular in the early stage in which reticular fibres first appeared at 40 days of gestation and formed network at 44 days of gestation but no collagen or elastic fibres were seen not coinciding with the present findings.

The intensity of these fibres as reported was general in all samples that reticulin assimilation was originating in neck and thigh region (Figure 1.A and 1.C) with intense, moderate in dorsal and flank (Figure 1.B and 1.E) with moderate and weak in ventral region of the skin (Figure 1.D) (Table 1).

As reported that hair follicular rows i,e superficial, middle and deep observed in the late group III samples (CR length upto 41 cm) follicles of middle layer exceeded in number and were of variable size though most of the follicles found intermediate size and were distributed either randomly or in groups of 2 to 4 with majority in groups of 3 happened to be oriented linearly, usually parallel to the free surface of the skin. However, occasionally these were arranged radially and obliquely. They are sparsely surrounded by a thin layer of reticulin following elastin outside. Collagen fascicles actually form the complete supporting base to all the skin components. The superficial layer of follicles placed in subepithelial position, consisted predominantly of small follicles bordered with reticulin which were flanked by the sebaceous glands encircled by reticulin on either side and were distributed randomly at place in the groups of three (Figure Fig 2. A-E). The elastin fibres amalgamated here in follicles and sebaceous glands layering outside of the reticular layer.

Group II (Neonate)

Reticulin fibres build the good supporting system of skin, gives skeletal support to connective line of epidermis with the dermal layer (Figure 3.A) along with hair follicles and arrector pili (Figure 3.A and 3.B) and all glands (Figure 3.C, 3.D and 3.E) found there in the skin.

As clear in fetal stages reticulin fibres consistently found intense in the neck and dorsal region (Figure 4.A and 4. B) gradually decreases moderate in thigh, flank and ventral region respectively (Figure 4.C, 4.D and 4.E) (Table 2).

In silver impregnated specimens, reticulin fibrils are densely coated with coarse metal particles, probably due to the high content of glycoprotein around the fibrils. These findings suggest in Gomori's reticulin method that the surface of the individual reticulin fibres was showing argyrophilic characteristics which become visible in specific places. Compactness of reticulin fibres significantly increased in the neonate group.

They lead to condensed distribution of fibril in specific areas in neonate skin. It's hairy fibril structure interlacing within themselves and arranged to fabricate the regular architecture for supporting basal layer and other skin components. The emerging reticulin fibres forming prominent hairy meshwork in dorsal sub-epithelial papillary region (Figure 3.A), where as in ventral region the fibres indicating comparatively less intense and very fine growth of light fibrils in papillary region (Figure 3.D).

These fibres are composed of randomly oriented collagen III fibrils lying in an amorphous dermal matrix. The arrangement of reticular fibres is important for its functional role in skin. They are not oriented in orderly form hence characterized by different arrays from other fibres. They are distinguished by their tendency to form fine meshed networks at their location (Figure 3 & 4). Reticulin fibres are shown around the hair follicle uniformly covering the follicular base right from the foetal stages . The fibres are found supporting each and every hair follicle along with its sebaceous glands in papillary and reticular region (Figure 3.A,B&E). Sparse layer of reticular fibres found ubiquitously around the sweat gland in reticular region (Figure 4).

Group III (Adult)

In adult samples reticulin builds a highly ordered fibrillar network and provides a skeletal support system. We characteristically used Gomori's reticulin stain process and corresponding results were found that the reticulin fibres were stained black and were

confined to the basal lamina of the epidermis (Figure 5.A & 5.C) around the hair follicles (Figure 5.A, 5.B & 5.D), sebaceous glands (Figure 5.E). Their distribution varied from sparse to dense.

Regional differences reported are general in all samples that reticulin assimilation was highly intense found in the neck (Figure 6.A) region of the skin. The fibres were found intense in dorsal & thigh (Figure 6.B & 6.C) region and moderate in other body regions such as ventral & flank (Figure 6.D & 6.E) (Table 3).

The arrangement of reticulin fibres as shown in light microscopy is important for understanding the functional role of the fibres in the skin. These findings show that it forms a delicate network of fine fibrils underlie the epithelial layer adjoining the dermal papilla, surrounding hair follicles and other skin glands and arrector pili muscles. The firm attachment of individual fibrils with the basal lamina shows that the fibril meshwork and the basal lamina, as a whole, form a distinct structural unit for the support of the skin. As a whole this fibre system plays a role in maintaining the skeletal support to various skin components. The result coincides with the findings of Razvi *et al.* (2015) on Bakarwal goat where he showed fine reticular fibres around sweat, sebaceous and hair follicles.

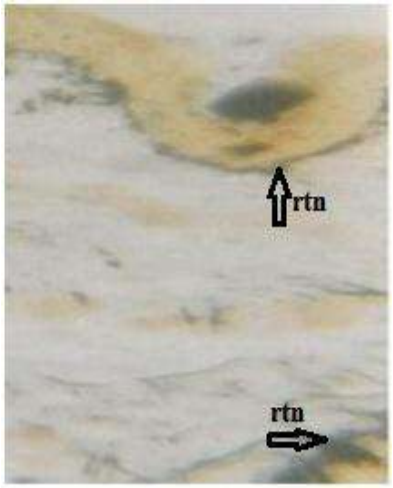
Reticulin fibres thus differ in structure arrangement and function from other fibrils, but many places are continuous with collagen fibres. In this scene, these two fibrous components are considered to form an extensive network of collagen fibrils as the collagen fibrillar system.

S. No.	Neck	Dorsal	Thigh	Ventral	Flank
1	+++	++ to +	+++	++ to +	++
2	+++	++	+++	+	++ to+
3	++	+	++	+	++
4	+++	++	++ to +	+	++
5	+++	+	++	+	+
6	++	++	+++	+	+
7	+++	++	++	+	+
8	++	++	+++	+	++
9	+++	++	+++	++ to +	++
10	+++	++ to +	++ to +	+	++ to +

++++ Highly intense, +++ Intense, ++ Moderate, + Weak, - Inconspicuous

Table 1: Histological analysis of cutaneous reticulin in foetuses goat of group I

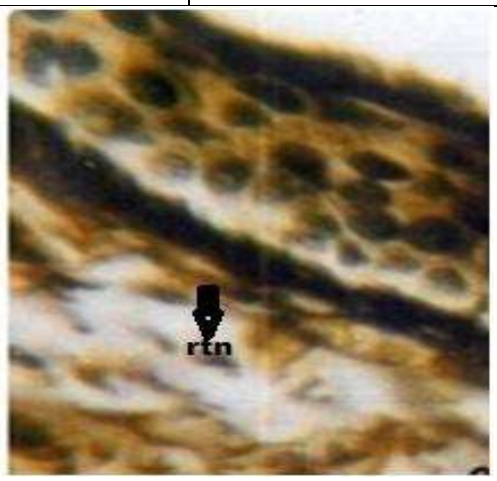
A.



B.

C.

D.



E.

Figure 1. (A-E): Micrograph of cross section of skin of foetal goat (group III), Gomori's method: (CR length 21 to 30 cm)

A. Neck region (CR length 25 cm), x 100 showing reticulin fibres underlie the epithelial layer adjoining the dermal papilla.

B. Dorsal region (CR length 23 cm), x 400 showing reticulin fibres underlie the epithelial layer adjoining the dermal papilla, surrounding hair follicles.

C Thigh region (CR length 30 cm), x 100 showing reticulin appeared around hair follicles, other skin glands such as sebaceous and sweat glands

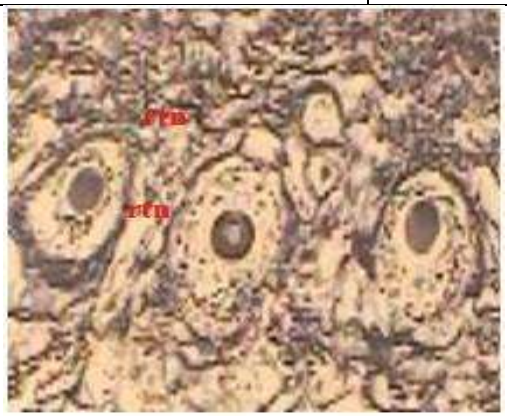
D. Ventral region (CR length 23 cm), x 400 showing reticulin fibres surrounding hair follicles.

E. Flank region (CR length 25 cm), x 100 showing reticulin fibres underlie the epithelial layer adjoining the dermal region..

A.

B.

C.



D.

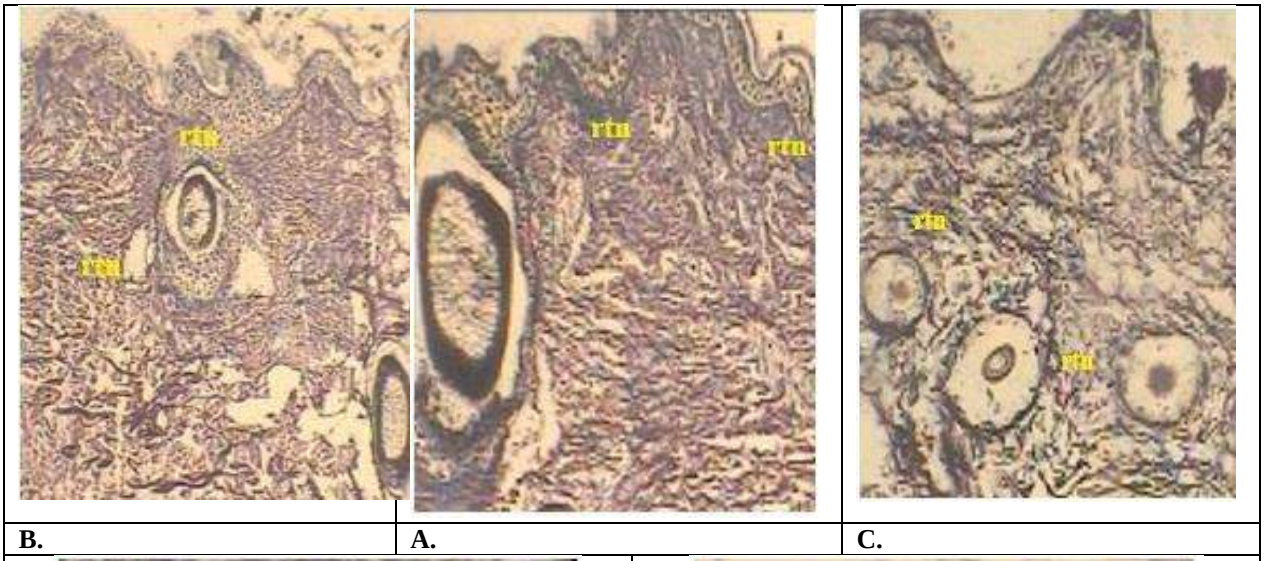
E.

Figure 2. (A-E): Micrograph of cross section of skin of foetal goat (group III), Gomori's method: (CR length from 31 cm and above)
A. Neck region, x 100 showing intense reticulin fibre.
B. Dorsal region, x 100 showing moderate reticulin fibre.
C. Thigh region, x 100 showing intense reticulin fibre.
D. Ventral region, x 100 showing weak reticulin fibre.
E. Flank region, x 100 showing moderate reticulin fibre.

S.No.	Neck	Dorsal	Thigh	Ventral	Flank
1	+++	+++	++	++	++
2	+++	+++	++	++	++
3	++	+++	++	++	++
4	+++	++	++	++	++
5	+++	+++	++	++ to +	++
6	++	++	++	++	++
7	+++	+++	++	++	++
8	+++	++	++	++	++
9	++	+++	++ to +	++	++
10	+++	+++	++	++	++ to +

++++ Highly intense, +++ Intense, ++ Moderate, + Weak, - Inconspicuous

Table 2: Histological analysis of cutaneous reticulin in neonatal goat



B.	A.	C.
D.		E.

Figure 3(A-E): Micrograph of cross section of skin of neonatal goat, Gomri's method:

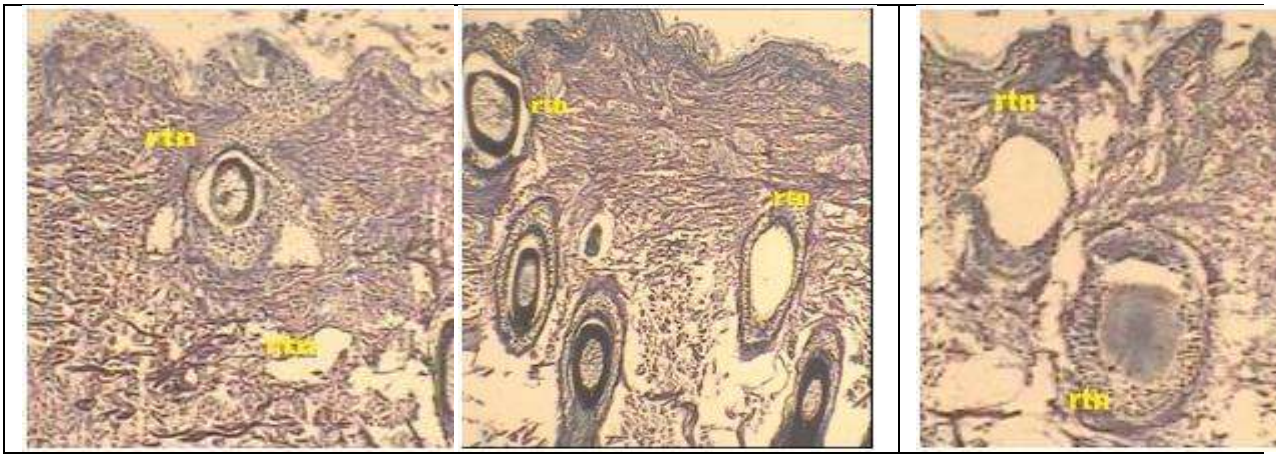
A. Neck region, x100 showing reticulin fibres underlie the epithelial layer adjoining the dermal papilla, surrounding hair follicles and arrector pili .

B. Dorsal region, x 100 showing reticulin fibres surrounding hair follicles.

C. Thigh region, x100 showing reticulin fibres presence near arrector pili

D. Ventral region, x 100 showing reticulin fibres surrounding sebaceous glands.

E. Flank region, x 100 showing reticulin fibres surrounding sweat glands.



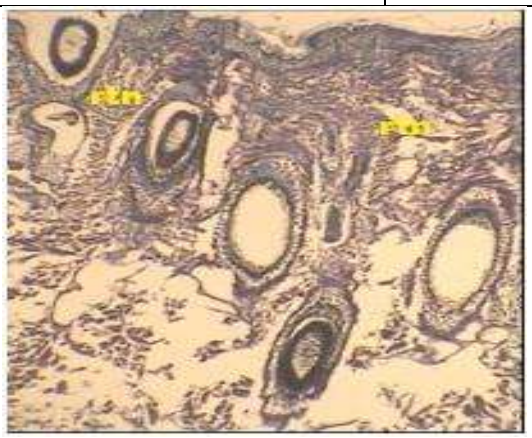
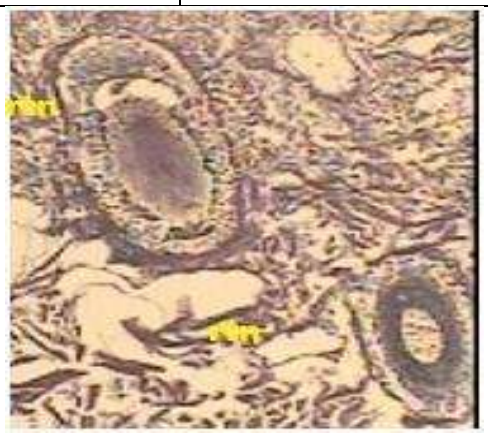
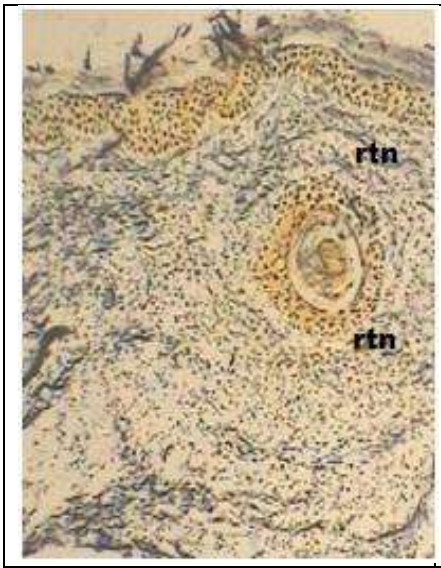
A.	B.	C.
		
D.	**E.**	

Figure 4. (A-E): Micrograph of cross section of skin of neonatal goat, Gomori's method:
A. Neck region, x 100 showing intense reticulin fibres.
B. Dorsal region, x 100 showing intense reticulin fibres.
C. Thigh region, x 100 showing moderate reticulin fibres.
D. Ventral region, x 100 showing moderate reticulin fibres.
E. Flank region, x 100 showing moderate reticulin fibres.

S. No.	Neck	Dorsal	Thigh	Ventral	Flank
1	++++	+++	+++	++	++
2	++++	++	+++	++	++ to+
3	++	+++	++	++	++
4	++++	++	+++	++	++
5	+++	+++	++	++ to +	++
6	++++	++	++	++	++
7	++++	+++	+++	++	++
8	++++	+++	+++	++	++
9	++	+++	+++	++ to +	++
10	++++	+++	++	++	++ to +

++++ Highly intense, +++ Intense, ++ Moderate, + Weak, - Inconspicuous

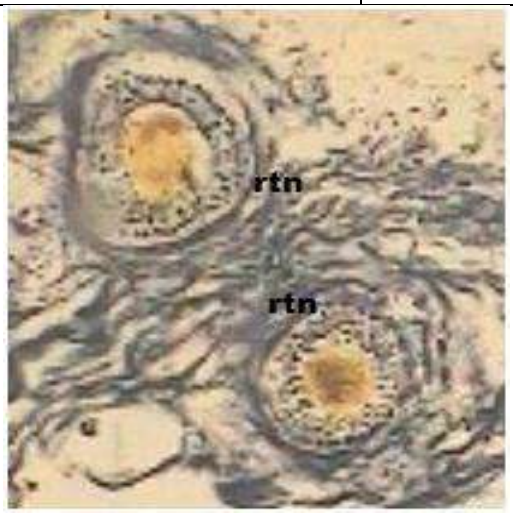
Table 3 : Histological analysis of cutaneous reticulin in adult goat



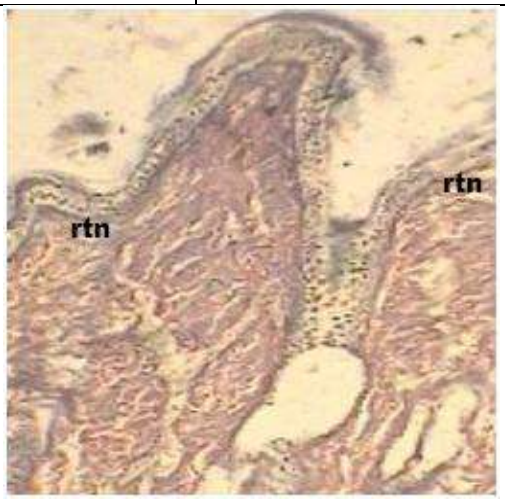
A.

B.

C.

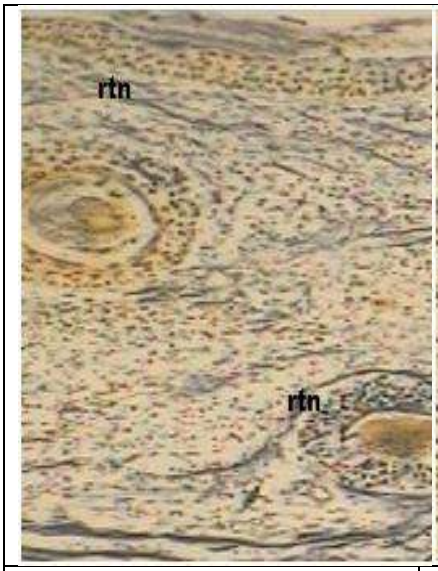


D.



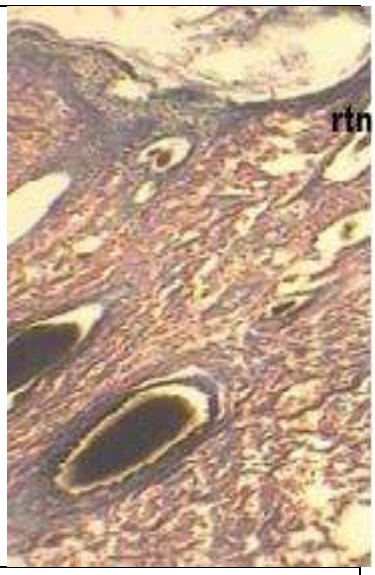
E.

Figure 5. (A-E): Micrograph of cross section of skin of adult goat, Gomri's method:
A. Neck region, x 100 showing showing reticulin fibres present in around hair follicles.
B. Dorsal region, x 100 showing hair follicles surrounded by reticulin fibre.
C. Thigh region, x 100 reticulin fibre present in basal lamina of the epidermis.
D. Ventral region, x 100 showing present of reticulin fibres near hair follicle.
E. Flank region, x 100 showing reticulin fibre present in basal lamina of the epidermis.

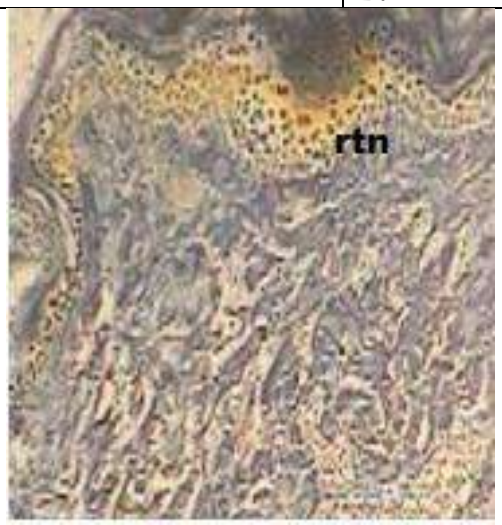


A.

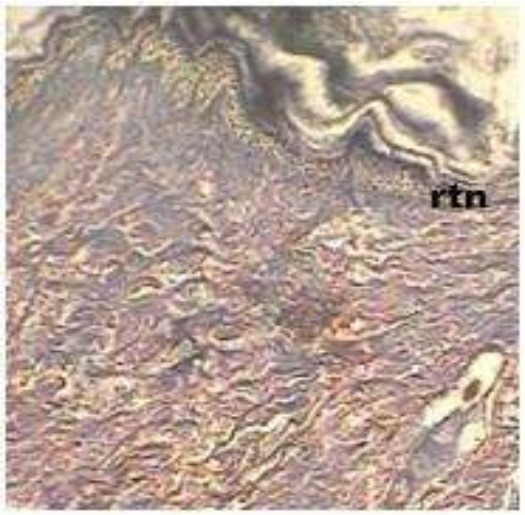
B.



C.



D.



E.

Figure 6. (A-E): Micrograph of cross section of skin of adult goat, Gomri's method:
A. Cross section of skin of adult goat, neck, Gomri's method, x 100 showing highest intense reticulin.
B. Cross section of skin of adult goat, dorsal, Gomri's method, x 100 showing intense reticulin.
C. Cross section of skin of adult goat, thigh, Gomri's method, x 100 showing intense reticulin.
D. Cross section of skin of adult goat, ventral, Gomri's method, x 100 showing moderate reticulin.
E. Cross section of skin of adult goat, flank, Gomri's method, x 100 showing moderate reticulin.

References

1. Arey, L.B. (1974) *Developmental anatomy: A text book and laboratory manual of embryology*.7th ed. W.B. Saunders Company, Philadelphia and London, pp. 439-453.
2. Bloom, W. and Fawcett, D.W. (1982) *A textbook of histology*. 10th ed. W.B. Saunders Company, Philadelphia, London, Toranto, Igakcu – Shion/Sounders Tokyo, 563-597.
3. Cheah KSE. (1985) Collagen genes and inherited connective tissue disease. Biochem. J. 229, 287-303.
4. Dellmann, H.D. and Brown, E.M. (1987) Textbook of veterinary histology." 3rd ed. lea and febiger, Philadelphia, 382-415.
5. Drury, R.A.B. and Wallington, E.A. (1980) *Carleton's histological technique*.5th ed. Oxford University Press.*Oxford New York*, Toronto pp.519-529.
6. Gomori's, G. (1937) Silver impregnation of reticulum in paraffin sections. *American Journal of Clinical Pathology*, **13**:993-1002.
7. Kumar, P., Prakash, A., Farooqui, M.M., Pathak, A., Singh, S. P. and Gupta, V. (2017) Histogenesis of skin in early prenatal goat *(Capra hircus)*. *Journal of Animal Research*, **7**(2): 377-384.
8. Lillie, R.D. and Fullmer, H.M. (1976) *Histopathologic technique and practical histochemistry*.4th ed. MnGraw-hill Book Company, *a* blakiston publication. 942.
9. Mahesh, R., Singh, G. and Kumar, P. (2014) Light and scanning electron microscopic studies on the rumen of goat *(Capra hircus).Veterinary Research International*, **2**(3): 74-80.
10. Montagna, W. (1956) *The Structure and Function of Skin*. Academic Press Inc, New York, 337.
11. Montagna, W. (1962) *The structure and function of skin*. 2nded. New York: Academic Press.
12. Montes, G.S., Krisztan, R.M., Shigihara, K.M., Tokoro, R., Mourao, P.A.S. and Junqueira, L.C.U. (1980) Histochemical and morphological characterization of reticular fibres. *Histochemistry*,**65**: 131-141.
13. Razvi, R., Shukla, P., Rajput, R. andPathak, V.(2015) Histological studies on prenatal skin of developing gaddi sheep foetus. *Journal of Cell and Tissue Research*,**15**(3) 5329-5334.

14. Razvi, R., Suri, S., Sarma, K. and Sharma R. (2015) Histomorphological and histochemical studies on the different layers of skin of Bakerwali goat. *Journal of Applied Animal Research*,**43**(2): 208–213.
15. Shumway, W. (1949) Introduction to vertebrate embryology 4th ed. john wiley and sons INC New York, Champan and Hall Limited London, 213-216.
16. Ushiki, T. (2002) Collagen fibers, reticular fibers and elastic fibers. a comprehensive understanding from a morphological viewpoint. *Arch. Histol. Cytol*,**65**(2):109-26.
17. Weinstein, G.D. and Boucek, R.J. (1960) Collagen and elastin of human dermis. *Journal of Investigative Dermatology*, **35**: 227-229.