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Histomorphological changes in the goat, *Capra hircus* L. rumen during different stages of gestation

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Understanding the fetal development is essential to treat different anomalies of livestock after birth. Biological and anatomical data on the developing rumen of goat is scanty. Here, we investigated histomorphological changes in the ruminal part of the fore stomach of the goat, *Capra hircus* L. to elucidate the transformation of rumen prior to its functional state. For this study, 36 developing rumen were collected from healthy and normal embryos/foeti of either sex of goat from the first stage of prenatal life until birth. Tissues were processed by paraffin embedding technique and sections were stained. Definite four layers of the tubular structure were elucidated first at 51 days of gestation. We observed the ruminal wall lined by undifferentiated stratified epithelium up to 100 days which gradually became stratified squamous keratinized epithelium at 145 days of foetal life. Throughout the gestation lamina muscularis was absent. Reticular fibers evident first followed by collagen and elastic fibers. The prenatal development of the goat ruminal mucosa evidenced a considerable resemblance with the post natal rumen; however, certain morphological changes are still incomplete in order to meet the functional demands in postnatal life.

Keywords: Fore stomach, Livestock, Prenata

Goat is important in arid, semiarid and mountainous region where crop and dairy farming are not economically feasible[1]. The ability to browse and optimise the use of grazing land has been linked to the peculiar nature of the stomach of ruminants[2]. The rumen appears to be the most important component of the fore-stomach as it is involved in breakdown of feed and serves as the primary site for microbial fermentation. Literature on development and growth of stomach has been carried in other ruminants[2-6]. However, these research findings cannot easily be adapted to our local breeds due to variation in genetic makeup, climate, vegetation and feeding regimen. It is necessary to have the fair knowledge of development of ruminant stomach to understand the normal histo-differentiation and maturation of different structures in different strata of this organ which in term will be useful for better understanding of sequential changes occurred during various stages of gestation. Therefore, here, we tried to investigate the accurate day of appearance of primordium of the rumen, its differentiation and maturation.

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Materials and Methods

The developing rumen was collected from 36 healthy and normal embryos/ foeti of either sex of non- descript goat of Mathura region. An approval was obtained from animal ethic committee of U.P. Pandit Deen Dayal Upadhyaya Pashu Chikitisha Vigyan Vishwavidhalaya Evam Go-Anusandhan Sansthan (DUVASU), Mathura, Uttar Pradesh prior to the commencement of the study. For this work, the embryos/foeti were collected from the local abattoir, cases of dystokia and abortion from clinics and farms. The embryos/foeti were ranged from 32 days to near full term. The age of embryos/foeti was ascertained by using the formula

$$W^{1/3} = 0.096\ (t\text{-}30)$$

where W = body weight of foetus in grams and t = age of foetus in days[7]. Embryos/foeti were assigned into three groups *viz.*, Gr. I (0-50 days of gestation), Gr. II (51-100 days of gestation) and Gr. III (101-150 days of gestation).

Developing rumen was harvested and small pieces of tissues were cut in groups II and III while in Gr. I whole of the stomach was collected. The tissues were fixed in 10 per cent neutral buffered formalin and were processed by routine paraffin embedding technique. Six micromolar thick sections were taken and stained with hematoxylin and eosin, Wilder's reticulin stain for reticular fibers, Verhoeff's stain for elastic fibers and Mallory's triple stain for collagen fibers[8,9]. Stained slides were observed under light microscope.

Micrometric observations were done on hematoxyline eosin stained sections by Leica DM750 computerized image. The data generated by the micrometrical observations were subjected to statistical analysis[10].

Results and Discussion

Histo-differentiation of different compartments of goat stomach took place at 38 days of gestation. The wall of foetal rumen was made up of three strata i.e., epithelium, pleuripotent blastemic tissue and serosa up to 49 days of fetal age and definite four layers of the wall *viz.*, epithelium, propria-submucosa, tunica muscularis and serosa were observed first at 51 days of gestation. In contrast to the study on histology of goat foetal rumen, the thickness of each stratum was increased with advancement of gestation accept propria submucosa[11].

Before differentiation of rumen, the wall of the digestive tube was irregularly thick with a narrow lumen. It consisted of three strata *viz.*, epithelium, pleuripotent blastemic tissue and serosa (Fig. 1). Several authors also observed similar stratification at 35 days (goat), 23-29 days (sheep) and 30 days of gestation (red deer)[2,12,13]. However, these authors reported that the wall of the cavity consisted of internal epithelium and external pluripotential blastemic tissue only. During this period the epithelium was undifferentiated stratified type (Fig. 1), whereas, it was pseudostratified, non ciliated cylindrical stratified type in sheep (23-29 day)[12] and stratified type in red deer (30 day) and goat (35 day)[2,13].

The pleuripotent blastemic tissue comprised of different shapes of mesenchymal cells with ground substance, blood vessels and immature red blood cells (Fig. 1) as in sheep at 32 and red deer at 30 days of gestation[12,13]. The wall of the stomach was surrounded by a single layer of squamous cells, the mesothelium.

Tunica Mucosa

Epithelium

The wall was lined by undifferentiated stratified epithelium in groups I and II which gradually became stratified squamous keratinized epithelium at 145 days. The epithelium of rumen in all groups was divided into darkly stained basal and lightly stained superficial zones. There was significant increase in the thickness of epithelium in groups II and III. Highest growth spurt was noticed in Gr. II (Table 1).

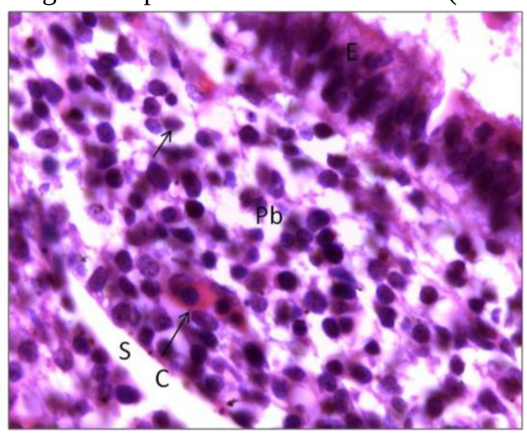

Fig. 1 — Photomicrograph of section of 32 day old goat embryo showing epithelium (E), pleuripotent blastemic tissue (Pb), serosa (S), capillary (C) and differentiating mesenchymal cell (arrow). H & E X1000

Table 1 — Micrometrical parameters (Mean ± SE) of rumen in prenatal goat in various stages of gestation

Parameters (in µm)	Groups	Group I (0-50 days)	Group II (51-100 days)	Group III (101 days-till term)
Thickness	Epithelium	71.18±9.71 (61.47-80.89)	122.27±23.51 (40.88-223.38)	153.85±26.59 (53.73-254.51)
	Lamina propria and submucosa	72.25±7.63 (55.10-86.00)	43.80±5.66 (27.67-68.15)	46.72±8.53 (30.66-79.25)
	Tunica muscularis		74.75±25.09 (12.07-211.29)	183.50± 61.89 (37.78-411.95)
	Tunica serosa	5.2	7.97±3.18 (3.48-14.14)	14.10±3.89 (9.1-25.56)
	Basal zone	-	17.42±0.79 (14.31-19.97)	11.50±1.21 (7.02-14.76)
	Superficial zone	-	88.61±16.26 (31.73-147.71)	86.75±13.66 (44.44-120.83)
Superficial cell	Height	7.36±0.58 (6.52-8.47)	12.67±1.23 (7.18-16.05)	11.51±1.31 (6.03-14.52)
	Width	4.84±0.20 (4.58-5.24)	6.64±0.89 (3.92-10.13)	6.80±0.84 (3.60-9.77)
	Diameter of nucleus	3.01±0.19 (3.76-3.57)	3.29±0.2 (2.43-4.01)	3.05±0.29 (2.18-367)
	Thickness of stratum corneum	-	-	3.7±0.029 (3.41-4)
Basal cell	Height	7.53±0.01 (7.532-7.532)	8.4±0.55 (7.00-11.18)	5.96±0.65 (3.06-7.52)
Nucleus of basal cell	Height	-	4.35±0.43 (3.33-5.50)	4.22±0.27 (3.44-4.61)
	Width/diameter	3.06±0.11 (2.75-3.29)	3.07±0.23 (2.22-3.77)	2.24 ±0.18 (1.75-2.69)
	Height of ruminal papilla	-	49.48±1.99 (40.44- 58.4)	103.51 ±46.58 (47.98-242.60)
	Width of ruminal papilla	-	13.10±1.63 (5.84-18.33)	43.75±20.69 (15.37-104.45)
	Interpapillary space	-	167.6±30.98 (85.71-288.01)	79.50 ±15.74 (34.82-104.56)

[n= 12. Figures in parenthesis indicate range, (-) could not be recorded]

Basal zone

The cells of deepest layer of basal zone were cuboidal to columnar throughout the gestation. Their nuclei were vesicular; placed at the center or towards apex and elongated or spherical shaped. The cells of other layers of basal zone were either low columnar or polyhedral in shape. The vesicular nuclei were spherical or round in shape and placed centrally. From 44 days onwards cells varied in shape from cuboidal to polygonal (Fig. 2 A-F). Contrast to present findings, in sheep reverse cytological characters was reported[12]. These authors mentioned that the cytoplasm of basal zone was lightly stained and cells were anucleated. In the present study, various mitotic figures and degeneration of few cells of basal zone were also noticed as in buffalo[14]. Vacuolations were found in infranuclear zone of few cells (Fig. 2A) as reported earlier in buffalo foetal rumen[15]. The number of layers of basal zone gradually decreased from 3-4 at 38 days to 1-2 layers at 145 days of gestation. Reduction in number of layers with advancement of age was also observed in buffalo[5,14,16]. Analysis of the data revealed that there was increase in height of basal cells in Gr. II and abrupt decrease in height in Gr. III (Table 1). The height of the cell reduced with advancement of gestation. This might be due to accommodate the different layers of rumen.

Superficial zone

The cells of this zone showed similar cytological characters in all stages of development except there was variation in number of layers. The number of cells of superficial zone increased (3-4 to 30-32 layers) from 38 to 145 days of gestation. The process of degenerative changes was enhanced and evidenced by lack of nuclei in these cells, could be spoken as the beginning of lumen formation (Fig. 2A). In early stages of gestation, most of the cells of superficial zone were polygonal in shape with distinct, highly eosinophilic cell boundaries. From 87 days onwards few of the cells of central zone became columnar in shape with a cytoplasmic process at their apical end. Condensation of cytoplasm took place resulting into formation of future stratum spinosum (Fig. 2F). The other cells of superficial zone were polyhedral in

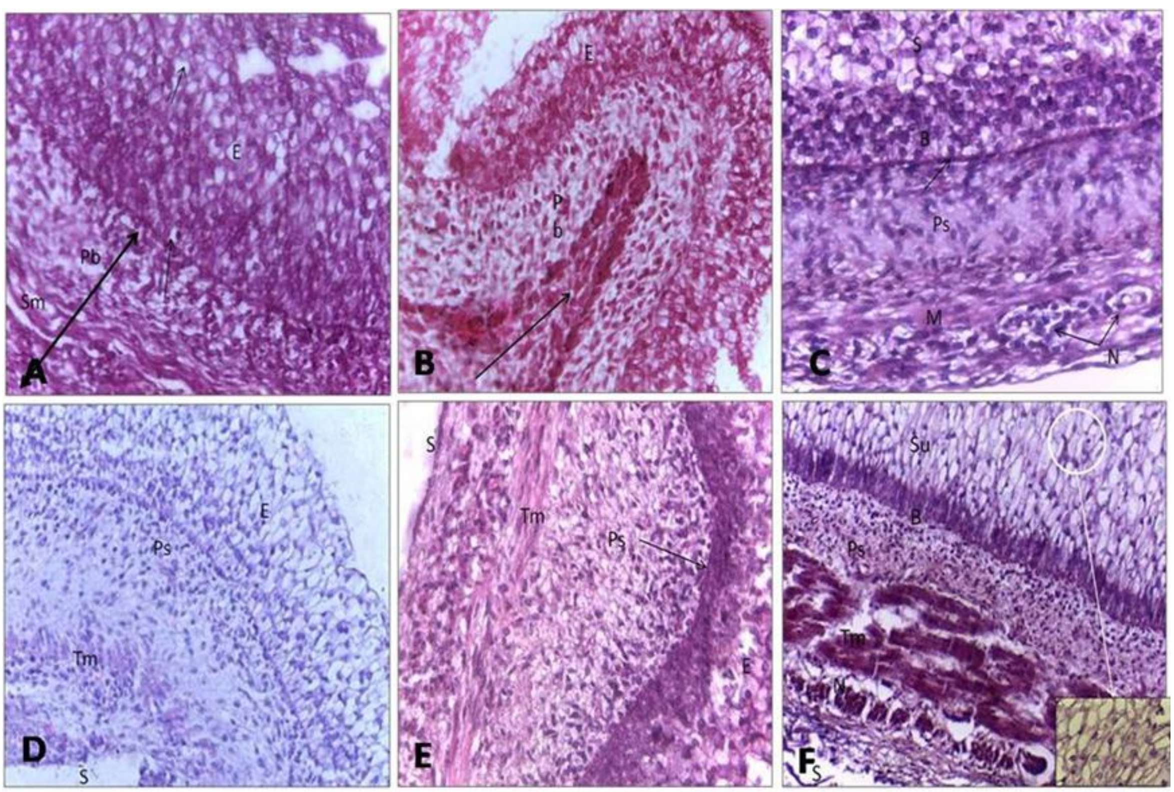

Fig. 2 — Photomicrograph of section of (A & B) 38; (C) 44; (D) 51; (E) 76; and (F) 87 days old goat foetal ruminal wall showing rumina epithelium (E), basal (B) and superficial zones (Su) of epithelium, pleuripotent blastemic tissue(Pb), differentiating myocytes (Sm), propria-submucosa (Ps), tunica muscularis (Tm) and serosa (S). Arrow showing, vacuolation in epithelial cells in A, basement membrane in C and ruminal papilla in E. [A-E- H&E X400, F- H&E X200 with higher magnification (1000X) of circled area showing formation of stratum spinosum (insight)]

shape. In contrast to the present findings, in buffalo centrally placed nuclei in epithelial cells of middle layer of superficial zone and eccentrically placed nuclei in basal layer were recorded[5]. The cytoplasm of the cells varied from lightly eosinophilic to pale. The cytological characters of the cells of superficial zone were in line with the earlier reports in buffalo and goat[2,5,14]. Distinct stratum corneum, stratum granulosum and indistinct lucidum spinosum layers were noticed in sheep foetal rumen between 53-79 days of gestation[12]. While, between 76-112 days of gestation in goat, the mucosa is comprised of strongly basophilic stratum basale, stratum granulosum, stratum spinosum and stratum corneum[2]. At the fag end of the mid gestation period (Gr. II), the superficial zone consisted of polyhedral, quadrangular and few flat cells (Fig. 3B). The flat cells were found in uppermost layer which stands similar to the observation in buffalo at 160 days of gestation[5].

Flattening and desquamation of upper layer cells were noticed in Gr. III. The progressive flattened and darkened cells of superficial epithelial layer were in the phase of being parakeratotic. Few cells of the topmost layer had lost their nuclei and had highly eosinophilic cytoplasm might be spoken as the presumptive stratum corneum (Fig. 4). In the present study future stratum corneum appeared at 145 days of gestation. keratinization was observed in at buffalo foeti of 74.0 cm CRL and at 120 days of gestation[5,14]. In the present study stratum corneum contained only 1-2 layers while in buffalo foetal rumen in keratinization, basal (2-3 cell layer), middle (1-2 layers) and apical layers (1-2 layers) were involved[16]. The stratum corneum may require some more time for its proliferation might be due to species differences or environmental or nutritional factors. The beginning of cornification indicates that the ruminal epithelium could acts as a protective layer against potentially sharp fibers consumed by an adult animal[17,18]. The presence of eccentrically placed nucleoli in different layers of rumen was suggestive of the absorptive and metabolic functions of rumen[19]. During entire gestation, eccentrically placed nuclei, process of degeneration with pyknotic nuclei and mitotic figures were constant phenomenon. Mitotic figures frequently observed in the epithelium suggested that new cells were required for the growth of epithelium. Height of the superficial cells decreased while width of the superficial cells increased with advancement of age (Table 1).

Lamina propria and Submucosa

Throughout the gestation the lamina propria and submucosa blended with each other without any line of demarcation to form propria- submucosa as lamina muscularis could not be observed. In early gestational period lamina propria, submucosa and tunica muscularis were non independent strata and were termed as pleuri-potent blastemic tissue. The pleuripotent blastemic

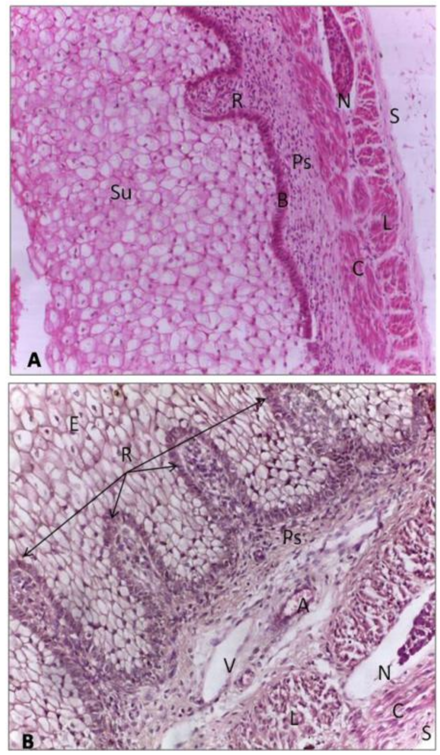

Fig. 3 — Photomicrograph of section of (A) 112; and (B) 134 days old goat foetal ruminal wall (dorsal sac) showing ruminal papillae (R), dark basal (B) and light superficial (Su) zones of epithelium (E), propria submucosa (Ps), neuronal element (N), serosa (S), inner circular (C) and outer longitudinal (L) arrangement of smooth muscle bundles of tunica muscularis, artery (A) and vein (V). [A: H & E X400 and B: H & E X200]

Fig. 4 — Photomicrograph of section of 145 day old goat foetal ruminal epithelium showing well developed ruminal papillae (R), stratum corneum (Sc) and core of the papillae (Pc). H & E X400

tissue had two clear-cut zones. The zone just below the epithelium was darker and more cellular while deeper zone was less cellular with more ground substance (Fig. 2A). On the contrary, a very thin and lightly stained lamina propria and darkly stained submucosa were noticed in buffalo[14]. Few differentiating myocytes were also noticed. These cells were fusiform shaped with elongated and fusiform vesicular nuclei which were arranged parallel to surface epithelium. Cytoplasm was more eosinophilic (Fig. 2A). Close to the serosa differentiating neuronal elements were noticed. Few mesenchymal cells also differentiated into future plasma cells. The cytological characters of these plasma cells were as identical to plasma cells of adult. These plasma cells are abundant in loose connective tissue of lamina propria of gastro intestinal tract[20,21]. Besides this, differentiating capillaries and immature RBC'S were also noticed between mesenchymal cells. Distinct basement membrane with a layer of differentiating fibroblasts was noticed just below the epithelium at 44 days of gestation (Fig. 2C). Process of proliferation of smooth muscle cells was enhanced after 48 days of gestation and clusters of 3-4 cells were noticed. These smooth muscle cells were directed obliquely, longitudinally or circularly. The differentiating tunica muscularis was well supported by capillary network and isolated reticular fibrils surrounding the clusters of smooth muscle cells appeared at 46 days of gestation (Fig. 5A). Reticular fibers in the lamina propria and submucosa near muscular coat were described at 3 month of gestation in bovine rumen[22]. These reticular fibers appeared as thin fibrils in first trimester of pregnancy in human foeti[23]. In the present study thin isolated reticular fibrils were first detected around myocytes were in partially in line with above description. This might be due to species differences. Neuronal elements showed two distinct types of cells[24]. Type I, ganglionic cells were large, spherical to ovoid in shape with an indistinct contour. Nuclear chromatin of these cells was evenly distributed and lightly stained.

Type II, were small with indistinct cell boundaries, referred as supporting cells. Nuclei of these cells were spherical in shape with darkly stained chromatin and their cytoplasm was pale. At this stage they were present in disorganized manner. The nerve tissue was recorded at 113-150 days of gestation in foetal goat[2]

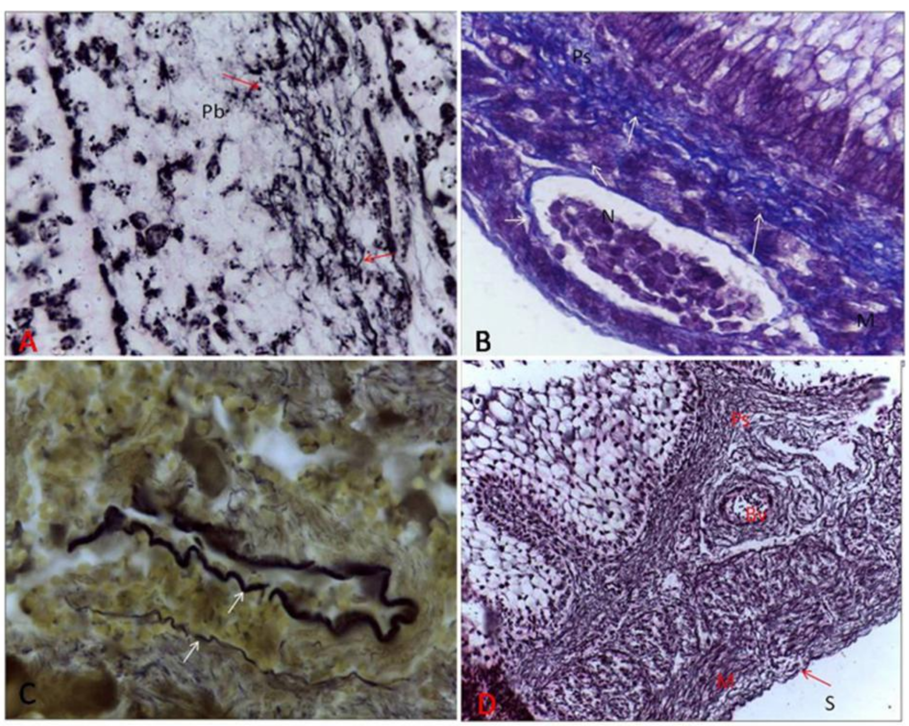

Fig. 5 — Photomicrograph of section of (A) 46; (B) 87; (C) 131; and (D) 134 day old goat foetal ruminal wall showing A- Reticular fibers in pleuripotent blastemic tissue (Pb) (arrow). Wilder's reticular stain X1000 B-Collagen fibers (arrow) in propria- submucosa (Ps), in between muscle bundles (M) and around neuronal element (N). Masson's trichrome stain X400. C- Elastic fibres (arrow) in inner and outer wall of blood vessel. Verhoeff's stain X1000; D- Reticular fibers (arrow) in the form of rete in propria- submucosa (Ps), in between muscle bundles (M), around blood vessel (Bv) and serosa (S). Wilder's reticular stain X200

and 19.6 cm CRL of buffalo foeti close to serosa[5]. Presence of nerve elements in between smooth muscle bundles is required to monitor the content of stomach and to control the smooth muscle contraction and secretion of digestive substances. Between 51-55 days of gestation, lamina propria became independent strata characterized by more cellular darkly stained with less amount of ground substance (Fig. 2D) while reverse was noticed in submucosa in foetal goat, sheep and buffalo[2,12,13]. Well developed differentiating fibroblast layer was observed just below the epithelium. Isolated reticular fibrils became coarser and their concentration was increased towards the submucosa. In later stages, rete formation of reticular fibers was noticed between 60-70 days. Fine, isolated thin reticular fibrils and immature fine isolated collagen fibrils were also found around the blood vessels from 60 days onwards.

With the advancement of age, connective tissue elements became denser and coarser with more number of fibroblasts and arranged regularly (Figs. 5 B and D). The mesenchymal cells were loosely arranged with regular interval and ground substance was lightly stained. The clusters of reticular fibers were noticed in terminal stage (134 day) in submucosa (Fig. 5D). The amount of collagen fiber was less in lamina propria, whereas coarser and wavy collagen fibers were found in submucosa particularly around blood vessels. Thin, isolated elastic fibers were noticed in blood vessels of lamina propria from 112 days of gestation onwards (Fig. 5C). The thickness of propria submucosa decreased with the advancement of age (Table 1). In the present study there was an increase in number of layer of epithelium and thickness of tunica muscularis, this might be causing the decrease in the thickness of propria submucosa. Further studies are suggested to elucidate the possible reasons for above findings. In partial harmony with present observation loosely arranged elastic and collagen fibers along with blood vessels, submucosa and around nerves in serosa were reported in goat, buffalo and sheep[2,5,12].

Ruminal papillae

At 51 days of gestation at few places condensation of cells of stratum basale and oscillation of the superficial layer of epithelium was noticed. This envagination was towards the future lumen, the beginning of formation of ruminal papillae. In the present study well defined papilla was first observed in 76 days old foetal rumen (Fig. 6). These papillae were crecenteric, cylindrical and tongue shaped at 76, 112 and 145 days of gestation. Similar observations have also been noticed in sheep at 61 and 103 days[12,25]; in goat at 126 days[16] ; in Dama dama from 85 days of gestation[15]; in bovine just before term[7,22] and 120 days[6] of gestation and in buffalo at 170 mm CRL[26]. The early appearance of the papillae recorded in the present study could possibly be due to shorter gestation period and species variations. The cytological characters of epithelium of the ruminal papillae were identical to the description of ruminal epithelium. The number of layers of basal zone was more at the origin of the papillae which were gradually decreased towards the sides and tip of the papillae (Fig. 3B). Height and number of papillae increased with the advancement of gestation while reverse trend was noticed as regards the interpapillary distance. Near full term the papillae was about to reach the luminal surface of epithelium. Ruminal papillae in goat reached to half of the height of the epithelium at 76 days and the papillae became longer and thicker at 113 day[2]. Similar observations were also reported by in buffalo[6,14]. However, age of the foeti was different. In Gr. II width of the papillae present in ventral sac was more, while, their height and interpapillary distance was more in dorsal sac. In Gr. III height, width as well as interpapillary distance was more in the papillae of ventral sac than dorsal sac. The height and width of the papillae increased with advancement of age. The tip of the papilla was crecenteric shaped in Gr. II which became cylindrical in shape in Gr. III.

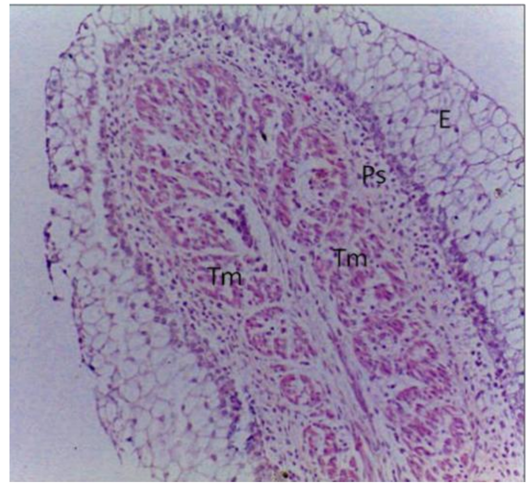

Fig. 6 — Photomicrograph of section of 134 day old goat foetal ruminal pillar showing epithelium (E), propria submucosa (Ps) and smooth muscle bundles of tunica muscularis (Tm). H & E X400

The core of the ruminal papillae was composed of differentiating mesenchymal cells, maturing fibroblasts and blood capillaries. These were the common component of lamina propria and core of the papillae. The base of the ruminal papillae was well supported by a layer of fibroblasts, the basement membrane. The ruminal papillae of sheep also had the submucosal contents in addition to lamina propria[12]. With the advancement of gestation few mesenchymal cells were transformed into fibroblasts and vascularization was also increased in the core of the papillae. These capillary networks pushed up the lamina propria to form the undulations as reported by in bovine foetal rumen[27]. The connective tissue elements comprised of reticular and collagen fibers. The concentration of reticular fibers was more at the base and tip of the papillae (Fig. 6D), whereas, collagen fibers were more only at the base. Elastic fibers were noticed from 112 days of gestation at the base of the papillae and also scattered in the central part of the papillae. The reticular fibers became branched and coarser at the tip of the papillae from 134 days onwards. The papillae formation was followed by secondary undulation incipient format at 152-160 days of gestation in bovine[10].

Tunica muscularis

The isolated smooth muscle cells were the part of pleuripotent blastemic tissue at early stages of gestation as recorded earlier in goat[2]. Arrangement of smooth muscle cells into bundles was noticed after 51 days of gestation. In between the smooth muscle bundles capillary network and connective tissue elements were noticed. Initially, inner longitudinal followed by outer obliquely or circularly directed muscle fibers were noticed (Fig. 2D). The present observations were partially in divergence with earlier reports of goat[2] in which the authors mentioned that smooth muscle fibers were arranged in two layers, inner circular and outer longitudinal at 50 days gestation. Similarly, in bovine foetus inner circular and outer longitudinal arrangement of tunica muscularis was noticed from 53 days of gestation[27]. Osman & Berg[14] observed circularly arranged smooth muscle fibers at 1.5 cm Crown Rump Length (CRL) while longitudinally directed fibers at 3.2 cm CRL in Egyptian water buffalo foeti. In present study inner obliquely to circularly and outer longitudinally directed muscle fibers were noticed between 69-87 days of gestation (Fig. 2E). Again at 94 days, the orientation of muscle fibers was identical to 51 days of gestation. With the advancement of age, the length of fibers as well as the length of their nuclei was found increased. The cytoplasm of these muscle cells was highly eosinophilic than other mesenchymal cells. Nuclei of the smooth muscle cells were initially oval or spherical in outline and became elongated or spindle shaped with the advancement of age as in human[28]. Reticular fibers were noticed between the muscle bundles which became branched and coarser from 100 days onwards (Fig. 6D). Thin collagen fibrils appeared between the smooth muscle bundles and around blood vessels between 69-87 days of gestation (Fig. 6B) and became coarser and wavy from 112 days of gestation. Amount of collagen fibers increased from 112 days onwards and arranged in parallel bundles between smooth muscle bundles and around blood vessels. Isolated, thin elastic fibers were noticed between smooth muscle bundles from 112 days onwards. Presence of connective tissue elements and blood vessels between smooth muscle bundles observed in the present study were also mentioned in terminal stages of gestation in buffalo[5]. The presence of connective tissue and blood capillaries in between smooth muscle bundles are required to provide strength and nutrition to the smooth muscle cells. At 94 days of gestation, the smooth muscle fibers were arranged inner longitudinal and outer circular manner in dorsal sac while reverse orientation was noticed in ventral sac of rumen. However, at 112-118 days of gestation the smooth muscle fibers were directed transversely and longitudinally in dorsal sac and reversely oriented in ventral sac. In between these two types of muscle bundles few oblique fibers were also encountered. Probably, the smooth muscle fibers were initially oriented either longitudinal or circular or oblique and therefore gave a different orientation pattern in same section. The tunica muscularis was thicker in ventral sac as compared to dorsal sac. This might be attributed to bear the increasing pressure of digestive viscera and ingesta. The thickness of tunica muscularis increased with advancement of age as reported in goat[2] and buffalo[14]. The tunica muscularis had been reported to play an important role in the mechanical mixing of ingesta and removal of gas through eructation, contraction and also for regurgitations.

Ruminal pillar

The epithelium and undulating blastemic tissue get evaginated towards the future lumen at the site of ruminal pillar formation (Fig. 2B). Rudimentary ruminal pillars were noticed in goat at 46 days, sheep at 42 days, red deer at 67 days, in fallow deer at 72 days and in bovine at 44 days of gestation[2,12,13,15,29]. In the present study, appearance of ruminal pillar was earlier as compared to previous studies and that could be possibly due to the species and breed differences; and foetal age determination technique. The core of the ruminal pillar was composed of loosely arranged mesenchymal cells, undifferentiating fibroblasts and scattered immature RBC'S. Differentiating myocytes were radially directed towards the tip of the ruminal pillar. The cells of neuronal elements were also encountered in the core of the pillar. With advancement of age the ruminal pillars became wider and denser and contained above mentioned structures. The dorsal and ventral sacs were partially separable with the formation of pillars. With the advancement of age connective tissue elements became coarser and thicker. The network of reticular fibers and bundles of collagen fibers were noticed around the blood vessels. The core of the ruminal pillar contained meager amount of propria submucosa and major amount of tunica muscularis and serosa (Fig. 6). Initially, tunica muscularis was arranged in the center of the core and with the advancement of gestation thickness of the tunica muscularis increased in the center as well as towards the sides of the core and arranged in bundles which were well supported by collagen, reticular fibers and blood capillaries. The main content of the ruminal pillar was tunica muscularis. All the layers of rumen had participated in the formation of ruminal pillar. The pillars of the rumen were extensive fold of entire wall which contained a core of muscle from tunica muscularis[30]. The pillars were devoid of ruminal papillae as observed earlier in goat, sheep, red deer and bovine[2,12,13,29]. The data indicated that there was an abrupt increase in the thickness of tunica muscularis within the ruminal pillar which was almost 1.5 times more in Gr. III as compared to Gr. II (Table 1).

Tunica serosa

The rumen was enveloped by a single layer of differentiating squamous cells, whose boundaries were indistinct, the mesothelium (Fig. 2C) from very early stage of its differentiation from other compartments of stomach. Present observations were in contrast to the previous reports in goat[2] in which the authors observed serosa at 58 days of gestation. At 51 days of gestation, mesothelium was well supported by loose vascular connective tissue which had major amount of ground substance. The present observation was in close proximity with goat at 58 days and bovine at 53 days of gestation[2,27]. Presence of loose connective tissue below the mesothelium helps to protect the stomach from friction because serosa secrets a thin watery secretion known as fluid. Neuronal elements containing two types of cells surrounded by connective tissue fibers were observed in present investigation. At 86 days of gestation fine reticular fibers and few collagen fibrils were noticed. With the advancement of gestation in Gr. III the fibers became coarser and abundant. The collagen fibers were arranged in thin wavy bundles. Elastic fibers were noticed from 112 days of gestation. The thickness of tunica serosa increased as age advanced but reverse was noticed in sheep and red deer[12,13].

Conclusion

Prenatal goat rumen acquire the character of a fully developed digestive tube with four definite stratum i.e., mucosa, propria submucosa, muscularis and serosa from 51 days of gestation. Ruminal papillae came into existence from 76 days of gestation. Stratification of epithelium was increased gradually with the advancement of age and about 145 days of gestation, keratinization was noticed at few places. It can be concluded that the histogenesis of rumen was almost completed in prenatal life. However, to become functional it still required more time as process of keratinization was yet to be completed.

Conflict of Interest

Authors declare no competing interests.

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