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# New Parameters for Studying Morphometric Indicators of the Splen

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# **Abstract**

The article presents the results of our own morphological, morphometric studies to study the influence of an immunomodulator of nucleic nature on the morphogenesis of peripheral immunocompetent organs (spleen), which further ensure the formation of cellular immunity; in the middle of pregnancy and upon its completion, it ensures an earlier development of structures responsible mainly for the humoral response.

# **Keywords:**

immunocompetent organs, spleen, numerous effects, differentiated functions.

In conditions of sharply increasing physiological loads on the animal's body, if the technology of housing and feeding is violated, especially during pregnancy, disturbances in the processes of formation, development and differentiation of systems that ensure the life activity of newborns may occur. The existing close relationship between the organisms of the mother and fetus allows these processes to be regulated by various pharmacological agents. The numerous effects of drugs of biological origin make it possible to use them at different stages of pregnancy, thus increasing the functional status of the female's body and ensuring the formation of adaptation systems in the postnatal period in the offspring [2, 6, 7].

In addition to their specific action, almost all immunomodulators have a number of identical pharmacological effects - activation of tissue regeneration processes, and also have an adaptogenic and growth-stimulating effect [1, 4, 13]. The mechanism of action of these drugs is based on the formation of a number of substances that carry out the metabolism and differentiated functions of various cells. In this regard, it seems relevant to study aspects of their impact on the structural components of internal organs.

Exposure to components of nucleic acid preparations can ensure proliferation of cellular elements of lymphoid tissue [8-11]. According to some authors [14], compensatory and adaptive processes in the organs of the immune system are manifested by an increase in the volume occupied by functionally active elements. Such transformations of the organs of the lymphoid system are necessary for the

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body during critical periods of development, especially in the early stages of postnatal ontogenesis.

Cells of the lymphoid system, due to their inherent barrier-phagocytic functions, are capable of nonspecifically absorbing macromolecules of exogenous RNA [3, 5]. When they are used in pregnant females, the functional activity of this system may increase in the body, which will have a positive effect on the further development of the structures of the lymphoid system of the offspring.

The spleen appears to be the only peripheral organ of the immune system located on the path of blood flow from the aorta to the portal vein system, which branches in the liver. In the marginal zone of the spleen, the formation of effector lymphocytes for cellular immunity reactions and plasma cells for humoral immunity reactions occurs and their release into the red pulp.

Material and research methods. The experiment used female Wistar rats and their offspring at different periods of postnatal ontogenesis - days 5, 15, 20. Pregnant animals were divided into four groups - three experimental and one control. Animals of the 1st experimental group were administered the nucleic acid drug "Vestin" intramuscularly once on the fourth day after fertilization. Females of the 2nd experimental group were administered the drug "Vestin" on the 14th day after fertilization. In the 3rd experimental group, "Vestin" was used intramuscularly once on the 18th day after fertilization. Females in the control group were injected intramuscularly with saline.

To take organs, rats and their offspring were removed from the experiment by decapitation, observing the "Rules for carrying out work using experimental animals" (M., 1998). In order to clarify the age and organotypic characteristics, histological, histochemical and morphometric studies of the spleen of the offspring of rats from the experimental and control groups were carried out. The taken material was fixed in a 10% solution of neutral formaldehyde, dehydrated and embedded in paraffin according to the generally accepted method. Using a microtome, sections 5-6  $\mu$ m thick were obtained and, after deparaffinization , they were stained with hematoxylin and eosin and Schiff's reagent.

The data obtained during the research were subjected to statistical processing, arithmetic mean values were compared using the Student's P reliability criterion.

Research results. The dynamics of morphological changes in the spleen in the offspring of rats from experimental groups in the early postnatal period of life was aimed at increasing the volume of lymphoid tissue.

At five days of age, the area occupied by the PALM of the spleen in the offspring in all experimental groups was less than that of the control. At this age, the width of the MZ of the spleen - the place on the border of the red and white pulp, where lymphocytes and macrophages enter from the blood and where they first receive information about antigens and then migrate into the white pulp, in rat pups

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of the 1st experimental group exceeded the indicators of other experimental groups. And the offspring of the 2nd and 3rd experimental groups had less than in the control. It can be assumed that slightly more lymphocytes entered the spleen MZ of rat pups of the 1st experimental group, but, as in the offspring of the 3rd experimental group, they were redistributed between the PALM and LN, which by this age had already been formed in the offspring of these groups. According to literary sources, the formation of lymphoid nodules in rats is observed only after 10 days of development. Their appearance in rat pups of the 1st and 3rd experimental groups in the early period of observation may be a reflection of earlier stimulation of the humoral immune system.

At fifteen days of age, the area of lymphoid structures in the spleen naturally increased in the offspring of all experimental groups. However, this increase occurred unequally in different structures of the white pulp of the spleen in analogues of all groups. In rat pups of the 1st experimental group, hyperplasia of the splenic PALM (T-zone) was observed with a decrease in the area of the lymph nodes and the width of the MH; in the 2nd experimental group, with a decrease in the PALM zone, the area of the lymph nodes (B-zone) and the MH increased; in the spleen of the offspring of the 3rd experimental group, the area of the PALM and LN increased with a simultaneous decrease in the size of the MH. In the control group of fifteen- day- old rat pups, an increase in the area of the lymph nodes was noted. At the same time, in the offspring in the control group, the PALM value remained significantly larger in comparison with the analogues of the experimental groups, while the width of the spleen MH decreased. The identified structural transformations occurred with a simultaneous increase in the relative mass of the organ in the offspring of all experimental groups compared to the control. Such changes may indicate the activity of the processes of proliferation and differentiation of cells entering the MH, and their transition to the structures of the white pulp, with subsequent activation of factors of both cellular and humoral immunity in the offspring of the experimental groups, which is typical for this age. The trend we identified can most likely be associated with the characteristics of the age period of the rat pups, in which their bodies experienced the effects of new substances supplied with food, as well as with a certain stimulation of the body of the experimental groups by biological factors. Our data are consistent with the results of studies by some authors [11, 15] on the effect of immunomodulators on the structural components of the white pulp of the spleen.

At twenty days of age, the offspring of the experimental groups experienced a decrease in the area of the PALM. Particularly significant was the decrease in this indicator in the spleen of rat pups in the 1st experimental and control groups, simultaneously with which there was an increase in the size of the lymph nodes. In the offspring of the 2nd and 3rd experimental groups, this trend towards a decrease in PALM was insignificant, but there was a decrease in the area of the LN. Changes in the size of the MH in rat pups of the 2nd and 3rd experimental groups compared to control analogues may indicate a slight slowdown in the transition of cells from the PALM and LN to the bloodstream.

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During this age period, the formation of secondary nodules with reproductive centers occurred in the rat pups of the 3rd experimental group. This corresponds to a high degree of differentiation of lymphoid tissue. Breeding centers perform different functions. They carry out the proliferation of cell clones that specifically respond to antigenic stimulation; plasma cell precursors are formed. The presence of a large number of blast forms of cells in the lymphoid nodules with reproduction centers also indicates high lymphocytopoietic potency [5, 12, 16]. The weight of the organ at this age in the offspring of the 1st and 3rd experimental groups was slightly lower than in the control, which could be associated with a decrease in the development of hyperplastic processes in the spleen of animals of these groups.

## **Conclusions**

The identified age-related changes in the spleen in all groups of offspring were natural. But when the test drug was administered to female rats in the early stages of gestation, the development of structures that subsequently ensured the formation of cellular immunity was predominant. The administration of "Vestin" in the middle of pregnancy and upon its completion ensures an earlier development of structures primarily responsible for the humoral response.

It should be noted that in the offspring of all experimental groups and in the control, at the age of five days, the development of zones responsible for the formation of cellular immunity factors was predominant, which is typical for the early period of postnatal development and, apparently, is associated with colostrum and milk feeding of rat pups.

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