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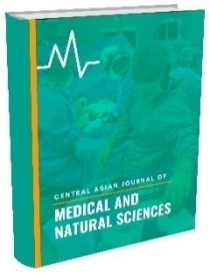

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Morphological Features of Spleen at Different Stages of Postnatal Ontogenesis

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Annotation: The aim of the study was to study the structural features of the morphology of the spleen in white rats at different stages of postnatal ontogenesis.

Materials and methods of research. The experiment was carried out on 65 male mongrel rats in a newborn and at the age of 3, 6 months. In the preparations, the structures of the spleen were examined morphometrically using an ocular micrometer. The diameter of the periarterial lymphatic couplings of the spleen, lymph nodes and their germinal centers, the width of the mantle, marginal and periarterial zones of lymph nodes, the relative area of connective tissue elements and white pulp (relative to the total area of the cut) were measured.

Results. The spleen of newborn rats is functionally immature, lymph nodes are not fully formed, it is difficult to distinguish all zones in them and germinative centers are not determined. By the six-month period, the spleen has the highest immune activity, manifested by an increase in the number of lymph nodes with a reproduction center and a large number of lymphocytes in them and in the periarterial lymphatic couplings.

Conclusions. It has been established that since the age of nine months, involutive changes are observed, characterized by an increase in the number of connective tissue elements in the spleen, a decrease in germinal centers in lymph nodes, the total number of lymphocytes in lymph nodes without reproduction centers, periarterial lymphatic couplings.

Keywords: immune system, spleen, lymphoid follicles, genetically postnatal ontogenesis.

All cellular elements involved in both innate and acquired immunity are subject to age-related remodeling [1]. The spleen is the largest peripheral organ of immunogenesis, which largely determines

the immune status: the state of innate and acquired immunity, its humoral and cellular links, the quality and quantity of lymphoid cells in humans and animals [2]. In the postnatal ontogenesis of mammals, the juvenile, sexually mature and involutional age periods are quite well distinguished [3]. On the 3rd day after birth, foci of myelopoiesis persist, mainly thrombocytopoiesis. From this time on, concentric lymphoid clusters begin to appear around some small arterial vessels - primitive periarterial lymphoid vaginas (PALV), containing mainly medium and small lymphocytes [4,5].

At the age of 7 days, the contours of the PALV in the spleen become clearer, which makes it possible to clearly distinguish the red and white pulp at this time, but the latter still lacks a B-cell compartment, the formation of which lags far behind the development of the T-zones of the PALV [6,7,8].

It is known that by the 10th day of the postnatal period, the outlines of a marginal zone appear in the white pulp, and by the 15th day, single primary lymph nodes are formed [9]. By 30 days after birth, the rat organ demonstrates a definitive microstructure with the presence of secondary lymph nodes with germinative centers [10]. At the suckling age, rats showed signs of intensive growth and morphofunctional development of the spleen, which consisted in a gradual increase in organometric parameters of the organ and morphometric parameters of the white pulp. By the end of the suckling age (21 days after birth), a qualitative transformation of the spleen parenchyma took place in the form of the formation of mature secondary lymph nodes and zones of periarterial lymphatic vaginas, which was a sign of the onset of functional maturity of the immune apparatus of the organ [11].

In adolescence, the size of lymphoid nodules decreases somewhat. At the same time, the size of the breeding centers, on the contrary, increases slightly. Only 10% of lymphoid nodes have such centers. With age, the periarterial lymphoid couplings noticeably expand and lengthen. From 4 to 6 months, the total number of lymph nodes decreases, they persist mainly in the subcapsular zone. At 6-7 months, the number of sinusoids and their sizes per unit area increase in the red pulp. Throughout adulthood, there is a further decrease in the relative number of lymphoid structures [12].

Starting from the period of puberty, immunomodulatory shifts in the spleen are of an ivolytive nature, affecting successively the PALV, LU and marginal zone and are manifested by hypoplasia and delimphatization of the white pulp. Against this background, the relative area of the B-dependent zones of the white pulp increases, which indicates the activation of the migration of recirculating T- and B-lymphocytes. This ensures a certain stability of the immunological status of an organism starting to age [13]. With age, lymphoid nodes with a germinative center are found in isolated cases, their sizes also gradually decrease. The content of connective tissue stroma increases in the spleen, and this process continues in the elderly and, especially, in old age [14]. During the period of pronounced pre-aging and senile changes in animals, there is a weakening of the immune defense of the body due to the involution of the immune system organs.

Thus, the decrease in the overall immune function of the spleen during ontogenesis is largely associated with the suppression of the humoral immune response, that is, with the suppression of B-cell immunity. On the other hand, a certain role is played by a certain weakening of the immune response of the cellular type, due to a reduction in the T-cell pool of spleen lymphocytes as well. In general, the detected changes have an appropriate effect on the state of both B- and T-cell immunity [15].

Materials and methods of research.

The experiment was carried out on 65 male mongrel rats. The rats were kept in the usual conditions of a vivarium. The rats were kept in quarantine for a week, after exclusion of somatic or infectious diseases, they were transferred to the usual vivarium regime. During the experiment, the behavior and physiological state of the animals were monitored. To identify morphological and morphometric

parameters of the spleen structure in postnatal ontogenesis, groups of animals were formed in the newborn and at the age of 3, 6 months.

The spleen and its parts were fixed in 10% neutralized formalin for 10 days, washed with running water for 2-4 hours and dehydrated in increasing concentrations of alcohols and chloroform, paraffin blocks were prepared according to generally accepted methods. Paraffin sections 5-8 microns thick were stained with hematoxylin - eosin and Van Gieson. In the preparations, the structures of the spleen were examined morphometrically using an ocular micrometer. The diameter of the periarterial lymphatic couplings of the spleen, lymph nodes and their germinal centers, the width of the mantle, marginal and periarterial zones of lymph nodes, the relative area of connective tissue elements and white pulp (relative to the total area of the cut) were measured. Measurements were carried out in five fields of view of each histological section. The fields of view were chosen randomly.

To study the cytoarchitectonics of the lymphoid structures of the spleen, the number of cells was calculated by oil immersion with a magnification of 10x100 on a NOVEL Model NLCD-307 microscope (China). The number of cells was calculated using a morphometric grid mounted on the eyepiece of a microscope. Per unit cross-sectional area, the total number of lymphocytes in lymph nodes without a reproduction center and periarterial lymphatic couplings were determined.

The results of research

Studies have shown that the mass of newborn animals ranges from 4.4g to 5.8g, on average 5.18 ± 0.15 g. The absolute mass of the organ is 0.02- 0.04 g, on average 0.032 ± 0.002 g. The mass index ranges from 0.545% to 0.674%, on average 0.617 ± 0.014%.

The length of the spleen of newborn baby rats ranges from 5.4 mm to 8.4 mm, on average - 7.2 ± 0.32 mm. The width of the spleen is in the range of 1.4-2.9 mm, on average - 2.12 ± 0.16 mm. The thickness of the spleen varied from 0.8 mm to 1.8 mm, on average - 1.2 ± 0.11 mm.

The relative area of the white pulp ranges from 14.2 -20.1%, on average - 17.16 ± 0.64%. The relative area of connective tissue elements varied from 5.2% to 6.5%, on average - 5.94± 0.16% (to the total area of the spleen section).

PALM diameter ranges from 90.2 microns to 109.2 microns, with an average of 100.2±2.05 microns. The diameter of the LU is 218.4- 252.2 microns, on average - 242.76 ± 3.65 microns. In a third of the total number of nodules, mantle and marginal zones can be distinguished.

The width of the mantle zone is 28.4 microns to 38.3 microns, on average 35.28 ± 1.07 microns. The width of the marginal zone ranges from 56.3 microns to 69.7 microns, on average 64.32 ± 1.45 microns. The width of the periarterial zone is 38.6 microns to 48.4 microns, on average 44.16 ± 1.06 microns.

The white pulp is formed mainly from lymphocytes at various stages of maturation. There are small, medium and large lymphocytes.

In three-month-old white rats, the spleen has already been formed. When examining the spleen of 3-month-old intact rats , the following data were obtained:

The mass of animals of 3 months of age ranges from 90 g to 130 g, on average 114.16 ± 3.68 g. The absolute mass of the organ is 0.3- 0.7 g, on average 0.52 ± 0.037 g. The mass index ranges from 0.333% to 0.551%, on average 0.455 ± 0.024%. The mass of animals compared with newborn baby rats increased by 22.04 times, and the absolute mass of the organ by 16.25 times.

The length of the spleen ranges from 22.4 to 29.2 mm, on average - 26.78 ± 0.63 mm. The growth rate is equal to- 272.0%. The width of the spleen is in the range of 4.6-7.4 mm, on average - 5.94 ± 0.26

mm. The growth rate is 180.2%. The thickness of the spleen varied from 1.8 mm to 3.9 mm, on average - 2.92± 0.19 mm. The growth rate is equal to- 143.3%.

In histopreparations of the spleen of 3-month-old intact rats, a clearer separation of the organ parenchyma into red and white pulp was observed. The relative area of the white pulp ranges from 19.8% to 26.2%, on average - 22.2 ± 0.59%. The relative area of the white pulp increased by 29.4% compared to newborn baby rats. The relative area of connective tissue elements varied from 5.0% to 6.1%, on average - 5.52±0.1% (to the total area of the spleen section)

In the white pulp, periarterial lymphatic couplings (PALM) and lymphoid nodules (LU) can be clearly distinguished. PALM diameter ranges from 122.6 microns to 139.6 microns, with an average of 132.14±1.56 microns. The growth rate is 31.9%. The diameter of lymph nodes increases by 92% compared to newborn baby rats and ranges from 341.8 microns to 486.05 microns, on average 466.05 ± 13.27 microns. LU can be visually divided into primary and secondary, which percentage ratio is 32% and 68%, respectively. In secondary LU, the formed germinative centers are determined. The diameter of the germinal centers ranges from 94.6 microns to 167.8 microns, on average 147.8 ± 6.73 microns.

In most cases, the LU zones are clearly distinguishable. The width of the mantle zone ranges from 39.7 microns to 49.45 microns, with an average of 45.32±0.89 microns. The width of the marginal zone ranges from 70.3 microns to 84.7 microns, with an average of 77.14± 1.32 microns. The width of the periarterial zone ranges from 81.9 microns to 89.4 microns, with an average of 85.04 ± 0.69 microns. The width of the mantle, marginal and periarterial zones increased by 28.45%, 20% and 92.6%, respectively, compared with newborn baby rats.

It was found that the total number of lymphocytes in the LU without breeding centers is 42-53, on average - 47.3 ± 1.01 cells. Lymphoid nodules without reproduction centers contain (per unit area) small lymphocytes - 30-38, on average - 34.0 ± 0.74 cells, medium lymphocytes - 10-12, on average - 11.0 ± 0.18 cells, large lymphocytes - 2-3, on average - 2.3 ± 0.1 cells.

The total number of lymphocytes in the periarterial lymphoid couplings of the white pulp of the spleen is 41-53, on average - 47.2 ± 1.1 cells. Periarterial lymphoid couplings contain (per unit area) small lymphocytes -29-37, on average - 33.0 ± 0.74 cells, medium lymphocytes - 9-11, on average - 10.25 ± 0.18 cells and large lymphocytes -3-4, on average - 3.5 ± 0.1 cells.

The weight of 6-month-old animals ranges from 190 g to 240 g, on average 220.2 ± 5.4 g. The absolute mass of the organ is 0.6- 0.9 g, on average 0.79 ± 0.032 g. The mass index ranges from 0.315% to 0.405%, on average 0.358 ± 0.01%. The mass of animals compared with 3-month-old rats increased by 1.93 times, and the absolute mass of the organ by 1.52 times.

In 6-month-old rats of the control group, the relative area of the white pulp of the spleen ranges from 18.2 to 24.6%, on average -20.54 ± 0.69%. The relative area of the white pulp decreased by 8.1% compared to 3-month-old rats. The relative area of connective tissue elements varied from 5.6% to 6.7%, on average - 6.21 ± 0.12% (to the total area of the spleen section).

PALM diameter ranges from 128.2 microns to 141.6 microns, with an average of 136.22±1.55 microns. The growth rate is 3.1%. The diameter of the lymph nodes ranges from 380.8 microns to 477.05 microns, on average 420.96 ± 10.44 microns. The percentage ratio of primary and secondary LU is 34% and 66%, respectively. The diameter of the germinal centers ranges from 122.4 microns to 147.7 microns, on average 135.08 ± 2.73 microns. The diameter of the LU and germinal centers decreased by 10.7% and 9.42%, respectively, compared with three-month-old rats. The LU of the white pulp has a rounded, oval and elongated shape.

In micropreparations, it is visually possible to distinguish all the zones of LU. The width of the mantle zone ranges from 40.5 microns to 50.4 microns, with an average of 46.56±1.06 microns. The width of the marginal zone ranges from 74.5 microns to 86.2 microns, on average 80.72 ± 1.26 microns. The width of the periarterial zone ranges from 84.9 microns to 94.7 microns, with an average of 89.42± 1.06 microns. (Fig.3.1.6). The width of the mantle, marginal and periarterial zones increased by 2.74%, 4.64% and 5.15%, respectively, compared with 3-month-old rats.

It was found that the total number of lymphocytes in the LU without breeding centers is 52-61, on average - 57.2 ± 0.97 cells. The total number of lymphocytes in the LU without breeding centers increased by 21.0% compared to 3-month-old rats.

Lymphoid nodules without reproduction centers contain (per unit area) small lymphocytes - 37-43, on average - 41.0 ± 0.65 cells, medium lymphocytes - 12-14, on average - 13.0 ± 0.22 cells, large lymphocytes - 3-4, on average - 3.2 ± 0.11 cells.

The total number of lymphocytes in the periarterial lymphoid couplings of the white pulp of the spleen is 53-61, on average - 58.4 ± 0.86 cells. The total number of lymphocytes in the periarterial lymphoid couplings of the white pulp of the spleen increased by 23.7% compared to 3-month-old rats.

Periarterial lymphoid couplings contain (per unit area) small lymphocytes - 38-43, on average - 41.0 ± 0.54 cells, medium lymphocytes - 11-13, on average - 12.0 ± 0.22 cells and large lymphocytes -5-6, on average - 5.4 ± 0.11 cells.

Conclusions. The spleen of newborn baby rats is not functionally immature, lymph nodes are not fully formed, it is difficult to distinguish all zones in them and germinative centers are not determined. By the six-month period, the spleen has the highest immune activity, manifested by an increase in the number of lymph nodes with a reproduction center and a large number of lymphocytes in them and in the periarterial lymphatic couplings. Starting at the age of nine months, involutive changes are observed, characterized by an increase in the number of connective tissue elements in the spleen, a decrease in germinal centers in lymph nodes, the total number of lymphocytes in lymph nodes without reproduction centers, periarterial lymphatic couplings.

Literature

1. Ahrorovna, K. D. (2020). Effect of a genetically modified product on the morphological parameters of the rat's spleen and thymus. European Journal of Molecular and Clinical Medicine, 7(1), 3364-3370.
2. Akhrorovna, K. D. (2021). ANATOMICAL CHARACTERISTICS OF THE RAT SPLEEN DURING THE INTRODUCTION OF A NON-GENETICALLY MODIFIED PRODUCT. Conferencea, 7-8.
3. Akhrorovna, K. D. (2021). MACROANATOMIC CHARACTERISTICS OF THE THYMUS GLAND IN RATS IN EARLY POSTNATAL ONTOGENESIS. Conferencea, 22-23.
4. Ahrorovna, K. D., & Rustamovna, U. R. (2021, August). GENMODIFIZIERTE PRODUKTE SEIN EINFLUSS AUF DIE MORPHOLOGIE von MILZ UND THYMUSDRÜSE BEI VERSUCHTIEREN. In INTERNATIONAL CONFERENCE ON MULTIDISCIPLINARY RESEARCH AND INNOVATIVE TECHNOLOGIES (Vol. 1, pp. 10-13).
5. Ahrorovna, K. D., & Dilshodovich, K. M. (2021). MORPHOGENESIS OF THE THYMUS OF LABORATORY ANIMALS UNDER THE INFLUENCE OF VARIOUS FACTORS. Web of Scientist: International Scientific Research Journal, 2(04), 483-488.

6. Arnon TI., Horton RM., Grigorova IL., Cyster JG. Visualization of splenic marginal zone B-cell shuttling and follicular B-cell egress // Nature.2013. Vol. 493. P. 684-688
7. Balogh P., Horvath G., Szakal A.K. Immunoarhitecture of distinct reticular fi broblastic domains in the white pulp of mouse spleen. J. Histochem. Cytochem. – 2004. – Vol. 52, № 10. – P. 1287–1298
8. Flores T., Forteza J., ArribasA., PirisMA.Splenic follicular lymphoma: clinicopathologic characteristics of a series of 32 cases // Am J Surg Pathol. 2009. Vol. 33(5). P. 730-738
9. Khasanova DA. Current problems of safety of genetically modified foods (literature review), 2020; 5 (45): 20-27
10. Khasanova D.A. Macroanatomy of payer's patches of rat's small intestine under the influence of antiseptic stimulatort dorogov faction 2 on the background of chronic radiation sickness// Тиббиётда янги кун. - 2020. -№1(30). - С.21-24.
11. Khasanova D.A., Teshaev Sh.J. Topografic-anatomical features of lymphoid structures of the small intestine of rats in norm and against the background of chronic radiation diseases // European science review. Medical science. Vienna, Austria. – 2018. Vol. 2. - №9-10. - P. 197-198.
12. Khasanova DA, Teshaev SJ. Effects of genetically modified products on the human body (literature review), 2020; 5(45): 5-19
13. Khasanova, D. A. (2021). MICROSCOPIC STRUCTURE OF THE RAT SPLEEN DURING THE INTRODUCTION OF A GENETICALLY MODIFIED PRODUCT. British Medical Journal, 1(1.2).
14. Khasanova, D. A. (2021). HISTOLOGICAL STRUCTURE OF THE RAT SPLEEN IN EARLY POSTNATAL ONTOGENESIS. Art of Medicine. International Medical Scientific Journal, 1(2).
15. Milićević N.M. TNF receptor-1 is required for the formation of splenic compartments during adult, but not embryonic life / N.M. Milićević [et al.] // J Immunol. – 2011. – Vol. 186, N 3. – P. 1486 - 1494