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Effect of Sodium Cyanide on Complement Hemolysis.

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The addition of various amounts of neutralized NaCN to the complex, red cells, specific hemolysin, and complement partially or completely inhibits hemolysis. (0.4 cc. or 0.5 cc. of 1% solution of NaCN in a total volume of 3 cc.) It makes no difference whether the cyanide is incubated first with the amboceptor or with the complement, for on the addition of the other reagents of the test the same degree of inhibition is noted.

The inhibitory action is not due to the HCN gas liberated by neutralization of the alkaline NaCN. The NaCN solution was adjusted to pH 7 by addition of HCl. This was tested by saturating all of the constituents of the test with HCN gas and comparing the time necessary for complete hemolysis, by reagents saturated with HCN gas with the time required by untreated reagents. The time was the same in both cases showing that the HCN gas was not inhibitory.

The cyanide does not effect the union of immune hemolysin with the red cells as washing the red blood cells after contact with hemolysin in the presence of cyanide in order to remove the cyanide, permits hemolysis to take place when complement is added.

These results are contradictory to those of L. Jarno and L. Suranyi,[1] who suggest that the action of NaCN is due to the inactivation of a cholic acid-like complex which they believe is associated with the properties of immune hemolysin.

Inasmuch as these workers fail to state whether they neutralized the cyanide their results might be due to the alkalinity of the cyanide solution. Experiments were carried out to show that the alkalinity of NaCN is sufficient to inactivate complement when used in amounts identical with those reported by the above workers. When unneutralized NaCN is used as above, the pH of the mixture was 11, but the union of amboceptor with red cells is not affected, as after washing red cells which have been treated with NaCN treated amboceptor, the addition of fresh complement results in hemolysis. On the other hand the presence of the unneutralized NaCN inhibits the action of the complement in mixtures of red cells, hemolysin and complement.

[1] *Z. für Immunitats.*, 1930, **69**, 298.

Apparently the effect of the cyanide in inhibiting hemolysis is upon the complement and not upon the antigen-amboceptor complex; and this effect is not dependent upon the pH of the cyanide.

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Observations on Serological Reactions with Albumose Preparations. II.

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In a former communication[1] reactions with immune sera produced by injections of heteroalbumose preparations were briefly described. In order to give some additional information the following experiments, which were carried out with 3 albumose fractions obtained from the products of peptic digestion of coagulated sheep serum, may be cited.

Heteroalbumose was prepared in the manner previously described.[2] The preparation was precipitable from 1% saline solution by about 33% alcohol or by one-half volume of saturated ammonium sulfate solution.

After removal of heteroalbumose, the albumoses which were precipitated between ½ and ⅔ saturation with ammonium sulfate were further fractionated at icebox temperature by precipitation with 65% alcohol at pH 5.1, fraction 1 being insoluble, fraction 3 remaining in solution. Fraction D 1 was precipitable by alcohol in a concentration above 50%, or by addition of 1 to 1.25 volumes of saturated ammonium sulfate solution. Fraction D 3 was precipitable by alcohol in a concentration above 80%, or by addition of 1.25 to 1.5 volumes of saturated ammonium sulfate solution.

Precipitin tests with a particularly strong antiserum obtained by injections of crude heteroalbumose mixed with charcoal[3] are given in Table I.

As already stated, by means of inhibition tests reactions could be

[1] Landsteiner, K., and van der Scheer, J., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 983.

[2] Landsteiner, K., and van der Scheer, J., *Z. Hyg. u. Infekt.*, 1931, **113**, 2.

[3] Landsteiner, K., and Jacobs, J., PROC. SOC. EXP. BIOL. AND MED., 1932, **29**, 570.