Relationships Between Protein Composition and Mixograph and Loaf Characteristics in Wheat[1]

H. DONG,[2] R. G. SEARS,[2] T. S. COX,[3] R. C. HOSENEY,[4]
G. L. LOOKHART,[5] and M. D. SHOGREN[5]

ABSTRACT

To study the relationships between protein composition and baking properties of 135 randomly selected wheat (*Triticum aestivum*) lines from a randomly mated population, we analyzed gliadin proteins by acidic buffer polyacrylamide gel electrophoresis (PAGE) and high molecular weight (HMW) glutenin proteins by PAGE in the presence of sodium dodecyl sulfate. The amount of protein in each gliadin band was estimated from densitometer scans, and presence or absence of particular glutenin subunits was recorded. Simple correlations were calculated among individual gliadin bands, Payne glutenin score, and each of five quality parameters: mixing time, mixing tolerance, water absorption, loaf volume, and crumb grain score. Five gliadin bands were correlated with loaf volume, two with crumb grain score, 12 each with mixing time and mixing tolerance, and six with water absorption. No correlations were detected between protein concentration and presence of any particular gliadin or glutenin proteins. Payne score was significantly correlated only with loaf volume and mixing time. Significant correlations were found between protein concentration and both loaf volume and absorption and between mixing tolerance and crumb grain score. No associations were found between total protein content and mixing properties. Multiple regression analyses were conducted for each of the functional tests, with the individual gliadin and/or HMW glutenin bands as independent variables. Prediction of 26–45% of the variation in quality parameters required eight to 11 gliadin and glutenin bands. Glutenin subunits 5+10 had the most consistently positive effect on most of the quality measurements. These biochemical methods can be used to identify wheat genotypes with specific HMW glutenin and gliadin composition in parental and early-generation selections, but phenotypic quality traits must be considered as well.

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Wheat (*Triticum aestivum*) flour mixed with water produces an elastic, cohesive dough. The unique properties of wheat dough are due to its water-insoluble proteins, as can be demonstrated by removing lipids, starch, and water-soluble carbohydrates from the flour; the remaining gluten, which is 80% protein, still forms a hydrated, rubbery mass (Wall 1979). The breadmaking potential, or intrinsic quality, of a wheat cultivar depends largely on its gluten quality and quantity.

Wheat breeding programs have selected for improved bread-baking quality for many years. The mixograph has been used to help predict functional dough mixing properties of wheat genotypes in many hard wheat breeding programs in the United States. The instrument is very useful for estimating important physical dough properties in early-generation progenies and can also be used to predict bread loaf volume, mixing requirement, dough oxidation requirement, and baking water absorption (Finney and Shogren 1972).

Polyacrylamide gel electrophoresis (PAGE) (Lookhart et al 1982) and sodium dodecyl sulfate PAGE (SDS-PAGE) (Payne et al 1981a) have been used to detect differences among wheat genotypes in protein composition. Determination of the association between gliadin or high molecular weight (HMW) glutenin subunits and mixograph mixing properties may provide a quick and simple way to evaluate the baking quality of germ plasm, parents in breeding programs, or selections (Campbell et al 1987).

Wheat gliadin and glutenin proteins are oligogenic traits controlled by genes or gene clusters located on homeologous group 1 and 6 chromosomes (Boyd et al 1967, Solari and Favret 1967). Specifically, genes controlling gliadin proteins are located on the short arms of chromosome groups 1 and 6 (Branlard 1983), and genes encoding glutenin proteins are located on the long arms of chromosome group 1 (Orth and Bushuk 1974, Payne and Lawrence 1983). Different cultivars usually have different band patterns of gliadin or glutenin proteins, which may affect baking performance.

Differences among cultivars in breadmaking potential result primarily from the quantitative and qualitative differences in gluten proteins in common wheat. HMW glutenin subunits 5 and 10 have been reported consistently to be strongly correlated with various quality measurements in different sets of wheat genotypes (Payne et al 1983; Moonen and Zeven 1985; Lawrence et al 1987, 1988; Ng and Bushuk 1988, Dong et al 1991). Glutenin subunits 17 and 18 were also reported to have a greater effect on baking quality than other B-genome-controlled subunits (Lawrence et al 1988). Pogna et al (1982) reported close associations between gliadin 43.5 and good baking quality and between gliadin 40 and poor quality.

Relatively little research has been done on the association between gluten components and mixograph characteristics in common wheat. We attempted to evaluate the effects of each group of proteins and their individual bands on dough mixing properties and to associate certain specific bands with mixing and baking parameters. We analyzed multiple regression models of the quality traits, with protein components or individual bands as independent variables and mixograph and baking criteria as dependent variables, for a sample of 135 randomly selected wheat lines from a randomly mated population.

MATERIALS AND METHODS

Wheat Selections

The 135 hard winter wheat lines used in a study of HMW glutenin subunits (Dong et al 1991) also were used in this study. These lines were randomly selected from a bulk population derived from 87 hard winter and spring wheat parents. Three-way crosses (winter/spring//winter) were made in 1980 and harvested as a bulk in 1981. Two subsequent generations were randomly mated with chemical hybridizing agents. In 1985, 135 F_4 individual plants were randomly selected, and in 1986 the F_4-derived F_5 lines were planted at the Agronomy Research Farm at Hutchinson, KS. A small amount of seed was ground for primary electrophoresis. In 1987, F_4-derived F_6 lines were planted at Ashland Research Farm near Manhattan, KS, and the harvested seeds were used for mixograph tests and gel electrophoretic analysis. In 1988, F_4-derived F_7 lines were increased at Manhattan and Hutchinson, and the grain harvested from both sites was bulked for each genotype and evaluated for baking quality.

[1]Contribution 90-487-J of the Kansas Agricultural Experiment Station.
[2]Department of Agronomy, Kansas State University, Manhattan 66506.
[3]U.S. Department of Agriculture, Agricultural Research Service (USDA-ARS) and Department of Agronomy, Kansas State University.
[4]Department of Grain Science, Kansas State University.
[5]USDA-ARS, U.S. Grain Marketing Research Laboratory, Manhattan, KS 66502.

Gliadin Extraction

Ground wheat (35 mg) was extracted with 100 μl of 70% aqueous ethanol. The samples were incubated at room temperature with occasional mixing for about 1 hr then centrifuged at 15,600 × *g* for 10 min. Five drops of glycerol was added to the supernatant. One drop of 1% methyl green solution was added as a tracking dye (Lookhart et al 1982).

Glutenin Extraction

Wheat flour (35 mg) was extracted in a conical centrifuge tube in 800 μl of 0.125*M* Tris buffer, pH 6.8, containing, per liter, 10 g of SDS, 100 g of glycerol, 0.1 g of bromophenol blue, and 50 g of 2-mercaptoethanol. The samples were allowed to stand at room temperature for 1 hr, then in boiling water for 2 min, followed by 1 hr at room temperature. They were then centrifuged at 15,600 × *g* for 10 min.

Electrophoretic Separation of Gliadin and HMW Glutenin Proteins

Electrophoretic separation of gliadin proteins was performed according to the procedures described by Lookhart et al (1982). Gliadin extract (10 μl) was loaded, and 20 samples per gel were electrophoresed at 500 V for 2 hr. The cultivar Marquis was used on each gel as a standard check for band mobility.

Gels were subsequently stained for 10–12 hr in 200 ml of solution composed of 50 ml of 50 ml L^{-1} trichloroacetic acid (TCA) and 6 ml of 10 g L^{-1} Coomassie Brilliant Blue R. The gels were then transferred to 200 ml of destaining solution, which had the same TCA concentration as the staining solution but no Coomassie Blue, for one day at room temperature. They were then put in a refrigerator for at least three days without changing the destaining solution. The gels were photographed with 4- × 5-in. (10.2- × 12.7-cm) Kodak technical pan film. The developed films were then printed to another film to get a positive image of the gel. The printed films were scanned on an LKB 2202 Ultroscan laser densitometer (LKB, Bromma, Sweden) to measure the density of the gliadin bands. The density of each band was expressed as a percentage of the total density of that line.

Seed from F_4-derived F_5 lines was used for gliadin PAGE. Forty F_4-derived F_6 lines were also electrophoresed to confirm the gliadin band patterns. No differences were detected between these two generations. The nomenclature of Bushuk and Zillman (1978) was used for gliadin band identification.

An SDS discontinuous system based on modifications of Laemmli (1970) was used for separating glutenin proteins. A 20-μl sample was loaded and run at a constant current of 60 mA for 5–5.5 hr. The stain, destain, and photography procedures were the same as for gliadin gels. The flours used for glutenin detection were F_4-derived F_7 lines. The HMW glutenin subunit designation followed Payne and Lawrence (1983).

Functional Tests

All functional tests were conducted at the U.S. Department of Agriculture Grain Marketing Research Laboratory in Manhattan, KS. Ten-gram mixographs (Finney and Shogren 1972) were performed. Mixing time (minutes to the peak of the curve), mixing tolerance (width of the curve 2 min after peak), and water absorption were measured. Protein concentration was measured by near-infrared reflectance in 1987 and by the Kjeldahl method in 1988, with a conversion factor of 5.7 between nitrogen and protein.

Ten-gram loaves were baked by the procedure of Shogren et al (1969) except that ascorbic acid was used instead of potassium bromate. Crumb grain was rated visually on a scale from 0 to 10, with 0 unsatisfactory, 2 unsatisfactory to questionable, 4 questionable, 6 questionable to satisfactory, 8 satisfactory, and 10 very satisfactory. Two loaves were baked for each sample, and the mean values were used in the analysis.

Statistical Analysis

We computed simple correlations between each group of proteins, individual gliadin bands, and average quality measurements. A score was assigned to each line based on the HMW glutenin composition of the line, as described by Payne (1986), and simple correlations were computed between these Payne scores and the quality parameters.

For the purpose of prediction, multiple regression models were established with densities of individual gliadin bands, presence or absence of glutenin bands, and protein concentration as independent variables. The prediction equation can be written as

$$y = a + b_1x_1 + b_2x_2 + \ldots + b_nx_n ,$$

where y is the quality characteristic to be estimated, a is the intercept, b_1–b_n are slopes, and x_1–x_n are the independent variables included in the regression equation. An F-test for significance was applied to the regression models to determine the predictive values.

Four quality indices were also constructed to estimate overall quality. The original quality variables were standardized as follows:

$$X_s = (X - \bar{X})/\sigma_X ,$$

where X_s is the standardized value of a variable, X is the original value of the variable, $\bar{X}$ is the mean of the variable over all the lines tested, and σ_X is the standard deviation of the variable.

The following four indices were constructed:

$$ID_1 = (X_{s\,MT} + X_{s\,MTL})/2 ,$$

$$ID_2 = (X_{s\,LV} + X_{s\,CG} + X_{s\,AB})/3 ,$$

$$ID_3 = (ID_1 + ID_2)/2 , \text{ and}$$

$$ID_4 = (X_{s\,LV} + X_{s\,CG} + X_{s\,MT} + X_{s\,MTL} + X_{s\,AB})/5 ,$$

where $X_{s\,LV}$, $X_{s\,CG}$, $X_{s\,MT}$, $X_{s\,MTL}$, and $X_{s\,AB}$ are the standardized observations for loaf volume, crumb grain score, mixing time, mixing tolerance, and absorption, respectively. ID_1 can be interpreted as a mixing index and ID_2 as a baking index. ID_3 and ID_4 are overall quality indices; ID_3 is the mean of ID_1 and ID_2, and ID_4 is an unweighted mean of all five traits.

RESULTS

Because of the limited amount of seed available, baking tests were performed only in 1988. Mixograms were done on material grown in both 1987 and 1988, providing two years of estimates for mixing time, mixing tolerance, absorption, and flour protein concentration. To estimate the relative effects of heredity and environment on the quality traits, we computed simple interyear correlations (equivalent to heritability in standard units [Frey and Horner 1957]). Correlations were highly significant (P = 0.001) for all four traits, but the correlation coefficients varied widely. Mixing time and mixing tolerance between years correlated relatively well (r = 0.65 and 0.60, respectively) compared to protein concentration and water absorption (r = 0.47 and 0.27, respectively). This result reflected stronger genotype × environment interaction for the latter traits.

Correlations Between Protein Concentration and Quality Measurements

Flour protein concentration was highly correlated (P = 0.001) with loaf volume (r = 0.58) and with absorption in both years (r = 0.71 in 1987 and 0.60 in 1988). No associations were found between flour protein concentration and mixing time, mixing tolerance, or crumb grain score in either year. Mixing time and mixing tolerance were also significantly correlated (P = 0.001) in both years (r = 0.69 in 1987 and 0.79 in 1988). Mixing time was not correlated with loaf volume.

Small but significant (P = 0.001) positive associations were found between mixing tolerance and crumb grain score (r = 0.30)

and between crumb grain score and absorption ($r = 0.28$). Water absorption was also significantly correlated ($P = 0.001$ except where noted otherwise) with loaf volume ($r = 0.56$), crumb grain score ($r = 0.28$), mixing time ($r = 0.18$ [$P = 0.05$]), and mixing tolerance ($r = 0.28$).

Associations Between HMW Glutenin Proteins and Quality Characteristics

There were low but significant ($P = 0.01$) correlations between Payne score and two quality traits, loaf volume and mixing time ($r = 0.20$ and 0.23, respectively). Payne score was not correlated with other traits. There were no significant correlations between the number of HMW glutenin bands present and any of the quality measurements.

Means over all lines carrying each of the possible HMW glutenin subunit combinations are shown in Table I. For A-genome-controlled subunits, lines with subunit 2* had greater loaf volume and mixing tolerance, but no allele showed significantly different effects compared to the population mean for any quality trait measured.

Among the B-genome-controlled subunits, the combination of subunits 6 and 8 had the lowest loaf volume, crumb grain score, mixing time, mixing tolerance, and absorption. Subunits 17 and 18 had the highest loaf volume, but only eight selections had this combination. Subunit combination 7 and 8 had a significantly longer mixing time ($P = 0.05$) and better mixing tolerance than the population mean.

Among the D-genome-coded glutenins, subunits 5 and 10 had higher means than the other subunit combinations for most of the quality traits, and the mixing time was significantly longer than the population mean, whereas combinations 2 and 12 and 3 and 12 had significantly shorter mixing times than the population mean.

In this set of randomly derived lines, Payne score had very weak correlations with all quality traits. Therefore, we used the individual HMW glutenin subunits (with 1 = present and 0 = absent) in all multiple regression computations. Using stepwise regression, HMW glutenin subunit combinations and flour protein were used as independent variables to maximize the coefficient of determination (R^2) for each quality trait (Table II). The prediction for loaf volume involved subunits 17 and 20 and protein concentration, with an R^2 value of 0.38. R^2 values for crumb grain score, mixing time, and mixing tolerance were low. None of the glutenin proteins significantly affected absorption.

The predictive model for the mixing index (ID_1) required three glutenins but had a low R^2 of 0.10 (Table III). The baking index (ID_2) equation contained glutenin subunits 12 and 20 and protein concentration, with an R^2 of 0.34. The equation for the overall index, ID_3, included glutenin subunits 5+10, 7, and 20 and protein concentration, with an R^2 of 0.21, and the model for ID_4 contained subunits 7 and 10 plus protein concentration, with an R^2 of 0.24. The equations for all four indices were significant to at least $P = 0.0004$, but none explained a major proportion of the variation in overall quality.

Associations Between Gliadin Proteins and Quality Traits

More than 50 gliadin bands were detected in the 135 wheat lines analyzed. Only 43 were used in the calculations because some bands appeared only in a very few lines. Each individual line had between 19 and 32 bands.

The gliadin proteins are often categorized into four groups, α, β, γ, and ω, according to their electrophoretic mobility, with α having the highest and ω the lowest mobilities. We computed correlations between the proportions of total gliadin protein in each of the four groups and the quality parameters. Because both positive and negative correlations were present among the individual bands within each group, most of the correlations between gliadin groups and quality traits were not significant. Small but significant associations were observed between α-gliadins and mixing time ($r = 0.28$**), β-gliadins and absorption ($r = 0.22$*), and γ-gliadins and mixing tolerance ($r = 0.20$*). There were significant negative correlations between ω-gliadins and mixing tolerance ($r = -0.22$*).

No correlation was found between protein concentration and any of the 43 gliadin bands, indicating that the percentage band intensity is independent of protein concentration. Of the 43 gliadin bands examined, 16 were significantly correlated with at least one of three quality traits measured in 1987, and 20 were associated with at least one of the five traits measured in 1988. Five bands were significantly correlated with loaf volume, three positively and two negatively. Three bands were correlated with crumb grain score. Twelve bands were associated with mixing time, eight of them negatively and four positively, in at least one year, and 11 of these plus one more were associated with mixing tolerance. Three bands were negatively and six positively related to water absorption.

Bands 25 and 46 showed the strongest association with mixing properties. The correlation coefficients between band 25 and mixing time and tolerance were 0.41*** and 0.31**, respectively, in 1987 and 0.27** and 0.22*, respectively, in 1988. The correlations between band 46 and mixing time and tolerance were

TABLE I
Means for Five Quality Traits over All Lines Carrying Each HMW Glutenin Genotype of Each Locus

Glutenin Genotype		No. of Lines	Loaf Volume (cm^3)	Crumb Grain Score	Mixing Time (min)	Mixing Tolerance (cm)	Absorption (%)
A	1	34	89.4	6.8	4.12	1.75	61.8
	2*	34	92.6	7.0	4.28	1.82	61.8
	Null	67	90.5	7.3	4.31	1.76	62.8
B	7	10	87.0	7.2	3.89	1.72	61.9
	7+8	27	90.6	6.9	4.71[a]	1.86	61.6
	7+9	71	91.5	7.3	4.24	1.79	61.7
	6+8	5	85.6	5.8	2.94[a]	1.39[a]	61.5
	17+18	8	92.8	6.4	3.82	1.70	62.2
	20	14	90.6	7.4	4.39	1.76	61.6
D	2+12	17	90.5	6.6	3.85[a]	1.68	61.9
	3+12	20	88.1	6.4	3.51[a]	1.64	61.8
	5+10	98	91.5	7.3	4.47[a]	1.82	61.6
Population mean			90.8	7.1	4.25	1.77	61.7

[a] Significantly different from the population mean ($P = 0.05$). (Significance of differences for loaf volume and crumb grain was not determined because data were from one year only.)

TABLE II
Significant HMW Glutenin Subunits and Protein Concentration (P), Intercepts, and Adjusted Coefficient of Determination (R^2) in the Prediction Equations for Five Quality Traits

Trait	Glutenins and P[a]	Intercept	R^2
Loaf volume	P(4.02); 17(3.14); 20(2.30)	36.82	0.38
Crumb grain	7(1.39); 5+10(0.76); 12(−1.32)	6.76	0.13
Mixing time	6(−0.81); 9(−0.42); 5+10(1.03)	3.61	0.10
Mixing tolerance	2(−0.16); 3(−0.15); 7(0.19); 5+10(0.28)	1.15	0.10
Absorption	P(1.01)	48.83	0.34

[a] Regression coefficients in parentheses.

TABLE III
Significant HMW Glutenin Subunits and Protein Concentration (P), Intercepts, and Adjusted Coefficient of Determination (R^2) in the Prediction Equations for Four Quality Indices

Index	Glutenins and P[a]	Intercept	R^2
ID_1	7(0.55); 9(−0.24); 5+10(0.64)	−0.84	0.10
ID_2	12(−0.35); 20(0.31); P(0.43)	−5.79	0.33
ID_3	7(0.39); 5+10(0.36); 20(0.20); P(0.23)	−3.78	0.21
ID_4	7(0.39); 5+10(0.31); P(0.27)	−4.18	0.24

[a] Regression coefficients in parentheses.

0.18* and 0.22*, respectively, in 1987 and 0.30** and 0.37***, respectively, in 1988. Bands 37.3 and 52.9 had significant negative relationships with mixing time and mixing tolerance. The correlations of band 37.3 with mixing time and tolerance were −0.27** and −0.23*, respectively, in 1987 and −0.29** and −0.44***, respectively, in 1988. The corresponding correlation coefficients for band 52.9 were −0.17* and −0.22* in 1987 and −0.23* and −0.26* in 1988.

Baking quality parameters were regressed against gliadin bands and protein concentration (Table IV). Forty-six percent of the variation in loaf volume could be estimated from knowledge of five gliadin bands, and 29% of the variation in mixing time was predicted from seven gliadin bands. Mixing tolerance involved six gliadin bands ($R^2 = 0.37$) and absorption three ($R^2 = 0.40$). The predictability of crumb grain score was very low ($R^2 = 0.10$), with four significant bands.

When all HMW glutenins, gliadins, and protein concentration were included as independent variables, the D-genome HMW glutenin combination 5+10 plus five and seven gliadin bands were required to explain approximately 40% of the variation in mixing time and mixing tolerance, respectively (Table V). The same glutenin subunits plus three gliadin bands and protein concentration accounted for 45% of the variation in loaf volume. Predicting 26% of the variation in crumb grain score involved five glutenin subunits and two gliadin bands.

The estimation equations for the four quality indices were also calculated with both gliadin and glutenin proteins as independent variables (Table VI). The HMW glutenin proteins 5+10 and 20, gliadins 25 and 37.3, and protein concentration were most often involved as predictors of the indices, and R^2 ranged from 0.39 to 0.44 for all equations. The predictor variables and their coefficients in individual trait equations differed from those in the index equations involving the traits. For example, the mixing index equation included more glutenins and different gliadins than the mixing time or tolerance equations.

TABLE IV
Significant Gliadin Proteins and Protein Concentration (P), Intercepts, and Adjusted Coefficient of Determination (R^2) in Optimum Prediction Equations for Five Quality Traits

Trait	Gliadin Bands and P[a]	Intercept	R^2
Loaf volume	24(−0.52); 31.5(0.90); 39.4(0.89); 50(0.60); P(3.59)	42.46	0.46
Crumb grain score	37.3(0.25); 42.5(0.21); 52.9(−0.31); 70(0.10)	6.55	0.10
Mixing time	25(0.36); 37.3(−0.22); 37.8(−0.18); 46(0.04); 53(−0.06); 68.5(0.09); 70(0.11); P(0.16)	1.26	0.29
Mixing tolerance	25(0.08); 37.3(−0.12); 42.5(−0.12); 46(0.02); 70(0.02); P(0.08)	0.26	0.37
Absorption	47(0.07); 54.5(0.07); 64(0.24); P(0.93)	48.13	0.40

[a] Regression coefficients in parentheses.

TABLE V
Significant Glutenins and Gliadins and Protein Concentration (P), Intercepts, and Adjusted Coefficient of Determination (R^2) in Optimum Prediction Equations for Four Quality Traits

Trait	Glutenin Subunits (Glu), Gliadin Bands (Gli), and P[a]	Intercept	R^2
Loaf volume	Glu5+10(2.01); Gli31.5(−0.64); Gli56.7(0.66); Gli58(−0.22); P(3.71)	40.29	0.45
Crumb grain score	Glu5+10(2.68); Glu7(1.09); Glu8(−0.45); Glu12(−1.36); Gli42.5(0.20); Gli53(−0.31)	7.29	0.26
Mixing time	Glu5+10(0.87); Gli25(0.38); Gli36(−0.22); Gli37.8(−0.24); Gli39.4(−0.21); Gli68.5(0.12); Gli70(0.12)	3.17	0.38
Mixing tolerance	Glu5+10(0.14); Gli21.6(0.08); Gli25 (−0.14); Gli42.5(−0.11); Gli46 (0.02); Gli70(0.02)	0.09	0.39

[a] Regression coefficients in parentheses.

DISCUSSION

The results of this study provide additional evidence for the existence of direct associations between breadmaking quality and certain HMW glutenin and gliadin subunits. Effects of low molecular weight (LMW) glutenin subunits were not determined. In general, correlations between two traits can be caused by pleiotropy, genetic linkage, or chance associations. Studies involving sets of cultivars may detect associations due to any of the three factors. This study involved randomly selected lines from a population with a broad genetic base after several generations of random intermating. The population's parentage comprised 87 genetically diverse hard wheat cultivars including spring and winter wheats adapted to the Great Plains of the United States. Random mating allowed different alleles of various protein genes to combine with each other, reducing the importance of all but direct genetic effects (pleiotropy) of the predictor loci.

Some of the HMW glutenin subunits have consistently shown their effects on baking quality. The associations of subunits 5+10 with good dough and baking quality and of subunits 2+12 and 3+12 with poor dough and baking quality are the most striking in our study and are consistent with previous results (Payne et al 1983, Branlard and Dardevet 1985b, Ng and Bushuk 1988, Dong et al 1991). The superiority of subunits 5+10 at the Glu-D1 locus was much greater for mixing traits and loaf volume than the difference between the other alternatives, 2+12 and 3+12. In comparing wheat isolines, Lawrence et al (1987) found that dough from genotypes with subunits 5+10 had higher resistance, while genotypes with 2+12 had higher extensibility in extensigraph tests. Seventy-five percent of the U.S. hard red wheat and most of the recent cultivars in the Great Plains, including Arkan, Eagle, Hawk, and Newton, have HMW glutenin subunits 5 and 10 (G. L. Lookhart et al, *unpublished*). Selection for baking quality by mixograph results probably has been responsible for increasing the frequency of 5+10.

Among the B-genome-controlled subunits, 17+18 had the strongest associations with loaf volume. Because only nine lines in our study had this subunit combination, the results should be considered preliminary. Lawrence et al (1988) reported that subunit combination 17+18 provided greater loaf volume and longer mixing time and was as effective as 5+10 for improving dough and baking quality; 17+18 also was related to high SDS sedimentation volume (Payne et al 1984, Branlard and Dardevet 1985b). The 17+18 combination is particularly prominent in Australian prime wheat, appearing in 34% of the 106 cultivars studied by Lawrence (1986). This combination is also present in 4% of cultivars from a world collection (Payne et al 1983). G. L. Lookhart et al (*unpublished*) found 17+18 in 5% and 14% of

TABLE VI
Significant Glutenins and Gliadins and Protein Concentration (P), Intercepts, and Adjusted Coefficients of Determination (R^2) in Prediction Equations for Four Quality Indices

Index	Glutenins (Glu) and Gliadins (Gli) and P[a]	Intercept	R^2
ID_1	Glu2(−0.46); Glu3(−0.50); Glu5+10(1.36); P(0.33); Gli25(0.22); Gli37.3(−0.24); Gli46(0.06); Gli53(−0.04)	−3.87	0.41
ID_2	Glu3(−0.27); Glu9(0.22); Glu20(0.31); P(0.42); Gli24(−0.07); Gli31.5(0.09); Gli37.3(−0.11); Gli37.8(−0.07); Gli50(0.03)	−5.73	0.44
ID_3	Glu5+10(0.91); Glu20(0.26); P(0.31); Gli25(0.15); Gli37.3(−0.19); Gli46(0.04); Gli50(0.03)	−5.77	0.42
ID_4	Glu2(−0.21); Glu3(−0.40); Glu5+10(0.25); P(0.33); Gli25(0.12); Gli37.3(−0.21); Gli37.8(−0.08)	−4.35	0.39

[a] Regression coefficients in parentheses.

U.S. winter and spring wheat cultivars, respectively, whereas Khan et al (1989) found it in only one of 44 spring wheats grown in North Dakota.

There were no correlations between A-genome-controlled subunits and baking quality in this study. Payne et al (1981b) reported a strong correlation ($r = 0.72$) between the presence of subunit 1 and sedimentation volume. We did not measure sedimentation; however, subunit 2* had higher overall quality values than subunit 1 in this study.

Several gliadin proteins were found to be correlated with baking performance in this experiment. However, the association between specific gliadin bands and quality measurements appears more complex than that between HMW glutenin subunits and quality, since there are probably thousands of permutations possible for gliadin variation and it is impossible to include all of them in one study. Moreover, because gliadins are encoded by six blocks of genes, some groups of bands with significant effects could have arisen from linked genes, or different proteins from linked genes may have opposite effects that offset each other. Determining cosegregation of gliadin blocks in this population of complex parentage was not possible. It would be very difficult to manipulate gliadin proteins in wheat breeding.

In our regression analysis, no equation using any combination of glutenins or gliadins, or both, had an R^2 above 0.50, and most R^2 values ranged from 0.35 to 0.45. These estimates are lower than those reported by Ng and Bushuk (1988), who, using only glutenins, reported R^2 values of 0.56–0.74 for various quality traits. Our values are similar to the estimates of Branlard and Dardevet (1985a, 1985b), who used glutenins or gliadins alone and reported R^2 values ranging from 0.20 to 0.54.

Our work further indicates that protein composition can explain only part of the quality variation observed in wheat flour; other proteins (such as LMW glutenins), lipids, carbohydrates, or even some minor compounds in wheat flour such as mineral content, as well as interactions among all components, may influence quality as well. Wheat quality characters are quantitative traits affected by some known genes (gliadin and glutenin loci) and many unidentified genes.

Mixograph selection is effective in quality improvement, but at least 15 g of seed is required for this test. In the F_2 and F_3 generations, very limited amounts of seed are available. SDS-PAGE of glutenins may be a reasonable tool for eliminating some progenies with undesirable glutenin subunit combinations in early-generation selection, considering that in a specific cross the progenies would have similar genetic background and HMW glutenin composition may make a difference. However, electrophoretic screening cannot remove the necessity for functional evaluation in later generations. This should be emphasized in light of the great variation in the prediction equations for quality traits and trait indices in this and other studies.

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