Predicting Wheat Quality Characteristics and Functionality Using Near-Infrared Spectroscopy

F. E. Dowell[1,2] E. B. Maghirang,[1] F. Xie,[3] G. L. Lookhart,[3] R. O. Pierce,[4] B. W. Seabourn,[5] S. R. Bean,[5] J. D. Wilson,[5] and O. K. Chung[5]

Cereal Chem. 83(5):529–536

ABSTRACT

The accuracy of using near-infrared spectroscopy (NIRS) for predicting 186 grain, milling, flour, dough, and breadmaking quality parameters of 100 hard red winter (HRW) and 98 hard red spring (HRS) wheat and flour samples was evaluated. NIRS shows the potential for predicting protein content, moisture content, and flour color b^* values with accuracies suitable for process control ($R^2 > 0.97$). Many other parameters were predicted with accuracies suitable for rough screening including test weight, average single kernel diameter and moisture content, SDS sedimentation volume, color a^* values, total gluten content, mixograph, farinograph, and alveograph parameters, loaf volume, specific loaf volume, baking water absorption and mix time, gliadin and glutenin content, flour particle size, and the percentage of dark hard and vitreous kernels. Similar results were seen when analyzing data from either HRW or HRS wheat, and when predicting quality using spectra from either grain or flour. However, many attributes were correlated to protein content and this relationship influenced classification accuracies. When the influence of protein content was removed from the analyses, the only factors that could be predicted by NIRS with $R^2 > 0.70$ were moisture content, test weight, flour color, free lipids, flour particle size, and the percentage of dark hard and vitreous kernels. Thus, NIRS can be used to predict many grain quality and functionality traits, but mainly because of the high correlations of these traits to protein content.

Quality characteristics of wheat (*Triticum aestivum* L.) whole grain, flour, dough, and bread can be measured by various qualitative and quantitative tests. These measurements are typically used to determine value or used to predict functionality and end use quality. There are standard or recommended measurement methods for many of these quality parameters such as those found in the Approved Methods of AACC International (2000) and the United States Department of Agriculture (USDA) Grain Inspection Handbook (USDA 2004). These methods are generally difficult and time-consuming, and most cannot be used to rapidly measure quality characteristics and functionality.

Near-infrared spectroscopy (NIRS) has been used as a rapid, accurate, and nondestructive technique for measuring many wheat quality parameters. Williams et al (1988) used NIRS to predict wheat strength from hard red spring (HRS) flour spectra with good accuracies. Their samples were selected to represent a wide range in dough strength, sedimentation volume, and loaf volume. Rubenthaler and Pomeranz (1987) showed good correlations of water absorption, mix time, and loaf volume of hard red winter (HRW) wheat to flour NIR spectra. Delwiche et al (1998) applied NIRS models of flour from pure HRW cultivars to predict glutenin and gliadin content, SDS sedimentation volume, and mixograph peak resistance. When using commercial HRW and HRS flour, Delwiche and Weaver (1994) predicted absorption, mix time, bake score, loaf height, and mix tolerance from NIR spectra. Pawlinsky and Williams (1998) further showed that, when scanning Canadian HRW and HRS wheat grain, they could predict functionality parameters for the identification of suitable material for advancement in breeding programs. Their tests were limited to predicting protein content, wet gluten content, Zeleny sedimentation volume, mixing time, and farinograph parameters. In a study using spectra from whole grain and flour, Millar (2003) developed NIRS calibrations from U.K. and French wheat and showed potential for predicting protein and moisture content, water absorption, and flour color using NIR spectra, but had poor results when attempting to predict loaf volume and crumb grain score. Sissons et al (2006) used NIR spectra from grain from durum (*Triticum turgidum* L.) breeding lines to predict kernel, flour, and dough characteristics for breeding programs. Their results showed potential for grouping samples into low, medium, and high categories for test weight, thousand kernel weight, semolina yield, semolina yellow color, semolina browning, grain hardness, and cooked pasta firmness.

Hruskova and Famera (2003) used flour NIR spectra for quantitative screening based on moisture and protein content, ash, and wet gluten content. However, related research showed that farinograph (Hruskova et al 2001) and alveograph (Hruskova and Smejda 2003) parameters were predicted poorly when using NIR spectra from flour. Devaux et al (1986) used NIRS models to assign French soft wheat samples into three breadbaking quality categories (good, unsuitable, and irregular), but actual quality measurements or predictions were not made.

Thus, although other researchers have examined the potential for NIRS to predict various quality parameters from flour and whole grain spectra, most were limited to small sample sets, pure cultivars, or predicting a few specific parameters. No previous research has attempted to predict multiple whole kernel, milling, flour, dough, and breadmaking quality from whole kernels and flour from samples representing those in commercial trade. Thus, the objective of this research was to evaluate the potential of NIRS to measure whole kernel, milling, flour, dough, and breadmaking quality characteristics from whole kernels and flour of HRW and HRS wheat samples selected to represent the quality range expected in U.S. commercial wheat. It is not the goal of this research to develop calibrations but to examine where NIRS may provide the grain industry with a potential rapid means to predict grain, flour, dough, and bread quality, and where to focus future calibration efforts.

[1] USDA ARS, Grain Marketing and Production Research Center, Engineering Research Unit, 1515 College Avenue, Manhattan, KS 66502. Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that may also be suitable.

[2] Corresponding author. Phone: 785-776-2753. Fax: 785-537-5550. E-mail: floyd.dowell@gmprc.ksu.edu

[3] Kansas State University, Department of Grain Science and Industry, Manhattan, KS 66506.

[4] USDA, Grain Inspection, Packers, and Stockyards Administration, Federal Grain Inspection Service, Kansas City, MO 64163.

[5] USDA ARS, Grain Marketing and Production Research Center, Grain Quality and Structure Research Unit, 1515 College Avenue, Manhattan, KS 66502.

DOI: 10.1094/CC-83-0529

MATERIALS AND METHODS

Wheat Samples

One hundred HRW and 100 HRS wheat samples (1 kg each) from the 2002 and 2003 crop years were provided by the USDA Grain Inspection, Packers, and Stockyards Administration (GIPSA), Federal Grain Inspection Service (FGIS), Kansas City, MO. Samples were selected primarily based on their protein content and were expected to result in a wide range of bread quality.

Two HRS wheat samples were discarded from the sample set due to insect infestation. The HRW protein content range was 9.2–15.8% (average 12.6%), and the HRS protein content range was 11.4–19.3% (average 14.6%). Maghirang et al (2006) reported the details on all quality factors for these samples, including their source.

NIR Instrumentation

Four NIR instruments were selected to determine whether different wavelength ranges or scanning technology would affect prediction accuracy. NIR instruments used to collect spectral data of whole grain and flour samples were the Cognis-QTA Bruker Optics FT-NIR (835–2,502 nm, Cognis, Cincinnati, OH), which measured NIR absorbance using an interferometer; the Foss Infratec 1241 (850–1,050 nm, Foss NIRSystems, Eden Prairie, MN), which has the narrowest wavelength range but is probably the most common NIR instrument used in the grain industry; the Foss NIRSystems 6500 (450–2,498 nm), which had the widest wavelength range and included the visible spectrum; and the Perten Diode Array (DA) 7200 (950–1,650 nm, Perten Instruments, Springfield, IL). For the Foss 6500, the full-rectangular scanning module was used for scanning grain, whereas the quarter-rectangular module was used for scanning flour. The Foss 1241 measured NIR absorbance using diffuse transmittance, whereas the other spectrometers used diffuse reflectance. All spectral data were collected at the USDA-ARS Grain Marketing and Production Research Center (GMPRC), Manhattan, KS, except for Foss Infratec 1241 whole grain spectra that were collected by GIPSA. Grain samples used for spectral data collection were passed through the Boerner sample divider (Seedburo Equipment Company, Chicago, IL), with the number of passes being dependent on the sample size requirement of the NIR instrument. This was done to ensure that representative subsamples from the original sample were being used.

For all spectrometers, data were collected according to the instrument manufacturers' recommendations. For the Cognis-QTA, 200 spectra were collected from a subsample with ≈150 g of grain or 80 g of flour and it was continuously stirred while the sample container rotated about a fixed paddle. The spectra were averaged into one spectrum. This was repeated for a second subsample and the spectra from the two subsamples were averaged. For the Foss 6500, 64 spectra were collected from a subsample with ≈ 250 g of grain or 60 g of flour as the sample cell moved past the detection optics, and the spectra were automatically averaged into one spectrum. This was repeated for a second subsample and the spectra from the two subsamples were averaged. For the Perten 7200, 100 spectra were collected as a sample with ≈260 g of grain or 160 g of flour rotating in the sample cup. The spectra were automatically averaged into one spectrum. This was repeated four times on the same subsample, and the four resulting spectra were averaged. For the Foss Infratec whole grain analysis, one spectrum was collected from each of 10 subsamples from 600 g of sample as the sample automatically fed through the spectrometer. The 10 spectra were then automatically averaged. For the flour samples analyzed on the Foss Infratec, a subsample of ≈10 g was placed in a sample cell and five spectra were collected and automatically averaged. This was repeated for a second flour subsample and the two spectra were averaged.

Wheat Quality Analysis

Maghirang et al (2006) described the testing methods. Whole grain quality characteristics included test weight, protein content, moisture content, single kernel hardness, average single kernel moisture content, and mean kernel diameter. In addition, the percentage of dark hard and vitreous kernels were measured on the HRS wheat samples.

Flour was milled on a Brabender Quadrumat Sr. mill using the procedures described by Maghirang et al (2006). Milling and flour quality indicators measured include flour yield, wheat and flour ash contents, flour protein content, brightness (L^*), amount of red and green color (a^*), amount of yellow and blue color (b^*), flour geometric mean diameter, starch geometric mean diameter, polyphenol oxidase content, falling number, SDS sedimentation volume, total gluten content, gluten index, insoluble and soluble glutenin protein contents, gliadin protein content, total glutenin protein content, free lipids, polar lipids, and nonpolar lipids.

Dough properties were evaluated using the mixograph, farinograph, and alveograph. Parameters measured by the mixograph were water absorption, mix time, and mixing tolerance. The parameters measured by the farinograph were water absorption, development time, stability, tolerance, and quality number. The parameters measured by the alveograph were peak height, length, swelling index, work, and configuration ratio. The breadmaking quality parameters measured for the pup loaf (100 g of flour) straight-dough procedures were baking water absorption, baking mix time, crumb grain score, loaf volume, specific loaf volume, and loaf volume potential.

All whole grain quality characteristics were analyzed by GIPSA. CII Laboratory Services, Kansas City, MO, conducted alveograph tests. The GMPRC conducted all other tests. Standard methods were used whenever an approved method was available and are described by Maghirang et al (2006).

Data Analyses

Forty-six HRW and 47 HRS whole grain, milling, flour, dough, and breadmaking quality characteristics were analyzed from both whole grain and flour spectra, resulting in a total of 186 predictions. Spectral data were analyzed using GRAMS/AI software (v. 7, Thermo Galactic, Salem, NH). Models were developed for each quality parameter by partial least squares (PLS) regression. The number of factors when the F-ratio probability level was ≈0.75 was used for the calibration model. A one-sample-out cross-validation was used for the PLS analysis.

Since many measured parameters are correlated to protein content, the PLS regressions were performed with and without the influence of protein content. To remove the influence of protein content on the regressions, each selected attribute was regressed on protein content and the residuals calculated. These residuals were then used in the PLS regressions. This was done only for the Foss 6500 data.

Mean-centered absorbance (Log 1/R) and the Savitzky-Golay first-derivative of the absorbance spectrum were analyzed. Other pretreatments were not tested. Small improvements in prediction accuracies can be achieved with some spectral pretreatments (Delwiche and Reeves 2004), but generally pretreatments will result in similar classification accuracies but with fewer factors. Statistics used to determine the ability of NIR to predict specific parameters were the coefficient of determination (R^2) and standard error of cross validation (SECV). When reporting results, only parameters with $R^2 > 0.70$ were considered important. This value was selected because the ratio of the standard deviation of the reference data to the SECV, which is similar to the RPD in Williams (2001), is ≈2 when $R^2 = 0.70$. A smaller R^2 indicates predictions using PLS will not be much better than predictions using the mean value of the reference data alone. Similarly, higher R^2 values indicate that NIR spectroscopy can predict quality parameters with more accuracy. Williams (2001) indicated that $R^2 = 0.70$–0.90 is

suitable for rough screening, R^2 = 0.90–0.97 is suitable for screening or quality control, R^2 = 0.97–0.99 is suitable for process control, and larger values are suitable for most applications.

RESULTS

Comparison of NIR Spectrometers

When comparing R^2 and SECV values for the four spectrometers for each of the HRS and HRW grain and flour quality predictions, the Foss 6500 had the highest R^2 for 68 of the quality predictions (Tables I–IV). The Foss 1241 had the highest R^2 for 46 of the quality predictions, followed by the Perten 7200 for 22 of the quality predictions, and the Cognis QTA for 14 of the quality predictions. There were 36 quality factors predicted with the same accuracy by two or more spectrometers.

There were no whole grain, flour, dough, or baking characteristics that were consistently predicted more accurately by a specific spectrometer, with the exception of flour color. The Foss 6500 had R^2 values for color a^* and b^* that were almost twice as high as the other spectrometers (≈0.96 when predicting flour color from flour and ≈0.70 when predicting flour color from whole grain), which can be expected because it was the only spectrometer that had a visible wavelength sensor in addition to an NIR sensor. The Cognis FT-NIR with its interferometer and ability to measure absorbance over very narrow wavebands did not show any advantages in predicting quality factors when compared with the other instruments. This is probably because the quality factors absorb over broad regions in the NIR, therefore negating any benefit of using the interferometer. The Foss 1241 with the narrowest wavelength range had higher R^2 values for more quality predictions when compared with the Cognis and Perten instruments. Because absorption overtones extend throughout the NIR region, perhaps the simplicity of this instrument offsets the noise introduced when using sensors that extend further into the NIR. A more complete comparison and description of these spectrometers and their advantages and limitations is reported by Armstrong et al (2006).

TABLE I
Summary of Partial Least Squares Regression Statistics for Predicting Grain, Flour and Baking Quality Characteristics from Spectra from Four NIR Spectrometers (Hard Red Winter Wheat Whole Kernels)

	Cognis FT-NIR		Foss 1241		Foss 6500				Perten 7200	
Quality Parameter	R^2	**SECV**	R^2	**SECV**	R^2	**SECV**	$R^{2\,c}$	**SECV**[c]	R^2	**SECV**
Test weight, lb/bu	0.69(7)	0.66	0.69(6)	0.66	**0.74(6)**	0.56	0.60(6)	0.60	**0.72(10)**	0.62
Grain protein content, 14% mb	**0.98(10)**	0.25	**0.99(9)**	0.18	**0.97(12)**	0.29	–	–	**0.98(9)**	0.25
Grain moisture content, %	**0.95(7)**	0.19	**0.96(8)**	0.19	**0.96(8)**	0.18	**0.95(8)**	0.20	**0.97(10)**	0.16
Single kernel hardness index	0.38(10)	3.86	0.47(10)	3.51	0.46(10)	3.54	0.38(10)	3.65	0.39(8)	3.79
Single kernel moisture, %	**0.93(7)**	0.26	**0.93(6)**	0.26	**0.92(10)**	0.26	**0.92(8)**	0.27	**0.94(5)**	0.22
Single kernel dia, avg mm	0.68(6)	0.07	0.55(11)	0.08	**0.71(8)**	0.07	0.45(11)	0.07	**0.71(6)**	0.06
Flour protein content, 14% mb	**0.92(9)**	0.45	**0.97(7)**	0.29	**0.97(12)**	0.30	–	–	**0.97(10)**	0.31
Falling number, sec	0.13(1)	105.5	0.21(9)	91.44	0.30(8)	86.16	0.22(9)	89.30	0.04(5)	100.9
Flour yield, extraction, %	0.37(6)	1.01	0.36(11)	1.00	0.34(6)	1.04	0.28(6)	1.03	0.39(5)	1.00
Wheat ash, 14% mb	0.43(8)	0.07	0.41(8)	0.07	0.43(8)	0.07	0.31(10)	0.07	0.33(5)	0.08
Flour ash, 14% mb	0.31(9)	0.03	0.27(8)	0.03	0.26(10)	0.03	0.24(10)	0.03	0.13(4)	0.03
Polyphenol oxi, au/min/mL	0.45(7)	0.06	0.41(7)	0.06	0.51(6)	0.06	0.50(8)	0.05	0.41(7)	0.06
SDS sedimentation vol, mL	**0.70(8)**	2.36	**0.70(11)**	2.36	**0.72(9)**	2.31	0.25(10)	2.20	0.68(7)	2.47
Brightness (color L^*)	0.35(5)	0.39	0.42(9)	0.37	0.21(2)	0.43	0.28(10)	0.22	0.33(5)	0.40
Red/green (color a^*)	0.50(8)	0.16	0.45(8)	0.17	0.69(9)	0.13	0.59(12)	0.12	0.56(10)	0.15
Yellow/blue (color b^*)	0.30(8)	0.57	0.32(12)	0.57	0.66(13)	0.39	0.66(13)	0.39	0.36(10)	0.55
Gluten content, g/10 g of flour	**0.92(10)**	0.16	**0.93(9)**	0.14	**0.88(10)**	0.19	0.26(6)	0.14	**0.92(10)**	0.15
Average gluten index, %	0.27(4)	2.83	0.38(12)	2.65	0.51(12)	2.05	0.17(9)	2.62	0.40(9)	2.59
Flour size GMD, 50% vol, µm	0.11(4)	2.32	0.25(10)	2.17	0.31(10)	2.06	0.31(10)	2.06	0.13(4)	2.30
Starch size GMD, 50% vol, µm	0.14(5)	1.33	0.07(8)	1.63	0.16(3)	1.63	0.11(4)	1.57	0.09(5)	1.38
Soluble glutenins, mg	**0.75(9)**	0.42	**0.77(8)**	0.40	**0.75(10)**	0.42	0.19(7)	0.39	0.61(5)	0.51
Soluble gliadins, mg	**0.85(9)**	0.76	**0.89(8)**	0.64	**0.86(10)**	1.72	0.17(5)	0.65	**0.88(10)**	0.65
Insoluble glutenins, mg	**0.85(9)**	0.64	**0.85(7)**	0.65	**0.84(10)**	0.66	0.09(9)	0.62	**0.84(10)**	0.67
Total glutenins, mg	**0.89(8)**	0.76	**0.93(7)**	0.59	**0.92(9)**	0.64	0.12(6)	0.56	**0.81(6)**	1.02
Free lipids, %	0.08(2)	4.67	0.46(12)	3.61	0.08(2)	4.68	0.03(6)	4.70	0.001(1)	4.90
Polar lipids, %	0.14(5)	3.25	0.15(5)	3.16	0.18(4)	3.11	0.04(5)	3.02	0.07(4)	4.46
Nonpolar lipids, %	0.05(2)	4.46	0.21(11)	3.94	0.05(1)	4.51	0.05(1)	4.51	0.001(1)	5.54
Mixograph absorption, %	**0.92(7)**	0.67	**0.92(7)**	0.69	**0.90(9)**	0.75	0.15(4)	0.64	**0.91(7)**	0.74
Mixo time, min	0.40(8)	0.53	0.44(9)	0.50	0.60(9)	0.43	0.42(10)	0.42	0.47(9)	0.49
Mixo tolerance score (0–6)	0.30(8)	0.78	0.26(12)	0.80	0.41(9)	0.71	0.42(4)	0.70	0.25(6)	0.79
Farinograph absorption, %	0.65(9)	1.35	0.65(12)	1.35	**0.76(13)**	1.12	0.51(14)	1.10	0.66(10)	1.32
Farino development time, min	0.32(8)	3.79	0.33(9)	3.73	0.23(6)	4.01	0.04(3)	3.75	0.36(9)	3.64
Farino stability, min	0.16(5)	3.76	0.30(14)	3.50	0.06(7)	4.03	0.04(3)	4.05	0.15(10)	3.92
Farino mixing tolerance, min	0.21(5)	10.12	0.24(10)	10.04	0.14(7)	10.71	0.02(4)	10.62	0.19(9)	10.31
Farino quality number	0.36(8)	47.39	0.34(9)	47.56	0.27(7)	50.18	0.13(2)	47.50	0.27(7)	49.19
Alveograph peak (P), mm	0.05(7)	15.20	0.20(11)	13.81	0.26(10)	13.34	0.22(10)	13.17	0.23(10)	13.74
Alveo length (L), mm	0.65(8)	18.47	0.69(7)	17.44	0.69(10)	17.45	0.10(10)	16.88	**0.70(9)**	17.33
Alveo swelling index, mL	0.64(7)	2.12	0.69(7)	1.94	**0.72(10)**	1.88	0.08(8)	1.86	**0.73(10)**	1.83
Alveo work, 10^{-4} J	0.66(7)	48.60	0.69(9)	43.63	**0.70(10)**	45.72	0.09(10)	44.28	0.69(8)	46.02
Alveo config ratio (P/L)	0.48(8)	0.42	0.47(7)	0.41	0.52(10)	0.40	0.05(7)	0.40	0.69(10)	0.32
Baking water absorption, %	0.48(9)	1.31	0.48(7)	1.28	0.37(6)	1.42	0.05(11)	1.25	0.43(8)	1.35
Baking mix time, min	0.37(8)	0.68	0.35(13)	0.70	0.41(9)	0.66	0.34(12)	0.63	0.38(9)	0.68
Crumb grain score (0–6)	0.27(8)	0.58	0.14(6)	0.61	0.20(6)	0.59	0.08(16)	0.56	0.23(8)	0.58
Loaf volume, cm^3	**0.78(7)**	39.27	**0.81(7)**	36.36	**0.82(9)**	36.11	0.05(5)	32.83	**0.82(10)**	35.85
Loaf specific vol, cm^3/g	**0.80(9)**	0.25	**0.80(9)**	0.25	**0.85(11)**	0.22	0.05(9)	0.23	**0.80(10)**	0.24
Loaf vol potential, cm^3/% protein	0.35(8)	3.98	0.27(7)	4.20	0.22(3)	4.30	0.07(4)	4.08	0.25(5)	4.26

[a] R^2 values ≥ 0.70 shown in bold font.
[b] Numbers in parentheses after the R^2 values refer to the number of factors of the PLS model.
[c] After protein covariate removal.

When spectra were preprocessed using a Savitzky-Golay first-derivative, no advantage in R^2 or SECV values over using the absorbance spectra was seen in the ability to predict any quality parameters (data not shown).

Predictions Using HRW Whole Kernel and Flour Spectra

When analyzing spectra from whole kernels, one or more spectrometers predicted HRW grain protein content, flour protein content, and grain moisture content with $R^2 \geq 0.97$ (Table I). Average single kernel moisture content, total gluten content, total glutenin content, and mixograph absorption were predicted with $R^2 \geq 0.90$. Test weight, average single kernel diameter, SDS sedimentation volume, soluble and insoluble glutenin content, soluble gliadin content, farinograph absorption, alveograph length, alveograph swelling index, alveograph work, loaf volume, and specific loaf volume were predicted with R^2 values of ≈0.70–0.90.

When analyzing flour spectra, one or more spectrometers predicted HRW grain and flour protein content, and color *b** with $R^2 \geq 0.97$ (Table II). Color *a**, total gluten content, total glutenin content, and mixograph absorption were predicted with $R^2 \geq 0.90$. The following attributes were predicted with R^2 of 0.70–0.90: SDS sedimentation volume, soluble and insoluble glutenin content, soluble gliadin content, alveograph length, alveograph swelling index, alveograph work, loaf volume, and specific loaf volume.

Predictions Using HRS Whole Kernel and Flour Spectra

Results similar to HRW wheat were seen when predicting quality traits of HRS wheat. When analyzing spectra from whole kernels, one or more spectrometers predicted grain and flour protein content, and grain moisture content with $R^2 \geq 0.97$ (Table III). Total gluten content was predicted with $R^2 \geq 0.90$. The attributes predicted with R^2 of ≈0.70–0.90 were test weight, average single kernel moisture content, dark hard and vitreous kernels, average single kernel diameter, color *a** and *b**, insoluble and total glutenin content, mixograph absorption, farinograph quality number, baking water absorption, loaf volume, and specific loaf volume.

TABLE II
Summary of Partial Least Squares Regression Statistics for Predicting Grain, Flour and Baking Quality Characteristics from Spectra from Four NIR Spectrometers (Hard Red Winter Wheat Flour)

	Cognis FT-NIR		Foss 1241		Foss 6500				Perten 7200	
Quality Parameter	R^2	SECV	R^2	SECV	R^2	SECV	R^2 [c]	SECV[c]	R^2	SECV
Test weight, lb/bu	0.38(8)	0.94	0.43(6)	0.88	0.55(6)	0.79	0.34(5)	0.77	0.47(6)	0.86
Grain protein content, 14% mb	**0.95(4)**	0.39	**0.98(5)**	0.25	**0.98(5)**	0.25	–	–	**0.98(6)**	0.30
Grain moisture content, %	0.38(8)	0.73	0.30(12)	0.78	0.47(9)	0.67	0.46(9)	0.67	0.29(11)	0.81
Single kernel hardness index	0.29(8)	4.15	0.14(5)	4.45	0.47(8)	3.49	0.39(8)	3.59	0.30(3)	4.00
Single kernel moisture, %	0.45(8)	0.70	0.38(12)	0.76	0.53(8)	0.65	0.52(8)	0.66	0.32(12)	0.82
Single kernel dia, avg mm	0.43(8)	0.09	0.37(6)	0.10	0.54(11)	0.08	0.27(11)	0.08	0.40(6)	0.09
Flour protein content, 14% mb	**0.96(7)**	0.35	**0.99(7)**	0.21	**0.98(6)**	0.22	–	–	**0.99(8)**	0.20
Falling number, sec	0.08(10)	105.7	0.08(8)	100.0	0.14(6)	95.83	0.10(6)	96.11	0.01(3)	111.7
Flour yield, extraction, %	0.27(10)	1.14	0.40(11)	1.00	0.36(6)	1.02	0.32(6)	1.01	0.24(10)	1.18
Wheat ash, 14% mb	0.36(9)	0.08	0.18(7)	0.09	0.24(7)	0.08	0.10(9)	0.09	0.31(8)	0.08
Flour ash, 14% mb	0.56(10)	0.02	0.14(12)	0.03	0.34(10)	0.03	0.29(10)	0.03	0.35(8)	0.02
Polyphenol oxi, au/min/mL	0.41(10)	0.06	0.24(10)	0.07	0.33(6)	0.07	0.21(3)	0.07	0.32(8)	0.07
SDS sedimentation vol, mL	0.67(4)	2.49	**0.70(7)**	2.39	0.69(3)	2.42	0.11(4)	2.37	**0.70(4)**	2.36
Brightness (color *L**)	0.31(7)	0.41	0.40(6)	0.37	0.32(4)	0.40	0.27(10)	0.20	0.37(10)	0.40
Red/green (color *a**)	0.41(9)	0.18	0.47(11)	0.17	**0.96(7)**	0.05	**0.94(10)**	0.05	0.53(12)	0.16
Yellow/blue (color *b**)	0.31(9)	0.58	0.51(12)	0.48	**0.97(7)**	0.11	**0.97(7)**	0.11	0.48(12)	0.49
Gluten content, g/10 g of flour	**0.91(6)**	0.16	**0.95(10)**	0.12	**0.93(5)**	0.14	0.14(3)	0.15	**0.94(9)**	0.13
Average gluten index, %	0.33(3)	2.69	0.42(10)	2.53	0.50(3)	2.02	0.36(5)	2.22	0.40(7)	2.55
Flour size GMD, 50% vol, µm	0.58(8)	1.59	0.40(7)	1.90	0.60(2)	1.55	0.58(2)	1.55	0.51(5)	1.71
Starch size GMD, 50% vol, µm	0.11(2)	1.34	0.05(3)	1.39	0.09(1)	1.36	0.03(6)	1.68	0.11(1)	1.34
Soluble glutenins, mg	**0.75(4)**	0.41	**0.78(8)**	0.39	**0.78(8)**	0.39	0.17(9)	0.40	**0.79(3)**	0.38
Soluble gliadins, mg	**0.84(6)**	0.77	**0.89(7)**	0.64	**0.86(5)**	0.71	0.16(9)	0.67	**0.88(7)**	0.67
Insoluble glutenins, mg	**0.82(5)**	0.71	**0.83(7)**	0.68	**0.86(4)**	0.63	0.13(7)	0.60	**0.86(4)**	0.63
Total glutenins, mg	**0.91(4)**	0.68	**0.93(6)**	0.62	**0.96(5)**	0.48	0.15(8)	0.55	**0.95(5)**	0.53
Free lipids, %	0.21(9)	4.57	0.48(11)	3.57	0.61(13)	3.07	0.58(13)	3.04	0.28(8)	4.26
Polar lipids, %	0.14(4)	3.27	0.23(6)	3.00	0.16(4)	3.14	0.04(2)	3.20	0.14(3)	3.18
Nonpolar lipids, %	0.16(5)	4.97	0.22(11)	3.93	0.33(13)	3.72	0.33(13)	3.72	0.13(10)	4.36
Mixograph absorption, %	**0.92(3)**	0.68	**0.95(7)**	0.55	**0.93(5)**	0.63	0.03(3)	0.72	**0.92(4)**	0.67
Mixo time, min	0.35(10)	0.56	0.36(10)	0.54	0.54(5)	0.45	0.43(6)	0.42	0.35(3)	0.54
Mixo tolerance score (0–6)	0.26(9)	0.83	0.28(14)	0.80	0.45(5)	0.67	0.46(5)	0.67	0.22(3)	0.81
Farinograph absorption, %	0.63(11)	1.40	0.69(12)	1.26	0.63(6)	1.37	0.32(11)	1.31	0.67(12)	1.32
Farino development time, min	0.29(3)	3.81	0.38(5)	3.56	0.29(2)	3.81	0.14(2)	3.83	0.37(3)	3.60
Farino stability, min	0.13(9)	4.15	0.06(5)	3.99	0.06(2)	3.97	0.03(2)	3.97	0.15(8)	3.90
Farino mixing tolerance, min	0.11(9)	11.74	0.17(7)	10.39	0.06(2)	11.05	0.04(2)	10.98	0.13(8)	11.11
Farino quality number	0.35(5)	46.85	0.37(5)	46.02	0.31(2)	48.37	0.07(2)	48.37	0.39(9)	46.49
Alveograph peak (*P*), mm	0.04(3)	15.06	0.11(8)	14.54	0.24(10)	13.72	0.06(2)	14.19	0.07(3)	14.66
Alveo length (*L*), mm	**0.72(7)**	16.55	**0.71(7)**	16.83	**0.71(3)**	16.91	0.09(2)	16.39	**0.71(3)**	16.50
Alveo swelling index, mL	**0.71(6)**	1.88	**0.72(8)**	1.86	**0.73(4)**	1.84	0.08(2)	1.81	**0.73(3)**	1.82
Alveo work, 10^{-4} J	**0.70(5)**	45.69	**0.70(7)**	45.63	**0.75(3)**	41.68	0.05(4)	44.26	**0.70(4)**	45.54
Alveo config ratio (*P/L*)	0.56(9)	0.38	0.50(8)	0.40	0.58(7)	0.32	0.16(7)	0.37	0.50(5)	0.40
Baking water absorption,%	0.51(4)	1.24	0.52(5)	1.23	0.53(4)	1.22	0.01(2)	1.30	0.54(8)	1.21
Baking mix time, min	0.20(5)	0.77	0.38(10)	0.68	0.40(3)	0.66	0.23(3)	0.68	0.38(10)	0.69
Crumb grain score (0–6)	0.19(3)	0.59	0.32(10)	0.55	0.34(2)	0.54	0.12(3)	0.54	0.21(5)	0.59
Loaf volume, cm^3	**0.78(6)**	39.85	**0.87(6)**	30.25	**0.83(5)**	35.16	0.02(3)	35.91	**0.83(4)**	34.66
Loaf specific vol, cm^3/g	**0.77(7)**	0.27	**0.84(6)**	0.22	**0.80(5)**	0.25	0.01(3)	0.25	**0.80(4)**	0.25
Loaf vol potential, cm^3/% protein	0.40(9)	3.87	0.45(10)	3.64	0.33(7)	4.03	0.01(2)	4.22	0.41(8)	3.79

[a] R^2 values ≥ 0.70 shown in bold font.
[b] Numbers in parentheses after the R^2 values refer to the number of factors of the PLS model.
[c] After protein covariate removal.

When analyzing spectra from flour, one or more spectrometers predicted HRS grain and flour protein content, and color b^* with $R^2 \geq 0.97$ (Table IV). Color a^* and total gluten content were predicted with $R^2 \geq 0.90$. The attributes predicted with R^2 of ≈0.70–0.90 were test weight, flour particle size, insoluble glutenin content, total glutenin content, free lipid content, mixograph and farinograph water absorption, baking water absorption, loaf volume, and specific loaf volume.

Influence of Protein Content on Predictions

While NIR spectroscopy shows potential to predict various HRW and HRS quality attributes, many of these attributes are correlated to protein content. Of those parameters predicted with $R^2 \geq 0.70$ for either HRW or HRS wheat, total gluten content, insoluble glutenin content (HRW), total glutenin content, soluble gliadin content (HRW), mixograph absorption, loaf volume, and specific loaf volume were correlated to protein content with $r >$ 0.90. SDS sedimentation volume, soluble glutenin content (HRW), insoluble glutenin content (HRS), alveograph length, alveograph swelling index, and alveograph work (HRW) were correlated to protein content with $r > 0.80$. Test weight, average single kernel diameter, baking water absorption, alveograph work (HRS), and farinograph absorption were correlated to protein content with $r >$ 0.70. The remaining parameters of moisture content, color a^* and b^*, flour particle size, lipid content, and dark hard and vitreous kernels showed no significant correlation to protein content.

When the influence of protein content was removed from the HRW grain and flour analyses, the only constituents predicted with $R^2 \geq 0.70$ were moisture content predicted from grain spectra, and color a^* and b^* values from flour (Tables I and II). Most constituents had R^2 values reduced to <0.20 when the influence of protein content was removed.

When the influence of protein content was removed from the HRS grain and flour analyses, the only constituents predicted

TABLE III
Summary of Partial Least Squares Regression Statistics for Predicting Grain, Flour and Baking Quality Characteristics from Spectra from Four NIR Spectrometers (Hard Red Spring Wheat Whole Kernels)

	Cognis FT-NIR		Foss 1241		Foss 6500				Perten 7200	
Quality Parameter	R^2	SECV	R^2	SECV	R^2	SECV	$R^{2\,c}$	SECV[c]	R^2	SECV
Test weight, lb/bu	**0.85**(8)	0.65	0.62(9)	1.05	**0.89**(12)	0.56	**0.73**(12)	0.63	**0.84**(10)	0.68
Grain protein content, 14% mb	**0.98**(10)	0.22	**0.99**(9)	0.20	**0.97**(12)	0.29	–	–	**0.98**(10)	0.23
Grain moisture content, %	**0.89**(7)	0.36	**0.98**(7)	0.17	**0.93**(6)	0.28	**0.92**(6)	0.29	**0.94**(8)	0.25
Dark hard vitreous, %	**0.79**(8)	7.78	**0.77**(12)	8.18	**0.84**(8)	6.85	**0.79**(6)	7.48	**0.79**(7)	7.89
Single kernel hardness index	0.50(11)	3.42	0.41(12)	3.76	0.57(12)	3.18	0.52(12)	3.37	0.49(11)	3.50
Single kernel moisture, %	**0.81**(6)	0.45	**0.81**(10)	0.45	**0.84**(6)	0.41	**0.82**(6)	0.42	**0.84**(5)	0.41
Single kernel dia, avg mm	0.68(8)	0.08	0.68(11)	0.08	**0.70**(10)	0.08	0.45(10)	0.08	**0.70**(7)	0.08
Flour protein content, 14% mb	**0.97**(10)	0.26	**0.98(8)**	0.24	**0.97(12)**	0.26	–	–	**0.98(9)**	0.24
Falling number, sec	0.40(8)	63.65	0.43(7)	61.49	0.43(9)	61.72	0.39(7)	62.00	0.37(7)	64.76
Flour yield, extraction, %	0.42(10)	1.62	0.63(12)	1.25	0.54(12)	1.42	0.35(12)	1.42	0.53(10)	1.41
Wheat ash, 14% mb	0.47(9)	0.08	0.37(7)	0.09	0.47(7)	0.08	0.37(7)	0.08	0.45(9)	0.08
Flour ash, 14% mb	0.43(7)	0.03	0.34(13)	0.03	0.38(8)	0.03	0.29(7)	0.03	0.37(11)	0.03
Polyphenol oxi, au/min/mL	0.44(5)	0.06	0.51(13)	0.06	0.46(10)	0.07	0.32(5)	0.07	0.52(11)	0.06
SDS sedimentation vol, mL	0.02(6)	4.63	0.01(3)	4.67	0.05(6)	4.58	0.02(6)	4.56	0.01(3)	4.71
Brightness (color L^*)	0.28(10)	0.37	0.11(6)	0.40	0.35(11)	0.35	0.31(8)	0.23	0.27(9)	0.37
Red/green (color a^*)	0.63(8)	0.14	0.63(12)	0.14	**0.70(11)**	0.12	0.62(11)	0.11	0.60(9)	0.14
Yellow/blue (color b^*)	0.58(6)	0.45	0.50(12)	0.42	**0.73(12)**	0.36	**0.72(12)**	0.36	0.67(11)	0.41
Gluten content, g/10 g of flour	**0.86(8)**	0.16	**0.90(7)**	0.14	**0.87(10)**	0.15	0.39(3)	0.13	**0.90(8)**	0.13
Average gluten index, %	0.15(2)	3.66	0.17(2)	3.61	0.13(3)	3.72	0.11(4)	3.66	0.17(4)	3.63
Flour size GMD, 50% vol, μm	0.49(12)	1.78	0.45(10)	1.84	0.60(12)	1.57	0.43(9)	1.87	0.46(9)	1.82
Starch size GMD, 50% vol, μm	0.31(5)	1.43	0.30(7)	1.44	0.35(11)	1.42	0.08(2)	1.38	0.44(11)	1.31
Soluble glutenins, mg	0.13(6)	0.70	0.13(6)	0.70	0.11(5)	0.70	0.08(2)	0.67	0.16(6)	0.68
Soluble gliadins, mg	0.33(8)	2.04	0.36(6)	1.97	0.20(6)	2.23	0.01(5)	2.01	0.21(5)	2.19
Insoluble glutenins, mg	**0.78(8)**	0.85	**0.76(10)**	0.89	**0.76(10)**	0.90	0.07(3)	0.77	0.38(7)	1.45
Total glutenins, mg	**0.81(8)**	0.88	**0.82(7)**	0.86	**0.76(9)**	1.00	0.01(7)	0.80	0.61(9)	1.29
Free lipids, %	0.40(4)	4.89	0.48(10)	4.53	0.50(9)	4.47	0.47(8)	4.39	0.66(10)	3.68
Polar lipids, %	0.27(8)	3.75	0.32(3)	3.55	0.30(5)	3.59	0.27(8)	3.55	0.45(11)	3.25
Nonpolar lipids, %	0.36(2)	3.94	0.36(7)	3.97	0.39(7)	3.88	0.37(7)	3.90	0.49(10)	3.56
Mixograph absorption, %	**0.86(8)**	0.79	**0.86(7)**	0.80	**0.85(11)**	0.83	0.06(5)	0.78	**0.89(9)**	0.69
Mixo time, min	0.63(8)	0.59	0.62(12)	0.61	0.66(8)	0.52	0.43(4)	0.60	0.61(7)	0.61
Mixo tolerance score (0–6)	0.51(8)	0.78	0.54(8)	0.77	0.58(6)	0.72	0.48(5)	0.72	0.52(8)	0.77
Farinograph absorption, %	0.66(11)	1.37	0.66(10)	1.35	0.66(10)	1.35	0.32(9)	1.39	0.69(10)	1.34
Farino development time, min	0.607(8)	3.98	0.60(12)	4.38	0.65(8)	4.07	0.35(7)	3.93	0.60(9)	4.39
Farino stability, min	0.35(8)	3.54	0.50(12)	3.10	0.48(8)	3.16	0.43(8)	3.01	0.46(4)	3.19
Farino mixing tolerance, min	0.21(9)	8.65	0.14(6)	8.87	0.29(8)	8.12	0.19(8)	8.04	0.27(10)	8.40
Farino quality number	0.66(8)	41.35	0.67(12)	41.61	**0.73(10)**	37.33	0.50(8)	35.7	0.68(9)	40.30
Alveograph peak (P), mm	0.03(3)	17.22	0.17(11)	16.30	0.39(10)	13.84	0.25(8)	15.21	0.21(12)	16.25
Alveo length (L), mm	0.42(10)	19.80	0.40(10)	19.13	0.51(10)	17.85	0.21(8)	18.25	0.43(10)	19.37
Alveo swelling index, mL	0.41(10)	2.00	0.41(10)	1.97	0.52(10)	1.79	0.21(8)	1.82	0.36(7)	2.04
Alveo work, 10^{-4} J	0.44(7)	79.64	0.44(6)	79.04	0.46(6)	77.37	0.05(2)	74.66	0.46(7)	77.21
Alveo config ratio (P/L)	0.27(10)	0.29	0.26(10)	0.29	0.48(10)	0.24	0.28(11)	0.26	0.34(11)	0.27
Baking water absorption,%	**0.70(8)**	0.95	0.67(9)	0.99	0.68(10)	0.97	0.32(2)	1.04	**0.70(10)**	0.96
Baking mix time, min	0.56(7)	0.87	0.50(12)	0.89	0.59(7)	0.78	0.44(7)	0.83	0.57(6)	0.86
Crumb grain score (0–6)	0.31(9)	0.55	0.21(3)	0.57	0.17(2)	0.59	0.15(2)	0.59	0.19(4)	0.58
Loaf volume, cm^3	**0.79(8)**	40.56	**0.79(8)**	40.58	**0.77(9)**	42.82	0.01(4)	37.68	**0.79(7)**	40.60
Loaf specific vol, cm^3/g	**0.76(8)**	0.28	**0.77(11)**	0.28	**0.73(9)**	0.30	0.01(4)	0.26	**0.78(8)**	0.27
Loaf vol potential, cm^3/% protein	0.11(4)	3.46	0.18(6)	3.32	0.06(6)	3.65	0.01(4)	3.41	0.14(5)	3.40

[a] R^2 values ≥ 0.70 shown in bold font.
[b] Numbers in parentheses after the R^2 values refer to the number of factors of the PLS model.
[c] After protein covariate removal.

with $R^2 \geq 0.70$ were test weight, moisture content, dark hard and vitreous kernels, and color b^* predicted from grain spectra, and color L^*, a^*, and b^* values, flour particle size, and free lipids predicted from flour spectra (Tables III and IV). As with HRW results, most constituents R^2 values were reduced to <0.20, which shows the strong influence of protein content on the ability of NIRS to predict many quality parameters.

Comparison of Predictions from Whole Kernels or Flour

Grain protein content was predicted from flour spectra with good accuracy ($R^2 = 0.98$), but that is because grain and flour protein content are highly correlated ($r = 0.99$). Except for HRS test weight, which could be predicted from flour spectra with $R^2 = 0.74$, no other grain characteristics could be predicted from flour spectra. There may be occasions when a miller or baker may wish to determine whole kernel characteristics from good or bad performing flour so similar grain lots can be obtained, or avoided. However, these results show it is difficult to determine grain characteristics from flour. As expected, most flour, dough, and baking parameters were predicted more accurately, or with similar accuracy, when using the flour spectra than when using whole kernel spectra. Exceptions were that some farinograph and mixograph parameters had slightly greater R^2 values when predicted from whole kernel spectra than from flour spectra.

DISCUSSION

Measuring protein content in grain and flour has been a successful application of NIRS because it has very strong and broad absorption bands throughout the NIR region (Williams 2001) and is a major wheat component. This is not the case for many other biochemical components that affect other grain quality attributes because they may be present in very small quantities and additionally highly correlated to protein content, as shown in this

TABLE IV
Summary of Partial Least Squares Regression Statistics for Predicting Grain, Flour and Baking Quality Characteristics from Spectra from Four NIR Spectrometers (Hard Red Spring Wheat Flour)

	Cognis FT-NIR		Foss 1241		Foss 6500				Perten 7200	
Quality Parameters	R^2	SECV	R^2	SECV	R^2	SECV	R^2 [c]	SECV[c]	R^2	SECV
Test weight, lb/bu	0.67(9)	0.97	0.66(11)	0.99	**0.74(8)**	0.86	0.40(8)	0.89	0.66(7)	0.99
Grain protein content, 14% mb	**0.98(7)**	0.24	**0.99(10)**	0.19	**0.98(7)**	0.23	–	–	**0.98(9)**	0.26
Grain moisture content, %	0.40(10)	0.85	0.43(12)	0.82	0.40(9)	0.83	0.39(8)	0.84	0.32(8)	0.90
Dark hard vitreous, %	0.34(8)	13.97	0.45(5)	12.67	0.45(5)	12.67	0.39(5)	12.70	0.37(6)	13.58
Single kernel hardness index	0.42(10)	3.68	0.31(12)	4.08	0.40(4)	3.69	0.39(4)	3.71	0.19(3)	4.29
Single kernel moisture, %	0.43(10)	0.80	0.35(11)	0.85	0.34(9)	0.85	0.29(10)	0.86	0.38(8)	0.83
Single kernel dia, avg mm	0.51(3)	0.10	0.58(12)	0.09	0.55(4)	0.09	0.23(3)	0.09	0.56(7)	0.09
Flour protein content, 14% mb	**0.99(3)**	0.15	**0.99(7)**	0.16	**0.99(4)**	0.16	–	–	**0.99(7)**	0.16
Falling number, sec	0.23(6)	71.64	0.37(4)	64.71	0.43(6)	61.39	0.39(6)	62.01	0.37(6)	64.94
Flour yield, extraction, %	0.53(7)	1.42	0.58(12)	1.34	0.63(9)	1.24	0.52(9)	1.22	0.56(8)	1.37
Wheat ash, 14% mb	0.32(10)	0.09	0.35(12)	0.09	0.32(12)	0.09	0.11(11)	0.10	0.20(8)	0.10
Flour ash, 14% mb	0.46(7)	0.03	0.47(10)	0.03	0.41(8)	0.03	0.38(8)	0.03	0.40(8)	0.03
Polyphenol oxi, au/min/mL	0.27(8)	0.08	0.31(10)	0.07	0.26(9)	0.08	0.15(9)	0.08	0.25(10)	0.08
SDS sedimentation vol, mL	0.30(6)	2.36	0.05(4)	4.54	0.04(3)	4.57	0.01(2)	4.51	0.06(4)	4.56
Brightness (color L^*)	0.40(9)	0.32	0.40(11)	0.33	0.49(7)	0.30	**0.75(5)**	0.14	0.43(6)	0.32
Red/green (color a^*)	0.61(8)	0.14	0.60(12)	0.14	**0.96(6)**	0.04	**0.90(6)**	0.06	0.57(8)	0.15
Yellow/blue (color b^*)	0.49(10)	0.50	0.38(12)	0.56	**0.97(6)**	0.12	**0.97(6)**	0.13	0.35(8)	0.59
Gluten content, g/10 g of flour	**0.91(9)**	0.13	**0.91(8)**	0.13	**0.90(5)**	0.13	0.45(6)	0.13	0.91(7)	0.13
Average gluten index, %	0.15(8)	3.78	0.16(12)	3.83	0.17(2)	3.63	0.17(3)	3.59	0.16(5)	3.67
Flour size GMD, 50% vol, mm	**0.70(3)**	1.33	**0.72(8)**	1.31	**0.78(11)**	1.16	**0.77(11)**	1.17	0.72(6)	1.31
Starch size GMD, 50% vol, mm	0.38(7)	1.26	0.24(4)	1.49	0.36(6)	1.38	0.24(9)	1.29	0.26(4)	1.48
Soluble glutenins, mg	0.07(3)	0.73	0.04(4)	0.73	0.09(4)	0.71	0.09(7)	0.68	0.10(3)	0.70
Soluble gliadins, mg	0.34(5)	1.99	0.33(5)	2.01	0.30(3)	2.06	0.02(3)	2.08	0.29(3)	2.06
Insoluble glutenins, mg	**0.81(10)**	0.79	**0.78(6)**	0.81	**0.86(7)**	0.68	0.02(3)	0.81	**0.80(3)**	0.82
Total glutenins, mg	**0.82(7)**	0.85	**0.86(7)**	0.76	**0.88(7)**	0.71	0.05(3)	0.82	**0.86(5)**	0.75
Free lipids, %	0.52(10)	4.57	**0.74(12)**	3.24	**0.73(11)**	3.29	**0.71(11)**	3.27	0.60(9)	4.11
Polar lipids, %	0.35(7)	3.60	0.35(5)	3.51	0.35(6)	3.52	0.28(6)	3.47	0.52(8)	3.03
Nonpolar lipids, %	0.22(9)	5.24	0.44(11)	3.74	0.37(11)	4.04	0.37(11)	4.05	0.35(11)	4.16
Mixograph absorption, %	**0.88(3)**	0.74	**0.88(7)**	0.72	**0.88(4)**	0.73	0.01(2)	0.72	**0.87(5)**	0.78
Mixo time, min	0.53(8)	0.69	0.66(12)	0.58	0.67(7)	0.57	0.47(8)	0.58	0.64(4)	0.59
Mixo tolerance score (0–6)	0.65(10)	0.66	0.58(12)	0.72	0.55(7)	0.75	0.41(8)	0.76	0.58(5)	0.72
Farinograph absorption, %	0.64(7)	1.39	0.75(10)	1.15	0.58(7)	1.50	0.32(9)	1.39	0.62(6)	1.42
Farino development time, min	0.60(8)	4.36	0.59(12)	4.47	0.67(9)	3.95	0.28(7)	4.14	0.62(6)	4.23
Farino stability, min	0.44(9)	3.29	0.43(9)	3.26	0.38(6)	3.41	0.24(4)	3.45	0.43(5)	3.26
Farino mixing tolerance, min	0.22(5)	8.50	0.22(4)	8.38	0.14(3)	8.82	0.02(2)	8.72	0.19(7)	8.78
Farino quality number	0.66(9)	42.21	0.58(7)	45.97	0.65(8)	42.41	0.36(11)	41.17	0.68(6)	40.37
Alveograph peak (P), mm	0.31(8)	14.77	0.41(10)	13.39	0.21(9)	15.83	0.22(9)	15.73	0.30(10)	15.30
Alveo length (L), mm	0.62(9)	15.66	0.66(11)	14.77	0.60(9)	16.02	0.37(9)	16.10	0.53(6)	17.27
Alveo swelling index, mL	0.60(7)	1.62	0.65(10)	1.51	0.62(9)	1.58	0.37(9)	1.61	0.55(6)	1.71
Alveo work, 10^{-4} J	0.49(2)	74.84	0.49(6)	75.02	0.52(3)	72.42	0.08(3)	72.50	0.50(8)	75.94
Alveo config. ratio (P/L)	0.50(8)	0.23	0.54(10)	0.22	0.48(9)	0.24	0.32(9)	0.24	0.50(10)	0.23
Baking water absorption,%	0.63(3)	1.05	**0.74(9)**	0.87	0.65(5)	1.03	0.06(3)	1.11	0.64(6)	1.04
Baking mix time, min	0.42(5)	1.01	0.55(13)	0.89	0.61(7)	0.82	0.44(8)	0.83	0.56(5)	0.87
Crumb grain score (0–6)	0.19(5)	0.59	0.19(2)	0.58	0.14(8)	0.61	0.16(8)	0.61	0.19(9)	0.62
Loaf volume, cm^3	**0.82(3)**	36.64	**0.83(7)**	36.59	**0.83(4)**	35.98	0.01(4)	37.47	**0.82(5)**	37.44
Loaf specific vol, cm^3/g	**0.80(4)**	0.25	**0.80(7)**	0.26	**0.80(4)**	0.26	0.02(4)	0.26	0.78(5)	0.27
Loaf vol potential, $cm^3 \cdot$% protein	0.16(3)	3.40	0.12(6)	3.44	0.17(4)	3.35	0.01(3)	3.27	0.14(7)	3.51

[a] R^2 values ≥ 0.70 shown in bold font.
[b] Numbers in parentheses after the R^2 values refer to the number of factors of the PLS model.
[c] After protein covariate removal.

study. Thus, it is difficult to measure these attributes using NIRS independent of their correlation to protein content. One exception is particle size analysis by NIRS because absorption increases with an increase in particle size (Approved Method 39-70A, near-infrared reflectance method for hardness determination in wheat) (AACC International 2000), and Tables I–IV show that particle size predictions are not influenced by the removal of protein content from prediction models.

Results obtained by Pawlinsky and Williams (1998) when scanning whole kernels to predict protein content, gluten content, and mixograph time were similar to those obtained in this study, but their R^2 values for farinograph parameters were higher than obtained in this study. Their better results may be because their samples were from pure cultivars grown at one location. Millar (2003) reported an R^2 value for protein content measured from whole grain that was similar to that reported here (R^2 = 0.99 vs. 0.97), but their water absorption and loaf volume R^2 values were much lower (0.68 vs. 0.90 for water absorption and 0.41 vs. 0.80 for loaf volume). When predicting these same parameters from flour, Millar (2003) reports protein content predictions that are similar to those achieved in this study (R^2 = 0.99), but farinograph water absorption predictions were higher (R^2 = 0.93 vs. 0.58–0.63), and loaf volume predictions were lower (R^2 = 0.62 vs. 0.83) than values reported here. Their samples had a much narrower range in quality when compared with the range of those used in this study, with the protein content and loaf volume ranges being only about half the range of samples in the study reported here.

Delwiche et al (1998) reported HRW R^2 values for predicting protein content that were similar to those reported here (0.99 vs. 0.97). The absolute amount of gliadins and glutenins is well correlated to protein content (Bean et al 1998; Delwiche et al 1998), and after the influence of protein content was removed from the predictions by Delwiche et al (1998), R^2 values, although poor, were similar or higher than those reported here for HRW wheat (0.53 vs. 0.17 for gliadins, 0.41 vs. 0.09 for glutenin content, 0.54 vs. 0.46 for mix time, and 0.45 vs. 0.42 for mix tolerance). Thus it may be possible to predict the quantity of gliadins and glutenins using NIRS only because of the correlation to protein content. The Delwiche et al (1998) sample set had a wider range of protein content (6.8–20.1% vs. 9.2–15.8%) and mixograph time (1.6–7.5 min vs. 2.5–6.1 min) than in this study. These statistics reported by Delwiche et al (1998) may be better than some of those reported here because their sample set consisted of pure cultivars grown in one region, whereas the commercial samples and blends used in this study were obtained from throughout the United States. These commercial samples include environmental influences and blends that may mask differences in protein quality. Protein quality measurements (quantity of gliadins and glutenins) by NIRS may be applicable to breeding programs as stated by Delwiche et al (1998) but may not be attainable on commercial samples.

Sissons et al (2006) showed the correlation of protein content to durum quality measurements but removed the influence of protein content only from the predictions of test weight and pasta firmness. The R^2 value for test weight of ≈0.90 was much higher than the best value of 0.73 obtained in our research. This may be because their samples consisted of pure breeding lines.

Delwiche and Weaver (1994) reported similar R^2 (≈0.65) for baking water absorption as achieved in our research but much lower values for mix time (R^2 = 0.25 vs. 0.61). Williams et al (1988) reported baking absorption, farinograph absorption, protein content, loaf volume, and alveograph work R^2 values similar to those obtained in this research. However, their farinograph stability R^2 values were much higher than ours (0.73 vs. 0.38). This may be due to their samples containing a much wider range in strength values because they were selected specifically based on strength parameters and included hard and soft wheat. Rubenthaler and Pomeranz (1987) predicted loaf volume of HRW wheat flours with similar accuracy as in this study, but their mix time and absorption R^2 values were much higher (0.72 vs. 0.40 for mix time and 0.81 vs. 0.53 for absorption). This may be due to the small number of unique samples analyzed by Rubenthaler and Pomeranz (1987). They analyzed 173 subsamples but these were mostly replicates from only 19 original samples.

Hruskova et al (2001) reported that no farinograph characteristics were predicted from NIR spectra with $R^2 < 0.40$, which they achieved with water absorption. The farinograph water absorption was predicted in our research with an R^2 of ≈0.70. Hruskova and Smejda (2003) did not achieve $R^2 \geq 0.25$ for any alveograph measurements, possibly due to their small sample sizes. In this study, we achieved HRW R^2 values of ≈0.70 for alveograph length, swelling index, and work. Hruskova and Famera (2003) reported Zeleny sedimentation R^2 of 0.11–0.50 when predicted from flour NIR spectra, whereas we achieved R^2 values of ≈0.20–0.70 in our studies.

This research did not attempt to predict starch damage using NIRS. However, Osborne et al (1982) used NIRS to predict starch damage as measured by the Farrand method with a SEP = 3.2. Morgan and Williams (1995) used NIRS to predict starch damage with SEP = 3 and R^2 = 0.92. However, Finney et al (1988) showed a high correlation (r = 0.89) between hardness and damaged starch, and this relationship was further reviewed by Pomeranz (1988). Thus, since damaged starch is not chemically different from undamaged starch, NIRS likely predicts starch damage because of correlations of starch damage to factors that have absorption bands in the NIR region.

The precision of the reference methods affects the potential for using NIRS to predict flour and grain attributes. The reference method precision for those characteristics predicted with $R^2 >$ 0.70 are discussed below. Williams (1975) reported that the moisture content reference method (Approved Method 44-15A, moisture-air oven method) (AACC International 2000) standard error was 0.069%. The SECV results reported here were 0.20–0.29% for predicting grain moisture content and 0.27–0.42% for predicting single kernel moisture content. These samples had equilibrated to ≈11% moisture content, with a standard deviation between samples of ≈1%. Thus, while the SECV was several times higher than the reference error, a lower NIR error may be achieved with a wider range of moisture content. Williams (1975) reported that the protein content reference method (Approved Method 46-10, crude protein improved Kjeldahl method) standard error was 0.098%. The SECV results reported here were 0.16–0.30%. Thus, protein content was predicted with an error of 2–3× that of the reference method. Oliver et al (1992) reported standard deviations for L^*, a^*, and b^* of 0.48, 0.12, and 0.78, respectively, when measuring 33 white flours. The reproducibility should be less than these values. The flour SECV values of 0.14–0.20, 0.05–0.06, and 0.11–0.13 for L^*, a^*, and b^*, respectively, indicate that the Foss 6500 is predicting color values with accuracies better than those reported previously.

This research showed that the free lipid reproducibility using two replicates per sample was ≈1.70% (data not shown), which agrees with repeatability reported by Hubbard et al (2004) of ≈2%. The NIR prediction SECV was 3.20% for HRS flour. Thus, the error for predicting free lipids using NIRS was about twice that of the reference method. All other lipid predictions were poor.

For flour particle size predictions, the reproducibility of our laser diffraction reference method was 0.336 μm (average standard deviation of two replicates from 99 samples) and CV = 0.4%. Hareland (1994) reported that the sieving reference method had a standard deviation = 7.2 μm and coefficient of variation (CV) of 8.9% for our approximate particle size range, and that the laser diffraction method had a standard deviation =3.8 μm and CV = 4.5%. They also used NIRS to predict flour particle size with a CV = 1.3% and SECV = 1.1 μm. These results agree with our

HRW CV = 1.3% and SECV = 1.17 μm. Thus, NIRS can be used to predict HRS flour particle size with accuracies similar to reference methods reported previously, but 3–4× higher than the reproducibility of the reference method used in this research.

HRS dark hard and vitreous kernel predictions had a SECV = 7.48%, which is about twice the standard deviation of 3.5 reported by Xie et al (2004) for detecting vitreous kernels in durum wheat. HRS test weight SECV was 0.63 lb/bu and was less than the standard error of 1.20 lb/bu for replicates reported by Troccoli and di Fonzo (1999). This lower NIRS error was unexpected and shows that the error in predicting test weight using NIRS is less than the error in measuring test weight in replicated samples. The HRW test weight error was similar in magnitude to the HRS error, but the HRW R^2 value was only 0.60.

While other researchers have reported the application of NIRS to predict various quality attributes, this is the first attempt to predict multiple whole grain, flour, dough, and bake quality attributes from whole grain, and then the same attributes from flour from the same samples. Most results agree with previous researchers. However, this is the first report of using NIRS to predict SDS sedimentation volume and alveograph parameters from whole grain.

NIRS shows the potential for using spectra from whole kernels for predicting protein content and bulk moisture content with accuracies suitable for process control ($R^2 > 0.97$). Test weight, average single kernel diameter and moisture content, SDS sedimentation volume, color a^* and b^* values, total gluten content, soluble gliadin content, soluble and insoluble glutenin content, total glutenin content, mixograph water absorption, farinograph water absorption, farinograph quality number, alveograph length, alveograph swelling index, alveograph work, loaf volume, specific loaf volume, baking water absorption, and dark hard and vitreous kernels had accuracies suitable for rough screening ($R^2 \geq 0.70$).

NIR spectra from flour can predict protein content and b^* value with accuracies suitable for process control. NIR spectra from flour can predict test weight, color a^*, total gluten content, soluble gliadin content, soluble and insoluble glutenin contents, total glutenin content, flour particle size, free lipid content, mixograph and farinograph water absorption, alveograph length, alveograph swelling index, alveograph work, baking water absorption, loaf volume, specific loaf volume, and SDS sedimentation volume with accuracies suitable for rough screening. However, when the influence of protein content on prediction models is removed, very few quality attributes could be predicted with accuracy, even for rough screening.

ACKNOWLEDGMENTS

We gratefully acknowledge the USDA, ARS, GMPRC Hard Winter Wheat Quality Laboratory for sample analysis and the USDA, GIPSA, FGIS Technical Center for sample analysis and partial funding for this research. We also thank Perten Instruments, Cognis, and Foss for loaning NIR instrumentation. We also thank Stephen Delwiche, agricultural engineer, USDA ARS Instrumentation and Sensing Laboratory, Beltsville, MD, and Mark West, area statistician, USDA ARS, Ft. Collins, Co., for comments on early versions of this manuscript.

LITERATURE CITED

AACC International. 2000. Approved Methods of the American Association of Cereal Chemists, 10th Ed. Methods 39-70A, 44-15A, and 46-10. The Association: St. Paul, MN.

Armstrong, P. R., Maghirang, E. B., Xie, F., and Dowell, F. E. 2006. Comparison of dispersive and Fourier-transform NIR instruments for measuring grain and flour attributes. Appl. Eng. Agric. 22:453-457.

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 proteins in flour. Cereal Chem. 75:374-379.

Delwiche, S. R., and Reeves, J. B., III. 2004. The effect of spectral pretreatments of the partial least squares modeling of agricultural products. J. Near Infrared Spectrosc. 12:177-182.

Delwiche, S. R., and Weaver, G. 1994. Bread quality of wheat flour by near-infrared spectrophotometry: Feasibility of modeling. J. Food Sci. 59:410-415.

Delwiche, S. R., Graybosch, R. A., and Peterson, C. J. 1998. Predicting protein composition, biochemical properties, and dough-handling properties of hard red winter wheat flour by near-infrared reflectance. Cereal Chem. 75:412-416.

Devaux, M. F., Bertrand, D., and Martin, G. 1986. Discrimination of bread-baking quality of wheats according to their variety by near-infrared reflectance spectroscopy. Cereal Chem. 63:151-154.

Finney, P. L., Kinney, J. E., and Donelson, J. R. 1988. Prediction of damaged starch in straight-grade flour by near-infrared reflectance analysis of whole ground wheat. Cereal Chem. 65:449-452.

Hareland, G. A. 1994. Evaluation of flour particle size distribution by laser diffraction, sieve analysis and near-infrared reflectance spectroscopy. J. Cereal Sci. 21:183-190.

Hruskova, M., and Famera, O. 2003. Prediction of wheat and flour Zeleny sedimentation value using NIR technique. Czech J. Food Sci. 21:91-96.

Hruskova, M., and Smejda, P. 2003. Wheat flour dough alveograph characteristics predicted by NIRSystems 6500. Czech J. Food Sci. 21:28-33.

Hruskova, M., Bednarova, M., and Novotny, F. 2001. Wheat flour dough rheological characterictics predicted by NIRSystems 6500. Czech J. Food Sci. 19:213-218.

Hubbard, J. D., Downing, J. M., Ram, M. S., and Chung, O. K. 2004. Lipid extraction from wheat flour using supercritical fluid extraction. Cereal Chem. 81:693-698.

Maghirang, E. B., Lookhart, G. L., Bean, S. R., Pierce, R. O., Xie, F., Caley, M. S. Wilson, J. D., Seabourn, B. W., Ram, M. S., Park, S. H., Chung, O. K., and Dowell, F. E. 2006. Comparison of quality characteristics and breadmaking functionality of hard red winter and hard red spring wheat. Cereal Chem. 83:520-528.

Millar, S. J. 2003. The development of near-infrared (NIR) spectroscopy calibrations for the prediction of wheat and flour quality. The Home-Grown Cereals Authority Project Report No. 310. HGCA: London.

Morgan, J. E., and Williams, P. C. 1995. Starch damage in wheat flours: A comparison of enzymatic, iodometric, and near-infrared reflectance techniques. Cereal Chem. 72:209-212.

Oliver, J. R., Blakeney, A. B., and Allen, H. M. 1992. Measurement of flour color in color space parameters. Cereal Chem. 69:546-551.

Osborne, B. G., Douglas, S., and Fearn, T. 1982. The application of near infrared reflectance analysis to rapid flour testing. J. Food Technol. 17:355-363.

Pawlinsky, T., and Williams, P. 1998. Prediction of wheat bread-baking functionality in whole kernels, using near infrared reflectance spectroscopy. J. Near Infrared Spectrosc. 6:121-127.

Pomeranz, Y. 1988. Composition and functionality of wheat flour components. Pages 219-370 in: Wheat Chemistry and Technology. Y. Pomeranz, ed. AACC International: St. Paul, MN.

Rubenthaler, G. L., and Pomeranz, Y. 1987. Near-infrared reflectance spectra of hard red winter wheats varying widely in protein content and breadmaking potential. Cereal Chem. 64:407-411.

Sissons, M., Osborne, B., and Sissons, S. 2006. Application of near infrared reflectance spectroscopy to a durum wheat breeding programme. J. Near Infrared Spectrosc. 14:17-25.

Troccoli, A., and di Fonzo, N. 1999. Relationship between kernel size features and test weight in *Triticum durum*. Cereal Chem. 76:45-49.

USDA. 2004. Grain Inspection Handbook, Book II. Washington, D.C.

Williams, P. C. 1975. Application of near infrared reflectance spectroscopy to analysis of cereal grains and oilseeds. Cereal Chem. 52:561-576.

Williams, P. C. 2001. Implementation of near-infrared reflectance technology. Pages 145-169 in: Near-infrared Technology in the Agricultural and Food Industries. P. Williams and K. Norris, eds. AACC International: St. Paul, MN.

Williams, P. C., El-Haramen, P. J., Ortis-Ferira, G., and Srivastava, J. P. 1988. Preliminary observations of the determination of wheat strength by near-infrared reflectance. Cereal Chem. 65:109-114.

Xie, F., Pearson, T., Dowell, F. E., and Zhang, N. 2004. Detecting vitreous wheat kernels using reflectance and transmittance image analysis. Cereal Chem. 81:594-597.

[Received January 25, 2006. Accepted June 26, 2006.]