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# Morphohistometrical study for prevntriculus in pre and post-hatching broiler chicks

#### Hind Hussein Kadhim

Department of Anatomy and Histology, College of Veterinary Medicine, Al-Qasim Green University, Babylon, Iraq Corresponding author email: hind.hussien@vet.uoqasim.edu.iq

#### Isam M. J. Zabiba

Department of Anatomy and Histology, College of Veterinary Medicine, Al-Qasim Green University, Babylon, Iraq

> Abstract --- The present work is designed for morphological and histological study to evaluate the development and growth of the proventriculus of pre-hatching and post-hatching chicks Ross 308 of the proventriculus in the 11th, 15th days of pre-hatching, and 7th, 14th days of post-hatching of broiler chicks Ross 308, the avian stomach has two chambers. The first chamber is known as the proventriculus (glandular), and it is where gastric juices are produced. The second chamber is a mechanical part known as the ventriculus or gizzard. and was undertaken on 60 chick embryos obtained from incubated broiler chick eggs Ross 308 (Gallus gallus domesticus) 30 chick embryos were collected at stage 37 (Hamburger and Hamilton, 1951) Hand H (11 days of pre-hatch ) and 30 chick embryos were collected( H and H) at stage 41 (15 days of pre-hatch). 60 chicks were collected at 7 days and 14 days post-hatching. The proventriculus color, shape, and position had been revealed by a morphological study. The length, width, and weight total of the proventriculus were progressively increased to use Electronic Vernia and electronic balance. Chicks of proventriculus were formed from four tunics in this histological study: tunica mucosa, tunica submucosa, tunica muscularis, and tunica serosa. The results reveal that the simple tubular glands of the proventriculus were poorly defined at embryonic days 11 and 15, while the framework of the compound tubular glands of the proventriculus was fully established. At embryonic days 11 and 15, simple cuboidal cells lined the secretory tubules of compound tubular glands and increase of the tunica muscularis from 2 to 3 layers during growth.

*Keywords*---prevntriculu, hatching broiler chicks, morphological study.

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## Introduction

Through its early stages of development, the vertebrate digestive tract is a simple tube composed of endodermal epithelium and surrounding mesenchyme. The anteroposterior axis defines distinct digestive organs, which the chicken stomach is separated from the caudal gizzard by a cranial proventriculus. The proventriculus is distinguished by the formation of compound glands that produce digestive enzymes. Around day 6 of pre-hatch, the epithelium begins to invade the surrounding mesenchyme to form simple glands observed by (Tabata and Yasugi, 1998).During the development of a chick's proventriculus, the gut endoderm differentiates into glandular and luminal epithelium digestive system organs (glandular stomach). The embryonic gastrointestinal tract undergoes significant structural and functional changes, which involve several processes such as endoderm induction and patterning, as well as mesoderm recruitment observed by (Grappin-Botton and Melton, 2000; Stainier, 2002). This begins with the formation of the intraembryonic tube, also revealed as the primitive gut, which also is lined by endoderm and covered by splanchnic mesoderm at either end of the embryo (Grappin-Botton and Melton, 2000). The primitive tube had been eventually divided into three parts: the foregut, midgut, and hindgut (McGeady et al., 2006), with the foregut forming the esophagus, crop, proventriculus, and gizzard, the midgut forming the small intestine, and the hindgut forming the large (Drucilla etal., 1998). The chicken digestive tract active transport food to the stomach. The proventriculus and gizzard are first important sites of enzyme activity. In general, the gastrointestinal tract of birds varies from one bird to the next depending on nutrition and strains that the birds belong to. When compared to mammals, the stomach of birds is modified depending on diet nature recorded (Davis, 2007). Because of glandular stomach is located ahead of the muscular stomach in birds, the shape of the glandular stomach differs (Thomson, 1996). In the chick, the proventricular stomach is spindly and connects to the esophagus (King and Mclelland, 1975). The glandular's internal surface (proventriculus) is characterized by raised gastric papillae on the upper surface This papillae secreted gastric juices, were also a combination of digestive enzyme, hydrochloride acid, and mucin (Ahmed et al., 2009 and Sherri, 2003). The proventriculus was categorized into 4 layer or tunica tunics depending on the type of ingested: tunica mucosa, tunica submucosa, tunica muscularis, and tunica serosa, and the diet composition, feeding habits, and quantity.

### **Materials and Methods**

An experiment is carried out on 120 commercially fertile broiler hatching eggs. The current study used broiler meat type (ROSS 308) obtained from the AL-Nasar company. We will use where it is placed in a small incubator and the incubation was performed in an automatic incubator at a controlled temperature  $(37.5^{\circ}C)$  and humidity (65%) Candling was used to examine the eggs. The chicks were separated into two groups: Pre-hatching and take 60 samples from the embryos of the chicks at ages (11,15) days, morphological study the sample pass through routine histological technique in order to perform histological slides: embryo will be examined proventriculus for these two main groups, and post-hatching 60 chicks divided into 2 groups at ages (7,14) days. Cervical dislocation killed all of the chicks after each sample of 30 chicks was examined proventriculus

morphological study, for these two groups the chicks were collected and fixed in fixative, formalin 10% the samples were pass through routine histological technique in order to perform histological slides.

## **Results and Discussion**

### Morphology Proventriculus at Pre -hatch and Post-hatch

Birds have two stomachs, one glandular known as the proventriculus and one muscular known as gizzard. The stomach of broiler chicks was divided into 2 based on morphological evaluation: the glandular compartment parts (proventriculus) and the muscular compartment (gizzard), in histological distinctions, proventriculus morphological was discovered in those other species (Zaher et al., 2012; Adel and Zabiba, 2021), these two cavities are separated by an intermediate zone (isthmus) and disagree (Hodge, 1974). who observed that the stomach of birds is divided into 3 compartments: proventricular, ventricular, and pyloric. This discovery was similar to the current observation of quail (Ahmed et al., 2011;Al-Khakani et al., 2020). The embryos of broiler chicks showed morphological development from pre-hatch 11 days to 14 days (7,14) post-hatch. The proventriculus was small tubular elongated to a spindle shape and creamy colored pre-hatching, changing to the color is reddish brown post-hatching. This result was similar to those observed by (Hassouna, 2001), in chickens, in which the demarcation between proventriculus and esophagus in pre and post-hatching chicks was not recognized by the smooth surface of the esophagus. In addition to thickness, the proventriculus wall was thicker than the esophagus wall in posthatch.

### **Morphometric measurements of Provetriculus**

The proventriculus in broiler chicks increases mean length, width, and weight gradually from day 11 pre-hatch to day 14 post-hatch, gradually. The proventriculus in 11 days, 15 days pre-hatch, 7 days, and 14 days post-hatch in the table(1), (Murakami *et al.*,1992), observed in broiler chickens that the digestive system growth between the 3 and 7 days post-hatch. Post-hatching length was 17.8  $\pm$ 1.788 mm, and width was 10.6 $\pm$ 0,894 mm. According to (Jassem et al., 2016 ; Al-Khakani *et al.*, 2018), the proventriculus in adult common moorhens was tubular in shape, with an average length and width of (20.00  $\pm$ 7906mm) and (10.48 $\pm$ 1.53194mm) respectively. Similarly, their mean lengths in 7 days were 14.47 0.21 mm in Kadaknath Fowl observed by (Das *et al.*, 2013).

### Histological Proventriculus Pre -hatch and Post-hatch

At stage 37 H&H (11 days of incubation),histological sections taken pre-hatch revealed that the proventriculus wall in broiler chicks was composed of 4 classic tunica or layers : tunica mucosa, tunica submucosa, tunica muscularis, and tunica serosa (Fig. 1). Shows the surface of tunica mucosa of proventriculus was lined by simple columnar epithelium (Fig.1). This is similar to the findings by (Hassan and Moussa ,2012) in pigeon and duck, and it differs from the research results of( Banks ,1992) in fowl, who stated that it is cuboidal. Connective tissue

and smooth muscle fibers scattered between the proventricular glands ,Simple tubular glands were found in the lamina propria, whereas compound or multilobular glands were found in the sub mucosa (Fig.3). The muscularis was composed of 2 clearly layers: an internal circular layer that was thin as well as an external longitudinal layer that was thick (Fig.2 ). Stage 41 H&H (15 days prehatch) .The diameter of the proventricular glands increased with age. The connective tissue and blood vessels, as well as the deep proventricular glands, make up the submucosa. these conic or pear-shaped glands account for a sizable proportion of the proventriculus wall thickness (Fig. 4,6). The recent study were similar in other birds such like quail (Attia, 2008) and broilers (Nasrin et al., 2012). The glands of the proventriculus they are separated by septa of connective tissue of various ages, the glands formed lobules, The epithelium of these tubules was cuboidal appearance, which converged near the surface into a common cavity (Fig.6). Previously, similar findings were discovered by (Ibrahim et al., 2020), in the Dandarawi chick. 7 days post- hatching, the proventriculus tunica mucosa was folded and lined by simple columnar epithelium (Fig.7). According to (Islam et al., 2006), the lamina propria in broiler and chickens from Bangladesh was composed of connective tissue and a simple tubular gland. Muscular fibers are distinct types of smooth muscle, the proventriculus glands were found in the tunica submucosa, where the proventriculus wall was the thickest (Fig.7). This result contradicts the findings of (Bradly and Grahom, 1960), who discovered that chicken proventriculus submucosa lacked proventriculus glands. The current study discovered that at 7 days post-hatch, the muscular layer of hicks was made up of two layers of smooth muscle fibers, one inner circularly and one outer longitudinaly (Fig.8). collagen fibers beneath mucosa, septum of gland, tunca muscularis and serosa (Fig.8). Previous research has showed that the number and arrangement of smooth muscle fibers in the tunica muscularis layer differentiated between birds (Hodges, 1974). This layer in quail was composed of 2 layers: internal circular and external longitudinal muscle fibers, according to (Attia,2008). (Abomandour,2014) developed a new arrangement in falcons composed of inner longitudinal and outer circular layers (Denbow, 2000), in parrots, just one layer of smooth muscle fibers was discovered to be circularly arranged. 14 days post-hatching, there are four tunica, Mucosal fold lining epithelia simple columnar epithelium, tunica submucosa was composed of connective tissue and contained proventricular glands lined by simple cuboidal epithelium, Tunica submucosa was made up of connective tissue and had proventricular glands lined by simple cuboidal epithelium (Fig.9). The tunica muscularis had been consists of 3 layers of smooth muscle fibers (inner circular, middle longitudinal, and outer circular) and connective tissue between the layers. (Fig.10). Mesothelium covered the entire tunica serosa (Fig.10). The same findings were made by (Banks, 993).

#### **Histometric Measurements of Provetriculus**

Tables (2) and (3) show comparative measurements of chick height epithelium, mucosa thickness, submucosa thickness, and muscularis thickness in prehatching and post-hatching chicks, during the present study highly statistical significant was (a) and statistical significant was (b) noted in table (2,3) histological parameters of proventriculus of broiler chicks from 11 days and 15 days pre-hatching and 7 days and 14 days post-hatching respectively. The height of epithelium were  $(9.74 \pm 1.68) \mu m, (13. 76 \pm 1.28) \mu m,$  were  $(37.14 \pm 2.11)$  $\mu$ m,(40. 76± 1.51)  $\mu$ m, the thickness of mucosa,(262.98 ± 2.42)  $\mu$ m, (351.08 ± 3.98)  $\mu$ m, (647. 14 ±2.71)  $\mu$ m, (752.42 ± 2.13)  $\mu$ m, thickness of sub mucosa were  $(1112.66 \pm 1.71) \mu m, (1129.44 \pm 2.270) \mu m, (2213.51 \pm 2.42) \mu m, (4117.34 \pm 2.89)$  $\mu$ m, the thickness of muscularis were,(149.81 ±1.33)  $\mu$ m,(217.42 ±3.43)  $\mu$ m  $(353.68 \pm 2.83) \mu m, (369.44 \pm 1.54) \mu m$ , the thickness of serosa were  $(11.24 \pm 0.79)$  $\mu$ m,(12.61 ±1.59)  $\mu$ m,(15.24 ±1.37)  $\mu$ m,(20.6 ± 0.89)  $\mu$ m, table(2,3). as according to (AL-Saffar and AL-Samawy, 2014), the mean thicknesses of the mucosa, submucosa, muscularis, and serosa in the striated scope owl were 3240 µm, 14280 µm, 960 m, and 120 µm, respectively.

| period                |              |                            |               |  |  |  |
|-----------------------|--------------|----------------------------|---------------|--|--|--|
| Age                   | Weight       | Length                     | Width         |  |  |  |
|                       | gm           | mm                         | mm            |  |  |  |
| 11<br>Pre-hatch<br>15 | 0.017 ± 0.3b | 5.4 ± 0.651 <mark>b</mark> | 2.44 ± 0.089b |  |  |  |

7.1 ± 1.244 a

14.8 ± 1.095b

17.8 ±1.788a

| Table (1) :This table shows the (weight gram , length mm and width mm)   | of the |
|--|--------|
| proventriculus in broiler chicks (Ross- 308) from pre-hatching post-hate | ching  |
| period   |        |

1.673 ± 0.348 a • a= highly statistical significant difference difference .

• b= statistical significant difference difference .

0.58 ± 0.016 a

0.997 ± 0.221b

p =≤ 0.05 ٠

Pre-hatch

7 Post hatch

14 Post-hatch

Table (2): This table shows the high of epithelium, thickness of (mucosa, submucosa, muscularis, serosa) of the proventriculus in broiler chicks(Ross- 308) at pre-hatching period

| Age       | High of<br>epithelium M<br>± SE | Thickness of<br>mucosa<br>M ± SE | Thickness of<br>sub-mucosa<br>M ± SE | Thickness of<br>muscularis<br>M ± SE | Thickness<br>of serosa<br>M ± SE |
|-----------|---------------------------------|----------------------------------|--------------------------------------|--------------------------------------|----------------------------------|
|           | μm                              | μm                               | μm                                   | μm                                   | μm                               |
| 11        | 9.74 ± 1.68 <sup>b</sup>        | 262.98 ± 2.42 <sup>b</sup>       | 1112.66±1.71 <sup>b</sup>            | 149.81±1.33 <sup>b</sup>             | 11.24±1.79 <sup>b</sup>          |
| Pre hatch |                                 |                                  |                                      |                                      |                                  |
| 15        | 13.76± 2.08 <sup>b</sup>        | 351.08± 3.98ª                    | 1129.44±2.270ª                       | 217.42±3.43ª                         | 12.61±3.59ª                      |
| Pre hatch |                                 |                                  |                                      |                                      |                                  |
|           |                                 |                                  |                                      |                                      |                                  |

• a= highly statistical significant difference difference .

• b= statistical significant difference difference .

p =≤ 0.05

 $3.9 \pm 0.651a$ 

9 ± 1.414b

10.6 ± 0894a

| post-hatching period |                                       |  |  |  |  |  |
|----------------------|---------------------------------------|--|--|--|--|--|
| Age                  | High of<br>epithelium M<br>± SE<br>µm | Thickness of<br>mucosa<br>M ± SE<br>µm | Thickness of<br>sub-mucosa<br>M ± SE<br>μm | Thickness of<br>muscularis<br>M ± SE<br>µm | Thickness of<br>serosa<br>M ± SE<br>µm |  |
| 7<br>Post hatch      | 37.14±2.11 <sup>b</sup>               | 647.14±1.71 b                          | 2213.51±2.42 <sup>b</sup>                  | 353.68±1.83 <sup>b</sup>                   | 15.24±1.37 <sup>b</sup>                |  |
| 14<br>Post hatch     | 40.76± 2.51ª                          | 752.42±2.13ª                           | 4117.34±3.89ª                              | 369.44±2.54ª                               | 20.6±0.89ª                             |  |

Table (3) : This table shows the high of epithelium, thickness of (mucosa, submucosa, muscularis, serosa) of the proventriculus in broiler chicks (Ross- 308) at

- a= highly statistical significant difference difference .
- b= statistical significant difference difference .
- p =≤ 0.05



Fig.(1): Cross histological section of broiler chicks proventriculus at (11 day)prehatch shows: lining epithelia (E), sub-mucosa (Sm), proventriculus gland (Pg), tunica musclaris (red arrow), tunica (yellow serosa arrow). H&E stain. X100.



Fig.(2): A: Cross histological section of broiler chicks proventriculus at (11 day) pre-hatch shows: sub-mucosa (Sm), proventriculus gland (G), muscularis (M), serosa (red arrow). H&E stain. X200. B: Magnified section. H&E stain. 400x.



Fig.(3): A: Cross histological section of broiler chicks proventriculus at (15day) pre-hatch shows: connective tissue (black arrow), lining epithelia mucosa (E), proventriculus gland (Pg) masson trichrom stain. 20X.



Fig.(4): A: Cross histological section of broiler chicks proventriculus at (15day) pre-hatch shows: mucosa (M), sub-mucosa (Sm), proventriculus gland (G), muscularis mucosa (Ms), serosa (yellow arrow), nerve supply (N), mucosal gland (yellow star).H&Estain.40x. B and C: magnified section. H&E stain.400x.



Fig.(5): A: Cross histological section of broiler chicks proventriculus at (15day) pre-hatch shows: muscularis inner layer (yellow arrow), muscularis outer layer (red arrow), blood supply (black arrow), nerve supply (blue arrow). H&Estain.200x. B: Magnified section. H&E stain.400x.



Fig.(6): A: Cross histological section of broiler chicks proventriculus at (15day) pre-hatch shows: connective tissue (black arrow), mucosa (blue arrow), muscularis (red arrow), muscularis inner layer (a), muscularis outer layer
(b),serosa (yellow arrow). masson trichrom stain. X40. B&C: Magnified section. masson trichrom stain. X400.



Fig.(7): Cross histological section of broiler chicks proventriculus at (7day) Posthatch shows: mucosa (A), mucosal fold (yellow arrow), sub-mucosa (B), muscularis (C), serosa (D). H&E stain. X40



Fig.(8): Cross histological section of broiler chicks proventriculus at (7 day) Posthatch shows: mucosa (A), sub-mucosa (B), proventriculus gland (G), muscularis (C), collagen bundles beneath(mucosa, septum of gland, muscularis) (black arrow). Masson trichrom stain. X100.



Fig.(9):A: Cross histological section of broiler chicks proventriculus at (14 day) Post-hatch shows: mucosa (A), sub-mucosa (B), muscularis (C), serosa (D). H&E stain x40. B: Mucosal fold (E), mucosal lining epithelia (black arrow). H&E stain. X400. C: Proventriculus gland lining epithelia (yellow arrow). H&E stain. X400.



Fig.(10):A: Cross histological section of broiler chicks proventriculus at (14 day)
Post-hatch shows: connective tissue in tunica mucosa (black arrow), connective tissue in tunica sub-mucosa (yellow arrow). Masson trichrom stain. X40. B:
Magnified section show: connective tissue between the layer of tunica muscularis.

H&E stain. X400.

### Conclusion

broiler The study describes the developing proventriculus in chicks in great detail, the proventriculus displayed distinct morphological and histological characteristics throughout its development. More molecular research is required to complete picture of this avian species' proventriculus development. The the lengths, widths, average weights, and other histological of proventriculus gradually increased. structures the were Based on findings the and discussion presented above. this study provides a clear picture 308 broiler of the growth rate of Ross chicks, particularly those in the proventriculus, which are also related to feeding digestion in birds.

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