Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2017; 5(6): 948-956

Received: 22-09-2017
Accepted: 25-10-2017

Luay O Hamza
Department of anatomy and histology College of Veterinary Medicine/ University of Baghdad, Iraq

Najlaa Awaied Al-Mansor
Department of anatomy and histology / college of veterinary medicine/ University of kufa, Kufa, Iraq

Correspondence
Luay O Hamza
Department of anatomy and histology College of Veterinary Medicine/ University of Baghdad, Iraq

Histological and histochemical observations of the small Intestine in the indigenous Gazelle (*Gazella subgutturosa*)

Luay O Hamza and Najlaa Awaied Al-Mansor

Abstract
The investigation of histological and histochemical characters of the small intestine in indigenous Gazelle was carried out during November into April 2017. The tissue samples were processed by paraffin technique and stained with the H and E, Masson's trichrom, PAS and Alcian blue stains. In duodenum the submucosa occupied by mucus and serous Brunner's glands. The jejunum had short villi with few goblet cells. The ileum had furthermore of goblet cell with submucosal payer's patches. The goblet cells of duodenum were positive with Alcian blue (2.5pH) stain. The Brunner's glands were weak PAS and negative with Alcian blue. In jejunum, the goblet cells of crypt were moderately and strongly with PAS and AB (pH2.5) respectively. In the ileum the goblet cells were more than the neutral in the crypts. This work concluded that the small intestine of Indigenous gazelle was similar those of small ruminant.

Keywords: Histology, histochemical, gazelle, duodenum, jejunum, ileum

Introduction
The digestive tracts in various species of mammals adapted to the most efficient use of the feed they consume [1]. The digestive tract is an essential in living organisms, and assumes a fundamental role in food processing and absorption [2, 3]. reported that the microscopic section of small intestine is contained a numerous "finger-like" projections known as villi, that increase the intestinal surface area to assists in nutrient absorption. The mucosal epithelium of small intestine was consisted of columnar cells. The mucosa was rich in lymphoid follicles, which aggregate to form Payer's patches in ileum [4]. The crypt was formed in the epithelium of small intestine by invagination of the villi which known as crypts of Lieberkühn that appeared more numerous and smaller than the villi. In the base of crypt has a lysozyme and cryptdin-producing Paneth cell [5]. The goitered or black-tailed gazelle (*Gazella subgutturosa*) was a widespread species, free living in northern Azerbaijan, eastern Georgia, some portion of every Iran, Iraq and southwestern Pakistan, southeastern Turkey, Afghanistan and the Gobi Desert [6] in Iraq *Gazella subgutturosa* has saved in several nature reserves located in many provinces including, the district of AL Madaen in the outskirts of the capital Baghdad, an area of (157) acres, where ksiab-reservoir to save the species and varieties from the risk of Extinction [7]. The present study was aimed to investigate the microscopic character and special histochemical identification for staining reaction to the cells and gland in each part of small intestine in the indigenous gazelle *Gazella subgutturosa*.

Materials and Methods
Six animals were obtained from (AL-Madaen Animal Reservoir) in Baghdad-Iraq, and the study was conducted in department of anatomy, histology in college of veterinary medicine-university of Baghdad, during a period extended from November into April 2017. After animal slaughtering the samples of small intestine were extirpate gently and washed up to get scour of any remnant, specimens from three parts of small intestine (Duodenum, jejunum and ileum) were taken and fixed in Bouin's solution for 48 hours [8]. The specimens were processed upgrading with ethanol alcohol for paraffin technique and sectioned serially at (5-6) µm. The prepared sections were stained with the Hematoxylin and Eosin, Masson's trichrom, PAS and Alcian blue stains [9, 10]. The histological observations were pictured by using the color digital camera [11].

Results and Discussion

Histologically Duodenum; had composed of four tunicae: mucosa, submucosa, muscularis and serosa (Fig. 1). The mucosa had finger like projections (villi) which heavy and crowded and lined by simple columnar epithelium, the lamina propria of each villus consisted of thin layer of cellular loose connective tissue, while the beneath villi was slightly thin layer contained crypts of Lieberkühn which contained the Paneth cells, these cells were occupied with acidophilic cytoplasmic granules. The muscularis mucosa was consisted of two layers of smooth muscle fibers (Fig. 2, 3). Similar findings were recorded in Dromedary Camels (Camelus dromedaries) by Althnain [12] and in the Persian squirrel (*Sciurus anomalus*) by Tootian [13]. But, the present result disagreed with [14] and [15] who noticed that there was not Paneth cell have been seen the epithelial villi. The present study finding revealed that the Brunner's glands was the most characteristic features observed in the duodenum, which appear numerous and extending to the plica cirrcularis, these glands were branched tubule-alveolar contained mucus & serous alveoli (Fig. 1, 2). These observations were similar to many previously finding by [16, 17, 18, 19. 20, 21]. The current study was disagreed with another study in in goat by [22] who mentioned that there were extensive mucous observed in the duodenum and numerous goblet cells due to the absence of Brunner's glands in the sub mucosa of duodenum. This suggests that these differences could be due to the type of food especially the rough, dry food consumed by goat. Current finding revealed that, the tunica muscularis composed of two layers; the inner thick circular smooth muscle fibers, and the thin longitudinal smooth muscle fibers, between them there was thick layer of collagen bundles that housed Auerbach's (Myenteric) nerve plexuses. The tunica serosa was a thick layer of loose connective which covered by mesothelium (Fig. 1 and 4), agreed with previously finding of the Opossum by El-Sayed and Alina [23, 24]. Intra mural common bile duct and duodenal papillae had wide lumen, which composed of tunica mucosa, sub mucosa and muscularis, the mucosa had very tall simple mucosal folds which lined with simple columnar epithelium that invaded with goblet cells and the lamina propria displayed groups of sub mucosa tubuloalveolar seromucous glands (Fig. 5 & 6). This result was agreed with [25] in small ruminant and [26] in indigenous buffalo. This observation revealed that the duodenal papillae have the same structure of duodenum except that it had thickest tunica muscularis (Fig. 5). These results was agree with Riches [27] in rat, but disagree with [28] in mouse, have been revealed that the epithelial lining the duodenal gland were shorter (cuboidal or columnar). Jejunum had the same structure of duodenum except that the tunica mucosa consist of short villi and lamina propria beneath villi was thick layer contained the secretory units of crypts of Lieberkühn which were small in size lined by simple columnar cells (Fig. 7 & 8). Similar observation was mentioned by Hassan [22] in goat and Kumar [29] in sheep. The tunica submucosa of jejunum have not seen Brunner's glands along its entire length except the anterior part of jejunum that show the presence the submucosal Brunner's glands (Fig. 9). This result was parallel to the observation in pigs and large herbivores by Verdiglione [30], in rabbit by Ergun [31] whom noticed that the duodenal sub mucosal glands were distributed to begin from the pyloroduodenal junction and to reach closer to the jejunum. Ileum**:** the pilica cirlcularis was little and the villi were short and leaf like projections and furthermore showed goblet cells (Fig. 10 and 11). These results were incompatible with Charlotte [32] in the African giant rat that showed few goblet cells in the short villi of ileum. Many previously result by Constantinescu [33] and Kumar [34] agreed with the recent result which revealed that the tunica submucosa was characterized in the ileum by the present of Peyer's patches (Fig. 10), in the ox it arranged as irregularly raised plaques or bands, varying in length from a few millimeters to more than 25 cm. while in horse and ruminants were relatively well developed and extend as a large bowel. In the pig, these patches were present in the jejunum and in the ileum. The duodenal histochemical results showed the goblet cells within epithelium of villi were positively reacted with Alcian blue (2.5pH) stain and the secretory cells within epithelial crypts were giving positive reaction for both PAS and Alcian blue (2.5pH). On other hand some secretory cells were containing both positive PAS and AB granules within their cytoplasm and were been mixed type of secretory units, the population of acidic reaction secreting cells were predominating than that of neutral reaction (Fig. 12). The present study disagreed with another previous result of [13] in the Persian squirrel, which revealed that the goblet cells of the small intestine were PAS-positive, and appeared red in color owing to the presence of neutral mucin. The secretory products of Brunner's glands showed weak reaction with PAS stains and negative for Alcian blue stain (Fig. 13). These findings were incompatible with the result of Andleeb [35] in Gaddi goat who reported that the secretion of Brunner's glands in showed PAS positive reaction with Alcian blue. This observation may be due to species difference and type of food that consuming by indigenous gazelle. In jejunum the stain's reactions revealed the presence of neutral and acidic mucin in the goblet cells of crypts which were moderately and strongly reacted with PAS (Fig. 14). These finding were in opposite with previous study of [35] in Gaddi goat, [13] in Persian squirrel, [20] in African giant rat and [29] in sheep. In ileum the acidic goblet cells of ileal villi and crypts showed the presence apparently acidic mucin more than the neutral (Fig. 15). Similar findings have been reported in sheep (*Ovis Aries*) by Kumar [34]. But incompatible with Andleeb [35] in Gaddi goat, the crypts showed a moderate reaction with PAS and a mild reaction with Alcian blue stain (pH 2.5) showing less amount of both sulphated and non-sulphated mucin. These results may be due to the type of food consuming by the indigenous gazelle.

Fig 1: Histological section of duodenum shows: tunica mucosa (Tm), Tunica submucosa (Sb), Bruner's gland (Bg), Inner circular (IL), myenteric plexus (red arrow) and Outer longitudinal layers (OL) of tunica muscularis, tunica serosa (Ts) and villi (arrows). H&E 40X.

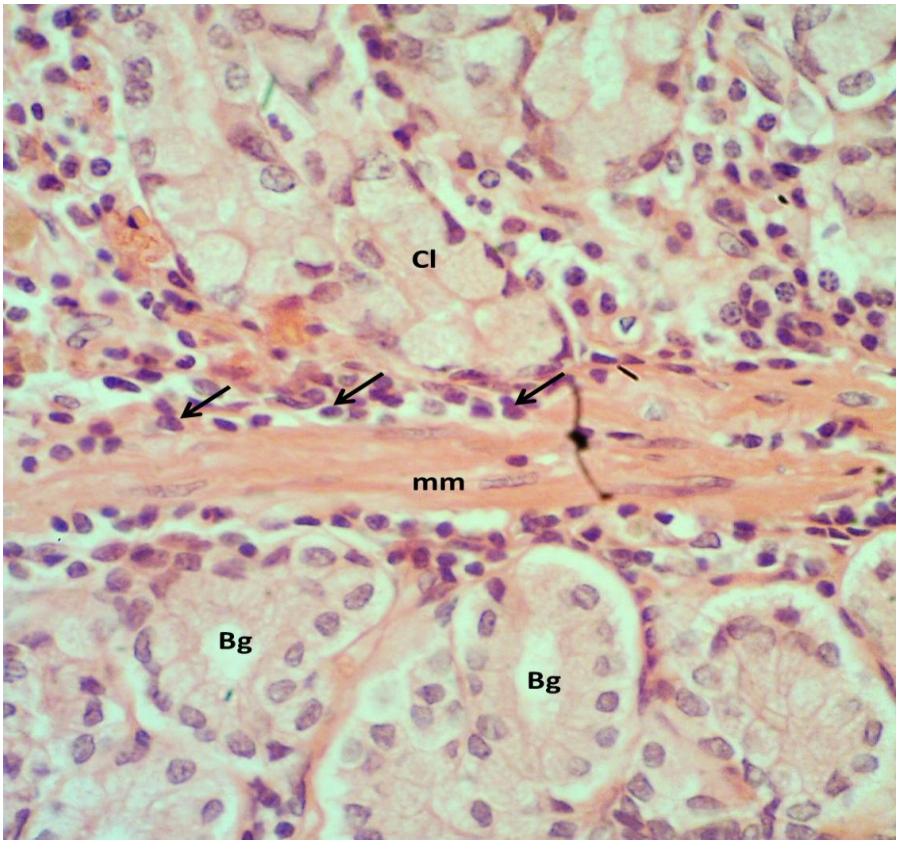

Fig 2: Magnified histological section of duodenum shows: muscularis mucosa (mm), crypt of Lieberkühn (Cl), Bruner's gland (Bg) and lymphocyte (arrows). H&E 400X.

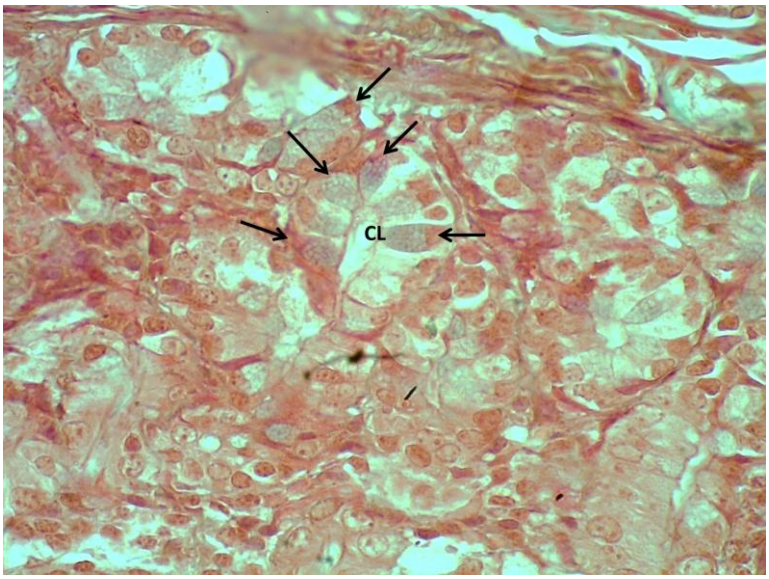

Fig 3: Magnified histological section shows: Paneth cell (arrows) and crypt of Lieberkühn (CL). Masson's trichrom stains 400X.

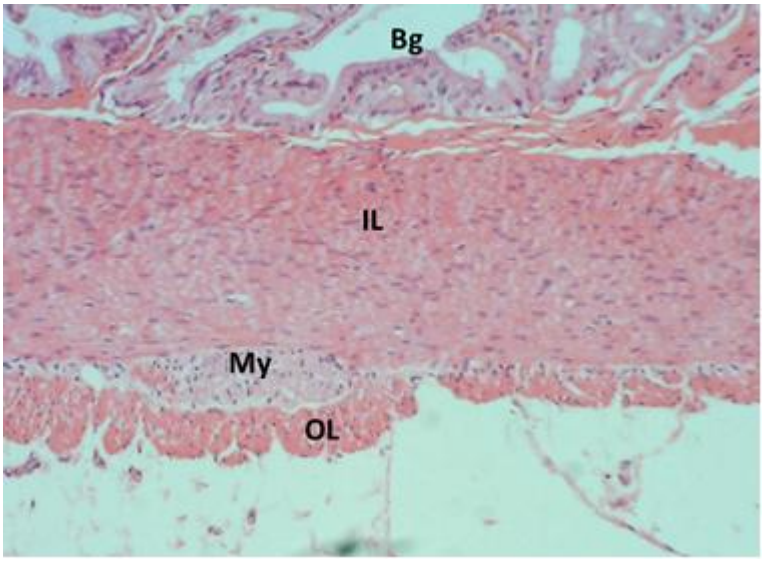

Fig 4: Histological section of wall of duodenum shows: Bruner's gland (Bg), myenteric plexus (My) and Inner circular (IL) and Outer longitudinal layers (OL) of tunica muscularis. H&E 100X.

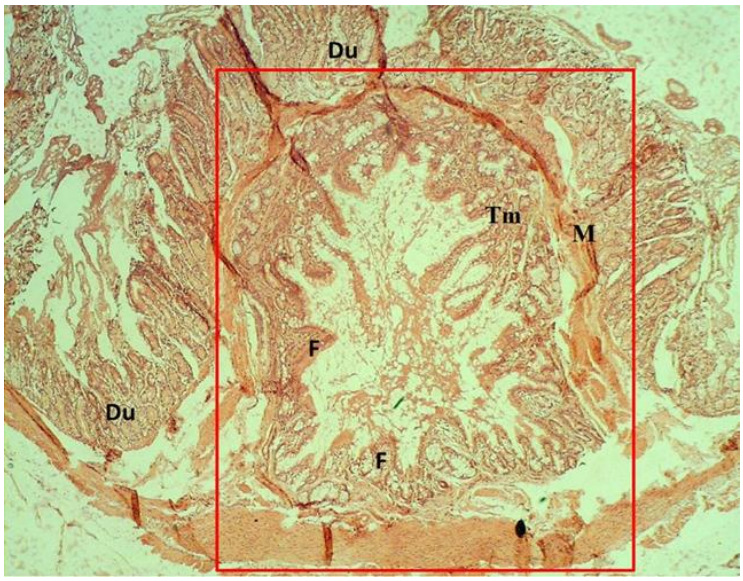

Fig 5: Section of intra mural common bile duct: shows; common bile duct (red square area), duodenum (Du), tunica mucosa (Tm), tunica muscularis (M) & folds of mucosa (F) H&E stains 40X.

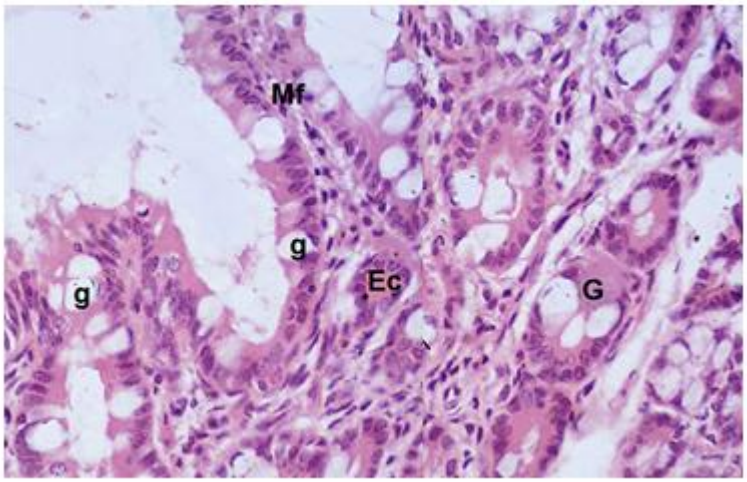

Fig 6: Magnified section of mucosa of intra mural common bile duct: shows: epithelial crypts (Ec), goblet cells (g), mucosal fold (Mf) and sub mucosal tubulo-alveolar glands (G). H&E stains 400X.

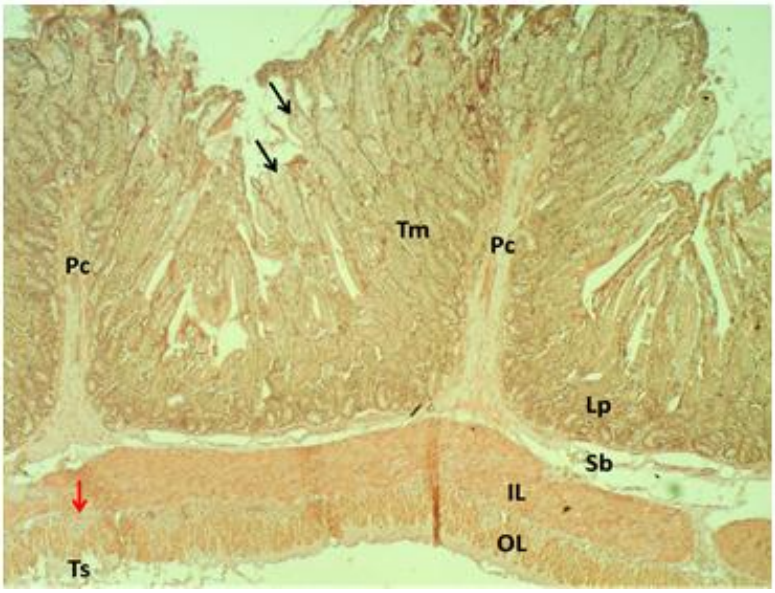

Fig 7: Histological section of jejunum shows: tunica mucosa (Tm), plica cirrcularis (Pc), lamina propria (Lp), tunica submucosa (Sb), Inner circular (IL) and Outer longitudinal layers (OL) of tunica muscularis, myenteric plexus (red arrow) and tunica serosa (Ts). H& E stains 4X.

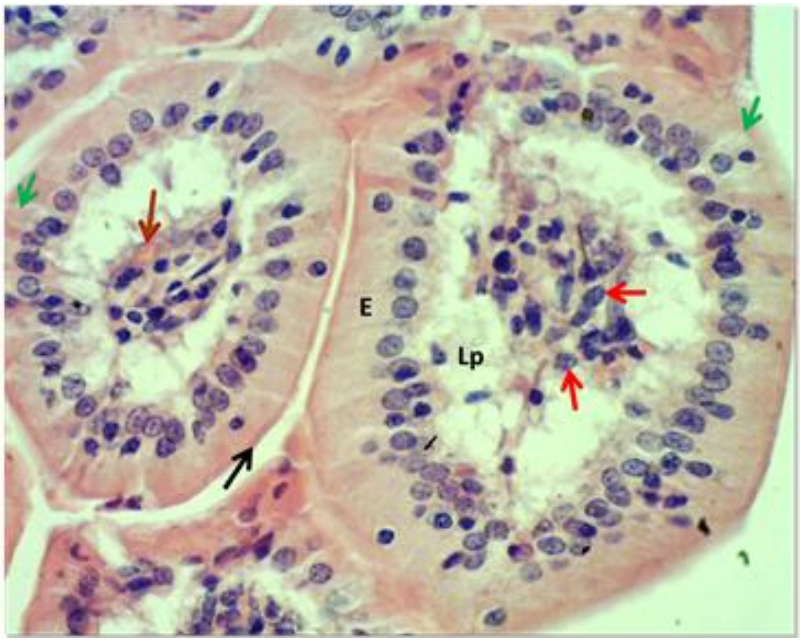

Fig 8: Magnified transverse section of jejunal villi shows: epithelium (E), lamina propria (Lp), and lymphocytes (red arrow), line of demarcation represent microvilli (black arrow), lacteal blood vessels (brown arrow) and goblet cells (green arrow). H&E 400X.

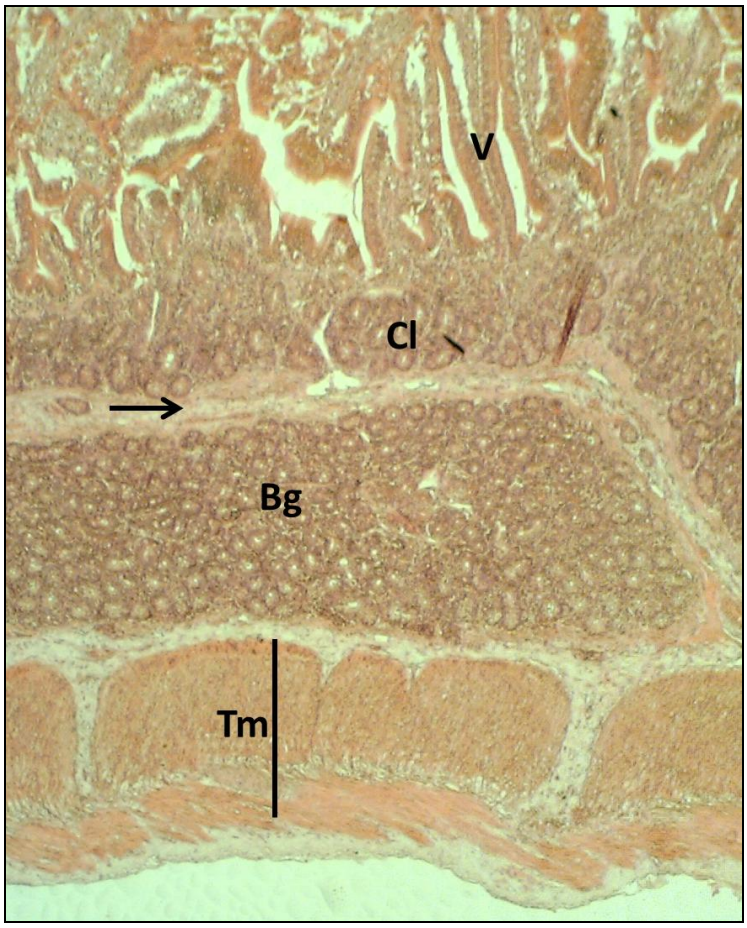

Fig 9: Histological section of anterior part of jejunum shows: villi (V), crypt of Lieberkühn (Cl), muscularis mucosa (arrows), Brunner's gland (Bg) and tunica muscularis (Tm). H&E. 40X.

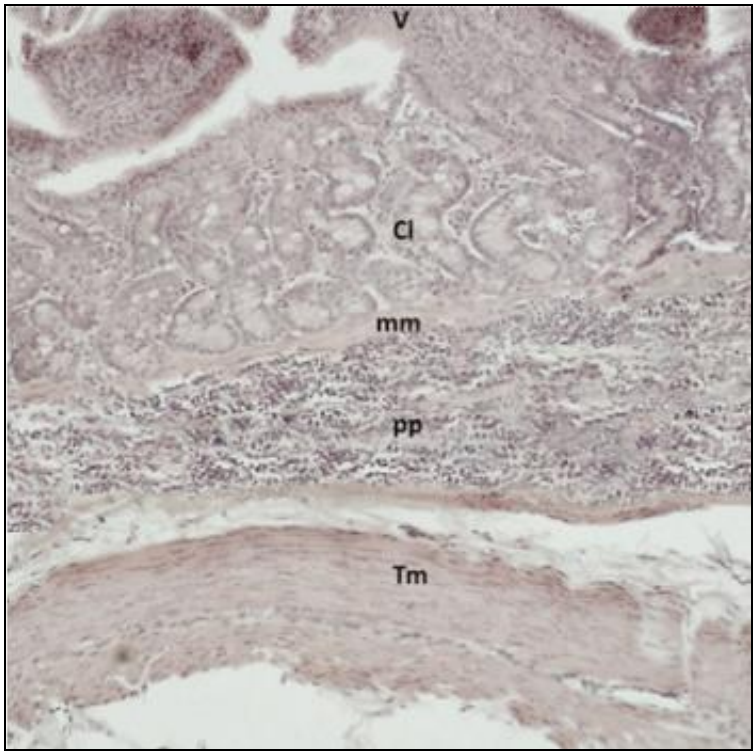

Fig 10: Histological section of ileum shows: villi (V), crypt of Lieberkühn (Cl), muscularis mucosa (mm), payer's patches (pp) and tunica muscularis (Tm). H&E. 40X.

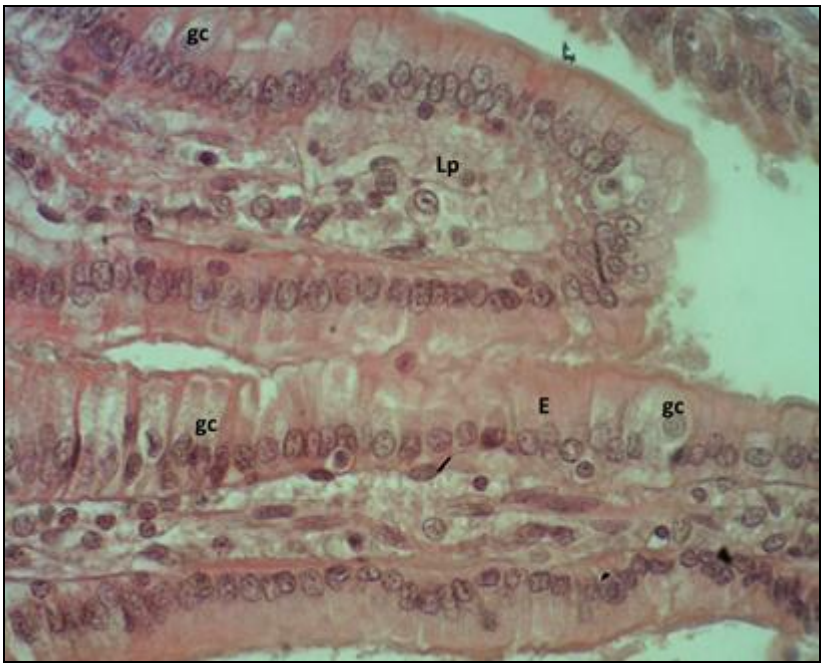

Fig 11: Magnified histological section of ileum villi shows: goblet cells (gc), epithelia (E) and lamina propria (Lp), H&E. 400X.

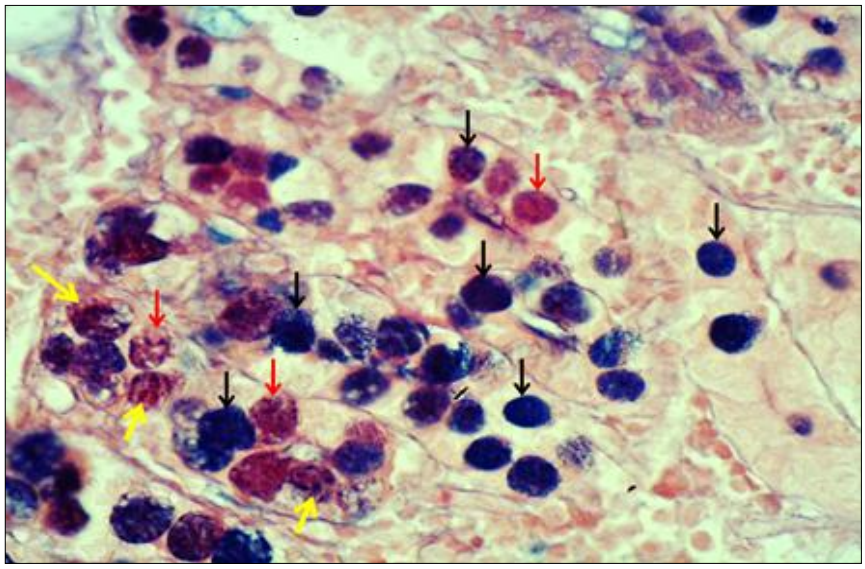

Fig 12: Magnified histochemical section of crypt of Lieberkühn (duodenum) shows: positive AB (2.5pH) (black arrows) and positive PAS stains (red arrows) & mixed secretory cell (yellow arrow). Combined AB & PAS stains 400X.

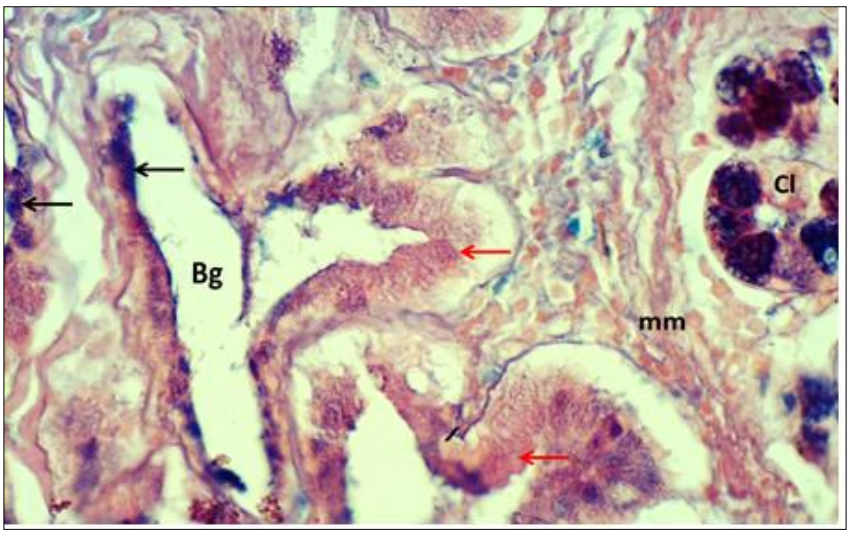

Fig 13: Section of Brunner's gland shows: weak positive PAS stains (red arrows) and negative for AB (2.5 pH). Combined AB & PAS stains 400X.

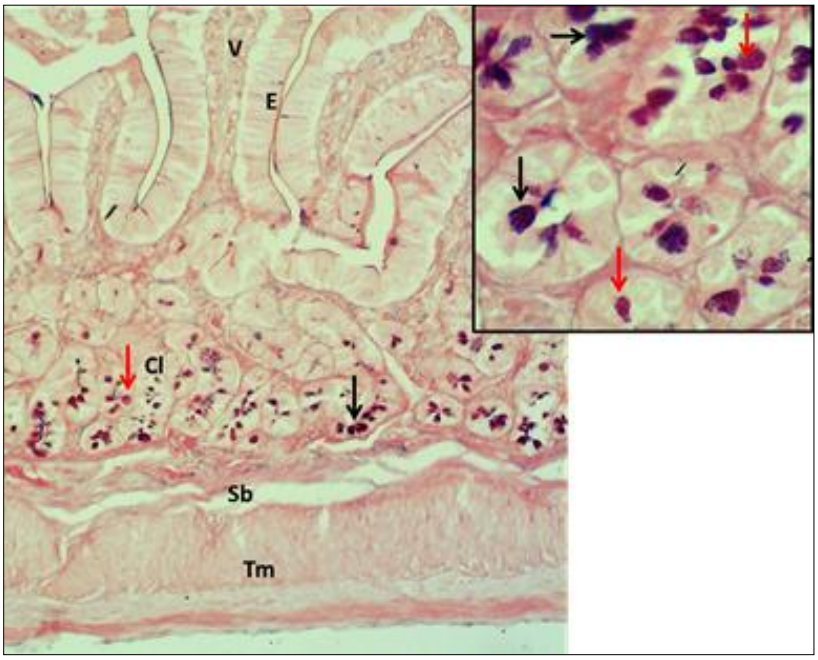

Fig 14: Section of jejunum shows: villi (V), epithelia (E), crypt (Cl), tunica submucosa (Sb), tunica mucosa (Tm), positive PAS stains (red arrows), positive AB (2.5 pH) (black arrow), Combined AB & PAS stains. 40X and magnified section 400X.

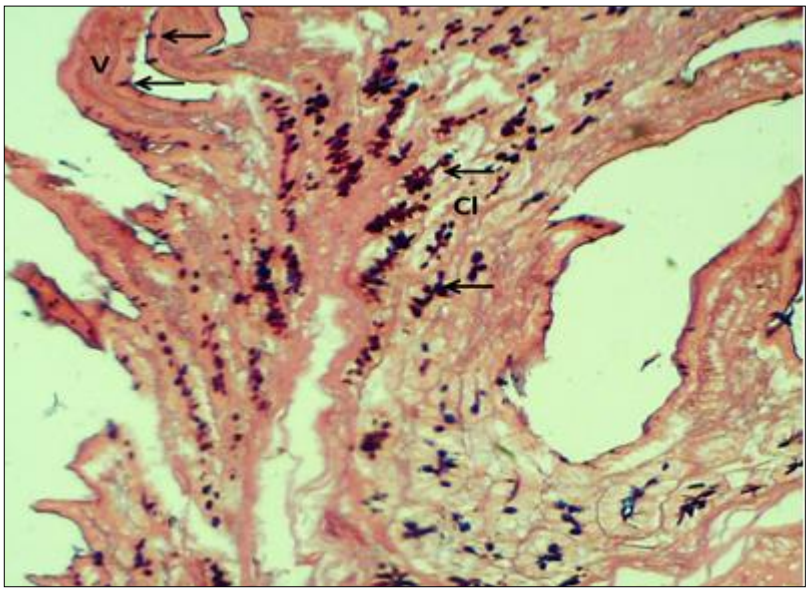

Fig 15: Section of ileum shows: villi (V), crypt (Cl), tunica submucosa, predominant positive AB (2.5 pH) stains of goblet cells (black arrows). Combined AB & PAS stains 40X.

Conclusion

The present experimental results conclude that the histological observation had the same structure with other animals except that the Brunner's glands in duodenum appeared as two types (mucus & serous) branched tubule-alveolar glands. The muscularis mucosa in jejunum was consisted of (3-4) layers of smooth muscle fibers. While histochemicaly, it was characterized by positive reaction of the goblet cell with Alcian blue (2.5pH) stain in duodenum, while moderately and strongly reacted with PAS and AB (pH2.5) in jejunum, in ileum showed the presence apparently acidic mucin in the goblet cells.

Acknowledgement

Author would like to acknowledge all technical staff your assistances in department of Anatomy, Histology &Embryology-College of veterinary Medicine at University of Baghdad.

References

1. Stufflebeam CE. Principles of Animal Agriculture. Englewood Cliffs, NJ: Prentice Hall, Inc. 1983.
2. Hill RW, Wyse GA, Anderson M. Animal Physiology. 2nd Ed., Sinauer Associates, Inc. 2008, 762.
3. Parish J. Ruminant Digestive Anatomy and Function. Beef Production Strategies. 2011.
4. Junqueira LC, Carneiro J. Organs associated with the digestive tract. In: Basic histology. Text & Atlas10th Ed., McGraw-Hill Publishing Co, Starnford. 2003, 332-344.
5. Karam SM. Lineage commitment and maturation of epithelial cells in the gut. Front. Biosci. 1999; (4)286-298.
6. Murtskhvaladze M, Gurielidze Z, Kopaliani N, Tarkhnishvili D. Gene introgression between Gazella subguturrosa and G. marica: limitations of maternal inheritance analysis for species identification with conservation purposes. Acta. Theriologica. 2012; DOI 10:1007.
7. Abood DA, Hussein ZM. Histological features of Vomer

nasal organ in Indigenous Gazelle (*Gazella subgutturosa*). Journal of Entomology and Zoology Studies. 2017; 5(4):598-604.
8. Carson FL, Hladik Ch. Histotechnology. A Self-Instructional Text. Third Ed., Chicago ASCP Press, 2009, 400.
9. Vacca LL. Laboratory Manual Histochemistry, raven press. Book, Ltd, New York. 1985.
10. Bancroft JD, Marilyn G. Theory and practice of histological technique 6th Ed., Elsevier Limited, London. 2008, 168-173.
11. Kiernan JA. Histological and histochemical methods. 4th Ed., Scion Publishing Ltd. 2008, 606.
12. Althnain TA, Alkhodair KM, Albokhadaim IF, Abdelhay M, Homeida AM, El-Bahr S. Histological and Histochemical Investigation on Duodenum of Dromedary Camels (*Camelus dromedarius*). Science International, 2013; 1(6):217-221.
13. Tootian Z, Sadeghinezhad J, Taghi Sheibani M, Fazelipour S, De Sordi N, Chiocchetti R. Histological and mucin histochemical study of the small intestine of the Persian squirrel (Sciurus anomalus). Anatomical science international, 2013; 88:38-45.
14. Ahmed YA, El-Hafez AAE, Zayed AE. Histological and Histochemical Studies on the Esophagus, Stomach and Small Intestines of Varanus niloticus. Journal of veterinary anatomy. 2009; 2(1):35-48.
15. Ergun E, Ergun L, Asti RN, Kurum A. Light and scaning electron microscopyof the paneth cells in the sheep small intestine. Revue de médecine vétérinaire, 2003; 154(5):351-355.
16. Oduor-Okelo D. Histochemistry of the duodenal glands of the cat and horse. Acta. Anatomica, 1976; 94(3):449-456.
17. Pfeiffer CJ, Dabareiner RM. Ultrastructure of Brunner`s glands in the horse. Journal of Submicroscopic Cytology and Pathology. 1992; 24:581-588.
18. Takehana K, Abe M, Iwasa K, Hiraga T, Miyata H. Carbohydrate histochemistry of bovine duodenal glands. The Journal of Veterinary Medical Science, 1991; 53:699-706.
19. Takehana K, Ueda Eerdunchaoluo H, Kobayashi A, Iwasa K, Sou K. A histochemical study of the camel (Camelus bactrianus) duodenal glands. The Journal of Veterinary Medical Science. 2000; 62:449-452.
20. Nzalak JO. Anatomical and Histochemical Studies of the Digestive System of the African Giant Rat (Cricetomys gambianus-Waterhouse). Ph.D. Thesis, Postgraduate School, Ahmadu Bello Universityphilosophy Veterinary Anatomy. Zaria, 2010.
21. Nzalak JO, Wanmi N, Imam J, Ali M, Umosen AD. Anatomical and Histological Studies of the Small Intestine of the African Giant Rat (Cricetomys gambianus-Water house) - II. Annals of Experimental Biology, 2015; 3(4):20-26.
22. Hassan AS, Moussa EA. Light and scanning electron microscopy of the small intestine of goat (Capra hircus). Journal of cell and animal Biology. 2015; 9(1):1-8.
23. El-Sayed H. Veterinary Histology. The Science of Cells and Tissues. Faculty of Veterinary Medicine, Mansoura University, Egypt, 2006.
24. Alina M, Codruța I, Cebzan C, Niculescu M, Niculescu V, Matusz P *et al*. Enteric nervous system development in cavitary viscera allocated to the celiac plexus Romanian. Journal of Morphology and Embryology. 2008; 49(1):63-67.
25. Al-Mayahi MS. Anatomical, radiological and histological study of the gall bladder and biliary duct system (extra and intra hepatic) in small ruminant. MSc thesis veterinary medicine college, University of Baghdad (anatomy and histology), 2008.
26. Al-Rekabi AS. Anatomical, radiological and histological study of the gall bladder and biliary duct system (extra and intra hepatic) in indigenous buffalo (*Bubalus bubalis*). MSc thesis, veterinary medicine college, University of Baghdad (anatomy and histology), 2011.
27. Riches DJ. Ultra-structural observation on the common bile duct epithelium of the rat. Journal Anatomica, 1972, 157-170.
28. Yamada K. The gland of the common bile duct in rat and mouse an electron microscope study. Acta Anatomica, 1970; 77:438-1453.
29. Kumar P, Kumar P, Singh G, Poonia A, Parkash T. Histological architecture and histochemistry of jejunum of sheep (*Ovis Aries*) Haryana, 2014; 53(1):55-57.
30. Verdiglione R, Mammola CL, Filotto U. Glycoconjugate histochemistry of bovine Brunner glands. Annals of Anatomy, 2002; 184:61-69.
31. Ergun E, Ergun L, Ozen A, Kurum A, Bayraktaroglu AG. Histomorphology of the Brunner gland in the Angora rabbit. Journal of Animal and Veterinary Advances. 2010; 9(5):887-891.
32. Charlotte LO. Veterinary Histology VMED 7123, In online lecture notes. 2004, 1-17.
33. Constantinescu GM. Guide to regional Ruminant Anatomy based on the Dissection of the Goat. Ames: Iowa State University Press, 2001.
34. Kumar P, Kumar P, Singh G, Poonia A, Parkash T. Histoarchitecture and histochemistry of the ileum of sheep (*Ovis Aries*). Haryana Vet. 2015; 54(1):50-52.
35. Andleeb RR, Bhardwaj RL, Sharma KB. Histochemical studies on the small intestine of Gaddi goat. Indian journal of animal physiology. 2009; 2:75-78.