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Light and electron microscopic examination of the cervical portions of the esophagus in the Muscovy duck (Cairina moschata)

Lutfi Takcı [1] İmdat Orhan [2]

[1] *Department of Anatomy, Faculty of Veterinary Medicine, Sivas Cumhuriyet University, Sivas, Türkiye*
[2] *Department of Anatomy, Faculty of Veterinary Medicine, Erciyes University, Kayseri, Türkiye*

Correspondence: *Lutfi Takcı (ltakci@cumhuriyet.edu.tr)*

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ABSTRACT

Objectives: The esophagus in poultry serves as a primary conduit for food transport and, in many species, a specialized organ for temporary storage and conditioning. This study aimed to elucidate the anatomical, histological, and surface ultrastructural features of the esophagus and crop in the Muscovy duck, focusing on regional specializations related to its feeding ecology.

Materials and Methods: Tissue samples were collected from ten healthy adult Muscovy ducks across three distinct regions: the cervical esophagus (Region 1), the crop (Region 2), and the post-crop esophagus (Region 3). The samples were processed for light microscopy (H&E and PAS staining) and scanning electron microscopy (SEM) to evaluate the mucosal architecture and glandular distribution.

Results: Histological examination revealed that the entire esophageal tract is lined with a protective stratified squamous epithelium. A significant regional variation was observed in the density of esophageal glands, which were markedly more abundant in Region 1, suggesting an intensive lubrication requirement during the initial stages of deglutition. In the crop (Region 2), the tunica muscularis was notably thinner, facilitating the organ's expansion for food storage. A unique finding was the close anatomical association between the mucosal glands and lymphoid follicles, indicating a specialized local immune barrier. SEM analysis demonstrated intricate microridge patterns on the epithelial surface of transport zones, whereas the crop exhibited prominent glandular openings and a dense mucociliary network designed for food softening.

Conclusions: The findings indicate that the Muscovy duck esophagus is a morphologically dynamic organ with distinct regional adaptations. The high glandular density and robust lymphoid presence highlight a dual specialization for both efficient food processing and mucosal defense, reflecting the evolutionary requirements of a surface-feeding waterfowl.

Keywords: *Muscovy duck, Cairina moschata, Esophagus, Crop, SEM, Histology*

INTRODUCTION

The digestive system of birds has evolved to balance the lightness required for flight capability with the energy demand necessitated by a high metabolic rate. During this adaptation process, the upper digestive tract, particularly the esophagus and the crop (ingluvies), exhibits significant morphological variations depending on the bird's feeding ecology and dietary habits (King and McLelland, 1984; El-naseery et al., 2021). Although the esophagus in poultry is fundamentally a muscular tube that ensures the transmission of swallowed food to the stomach, in many species, it

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is specialized with the structure of the crop, where food is temporarily stored, softened, and in some species, special secretions such as "crop milk" are produced (Mahdy and Mohammed, 2024).

The shape of the crop varies significantly among poultry species depending on their feeding habits. (Kierończyk et al., 2016). It has been described as a large, glandless sac forming diverticula on either side of the esophagus in pigeons, doves, and rock pigeons (Gussekloo, 2006; Konig, 2016; Shawki et al., 2021). In chickens, the crop is described as a ventral widening of the esophagus (Denbow, 2015). It has been likened to a needle-shaped expansion in ducks and geese. (Konig, 2016). Garganeys, some water birds, grey-backed shrikes, gulls, penguins, and owls do not have a crop (Zhu, 2015; Singh, 2018; Hanafy et al., 2020; Hanafy, 2021). In waterfowl (Anseriformes), the esophagus and crop stand out due to their abundance of mucous glands and capacity for expansion, specifically aiding in the rapid swallowing of food along with water (Madkour et al., 2018; El-naseery et al., 2021).

At the histological level, all regions of the avian esophagus are generally lined with stratified squamous epithelium resistant to mechanical abrasion. However, the degree of keratinization of this epithelium and the density of the esophageal glands (glandulae esophageae) in the underlying layers differ interspecifically according to the nature of the food. Scanning Electron Microscopy (SEM) studies have determined that microridges on the epithelial surface provide mechanical protection, while mucosal folds and gland openings form a secretory network that facilitates food passage (Alsanosy et al., 2021; Ali et al., 2023). Developmental studies conducted on Muscovy ducks reported that junction points in the digestive tract (such as the esophago-proventricular junction) undergo rapid morphological change during the post-hatching period, and lymphoid infiltrations concentrate in these areas for immune response (Madkour et al., 2018; Madkour et al., 2022).

Although the Muscovy duck is an important part of aquatic ecosystems with both domestic and wild populations, and some developmental studies on its digestive system exist (Madkour et al., 2022), the ultrastructural details of the cervical region of the esophagus, the structure of the crop, and the transition between these two regions in adult individuals of this species have not yet been fully elucidated. Studies utilizing both light microscopy and high-resolution scanning electron microscopy (SEM) are particularly critical for understanding the functional morphology of these specialized regions.

The aim of this study is to examine the anatomical, histological, and surface ultrastructure (SEM) of the cervical portion of the esophagus and the crop region in the Muscovy duck (Cairina moschata), and to reveal the relationship between these structures and aquatic life and feeding biology. The data obtained from this study will contribute to the comparative anatomy of poultry, as well as establish a fundamental reference for the physiology and pathologies of the digestive system in waterfowl.

MATERIALS and METHODS

The digestive systems of 10 Muscovy ducks (5 Male, 5 Female) slaughtered at a commercial facility were removed using appropriate dissection methods and transferred to a pre-prepared 10% formaldehyde solution. After the fixation of the tissues in formaldehyde was completed, tissue samples were taken from the regions of the esophagus indicated by arrows in Figure 1 and the necessary procedure for light and scanning electron microscopy (SEM) was applied (Figure 1: 1, 2, 3).

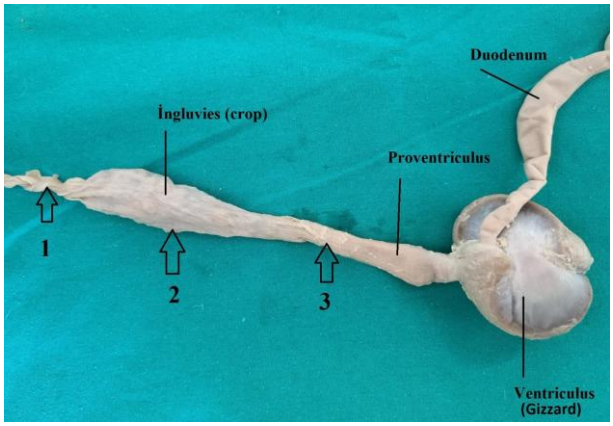


Figure 1. Regions from which tissue samples were taken from the esophagus. Arrows 1: cervical portion of the esophagus. 2. Crop. 3. Thoracal portion of the esophagus (between the crop and the proventriculus)

All tissue samples taken for light microscopy were fixed in 10% buffered formalin solution for 24 hours at room temperature. The tissue samples, processed through routine histological steps, were embedded in paraplast blocks. Serial sections 6 μm thick were obtained from the resulting paraplast blocks using a microtome (Leica RM2125RTS). The slides stained with Mallory's triple stain were examined under a research microscope (Zeiss Axio 500), and

photographs of the necessary areas were taken (Bancroft and Cook, 1984).

Samples taken for scanning electron microscopy were kept in 10% formalin solution for 24 hours. After washing twice for ten minutes in phosphate-buffered saline (0.1 M PBS), they were kept in 2.5% glutaraldehyde for 5-6 hours. Following five repeated washes with the buffer solution, they were kept in 70%, 80%, 96%, and 100% alcohol series for dehydration. The resulting samples were dried in a critical point dryer at the Erciyes University Nanotechnology Application and Research Center in Kayseri, then coated with gold and examined under a Zeiss Evo LS10 scanning electron microscope.

Ethical approval

This study was approved by the Sivas Cumhuriyet University Animal Experiments Local Ethics Committee with decision number 65202830-050.04.04-125 dated 26.12.2025.

RESULTS

In this study, the esophagus was examined in three sections for both histological and electron microscopy studies: the pre-crop cervical part, the crop, and the crop-proventriculus transition (Figure 1 1, 2, 3). Throughout its entire course, the organ is covered by cutaneous mucosa and has a four-layered wall structure. The lamina epithelialis consists of stratified squamous keratinized epithelium (Figure 2 A, B, C). The lamina propria exhibited the characteristics of dense connective tissue, composed of elastic and collagen fibers containing blood and lymph vessels and nerve fibers. It was determined that the layer was supported by a relatively small number of lymphocyte infiltrations and lymphatic cords. Within this layer, numerous mucous tubuloalveolar branched glands were observed in poultry and ducks, unlike in mammals. It was determined that these glands were mostly located in close proximity to lymph follicles or lymphoid cell infiltrations. The glands located in all three of these regions were found individually or as double gland units (Figure 2 A, B, C). While the number of glands was higher in the first region compared to the other regions, it was noted that the sections taken from the second region were very thin compared to the 1st and 3rd regions, resulting in a numerically lower count of glands. The lamina muscularis, composed of smooth muscle, was observed in places. The tunica submucosa was distinguished as having a loose connective tissue structure. In the prepared slides, the submucosa appeared wider in the proximal and distal sections, while it was very narrow in the medial section sections. The tunica muscularis layer consisted of smooth muscles in all three regions examined, but it was determined to be thicker in the 1st and 3rd region sections and thinner in the 2nd region (Figure 2: A, B, C).

In the cervical section of the esophagus (Zone 1), folds parallel to the long axis and pits between these folds were observed. The fold surface was found to be covered with micro-protrusions. In the muscular layer, an irregular undulation of muscle fibers was detected. Openings for glands to enter the lumen were found between the muscle fibers (Figure 3).

Consistent with its storage function, the surface folds in the crop region are seen to exhibit a wider and flatter structure. Crater-like circular glandular openings, where the glands producing the secretion necessary for softening the food open into the lumen, are noteworthy in this region. The epithelial surface is covered with a dense layer of mucus that envelops the stored food mass (Figure 4).

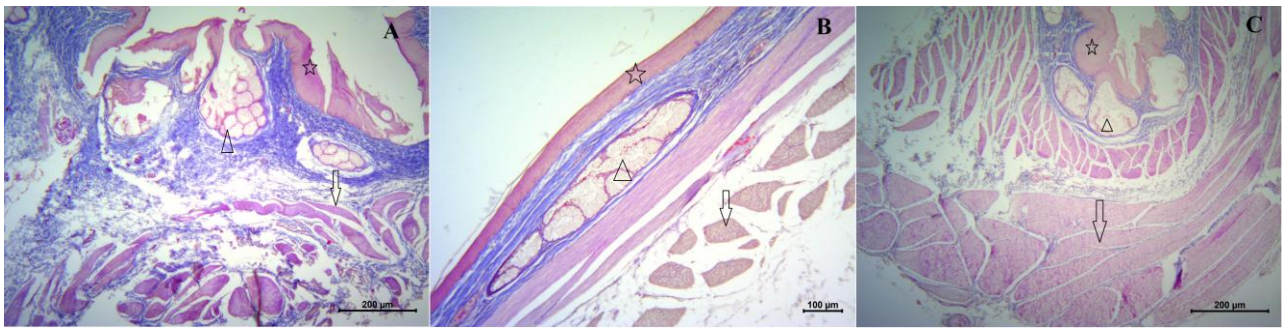


Figure 2. A: Esophagus 1st region; asterisk: lamina epithelialis consisting of stratified squamous epithelium, triangle: esophageal glands, arrow: tunica muscularis consisting of smooth muscles. A scale bar 200 µm. **B:** Esophagus 2nd region; asterisk: lamina epithelialis, triangle: esophageal glands, arrow: tunica muscularis B scale bar 100 µm. **C:** Esophagus 3rd region; asterisk: lamina epithelialis, triangle: esophageal glands, arrow: tunica muscularis. C scale bar 200 µm A-B-C Triple staining, A-C scale bar 200 µm. B scale bar 100 µm.

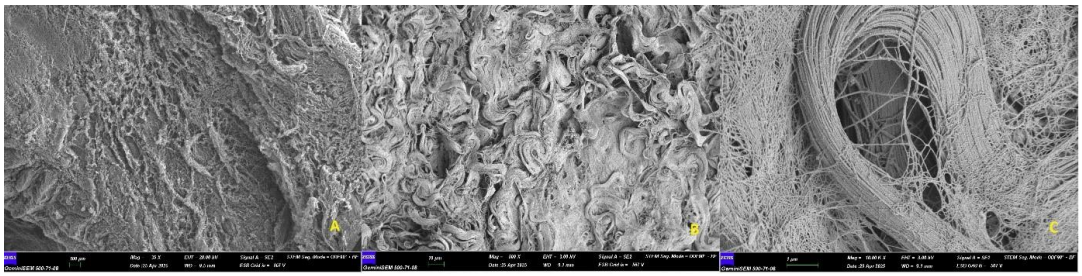


Figure 3. Scanning electron microscopic (SEM) view of the cervical esophagus (Region 1) of the Muscovy duck (Cairina moschata). **A:** Folds covered with micro-protrusions. **B:** Undulation observed in the tunica muscularis. **C:** Glandular openings between muscle fibers.

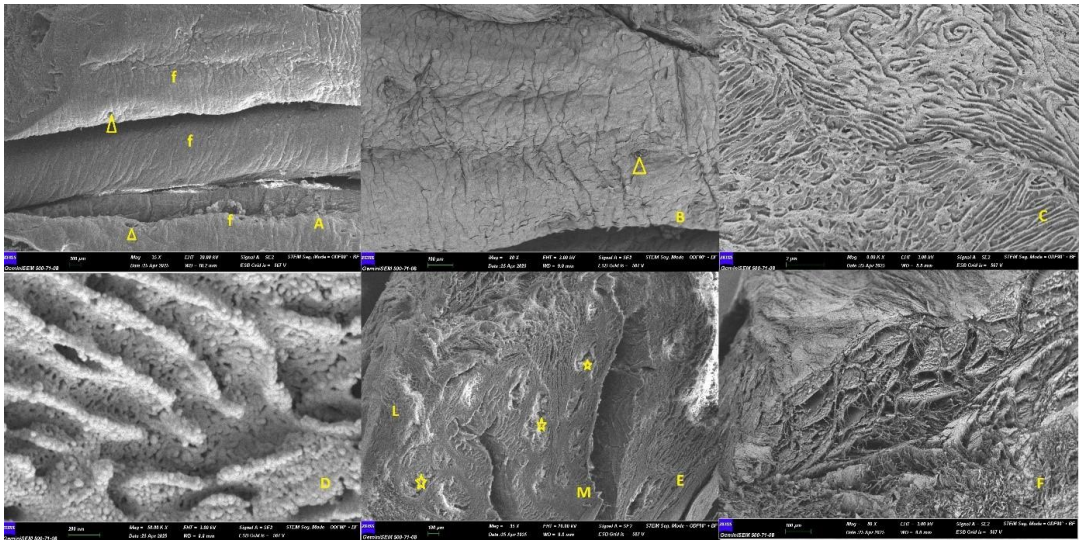


Figure 4. Scanning electron microscopic (SEM) view of the crop region (2nd Region) of the Muscovy duck. **A.** Longitudinal folds and pits in the crop. f: folds, Triangles: openings for crop glands. **B.** Magnified view of the fold, Triangle: Openings for crop glands. **C.** Microprotruding surface of the fold. **D.** Microprotruding surface of the fold (closer view of C). **E.** Cross-section of the crop wall. L: Tunica mucosae, M: Tunica muscularis, Stars: Crop glands located in the submucosa. **F.** Lower magnification view of the cross-section.

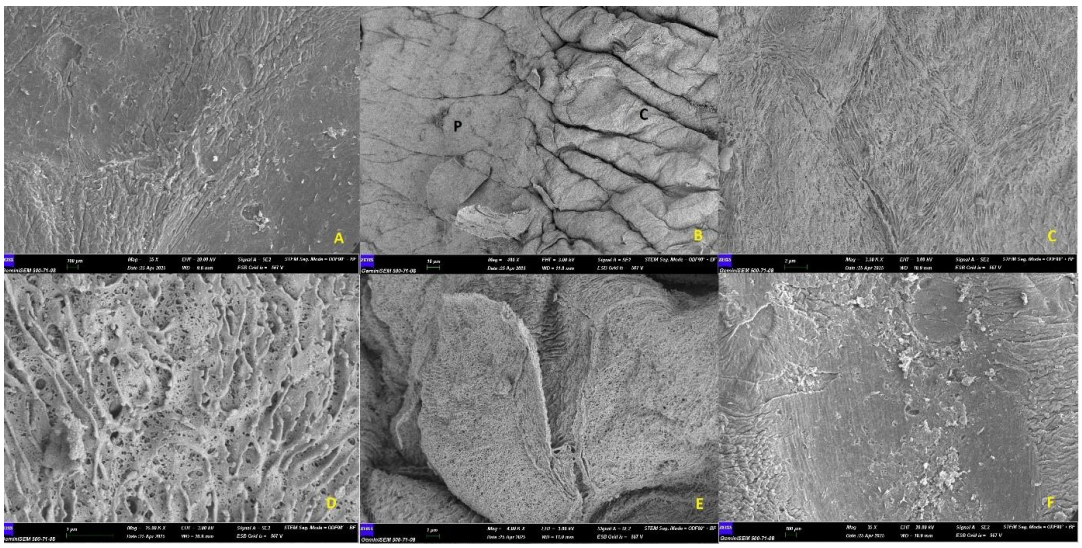


Figure 5. Scanning electron microscopic (SEM) view of the post-crop esophagus (3rd Region) of the Muscovy duck. **A.** Folded and flat areas in the thoracic section of the esophagus. **B.** Esophageal-proventriculus transition. P: proventriculus, C: esophagus. **C.** Microprotruding surface of the folds. **D.** Microprotruding surface of the folds (closer view of C). **E.** Transverse section of the post-crop esophageal wall. **F.** Folded and smooth areas in the esophageal lumen.

In this section, which facilitates the transition from the crop to the proventriculus, the luminal surface is observed to narrow again, consistent with its transport function, and the longitudinal folds become more prominent. In the surface morphology, actively secreting glandular openings and protective epithelial microstructures are observed, which facilitate the passage of food into the stomach. The thickening of the tunica muscularis in this region supports the propulsion of food towards the proventriculus via peristaltic movements (Figure 5).

DISCUSSION

In this study, the cervical part of the esophagus, the crop, and the transitional regions in the Muscovy Duck were morphologically examined; the findings obtained were correlated with the feeding ecology and food transport strategies of this waterfowl. The structure of the stratified squamous epithelium observed in all parts of the esophagus in our study is consistent with the general histological characteristics of poultry (King and McLelland, 1984). However, the thickness of the epithelium and the level of keratinization showed significant differences between regions.

One of the most striking findings of our study is the higher number of glands in Region 1 (cervical esophagus) compared to other regions. This indicates that the need for lubrication of the material ingested with water during swallowing is highest in this area. El-naseery et al. (2021), in their comparative study of ducks and pigeons, stated that esophageal glands in ducks are more developed than in other species, correlating this with their aquatic lifestyle (El-naseery et al., 2021). Our findings contrast with the simpler glandular structure reported by Alsanosy et al. (2021) for the common kestrel *(Falco tinnunculus)*, supporting the high secretory requirement of waterfowl (Alsanosy et al., 2021).

The thinner wall structure and tunica muscularis in the second region (crop) compared to other regions can be explained by the primary function of this area being an expansible storage space rather than transport. Mahdy and Mohammed (2024) reported that the crop in pigeons specializes into two lateral diverticula and has a relatively thin wall structure (Mahdy and Mohammed, 2024). Although a more uniform expansion was observed in the American Muscovy Duck in our study instead of a distinct bilobular structure like in pigeons, the histological thinning of the wall was evaluated as a common adaptation for the storage function (Konig, 2016).

The close proximity of the esophageal glands to lymph follicles and lymphoid cell infiltrations is among the unique findings of our study. Madkour et al. (2018) observed dense lymphoid infiltrations at the esophago-proventricular junction in post-hatching Muscovy Ducks and described this as part of the regional immune defense (Madkour et al., 2018). Our findings also suggest that these entry regions of the digestive tract are under a continuous defense mechanism against antigenic stimuli and that the gland-lymphoid relationship plays a critical role in mucosal immunity (GALT).

The microridges and glandular openings observed on the surface in our Scanning Electron Microscopy (SEM) examinations parallel other findings in the literature. Shawki et al. (2021) stated that these structures in rock pigeons increase mucus retention, thereby protecting the epithelial surface (Shawki et al., 2021). The dense secretory network observed in the American Muscovy Duck in our study was interpreted as a vital adaptation for maintaining mucosal integrity, especially during the passage of hard food items or aquatic microorganisms.

The tunica muscularis being thicker in Regions 1 and 3, and thinner in Region 2 (crop), is directly related to the organ's need for peristaltic activity. While the cervical region requires a strong muscle mass to propel food, and the post-crop region needs it to transfer food controllably to the proventriculus, the crop region focuses on passive expansion. This finding functionally contrasts with the trend of muscle thickening reported by Madkour et al. (2022) in ventriculus development, but it proves that each segment is differentiated appropriately for its specific mechanical task (Madkour et al., 2022).

CONCLUSION

In conclusion, the esophageal transit lines in the American Muscovy Duck exhibit a dynamic structure in terms of epithelial thickness, gland density, and lymphoid organization. This morphological diversity optimizes the species' feeding success in the aquatic environment and the defense strategies in the upper parts of the digestive tract.

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