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MORPHOLOGICAL FEATURES OF MESENTERIAL LYMPH NODES IN EXPERIMENTAL HYPOTHYROIDIS IN THE MOTHER

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ABSTRACT

According to statistics, half of the world's population has health problems related to the thyroid gland.

The effect of experimental hypothyroidism in female rats on the development, formation and growth of mesenteric lymph nodes (MLN) of the offspring was studied.

Hypothyroidism in female rats was induced by oral administration of the antithyroid drug Mercazolil at a dose of 0.5 mg per 100 g of body weight for 21 days. After establishing a stable decrease in the concentration of free thyroxine in the blood serum, the females were fertilized by healthy males. During pregnancy and lactation, females continued to be administered a maintenance dose of the drug.

At 3, 7, 14, 21 and 30 days after birth, the development and formation of MLN offspring obtained from females with experimental hypothyroidism were morphologically studied.

It has been established that in offspring born under conditions of maternal hypothyroidism, the time parameters for the formation of structural and functional zones of MLN, especially its T-dependent zones, significantly lag behind the control.

Key words: hypothyroidism in pregnant women, offspring, postnatal ontogenesis, mesenteric lymph nodes.

INTRODUCTION

The close relationship between the immune and endocrine systems has been shown in a number of works [1,2,10]. It has been established that various lesions of the endocrine organs are accompanied by certain immune changes in the body, and, in turn, certain immune disorders lead to disturbances in the endocrine

functions of the body. In this regard, the relationship between the thyroid gland and the immune system deserves special attention. Experimental studies have confirmed that thyroid hormones T4 and T3, as well as thyroid-stimulating hormone, have a modulating effect on immunocompetent cells, including macrophages, dendritic cells and subpopulations of T and B lymphocytes [4,6,7]. Clinical observations indicate that with diseases of the thyroid gland, various immune disorders develop in the body of patients, the nature and severity of which largely depends on the level of thyroid hormones [9].

Among thyroid dysfunctions, a special place is occupied by the so-called “maternal hypothyroidism” (pregnant hypothyroidism or gestational hypothyroidism), which is observed in 2-2.5% of all pregnant women and can manifest itself in subclinical or manifest forms [12, 14]. The relevance of the problem of hypothyroidism in pregnant women in the practice of doctors of various specialties is undoubted, since with a deficiency of thyroid hormones, necessary for the normal development and functioning of almost every cell of the human body, severe changes develop in all organs and systems without exception [5, 6].

An analysis of the literature showed that the main attention of researchers is paid to the study of the nervous system of offspring who were born from mothers with pregnant hypothyroidism. Numerous clinical observations show that children born from mothers with hypothyroidism have reduced cognitive function of the brain and indices of intellectual abilities were also lower compared to their peers born from healthy mothers [11,16]. There are only a few studies indicating the unfavorable effect of experimental hypothyroidism in pregnant female rats and on some immunological and hematological parameters of the offspring. The authors note that hypothyroidism induced by methimazole and propylthiouracil caused a decrease in body weight, as well as in the weight of the thymus and spleen [3,8,13,15]. A disproportion was observed in the subpopulations of T and B lymphocytes: a decrease in the number of B cells was noted with a relative increase in the number of inactive T lymphocytes. At the same time, the structural and functional mechanisms of postnatal growth and development of immune system organs remain practically unclear. The extreme urgency of the problem of hypothyroidism in pregnant women, the uncertainty and insufficient knowledge of its negative consequences on the immune system of the offspring determine the high degree of relevance of further research in this direction.

The purpose of the study was to identify the structural and functional features of the postnatal development of mesenteric lymph nodes (MLN) of

offspring obtained under conditions of experimental hypothyroidism in pregnant women in the mother.

Material and methods. The experiments were carried out on 50 white outbred sexually mature nulliparous female rats weighing 160-180 g, as well as 369 (222 - control group, 147 - experimental group) rat pups obtained from them. All animals were kept under standard vivarium conditions with the same diet. The study was carried out in two stages. At the first stage, after excluding somatic and infectious diseases, all female rats were divided into two groups: females of the 1st group (experimental - 25) were administered the antithyroid drug Mercazolil at a dose of 0.5 mg per 100 g of body weight daily for 21 days and experimental hypothyroidism was induced. The 2nd group of females (intact – 25) received an equal volume of physiological solution. Animals of both groups were under observation. It should be noted that 2 weeks after the administration of Mercazolil, the majority of female rats of the 1st group showed a slight decrease in motor activity and appetite, drowsiness, and dulling of the coat. In order to determine the functional state of the thyroid gland, on the 15th and 22nd days of the experiment, the concentration of thyroid-stimulating hormone (TSH) and free thyroxine (T4) was determined in the blood serum of 10 animals from each group. After establishing a persistent decrease in the concentration of free T4, we moved on to the second stage of the experiment: females from both groups were paired with healthy males for fertilization.The onset of pregnancy was monitored by detecting sperm in vaginal smears. After pregnancy, the females were separated from the males and placed in separate cages for further research. During periods of pregnancy and feeding of cubs, females continued to be administered a maintenance dose of the drug at the rate of 0.25 mg per 100 g of weight, i.e. In the rats of the experimental group, the periods of fertilization, pregnancy and lactation took place under conditions of mercazolil-induced hypothyroidism.

147 rat pups were born from females with experimental hypothyroidism, and 222 from intact females. Within 7 days after birth, 6 (2.7%) of 222 rat pups in the control group and 11 (7.5%) of 147 rat pups in the experimental group died

After birth, litters of pups were kept with their mother rats for one month before being transferred to other cages. Rats born from mothers with experimental hypothyroidism made up the experimental group (141 rat pups), and rat pups born from intact rats made up the control group (211 rat pups).

The study used general morphological, morphometric and statistical methods.

3-, 7-, 14-, 21- and 30-day-old experimental and control rat pups were sacrificed in the morning, on an empty stomach, by decapitation under light ether anesthesia. For morphological studies, MDR were fixed in Bouin's solution, then

pieces of the organ were dehydrated in alcohols of increasing concentrations and embedded in paraffin. Sections 5-7 μm thick, stained with hematoxylin and eosin, were examined using morphological and morphometric methods.

To carry out morphometric studies, we used the Avtandilov grid and the morphometric computer program Nano Zoomer.

All digital data were processed using the variation statistics method. Calculations and statistical analysis were carried out using a statistical package for Window`s. All data were presented as mean ± standard deviation (SD). The statistical significance of differences between the control and experimental groups was compared using the Student's t test, and P values <0.05 were considered significant

Results and discussion. The results of the study showed that in newborn rat pups, the MDR parenchyma contained only diffuse lymphoid tissue, where the structural and functional zones were not differentiated.

On day 3, sinuses were quite clearly formed under the capsule, and lymphoblasts, medium and small lymphocytes were distinguished among the parenchymal cells.

On the 7th day, along with an increase in the area and volume of the node, the cortex and medulla were detected for the first time. On the 14th day of postnatal development, further differentiation of the lymph node parenchyma and the formation of its stroma occurred. Primary lymphatic follicles with clear edges appeared in the cortex. The cell population of lymphatic follicles consisted mainly of small and medium-sized lymphocytes. Among them there were individual lymphoblasts, prolymphocytes and mitotically dividing cells. By this time, the formation of the brain matter was quite well expressed. The cerebral sinuses had clearly defined boundaries and were covered with coastal cells. In the lumen of the sinuses, individual lymphoblasts and macrophages were found among small and medium-sized lymphocytes. Brain cords mainly contained small and medium-sized lymphocytes, single macrophages and a moderate number of plasma cells.

On the 21st day of postnatal ontogenesis, the predominant development of the cortical substance was noted. In the cortex at this time, it was possible to distinguish both primary and secondary lymphatic follicles, the cortical plateau and the paracortical zone. It should be noted that in individual lymphatic follicles at this time germinal centers were clearly expressed, where lymphoblasts, prolymphocytes, large lymphocytes, single mitotically dividing cells and macrophages were detected. The cortical plateau contained mainly small and medium-sized lymphocytes and reticular cells. The paracortical zone, located between the lymphatic follicles and the medulla, contained medium and significant

numbers of small lymphocytes, postcapillary venules with high endothelial cells. In the lumen of the intermediate cerebral sinuses, single small and medium-sized lymphocytes and reticular cells were found. The brain cords contained medium and large lymphocytes, plasmablasts, plasma and reticular cells, and single macrophages.

Further growth of the organ was accompanied by quantitative and qualitative changes. By the 30th day of postnatal ontogenesis, MLN acquired a structure characteristic of the lymph nodes of adult animals.

Thus, the postnatal development of MLN is a complex, genetically determined process. The critical period for the differentiation of structural and functional thymus-dependent (T) and thymus-independent (B) zones of mesenteric lymph nodes is days 7-14 of postnatal life. It is during this period that significant structural and functional changes occur, associated with the specialization of the organ to participate in immune processes. This period is characterized by differentiation of the cortex and medulla, the formation of lymphatic follicles, and an increase in the relative and absolute content of immunocompetent cells. This period is characterized by differentiation of the cortex and medulla, the formation of lymphatic follicles, and an increase in the relative and absolute content of immunocompetent cells. During this period, the proliferative and migratory activity of immunocompetent cells populating the corresponding compartments of the organ increases. These changes are mainly stabilized by the 3rd week of life of animals, when they switch to definitive nutrition. This period was distinguished by the differentiation of the parenchyma into the cortex and medulla, the formation of all their structural components, and the appearance of intercellular cooperation of macrophages with lymphocytes and plasma cells.

Experimentally induced hypothyroidism in females led to certain disturbances in the process of postnatal growth and the development of MLN in the offspring. A significant decrease in the rate of formation of structural and functional zones of the organ was noted. Thus, if in control rat pups, by 14 days after birth, primary lymphatic follicles could be identified in the MLN, and by 21 days, lymphatic follicles with germinal centers, then in 14-day rat pups of the experimental group, no formed follicles were found. The absolute area of the follicles of the lymph nodes remained significantly smaller during all observation periods compared to the control. Morphometric analysis showed that in conditions of hypothyroidism in the mother in the MDR offspring, the formation of thymus-dependent (cortical plateau and paracortical) zones of the organ suffered the most. The area of T-dependent zones was 25-29% smaller compared to the control, which is likely due to disturbances in the postnatal formation of the thymus. Data obtained in our

laboratory showed that maternal hypothyroidism significantly disrupts the postnatal formation of the thyroid gland and can lead to the development of secondary hypothyroidism in the offspring [2,8]. This, in turn, contributes to disruption of the regulatory activity of the thymus in relation to the processes of proliferation, migration and settlement of T-lymphocytes in the corresponding compartments of the peripheral organs of the immune system, including MLN.

Thus, maternal hypothyroidism had a negative impact on the course of postnatal development and the formation of MLN offspring. There was a pronounced decrease in the rate of growth and formation of structural and functional zones of the organ. These changes are relatively more pronounced in the thymus-dependent areas of the lymph nodes and manifest themselves in the form of a slowdown in their formation. All this can ultimately play a significant role in the pathogenesis of immunodeficiency in infants and children born under conditions of maternal hypothyroidism.

Conclusions:

Maternal hypothyroidism has a negative impact on the process of postnatal development and the formation of MDR lymphoid tissue in the offspring. In offspring born under conditions of maternal hypothyroidism, the time parameters for the formation of structural and functional zones of MDR, especially its T-dependent zones, significantly lag behind the control.

The slowdown in the rate of formation of MLN T-zones is based on an imbalance between the processes of cell proliferation and apoptosis, caused by hypothyroidism and a violation of the regulatory function of the thymus.

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