Intra-Varietal Polymorphism of Gliadins and Glutenins within Wheat Varieties Grown in the Czech Republic and its Impact on Grain Quality

Václav DVOŘÁČEK[1], Jana BRADOVÁ[1], Ivana CAPOUCHOVÁ[2], Anna PROHASKOVÁ[1] and Ludmila PAPOUŠKOVÁ[1]

[1]Department of Gene Bank, Crop Research Institute, Prague-Ruzyně, Czech Republic;
[2]Department of Crop Production, Faculty of Agrobiology, Food and Natural Sciences, Czech University of Life Science Prague, Prague, Czech Republic

Abstract

Dvořáček V., Bradová J., Capouchová I., Prohasková A., Papoušková L. (2013): **Intra-varietal polymorphism of gliadins and glutenins within wheat varieties grown in the Czech Republic and its impact on grain quality**. Czech J. Genet. Plant Breed., **49**: 140–148.

Using vertical electrophoresis, a set of 22 biotypes heterogeneous according to their gliadin alleles as well as their low-molecular-weight (LMW) and high-molecular-weight (HMW) glutenin subunits were identified in 10 winter wheat varieties registered in the Czech Republic. The effects of individual biotypes and their specific allelic compositions on 16 grain quality parameters were investigated. Inter-varietal differences in particular quality parameters (Zeleny sedimentation, farinograph water absorption, several values of the solvent retention capacity test) were significantly greater than the differences detected among biotypes of each variety. Special attention was given to the LMW glutenin subunits and gliadin alleles and to mutual interactions responsible for significant differences in the tested grain parameters. The results revealed at least one case of significant differences in grain quality parameters among biotypes of eight heterogeneous wheat varieties. This work unambiguously indicates that the high prevalence of wheat biotype(s) with significantly poorer values in some grain parameters can also decrease the expected technological quality of the original wheat variety. In particular, multi-line wheat varieties carrying alleles *Glu-B1* (6+8) and *Glu-B1* (7+9) or *Glu-B3j* and *Glu-B3g* can indicate the possibility of some significant changes in grain quality parameters.

Keywords: HMW- and LMW-glutenin subunits; storage proteins; technological parameters; wheat biotypes

Wheat storage proteins (i.e. gliadins, high-molecular-weight glutenin subunits (HMW-GS), and low-molecular-weight glutenin subunits (LMW-GS)) have long been used as genetic markers for identifying wheat varieties, characterizing genetic diversity, and predicting bread-making quality (Branlard *et al.* 2001; Bradová & Šašek 2005).

Allelic variation of wheat storage proteins, which directly form a part of wheat gluten, provide a basis for studying relationships between the particular gluten proteins and wheat's bread-making properties. Although protein markers have been supplanted by DNA markers in many cases and various molecular methods are regularly used as breeding tools in marker-assisted selection (MAS) systems, protein alleles remain highly effective for wheat breeding purposes as genetic markers for the prediction of technological (baking) parameters (Zheleva *et al.* 2007).

Genetic potential for dough properties has been estimated using HMW-GS composition and the *Glu-1* quality scoring system developed by Payne *et al.* (1987) and further refined by Cornish *et al.* (2005) according to the alleles' various effects on the technological parameters. No corresponding scoring system for LMW-GS and gliadins in relation to baking quality is available, and the findings of some researchers have confirmed varying effects of some LMW-GS on wheat technological parameters (Branlard *et al.* 2001; Eagles *et al.* 2004). The cumulative effect of HMW- and LMW-GS on dough quality can also differ within tested wheat genotypes. This likely is caused by their interactions with other components of their different genetic backgrounds in particular wheat varieties (Ito *et al.* 2011).

Wheat biotypes with differing gliadin and HMW-GS compositions, defined as naturally occurring variants found within a variety and likely being segregants from the original cross, have been previously detected in common wheat varieties (Mecham *et al.* 1985; Lawrence *et al.* 1987). Cornish *et al.* (2005) reported that wheat biotypes obtained from one variety are ideal materials for evaluating the effects of allelic differences on grain quality parameters. Recent studies by Vyhnánek and Bednář (2003) and Bradová and Šašek (2005) also revealed that some heterogeneous wheat varieties registered in the Czech Republic each consist of multiple protein biotypes.

Despite the detection of many wheat biotypes within cultivated wheat varieties, these are not yet regularly monitored even though they could have an essential impact on possible changes in the grain quality of a given wheat variety during its maintenance and distribution among farmers.

The aim of our research was to analyse a potential risk for decrease in technological quality in heterogeneous wheat varieties cultivated in the Czech Republic. In parallel, this research also included identifying key LMW-GS and gliadin alleles responsible for these changes while evaluating their significance in conditions of the varying genetic backgrounds of wheat varieties.

MATERIAL AND METHODS

Wheat biotype preparation. An overview of 22 evaluated wheat biotypes identified using electrophoresis from 10 winter wheat varieties heterogeneous according to their gliadin, LMW- and HMW-GS allele compositions (Bradová & Šašek 2005) are shown in Table 1. The biotypes' *Glu-1* scores are also included there. The original wheat varieties were obtained from the Czech Republic's Central Institute for Supervising and Testing in Agriculture (ÚKZÚZ) and were sown by hand onto two-row parcels in experimental fields at the Crop Research Institute (CRI) in Prague. Thirty spikes of each variety were collected from individual wheat varieties at full maturity. The electrophoretic analyses of gliadin, LMW- and HMW-GS alleles were carried out for two grains from each spike. Following the analyses, the spikes, which had identical alleles for both grains, were divided according to differing storage protein composition into individual biotypes. After that single spike progenies were cultivated in small experimental plots during two subsequent harvest years (2007 and 2008) in order to obtain the amount of seed required for field trials.

Electrophoretic methods. Gliadins and glutenins were extracted from single crushed wheat grains. Electrophoretic patterns of gliadins were determined by vertical electrophoresis in starch gel columns (Šašek & Sýkorová 1989). The method of Singh *et al.* (1991) was used for extracting LMW- and HMW-GS, and their electrophoretic patterns were determined using SDS-PAGE (Laemmli 1970). The allelic gliadin blocks of bands were separated from the electrophoretic patterns of gliadins according to a previously published method (Šašek & Sýkorová 1989). LMW- and HMW-GS were identified by comparison with published references (Payne & Lawrence 1983; Jackson *et al.* 1996).

Field experiments. The multiplied wheat biotypes were cultivated at the Uhříněves Experimental Station affiliated with the University of Life Sciences in Prague during 2009–2011. Plots (10 m^2) were arranged in a randomized plot design in two (2009) and three (2010, 2011) replications. The treatment of experimental plots was carried out according to standard agronomic procedures appropriate for winter wheat.

Grain quality characteristics. Approximately 1500 g of grains from each replication were sampled. A grain mixture from each individual wheat biotype was prepared and then used for the subsequent grain analyses. The wheat quality parameters were defined as yield of grain (YG), thousand grain weight (TGW), test weight (TW), crude protein content (CP) (ČSN EN ISO 5983-1 2005),

Table 1. Allelic composition of gliadin, high-molecular-weight glutenin subunits (HMW-GS) and low-molecular-weight glutenin subunits (LMW-GS) of evaluated wheat biotypes

Variety/ Category of BMQ	Biotype designation	Gliadins							LMW-GS			HMW-GS			*Glu-1* score
		Gld-1-1A	*Gld-2-1A*	*Gld-1B*	*Gld-1D*	*Gld-6A*	*Gld-6B*	*Gld-6D*	*Glu-A3*	*Glu-B3*	*Glu-D3*	*Glu-A1*	*Glu-B1*	*Glu-D1*	
Asta/B	*A*	4	0	4	1	2	2	19	*a*	*f*	*c*	0	7+9	5+10	8
	B	4	0	4	1	2	2	19	*a*	*f*	*c*	0	6+8	5+10	6
Astella/B	*A*	2	0	1	2	1	1	1	*d*	*b*	*c*	2*	7+9	5+10	10
	B	2	0	1	2	1	1	4	*d*	*b*	*c*	2*	7+9	5+10	10
Ilona/E	*A*	3	0	1	13	1	1	6	*e/f*	*b*	*b*	0	7+9	5+10	8
	B	12	0	1	13	1	1	6	*e/f*	*b*	*b*	0	7+9	5+10	8
Karolinum/A	*A*	9	0	3	1	3	1	2	*a*	*j*	*c*	1	17+18	5+10	10
	B	9	0	4	1	3	1	4	*a*	*g*	*c*	1	17+18	5+10	10
Meritto/B	*A*	4	0	4	1	2	1	1	*a*	*g*	*c*	0	6+8	2+12	4
	B	4	0	4	1	2	4	1	*a*	*g*	*c*	0	6+8	2+12	4
Mladka/C	*A*	4	0	1	9	3	1	2	*e/f*	*b*	*c*	0	7+9	2+12	6
	B	4	0	4	9	3	1	2	*e/f*	*f*	*c*	0	7+9	2+12	6
Niagara/A	*A*	2	0	1	1	1	1	1	*d*	*b*	*c*	0	7+9	5+10	8
	B	2	0	4	1	1	1	1	*d*	*g*	*c*	0	7+9	5+10	8
	C	2	0	4	1	1	1	9	*d*	*g*	*c*	0	7+9	5+10	8
Sepstra/C	*A*	9	0	4	1	2	2	1	*a*	*g*	*c*	0	7+9	2+12	6
	B	9	3	4	1	2	2	1	*a*	*g*	*c*	0	7+9	2+12	6
Solara/B	*A*	2	0	1	1	1	3	1	*d*	*b*	*c*	0	7+9	5+10	8
	B	2	0	1	3	1	3	1	*d*	*b*	*a*	0	7+9	5+10	8
Windsor/C	*A*	2	0	4	1	3	12	5	*d*	*f*	*c*	0	6+8	2+12	4
	B	2	0	3	1	3	12	5	*d*	*j*	*c*	0	6+8	2+12	4
	C	9	0	4	1	3	12	5	*a*	*f*	*c*	0	6+8	2+12	4

BMQ – bread-making quality as declared by the Czech Republic's Central Institute for Supervising and Testing in Agriculture (ÚKZÚZ) at the time of variety registration; E – elite to very good, improving; A – high-quality to good, separately workable; B – bread-making to additive, used in blends; C – unsuitable for yeast dough production

falling number (FN) (ICC Standard No. 107/1:1995), wet gluten content (WG), gluten index (GI) (ICC Standard No. 155:1994) and Zeleny sedimentation (ZS) (ČSN ISO 5529:2000). The dough rheological properties were examined by Brabender farinograph and included water absorption (WA), dough development time (DDt), dough stability (DS) and degree of softening (DeS) (AACC 54-21). The solvent retention capacity (SRC) test was used to assess water retention (SRC_w), 5% lactic acid (SRC_l), 50% sucrose (SRC_s), and 5% sodium carbonate (SRC_c) (AACC 56-11:2000). Grain analyses were carried out in two replications.

Statistical methods. Basic descriptive statistics (mean, minimum, maximum and standard deviation) as well as analysis of variance (Main Effects ANOVA) with genotype and environment (year) as fixed factors, including subsequent Tukey's HSD test, were calculated using Statistica 7.0 CZ statistical software (StatSoft, Tulsa, USA). Effects of genotype (G) and environment (E) were expressed as percentages of total sums of squares in accordance with Gomez-Becerra *et al.* (2010). A dendrogram of distances among biotypes was constructed on the basis of a prepared binary matrix describing the presence or absence of the specific allele within all known combinations. The linkage rule (unweighted pair group method with arithmetic mean (UPGMA)) and Euclidean distances as a distance measure were applied in constructing the final dendrogram using the Statistica 7.0 software.

RESULTS AND DISCUSSION

This study confirmed the highest allelic variability at gliadin loci compared to variability of alleles encoding HMW- and LMW-GS in the investigated set of 22 wheat biotypes (Table 1). In total, 30 gliadin, 10 LMW-GS, and 8 HMW-GS alleles were identified. The gliadin allele *Gld 2-1A* (0) showed the highest frequency. In the cases of HMW-GS and LMW-GS, respectively, the highest occurrences were found for *Glu-A1* (0) and *Glu-D3c*. Some alleles were rare. Found in only one biotype each were *Gld-1-1A* (3), *Gld-1-1A* (12), *Gld-2-1A* (3), *Gld-1D* (3), *Gld-6B* (4), *Gld-6D* (9) and *Glu-D3a*. The allelic profiles of HMW-GS, LMW-GS and gliadins enabled us unambiguously to distinguish all biotypes from one another (Table 1, Figure 1).

Three-year average values for the evaluated wheat grain quality parameters are shown in Table 2. The detected ranges of tested parameters were generally affected by both genotypic (biotype) and environmental (growing season) factors. Strong dependence on environmental factors was confirmed in thousand grain weight (TGW), yield of grain (YG), crude protein content (CP), falling number (FN) and degree of softening (DeS), where the percentage effects of growing conditions on the parameter variability exceeded 74%. Lower average values with high annual variations were found especially in FN and DeS. This was mainly caused by adverse weather conditions when wheat was at full maturity

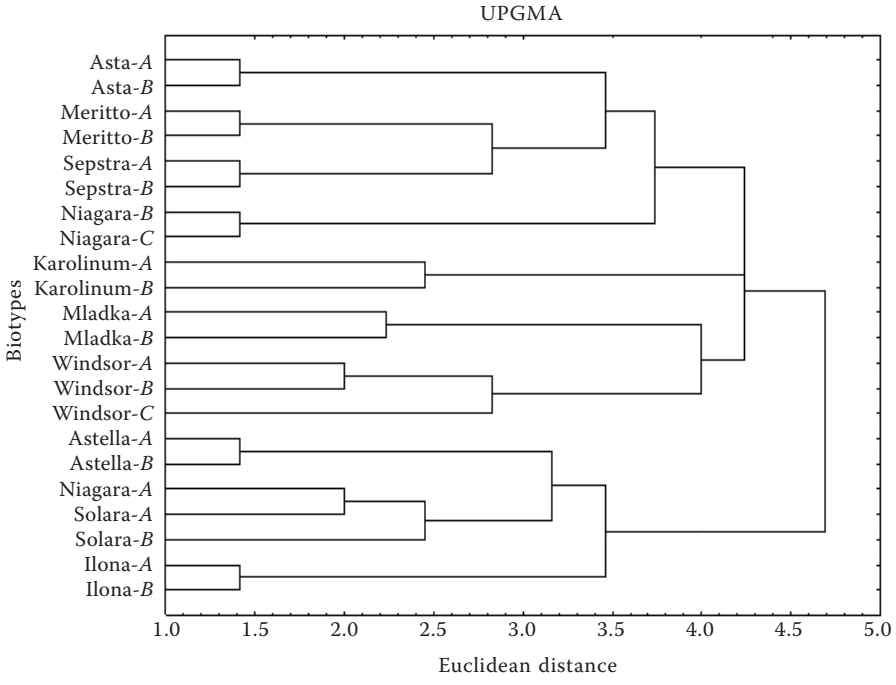

Figure 1. Dendrogram of 22 wheat biotypes based on genetic similarity calculated from data of gliadins, low-molecular-weight glutenin subunits (LMW-GS) and high-molecular-weight glutenin subunits (HMW-GS) using the unweighted pair group method with arithmetic mean (UPGMA) clustering method

Table 2. Variability of grain quality parameters in evaluated wheat biotypes (2009–2011)

Parameters	Mean	Min	Max	SD	Effect of genotype	Effect of environment
					(%)	
Thousand grain weight (TGW, g)	43.58	40.02	49.49	6.60	16.57**	74.65**
Yield of grain (YG, t/ha)	8.03	7.27	8.92	1.61	ns	77.15**
Test weight (TW, kg/hl)	72.55	68.62	75.67	4.27	18.64**	69.40**
Crude protein content (CP, %)	12.44	12.00	13.60	1.23	10.05*	80.98**
Wet gluten content (WG, %)	28.99	25.58	34.25	3.22	ns	20.25**
Gluten index (GI)	80.67	54.67	98.67	15.86	81.73**	ns
Zeleny sedimentation (ZS, ml)	34.96	22.27	50.22	8.62	64.58**	23.54**
Falling number (FN, s)	198.30	88.83	256.67	114.32	13.90**	74.86**
Water absorption (WA, %)	51.73	45.47	55.47	2.78	75.73**	ns
Dough development time (DDt, min)	1.85	1.33	2.50	0.51	52.90**	ns
Dough stability (DS, min)	4.06	2.00	5.67	1.52	62.29**	ns
Degree of softening (DeS, BU)	143.33	121.67	186.67	86.00	ns	83.79**
Solvent retention capacity for water (SRC_w, %)	59.85	53.20	66.97	5.89	59.65**	29.82**
Solvent retention capacity for lactic acid (SRC_l, %)	123.39	100.37	145.63	17.26	51.82**	37.31**
Solvent retention capacity for sucrose (SRC_s, %)	104.84	96.17	115.43	8.31	57.33**	22.14**
Solvent retention capacity for sodium carbonate (SRC_c, %)	79.58	68.70	92.93	8.62	55.88**	31.04**

SD – standard deviation; *significant at level $P \leq 0.05$; **significant at level $P \leq 0.01$; ns – not significant

in 2010 (more than 100 mm of precipitation in a single week), which postponed the optimal harvest time by about a fortnight. The weather conditions in 2010 influenced all grain parameters negatively, and especially YG, TGW and TW.

In spite of the strong pressure of environmental factors, significant genetic dependence on parameters GI, ZS, water absorption (WA), dough development time (DDt) and dough stability (DS) was confirmed. The mutual effects of both factor types – albeit with a predominating genotypic influence – were found in the SRC test parameters (Table 2). Significant correlations among WA, SRC_w, SRC_c and hardness of grains mentioned by Morris *et al.* (2013) probably indicate that our found differences in these parameters could also be caused by different grain hardness among tested wheat biotypes. Nevertheless, this parameter was not assessed because grain hardness is still not accepted as a standard trait for prediction of bread-making quality in the Czech Republic.

Inter-varietal differences of particular quality parameters (e.g. ZS, WA, SRC_1 and SRC_w) were significantly greater than detected differences among biotypes of each variety. This corresponds to the higher variability of gliadin and glutenin alleles among different varieties than among derived biotypes of an individual variety (Figure 1).

The detected spectrum of parameters with significant differences among biotypes of a single variety was not wide. Nevertheless, the results revealed at least one case of significant differences in grain quality parameters among the biotypes of eight heterogeneous wheat varieties (Table 3). In all biotypes selected from varieties with the highest declared bread-making quality (Karolinum, Ilona and Niagara), several significant differences were detected in the tested parameters. Farinograph dough stability (DS) showed a significantly higher value (5.50 min) in the Ilona-*A* biotype with the *Gld 1-1A* (3) allele compared to Ilona-*B* (4.67 min) containing the *Gld 1-1A* (12) allele. Significantly lower values of ZS, CP and SRC_1 were detected in the Karolinum-*A* biotype with *Gld-1B* (3), *Gld-6D* (2) and *Glu-B3j* alleles, whereas Karolinum-*B* contained *Gld-1B* (4), *Gld-6D* (4) and *Glu-B3g* alleles. In the case of Niagara biotypes, significant differences were confirmed in the parameters DDt and SRC_c (Table 3). In these cases, a possible quality reduction can mean significant economic losses, especially for wheat growers and seed producers. Cornish *et al.* (2005) had also confirmed that a prevalence of one biotype in multi-

line wheat varieties can cause significant changes in technological parameters that will not correspond with the declared parameters of the original variety.

The significant variation of technological parameters found in biotypes derived from wheat varieties with lower bread-making quality, such as Mladka

Table 3. Documented significant differences in values of grain quality parameters among wheat biotypes (2009–2011)

Wheat biotype	Allele differences	Parameters
Asta-*A*	*Glu-B1* (7+9)	GI = 95.33[a]
Asta-*B*	*Glu-B1* (6+8)	GI = 89.33[b]
Astella-*A*	*Gld-6D* (1)	SRC_w = 54.00[a]
Astella-*B*	*Gld-6D* (4)	SRC_w = 55.43[b]
Ilona-*A*	*Gld-1-1A* (3)	DS = 5.50[a]
Ilona-*B*	*Gld 1-1A* (12)	DS = 4.67[b]
Karolinum-*A*	*Gld-1B* (3); *Gld-6D* (2); *Glu-B3j*	CP = 12.37[a] ZS = 33.40[a] SRC_l = 112.40[a]
Karolinum-*B*	*Gld-1B* (4); *Gld-6D* (4); *Glu-B3g*	CP = 12.92[b] ZS = 41.77[b] SRC_l = 127.93[b]
Mladka-*A*	*Gld-1B* (1); *Glu-B3b*	GI = 85.33[a] SRC_w = 59.47[a] SRC_l= 132.50[a] SRC_s = 104.63[a]
Mladka-*B*	*Gld-1B* (4); *Glu-B3f*	GI = 69.67[b] SRC_w = 55.50[b] SRC_l = 124.53[b] SRC_s = 98.53[b]
Niagara-*A*	*Gld-1B* (1); *Gld-6D* (1); *Glu-B3b*	DDt = 1.83[a] SRC_c = 82.77[ab]
Niagara-*B*	*Gld-1B* (4); *Gld-6D* (1); *Glu-B3g*	DDt = 2.50[b] SRC_c = 85.23[b]
Niagara-*C*	*Gld-1B* (4); *Gld-6D* (9); *Glu-B3g*	DDt = 1.83[a] SRC_c = 81.10[a]
Sepstra-*A*	*Gld-2-1A* (0)	ZS = 42.95[b] DS = 4.83[b] SRC_l = 123.60[b]
Sepstra-*B*	*Gld-2-1A* (3)	ZS = 34.57[a] DS = 3.67[a] SRC_l = 112.07[a]
Windsor-*A*	*Gld-1-1A* (2); *Gld-1B* (4); *Glu-A3d*; *Glu-B3f*	SRC_l = 116.33[b]
Windsor-*B*	*Gld-1-1A* (2); *Gld-1B* (3); *Glu-A3d*; *Glu-B3j*	SRC_l = 100.37[a]
Windsor-*C*	*Gld-1-1A* (9); *Gld-1B* (4); *Glu-A3a*; *Glu-B3f*	SRC_l = 103.20[a]

GI – gluten index; CP – crude protein content (%); ZS – Zeleny sedimentation (ml); DDt – dough development time (min); DS – dough stability (min); SRC_w – solvent retention capacity test for water (%); SRC_l – solvent retention capacity test for lactic acid (%); SRC_s – solvent retention capacity test for sucrose (%); SRC_c – solvent retention capacity test for sodium carbonate (%); significantly different values (Tukey's HSD test, $P \leq 0.05$) in the same parameters of related biotypes are marked with various letters

(GI, SRC_w, SRC_l and SRC_s) and Sepstra (ZS, DS and SRC_l) should not play an important economic role as they are predominantly used for livestock feeding purposes and due to the absence of official technological criteria (Dvořáček *et al.* 2008). Such varieties may also be used for pasta and biscuit production, where demands on technological quality are not as strict as they are for bread production (Bushuk 1998).

Potential decrease of wheat technological quality will be associated with very high prevalence of those biotype(s) having unsuitable compositions of glutenin and gliadin alleles. With appropriate variety maintenance and seed propagation, the risk for changes of biotype participation within a given variety does not appear to be very high. However, this possibility likely did occur in the wheat variety Karolinum, which was reclassified after the three-year registration from the A to the B baking class due to a gradual deterioration in its bread-making parameters (Horáková *et al.* 2005, 2006).

In our study, it is possible to divide the detected allelic effects on changes of technological parameters into three categories. The first category includes an effect of an individual allele the incidence of which significantly decreases or increases values of some technological parameters. In accordance with Branlard *et al.* (2001), these were *Gld-1B* (3) and *Glu-B3j* alleles, which were identified in biotypes derived from Karolinum and Windsor varieties. According to Jackson *et al.* (1996), these are connected with occurrence of the rye translocation 1BL/1RS and their incidence showed unfavourable effects on ZS and SRC_l. A positive effect on the values of GI parameters was confirmed for the *Glu-B1* (7+9) allele only in Asta biotypes (Table 3). This allele has long been known to have an effect of improving baking parameters (Payne *et al.* 1987; Shewry *et al.* 2001).

The next category includes mutual interactions of individual alleles and the genetic background of each biotype and which caused significant parameter differences. These cases were observed in the presence of alleles *Gld-1B* (4), *Glu-B3f* and *Glu-B3g*. The allele *Gld-1B* (4) in combination with *Gld-6D* (4) and *Glu-B3g* in the Karolinum-*B* biotype was associated with significantly higher values of ZS (41.8 ml) and SRC_l (127.9%). On the other hand, the presence of *Gld-1B* (4*)* together with *Glu-B3f* was associated with reduction in the value in Mladka-*B*, for example, of GI (69.7) and SRC_l (124.5%). Two Windsor biotypes (*A* and *C*) with *Gld-1B* (4) and *Glu-B3f* alleles also showed significantly different values of SRC_l in relation to the interaction with *Gld 1-1A* (2) and *Glu-A3d* or *Gld 1-1A* (9) and *Glu-A3a* alleles (Table 3). The results obtained support possibilities for drawing mutually controversial conclusions as to the effects of some individual LMW-GS and gliadin alleles on rheological and gluten properties as published by Branlard *et al.* (2001), Cornish *et al.* (2001), Eagles *et al.* (2004) and Tsenov e*t al.* (2009).

Other mutual interactions of LMW-GS on *Glu-A3* and *Glu-B3* loci with significant impacts on dough properties as noted by Ito *et al.* (2011) can be partially documented by our model example. Table 4 shows eight selected biotypes derived from four different varieties with identical HMW-GS composition of *Glu-A1null*, *Glu-B1* (7+9) and *Glu-D1* (5+10) plus the identical *Glu-1* score of 8 (Table 1). This should indicate a similar level of technological parameters. We nevertheless found significant differences in seven technological parameters, with relative percentage differences ranging from 7% to 47% for WA and ZS values, respectively. In the case of ZS, the maximum detected differences in the range of 32–50 ml (biotype Solara-*A* vs. Niagara-*C*) even included three categories of wheat baking quality (E, A and B) as declared by ÚKZÚZ (2012). These interactions among LMW-GS and gliadin alleles plus the specific genetic backgrounds of the varieties (expressed as effect of genotype) significantly influenced the final variability of the aforementioned parameters in the range 45–74% (Table 4). Considering the identical composition of HMW-GS in our wheat biotypes, these values were higher compared to the results of Branlard *et al.* (2001), who reported in a large number of different wheat genotypes the percentage effect of LMW-GS and gliadin alleles on Zeleny sedimentation and rheological parameters in the range of 18–33%.

The final category of allelic effects where genetic background probably played the main role was detected in comparing the two biotypes Solara-*A* and Niagara-*A*. They had identical composition of HMW- and LMW-GS but significant differences in ZS, WA, SRC_s and SRC_c. The significantly higher values of ZS, WA, SRC_s and SRC_c in the Niagara-*A* biotype were probably caused not only by the one allelic difference in gliadins (*Gld*-6B (1) in Niagara-*A* and *Gld*-6B (3) in Solara-*A*), but mainly by mutual interactions of all allelic systems with the specific genetic backgrounds of the particular varieties.

In conclusion, it is appropriate to emphasize that significant changes in grain's technological param-

Table 4. Detected significantly different grain quality parameters in wheat biotypes with identical high-molecular-weight glutenin subunits (HMW-GS) composition (0; 7+9; 5+10) (2009–2011)

Biotype	ZS (ml)	GI	WA	SRC_w	SRC_l	SRC_s	SRC_c
			(%)				
Asta-A	34.90^{a}	95.33^{ab}	53.90^{ab}	63.47^{a}	108.97^{e}	107.03^{ab}	83.50^{b}
Ilona-A	36.60^{a}	91.33^{ab}	54.20^{ab}	60.70^{a}	124.03^{a}	104.07^{abc}	77.37^{a}
Ilona-B	34.99^{a}	90.67^{a}	54.63^{b}	61.57^{a}	127.50^{ab}	103.90^{ac}	79.47^{ab}
Niagara-A	45.08^{b}	97.33^{a}	52.70^{ab}	60.17^{ac}	141.90^{cd}	112.30^{ab}	82.77^{ab}
Niagara-B	47.07^{b}	98.67^{a}	52.97^{ab}	62.53^{a}	145.63^{d}	114.10^{b}	85.23^{b}
Niagara-C	50.22^{b}	98.00^{ab}	52.37^{ab}	61.57^{a}	141.63^{bcd}	109.77^{ab}	81.10^{a}
Solara-A	31.69^{a}	97.67^{ab}	49.93^{c}	54.87^{bc}	130.37^{abc}	96.40^{c}	68.70^{c}
Solara-B	34.10^{a}	84.00^{b}	48.93^{c}	53.43^{b}	129.73^{abc}	96.37^{c}	70.60^{c}
Effect of genotype (%)	56.42**	48.18*	74.11**	45.46*	48.95**	48.72*	60.55**
Effect of environment (%)	27.50**	27.04**	ns	31.66**	36.05**	26.75**	27.12**

Values of parameters marked by different letters are significantly different at $P \leq 0.05$ (Tukey's HSD test); *significant at level $P \leq 0.05$; **significant at level $P \leq 0.01$; ns – not significant; WA – water absorption; SRC_w – solvent retention capacity test for water; SRC_l – solvent retention capacity test for lactic acid; SRC_s – solvent retention capacity test for sucrose; SRC_c – solvent retention capacity test for sodium carbonate

eters can occur in wheat varieties composed of multiple biotypes. Greater risk can be expected in wheat varieties composed of wheat biotypes carrying different alleles with contrasting effect on technological parameters (e.g. *Glu-B1* (6+8) vs. *Glu-B1* (7+9); *Glu-B3j* vs. *Glu-B3g*). The results of this study also confirm a substantial contribution from other allelic groups (LMW-GS and gliadins) to the estimation of technological quality in wheat varieties and their specific interaction with HMW-GS and the genetic backgrounds of particular wheat varieties in some cases. It is also necessary to take into account that complex electrophoretic evaluation of protein alleles can contribute not only to technological stability of the registered wheat varieties during their maintenance, but it also can be an appropriate tool for use in preserving varietal authenticity in the context of providing legal protection for wheat varieties.

Acknowledgements. This work was supported by the Ministry of Agriculture of the Czech Republic, Projects No. 0002700604 and QJ1310219.

References

AACC 54-21 (1999): Farinograph Method for Flour. Approved Methods of Analysis. Vol. 2, American Association of Cereal Chemists, St. Paul, 1–7.

AACC 56-11 (2000): Solvent Retention Capacity Profile. Approved Methods of Analysis. Vol. 2, American Association of Cereal Chemists, St. Paul, 1–2.

Bradová J., Šašek A. (2005): Diversity of gliadins and HMW glutenin subunits in Czech registered wheat varieties. Czech Journal of Genetics and Plant Breeding, **41**: 160–163.

Branlard G., Dardevet M., Saccomano R., Lagoutte F., Gourdon J. (2001): Genetic diversity of wheat storage proteins and bread wheat quality. Euphytica, **119**: 59–67.

Bushuk W. (1998): Wheat breeding for end-product use. Euphytica, **100**: 137–145.

Cornish G.B., Békés F., Eagles H.A., Payne P.I. (2005): Prediction of dough properties for bread wheats. In: Gliadin and Glutenin: The Unique Balance of Wheat Quality. American Association of Cereal Chemists, St. Paul, 243–280.

ČSN ISO 5529 (2000): Wheat – Assessment of Sedimentation Index – Zeleny test. Czech Standards Institute, Prague, 1–9. (in Czech)

ČSN EN ISO 5983-1 (2005): Animal Feeding Stuffs – Determination of Nitrogen Content and Calculation of Crude Protein – Part 1: Kjeldahl Method. Czech Standards Institute, Prague, 1–10.

Dvořáček V., Kodeš A., Stehno Z., Hučko B., Mudřík Z. (2008): Nutritive effect of protein composition and other grain properties of doubled haploid wheat lines with/without translocation 1B/1R in a model feeding test. Czech Journal of Animal Sciences, **53**: 487–498.

Eagles H.A., Eastwood R.F., Hollamby G.J., Martin E.M., Cornish G.B. (2004): Revision of the estimated of

glutenin gene effects at the *Glu-B1* locus from southern Australian wheat breeding programs. Australian Journal of Agricultural Research, **55**: 1093–1096.

Gomez-Becerra H.F., Erdem H., Yazici A., Tutus Y., Torun B., Ozturk L., Cakmak I. (2010): Grain concentrations of protein and mineral nutrients in a large collection of spelt wheat grown under different environments. Journal of Cereal Science, **52**: 342–349.

Horáková V., Beneš F., Mezlík T. (2005): Classification of varieties into categories baking quality. In: List of Varieties 2005. Central Institute for Supervising and Testing in Agriculture, Brno, 24–35. (in Czech)

Horáková V., Beneš F., Mezlík T. (2006): Classification of varieties into categories baking quality. In: Recommended List of Varieties. Central Institute for Supervising and Testing in Agriculture, Brno, 22–33. (in Czech)

ICC Standard No. 107/1 (1995): Determination of the "Falling Number" According to Hagberg – as a Measure of the Degree of Alpha-Amylase Activity in Grain and Flour. International Association for Cereal Science and Technology, Vienna.

ICC Standard No. 155 (1994): Determination of Wet Gluten Quantity and Quality (Gluten Index ac. to Perten) of Whole Wheat Meal and Wheat Flour (*Triticum aestivum*). International Association for Cereal Science and Technology, Vienna.

Ito M., Fushie S., Maruyama-Funatsuki W., Ikeda T.M., Nishio Z., Nagasawa K., Tabiki T., Yamauchi H. (2011): Effect of allelic variation in three glutenin loci on dough properties and breadmaking qualities of winter wheat. Breeding Science, **61**: 281–287.

Jackson E.A., Morel M.H., Sontag-Strohm T, Branlard G., Metakovsky E.V., Redaelli R. (1996): Proposal for combining the classification systems of alleles of *Gli-1* and *Glu-3* loci in bread wheat. Journal of Genetics and Breeding, **50**: 321–336.

Laemmli V.K. (1970): Cleavage of structural proteins during assembly of the head bacteriophag. Nature, **227**: 680–685.

Lawrence G.J., Moss H.J., Shepherd K.W., Wrigley C.W. (1987): Dough quality of biotypes of eleven Australian wheat cultivars that differ in high-molecular-weight glutenin subunits composition. Journal of Cereal Science, **6**: 99–101.

Mecham D.K., Kasarda D.D., Qualset C.O. (1985): Identification of U.S. western wheat varieties by polyacrylamid gel electrophoresis of gliadin proteins. Hilgardia, **53**: 1–32.

Morris C.F., Anderson J.A., King G.E., Bettge A.D., Garland-Campbell K., Allan R.E., Fuerst E.P, Beecher B.S. (2013): Characterization of a Unique "Super Soft" Kernel Trait in Wheat. Cereal Chemistry, **90**: 47–57.

Payne P.I., Lawrence G.J. (1983): Catalogue of alleles or the complex loci, *Glu-A1*, *Glu-B1* and *Glu-D1* which coded for high-molecular-weight subunits of glutenin in hexaploid wheat. Cereal Research Communications, **11**: 29–35.

Payne P.I., Nightingale M.A., Krattiger A.F., Holt L.M. (1987): The relationship between HMW glutenin subunit composition and the bread making quality of British-grown wheat varieties. Journal Science Food and Agriculture, **40**: 51–65.

Šašek A., Sýkorová S. (1989): Standardisation of vertical electrophoresis in starch gel columns and characterization of gliadin allelic blocks. Scientia Agriculturae Bohemica, **21**: 99–108.

Shewry P.R., Tatham A.S., Fido R., Jones H., Barcelo P., Lazzeri P.A. (2001): Improving the end use properties of wheat by manipulating the grain protein composition. Euphytica, **119**: 45–48.

Singh N.K., Shepherd K.W., Cornish G.B. (1991): A simplified SDS PAGE procedure for separating LMW subunits of glutenin. Journal of Cereal Science, **14**: 105–109.

Tsenov N., Atanasova D., Todorov I., Ivanova I., Stoeva I. (2009): Allelic diversity in Bulgarian winter wheat varieties based on polymorphism of glutenin subunit composition. Cereal Research Communications, **37**: 551–558.

ÚKZÚZ (2012): Recommended List of Varieties. Available at http://www.ukzuz.cz/Articles/34146-2-Recommended+List+of+Varieties+(SDO)+.aspx (accessed September 7, 2012)

Vyhnánek T, Bednář J. (2003): Detection of the varietal purity in sample of harvested wheat and triticale grains by prolamin marker. Plant, Soil and Environment, **49**: 95–98.

Zheleva D., Todorovska E., Christov N., Ivanov P., Ivanova I., Todorov I. (2007): Assessing the genetic variation of Bulgarian bread wheat varieties by biochemical and molecular markers. Biotechnology and Biotechnological Equipment, **21**: 311–321.

Received for publication November 27, 2012
Accepted after corrections September 11, 2013

Corresponding author:

Ing. Jana Bradová, Výzkumný ústav rostlinné výroby, v.v.i., Oddělení genové banky, Drnovská 507, 161 06 Praha 6-Ruzyně, Česká republika
e-mail: bradova@vurv.cz