THE K. INOUYE LABORATORY

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Taking as our starting point the metabolism of liver disease, we have reached the idea that the nutritional factor cannot be neglected. Therefore, our principal course of examination was gradually concentrated to find out the nutritional status of diseased personnels with regard to the function of liver or other endocrine organs.

In the clinical observation of many malnutritional patients prevailed in the wartime, for example, we have demonstrated that the interrelationship of nutrition to the individual constitutional moment, especially to the endocrine functions, is very important. Details have been shown in the volume edited by K. Inouye: "The Nutrition of Japanese People." (1948). The outline of some researches will be shown in the following:

Studies on the Metabolism in Malnutriton

By **K. Suzawa**

In- and post-war times, many persons in this country suffered from malnutrition or semi-starvation. Our laboratory investigated this disease, and many researches were carried out. One of our problems was the metabolism of various vitamins in this disease. The results concerned with thiamin metabolism were as follows: Most patients showed a low level of B_1 content in the blood, while in urine much amount was excreted. In fact, even when much drug administered, the normal level could not be attained. Therefore, these patients were not simply difficient in B_1. In these patients the function to accept and utilize the thiamin might have been disturbed. The hormonal organs might have played a rôle in causing a malnutritional state, because at the autopsy we found the endcrine glands were intensively affected. Consequently, in the treatment, we used cocarboxylase, or thiamin together with the cortical hormon instead of free thiamin alone. Thus we could obtain better results.

Polarograpic Studies of Serum Protein

By **T. Sasai**

In order to find out the pathological changes of the thiol groups contained in serum protein, we studied the polarographic protein wave, which was found by Brdička in ammonium buffered cobaltous or cobaltic solution. Results were as follows.

1. Problems Concered with Serum Protein itself

a) Influence of denaturating agents on serum protein. The effect of denaturation, such as of alkali or of the heating, on serum protein consists of two phases of

reactions polarographically. The height of protein wave increases at first rapidly and then decreases slowly with the lapse of time, and finally disappears. However, under an appropriate temperature for the denaturation, for example in N/5 KOH below 20°C, no phase corresponding to the second decreasing was found. Such a condition may be used for clinical investigation.

b) Experiments on isolated protein fractions.

Using the fractions which were isolated by various methods (e. g. ethanol-fractionation in temp. -5°C or the salting-out method by Na_2SO_4), the relationship between the wave-height and protein concentration was studied both in natural and in denaturated state. We found that albumin revealed a higher wave than globulin and, when denaturated, the wave from albumin becomes very high, such as 2 fold, while the wave from globulin showed almost the same. The wave-forms of proteins were characteristic of fraction albumin or globulin and for their origin (humane or horse), and not of the isolating method. From the facts above mentioned, regarding either the height or the form of protein wave, it is apparent that the wave obtained from serum protein means a combined wave of the albumin and globulin contained in sera.

Clinical evidence that in the cases of some diseases, including cancer and inflammatory diseases, the wave is obviously lower than the normal, are due to two facters, namely the quality and quantity of serum protein. To find the nature of this test, we compared the wave heights from the definite serum-dilution of equal amount of protein, thus putting the quantity out of consideration, with the value of A/G ratio of each serum obtained by Kjeldahlometry.

Thus it was revealed that at least in the cases of the liver disease the correlation between the two values was very intimate. In the other diseases, e. g. in blood disease and in the acute nephritis, there were, however, some cases which showed no relation between two values. In a word we opine that in essentials the polarygraphic test of the serum protein represents the test with regard to A/G ratio, but some clinical materials may contain other high molecular and surface-active substances, which do not show their proper wave but interfere the protein wave.

2. Problems Concerning the Serum Filtrate from Sulfosalicylic Acid

The protein wave is also obtainable from the serum filtrate, which contains protein-like substances, soluble in sulfosalicylic acid and insoluble in tungstic acid and in saturated $(NH)_2SO_4$ solution. Clinical evidence is that the filtrates, obtained from the sera of cancer or inflammatory diseases give a much higher wave than the normal. In general the denaturating procedure of serum before its deproteination has been done by many authors for the reason that by this procedure the filtrate wave becomes higher and the difference between the normal and disease states becomes more evident. From these facts they (Brdička and his followers) believed that the filtrate wave would represent the split products of serum protein.

In our experiments it was, however, found that the increase of soluble substances in the filtrate, caused by alkaline effect, is usually the secondary change,

and rather the initial change should be the decrease, and this variation is due to the unkowen factor of individual serum protein. Therefore, in the clinical evalution of the filtrate wave, it is more effectual to compare the wave height from non denaturated sera than that from denaturated sera.

As new clinical findings we must point out the fact that in the cases of parenchymatic liver damage, such as liver cirrhosis, hepatic coma or hepatitis, the heights of the filtrate wave are usually lower than the normal; especially in the cases of cirrhosis the grade of lowerness was so obvious that it should be a pathognomonic sign for the liver disturbances, because the phenomenon of this kind was never found in other diseases.

In nephrotic patients, the filtrate wave did not show any constant results. However, in nephrosis or in hypoproteinemia of other origin, it seemed interesting that the filtrate wave became much higher in the reconvalescent stadium, namely directly before the period when the reëstablishment of serum protein occurs.

It was demonstrated by R. J. Winzler and K. Mayer, that this filtrate substance is mucoprotein. We found that this is a constant constituent of serum, tissue and urine. And we believe this protein may play an important rôle in the protein metabolism of human being.

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