PROTEIN AND AMINO ACID COMPOSITIONS OF DRY-MILLED AND AIR-CLASSIFIED FRACTIONS OF TRITICALE GRAIN

A. C. STRINGFELLOW, J. S. WALL, G. L. DONALDSON, and R. A. ANDERSON, Northern Regional Research Laboratory[1], Peoria, IL 61604

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Protein contents and amino acid compositions were determined on fractions obtained by roller milling, fine grinding, and air classifying one spring and two winter triticale varieties. Because the triticale grains have protein levels equivalent to the hard wheats but higher lysine contents, the grain fractions were explored as sources of high-protein flours suitable for use as supplements in cereal products. The three triticale grains were first processed in a laboratory roller mill through three break and three reduction steps to give a total flour extraction of 62–65%. Variations in the protein and lysine contents of the flour milling fractions reflected differences in the distribution of proteins in the endosperm. Bran fraction and shorts were high in protein and lysine due to their germ and aleurone content. The triticales, especially the Texas-grown spring variety, behaved like soft wheats upon fine grinding and air classification. Yields of the high-protein fractions and their protein levels were generally superior to those of hard wheats. Low levels of coarse residues from the reground flour remained after seven steps of air classification. The high-protein fractions of triticale flours have lysine contents comparable to that of the initial flour.

Triticale has generated considerable interest because it has potential for better yield in some areas and for higher protein content and nutritional value than many wheats. Triticale is a man-made species produced by crossing wheat (*Triticum*) and rye (*Secale*) in a manner that combines the genetic complements of both grains. Current success in triticale stems from hexaploid lines produced by breeding of crosses of durum wheat and rye selections at the University of Manitoba (1). Further improvements in triticale are the day-length insensitive and highly fertile lines developed by Zillinsky and Borlaug (2) in Mexico. Triticales have been crossed with hexaploid bread wheats to help overcome problems of winter hardiness, lodging, and disease susceptibility.

Triticale has been milled to yield flour which was tested in baking applications. Unrau and Jenkins (3) reported that mixing strength and loaf volumes of doughs prepared from the flour were generally lower than those from bread wheat flours. Satisfactory baking results were obtained when some triticale flours were blended with flours from strong wheats. Lorenz *et al.* (4) reported that adjusting mixing time and absorption caused some triticale flours to yield breads of acceptable quality. Flours from winter triticales produced better loaves than those from spring triticales. The grains of the spring and winter triticales differed also in appearance and hardness.

Villegas *et al.* (5) observed that the protein and lysine contents of triticales are generally higher than those of wheat, possibly because of the rye parent. Chen and Bushuk (6) demonstrated that the proteins of triticale consist of components similar to those of both wheat and rye. Results of feeding experiments with

[1]Agricultural Research Service, U.S. Department of Agriculture, Peoria, IL 61604.
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triticale generally confirm its nutritionally better protein compared to wheat (7,8).

In response to current demand for additional sources of vegetable protein of good nutrient quality, we explored the possibility of producing protein concentrates from triticale by dry-milling and air-classifying methods. A preliminary study by Anderson *et al.* (9) revealed that a spring triticale variety responded well to fine grinding and air classification and yielded fractions containing an increased concentration of flour protein. We studied both spring and winter triticales that differed in milling response. Yields, obtained by milling and air-classifying methods, proximate analyses, and amino acid compositions of wheat and flour fractions are presented.

MATERIALS AND METHODS

Triticale Grain

Three triticale varieties were supplied by Farm Management Services, Inc., Wichita, Kans. Fas Gro 204 is a spring hexaploid grown in Texas, whereas Fas Gro 131 and Fas Gro 385 are winter hexaploids grown in Kansas. Two lots of Fas Gro 131 that differed slightly in composition and were designated Fas Gro 131A and Fas Gro 131B were milled. Varieties 204 and 385 were also used by Lorenz *et al.* (4) in their baking studies.

Milling and Fractionating

Grain of 12.5% moisture content was tempered at 76° F overnight to 14.5% moisture and then raised to 15.5% 30 min before milling. Milling was conducted on a Buhler pneumatic laboratory mill, type MLV-202. Roll settings and sieves used were those recommended by the manufacturer for hard wheats. Flour was collected from three break stages and three reduction stages and combined to yield a straight-grade flour.

The straight-grade flour was further ground by passing it three times through an Alpine Kolloplex pin mill (Model 160-Z) operating at 14,000 rpm, conditions we normally use for hard wheat flour.

Air classification of this finely ground flour was carried out in a Pillsbury laboratory model classifier. Flour was separated into eight fractions by collecting a fine fraction, then readjusting the classifier for a coarser cut and reclassifying the coarse fraction until seven fine fractions and one coarse fraction remained. Classifier cut points were set to yield fractions with particle sizes (reported in terms of mass median diameter; *i.e.*, diameter at which 50% of weight of sample is undersize) of about 11, 12, 15, 18, 21, 23, and 25 μ. Because of the large number of fractions taken and the intensive flour regrinding imposed, separations are near maximum for this technique (10).

Analytical Procedures

Proximate analyses were made according to procedures described in the Approved Methods of the AACC (11). Per cent protein was calculated as 5.7 $\times$ per cent nitrogen.

For amino acid analysis, samples of the grain or flour were hydrolyzed by refluxing in 6*N* HCl for 24 hr. Sample size was chosen to contain 1.0 mg of nitrogen and the volume of acid used for hydrolysis was 2 ml/mg of sample. A

Beckman Spinco Model 121 amino acid analyzer was used to analyze the hydrolysates. Chromatographic peaks were integrated by an Infotronics CRS-210 unit and amino acid values calculated on an IBM 1130 computer according to the program of Cavins and Friedman (12).

RESULTS AND DISCUSSION

Composition of Grains

The spring variety of triticale, Fas Gro 204, was higher in protein than either of the winter varieties, 131 or 385 (Table I). While the protein content of Fas Gro 204 is comparable to that of most hard red spring wheats, Fas Gro 131 and 385 are similar to rye and hard red winter wheats. Fas Gro 204 was also higher in crude fiber than the other triticales. Fat content varied slightly among the three varieties. The lower nitrogen-free extract content of Fas Gro 204 may be related to its softer wrinkled structure compared to the plumper and harder kernels of the winter varieties.

Yields and Compositions of Milling Fractions

Overall extraction of straight-grade flour was similar for all three varieties when milled in a Buhler mill (Table II). Yield of break flour was higher for varieties 131 and 204 than for 385. In all three varieties, average protein content of the break flours was less than that of the reduction flours. Protein content increased in flours from successive break and reduction steps. Although the increase was more pronounced in the triticales than in hard wheats (13), it was similar to results obtained on milling soft wheats (13). Both 3rd break and 3rd reduction flours contained more ash than the other milling fractions.

Yields of the straight-grade flour (62–65%) were lower than those for hard red winter wheats (68–70%). Poor release of flour from shorts and bran when milling triticale was also noted by Unrau and Jenkins (3) who remilled shorts and bran to increase yields of flour. Lorenz *et al.* (4) obtained low yields of flours (33–45%) from triticale when they used a Brabender Quadrumat Junior mill. The low protein contents of their flours undoubtedly resulted from the limited extractions, a conclusion supported by our observed differences in protein contents of successive break and reduction fractions.

TABLE I
Composition of Triticales[a]

	Triticale Variety			
	Fas Gro	Fas Gro 131		Fas Gro
Component	204	A	B	385
Crude protein (N × 5.7)	14.7	13.2	12.0	12.0
Crude fat	1.5	1.5	1.6	1.3
Crude fiber	2.7	2.1	2.1	2.1
Ash	1.8	1.8	1.7	1.7
Nitrogen-free extract	65.3	67.4	68.6	69.0

[a]14% moisture basis.

TABLE II
Yields of Fractions from Triticale Varieties Obtained with Buhler Mill

Fraction	Yield % of Recovered Products			Protein, % (14% Moisture)			Ash, % (14% Moisture)		
	Fas Gro 204	Fas Gro 131A[a]	Fas Gro 385	Fas Gro 204	Fas Gro 131A	Fas Gro 385	Fas Gro 204	Fas Gro 131A	Fas Gro 385
Break flour									
1st	3.8	4.0	2.4	8.6	7.5	7.5	0.54	0.54	0.56
2nd	5.2	5.9	4.9	10.6	9.5	9.2	0.56	0.62	0.59
3rd	1.6	1.8	1.7	13.1	11.3	10.7	0.72	0.81	0.79
Reduction flour									
1st	27.8	22.2	25.8	11.6	10.4	9.6	0.51	0.48	0.48
2nd	18.0	20.1	22.5	13.4	12.0	11.3	0.52	0.48	0.46
3rd	7.4	7.9	7.6	14.8	13.6	11.9	0.64	0.65	0.66
Straight-grade flour	63.8	61.9	64.9	12.2	11.1	10.4	0.54	0.52	0.52
Shorts	9.9	12.4	13.5	17.4	15.7	14.9	2.45	2.52	2.45
Bran	26.3	25.7	21.5	19.7	17.5	17.3	4.87	4.77	4.40

[a]Yield of products from Fas Gro 131B was similar to that from Fas Gro 131A.

Milling of Fas Gro 204 produced the largest bran fraction (Table II) consistent with the higher fiber content of that grain. With our milling system, germ was primarily retained with the bran fraction while the aleurone layer was directed to the shorts fraction. Lower extraction resulted in much endosperm tissue falling into the shorts. Protein contents of the bran and shorts exceeded those of the flours of all three varieties. More than 45% of the grain protein was contained in bran and shorts fractions. Among the three varieties, Fas Gro 204 gave flour and by-product fractions having the highest protein content. The difference in protein content of the whole grains (Table I) and straight-grade flours (Table II) was about two percentage units, about twice that usually found for hard wheats of similar protein contents. Poor extraction or large germ and aleurone components, or both, in triticale may account for that difference.

Amino Acid Composition of Milling Fractions

Content of lysine and of other amino acids for Fas Gro 204 and 385 varieties (Table III) was similar for both whole grains. The level of lysine in the protein of those grains was 10–25% higher than that for wheat grains of different types as summarized by Kasarda *et al.* (14).

The bran, shorts, and flour fractions differed markedly in their amino acid contents. Corresponding fractions of the two varieties were similar in amino acid composition. Bran was higher in lysine, arginine, and aspartic acid than flour, which had more glutamic acid and proline and slightly more sulfur-containing amino acids. Shorts were intermediate in composition between flour and bran. Those preparations of triticale shorts were not so rich in lysine as commercial wheat shorts, whose analyses were reported by Kasarda *et al.* (14). The lower

TABLE III
Amino Acid Composition of Triticale Grain and Buhler-Milled Fractions
(g/16 g N)

	Grain		Bran		Shorts		Flour	
Amino Acid	**Fas Gro 204**	**Fas Gro 385**	**Fas Gro 204**	**Fas Grow 385**	**Fas Gro 204**	**Fas Gro 385**	**Fas Gro 204**	**Fas Gro 385**
Lysine	3.4	3.4	4.4	4.4	3.6	3.7	2.5	2.4
Histidine	2.3	2.4	2.9	2.7	2.4	2.4	2.2	2.2
Ammonia	3.8	3.7	2.8	2.9	3.0	3.3	3.6	3.6
Arginine	5.6	5.6	8.4	7.4	6.6	6.3	3.9	4.0
Aspartic acid	6.5	5.9	8.0	5.9	6.7	6.7	4.9	5.1
Threonine	2.9	3.1	3.5	3.0	3.2	3.2	2.8	2.9
Serine	4.7	4.5	4.6	4.5	4.4	4.5	4.7	4.7
Glutamic acid	31.9	28.5	21.2	21.3	23.7	25.1	33.3	34.9
Proline	9.5	9.6	9.6	9.6	7.9	8.6	11.5	11.5
Glycine	4.3	4.2	5.4	4.2	4.4	4.6	3.5	3.6
Alanine	3.9	3.9	5.0	3.8	4.3	4.3	3.2	3.3
Half-cystine	1.3	1.3	1.2	1.3	1.2	1.2	1.4	1.7
Valine	4.5	4.6	4.8	4.6	4.7	4.9	4.4	4.4
Methionine	1.5	1.6	1.5	1.1	1.8	1.8	1.4	2.0
Isoleucine	3.5	3.5	3.3	3.5	3.4	3.6	3.5	3.5
Leucine	6.9	6.2	6.2	6.3	6.2	6.3	7.0	6.4
Tyrosine	3.0	3.1	3.2	3.1	3.0	3.1	3.0	3.5
Phenylalanine	4.6	4.8	4.4	4.6	4.4	4.6	4.9	5.2

lysine content of triticale shorts probably was attributable to our poor flour extractions and the contamination of shorts with other endosperm material. However, since both the shorts and bran contained high protein levels and good amounts of essential amino acids, those fractions may be nutritionally beneficial in foods and feeds. Use of triticale mill feeds as a source of protein concentrate has been explored (15).

The difference in amounts of lysine in whole grain and flour was greater for triticale than for wheat as listed by Kasarda *et al.* (14). That observation provides further evidence that germ and aleurone may contribute more to the grain protein in triticale than in wheat.

Break and reduction flours of Fas Gro 385 triticale differed slightly in their amino acid content (Table IV). The break flour, being derived from the floury part of the kernel, may have a lower proportion of water-insoluble protein and thus a slightly higher average lysine value. There was a small successive decrease in lysine contents of the break flours proceeding from 1st to 3rd break stages (Table IV). The 2nd reduction flour had the lowest lysine content and highest glutamic acid level. It probably had the least amount of floury endosperm or aleurone impurity. On the basis of protein levels and amino acid contents, the separate use of roller-milling fractions of triticale flour offers no significant economic or nutritional advantages.

Yields and Composition of Air-Classified Fractions

The finely ground flours from the three triticale varieties followed air-classification patterns similar to those of various wheats; namely, fractions 1–3 with particle sizes below 15 μ had much higher protein contents than those of the

TABLE IV
Amino Acid Composition of Break and Reduction Flours of Fas Gro 385 Triticale (g/16 g N)

	Break Flours			Reduction Flours		
Amino Acid	**1st**	**2nd**	**3rd**	**1st**	**2nd**	**3rd**
Lysine	2.8	2.6	2.4	2.4	2.3	2.5
Histidine	2.1	2.1	2.1	2.0	2.1	2.0
Ammonia	3.7	3.8	4.1	4.0	3.9	3.6
Arginine	4.3	4.2	4.2	4.0	3.9	4.2
Aspartic acid	5.3	4.9	4.7	4.6	4.6	5.0
Threonine	2.9	2.8	2.7	2.7	2.8	2.9
Serine	4.6	4.5	4.4	4.5	4.5	4.6
Glutamic acid	30.9	32.0	31.7	32.8	33.6	32.9
Proline	11.0	11.6	11.4	11.4	11.7	11.6
Glycine	3.6	3.5	3.4	3.4	3.4	3.7
Alanine	3.9	3.3	3.2	3.1	3.1	3.4
Half-cystine	1.7	1.7	1.6	2.0	1.7	1.8
Valine	4.4	4.3	4.1	4.2	4.2	4.4
Methionine	1.6	1.6	1.6	1.6	1.6	1.6
Isoleucine	3.6	3.6	3.4	3.5	3.6	3.7
Leucine	6.4	6.3	6.1	6.3	6.3	6.4
Tyrosine	3.3	3.2	3.0	3.2	3.2	3.2
Phenylalanine	4.9	5.0	4.8	5.1	5.2	5.1

original flours (Table V). The highest protein content was in fraction 1 of triticale flour 204 (34.3%). The finer fractions also contained high fat and ash analyses that appeared to parallel the high protein contents.

The starchy or low-protein fractions 4 through 7 varied in quantity from 12 to 23% and in protein content from 4.4 to 6.8%.

The coarse residues are more like the parent flours in composition than any of the corresponding fractions (Table V).

In Table VI, fractionation results obtained from air-classifying the three triticale flours are compared to those of a rye and two different types of wheat.

Maximum range of protein contents among the eight classified fractions was from Fas Gro 204, 4.4 to 34.3%. The greater ease of fractionation is also reflected in the highly significant protein shift of 73% in 204 flour as compared to 56 to 58% from other triticale flours, 40% for rye flour, and 29 to 50% for the two hard wheats. Fas Gro 204 behaved much like the soft wheat that had a high protein shift (82%). Total protein shift is the sum of the protein shifted into the high-protein fractions and out of the low-protein fractions, expressed as percentage of the total protein present in the flour, as described by Gracza (10).

Apparently Fas Gro 204 is less vitreous than the other two triticale varieties and responds more readily to fine grinding and air classification. It gave a 25% yield of the three high-protein fractions—slightly less than that of soft wheat flour (29%), but equal to that of rye flour (26%), and somewhat greater than hard wheat flours (19–21%). Protein content of the three finest fractions (29%) from

TABLE V
Yield and Composition of Fractions Produced by Fine Grinding and Air Classification of Triticale Flours (14% Moisture basis)

Kind of Flour and Fraction Number	Yield %	Protein %	Ash %	Fat %
Fas Gro 204				
Straight-grade flour	100.0	12.7	0.57	0.69
Fraction 1	10.7	34.3	1.2	1.6
Fraction 2	8.0	28.4	0.9	1.1
Fraction 3	6.4	20.7	0.7	0.9
Fraction 4	22.8	6.7	0.5	0.4
Fraction 5	14.0	4.8	0.5	0.3
Fraction 6	16.8	4.4	0.4	0.3
Fraction 7	11.7	5.6	0.5	0.3
Fraction 8 (coarse residue)	9.6	13.3	0.5	0.4
Fas Gro 385				
Straight-grade flour	100.0	10.4	0.58	0.5
Fraction 1	7.1	30.2	1.7	1.6
Fraction 2	6.0	24.3	1.2	1.2
Fraction 3	5.4	17.4	0.8	1.0
Fraction 4	21.2	6.5	0.4	0.5
Fraction 5	13.6	5.2	0.3	0.4
Fraction 6	16.8	5.1	0.3	0.4
Fraction 7	12.6	6.8	0.3	0.4
Fraction 8 (coarse residue)	17.3	12.4	0.4	0.5

TABLE VI
Comparison of Fractionation Responses of Reground Flours from Triticale, Rye, and Wheat
(Expressed as per cent on 14% moisture basis)

	Variety Fas Gro				Wheat		
Sample	**204**	**131B**	**385**	**Rye[a], Commercial Mix**	**Hard red spring Selkirk[b]**	**Hard red winter Wichita[c]**	**Soft red winter Vermillion[d]**
Straight-grade flour, protein	12.7	10.2	10.4	10.9	12.3	10.9	9.4
Maximum range of protein	34.3–4.4	27.6–4.7	30.2–5.1	21.5–6.2	23.7–7.6	29.4–5.5	26.7–2.3
Combined high-protein fractions 1–3							
Yield	25.1	21.7	18.5	25.8	18.8	21.0	29.4
Protein	29.0	22.1	24.5	19.8	19.4	24.4	21.4
Combined starchy fractions 4–7							
Yield	65.3	64.6	64.2	56.8	49.5	52.9	64.5
Protein	5.5	5.6	5.9	7.4	8.7	6.5	3.3
Coarse residue fraction 8							
Yield	9.6	13.7	17.3	17.4	31.7	26.1	6.1
Protein	13.3	12.8	12.4	9.6	14.2	9.8	6.2
Protein shifted, total	73	58	56	41	29	50	82

[a]Unpublished data.
[b]Peplinski *et al.* (16).
[c]Stringfellow and Peplinski (17).
[d]Peplinski *et al.* (18).

204 was considerably more than either the rye fractions (20%) or the hard and soft wheat fractions (19–24%). Yields of the three high-protein fractions for the other two triticale flours, 131 and 385, were similar to the hard wheat flours. Also, protein contents of fractions 1 to 3 from those two triticales were comparable to those of the same fractions from hard wheats.

Yields of the combined starchy fractions 4–7 from the three triticale flours were about the same as similar fractions from soft wheat but higher than those from rye and hard wheat. Protein contents of triticale fractions 4–7 were lower than those from either rye or hard wheats and higher than those from comparable soft wheat fractions.

Yields of the coarse residues remaining after fractionation of the three triticale flours ranged from 9.6 to 17.3% and protein contents from 12.4 to 13.3%. The small amounts of coarse residues point to ease of fractionating triticale flours compared to hard wheats. Fas Gro 204, with only 9.6% coarse residue, responded better to air classification than either 131 or 385, but all three flours had a much better response than usual for hard wheats.

In general, fractionation results indicate that triticales behave much like soft wheat flours when finely ground and air classified, but they contain more protein in their fractions than in those from soft wheat flours. Smaller amounts of coarse residue from the triticales, and high yields of starchy fractions, also reflect the similarity to soft wheats. These characteristics of triticale make it more suitable for air classification.

Amino Acid Contents of Air-Classified Fractions

No major variations occurred in the amino acid composition of fractions from Fas Gro 385 flour (Table VII). Concentration of protein into the finer fractions does not reduce the nutritional quality of the triticale flour protein in those fractions. Thus, air classification does not selectively concentrate specific protein classes in different fractions.

The better nutritional quality of triticale protein concentrates compared to high-protein wheat fractions (2.5 *vs.* 1.5 g lysine per 16 g nitrogen) suggest use of such fractions in breakfast cereals, snack foods, and pancake flours where improvement in protein level and quality is desirable. The low-protein fractions

TABLE VII
Some Essential Amino Acids in Air-Classified Fractions of Fas Gro 385 Triticale Flour

	g Amino Acid/16 g Nitrogen								
	Parent	Fractions							
Amino Acid	Flour	1	2	3	4	5	6	7	8
Lysine	2.5	2.7	2.5	2.4	2.6	2.9	2.7	2.7	2.5
Threonine	2.9	3.0	3.0	2.9	3.1	3.0	2.9	2.9	3.0
Valine	4.6	4.8	4.5	4.5	4.6	4.6	4.7	4.7	4.7
Methionine	2.0	1.8	1.9	1.8	2.0	1.9	1.8	1.8	1.7
Isoleucine	3.7	3.7	3.6	3.7	3.9	3.8	3.9	3.9	4.0
Leucine	6.5	6.8	6.6	6.6	6.6	6.8	6.7	6.9	7.0
Phenylalanine	5.4	5.4	5.5	5.4	5.4	5.4	5.3	5.7	5.7

should have applications in many industrial flour uses such as adhesives or coatings.

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