

During the first day of cultivation a decrease was observed both in the total number of cells and in the number of CFU_c. On the fifth day of cultivation the number of CFU_c in the explant increased, but did not reach its initial level, and later it decreased until the 50th day. The total number of living cells also fell at this time, and by the 50th day hematopoiesis was virtually exhausted. The number of CFU_c decreased more than the number of living cells (Table 2).

The results thus indicate that hematopoiesis of erythroid type can continue for a long time (over 1.5 months) in organ cultures of human embryonic liver. In cultures of mouse embryonic liver the erythroid cells were observed [3] to disappear by the 10th-14th day of cultivation, whereas the cells of the myeloid series persisted for a long time.

Differences in the type of hematopoiesis during cultivation of mouse and human embryonic liver may be associated with species differences. The possibility cannot be ruled out that the conditions of cultivation of mouse embryonic liver were inadequate to maintain erythroid hematopoiesis. Human embryonic liver cells may also be more sensitive to the presence of exogenous erythropoietin in the nutrient medium. Another possibility is that human embryonic liver cells can produce their own erythropoietin.

Research into the study of erythroid cells in organ culture of human embryonic liver is continuing at the present time.

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CHARACTERISTICS OF HEMATOPOIESIS IN NUDE MICE

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The state of the bone marrow, peripheral blood, and spleen was studied from the age aspect in nude mice, characterized by the presence of the recessive nu mutation. In athymic homozygous (nu/nu) individuals, various features were found in the blood system that distinguished them from heterozygous (nu/+) animals: a low lymphocyte count in the peripheral blood, bone marrow, and spleen, inhibition of erythropoiesis in the bone marrow, and hyperplasia of erythroid elements in the spleen.

KEY WORDS: *nude mice; peripheral blood; bone marrow; thymus; spleen.*

The question of the role of the thymus in hematopoiesis has not yet been fully explained. A very interesting aspect of this problem is the study of nude mice, which were described previously [1]. These mice are characterized by the almost complete absence of hair, as the result of a recessive nu mutation. Homozygous (nu/nu) individuals are also distinguished by absence of the thymus and by certain other features [3]. The leukocyte

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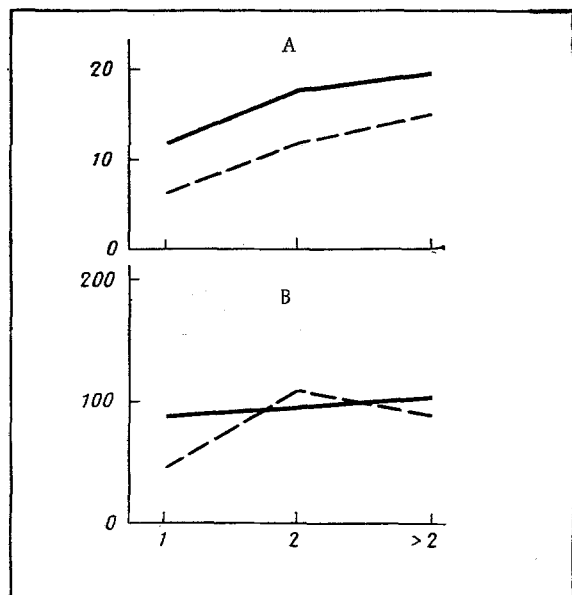


Fig. 1

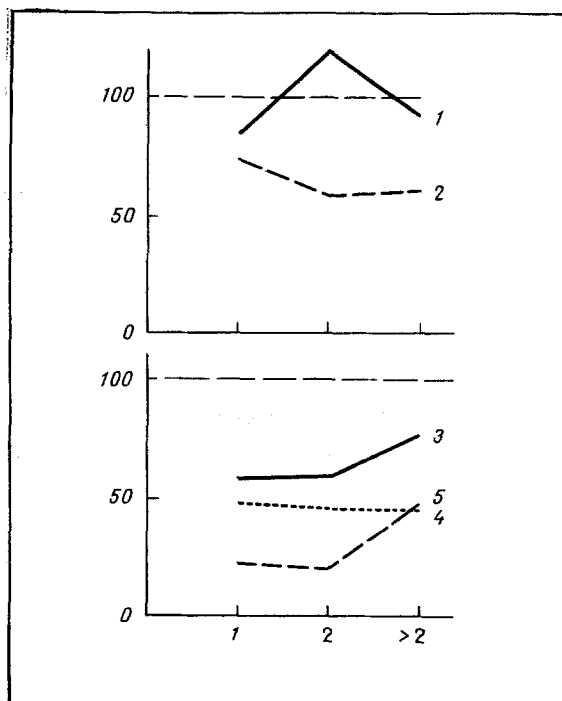


Fig. 2

Fig. 1. Age dynamics of body weight and weight of spleen in homozygous and heterozygous nude mice. Abscissa, age (in months); ordinate: A) body weight (in g); B) weight of spleen (in mg). Broken line denotes homozygous, continuous line heterozygous mice.

Fig. 2. Age dynamics of state of peripheral blood and bone marrow of nude mice. Abscissa, age (in months); ordinate, number of cells in homozygous mice (in % of corresponding number in heterozygous mice); 1) leukocytes; 2) lymphocytes; 3) myelokaryocytes; 4) lymphocytes; 5) erythroid bone marrow cells.

count in the peripheral blood of athymic mice has been shown to be extremely low [3], the lymph nodes and spleen are considerably reduced in size [4], and phenomena of hematopoiesis are found in the spleen [2]. The state of hematopoiesis in nude mice has not previously been studied.

EXPERIMENTAL METHOD

Altogether 53 nude mice (mainly females) obtained from the animal house of the Institute of Cytology and Genetics, Siberian Branch, Academy of Sciences of the USSR, were investigated. Thirty of the animals were homozygous (nu/nu) and 23 were heterozygous (nu/+) sibs. The mice were divided into three groups (6-17 individuals in each group): aged 1 month (20-31 days), 2 months (34-59 days), and over 2 months (68-124 days). The body weight and also the absolute and relative weight of the spleen were determined in the animals; the erythrocyte, reticulocyte, platelet, and total and differential leukocyte count were determined in blood from the caudal vein, and the total myelokaryocyte count (per femur) and the myelogram were investigated in the bone marrow; the overall mitotic index for the bone marrow cells was recorded and the splenogram calculated from squash preparations of the spleen.

EXPERIMENTAL RESULTS

The body weight and the weight of the spleen of the athymic mice aged 1 month were only half those of the heterozygous mice ($P < 0.05$). With an increase in age the differences in the body weight of the animals persisted (Fig. 1A), whereas differences in the weight of the spleen disappeared (Fig. 1B). The relative weight of the spleen in the athymic animals over 1 month old was thus considerably greater than for the nu/+ individuals.

The erythrocyte count in the blood of the mice increased with age in both groups. The reticulocyte count in the peripheral blood fell with age; in the first two age groups there

was no difference in their number in homozygous and heterozygous individuals, whereas in nu/nu mice over 2 months old they were almost twice as numerous as in nu/+ mice.

In mice of both groups a tendency was observed for the total leukocyte count to rise with age; in the nu/+ mice this increase took place on account of both neutrophilic granulocytes and lymphocytes, whereas in the nu/nu mice it took place purely on account of mature neutrophils. The mean leukocyte count in homozygous and heterozygous animals was almost the same, but mature granulocytes were much more numerous in nu/nu than in nu/+ mice, especially in the first two age groups. The lymphocyte count was much smaller in the blood of all the athymic mice (Fig. 2A).

The total number of bone marrow cells was increased in the mice of both groups, but in athymic individuals it was smaller than in the heterozygous animals. Analysis of the myelograms showed a marked decrease in the number of lymphocytes in the bone marrow of the nu/nu mice. For instance, in the first two age groups their absolute number was less than one-fifth of that in the nu/+ mice. In animals over 2 months old, although the lymphocytes were more numerous, they reached only about 50% of their level in nu/+ mice. A similar relationship also was found with respect to the erythroid cells. Granulocytopoiesis was predominant in the bone marrow of the athymic animals. Whereas in the heterozygous mice the leukoerythroblastic ratio was between 1.5 and 2.2, in homozygous mice aged 1 month it was 2.4, in those aged 2 months it was 4.1, and over 2 months 4.6. The overall mitotic index of the bone marrow cells was almost the same in the mice of both groups and it was independent of age.

An increase in the number of reticular cells, monocytes, and macrophages was found in squash preparations from the spleen of the athymic mice, together with a marked decrease in the number of small lymphocytes and their precursors, marked erythropoiesis, and intensification of granulopoiesis and of megakaryocytopoiesis.

Athymic mice (nu/nu) also differed from the nu/+ mice in their external appearance; they were characterized by absence of hair, and the skin was dry and resembled parchment. Direct correlation was found between the number of myelokarocytes in the bone marrow and the body weight of the athymic mice; for corresponding age groups the value of r was 0.75 ($P < 0.05$), 0.74 ($P < 0.01$), and 0.9 ($P < 0.01$).

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