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HISTOMORPHOLOGICAL AND HISTOMETRICAL STUDY OF SMALL INTESTINE OF THE GUINEA FOWL, *NUMIDIA MELEAGRIS*

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ABSTRACT : The current study was aimed to explore the histological structures of small intestine in male and female of guinea fowl. The histological structure of the small intestine was similar in both male and female guinea fowl and the wall of small intestine along the entire length contained four tunicae named tunica Mucosa, Submucosa, Muscularis and Serosa from inner to outer. The mucosa of the small intestine was thrown into projections known as villi. The shape and pattern of small intestinal villi was varied in different segments of the small intestine and these were lined by columnar and goblet cells. The density of goblet cells were increase from duodenum toward the ileum. The Lamina propria formed the bulk of core of villus and contained connective tissue fibers and cells. The submucosa was thin and poorly developed and almost difficult to disguished except it noticed when the large blood vessels were present and separated the mucosa from the underlying muscularis externa. Brunner glands not observed in the duodenal submucosa. Tunica muscularis presented in an inner circular and outer longitudinal smooth muscle bundles. *Tunica serosa* was predominantly composed of collagen fibers. The tunicae thickness in different regions of the small intestine showed significant variation at ($p \leq 0.05$) between male and female guinea fowl.

Key words : *Numidia meleagris*, intestine, histometrical study.

INTRODUCTION

Guinea fowl belongs to the family Numididae, order Galliformes (Dyke *et al,* 2003; Haaroma, 2003). Guinea fowls considered as a source of high-value of meat and eggs and more resistance for disease than other types of birds (Zvakare *et al*, 2017). Its easy maintenance, early sexual maturity, shorter generation interval and high rate of egg production has become a pilot animal in the field of research. Small intestine of birds plays a very important role in much of digestion and all of the absorption (McLelland, 1979). As a general the digestive system anatomy and histology of domestic bird is quite different from those of mammals. In addition, there are many structural differences among avian species according to their feeding habits (Getty, 1975; Nickel *et al*, 1977; Karadað and Nur, 2002; Haligür, 2008; Elsheikh *et al*, 2017).

The small intestine is the heaviest structure within the gastrointestinal tract and are located near the bird's center of gravity within the abdominal cavity. The bird intestine has a major influence on growth performance as it affects feed digestion, nutrient absorption and mortality, the small intestine which consisted of duodenum, jejunum and ileum, is relatively simple and short but highly efficient nevertheless (Nasrin *et al*, 2012). The digestive system of multi-cellular organisms converts their ingested food material into nutrients that require for their maintenance, growth and production. In birds, the process of digestion takes place by mechanical and chemical action on their ingested food material. The current study aimed to explore histomorphological and histometrical of the small intestine of guinea fowl.

MATERIALS AND METHODS

The current study was carried out on 10 adult birds of both sex (male and female). The coelomiccavity of the birds were opened under anesthesia by pentobarbitone (80 mg/kg body weight), then the birds of both sex were sacrificed and the small intestine was collected and flushed with fresh normal saline. Small tissue pieces of 1cm length from the middle region of duodenum, jejunum and ileum were collected, and fixed with 10% neutral buffered formalin, dehydrated in ascending grades of alcohol, cleared in xylene and embedded in paraffin. Then the sections of 6-7 µm were stained with Mayer's hematoxylin

and eosin stain for general histological structure, Masson·sTrichrom stain for collagen fibers (Luna, 1968; Singh and Sulochana, 1996; Suvarna *et al*, 2013). The layer thickness of small intestine segments were done by using ocular micrometer after calibration and the Statistical analysis was done by t test.

RESULTS

The microscopic examination of the entire small intestine (duodenum, jejunum and ileum) of the male and female guinea fowl showed the presence of four layers of a tubular organ that were: Mucosa, Submucosa, Muscularis and Serosa (Fig. 1). As reported by Hamdi *et al* (2013) in black winged kite and Al-Bideri and Jawad (2015) in rock dove. The duodenal tunica mucosa presented in a finger like villi covered by a simple columnar epithelium with few of goblet cells (Fig. 3) followed by lamina propria. themuscularis mucosae was arranged into longitudinal smooth muscle bundles (Fig.3) this findings were similarly documented in previous study in brown falcon by Al-Taee (2017).

The characteristic features of the duodenal mucosa were the villi and the intestinal glands the villi appeared as tall finger –like mucosal projections and others appears as a leaf like projections, its corewere filled by the lamina propria, longitudinal bundles of smooth muscle fibers and blood vessels. There is no significant difference in between male and femalein the villus height (Tables 1, 2, 3), as observed by Mohamed and Hassasn (2008). The mean crypts depth in female was higher than in male. The intestinal epithelial cells are change constantly and compensate villi cells losses through proliferation and maturation inside crypts and upward migration. The crypts depth was correlated with the intestinal cells turnover rate and the increase in crypts depth indicates the need for enterocyte replacement and higher tissue turnover (Oliveira *et al*, 2009). The crypt depth may be an important factor that determines the ability of the crypts to sustain the increase in the villus height as well as to maintain the villus structure (Poole *et al*, 2003). The ratio of villus height to crypts depth was slightly hieghr in female than in male. The increase of villus height to crypts depth associated with better nutrient absorption and faster growth (Wu *et al*, 2004).

Mucosa of the jejunum was modified into different size and shapes of villi display as leaf like projection, arranged in azig-zag design or displayed blunt or pointed apical end with wide basal portion (Fig. 4) and they appears as a wider or shorter than that of that observed in the duodenum as found by Al-Saffar and Al-Samawy

Table 1 : The Microscopic measurement (Mean ±SE) of the height of columnar cells, villus length, crypts depth, the ratio of villus height to crypts depth and villus width of duodenum in males and females guinea fowl.

Duodenal measurements (µm)	Male (Mean±SE)	Female (Mean ±SE)	T test
Height of columnar cells	28.2±0.20[a]	28.8±0.10[a]	0.640
Villus height	998±2.9[a]	999.2± 2.10[a]	0.339
Crypts depth	174±1.11[a]	174.2± 0.44[a]	0.125
Ratiovillus height crypts depth	5.71[a]	5.73[a]	0.106
Villus width	102.2±0.44[a]	101.8±0.22[a]	0.284
Thickness of *Tunica mucosa*	1270±5.59[a]	1268.6± 6.67[a]	0.053
Thickness of *Tunica submucosa*	26.2±0.20[a]	25.8± 0.49[a]	0.775
Thickness of *Tunica musclaris*	1739.6± 1.37[a]	1736.6± 0.67[a]	0.298
Thickness of *Tunica serosa*	59.8±0.56[a]	60.4±0.44[a]	0.341

The similar letters mean non-significant differences (P≤ 0.05) between male and female guinea fowl.

Table 2 : The Microscopic measurement (Mean± SE) of the height of columnar cells, villus length, crypts depth and the ratio of villus height to crypts depth of jejunum in males and females guinea fowl.

Jejunal measurements (µm)	Male (Mean ±SE)	Female (Mean ±SE)	T test
Height of columnar cells	26.8±0.22[a]	26.4±0.86[a]	0.066
Villus height	497.2±0.99[a]	499.4±0.13[a]	0.570
Crypts depth	147.0±1.32[a]	147.4±0.89[a]	0.464
Ratiovillus heightcrypts depth	3.382[a]	3.388[a]	0.012
Villus width	122.2±2.78[a]	124.6±1.56[a]	0.876
Thickness of *Tunica Mucosa*	766.6±4.03[a]	767.2±4.24[a]	0.196
Thickness of *Tunica submucosa*	13.9±0.22[a]	14.4±0.09[a]	0.017
Thickness of *Tunica musclaris*	856.4±0.89[a]	857.2±0.14[a]	0.122
Thickness of *Tunica serosa*	51.4±0.67[a]	51.2±0.44[a]	0

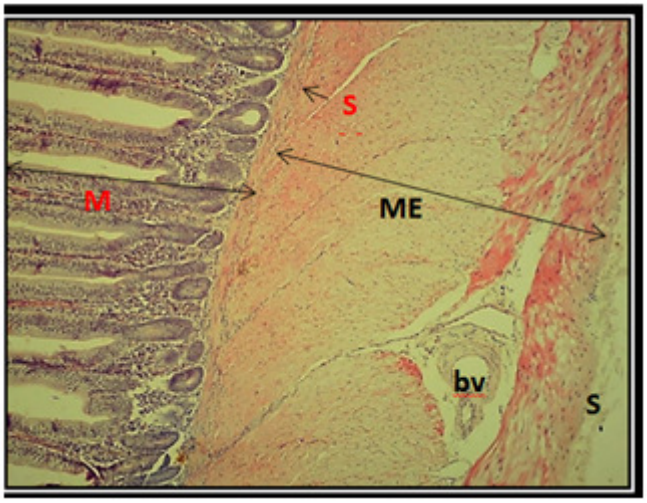

Fig. 1 : Histological section of the layers of the duodenum of male guinea fowl shows: Mucosa(M), Sub mucosa(SM), Muscularis Extern (ME), blood vessels(bv) and Serosa(S) (H&E) (X 100).

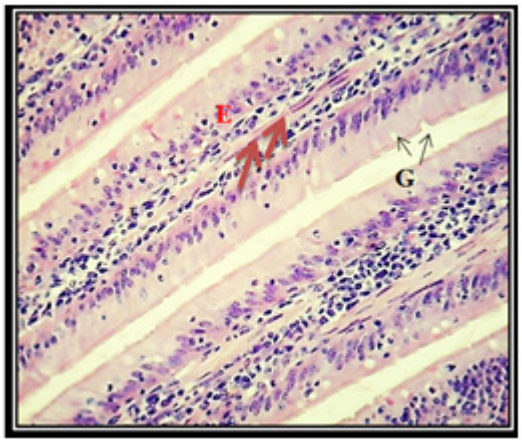

Fig. 2 : Histological section of the duodenum in female guinea fowl shows: Simple Columnar Epithelium (E), smooth muscle fibers (red arrow) and Goblet cells (G) (H & E) (X400).

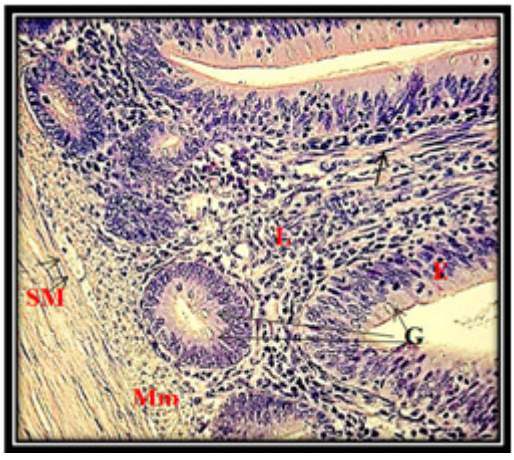

Fig. 3 : Histological section of the duodenum in male guinea fowl shows: Epithelium (E), Goblet cells (G), lamina propria (L), *Muscularis mucosa* (Mm), smooth muscle fiber (black arrow) and Intestinal glands (I) (H & E) (X400).

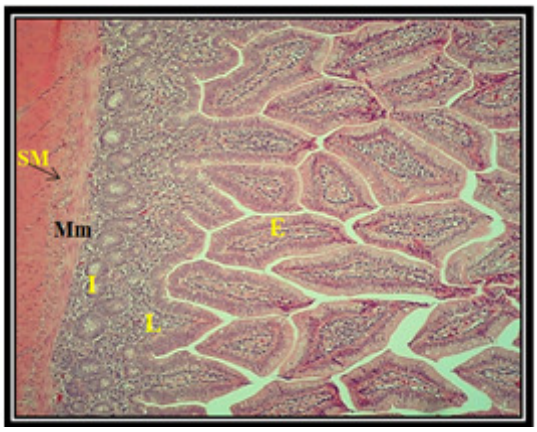

Fig. 4 : Histological section of leaf- like villi of the jejunum in female guinea fowl shows: Epithelium (E), lamina propria (L), Intestinal gland (I) and Musclaris mucosa(Mm), Submucosa (SM) (H&E) (X100).

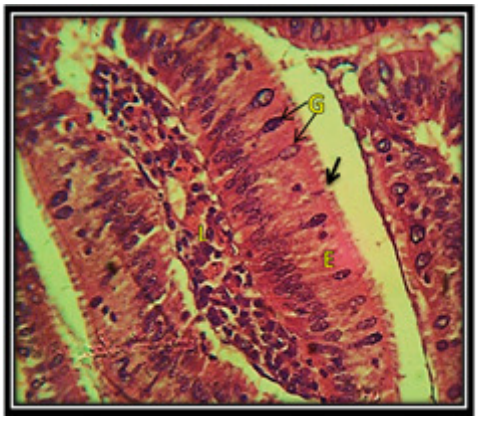

Fig. 5 : Histological section of the jejunum in female guinea fowl shows: Epithelium (E), Goblet cells(G), *Lamina propria* (L) and Brush border (black arrow) (PAS) (X400).

(2016) and Moreki (2009) in owl and ostrich.

The mucosa of the small intestine presented villi, which varied in size and shape in different segments as observed by Iji *et al*, (2001).

The epithelium of villi in all segments of the small intestine and glands was lined by single layer of columnar cells which contained oval to elongated nucleus towards the base of the cells. The free borders of these cells with brush border (Fig. 5). In between columnar cells, typical goblet cells were observed which increased in its density from the duodenumtoward the ileum (Fig. 5). The free borders of these cells were striated due to presence ofbrosh border as reported earlier by Iji *et al* (2001) in chicken in coturnixquails (Ahmad *et al,* 2012), fowl (Hodges, 1974). A simple tubular intestinal glands were present in base of villi and opened between the bases of the villi. The epithelium lining of these glands was the same epithelium covering the villi, which was simple columnar cells and goblet cells (Fig. 3) as reported by AL Sheshani (2006) in *Accipiter nisus* Linnaeus.

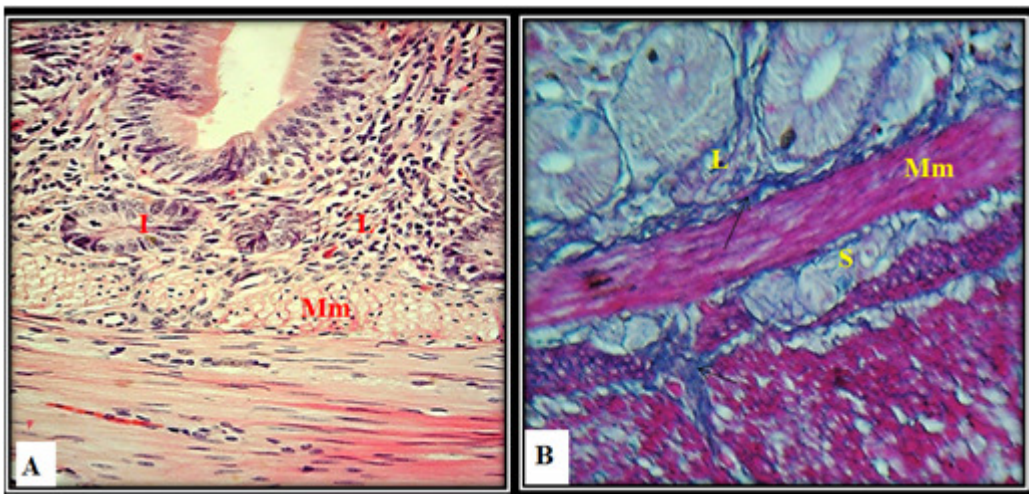

Fig. 6 : Histological section of the ileum (A & B)in male guinea fowl shows: collagen fiber (black arrow), *Lamina propria* (L), Musclaris mucosa (Mm) and Submucosa (SM), A(H&E), B (Masson,sTrichrom) (X400).

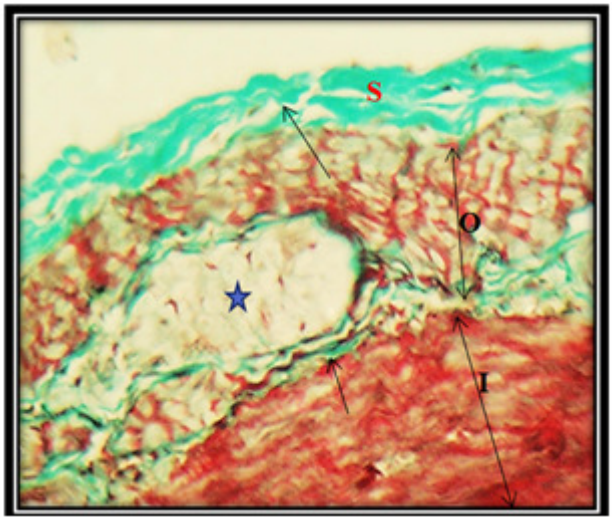

Fig. 7 : Histological section of the ileum in female shows: Collagen fibers (black arrow), Serosa(S), Inner circular(I), Outer longitudinal(O) and Nerve Plexus(blue star) (Masson's Trichrom) (X 400)

Lamina propria of all segments of the small intestine was consisted of cellular connective tissue with a few collagen fires, smooth muscle fibers and numerous capillaries. Underneath the mucosa, the submucosa was poorly developed and when observed it structured of thin layer of loose connective tissue contained blood vessels (Fig. 6). This finding was a good agreements with that observed by McLelland (1979), Hodges (1974) in fowl and (Kachave, 2009) in broiler and layers (Al-Samawy, 2015) in pigeon, but different in the mallard and owl in which this layer was appeared as a thick layer. The mean thickness of submucosa was lesser than that observed by Al-Samawy (2015), Al-Saffar and Al-Samawy (2016) in owl and pigeon and higher than recorded in blue and yellow macaws, mallard, barn owl and Brown falcon recorded by Rodrigues *et al* (2012), Dawood (2013), Al-Taee (2017), the thickness of this layer was higher in male than in female Table 1. The absence of Brunner gland in the duodenal submucosa was confirmed the

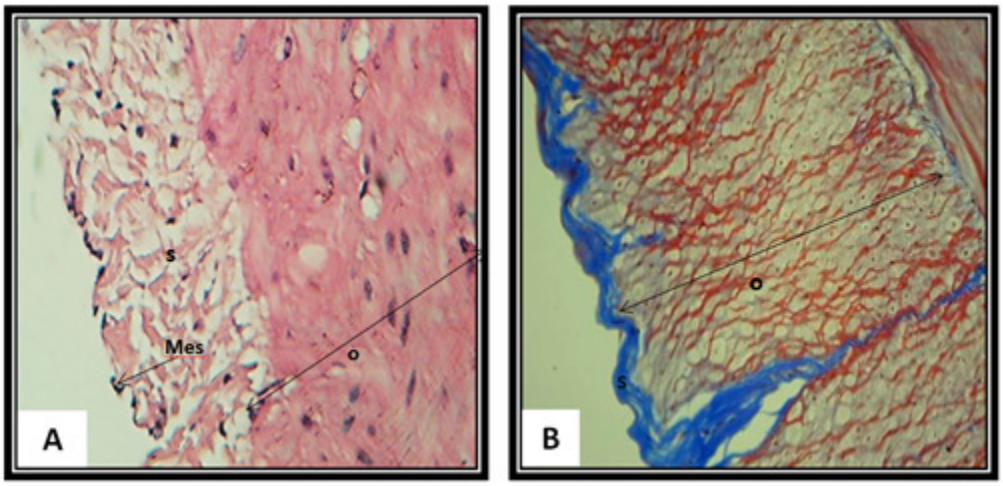

Fig. 8 : Histological section of the duodenum in male guinea fowl shows: Inner circular (I), Outer Longitudinal (O) Mesothelium (Mes) and Serosa (S) (A H&E) and (B Masson's Trichrom) (X 400).

Table 3 : The Microscopic measurement (Mean± SE) of the height of columnar cells, villus length, crypts depth and the ratio of villus height to crypts depth of Ileum in males and females guinea fowl.

Ileum measurementsμm	MaleMean ±SE	FemaleMean ±SE	T test
Height of columnar cells	26.2±0.67[a]	25.6±0.22[a]	0.775
Villus height	329±3.94[a]	328.8± 4.24[a]	0.017
Crypts depth	60±0.44[a]	59.8± 0.67[a]	0.083
Ratiovillus heightcrypts depth	5.48[a]	5.49[a]	0.016
Villus width	122±2.42[a]	123.8±1.11[a]	0.605
Thickness of Tunica mucosa	422.6± 1.11[a]	423.2±3.35[a]	0.133
Thickness of Tunica submucosa	15± 0.64[a]	15.8 ±0.44[a]	0.749
Thickness of Tunica musclaris	465± 0.64[a]	463± 1.52[a]	0.125
Thickness of Tunica serosa	52± 1.38[a]	52.6± 1.11[a]	0.178

The similar letters mean non-significant differences (P≤ 0.05) between male and female guinea fowl.

previous finding in chicken by Aitken (1958) and Kalita *et al* (2012) in kadaknth fowl who mentioned lacking of duodenal Brunner's glands and their mucus secreting role was carried out by numerous goblet cells present between the enterocytes of the surface epithelial villi and the intestinal glands.

The muscularis mucosa was arranged in one longitudinal layer of smooth muscle fibers (Fig. 6) as reported earlier in quails (Fitzgerald, 1969) and fowl (Hodges, 1974; McLelland, 1979) as reported in mallaredand kadaknath fowl by Dawood (2013) and Kalita *et al* (2012) also in owl which the muscularis mucosa arranged in one longitudinally arranged of smooth muscle fibers, but differently observed in African pied crow the muscluris mucosa was absent (Igwebuike and Eze, 2010).

Tunica muscularis in all segments of the small intestine was made up of two layers of smooth muscle fibers arranged in a well-developed inner circular layer which was appeared thicker than the outer thin longitudinal layer. Between the two layers a narrow connective layer contained blood and lymphatic vessels as well as a nerve plexus (Fig. 7). Thickness of tunica muscularis appeared to be decreased from duodenum to ileum and in there is no significant difference between male and female Tables 1, 2, 3. This muscular coat was constructed of an thick inner circularly and an thin outer longitudinally arrangement of smooth muscle bundles.

Thickness of tunica muscularis appeared to be higher in duodenum than jejunum and ileum as observed in fowl (Hodges, 974). However, Sivakumar and Vijayaragavan (1989) reported an increased thickness of tunica muscularis from duodenum to ileum in Japanese quail.

Tunica serosa was relatively a thin layer of connective tissue especially contain collagen fibers, adipose tissue and blood vessels (Fig. 8). *Tunica serosa* was relatively a thin layer of loose connective tissue contains a collagen fibers, adipose tissue and blood vessels covered by mesothelium as reported by by Khaleel and Atiea (2017) in mallard.

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