Whether or not the incidence of furunculosis among kelts in the Scottish rivers in 1958 was unusually high, the two sets of results suggest that the normal incidence of this disease in kelts is high enough to warrant further investigation. As this might be an important factor in the spread of the disease, these observations will be continued.

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¹ Final Report of the Furunculosis Committee (H.M. Stationery Office, 1935).

Genetical and Biochemical Studies of the Immobilization Antigens of Paramecium aurelia

HIGHLY purified preparations of the immobilization antigens of Paramecium aurelia, about which extensive genetic studies have been described previously¹, are now known to consist largely of protein (ref. 2 and unpublished work by J. O. B.). With the aim of obtaining information on genetically controlled protein variations, we have made a preliminary study of the physicochemical properties of some of these antigens, using various stocks of P. aurelia (variety 1). The antigens studied are denoted 60D, 90D, 103D. 33D, 60G, 60T. Formation of the first four is governed by a series of alleles denoted D⁶⁰, D⁹⁰, etc., and of the fifth by a non-allelic gene G^{60} in conjunction with a different cytoplasmic state. The type 60T is produced in conjunction with a third cytoplasmic state; but the genic basis of this type has not been investigated.

The immunological relationships between the different types of antigen appeared to vary according to the kind of test which was performed. By the immobilization test very little cross-reaction was found when any heterologous combination of the abovementioned antigens and the various antisera was made. Other tests, however (absorption of immobilizing antibodies by whole *Paramecia* or migration of precipitin bands in agar-gel diffusion experiments), showed that the four D antigens are immunologically related to each other, and are unrelated to the 60G type (J. O. B., unpublished work).

Electrophoretic behaviour of the antigens was studied on starch-gel at pH 9.6 by a modification of the method of Smithies⁴. A gradient of 30-40 V. per cm. was found to be necessary to get appreciable movement of the antigens and the runs usually lasted 6-8 hr. After this time a slice of the gel was removed and stained with nigrosin, showing the presence of a single band of protein near the origin on the anode side. All other mobile proteins had passed into the anodic buffer vessel by this time. Correspondence of the stained band with antigen was shown by taking a parallel slice of gel and testing the eluate obtained from it for specific serum-blocking activity.

In this way the electrophoretic mobilities of the antigens were compared and it was found that type 60G moved toward the anode significantly more slowly than any of the D types (see Fig. 1), and type 60T was the fastest. All four (allelic) D types were apparently identical in mobility under these conditions.

Some of the antigens were also compared in regard to their adsorption-desorption behaviour on a column of calcium phosphate, using the technique of Tiselius *et al.*⁵. Partially purified preparations of various

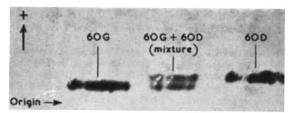


Fig. 1. Photograph of starch-gel slice containing three samples of antigen, stained with nigrosin after 4 hr. electrophoresis at 30 V. per cm., pH 9.6

antigens were applied to the column and elution was then carried out with phosphate buffer at pH 6.8, starting with an ionic strength of 0.001 M and increasing stepwise to 0.05 M. Protein in the samples of eluate was measured spectrophotometrically by ultra-violet absorption at 280 mµ and antigenic activity assayed by serum-blocking. It was found that antigen 60G was eluted by buffer at an ionic strength of 0.005 M, 60D and 60T at 0.01 M and 90D at 0.05 M. When two antigens (60D and 90D) were mixed together and chromatographed on calcium phosphate, two peaks of protein-each containing only one antigen-were obtained at positions on the elution diagram corresponding exactly to those found when the two antigens were chromatographed separately.

We therefore conclude that the electrophoretic comparisons of the different antigens studied to some extent parallel the immunological and genetical relationships: antigens which are unrelated immunologically and controlled by genes at different loci (for example, the G and D types) differ electrophoretically; antigens which are immunologically closely related and controlled by allelic genes at a single locus do not show electrophoretic differences under the conditions chosen, but do differ in regard to their adsorption-desorption behaviour on calcium phosphate.

More detailed accounts of the work referred to here will be published elsewhere. Thanks are due to the Medical Research Council for a grant supporting this work.

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- ³ Oudin, J., Meth. Med. Res., 5, 335 (1952).
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BACTERIOLOGY

Mechanism of Immunity in Hæmorrhagic Septicæmia

HEMORRHAGIC septicemia of cattle and buffaloes caused by *Pasteurella multocida* type I kills many thousands of animals in Asia each year. It is possible to immunize against the disease effectively with improved vaccines, and in addition 5–10 per cent of animals possess a naturally acquired immunity. Both kinds of immune animals have in their sera circulating antibodies which are detectable by the passive mouse protection test. Hitherto, it has been