NITROGEN BALANCE STUDIES IN
ADULT SOUTH INDIAN COLLEGE GIRLS

Thesis submitted to
Sri Venkateswara University
for the award of the Degree of
Doctor of Philosophy.

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INTRODUCTION
ACKNOWLEDGEMENTS

It has been my privilege to carry out this investigation under the able guidance of Dr. (Miss) Philomena Royappa Reddy, M.Sc., Ph.D. (Cornell). I am deeply indebted to her for her constant help and encouragement for the completion of this research project and for the preparation of this manuscript.

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meal frequency at a low protein intake with most of the protein provided by plant sources such as cereals, pulses and vegetables. The diet supplied enough protein to meet the minimal protein needs of the subjects, who were adult South Indian college girls. The effect of feeding the protein in two or six meals on nitrogen retention was assessed and the results have been presented in the first chapter.

Supplementation of protein concentrates or blends to diets and amino acid fortification and supplementation of foods have been widely advocated as a solution to the problems of protein nutrition. Innumerable studies have been carried out along these lines. Amino acid supplementation may, however, be considered a costly venture for developing countries. Clark et al (1956) presented data indicating that the relative proportions in which amino acids are utilized most efficiently by man are not known and that the effect of supplementing foods with amino acids cannot be predicted with certainty. Recently Hegsted (1972) observed that the information available on and the basic understanding of amino acid requirements and the relationship of these to protein quality are not satisfactory.

Of the many amino acids, eight are known to be essential for humans and of these, two, namely, lysine and methionine, are especially critical and limiting. A perusal of the amino acid composition of the various food stuffs, shows
that the proteins of egg, meat and milk are of high biological value, whereas those of vegetable origin are not only present in low amounts but are also deficient in lysine and methionine. The proteins of rice and wheat are particularly deficient in lysine and those of legumes or dhal in methionine. Combinations of the two are better than either of them singly, since the amino acids complement each other. Pulses were found to be good supplements to cereals in improving the quality of dietary protein (Phansalkar and Patwardhan 1956). If the inhabitants of Asian countries like Japan and India have survived the centuries, it is probably because the combinations of rice and dhal or rice and fish yield protein of reasonably good quality even if low in quantity.

Mutual amino acid supplementation can thus be achieved by a judicious combination of available foods. This approach was tried in a second experiment on adult college girls. Apart from a judicious combination of foods, the distribution of animal protein between meals was also considered. The experiment was done at minimal protein intake. The results are presented in the second chapter.

When the basic principles of nutrition were being established, it was essential to consider nutrients independently in order to identify their functions and to determine their quantitative needs. This approach, however, may overshadow the obvious fact that individual nutrients are not
consumed in isolation but as part of a diet providing a large number of variable components. For example, the nutritive value of proteins is usually determined under conditions ensuring their maximal utilization. Such studies do not adequately represent the fate of protein in natural diets, in which not only are the absorbed amino acids derived from several foods, but the nutritive value of the mixture is affected by the amounts of other dietary constituents, such as the energy yielding nutrients, the vitamins and the minerals. In the present investigation, protein utilization under these practical conditions has been assessed on adult college girls in the third, fourth, fifth and sixth experiments at a low protein intake. The results are presented in the third and fourth chapters.

Low protein intake in population groups often exists in parallel with low calorie intake. This can be explained by the fact that the chief sources of protein and calories in Indian diets are the staple food stuffs such as cereals and pulses. An increase or decrease in their consumption is bound to cause a parallel movement in both calories and protein. In the present context, the effect of varying the calorie intake alone on the utilization of protein was tested on adult college girls. Details of this study are presented in the third chapter. Some of the subjects in the first experiments were found to maintain positive nitrogen balance even at protein intakes which were less than their minimum requirement levels as per body weight. Hence, in
the third and fourth experiments the subjects were fed diets supplying minimum and less than minimum requirement levels of protein and an alteration was made in their calorie intakes. The effect of this on nitrogen retention was assessed.

The above mentioned studies were carried out with diets supplying 80 per cent of protein from foods of vegetable origin and 20 per cent from skimmed milk powder. In the fifth and sixth experiments, however, pure vegetarian diets commonly consumed by the poor population groups of South India were fed at minimal protein intakes. The effect of vitamin and mineral supplementation and of supplementing 300 mg of ascorbic acid to this diet was assessed. Nitrogen balances were determined. Details of these studies are presented in the fourth chapter.

An attempt was made to determine the endogenous nitrogen excretion of South Indian women as this has not been reported earlier. The protein free diet based on corn starch was not easily accepted by the subjects and addition of small amounts of other foods to make the diet acceptable resulted in an increase in the dietary protein level. Hence this diet could not be considered as protein free. The nitrogen balances were determined at a low protein level of 12 g. Data relating to this study is presented in the fifth chapter.

General discussion of the results of all the experiments is covered in the sixth chapter. The studies are presented in the following order.
1. The effect of meal frequency on the nitrogen balance of adult college girls consuming a diet satisfying minimum protein needs.

2. The effect on the nitrogen balance of adult college girls of a proper combination and distribution of foods between the meals of a diet, bringing about a mutual amino acid supplementation at the minimum protein intakes.

3. The effect on nitrogen balances of adult college girls of increasing the energy intake of a diet at minimum protein requirement levels.

4. The effect on the nitrogen balance of adult college girls of increasing the energy intake of a diet supplying protein at a level which is less than the minimum requirement.

5. The effect on the nitrogen balance of adult college girls of supplementing a vitamin mineral capsule to a South Indian vegetarian diet at minimum protein requirement level.

6. The effect on the nitrogen balance of adult college girls of supplementing 300 mg ascorbic acid to a South Indian vegetarian diet at minimum protein requirement level.

7. The nitrogen balances obtained on feeding a low protein diet to adult college girls.
Some subjects participated in more than one experiment, making it possible to compare the effects of more than one treatment. Conclusions were drawn based on responses obtained on the above mentioned treatments.

With more than 40 dietary constituents now established as being utilized by one species or another, the number of combinations that might be studied by a research worker is virtually infinite. However, it is important to choose a problem that may contribute to a certain extent to the improvement of the nutritional status of the people. In a developing country like India, the aim of the Nutrition Research Institutes such as the National Institute of Nutrition has been to "—discover the true nature of our serious nutritional problems and then to find a means of dealing with them" (Gopalan 1970). The food problem or calorie insufficiency happens to be a major issue in this country. This, coupled with the protein inadequacy, is the basic cause of several nutritional disorders prevalent among the vulnerable groups of the Indian population unlike in developed countries such as the United States (Goldsmith 1965). The best utilization of the limited supplies of food stuffs is important if the needs of the vulnerable groups of the Indian population, which has a low purchasing power, are to be met.

Considering that a minimum amount of protein is
available to the average Indian woman, attempts were made to assess the best way of feeding this available protein for maximal nitrogen retention. The attempts made have been listed above. Adult South Indian college girls were the subjects of this investigation. Similar studies at low protein intakes have not been reported with regard to South Indian women.

Human subjects were selected for these studies, as it was felt to be a worthy experience to face the problems and pleasures involved in human experiments and to obtain information relevant to human nutrition. Putting it in the words of Hegsted (1964), "—the studies done with man are obviously the most relevant to man, but usually have a very poor and undetermined degree of accuracy. Thus, the method may be insufficient to measure the differences expected. Studies with animals may or may not be more accurate, although they can be made so, but suffer from the fact that the results may not be applicable to human nutrition".

Nitrogen balance studies were done as nitrogen balance is a sensitive index to the state of protein nutrition (Munro 1964).