

Macro and Microanatomy of the Visceral Organs of a White-Breasted Waterhen (Dahuk) bird (*Amaurornis phoenicurus*)

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With 7 figures.

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Abstract

The white-breasted waterhen is a widely distributed wetland species that inhabits waterlogged environments consistently throughout the year. The small water birds are good hunters of worms, insects, and small fish. This bird uttered loud calls during the periods of its courtship display, incubation, and caring for the young. Although several studies have been done on the white-breasted Waterhen bird, little attention has been paid by ornithologists to its visceral organs. Therefore, this study

aimed to describe the macro- and microanatomical features of a white-breasted waterhen bird's visceral

organs and compare them with domestic birds. This study showed that the bird had a black coat color on the dorsal part of the body, but the white color extended from the base of the beak to the vent as the ventral region of the body through the breast. The beak and legs display a subtle pale-yellow hue extending along their length. Legs and fingers are long and bony. At the bifurcation of the trachea, there was a laterally swollen structure called the syrinx, more well-developed than domestic chickens and capable of producing the loud call. The esophagus was a long, thin-walled, dilatable tube without any crop. Gizzard was comparatively softer in consistency.

The length of the duodenum and jejunum is relatively smaller than that of other domestic avian species. There was a poorly developed caecum, and the colo-rectum was shorter in length. Interestingly, the spleen was elongated, different from that of other avian species. The right hepatic lobe was observed to be larger than the left as usual. All these anatomical modifications might be due to their environmental adaptation. Thus, this research will help our clinicians and conservationists take further steps toward disease diagnosis and monitor the conservation of this species.

Keywords: Waterhen, syrinx, crop, intestine, and spleen.

Introduction

In Bangladesh, there are 50 species of wetland birds, 9 of which are threatened in different categories (IUCN, 2000). Wetland birds are the most appealing birds and live in freshwater marshes, rivers, lakes, and coastal areas. Common aquatic birds observed in Bangladeshi freshwater habitats comprise herons (*Ardeola grayii*, *Butorides striatus*) and egrets (*Egretta garzetta*, *Bubulcus ibis*, *Casmerodius albus*), cormorants (*Phalacrocorax spp.*), bronze winged jacanas (*Metopidius indicus*), white-breasted waterhens (*Amaurornis phoenicurus*), whistling ducks (*Demigryna spp.*), and so on (Akhtar et al., 2013). The waterhen (*Amaurornis phoenicurus*) belongs to the Rallidae family, which

includes rail and crane birds. The population of waterhens has expanded its range across regions in South and Southeast Asia. (Sadam et al., 2022). 'White-breasted Waterhen', or simply 'Dahuk' in Bengali, is one of these birds. It is available in Bangladesh, Sri Lanka, the Maldives, India, Pakistan, and Myanmar. Although birds are essential for controlling pests, scavenging, pollinating, and providing food for humans, their habitat is highly susceptible to changes, and their diversity is rapidly declining due to people's lack of awareness of the need to protect and conserve them (Sarker et al., 2001). The degradation of habitat and its subsequent impact on food and shelter are the leading causes of Bangladesh's alarming decline in bird populations (Hussain, 2008). Plenty of information is available from past research on white-breasted waterhens, such as their habitats, dietary preferences, and activity patterns. However, some research has provided information on some of its anatomical features. This thorough analysis considers macro and micro-anatomical characteristics, but there is still a lack of knowledge regarding the anatomy of their internal organs. This article aims to fill this gap by thoroughly examining the White-Breasted Waterhen's structural traits at both the organ and cellular levels. The main objective of this research was to find out about the macro and microanatomy of the white-breasted waterhen's visceral organs. This may help better understand the

physiological and pathological conditions of this species.

Materials and Methods:

Bird

A lifeless white-breasted waterhen was discovered resting near a waterlogged area within the forested hills of Chattogram district, Bangladesh. The dead bird was wrapped in a polythene bag and brought to the Anatomy lab in the Department of Anatomy and Histology, Chattogram Veterinary and Animal Sciences University (CVASU), in a plastic container with ice within 2 hours. This dead bird was evaluated by two avian specialists who also conducted the postmortem assessment following the standard procedure or protocol described by the Atlas of Avian Necropsy (Majó & Dolz, 2011). The bird was thoroughly cleaned and then dissected for detailed anatomical study. They examined every bodily system chronologically. It was 60 cm in length, and this bird was identified as a white-breasted waterhen by the characteristic features of the bird, such as long legs, the black color of the whole body, and a wider white mark in the breast area. It was identified as a female bird by finding the ova during dissection. The cause of death was unknown. No distinctive pathognomonic lesions were detected during the postmortem examination.

Measurement and Sample Collection

Samples from internal organs were taken for gross anatomical and histological analysis. The length and width of several visceral organs were measured using a measuring scale. Tissue samples were preserved in 10% neutral buffered formalin for histological examination, and paraffin blocks were prepared following the protocol. The paraffin blocks underwent sectioning at a thickness of 5 μ m, and they were stained with Hematoxylin and Eosin stain according to standard protocol (Cardiff et al., 2014).

Histomorphometry

To obtain histomorphometries, a microscopic study of the different parts of the uterus was done, and photomicrographs were taken using a photomicroscope (AmScope Trinocular Compound Microscope with 1.3 MP Camera, Model T490 B-MT) and AmScope image measuring software (x86, 3.7.3036 version).

Result:

External morphology

The bird had a black coat color on the whole body, but the white color extended from the base of the beak to the vent as the ventral region of the body through the breast (Fig 1). The beak and legs were faded yellow in color. The beak was 3 cm long. From the point of the beak to the tip of the tail,

the length of the bird was 32 cm. From the point of the beak to the base of the tail, the length was 24 cm. The leg length was 31 cm for both the right and left legs. The pes length was 12 cm.

Macro and microanatomy of the visceral organs

Respiratory system

Larynx and Trachea

The larynx was 1 cm long and 0.51 cm wide. The trachea was 9.9 cm and 0.49 cm wide. The well-developed syrinx was located between the junction of the trachea and the primary bronchus or the bifurcation of the trachea (Fig 2). The complete tracheal cartilage encircled the airway. The length of the primary bronchus was 0.85 cm. Histologically, the tracheal wall examined had four distinct tunicae: mucosa, submucosa, fibrocartilage, and adventitia according to H&E-stained cross-sections of the trachea. The tunica mucosa was lined by a pseudo-stratified columnar epithelium consisting of ciliated, goblet, and basal cells. The submucosa was closed by the hyaline cartilage. The trachea-lateralis muscles were firmly connected to the outer adventitia. (Fig 5).

Lungs

The length of the lungs was 3.20 cm, and the width was 2.18 cm at the mid position. The rib impressions were found on the dorsal surface of each lung (Fig 2). Histological examination revealed that the lungs comprised

para-bronchi, which were situated centrally within the pulmonary lobules and originated from the secondary bronchi (Fig 5).

Digestive system

Esophagus

The length of the esophagus was 9.7 cm, and the diameter was 0.35 cm. The esophagus extended from the oropharynx all the way to the proventriculus (Figure 3). It was a long, elastic, thin-walled tube. Cranially, it was located in the midline dorsal to the larynx and trachea but caudal to the 5th cervical vertebra, and it was on the right side of the neck to the trachea. The crop was absent. Histologically, the esophagus consisted of four layers: tunica mucosa, submucosa, tunica muscularis, and tunica adventitia/serosa. The tunica mucosa contained several longitudinal folds lined by non-keratinized squamous epithelium. The esophageal glands were present in the proprio-submucosa. The tunica muscularis consisted of a thick inner circular layer and a thin longitudinal layer of smooth muscle fibers (Fig 6).

Proventriculus

The proventriculus measured 1.2 cm in length, with a width of 0.56 cm at its midpoint. It was an elongated organ directed cranio-caudally (Fig 3). Under the microscope, the wall of the proventriculus was composed of four distinct layers: the mucous membrane (tunica mucosa), the submucosa (tunica submucosa), a muscular layer (tunica

muscularis), and an outer serous layer (tunica serosa). The numerous tubular glands were in the submucosa. The tunica muscularis was composed of two layers of smooth muscle fibers: an inner layer arranged longitudinally, and an outer layer arranged circularly (Fig 6).

Gizzard

The gizzard was 2.3 cm in length and 2.0 cm in width (Fig 3). Microscopically, the branched tubular mucosal glands were in the proprio-submucosa, and thick smooth muscle layers were in the tunica muscularis layers (Figure 6).

Small intestine

The duodenum measured 7.4 cm in length and had a width of 0.15 cm. The duodenum had an ascending and descending parts surrounding the pancreas. The Jejunum and ileum were 18.2 cm & 4.7 cm in length and 0.13 cm and 0.16 cm in width, respectively (Fig 3). Histologically, the tunica mucosa layer was damaged, and this cannot be explained. The small intestine contains intestinal glands in the submucosa layer. The tunica muscularis was made up of an inner layer of smooth muscle fibers arranged in a circular pattern and an outer layer of smooth muscle fibers arranged longitudinally (Fig 6).

Large intestine

The large intestine of this bird consists of the paired cecae and small straight colorectum to the continuation of the ileum. The caecum was 2.1 cm in length and 0.14 cm in width. The colorectum was 4.2 cm in length and 0.24 cm in width (Figure 3). Histologically, the tunica mucosa layer was damaged, and this cannot be explained. The small intestine contains intestinal glands in the submucosa layer. The tunica muscularis consisted of two smooth muscle layers: an inner layer with fibers arranged circularly and an outer layer with fibers oriented longitudinally (Fig 6).

Liver

The liver has right and left lobes that join cranially with each other. The right lobe was generally larger than the left lobe. The liver was 3.6 cm in length and 2.3 cm in width (Fig 4).

Heart

The heart was 2.0 cm in length and 1.1 cm in width (Fig 4). The heart apex is narrower than the base, as the greater part of the heart lies on the left side of the median plane.

Kidney

Both kidneys consist of a cranial, middle, and caudal lobes and are flattened and embedded in the ventral surface of the synsacrum bone. The kidney was 3.2 cm in length, and the cranial, middle, and caudal widths were 0.87, 0.5, and 0.53, respectively. In histological observation, the kidneys of all the birds examined were composed of two

main zones: the cortex and the medulla. The cortex makes up the vast area of the kidney, with only a small portion of it being the medulla. The renal corpuscle was made up of an outer Bowman's capsule, which was separated from the centrally positioned glomerulus by Bowman's space. The glomeruli consisted of the tightly packed central core of mesangial cells, surrounded by capillary loops (Fig 7).

Spleen

The spleen was elongated in shape. The spleen was 1.6 cm in length and 0.33 cm in width (Figure 4). The spleen was histologically divided into two main parts: the white pulp and the red pulp. No identifiable marginal zone was observed in the spleen (Fig 7).

Discussion

The structure of the different organs of the waterhen bird indicates some similarities and dissimilarities with other avian species. The male (28 cm) birds were comparatively larger than the female (27 cm). The adult bird was black or dark grey, and the face and breast were milky white in color. The eyes were black, and the beak was faded yellow (Sarkar, 2024). Similar to the syrinx of other singing birds, the waterhen also possesses a well-developed syrinx, which controls labial tension and airflow (Riede & Goller, 2010). The sound was unpleasant and harsh, with the call lasting typically for about 15 to 20 minutes at a stretch. (Sarkar, 2024). The dorsal surface of the lungs

was convex, and the impressions of ribs were similar to other avian species (Doğan & Takıcı, 2018).

The esophagus of the waterhen was long, elastic, and located dorsally on the right side of the neck to the trachea, similar to the esophagus of other avian species (Mobini, 2014). The length of the esophagus was 9.7 cm, whereas the lengths of a kingfisher and broiler chicken were 7.3 ± 0.07 cm and 18.8 ± 4 cm, respectively (Elshaer, 2018; Koko-szyński et al., 2017). The average length of the esophagus was 14.6 cm in ducks (Działo-Szczepańczyk & Wesolo-wska, 2008). In avian species, the most important function of the crop is the storage of ingested feed (Bolton, 1965) along with absorptive and digestive capability, but the crop in the waterhen was absent. Avian crops play an important role in the temporal storage of feed, moistening, and a functional barrier for pathogens through decreasing pH value by microbial fermentation (Kierończyk et al., 2016). Analysis of the crop contents in fowls has demonstrated that starch undergoes extensive breakdown into sugars, with a significant portion of the sugar produced being lost from the crop. This loss can occur through absorption by the crop lining, microbial activity, or leaching out of the crop (Bolton, 1965).

The waterhen mostly relies on aquatic species for feeding. They feed on worms, insects, different types of seeds, small fish, etc. (Sarkar, 2024).

It may have little importance for fiber digestion in crops. Although food may be stored throughout the length of the esophagus of avian species that have no crop (Ziswiler & Farner, 1972), no food remains were observed in the esophageal lumen of the waterhen examined. The gizzard of a chicken is keratinous and horny (Nasrin et al., 2012), but the gizzard of a waterhen appeared to be soft in consistency. In the case of the small intestine of the waterhen, the length of the duodenum and jejunum were 7.4 cm and 18.2 cm, respectively. But in the case of chicken, the average length of the duodenum and jejunum was 34.13 cm and 123.50 cm, respectively (Nasrin et al., 2012). Diet played a key role in explaining the differences observed in the length and nominal surface area of the small intestine among birds (Duque-Correa et al., 2022; Lavin et al., 2008). The average length of the caecum was 18.30 ± 1.20 cm in broiler chickens (Rani et al., 2020), and in the waterhen, the length of the caecum was 2.1 cm. A significant relationship between avian cecal lengths and food habits, and the longest ceca occurred in herbivorous species (Naik & Dominic, 1962; Pinchon, 1942). Certain bird orders, such as Anseriformes, Galliformes, Gruiformes, Cuculiformes, Strigiformes, Caprimulgiformes, and Trogoniformes, typically have well-developed ceca. However, the development of the ceca is less predictable in other bird groups and appears to be influenced mainly by

diet. Herbivorous birds consuming large amounts of cellulose tend to have well-developed ceca, while species with diets rich in soluble sugars and proteins usually have small or absent ceca (DeGouler et al., 1999). The avian caecum plays an important function in the prevention of colonization of pathogens, eliminating toxic compounds, digesting nutrients, and absorbing ingested nutrients (Tan et al., 2019), but the caecum was poorly developed in the waterhen because of their protein-trend diet; it may have little importance for fiber digestion.

The right lobe of the liver was usually larger than the left lobe, and there were no further lobular subdivisions similar to the liver of other avian species (Bailey et al., 1997).

The spleen of birds has differences in size (Khan et al., 2014; Millán et al., 2001). In the anatomic analysis of the present study, the spleen was elongated in shape and lay close to the right side of the junction between the proventriculus and gizzard. This finding is dissimilar to the observation of (Paul & Sarder, n.d.) in broiler chickens. Further reported that the spleen was a rounded, reddish-brown organ. The spleen of the duckling had a triangular shape, featuring a dorsal surface and a curved ventral surface (Sultana et al., 2012). Kara et al., (2021) reported the spleen of Red-legged partridges as dark red in color and elliptical in shape. It was located close to the proventriculus, ventriculus, and liver

on the right side of the gastric isthmus. (Song et al., 2012) reported that the spleen in ostriches is dark red, elliptical in shape, and positioned wedged against the right kidney, the posterior of the caudal vein, and the proventriculus.

Conclusion

This research detailed the anatomical characteristics of the internal organs of a white-breasted waterhen (Dahuk) bird. A postmortem examination is the main technique used to diagnose avian diseases. This study could be used as a basis for research on the pathophysiology of this species. The liver, lungs, spleen, cecum, duodenum, jejunum, and colorectum are the key organs examined during post-mortem to assist in disease diagnosis. Many infectious and noninfectious diseases bring pathognomonic lesions to these visceral organs of birds, concerning changes in color, size, shape, and consistency. To identify these changes in those visceral organs, we must know their normal color, size, shape, and consistency at different stages. Moreover, this data may contribute to a better understanding of the postmortem examination and disease diagnosis and thus improve the strategies for the conservation of this species.

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Author contribution: MRP and SKS conceived and designed the study with comments from all authors. MRP wrote the original manuscript, which SKS, ALA, and MLR edited. MRP, MSHT, and FA performed the experiments. SKY transported the dead bird to the Department of Anatomy and Histology, CVASU.

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Conflict of Interest

The authors declare that there is no conflict of interest.

Ethics approval statement: It was a dead waterhen bird. Nevertheless, the study was conducted according to the guidelines provided by the ethical committee of the Chattogram Veterinary and Animal Sciences University, Bangladesh.

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Fig (1): External morphology of a white-breasted waterhen bird. A) Dorsal view, B) Ventral view;

a) light yellow color beak, b) black coat color dorsal part of the body, c) white coat colored ventral part of the body, d) light yellow colored leg, e) light yellow colored fingers.



Fig (2): Organs of the respiratory system of a white-breasted waterhen bird.

a) larynx, b) Trachea, c) Syrinx, d) Primary bronchus, e) lung, f) Rib impression on the lung.

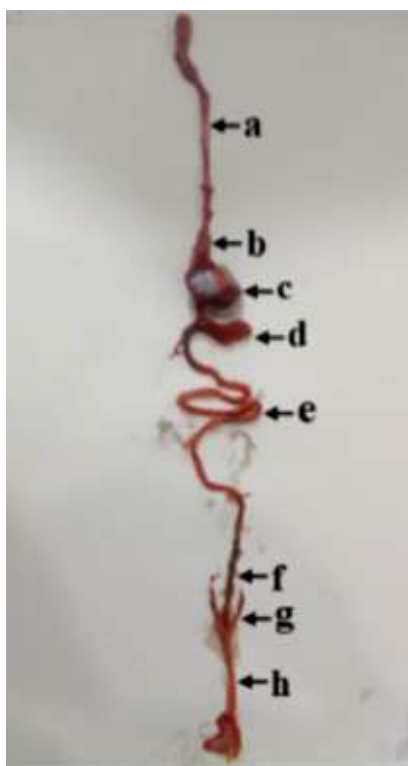


Fig (3): Organs of the digestive system of a white-breasted waterhen bird.

a) Esophagus, b) Proventriculus, c) Gizzard, d) Duodenum, e) Jejunum, f) Ileum, g) Caecum, and h) Colo-rectum.



Fig (4): Organs of a white-breasted waterhen bird.

A) Liver, a) Right lobe, b) Left lobe; B) Heart; C) Spleen.

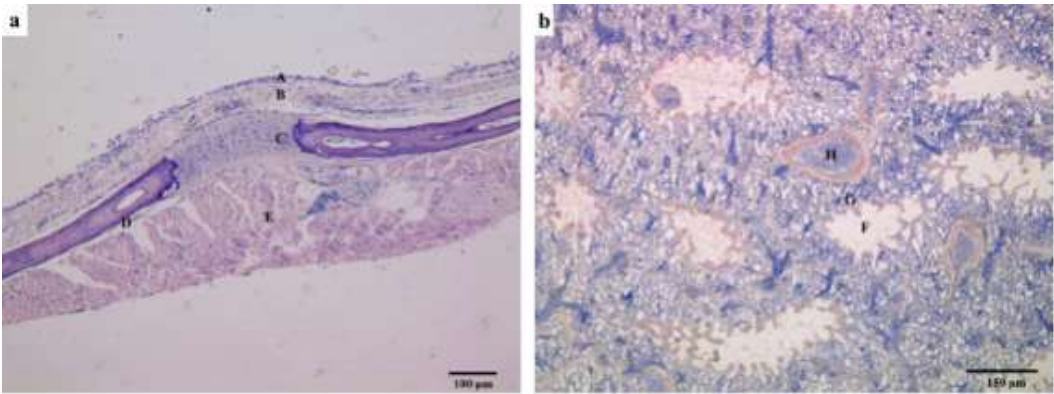


Fig (5): Hematoxylin and eosin staining of the trachea (a) and lung (b) from the respiratory system of a white-breasted waterhen bird.

A, Lining epithelium; B, Propio-submucosa; C, Hyaline cartilage; D, Tunica adventitia; E, Skeletal muscles; F, Parabronchial lumen; G, Parabronchial wall; H, Blood vessel. Scale for low magnification: 150 µm, for high magnification: 100 µm.

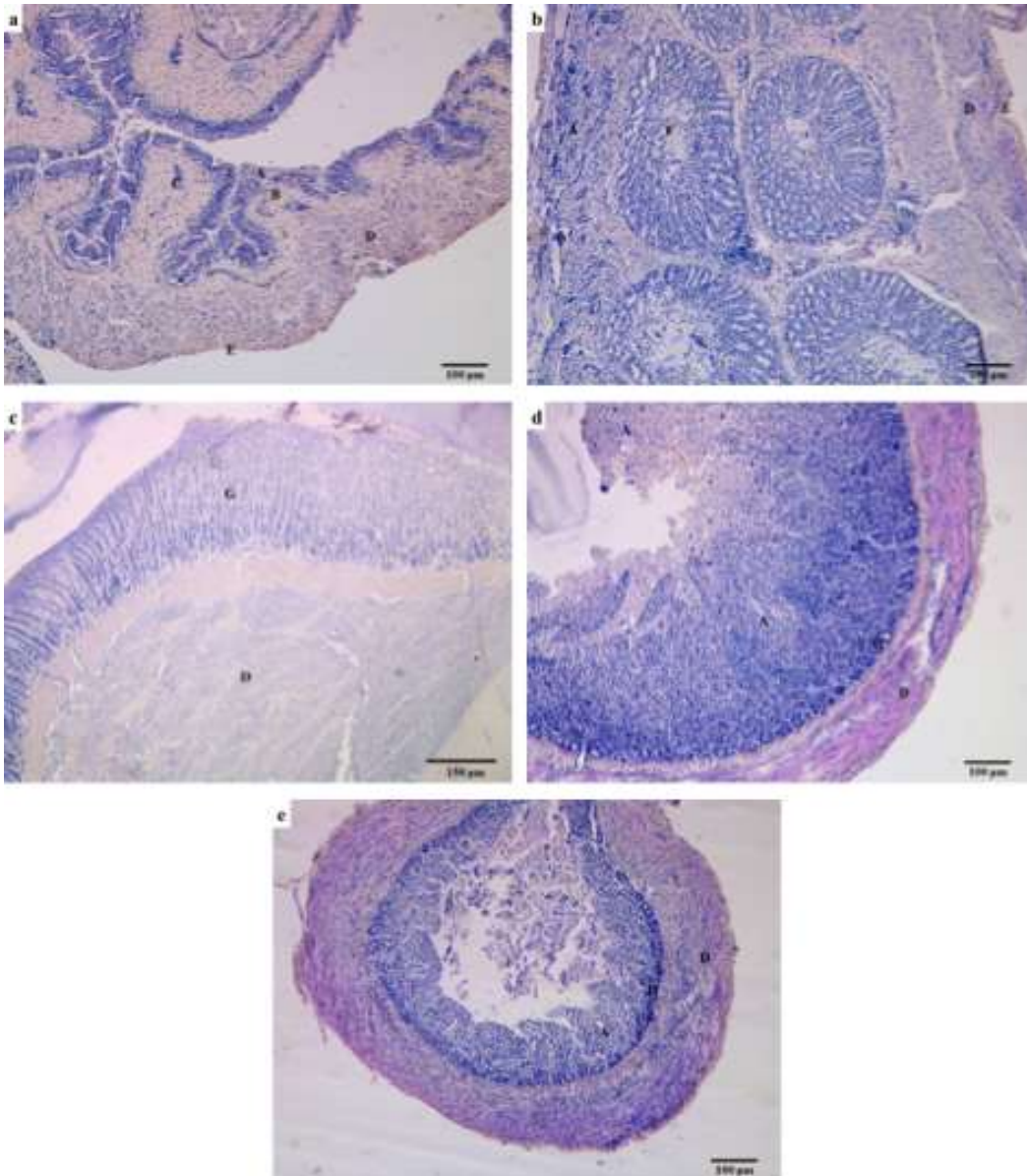


Fig (6): Hematoxylin and eosin staining of the cervical part of the esophagus (a), pro-ventriculus (b), gizzard (c), small intestine (d), and large intestine (e) from the digestive system of a white-breasted waterhen bird.

A, Mucous membrane; B, Propio-submucosa; C, Esophageal gland; D, Tunica muscularis; E, Tunica adventitia; F, Proventricular gland; G, Simple tubular gland; H, Intestinal gland.

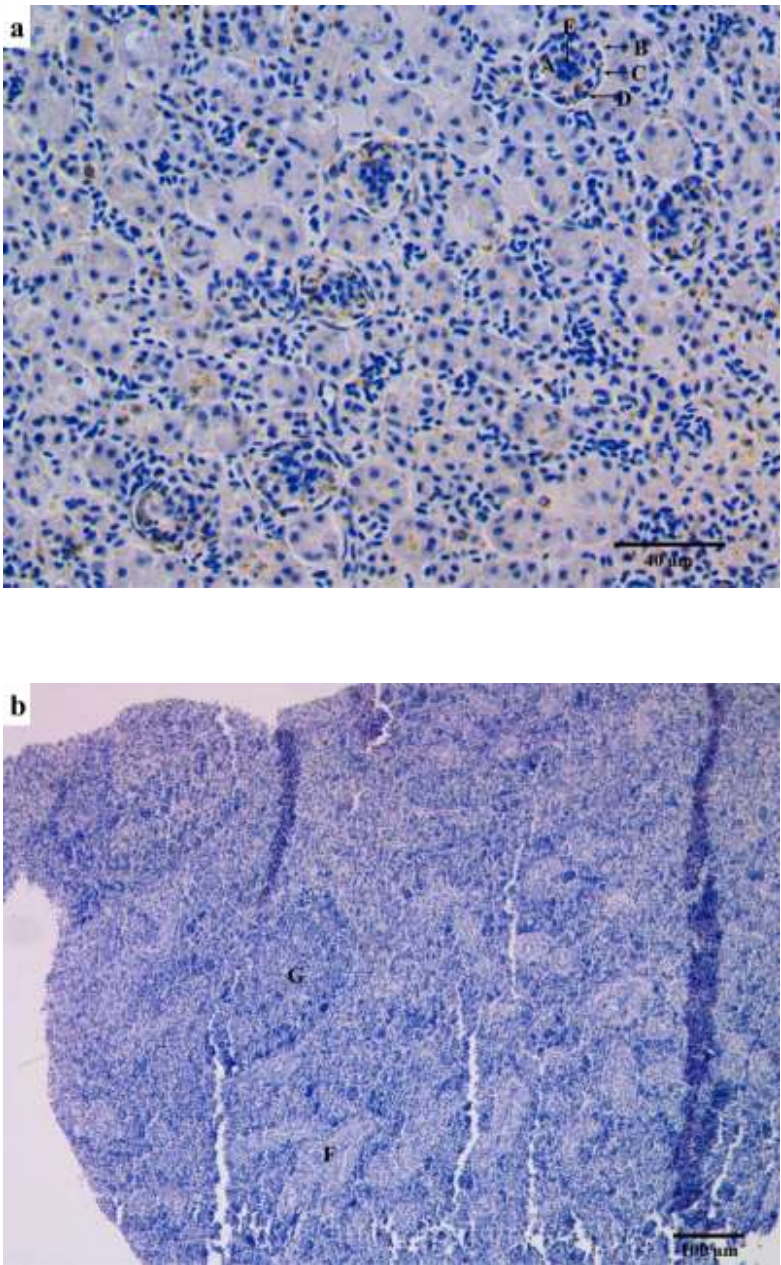


Fig (7): Hematoxylin and eosin staining of the kidney (a) and spleen (b) of a white-breasted waterhen bird. A, Glomerulus; B, Bowman's capsule; C, Visceral stratum; D, Bowman's space; E, Mesangial cells; F, white pulp; G, Red pulp.