Variations in Total Amino Acid Content of Peanut Meal^{1,2}

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ABSTRACT

The amino acid levels of oil-free peanut meal prepared from 16 varieties of peanuts that had 24-30% protein content of the kernels were examined. Nearly two-fold variations in the limiting essential amino acids (lysine, isoleucine, methionine, threonine, and valine) were found and are reported. Data for the remaining amino acids also are included. Utilization of these variations in amino acid composition should assist the development of peanut protein of improved quality. An analytical procedure to measure amino acid composition of peanut meal with a high precision on duplicate samples and which requires 15 hr for hydrolysis for best accuracy is described.

INTRODUCTION

There is a growing demand throughout the world for a balanced dietary source of protein. With the recent automation of ion-exchange chromatography using the Spackman et al. (1) technique, it has been possible to obtain rapid and accurate values for the amino acid composition of food products. In a summary article, Hoffpauir (2) in 1953 reported the amino acid composition of peanuts. Several other publications (3-5) have since reported conflicting values in the total amino acid composition of peanuts. Two

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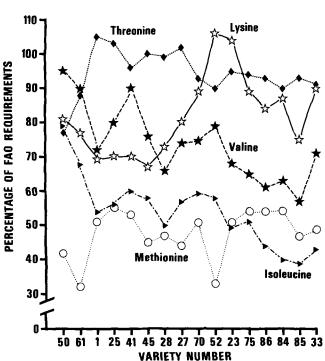


FIG. 1. Variation in lysine, isoleucine, methionine, threonine, and value content of 16 varieties of peanuts. Values are expressed as a percentage of the Food and Agriculture Organiation of the United Nations (FAO) requirement (18) for each of these essential amino acids shown to be deficient in peanut meal. \circ = Methionine, \blacktriangle = isoleucine, \bigstar = value, \blacklozenge = threonine, and \Leftrightarrow = lysine.

of these (5,6) reported on several varieties and generally concluded that large varietal differences did not exist, although it was stated (5) that small, but significant, differences of nitrogen, serine, glutamic acid, proline, alanine, leucine, phenylalanine, tyrosine, lysine, methionine, and cystine content were found. The differences in nitrogen content of oil-free meal from these nine varieties varied only from 10.69-10.81%. Results of Young and Holley (7) showed variations in the nitrogen content among peanut kernels of several varieties ranging from 4.72-5.45%.

In this study, results were obtained for the amino acid patterns in peanut flour of 16 varieties. Varieties with widely differing protein content were used. The effect of hydrolysis time on the amino acid values found for peanut protein was investigated, and an improved procedure is reported. The data were from varieties obtained using the improved analytical procedure, as described by Young (8), and which was somewhat similar to those used in studies on wheat (9).

EXPERIMENTAL PROCEDURES

Amino acid analyses were performed by the ion-exchange column chromatography technique of Spackman et al. (1), with a Beckman Model 120-C Amino Acid Analyzer using the P-28 resin for acidic and neutral amino acids and the P-35 resin for basic amino acids.

Peanut samples were selected from those grown in experimental plots at Tifton, Ga. These were hand shelled and selected for sound mature kernels, i.e. those peanuts having dark colored interior pericarp surfaces with thin faded testa. In most varieties, the sound mature kernels were smooth with little or no wrinkling of the testa surface. The peanut meal was prepared by Soxhlet extraction to remove the oil and by grinding in a Wiley laboratory mill. Nitrogen was determined by the Macro-Kjeldahl method (10) and reported on an oven-dry basis. Nitrogen was converted to protein by using the conversion factor of 5.46 (11).

Ca. 20 mg fat-free peanut meal (containing skins and hearts) was weighed accurately on a micro analytical balance into the hydrolyzate tubes. Two milliliters of 6N HCl was added, the sample cooled to -78 C, evacuated with

TABLE I

Nitrogen Content of Peanut Kernels and Peanut Meal

Variety	Variety or strain	Kernels	Peanut	
no.	OI SU ain	Refficio	mean	
		mgN/100 mg		
01	Dixie Spanish	4.80	8.95	
23	Tennessee Red	4.88	9.07	
25	Ga. 61-42	4.91	9.63	
27	Nambyguarae, PI 221068	4.56	7.74	
28	Va. Bunch 67	4.61	8.85	
33	Argentine	4.84	8.99	
41	Jenkins Jumbo	5.46	9.80	
45	Early Runner	4.59	9.25	
50	Conagin Macrocarpa	4.69	8.54	
52	Florida Jumbo	5.38	10.09	
61	McEachern Jumbo	5.50	9.54	
70	Bynum Runner	5.46	9.32	
75	NC-5	4.38	8.45	
84	Tarapota, PI 259747	4,40	8.71	
85	F 334A-B-14	4.34	8.67	
86	Ga.186-28	4.49	8.75	

TABLE II

Amino Acid Composition of Meal from 16 Varieties of Peanuts

Amino acid	Variety number											
		1	23	25	27	28	33	41	45	50		
	% of Total (by wt)											
Aspartic acid		15.43	11.67	13.15	15.03	13.08	12.35	13.28	13.76	8.83		
Threonine		2.73	2.46	2.69	2.66	2.57	2.37	2.50	2.60	2.01		
Serine		5.89	2.31	5.71	5.38	5.27	5.74	5.60	5.69	5.52		
Glutamic acid		20.16	19.26	21.59	19.15	21.60	19.59	20.60	21.18	22.39		
Proline		4.7 7	4.28	4.85	4.73	4.75	4.60	4.90	4.89	4.96		
Glycine		6.41	6.03	6.69	7.83	7.51	6.69	5.85	7.22	7.29		
Alanine		4.02	3.55	3.92	3.91	3.98	4.16	3.73	3.82	4.04		
Half cystine		2.45	2.62	3.31	2.64	2.77	2.44	2.41	2.42	2.22		
Valine		3.04	2.85	3.36	3.11	2.79	2.99	3.78	3.19	3.99		
Methionine		1.12	1.12	1.21	0.96	1.03	1.08	1.17	0.98	0.92		
Isoleucine		2.26	2.07	2.34	2.41	2.12	1.81	2.52	2.43	3.33		
Leucine		6.15	5.93	6.70	6.17	6.32	5.77	6.73	6.40	6.79		
Tyrosine		3.64	3.58	3.79	3.80	3.80	3.72	3.96	3.70	3.53		
Phenylalanine		4.91	4.64	5.06	4.91	5.19	4.89	5.19	5.00	5.16		
Lysine		2.88	4.38	2.96	3.35	3.07	3.79	2.94	2.81	3.41		
Histidine		1.97	2.94	2.05	2.28	2.06	2.59	2.13	2.15	2.22		
Ammonia		1.73	1.77	1.30	1.50	1.59	2.60	1.61	1.26	2.06		
Arginine		10.40	15.49	10.35	11.13	10.44	12.82	11.07	10.50	11.33		
Amino acid	52	61	70	/ariety nu 75	mber 84	85	86	R	ange	FAOa		
	% of Total (by wt)											
Aspartic acid	12.37	12.72	12.77	13.19	12.73	13.95	12.87	8.8	3-15.43	12.09		
Threonine	2.35	2.30	2.43	2.44	2.35		2.43		1- 2.73	2.77		
Serine	4.74	5.11	4.85	5.07	5.26		5.30		4- 6.04	5.08		
Glutamic acid	17.20	20.78	20.34	20.99			22.46		0-22.46	19.38		
Proline	4.79	5.57	5.20	5.45			6.36		8- 6.36	4.62		
Glycine	5,42	6.08	5.33	6.91	6.38		6.77		3- 7.83	5.92		
Alanine	3,54		3.54	3.62	3.56		3.41		1- 4.16	4.13		
Half cystine	2.12	2.29	2.73	1.97	2.90		2.41		7- 3.31	1.32		
Valine	3.32	3.76	3.14	2.74	2.90		2.55		9- 3.99	4.43		
Methionine	0.73		1.12	1.18	1.18		1.18		1- 1.21	1.22		
Isoleucine	2.44		2.47	2.15	1.10		1.10		4- 3.33	3.58		
Leucine	6.10		6.16	6.06			6.12		0- 6.79	6.79		
Tyrosine	3.50		3.60	3.39	3.36		3.57		6- 3.96	6.79 4.14		
Phenylalanine	4.61	5.06	4.82	5.50	5.54		4.62					
Lysine	4.45	3.23	3.73	3.30	5.54 3.66		4.62		6-5.54 8-4.45	5.28 3.75		
Histidine	3.17	2.45	2.68	2.49	2.51		2.20		4- 3.17	2.51		
Ammonia	1.98		1.88	2.49	2.51		2.20		6- 2.60	2.51		
Arginine	17.17	11.89	13.21	11.66	12.91	10.31	10.95		1-17.17	11.84		
				11.00	12.91		10.93	10.5		11.04		

^aCalculated from data tabulated by Food and Agriculture Organization of the United Nations (15).

a water aspirator, and sealed. The sealed samples were placed in a 110 C oven for 15 hr. Each hydrolyzed sample was transferred to a water moistened filter paper (Whatman No. 1) and filtered to remove humin. The filter paper was washed four times, the filtrate evaporated to dryness on a rotary evaporator, dissolved in 10 ml pH 2.2 citrate buffer, and stored at -20 C until analyzed. Each of the two columns of the analyzer required 0.5 ml buffered sample for determination of the amino acids. Tryptophan was not determined.

A series of standards from Spinco Division, Beckman Instruments, Inc., Palo Alto, Calif., was analyzed to obtain a measure of accuracy of the instrument and methodology (including variation due to application of sample to column). Standards were included for comparison each time samples were analyzed.

RESULTS AND DISCUSSION

Preliminary studies indicated 10-15 mg peanut meal to be insufficient material for analysis. The sample size was increased to 20 mg in studies of amino acid recoveries as a function of hydrolysis time and for determining the best procedure to analyze for varietal variation in amino acid composition. A 6.5 hr hydrolysis time was found to be sufficient for methionine only. Within 12-30 hr, only small differences in yield of individual amino acids were observed. In preliminary studies, hydrolysis periods longer than 30 hr decreased the amount of many of the amino acids, a finding in agreement with results reported on wheat (9). Thus, the decision was made to hydrolyze the peanut meal for 15 hr rather than the normally accepted practice of 24 hr. Amino acid standards in this study gave an average recovery of $100 \pm 1.62\%$ or better than the normal expected recovery of $100 \pm 3\%$ (12). In the hydrolyzate and variety studies, there was considerable variation in the ammonia analyses. Tkachuk and Irvine (9) pointed out that filter paper must be prewashed to remove ammonia, which may have accounted for the problem with reproducibility. At the time these samples were analyzed, the aspartic acid content was subject to considerable variation. Other than possible equipment or column malfunction, no explanation can be offered. Thus, aspartic acid and ammonia values are not included in the following discussion. The average variation between duplicate analyses for the first five varieties was $\pm 1.63\%$. This compares favorably with the $\pm 1.62\%$ value determined for the standards and is smaller than the $\pm 2.47\%$ variation observed in the hydrolyzate study.

Table I identifies the 16 varieties studied and shows the nitrogen content of the peanut kernels and of peanut meal. These varieties were selected because of their wide range of protein content, as determined in earlier studies (7,13,14). These 16 varieties also represent large genotypic variations

of the kernels, such as seed size, shape, and testa color (14). The amino acid compositions of these meals as percent-

ages of total, measured on a wt basis, are shown in Table II. The range of values for each amino acid is shown along with a calculated value (Food and Agriculture Organization of the United Nations [FAO]) using the data previously tabulated by the FAO (15). Of the seven essential amino acids determined in this study (tryptophan was not measured), only leucine and phenylalanine were adequate in all varieties examined. Histidine, which is essential for rats, was deficient in several varieties, especially Dixie Spanish (No. 1) and F 334A-B-14 (No. 85). This also might contribute to the low protein efficiency ratio (PER) values obtained when feeding rats. Valine usually is not reported as deficient but was low in all varieties analyzed. The other four essential amino acids (lysine, isoleucine, methionine, and threonine) usually are considered deficient (6,16,17). Using the FAO recommended levels (18) of each of these five amino acids, the percentages supplied by each variety were calculated and plotted in Figure 1 to illustrate the large variety differences encountered in this study.

The differences among varieties were large enough to be of important significance to a plant breeder attempting to increase any of these five essential amino acids.

The much lower levels of isoleucine in peanut meal (about one-half than previously reported) should be noted. This observation has occurred consistently in related studies, and an explanation still is lacking. Collaborative studies with another laboratory are being initiated in an effort to determine whether the variance observed is an analytical artifact or does indeed describe these peanuts. These data would suggest that isoleucine is the limiting amino acid in most varieties and is deficient in all varieties. Since one-third to one-half methionine requirement can be supplied by cystine, several varieties might have sufficient methionine. Assuming that cystine could supply one-half of the methionine requirement, Conagins Macrocarpa (No. 50) would appear to have the best balance of essential amino acids, although still deficient in these five, as shown in Figure 1.

This study, for the first time, has shown clearly that the variation in total amino acids was present in peanuts, thereby providing the opportunity for the genetic development of a superior protein quality. These results contrast those of Chopra and Sidhu (5) who found that the nine varieties they examined probably would not permit development of a variety of superior protein quality.

Aspartic acid, glutamic acid, and arginine account for ca. 45% total amino acids present. Thus, a significant reduction of these three would produce significant increases in other amino acids. Certainly the improvement of peanut protein quality by breeding will be a complicated and challenging endeavor.

Lastly, the 16 varieties reported herein are only a small portion of more than 3,000 accessions of cultivated varieties in the Plant Introduction Station (USDA) and of more than 20,000 different breeding lines of cultivated peanuts available in this country (19). Within these materials, it is possible that a peanut genotype with a superior protein quality already exists.

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