EFFECT OF EXTRACTION OF LARGE AMOUNTS OF BONE MARROW ON THE BLOOD SYSTEM AND SURVIVAL

I. A. Rudakov, V. M. Zyablitskii, N. A. Zhukova, and S. F. Rudakova

UDC 612.014.482+612.419-089.87-07: 612.119+612.013

Removal of marrow and medullary puncture improve the chances of survival of irradiated rats.

* * *

The large-scale application of transplantation of hemopoietic tissues for treatment of radiation sickness is being impeded at the present time by a number of difficulties. Autotransplantation, for example, is possible only when the irradiation is planned beforehand and when unirradiated marrow can be obtained [4]. The use of homografted marrow requires very careful choice of donor in order to minimize the risk of onset of immunologic conflict [3, 6, 8, 9].

This lends interest to the study of the therapeutic value of autologous bone marrow obtained from the totally and uniformly irradiated organism. Only isolated investigations have been conducted to study the possibility of using this method in treatment of radiation sickness [7]. However, because this treatment requires the extraction of a large amount of marrow tissue in the post-irradiation period, it was decided to study the effect of massive loss of marrow on the blood system and the survival rate of irradiated rats.

EXPERIMENTAL METHOD

Experiments were conducted on 65 August rats weighing 190-210 g. In series I the effect of massive extraction of marrow on the hemopoiesis of healthy animals was studied. After preliminary investigation of the blood, marrow (on the average 17×10^6 nucleated cells per animal) was extracted from the rats by femoral puncture under ether anesthesia. The peripheral blood morphology was investigated on the 1st, 3rd, 5th, 7th, 15th, and 30th days after extraction of the marrow.

In the experiments of series II the effect of extraction of marrow on hemopoiesis of irradiated rats was studied. After preliminary investigation of the blood the animals were divided into 4 groups and irradiated on a "Gammacel-220" apparatus in a dose of 550 R at the rate of 77 R /sec. During the 3 h after irradiation marrow was extracted from one femur of the rats of group 1, and from both femora of the rats of group 2. A mock puncture of the femora was performed on the rats of group 3. The animals of group 4 were irradiated and then merely anesthetized with ether; these rats acted as controls. Blood was taken for investigation on the 5th, 11th, 20th, and 30th days after irradiation.

In series III the effect of stimulation of liberation of hemopoietic cells into the blood stream on the survival rate of the irradiated rats was studied. During the 3 h after irradiation in a dose of 600 R, a mock puncture of one femur was performed on the rats of group 1 and 0.5 ml physiological saline was injected into the other. The animals of group 2 were simply anesthetized with ether and acted as controls.

EXPERIMENTAL RESULTS

The leukocyte count of the unirradiated animals was increased by 50% on the 1st-3rd days after marrow extraction chiefly on account of neutrophils. By the end of the 1st week the number and composition of the leukocytes were normal again. The changes in the red blood cells were more marked. A decrease in the erythrocyte count and hemoglobin concentration was observed in the blood 24 h after marrow extraction, reaching a minimum on the 3rd-5th day (70-80% of the initial level). The anemia was persistent, and not until the 30th day were the initial values of hemoglobin concentration and erythrocyte count regained.

Laboratory of Experimental Hematology, Department of Pathophysiology, Institute of Medical Radiology, Academy of Medical Sciences of the USSR, Obninsk (Presented by Active Member of the Academy of Medical Sciences of the USSR P. D. Gorizontov). Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 65, No. 1, pp. 40-42, January, 1968. Original article submitted February 21, 1966.

TABLE 1. Survival Rate of Rats after Irradiation in Dose of 550 R Depending on Extent of Loss of Marrow

Operation performed	No. of cells taken	Number of rats				
		Total	surviving		dying	
			absolute	%	absolute	%
Puncture of one femur	$21.7 \cdot 10^6$	10	8	80	2	20
Puncture of both femora	$33.1 \cdot 10^6$	9	7	78	2	22
Mock puncture	–	9	7	78	2	22
Control	–	9	5	56	4	44

Throughout the period of observation erythropoiesis was intensified as shown by increases in the number of large erythrocytes and in their mean diameter.

In series II the clinical manifestations of radiation sickness and the hematological disturbances were somewhat more severe in the animals of the control group than in the rats undergoing medullary puncture. The number of control rats surviving until the 30th day was 56%, compared with a mean value of 79% for the rats of the experimental groups (see Table 1).

In series III the clinical manifestations of radiation sickness were also more severe in the animals of the control group. By the 30th day after irradiation 4 of the 10 control rats had died, but all the experimental animals were alive. The dynamics of the hematological changes was the same in the rats of both groups.

The cause of the decrease in the erythrocyte count in the blood of healthy animals after removal of marrow is unknown; a similar phenomenon, but in a more severe form, has been observed by other authors [1, 2]. By itself, the removal of some of the erythropoietic elements from the hemopoietic tissue cannot give rise to anemia, bearing in mind the long life span of the erythrocytes, developing in the course of a few days. The blood loss accompanying puncture was slight (the volume of material aspirated did not exceed 1% of the animal's total blood volume), and could not cause anemia as a result of subsequent destruction of the erythrocytes [5]. Perhaps the stimulation of the interoceptors of the bone marrow and destruction of marrow cells accompanying puncture could cause action of blood destruction, giving rise to anemia despite the intensified erythropoiesis. The fact that marrow extraction did not impair the hemopoiesis of the irradiated animals was evidently attributable to the action of mechanisms, by marrow puncture after irradiation, having a beneficial effect on the state of the animal. Injection of the needle into the medullary canal, for example, is accompanied by partial damage to the structure of the marrow and by liberation of many hemopoietic cells into the blood stream. These cells are taken by the blood flow throughout the body, and if they settle in other organs they may find more favorable conditions for their existence and produce extramedullary foci of hemopoiesis. The appearance of extramedullary hemopoiesis is demonstrated, in particular, by growth of macroscopically visible colonies on the spleen of irradiated mice [10, 11]. Such a mechanism is, of course, regarded in this case as only a possibility, and the participitation of other processes is not ruled out.

LITERATURE CITED

1. M. A. Kotovshchikova et al., Probl. Gematol., No. 10, 35 (1963).
2. K. T. Nikolau et al., Probl. Gematol., No. 2, 38 (1962).
3. M. O. Raushenbakh and M. P. Khokhlova, Med. Radiol., No. 5, 67 (1960).
4. G. S. Strelin et al., Radiobiologiya, 2, No. 2, 561 (1962).
5. Ya. G. Uzhanskii, Arkh. Pat., No. 6, 29 (1956).
6. I. L. Chertkov, M. N. Novikova, L. S. Rogacheva, et al., Med. Radiol., No. 6, 51 (1963).
7. O. Costacel and J. Corneci, In the book: Proceedings of the 2nd United Nations International Conference on the Peaceful Uses of Atomic Energy, Geneve, 26, 108 (1958).
8. G. Mathe, J. L. Amiel, L. Schwarzenberg, et al., Blood, 22, 44 (1963).
9. G. Mathe, Med. Radiol., No. 8, 32 (1965).
10. E. A. McCulloch and J. E. Till, Radiat. Res., 13, 115 (1960).
11. J. E. Till and E. A. McCulloch, Ibid., 14, 213 (1961).