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Comparison of nutritional and technological quality of winter wheat (*Triticum aestivum* L.) and hybrid wheat (*Triticum aestivum* L. x *Triticum spelta* L.)

Porovnanie nutričnej a technologickej kvality pšenice letnej, formy ozimnej (*Triticum aestivum* L.) a hybridnej pšenice (*Triticum aestivum* L. x *Triticum spelta* L.)

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Abstract

Wheat is one of the most important food crop in the world. Gluten proteins represent major protein fraction of the grain and are responsible for unique properties of the dough. The aim of this study was to analyze one genotype of the winter wheat (*Triticum aestivum* L.) and one genotype of hybrid wheat (*Triticum aestivum* L. x *Triticum spelta* L.) based on protein polymorphism and to compare their nutritional and technological quality. According to Kjeldahl and Golenkov, content of total nitrogen and protein fraction composition of analyzed samples were determined. Higher content of albumins and globulins (26.39%) was detected in hybrid wheat genotype PS Lubica with total nitrogen 2.02% and crude protein content 11.49%. Winter wheat genotype Elinor showed higher content of prolamins and glutelins (70.68%) and higher coefficient of nutritional quality (91.79%) than analyzed hybrid wheat. Higher total content of amino acids was determined in hybrid wheat genotype PS Lubica (122.02 $mg*g^{-1}$ dry weight). For the separation of glutenin protein subunits, SDS-PAGE method by ISTA was used. On the basis of molecular weight, glutenins were separated in SDS-PAGE into HMW-GS (Elinor 10.7%; PS Lubica 3.78%) and LMW-GS (Elinor 58.4%; PS Lubica 67.28%). Electrophoretic profile of genotype Elinor was 0, 7+9, 5+10 with Glu Score 7 and composition of HMW-GS in genotype PS Lubica was 0, 20, 2+12 with Glu Score 4. According to the results, better nutritional and technological quality was proved in winter wheat genotype Elinor. According to identical results in 5 biological replicates, hybrid wheat genotype PS Lubica was not recommended for bread-making.

Keywords: nutritional quality, storage proteins, technological quality, wheat

Abstrakt

Pšenica je jednou z najdôležitejších potravinárskych plodín na svete. Lepkotvorné bielkoviny predstavujú hlavnú bielkovinovú frakciu zrna a sú zodpovedné za jedinečné vlastnosti cesta. Cieľom tejto práce bolo analyzovať 1 genotyp pšenice letnej, formy ozimnej (*Triticum aestivum* L.) a 1 genotyp hybridnej pšenice (*Triticum aestivum* L. x *Triticum spelta* L.) na základe polymorfizmu bielkovín a porovnať ich nutričnú a technologickú kvalitu. Podľa Kjeldahla a Golenkova bol stanovený obsah celkového dusíka a frakčnej skladby bielkovín analyzovaných vzoriek. Obsah hrubých bielkovín a koeficient nutričnej kvality boli vypočítane. Väčší obsah albumínov a globulínov (26,39%) bol detekovaný v genotype hybridnej pšenice PS Lubica s celkovým dusíkom 2,02% a obsahom hrubých bielkovín 11,49%. Genotyp pšenice letnej, formy ozimnej Elinor preukázal vyšší obsah prolamínov a glutelínov (70,68%) a vyšší koeficient nutričnej kvality (91,79%) ako analyzovaná hybridná pšenica. Vyšší celkový obsah aminokyselín bol stanovený v genotype hybridnej pšenice PS Lubica (122,02 mg*g^{-1} sušiny). Na separáciu glutenínových podjednotiek bielkovín bola použitá metóda SDS-PAGE (ISTA). Na základe molekulovej hmotnosti boli gluteníny separované v SDS-PAGE na HMW-GS (Elinor 10,7%; PS Lubica 3,78%), LMW-GS (Elinor 58,4%; PS Lubica 67,28%) a zvyškové albumíny a globulíny. Elektroforetický profil genotypu Elinor bol 0, 7+9, 5+10 s Glu skóre 7 a zastúpenie HMW-GS v genotype PS Lubica bolo 0, 20, 2+12 s Glu skóre 4. Vzhľadom na výsledky, lepšia nutričná a technologická kvalita bola dokázaná v genotype pšenice letnej, formy ozimnej Elinor. Vzhľadom na zhodné výsledky 5 biologických opakovaní, genotyp hybridnej pšenice PS Lubica sa neodporúča na výrobu chleba.

Kľúčové slová: nutričná kvalita, pšenica, technologická kvalita, zásobné bielkoviny

Introduction

Cereals are very important crops for production of feed and food (Belitz et al., 2009). Wheat is one of the most grown cereal in the world. In human nutrition, it is very important cereal crop which is main source of proteins, energy and dietary fiber. Wheat grain is composed of 8% to 20% of proteins (Anjum et al., 2007).

Major determinants of wheat quality are storage proteins. Proteins from wheat flour combined with water formed gluten, which holds gas produced by yeast during baking. Gluten proteins are gliadins and glutenins. Wheat has three genomes, namely A, B and D. Gliadins are the major storage proteins of the wheat grain endosperm with molecular weight from 30 kDa to 60 kDa. They are categorized into *α-, β-, γ-* and *ω*-gliadins. Glutenins include high molecular weight glutenin subunits (HMW-GS) and low molecular weight glutenin subunits (LMW-GS) (Ram and Mishra, 2010; Katyal et al., 2016).

HMW-GS (80-130 kDa) are encoded by Glu-A1, Glu-B1 and Glu-D1 loci located on the long arm of chromosomes. Glu-1 loci contains genes, which encode x-type and y-type glutenin subunits based on their molecular weight. HMW-GS play important role in determining wheat gluten and dough elasticity (Shewry et al., 2002; Henkrar et

al., 2017). Dough properties and bread-making quality are influenced in negative or positive way by HMW-GS. Alleles 5+10 and 17+18 have a positive influence whereas allele Null and 2+12 have a negative effect (Liu et al., 2009).

LMW-GS (10-70 kDa) influence quality of the wheat products, effect strength of gluten and extensibility of dough. On the short arm of chromosomes are Glu-A3, Glu-B3 and Glu-D3 loci, which encode LMW-GS (Henkrar et al., 2017).

One of the main methods used to analyze cereal proteins is gel electrophoresis, which is used to separate macromolecules of proteins based on their electric charge, particle size and molecular weight (The United States Pharmacopeial Convention, 2014). Components of the wheat gluten are separated by SDS-PAGE or two-dimensional gel electrophoresis (Shewry et al., 2002).

Materials and methods

Plant material

Grains of 1 winter wheat (*Triticum aestivum* L.) genotype Elinor and 1 hybrid wheat (*Triticum aestivum* L. x *Triticum spelta* L.) genotype PS Lubica were obtained from Research and Breeding Station at Vígľaš-Pstruša – The Research Institute of Plant Production, Piešťany.

Determination of total nitrogen (Kjeldahl)

Determination of the total nitrogen content of analysed samples was performed according to Kjeldahl (1983) in 5 biological replicates for each plant sample. Mineralisation of 0.5 g plant material was carried at 420 °C for 20 minutes in presence of catalyst ($CuSO_4$ + K_2SO_4) and concentrated sulphur acid H_2SO_4. Distillation of ammonia from analysed samples was carried for 5 minutes. Titration of the NH_3 distilled into the H_3BO_3 solution was performed by using of Tashiro‘s indicator with 0.05 mol*dm^{-3} H_2SO_4.

Determination of protein fraction composition (Golenkov method)

Extraction of protein fractions (albumins, globulins, prolamins, and glutelins) from plant samples was carried according to Golenkov, with modifications of procedure reported by Michalík (2002). The protein content of individual fractions was measured in five biological replicates and calculated according to Kjeldahl.

Electrophoretic separation of proteins in SDS-PAGE

Seed storage proteins were isolated from individual, whole, dry, mature grains according to standard method by ISTA (Wrigley, 1992). Storage protein separation was carried in polyacrylamide gel in the presence of sodium dodecyl sulfate. Electrophoresis was performed in the vertical discontinual electrophoretic system Hoefer SE 600 DeLuxe by Thermo Fisher Scientific by the standard reference electrophoretic method by ISTA (Wrigley, 1992). Polyacrylamide gels were stained

with Coomassie Brilliant Blue R250 and visualised in photo device with black-white CCD camera. Aquired images were analyzed in Doc-It LS Image analysis UVP software. The Glu Score was calculated according to the catalogue of alleles for HMW-GS (Payne et al., 1987 cited in Lásztity, 2003).

Two dimensional gel electrophoresis (2-DE)

Dry mature grains were used for isolation of storage proteins. Extraction of proteins was performed according to Hurkman and Tanaka (1986). Determination of protein concentration after extraction was carried by Bradford (1976). Protein samples were incubated with ReadyStripTM IPG Strips 11 cm, 3–10 pH (Bio-Rad) after addition of appropriate volume of IEF buffer. Isoelectric focusing was carried in Bio-Rad PROTEAN ® IEF CellUnit with linear pH gradient 3–10. Electrophoretic separation of proteins was performed in 10% SDS polyacrylamide gels at 30 mA and 10 °C for 6-10 hours. Polyacrylamide gels were stained with Coomassie Brilliant Blue R250. Gels were visualised with scanner Bio-Rad GS-800 Calibrated Densitometer and aquired images were edited in Quantity One program (Bio-Rad). Images were analysed in program PDQuest™ 2-D Analysis Software (Bio-Rad).

Results and discussion

Cereals are one of the most important food sources. Protein content of cereal grain indicates nutritional and technological quality. Quantity and quality of the grain storage proteins are the main factor, which influence technological quality of the grain. On the other hand, nutritional quality depends on content of essential amino acids (Payne et al., 1983; Petrovičová et al., 2013).

Table 1. Average values of nitrogen and protein content

	Elinor[1]	PS Lubica[2]
Total nitrogen [%]	1.74	2.02
Crude protein content [%]	9.9	11.49
Coefficient of nutritional quality [%]	91.79	88.29

[1]Elinor – genotype of winter wheat *Triticum aestivum* L., [2]PS Lubica – genotype of hybrid wheat *Triticum aestivum* L. x *Triticum spelta* L.

Measurement of total nitrogen, crude protein content and coefficient of nutritional quality was performed in 5 repetitions. In the Table 1 were presented average values of measuring data. Higher value of total nitrogen was detected in hybrid wheat PS Lubica (2.02%). Winter wheat (genotype Elinor) contained 1.74% of total nitrogen. Average content of crude protein was 9.9% for Elinor and 11.49% for PS Lubica. Coefficient of nutritional quality was calculated according to individual protein fractions of the analyzed wheat grains. Higher average value of coefficient of

nutritional quality was detected in Elinor (91.79%). Coefficient of nutritional quality detected in hybrid wheat PS Lubica was 88.29%.

Comparable results of the average crude protein content detected Gálová et al. (2011) who analyzed variously wheat varieties in work. The average crude protein content in cereals was 9.13%. Wheat contained 8% to 11.35%, which was consistent with this results. According to Socha (2011), the content of crude wheat proteins ranged from 10.37% to 14.08%, which was not fully correspond to this results.Wheat endosperm proteins were classified by Osborne (1907), based on their different solubility, into albumins soluble in water, globulins soluble in saline solutions, prolamins soluble in 70% to 90% alcohol and glutenins soluble in dilute alkalis and acids.

In the Table 2 were presented average values of protein fraction composition of analyzed seeds. Measurement of individual protein fractions was performed in five repetitions. Nutritional quality of wheat is positively influenced by higher levels of albumins and globulins. Higher content of albumins and globulins was detected in hybrid wheat PS Lubica (26.39%). Content of albumins and globulins in winter wheat Elinor was 21.97%. For bakery industry, content of wet gluten in minimal value 25% is very important. The technological quality of cereals is influenced by the quality and content of gluten proteins, namely prolamins and glutelins. Their higher content affects the technological quality in positive way and lower content in negative way. Higher content of prolamines and glutelines (70.68%) was detected in winter wheat, genotype Elinor. According to contain of prolamins and glutelins (65.17%), hybrid wheat, genotype PS Lubica showed lower technological quality in comparison to winter wheat. Socha (2011) showed that the percentage of albumin and globulin fraction in wheat was 23.03% and the percentage of prolamine and gluteline fraction in wheat was 67.41%. Results are closed to percentages of the fractional composition of proteins obtained in this study.

Table 2. Average fractional composition of proteins in analyzed seeds

	Elinor[1]	PS Lubica[2]
Albumins+globulins [%]	21.97	26.39
Prolamins+glutelins [%]	70.68	65.17

[1]Elinor – genotype of winter wheat *Triticum aestivum* L., [2]PS Lubica – genotype of hybrid wheat *Triticum aestivum* L. x *Triticum spelta* L. [1]Elinor – genotyp pšenice letnej *Triticum aestivum* L., [2]PS Lubica – genotyp hybridnej pšenice *Triticum aestivum* L. x *Triticum spelta*.

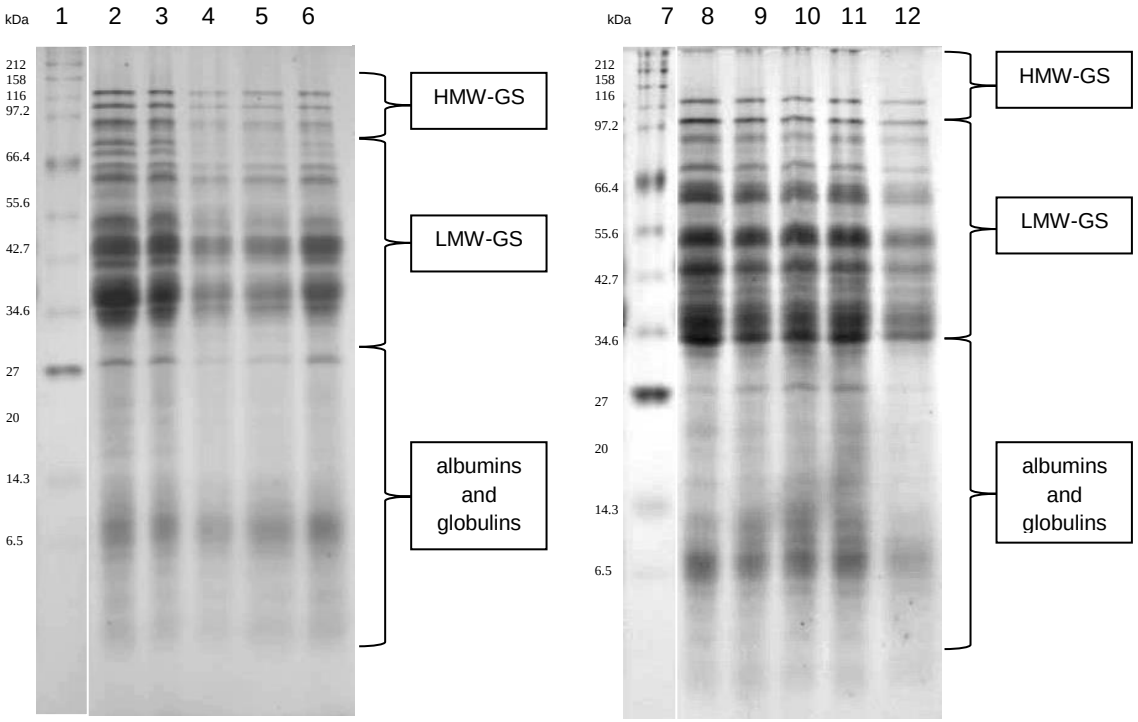
Table 3. Amino acid composition of proteins in analyzed wheat grains

Amino acid [mg*g^{-1} dry weight]	Elinor[1]	PS Lubica[2]
Aspartic acid	7.87	8.52
Threonine	4.36	4.41
Serine	5.71	6.12
Glutamic acid	38.92	41.2
Proline	11.5	13.45
Glycine	4.72	4.93
Alanine	3.74	4.24
Valine	4.04	4.58
Isoleucine	3.06	3.42
Leucine	7.59	8.22
Tyrosine	2.68	2.61
Phenylalanine	4.73	5.04
Histidine	3.3	3.35
Lysine	2.95	3.24
Arginine	4.8	4.84
Cystine	2.82	2.44
Methionine	1.52	1.41
Sum total	114.28	122.02

[1]Elinor – genotype of winter wheat *Triticum aestivum* L., [2]PS Lubica – genotype of hybrid wheat *Triticum aestivum* L. x *Triticum spelta* L. [1]Elinor – genotyp pšenice letnej *Triticum aestivum* L., [2]PS Lubica – genotyp hybridnej pšenice *Triticum aestivum* L. x *Triticum spelta* L.

Composition of amino acids in prolamins and glutelins is very different to albumins and globulins. From a nutritional point of view, prolamins contain more non-essential amino acids. On the other hand, albumins and globulins are important fractions of proteins because of higher content of essential amino acids (Alvarez-Jubete et al., 2010).

In collaboration with Research Institute for Animal Production in Nitra, the content of some amino acids (Table 3) in analyzed samples was determined. According to results, the most abundant amino acids in winter wheat and hybrid wheat were non-essential glutamic acid (38.92 mg*g^{-1} dry weight; 41.2 mg*g^{-1} dry weight) and proline (11.5 mg*g^{-1} dry weight; 13.45 mg*g^{-1} dry weight). The lowest abundant amino acids in winter wheat and hybrid wheat were essential amino acids methionine (1.52 mg*g^{-1} dry weight; 1.41 mg*g^{-1} dry weight) and lysine (2.95 mg*g^{-1} dry weight; 3.24 mg*g^{-1} dry weight) and non-essential cystine (2.82 mg*g^{-1} dry weight; 2.44 mg*g^{-1} dry weight). Hybrid wheat genotype PS Lubica showed higher total content of amino acids (122.02 mg*g^{-1} dry weight) in comparison to winter wheat genotype Elinor (114.28 mg*g^{-1} dry weight).



HMW-GS – high molecular weight glutenin subunits, LMW-GS – low molecular weight glutenin subunits, 1 – molecular marker P7702S, 2-6 – *Triticum aestivum* L. genotype Elinor , 7 – molecular marker P7702S, 8-12 – *Triticum aestivum* L. x *Triticum spelta* L. genotype PS Lubica.
HMW-GS – vysokomolekulárne glutenínové podjednotky, LMW-GS – nízkomolekulárne glutenínové podjednotky, 1 – molekulárny marker P7702S, 2-6 – *Triticum aestivum* L. genotyp Elinor , 7 – molekulárny marker P7702S, 8-12 – *Triticum aestivum* L. x *Triticum spelta* L. genotyp PS Lubica.

Figure 1. Electrophoretic spectrum of wheat storage proteins in SDS-PAGE

Differences in fractional composition of storage proteins influence nutritional and technological quality of grains of different wheat varieties (Gálová et al., 2012). The most widely used method for identification of high and low molecular weight glutenin subunits is polyacrylamide gel electrophoresis in the presence of sodium dodecyl sulfate (SDS-PAGE). This method is based on electrophoretic mobility of

proteins. It is simple, cheap and straight-forward technique but it has some negatives such as low resolution and overestimation of molecular mass (Liu et al., 2009; Chňapek et al., 2015).

By SDS-PAGE, storage proteins of analyzed wheat genotypes were separated, according to their molecular weight, into three different groups, high molecular weight glutenin subunits (HMW-GS) which were separated in the upside, low molecular weight glutenin subunits (LMW-GS) which were separated in the middle part of the polyacrylamide gel and the residual albumins and globulins (Figure 1).

Table 4. Molecular weight of protein subunits

Fractions of storage proteins	Molecular weight of protein subunits [kDa]
HMW-GS[1]	140-80
LMW-GS[2]	69-32
Alb+glo[3]	27-3

[1]HMW-GS – high molecular weight glutenin subunits, [2]LMW-GS – low molecular weight glutenin subunits, [3]alb+glo – albumins and globulins. [1]HMW-GS – vysokomolekulárne glutenínové podjednotky, [2]LMW-GS – nízkomolekulárne glutenínové podjednotky, [3]alb+glo – albumíny a globulíny.

In analyzed genotypes of wheat, the molecular weight (Table 4) of HMW-GS was from 140 kDa to 80 kDa. Molecular weight of LMW-GS was in the range of 69 kDa to 32 kDa and residual albumins and globulins 27 kDa to 3 kDa.

Socha (2011) detected comparable results of wheat protein subunits. Molecular weight of HMW-GS was in the range of 80 kDa to 140 kDa and LMW-GS 30 kDa to 80 kDa.

According to SDS-PAGE, percentage of high and low molecular weight glutenin subunits and residual albumins and globulins was determined (Table 5) by analysis of individual electrophoreograms. Some differences were detected in content of glutenin subunits between winter wheat and hybrid wheat.

In all analyzed samples, low molecular weight glutenin subunits presented the highest percentage of proteins, followed by residual albumins and globulins and the lowest percentage showed high molecular weight glutenin subunits (Table 5, 6).

Table 5. Percentage of HMW-GS, LMW-GS and residual cytoplasmatic proteins in analyzed genotype of winter wheat (*Triticum aestivum* L.)

Genotype	Repetition	HMW-GS[1] [%]	LMW-GS[2] [%]	alb+glo[3] [%]
	I	10.75	61.34	27.91
	II	10.98	61.57	27.45
Elinor	III	10.77	55.93	33.3
	IV	10.95	54.93	34.12
	V	10.03	58.27	31.7
X[4]		10.7	58.4	30.9
σ[5] [%]		0.35	2.71	2.74
Min		10.03	54.93	27.45
Max		10.98	61.57	34.12
V[6] [%]		3.23	4.65	8.88

[1]HMW-GS (%) - high molecular weight glutenin subunits; [2]LMW-GS (%) – low molecular weight glutenin subunits; [3]alb+glo (%) – albumins and globulins; [4]x - average; [5]σ (%) - standard deviation; [6]V (%) - coefficient of variation. [1]HMW-GS (%) – vysokomolekulárne glutenínové podjednotky; [2]LMW-GS (%) – nízkomolekulárne glutenínové podjednotky; [3]alb+glo (%) – albumíny a globulíny; [4]x – priemer; [5]σ (%) – smerodajná odchýlka; [6]V (%) – variačný koeficient.

Results showed, that in winter wheat (genotype Elinor), content of HMW-GS was detected in the range of 10.03% to 10.98% with an average 10.7%. The highest content of proteins reached LMW-GS with the average content 58.4%. Content of LMW-GS was 54.93% to 61.57%. Content of residual albumins and globulins achieved 27.45% to 34.12% with average content 30.9% (Table 5).

Content of HMW-GS (Table 6) in hybrid wheat genotype PS Lubica, was in the range of 2.32% to 4.73% with average content of 3.78%. Average percentage of LMW-GS reached 67.28%, with minimum at 64.51% and maximum at 69.59%. Content of residual albumins and globulins varies from 26.22% to 33.17% with mean value of 28.95%.

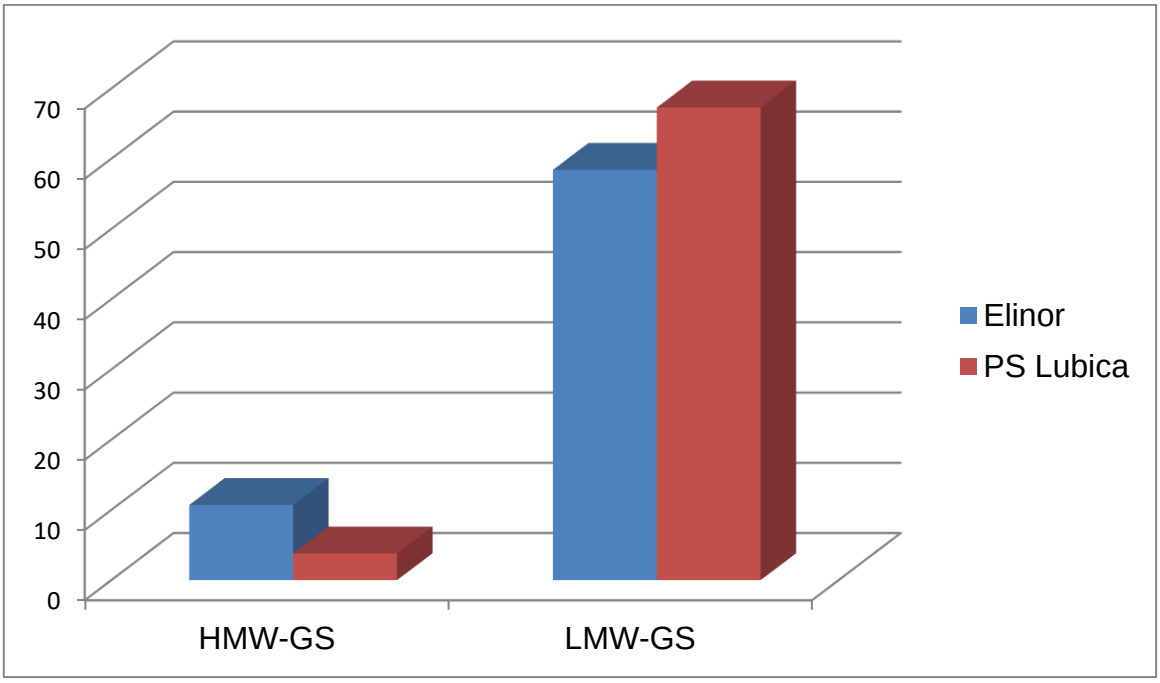
Table 6. Percentage of HMW-GS, LMW-GS and residual cytoplasmatic proteins in analyzed genotype of hybrid wheat (*Triticum aestivum* L. x *Triticum spelta* L.)

Genotype	Repetition	HMW-GS[1] [%]	LMW-GS[2] [%]	alb+glo[3] [%]
	I	4.73	68.16	27.12
	II	4.19	69.59	26.22
PS Lubica	III	3.37	69.45	27.19
	IV	2.32	64.51	33.17
	V	4.27	64.68	31.05
X[4]		3.78	67.28	28.95
σ[5] [%]		0.85	2.25	2.69
Min		2.32	64.51	26.22
Max		4.73	69.59	33.17
V[6] [%]		22.48	3.34	9.28

[1]HMW-GS (%) - high molecular weight glutenin subunits; [2]LMW-GS (%) – low molecular weight glutenin subunits; [3]alb+glo (%) – albumins and globulins; [4]x - average; [5]σ (%) - standard deviation; [6]V (%) - coefficient of variation. [1]HMW-GS (%) – vysokomolekulárne glutenínové podjednotky; [2]LMW-GS (%) – nízkomolekulárne glutenínové podjednotky; [3]alb+glo (%) – albumíny a globulíny; [4]x – priemer; [5]σ (%) – smerodajná odchýlka; [6]V (%) – variačný koeficient.

Detection of gluten proteins in two analysed genotypes showed some differences between amount of HMW-GS and LMW-GS (Figure 2). Winter wheat genotype Elinor reached higher content of HMW-GS (10.7%) in comparison to hybrid wheat genotype PS Lubica (3.78%). On the other hand, detection of LMW-GS in analysed genotypes showed that winter wheat genotype Elinor reached lower content of LMW-GS (58.4%) than hybrid wheat PS Lubica (67.28%).

Miháliková et al. (2016) determined in wheat higher content of HMW-GS (15.13%) and similar content of LMW-GS (65.89%) as these reached results. Comparable results showed Gálová et al. (2011), who determined content of HMW-GS in the range of 2.6% to 28.41% and LMW-GS 54.5% to 83.88%.



HMW-GS – high molecular weight glutenin subunits, LMW-GS – low molecular weight glutenin subunits. HMW-GS – vysokomolekulárne gluteníové podjednotky, LMW-GS – nízkomolekulárne gluteníové podjednotky.

Figure 2. Average percentage of HMW-GS and LMW-GS in analyzed genotypes

HMW-GS are encoded by genes at GLU-1 loci on the long arm of chromosomes of the A, B and D-genomes. Bread-making quality of wheat flour related to allelic variation. Subunits 1, 2*, 17+18 and 5+10 improve bread-making quality. On the other side, subunit pair 2+12 have negative effect on technological quality of the wheat grain (Rhazi et al., 2009).

Electrophoretic profile of winter wheat genotype Elinor was different from composition of glutenin subunits in hybrid wheat genotype PS Lubica (Table 7). On the GLU-A1 loci of genotype Elinor was detected allele Null, on the GLU-B1 loci was determined HMW-GS 7+9 and on the GLU-D1 loci was detected HMW-GS 5+10. Allele 7+9 and 5+10 have a positive effect on technological quality of the wheat grain.

Composition of glutenin subunits in genotype PS Lubica was very specific (Table 7). On the GLU-A1 loci was detected allele Null, on the GLU-B1 loci was determined allele 20, which can be typically found in spelt wheat. On the GLU-D1 loci was detected HMW-GS 2+12. HMW-GS 20 and 2+12 influence the guality of the wheat grain in negative way.

Tahir (2009) and Miháliková et al. (2016) determined similar electrophoretic profiles of winter wheat (0, 7+9, 5+10). Chňapek et al. (2014) detected allele 20 on the Glu-B1 loci in spelt wheat.

According to the composition of individual HMW-GS, quality of the wheat grain can be predicted by calculation of Glu Score with highest possible value 10. Wheat grains with Glu score higher that 7 are genotypes with good technological quality (Chňapek et al., 2014).

On the basis of Glu Score calculation (Table 7), better technological quality was proved in winter wheat genotype Elinor with Glu Score 7, which mean, this genotype can be characterized as a wheat with good technological quality. Hybrid wheat genotype PS Lubica with Glu Score 4 can not be recommended for bread-making and production of other fermented bakery products. It is suitable for non-fermented bakery products, for example cookies, crackers and biscuits.

Table 7. Composition of glutenin subunits and Glu Score

Genotype	Country of origin	The year of registration	GLU-A1[1]	GLU-B1[2]	GLU-D1[3]	Glu Score
Elinor	SR	2014	0	7+9	5+10	7
PS Lubica	SR	2014	0	20	2+12	4

[1]GLU-A1, [2]GLU-B1, [3]GLU-D1 – loci coded HMW-GS. [1]GLU-A1, [2]GLU-B1, [3]GLU-D1 – lokusy kódujúce HMW-GS.

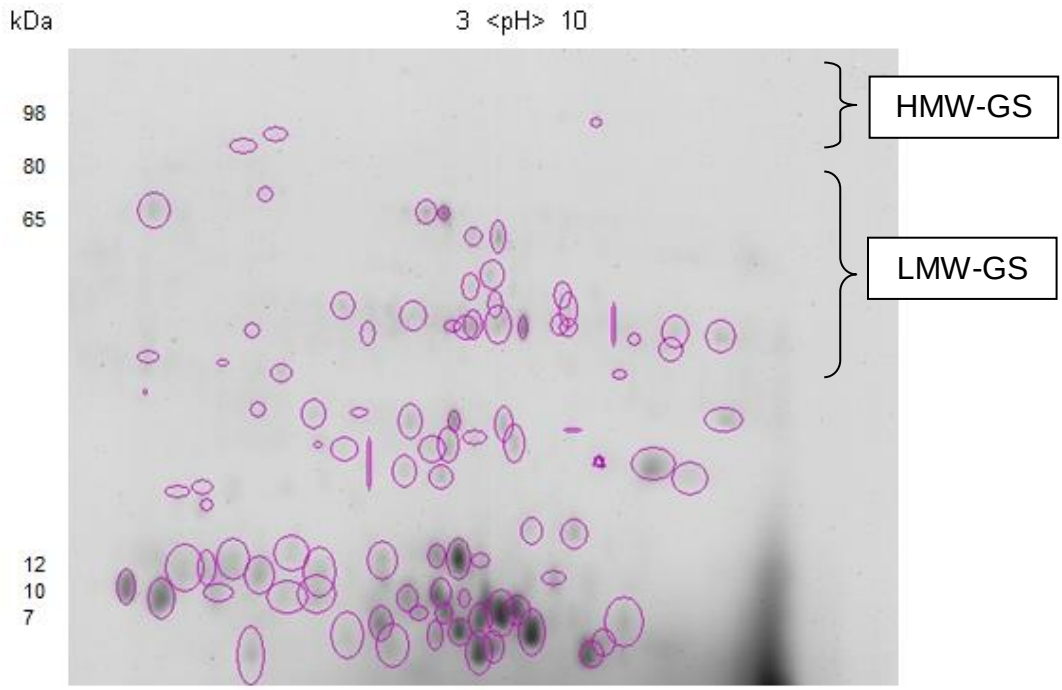


Figure 3. 2D gel of grain of winter wheat (*Triticum aestivum* L.), genotype Elinor

Two-dimensional polyacrylamide gel electrophoresis is one of the techniques used to analyze proteins, which are extracted from various biological sources. Proteins are separated according to their isoelectric point and molecular weight (Rodriguez et al., 2014).

2D gel of winter wheat genotype Elinor showed distribution of proteins in range of pH 3–10. In regard to the interpretation of results by programme PDQuest™, in Figure 3,

96 proteins with molecular weight from 98 kDa to 4 kDa was tagged. This proteins were located in area with pH from 4.5 to 9.5. HMW-GS can be described as proteins with size of 80–140 kDa. These were detected at range of pH 4.7-7.8. LMW-GS had size of 32 kDa–69 kDa and in gel they were distributed in pH range of 4.5–9.

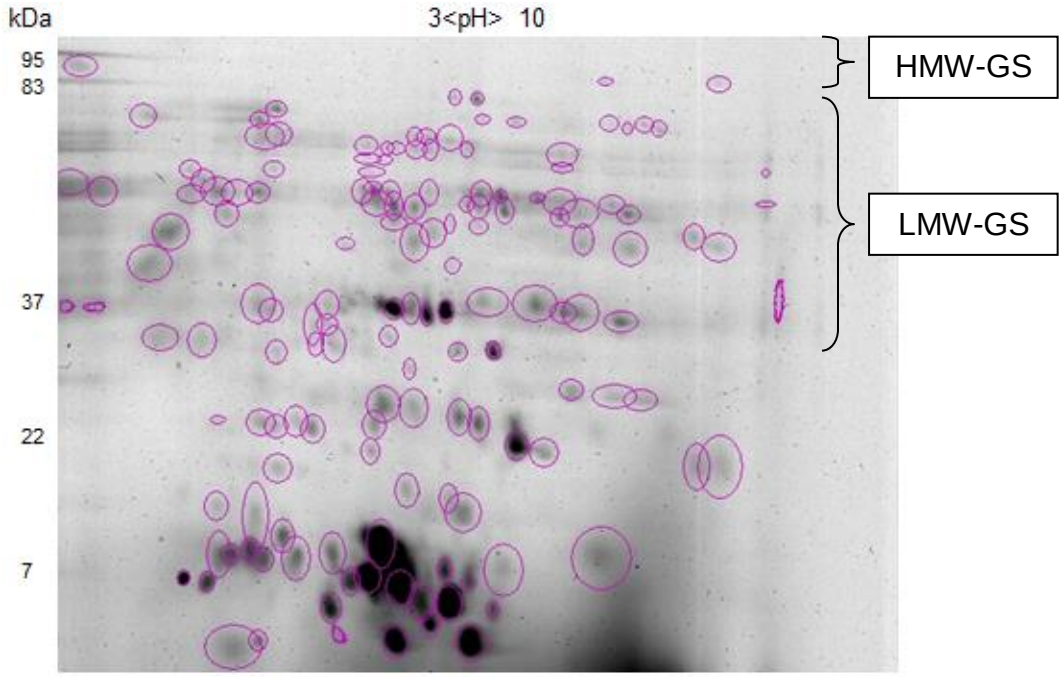


Figure 4. 2D gel of whole grain of hybrid wheat (*Triticum aestivum* L. x *Triticum spelta* L.), genotype PS Lubica

Analysis of grain proteins from genotype PS Lubica via 2D electrophoresis showed distribution of proteins in range of pH 3–10. In the 2D gel of genotype PS Lubica (Figure 4), programme PDQuest^{TM} was tagged 151 proteins with molecular weight in the range of 95 kDa to 4 kDa in the area with pH 4.2 to 9.7. Proteins with size of 80 kDa–140 kDa (HMW-GS) were found in range of pH 4–9. LMW-GS with size of 32 kDa–69 kDa were detected in pH range 4–9.5.

This results were similar with study of Gálová et al. (2014) and Skylas et al. (2000), who analyzed samples of wheat proteins by 2-dimensional gel electrophoresis. On the 2D gel, Gálová et al. (2014) detected gluten proteins with molecular weight 40 kDa to 200 kDa in the area with pH 9 to 11. Skylas et al. (2000) found on 2D gel group of HMW-GS with molecular weight 66 kDa to 100 kDa in the area with pH 5.5 to 6.5 and group of *ω*-gliadins with molecular weight 45 kDa to 55 kDa in area with pH 5 to 6.

Conclusions

Comparison of protein composition of analyzed genotypes showed that better technological quality was proved in winter wheat genotype Elinor, which showed higher content of prolamins and glutelins (70.68%) and higher coefficient of

nutritional quality (91.79%) than analyzed hybrid wheat genotype PS Lubica. HMW-GS, determinants of technological quality, influenced functional properties of gluten proteins. Average percentage of HMW-GS in genotype Elinor (10.7%) was higher than in hybrid wheat genotype PS Lubica (3.78%). On the other side, average percentage of LMW-GS was higher in genotype PS Lubica (67.28%) compared to genotype Elinor (58.4%). Electrophoretic profile of genotype Elinor was 0, 7+9, 5+10. HMW-GS 7+9 and 5+10 influenced technological quality of the grain in positive way. Glu Score of the winter wheat genotype Elinor was 7, what was a wheat with good technological quality. Composition of HMW-GS in genotype PS Lubica was 0, 20, 2+12, which had negative effect on technological quality of wheat grain. Hybrid wheat genotype PS Lubica with Glu Score 4 was not recommended for bread-making.

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