

The Morphological Description and Histological Structure of the Liver in Marsh Harrier Bird in Iraq (*Circus aeruginosus*)

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Abstract: The liver of marsh harrier grossly appeared as large, bi-lobed organ divided into left and right lobes, which are approximately equal in size and not divided into secondary lobes. Histologically, the liver of marsh harrier was found to contain numerous lobules which are not well defined by the connective tissue of the septa except that surrounded the portal triads. The parenchyma of liver composed of irregular branching cords of hepatocytes organized in double rows alternating with tortuous path sinusoids which are lined with flattened endothelial cells and large, irregular outlined kupffer cells. Hepatic cords arranged in a radial pattern around the central vein of the liver lobule while in a subcapsular region they run parallel to the capsule. Portal areas contained a large branch of the portal vein that usually accompanied by another smaller one, hepatic artery and 1-4 branches of variable size bile duct that lined by simple cuboidal epithelium and contained abundant secretory material in their lumen. Portal triad structures are surrounded by moderate amounts of connective tissue, which contains few smooth muscle fibers, the later was also observed within the capsule that surrounded the liver lobes. Glycogen granules stored in liver cells stained moderately with PAS stain technique.

Keywords: morphology, histology, liver, harrier, bird

1. Introduction

The liver is the largest mass of the glandular tissue in the body and is intimately associated with the digestive tract [1]. It plays an important role in the intermediate metabolism of proteins, carbohydrates, and lipids, and also functions as a storage organ for fat-soluble vitamins (A, D, E, and K) and glucose in the form of glycogen. In addition, the liver is an extremely powerful organ that responsible for detoxification and removal of toxin compounds, and other materials, and production of the most plasma proteins like albumin as well as the production of the bile [2].

Anatomically, avian liver consists of two lobes, called the right and left lobes, which are connected cranially at the midline by a small narrow isthmus. The right lobe usually being the larger in most avian species like the pigeon, and ostrich, while the two lobes can be equal sizes in fowl, and rarely the left lobe of the liver is larger than the right in some species. Additional subdivision into secondary lobes may occur in right or left lobes [3]. Histologically, the liver of avian similar to that in mammals but there are some differences such as absent connective tissue septa between lobules except that bounded the triad [4]. The marsh harrier bird is one of the wild carnivorous birds that belong to the family Accipitridae [5], and this type of birds recorded as a breeding resident; winter visitor and passage migrant in Iraq [6]. The aim of the present study was to investigate the morphological and histological features of the liver in marsh harrier in order to show some possible differences with respect to other avian species.

2. Materials and Method

A total of six healthy mature marsh harrier birds of both sexes were used in this study, purchased from specific markets at Baghdad province. These birds were euthanized prior to its dissection by diethyl ether inhalation. After death,

the body cavity was open by made a longitudinal incision through the mid-ventral surface of each bird to expose internal visceral organs. After that, macroscopic observations about the location, color and shape of the livers recorded and photographs were taken by a digital camera for the liver within the body of the bird. For histological study the specimens were taken from different area of the right and left lobes and immersed in 10% formalin or Bouin's solution for 24 hours, after fixation the specimens were dehydrated by (70%, 80%, 90%, and 100%) ethanol and then clearing process was done by using xylene for one hour. Tissue samples were embedded in paraffin wax and sectioned serially at 5-7 µm thickness and stained by Hematoxylin & Eosin stains to demonstrate general features, and Periodic acid-Schiff (PAS) Stain to demonstrate collagen materials and Masson's trichrome stain to demonstrate collagen fibers [7]. Later, photographs of the sections were taken by digital camera connected to Olympus microscope.

3. Results and Discussion

The liver grossly appeared as large organ had reddish brown to dark brown color. It is located in the anterior portion of the body cavity and situated ventral and caudal to the heart and cranial to the gizzard (Fig 1), such description for the position and color of the liver in marsh harrier was similar to that recorded by [8] in local Moorhen and by [9] in the male indigenous turkey. According to [10] the nutritional state of the adult bird effect on the normal color of the liver, it usually has a dark red to red-brown color that changes to a yellow-tan if the bird on a high-fat diet. The macroscopic examination also revealed that the liver of harrier was a bi-lobed organ divided into left and right lobes, which are approximately equal in size and undivided into secondary lobes (Fig. 1). These findings were differing from those observed by [11] who reported that the liver of local coat bird has two lobes, the right lobe was larger than the left lobe which is divided into two secondary lobes, while it was

in accordance with [12] who declared that there were no further lobar subdivisions are present in the liver of bustard. A small interconnecting portion was seen cranially at midline connected the two lobes of the liver, which was also observed in other avian species. The gallbladder located on the inferior surface of the right lobe of the liver, which was absent in many other species such as ostrich and pigeons and many parrots [3; 13].

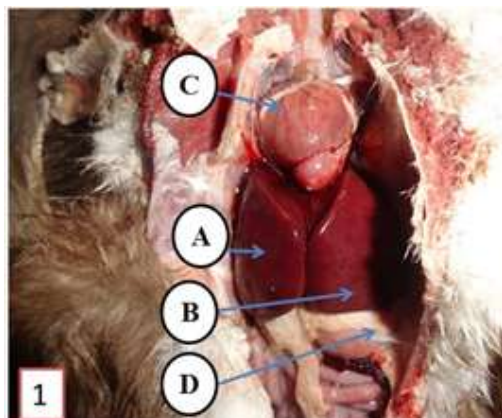


Figure 1: Showed the right and left lobes of marsh Harrier which are nearly equal in size and its position in body cavity: right lobe of liver (A), left lobe of liver (B), heart (C) and gizzard (D).

Histological analysis showed that the outer surface of the liver lobes was covered by a thin layer of flattened mesothelial cells, the capsule serosa, below which there was a dense regular connective tissue layer, which is named as Glisson's capsule as that documented in other avian species such as the liver of the ostrich, Iraqi local duck, and coot bird [11; 14; 15]. The later capsule (Glisson's capsule) mainly was composed of collagen fibers along with smooth muscle fibers which were present, especially in the deeper parts and also contain fibroblasts and blood vessels (Fig. 2). This structure of the capsule is similar to that observed by [14] in the ostrich. [14] suggested that the serosal coat might be an additional source of blood supply to the liver, especially for the adjacent parts of the parenchyma.

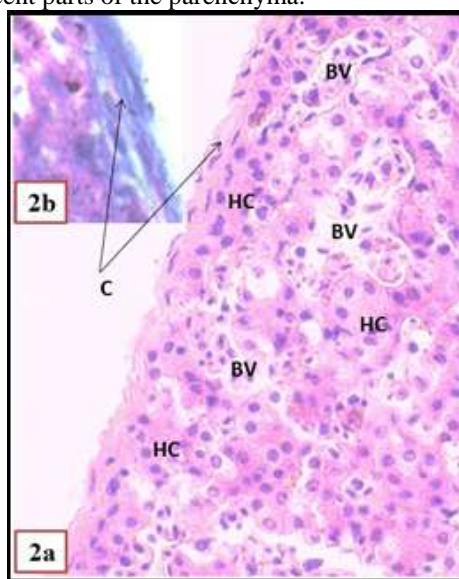


Figure 2: Cross section of the Liver of marsh harrier showed Capsule (C), blood vessels (BV) and hepatic cords (HC) (a: H & E, X400, b: Masson's trichrome stain).

The parenchyma of the liver in marsh harrier birds is divided into many lobules which are not clearly defined and blended with each other at the peripheries due to the poorly developed or absence of the interlobular connective septa, except in the portal canals which considered the only signs that clearly demarcated the liver lobules (Fig. 3). This observation is in accordance with the findings obtained from prior studies performed in other species of birds like ostrich, duck, and turkey [9; 13; 15], whose results showed that the lobulation of the liver was indistinct. The hepatic parenchymal tissue was constituted by numerous epithelial cells called hepatocytes which are arranged in irregular branching cords or plates, each one of them contains two rows of hepatocytes and follow the course of the sinusoids throughout the liver tissue (Fig. 4, 5). This finding is in agreement with those reported by [9] in male indigenous turkey, by [8] in local Moorhen birds and by [16] in Ruddy Shelduck and Collared Dove and in contrast with [17] who indicated that hepatic laminae composed of (1-2) cells in thickness in *passer domesticus* and also [8] in the three species, *Laruscanus*, *Agaporinsfischeri* and *Numidameleagris*. The current study revealed that the hepatic cords present in the subcapsular region arranged parallel to the capsule while those present inward (within lobules) were organized in a radial manner around the central vein which occupied the core of each hepatic lobule (Fig. 2, 3a, 3b) and such parenchymal arrangement resembles that described by [8; 18].

The central vein varied in size and has circular or tortuous lumen that lined with a single layer of squamous endothelial cells supported by a scanty collagenous connective tissue and contains nucleated erythrocytes (Fig. 3b). A similar observation was made earlier by [8; 18]. Hepatocytes are seen as a large, polygonal cells with unclear cell boundaries. Their cytoplasm intensely stained with eosin and contains a few granules as well as small or tiny vacuoles. Nuclei appeared round with one or two prominent nucleoli and most of them lightly stained with hematoxylin and eccentrically placed in the cells (Fig. 4, 5). The sections of liver stained with PAS showed moderately positive reaction (Fig. 5) indicating the presence of glycogen granules that are stained purple or red due to the neutral mucous substance nature of these granules. These granules distributed randomly within hepatocytes with a tendency to concentrate peripherally.

The hepatic sinusoids were coursing among the hepatocyte plates as irregular, thin-walled tubular channels, that had wide lumina contains nucleated red blood cells and few leukocytes. The lining of these vascular capillaries mainly formed from a discontinuous sheet of small, flattened endothelial cells, which contain dark elongated nuclei as shown in figure (Fig. 4, 5). The structure and the tortuous course of sinusoids throughout the liver tissue allows for an efficient exchange of materials and solutes between hepatocytes and blood [2]. Kupffer cells represent the second type of cells that contribute in forming of the sinusoidal lining in the liver and observed either in close contact with sinusoidal endothelial or sometimes laying free in the sinusoidal lumen. These cells appeared as large, and irregular in outline with several cytoplasmic processes. Their nuclei are oval or rounded in shape, lightly stained and

surrounded by granular lightly eosinophilic cytoplasm (Fig 4,5).[13] reported that Kupffer cells, distinguish morphologically from the endothelial cells, by their strongly protruding into the sinusoidal lumen and their satellite appearance with many microvillus pseudopods in the liver of ostrich. Kupffer cells are specialized liver phagocytes that

are the predominant cell type of the hepatic reticuloendothelial system. These cells filter and phagocytose damaged erythrocytes, infectious agent, cellular debris as well as the particulate material from systemic circulation.[2; 19].

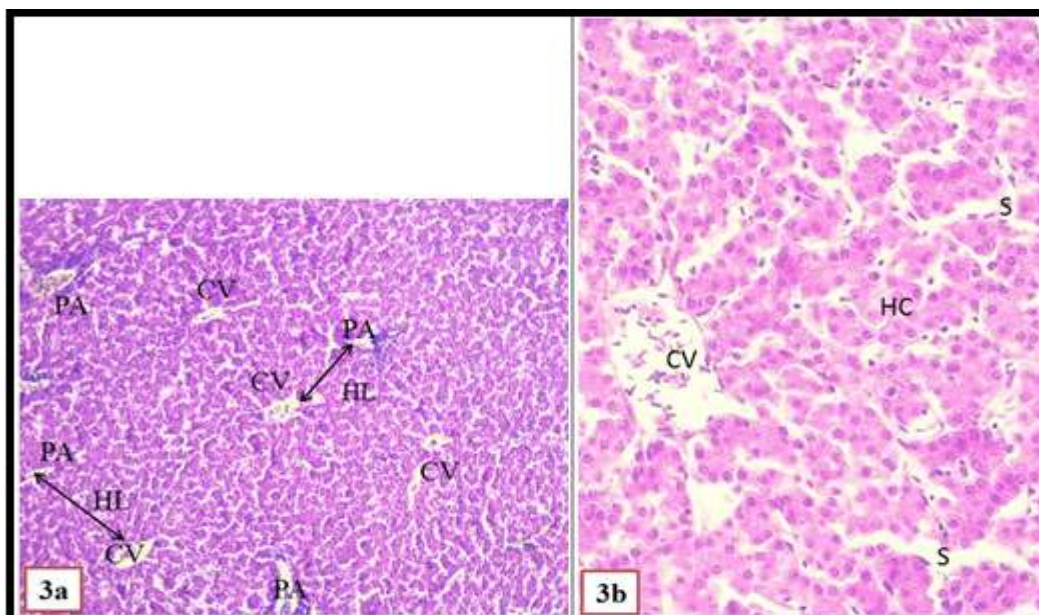


Figure 3: Transverse section of marsh harrier liver showing hepatic lobules(HL), Central vein(CV), portal area(Pa), liver sinusoid (S) (H&E, a: X10, b:X400). Note RBC in central vein and sinusoids

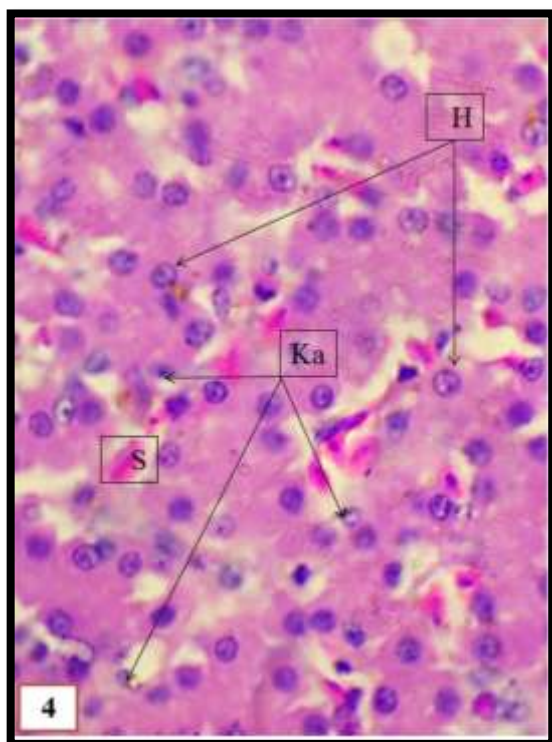


Figure 4: Transverse section of marsh harrier liver showed hepatocytes (H) , liver sinusoids (S) , Kupffer's cells (Ka) (H&E, X1000)

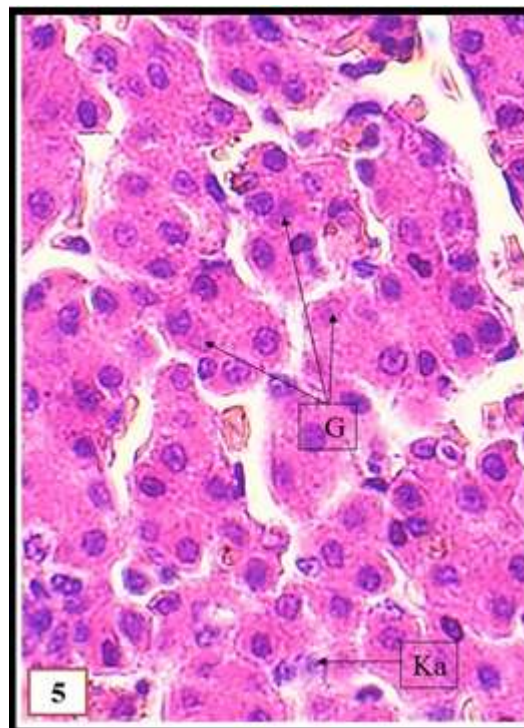


Figure 5: Transverse section of marsh harrier liver showed glycogen granules(G) stored in hepatocytes, Kupffer's cells (Ka) (PAS, X1000)

Portal tracts are formed by the interlobular connective tissue septa and distributed randomly at junctional sites between the adjoining lobules. It contains a moderate amount of collagenous connective tissue along with few smooth muscle fibers surrounding a branch of portal vein, a branch of the

hepatic artery and 1 to 4 branches of bile duct (Fig.6a,6b). In addition to a large branch of portal vein, a small branch of portal vein also usually presents in the portal area. In contrast, [14] documented that small branches of a hepatic artery, portal vein and bile ducts were observed throughout the hepatic parenchyma instead of the typical portal triads that were absent in the liver of ostrich. The current study indicated that both the wide and narrow lumens of the interlobular vein and interlobular artery, respectively, were contained nucleated red blood cells as well as few leukocytes and outlined by a single sheet of endothelial cells rested on a basal lamina and bounded by a layer of smooth muscle fibers that appears thicker in artery in comparison to the corresponded vein (Fig.6a).

The interlobular bile duct in this study was found to be lined by a cuboidal cell layer covered by a loose connective tissue with smooth muscle fibers present around larger ducts. This result agrees with [16] who mentioned that the bile ducts are lined by simple cuboidal epithelium and surrounded by a

loose connective tissue with smooth muscles around the larger ducts in the liver of Collared dove and Ruddy Shelduck. The elliptical nuclei were euchromatic and filled most of the cell; they had a thick conspicuous nuclear membrane and usually contained one prominent nucleolus located eccentrically. The cytoplasm was less intensely stained with eosin. These results partially agreed with [14] who reported that cells lined bile duct contained centrally located nuclei and faint acidophilic cytoplasm. The lumen of bile ducts contained abundant secretory material (Fig.6a) and such a result suggests that the liver cells were very active in the production of the bile which was very necessary to facilitate digestion of fats in the food of this carnivorous bird. The present study showed the presence of numerous lymphocyte aggregations in the liver parenchyma of the harrier (Fig.7) similar to that noted by [13] in African ostrich. According to [13; 14], these patches of lymphocytes may attribute to a focal area of lymphocytes as immune patches.

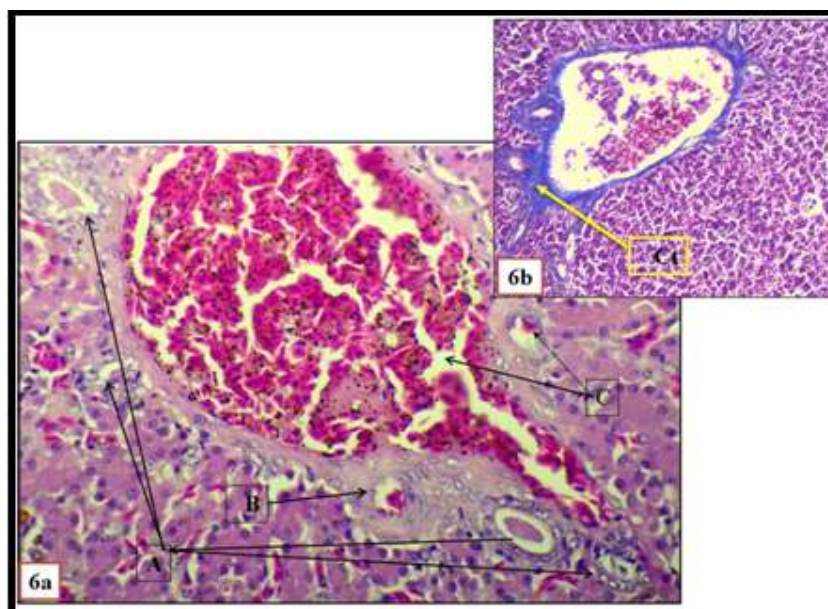


Figure 6: Transverse section of liver in marsh harrier. Note portal area: Bile duct (A), Hepatic portal artery (B), Hepatic portal vein (C), Connective tissue, (a: H & E X 400, b: Masson's trichrome stain, X10).

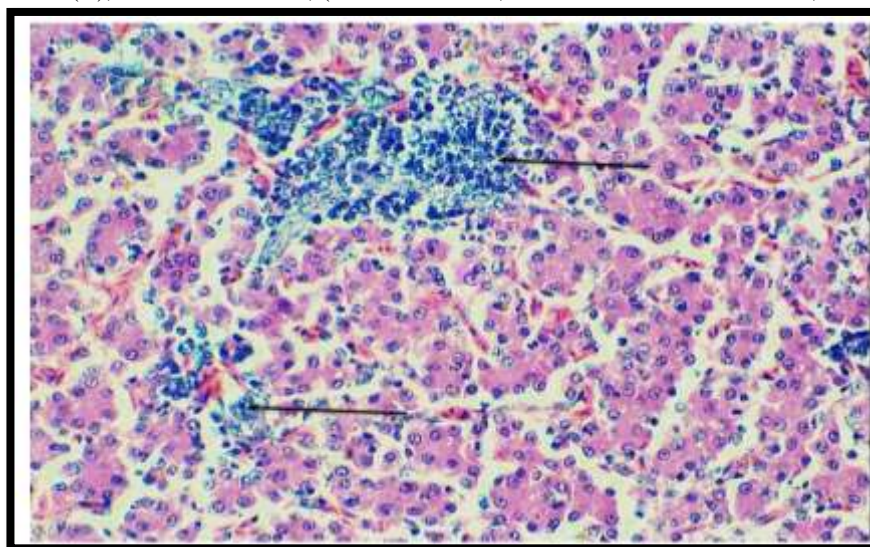


Figure 7: Cross section in the liver of marsh harrier showing patches of lymphocytes between the hepatic cords (arrow) (H&E, X400)

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