

How to Cite:

Thaker, S. M., & Haba, M. K. (2022). Histological and histochemical study of the intestine in invasive species *Acridotheres tristis* in Iraq. *International Journal of Health Sciences*, 6(S5), 5380–5387. <https://doi.org/10.53730/ijhs.v6nS5.9802>

Histological and histochemical study of the intestine in invasive species *Acridotheres tristis* in Iraq

Saja M. Thaker

Mukhtar K. Haba

Abstract---The current study was conducted to find out the most important histological and histochemistry differences in the bird *A. tristis* intestine. The bird samples were dissected; to remove and fixed in 10% formalin, preparing them for histological and histochemical study. The results showed there was a histological differences in the bird intestine. Histological sections showed that the mucosa of *A. tristis* small intestine was presented numerous long villi which lined by simple columnar epithelium. In the bird large intestine the mucosa was presented numerous villi which lined by simple columnar epithelium. The histochemical sections of *A. tristis* small and large intestine showed that with Alcian blue (pH2.5) stain the epithelium of villi and intestinal crypts showed that the goblet cells contained acidic mucopolysaccharide. While the enterocytes showed negative reaction. With PAS stain, the sections of villi showed strong neutral mucopolysaccharid within goblet cells.

Keywords---*Acridotheres tristis*, invasive species, intestine.

Introduction

Biological invasion by non-native species is recognized as one of the major threats to native species and ecosystems (Pimentel et al 2000, Sala et al 2000). Such invasions into native communities are among the prime global change factors contributing to biodiversity decline (Sala et al 2000). According to the Fifth National Report to the Convention on Biological Diversity Iraq that consider species Myna and Tilapia as invasive species in the Iraqi environment (Haloob, 2014). One such invader is the Myna showing recent dramatic increases in its global distribution range (Holzapfel, 2006). The early record of Tilapia in the Iraqi waters was in Euphrates River near Musaib City, Centre region of Iraq (Saleh, 2007).

Omnivores that eat greater amounts of animals in their diet have a large stomach and a longer intestine. Omnivores that eat a great amount of plants typically have a smaller stomach and a long intestine (Zandonà, 2015). The avian alimentary canal has undergone a physiological structure in apposite to other animals to accommodate physical and chemical features of a wide variety of food types (Klasing 1999), and requirements for flight (Denbow 2015). The morphology of an organ system varies according to the feeding habit, habitat and nature of their life-style. This phenomenon is called adaptation (Tomar 2015). This study aimed to identify the histological and histochemical changes of the intestine in invasive species *Myna A. tristis*

Material and Methods

Adult samples of *Acridotheres tristis* from both sexes were used. The birds were obtained from pet stores, and checked for their health status before being euthanized. Intestine were immediately placed in a 10% buffered Formalin solution. After fixation, the tissue was dehydrated by transferring through a series of alcohols with increasing concentrations, cleared with xylene and embedded in paraffin. Sections of 5–7 µm in thickness were obtained. The sections were then subjected to haematoxylin and eosin (HE) staining for histological features, Masson's trichrome for connective tissue, Periodic Acid Schiff (PAS) for neutral mucins, Alcian blue (pH 2.5) for acidic mucins. Photomicrographs of the sections were captured using a digital camera attached to microscope.

Results

Anatomically the intestine is considered as the longer part of the digestive tract in *Myna*. The intestine located after the gizzard, the current study showed the total length of its intestine is about 30.5 cm, the intestine divided into two parts, small and large intestine each of which have a specialized function. The intestine is ended in a very narrow muscular tube known as the rectum which opened out the body in the anus in *A. tristis*.

The mucosa of *A. tristis* small intestine was presented numerous long villi which lined by simple columnar epithelium that composed of two types of cells: goblet cells and enterocytes (fig. 1A & 1B). The enterocytes of villi showed eosinophilic cytoplasm with marked brush border and the goblet cells showed clear lightly stained cytoplasm (fig. 1B).

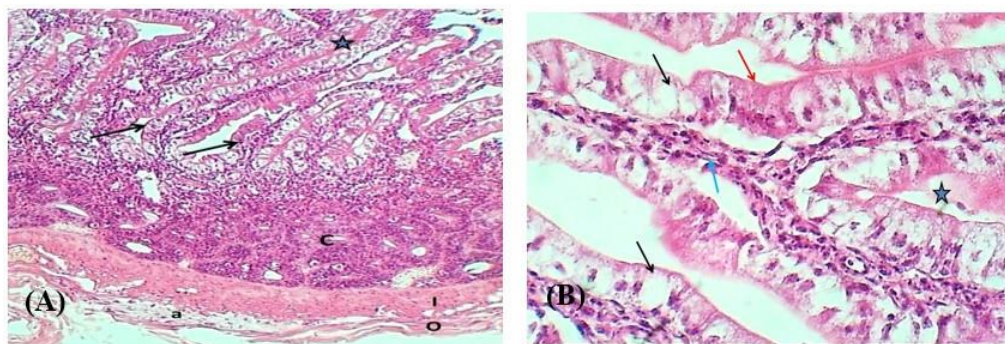


Figure 1: (A) Section of small intestine (duodenum) (Bird) shows: Villus of (asterisk), epithelium (Arrow), intestinal gland (C,), inner circular layer of muscularis (I) & outer longitudinal layer of muscularis (O) & aurbach plexuses (a) . H&E stain.40x. (B) Section of villus small intestine (Bird) shows: goblet cells (Black arrows), eosinophilc enterocyte (Red arrow), intestinal crypt (asterisk,), and fibrous lamina propria (blue arrow). H&E stain.400x.

In *A. tristis* the lamina propria was very thick layer that occupied by compound tubular glands (intestinal crypts), epithelial crypts were lined by the same population of epithelial cells (Fig. 1B). The tunica muscularis of *A. tristis* was presented inner circular smooth fibers and externally was presented longitudinally arranged smooth muscular fibers (fig. 4).

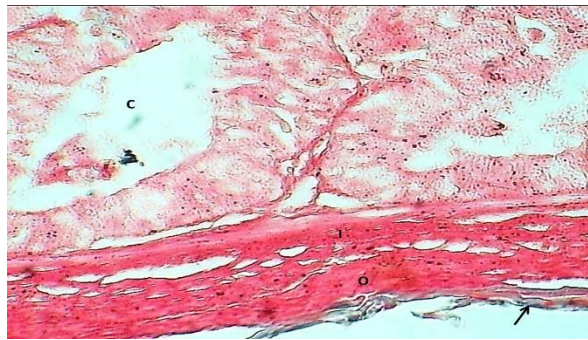


Figure 4: Section of wall of small intestine (Bird) shows: epithelial crypt (C), inner circular muscular layer (I), outer longitudinal muscular layer. (O) & tunica serosa (Arrow). Masson trichrom stain.400x.

The histochemical sections of *A. tristis* small intestine showed that with Alcian blue (pH2.5) stain the epithelium of villi and intestinal crypts showed that the goblet cells contained acidic mucopolysaccharide. While the enterocytes showed negative reaction (fig. 6). With PAS stain, the sections of villi showed strong neutral mucopolysaccharid within goblet cells. The (figs. 7) show weak neutral cytoplasm within enterocytes that stained with PAS.

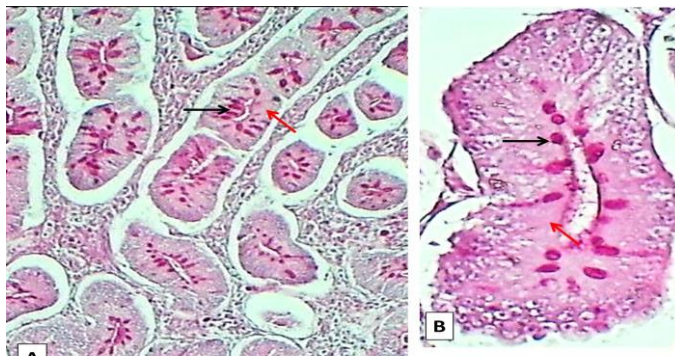


Figure 6: section of mucosa of small intestine (Bird): Micrographs (A & B) shows strong neutral secretion within goblet cells (Black arrows) & weak neutral secretion within enterocytes (Red arrows). PAS stain. 100 & 400x.

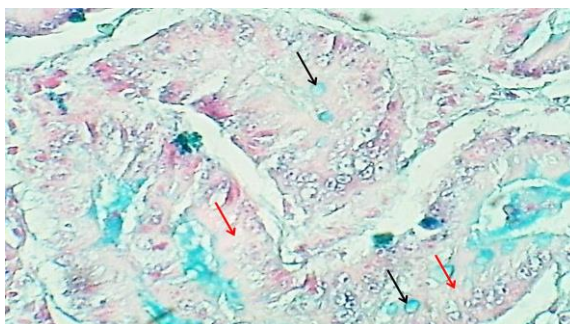


Figure 7: section of mucosa of small intestine (Bird) shows acidic mucopolysaccharid within goblet cells (Black arrows) & negative reaction with secretion enterocytes (Red arrows). Alcian blue (pH 2.5) stain. 400x.

The mucosa of the *A. tristis* large intestine was presented numerous villi which lined by simple columnar epithelium that composed of two type of cells: goblet cells and enterocytes (fig. 10A & 10B). The enterocytes were few in numbers and had marked eosinophilic cytoplasm with marked brush border while the goblet cells were predominated and showed clear lightly stained cytoplasm (fig. 10B).

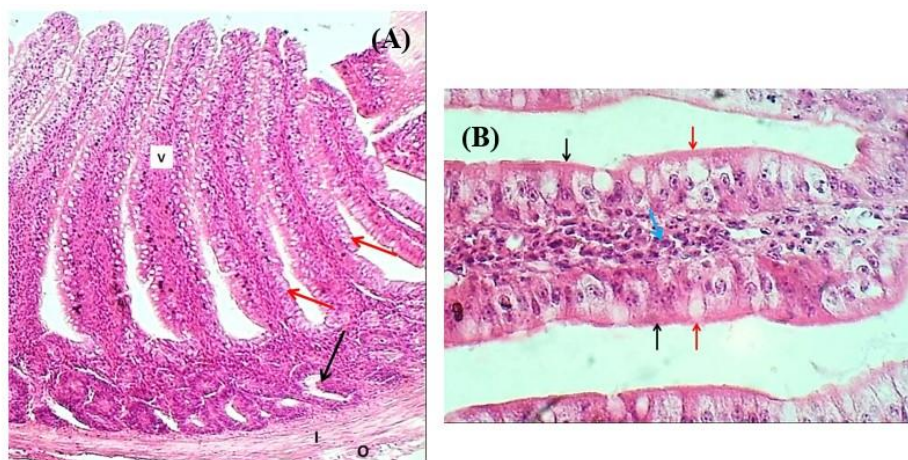


Figure 10: (A) Section of large intestine (Colon) (Bird) shows: Villus of (asterisk), epithelium (Red arrows), and thin layer of intestinal gland (black arrow), inner circular layer of muscularis (I) & outer longitudinal layer of muscularis (O). H&E stain. 100x. (B) Section of villus of large intestine (Bird) shows: goblet cells (Red arrows), eosinophilic enterocyte (Black arrow), crypt (asterisk), and cellular loose connective of lamina propria (blue arrow). H&E stain. 400x.

Lamina propria of *A. tristis* villus was occupied by compound tubular glands (intestinal crypts) which showed few goblet cells (Fig. 13). The tunica muscularis of *A. tristis* was continuous for that in small intestine (fig. 14).

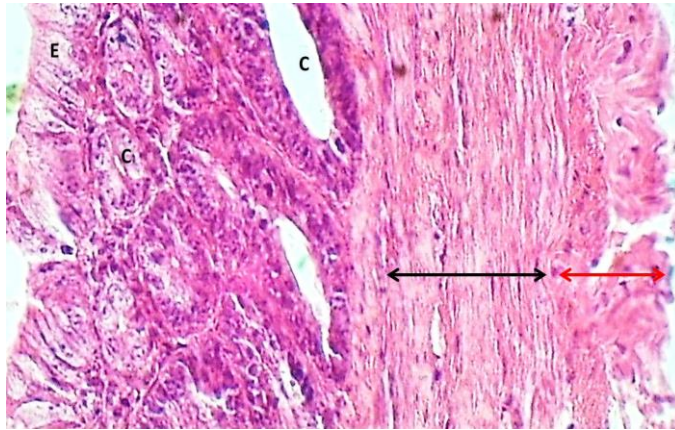


Figure 13: Section of wall of large intestine (Bird) shows: epithelium (E), epithelial crypt (C), inner circular muscular layer (black double arrow), outer longitudinal muscular layer (Red double arrow).H&E stain.400x.



Figure 14: Section of wall of large intestine (Bird) shows: epithelial crypt (C), inner circular muscular layer (black double arrow), outer longitudinal muscular layer (Blue double arrow).Masson trichrom stain.400x

The histochmical sections of *A. tristis* large intestine showed that with Alcian blue (pH2.5) stain the epithelium of villi and intestinal crypts showed that the goblet cells contained acidic mucopolysaccharide while the enterocytes showed negative reaction (fig. 16). Meanwhile with PAS stain, the sections of villi showed strong neutral secretion within goblet cells and no reaction within cytoplasm of enterocytes (fig.16).

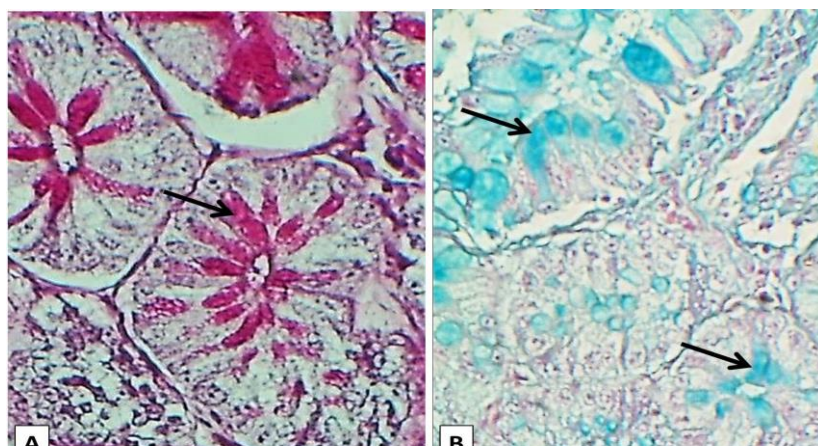


Figure 16: Section of mucosa of large intestine (Bird): Micrographs (A) with PAS stain shows strong neutral mucopolysaccharid within goblet cells (Black arrows) & weak neutral secretion within enterocytes. 400x. Micrographs (B) with Alcian blue shows strong acidic mucopolysaccharid within goblet cells (Black arrows) & no reaction within enterocytes. 400x.

Discussion

The intestine length of *A. tristis* is about 30 cm, this result can be explain according to the food texture that the samples feed on it, since the bird *A. tristis* consider as omnivorous that feed on insects, grains, fruits. The mucosa of the small intestine was presented numerous long villi which lined by simple columnar epithelium as showed by (Hanafy, 2020) when he studied on the intestine of Garganey that composed of two type of cells: goblet cells, the function of goblet cells (mucocytes) to producing a secretion that contains mucopolysaccharides have an important protective role against chemical irritants and microorganisms (barrier function) as well as a transporting role of substances between the intestinal lumen and epithelial microvilli (Duritits, 2016). The histochemical sections in the recent study of the small intestine showed that with Alcian blue (pH2.5) stain the epithelium of villi and intestinal crypts showed that the goblet cells contained acidic mucopolysaccharide, The acid mucopolysaccharides possess a higher viscosity and they play a very important role in the formation of a protective barrier against microorganisms and ensure protection against mechanical irritation (Piel, 2005). While the enterocytes showed negative reaction with Alcian blue stain. The villi with PAS stain showed strong neutral mucopolysaccharid within goblet cells, the neutral mucopolysaccharides are less viscous, and facilitate an easier feed mass transit along the alimentary tract (Piel, 2005). The same results are showed in the large intestine of *Acridothores tristis* in which the mucosa have numerous villi which lined by simple columnar epithelium that composed of two type of cells: goblet cells and enterocytes, the enterocytes were few in numbers and had marked eosinophilic cytoplasm with marked brush border while the goblet cells were predominated. The sections of large intestine showed that with Alcian blue (pH2.5) stain the epithelium of villi and intestinal crypts showed that the goblet cells contained acidic mucopolysaccharide while the enterocytes showed negative

reaction. Meanwhile with PAS stain, the sections of villi showed strong neutral secretion within goblet cells and no reaction within cytoplasm of enterocytes. The lamina propria of villus was occupied by compound tubular glands (intestinal crypts) which have few goblet cells and, the tunica muscularis was continuous for that in small intestine. According to Caspary (1992) the increased villus height suggests an increased surface area capable of greater absorption of available nutrients. It is understood that greater villus height and numerous cell mitoses in the intestine are indicators that the function of the intestinal villi is activated

Intestinal length and the number of villi increase the number of enterocytes. Enterocytes are often attached to each other by tight junctions. Together enterocytes and tight junctions form a continuous barrier that regulates both trans-cellular and para-cellular diffusion of molecules, thus constituting the principal component of the intestinal primary barrier (Okuthe, 2021).

References

- Caspary WF. Physiology and pathophysiology of intestinal absorption. *The American journal of clinical nutrition*. 1992 Jan 1;55(1):299S-308S.
- Denbow DM. Gastrointestinal anatomy and physiology. In *Sturkie's avian physiology* 2015 Jan 1 (pp. 337-366). Academic Press.
- Duritis I, Mugurevics A. Distribution and Characterisation of Goblet Cells in the Large Intestine of Ostriches during the Pre-and Post-Hatch Period. *Anatomia, histologia, embryologia*. 2016 Dec;45(6):457-62.
- Haloob A. Fifth National report to the Convention on Biological Diversity Iraq, 2014
- Hanafy BG, Abumandour MM, Bassuoni NF. Morphological features of the gastrointestinal tract of Garganey (*Anas querquedula*, Linnaeus 1758)—Oesophagus to coprodeum. *Anatomia, histologia, embryologia*. 2020 Mar;49(2):233-50.
- Holzappel CL, Levin NO, Hatzofe OH, Kark SA. Colonisation of the Middle East by the invasive Common Myna *Acridotheres tristis* L., with special reference to Israel. *Sandgrouse*. 2006 Jan 1;28(1):44.
- Khanna SS, Mehrotra BK. Morphology and histology of the teleostean intestine. *Anatomischer Anzeiger*. 1971;129(1):1-8.
- Klasing KC. Avian gastrointestinal anatomy and physiology. In *Seminars in avian and exotic pet medicine* 1999 Apr 1 (Vol. 8, No. 2, pp. 42-50). WB Saunders.
- Okuthe GE, Bhomela B. Morphology, histology and histochemistry of the digestive tract of the Banded tilapia, *Tilapia sparrmanii* (Perciformes: Cichlidae). *Zoologia (Curitiba)*. 2021 Jan 18;37.
- Piel C, Montagne L, Sève B, Lallès JP. Increasing digesta viscosity using carboxymethylcellulose in weaned piglets stimulates ileal goblet cell numbers and maturation. *The Journal of nutrition*. 2005 Jan 1;135(1):86-91.
- Khidoyatova, M. R., Kayumov, U. K., Inoyatova, F. K., Fozilov, K. G., Khamidullaeva, G. A., & Eshpulatov, A. S. (2022). Clinical status of patients with coronary artery disease post COVID-19. *International Journal of Health & Medical Sciences*, 5(1), 137-144. <https://doi.org/10.21744/ijhms.v5n1.1858>
- Pimentel D, Lach L, Zuniga R, Morrison D. Environmental and economic costs of nonindigenous species in the United States. *BioScience*. 2000 Jan;50(1):53-65.

- Sala OE, Chapin FS, Armesto JJ, Berlow E, Bloomfield J, Dirzo R, Huber-Sanwald E, Huenneke LF, Jackson RB, Kinzig A, Leemans R. Global biodiversity scenarios for the year 2100. *science*. 2000 Mar 10;287(5459):1770-4.
- Tomar MP, Joshi HR, Ramayya PJ, Vaish R, Shrivastav AB. Avian Esophagus: A comparative microscopic study in birds with different feeding habits. *International journal of Medical and Health sciences*. 2015;9(8):6.
- Suryasa, I. W., Rodríguez-Gámez, M., & Koldoris, T. (2022). Post-pandemic health and its sustainability: Educational situation. *International Journal of Health Sciences*, 6(1), i-v. <https://doi.org/10.53730/ijhs.v6n1.5949>
- Zandonà E, Auer SK, Kilham SS, Reznick DN. Contrasting population and diet influences on gut length of an omnivorous tropical fish, the Trinidadian guppy (*Poecilia reticulata*). *PLoS One*. 2015 Sep 11;10(9):e0136079.