Analysis of Relationships Between Flour Quality Properties and Protein Fractions in a World Wheat Collection

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ABSTRACT

The relationships among and between physical dough properties, baking and related quality properties, and protein fractions obtained by solvent fractionation and gel filtration chromatography have been analyzed for a world wheat collection of 33 hard-textured cultivars. Strong significant correlations were obtained among farinograph measurements and among baking quality measurements using four baking procedures, but little or no relationship was obtained between dough strength and baking quality parameters. Nonsignificant correlations were obtained between farinograph properties of nonfermented doughs and remix peak mixing time, which provides an indication of fermented dough strength. Protein content was highly correlated to loaf volumes and to loaf volumes per unit of protein. Sodium dodecyl sulfate sedimentation value and glutenin (Glu-1) score were generally poor predictors of dough strength and baking quality. However, Glu-1 score was strongly correlated to remix peak loaf volume and to loaf volume per unit of protein, where mixing and oxidation requirements were optimized. Correlation and principal component analysis showed that protein content and the amount of peak 3 lower-molecular-weight gliadins, a redundant protein content predictor, were most closely associated with loaf volume, whereas salt-soluble flour proteins were most closely associated with farinograph properties and loaf volumes per unit of protein for the two AACC modified straight-dough procedures. Osborne residue and acetic acid-insoluble protein fractions and Glu-1 score were most closely associated with fermented dough strength (remix peak mixing time) and remix peak loaf volume per unit of protein.

The physical dough properties and breadmaking properties of wheat flours are primarily determined by their proteins. Protein content has long been known to be closely related to loaf volume, a primary measure of baking quality (Finney and Barmore 1948). Its influence upon dough strength properties is less evident, although in general a positive relationship between these properties is obtained (Fowler and DeLaRoche 1975). At present, most evidence suggests that physical dough properties, especially those associated with dough strength, and baking properties are mainly determined by the qualitative and quantitative properties of the glutenin proteins. This quantitative relationship has been demonstrated by reconstitution studies (Lee and MacRitchie 1971; MacRitchie 1973, 1987), solvent fractionation studies (Pomeranz 1965, Orth and Bushuk 1972, Orth and O'Brien 1976), and molecular weight fractionation by gel filtration (Huebner and Wall 1976, Singh et al 1990) and high-pressure liquid chromatography (Huebner and Bietz 1985, Dachkevitch and Autran 1989, Singh et al 1990). In most cases, the proportion or absolute quantity of glutenin, and in particular, the proportion of high-molecular-weight or insoluble glutenin was found to be positively related to dough strength properties and/or baking quality. However, these relationships are influenced by extraction conditions (Singh et al 1990). In several studies the proportion of solubilized or high-molecular-weight glutenin was found to be negatively related to dough strength properties (Orth and Bushuk 1972, Bietz 1986). The qualitative relationship between various glutenin subunits and dough strength and/or baking properties has also been demonstrated by electrophoresis (Payne et al 1979, 1981) and high-pressure liquid chromatography (Huebner and Bietz 1985, Wieser et al 1989). This work has resulted in the development of a glutenin subunit scoring system (Glu-1 score) to predict these quality parameters (Payne et al 1987).

Studies have also shown that gliadin protein fractions appear to be related to both dough strength properties and baking quality. Both quantitative and qualitative properties of gliadins have been shown to be associated with dough strength properties (Hamada et al 1982, Wrigley et al 1982, Branlard and Dardevet 1985, MacRitchie 1987). These proteins may also play an important role in determining baking quality (Branlard and Rousset 1980, Huebner and Bietz 1986). Reconstitution studies by Finney and co-workers demonstrated that gliadin proteins are mainly responsible for the control of loaf volume (reviewed by Hoseney and Finney 1971 and Finney 1985). Salt-soluble proteins (albumins and globulins) also appear to be related to these quality parameters (Marais and D'Appolonia 1981a,b; MacRitchie 1987; Singh et al 1990).

The differences in the relative influence of flour protein fractions and individual protein subunits upon physical dough and baking quality evident in the above studies can be attributed to a number of factors. As noted above, extraction conditions used to isolate various fractions can dramatically change the relationships (correlations) between these fractions and quality parameters. Similarly, the choice of methodology used to measure quality parameters can have a strong effect upon results (Marais and D'Appolonia 1981a,b; Hamada et al 1982; Campbell et al 1987). Relationships between and among protein fractions and quality parameters may also be strongly influenced by the choice of samples. In many of the studies cited above, relationships may be biased due to insufficient numbers of samples or the inclusion of soft, low-protein wheats of poor baking quality and weak dough properties that would normally not be included in bread-wheat breeding programs, as pointed out by Wrigley et al (1982) and Branlard and Dardevet (1985).

The objective of the present study was to determine the interrelationships among and between flour quality parameters (with emphasis on physical dough and baking properties) and protein fractions while minimizing the effects of the factors discussed above. To meet this objective, 33 wheat samples with hard kernel texture were selected from a world wheat collection and subjected to a wide range of quality testing, protein solubility and molecular weight fractionation techniques and other quality prediction tests. Data were assessed by correlation and principal component analysis (PCA). The latter technique was included because of its ability to reduce the complexity of data sets to a small number of independent (orthogonal) principal components representing linear combinations of the original variables. This approach allows an assessment of the association between groups of variables and a more basic understanding of the primary components contributing to the underlying variability of a data set.

MATERIALS AND METHODS

Seed Source

Thirty-three wheat samples, grown in 14 countries (as described below), were selected on the basis of their visual appearance, hardness, and electrophoretic homogeneity from a collection

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maintained by the Canadian International Grains Institute, Winnipeg. All samples that showed visually sound kernel characteristics, had particle size index values less than 63 (indicating hard-kernel types), and showed uniform endosperm protein composition as assessed by single-kernel acid (Lukow et al 1990) and sodium dodecyl sulfate (SDS) (Lukow et al 1989) polyacrylamide gel electrophoresis were included. Selection of samples on the basis of the two former parameters was designed to reduce variation in physical dough and baking qualities related to differences in milling efficiency. Exclusion of softer wheats was also designed to reduce bias in the statistical analysis. Soft wheats, which normally are bred for low protein content and weak dough properties, would tend to create a high concentration of data points over a narrow range for these parameters.

The sample set consisted of two varieties grown in Argentina (Klein Toledo and Dekalb Lapacho), four grown in Canada (Neepawa, Katepwa, and two NB311 selections), one grown in Bulgaria (Sadova #1), two grown in Czechoslovakia (Sava and Jubilejni), four grown in Hungary (MV3, MV4, Bezostaya, and Jubejnaja), one grown in Israel (Miriam), four grown in New Zealand (Aeota, Takahe, Hilgonoorf, and Kamaru), six grown in Pakistan (SA75, Sandel3, Chenab70, Pari73, Lyp73, and Punjab86), one grown in Portugal (Siete Cerros), one grown in Syria (Mexican), one grown in Turkey (Bezostaya), three grown in England (Bouquet, Flinor, and Maris Huntsman), two grown in Uruguay (LE505 and Dakuru), and one grown in West Germany (Kolibri). These varieties include spring, winter, and indeterminate growing types. The countries where the varieties were grown are not necessarily their countries of origin but represent regions in which they are well adapted agronomically.

Quality Testing

Wheat moisture, kernel weight, and wheat ash were determined as described by Preston et al (1988). Wheat protein was determined by a modified Kjeldahl method (Williams 1973). SDS sedimentation values were obtained by the procedure of Preston et al (1982a). Wheat hardness was assessed by the particle size index method using a Udy Cyclone grinder (Udy Corp., Fort Collins, CO) as described by Williams and Sobering (1986) and by grinding time as described by Kosmolak (1978). With both these methods, higher values indicate softer kernel texture. Glu-1 score was determined by the system of Payne et al (1987) from SDS polyacrylamide gel electrophoresis patterns obtained as described by Lukow et al (1989).

Milling was done on a Buhler pneumatic laboratory mill after cleaning and tempering wheat samples to 16.5% moisture (Black et al 1980).

Flour moisture, protein, ash, color, gassing power, and amylograph peak viscosity were determined as previously described (Preston et al 1988). Starch damage was determined on a 5-g flour sample by the method of Farrand (1964).

Farinograph properties were obtained using a method similar to AACC Method 54-21 (AACC 1983), as described by Preston et al (1982b). Remix (constant 2.5 min remix time) and remix peak baking properties were assessed by the procedure of Kilborn and Tipples (1981). AACC baking properties were determined with 10 and 0 ppm bromate by the modified procedure of Lukow et al (1990). Loaf volumes for all baking procedures were determined by rapeseed displacement from loaves made from 100 g of flour.

All wheat test results were calculated on a 13.5% moisture basis and all flour test results were reported on a 14.0% moisture basis. All analyses were made in duplicate.

Flour Protein Extraction

Flour protein extractability (in duplicate) in 0.05*M* acetic acid (HAc) or 1.0*M* sodium thiocyanate (NaSCN) was determined by mixing 3.0 g of flour with 25 ml of solvent in a 50-ml capped centrifuge tube on a vortex mixer. The tube was then rotated for 60 min at room temperature on a rotator at 50 rpm and centrifuged at 40,000 × *g* for 15 min at 10°C. The pellet was suspended in 25 ml of solvent and the extraction process repeated. A third extraction was made on the resulting pellet with an additional 25 ml of solvent. Supernatants were combined and dialyzed against water to remove small peptides and amino acids (and NaSCN, if necessary), then freeze-dried. Pellets (insolubles) were also dialyzed against water and freeze-dried.

Flours were fractionated (in duplicate) by a modified Osborne procedure (Chen and Bushuk 1970). Fractions included salt-extractable (0.5*M* NaCl), 70% ethanol-extractable, 0.05*M* HAc-extractable and nonextractable (residue) proteins.

The protein content of fractions ($N \times 5.7$) was determined (in duplicate) by digestion of samples with sulfuric acid using selenium catalyst in 100-ml digestion tubes placed in a Tecator System 12 digestion unit (Tecator AB, Hoganas, Sweden). Digests were made up to 100 ml with water and analyzed by automated measurement of ammonium ion with alkaline sodium phenate in the presence of sodium hypochlorite, as described by Mitcheson and Stowell (1970). Recoveries of fractions are reported as percent of total recovery.

Gel Filtration Chromatography

Gel filtration of 1.0*M* NaSCN-extractable flour proteins was performed with Sephacryl S-300 superfine (Pharmacia) in a 2.5- × 100-cm column using 2.0*M* NaSCN as eluant at an upward flow rate of 35 ml/hr (Preston 1984). Samples were prepared from single extracts (3.0 g of flour and 25 ml of solvent) as described above. Peak areas were determined by absorbance at 280 nm as outlined by Salomonsson and Larsson-Raznikiewicz (1985). Each sample was extracted and subjected to column analysis in duplicate. Total recovery of fractions ranged from 93 to 101%, based upon the absorbance of the initial extract. Recoveries of fractions are reported as percent of total recovery.

Analysis of Data

Correlation (parametric) analysis and PCA (using the correlation matrix technique) were done using SAS procedures (SAS 1987) on a 286 microcomputer. Parameters with loading factors equal to or greater than 0.25 were considered to be important contributors to each principal component (Sinha 1977). Spearman's rank-order correlation procedure was used to determine correlations between Glu-1 score, a nonparametric measure, and other parameters. For PCA analysis, Glu-1 score was treated as a numeric variable to estimate its effect.

RESULTS

Table I gives codes, means, standard deviations, coefficients of variability, and ranges for quality tests and protein fractionation results for the 33 wheat samples. Wide variation was evident in most of the quality parameters measured. This variation can probably be attributed to the presumably wide genetic diversity of the wheat varieties and to the fact that they were grown in different locations (with differences in growth habit and adaptability). The former property should maximize the "robustness" of the sample set by reducing the number of genetically closely related varieties. However, as pointed out by Huebner (1989), differences in environmental growing conditions would tend to reduce the degree of association among measured parameters.

Interrelationships Among Quality Parameters

Correlation coefficients among quality parameters are given in Table II. Dough strength was assessed by farinograph measurements, including dough development time, mixing tolerance index, and stability. Dough strength was also determined by measurement of the optimum (peak) mixing time of full-formula fermented doughs for the remix peak baking procedure. The farinograph measurements were highly intercorrelated ($P < 0.01$), with correlation coefficients varying from plus or minus 0.70 to 0.84 (Table II). Flour protein content was significantly correlated to these farinograph parameters, but values were much lower. However, surprisingly, none of these measurements was significantly correlated to remix peak mixing

TABLE I
Means, Standard Deviations (SD), Coefficients of Variability (CV), and Ranges for Flour Quality and Protein Fractionation Values of 33 World Wheat Samples[a]

Attribute	Code	Mean	SD	CV	Range
Protein, %	FPRO	11.2	1.6	14.3	8.2–14.1
Farinograph values					
Absorption, %	FABS	58.3	3	5.1	52.4–65.2
Dough development time, min	FDDT	6.15	5.17	84.1	1.0–24.0
Mixing tolerance index, BU	FTOL	39.7	24.1	60.7	5–110
Stability, min	FSTAB	11.4	8	70.2	1.5–30.5
Remix baking values					
Loaf volume, cm^3	RVOL	591	132	22.3	340–845
Per unit of protein	RPRO	52.8	8.2	15.5	34.0–69.4
Peak loaf volume, cm^3	PVOL	662	142	21.5	330–875
Per unit of protein	PPRO	59.2	9.5	16	33.7–73.2
Peak mixing time, min	PTIME	3.6	1	27.8	1.8–6.3
AACC baking test values					
Loaf volume (10 ppm bromate), cm^3	BRVOL	558	131	23.5	300–905
Per unit of protein	BRPRO	49.9	7.8	15.6	36.6–66.0
Loaf volume (0 ppm bromate), cm^3	NOVOL	609	100	16.4	370–820
Per unit of protein	NOPRO	54.6	5.3	9.7	44.8–67.1
Insoluble protein					
1.0*M* NaSCN,[b] %	NASCN	36.8	4.2	11.4	29.8–47.2
0.05*M* Acetic acid,[b] %	HAC	28.2	4.1	14.5	20.4–38.5
Gel filtration					
Peak 1,[c] %	PK1	28.8	4.5	15.6	16.3–39.6
Peak 2,[c] %	PK2	37.2	4	10.8	29.3–44.2
Peak 3,[c] %	PK3	23.8	3.8	16.0	15.9–31.6
Peak 4,[c] %	PK4	5.1	1.6	31.4	2.4–9.4
Peak 5,[c] %	PK5	1.9	1.3	68.4	0.4–6.5
Peak 6,[c] %	PK6	3.2	2.4	75.0	0.1–12.4
Ratio PK1:PK2	RAT1	0.79	0.18	22.8	0.46–1.21
Ratio PK1:PK3	RAT2	1.25	0.31	24.8	0.52–1.89
Ratio PK2:PK3	RAT3	1.62	0.41	25.3	1.1–2.7
"Osborne" fractions					
Salt-soluble,[b] %	SALT	17.1	2.3	13.5	13.4–24.7
70% Alcohol-soluble,[b] %	GLI	34.1	3.6	10.6	26.1–40.7
0.05*M* Acetic acid-soluble,[b] %	GLU	8.9	1.8	20.2	5.3–12.1
0.05*M* Acetic acid-insoluble,[b] %	RES	39.9	3.1	7.8	33.3–45.6
Ratio SALT:GLI	SAGI	0.51	0.1	19.6	0.33–0.82
Ratio SALT:GLU	SAGU	2.01	0.58	28.9	1.23–3.95
Ratio SALT:RES	SARES	0.43	0.08	18.6	0.34–0.74
Ratio GLI:GLU	GIGU	4.03	1.1	27.3	2.2–7.3
Ratio GLI:RES	GIRES	0.87	0.14	16.1	0.59–1.21
Ratio GLU:RES	GURES	0.22	0.05	22.7	0.13–0.31
SDS sedimentation volume, ml	SDS	43.5	13.3	30.6	23.0–82.5
GLU-1 score	GSCORE	7.9	2.1	26.6	4.0–10.0

[a]All quality data corrected to 14.0% moisture basis.
[b]Values represent percent of nitrogen recovery (micro-Kjeldahl).
[c]Values represent percent of total recovery at 280 nm.

TABLE II
Correlation Coefficients Among Dough Strength, Baking Quality Parameters, and Flour Protein Content for 33 World Wheat Flour Samples[a,b]

	FDDT	FTOL	FSTAB	PTIME	RVOL	PVOL	BRVOL	NOVOL	RPRO	PPRO	BRPRO	NOPRO	FPRO	SDS	GSCORE[c]
FDDT	1.00	−0.70**	0.84**	...	...	0.43*	...	...	...	...	...	−0.42*	0.51**	...	0.43*
FTOL		1.00	−0.83**	...	...	−0.49**	...	...	...	...	...	...	−0.46**	−0.39*	...
FSTAB			1.00	...	...	0.38*	...	...	...	...	...	...	0.36*	0.37*	...
PTIME				1.00	...	...	...	...	...	0.50**	...	...	...	...	0.38*
RVOL					1.00	0.82**	0.89**	0.90**	0.79**	0.51**	0.66**	0.50**	0.71**	0.39*	0.42*
PVOL						1.00	0.75**	0.77**	0.57**	0.77**	0.47**	...	0.71**	0.41*	0.78**
BRVOL							1.00	0.95**	0.61**	0.38*	0.79**	0.53**	0.74**	...	0.45**
NOVOL								1.00	0.57**	0.35*	0.66**	0.52**	0.82**	...	0.37**
RPRO									1.00	0.67**	0.77**	0.78**	...	...	...
PPRO										1.00	0.49**	0.48**	...	...	0.63**
BRPRO											1.00	0.86**	...	...	...
NOPRO												1.00	...	...	...
FPRO													1.00	...	0.45*

[a]Correlation coefficients are shown if significant at the 5% ($P < 0.05$) or 1% ($P < 0.01$) level. The significance is indicated by * for the 5% level and ** for the 1% level.
[b]FDDT = farinograph dough development time, FTOL = farinograph mixing tolerance index, FSTAB = farinograph stability, PTIME = remix peak mixing time, RVOL = remix loaf volume, PVOL = remix peak loaf volume, BRVOL = AACC loaf volume (10 ppm bromate), NOVOL = AACC loaf volume (0 ppm bromate), RPRO = remix loaf volume per unit of protein, PPRO = remix peak loaf volume per unit of protein, BRPRO = AACC loaf volume (10 ppm bromate) per unit of protein, NOPRO = AACC loaf volume (0 ppm bromate) per unit of protein, FPRO = flour protein, SDS = sedimentation by sodium dodecyl sulfate electrophoresis, GSCORE = glutenin score.
[c]Correlation coefficients between glutenin score and other parameters were calculated by Spearman's rank-order technique.

time, a measure presumably related to fermented dough strength.

Baking quality was assessed by measurement of loaf volume and loaf volume per unit of protein, using four straight-dough procedures (remix, remix peak, AACC-10 ppm bromate, and AACC-0 ppm bromate). The former procedures involve a remixing stage after fermentation, which aids in the dough development of strong flours and optimizes oxidation response (Kilborn and Tipples 1981). Highly significant positive correlations ($P < 0.01$) were obtained for loaf volume among the four procedures, as shown in Table II. Flour protein was also closely related to loaf volume, although correlation values (r) were lower than those obtained between baking procedures. This result is consistent with previous studies (Finney and Barmore 1948) showing the close relationship between protein content and loaf volume for individual varieties or classes of wheat. Correlations between loaf volume per unit of protein were also highly significant ($P < 0.01$) and positive for the four baking methods. However, correlation coefficients between remix peak values and those obtained by the AACC procedures were relatively low in magnitude ($r < 0.5$), reflecting differential response among varieties to baking procedure. Interrelationships between loaf volume and loaf volume per unit of protein were also highly significant in most cases, showing that protein quality, as well as quantity, was important in determining loaf volume.

Correlations between dough strength properties and baking quality parameters were nonsignificant or were significant but with r values less than 0.5. Significant positive values included those between remix peak loaf volume and farinograph dough development time and stability (negative for mixing tolerance index) and that between remix peak loaf volume per unit of protein and remix peak mixing time. A significant negative correlation was obtained between loaf volume per unit of protein with the AACC no-bromate procedure and farinograph dough development time. This negative relationship may be the result of lower oxidation requirements for the doughs mixed for shorter times.

SDS sedimentation volume, which has previously been used to predict or assess dough strength and baking quality (Axford et al 1979, Payne et al 1987), gave significant positive correlations ($P < 0.05$) with farinograph stability and remix loaf volume and peak loaf volume. However, correlation coefficients were very low ($r < 0.41$), indicating poor predictability. The Glu-1 score showed low but significant positive correlations with flour protein content, farinograph dough development time, remix peak time, and remix and AACC (0 and 10 ppm) bread volumes. However, much higher positive correlations were obtained with remix peak volume ($r = 0.78$, $P < 0.01$) and remix peak volume per unit of protein ($r = 0.63$, $P < 0.01$).

Parameters related to milling (kernel weight, milling yield, wheat and flour ash, and color) were not significantly correlated with any of the dough strength or baking quality data. Tests related to α-amylase (amylograph peak viscosity and gassing power) also showed no apparent relationship to these parameters. Thus, no bias appears to have been introduced into the data set from milling quality or amylase activity (data not shown). Quality parameters related to kernel texture (particle size index, grinding time, and flour starch damage) showed low but generally significant ($P < 0.05$) relationships to a number of the dough strength and baking quality results (data not shown). These significant correlations are probably related to breeding methodology, in which hard wheats tend to be selected for high dough strength and superior baking quality, whereas many softer or semihard wheats are selected on the basis of other parameters. As expected, wheat protein content was closely related to flour protein content ($r = 0.98$) and showed correlations with quality measurements similar to those of the latter (data not shown).

The relationships among the quality parameters measured for the 33 wheat samples were evaluated by PCA to determine the source of the underlying variability. For dough strength, baking, and flour protein parameters, the first three principal components accounted for 86.6% of the variability (Table III). The first principal component, representing 46.3% of the variability, was closely related to all the baking parameters and flour protein, as indicated by loading factors (a measure of association between the parameters and the principal component) of 0.25 or higher. This result is consistent with the highly significant correlations

TABLE III
Principal Component (PC) Analysis of Flour Quality Data for 33 World Wheat Flour Samples[a]

PC 1 (46.3%)		PC 2 (26.3%)		PC 3 (14.0%)	
Quality[b] Test	Loading[c] Factor	Quality[b] Test	Loading[c] Factor	Quality[b] Test	Loading[c] Factor
RVOL	0.39	FDDT	0.49	PTIME	0.60
BRVOL	0.38	FSTAB	0.45	PPRO	0.55
NOVOL	0.38	FTOL	−0.44	FPRO	−0.36
PVOL	0.36	NOPRO	−0.34		
RPRO	0.32	BRPRO	−0.30		
BRPRO	0.31	FPRO	0.25		
FPRO	0.27				
PPRO	0.26				
NOPRO	0.25				

[a]Principal components representing over 10% of the variability of the data set shown.
[b]RVOL = remix loaf volume, BRVOL = AACC loaf volume (10 ppm bromate), NOVOL = AACC loaf volume (0 ppm bromate), PVOL = remix peak loaf volume, RPRO = remix loaf volume per unit of protein, BRPRO = AACC loaf volume (10 ppm bromate) per unit of protein, FPRO = flour protein, PPRO = remix peak loaf volume per unit of protein, NOPRO = AACC loaf volume (0 ppm bromate) per unit of protein, FDDT = Farinograph dough development time, FSTAB = farinograph stability, FTOL = farinograph mixing tolerance index, PTIME = remix peak mixing time.
[c]Loading factors (eigenvectors) of 0.25 or higher shown (positive or negative values).

TABLE IV
Correlation Coefficients Between Protein Fractions and Dough Strength Parameters, Flour Protein, Sodium Dodecyl Sulfate Sedimentation, and Glutenin Score for 33 World Wheat Samples[a,b]

	FDDT	FTOL	FSTAB	PTIME	FPRO	SDS	GSCORE[c]
NASCN				0.42*			
HAC				0.65**			0.38*
PK1							
PK2					0.63**	0.38*	
PK3					−0.69**		−0.38*
PK4							
PK5							
PK6							0.36*
RAT1					−0.40*		
RAT2					0.36*		
RAT3					0.75**		
SALT	−0.38*	0.60**	−0.42*				
GLI							
GLU							
RES				0.50**			0.56**
SAGI		0.42**					
SAGU	−0.38*	0.52**	−0.44*				
SARES		0.58**	−0.38*				−0.36*
GIGU							
GIRES							−0.47**
GURES				−0.46**			

[a]Correlation coefficients are shown if significant at the 5% ($P < 0.05$) or 1% ($P < 0.01$) level. The significance level is indicated by * for the 5% level and ** for the 1% level.
[b]FDDT = Farinograph dough development time, FTOL = farinograph mixing tolerance index, FSTAB = farinograph stability, PTIME = remix peak mixing time, FPRO = flour protein, SDS = sedimentation by sodium dodecyl sulfate electrophoresis, GSCORE = glutenin score, NASCN = NaSCN-insoluble protein, HAC = HAc-insoluble protein, PK = gel filtration peak, RAT1 = ratio PK1:PK2, RAT2 = ratio PK1:PK3, RAT3 = ratio PK2:PK3, SALT = salt-soluble protein, GLI = gliadin protein, GLU = glutenin protein, RES = residual protein, SAGI = ratio SALT:GLI, SAGU = ratio SALT:GLU, SARES = ratio SALT:RES, GIGU = ratio GLI:GLU, GIRES = ratio GLI:RES, GURES = ratio GLU:RES. See Table I for more exact definitions.
[c]Correlation coefficients between glutenin score and other parameters were calculated by Spearman's rank-order technique.

among baking parameters and their close relationship to protein content discussed above. The second principal component accounted for 26.3% of the variability. High loading factors were obtained for farinograph dough strength measurements. Loaf volumes per unit of protein for the AACC baking tests also gave loading factors above 0.30, indicating a qualitative relationship with dough strength. This relationship was not readily evident with correlation coefficients, probably due to the overriding quantitative effect of protein content on baking quality. Protein content also contributed to the second principal component, indicating a qualitative relationship between protein content and dough strength, consistent with the significant positive correlation coefficients between these parameters.

The third principal component represented 14.0% of the variability. Very high loading factors were obtained for remix peak time and loaf volume per unit of protein for the remix peak method. Flour protein also showed a high loading factor indicating, as with the second component, a qualitative relationship. The qualitative relationship of protein content to the other parameters may be a result of breeding strategies in which bread wheats are selected for both high quality and high protein.

When SDS sedimentation values and Glu-1 scores were added to the PCA data set, the former did not show any apparent effect. In contrast, the Glu-1 score showed loading factors of 0.25 for both the first and third components. This result is consistent with the significant correlation coefficients obtained between Glu-1 score and quality parameters present in the first, and in particular, the third principal component. No effect of Glu-1 score upon the second component was evident; this component contained high loading factors for farinograph dough strength parameters.

Addition of the remaining flour quality measurements (the milling- and hardness-related parameters shown in Table I) had little effect upon the quality parameters contributing to the first and second principal components (results not shown). For the third principal component, flour protein was replaced by gassing power and starch damage (hardness-related measurements). The presence of these parameters may also be a result of breeder selection, in which bread-wheats are selected for hard kernel texture in order to obtain increased water absorption.

Relationships Between Quality Parameters and Protein Fractions

Flour protein was fractionated on the basis of extractability and molecular size to determine the relationship of the fractions to physical dough and baking properties. The former included measurement of 0.05*M* HAc- and 1.0*M* NaSCN-insoluble protein and modified Osborne fractionation (0.05*M* salt-soluble, 70% ethanol-soluble, 0.05*M* acetic acid-soluble, and residue). Proteins extractable in 1.0*M* sodium thiocyanate (nonreduced) were also separated into six fractions by gel filtration on Sephacryl S-300 to obtain molecular size distribution. Previous studies (Preston 1984) have shown that the first four fractions had apparent molecular weights of greater than 300,000 (glutenin), 38,000 (gliadin), 18,000 (mainly gliadins with high electrophoretic mobility), and 14,500 (similar to fraction 3). The last two fractions had lower apparent molecular weights (less than 10,000). The first three fractions (greater than 300,000, 38,000 and 18,000) on average, accounted for approximately 90% of the total extractable protein.

Tables IV and V show correlations between the distribution of protein fractions (and ratios of selected fractions) and physical dough and baking quality parameters. Flour protein, SDS sedimentation value, and Glu-1 score were also included. Very few of the protein fractions showed significant correlations with unfermented dough strength parameters (Table IV). Osborne salt-soluble (albumins and globulins) proteins and the ratios of salt-soluble proteins to glutenins and salt-soluble proteins to residue protein showed significant ($P < 0.05$ or < 0.01) correlations with farinograph measurements, with the highest values obtained for farinograph mixing tolerance index. The sign of these relationships indicated a negative relationship between salt-soluble protein and dough strength. For fermented dough (remix peak method), remix peak mixing time was significantly positively correlated with proteins insoluble in 1.0*M* NaSCN and 0.05*M* acetic acid as well as with "Osborne" 0.05*M* acetic acid-insoluble protein and were

TABLE V
Correlation Coefficients Between Protein Fractions and Baking Quality Parameters, Flour Protein, Sodium Dodecyl Sulfate Sedimentation, and Glutenin Score for 33 World Wheat Samples[a,b]

	RVOL	PVOL	BRVOL	NOVOL	RPRO	PPRO	BRPRO	NOPRO	SDS	GSCORE[c]
NASCN	−0.45**		−0.49**	−0.45**	−0.37*		−0.48**			
HAC						0.40*				0.38*
PK1										
PK2	0.50**		0.49**	0.53**					0.38*	
PK3	−0.73**	−0.58**	−0.73**	−0.73**	−0.44*		−0.43**			−0.38*
PK4						−0.40*				
PK5				0.37*						
PK6										−0.36*
RAT1										
RAT2	0.54**	0.43*	0.55**	0.51**	0.45**		0.48**	0.35*		
RAT3	0.74**	0.55*	0.77**	0.75**	0.38*		0.42*			
SALT										
GLI						−0.39*				
GLU										
RES		0.43**				0.52**				0.56**
SAGI										
SAGU										
SARES										−0.36*
GIGU										
GIRES		−0.39*				−0.49**				−0.47**
GURES										

[a]Correlation coefficients are shown if significant at the 5% ($P < 0.05$) or 1% ($P < 0.01$) level. The significance level is indicated by * for the 5% level and ** for the 1% level.

[b]RVOL = remix loaf volume, PVOL = remix peak loaf volume, BRVOL = AACC loaf volume (10 ppm bromate), NOVOL = AACC loaf volume (0 ppm bromate), RPRO = remix loaf volume per unit of protein, PPRO = remix peak loaf volume per unit of protein, BRPRO = AACC loaf volume (10 ppm bromate) per unit of protein, NOPRO = AACC loaf volume (0 ppm bromate) per unit of protein, SDS = sedimentation by sodium dodecyl sulfate electrophoresis, GLU-1 = glutenin score, NASCN = NaSCN-insoluble protein, HAC = HAc-insoluble protein, PK = gel filtration peak, RAT1 = ratio PK1:PK2, RAT2 = ratio PK1:PK3, RAT3 = ratio PK2:PK3, SALT = salt-soluble protein, GLI = gliadin protein, GLU = glutenin protein, RES = residual protein, SAGI = ratio SALT:GLI, SAGU = ratio SALT:GLU, SARES = ratio SALT:RES, GIGU = ratio GLI:GLU, GIRES = ratio GLI:RES, GURES = ratio GLU:RES. See Table I for more exact definitions.

[c]Correlation coefficients between glutenin score and other parameters were calculated by Spearman's rank-order technique.

negatively correlated with the ratio of "Osborne" glutenin to residue. Flour protein showed no significant relationship to the above protein fractionation parameters but was significantly related to a number of the gel filtration fractions. Glutenin score was positively correlated ($P < 0.05$) with gel filtration peak 6 and with 0.05M acetic acid-insoluble and "Osborne" insoluble protein and negatively related to peak 3 and the ratios of "Osborne" salt-soluble protein and 70% alcohol-soluble protein to residue protein. The SDS sedimentation value showed a significant correlation only with peak 2 protein from gel filtration.

The relationships between protein fractions and baking quality (Table V) were different than those between protein fractions and dough strength measurements. Loaf volumes were highly correlated ($P < 0.01$) with 1.0M thiocyanate-insoluble protein, gel filtration peaks 2 and 3, and the ratios of peaks 1 to 3 and 2 to 3. High r values (over 0.7) were obtained between three of the baking procedures and peak 3 and the ratio of peak 2 to 3. However, these high values, with the exception of 1.0M thiocyanate-insoluble protein, may be the result of the close relationship between these parameters and flour protein content (Table IV), which also gave very high correlations with bread volume. No highly significant ($P < 0.01$) correlations were evident between loaf volumes and "Osborne" fractions. However, a significant ($P < 0.05$) correlation was obtained between remix peak volume and "Osborne" residue protein.

Nonsignificant or significant correlations with low r values ($P < 0.5$) were evident between some protein fractions and loaf volumes per unit of protein. In general, prediction parameters that showed a significant relationship to remix peak loaf volume per unit of protein did not show a significant relationship to the other three baking procedures and vice versa. This result appears to be consistent with the lower correlations between remix peak values and those obtained with the other baking methods (Table II).

The results of PCA of the relationships among quality and protein fractionation parameters, given in Tables IV and V, are shown in Table VI. The first four principal components accounted for 67.2% of the total variability in the sample set. The first three principal components, accounting for 26.3, 15.6, and 14.7% of the total variability, respectively, showed quality parameters (loading factor > 0.25) for each component similar to those obtained in the absence of protein fractionation results (Table III). The fourth principal component was related to the associated variability among the "Osborne" fractions but was not related to any quality parameters.

The first principal component indicated that gel filtration peak 3 and the ratio of peak 2 to peak 3 were closely related to loaf volumes from all four baking procedures and to flour protein content. This is consistent with the high correlations found among these measurements, indicating that these gel filtration parameters and flour protein content are quantitative measures of loaf volume.

The second principal component was associated with farinograph dough strength properties and loaf volumes per unit of protein for the modified AACC baking procedures. The fractions with the highest loading factors on this component included "Osborne" salt-soluble proteins and the ratio of "Osborne" salt-soluble protein to "Osborne" residue protein. The third principal component appeared to be associated with the strength (remix peak time) of fermented dough and remix peak loaf volume per unit of protein. Glu-1 score, 0.05M acetic acid-insoluble protein, "Osborne" 0.05M acetic acid-insoluble protein, and the ratio of gliadin to residue protein also showed high loading factors for this component. SDS sedimentation value did not show high loading factors for any of the above principal components, consistent with its lack of significant correlation with physical dough and baking parameters (Table II).

DISCUSSION

Dough strength properties of both unfermented and fermented doughs showed little relationship to baking quality based upon correlations. This may have been partially due to the very strong influence (high correlations) of protein content on loaf volume with all four baking procedures. The high average dough strength of the sample set (Table I) may have also reduced the influence of these parameters on baking quality. Previous studies (Bushuk et al 1969, Hamada et al 1982) suggested that, above a minimum dough strength, further increases in this property may not result in improved baking quality. PCA analysis of the quality data (Table III) confirmed the strong influence of protein content on baking properties. However, after apparent removal of this influence, there appeared to be an association between dough strength and loaf volume per unit of protein (principal components 2 and 3, Table III). It is interesting to note that the dough strength properties of the unfermented doughs were associated with a different principal component and baking tests than those of fermented doughs and that no significant relationship was evident between the dough strength properties of unfermented and fermented doughs. At present, little information is available on the relationship of these properties and their relative influence on baking quality. However, work in our laboratory (Casutt et al 1984) and by Hoseney and co-workers (Wu and Hoseney 1989) indicated that wheat flours can vary in their fermentation response on the basis of extensigraph results and that these changes may influence baking properties.

Salt-extractable "Osborne" protein (and ratios containing this fraction) was the only protein fraction showing a significant correlation with farinograph dough strength parameters. PCA

TABLE VI
Principal Component (PC) Analysis of Flour Quality Data, Protein Fractionation Data, SDS Sedimentation, and Glutenin Score for 33 World Wheat Flour Samples[a]

PC 1 (26.3%)		PC 2 (15.6%)		PC 3 (14.7%)		PC 4 (10.6%)	
Variable[b]	Loading[c] Factor	Variable[b]	Loading[c] Factor	Variable[b]	Loading[c] Factor	Variable[b]	Loading[c] Factor
RVOL	0.29	SALT	0.30	HAC	0.33	GLI	0.38
NOVOL	0.29	SARES	0.28	PPRO	0.33	GIGU	0.40
BRVOL	0.28	BRPRO	0.28	PTIME	0.33	GLU	−0.34
PVOL	0.27	NOPRO	0.28	RES	0.30	SAGI	−0.32
PK3	−0.27	FTOL	0.28	GIRES	−0.26	GIRES	0.30
RAT3	0.26	FDDT	−0.26	GSCORE	0.26		
FPRO	0.25	FSTAB	−0.25				

[a] Principal components representing over 10% of the variability of the data set shown.
[b] RVOL = remix loaf volume, NOVOL = AACC loaf volume (0 ppm bromate), BRVOL = AACC loaf volume (10 ppm bromate), PVOL = remix peak loaf volume, PK3 = gel filtration peak 3, RAT3 = ratio gel filtration peak 2 to peak 3, FPRO = flour protein, SALT = salt-soluble protein, SARES = ratio SALT to residual (RES) protein, BRPRO = AACC loaf volume (10 ppm bromate) per unit of protein, NOPRO = AACC loaf volume (0 ppm bromate) per unit of protein, FTOL = farinograph mixing tolerance index, FDDT = farinograph dough development time, FSTAB = farinograph stability, HAC = HAc-insoluble protein, PPRO = remix peak loaf volume per unit of protein, PTIME = remix peak mixing time, GIRES = ratio gliadin protein (GLI) to RES, GSCORE = glutenin (GLU) score, GIGU = ratio GLI:GLU, SAGI = ratio SALT:GLI, GIRES = ratio GLI:RES. See Table I for more exact definitions.
[c] Loading factors (eigenvectors) of 0.25 or higher shown (positive or negative values).

analysis also showed that this fraction was associated with unfermented dough strength (principal component 2, Table VI) and with loaf volume per unit of protein with the modified AACC baking procedures. The former association could be related to the effects of albumins (Marais and D'Appolonia 1981b) and/or globulins (MacRitchie 1987) upon mixing requirements or to the presence of nonprotein components such as glutathione and similar compounds that reduce dough strength. The latter association could be related to the effect of albumins or globulins present in this fraction on bromate response (Marais and D'Appolonia 1981a) or could be the result of phosphoric acid present in the salt-soluble fraction (Hoseney et al 1972). With the modified AACC baking procedures, the use of constant bromate levels would increase the sensitivity of the flours to this influence. In contrast, the remix baking procedures are generally insensitive to varietal differences in oxidation requirements (Kilborn and Tipples 1981).

The lack of association between unfermented dough strength (farinograph properties) and protein fractions normally associated with high-molecular-weight and/or insoluble glutenins (0.05M NaSCN-insoluble, 1.0M acetic acid-insoluble, and "Osborne" acetic acid-insoluble) is surprising in view of previous studies indicating the opposite (Pomeranz 1965, Orth and Bushuk 1972, Orth and O'Brien 1976). However, these protein fractions were significantly correlated with the remix peak time of fermented doughs; the acetic acid-insoluble fraction ($r = 0.65$, $P < 0.01$) and the "Osborne" 0.05M acetic acid-insoluble protein ($r = 0.50$, $P < 0.01$) had the highest values. PCA also demonstrated an association between the latter two fractions, Glu-1 score, and remix peak time (principal component 3, Table VI), as well as remix peak loaf volume per unit of protein. Although speculative, these results suggest that the quantitative and qualitative (Glu-1 score) properties of the high-molecular-weight glutenins may play an important role in determining fermentation tolerance. Varieties lacking fermentation tolerance would result in weaker fermented doughs that lack optimum gas retention properties. This would result in lower bread volumes compared to those from varieties possessing good fermentation tolerance. These differences would be most evident with the remix peak baking procedure, where other factors including oxidation and mixing requirements are optimized.

LITERATURE CITED

AMERICAN ASSOCIATION OF CEREAL CHEMISTS. 1983. Approved Methods of the AACC, 8th ed. Method 54-21, approved April 1961, reviewed October 1982. The Association: St. Paul, MN.

AXFORD, D. W. E., McDERMOTT, E. E., and REDMAN, D. G. 1979. Note on the sodium dodecyl sulphate test of breadmaking quality: Comparison with Pelshenke and Zeleny tests. Cereal Chem. 56:582.

BIETZ, J. A. 1986. High-performance liquid chromatography of cereal proteins. Pages 105-107 in: Advances in Cereal Science and Technology, Vol. 8. Y. Pomeranz, ed. Am. Assoc. Cereal Chem.: St. Paul, MN.

BLACK, H. C., HSIEH, F.-H., TIPPLES, K. H., and IRVINE, G. N. 1980. The GRL sifter for laboratory flour milling. Cereal Foods World 25:757.

BRANLARD, G., and DARDEVET, M. 1985. Diversity of grain proteins and bread wheat quality: II. Correlation between high molecular weight subunits of glutenin and flour quality characteristics. J. Cereal Sci. 3:345.

BRANLARD, G., and ROUSSET, M. 1980. Les caracteristiques eletrophoretiques des gliadines et la valeur en panification du ble tendres. Ann. Amelior. Plant. 30(2):133.

BUSHUK, W., BRIGGS, K. G., and SHEBESKI, L. H. 1969. Protein quantity and quality as factors in the evaluation of bread wheats. Can. J. Plant Sci. 49:113.

CAMPBELL, W. P., WRIGLEY, C. W., CRESSEY, P. J., and SLACK, C. R. 1987. Statistical correlations between quality attributes and grain-protein composition for 71 hexaploid wheats used as breeding parents. Cereal Chem. 64:293.

CASUTT, V., PRESTON, K. R., and KILBORN, R. H. 1984. Effects of fermentation time, inherent flour strength, and salt level on extensigraph properties of full-formula remix-to-peak processed doughs. Cereal Chem. 61:454.

CHEN, C. H., and BUSHUK, W. 1970. Nature of proteins in triticale and its parental species. I. Solubility characteristics and amino acid composition of endosperm proteins. Can. J. Plant Sci. 50:9.

DACHKEVITCH, T., and AUTRAN, J.-C. 1989. Prediction of baking quality of bread wheats in breeding programs by size-exclusion high-pressure liquid chromatography. Cereal Chem. 66:448.

FARRAND, E. A. 1964. Flour properties in relation to the modern bread processes in the United Kingdom, with special reference to alpha-amylase and starch damage. Cereal Chem. 41:98-111.

FINNEY, K. F. 1985. Experimental breadmaking studies, functional (breadmaking) properties, and related gluten protein fractions. Cereal Foods World 30:794.

FINNEY, K. F., and BARMORE, M. A. 1948. Loaf volume and protein content of hard red winter and spring wheats. Cereal Chem. 25:291.

FOWLER, D. B., and DeLaROCHE, I. A. 1975. Wheat quality evaluations. 2. Relationships among prediction tests. Can. J. Plant Sci. 55:251.

HAMADA, A. S., McDONALD, C. E., and SIBBITT, L. D. 1982. Relationship of protein fractions of spring wheat flour to baking quality. Cereal Chem. 59:296-301.

HOSENEY, R. C., and FINNEY, K. F. 1971. Functional (breadmaking) and biochemical properties of wheat flour components. XI. A review. Baker's Dig. 45(4):30.

HOSENEY, R. C., FINNEY, K. F., and SHOGREN, M. D. 1972. Functional (breadmaking) and biochemical properties of wheat flour components. X. Fractions involved in the bromate reaction. Cereal Chem. 49:372.

HUEBNER, F. R. 1989. Assessment of potential breadmaking quality of hard spring wheats by high-performance liquid chromatography of gliadins—Year two. Cereal Chem. 66:333.

HUEBNER, F. R., and BIETZ, J. A. 1985. Detection of quality differences among wheats by high-performance liquid chromatography. J. Chromatogr. 327:333.

HUEBNER, F. R., and BIETZ, J. A. 1986. Assessment of the potential breadmaking quality of hard wheats by reversed-phase high-performance liquid chromatography of gliadins. J. Cereal Sci. 4:379.

HUEBNER, F. R., and WALL J. S. 1976. Fractionation and quantitative differences of glutenin from wheat varieties varying in baking quality. Cereal Chem. 53:258.

KILBORN, R. H., and TIPPLES, K. H. 1981. Canadian test baking procedures. I. GRL remix method and variations. Cereal Foods World 26:624.

KOSMOLAK, F. G. 1978. Grinding time—A screening test for kernel hardness in wheat. Can. J. Plant Sci. 58:415.

LEE, J. W., and MacRITCHIE, F. 1971. The effect of gluten protein fractions on dough properties. Cereal Chem. 48:620.

LUKOW, O. M., PAYNE, P. I., and TKACHUK, R. 1989. The HMW glutenin subunit composition of Canadian wheat cultivars and their association with bread-making quality. J. Sci. Food Agric. 46:451.

LUKOW, O. M., ZHANG, H., and CZARNECKI, E. 1990. Milling, rheological, and end-use quality of Chinese and Canadian spring wheat cultivars. Cereal Chem. 67:170.

MacRITCHIE, F. 1973. Conversion of a weak flour to a strong one by increasing the proportion of its high molecular weight gluten protein. J. Sci. Food Agric. 24:1325.

MacRITCHIE, F. J. 1987. Evaluation of contributions from wheat protein fractions to dough mixing and breadmaking. J. Cereal Sci. 6:259.

MARAIS, G. F., and D'APPOLONIA, B. L. 1981a. Factors contributing to baking quality differences in hard red spring wheat. I. Bases for different loaf volume potentials. Cereal Chem. 58:444.

MARAIS, G. F., and D'APPOLONIA, B. L. 1981b. Factors contributing to baking quality differences in hard red spring wheat. II. Bases for different mixing properties. Cereal Chem. 58:448.

MITCHESON, R. C., and STOWELL, K. C. J. 1970. Application of new analytical techniques to routine malt analysis. I. Determination of barley and malt nitrogen content using an autoanalyser technique. J. Inst. Brew. 76:335.

ORTH, R. A., and BUSHUK, W. 1972. A comparative study of the proteins of wheats of diverse baking qualities. Cereal Chem. 49:268.

ORTH, R. A., and O'BRIEN, L. J. 1976. A new biochemical test of dough strength of wheat flour. J. Aust. Inst. Agric. Sci. 42:122.

PAYNE, P. I., CORFIELD, K. G., and BLACKMAN, J. A. 1979. Identification of a high-molecular-weight subunit of glutenin whose presence correlates with bread-making quality in wheats of related pedigree. Theor. Appl. Genet. 55:153.

PAYNE, P. I., CORFIELD, K. G., HOLT, L. M., and BLACKMAN, J. A. 1981. Correlations between the inheritance of certain high-molecular weight subunits of glutenin and bread-making quality in progenies of six crosses of bread wheat. J. Sci. Food Agric. 32:51.

PAYNE, P. I., NIGHTINGALE, M. A., KRETTIGER, F., and HOLT,

L. M. 1987. The relationship between HMW glutenin subunit composition and the bread-making quality of British-grown wheat varieties. J. Sci. Food Agric. 40:51.
POMERANZ, Y. 1965. Dispersibility of wheat proteins in aqueous urea solutions—A new parameter to evaluate breadmaking potentialities of wheat flours. J. Sci. Food Agric. 16:586.
PRESTON, K. R. 1984. Gel filtration and characterization of neutral salt extracted wheat gluten proteins varying in hydrophobic properties. Cereal Chem. 61:76.
PRESTON, K. R., MARCH, P. R., and TIPPLES, K. H. 1982a. An assessment of the SDS-sedimentation test for the prediction of Canadian bread wheat quality. Can. J. Plant Sci. 62:545.
PRESTON, K. R., KILBORN, R. H., and BLACK, H. C. 1982b. The GRL pilot mill. II. Physical dough and baking properties of flour streams milled from Canadian red spring wheat. Can. Inst. Food Sci. Technol. J. 15:29.
PRESTON, K. R., MORGAN, B. C., and TIPPLES, K. H. 1988. A review and analysis of export cargo quality data for Canada Western Red Spring wheat: 1973–1986. Can. Inst. Food Sci. Technol. J. 21:520.
SALOMONSSON, L., and LARSSON-RAZNIKIEWICZ, M. 1985. Method for fractionation and characterization of proteins in wheat. Swed. J. Agric. Res. 15:123.
SAS INSTITUTE. 1987. SAS/STAT Guide for Personal Computers, Version 6 Edition. The Institute, Inc.: Cary, NC.
SINGH, N. K., DONOVAN, R., and MacRITCHIE, F. 1990. Use of sonication and size-exclusion high-performance liquid chromatography in the study of wheat flour proteins. II. Relative quantity of glutenin as a measure of breadmaking quality. Cereal Chem. 67:161.
SINHA, R. N. 1977. Use of multivariate methods in the study of stored-grain ecosystems. Environ. Entomol. 6:185.
WIESER, H., SEILMEIER, W., and BELITZ, H.-D. 1989. Reversed-phase high-performance liquid chromatography of ethanol-soluble and ethanol-insoluble reduced glutenin fractions. Cereal Chem. 66:38.
WILLIAMS, P. C. 1973. The use of titanium dioxide as a catalyst for large scale Kjeldahl determination of the total nitrogen content of cereal grains. J. Sci. Food Agric. 24:343.
WILLIAMS, P. C., and SOBERING, D. C. 1986. Attempts at standardization of hardness testing of wheat. I. The grinding/sieving (particle size index) method. Cereal Foods World 31:359.
WRIGLEY, C. W., LAWRENCE, G. J., and SHEPHERD, K. W. 1982. Association of glutenin subunits with gliadin composition and grain quality in wheat. Aust. J. Plant Physiol. 9:15.
WU, J. Y., and HOSENEY, R. C. 1989. Rheological changes in cracker sponges during an 18-hour fermentation. Cereal Chem. 66:182.

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