A Rapid, Small-Scale Sedimentation Method to Predict Breadmaking Quality of Hard Winter Wheat

Bradford W. Seabourn,* Zhihua S. Xiao, Michael Tilley, Thomas J. Herald, and Seok-Ho Park

ABSTRACT

Breeders and processors are always looking for rapid and accurate methods to evaluate wheat (*Triticum aestivum* L.) quality. The objective of this study was to develop a rapid, small-scale method to accurately determine breadmaking quality for early generation hard winter wheat (HWW) breeding lines by combining the solutions used in the sodium dodecyl sulfate (SDS) sedimentation method (American Association of Cereal Chemists [AACC] 56-70 [AACC, 2000]) and the centrifugation process found within the solvent retention capacity (SRC) method (AACC 56-11 [AACC, 2000]). A preliminary test of the hybrid SDS-SRC method was performed on eight HWW varieties and compared to AACC (56-70) and in-house Hard Winter Wheat Quality Laboratory (HWWQL) methods to show proof of concept. Further validation of the hybrid method was conducted on a diverse set of 53 HWW varieties. The hybrid method was performed in 66% less time than AACC 56-70 and HWWQL methods. Furthermore, sample size was reduced from 6 g for the AACC method to 1 g for the hybrid method. Results obtained from the hybrid method exhibited a higher correlation to bread loaf volume ($r \geq 0.84$) compared to results from the AACC method ($r > 0.42$) and HWWQL method ($r \geq 0.64$) for wheat flour. Due to enhanced speed, accuracy, and simplicity the hybrid SDS-SRC sedimentation method may prove useful in breeding programs, grain elevators, and other scenarios where rapid assessment of end-use quality determination is required.

B.W. Seabourn, Z.S. Xiao, M. Tilley, T.J. Herald, USDA-ARS, Center for Grain & Animal Health Research, Hard Winter Wheat Quality Laboratory, 1515 College Avenue, Manhattan, KS 66502; Seok-Ho Park, International Delights LLC, 230 Brighton Road, Clifton, NJ 07012. Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the USDA. The USDA is an equal opportunity provider and employer. Received 15 Aug. 2011. *Corresponding author (brad.seabourn@ars.usda.gov).

Abbreviations: AACC, American Association of Cereal Chemists; HWW, hard winter wheat; HWWQL, Hard Winter Wheat Quality Laboratory; mb, moisture basis; rpm, revolutions per minute; SDS, sodium dodecyl sulfate; SRC, solvent retention capacity; WQC, Wheat Quality Council.

One important goal in wheat (*Triticum aestivum* L.) breeding is to develop high-yielding cultivars that possess the functional attributes demanded by producers, processors, and consumers. This is no small feat because producers strive for high yielding crops, processors require highly functional flour, and consumers demand and expect excellent sensory qualities in the end products.

Development of a commercial wheat cultivar requires 10 to 12 yr at an estimated cost of US$2 million. Wheat breeders welcome new technologies that offer a means to predict processing and end-product quality of early generation lines that may reduce the investment (time and money) to bring a cultivar to the final stages of commercialization.

Historically, breeders have relied on the fact that higher protein content in wheat is moderately correlated to higher loaf volume in the final product. Finney (1943) and Finney and Barmore

Published in Crop Sci. 52:1306–1315 (2012).
doi: 10.2135/cropsci2011.04.0210

(1948) found that when an optimized baking formula was used, the relationship between protein content and loaf volume was linear for a given wheat cultivar over the protein range examined (8–18%). Over the years, various methods have been developed to better predict the quality of flours for breadmaking. Currently, breeders use a number of more sophisticated wheat functionality tests to assist them in making decisions on whether or not to proceed with the development of a given experimental line.

Over the past several decades, hard winter wheat (HWW) breeders have routinely relied on the well-established American Association of Cereal Chemists (AACC) 56-70 sodium dodecyl sulfate (SDS) sedimentation method (AACC, 2000) for predicting breadmaking quality of wheat since the test does not require a large amount of sample, which is typically a limiting factor in breeding programs. The SDS sedimentation test is a good indicator of gluten quantity and quality because they are related to end-product quality (Axford et al., 1978, 1979; Preston et al., 1982; Dick and Quick, 1983). Although the SDS sedimentation method is highly regarded, the test does have limitations, namely (i) the SDS sedimentation values are not linearly correlated with bread loaf volume at high flour protein levels (>140 g kg^{-1}) (Preston et al., 1982; Ayoub et al., 1993) and (ii) the method is sensitive to processing variables and therefore are prone to operator error; for example, a slight change in shaking time can considerably alter the results.

The original sedimentation test developed by Zeleny (1947) measured the rate of wheat flour sedimentation from an acidic suspension. Over time, the sedimentation method was generally accepted to be a useful indicator of gluten quality for wheat products. Many modifications of the sedimentation procedure have occurred over the last fifty years leading up to the current approved method.

The AACC method 56-70 requires approximately 30 min to conduct (not including the time to weigh sample). Although the AACC 56-70 sedimentation test has been used effectively, breeders and quality labs are always developing improved methods to reduce analysis time while maintaining accuracy and reproducibility.

Another method that is used to test wheat gluten quality is the solvent retention capacity (SRC) test (AACC method 56-11; AACC, 2000), which uses 5% lactic acid as a solvent. The SRC method employs centrifugation to assist separating the supernatant from the flour biomacromolecular precipitate (starch and gluten) (Gaines, 2000). Guttieri et al. (2004) reported that the SRC test was successful in predicting the quality of soft wheat products. Xiao et al. (2006) reported a strong correlation (r = 0.83) between the SRC method using 5% lactic acid and HWW bread loaf volume. Unfortunately, the limiting factor of the SRC method is that 50 min is required to perform the analysis.

Therefore, the objective of this study was to develop a rapid, small-scale method to accurately determine breadmaking quality for early generation HWW breeding lines by combining the solutions used in the SDS sedimentation method (AACC 56-70) and the centrifugation process found within the SRC method (AACC 56-11).

MATERIALS AND METHODS

Sample and Sample Preparation

A set of eight HWW samples selected from samples submitted to the 2006 Wheat Quality Council (WQC) milling and baking test evaluations were used ('Overley' and 'Fuller' from Kansas, NE01643 from Nebraska, OK93P656H3299-2C04, OK01420, and OK02405 from Oklahoma, SD01W064 from South Dakota, and TX01D3232 from Texas) to develop a rapid and small-scale hybrid SDS sedimentation–SRC method. Wheat samples were milled into straight-grade flour using a Miag Multomat mill (Buhler, Duisburg, Germany). Ground whole wheat meal (hereafter called "wheat meal") was prepared from approximately 18 g of wheat kernels using a Udy cyclone mill (Udy Corp., Fort Collins, CO). Kernels were finely ground to a uniform size for passage through a 1-mm screen (Guttieri et al., 2004). The ground meal was placed in glass jars and further mixed for 5 min to provide a homogeneous mixture.

A set of 53 HWW samples was used in the validation study and had greater variation in quality characteristics. Samples were provided by the Hard Winter Wheat Quality Laboratory (Manhattan, KS) (HWWQL), which is one of four federal wheat quality laboratories within the USDA and whose mission is to assist U.S. HWW breeders in their screening decisions by providing end-use quality evaluation of advanced experimental wheat lines before commercial release. Samples were randomly selected from 600 breeding lines harvested in 2006, including experimental lines from the USDA Regional Performance Nursery as well as entries from the WQC HWW evaluations. The WQC is the nonprofit organization in the United States whose goal is to improve the value of all U.S. wheat classes for producers, millers, and processors through the annual evaluation of the end-use quality of advanced breeding lines. Wheat samples were then milled into straight-grade flour using a Brabender Quadrumat Senior experimental mill (Brabender, GmbH & Co., Duisburg, Germany). Wheat meal was prepared as described above.

Proximate Analyses

Protein content was estimated by near-infrared analysis (AACC-approved method 39-11; AACC, 2000). Ash and moisture contents were determined according to AACC-approved methods 08-01 and 44-15A (AACC, 2000), respectively. Analytical data are reported on a 140 g kg^{-1} moisture basis (mb).

Official AACC 56-70 SDS Sedimentation Method

In the AACC method 56-70, 6.3 g of sample was dispersed in 50 mL of distilled and deionized water and shaken 6 min. Then 50 mL of a solution containing 3% (w/v) SDS and 0.002 g mL^{-1} lactic acid was added. The solution was then shaken for an additional 4 min. The final concentration of lactic acid was 0.001 g mL^{-1} and SDS was 0.015 g mL^{-1}. The contents were allowed to settle for 20 min after which the volume of sediment was recorded. The total time required to complete the analysis was 30 min. The official AACC method 56-70 was conducted in the preliminary test to be used as a benchmark when evaluating the feasibility of the in-house HWWQL and the proposed hybrid sedimentation methods.

In-House HWWQL Sedimentation Method

Since 1994, the HWWQL has used an in-house sedimentation method based on modifications to AACC method 56-70 to address the limited sample quantity often seen with breeding programs. Xiao et al. (2006) described the modified AACC method and reported the correlation between the results obtained using the modified method and breadmaking quality. The in-house HWWQL method was used in both preliminary and validation tests. Modification of AACC 56-70 included a reduced sample analysis weight (2 g) and a 50% reduction in solvent as well as implementation of a continuous (automated) shaking process throughout the assay. The in-house HWWQL method requires 36 min (16 min shaking time and 20 min sedimentation time). The samples evaluated using the in-house HWWQL method were prepared as follows: 2.0 g (140 g kg^{-1} mb) sample of wheat flour or wheat meal was suspended in 20 mL of aqueous bromophenol blue (0.004 mg mL^{-1}) solution and shaken vigorously by hand for 5 sec in a 100 mL sealed graduated cylinder followed by shaking for 4 min on an automated wrist-action (table-top) shaker at 40 revolutions per minute (rpm) (Merkle-Korff Gear Co., Chicago, IL). Then 20 mL of 2.5% (w/w) SDS solution was added, and the resulting solution was mixed by automated wrist shaker for an additional 6 min at 40 rpm. This was followed by the addition of 10.0 mL 0.005 g mL^{-1} lactic acid solution and further mixing by automated shaking for another 6 min. The final concentration of the solution containing lactic acid was 0.001 g mL^{-1} and 0.01 g mL^{-1} SDS. The cylinder contents were allowed to settle for 20 min after which the volume of sediment was recorded.

Hybrid SDS-SRC Sedimentation Method

This new method is based on a combination of the HWWQL in-house method and SRC test AACC method 56-11. Four combinations of processing conditions were studied that varied shaking and centrifugation times (2 min shaking with 1 min centrifugation, 2 min shaking with 2 min centrifugation, 4 min shaking with 1 min centrifugation, and 4 min shaking with 2 min centrifugation).

Reagent Preparation

Twenty-five milliliters of 85% lactic acid solution (Fisher Scientific, St. Louis, MO) was added to 200 mL water resulting in a lactic acid concentration of 0.09 g mL^{-1}. This "stock" solution was refluxed for 6 h to reduce the aggregation tendency of the lactic acid molecules. A lactic acid "working" solution was made by mixing the stock solution with water at a ratio of 50 mL to 950 mL water, in which the final concentration of working solution was 0.0047 g mL^{-1}. A 1.25% solution of SDS was prepared by dissolving 12.5 g of SDS in 987.5 mL water. The final test solution is a mixture of the lactic acid working solution and the SDS solution and contains 0.001 g mL^{-1} lactic acid and 0.01 g mL^{-1} SDS, which is the same as the in-house HWWQL method.

Sample Preparation and Processing Conditions Used in the Hybrid SDS-SRC Sedimentation Method

The hybrid SDS-SRC method required a 1.0-g sample size. This is a 50% reduction compared to the in-house HWWQL method. The solution concentration used in the hybrid SDS-SRC method was the same as that used for the in-house HWWQL method, although the solution volume required to conduct the analysis was reduced from 50 to 25 mL. In addition, the hybrid SDS-SRC method utilized centrifugation rather than sedimentation to analyze results. The procedure was as follows: 1.0 g wheat meal or flour was weighed and placed in a preweighed 50 mL polypropylene tube followed by the addition of 5 mL of 0.47% lactic acid. After capping the tube, samples were mixed on a small vibratory mixer for 6 sec and then 20 mL of 1.25% SDS solution was added. The tubes were capped and mixed again for 6 sec. The tubes were shaken on a platform shaker (Innova 2000, New Brunswick Scientific, Edison, NJ) at 300 rpm for either 2 or 4 min (according to the experimental design). The tubes were centrifuged at 3200 × *g* in a swinging bucket rotor (Eppendorf 5810R, Brinkmann Instruments Inc., Westbury, NY) for either 1 or 2 min (according to the experimental design). Following centrifugation, the supernatant was carefully removed by decanting so as not to disturb the precipitate. A paper towel was used to wipe any remaining foam that was visually identified inside the tube. The tube was capped and weighed to determine the sedimentation weight value, which was calculated using the following formula similar to AACC 56-10 and AACC 56-11 (AACC, 2000):

$$\text{weight value (\%)} = ((\text{pellet weight}/\text{flour weight}) \times \{[86/(100 - \text{percent flour moisture})]\} - 1) \times 100.$$

Straight Dough Baking Method

An optimized straight dough baking method (AACC method 10-10B; AACC, 2000) was used for the experimental breadmaking test. The bread formulation contained 100 g flour, 11 mL of a solution containing 6 g sucrose and 1.5 g sodium chloride, and 5 mL of aqueous malt mixture containing (0.25 g) dried malt, dry active yeast (1 g), shortening (3 g), and ascorbic acid (5 mg). Bake water absorption and mix time were estimated based on mixograph data and adjusted to optimum as determined by the appearance and feel of the dough by an experienced baker. Dough was fermented for 90 min in a proofing cabinet set at 30°C with a relative humidity of 86%. Loaves were baked at 218.3°C (425°F) for 18 min and were weighed immediately after removal from the oven. The bread volumes were measured by rapeseed displacement.

A trained baker graded the bread for crumb grain on a scale from 0 to 6: 0 for "unsatisfactory," 2 for "questionable," 4 for "satisfactory," and 6 for "outstanding" as described by Park et al. (2004).

Statistical Analysis

Statistical analysis of data was performed using the Statistical Analysis System (v. 8.0; SAS Institute, 1999). Statistical methods utilized were simple correlation coefficient (r), coefficient of determination (R^2), $p < 0.05$, $p < 0.01$, $p < 0.001$, $p < 0.0001$, and linear regression. The reproducibility of each method was assessed by comparing the results of triplicates of sample analysis in the preliminary test and duplicates for the validation test; in all cases, the mean value (not the individual analysis) of each sample for each method (e.g., shaking time) was correlated with the common quality traits using the Pearson simple correlation analysis.

RESULTS AND DISCUSSION

Preliminary Investigation of Processing Parameters

The first phase of the research was to determine the processing parameters for the hybrid SDS-SRC method (shaking speed, shaking time, and centrifugation time). This was performed on a set of eight HWW samples selected from the 2006 WQC. Test weights for this group ranged from 726 to 795 kg hL^{-1} (mean 766 kg hL^{-1}) while the variation in wheat protein content (140 g kg^{-1} mb), flour protein content (140 g kg^{-1} mb), and ash content (140 g kg^{-1} mb) were 120 to 148 g kg^{-1} (mean 132 g kg^{-1}), 104 to 133 g kg^{-1} (mean 118 g kg^{-1}), and 3.7 to 4.6 g kg^{-1} (mean 4.0 g kg^{-1}), respectively. Variation in breadmaking parameter results for water absorption, mixograph mix time, mixing tolerance (0–6 scale), loaf volume, and crumb grain score (0–6 scale) were 617 to 656 g kg^{-1} (mean 634 g kg^{-1}), 2.88 to 5.88 min (mean 4.36 min), 0.0 to 5.0 (mean 4.0), 745 to 1000 cm^3 (mean 876 cm^3), and 2.3 to 4.2 (mean 3.6), respectively.

Experimental data showed the optimal shaker speed to be 300 rpm. In this proof of concept stage, the hybrid SDS-SRC method was initiated by employing a 10 min shake time as described in AACC 56-70 and slowly reduced the shaking time at 2 min intervals from 10 min down to 2 min. Therefore, shaking times of 2, 4, 6, 8, and 10 min were investigated at a fixed centrifugation time of 1 min. Centrifugation was employed as a means to expedite the sedimentation rate as described in AACC 56-11. The minimum amount of time that the centrifuge could be programmed was 1 min. To determine if additional centrifugation time improved sedimentation, a 2 min centrifugation time was used for comparison.

No significant differences were observed among all shake times (data not shown). Therefore, the 2 and 4 min shake times were selected since both exhibited an r value above 0.90 between the hybrid SDS-SRC method and bread loaf volume. This phase of the research suggested that a reduction in processing time was successful.

Evaluation of Processing Conditions for the Hybrid SDS-SRC Method

The correlation coefficients between results from the hybrid SDS-SRC method and bread loaf volume using wheat flour and wheat meal from eight test samples were all greater than 0.90 for the 2 or 4 min shaking time and 1 min centrifugation (Table 1). It was observed that the 2 min shake time resulted in a weaker pellet more often than the 4 min shaking time even though the correlations coefficients were similar for the wheat flour samples. A firmer pellet (a sample that did not fall when inverted) was observed more frequently for wheat meal samples compared to wheat flour samples under the same processing conditions. Increasing the centrifugation time from 1 to 2 min under the same conditions described did not substantially change the correlation coefficient ($r > 0.90$) between loaf volume and sedimentation values (Table 1). However, the 2 min centrifugation time resulted in firmer pellets for the eight wheat flour and meal samples compared to 1 min centrifugation. Furthermore, the wheat meal samples exhibited firmer pellets than the flour samples under the same processing conditions. Therefore, to obtain higher consistency the 2 min centrifugation time was used for the remainder of the investigation. A total assay time of

Table 1. Comparison of three selected sedimentation methods to predict bread loaf volume using eight different hard winter wheat varieties.†

Sedimentation methods‡	Processing condition: Shaking time (min)	Sedimentation time (min)	Centrifugation time (min)	Sample size (g)	Correlation (r), Bread loaf volume: Wheat flour§	Wheat meal¶
AACC method 56-70 (AACC, 2000)	10	20	0	6.3	0.56	0.94
In-house HWWQL#	16	20	0	2.0	0.60	0.76
Hybrid SRC-SDS	2	0	1	1.0	0.90	0.95
	2	0	2	1.0	0.92	0.93
	4	0	1	1.0	0.94	0.91
	4	0	2	1.0	0.95	0.95

† $n = 8$.

‡ AACC, American Association of Cereal Chemists; HWWQL, Hard Winter Wheat Quality Laboratory, Manhattan, KS; SDS, sodium dodecyl sulfate; SRC, solvent retention capacity.

§ Correlation coefficients describe the relationship between the sedimentation value of wheat flour and bread loaf volume.

¶ Correlation coefficients describe the relationship between the sedimentation value of wheat meal and bread loaf volume.

In-house HWWQL method is a modified method based on AACC method 56-70 (AACC, 2000).

less than 10 min with the hybrid method represents a considerable shortening of overall assay time from the 30 min required for AACC 56-70 and 36 min for the in house HWWQL assay. Wang and Kovacs (2002) reported similar correlation coefficient values ($r > 0.90$) but a considerably longer shaking and centrifugation process totaling over 25 to 45 min depending on swelling time.

Correlation and Reproducibility among AACC 56-70, In-House HWWQL, and Hybrid SDS-SRC Sedimentation Methods

Results from the hybrid SDS-SRC method exhibited a stronger correlation to bread loaf volume in flour compared to results from the other methods evaluated. Regardless of processing conditions, results from the hybrid SDS-SRC method were highly correlated to bread loaf volume ($r > 0.90$) whereas results from the AACC method 56-70 and in-house HWWQL methods exhibited r values of 0.56 and 0.60, respectively, for the eight wheat flour samples (Table 1). Xiao et al. (2006) analyzed and correlated bread loaf volume to sedimentation values using the in-house HWWQL method and reported $r = 0.72$ for 116 HWW flour samples that exhibited a large range in breadmaking properties. Axford et al. (1979) reported $r = 0.80$ and Oelofse (2008) reported $r = 0.53$ between loaf volume and AACC method 56-70. Compared to the AACC 56-70 method, the hybrid SDS-SRC method was conducted in 66% less time, used an 84% smaller sample size, and required 75% less lactic acid and SDS solution.

When using wheat meal as the test sample, results from the hybrid SDS-SRC method exhibited similar correlation values to loaf volume when compared to results using AACC method 56-70 ($r > 0.90$). The in-house HWWQL sedimentation method provided data whose correlation value for loaf volume was 0.76 for wheat meal (Table 1). Sapirstein and Suchy (1999) reported similar results ($r = 0.97$) for the correlation between wheat meal and bread loaf volume using AACC method 56-70.

The reproducibility of data derived from the hybrid SDS-SRC method was determined by evaluating the eight samples used in Table 1 for both flour (Table 2) and wheat meal (Table 3) in triplicate. Analysis of wheat flour using the hybrid SDS-SRC method produced results with similar reproducibility and CVs averaged 0.58 and 1.63 for 2 min shaking and 4 min shaking tests, respectively, compared to a CV of 0.89 for the AACC method 56-70. The HWWQL in-house test produced values that showed the greatest variability with an average CV of 2.22. In general, wheat meal displayed superior repeatability of test data resulting in less variability between samples and with a lower average CV for both hybrid test conditions than the AACC and in-house methods (Table 3).

Validation Study

To confirm our preliminary results, a larger, more diverse set of 53 HWW flour and meal samples milled from early generation lines was used in the validation study (Table 4). The AACC method 56-70 and the in-house HWWQL SDS sedimentation methods were used as methods of comparison in the validation study. The data suggested that the selected sample set represents a broad range of HWW quality. The sample set showed a large variation in protein content (105–168 g kg^{-1} for wheat protein and 93–161 g kg^{-1} for flour protein), and test weights ranged from 694 to 805 kg hL^{-1} (mean of 755 kg hL^{-1}). Furthermore, samples exhibited variability in mixograph characteristics including water absorption (58.3–69.8%), mix time (1.5–8.6 min), and tolerance (0–6). Consequently, bread baking results showed a wide range in loaf volumes (670–1200 cm^3) and crumb grain scores (1.0–5.0). These data are consistent with the ranges observed in other breeder samples (Xiao et al., 2006).

Table 2. Assessment of selected sedimentation methods for determining hard winter wheat flour quality on eight wheat varieties.†

Method	AACC‡ method 56-70 (AACC, 2000)			In-house HWWQL§			2 min shaking and 2 min centrifugation			4 min shaking and 2 min centrifugation		
Sample	Sedimentation volume (mL)	SD	CV (%)	Sedimentation volume (mL)	SD	CV (%)	WV¶ (%)	SD	CV (%)	WV (%)	SD	CV (%)
1	88.83	1.04	1.17	32.67	1.04	3.19	393.00	1.18	0.30	424.65	10.62	2.50
2	91.93	0.12	0.13	36.40	0.69	1.90	417.70	0.62	0.15	435.43	4.80	1.10
3	83.17	1.53	1.84	23.50	0.50	2.13	306.48	1.27	0.41	326.90	7.82	2.39
4	87.07	0.40	0.46	31.43	0.51	1.63	327.55	1.96	0.60	329.23	6.07	1.84
5	87.93	0.60	0.69	35.33	0.31	0.86	345.65	3.80	1.10	358.77	0.57	0.16
6	85.37	0.35	0.41	24.33	0.58	2.37	315.71	3.03	0.96	337.45	5.53	1.64
7	94.83	1.44	1.52	39.03	0.65	1.67	392.52	3.53	0.90	393.15	6.50	1.65
8	87.10	0.79	0.91	38.33	1.53	3.98	408.43	0.87	0.21	406.21	6.95	1.71
Average			**0.89**			**2.22**			**0.58**			**1.63**

† Values represent triplicate measurements.

‡ AACC, American Association of Cereal Chemists.

§ HWWQL, Hard Winter Wheat Quality Laboratory, Manhattan, KS.

¶ WV, weight value.

Table 3. Assessment of selected sedimentation methods for determining hard winter wheat meal quality on eight wheat varieties.†

Method Sample	AACC‡ method 56-70 (AACC, 2000) Sedimentation volume (mL)	SD	CV (%)	In-house HWWQL§ Sedimentation volume (mL)	SD	CV (%)	2 min shaking and 2 min centrifugation WV¶ (%)	SD	CV (%)	4 min shaking and 2 min centrifugation WV (%)	SD	CV (%)
1	95.67	0.58	0.60	27.10	0.66	2.42	333.26	2.95	0.89	339.42	2.38	0.70
2	96.17	0.29	0.30	28.60	0.17	0.61	346.37	2.26	0.65	345.71	4.88	1.41
3	72.83	0.29	0.40	18.00	0.00	0.00	273.24	3.13	1.15	275.25	1.35	0.49
4	73.90	1.35	1.83	21.87	0.12	0.53	270.23	2.00	0.74	276.36	1.38	0.50
5	80.23	1.12	1.40	23.17	0.65	2.81	283.00	2.02	0.71	287.05	1.74	0.61
6	77.17	1.04	1.35	22.00	0.20	0.91	279.11	1.98	0.71	281.82	1.72	0.61
7	87.33	0.58	0.66	22.57	0.40	1.79	304.37	1.54	0.51	306.35	0.29	0.09
8	94.77	0.93	0.98	27.10	0.36	1.33	312.65	1.26	0.40	311.89	5.20	1.67
Average			**0.94**			**1.30**			**0.72**			**0.76**

† Values represent triplicate measurements.
‡ AACC, American Association of Cereal Chemists.
§ HWWQL, Hard Winter Wheat Quality Laboratory, Manhattan, KS.
¶ WV, weight value.

Table 4. Composite quality parameters of 53 hard winter wheat cultivars used in the validation study selected from the hard winter wheat growing region during 2006 using the Hard Winter Wheat Quality Laboratory (HWWQL) (Manhattan, KS) in-house sedimentation method.†

Quality parameter‡	Mean	Range
Wheat characteristics		
Wheat protein content, g kg^{-1}	137	105–168
Flour protein content, g kg^{-1}	127	93–161
Ash content, g kg^{-1}	03.9	03.2–04.7
Mixograph characteristics		
Water absorption, g kg^{-1}	640.1	583–698
Mix time, min	3.6	1.5–8.6
Mixing tolerance	2.9	0.0–6.0
AACC method 56-70 (AACC, 2000)		
Sedimentation volume (wheat flour, mL)	87.4	20–96
Sedimentation volume (wheat meal, mL)	78.3	21–97
SDS sedimentation		
Sedimentation volume (wheat flour, mL)	37.3	21.5–43.5
Sedimentation volume (wheat meal, mL)	22.1	9.0–32.0
Bread characteristics		
Loaf volume, cm^3	914.4	670–1200
Crumb grain score	3.6	1.0–5.0

† Values are on a 140 g kg^{-1} moisture basis.
‡ AACC, American Association of Cereal Chemists; SDS, sodium dodecyl sulfate.

Correlation coefficients for the various processing conditions used to evaluate the 53 HWW varieties exhibited coefficients above 0.90 for both wheat flour and wheat meal (data not shown). The high correlation coefficients suggest that test results derived from the hybrid SDS-SRC method were consistent and repeatable throughout the diverse range of HWW samples analyzed using either processing condition.

Correlation Between Hybrid SDS-SRC Sedimentation Method and Wheat Flour Characteristics

Test results utilizing the hybrid SDS-SRC method provided superior correlation ($n = 53$) to a number of quality parameters such as protein content, mixogram water absorption and mix time, and bread characteristics (including loaf volume and crumb grain) when compared to results from AACC method 56-70 and the in house SDS sedimentation assay (Table 5).

The correlation coefficient (r) for wheat and flour protein content and results obtained from using the hybrid SDS-SRC method on flour and wheat samples ranged from 0.60 to 0.65 ($p = 0.0001$). The correlation coefficient (r) for wheat and flour protein content and the AACC 56-70 sedimentation method value on wheat meal ranged from 0.53 to 0.57 ($p = 0.001$).

Correlation of results obtained from AACC method 56-70 using flour showed no significance to both wheat and flour protein content, whereas the r value for wheat and flour protein content and the in-house HWWQL sedimentation method value on flour are 0.42 and 0.43 ($p = 0.01$), respectively. However, results from wheat meal analysis using the in-house assay produced r values of 0.61 and 0.63 for wheat and flour protein content, respectively, and were found to be significant ($p = 0.0001$). This data is similar to results reported by Khatkar et al. (1996) and Colombo et al. (2008) regarding SDS sedimentation and protein contents.

Test results obtained from the hybrid SDS-SRC method exhibited a significant correlation ($p = 0.0001$) to mixogram water absorption values for all the process conditions evaluated. Finney and Shogren (1972) reported that the water absorption and mixograph data provided a quantitative guide

Table 5. Correlation coefficients between some wheat quality parameters and sedimentation results of different processing and analytical conditions (n = 53).

Quality parameter	Hybrid SDS-SRC† sedimentation method: 4 min shaking and 2 min centrifuge	2 min shaking and 2 min centrifuge	4 min shaking and 2 min centrifuge	2 min shaking and 2 min centrifuge	AACC‡ 56-70 (AACC, 2000)		In-house HWWQL§	
	Wheat flour	Wheat flour	Wheat meal	Wheat meal	Wheat flour	Wheat meal	Wheat flour	Wheat meal
Wheat characteristics								
Wheat protein content, g kg^{-1}	0.66***	0.65***	0.59***	0.60***	–0.02 NS¶	0.57***	0.42**	0.61***
Flour protein content, g kg^{-1}	0.67***	0.65***	0.61***	0.63***	0.03 NS	0.53***	0.43**	0.63***
Mixograph characteristics								
Water absorption, g kg^{-1}	0.70***	0.65***	0.59***	0.61***	–0.01 NS	0.55***	0.45***	0.66***
Mix time, min	0.40**	0.36**	0.28*	0.22*	0.11 NS	0.16 NS	0.29*	0.25*
Mix tolerance	0.19 NS	0.19 NS	0.1 NS	0.05 NS	0.17 NS	0.01 NS	0.26*	0.08 NS
AACC method 56-70								
SDS sedimentation (wheat flour), mL	0.49***	0.56***	0.64***	0.65***	1	1	0.49***	0.44**
SDS sedimentation (wheat meal), mL	0.81***	0.84***	0.87***	0.88***	0.61***	0.61***	0.74***	0.78***
In-house HWWQL								
SDS sedimentation (wheat flour), mL	0.82***	0.80***	0.75***	0.74***	0.49***	0.74***	1	0.87***
SDS sedimentation (wheat meal), mL	0.90***	0.88***	0.87***	0.85***	0.44**	0.78***	0.87***	1
Bread characteristics								
Loaf volume, cm^3	0.85***	0.86***	0.87***	0.85***	0.42**	0.73***	0.64***	0.79***
Crumb grain score	0.65***	0.68***	0.67***	0.66***	0.39**	0.68***	0.64***	0.60***

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
*** Significant at the 0.001 probability level.
† SDS, sodium dodecyl sulfate; SRC, solvent retention capacity.
‡ AACC, American Association of Cereal Chemists.
§ HWWQL, Hard Winter Wheat Quality Laboratory, Manhattan, KS.
¶ NS, r value is not significant at p = 0.05.

for the level of water required to optimize dough for breadmaking. Mixing time was significantly correlated to hybrid SDS-SRC method values using wheat flour (p = 0.01). However, results obtained when using wheat meal resulted in lower correlations (p = 0.05). Marginal correlations to mix time were observed when using results from the in-house assay, whereas AACC method 56-70 test values showed no correlation to mixing time when using wheat flour or wheat meal. Oelofse et al. (2010) reported a correlation of 0.37 between results derived from AACC method 56-70 and mixograph mix time of 11 South African wheat lines grown over eight locations. None of the three methods produced test data that showed a significant correlation with mixing tolerance. Mixing tolerance is an indication of how rapidly dough loses consistency (breakdown) on over-mixing (Ohm and Chung, 1999).

Correlation Between Bread Quality Parameters (Loaf Volume and Crumb Grain) and Test Value

Comparisons were made among the correlation coefficients (n = 53) of loaf volume with results from the three sedimentation methods (Table 5). All of the correlation coefficients comparing loaf volume to results from the hybrid SDS-SRC method were at least 0.85, whereas the AACC method 56-70 and in-house methods produced results that exhibited lower r values with considerable variation between results obtained using flour and wheat meal. However, wheat meal had higher correlations.

Figures 1A and 1B depict the relationships (n = 53) between loaf volume and wheat flour protein (r = 0.66) and wheat meal protein (r = 0.64), which has historically been used as an indicator of end-use quality in bread wheats.

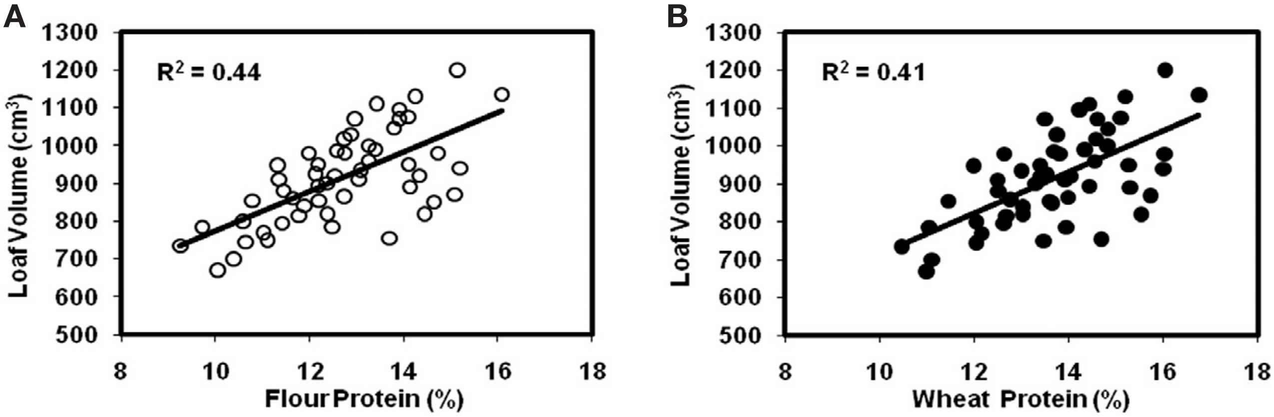

Figure 1. Relationship between wheat flour (A) and wheat meal (B) protein content vs. loaf volume (n = 53).

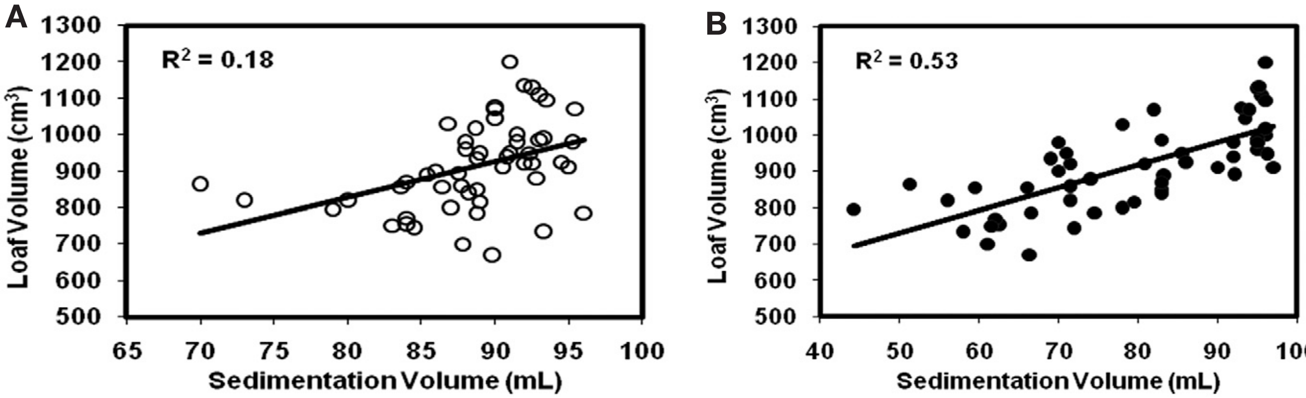

Figure 2. Wheat bread loaf volume as a function of sedimentation volume from wheat flour (A) and wheat meal (B) using American Association of Cereal Chemists method 56-70 (AACC, 2000) (n = 53).

The figures confirm a significant correlation between loaf volume and protein content in the 53 samples used in this study (Finney, 1943; Finney and Barmore, 1948). Similar results were reported by Colombo et al. (2008).

The correlation between loaf volume and results obtained using AACC method 56-70 for wheat flour and wheat meal is depicted in Fig. 2A and 2B. Similar to the results of our preliminary test, the sedimentation value showed a significant correlation with loaf volume for wheat meal (r = 0.73) that was higher than the correlation of loaf volume to wheat meal protein content but was lower for wheat flour (r = 0.42) and considerably lower than the correlation of loaf volume to wheat flour protein content.

The correlations between loaf volume and results obtained using the in-house HWWQL sedimentation method for wheat flour and wheat meal are shown in Fig. 3A and 3B. Wheat meal resulted in more consistent results with stronger correlations than results obtained from wheat flour. Figure 3A reveals that as the sedimentation volume increased to a threshold point, the correlation to loaf volume decreased as observed in Fig. 2A. This may indicate that the sedimentation method is limited in use as a predictor of bread loaf volume. Wheat flour correlations exhibited a ceiling effect, which implies that the samples with stronger and higher protein contents conferred sedimentation volumes beyond the test's sensitivity for satisfactory discrimination (Preston et al., 1982; Morris et al., 2007). Ayoub et al. (1993) reported a similar phenomenon with eastern Canadian bread wheat that had high protein content. Moreover, sedimentation results from AACC method 56-62 (AACC, 2000) also displayed a ceiling effect from wheat with high protein content. Xiao et al. (2006) reported an R^2 of 0.52 for sedimentation value versus loaf volume when using the in-house HWWQL sedimentation method on 116 HWW flour samples. The ceiling effect was minor in the wheat meal samples (Fig. 2B and 3B).

The correlation between loaf volume and results from the hybrid SDS-SRC method with 2 min shaking and 2 min centrifugation for wheat flour and wheat meal are shown in Fig. 4A and 4B. There is a strong linear correlation between loaf volume and the hybrid SDS-SRC method value for both wheat meal and wheat flour. When comparing results from AACC method 56-70 and the hybrid SDS-SRC method, R^2 increased from 0.18 to 0.75 for wheat flour and 0.53 to 0.73 for wheat meal. Comparison of results from the in-house HWWQL sedimentation method and the hybrid SDS-SRC method show that R^2 increased from 0.40 to 0.75 for wheat flour and 0.64 to

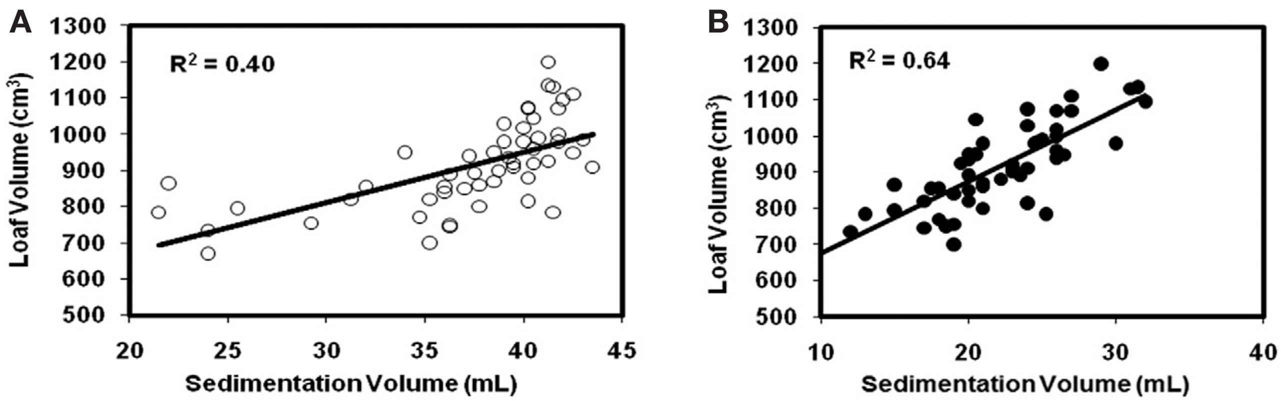

Figure 3. Wheat bread loaf volume as a function of sedimentation volume from wheat flour (A) and wheat meal (B) using the in-house modified American Association of Cereal Chemists method 56-70 (AACC, 2000) (n = 53).

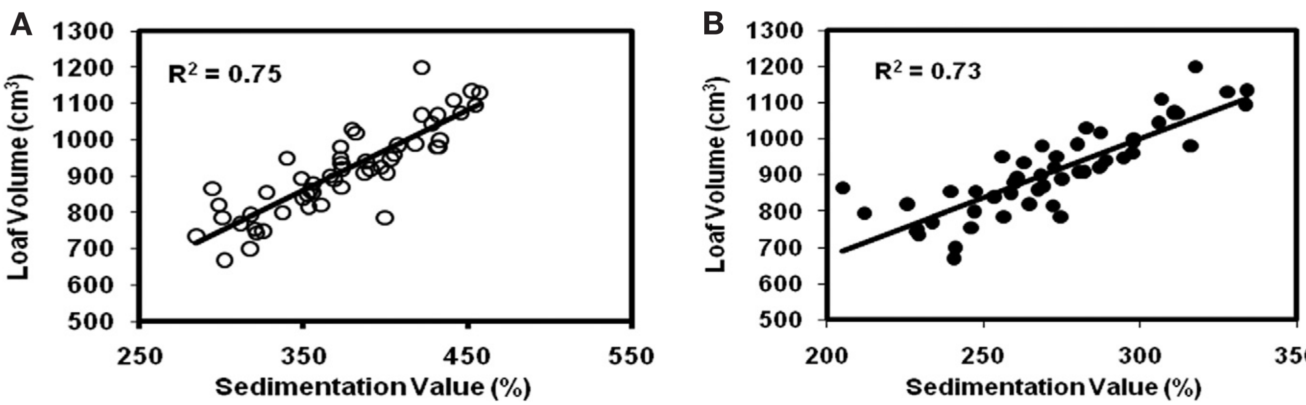

Figure 4. Relationship between new sodium dodecyl sulfate sedimentation value of wheat flour (A) or wheat meal (B) using 2 min shaking and 2 min centrifuge flour protein vs. loaf volume (n = 53).

0.73 for wheat meal. No ceiling effect was observed with regard to protein content on either wheat flour and wheat meal analyzed by the hybrid method. The results using a 4 min shaking and 2 min centrifugation similarly show a strong linear correlation between loaf volume and test data from the hybrid SDS-SRC method for wheat flour and wheat meal (Table 5). In addition, the hybrid SDS-SRC method exhibited correlations between loaf volume and sedimentation values for both wheat meal and flour that were higher than using those between loaf volume and wheat and/or flour protein contents.

The correlation coefficients between sedimentation methods and bread crumb score were determined for wheat meal and flour at 2 min shake time and 2 min centrifugation time. The correlation coefficients ranged from r = 0.66 to 0.68 for flour and wheat (Table 5). The 4 min shake time and 2 min centrifugation time produced results similar to the processing conditions reported above. The in-house method provided values that showed good correlation in both wheat meal and flour (r = 0.60 and 0.64). In contrast, results obtained using AACC method 56-70 showed equally strong correlations to crumb grain score in wheat meal samples (r = 0.68) but not in flour (r = 0.39). Bread crumb grain scores were only marginally correlated with flour protein content (r = 0.39; p = 0.05; data not shown). Crumb grain scores usually do not show high correlation with other quality parameters as compared to loaf volume (Park et al., 2004). Previous reports suggest that bread crumb grain score is an important parameter used to describe the appearance of sliced bread (Kamman, 1970; Hayman et al., 1998; Park et al., 2004; Xiao et al., 2006).

CONCLUSIONS

The hybrid SDS-SRC method showed similar performance on wheat meal compared to AACC method 56-70 and the in-house HWWQL methods for predicting bread quality parameters. The major advancement of the hybrid SDS-SRC method over the other two methods evaluated was the 60% reduction in analysis time. Furthermore, the hybrid SDS-SRC method may be more convenient for breeders for use on early generation lines since whole meal and milled flour are typically available in only limited quantities. Moreover, test results obtained from the hybrid SDS-SRC method showed improved correlation with loaf volume when using flour compared to results derived from AACC method 56-70 and the in-house HWWQL method. An additional advantage of the hybrid SDS-SRC method is that the method is not limited simply to wheat meal and provides similar values regardless of sample type (wheat meal or flour). This study suggests that either a 2 or 4 min shaking with 2 min centrifugation may achieve optimum results for both flour and wheat meal samples. For more rapid screening tests on both wheat meal and flour samples, 2 min shaking followed by 2 min centrifugation may be used but with a minor loss in accuracy compared to the 4 min shake time. The more rapid 2 min screening method could also be used in other settings, such as grain elevators, where a quick utilitarian determination of end-use quality might be useful.

While results from this study suggest that this modified method has broad application, further study is needed. Future research will focus on a larger test population that includes greater variation in environmental conditions (and stresses) as well as greater variation in quality factors such as test weight, protein quantity and quality, and loaf volume.

References

American Association of Cereal Chemists International (AACC). 2000. Approved methods of the American Association of Cereal Chemists, 10th ed. AACC International, St. Paul, MN.

Axford, D.W.E., E.F. McDermott, and D.G. Redman. 1978. Small-scale tests of breadmaking quality. Mill. Feed Fert. 16l:18.

Axford, D.W.E., E.F. McDermott, and D.G. Redman. 1979. Note on the sodium dodecyl sulfate test of breadmaking quality: Comparison with Pelshenke and Zeleny tests. Cereal Chem. 56:582–584.

Ayoub, M., J. Fregeau-Reid, and D.L. Smith. 1993. Evaluation of the SDS-sedimentation test for the assessment of eastern Canadian bread wheat quality. Can. J. Plant Sci. 73:995–999. doi:10.4141/cjps93-130

Colombo, A., G.T. Pérez, P.D. Ribotta, and A.E. León. 2008. A comparative study of physiochemical test for quality prediction of Argentine wheat flours used as corrector flours and for cookie production. J. Cereal Sci. 48: 775–780. doi:10.1016/j.jcs.2008.05.003

Dick, J.W., and J.S. Quick. 1983. A modified screening test for rapid estimation of gluten strength in early-generation durum wheat breeding lines. Cereal Chem. 60:315–318.

Finney, K.F. 1943. Fractionating and reconstituting techniques as tools in wheat flour research. Cereal Chem. 20:381–396.

Finney, K.F., and M.A. Barmore. 1948. Loaf volume and protein content of hard red spring wheats. Cereal Chem. 25:283–390.

Finney, K.F., and M.D. Shogren. 1972. A ten-gram mixograph for determining and predicting functional properties of wheat flour. Bakers. Dig. 46(2):23.

Gaines, C.S. 2000. Collaborative study of methods for solvent retention capacity profile –Method 56-11. Cereal Foods World 45:303–306.

Guttieri, M.J., C. Becker, and E.F. Souza. 2004. Application of wheat meal solvent retention capacity tests within soft wheat breeding populations. Cereal Chem. 81:261–266. doi:10.1094/CCHEM.2004.81.2.261

Hayman, D., R.C. Hoseney, and J.M. Faubion. 1998. Bread crumb grain development during baking. Cereal Chem. 75:577–580. doi:10.1094/CCHEM.1998.75.5.577

Kamman, P.W. 1970. Factors affecting the grain and texture of white bread. Baker's Dig. 44(2):34–38.

Khatkar, B.S., A.E. Bell, and J.D. Schofield. 1996. A comparative study of the inter-relationships between mixograph parameters and break-making qualities of wheat flour and glutens. J. Sci. Food Agric. 72:71–85. doi:10.1002/(SICI)1097-0010(199609)72:13.0.CO;2-4

Morris, C.F., B. Paszcznska, A.D. Bettge, and G.E. King. 2007. A critical examination of the sodium dodecyl sulfate (SDS) sedimentation test for wheat meals. J. Sci. Food Agric. 87:607–615. doi:10.1002/jsfa.2740

Oelofse, R.M. 2008. The relationship of sodium dodecyl sulfate sedimentation test values to bread making quality of early generation dryland wheat. Ph.D. diss. University of the Free State, Bloemfontein, South Africa.

Oelofse, R.M., M.T. Labuschagne, and C.S. van Deventer. 2010. Influencing factors of sodium dodecyl sulfate sedimentation in bread wheat. J. Cereal Sci. 52:96–99. doi:10.1016/j.jcs.2010.03.010

Ohm, J.B., and O.K. Chung. 1999. Gluten, pastry, and mixograph parameters of hard winter wheat flour in relation to bread making. Cereal Chem. 76:606–613. doi:10.1094/CCHEM.1999.76.5.606

Park, S.H., J.D. Wilson, O.K. Chung, and P.A. Seib. 2004. Size distribution and properties of wheat starch granules in relation to crumb grain score of pup-loaf bread. Cereal Chem. 81:699–704. doi:10.1094/CCHEM.2004.81.6.699

Preston, K.R., P.R. March, and K.H. Tipples. 1982. An assessment of the SDS-sedimentation test for the prediction of Canadian bread wheat quality. Can. J. Plant Sci. 62:545–555. doi:10.4141/cjps82-083

Sapirstein, H.D., and J. Suchy. 1999. SDS-protein gel test for prediction of bread loaf volume. Cereal Chem. 76:164–172. doi:10.1094/CCHEM.1999.76.1.164

SAS Institute. 1999. The SAS system for Windows. Release 8.0. SAS Inst., Cary, NC.

Wang, C., and M.I.P. Kovacs. 2002. Swelling index of gluten test. I. Method and comparison with sedimentation, gel-protein, and insoluble glutenin tests. Cereal Chem. 79:183–189. doi:10.1094/CCHEM.2002.79.2.183

Xiao, Z.S., S.H. Park, O.K. Chung, M.S. Caley, and P.A. Seib. 2006. Solvent retention capacity values in relation to hard winter wheat and flour properties and straight-dough breadmaking quality. Cereal Chem. 83:465–471. doi:10.1094/CC-83-0465

Zeleny, L. 1947. A simple sedimentation test for estimating the bread-baking and gluten qualities of wheat flour. Cereal Chem. 24:465–475.