

# Factors Contributing to Baking Quality Differences in Hard Red Spring Wheat. I. Bases for Different Loaf Volume Potentials<sup>1</sup>

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## ABSTRACT

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When baked at a fixed level of bromate (10 ppm), the flour from two hard red spring wheat varieties, Coteau and Butte, had the same loaf volumes. Interchanges of the starch, gluten, tailings, and water-soluble fractions showed little indication that these fractions differ to any great extent. The known superior loaf volume potential of Coteau resides in its ability to respond to bromate. Both the albumin and globulin protein classes are

implicated as an effect. No evidence was found indicating that the small variations in pentosan contents in the two varieties affected bromate requirements. A major portion of the variability in bromate requirement could not be explained by the variables that were measured. At optimum bromate levels, loaf volume correlated positively with total protein content but negatively with the percentage of glutenin and residue protein.

Wheat flour is unique in its ability to form a dough that will expand by trapping gas produced during fermentation. Different flours vary widely in this ability (Hoseney et al 1972), and efforts to elucidate the causes for different flour functional properties have kept many cereal chemists at work for more than 50 years. As indicated by Pomeranz (1973), several factors complicate efforts to relate chemical composition and structure of wheat flour components to processing properties. These are the large number of components, high molecular weights, limited solubility and the difficulty of separating or isolating pure components without altering them, and the interaction of components during dough mixing, fermentation, and baking.

A number of studies have attempted to link loaf volume potential to chemical properties (D'Appolonia et al 1970; Hoseney and Finney 1971; Orth and Bushuk 1972, 1973; Wall 1971). The use of different baking procedures apparently complicates the subject and generalization of results.

Obviously, a better understanding of factors involved in the manifestation of quality differences among genotypes in a quality evaluation program is useful. This is especially true for loaf volume, for which methodology differs in most laboratories. The present investigation studied the factors causing different loaf volume potentials (as measured in this laboratory) among the parents and progeny of a cross between the common wheat varieties Coteau and Butte. Field plot variety trial data accumulated by the Department of Cereal Chemistry and Technology in 1975 and 1976 from over six localities per year<sup>3</sup> indicated that Coteau has farinograph characteristics similar to those of Butte, yet shows a higher flour protein content and loaf volume potential.

## MATERIALS AND METHODS

### Wheat Samples

Grain of the varieties Coteau and Butte from experimental plots grown at two localities (Fargo and Casselton, ND) in 1977 and 1978 were bulked and Buhler-milled in each year to provide flour for fractionation and reconstitution studies.

In 1978, bulked samples from replicated plots for 28 F<sub>2</sub>-derived F<sub>4</sub>-lines and from the two parental varieties were Buhler-milled for correlation studies.

### Baking

A 25-g baking method with a 2-hr fermentation period was used. Absorption and mixing time were determined by dough feel and

appearance. The baking formula (on a flour basis) was as follows: flour, 25 g; sugar, 5.0%; salt, 1.0%; yeast, 3.0%; malt, 0.3%; and bromate, variable.

### Pentosan Content

Total pentosan content was estimated according to the method of Dische and Borenfreund as modified by Cracknell and Moye<sup>4</sup> and further modified by MacArthur and D'Appolonia (1977).

### Damaged Starch

The procedure described by Williams and Fegol (1969) was used.

### Fractionation

For both years, Buhler-milled flours of Coteau and Butte were fractionated into gluten, starch, tailings, and water-soluble fractions. Three fractionations were made per variety, and the various subfractions were combined into the composite fractions used for reconstitution. The water-soluble and tailings fractions of the 1978 samples were further subfractionated.

**Flour.** Three hundred grams of flour and 180 ml of water were mixed in a laboratory mixer for 1 min. The gluten was handwashed from the flour using four 500-ml portions of water and a final wash with 300 ml of water. The gluten ball was chilled in a freezer for 25–30 min and then lyophilized. The washings, after removal of the gluten, were centrifuged and the supernatant (water solubles) decanted, shell frozen, and lyophilized. The layer of material on top of the starch (tailings) was removed with a spatula and the starch was reslurried and centrifuged. Additional tailings were removed, combined with the previous tailings, frozen, and lyophilized. The starch was air dried and passed through a 70-mesh sieve; the gluten and tailings were passed through a 40-mesh sieve.

**Tailings.** Fractionation of the tailings was based on the method described by Kulp and Bechtel (1963). The wet tailings fraction was reslurried and passed through a 400-mesh sieve. The overs (high pentosan fraction) and throughs (high starch fraction) were subsequently frozen, lyophilized, and passed through a 40-mesh sieve.

**Water-Solubles.** The procedure was similar to that described by Hoseney et al (1972). Four grams of lyophilized water solubles was dissolved in 100 ml of water. The solution was dialyzed against distilled water at 4°C for three days, with the water changed daily. The globulin precipitate was removed by centrifugation at 1,000 × g. The dialysate, albumin-rich (supernatant), and globulin-rich (insoluble) fractions were subsequently freeze-dried. A pentosan-rich fraction was also prepared by boiling the albumin-rich supernatant. After centrifugation at 1,000 × g, the supernatant was freeze-dried.

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<sup>3</sup>Unpublished data.

<sup>4</sup>R. L. Cracknell and C. J. Moye. 1970. A colourimetric method for the determination of pentosans in cereal products. 20th Annual Conference, Royal Australian Chemical Institution.

## Protein Solubility Fractions

The levels of the various protein fractions in each flour were determined, using the modified Osborne procedure described by Chen and Bushuk (1970). Residue protein contents were also determined using the rapid procedure of Orth and O'Brien (1976). These values were expressed as percentage of the total flour protein.

## Protein Content

The AACC macro-Kjeldahl procedure was used to measure protein.

## Kernel Hardness

The pearling index described by McCluggage (1943) was used to measure kernel hardness.

## Physical Dough Properties

The farinograph was used to measure dough properties, with dough consistency centered on the 540-BU line.

## Flour Reconstitution

From 1977 samples were used to recombine fractions from the two sources in all possible combinations of a 2<sup>4</sup>-factorial experiment (based on the Butte fraction yields). In addition, a parental combination was prepared using the Coteau fractions and fraction yields. No other combinations could be made because of the limited amount of flour. Three replicates of each combination were subsequently baked with 10 ppm bromate.

The 1978 material was used to conduct three experiments.

*Experiment 1.* A 50:50 blend of starches from the two parents were used in a factorial experiment in which the gluten, tailings, and water-soluble fractions were interchanged, based on the Butte fraction yields.

*Experiment 2.* Two composite fractions were prepared using the two sets of gluten, starch, and water solubles and the corresponding Butte fraction yields. These were recombined with the starch-rich and pentosan-rich tailings fractions in a 2<sup>3</sup>-factorial experiment.

*Experiment 3.* A single composite of gluten, starch, and tailings from Coteau was prepared based on the Coteau fraction yields. To this was added recombinations of the albumin, globulin, and dialysate fractions from the water solubles. Fractions were added to combinations in the amounts in which they were isolated.

## Statistical Analyses

The factorial experiments were analyzed assuming a fixed model (Steel and Torrie 1960).

## RESULTS AND DISCUSSION

### Reconstitution Study of 1977

The aim of this experiment was to identify the major flour components housing factors that cause differential loaf volume responses in the two flours. Results of fractionation of the parental flours into gluten, starch, tailings, and water solubles are shown in Table I. The gluten protein contents were relatively low (about 70% on a dry basis) but were similar for both varieties and years and should not have influenced conclusions to a great extent. The analysis of variance of the reconstitution experiment is summarized in Table II.

The loaf volumes of the reconstituted systems (based upon each variety's own fraction yields) were markedly lower than those of the original flours; however, the relative volumes with respect to one another were maintained. The loaf volumes, using 10 ppm bromate with original and reconstituted flours, were 194 and 169.5 cc, respectively, for Coteau and 196 and 169.0 cc, respectively, for Butte.

The reconstituted loaves also had a slightly "older" appearance than the original flours. (The term "old" is used here to distinguish an overoxidized loaf appearance from an underoxidized or "young" appearance.)

The analysis of variance (Table II) indicated that the flour components caused differential effects upon interchange. Three interactions also appeared to be important. In all cases, the component originating from parent Butte had an enhancing effect upon loaf volume (Table III).

### Reconstitution Studies of 1978

Results on fractionations are summarized in Table I. The aim of these experiments was: 1) to see whether the 1978 flours gave more or less the same results as the 1977 flour, 2) to study the tailings effect, and 3) to study the water solubles effect.

As with the 1977 experiments, no significant differences could be detected between the two flours (either unfractionated or reconstituted).

Analyses of variance showed that the only significant effect was associated with the water solubles interchange in Experiment 1.

TABLE I  
Fractionation Data<sup>a</sup> on Wheat Varieties Butte and Coteau

Flour and Fractions	Yield (percent of total flour)		Contents						
	Butte	Coteau	Protein (%)		Pentosan (%)		Damaged Starch (absorbance × 10)		
			Butte	Coteau	Butte	Coteau	Butte	Coteau	
1977 material									
Gluten	0.20	0.23	62.5	59.4	...	...	...	...	...
Starch	0.39	0.43	0.1	0.1	...	...	...	...	...
Tailings	0.36	0.29	0.7	0.8	2.3	2.8	...	...	...
Water solubles	0.05	0.05	24.9	23.7	10.6	10.6	...	...	...
Unfractionated flour	100.0	100.0	14.1	15.0	1.4	1.4	6.65	4.84	...
1978 material									
Gluten	0.20	0.21	62.4	61.9	...	...	...	...	...
Starch	0.42	0.43	0.3	0.3	...	...	3.16	2.59	...
Tailings	0.33	0.30	0.7	0.8	2.1	2.2	...	...	...
Starch-rich	0.31	0.29	0.6	0.5	0.7	1.0	4.04	4.89	...
Pentosan-rich	0.02	0.02	3.7	4.0	25.2	27.2	16.02	13.92	...
Water solubles	0.049	0.050	24.5	23.6	9.5	8.8	...	...	...
Albumin-rich <sup>b</sup>	0.018	0.018	49.8	46.7	24.7	25.6	...	...	...
Globulin-rich <sup>b</sup>	0.001	0.001	55.5	53.7	...	...	...	...	...
Dialysate <sup>b</sup>	0.030	0.031	8.8	10.2	...	...	...	...	...
Pentosan-rich <sup>b</sup>	0.017	0.016	48.1	38.5	26.8	31.3	...	...	...
Unfractionated flour	100.0	100.0	14.1	14.9	1.3	1.2	...	...	...

<sup>a</sup>14% mb, except where noted.

<sup>b</sup>As is basis.

Part of this may be related to the fact that fewer observations were involved in this set of experiments. However, other explanations are apparent.

### Conclusions from the Reconstitution Studies

The 1977 parental recombinations of flour fractions based on Butte fraction yields resulted in a loaf volume of 167.7 cc for Butte and 157.7 cc for Coteau (data not shown). This difference exceeds the 5% least significant difference of 7.54. Because reconstitution to each parent's own fraction yields resulted in about the same loaf volumes for the parental combinations, we concluded that the differential responses of the fractions can largely be attributed to the fact that an isolated Coteau fraction was not reconstituted in its natural amount.

Table I shows that when the Butte yield was applied, gluten contributed 12.5% protein to the factorial combinations, whereas Coteau gluten contributed 11.9%. Thus the small main effect associated with the gluten may result from the larger amount of gluten protein included in combinations receiving Butte gluten.

**TABLE II**  
Analysis of Variance of a Factorial Experiment<sup>a</sup> (1977 Material)

Source of Variation	Degrees of Freedom	Mean Square
Replications	2	57.90
Treatments	15	93.98 <sup>b</sup>
S	1	123.52 <sup>c</sup>
G	1	368.52 <sup>b</sup>
S × G	1	0.52
T	1	99.19
S × T	1	1.02
G × T	1	46.02
S × G × T	1	4.69
W	1	180.19 <sup>b</sup>
S × W	1	1.69
G × W	1	295.02 <sup>b</sup>
S × G × W	1	4.69
T × W	1	93.52 <sup>c</sup>
S × T × W	1	11.02
G × T × W	1	130.02 <sup>c</sup>
S × G × T × W	1	50.02
Error	30	20.45
Total	47	...

<sup>a</sup> S = starch, G = gluten, T = tailings, W = water soluble.

<sup>b</sup> P = 0.01.

<sup>c</sup> P = 0.05.

**TABLE III**  
Average<sup>a</sup> Volumes (cc) from Coteau and Butte Flour Fractions in Reconstituted Flours, 1977 Factorial Experiment

Fraction	From Variety	
	Coteau	Butte
Starch	164.8	168.0
Gluten	163.6	169.1
Tailings	164.9	167.8
Water solubles	164.4	168.3

<sup>a</sup> Of 24 loaves.

**TABLE IV**  
Bromate Responses of Unfractionated Coteau and Butte Flours, Measured as Loaf Volume Changes (cc)

Season	Variety	Bromate Level (ppm)			
		0	10	20	30
1977	Butte	188	196	191	...
	Coteau	180	194	220	...
1978	Butte	192	206	215	199
	Coteau	179	209	218	235

The conclusion that a gluten quality difference is not involved is confirmed by Experiment 1 on the 1978 material. In this case, the Butte gluten fraction contributed 12.5% protein to factorial combinations and gave an average loaf volume (eight loaves) of 207.9 cc. The Coteau gluten fraction contributed 12.4% protein and gave an average of 207.1 cc.

In the 1977 experiment, Butte starch fractions probably contributed more damaged starch than did Coteau starch, which probably accounts for the beneficial effect associated with the Butte starch fraction. This wheat had a somewhat higher pearling index than did Coteau (62.5 and 61.0, respectively) and also showed a higher degree of starch damage upon milling (Table I).

In the 1977 experiment, the Butte tailings contributed 0.83% pentosans and Coteau tailings 1.01% pentosans to the factorial combinations. On the other hand, the Butte tailings probably contributed more damaged starch. Previous studies (Ali 1978, Patil et al 1976) showed pentosans to have an oxidizing effect in baking. The present differences in pentosan content did not cause visible differences in internal and external loaf appearances, which were probably too small to be noticeable. The small beneficial effect of the Butte tailings fraction may be the result of an adverse pentosan effect or a beneficial starch effect or some other factor. No significant tailings effect was observed in the 1978 experiments. Fractionation of the tailings into pentosan-rich and starch-rich subfractions and interchanging these subfractions in the Butte ratios caused little variation as far as percent pentosan and damaged starch contributions in the factorial combinations are concerned (Table I).

Water solubles were the only components showing significant effects in both years. Table I shows that the amounts of water-soluble pentosan and protein added to factorial combinations through the inclusions of both sources of water solubles were rather similar. In both years, the Butte water solubles caused an enhancing effect on loaf volumes. The factorial experiment involving the fractions albumin, globulin, and dialysate failed to indicate a source for such a difference. Crude water-soluble pentosan preparations of the two parents were also interchanged against a reconstituted Coteau background (gluten, starch, tailings, globulin, and dialysate) but failed to show any differences in loaf volume. Rather, removal of some of the albumin fraction appeared to cause more "oldness" in the loaves, thus implicating these

**TABLE V**  
Characteristics Measured in Correlation Study and Ranges

Characteristic	Range
Contents (%)	
Protein	13.6–15.8 <sup>a</sup>
Albumin	11.9–14.0
Globulin	2.4–3.3
Gliadin	25.9–34.4
Glutenin	9.0–20.4
Residue protein	25.9–40.7
	22.2–29.1 <sup>b</sup>
Ratio	
Albumin/globulin	3.82–5.53
Gliadin/glutenin	1.41–3.53
Total pentosan content (%) <sup>a</sup>	0.92–1.32
Bromate requirement (ppm)	0–40

<sup>a</sup> 14% moisture basis.

<sup>b</sup> Determined by the Orth and O'Brien procedure (1976).

**TABLE VI**  
Stepwise Regression Analysis

Characteristic	Variables in Model	r
Bromate requirement	Albumin/globulin ratio (–)	0.53
Loaf volume		
10 ppm bromate	Protein content (+)	0.50
Optimum bromate	Protein content (+), glutenin content (–), Orth and O'Briens' residue protein (–)	0.81

proteins in the bromate reaction. If the averages obtained in this factorial are considered, however, the superiority of the Butte water-solubles appears to be the result of cumulative small effects provided by the various subfractions. None of these effects is strong enough to cause a significant effect under the conditions of the test, but the Butte fractions consistently cause small positive effects. The two parental combinations differ significantly in loaf volume (5% level), further substantiating this observation.

### Bromate Response

Apart from these main effects and interactions, we also observed that the inclusion of different water-soluble and gluten fractions markedly affected the internal and external loaf appearance. Combinations receiving Butte water solubles and gluten had an old appearance, and those receiving the corresponding Coteau fractions had a young appearance. Interchanges of these fractions resulted in slightly old loaves. Thus, the Butte water-soluble and gluten fractions resulted in an overoxidation effect from the fixed bromate addition, whereas the inclusion of the corresponding Coteau fractions seems to have resulted in higher bromate requirements. No such differences could be detected upon interchange of the water-soluble subfractions (1978 study). However, this might have resulted from the smaller differences encountered in this material. This differential bromate response was also exhibited by unfractionated flours of both wheats (Table IV).

At the 10-ppm bromate level applied in the preceding set of experiments, essentially no differences could be observed in loaf volume. However, at optimum bromate levels Coteau had a higher loaf volume than Butte.

### Correlation Studies

In order to further investigate the factors that govern loaf volume responses, 28 F<sub>2</sub>-derived F<sub>4</sub> lines showing diverse loaf volume responses were selected from the progeny of these two varieties. Pup loaves at 0, 10, 20, 30, and 40-ppm bromate levels were baked. The bromate levels producing optimum loaf characteristics were subsequently determined. In addition, the protein of each line was separated into five solubility classes according to a modified Osborne technique (91% or more of the total protein was recovered). Further measurements on these samples included total pentosan content and Orth and O'Brien's (1976) residue protein content. The range of results obtained is given in Table V.

These data were subsequently used to determine by stepwise regression the best fitting models for prediction of loaf volume and bromate requirement. Only variables contributing significantly (5% level) to the regression sum of squares were retained in the final models (Table VI).

### SUMMARY

With regard to this study, the following conclusions can be drawn. When baked with 10-ppm bromate, the two varieties Coteau and Butte have very similar loaf volumes. If individual fractions are compared, differential responses result, probably from the amount rather than the type of gluten. Butte, being a harder wheat, produced more damaged starch upon milling, probably accounting for the beneficial effects observed with the Butte starch and tailings fractions. The beneficial effect of the Butte water solubles apparently resides in all three soluble subfractions rather than in one specific subfraction. However, all these effects are nullified upon reconstitution to original fraction yields. The water-soluble and gluten fractions probably affect the bromate requirements in this material.

A correlation study associated the albumin and globulin with bromate requirements; however, this could account for only 28% of the total variation. Hoseney et al (1972) found that phosphoric acid in the pH 6.1-soluble gluten was involved in the bromate reaction and also that the pH 6.1-nondialyzable gluten proteins affected the bromate requirement. Thus the present chemical determinations apparently do not accurately monitor the majority of factors

involved in this situation. No evidence indicates that the small differences in pentosan content exerted any significant oxidative effects.

We also conclude that when this baking procedure is used in conjunction with optimized bromate levels, the acetic acid-soluble and insoluble glutenins become a limiting factor in the expression of loaf volume. Orth and Bushuk (1972) found glutenin content to correlate negatively and residue protein content to correlate positively with loaf volume using the remix baking test. Hoseney and Finney (1971) concluded that gliadin rather than glutenin controlled loaf volume potential. MacRitchie (1980) found that the more easily extractable glutenin appears to be more effective than the residue protein in promoting loaf volume. In the present study, a correlation of -0.38, significant at the 5% level, was observed between residue protein content and loaf volume (at optimum bromate). The baking test used in this study employs markedly less mixing and fermentation than does the remix method. It also has a shorter fermentation time than the method used by Hoseney and Finney (1971). Therefore, the residue protein may promote loaf volume under the conditions of the remix test but be limiting to loaf volume at certain levels under the mild conditions of the baking test employed.

The fact that related material was involved in the correlation study might have introduced some bias in the conclusion.

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