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MUTUAL AND AMINO ACID SUPPLEMENTATION OF PROTEINS

I. EFFECT OF SUPPLEMENTATION OF BLENDS OF GROUNDNUT, SOYA BEAN, SESAME AND COCONUT WITH THE LIMITING AMINO ACIDS ON THE NUTRITIVE VALUE OF THE PROTEINS

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During recent years, considerable amount of work has been carried out by many workers on the use of oil seed meals for overcoming protein deficiency in the diets of childern in developing countries.[1-5] The most abundant and promising sources of proteins in several developing countries are the meals obtained from groundnut, soya bean, cottonseed, sesame and coconut. The proteins of oilseed meals are partially deficient in one or more essential amino acids e.g. groundnut in lysine, methionine and threonine; soya bean in methionine; cottonseed in lysine and methionine; sesame in lysine and coconut in lysine, methionine and threonine.[6,7] Studies carried out by some workers have shown that sesame proteins supplement those of soya bean and groundnut and coconut proteins supplement those of groundnut to a significant extent.[8] Fortification of these proteins with the limiting essential amino acids have been shown to bring about a marked improvement in their nutritive value.[9] Methods for the large scale production of l-lysine, dl-methionine and dl-threonine at low cost, are available and fortification with these amino acids will increase only to a small extent the cost of the protein foods.[10, 11] It is evident from the above account that a vegetable protein supplement of high nutritive value can be prepared by judicious blending of two or more vegetable proteins and by fortification with the limiting essential amino acids. Since groundnut is the most abundant oilseed in India, the object of the present investigation was to study the nutritive value of protein blends based mainly on groundnut flour and containing varying amounts of soya bean, sesame and cocount meals and fortified with certain limiting essential amino acids.

Experimental

Materials : The oilseed meals and protein blends used in this study were prepared as follows :

Groundnut flour : This was prepared from good quality decuticled groundnut kernels free from spoilt seeds according to Subrahmanyan *et al.*[12]

Full-fat soya flour : This was prepared according to Narayana Rao *et al.*[13]

Sesame flour : This was prepared according to Subrahmanyan *et al.*[12]

Coconut meal : A lot of screw pressed cake of good quality similar to that used by Krishnamurthy *et al.*[13a] was obtained from Tata oil mills ltd, Ernakulam, India.

Protein blends : Protein blend I consisted of 60 parts of groundnut flour, 24 parts of soya flour and 16 parts of sesame flour. The proportions of proteins from the above sources were as 6:2.5:1.5. Protein blend II consisted of 52 parts of groundnut flour, 20 parts of soya flour and 28 parts of coconut meal. The proportions of proteins from the above sources were as 6:2.5:1.5 Protein blend III consisted of 39 parts of groundnut flour, 20 parts of soya flour, 13 parts of sesame fiour, and 28 parts of coconut meal. The proport-

ion of proteins from the different meals were as 4.5 : 2.5 : 1.5 : 1.5.

Fortification with amino acids : L-lysine monohydrochloride (AEC, France), dl-methionine (E. Merck) and dl-threonine (AEC, France) were used for the fortification of the proteins. The fortified protein blends contained 6.6g/16gN of lysine, 4.4g/16gN of the sulphur amino acids and 4.9g/16gN of threonine. Twice the quantity of dl-threonine was added as this has only half the biological activity of l-threonine. The compositions of the different protein blends are given in Table I.

Table I. *Percentage composition of the different protein blends*

Ingredients	Blend 1	Blend 2	Blend 3
Low-fat groundnut flour	60	52	39
Processed soya flour	24	20	20
Coconut meal	—	28	28
Sesame meal	16	—	13
Protein (Nx 6.25) %	47.0	40.1	39.9

Methods : *Essential amino acid composition of protein blends* : Lysine, methionine cystine and threonine contents of the oilseed meals were determined by microbiological methods.[14] The contents of these amino acids in the protein blends were calculated and the data are given in Table II.

Protein efficiency ratio : The protein efficiency ratios (PER) of the different protein blends were determined by the method of Osborne *et al.*[15] Freshly weaned male albino rats (21 days old) from the laboratory stock colony (Wistar strain) were allotted to different groups (8 per group) in a randomised block design The composition of the experimental diets was similar to that used by Panemangalore *et al.*[16] The diets contained about 19% protein from the different foods The procedure used for the preparation of the diets and the feeding of experimental animals was the same as that of Tasker *et al.*[17] The rats were weighed weekly and records of individual food intake of rats were maintained. The protein efficiency ratios were calculated from the data obtained. The results are given in Table III.

Results

PER of protein blends : The mean protein efficiency ratios of the three protein blends were 2.47, 2 20 and 2 38 respectively and were significantly higher than those of the proteins of groundnut, sesame and

Table II. *The lysine, methionine, cystine and threonine contents of the unfortified and amino acid fortified blends of groundnut, soya, sesame and coconut meals as compared with those of cow's milk, human milk and egg proteins (g 16gN)*

Sl. No.	Protein source	Lysine	Methionine	Cystine	Total sulphur amino acids	Threonine
1.	Low-fat groundnut flour	3.5	0.8	1.5	2.3	2.8
2.	Processed soya flour	6.3	1.4	1.7	3.1	4.0
3.	Coconut meal	4.0	1.7	1.6	3.3	3.0
4.	Sesame meal	2.7	2.8	2.3	5.1	3.2
5.	Protein blend I (groundnut+soya bean+sesame)	4.0	1.2	1.7	2.9	3.2
6.	,, ,, + lysine + methionine	6.5	2.6	1.7	4.3	3.2
7.	,, ,, + *threonine*	6.5	2.6	1.7	4.3	5.0
8.	Protein blend II (groundnut + soya bean + coconut)	4.2	1.2	1.6	2.8	3.0
9.	,, ,, + lysine + methionine	6.5	2.9	1.6	4.5	3.0
10.	,, ,, + threonine	6.5	2.9	1.6	4.5	4.8
11.	Protein blend III (groundnut +soya bean + sesame+ coconut)	4.1	1.5	1.8	3.3	3.4
12.	,, ,, + lysine + methionine	6.6	2.7	1.8	4.5	3.4
13.	,, ,, + threonine	6.6	2.7	1.8	4.5	5.1
14.	Skim milk powder	8.1	2.0	1.1	3.1	4.8
15.	Human milk[18]	6.6	2.3	2.0	4.3	4.5
16.	Egg, hen[18]	6.6	3.1	2.3	5.4	5.0

Table III. *Protein efficiency ratios of unfortified and amino acid fortified blends of groundnut, soya sesame and coconut meals as compared with skim milk powder*
(Level of protein in the diet, 10%; 8 male rats per group ; duration of experiment, 4 weeks)

Group No.	Source of protein in the diet	Initial body weight (g)	Gain in body weight (g)	Protein intake (g)	PER
1.	Groundnut flour	36.4	34.5	19.5	1.75
2.	Soya bean flour	36.9	51.4	21.3	2.40
3.	Sesame flour	36.5	34.9	20.0	1.74
4.	Coconut meal	36.6	36.0	16.9	2.10
5.	Protein blend I (groundnut+soya bean + sesame)	36.4	51.8	21.0	2.47
6.	,, ,, + lysine + methionine	36.4	60.0	22.3	2.68
7.	,, ,, + threonine	36.8	80.6	25.7	3.13
8.	Protein blend II (groundnut + soyabean + coconut)	37.1	45.9	20.7	2.20
9.	,, ,, + lysine + methionine	36.5	49.3	20.5	2.43
10.	,, ,, + threonine	36.6	70.4	23.8	2.94
11.	Protein blend III (groundnut +soya bean+ sesame+ coconut)	36.9	48.9	20.5	2.38
12.	,, ,, +lysine+ methionine	36.5	52.8	20.4	2.59
13.	,, ,, ,, + threonine	36.8	76.9	24.3	3.16
14.	Skim milk powder	36.9	73.9	22.5	3.29
	Standard error of the mean (91 df)				±0.08
	Critical difference at 5%			0.19	0.23
	,, ,, 1%			0.27	0.30
	,, ,, 0.1%			0.37	0.39

a — One tailed test b — Two tailed test

coconut (except protein blend II) and were nearly equal to that of soya bean (2.40).

PER of protein blends fortified with limiting amino acids : Fortification of the protein blends with l-lysine and dl-methionine brought about only a slight improvement while addition of dl-threonine along with l-lysine and dl-methionine brought about a highly significant increase in the PER of the blends. It is of interest to note that the PER's of the protein blends fortified with the three limiting amino acids (3.13, 2.94 and 3.16) are nearly equal to that (3 29) of milk proteins.

Discussion

The results reported here have shown that the PER of protein blends (2.20 to 2.47), containing 40 to 60% of the proteins from groundnut flour and the remaining from a mixture of soya, coconut and sesame flour, was higher than that of groundnut protein (1.75) and nearly equal to that (2.40) of soya proteins. In view of the fact that the production of soya bean in India is very much less than that of groundnut it could best be used in combination with groundnut flour. Further, sesame and coconut meals can be incorporated only in small amounts in a protein blend as the former has a slight bitter taste and the latter contains too much of fibre. Fortification of the protein blends with the limiting amino acids viz. lysine, methionine and threonine brought about a highly significant increase in the PER (2.94 to 3.16) to about the same level as that (3.29) of milk proteins, while fortification of the blends with lysine and methionine brought about only a slight improvement. These results indicate that all the three amino acids are equally limiting in the protein blends.

Summary

1. The protein efficiency ratios of the following protein blends (*Protein blend I*: 60 parts of groundnut flour, 24 parts of soya flour and 16 parts of sesame flour; *Protein blend II*: 52 parts of groundnut flour, 20 parts of soya flour and 28 parts of coconut meal; and *Protein blend III*: 39 parts of groundnut flour, 20 parts of soya flour, 13 parts of sesame flour and 28 parts of coconut meal) determined at 10% level of protein intake over a period of 4 weeks were 2.47, 2.20 and 2.38, as compared with values of 1.75, 1.74, 2.10 and 2.40

obtained for the proteins of groundnut, sesame, coconut and soya bean respectively. The results indicate that the proteins of the blends mutually supplement each other.

2. Fortification of the protein blends with the limiting amino acids viz lysine, methionine and threonine brought about a highly significant increase in the protein efficiency ratio ; the values for the PER of the fortified blends were 3.13, 2.94 and 3.16 as compared with 3.29 obtained for milk proteins.

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