

coagulum with fibrinogen in a few minutes. As thrombin is made with 8 per cent. NaCl solution, it was diluted to approximate isotonicity, m/6. In order to still further control any possible osmotic effects from hypotonicity, solutions of m/5 and m/4 concentrations were also used to dilute the serum.

The system consisted of 1 c.c. of 1-100 immune serum, plus 0.1 c.c. washed ox corpuscles (5 per cent. emulsion), plus thrombin in amounts varying from 0.05 c.c. to 0.3 c.c. As a control one tube was always prepared with normal serum (alexin) instead of thrombin. The tubes were kept at a temperature of 36-37° C.

The results can be stated in a few words. The control tubes showed complete hemolysis in from fifteen to thirty minutes. In no instance was there a trace of hemolysis even after several hours, in the tubes containing thrombin. The corpuscles settled out of the solution and left a clear supernatant fluid, not even tinged with hemoglobin. Having obtained this negative result with thrombin, I then tried solutions of fibrinogen and of serum-globulin, in order to make the matter complete. Exactly the same results were obtained, and I think we may conclude that whatever alexin may be, it is certainly *not* identical with thrombin or the globulins.

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On the nature of the union of alexin with specific precipitates.

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In continuing studies on the nature of alexin fixation by mixtures of unformed proteins and their antisera, it occurred to the writer to examine whether the alexin fixation which is exerted by specific precipitates was subject to the same conditions that prevail in the case of sensitized cell complexes in their relations to the alexin fractions as first obtained by Ferrata. It is well-known, of course, that by dialysis, by dilution with weak acid in distilled water and by a number of other methods of globulin precipitation, the alexin or complement can be divided into two functional parts, one which comes down with the globulins, the

so-called "midpiece" and the other which remains in the albumin fraction, the so-called "endpiece." Neither of these can produce hemolysis of sensitized cells alone. Together they functionate. The globulin fraction can be bound to sensitized cells, forming the so-called "persensitized cells" which are now hemolyzable by the endpiece alone. The albumen fraction does not become attached or fixed to the sensitized cells except in the presence of the globulin fraction. (The terms midpiece and endpiece are used for the sake of clearness since they are terms which have become established in the German literature. Owing to studies which are being made by Mr. Maltaner in this laboratory we feel that a definite nomenclature which assumes an intermediate function of the globulin, fraction, is premature.)

In experiments in which alexin fractions, produced by both the method of Ferrata and by that of Sachs and Altmann, were exposed to union with precipitates, formed in mixtures of beef serum and its antiserum, we have found that the conditions which prevail are entirely analogous to those which govern the attachment of the alexin fractions to the sensitized cells. A specific precipitate may fix the globulin fraction (midpiece) alone. It may fix the albumin fraction (endpiece) in the presence of the globulin fraction. It does not however, fix the albumin fraction (endpiece) by itself. The experiments were in every case controlled by titrations of the alexin fractions and the whole alexin in tubes set up parallel with the main experiment.

The writer believes that these results have considerable theoretical importance in bearing out his previously expressed view that the "precipitin" may be regarded as a protein sensitizer, the fact of visible precipitation being a merely secondary occurrence due to the union of two colloids under conditions of quantitative relations and environment which favor precipitation.