European Cereals Genetics Co-operative Newsletter 2019

Proceedings of the 17th International EWAC Conference

3 - 8 June 2018

Bucharest, Romania





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Leibniz-Institut für Pflanzengenetik und Kulturpflanzenforschung, Gatersleben, Germany

and

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Edited by

A. Börner and M. Ciucă

Leibniz-Institut für Pflanzengenetik und Kulturpflanzenforschung, Seeland/OT Gatersleben, Germany and National Agricultural Research and Development Institute, Fundulea, Romania

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Preface

A. Börner

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The 17th EWAC International Conference was organised by Matilda Ciucă and her colleagues in Bucharest, Romania from June 3 – 8, 2018. The organizing bodies were the National Agriculture Research and Development Institute, Fundulea and Ministry of Agriculture and Rural Development by the ADER116 project. It was the first EWAC conference in Romania since it was founded in 1967 (Table 1).

Table 1: Years and venues of EWAC Conferences.

1		
1967	Cambridge	UK
1970	Weihenstephan	Germany
1974	Novi Sad	Yugoslavia
1979	Cambridge	UK
1981	Wageningen	The Netherlands
1984	Versailles	France
1987	Martonvasar	Hungary
1991	Cordoba	Spain
1994	Gatersleben	Germany
1997	Viterbo	Italy
2000	Novosibirsk	Russia
2002	Norwich	UK
2005	Prague	Czech Republic
2007	Istanbul	Turkey
2011	Novi Sad	Serbia
2015	Lublin	Poland
2018	Bucharest	Romania

Since 2016 EWAC is a working group 'Cereals Genetic Stocks' of the Cereals Section of the 'European Association for Research on Plant Breeding' (EUCARPIA). Fifty-five participants from 12 countries did attend the conference comprising 22 lectures and 15 poster presentations.

Under the general Motto 'Cereals for Tomorrow' two main subjects were discussed:

- Genetic gains through novel diversity and tools
- New approaches for cereals improvement and the future contribution of genetic stocks

The scientific programme but also the local organisation of the conference were excellent. Many thanks to Matilda Ciucă and her team for preparing and running this successful conference in a very kind and friendly atmosphere. We did enjoy the days in Bucharest and Fundulea very much.

Just before the conference Elena Khlestkina offered to organise the next EWAC Conference at the Vavilov Research Institute of Plant Genetic Resources in St. Petersburg in 2021.

We are looking forward to the 18th EWAC Conference.

Phenotypic and molecular variability of Serbian and Austrian winter wheat varieties

S. Mikić¹, A. Kondić-Špika¹, D. Trkulja¹, M. Mirosavljević¹, V. Takač¹, N. Buha¹, H. Grausgruber²

Summary

Genetic variability of locally adapted Serbian and Austrian winter wheat varieties was evaluated in order to assess their potential as a genetic material that can be exploited in crosses between two different European breeding pools. A field trial with 20 elite wheat varieties from each country was set at the Institute of Field and Vegetable Crops, Novi Sad, Serbia in a row-column design with three replications during the 2016/2017 season. The genotypes were phenotyped for tillering, heading and flowering time, plant height, ear length, number of spikelets per spike, number of grains per spike, thousand-kernel weight, chlorophyll content and resistance to prevalent wheat diseases. Additionally, the varieties were genotyped with 30 microsatellites. The varieties from two geographic regions and different release periods were clearly differentiated with population structure obtained from marker data. A significant phenotypic variation was found for most of the traits. Coefficients of variation were the largest for chlorophyll content (16.5%) and plant height (10.1%). Generally, the early genotypes were more susceptible to leaf rust (r = -0.6), while the late maturing genotypes produced more grains per spike (r = 0.4). The Serbian varieties had earlier tillering (p < 0.03), heading (p < 0.00) and flowering (p < 0.00) dates, shorter plant stems (p < 0.00), higher chlorophyll content (p < 0.00) and were more susceptible to leaf rust (p < 0.00) than the Austrian ones. The principal component analysis indicated general properties of the groups that would facilitate the choice of parent combinations for crossings.

Introduction

The effects of climate change, such as extremely high temperatures, low relative humidity, uneven rainfall distributions, strong insolation and droughts, are becoming very frequent in Serbia (Gocic & Trajkovic 2013). Similarly, warmer and dry summers are observed in plain terrains of the western parts of Austria (Alexandrov et al. 2002). A natural mechanism of plants to respond to changing environments, known as phenotypic plasticity, can be used to tackle climate change. Phenotypic plasticity is the ability of a genotype to express different variations of a trait when environmental conditions change or when a genotype is grown in a range of differing environments (Fusco & Minelli 2010). It is believed that the genotypes with high phenotypic plasticity can better adapt to different conditions and expand to diverse environments. Estimating plasticity and genetic variability of important wheat agronomic traits may help breeders to identify cultivars that are more suitable for production in less favourable, risk-prone environments and to distinguish them from cultivars suitable for production in optimal, non-stressed environments, as well as to define breeding strategies to broaden genetic diversity of elite wheat varieties from different regions. The geographical specificities of Serbia and Austria make them distinct and characteristic testing environments for field trials. The aims of this study were: 1) to assess genetic variability of locally adapted elite Serbian and Austrian winter wheat varieties, 2) to evaluate the most important morphological, phenological and agronomic characteristics of the selected varieties, and 3) to assess their potential that can be

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exploited in crosses between two different European breeding pools.

Materials and methods

The field trial was set at the Institute of Field and Vegetable Crops, Rimski sancevi (45°20' N, 19°51' E, 84 m a.s.l.), Serbia, in a row-column design with three replications during the season 2016/2017. In total, 40 elite winter wheat varieties were selected, 20 from Austria and 20 from Serbia together with two check varieties, Bezostava 1 and Amadeus. The phenotypic evaluation was performed for ten morphological, phenological and agronomic yield-related traits, namely tillering time (days from sowing), heading time (days from sowing), flowering time (days from sowing), plant height (cm), ear length (cm), number of spikelets per spike, number of grains per spike, thousand-kernel weight (g), chlorophyll content (CCI = % transmittance at 931nm. / % transmittance at 653 nm) at flowering and resistance to prevalent diseases. Genomic DNA was extracted from the seedlings using CTAB protocol. The genotyping was done with 30 microsatellite markers (Table 1). Total PCR mix contained 25 ng genomic DNA, 0.2 mM dNTP, 1×Taq buffer with KCl, 2 mM MgCl2, 1 U Taq polymerase, 0.5 pmol of fluorescently labelled forward primer and 0.5 pmol of reverse primer. PCR began with DNA denaturation at 94 °C for 5 min, followed by 38 cycles at 94 °C for 30 s, 52-62 °C for 45 s, 72 °C for 45 s and the final extension for 7 min at 72 °C. The 10 µL reaction volume for fragment analysis contained: 2 µL of differently labelled PCR products mixture, 0.2 µL GeneScan500 LIZ size standard and 7.8 µL Hi-Di formamide. The PCR products were separated by capillary electrophoresis on ABI Prism 3130 and their sizes were determined with Gene Mapper Software Version 4.0 (Applied Biosystems).

Table 1: Names, chromosome positions and repeat motifs of 30 analysed SSR markers.

No	SSR	Chr.	Repeat	No	SSR	Chr.	Repeat
1	barc102	3B	(TAA)20	16	gwm413	1B	(GA)18
2	barc1096	4B	(CT)10	17	gwm458	1D	(CA)13
3	barc110	5B	(ATT)28	18	gwm495	4B	(GA)20
4	barc 187	1B	(CT)26	19	gwm513	4B	(CA)12
5	barc3	6A	(CCT)17	20	gwm577	7B	(CA)14(TA) 6
6	cfa2149	4B, 5A	(TG)20	21	gwm636	2A	(GA)28imp
7	gdm63	5D	(CT)20	22	gwm639	5A, 5B, 5D	(GA)19
8	gpw3017	4B	(GA)33	23	wmc125	4B	(GT)11(GT) 20
9	gwm261	2D	(CT)21	24	wmc14	7D	(CT) (CA)
10	gwm291	5A	(CA)35	25	wmc25	2B, 2D	(GT)26
11	gwm296	2A, 2D, 7D	(CT)28	26	wmc262	4A	(GA)29
12	gwm325	6D	(CT)16	27	wmc317	2B	(GT)23
13	gwm350	7A, 7D	(GT)14	28	wmc410	5A	(CA)26
14	gwm371	5B, 5D	(CA)10(GA)32	29	wmc601	2D	-
15	gwm408	5B	(CA)22(TA)(CA)7(TA)9	30	wmc617	4A, 4B, 4D	-

The molecular diversity parameters were analysed in GenAlEx 6.5. Population structure was calculated using model-based clustering method based on parametric model of frequency distribution with unknown number of subpopulations integrated into STRUCTURE software. The analysis of molecular variance (AMOVA) was applied to partition genetic variation among

groups based on the codominant allelic distance matrix. The phenotypic data were used to perform analysis of variance and principal component analysis.

Results and discussions

In total, 209 alleles were detected in 30 SSR loci with the mean number of 6.4 alleles per locus. The proportion of heterozygous individuals was 1%. The average PIC value was 0.61, while twelve microsatellites had PIC values above 0.70, indicating relatively high discriminatory power of microsatellite markers. In previous studies, similar PIC values of 0.64 were obtained with 39 SSRs (Roussel et al. 2005) and 0.67 with 19 SSRs (Röder et al. 2002) for a larger number of European wheat varieties. The smaller number of detected alleles and the average number of alleles per locus obtained in our study comparing to the previous ones, was due to much smaller sample size and narrower time span of variety release periods.

Population structure analysis performed with the programme Structure divided the genotypes distinctly by the country and the average year of release (Figure 1). Although the two Austrian groups were closer in terms of their average release periods (8 years difference), than the two Serbian (16 years difference), the groups were denoted as Austrian older with 11 varieties, Austrian new with 9 varieties, Serbian older with 14 varieties, and Serbian new with 6 varieties. El-Esawi et al. (2018) also showed an importance of geographic origin in a wheat diversity study, finding significant difference between western and central European winter wheat variety groups.

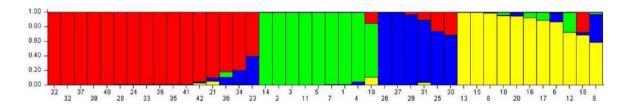


Fig. 1: Population structure of 42 wheat varieties estimated with SSRs. Red - Serbian older, green - Austrian new, blue - Serbian new, yellow - Austrian older varieties.

Analysis of molecular variance showed that genetic variation was much higher within the groups, accounting for 80%, than the variance among the groups (20%). Similar distribution of variance, where the majority of the diversity was attributed to differences among varieties within populations was obtained in Austrian and Belgian breeding pools (El-Esawi et al. 2018). The varieties were more differentiated by their geographical origin than by their release period, which is in accordance with the finding of Roussel et al. (2005), who demonstrated significant geographical variation between the wheat varieties from western and south-eastern European countries and, to less extent, temporal variation among the wheat varieties from different breeding periods.

A significant phenotypic variation was found for most of the traits (Table 2). Coefficients of variation were the largest for chlorophyll content (16.5%) and plant height (10.1%). Tukey's honest significant difference tests were used for multiple comparisons of means. The Serbian varieties had earlier tillering, heading and flowering dates than the Austrian. It is worth noting that while there were no differences in heading and flowering between the old and new Serbian varieties, old and new Austrian varieties significantly differed. Heading and flowering dates of the new Austrian varieties were recorded earlier than in old Austrian varieties, indicating a shift

in breeding towards earlier genotypes. This shift in wheat breeding has been observed in countries with frequent terminal drought as a strategy to minimize of the risk of drought stress (Shavrukov et al. 2017).

Table 2: Analysis of variance and comparison of means for nine phenotypic traits of four groups of winter wheat.

Group	Al	A2	S1	S2	Mean	CV (%)
Tillering	122.4 a	122.4 a	119.9 b	119.8 b	121.1	1.1
Heading	183.9 a	180.8 b	176.5 c	177.6 c	179.5	2.0
Flowering	186.2 a	183.6 b	180.6 c	181.2 c	182.7	1.5
Ear length	10.5 a	10.2 a	9.7 a	10.5 a	10.1	9.7
Plant height	77.2 a	76.4 a	67.5 b	67.2 b	72.4	10.1
Chlorophyll index	28.8 b	32.1 b	38.2 a	31.4 b	33.4	16.5
Grains per spike	51.0 a	51.8 a	49.0 a	53.0 a	51.2	6.2
Spikelets per spike	19.3 a	19.4 a	19.4 a	20.2 a	19.6	8.4
Thousand-kernel weight	48.7 a	48.5 a	48.2 a	48.8 a	48.5	5.9

A1 - Austrian older, A2 - Austrian new, S1 Serbian older, S2- Serbian new varieties

No significant differences among the groups were found for the average values of ear length, number of grains per spike, number of spikelets per spike and thousand-kernel weight. On average, the Austrian genotypes were higher than the Serbian ones, but there were no differences between the older and new varieties. The old Serbian varieties had significantly higher chlorophyll content index at the flowering then the other groups. This is the only trait showing differences between two groups of Serbian varieties.

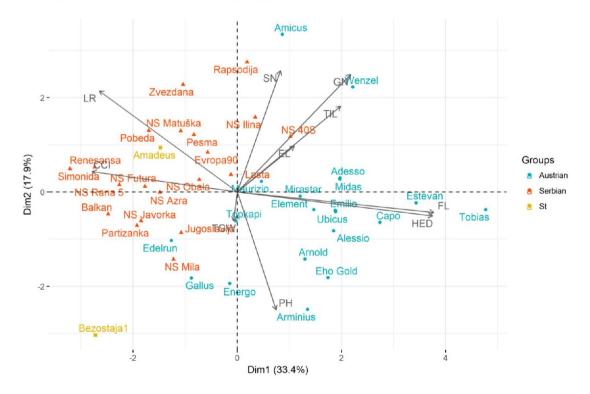


Fig. 2: Principal component analysis biplot of Austrian and Serbian winter wheat varieties.

The principal component analysis differentiated the varieties from Serbia and Austria (Fig. 2). The Austrian varieties were in general later maturing and higher than the Serbian, grouping around the vector for flowering and heading. On the other hand, Serbian varieties were more susceptible to rust and had higher chlorophyll content. The Austrian and Serbian varieties did not differ in the number of spikelets, number of grains per spike or thousand-kernel weight. The PCA biplot showed positive correlations between flowering date and number of grains per spike, as corroborated with the Pearson's coefficient (r = 0.4; p < 0.01). Pearson's correlations among the traits showed that the early genotypes were more susceptible to leaf rust (r = -0.6; p < 0.0001) and had higher chlorophyll content (r = -0.5; p < 0.001) than the late ones. In other studies, a positive significant correlation (Neumann et al. 2011) and no correlation between leaf rust and flowering time were found (Gao et al. 2016). Since rust infections before or at the flowering time of cereals are most damaging (Agrios 2005), it seems that early seasonal occurrence of the disease in 2017 concurred with flowering stage of early genotypes, causing severer symptoms in early that in late genotypes. The integration of data from the trial in Austrian environment and from two experimental years is expected to give a more detailed insight into phenotypic and molecular variability of the analysed wheat varieties and facilitate the choice of parent combinations for crossings.

Acknowledgements

This research was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia (project TR-31066).

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