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Protein Quality of Four Peanut Cultivars Grown at Two Locations

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ABSTRACT

Four cultivars of peanuts (Florigiant, Florunner, Starr and Tamnut) grown at two locations (Lewiston, North Carolina (NC) and Stephenville, Texas (TX)) in the National Regional Variety Trials were evaluated for protein nutritional quality. Peanuts, blanched with a minimum of heat treatment and partially defatted on a Carver press, were extracted with hexane at room temperature. Rats were fed these peanut meals to provide 10% of dietary protein (N x 6.25). PER was calculated as the ratio of weight gained to protein consumed for the 28-day feeding period. Adjusted PER values of the 8 peanut meals ranged from 1.3 to 1.7 compared to a value of 2.5 for the casein control diet. No statistically significant differences existed in PER among the 4 cultivars grown in NC. Florunner and Tamnut produced in TX had lower PER values than Florigiant and Starr grown at the same location. Florigiant, Florunner, and Tamnut grown in NC had lower PER values than the same varieties from TX. Rats were fed diets with Florunner peanut meal supplemented with methionine, lysine, and threonine, singly and in all combinations, for 7-day periods. Differences in protein quality between NC and TX-produced peanuts were overcome by addition of methionine to the diets. When all three amino acids were added, peanut meal from both locations supported growth of young rats equal to that of casein.

Key Words: Protein quality, PER, Limiting amino acids, Methionine, Lysine, Threonine.

Variations in composition of peanuts associated with genotype and with environmental conditions and cultural practices during growth have been of concern to those involved in production and utilization of the crop for some time.

The proteins of peanuts, like those of most plants, will not supply all of the essential amino acid requirements of man and some other mammals when fed at low or moderate dietary levels (6). Surveys of the amino acid profile of several varieties of peanuts led to the suggestion that gentic development of a peanut with improved protein quality might be possible (14, 15). Holaday and Pearson (4) reported differences in total protein content of peanuts associated with year and location of production as well as with variety.

Defatted meals made from seven cultivars of peanuts were incorporated into diets to provide 12, 16, and 20% of dietary protein (7). Statistically significant differences between varieties occurred in growth performance of rats fed the lower levels of protein, but these were not considered to be of practical importance. However, they did suggest that protein quality of the peanut might be improved by breeding. Mir and Hill (8) speculated that nutritional quality of peanuts grown in Ontario might not be comparable to that of peanuts produced in the less severe climate of the United States. However, a biological evaluation by protein efficiency ratio (PER) with rats showed no difference in protein quality of Comet type peanuts grown in Ontario and those produced in the United States (8).

Analysis of peanuts grown in the National Regional Peanut Variety Trials had indicated differences between varieties and between locations in amino acid composition of the samples (15). The major objective of the studies reported here was to determine if these differences in amino acid composition as determined by chromatography would result in significant differences in growth of rats fed diets containing meals from different genotypes and varieties of peanuts as the source of protein. When results of the first study showed that the PERs for three of the four varieties tested were greater for one location than for the other, Florunner cultivar was selected for further examination by dietary supplementation of amino acids.

Materials and Methods

Peanuts, from a common seed source for each variety, were grown under conventional cultural methods as part of the National Regional Peanut Variety Trials in the 1975 crop year. They were produced from a common seed source for each variety using conventional cultural methods at each location. After harvesting and drying by recommended procedures, they were shipped to the laboratory at Dawson, Georgia, where they were shelled, sized on a 0.635 cm slotted screen, and stored in plastic bags at 4 C until used for the protein quality studies in the summer of 1979.

The four varieties were selected for study because of commercial importance and differences in amino acid composition by ion-exchange chromatography. Stephenville, Texas (TX) and Lewiston, North Carolina (NC) were chosen as the locations for the first studies due to availability of sufficient material for the analyses.

Peanuts were subjected to mild heat treatment, to loosen the testa, in a 13.6 kg capacity oven (Preedit Electric Roaster, model 37) preheated to 157 C and loaded with peanuts at room temperature. The peanuts were heated until air in the rotating oven reached 105 C (about 15 min) and then cooled rapidly in a draft of ambient air. The skins were removed with a model EX Ashton Food Machinery blancher. Neucere et al. (10) have shown that dry heating peanuts at 155 C for 1 hour did not result in losses of methionine, cystine, lysine or threonine. Approximately 50% of the oil was pressed out of the blanched nuts by passing them three times through a Carver press. The final extraction was carried out by passing hexane through the ground pressed meal in a chromatography column (8 cm diameter) connected to a vacuum. One-kg batches of the meal, which filled the column to about 50 cm, were washed with approximately 2.5 l of hexane. The column remained connected to the vacuum until the last of the visible solvent was eluted from the meal (usually overnight). The meal was spread in a thin layer on a tray in a hood and stirred several times during the next 24 hrs to allow remaining hexane to evaporate. Peanuts, peanut meal, and mixed diets were kept at 5 C at all times when not in use.

The peanut meals were analyzed for moisture by drying for 2 hrs at 110 C in a forced draft oven, for nitrogen by Keldahl procedure, and for residual lipids by gravimetric determination of the material extracted by the Bligh and Dyer (1) methods. Samples of air dried peanut meal were hydrolyzed for amino acid analysis by a modification of the method of Roach and Gehrke (11). In screw capped tubes, 100 mg of peanut meal, or 25 mg of casein, in 20 ml of 6N HCl were flushed with nitrogen and

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heated at 145 C for 2 hr. The pH was adjusted to 2.1-2.2 with 12N NaOH and the sample diluted to 100 ml with citrate buffer at pH 2.2. Amino acids were quantified by ion-exchange chromatography as described by Spackman et al. (12) using a Durrum Model D-500 with a 1.75 mm x 48 cm column packed with Durrum high-resolution cation exchange resin (bead diameter of 8 ± 1 microns).

All diets contained, per kg, 22 g Vitamin Fortification Mixture (ICN Life Science Group), 35 g salt mixture UCB-1Rb (3), and sufficient soybean oil to make the total lipid content 80 g (including the residual peanut lipid). Peanut meal or casein (ANRC, Humko Sheffield) was added to give 16 g nitrogen and corn starch (Corn Products Co. No. 3401) to make 1000 g. Composition of the diet with meal made from Florigiant peanuts grown at Raleigh is shown as an example in Table 1.

Table 1. Composition of rat diet made with meal from Florigiant peanuts grown in North Carolina in 1975.

Ingredient	Gm/kg
Peanut meal (9.7% N)	164.9
Soybean oil	77.7
Vitamin mix	22.0
Salt mix	35.0
Corn Starch	700.4

Weanling male rats (Sprague-Dawley CD^R, Charles River Breeding Laboratories) were housed individually in stainless steel cages with wire mesh floors. Feed and deionizd water were provided ad libitum for 28 days to 10 rats in each treatment group. Feed intake was measured every second day and animals were weighted once weekly. PER was calculated as weight gained per gram of protein (nitrogen x 6.25) consumed. Values for diets containing peanut meal were adjusted to a fixed PER of 2.5 for the casein deit by multiplying the calculated PERs by the ratio of 2.5 to the calculated PER of the casein diet in each experiment.

For the second and third studies with Florunner peanuts from the 1975 and 1979 crops, respectively, diets were supplemented with amino acids at the expense of corn starch. Amino acids were supplied as the l-isomers to provide, in g/kg of diet: lysine, 3.2 (as lysine HCl, 4.0); threonine, 2.75; and methionine, 4.5. In these studies the diets were fed for 7 days only to 8 rats in each treatment.

Results and Discussion

Nitrogen, lipid, and moisture content of the peanut meals are given in Table 2. Amino acid composition of the meals and the rat's requirement for some dietary essential amino acids are shown in Table 3.

Calculated PER values for the casein diets were 3.34 in experiment 1, 3.61 in experiment 2, and 4.15 in experiment 3. Adjustment of a fixed value of 2.5 for casein PER has been recommended in order to facilitate comparisons among data from experiments conducted at various times and in different laboratories. Adjusted values of PER for the peanut meals fed in the first experiment are given in Table 4.

The adjusted PERs of 1.5 to 1.7 for meal made from defatted, blanched peanuts are similar to values reported previously from this laboratory (6, 7) and from other laboratories (5, 8). Florunner and Tamnut peanuts grown in TX gave somewhat lower adjusted PERs of 1.3 than usually reported for meals made from seeds that have had only mild heat treatment.

Table 2. Composition of meals made from peanuts grown in North Carolina (NC) and Texas (TX)

	Nitrogen, %		Lipid, %		Water, %	
	NC	TX	NC	TX	NC	TX
			1975			
Florigiant	9.7	9.3	1.4	1.2	7.8	10.0
Florunner	9.3	8.7	1.2	1.5	8.5	10.3
Starr	9.8	9.4	3.0	1.7	7.4	9.0
Tamnut	10.1	9.4	2.0	1.6	7.8	10.0
			1979			
Florunner	9.3	9.4	1.0	1.4	10.0	7.2

For three of the four varieties of peanuts included in this study, the ratio of weight gain to protein intake was significantly greater in rats fed meal made from seeds grown in NC than in those fed meal from peanuts produced in TX. No reports have been found in the literature that document a difference in performance of rats associated with the location in which their source of dietary protein was grown. Mir and Hill (8), for example, found no significant differences in weight gain, PER, or net protein ratio of rats fed meals made from Comet peanuts produced in the United States and in Ontario.

The data in Table 3 indicated that sulfur amino acids were the most limiting for rats in diets containing enough peanut meal to supply 1.6% nitrogen. About 25% of the rat's requirement (9) for sulfur amino acids was supplied by the meals while about 45% of its needs for lysine and threonine, the next most limiting amino acids, was provided. The amino acid data, however, did not appear to explain the differences in PER between varieties of peanuts and the location at which they were grown. Meal made from Florunner peanuts grown in TX supplied the least amount of sulfur amino acids and gave the lowest PER value, but meal from Starr peanuts produced in NC had almost the same concentration of sulfar amino acids and yielded one of the higher PER values.

Limitations of amino acids in the meals were investigated further by supplementing diets with methionine, lysine, and threonine singly and in all possible combinations (Table 5). Florunner was selected for more detail study because it was the variety having the greatest difference in PER values associated with location in which the peanuts were grown.

Most PER values obtained for meals made from Florunner peanuts grown in 1979 were lower than those obtained with the 1975 crop. The compositional data in Tables 2 and 3 offer no ready explanation for the difference in the rats' responses to peanut meals from the two crop years. Rats fed diets made with peanut meals from the 1979 crops, especially the diets without methionine supplementation, generally ate somewhat less food and gained even less weight per gram of food eaten than rats fed corresponding diets made with peanut meals from the

Table 3. Amino acid composition of meals made from peanuts grown in North Carolina (NC) and Texas (TX), g/16 g nitrogen (per kg diet).

		Florigiant		Florunner		Starr		Tamnut		Florunner	
		1975								1979	
		NC	TX	NC	TX	NC	TX	NC	TX	NC	TX
Alanine		4.5	4.7	4.7	5.0	4.6	4.9	4.6	4.5	3.5	3.9
Arginine	(6)[1]	12.2	10.7	11.9	10.5	11.2	12.5	11.3	11.2	9.3	10.5
Aspartic Acid	(4)	12.5	11.9	12.2	12.2	11.9	13.1	12.2	12.2	10.9	12.2
Glutamic Acid	(40)	23.3	21.8	22.6	21.9	22.0	24.5	22.7	22.6	20.1	22.1
Glycine		6.2	5.5	6.1	5.8	5.3	2.8	5.2	5.6	5.6	6.5
Histidine	(3)	2.3	2.8	2.3	2.3	2.3	2.5	2.3	2.5	2.3	2.3
Isoleucine	(5)	2.9	2.8	2.6	2.7	2.6	2.9	2.7	3.3	2.9	3.3
Leucine	(7.5)	6.9	6.4	6.9	6.8	6.9	7.1	6.6	6.4	6.1	6.7
Lysine	(7)	3.3	3.2	3.4	3.4	3.1	3.3	3.1	3.0	3.0	3.3
Methionine	(6)	0.7	0.6	0.7	0.6	0.5	0.8	0.6	0.7	0.7	0.6
Cystine		1.0	0.9	1.0	0.8	0.8	1.1	1.0	0.9	1.2	1.2
Phenylalanine	(8)	4.4	4.2	4.5	4.6	5.0	4.9	4.7	4.9	4.5	5.1
Tyrosine		3.9	3.6	3.9	3.9	3.8	4.3	3.8	4.1	3.8	4.1
Proline	(4)	3.5	3.5	3.4	3.5	3.2	3.5	3.3	4.1	4.1	4.4
Serine		4.5	4.4	4.4	4.5	4.4	4.8	4.4	4.7	3.9	4.3
Threonine	(5)	2.4	2.2	2.3	2.3	2.3	2.4	2.2	2.5	2.1	2.4
Valine	(6)	3.8	3.7	3.8	3.9	4.5	4.6	4.5	4.1	3.7	4.1
TOTAL		99.5	94.2	98.4	96.2	95.9	104.4	96.8	98.6	89.1	98.3

[1]Values in parentheses are dietary requirements for amino acids (g/kg diet) for the rat (9).

Table 4. Adjusted[1] protein efficiency ratio (28-day feeding period) of meals made from four varieties of peanuts grown at two locations in 1975[2].

	North Carolina	Texas	t[3]
Variety			
Florigiant	1.71 ± 0.09 a	1.51 ± 0.20 a	2.99
Florunner	1.66 ± 0.17 a	1.31 ± 0.15 b	4.89
Starr	1.60 ± 0.13 a	1.50 ± 0.18 a	1.47
Tamnut	1.57 ± 0.14 a	1.33 ± 0.20 b	3.13

[1]Adjusted to a fixed value of 2.5 for PER of casein.

[2]Values in a column, within an experiment followed by a common letter are not significantly different at $P \leq 0.05$.

[3]Values of t for significance of difference between locations.

1975 crop. Differences between crop years in the rat's responses were usually less for diets supplemented with methionine, and disappeared altogether for the diets supplemented with methionine, lysine, and threonine. One possible explation is that less methionine was biologically available to the rats from the peanuts produced in 1979 than from those grown in 1975. The two feeding trials comparing meals made from peanuts grown in the different years were conducted at different times and with rats purchased in two separate lots. Although these animals are bred for uniformity of response to nutritional factors, it is well known that identical results may not be obtained with different groups of rats. The calculated values of 3.61 and 4.15 for the actual PER of rats fed the standard casein diet in the two experiments not only point out such a difference in response but also account for some of the difference in adjusted values of PER for the peanut meal diets.

Differences between locations were signifiant for unsupplemented meals in 1975 and for those to which either lysine or threonine was added in both years. When methionine was added to the diets, alone or in combination with lysine and threonine, differences between locations were significant only in the case of methionine and lysine addition to meals made from the 1979 crop. Thus, the general superiority of protein quality of meals made from peanuts grown in NC over that of TX meals was overcome by supplementation of the diets with methionine.

The difference between analyzed values for sulfur amino acids (Table 3) for the meals made from Florunner peanuts grown at the two locations seems small to account for such significant differences in growth performance of the rats. The rat's dietary requirement forsulfur amino acids is estimated to be 6 mg per kg of diet (9). Diets made with meal from peanuts grown in TX and NC provided 22% and 28%, respectively, of the reputed requirement. It is possible that differences in biological availability of the sulfur amino acids in the two meals were responsible for the significant difference in growth response that was obtained in these studies.

Methionine, alone or in any combination with the other amino acid supplements, improved protein quality of peanuts grown in both years in TX. Peanuts grown in NC in 1975 were not improved by methionine alone or with either lysine or threonine. For the 1979 crop produced in NC, methionine or methionine and threonine supplementation of the diets resulted in increased PER values

Talbe 5. Adjusted[1] protein efficiency ratio (7-day feeding period) of meals from Florunner peanuts grown at two locations in two years and supplemented with amino acids[2].

Amino Acid	1975 Crop			1979 Crop		
Supplement	North Carolina	Texas	t[3]	North Carolina	Texas	t
none	1.30 ± 0.35 cde	0.84 ± 0.25 d	3.01	0.66 ± 0.42 d	0.58 ± 0.30 de	0.42
lysine (lys)	1.36 ± 0.33 cd	0.86 ± 0.39 d	2.81	0.86 ± 0.36 cd	0.34 ± 0.51 e	2.39
threonine (thr)	1.75 ± 0.12 b	0.47 ± 0.32 e	10.55	1.09 ± 0.47 cd	0.68 ± 0.29 d	2.17
lys + thr	1.00 ± 0.40 e	0.64 ± 0.38 de	1.80	0.61 ± 0.26 d	0.37 ± 0.20 e	2.04
methionine (met)	1.50 ± 0.37 bcd	1.62 ± 0.39 b	0.64	1.27 ± 0.54 bc	1.54 ± 0.33 b	1.21
met + lys	1.21 ± 0.42 de	1.26 ± 0.46 c	0.16	1.00 ± 0.75 cd	0.98 ± 0.24 c	0.10
met + thr	1.64 ± 0.24 bc	1.73 ± 0.07 b	0.97	1.66 ± 0.23 b	1.42 ± 0.21 b	2.20
met + lys + thr	2.69 ± 0.24 a	2.80 ± 0.19 a	1.00	2.78 ± 0.36 a	2.74 ± 0.08 a	0.33

[1]Adjusted to a fixed value of 2.5 for PER of casein.

[2]Values in a column, within an experiment followed by a common letter are not significantly different at $P \leq 0.05$.

[3]Value of t for difference between locations: for 14 d.f.; t.05 = 2.14; t.01 = 2.98.

over the unsupplemented meal.

Thus, methionine appeared to be the most limiting amino acid in peanuts grown in TX in both crop years and in peanuts produced in NC in 1979. In these crops, lysine and threonine were equally second limiting to the sulfur amino acids. The three amino acids were all equally limiting in the 1975 crop of peanuts grown in NC. For both locations and both crop years, the addition of all three amino acids, methionine, lysine,and threonine, gave PER values greatly superior to any diet with unsupplemented or partially supplemenmted peanut meal. Diets containing peanut meals supplemented with the three amino acids were equivalent to the diet containing casein in their capacity to support growth of weanling rats.

We found the three amino acids to be equally limiting in Florunner peanuts grown in Georgia in 1975 (6), and McOsker (5) also reported them to be equally limiting in raw peanut protein. Whether methionine is most limiting or equally limiting with lysine and threonine appears to vary with different lots of peanuts. Among the peanut meals that we have evaluated, those in which methionine was most limiting gave particularly low PER values.

None of the published data provide any evidence as to whether differences among lots of peanuts of the same variety are due to events that occur during growth of the seeds or to post-harvest factors. The sulfur amino acids of peanut proteins may be particularly susceptible to heat damage. Whatever the cause, various lots of peanut meal differ significantly in their capacity to supply growing rats with biologically available methionine.

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