

## EFFECT OF VARIOUS SUPPLEMENTS ON THE PROTEIN VALUE OF PAKISTANI DIETS\*

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### Introduction

Survey figures from a number of developing countries of Africa, Asia and South America have shown the diets of the common people to be inadequate in respect of a number of dietary constituents, especially protein. International nutritional and health agencies such as Food and Agricultural Organization and the World Health Organization of the United Nations have stressed the need for developing new sources of protein rich foods for inclusion in human diets. A few recent efforts to improve low protein diets are the activities of I. N. C. A. P.,<sup>1</sup> the group under Scrimshaw in Gutemala, enrichment of maize with various natural foods and amino acids,<sup>2,3</sup> peanut flour as supplementary food,<sup>4</sup> enrichment of white flour with fish protein and its acceptability after baking,<sup>5</sup> leaf protein as human food,<sup>6</sup> multipurpose food based on a mixture of Bengal gram flour, peanut and sesame meal.<sup>7</sup>

In experimental studies with rats, Fox<sup>8</sup> reported improvement of certain Jamican diets with yeast, leaf protein and pulses. Frankul<sup>9</sup> studied the effect of addition of 'kushik' (fermented par-boiled wheat, yogurt mixture) on poor Iraqi diets. Koksai<sup>10</sup> made trials with eggs, sesame, yogurt and dried meat on some Turkish diets and infant food.

In the present study the effect of addition of Glaxo salt mixture, whole milk powder, leaf protein and vacuum oil dehydrated fish and meat on the protein value of East and West Pakistani diets has been investigated.

### Materials and Methods

Dehydrated meat and fish were prepared in a Vacuum Oil Dehydration Unit, developed at the Human Nutrition Research Unit, London. Leaf protein was obtained through the courtesy of Mr. N.W. Pirie, Rothamsted Experimental

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Station, Herts., England. Glaxo salt mixture and whole milk powder were made available at the Human Nutrition Unit.

The East and West Pakistan diets were prepared as described earlier.<sup>11</sup> The various supplements were thoroughly mixed with the diets for 2 hours in an electric food mixer.

The methods used in these investigations were the same as reported previously.<sup>12</sup>

### Results

The protein content of the diets and various supplements used are given in Table 1. The effects of various supplements on the protein values of the East and West Pakistan diets are shown in Tables 2 and 3.

It is seen from Tables 2 and 3 that the addition of minerals to the two national diets have not effected any improvement, showing that there is no gross deficiency of any particular mineral or trace element. This experiment also suggests that a diet having a low protein value cannot be improved by the addition of minerals. Such diets perhaps require a proportionately low mineral content to satisfy the growth requirements of an animal on this diet.

The addition of leaf protein, although it raised the protein values of the two diets, was less effective as compared with meat or fish. This is more noticeable in the case of the West Pakistan diet. Addition of 6% leaf protein has increased the N.D.p. cal. % of the West Pakistan diet, from 7.6 to 8.6, making it suitable for children and adolescents but not for nursing mothers who require N.D.p. cal. % = 9.5.<sup>13</sup> Meat or fish, when added to the extent of 6% (about 3% on fat free dry weight basis) increases the protein value of the West Pakistan diet from 7.6 to 9.5 and 10.1,

TABLE 1.—NITROGEN CONTENT OF THE PAKISTANI DIETS AND THE PROTEIN CONCENTRATES USED FOR THE IMPROVEMENTS OF THE DIETS.

	N. %	
East Pakistan diet	..	1.49
West Pakistan diet	..	2.26
Leaf Protein	..	7.40
Vacuum oil dehydrated fish	..	9.37
Vacuum oil dehydrated meat	..	7.42
Freeze dried egg	..	7.35
Whole milk powder	..	4.16

TABLE 2.—EFFECT OF THE ADDITION OF SALT MIXTURE AND PROTEIN CONCENTRATES ON THE PROTEIN VALUE OF EAST PAKISTAN DIET.

	No. of rats	Protein cal. %	NPU (op) %	NDp cal. %
East Pakistan diet alone	32	9.0	59	5.3
East Pakistan diet + 5% Glaxo Salt Mixture	16	8.9	55	4.9
East Pakistan diet + 6% leaf protein	8	11.0	62.5	6.9
East Pakistan diet + 6% Vac.-oil dehydrated fish (3.4% fat free dry weight basis)	8	11.7	62	7.3
East Pakistan diet + 6% vac.-oil dehydrated meat (3% fat free dry weight basis).	8	11.0	64	7.0
East Pakistan diet + 15% whole milk powder	8	12.0	68	8.2

TABLE 3.—EFFECT OF ADDITION OF SALT MIXTURE AND PROTEIN CONCENTRATES ON THE PROTEIN VALUE OF WEST PAKISTAN DIET.

Diet	No. of rats	Protein cal. %	NPU (op) %	NDp cal. %
West Pakistan diet alone	16	13.4	57	7.6
West Pakistan diet + 5% Glaxo Salt Mixture	16	12.7	57	7.3
West Pakistan diet + 6% leaf protein	8	14.9	58	8.6
West Pakistan diet + vac.-oil dehydrated meat (3.4% fat free dry-weight basis) fish	8	15.6	65	10.1
West Pakistan diet + 6% vac.-oil dehydrated fish (3.4% fat free dry-weight basis)	8	14.9	64	9.5
West Pakistan diet + 12% whole milk powder	8	14.8	62	9.2

respectively, which makes it suitable to meet the protein needs of nursing mothers.

Whole milk powder raised the N.P.U. and N.D.p. Cals.% in both the diets but almost double the amount is required to achieve the same degree of improvement as with meat or fish. Skimmed milk powder in which protein calories are increased due to the absence of fat is considered to be a better supplement than the whole milk powder.

### Discussion

The above results show that the factors which are basically involved in improving the protein value of a diet are: (a) protein concentration and b) net protein utilization. Protein supplements containing greater quantity of protein will be more effective than those containing protein diluted with fat or carbohydrate. Thus whole milk powder is considerably less effective as a protein source as compared with dehydrated meat or fish which contain a higher concentration of

protein. Increase in net protein utilization is subject to mutual supplementation of amino acids. The efficacy of animal protein supplements is due to the high content of certain essential amino acids such as lysine in which cereal proteins are generally deficient. Leaf protein is perhaps deficient in one or more essential amino acids, which though increasing the protein calories in the diet, does not raise NPU appreciably. Blending of leaf protein with other protein supplements would perhaps be more beneficial than using the leaf protein alone.

The mutual supplementation of amino acids could also be achieved in devising suitable vegetable protein mixtures. It will be, therefore, an interesting field of study to utilize hitherto untapped resources of protein-rich foods such as oil-seed meals and legumes, besides increasing the supply of animal proteins.

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#### A NOTE ON THE RELATIVE NUTRITIVE VALUE AND THE TOTAL AND "AVAILABLE" METHIONINE TRYPTOPHANE AND HISTIDINE IN SOME FISH FLOURS†

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FAO<sup>1</sup> has been able to formulate a chemical score technique as a result of certain observations in the field of protein nutrition. The factors leading to this technique are many and include the limiting factors for the utilization of a protein in a body, the amount of these limiting amino acids in any food correlating well with the nutritive value.

The protein value of any foodstuff can be predicted without taking recourse to experiments on animal. Carpenter et al<sup>2-4</sup> have shown that the available lysine rather than the total lysine is the limiting factor for the determination of the nutritive value and has developed a chemical technique for the measurement of the 'available' lysine. The assessment of the available lysine is important for those foods stuffs in which there is a chance of damage to proteins due to heat treatment and cooking.

Recently, Ford<sup>5-6</sup> has developed a microbiological technique, using the organism *S. zymogenes*, by which both the total and the available amino acids, specially the limiting ones and the Relative Nutritive Value (R.N.V) can be easily determined. The technique is based on the inheritance of the property of its growth by utilization of the amino acids in a similar manner as the test laboratory animals and of possessing the proteolytic character like the higher organisms. By the application of this technique on a number of various natural and processed foodstuffs and meals it has been observed that the available methionine, histidine etc., fairly correlate with the Relative Nutritive value and the latter on the other hand with the nutritive value (GPr and NPU) as evaluated by experiments<sup>4</sup> with animals.

†The work was carried out during study tour of the author in U.K.  
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The author has availed the opportunity of examining the samples of three freshwater fish flours by application of the above microbiological assay technique. These flours were processed in the Nutrition Section of the Food and Fruit Research Division of the East Regional Laboratories Dacca, for the preparation of the formulated high-protein diets for children and the other vulnerable groups of the population suffering from protein deficiencies. A report on their Chemical composition and the storage quality was submitted to the F.A.O. Regional Food Technology Seminar.<sup>7</sup> The present report gives a brief description of the results on the total and available methionine, histidine and tryptophane and the Relative Nutritive Value (R.N.V.) of these flours as evaluated by the microbiological technique.

The test organism *S. zymogenes*, obtained from the National Type Culture Collection of the Institute was maintained in the basal medium with all the salts and the vitamins as prescribed by Ford (loc cit) and thereafter grown in the amino acid medium of the similar composition as that of casein modified with the omission of the amino acid which is to be measured in the foodstuff—both the total and the available ones. For the estimation of total methionine, histidine and tryptophane the fish was hydrolysed in an autoclave and after adjustment to pH 7.6 and after necessary dilution, added to the culture tubes in graded doses and the tubes incubated for 28 hours. For each amino acid separate series of tubes were arranged against the control batches in which the amino acid under analysis was added in graded doses.

For the study of the available amino acid and Relative Nutritive Value, the flours were pre-digested with pepsin so as to make the available amino acid fully utilisable by the organism. In this case also, the graded doses of the digested mixture and the amino acids were incubated with the organism in the modified amino acid medium as mentioned above. For the study of the Relative Nutritive Value the amino acid mixture of the medium was made complete without any omission of any of the amino acids as done in the case of the study of the total and available amino acid.

The growth of the organism was measured both by turbidimetric and acidity determinations and the values of the total and available methionine, tryptophane and histidine and of the HNV were then calculated from the growth—dose response curve of the known amino acid mixture after incubation with organism. The values, so far obtained are summarised in Table 1.

The results indicate a very high proportion of the available methionine ranging from 2.40 to 2.86 g/16g. N and this comprises about 72 to 80 per cent of the total methionine. The higher proportion of the available methionine and also histidine and tryptophane in these samples is perhaps the limiting factor for elaborating higher Relative Nutritive Value (R.N.V.) as well. Some samples of fish meal prepared from herring, white, cod and pilchard examined (Ford) showed lower proportion of available methionine (1.4 to 1.9 gm) and less R.N.V. (60 to 85) as compared to the values obtained with the present samples. It so appears that the improved tech-

TABLE 1.—SHOWING THE "TOTAL AND AVAILABLE" METHIONINE, HISTIDINE AND TRYPTOPHANE CONTENTS AND THE RELATIVE NUTRITIVE VALUE (R.N.V.).

Nature of fish flour	Nitrogen percent	Methionine (g./16g. N)		Histidine (g./16g. N)		Tryptophane (g./16g. N)	Relative Nutritive* Value (R.N.V.)		
		Total	Available		Total			Available	
			Net	as percent of the total				Net	as percent of the total
Shoul fish flour ( <i>Ophicephelus striatus</i> )	12.5	3.56	2.36	74.6	3.23	2.67	82.6	0.89	112
Gazar fish flour ( <i>Ophicephalus marulius</i> )	13.2	3.64	2.64	72.5	3.20	2.41	75.3	0.82	112
Boyal fish flour ( <i>Osteriophysis, wallangu Atu</i> )	12.2	3.02	2.40	80.0	2.68	1.60	60	0.50	91

\*Relative to casein=100. The nutritive value of casein (NPU) with respect to egg is 75 p.c.

nique applied in processing of the fish flours under examination have not caused any damage to the quality of their proteins. Detailed results will be communicated in due course.

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## BOOK REVIEWS

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**History of Fishes.** P.H. Green Wood. 398 pp. 142 figs. and 7 plates. Ernest Beon Ltd., London, 1963. Price 21s.

Just as a sightseer visiting a city wishes to see relics of its past glory as also the busiest street throbbing with commercial life, the reader of the book under review is being presented with a survey of fish life including its past, present and even future, which deals with the utilization of fish products. Besides discussing fishes, dead and buried forever, there is mentioned an archaic type hitherto taken as extinct but which made its appearance as a "living fossil" (*Latimeria chalumnae*) first in 1938 in South Africa. There are cunning fishes able to disguise themselves to match with their surroundings and thus outwit their enemies (*Bothus podas*). There are dainty fishes, rainbow coloured, which can be called fairies in the sea. There are others misshaped and monstrous which strike as devils of the sea. There are luminescent fishes bearing miniature torches like fireflies on land. Some fishes can generate electric current perhaps to electrocute their assassins. For a credulous Muslim, there

is even a fish bearing eloquent testimony to the powers of the Almighty who has sent it into the world bearing a written document to the glory of God, and the message is specifically written in Arabic.

The book first appeared in 1931 and its second edition in 1963. The author is on the staff of the British Museum, a centre as much esteemed as any in the world. The duration of over 30 years, in between the two editions, must have further given all opportunities to make the book as ideal as any author could wish. Nevertheless a book of this standard should have had at least a couple of coloured plates, for the reviewer, with many others, does feel that some fishes make a better aesthetic appeal than even butterflies. One therefore misses coloured pictures. D'Arcy Thompson has ingeniously applied the principle of Cartesian coordinates to the problem of animal forms and explains how a teleost fish like *Diodon* in outline can be equated with the Sunfish *Orthogarisiscus*. Such a biomathematical study of fish life should have been represented. What the prospective purchaser would be happy to hear is that the