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THE WHO PROGRAMME OF TESTING OF PROTEIN-RICH FOOD MIXTURES

by

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Protein-calorie malnutrition is highly prevalent and is an important public health problem in many parts of the world. This deficiency is especially frequent among pre-school-age children whose protein requirements are higher per unit of weight than any other age-group. An inadequate dietary protein intake is responsible for this deficiency and any increase in production hand in hand with better distribution will be a major factor in preventing it. Other factors, such as control of the infectious diseases of childhood, better sanitary conditions, better education of mothers and **improvement** of socio-economic conditions are also aspects of prevention which must not be neglected. It is therefore obvious that prevention, to be really effective, must be based on a multi-disciplinary approach to the problem.

It is, however, not within the scope of this paper to review all the aspects of prevention and we should like to limit ourselves today to the question of protein foods. Since pre-school age children are the most vulnerable population group because of their relatively high protein requirements, WHO, FAO and UNICEF therefore have concentrated first of all on the protection of this special group, and programmes for the development of protein-rich foods specially intended for young children have been set up. At the outset of the programme i.e. in 1955

these organizations considered that flours prepared from oilseed press-cakes, fish, and pulses offered excellent prospects for the development of weaning or supplementary foods intended for this age-group. Various food mixtures based on cereals and protein concentrates or pulses were thus developed containing about 20 per cent of high quality proteins. Incaparina whose composition was worked out by Incap some ten years ago, is an example of a mixture of this type. It contains 27 per cent protein its main ingredients being maize flour and cottonseed flour.

Before these mixtures are accepted for human nutrition, it is evident that they must be tested for safety and suitability.

The procedure which is rather long includes the precise identification of the foodstuffs and of the various treatments they have undergone; bacteriological and chemical analysis, study of the effects of storage; study of possible toxicity using laboratory animals; and carefully controlled tests with children. These are followed by tests with large population groups. Some of these stages are well-known and therefore a few of them will only be reviewed here.

Firstly, it must be made sure that the various ingredients used in the preparations of the mixtures are of good quality and do not contain toxic concentrations of pesticides, fungicides or solvent residues. In addition, the ingredients should not contain toxins derived from moulds. If solvents have been used in their preparation, as is often the case with oilseed press-cakes, the residues should have been removed as completely as possible and any remaining amount should be below the toxic level. Whenever the ingredients used for producing these food mixtures have been treated with pesticides or fungicides, it must be made sure that such treatments have been carried out in accordance with the rules accepted in agriculture. If there is any doubt on this point or if it is impossible to obtain the necessary information, then the quantities present should be determined.

If one of the ingredients contains an antimetabolite this should be inactivated, destroyed or extracted. In the case of soya flour, for example, the anti-tryptic factor can be inactivated by heating. In the case of cottonseed flour, it is possible to extract the gossypol using selective solvents or azeotropic mixtures. Finally, certain pulses may contain haemagglutinins, haemolysins or other factors which are toxic for the nervous system and which should be borne in mind when such products are included in these food mixtures. Among the different toxins deriving from mould, aflatoxin produced by Aspergillus flavus is probably the most frequent, and the aflatoxin level in the food mixtures must be kept as low as possible. It is, however, often impossible under present production and storage conditions to ensure complete freedom from aflatoxin so that the Protein Advisory Group has recommended that its level, as measured by the methods developed at the Tropical Products Institute of London and by the United States Food and Drug Administration, should not exceed 30 parts per 1 000 million i.e. 30 micrograms per kg. This quantity was calculated on the assumption that a pre-school-age child would eat an average of 100 g per day of the food mixture containing aflatoxin.

Various chemical reactions may occur during processing. Thus in the Maillard reaction the amount of available lysine decreases and this, of course, affects the nutritional value of the product. Its bacteriological quality is also influenced by the cooking time.

By reason of the very different compositions and types of preparation of these food mixtures it is difficult to define bacteriological criteria which can be applied to each and all of them. While awaiting more ample information in this field the following maximal criteria are employed:

Micro-organisms

Aerobic mesophilic organisms	$\leq 10^5/g$
Enterobacteriaceae	$\leq 10^2/g$
Staphylococci	$\leq 10/g$

Concerning the enterobacteriaceae it is advisable to test for these first with a view to decreasing the number of subsequent tests required for Salmonella. When they are present then Salmonella should also be tested for and should be absent from ten 25 g samples. Finally, when fish or preparations based on fish are present in the food mixtures it is also advisable to test for the spores of Clostridium botulinum type E.

It is only when a food mixture has been prepared from ingredients recognized as suitable in such a way that there is no doubt in regard to safety of the product and when all the tests it has undergone have given a favourable result that the mixture can be tried out on children. Such trials on children include checks for acceptability and tolerance and, if necessary, an assessment of the nutritional value of the product. This comprises a study of the growth curve for infants or young children, of the nitrogen balance and possibly of other biochemical parameters.

WHO assumes responsibility for human tests on all products developed or distributed as part of the international FAO/WHO/UNICEF programme for the development of protein-rich foods. FAO and UNICEF share the responsibility for the tests preceding the human trials. Five centres all over the world are at present collaborating with WHO and testing these new protein-rich foods with children

and infants. Among those which have already undergone such trials or will do very soon, the following food mixtures may be mentioned.

Of the food mixtures based on wheat, five were developed jointly by UNICEF and FAO and a sixth by the U.S. Department of Agriculture and the U.S. Agency for International Development for possible distribution by UNICEF and other organizations. Pulses, soya flour or a mixture of soya flour and pulses are the chief sources of proteins in these foods. Some of them also contain a small proportion of skim milk. The mixtures are pre-cooked and their nutritive value as measured on rats is good. Three mixtures have already undergone clinical trials and their acceptability varied from good to excellent; they were well tolerated (see table I).

Seven mixtures of maize and soya have also been developed, including one by the U.S. Department of Agriculture and the U.S. Agency for International Development for distribution by UNICEF and other organizations. Some of these mixtures contain small amounts of skim milk powder or fish flour. They are pre-cooked and roller-dried, apart from one prepared by the extrusion cooking method. Their nutritional value is good. Three of these mixtures have already been tested on children and the results range from very good to excellent. They are also well tolerated (see table III).

Three mixtures of the Incaparina type have also been developed by UNICEF. The addition of five per cent of fish flour to the formula appears to increase the nutritional value more than does enrichment with 0.25 per cent lysine. It is also possible that the extrusion cooking method results in a slightly higher nutritional value than the pre-cooking and roller-drying procedure (see table 4). In general, however, these mixtures of maize and cottonseed flour have a lower nutritional value than mixtures of maize-soya, wheat-soya or wheat-pulses.

Clinical trials of a mixture of soya flour and whey have also been carried out. Although acceptability was good, there were certain problems in regard to tolerance; possibly the high lactose concentration in the product was responsible for the attacks of diarrhoea observed among the children. Another mixture, consisting of cat flakes and soya flour, has also been tried out with good results

as concerns acceptability and tolerance (see **table V**).

The programme has not yet been completed and several more months will be necessary before the full results for the different mixtures being tested as part of the FAO/WHO/UNICEF programme are available. The trials will have to be extended to large population groups using the mixtures which have given the best results before it will be possible to try to market these products. Some of them have nevertheless already reached the commercial or mass distribution stage, namely Incaparina, in Guatemala and Columbia; Superamine, in Algeria; and CSM which, although not sold commercially, is distributed on a very large scale throughout the world.

It is probable that in the future this testing programme will have to be extended still further and that other proteins, chiefly non-conventional proteins such as those derived from unicellular organisms, will also have to undergo rigorous testing.

Finally, more information should certainly be collected on the validity of methods for the fortification of cereals with pure amino acids. It has been shown in carefully controlled experiments that it is possible to increase the nutritional value of cereals by enriching them with pure amino acids but it remains to be proved that this can be applied on the scale of a whole population and that the method is economical and nutritionally valid when applied on such a scale.

In conclusion, it is obvious that more remains to be done, but this is a good sign for it shows that there are many possible lines of research in the attempt to overcome the protein gap and to prevent protein-calorie malnutrition.

TABLE I  
NET WORLD FOOD SUPPLY 1960-1962

	Annual food supply (millions of tons)	Annual protein supply (millions of tons)	Annual protein supply per person (kg)
Cereals, net <sup>a</sup>	423.8	38.84	12.30
Wheat, net <sup>a</sup>	145.9	17.08	5.41
Rice, net <sup>a</sup>	156.3	10.48	3.32
Coarse grains, net <sup>a</sup>	121.3	11.27	3.57
Starchy roots	255.8	3.06	0.97
Sugar	60.9	-	-
Pulses and nuts <sup>b</sup>	46.1	10.39	3.29
Vegetables	156.0	2.18	0.69
Fruits	127.9	0.88	0.28
Fats and Oils	28.1	-	-
Vegetables oils	15.8	-	-
Butter	5.4	-	-
Others	6.6	-	-
Meat	80.5	9.66	3.06
Beef and veal	27.8	4.10	1.30
Mutton, lamb	6.0	0.73	0.23
Pork	26.8	2.62	0.83
Poultry	8.2	0.98	0.31
Others	9.8	1.17	0.37
Eggs	13.6	1.48	0.47
Fish	30.0	5.65	1.79
Milk	210.7	7.36	2.33
Total		79.50	25.18

<sup>a</sup> After extraction

<sup>b</sup> Includes peanuts and other oilseeds used as well as coconuts.

TABLE II

## WHEAT BASED PROTEIN-RICH MIXTURES

	Tunisia	Algeria	Turkey	66/10/202	66/10/201	WSB
Wheat	31	28	40	32 (whole)	52 (whole)	73 (wheat fractions)
Chickpeas	34	38	20	30		
Lentils	26	18.5				
Soya flour			20	30 (full fat)	40 (full fat)	20
Skim milk		10	10			
Sugar	8	5	8	7	7	
Soybean oil						4
Vitamins, minerals, flavour	1	0.5	2	1	1	3
Process	Precooked	Precooked	Roller dried	Precooked Roller dried	Precooked Roller dried	
Protein content	20.9	20.2	25.4	25.8	26.6	22.0
PER (Stand)	2.08	2.40	2.27	2.21	2.15	2.31
NPU	60	71	61	63	63	65
Acceptability		+++		++	++	
Tolerance		good		good	good	



TABLE III  
CORN-SOYA PROTEIN-RICH MIXTURES

	CSM	66/10/203	Mx-49	Mx-50	Mx-51	Mx-52	Mx-60
Corn	68	43	40	58.5	41	48	41
Soya flour	25	40 (full fat)	38 (full fat)	35	42 (full fat)	30 (full fat)	42
Skin milk	5		5	5			
Fish protein concentrate						5	
Sugar		15.5 (white + raw sugar)	15.5		15.5	15.5	15.5
Vitamins, minerals flavour	2	1.5	1.5	1.5	1.5	1.5	1.5
Process	dry blending	Precooked Roller Dried	Precooked Roller Dried	Precooked Roller Dried	Precooked Roller Dried	Precooked Roller Dried	Cooking extrusion
Protein content	20.9	20.3	21.7	21.8	22.1	21.1	20.6
PER (Stand)	2.36	2.35	2.5	2.8	2.49	2.59	2.45
NPU	69	65	66	68	59	-	60
Acceptability	++	+++	+++	+	++	++	++
Tolerance	good	good	good	good	good	good	good difficult to prepare

TABLE IV

## CORN-COTTONSEED PROTEIN-RICH MIXTURES

	Mx-62	Mx-71	Mx-72
Corn	45.5	47	45.5
Cottonseed flour	32	26	32
Fish protein concentrate		5	
Lysine	0.25		0.25
Sugar	15.75	15.5	15.75
Cottonseed oil	5	5	5
Vitamins, minerals, flavour	1.5	1.5	1.5
Process	Cooking Extrusion	Precooked Roller Dried	Precooked Roller Dried
Protein content	22.9	22.2	21.2
PER (Stand)	1.98	2.16	1.59
NPU	57	63	53
Acceptability	++		
Tolerance	Good. Some difficulty in the preparation		

TABLE V  
MISCELLANEOUS

	Mx-37	Mx-39
Soya	33 (full fat)	15
Whey	66	
Rolled oats		85
Process	Concentration in vacuo spray drying	
Protein content	22	20.7
PER (Stand)	2.14	2.25
NPU	84	74
Acceptability	++	+++
Tolerance	Bouts of Diarrhoea	good