

## Variation in Total Amino Acid Percentage in Different Portions of Peanut Cotyledons<sup>1</sup>

Allan R. Hovis,<sup>2</sup> Clyde T. Young\*<sup>2</sup>, and Peter Y. P. Tai<sup>3</sup>

### ABSTRACT

Six peanut (*Arachis hypogaea* L.) varieties were analyzed for amino acid concentration among four consecutive sections across the two cotyledons. Significant differences were found among varieties (average 60% of total variation), among seeds (average 15% of total variation), and for some amino acids between sections (average 2.7% of total variation). With the exception of glutamic acid, proline, and histidine, varietal differences accounted for most of the variability found. Therefore, it appears that partial seed analysis for amino acids may be useful in genetic studies and for breeding selections.

Key Words: peanuts, *Arachis hypogaea* L., amino acids, groundnuts.

Due to low levels of several essential amino acids, the protein of the peanut (*Arachis hypogaea* L.) is considered

<sup>1</sup>Paper No. 6158 of the Journal Series of the North Carolina Agricultural Research Service, Raleigh, N.C.

<sup>2</sup>Department of Food Science, P. O. Box 5992, North Carolina State University, Raleigh, N. C. 27650.

<sup>3</sup>USDA, ARS, Sugarcane Field Station, Canal Point, FL.

to be of poor quality (2, 3). It is hoped that the amino acid distribution might be improved through plant breeding.

Young et al. (9) found varietal differences in the amino acid composition of sixteen peanut lines. These genetic differences for amino acid composition suggest that varieties with superior protein quality can be developed.

Various chemical analyses have been reported on portions of the peanut cotyledons by Tai and Young (7). Highly significant differences for the effects of variety, seed, section, and section x variety interaction were found in percentage protein in four cross sections along the longer axis of seeds from six peanut varieties. They concluded that analysis of half-cotyledons may be used for determining the protein percentage of peanut varieties on an individual seed basis.

The purpose of this study was to determine the usefulness of partial seed analysis for amino acids in peanut cotyledons from six peanut varieties. If the seed half opposite the embryonic end can be used for representative amino acid analysis, the remaining half can be germi-

nated. This would allow for more rapid selection of a breeding line containing the desired amino acid levels.

### Materials and Methods

Six peanut varieties, consisting of two spanish (*A. hypogaea* ssp. *fastigiata* var. *vulgaris*) types, Tifspan and Spancross, two valencia (*A. hypogaea* ssp. *fastigiata* var. *fastigiata*) types, Tennessee Red and White Manyema (PI 270773), and two virginia (*A. hypogaea* ssp. *hypogaea* var. *hypogaea*) types Florunner and F 334-A-B-14 were used. The peanuts were field grown at Tifton, Georgia in 1973, stack-cured, hand-shelled selecting only sound mature seeds by kernel and pod characteristics, and stored at -18 C until analyzed.

The sections of five sound mature seeds per variety were prepared as reported by Tai and Young (7). The sections were numbered consecutively from one to four with section four at the embryonic end of the seed.

The samples were defatted by placing each section in a 20 ml vial containing 15 ml of diethyl ether and crushing with a glass rod. The vial was allowed to stand until the solution was clear, and the ether was then decanted. The procedure was repeated twice and the residue allowed to dry.

For amino acid analysis, samples of the peanut sections were hydrolyzed by a modification of the method of Roach and Gehrke (4). Twenty-five mg of residue was weighed into a screw cap tube to which 20 ml of 6N HCl was added. After flushing with nitrogen gas, the tubes were heated at 140 C for 2 hr. The pH was then adjusted to 2.2 with 12N NaOH and the sample diluted to 50 ml in a volumetric flask with pH 2.2 citrate buffer. The amino acids were quantitated by ion exchange chromatography as described by Spackman et al. (6) using a Durrum Model D-500 with a 1.75 mm x 48 cm column packed with Durrum high-resolution cation exchanger (bead diameter, 8 ± 1 µm). Each sample was analyzed twice on the amino acid analyzer. The amino acids eluted in the following order: aspartic acid (ASP), threonine (THR), serine (SER), glutamic acid (GLU), proline (PRO), glycine (GLY), alanine (ALA), cystine (CYS), valine (VAL), methionine (MET), isoleucine (ILE), leucine (LEU), tyrosine (TYR), phenylalanine (PHE), histidine (HIS), lysine (LYS), ammonia (NH<sub>4</sub>), and arginine (ARG). Prior to statis-

tical analysis, the raw data were converted to percent of total amino acid residues to minimize effects due to differences in protein content. The data were analyzed by the analysis of variance (5) and Waller-Duncan procedures (8). The error term for testing differences among varieties was seed within varieties; for part and variety x part it was part by seed within variety. The residual mean square was used as the error term for the remaining effects.

### Results and Discussion

Table 1 shows that although a large amount of variation in amino acid content in peanuts is due to differences

Table 1. Mean squares with level of significance for components of analysis of variance for amino acid composition.

df =	V 5	S(V) 24	P 3	YXP 15	PXS(V) 72
ASP	1.568 **	0.109 **	0.189 **	0.197 **	0.029 **
THR	0.522 **	0.021 **	0.020 *	0.016 **	0.006 **
SER	2.724 **	0.080 **	0.017 NS	0.034 NS	0.034 **
GLU	1.270 **	0.264 **	0.296 NS	0.172 NS	0.124 **
PRO	2.138 **	0.496 *	0.513 NS	0.291 NS	0.245 NS
GLY	6.694 **	0.345 **	0.428 *	0.266 *	0.141 **
ALA	0.448 **	0.024 **	0.106 **	0.040 **	0.006 **
CYS	1.444 **	0.073 **	0.529 **	0.213 **	0.023 **
VAL	2.063 **	0.056 **	0.048 NS	0.017 NS	0.030 **
MET	1.333 **	0.052 **	0.236 **	0.082 **	0.015 **
ILE	7.321 **	0.230 **	0.024 NS	0.061 NS	0.049 NS
LEU	5.130 **	0.116 **	0.022 NS	0.052 NS	0.029 NS
TYR	2.118 **	0.072 **	0.109 NS	0.098 *	0.044 NS
PHE	3.499 **	0.058 **	0.055 NS	0.020 NS	0.024 NS
HIS	5.310 NS	2.486 **	0.021 NS	0.283 NS	0.236 **
LYS	1.367 **	0.092 **	0.052 NS	0.044 NS	0.027 **
NH <sub>4</sub>	15.162 **	0.298 **	0.168 NS	0.133 *	0.067 **
ARG	10.170 **	0.316 **	0.826 **	0.434 **	0.186 **
SUM	143780 **	5588 **	8128 **	1473 NS	1733 **

\*\* = Significant at the 0.01 level  
 \* = Significant at the 0.05 level  
 NS = Not significant  
 V = Variety  
 P = Part = section  
 S = Seed

Table 2. Variety and part means with the Waller-Duncan multiple range test for each amino acid (percent of total).

	Variety <sup>1</sup>						Seed Part			
	1	2	3	4	5	6	1	2	3	4
ASP	12.43 a	12.35 a	12.17 b	12.43 a	11.96 c	12.06 bc	12.30 a	12.26 ab	12.21 bc	12.17 c
THR	2.66 a	2.68 a	2.65 a	2.54 b	2.41 c	2.46 c	2.55 b	2.58 a	2.58 ab	2.55 b
SER	5.20 b	5.33 a	5.17 b	4.58 d	5.02 c	4.97 c	5.05 a	5.03 a	5.03 a	5.07 a
GLU	16.74 b	16.82 ab	16.60 bc	16.99 a	16.48 c	16.63 bc	16.81 a	16.71 a	16.68 a	16.64 a
PRO	6.07 ab	5.80 b	5.88 b	6.23 a	6.36 a	6.32 a	6.04 a	6.22 a	6.15 a	6.02 a
GLY	6.32 b	6.17 b	6.12 bc	5.88 c	6.61 a	5.41 c	6.04 b	6.07 ab	6.21 a	6.03 b
ALA	4.12 a	4.07 ab	4.03 b	4.03 b	3.87 c	3.86 c	4.00 b	4.04 a	4.02 ab	3.94 c
CYS	1.37 c	1.41 c	1.40 c	1.72 ab	1.80 a	1.67 b	1.54 b	1.50 b	1.51 b	1.70 a
VAL	4.18 bc	4.14 c	4.24 b	4.51 a	3.80 d	4.20 bc	4.15 a	4.22 a	4.17 a	4.18 a
MET	1.43 b	1.62 a	1.64 a	1.13 c	1.47 b	1.41 b	1.44 b	1.39 c	1.44 b	1.54 a
ILE	3.86 a	3.82 ab	3.89 a	2.76 d	3.53 c	3.65 bc	3.57 a	3.59 a	3.61 a	3.58 a
LEU	6.92 a	6.97 a	6.93 a	6.02 c	6.64 b	6.60 b	6.70 a	6.65 a	6.68 a	6.69 a
TYR	4.04 b	4.00 b	4.01 b	4.54 a	3.89 c	4.06 b	4.10 a	4.13 a	4.10 a	4.03 a
PHE	5.44 c	5.38 c	5.35 c	5.93 a	5.04 d	5.59 b	5.43 a	5.50 a	5.44 a	5.45 a
HIS	3.19 ab	3.00 ab	3.45 a	2.41 b	2.75 ab	3.11 ab	2.97 a	3.01 a	2.98 a	2.98 a
LYS	3.47 ab	3.49 ab	3.53 ab	3.46 b	3.60 a	3.08 c	3.43 a	3.43 a	3.48 a	3.41 a
NH <sub>4</sub>	0.59 b	0.74 b	0.82 b	1.79 a	1.81 a	1.89 a	1.26 a	1.22 a	1.28 a	1.34 a
ARG	11.98 c	12.21 b	12.08 bc	13.04 a	12.93 a	13.01 a	12.62 a	12.45 b	12.44 b	12.67 a
SUM	439.52 c	422.39 c	421.44 c	542.14 a	510.13 b	549.43 a	490.84 a	473.58 b	468.30 b	490.66 a

Values with the same letter within rows for variety or seed part are not significantly different at the 0.05 level.

<sup>1</sup>Variety identification: 1 = Tennessee Red; 2 = Tifspan; 3 = Spancross; 4 = F 334-A-B-14; 5 = Florunner; 6 = White Manyema.

among varieties, there is also much seed to seed variability within varieties. The significant difference between seeds within a variety suggests that partial seed analysis can be useful in improving the amino acid profile within a variety while recognizing that environmental as well as genetic effects may be involved.

Significant differences between seed sections were found for ASP, THR, ALA, CYS, MET, and ARG. The variation due to different seed sections contributed only as average of 2.7% to the variability between samples.

Based on previous results of Young et al. (9), varietal differences in amino acid concentrations were expected (Table 2). Florunner and F 334-A-B-14 (both virginia types) along with White Manyema (valencia type) showed higher levels of amino acids indicating a higher protein content than the other three varieties. When the percentages were totaled for the essential amino acids (1) (except tryptophan which was not measured), the two virginia types (Florunner and F 334-A-B-14) were 3% lower than the other four varieties.

Variety by part interactions were nonsignificant for half of the amino acids (Table 1) indicating that changes in these amino acid levels between seed parts were similar. Since the variety by part interaction was significant for the remaining amino acids, no general pattern for amino acid content could be applied to all varieties. Even though the part by seed within variety interaction was significant for all the amino acids except PRO, ILE, LEU, TYR, and PHE, its contribution to the variability between samples was minor.

The statistical model, on the average, accounted for 92% of the experimental variability resulting in a small

error term. This allowed very small differences to be seen as statistically significant. With the exception of GLU, PRO, and HIS, varietal differences accounted for the majority of the variability with the effect of seed part being very small.

Therefore, it appears that part seed amino acid analysis can be useful in genetic studies and breeding selections especially for initial screening to improve the amino acid balance of new releases.

### Literature Cited

1. FAO/WHO. 1973. Energy and protein requirements. FAO Nutr. Meeting Report Series #52. Rome.
2. Hegsted, D. M., R. Neff, and J. Worcester. 1968. Determination of the relative nutritive value of proteins. Factors affecting precision and validity. *J. Agr. Food Chem.* 16, 190.
3. Neucere, N. J., E. J. Conkerton, and A. N. Booth. 1972. Effect of heat on peanut proteins. II. Variations in nutritional quality of the meals. *J. Agr. Food Chem.* 20, 256.
4. Roach, D., and C. W. Gehrke. 1970. The hydrolysis of proteins. *J. Chromatogr.* 52, 393.
5. Snedecor, G. W. and W. G. Cochran. 1967. *Statistical Methods*, Sixth Ed. Ames, Iowa: The Iowa State University Press.
6. Spackman, D. H., W. H. Stein, and S. Moore. 1958. Automatic recording apparatus for use in chromatography of amino acids. *Anal. Chem.* 30, 1190.
7. Tai, Y. P., and C. T. Young. 1974. Variation in protein percentage in different portions of peanut cotyledons. *Crop Sci.* 14, 227.
8. Waller, R. A., and D. B. Duncan. 1972. A bayes rule for the symmetric multiple comparison problem. *Journal of the American Stat. Assoc.* 64, 1484. (Corrigenda 1972. Vol. 67, p. 253.)
9. Young, C. T., G. R. Waller, and R. O. Hammons. 1973. Variations in total amino acid content of peanut meal. *J. Amer. Oil Chem. Soc.* 50, 521.

Accepted April 1, 1982