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Morphological Features of Bone Marrow Cells Under the Antigenic Effect

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Summary: The bone marrow in the adult body is a universal hematopoietic central organ of B-lymphopoiesis. Therefore, one of the reactions of the bone marrow to antigenic effects is a reaction from lymphopoiesis. With salmonella exposure, activation of lymphopoiesis in the bone marrow is accompanied by parallel pronounced absolute and relative lymphocytosis.

Key words: Bone marrow, antigen, salmonellosis, infection.

Relevance: The study of the structural and functional foundations of adaptive changes in the organs of the immune system is one of the urgent problems of modern medicine and biology in general. The choice of the model was determined to a certain extent by the need to conduct such studies in connection with the urgency of the problem of exposure to antigens and the problem of salmonellosis in our region.

The red bone marrow of an adult under normal physiological conditions contains hematopoietic stem cells. In the hematopoietic tissue of the bone marrow, differentiation of all hematopoiesis sprouts is carried out. The red bone marrow as a full-fledged organ of hematopoiesis is formed for the first time in amphibians, their hematopoiesis is activated during the summer periods of life. In reptiles and birds, the red bone marrow is also a universal hematopoietic organ (10,11,14,15).

The bone marrow of almost all representatives of vertebrates is built the same way. Its stroma is made up of reticular tissue. The hematopoietic tissue of the red bone marrow is a heterogeneous population of cells, where both undifferentiated and mature hematopoiesis cells are found (1,2,4,6,7).

The reticular tissue of the bone marrow stroma is a network of cells that are heterogeneous in morphological and histogenesis respects. These include reticular cells, fibroblasts, endothelial and fat cells, which together create a microenvironment for the differentiation of myeloid and lymphoid germ cells (3,5,8,9,12,13).

Material and methods: The experiments were carried out on white sexually mature mongrel male rats with an initial weight of 120-130 grams, which were on a regular laboratory diet. After the examination, a culture was taken on a medium of Ploskirev and bismuth sulfate agar from the contents of the ileum and colon for bacteriological studies. The analyses of these studies showed the absence of growth of salmonella and other pathogenic microbes.

The experimental animals were divided into three groups. The first group consisted of 32 intact rats. The second group is experimental (218 rats). After 48 hours of fasting, 2 ml of whole cow's milk was injected into their stomach through a probe to neutralize gastric juice, 30-35 minutes after that, the animals were infected with the pathogenic strain of salmonella mouse type No. 57775 (Salm. tyhmurium) at a dose of 2 billion microbial bodies in 2 ml of saline solution.

The third group consisted of 100 control rats. After 48 hours of fasting, 2 ml of whole cow's milk was injected into their stomach through a probe, and then 30 to 35 minutes later, 2 ml of sterile saline solution was injected into their stomach.

Blood smears and bone marrow grains served as the material for research.

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

The study of peripheral blood and bone marrow was carried out according to the following basic tests:

1. Cytological analysis of peripheral blood and bone marrow with calculation of hemogram and myelogram;

2.Cytochemical study of peripheral blood neutrophils with a semi-qualitative assessment of the activity of peroxidase, acid and alkaline phosphatases (K.Butenko et al., 1974).

3.Electron microscopic cytochemistry of peroxidase by the method of R.Gracham,M.Karnovskiy (1966).

4. Scanning electron microscopy of blood cells using the Yu.A. method.Rovensky (1979).

5.Immunocytochemical study of peripheral immunoglobulin-bearing (p-IG) lymphocytes at the level of electron microscopy.

For electron microscopic studies, pieces of thymus were fixed in 2.5% glutaraldehyde solution at 4 ° C for 40 minutes, followed by additional fixation in 1% osmic acid solution for I hour at 4 ° C. The materials were dehydrated in alcohols of increasing concentration, poured into araldite and epon-812. Ultrathin sections were obtained after taking and appropriately coloring the sighting semifine sections (E.Encuses, F.Ehrenpreis 1980) on an ultramicrotome from LKB (Sweden). The contrast was carried out with uranyl acetate and lead citrate, after which the sections were viewed in an electron microscope JEM-100S from Geol (Japan).

The results of the study and their discussion:

Our studies have shown that the structural and functional rearrangements of bone marrow and blood cells in experimental salmonella infection have a certain dynamics, which can be divided into three periods:

1. The early period (3-12 hours after infection);
2. The period of peak infection (I - 7 - day);
3. The period of reconvalescence (14-21 days)

Table No. 1

THE STATE OF RED BLOOD IN THE EXPERIMENTAL SALMONELLOSIS (M±m)

Experiment	Hemoglobin (G/L)	Red Blood Cells (1012 G/L)	Color indicator
Control	158,O+ 1,2	5,7±0,1	0,8
3 ч	161,0 ± 0,7	5,8±0,1	0,8

6 ч	154,0±2,6	5,5±0,1	0,8
12 ч	151,0±1,4	5,6±0,1	0,8
24 ч	142,0±1,1^{+}	6,0±0,2	0,7
3 с	120,0±1,3^{+}	5,0±0,1	0,7
5 с	134,0±1,2^{+}	4,6±0,1^{+}	0,8
7 с	135,0±16,6^{+}	4,9±0,1^{+}	0,8
14 с	140,0±5,6^{+}	5,1±0,2	0,8
21 с	152,0±4,3	5,3±0,2	0,8

During the period of early changes on the part of peripheral blood cells, no special quantitative changes were found (see Table I).As can be seen from table No. I, the indicators of red blood in 3-12 hours after infection with salmonella do not differ from those in the control. However, when calculating the leukocyte formula, some tendency to leukocytosis with segmented neutrophilosis is revealed (see Table 2). Table 2 shows that the number of segmented neutrophils 12 hours after infection with salmonella in rats is 3.55 ± 0.21* 109/l versus 2.93 ±0.14*109/l in controls.

Table No. 2

DYNAMICS OF THE REACTION OF NEUTROPHILIC GRANULOCYTES OF BLOOD ON EXPERIMENTAL SALMONELLA EXPOSURE (M= m 109\L)

The timing of the experiment	The total number of Ley-kotsit	Neutrophils		
		Segmento nuclear	Stick-core	Young
Control	9,45±0,46	2,93±0,14	0,19±0,01	
3 ч	9,00±0,43	2,61±0,13	0,09±0,004	
6 ч	9,50±0,18	2,47±0,05	0,19±0,004	
12 ч	9,60±0,56	3,55±0,21	0,10±0,01	
24 ч	15,50±1,20^{+}	8,53±0,65	0,16±0,01	0,16±0,01
3 с	20,03±0,85^{+}	9,20±0,39^{+}	0,60±0,03^{+}	0,40±,02
5 с	22,01±0,96^{+}	6,16±0,27	0,88±0,04	0,66±0,03
7 с	18,80±1,14^{+}	4,68±0,29^{+}	0,56±0,03^{+}	0,19±0,01
14 с	13,90±0,86^{+}	2,92±,18	0,14±0,01	
21 с	8,80±0,42	2,02±0,10	0,09±0,004	

One of the characteristic signs of the period of early changes is an increase in the cytochemical parameters of neutrophil granulocytes (Table Z).The activity of alkaline phosphatase in the early period of the experiment gradually increases, amounting to 195.0 ± 3.0 conl.units after 321.4 ± 3.0 conl.units after 6 and 242.0 ± 2.0 conl.units after 12 hours of the study against 181.0 + 2.0 conl.units in the controls. By 12 hours of the study, the activity of myeloperoxidase also significantly increased (190.0±1.0 versus 132.0+2.0 conl.units in the controls).

Table No. 3

The duration of the study	Alkaline phosphatase	Acid phosphatase	Myeloperoxidase
Control	181,0±2,0	83,0±2,0	132,0±2,0
3 ч	195/,0±3,0	79,0±4,0	137,0±4,0
6 ч	214,0±3,0	81,0±1,0	140,0±3,0
12 ч	242,0±2,0	87,0±2,0	190,0±1,0
24 ч	259,0±7,0	85,0±2,0	219,0±2,0
3 с	311,0±4,0	90,0±2,0	180,0±2,0
5 с	209,0±5,0	108,0±3,0	178,0±5,0
7 с	203,0±2,0	132,0±2,0	135,0±1,0
14 с	197,0±4,0	123,0±3,0	134,0±1,0
21 с	191,0±2,0	96,0±2,0	134,0±5,0

We did not find any significant changes in the quantitative composition in the myelogram in the early period.

Ultrastructural studies of bone marrow in the early period of experimental salmonellosis revealed a number of changes in the submicroscopic organization of its cellular components. One of the earliest signs is disorders of the microcirculatory system, manifested in the form of dilation of capillaries, arterioles, venules, capillarostasis. Clusters of red blood cells and other blood cells are detected in the lumen of dilated capillaries. Destructive changes in the cellular components of the bone marrow are often found. Moreover, the destruction of subcellular organoids of cells of almost all hematopoiesis germs is determined.

The most pronounced structural and functional rearrangements of blood and bone marrow cells are observed in the period of the height of experimental salmonella infection (1-7 days of the experiment). Anemia is noted in the blood, which is associated with a parallel decrease in hemoglobin and the number of red blood cells. Absolute leukocytosis is also characteristic. As shown in Table 3, leukocytosis is accompanied by a significant increase in the number of neutrophils (segmented, rod-shaped, juvenile) eosinophils, basophils, monocytes and lymphocytes.

Conclusions: 1). Structural and functional bone marrow rearrangements in the dynamics of experimental salmonellosis are characterized by periodicity. There are periods of early changes (3-12 hours of experiments), peak perestroika (1-7 days) and long-term changes (14-21 days).

2). Each of these periods is characterized by structural, functional and quantitative features, which together determine the essence of adaptive reactions of the immune system in response to salmonella exposure.

3). The most pronounced changes they are observed at the peak of experiments. They manifest themselves in the form of:

-leukocytosis with segmented neutrophilosis, increased cytochemical parameters of neutrophilic granulocytes (alkaline phosphatase, myeloperoxidase);

-microcirculatory disorders of the capillaries of the bone marrow, destructive changes in part of the stromal and hematopoietic cells in it.

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