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Shevchuk M. M.

MACRO- AND MICROSTRUCTURAL LIVER ARRANGEMENT IN WHITE RATS IN HEALTH

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Macro- and microstructural study of the liver of white rats was carried out. It was established that weight of the liver in a white mature rat weighing 180-230 g is 11-16 g, and weight of the liver in immature rats is 6-8 g. It consists of 6 lobes, externally covered with connective tissue capsule, which is tightly fused with the visceral layer of the peritoneum. On the visceral surface, there is a porta hepatis with portal vein, own hepatic artery and hepatic ducts forming the common bile duct. Rats have no gallbladder. The internal structure of the liver is formed by parenchyma and stroma. The liver parenchyma is represented by hepatic lobules, which are formed by two rows of polygonal hepatocytes. Hepatic tubules are formed by hepatocytes. Hepatocytes are polygonal in shape, they are located in tubules in two rows. Hepatocytes are heteromorphic depending on the hepatic lobe and the region and size within a specific region of the lobule. Interlobular connective tissue trabeculae are poorly developed. Interlobular blood vessels and bile ducts in portal tracts form large vascular bundles consisting of 2-3 portal vein branches, 2-3 hepatic artery branches, and 3-4 bile ducts. Central veins were located in the center of classical hepatic lobules. Endothelial layer in the walls of the central veins was solid. Endotheliocytes were homogeneous in structure, which was characteristic for these cells.

Key words: *rat, liver, hepatocyte.*

Connection of the study with planned scientific research projects. The results of these studies were obtained by the author during the research project of Danylo Halytskyi Lviv National Medical University (Department of Pathological Anatomy and Forensic Medicine) on "Study of pathomorphological features of diseases of thyroid gland, cardiovascular digestive, urinary and reproductive systems, and the perinatal period, for improving their morphological diagnosis" (state registration № 0118U000100).

Introduction. Studies of the liver in recent years indicate functional and morphological hepatic heterogeneity [1, 2]. Heterogeneity in the structure of the liver is associated with differences between the lobes of the liver and different zones of their lobules [3]. Studies suggest that morphofunctional hepatic heterogeneity is due to the fact that pathological changes and the dynamics of their spread in the liver under various pathological conditions at the initial stages have a certain characteristic localization, which, in particular, is connected with the specific nature of its structure [4, 5]. Special mention in this regard should be made of subcapsular area of the liver because it was scantily studied [6, 7], as well as due to statistically frequent cases of its involvement in the damage area during various pathological processes.

Aim of the study is to investigate and describe morphological features of the rat liver in health.

Object and research methods. Rats were kept on a standard vivarium diet with free and unrestricted access to water. All animals were kept in the conditions of the vivarium of Danylo Halytskyi Lviv National Medical University, the experiments were conducted in accordance with the provisions of the European Convention for the Protection of Vertebrate Animals Used for Experimental and

other Scientific Purposes (Strasbourg, 1986), Council of Europe Directive 86/609/EEC (1986), Law of Ukraine No. 3447-IV "On the Protection of Animals from Cruelty". The experiments were conducted in accordance with Minutes No. 7 dated 29.08.2022 of the Commission on the ethics of scientific research, experimental developments and scientific papers of Danylo Halytskyi Lviv National Medical University. Sampling for this research was carried out under ether anesthesia. Liver biopsy specimens of male rats weighing 180-230 g were the material for the study. Since liver is a large organ, tissue samples from each animal were collected simultaneously for histological examination and for electron microscopy, which allowed optimization of the use of biological material and reduction in the number of animals required for the study. Liver samples were taken randomly, at least 5 fragments from different lobes for making histological preparations and for electron microscopy. Thickness of samples for histological preparations did not exceed 4 mm, while thickness of samples for electron microscopy was no more than 1 mm. Samples for making histological preparations were fixed in 10% buffered formalin. Afterwards, these samples were dehydrated by processing in increasing concentration of ethyl alcohol (60%, 70%, 80%, 90%, 95%, for 2 hours, and twice 100% for 30 minutes), and embedded in paraffin in a thermostat at a temperature of 60°C with an intermediate processing in alcohol-xylene (50 to 50) and 100% xylene, 30 minutes in each solution. For morphometry, sections with a thickness of 3 μm were stained with hematoxylin-eosin [8]. Immediately after sampling, material for electron microscopy was placed in 2% osmium tetroxide solution in phosphate buffered saline (0.1M, pH 7.36) with sucrose. The strips were re-transferred into a drop of the same solution placed on wax on an ice slab, and blocks of 1 mm^3 were cut out. Next, these blocks were fixed also with 2% osmium tetroxide solution in phosphate buffered saline (0.1 M, pH 7.36) with the addition of sucrose for 2 hours in an iced bath. Afterwards, these blocks were washed with buffered solution 4 times for 15 minutes. After washing, dehydration was carried out before pouring into non-water-soluble resins. For this purpose, samples were processed in a series of ethyl alcohol solutions: three portions of 40% for 10 minutes, three portions of 70% for 10 minutes, two portions of 96% for 20 minutes. Next, they were processed in pure acetone 6 times for 15 minutes, followed by processing in a mixture of resins and acetone: two hours in a mixture of acetone and resin at the ratio of 3 to 1, two hours in a mixture of acetone and resin at the ratio of 1 to 1, two hours in a mixture of acetone and resin at the ratio of 1 to 3. Finally, the blocks were placed in pure resin for 12 hours. Resin was prepared at the following ratio of components: 5 ml of epon 812, 3 ml of Araldite M, 11 ml of DDSA, 0.4 ml of dibutyl phthalate, 15 drops of DMP-30. Slow-speed centrifuge was used during processing in the acetone-resin mixture. For the purpose of polymerization, the blocks were placed by self-immersion in epon-araldite in glycerine capsules, which were placed in thermostat for resin polymerization and kept for 24 hours at the temperatures of 36°C, 45°C, and 60°C. Ultrathin sections were prepared with the help of UMTP-3M microtome, glass knives were made using SSN-1 device. Sections were contrasted in 2% uranyl acetate and lead citrate solution, studied and photographed using UEMV-100K microscope (Ukraine) at an accelerating voltage of 75 kV and 2000x-10000x screen magnification.

Research results and their discussion. Normal weight of the rat liver is 11-16 g in mature animals, and 6-8 g in immature animals, it consists of 6 lobes: right lateral, left lateral, right central, left central, caudate lobe and additional lobe. The rat liver is externally covered with a connective tissue capsule, which is tightly fused with the visceral layer of the peritoneum. The ligament apparatus is poorly pronounced. On the visceral surface, there is a porta hepatis with portal vein, own hepatic artery and hepatic ducts forming the common bile duct. Rats have no gallbladder. The internal structure of the liver is formed by parenchyma and stroma. Hepatic parenchyma is represented by hepatic lobules. Hepatic lobules have polygonal 5-6-sided shape, and consist of hepatic tubules and interlobular sinusoidal capillaries. The center of the lobule is the central vein. A small space called the space of Mall is found around the stromal elements of triads. Hepatic tubules are formed by hepatocytes, and are located in the radial direction. Hepatocytes in tubules are located in two rows, and are connected to each other by desmosomes. Hepatocytes are polygonal in shape. Sometimes solitary segmental triads **(fig. 1)** with pronounced perivascular space are located between the lobes. Interlobular connective tissue trabeculae are poorly developed. Margins of classical hepatic lobes were determined by the location of portal triads, hepatic plates and sinusoidal capillaries, which had a radial direction, from the central vein to the portal tracts **(fig. 1)**. Between the classical hepatic lobules, layers of loose connective tissue are found, in which the components of the hepatic triad are located, such as branches of hepatic artery, portal vein, lymphatic vessels and bile ducts. Fibroblasts, as well as solitary lymphocytes and macrophages are located in the cellular composition of loose connective tissue of the hepatic triads. Hepatic parenchyma is represented by interlobular connective tissue. In rats, margins of the hepatic lobules are visualized only in the direction of the hepatic tubules, and are characterized by regions with randomly located hepatocytes and porto-portal shunts.

More pronounced layers of connective tissue are observed only around interlobular vessels and large diameter vessels. Sinusoidal capillaries of the liver of intact animals have a radial direction from the periphery of the liver lobules to the central vein, they are somewhat tor-

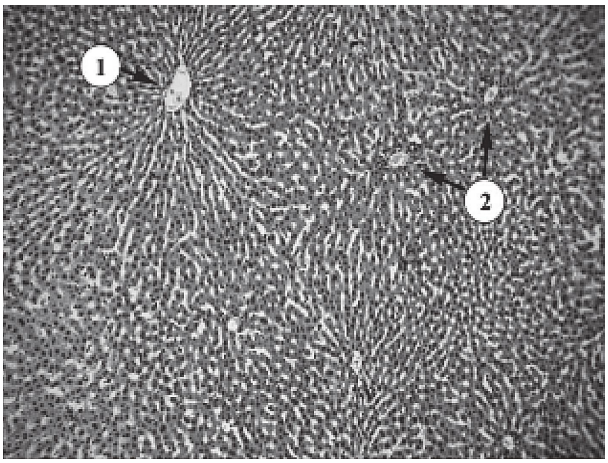

Figure 1 – Rat liver. Micrograph. Hematoxylin and eosin staining. Magnification: ×200. Designation: 1 – classical hepatic lobules with central vein; 2– hepatic triads.

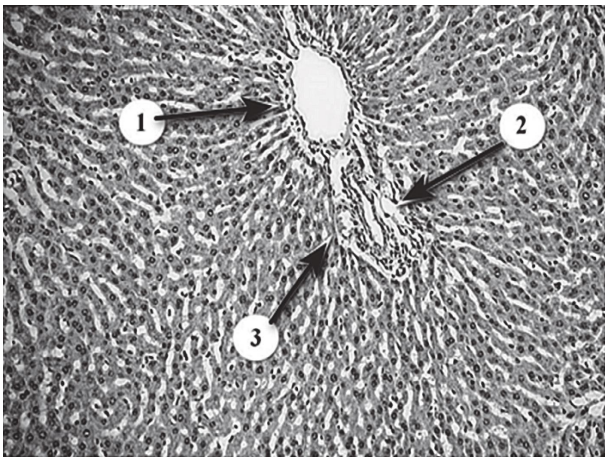

Figure 2 – Interlobular hepatic triad in intact rat. Micrograph. Hematoxylin and eosin staining. Magnification: ×200. Designation: 1 – branch of the portal vein; 2 – bile duct; 3 – branch of the own hepatic artery.

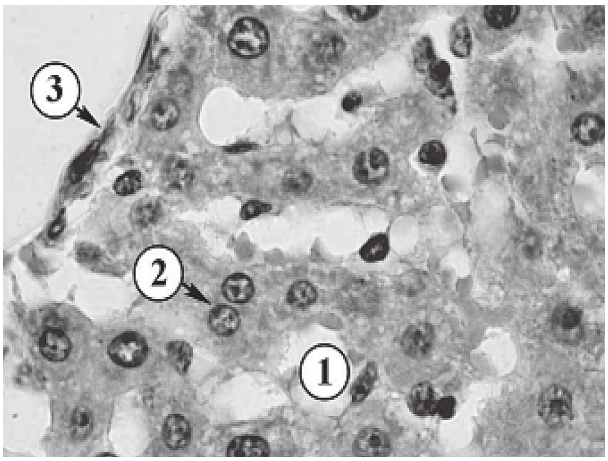

Figure 3 – Subcapsular region of the liver in intact rat. Micrograph. Hematoxylin and eosin staining. Magnification:×1000. Designation: 1 – lumen of sinusoidal capillary; 2 – binuclear hepatocyte; 3 – liver capsule.

tuous, and have a uniform diameter of 4.02 [3.58; 4.64] μm in mature animals ranging from a minimum value of 2.49 μm to a maximum value of 7.33μm. There are no porto-portal connective tissue bridges, these areas were visually indistinguishable from others. Interlobular blood vessels and bile ducts in portal tracts form large vascular bundles consisting of 2-3 portal vein branches, 2-3 hepatic artery branches, and 3-4 bile ducts **(fig. 2)**. On cross-sections, hepatic plates in most classical lobes of the liver had a radial direction.

Sinusoidal capillaries were located between hepatic plates. In intact rats, sinusoidal capillaries are tortuous, slightly narrowed from the portal tracts to the central vein, with solitary erythrocytes and leukocytes found in the lumens of these sinusoidal capillaries, among which large lymphocytes were identified, which nuclei were intensely colored, and granules with dense center were located in the cytoplasm. The diameter of sinusoidal capillaries was 9.6 (8.51; 11.33) μm, ratio of the sectional area of sinusoidal capillaries to the sectional area of the parenchyma was 0.20±0.02. In the subcapsular zone, sinusoidal capillaries with a large diameter of up to 20 μm were often observed, the average diameter of subcapsular sinusoidal capillaries was 13.20±0.30 μm. Walls of sinusoidal capillaries consisted of endotheliocytes and stellate macrophagocytes. Solitary elastic and collagen fibers, processes of hepatocytes and hepatic stellate cells, and solitary hepatic stellate cells themselves: stellate cells, perisinusoidal cells, Ito cells were observed in the perisinusoidal spaces of Disse. These cells are small-sized, their differentiation was carried out by heterogeneous, cleared cytoplasm with vacuole-like inclusions of fat located in the perinuclear space.

Central veins were located in the center of classical hepatic lobules. Lumens of central veins in intact rats were moderately plethoric. Endothelial layer in the walls of the central veins was solid. Endotheliocytes were homogeneous in structure, which was characteristic for these cells. Shape of hepatocytes in classical hepatic lobules of intact rats was polygonal. Cytoplasm of most cells is cleared and uniformly colored. Hepatocytes are heteromorphic, and were found to be mostly uniform in their shape, intensity of cytoplasm coloring, and size within a specific region of the lobule. Small elongated hepatocytes predominated in centrolobular regions, large hepatocytes of mostly polygonal or rounded shape were found in the regions surrounding portal tracts. A comparative examination of the histological preparation of the rat liver in subcapsular region and in the depth of the hepatic parenchyma revealed differences in the morphology of these regions. In particular, hepatocytes in subcapsular region are characterized by pronounced morphological heterogeneity without zonal regularity, compared to the regions in the depth of the parenchyma. In addition, hepatocytes with cleared cytoplasm, rounded and giant cells with a diameter of up to 30-35 μm are more common in subcapsular region **(fig. 3).**

The diameter of sinusoidal capillaries of subcapsular region in intact rats was (13.40±0.30) μm, while the diameter of sinusoidal capillaries in the depth of the parenchyma was (10.0±0.20) μm.

Solitary binuclear and multinuclear hepatocytes were observed throughout the hepatic parenchyma. Solitary cells with apoptotic changes or necrotized cells were detected in centrolobular region of hepatic lobules, sometimes there were cells with signs of adipose degeneration.

Conclusions.

1. Normal weight of the rat liver is 11-16 g in mature animals, and 6-8 g in immature animals. Rats have no gallbladder.

2. There are 6 lobes in the rat liver: right lateral, left lateral, right central, left central, caudate lobe and additional lobe.

3. In rats, margins of the hepatic lobules are visualized only in the direction of the hepatic tubules, and are characterized by regions with randomly located hepatocytes and porto-portal shunts.

4. Hepatocytes are heteromorphic depending on the hepatic lobe and the region, and are found to be mostly uniform in their shape, intensity of cytoplasm coloring, and size within a specific region of the lobule.

5. The diameter of sinusoidal capillaries was 9.6 (8.51; 11.33) μm, ratio of the sectional area of sinusoidal capillaries to the sectional area of the parenchyma was 0.20±0.02.

Prospects for further research. Materials of our research will serve a morphological basis for further experimental studies, which will be conducted on experimental animals (white rats).

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МАКРО- ТА МІКРОСТРУКТУРНА ОРГАНІЗАЦІЯ ПЕЧІНКИ БІЛОГО ЩУРА В НОРМІ

Шевчук М. М.

Резюме. Вага печінки щурів у нормі становить: у статевозрілих тварин 11-16 г та 6-8 г у статевонезрілих. Складається з 6 часток: права бокова, ліва бокова, права центральна, ліва центральна, хвостова частка та додаткова частка. Печінка щура зовні покрита сполучно-тканинною капсулою, яка щільно зрощена з вісцеральним листком очеревини. Зв'язковий апарат слабо виражений. На вісцеральній поверхні розташовані ворота печінки із воротною веною, власною печінковою артерією та печінковими протоками, що формують спільну жовчну протоку. Жовчний міхур у щурів відсутній. Внутрішня будова печінки утворена паренхімою та стромою. Паренхіма печінки представлена печінковими часточками. Печінкові часточки полігональної 5-6-тигранної форми та складаються з печінкових балок і міжчасточкових синусоїдних капілярів. В центрі часточки знаходиться центральна вена. Навколо строми елементів тріад визначають невеликий простір – простір Мола. Поміж класичними печінковими часточками визначаються прошарки пухкої сполучної тканини, в яких розташовуються компоненти печінкової тріади – гілки печінкових артерії, воротної вени, лімфатичні судини та жовчні протоки. У клітинному складі пухкої сполучної тканини печінкових тріад розташовувались фібробласти, а також поодинокі лімфоцити та макрофаги. Печінкові балки утворені гепатоцитами та розташовані в радіальному напрямку. Гепатоцити за формою – багатогранні, в балках розташовуються двома рядами і пов'язані між собою десмосомами, гетероморфні в залежності від частки печінки та зони, при чому в межах конкретної зони часточки виявляли гепатоцити переважно однорідні за формою, інтенсивністю забарвлення цитоплазми та розміром. Міжчасточкові сполучнотканинні трабекули розвинені слабо. Межі класичних печінкових часточок визначалися через розташування портальних тріад, печінкових пластинок та синусоїдних капілярів, які мали радіальний напрямок, від центральної вени до портальних трактів.

Ключові слова: щур, печінка, гепатоцит.

MACRO- AND MICROSTRUCTURAL LIVER ARRANGEMENT IN WHITE RATS IN HEALTH

Shevchuk M. M.

Abstract. Normal weight of the rat liver is 11-16 g in mature animals, and 6-8 g in immature animals. It consists of 6 lobes: right lateral, left lateral, right central, left central, caudate lobe and additional lobe. Rat liver is externally covered with connective tissue capsule, which is tightly fused with the visceral layer of the peritoneum. The ligament apparatus is poorly pronounced. On the visceral surface, there is a porta hepatis with portal vein, own hepatic artery and hepatic ducts forming the common bile duct. Rats have no gallbladder. The internal structure of the liver is formed by parenchyma and stroma. The liver parenchyma is represented by hepatic lobules. Hepatic lobules have polygonal 5-6-sided shape, and consist of hepatic tubules and interlobular sinusoidal capillaries. The center of the lobule is the central vein. A small space called the space of Mall is found around the stromal elements of triads. Between the classical hepatic lobules, layers of loose connective tissue are found, in which the components of the hepatic triad are located, such as branches of hepatic artery, portal vein, lymphatic vessels and bile ducts. Fibroblasts, as well as solitary lymphocytes and macrophages are located in the cellular composition of loose connective tissue of the hepatic triads. Hepatic tubules are formed by hepatocytes, and are located in the radial direction. Hepatocytes are polygonal in shape, they are located in tubules in two rows, and are connected to each other by desmosomes. Hepatocytes are heteromorphic depending on the hepatic lobe and the region, and are found to be mostly uniform in their shape, intensity of cytoplasm coloring, and size within a specific region of the lobule. Interlobular connective tissue trabeculae are poorly developed. Margins of classical hepatic lobes were determined by the location of portal triads, hepatic plates and sinusoidal capillaries, which had a radial direction, from the central vein to the portal tracts.

Key words: rat, liver, hepatocyte.

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