SPECIAL ARTICLES

THE AGGLUTINATING ACTION OF AGAR ON BACTERIA

THE recent statement by C. P. Fitch and associates[1] that small amounts of agar have some influence on thermo-agglutination of the *Brucella abortus* group leads me to state briefly some facts in reference to the same or a similar phenomenon not associated with heat which was first observed a dozen years ago in the study of *Brucella abortus*. Though the phenomenon may be well known, the writer has not seen any reference to it until the appearance of the article cited. Examination of recent hand-books, such as the "System of Bacteriology" by English authors and the latest edition of the German hand-book by Kolle, Kraus and Uhlenhuth, has not revealed any statement bearing on this property of a universally used culture material.

The custom of having a small amount of condensation water left in the sloped agar tube brought this phenomenon to the surface. When growth from the agar surface is suspended in water, normal saline or bouillon, *Brucella abortus* exhibits active Brownian motion. When the same growth is suspended in a small drop of condensation water from the same or a sterile agar tube, the clumping is so prompt that all bacteria appear in dense cloudlike masses no matter how quickly the slide is placed under the microscope. At first the clumping was referred to specific agglutinins in animal tissues placed in the tube but soon found to be inherent in the agar itself. The clumping persists in the condensation water of a culture medium consisting of 2 per cent. agar only. Acid agglutination was eliminated, inasmuch as the clumping took place in a neutral medium. It does not occur among bacteria from the sloped agar surface itself when they are suspended in bouillon, normal saline or water.

If the original condensation water formed after the agar tubes have been sloped and then placed upright be removed and replaced by either bouillon, normal saline or water, the added fluids acquire within a few days clumping properties. A second replacement acts in the same way. Even when enough fresh fluid is added to the upright tube to cover most of the slope, the fluid acquires clumping properties. This removal and replacement was carried out four successive times by covering the agar surface each time with the fresh fluid. The water clumped after the second exhaustion completely, only partially after the third, and not after the fourth. The clumping agent is present in the water in which agar shreds are allowed to swell up. After repeated washings of the shreds over night the fluid failed to clump. Made into ordinary nutrient agar, the condensation water which formed again clumped, but the clumps were small as compared with the large cloudlike masses of the usual supply. Allowed to stand for several weeks, the agar tubes made with washed agar failed to clump. When a thin film of bouillon was allowed to rest on the entire slope, the clumping reappeared next day. Boiling and resloping brought clumping back promptly, however.

Spontaneous agglutination was first observed among members of the non-motile Brucella group and was at first regarded specific for that group. All races acted alike. Another species similarly affected is the motile paratyphoid bacillus, including many races thus far examined. A strain from the surface of an agar growth when stirred in condensation water becomes clumped, but not wholly. About one third of the rods remain single and in motion. The rest are in clumps in which the rods are ranged side by side. These bundles measure up to 10 μ in breadth. Since the bacilli in young cultures are all in active motion the clumping is not selective with reference to motility. Clumping has also been observed in certain strains of staphylococci, streptococci, *B. pullorum* and *B. bronchisepticus*. There are indications that capsulated bacteria, not clumped themselves by such capsular material, are not affected by the agar agent. The concentration of the agglutinating substance in the condensation water varies with the time of exposure of the agar mass to it. Clumping was still present in dilutions of 1 to 8 in many observations.

It would require more space than this communication is entitled to to go into further details or to try to correlate our observations with those presented by Fitch and associates. Further analysis of the phenomenon has been under way.

THEOBALD SMITH
DEPARTMENT OF ANIMAL PATHOLOGY,
THE ROCKEFELLER INSTITUTE FOR MEDICAL RESEARCH,
PRINCETON, NEW JERSEY

[1] *Tech. Bul.* 73, 47, Univ. Minn. Agri. Exp. Sta., 1930.

INTELLIGENCE AND BODY CHEMISTRY[1]

IN a recent communication to SCIENCE,[2] H. D. Powers reports a series of observations of the calcium and the inorganic phosphorus of the blood in idiots and in normal or superior persons. The writer has certain data bearing on this point to which he desires to call attention. These include material which has not previously been published because the essentially negative nature of the results indicated the need for further work.

The data presented below were obtained from a random sample of children referred to the Illinois

[1] Publications of the Institute for Juvenile Research; Paul L. Schroeder, M.D., Director. Series B—No. 175.
[2] H. D. Powers, SCIENCE, 73, 316, 1931.