RESPONSE OF COLONY-FORMING BONE MARROW CELLS OF IRRADIATED MICE TO SOME PHLOGOGENIC FACTORS

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Experiments of irradiated mice (600 and 800 R) showed that the numbers of colonies in the spleen is increased under the influence of phlogogenic factors and their morphological composition is changed. It is suggested that under the influence of a noxious agent a "hypothetical factor" causing the above-mentioned changes is formed in the skin.

KEY WORDS: colony-forming cells; inflammation.

In the irradiated individual the character of an inflammatory process is substantially changed on account of inhibition of the exudative and cellular components [4, 8]. Weakening of the cellular reaction ultimately leads to delay or disturbance of tissue regeneration [2, 5, 8]. An inflammatory focus in an irradiated individual may thus be an additional source for the entry of products of cell breakdown and of newly formed biologically active substances into the blood stream. This aggravates the disturbance of hematopoiesis in the irradiated individual [3, 10] and probably has an additional unfavorable effect on hematopoietic stem cells.

It was accordingly decided to study the ability of hematopoietic stem cells to form colonies in the spleen of irradiated mice exposed to the action of certain physical and chemical phlogogenic factors.

EXPERIMENTAL METHOD

Experiments were carried out on 230 sexually mature albino laboratory mice weighing 20-24 g, irradiated with ^{60}Co γ-rays in a dose of 600 or 800 R (dose rate 21 R/sec). There were three series of investigations. In series I, on the animals of group 1, a thermal burn was inflicted on the previously (2-3 days beforehand) depilated dorsal skin by application of a metal cylinder, with an area of 3 cm^2, heated to 103°C for 5 sec. The animals of group 2 received a subcutaneous injection of 0.1 ml xylene in the dorsal region, and the animals of group 3 a similar injection of 0.1 ml turpentine. In the experiments of series II, 24 h after irradiation in a dose of 800 R burned or unburned animals received an intravenous injection of 10^5 isologous karyocytes from healthy animals. In the experiments of series III the animals were given four intraperitoneal injections (once 1 h before irradiation and at intervals of 15 min thereafter) of 5-hydroxytryptophan (50 mg/kg) or serotonin (0.6 mg/kg). In

TABLE 1. Effect of Phlogogenic Factors on Weight of Spleen and Number and Morphological Composition of Endogenous Colonies in Irradiated Mice (M ± m)

Experimental conditions	Wt. of spleen mg/10 g body weight	No. of colonies per spleen	Composition of colonies, %				
			erythroid	granulocytic	megakaryocytic	undifferentiated	mixed
Irradiation 600 R	20,5±1,3	4,5±0,3	47,1±5,3	16,1±3,9	19,5±4,2	14,9±3,8	2,3±1,6
Irradiation and burns	28,0±1,4*	16,0±3,0*	38,0±4,0	26,7±3,6†	6,7±2,1*	15,3±2,9	13,3±2,8*
Irradiation and turpentine	33,0±2,6*	14,9±4,8*	25,0±4,0*	22,4±3,9†	6,0±2,2*	6,0±2,2*	40,5±4,6*
Irradiation and xylene	31,0±4,9*	9,5±5,0	33,6±3,9	20,1±3,8‡	9,6±2,9	12,5±3,2	24,0±4,2*

*Differences from control significant ($P < 0.05$).

†All colonies eosinophilic.

‡Composition of colonies: neutrophilic 8.6 ± 2.8; eosinophilic 11.5 ± 3.0.

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TABLE 2. Effect of Burns and Mediators of Inflammation on Weight of Spleen and Number of Colonies of Irradiated Mice (M ± m)

Experimental conditions	Wt. of spleen mg/10 g body weight	No. of colonies per spleen
Exogenous test		
irradiation 800 R	16,0±1,5	22,3±5,0
irradiation and burns	26,0±1,6*	21,0±7,6
Endogenous test		
irradiation 600 R	20,5±1,3	2,3±0,8
irradiation 600 R + 5-hydroxytryptophan	20,6±2,9	2,0±0,8
irradiation 600 R and serotonin	20,3±2,1	2,5±1,1

*Differences from control significant ($P<0,05$).

all cases animals subjected only to irradiation were used as the control. The animals were killed 8 days later, the spleen removed and weighed, and the number and morphological composition of the colonies were determined as recommended in [7, 11].

EXPERIMENTAL RESULTS

The results are summarized in Tables 1 and 2.

As Table 1 shows, under the influence of injury to the skin by a high temperature, turpentine, or xylene, there was a marked increase in the weight of the spleen and in the number of colonies. The additional extremal factors evidently led to migration of stem cells from the bone marrow or to their increased uptake by the spleen.

The inflammatory agent caused considerable changes not only in the number of colonies, but also in their morphological composition. This was manifested most clearly after irradiation and injection of turpentine. The relative number of erythroids, megakaryocytic, and undifferentiated colonies was found to be reduced whereas the number of colonies of mixed type was increased. Furthermore, among the colonies of granulocytic type, most were colonies of eosinophils (no colonies of this type were found in the control). In the remaining cases the changes were similar.

After transplantation of isologous bone marrow (Table 2) the number of colonies in the spleen of the mice of the experimental and control groups was roughly the same. It was accordingly concluded that the considerable increase in the number of colonies in the experiments of series I was due, not to increased ability of the spleen to retain stem cells, but to their migration from the bone marrow.

It might be supposed that the increase in the number of colonies in the spleen was connected with the effect of biologically active substances (histamine, serotonin) entering the blood stream from the mast cells of the skin and connective tissue of the damaged region, all with activation of the pituitary-adrenal system and a consequent rise in the blood levels of various hormones [6, 9]. However, experiments showed (Table 2) that neither serotonin nor histamine had any significant effect on the number of colonies, and stimulation of the adrenal cortex is known [1] to inhibit the migration of colony-forming cells from the bone marrow. The increase in the number of colonies observed in the present experiments and the change in their morphological composition were evidently due to the fact that, under the influence of the noxious agent, a "hypothetical factor" is formed, leading to increased migration of stem cells from the bone marrow and modification of the pathway of their differentiation. However, further investigations are necessary to reveal the nature of this factor and the mechanism of its action.

LITERATURE CITED

1. G. I. Bezin, R. M. Khaitov, B. B. Moroz, et al., Radiobiologiya, No. 2, 193 (1975).
2. L. B. Berlin, "Regeneration of the epidermis after burns and transplantation," Doctoral Dissertation, Leningrad (1962).

3. L. G. Vasil'eva, in: Burns. Pathogenesis, Clinical Picture, and Treatment. Proceedings of a Scientific Conference on the Problem of Burns [in Russian], Leningrad (1960), p. 281.
4. V. G. Garshin, Vestn. Rentgenol., 19, 355 (1938).
5. K. K. Zaitseva, "Thermal burns of the skin and free skin autografting in acute radiation sickness," Candidate's Dissertation, Leningrad (1960).
6. V. I. Kandror, The Pituitary and Adrenals in Radiation Injuries [in Russian], Moscow (1965).
7. A. G. Konoplyannikov, Technical Recommendations for Determination of the Number of Hematopoietic Colony-Forming Units (CFU) by Tests of Exogenous and Endogenous Splenic Colonies [in Russian].
8. N. A. Kraevskii and A. E. Ivanov, Arkh. Patol., No. 8, 3 (1963).
9. B. B. Moroz and I. N. Kendysh, The Radiobiological Effect and Endocrine Factors [in Russian], Moscow (1975).
10. N. A. Fedorov, S. V. Skurkovich, E. V. Shamshina, et al., in: All-Union Scientific Conference on Combined Radiation Injuries [in Russian], Moscow (1958), p. 49.
11. J. L. Curry and J. J. Trentin, Develop. Biol., 15, 395 (1967).

GENESIS OF AMNESIAS INDUCED BY ELECTROCONVULSIVE SHOCK

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The effect of a series of repeated electric shocks (ES) on preservation of conditioned reflexes and on the concentration of free amino acids (AA) in the rat brain was studied. Under the influence of repeated ES, the first of which was applied 24 h after irradiation, the rats developed amnesia. The excitatory AA concentration in the brain was unchanged, but the concentrations of inhibitory AA and also of phenylalanine and tyrosine fell sharply. The AA concentration in the blood plasma rose. It is suggested that the onset of amnesia was due to a change in the balance between excitatory and inhibitory AA in favor of the former. A definite role in the genesis of these disturbances may be played by changes in the functional state of the blood-brain barrier.

KEY WORDS: amnesia; free amino acids in the brain; blood-brain barrier.

Despite numerous investigations into the nature of retrograde amnesia induced by electroconvulsive shock (ECS) the genesis of this phenomenon is still largely unexplained [6]. Nevertheless, the elucidation of the mechanism of development of amnesias induced by ECS is of great importance not only for the solution of many fundamental problems in the neurobiology of memory, but also for the further theoretical study and effective use of electroconvulsive therapy in clinical practice. One effective way of studying the mechanisms of amnesias induced by ECS is the investigation of metabolic changes in the brain. In this respect the investigation of the content of free amino acids (AA) is of considerable interest, for besides their participation in protein metabolism, AA perform the role of neuromediators and participate in the synthesis of the "classical" neuromediators. One of us (L.G.P.) showed previously that a single ECS, causing retrograde amnesia, leads to marked changes in the absolute content and relative proportions of AA, detectable both at once and, in particular, 24 h after ECS [3]. It was suggested that the most likely cause of the retrograde amnesia is a disturbance of engram recall.

The object of the present investigation was to study the effect of, not one, but repeated ECS on preservation of the temporary connections and on the AA content in the brain. To test the hypothesis that ECS affects predominantly the operation of recall, the first of a

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