Half-Life of Murine IgE Antibodies in the Mouse

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Abstract

The half-life of intravenously injected murine anti-DNP IgE antibody was determined in recipient mice by sequential bleeding and was found to be 12 h. Intradermally injected IgE antibody persisted much longer, the half-life was 6 days.

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The half-life of circulating murine IgE antibodies was measured after passive intravenous administration of known amounts of mouse monoclonal antibody or serum of mice containing IgE antibody obtained by conventional immunization and the half-life of intradermally injected antibody was also measured in passively sensitized animals.

Female BALB/c mice raised in our animal facilities were used for the maintenance of hybridoma B 53 producing monoclonal anti-DNP antibody [1] and for immunization for production of conventional anti-DNP IgE antibody. Antigens and immunization were described [2]. Briefly, 0.2 µg dinitrophenylated oval-bumin (DNP-OVA) was injected intraperitoneally into 8 to 10-week-old mice with 1 mg Al(OH)₃ as adjuvant. 4 weeks later, the spleen cells of these donor mice were harvested and 1.5×10^7 spleen cells were injected intravenously into irradiated (600 R) syngeneic recipients which were then boosted intraperitoneally with the same dose of antigen as that used for the immunization of the donors. Adoptive transfer was used as we had shown previously [2, 3] that a higher amount of IgE antibody is produced in these mice than in the original donors. The animals were bled 2 weeks later, the sera pooled and titrated by passive cutaneous anaphylaxis (PCA) in 300 g male Sprague-Dawley rats obtained from Blue Spruce Farms (Alta-mount, N.Y.) as described [4].

1 This work was supported by a grant (AI-03075) from the National Institute of Health. Known amounts of monoclonal antibody or sera from immunized mice containing known amounts of anti-DNP antibody were injected intravenously in 0.2 vol into normal syngeneic recipients. These mice were then periodically bled from the retroorbital sinus as described [5, 6] for titration of anti-DNP IgE antibody. The sera were titrated individually. 6 BALB/c recipients and 6 C57BL/6 recipients were used for the monoclonal antibody. Only BALB/c recipients were used for sera from immunized mice. The C57BL/6 mice were obtained from Jackson Laboratory, Bar Harbor, Me. The batch of ascites used in these experiments contained 80 µg monoclonal anti-DNP IgE/ml. For easier comparison with the antiserum obtained by immunization, we express the anti-DNP content as the minimal dilution giving threshold PCA reactions which is around 10 ng IgE/ml.

Table I shows the results of a typical experiment with monoclonal anti-DNP antibody and table II the results of a typical experiment with conventionally obtained anti-DNP antibody.

In spite of the fact that the amount of monoclonal antibody was much higher than the amount of conventionally obtained antibody, the rate of elimination from circulation was the same, and in both cases, the half-life was calculated to be 12 h. If the fixation of IgE to Fcε receptors were to play an important role, it would be expected that the lower amount of injected antibody would disappear faster from circulation. This, however, was not the case.

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Table I. Half-life of intravenously injected monoclonal anti-DNP IgE

Mouse Day

0 12 3

‘ Last dilution giving PCA in rats. 0.2 ml ascites with monoclonal anti-DNP B 53 titer of this preparation 1/40,000/ml expressed as last dilution giving PCA in rats. Recipients: 1–6 BALB/c; 7–12 C57BL/6. Day 0 bleeding in mice 1–3 and 7–9 was 15 min after antibody injection in mice 4–7 and 10–12 was 1 h after antibody injection.

Table II. Half-life of intravenously injected anti-DNP IgE obtained from immunized mice

Mouse Day 0 Day 1 Day 2

15 min 24 h 48 h

later later later

120’ 30 ND

120 30 ND

120 15 ND

1 Last dilution giving PCA in rats. ND = Not done. 0.2 ml anti-DNP IgE intravenously, titer of this preparation: 1/1,200/ml expressed as last dilution giving PCA reaction in rats.

The persistence in intradermally injected sites was investigated in CFW mice obtained from Charles River Breeding Laboratories (Wilmington, Mass.). 4 mice were used for each sensitization period and challenged with 0.5 mg DNP mouse albumin in 0.2 ml 0.5% Evans blue dye. Table III shows a typical experiment. It is interesting to note that even after a sensitization period of 2 h, PCA reactions were obtained with dilutions of 1/4,000; the maximum was after 1 day, 1/8,000. With IgE antibody obtained by immunization, generally 1 or 2 days of sensitization period is necessary to obtain maximum sensitization and shorter sensitization periods were not thoroughly investigated. The half life of persistence in the intradermally injected site was shown to be 6 days.

Very similar results were obtained by Tada et al. [7] in the rat with rat IgE antibody. They found that the half-life of the circulating IgE in the rat was 12 h and the persistence in the skin of intradermally injected IgE was 7.4 days.

The persistence of murine IgE in rat skin could not be investigated as, after 4–6 days, rats mounted an immune response to murine IgE antibody and the intradermally injected murine antibody could not be demonstrated. The sera of these rats contained anti-murine IgE which could be shown by reverse PCA [8] or by passive hemagglutination, using sheep erythrocytes coated with DNP as described [9], then covering them with nonagglutinating amounts of monoclonal anti-DNP IgE. One drop of 0.1% coated sheep erythrocytes and one drop of rat serum dilution were mixed. The diluent was 0.15 M NaCl.

We can conclude that murine IgE antibody persists for a short time in the circulation of the mouse (half-life 12 h), but much longer when the antibody is injected intradermally (half-life 6 days).

Table III. Half-life of intradermally injected monoclonal anti-DNP IgE

1 Last dilution giving PCA in rats. 0.03 ml of an ascites containing 80 µg anti-DNP IgE was injected intradermally. The animals were challenged with 0.2 ml 0.5% Evans blue containing 500 µg of DNP mouse albumin.

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