CHANGES IN HEMATOPOIESIS IN RATS AFTER DESTRUCTION OF THE POSTERIOR HYPOTHALAMIC NUCLEI

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The effect of bilateral electrical coagulation of the posterior (nuclei of the mammillary body and the posterior hypothalamic nucleus) part of the hypothalamus on hematopoiesis was studied in rats. Inhibition of erythropoiesis was observed for 28 days after destruction of the nuclei of the posterior hypothalamus: a decrease in proliferative activity and a percentage of erythroid cells in the bone marrow, a decrease in the number of erythrocytes and hematocrit index and an increase in the number of reticulocytes in the peripheral blood. Meanwhile stimulation of granulocytopoiesis was observed: an increase in the proliferative activity and percentage of immature granulocytes in the animals' bone marrow.

KEY WORDS: hypothalamus; erythrocytes; reticulocytes; hematocrit; leukocytes; leukocytic formula; bone marrow.

Despite numerous investigations into the role of the nervous system in the regulation of hematopoiesis the problem of the influence of the nervous system on hematopoiesis is still far from solution. Moreover, there is no clear idea of the importance of the hypothalamus as a regulator of automatic function.

Evidence has now been obtained to suggest that the hypothalamus plays a role in the regulation of the blood system. Investigations have demonstrated conclusively the influence of the hypothalamus (mainly its posterior part) on the erythrocytic composition of the blood [2, 3, 5-8]. The effect of the hypothalamus of the morphological composition of the bone marrow and, in particular, on leukopoiesis, has been inadequately studied.

In the investigation described below the effect of destruction of the posterior hypothalamic nuclei on the morphological composition of the peripheral blood and bone marrow was studied at different times after the operation.

EXPERIMENTAL METHODS

Experiments were carried out on 37 male Wistar rats weighing 250-350 g (20 experimental and 17 control animals).

Under pentobarbital anesthesia the nuclei in the posterior part of the hypothalamus (nuclei of the mammillary body and posterior hypothalamic nucleus) were destroyed bilaterally. Platinum or nichrome electrodes 100 μ in diameter were used. The coordinates were taken from a stereotaxic atlas of the rat's brain [4]. Electrical coagulation was carried out with a direct current of 2 mA for 5 sec.

Before the operation, daily during the first 3 days after the operation, and subsequently once a week for 14-28 days, blood was taken from the caudal vein of the animals to determine the erythrocyte, leukocyte, and reticulocyte counts, the hematocrit index, and the leukocytic formula. Bone marrow was taken from the long bones before the operation and on the 1st, 3rd, 7th, 14th, 21st, and 28th days after the operation.

After the end of the experiment the vessels of the rat's brain were perfused initially with physiological saline, and later with a 10% solution of neutral formalin for 2 weeks to 1 month, and then embedded in gelatin on increasing concentration (from 12 to 24%). Serial sections 25-30 μ in thickness in the frontal plane of the

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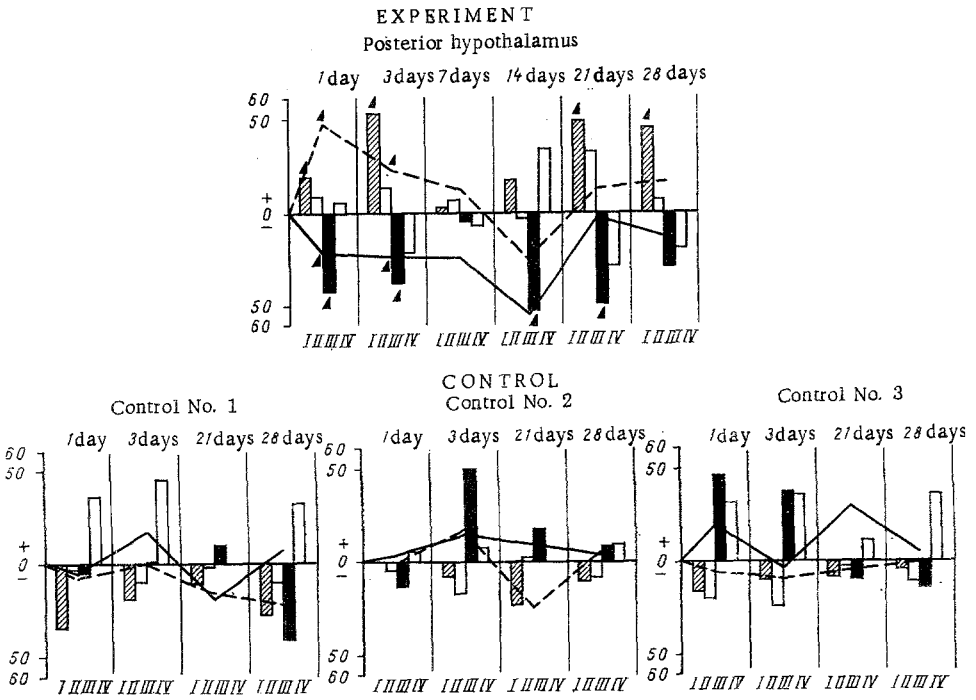

Fig. 1. Changes in morphological composition of bone marrow of rats during 28 days after destruction of part of posterior hypothalamus. I) Immature granulocytes; II) mature granulocytes; III) erythroid cells; IV) lymphoid cells. Continuous line shows mitotic index of erythroblasts; broken line the mitotic index of granulocytes. Abscissa, time (in days); ordinate, % of initial level.

brain were cut on a freezing microtome. To detect the focus of destruction, the sections were stained by Nissl's method.

Control tests were carried out on three groups of animals: control No. 1) intact animals after general anesthesia; control No. 2) animals with electrodes introduced into the nuclei of the posterior hypothalamus but without electrical coagulation; control No. 3) animals in which basal structures outside the zone of the hypothalamus (the lateral geniculate body) were destroyed.

The numerical results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

During the first 14 days after destruction of nuclei of the posterior part of the hypothalamus the number of erythrocytes in the peripheral blood fell. The erythocyte count reached a minimum on the 2nd day after the operation, when it was reduced on average by 780,000 ($P < 0.02$). In controls Nos. 1 and 3 a transient or very slight decrease (by a maximum of 29,000 on the 3rd day) in the erythrocyte count was observed. In control No. 2 the decrease in the erythrocyte count continued for 2 weeks, was not significant, and was maximal (by 690,000) on the 3rd and 7th days of observation.

At all times after the operation the reticulocyte count in the peripheral blood was increased in the experimental rats, and the increase was greatest one day after the operation: on average by 23% ($P < 0.05$). No regular or significant changes in the reticulocyte counts were found in the control animals.

In animals with destruction in the posterior hypothalamus the hematocrit index was reduced for 28 days, and the decrease was maximal on the 2nd and 3rd days after the operation, on average by 11% ($P < 0.01$). A less marked decrease, not significant, in the hematocrit index was found in controls Nos. 2 and 3 during the first 3 days of observation.

In both the experimental and control rats leukocytosis was observed at all times of observation on account of an increase in the absolute numbers of granulocytes and lymphocytes. Compared with the control animals, the increase in the number of granulocytes on the first day after the operation was greater in the experimental animals and the shift of the leukocytic formula to the left was more marked. For instance, the

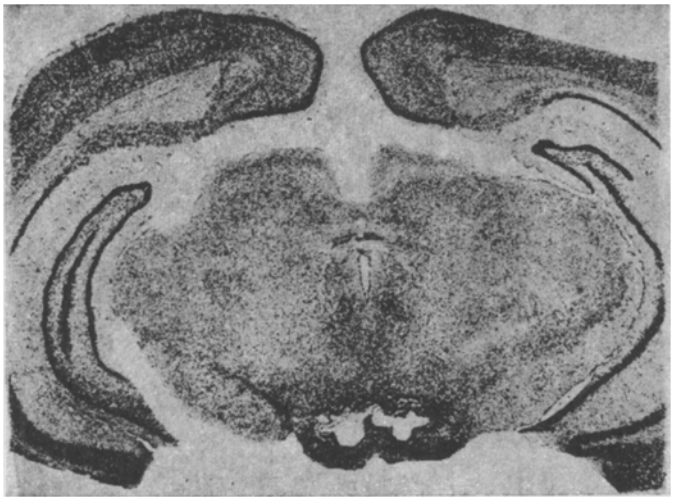

Fig. 2. Frontal section through brain of rat No. 50-4 with bilateral focus of destruction in medial and supramammillary nuclei of the mammillary body. Nissl's stain, 24×.

number of stab neutrophils in the rats one day after destruction of the posterior hypothalamus was significantly increased almost threefold, and it remained two or three times higher than initially at all times of observation. No marked changes in the number of these cells could be found in the control animals of all groups. The absolute number of polymorphs also was doubled in the experimental animals on the first day after the operation ($P < 0.05$). The absolute number of polymorphs in control No. 1 was increased at this time by only 580 cells ($P < 0.05$), and in controls Nos. 2 and 3, by 50-100% on the 21st and 28th days.

Marked inhibition of erythropoiesis was observed in the bone marrow of the experimental rats (Fig. 1). The number of erythroid cells on the first day after destruction of the posterior part of the hypothalamus was reduced on average by 44% ($P < 0.01$) and remained below the initial level until the end of observation. The number of mitoses in the erythroblasts was reduced. The maximal decrease in proliferative activity of the erythroblasts (on average by 23%; $P < 0.05$) was observed on the third day after the operation.

On the first day after destruction of part of the posterior hypothalamus the number of mitoses in granulocytes was significantly increased (on average by 41%) and it remained higher than initially at all times of observation except the 14th day. The number of immature granulocytes at all times after the operation was increased, maximally on the 3rd day, on average by 54% ($P < 0.001$).

No significant changes in the number of mature granulocytes and lymphocytes were observed after destruction of part of the posterior hypothalamus.

No analogous changes in the number of bone marrow cells were observed in the control groups at the same times of observation (Fig 1).

The effect of destruction of nuclei of the anterior (anterior hypothalamic nucleus) and middle (dorsomedial and ventromedial nuclei) parts of the hypothalamus on hematopoiesis also was studied. In experiments with destruction of the anterior hypothalamus no marked changes were observed either in the erythrocytic composition of the peripheral blood or in the morphological composition of the bone marrow. After injury to nuclei of the middle part of the hypothalamus the erythrocyte count in the peripheral blood was reduced and the reticulocyte count increased. An increase in the number of immature granulocytes in the bone marrow was observed at all times of observation, whereas the number of erythroid cells was reduced on the 1st and 3rd days after the operation on average by 21 and 16% respectively. However, the changes observed in the bone marrow after destruction of nuclei of the middle part of the hypothalamus were not significant.

Considering that changes in the erythrocyte and leukocyte counts in the peripheral blood may be secondary in character, during assessment of the results most attention was paid to changes in the morphological composition of the bone marrow.

Distinct changes in hematopoiesis were thus found after destruction of part of the posterior hypothalamus. Proliferative activity of the erythroid cells and their relative percentage in the bone marrow were reduced. The reticulocyte count was increased in the peripheral blood. Destruction of this zone of the hypothalamus evidently leads to delay of maturation of reticulocytes.

Comparison of the results of the hematological investigation with those of morphological examination of the focus of injury in the posterior hypothalamus (Fig. 2) showed that if the lesion was mainly in the mammillary body the inhibition of erythropoiesis was well marked and lasted longer. In these cases the number of erythroid cells fell by 70-80% on the 1st and 3rd days after electrical coagulation. These results correlate with those of electrophysiological investigations [1, 3] which demonstrated the influence of nuclei in the posterior hypothalamus on erythropoiesis. This dynamic study of the morphological composition of the bone marrow over a period of 28 days revealed considerable and prolonged inhibition of erythropoiesis after destruction of nuclei in the posterior hypothalamus. Besides inhibition of erythropoiesis, after destruction of the posterior part of the hypothalamus proliferative activity and the number of immature granulocytes in the bone marrow were increased. Stimulation of granulocytopoiesis after destruction of the nuclei in this part of the hypothalamus evidently is not due to necrotic changes actually in the brain tissue itself, for electrical coagulation of basal structures in the brain lying outside the hypothalamus in control animals (control No. 3) did not lead to the corresponding changes.

It can thus be concluded from this investigation that the posterior part of the hypothalamus participates in the regulation of erythropoiesis and also, possibly, of leukopoiesis.

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