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# COMPARATIVE MORPHOLOGICAL CHARACTERISTICS OF THE PROSTATE IN JUVENILE RATS AND RATS WITH CHRONIC ALCOHOLISM

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Article history:		Abstract:
Received: Accepted: Published:	March 10 <sup>th</sup> 2023 April 14 <sup>th</sup> 2023 May 20 <sup>th</sup> 2023	The article presents the results of a study on the morphological features of the prostate of juvenile rats and structural changes in rats with chronic alcoholism. In rats with chronic alcoholism, compared with the control, there is polymorphism of the terminal sections of the glands, an increase in the volume fraction of acini that do not contain a secret, and a decrease in the volume fraction of the glandular parenchyma in the structure of the organ. The experiment revealed acini with foci of epithelial stratification, vascular changes, mild and moderate lymphocytic infiltration, expansion of connective tissue layers, proliferation of fibrous connective tissue and compaction of bundles of collagen fibers.

**Keywords:** rat prostate, gland, morphometry, chronic alcoholism

#### INTRODUCTION

Chronic alcohol consumption is widespread in modern society. Alcohol is a dangerous toxic effect on most organs and systems. Alcohol abuse is the world's third risk factor for diseases and disability, and is the cause of 60 types of diseases [13].

The toxic effect of alcohol has been proven in the development of pathologies of the liver, hollow organs of the gastrointestinal tract, mucous membranes, cardiovascular and genitourinary systems, which, depending on its concentration, leads to the development of dystrophic and destructive changes [1,2,4,5,6,8,9,10,11,12].

It should be noted that the morphology of the prostate in alcoholism is practically not studied. In this regard, the study of the morphological features of the organ in conditions of chronic alcoholism is of undoubted interest for theoretical and practical medicine.

#### **MATERIALS AND METHODS**

The study was performed on 24 outbred white male rats at the age of 3 months. 2 experimental groups were formed: 1st - control (n=12); 2nd - experimental group (n=12).

In the experimental group, for modeling chronic alcoholism, forced alcoholization of animals was used using a 40.0% ethanol solution [7]. The solution was administered intragastrically using a metal probe 1 time per day at a total dose of 7 g/kg of body weight for 1 month before the study age. Control animals received intragastrically equal volumes of 0.9% NaCl solution. Rats were sacrificed by instantaneous decapitation under ether anesthesia, according to approved rules [3].

For histological examination, pieces of the prostate were fixed in 10% buffered formalin and embedded in paraffin according to the standard method. Histological sections obtained from paraffin blocks, 5-7  $\mu$ m thick, were stained with hematoxylin and eosin for review purposes, collagen fibers were detected by van Gieson staining.

With a microscope magnification of 70 times (7x10), the sections were determined:

- the shape of the lumen of the glands, the number of terminal sections of the glands in the field of view, the volume fraction of acini with and without secretion (in %), the number of acini with desquamated epithelial cells in the field of view, in the intralobular stroma, the number of intraorgan vessels in the field of view was counted.

In the preparations, at a magnification of 280 times (7x40), using an eyepiece micrometer, the diameter of the lumen of the glands, the height of the epithelium, the inner diameter and wall thickness of intraorganic vessels were measured. In addition, the thickness of collagen fibers and their distribution in the tissues of the gland were determined.

In the field of view (7x40), the presence and severity of lymphocytic infiltration in the tissues of the gland was assessed. When distributing lymphocytes by severity (cell density), the classification of the North American Chronic Prostatitis Collaborative Research Network and the International Prostatitis Collaborative Network was used



1) mild degree - single lymphocytic cells separated by distinct intermediate zones;

2) moderate degree - confluent fields of lymphocytic cells without tissue destruction and / or lymphoid nodular / follicular formation;

3) severe degree - confluent fields of lymphocytic cells with tissue destruction and / or lymphoid nodular / follicular formation.

To assess the severity (fibrosis) of the proliferation of connective tissue using an eyepiece micrometer with a magnification of the objective x40, eyepiece x7 in the field of view, the thickness of the stroma layers between the glands was measured.

The degree of compaction of the connective tissue was determined by the appropriate method (Gorbunova E.N., Davydova D.A., Krupin V.N., 2011) as follows: 1) mild form (increase in the thickness of stromal septa up to 2 times in 2-4 fields of view out of 10); 2) moderate form (the thickness of the stromal septa is increased up to 2 times in more than 4 fields of view or a sharp thickening - more than 3 times and is present in single (1-2) fields of view); 3) pronounced form (stromal septa are enlarged up to 3 times or more in 7-10 fields of view).

Conducted a study of the volume fractions of glandular and stromal elements (in%). To do this, using the morphometric grid G.G. Avtandilov (with the number of intersections 100) using an eyepiece x10, a lens x10 in each preparation of the prostate in 10 fields of view, the number of intersections falling on the stromal and glandular (including the lumen of the gland) elements was counted to determine their ratios.

## **RESULTS AND DISCUSSION**

The study showed that in 3-month-old rats, the prostate has a normal structural plan, consists of numerous separate alveolar-tubular glands and muscular-elastic stroma in the form of loose fibrous connective tissue, bundles of smooth myocytes and vessels.

In survey microscopy, the terminal secretory sections or acini in most cases (75.0%) have a folded appearance, are represented by highly prismatic epithelium with high columnar and basal cells that rest on a clearly visible basement membrane. The height of the epithelial layer varies from 8.4 to 21.0 µm, on average 16.5±0.6 µm. Acini have oval and rounded shapes (Fig. 1). The diameter of the lumen of the glands ranges from 105.0 to 298.2 microns, on average -  $198.7 \pm 8.0$  microns. The number of acini in the field of view ranges from 38 to 66, averaging 51.8±1.5. The volume fraction of acini with a secret is in the range of 70-100%, on average - 83.5±1.6. The proportion of acini without a secret is 8-30%, on average 16.5±1.2. The number of acini with desguamated epithelial cells in the lumen varies up to 4 per field of view, on average 1.7±0.2.

In the periglandular stroma, single lymphocytes are determined, separated by clear intervals. Their number in the field of view is in the range of 4-9, on average  $5.7\pm0.3$ . The thickness of the stromal septa between the acini ranges from 8.4 to  $25.2 \mu$ m, averaging  $15.5\pm0.9 \mu$ m.

The number of stromal vessels in the field of view is in the range of 3-8, averaging  $5.0\pm0.3$ . The inner diameter of the venules is in the range from 16.8 to 19.4 microns, on average -  $20.6\pm0.67$  microns. The thickness of their wall ranges from 4.2 to 8.4 microns, on average -  $4.7 \pm 0.21$  microns. The diameter of the capillaries varies from 4.2 to 12.6 µm, on average  $8.9\pm0.21$  µm. The wall thickness is in the range of 2.1-4.2 microns, on average -  $4.0\pm0.13$  microns. The inner diameter of arterioles ranges from 8.2 to 16.4 µm, averaging  $13.8\pm0.46$  µm. Their wall thickness varies from 4.2 to 8.4 µm, on average 7.5\pm0.21 µm.



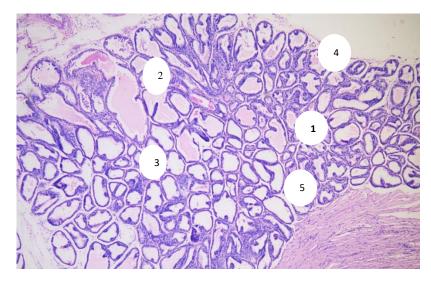


Fig.1. Prostate of a 3-month-old rat. Stained with hematoxylin-eosin. 1-secretory terminal sections (acini), 2intralobular stroma, 3-acinus with intraluminal secretion, 4-gland capsule, 5-bundles of smooth myocytes. OK. 10 x vol. 10.

The volume fraction of glandular tissue is 62-80%, on average  $71.0\pm1.0\%$ . The proportion of stromal tissue ranges from 20-38%, averaging  $29.0\pm1.0\%$ .

Collagen fibers are located in the stroma of the prostate, forming a small-loop network, most of the

fibers lie under the epithelium and surround the prostatic acini (Fig. 2). The thickness of collagen fiber bundles varies from 4.2 to 12.6  $\mu$ m, averaging 8.2±0.46  $\mu$ m.

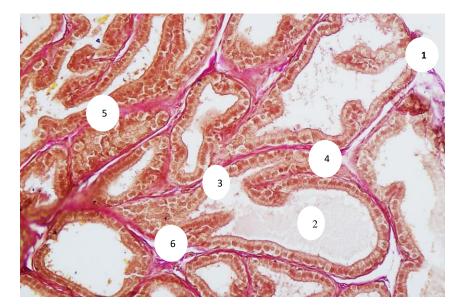


Fig.2. Prostate of a 3-month-old rat. Van Gieson coloring. 1-capsule of the gland, 2-acini, 3-cylindrical epithelium, 4fibrous-muscular stroma, 5-bundles of collagen fibers around the secretory sections, 6-stromal small-loop network formed by collagen fibers. OK. 10 x vol. 20.



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It has been established that in 3-month-old rats of the experimental group, acini in 50% of cases have a folded appearance, are represented by low-prismatic epithelium, and high-prismatic epithelium is determined in some places. The height of the epithelium varies from 4.2 to 16.8 µm, on average, 9.1  $\pm$  0.6 µm. In places in the epithelium, foci of epithelial stratification are defined, known as prostatic intraepithelial neoplasia (PIN). At the same time, the rows of layers in the epithelium are disturbed, cell polymorphism and the presence of large and multiple nucleoli in the nuclei are noted (Fig. 3). Acini have a polygonal shape (Fig. 4). The diameter of the lumen of the glands ranges from 25.2 to 126.0 microns, on

average - 75.2  $\pm$  0.4 microns. The number of acini in the field of view ranges from 54 to 96, averaging 74.2 $\pm$ 2.3. The lumens of the acini are partially filled with a homogeneous secret. The volume fraction of acini with a secret is in the range of 15-46%, on average - 29.2 $\pm$ 1.7. The proportion of acini without a secret is 54-85%, on average 70.8 $\pm$ 1.7. In the lumen of individual acini, there are many fragments of desquamated, desquamated cells, their number varies from a few cells to small cell conglomerates (Fig. 5). In the field of view, the number of acini with desquamated epithelium varies from 4 to 10, on average 7.5 $\pm$ 0.3.

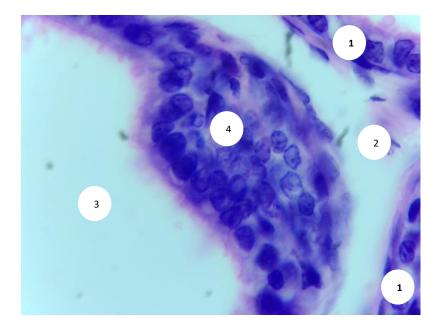


Fig.3. Prostate of a 3-month-old rat of the experimental group. Stained with hematoxylin-eosin. 1-normal epithelial lining of acini, 2-interglandular stroma, 3-acinus lumen, 4-focus of epithelial stratification (PIN). OK. 10 x vol. 40.

In the interacinar stroma, chaotically scattered single lymphocytes are determined in places. In most preparations, merging fields of lymphocytes are visualized; they envelop the end sections of the glands in the form of chains of round or oval shapes. In addition, accumulations of lymphocytes are found inside the lumen of the acini and around the vessels of the interlobular stroma, which infiltrate the walls of the vessels. At the same time, the integrity of the epithelial lining is not broken, tissue destruction and lymphoid nodules are not observed (Fig. 5). The number of lymphocytes in the stroma (in the field of view) ranges from 15 to 40, on average  $26.0\pm 1.4$ . The thickness of the stromal septa ranges from 21.0 to 75.6 µm, averaging  $40.7\pm 2.94$  µm.

The number of stromal vessels in the field of view is in the range of 6-12, averaging  $9.1\pm0.3$ . The

inner diameter of the venules ranges from 16.8 to 37.8  $\mu$ m, on average 25.2±1.13  $\mu$ m. The thickness of their wall ranges from 2.1 to 4.2 microns, on average - 3.57 ± 0.13 microns. The diameter of the capillaries varies from 8.4 to 16.8  $\mu$ m, on average 12.6±0.46  $\mu$ m. The thickness of their wall is in the range of 2.1-4.2 microns, on average - 3.49 ± 0.13 microns. The inner diameter of arterioles ranges from 12.6 to 21.0  $\mu$ m, averaging 17.5±0.46  $\mu$ m. Their wall thickness varies from 4.2 to 8.4  $\mu$ m, on average 5.59±0.21  $\mu$ m.

Morphometry of the parenchymal-stromal ratio showed that the relative area of its parenchyma varies within 32-55%, averaging  $40.6\pm1.24\%$ . The proportion of stromal tissue ranges from 45-68%, averaging  $59.4\pm1.24\%$ .

In the experiment, numerous collagen fibers occupy all interepithelial regions of the stroma. They



are found around the acini and ducts of the gland, where they densely braid the smooth myocytes of the stromal layer. In places, a coarse network of collagen fibers is formed in the interacinar stroma. The thickness of collagen fiber bundles varies from 4.2 to 8.4  $\mu$ m, averaging 6.13±0.21  $\mu$ m.

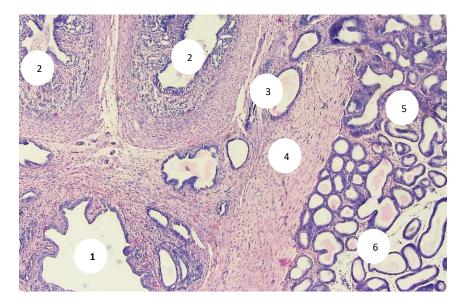


Fig.4. Prostate of a 3-month-old rat of the experimental group. Stained with hematoxylin-eosin. 1-prostatic urethra, 2ejaculatory ducts, 3-glandular ducts, 4-smooth muscle tissue, 5-polygonal secretory terminal sections (acini), 6intralobular stroma. OK. 10 x vol. 10.

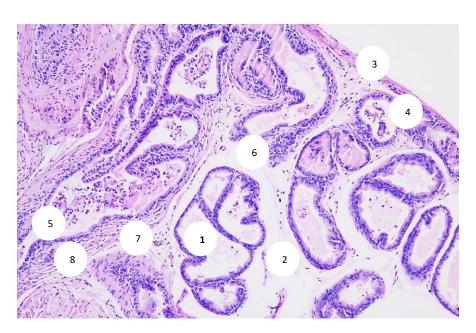


Fig.5. Prostate of a 3-month-old rat of the experimental group. Stained with hematoxylin-eosin. 1-secretory end sections (acini), 2-dilated stromal septa, 3-gland capsule, 4-intraluminal desquamated epithelial cells, 5-lymphocytes inside the lumen of the glands, 6-diffuse accumulation of lymphocytes in the subepithelial layer of the stroma, 7-accumulation of lymphocytes around the stromal vessels, 8- bundles of smooth myocytes. OK. 10 x vol. 20.



# CONCLUSION

The prostate of juvenile rats has the usual structural plan, typical of an organ with complete differentiation of glandular-stromal elements.

In animals with chronic alcoholism, glandular polymorphism, a decrease in the size and folding of acini, an increase in the volume fraction of acini that do not contain a secret, and a decrease in the volume fraction of glandular parenchyma in the organ structure are observed. In places, acini are detected with foci of epithelial stratification, which can subsequently cause a malignant neoplasm.

In the experiment, mild and moderate diffuse focal periductal lymphocytic infiltration, vascular and alterative changes in the form of focal desquamation of varying severity are noted, which indicates accelerated elimination (extinction) of glandular epithelial cells.

The effect of alcohol leads to expansion of connective tissue layers, growth of fibrous connective tissue and thickening of bundles of collagen fibers, which can be considered as a reaction of the body aimed at isolating the focus of damage from surrounding tissues and systemic blood flow.

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