# PROTEIN CONTENT AND AMINO ACID COMPOSITION OF BARLEYS FROM THE WORLD COLLECTION[1]

Y. POMERANZ[2], G. S. ROBBINS[3], R. T. SMITH[4], J. C. CRADDOCK[4], J. T. GILBERTSON[3], and J. G. MOSEMAN[4], National Barley and Malt Laboratory, Madison, Wis.

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## ABSTRACT

Kernel weight, protein content, and amino acid composition were determined for 113 barley cultivars from the USDA world collection. Two-row covered spring, six-row covered winter, six-row hull-less spring, and six-row covered spring types were represented. Two-row covered spring barleys were highest and six-row hull-less spring barleys were lowest for kernel weight. Protein content was highest in six-row hull-less spring barleys and lowest in six-row covered winter barleys. Six-row covered winter barleys were highest for lysine and threonine as percentage of protein, and six-row hull-less spring barleys were highest for lysine and threonine as percentage of sample. Correlations for all barley types were negative between protein and leucine, and positive between isoleucine and valine. Representative cultivars were selected, for each of the four barley types, on the basis of kernel weight, protein content, and contents of essential amino acids. Large differences among the barley types indicated that the cultivars tested would provide a rich source of germplasm for plant breeders.

Investigations of Munck *et al.* (1) have led to the discovery of a high-protein, high-lysine barley line (Hiproly) with improved nutritional value. The Hiproly barley, C.I. 3947, is of Ethiopian origin; it is an erectoid type with naked, slightly shriveled seeds, and requires a long photoperiod. A sister line to Hiproly, C.I. 4362, has a similar growth habit, but has smoother, heavier seeds. Both lines are high in protein, but C.I. 3947 has substantially more lysine in the protein and a higher nutritional value than C.I. 4362. Hiproly, with its high protein content, provides more of the essential amino acids than commercially grown oats or any other cereal grain, including the best maize mutants (2). However, the poor agronomic characteristics of Hiproly preclude its commercial cultivation. Plant breeders are highly interested in developing acceptable crosses between Hiproly and commercially grown cultivars. Hopefully, such crosses would combine the excellent amino acid balance of Hiproly with overall acceptable agronomic characteristics in cultivars useful for food, feed, and malting purposes.

In a recent study (3), kernel weight, protein content, and amino acid composition were determined in isogenic pairs of two-row and six-row barleys and in two-row and six-row backcrosses to a two-row barley cultivar (Bonneville). The two-row selections were higher both in kernel weight and in

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[2]Chemist in Charge, Barley and Malt Laboratory, USDA, and Professor, Agronomy Department, University of Wisconsin, Madison. Present address: Director, Grain Marketing Research Center, ARS, USDA, Manhattan, KS 66502.

[3]Respectively: Research Chemist and Research Technician, Barley and Malt Laboratory, Madison, WI 53705.

[4]Respectively: Research Agronomist, Research Agronomist, and Chairman, Plant Genetics and Germplasm Institute, Agricultural Research Service, USDA, Beltsville, MD 20705.

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**TABLE I**
**Means and Least Significant Differences[a] for Kernel Weight, Protein Content,[b] and Amino Acid Composition[c] for Four Barley Types**

| Metameter | Metameter Means by Barley Type: Two-Row spring covered n=26 | Six-Row winter covered n=12 | Six-Row spring hull-less n=9 | Six-Row spring covered n=66 | L.S.D. P=0.05 | General Mean n=113 | Coefficient of Variation % |
|---|---|---|---|---|---|---|---|
| Kernel weight (mg) | 47.05 | 41.98 | 33.10 | 43.33 | 3.57*[d] | 43.23 | 16.17 |
| Protein (%) | 14.17 | 10.34 | 15.27 | 13.77 | 0.89* | 13.62 | 12.94 |
| Lysine | 3.24 | 3.75 | 3.35 | 3.33 | 0.14* | 3.35 | 8.12 |
| Histidine | 2.06 | 2.20 | 2.10 | 2.07 | 0.05* | 2.09 | 4.18 |
| Ammonia | 3.33 | 3.10 | 3.26 | 3.27 | 0.08* | 3.26 | 4.64 |
| Arginine | 4.28 | 4.53 | 4.21 | 4.40 | 0.18* | 4.37 | 7.90 |
| Aspartic acid | 6.14 | 6.85 | 6.05 | 6.25 | 0.26* | 6.27 | 8.12 |
| Threonine | 3.06 | 3.32 | 3.03 | 3.06 | 0.08* | 3.09 | 5.10 |
| Serine | 3.45 | 3.58 | 3.47 | 3.44 | 0.13 | 3.46 | 7.37 |
| Glutamic acid | 27.75 | 25.83 | 28.10 | 27.36 | 0.62* | 27.35 | 4.41 |
| Proline | 12.58 | 11.25 | 12.38 | 12.40 | 0.71* | 12.32 | 11.40 |
| Half-cystine | 1.21 | 1.06 | 1.18 | 1.17 | 0.09 | 1.17 | 14.74 |
| Glycine | 3.72 | 4.12 | 3.73 | 3.78 | 0.14* | 3.81 | 6.91 |
| Alanine | 4.05 | 4.48 | 3.97 | 4.06 | 0.14* | 4.10 | 6.68 |
| Valine | 4.86 | 5.26 | 4.92 | 4.97 | 0.12* | 4.97 | 4.57 |
| Methionine | 2.48 | 2.68 | 2.52 | 2.59 | 0.21 | 2.57 | 15.71 |
| Isoleucine | 3.59 | 3.76 | 3.56 | 3.59 | 0.06* | 3.61 | 3.46 |
| Leucine | 6.47 | 6.82 | 6.51 | 6.50 | 0.10* | 6.53 | 3.08 |
| Tyrosine | 2.54 | 2.43 | 2.42 | 2.54 | 0.18 | 2.52 | 14.10 |
| Phenylalanine | 5.27 | 5.06 | 5.35 | 5.24 | 0.11* | 5.24 | 4.06 |
| REN[e] | 87.67 | 87.21 | 87.98 | 88.24 | 2.54 | 87.98 | 5.67 |

[a]Single class analysis of variance with unequal samples; L.S.D. divisor = 29.
[b]Nitrogen × 6.25, moisture-free basis.
[c]Per cent of protein (g amino acid/100 g recovered).
[d]*F ratio significant at $\lesseqgtr$ 0.05 probability level.
[e]Per cent recovered nitrogen.

**TABLE II**

**Means and Least Significant Differences[a] for Amino Acids Expressed as Percentage of Sample for Four Barley Types**

| Metameter | Metameter Means by Barley Type | | | | | | |
|---|---|---|---|---|---|---|---|
| | Two-Row spring covered n=26 | Six-Row winter covered n=12 | Six-Row spring hull-less n=9 | Six-Row spring covered n=66 | L.S.D. P=0.05[b] | General Mean n=113 | Coefficient of Variation % |
| Lysine | 0.46 | 0.38 | 0.51 | 0.46 | 0.03 | 0.45 | 11.62 |
| Histidine | 0.29 | 0.23 | 0.32 | 0.29 | 0.02 | 0.28 | 11.07 |
| Ammonia | 0.47 | 0.32 | 0.50 | 0.45 | 0.04 | 0.45 | 15.93 |
| Arginine | 0.60 | 0.46 | 0.64 | 0.60 | 0.03 | 0.59 | 11.07 |
| Aspartic acid | 0.86 | 0.70 | 0.92 | 0.86 | 0.05 | 0.85 | 11.25 |
| Threonine | 0.43 | 0.34 | 0.46 | 0.42 | 0.02 | 0.42 | 10.66 |
| Serine | 0.49 | 0.37 | 0.53 | 0.47 | 0.03 | 0.47 | 13.20 |
| Glutamic acid | 3.95 | 2.69 | 4.31 | 3.78 | 0.31 | 3.75 | 16.22 |
| Proline | 1.81 | 1.18 | 1.90 | 1.72 | 0.18 | 1.70 | 20.63 |
| Half-cystine | 0.17 | 0.11 | 0.18 | 0.16 | 0.02 | 0.16 | 18.17 |
| Glycine | 0.52 | 0.43 | 0.57 | 0.52 | 0.03 | 0.51 | 9.69 |
| Alanine | 0.57 | 0.46 | 0.60 | 0.56 | 0.03 | 0.55 | 9.78 |
| Valine | 0.69 | 0.54 | 0.75 | 0.68 | 0.04 | 0.67 | 11.94 |
| Methionine | 0.35 | 0.28 | 0.38 | 0.36 | 0.03 | 0.35 | 17.67 |
| Isoleucine | 0.51 | 0.39 | 0.54 | 0.50 | 0.03 | 0.49 | 12.35 |
| Leucine | 0.92 | 0.70 | 0.99 | 0.89 | 0.05 | 0.89 | 11.23 |
| Tyrosine | 0.36 | 0.25 | 0.37 | 0.35 | 0.03 | 0.34 | 16.20 |
| Phenylalanine | 0.75 | 0.52 | 0.82 | 0.72 | 0.05 | 0.72 | 14.95 |

[a]Single class analysis of variance with unequal samples; L.S.D. divisor = 29.
[b]F ratios from all analyses of variance were significant at $\leq$ 0.05 level of probability.

protein content than the six-row selections. In addition, the proteins in the two-row selections contained more glutamic acid and proline (and less of most of the other amino acids) than the proteins of the six-row selections. Amino acids were determined in proteins of 15 two-row and 21 six-row cultivars (grown for 2 years), each from two locations which consistently produced barleys varying widely in protein content. The results showed that the amino acid composition in the two types of barleys (two-row and six-row) depended on their protein contents. It was concluded that amino acid composition in proteins of two-row and six-row barleys was governed by the total protein content, rather than by the type of barley. There was a highly significant positive correlation between the lysine and aspartic acid contents of proteins in all types and groups of barleys that were studied.

This study was conducted to survey protein and amino acid composition and kernel weight of barleys from the USDA world collection of barley maintained in the Plant Genetics and Germplasm Institute at the Beltsville Agricultural Research Center, Beltsville, Md. Emphasis was placed on selection of barleys from foreign sources. The objectives were to select potential breeding stocks, to depict reliable differences within and between barley types, and to measure intradependencies and relationships among amino acids and with protein content and kernel weight.

## MATERIALS AND METHODS

The 113 barley samples selected for this study originated from the following countries: Algeria—3, Argentina—1, Bolivia—1, Bulgaria—3, People's Republic of China—12, Cyprus—1, United Arab Republic—9, England—1, Ethiopia—27, Finland—1, France—1, Hungary—2, India—7, Iran—4, Iraq—5, South Korea—1, Morocco—1, Republic of South Africa—1, Spain—3, Syria—2, Tunisia—1, Turkey—3, U.S.S.R.—21, and U.S.—2.

Classification according to type gave: 26 two-row covered spring, 66 six-row covered spring, 12 six-row covered winter, and 9 six-row hull-less spring barleys.

Crude protein (Kjeldahl) and amino acid analyses of acid hydrolysates (on a Beckman 121 automatic analyzer) were determined as described elsewhere (4). The amino acids that were determined did not include tryptophan. Crude protein was estimated by the product of nitrogen $\times$ 6.25 and is reported on a moisture-free basis.

## RESULTS AND DISCUSSION

Much of our results and discussions emphasize protein content and amino acid composition (primarily the limiting lysine and threonine and the other essential amino acids) which are of main interest to nutritionists and plant breeders. We have included kernel weight in the results as it is a component of yield (5) which is of primary interest to the farmer.

Means and least significant differences for kernel weight, protein content, and amino acid composition (expressed as either percentage of protein or as percentage of sample) for each of the four barley types are summarized in Tables I and II.

Kernel weight was highest for the two-row covered spring cultivars and lowest

TABLE III

**Linear Correlations for Protein Content and Kernel Weight for All Amino Acids (% of Protein) Tested for Each Barley Type**

| Metameter | Correlation Coefficients by Barley Type: Two-Row spring covered (d.f.=24) | | Six-Row winter covered (d.f.=10) | | Six-Row spring hull-less (d.f.=7) | | Six-Row spring covered (d.f.=64) | |
|---|---|---|---|---|---|---|---|---|
| | Protein | Kernel wt | Protein | Kernel wt | Protein | Kernel wt | Protein | Kernel wt |
| Kernel weight | 0.513*[a] | | 0.471 | | 0.042 | | 0.001 | |
| Lysine | −0.747* | −0.662* | −0.737* | −0.281 | 0.257 | −0.547 | −0.637* | −0.183 |
| Histidine | −0.651* | −0.665* | −0.715* | −0.062 | −0.922* | −0.239 | −0.528* | −0.134 |
| Ammonia | 0.597* | 0.478* | 0.600* | 0.537 | 0.826* | −0.447 | 0.540* | 0.139 |
| Arginine | −0.634* | −0.510* | −0.711* | 0.244 | −0.459 | −0.355 | −0.488* | −0.043 |
| Aspartic acid | −0.800* | −0.621* | −0.684* | −0.273 | −0.858* | −0.232 | −0.311* | −0.059 |
| Threonine | −0.749* | −0.731* | −0.826* | −0.307 | −0.659 | −0.429 | −0.547* | −0.247* |
| Serine | −0.351 | −0.286 | −0.299 | 0.340 | 0.021 | −0.061 | −0.362* | −0.173 |
| Glutamic acid | 0.659* | 0.608* | 0.916* | 0.432 | 0.761* | 0.244 | 0.717* | −0.133 |
| Proline | 0.854* | 0.556* | 0.603* | −0.132 | 0.292 | 0.264 | 0.384* | −0.057 |
| Half-cystine | 0.181 | −0.088 | 0.345 | 0.492 | 0.033 | −0.075 | −0.318* | 0.177 |
| Glycine | −0.778* | −0.622* | −0.843* | −0.352 | −0.893* | −0.068 | −0.590* | −0.169 |
| Alanine | −0.791* | −0.602* | −0.761* | −0.189 | −0.755* | 0.145 | −0.657* | −0.015 |
| Valine | −0.667* | −0.465* | −0.762* | −0.230 | −0.409 | 0.450 | −0.565* | 0.025 |
| Methionine | −0.176 | −0.306 | −0.112 | −0.436 | −0.473 | −0.016 | −0.254* | 0.219 |
| Isoleucine | −0.350 | −0.186 | −0.608* | −0.021 | −0.120 | 0.301 | −0.277* | −0.122 |
| Leucine | −0.716* | −0.579* | −0.857* | −0.298 | −0.831* | 0.315 | −0.589* | −0.293* |
| Tyrosine | −0.472* | −0.263 | −0.321 | 0.394 | −0.039 | 0.377 | −0.280* | 0.035 |
| Phenylalanine | 0.229 | 0.202 | 0.568 | 0.121 | 0.041 | −0.021 | 0.536* | −0.247* |

[a]*Correlation coefficient significant at ≤0.05 level of probability.

**TABLE IV**
**Selected Cultivars that Typify[a] each Barley Type for Kernel Weight, Protein Content, and Essential Amino Acids Expressed as % of Protein**

| Type, Name, and Source | Kernel Weight mg | Protein % | Lysine | Threonine | Methionine | Valine | Isoleucine | Leucine | Phenylalanine |
|---|---|---|---|---|---|---|---|---|---|
| Two-Row spring covered | | | | | | | | | |
| Dentil—Syria | 54.4 | 15.6 | 2.8 | 2.9 | 2.8 | 4.4 | 3.5 | 6.3 | 5.3 |
| C.I. 3955[b]—U.A.R. | 59.5 | 16.9 | 2.7 | 2.9 | 1.5 | 4.5 | 3.5 | 6.4 | 5.5 |
| C.I. 4968—Iraq | 53.5 | 15.4 | 2.8 | 3.1 | 1.8 | 4.5 | 3.5 | 6.3 | 5.4 |
| Six-Row winter covered | | | | | | | | | |
| C.I. 3882—Algeria | 34.0 | 8.0 | 4.4 | 3.6 | 3.1 | 5.5 | 3.8 | 7.1 | 5.0 |
| C.I. 4327—U.S.S.R. | 41.7 | 8.9 | 4.1 | 3.4 | 3.0 | 5.6 | 3.9 | 7.1 | 5.1 |
| Derbent—U.S.S.R. | 37.9 | 8.3 | 4.0 | 3.6 | 1.9 | 5.5 | 3.8 | 7.0 | 5.0 |
| Six-Row spring hull-less | | | | | | | | | |
| C.I. 4339—China | 33.3 | 14.3 | 3.2 | 3.0 | 2.6 | 5.1 | 3.5 | 6.6 | 5.6 |
| Kharsila—India | 31.7 | 14.4 | 3.1 | 3.2 | 2.8 | 4.6 | 3.4 | 6.6 | 5.3 |
| Hiza Hacha—Bulgaria | 22.7 | 13.3 | 3.3 | 3.1 | 2.7 | 4.9 | 3.6 | 6.5 | 5.4 |
| Six-Row spring covered | | | | | | | | | |
| C.I. 5631—U.S.S.R. | 26.5 | 16.3 | 3.2 | 2.9 | 2.4 | 4.7 | 3.6 | 6.4 | 5.5 |
| C.I. 12622—Ethiopia | 39.5 | 13.9 | 3.5 | 3.1 | 2.5 | 5.2 | 3.6 | 6.6 | 5.3 |
| C.I. 12800—Ethiopia | 41.2 | 14.8 | 3.1 | 2.9 | 2.6 | 4.8 | 3.5 | 6.5 | 5.6 |

[a]Posterior probability of membership in each group >0.95 determined by discriminant analysis.
[b]USDA cereal accession number.

for the six-row hull-less spring cultivars (Table I). Both the small-kernelled six-row hull-less spring and the large-kernelled two-row covered spring barleys contained much more protein than the six-row covered winter barleys. The low average protein content of the six-row covered winter barleys may reflect an environmental response, since all the cultivars were grown at a location favoring spring types. The rest of the data in Table I concerns the amino acid composition, or quality, of the barley proteins. There were significant differences among the barley types, except for serine, tyrosine, and the sulfur-containing amino acids. The lack of significance for the latter may be related to their large coefficients of variation which, in part, may be the result of relatively lowered analytical precision.

The results in Table II show significant differences in amounts of all amino acids provided by barleys from the four types. Whereas there are large differences in coefficients of variation for the amino acid data in Table I, the coefficients of variation for practically all essential amino acids in Table II are relatively more comparable.

The data in Table III show correlations of amino acid composition with protein content and kernel weight, both of which must be considered in a sound plant-breeding program. The data are given for the four types of barley. As the chi-square test for homogeneity of correlation coefficients was significant, a pooled correlation over barley types was not appropriate. The increase in protein content is associated mainly with an increase in the nonessential amino acids glutamic acid and proline, major components of barley hordeins and glutelins. The increase in protein is accompanied by decreases in practically all essential amino acids. There is a significant low positive correlation between kernel weight and protein only for the two-row covered spring barleys. Consequently, an

**TABLE V**
**Within Barley-Type Symbolic[a] Correlations**

| Metameters[b] | Kernel Weight | Protein | Threonine | Lysine | Valine | Methionine | Isoleucine |
|---|---|---|---|---|---|---|---|
| Protein | A | | | | | | |
| Threonine | −(AD) | −(ABD) | | | | | |
| Lysine | −A | −(ABD) | ABD | | | | |
| Valine | −A | −(ABD) | AD | ABD | | | |
| Methionine | | −D | | | | | |
| Isoleucine | | −(BD) | | D | ABCD | | |
| Leucine | −(AD) | −(ABCD) | ABD | ABD | ABD | | ABD |
| Phenylalanine | −D | D | −D | −D | −(AD) | −D | |

[a]Significant positive or negative correlation coefficients at $P \leq 0.05$ for: A = two-row spring covered, d.f. = 24; B = six-row winter covered, d.f. = 10; C = six-row spring hull-less, d.f. = 7; and D = six-row spring covered, d.f. = 64.
[b]Amino acid as % of protein.

increase in kernel weight (and associated yield) may not necessarily impair amino acid composition of the six-row spring barleys.

While the value of correlation coefficients is limited, especially for comparison among groups varying widely in sample numbers, the coefficients are frequently used indices of associations among measured variables for this type of data.

Table IV was constructed to select representative cultivars for each barley type. Three cultivars were selected for each type, to provide appropriate germplasm for studies on chemical and/or genetic differences. Only those metameters of primary interest to the plant breeder and nutritionist (kernel weight, protein, and essential amino acids) were included in the multiple measurement analysis (6). The cultivars are identified by C.I. number (or name) and source; values for kernel weight, protein, and essential amino acid metameters are shown in the table. Seed can be requested from J. C. Craddock, Curator of the World Collection of Barley, Plant Genetics and Germplasm Institute, Beltsville Agricultural Research Center (West), Beltsville, MD 20705.

Results given in Table V summarize all possible significant correlations for kernel weight, protein content, and essential amino acids. There is a positive relationship between valine and isoleucine and a negative relationship between protein and leucine for all barley types. For additional studies of relationships between different variables, cultivars listed in Table IV could be selected.

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