

as a route of infection. But it seems very unlikely that a similar process could be of nutritional significance.

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¹Payne, J. M., Sansom, B. F., Garner, R. J., Thomson, A. R., and Miles, B. J., *Nature*, **183**, 586 (1960).

WE are grateful to Prof. R. B. Fisher for his comments but must point out that no claim was made that "similar processes may be of nutritional significance". We presume the sentence under question is: "We suggest that the use of radioactive particles of this kind may prove to be a sensitive method for the study of various problems of infectious disease and possibly also in work on nutrition and nutritional allergy"; but this can scarcely be taken as implying that the absorption of particles is of nutritional significance.

It seemed to us that the demonstration of particulate absorption, however small, might be of interest to those working on the absorption of relatively insoluble elements required in only trace amounts. In this connexion it may be of interest to record that we are endeavouring to determine the relative uptake of particles of different sizes. Experiments of this kind, attempting to measure the 'porosity' of the alimentary canal, may well find some application in work on nutrition although we are mainly interested in the pathogenesis of infectious disease.

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HÆMATOLOGY

Antigenicity of A₂ Hæmoglobin

THE heterogeneity of adult human hæmoglobin is a well-established fact¹⁻³. The present work was designed to determine if the slow electrophoretic fraction A₂ has antigenic specificity and if this fraction can be detected immunologically in cord blood and in the presence of hæmoglobin S and C. Previously described⁴ techniques were used for the preparation and the agar-gel electrophoresis of the hæmolyates. Following electrophoresis the agar gel was cut into sections containing the hæmoglobin fractions to be tested for antigenicity. These sections were then frozen, thawed and filtered through fritted Büchner funnels of medium porosity. The short duration (90 min.) and the use of large glass plates as the base of the agar gel made it possible to obtain quantities of eluates sufficient for immunization of rabbits. Eluates of the following fractions were used for this purpose: the main component (A₁) of normal blood and of blood from a patient with thalassaemia minor; the respective A₂ fraction and the non-hæmoglobin fraction which we have previously⁴ designated 'unidentified component I' and which probably is identical with Derrien's X₁ fraction². In addition the most cathodic part of the fetal hæmoglobin from cord blood (Fig. 1b, area between brackets) was eluted from agar gel because of the possibility that this part contains A₂ hæmoglobin. Each eluate containing approximately 3 mgm. of protein was combined with an equal volume of Freund's adjuvant

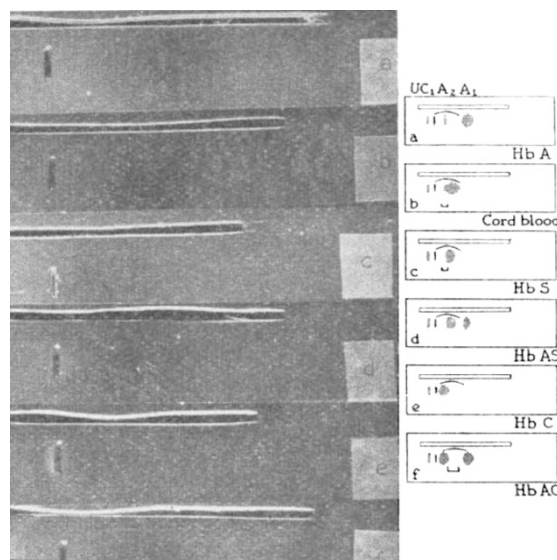


Fig. 1. Immuno-electrophoretic precipitin reaction of A₂-antiserum and hæmolyates. The brackets in b, c, f indicate the areas which were isolated and eluted for immunological studies

and injected into adult male albino rabbits on three or four different sites.

Twenty-one days following the injection blood was obtained from the ear artery and this procedure was repeated at weekly intervals. Serum from these samples was tested for antibody content by applying the Ouchterlony agar-gel diffusion technique as modified in this Laboratory⁵ and immuno-electrophoresis in agar gel on microscope slides. As test antigens the following material was used: (a) agar-gel eluate of the A₁, A₂ and unidentified component I fractions of normal and thalassaemic hæmoglobin; (b) whole hæmolyates of normal adult and cord blood and of blood from patients with thalassaemia minor, sickle cell anæmia, sickle cell thalassaemia, A-S and A-C hæmoglobin; (c) eluates of sections of electrophoretograms of fetal, S, and A-C hæmoglobin which correspond to the location of the A₂ fraction (Fig. 1b, c, f, areas between brackets).

Antibody to A₂ hæmoglobin appeared three weeks after a single injection of the A₂ fraction. This antibody reacted with hæmolyates of normal adult and fetal hæmoglobin, hæmoglobin S, A-S, A-C (Fig. 1) and S thalassaemia. The precipitation line was always in a position opposite to a place on the electrophoretogram corresponding to the A₂ fraction. On Ouchterlony plates all hæmolyates reacted with the antiserum, but when eluates of the isolated fractions were used as test antigens, only A₂, the slowest section of S hæmoglobin, and the eluate of the interspace of A and C reacted, which are the areas corresponding to the location of the A₂ component. Antisera to the slowest section of fetal hæmoglobin reacted in the same manner as the antiserum to A₂ from thalassaemic and normal blood, suggesting immunological identity of these fractions.

Antibody to A₁ and to unidentified component was demonstrated only following one or two booster injections. A₁ was only weakly, but the unidentified component I was strongly, antigenic (Fig. 2). Anti-unidentified component I serum cross-reacted slightly with A₂ but not with A₁. A clearly defined precipita-