DEPRESSION OF THE SERUM SPECIFIC IgE ANTIBODY LEVEL IN MICE AND OF IgE PRODUCTION BY BONE MARROW CELLS

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The wide distribution of allergic atopic diseases necessitates the study of the particular immunologic mechanisms of this pathology [1]. Atopic diseases, according to ideas on "allergic break-through" [8], develop when the regulation of the IgE response is disturbed. The bone marrow is an important organ regulating the immune system [3, 6, 13]. Bone marrow cells have been shown to both stimulate and inhibit the processes of antibody formation [4]. This activity is realized through the production of humoral meditators, which act on the inductive and productive phases of the immune response [5]. How these mechanisms act on the formation and development of the IgE response is not completely clear. To study this effect we have developed a model in which the action of bone marrow cells on the IgE response has been investigated.

EXPERIMENTAL METHOD

Experiments were carried out on (CBA × C57BL/6)F_1 mice weighing 18-20 g. The IgE response was induced by intraperitoneal injection of 4-times recrystallized ovalbumin in a dose of 5 μg per mouse. Aluminum hydroxide in a dose of 5 mg per mouse, obtained from potassium alum, was used as the adjuvant. Suspensions of bone marrow cells of syngeneic donors were injected intravenously into the mice in 0.5 ml of Hanks' solution. The suspensions were preppared by the usual methods. Bone marrow cells were freed from erythrocytes by hemolysis with NH_4Cl and separation in a Ficoll density gradient: $\rho = 1.09$ [7]. Adherent cells were removed by incubation on plastic Petri dishes with an internal diameter of 100 mm, from the Leningrad Medical Polymers Factory [11]. T lymphocytes were removed by the double cytotoxic reaction with anti-Thy-1,2-serum, generously provided by B. V. Nikonenko, and by guinea pig complement [2]. The level of IgE antibodies in the mice was determined in the 1st and 2nd weeks after immunization by the passive cutaneous anaphylaxis (PCA) and heterologous adoptive transfer (HAT) methods [9, 12].

EXPERIMENTAL RESULTS

The experiments showed that bone marrow cells, in a number of (20-30) × 10^6 cells per mouse, when injected intravenously at various times after immunization, suppress the IgE response to ovalbumin. The maximal effect was achieved by injection of the cells simultaneously with immunization. This served as the basis for the use of the following model in subsequent experiments: injection of bone marrow cells in a number of (20-30) × 10^6 cells per mouse simultaneously with immunization with ovalbumin and determination of the IgE antibody level in the 1st and 2nd weeks after immunization.

The use of this model showed that the fall in the serum IgE level of the mice, determined by the PCA method, corresponded to a decrease in IgE production by the bone marrow cells, determined by the HAT method (Fig. 1). The serum IgE antibody level fell by 1.5 dilutions. This could be the result of suppression of IgE antibody production by different cells, including bone marrow cells, as is confirmed by the data in Fig. 1.

It was next shown that the suppressor action of bone marrow cells does not depend on the presence of erythrocytes, monocytes, and T lymphocytes, for the effect remained after removal of these cells from the injected suspension. It was absent in the mice in the control and after injection of thymus cells, and it was not observed after

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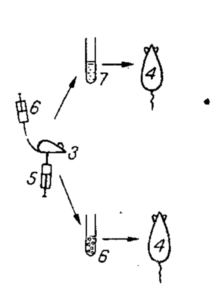

Fig. 1

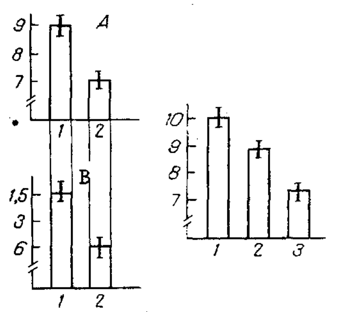

Fig. 2

Fig. 1. Lowering of serum IgE antibody level in mice during inhibition of IgE antibody production by bone marrow cells. A) Lowering of serum IgE level on injection of bone marrow cells (ordinate, $\log_2$ of titer of PCA reaction; B) inhibition of IgE production on injection of bone marrow cells (ordinate, number of bone marrow cells in HAT (in 10^6/ml); 1) control group receiving ovalbumin, 5 μg/mouse, and 0.5 ml of Hanks' solution; 2) experimental group receiving ovalbumin, 5 μg/mouse, and suspension of intact bone marrow cells in a dose of (20-30) × 10^6 cells per mouse in 0.5 ml of Hanks' solution; 3) (CBA × C57BL/6)F_1 mice; 4) noninbred male rats; 5) ovalbumin, 5 μg/mouse; 6) mouse bone marrow cells; 7) mouse blood serum.

Fig. 2. Effect of immunization with ovalbumin on suppressor activity of mouse bone marrow cells. Ordinate, $\log_2$ of titer of PCA reaction; 1) control group receiving only ovalbumin in a dose of 5 μg/mouse and Hanks' solution in a dose of 0.5 ml/mouse; 2) experimental group receiving ovalbumin in a dose of 5 μg/mouse and suspension of intact bone marrow cells in a dose of (20-30) × 10^6 cells per mouse in 0.5 ml of Hanks' solution; 3) experimental group receiving ovalbumin in a dose of 5 μg/mouse and suspension of immune bone marrow cells in a dose of (20-30) × 10^6 cells per mouse in 0.5 ml of Hanks' solution.

injection of unfractioned bone marrow cells, bone marrow cells deprived of erythrocytes, nonadherent bone marrow cells without erythrocytes, and nonadherent bone marrow cells deprived of T lymphocytes and erythrocytes. Depression of the serum IgE antibody level corresponded to depression of IgE antibody production by bone marrow cells. It can be tentatively suggested on the basis of the data on preservation of suppression of the IgE response afer removal of erythrocytes, monocytes, and T lymphocytes that the action was due to cells of the B series: suppressor B cells [14] or what are called natural suppressors [10]. Determination of the precise phenotype of the active cells will help to solve this problem finally. Since the role of bone marrow suppressors is to limit the immune response by inhibiting proliferation of precursors of antibody producers [6], it was suggested that the suppressors are most active at the stage of decline of antibody production, after the peak of the immune response. To test this hypothesis, the action of intact bone marrow cells of mice immunized with ovalbumin 4 days before removal of the bone marrow was compared (the level of IgE-antibodies in the bone marrow donors in the 4th week was in the stage of decline). The experiments showed that immune bone marrow taken from donors at the time of decline of the IgE antibody level had a stronger suppressor action than intact bone marrow (Fig. 2). This confirms the view of the role of suppressors in the mechanism of limitation of the immune response and is evidence that bone marrow cells may be among the suppressors that are involved in regulation of the IgE response.

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