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The Histological Study of the Ileum in the Caucasian Squirrel *Sciurus anomalus*

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Abstract

The current study was designed to identify the histological composition of the ileum in the Caucasian squirrel *Sciurus anomalus*. Ten adult squirrels were used for the present study. Animals were anesthetized, anatomized, fixed the study specimens in 10% formalin solution, and conducted a series of histological staining techniques. The ileum consists of four main tunicae represented by mucosa, submucosa, muscularis, and serosa. The tunica mucosa consists of three secondary layers: the epithelial lining layer, lamina propria, and muscularis mucosa. The epithelial lining comprises simple columnar epithelial tissue composed of columnar cells and goblet cells. The lamina propria contains simple straight tubular glands called the intestinal glands or the crypt of Lieberkuhn. The muscularis mucosa is composed of a very thin, single layer of smooth muscle fibers arranged circularly. The submucosa consists of dense connective tissue, while the tunica muscularis appears composed of two layers of smooth muscle fibers. The tunica serosa consists of a loose connective tissue that is covered by a mesothelium.

Keywords: Histological study, Ileum, Caucasian squirrels.

1.Introduction

Caucasian squirrel (*Sciurus anomalus*) (Gmelin,1778), the subject of the current study, belongs to the Sciuridae family of Rodentia, and it appears covered by thick fluffy hair, soft fur, and reddish brown or chestnut color. It is gray-brown on the dorsal side[1]. Some studies dealt with the gastrointestinal tract in various vertebrates. Dos Santos *et al.* [2] studied the gastrointestinal tract's morphology, histology, and histochemistry in *Trachelyopterus striatalus*. Jedeje *et al.* [3] presented an anatomical study of the gastrointestinal tract in the striped stood snake (*Psammophis sibilans*). Sauer *et al.* [4] examined an anatomical study of the gastrointestinal tract of two types of antelope, black buck (*Antilope cervicapra*) and Arabian sand Gazella (*Gazella subgutturosa marica*). Ahmed and Mnati [5] worked in the histological and histochemical study of the duodenum in the Caucasian squirrel (*Sciurus anomalus*). The small intestine has many functions. It digests and absorbs nutrients and many enzymes that aid digestion [6]. The small intestine consists of three main parts: duodenum, jejunum, and ileum [7]. This study aimed to identify the histological structure of the ileum in the Caucasian squirrel.

2.Materials and Methods

Samples collection:

The current study required the collection of 10 live adult squirrels, regardless of their sex, they were purchased from the Al-Gazal market in Baghdad, then euthanized, and dissected. The specimens of the ileum were collected and fixed in 10% formalin; then, the histological staining techniques were conducted.

Histological preparation

For the histological study, the samples were fixed with 10% formalin solution for 48 hours and then passed through an ascending series of ethyl alcohol (70, 80, 90, 100%) embedded with paraffin wax, cut blocks of 6 micrometers thickness, and stained with Hematoxylin-Eosin stain. Then, we examined and photographed selected sections using a compound light microscope with a camera [8].

Results

The results of the histological study in the Caucasian squirrel *Sciurus anomalus* showed that the ileum wall consisted of four main tunicae divided from the inside to outside (mucosa, submucosa, muscularis, and serosa) (**Figure 1**).

Tunica Mucosa

The tunica mucosa consists of three secondary layers: the epithelial lining, the lamina propria, and the muscularis mucosa, the epithelial lining consists of a simple columnar epithelial tissue composed of columnar cells and goblet cells, and the outer surface of the columnar cells appeared with a striated border due to the presence of the microvilli (**Figure 2**), the epithelial lining of the ileum part transforms into many projections known as villi and that show in different shapes, they may look leaf-shaped and shorter and with pointed ends or rounds, while some of them appear cylindrical, and a little mace shapes(**Figure 3,4**). Columnar cells are characterized by long compact cells with cytoplasm and large oval nuclei located near the base of the cell (hyponucleated) and rest on the basal membrane and the apical end of these cells with a striated border (**Figure 2**). The goblet cells are characterized by a unicellular gland that appears scattered between columnar cells with apical extension and a narrow base with an oval or flat-shaped nucleus close to the cell's base. They are many numbers in this part (**Figure 2**).

The lamina propria consists of loose connective tissue interspersed with many connective tissue nucleus cells, smooth muscle fibers, blood vessels, and lymphatic vessels (**Figure 3**). The epithelial lining at the base of the villi forms glandular units known as the intestinal glands. The intestinal glands spread in the lamina propria layer, appear in the form of simple straight tubular glands, and extend to the muscularis mucosa layer. The wall of the intestinal glands is formed by some cells represented by columnar cells and goblet cells (**Figure 5**).

In addition, another type of cell is undifferentiated cells (stem cells), concentrated in the basal part of the intestinal glands. They are located adjacent to or between the Paneth cells, which are close to the shape of columnar cells with pale cytoplasm and a spherical or oval-shaped base-located, euchromatin, surrounded by a clear nuclear envelope. These three types of cells are based on the basal membrane (**Figure 5**).

The intestinal glands of the ileum are characterized by some pyramidal cells, with a broad base and narrow towards the apical. The nucleus is characterized by an oval to a spherical shape. Its presence was noted in abundance in the basal and cervical area of the crypts and lack of presence in the villi. The cytoplasm of the apical area contains rough spherical granules with acidic secretion represented by Paneth cells (**Figure 5**). While the muscularis mucosa layer appears composed of a very thin and single layer of smooth muscle fibers arranged circularly, this layer is located directly under the lamina propria. It extends vertically within the villi and between the units of the intestinal glands as it separates the lamina propria from the tunica submucosa under it (**Figure 5**).

Tunica Submucosa

Tunica submucosa is composed of dense connective tissue and contains bundles of collagenous fibers, which also represent the dominant type of connective tissue fibers. This tunica contains smooth muscle fibers and is interspersed with many blood vessels, lymphatic vessels, and Meissner's plexuses, which appear scattered during this tunica (**Figure 5, 6**).

Tunica Muscularis

This tunica consists of two secondary layers of smooth muscle fibers, as the inner layer fiber is formed circularly. The circular inner layer looks thicker than the longitudinal outer layer. In contrast, the outer layer fiber is longitudinally arranged and appears in irregular muscle bundles. A thin strip of fibrous connective tissue separates between the two layers containing Aurebach's plexuses, ganglion cells, collagenous fibers, and a few elastic fibers (**Figure 6**).

Tunica Serosa

It represents the last tunica and is composed of loose connective tissue covered from the outside by a single row of simple squamous epithelial tissue. Many blood vessels and clusters of adipose tissue are scattered (**Figure 7**).

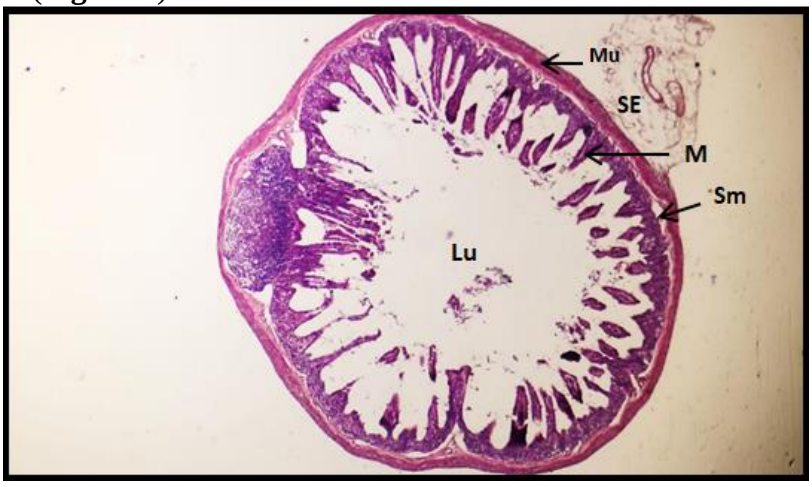

Figure 1. Cross section in the wall of the ileum shows the four tunicae: mucosa (M), submucosa (Sm), muscularis (Mu), serosa (Se), lumen (Lu), (H&E stain, 4x).

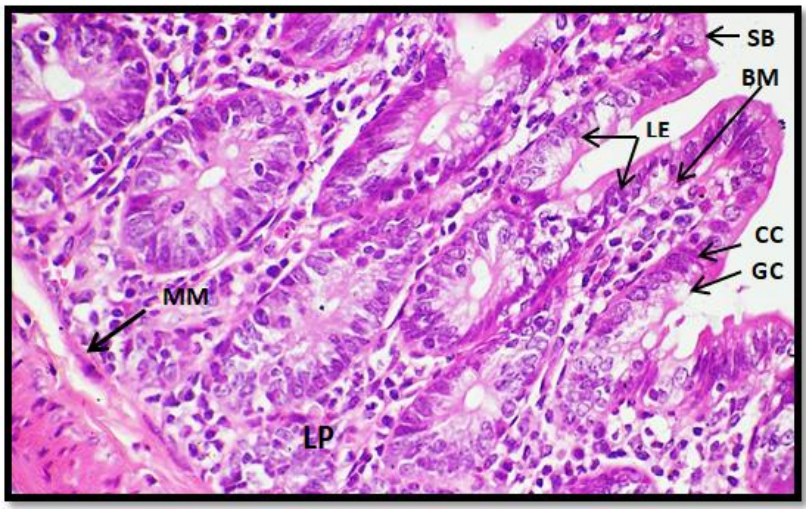

Figure 2. Longitudinal section in the wall of the ileum shows secondary layers that component the tunica mucosa: lining epithelial (LE), lamina propria (LP), Muscularis mucosa (MM), and component lining epithelium: Columnar cell(CC), Goblet cells (GC), Basal membrane(BM), Striated border (SB) (H&E stain, 40x).

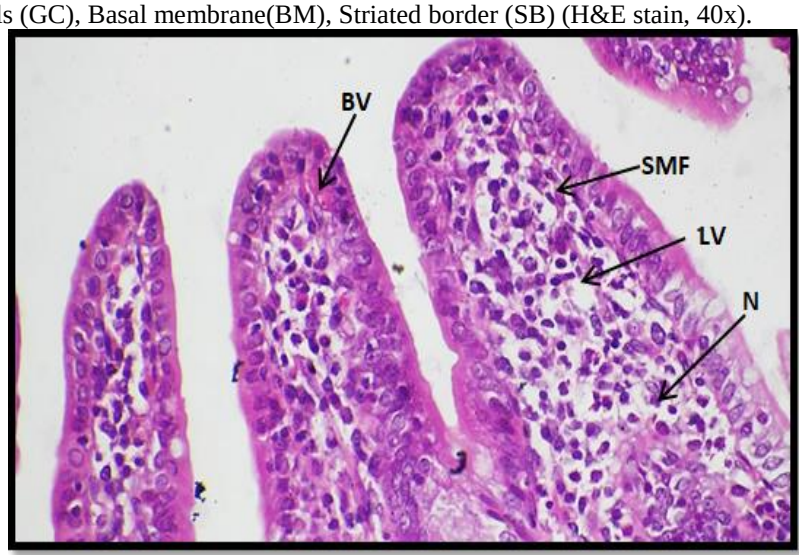

Figure 3. Cross section in the wall of ileum shows the leaf shape of villi and components of the lamina propria, nuclei of connective tissue cell (N), smooth muscle fiber (SMF), blood vessels (BV), lymphatic vessels (LV) (H&E stain, 40x).

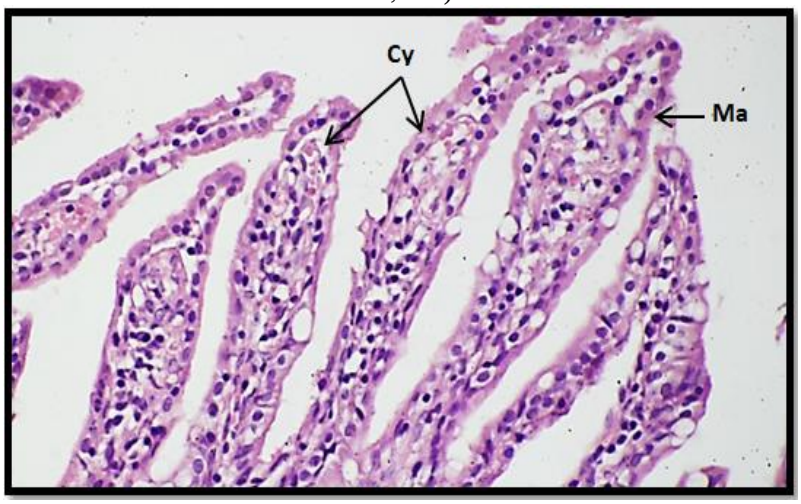

Figure 4. Cross section in the wall of the ileum shows the cylindrical shape (Cy) of villi and Mace shape(Ma) of villi) (H&E stain, 40x).

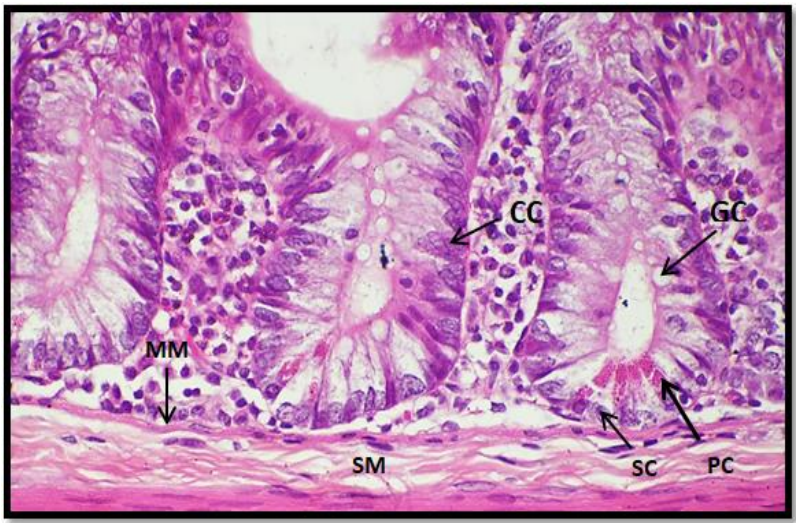

Figure 5. Cross section in the wall of ileum show intestinal glands constituent cells, Columnar cell (CC), goblet cell (GC), Stem cell (SC), Paneth cell (PC), Musculares mucosa (MM), Tunica submucosa (SM) (H&E stain, 40x).

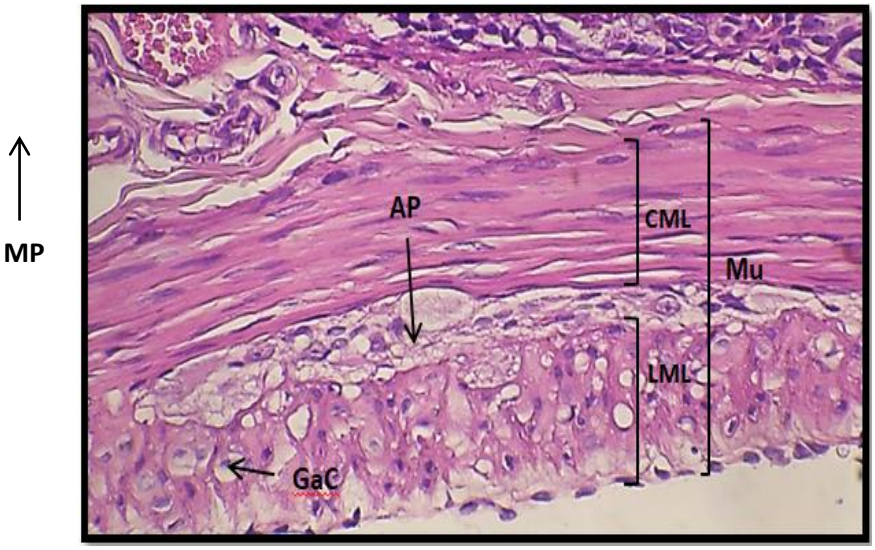

Figure 6. Cross section in the wall of the ileum shows tunica summucosa (SM), tunica muscularis (Mu) with two parts, circular muscular layer (CML) and longitudinal muscular layer (LML), Ganglion cells (GaC), Auerbach's plexus(AP), Meissner's plexuses (MP) (H&E stain, 40x).

Figure 7. cross section in the wall of ileum shows tunica serosa (Se), and Artery (A), Vein (Ve), adipose tissue (AT), Nerve (N) (H&E 100x).

3. Discussion

The current study results showed that the tunica mucosa of the ileum in the Caucasian squirrel *Sciurus anomalus* consisted of three secondary layers: the epithelial lining, the lamina propria, and muscularis mucosa. The epithelial lining was composed of a simple columnar epithelial tissue composed of columnar cells and goblet cells. The outer surface of the columnar cells has striated border due to the presence of microvilli, which is consistent with the results of many previous studies on the histological composition of mammalian species [6,9].

The epithelial lining of the ileum part transforms into many projections known as villi, which show in different shapes. They may look leaf-shaped and shorter with pointed ends or rounds, while some appear cylindrical. A little mace shapes, and this result is consistent with the results of the study of Bat(*Eidolon helvum*) and Pangolin(*Manis tricuspis*) [10] and [11] in *Gazella subgutteroza.* , While the results of the current study differ from the results of Sheep(*Ovis aries*) and Goat(*Capra hircus*), which showed the villi in ileum shortest and tongue shape[12].

The lamina propria consists of loose connective tissue. The epithelial lining is curved at the base of the villi to form glandular units known as the intestinal glands. These glands spread in the layer of the lamina propria and appear in the form of a simple straight tubular gland with a clear lumen and extend to the muscular mucosa layer. The wall of intestinal glands is formed by many cells represented by columnar and goblet cells, and this result is consistent with many sources and research [13,14,11]. At the same time, the results of the current study differ from the results of the sheep (*Ovis aries*) study, showing the intestinal glands characterized by simple and branched tubular glands, coiled and lined with a simple cuboidal epithelium to a low columnar [15]. The functional importance of columnar cells lies in their ability to absorption of water, nutrients, and other valuable substances [13].

In comparison, goblet cells are characterized by the secretion of mucin and the preservation of the surface epithelium of the mechanical and chemical effects that the digested enzymes act within the small intestinal lumen [16]. In addition, another type of cell is represented by undifferentiated cells (stem cells) that are aggregated in the basal part of the intestinal glands and located in the adjacent or between Paneth cells and characterized by frequent division. This result is consistent with the results of the human study [17].

The intestinal glands' basis of the part of the ileum is characterized by several pyramidal cells, with a broad base and narrow towards the apical, their nucleus characterized by an oval to a spherical shape. The cytoplasm of the apical area contains rough spherical granules with acidic secretions, represented by Paneth cells. This result agrees with several studies on different types of mammalians [18,19]. The current study's results disagree with [20] study that included dogs and pigs, which showed that the small intestine does not have Paneth cells. They noted presence in abundance in the basal and cervical area of the crypt and a lack of presence in the villi.

While the muscularis mucosa is composed of a very thin, single layer of smooth muscle fibers arranged circularly, this layer is located directly under the lamina propria. It extends vertically within the villi and between the units of the intestinal glands. This result is in line with some studies on the histological composition of different mammalians [21,22]. In contrast, the current results disagree with [23] in *Isoodon macrourus* that showed the tunica muscularis mucosa consists of two layers of smooth muscle fibers, the first represented by the Longitudinal outer layer and the second is a circular inner layer of arrangement.

While the tunica submucosa is composed of dense connective tissue, bundles of collagenous fibers, and is interspersed with many blood vessels, lymphatic vessels, and neural plexuses, or Meissner's plexuses. This result is in accordance with the Iranian squirrel *Sciurus anomalu s*[24]. While this result is in disagreement with [25] in the rabbit *Oryctolagus cuniculus,* showing that this tunica is composed of fibrous connective tissue. There are some circular folds known as Kerkring valves in the mucosa and submucosa tunica of the ileum, which are specialized compositions to facilitate digestion and absorption and increase the surface area of digestion and absorption[6]. In contrast, the results of the mice study are contrary to the current results, which showed that the small intestine is free of these folds [26].

The tunica muscularis consists of two secondary layers of smooth muscle fibers, as the fiber is arranged in the inner layer circularly and thickly. It appears in irregular muscle bundles and separates a thin strip of fibrous connective tissue between the two layers containing the neural plexuses represented by Auerbach's plexuses, ganglion cells, and collagenous fibers. In contrast, the fiber showed that the outer layer was arranged longitudinally and was less thick—this resulted from the Gaddi sheep [27] and *Oryctolagus cuniculus* [25].

The tunica serosa is the last layer and is composed in turn of loose connective tissue. This tissue is covered from the outside by a row of simple squamous epithelial tissue spread by many blood vessels and clusters of adipose tissue. This result corresponds to the results [11] in ghazal *Gazella subguturosa*. In contrast, the results of the current study disagreed with [22] study in indigenous rabbits (*Oryctolagus cuniculus*), which explained that this tunica consists of regular dense connective tissue.

4. Conclusion

The current study showed that the villi of ileum show in different shapes and contain goblet cells.

References

1. Al-Sheikhly, O.F.; Haba, M.K. The field guide of land mammalian in Iraq. Publications of College of Science for woman, University of Baghdad. **2014**, 89 pp.
2. Dos Santos, M.L.; Arantes, F.P.; Pessali, T.C.; Dos Santus, J.E. Morphological, histological and histochemical analysis of the digestive tract of *Trachelyopterus stritulus* (Siluriformes: Auchenipteridae). *Zoologica*. **2015**, *32*,4, 296-305.
3. Jedeje, H.O.; Sonfada, M.L.; Salami, S.O. Anatomical studies of the gastrointestinal tract of the striped sand snake (*Psammophis sibilanus*). *Nig. Vet. J.* **2015**, *36*, *4*, 1288-1298.
4. Sauer, C.; Bertelsen, M.F.; Hammar, S.; Lund, P.; M.R.; Clauss, M. Macroscopic digestive tract anatomy of two Small antelopes, The Blackbuck (*Antilope cervicapra*) and The Arabian and Gazelle (*Gazella subgutturosa marica*). *J. Veter. Medi*. **2015**, *2*, *5*, 1-7
5. Ahmed, M.A. and Mnati,I.M. Histological and histochemical study of the duodenum in the Caucasian squirrel, *Sciurus anomalus*. *Biochem. Cell. Arch*. **2020**, *20*, *2*, 3859-3864 pp.
6. Kumar, B. Histology text and atlas.1st ed .Wolters Kuwer Health (India). *Haryana*. **2013**, 392 pp.
7. Mckinley, M.; O'loughlin, V.D. *Human anatomy*. 1sted. MC GRAW-Hill companies. United states. **2006**, 885 pp.
8. Bancroft, J.D. and Stevens, A. *Theory and practice of histological techniques*. 4th ed. Churchill Livingston. London . **2010**, 726 pp.
9. Strobel, S.; Encarnaco, J.A.; Becker, N.T.; Trenzek, T.E. Histological and histochemical analysis of the gastrointestinal tract of the common pipistrelle bat (*Pipistrellus pipistrellus*). *Europ. J. Histochem*. **2015**, *59*, 2477, 107-115.
10. Ofusori, D.A.; Caxtonomortins, E.A.; Komolafe, O.O; Oluyemi, K.A.; Adee, O.A; Ajayi, S.A; Oluwayinka, P.O; Adelakum, E.A; Keji, S.T.; Adesanya, O.A. Comparative study of ileum in rat (*Rattus novegicus*), Bat(*Eidolon helvum*) and Pangolin (*Mais tricuspis*) as Insectigated using Histological methods. *Int. J. Morphol*. **2008**, *26*, *1*, 137-141.
11. Hamza, L.O.; Al-Mansor, N.A. Histological and Histochemical observations of the Small intestine in the indigenous Gazella (*Gazella Subgutturosa*). *J. Entomol. Zool. Studies*. **2017**. *5*, 6, 948-956.
12. Maruti, G.M. Comparative gross anatomical and histological studies on small intestine in Sheep (*Ovis aries*) and Goat (*Capra hircus*). M.Sc. Thesis, Collage of veterinary Science, Maharashtra and Fishery science university. **2017**, xxxvii pp.
13. Gartner, L.P.; Haitt, J.L. *Color Textbook of Histology*. 3rd ed. Sauners, an in print of Elsevier, China. **2007**, 572pp.

14.Cui, D.; Naftel, J.P.; Lynch, C.J.; Yang, G.; Daley, W.P.; Haines, D.E.; Fratkin, J.D. *Histology with functional and clinical correlation*.1st ed. Lippincott Willians and Wilkins. London. **2011**; 456 pp.

15. Parveen, K.; Pawan, K.; Gurdial, S.; Amit, P. Histological architecture and histochemistry of duodenum of the sheep (*Ovis aries*). *Ind. J. Vet. Anat*. **2013**, *25, 1*, 30-32.

16. Kim, J.J.; Khan, W.I. Goblet cells and mucins: Role in innate defense in enteric in infections. *J. Pathog*. **2013**, *2*, 55-70.

17. Griffiths, M. *Gastrointestinal system*. 4th ed. Elsevier Ltd. China. **2012,** 203 pp.

18. Eurell, J.N.; Frappier, B.L. *Textbook of Veterinary Histology*. 6th ed. Black well publication, Asturalia. **2006**, 419 pp.

19.Mescher, A.L. *Junqueira Basic histology*.13rded. MC GRAW-Hill education. United states. **2013**, 559 pp.

20.Dellman, H.D.; Eurell, J.A. *Text book of veterinary histology*. 5thed. Lippincott Williams and Wilkins. Baltimore. **1998**, 368 pp.

21. Selim, A. and El-Nahas, E. Comparative histological studies on the intestinal wall between the prenatal ,the postnatal and the adult of the two species of Egyptian bats (*Frugivorous Rousettus aegyptiacus and intsectivorus taphozous nudiventris*). *J. Basic. App. Zool.* **2015**, 70, 25-32.

22. Alhaaik, A.G.M. Histomorphological and Immunohistological postnatal developmental changes in the small intestine and colon of the Indogenous Rabbits (*Oryctolagus cuniculus*). Ph.D. Thesis. Collage of veterinary medicine, Baghdad university. **2016**, 188 pp.

23.Hara, P.J.; Murry, P.J.; Klieve, A.V. Histology of gastrointestinal tract of the northern brown bandicoot, *Isoodon macrourus* (Marsupialia: Peramelidae). *Austeral. Mammal*. **2011**, *33*, 44-46.

24.Tootian, Z.; Sadeghinezhad, J.; Sheibani, M.T.; Fazelipour, S.; Sorde, N.D.; Chiocchetti, R. Histological and mucin histochemical study of the small intestine of the Persian squirrel (*Sciurus anomalus*). *Ant. Sci. Int.* **2013**, *88*, 38-45.

25. Ranjan, R. Anatomical exploration of gastrointestinal tract and express ion pattern of calcium channels in rabbit gut epithelium. Ph.D. Thesis. West Bengal university of animal and fisher science. **2014**, 178 pp.

26.Treuting, P.M.; Arends, M.J.; Dintzis, S.M. Upper Gastrointestinal tract .In: Treuting, P.M.; Dintzis, S.M. and Montine, K.S. (eds.). *Comparative anatomy and histology a Mouse, Rat, Human Atlas*. 2nd ed. Elsevier, London. **2018**, 211-231.

27.Rajput, R. Anatomical studies on the intestine of Gaddi Sheep. Ph.D thesis Collage of veterinary and animal science, csk himachal Pradesh krishi vishvavidyalaya. Palampur. **2006**, 160pp.