Identification of Molecular Markers for Resistance to Pre-Harvest Sprouting in *Triticum aestivum*

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**Abstract** Pre-harvest sprouting (PHS) represents a worldwide major problem in wheat production, because it causes huge loss of grain quality. PHS occurs when germination of grains takes place in the spike before harvest and it is influenced by high humidity.

The goal of this study was to find QTLs (quantitative trait loci) on chromosomal regions from Romanian wheat and the identification of specific markers that can be used for plant breeding in order to improve resistance to pre-harvest sprouting.

The parental lines with different sensibility to pre-harvest sprouting (Turda 95-sensitive, Turda 18-94-resistant, Lovrin 32-sensitive, Fundulea 29 – resistant) were used for the genotypic characterization using microsatellite markers.

A total of 119 from 640 SSR (simple sequence repeats) markers used on the parental lines indicated polymorphism. The initial primer test with the polymorphic markers on the cross lines: Turda 95(S) x Turda 18-94(R), Turda 95(S) x Fundulea 29 (R), Turda 18-94(R) x Lovrin 32 (S) showed that the cross between Turda 95(S) x Turda 18-94(R) had the highest number of segregating markers in the population. 111 F2 lines of the cross Turda 95(S) x Turda 18-94(R) now are being genotypic and phenotypic characterized in order to detect the QTLs underlying pre-harvest sprouting resistance.

**Keywords**: pre-harvest sprouting, QTL, SSR markers, wheat

**INTRODUCTION**

Wheat (*Triticum aestivum*) is a cereal with particular importance, given that bread made from wheat flour is the basic food for nearly half the world's population. Pre-harvest sprouting (PHS) (Fig.1) in winter wheat (*Triticum aestivum*) is a damage which causes losses in yield because of the premature germination of the seed while still in the head in field.

This damage causes the limits of end uses in the flour and substantial loss in yield and price (Mares et al. 2004). Pre-harvest sprouting in winter wheat represents a major problem in the world especially in places where the humidity is high in the time of maturity. Wheat quality in Romania is also affected by climatic factors.

Quantitative Trait Locus (QTL) mapping of Romanian winter wheat helps to improve by breeding the quality of this crop plant. QTL analysis it is a statistical method that links two types of information the phenotypic data (trait measurements) and genotypic data (with molecular markers in our case SSRs also called microsatellites markers).
The goals of the study are to find chromosomal regions carrying QTL and identify linked markers for plant breeding on Romanian wheat.

MATERIAL AND METHOD

The genotypic analysis was started with the parental testing of the four parental lines with different sensibility for pre-harvest sprouting (Turda 95-sensible, Turda 18-94-resistant, Lovrin 32-sensible and Fundulea 29-resistant) with SSR markers for polymorphism. The plant material was from the ARDS Turda, Romania and the research was made in Leibniz Institute of Plant Genetics and Crop Plant Research (IPK) Gatersleben, Germany.

The F2 population is composed by the three crossings between the four parental lines, Turda 95(S) x Turda 18-94(R), Turda 95(S) x Fundulea 29 (R), Turda 18-94(R) x Lovrin 32 (S).

DNA isolation from parental lines and F2 population was made from two week old leaves after Doyle and Doyle (1990) protocol.

The Polymerase Chain Reaction (PCR) was carrying out with SSR (Simple Sequence Repeats or Microsatellite) markers (gwm, wmc and barc) specific for wheat (Röder et al., 1998; Ganal and Röder, 2007) and a Viviparous-1 allelic variant primer (Vp1-B) (Yang et al. 2007). The fluorescent labeling of the left primer was recognized by the laser from the Automated Laser Fluorescent (ALF) fragment analyzer.

RESULTS AND DISCUSSIONS

Pre-harvest sprouting is largely studied in all countries of the world such as China (Chen et al., 2008), France (Groos et al., 2002), Germany (Lohwasser et al., 2002) or in the USA (Liu et al., 2008).

In the Romanian winter wheat it is a lack of information about the QTL (Quantitative Trait Locus) linked to pre harvest sprouting. The goal of this study is to identify QTLs responsible for pre harvest spouting (Fig. 1).

In QTL analysis in wheat it is necessary to use a high number of polymorphic primers to have the chance to find a QTL for pre-harvest sprouting. A total of 640 SSR markers where used for parental testing, from these markers 119 were polymorphic between the parental lines.

These primers will be use in the next genotypic studies on F2 population. The cross Turda 95 and Turda 18-94 had the highest percent of polymorphism (Tab.1) this means that the genetic difference between this two parental lines is the highest.

Tab.1

<table>
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<tr>
<th>The percent of polymorphism of the four parental lines</th>
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<td>Turda 95 x Turda 18-94</td>
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<td>14 %</td>
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CONCLUSION

These can be use for further genotypic analysis with the polymorphic markers and in direct phenotypic analysis.

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REFERENCES


Fig.1. Pre-harvest sprouting in wheat