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Histological and histochemical studies on the esophagus of pigeon (*Columba livia domestica*)

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Abstract
The present research work was done on the histology and histochemistry of the oesophagus of pigeons and found that the oesophagus was composed of 3 parts: cervical part, crop (middle lobe and lateral lobe) and thoracic part. Histologically, the mucosa of cervical part, crop and thoracic part was lined by stratified squamous non-keratinized to partially keratinized epithelium. The oesophageal glands were absent in cervical part and crop whereas, in the thoracic part these glands were present and their duct open toward the lumen of the oesophagus. The mucosal folds were well developed in the cervical part, lateral lobe of the crop and thoracic part but absent in middle lobe of oesophagus. Histochemically, the epithelium of oesophagus showed PAS positive reaction that indicated presence of neutral mucopolysaccharides, whereas the stratum corneum showed Alcianophilic reaction that indicated presence of acidic mucosubstances by PAS-AB method. The oesophageal glands showed strong Alcianophilic reaction by PAS-AB method and AB method.

Keywords: Oesophagus, pigeon, histology, histochemistry

Introduction
The domestic pigeon (*Columba livia domestica*) is one of the commonest bird kept and bred by peoples as ornamental bird, messenger and for meat purpose. It is a member of the bird family Columbidae (dives and pigeons) Gibbs *et al*., (2007) [6] and Khan, (2004) [9]. The birds have coelomic cavity without diaphragm. The avian esophagus is long distensible tube that extended from the oropharynx to the proventriculus. It lies on the right side of the neck dorsal to the trachea. Immediately cranial to the thoracic entrance the esophagus of birds returns to the median line and expand ventrally to form the crop Dyce *et al*., (2010) [5] in birds, Shehan, (2012) [14] in geese and Rus *et al*., (2000) [13] in white stork whereas according Rodrigues *et al*., (2012) [12] in rhea and Bailey *et al*., (1997) [3] in captive bustards mention no form the crop. Avian esophagus consists of three parts the cervical part, crop and thoracic part (Dyce *et al*. 2010) [5]. The luminal surfaces of the oesophagus presented longitudinal folds and they are lined by stratified squamous non keratinized to partially keratinized epithelium in fowl (Banks, 1992) [4]. The wall of the oesophagus in birds consists of four tunicae i.e. mucosa, sub mucosa, muscularis and adventitia/serosa. The histological features of oesophagus varied according species to species and there are very few literature found on histology and histochemistry of oesophagus in pigeon.
So the aim to conduct the present study was to observe the histological and histochemical features of the oesophagus in pigeon. It will helpful for students, researchers, veterinarian and pigeon farmers.

Materials and Methods
Sample Collection
The present histological and histochemical study of the oesophagus was carried out on the six dead pigeons. Those were collected from two sources: the Namo Namah Parivar Bird Camp - 2020, held in Ahmedabad during the kite festival, and the Department of Veterinary Surgery and Radiology, College of Veterinary Science and Animal Husbandry, K. U., Anand. The entire research work was done in the Department of Veterinary Anatomy and Histology, College of Veterinary Science and Animal Husbandry, Anand, Gujarat.

Dissection
After collection of dead pigeons the defeathering of the entire carcass was carried out for a better accessibility of oesophagus to have a clean dissection. A sharp dissection scissor was used to cut through the abdominal wall along the posterior margin of the sternum and carefully reflected by further cutting of the thoracic region. The cervical region was incised through the mid-ventral incision on the neck and thus the cervical part, crop and thoracic part of the oesophagus were exposed then removed from the body.

Washing and Fixation
The lumen of the oesophagus was washed through 0.5 percent phosphate buffered saline solution and then fixed in 10% neutral buffered formalin solution for 48 hours.

Histological and Histochemical Study
The fixed tissues were processed out for routine paraffin blocks making and sectioning techniques. The paraffin sections were cut of 5-6 µm in thickness with the help of microtome machine and stained with following staining techniques:

1. Harris' hematoxylin and eosin stain for general architecture (Luna, 1968) [10].
2. Weigert's method for elastic fibres (Luna, 1968) [10].
3. Alcian blue method for muco-substances (pH 2.5) (Luna, 1968) [10].
4. PAS-Alcian blue method for acidic and neutral mucosubstances (pH 2.5) (Luna, 1968) [10].

The photomicrographs of histological and histochemical slides were taken by Mag Vision Soft ware.

Result and Discussion
Cervical part (pre-crop part)
The cervical part of oesophagus was comprised of four different tunics i.e. the tunica mucosa, tunica submucosa, tunica muscularis and tunica adventitia (Figure 1). The luminal surface of the oesophagus presented primary folds. The free surface of these primary folds presented some small sized secondary folds which were of varying shape and size (Figure 1). These folds were lined by stratified squamous non-keratinized to partly keratinized epithelium having varying numbers of rows in different strata (Figure 2). These observations are in agreement with the observations reported in *Elanus caeruleus* (Hamdi *et al*., 2013) [7] and in male and female homing pigeon (Kadhim and Mohamed, 2015) [8]. On the other hand, the present study findings are in disagreement with the observations reported in common wood pigeon (Rasha *et al*., 2015) [11] in which the mucosal surface was lined by stratified squamous keratinized epithelium. The size of the nuclei generally increased as moved toward the superficial layer. The density of the nuclei was more toward the deeper portion and decreased toward superficial portion. The lamina propria was having loose irregular connective tissue. It was found in the sub-epithelial portion, in the core of the tip of primary oesophageal folds and in the core of secondary oesophageal folds (Figure 2). Abdelnaeem *et al*., (2019) [1] reported the goblets cells were found in lamina propria layer in hoopoe where as in present study the goblet cells were not observed in the lamina propria layer in pigeon. The lamina muscularis was the thick layer and comprised of bundles of smooth muscle fibers. It was present deep to the lamina propria and formed a layer that extend and formed the core of primary oesophageal folds (Figure 1, 2). The tunica submucosa was very thin and composed of loose irregular connective tissue. Abdelnaeem *et al*., (2019) [1] reported the tunica submucosa was absent in Kingfisher. The tunica muscularis was very thick and comprised of circularly arranged bundles of smooth muscles fibers. In between muscle fasciculi fine connective tissue fibers were also observed. The tunica adventitia was having loose irregular connective tissue and fine blood vessels of varying size (Figure 1).

Crop (Lateral lobes and Middle lobe)
The lateral lobes were comprised of all the four tunics. The mucosa was lined by stratified squamous non-keratinized to partly keratinized epithelium. The keratinized layer was desquamating irregularly at different places. These findings are coincide with the observations reported in blue and yellow macaws (Rodrigues *et al.*, 2012) [12], geese (Shehan, 2012) [14] and barn owl (Al-Juboory *et al.*, 2015) [2]. The luminal surface was very irregular because of presence of folds of varying shape and size (Figure 3, 4). The lamina propria was thick and having loose irregular connective tissue and fine blood capillaries. It was present behind the epithelium and in the core of folds (Figure 3, 4). The mucosal glands were present in lamina propria layer as reported in common quail (Zaher *et al.*, 2012) [15], geese (Shehan, 2012) [14] and barn owl (Al-Juboory *et al.*, 2015) [2] where as in present study these glands were absent in crop of pigeon. The lamina muscularis was composed of thick layer of smooth muscle fibers which was arranged obliquely and interrupted in nature (Figure 3, 4). The tunica sub-mucosa was very thick layer and composed of loose irregular connective tissue and fine blood capillaries (Figure 3, 4). The present study findings are showing non-coincide with the observations reported in common wood pigeon (Rasha *et al*., 2015) [11] in which the adipose tissue was present in the tunica submucosa layer of crop. While in the present study the adipose tissue was not observed in tunica submucosa layer of crop in pigeon. The tunica muscularis was comprised of circularly arranged bundles of smooth muscles fibers which were interrupted in nature. In between the muscles fasciculi, fine blood capillaries and connective tissue were also observed. The outer most tunica adventitia was observed which was having loose irregular connective tissue and few small sized blood vessels (Figure 3, 4).
In middle lobe the all features are same except the layers are thin as compared lateral lobes (Figure 5).

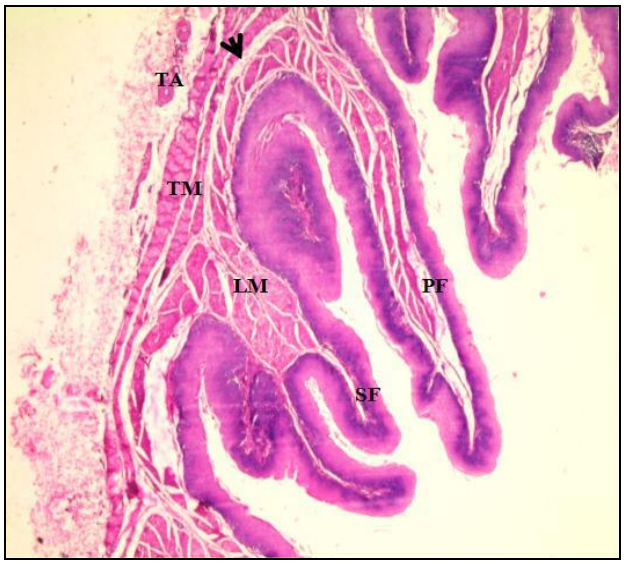

Fig 1: Photomicrograph of cranial part of oesophagus showing Primary Fold (PF), Secondary Fold (SF), Lamina Muscularis (LM), Tunica Submucosa (Black Arrow), Tunica Muscularis (TM) and Tunica Adventitia (TA). Haematoxylin & Eosin stain, 40X

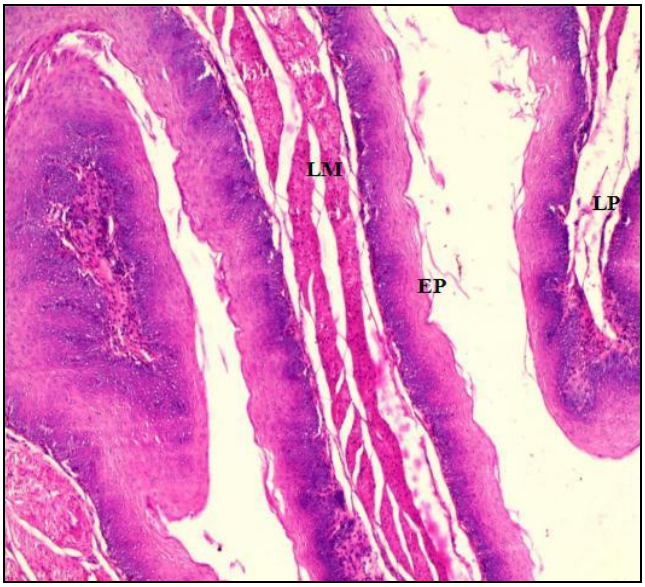

Fig 2: Photomicrograph of cranial part of oesophagus Showing Epithelium (EP), Lamina Propria (LP) and Lamina Muscularis (LM). Haematoxylin & Eosin stain, 100X

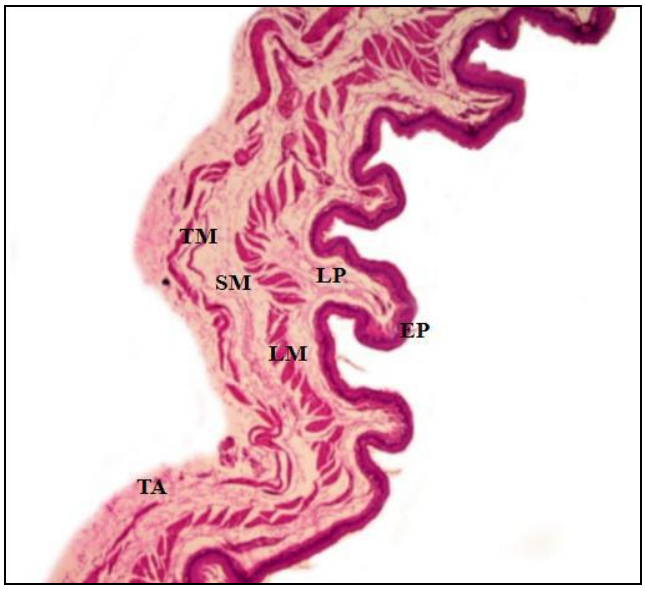

Fig 3: Photomicrograph of lateral lobe of crop Showing Epithelium (EP), Lamina Propria (LP), Lamina Muscularis (LM), Tunica Submucosa (SM), Tunica Muscularis (TM) and Tunica Adventitia (TA). Haematoxylin & Eosin stain, 40X

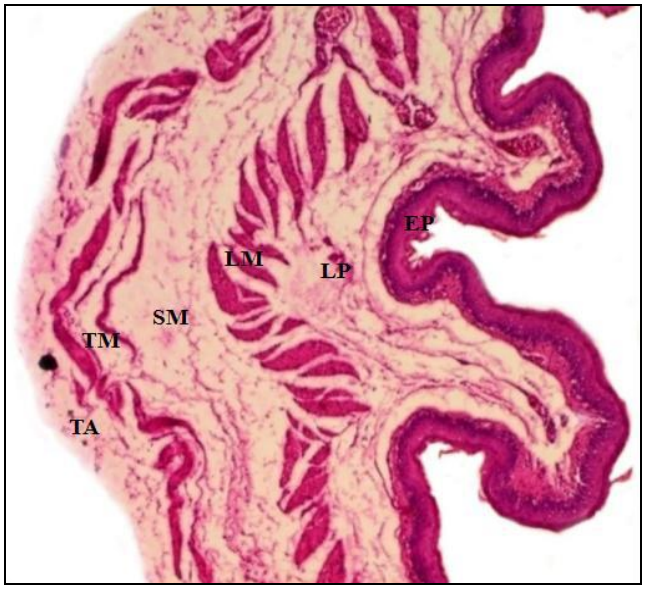

Fig 4: Photomicrograph of lateral lobe of crop showing Epithelium (EP), Lamina Propria (LP), Lamina Muscularis (LM), Tunica Submucosa (SM), Tunica Muscularis (TM) and Tunica Adventitia (TA). Haematoxylin & Eosin stain, 100X

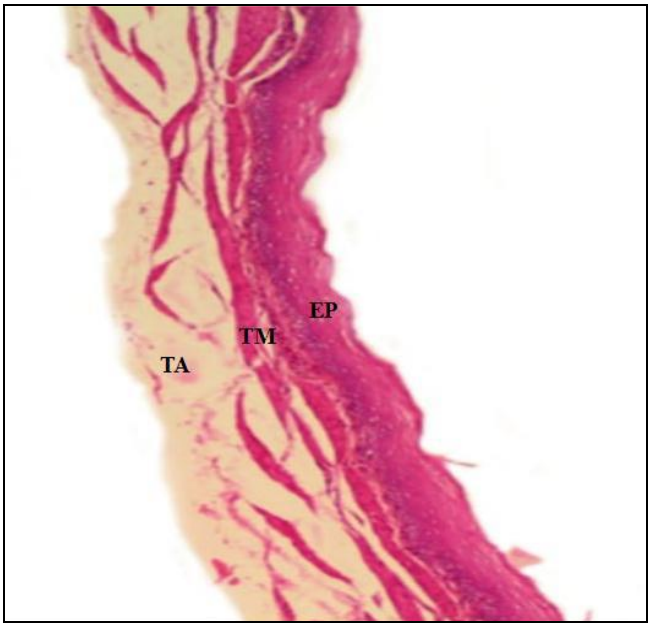

Fig 5: Photomicrograph of middle lobe of crop showing Epithelium (EP), Tunica Muscularis (TM) and Tunica Adventitia (TA). Haematoxylin & Eosin stain, 100X

Thoracic part (post-crop part)

The thoracic part of oesophagus was comparatively more developed as compared to the cervical part of oesophagus. The luminal surface presented well developed large sized oesophageal folds. The epithelium was lined by stratified squamous non-keratinized type except at few places where partly keratinization was observed (Figure 6).The lamina propria had loose irregular connective tissue and fine blood capillaries. The maximum portion of lamina propria was occupied by the mucous type of glands which were varying shapes and size and separated from those of adjacent one by the connective tissue (Figure 6, 7). The glandular ducts coursed towards the surface of the epithelium and opened on the free surface. The present findings were coinciding with the observations reported in male and female homing pigeon (Kadhim and Mohamed, 2015) [8] and in common wood pigeon and barn owl (Rasha *et al*., 2015) [11]. The lamina muscularis was well developed and interrupted in nature. It was found below the lamina propria and in the core of folds (Figure 6, 7). The tunica sub-mucosa was thin and formed by loose irregular connective tissue. The tunica muscularis was comprised of an inner circular and an outer longitudinal layer of smooth muscle fibers. The inner circular layer was as much as 5-6 times thicker than that of outer longitudinal layer. The present findings were coinciding with the observations reported in male and female homing pigeon (Kadhim and Mohamed, 2015) [8] and in common wood pigeon and barn owl (Rasha *et al*., 2015) [11]. The present findings not coincide with the observations reported in Elanus caeruleus (Hamdi *et al*., 2013) [7] in which the tunica muscularis was composed of inner longitudinal and outer circular layer of smooth muscle fibers. The outer most layer tunica serosa was formed by loose irregular connective tissue, few mesothelial cells, fine blood capillaries and blood vessels of varying shapes and size (Figure 6).

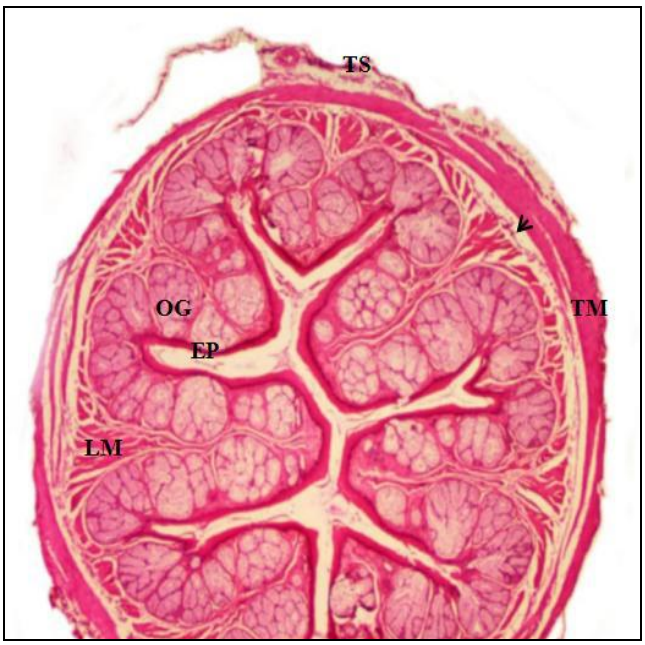

Fig 6: Photomicrograph of thoracic part of oesophagus showing Epithelium (EP), Oesophageal Gland (OG), Lamina Muscularis (LM), Tunica Submucosa (Black Arrow), Tunica Muscularis (TM), Tunica Serosa (TS). Haematoxylin & Eosin stain, 40X

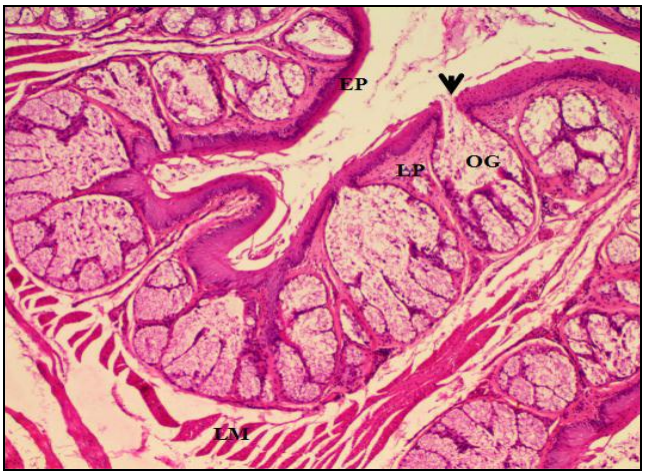

Fig 7: Photomicrograph of thoracic part of oesophagus showing Epithelium (EP), Lamina Propria (LP), Oesophageal Gland (OG), opening of oesophageal gland (Black Arrow) and Lamina Muscularis (LM). Haematoxylin & Eosin stain, 100X

Histochemistry

Cervical part (pre-crop part)

Epithelium showed moderate PAS positive reaction whereas, the lamina propria showed strong PAS positive reaction that indicated presence of neutral mucopolysaccharides. The lamina muscularis, tunica submucosa, tunica muscularis and tunica adventitia showed PAS positive reaction along with weak Alcianophilic reaction that indicated presence of neutral mucopolysaccharides and traces of acidic mucosubstances (Figure 8). These findings were coincides with the observations reported in *Coturnix coturnix* (Zaher *et al*., 2012) [15], *Elanus caeruleus* (Hamdi *et al*., 2013) [7] and kingfisher and hoopoe (Abdelnaeem *et al*., 2019) [1]. The lamina muscularis, tunica submucosa, tunica muscularis and tunica adventitia showed weak Alcianophilic reaction by AB method. (Figure 9).The blood vessels were present in propria-submucosa layer showed the presence of elastic fibers stained by Weigert's method.

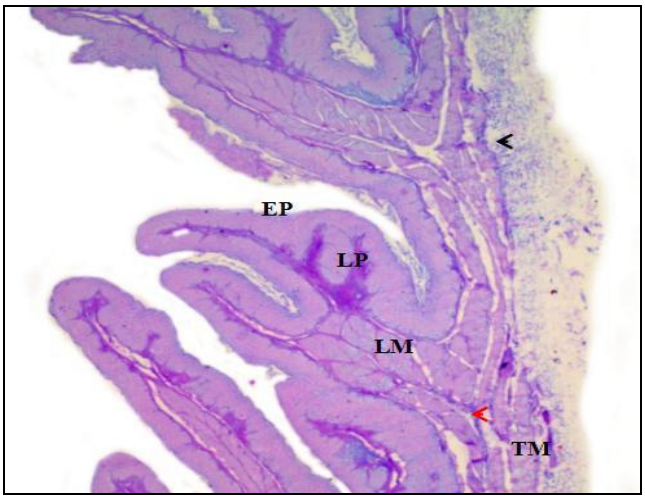

Fig 8: Photomicrograph of cervical part of oesophagus showing neutral mucopolysaccharides in epithelium (EP), lamina propria (LP), lamina muscularis (LM), tunica submucosa (Red Arrow), tunica muscularis (TM), tunica adventitia (Black Arrow) and traces of acidic mucosubstances in lamina muscularis (LM) and tunica submucosa (Red Arrow). PAS-AB stain, 40X

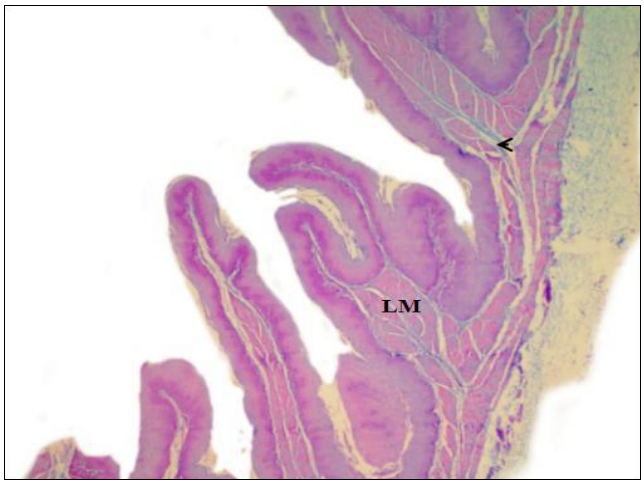

Fig 9: Photomicrograph of cervical part of oesophagus showing traces of acidic mucosubstances in lamina muscularis (LM) and tunica submucosa (Black Arrow). AB stain, 40X

Crop (Lateral lobes and Middle lobe)

The epithelium, lamina muscularis and tunica muscularis were PAS positive indicated presence of neutral mucopolysaccharides in PAS-AB method. The lamina propria, tunica submucosa and tunica adventitia showed weak Alcianophilic reaction that indicated presence of traces of acidic mucosubstances (Figure 10, 11, 13). AB method also showed presence of acidic mucosubstances in the lamina propria, tunica muscularis and tunica adventitia indicated by weak Alcianophilic reaction (Figure 11, 14). The blood vessels were present in tunica adventitia stain by Weigert's method (Figure 12).

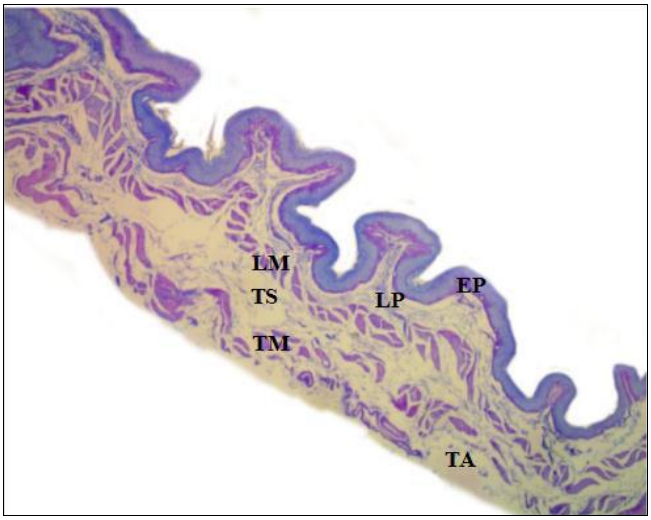

Fig 10: Photomicrograph of lateral lobe of crop showing neutral mucopolysaccharides in epithelium (EP), lamina propria (LP), lamina muscularis (LM), tunica submucosa (TS), tunica muscularis (TM), tunica adventitia (TA) and traces of acidic mucosubstances in lamina propria (LP), tunica submucosa (TS) and tunica adventitia (TA). PAS-AB stain, 40X.

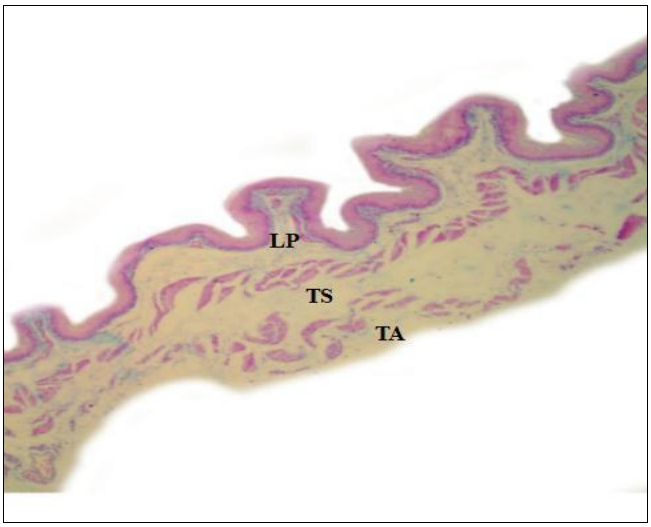

Fig 11: Photomicrograph of lateral lobe of crop showing traces of acidic mucosubstances in lamina propria (LP), tunica submucosa (TS) and tunica adventitia (TA). AB stain, 40X.

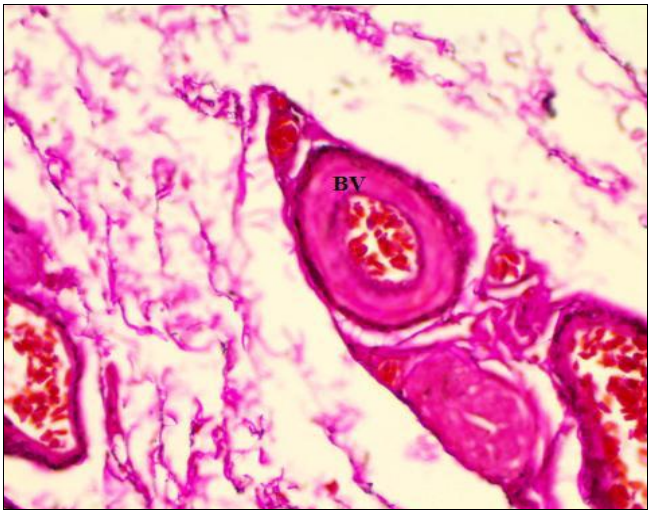

Fig 12: Photomicrograph of lateral lobe of crop showing blood vessel with elastic lamina in tunica adventitia (TA). Weigert's Stain, 100X

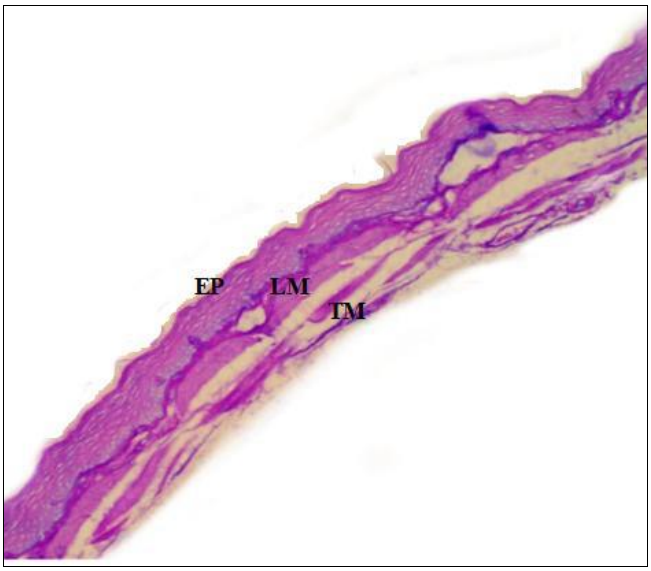

Fig 13: Photomicrograph of middle lobe of crop showing neutral mucopolysaccharides in epithelium (EP), lamina muscularis (LM) and tunica muscularis (TM). PAS-AB stain, 40X

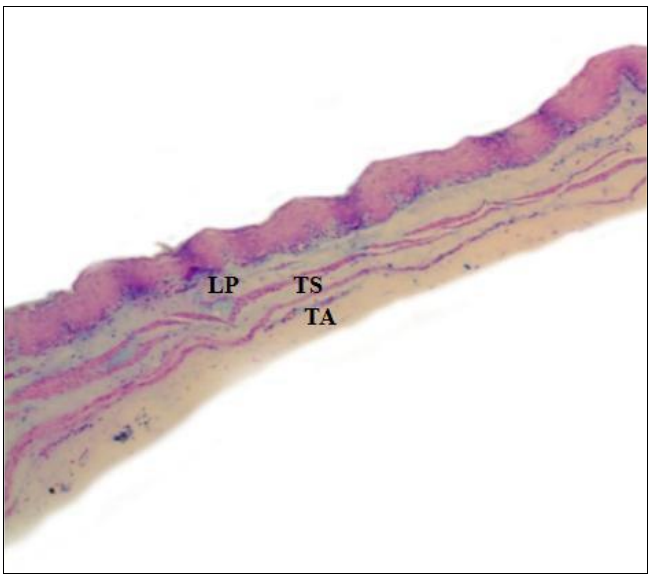

Fig 14: Photomicrograph of middle lobe of crop showing traces of acidic mucosubstances in lamina propria (LP), tunica submucosa (TS) and tunica adventitia (TA). AB stain, 40X

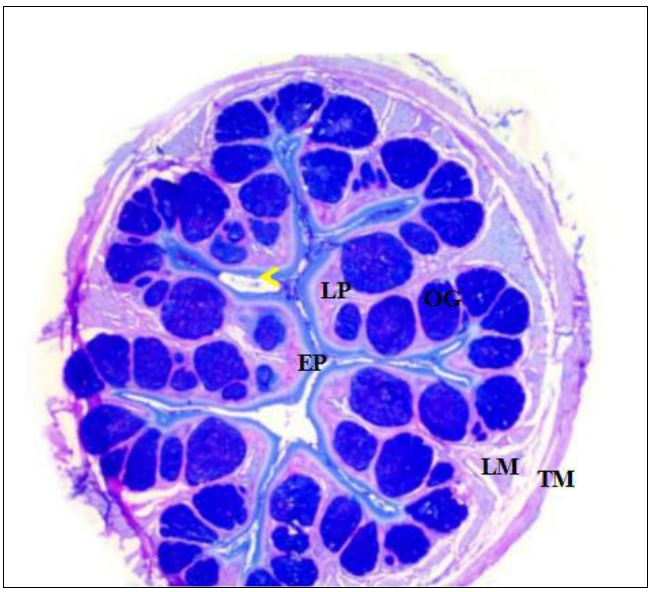

Fig 15: Photomicrograph of thoracic part of oesophagus showing neutral mucopolysaccharides in epithelium (EP), lamina propria (LP), lamina muscularis (LM), tunica muscularis (TM) and acidic mucosubstances in luminal surface of epithelium (Yllow Arrow), oesophageal glands (OG). PAS-AB stain, 40X

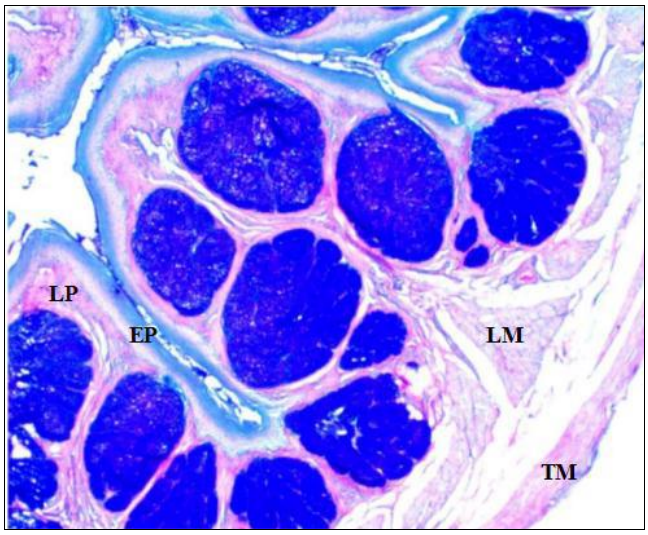

Fig 16: Photomicrograph of thoracic part of oesophagus showing neutral mucopolysaccharides in epithelium (EP), lamina propria (LP), lamina muscularis (LM), tunica muscularis (TM) and acidic mucosubstances on luminal surface of epithelium, oesophageal glands (OG), lamina muscularis (LM). PAS-AB stain, 100X

Thoracic part (post-crop part)

The epithelium showed PAS positive reaction that indicated presence of neutral mucopolysaccharides, whereas the stratum corneum showed Alcianophilic reaction that indicated presence of acidic mucosubstances by PAS-AB method (Figure 15, 16). The lamina propria showed strong PAS positive reaction that indicated presence of neutral mucopolysaccharides. The oesophageal glands showed strong Alcianophilic reaction. These findings were coincides with the observations reported in *Coturnix coturnix* (Zaher *et al*., 2012) [15], *Elanus caeruleus* (Hamdi *et al*., 2013) [7] and Kingfisher and Hoopoe (Abdelnaeem *et al*., 2019) [1]. The lamina muscularis, tunica submucosa and tunica serosa showed PAS positive reaction with weak Alcianophilic reaction indicated presence of neutral mucopolysaccharides and traces of acidic mucosubstances. Tunica muscularis showed PAS positive reaction (Figure 15, 16).

References

1. AbdElnaeem A, Elshaer F, Rady M. Histological and histochemical studies of the esophagus and stomach in two types of birds with different feeding behaviors. International Journal of Development. 2019 Dec 25;8(1):23-40.
2. Al-Juboory RW, Daoud HA, Al-Arajy AS. Comparative anatomical, histological and histochemical studies of the oesophagus in two different Iraqi birds (*Columba palumbus and Tytoalba*). International Journal of Advanced Research in Biological Sciences. 2015;2(12):188-199.
3. Bailey TA, Mensah-Brown EP, Samour JH, Naldo J, Lawrence P, Garner A. Comparative morphology of the alimentary tract and its glandular derivatives of captive bustards. The Journal of Anatomy. 1997 Oct;191(3):387-98.
4. Banks WJ. Histologia veterinária aplicada. 2nd ed. São Paulo: Manole; c1992.
5. Dyce KM, Sack WO, Wensing CJG. Textbook of Veterinary Anatomy, 4th ed. The anatomy of birds. W B. Saunders company. Philadelphia; c2010. p. 794-796.
6. Gibbs D, Eustace B, John C. Pigeon and Doves. A Guide to the Pigeons and Doves of the World. United Kingdom: Pica Press; c2007. p. 624. ISBN 1873403607.
7. Hamdi HA, Wahab E, Ghareeb MZ, Abu Amod F. Anatomical, histological and histochemical adaptations of the avian alimentary canal to their food habits: II *Elanus caeruleus*. International Journal of Scientific and Engineering Research. 2013;(4):2229-5518.
8. Kadhim, Mohamed A. Comparative anatomical and histological study of the esophagus of local adult male and female homing pigeon (*Columba livia domestica*). Al-Qadisiyah Journal of Veterinary Medicine Sciences. 2015;14(1):80-87.
9. Khan MS. Technical report on the status, trends, utilization and performance of FANGR and their wild relatives in Pakistan. Department of Animal Breeding& amp; Genetics, University of Agriculture Faisalabad, GEF-UNDP Project 2715-03-4709; c2004. p. 11-12.
10. Luna LG. Mannual of Histologic Staining Methods of Armed Forces Institute of Pathology (3rd edn.). New York: Mcgraw- Hill Book, Co., New York; c1968.
11. Rasha WAJ, Hussein AMD, Ali SAA. Comparative anatomical, histological and histochemical studies of the oesophagus in two different Iraqi birds (*Columba palumbus* and *Tyto alba*). International Journal of Advanced Research in Biological Sciences. 2015;2(12):188-199.
12. Rodrigues MN, Oliveira GB, Silva R, Tivane CT, Albuquerque JFG, Mi-glino MA, *et al*. Gross morphology and topography of the digestive apparatus in rheas (*Rhea americana americana*). Pesquisa Veterinria Brasileira. 2012;32(7):681-686.
13. Rus V, Miclauş V, Nadas GC, Cadar D. Structural particularities of the White stork (*Ciconia ciconia*) esophagus. Annals of RSCB. 2000;14(1):177-179.
14. Shehan NA. Anatomical and histological study of esophagus in geese (*Anseranser demesticus*). Bas. J Vet. Res. 2012;11(1):13-22.
15. Zaher M, El-Ghareeb AW, Hamdi H, AbuAmod F. Anatomical, histological and histochemical adaptations of the avian alimentary canal to their food habits: I-Coturnix coturnix. Life Sci J. 2012 Jan 1;9(3):253-75.