Electrophoretic Evaluation of the Serum Proteins in Malignant Disease

It is well known that many diseases produce a marked alteration in the level of the total serum protein. Furthermore, it has been shown by electrophoretic analysis that certain diseases bring about distinctive changes in the relative concentrations of the different serum proteins. In early active tuberculosis[1] there is a rise in the γ-globulin fraction of the serum protein and a corresponding fall in the albumin-level. As the disease progresses the α^2-globulin concentration also increases and in advanced tuberculosis all the globulin concentrations are raised. In contrast, patients with nephrosis[2] show an increase in the α^2-globulin concentration which is usually associated with a decrease in the γ-globulin fraction.

In malignant disease there is an excessive protein demand by the rapidly growing tumour cells, which can only be met by utilizing the host's dispensable protein and the labile serum proteins. This drain on the body's store of protein is bound to be reflected as a change in the concentration of the different serum proteins. Mider *et al.*[3], using the Tiselius electrophoresis apparatus with a veronal buffer *p*H 8·5, found that the total plasma protein-level in malignant disease was below normal, the albumin-level was reduced but there was a significant rise in the level of the α-globulins; the β- and γ-globulin fractions also rose but the change in concentration was much less. Peterman and Hogness[4], using a similar method, found that in patients with carcinoma of the stomach the albumin-level was low, the α-globulins were raised, but the β- and γ-globulin-levels were normal. Gross and Snell[5] reported that there was no significant difference between the serum γ-globulin-levels of patients with malignant disease and those of healthy controls.

The present report concerns the results obtained from an electrophoretic evaluation of the serum protein-levels of twenty-four patients from the Dartford Group of Hospitals, Kent, suffering from malignant disease, which had been verified by histological examination. The patients were chosen at random, and their ages varied from thirty-one to seventy years. The majority of the tumours were situated in the genital or intestinal tracts and were at different stages of development, some being localized while others had metastasized widely. Blood samples were taken from each patient on admission and only the blood from those patients having normal temperatures, total and differential leucocyte counts, erythrocyte counts and hæmoglobin-levels were used. Blood samples were also taken from twenty-one healthy females of the same age-group to serve as controls.

The protein-level of each of the sera was estimated by first separating the different fractions by paper electrophoresis at *p*H 8·5 using a veronal buffer. The filter paper strips were then treated with bromphenol blue and the optical density of the protein dye complex was measured by means of a photoelectric cell. In this manner the percentages of the different proteins in relation to the total protein in the serum could be assessed.

The results showed that the sera of the patients with malignant disease had no distinctive electrophoretic pattern. The difference between the α^1-, α^2- and β-globulin-levels of the patients with malignant disease and those of the healthy controls was not statistically significant. The level of the serum albumin, however, was lower in the patients with malignant disease than in the controls. Although these findings are not in complete agreement with those of other workers it must be appreciated that only those patients having normal temperatures and blood pictures were used for the present investigation and the stage of development of the malignant tumour in each case was not taken into account.

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Intracellular Site of Developing Herpes Virus

A study of the early hours of multiplication of herpes virus in chick embryo cells[1] showed that some 90 per cent of the virus present in the cells at the beginning of incubation could not be detected 3 and 6 hr. later. The method of cell homogenization employed in this work to extract virus for titration left the cell nuclei intact, and the findings of Gray and Scott[2] suggested that the virus lost in the early hours of incubation might have made its way into the nuclei, and so escaped detection. This possibility was investigated.

Chorio-allantoic membranes of 10-day fertile hens' eggs were infected with herpes virus. After 2, 4, 6, 8 and 10 hr. incubation, membranes were excised, washed and put in diluted anti-herpes serum overnight at 4° C. The membranes were then washed, homogenized and diluted. Fixed and stained smears of diluted homogenate showed intact cell nuclei stripped of cytoplasm. The nuclei were deposited by centrifugation and the virus titre of the supernatant 'cytoplasmic fraction' measured. The nuclei were repeatedly washed, then half were suspended in dilute rabbit serum with 0·85 per cent sodium chloride and half in dilute rabbit serum with 6 per cent sodium chloride. These two suspensions were left overnight at 4° C. and then centrifuged, the supernatant containing 6 per cent sodium chloride being regarded as the 'nuclear fraction' and the other as a control. The virus titres of both were measured. Stained smears of the deposits showed that the 6 per cent sodium chloride solution had removed all basophilic material from the nuclei, leaving none intact except for a very few from erythrocytes; in the control, intact nuclei were numerous. The results of the virus assays, made by the pock-count method on chorio-allantoic membranes, are shown in Table 1.

It will be seen that at each interval the nuclear fraction contained more virus than the control, but less than one-tenth of that found in the cytoplasm. In three repetitions of this experiment the distribution of virus between the fractions has been the same as in this instance. If all the virus lost from infected cells soon after the onset of incubation[1] had entered nuclei these should contain up to nine times as much as the cytoplasm.