## SPECIAL ARTICLES

## THE AGGLUTINATING ACTION OF AGAR ON BACTERIA

THE recent statement by C. P. Fitch and associates<sup>1</sup> 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 eited. 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 con-

<sup>1</sup> Tech. Bul. 73, 47, Univ. Minn. Agri. Exp. Sta., 1930.

densation 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  $10 \,\mu$  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.

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## INTELLIGENCE AND BODY CHEMISTRY<sup>1</sup>

IN a recent communication to SCIENCE,<sup>2</sup> 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

<sup>&</sup>lt;sup>1</sup> Publications of the Institute for Juvenile Research; Paul L. Schroeder, M.D., Director. Series B-No. 175. <sup>2</sup> H. D. Powers, SCIENCE, 73, 316, 1931.

Institute for Juvenile Research, being limited only by the technician's ability to obtain from the patient a sufficiently large sample of blood. They included intelligence levels ranging from the imbecile to the superior. Calcium of the blood was determined by the Clark-Collip modification of the Kramer-Tisdale method; inorganic phosphorus by the Fiske-Subarrow method, and total and lipoid phosphorus by similar procedure following digestion and alcohol-ether extraction, respectively. The intelligence quotient was obtained in each case by an individual mental test.

The coefficients of correlation obtained between the I.Q.'s and the chemical determinations are given below, together with the number of cases upon which each is based (in parenthesis).

	Correlation with I.Q.	
Calcium	+.06	(69)
Inorganic Phosphorus	+.07	(245)
Lipoid Phosphorus	04	(77)
Total Phosphorus	+.06	(118)
Lipoid P÷Total P	+.19	(63)

Our results agree with those of Powers with respect to the calcium of the blood. There is no evidence that it bears any relation to the individual's intelligence.

With respect to the inorganic phosphorus of the blood, on the other hand, we differ markedly from Powers. The data obtained from 245 subjects show no relationship between intelligence, as measured by the I.Q., and the phosphorus content of the blood. There is, of course, a difference in method. We worked with persons ranging from subnormal to superior intelligence, instead of extreme groups, and did not include cases of idiocy. If, however, the difference obtained in such extreme groups is valid outside of the very lowest ranges of intelligence, a correlation will necessarily be apparent when a wide range of intelligence is considered. Although our subjects were children, age does not appear to be a factor, for we have found that it correlates with the inorganic phosphorus content of the blood only to the extent of -.14. Moreover, the determinations of lipoid and of total phosphorus show no higher relationships to intelligence than do those for inorganic phosphorus. The ratio of lipoid phosphorus to total phosphorus does, indeed, correlate more highly with intelligence, but, as this relation is vitiated by a correlation of +.30 between this ratio and chronological age, further work with controlled age groups is essential for its interpretation.

The relationship of phosphorus metabolism to mental phenomena is a problem which is as yet far from solution. We have noted elsewhere<sup>3</sup> a correlation of

<sup>3</sup>G. J. Rich, Jour. Abnorm. and Social Psychol., 23: 172, 1928.

-.51 between intelligence and the excretion of phosphorus per unit of body weight. As the significance of this figure is lessened by the fact that it was obtained from only 28 subjects, it was suggested that verification was most desirable. This definitely positive result from a study of urinary excretion, the negative results mentioned above, and Powers' positive findings, taken together, suggest a complexity of relationship that urgently calls for further work. Our interest has been primarily in connection with certain non-intellectual traits, with which both the calcium and the various types of phosphorus in the blood likewise failed to show any significant correlations.

In his first paragraph, Powers makes reference to the lack of previous work on the relationships of body chemistry to mental phenomena, and characterizes the work that has been done as "vague." This broad statement can not be allowed to pass unchallenged. For example, the literature contains a group of studies, too numerous to mention in detail here, on the differential chemistry of the psychoses and neuroses.<sup>4</sup> In general, the procedures have been similar to that of Powers, save that psychotics or neurotics, instead of idiots, were compared to normals. Many of these studies have yielded negative results and they have often contradicted one another, but they have hardly been "vague." Such work as that of Starr<sup>5</sup> and Stratton<sup>6</sup> on stammerers likewise reached definite conclusions, and the writer<sup>7</sup> has published correlational values. One fears that Powers has been somewhat overhasty in drawing his indictment, for he has himself published nothing more definite than can be found elsewhere.

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## BOOKS RECEIVED

- Anales de la Dirección de Meteorologia. Tomo XIX: Conteniendo las observaciones practicadas en al año 1928. Volumen I: Resultados de las Observaciones Aerológicas Efectuadas con Globos-Pilotos en el Observatorio Regional Buenos Aires. Ministerio de Agricultura de la Nacion, Buenos Aires.
- Boletim do Museu Nacional. Volume VI; No. 2. Pp. 61+147; No. 3. Pp. 149+264; No. 4. Pp. 265-320. Illustrated. The Museum, Rio de Janeiro.
- SPEERSCHNEIDER, C. I. The State of Ice in the Arctic Seas: 1930. Pp. 17. Danish Meteorological Institute.

<sup>4</sup> Reviews of the earlier of these papers may be found in: A. Schaefer, *Monatschr. f. Psychiat.*, 2: 157-162, 229-242, 377-386, 443-451, 1897; F. M. Barnes, *Amer. Jour. Insanity*, 68: 431-472, 1912; and K. M. Bowman, *Amer. Jour. Psychiat.*, 2: 379-408, 1923.

<sup>5</sup> H. E. Starr, Amer. Jour. Psychol., 33: 394-418, 1922. <sup>6</sup> L. D. Stratton, Jour. Compar. Psychol., 4: 325-346, 1924.

<sup>7</sup>G. J. Rich, Jour. Abnorm. and Social Psychol., 23: 158–175, 1928; Arch. Neurol. and Psychiat., 20: 589– 594, 1928.