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Morphometric Characteristics Of The Spleen Of White Rats In Normal And In Chronic Radiation Disease

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ABSTRACT

In an experimental study, the morphofunctional features of the spleen of 6-month-old white rats were studied in normal conditions and in chronic radiation sickness. The study found that in response to the action of a chronic radiation factor in the spleen of white rats, there is a decrease in structural parameters. This is reflected in the morphological parameters of the organ. As a result, the functional activity of the spleen's lymphoid tissue decreases.

KEYWORDS

Immune system, spleen, lymphoid follicles, chronic radiation sickness.

INTRODUCTION

A living organism cannot be imagined without a protective immune system. One of the most dynamic systems of the body is the immune system of humans and animals, which quickly reacts to the effects of external adverse factors. The immune organs create protection against various pathogenic influences [1,3,8].

Among the peripheral organs of the immune system, the largest and most complex organ is

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the spleen, which performs filtration, cleansing, immune, hematopoietic and depositing functions [6,9,11].

At the present time, a disease associated with damage to the immune system is increasing in the world. According to researchers from different countries, an increase in the number of such morbidity is associated with environmental pollution, leading to a violation of the protective and adaptive processes of the body [7,12].

Radiation occupies a special place among external unfavorable factors. Radiation has the most destructive effect on the human body and causes profound changes in all organs and systems [2,4,5,10].

Today, an urgent and important problem in science and health is the study of the state of health of a person exposed to radiation from various sources of ionizing radiation. Considering the above, we have set the task of studying the morphological parameters of the spleen of white rats in normal conditions and in chronic radiation sickness.

PURPOSE OF THE STUDY

To study the morphofunctional features of the lymphoid structures of the spleen in 6-month-old white rats under normal conditions and with chronic radiation sickness.

MATERIAL AND METHODS OF RESEARCH

The study was carried out in 22 outbred white male rats weighing from 90 to 130 g, which were kept under standard vivarium conditions. The rats were divided into 2 groups: control group (n = 10), experimental group (n = 12). Irradiation of rats was carried out using the AGAT P1 apparatus (Baltiets plant Narva, Estonia, 1991 release, operation since 1994, recharge in 2007) with a capacity of 25.006 sGr / min for 20 days at a dose of 0.2 Gr. The total radiation dose for rats up to 90 days of age was 4.0 Gr. All experimental studies on animals were carried out in accordance with the "Rules for work using experimental animals."

The animals were weighed and removed from the experiment at 90 days of age by instant decapitation under ether anesthesia.

The spleen was removed from the abdominal cavity. To carry out the morphological and morphometric study of the study, spleen fragments were fixed in a 10% formalin solution, passed through a battery of alcohols, and embedded in paraffin blocks according to standard techniques. Paraffin sections 5-8 μm thick were stained with hematoxylin-eosin and according to Van Gieson. Sections were examined morphometrically, using an eyepiece micrometer DN-107T / Model NLCD-307B (Novel, China), the diameter of the periarterial lymphatic muffs, lymph nodules and their germinal centers, the width of the mantle, marginal and periarterial zones, the relative area of the white pulp and connective tissue elements were measured. Spleen to the total cut area. Measurements were performed in five fields of view of each histological section. The fields of view were chosen at random.

In order to study the cytoarchitectonics of the lymphoid structures of the spleen, the cells were counted using a NOVEL Model NLCD-307 microscope, at a magnification of 10x90, under oil immersion. Cell counting was carried out using a morphometric grid mounted in the eyepiece of the microscope.

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The total number of lymphocytes, the number of large, medium and small lymphocytes per unit section area in PALM, in lymphoid nodules without a proliferation center were counted.

With the help directly from the general data matrix "Excel 7.0" on a personal computer Pentium-IV carried out mathematical processing, determined the indices of the standard deviation and the error of representativeness.

THE RESULTS OF THE STUDY AND THEIR DISCUSSION

When studying the spleen of 6-month-old intact rats, it was found that the weight of the animals is in the range from 190 g to 240 g, on average 220.2 ± 5.4 g. The absolute weight of the organ is 0.6-0.9 g, in average – 0.79 ± 0.032 g. Mass index ranges from 0.315% to 0.405%, on average 0.358 ± 0.01%. The weight of the animals in comparison with 3-month-old rats increased by 1.93 times, and the absolute weight of the organ increased by 1.52 times.

The length of the spleen ranges from 26.4mm to 35.7mm, with an average of 31.76 ± 1.0mm. The growth rate is 18.6%. The width of the spleen is in the range of 4.9-7.7 mm, on average – 6.34 ± 0.03 mm. The growth rate is 6.73%. The spleen thickness varied from 2.4 mm to 4.2 mm, on average – 3.12 ± 0.19 mm. The growth rate is equal to 6.85%.

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%. (fig. 1). 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).

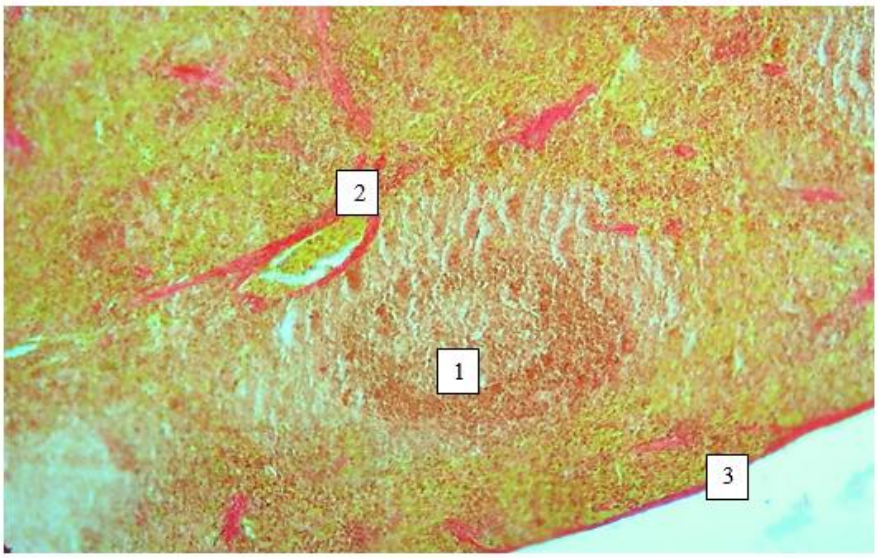

Fig. 1. Spleen of a 6-month-old rat of the control group. Coloring by Van Gizon. ok. 10 x vol. 20. 1-white pulp, 2–trabecula, 3–capsule Spleen.

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

All LN zones can be visually distinguished in microslides. The width of the mantle zone is from 40.5 microns to 50.4 microns, on average 46.56 ± 1.06 microns. The width of the marginal zone ranges from 74.5 µm to 86.2 µm, on average 80.72 ± 1.26 µm. The width of the periarterial zone ranges from 84.9 µm to 94.7 µm, on average 89.42 ± 1.06 µm. (fig. 2). The width of the mantle, marginal and periarterial zones increased by 2.74%, 4.64%, and 5.15%, respectively, in comparison with 3-month-old rats.

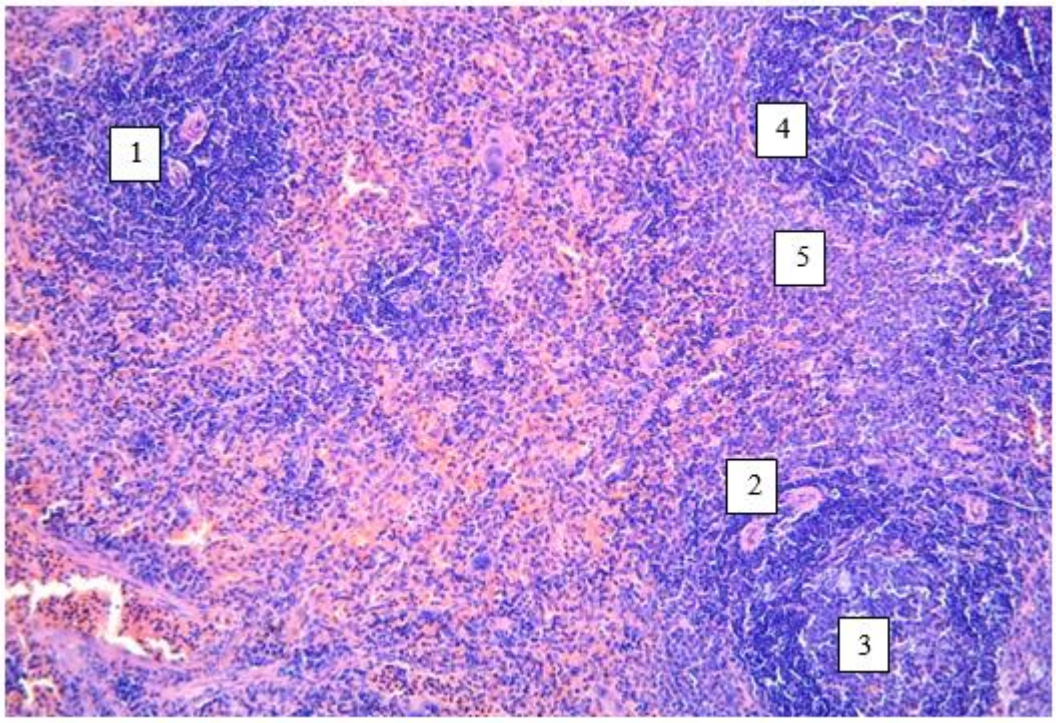

Fig. 2. Spleen of a 6-month-old rat of the control group. Coloration Hematoxylin – eosin. ok. 10 x vol. 20. 1- lymph node, 2-periarterial zone, 3-germinal center, 4-mantle zone, 5-marginal zone.

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It was found that the total number of lymphocytes in LN without proliferation centers is 52-61, on average, 57.2 ± 0.97 cells. The total number of lymphocytes in LN without proliferation centers increased by 21.0% in comparison with 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 muffs 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 muffs of the white pulp of the spleen increased by 23.7% compared with 3-month-old rats.

Periarterial lymphoid clutches 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.
The mass of 6-month-old laboratory animals with chronic radiation sickness is in the range from 170 to 250 g, on average 217.8 ± 7.36 g. The absolute mass of the organ is 0.54 to 0.86 g, on average – 0.73 ± 0.03 g. The mass index ranges from 0.277% to 0.392%, on average 0.335 ± 0.01%. The weight of the animals in comparison with 3-month-old irradiated rats increased by 2.02 times, and the absolute weight of the organ increased by 1.7 times.

The length of the spleen ranges from 27.4 mm to 35.8 mm, with an average of 31.3 ± 0.77 mm. The growth rate is 25.2%. The width of the spleen is in the range of 5.0-7.4 mm, on average – 6.16 ± 0.22 mm. The growth rate is -9.8%. The spleen thickness varied from 2.0 mm to 3.8 mm, on average 3.02 ± 0.16 mm. The growth rate is 12.0%.

According to our data, in 6-month-old irradiated rats, the relative area of the white pulp of the spleen ranges from 13.2% to 20.4%, on average -16.97 ± 0.66% (Fig. 3). The growth rate is 16.23%. The relative area of connective tissue elements varied from 5.8% to 7.0%, on average – 6.53 ± 0.11% (to the total area of the spleen section).

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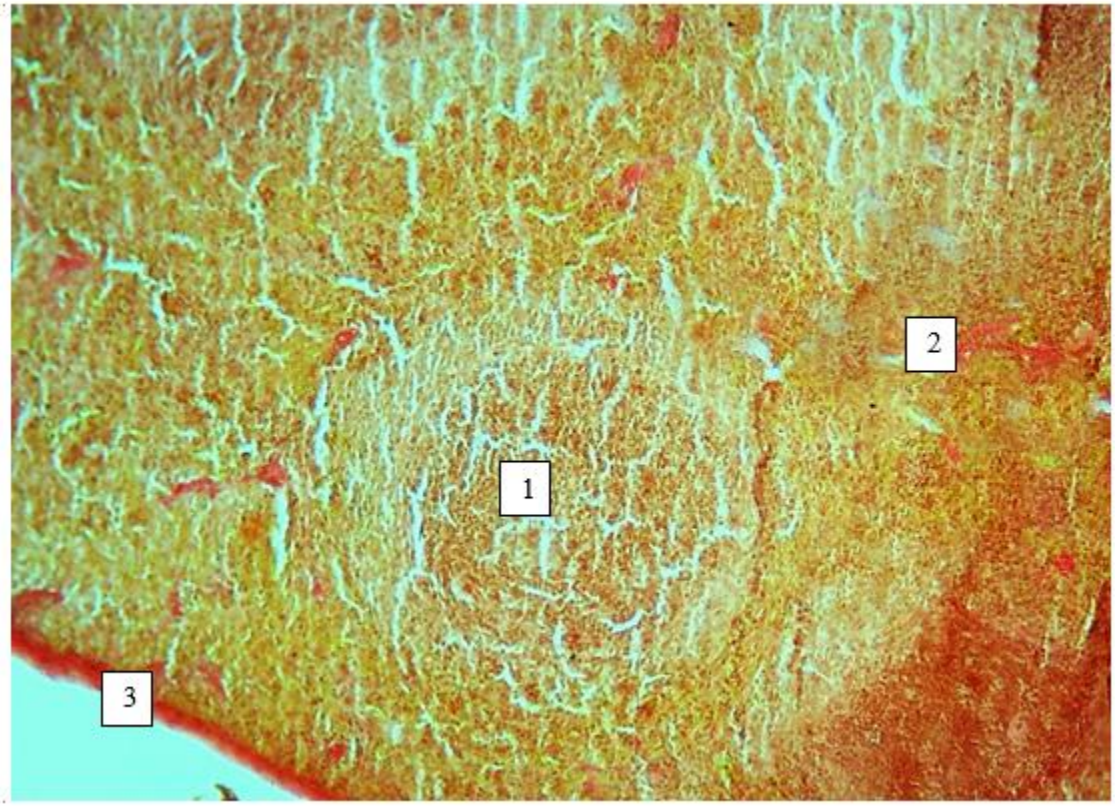

Fig. 3. The spleen of a 6-month-old rat with chronic radiation sickness. Staining by Van Gizon. ok. 10 x vol. 20. 1 – white pulp, 2 – trabecula, 3 – spleen capsule.

The PALM diameter ranges from 120.6 μm to 128.8 μm, on average 123.83 ± 0.75 μm. The growth rate is equal to 24.64%. The diameter of the lymph nodules ranges from 370.3 μm to 436.7 μm, with an average of 399.87 ± 6.1 μm. The growth rate is 104.2%. LUs have HZs. The percentage of primary and secondary LUs is 46% and 54%, respectively. The diameter of the germinal centers ranges from 96.3 μm to 122.8 μm, on average 106.09 ± 2.44 μm. Lymphoid nodules are generally round – oval, elongated (91.8%) and less often irregular (8.2%).

In most cases, the LN zones are clearly visible. The width of the mantle zone ranges from 38.4 μm to 47.6 μm, on average 43.64 ± 0.84 μm. The width of the marginal zone ranges from 69.2 microns to 79.8 microns, on average 74.81 ± 0.98 microns. The width of the periarterial zone ranges from 78.2 microns to 87.4 microns, on average 82.32 ± 0.84 microns. (fig. 4). The rate of increase in the width of the mantle, marginal and periarterial zones is 19.43%, 24.14% and 37.47%, respectively, in comparison with the 3-month-old rats of the irradiated group.

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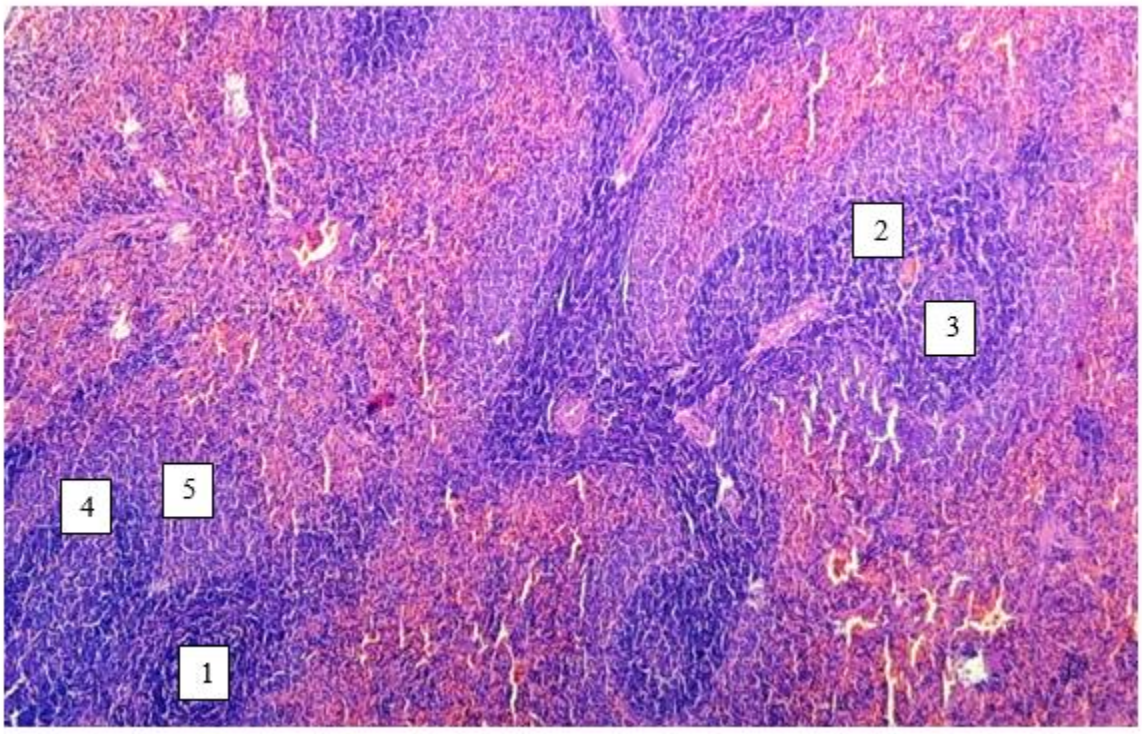

Fig. 4. The spleen of a 6-month-old rat with chronic radiation sickness. Staining with hematoxylin – eosin. ok. 10 x vol. 20. 1- lymph node, 2-periarterial zone, 3-germinal center, 4-mantle zone, 5- marginal zone.

It was found that the total number of lymphocytes in LN without centers of reproduction is 48-57, on average, 53.4 ± 0.83 cells. The total number of lymphocytes in LN without multiplication centers increased by 52.1% compared to 3-month-old irradiated rats.

Lymphoid nodules without reproduction centers contain (per unit area) small lymphocytes – 35-41, on average – 38.7 ± 0.55 cells, medium lymphocytes – 11-13, on average – 12.0 ± 0.18 cells and large lymphocytes – 2-3, on average – 2.7 ± 0.1 cells.

The total number of lymphocytes in the periarterial lymphoid muffs of the white pulp of the spleen is 50-60, on average, 54.2 ± 0.92

cells. The total number of lymphocytes in the periarterial lymphoid muffs of the white pulp of the spleen increased by 54.0% in comparison with 3-month-old irradiated rats.

Periarterial lymphoid clutches contain (per unit area) small lymphocytes -36-43, on average – 38.4 ± 0.64 cells, medium lymphocytes – 10-12, on average – 11.2 ± 0.18 cells and large lymphocytes - 4-5, on average – 4.6 ± 0.1 cells.

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CONCLUSIONS

In the spleen and structural formations of the white pulp of white rats with chronic radiation sickness, quantitative changes are observed, which are expressed in the morphological and morphometric parameters of the spleen. Irregular shapes (8.2%) of lymphatic nodules are revealed, which are not detected in the spleen of healthy rats. The width of the functional zones of the lymph nodules decreases, as well as the total number of lymphocytes in the lymph nodules without the center of proliferation and periarterial lymphatic muffs by 1.07 and 1.08 times, respectively. This indicates a negative effect of radioactive radiation on the lymphoid structures of the spleen, causing the development and formation of immunodeficiency.

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