Ultracentrifugal and Electrophoretic Studies on Antibodies

MANY attempts have been made to obtain from immune sera, substances carrying the antibody function. Protein solutions specifically precipitable to the extent of 40–60 per cent (nitrogen precipitated by pneumococcus specific polysaccharide/total nitrogen) have thus been prepared by the Felton method[1]. By dissociation of specific precipitates with strong sodium chloride, antibody solutions precipitable to the extent of 90 per cent have been prepared[2]. An investigation of the physico-chemical properties of these systems, especially of those bearing upon chemical homogeneity and the relation to the components of normal sera, would seem to be of interest.

To this end, preparations of horse and rabbit antibodies against pneumococcus type specific polysaccharides, as well as rabbit antibody against crystallized egg albumin, have been studied by ultra-centrifuge and electrophoresis methods[3,4], together with whole and fractionated normal sera. The simplest behaviour was shown by a horse serum preparation of type I pneumococcus anticarbohydrate[2] in which 61 per cent of the nitrogen was specifically precipitable. This showed almost homogeneous sedimentation, with sedimentation constant $s_{20} = 17{\cdot}2 \times 10^{-13}$ as compared with that of normal globulin from mammalian sera, s_{20} about 7×10^{-13}, previously found at Uppsala. However, normal sera regularly contain small amounts of a minor component of sedimentation constant about 17×10^{-13}. The electrophoresis of the above antibody preparation was homogeneous, with an isoelectric point as acid as *p*H 4·8.

In the case of rabbit sera, homogeneous sedimentation was found with no appreciable difference between the sedimentation constants of the globulin fractions of normal serum and immune serum, or of normal globulin and immune globulin containing up to 50 per cent of anti-egg albumin, or of antibody to the type III pneumococcus polysaccharide containing 90 per cent of precipitin[2], all showing s_{20} about 7×10^{-13}. Only the last mentioned preparation was investigated electrophoretically, giving inhomogeneous migration and an isoelectric point of about *p*H 6·6.

Preparations from antipneumococcus I horse sera (Felton) showed the same principal component as did the horse antibody above, together with varying amounts of slower and faster sedimenting components; whereas a Felton solution from *normal* horse serum was quite different, with a predominating component of $s_{20} = 7{\cdot}8 \times 10^{-13}$. Electrophoresis was rather inhomogeneous, with isoelectric points of about *p*H 5–5·2 for the immune and *p*H 5·7 for the normal preparations.

In a few runs with horse antibodies, the centrifuge was stopped when all of the heavier components had settled to the bottom, although a considerable amount of the lighter substances were still in solution. Samples taken out indicated that almost all specifically precipitable protein was on the bottom, showing that the antibody function is connected with the heavy component. This is also in accordance with previous experiments[5,6].

Preparations made from antipneumococcus horse sera, preserved with phenol and ether, although containing up to 87 per cent specifically precipitable nitrogen, were characterized by their inhomogeneity both in the ultracentrifuge and in their electrophoretic behaviour. The isoelectric point was *p*H 5·9, but a component of an isoelectric point of about *p*H 4·9 was also present.

From these data it is evident that protein preparations from immune sera carrying a high percentage of the antibody function may show all the signs of being chemical individuals, so far as the methods referred to are decisive. Nevertheless, methods of preparation and preservation may considerably influence the result (by denaturation ?) and give rise to less uniform systems, which, however, may still show a very high specific precipitability. It is also clear that different fractions of the serum proteins carry the antibody function in the horse and in the rabbit. It would thus appear that the rabbit produces antibody from the principal globulin component, while the horse develops pneumococcus I anticarbohydrate, at least, from an otherwise minor component.

The analytical methods for determining the antibody content of the solutions have been described in earlier publications[7]. Details of the experiments will be given and some of the many questions raised by this work will be discussed in forthcoming communications.

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Two Unusual Modifications of Eye Colour in *Drosophila melanogaster*

IN the course of a series of investigations now in progress concerning the modification of eye colour in the fruit fly *Drosophila melanogaster* under X-irradiation, undertaken in connexion with an attempted determination of the volume of the locus 'white', two changes of colour have been observed which seem of sufficient interest to be worthy of a note at the present time.

The first change involves an apparent reverse genovariation, under X-irradiation, similar to those observed by Timofeef-Ressovsky[1], the change being from white to red eye.

The white stock concerned was obtained by suitable breeding from a few white individuals arising as spontaneous mutants in a pure culture of Florida wild-type *Drosophila* obtained about two years ago through the courtesy of Dr. Demerec. It has been bred for three months in this laboratory in pure