The Interactions of IgM Antibody with Immunoadsorbants

by

Brian L. Clevinger

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April 21, 1975
Date

D.C. Bauer, Ph.D.
Professor of Microbiology

Joseph S. Ingraham, Ph.D.
Professor of Microbiology

Ann G. Matthysse, Ph.D.
Assistant Professor of Microbiology

SUMMARY

The purpose of this investigation was to devise an efficient method for eluting IgM antibody from an immunoadsorbant. Bromoacetyl cellulose conjugated with bovine serum albumin was used to isolate anti-BSA IgM from a globulin fraction of a pool of primary anti-BSA sera. IgM was the primary hemagglutinating antibody in the pool and this activity was used to follow the recovery of IgM from the immunoadsorbant. A variety of physical-chemical parameters were tested for their effects on eluting hemagglutinating activity from the BSA-BAC. These included the type of eluting agent, the amount of immunoadsorbant used, and the time the globulin fraction was exposed to the adosrbant. The conclusion drawn from these experiments was that anti-BSA IgG could be recovered from the BSA-BAC almost quantitatively (90%) and that as much as 80% of the anti-BSA IgM was irreversibly bound to the immunoadsorbant. A second immunoadsorbant, carboxymethyl cellulose conjugated with sulfanilic acid, was prepared, and attempts were made to reduce the irreversible binding of IgM to this immunoadsorbant by decreasing the number of SA molecules to which an IgM molecule could bind. These attempts included the preparation of three SA-CMC immunoadsorbants with decreasing degrees of SA substitution and the blocking of SA sites with anti-SA IgG. The experiments showed that reducing the number of available SA sites decreased the number of IgM molecules that adsorbed tothe immunoadsorbant but did not increase the yield of IgM that did bind. A possible explanation for these results was that an IgM molecule must bind

by a minimum number of its binding sites to adsorb to an immunoadsorbant. Binding by this number of sites, however, gives a highly avid interaction which is not disrupted by the elution techniques employed in this study.

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