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Liver histological structure in adult European bison. *Bison bonasus* (Linnaeus, 1758)

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The histological structure of the liver in 12 European bison, 2–8 years old, was presented. The study of the hepatic lobule showed that in Bison bonasus *there were no connective septa around the hepatic lobules. On the hepatic lobule territory, solitary bile ducts, respectively solitary arterioles, were sometimes observed. Every one of these vessels was isolated from the hepatic parenchyma by thick coats of collagen. In the adventitia of the hepatic sublobar vessels, capillaries of the vasa vasorum system were found. The thick capsule of Glisson presented an outer part lacking in blood vessels and an inner part vascularised with arterioles and capillaries originating from the subcapsular portal tracts. The ligament insertion in the liver parenchyma was described.*

key words: capsule of Glisson, hepatic ligament, hepatic lobule, portal tract, solitary arteriole, solitary bile duct, vasa vasorum

INTRODUCTION

The histological liver structure of the representatives of the suborder Ruminantia has been little studied. This fact is surprising, considering the economic importance of a part of the species of this taxonomic group and also the great hunting interest in other species belonging to this group.

The liver of *Bison bonasus* was a massive organ, with the medium weight of 9–10 kg. It was constituted of 4 main lobes. The hepatic parenchyma was distributed unequally between these lobes; therefore, some liver morphologic features presented individual variability [11].

An anatomic study on the hepatic venous system in the bison liver was realised with the aim to define the conspicuous origin of the hepatic veins and their pathways through the hepatic lobes [12].

Until now, only one paper has presented the histological structure of the European bison liver [15]. This paper was realised after the study of one 10-year-old bison and presented a brief description of the hepatic lobule surrounded by "peripheral trabecula" and some data about the hepatocyte dimensions.

In the present paper, after the study of the liver of 12 adult European bison, the hepatic lobule structure lacked interlobular connective septa, the capsule of Glisson structure and the ligament insertion on the liver parenchyma were described.

MATERIAL AND METHODS

The liver samples of 12 European bison of both sexes, 2–8 years old, were collected between December 1999 — February 2000, on the territory of Białowieża Primeval Forest (Poland). The animals were culled by shooting, for the stabilisation of the population.

The liver samples were collected and fixed for histological investigations by workers of the Mam-

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mal Research Institute of the Polish Academy of Sciences and Białowieża National Park. The bison liver samples were fixed in 8% formaldehyde in saline. After the routine histological procedures, the blocks in paraffin were sectioned at 5–7 μm and stained with Hemalum-Eosin (H-E), Perls method for ferric ion evidentiation and with picro-Sirius red stain (PSR) [8] — the specific histochemical stain for collagen fibres. Some liver samples were serially sectioned to more precisely state the micro-anatomical connections between different hepatic structures.

For this study, 50 liver samples embedded in paraffin were processed and about 2,200 histological sections were obtained. Special attention was paid to the capsule of Glisson and the histological structure of the ligament insertion in the hepatic parenchyma.

RESULTS

The Histology of the Hepatic Lobule

In the bison liver, the hepatic lobule can be identified by the presence of the central vein and the existence of 3–6 afferent portal profiles in cross, oblique or longitudinal section. Between the portal tracts defining the limits of a hepatic lobule, there were no fibrous septa (Fig. 1). The lobule may be marked by terminal portal tracts with barely distinct lumens of the portal vein, arteriole and bile duct but also by great portal tracts with great or medium sized portal vessels (Figs. 1–3). Infrequently, hepatic lobules outlined by thick collagenous septa appeared. This feature is not typical for the hepatic lobule organisation in the bison liver. It may be observed only in the parenchyma of the liver lobe edges.

The striking feature of the portal system in the bison liver is the pronounced thickness of the collagen sheaths of the great and medium sized veins. The portal bile ducts were surrounded by many collagen fibres. The portal artery was separated from the other portal vessels by a thick coat of connective tissue. Every principal or intermediate portal tract was separated towards the hepatic parenchyma by a thick layer of connective tissue (Fig. 2).

The hepatic parenchyma was constituted of rows of hepatocytes, forming hepatic cords radially orientated towards the central vein and of the hepatic sinusoids with irregular outlines. Picro-Sirius red stain indicated the presence of an important amount of collagen situated perisinusoidally. In some zones of the hepatic lobes, especially the subcapsular zone, there were more collagen fibres. Also, in these zones, the nuclei of the hepatocytes were hyperchromatic.

The hepatocyte was a small cell of polyhedrical-ovoid shape, with a great vesiculiform nucleus which occupied the central zone of the hepatocyte. Binucleated hepatocytes were not frequent.

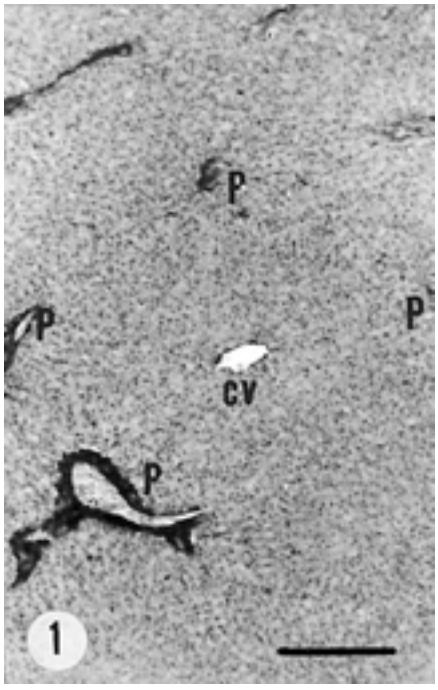

Figure 1. Typical hepatic lobule lacking in fibrous septa. Central vein (cv) and different sized portal tracts (P). PSR, scale bar, 200 μm.

Figure 2. Great sized portal tract (P) with thick sheath of collagen. H-E, scale bar, 150 μm.

Figure 3. Medium sized portal tract (P). PSR, scale bar, 50 μm.

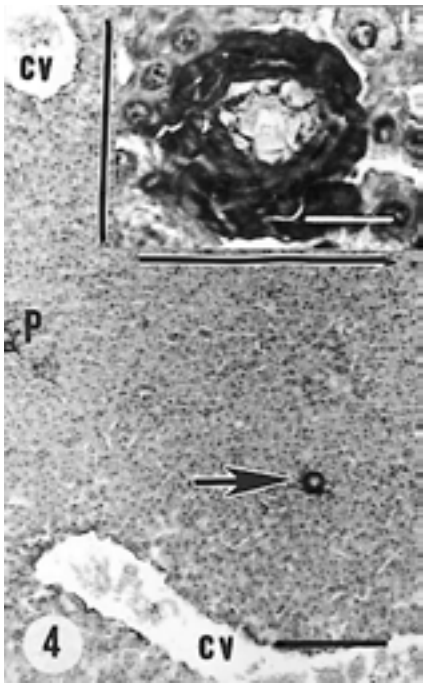

Figure 4. Solitary bile duct in the hepatic lobule (arrow). Note, two central veins (cv), portal tract (P). PSR, scale bar, 200 μm. **Inset:** Solitary bile duct (detail). PSR, scale bar, 20 μm.

The rare perisinusoidal stellate cells presented great lipid vacuoles distorting the nucleus.

The Kupffer cells were present in the endothelial lining of the sinusoid. As a rule, they did not present in the cytoplasm Perls-positive secondary lysosomes containing ferric ion accumulations. This reaction was positive for small groups of hepatocytes scattered in the liver parenchyma.

An interesting structure was the solitary bile duct encountered sometimes in the liver parenchyma (Fig. 4 and inset). These ducts were formed by typical biliary cells and were always surrounded by a thick layer of collagen fibres. In this collagenous mass, 1–2 cross--cut capillary lumens may be visible. The solitary bile ducts had a sudden appearance right in the hepatic lobule, making the connection between the bile canalicles with the portal bile ducts.

Other independent vessels which travel long lines through the bison hepatic lobules were the hepatic arterioles or capillaries (Fig. 5). They were protected towards the neighbouring hepatocytes and sinusoids by thick Sirius red positive coats of collagen fibres.

The hepatic sublobar veins presented a large adventitia with rare capillaries of the vasa vasorum system (Fig. 6 and inset).

The central veins presented a moderate amount of collagen in the wall structure (Figs. 1, 4). At the level of the central veins capillaries of the vasa vasorum system were never observed.

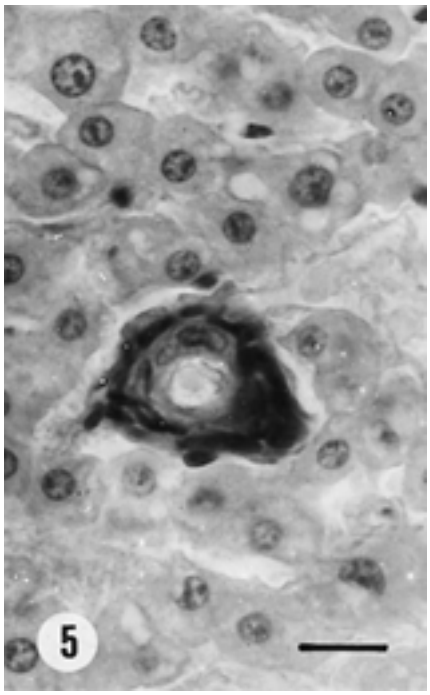

Figure 5. Independent arteriole surrounded by thick coat of collagen (C). PSR, scale bar, 20 μm.

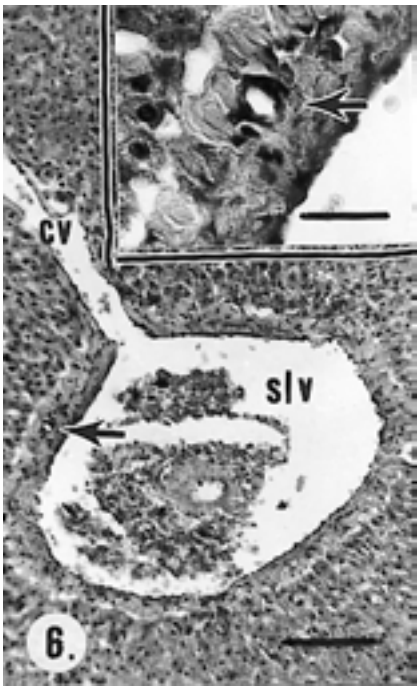

Figure 6. Sublobar vein (slv) communicates with a terminal central vein (cv). Note large adventitia and a capillary (arrow) of the vasa vasorum system. H-E, scale bar, 100 μm. **Inset:** capillary of the vasa vasorum system (arrow). Detail. H-E, scale bar, 50 μm.

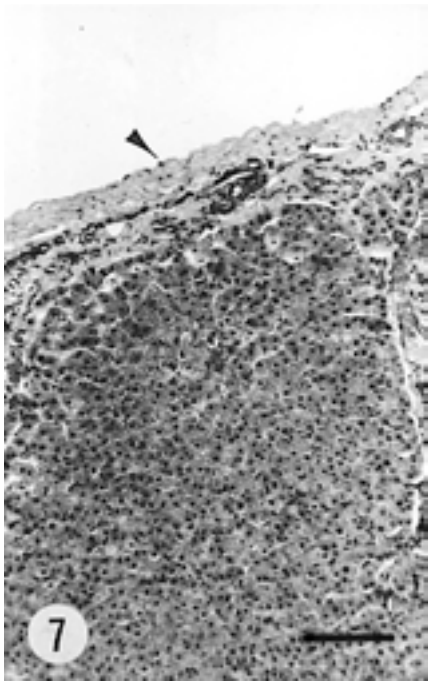

Figure 7. Capsule of Glisson covered by a flat thin mesothelium (arrow head). In the inner part of the Glisson capsule there are numerous profiles of arterioles and capillaries. H-E, scale bar, 100 μm.

The Capsule of Glisson

The bison liver was covered by a well fibrosed capsule. At the outermost level, the capsule of Glisson was covered by the corresponding segment of visceral membrane of the peritoneum, formed by mesothelial flat cells (Fig. 7).

The capsule of Glisson of 100–300 μm thickness was constituted by two distinct parts:

— the outer part was constituted by a few connective cells and many layers of collagen fibres with faint Sirius red positivity (Figs. 7, 8);

— the inner part was constituted by many connective cells and a great number of collagen fibres, with good affinity for Sirius red stain. The inner part of the capsule of Glisson was supplied by the subcapsular arterioles which branched into capillaries. The inner part of the capsule of Glisson would be considered as a generating layer of the capsule.

Numerous lymphatic vessels (Figs. 7, 8) were noted at the limit between the two parts of the capsule of Glisson.

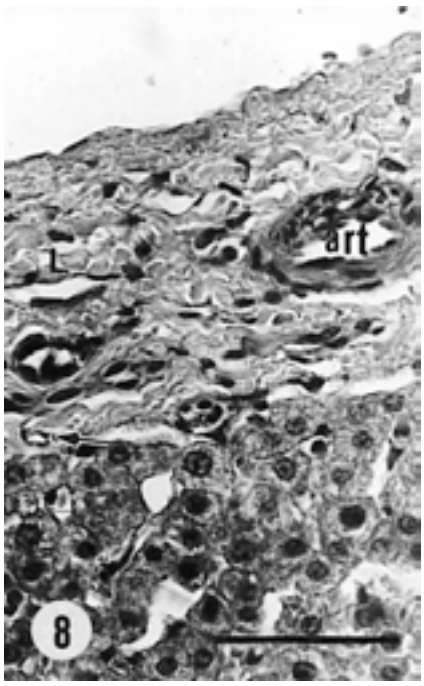

Figure 8. Capsule of Glisson presented an outer layer of collagen fibres and an inner layer with many arterioles (art), lymphatic vessels (L) and capillaries (arrow). H-E, scale bar, 50 μm.

The Histological Structure of the Ligaments

The connective fascicles of the ligaments were organised in parallel planes oriented towards the traction strengths applied on the whole liver.

The histological sections through the hepatic parenchyma, situated between the deep ramifications of the coronary hepatic ligament, revealed that

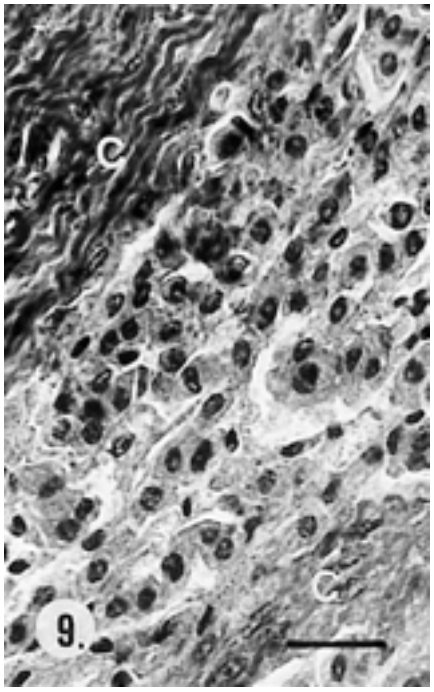

Figure 9. Fragment of hepatic parenchyma situated between two profound ligament ramifications. Hepatocytes are not organised in hepatic cords (C — collagen). PSR, scale bar, 20 μm.

the connective structure of this ligament grew proportionally with the hepatic lobe growth. Between two or more fibrous structures of the ligament, zones of the altered hepatic parenchyma remained arrested. Between two profound, compact beds of the coronary hepatic ligament, the hepatic tissue appeared disorganised: the hepatocytes did not remain bound in cords (Fig. 9).

The vascularisation of the connective tissue of the ligament was realised by the portal arteries. The fibrous walls of the portal tracts were in continuity with some profound fascicles of the ligament, contributing to the enlargement of the ligament insertion area.

DISCUSSION

The histological structure of the liver in *Bison bonasus* has been little studied [15]. Also, little attention has been paid to the liver structure of the breeds of zootechnical importance belonging to the species *Bos taurus*.

Three decades ago, it was considered [1, 7] that the connective septa separating more or less completely the hepatic lobules began from the capsule of Glisson and met the perivascular connective tissue marking the limits of the numerous hepatic lobules.

Our observations on the histological sections showed that in the bison liver the relations between the periportal connective tissue and the capsule Glisson were realised by the proliferation of the subcapsular portal arterioles until the inner side of the capsule of Glisson. These arterioles were always accompanied by thick layers of collagen. We never saw the least sign which could demonstrate that the connective perivascular tissue originated in the capsule of Glisson.

The conceptions about the capsular origin of the connective tissue of the interlobular septa and portal area have an old tradition in hepatology [1, 3, 6]. The structural and functional similitude between the connective tissue of capsule of Glisson with the connective tissue of the portal tract was discussed by Cormack [2].

Ekataksin [4] made a brief presentation of the arterial vascularisation of the capsule of Glisson and the arteriolar supply in the hepatic veins walls in the ox. The presence of the arterial vascular structures in the capsule of Glisson and in the adventitia of the hepatic veins was confirmed also in *Bison bonasus* by the descriptions and illustrations of this paper.

The presentation in this paper of the isolated arterioles with intralobular, extraportal pathways in the bison liver represented the first description for this species. Also, the intralobular, extraportal presence of some solitary bile ducts was described here for the first time.

The structure of the bison hepatic lobule was similar to that of the ox [1]. Only exceptionally, a few groups of hepatic lobules situated at the lobe edges presented incomplete perilobular connective septa. The typical hepatic lobule of *Bison bonasus* was punctually marked by 3–6–7 portal area in oblique or cross sections. In the centre of the hepatic lobule was a terminal or subterminal hepatic vein with slightly fibrosed wall.

In the frame of the order Artiodactyla, the most studied liver was that of the species *Sus scrofa* [13, 14]. The striking feature of this liver was the existence of the simple or compound lobules separated by continuous connective septa, where the portal triads and the sublobar hepatic veins were housed [5, 13, 14].

In the liver of the suborders Suiformes and Ruminantia, the capsule of Glisson presented the same striking features: the thickness of the capsule due to the great quantity of the collagen fibres and the high degree of arterial vascularisation. So, the capsule of Glisson contributed to the resistance structure for supporting the great amount of hepatic parenchyma.

A comparison with the capsule of Glisson of the *Ursus arctos* liver (our unpublished data) revealed the possibility that the important liver dimensions in the order Carnivora might be supported by a thinner and less vascularised capsule of Glisson than in Ruminantia.

Unlike the pig liver, which presented interlobular connective septa, in the ox and bison liver there was noted the presence of a great amount of connective tissue concentrated along the portal tracts and hepatic veins. Such a connective superstructure of the main hepatic vessels formed an inner resistant and elastic skeleton capable of maintaining the functionality of the great mass of hepatic parenchyma.

The microvascular lesions described in steers [9], defined as pretelangiectasia and telangiectasia, were never observed in individuals of *Bison bonasus*. Hepatic lesions similar to those signalled by Piusiński et al. [10] and considered to be due to parasitosis with *Fasciola hepatica* were observed only in one of the 12 bison studied.

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