# Comparison of Gluten Quality in Triticale: A Fractionation-Reconstitution Study<sup>1</sup>

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#### **ABSTRACT**

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The gluten quality of four agronomically acceptable secondary hexaploid triticales, one hard red spring wheat (Marquis), and one soft wheat (Fredrick) were examined. Whereas flour protein contents were similar, gluten protein contents and falling number values for the triticales were variable and lower than those for both wheats, especially Marquis. Mixing and baking studies of flours reconstituted on an equal gluten protein basis indicated that a considerable range of gluten quality existed among the triticales and between the hard and soft wheats. A linear

relationship between peak time and peak area with gluten stretching values for the triticales was demonstrated. When flours were reconstituted with equal amounts of gluten protein from each of the triticales, the rheological properties of these flours were linearly related to the gluten strength of the parental gluten. Loaf volume increased for all the flours reconstituted at the higher gluten levels. On the basis of these analyses, Impala triticale gluten appeared to be similar in quality to that found in Marquis wheat.

The future commercial success of triticale as a crop depends upon the development of triticale lines with good breadmaking quality. Many of the studies to evaluate the breadmaking quality of triticales were with primary hexaploid triticales. Generally, the triticales had inferior dough strength and different rheological properties when compared to wheat (Unrau and Jenkins 1964, Lorenz et al 1972, Tsen et al 1973, Haber et al 1976). Factors that contribute to the inferior properties include high amylolytic (Welsh and Lorenz 1974) and proteolytic activities (Madl and Tsen 1973) that are known to have detrimental effects on baking performance. It has also been observed that triticales have a higher level of water-soluble proteins and a correspondingly lower proportion of the gluten proteins (Chen and Bushuk 1970), although actual gluten values are not reported.

Secondary hexaploid triticales are obtained by crossing a primary hexaploid triticale with either an octaploid triticale or a hexaploid wheat. Unlike the parents of a primary hexaploid

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triticale, one of the parents of the secondary hexaploid triticale can be a bread wheat. Thus, the potential for introducing breadmaking quality is considerably greater with the secondary triticales. Among the recently developed secondary hexaploid triticales, lines with lower  $\alpha$ -amylase-activity (Peña 1979) and better dough strength (Lorenz and Welsh 1977, Peña 1979) have been identified.

The object of the present study was to evaluate the gluten quality of four agronomically acceptable secondary hexaploid triticales. The effects of high and variable enzyme levels in the triticales were minimized by using a no-time baking procedure. This approach was justified because the enzyme activities and the protein quality problems are due to genetically separate events. Mixing and baking qualities were evaluated for reconstituted flours to compare the gluten quality of the samples on an equal gluten level.

#### **MATERIALS AND METHODS**

## **Grain and Flour Samples**

Four secondary hexaploid triticales—three grown in Sonora, Mexico, during the 1980-1981 season as part of the International Maize and Wheat Improvement Center (CIMMYT) yield nurseries, and the fourth (cultivar Carman) grown at the University of Manitoba, Winnipeg, Canada, during the summer of 1980—were all harvested under dry conditions. Hard red spring

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wheat (cultivar Marquis) of good dough strength was also grown at Winnipeg in 1980-1981. Flour from a soft wheat (cultivar Fredrick) was obtained from a local milling company.

The wheat and triticale samples were tempered (to 15.5 and 14.5% moisture, respectively) overnight and milled on a Buhler experimental mill. The wheat flour streams were blended and rebolted through a 70-gg (236- $\mu$ m) sieve. For triticale flours the last flour stream from the reduction system was first sifted through an 11×× (125- $\mu$ m) sieve to reduce ash content of the flour before blending and rebolting as with the wheat flour.

# Flour Analysis

Ash, moisture, protein (N $\times$ 5.7), falling number value, and the Zeleny sedimentation value, were all determined in duplicate or triplicate using AACC methods 08-01, 44-15A, 46-12, 56-81b, and 56-60, respectively (AACC 1976). Protein contents of gluten and nongluten fractions were determined by the micro-Kjeldahl method 46-13 (AACC 1976).

# **Mixing Characteristics**

Mixing characteristics of original flours were determined with the Swanson and Working mixograph (National Mfg. Co.) using 35 g of flour (14% mb), at 60% absorption for the wheat Marquis and at 59% absorption for the soft wheat and the triticales. The gauge (spring) was set in position 8. Mixing characteristics of original and fractionated-reconstituted flours (5-g samples, 14% mb) were evaluated from 5-g mixographs using the electronic recording dough mixer (Voisey et al 1966) equipped with a Dynamaster recorder (The Bristol Company of Canada, Toronto). The mixer speed was 95 rpm. Water absorption was as noted later. Peak time, peak height, and peak area (the area under the curve between the origin and the peak time) were all measured in 5-g mixographs.

#### Gluten Stretching Test

Glutens were obtained from flour using the Theby glutenwashing apparatus according to AACC method 38-11 (1976). Gluten stretching values were determined in duplicate by measuring the force required to stretch and break a strand of fresh wet gluten using the apparatus and technique of Matsuo (1978).

# **Baking Procedure**

The baking procedure used is referred to as the no-time/sheeting procedure. The baking formula included 25 g of flour (14% mb), 0.625 g of sugar, 0.25 g of sodium chloride, 0.75 g of shortening, 1.0 g of fresh yeast, 0.025 g of ammonium phosphate, 70 ppm of ascorbic acid, 30 ppm of potassium bromate, and water to a fixed absorption. These dough ingredients were premixed for 1 min in a National mixer (National Mfg. Co.) and then developed by passing them 15 times through sheeting rolls (National Mfg. Co.) at a 5/64-in. gap. The gap distance between rolls was chosen to be the minimum without tearing the dough. After each pass, the dough was shaped into a cylinder and rotated 90° for the next pass. Doughs were kept in a fermentation cabinet (32.2°C, 90–95% rh) for an intermediate proofing period of 20 min following the recommendations of Kilborn and Tipples (1979). Without folding between passes, the dough was next sheeted three times at successive gap distances of 7/32, 5/32, and 5/64-in. The sheeted dough was rolled into a cylinder, panned, proofed for 60 min, and baked for 25 min at 216° C. Loaf volume was measured by rapeseed displacement. The baking formula and other conditions were established after considering several baking formulas used by different investigators (Kilborn and Tipples 1974, Stenvert et al 1979, Moss 1980, Kilborn et al 1981) for the no-time baking system.

## Flour Fractionation

Prior to fractionation, flour (200 g, moisture as is) from each of the six samples was partially defatted by extracting overnight (16 hr) with n-hexane (1 L) in a shaking water bath at  $22-26^{\circ}$  C. The slurry was filtered on a Buchner funnel. The residue was then reextracted with 0.5 L of solvent for an additional hour, recovered

by filtration, and washed with a small portion of solvent. Lipids were recovered from the combined extracts by solvent evaporation with a rotary evaporator and were stored at  $-18^{\circ}$  C until used for reconstitution of flours. The flour was air-dried at room temperature until solvent odor was no longer detected.

The partially defatted flours were fractionated into gluten and a starch plus water solubles (S-WS) fraction as follows: flour (50 g as is) was extracted with 0.001 M NaCl solution (100 ml) for 5 min and then centrifuged (15,000 × g) for 10 min at 4° C. The supernatant was kept at 0-4° C. The flour residue was made into a dough by mixing for 1 min in a GRL mixer, placed in a metallic container, and kneaded in the presence of a small portion of the salt solution for 1-2 min to gradually remove starch and remaining soluble material. The suspension obtained while kneading the dough was percolated through an  $11\times\times$  (125- $\mu$ m) sieve. The kneading operation was repeated until the gluten mass was formed and the washing solution was relatively clear and not milky. The gluten was allowed to relax in distilled deionized water for 30 min before it was frozen and freeze-dried. The S-WS suspension and the supernatant obtained at the beginning of the operation were combined, frozen, and freeze-dried. The freeze-dried gluten and S-WS fraction were separately ground to pass a  $9\times\times$  (156- $\mu$ m) sieve and the moisture and protein content determined. The fractions were stored at 0-4° C.

#### Flour Reconstitution

Ground gluten and S-WS fractions were blended in the desired proportions before the extracted lipid material was added. Reconstituted flours were rehydrated in a fermentation cabinet for 32 hr at room temperature. The humidity control (giving 90–95% rh) was turned on for 15 min three times during the rehydration period. The samples reached a moisture content between 11 and 13%. All original and reconstituted flours were tested in duplicate for mixing characteristics and for baking quality using the notime/sheeting procedure. For reconstitution I. flour fractions were reconstituted into flours having the same constituents and constituent levels as the original flours. For reconstitution II, a composite S-WS fraction was obtained by blending the S-WS fractions from all flours (20% from each of Impala, Carman, Marquis, and Fredrick; only 10% from each of 4T and 11T). Similarly a composite lipid fraction was prepared by combining equal amounts of the extracted lipids from each sample. The composite lipid fraction (added at 0.85% weight of reconstituted flour, the average free lipid content of all flours) and the composite S-WS fraction were blended with gluten isolated from each of the triticale and wheat samples. These reconstituted flours differed only in their source of gluten as they had the same S-WS and lipid fractions and the same amount of gluten protein (11.5% dry basis). The gluten protein level was selected to be similar to what is found in a good baking quality wheat sample.

# **RESULTS AND DISCUSSION**

## Characteristics of Samples

The chemical and rheological comparisons that were made on the samples are presented in Table I. The particular samples of the four triticales and two wheats were selected to be as similar in protein content as possible. The ash contents of all flours were similar, although those of 4T and Fredrick were somewhat higher. From the falling number values it was concluded that the amylolytic activity varied among the triticales from moderate (11T) to high (4T). Based on the fact that all the samples were harvested under dry conditions and that another study with the same lines has shown that amylase activity increases from as early as 20 days post-anthesis (Macri et al 1986), it was assumed that the variable falling number values were not caused by sprouting.

The Zeleny sedimentation test and mixing times varied among the triticales. Both of these tests are affected by gluten quantity and quality factors. Lukow and Bushuk (1984) showed that with wheat, both values decline with germination. Whether this is caused by amylase activity or a breakdown of the gluten complex by proteases is not known. Macri et al (1986) reported that protease

activities in these cultivars decline during development through to

The gluten protein content of the triticale cultivars varied substantially, ranging from 50% (4T) to 69% (Impala) of the defatted flour protein. This compares to 78 and 86% for Fredrick and Marquis, respectively. The difference in the proportion of gluten protein present in wheat and in the various triticales explains in part the generally weaker rheological properties of triticale doughs. These results support a similar effect reported by Tsen et al (1973). Whereas the differences in gluten protein content among the triticales precluded any direct comparison of gluten

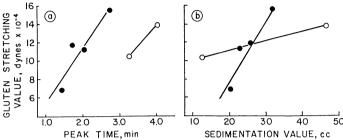


Fig. 1. Comparison of (a) 35-g mixograph mixing times and (b) sedimentation values for the original flours with gluten stretching values (triticales, •; wheats, 0).

quality in the dough system, the strength of the glutens was evaluated by a gluten stretching test (Table I). With the exception of 11T, the glutens were similar in their protein content. The marginally lower protein content of 11T gluten would not, however, explain why the gluten from 11T appeared to be the weakest gluten. Impala and Marquis had the highest stretching values. Among the triticales, the gluten stretching values were linearly related to both mixing time and sedimentation values. The wheats and triticales (Fig. 1) appear to differ from one another in both these relationships.

Although the results of several of the above tests suggest that these triticale samples have variable breadmaking potential, this characteristic can only be effectively evaluated in a breadmaking system. An evaluation of the gluten quality as it relates to breadmaking potential was complicated by the different levels of both gluten and amylase activity in the original samples. Because the no-time/sheeting procedure described in the methods was found to be less affected by moderate levels of amylase activity than the AACC straight-dough procedure (preliminary results) this method was used throughout.

## Gluten Quality of Reconstituted Flours

The variable levels of important constituents were normalized through a fractionation-reconstitution (F-R) study; however, this treatment itself was likely to have some effects on both rheological and baking characteristics. To evaluate the magnitude of the F-R

TABLE I
Analysis of Flours and Gluten Samples

Quality		]	Friticale		Wi	LSDa		
Characteristic	4T	11T	Carman	Impala	Marquis	Fredrick	(P = 0.05) $0.5$	
Protein <sup>b</sup> (%)	11.3	11.9	11.8	12.4	12.6	10.0		
Defatted flour protein <sup>b</sup> (%)	11.8	12.0	12.4	12.9	13.3	10.1	0.4	
Falling number value (sec)	66	221	129	169	404	274	18	
Ash <sup>b</sup> (%)	0.56	0.44	0.45	0.46	0.46	0.52	0.01	
Free lipid <sup>b,d</sup> (% of flour)	0.64	0.63	0.81	1.00	1.13	0.93		
Zeleny sedimentation (cm <sup>3</sup> )	23	20	26	32	46	12	1.6	
35-g Mixograph peak time <sup>c</sup> (min)	2.0	1.4	1.7	2.7	4.0	3.2	0.2	
Gluten content (%)								
in defatted flour <sup>b,d</sup>	6.3	8.6	8.5	10.0	12.7	8.6		
Protein in gluten <sup>b</sup> (%)	93.5	86.5	92.6	89.3	89.9	91.7	1.1	
Gluten protein (%)								
in defatted flour <sup>b</sup>	5.9	7.4	7.9	8.9	11.4	7.9	0.2	
Gluten stretching value								
$(dynes \times 10^{-4})$	11.3	6.8	11.8	15.5	13.8	10.3	1.5	

<sup>&</sup>lt;sup>a</sup> Least significant difference.

TABLE II
Effect of Fractionation-Reconstitution (Reconstitution I) on Functional Properties of Triticale and Wheat Flours

Quality Characteristic		Triticale							Wheat				
	4T		11T		Carman		Impala		Marquis		Fredrick		LSDb
	Oa	Rª	0	R	0	R	0	R	0	R	0	R	(P=0.05)
Flour protein <sup>c</sup> (%)	11.3	11.6	11.9	12.2	11.8	12.1	12.4	12.5	12.7	13.3	10.0	10.3	0.5
Falling number (sec)	66	65	221	203	129	129	169	180	404	418	274	280	8.0
5-g Mixograph <sup>d</sup>													
Peak time (min)	1.3	1.4	1.0	1.0	1.1	1.4	1.6	2.0	2.6	2.1	1.8	1.6	0.2
Peak height (cm)	12.6	12.1	12.0	12.0	12.8	12.0	12.8	11.0	10.9	12.8	9.3	9.8	0.2
Peak area (cm <sup>2</sup> )	25.0	36.1	22.2	22.7	28.4	32.2	43.0	41.6	56.0	52.3	32.6	29.2	3.2
Breadmaking <sup>e</sup>													
Loaf volume (cm <sup>3</sup> )	134	118	160	155	174	159	203	189 <sup>g</sup>	225	$212^{g}$	176	156	4.0
Crumb appearance <sup>f</sup>	U	U	U	U	U	U	S	S	S	S	U	U	
Crumb structure <sup>f</sup>	U.C.SY	U.C.SY	U,O	U,O	U,O	U,O	S	S	S	S	U,SO	U,O	

<sup>&</sup>lt;sup>a</sup> O = Original, R. = reconstituted.

<sup>&</sup>lt;sup>b</sup>Dry weight basis.

<sup>&</sup>lt;sup>c</sup>Swanson and Working mixograph. Water absorption was 60% for Marquis flour and 59% for the other flours.

<sup>&</sup>lt;sup>d</sup>Data from single determinations.

<sup>&</sup>lt;sup>b</sup>LSD = Least significant difference.

<sup>&</sup>lt;sup>c</sup>Dry weight basis.

<sup>&</sup>lt;sup>d</sup>Water absorption was 62% for Marquis and 58% for all other cultivars.

<sup>&</sup>lt;sup>e</sup>Water absorption was 63% for Marquis and 58% for all other cultivars.

S = Satisfactory, U = unsatisfactory, O = open, SO = slightly open, SY = sticky, C = compact.

<sup>&</sup>lt;sup>8</sup>Single data point.

treatment effect, the individual flours were initially reconstituted to their original composition (reconstitution I) and the properties of the original and reconstituted flours were compared (Table II). Comparison of falling numbers and protein values for the reconstituted flours with those of the original flours indicated that the major components have been quantitatively reconstituted. Although the F-R procedure did not generally alter the mixograph patterns relative to those of the unfractionated flours, variations in the mixing characteristics measured were observed for some of the samples (Table II). The F-R treatment also resulted in a 3-11% reduction in loaf volume. Reductions in loaf volume with similar treatments were also observed in previous studies (Marais and D'Appolonia 1981a, Finney et al 1982). The crumb characteristics of the reconstituted flours were similar to those of the unfractionated flours.

To evaluate the quality of the glutens from the six different sources on an equal quantitative basis, a second reconstitution experiment (reconstitution II) was conducted. An equal amount of gluten protein from each of the six sources was added to the common composite S-WS fraction plus composite lipids. The desired reconstitution was achieved based on the uniform protein contents of those six reconstituted flours (Table III).

The mixing characteristics of the reconstituted flour doughs differed significantly (Table III) indicating gluten quality variation among the triticales and wheats. The peak times and peak areas of the reconstituted (II) flours on an equal gluten basis were linearly related to those of either the original samples or reconstituted (I) flours that had significantly different gluten contents. Thus gluten quality is an important factor in controlling dough mixing properties. This observation agrees with Marais and D'Appolonia (1981b) who reported that gluten and water-solubles had major effects on mixing properties of wheat flour doughs. Both peak time and peak area showed a positive linear relation with gluten stretching values (Figs. 2 and 3). The original triticale and wheat samples are represented by two divergent lines (Fig. 2). However in the reconstituted (II) flours where all samples were quantitatively similar, differing only in the gluten sources, the difference between wheat and triticale was not apparent (Fig. 3). All samples with the

exception of the Marquis sample increased in gluten level relative to the original samples. This would be expected to cause longer mixing times. Furthermore the mixing time for the Marquis sample decreased, presumably as a result of the F-R treatment as was observed in reconstitution I (Fig. 2). The common S-WS and lipid fractions would also likely diminish differences observed in the original samples.

The bread dough is the system in which the gluten interacts with other functional components to fully express its quality in terms of loaf volume and crumb characteristics. Baking quality varied among the reconstituted flours tested, again indicating a wide variation in gluten quality among the secondary hexaploid triticales and between the hard and soft wheats (Table III). The gluten stretching value of the Impala sample and the 5-g mixograph and breadmaking characteristics of the reconstituted (II) flour with Impala gluten demonstrated the Impala gluten to be as strong as that from a good bread wheat flour, Marquis.

The rheological parameters of dough (expressed as the peak time and peak area of the 5-g mixograph) and the gluten strength (expressed as stretching values) were, in general, related well with breadmaking characteristics except for the 4T triticale. Extremely low falling number (66) and gluten protein content (5.89% of defatted flour) of 4T may explain this discrepancy. The flour reconstituted (II) with 4T gluten produced bread with a much smaller loaf volume than that anticipated from the dough mixing properties and its gluten stretching value. It may indicate an extremely unusual loss of gliadin so that the gliadin/glutenin ratio might have decreased in the 4T gluten. The inferior loaf volume of 4T relative to that of 11T may again reflect the greater resistance to extension, which may in turn prevent proper expansion of the dough during the fermentation and oven spring stages.

The four triticales examined in this study had lower falling numbers, sedimentation values, and gluten protein contents than standard bread wheat, Marquis. Whereas all the triticales, including Impala, were deficient in the proportion of gluten protein in their flours, the gluten quality of Impala was equivalent or slightly superior to that found in Marquis. The present results indicate that the breadmaking potential of triticales with Impala-

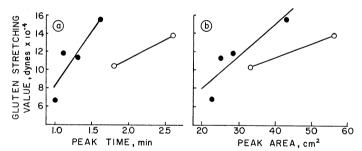


Fig. 2. Relationship between 5-g mixograph characteristics (peak time, a; peak area, b) of original flours and gluten stretching values (triticales, •; wheats, 0).

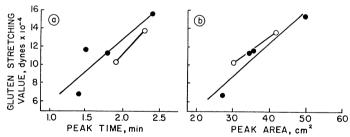


Fig. 3. Relationship between 5-g mixograph characteristics (peak time, a; peak area, b) of reconstituted (II) flours and gluten stretching values (triticales, •; wheats, 0).

TABLE III
Influence of Gluten Quality on Functional Properties of Reconstituted Flours (Reconstitution II) Having the Same Gluten Protein Content (11.5% db)

Quality Characteristic			<b>Friticale</b>		WI	LSD <sup>a</sup>	
	4T	11T	Carman	Impala	Marquis	Fredrick	(P=0.05)
Flour protein <sup>b</sup> (%)	14.5	14.3	14.3	14.3	14.5	14.6	0.32
5-g Mixograph <sup>c,d</sup>							
Peak time (min)	1.8	1.4	1.5	2.4	2.3	1.9	0.2
Peak height (cm)	10.0	10.4	13.2	11.1	9.5	8.5	0.8
Peak area (cm²)	34.6	27.4	35.7	50.2	42.2	30.4	5.5
Breadmaking <sup>c</sup>							
Loaf volume (cm <sup>3</sup> )	169	193	198	225	217	204	10.0
Crumb appearance <sup>e</sup>	U	Q	U	S	S	0	
Crumb structure <sup>e</sup>	U,O	Q,SO	U,O	S	S	Q,ŠO	

<sup>&</sup>lt;sup>a</sup>LSD = Least significant difference.

<sup>&</sup>lt;sup>b</sup>Dry weight basis.

<sup>°</sup>All samples tested at 60% water absorption for mixograms and at 61% for baking.

<sup>&</sup>lt;sup>d</sup>Electronic recording dough mixer.

<sup>&</sup>lt;sup>e</sup>S = Satisfactory, Q = questionable, U = unsatisfactory, O = open, SO = slightly open.

type gluten could be further improved if both protein content and the proportion of gluten protein in the flour increased.

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