## Triticale Protein Concentrate: Preparation, Composition, and Properties

Y. Victor Wu,\* Kenneth R. Sexson, and Joseph S. Wall

An alkaline extraction process gives protein concentrates and starch from ground triticale. Optimum extraction was at pH 10.8 in 0.05 N sodium hydroxide with 150 g of triticale per 900 ml of solvent. The triticale was extracted twice with sodium hydroxide solutions. After centrifugation each of two alkaline extractions was adjusted to pH 4.6 to yield a precipitate and a supernatant. Bran was removed from starch and protein by screening the second alkaline dispersion. Protein content (nitrogen  $\times$  5.7) of the concentrates varied between 82 and 87%, depending on the amount in the original grain, accounted for 53 to 59% of total triticale protein, and had from 3.2 to 3.3 g of lysine and 3.5 to 3.8 g of total sulfur amino acids per 16 g of nitrogen. Minimum nitrogen solubility of the concentrates was 8 to 9% near pH 6, and solubility was 70 to 83% at pH 2.3. All protein concentrates had good functionality relative to their hydration capacity (near 4), emulsifying activity (near 90%), and emulsion stability (around 85%).

Triticale is a cross between wheat and rye. Plant breeders are seeking to combine the quality and uniformity of wheat with the hardiness, vigor, yield capacity, and disease resistance of rye. Current commercial lines of triticale are hexaploid, containing chromosomes of a durum wheat and those of rye. Some triticale lines combine the high protein content of wheat and the high lysine content of rye. In some locations, triticale has outyielded wheat. CIMMYT (1974) reports that several lines of triticale have a lysine content equal to that of high-lysine corn. Both grain plumpness and grain density are being improved each year in Mexico; earlier problems such as excessive sensitivity to length of day, low grain fertility, late maturity, low yield, lodging, and susceptibility to certain diseases have been reduced considerably (Hulse and Spurgeon, 1974).

Under standard dough processing and baking conditions, triticale flour alone produces bread with poor loaf volume because of the low viscoelastic strength of triticale gluten. But substituting triticale flour for 30 to 40% wheat flour did not markedly decrease loaf volume and internal characteristics of breads (Unrau and Jenkins, 1964; Rooney et al., 1969). The successful production of 100% triticale breads by modifications of mixing speeds and fermentation times has been reported (Muntzing, 1966; Lorenz et al., 1972; Lorenz, 1972; Tsen et al., 1973). Baking quality varies more among triticales than among wheats. Loaf volume from 100% triticale flour doughs was improved by adding dough conditioners according to Tsen et al. (1973).

Dry milling triticale has been reported by a number of people (Lorenz, 1972; Madl and Tsen, 1973; Rooney et al., 1969; Anderson et al., 1972; Kaltsikes and Larter, 1970). Triticales produced lower yields of flour (50.5 to 64.0%) than wheat varieties (66.8 to 73.0%). Lysine content of the flour was reduced compared with that of whole grain. Air classification indicated that triticale flours behave somewhat like soft wheat flours and give a good shift of protein into high protein fractions; however, air-classified triticale fractions contain more protein (Anderson et al., 1974).

Wet milling triticale flour by a dough ball process resulted in gluten and starch in yields similar to that from soft and hard wheat flours (Anderson et al., 1974). The triticale gluten contained 70% protein, and the gluten fraction represented 81% of the protein present in the flour. However, triticale yielded a soft, weak gluten ball, and the dough could be worked only with care. The leaching of water-soluble proteins from gluten during wet processing of flour decreased lysine content to 2.3 g/16 g of nitrogen.

Anderson et al. (1974) wet milled triticale grain by a process similar to the one they used with corn. First, grain was steeped in 0.3% sulfur dioxide for 24 h at 100 °F. The steeped grain was then processed into gluten, starch, and fiber fractions. Gluten recovered has reduced elasticity compared with gluten made from triticale flour by the dough ball process.

A protein concentrate was made by wet processing bran at different pH values (Saunders et al., 1974). The yield of protein concentrate from bran was 8.3 to 21.4%, and the protein content (nitrogen  $\times$  6.25) of the concentrate was 61 to 43%. If starch was removed during processing, the concentrate had a protein content of 72 to 74% but at a considerably lower yield from 6.7 to 5.9%.

Since whole triticale has a higher protein content and a better amino acid composition than triticale flour (Anderson et al., 1974) and since no practical process has been described to make gluten or protein concentrate from whole triticale, we investigated a number of factors affecting extraction of protein concentrates from whole triticale having different protein contents.

The protein concentrate produced by our alkaline procedure not only contains much of the soluble protein of triticale, but also has a lysine content comparable to the whole grain and considerably higher than gluten made by the dough ball process (Anderson et al., 1974). Since commercial potential depends on composition and functional properties, we have determined protein, starch, fat, fiber, ash, amino acid composition, and total neutral carbohydrates of both triticale protein concentrate and by-products, as well as nitrogen solubility, hydration capacity, emulsifying activity, and emulsion stability of the concentrate only.

### MATERIALS AND METHODS

**Triticale.** The two triticale varieties used were supplied by Farm Management Services, Inc., Wichita, Kan. Fas Gro 204, a spring hexaploid grown in Texas, had a protein content of 17.7% (nitrogen  $\times$  5.7), dry basis. Fas Gro 385, a winter hexaploid also grown in Kansas, had a protein content of 15.0%, dry basis.

Northern Regional Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Peoria, Illinois 61604.

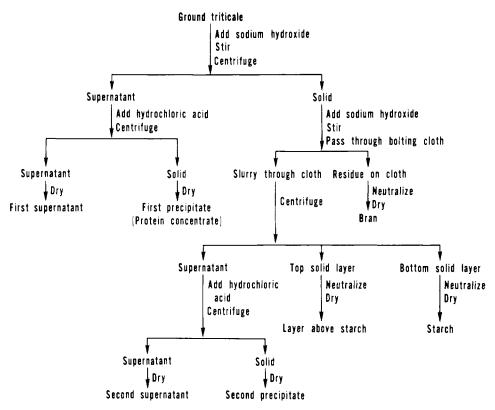


Figure 1. Schematic diagram for preparing protein concentrate and by-products from ground triticale.

Each triticale was ground twice in a hammer mill equipped with a screen containing 1/16-in. holes; 58% of the twice-ground Fas Gro 385 passed through a 100-mesh screen.

**Protein Extraction.** To test the effect of various solvents on extraction, ground triticale 204 was mixed with a solvent at a specified weight-to-volume ratio, stirred magnetically for 25 min, and then centrifuged for 10 min at 3300g in a Sorvall laboratory centrifuge. A portion of the supernatant from centrifugation was analyzed for nitrogen by a micro-Kjeldahl method, and the remaining supernatant was freeze-dried.

**Precipitation pH.** To determine the pH value where the greatest precipitation of protein occurred, an alkaline extract (7 ml) of triticale 204 was pipetted into each of six centrifuge tubes, and hydrochloric acid solution was added dropwise to each tube until pH values ranged from 4.0 to 6.5. The mixture in each tube was stirred magnetically and then centrifuged at 3300g in a Sorvall laboratory centrifuge for 15 min. A portion of each supernatant after centrifugation was analyzed for nitrogen by a micro-Kjeldahl method. Subtracting the amount of protein in the supernatant from that in the alkaline extract gave the amount of protein precipitated at each pH level.

**Protein Concentrate.** Sodium hydroxide solutions of 0.04 and 0.05 N were used to make protein concentrates and by-products at pH 10 and 10.8. Ground triticale (150 g) and 900 ml of 0.05 N sodium hydroxide were stirred for 25 min magnetically (Figure 1); the slurry pH was adjusted to 10.8 by addition of sodium hydroxide or hydrochloric acid solution, if needed. The slurry was centrifuged at 3300g in a Lourdes centrifuge for 15 min, and the supernatant was decanted and adjusted to pH 4.6 with 6 N hydrochloric acid to precipitate almost all the protein. The mixture was centrifuged at 3300g for 15 min to yield a precipitate and a supernatant, which were freeze-dried separately as the first precipitate (protein concentrate) and supernatant.

The alkaline residue from the first centrifugation was redispersed to original volume and pH by addition of water and sodium hydroxide solution (Figure 1). This slurry was stirred magnetically for 25 min and passed through 100-mesh bolting cloth to remove bran. Slurry that passed through the cloth was centrifuged at 3300g for 15 min to give a supernatant, a starch layer, and a layer above starch. The supernatant was adjusted to pH 4.6 by adding 6 N hydrochloric acid to precipitate almost all the protein. The mixture was centrifuged at 3300g for 15 min to yield a precipitate and a supernatant, which were freeze-dried separately as the second precipitate and supernatant. Starch, the layer above the starch, and bran that remained on the bolting cloth were each neutralized with 6 N hydrochloric acid and freeze-dried.

**Composition and Properties.** Protein content was calculated from duplicate micro-Kjeldahl analyses by multiplying percentage nitrogen by 5.7 and correcting to a moisture-free basis. Total neutral carbohydrates of acid-hydrolyzed samples of protein concentrate and by-products were determined by a gas-liquid chromato-graphic (GLC) procedure (Sloneker, 1971), and the cellulose fraction was analyzed by the same GLC procedure after other components were solubilized and removed (Sloneker, 1971). Fiber, ash, and hydration capacity were determined according to AACC Approved Methods (1971), and starch was determined by a polarimetric method (Garcia and Wolf, 1972). Fat was measured by a GLC procedure (Black et al., 1967) as well as by petroleum ether extraction (EE).

Samples for amino acid analyses were hydrolyzed for 24 h by refluxing in 6 N hydrochloric acid. A portion of the hydrolysate solution was analyzed in a Beckman Spinco Model 121 amino acid analyzer, and data were computed automatically (Cavins and Friedman, 1968).

Nitrogen solubility was determined by mixing 0.1 g of protein concentrate with 10 ml of water, and either sodium hydroxide or hydrochloric acid solution was added

Table I.	Extraction of	Triticale Protein <sup>a</sup>
with Vari	ous Solvents	

Solvent	pH of slurry	% of triticale protein ex- tracted	Protein in ex- tracted solids, %
Water	6.2	18	32
Hydrochloric acid <sup>b</sup>	3.5	20	34
0.1 N acetic acid	4.4	25	35
1 N acetic acid	3.4	34	46
0.03 N sodium hydroxide	9.9	53	57
0.04 N sodium hydroxide	10.5	69	63
0.05 N sodium hydroxide	10.9	79	67
0.08 N sodium hydroxide	11.6	81	63

<sup>a</sup> Fas Gro 204, a spring hexaploid triticale grown in Texas, with a protein content of 17.7% (nitrogen  $\times$  5.7). Solid-to-solvent ratio, 1:6, dry basis. <sup>b</sup> Hydrochloric acid (1 N) was added dropwise to the slurry until pH 3.5; exact normality not known.

Table II. Solid-to-Solvent Ratio Influenced Extraction of Triticale Protein $^a$ 

Solid:solvent ratio	[Solvent] (NaOH), N	Protein extracted, %
1:3	0.1	47
1:4	0.075	58
1:6	0.05	75
1:10	0.03	82

<sup>a</sup> Triticale 204; slurry pH 10.9.

dropwise to give a range of pH values from 2.2 to 10.2. The mixture was stirred magnetically for 25 min and then centrifuged at 1300g (or at 3300, 12100, or 27000g, if needed) for 20 min to separate solid and supernatant satisfactorily. The supernatant was analyzed for nitrogen by micro-Kjeldahl, and the percentage of nitrogen soluble was calculated at each pH level. Emulsifying activity and emulsion stability were determined by the method of Yasumatsu et al. (1972) for a simple system, in which only soybean oil and water were added to the protein concentrate.

#### RESULTS AND DISCUSSION

Solvent. A number of solvents were used to extract triticale 204 protein at a solid-to-solvent ratio of 1:6 (Table I). Water removed 18% of the total protein from ground triticale, and the extracted solid (the supernatant from centrifugation and then freeze-dried) contained 32% protein. Dilute hydrochloric acid at pH 3.5 extracted about the same amount of triticale protein as water, but acetic acid solubilized more triticale protein than did hydrochloric acid at comparable pH. As slurry pH increased from 9.9 to 11.6 in sodium hydroxide solutions, the percentage of protein extracted increased from 53 to 81%, but the protein content in extracted solids reached a maximum of 67% at pH 10.9 where 79% of the triticale protein was extracted. The optimum pH for dissolving triticale protein was around 10.9. At higher pH values the percentage of triticale protein extracted increased slightly; also, there was more risk of modifying the triticale protein and more chance of starch gelatinization.

Solid-to-Solvent Ratio. Ground triticale 204 was extracted with sodium hydroxide solutions at various

Table III

pH	Protein precipitated, %	pН	Protein precipitated, %
4.0	64	5.5	71
4.5	72	6.0	64
5.0	71	6.5	45

Table IV. Products from Tritical	es 204 and 385 <sup>a</sup>
----------------------------------	-----------------------------

		Yield,	%	to	otein	
	2	04	385,	2	204	
Product	pH 10	pH 10.8	pH 10.8	рН 10	pH 10.8	385, pH 10.8
Protein concentrate (first precipitate)	9	11	11	42	53	59
First supernatant	9	9	9	15	18	17
Second precipitate	2	2	1	8	6	4
Second supernatant	3	2	2	9	5	3
Bran	<b>25</b>	22	28	15	10	13
Layer above starch	9	12	17	4	4	2
Starch	42	38	29	0	1	0
Total	99	96	97	93	97	98

<sup>a</sup> Solid-to-solvent ratio was 1:6, dry basis. <sup>b</sup> Nitrogen  $\times$  5.7.

solid-to-solvent ratios of 1:3 to 1:10 (Table II). Since the percentage of protein extracted by sodium hydroxide solutions depends on pH of the slurry (Table I), normality of the sodium hydroxide was adjusted to the same pH value of 10.9. At this constant pH, the protein extracted increased from 47 to 82% when the solid-to-solvent ratio increased from 1:3 to 1:10. The largest increase in percentage of protein extracted occurred when the solid-to-solvent ratio increased from 1:4 to 1:6. An increase in the percentage of protein extracted was smaller when this ratio was further increased from 1:6 to 1:10. Probably, a solid-to-solvent ratio of 1:6 is a good compromise between the highest percentage of protein extracted and a minimum amount of extractant needed (Table II) and that ratio was always used unless otherwise specified.

**Precipitation pH.** The effect of precipitation pH on the alkaline extract of triticale 204 was determined at six pH values between 4 and 6.5. The amount of protein precipitated ranged from 45 to 72% (Table III) and this large difference demonstrated the importance of proper pH value for precipitating triticale protein from the alkaline extract. The maximum amount of protein precipitated, 72% at pH 4.5, was essentially maintained between pH 5.0 and 5.5. The amount of protein precipitated dropped substantially to 64% at pH 4.0 and at 6.0, and a large drop was observed when precipitation pH was 6.5. A pH value of 4.6 was chosen, therefore, to precipitate the triticale protein from the alkaline extract.

**Products from Triticales.** All figures in Table IV for the seven fractions from alkaline extraction of ground triticale have been rounded off to the nearest percent. Note that the yield of protein concentrate increased from 9 to 11% when the pH was raised from 10 to 10.8 for triticale 204; also, the total protein accounted for by the concentrate increased from 42 to 53%. The yields of first supernatants, second precipitates, and second supernatants were about the same at pH 10 and 10.8, but the yield of bran at the higher pH dropped. The yield of layer above starch was higher at pH 10.8 than at 10, but the trend was reversed for starch.

Both protein content and variety affect extraction at pH 10.8 (Table IV, triticales 204 and 385). Although the yield of protein concentrate was the same for both, the percentage of total protein accounted for by the concentrate

Table V	Composition of Protein Co	oncentrate and By-products from	Triticales Fas Gro	204 and 385 (% Dr	v Basis P
Table V.	composition of flotein co	incentiate and by products nom	Tinucales Fas Giu	204 and 000 (70 DI	y Dasis j

						Fat										
	Protei	n (N ×	5.7)		204		3	85	Fil	ber		Ash		5	Starch	
	20	4	<b>3</b> 85,	F	EE	GLC.	EE.	GLC.	204.	385.	20	)4	385.	20	4	<b>3</b> 85.
Material	pH 10	10.8	10.8	10	10.8	10.8	10.8	,	10.8		10	10.8	10.8	10	10.8	10.8
Ground triticale	17.7	17.7	15.0	1.7	1.7	1.7	1.3	2.0	2.8	2.6	2.1	2.1	1.8	56.5	56.5	60.5
Protein concentrate (first precipitate)	80.0	86.5	82.3	2.0	3.1	6.7	1.7	8.4	0.1	0.1	2.2	1.5	2.1			
First supernatant	31.3	34.7	28.4	0.2	0.3	0.2	0.1	0.1	0.2	0	15.3	17.3	22.3			
Second precipitate	79.7	68.0	69.3	8.2	24.5	23.3	12.5	17.3			1.0	1.0	1.5			
Second supernatant	58.1	44.2	29.1	0.2	0.1		0.1				9.1	24.4	20.1			
Bran	10.6	8.1	7.1	1.1	1.4	1.7	0.5	1.3	11.2	8.4	3.4	3.3	2.9	36.8	38.2	46.0
Layer above starch	9.0	5.9	1.8	0.4	0.9	0.7	0.1	0.3	2.7	1.6	4.2	4.4	1.8	72.0	67.3	88.2
Starch	0.2	0.2	0.2	0	0	0.1	0	0.1	0.1	0.1	1.1	0.5	1.4	101	93.3	92.3

<sup>a</sup> Key to abbreviations: EE, petroleum ether extraction; GLC, gas-liquid chromatography.

was lower for 204 compared with 385. Yields of the first supernatant, the second precipitates, and second supernatants were about the same for both varieties, but yields of the 204 bran and layer above starch were considerably lower than that from 385. The bran fraction contains a portion of the starch in the kernel. The yield of starch for 204 is about the same as that from conventional wet milling of triticale (Anderson et al., 1974) and of hard red spring wheat (Slotter and Langford, 1944). A much higher yield of starch was observed for 204 compared with 385.

The two samples of triticale were also used in air classification studies by Anderson et al. (1974). They found triticale 204 is less vitreous than 385 and responds more readily to fine grinding and air classification. The lower yield of bran and higher yield of starch for 204 compared with 385 (Table IV) also indicate that 204 responds better than 385 to wet processing.

Composition. The proximate analyses and starch contents of protein concentrate and by-products extracted at two pH values from triticales 204 and 385 appear in Table V. In addition to protein, fat, fiber, ash, and starch, triticale also contains sugars (Vaisey and Unrau, 1964) and pentosans (Heinrichs and Hill, 1971). For triticale 204 extracted at pH 10.8, the concentrate had a protein content of 86.5% compared with 17.7% for ground triticale; in addition, the concentrate had low fiber (0.1%), ash (1.5%), and fat (3.1%) (EE). The second precipitate had 68% protein but higher fat and lower ash than the protein concentrate. The first and second supernatants had from 35 to 44% protein, low levels of fat and fiber, but high ash. These two supernatants contained the albumins, globulins, salt, sugars, minerals, and other water-soluble materials. Their high ash content is partly due to sodium chloride formed by neutralizing the sodium hydroxide solution with a hydrochloric acid solution. The bran fraction had 8.1% protein, 1.4% fat, 3.3% ash, and 8.4% fiber. Saunders et al. (1974) reported that after dry milling triticale having 18.4% protein, the bran fraction had 24.1% protein, 5% fat, 6% fiber, and 5.3% ash. After extraction with alkali our bran fraction was considerably lower in protein, fat, and ash and higher in fiber. Although the layer above starch consists of both bran and starch, but predominantly starch, it had lower percentages of protein, fat, and fiber but higher ash and starch compared with the bran fraction. The starch fraction had a low protein content (0.2%)making it a high-quality starch. The absence of fat, the low content of fiber, as well as the high content of starch are also consistent with a high-quality material.

Extraction of triticale 204 is affected by pH (Table V). At pH 10 extraction of concentrate was less efficient in terms of protein content (80 vs. 86.5%) and yield (9 vs. 11 in Table IV). The concentrate extracted at pH 10 had

lower fat and higher ash compared with that extracted at pH 10.8, and the first supernatant also had a lower protein content at pH 10 compared with pH 10.8. Since more protein is available for the second alkaline extraction, protein in the second precipitate and supernatant at pH 10 was higher than at an extraction of pH 10.8. Protein contents of bran and layer above starch at pH 10 were higher than at pH 10.8, and this difference in protein contents again indicated that extraction of protein concentrate was more efficient at pH 10.8 than at 10. Despite differences in efficiency, the starch fraction was low in protein (0.2%) at both pH values. In general, proximate compositions and starch contents of protein concentrate and by-products follow the same trend at pH 10 as at 10.8.

The protein concentrate from triticale 385 had a lower protein content than 204 (Table V) because 385 had a lower initial protein content. The second 385 precipitate had a little higher protein content than that from 204, but the first and second supernatants, bran, and layer above starch from 385 had lower protein contents than corresponding 204 fractions. The lower fiber and higher starch contents of 385 grain compared with 204 are, in general, reflected in lower fiber and higher starch for all 385 fractions compared with corresponding 204 fractions. Ash contents of protein concentrate, first supernatant, second precipitate, and starch from 385 were higher than those from 204, but the trend was reversed for second supernatant, bran, and layer above starch. Fat contents (EE) of protein concentrate, first supernatant, second precipitate, bran, and layer above starch were lower than the corresponding 204 fractions because 385 grain had a lower fat content (EE).

Neutral Carbohydrates. The amount and kinds of neutral carbohydrates from acid hydrolysates of protein concentrate and by-products from triticales 204 and 385 are shown in Table VI. For ground triticale 385 the hydrolysate contained small amounts of arabinose and xylose, in addition to a large amount of glucose. Arabinose and xylose are derived from pentosans (Heinrichs and Hill, 1971) and hemicellulose (Vaisey and Unrau, 1964). Almost all the glucose is derived from starch because triticale 385 is 60.5% starch (Table V). The rest of the glucose is primarily from such oligosaccharides as maltotriose, maltotetraose, and maltopentaose (Vaisey and Unrau, 1964), although smaller amounts of glucose, sucrose, and maltose also contribute (Vaisey and Unrau, 1964).

The 385 concentrate yielded no neutral sugar on acid hydrolysis, and the second precipitate produced 4% glucose. Arabinose and xylose from acid hydrolysates of first and second supernatants of 385 are derived probably from water-soluble pentosans, whereas glucose from the hydrolysates of supernatants came primarily from mal-

Table VI. Neutral Carbohydrates from Acid Hydrolysates of Protein Concentrate and By-products from Triticales 204 and  $385^{a,b}$ 

	L-Arabinose		D-Xylose		D-Galactose		D-Glucose	
Material	204	385	204	385	204	385	204	385
Ground triticale	0	2.5	3.4	3.7	0	0	80.3	72.5
Protein concentrate (first precipitate)	0	0	0	0	0	0	1.5	0
First supernatant	2.6	3.0	2.7	3.6	1.6	0	17.3	10.5
Second precipitate	0	0	0	0	0	0	3.7	4.1
Second supernatant	1.3	3.8	1.8	4.7	0	1.1	9.1	8.9
Bran	11.1	7.4	16.4	11.7	Ó	0.8	57.7	58.3
Layer above starch	0	1.3	2.7	1.0	Ō	0	81.6	91.5
Starch	Ó	0	0	0	Ō	Ō	105	107

<sup>a</sup> Extraction pH was 10.8 for triticale 385; 10 for triticale 204. The neutral carbohydrates are expressed as percent of material on extreme left for each horizontal line, dry basis. <sup>b</sup> D-Mannose not detected in any fraction.

Table VII.	Amino Acid Composition of Protein	Concentrate and By-products:	from Triticales 204 and 385 <sup>a</sup>

	Ground		Protein concentrate (first precipitate)			First		Second		Second				
		cale	pH 10,	pН	10.8		supernatant		precipitate		supernatant		Bran	
Amino acid	204	385	204	204	385	204	385	204	385	204	385	204	385	
Lysine	3.2	3.4	2.9	3.3	3.2	2.6	3.0	2.3	3.4	1.7	2.0	3.7	4.3	
Histidine	2.3	2.4	2.2	2.5	2.5	2.0	2.5	2.0	2.4	1.7	2.1	2.2	2.6	
Ammonia	4.1	3.9	3.7	3.5	3.7	4.7	4.1	4.3	4.0	5.2	4.7	4.7	3.1	
Arginine	5.8	5.5	5.2	5.9	5.2	4.8	5.6	4.3	5.4	3.8	4.6	6.7	7.3	
Aspartic acid	6.0	6.2	6.2	5.5	5.4	9.3	9.0	4.7	5.2	5.8	6.2	7.6	7.2	
Threonine	3.1	3.1	3.5	3.1	3.1	3.3	3.1	3.0	3.1	2.7	2.6	3.8	3.7	
Serine	4.5	4.4	5.2	4.6	4.5	4.2	4.2	4.7	4.5	4.3	4.2	4.7	4.7	
Glutamic acid	28.3	29.5	30.8	31.0	31.9	26.2	28.0	32.9	29.2	33.3	33.1	18.9	20.7	
Proline	9.7	9.4	9.0	8.8	10.0	10.3	9.1	13.1	10.0	11.6	12.5	4.4	8.9	
Glycine	4.2	4.3	4.9	4.6	4.2	3.9	4.1	3.6	4.2	2.9	3.1	5.3	5.4	
Alanine	3.9	3.9	4.1	3.8	3.7	4.5	4.0	3.5	3.8	3.3	3.1	5.5	5.2	
Half-cystine	1.7	1.7	1.1	1.5	1.8	2.1	3.3	0.5	1.7	1.3	2.2	0.8	1.3	
Valine	4.7	5.0	5.3	4.8	4.8	4.4	4.3	4.8	4.8	4.3	3.9	5.5	5.5	
Methionine	1.8	1.8	1.3	2.0	2.0	2.0	1.8	2.0	1.9	1.7	1.7	2.0	2.0	
Isoleucine	3.7	3.9	3.8	3.8	4.0	3.4	3.2	4.0	4.0	3.8	3.6	3.8	3.9	
Leucine	6.7	6.4	7.1	6.9	6.8	6.0	5.3	7.3	7.1	6.5	5.8	7.1	7.0	
Tyrosine	3.2	3.3	4.1	4.0	3.6	2.3	2.8	2.7	3.3	2.2	2.1	2.7	3.2	
Phenylalanine	4.7	4.9	5.1	5.2	5.4	3.7	3.7	5.5	5.2	5.3	5.1	4.4	5.1	

 $^a$  Amino acid expressed in grams of amino acid/16 g of nitrogen recovered. Extraction pH was 10.8 unless otherwise specified.

totriose, maltotetraose, and maltopentaose with some contribution from glucose, sucrose, and maltose. The second supernatant and bran from 385 yielded a small amount of galactose, which was not detected in the starting triticale because of differences in concentration; the respective yields of the second supernatant and bran were only 2 and 28% (Table IV). Among all the fractions, bran produced the highest amount of arabinose and xylose, which were likely derived mostly from water-insoluble pentosans. According to Table V, 385 bran had 46% starch which, after hydrolysis, yielded most of the glucose for that fraction in Table VI. The layer above starch produced mostly glucose, derived primarily from starch (88%, Table V), and the small amounts of arabinose and xylose were probably produced from water-insoluble pentosans. The starch fraction yielded glucose without any other neutral carbohydrate.

The neutral carbohydrates from acid hydrolysates of 204 and its fractions were, in general, similar to the corresponding ones in 385.

Amino Acid Composition. The amino acid compositions of protein concentrate and by-products from the two triticales (Table VII) were corrected to 100% nitrogen recovery and expressed in grams of amino acid per 16 g of nitrogen recovered. Only significant differences in amino acid composition are discussed here. The two triticale grains have similar amino acid compositions, and their grain protein had high levels of glutamic acid and proline. The large amount of ammonia on a molar basis indicates that most of the glutamic and aspartic acids are present as glutamine and asparagine residues. High levels of glutamic acid, proline, and ammonia present in triticales were close to those reported for hard wheat (Horn et al., 1958), but the lysine content of the triticales was considerably higher than that of hard wheat (Horn et al., 1958).

Protein concentrate extracted at pH 10 from triticale 204 had a similar amino acid composition as 204 grain except the concentrate had higher threonine, serine, glycine, valine, and tyrosine but lower arginine, half-cystine, and methionine than the grain (Table VII). The 204 protein concentrate extracted at pH 10.8 had higher lysine, histidine, arginine, half-cystine, and methionine but lower aspartic acid, threonine, and serine than that extracted at pH 10. Lysine and total sulfur amino acids of the 204 concentrate extracted at pH 10.8 were essentially equal to those of the 204 grain. The lysine content of 3.3 g per 16 g of nitrogen for 204 concentrate extracted at pH 10.8 was much higher than the lysine content of 2.3 for gluten prepared from 204 flour by wet milling (Anderson et al., 1974).

The first supernatant from 204 had lower lysine, histidine, arginine, glutamic acid, glycine, isoleucine, leucine, tyrosine, and phenylalanine and higher aspartic acid, proline, alanine, and half-cystine than 204 concentrate extracted at pH 10.8 (Table VII). The second 204 pre-

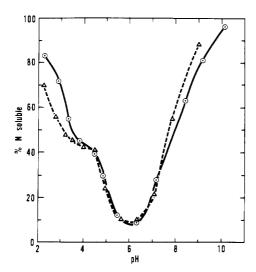


Figure 2. Nitrogen solubility of triticale protein concentrate at various pH values. Protein concentrate (0.1 g) was stirred with 10 ml of water to which either hydrochloric acid (below pH 4) or sodium hydroxide (above pH 4.5) solution was added to arrive at the desired pH: ( $\odot$ ) Fas Gro variety 204; ( $\triangle$ ) Fas Gro variety 385. Both concentrates were extracted at pH 10.8.

cipitate had lower lysine, histidine, arginine, aspartic acid, glycine, half-cystine, tyrosine, and higher proline than the 204 concentrate extracted at pH 10.8. The second 204 supernatant had lower lysine, histidine, arginine, threonine, glycine, alanine, valine, methionine, tyrosine, and higher proline than the 204 concentrate extracted at pH 10.8. The 204 bran had higher lysine, arginine, aspartic acid, threonine, glycine, alanine, and valine and lower histidine, glutamic acid, proline, half-cystine, tyrosine, and phenylalanine than the 204 concentrate extracted at pH 10.8.

The amino acid composition of the protein concentrate and by-products from triticale 385 did not differ greatly from the corresponding 204 fractions (Table VII). The 385 concentrate had lower arginine but higher proline and half-cystine than the 204 concentrate, and the first supernatant from 385 had higher lysine, histidine, arginine, and half-cystine, and lower proline, alanine, and leucine than that from 204. The second 385 precipitate had higher lysine, histidine, arginine, glycine, half-cystine, and tyrosine, and lower glutamic acid and proline than that from 204, and the second supernatant from 385 had higher lysine, histidine, arginine, and half-cystine, and lower leucine than that from 204. The 385 bran had higher lysine, histidine, proline, half-cystine, tyrosine, and phenylalanine than 204 bran. Amino acid compositions of unextracted protein of 204 and 385 brans were close to that of triticale bran from dry milling (Saunders et al., 1974) except their bran had higher glutamic acid and half-cystine when their data were calculated to 100% nitrogen recovery.

Nitrogen Solubility. The triticale protein concentrate used for nitrogen solubility studies was extracted at pH 10.8, and the percentage of nitrogen soluble at a number of pH values from 2.2 to 10.2 for triticale 204 and 385 concentrate is plotted in Figure 2. The minimum solubility of 204 concentrate was near pH 6 where around 8% of the nitrogen was soluble. When the triticale protein concentrate was prepared by precipitation at pH 4.6, the ionic strength of the solution was higher than that used here due to salt present originally in triticale and due to neutralization of alkali. The solubility of 204 concentrate in 0.25 M KCl decreased drastically to 3% at pH 4.7 (not shown in Figure 2), but was essentially unchanged at pH

Table VIII. Some Functional Properties of Triticale Protein Concentrate Compared with a Soy Protein

Protein concentrate	Extrac- tion pH	Hydra- tion capacity	sifying	sion
Triticale 204	10	4.1	91	87
Triticale 204	10.8	3.7	85	81
Triticale 385	10.8	3.9	90	85
Soy protein isolate			45	45

5.3 and 5.6 compared with the values in Figure 2 where no salt was added. The difference in ionic strength accounts for the different pH values (4.6 and 6) for minimum solubility of protein concentrate, and the lower ionic strength used in Figure 2 makes the concentrate somewhat soluble at pH 4.6 to 6 instead of insoluble. Solubility of 204 concentrate increased rapidly as pH increased beyond 7; almost all the nitrogen was soluble at pH 10.2. Nitrogen solubility below pH 5.5 increased as pH decreased, and a shoulder in the solubility curve was observed around pH 4.2. Another rapid increase in solubility was seen below pH 3.5, and solubility reached 83% at pH 2.3.

The nitrogen solubility curve of triticale 385 concentrate was, in general, similar to that of the 204 concentrate (Figure 2), and the minimum solubility of the 385 concentrate was also near pH 6 where around 9% of the nitrogen was soluble. The nitrogen solubility of the two concentrates was close between pH 4.2 and 7.3, but above pH 7.3 solubility of the 385 concentrate was higher, and almost all nitrogen in 385 concentrate was soluble at pH 9. A shoulder in the nitrogen solubility curve was also observed for the 385 concentrate around pH 4. However, its solubility was lower than that of the 204 concentrate below pH 4, and 70% of the nitrogen in the 385 concentrate was soluble at pH 2.2.

**Hydration Capacity.** The hydration capacity (weight of sediment per weight of sample) of the triticale 204 concentrate (Table VIII) decreased from 4.1 to 3.7 when the extraction pH of the concentrate increased from pH 10 to 10.8. The triticale 385 concentrate had a higher hydration capacity than the 204 concentrate when both concentrates were extracted at pH 10.8.

Emulsifying Activity and Emulsion Stability. When extraction pH of the triticale 204 concentrate increased from 10 to 10.8 (Table VIII), emulsifying activity and emulsion stability decreased 6%. Emulsion stability was 4% lower than the corresponding emulsifying activity at pH 10 and 10.8. Triticale 385 concentrate had a higher emulsifying activity and emulsion stability than 204 concentrate when both concentrates were extracted at pH 10.8. The emulsion stability of the 385 concentrate was 5% lower than its emulsifying activity. A commercial soy protein isolate gave emulsifying activity and emulsion stability values of 45% under the same experimental conditions as triticale concentrate. Emulsifying activity and emulsion stability of the triticale concentrate are remarkably good, and they are much better than those of the commercial soy isolate.

Potential Uses of Protein Concentrate and Byproducts. Triticale concentrate may find application in foods as a protein ingredient. The attractive water-binding capacity, excellent emulsifying activity, and emulsion stability of the concentrate suggest possible use as fat emulsifiers and water-absorbing agents in prepared foods. Since triticale whole grain, shorts from milling grain, and partially dehulled grain have been successfully extruded (Anderson et al., 1974), the residue after one protein extraction presumably can be extruded into breakfast

#### LITERATURE CITED

- American Association of Cereal Chemists, AACC Approved Methods, Revised, St. Paul, Minn., 1971.
- Anderson, R. A., Stringfellow, A. C., Griffin, E. L., Jr., Northwest. Miller 279(2), 10 (1972).
- Anderson, R. A., Stringfellow, A. C., Wall, J. S., Griffin, E. L., Jr., Food Technol. 28(11), 66 (1974).
- Black, L. T., Spyres, G. G., Brekke, O. L., Cereal Chem. 44, 152 (1967).
- Cavins, J. F., Friedman, M., Cereal Chem. 45, 172 (1968).
- Centro Internacional de Mejoramiento de Maiz y Trigo, CIMMYT Report on Wheat Improvement, 1973, El Batan, Mexico, 1974.
- Garcia, W. J., Wolf, M. J., Cereal Chem. 49, 298 (1972).
- Heinrichs, E. R., Hill, R. D., Abstract 103, Cereal Sci. Today 16, 306 (1971).
- Horn, M. J., Fifield, C. C., Blum, A. E., Warren, H., Cereal Chem. 35, 411 (1958).
- Hulse, J. H., Spurgeon, D., Sci. Am. 231(2), 72 (1974).
- Kaltsikes, P. J., Larter, E. N., Wheat Newslett. 17, 30 (1970).
- Lorenz, K., Food Technol. 26(11), 66 (1972).
- Lorenz, K., Welsh, J., Normann, R., Maga, J., Cereal Chem. 49, 187 (1972).
- Madl, R. L., Tsen, C. C., Cereal Chem. 50, 215 (1973).

- Muntzing, A., Proc. Int. Wheat Genet. Symp., 2nd (1963); Hereditas Supp. 2, 291 (1966).
- Rooney, L. W., Gustafson, C. B., Perez, N., Porter, K. B., "Agronomic Performance and Quality Characteristics of Triticale Grown in the Texas High Plains", Texas Agricultural Experiment Station, College Station, Tex., Sept 1969.
- Saunders, R. M., Betschart, A. A., Connor, M. A., Edwards, R. H., Kohler, G. O., "Triticale: First Man-Made Cereal", Tsen, C. C., Ed., American Association of Cereal Chemists, St. Paul, Minn., 1974, p 280.
- Sloneker, J. H., Anal. Biochem. 43, 539 (1971).
- Slotter, R. L., Langford, C. T., Ind. Eng. Chem. 36, 404 (1944).
  Tsen, C. C., Hoover, W. J., Farrell, E. P., Cereal Chem. 50, 16 (1973).
- Unrau, A. M., Jenkins, B. C., Cereal Chem. 41, 365 (1964).
- Vaisey, M., Unrau, A. M., J. Agric. Food Chem., 12, 84 (1964).
- Yasumatsu, K., Sawada, K., Moritaka, S., Misaki, M., Toda, J., Wada, T., Ishii, K., Agric. Biol. Chem. 36, 719 (1972).

Received for review October 30, 1975. Accepted January 26, 1976. Presented at the Division of Agricultural and Food Chemistry, 170th National Meeting of the American Chemical Society, Chicago, Ill., Aug 24–29, 1975. Mention of firm names or trade products does not imply that they are endorsed or recommended by the Department of Agriculture over other firms or similar products not mentioned.

# Influence of Suspension Medium and pH on Functional and Protein Properties of Defatted Peanut Meal

Kay H. McWatters,\* John P. Cherry,<sup>1</sup> and Mac R. Holmes

Defatted peanut (Arachis hypogaea L. C.V. Florunner) meal was blended with either water, 0.1 M NaCl, or 1.0 M NaCl and the pH of each suspension adjusted to either 1.5, 4.0, 6.7, or 8.2; two-step sequential adjustments from 6.7 to 4.0 to 6.7 and from 6.7 to 4.0 to 8.2 were also included. All suspensions had similar viscosities. Those at pH 4.0 produced soluble extracts with lowest percentage protein and failed to form emulsions. Suspensions at pH 6.7 varied widely in percentage protein, produced the least increase in foam volume, and formed poor emulsions. The most desirable emulsions and foams were produced by peanut meal-water suspensions adjusted from pH 6.7 to 4.0 to 8.2 and from pH 6.7 to 1.5. Gel electrophoresis of soluble proteins and multiple regression analysis showed that functionality of peanut meal was influenced by complex interactions involving suspension medium, pH, and level and character of soluble proteins.

Peanuts have traditionally been consumed in the form of peanut butter and in candies, salted nuts, and snack-type crackers because of their highly acceptable roasted flavor. In recent years, interest has developed in high-protein products such as defatted peanut flour, concentrates, and isolates as potential ingredients having the capacity to perform specific functions in food systems. Oilseed protein products act as emulsifiers and extenders in meat products, fat and water absorption agents in meats and bakery products, thickeners in soups and gravy products, gelling agents in meat products, color control agents in bread products, and whipping agents in toppings, chiffon mixes, and confections (Wolf and Cowan, 1971).

The level or proportion of soluble proteins has been used as a measure of the availability of these components for functional uses (Johnson, 1970; Mattil, 1971; Wolf and Cowan, 1971; Cherry et al., 1975). For example, processing techniques such as moist heat may be applied for the express purpose of modifying the protein components of oilseeds to fit specific product applications. The application of moist heat to alter certain physicochemical and solubility properties of peanut proteins and thus change their functional properties has been discussed by Cherry et al. (1975). These workers found that water-soluble proteins of moist-heated peanut seeds were altered sequentially to various structural components, aggregates, and insoluble forms. Arachin, the major storage globulin of peanut seeds, was altered to these denatured forms at at slower rate than nonarachin proteins. Further studies

Department of Food Science (K.H.M., J.P.C.) and Agricultural Economics (M.R.H.), University of Georgia, College of Agriculture Experiment Stations, Experiment, Georgia 30212.

<sup>&</sup>lt;sup>1</sup>Present address: Southern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, New Orleans, La. 70179.