Changes in Some Parameters of Immune Response under the Effect of Adrenoceptor Blockers

E. Yu. Sherstoboev, A. P. Babenko, N. V. Masnaya, and O. S. Borsuk

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We studied the effect of α- and β-adrenoceptor blockers on humoral and cell immune response in $(CBA \times C57Bl/6)F_1$ mice under normal conditions and during stress. The suppressive effect of α-adrenoceptor blocker dihydroergotamine on humoral immune response against the background of both immunization and combined effects of immobilization and injection of a thymus-dependent antigen was more pronounced compared to that of β-adrenoceptor blocker propranolol. Dihydroergotamine suppressed proliferative activity of T cells, stimulated proliferation of B cells at late terms after immunization, but inhibited it against the background of stress and antigen treatment. Propranolol stimulated proliferative activity of T and B cells at early terms after both immunization alone and stress with antigen treatment, but decreased the proliferation stimulation index of lymphoid cells at late terms after treatment (days 7-10).

Key Words: *humoral immune response; cellular immune response; α- and β-adrenoceptor blockers; immobilization stress*

The interaction between the nervous, endocrine, and immune systems is proven in various experimental and clinical studies [6,8,11,12]. Stress (*e.g.* immobilization) reduces immune reactivity of the organism [4,15]. It was found that immunobiological reaction depends on activity of neurotransmitter systems [2]. Catecholamines secreted by nerve terminals of the sympathetic nervous system modulate proliferation and differentiation of immunocompetent cells via specific receptors on their membranes [1]. However, the effects of catecholamines can vary during local responses under conditions of specific environment.

The aim of the present study was to evaluate the effects of α- and β-adrenoceptor blockers (AB) on humoral and cell immunity under normal conditions and during stress.

Institute of Pharmacology, Tomsk Research Center, Siberian Division of Russian Academy of Medical Sciences

MATERIALS AND METHODS

The experiments were carried out on 150 female $(CBA \times C57Bl/6)F_1$ mice (age 2.0-2.5 months, weight 18-20 g) obtained from Laboratory of Experimental Biomodeling, Institute of Pharmacology, Tomsk Research Center, Russian Academy of Medical Sciences). All animals except the background group (n=6) were immunized with corpuscular thymus-dependent antigen sheep erythrocytes (SE, Microgen company). SE were washed 3 times with sterile physiological saline and injected intraperitoneally (0.2 ml 15% erythrocyte suspension per animal).

For evaluation of the effect of stress on functional activity of immune cells, the mice of the experimental group were subjected to 24-h immobilization in closed cylinders before immunization. Some animals (n=48) received subcutaneous injection of α-AB dihydroergotamine (3.9 mg/kg, Galena) or β-AB propranolol (5 m/kg, Isis Pharma) 3-5 min before immobilization (experimental group)

or immunization (control group) and 6 h after the start of exposure. The drugs were dissolved in sterile physiological saline immediately before use. The mice were decapitated under ether narcosis. The material was collected on days 1, 4, 7, and 10 after immunization with SE.

The reaction of the immune system in mice receiving α- or β-AB was compared to the corresponding values in immunized animals (n=24) receiving an equivalent volume of physiological saline.

Phagocytic activity of peritoneal macrophages (PM) was determined by the intensity of incorporation of ink particles [5]. The absolute number of antibody-forming cells (AFC) in the spleen was evaluated by the method of local hemolysis [7], the level of total serum immunoglobulins was determined by the reaction of hemagglutination [3]. Cell immunity was evaluated by changes in proliferative activity of T and B cells in the blast-transformation reaction [14].

The data were processed statistically using Student's t test after verification of normal distribution.

RESULTS

The number of active phagocytes increased on days 4, 7, and 10 (Fig. 1, *a*). The index of phagocytosis decreased under these conditions (Fig. 1, *b*), which was probably related to excessive activation of PM aimed at antigen processing and production of cytokines. Treatment with α-AB in the control group significantly increased the number of active PM on day 1 of the experiment compared to mice receiving physiological saline. Then, the absolute number of PM decreased and was significantly lower than in animals receiving SE to the end of the observation period (Fig. 1, *a*). Injection of α-AB against the background of immunization increased the index of phagocytosis compared the corresponding values in immunized animals (Fig. 1, *b*). Injection of β-AB propranolol had no effect on the number of phagocytizing PM and their functional activity (Fig. 1, *a*, *b*).

After immunization with SE, the absolute number of AFC in the spleen of mice surpassed the background values throughout the experiment and peaked on day 4. The total titer of hemagglutinins peaked on day 7 after immunization. The content of AFC in the spleen and the level of total immunoglobulins in the serum of immunized mice receiving α-AB were lower than in immunized mice receiving physiological saline throughout the experimental period (Fig. 1, *c, d*). Injection of β-AB also significantly decreased the content of AFC (except day 4) and the level of total immunoglobulins in the serum of control mice (Fig. 1, *c, d*).

Injection of SE increased proliferative activity of both T and B cells on days 1, 7, and 10 (although the changes from the background values were insignificant). Injection of α-AB to immunized mice reduced proliferative activity of T cells on day 7 of the experiment and increased the index of B cell stimulation on day 10. After injection of β-AB proliferative activity of T cells decreased on days 7 and 10, B cells on day 7, and index of B cell stimulation increased on day 4 (Fig. 1, *e, f*).

Immobilization stress reduced the content of phagocytizing PM on day 4 of the experiment compared to the control group. The index of PM phagocytosis decreased similarly to the control group, but less markedly. The absorption capacity and the absolute number of phagocytizing PM in experimental mice receiving α-AB increased compared to the corresponding parameters without α-AB treatment only on day 1 of the experiment, *i.e.*, during the action of the drug. Injection of β-AB had no effect on the number of phagocytizing PM, but decreased the index of phagocytosis on days 4 and 10 (Fig. 2, *a*, *b*).

In experimental mice, the absolute number of AFC significantly decreased compared to the corresponding parameters in control animals, while the peak of AFC accumulation was shifted towards day 10 (Fig. 2, *c*). The total titer of hemagglutinins significantly decreased on days 4 and 7 (Fig. 2, *d*). AB suppressed accumulation of AFC in mouse spleen against the background of stress and antigenic stimulation throughout the observation period compared to the corresponding values obtained without AB (Fig. 2, *c*). Functional activity of AFC decreased, which manifested in lowered serum hemagglutinin titer (Fig. 2, *d*).

In the experimental group, the index of stimulation of T and B cell proliferation decreased at early terms of the experiment compared to the control values. Compensatory hyperstimulation of these cells was observed by day 7. Injection of α-AB to experimental mice reduced proliferative activity of T cells on days 7 and 10 and B cell on day 7. However, the index of B cell stimulation increased on day 4. Injection of β-AB under these conditions led to wave-like changes in proliferative activity of lymphoid cells. For instance, proliferation of T and B cells increased at early terms (days 1-4), but decreased on days 7 and 10 (Fig. 2, *e, f*).

Injection of α-AB suppressed the humoral immune response against the background of both immunization and combined use of immobilization and SE administration, but produced a stimulatory effect on functional activity of PM, although the number of phagocytizing cells decreased under these

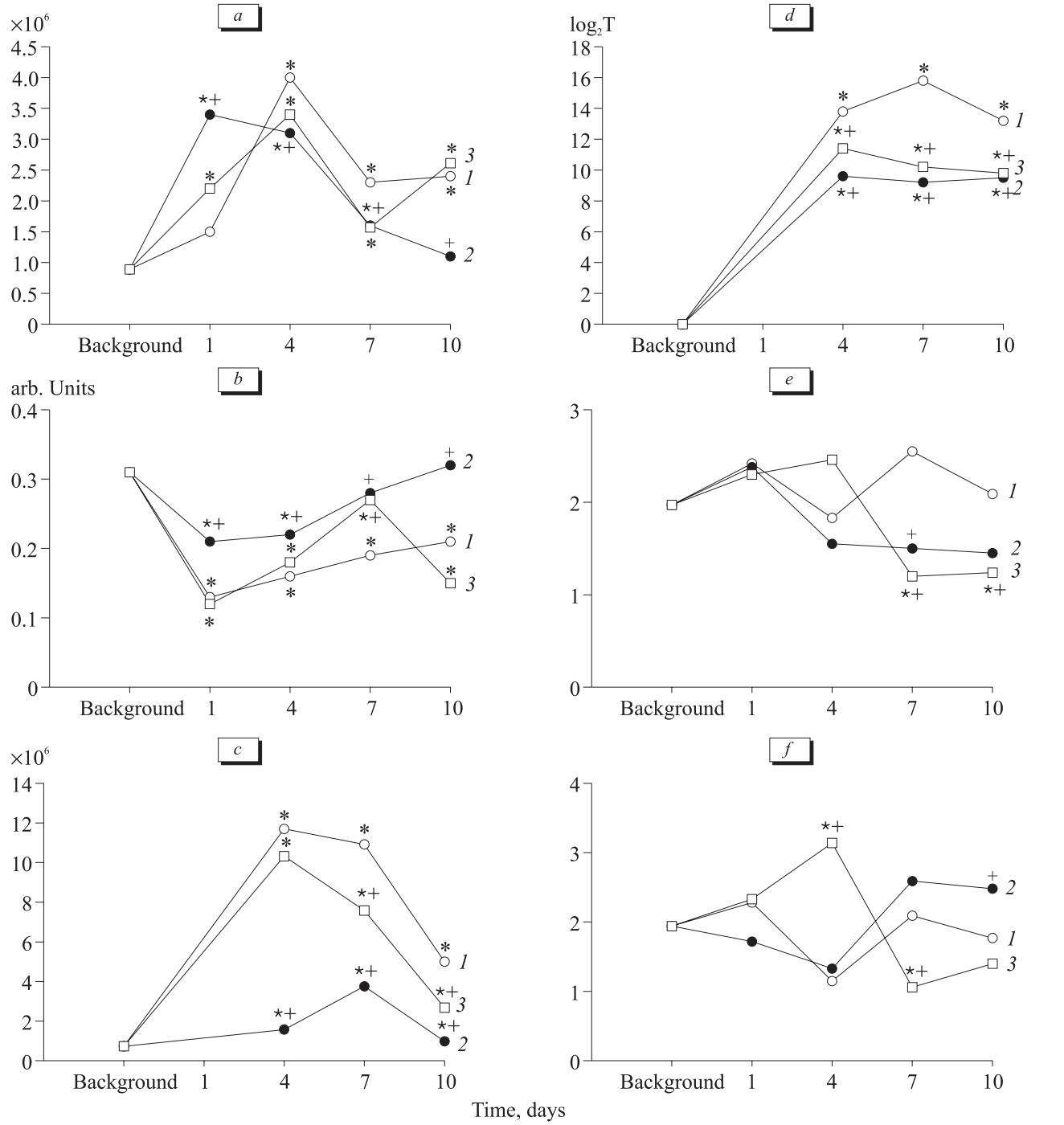

Fig. 1. Effect of immunization (control) on parameters of immune response in (CBA×C57Bl/6)F_1 mice. Here and on Fig. 2: *a*) total number of phagocytizing PM; *b*) phatocytic index; *c*) number of AFC in the spleen; *d*) titer of hemagglutinins, *e*) index of stimulation of T cell proliferation; f) index of stimulation of B cell proliferation. *1*) physiological saline; *2*) α-AB; *c*) β-AB. **p*<0.05 compared to background values, ^{+}p<0.05 compared to *1*.

conditions. α-Adrenoceptors are present on PM and alveolar macrophages, while peripheral monocytes express only β-adrenoceptors; however, modulation of α_1- or α_2-adrenoceptors can modulate the immune response [9,10]. Treatment with β-AB reduced the efficiency of humoral immune response, but to a lesser extent than α-AB. This is probably related to the fact that β-AB prevents inhibition of Th1 functions by catecholamines and shifts the cytokine balance towards Th1, thus promoting the cell response, because it is known that propranolol blocks the inhibitory effect of catecholamines on cytokine-producing cells and increases lipopolysaccharide-stimulated synthesis of TNF-α and IL-12

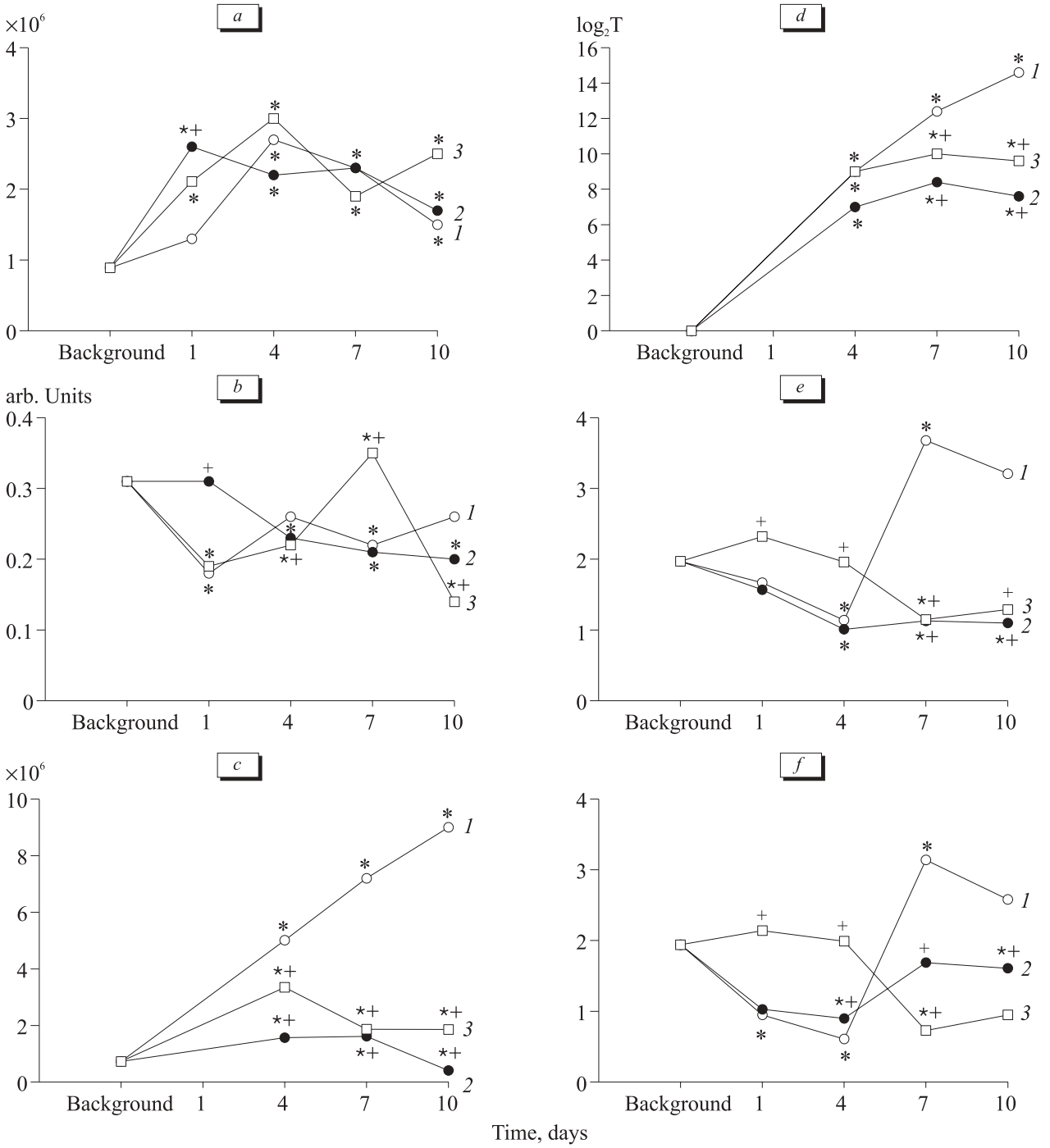

Fig. 2. Effect of immobilization and immunization (experiment) on parameters of immune response in (CBA×C57Bl/6)F_1 mice.

in mice [9,13]. Thus, treatment with α- and β-AB can modulate the reaction of both humoral and cellular immunity by producing opposite effects on the functions of immunocompetent cells.

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