The Botanical Constituents of Wheat and Wheat Milling Fractions. II. Quantification by Amino Acids

S. A. JENSEN[1] and H. MARTENS[2]

ABSTRACT

The amino acid composition of manually dissected pericarp, testa, aleurone, endosperm, and germ from wheat was determined and compared to the amino acid composition of 50 wheat milling fractions. The data obtained were used in a multiple linear regression to quantify the botanical composition of pericarp, aleurone, endosperm, and germ in the wheat milling fractions. Comparison for pericarp, aleurone, and endosperm to an earlier method based on autofluorescence characteristics showed good agreement for pericarp and endosperm, whereas that of aleurone showed a lower correlation coefficient. Correlation to chemical constituents ash, fiber, starch, and niacin of both sets of measurements established that the differences between the two independent methods were due to inadequacies in the amino acid model rather than in the fluorescence method.

Cereal Chem. 60(2):172–177

Wheat consists of botanically defined components with widely different physical and chemical characteristics of importance in food technology as well as in nutrition. In an earlier article (Jensen et al 1982), the autofluorescence spectra of pericarp, aleurone, and endosperm were used to quantify these components in wheat milling fractions. The method was initially calibrated against mixtures of manually dissected fractions critically controlled for purity under the fluorescence microscope. We further analyzed the pure botanical fractions for their amino acid composition.

Amino acid data from various pure foods (Lindquist et al 1975, Martens 1979, Martens and Lea 1981) were previously used to determine the composition of mixed foods. This article describes how a similar statistical multivariate technique used amino acid data to determine the composition of wheat milling fractions in terms of pericarp, aleurone, endosperm, and germ. The wheat milling fractions are identical to those analyzed by the fluorescence method (Jensen et al 1982).

The present research, therefore, had two purposes: to define the amino acid composition of pure, manually dissected botanical fractions, and to use this information to estimate the botanical composition of the wheat milling fractions as an independent control of the results obtained by the fluorescence method.

MATERIALS AND METHODS

Raw Materials

The wheat fractions analyzed were identical to samples described previously (Jensen et al 1982).

Amino Acid Analysis

Acid Hydrolyses. Direct hydrolysis of the wheat samples with 6*N* HCl was performed to obtain hydrolysates suitable for all the amino acids present, except for cystine and tryptophan, and for the amides of glutamic and aspartic acid (Jonassen 1980).

Basic Hydrolysis. Samples for tryptophan determination were hydrolyzed with 5*N* $Ba(OH)_2$ according to the method described by Eggum (1968).

All amino acid results are based on duplicate determinations (for tryptophan single analyses) and expressed as a percentage (by weight) of the total amino acids measured (Table I). The data can be transformed to grams of amino acid per 100 g of protein by use of percent protein (N × 5.7) and the factor f (grams of amino acids per 100-g sample, dry basis). For aspartic acid in wheat, for example (Table III), with 12.0% protein and 11.35 g of amino acids per 100-g sample, our result was 5.4%. This is equivalent to (0.054 × 11.35)100/12.0 = 5.1 g of aspartic acid per 100 g of protein.

Determination of Moisture, Protein, and Niacin

The moisture content of all the wheat samples was measured by oven drying for 2 hr at 130° C. The protein content in the milled wheat samples was measured by the Kjeldahl method (Kjeldahl 1883), using the factor 5.7. Total nitrogen and niacin in the 10 decortication fractions were determined by the microbiological method (AACC 1979). The results for protein and niacin are reported on a dry weight basis.

Statistical Procedures

Amino Acid Pattern of Manually Dissected Wheat Fractions. Pericarp, aleurone, endosperm, and germ (referred to as pure components) were used in a weighted multiple linear regression (Martens 1979) to estimate the composition of these components in the milled wheat samples. When the different proteins behave additively in the amino acid analysis of wheat, the linear mixture model is written:

$$Y_{ik} = \Sigma_J^J = 1\ X_{ij} \times P_{jk} \qquad (1)$$

where Y_{ik} represents experimental data for amino acid variable i (i = 1, 2, ..., I) in sample k (k = 1, 2, ..., K); X_{ij} is the relative contribution of variable i in pure component j(j = 1, 2, ..., J); and P_{jk} is the amount of pure component j in sample k.

A weighted least squares estimate of P was chosen (Martens and Bach Knudsen 1980). This corresponds to a weighted multiple linear regression without offset term. Each amino acid in the model was weighted by its corresponding measurement noise (variance between parallels, Table I).

Amino acid data may be reported in a variety of forms. The chemical units of X and Y, however, must be consistent with one another. All the amino acid data in X and Y are expressed here as percentages by weight of the total amino acids measured; ie, each amino acid spectrum totals 100%. These units have the advantage of reducing measurement noise caused by sample weighting, providing for easy graphic analysis of the data. With these units for X and Y, however, the unit for P is given as weight fraction of the pure component proteins rather than as weight percent of the pure components. To obtain results in the latter unit, the following inverse weighting of P was performed. If f_j is the number of grams of amino acids recovered in 100 g of sample (dry basis) of pure component j, then:

$$P'_{jk} = P_{jk} \times (1/f_j)/\ \Sigma_{m=1}^{J} 1/f_m \qquad (2)$$

Finally, the component sum for each sample was forced to 100%:

$$P''_{jk} = P_{jk}/\Sigma_{m=1}^{J} P'_{mk} \times 100 \qquad (3)$$

Thus, the final estimates for botanical composition P_{jk} are given in weight percent of the pure components and are therefore

[1]Carlsberg Research Laboratory, Dept. of Biotechnology, Gamle Carlsberg Vej 10, DK-2500 Valby, Copenhagen, Denmark.
[2]Norwegian Food Research Institute, Dept. of Food Technology, P.O. Box 50, 1432 Aas, Norway.

comparable to the results obtained by the fluorescence method (Jensen et al 1982).

The aim of the present mathematical analysis was to estimate the composition of pericarp, aleurone, endosperm, and germ in the 50 wheat milling samples; thus $J = 4$, and $K = 50$. To balance measurement uncertainty against information, only those amino acids that showed good signal to noise ratios over the four botanical components were included in the model. This criterion was satisfied by seven amino acids (aspartic acid, glutamic acid, proline, glycine, alanine, lysine, and arginine); thus, $I = 7$. All calculations were made on a Hewlett-Packard 9825T computer.

RESULTS AND DISCUSSION

Amino Acid Composition

Manually Dissected Botanical Fractions. In contrast to the abundant information regarding the amino acid composition of commercial wheat mill streams, very few published investigations have defined the amino acid composition of the individual botanical constituents of wheat. This is primarily because separation of the botanical components of mature wheat is difficult and time consuming.

The amino acid pattern of the manually dissected botanical components is shown in Table I. The composition of testa was estimated from the difference of the amino acid compositions of aleurone and aleurone plus testa, assuming a weight proportion at 7:3 between aleurone and testa (Hinton 1947). These results indicate that testa showed an amino acid composition close to that of pericarp and aleurone for aspartic acid, proline, glycine, alanine, and histidine, whereas testa differs from all the other botanical components in its very low content of tryptophan (0.7%).

The results for pericarp and testa seem to be the first report for quantitative amino acid analysis of these individual components.

TABLE I
Amino Acid Composition of Botanical Components of Wheat

	Molecular Weight	Pericarp	Testa	Aleurone + Testa	Aleurone	Endosperm	Germ	Mean	$\overline{CV}$[a]
Aspartic acid	133	9.5[b]	7.9	7.9	7.9	4.2	10.4	8.0	3.66
Threonine	119	4.0	3.5	3.1[b]	2.9	2.2	4.0	3.2	2.02
Serine	105	3.9	4.0	3.2[b]	2.9	2.7	3.0	3.1	4.62
Glutamic acid	147	15.8	22.6	21.4	20.9	35.2	13.9	21.4	1.51
Proline	115	6.6[c]	7.6	6.7	6.3	12.9	4.8	7.5	1.10
Glycine	75	7.9	6.5	6.0	5.8	3.6	7.4	6.2	2.63
Alanine	89	6.6[c]	5.9	5.9	5.9	3.5	7.7	5.9	2.49
Valine	117	5.5[c]	4.3	5.0	5.3	4.2	6.5	5.5	1.62
Methionine	149	2.4	1.6	1.6	1.6	1.6[c]	2.0	1.8	1.70
Isoleucine	131	5.1	4.3	3.8	3.6	4.0[c]	4.1	3.1	1.85
Leucine	131	8.4	8.8	7.2	6.5	7.3	7.5	7.4	1.44
Tyrosine	181	3.7[c]	3.6	3.4	3.3	3.7	3.2	3.5	4.49
Phenylalanine	165	5.4	5.5	4.3	3.8	5.3	4.1	4.6	2.97
Histidine	155	1.6[c]	2.7	3.2	3.4	2.0	2.9	2.6	2.54
Lysine	146	4.6	4.1	4.6	4.8	2.1	8.3	4.9	3.53
Arginine	174	5.1[c]	6.5	9.7	11.1	3.6	8.7	7.4	1.44
Tryptophan	204	4.0	0.7	3.0	4.0	2.0	1.7	2.9	...
Sum		100.1	100.1	100.0	100.0	100.1	100.2		
f[d]		2.63	4.76	13.85	17.95	8.75	31.84		

[a] Coefficient of variation; standard deviation between parallels in percent of mean value.
[b] $CV_{ik} > 3 \times \overline{CV}$.
[c] $CV_{ik} > 2 \times \overline{CV}$.
[d] f = Grams of amino acids per 100-g of sample, dry basis.

TABLE II
Amino Acid Composition of the Botanical Components of Wheat

	Bran Fractions from Milling		Aleurone[a]	Endosperm[a]	Manually Dissected Germ
	Manually Dissected Bran	Hepburn et al (1960)	Tkachuk and Irvine (1969)	Fulcher et al (1972)	Fulcher et al (1972)	Tkachuk and Irvine (1969)
Aspartic acid	8.7	8.1	8.1	9.3	5.7	9.8
Threonine	3.6	3.5	3.8	3.8	3.1	4.6
Serine	3.6	5.6	5.7	5.0	6.1	5.1
Glutamic acid	18.5	19.8	22.2	18.3	33.5	16.0
Proline	6.7	7.5	7.5	4.6	11.5	4.5
Glycine	7.0	6.3	6.1	7.0	4.6	6.5
Alanine	6.3	5.7	5.0	6.5	3.9	6.8
Valine	5.3	5.7	4.8	5.5	4.1	6.6
Methionine	2.0	1.8	1.3	0.4	0.4	1.9
Isoleucine	4.5	4.0	3.6	3.2	3.5	3.8
Leucine	7.8	6.7	6.7	6.7	7.0	6.7
Tyrosine	3.6	3.5	3.2	2.8	3.1	2.9
Phenylalanine	4.9	4.4	4.1	4.2	4.4	3.7
Histidine	2.4	2.7	3.3	4.3	2.3	2.8
Lysine	4.6	4.6	4.9	5.9	3.1	7.5
Arginine	7.2	8.1	7.8	12.3	4.0	9.0
Tryptophan	3.5	1.9	2.2	...	...	1.7
Sum	100.2	100.3	100.2	99.8	100.3	99.9

[a] Separated by air-classification and centrifugation.

Numerous reports give the amino acid composition of wheat bran (Hepburn et al 1960, Kohler and Patter 1967, Tkachuk and Irvine 1969). Wheat bran includes pericarp, testa, and aleurone. In Table III, previously published data for bran are compared to our results for bran, where the amino acid composition was constructed by the amino acid composition of pericarp and aleurone plus testa, assuming the proportion to be 0.51:0.49 (Jensen et al 1982). The results are in reasonable agreement with one another.

Amino acid analysis of isolated aleurone cell content from wheat (Fulcher et al 1972) showed a high content of basic amino acids, especially arginine (Table II). The same tendency is shown in the data for aleurone (Table I), although deviations occur for aspartic acid, threonine, glutamic acid, proline, and lysine. This might be because the former study was performed on aleurone cell contents isolated from air-classified flour by differential centrifugation in a nonaqueous medium, and not on whole aleurone cells (ie, cell walls plus contents). This indicates differences in the amino acid compositions of aleurone cell walls and of aleurone cell contents.

In the same studies (Fulcher et al 1972), endosperm protein, isolated by air-classification and centrifugation, showed a high content of gliadin/glutenin protein, as characterized by a high content of glutamic acid and proline (Table II). That analysis agrees with the present work on manually dissected endosperm (Table I).

Amino acid data (Tkachuk and Irvine 1969) on manually dissected wheat germ (Table II) agreed with our data (Table I) in which germ was shown to be rich in basic amino acids and in aspartic acid and low in glutamic acid and proline.

Whole Wheat and Milled Wheat Fractions. The decortication experiment (Jensen et al 1982) was performed to produce a series of decorticated fractions (with successive variation in botanical components) from pericarp, germ, testa, and aleurone to endosperm. This morphological sequence is reflected in the decortication fractions by differences in the relative proportions of individual amino acids (Table III). The content of aspartic acid in the decortication fractions, for example, decreased significantly from fraction 1 to fraction 10, with an even lower content in decorticated wheat. A similar tendency was observed for other amino acids (Table III).

A qualitative distribution of the botanical constituents in the decortication fractions is illustrated two-dimensionally in Figs. 1 and 2, in which the arginine content (indicative of aleurone) is plotted against aspartic acid (indicative of pericarp and germ) and against glutamic acid content (indicative of endosperm). In Fig. 1, the composition of fraction 1 is shown to be very close to that of pericarp, whereas the composition of fractions 2 and 3 approaches that of germ and aleurone. The remaining decortication fractions have compositions such that they successively approach the composition of endosperm. The position of whole wheat implies that its composition is close to that of fractions 9 and 10; the composition of decorticated wheat places it close to endosperm. A similar tendency is shown in Fig. 2, in which the arginine content is related to the glutamic acid content. In this case, however, the position of fraction 1 indicates that this decortication fraction (mainly pericarp) contains some endosperm impurities. The remaining fractions asymptotically approach a tangent between aleurone-germ and endosperm. This indicates a delay in the removal of aleurone-germ by the decortication process.

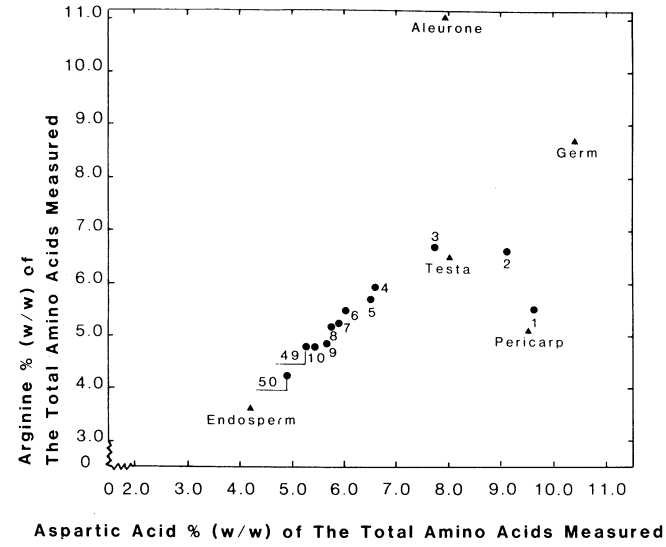

Fig. 1. Aspartic acid and arginine in manually dissected wheat fractions (▲), decortication fractions (1–10), whole wheat (49), and decorticated wheat (50).

TABLE III
Amino Acid Composition of Wheat, Decorticated Wheat Fractions, and Decorticated Wheat

		Decorticated Wheat Fractions										Decorticated		
	Wheat	1	2	3	4	5	6	7	8	9	10	Wheat	Mean	CV[a]
Aspartic acid	5.4	9.6	9.1[b]	7.7[b]	6.6	6.5	6.0	5.9	5.8	5.7	5.4	4.9	6.6	3.21
Threonine	2.7	3.8	3.3	3.0	2.8	2.7	2.9	2.5	2.5	2.4	2.4	2.5	2.8	2.16
Serine	3.5	3.5[c]	3.0[c]	3.1	3.5	3.0	3.8[c]	2.8	2.7	2.7	2.7[c]	3.4	3.1	3.92
Glutamic acid	32.5	18.4	21.2	25.0	28.6	28.9	29.9	31.0	31.2	32.3	33.1	34.0	28.8	1.88
Proline	10.8	6.5	6.9	7.9	9.3	9.7	10.1	10.1	10.3	10.5	10.8	11.7	9.6	1.27
Glycine	4.4	6.5	6.7	5.9[c]	5.1[b]	5.1	4.8	4.7[b]	4.7	4.5	4.3	3.9	5.1	3.16
Alanine	3.8	7.0	5.9	5.0	4.4	4.3	4.2	4.0	3.9	3.8	3.6	3.3	4.4	2.00
Valine	4.5[c]	5.3[c]	5.4	5.2	4.8	4.8	4.7	4.6	4.7	4.5	4.4	4.3	4.8	1.38
Methionine	1.9	2.0	1.8	1.8[c]	1.7	1.7	1.7	1.9	1.8	1.8	1.9	1.8	1.8	2.21
Isoleucine	4.2	5.0	4.5	4.2	4.1	4.1[c]	4.0	4.2	4.1[c]	4.3	4.2	4.1	4.3	1.26
Leucine	7.6	9.4[c]	7.9	7.6	7.3	7.4	7.2	7.4	7.4	7.5	7.5	7.4	7.6	2.01
Tyrosine	3.3	3.2	3.0	3.2	3.2	3.1	3.3	3.3	3.2	3.4	3.4	3.3	3.2	3.26
Phenylalanine	5.3	5.6	5.2	5.1	4.9	5.0	4.8	5.2	5.2[c]	5.2	5.2	5.4	5.2	1.89
Histidine	1.2	2.4[c]	2.9	2.9	2.7	2.7[c]	2.6	2.5	2.5	2.4	2.4	2.1	2.4	3.01
Lysine	2.9	4.9	4.9	4.2	3.5	3.6	3.3	3.3	3.3	2.9[c]	2.9	2.4	3.5	4.67
Arginine	4.8	5.5	6.6	6.7[c]	5.9	5.7	5.5	5.2	5.2	4.8	4.8	4.2	5.4	1.68
Tryptophan	1.1	1.4	1.8	1.7	1.5	1.7	1.4	1.4	1.3	1.3	1.2	1.2	1.4	...
Sum	99.9	100.0	100.1	100.2	99.9	100.0	100.2	100.0	99.8	100.0	100.2	99.9		
Percent protein[d]	12.0	6.8	9.7	14.2	14.9	15.4	14.9	13.7	13.1	12.6	12.0	10.9		
f	11.35	6.21	8.75	13.20	14.52	15.73	15.23	13.88	13.74	12.91	12.05	9.73		

[a] Coefficient of variation; standard deviation between parallels in percent of mean value.
[b] $CV_{ik} > 3 \times \overline{CV}$.
[c] $CV_{ik} > 2 \times \overline{CV}$.
[d] Dry basis.

In Fig. 3, a similar delineation (arginine vs glutamic acid) was performed for the sieving fractions. Four of the decortication fractions are illustrated in which fraction 1 is separated to 11 and 12, fraction 2 is separated to 13, 14, 15, and 16, fraction 4 is separated to 21, 22, 23, and 24, and fraction 7 is separated to 33, 34, 35, and 36. The results clearly indicate that the sieving fractions greater than 125 μ (12, 16, 24, and 36) contain considerable amounts of pericarp and germ proteins and minor amounts of aleurone proteins, whereas the fractions between 32 and 63 μ (14, 22, and 34) seem to have the highest content of endosperm proteins.

The amino acid distribution of the milled wheat fractions contains considerable information regarding the distribution of the botanical components. This information is used in the multivariable model to quantify the botanical components.

Botanical Composition of Milled Wheat Fractions

The linear relationship between the botanical composition (pericarp, aleurone, and endosperm), estimated by the amino acid pattern and by the fluorescence method (Jensen et al 1982), is illustrated in Figs. 4, 5, and 6 for the 50 wheat milling fractions. For pericarp (Fig. 4), the correlation coefficient was 0.96, indicating agreement between the two independent determinations. Generally, the pericarp content estimated by the amino acid pattern was slightly overestimated compared to results by the fluorescence method. Closer inspection of the individual residuals (Fig. 4) showed that the overestimation was due to positive deviations observed in some of the sieving fractions greater than 125 μ (12, 24, 28, 32, 36, and 40). The correlation coefficient for endosperm (Fig. 6) was also high, 0.95, demonstrating that the two methods are in accordance with one another. Large negative residuals, however, were observed in the same sieving fractions as for pericarp, except for fraction 24. Considering the aleurone content (Fig. 5), the correlation coefficient was significantly lower, 0.76 in this case, and large residuals were observed for fractions 25, 37, 42, 43, and 44. This indicates a lack of fit between the two methods because the deviations were independent with regard to type of fraction.

The differences between the two methods of estimating botanical components may result from inadequate modeling in one or both of the methods. To elucidate the results, correlations were performed between chemical constituents (ash, fiber, and starch, previously published [Jensen et al 1982], and niacin), and botanical components estimated by both methods (Table IV). Niacin is a more specific indicator of aleurone than ash (Heathcote et al 1952). The correlation coefficients for aleurone estimated by the amino acid pattern decreased significantly for ash as well as for fiber and starch compared to the corresponding results obtained by the fluorescence method. In contrast, those of paricarp and endosperm are almost identical, showing high positive correlation between pericarp and fiber, as well as high positive correlation between endosperm and starch.

Considering the correlation between niacin and aleurone, a correlation coefficient of 0.828 was observed for aleurone estimated by the amino acid model, whereas the same correlation for the fluorescence model was relatively high, 0.927. These results indicate that the fluorescence model is the most reliable, because 82% of the niacin in the wheat kernel is concentrated in the aleurone layer (Heathcote et al 1952). Closer inspection of the individual residuals between pericarp and endosperm estimated by the amino acid model and fiber and starch revealed systematic deviations for the sieving fractions greater than 125 μ. This indicates that the amino acid model yielded erroneous results for these fractions. The correlation coefficients for germ, only estimated by the amino acid pattern, yielded positive correlation with ash and fiber and negative correlation with starch (Table IV). The correlation between niacin and germ is relatively high because the abrasive decortication equipment separated both germ and aleurone from the endosperm.

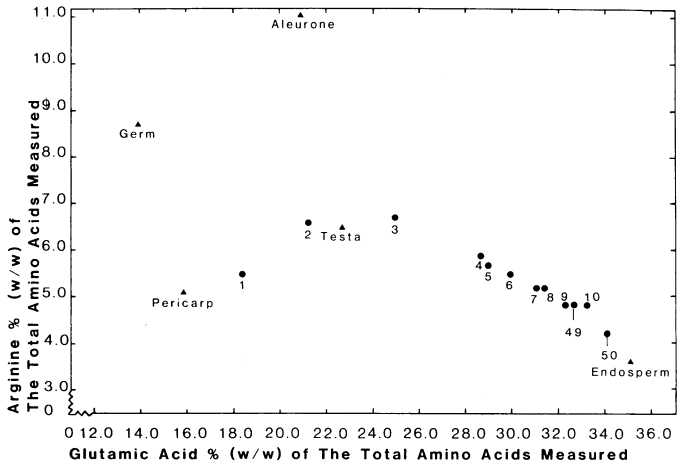

Fig. 2. Glutamic acid and arginine in manually dissected wheat fractions (▲), decortication fractions (1–10), whole wheat (49), and decorticated wheat (50).

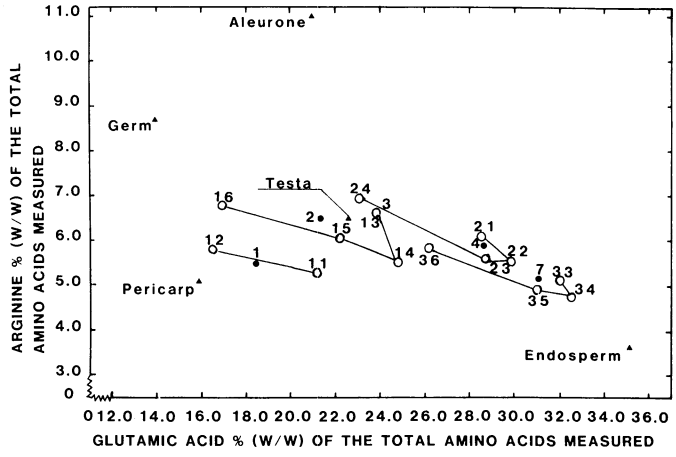

Fig. 3. Glutamic acid and arginine in manually dissected wheat fractions (▲), decortication fractions (1, 2, 4, and 7), and corresponding sieving fractions (o). Lines connect decortication fractions with their derived sieving fractions.

TABLE IV
Correlation Coefficients Between Botanical and Chemical Constituents (N = 50)

	Botanical Constituents						
	Estimated by Fluorescence Method[a]			Estimated by Amino Acid Pattern			
	Pericarp	Aleurone	Endosperm	Pericarp	Aleurone	Endosperm	Germ
Ash	0.370	0.891	−0.541	0.427	0.779	−0.561	0.561
Fiber	0.974	0.468	−0.966	0.974	0.270	−0.944	0.422
Starch	−0.942	−0.741	0.978	−0.937	−0.373	0.968	−0.371
Niacin[b]	0.420	0.927	−0.402	0.280	0.828	−0.398	0.567

[a] Jensen et al 1982.
[b] Measured in the main decortication fractions (N = 10).

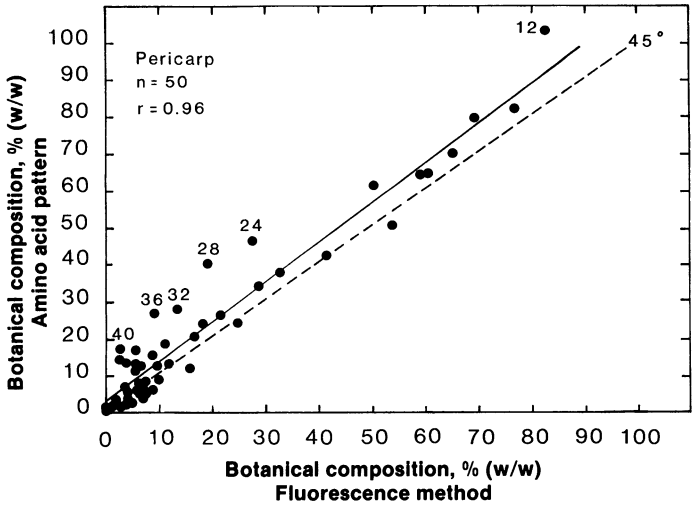

Fig. 4. The relationship between pericarp estimated by the fluorescence method and by the amino acid pattern.

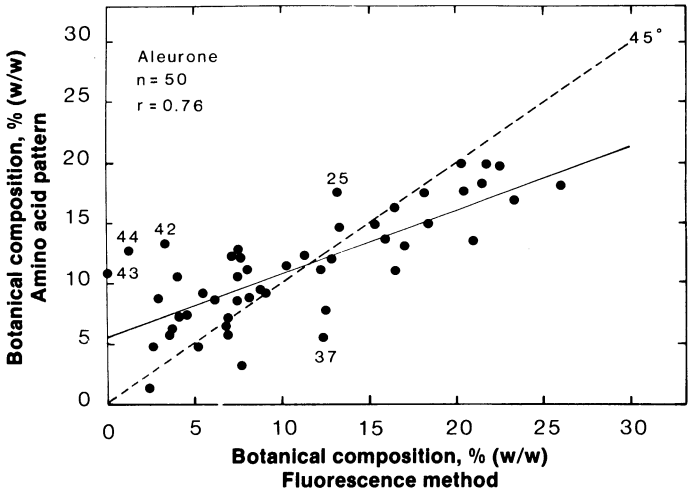

Fig. 5. The relationship between aleurone estimated by the fluorescence method and by the amino acid pattern.

The apparent problem with the amino acid model may be explained by uneven protein distributions within the individual botanical components. Kent and Evers (1969) and McDermott and Pace (1960) reported significant variation in amino acid composition within the endosperm of wheat. In our study, the manually dissected endosperm fraction was considered homogeneous, although endosperm may be subdivided into subaleurone vs inner endosperm or into vitreous and mealy parts. Subdivision of these constituents of the starchy endosperm during milling will thus contribute to erroneous results. Similarly, a subdivision of pericarp into outer and inner pericarp and of whole aleurone cells into cell walls and cell contents will contribute to erroneous results. The influence from the latter factor is confirmed by the amino acid results obtained by Bacic and Stone (1981) who used wheat aleurone cell wall and intercellular contents isolated by air-classification and differential centrifugation in a nonaqueous medium. In this study, significant deviations were observed between these subfractions of the aleurone layer regarding contents of lysine, arginine, glutamic acid, and proline.

Generally, the amino acid model agrees well with the fluorescence model. The differences observed, particularly for aleurone composition, appear to be inadequacies in the amino acid model rather than in the fluorescence model. It is, therefore, feasible to develop the application of the fluorescence method for practical use.

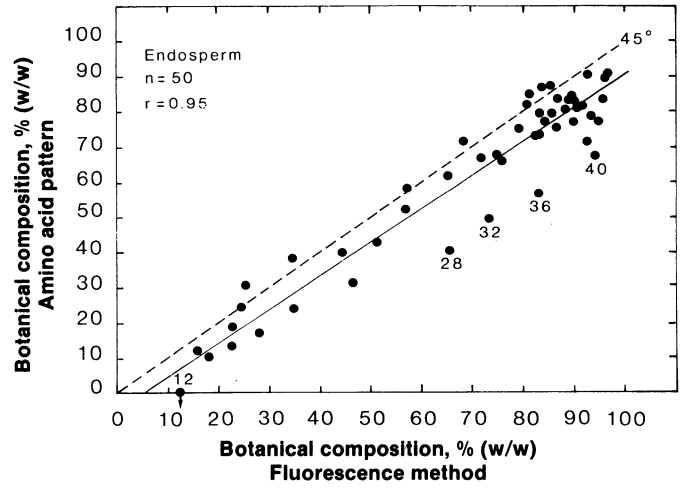

Fig. 6. The relationship between endosperm estimated by the fluorescence method and by the amino acid pattern.

ACKNOWLEDGMENTS

We wish to thank L. Munck and J. Mundy for critical review of the manuscript. Part of the work of Sv. A. Jensen was supported as a research project by the Danish Academy of Technical Sciences under the direction of O. B. Jørgensen (Dept. of Technical Biochemistry, The Technical University of Denmark) and L. Munck. Part of the work of H. Martens was financed by a grant from the Norwegian Society for Agriculture Food Research.

LITERATURE CITED

AMERICAN ASSOCIATION OF CEREAL CHEMISTS. 1979. Approved Methods of the AACC. Method 86-51, approved April 4, 1968. The Association, St. Paul, MN.

BACIC, A., and STONE, B. A. 1981. Chemistry and organization of aleurone cell wall components from wheat and barley. Aust. J. Plant Physiol. 8:475.

EGGUM, B. O. 1968. Determination of tryptophan. Acta Agric. Scand. 18:127.

FULCHER, R. G., O'BRIEN, T. P., and SIMMONDS, D. H. 1972. Localization of arginine-rich proteins in mature seeds of some members of the gramineae. Aust. J. Biol. Sci. 25:487.

HEATHCOTE, J. G., HINTON, J. J. C., and SHAW, B. 1952. The distribution of nicotinic acid in wheat and maize. R. Soc. London B. 139:276.

HEPBURN, F. N., CALHOUN, W. K., and BRADLEY, W. B. 1960. The distribution of the amino acids of wheat in commercial mill products. Cereal Chem. 37:749.

HINTON, J. J. 1947. The distribution of vitamin B_1 and nitrogen in the wheat grain. Proc. R. Soc. London B. 134:418.

JENSEN, S. Aa., MUNCK, L., and MARTENS, H. 1982. The botanical constituents of wheat and wheat milling fractions. I. Quantification by autofluorescence. Cereal Chem. 59:477.

JONASSEN, I. 1980. Characteristics of hiproly barley. I. Isolation and characterization of two water-soluble proteins. Carlsberg Res. Commun. 45:47.

KENT, N. L., and EVERS, A. D. 1969. Variation in protein composition within the endosperm of hard wheat. Cereal Chem. 46:293.

KJELDAHL, J. T. 1883. En ny metode til kvaelstofbestemmelse i organiske stoffer. (In Danish.) Medd. Carlsberg Lab. II:1.

KOHLER, G. O., and PALTER, R. 1967. Studies on methods for amino acid analysis of wheat products. Cereal Chem. 44:512.

LINDQUIST, B., OSTGREN, J., and LINDBERG, I. 1975. A method for the identification and quantitative investigation of denatured proteins in mixtures based on computer comparison of amino acid patterns. Z. Lebensm. Unters. Forsch. 159:15.

MARTENS, H. 1979. Factor analysis of chemical mixtures. Anal. Chim.

Acta 112:423.
MARTENS, H., and BACH KNUDSEN, K. E. 1980. Fractioning barley protein by computer factor analysis. Cereal Chem. 57:97.
MARTENS, H., and LEA, P. 1981. Bestemmelse av Fremmedprotein i Kjattvarer ved Statistisk Analyse av Aminosyre-Data (in Norwegian). NINF Report 51601, Norwegian Food Res. Inst., Oslo, Norway.
McDERMOTT, E. E., and PACE, J. 1960. Comparison of the amino acid composition of the protein in flour and endosperm from different types of wheat, with particular reference to variation in lysine content. J. Sci. Food Agric. 11:109.
TKACHUK, R., and IRVINE, G. N. 1969. Amino acid composition of cereals and oilseed meals. Cereal Chem. 46:206.

[Received May 18, 1982. Accepted October 8, 1982]