PROTEIN METABOLISM

INFLUENCE OF GROWTH HORMONE, ANABOLIC STEROIDS, AND NUTRITION IN HEALTH AND DISEASE

AN INTERNATIONAL SYMPOSIUM

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AN INTERNATIONAL SYMPOSIUM

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Preface

The Symposium on "Protein Metabolism: Influence of Growth Hormone, Anabolic Steroids, and Nutrition in Health and Disease" is the fourth in the series of International Symposia sponsored by CIBA Limited, Basle. As in the case of the previous conferences, it was planned and organised with the help of experts in the field concerned. Special thanks are due to Prof. A. QUERIDO and Dr. A. A. H. KASSENAAR who, once the idea of the Symposium had been conceived in the course of joint discussions, embarked upon the project with enthusiasm and inspiration, although they must have known full well what a great deal of time and trouble the organisation of such a meeting would inevitably cost them. For their untiring efforts, for the judicious manner in which they contrived to select precisely those subjects on which interest is chiefly centred today, and — last but not least — for their success in finding competent specialists to participate in the proceedings, we wish to assure them of our sincere gratitude. To all the members of the Department of Clinical Endocrinology and Diseases of Metabolism, at the University Hospital in Leyden, who helped in preparing the meeting, we would likewise extend a warm vote of thanks.

The fact that the present volume, featuring the papers and discussions of the Symposium, has been published only a few months after the event, was made possible thanks to the co-operative help of all who participated. In this connection, we are particularly indebted to Mr. H. D. PHILPS, M. A., and Miss S. R. NAEGELI, who devoted all their available time to the task of meeting the dead-line which we had set ourselves, and to Dr. WILTRUD HATZINGER, who kindly prepared the subject index for us. We should also like to express our appreciation to Dr. H. GÖTZE

for the co-operation and understanding with which he and his associates of Springer-Verlag met all our demands.

One difficulty with which we were faced was that of the difference between English and American spelling. Lest we should be accused by our American readers of ignoring the welcome simplifications which in their country have been introduced into the complexities of English orthography, we have adhered to the American usage in the case of the papers read by American authors.

Basle, December, 1962 F. G.

Corrigendum

Fig. 1 A and 1 C on page 263 and 265 have been interchanged by error.

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Opening remarks

By

A. QUERIDO

"However elegant and memorable, brevity can never in the nature of things do justice to all the facts of a complex situation".

ALDOUS HUXLEY*

Many of the participants, when invited to attend this Symposium, were kind enough to praise the programme. I can assure you that it was not so difficult, with the efficient aid of Dr. GROSS and Dr. KASSENAAR, to arrive at the final scheme. It was, though, more difficult to think of a concise title. We finally chose "Protein Metabolism", adding, however, the subtitle "Influence of growth hormone, anabolic steroids, and nutrition in health and disease".

You might consider these remarks futile for this distinguished gathering, but they are made to underline a rather important point. The title of this Symposium does not clearly indicate what the organisers were driving at when they invited you to participate.

What, then, was the idea behind this programme? To say it in a risky way, it was to assemble together a number of experienced workers in the field of protein metabolism, in order to *discuss* biological mechanisms and conditions related to protein metabolism at the cellular level and in the organism in a state of health, in a state of disease, and in a state of repair. It is risky for me to put it that way, because I have only mentioned the workers and not their papers — even though the latter have undoubtedly been composed with great care, if perhaps grudgingly. I hope that, having examined these papers thoroughly, we shall consider them only as a framework for the development of our thought. Unfortunately, I must dispel any impression you may have that you had finished your task once you had prepared and handed in your

* "Brave New World Revisited" (Harper, New York).

manuscripts, because I sincerely hope that a major and essential part of the work will be done during the actual meeting.

However, it is quite clear that we cannot discuss all aspects of protein metabolism, and the action of physiological and pharmacological agents upon it. I shall therefore try to put forward some thoughts and problems that may be relevant to this Symposium.

It is not necessary before this audience to stress the significance of proteins for life. A short quotation from PAULING (*6*) may serve our purpose here: "The molecules that compose the body of a human being may be conveniently divided into two classes: small molecules and large molecules. Small molecules are molecules containing 10 or 20 or perhaps 100 atoms" . . . " large molecules are molecules containing hundreds or thousands or tens of thousands of atoms; examples are the proteins and the nucleic acids." . . . "The large molecules are especially important, because it is they that carry biological specificity." To list a few functions which proteins fulfil: they are enzymes or essential parts of enzymes; they take part in many different forms in the intracellular and extracellular structure of the body. Antibodies are proteins, as are a number of pituitary hormones. In combination with nucleic acids they harbour inheritance factors. Another important aspect of proteins is that small alterations in their structure may lead to disease.

Despite all these different functions, proteins have some characteristics in common. All are constructed with a limited assortment of building blocks, the amino acids. They are all manufactured by the cells in the same way, and they are in a dynamic state of break-down and renewal. This dynamic state, so clearly expounded by BORSOOK (*2*) and SCHOENHEIMER (*8*) more than 20 years ago, which shows highly different rates of renewal and breakdown for the different proteins and under different conditions, makes the study of the organism as a whole extremely difficult.

Table 1. *Large protein stores in the body* ♂, 168.5 cm., 53.8 kg. From FORBES et al.: J. Biol. Chem. (U.S.A.) **208**, 359 (1953)

	g.
Total protein (N × 6.25)	10,006
Striated muscle	4,680
Skeleton	1,864
Skin	924
Adipose tissue	361
Estimate of blood:	
Haemoglobin	750
Albumin	250

Table 1, compiled from one of the few available analyses which have been made of the human body, lists the body's major protein stores.

The total amount of body protein is approximately 19% of the fresh weight. 45% of this protein is present in the muscle and 18% in the skeleton, while skin and adipose tissue account for another 10% and 4% respectively. These figures are quite interesting, but, upon closer examination, provide very little information. They would gain in importance if it were known how much is renewed daily, and what factors regulate the renewal rate. Then it would be possible to obtain an estimate of the extent to which these stores participate in daily protein metabolism, since their amino acids have to enter and mix with the amino-acid pool. Surprisingly little exact information on this question is available.

When talking about the renewal of proteins, we have to distinguish between the dynamic state of protein intracellularly or extracellularly and the renewal of whole cells, i.e. replacement of dying cells. The red blood cell may serve as a nice example of this last form of renewal. Haemoglobin seems to be stable during the life-span of an erythrocyte, but $^1/_{120}$ of our haemoglobin is renewed daily. Some cells have a very short life — as in the jejunal mucosa, for example, where most of the cells do not live longer than one day. There are indications that, once their protein is formed, this protein is stable for the rest of the day (*3*). This is certainly not the case in, for instance, the liver, where the life-span of the cell seems to be very long and the half-life of the protein *mixture* in the cells is not more than 4 days; here, therefore, we may speak of a real dynamic state.

If we come then to consider large protein masses, such as collagen, which has been estimated to constitute $^1/_3$ of our body protein, estimates of renewal rate are very complicated.

Neuberger et al. (*5*), about a decade ago, came to the conclusion that collagen of the tendon in rats is probably metabolically inert, with a half-life of several hundred days. He made some very cautious suggestions with relation to collagens in other sites. However, in recent years (*9*), at least 3 biochemically different fractions of collagen have been recognised, of which one may even have a half-life of 2 days. Furthermore, the pregnant uterus shows rapid deposition and, upon involution, rapid removal of collagen.

Before ending our remarks on the dynamic state of proteins, attention should be drawn to the opinion of several authors that only a minor part of the body protein is rapidly renewed. This, however, does not imply that the participation of large deposits of proteins (e.g. muscle proteins) in the amino-acid pool is small. Assuming an integrated half-life of 100 days for muscle proteins,

the amount of amino acids participating daily in intermediary metabolism would be 20 g.

If we now turn to the methods available for investigating protein metabolism in human beings in health or disease, the most widely used technique is the study of the nitrogen balance, based on the concept of dynamic equilibrium. In view of what I have

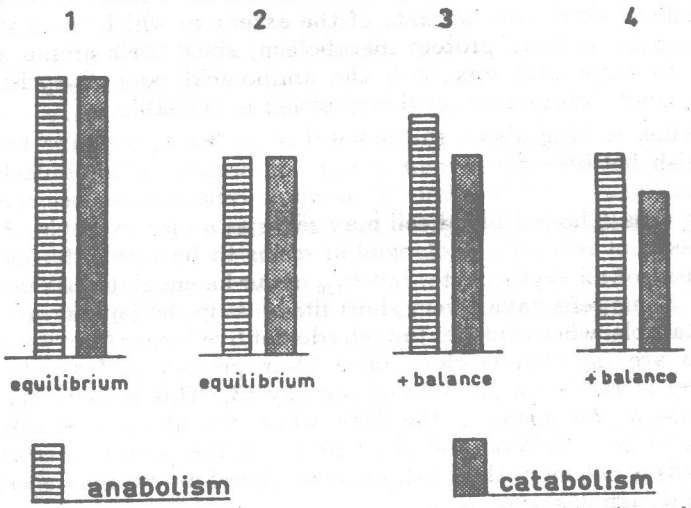

Fig. 1. Theoretical changes in anabolism and catabolism leading to a positive nitrogen balance

already said, it is clear that the information obtained with this technique will be limited. The results indicate only a difference between intake and output, being the integrated outcome of the anabolism and catabolism of many different protein pools. There is no evidence nor reason to assume that all these protein pools will move in the same direction and that the result of the balance study is representative of the trend in the body. Another relevant point is that a similar difference between anabolism and catabolism in a particular pool can be achieved at varying levels of anabolism, for example, as is shown in Fig. 1. It is probable that such explanations may account for ALLISON's classic observations (*1*) which indicate that achievement of nitrogen balance is dependent on the adaptation of the body to previous feeding periods.

This simple statement — to the effect that the body contains proteins with different renewal rates, which may be affected by disease or by active agents in a *different* way — already demonstrates the complexity of the study of protein metabolism. We should of

course like to define in cases of disease *which* proteins are affected in their metabolism and whether all the proteins of the body are involved or only special ones. With some diseases we have the clinical impression that most proteins undergo changes — as, for example, in Cushing's syndrome. The big protein stores seem to become reduced: skin and muscles are atrophic and the skeleton breaks down. Experimental protein depletion, however, shows a preference for certain organs. In his able monograph, ALLISON summarised the available information on the effect of depletion as follows: "In general the data reveal that total protein and enzymes of the liver are the most labile, while the total protein and the enzymes of the brain are most resistant. The total protein and enzymes of the ventricle of the heart are more resistant than those of the kidney, spleen or skeletal muscle and almost as resistant as those of the brain."

Recently it has been shown that in mice placed on a diet containing enough calories, but no protein, the colon and other tissues atrophy markedly, but the jejunum does not. This again also shows the danger of generalisations when the tissues are not sharply defined. To explain this type of result the term "labile protein reserves" is sometimes used, an expression which to me is not clearly defined. Perhaps some clarification on this point can be achieved during our conference.

It seems to me that the example of Cushing's disease, where the skin, too, is affected in such a marked way that it is paper-thin and shows ruptures appearing as large striae, raises the question as to why this phenomenon is not seen in other disease states, such as malnutrition. This brings us to the problem of how factors responsible for changes in protein metabolism operate. *Changes* in the amount of a protein present in the body at a given moment must be dependent on the amount synthesised per unit time and the amount broken down per unit time. If we assume that both processes are slowed down, the total amount present does not necessarily change. If, however, break-down is accelerated and synthesis does not increase or does not accelerate enough, large changes in the amount of protein will occur. The second possibility might well apply in Cushing's disease. The catabolic action of corticosteroids may affect the body as a whole, and the counter-regulation of increasing synthesis may be insufficient. Studies of the half-life of albumin during treatment with corticosteroids seem to support this hypothesis. The half-life of albumin under these conditions is shortened; synthesis, however, is also increased, but not enough to neutralise the increased degradation (7).