Effect of Blending Plant Materials on Protein Quality. I. Peanut and Citrus Seed Proteins¹

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ABSTRACT

Oilseeds are receiving much attention as sources of edible protein, even though some are generally low in certain essential amino acids such as lysine, methionine, tryptophane, and/or isoleucine. One of the best ways for correcting the amino acid balance is to blend two or more oilseed proteins. Defatted peanut flour (low in methionine) and a citrus seed flour reportedly high in methionine were blended and evaluated chemically for protein solubility, amino acid composition, gel electrophoretic protein patterns and methionine and available lysine levels. Solubility of peanut proteins is much higher than that of citrus seed proteins, precluding the use of blends in preparation of protein co-isolates for some types of beverages. Because of their properties, these blends would probably find better use in cloudy, fruit-flavored, or milktype beverages or in solid food items such as meat extenders, bakery goods, dry soup, or gravy mixes.

Keywords: peanut flour, citrus seed flour, peanut-citrus protein blends, protein solubility, methionine.

Projected shortages of food proteins throughout the world have increased the importance of plant materials as economical, readily available sources of protein for humans (6). Oilseeds and legumes are particularly important, although most are poor sources of methionine, one of the essential amino acids. All plant or animal proteins added to foods to improve nutritive value, to add functional properties, or to extend the protein already present have unique functional, chemical, and nutritional characteristics. There is no one protein that possesses all of the functional or nutritional properties desirable for its exclusive use in foods or beverages. To provide good amino acd balance where plants are the sole source of protein in the diet, many nutritionists blend two or more proteins in ratios that raise the levels of limiting amino acids to an optimum range.

Peanut proteins are low in methionine (8, 12) but they have other functional properties that make them desirable as a source of edible plant protein (2, 8, 15). Conkerton and Ory (8) reported 1.0 g of methionine per 16 g nitrogen for defatted peanut meal. In 1972, Braddock and Kesterson (4) reported that defatted grapefruit seed meal contained more methionine than orange seed meal (2.6 g/16 g N vs. 1.8 g/16 g N, respectively) and both of these were higher than soybean meal (1.3 g/16 g N). Florida alone produces over 38 million lb of citrus seed flour anually, but this is currently sold for animal feed supplements (4). The potential application in foods has never been examined.

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Peanut proteins have been blended with many plant products for human consumption: chickpea flour in India and Ethiopia (18), wheat flour for breadmaking (13), wheat, Ragi, sorghum, or soy flour (9, 19), cottonseed flour (3); wheat, cottonseed, and/or soy flour, fortified cake donuts (14), and rice (21, 22). Free methionine added to food products raises the nutritional levels, but introduces flavor problems through the formation of thiol compounds during processing. Dunlap et al. (10) reported that methionine supplementation of pinto beans in tomato sauce introduced off-flavor in the cooked product. The possibility of blending citrus seed meal with peanut meal to improve the methionine balance for food and/or beverage applications seemed worthy of attention. This report describes studies of some chemical properties of different blends of these two seed proteins that could affect their utilization in food or beverage products.

Materials and Methods

Defatted flour was prepared from white skin Pearl variety peanuts as described by Conkerton and Ory (8). Citrus seed flour prepared from defatted grapefruit seed meal was a gift from Dr. R.J. Braddock, University of Florida. Peanut: citrus seed flour blends were prepared by accurately weighing the appropriate amounts for 3:1, 1:1, and 1:3 blends. The flours were first mixed thoroughly with a spatula on aluminum foil on a flat surface to break up any clumps, then transferred to sealed jars and agitated for at least 10 min. All reagents used for buffers, protein solubility studies, gel electrophoresis, etc., were obtained from commercial sources.

Amino acids were determined by gas chromotography as described by Conkerton (7). Methionine was determined by gas chromotography and by the cyanogen bromide method of Finlayson and MacKenzie (11). Polyacrylamide gel electrophoresis was conducted by the method of Ornstein (17) as modified by Cherry *et al.* (5) for peanut proteins. For nitrogen solubility studies, 1 g of the flour was mixed in the desired buffered solution (1:10, w:v) and stirred at room temperature for 1 hr then centrifuged at 14,000 x g for 30 min at 23° C. Nitrogen in the supernatant was determined by macroKjeldahl analysis. Available lysine values were determined under contract by a commercial analytical laboratory.

Results and Discussion

There were only slight differences in the color of the peanut flour, peanut-citrus flour blend, and the citrus seed flour. The peanut flour is whiter than the creamy-colored citrus flour and the 1:1 peanut-citrus flour blend. Figure 1 illustrates the nitrogen solubility curves of peanut flour, citrus seed flour, and a 1:1 peanut-citrus flour blend. Peanut proteins are much more soluble (A) than citrus seed proteins (C). Solubility of the 1:1 blend of peanut and citrus seed proteins (B) is intermediate. It was surprising to find that protein solubility of citrus seed flour was so low at all pH's between 2 and 10. This low solubility of citrus proteins could limit their use in blends with peanut flour in certain types of beverages.

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Fig. 1. Solubility curves for peanut and citrus seed proteins. A, peanut proteins; B, a 1:1 blend of peanut and citrus seed proteins; C, citrus seed proteins. Conditions described in Materials and Methods.

The differences in solubility of peanut and citrus seed proteins are vividly depicted in the gel electrophoretic patterns in Figure 2. The lower solubility of citrus proteins shows up as a smaller major band and two or three minor bands near the origin. The peanut-citrus seed blend (1:1) shows a typical peanut protein pattern. It is obvious that the greater solubility of peanut proteins masks that of the citrus seed proteins in blends.

Table 1 lists the essential amino acids of the peanut flour, peanut: citrus seed flour blends (3:1, 1:1, and 1:3), and the citrus seed flour. The methionine content of peanut meal is lower than that of citrus seed flour. Blends of peanut and citrus seed flour were about equal to the citrus flour alone in methionine content or slightly higher than that of peanut flour. Cystine was also lower in peanut flour compared to the citrus seed flour, with intermediate values for the blends. The total sulfur amino acids content in citrus seed flour is much higher than that of peanut flour, indicting a possible means for raising methionine levels in peanut protein supplemented products.

The methionine values in Table 2 were obtained by the gas chromotographic method of Conkerton (7)



Fig. 2. Polyacrylamide gel patterns of peanut and citrus seed proteins. P, peanut proteins; P:C, a 1:1 blend of peanut and citrus seed proteins; C, citrus seed proteins. Migration is from top to bottom. Other conditions listed in Materials and Methods.

Table 1. Essential	l amino acid	contents o	f peanut	and	citrus	seed
protein blends ((g/16g N).					

Amino	P	P:C	P:C	P:C	С
Acid	100%	3:1	1:1	1:3	100%
ISOLEU	3.3	3.3	2.6	1.9	3.0
LEUC	6.2	7.6	6.8	6.7	5.4
LYS	3.1	3.3	3.3	3.5	3.1
METH	0.9	1.3	1.1	1.2	1.3
CYST	0.9	1.4	1.6	1.4	1.8
PHEN	5.2	5.2	4.8	4.9	4.7
TYRO	4.0	3.3	3.6	3.8	2.6
THRE	2.4	3.1	2.8	2.5	3.2
VAL	4.0	4.6	3.6	5.6	4.3
ARG	11.1	8.0	10.8	10.3	8.3
HIST	2.1	1.7	2.0	1.9	1.5

P = peanut flour; C = citrus seed flour.

and by the cyanogen bromide method of Finlayson and Mackenzie (11), a superior method that employs cyanogen bromide to release the methyl group of

Table 2. Methionine assays of peanut and citrus seed protein blends by two methods (g/16g N).

	Amino Acid	CNBr	Literature	
Sample	Analysis	Method	Value	
P (100%)	0.9	1.2	0.9*	
P:C :: 3:1	1.3	1.5		
P:C :: 1:1	1.1	1.5		
P:C :: 1:3	1.2	1.5		
C (100%)	1.3	1.9	2.6**	

* This report.

** Braddock and Kesterson, 1972.

 Table 3. Theoretical nutritional evaluation of peanut and citrus seed protein blends.

Sample	Simplified Chemical Score*	Available Lysine	
		%	
P (100%)	39	1.8	
P:C :: 3:1	54		
P:C :: 1:1	57	2.2	
P:C :: 1:3	54		
C (100%)	67	2.5	

* Only lysine, methionine (values from CNBr Method,

Table II) and cystine were used to calculate the

chemical score.

methionine, forming methyl thiocyanate which can be measured directly in the gas chromatograph. With this procedure there is no loss of methionine due to oxidation during hydrolysis and the method yields more accurate values. As seen in Table 2, values obtained by the cyanogen bromide method for these flours and blends are approximately 25% higher than those obtained by gas chromatographic analysis.

Several methods have been reported for predicting theoretical nutritional values of plant protein mixtures, based on average values for the essential amino acids, to give what is designated as a "chemical score." Table 3 illustrates a simplified chemical score for peanut flour, peanut-citrus blends, and citrus seed flour, determined by the method of McLaughlan *et al.* (16), that includes lysine, methionine, and cystine values only. Sawar *et al.* (20) determined the chemical score for legumecereal blends with this method and found that the values correlated well with those obtained by the Essential Amino Acid Index, Protein Efficiency Ratio, and Total Biological Value methods. Our calculations suggest that the high methionine citrus flour has a higher nutritional value than that of peanut flour alone, but peanut-citrus blends should have values between these two. They suggest that citrus seed flour, when blended with peanut flour, might be potentially useful to raise the nutritional values based on lysine, methionine and cystine contents.

Since one protein alone will rarely serve as the sole source of dietary amino acids, properly processed oilseed flours blended together could be used as protein sources. The relatively poorer solubility of citrus seed proteins compared to peanut proteins would preclude their use in clear or nonopaque beverages. However, defatted citrus meal has been used as a clouding agent for beverage bases (4), and Conkerton and Ory (8) showed that peanut proteins were soluble enough to supplement the protein concentration of pineapple juice, an acid beverage. It might be possible, therefore, to use peanut-citrus protein blends in fruit-flavored drinks or beverages such as pineapple juice or citrus drinks, to provide both protein fortification and the cloudiness of the natural products.

Because of the low solubility of citrus proteins at all pH's examined, the most likely application of citrus-peanut blends would be in bread or baked items. Peanut flour was used successfully in protein fortification of bread (13) and cake donuts (14). These blends should also be useful as meat extenders. Research on beef and protein-extended meats has shown that nutritive values of blends containing protein extenders from animal or plant sources can almost equal that of 100% lean beef (1).

The apparent higher methionine content of the citrus flour suggests a potential application for raising methionine levels of peanut flour. The current surplus of citrus seeds should also mean lower initial cost for the citrus seed flour. Its slight creamy color indicates that it could be blended with peanut flour and added to virtually all solid food products without creating a color problem. Raising the methionine level by blending with citrus seed flour should also have less effect on flavor of the final products than by supplementing with free methionine.

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Literature Cited

- 1. Anonymous. 1975. Research on nutrition of meat and extender blends shows PER of mixtures close to that of lean beef. Food Prod. Develop. 9: 72-74.
- Ayres, J.L., and B.L. Davenport. 1977. Peanut protein: A versatile food ingredient. J. Am. Oil Chem. Soc. 54: 109A-111A.

- 3. Berardi, L.C., and J.P. Cherry. 1977. Functional characteristics of co-precipitated protein isolates from mixtures of cottonseed, soybean, and peanut flours. Abs. 221, 37th Ann. Meeting, Inst. Food Technol., Philadelphia, Pa., June 5-8.
- 4. Braddock, R.J., and J.W. Kesterson. 1972. Amino acids of citrus seed meal. J. Am. Oil Chem. Soc. 49: 671-672.
- Cherry, J.P., J.M. Dechary and R.L. Ory. 1973. Gel electrophoretic analysis of peanut proteins and enzymes. I. Characterization of DEAE-cellulose separted fractions. J. Agr. Food Chem. 21: 652-655.
- Childers, A.B. 1972. Vegetable protein foods A review. J. Milk Food Tech. 35: 604-606.
- Conkerton, E.J. 1974. Gas chromatographic analysis of amino acids in oilseed meals. J. Agr. Food Chem. 22: 1046-1049.
- 8. Conkerton, E.J., and R.L. Ory. 1976. Peanut proteins as food supplements: A compositional study of selected Virginia and Spanish peanuts. J. Am. Oil Chem. Soc. 53: 754-756.
- 9. Daniel, V.A., D. Narayanaswamy, S. Kurien, M. Swaminathan, and H.A.B. Parpia. 1972. The protein efficiency ratio of protein enriched cereal foods based on blends of different cereals, oilseed meals, and legumes. Nutrition Reports Int. 5: 349-356.
- Dunlap, C.J., D.G. Guadogni, J.C. Miers, and J.R. Wagner. 1974. Methionine supplement alters flavor, PER of pinto beans canned in tomato sauce. Food Prod. Dev. 8, 88, 91-92.
- Finlayson, A.J., and S.L. MacKenzie. 1976. A rapid method for methionine determination in plant materials. Anal. Biochem. 70: 397-402.
- 12. Hudson, G.A., and J.L. Heinis. 1976. Microbiological and

chemical determination of tryptophan, methionine, and niacin in peanuts. Proceedings, APREA 8: 73.

- 13. Khan, M.N., and L.W. Rooney. 1975. Fortifying bread with oilseed protein. Tex. Agric. Prog. 21: 10-11.
- Lawhon, J.T., C.M. Cater, and K.F. Mattil. 1975. Sensory, analytical evaluation of cake doughnuts fortified with protein from oilseed flours. Food Prod. Dev. 9: 110-118.
- 15. Mattil, K.F. 1973. Considerations for choosing the right plant protein. Food Prod. Dev. 7: 40-44.
- McLaughlan, J.M., C.G. Rogers, D.G. Chapman, and J.A. Chapman. 1959. Evaluation of protein in foods. IV. A simplified chemical score. Can. J. Biochem. Physiol. 37: 1293-1299.
- 17. Ornstein, L. 1964. Disc electrophoresis. I. Background and theory. Ann. N.Y. Acad. Sci. 121: 321-349.
- Rechcigl, M., Jr., 1973. Man, food, and nutrition. Chemical Rubber Co. Press, Cleveland, Ohio, pp. 257-258.
- Sahni, S.K., K. Krishnamurthy, and G.K. Girish. 1975. Development of high protein bread. I. Groundnut flour and groundnut protein isolate utilization. J. Food Sci. Tech. 12: 283-289.
- Sawar, G., F.W. Sosulski and N.W. Holt. 1975. Protein nutritive value of legume-cereal blends. Can. Inst. Food Sci. Tech. J. 8: 170-175.
- 21. Wakefield, L.M., and R. Rowlands. 1973. Protein quality of rice polish and combinations with peanut flour, fish protein concentrate, and lysine. Cereal Chem. 50: 428-434.
- 22. Yayathi, T., and G.L. Brinkman. 1975. Complementary and supplementary value of peanut protein to rice when fed to weanling rats. Nutrition Reports Int. 12: 359-367.