PROCEEDINGS OF THE BIOCHEMICAL SOCIETY

The 492nd Meeting of the Society was held jointly with the Nutrition Society at the University of Reading on Thursday and Friday, 27 and 28 March 1969, when the following papers were presented:

SYMPOSIUM ON 'BIOCHEMICAL ASSESSMENT OF PROTEIN NEEDS'*

The Absorption of Protein From the Intestine

By K. J. CARPENTER. (*School of Agriculture, University of Cambridge*)

The present state of knowledge about absorption of protein from the intestine will be reviewed and an attempt will be made to relate it to the other topics dealt with in this symposium.

Adaptation of Mammalian Protein Metabolism to Amino Acid Supply

By HAMISH N. MUNRO. (*Physiological Chemistry Laboratories, Department of Nutrition and Food Science, Massachusetts Institute of Technology, Cambridge, Mass., U.S.A.*)

Animals are equipped to make two major types of metabolic adaptation to the supply of nutrients. First, there are transient physiological adaptations to the intermittent intake of nutritionally adequate meals. These account for a large component of the diurnal variations that have been described in the protein metabolism of mammals (Wurtman, 1969). Thus liver polysome aggregation and the activities of some liver enzymes with short half-lives show rhythmic variations with a 24 hr. periodicity that appears to be related to the intermittent intake of protein in each meal. On the other hand, plasma amino acid concentrations undergo an independent diurnal cycle, which may be partly accounted for by amino acid deposition in muscle resulting from the influx of carbohydrate after each meal. Secondly, long-term adaptation of protein metabolism occurs when there is a decrease in the availability of essential amino acids in the diet. Under these circumstances, tissue constituents are lost to varying extents from different parts of the body. It is possible that changes in protein metabolism induced by prolonged amino acid deficiency may represent exaggeration of the normal diurnal metabolic adaptation. It is proposed to discuss mainly the subcellular mechanisms that allow short-term physiological adaptation to variations in amino acid supply.

* The full papers are published in the Proceedings of the Nutrition Society (1969) **28**, No 2.

During the absorptive period after consumption of protein the liver is subjected to an extensive increase in amino acid supply, much of which is diverted within the liver to hepatic protein formation, to plasma protein secretion and into catabolic pathways, so that only a relatively small part passes to the peripheral circulation as free amino acids (Elwyn, 1969). This increase in liver protein synthesis is accompanied by accumulation of polysome aggregates and diminution in dimer and monomer ribosomes and ribosome sub-units. There is evidence to suggest that the turnover of ribosomal RNA is retarded by this increase in the proportion of ribosomes present as polysomes; such an effect would account for the parallel changes in liver RNA and protein contents observed when amino acid supply is varied (Munro, 1968). The response of polysomes to amino acid influx is dependent on the mixture of amino acids undergoing absorption. Thus administration of various amino acid mixtures to rats after a brief period without food shows that polysome aggregation is dependent on the inclusion of tryptophan in the mixture (Proneczuk, Baliga, Triant & Munro, 1968). However, pre-treatment of the animals to alter the intracellular amino acid pools allows the administration of other amino acids to determine polysome aggregation in the liver (A. W. Proneczuk, Q. R. Rogers & H. N. Munro, unpublished work). The regulation of polysome aggregation in relation to amino acid supply appears to be effected by a cytoplasmic mechanism and can consequently be reproduced in cell-free systems (Baliga, Proneczuk & Munro, 1968).

In tissues that undergo cell multiplication, amino acid supply appears to influence cell division. Thus the rate of DNA synthesis in the intestinal mucosa is decreased by brief exposure to a protein-free diet (Munro & Goldberg, 1964), and multiplication of antibody-producing cells in the spleen in response to the challenge of an antigen is also inhibited (Kenney, Roderuck, Arnrich & Piedad, 1968; W. C. Cooper & H. N. Munro, unpublished work). Finally, protein deficiency at certain critical stages in the development of the rat affects the subsequent growth of cell numbers in vulnerable tissues (Winick

& Noble, 1966). This observation indicates that nuclear programming is influenced by amino acid supply.

- Baliga, B. S., Pronczuk, A. W. & Munro, H. N. (1968). *J. molec. Biol.* **34**, 199.
- Elwyn, D. (1969). In *Mammalian Protein Metabolism*, vol. 4, chapter 37. Ed. by Munro, H. N. New York: Academic Press, Inc. (in the Press.)
- Kenney, M. A., Roderuck, C. E., Arnrich, L. & Piedad, F. (1968). *J. Nutr.* **95**, 173.
- Munro, H. N. (1968). *Fed. Proc.* **27**, 1231.
- Munro, H. N. & Goldberg, D. M. (1964). In *The Role of the Gastro-Intestinal Tract in Protein Metabolism*, p. 189. Ed. by Munro, H. N. Oxford: Blackwells Scientific Publications.
- Pronczuk, A. W., Baliga, B. S., Triant, J. W. & Munro, H. N. (1968). *Biochim. biophys. Acta*, **157**, 204.
- Winick, M. & Noble, A. (1966). *J. Nutr.* **89**, 300.
- Wurtman, R. J. (1969). In *Mammalian Protein Metabolism*, vol. 4, chapter 36. Ed. by Munro, H. N. New York: Academic Press, Inc. (in the Press.)

Assessment of Protein Requirements by Nitrogen Balance

By D. S. MILLER (*Queen Elizabeth College, London W.8*) and P. R. PAYNE (*London School of Hygiene and Tropical Medicine, London W.C.1*)

During the last 100 years, published estimates of the requirements of adult man for protein have fallen from 120 g./day to around 30 g./day. Early estimates were intended, for example by Voit, to be used as dietary standards for workers and were based on recorded intakes of labourers. However, the tradition soon arose that liberal allowances of protein in the diet were essential for the maintenance of mental and bodily vigour, and as a protection against disease. Subsequent research has failed to establish the need for protein over and above that necessary to replace inevitable losses from the tissues and to provide for growth, and thus the criterion of nitrogen balance has come to be generally adopted as an indication of dietary adequacy.

A factorial method for calculating protein requirements will be described. An addition is made of the losses of nitrogen from the body by all routes including urine, faeces, skin, and where appropriate the products of conception, and milk. To these are added calculated nitrogen gains during growth. Allowance can be made for possible variations between individuals, to provide a total nitrogen requirement that can be expressed in terms of an ideal perfectly utilized protein.

Our understanding of the dependence of protein quality on amino acid composition now has a quantitative basis, and other factors that influence the utilization of dietary proteins have been studied and evaluated. In particular the interrelationship

between energy and protein metabolism will be examined in view of its importance to the world food problem.

The utilizable protein content of diets can be estimated, and evidence will be presented that man can survive in apparent good health on diets that provide sufficient to meet his estimated needs for nitrogen balance. No claim is made that such diets are culturally acceptable or that higher levels of protein are necessarily detrimental; merely that they confer no known benefit.

During the next 30 years the world's population will double and so will mankind's total nutrient requirements. The lowered estimates for protein allowances do not in any way diminish the magnitude of the task of meeting this growing demand, nor do they represent a lowering of dietary standards, but they should be seen as directing our efforts towards the production of more palatable foods of all kinds rather than searching for some apparently easier solution to the problem by increasing the supply of specific nutrients such as protein.

Protein Nutrition and Enzymes

By J. C. WATERLOW and JOAN M. L. STEPHEN. (*Medical Research Council Tropical Metabolism Research Unit, University of the West Indies, Kingston, Jamaica*)

In this paper we deal mainly with studies that have been made in man, because this Symposium is concerned with human protein nutrition.

One way of looking at the relationship between protein nutrition and enzyme activity is to regard enzymes as proteins whose specific enzymic activity serves as a natural label or marker. It is not, of course, strictly accurate to equate the activity of an enzyme with its amount. The two things may well be different *in vivo*, but when assays are made *in vitro*, under so-called optimum conditions, it is probably reasonable to take activity as a measure of amount.

In studies on human subjects measurements have most often been made on serum or plasma for obvious practical reasons. Enzymes that are found in the plasma must have been synthesized elsewhere; sometimes they are specific for a particular organ or tissue. Examples of plasma enzymes that are decreased in malnutrition with their probable tissues of origin are pseudocholinesterase (acylcholine acyl-hydrolase, EC 3.1.1.8) (liver), alkaline phosphatase (orthophosphoric monoester hydrolase, EC 3.1.3.1) (bone), amylase and lipase (pancreas) (Waterlow, 1969).

In theory a fall in the amount of a circulating enzyme, as measured by its activity, could result