

CONTENT OF ACID AND ALKALINE PHOSPHATASES  
IN LYMPHOCYTES OF PERIPHERAL BLOOD  
AND HEMATOPOIETIC ORGANS OF INTACT RATS AND MICE

E. D. Gol'dberg, G. V. Karpova,  
E. V. Melik-Gaikazyan, and G. N. Pakhryaeva

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The content of acid and alkaline phosphatases was studied in the lymphocytes of the peripheral blood, bone marrow, and lymphoid organs (thymus, spleen, lymph node) of intact noninbred rats. The mean percentage of cells containing the enzymes was determined. Their organ and species characteristics are described.

KEY WORDS: lymphocytes; acid and alkaline phosphatase; bone marrow; lymphoid organs.

Data in the literature on the phosphatase content in rat and mouse lymphocytes are few in number and contradictory in nature. It is generally accepted [1, 3] that alkaline phosphatase cannot be detected histochemically in the lymphocytes and that acid phosphatase activity varies from weak to moderate. Meanwhile, alkaline phosphatase detectable by the azo-coupling method with substituted naphthol has been found [7] in the peripheral blood lymphocytes of mice, guinea pigs, and man. In rats the reaction for alkaline phosphatase in the lymphocytes was negative. On electron-cytochemical investigation, alkaline phosphatase activity has been found in guinea pig thymocytes [9].

The determination of the concentration of acid and alkaline phosphatases is of considerable interest in connection with new data showing that alkaline phosphatase is a marker of B lymphocytes and acid phosphatase a marker of the T cells [4, 8].

#### EXPERIMENTAL METHOD

This paper gives the results of investigation of the content of acid and alkaline phosphatases in lymphocytes of the peripheral blood, bone marrow, and lymphoid organs (thymus, spleen, lymph node) of intact, noninbred, sexually mature animals of both sexes (55 rats and 20 mice). Thin films of bone marrow and lymphoid organs were prepared, using serum of the same animals, whose peripheral blood was obtained from the caudal vein, as the diluting fluid. The content of acid phosphatase was determined as in [5] and of alkaline phosphatase by the azo-coupling method in the modification described in [2]. The results were assessed by a semiquantitative method [6], with determination of the percentage of lymphocytes containing the enzyme.

#### EXPERIMENTAL RESULTS

The highest percentage of lymphocytes giving a positive reaction for acid phosphatase in rats was found in the thymus (Table 1). Next followed the spleen and lymph node, in which it was equally high. About 60% of peripheral blood lymphocytes contained granules of acid phosphatase. The percentage of bone marrow cells containing the enzyme was the lowest of all. It should be noted that activity of the enzyme in the lymphocytes of nearly all organs tested was low. It was minimal in bone-marrow and lymph-node lymphocytes, and in the thymus there was a small percentage of thymocytes (about 3) with a moderate enzyme content. Only in the spleen and peripheral blood were solitary lymphocytes with high acid phosphatase activity found.

In mice the lymphocytes were characterized by extremely low acid phosphatase activity (homogeneous staining of one or two granules). Although the percentage of lymphocytes giving a positive reaction for the enzyme was lower in mice than in rats, the character of distribution of the enzyme among the organs was the

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TABLE 1. Number of Lymphocytes (in %) Positive for Acid and Alkaline Phosphatase in Intact Animals ( $M \pm m$ )

| Animals              | Bone marrow     | Peripheral blood | Thymus         | Spleen         | Lymph node     |
|----------------------|-----------------|------------------|----------------|----------------|----------------|
| Acid phosphatase     |                 |                  |                |                |                |
| Rats                 | 41,4 $\pm$ 2,8  | 59,1 $\pm$ 3,2   | 90,4 $\pm$ 1,5 | 70,7 $\pm$ 2,8 | 71,4 $\pm$ 2,7 |
| Mice                 | 13,3 $\pm$ 1,7  | 28,3 $\pm$ 3,9   | 76,2 $\pm$ 5,1 | 49,3 $\pm$ 8,1 | 48,2 $\pm$ 5,4 |
| Alkaline phosphatase |                 |                  |                |                |                |
| Rats                 | 26,0 $\pm$ 10,5 | 0,7 $\pm$ 0,3    | 4,0 $\pm$ 0,8  | 8,6 $\pm$ 2,4  | 6,8 $\pm$ 2,4  |
| Mice                 | 21,1 $\pm$ 8,3  | 1,6 $\pm$ 0,7    | 14,8 $\pm$ 5,6 | 8,3 $\pm$ 1,2  | 17,0 $\pm$ 3,9 |

same. For instance, the highest percentage of lymphocytes with a positive reaction for acid phosphatase was found in the thymus, followed by the spleen and lymph node, and the lowest percentage in the peripheral blood and bone marrow.

As regards alkaline phosphatase, both in rats and in mice lymphocytes were found in all organs studied with either extremely low enzyme activity in the cytoplasm of most cells (staining of one or two small granules), or less frequently, moderate activity and only isolated cells (mainly bone marrow lymphocytes) with a high content of the enzyme. The highest percentage of lymphocytes containing alkaline phosphatase was found in the bone marrow of rats and mice. Enzyme-positive lymphocytes were extremely rare in the peripheral blood of the rats. Their number in the spleen and lymph node was a little higher than in the thymus. In mice, the percentage of lymphocytes containing alkaline phosphatase granules in the lymphoid organs and peripheral blood was much higher than in rats (Table 1).

The study of the lymphocytes of the peripheral blood, bone marrow, and lymphocytes thus showed that a certain percentage of them contains low or moderate enzyme activity in the form of discrete granules. The percentage of lymphocytes containing acid phosphatase is lower in mice than in rats. The largest number of cells giving a positive reaction for acid phosphatase is found in the thymus, spleen, and lymph node, and the smallest number in the bone marrow. The percentage of lymphocytes containing alkaline phosphatase was appreciably higher in the lymphoid organs and peripheral blood of mice.

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