

Gluten, Pasting, and Mixograph Parameters of Hard Winter Wheat Flours in Relation to Breadmaking¹

J. B. Ohm^{2,3} and O. K. Chung^{2,4}

ABSTRACT

Cereal Chem. 76(5):606–613

Flour gluten, pasting, and mixogram characteristics of 12 hard winter wheat cultivars grown in six counties in Kansas were analyzed using the Glutomatic System, a Rapid Visco-Analyser, and MIXSMART computer software, respectively, to investigate their relationships with breadmaking. Gluten contents and hydration amounts had significant correlations with water absorption. In addition, gluten parameters were significantly correlated to kernel hardness. One of the most difficult challenges in mixograph usage is to find the optimum water absorption of a given flour. Flour protein contents (FP) and near-infrared hardness scores or FP and gluten

parameters could predict mixograph water absorptions, showing R^2 values of 0.842 or 0.814, respectively, by multiple regression analysis. For our set of 72 wheat samples, computer-analyzed mixograph parameters were significantly correlated to conventional parameters. Computer-analyzed mixograph midline peak times and bandwidths at 6 min were highly correlated to conventional mixograph mix times and mixing tolerances, respectively. Flour pasting temperatures complemented FP in predicting loaf volumes. The ratios of FP to pasting temperatures had a significant curvilinear relationship with loaf volumes showing an R^2 of 0.725.

Proteins are among the primary factors responsible for the variation in wheat flour quality. Flour protein content (FP) alone is generally measured by the analysis of nitrogen. Recently, wheat protein quantity and quality have been determined by measuring gluten content and gluten index (GI) using the Glutomatic System (Pertin 1990). Gluten parameters show potential as a quality factor. Chung and Seabourn (1990) reported that GI was related linearly to loaf volume (LV) potentials (LV regression slopes: change in LV [cm³]/1 percentage point of FP) and curvilinearly to bake mix times. Some other gluten properties can be obtained from the Glutomatic System in addition to gluten content and GI. They need to be investigated to determine whether they have any significant relationship with baking parameters.

Flour pasting properties have been measured by changes in viscosity during heating using a Brabender Amylograph. The pasting properties of flour are related closely to those of starch. Flour pasting properties are also influenced by other flour components including proteins, pentosans, and lipids; added ingredients such as sugar and salt; and α -amylase activity (Hutchinson 1966, Mathewson and Pomeranz 1978, Olkku et al 1978). Recently, the Rapid Visco-Analyser (RVA) was developed for detection of sprout damage. We used the RVA to evaluate the pasting properties of flour and starch (Ross et al 1987). The RVA provides a pasting curve that is sensitive enough to detect the differences in viscosity of flours with a small sample size in a short time (Walker et al 1988, Deffenbaugh and Walker 1989). Research on relationships between flour pasting properties and breadmaking is limited, and the RVA appears to have potential to study these relationships.

The mixograph was developed in response to a need to evaluate the strong, high-protein flours produced in the Great Plains of the United States (Shogren 1990). When Swanson and Working (1933)

introduced a basic mixograph system requiring 400 g of flour, the National Manufacturing Company (Lincoln, NE) developed a mixograph requiring 35 g of flour in 1939. Other mixographs using 10-, 5-, and 2-g of flour have been developed subsequently with simplified and standardized procedures (Finney and Shogren 1972, Finney 1989, Rath et al 1990). Mixograph procedures have also been improved by computerization, which facilitates data handling and allows objective interpretation of the mixogram (Rubenthaler and King 1986, Pon et al 1989, Gras et al 1990, Navickis et al 1990, Sterns and Barta 1990). Mixograph parameters measured by a computerized system were significantly correlated with those by conventional method (Pon et al 1989, Gras et al 1990).

Several researchers have used computers to interpret mixograph parameters (Buckley et al 1990, Neufeld and Walker 1990, Wikström and Bohlin 1996, Martinant et al 1998). It is possible to obtain many mixogram parameters in a short time using MIXSMART commercial software, (Version 3.40, National Manufacturing Division, TMCO, Lincoln, NE). These parameters needed to be evaluated for testing flour quality.

Because the Glutomatic System, RVA, and mixograph are simple, rapid, and safe procedures that require small sample amounts and no hazardous organic chemicals, they could be very helpful for flour quality evaluation in breeding programs and industry where there is a need to analyze many samples in a short time. Using these instruments, new parameters can be obtained from gluten, pasting curve, and mixogram analyses that are potential quality factors for breadmaking. Thus, we investigated whether the Glutomatic System, RVA, and MIXSMART could be applied to quality evaluation of hard winter wheats produced in Kansas, one of the major wheat production areas in the United States.

MATERIALS AND METHODS

Wheat samples from 12 hard winter wheat cultivars harvested in six counties in Kansas were analyzed for gluten, pasting, and mixograph properties. Information on wheat cultivars, growing regions, and other quality data were described previously (Ohm et al 1998).

Gluten Properties

The gluten properties of flour were determined using a Glutomatic System (Falling Number, Huddinge, Sweden) according to Method 106/2 (Standard No. 137/1) (ICC 1995). Wet gluten (WG) was washed and separated from 10 g of flour on a 14% flour moisture basis (mb) with 2% NaCl buffer (pH 5.95) using a Glutomatic 2100 Automatic Washing System. The WG was centrifuged using the GI sieve cassettes at 6,000 rpm for 1 min. After the

¹ Cooperative investigations, U. S. Department of Agriculture, Agricultural Research Service and the Department of Grain Science and Industry, Kansas State University, Contribution No. 97-412-J from the Kansas Agriculture Experiment Station, Manhattan, KS 66506. Mention of firm names or trade products does not constitute endorsement by the U. S. Department of Agriculture over others not mentioned.

² Graduate research assistant and professor, respectively, Department of Grain Science and Industry, Kansas State University, Manhattan, KS 66506.

³ Present address: Postdoctoral research chemist, USDA-ARS, Grain Marketing and Production Research Center, Manhattan, KS 66502.

⁴ Supervisory research chemist, USDA-ARS, Grain Marketing and Production Research Center, Manhattan, KS 66502. Corresponding author. Phone 785/776-2703. Fax: 785/776-2792. E-mail: okchung@usgmrl.ksu.edu

amount of gluten that passed through the sieve was weighed, the weight of total WG was measured. The weight of dry gluten (DG) was determined after drying WG at 150°C for 4 min. The GI was calculated as:

$$GI = 100 \times [WG (14\% \text{ flour mb}) - \text{gluten passed through sieve (14\% flour mb)}] / WG (14\% \text{ flour mb})$$

Gluten hydration amount (GHA) and gluten hydration capacity (GHC) were estimated as:

$$GHA = WG - DG$$

and

$$GHC = GHA/DG$$

The ratio of DG to FP (DG/FP) was also estimated.

Pasting Properties

Pasting properties of flour were measured by a Rapid Visco-Analyser (RVA-4, Newport Scientific, Australia) according to Standard Procedure 1 described in the Operations Manual supplied by the manufacturer. RVA analysis was performed with 25.0 mL (± 0.1 mL) water and 3.5 g of flour (14% mb) weighed to the nearest 0.01 g in a canister. The RVA was warmed up 30 min at 50°C before analysis. After a sample was equilibrated at 50°C for 1 min, temperature was increased to 95°C at a rate of 12°C/min, maintained at 95°C for 2.5 min, decreased to 50°C at a rate of 12°C/min, and held at 50°C for 2 min. Thus, the total analysis took 13 min per sample. Paddle speed was adjusted to 960 rpm for the first 10 sec to mix flour with water and then maintained at 160 rpm to the end of the analysis. The pasting profile was recorded and characterized using Thermoline software for Windows (Version 1.0, Newport Scientific 1995). The measured parameters of the RVA pasting curve included pasting temperature (PT), peak viscosity (PV), trough viscosity (TV), final viscosity (FV), breakdown (BD), and setback (SB). Viscosity was measured in RVA viscosity units (RVU).

Mixograph Procedure

The mixogram was obtained using a 10-g bowl mixograph according to the procedure of Finney and Shogren (1972). Flour (10 g) was mixed with the optimum amount of water for 8 min or until mix time could be determined at 77°F in a mixograph bowl. Optimum water absorption was empirically determined by the feel of the dough. A mixograph pattern was analyzed to provide mixing requirement, mixing tolerance, and optimum water absorption. The mixograph pattern was also evaluated using MIXSMART computer software (Version 3.40, National Manufacturing Division, TCMCO, Lincoln, NE). Computer-analyzed parameters of the mixograph included time to peak (midline peak time, MPT) and bandwidth at the MPT; receptive time; slope and bandwidth of the left or right of the envelope peak; time and width of envelope peak; and the mixograph bandwidth at 6 min (B6M). Computer-analyzed mixograph parameters are described in detail in the MIXSMART user's manual (National Cereal Chemistry Equipment, Lincoln, NE).

Statistical Analysis

Statistical analysis was performed using SAS statistical methods (Version 6.11, SAS Institute, Cary, NC). Analysis of variance was performed considering effects of cultivar and growing region as random. Phenotypic correlation (r_p , $n = 72$) was calculated from data across all combinations of 12 cultivars and six regions using the average of two replicates. Correlation coefficient among cultivars (r_c) ($n = 12$) was estimated using each mean performance of 12 cultivars across six counties (Souza et al 1993). Correlation among growing regions (r_r) ($n = 6$) was calculated using each mean performance of six counties. Multiple regression was computed with parameters that are significant at $P < 0.05$.

Means and Ranges for Gluten, Pasting, and Mixograph Parameters

Table I shows mean, standard deviation, and range for the FP, gluten and pasting properties, and mixogram parameters of the 72 samples analyzed. Ranges for FP, WG, DG, and GHA were 7.8–14.8%, 15.8–40.8%, 5.8–14.3%, and 10.0–27.1% (14% mb), respectively. The GI value ranged from 74.0 to 99.5%. The GHC values indicated that gluten could take up to $\approx 1.9\times$ of its weight in water, on an average, ranging from 1.5 to 2.1. All GHC values were within the reported range of 1–3 kg of water absorbed/1 kg of dry flour protein (Bloksma and Bushuk 1988). The DG/FP ratio, indicating the gluten-forming ability of flour protein, ranged from 0.68 to 1.24, with an average of 0.94. In general, gluten containing $\approx 80\%$ of the total flour protein consists of 80% proteins, 5–8% lipids, and 12–15% occluded starch, nonstarch carbohydrates, and other components (Wrigley and Bietz 1988).

For the pasting properties, PT ranged from 65.7 to 87.7°C with a mean of 80.5°C (Table I). The mean values of PV, TV, FV, BD, and SB were 187.5, 121.2, 190.9, 66.3, and 69.7 RVU, respectively. Among the 72 flours, some had significantly lower viscosity, which might have been caused by sprout damage. Sprout damage was considered to affect flours from TAM 200, Arlin, and Discovery cultivars grown in Franklin County and Scout 66 and Arlin cultivars grown in Stafford County (data not shown). Pasting viscosity decreases as sprouting increase due to high α -amylase activity (Kruger and Lineback 1987).

For conventional mixograph data, the range was 56.4–67.2% (14% flour, mb) for water absorptions, and 2.9–9.2 min for mix times (Table I). Mix tolerance ranged from 2.0 to 5.0 with a mean value of 3.8. For the computer-analyzed mixograph parameters, the MPT ranged from 3.7 to 9.7 min with a mean value of 6.2 min. The envelope peak time (EPT) values were generally lower than the MPT values with a range from 0.7 to 9.2 min and a mean value of 4.9 min. The B6M value ranged widely from 7.4 to 26.4. Other parameters also showed wide ranges of variation (Table I).

Analysis of variance indicated that the effects of cultivars, growing regions, and their interactions were estimated to be significant ($P < 0.001$) for FP and gluten characteristics including WG, DG, and GHA (Table II). The variation in GI values among cultivars was significant at $P < 0.10$ but not at $P < 0.05$, probably because the genetic variations of wheat were limited in the hard winter wheat cultivars used in this study. Among pasting parameters, cultivars, growing regions, and their interactions affected variations in PT and TV at $P < 0.05$. However, cultivar effect was insignificant on PV, FV, and BD, suggesting that growing regions strongly affected variations in these parameters. The SB values were not affected by cultivar or growing region. Mixograph water absorptions, mix times, and mixing tolerances were significantly affected (at $P < 0.01$) by cultivars, growing regions, and their interactions. Many computer-analyzed parameters were also affected by cultivars, growing regions, and their interactions.

Relationships Between Protein, Gluten, and Pasting Properties

Significant linear correlation coefficients (r_p for phenotypic, r_c for cultivar, r_r for growing region) occurred among gluten parameters. The genetic and environmental variations in protein contents had significant effects on gluten quantity as indicated by correlations between FP and DG ($r_p = 0.90$, $r_c = 0.78$, $r_r = 0.97$; $P < 0.01$). A similar relationship was previously reported (Doekes and Wennekes 1982). The high correlations of GHA with WG ($r_p = 0.99$, $r_c = 0.99$, $r_r = 0.99$; $P < 0.01$) and with DG ($r_p = 0.97$, $r_c = 0.93$, $r_r = 0.99$; $P < 0.01$) suggest that the amount of water held by gluten can be affected greatly by the quantitative variations in gluten. The high r_c and r_r values indicated significant variations were caused by both cultivars and growing regions. The GI values were negatively correlated with DG ($r_p = -0.28$, $r_c = -0.63$; $P < 0.05$).

and WG ($r_p = -0.24$, $r_c = -0.66$; $P < 0.05$). However, the gluten-forming abilities (DG/FP ratios) were positively correlated to both WG ($r_p = 0.68$, $r_c = 0.83$; $P < 0.01$) and DG ($r_p = 0.65$, $r_c = 0.76$; $P < 0.01$). The high r_c values of DG with DG/FP and with GI showed that both gluten quantity and quality were affected by wheat cultivars. Significant correlation coefficients also occurred among flour pasting properties analyzed by RVA. PV values were significantly ($P < 0.01$) correlated with TV ($r_p = 0.92$, $r_c = 0.96$, $r_r = 0.90$) and FV ($r_p = 0.96$, $r_c = 0.97$, $r_r = 0.99$).

Relationships Between Conventional and Computer-Analyzed Mixograph Parameters

Mixograph water absorptions had significant phenotypic correlations with many computer-analyzed mixograph parameters including midline peak height (MPH), midline peak width (MPW), slope of left of envelope peak (SLE), bandwidth of right of envelope peak (BRE), and B6M values (Table III). In addition, mixograph water absorptions significantly influenced MPH ($r_r = 0.99$, $P < 0.01$), SLE ($r_r = 0.99$, $P < 0.01$), and MPW ($r_r = 0.81$, $P < 0.05$), suggesting growing environmental influences were highly significant on these parameters.

Although conventional mixograph mix times showed the highest phenotypic correlation with MPT, MPT values alone were not enough to explain all the variations in conventional mixograph mix times (Table III). Conventional mixograph mix times and MPT also showed significant correlations among cultivars ($r_c = 0.92$, $P < 0.01$) and among growing regions ($r_r = 0.87$, $P < 0.05$). The B6M values had a significant r_p value (0.58 at $P < 0.01$) and r_c value (0.90 at $P < 0.01$) with conventional mix tolerances, suggesting that B6M could be helpful in evaluating mixing tolerance. The significant correlations between conventional and computer-analyzed mixo-

graph parameters strongly suggest that the objective parameters from computer analysis can be used to evaluate flour quality.

Relationships Between Wheat Hardness and Gluten Parameters

Using the data on wheat kernel hardness parameters reported by Ohm et al (1998), r_p values between near-infrared reflectance hardness score (NIR-HS) with a range of 38–86 (a mean value of 62) and gluten parameters were calculated (Table IV). Though there was no significant relationship between NIR-HS and FP, the WG, DG, and GHA values showed significant correlations with NIR-HS. The relationship between gluten parameters and NIR-HS was investigated further using multiple regression analysis. The regression equation showed an R^2 value of 0.543 ($N = 72$) ($r_c = 0.72$, $r_r = 0.93$; $P < 0.01$): $\text{NIR-HS} = -0.5 + 1.2 \times (\text{DG})^* + 229.2 \times (\text{DG/GI})^{**} + 1869.0 \times [\text{GHA}/(\text{FP} \times \text{GI})]^{**}$ (*, ** = significant at $P < 0.05$ and $P < 0.01$, respectively). Thus, gluten parameters can be responsible for 54.3% of the variation in NIR-HS values. The ratios of GHA/(FP \times GI) had a significant correlation with the NIR-HS ($r_p = 0.66$, $r_c = 0.77$, $r_r = 0.86$; $P < 0.01$). The correlation between DG/GI and NIR-HS was phenotypically significant ($r_p = 0.37$, $P < 0.01$). Starch surface proteins and lipids have been reported to be responsible for the variations in hardness among durum, hard, and soft wheats (Greenwell and Schofield 1986, Greenblatt et al 1995). In the hard winter wheat sample set that we studied, significant correlations between hardness and gluten parameters suggest that gluten quantity and quality could be partly responsible for the variation in hardness. A significant relationship between gluten proteins and kernel hardness was reported within a wheat class. Huebner and Gaines (1992) showed that a gliadin fraction was related to hardness in hard red spring wheats. The GI

TABLE I
Mean and Standard Deviation (SD) of Gluten, Pasting, and Mixograph Parameters^a

Quality Parameter	Abbreviation	Mean	SD	Minimum	Maximum
Flour protein content (%) ^b	FP	10.0	1.6	7.8	14.8
Gluten properties					
Wet gluten content (%)	WG	27.4	5.8	15.8	40.8
Dry gluten content (%)	DG	9.5	2.0	5.8	14.3
Gluten hydration amount (%)	GHA	17.9	3.8	10.0	27.1
Gluten index (%)	GI	93.7	4.9	74.0	99.5
Gluten hydration capacity (%/%)	GHC	1.9	0.1	1.5	2.1
Dry gluten/flour protein (%/%)	DG/FP	0.94	0.09	0.68	1.24
Pasting properties					
Pasting temperature (°C)	PT	80.5	8.0	65.7	87.7
Peak viscosity (RVU) ^c	PV	187.5	27.8	28.2	207.2
Trough viscosity (RVU)	TV	121.2	31.9	1.4	167.5
Final viscosity (RVU)	FV	190.9	37.6	2.8	252.0
Breakdown (RVU)	BD	66.3	12.9	26.8	93.9
Setback (RVU)	SB	69.7	16.3	1.4	118.1
Conventional mixograph					
Water absorption (%)	...	60.6	2.7	56.4	67.2
Mix time (min)	...	5.2	1.2	2.9	9.2
Mix tolerance	...	3.8	0.6	2.0	5.0
Computer-analyzed mixograph					
Midline peak					
Time (min)	MPT	6.2	1.2	3.7	9.7
Height	MPH	37.3	5.8	27.5	50.3
Width	MPW	13.6	3.4	7.6	26.8
Left of envelope peak					
Slope	SLE	4.6	2.5	0.1	11.5
Bandwidth	BLE	16.6	3.5	10.4	27.9
Envelope peak					
Time (min)	EPT	4.9	1.7	0.7	9.2
Width	EPW	16.9	4.0	10.6	30.2
Right of envelope peak					
Slope	SRE	-3.4	1.6	-8.5	-0.3
Bandwidth	BRE	15.0	5.5	7.4	31.1
Bandwidth at 6 min	B6M	13.0	3.5	7.4	26.4

^a Mean and standard deviation were calculated from mean of two replicates ($n = 72$).

^b 14% moisture basis of flour.

^c Rapid Visco-Analyser viscosity unit.

values showed a negative correlation with wheat hardness (Table IV) and also with quantitative parameters of glutes (data not shown). All gluten parameters except GI had positive correlations with wheat hardness (Table IV). Thus, the negative correlation between GI and wheat hardness scores might have resulted from the quantitative aspects of gluten. An increase in gluten quantity can accompany an increase in gliadins, which could contribute negatively to GI but positively to wheat hardness. The NIR-HS and GI also had significant r_c and r_r values ($r_c = -0.74$, $P < 0.01$; $r_r = -0.85$, $P < 0.05$) and DG/FP ratios ($r_c = 0.70$, $P < 0.05$; $r_r = 90$, $P < 0.05$).

TABLE II
Analysis of Variance Results for Gluten, Pasting, and Mixograph Parameters^a

Quality Parameter	Cultivar (C)	Growing Region (R)	C × R
Flour protein content (%) ^b	***	***	***
Gluten properties			
Wet gluten content (%)	***	***	***
Dry gluten content (%)	***	***	***
Gluten hydration amount (%)	***	***	***
Gluten hydration capacity (%/%)	**	**	ns
Dry gluten/flour protein (%/%)	*	***	***
Pasting properties			
Pasting temperature (°C)	**	***	***
Peak viscosity (RVU) ^c	ns	***	***
Trough viscosity (RVU)	**	***	***
Final viscosity (RVU)	ns	***	***
Breakdown (RVU)	ns	***	***
Setback (RVU)	ns	ns	***
Conventional mixograph			
Water absorption (%)	**	***	***
Mix time (min)	***	***	***
Mix tolerance	**	***	***
Computer-analyzed mixograph			
Midline peak			
Time (min)	***	***	***
Height	**	***	***
Width	**	***	*
Left of envelope peak			
Slope	***	***	***
Bandwidth	ns	***	**
Envelope peak			
Time (min)	***	ns	***
Width	*	***	**
Right of envelope peak			
Slope	ns	ns	***
Bandwidth	*	**	***
Bandwidth at 6 min	***	***	***

^a *, **, *** = *F*-value is significant at $P < 0.05$, 0.01 , and 0.001 , respectively; ns = not significant at $P < 0.05$.

^b 14% moisture basis of flour.

^c Rapid Visco-Analyser viscosity unit.

TABLE III
Correlation Coefficients Between Conventional and Computer-Analyzed Mixograph Parameters^a

Computer-Analyzed Parameter ^b	Conventional Mixograph Parameter		
	Water Absorption (%) ^b	Mix Time (min)	Mix Tolerance (min)
Midline peak			
Time (min)	ns	0.81	0.61
Height	0.84	ns	ns
Width	0.45	0.58	0.47
Left of envelope peak			
Slope	0.77	ns	ns
Envelope peak			
Time (min)	ns	0.75	0.37
Width	ns	0.31	0.45
Right of envelope peak			
Bandwidth	0.63	0.31	0.35
Bandwidth at 6 min	0.36	0.70	0.58

^a $n = 72$; $P < 0.01$; ns = correlation coefficient is not significant at $P < 0.01$.

^b 14% moisture basis of flour.

Relationships Between Wheat Hardness and Mixograph Parameters

The NIR-HS and mixograph water absorptions showed significant r_p values (Table IV). The effects of hardness parameters on mixograph parameters are probably related to damaged starches, which are influenced greatly by wheat kernel hardness. Damaged starches increase flour-water absorption because their water-absorbing capacity is 3× as high as that of undamaged starch granules (Farrand 1964).

However, the NIR-HS had negative phenotypic correlations with mixograph mix times ($r_p = -0.42$, $P < 0.01$) and MPT ($r_p = -0.50$, $P < 0.01$), though Martinant et al (1998) reported insignificant correlations between wheat kernel hardness and MPT values. These seemingly contradictory results may be due to the differences in genetic variations and the influence of growing environment between the sample sets. The significant correlations of NIR-HS with GI and DG/FP in this sample set suggest that the relationship of mixograph mix times and NIR-HS might have also resulted from variation in gluten quality. The variation in flour protein composition has been known to influence dough development and strength (Dachketvitch and Autran 1989, MacRitchie 1989, Martinant et al 1998).

Relationships Between Protein, Gluten, and Mixograph Parameters

The FP showed significant phenotypic correlations with mixograph water absorption and computer-analyzed parameters (Table V).

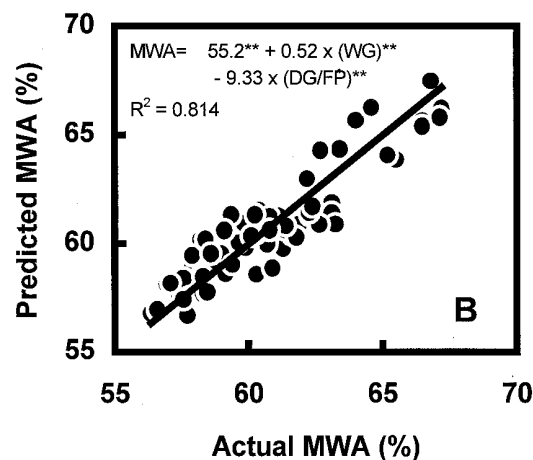
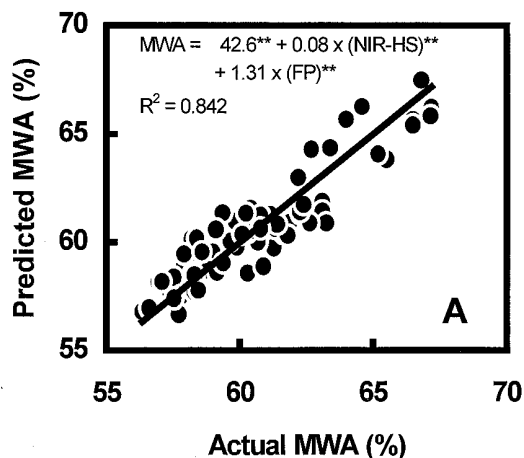


Fig. 1. Plots of actual mixograph water absorption (MWA) and predicted absorption by flour protein content (FP) and near-infrared hardness score (NIR-HS) (A) and gluten parameters (B). WG = wet gluten content (14% flour mb), DG = dry gluten content (14% flour mb). ** = significant at $P < 0.01$.

Optimum water requirements for mixing were correlated linearly with FP (Merritt and Stamborg 1941, Navickis et al 1990), which had the most influence on variation in mixograph parameters within a cultivar (Swanson 1941). Mixograph water absorptions and computer-analyzed mixograph parameters were significantly influenced by FP. In addition, with computer-analyzed mixograph parameters, FP had high r_c and r_r values including MPH ($r_c = 0.82$, $P < 0.01$; $r_r = 0.90$, $P < 0.05$), midline peak bandwidth (MPB) ($r_c = 0.67$, $P < 0.05$; $r_r = 0.96$, $P < 0.01$), SLE ($r_c = 0.60$, $P < 0.05$; $r_r = 0.94$, $P < 0.01$) and BRE ($r_c = 0.67$, $P < 0.05$; $r_r = 0.74$, $P < 0.10$).

Mixograph water absorptions had significant phenotypic correlations with gluten parameters such as WG, DG, and GHA. In addition, it showed significant r_c and r_r values with WG ($r_c = 0.92$, $r_r = 0.97$; $P < 0.01$), DG ($r_c = 0.89$, $r_r = 0.96$; $P < 0.01$), and GHA ($r_c = 0.91$, $r_r = 0.97$; $P < 0.01$). Insignificant correlations between mixograph mix times and gluten contents might have resulted from the nonlinear relationship of the mixograph mix time with FP (Johnson et al 1943) and the variations in flour protein composition that influence dough development and strength (MacRitchie 1989, Dachketvitch and Autran 1989, Martinant et al 1998).

The SLE values represent the dough development rate (Swanson and Johnson 1943). The positive r_p values of SLE with FP, WG, DG, and GHA suggest the influence of FP and gluten contents on SLE. The SLE values also had positive r_r values with FP ($r_r = 0.94$, $P < 0.01$), WG ($r_r = 0.95$, $P < 0.01$), DG ($r_r = 0.95$, $P < 0.01$), and GHA ($r_r = 0.95$, $P < 0.01$).

The GHC had negative correlations with mixograph mix times ($r_p = -0.52$, $r_c = -0.72$, $r_r = -0.92$; $P < 0.01$), tolerances ($r_p = -0.50$, $P < 0.01$; $r_c = -0.76$, $P < 0.01$; $r_r = -0.81$, $P < 0.05$), and MPT ($r_p = -0.42$, $P < 0.01$; $r_c = -0.71$, $P < 0.05$; $r_r = -0.79$, $P < 0.10$). Low hydration of gluten could be caused by its strong hydrophobic interaction, which was considered to enhance gluten quality for bread-making (Nakai and Li-Chan 1993).

Prediction of Optimum Mixograph Water Absorption

As stated earlier, the FP and kernel hardness greatly influenced the variation in water absorption. The NIR-HS values supplemented FP for the estimation of mixograph water absorptions of hard winter wheats. The estimation equation of mixograph water absorption was attained using FP (14% mb) and NIR-HS values. Optimum mixograph water absorption = $42.6 + 0.08 \times (\text{NIR-HS}) + 1.31 \times (\text{FP})$. Parameters in this equation are significant at $P < 0.01$. The equation showed an R^2 of 0.842 ($n = 72$) and an RMSE of 1.10 ($r_p = 0.92$, $r_c = 0.78$, $r_r = 0.97$; $P < 0.01$) as indicated in Fig. 1A.

Significant correlations that occurred between gluten parameters, wheat kernel hardness, and water absorptions (Table IV) also suggested that mixograph water absorptions could be estimated using gluten parameters. The estimation equation of mixograph water

absorption was calculated using FP and gluten parameters. Optimum mixograph water absorption = $55.2 + 0.52 \times (\text{WG}) - 9.33 \times (\text{DG/FP})$. Parameters in this equation are significant at $P < 0.01$. The equation indicated an R^2 of 0.814 ($n = 72$) and an RMSE of 1.19 ($r_c = 0.90$, $r_r = 0.96$; $P < 0.01$) as indicated in Fig. 1B. Therefore, we could estimate optimum water absorptions for given flours if NIR-HS or gluten values and FP were known. At a wheat quality laboratory like ours, it is easier to use the NIR-HS values rather than gluten parameters because most of our samples are received as wheats that we experimentally milled into flours.

Relationships Between Bake and Gluten or Pasting Parameters

Bake water absorptions and LV had significant positive phenotypic correlations with WG, DG, and GHA (Table VI), suggesting that the large LV could be obtained by increasing gluten content as the effect of FP (Graybosch et al 1993). Correlation coefficients were significant among growing regions between LV and WG ($r_r = 0.89$, $P < 0.05$), DG ($r_r = -0.92$, $P < 0.01$), and GHA ($r_r = 0.88$, $P < 0.05$) whereas they were not significant among cultivars, probably because of relatively limited genetic variations in gluten quantity for our sample set.

Negative correlations were shown between LV regression (LVR) and WG, DG, and GHA (Table VI). The term LVR was defined as a change in LV (cm^3)/change in 1 percentage point of FP (Finney 1985). In addition to the negative phenotypic correlations, there were negative correlations among cultivars and growing regions between LVR and WG ($r_c = -0.63$, $r_r = -0.86$; $P < 0.05$), DG ($r_c = -0.65$, $r_r = -0.86$; $P < 0.05$), and GHA ($r_c = -0.61$, $r_r = -0.85$; $P < 0.05$). Other researchers reported that an increase in gliadin protein is responsible for the increase in wheat protein content and weakening gluten elasticity (Doekes and Wennekes 1982, Huebner and Bietz 1988, Janssen 1990, Gupta et al 1992, Blumenthal et al 1993). Flours with lower monomeric-to-polymeric protein ratios had larger bread LV than flour with a higher ratio (MacRitchie 1989, Bean et al 1998). Thus, the negative correlations between gluten contents and LVR could be, at least in part, caused by increases in gliadin-to-glutenin or monomeric-to-polymeric protein ratios resulting from increases in protein quantity. The GI, representing gluten elasticity, showed negative correlations with DG and WG in this sample set. Chung and Seabourn (1990) reported a significant positive relationship between GI and LVR in hard winter wheat breeding lines with GI values ranging from 53.5 to 98.6% ($r = 0.643$, $n = 38$). However, our sample set did not show a significant correlation between GI and LVR, probably due to a narrower variation in GI (74.0–99.5%) or gluten quality.

Flour pasting properties showed lower correlation coefficients with baking parameters than gluten parameters did. The PT that

TABLE IV
Correlation Coefficients Between Gluten or Mixograph Parameters and Near-Infrared Hardness Scores^a

Quality Parameter	Near-Infrared Reflectance Hardness Score
Flour protein content (%) ^b	ns
Gluten parameters	
Wet gluten content (%)	0.53
Dry gluten content (%)	0.48
Gluten hydration amount	0.55
Gluten index (%)	-0.44
Gluten hydration capacity (%/%)	0.32
Dry gluten/flour protein (%/%)	0.63
Conventional mixograph	
Water absorption (%) ^b	0.53
Mix time (min)	-0.42
Computer-analyzed mixograph	
Midline peak time (min)	-0.50

^a $n = 72$; $P < 0.01$; ns = correlation coefficient is not significant at $P < 0.01$.

^b 14% moisture basis of flour.

TABLE V
Correlation Coefficients Between Gluten and Mixograph Parameters (%)^{a,b}

Mixograph Parameter	Flour Protein	Gluten Parameter		
		Wet Gluten	Dry Gluten	Gluten Hydration
Water absorption (%)	0.86	0.87	0.86	0.87
Computer-analyzed mixograph				
Midline peak				
Height	0.81	0.76	0.77	0.75
Width	0.68	0.42	0.48	0.38
Left of envelope peak				
Slope	0.75	0.71	0.71	0.70
Bandwidth	0.49	0.41	0.43	0.40
Right of envelope peak				
Bandwidth	0.74	0.59	0.63	0.56

^a $n = 72$; $P < 0.01$.

^b 14% moisture basis of flour.

represents the first viscosity increase in the pasting curve showed negative phenotypic correlations with LV ($r_p = -0.35$, $P < 0.01$) and LVR ($r_p = -0.33$, $P < 0.01$). The PT had a significant r_r value of -0.91 ($P < 0.05$) and an r_c value of -0.71 ($P < 0.01$) with LVR. These significant correlation coefficients suggest that the variation in PT, affected by cultivars or growing environments, can contribute to a variation in LV potential. The PT values were important enough to supplement FP (14% mb) for the evaluation of flour baking quality. The FP/PT ratios had a curvilinear relationship with LV with an R^2 of 0.726 ($n = 72$) and an RMSE of 45 ($r_c = 0.73$, $r_r = 0.99$; $P < 0.01$); $LV = 2,089 - 36,728 \times (FP/PT) + 309,221 \times (FP/PT)^2 - 775,156 \times (FP/PT)^3$. Parameters in this equation are significant at $P < 0.01$. The optimum FP/PT ratio for producing large LV was in the range of 0.17–0.18. This result suggests that interactions of proteins with starch granules that were swelling during baking could play a role in determining bread LV. Starch granules become partially soluble and hydrophilic as they swell in heated water, which contributes to increases in viscosity at the initial stage of pasting (Hoseney et al 1977). During swelling, starch granules can interact with gluten proteins, forming a starch-gluten matrix. Swollen starch granules have been observed by microscope to interact with gluten proteins (Bechtel et al 1978, Pomeranz et al 1984). The starch-gluten matrix surrounds and reinforces the liquid film of gas cells at the initial stage of dough expansion, contributing to increasing LV (Gan et al 1990). Thus, an optimum increase in swelling of starch granules accompanied with high FP can increase LV. Excessive swelling of starch granules can limit free water available to gluten proteins and result in low LV caused by an early rupture of gas cells by setting the liquid film to a rigid state (Sanstead 1961). We also found a decrease in LV at high FP/PT ratios.

Relationships Between Mixograph and Baking Parameters

Bake water absorptions were significantly correlated with conventional mixograph water absorptions (Table VI). A significant phenotypic correlation occurred between the mixograph and bake mix times. Bake mix times also had significant phenotypic correlations with computer-analyzed mixograph parameters including MPT, MPW, BRE, and B6M. Among cultivars, bake mix times and MPT showed a significant r_c value of 0.84 ($P < 0.01$). Significant correlations among cultivars and regions also occurred between

bake mix times and computer-analyzed mixograph parameters including MPW ($r_c = 0.83$, $P < 0.01$; $r_r = 0.85$, $P < 0.05$) and B6M ($r_c = 0.89$, $P < 0.01$; $r_r = 0.91$, $P < 0.05$).

Mixograph parameters were significantly correlated to LV except for mixograph mix time and MPT (Table VI), probably due to the influence of protein on these parameters. Slaughter et al (1992) and Souza et al (1993) also reported the significant correlations of LV with mixograph parameters. The LV were correlated to MPH ($r_r = 0.86$, $P < 0.05$), SLE ($r_r = 0.87$, $P < 0.05$), MPW ($r_r = 0.90$, $P < 0.05$), and B6M ($r_r = 0.90$, $P < 0.05$) among growing regions but not among cultivars. In this experiment, the LVR generally had negative correlations with mixograph parameters. Those mixograph parameters are also possibly influenced by the quantitative effect of proteins. Significant correlation coefficients between baking and conventional and computer-analyzed mixograph parameters indicate that computer-analyzed mixograph parameters can be applied to evaluate baking quality, especially for breeding lines.

CONCLUSIONS

A multiple regression equation calculated from FP and gluten parameters explained the 54.3% of variation in NIR-HS, suggesting the influence of gluten protein quality and quantity on hard winter wheat kernel hardness. The NIR-HS had a positive correlation with mixograph water absorptions, but a negative correlation with mixograph mixing times. Multiple regression indicated that 84 and 81% of the variations in mixograph water absorptions resulted from FP and NIR-HS and FP and gluten parameters, respectively.

Correlation coefficients between mixograph parameters suggested that the conventional analysis of a mixogram could be replaced with an objective computer analysis. The conventional mixograph mix time and mixing tolerance can be replaced by the MPT and B6M, respectively.

Gluten parameters (WG, DG, and GHA) were significantly correlated to bake water absorptions and LV. Among pasting parameters, the PT had the most significant negative correlations with LV and LVR. The FP/PT ratios had a curvilinear relationship with LV ($R^2 = 0.725$, $n = 72$), suggesting the beneficial influence of the interactions between proteins and starch granules during swelling on LV. The optimum FP/PT ratio was in the range of 0.17–0.18 for

TABLE VI
Correlation Coefficients Between Baking and Gluten and Mixograph Parameters^{a,b}

Quality Parameter	Baking Parameters			
	Water Absorption (%)	Mix Time (min)	Loaf Volume	Loaf Volume Regression (cm ³ /% FP) ^c
Protein content (%)	0.82	0.39	0.77	-0.56
Gluten parameters				
Wet gluten content (%)	0.82	ns	0.61	-0.55
Dry gluten content (%)	0.83	ns	0.65	-0.55
Gluten hydration amount (%)	0.81	ns	0.58	-0.54
Conventional mixograph				
Water absorption (%)	0.86	ns	0.58	-0.56
Mix time (min)	ns	0.83	ns	ns
Computer-analyzed mixograph				
Midline peak				
Time (min)	ns	0.64	ns	ns
Height	0.77	ns	0.64	-0.43
Width	0.44	0.70	0.51	-0.42
Left of envelope peak				
Slope	0.69	ns	0.60	-0.38
Right of envelope peak				
Bandwidth	0.65	0.49	0.60	-0.40
Bandwidth at 6 min	0.41	0.73	0.49	-0.37

^a $n = 72$; $P < 0.01$; ns = correlation coefficient is not significant at $P < 0.01$.

^b 14% moisture basis of flour.

^c FP = flour protein content on 14% moisture basis.

producing large LV. Baking parameters had significant correlations with computer-analyzed mixograph parameters. Our results indicate that the Glutomatic System, RVA, and MIXSMART could be considered as valuable tools for flour quality evaluation of hard winter wheats. Because correlation coefficients in this sample set were estimated based on hard winter wheat cultivars grown in Kansas, there have been relatively small variations in quality parameters. Thus, further research may be needed to investigate applications to other wheat sets that have wider genetic and environmental variations.

LITERATURE CITED

- Bean, S. R., Lyne, R. K., Tilley, K. A., Chung, O. K., and Lookhart, G. L. 1998. A rapid method for quantitation of insoluble polymeric protein in flour. *Cereal Chem.* 75:374-379.
- Bechtel, D. B., Pomeranz, Y., and De Francisco, A. 1978. Bread making studied by light and transmission electron microscopes. *Cereal Chem.* 55:392-401.
- Blokma, A. H., and Bushuk, W. 1988. Rheology and chemistry of dough. Pages 131-218 in: *Wheat: Chemistry and Technology*. Vol II. Y. Pomeranz, ed. Am. Assoc. Cereal Chem.: St. Paul, MN.
- Blumenthal, C. S., Barlow, E. W., and Wrigley, C. W. 1993. Growth environment and wheat quality: The effect of heat stress on dough properties and gluten proteins. *J. Cereal Sci.* 18:3-21.
- Buckley, D. J., Lukow, O. M., Lalonde, J. L. M., and Woods, S. M. 1990. A multilevel computer system for dough quality analysis and report distribution. *Cereal Foods World* 35:472-478.
- Chung, O. K., and Seabourn, B. W. 1990. Effect of growing years on gluten quality of hard winter wheats from two nurseries. Pages 724-739 in: *Gluten Proteins 1990*. W. Bushuk and R. Tkachuk, eds. Am. Assoc. Cereal Chem.: St. Paul, MN.
- Dachketvitch, T., and Autran, J.-C. 1989. Prediction of baking quality of bread wheats in breeding programs by size-exclusion liquid chromatography. *Cereal Chem.* 66:448-456.
- Deffenbaugh, L. B., and Walker, C. E. 1989. Comparison of starch pasting properties in the Brabender Viscoamylograph and the Rapid Visco-Analyzer. *Cereal Chem.* 66:493-499.
- Doekes, G. J., and Wennekes, M. J. 1982. Effect of nitrogen fertilization on quantity and composition of wheat flour protein. *Cereal Chem.* 59:276-278.
- Farrand, E. A. 1964. Flour properties in relation to the modern bread process in the United Kingdom, with special reference to α -amylase and starch damage. *Cereal Chem.* 41:98-111.
- Finney, K. F. 1985. Experimental breadmaking studies, functional (bread-making) properties, and related gluten protein fractions. *Cereal Foods World* 30:794-801.
- Finney, K. F. 1989. A 5-g mixograph to determine and predict functional properties of wheat flours. *Cereal Chem.* 66:527-530.
- Finney, K. F., and Shogren, M. D. 1972. A 10-g mixograph for determining and predicting functional properties of wheat flours. *Baker's Dig.* 46(2):32-42, 77.
- Gan, Z., Angold, R. E., Williams, M. R., Ellis, P. R., Vaughan, J. G., and Galliard, T. 1990. The microstructure and gas retention of bread dough. *J. Cereal Sci.* 12:15-24.
- Gras, P. W., Hibberd, G. E., and Walker, C. E. 1990. Electronic sensing and interpretation of dough properties using a 35-g mixograph. *Cereal Foods World* 35:568-571.
- Graybosch, R., Peterson, C. J., Moore, K. J., Stearns, M., and Grant, D. L. 1993. Comparative effects of wheat flour protein, lipid, and pentosan composition in relation to baking and milling quality. *Cereal Chem.* 70:95-101.
- Greenblatt, G. A., Bettge, A. D., and Morris, C. F. 1995. Relationship between endosperm texture and occurrence of friabilin and bound lipids on wheat starch. *Cereal Chem.* 72:172-176.
- Greenwell, P., and Schofield, J. D. 1986. A starch granule protein associated with endosperm softness in wheat. *Cereal Chem.* 63:379-380.
- Gupta, R. B., Batey, I. L., and MacRitchie, F. 1992. Relationship between protein composition and functional properties of wheat flours. *Cereal Chem.* 69:125-131.
- Huebner, F. R., and Bietz, J. A. 1988. Quantitative variation among gliadins of wheats grown in different environments. *Cereal Chem.* 65:362-366.
- Huebner, F. R., and Gaines, C. S. 1992. Relation between kernel hardness, environment, and gliadin composition. *Cereal Chem.* 69:148-151.
- Hutchinson, J. B. 1966. The paste viscosities of wheat starch and flour-water mixtures on cooking. I. An interaction of the Hagberg falling number, Brabender Amylograph and α -amylase activities of Australian and English flour. *J. Sci. Food. Agric.* 17:198-201.
- ICC. 1995. Standard Methods of the International Association for Cereal Science and Technology. Method 106/2, approved 1984; Standard 137/1, approved 1982 and revised 1994. The Association: Vienna.
- Janssen, A. M., Vereijken, J. M., Beintema, J. J., Witholt, B., and Van Vliet, T. 1990. Rheological studies on gluten. Page 180-186 in: *Gluten Proteins 1990*. W. Bushuk and R. Tkachuk, eds. Am. Assoc. Cereal Chem.: St. Paul, MN.
- Johnson, J. A., Swanson, C. O., and Bayfield, E. G. 1943. The correlation of mixogram with baking results. *Cereal Chem.* 20:625-644.
- Kruger, J. E., and Lineback, D. R. 1987. Carbohydrate-degrading enzymes in cereals. Page 117-140 in: *Enzymes and Their Role in Cereal Technology*. J. E. Kruger, D. Lineback, and C. E. Stauffer, eds. Am. Assoc. Cereal Chem.: St. Paul, MN.
- MacRitchie, F. 1989. Identifying the baking quality related components of wheat flours. *Cereal Foods World* 34:548-552.
- Martinant, J. P., Nicolas, Y., Bouguennec, A., Popineau, Y., Saulnier, L., and Branland, G. 1998. Relationships between mixograph parameters and indices of wheat grain quality. *J. Cereal Sci.* 27:179-189.
- Mathewson, P. R., and Pomeranz, Y. 1978. Hot-paste viscosity and α -amylase susceptibility of hard red winter wheat flour. *J. Food Sci.* 43:60-63.
- Merritt, P. P., and Stamborg, O. E. 1941. Some studies on flour absorption. *Cereal Chem.* 18:632-639.
- Nakai, S., and Li-Cahn, E. 1993. Recent advances in structure and function of food proteins: QSAR approach. *Crit.Rev. Food Sci. Nutr.* 33:477-512.
- Navickis, L. L., Rubenthaler, G. S., Nelsen, T. C., and Butterfield, R. O. 1990. Evaluating mixogram parameters by electronic torque sensing. *Cereal Foods World* 35:493-496.
- Neufeld, K. J., and Walker, C. E. 1990. Evaluation of commercial wheat gluten using the mixograph. *Cereal Foods World* 35:667-669.
- Ohm, J. B., Chung, O. K., and Deyoe, C. W. 1998. Single kernel characteristics of hard winter wheats in relation to milling and baking quality. *Cereal Chem.* 75:156-161.
- Olkku, J., Fletcher, III, S. W., and Rha, C. 1978. Studies on wheat starch and wheat flour model paste system. *J. Food Sci.* 43:52-59.
- Perten, H. 1990. Rapid measurement of wet gluten quality by the gluten index. *Cereal Foods World* 35:401-402.
- Pomeranz, Y., Meyer, D., and Seibel, W. 1984. Wheat, wheat-rye, and rye dough and bread studied by scanning electron microscopy. *Cereal Chem.* 61:53-59.
- Pon, C. R., Lukow, O. M., and Buckley, D. J. 1989. A multichannel, computer-based system for analyzing dough rheology. *J. Texture Stud.* 19:343-360.
- Rath, C. R., Gras, P., Wrigley, C. W., and Walker, C. E. 1990. Evaluation of dough properties from 2 g of flour using the mixograph principles. *Cereal Foods World* 35:572-574.
- Ross, A. S., Walker, C. E., Booth, R. I., Orth, R. A., and Wrigley, C. W. 1987. The Rapid Visco-Analyzer: A new technology for the estimation of sprout damage. *Cereal Foods World* 32:827-829.
- Rubenthaler, G. L., and King, G. E. 1986. Computer characterization of mixogram and their relationship to baking performance. Pages 131-168 in: *Fundamentals of Dough Rheology*. H. Fardi and J. M. Faubion, eds. Am. Assoc. Cereal Chem.: St. Paul, MN.
- Sandsted, R. M. 1961. The function of starch in the baking of bread. *Baker's Dig.* 35 (3):36-42, 44.
- Shogren, M. D. 1990. A short history of the mixograph. *Cereal Foods World* 35:480-482.
- Slaughter, D. C., Norris, K. H., and Hruschka, W. R. 1992. Quality and classification of hard red wheat. *Cereal Chem.* 69:428-432.
- Souza, R., Tyler, J. M., Kephart, K. D., and Kruk, M. 1993. Genetic improvements in milling and baking quality of hard red spring wheat varieties. *Cereal Chem.* 70:280-285.
- Sterns, M. M., and Barta, S. 1990. Mixograph standardization/digital data acquisition. *Cereal Foods World* 35:485-488.
- Swanson, C. O. 1941. Factors which influence the physical properties of dough. III. Effect of protein content and absorption on the pattern of curves made on the recording dough mixer. *Cereal Chem.* 18:615-627.
- Swanson, C. O., and Johnson, J. A. 1943. Description of mixograms.

Cereal Chem. 20:39-42.

Swanson, C. O., and Working, E. B. 1933. Testing of the quality of flour by the recording dough mixer. Cereal Chem. 10:1-29.

Walker, C. E., Ross, A. S., Wrigley, C. W., and McMaster, G. J. 1988. Accelerated starch-paste characterization with the Rapid Visco-Analyzer. Cereal Foods World 33:491-494.

Wikström, K., and Bohlin, L. 1996. Multivariate analysis as a tool to predict bread volume from mixogram parameters. Cereal Chem. 73:686-690.

Wrigley, C. W., and Bietz, J. A. 1988. Proteins and amino acids. Pages 159-275 in: Wheat Chemistry and Technology. Vol. 1. Y. Pomeranz, ed. Am. Assoc. Cereal Chem.: St. Paul, MN.

[Received June 18, 1998. Accepted March 31, 1999.]