### FOOD PROTEIN EVALUATION

# Amino Acid Proportions in Food Proteins Compared to Proportions Utilized in Rat Growth

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By microbiological assay the essential amino acid content of 30 foods associated with the southwestern part of the United States was determined. On the basis of the essential amino acid requirements of the rat, the following conclusions were made for the food groups analyzed. Cheese protein was fairly well balanced, except for a low proportion of methionine. Nut protein had a very high proportion of arginine and a low proportion of methionine, lysine, and tryptophan. Fresh vegetable protein had a low proportion of methionine, lysine, and histidine. Methionine was deficient in all the food groups analyzed. Coconut meat required preliminary treatment to give good microbiological assay results.

**P**ROTEIN IS A LIMITING FACTOR in the diets of many peoples in many countries. Even in the United States some people receive too little protein or protein too poor in quality for good nutritional status. As the population of the United States increases, protein foods will become more and more limited, and all available sources of protein will eventually have to be utilized. A knowledge of the amount of protein in foods and their amino acid contents will then become even more important than it is now.

Publications on the amino acid contents of food proteins are numerous. But, for an accurate knowledge of the quality of proteins in foods, information on the amino acid content of many foods from many parts of the country is needed. The essential amino acid content of ten foods grown in the southwestern part of the United States was reported in previous papers from this laboratory (6, 8). In the present paper the essential amino acid content of 30 additional foods, most of which were grown or produced in the southwestern part of the United States, is reported. A few foods for which only limited data on amino acids were available were also assayed.

#### Experimental

The foods listed in Table I were assayed for arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine. Most of these foods are well known and widely consumed in the United States, although several are used only in the West or Southwest. Jack cheese is typically western and is a semihard, naturally ripened, whole milk cheese. Mexican cheese is common only in the Southwest and Mexico and is consumed largely by the Spanish Americans. It is a semihard, unripened skim milk cheese with a relatively high salt and moisture content. Of the legumes, cranberry beans and garbanzos are consumed mainly in the Southwest.

The fresh vegetables were prepared for assay as described in a previous paper (8). The mature legumes were ground in a hammer mill and stored at room temperature. The cheeses and nuts were ground in a blender and portions of each sample stored at  $0^{\circ}$  C. for subsequent fat, ash, and moisture determinations. The remainder of the sample was extracted three times with Skellysolve B in a blender, dried at room temperature, and stored in desiccators.

Hydrolysis and microbiological assay of samples were done in the manner previously described (6, 8). Horn and others (7) reported that hydrolyzate humin of cereals and legumes contained substances that increased microbiological response and thus caused incorrect amino acid values. These substances were removable by filtration at pH 4.0 but not at pH 7.0. With the media and procedures employed in the work reported, there were no differences in results obtained by filtration at pH 4.0 and 7.0.

Coconut required special treatment before hydrolysis to remove the major part of the soluble carbohydrates. After grinding in a blender, the coconut was dried at 60° C. and extracted in a Soxhlet extraction apparatus with Skellysolve B for 16 hours to remove fat. This fatfree sample was ground to a fine powder and stirred in water for 30 minutes. The pH was adjusted to 1.5 and 10% sodium tungstate was added slowly. The pH was readjusted to 2.7, the mixture centrifuged, and supernatant liquid decanted.

# Table I. Essential Amino Acid Content of Thirty Foods

					Average % Amino Acid Content						
Food	Basis of Calculation	Arginine	Histidine	Isoleucine	Leucine	Lysine	Methionine	Phenyl- alanine	Threonine	Trypto- phan	Valine
				Amino Aci	d Content	of Cheese					
Blue cheese	Fresh weight	0.537	0.558	1.07	1.75	1.35	0.540	0.963	0,749	0.292	1.61
Jack cheese	Fresh weight	2.75 0.733 3.74	2.80 0.598 3.05	5.51 1.15 5.86	8.98 1.99 10.14	0.92 1.50 7.67	0.494	4.94 1.05 5.37	0.779 3.97	0.251	8.25 1.76 8.96
Mexican cheese	Fresh weight	1.26	0.835	1.78	2.83	2.10 7.84	0.654	1.50	1.12	0.307	2.00
Oregon Cheddar	Fresh weight	0.932	0.744	1.54	1.80	1.97	0.668	1.59	1.04	0.307	1.89
Swiss cheese	Fresh weight	0.970	0.918	1.66	3.03	2.16	0.685	1.51	1.08	0.346	2.14
Tillamook	Fresh weight Protein	1.17	0.861	1.65	2.04	2.10	0.722	1.62	1.19	0.343	2.12
Velveeta	Fresh weight Protein	0.533 3.85	0.458 3.31	0.874 6.31	1.49 10.79	0.977 7.06	0.413	0.798 5.77	0.625 4.52	0.160 1.16	1.17 8.47
				Amino A	cid Conten	t of Nuts					
Acorns	Fresh weight Protein	0.615 5.88	0.213 2.04	0.479 4.58	0.690 6.60	0.541 5.18	0,118 1,13	0.404 3.86	0.370 3.54	0.107 1.02	0.613 5.86
Almonds	Fresh weight Protein	2.32 10.12	0.530 2.31	0.892 3.89	1.48 6.48	0.593 2.59	0.266 1.16	1.17 5.10	0.623 2.72	0.1 <b>81</b> 0.789	1.15 5.01
Brazil nuts	Fresh weight Protein	2.12 13.23	0.347 2.17	0.601 3.76	1.08 6.73	$\begin{array}{c} 0.341\\ 2.13 \end{array}$	0.871 5.45	0.526 3.29	0.364 2.28	0.153 0.957	0.774 4.84
Cashew nuts	Fresh weight Protein	1.82 9.61	0.362 1.91	1.06 5.60	1.32 6.97	0.688 3.63	0.306 1.62	0.826 4.34	0.639 3.38	0.410 2.16	1.38 7.29
Coconuts	Fresh weight Protein	0.499 14.59	0.067 1.96	0.127 3.71	0.245 7.16	0.116 3.39	0.068 1.99	0.154 4.50	0.118 3.45	$\begin{array}{c} 0.024\\ 0.702 \end{array}$	0.209 6.11
Filberts	Fresh weight Protein	1.66 13.60	0.266 2.18	0.552 4.52	0.784 6.42	0.295 2.42	0.146 1.20	0.457 3.74	0.362 2.96	0.242 1.98	0.723 5.92
Pecans	Fresh weight Protein	1.04 11.46	0.190 2.10	0.456 5.04	0.594 6.56	0.323 3.57	0.109 1.20	0.430 4.75	0.280 3.09	0.098 1.09	0.487 5.38
Walnuts	Fresh weight Protein	$\begin{array}{c} 2.10\\ 12.98\end{array}$	0.375 2.29	0.707 4.33	1.13 6.94	0.408 2.50	0.280 1.72	0.707 4.33	0.544 3.33	0.163 1.00	0.897 5.50
			Ami	no Acid Co	ntent of M	ature Leg	umes				
Cranberry beans	Fresh weight	1.02	0.631	1.18	1.67	1.63	0.216	1.19	0.910	0.272	1.21
Garbanzos	Fresh weight	1.93	0,559 2,61	1.09 5.10	9.00 1.59 7.42	0.04 1.36 6.34	0.375	0.45 1.46 6.77	4.95 0.791 3.69	1.40 0.155 0.722	1.35
Kidney beans	Fresh weight Protein	1.12	0.661	1.16	1.93	1.52	0.252	1.37	0.950	0.226	1.50
Lentils	Fresh weight Protein	2.27	0.620	1.16	1.88	1.43	0.201	1.12	0.940	0.202	1.58
Lima beans	Fresh weight Protein	1.31	0.731	1.28	1.78	1.25	0.311	1.32	1.01	0.188	1.66
Pink beans	Fresh weight Protein	0.957	0.667	1.17	1.63	1.22	0.210	1.22	0.945	0.220	1.17
Split peas, green	Fresh weight Protein	2.25	0.519	1.04	1.66	1.40	0.163	1.04	0.835	0.201	1.20
Split peas, yellow	Fresh weight Protein	2.10	0.477	1.00	1.59	1.41	0.159	0.991	0.805	0.189	1.16
White beans, large	Fresh weight Protein	1.25	0.677	1.36	1.78	1.76	0.231	1.28	0.880	0.254	1.26
White beans, small	Fresh weight Protein	1.14 5.83	0.581 2.97	1.28 6.54	1.66 8.48	1.12 5.72	0.220 1.12	1.33 6.80	1.05 5.36	0.265 1.35	1.25 6.39
			Ami	no Acid Co	ontent of Fr	esh Veget	ables				
Fresh asparagus tips	Fresh weight Protein	0.163 5.44	0.042	0.107	0.100	0.131	0.048	0.087 2.92	0.091	0.026	0.137 4.59
Fresh Brussels sprouts	Fresh weight	0.266	0.100	0.188	0.180	0.179	0.046	0.160	0.130	0.033	0.173
Fresh Lima beans	Protein Fresh weight	6.48 0.294	2.41 0.203	4.58 0.415	4.38 0.580	4.35 0.383	1.12 0.088	3.89 0.376	3.18 0.307	0.801 0.091	4.21 0.435
Fresh green peas	Protein Fresh weight	4.39 0.659	3.03 0.112	6.19 0. <b>314</b>	8.66 0.289	5.72 0.379	1.32 0.047	5.61 0.274	4.58 0.203	1.36 0.059	6.49 0.267
Fresh green beans	Protein Fresh weight	12.09 0.090	2.06 0.037	5.78 0.094	5.28 0.112	6.96 0.0 <u>9</u> 3	0.853	5.01 0.049	3.72 0.077	1.08	4.90 0.096
Whole egg protein (	Protein 3)	4.35 6.6	1.80 2.4	4.51 7.7	5.39 9.2	4.47 7.0	1.19 4.0	2.36 6.3	3.71 4.3	1.93 1.5	4.65 7.2
<sup><i>a</i></sup> Nitrogen $\times$ 6.2	5.										
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#### Table II. Analyses of Food

(Per cent of fresh weight)

Food	Protein (N X 6,25)	Moisture	Fat	Ash
Cheese	(			
Blue cheese Jack cheese Mexican cheese Oregon Cheddar Swiss cheese Tillamook Velveeta	19.53 19.61 26.80 24.65 26.79 26.92 13.82	47.27 41.44 48.47 37.08 32.46 33.75 45.05	24.14 31.12 11.94 29.87 30.50 30.51 27.04	5.52 3.78 6.54 3.34 5.08 3.19 5.40
Nuts				
Acorns Almonds Brazil nuts Cashew nuts Coconuts Filberts Pecans Walnuts	10.4522.9315.9918.933.4212.219.0516.31	4.12 4.07 4.53 4.76 54.62 7.09 3.57 3.85	33.03 53.84 67.92 49.30 21.77 63.88 72.69 65.94	3.08 2.75 2.96 2.63 0.70 2.25 1.48 1.98
Mature legumes				
Cranberry beans Garbanzos Kidney beans Lentils Lima beans Pink beans Split peas, green Split peas, yellow White beans, large White beans, small	18.44  21.46  21.04  24.52  21.34  18.17  21.09  19.79  20.24  19.57	8.42 8.00 11.62 8.64 9.36 8.70 9.44 9.40 8.83 7.14	0.64 6.02 1.24 0.68 1.59 1.70 1.09 1.04 1.89 2.00	4.26 1.96 2.96 3.29 4.10 2.74 2.78 3.62 3.94
Fresh vegetables Asparagus tips Brussels sprouts Lima beans Green peas Green beans	2.99 4.11 6.70 5.45 2.07	91.83 88.32 69.92 79.14 90.91	••••	0.68 0.84 1.32 0.71 0.69

The solids were hydrolyzed with 20% hydrochloric acid ( $\delta$ ) and the hydrolyzate was combined with the supernatant liquid. This solution of hydrolyzate and supernatant liquid was used for the microbiological assay.

#### **Results and Discussion**

The percentages of essential amino acids, calculated on the basis of fresh weight and protein content of 30 foods, are given in Table I. The percentages of proteins, moisture, fat, and ash are given in Table II. Although the amino acid composition of foods is usually expressed on a basis of per cent protein (N=16), percentages of fresh weight are also included to correspond, in part, to U. S. Department of Agriculture Handbook 8 (17), which lists the food components in terms of raw, processed, and prepared edible portions. This makes Table I of more value for diet preparation.

The amounts of essential amino acids in the proteins of foods do not give an accurate assessment of the nutritive value of the proteins. The proportions in which the amino acids are contained in the proteins are highly important. This has been amply demonstrated by the work of Rose (15, 16) on the amino acid requirements of rats; and by the work of Loosli (10) and Mertz, Beeson, and Jackson (11) on the amino acid requirements of swine. According to Albanese (1), the human infant up to 9 months of age requires lysine, isoleucine, and tryptophan in approximately the same proportions as Rose (15, 16) found for rats. Thus the nutritive values of the proteins in the foods in Table I can be better evaluated by determining the relative proportions of the ten amino acids in the proteins.

Table III compares the proportions of amino acids in the food proteins with the proportions that Rose (15) found to be required by the rat, and includes values for foods from a previous publication ( $\delta$ ). The proportions were calculated from the analytical data by a procedure similar to one recently reported by Flodin (4). In this procedure the content of tryptophan because of its low value in most food proteins, is made equivalent to 1.0 and the rest of the amino acids are proportioned to it. However, tryptophan was contained in such low percentages in most of the foods that it could not logically be used as a reference acid with the value 1. Therefore, threonine was used as a reference and given a value of 2.5, which is equivalent to the proportion, as compared to tryptophan, that is required by the rat.

Included in Table I are the percentages of essential amino acids in whole egg protein, to which reference may also be made for the amounts of individual amino acids present in the foods analyzed. The use of a standard protein (whole egg) was pioneered by Mitchell and Block (12).

Using the proportions of amino acids (Table III) as a measure for protein quality, all the proteins in the foods reported in this paper are deficient in methionine, except Brazil nuts. Doubtless, the proteins in some of the foods contain cystine which would augment the methionine and niacin which would augment the tryptophan (9). As would be expected, cheese protein is fairly well balanced in all the amino acids except methionine. Nut protein is off balance, with a very high proportion of arginine, a low proportion of lysine and, except for Brazil nuts, a low proportion of methionine. The protein of both fresh and dried legumes contains low proportions of lysine, methionine, and, with the exception of snap beans, tryptophan. The fresh vegetable proteins contain low proportions of methionine, histidine, and lysine.

In another method of protein evaluation, proposed by Oser (1-1), the essential amino acids in food protein are compared to the essential amino acids in whole egg protein to obtain an essential amino acid index (EAA). From these values the biological value (BV), may be estimated. The biological values, thus obtained, are reported to correspond closely to the values obtained mainly from rat studies. In Table IV are presented the values calculated by Oser's method for the various food groups reported in this paper.

In human dietaries several food proteins are usually consumed during one meal. Therefore, a deficiency of an amino acid in one food may be offset by a more than adequate amount of the same amino acid in some other food, or the deficiency may be at least partly overcome by increasing the intake of the deficient food protein. The former method of balancing amino acid intake depends upon the assembling of a diet through the knowledge of food components and their interrelationships. The latter method has been demonstrated by Mulford and Griffith (13), who found that by increasing the casein content of rat diets methionine deficiency could be overcome. Almquist (2) has generalized the gain in efficiency obtainable by the increased intake of deficient proteins; however, Grau (5) has shown that

					,	11.0	<b>B</b> 1.4	~,	-	
Rat growth	Arg. 1 0	<i>п</i> із. 2 ∩	1si. 2 5	1.eu. 4.0	Lys.	Mer.º	701.º	1hr. 25	1 ry.	Val. 35
Cheese	1,0	2.0	2.5	4.V	5.0	5.0	5.5	2,5	1.0	5.5
Jack	1.8 2.4	1.9 1.9	3.6 3.7	5.8 6.4	4.5 4.8	1.8 1.6	3.2 3.4	2.5 2.5	$1.0 \\ 0.8$	5.4 5.6
Mexican	$\frac{2.8}{2.2}$	1.9	4.0	6.3	4.7	1.5	3.4	2.5	0.7	4.5
Swiss	2.2	2.1	3.8	7.0	5.0	1.6	3.5	2.5	0.8	4.5 5.0
Tillamook Velveeta	2.5 2.1	$1.8 \\ 1.8$	3.5 3.5	4.3 6.0	4.4 3.9	1.5 1.6	3.4 3.2	2.5 2.5	0.7 0.6	4.5 4.7
Average	2.3	1.9	3.7	5.7	4.6	1.6	3.4	2.5	0.8	4.9
Nuts										
Acorns	4.2	1.4	3.2	4.7	3.7	0.8	2.7	2.5	0.7	4.1
Brazil	14.5	2.4	4.1	7.4	2.3	5.6	3.6	2.5	1.0	5.0
Cashew Coconuts	7.1 10.6	1.4	$\frac{4.1}{2.7}$	$\frac{5.2}{5.2}$	2.7	1.2	3.2	2.5	1.6	5.4 4.4
Filberts	11.5	1.8	3.8	5.4	2.0	1.0	3.2	2.5	1.7	5.0
Peanuts, raw <sup>o</sup> Peanuts, roasted <sup>b</sup>	9.1 8.5	2.4	3.9	4.3 4.6	3.3	$0.8 \\ 0.8$	4.8	$\frac{2.5}{2.5}$	$1.0 \\ 1.0$	4.7
Pecans	9.2	1.7	4 1	5.3	2.9	1.0	3.8	2.5	0.9	4.4
Average	9.7 9.4	1.7	3.3 3.7	5.2 5.3	1.9 2.8	1.5	3.3 3.7	2.5 2.5	0.8 1.0	4.1 4.7
Mature legumes										
Cranberry beans	2.8	1.7	3.2	4.6	4.5	0.6	3.3	2.5	0.8	3.3
Blackeyed peas <sup>9</sup> Garbanzos	4.0 6.1	$\frac{2.0}{1.8}$	3.0	4.9 5.0	4.0	0.8	3.2	2.5	0.6	3.6
Kidney beans	2.9	1.7	3.0	5.1	4.0	0.7	3.6	2.5	0.6	3.9
Lima beans	6.0 3.2	$1.6 \\ 1.8$	$\frac{3.1}{3.2}$	5.0 4.4	3.8 3.1	$0.5 \\ 0.8$	3.0 3.3	2.5	0.5	4.2
Pink beans Pinto beansh	2.5	1.8	3.1	4.3	3.2	0.6	3.2	2.5	0.6	3.1
Split peas, green	6.7	1.6	3.1	4.8 5.0	5.9 4.2	0.6	3.5 3.1	2.5	0.6	3.6
Split peas, yellow White beans,	6.5	1.5	3.1	4.9	4.4	0.5	3.1	2.5	0.6	3.6
White beans,	5.0	1.9	5.9	5.0	5.0	0.6	3.0	2.5	0.7	3.0
small	2.7	1.4	3.0	4.0	2.7	0.5	3.2	2.5	0.6	3.0
Average	4.2	1.7	3.2	4.8	2.9	0.7	3.4	2.5	0.6	3.6
Fresh legumes Blackeved peas <sup>b</sup>	4 0	2.2	3 1	4 4	4 2	0.0	37	2 5	0.6	35
Lima beans	2.4	1.6	3.4	4.7	3.1	0.7	3.1	2.5	0.7	3.5
Green peas Snap beans	8.1 2.9	1.4 1.2	3.9 3.0	3.5 3.6	4.7 3.0	0.6 0.8	3.4 1.6	2.5 2.5	0.7 1.3	3.3 3.1
Average	4.4	1.6	3.4	4.0	3.8	0.8	3.0	2.5	0.8	3.4
Fresh vegetables	4 -	1 2	2 0	2 7	2 (		<b>.</b> .			•
Asparagus tips Broccoli, heads <sup>b</sup>	4.5 4.4	1.2	$\frac{3.0}{2.7}$	2.7	3.6 1.8	$1.3 \\ 0.8$	2.4 2.7	2.5	0.7 0.8	3.8
Broccoli, stalks <sup>b</sup>	3.5	1.2	2.5	2.9	3.3	0.9	2.4	2.5	0.7	3.7
Potato, white <sup>b</sup>	3.2	0.9	2.7	3.4	3.5	0.9	3.1 3.1	2.5	0.6 0.07¢	3.3 3.7
Spinach <sup>b</sup> Sweet potato <sup>b</sup>	2.9 2.3	1.3	2.4 2.2	3.7 3.6	3.4 2.4	0.9 1.1	2.5 2.7	2.5	0.9	3.2 4.1
Average	4.0	1.3	2.7	3.2	3.1	1.0	2.7	2.5	0.8	3.7
		1								

Table III. Amino Acid Proportions in Food Protein Compared to Rat **Growth Requirements** 

at very high levels of protein intake efficiency of lysine utilization is diminished

#### Summary

Thirty foods were analyzed for their essential amino acid content-- seven cheeses, eight nuts, five fresh vegetables, and ten mature legumes. Values for the ten essential amino acids are presented on a basis of fresh weight and proteins. The amino acid proportions in the proteins of the foods are compared with the requirements of the growing rat.

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u.	stine may in part replace methionine and tyrosine replace phenylalanin	e.
b	Calculated from previous publication (3).	

<sup>e</sup> Not included in average.

Table IV. Essential Amino Acid Index and Biological Values of Food **Proteins**<sup>a</sup>

	Cheese	Nuts	Mature Legumes	Fresh Legumes	Fresh Veg.	Whole Egg
EAA	86	67	80	74	70	100»
BV	90	70	83	76	72	106¢

<sup>a</sup> Values calculated on methionine and not on methionine plus cystine.

<sup>b</sup> Assumed. Calculated.