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Histological structures of liver and Bursa of Fabricius in two-line selection female and male Japanese quail *Coturnix japonica*

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ABSTRACT

Japanese quail (*Coturnix japonica*) is a common avian species used as a model for studying reproductive and physiological functions. Liver and Bursa of Fabricius are two important organs involved in various metabolic and immunological processes. Previous research has shown that these organs exhibit sexual dimorphism and may be affected by genetic selection for certain traits in quail. This study aims to investigate the histological structures of liver and Bursa of Fabricius in two-line selection female and male Japanese quail. Liver and bursa were collected from both lines at 35 days of age. The hypatocytes in both strains is same in ranged and it sphirical in female but polyhidral in male , the sinosoids seen narrow in brown female and male compared with black, the artary in black strain seen more wide than in brown, the portal vein also had a large lumen in black than brown. The two strain have same characters of tissues in bursa of Fabricius but the cortex of black is larger than brown (female and male)and more eosinophilic in addition to present of adipose cells distributed in the bursa of Fabricius of black strain more than in the brown.

التركيب النسجي لكبد وجراب فابريشيوس في سلالتين مختارتين من اناث وذكور السمان الياباني

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الخلاصة

يعتبر السمان الياباني نوع شائع من الطيور المستخدم كنموذج لدراسة الوظائف التكاثرية والفسيولوجية.وان الكبد وجراب فابريسيوس هما عضوان مهمان يشاركان في مختلف العمليات الايضية والمناعية. أظهرت الأبحاث السابقة ان لهذه الأعضاء ازدواج الشكل الجنسي وقد تتأثر بالنتقاء الجيني لسمات معينة في السمان. تهدف هذه الدراسة الى التحقق في التراكيب النسجية للكبد وجراب فابريشا لسلالتين من السمان الياباني الاناث والذكور، وتم جمع العينات من كلا السلالتين بعمر 35 يوماً. الخلايا الكبدية في كلتا السلالتين هي نفسها من حيث الترتيب وهي كروية في الاناث ولكنها متعررة السطوح في الذكور,الجيبانيات الدموية تكون ضيقة في الاناث والذكور البنية مقارنة بالسلالة السوداء, والشريان الكبدي في السلالة السوداء يظهر أوسع من البنية, وكان للوريد البابي تجويف اكبر بالسلالة السوداء بالنسبة للبنية. السلالتان لهما نفس الانسجة في جراب فايبريسيوس ولكن القشرة بالسوداء اكبر من البنية (انثى وذكر) وخلايا حمضة اكثر بالإضافة الى وجود الخلايا الدهنية الموزعة في جراب فابريسيوس في السلالة السوداء اكثر من البنية.

الكلمات المفتاحية : جراب, انثى, ذكر, سمان ياباني, كبد

INTROUCTION

Japanese quail (*Coturnix japonica*) is a preferred species for studies due to its short generation time and different genetic strains (Jacobsen *et al.,* 2017). The limited spatial needs and husbandry costs make the bird a convenient another model species (Jaspers, 2015). Onagbesan *et al.,* (2009) showed that quails reach sexual maturity at six to seven weeks of age. Duymus *et al.,* (2013) noted that quail plumage colours are grey, brown, white, golden, and black, and the plumage is one of the important traits of thquail (Nadeau, 2006). On other hand, Bed'hom *et al.,* (2012) found that the colour of plumage has a relationship with some productive traits, including body weight. The liver and Bursa of Fabricius are two important organs in poultry involved in various metabolic and immunological processes (Sorour *et al.,* 2021).

The liver is responsible for detoxification, metabolism of nutrients, and synthesis of proteins and hormones, while the Bursa of Fabricius plays a key role in the development and maturation of B lymphocytes and the production of antibodies (Zaefarian *et al.*, 2019; Behboodi *et al.*, 2021). In poultry species, liver is larger than in animals when compared to body size. The size and colour of the liver depend on age and body weight (Lumeij, 1994). According to Pu *et al.*, (2020) the liver plays a central role in metabolism, glycogen storage and the decomposition of red blood cells.

Bursa of Fabricius a lymphoid organ of immunological significance that is specific to avian species. It is a transient lymphoepithelial organ of birds that develops fully at hatching and totally involutes after reaching sexual maturity in the majority of species (Onyeanusi *et al.*, 1993).The microenvironment for B-lymphocyte development, maturation, differentiation, and antibody diversification is provided by Bursa of Fabricius ,which supports humoral immune response in avian species (Arakawa *et al.*, 2002). In addition, Delmore *et al.* (2016) demonstrated that there is a high probability of linking the different colors of plumage to some quantitative traits that need to be studied.

This study suggested there is some different information on the gross anatomical changes in the liver and Bursa of Fabricius in two strains of female and male Japanese quail (*Coturnix japonica*) (Black and Brown), and also paves the way to understand the differences in the gross anatomy of the liver and bursa in female and male Japanese quail (Coturnix japonica).

MATERIAL AND METHODS

Birds Managements

The ethical committee of animal research of the department of animal production permitted all experimental protocols (Tikrit University). Female and male Japanese quails were hatched and reared in a breeding colony in the poultry housing at poultry housing in college of Agriculture. A total of 20 females, 10 males (black and brown plumage) were compared and placed at five weeks of age in a breeding facility with pens measuring (40 x 40 cm) . Birds were fed ad libitum a profitablquail food (NRC,1994), starter diet (24% CP and 2976 kcal ME/kg), and production diet (20% CP and 2850 kcal ME/kg) during the experment periods. Thquails were housed in cages in a well-ordered environment.

Histological samples

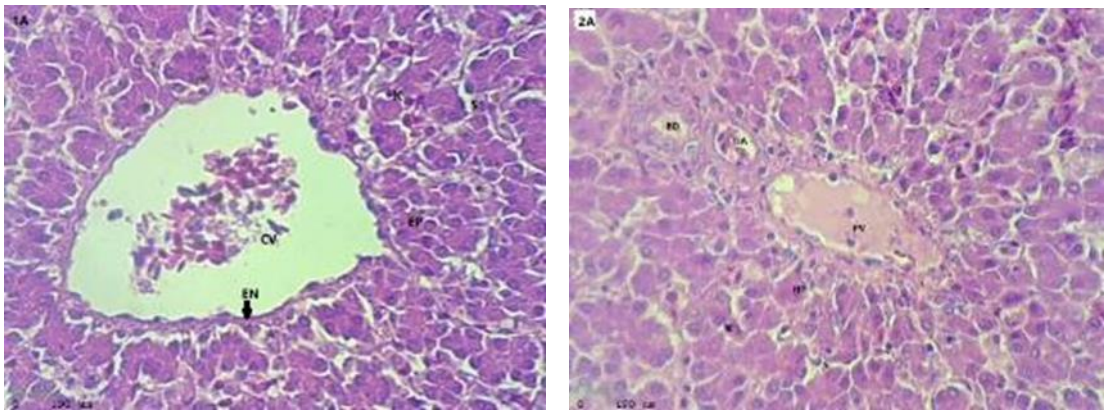
The liver and bursa are harvested from the brown and black female and male Japanese quail by dissections of the abdominal and collected the liver, while the bursa of Fabricius is removed by the cervical area dislocated and both organs fixed for routine paraffin method. The organs were fixed in formalin 10 percent prepared by taking 10 ml formaldehyde and diluting it with 90ml tap water, left the organs in it for 24 hours and then washed in tap water and distal water each one for 30 min, removed the water from the tissues by dehydration with ethanol alcohol in ascending concentration 30, 50, 70, 80, 90, 95, 100 and100 percent each concentration for 30 min, clearing it by xylol for 30 min twice times and embedding it in paraffin for twice times and molding in stainless steel casting molds, leave it to hardens then make a sectioning Rotary Microtome Cut4060 - Germany) in 5-7 µ thickness leave at the hot plate at 40C^{o} to dry and dying with ematoxylinin and eosin (Bancroft and Gamble,2008). Amounted by DPX and examined by light microscope (OBL-137C832DIGITAL MICROSCOPE SET 4x-100x with 3W LED (transmitted light), with Tablet camera 5mp, WLAN, USB 2.0, HDMI, SD,CMOS 1/2,5" inclusive of C-MOUNT ADAPTER).

RESULTS AND DISSCUSION

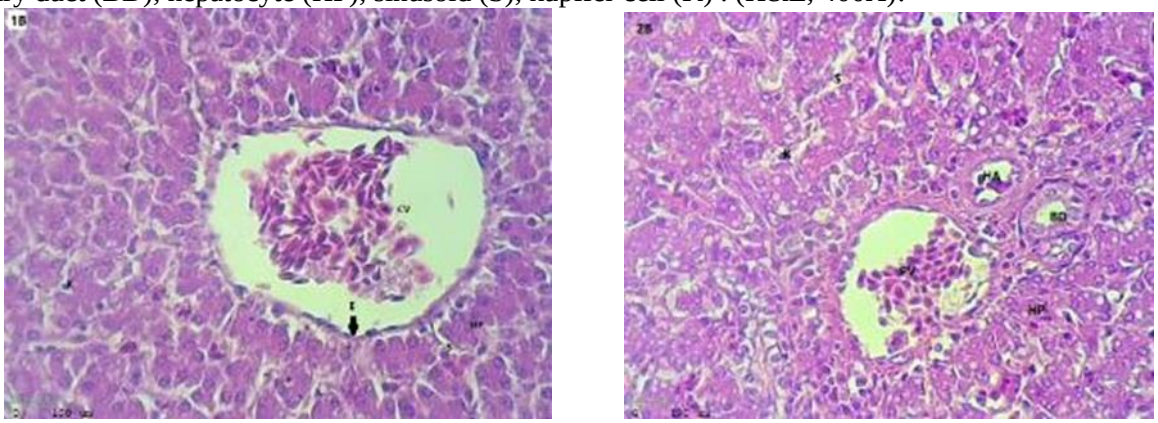
Liver and Bursa of Fabricius Histological structures

Liver tissues of brown and black femal and male Japanese quail *Coturnix japonica*

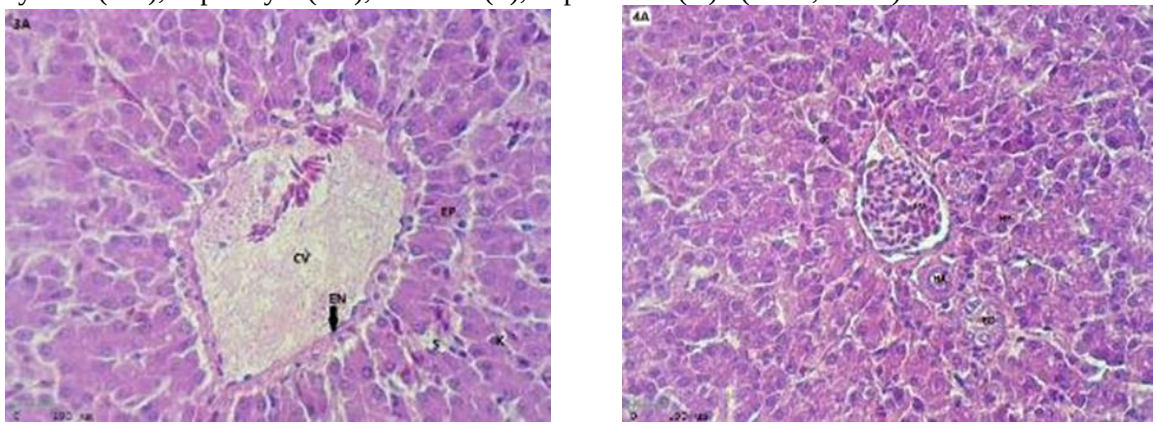
The hepatocyte in both strains brown and black female and male called hepatic parenchyma organized as irregular cords of two or more cells around the central vein, the shape of hepatocyte is same, it oval or spherical in the female but it polyhedral in male in both brown and black . the sinusoid in the female and male present between the hepatocytes and it seen narrower in brown compared with black this results agree with El-Zoghby,(2005) in quail ,Khaleel et al.(2017) in Mallard duck , it lining with flattened endothelial cells and number of kupffer cells appear in the sinusoid, the portal vein presented with hepatic artery and bile duct as a portal triad , the bile duct lining by simple cuboidal cells, the hepatic artery have thick wall, small and narrow lumen lining by endothelial cells the bile duct and hepatic artery in black seen more wide than in brown, the portal vein in also had a large lumen in black than brown and lining by endothelial cells. (Fig A).that resembled with the result of Hussein and Hussein (2016) when descripted the lumen of sinusoid moorhen, domestic fowl, and many other domestic birds and vertebrates (Elizabeth and Frye, 2001) and (Al-Abdulla,2015).



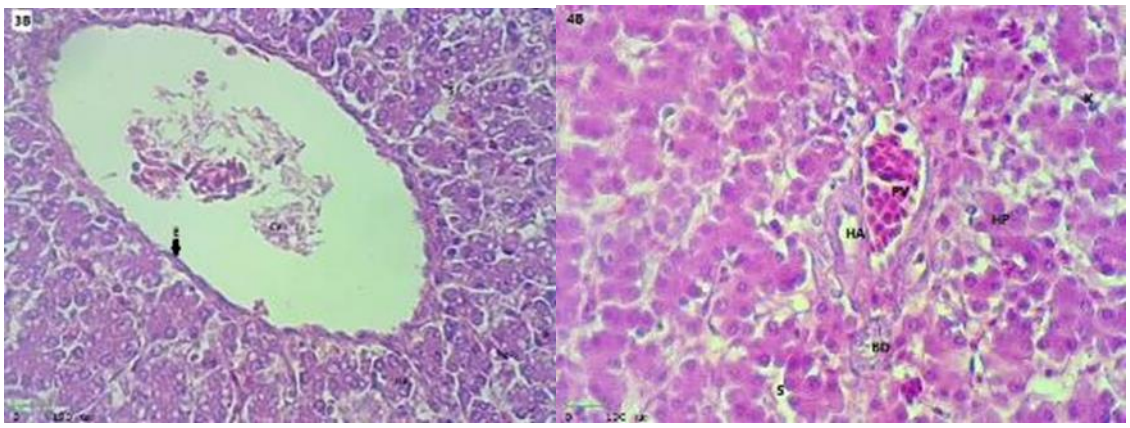
Figure(A): the liver of Brown female *Coturnix japonica* showed (1A) central vein with blood cells (CV), endothelial (E), hepatocytes (H), sinusoid(S), kupffer cell(K), (2A) portal vein (PV), hepatic artery (HA), biliary duct (BD), hepatocyte (HP), sinusoid (S), kupffer cell (K) . (H&E, 400X).



Figure(B): the liver Black female *Coturnix japonica* showed (1B) central vein with blood cells (CV), endothelial (E), hepatocytes (H), sinusoid(S), kupffer cell(K), (2B) portal vein (PV), hepatic artery (HA), biliary duct (BD), hepatocyte (HP), sinusoid (S), kupffer cell (K) . (H&E, 400X).



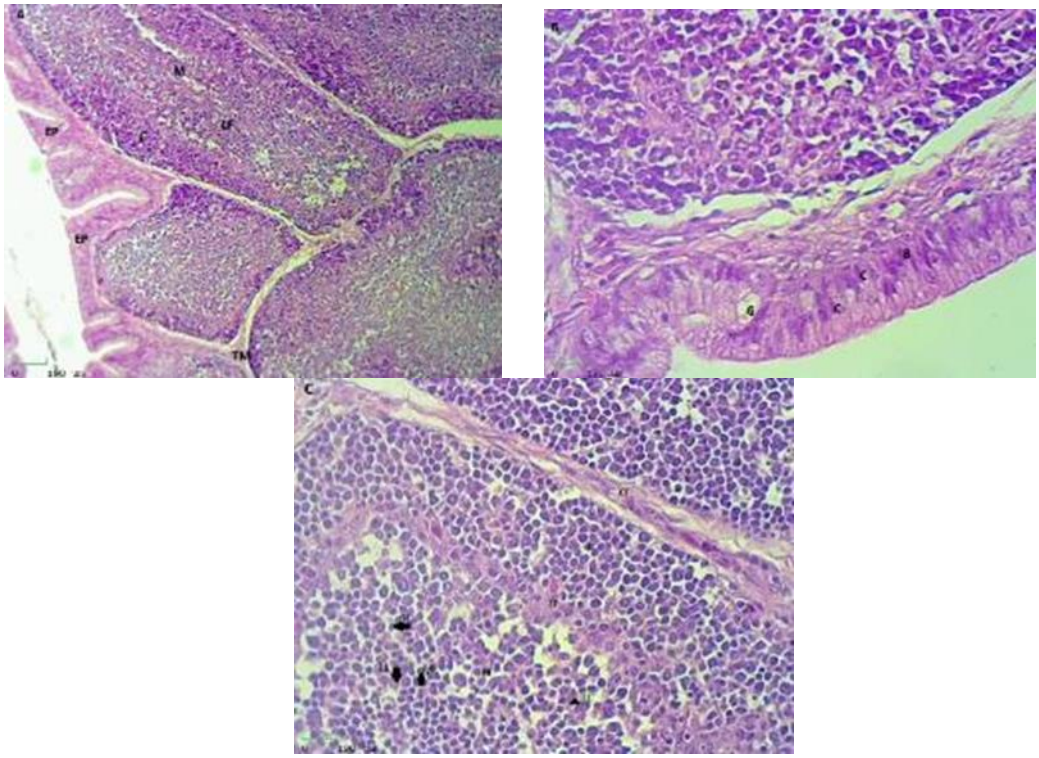
Figure(A): the liver of Brown male Coturnix japonica showed (3A) central vein with blood cells (CV), endothelial (E), hepatocytes (H), sinusoid(S), kupffer cell(K), (4A) portal vein (PV), hepatic artery (HA), biliary duct (BD), hepatocyte (HP), sinusoid (S), kupffer cell (K) . (H&E, 400X).



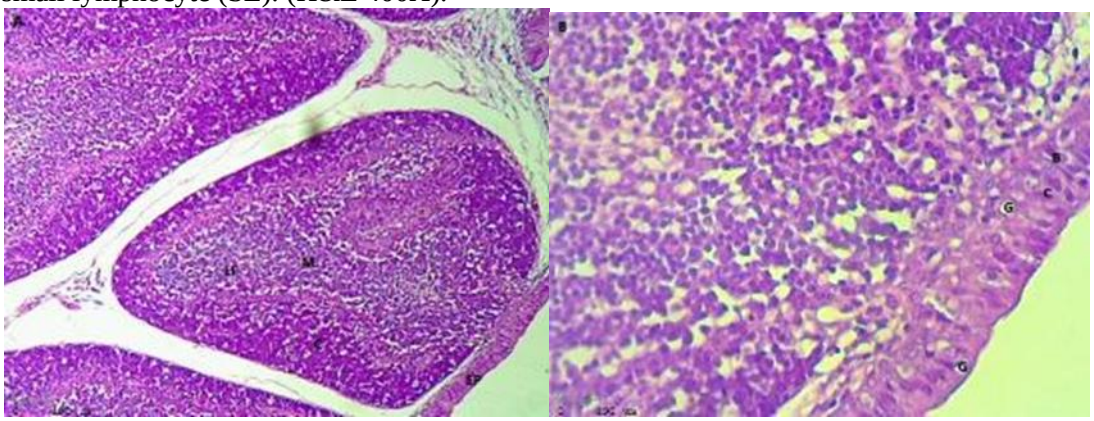
Figure(B): the liver Black male Coturnix japonica showed (3B) central vein with blood cells (CV), endothelial (E), hepatocytes (H), sinusoid(S), kupffer cell(K), (4B) portal vein (PV), hepatic artery (HA), biliary duct (BD), hepatocyte (HP), sinusoid (S), kupffer cell (K) . (H&E, 400X).

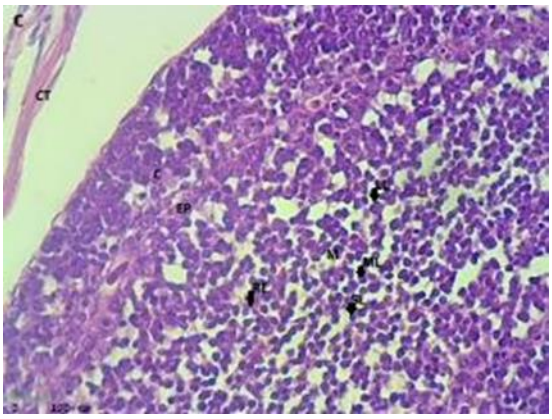
Bursa of Fabricius tissues of brown and black female and male Japanese quail *Coturnix japonica*

The bursa of fabricia contain of lymphoepithelial follicles, these follicles separated by interstitial dense connective tissue and lining by epithelial tissue that contain three types of cells 1-columnar cells,2- basal cells 3- goblet cells among the columnar cells, the follicles contain of two region cortex near the connective tissue and medulla in the center there is epithelial cells and blood separated them these result agreement with (Hossaint et al.,1986) and similar finding with Mazzone et al.(2003)in leghorn hens, in the medulla there are various sizes of lymphocytes can recognized (large, middle and small lymphocytes) similar result were observed by King (1977) in duck,and Kumar et al.(2014) khaki campbell duck. also there are riticoloepithilial cells in the medulla. the two strains have this contain as a same characters but the cortex in the black female is larger than brown female and more eosinophillic and there are adipose cells distributed in the bursa of fabricia (cortex and medulla) of the black female more than in brown female.it agree with (Jeyachandra et al.,2017) and (Deka et al.,2020) in Pati duck. The same characters is found in the Bursa of Fabricius tissues of brown and black male Japanese quail Coturnix japonica as showed in the figures below .

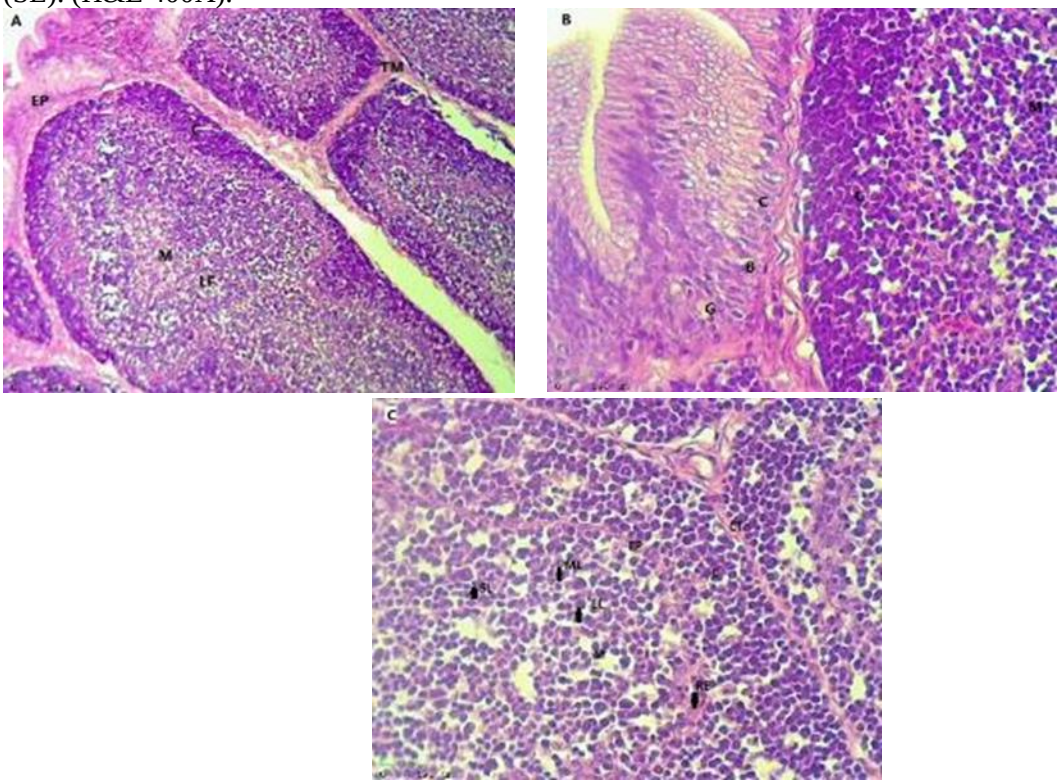


Figure(A,B,C): bursa of fabricias tissues in brown female Coturnix japonica showed (A) panoramic view, Tunica muscularis (TM), Cortex (C), Medulla(M), Epithelium (EP), Lymphoid follicle(LF). (100X H&E). (B) three types of cells in the epithelium; columnar cells (C), basal cell(B), goblet cell(G). (400X H&E). (C) the interstitial dense connective tissues that separated the follicles from each other (CT), cortex (C), medulla (M), epithelial cells(EP), reticloepithelial cells (RE), large lymphocyte (LL), middle lymphocyte (ML), small lymphocyte (SL). (H&E 400X).

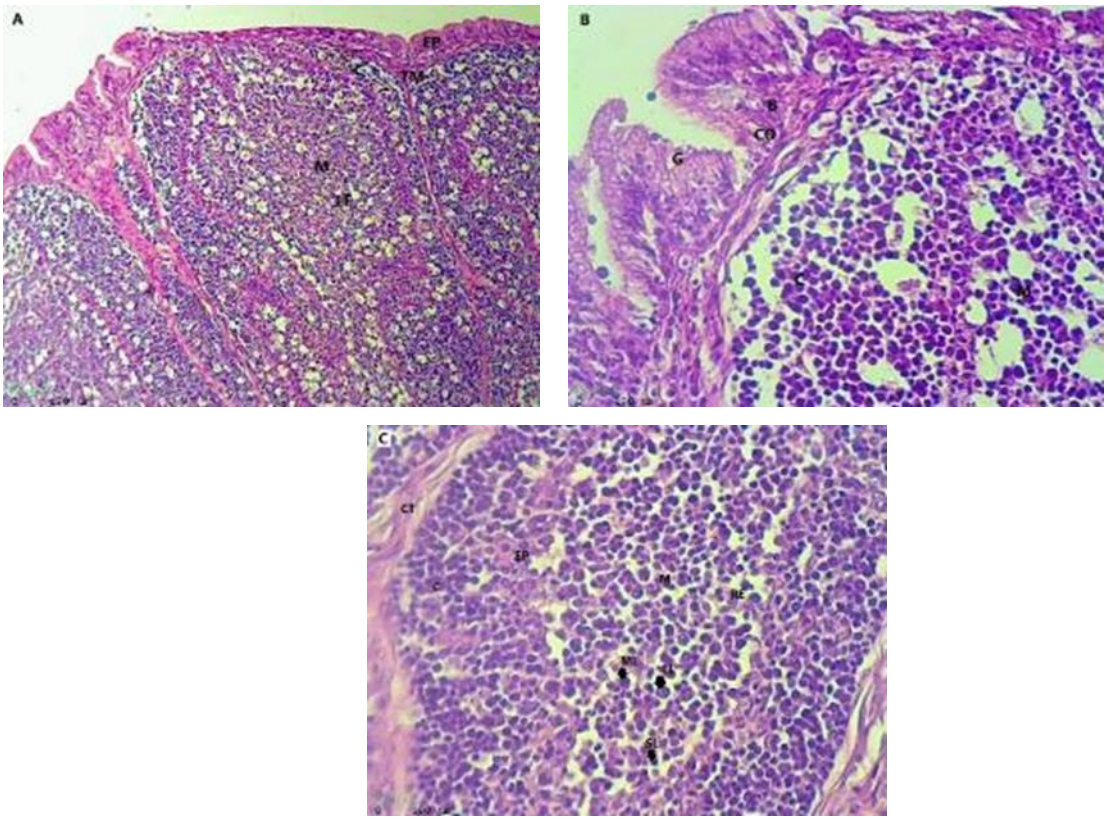




Figure(A,B,C): bursa of fabricia tissues in black female Coturnix japonica showed (A) panoramic view, Cortex (C), Medulla(M), Epithelium (EP), Lymphoid follicle(LF). (100X H&E). (B) Three types of cells in the epithelium; columnar cells (C), basal cell(B), goblet cell(G). (400X H&E). (C) the interstitial dense connective tissues that separated the follicles from each other (CT), cortex (C), medulla (M), epithelial cells(EP), reticloepithelial cells (RE), large lymphocyte (LL), middle lymphocyte (ML), small lymphocyte (SL). (H&E 400X).



Figure(1-A,B,C): bursa of fabricias tissues in brown male Coturnix japonica showed (A) panoramic view, Tunica muscularis (TM), Cortex (C), Medulla(M), Epithelium (EP), Lymphoid follicle(LF). (100X H&E). (B) three types of cells in the epithelium; columnar cells (C), basal cell(B), goblet cell(G). (400X H&E). (C) the interstitial dense connective tissues that separated the follicles from each other (CT), cortex (C), medulla (M), epithelial cells(EP), reticloepithelial cells (RE), large lymphocyte (LL), middle lymphocyte (ML), small lymphocyte (SL). (H&E 400X).



Figure(2-A,B,C): bursa of fabricias tissues in Black male Coturnix japonica showed (A) panoramic view, Tunica muscularis (TM), Cortex (C), Medulla(M), Epithelium (EP), Lymphoid follicle(LF). (100X H&E). (B) three types of cells in the epithelium; columnar cells (C), basal cell(B), goblet cell(G). (400X H&E). (C) the interstitial dense connective tissues that separated the follicles from each other (CT), cortex (C), medulla (M), epithelial cells(EP), reticloepithelial cells (RE), large lymphocyte (LL), middle lymphocyte (ML), small lymphocyte (SL). (H&E 400X).

CONCLUSION

It can be concluded from the current study there was a significant difference between the two strains in body weight and liver weight. The data did not show any significant difference between the two strains in the bursa of Fabricius weight. The normal histological study of the liver and bursa of Fabricius showed that there are differences in tissue composition between the two strains, and this could give researchers additional information about these two organs and their role in body functions.

CONFLICT OF INTEREST

The authors declare no conflicts of interest associated with this manuscript.

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