Breeding for breadmaking quality using HMW glutenin subunits in wheat (*Triticum aestivum* L.)

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The molecular markers can be used to facilitate the selection of chromosome segments including useful agronomic traits during the breeding process. These markers are particularly useful for incorporating genes that are highly affected by the environment, genes for resistance to diseases that cannot be easily screened, and to accumulate multiple genes for resistance to specific pathogens and pests within the same cultivar, a process called gene pyramiding. An additional advantage of the incorporation of MAS into breeding programs is that very different types of traits, e.g. a disease resistance gene or a genes predict quality wheat are identified. The MAS strategy is a way to capitalize on available markers and to incorporate valuable traits into elite lines that are suitable for cultivar release. MAS have the potential to facilitate the transfer of valuable genes identified in basic research programs into public wheat varieties.

The aims:

Applied – implement Marker Assisted Selection (MAS) in wheat breeding programes

Research – identify and study new alleles and develop new markers

I. MAS programme for pyramiding resistance genes

Leaf rust

Leaf rust, caused by *Puccinia triticina* is globally important fungal disease of wheat that cause significant annual yield losses. Growing resistant varieties is an efficient and economical method of reducing losses to leaf rust.

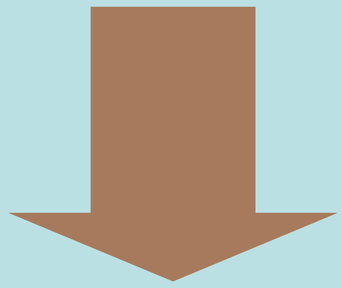
We have used MAS for the transfer and pyramiding of alien genes *Lr19*, *Lr24* and *Lr35* for leaf rust resistance to elite varieties.

Closely linked markers

STS marker linked to the gene *Lr19*, has been used for screening of plants possessing this gene.
The sequences of primers (CATCCTTGGGGACCTC - forward primer, CCAGCTCGCATACATCCA- reverse primer.
PCR-based DNA-STS marker, linked to the gene *Lr24: t*he sequences of primers TCTAGTCTGTACATGGGGGC - forward primer, TGGCACATGAACTCCATACG - reverse primer.
SCAR marker linked to the gene *Lr35*: the sequences of primers AGAGAGAGTAGAAGAGCTGC - forward primer, AGAGAGAGAGCATCCACC - reverse primer

The hundred plants are analysed by electroforesis methods for the present markers for genes *Lr19, Lr24* and *Lr35*

Segregation of DNA-STS specific marker linked to *Lr24* in BC_5 plants from the cross (Hana x Thatcher/*Lr24*)



R the resistant hybrid
P1 the susceptible recurent parent
P2 the resistant Thatcher /*Lr24*
as a donor *Lr24* gene

1 NIL Thatcher/*Lr35*
2 BC_3
3 Astella (elite cultivar)
(as a recurent parent)

II. MAS programme for quality of wheat

Glu alleles

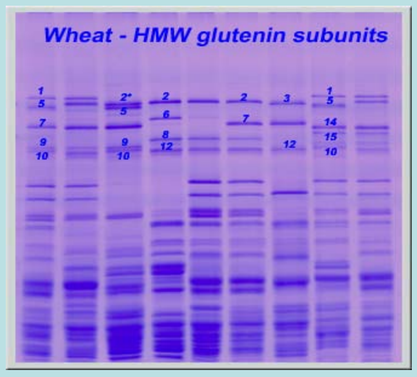
The analysis of seed storage proteins is a useful tool for plant breeding, due to their relationship with the technological properties of wheat. High molecular weight (HMW) glutenin subunits are wheat endosperm proteins arise from three loci (Glu-A1, Glu-B1, Glu-D1) located on the long arms of wheat chromosomes 1A, 1B and 1D. Several alleles, responsible for the production of diverse subunits, have been described for each locus. Variation in HMW glutenin subunit composition (e.g. allelic composition) contributes to genetic differences in breadmaking quality observed among varieties. The results from identification of HMW–GS alleles are useful for wheat-improvement programmes aimed at varietal identification and breeding for good quality parameters.

Direct markers

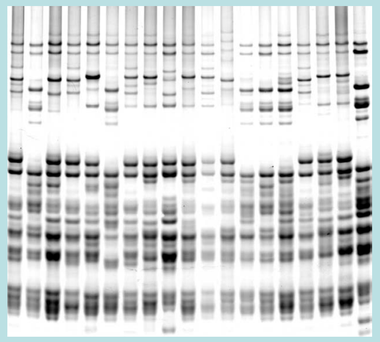
HMW-GS alleles (MAS) for calculate of *GLU – 1* quality score by SDS –PAGE and A-PAGE methods

Glu-1 quality score	Chromosome		
	1A	1B	1D
4	-	-	5+10
3	1	-	-
3	2*	-	-
3	-	17+18	-
3	-	7+8	-
3	-	13+16	-
2	-	7+9	-
2	-	-	2+12
1	null	-	-
1	-	7	-
1	-	6+8	-
1	-	20	-

SDS-PAGE



Glu-1 quality score between:	Points to be substracted due to 1BL/1RS presence	Rye-adjusted quality score
8-10	-3	5-7
5-7	-2	3-5
3-4	-1	2-3



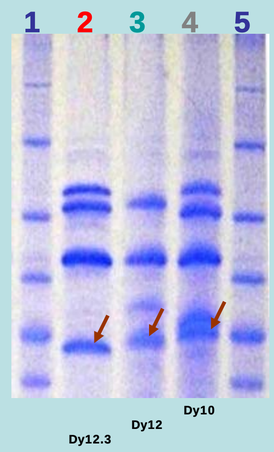
Identification of the New (unknown) HMW-GS alleles in wheat genotypes

I. New allele with electrophoretic mobility between HMW-GS 8 and 9 was detected in one of the lines of landrace **Eritrospermum 917**, located at the **loci *Glu-1B***.

II. New HMW-GS pair with electrophoretic mobility between HMW-GS 7 and 8 was detected in one of the lines of landrace **Kotte**, likely located at the loci ***Glu-1B*** and its relative molecular weights were calculated 104 kDa and 120 kDa.

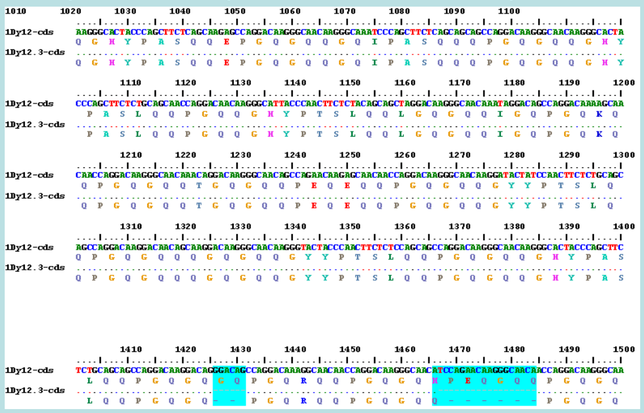
III. New allele at the loci ***Glu-1B***

IV. New allele at the loci ***Glu-1D*** in **Noe**



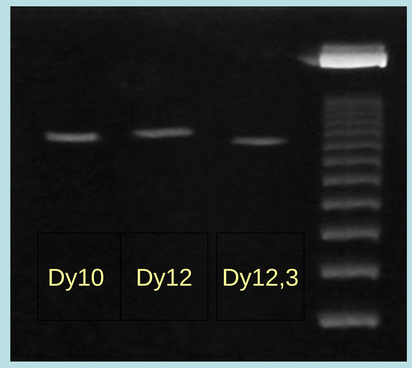
Verification of New HMW-GS allele at the loci *Glu-1D*

New HMW-GS allele at the loci ***Glu-1D***: Dy type HMW subunit protein (1Dy12.3) gene, complete cDNA (GeneBank EF472958)



III. Development of DNA marker (MAS)

MAS for
Dy10 =599 bp
Dy12 =605 bp
Dy12.3=581 bp



The application of MAS to improve wheat quality and enhance disease resistance will keep Slovak wheat competitive in the cultivar market