If the proposed mechanism is correct, it seems likely that it generally governs at least in part the textural properties of fully processed potato products. The factors that determine the texture of processed potato tubers are, according to this view, the thermal lability of the cytoplasmic membrane, the ion concentration of the cell sap (predominantly K), the amount of PME present in the tissues, and the amount of available alkaline earths.

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Received for review June 9, 1971. Accepted September 3, 1971. Purdue University Agricultural Experiment Station Journal Article No. 4415.

Amino Acid Composition of Buckwheat

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Crude protein and 17 amino acids were determined in ten samples of genetically diverse buckwheats, in buckwheat fractions from a commercial mill, and in the germ and degermed groats. The buckwheat proteins were particularly rich in lysine (6.1%), and contained less glutamic acid and proline and more arginine and aspartic acid than cereal proteins. About 56% of glutamic and aspartic acids were in the form of amides. Whereas correlations among basic or neutral and acidic amino acids were positive, correlations between basic and acidic or neutral amino acids were negative. Dark flour and feed fractions contained more protein than the whole kernel or the groat, but the amino acid patterns differed little. Distribution of amino acids in buckwheat tissues differed significantly from distribution in tissues of cereal grains. The pattern of essential amino acids in buckwheat is compared to that of cereal grains and egg reference patterns. Chemical analyses of the buckwheat hydrolyzates indicated that the amino acid composition was nutritionally superior to that of cereal grains.

Buckwheat (*Fagopyrum esculentum* Möench) is not a true cereal. It belongs to the *Polygonaceae* (or buckwheat) family, but like the cereals, the grain of buckwheat is a dry fruit (Winton and Winton, 1945; Marshall, 1969). The black hulls of the triangular fruit are not suited for human food. Structurally, they have little in common with bran coats of the cereals. The seed proper (groat) is similar to that of cereals in that it consists of starchy endosperm and oily embryo.

Feeding experiments of Sure (1955) have shown that the proteins in buckwheat are the best known source of high biological value proteins in the plant kingdom, having 92.3% of the value of nonfat milk solids and 81.4% of whole egg solids. The proteins of buckwheat were shown to have excellent supplementary value to the cereal grains (Sure, 1955; Wyld *et al.*, 1958).

Sokolov and Semikhov (1968) fractionated proteins of diploid and tetraploid forms of buckwheat; globulins were a main component of both. However, seeds of the tetraploid form contained more globulins and less albumins. Polyacrylamide gel electrophoresis showed no qualitative differences in composition of albumin, globulin, and glutelin fractions of diploid and tetraploid seeds. Similarly, Jacko and Pleskov (1968) found little difference in albumins and globulins, separated by chromatography on tetraethylaminoethylcellulose and by electrophoresis in polyacrylamide gel, in diploid and tetraploid buckwheat.

Amino acid composition of whole buckwheat was determined by several investigators, including Lyman *et al.* (1956), who used a microbiological assay method, and more recently by Tkachuk and Irvine (1969), who used an ion-exchange procedure. Zebrok *et al.* (1966) found no significant difference in the amino acid composition of the total protein between diploid and tetraploid buckwheat.

Gross composition of milled buckwheat products was reported by Coe (1931) and by Watt and Merrill (1963).

The present work is concerned with the amino acid composition of buckwheat grown in the United States, of commercially milled buckwheat fractions, and of fractions from seeds dissected by hand.

MATERIALS AND METHODS

Ten buckwheat seed samples of various origins were obtained from H. G. Marshall, Research Agronomist of the United States Department of Agriculture, at Pennsylvania State University, University Park. Those samples are described in Table I.

The samples were from seed lots grown in different years at

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University Park. A sample of buckwheat seed and samples of its commercially milled products were obtained from J. W. Bailey (The Birkett Mills, Penn Yan, N.Y.). The groats were separated into germ and degermed fractions by dissection or by manual separation after pearling on a Strong-Scott barley pearler. For analyses, all samples were ground in a micro-Wiley mill to pass through a 20-mesh sieve. Nitrogen was determined by the Kjeldahl method on 1.0 g of kernels and commercially milled samples, and on 0.1 g of the hand-separated fractions. Crude protein was estimated by multiplying the nitrogen concentration by 6.25, and is reported on a moisture-free basis.

Amino acid analyses of hydrolyzates were performed with a Beckman 121 automatic amino acid analyzer by an assay method described elsewhere (Robbins *et al.*, 1971). Values are expressed in grams of amino acid per 100 g of amino acid recovered. Recoveries were at least 80% and averaged 85%. Most results are rounded off to the first decimal point, though in actual statistical computations at least three figures after the decimal point were used. In addition to evaluating the data on the individual amino acids, the sum of lysine, threonine, and methionine (SLTM), which are the main limiting amino acids in cereals, was used for statistical computations and comparisons among cultivars and milled fractions of buckwheat.

RESULTS AND DISCUSSION

Means, ranges, standard deviations, and coefficients of variations of crude protein and amino acid compositions of the ten samples described in Table I are given in Table II. None of the tested samples differed by three standard deviations (or more) from the mean.

There were no consistent differences in the protein content or in the amino acid composition of the diploid and tetraploid cultivars. This is in agreement with the results of Zebrok *et al.* (1966). The main outstanding characteristic of the buckwheat proteins is the high lysine content (average of 6.1%) which is higher than in any of the cereal grains. Oat proteins are richest among the cereals in lysine, with an average of 4.2% of this amino acid (Robbins *et al.*, 1971). Compared to cereal grains, buckwheat proteins contain relatively low concentrations of glutamic acid and proline, but are rich in arginine and aspartic acid. Assuming that the source of ammonia is mainly from glutamic and aspartic acids, about 56% of those amino acids was in form of amides; the corresponding values were 85% in wheat and 69% in oat proteins (Waggle *et al.*, 1966; Robbins *et al.*, 1971).

Correlations among amino acids and between amino acids and protein contents of the whole buckwheat are summarized in Table III. Correlation coefficients of at least 0.632 and 0.765 were significant at the 5 and 1% levels, respectively. None of the amino acids was significantly correlated with protein content. This indicates that increasing protein content probably does not impair the excellent amino acid pattern of buckwheat proteins, and that plant breeding and cultural practices to increase protein content might not decrease the biological value of buckwheat. Such decreases were observed in wheat (Johnson *et al.*, 1970) and barley (Mosse and Baudet, 1969).

The concentration of lysine increased as the concentrations of histidine and arginine increased, and as concentration of glutamic acid decreased. Histidine was positively correlated with arginine and negatively with glutamic acid, isoleucine, and leucine. Negative correlations were found between arginine and aspartic acid, threonine, glutamic acid, glycine, and alanine. Concentration of aspartic acid increased as alanine increased. Threonine was correlated positively with serine, glycine, and alanine; glutamic acid was correlated with glycine, alanine, and phenylalanine; glycine was correlated with alanine and phenylalanine; and isoleucine was correlated with leucine and valine.

Correlations among basic amino acids or among neutral and acidic amino acids were positive; correlations between basic amino acids and acidic or neutral amino acids were negative. The results point to the presence of proteins that are consistently rich in amino acids of a specific type (basic, neutral, or acidic).

The protein content and amino acid composition of whole buckwheat and milled buckwheat products are shown in Table IV. As in debranning of cereal grains (Pomeranz and MacMasters, 1970), dehulling of buckwheat increased the protein content. Similarly, the dark flour and buckwheat feed contained more protein, the light flour less, and hulls

Table I. Buckwheat Samples Used in the Survey

Identifying no.	Name	Origin	Description
C.I. 16	Pennquad	Pennsylvania (USA)	Tetraploid, Japanese type
C.I. 10	Tokyo	Canada	Diploid, Japanese type
Pa C156	Kasho	Japan	Diploid, Japanese type
Pa 66G99w		Pa. Exp. Sta. (USA)	Tetraploid, silverhull, experimental
C.I. 2		Wisconsin (USA)	Diploid, silverhull, local strain
Pa C29	LaHarpe	France	Diploid, silverhull
Pa C146		USSR	Tetraploid, Japanese type
C.I. 23		USSR	Tetraploid, Japanese type
Pa C154		New York (USA)	Diploid, Japanese type, local strain
Pa C155		Minnesota (USA)	Diploid, Japanese type, local strain

Table II. Mean, Maximum Value (max), Minimum Value (min), Standard Deviation(s), and Coefficient of Variability (CV) for Crude Protein and Amino Acid Composition of Ten Buckwheat Samples

Parameter	Max	Min	Mean	s	CV
Percent protein (PcP)	15.4	12.6	13.7	0.851	6.2
Lysine (Lys)	7.0	5.0	6.1	0.460	7.5
Histidine (His)	3.1	2.3	2.7	0.196	7.1
Ammonia (NH_3)	2.3	1.7	2.1	0.164	7.8
Arginine (Arg)	11.6	8.5	9.7	0.837	8.6
Aspartic acid (Asp)	12.1	10.8	11.3	0.331	2.9
Threonine (Thr)	4.1	3.6	3.9	0.138	3.5
Serine (Ser)	5.2	4.5	4.7	0.198	4.2
Glutamic acid (Glu)	19.4	17.8	18.6	0.404	2.2
Proline (Pro)	4.3	3.2	3.9	0.353	8.9
Half cystine (Cys)	1.8	1.2	1.6	0.162	10.1
Glycine (Gly)	6.5	5.9	6.3	0.186	2.9
Alanine (Ala)	4.7	4.2	4.5	0.133	2.9
Valine (Val)	5.4	4.8	5.1	0.166	3.2
Methionine (Met)	3.0	1.8	2.5	0.344	13.5
Isoleucine (Ile)	4.0	3.6	3.8	0.098	2.5
Leucine (Leu)	6.6	6.1	6.4	0.156	2.4
Tyrosine (Tyr)	2.5	1.8	2.1	0.235	11.1
Phenylalanine (Phe)	5.0	4.6	4.8	0.127	2.6
Sum of Lys + Thr + Met (SLTM)	13.0	11.7	12.5	0.389	3.1

Table III. Correlations Among Amino Acids and Between

	PcP	Lys	His	NH_3	Arg	Asp	Thr	Ser	Glu	Pro
PcP	**1.000**									
Lys	0.005	**1.000**								
His	−0.072	**0.974**[a]	**1.000**							
NH_3	0.220	**0.848**	**0.815**	**1.000**						
Arg	0.117	**0.813**	**0.852**	*0.730*	**1.000**					
Asp	−0.143	−0.452	−0.526	−0.424	**−0.769**	**1.000**				
Thr	−0.007	−0.548	−0.525	−0.370	**−0.797**	*0.651*	**1.000**			
Ser	0.238	−0.321	−0.262	−0.101	−0.447	0.420	**0.877**	**1.000**		
Glu	0.145	**−0.897**	**−0.904**	**−0.769**	**−0.866**	*0.642*	*0.649*	0.425	**1.000**	
Pro	−0.097	0.095	0.143	−0.217	0.083	−0.141	−0.111	0.057	−0.188	**1.000**
Cys	−0.462	−0.461	−0.446	−0.627	−0.257	0.158	−0.211	−0.550	0.381	−0.250
Gly	−0.026	*−0.703*	*−0.723*	−0.610	**−0.899**	*0.651*	**0.778**	0.473	**0.870**	−0.288
Ala	−0.143	*−0.670*	*−0.706*	−0.538	**−0.923**	**0.840**	**0.851**	0.537	**0.798**	−0.353
Val	0.364	−0.536	*−0.653*	−0.127	−0.620	0.415	0.494	0.311	0.575	−0.506
Met	−0.150	−0.251	−0.252	−0.297	0.102	−0.514	−0.495	*−0.676*	−0.042	−0.065
Ile	0.255	*−0.635*	**−0.775**	−0.368	*−0.681*	0.445	0.347	0.057	0.628	−0.422
Leu	−0.042	*−0.696*	**−0.823**	−0.622	*−0.748*	0.610	0.275	−0.104	*0.667*	−0.266
Tyr	−0.358	−0.505	−0.367	−0.463	−0.202	−0.149	0.234	0.197	0.159	0.214
Phe	0.079	*−0.734*	*−0.714*	−0.554	*−0.667*	0.476	0.531	0.330	**0.846**	−0.454
SLTM	−0.127	*0.760*	*0.736*	0.598	*0.764*	*−0.759*	*−0.733*	*−0.669*	**−0.862**	0.023

Degrees of freedom for above correlations (N − 2) = 8.

[a] All above figures above 0.632 are in italics and all figures above 0.765 are in bold face to indicate statistical significance.

the least protein among the milling fractions. Similar results were reported by Coe (1931).

The amino acid pattern of milled buckwheat fractions differs from the pattern of milled cereal grains. In wheat, proteins in white flour contain substantially less lysine and in dark flour slightly less than in the whole kernel. Commercial bran and germ proteins contain, respectively, about 1.4 and 1.9 times more lysine than the whole wheat proteins. Milling wheat shifts the concentration of the main amino acid of storage proteins, glutamic acid. It increases from about 28.5% in the whole kernel to 34.5% and 29.6% in white and dark flours, respectively, and decreases to 16.2% in bran and to 14.0% in germ. Similar changes to those in glutamic acid occur in proline of wheat products (Bradley, 1967).

Milled fractions of buckwheat differed little in amino acid composition (Table IV). Only the hulls, which are unsuited for human consumption, differed in amino acid composition from that in the edible fractions. There were only minor differences in concentrations of lysine, glutamic acid, and proline in the whole groat, the dark flour, the light flour, and the feed fraction. The light flour would be expected to contain primarily particles from the starchy endosperm; the dark flour is rich in germ, and the feed fraction is a mixture that includes, in addition to the above, some screenings, broken kernels, and limited amounts of the hull. Separations in commercial milling are rather arbitrary, they vary among mills, and are based primarily on differences in physical properties of various tissues in the kernel. Despite imperfections of the milling process, the data in Table IV suggest that, contrasted with wheat, there are only small differences in amino acid composition of proteins among the tissues of the dehulled buckwheat groat.

The latter assumption is confirmed, in part, by the results

Table IV. Protein Content and Amino Acid Composition of Commercially Milled Buckwheat

Parameter	Whole buckwheat	Buckwheat groats	Light flour	Dark flour	Buckwheat feed	Hulls
PcP	13.8	16.4	7.4	19.0	18.9	4.0
Lys	6.0	5.9	5.7	5.2	5.2	6.3
His	2.6	2.6	2.7	2.4	2.8	3.9
NH_3	2.1	1.9	2.2	1.7	2.3	3.0
Arg	9.2	10.0	7.9	8.7	9.7	5.4
Asp	11.4	11.4	10.5	11.8	10.8	10.7
Thr	4.0	3.8	4.1	4.1	3.9	5.4
Ser	4.9	4.6	4.7	5.5	5.0	5.7
Glu	18.5	19.3	17.6	20.1	19.2	13.3
Pro	3.8	3.8	5.1	4.1	4.0	4.2
Cys	1.6	1.8	1.8	1.5	1.5	0.5
Gly	6.6	6.2	6.2	6.6	6.2	7.5
Ala	4.3	4.4	4.5	4.1	4.2	5.0
Val	5.3	4.9	5.4	5.1	5.1	6.6
Met	2.3	2.8	2.8	1.9	2.3	2.9
Ile	4.0	3.7	4.2	3.9	4.0	5.1
Leu	6.7	6.2	7.0	6.5	6.7	8.3
Tyr	2.0	2.1	2.9	2.1	2.3	1.8
Phe	4.8	4.8	4.8	4.9	4.7	4.3
SLTM	12.4	12.4	12.5	11.2	11.5	14.6

Table V. Protein Content and Amino Acid Composition of Hand-Separated Buckwheat Fractions

Parameter	Germ	Degermed buckwheat	Germ from pearling	Endosperm from pearling
PcP	55.9	10.1	50.3	3.3
Lys	5.6	6.1	5.6	6.0
His	2.6	2.3	2.6	2.1
NH_3	1.9	2.0	2.1	1.8
Arg	11.9	9.5	12.5	5.6
Asp	10.9	10.9	11.0	10.6
Thr	3.7	3.9	3.5	4.5
Ser	5.0	4.3	4.9	4.2
Glu	19.3	17.2	19.8	13.4
Pro	3.8	4.9	4.0	4.3
Cys	2.2	1.8	1.8	1.9
Gly	6.0	6.1	5.6	6.9
Ala	3.6	4.9	3.7	6.4
Val	4.8	5.1	4.6	6.5
Met	2.0	3.1	1.8	3.7
Ile	3.5	3.9	3.4	5.0
Leu	5.8	6.6	5.5	9.0
Tyr	2.8	2.9	2.8	3.5
Phe	4.7	4.7	4.8	4.6
SLTM	11.3	13.1	11.0	14.3

Amino Acids and Protein Content of Ten Buckwheat Samples

	Cys	Gly	Ala	Val	Met	Ile	Leu	Tyr	Phe	SLTM
PcP										
Lys										
His										
NH_3										
Arg										
Asp										
Thr										
Ser										
Glu										
Pro										
Cys	**1.000**									
Gly	0.270	**1.000**								
Ala	0.221	**0.913**	**1.000**							
Val	−0.056	0.583	0.613	**1.000**						
Met	0.589	−0.142	−0.282	0.032	**1.000**					
Ile	0.236	0.593	0.610	**0.919**	0.272	**1.000**				
Leu	0.537	0.581	*0.651*	0.672	0.316	**0.887**	**1.000**			
Tyr	0.203	0.001	0.062	−0.081	0.375	−0.051	0.045	**1.000**		
Phe	0.424	**0.831**	*0.733*	0.509	−0.008	0.524	0.493	−0.009	**1.000**	
SLTM	−0.095	*−0.678*	*−0.742*	−0.432	0.414	−0.385	−0.442	−0.183	*−0.686*	**1.000**

in Table V. Although the germ contained over 50% protein (on a moisture-free basis) and the central endosperm only 3.3% protein, they differed little in lysine concentration. Proteins of the wheat endosperm are rich and the germ is low, in glutamic acid; the opposite is true in fractions of buckwheat. The only gross similarity between the germ of wheat and buckwheat was that they contained more arginine than the respective endosperms. The high lysine, relatively low glutamic acid and proline, and relatively low ammonia content of buckwheat hydrolyzates confirm reports on high concentrations of soluble proteins; such soluble proteins are rich in lysine and may account for the excellent amino acid composition of buckwheat.

Several scoring procedures have been recommended as preliminary screening methods for predicting the limiting amino acid(s) and the approximate amino acid balance of a food. The procedures give different estimates of the nutritive value of the proteins (National Research Council, 1963; FAO/WHO, 1965).

Nutritional values of the proteins of buckwheat (mean of samples used in this study) are compared with wheat, oats, normal and opaque-2 corn, and an egg-reference pattern in Table VI. The data in Table VI are based on a scheme proposed by Kasarda *et al.* (1971). The ratio of essential to total amino acids (E/T) in all cereals is lower than in the egg-reference pattern. The E/T ratio in eggs is actually twice as high as would be needed for most efficient use of its essential amino acids. Consequently, the E/T ratio is of limited value in direct evaluation of nutritional adequacy. A more meaningful evaluation can be obtained by calculating the ratio of specific essential amino acids to the sum of the essential amino acids (A/TE).

A comparison of the A/TE ratio with the egg-reference pattern yields a chemical score which indicates the limiting amino acids; the lower the value, the more limiting the amino acid. Thus, in wheat, lysine is the first limiting amino acid; the next are threonine and isoleucine. In oats, isoleucine, lysine, and threonine are the first limiting amino

Table VI. Nutritional Values of the Proteins of Buckwheat Compared with Several Cereal Grains and with Whole Egg Protein as a Reference

	E/T values[a]					
					Corn[d]	
Essential amino acid	Egg reference pattern[b] 3.22	Wheat[b] 1.99	Oats[c] 2.38	Buckwheat 2.41	Normal 2.65	Opaque-2 2.54
	A/TE values[e]					
Isoleucine	129	122(95)[h]	102(79)	99(77)	94(73)	93(72)
Leucine	172	213	194	166	328	241
Lysine	125	82(66)	110(88)	158	66(53)	116(93)
Tyrosine and phenylalanine	195	243	220	179(92)	217	206
Cystine and methionine	107	196	107	106	76(71)	81(76)
Threonine	99	93(94)	86(87)	101	85(86)	96
Tryptophan	31	41	42[f]	60[f]	17(55)[g]	32[g]
Valine	141	150	139	132(94)	118(84)	135

[a] Grams essential amino acids per gram total N.. [b] From Kasarda *et al.* (1971). [c] From Robbins *et al.* (1971). [d] From Mertz *et al.* (1965). [e] Milligrams specific amino acid per gram of total essential amino acids. [f] From Tkachuk and Irvine (1969). [g] From Johnson *et al.* (1970). [h] Values in parentheses are A/TE for specific amino acid/A/TE for egg reference pattern × 100. The lowest value under a commodity shows the first limiting amino acid and gives a chemical score.

acids, though the balance between the essential amino acids is much better than in wheat. Similarly, opaque-2 is superior to normal corn. In agreement with feeding tests of Sure (1955), the chemical amino acid assays indicate that buckwheat has a better balance and better potential than cereal grains of supplementing foods which are low in lysine.

There is presumptive evidence that concentrations of the essential amino acids of egg protein are higher than concentrations required by man (FAO/WHO/UNICEF, 1970). Isoleucine and methionine are particularly high, and the use of egg as a reference may overestimate the extent to which those amino acids are limiting and may underestimate the quality of a protein for human use. Consequently, the nutritional value of opaque-2 corn, oat, and buckwheat proteins is probably higher than indicated in the data given in Table VI. On the other hand, amino acid analyses do not measure one of the most important parameters that determine nutritive value of a food, its digestibility, and "chemical scores" should be considered primarily as a powerful and convenient screening tool.

ACKNOWLEDGMENT

The technical assistance of J. T. Gilbertson with the amino acid analyses is gratefully acknowledged. We thank J. W. Bailey (The Birkett Mills, Pen Yan, N.Y.) for the commercially milled samples, and H. G. Marshall for the samples of buckwheat and for useful suggestions in preparation of the manuscript.

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Received for review June 11, 1971. Accepted September 8, 1971. This work was a cooperative investigation of the Plant Science Research Division, Agricultural Research Service, USDA, and the Wisconsin Agricultural Experiment Station, Madison, Wisconsin. Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, and does not imply its approval to the exclusion of other products that may also be suitable.

Spectral and Gas Chromatographic Characteristics of Some *N*-Nitrosamines

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The mass spectral, infrared spectral, and gas–liquid chromatographic retention time data of 25 *N*-nitrosamines are reported, together with general procedures for the synthesis of *N*-nitrosamines not commercially available.

Since the discovery of the toxic and later the carcinogenic properties of dimethylnitrosamine (Barnes and Magee, 1954; Magee and Barnes, 1956) many investigators have been working with nitrosamines. The most extensive review on the properties of *N*-nitroso compounds has been reported by Druckrey *et al.* (1967).

The report of dimethylnitrosamine and its causal relationship to liver damage in sheep (Ender *et al.*, 1964) has raised the question of the occurrence of various nitrosamines in the food supply. The only confirmed reports of a nitrosamine in food products have been those of dimethylnitrosamine in herring fish meal (Ender *et al.*, 1964), in a South African Bantu food plant (DuPlessis *et al.*, 1969), and in various fish products (Fazio *et al.*, 1971). However, there have been other reports of the presence of nitrosamines in foods: wheat meal (Kroeller, 1967; Marquardt and Hedler, 1966), cheese (Freimuth and Gläser, 1970; Kroeller, 1967; Marquardt and Hedler, 1966), milk (Marquardt and Hedler, 1966), mushrooms (Ender and Ceh, 1968), African alcoholic beverage (McGlashan *et al.*, 1968), meat products (Ender and Ceh, 1968; Freimuth and Gläser, 1970), and fish products (Ender and Ceh, 1968; Howard *et al.*, 1970; Sen *et al.*, 1970). The accuracy of many of these reports is questionable, as specific methods for con-

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