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MORPHOMETRIC FEATURES OF THE INGUINAL AND POPLITEAL LYMPH NODES IN WHITE OUTBRED RATS WITH EXPERIMENTAL RHEUMATOID ARTHRITIS

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Abstract. Rheumatoid arthritis is accompanied by systemic immunomorphological changes affecting peripheral lymph nodes. The aim of the study was a comparative assessment of morphometric parameters of the inguinal and popliteal lymph nodes in white outbred rats with experimental rheumatoid arthritis. The results demonstrated that the inguinal lymph nodes were characterized by more pronounced proliferative activity of lymphoid tissue, whereas the popliteal lymph nodes showed a relative enlargement of the medullary zone and stromal component. The obtained data reflect regional features of the immune response during the inflammatory process.

Keywords: rheumatoid arthritis, lymph nodes, morphometry, immunomorphology, experiment.

Introduction. The lymphatic system represents a complex morphofunctional structure that plays a key role in maintaining the immune defense of the organism. It participates in the removal of excess interstitial fluid, transport of immunocompetent cells, antigen recognition, and the formation of immune responses [1]. Lymph nodes, as peripheral organs of the immune system, function as biological filters, retaining microorganisms, foreign antigens, and cellular degradation products carried by the lymph flow [2].

Within lymph nodes, antigen presentation to immunocompetent cells occurs, initiating the processes of adaptive immunity formation and ensuring an effective protective response against infectious and inflammatory factors [3]. Peripheral lymph nodes play an essential role in maintaining immunological homeostasis by facilitating interactions between antigen-presenting cells and lymphocytes [4].

Regional lymph nodes, including the inguinal and popliteal nodes, are of particular importance as they provide immune surveillance of the lower extremity tissues and respond to local inflammatory processes through changes in their morphological structure [5]. In these nodes, lymphocyte activation, clonal proliferation, and differentiation into effector immune cells take place [6].

Histologically, a lymph node consists of cortical, paracortical, and medullary zones, each characterized by a specific cellular composition and functional specialization [7]. The cortical zone contains lymphoid follicles rich in B lymphocytes, whereas the paracortical area is predominantly composed of T lymphocytes responsible for cellular immunity [8]. The medullary zone includes plasma cells, macrophages, and elements of the reticular stroma involved in the final stages of the immune response [9].

In chronic inflammatory diseases, including rheumatoid arthritis, lymph nodes undergo pronounced structural remodeling reflecting the degree of immune system activation [10].

Morphometric studies of regional lymph nodes allow an objective assessment of immune response characteristics and adaptive-compensatory processes developing under conditions of experimental inflammation [11].

Aim of the Study.

The aim of the present study was a comparative investigation of the morphological and morphometric features of the inguinal and popliteal lymph nodes in white outbred rats with experimental rheumatoid arthritis in order to assess regional characteristics of the immune response and structural remodeling of lymphoid tissue under conditions of a chronic inflammatory process.

Materials and Methods. The study was conducted in the research laboratory of the Center for Scientific and Experimental Biomedicine of the Bukhara State Medical Institute. Experimental investigations were performed on white outbred male rats aged 3 months, maintained under identical vivarium conditions. The body weight of the animals ranged from 350 to 370 g.

All laboratory animals were housed under standard vivarium conditions with relative air humidity of 50–60%, temperature of 19–22 °C, and a 12-hour light/12-hour dark cycle. Vivarium facilities were subjected to daily sanitary treatment in accordance with hygienic requirements. Animals were fed according to the standard laboratory animal diet.

The animals were divided into experimental groups. The control group consisted of intact rats maintained on a standard diet. In the experimental group, rheumatoid arthritis was induced using an adjuvant-induced inflammation model. For this purpose, 0.1 ml of Freund's adjuvant was administered once subcutaneously at the base of the tail. The adjuvant contained mannide monooleate, mineral oil, and inactivated *Mycobacterium tuberculosis*.

During the first three days after administration, local edema and hyperalgesia at the injection site were observed. On days 7–10, inflammatory changes developed in the distal joints, confirming the formation of the experimental rheumatoid arthritis model.

At the end of the experiment, animals were euthanized by decapitation in the morning under fasting conditions. Peripheral lymph nodes — inguinal and popliteal — were isolated for morphological examination. Their macroscopic dimensions were recorded, after which the material was fixed for further histological and morphometric analysis.

Histological specimens were prepared using standard techniques and stained with hematoxylin and eosin. Morphological examination was performed using light microscopy. The following morphometric parameters of the lymph nodes were evaluated: total diameter, thickness of cortical and paracortical zones, number and diameter of lymphoid follicles, proportion of germinal centers, relative area of the medullary zone, and volume fraction of the stromal component.

The obtained data were subjected to statistical analysis with calculation of mean values (M) and standard error of the mean (±Δ). The results are presented as mean values using methods of variational statistics.

Results. The conducted morphometric study demonstrated that experimental rheumatoid arthritis is accompanied by pronounced structural and functional changes in peripheral lymph nodes, manifested by regional features of lymphoid tissue remodeling.

Analysis of the inguinal lymph nodes in 3-month-old rats of the experimental group revealed an increase in the morphofunctional activity of the lymphoid apparatus. The total diameter of the lymph node ranged from 2.50 to 3.10 mm, with a mean value of 2.80 ± 0.075 mm, indicating activation of immunocompetent structures. The thickness of the cortical zone reached 580.25 ± 6.91 µm, while the paracortical zone measured 524.55 ± 6.46 µm, reflecting enhancement of both humoral and cellular components of the immune response.

The number of lymphoid follicles per section averaged 20.0 ± 0.25, with a mean follicular diameter of 259.4 ± 3.84 µm. A high proportion of germinal centers (65.1 ± 0.93%) indicates intensive B-lymphocyte proliferation and active development of the immune response. The relative area of the medullary zone was 18.4 ± 0.30%, whereas the volume fraction of the stromal component reached 28.2 ± 0.40%, suggesting preservation of the structural organization of lymphoid tissue despite inflammatory stimulation.

In the popliteal lymph nodes of the same animals, somewhat different morphometric characteristics were observed. The total lymph node diameter was 2.40 ± 0.050 mm, which was smaller than that of the inguinal lymph nodes. The thickness of the cortical zone measured 520 ± 7.58 µm, and the paracortical zone measured 480.05 ± 6.16 µm, indicating less pronounced proliferative activity of lymphoid elements (Fig. 1).

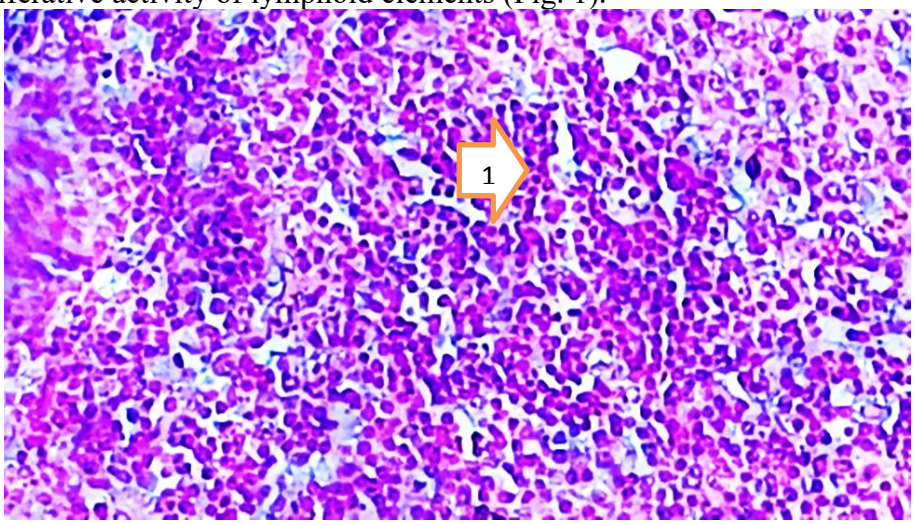

Figure 1. Histochemical analysis of changes in the popliteal lymph node in 3-month-old white outbred rats after experimental rheumatoid arthritis. Alcian blue staining. Objective lens ×20, eyepiece ×10. 1 — mucopolysaccharides.

The mean number of lymphoid follicles was 19 ± 0.25 per section, and their average diameter was 240.1 ± 3.59 µm. The proportion of germinal centers reached 60.25 ± 0.87%, indicating preservation of immune activity; however, its intensity was lower compared with the inguinal lymph nodes. At the same time, the relative area of the medullary zone increased to 20.10 ± 0.33%, while the volume fraction of the stromal component reached 28.93 ± 1.12%, reflecting a tendency toward enhancement of the stromal component and the development of compensatory-adaptive processes.

Comparative analysis demonstrated that in experimental rheumatoid arthritis, inguinal lymph nodes are characterized by more pronounced hyperplasia of lymphoid follicles and higher proliferative activity of germinal centers. In contrast, the popliteal lymph nodes showed a relative enlargement of the medullary region and stromal component, which may indicate regional differences in immune response and specific features of lymph circulation.

The obtained results indicate that the inflammatory process in rheumatoid arthritis induces heterogeneous morphofunctional remodeling in different groups of peripheral lymph nodes, reflecting adaptive mechanisms of the immune system under conditions of chronic inflammation.

Conclusion. Thus, experimental rheumatoid arthritis is accompanied by pronounced morphometric changes in regional lymph nodes of white outbred rats. The inguinal lymph nodes are characterized by larger size, thickening of cortical and paracortical zones, increased diameter of lymphoid follicles, and a higher proportion of germinal centers, indicating activation of the humoral immune response.

In the popliteal lymph nodes, a relative enlargement of the medullary zone and stromal component is observed, which may reflect the development of compensatory-adaptive and remodeling processes under chronic inflammatory conditions.

The identified morphometric features reflect the regional specificity of immune reactions and may serve as a morphological basis for further investigation of mechanisms underlying immune remodeling of lymphoid tissue in rheumatoid arthritis.

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