

# Structural and functional features of immune system cells in the dynamics of experimental temperature exposure

Shaxnoza Davronova\*, Rakhmon Davronov, and Jurat Bakhronov

Bukhara State Medical Institute, Bukhara, Uzbekistan

**Abstract.** The body's immune response to temperature exposure is a complex multi-stage process that is controlled by a variety of reactions developing at various levels of the body. The relationship of individual cell types is the most important link in the immune regulatory mechanism. The structural changes occurring in the organs of immunity under thermal exposure are in principle identical, but the involvement of one or another organ in the process depends on the activity and strength of temperature.

## 1 Introduction

The immune system, which includes central (thymus gland, bone marrow) and peripheral (spleen, lymph nodes, all diffuse lymphoid tissue of mucous membranes) organs, as well as effector cells - T, -B lymphocytes macrophages (A cells) in unity and in interaction with each other, provides immune homeostasis of the body [1, 2, 3, 4].

The initial stage after exposure to exogenous factors in the body is their recognition by T lymphocytes, which, migrating to the zone of primary affect, either destroy them or react with them by producing various kinds of soluble factors [7, 9, 10].

In recent years, a large number of works have appeared in the literature concerning changes in the cellular components of the immune system under various effects on the body. Nevertheless, there are few works by studying of immune organs under thermal effects on the body by general morphological, histochemical and electron microscopic research methods [5, 6, 7, 8].

## 2 Research materials and methods

All experiments were conducted on white mongrel male rats with an initial weight of 120-140 grams, who were on normal laboratory nutrition. All experimental animals were obtained from the same vivarium, the age of the animals, their weight and other indicators were under the same conditions.

Prior to the start of the experiment, 20 rats under ether anesthesia, under sterile conditions, underwent laparotomy and thoracotomy for the purpose of macroscopic examination of all internal organs and lymphoid formations of the gastrointestinal tract. After the examination,

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\* Corresponding author: shaxnozada9@gmail.com

a culture was taken on a medium of Ploskirev and bismuth sulfate agar from the contents of the ileum and colon for bacteriological studies. Analyses of these studies have shown the absence of growth of pathogenic microbes.

The experimental group of animals was quarantined for 1 month, and veterinary examinations were carried out regularly.

The experimental group of animals was kept for 1 hour in the hot climate of the Bukhara region, the cages with laboratory animals were outside under direct sunlight. To ensure the representativeness and randomization of the study, the same conditions for the action of environmental factors, control over laboratory conditions, including animal nutrition, are provided. After temperature exposure, the animals were transferred to normal laboratory conditions with unlimited access to water. Moreover, all experiments were conducted in the summer season (June, July, August) at a temperature of +390 +430C.

Experimental and control animals were slaughtered by decapitation, on an empty stomach, after 3,6,12,24 hours,3,5,7,14 ,21 days after infection.

Blood smears, bone marrow grains and pieces of thymus, spleen, mesenteric lymph nodes served as the material for research.

For the purpose of morphological studies, whole spleens and lymph nodes were taken from some control and experimental rats, pre-perfused with sterile saline solution, then 4% formalin solution, through the left ventricle of the heart under ether anesthesia.

For light-optical studies, the materials were fixed in 10% formalin, in Buena and Carnois liquids. The pieces of organs were poured into paraffin after appropriate treatment. The dewaxed sections were stained with Hemotoxylin-eosin, on the Brush RNA. Blood smears from the bone marrow were stained according to Romanovsky-Gimza.

Ultrastructural studies were carried out according to a generally accepted method. The ultrathin sections were viewed using a JEM-100S electron microscope (Japan).

Statistical processing of the obtained digital results was carried out in calculating the arithmetic mean -  $M$ , average deviations -  $\sigma$ , standard error - $m$ . We found the statistical significance of the data obtained by the Student's criterion - $t$  with the calculation of the probability of error -  $P$  when checking the normality of the distribution and the equality of the general variances  $F$  – Fisher's criterion. To assess the statistical significance of the identified indicators, the significance levels -  $R$ . Three main levels of significance were taken as statistically significant changes: average –  $p < 0.01$ , low –  $p < 0.05$ , insignificant –  $p < 0.050$ .

### **3 The results of our own research**

All our studies have shown that the organs of the immune system have a certain dynamics when exposed to temperature, which is divided into three types:

1. Early term (up to 12 hours after temperature exposure);
2. The peak period (I - 7 – day of research);
3. The recovery period (14-21 days of experiments).

No significant quantitative changes were found in the timing of early changes on the part of peripheral blood cells (see Table I). The indicators of the red part of the blood in 3-12 hours after exposure to temperature do not differ from those in the controls. However, when calculating the leukocyte formula, some tendency to leukocytosis with segmented gyperneutrophills is revealed (see Table 2). Table 2 shows that the number of segmented neutrophils 12 hours after temperature exposure to salmonella infection in rats is  $3.55 \pm 0.21 \cdot 10^9/l$  versus  $2.93 \pm 0.14 \cdot 10^9/l$  in controls.

**Table 1.** The condition of the red bone at temperature exposure (M±M).

Experiment	Hemoglobin (G/L)	Red Blood Cells ( $10^{12}$ G/L)	Color indicator
<b>Control</b>	158,0±1,2	5,7±0,1	0,8
12 h	151,0±1,4	5,6±0,1	0,8
24 h	142,0±1,1 <sup>+</sup>	6,0±0,2	0,7
3 d	120,0±1,3 <sup>+</sup>	5,0±0,1	0,7
5 d	134,0±1,2 <sup>+</sup>	4,6±0,1 <sup>+</sup>	0,8
7 d	135,0±16,6 <sup>+</sup>	4,9±0,1 <sup>+</sup>	0,8
14 d	140,0±5,6 <sup>+</sup>	5,1±0,2	0,8
21 d	152,0±4,3	5,3±0,2	0,8

Note: Here and in the following tables, the + sign is marked statistically significant differences compared with the control (P<0.05).

**Table 2.** Dynamics of the reaction of neutrophil granulocytes of bone marrow at temperature exposure (M± M).

Experiment	Total number of leukocytes	Neutrophils		
		Segmento nuclear	Stick- core	Young
<b>Control</b>	9,45±0,46	2,93±0,14	0,19±0,01	
12 h	9,60±0,56	3,55±0,21	0,10±0,01	
24 h	15,50±1,20 <sup>+</sup>	8,53±0,65	0,16±0,01	0,16±0,01
3 d	20,03±0,85 <sup>+</sup>	9,20±0,39 <sup>+</sup>	0,60±0,03 <sup>+</sup>	0,40±,02
5 d	22,01±0,96 <sup>+</sup>	6,16±0,27	0,88±0,04	0,66±0,03
7 d	18,80±1,14 <sup>+</sup>	4,68±0,29 <sup>+</sup>	0,56±0,03 <sup>+</sup>	0,19±0,01
14 d	13,90±0,86 <sup>+</sup>	2,92±,18	0,14±0,01	
21 d	8,80±0,42	2,02±0,10	0,09±0,004	

Morphometry of sections of the thymus gland of control animals revealed that 71% is the area of the cortical, 21% is the medullary zone, 5% is the cortico-medullary zone, 3% falls on the area of the connective tissue of the capsule and interlobular septa (Table No. 3).

**Table 3.** Indicators of the areas of the thymus gland in dynamics temperature exposure.

Experiment	THYMUS ZONES				Cortico-medullary (rel. units %)	
	Cortical (rel. units, %)		Medullary (rel. units, %)			
<b>Control</b>	45,4± 0,5	71	16,4±0,6	21	2,2±0,16	5
12 h	36 ±0,5*	57	27,6±0,5*	34	6,2±0,6*	9
24 h	35,2±0,3*	55	23,5±0,3*	37	5,3±0,4*	8
3 d	40,3±0,7*	63	21,3±0,8*	33	2,3±0,2	4
5 d	44,7±0,2	70	16,2±0,2*	24	4,1±0,1*	6
7 d	32,6±0,5*	51	26,1±0,3*	41	5,3±0,1*	8
14 d	37,1±0,3*	58	21,8±0,4*	33	5,6±0,2*	9

Immunomorphological rearrangements of the spleen in the dynamics of temperature exposure are generally of the same type as those in mesenteric lymph nodes. However, in terms of time parameters, these changes in both bodies differ somewhat from each other. Thus, pronounced immunomorphological rearrangements in mesenteric lymph nodes are detected on the 3-7 day of experiments, whereas in the spleen they occur on the 3-14 day of experiments.

And so, in the early period of the experiments, changes in almost all components of the microcirculatory bed were noted (vascular dilation, blood stasis, increased cell migration) and destructive changes in some cells (expansion of perinuclear spaces, swelling and lysis of mitochondria, destruction of cells themselves).

Hypertrophy and functional tension of immune cells were observed on 1-7 days of experiments. In these terms, the number of macro organs, lymphopoiesis cells with activation of their intracellular components, and high plasmatisation were increased.

On the 14th-21st day of the experiments, the functional tension of the immune cells somewhat subsides, however, the number and functional activity are observed from the stromal elements of the immune system. Moreover, fibroblasts are activated, areas of sclerosis and elements of fatty degeneration appear in the organs.

## 4 Conclusions

Immune organs under temperature exposure are characterized by periodicity. There are periods of early changes (up to 12 hours of experiments), peak of perestroika (1-7 days of experiments) and recovery period (14-21 days).

The period of early changes is characterized by:

- leukocytosis with segmented neutrophilosis;
- microcirculatory disorders in mesenteric lymph nodes, especially in the postcapillary venules of paracortical zones, increased emigration of lymphocytes, which lead to a decrease in the areas of T-dependent zones.

The peak period (1 - 7 - day) is characterized by:

- a high degree of plasmatisation of the bone marrow, destructive changes in organelles in developing cells of the granulocyte and erythrocyte series, increased functional activity of bone marrow macrophages;

- a low number of small and medium-sized lymphocytes of the cortical and corticomedullary zones of the thymus;

- hypertrophy and hyperplasia of lymphoid follicles of mesenteric lymph nodes and spleen, an increase in the areas of B - dependent zones with an increase in proliferating cells in them, the degree of plasmatisation, the number and functional activity of macrophages.

The recovery period (14-21 days) is characterized by a tendency to normalize quantitative and qualitative changes in cells of various structural zones of the immune system, a decrease in the proliferative activity of cells in them. However, the tension of the subcellular structures of immuno-competent cells still persists, there is an increase in the number and functional activity of stromal cells, primarily fibroblasts.

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