SOME CYTOCHEMICAL AND FUNCTIONAL INDICES OF THE STATE OF THE MICROPHAGES IN RABBITS WITH EXPERIMENTAL ALLERGIC ARTHRITIS

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Regular changes in the activity of acid and alkaline phosphatases and myeloperoxidase and in the glycogen concentration together with a decrease in phagocytic activity of the microphages (pseudoeosinophilic leukocytes) were found in rabbits with allergic arthritis. The observed changes were perhaps linked with the development of degenerative changes in the microphages following contact between these cells and antigen. The development of experimental allergic arthritis was shown to be accompanied by dysfunction of the microphages, with a consequent decrease in the protective properties of these cells.

KEY WORDS: *experimental allergic arthritis; microphages; cytochemical and functional indices.*

Previous investigations showed [3, 4] that in rabbits with experimental allergic arthritis changes arise in the heart muscle, the activity of the properdin system is lowered, and degranulation of basophilic leukocytes is observed.

The object of the present investigation was to study enzymic and phagocytic activity of the peripheral blood microphages in the course of development of experimental allergic arthritis in rabbits.

EXPERIMENTAL METHOD

Allergic arthritis was induced in animals previously sensitized with horse serum by injection of the reacting dose of the same antigen into a joint. Peripheral blood microphages were investigated in 40 rabbits in the initial state and during the development of arthritis. Changes in the joint were monitored by measurement of the joint in two projections and also histologically and radiologically. To assess the changes in phagocytic power of the microphages, against Escherichia coli (strain K12S), Hamburger's index (HI), the phagocytic number (PN), and the completeness of phagocytosis (by the method of Berman and Slavskaya [2]) were determined. To assess the digestive power, the digestive index (DI) and the percentage of leukocytes completing phagocytosis (LCP) were calculated. Activity of alkaline (ALP) and acid (ACP) phosphatases was demonstrated by the azo-coupling [7, 10], myeloperoxidase (MP) activity by Sato's method [15], and glycogen (G) by Shabadash's method [9]. Kaplow's principle [12] was followed during assessment of the glycogen content and enzyme activity. In control tests the activity of the enzymes and glycogen content were determined under similar conditions but without addition of the bacteria. The results were subjected to analysis by variance and correlation methods.

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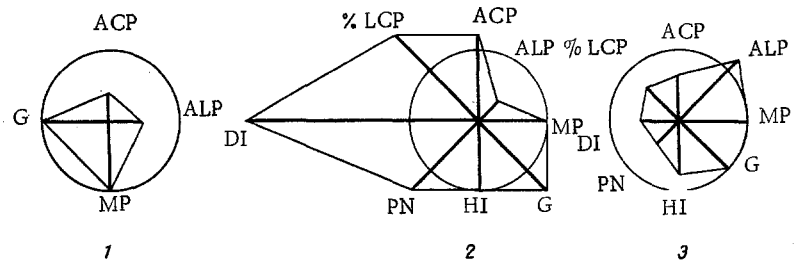

Fig. 1. Changes in cytochemical indices and phagocytic activity of blood microphages of rabbits during development of allergic arthritis: 1) reacting injection of serum in control tests; 2) final stage of phagocytosis in intact rabbits; 3) final stage of phagocytosis after reacting injection of antigen. In all cases circle describes level of cytochemical indices and functional activity of blood microphages in initial state. For meaning of symbols, see description of experimental method.

EXPERIMENTAL RESULTS

The period of sensitization was shown to be accompanied by a significant ($P < 0.01$) increase in ALP activity by 27%, an increase in the G concentration by 42%, and a decrease of 42% in ACP activity ($P < 0.001$); MP activity showed no significant change. The reacting injection of serum (antigen) led to a sharp fall in phosphatase activity and a decrease in the G concentration to the initial level; MP activity again was not significantly changed (Fig. 1).

At the height of the clinical manifestations of arthritis, i.e., on the 3rd-8th day after the reacting injection of antigen, ACP activity and the G concentration were at their initial level but the ALP activity remained low. During the recovery period ALP activity was the first index to return to normal, but MP activity fell. The changes in microphagal enzyme activity were shown to be due to repeated contact between the sensitized organism and the antigen ($F\phi > Ft$). In the original state and in the initial phase of phagocytosis MP activity decreased; both the number and the intensity of staining of the granules containing the enzyme diminished. In some bacteria engaged in phagocytosis of the bacteria, MP granules disappeared. As fragmentation of the bacteria progressed, the activity of this enzyme was restored. Attraction of the bacteria in the initial phase of phagocytosis was observed in microphages with average ACP activity. In the final stage of phagocytosis ACP activity increased, and nonviable bacteria were found more frequently in microphages with high ACP activity. In the course of digestion of the bacteria, ALP activity fell. During the course of the phagocytic reaction the G concentration rose and the staining changed from diffuse to granular. The granules accumulated more often at the periphery of the cell and evaginations were formed in some microphages. During the period of sensitization and, in particular, after the reacting injection of the antigen, the phagocytic activity of the microphages changed at the height of the clinical manifestations of arthritis (the number of microphages engaged in phagocytosis decreased, their ability to ingest and digest bacteria declined), and at this stage ALP activity in the microphages was higher but ACP and MP activity and the G concentration were lower than in the original state (Fig. 1).

The increase in ACP activity found during phagocytosis can be explained by an increase in permeability of the lysosomal membranes. Endotoxin of bacteria of the enteric group are known to possess this property [16]. The divergent character of the changes in ACP and ALP activity in the final stage of phagocytosis can probably be explained by the fact that they are contained in granules of different types, which do

not join the phagosome simultaneously [11, 14]. Changes in MP activity in the initial and final stage of phagocytosis can be understood in the light of data [8, 13] showing that the intraleukocytic MP—H_2O_2 system has a bactericidal action on ingested bacteria. The increase in the G concentration can be explained by the development of the initial features of clasmatosis, as a result of oxygen deprivation of the cell and blocking of its glycolytic enzymes [1].

Considering earlier observations that the digestive power of the microphages correlates with their phosphatase activity [5, 6] it can be concluded that the cytochemical changes in the microphages observed in allergic arthritis are the cause of the sharp fall in their phagocytic function. A decrease in the functional activity of the microphages must inevitably lead to a decline in nonspecific immunologic reactivity of the organism.

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