EMIGRATION OF CELLS FROM THE SPLEEN UNDER NORMAL CONDITIONS AND IN STRESS

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According to determination of the mitotic activity of the cells, emigration of lymphocytes of the spleen under normal conditions amounts to $0.1 \cdot 10^9$ daily (10% of the total number of cells). In response to stress produced by electrical stimulation and immobilization, emigration of lymphocytes from the spleen is increased by 7-8 times. The decrease in the number of cells in the spleen after both these procedures is due to the extent of 90% to increased emigration and of 10% to a decrease in the formation of new lymphocytes.

One of the most rapid responses of the spleen is its atrophy during stress. Previous work [1] has shown that destruction and lysis of the lymphocytes play by no means the dominant role in the decrease in number of cells in the spleen in response to stress. Consequently, the question of the importance of cell migration in the decrease in the cell population of the spleen must be considered. This empyting of the spleen may be due to natural or to increased emigration of cells from it. Everything depends on the extent to which cells emigrate under normal conditions and the depth of inhibition of proliferation.

In the present investigation a quantitative analysis was made of emigration of cells from the spleen under normal conditions and in response to stress, and the result showed that emigration of cells in stress is sharply increased.

EXPERIMENTAL METHOD

Experiments were carried out on 156 10-week Wistar rats of both sexes weighing 160-180 g. The state of stress was reproduced by measured electrical stimulation [2] (duration of the series of pulses 3 sec, interval between series 1.5 min, current 2.5 mA, frequency 2000 Hz, duration of stimulation 3 h). Parallel tests were carried out on rats immobilized for 6 h on their backs. The rats were decapitated. The number of cells in the spleen was determined as follows: the organs were isolated and placed in 5% acetic acid solution, the volume of which was made up to 10 ml. The spleen was reduced to a suspension of single cells by means of a homogenizer with a glass pestle. The cell suspension was drawn into a pipette for counting leukocytes (dilution 1:10). Cells were counted in five large squares of a Goryaev's chamber. The formula for the calculation was:

$$x = a/2 \cdot 10^7$$
.

where x is the number of cells in the organ and a the number of cells in the Goryaev's chamber.

The mitotic activity of the spleen cells was studied in films obtained from homogenates of pieces of the organs stained by the method of Kellner and Stokinger [4].

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TABLE 1. Changes in Mitotic Index (in promille) in the Spleen after Electrical Stimulation for 3 h and Immobilization for 6 h $(M\pm m)$

| Type of procedure | Control | Time (in h) after beginning of procedure | | | | |
|------------------------|-----------------|--|------------------|---------------------------|------------------|---------------------------|
| | | 3 | 6 | 9 | 12 | 24 |
| Electrical stimulation | $1,3\pm0,22$ | | | | | |
| Immobilization | 2,0±0,43 (8) | $1,0\pm0,28$ (5) | 0,81±0,12 (5) | 0,80 <u>±</u> 0,09 (5) | 0,75±0,19 (5) | 0,41 <u>+</u> 0,16 (6) |

Note. Number of animals given in parentheses.

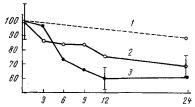


Fig. 1. Content of cells in spleen:
1) during total suppression of mitosis (theoretical curve); 2) after electrical stimulation for 3 h; and
3) after immobilization for 6 h. Abscissa, time in hours after beginning of procedure; ordinate, number of cells in spleen (in %).

EXPERIMENTAL RESULTS AND DISCUSSION

Electrical stimulation for 3 h and immobilization of the animals for 6 h led to a marked increase in the number of cells in the spleen (Fig. 1). The mitotic activity of the spleen cells also was reduced 3-6 h after the beginning of exposure to both procedures (Table 1).

Under near-physiological conditions the spleen is a cell population in equilibrium. The stability of the number of cells in the spleen is maintained by equality between the intensity of proliferation and of migration of cells from the organ. Should either of these two mutually balanced processes decrease in intensity, the number of cells in the organ will either increase or decrease at a rate corresponding to the rate at which new cells are formed or at which they leave the organ.

The number of cells leaving the spleen can be determined by simple calculations based on the mitotic index and the duration of mitosis, using the formula:

$$n \cdot I \cdot t/\tau_M$$

where n is the number of cells in the organ, I the mitotic index, $\tau_{\rm M}$ the duration of mitosis, which is approximately 20 min [3], and t the time.

Estimation of the mitotic activity of the cells in the spleen of the control animals showed that the emigration of lymphocytes from the spleen under normal conditions is $0.1 \cdot 10^9$ cells/day (10% of the initial number present). Even if the inhibition of proliferation in the spleen during stress were total and the decrease in the total number of cells in the organ were determined entirely by natural elimination (destruction of the cells could not be found after either procedure), the number of cells in the spleen would only be reduced by 5-7% 12 h after the beginning of immobilization and electrical stimulation. In fact, 9-12 h after the beginning of the procedures the number of cells in the organ was 60 and 70%, respectively, of the original level, consequently, emigration of lymphocytes from the spleen under these conditions of stress was increased by 7-8 times above the normal level.

The decrease in the number of cells in the spleen after exposure for 3 h to electrical stimulation and for 6 h to immobilization was 90% caused by increased emigration and 10% caused by decreased formation of new lymphocytes.

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