

obtained from the other strains. The strict specificity of these substances makes it difficult to attempt any typing by this means in such a heterologous group of organisms.

A similar substance isolated from encapsulated *B. coli* reacted also in very high dilution with homologous immune sera and showed the same narrow type specificity.

To determine whether these substances were antigenic, rabbits were injected in the same way as with the bacterial culture, but with considerably larger quantities of dry material. Precipitations were not produced.

Protein free hapten was isolated from a strain of *B. lactis aerogenes*. This substance consists chiefly of polysaccharides, but in spite of several attempts at further purification, its nitrogen content could not be reduced below 0.9 per cent. Similar substances obtained from several other aerogenes strains and from encapsulated *B. coli* showed a strict individual specificity, no cross reactions were observed.

¹ Emmerling, O., *Ber. d.d. chem. Ges.*, 1900, xxxiii, 2477.

² Schardinger, F., *Cent. f. Bakt.*, Abt. II, 1902, viii, 144.

³ Toeniessen, E., *Cent. f. Bakt.*, Orig., 1920-21, lxxv, 225.

⁴ Heidelberger, M., Goebel, W. F., Avery, O. T., *J. Exp. Med.*, 1925, xlii, 701.

3581

Anaphylactic Shock Produced by a Soluble Specific Substance Largely Carbohydrate in Nature.

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In order to study the relation of the precipitin test to anaphylaxis, and to determine whether a substance largely carbohydrate in nature and failing to give protein reactions would produce shock, the following experiments were made. The aerogenes specific substance described in the previous paper, containing 0.9 per cent N and giving a precipitate with immune serum when diluted 1:500,000, was used for the tests.

Twenty guinea pigs were sensitized by the intraperitoneal injection of 1 cc. aerogenes immune serum, and the response of their uteri tested, using the Schultz-Dale method. The uteri of all these animals reacted to the specific substance when tested from 2 hours to

12 days after the injection of serum. The smallest amount of specific substance that caused a distinct but not maximal contraction, was 0.00000625 gm. added to a bath of 123 cc. so that the final concentration of the substance was 1:20,000,000. Desensitization of the uteri was demonstrated after the contraction due to one addition of 0.005 gm. of the substance to the bath.

When tested for sensitiveness to specific substances obtained from other strains of aerogenes there was no response, so that the results correspond to those obtained with the precipitin reaction.

As controls, the uteri from 6 normal guinea pigs were tested and not one gave a reaction following the addition of as much as 0.01 gm. of specific substance to the bath.

In vivo tests were made on 11 guinea pigs 24 hours after the intraperitoneal injection of from 1 to 4 cc. immune serum. Ten of these animals died showing the typical symptoms and signs of anaphylaxis. The M. L. D. of the specific substance for these sensitized animals was 0.000033 gm. The animal receiving 0.00002 gm., showed symptoms, but survived. Control tests on untreated animals show that 0.001 gm. failed to cause any reaction.

Attempts to sensitize guinea pigs with the specific substance alone were negative.

In this, which is a preliminary report, it is shown that in passively sensitized guinea pigs anaphylactic shock is produced by very high dilutions of a substance that is largely carbohydrate in nature. The presence of the small amount of nitrogen, presumably as an impurity, prevents us from concluding definitely that shock may be produced by carbohydrate alone. Further work with specific substances from other organisms is in progress.

3582

Optimal Proportions of Hapten and Immunserum in the Precipitation and Complement Fixation Reactions

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In a recent paper Dean and Webb¹ have shown that just as in the Ramon test there is an optimum zone in mixtures of protein and its antiserum where precipitation occurs more quickly, and that the indicator or first tube in which a precipitate appears contains neither