

Vegetable protein mixtures for human consumption

The Development and Nutritive Value of INCAP Mixture 15, Based on Soybean and Cottonseed Protein Concentrates ¹

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SUMMARY

A series of studies were designed to develop and test vegetable protein mixtures based on soybean and cottonseed protein concentrates. The results obtained indicated that cottonseed and soybean protein complement each other when each provided the same concentration of protein in the diet. From this, INCAP vegetable Mixture 15 containing 25% crude protein, was formulated with 19% cottonseed flour, 19% soybean flour, 58% corn flour, 3% torula yeast, 1% calcium phosphate and 4500 I. U. vitamin A.

The mixture was tested in rats and dogs with good results. It has 80% of the nutritive value of casein. A significant improvement in protein quality was obtained when the mixture was supplemented with 0.2% methionine and 0.1% lysine or with 0.2% each of lysine, methionine and threonine. The corn can be replaced by other sources of calories such as cornstarch or banana flour, cornstarch and corn, processed in different ways. The studies also present information on mixtures based on cottonseed, soybean and skim milk.

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INTRODUCTION

Several laboratories in different parts of the world are attempting to develop and introduce protein-rich foods or mixtures made of locally produced materials, as part of the programs to prevent protein-calorie malnutrition in areas affected by this problem (1-4).

Previous work in the Institute of Nutrition of Central America and Panama (INCAP) dealt with vegetable protein mixtures based on the combination of cereal grains with sesame flour as the main source of protein (5, 6). This was followed by the development of mixtures based on cereal grains and cottonseed flour (7, 8) or cereal grains and soybean flour (9), where the combination between the components was derived from the results of biological tests with experimental animals. The combination selected is the best in terms of protein value, which can be obtained between the ingredients. It is superior to the protein quality of mixtures where consideration is given only to final protein concentration, and the amino acid balance is left to follow by itself.

The above formulations have been extensively tested in experimental animals, and the results confirm their relatively high nutritional quality. In most cases, corn was the cereal grain used in combination with the vegetable protein concentrate, but other cereal grains proved as effective or slightly better (10, 11). This finding increases their applicability, since there are areas where other cereal grains are more economical than corn. More versatile formulations are necessary, however, with respect to the main protein source as well as to the use of foods richer than cereal grains in carbohydrate content, which would still be high in quantity and quality of protein.

This paper describes the development and testing of a vegetable protein mixture, numbered INCAP 15, the basic protein of which consists of a combination between cottonseed and soybean protein concentrates. Different sources of additional calories were also tested.

MATERIALS AND METHODS

Description of ingredients.—The protein concentrates used for the development of the protein base of the mixture were

a pre-press hexane extracted cottonseed flour⁶ and soybean flour⁷. The cereal used in most of the studies was white corn meal, or toasted corn, later ground to a flour. The other sources of calories tested as replacements for corn were cornstarch and banana flour. Torula yeast⁸ served as a source of B-vitamins. All ingredients were analyzed for their nitrogen content by the Kjeldahl method.

BIOLOGICAL TESTS

a) *Development of basic mixtures.*—Six diets were prepared, of which Nos. 1 and 6 contained cottonseed or soybean flour, respectively, as the only source of protein, in a quantity to provide 10 or 20% protein in the diets. The rest of the diets consisted of mixtures in which soybean flour replaced cottonseed flour isoproteically in different proportions. The diets were supplemented with 3% torula yeast, 4% mineral mixture (12), 1% cod liver oil and 5% refined cottonseed oil. Cornstarch was used to adjust to 100%. All diets were supplemented with 5 ml of a complete vitamin solution (13) per 100 g and analyzed for their nitrogen content by the Kjeldahl method before feeding.

b) *Amino acid supplementation studies.*—From the previously described studies, Mixture 15 was formulated. This formula was then studied in order to determine its amino acid deficiencies. The tests were carried out at two levels of dietary protein. The basal diet providing 10 or 20% protein was supplemented with lysine, methionine and threonine added alone or in combinations.

c) *Nitrogen balance index in dogs.*—Four-month old dogs weighing 3.4, 3.7, 3.6 and 3.5 kg were used for the study. Two were fed casein and two were fed formula 15. The casein diet contained, in percentage: casein, 25; hydrogenated vegetable oil, 10; mineral mixture (12), 5; cod liver oil, 1; dextrin, 25; dextrose, 34. It was enriched with 5 ml of a complete vitamin solution (13) per 100 g of diet. Protein intake was decreased every 8 days from 6 g/kg body weight/day to zero, while the intake of calories remained constant at 130 cal/kg/day. After

6 Kindly supplied by Borgonovo Hnos., Zacatecoluca, El Salvador.

7 Kindly supplied by General Mills, Minneapolis, Minn., U. S. A.

8 Kindly supplied by Lake State Yeast Corp., Rhinelander, Wis.

allowing 15 days for protein repletion, the above was repeated. This time, however, the dogs fed casein were fed formula 15 and viceversa, so that all four animals had received the two protein sources.

The dogs were placed in individual metabolism cages and weighed daily. Each level of protein was fed for 8 days, obtaining 2 four-day balance periods. Food was provided every morning mixed with 600 cc of warm water. Urine was collected in dark bottles containing 1 cc of concentrated acetic acid. Feces were also collected daily. Upon completion of each 4-day balance period, samples of urine and the homogenized feces as well as of the diet were analyzed for total nitrogen by the Kjeldahl method. From these values, nitrogen retention and also the nitrogen balance index (14) of the two proteins were calculated.

d) *Biological tests with raw and toasted corn, cornstarch and banana flour.*—From the results of the study described under a), the best combination in terms of protein value between cottonseed and soybean protein concentrate was used to test variations of formula 15. In one experiment, raw corn in the formula was replaced by toasted corn, cornstarch or banana flour with and without sugar. In a second, the mixture with whole corn flour or with cornstarch was cooked, treated with amylolytic enzymes⁹ and dried by freeze drying or by the use of hot air. Cooking was carried out by mixing one part of mixture 15 with 5 parts of water and heating to boiling. The enzyme treatment was carried out by adding to the above mixture 0.1% of the enzyme. The protein in the experimental diets was diluted to 10% for PER assay in one experiment, while in a second experiment the mixtures were tested with minimum dilution of their protein content.

e) *Testing of commercially produced mixtures and variations.*—In order to obtain information on the nutritive value of the commercially produced mixture as compared to the experimental one, as well as to learn whether or not protein quality could be increased, a study was carried out in which the commercial mixture, the experimental formula, a variation of same, and a milk supplemented formula with casein as control, were all tested at the 10% protein level in the diet. The composition of the commercial and experimental for-

⁹ Enzyme Rhozyme H-39, kindly donated by Rohm & Hass, Philadelphia, Pa.

mulas was the same, with the exception that soybean flour used in the commercial mixture was manufactured in Colombia. The variation tested was formulated to contain 30% soybean flour, 20% cottonseed flour, 3% torula yeast, 46% corn flour and 1% calcium carbonate. The skim milk supplemented formula consisted in 88 parts of the commercial formula and 12 parts of skim milk.

f) *Procedure used with rats.*—Weanling white rats from the Wistar strain of the INCAP colony were used in all these experiments. Each diet was fed to groups of 8 rats. The animals were distributed among the experimental groups according to sex and weight so that all groups contained equal number of males and females and the average initial weight was the same. The rats were placed in individual all-wire screen cages with raised screened bottoms. Feed and water were provided *ad libitum*. Weekly records of food intake and weight changes were kept over a period of 28 days.

RESULTS

Table 1 presents the results of the study of different combinations between cottonseed and soybean flour. From PER and weight gain data at the 10% protein level in the diet, it appears that optimum complementation takes place when 60% of the dietary protein is derived from cottonseed flour and 40% from soybean flour. At the higher level of dietary protein, however, the differences in PER between the various combinations on the two protein sources tend to disappear. However, for practical purposes, and because the results suggested better protein quality with higher levels of soybean protein, formula 15 was formulated so that cottonseed and soybean protein would contribute equal amounts of protein to the final mixture 50% each.

For a 20% protein contribution to the mixture from the two components, the formula should contain 19% by weight of each. The amounts of the other components of the final mixture were formulated at 58% corn flour, 3% torula yeast and 1% calcium phosphate. In this mixture, 80% of the protein is derived from both cottonseed and soybean flour and 20% from the other components.

TABLE 1

**EFFECT OF REPLACING COTTONSEED FLOUR WITH SOYBEAN FLOUR ON WEIGHT GAIN,
AND PROTEIN EFFICIENCY RATIO IN RATS**

Percentage distribution of protein from		Protein distribution in diet ¹ g		Weight distribution in diet g		EXPERIMENT No. 1 ²		EXPERIMENT No. 2 ³	
CSF	SBF	CSF	SBF	CSF	SBF	Weight gain ⁴ g	PER	Weight gain ⁵ g	PER
100	0	19	0	38.0	0	67	1.91	154	1.74
80	20	15.2	3.8	30.4	7.6	86	2.31	153	1.82
60	40	11.4	7.6	22.8	15.2	101	2.50	156	1.84
40	60	7.6	11.4	15.2	22.8	102	2.46	153	1.92
20	80	3.8	15.2	7.6	30.4	109	2.60	162	1.93
0	100	0	19	0	38.0	101	2.65	154	1.93

¹ All diets contained 3% torula yeast, 4% mineral mixture (12), 1% cod liver oil, 5% vegetable oil and 5 ml of a vitamin solution per 100 g (13).

² Protein level in diet, 10%.

³ Protein level in diet, 20%.

⁴ Initial weight, 52 g.

⁵ Initial weight, 47 g.

TABLE 2

AMINO ACID SUPPLEMENTATION OF VEGETABLE MIXTURE 15

Amino acid supplement	Protein in diet ¹ %	Weight gained ² g	PER	Protein in diet ¹ %	Weight gained ² g	PER
None	10.1	101	2.52	17.3	157	2.07
L-lysine HCl (0.2%)	10.4	116	2.66	17.9	168	2.07
L-threonine (0.2%)	10.4	96	2.35	17.4	160	2.09
DL-methionine (0.2%)	10.2	109	2.43	17.5	159	2.04
L-lysine HCl (0.2%) + L-threonine (0.2%)	10.2	120	2.64	18.9	151	1.95
L-lysine HCl (0.2%) + DL-methionine (0.2%)	10.1	113	2.53	19.9	155	1.73
L-threonine (0.2%) + DL-methionine (0.2%)	10.0	114	2.64	18.6	151	1.91
L-lysine HCl (0.2%) + L-threonine (0.2%) + DL-methionine (0.2%)	10.7	144	2.86	19.3	167	1.91

¹ The protein 10% diet contained 40% of formula 15, while the higher protein diet contained 80%. Other supplements were 4% mineral mixture (12), 1% cod liver oil, 5% vegetable oil and 5 ml of a complete vitamin mixture (13) per 100 g.

TABLE 3
AMINO ACID SUPPLEMENTATION OF INCAP VEGETABLE
MIXTURE 15

Treatment	Protein In diet %	Average weight ¹ gain, g	PER
Vegetable Mixture 15	10.9	88	2.22
+ 0.1% L-lysine HCl	10.9	99	2.36
+ 0.2% L-lysine HCl	10.9	102	2.31
+ 0.1% DL-methionine	10.8	108	2.45
+ 0.1% L-lysine HCl + 0.1% DL-methionine	10.7	100	2.30
+ 0.1% L-lysine HCl + 0.2% DL-methionine	10.4	121	2.68
+ 0.2% L-lysine HCl + 0.1% DL-methionine	11.3	96	2.34
+ 0.1% L-lysine HCl + 0.1% DL-methionine + 0.1% L-threonine	10.8	123	2.72
+ 0.2% L-lysine HCl + 0.1% DL-methionine + 0.1% L-threonine	11.2	119	2.64
+ 0.2% L-lysine HCl + 0.2% DL-methionine + 0.2% L-threonine	10.9	140	2.95
Casein	11.7	121	2.87

¹ 28 days average initial weight 47 g/8 animals/group.

TABLE 4

THE EFFECT OF ADDING DIFFERENT SOURCES OF CALORIES TO THE BASIC MIXTURE OF SOYBEAN AND COTTON SEED PROTEIN CONCENTRATE OF MIXTURE 15

Calorie source added to basic soybean-cottonseed flour mixture ¹	Protein in diet %	Weight gain ² g	PER	Protein in diet %	Weight gain ² g	PER
Raw corn	12.0	113	2.24	24.4	157	1.51
Toasted corn	11.9	97	2.12	24.2	186	1.68
Cornstarch	12.6	108	2.14	24.0	184	1.67
Banana flour	12.4	109	2.00	23.9	111	1.21 ³
Cornstarch + sugar	12.0	105	2.15	24.6	160	1.47
Banana flour + sugar	12.2	107	2.03	23.5	155	1.46

¹ 58% of each calorie source was used in experimental mixture in place of an equal weight of raw corn. The diets contained 19% cottonseed flour, 19% soybean flour and 3% torula yeast. The protein was diluted with cornstarch to 10 and 24% protein. Other ingredients added were as before (see Table 2).

² Average initial weight, 44 g.

³ Two animals died of diarrhea.

TABLE 5

EFFECT OF COOKING AND DRYING WITH AND WITHOUT THE ADDITION OF AMYLOLYTIC ENZYME
ON THE NUTRITIVE VALUE OF VARIATIONS OF VEGETABLE MIXTURE 15

Treatment	Calorie source	Protein in diet %	Weight gain ¹ g	PER
Cooked-lyophilized	Cornstarch	12.6	121	2.22
Cooked-hot air drying	Cornstarch	12.5	132	2.25
Cooked-enzyme lyophilized	Cornstarch	13.2	118	2.07
None	Corn	12.1	131	2.23
Cooked-hot air drying	Corn	13.7	108	1.83
Cooked-enzyme lyophilized	Corn	12.3	116	2.30
Cooked-lyophilized	Corn	12.1	125	2.44

¹ Average initial weight, 51 g.

TABLE 6

PROTEIN COMPLEMENTATION BETWEEN VEGETABLE MIXTURE 15 AND SKIM MILK
AT TWO LEVELS OF PROTEIN IN THE DIET

PROTEIN DISTRIBUTION IN DIET		Protein in diet %	Weight gain ¹ g	PER	Protein in diet %	Weight gain ¹ g	PER
Vegetable Mixture 15	Skim milk						
100	0	10.9	118	2.42	20.0	188	1.88
80	20	11.5	136	2.52	20.3	178	1.88
60	40	10.6	156	2.98	20.0	168	1.93
40	60	10.3	135	3.00	20.4	161	1.78
20	80	11.4	148	2.86	19.8	157	1.93
0	100	11.0	142	3.03	20.8	156	1.95

¹ Average initial weight, 48 g.

The mixture was then subjected to testing. Table 2 shows the results of amino acid supplementation. None of the three amino acid, lysine, methionine or threonine, added alone, produced a significant increase in PER. Threonine addition caused a slight decrease in protein quality. Likewise, none of the combinations of two amino acids caused an increase in PER. The addition of the three, however, produced at the 10% protein level a significant increase in weight gain and protein efficiency. The addition of the three amino acids alone or combined when the diet contained close to 20% protein, did not result in any improvement.

Table 3 shows the results of a second study in which mixture 15 was supplemented with two levels of lysine and methionine alone and in combination. It can be seen that 0.1 or 0.2% L-lysine HCl produces similar weight gain and PER. The combination of 0.1% L-lysine HCl and 0.2% DL-methionine caused a significant increase in weight gain and PER over the results obtained with the control group. The response obtained with this combination was similar to that obtained when mixture 15 was supplemented with 0.1% each of lysine, methionine and threonine. Best results were observed as before when mixture 15 was supplemented with 0.2% each of lysine, methionine, and threonine.

Table 4 presents a series of results in which corn was replaced by toasted corn or by starch, and by banana flour alone or with sugar added. None of the substitutions changed the nutritive value of the mixture when the protein in the diet was around 12%. However, at a higher level of dietary protein, the animals fed the mixture with banana flour showed lower gains owing to a decreased intake which resulted in a lower PER. The effect of cooking and drying on the protein quality of mixture 15 is shown in Table 5. None of the treatments applied changed the PER of the mixture. As an exception, the group of animals fed the mixture prepared with corn, cooked and dehydrated by hot air, gave a slightly lower weight gain and PER.

The results concerning the complementation between the proteins of mixture 15 and skim milk are shown in Table 6 at two levels of dietary protein. At the lower level, replacement of the protein from mixture 15 with milk resulted in an increase in PER up to the point where 60% of the protein

of the diet was derived from mixture 15 and 40% from skim milk. Replacement of higher amounts of mixture 15 protein by skim milk did not increase or decrease PER. Similar results can be observed at the higher level of dietary protein, although the differences in PER between diets are smaller than when the diets contained 10% protein.

Table 7 shows the results of the tests carried out with the commercial and experimental formulas and other variations. Protein efficiency ratios were very similar among all formulations tested and lower than the value found for casein. However, weight gain was significantly higher from the formula using 30% soybean flour and 20% cottonseed flour and from the skim milk supplemented formula than that found from the commercial and experimental formula.

Table 8 presents the average nitrogen balance values of the dogs fed casein, or vegetable mixture 15, at decreasing levels of protein intake. At all levels studied, better nitrogen

TABLE 7
PROTEIN QUALITY OF COMMERCIAL FORMULA 15
AND VARIATIONS

Formula	Protein in diet %	Average weight gain, g	PER
Experimental ¹	10.8	87	2.22
Commercial ²	11.9	83	2.00
Variation No. 1 ³	12.9	114	2.25
Skim milk supplemented ⁴	12.3	103	2.19
Casein	11.9	116	2.52

¹ Contained, in percent: soybean flour, 19; cottonseed flour, 19; corn flour, 58; torula yeast, 3, and calcium carbonate, 1.

² Contained, in percent: same as above, except that the soybean flour was manufactured in Colombia.

³ Contained, in percent: soybean flour, 30; cottonseed flour, 20; torula yeast, 3; corn flour, 46, and calcium carbonate, 1.

⁴ Mixture of 88 parts of experimental formula and 12 parts skim milk.

TABLE 8

NITROGEN BALANCE DATA ON DOGS FED CASEIN AND VEG-
ETABLE MIXTURE 15 AT DECREASING LEVELS OF
NITROGEN INTAKE (Average 4 dogs)

CASEIN Nitrogen			MIXTURE 15 Nitrogen		
Intake	Absorbed mg/kg/day	Retained	Intake	Absorbed mg/kg/day	Retained
795	738	346	790	585	274
717	659	297	708	491	168
642	590	264	642	460	193
564	515	268	549	385	162
481	438	186	486	337	138
407	363	161	410	283	104
326	225	107	319	223	78
252	213	51	237	160	32
163	129	2	163	97	-14
91	59	-48	—	—	—
47	25	-61	67	28	-64
0	0	-105	0	—	-113

retention values were obtained with casein than with mixture 15, at comparable levels of nitrogen intake. The data likewise shows that nitrogen absorbed was also higher with casein. The same is true when nitrogen retention values are expressed on the basis of nitrogen intake. However, when nitrogen retention is expressed on the basis of the nitrogen absorbed, the values of casein and mixture 15 are similar.

The regression of nitrogen intake to nitrogen retention was $NR = -104.95 + 0.60 NI$ for casein and $NR = -93.17 + 0.47 NI$ for mixture 15, suggesting that mixture 15 has about 80% of the nutritional value of casein.

DISCUSSION

The results of the present investigation on the formulation of vegetable protein mixtures, using soybean and cottonseed protein concentrates as the main protein sources, indicate that

the amino acid pattern of these two proteins complement each other efficiently when each contributes an equal amount of protein to the mixtures. The type of complementation is one in which the nutritional quality improves to a certain point, when one protein is replaced by the other at isonitrogenous levels of dietary protein. Further changes in the ratio between cottonseed and soybean protein do not increase or decrease the protein value of the mixture. In other words, the protein value of soybean protein remains the same up to a certain point, when it is replaced by cottonseed protein and it decreases as cottonseed increases in the diet, at equal levels of protein intake. These results can be explained on the basis of the higher levels of lysine in soybean proteins as compared to the level for the same amino acid in cottonseed protein. On the other hand, cottonseed and soybean protein contain similar levels of sulphur containing amino acids. The decrease in the protein value of soybean, when it is replaced by cottonseed protein occurs when lysine and the methionine in the mixture balance each other. However, both amino acids are not present in sufficient amounts. This is indicated by the results of amino acid supplementation of the mixture, which indicated that the lysine addition alone caused a slight increase in protein quality, but in the presence of 0.2% methionine resulted in a significant increase in PER of the mixture.

It is of interest to note also that threonine is the third limiting amino acid after lysine and methionine, particularly when the higher levels of these two amino acids were added. This is a good example of the care which should be taken when proteins are supplemented with their limiting amino acids, as discussed by Rosenberg (15) and Harper (16). It should be pointed out that the cost of the mixture would not increase significantly if it were supplemented with 0.1% when proteins are supplemented with their limiting amino were added, not only would it be more expensive, but in this case threonine would also have to be added.

Formula 15 was developed to contain 19% cottonseed and 19% soybean protein, because 25% was the level of protein chosen for the final mixture using corn as calorie source. If the calorie sources are materials which do not provide protein, such as cornstarch, the desired levels can be obtained

by modifying the amounts of cottonseed and soybean protein in the mixture. Mixtures with any level of protein concentration can be prepared up to the limit provided by combinations of the protein sources which give the best protein value. For example, a protein concentrate containing 50% protein could be formulated with 50 g of cottonseed protein and 50 g of soybean, since each one of these two concentrates contains 50% protein. The protein efficiency will be maintained in all the mixtures; this indicates the advantage of formulating vegetable protein mixtures through the use of studies in rats, as indicated in this report.

The results also indicate that several foods can be used as calorie sources in mixture 15, without changes in the protein efficiency ratio as was reported before for other mixtures (6, 10, 11). Furthermore, treatment of the cereal grain with hydrolytic enzymes for carbohydrate does not change its quality.

When the tests were carried out at a 20% protein level in the diet, the PER values were lower than when the tests were performed at 10% protein in the diet. This is to be expected since it has been shown that lower PER values are obtained as the protein level in the diet increases (17-19).

The same type of complementation studies was performed between mixture 15 and skim milk as those carried out between cottonseed and soybean protein. The results obtained can be used to formulate mixtures of cottonseed, soybean and skim milk proteins. From the results obtained in this study, a mixture of high protein quality and with 29% protein can be formulated, consisting of: 40% corn flour, 14% soybean protein concentrate, 14% cottonseed protein concentrate and 32% skim milk. The PER value of such a mixture would be close to 3.0.

From the results obtained when the commercial and experimental formulas were tested, it can be concluded that the quality of the soybean flour produced in Colombia is similar to that employed in the experimental formula. The two modifications did not show a higher PER value although better weight gain was obtained probably due to a higher food intake.

The nitrogen balance results indicated that mixture 15 has 80% of the protein quality of casein, which is an adequate figure for human feeding. When the value is calculated on

the basis of the nitrogen absorbed, mixture 15 has the same quality as casein. This difference is due to the lower amounts of nitrogen absorbed when mixture 15 was fed as compared to casein. Similar results in animals and humans have been reported before for INCAP mixtures 9 and 14 (7, 9). These results can be interpreted to mean that the quality of the nitrogen absorbed is higher. In other words, the amino acid balance absorbed is as high as that from casein. There is no available information to explain this behavior of vegetable protein mixtures. The lower absorption from mixture 15 is probably due to indigestible protein resulting from the processing to which the protein sources were subjected. On the other hand, urinary nitrogen excretion was higher when casein was fed as compared to mixture 15. These results suggest that casein is catabolized faster than the protein from mixture 15. This problem is under investigation.

Finally, mixture 15 has been tested in children; the protein value of mixture 15 proved to be similar to that of skim milk when fed at levels of intake from 1 to 2 g protein/kg/day; such studies are to be reported elsewhere. Mixture 15 is already in commercial production with good acceptability in two Latin American countries.

RESUMEN

Mezclas de proteínas vegetales para consumo humano. Desarrollo y valor nutritivo de la mezcla INCAP 15 basada en concentrados de proteínas de soya y algodón.

Se diseñaron varios estudios conducentes al desarrollo de mezclas vegetales a base de concentrados proteicos de frijol de soya y de semilla de algodón. Los resultados obtenidos indican que las proteínas de ambas semillas se complementan entre sí cuando cada una aporta a la dieta la misma concentración proteica. Así, la Mezcla Vegetal INCAP 15, que contiene 25% de proteína cruda, se elaboró de acuerdo con la siguiente fórmula: 19% de harina de semilla de algodón, 19% de harina de frijol de soya, 58% de harina de maíz, 3% de levadura torula, 1% de fosfato de calcio y 4.500 U. I. de vitamina A.

Utilizando perros y ratas se llevaron a cabo pruebas del valor nutritivo de la mezcla, cuyo valor proteico asciende a 80% del de la caseína, con buenos resultados. Al suplementar la mezcla que proporcionaba niveles bajos de proteína con 0.2% de metionina y 0.1% de lisina, o con 0.2% de cada uno de los aminoácidos lisina, metionina y treonina, se logró una mejoría significativa. Los estudios confirman que el maíz puede ser

reemplazado con otras fuentes de calorías, tales como harinas de almidón de maíz, de banano y de maíz procesado en diferentes formas. Asimismo, proporcionan datos referentes a mezclas elaboradas a base de semilla de algodón, frijol de soya y leche descremada.

BIBLIOGRAPHY

- (1) Bressani, R. & Béhar, M.—The use of plant protein foods in preventing malnutrition. In: *Proc. 6th Internat. Cong. Nutrition*, Edinburgh 9-15 August, 1963. Edinburgh, E. & S. Livingstone Ltd., 1964, p. 181-206.
- (2) Milner, M.—Peanuts as a protein resource in international feeding programs. *Food Tech.* 16 (7): 46-53, 1962.
- (3) Bressani, R.—The use of cottonseed protein in human foods. *Food Techh.* 19: 1655-1662, 1965.
- (4) Altschul, A. M.—Edible seed protein concentrates: their role in control of malnutrition. *Israel J. Med. Sci.* 1: 471-479, 1965.
- (5) Squibb, R. L., Wyld, M. K., Scrimshaw, N. S. & Bressani, R.—All-vegetable protein mixtures for human feeding. I. Use of rats and baby chicks for evaluating corn-based vegetable mixtures. *J. Nutrition* 69: 343-350, 1959.
- (6) Bressani, R., Aguirre, A. & Scrimshaw, N. S.—All-vegetable protein mixtures for human feeding. II. The nutritive value of corn, sorghum, rice and buckwheat substituted for limetreated corn in INCAP Vegetable Mixture Eight. *J. Nutrition* 69: 351-355, 1959.
- (7) Bressani, R., Elías, L. G., Aguirre, A. & Scrimshaw, N. S.—All-vegetable protein mixtures for human feeding. III. The development of INCAP Vegetable Mixture Nine. *J. Nutrition* 74: 201-208, 1961.
- (8) Scrimshaw, N. S., Béhar, M., Wilson, D., Viteri, F., Arroyave, G. & Bressani, R.—All-vegetable protein mixtures for human feeding. V. Clinical trials with INCAP Mixtures 8 and 9 and with corn and beans. *Am. J. Clin. Nutrition* 9: 196-205, 1961.
- (9) Bressani, R. & Elías, L. G.—All-vegetable protein mixtures for human feeding. The development of INCAP Vegetable Mixture 14 based on soybean flour. *J. Food Sci.* 31 (in press).
- (10) Bressani, R., Aguirre, A., Elías, L. G., Arroyave, R., Jarquín, R. & Scrimshaw, N. S.—All-vegetable protein mixtures for human feeding. IV. Biological testing of INCAP Vegetable Mixture Nine in chicks. *J. Nutrition* 74: 209-215, 1961.
- (11) Bressani, R., Elías, L. G. & Scrimshaw, N. S.—All-vegetable protein mixtures for human feeding. VIII. Biological testing of INCAP Vegetable Mixture Nine in rats. *J. Food Sci.* 27: 203-209, 1962.
- (12) Hegsted, D. M., Mills, R. C., Elvehjem, C. A. & Hart, E. B.—Choline in the nutrition of chicks. *J. Biol. Chem.* 138: 459-466, 1941.
- (13) Manna, L. & Hauge, S. M.—A possible relationship of vitamin B₁₂ to orotic acid. *J. Biol. Chem.* 202: 91-96, 1953.
- (14) Allison, J. B. & Anderson, J. A.—The relation between absorbed nitrogen, nitrogen balance, and biological value of proteins in adult dogs. *J. Nutrition* 29: 413-420, 1945.

- (15) Rosenberg, H. R.—Amino acid supplementation of foods and feeds. In: **Protein and Amino Acid Nutrition**, edited by A. A. Albanese. New York, Academic Press, 1958, p. 381-417.
- (16) Harper, A. E.—Balance and imbalance of amino acids. **Ann. New York Acad. Sci.** 69: 1025-1041, 1957-58.
- (17) Morrison, A. B. & Campbell, J. A.—Evaluation of protein in foods. V. Factors influencing the protein efficiency ratio of foods. **J. Nutrition** 70: 112-118, 1960.
- (18) Barnes, R. H. & Bosshardt, D. K.—The evaluation of protein quality in the normal animal. **Ann. N. Y. Acad. Sci.** 47: 273-296, 1946.
- (19) Henry, K. M. & Kon, S. K.—Effect of level of protein intake and of age of rat on the biological value of proteins. **Brit. J. Nutrition** 11: 305-313, 1957.