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PREFERENCE OF MOLECULAR MARKERS FOR DETECTING ALLELIC VARIANTS OF GLIADINS

The main method of studying allelic variants of gliadins is electrophoresis in acidic PAGE, which actually shows the phenotype determined by the set of genes of the gliadin-encoding locus. This method is difficult to use, because peptides encoded by all gliadin-encoding loci are present on one lane of the gel and may overlap. Therefore, there is a need for molecular markers that would allow the determination of allelic variants of gliadins using PCR. This will help to use gliadins for marker-assisted selection, to identify allelic variants of gliadins even before obtaining grain suitable for electrophoresis of storage proteins. In this regard, the aim of study was to analyze polymorphism of *Gli-1* loci using allele-specific molecular markers and microsatellites and find suitable molecular markers for detecting allelic variants of gliadins by PCR method.

The collection of bread wheat cultivars from different countries, which reflects the maximum diversity of allelic variants of gliadins encoded by the *Gli-B1* locus (provided by Dr. E. Metakovsky) and collection of the modern Ukrainian wheat cultivars and lines from different breeding centers were analyzed using allele-specific primers to loci *Gli-A1*, *Gli-B1* and *Gli-D1* developed by Zhang et al. (2003) and *Taglgap* microsatellite marker described by Devos et al. (1995).

PCR with allele-specific primers to the *Gli-B1* locus permit to differentiate wheat genotypes with the *Gli-B1.1* or *Gli-B1.2* alleles, but we also revealed polymorphism within *Gli-B1.1* or *Gli-B1.2* alleles caused by a microsatellite within the amplified sequence that have been described Devos et al. (1995). Seven alleles were sequenced, and the presence of microsatellite with a CAA cor-motif which vary in number of repeats was confirmed. By using *Taglgap* microsatellite marker we revealed 12 alleles in the investigated wheat collections. The correspondence between "SNP allele of the *Gli-B1* locus – allele of the *Taglgap* microsatellite – allelic variant of gliadins" have been shown.

PCR analyze of the *Gli-A1* locus with two pairs of allele-specific primers developed by Zhang et al. (2003) permit to revealed *Gli-A1.1* and *Gli-A1.2* alleles which were of the same length. Based on the results obtained for the *Gli-A1* locus, correspondence between the *Gli-A1.1/Gli-A1.2* alleles and allelic variants of gliadins encoded by the *Gli-A1* locus was established. Bioinformatics analysis of the sequences coding

Gamma gliadin-A1 gene help to develop a pair of primers MsA1 to microsatellite in this locus. Eight different alleles that correspond to allelic variants of gliadins encoded by the *Gli-A1* locus were identified by using MsA1-primers.

Correspondence between alleles determined in PCR with allele-specific primers to the *Gli-D1* locus has not been established.

As a conclusion, we recommend to use *Taglgap* primers for detecting allelic variants of gliadins encoded at *Gli-B1* and new developed primers MsA1 for detecting allelic variants of gliadins encoded at *Gli-A1* locus.

References

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2. Devos K. M., Bryan G. J., Collins A. J., Stephenson P., & Gale M. D. (1995). Application of two microsatellite sequences in wheat storage proteins as molecular markers. *Theor. Appl. Genet*, 90, 247–252.