

Molecular Breeding for Durable Blast Disease Resistance in Rice

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Abstract

Rice blast disease caused by the fungus, *Magnaporthe grisea* is a serious threat to rice production in irrigated temperate rice as well as in the moisture stress condition of upland rice ecosystem. Blast resistance conferred by most of the identified genes is race specific. Resistance of high yielding *indica* and *japonica* cultivars breaks down within a short period of time because of variable virulence properties of *M. grisea*. Identification and use of major resistance genes conferring broad-spectrum durable resistance is one of the major rice breeding strategies to control the blast disease. A new blast resistance gene, *Pi40* was identified in an *indica* introgression line IR65482-4-136-2-2 that confers strong resistance to blast strains of South Korea compared to other known resistance genes like *Pi2*, *Piz-t*, *Pi9* and *Pi5*. The *Pi40* gene was located on the short arm of chromosome 6 and associated with the DNA marker, 9871.T7E2b. Using marker-assisted selection (MAS) strategy through backcross breeding the *Pi40* gene was introduced into two *japonica* cultivars. The advanced backcross breeding lines expressed resistance to blast strains of Korea as well as Philippines. Some advanced breeding lines with the *Pi40* gene and monogenic differential lines with other known resistance genes were tested against 29 field blast isolates of Korea by sequential planting method. Our results revealed strong resistance of the lines with *Pi40* gene to all the blast isolates except one during the seven cycles of evaluation indicating its potential for durable resistance to blast. The breeding lines possessing the *Pi40* gene have desirable agronomic traits and are useful for blast resistance breeding.

Media summary

Identification of a new blast resistance gene, *Pi40* and its incorporation into blast susceptible japonica rice cultivars using marker-assisted selection (MAS) will provide opportunity to develop broad-spectrum durable blast resistant cultivars.

Key Words

DNA marker, blast disease, durable resistance, marker-assisted selection, breeding, rice

Introduction

Rice production and productivity is significantly affected by several biotic stresses. Rice blast caused by the vicious fungal pathogen, *Magnaporthe grisea* affects rice production in temperate as well as tropical regions worldwide except Australia. Host plant resistance is the most economical and environment friendly approach to control this devastating disease of rice. There are about 40 major genes and several QTLs identified for blast resistance even though the genetics of blast resistance is complex by nature (Ou 1985, Jeung et al. 2007).

The blast pathotypes/ races in South Korea are highly virulent and different from other country pathotypes. Most of the blast resistance genes like, *Pib*, *Pita*, *Pil*, *Piz*, and *Pik* are susceptible to blast isolates of Korea. However, the identification of a new source of blast resistance (R) gene in the IRRI breeding line, IR65482-4-136-2-2 and subsequent mapping of the resistance gene (*Pi40*) on the chromosome 6, have provided opportunities to use the gene for blast resistance breeding. Marker-assisted backcross breeding (MAB) is an ideal approach to successfully introduce the new genes into diverse rice cultivars (Collard and Mackill 2007). The objective of this study is to introduce the new gene *Pi40* into elite Japonica cultivars through marker-assisted selection and backcross breeding and develop improved blast resistant breeding lines.

Methods

This study involved two Japonica cultivars, Jinbubyeo and Junambyeo which were susceptible to most of the blast isolates in Korea. The donor for the new resistance gene, *Pi40* was IR65482-4-136-2-2. BC₃F₄ progenies were produced from crosses of Jinbubyeo x IR65482-4-136-2-2, and Junambyeo x IR65482-4-136-2-2. Early generation and advanced generation BC progenies were planted in the experimental field of the Genetics and Breeding Division, National Institute of Crop Science, RDA at Suwon. BC progenies were selected for the presence of the *Pi40* gene and desirable agronomic traits by a dual selection method (foreground selection using the STS marker, 9871.T7E2b and recurrent parent traits). Selected BC progenies were selfed. Blast resistant breeding lines in japonica genetic background were evaluated in different blast hotspot areas in Korea. Four *Pi40* derived blast resistant lines, recurrent parents and monogenic differential lines with known resistance genes were evaluated at the greenhouse

by sequential planting method for seven cycles with 29 field blast isolates inoculation. SSR marker analysis was carried out for background genotyping of selected breeding lines.

Results

The DNA marker, 9871.T7E2b tightly linked to the *Pi40* gene was validated on the advanced BC progenies in the genetic background of two blast susceptible japonica cultivars Jinbubyeo and Junambyeo by gene-marker association analysis (Fig. 1). The new R gene, *Pi40* was also evaluated for its broad-spectrum resistance by using ten virulent isolates available at IRRI, Philippines. The resistant donor line, IR65482-4-136-2-2 carrying the *Pi40* gene expressed true resistance to all the Philippines isolates tested, and further confirmed broad-spectrum resistance of the R gene, *Pi40*.

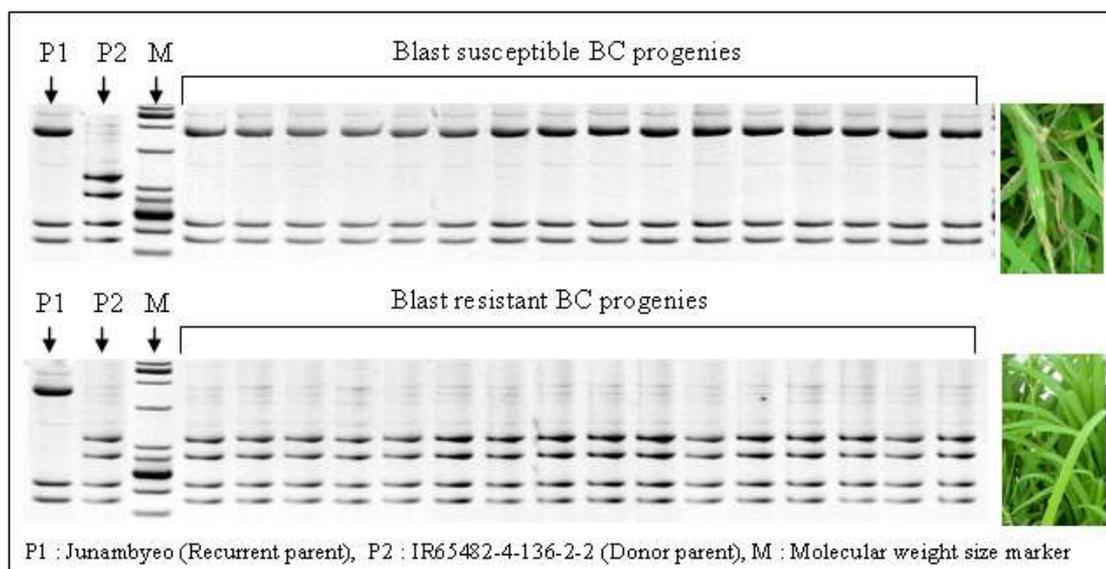


Fig. 1. MAS validity test of blast-resistant BC progenies in susceptible japonica cultivars (P1) genetic background. Gel image of detected marker genotypes on the 9871.T7E2b locus among selected BC progenies derived from a cross between Junambyeo and IR65482-4-136-2-2 (P2).

50 breeding lines were produced in the genetic background of Jinbubyeo and Junambyeo by 3 back crosses and 4-5 generations of selfing. Most of the breeding lines were selected for recurrent parent phenotype possessing the *Pi40* gene using marker-assisted selection (MAS) and evaluation of desirable agronomic traits (Fig. 1). These lines were tested for leaf and neck blast resistance at two blast hotspot sites in Korea (Cheolwon and Jecheon) during 2006 and 2007. All of BC progenies having the *Pi40* gene showed resistance to leaf and neck blast in

both locations. Our results on testing durable resistance potential of the *Pi40* gene by sequential planting revealed that the donor parent as well as four breeding lines with the *Pi40* gene expressed blast resistance consistently. However, other blast resistance genes as well as the recurrent parents were highly susceptible suggesting the potential of the *Pi40* gene for durable resistance breeding. These breeding lines possessed agronomic traits comparable to the recurrent parents (Table 1).

Table 1. Major agronomic traits of promising breeding lines carrying the *Pi40* gene

Line/variety	DTH* (days)	CL* (cm)	PL* (cm)	PN*	1,000 grain wt. (g)
Jinbubyeo (RP)*	95	85±3.3	21±0.3	13±1.4	21±0.2
IR83243-2-1-12-1-18-2-3	97	81±4.1	20±0.8	13±1.9	22±0.4
Junambyeo (RP)*	117	72±3.3	20±0.3	12±1.2	23±0.9
IR83260-1-1-1-2-1-8-1	100	65±2.1	23±0.8	11±1.8	23±0.4
IR83260-1-1-1-3-2-1-1	113	72±3.4	23±0.5	11±1.1	22±0.6
IR83260-1-1-12-1-1-3	116	80±2.6	19±0.6	12±1.8	21±1.0

*RP: Recurrent parent; DTH: Days to heading; CL: Culm length; PL: Panicle length; PN: Panicle number

Conclusion

This study suggested that DNA markers for the *Pi40* gene were reliable for marker-assisted blast resistance breeding in rice. Marker-assisted backcrosses realized rapid recovery of recurrent parent genotype with integration of *Pi40* gene. Detection of consistance resistance potential of the *Pi40* gene in the field and sequential planting in the greenhouse indicated its use for durable blast resistance breeding in temperate japonica and tropical indica rice improvement.

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