

Microsatellite Markers for Improvement of Rice Blast Resistance

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Short Communication

Rice blast, caused by the pathogen *Pyricularia grisea*, is the primary limiting biotic factor for rice production throughout the world. The use of blast resistant cultivars is the most effective and economical way to control the disease. Therefore continuous breeding efforts to develop novel disease resistant cultivars should be the top priority for rice breeding programs. One of the major challenges which breeders face during the development of resistant cultivars is the incorporation of disease resistance gene into the target cultivar. The integration of plant genomics and gene-based marker assisted selection (MAS) being particularly effective, is a powerful method for efficient selection. Pyramiding several major genes for resistance into the genetic background of the recipient parent has been made easier with the help of marker-assisted selection.

Identifying resistance genes using molecular markers is the basic prerequisite for performing MAS in resistance breeding programs [1]. The use of molecular markers is still prohibitively expensive for most large-scale applications in rice breeding programs. Therefore, MAS methods are currently used for more targeted applications. Among all the biomolecular or DNA markers PCR-based microsatellite markers otherwise known as SSR markers (Simple Sequence Repeats) have attracted a great deal of attention by virtue of their key advantages such as hypervariability, high reproducibility and broad and uniform distribution throughout the rice genome. Furthermore, microsatellites are multi-allelic, highly polymorphic, abundant in the genome and cost effective. However, significant drawbacks do exist with respect to using microsatellite-based markers including unsuitability across species, high development costs and the significant effort required to design primer sets for a given study species.

Currently, there are no reliable estimates of the number of (AT)_n or (GC)_n sites in rice due to the difficulty of the hybridization-based screening methods used to detect these motifs. The size of the rice genome is ~ 0.45 × 10⁹ base pair which suggests that there should be one (AC)_n site approximately in every 360-450 kb and one (GA)_n motif every 225-330 kb in rice. 323 microsatellite markers identified by library screening and GeneBank searches of rice sequences have been localized on the rice genetic map. The (CGG)_n motif has been reported to be the abundant type in rice and is distributed throughout the genome. The degree of polymorphism shown by microsatellites ultimately determines their usefulness in genomic analyses.

Many Pi genes have been discovered that confer blast resistance to rice against a wide spectrum of pathotypes and it is often difficult to detect the presence of individual resistance genes and pyramid these in breeding lines. Biomolecular markers linked to many of the Pi genes have been localized on rice chromosomes, as well as markers for Pi-ta [2] and Pi-b [3]. Unfortunately,

majority of DNA markers for rice blast resistance are RFLPs, which are relatively labor intensive to analyze for use in breeding programs.

The gene *Pi-z*, which is present in the rice genotypes Zenith and Fukunishiki, represents a potential source of blast resistance for the northwestern Himalayan region of India. Genomic resources in rice and markers available in the public domain were exploited in order to develop a new set of microsatellite markers linked to *Pi-z* gene. Of the three reported markers for *Pi-z*, only MRG5836 was found to be suitable for MAS. A survey conducted for allelic diversity at the *Pi-z*-linked microsatellite marker loci revealed that the Fukunishiki and Zenith type alleles were not present in most of the *indica* rice genotypes. So these polymorphic markers between the *Pi-z* donors and majority of local *indica* rice strains can be used as selection tools in rice breeding programs aimed at improving blast resistance in local cultivars [4]. Three microsatellite markers were mapped at a distance of 0.0-11.5 cM from *Pi-z* in several different crosses segregating for the gene [5]. *Pi-z* has been reported to be allelic with, or at least closely linked to, three other blast resistance genes, Pi-2, *Pi-zt* and Pi-9, which map close to the centromere of chromosome 6 [6].

The *Pi20(t)* gene was identified from 160 Chinese *Magnaporthe oryzae* isolates, and among these, isolate 98095 can specifically differentiate the *Pi20(t)* gene present in cv. IR24. Two flanking and three co-segregating microsatellite markers for *Pi20(t)*, which is located near the centromeric region of chromosome 12, were identified using 526 highly susceptible F₂ plants derived from a cross between Asominori (highly susceptible) and IR24 (resistant). The microsatellite OSR32 was mapped at a distance of 0.2 cM from *Pi20(t)*, and the microsatellite RM28050 was mapped to the other side of *Pi20(t)* at a distance of 0.4 cM. The other three microsatellite markers, RM1337, RM5364 and RM7102, were observed to co-segregate with *Pi20(t)*.

In conclusion, molecular mapping of rice populations is a prerequisite to identify molecular markers tightly linked to the desirable Pi resistance gene. Microsatellite markers have turned out to be the most important and popular among the many type of molecular markers existing within the rice genome. They have played a pivotal role in the identification of numerous important genetic loci in different cereal crops. Microsatellite markers are also widely used in MAS programs to develop durably resistant cultivars against specific diseases. Microsatellites have been found to be highly polymorphic, genome-specific, and abundant and co-dominant and they have become important genetic markers in rice breeding programs for improving blast resistance in the recent years.

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