Detection of Fcγ-Receptor Blocking Antibodies in the Serum following Vaccination

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Abstract

In the current series of experiments the Fcγ-receptor blocking serum activity (EAI) was studied before and after vaccination with tetanus toxoid and rDNA HBsAg vaccines in two groups of 6 healthy persons. The analysis of the individual serum samples (on three different allogeneic and on autologous B lymphocytes) suggests that two types of EAI antibodies were produced: allo- and autoantibodies. In all cases an increase of EAI activity was noticed after immunization (in the cases of tetanus toxoid vaccination from 17.7 ± 9.7 to 39.3 ± 16.0 and in the cases of virus vaccination from 67.4 ± 16.5 to 80.2 ± 4.5).

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Fcγ-receptor (FcγR) blocking antibodies have been demonstrated in patients’ sera following transfusions [1], preparation of anti-Rh(D) hyperimmune globulin [2], in hyperimmune normal pooled gamma-globulin [3], and in intravenous gammaglobulin [4]. Earlier studies demonstrated that there are two types of FcγR blocking antibodies: alloreactive or autoreactive. These antibodies probably have regulatory functions on immune response [4]. These noncytotoxic, IgG class antibodies do not bind directly to Fc-receptors but rather to antigen structures on the lymphocyte membrane closely associated with Fc-receptors [5]. They can be detected by inhibition of rosette formation (EAI) with IgG sensitized erythrocytes and B lymphocytes (EA rosettes). This assay has been previously described in details [6]. Briefly, T cell-depleted lymphocyte fractions, referred to as B cells, were incubated with blocking serum samples. After washing, the cells were incubated overnight at 4 °C with 2% human Rh(D) erythrocytes preincubated with anti-D antibodies. Rosettes were counted and expressed as:

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\text{EA rosettes in the test} \]

The inhibition of EA rosette formation by serum samples (inactivated by heat treatment at 56 °C for 30 min, and then ultracentrifuged at 105 g for 90 min) before and after vaccination was tested (table 1). The patients have never received blood transfusions or ever been pregnant. Some patients (1–6) were vaccinated with tetanus toxoid (Human, Budapest) while others (7–12) were vaccinated with recombinant DNA-HBsAg vaccines (Engerix-B, Smith Kline RIT, Belgium), respectively. Serum samples were collected before vaccination and on the 10th day after vaccination in the first group (1–6) and on days 4, 7 or 14 after vaccination in the second group (7–12). Data are expressed as a mean of EAI activity tested on three different lymphocytes. They suggested that EAI activity of serum samples was independent of the serologically detected HLA type of the lymphocytes: (1) lymphocyte: HLA-A 23, B 8,40, DR 1,7; (2) lymphocyte: HLA-A 29,33, B
14,39, DR 3,5, and (3) lymphocyte: HLA-A 2,9, B 7, DR 4, 5. The EAI activity measured with autologous cells was lower than that tested with allogeneic cells. EAI activity induced by tetanus toxoid was lower as compared to the activity induced by virus vaccination. Both EAI activities (autoreactive and alloreactive) belong to the IgG fraction as shown by the persistence of EAI activity following inactivation of IgM by DTT (dithiothreitol) treatment of sera (table 1). On the other hand, after cold ethanol precipitation, the serum samples did not show any EAI activity (data not presented).

These findings, besides their theoretical interest, have some practical implications. If EAI activity is im-

Table 1. Mean (%) and range of EAI activity in the serum produced by vaccination

portant in the survival of transplanted kidney [1], one possibility to induce these antibodies might be vaccination. Kreisler et al. [7] reported on induction of cold-reactive lymphocytotoxic antibodies following immunization with viral vaccines, and their association with a good kidney graft survival.

References


