Variation in quality traits of newly developed Serbian wheat cultivars under different environmental conditions of Pannonian plain

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Abstract

Introduction of new high-yielding wheat cultivars and intensification of high year-to-year variability require an additional update of information about environmental effects on main wheat quality parameters that is lacking for newly released cultivars in Serbia. The aim of this study was to determine the effect of growing seasons on the technological properties of new Serbian wheat cultivars, and to examine the relationships among the quality traits in different growing seasons. The experiment was set up in three growing seasons (2016-17, 2017-18 and 2018-19). A total of 13 recently developed wheat cultivars in Serbia and five most widespread local cultivars were evaluated to compare differences in end-use quality. Also, the influence of climatic conditions on the technological quality was examined as well as relations among them. Modern Serbian cultivars have improved grain yield but lower technological quality than the widespread Serbian cultivars. Changes in farinograph resistance and degree of softening were related to the differences growing seasons, cultivars and their interaction, while growing seasons had the highest effect on bread volume and extensograph parameters energy and ratio resistance and extensibility variation. Moreover, a strong positive correlation was found between protein content, sedimentation value, wet gluten content and water absorption with extensograph extensibility, and negative correlation with the second parameter of extensograph, ratio of resistance and extensibility.

Introduction

Wheat is the most important winter cereal crop in the world. Furthermore, it is one the most widely cultivated field crops in Europe and Serbia, with average 2014-2018 production ranging from 254,716.6 to 2,583.6 thousand t, respectively (http://www.fao.org/faostat/en/#data/FBS). In the last decade in Europe, a fluctuation in wheat yields has been recorded from 3.5 t ha\(^{-1}\) to 4.4 t ha\(^{-1}\) (http://www.fao.org/faostat/en). Similarly, in the same period, the average grain yield in Serbia varied notably from 3.4 t ha\(^{-1}\) in 2010 to 4.4 t ha\(^{-1}\) in 2019 (Statistical Office of the Republic of Serbia, https://data.stat.gov.rs/Home/Result/130102?languageCode=en-US) showing remarkable
instability. Next to the high year-to-year grain yield variability, several studies (Bilgin et al., 2016; Mastilović et al., 2018; Liu et al., 2019) also confirmed a significant fluctuation of bread-making quality of wheat cultivars among different growing seasons. Furthermore, Fleitas et al. (2020) reported as one of the aims to identify which cultivars from a wide set possess both high yielding potential and stable bread-making quality under heat stress. Since most of the produced Serbian wheat is intended for local milling, baking sector and export, it is necessary to ensure stable production of high quality wheat.

Grain quality of winter wheat is under strong influence of environmental conditions as extreme weather events negatively affect starch and storage protein metabolism (Tomás et al., 2020). Different environmental factors, such as temperature, amount and distribution of precipitation, occurrence of heat and drought stress periods during plant development and grain filling period, have significant effects on wheat grain yield and quality (Balla et al., 2011; Malik et al., 2012; Dencic et al., 2013; Kondić-Špika et al., 2019). Heat and drought stress during anthesis and grain-filling period significantly decrease wheat grain yield by reducing grain number per plants and grain weight (Akter et al., 2017). Also, heat stress, temperatures above 35 °C during grain filling, has a negative effect on gluten extensibility (Hernández-Espinosa et al. 2018), although some genotypes retain high end-use quality. A large amount of precipitation before harvest can trigger pre-harvest sprouting, and even a small amount of germinated grains can cause a significant reduction of dough strength (Barbeau et al., 2006).

Different climate scenarios predict a rise of the average temperature by 2.0-4.9 °C until the end of this century due to climate changes (Asseng et al., 2015), as well as more frequent dry periods, heat stress and heavy rainfall (Trnka et al., 2014). Among the main wheat production regions in Europe, the Pannonian Plain has been identified as one of the most sensitive regions to the influence of further climate changes (Olesen et al., 2011). Therefore, predicted climate variability will further disturb high and stable production of quality wheat.
Apart from the environmental influence, wheat grain yield and quality are under the control of genotype, production management and their interaction (Rozbicki et al., 2015, Laidig et al. 2017). However, the influence of these factors on grain quality is inconsistent. Some traits, such as Zeleny sedimentation or flour yield, are more related to the genotypic effect (Carson and Edwards, 2009), while others, such as falling number or wet gluten, are under higher environmental control (Kong et al., 2013). Moreover, genotype by environment interaction has a strong influence on wheat quality, and variable genotypic response to different environments further complicates the production of high-quality wheat (Hristov et al., 2010, Rozbicki et al., 2015). However, in some long-term studies (Laidig et al. 2017), genotype by environment interaction was not so pronounced. Maintaining high quality of wheat cultivars across different growing seasons is crucial not only for human nutrition, but also for wheat breeders, producers and processing industry (Denčić et al., 2012).

In addition, grain quality is defined by several factors, such as physical and rheological traits strictly defined by end-users from different markets. Protein content is a basic indicator of wheat quality, strongly affected by environmental factors (Malik et al., 2013). Protein quality is highly related to the composition of gluten molecules and its main fractions, gliadins and glutenin subunits (GS) - high molecular weight (HMW) and low molecular weight (LMW), the specific composition of which is mainly determined by genotype (Johansson et al. 2013). Among them, HMW-GS are mostly related to the dough rheological properties of all proteins and their specific composition has a significant role in end-use quality of wheat. The difference in dough rheological properties is that the result of different composition of the HMW-GS amino acid sequences possess from four to seven cysteine residues at the N-terminal and C-terminal end (Shewry et al. 1992). Also, subunit HMW-GS 5 encoded by the D loci possesses an extra cysteine residue at the N-terminal end that is absent in HMW-GS 2 subunit encoded by the same loci (Gras et al. 2001). Liang et al. (2010) showed that LMW-GS also affected end-use quality of and acted as polymer chain terminators (Schmid et al. 2016). Furthermore, Guo et al. (2018) recently showed that
gliadins increase viscosity of gluten molecules and decrease glutenin elasticity resulting in actually softer gluten. In general, dough strength and elasticity are determined by glutenin. Furthermore, different important wheat rheological traits are closely related to the content and quality of wet gluten, and sedimentation value (Yang et al., 2014). Previous studies about the effects of genotype by environmental interaction on quality traits focused mainly on a limited set of generally outdated cultivars (Zečević et al., 2009; Živančev et al., 2016; Kondić et al., 2020). Introduction of new high-yielding wheat cultivars and intensification of high year-to-year variability require an additional update of information about environmental effects on main wheat quality parameters. Therefore, the main objectives of this study were a) to examine the effect of growing seasons on the main quality traits of thirteen newly released wheat cultivars in Serbia and five of the most commercially widespread wheat cultivars, and b) to analyse the relationships among the grain, flour, dough and baking traits across different growing seasons.

Materials and methods

Cultivars and experimental design

The study was conducted at the experimental field of the Institute of Field and Vegetable Crops, Novi Sad, Serbia (45°20’N and 19°51’E), during three growing seasons (2016-17, 2017-18 and 2018-19). The location is characterized by haplic chernozem aric soil (IUSS Working Group WRB, 2014) and a long-term three-year crop rotation of small grains, maize and legume crops. A total of 18 winter wheat cultivars developed at the Institute of Field and Vegetable Crops were selected for this study (Tab. 1). The main criterion for cultivar selection was to include five of the most commercially widespread cultivars (Simonida, NS 40S, Zvezdana, NS Futura and NS Ilina), as well as newly released high yielding cultivars (NS Mila, NS Obala, NS Javorka, NS Moma, NS Tajna, NS Jevrosima, NS Klica, NS Rani otkos, NS Vlajna, NS Epoha, NS Igra, NS Grivna and NS Todorka). The allelic composition of the high molecular weight glutenin subunits (HMW-GS) in the studied cultivars was obtained by the procedure described in Živančev et al. (2016) using
chip electrophoresis technique on an Agilent 2100 Bioanalyzer (Agilent Technologies, Santa Clara, CA, USA) with Protein 230 Lab-on-a-Chip kit for separation of proteins and presented in Table 1.

The experiment was set up as a completely randomized block design in three replications on plots 10 m long and 1 m wide with 10 rows per plot. Crops were sown on 17 October 2016, 23 October 2017, and 18 October 2018, providing the recommended sowing density for wheat production in southern Pannonian Plain of 500 plants per m². Basic soil properties were determined from the soil samples taken in each growing season from the topsoil layer (0-30 cm) before ploughing (Table 2). Based on these analyses, average doses of 60 kg ha⁻¹ of phosphorus (P) and 60 kg ha⁻¹ of potassium (K) were applied before sowing, along with 60, 45 and 50 kg of nitrogen (N) ha⁻¹ in 2016, 2017 and 2018, respectively. To avoid N deficiency during intensive plant growth, ammonium nitrate (34% N) was top-dressed based on the N-min analysis (Wehrmann and Scharpf, 1979) in early spring (Table 2). During the growing seasons, pests, diseases and weeds were controlled by recommended application of chemicals, when required.

**Grain, flour and bread quality traits**

Plots were combine-harvested at the grain ripening stage (late June), and grain yield (GY) was calculated at 13% moisture level. After harvest, the samples were threshed and cleaned to remove remaining parts of stalks and chaffs. The protein content (PC) in grain was determined according to the Kjeldahl method by Kejltec 2300 (Foss, Hillerød, Denmark) by AACC (2000) method 46-10 and expressed on a grain dry weight basis. Wheat samples were tempered for 24 h prior to milling to 13.5% moisture. Half an hour before milling, samples were prepared to the final tempering moisture of 15% and milled by a Bühler laboratory mill MLU-202 (Uzwil, Switzerland). Wet gluten content (WG) was obtained according to the ICC methods 137/1 using Glutomatic 2200 (Perten Instruments, Huddinge, Sweden). Falling number (FN) was determined with a Falling Number 1800 instrument (Perten Instruments, Huddinge, Sweden), according to
Hagberg, using the 107/1 method (ICC 2011). Sedimentation value (SV) of wheat samples was determined according to AACC (2000) method 56-62.01. Dough properties water absorption (WA), farinograph resistance (FR) and softening degree (SD) were obtained using a farinograph (Brabender OHG, Duisburg, Germany), according to the Hungarian Standard method 6369/653 (MSZ 1988). Extensograph energy (E), extensibility (EE) and resistance/extensibility ratio (R/EE) were measured on an extensograph (Brabender OHG, Duisburg, Germany) according to the 114/1 method (ICC 2011), while alveograph Alveograph deformation energy (W) was obtained using alveograph (Chopin, Paris, France) according to the 121 method (ICC 2011). The bread volume (BV) was measured by displacement method with millet, 24 h after baking, according to the internal procedure for evaluating wheat cultivars (Kaluderski and Filipović, 1998).

**Weather conditions**

The weather data for October to April in 2016-2017, 2017–2018 and 2018-2019 were obtained from the meteorological station located near the experimental field (Fig. 1).

Weather conditions during flowering and grain-filling stages, which in Serbia occur in May and June, respectively, are crucial for good grain quality performance. For this reason, the average temperatures and rainfalls were recorded on a daily bases starting from the flowering time (Fig. 2A-D).

**Statistical analysis**

The studied wheat quality parameters were subjected to analysis of variance (ANOVA) using Infostat (Di Rienzo et al. 2011). ANOVA was performed using a generalized linear model (GLM) including environments (growing seasons—GS), replications within environments, blocks within environments and their interactions. The components of variance were calculated to estimate the share of the factors and their interaction in the total variation of studied parameters. Linear
regression and Pearson’s correlation were calculated by averaging three replicates in the software InfoStat (Di Rienzo et al., 2011).

Results and discussion

Mid-daily air temperatures in October and November did not vary considerably among the growing seasons (Figure 1). On the other hand, precipitation amounts varied among growing season in autumn, when high, normal and low rainfall were measured in 2016-17, 2017-18 and 2018-19, respectively. Winter period in 2016-17 was characterized by the lowest average monthly temperatures and low rainfalls, while 2018-19 and 2017-18 were mild with moderate rainfall levels. February and March coincided with mid- and late-tillering period, and were relatively drier in 2016-17 and 2018-19 than in 2017-18. Plants were in stem elongation stage in April. All seasons were characterized by high rainfall and moderate temperatures, except 2017-18 which had higher average monthly temperature. The season 2017-18 was characterized by relatively higher temperatures during the first half of grain filling period, while the lowest daily temperatures were recorded in 2018-19 (Figure 2). The second half of grain filling period was characterized by high day-to-day temperature variability in 2017-18, relatively high daily temperatures in 2017-18 and 2018-19, and moderate daily temperatures in 2016-17. Daily precipitation was unequally distributed during grain filling period. There were several rainy days during mid-grain filling period in 2016-17 (Fig. 2B), while the best rainfall distribution was observed in 2018-19 (Fig. 2D). The results showed that almost half of grain yield (GY) variation (45.7%) was under genotypic effect (C), while the influence of year by cultivar interaction (Y × C) and year (Y) explained 32.4% and 21.9% of the total variation, respectively (Tab. 3). Although Serbian cultivars used in this study have been released in the last 15 years, high percentage of total variation explained by genotypic effect indicated diverse pedigrees of the cultivars reflected in differences in yield performance. The first six high yielding cultivars were the newly released, whereas four of the five most commercially widespread wheat cultivars (Simonida, Zvezdana, NS Futura and NS Ilina)
ranked from 12 to 15. Across the growing seasons, Serbian wheat cultivars achieved the highest grain yield in 2017-18, while the lowest GY was reported in 2016-17 (Tab. 4). According the Figure 1, the 2017-18 was characterized by high amount of precipitation that was equally distributed in spring, favouring crop growth and development. In small grain cereals, weather conditions during pre-anthesis and anthesis period are crucial for grain number and grain yield determination, as previously described by Slafer et al. (2014).

Growing season (Y), C, and Y × C had a significant (p<0.01) effect on PC (Tab. 3). More than half of variation in PC was explained by the influence of Y, followed by the influence of C and Y × C interaction. PC ranged from 11.6% in 2017-18 to 13.8% in 2018-19, with the total average protein content of 12.8% (Tab. 4). Also, three of the five the most commercially widespread wheat cultivars (Simonida, Zvezdana and NS Futura) ranked among the first top five based on PC. The analysed Serbian cultivars had higher PC on average than 20 most widely grown cultivars and advanced inbred lines from the International Winter Wheat Improvement Program in Turkey (Kaya and Akcura, 2014), and slightly lower PC than the average of 16 most widely grown cultivars in Spain (Sanchez-Garcia et al., 2015). Moreover, a significant variation in PC across different growing seasons has been reported for other environments, such as Mediterranean (Kaya and Akcura, 2014) or central Europe (Hlisnikovský et al., 2020). In our study, the lowest protein content was recorded in 2017-18 that was characterized by the highest average grain yield. This is not surprising as a strong negative correlation between PC and GY is generally known (Da Silva et al., 2014). The weather conditions during the grain filling period had significant effect on grain nitrogen accumulation and PC (Pan et al., 2006). The second part of grain filling period was characterized by higher temperatures in 2018-19 than in 2016-17 and 2017-18. High temperatures tend to shorten grain filling, thus decreasing grain carbohydrates accumulation in the developing grain, and resulting in more nitrogen per grain weight (Stone and Nicolas, 1998). The highest average PC was recorded in the cultivar NS Todorka (13.9%, recently developed), followed by NS Jevrosima (13.7%, recently developed), both of which had the same combination of HMW-
GS 2*, 7+9 and 2+12 with Glu score 7. On the other hand, the lowest PC was recorded in the cultivars NS Klica (11.6%, recently developed) and NS Grivna (11.9%, recently developed), which were characterized by the same combination of HMW-GS 2*, 7+9 and 5+10 and with Glu score 9. This is in accordance with the results of Johansson et al. (2020) and Blumenthal et al. (1995), who showed that some wheat cultivars with HMW–GS 5+10 at the Glu-D1 locus had lower values of technological parameters quality than cultivars with HMW–GS 2+12. Previous studies indicated higher variation in PC among different wheat cultivars, ranging between 10.8 and 15.7% in Romania (Liana et al., 2012), or between 12 and 16% in Croatia (Horvat et al., 2015). However, cultivars used in our study are mostly modern Serbian cultivars developed recently for intensive high-yielding production, showing less variability in PC. Generally, breeding activities in the past century resulted in development of high-yield cultivars with decreased PC (Mirosavljević et al., 2020) and such a trend was noted in our research.

Variations in WG and SV of the examined cultivars mostly related to differences among genotypes and were followed by the influence of the growing season. This is in accordance with Laidig et al. (2017) who found that SV was highly influenced by cultivars (variation – 69%). Furthermore, Rozbicki et al. (2015) showed that in addition to the significant influence of genotype on WG, Y × C interaction had a significant influence on the same parameter. The SV and WG content were higher in the years characterized by increased PC content. The highest SV was recorded in 2016-17, while WG was similar in 2016-17 and 2018-19. There was a high positive correlation of WG and SV with a PC (Table 6). The SV ranged between 22.0-41.7 ml, while WG varied between 24.9-38.8% (Tab. 4). The highest SV and WG content were recorded in NS Todorka, while NS Mila was characterized by the lowest value of SV and WG, whereas Simonida and Zvezdana ranked among first top six places based on these two parameters. Previously, Denčić et al. (2011) reported lower average WG content and its higher variation (21.3-40.6%) in winter wheat cultivars of different origins grown under conditions of the Pannonian Plain. On the other hand, Surma et
al. (2012) reported that SV varied between 34.1-53.6 ml and that was mainly under environmental control in Poland.

Farinograph trait WA significantly varied due to the influence of Y and C, which was in accordance with results of few studies (Linina et al., 2014; Rozbicki et al., 2015). However, Y × C interaction did not significantly influence WA in the examined Serbian cultivars, which was not the case in the study Rozbicki et al. (2015). FR and SD were under significant effect of Y, C and their interaction, whereas in Rozbicki et al. (2015) this effect was not significant. Variation in WA of the examined Serbian cultivars was mostly related to the differences among genotyped, whereas SD was mostly related to the influence of growing season. On the other side, almost half of variation of the FR parameter was explained by the influence of Y × C interaction. The best properties of dough during mixing were associated with the increased WA and FR, and decreased SD. Among the examined Serbian cultivars, the WA ranged from 53.9 to 61.2%, FR varied between 2.3 and 7.3 min, and SD from 51.7 to 108.3 BU (Tab. 4). Two of the five most commercially widespread wheat cultivars (Zvezdana and NS Futura) possessed the highest values of WA and FR. In addition, there were high positive relationships among protein parameters (PC and WG) with following farinograph parameters WA and FR (Table 4). Also, increase in WA with PC is in accordance with the one reported in Constantinescu et al. (2011). The results from other studies showed higher WA among different wheat cultivars with average content of 61.8% (high level of nitrogen fertilization) and 60.0% (low level of nitrogen fertilization) in Croatia (Šarčević et al., 2014) or between 58.2-65.1% in Lebanon (Sakr et al., 2021), whereas SD was on a similar level.

Growing season (Y), cultivar (C), and Y × C interaction had a significant (p<0.01) influence on FN (Tab. 3). This is similar to Rozbicki et al. (2015) who found that years and genotype possessed significant influence on the parameter, but not year × genotype interaction. More than 70% of variation in FN was explained by the influence of Y, followed by the influence of C and Y × C interaction. The second and third highest FN were recorded in Simonida and Zvezdana belonging
to the five most commercially widespread wheat cultivars. The lowest FN in the examined Serbian cultivars was recorded in 2016-17 (158 s), which is quite lower than 250 s minimum of optimal value, although the period of grain ripening (forty days after anthesis) was not characterized by high amount of precipitation (Fig 1. B). High fluctuations in day and night temperatures during grain maturation in 2016-17 could be a reason for premature increased production of first isomorph of wheat alpha-amylase in a small percentage of the cells in the aleurone layer without visible germination (Mares and Mrva, 2008), the effect known as late maturity α-amylase activity (LMA) triggered by environmental extremes (Farrell and Kettwell, 2008). In grain aleurone layer, the first isomorph of wheat α-amylase remains active, without any effect on kernel shape and size. On the other hand, 2018-19 growing season was characterized with the highest FN (>400 s), indicating insufficient α-amylase activity in wheat grain. In baking formulations with high FN flour, it is necessary to add α-amylase for bread loaf volume improvement (Posner and Hibbs, 2005). The obtained FN values in 2017-18 and 2018-19 were similar to the results of Šarčević et al. (2014) and Sakr et al. (2021).

Extensograph traits E, EE and R/EE, as well as alveograph parameter W, also significantly varied due to influence of Y, C and their interaction. However, influence varied among these extensibility traits, since E, EE, R/EE and W were mostly explained by the influence of C, Y and Y × C interaction, respectively. Variation in E and R/EE of the Serbian cultivars was mostly related to the differences between growing seasons, whereas EE and W were mostly related to the influence of cultivars. This is similar to Hernández-Espinosa et al. (2018) who reported that genotype variation had the highest contribution to W than environment, year and all interaction. Contrary to our findings, E and R/EE of Chinese cultivars were predominantly influenced by the genotype (Kun et al., 2020). The best extensibility dough properties were in 2018-19, since value of R/EE was in the optimum range between 1.5 and 2.5 (Kovačević, 2011), and W was the highest. The E ranged from 78.4 to 122.3 cm², while EE varied between 137 and 159 min, and R/EE from 2.0 to 3.7. Also, W varied from 176 to 226 J × 10⁻⁴ (Tab. 4). Moreover, Simonida, Zvezdana and NS
Futura possessed statistically higher value of W in comparison to the most 13 recently developed wheat cultivars except for NS Javorka, NS Moma and NS Jevrosima. The average results from the study of Šarčević et al. (2014) showed higher EE and lower R/EE among different wheat cultivars, especially in the case of high level of nitrogen fertilization - 190 mm and 1.3, respectively. Also, the values of alveograph parameter W were somewhat lower than the Lebanese wheat cultivars (Sakr et al., 2021).

Parameters Y, C and Y × C had a significant (p<0.01) influence on bread volume (BV) (Tab. 3). However, Rozbicki et al. (2015) found that only Y had a significant (p<0.05) influence on bread volume BV. Almost half of variation in BV of the Serbian cultivars was explained by the influence of Y, followed by the influence of C and Y × C interaction. Contrary to our findings, in Hernández-Espinosa et al. (2018) more than half of variation in BV was explained by the influence of genotype, followed by the influence of year and year × genotype interaction. The highest BV was recorded in 2016-17 (1441 cm$^3$) and was statistically higher than in 2017-18 (1255 cm$^3$) and 2018-19 (1269 cm$^3$). According to Ral et al. (2016), there were no results about LMA detrimental effect on the end-product quality. Although the best rheological properties were recorded in 2018-2019, they could not have a substantial effect on BV since our test baking method contained no sugar or addition of malt, regardless of the FN> 400 s high value. Furthermore, the three highest BV were recorded in Simonida, Zvezdana and NS Futura.

In order to determine whether some traits are stable across the environments, correlation coefficients were made between all growing seasons (Table 5). There were strong positive correlations of PC, SV, WG and E parameters among all three growing seasons. In the case of WA, FR, SD, FN parameters and correlations exist only between some growing seasons. This indicates that WA, FR, SD, FN and W parameters were dependent on weather conditions in the growing seasons.

The two most important wheat parameters GY and PC of the examined Serbian cultivars were in a strong negative correlation (Tab. 6). This was in agreement with results of Marinciu and Săulescu.
(2008) and Da Silva et al., (2014). Also, PC, SV and WG were strongly positively correlated with WA, which was in accordance with results of a study Rozbicki et al. (2015). Moreover, PC, SV, WG and WA were strongly positively correlated with EE, while negatively correlated with the second parameter of uniaxial dough extensibility R/EE. Contrary to our findings, the study of Kun et al. (2020) did not show any strong correlation among these parameters in individual years. The alveograph trait W of Serbian cultivars was in strong positive correlations with PC, SV, WG, WA and FR. Also, the W was in strong correlations of uniaxial extensibility dough traits; positive with EE and negative with R/EE. Furthermore, bread volume (BV) was strongly positively correlated with the most important technological properties PC, SV, WG, WA, and FR, while negatively correlated with R/EE. These results are in accordance with the previous findings of Wrigley and Bekes (2002) who showed a positive relationship between PC and BV, but not in accordance with Rozbicki et al. (2015) who showed a negative relationship between WA and BV. This correlation is used in the industry to help predict the suitability of flour for different end products.

Conclusions

The results of our study showed that recently developed Serbian wheat cultivars possess higher yield potential, but lower end-use quality compared to most widespread local cultivars, especially cultivars Simonida, Zvezdana and NS Futura. Also, significant influence of cultivar, year and their interaction on variation of farinograph parameters FR and SD was found. Furthermore, weather conditions that prevailed in the growing seasons mostly influenced the extensograph parameters E and R / E. The year had a statistically significant impact on most important parameter of end-use quality BV and almost half of variation in the BV was explained by the influence of year. Moreover, strong positive correlations among PC, SV, WG and WA with EE, and negative correlation with the second parameter of uniaxial dough extensibility R/EE were found. Therefore, further investigation of the studied technological traits variation is necessary in order to enable stable production of quality wheat cultivars.
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Table 1. The analysed cultivars, year of release and allelic composition at the high molecular weight glutenin subunits (HMW-GS) with GLU score.

| Cultivar     | Year of release | HMW-GS     | GLU score |
|--------------|----------------|------------|-----------|
|              |                | Glu-A1     | Glu-B1    | Glu-D1    |
| Commercially widespread cultivars |                |            |            |           |
| Simonida     | 2003           | N          | 7+9       | 2+12      | 5         |
| NS 40S       | 2006           | N          | 7+9       | 5+10      | 7         |
| Zvezdana     | 2005           | N          | 7+9       | 2+12      | 5         |
| NS Futura    | 2010           | 2*         | 7+9       | 5+10      | 9         |
| NS Ilina     | 2010           | N          | 7+8       | 5+10      | 8         |
| Newly released high yielding |                |            |            |           |
| NS Mila      | 2014           | N          | 7         | 5+10      | 6         |
| NS Obala     | 2015           | N          | 7+9       | 5+10      | 7         |
| NS Javorka   | 2015           | N          | 7+9       | 5+10      | 7         |
| NS Moma      | 2016           | N          | 7+9       | 2+12      | 5         |
| NS Tajna     | 2016           | N          | 7+9       | 5+10      | 7         |
| NS Jevrosima | 2016           | 2*         | 7+9       | 2+12      | 7         |
| NS Klica     | 2016           | 2*         | 7+9       | 5+10      | 9         |
| NS Rani otkos | 2017          | 2*         | 7+9       | 2+12      | 7         |
| NS Vlajna    | 2014           | N          | 7+9       | 5+10      | 7         |
| NS Epoha     | 2016           | 2*         | 7+8       | 5+10      | 10        |
| NS Igra      | 2016           | 2*         | 7+9       | 3+12      | 7         |
| NS Grivna    | 2016           | 2*         | 7+9       | 5+10      | 9         |
| NS Todorka   | 2016           | 2*         | 7+9       | 2+12      | 7         |

Table 2. Basic soil properties and amounts of applied nutrients.

| Soil properties                              | 2016/17 | 2017/18 | 2018/19 |
|----------------------------------------------|---------|---------|---------|
| pH (KCl)                                     | 7.33    | 7.17    | 7.41    |
| CaCO₃ (%)                                    | 5.17    | 5.08    | 4.85    |
| P₂O₅ (Egner-Riehm, AL method) (mg 100g⁻¹)    | 24.5    | 28.4    | 20.8    |
| K₂O (Egner-Riehm, AL method) (mg 100g⁻¹)     | 18.3    | 19.1    | 17.6    |
| Humus (Tyurin method) (%)                    | 2.7     | 2.9     | 2.5     |
| Available N (N-min method) (kg ha⁻¹; 0-90 cm) | 80      | 70      | 50      |

| Fertilization                               |         |         |         |
| P₂O₅ (kg ha⁻¹)                              | 60      | 50      | 70      |
| K₂O (kg ha⁻¹)                               | 60      | 50      | 70      |
| N (kg ha⁻¹) prior to sowing                 | 60      | 45      | 50      |
| N (kg ha⁻¹) topdressing (in spring)         | 80      | 98      | 120     |
Table 3. Relative contributions to total sum of squares and significance level of source of variance for the studied traits of winter wheat.

| Source       | GY     | PC     | WG     | SV     | WA     | FR     | SD     | FN     | E      | EE     | R/EE   | W      | BV     |
|--------------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| Year         | 10.3** | 69.7** | 1090** | 791**  | 134**  | 112**  | 27613**| 1036614**| 26502**| 6296** | 40.6** | 42850**| 581376**|
| Cultivar     | 2.5**  | 4.2**  | 146**  | 173**  | 29**   | 13**   | 1640** | 24488**| 2761** | 2195** | 3.5**  | 7973** | 50413**|
| Y x C        | 0.9**  | 0.8**  | 24**   | 31**   | 10**   | 11**   | 1366** | 12250**| 464**  | 714**  | 1.3**  | 3567** | 15837**|
| Error        | 0.1    | 0.3    | 2      | 2      | 6      | 0.04   | 1      | 21      | 4      | 0.02   | 73     | 3436   |

% of SS

- Year: 21.9, 58.8, 39.8, 28.3, 24.5, 27.2, 42.6, 71.3, 45.8, 17.0, 44.1, 25.0, 45.5
- Cultivar: 45.7, 30.4, 45.3, 52.8, 45.7, 27.1, 21.5, 14.3, 40.6, 50.3, 32.2, 39.6, 33.5
- Y x C: 32.4, 10.9, 14.8, 18.9, 29.8, 45.7, 35.8, 14.3, 13.6, 32.7, 23.7, 35.4, 21.0

GY – grain yield; PC – protein content; WG – wet gluten content; SV – sedimentation value; WA – water absorption; FR – farinograph resistance; SD – softening degree; FN – falling number; E – extensograph energy; EE – extensibility; R/EE – resistance/extensibility ratio; W – alveograph deformation energy; BV – bread volume; Y – year; C – cultivar; Y x C – year by cultivar interaction; SS – sum of squares; ** – significant at the 0.01 probability level; * – not significant.

Table 4. Average values of grain yield (GY), protein content (PC), wet gluten (WG), sedimentation value (SV), water absorption (WA), farinograph resistance (FR), softening degree (SD), falling number (FN), extensograph energy (E), extensibility (EE), resistance/extensibility ratio (R/EE), alveograph deformation energy (W) and bread volume (BV) in 18 wheat cultivars during three growing seasons (2016-17, 2017-18 and 2018-19).

| Cultivar     | GY     | PC     | WG     | SV     | WA     | FR     | SD     | FN     | E      | EE     | R/EE   | W      | BV     |
|--------------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| Simonida     | 7.47** | 13.5** | 35.2** | 36.2** | 58.8** | 3.8**  | 78.3** | 376**  | 120.7**| 173**  | 2.1**  | 236**  | 1433**|
| NS 40S       | 7.75** | 12.1** | 23.3** | 30.7** | 56.7** | 2.3**  | 108.3**| 260**  | 133.2**| 14.4** | 134**  | 14**   | 1200**|
| Zvezdana     | 7.40** | 13.4** | 34.8** | 34.3** | 59.6** | 5.3**  | 75.0** | 353**  | 104.0**| 169**  | 1.9**  | 221**  | 1413**|
| NS Futura    | 7.36** | 13.3** | 32.6** | 32.0** | 58.2** | 7.3**  | 51.7** | 286**  | 103.1**| 149**  | 2.4**  | 224**  | 1427**|
| NS Ilina     | 7.46** | 12.7** | 28.9** | 31.2** | 56.9** | 2.7**  | 80.0** | 324**  | 108.8**| 149**  | 2.7**  | 181**  | 1353**|
| NS Mila      | 7.30** | 12.0** | 24.9** | 22.0** | 56.0** | 2.7**  | 83.3** | 332**  | 64.3** | 115**  | 3.5**  | 154**  | 124**  |
| NS Obala     | 7.54** | 12.6** | 28.9** | 28.5** | 56.2** | 3.5**  | 85.0** | 237**  | 102.4**| 138**  | 3.2**  | 180**  | 1287**|
| NS Javorka   | 7.19** | 12.6** | 30.3** | 28.3** | 56.7** | 3.0**  | 83.3** | 380**  | 90.9** | 127**  | 3.4**  | 207**  | 1273**|
| NS Moma      | 7.77** | 13.1** | 33.8** | 33.3** | 58.7** | 4.3**  | 90.0** | 340**  | 98.0** | 157**  | 2.6**  | 208**  | 1360**|
| NS Tajna     | 8.52** | 12.0** | 27.6** | 27.5** | 53.9** | 2.8**  | 85.0** | 334**  | 85.3** | 136**  | 2.9**  | 176**  | 1253**|
| NS Jevrosima | 7.48** | 13.7** | 35.0** | 34.5** | 58.4** | 4.3**  | 103.3**| 229**  | 99.9** | 159**  | 2.2**  | 221**  | 1380**|
| NS Klica | 8.37<sup>ab</sup> | 11.6<sup>f</sup> | 27.8<sup>f</sup> | 27.5<sup>i</sup> | 54.0<sup>d</sup> | 4.0<sup>ef</sup> | 75.0<sup>i</sup> | 327<sup>d</sup> | 87.3<sup>th</sup> | 131<sup>hi</sup> | 3.2<sup>cd</sup> | 155<sup>f</sup> | 1187<sup>h</sup> |
| NS Rani otkos | 7.28<sup>de</sup> | 13.2<sup>b</sup> | 34.8<sup>c</sup> | 36.0<sup>b</sup> | 58.0<sup>d</sup> | 4.9<sup>ef</sup> | 97.1<sup>ac</sup> | 191<sup>i</sup> | 83.1<sup>hi</sup> | 154<sup>cd</sup> | 1.9<sup>k</sup> | 229<sup>h</sup> | 1368<sup>cd</sup> |
| NS Vlajna | 7.58<sup>e-e</sup> | 13.2<sup>b</sup> | 33.5<sup>a</sup> | 29.7<sup>i</sup> | 57.7<sup>a-d</sup> | 4.7<sup>cd</sup> | 87.4<sup>d-f</sup> | 284<sup>ef</sup> | 76.3<sup>i</sup> | 140<sup>f-h</sup> | 2.4<sup>l</sup> | 176<sup>df</sup> | 1333<sup>b-f</sup> |
| NS Epoha | 8.89<sup>bc</sup> | 13.0<sup>-d</sup> | 32.9<sup>ac</sup> | 34.8<sup>bc</sup> | 58.0<sup>d</sup> | 4.2<sup>ef</sup> | 75.0<sup>i</sup> | 271<sup>ef</sup> | 126.0<sup>ab</sup> | 153<sup>c-e</sup> | 3.0<sup>de</sup> | 213<sup>cd</sup> | 1360<sup>-f</sup> |
| NS Igra | 8.89<sup>a</sup> | 12.2<sup>def</sup> | 29.6<sup>df</sup> | 30.7<sup>f-h</sup> | 56.3<sup>b-d</sup> | 4.3<sup>de</sup> | 61.7<sup>i</sup> | 328<sup>d</sup> | 107.8<sup>d</sup> | 143<sup>de</sup> | 3.0<sup>de</sup> | 191<sup>e</sup> | 1280<sup>f-h</sup> |
| NS Grivna | 8.84<sup>a</sup> | 11.9<sup>ef</sup> | 29.0<sup>d</sup> | 29.2<sup>-i</sup> | 58.0<sup>d</sup> | 3.2<sup>h</sup> | 93.3<sup>cd</sup> | 332<sup>d</sup> | 83.2<sup>h</sup> | 144<sup>de</sup> | 2.6<sup>de</sup>-<sup>ij</sup> | 167<sup>lg</sup> | 1267<sup>h</sup> |
| NS Todorka | 7.70<sup>bc</sup> | 13.9<sup>a</sup> | 38.8<sup>g</sup> | 41.7<sup>a</sup> | 61.2<sup>a</sup> | 4.8<sup>c</sup> | 85.0<sup>g-z</sup> | 275<sup>ef</sup> | 100.9<sup>de</sup> | 173<sup>a</sup> | 1.9<sup>j</sup> | 217<sup>bd</sup> | 1367<sup>a-d</sup> |

| Years |  |  |  |  |  |  |  |  |  |  |  |  |  |
|-------|---|---|---|---|---|---|---|---|---|---|---|---|
| 2016/17 | 7.39<sup>c</sup> | 13.0<sup>b</sup> | 33.3<sup>a</sup> | 34.1<sup>a</sup> | 57.2<sup>a</sup> | 4.8<sup>a</sup> | 107.5<sup>a</sup> | 158<sup>b</sup> | 78.4<sup>c</sup> | 159<sup>a</sup> | 2.0<sup>c</sup> | 182<sup>a</sup> | 1441<sup>a</sup> |
| 2017/18 | 8.24<sup>a</sup> | 11.6<sup>c</sup> | 26.0<sup>e</sup> | 27.1<sup>c</sup> | 55.9<sup>g</sup> | 2.4<sup>c</sup> | 79.4<sup>a</sup> | 317<sup>b</sup> | 122.3<sup>a</sup> | 137<sup>c</sup> | 3.7<sup>a</sup> | 174<sup>c</sup> | 1255<sup>b</sup> |
| 2018/19 | 7.68<sup>b</sup> | 13.8<sup>a</sup> | 34.3<sup>a</sup> | 33.4<sup>bc</sup> | 59.1<sup>a</sup> | 4.9<sup>a</sup> | 62.8<sup>c</sup> | 434<sup>a</sup> | 95.2<sup>b</sup> | 146<sup>a</sup> | 2.5<sup>a</sup> | 226<sup>a</sup> | 1269<sup>b</sup> |

Means with at least one common letter were not significantly different at P = 0.05 by the Tukey’s test.
Table 5. Correlation matrix of 18 wheat cultivars for studied quality traits in three growing seasons.

|     | GS 2017 | PC 2017 | SG 2018 | SV 2017 | WG 2018 | GA 2017 | PC 2018 | SG 2018 | WG 2018 |
|-----|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| GS 2017 | 0.675** | 0.718** | 0.597** | 0.597** |
| GS 2018 | 0.592** | 0.550** | 0.601** | 0.607** |
| WA 2018 | 0.278 | 0.033 | -0.473’| 0.014 | 0.049 | -0.110 | 0.050 | -0.531’|
| WA 2019 | 0.580’ | 0.359 | 0.041 | 0.356 | 0.230 | 0.563’|
| E 2018 | 0.671** | 0.478’ | 0.283 | 0.014 | 0.049 | -0.110 | 0.050 | -0.531’|
| E 2019 | 0.542’ | 0.657** | 0.466 | 0.298 | 0.681** | 0.214 |
| BV 2018 | 0.537’ | 0.336 | 0.058 | 0.411 | 0.065 | 0.150 | 0.576 |
| BV 2019 | 0.357’ | 0.417 | -0.058 | 0.150 | 0.106 | 0.576 |

GS – growing season; PC – protein content; SV – sedimentation value; WG – wet gluten content; WA – water absorption; FR – farinograph resistance; SD – softening degree; E – extensograph energy; EE – extensibility; R/EE – resistance/extensibility ratio; BV – bread volume; FN – falling number; W – alveograph deformation energy; *, ** Significant at the 0.05 and 0.01 probability level, respectively.

Table 6. Correlation between studied quality traits of 18 wheat cultivars in averaged across growing seasons.

|     | PC 2017 | SV 2017 | WG 2017 | WA 2017 | FR 2017 | SD 2017 | E 2017 | EE 2017 | R/EE 2017 | BV 2017 | FN 2017 | W 2017 |
|-----|---------|---------|---------|---------|---------|---------|--------|---------|-----------|---------|---------|--------|
| SV 2017 | 0.801** | 0.895** | 0.836** | 0.841** | 0.805** | 0.796** | 0.600** | 0.492’ | -0.014 | 0.049 | -0.110 | 0.050 | -0.531’|
| WG 2017 | 0.812’ | 0.702’ | 0.745’ | 0.811’ | 0.794’ | 0.478’ | 0.007’ | 0.489 | 0.772’ |
| WA 2017 | 0.692’ | 0.682’ | 0.670’ | 0.628’ | 0.114 | 0.121 | 0.772’ |
| FR 2017 | 0.923’ | 0.811’ | 0.794’ | 0.478’ | 0.007’ | 0.489 |
| SD 2017 | -0.014 | 0.049 | -0.110 | 0.050 | -0.531’ | 0.772’ |
| E 2018 | 0.240 | 0.492 | 0.082 | 0.265 | 0.022 | 0.061 |
| EE 2018 | 0.776’ | 0.923’ | 0.811’ | 0.794’ | 0.478’ | 0.007’ | 0.489 |
| R/EE 2018 | 0.741’ | 0.692’ | 0.882’ | 0.670’ | 0.628’ | 0.114 | 0.121 | 0.772’ |
| BV 2018 | 0.889’ | 0.695’ | 0.824’ | 0.758’ | 0.661’ | 0.269 | 0.246 | 0.784’ | 0.796’ |
| FN 2018 | -0.275 | -0.292 | -0.154 | -0.132 | -0.232 | 0.379 | 0.072 | -0.113 | 0.124 | -0.085 | 0.885’ |
| W 2018 | 0.825’ | 0.723’ | 0.870’ | 0.655’ | 0.642’ | 0.273 | 0.197 | 0.723’ | 0.752’ | 0.063 | 0.168 | 0.481’ | 0.201 | 0.397 |

PC – protein content; SV – sedimentation value; WG – wet gluten content; WA – water absorption; FR – farinograph resistance; SD – softening degree; E – extensograph energy; EE – extensibility; R/EE – resistance/extensibility ratio; BV – bread volume; FN – falling number; W – alveograph deformation energy; GY – grain yield; *, ** Significant at the 0.05 and 0.01 probability level, respectively.
Figure 1. Average monthly rainfall and temperature during pre-anthesis developmental phases in three growing seasons.

Figure 2. Average daily temperatures during three growing seasons (A) and daily rainfall in 2016-17 (B), 2017-18 (C) and 2018-19 (D) after flowering.