

HISTOLOGICAL STUDIES ON THE RETICULUM OF THE SHEEP (OVIS ARIES)

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ABSTRACT

The present work was conducted to study the histopathological architecture of reticulum of 10 young sheep of 6-9 months of age. The reticulum was lined by stratified squamous keratinized epithelium comprising of strata basale, spinosum, granulosum and corneum. The lamina propria mucosae had a mixed distribution of collagen, reticular and elastic fibres. The lamina muscularis mucosae was present and extended towards the tip of the papillae of the primary fold, however, it was interrupted in the reticular lip. A thicker inner circular and a thinner outer longitudinal muscle layer constituted the tunica muscularis. The tunica serosa was present.

Key words: Histology, reticulum, sheep

Reticulum, the smallest among the four compartments of stomach, is situated most cranially just behind the diaphragm and more towards the left side of the median plane. Literature is available on the anatomy and histology of stomach of buffalo (Sengar and Singh, 1970) and goat (Chungath *et al.*, 1985). An attempt has been made to study the detailed histological structure of the reticulum of sheep.

MATERIALS AND METHODS

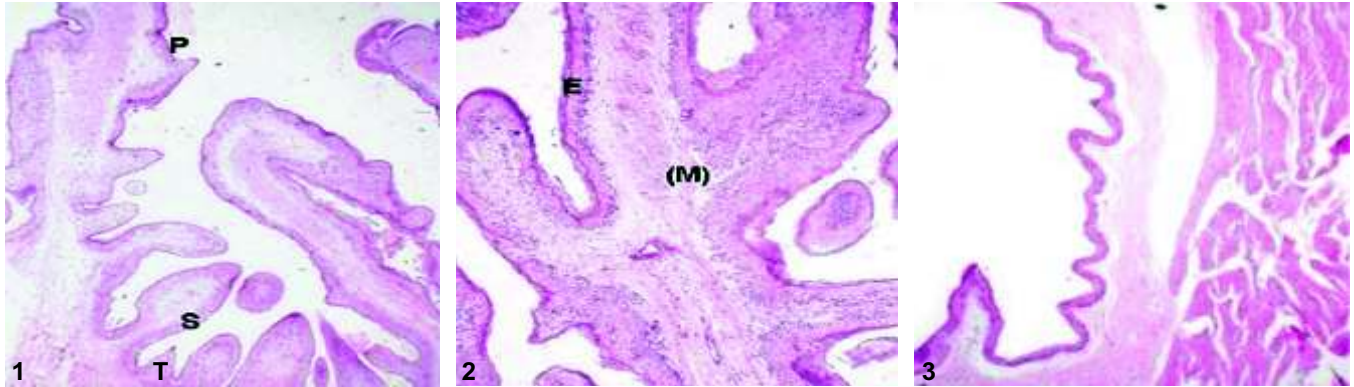
The study was conducted on ten young sheep (6-9 months age) of either sex of local breed. The stomach along with a small portion of oesophagus and duodenum were procured from the local slaughter house immediately after dressing. A total of three tissues (reticular cell small, reticular cell large and reticular groove with lip) were collected from reticulum of each animal. The tissues were fixed in 10% neutral buffered formalin and processed for routine paraffin technique of light microscopy. The paraffin sections of 5-6 μ were cut and stained with routine Harris' hematoxylin and eosin stained for general histoarchitecture, Gomori's method for reticular fibres, Weigert's method for elastic fibres, Ayoub-Shklar method for keratin and pre-keratin (Luna, 1968) and Crossman's trichrome stain for

collagen fibres (Crossman, 1937). Glycogen was demonstrated by McManus' method whereas, mucopolysaccharides were demonstrated by PAS-Alcian blue method and Alcian blue method (pH 2.5) (Luna, 1968).

RESULTS AND DISCUSSION

The reticulum presented honeycomb like appearance having small and large reticular cells. The small and large reticular cells presented primary, secondary and tertiary folds which were lined by stratified squamous keratinized epithelium (Figs. 1, 2). The primary folds were the largest whereas the tertiary folds were the smallest in dimensions. The reticular groove presented undulating surface because of the presence of small sized folds and few small sized papillae with blunt tips (Fig. 3). The reticular lip presented folds which were comparable to primary folds of the large and small reticular cells. These had varying shape and size because of uneven surface of folded mucosa (Fig. 4). Samuelson (2007) reported varied size papillae, which arose between the crests as well as extended from them. Mahesh (2008) observed that the reticular groove had uniform stratified squamous keratinized epithelium due to the absence of projections in the form of papillae whereas in the reticular lip, the epithelium had uneven surface of

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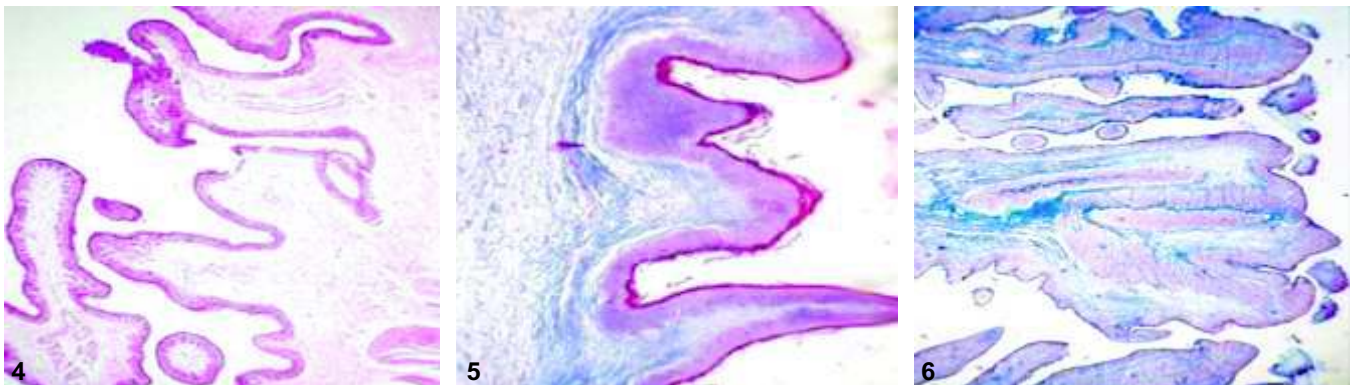


Figs 1-3. 1. Photomicrograph of small reticular cell showing primary fold (P) and secondary (S) and tertiary (T) papillae. (H. & E. x 40). 2. Photomicrograph of small reticular cell showing the stratified squamous keratinized epithelium (E) and lamina muscularis mucosae (M). (H. & E. x 100). 3. Photomicrograph showing the small sized papillae giving uneven surface appearance to the reticular groove. (H. & E. x 40)

folded mucosa projected to form small papillae with blunt tips.

The stratified squamous keratinized epithelium was comprised of the different cell layers (Figs. 1-4). The cells of the stratum basale had round to oval nuclei. The chromatin material was aggregated into smaller clumps irregularly throughout the nucleoplasm. One to two nucleoli were centric or eccentric in position. The cytoplasm of these cells was finely granular and eosinophilic. The rest of the layers i.e. stratum spinosum and stratum granulosum showed varying number of rows of nuclei at varying places in these folds. The nuclei of the stratum spinosum were round to oval shaped and were larger than those of stratum basale. The chromatin material was distributed evenly throughout the nucleoplasm, except a few

nuclei close of stratum basale where condensation of chromatin material was observed in smaller clumps. Some of the nuclei were very lightly stained whereas a few others were having dense nuclei masking the presence of nucleoli. Generally one to two nucleoli were centric or eccentric in position. A few vacuolated areas surrounding the nuclei were also present. The cytoplasm was finely granular and eosinophilic. The cytoplasmic processes of the cells intermingled with those of adjacent ones. The nuclei of stratum granulosum were also round to oval with few irregular surfaces. The chromatin material was distributed uniformly except a few where smaller clumps were observed towards the centre. Most of the nuclei were single and darkly stained with centric or eccentric nucleoli. A few large vacuolated areas surrounding the



Figs 4-6. 4. Photomicrograph of reticular lip showing large sized papillae along with a few small sized papillae. (H. & E. x 40). 5. Photomicrograph showing the well defined keratin layer in the stratified squamous keratinized epithelium in the reticular fold of large reticular cell. (Ayoub Shklar stain x 100). 6. Photomicrograph showing distribution of collagen fibres in the lamina propria mucosae of the small reticular cell. (Crossman's trichome stain x 40)

nuclei were also observed. A few cells had cytoplasm which was granular and eosinophilic. The stratum corneum was constituted by 2-3 cell layers having elongated darkly stained nuclei with more eosinophilic cytoplasm.

The secondary and tertiary folds were of varying shapes with uneven surfaces and dimensions. The cellular features of different cell layers were almost same as in the primary folds. The reticular epithelium demonstrated well defined keratinization (Fig. 5). At reticular lip, keratin layer was thick and disrupted. Earlier, Tiwari and Jamdar (1969) demonstrated iron pigment in the keratinized layer of epithelium of reticulum in 4 weeks old buffalo calves.

The lamina propria mucosae had loose irregular connective tissue alongwith collagen, reticular and elastic fibres, few connective tissue cells, small blood vessels and fine blood capillaries (Fig. 6). These connective tissue fibres were oriented parallel to the longitudinal axis of primary fold whereas at the base the connective tissue merged with that of the submucosa and was horizontally placed. The connective tissue was very less in secondary and tertiary folds. The lamina muscularis mucosae was seen in the form of small bundles of smooth muscles towards the apical portion of primary fold, only a few smooth muscle fibres in some of the secondary folds and was absent in tertiary folds (Figs. 1, 2). The primary crests had a lamina muscularis mucosae upto apical portion which when contracted reduced the heights of these crests (Samuelson, 2007). It was suggested that contraction of these smooth muscles reduced the epithelial height, facilitated the movement of blood away from the primary folds after its vessels had become filled during digestion, absorption of volatile fatty acids, ammonia, sodium, potassium, etc. The lamina propria mucosae merged with the submucosa forming propria submucosa in secondary crests/folds and tertiary papillae as reported earlier (Samuelson, 2007). The reticular groove had the lamina muscularis mucosae which was interrupted and separated the lamina propria mucosae and tunica submucosa in the reticular lip. It was observed as small bundles of smooth muscles at few places in the reticular groove.

Eurell and Frappier (2006) reported the presence of a condensed connective tissue layer in place of lamina muscularis mucosae in the reticulum. However, Sengar and Singh (1970), Tiwari and Jamdar (1970), Ramakrishna and Tiwari (1979) and Chungath *et al.* (1985) while indicating the absence of lamina muscularis mucosae had not mentioned the condensed connective tissue layer and had described the propria submucosa together without any distinct line of demarcation between the two. However, Taluja and Saigal (1987) reported an interrupted lamina muscularis mucosae in the base of the fold in buffaloes over 1½ years of age.

The epithelium lining both the surfaces of primary folds was the same as that of secondary and tertiary folds. Ayoub Shklar method demonstrated the presence of keratin in all types of folds (Fig. 5). The reticular fibres formed the basement membrane. The loose connective tissue of lamina propria mucosa was very less. The contents of propria and submucosa were similar to earlier descriptions except that in the primary folds the elastic fibres were present above and below the level of lamina muscularis mucosae towards the tip of the primary fold. The elastic fibres were large and vertically oriented in the core of the papillae. The bundles of the collagen fibres were present in subepithelial portion of lamina propria mucosae. These bundles surrounded the smooth muscles of the apical portion in primary fold. The connective tissue of submucosa was also loose, irregular having collagen, reticular and elastic fibres and fine blood capillaries. The connective tissue was denser towards the muscular layer.

The tunica muscularis was constituted by thicker inner circular and thinner outer longitudinal muscle layers as reported earlier by Chungath *et al.* (1985). However, Sengar and Singh (1970), Tiwari and Jamdar (1970) and Taluja and Saigal (1987) reported that the tunica muscularis of buffalo reticulum comprised of three layers. Large nerve bundles of myenteric plexus were present at the junction of these two layers during present study. A few collagen and reticular fibres were also horizontally placed in between the fasciculi of the smooth muscles. In

addition, a few fine blood capillaries and small blood vessels were also present. In reticular lip, the outer longitudinal muscle layer was absent, however, the muscle fibres were oriented in different planes of circular, oblique and longitudinal muscle at the junction of lip with reticular folds. In between these rows, nerve bundles were present. Towards free portion of the lip, the inner circular layer of both inner and outer surfaces was continuous with each other and in between the two layers a large number of small blood vessels, fine blood capillaries, fatty tissue, and nerve bundles were present along with collagen, reticular and elastic fibres.

The tunica serosa was present more superficially as a layer of flat mesothelial cells being supported by loose irregular connective tissue with large number of small sized blood vessels, fine blood capillaries, few nerve fibres and fatty tissue. This finding was in accordance with that of Sengar and Singh (1970). A mixed distribution of collagen, reticular and elastic fibres was also observed. The fine reticular fibres formed the basement membrane.

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