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Morphological and Histopathological Study of the Liver of Three Species of Syrian Birds (*Tyto Alba*, *Turdus Merula*, and *Carduelis Spinus*)

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Abstract— The current research was conducted to study histopathologically and morphologically the liver of three species of Syrian birds (*Tyto alba*, *Turdus merula*, and *Carduelis spinus*).
The results showed that the liver occupies the upper right part under the heart from the thoracic abdominal cavity. Its color ranges from pinkish red to dark red. Its weights ranged between 0419-4.453 g depending on the type studied. A thin capsule surrounds the liver cosisting of fibrous connective tissue , divides the liver into two main lobes (right and left) that differ in size and shape of the edges and the presence of the secondary lobes.
The histological study of the liver showed that the barriers between the lobules are not clear in the studied species, but the beginning of these septa was clear to the blackbird. In both owls and Eurasian siskin birds, the liver tissue is chordate, while this tissue is reticulated in the blackbird, with an observation of the beginning of the chordal alignment of multifaceted cells, with spherical near-central nuclei containing one or two nuclei. The blood sinuses are abundant. The adipocytes were observed around the sinuses, as well as, the Kupffer cells were found within the lumen of the endothelial boundaries. The portal space (hepatic triad) appeared clearly within the liver tissue of the owls, but the matter differs in both Eurasian siskin and the blackbird as they have a semi-portal distance (which means divergence among each artery, vein, and bile duct).

Keywords— histological study, morphological study, liver, Tyto alba, Turdus merula, and Carduelis spinus.

I. INTRODUCTION

The liver is larg gland, two-lobed, located at the front end of the body cavity. Its size is proportional to the circumference of the inner surfaces of the body wall and the adjacent and closed organs (such as the heart, pericardial cavity, spleen, gallbladder, intestinal loop and lungs). This organ holds in its place numerous ligamentous attachments and its surface is covered by the visceral peritoneum.

The liver tissue differs between organisms, but there are common characteristics. surrounding the liver a capsule varying thickness, which varies according to the species (glissonscapsule). The glissonscapsule continues into the liver and divides it into hexagonal functional units of connective tissue (hepatic lobes). Each lobe divides into approximately 100,000 hepatic lobules. Each lobule consists of a hepatocyte which occupies around 80% of the total volume of the liver and performs many of its functions. There are also two types of cells (the endothelial cells that line the blood sinuses and macrophages). Macrophages are referred to as Kupffer cells.

The lobes differ in number and size depending on the species. The right lobe is larger in domestic birds and most species, its size and color vary according to the food type [1], [2], [3].

Located between the hepatic lobules there is the portal triad, which is composed of the portal vein, the hepatic artery, and the bile duct, in addition to the presence of the lymph vessel and the nerves. These vessels are surrounded by connective tissue.

In vertebrates, the liver consists of hepatic cells, bile ducts and blood follicles. The structural differences among the species appear in the parenchyma, as there are three types of liver organization. In the first type, the hepatic cells are arranged radially around a central vein in the form of lamellae of two cell thicknesses with blood genomes separating the hepatic laminae. The bile ducts are located between the adjacent liver cells. This type is described in birds and mammals.

In the second type, the blood genomes form a network that surrounds the hepatic cord, while the bile ducts form the center of the hepatic cord around the central vein. This type was described in hagfish *Myxine glutinosa*. In the third type, the hepatic cells are arranged as laminae. This type was described in rainbow trout Salmo Gairdner [1], [4], [5].

There is a similarity between the livers of birds and mammals in terms of histological features and functions. The septum between the lobules does not appear in the birds' livers, but it can be observed in mammal's livers. However, each unit is accepted as having a central vein in the form of a lobule in the birds livers. Birds' livers have less connective tissue than mammalian livers and lack true structure. In bird species, the liver is large compared to the size of the bird ,unlike mammals, but it is functionally similar to mammals in that it is a biochemical factory responsible for metabolism, detoxification, and regulation of the production, storage and release of fats, carbohydrates and proteins. The liver produces a variety of proteins, such as blood proteins, enzymes, hormones, clotting factors, and immune factors. It acts as an endocrine gland as it is stores vitamins (A, D, K, and E) and B12 in addition to its role in the phagocytosis process due to the presence of Kupffer cells [6], [7], [8], [9].

II. Related Work

This is the first a histopathological study of the liver has been conducted in Syrian birds (owls, blackbirds, and Eurasian siskin).

This study constitutes a new addition to biological studies that contribute to increasing information about bird biodiversity in Syria.

The research was conducted to study the liver of the three species (owls, blackbird, and Eurasian siskin) anatomically and morphologically, and to Histological studying of the liver of these species using hematoxylin-eosin.

III. Methodology

Materials

The research included the following materials:

a. Glass slides from the Chinese company E.S.L.C.

b. Colorants with conventional Eosin-Haematoxylin Colorants.

c. Alcohol in different concentrations (absolute alcohol, 90%, 75%, 70%).

d. Xylol.

e. canada balsam.

f. Thin glass screens type. Citoglas.

g. Slicing tray

In addition to the following devices: microtome for thin-thickness tissue slicing (4-5 micron) (Meditome), oven (BINDER), optical microscope (Nikon Eclipse Ni) equipped with a digital camera, and preparing paraffin molds device (MEDITE).

Samples collection and screening:

The current study has been completed in the Postgraduate Laboratory in the Department of Biology / Faculty of Science / Tishreen University during 2019-2020. For each species studies, five individuals were collected from the Latakia city between. Samples were obtained alive and transferred to the laboratory.

They were anesthetized using Chloroform, and dissected. After dissection, the liver was isolated and kept in a 10% formalin solution for a few days before using them and preparing for histological study. Samples were prepared for histological sections at the histopathology department - Tishreen Hospital.

The method of re.[10] was followed. Liver samples were passed in the following solutions: Formalin 10% for an hour, different concentrations of Ethanol (70%, 75%, 90%), respectively, for an hour and absolute alcohol 100% twice for 2 hours in each time for samples dewatering. Then liver samples were passed in xylene thrice for 1 hour in each time, paraffin twice for 2 hours in each time.

Samples were embedded with paraffin wax after being placed in suitable blocks. The tissue sections were prepared with a thickness of 4-5 microns using the automatic microtome. These sections were placed in a 45-40 ° C water bath. Then the sections impregnated with paraffin were placed on glass slides, and placed at 80 ° C for 12 minutes to dissolve the wax from the preparations. The sections were immerged in the following solutions: xylene (three time), absolute alcohol (twice), commercial alcohol at the following concentrations 90%, 75% and 70%, respectively, to expel the xylene from the tissues (5 minutes for each time). The slides were washed with water and placed in Haematoxylin for a few minutes.

The slides were re-washed with water to remove the extra hematoxylin. Then they were soaked in Eosin for a minute and re-washed with water. The slides were immerged in the following concentrations of alcohol 70%, 75%, 90%, respectively, for 5 minutes in each time, and then placed in two basins of absolute alcohol for 5 minutes in each basin to push out water from the tissues. Slides were passed in two basins of xylene for 5 minutes in each basin to de-alcohol from the tissues. Canada balsam was applied and then the slides were covered with a coverslip. These sections were studied by light microscopy (Nikon Eclipse Ni).

Morphological characters of the species studied

The owl bird (*Tyto alba*)

It is called the warehouse owl or the white owl. Its length is 34 cm, the wing length is 89 cm. Its body is more straight than the rest of the species. The feathers are of varying color, the upper parts are light sandy color, and mostly white in color, and its wings appear silent, rounded, smooth, its face in the shape of a heart, and the legs and feet are covered White feathers to the claws emit a loud double shriek or squawk [11].

The blackbird (*Turdus merula*)

This species is a songbird, 25 cm long, dark black in color with a yellow beak. Female is dark brown in the top and its chest is pale gray. Female beak is dark in color. This bird

lives in bushes, gardens, orchards rich in short plants and often near agricultural areas, nesting in dense shrubs or trees. Most of these birds are cutters that spend the summer in our country and then migrate in the winter to neighboring countries. Its food is as varied as insects, worms and some types of fruits. Their eggs are green or greenish [11].

The Eurasian siskin (*Carduelis spinus)*
It is called Eurasian siskin. Its length is 12 cm. The body end is yellow. The chest is black in tail and its wings. Its back is greenish, black in the crown and the chin in males. The female is olive-brown on top and the bottom is white in color, it does not have a black color on the head like the male. It is a resident bird that spreads in Jordan, Iran, east and north and in central Arabia and is considered a stray bird in Syria. It lives in coniferous forests as it is seen on the coasts and next to mixed forests, feeding on thistle seeds and some trees such as pine, fir and mulberry [11].

IV. Results and Discussion

Morphological description of liver
The owl bird (*Tyto alba*)
The liver in the species *Tyto alba* occupies the upper right part of the lower of heart from the thoracic abdominal cavity. It is dark red in color, and it weighs about 4.263-4.453 g. It consists of two main lobes (right and left), the right lobe is slightly larger than the left, and both of them appear with complete edges (Figure 1).

The blackbird (*Turdus merula*)
It occupies the upper right under the heart of the thoracic abdominal cavity. It is dark red-brown in color. it weighs about 3.291 g. It is divided into two main lobes (right and left). The right lobe is larger than the left, and both of them appear with serrated edges. On the Dorsal side, it is notable a touch of atrophied lobes (Figure 2, 3).

The Eurasian siskin (*Carduelis spinus)*
It fills the upper part of the thoracic abdominal cavity below the heart. It is red, pinkish-brown in color. It weighs about 0.419-0.440 g. It is divided into two main lobes (right and left). The difference in size between them is observed as the right is larger with complete edges. As for the left, it has serrated edges. There are atrophic lobes on the dorsal side (two lobes), each of which is connected with one of the main lobes (Figure 5, 4).

Histological structure of liver
The owl bird (*Tyto alba*)
It is surrounded by a thin capsule. The inter-lobular septa is unclear. The chordate tissue consists of cells lined up in the form of multifaceted cords, with proxcentric globular nuclei containing one or two nuclei. The blood sinuses are abundant and the adipocytes are clearly observed around the blood sinuses. Inside the lumen of the blood sinuses there are kupffer cells in the boundaries of the endothelium cells, and the portal space appears within the hepatic tissue (Figure 6, 7, 8).

The blackbird (*Turdus merula*)
It is surrounded by a thin capsule. The beginning of the inter-lobular septa is noted, but it is not clear and continuous into the parenchyma. The tissue is as a network, where the cells locate randomly. The cells are multi-faceted with large globular nucleus near centrosporis, containing one or more nuclei. Fat cells and kupffer cells with spherical nuclei are clearly observed in separate places. The blood sinuses are small and abundant, the endothelial cells located inside the sinuses. The portal space also appeared within the liver tissue. There is a divergence between the artery, vein and the bile duct (semi-portal space) (Figures 9, 10, 11).

The Eurasian siskin (*Carduelis spinus)*
It is surrounded by a thin capsule, no inter-lobular septa. The eosinophilic coloration is severe. The tissue is chordate, hepatocytes line up in cords. The cells are multi-surface with near-central spherical large nucleus, containing one or more nuclei. Fat cells in addition to both globular nucleus kupffer cells and endothelial cells were observed inside the blood sinuses which were clearly small in size. The portal space also appeared clear as its components (the artery, vein, and bile duct) diverge (semi-portal space) (Figure 12, 13).

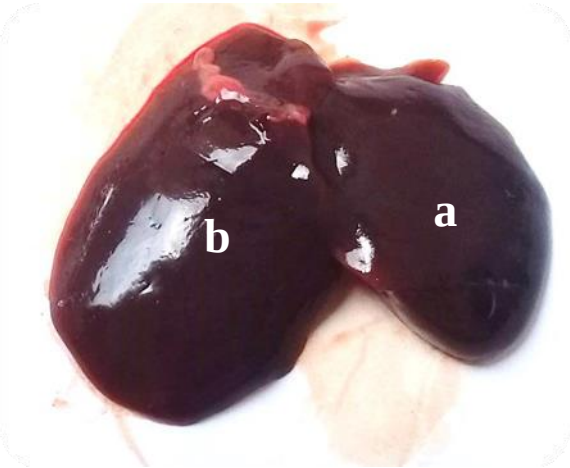

Figure (1): The liver of the owl bird (Tyto alba) a: (right lobe), b: (left lobe)

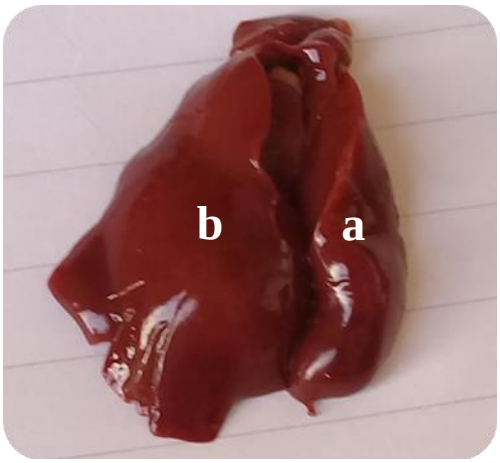

Figure (2): The liver of the blackbird (Turdus merula)

a: (right lobe), b: (left lobe)

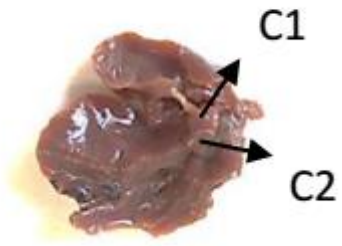

Figure (3): The atrophic lobes in the blackbird (Turdus merula), C_1 and C_2 : Connective lobes

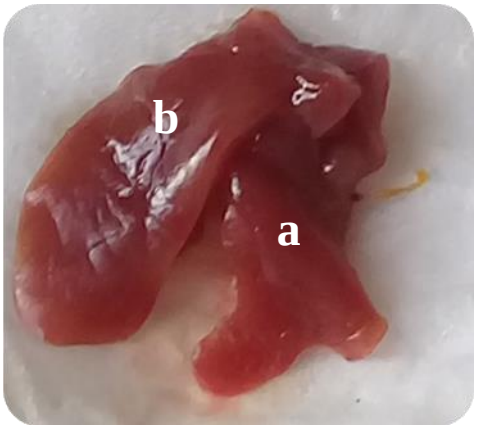

Figure (4): The liver of the Eurasian siskin (Carduelis spinus)

a: (right lobe), b: (left lobe)

Figure (5): The atrophic lobes of liver in the Eurasian siskin (Carduelis spinus)

, C_1 and C_2 : Connective lobes

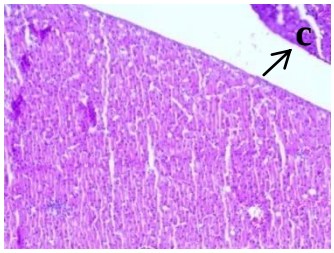

Figure (6): The capsules (C)and the cord line of the hepatic cells

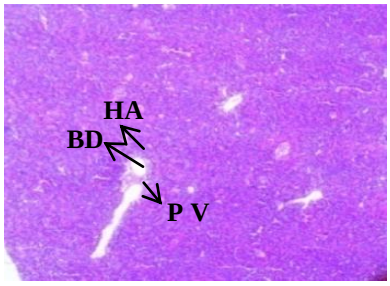

Figure (7): The porta; area (PV: portal hepatic vein, HA: hepatic artery, BD: bile duct).

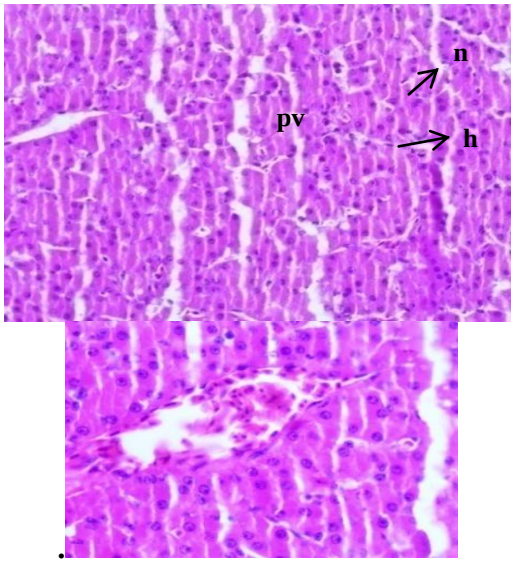

Figure (8): The radial located of the cells around the portal hepatic vein (pv), the multi-faces of hepatic cells (hepatocytes) (h), the near center nuclei (n), the sinusoid space (s), endothelial cell (e), and kupffer cell (k).

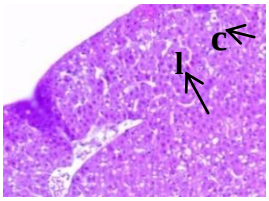

Figure (9): The capsule (C)and the beginning of septa forming between the lobes (I)

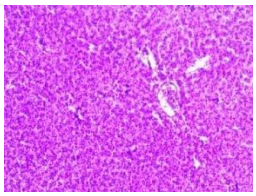

Figure (10): The network localization of the cell within the hepatic tissues

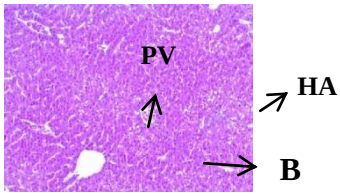

Figure (11): The portal area within the hepatic tissue (PV: portal hepatic vein, HA: hepatic artery, B: bile duct).

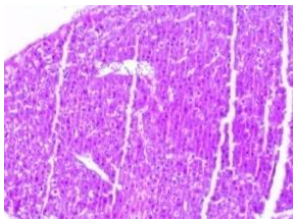

Figure (12): The capsule (C), and the cord line of the hepatic cells

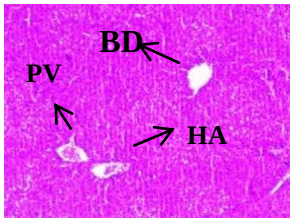

Figure (13): The cord localization of hepatic cells and the portal area (PV: portal hepatic vein, HA: hepatic artery, BD: bile duct).

Discussion

Results of the anatomical study showed the simple differences between the three species in terms of liver development.

The number of lobes decreases the more developed they are according to re. [12]. This agree with our findings. The atrophic lobes were appeared in the liver in both Eurasian siskin and Blackbird and absent in owls. In addition to the differences in size and color due to the difference in the size of each bird and its diet type.

The present study agree with other studies about the liver in different species of birds such as the number of lobes, fragmentation and size [13], [14]. But this study differ with some studies as [15], [16] which were performed on ostrich liver, and showed that it is divided into two main lobes but the right lobe is divided into sub-lobes, and this is not clearly seen in our study. Also the present study differs with re.[1] on seagull liver, as the left lobe is larger than the right lobe and both are segmented.

This is similar to the study on quail and falcon birds, where the left lobe is larger, in addition to the fact that it is divided into two parts for the quail [17]. Besides the study of re.[18] on the local Moorhen birds, which confirmed that the liver has a left and right lobes, but the left lobe is larger in size, and this is different to the liver of the species in the current study.

In the study of re.[2], the liver of Guinea fowl has a segmented left lobe. has, and this is contrary to what we found in our study, but it was agreed that there was a yellow cyst in Guinea fowl. Also, in a study on the liver of an orphan parrot, it consists of two not segmented lobes, left and right. The two lobes have the same size. It is notable that the yellow cyst in an orphan parrot is absence [2].

The study of re. [19] showed that the liver of racing pigeons is divided into two segmented left and right lobes, and this is contrary to what the current study has presented. As in a study on the liver of the kestrel, it has the left lobe which is larger than the right lobe [19].

According to the above results, results on histological study of the three species, it was found that there is a similarity in the basic structure with clear differences manifested in . (the lobular barriers, the hepatic tissue, and the portal space (the hepatic triad). The capsular appears thin in each of the studied species, and the barriers between the lobules are not clear. In the blackbird, the capsular appears divided by non-continuous septa into the inside of the liver tissue. The liver cells located as the chordate position in both owls and Eurasian siskin. In the blackbird, the tissue appears reticulocyte as it was observed that the random positioning of cells and the beginning of the chordal alignment. The portal space is clearly developed in the owl bird, while in the other two species it appears a quasi-portal space where a divergence is observed between its components of the artery, vein and bile duct.

This study agrees with the studies of the liver on the domestic sparrow [20], [21], the seagull [1], the budgerigar [2], the starling bird [13], local Moorhen birds [18], the racing pigeon [19], and the kestrel and the laughing pigeon and the white-throated kingfisher [22] in terms of histology structure of each of the studied species and the shape and position of the cells, as is the case for the chordal alignment of cells in the hepatic tissue of both Eurasian siskin and owls, but this is inconsistent with our results on the livers of the blackbird with retinal tissue, and our study also supported the idea of the inability to distinguish the barriers between the hepatic lobules.

Some differences were found in the histological study of the liver as the following:

-The thickness of the capsule in the present study was thin unlike re. [1] on the liver of the seagull, which showed that the capsule in the seagull is thick, and the same re. [2] on orphaned parrots and Guinea fowl which have medium thickness capsule.

-In the present study, the hepatic cell contains more than one nuclei in the three studied species, while in re.[2], the hepatic cell of budgerigar contain a spherical or oval nucleus with a single central nuclei.

V. CONCLUSION AND FUTURE SCOPE

The comparative histological and anatomical study of the liver of the three species , it was found that there were simple differences between them, which were manifested in (the difference in the color and size of the liver depending on the type , the presence and absence of atrophic lobes and barriers between lobules ,the development of the portal triad and the hepatic tissue that ranged between reticular and reticular).

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