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in

Molina-Cano J.L. (ed.), Christou P. (ed.), Graner A. (ed.), Hammer K. (ed.), Jouve N. (ed.), Keller B. (ed.), Lasa J.M. (ed.), Powell W. (ed.), Royo C. (ed.), Shewry P. (ed.), Stanca A.M. (ed.).
Cereal science and technology for feeding ten billion people: genomics era and beyond

Zaragoza : CIHEAM / IRTA
Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 81

2008
pages 67-69

Landjeva S., Korzun V., Ganeva G. **Temporal trends in the microsatellite-based genetic diversity of 91 bread wheat varieties released in Bulgaria since 1925.** In : Molina-Cano J.L. (ed.), Christou P. (ed.), Graner A. (ed.), Hammer K. (ed.), Jouve N. (ed.), Keller B. (ed.), Lasa J.M. (ed.), Powell W. (ed.), Royo C. (ed.), Shewry P. (ed.), Stanca A.M. (ed.). *Cereal science and technology for feeding ten billion people: genomics era and beyond.* Zaragoza : CIHEAM / IRTA, 2008. p. 67-69 (Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 81)

Temporal trends in the microsatellite-based genetic diversity of 91 bread wheat varieties released in Bulgaria since 1925[1]

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SUMMARY – Patterns of the molecular genetic diversity changes within groups of old and modern Bulgarian bread wheat varieties were analyzed using 19 wheat microsatellite markers and one secalin-specific sequence tagged site marker. A total of 91 genotypes representing almost the complete spectrum of varieties released in Bulgaria during the period 1925-2003 were included in the study. Analysis of microsatellite allele polymorphisms revealed quantitative (different allele frequency) and qualitative (loss of alleles and/or appearance of novel alleles) changes in genetic diversity over decades of variety release. Genetic diversity was high among both old and modern varieties with an average of 0.65. An average heterogeneity of 10.1% was established, this being highest in the old varieties. The genetic diversity was affected by the interaction between the breeding centre and the applied breeding strategy directed towards increasing the productivity or grain quality improvement.

Public concern over potential decline in crop diversity and the necessity of developing appropriate science-based breeding strategy motivated the assessment of current levels of and temporal trends in genetic diversity of a number of crops, including bread wheat (Donini *et al*., 2000; Chebotar *et al*., 2003; Khlestkina *et al.,* 2004). Quantitative estimation of the genetic diversity and characterization of diversity trends over time is fundamental for the efficient conservation, maintenance and rational utilization of genetic resources. The present study is an extended analysis of dimensions of genetic diversity and temporal trends in the diversity changes over periods of variety release among Bulgarian bread wheat germplasm as revealed by microsatellites.

The experimental material included 91 winter bread wheat (*Triticum aestivum* L.) varieties released in Bulgaria during the period 1925-2003 at the main breeding centres: Dobrudzha Agricultural Institute (DAI), General Toshevo; Institute of Plant Genetic Resources (IPGR), Sadovo, and Institute of Genetics (IG), Sofia. A total of 19 wheat microsatellite markers covering 17 chromosomes and one secalin-specific STS-marker for rye chromosome arm 1RS were used. A modification of the PCR protocol by Röder *et al.* (1998) was followed. The patterns of genetic variation were analyzed using Nei's measure of genetic diversity (Nei, 1987).

Microsatellite alleles polymorphisms

Analysis of microsatellite allele polymorphisms revealed quantitative and qualitative changes in the number of alleles per locus and the favoured alleles within the groups of old and modern varieties registered before 1960 and afterwards, respectively (Table 1). For six markers the dominating alleles have been maintained during the whole period under investigation, suggesting their importance and selective advantage. For six other, the prevailing allele among the sub-groups of modern releases is different in comparison to the early years of breeding. For the rest, the alleles which dominate within the group of old varieties prevail in the modern varieties at random. The total number of alleles detected in varieties from each period shows changes over time with the highest number of alleles found in varieties registered after 1990, and the least number of alleles present in 1960s-varieties. Four different patterns of changes in the number of alleles per locus over decades of variety release were recorded. Alleles detected with markers WMS46, WMS155, WMS389, WMS577 and WMS631 displayed a random pattern of change over time, meaning that alleles found in the early years of wheat breeding were randomly present in those registered later on. For markers WMS165, WMS357 and WMS458, a shifting pattern of predominance in two or three of their alleles was displayed. Seven

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[1] The partial support of the Bulgarian NSF (contract B-1538/05) is acknowledged.

markers (WMS18, WMS190, WMS408, WMS437, WMS513, MW1B002, and SR1R001) showed an increasing number of alleles. Two other markers displayed a pattern of decreasing number of alleles in modern varieties. This is the case for marker WMS261, for example, which is in tight linkage with the region on chromosome arm 2DS where the semi-dwarfing gene *Rht*8 and the *Ppd*-D1 gene determining response to day length are located (Korzun *et al*., 1998). The decrease in the number of WMS261 alleles is associated with the intensive introduction of semi-dwarfing genes into Bulgarian wheat germplasm after 1960 aiming to reduce final plant height and to increase the yield (Ganeva *et al*., 2005).

Table 1. Microsatellite alleles and their frequencies among Bulgarian bread wheat varieties, released in the period 1925-2003

Marker	Chromosome	Total no. of alleles	Range of alleles size (bp)	Dominating allele (allele frequency)				
				Old varieties	Modern varieties released during:			
					1960s	1970s	1980s	1990s
SR1R003	1RS	2	Null, 97	Null (1.00)	Null (0.67)	Null (0.90)	Null (0.74)	Null (0.63)
MW1A001	1A	9	Null, 131-154	137 (0.50)	137 (0.67) 140 (0.67)	140 (0.85)	140 (0.63) 143 (0.63)	140 (0.77)
MW1B002	1B	9	Null, 212-268	235 (0.50)	214 (0.33) 235 (0.33) 268 (0.33)	214 (0.60)	214 (0.47)	235 (0.37)
WMS003	3DL	5	74-82	76 (0.62)	76 (0.67)	76 (0.70)	76 (0.89)	76 (0.71)
WMS0018	1BS	8	Null, 183-197	187 (0.57)	183 (0.33) 185 (0.33) 187 (0.33)	185 (0.45)	185 (0.53)	189 (0.34)
WMS0046	7BS	10	145-179	173 (0.29)	173 (0.67)	173 (0.65)	173 (0.63)	173 (0.49)
WMS0095	2AS	6	106-124	118 (0.79)	116 (0.33) 118 (0.33) 120 (0.33)	118 (0.45) 120 (0.45)	120 (0.58)	120 (0.60)
WMS0155	3AL	6	126-148	142 (0.43) 146 (0.43)	146 (0.67)	140 (0.55)	142 (0.37)	140 (0.37)
WMS0165	4AS	6	Null, 182-194	192 (0.93)	Null (0.67) 192 (0.67)	192 (0.90)	192 (0.95)	192 (0.86)
WMS0165	4DL	3	196-200	196 (0.71)	196 (0.67)	196 (0.5)	196 (0.53)	196 (0.54)
WMS0190	5DS	6	197-212	210 (0.86)	210 (1.00)	210 (0.95)	210 (0.68)	210 (0.77)
WMS0261	2DS	6	165-215	174 (0.64)	192 (0.67)	192 (1.0)	192 (0.95)	192 (0.77)
WMS0357	1AL	4	120-126	122 (0.79)	120 (0.67)	120 (0.65)	122 (0.580	120 (0.54)
WMS0389	3B	7	Null, 116-138	132 (0.43)	118 (0.67)	Null (0.45)	Null (0.37) 118 (0.37)	118 (0.40)
WMS0408	5BL	8	147-193	147 (0.50)	179 (0.67)	179 (0.70)	179 (0.53)	179 (0.60)
WMS0437	7DL	11	89-130	113 (0.43)	89 (0.33) 115 (0.33) 120 (0.33)	89 (0.55)	89 (0.37) 115 (0.37)	89 (0.23)
WMS0458	1DL	3	108-112	110 (0.64) 112 (0.64)	110 (1.00)	110 (0.75)	110 (0.74)	110 (0.63)
WMS0513	4BL	5	141-151	143 (0.79)	143 (0.67)	143 (0.6)	143 (0.58)	143 (0.43)
WMS0577	7BL	10	Null, 129-167	158 (0.29) 167 (0.29)	Null (0.67)	163 (0.50)	163 (0.53)	163 (0.57)
WMS0631	7A	6	187-211	187 (0.29)	189 (0.67)	189 (0.60) 197 (0.60)	197 (0.68)	189 (0.66)
WMS0680	6B	6	108-136	132 (0.43)	132 (0.67)	122 (0.85)	122 (0.74)	122 (0.77)

Genetic diversity and heterogeneity

The genetic diversity as revealed by microsatellites was high within both old and modern varieties with an average of 0.65. A non-significant trend for diversity decrease, especially in 1970s, was observed, after which its value increased gradually to reach the levels of 0.71 in 1990s. For some markers, more than one allele was identified in a single variety, suggesting the existence of internal heterogeneity. Considering all variety/marker combinations, an average level of 10.1% of heterogeneity was established. A high proportion of the studied varieties were heterogeneous with respect to one (31%) or two (17%) markers, 26% were non-uniform with more than two markers, and only 26% showed homogeneity with all markers. These results generally agree with those obtained in a survey of over 500 varieties by Röder *et al.* (2002), who found the highest levels of heterogeneity for the South-Eastern European germplasm. Patterns of genetic diversity were further subjected to analysis of variance. No significant differences were observed in diversity with respect to the period of release, breeding centre, genomes, and their interactions. The impacts of breeding strategy to the genetic diversity was investigated by comparing the diversity values for varieties released in the period 1961-1990, when emphasis was placed on breeding for high productivity, and for varieties released after 1990, when efforts have been directed primarily towards improvement of grain quality. Although no significant differences were observed in the overall diversity due to breeding strategy, our data showed significant interactions between breeding centres and breeding strategy (F=9.51***), particularly between different centres during the first period (F=19.4***), and within centres between different breeding strategy periods (F=9.77** for IG; F=6.51* for DAI).

Genetic relationships

The hierarchical clusters were in good agreement with the origin of varieties and their pedigree information as far as was known. Three major groups were distinguished: Group 1 clusters the majority of old varieties. Group 2 encompasses the majority of varieties, developed at IPGR. In this group, two sub-clusters of the old and modern IPGR-derived varieties are further revealed. Group 3 clusters mostly DAI-derived varieties. The majority of 1RS.1BL varieties form a distinct sub-cluster, which, in turn, includes a sub-sub-cluster of varieties, developed at the IG.

The high genetic diversity levels established for Bulgarian varieties isolated before 1960 have been maintained almost unchanged afterwards. The high yield, earliness, resistance to stem lodging and rusts, tolerance to drought and low winter temperatures were among the most important characteristics, which have been genetically improved by breeders after 1960s. Further breeding activities aimed at achieving optimal combination of high productivity and ecological plasticity to guarantee sustainable yields under different conditions (Panayotov *et al*., 1994). The transition in 1990s to a new strategy which was focused on the improvement of grain quality (Panayotov, 2000) has been an important driver of additional diversity increase. The observed tendency towards enhancement of genetic diversity in 1990s is mainly due to the utilization of new diverse germplasm to introduce gibberellin-insensitive height-reducing genes (Ganeva *et al.,* 2005) and to achieve optimal combination between high productivity and high bread-making quality (Panayotov, 2000).

References

Chebotar, S., Röder, M., Börner, A. and Sivolap, Yu. (2003). *Proc. 10th Wheat Genet. Symp*., Paestum (Italy).

Donini, P., Law, J.R., Koebner, R.M.D., Reeves, J.C. and Cooke, R.J. (2000). *Theor. Appl. Genet*., 100: 912-917.

Ganeva, G., Korzun, V., Landjeva, S., Tsenov, N. and Atanasova, M. (2005). *Euphytica*, 145(3): 307-317.

Khlestkina, E.K., Röder, M.S., Efremova, T.T., Börner, A. and Shumny, V.K. (2004). *Plant Breed*., 123: 122-127.

Korzun, V., Röder, M., Ganal, M., Worland, A.J. and Law, C.N. (1998). *Theor. Appl. Genet*., 96: 1104-1109.

Nei, M. (1987). *Molecular Evolutionary Genetics.* Columbia University Press, New York.

Panayotov, I. (2000). *Bulg. J. Agric. Sci*., 6(5): 513-523.

Panayotov, I., Boyadjieva, D., Todorov, I., Stankov, I., Dechev, D. and Tsvetkov, S. (1994). *Rasteniev. Nauki*, 31(3-4): 48-57 (In Bulgarian).

Röder, M., Korzun, V., Wendehake, K., Plaschke, J., Tixier, M.-H., Leroy, P. and Ganal, M. (1998). Genetics, 149: 2007-2023.

Röder, M., Wendehake, K., Korzun, V., Bredemeijer, G., Laborie, D., Bertrand, L., Isaac, P., Rendell, S., Jackson, J., Cooke, R., Vosman, B. and Ganal, M. (2002). *Theor. Appl. Genet*., 106: 67-73.