Mechanisms of Hemopoiesis in Athymic Mice

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Cell elements of peripheral blood and bone marrow and splenic hemopoiesis were studied in athymic BALB/c mice (nude or thymectomized animals). In athymic animals lympho- and erythropoiesis were suppressed, while granulocyto- and monocytopoiesis were activated. These changes were more pronounced in nude mice.

Key Words: *hemopoiesis; thymus; T cells; thymectomy, nude mice*

It is now established that thymus-derived lymphocytes play an important role in the regulation of proliferation and differentiation of hemopoietic cells. In some experimental situations (immobilization stress, cytostatic and radiation treatments) T-lymphocytes realize their regulatory effects at the level of committed precursors either directly or via interaction with other elements of hemopoietic microenvironment [1-3,5]. In light of this, the study of this problem requires new experimental approaches suitable for evaluation of the role of lymphocytes in the regulation of hemopoiesis. Athymic nude mice and thymectomized mice can serve as these models. Here we compare hemopoiesis in these animals.

MATERIALS AND METHODS

Three-month-old BALB/c mice (n=37) obtained from the Institute of Pharmacology were used in the experiments. Experimental groups consisted of 10 homozygous nude mice and 15 thymectomized (at the age of 2 months) mice, other animals served as controls.

Specimens were obtained 1 month postoperation. The total erythrocyte and leukocyte counts in the peripheral blood were determined by standard hematological techniques [4,6]. Differential leukocyte count was determined using Nokht—Maksimov-stained smears. Bone marrow cells (from the femur) and splenocytes were counted in a Goryaev chamber, smears were prepared for morphological analysis. Colony-forming activity of myelokaryocytes was analyzed by cloning nonadhesive bone marrow cells in a semisolid medium. Committed erythro- (CFU-E) and granulomonocytopoiesis (CFU-GM) precursors were counted [4].

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RESULTS

Homozygous nude mice demonstrated peripheral blood leukopenia due to a significant decrease in lymphocyte count compared to nonmutant mice (Fig. 1). Similar changes were found in thymectomized animals. Leu-

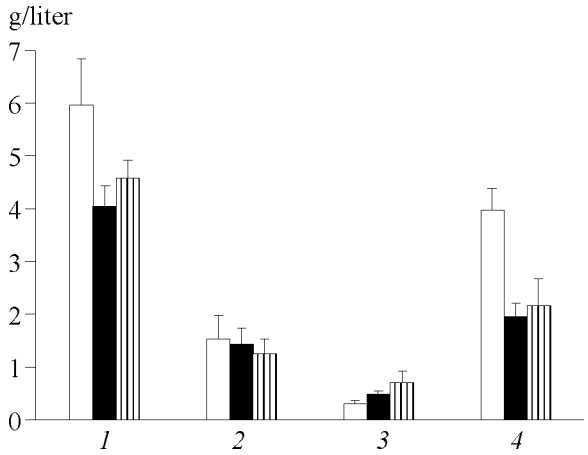

Fig. 1. Total number of leukocytes (*1*), segmented neutrophils (*2*), monocytes (*3*), and lymphocytes (*4*) in nude (dark bars) and thymectomized (shaded bars) BALB/c mice. Here and in Fig. 2: open bars: intact animals.

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TABLE 1. Bone Marrow Myelogram ($\times 10^6$/femur) and Number of CFU (per Femur) in Nude and Thymectomized BALB/c Mice.

Parameter	Intact	Thymectomized	Nude
Myeloblasts	0.301±0.030	0.349±0.050	0.125±0.030*+
Segmented neutrophils	1.921±0.540	2.772±0.660*	3.381±0.670*+
Lymphocytes	6.642±0.460	7.053±0.820	1.289±0.400*+
Monocytes	0.295±0.200	0.234±0.280	1.010±0.310*+
Erythrocytes	0.203±0.030	0.166±0.030	0.021±0.010*+
Polychromatophilic normoblasts	0.406±0.050	0.245±0.030*	0.125±0.040*+
CFU-E	18.879±2.200	37.871±4.880*	17.869±2.180+
CFU-GM	546.263±67.830	82.828±8.520*	462.526±66.970+

Note. $p<0.05$ *compared to intact and +thymectomized animals.

kocyte and lymphocyte counts did not differ in nude and thymectomized mice, while monocytosis was most pronounced (by 45%) in thymectomized animals.

No significant differences in erythrocyte and reticulocyte content between nude and thymectomized mice were found.

Analysis of myelograms revealed no changes in the total myelokaryocyte count in athymic mice compared to the control (Table 1), while the content of segmented neutrophils in the bone marrow increased in nude and thymectomized mice by 270 and 76%, respectively. The content of myeloblasts in nude mice significantly decreased compared to intact and thymectomized animals. The content of monocyte precursors increased in nude mice, but not in thymectomized animals (Table 1). Inhibition of erythropoiesis was found in all athymic animals, but in nude mice these changes were more pronounced (Table 1). The content of erythroblasts also decreased. The content of lymphoid elements in the bone marrow of nude mice was 4.15-fold below the control.

The number of CFU-E increased and the number of CFU-GM decreased in thymectomized mice compared to intact and nude mice (Table 1).

The total content of splenocytes was similar in all groups. The number of erythroid cells in the spleen increased in both nude and thymectomized mice. In nude mice a decrease in the number of large (by 99 and 83% compared to control and thymectomized animals, respectively) and medium (by 89 and 82%, respectively) lymphocytes was found, while the content of monocytes and mature and immature granulocytes in these groups increased (Fig. 2).

Thus, inhibition of lymphoid hemopoiesis was characteristic of all athymic animals, but in nude mice these changes were more pronounced and involved also bone marrow and spleen. Thymus and thymus-derived lymphocytes regulate all hemopoietic stems, which is confirmed by activation of granulocyto- and monocytopoiesis in athymic animals. Inhibition of

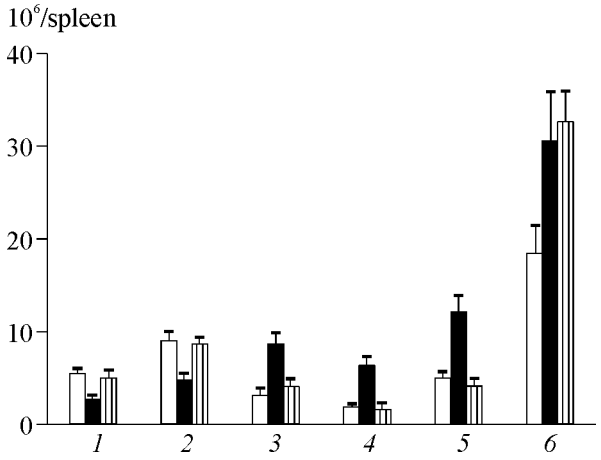

Fig. 2. The number of large (*1*) and medium lymphocytes (*2*), monocytes (*3*), immature (*4*) and mature neutrophils (*5*), and erythroid cells (*6*) in nude (dark bars) and thymectomized (shaded bars) BALB/c mice.

erythropoiesis in the bone marrow and compensatory stimulation of this process in the spleen were also noted.

In nude mice, the described changes in hemopoiesis were maximum and involved deep reserves of hemopoiesis (precursors), which suggests that, in contrast to thymectomized animals, impairment of their blood system depended largely on changes in the regulatory apparatus. Since interaction with T cells plays a key role in functioning of hemopoiesis-inducing microenvironment [1-3,5,7], changes in this component of hemopoiesis regulation are probably responsible for the observed hematological changes in athymic animals.

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