Relationships of Quality Characteristics with Size-Exclusion HPLC Chromatogram of Protein Extract in Soft White Winter Wheats

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ABSTRACT

This study investigated relationships between molecular weight distributions of unreduced grain proteins and grain, flour, and end-use quality characteristics of soft white winter wheats grown in Oregon. Absorbance area and area percentage values of protein fractions separated by size-exclusion HPLC (SE-HPLC) showed significant correlations with quality characteristics, indicating associations of molecular weight distributions of proteins with quality characteristics. Specifically, high molecular weight polymeric protein fractions appeared to have a detrimental effect on soft wheat quality. This was shown by significant positive correlations with single kernel hardness index, and mixograph water absorption and tolerance, and negative correlations with break flour yield, cookie diameter, and cake volume. Higher proportions of soluble monomeric protein fraction eluted after the main gliadin peak, were associated with soft wheat quality due to negative associations with single kernel hardness index and mixograph water absorption and tolerance, and positive associations with break flour yield, cookie diameter, and cake volume. Calibration models were developed by the application of multivariate analyses to the SE-HPLC data. These models explained >90% of the variation in mixograph water absorption and cookie diameter and thickness.

Baking quality of soft wheat is generally evaluated by experimental sugar-snap cookie making. It is commonly assumed that flour that produces sugar-snap cookies with larger diameters and with a more obvious and uniform cracking pattern on the top surface is associated with increased tenderness and better quality in most soft wheat products, except for crackers (Wade 1988; Miller and Hoseney 1997).

One of the most important quality factors used to select wheat for cookie making is kernel hardness. Wheat kernel hardness affects break flour yield, flour particle size distribution, and damaged starch content of flour (Wade 1988). When compared with hard wheat, soft wheat cultivars, in general, produce a higher yield of break flour with a smaller particle size distribution, and lower protein and starch damage, all of which are usually desirable for cookie and cake baking (Rogers et al 1993; Labuschagne et al 1997; Faridi et al 2000). Wheat hardness is affected by variations in puroindolines and lipids associated with starch granule surfaces (Greenblatt et al 1995; Giroux and Morris 1997). However, puroindolines are not fully responsible for variation of kernel texture when the tested wheat cultivars are limited within a class (Campbell et al 1999; Giroux et al 2000; Morris et al 2004, 2005). Significant genetic variation in hardness has been attributed to factors other than puroindolines such as water solubles, lipids, and pentosans (Bettge and Morris 2000; Giroux et al 2000; Ohm and Chung 2002). Wheat hardness is also reported to have a significant association with viscosity of water extractable arabinoxylans (Bettge and Morris 2000; Igrejas et al 2002a).

Variation in proteins has been also considered as an important factor affecting wheat hardness and consequently cookie quality (Huebner and Gaines 1992; Ohm et al 1999, 2006; Giroux et al 2000). Although cookie dough mixing either does not or only minimally develops the gluten network, baking the dough leads to formation of a gluten network as dough temperature exceeds the glass transition point of the gluten proteins due to the increased unfolding, hydration, and aggregation of proteins (Doescher et al 1987; Gaines 1990). Gluten network formation during baking accompanies a decrease of water mobility and a drastic increase of dough viscosity that slows spread and eventually results in elastic shrinkage of the baking cookie dough, preventing the expansion of gas cells (Doescher et al 1987; Slade et al 1989; Miller and Hoseney 1997). Thus, desirable gluten properties for cookie making include weak network development with low elasticity/high extensibility, minimal oxidative gelation between proteins for low dough viscosity, and high glass transition temperature (Doescher et al 1987; Slade et al 1989; Pedersen et al 2004; Bettge and Morris 2007). These studies suggest that qualitative variations in flour protein affect cookie quality as well as the quantitative variations. Qualitative variations in the main gluten proteins such as high molecular weight glutenin subunits were reported to have significant associations with cookie quality among soft wheat cultivars (Souza et al 1994; Labuschagne and van Deventer 1995; Hou et al 1996a,b; Igrejas et al 2002a,b). Huebner et al (1999) also reported a negative correlation between soft wheat quality characteristics and the quantities of individual protein fractions of gliadin and reduced glutenin analyzed by size-exclusion high performance liquid chromatography (SE-HPLC).

Variations in molecular weight distributions of native flour proteins have routinely shown significant associations with wheat quality characteristics and so their inclusion in predictive algorithms has a strong potential to improve quality evaluations (Bangur et al 1997; Morel et al 2000; Ohm et al 2006, 2008). For example, Ohm et al (2006, 2008) demonstrated that quality characteristics have distinct associations with specific protein fractions separated according to molecular weight by presenting collective correlation coefficients estimated using absorbance area (AA) and area percentage data of SE-HPLC at 0.05-min retention time intervals across the whole elution profile. These results also suggested that SE-HPLC data from flour native proteins could be applied to develop prediction models of wheat quality characteristics by multivariate analysis methods (Ohm et al 2006, 2008).

Despite significant associations of proteins with soft wheat quality characteristics (Souza et al 1994; Labuschagne and van Deventer 1995; Hou et al 1996a,b; Igrejas et al 2002a,b), no research has been conducted to investigate their relationships with variations in molecular weight distributions of whole-wheat proteins. This study was performed to investigate whether SE-HPLC chromatograms of whole-wheat protein extracts could be used for quality evaluations in wheat breeding programs and in industry. The use of whole wheat protein extract is expected to help greatly to evaluate wheat quality rapidly because the laborious and time-consuming flour milling is not required. The specific objectives

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were to study relationships between variations in protein molecular weight distributions and quality characteristics, and to investigate whether prediction models of quality characteristics could be calculated using molecular weight distribution data from wheat protein extracts in soft wheats. This study was conducted using proteins extracted from whole wheat after high-temperature treatment to decrease protease activity that prevents SE-HPLC analysis of native proteins.

MATERIALS AND METHODS

The total of 45 wheat samples included 44 soft white common winter wheat samples and 1 club wheat sample harvested in 2003 at Corvallis, OR (21 cultivars) and Hermiston, OR (24 cultivars). Single kernel (SK) hardness index was measured with a single kernel characterization system (model 4100, Perten Instruments, Huddinge, Sweden) according to Approved Method 55-31 (AACC International 2000). Flour was milled using a Quadrumat Sr mill (C.W. Brabender) (AACC Approved Method 26-50) after conditioning wheat to 14% moisture content for 24 hr.

Nitrogen was determined by nitrogen combustion analysis (AACC Approved Method 46-30) using a Dumas nitrogen analyzer (Leco, St. Joseph, MI) and protein content was calculated as N × 5.7 on 12 and 14% moisture bases for wheat and flour, respectively. Flour mixing characteristics were analyzed using 10-g mixograph (National Mfg., Lincoln, NE) according to AACC Approved Method 54-40A. Optimum water absorption and mixing tolerance were determined by standard methods of the Western Wheat Quality Laboratory. Experimental cookie baking was performed for sugar-snap (AACC Method 10-52) and wire-cut cookies (AACC Method 10-54). Wire-cut cookie hardness was determined by measuring the maximum force required to break a cookie using the a texture analyzer (TAXTPlus, Stablemicrosystems, UK). Cookie maximum breaking force was measured by pressing a cookie using a blade 70 mm wide and 3 mm thick at a crosshead speed of 2.0 mm/sec on a 3-point bend rig. The base gap of the 3-point bend rig was adjusted to 50 mm. Sponge cakes were baked according to the procedure of the Western Wheat Quality Laboratory.

Extraction and SE-HPLC of Proteins

Whole grains were ground using a cyclone sample mill (Udy, Fort Collins, CO) using a 0.5-mm sieve. Whole wheat flour proteins were extracted by the procedure of Morel et al (2000) with minor modifications (Ohm et al 2006). Wheat protein was extracted from a sample of 160 ± 0.05 mg (adjusted to 14% moisture content) by sonication (Sonic Dismembrator 100, Fisher Scientific). In the preliminary experiment, some samples had a protein degradation problem in SE-HPLC analysis, even after attempting to reduce protease activity using the heating procedure employed by Larroque et al (2000). The protein degradation problem may have been related to the different protein extraction procedures or excessive enzyme activity in the samples used in this experiment. A different heating procedure was performed to eliminate the enzyme activity. Whole grain flour samples in 20 mL of 1% SDS and 0.1*M* sodium phosphate buffer (pH 6.9) were agitated for 30 min at 98°C in 50-mL centrifuge tubes to reduce endogenous enzymatic activity (Saint Pierre et al 2008). After cooling to room temperature, samples were sonicated for 3 min at a power setting of 30% (5W) (Morel et al 2000). The mixture was then centrifuged for 30 min at 30,000 × *g* (RC5 Superspeed refrigerated centrifuge, Du Pont Instrument, Newtown, CT) and the supernatant was filtered through a membrane filter (0.45 μm HV Millipore, DuraPore). HPLC was performed using a separation module (2695, Waters, Milford, MA) and a BIOSEP SEC S4000 size-exclusion column (600 × 7.5 mm, Phenomenex, Torrance, CA) with a guard column (75 × 7.5 mm) (Batey et al 1991). Filtered supernatant (20 μL) was injected and eluted by 50% acetonitrile in water with 0.1% trifluroacetic acid for 30 min with a flow rate of 1 mL/min. Solutes were detected at 214 nm using a photodiode array detector (2996, Waters, Milford, MA).

TABLE I
Mean and Standard Deviation (SD) Values of Quality Parameters

Quality Parameters	Mean	SD
Single kernel hardness index	39.3	8.1
Single kernel weight (mg)	39.7	4.8
Wheat protein (%, 14% mb)	9.5	0.8
Flour yield (%)	66.6	2.2
Break flour yield (%)	32.4	2.0
Flour protein (%, 14% mb)	7.5	0.6
Flour ash (%, 14% mb)	0.42	0.03
Solvent retention capacity		
Water	56.4	2.7
Sucrose	82.5	4.7
Sodium carbonate	67.8	3.7
Lactic acid	91.0	17.3
SDS sedimentation (cm)	43.7	10.4
Mixograph		
Water absroption (%, 14 % mb)	53.8	2.3
Tolerance	3.1	1.4
Sugar-snap cookie diameter (cm)	9.3	0.2
Wire-cut type cookie		
Diameter (cm)	7.6	0.2
Thickness (mm)	2.9	0.1
Hardness (g)	2,117	276
Sponge cake volume (cm^3)	1,303	34

Data Analyses

Mean and standard deviation values and correlation coefficients among quality parameters were calculated using the PROC CORR procedure (v.8.0, SAS Institute, Cary, NC). Absorbance data from SE-HPLC chromatograms of protein extracts were transformed and analyzed using an inhouse program that was developed using MATLAB (v.6, The MathWorks, Natick, MA) (Ohm et al 2006). Absorbance data were interpolated to 0.01-min intervals by spline methods and used to calculate absorbance area (AA). The AA was calculated by mean absorbance × time interval (0.01 min). The sum of AA for each retention time interval of 0.05 min between 12.0 and 24.5 min of runtime was used for data analysis. The percentage of AA (A%) for each 0.05-min time interval was obtained by calculating percentage of AA of each time interval over the sum of all AA values between 12.0 and 24.5 min of runtime. The A% could represent AA adjusted to the same level of protein content. Simple linear correlation coefficients (r) were calculated between both AA and A% and quality parameters for each 0.05-min retention interval and shown as a continuous spectrum over retention time (correllogram).

Prediction models were developed by continuum regression, using PLS_Toolbox (v.2.1, Eigenvector Research, Manson, WA) and MATLAB software (The MathWorks) (Stone and Brooks 1990) using standardized data as described by Ohm et al (2006, 2008). Model performance was tested by cross-validation and evaluated by coefficient of determination (R^2) and root mean square error (RMSE).

RESULTS AND DISCUSSION

Relationships Between Wheat Quality and Cookie Characteristics

The sample set of soft white winter and club wheat breeding lines and cultivars showed kernel, flour, and baking characteristics within the ranges expected of typical Pacific Northwest soft wheat cultivars and varieties (Bettge and Morris 2007) (Table I). The relationships between quality characteristics were shown by simple linear correlation coefficients (Table II). Sugar-snap cookie diameter had significant ($P < 0.001$) correlations with

wire-cut cookie diameter and thickness. Notably, the relationship between the hardness of wire-cut cookies and sugar-snap cookie diameter had a numerically small correlation coefficient, even though it was significant at $P < 0.05$. This suggested that testing sugar-snap cookie diameter alone might be insufficient to evaluate the potential of a flour to produce tender wire-cut type cookies, even though flours producing cookies with larger diameters produce more tender textures in most other soft wheat products (Wade 1988; Miller and Hoseney 1997). SK hardness index did not show a significant correlation with break flour yield possibly due to the narrow range of hardness index in this sample set. Bettge and Morris (2007) reported a greater range of 13.7–52.1 for SK hardness index than the 20.4–51.4 range observed in this sample set. Despite the narrower range, SK hardness index was negatively associated with both sugar-snap and wire-cut cookie diameters, and with sponge cake volume ($r = –0.38$, $P < 0.05$). Wheat kernel hardness has affected flour particle size distribution, damaged starch content, and viscosity of water-extractable arabinoxylan (Wade 1988; Igrejas 2002a). Break flour yield had significant positive correlations with sugar-snap and wire-cut cookie diameters and negative associations with wire-cut cookie thickness and hardness. This result supports the generally known association of high break flour yield with the production of high-quality cookies (Faridi et al 2000).

Wheat and flour protein contents did not show significant correlations with cookie characteristics, except for cookie hardness (Table II), despite the commonly observed negative association between cookie diameter and protein content (Wade 1988; Bettge and Morris 2007). Nonsignificant associations of protein content with cookie diameter also have been reported, specifically when test cultivars were limited to only soft wheats. (Abboud et al 1985; Nemeth et al 1994; Fustier et al 2007). Significant correlations occurred between SDS sedimentation volume and cookie characteristics (Table II). This result suggests that qualitative variation in protein could affect cookie characteristics more than quantitative variation when the range of protein contents is as narrow as in the present research.

Mixograph water absorption and tolerance had negative correlations with cookie diameters and positive correlations with cookie thickness and hardness (Table II). Significant correlation coefficients indicated that wheat flours with high SDS sedimentation volume, mixograph water absorption, and tolerance have poor quality for cookie making, probably because of viscosity increases related to oxidative gelation or gluten network formation during baking, both of which retard or even shrink cookie spread (Slade et al 1989; Bettge and Morris 2000; Pedersen et al 2004).

Kernel Hardness, Milling Yields, and SE-HPLC

Although variations in protein quantity did not affect kernel hardness as indicated by insignificant associations of SK hardness index with wheat and flour protein contents in this sample set (Table II), SK hardness index showed significant ($P < 0.05$) and positive correlations with A% of protein fractions eluted at 12.6–12.8 min, 16.8–18.8 min, and 24.2–24.7 min, and negative correlations with protein fractions eluted at 13.3–14.4 min, and 21.3–21.9 min (Fig. 1A). Significant correlations of A% values with those protein fractions suggested that compositional variations in those protein fractions could affect kernel hardness among the soft wheats used in this research. Specifically, the fractions that eluted at 21.3–21.9 min, the main components of which are reported to be low molecular weight albumins and globulins (Larroque et al 1997), also affected variation of kernel hardness between hard and soft white winter wheats (Ohm et al 2006). In addition to A%, the AA of protein fractions eluted at 24.2–24.7 min was also positively associated with kernel hardness, suggesting that both the quantitative and qualitative variations of this soluble protein fraction affect variation of wheat kernel hardness in this sample set. Simmonds et al (1973) also reported associations of soluble proteins with kernel hardness. However, the monomeric gliadin fraction that showed significant differences between hard and soft wheats by Ohm et al (2006) did not have any significant association with SK hardness index, probably due to the narrower range of kernel hardness of the samples used in this study.

Break flour yield showed significant negative correlations with AA values of protein fractions eluted at 12.5–13.1 min and 19.5–21.0 min (Fig. 1B). Protein fractions at 12.5–13.1 min are mainly composed of high molecular weight polymeric proteins that have close relationships with dough strength (Morel et al 2000; Ohm et al 2006). The negative correlations could be due to selection pressure for low dough strength and high break flour yield in soft wheat breeding programs. Relative amounts of gliadin fractions separated by HPLC have significant correlations with flour particle size distribution, another index of wheat hardness (Simmonds et al 1973; Huebner and Gaines 1991; Rogers et al 1993; Ohm et al 2006). Absorbance area and A% of protein fractions that eluted at 19.8–20.2 min, which are mainly composed of gliadins (Larroque et al 1997), were also significantly and negatively correlated with break flour yield in this research. This result supported the significant association of gliadins with flour particle size distribution because flours with finer particle size distributions are generally produced from wheats with higher break flour yields (Faridi et al 2000).

TABLE II
Simple Linear Correlation Coefficients Between Quality Parameters and Cookie Characteristics[a]

		Wire-Cut		
Quality Characteristics	**Sugar-Snap Diameter (cm)**	**Diameter (cm)**	**Thickness (mm)**	**Hardness (g)**
Wire-cut type cookie characteristics				
Diameter	0.822***			
Thickness	–0.645***	–0.809***		
Hardness	–0.322*	–0.444**	0.396**	
Single kernel hardness index	–0.644***	–0.505***	ns	ns
Single kernel weight (mg)	0.306*	0.395**	ns	–0.342*
Break flour yield (%)	0.522***	0.515***	–0.550***	–0.325*
Protein content (%)				
Wheat (12 % mb)	ns	ns	ns	0.515***
Flour (14 % mb)	ns	ns	ns	0.372*
SDS sedimentation (cm)	–0.443**	–0.540***	0.582***	0.533***
Mixograph characteristics				
Water absorption (%)	–0.687***	–0.858***	0.722***	0.393**
Tolerance	–0.555***	–0.559***	0.518***	0.304*

[a] *, **, and ***, correlation coefficient is significant at $P < 0.05$, 0.01, and 0.001, respectively; ns, correlation coefficient is not significant at $P < 0.05$.

Area % of protein fractions at 21.4–22.4 min, which also had significant negative associations with wheat kernel hardness (Fig. 1A), showed positive correlations with break flour yield (Fig. 1B). This result suggests that these protein fractions could be associated with negative correlations between wheat kernel hardness and break flour yield that have been reported as statistically significant (Ohm et al 1998).

Flour yield had a significant negative correlation with AA and A% at 13.0–13.5, and 19.9–21.1 min (Fig. 1C). Positive correlations occurred between flour yield and A% at 16.3–19.1 and 23.2–24.7 min. The AA and A% values of these protein fractions also showed significant correlations with hardness and break flour yield. These results suggest that variation in protein fractions could affect flour yield through associations with kernel characteristics.

Mixing Characteristics and SE-HPLC

Mixograph water absorption and mixing tolerance had significant correlations with AA and A% of high molecular weight polymeric protein fractions (Fig. 2A and B). This observation is consistent with the reports of other researchers (Bangur et al 1997; Morel et al 2000; Ohm et al 2006). These results confirm that SE-HPLC analysis of wheat proteins is applicable for the evaluation of flour mixing characteristics. The low molecular weight monomeric protein fractions were also significantly correlated with mixing characteristics (Fig. 2A and B) as they were with SK hardness index (Fig. 1A). The A% of protein fraction at 21.5–22.3 min had negative correlations with mixograph water absorption and tolerance (Fig. 2) as it did for SK hardness index (Fig. 1A). These results suggest that these protein fractions are related to the association between soft wheat kernel texture and low water absorption and low dough strength.

Cookie Characteristics and SE-HPLC

Diameters of both sugar-snap and wire-cut cookies were negatively correlated with the high molecular weight polymeric and very low molecular weight monomeric protein fractions (Fig. 3A and B) that were also significantly associated with mixograph characteristics (Fig. 2A and B). These protein fractions were also positively associated with cookie thickness, suggesting that the variations in absolute and relative quantities of these protein fractions are associated with variation in cookie diameter and thickness. Correlations of sponge cake volume with AA and A% values showed a similar pattern to those of cookie diameter, but the coefficients were rarely significant ($P < 0.05$) (data not shown). Area% values of protein fractions eluted at 13.0–14.3, and 21.3–22.3 min also associated positively with cookie diameter and negatively with cookie thickness (Fig. 3C), indicating that, with other things being equal, a high percentage of these protein fractions in flour protein could contribute to improvement of cookie spread. The contrasting association of these protein fractions,

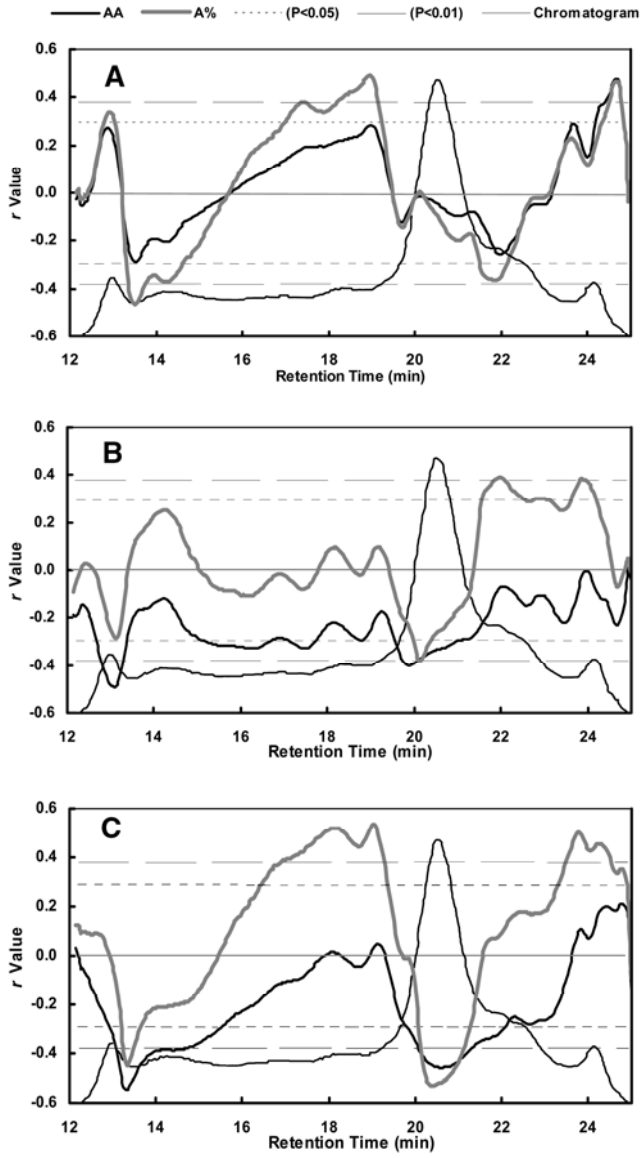

Fig. 1. Linear correlation coefficients (r) of single kernel hardness index (**A**), break flour yield (%) (**B**), and flour yield (%) (**C**) with absorbance area (AA) and area percentage (A%) of protein extracts separated by size-exclusion HPLC.

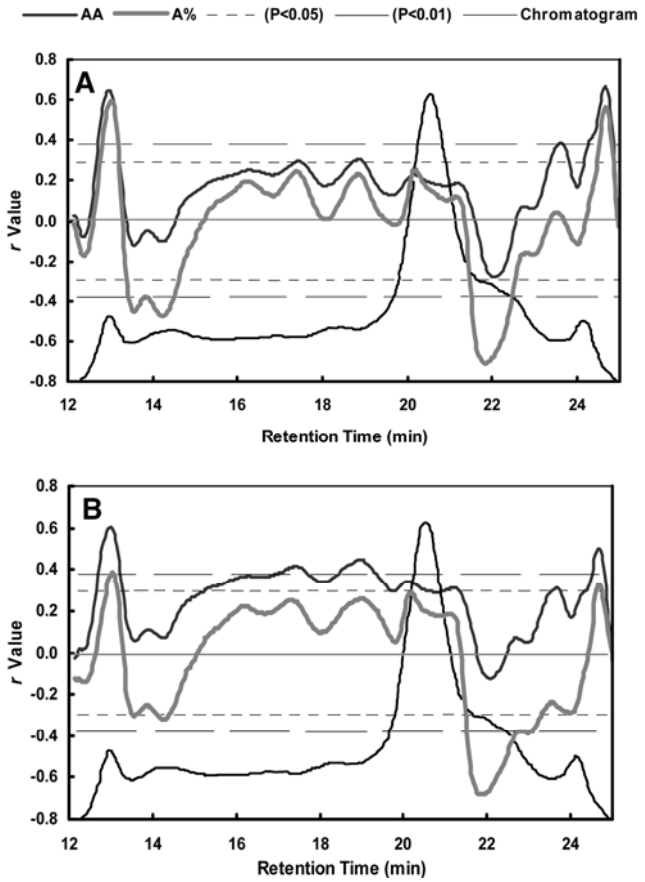

Fig. 2. Linear correlation coefficients (r) of mixograph water absorption (%) (**A**) and tolerance (**B**) with absorbance area (AA) and area percentage (A%) of wheat protein extracts separated by size-exclusion HPLC.

specifically the high molecular weight polymeric protein fraction, with cookie diameter and mixograph tolerance represent a generally known relationship that flour that forms strong dough is not appropriate for cookie making due to the elastic gluten network formation that could cause shrinkage of cookie spread during baking (Slade et al 1989). The A% of the protein fraction at 20.0 –21.0 min had positive correlations with cookie thickness although it did not have a significant association with cookie diameter. This result suggested that a high proportion of those protein fractions could act to increase cookie thickness without a significant effect on cookie diameter.

Hardness values of wire-cut cookies showed positive correlations with AA values (Fig. 3D) of almost all protein fractions, implying that quantitative variations in proteins mainly affect hardness of cookies, as shown by significant correlations with protein contents (Table II). Specifically, high molecular weight polymeric proteins appeared to affect cookie hardness more than other protein fractions due to the higher correlation coefficients with cookie-breaking forces (Fig. 3D).

Developing Prediction Models Using SE-HPLC

Analaysis of variance indicated that growing location and cultivar significantly affected peak area values of the HPLC chromatograms (data not shown). Specifically, effect of growing location was associated primarily with variations in AA value, while cultivar was associated primarily with variations of A% values. The variation in HPLC profiles among the 45 samples were considered large enough to allow use of the SE-HPLC data to develop models for the prediction of soft wheat quality characteristics. Ohm et al (2006) also reported significant variations of SE-HPLC data for hard winter wheats. Through principal component analysis, 15 principal component scores were calculated and explained 98% of the variation of AA and A% data of SE-HPLC (data not shown). The principal component scores and transformed principal component scores were used for model calibration by continuum regression. Coefficients of determination and RMSE values for calibration models and cross-validations are shown in Table III. The calibration model of single kernel hardness index showed an R^2 value of 0.87 and cross-validation indicated an R^2 value of 0.77. In calibration models, R^2 = 0.89 for flour yield and R^2 = 0.85 for break flour yield, and R^2 = 0.82 and R^2 = 0.77 for cross-validations, respectively. These results indicate that SE-HPLC data of whole wheat protein could be used to estimate milling quality when sample amount is very limited, without recourse to additional laboratory milling analyses.

The calibration model of mixograph water absorption showed good predictive capacities with $R^2 > 0.91$. Mixing tolerance showed a $R^2 > 0.73$ and the same RMSE values of 0.7 for both calibration and cross-validation (Table III). These results suggest the possibility of applying SE-HPLC data for mixing quality evaluation of soft wheats, as shown for the hard wheat flours by Ohm et al (2006).

Although cookie diameters were not significantly correlated with protein content (Table II), high $R^2 > 0.93$ for calibration models and $R^2 > 0.89$ for cross-validation (Table III) were estimated for cookie diameter values and thickness, thus SE-HPLC data from whole wheat flours could be used to predict cookie quality of soft wheats without flour milling and cookie baking. When compared with other cookie characteristics, prediction of cookie hardness was considered more difficult, but the $R^2 > 0.68$ for calibration model and cross-validation still indicated that the calibration model could be used for the evaluation of soft wheat breeding lines. When considering the rarely significant correlation of cake volume with SE-HPLC data, sponge cake volume here showed unexpectedly high $R^2 > 0.83$ for calibration and $R^2 > 0.73$ for cross-validation, probably due to the use of multivariate analysis of data with transformation that helped to explain variations that did not have linear relationships with AA and A%. The multivariate methods used here have advantages for model development such as avoidance of multicollinearity and the addition of the transformed data that help explain nonlinear and interaction relationships (Ohm et al 2006).

SUMMARY

This study was performed to investigate the relationships among quality characteristics of soft white winter wheat with molecular weight distribution of proteins. Although protein con-

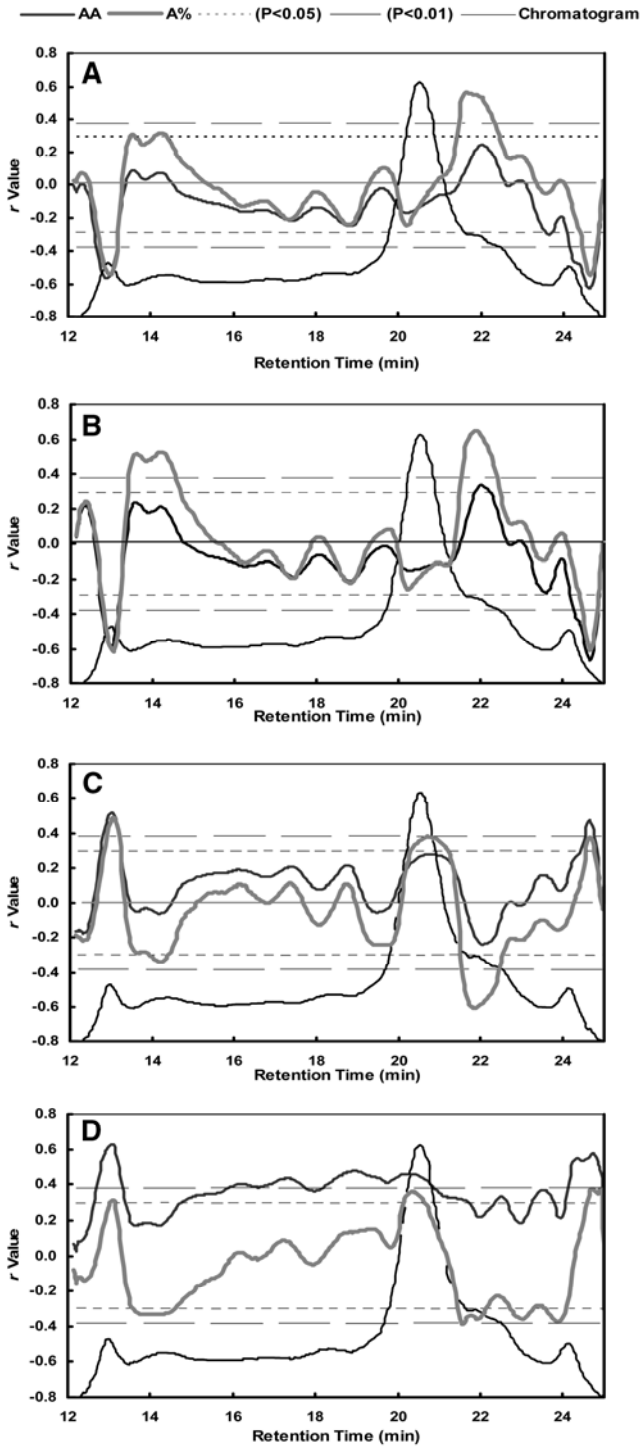

Fig. 3. Linear correlation coefficients (r) of sugar-snap cookie diameter (**A**) and wire-cut cookie diameter (**B**), thickness (**C**), and hardness (**D**) with absorbance area (AA) and area percentage (A%) of wheat protein extracts separated by size-exclusion HPLC.

TABLE III
Coefficient of Determination (R^2) and Root Mean Squares of Errors (RMSE) of Calibration Models and Cross-Validation for Quality Parameters

Quality Parameters	Latent Variables	Calibration		Cross-Validation	
		R^2	RMSE	R^2	RMSE
SK Hardness Index	3	0.865	3.4	0.771	3.8
Flour yield (%)	2	0.890	0.9	0.824	0.9
Break flour yield (%)	3	0.850	0.9	0.772	1.0
Mixograph					
Water absorption (%)	3	0.952	0.5	0.916	0.6
Tolerance	3	0.810	0.7	0.739	0.7
Sugar snap cookie					
Width (cm)	3	0.935	0.1	0.893	0.1
Wire cut cookie					
Diameter (cm)	3	0.952	0.1	0.932	0.1
Thickness (mm)	2	0.972	0.3	0.925	0.3
Hardness (g)	3	0.833	116.0	0.680	140.1
Sponge cake volume (cm^3)	3	0.839	15.8	0.736	17.7

tent did not have significant correlations with cookie characteristics, except for cookie hardness, significant association of SDS sedimentation volume with cookie characteristics suggests that qualitative characteristics of protein affected soft wheat quality. The AA and A% of protein fractions separated by SE-HPLC showed significant correlations with specific quality characteristics, confirming that compositional variations of wheat kernel proteins, as described by changes in molecular weight distribution, also affected soft wheat quality. This result also suggested that SE-HPLC chromatogram data could be applied to the development of calibration models for soft wheat quality characteristics, as already shown for hard wheats (Ohm et al 2006, 2008). The calibration models that were developed with the application of multivariate methods, such as principal component analysis and continuum regression, showed $R^2 > 0.81$, indicating that those models could be possibly be applied to quality evaluation of soft wheat cultivars. Because proteins were extracted specifically from whole wheat flour after treating at high temperature (95°C for 30 min) to prevent enzyme activity, the analytical procedures performed in this experiment could be applied for quality evaluation without recourse to labor-intensive flour sample preparation such as experimental flour milling, specifically when sample amount is very limited.

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