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COMPOSITION OF GLUTEN PROTEINS AND QUALITY PARAMETERS OF WINTER TRITICALE HYBRIDS

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Key words: triticale hybrid strains, electrophoresis of gliadins and glutenins, content of protein, content of soluble pentosans, falling number

The HMW glutenin subunits (SDS-PAGE) and gliadin subunits (A-PAGE) were analysed in selected triticale hybrids and their parental forms. In order to enable further selection of new high-quality hybrid strains of winter triticale determinations were also carried out for protein content, falling number and soluble pentosans level. Among the identified glutenin subunits coded by loci *Glu A1* both in the parental forms and in hybrids the qualitatively advantageous subunit 2* usually occurred. Glutenin subunits coded by *Glu-B1*: 7+25, 7+8, 6+8 for hybrids strains: IGS 5101 x LAD 122, FDT 975 x LAD 122 and Alzo x F 8063 were consistent with maternal forms. In the Alzo x F 8063 strains the additional subunits not detected in parental forms were identified as well. Electrophoretic analysis of gliadin proteins showed intralinear polymorphism of these proteins.

The studies pointed to parental genotypes as the most valuable ones because of the quality of grain of hybrids: the maternal FDT 975 and paternal LAD 122. The hybrid strain FDT 975 x LAD 122 was characterised by presence of qualitatively good glutenin subunits, the highest falling number, and a higher content of soluble pentosans in the grain.

INTRODUCTION

Triticale, the corn whose harvest potential and grain quality come from wheat, and resistance to pathogens from rye [Tohver *et al*., 2005], can be used as feed or consumer grains. Flour obtained from some varieties of triticale may be an alternative to wheat flour for baking cookies or crackers or, under proper conditions of making the dough (low speed of stirring, shortened time of fermentation) it may be used for baking bread that meets quality requirements [Perez *et al*., 2003; Varughese *et al*., 1996]. Quantity and quality of gluten proteins is largely responsible for baking properties of a grain. Proteins are characterised by the mode of inheritance that is independent from the environmental conditions. Allelic variants of genes coding high-molecular-weight glutenin subunits are particularly important for defining elasticity of wheat gluten and of dough. In hexaploid wheats subunits of HMW glutenins are coded by genes localized on chromosomes 1A, 1B, 1D, and in hexaploid triticale on chromosomes 1A, 1B and 1R. Triticale will be characterised by a higher bread-making quality, having valuable glutenin subunits coded by genes coming from wheat. Secalins from rye make the baking quality of triticale lower, causing deterioration in rheological properties and total gluten strength and an increase in dough viscosity [Tohver *et al.*, 2005; Payne *et al.*, 1987; Makarska, 2000].

In the case of triticale a considerable tendency of this species of grain to preharvest sprouting is still a problem; the tendency being connected with a high activity of α-amylase in the grain. Germinating also has an unfavorable influence on the baking value, *i.e.* it results in a decrease in water absorption, resistance of the dough to kneading, and an increase in soaking [Dojczew *et al.,* 2004]. Breeders try to solve the problem of vulnerability to germination of triticale grains by using the methods of selection of better varieties and strains that are more resistant to germination. At least two regulator genes can influence the level of transcription of structural genes of α-amylase: the activator that is controlled by gibberellin and the inhibiting gene controlled by abscisic acid [Masojć, 1997].

Usefulness of cereal for baking is influenced by the content of various components that sometimes are present in the grain in small quantities, including pentosans (arabinoxylans), both those soluble and insoluble in water. During the kneading they are responsible for water absorption in the dough, and hence for the increase in wetness of the bread and the structure of its crumb [Michniewicz *et al*., 1992; Michniewicz, 1995].

The aim of the study was to evaluate quality parameters as well as identify and characterize gluten protein of new winter triticale hybrid strains and parental components in order to enable the selection of the best strains and varieties.

MATERIALS AND METHODS

Kernels of 5 hybrid strains of winter triticale and their parental forms morphologically established and selected at the Institute of Genetics, Plant Breeding and Biotechnology of the Agricultural University in Lublin were the material of the study (Table 1). Straight strains with good crops were chosen for qualitative investigations.

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TABLE 1. Material of study.

Strains	variety
Maternal form	IGS 5101
Maternal form	FDT 975
Maternal form	Alzo
Paternal form	LAD 122
Paternal form	F 8063
Hybrid	IGS 5101 × LAD 122
Hybrid	IGS 5101 × F 8063
Hybrid	FDT 975 × LAD 122
Hybrid	Alzo × LAD 122
Hybrid	Alzo × F 8063

In order to identify gluten subunits two wheat varieties, Zebra and Clever, were used as standards.

Polymorphism of glutenins was analysed on the basis of electrophoretic separation in alkaline medium with the addition of sodium dodecyl sulphate (SDS-PAGE) according to Brzeziński & Łukaszewski [1998]. Characterization of gliadin and secalin fractions was conducted with the method of acid electrophoresis (A-PAGE) according to Brzeziński *et al*. [1989]. Separated proteins were dyed with Coomassie Blue R-250. Protein content was determined with the Kjeldahl method on a Kjel-Foss apparatus. The falling number was determined according to Polish Standard [PN-ISO: 3093, 1996]. Extraction and the level of soluble pentosans were determined by means of the Hashimoto *et al.* [1987] method. Data were compared using statistical analysis of the Tukey test.

RESULTS AND DISCUSSION

Plants which are characterised by morphological equalization, good crops and quality already in early generations may give rise to a new variety.

One of the most important markers of the bread-making quality is the quantity and quality of certain gluten proteins [Gianibelli *et al*., 2001; Payne *et al*., 1987; Waga, 1997].

Among the subunits of high-molecular-weight glutenins coded by loci *Glu A1* (derived from wheat) both in the parental forms and in hybrids the qualitatively advantageous subunit 2* usually occurred (Figure 1, Table 2). Quantitatively it predominated its allelic variants from the chromosome 1A, *i.e.* Glu A1-1 and Glu A1-null. The maternal form IGS 5101 coding the subunit 1 was an exception. In the hybrid strains derived from this maternal form the Glu A1-null subunit was found to occur. The presence of glutenins coded in the so-called uncoding variant of the null type is often identified in wheat varieties and strains and is an unfavorable phenomenon for its technological quality [Gianibelli *et al*., 2001; Waga, 1997; Ciołek & Makarska, 2004; Makarska & Szwed-Urbaś, 2005]. A greater polymorphism in the studied strains was found for glutenin fractions determined by the 1B chromosome with five allelic forms on each locus Glu B1. The analysed maternal forms were identified to contain subunits coded by *Glu B1*: 7+25, 7+8 and 6+8, whereas in the paternal forms: 7+9 and 7+26 (Figure 1). The variability of subunits coded by loci

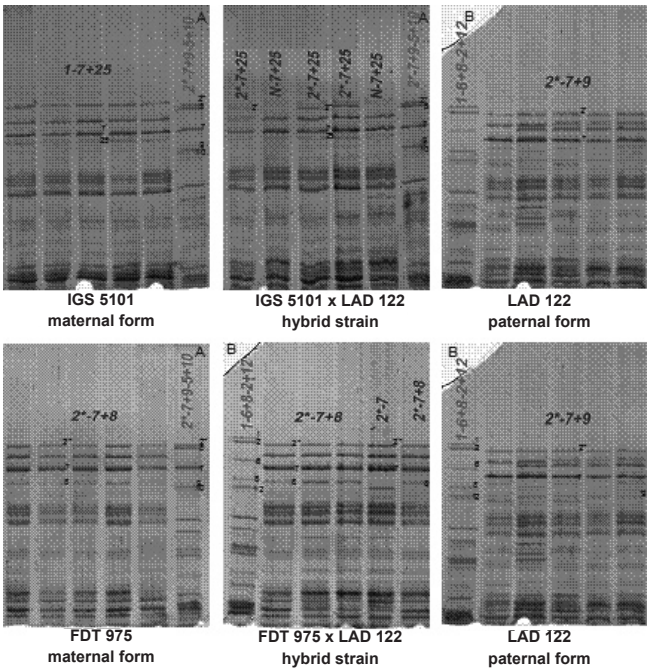

FIGURE 1. Electrophoregrams (SDS-PAGE) of glutenin subunits from hybrid strains and parental components of winter triticale (hybrid strains 5101 x LAD 122 and parental components, hybrid strains FDT 975 x LAD 122 and parental components) and from standard cultivars of wheat: (A) Zebra, (B) Clever.

TABLE 2. Composition of high-molecular-weight glutenin subunits of triticale strains and standard wheat cultivars.

Strain/cultivar		Composition of subunits coded by loci		
		Glu A1	*Glu* B1	*Glu* D1
Maternal forms	IGS 5101	1	7+25	-
	FDT 975	2*	7+8	-
	Alzo	2*	6+8	-
Patertal forms	LAD 122	2*	7+9	-
	F 8063	2*	7+26 (7+8)	-
Hybrids	IGS 5101 × LAD 122	2*, N	7+25	-
	IGS 5101 × F 8063	N	7+9	-
	FDT 975 × LAD 122	2*	7+8	-
	Alzo × LAD 122	2*	7+9	-
	Alzo × F 8063	2*	7+8, 6+8, 6+7+8	-
Wheat patterns	Zebra	2*	7+9	7+9
	Clever	1	6+8	2+12

Glu B1 for three hybrid strains, *i.e.* IGS 5101 × LAD 122, FDT 975 × LAD 122 and Alzo × F 8063 was derived from the maternal form. It has to be noted that in the Alzo × F 8063 strain the subunits not occurring in the parental forms were also identified. The presence of additional HMW subunits in triticale hybrids, not detected in parental forms, has previously been described in literature [Igrejas *et al*,. 1999]. For the IGS 5101 × F 8063 hybrid strain only the subunits not

detected in parental forms were found. These subunits were attributed to both the *Glu A1* and *Glu B1*. It probably shows the lack of levelling off with respect to these proteins, which proves differentiation on long arms of chromosomes of the first homologous group. The studied samples of hexaploid triticale did not have subunits coded by loci *Glu D1*. Waga & Grzesik [2003] connect the presence or lack of subunits of the x type (heavier, derived from wheat) to a higher technological value of triticale. Subunits of this type were found to occur in all the studied forms. Electrophoretic analysis of gliadins and secalins of the studied hybrid strains showed intralinear polymorphism with respect to the above mentioned proteins. Figure 2 shows the differentiation of electrophoregrams of proteins obtained from 5 individual kernels within one genotype. For example, the hybrid strains FDT 975 × LAD 122 and parental components were shown. In the case of triticale, the assessment of baking qualities on the basis of the composition of gluten proteins only is not so unambiguous as in wheats because of the substitution of subunits having 1D with secalins having 1R from rye [Payne *et al.*, 1987; Makarska, 2000]. Amiour *et al.* [2002] demonstrated that even with a high frequency of occurrence of alleles correlated with a good baking quality in triticale, the quality is deteriorated by a low efficiency and quality of gluten and a high level of α-amylase.

The choice of the parental components of 3 hybrid strains (IGS 5101 × LAD 122, IGS 5101 × F 8063, FDT 975 × LAD 122) did not affect an increase in protein level of grain. The Alzo × LAD 122 strain surpassed the maternal form and the Alzo × F 8063 strain both of the parental components in protein level.

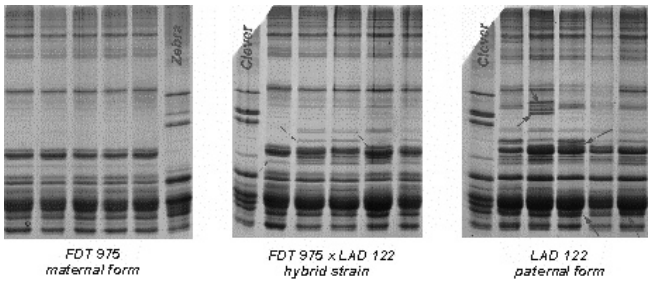

FIGURE 2. Electrophoregrams (Acid –PAGE) of gliadins and secalins of winter triticale (5 single kernels) and standard cultivars of wheat (Zebra, Clever): maternal form FDT 975, hybrid FDT 975 x LAD 122, paternal form LAD 122.

The falling number supplies the information about the level of liquefaction of starch in the endosperm as a result of the action of α-amylase [Sodkiewicz & Sodkiewicz, 2003]. A low falling number, proving a great activity of amylolytic enzymes in triticale grain, indicates that dough made of it will by too liquid for breadmaking [Tohver *et al*., 2005]. A statistically significant ($p \leq 0.05$) difference of the falling number in maternal forms, from a low one amounting to 100 s in the case of the IGS 5101 strain to 196 s in the FDT 975 strain, was not reflected in the tendency to germinate in the hybrid strains grain. Latent germination (without visible signs of germinating) expressed by low value of the falling number below 100 s, was shown in all the parental forms and hybrid strains except the FDT 975 x LAD 122 strain. Among the studied samples this strain was characterised by the highest falling number – 205 s, similar to that of its maternal form. Partial domination of low-activity α-amylase in the grain of the hybrid strain FDT 975 x LAD 122 depending on the combination of parental genotypes is analogous with the studies by Masojć [1997]. In this investigation, the feature of low activity of α-amylase in triticale hybrids expressed as a domination degree was changing in dependence on parental forms selection, regardless whether the resistant strains were maternal or parental forms.

The content of soluble pentosans in kernels of the studied strains varied greatly– from 0.98 to 2.67% d.m. (Table 3). Maternal forms were characterised by the lowest level of these components (0.98-2.04%). The assessed hybrids were usually characterised by a higher content of soluble pentosans as compared to their parental forms. The highest level of the discussed polysaccharides was found in the Alzo × F 8063 hybrid. The dependency of a higher percentage level of pentosans in hybrid strains of triticale with *Aegilops* sp. on their parental components was also found by Makarska *et al*. [1999]. This dependency was maintained in the subsequent generations of the plants.

Varughese *et al*. [1996] state that the content of pentosans in triticale is similar or higher than in wheat and much lower

TABLE 3. Quality parameters of hybrid strains and parental components of winter triticale grain.

Strain/cultivar		Crude protein (% d.w.)	Falling number (s)	Soluble pentosans (% d.w.)
Maternal forms	IGS 5101	$12.21^{cbd} \pm 0.15$	$100^{a} \pm 41.7$	$1.62^{b} \pm 0.01$
	FDT 975	$11.28^{abc} \pm 0.16$	$196^{b} \pm 25.5$	$0.98^{a} \pm 0.35$
	Alzo	$11.76^{a} \pm 0.31$	$167^{b} \pm 15.6$	$2.04^{bc} \pm 0.08$
Paternal forms	LAD 122	$12.71^{d} \pm 0.14$	$62^{a} \pm 1.4$	$2.19^{c} \pm 0.35$
	F 8063	$11.49^{ab} \pm 0.14$	$62^{a} \pm 0.0$	$2.06^{bc} \pm 0.03$
Hybrids	IGS 5101 × LAD 122	$11.58^{abc} \pm 0.23$	$63^{a} \pm 0.7$	$2.19^{c} \pm 0.00$
	IGS 5101 × F 8063	$11.28^{a} \pm 0.16$	$62^{a} \pm 1.4$	$2.17^{c} \pm 0.04$
	FDT 975 × LAD 122	$11.16^{a} \pm 0.11$	$205^{b} \pm 5.7$	$2.48^{cd} \pm 0.00$
	Alzo × LAD 122	$12.25^{cd} \pm 0.08$	$62^{a} \pm 0.0$	$2.42^{cd} \pm 0.02$
	Alzo × F 8063	$11.64^{abc} \pm 0.16$	$77^{a} \pm 4.2$	$2.67^{d} \pm 0.04$

Values are mean ± standard deviation; a,b,c,d- mean values with the same letter do not differ at $p \leq 0.05$

than in rye. A higher level of pentosans in the grain of the studied hybrid strains of triticale is desirable because of their favorable influence on rheological qualities of the dough and on slowing down the processes of products staling [Michniewicz *et al*., 1992]. The strains IGS 5101 × LAD 122 and IGS 5101 × F 8063, with the lowest content of arabinoxylans, may be used for feeds.

CONCLUSIONS

1. The estimation of the studied parameters pointed to parental genotypes: the maternal FDT 975 and paternal LAD 122, as the most valuable ones due to the quality of grain of hybrid strains.

2. Among the studied hybrid strains, the strain FDT 975 × LAD 122 was characterised by the presence of qualitatively good glutenin subunits, the highest falling number, and a higher content of soluble pentosans in the grain.

3. As a result of crossing and selection of chosen triticale strains the hybrids with more efficient nutritional parameters in relation to their parental components can be obtained..

REFERENCES

1. Amiour N., Dardeved M., Khelifi D., Bouguennec A., Branlard G., Alleic variation of HMW and LMW glutenin subunits, HMW secalin subunits and gamma-secalins of hexaploid triticale. Euphytica, 2002, 123, 179–186.
2. Brzeziński W., Łukaszewski A.J., Allelic variation at the Glu-1, Sec-2, Sec-3 loci in winter triticale. Proc. 4th Int. Triticale Symp. Red Deer, Alberta, 1998, 2, 6–12.
3. Brzeziński W., Van Gelder W.M.J., Mendelewski P., Kolster P., Polyacrylamide gel electrophoresis of wheat gliadins: the use of a moving boundary for improved resolution. Euphytica, 1989, 40, 207–212.
4. Ciołek A., Makarska E., Estimation of quality of new lines of durum wheat on the basis of gliadin and glutenin characterization under conditions of different nitrogen fertilization supply. Acta Sci. Pol. Technol. Aliment., 2004, 3, 147–155 (in Polish).
5. Dojczew D., Sobczyk M., Grodzicki K., Haber T., The influence of pre-harvest sprouting grains on the breadmaking wheat, rye, and triticale flour. Acta Sci. Pol. Technol. Aliment., 2004, 3, 127–136 (in Polish).
6. Gianibelli M.C., Larroque O.R., MacRitchie F., Wrigley C.W., Biochemical, genetic and molecular characterization of wheat glutenin and its component subunits. Cereal Chem., 2001, 78, 635–646.
7. Hashimoto S., Shogren M., Pomeranz Y., Cereal pentosans: estimation and significance. I. Pentosans in wheat and milled wheat products. Cereal Chem., 1987, 79, 880–884.
8. Igrejas G., Guedes-Pinto H., Carnide V., Branlard G., Seed storage protein diversity in triticale varieties commonly grown in Portugal. Plant Breeding, 1999, 118, 303–306.
9. Makarska E., The attempt to connect the glutenin composition with evaluation of the baking quality of winter triticale grains. Pol. J. Food Nutr. Sci., 2000, 9/50, 21–24.
10. Makarska E., Gruszecka D., Wojciechowska M., Biologically active non-nutrient compounds in grain of triticale and its hybrids with *Aegilops* sp.. Biul. IHAR, 1999, 211, 177–183 (in Polish).
11. Makarska E., Szwed-Urbaś K., Genetic conditionings of the quality of grain of new lines of *Triticum durum* Desf. Int. Agrophysics, 2005, 19, 147–152.
12. Masojć P., Genetic control of high alpha-amylase activity in triticale grain. Zesz. Nauk. AR Szczecin, 1997, 175, 259–263 (in Polish).
13. Michniewicz J., Pentosans in grain technology. 1995, Dissertation of habilitation. AR Poznań, p. 261 (in Polish).
14. Michniewicz J., Biliaderis C.G., Bushuk W., Effect of added pentosans on some properties of wheat bread. Food Chem., 1992, 43, 251–257.
15. Payne P.J., Nightingale M., Krattiger A., Holt L., The relationship between HMW glutenin subunit composition and the breadmaking quality of British-grown wheat varieties. J. Sci. Food. Agric., 1987, 40, 51–65.
16. Perez G.T., Leon A.E., Ribotta P.D., Aguirre A., Rubiolo O.J., Anon M.C., Use of triticale flours in cracker-making. Eur. Food Res. Technol., 2003, 217, 134–137.
17. PN – ISO 3093: 1996. Cereal. Falling number estimation (in Polish).
18. Sodkiewicz W., Sodkiewicz T., Inhibition of alfa-amylase acting in hexaploid triticale lines by exogenous abscisic acid. Biologia Plantarum, 2003, 46, 419–422.
19. Tohver M., Kann A., Taht R., Mihhalevski A., Hakman J., Quality of triticale cultivars suitable for growing and bread-making in northern conditions. Food Chem., 2005, 89, 125–132.
20. Varughese G., Pfeiffer W.H., Pena R.J., Triticale: A successful alternative Crop (Part 2). Cereal Foods World, 1996, 41, 635–645.
21. Waga J., Polymorphism of gliadin and glutenin proteins and their relation to quality of winter wheat (*Triticum aestivum* L). Biul. IHAR, 1997, 204, 205–218 (in Polish).
22. Waga J., Grzesik H., Electrophoretic analysis of storage proteins in some winter triticale strains resistant to preharvest sprouting. Biul IHAR, 2003, 230, 157–163(in Polish).

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