



GROSS AND HISTOMORPHOMETRIC STUDY OF FORESTOMACH OF JATTAL AND NACHI GOAT BREEDS

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Abstract

The digestive mechanism of goats, particularly the forestomach, allows converting low-quality forages into high-nutritional-value products. As a result, understanding the functional architecture of the forestomach is critical since it is intimately involved with the conversion of food to valuable products like as milk and meat. A total of 30 goats, 15 each of the Nachi and Jattal breeds, were chosen from the slaughter house, the Punjab Agriculture and Meat Company (PAMCO), Lahore, Pakistan for this investigation. All of the animals were between the ages of 1 and 2 years. The rumen was sampled from five distinct sites including cranial, dorsal, ventral sac, caudo-dorsal blind sac, and caudo-ventral blind sac. Omasal specimens were collected from several areas including reticulo-omasal, middle omasal, and omaso-abomasal junction. For the reticulum, samples were obtained from the medial wall of the reticulum. The data was analysed using an unpaired T-test through SPSS 20.0 software. The level of significance was $p \leq 0.05$. The results showed that there was a significant difference in gross anatomical and histomorphometric parameters of rumen, reticulum and omasum of Nachi and Jattal breeds. However, in gross anatomical parameters, more relative weight of filled rumen and omasum was found in Jattal breeds. In histomorphometric parameters, muscularis thickness, papillae length, thickness of keratinized epithelial layer and total epithelial thickness of rumen was more in Nachi breeds. It was concluded that Gross anatomical features of forestomach in Nachi breed are greater than those of Jattal breed. However, histomorphometric parameters are greater in Jattal than in Nachi breed. This study will assist to differentiate the anatomical and histomorphometric parameters of Jattal and Nachi goat breeds from other analogous small ruminants.

Key Words: Forestomach, Jattal and Nachi breed, Rumen, Reticulum, Omasum

Introduction

Goats are animals with ungulated hooves. They are classified as members of phylum Chordata, class Mammalia, order Artiodactyla (cloven hooves are their distinguishing feature), suborder Ruminantia (animals that regurgitate their food), family *Bovidae* and subfamily *Capra*. The domestication of goat is from *Capra argagrus hircus* species (Suárez et al. 2006). They were one of the first

domesticated animals in Western Asia, some 10,000 years ago. The production of milk, meat and hides is currently from over 300 different domestic goat breeds with a wide range of physical traits, sizes, and weight. Goats are friendly, curious, kind, sensibly active, tidy, unharmed and sturdy animals; making them appealing and practical animal models (Amit et al. 2011). Goats are one of the fastest-growing species of livestock in Pakistan and at present, there are about 76.1 million goats of various breeds in the country. Pakistan has both milk and meat-producing goat breeds and they contribute immensely to the livestock sector. They produce 762-tons of mutton and 940-tons of milk annually (Wasti et al. 2019). In Pakistan, different goat breeds are present in different climatic conditions among which, Nachi and Jattal are of significant importance as a source of livelihood for people of two different geographical regions. Nachi breed is found in hot climatic conditions of district Multan, Bahawalpur, and Bahawalnagar whereas Jattal goat is present in cold climatic conditions of Mirpur and Kotli. These two goat breeds are used for meat purposes. The adult weight of the male goat of Nachi is 38 kg and that of the female is 32 kg while the adult weight of the male goat of Jattal is 23 kg and that of the female is 19 kg (Yamamoto et al. 2018).

Ruminants are divided into two categories: grazers and browsers. The grazers have a smaller veritable abomasum, caecum, and intestines. Whereas, the browsers have a sizeable foregut. Because of this physical difference, browsers can digest the fibrous parts of the plant whereas grazers can digest high-quality feed (Membrive, 2016). Goat falls in the category of browsers and is the one that makes good use of pasture land (Mutoh, Wakuri, 2020). Although all the ruminants have this ability, the low-quality forages are transformed by them into products of great nutritious importance and this is due to their digestive system structure. This particular ability of grazing is related to the unique morphology of the ruminant stomach (Soni et al. 2018).

Forestomach plays crucial roles such as those of fermentation (rumen), absorption of fatty acids (reticulum), absorption of water and other nutrients (omasum) and in general microbial fermentation of ingesta to produce volatile fatty acids. The substantial fraction of volatile fatty acids not absorbed through the wall of rumen, as well as water and electrolytes such as sodium and potassium, are absorbed through the omasal laminae (Tamate et al. 2020). Histologically, the tunica mucosa, submucosa, muscularis, and serosa are the four tunics that make up the forestomach wall. The stratified squamous epithelium, lamina propria, and lamina muscularis mucosa make up the tunica mucosa. The lamina muscularis mucosae is absent from the ruminal mucosa (Garcia et al. 2013).

The reticulum along with rumen is regarded as a functional compartment. Because of its honeycomb-like lining, the reticulum is known as the "honeycomb." The ingesta readily passes between the reticulum and the rumen.

The fundamental function of the reticulum is to collect and transport smaller particles to the omasum, while larger particles remain in the rumen for further digestion (Phillipson et al. 2018). Omasum is the third and terminal compartment of the ruminant forestomach. Omasum contains numerous collateral laminae of contrasting sizes which are made up of thin muscular layers and a mucous membrane that is non-glandular with numerous papillae on the surface. Omasal laminae of numerous shapes are arranged in 4-5 orders and they also differ by their distribution and direction.

These longitudinal folds called laminae are flecked with papillae, giving them a cabbage leaf look. The epithelium of the omasum is stratified squamous keratinized composed of four layers (basale, granulose, spinose, and corneum) (EL-Gendy, Derbalah, 2016). Solids and liquids are retained in the omasum for a very short period due to their limited capacity.

However, the omasal laminae's broad surface area indicates that the omasum is an absorptive organ (Chandrasekar et al. 2019). To date, no comprehensive study about histology and histomorphometry of forestomach of Jattal and Nachi breed has been carried out. The present study was aimed to thoroughly explore and describe the morphology of forestomach compartments of adult healthy goats of Nachi and Jattal breeds to report the gross anatomical and histomorphometric features in these breeds.

Materials and Methods

The total 30 goats were selected for this study; 15 each of Nachi and Jattal breeds. The goats were accessed through the Punjab Agriculture and Meat Company (PAMCO) slaughterhouse. After accessing their data, it was ensured that all the animals were disease-free and without any recent or existing gastrointestinal disease. Animals were of age 1-2 years. Age, average weight and health of animals were assessed through antemortem examination.

The age of the goats were determined by dentition. Number of erupted permanent incisors were counted for determining the age in years. There were two permanent incisors in each goat that ensured each goat was of 2 years. The dental formula of goat is:

Deciduous teeth: $2(I\ 0/4\ P3/3)=20$

Permanent teeth: $2(I\ 0/4\ P3/3\ M3/3)=32$

The weight of the goats was ascertained by weighing balance. Average weight of Nachi goat was 35.69 kg and of Jattal was 24.91 kg. All animals were apparently healthy. Health status of goats, as assessed from their hair coat, body secretions, absence of inflammation, and other signs of distress, was evaluated during ante-mortem inspection of goats in lairage.

The inspection of live animals prior to slaughter is known as ante-mortem inspection. An ante-mortem inspection is required for all livestock brought for slaughter. In PAMCO Lahore, ante-mortem examination was performed under the supervision of the Veterinary Officers. Animals with existing or recent gastrointestinal disease or any systemic disease that leads to debilitation were excluded during ante-mortem examination in lairage. Samples were taken from five different locations of the rumen (cranial, dorsal, ventral sac, caudo-dorsal blind sac and caudo-ventral blind sac). The rumen's atrial sac has a spacious connection with the reticulum and can be detected (Chandrasekar et al. 2019). For the reticulum, samples were taken from the reticulum's medial wall (Franco et al. 2012). For omasum, samples were taken from different regions of omasum (reticulo-osamal, middle omasal and omaso-abomasal junction) (Malik et al. 2012).

Gross Observations

After slaughtering the animals, the weights of empty and filled rumen, reticulum, and omasum were measured with the help of weighing balance (SF-400A electronic Compact Scale) in grams. By cutting lateral to the cranial coronary pillar, the cranial sac was separated from the dorsal sac. By cutting across the periphery cranial to the dorsal and ventral coronary pillars, the caudodorsal blind sac and caudoventral blind sac were detached, leaving the caudal pillar linked to the ventral sac. By cutting dorsal to the right longitudinal and cranial pillars, the ventral sac was separated from the dorsal sac. Circumference of forestomach compartments was measured by closing the inlet and outlet openings by ligatures. Circumference was measured with the help of measuring tape (Jelínek et al. 2005).

The compartments were then emptied, washed in running tap water for 10 minutes and all the ingesta was removed. Subsequently, the internal surface of all the three compartments was examined. In the rumen, the papillae height was recorded in its different regions (ventral sac and caudoventral sac (blind) with the help of digital Vernier Calliper (Precision measuring digital calliper 150mm, China). The papillae of ventral and dorsal sacs. In reticulum, reticular cells area (area of the reticular polygonal cell) was recorded by measuring all the sides of a polygon and by taking apothem of the polygon with the help of digital vernier calliper (Bancroft et al. 2013). The area of polygonal cell of reticulum was calculated by using the following formula:

$A = 1/2 \times \text{apothem} \times \text{perimeter}$

For omasum, after taking empty and filled weight omasal fold number, height, and order (primary, secondary, tertiary) of profuse laminae were measured grossly (EL-Gendy, Derbalah, 2016).

Histological Observations

Rumen

For histomorphometry of rumen, rumen was dissected and samples of about 2 x 2 cm each were taken from the 5 different locations of rumen i.e. dorsal sac, cranial sac, ventral sac, caudo-ventral

sac (blind), caudo-dorsal sac (blind), and finally the cranial sac (blind) (Soni et al. 2016). Immediately after sampling tissues were immersed for 24- hours in a 10% neutral buffered formalin solution for fixation. The sample were then processed through the paraffin embedding technique and stained with H&E staining technique as described by (Clauss et al. 2010).

Paraffin Embedding Technique

After fixation of tissues for 24 hours in neutral buffered formalin (10%), tissues were washed with tap water for 2 hours. The first step after fixation is dehydration. Ascending series of ethyl alcohol was used for tissue dehydration. Samples were placed in tissue cassettes, which were placed for two hours each in 70%, 80%, 90%, and 2 concentrations of absolute (100%) alcohol solution for completion of dehydration procedure. Tissue cassettes were held in Xylene I and Xylene II for two hours each for clearing. Clearing is necessary for eliminating alcohol and allowing paraffin wax to infiltrate the tissue. Paraffin wax was used for infiltration. Tissue cassettes were immersed in a beaker containing paraffin wax and placed into an automatic tissue processor (RH-12EP2 Japan) for 6 hours at 58°C temperature. After completion of the infiltration procedure, paraffin blocks were formed by using metallic molds. Three slides per sample were studied for histomorphometry which include thickness of tunica mucosa, submucosa, muscularis, serosa, entire wall thickness and thickness of keratinized and non-keratinized layer of epithelium. Connective tissue and muscular component within tunics were also studied histologically [14]. All histomorphometric measurements are shown in Fig 1-11.

Statistical Design

The normal distribution of the data was checked by using the Kolmogorov-Smirnov test (Mahesh et al. 2014). Descriptive statistics were used to describe individual parameters of two breeds, whereas the comparison between two breeds was made by using an unpaired T-test (SPSS 20.0, Chicago IL, USA) (Tamate et al. 2020). The level of significance was $p \leq 0.05$.

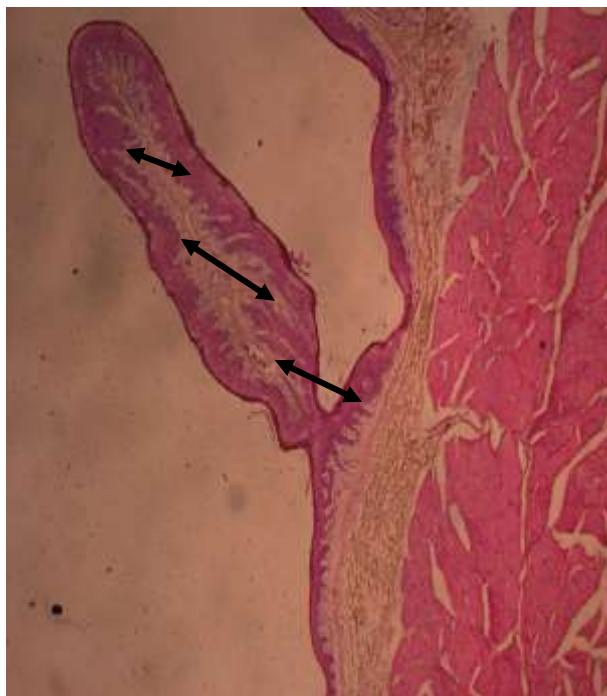


Fig 1: Rumen Tunics

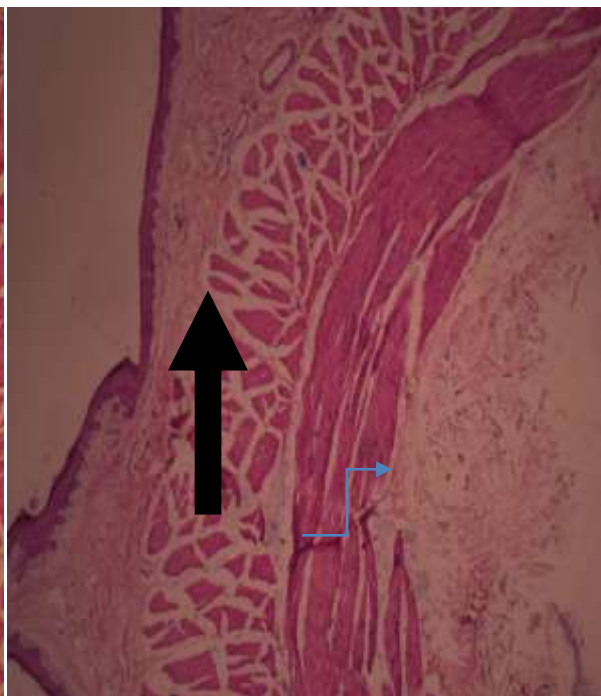


Fig 2: Ruminal Papilla Length and Width

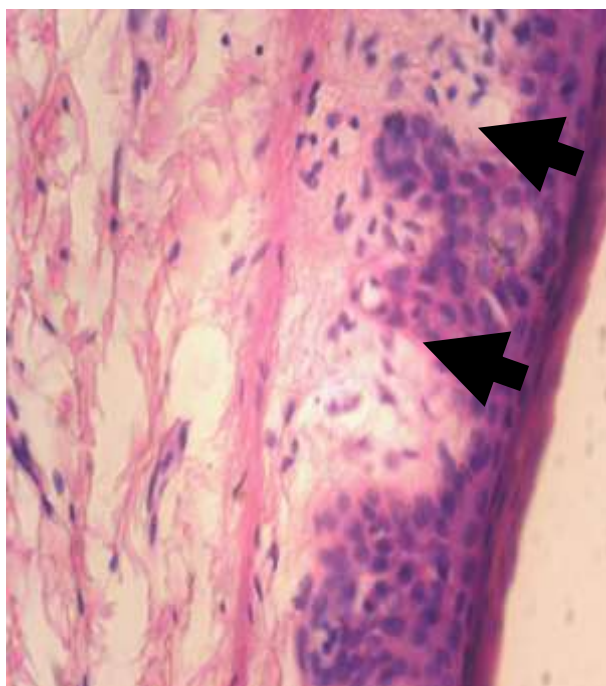


Fig 4: Rumen Epithelium Thickness of Keratinized Layer and Total Epithelial Thickness

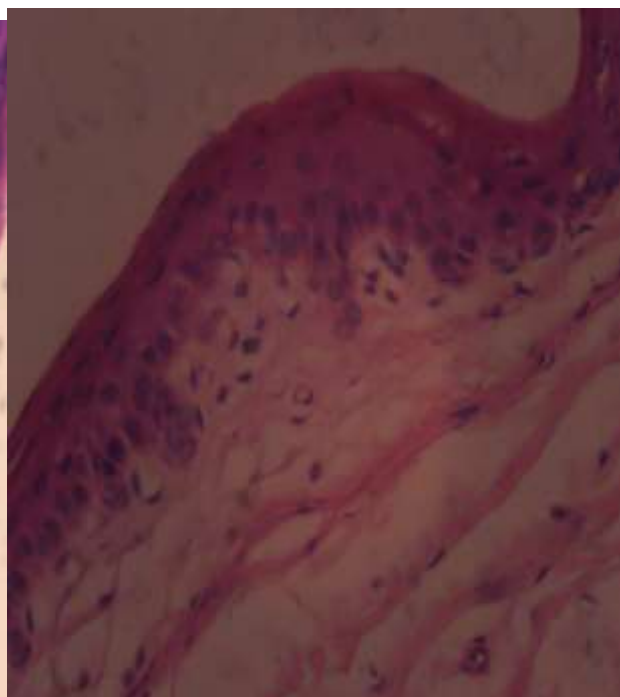


Fig 3: Ruminal Epithelium

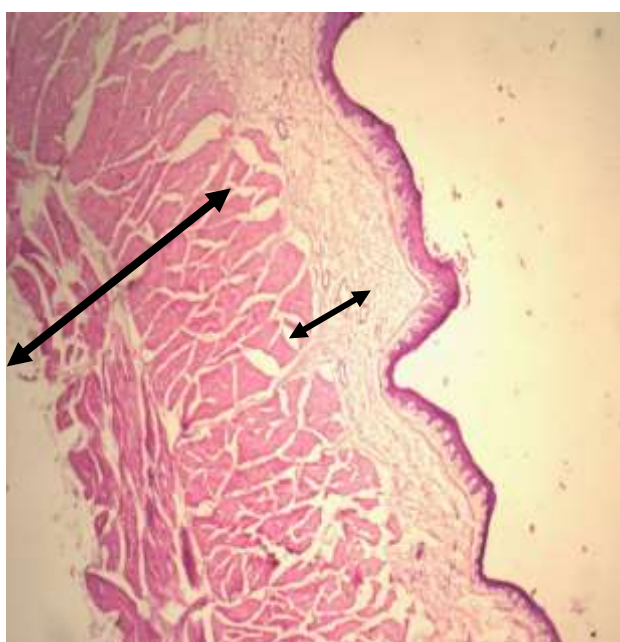


Fig 6: Reticulum Tunics

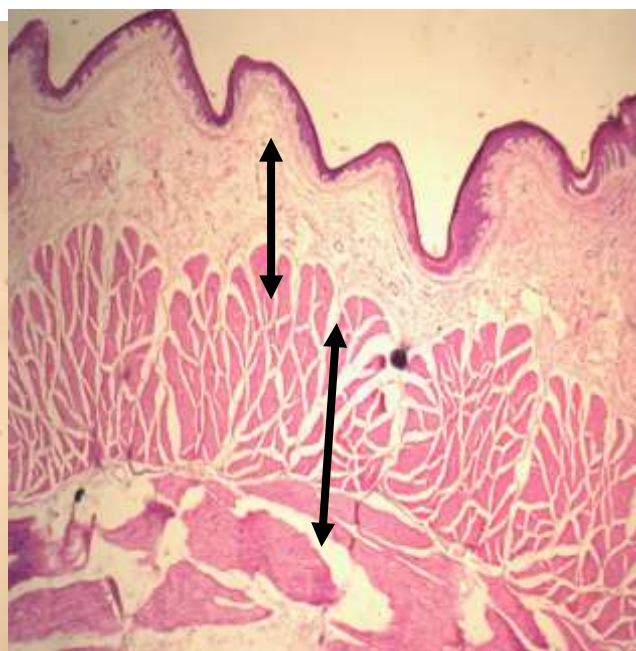


Fig 5: Rumen Tunics Thickness (Epithelium, Propria Mucosa and Muscular layer)

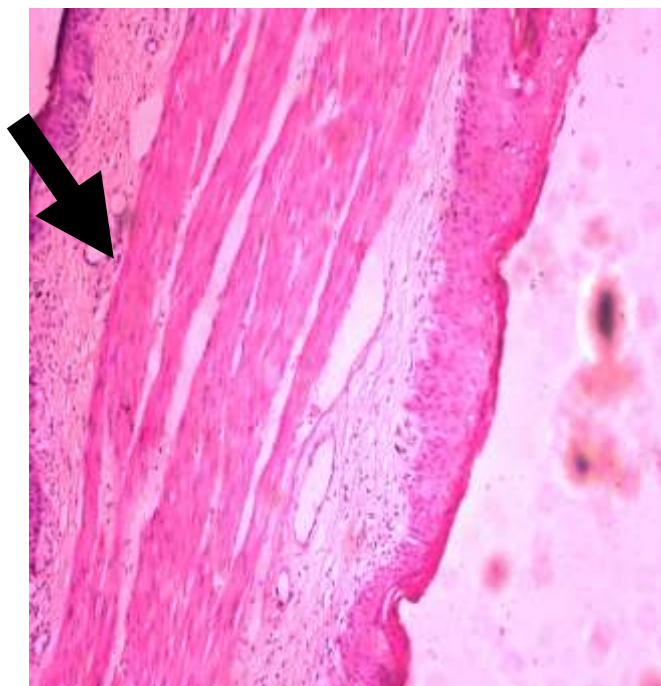


Fig 8: Omasum Tunics Thickness (Tunica Mucosa, Submucosa, Muscular Layer Thickness)

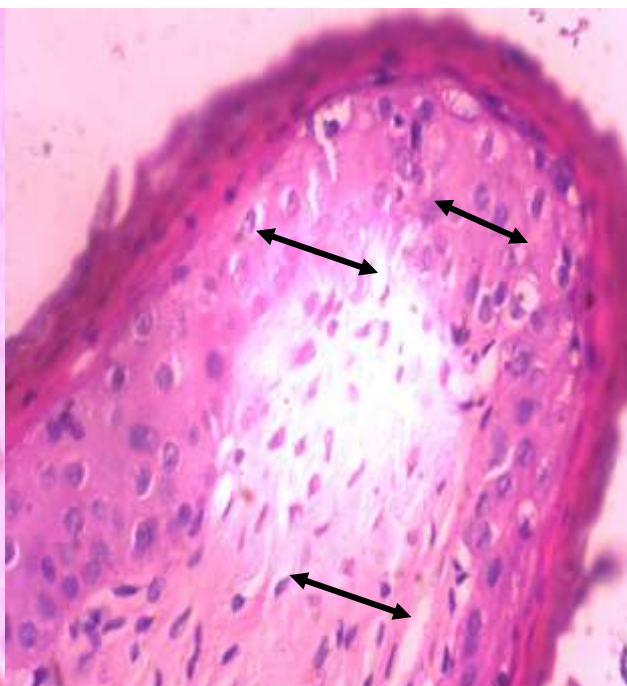


Fig 7: Reticulum Epithelium Keratinized Layer and Total Layer Thickness

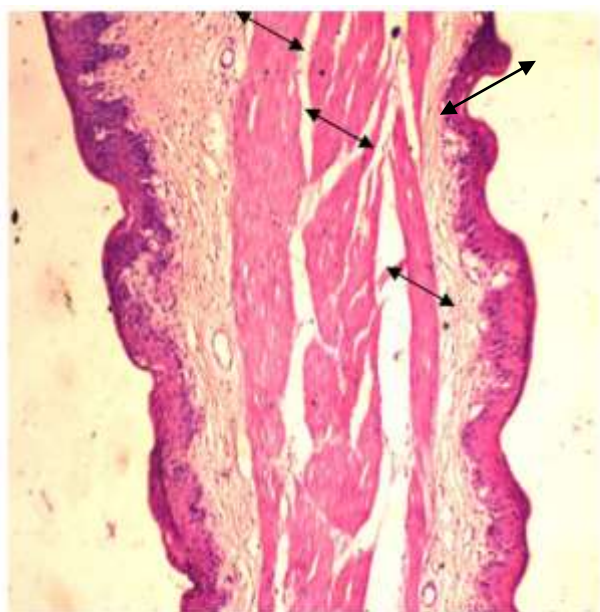


Fig 9: Omasum Tunics

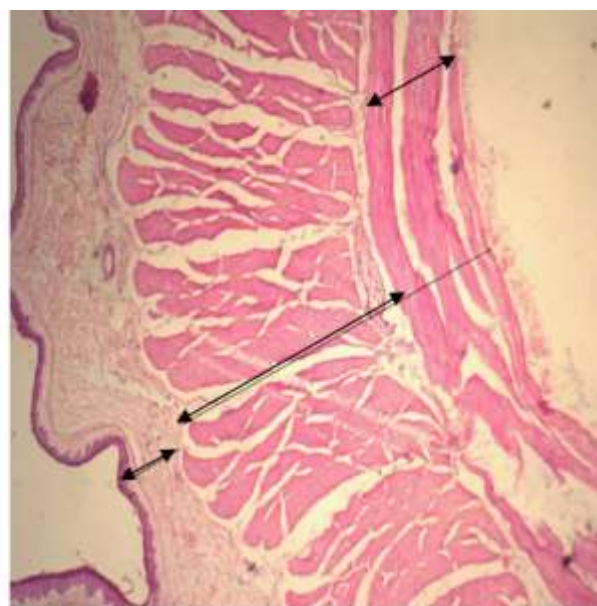


Fig 10: Histomorphometry Tunics Rumen (Epithelium Thickness, Propria-Submucosa and Muscular Layer Thickness)

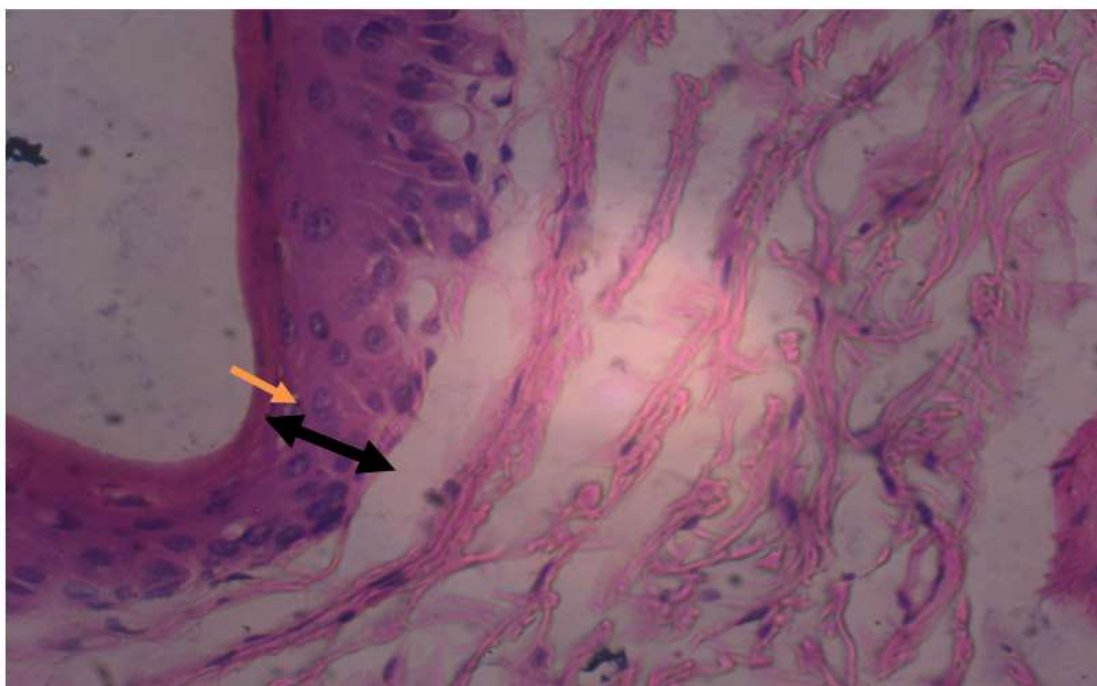


Fig 11: Histomorphometry Epithelium (Keratinized Epithelial Layer and Total Epithelial Layer Thickness). Yellow Arrow is Showing Total Epithelial Thickness While Black Arrows Shows Keratinized Layer and Non-keratinized Layer Thickness of Epithelium

Results

Gross Anatomical Parameters of Forestomach

The relative empty weight of the rumen was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. In contrast, absolute empty and filled weight of rumen, absolute rumen circumference and lengths of papillae in ventral and caudo-ventral sacs were greater ($p \leq 0.05$) in Nachi breed as compared to Jattal. Whereas no difference ($p > 0.05$) was observed in relative filled weight of rumen of Jattal and Nachi goat breeds. The relative empty and filled weights of the reticulum were greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. Whereas, absolute empty and filled weight of reticulum, absolute reticular circumference and reticular cell area were greater ($p \leq 0.05$) in Nachi breed as compared to Jattal.

Histomorphometric Parameters of Rumen (Cranial and Ventral Sac).

All the histological parameters like thickness of kaeratinized and non- keratinized epithelial layers, muscularis, papillae length and width was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed except lamina propria-submucosa thickness which was higher in Jattal than Nachi breed. All the histological parameters like thickness of keratinized and non- keratinized epithelial layers, muscularis was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed except lamina propria-submucosa thickness which was higher in Jattal than Nachi breed.

Histomorphometric Parameters of Rumen (Caudo-Ventral Sac).

The thickness of tunica muscularis was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, thickness of lamina propria-submucosa was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. Whereas, no difference ($p > 0.05$) in thickness of keratinized and non-keratinized epithelial layers was observed between Jattal and Nachi goat breeds.

Histomorphometric Parameters of Reticulum.

The thickness of tunica mucosa, muscularis and keratinized and non-keratinized epithelial layer was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, thickness of submucosa was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed.

Histomorphometric Parameters of Omasum (Middle Omasal Region)

The thickness of submucosa, and non-keratinized epithelial layer was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, thickness of tunica muscularis was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. Whereas, no difference ($p > 0.05$) in thickness of mucosal layer and thickness of keratinized was observed between Jattal and Nachi goat breeds.

Histomorphometric Parameters of Omasum (Omaso-Abomasal Junction)

The thickness of mucosa, muscularis and non-keratinized epithelial layer was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, thickness of submucosa was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. Whereas no difference between two breeds was observed in thickness of keratinized epithelial layer.

Relative Lengths of Ruminal Papillae Against Empty Organ Weight

The relative length of ruminal papillae (caudo-dorsal sac) was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, lengths of ruminal papillae in ventral and cranial sacs was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed. Whereas no difference between two breeds was observed in length of ruminal papillae of dorsal sac and caudo-ventral sac.

Relative Values of Omasal Folds Height and Reticular Cell Area Against Empty Organ Weight

The relative height of omasal primary and tertiary fold and reticular cell area was greater ($p \leq 0.05$) in Nachi breed as compared to Jattal breed. In contrast, Relative height of omasal secondary fold was greater ($p \leq 0.05$) in Jattal breed as compared to Nachi breed.

All the gross anatomical parameters of rumen, reticulum and omasum of Jattal and Nachi breeds are reported in Table 1, histomorphometric parameters of rumen in Table 2 and histomorphometric parameters of reticulum and omasum in Table 3.

Table 1: Gross anatomical Parameters of Rumen, Reticulum, and Omasum of Jattal and Nachi Goat Breeds (Mean \pm SEM).

Compartments	Parameters	Jattal	Nachi	P-Value
Rumen	Absolute weight of empty rumen (gm)	545.80 \pm 7.30 ^b	615.60 \pm 2.32 ^a	0.000
	Relative weight of empty rumen (gm)	2.15 \pm 0.51 ^a	1.68 \pm 0.009 ^b	0.000
	Absolute weight of filled rumen (gm)	3308.73 \pm 52.12 ^b	4503.73 \pm 7.87 ^a	0.000
	Relative weight of filled rumen (gm)	13.07 \pm 0.18	12.97 \pm 0.37	0.668
	Rumen circumference (inches)	24.26 \pm 0.44 ^b	30.00 \pm 0.35 ^a	0.000
	Papillae length (ventral sac) (mm)	6.13 \pm 0.09 ^b	7.12 \pm 0.14 ^a	0.000
	Papillae length (caudo-ventral sac) (mm)	4.89 \pm 0.106 ^b	5.54 \pm 0.095 ^a	0.000
Reticulum	Absolute weight of empty reticulum (gm)	116.06 \pm 2.89 ^b	136.26 \pm 0.89 ^a	0.000
	Relative weight of empty reticulum (gm)	0.43 \pm 0.12 ^a	0.33 \pm 0.11 ^b	0.000
	Absolute weight of filled reticulum (gm)	410.00 \pm 14.44 ^b	492.73 \pm 2.45 ^a	0.000
	Relative weight of filled	1.67 \pm 0.46 ^a	1.39 \pm 0.01 ^b	0.000

	reticulum (gm)			
	Reticulum circumference (inches)	11.73±0.31 ^b	14.26±0.27 ^a	0.000
	Reticular cell area (mm ²)	84.70±0.43 ^b	91.83±1.21 ^a	0.000
Omasum	Absolute weight of empty omasum (gm)	65.86±1.06 ^b	86.06±0.93 ^a	0.000
	Relative weight of empty omasum (gm)	0.24±0.005 ^a	0.21±0.003 ^b	0.000
	Absolute weight of filled omasum (gm)	194.93±2.87 ^b	216.33±0.99 ^a	0.000
	Relative weight of filled omasum (gm)	0.79±0.01	0.75±0.02	0.274
	Omasum circumference (inches)	5.76±0.11 ^b	8.20±0.15 ^a	0.000
	Omasal primary fold height (mm)	21.60±0.31 ^b	27.36±0.31 ^a	0.000
	Omasal secondary fold height (mm)	14.84±0.14 ^b	18.03±0.17 ^a	0.000
	Omasal tertiary fold height (mm)	5.17±0.12 ^b	5.76±0.13 ^a	0.003
	Omasal fold number	22.86±0.25 ^b	28.26±0.206 ^a	0.000

Table 2: Histomorphometric Parameters (µm) of Rumen (Dorsal Sac), (Caudo-Dorsal Sac), (Cranial Sac), (Ventral Sac), and (Caudo-Ventral Sac), of Jattal and Nachi Goat Breeds (Mean±SEM)_z

Compartments	Parameters	Jattal	Nachi	P-Value
Rumen (dorsal sac)	Thickness of keratinized epithelial layer	17.94±0.15 ^b	19.37±0.09 ^a	0.000
	Total epithelial thickness	70.13±0.29 ^b	85.81±0.24 ^a	0.000
	Lamina propria-submucosa thickness	303.40±0.25 ^b	610.67±0.76 ^a	0.000
	Muscularis thickness	1245.50±0.94	1252.79±8.51	0.409
	Total wall thickness	1641.55±8.63 ^b	1926.38±1.25 ^a	0.000
	Papillae length	1290.36±17.60	1328.15±8.80	0.065
	Papillae width	365.51±5.15 ^b	390.57±5.23 ^a	0.002
Rumen (caudo-dorsal sac)	Thickness of keratinized epithelial layer	12.82±0.13 ^b	18.66±0.18 ^a	0.000
	Total epithelial thickness	44.43±0.27 ^b	83.43±0.25 ^a	0.000
	Lamina propria-submucosa thickness	276.31±0.42 ^b	576.91±0.38 ^a	0.000
	Muscularis thickness	1045.05±3.44 ^b	1566.39±0.56 ^a	0.000
	Total wall thickness	1404.68±3.54 ^b	2188.03±0.84 ^a	0.000
	Papillae length	1220.12±14.69 ^b	1829.28±11.7 ^a	0.000
	Papillae width	310.38±4.45 ^b	359.78±28.07 ^a	0.000
Rumen (cranial sac)	Thickness of keratinized epithelial layer	10.12±0.13 ^b	21.18±0.12 ^a	0.000

	Total epithelial thickness	38.65±0.29 ^b	86.70±0.21 ^a	0.000
	Lamina propria-submucosa thickness	491.72±0.21 ^a	280.24±0.30 ^b	0.000
	Muscularis thickness	1327.58±0.47 ^a	1365.72±4.64 ^b	0.000
	Total wall thickness	1895.95±0.73 ^a	1694.48±4.57 ^b	0.000
	Papillae length	1192.89±16.21 ^b	1794.26±14.66 ^a	0.000
	Papillae width	319.88±2.78 ^b	380.27±5.94 ^a	0.000
Rumen (ventral sac)	Thickness of keratinized epithelial layer	14.01±0.20 ^b	24.08±0.22 ^a	0.000
	Total epithelial thickness	66.27±0.26 ^b	91.20±0.23 ^a	0.000
	Lamina propria-submucosa thickness	537.54±0.94 ^a	497.18±0.51 ^b	0.000
	Muscularis thickness	1265.70±0.74 ^b	1341.59±2.11 ^a	0.000
	Total wall thickness	1869.50±1.48 ^b	1930.44±2.05 ^a	0.000
Rumen (caudo-ventral sac)	Thickness of keratinized epithelial layer	14.17±0.15	15.86±0.13	0.166
	Total epithelial thickness	61.78±0.51	62.73±0.30	0.094
	Lamina propria-submucosa thickness	494.76±0.57 ^a	493.26±0.21 ^b	0.000
	Muscularis thickness	1225.70±0.61 ^b	1270.17±1.83 ^a	0.000
	Total wall thickness	1782.61±0.75 ^b	1825.16±1.85 ^a	0.000

Table 3: Histomorphometric Parameters (µm) of Rticulum and Omasum(Medial Omasal Region) of Jattal and Nachi Goat Breeds (Mean±SEM)

Compartments	Parameters	Jattal	Nachi
Reticulum	Mucosa thickness	85.39±0.58 ^b	122.94±0.39 ^a
	Submucosa thickness	574.67±0.62 ^a	555.27±6.03 ^b
	Muscularis thickness	1274.82±0.38 ^b	1360.96±3.51 ^a
	Total wall thickness	1934.88±1.08 ^b	2039.17±6.23 ^a
	Thickness of keratinized layer	15.37±0.14 ^b	19.07±0.10 ^a
	Total epithelial thickness	67.00±0.24 ^b	76.20±0.25 ^a
Omasum	Mucosa thickness	113.48±0.57	115.32±1.01
	Submucosa thickness	123.38±0.65 ^a	160.82±0.50 ^b
	Muscularis thickness	185.66±0.46 ^a	166.52±0.53 ^b
	Total wall thickness	416.53±0.79 ^a	443.68±1.19 ^b
	Thickness of keratinized layer	14.99±0.20	15.50±0.12
	Total epithelial thickness	82.81±0.23 ^a	107.50±0.24 ^b

Discussion

The present study revealed that forestomach in Nachi and Jattal goat breeds consisted of three compartments rumen, reticulum and omasum. These three compartments were easily identified. Rumen took approximately half of the abdominal cavity on the left side and extended well beyond the median plane ventrally. Its long axis extended almost to the pelvic inlet from the point opposite the ventral section of the seventh or eighth intercostal space. The 5 rumen sacs were also identified; dorsal sac, ventral sac, caudo-dorsal blind sac, caudo-ventral blind sac, and cranial sac, Similar findings was reported by Tamate et al. (2020) who identify same 5 rumen sacs in goats. Reticulum

was seen between the sixth and seventh ribs, more cranially. The rumino-reticular junction connected it to the rumen, while the reticulo-omasal junction connected it to the omasum. The omasum, on the other hand, was ellipsoidal in shape and somewhat compressed between the parietal and visceral surfaces. It was clearly distinguished from the rest of the compartments. It was located primarily to the right of the median plane, just across from the seventh and eleventh ribs. Our findings were similar to those of Clauss et al. (2010) who reported that rumen is located opposite to the ventral sections of seventh and eighth intercostal space, reticulum is located between sixth and seventh ribs and omasum is present across seventh and eleventh ribs.

The caudal blind sacs were marked by lesser coronary pillars, which were evident as grooves on the rumen's exterior surface. Our findings were comparable to those of (Mahesh et al. 2014) who studied the rumen of sheep and reported that sheep rumen had 5 ruminal portions and exteriorly these subdivisions appeared as grooves. Main ruminal pillar split the organ into dorsal and ventral sacs whereas caudal blind sacs were marked by lesser coronary pillars. Empty weight of the rumen relative to the body weight was greater ($p \leq 0.05$) in Jattal breed (2.15 ± 0.51 g) as compared to Nachi breed (1.68 ± 0.009 g). In contrast, absolute circumference of rumen was greater ($p \leq 0.05$) in Nachi breed (30.00 ± 0.35 inches) as compared to Jattal (24.26 ± 0.44 inches). Ruminal papillae were seen most prominently on ventral and caudo-ventral blind sac. Absolute height of ruminal papillae was taken by digital vernier calliper. Papillae height of ventral and caudo-ventral blind sac was greater ($p \leq 0.05$) in Nachi breed (7.12 ± 0.14 mm and 5.54 ± 0.095 mm) as compared to Jattal (6.13 ± 0.09 mm and 4.89 ± 0.106 mm) respectively. Whereas no difference ($p > 0.05$) was observed in relative filled weight of rumen of Jattal (13.07 ± 0.18 g) and Nachi goat (12.97 ± 0.37 g). Absolute empty and filled weight of rumen of Nachi (615.60 ± 2.32 g and 4503.73 ± 7.87 g) was greater ($p \leq 0.05$) as compared to Jattal (545.80 ± 7.30 g and 3308.73 ± 52.12 g) respectively. There exists a comparison between these results and those of Jelinik, (2005) in his study on goats. His findings for absolute empty and filled weight of rumen were (603.00 ± 36.09 g and 4500 ± 31.42 g) respectively. For absolute circumference of rumen, his findings were (28.36 ± 0.65 inches). According to his findings height of ruminal papillae of ventral sac was (6.50 ± 0.89 mm) and of caudo-ventral sac was (5.24 ± 0.43 mm). These findings are comparable to our current findings on Jattal and Nachi goats. The difference in our findings of two breeds Jattal and Nachi may be due to the reason that, two goat breeds under study are different from many aspects i.e. body size and geographical origin, and nutrition, Nachi is large sized breed to meet the energy and nutritional requirements of large sized goats, its stomach function is more apparent and so are the stomach structures. Papillae heights in our study were higher in ventral sacs compared to other sacs in both breeds. Possible reason of this change is evident from the findings of Tamate et al. (2020) who said that exposure to the nutrients and the concentration of short chain volatile fatty acids determine the size, quantity, and surface area of papillae.

The reticulum was much smaller than the rumen, being immediately cranial to it and in contact with the diaphragm's caudal surface. It was located above the xiphoid process of the sternum, just ventral to the esophageal-gastric junction. The reticular mucosa was similar to the ruminal mucosa in that it was non-glandular and had a stratified epithelium. Ridges that delineated 4, 5, and 6-sided cells provided a characteristic honeycomb appearance. Short papillae ran between these ridges and the cell floors. The smooth muscle of the ruminoreticular wall was divided into two layers, with the outer layer being thinner and the inner layer being thicker, with the fibres oriented at a more or less perpendicular angle to one another. These findings match those of **Beharka et al. (2018)** who reported that in goats the reticulum is located above the xiphoid process of sternum and he also discussed the honey comb like structure of reticulum. Relative empty and filled weights of the reticulum were greater ($p \leq 0.05$) in Jattal breed (0.43 ± 0.12 g and 1.67 ± 0.46 g) as compared to Nachi breed (0.33 ± 0.11 g and 1.39 ± 0.01 g) respectively. Present study revealed that the omasum of ruminants was located to the right of the rumino-reticular compartment in the intrathoracic region of the abdomen. It was shaped like a bean. The interior was occupied by a multiplicity of several laminae that protruded from the top and sides and projected to the bottom though provided enough space for the omasal canal. The crescentic laminae were a series of small recesses that partitioned

the lumen into different lengths and diameters. The laminae were thin muscle sheets that were covered with a non-glandular mucosa that contained small papillae. The omasal laminae appeared as folds. Similar findings were reported by (Shoeib et al. 2015). These findings are comparable to those of (Malik et al. 2011) in his study on omasum of goats who told that absolute empty omasal weight was $(80.02 \pm 1.21\text{g})$ and absolute filled omasal weight was $(213.70 \pm 14.09\text{g})$ whereas his findings for omasal circumference was $(8.13 \pm 1.09\text{inches})$. it was also reported that the omasal fold number 28 in goats was similar to our findings in Nachi, whereas 33 omasal folds in cattle were reported by Scocco et al. (2016) while 24 folds were reported by Soni et al. (2018) in his study on lambs.

The rumen is the most important compartment of the forestomach because it is a fermentative chamber. Through microbial fermentation, it digests cellulose and hemicellulose to generate volatile fatty acids, which are the primary source of energy for ruminants (Scocco et al. 2016). Interior of the rumen is covered by papillae which are the projections protected by stratified squamous keratinized epithelium and they have a vital contribution in increasing the surface area of the rumen to increase the absorption as reported by (Tamate et al. 2020). The present study revealed that papillae of different sizes and shapes appeared in rumen of Jattal and Nachi goats. The appearance of various sized papillae that were elongated and cylindrical in shape revealed that the mucosal surface of the rumen was uneven. Sheep's rumen papillae were often conical or tongue-shaped (Emam et al. 2016). The cow's ruminal mucosa had three forms of papillae: elongated filiform, short conical, and short fungiform Scala et al. (2016) whereas the buffalo's ruminal mucosa had only two types: long foliaceous and blunt conical papillae (Scocco et al. 2016). These papillae offered a greater surface area for metabolic activities such as fatty acid absorption, water absorption, and electrolyte absorption (Chandrasekar et al. 2019). The reticulum is also considered a functional chamber of the forestomach. Histological studies of epithelium and lamina propria, the muscularis and serosa, have been carried out on the reticulum of Jattal and Nachi goat breeds. Our findings revealed that the hexagonal honey comb-like shape of the reticulum of Nachi and Jattal goat breeds was characterized by small and large honey comb cells lined by stratified squamous keratinized epithelium, as reported in sheep (Endo et al. 2011). Reticular fold was formed by folding of the epithelial mucosa of large cells. Secondary and tertiary papillae of various shapes and sizes arose from the folds and neighboring reticular mucosa as reported earlier (Scott and Gardner, 2013). According to Endo et al. (2011), the serosa was a thin, stiff covering that contain loose connective tissue, blood vessels, and nerves in buffaloes. Similar results were reported by Beharka et al. (2018) in his study on goats. Histomorphometric measurements of reticulum of Jattal and Nachi goat breeds showed that thickness of tunica mucosa, muscularis and keratinized epithelial layer was greater ($p \leq 0.05$) in Nachi breed (122.94 ± 0.39 , 1360.96 ± 3.51 and $19.07 \pm 0.10\mu\text{m}$) as compared to Jattal breed (85.39 ± 0.58 , 1274.82 ± 0.38 and $15.37 \pm 0.14\mu\text{m}$) respectively. In contrast, thickness of submucosa was greater ($p \leq 0.05$) in Jattal breed (574.67 ± 0.62) as compared to Nachi breed (555.27 ± 6.03) respectively. The difference is might be due to the different diets and different climate conditions of areas of two breeds. Our findings are comparable to results of Bancroft et al. (2016) who reported the thickness of tunica mucosa in goat ($88.56 \pm 0.87\mu\text{m}$), submucosa ($582.74 \pm 0.37\mu\text{m}$), and muscularis ($1289.65 \pm 4.86\mu\text{m}$). Contrarily, Jelínek, (2005) reported different findings during his study on reticulum of goats during prenatal period. He reported the thickness of tunica mucosa, submucosa and serosa (556.32 ± 2.65 , 185.98 ± 0.36 and $186.42 \pm 0.16\mu\text{m}$) respectively. This difference might be due to the difference in the development of structures from prenatal to postnatal life. There exists comparison between our findings and those of Endo et al. (2011) who reported the thickness of keratinized epithelial layer of goat reticulum as ($18.68 \pm 4.87\mu\text{m}$) respectively. The epithelium of the omasum is stratified squamous keratinized composed of four layers (basale, granulosum, spinosum, and corneum (Scocco et al. 2016). In present study omasum of Jattal and Nachi goat breeds had primary secondary and tertiary laminae which were named as I, II, III depending upon their sizes, Scocco et al (2016) reported same 3 orders of laminae. However, Scala et al. (2016) reported 5 types of laminae in lambs and 28 in buffalo calves. In contrast, four types of laminae have been reported in Baladi goats (Scala et al.

2016). The type I and II order laminae had the papillae. Few papillae were also observed in the III order laminae as reported in sheep (Scocco et al. 2016). The study on sheep omasum showed two types of papillae in the laminae: larger conical papillae that prevented the digesta from passing between the laminae allowing the final divided material to pass distally and smaller blunt papillae with a thin horny covering that were associated with water and electrolyte absorption (Chandrasekar et al. 2019). Other studies showed that in Baladi goat omasal laminae of many shapes were arranged in 4-5 orders and they also differed by their distribution and direction (Malik et al. 2011).

Conclusion

Gross anatomical features of forestomach in Nachi breed are greater than those of Jattal breed, which is in accordance with the larger size of Nachi breed. However, histomorphometric parameters are greater in Jattal than in Nachi breed, which is associated with greater functional ability of their forestomach.

Future Outcomes

This study will contribute to discriminate the morphology and histomorphometry of two goat breeds (Jattal and Nachi) from other analogous small ruminants. This study will also contribute to the body of scientific knowledge and provide the baseline anatomical and histological data for future research. It will help to correlate the structures with its expected functions.

References

1. Amit P, Pawan K, Parveen KJ. Histomorphological studies on the rumen of the sheep (Ovisaries). Haryana Veterinary Journal 2011; 50: 49-52.
2. Bancroft D, Maierl R, Lecchi T. Histological processing techniques. Cell Tissues Research 2013; 124(3): 3532-3845.
3. Beharka A, Nagaraja T, Morrill J, Kennedy G, Klemm RJ. Effect of form of diet on anatomical, microbial and fermentative development of rumen of neonatal calves. International Journal of Dairy Science 2018; 81(7): 1946-1955.
4. Chandrasekar V, Narayanan P, Lalitha P, Vijayaragavan C. Comparative gross anatomy of the omasum of sheep and goats. Small Ruminant Research 2019; 9(4): 377-388.
5. Clauss M, Hofmann R, Streich W, Fickel J, Hummel J. Convergence in the macroscopic anatomy of the reticulum in wild ruminant species of different feeding types and a new resulting hypothesis on reticular function. Journal of Zoology 2010; 281 (1): 26-38.
6. Chandrasekar V, Narayanan P, Lalitha P, Vijayaragavan C. Comparative gross anatomy of the omasum of sheep and goats. Small Ruminant Research 2019; 9(4): 377-388.
7. EL-Gendy SA, Derbalah ABS. Macroscopic and microscopic anatomy of the omasum of the Baladi goat. Journal of Applied Biological Sciences 2016; 4(3): 37-45.
8. Franco A, Masot J, García A, Redondo EJ. Ontogenesis of the Reticulum with Special Reference to Neuroendocrine and Glial Cells: A Comparative Analysis of the Merino Sheep and Iberian Red Deer. Anatomia, Histologia, Embryologia 2012; 41(5): 362-373.
9. Emam M, Abugherin B. Immunohistochemical Study on the Ruminal Wall of Adult Baladi Goats (Capra hircus). Small Ruminant Research 2016; 104(3): 3632-3645.
10. Endo H, Sasaki M, Kogiku H, Hayashi Y, Komiya T, Narushima E, Arishima K, Yamamoto MJMS. Anatomy and histology of the stomach in a newborn pygmy hippopotamus (Choeropsis liberiensis). Mammal Study 2011; 26(1): 53-60.
11. Garcia A, Masot J, Franco A, Gazquez A, Redondo E. Histomorphometric and immunohistochemical study of the goat reticulum during prenatal development. Small Ruminant Research Journal 2013; 437-448.
12. Jelínek K. Development of the Forestomach of the Goat (Capra aegagrus f. hircus) in the Postnatal Period. Acta Veterinaria Brno 2005; 64(1): 49-61.
13. Mahesh R, Singh G, Kumar PI. Light and scanning electron microscopic studies on the rumen of goat (Capra hircus). Veterinary Research International 2014; 2(3): 74-80.

14. Malik MI, Rashid MA, Yousaf MS, Naveed S, Javed K, Nauman K, Rehman HRR. Rumen morphometry and sorting behavior of fattening male goat fed pelleted and unpelleted TMR with two levels of wheat straw. *Small Ruminant Research* 2011;196: 106-116.
15. Membrive C. Anatomy and Physiology of the Rumen. *Rumenology* 2016; 1-38.
16. Mutoh K, Wakuri H. New glands found at the Omasal end in the goat. *Australian Veterinary Journal* 2020;64(6): 351-36.
17. Phillipson A. Physiology of digestion and metabolism in the ruminant. *International Journal of Physiology* 2018; 19(6):235-258.
18. Soni T, Goswami H, Panchal KJAiLS. Prenatal Development of Fore-Stomach in Small Ruminants. *Small Ruminant Research* 2016; 5: 10209-10215.
19. Soni T, Panchal KJ. Gross, biometrical and histological study of reticulum and oesophageal groove of Surti goat (*Capra hircus*). *Indian Veterinary Journal* 2018; 27(2): 21-27.
20. Suárez BJ, Van Reenen CG, Gerrits WJ, Stockhofe N, Van Vuuren AM, Dijkstra J. Effects of supplementing concentrates differing in carbohydrate composition in veal calf diets: II. Rumen development. *Journal of Dairy Science* 2006;89(11):4376-86.
21. Shoeib MB, Hassanin A, Elnasharty MV. Morphological and morphometric characteristics of gastric mucosa in western grey kangaroo (*Macropus fuliginosus*). *Journal of Advanced Veterinary and Animal Research* 2015; 2(1): 40-48.
22. Scocco P, Mercati F, Tardella FM, Catorci AJ, technique. Increase of forage dryness induces differentiated anatomical response in the sheep rumen compartments. *Microscopy Research and Technique* 2016; 79(8): 738-743.
23. Scala G, Corona M, Maruccio LJA. Structural, histochemical and immunocytochemical study of the forestomach mucosa in domestic ruminants. *Anatomia, Histologia, Embryologia* 2016; 40(1): 47-54.
24. Scott A, Gardner ICJ. Papillar form in the forestomach of the sheep. *Journal of anatomy* 2013; 116(2): 255.
25. Tamate H, McGilliard A, Jacobson N, Getty RJJods. Effect of various dietaries on the anatomical development of the stomach in the calf. *International Journal of Dairy Science* 2020; 45(3): 408-420.
26. Wasti E, Arslan F, Haseeb KL. Pakistan Economic Survey. *SMEDA Research Journal* 2019; 65(3): 908-1220.
27. Yamamoto Y, Atoji Y, Agungpriyono S, Suzuki YJ. Morphological study of the forestomach of the Japanese serow (*Capricornis capensis*). *Anatomia, Histologia, Embryologia* 2018; 27(2): 73-81.