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STRUCTURAL FEATURES OF THE NORMAL MAJOR SALIVARY GLANDS STROMA OF RATS

The aim of the study was to determine the stroma structural features in the major salivary glands of rats in normal state. The study was carried out on 20 adult white male Wistar rats with body weight 260-300 grams, aged 10-12 months. Histological examination of the parotid stroma determined that the intralobular connective tissue was represented by amorphous substance, collagen fibers and fibroblast processes between adjacent terminal parts. The fibroblast bodies were located in the nodular interstitial compartments – the contact points of 3-4 terminal parts of the gland lobules as well as collagen fibers and blood vessels of the hemomicrocirculatory bed – capillaries and postcapillaries. Postcapillaries and venules were revealed in periductal connective tissue. The local protective barrier of the major salivary glands stroma in rats is predominantly represented by plasmocytes and macrophages in the interacinar interstitium as well as macrophages and mast cells in the periductal connective tissue.

Key words: salivary glands, stroma, rats.

Nowadays, various laboratory animals from protozoa to mammals are widely used for conducting experimental medical and biological studies [5, 7]. Correct data interpretation of the experimental morphological studies involving laboratory animals, rats in particular, requires the knowledge of possible specific features of their anatomical structure [12]. A number of experimental studies with rats were carried out to determine the structural and functional changes in salivary glands caused by the effect of various factors [3, 4, 6, 9, 10]. The relevance of mentioned studies is explained by the prevalence of salivary gland diseases, which account for 11.7% of all surgical dental diseases; sialadenosis and sialolithiasis are diagnosed more often [2]. The profound knowledge of the histological structure of the normal major salivary glands in rats includes the thorough investigation of the morphofunctional features of their stroma. Connective tissue is involved in stroma formation; polyfunctional nature of connective tissue is determined by the complexity of composition and structure. Connective tissues perform trophic, protective, supporting, plastic, morphogenetic functions. The trophic function is associated with nutritional regulation of various tissue structures, participation in metabolism and maintenance of homeostasis in the internal environment of the body. The protective function provides prevention of body damage from non-physiological mechanical effects (physical protection) and foreign substances neutralization (phagocytic activity of macrophages and immunocompetent cells involved in cellular and humoral immunity responses). Supporting (biomechanical) function is provided primarily by collagen and elastic fibers, which form the fibrous base of all organs, composition and physicochemical properties of the intercellular substance of skeletal tissues (mineralization). The plastic function of connective tissue provides adaptation to the changeable conditions, regeneration, participation in the replacement of organ defects in case of their damage [1].

Research porpoise - to determine the stroma structural features in the major salivary glands of rats in normal state.

Material and methods. The study was carried out on 20 adult white male Wistar rats with body weight 260-300 grams, aged 10-12 months. The animals were kept under standard laboratory conditions and received usual ration of the vivarium. Animals were euthanized with intraperitoneal introduction of sodium thiopental at a dose of 50 mg / kg body weight. The study complies with the requirements of the international principles of the "European Convention for the Protection of Vertebrate Animals used for Experiment and Other Scientific Purposes" (Strasbourg, 1985), the corresponding Law of Ukraine "On the Protection of Animals against Cruelty" (No. 3446-IV as of 21.02.2006, Kyiv), the Ethical Code of Ukrainian Physician and the Code of Ethics for a Scientist in Ukraine. The major salivary glands were immersed in cold 2.5% glutaraldehyde solution in phosphate buffer at pH 7.4 and stored at refrigerator temperature +4° C for 24 hours to obtain semithin sections. Parotid, submandibular and sublingual glands were embedded in Epon-812 according to the generally accepted technique, then material was placed in gelatin capsules and filled with resin, followed by polymerization at temperatures (35, 45, 60)° C each for 24 hours [8]. Semithin sections were obtained applying ultramicrotome UMTP-7. Staining of the sections was carried out with 0.1% solution of toluidine blue in phosphate buffer at pH 7.8 and polychrome dye.
After staining sections were enclosed in polystyrene under cover glasses and after polymerization were examined applying the light microscope.

**Results and their discussion.** Histological examination of the parotid stroma determined that the intralobular connective tissue was represented by amorphous substance, collagen fibers and fibroblast processes between adjacent terminal parts. The fibroblast bodies were located in the nodular interstitial compartments – the contact points of 3-4 terminal parts of the gland lobules as well as collagen fibers and blood vessels of the hemomicrocirculatory bed – capillaries and postcapillaries. Postcapillaries and venules were revealed in periductal connective tissue. Erythrocytes were visualized in the lumens of capillaries and postcapillaries; lumens of venules did not contain the formed elements of blood.

Mastocytes, mainly with eccentric localization of nucleus, with heparin containing granules were detected in the interstitial connective tissue around the terminal parts of the parotid salivary gland lobules. Mastocytes with the central nucleus location prevailed in the periductal connective tissue, which morphologically determined the presence of histamine secretory granules in their composition. The largest number of such mast cells was observed around the striated ducts (Fig. 1). The prevalence of mastocytes in the periductal connective tissue with the central nucleus location indicated their active regulation of the vascular wall permeability of postcapillaries and venules in state of nutrition dormancy.

The intralobular connective tissue of the submandibular glands of rats was represented by collagen fibers and fibroblasts. The vessels of the microcirculatory bed were determined in interstitium; macrophages, plasmocytes and mastocytes were located perivascularly. The study of the structure of hemomicrocirculatory bed vessels revealed that the arteriolar walls consisted of three layers; inner layer of endotheliocytes was located on the basement membrane. The inner elastic membrane separated the middle layer of smooth muscle cells. Adventitial cells were located externally. Precapillaries provided the blood supply to lobules parenchyma. Capillaries of the submandibular gland of rats provided blood supply of the terminal parts. The capillary wall of submandibular gland lobules was formed by non-fenestrated type of endotheliocytes, surrounded by continuous basal membrane and discontinuous layer of pericytes. Postcapillaries in the submandibular gland lobules of rats were localized in the periductal connective tissue of the inner area of "nodular" interstitial compartments around the inserted and striated ducts. Transverse sections presented the postcapillary lumen surrounded by 2-3 fenestrated endotheliocytes with thin cytoplasm. Continuous basal membrane separated the endothelial layer from perivascular fibroblasts. Mastocytes in degranulation state were determined along with revealed vessels.

Venules in the composition of the submandibular gland lobules were detected in connective tissue around the striated and intralobular secretory ducts. Their wall was characteristic of this microvessel structure type; formed elements of blood were evident in the lumens (Fig. 2).

In the sublingual salivary glands of rats the hemomicrocirculatory bed vessels were also detected in the connective tissue layers. The inner layer formed by endotheliocytes on the basal membrane was determined in the arteriolar wall at high magnification, visualizing as thin weakly basophilic band. The nuclei of spindle-shaped cells had distinct localization along the vessel (Fig. 3). Smooth myocytes formed continuous layer in the middle area. Considering the nuclei shape on the slices, the myocytes had the circular orientation. The outer layer was formed by loose connective tissue. The precapillary arterioles system was located both periductal and in the "nodal" interstitial compartments of lobules. Capillary vessels of sublingual gland were located periacinary and around the inserted and striated ducts. The capillary wall was formed by endotheliocytes of the flattened form of non-fenestrated type and basal membrane.
The pericyte layer was discontinuous. Capacitive microvessels – postcapillaries and venules were identified around the striated ducts. Their wall was formed by endotheliocytes on the basal membrane and adventitial cells; perivascular fibroblasts were localized externally. Macrophages, lymphocytes and plasmocytes were detected among the migrant cells of the interstitial connective tissue in the sublingual salivary glands of rats. Plasmocytes, macrophages and mast cells were constantly observed in the periductal connective tissue. Connective tissue included single mast cells around the striated and intralobular collecting ducts. The identified structural and functional features of the major salivary glands stroma in rats should be considered in experimental studies, since their neglection can lead to incorrect data interpretation.

Conclusions

1. The major salivary glands stroma of rats is represented by amorphous substance, collagen fibers and fibroblast processes between adjacent terminal parts of the lobules; fibroblast bodies are located in the nodular interstitial compartments – the contact points of 3–4 terminal parts.

2. Capillaries and post capillaries prevail around the terminal parts of the lobules among the hemocirculatory bed vessels; postcapillaries and venules are mostly observed in the periductal connective tissue.

3. The local protective barrier of the major salivary glands stroma in rats is predominantly represented by plasmocytes and macrophages in the interacinar interstitium as well as macrophages and mast cells in the periductal connective tissue.

Prospects for further research include the quantitative analysis of the immunocompetent cells number in the major salivary glands stroma of rats.

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Реферати

**СТРУКТУРНІ ОСОБЛІВОСТІ СТРОМИ ВЕЛИКИХ СЛЮННИХ ЗАЛОЗ У НОРМІ**

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Метою роботи стало встановлення особливостей структурної організації строми великих слюнних залоз щурів у нормі. Дослідження було виконано на 20
Notwithstanding the widespread use of opioid analgesics in medical practice and the increased incidence of drug addiction, especially among teenagers, the issue of the influence of opioids on the immune organs remains relevant.

52 white male rats of reproductive age with an initial body weight of 140–150 g were involved in study. Nalbuphine was administered intramuscularly once daily at 10:00 –11:00 hours during 42 days, increasing the dose every 7 days. The material was collected in accordance with conventional technique.

After one week of the introduction of Nalbuphine a moderate thickening of the basal membrane and enlargement of the lumen of blood capillaries was observed. Within the two weeks the lumen of arteries, arterioles and venules was slightly dilated, the nuclei of the endotheliocytes were slightly enlarged, occupying a significant portion of the cytoplasm; invaginations were formed by the karyolemma. After three to four weeks the veins and venule vascular wall were enlarged, the basal membrane was damaged, deformed red blood cells were found in the lumen of the hemocapillaries, arranged in the “coin column”, often attached to the luminal surface of the endotheliocytes. The wall of arteries and arterioles were thickened due to the edema of endotheliocytes and initial signs of sclerosis. After five to six weeks the perivascular edemas and hemorrhages into the parenchyma thymus was detected. The vast majority of vessels were “empty”. One week after the drug was discontinued, no irreversible changes were found.

Key words: nalbuphine, rat, vein, artery, blood capillary, endothelial cell, pericyte

The research is a part of complex topics, entitled “Features of the structural organization of the lymphoid organs and vascular bed in ontogenesis in the norm and the regularities of their reorganization under the action of antigens, chemical and physical factors on the organism” – the State registration number 0115U003903 and “Structure of organs and their bloodstream in ontogenesis, under the influence of laser irradiation and pharmaceuticals, with violations of blood supply, reconstructive operations and diabetes” – the State registration number 0110U001854.

For a long time opioid analgesics have been used for therapeutic purposes in medical practice [6, 9, 10]. Recently, experimental studies have been conducted on the impact of these drugs on certain organs and tissues (eyeball, tongue, cerebellum, ulcer, pancreas, skin) [1, 3, 5, 7, 8]. However, in the scientific literature, no data on the effect of narcotic analgesics on the organs of the immune system have been found to date. The primary immune organs include the thymus, where antigen-independent proliferation and differentiation of subpopulations of T-lymphocytes occurs. From thymus T-lymphocytes enter into the vascular bed, spreading to T-dependent zones of the secondary immune organs [4]. The latter provide an adequate response of the body to the penetration of foreign antigens. Therefore, the study of the effect of opioids on the structure of the thymus is the relevant medical issue.

The paper is aimed at the study of the features of submicroscopic changes of the components of the thymus vascular bed of white male rats of reproductive age in the dynamics of six-week exposure to opioid nalbuphine and within one week after its discontinuation.

Material and methods. 52 white male rats of reproductive age with initial weight of 140-150 g were involved into study. Nalbuphine was administered intramuscularly once daily at the same period of

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NALBUPHINE-INDUCED SUBMICROSCOPIC CHANGES IN THE COMPONENTS OF THE THYMUS VASCULAR BED
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