

Solubilities of Protein-Antigen/Rabbit-Antibody Complexes as a Measure of Serum Avidity

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Abstract

Several immunizing schedules were examined for the production of rabbit antibodies to human serum albumin, ovalbumin and lysozyme, having lowered affinities for respective antigens. From those sera which contained precipitating antibodies, antigen/antibody complexes were made at optimum proportions at pH 7.0. The solubilities of such complexes were measured in buffers of constant ionic strength (0.26) and of pH ranging from 2.5 to 10.0. The use of large quantities of alum-adsorbed antigen, administered in small doses (schedule I) yielded sera, complexes derived from which were the most soluble at least extreme pH values. Sera raised with complete Freund's adjuvant (CFA) yielded the least soluble complexes. Administration of large quantities of alum-adsorbed antigens in two doses produced sera whose complexes were similar to those of CFA sera. Sera taken early in the response to antigen and CFA yielded complexes similar to those of schedule I sera. Attention was focused on the effect of citrate buffer pH 4.0, in dissociating antigen/antibody complexes since at this pH value maximum solubility differences were observed between complexes derived from the greatest number of sera. Where technically possible antisera yielding complexes soluble at pH 4.0 were shown to yield dissociable complexes (by ultracentrifugation), to contain non-precipitating antibodies (by precipitin analysis), and to be non-avid (by the technique of Farr (1985)).