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ESSENTIAL AMINO ACID COMPOSITION OF SOY BEAN MEALS PREPARED FROM TWENTY STRAINS OF SOY BEANS*

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The specific varieties of the most important food plants which are grown today were obtained through careful selection and breeding. Breeding experiments have demonstrated that the protein content of seed crops is subject to variation, and therefore suggest the possibility that the nutritional quality of seed proteins might be influenced by breeding. A change in distribution of the essential amino acids, as a result either of the production of different proteins or of variation in the relative proportions of the individual proteins in the seeds of different varieties, could definitely alter the nutritive value of the total seed protein.

Investigations with different strains of soy beans have demonstrated considerable variation in the total protein content of the seed. Cartter and Hopper (1) examined ten varieties and found protein values ranging from 36.6 to 53.2 per cent. Less extreme variations in the protein content within a given strain as a result of environmental factors have also been observed (1–6). Since soy bean culture is limited to a small group of select varieties which are grown principally in the corn belt states and are commonly pooled in the processing of the seed for oil and meal production (7), variations which result from environmental conditions probably have little effect on the protein content and composition of commercial soy bean meals. However, in view of the wide variation noted in protein content among different strains of soy beans one might expect significant variations to occur in amino acid composition of different strains. Csonka and Jones (8) reported variations in the cystine, tryptophan, and tyrosine content of soy bean seeds of six varieties in 1934 and suggested that "varieties should be selected for planting which produce more and better quality protein from the nutritional standpoint."

Doty *et al.* (9) have recently reported analyses of corn single crosses for tryptophan, tyrosine, cystine, arginine, and histidine and have concluded

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that the amount of these amino acids in the corn protein is related to the genetic constitution of the plant.

Microbiological methods of assay for the essential amino acids can now be conveniently applied to this problem. A study was therefore made of the essential amino acid content of soy bean meals prepared from twenty different strains of soy beans to determine whether there may be outstanding differences in composition of nutritional importance.

Materials and Methods

Preparation of Soy Bean Meals—The soy bean meals analyzed in this investigation were prepared by The Procter and Gamble Company from soy beans obtained through the cooperation of the United States Regional Soybean Laboratory at Urbana. Carefully composited samples were prepared so that location and soil differences would cancel as nearly as possible. The group of twenty strains includes the most promising varieties that are now being grown and others that are being considered for release.

The meals were all prepared by solvent extraction under comparable conditions and were supplied as raw and toasted flakes. Toasting was accomplished by heating the raw flakes in a steam-jacketed container at atmospheric pressure for 30 minutes. The temperature ranged from 21–88° during the first 12 minutes, 6 minutes later reached 104°, and was held between 104–110° for 12 additional minutes. The original moisture content of the toasted flakes was 20 per cent; the final moisture content of the toasted flakes was approximately 11 per cent.

Methods of Analysis—Microbiological methods of amino acid assay were employed. Valine, leucine, isoleucine, and glutamic acid were determined with *Lactobacillus arabinosus* 17-5 (10, 11). *Leuconostoc mesenteroides* P-60 was used to determine methionine (12), arginine, lysine, and phenylalanine.[1] *Streptococcus faecalis* R was used to determine threonine, histidine (13), and tryptophan (14).

Hydrolysates for tryptophan analyses were prepared with NaOH and cysteine as previously described (14). Acid hydrolysates used for the determination of the other amino acids were prepared by suspending 1 gm. of sample in 100 ml. of 6 N HCl and gently refluxing for 24 hours. After removing the excess acid by vacuum distillation, the hydrolysates were diluted to 100 ml., filtered, and refrigerated. Aliquots were neutralized for analysis.

RESULTS AND DISCUSSION

Microbiological methods of amino acid analysis are particularly useful to the nutritionist, because they can be applied directly to the analysis of

[1] Kuiken, K. A., and Lyman, C. M., unpublished methods.

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complex foodstuffs. Since it is not necessary to isolate specific protein fractions, it is possible to evaluate the total amino acid content of a foodstuff which contains a mixture of individual proteins in different proportions. Difficulty is experienced, however, in arriving at a satisfactory method of expressing analytical data for such products. If one cites the amino acid content of the foodstuff, he must also provide other data on gross composition to facilitate comparison of different samples. This can best be done by calculating all data to some common nitrogen basis. The practice of calculating to 16 per cent nitrogen has been widely used even though it is now recognized that the nitrogen content of most proteins is not 16 per cent. Glycinin, the principal protein of soy beans, contains 17 per cent nitrogen (15). Smiley and Smith (16) found from 16.26 to 16.90 per cent nitrogen in "soybean protein." Nevertheless, the exact nitrogen content of the total soy bean seed protein is not known and there seems little value in adopting a new factor for calculating the data. An alternative method of expressing the data is to calculate the ratio of amino acid nitrogen to total nitrogen. This avoids an empirical assumption and permits comparison of different samples. However, this method does not in itself provide a measure of the amino acid content of the sample and is not as useful to the nutritionist as data calculated on the crude protein basis. In view of these considerations, the authors have expressed all amino acid data in this paper in terms of 16 per cent nitrogen.

The soy bean meals were analyzed for the ten essential amino acids and glutamic acid. The content of these amino acids in raw soy bean meals prepared from twenty different strains of soy beans is given in Table I. No outstanding differences in individual amino acid content were observed. The greatest variation occurred in methionine content, of which the highest value was 19 per cent above the lowest. With most of the other amino acids, the variations were considerably smaller. The data suggest a great degree of uniformity in amino acid composition of the soy bean seed, rather than extensive variation as suggested by the work of Csonka and Jones (8).

Portions of a second lot of soy bean meals were toasted in order to inactivate the trypsin inhibitor shown to be present in raw soy beans by Bowman (17) and Ham and Sandstedt (18). The essential amino acid content of the toasted samples was compared with identical raw controls. As is shown in Table II, there was practically no change in amino acid content as a result of toasting, with the possible exception of lysine. The average data indicate that about 5 per cent of the lysine in the soy bean meals may have been destroyed by the heat treatment. The observations with lysine were, therefore, extended to include analyses of raw and toasted samples of soy bean meals representing twenty-one strains of soy beans. The additional data (Table III) indicate a similar small loss of lysine which

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TABLE I

Amino Acid Content of Raw, Solvent-Extracted Soy Bean Meals Prepared from Twenty Strains of Soy Beans

The values are expressed as the percentage of the amino acid in the crude protein (equivalent to calculating to 16 per cent N).

Soy bean strain	Crude protein (N × 6.25)	Arginine	Glutamic acid	Histidine	Isoleucine	Leucine	Lysine	Methionine	Phenyl-alanine	Threonine	Tryptophan	Valine
Acadian....	48.89	7.75	18.1	2.29	5.34	7.75	6.52	1.43	4.95	3.95	1.51	5.44
Arksoy.....	50.92	7.56	18.9	2.30	5.30	7.86	6.54	1.39	5.11	3.87	1.45	5.30
A3-176.....	48.84	7.68	18.6	2.25	5.34	7.90	6.70	1.43	5.02	3.93	1.56	5.43
A3K-884....	47.24	8.09	18.5	2.26	5.42	7.85	6.88	1.40	4.87	4.06	1.50	5.31
A4-107-12...	47.95	8.01	18.2	2.23	5.22	7.93	6.53	1.34	4.84	3.84	1.48	5.28
Chief.....	48.23	7.82	18.4	2.30	5.20	7.86	6.59	1.48	5.04	3.96	1.56	5.41
C. N. S....	50.84	7.87	18.4	2.36	5.19	7.59	6.00	1.31	5.11	3.72	1.55	5.35
C-463.....	47.24	7.54	18.0	2.37	5.39	7.96	6.90	1.45	5.07	3.96	1.61	5.40
Earlyana...	48.81	7.72	18.7	2.29	5.31	7.95	6.74	1.37	5.20	3.98	1.56	5.31
Gibson.....	48.19	7.49	18.6	2.30	5.35	8.13	6.91	1.41	5.08	3.82	1.54	5.31
H-5.....	49.42	7.22	17.9	2.16	5.24	7.97	6.51	1.38	5.04	3.58	1.54	5.22
Lincoln.....	48.86	7.53	18.4	2.29	5.32	8.08	6.67	1.40	5.22	3.91	1.64	5.42
Lincoln No. 3.....	46.38	7.72	19.2	2.33	5.43	8.45	6.73	1.53	5.17	4.03	1.60	5.48
Mamloxi....	49.49	7.96	18.3	2.40	5.53	7.94	7.07	1.50	5.23	3.86	1.44	5.34
N44-92.....	48.96	7.60	18.5	2.49	5.29	8.17	6.70	1.35	5.17	4.04	1.49	5.47
N44-774....	46.38	7.85	17.9	2.37	5.28	8.04	6.73	1.35	5.13	3.99	1.57	5.17
Ogden.....	47.81	7.49	18.5	2.34	5.48	8.16	6.76	1.28	5.31	3.83	1.42	5.31
Richland...	46.38	8.30	18.4	2.35	5.18	7.98	6.47	1.37	4.80	3.84	1.57	5.20
Roanoke....	47.40	7.64	19.0	2.47	5.15	8.02	6.48	1.41	5.23	3.88	1.48	5.23
S-100.....	49.98	7.56	18.6	2.52	5.32	7.98	6.54	1.42	5.12	3.76	1.46	5.32
Average*.		7.72 ±0.055	18.4 ±0.07	2.33 ±0.020	5.31 ±0.023	7.98 ±0.039	6.65 ±0.050	1.40 ±0.013	5.08 ±0.031	3.90 ±0.026	1.53 ±0.013	5.34 ±0.020

* ± standard error.

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TABLE II

Effect of Toasting on Amino Acid Content of Solvent-Extracted Soy Bean Meals

The values are expressed as the percentage of amino acid in crude protein.

Amino acid	A3K-884		C-463		Mamloxi		Memphis blank		Richland		Average	
	Raw	Toasted	Raw	Toasted	Raw	Toasted	Raw	Toasted	Raw	Toasted	Raw	Toasted
Arginine	7.92	7.81	7.56	7.63	7.70	7.86	8.03	8.17	8.10	8.09	7.86	7.91
Glutamic acid	18.2	18.3	18.2	18.2	18.5	18.6	18.8	18.4	18.2	18.1	18.4	18.3
Histidine	2.37	2.43	2.48	2.51	2.50	2.48	2.40	2.26	2.47	2.37	2.44	2.41
Isoleucine	5.42	5.38	5.39	5.42	5.48	5.34	5.28	5.21	5.18	5.23	5.35	5.32
Leucine	7.64	7.77	7.73	7.71	7.80	7.77	7.66	7.64	7.57	7.47	7.68	7.67
Lysine	6.50	6.23	6.90	6.38	7.07	6.49	6.56	6.40	6.35	6.32	6.68	6.36
Methionine	1.38	1.39	1.44	1.48	1.45	1.39	1.43	1.40	1.38	1.40	1.42	1.41
Phenylalanine	4.78	4.80	4.92	5.00	5.30	5.32	4.80	4.86	4.85	4.75	4.93	4.95
Threonine	4.06	4.07	3.96	3.89	3.95	4.17	3.95	3.80	3.81	3.95	3.95	3.98
Tryptophan	1.50	1.50	1.61	1.65	1.45	1.49	1.55	1.56	1.43	1.48	1.51	1.55
Valine	5.23	5.24	5.22	5.25	5.32	5.28	5.32	5.38	5.16	5.12	5.25	5.25

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would not be expected to affect significantly the nutritive value of the toasted material. However, since the difference in the mean values for the paired raw and toasted samples exceeded the standard error of the difference between the two means by a factor of 5, the result is highly significant statistically (19). The work of other authors indicates that the lysine in soy bean meals can be extensively destroyed by overheating. Riesen *et al.* (20) found that about 50 per cent of the lysine in a soy bean meal sample was destroyed by autoclaving for 4 hours at 15 pounds pressure. Evans and McGinnis (21) observed 30 per cent destruction of lysine in a meal that was autoclaved for 1 hour.

TABLE III

Effect of Toasting on Lysine Content of Solvent-Extracted Soy Bean Meals

The values are expressed as the percentage of lysine in crude protein.

Soy bean strain	Raw	Toasted	Soy bean strain	Raw	Toasted
Acadian	6.52	6.21	H-5	6.51	6.11
Arksoy	6.54	6.43	Lincoln	6.67	6.21
A3-176	6.70	6.55	Lincoln No. 3	6.73	6.06
A4-107-12	6.53	6.38	N44-92	6.70	6.18
Chief	6.59	6.89	N44-774	6.73	6.04
C. N. S.	6.00	6.07	Ogden	6.76	6.22
Earlyana	6.74	6.23	Roanoke	6.48	6.26
Gibson	6.91	6.42	S-100	6.54	6.21
Average*				6.60	6.28

* Composite averages ± standard errors for lysine in the total of twenty-one samples represented in Tables II and III are 6.62 ± 0.048 and 6.30 ± 0.043 for the raw and toasted meals respectively.

Recent values from the literature for the essential amino acid content of soy bean meal are given in Table IV. Only values obtained by microbiological methods are recorded. In general, there is very good agreement with the values obtained in this investigation. The most striking discrepancy is in the methionine value reported by Stokes (23). The application of Stokes' method in the authors' hands has in general given low values for methionine. Lysine values reported by Stokes (23), Baumgarten (22), and Schweigert (24) are lower than those by Riesen (20) and the authors. Perhaps this is an indication of slight overheating in the samples, which gave low values. The authors' data and those of Riesen refer to raw preparations. It is conceivable that lysine assays might be used to detect overheating of soy bean meal. The high values for tryptophan reported by the present authors were obtained by an improved method of hydrolysis

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(Kuiken *et al.* (14)). It is evident that soy bean meals sampled independently tend to have identical amino acid composition.

The relatively small variations in the content of the individual amino acids in the protein of the different varieties of soy beans is paralleled by the authors' (26) findings with respect to cottonseed meal. Eight samples of cottonseed meal collected from different parts of the country were found to have practically the same amino acid composition when the data were calculated to 16 per cent nitrogen.

TABLE IV

Comparison of Present Data with Recent Literature Values for Essential Amino Acid Content of Soy Bean Meal

All data were obtained by microbiological methods and are expressed here as the percentage of amino acid in the crude protein.

Amino acid	Present paper	Riesen (20)	Baumgarten (22)	Stokes (23)	Schweigert (24, 25)
Arginine	7.7	7.4	5.3	7.1	7.3
Histidine	2.3	2.8	3.0	2.3	2.2
Isoleucine	5.3	5.1	6.2	4.5	4.8
Leucine	7.9	7.6	7.9	7.4	7.1
Lysine	6.6	6.6	5.3	5.4	5.7
Methionine	1.4	1.4	1.6	0.8	
Phenylalanine	5.1	5.1	4.9	5.3	4.5
Threonine	3.9	3.8	3.7	3.9	3.3
Tryptophan	1.5	1.2	1.1	1.2	
Valine	5.3	5.4	5.3	4.6	5.2

SUMMARY

Soy bean meals prepared from twenty strains of soy beans were analyzed for the essential amino acids and glutamic acid by microbiological methods. The amino acid distribution in the crude protein of the different samples was in general quite uniform. The greatest variation occurred in the case of methionine, in which the highest value was 19 per cent above the lowest.

Toasting the meals as practiced by the industry in order to inactivate the proteolytic inhibitor of raw soy beans did not result in the loss of any amino acids except lysine. The loss of lysine indicated by the data was too small to be of nutritional significance.

BIBLIOGRAPHY

1. Cartter, J. L., and Hopper, T. H., *U. S. Dept. Agr., Tech. Bull. 787* (1942).
2. O'Kelly, J. F., and Gieger, M., *Mississippi Agr. Exp. Sta., Tech. Bull. 24* (1937).
3. Lipman, J. G., and Blair, A. W., *Soil Sc.*, **1**, 171 (1916).
4. Fellers, C. R., *Soil Sc.*, **6**, 81 (1918).

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5. Stark, R. W., *J. Am. Soc. Agron.*, **16,** 636 (1924).
6. Smith, C. D., and Robison, F. W., *Michigan Agr. Exp. Sta., Bull. 224* (1905).
7. Hayward, J. W., The composition and nutritive properties of soybeans and soybean oil meal, Soybean Nutritional Research Council, Chicago, 13 (1938).
8. Csonka, F. A., and Jones, D. B., *J. Agr. Res.*, **49,** 279 (1934).
9. Doty, D. M., Bergdoll, M. S., Nash, H. A., and Brunson, A. M., *Cereal Chem.*, **23,** 199 (1946).
10. Kuiken, K. A., Norman, W. H., Lyman, C. M., Hale, F., and Blotter, L., *J. Biol. Chem.*, **151,** 615 (1943).
11. Lyman, C. M., Kuiken, K. A., Blotter, L., and Hale, F., *J. Biol. Chem.*, **157,** 395 (1945).
12. Lyman, C. M., Moseley, O., Butler, B., Wood, S., and Hale, F., *J. Biol. Chem.*, **166,** 161 (1946).
13. Lyman, C. M., Kuiken, K. A., and Hale, F., *J. Biol. Chem.*, **171,** 233 (1947).
14. Kuiken, K. A., Lyman, C. M., and Hale, F., *J. Biol. Chem.*, **171,** 551 (1947).
15. Pucher, G. W., and Nolan, L. S., unpublished data cited by Vickery, H. B., *Physiol. Rev.*, **25,** 347 (1945).
16. Smiley, W. G., and Smith, A. K., *Cereal Chem.*, **23,** 288 (1946).
17. Bowman, D. E., *Proc. Soc. Exp. Biol. and Med.*, **57,** 139 (1944).
18. Ham, W. E., and Sandstedt, R. M., *J. Biol. Chem.*, **154,** 505 (1944).
19. Pearson, F. A., and Bennett, K. R., Statistical methods applied to agricultural economics, New York, 311 (1942).
20. Riesen, W. H., Clandinin, D. R., Elvehjem, C. A., and Cravens, W. W., *J. Biol. Chem.*, **167,** 143 (1947).
21. Evans, R. J., and McGinnis, J., unpublished data cited by Stevens, J. M., and McGinnis, J., *J. Biol. Chem.*, **171,** 431 (1947).
22. Baumgarten, W., Mather, A. N., and Stone, L., *Cereal Chem.*, **23,** 135 (1946).
23. Stokes, J. L., Gunness, M., Dwyer, I. M., and Caswell, M. C., *J. Biol. Chem.*, **160,** 35 (1945).
24. Schweigert, B. S., *Poultry Sc.*, **27,** 223 (1948).
25. Schweigert, B. S., *J. Nutr.*, **33,** 553 (1947).
26. Lyman, C. M., Kuiken, K. A., and Hale, F., *Texas Agr. Exp. Sta., Bull. 692* (1947).

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