Theor Appl Genet (2006) 113:697–704
DOI 10.1007/s00122-006-0337-8

ORIGINAL PAPER

Pi35(t), a new gene conferring partial resistance to leaf blast in the rice cultivar Hokkai 188

T. T. T. Nguyen · S. Koizumi · T. N. La · K. S. Zenbayashi · T. Ashizawa · N. Yasuda · I. Imazaki · A. Miyasaka

Received: 7 March 2006 / Accepted: 3 June 2006 / Published online: 13 July 2006

Abstract The japonica rice cultivar Hokkai 188 shows a high level of partial resistance to leaf blast. For mapping genes conferring the resistance, a set of 190 F_2 progeny/F_3 families was developed from the cross between the indica rice cultivar Danghang-Shali, with a low level of partial resistance, and Hokkai 188. Partial resistance to leaf blast in the F_3 families was assessed in upland nurseries. From a primary microsatellite (SSR) linkage map and QTL analysis using a subset of 126 F_2 progeny/F_3 families randomly selected from the above set, one major QTL located on chromosome 1 was detected in the vicinity of SSR marker RM1216. This QTL was responsible for 69.4% of the phenotypic variation, and Hokkai 188 contributed the resistance allele. Segregation analysis in the F_3 families for partial resistance to leaf blast was in agreement with the existence of a major gene, and the gene was designated as *Pi35*(t). Another QTL detected on chromosome 8 was minor, explained 13.4% of the phenotypic variation, and an allele of Danghang-Shali increased the level of resistance in this QTL. Additional SSR markers of the targeted *Pi35*(t) region were further surveyed in the 190 F_2 plants, and *Pi35*(t) was placed in a 3.5-cM interval flanked by markers RM1216 and RM1003.

Keywords Rice · Leaf blast · Partial resistance · QTL analysis · SSR marker

Communicated by Q. Zhang

T. T. T. Nguyen · S. Koizumi (✉) · T. N. La · N. Yasuda · I. Imazaki · A. Miyasaka
Fungal Disease Laboratory, Department of Plant Pathology, National Agricultural Research Organization, Kannondai 3-1-1, Tsukuba, Ibaraki 305-8666, Japan
e-mail: skoizumi@affrc.go.jp

T. T. T. Nguyen · T. N. La
Molecular Biology Department, Agricultural Genetics Institute, Hanoi, Vietnam

K. S. Zenbayashi · T. Ashizawa
National Agricultural Research Center for Tohoku Region, Yotsuya, Omagari, Akita 014-0102, Japan

Introduction

Blast, caused by *Pyricularia grisea* (Cooke) Sacc., teleomorph *Magaporthe grisea* (Hebert) Barr (Rossman et al. 1992), is a serious and widespread disease of rice in both tropical and temperate regions. The blast pathosystem consists of two major interrelated phases: leaf blast and panicle blast, with the former providing inoculum for the later (Ou 1985). The use of resistant rice cultivars is an effective way to control the disease, and genetic analyses for the disease resistance have been conducted for developing resistant cultivars.

Blast resistance in rice has been generally classified into two types: complete (qualitative) or true and partial (quantitative) or field resistance (Ezuka 1972). Complete resistance, caused by incompatible combinations between the host and pathogen strains, prevents reproduction of the pathogen, and the resistance is usually controlled by a major gene. Another form of resistance, partial resistance, reduces the extent of pathogen reproduction in the compatible interaction, and the resistance is generally non-race specific and polygenic (Kiyosawa 1981).

Major genes conferring complete resistance have been introduced into many rice cultivars to control blast. However, the complete resistance of these cultivars has broken down within a few years after their release due to the increase in new, virulent blast races to the resistance (Bonman 1992; Kiyosawa 1982). On the other hand, partial resistance is commonly stable to different blast races. Therefore, the improvement in levels of partial resistance is considered to be one of the major goals in rice breeding (Kiyosawa 1981).

The genetics of partial resistance has been characterized using different rice cultivars, and QTLs for partial resistance to both leaf and panicle blast have been identified on different chromosomes of rice (Wang et al. 1994; Prashanth et al. 2000; Chen et al. 2000; Fukuoka and Okuno 2001; Miyamoto et al. 2001; Tabien et al. 2002). These studies have found that inheritance of partial resistance was most likely polygenic. However, major genes controlling partial resistance to blast such as *Pif* (Toriyama et al. 1968), *Pb1* (Fujii et al. 2000) and *Pi34*(t) (Zenbayashi et al. 2002) have been identified. Although some of the resistance genes for partial resistance have been mapped on rice chromosomes, the genetic basis of partial resistance to blast in most of rice cultivars has not been clarified yet.

The *japonica* rice cultivar Hokkai 188 was developed from a *japonica* Chinese cultivar Reishiko planted in central China, which has an extremely high level of partial resistance to blast, and has been used as a donor for blast resistance (Koyama 1952; Watanabe 1980, Fig. 1). Hokkai 188 was developed in 1961 at the Hokkaido National Agricultural Experiment Station, has a stable and high level of partial resistance to leaf blast in Japan, and has been used as a donor for developing blast-resistant cultivars (Mikami et al. 1990). The cultivar Fukei 138 is one of the blast-resistant cultivars developed from Hokkai 188, and it also shows a high level of partial resistance to leaf blast (Mikami et al. 1990, Fig. 1). Mikami et al. (1990) conducted a genetic analysis of the partial resistance in Fukei 138, and clarified that partial resistance in this cultivar is controlled by a major gene. Moreover, they presumed that the high level of partial resistance in Fukei 138 was derived from Hokkai 188. However, genetic analyses of partial resistance in Hokkai 188 have not been performed.

In this study, we conducted a genetic analysis for partial resistance to leaf blast in Hokkai 188, and identified a new resistance gene.

Materials and methods

Plant materials

A set of 190 F_2 progeny/F_3 families was developed from a cross between Danghang-Shali, an *indica* lowland rice cultivar with a low level of partial resistance to leaf blast and Hokkai 188, a *japonica* lowland rice cultivar with a high level of partial resistance to the disease at the Tohoku National Agricultural Experiment Station (present name - National Agricultural Research Center for Tohoku Region) from 1999 to 2001.

Both of the parental cultivars are lacking effective complete-resistance genes and are susceptible to most pathogenic races of rice blast fungus distributed in Japan (Naito et al. 1999), showing susceptible reactions to the Japanese blast races, 001.2, 003.0, 005.0 and 007.0.

Evaluation of partial resistance to leaf blast

Evaluation of partial resistance to leaf blast in the 190 F_3 families was conducted in an upland nursery trial in 2002 at the National Agricultural Research Center for Tohoku Region, in Omagari, Akita, Japan. About fifty seeds of each of the F_3 families and the parental cultivars were sown in a 40 cm-length row with 10 cm spacing on June 6, 2002. The respective F_3 families were planted in alternate rows, and the parental cultivars were drilled alternately between them. A complete randomized block design was used with three replications. To induce leaf blast development, the rice cultivar Sasanishiki with

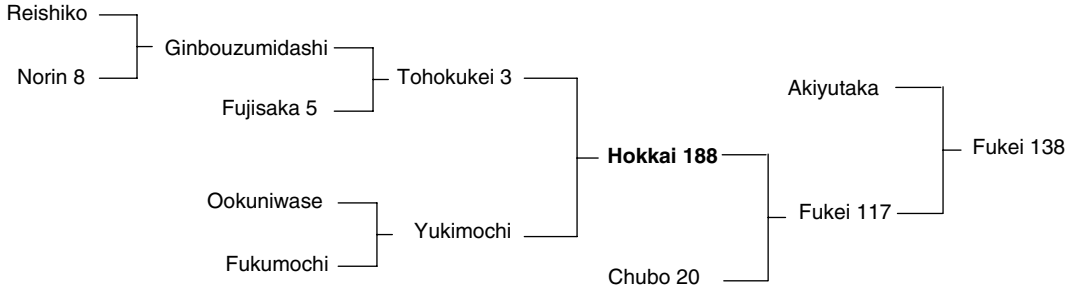

Fig. 1 The pedigree of the rice cultivar Hokkai 188

a low level of partial resistance to blast and susceptible to the blast race 007.0 was used as a spreader by planting this cultivar in ten rows at both sides of each block. The diseased leaves of the rice cultivar Sasanishiki, inoculated with the race 007.0, were scattered on the experiment plots on July 5, 2002. The disease severity of leaf blast in each of the F_3 families and the parental cultivars was evaluated on July 19 and 23 based on visual assessment of disease severity described by Asaga (1981). The scores ranged from 0 (no lesion) to 10 (all plants killed by the disease). Nitrogen was applied at 2 kg/100 m^2 in the trial field as a basal fertilizer and top-dressed at 0.5 kg/100 m^2 on June 27.

DNA markers

Leaf samples of the 190 F_2 individuals and the parental cultivars were collected from a paddy field for total DNA extraction according to the CTAB method described by Murray and Thompson (1980). A population of 126 F_2 individuals was randomly selected from the 190 F_2 individuals and used for primary map construction and QTL analysis. The 1364 SSR primers provide by Rice Genome Center (RGP, Japan) were used for polymorphism analysis of the parents. The polymorphic markers were then used for segregation analysis of the 126 F_2 individuals. DNA bands were visualized via ethidium bromide staining on 3% agarose gel. For PCR amplification, the following conditions were used: 20 µl reaction mixture containing 50 ng template DNA, 0.5 µM of each primer, 200 µM of each dNTPs, 1.5 mM $MgCl_2$, 0.5 unit Takara Ex Taq^{TM} polymerase (TaKaRa Shuzo Co., LTD) and 2 µl of 10X Ex Taq^{TM} buffer (Mg^{+2} free).

Map construction and QTL analysis

A primary genetic linkage map was constructed from segregating data of the 126 F_2 individuals using MapMaker Macintosh V2.0. The map distance was calculated based on the Kosambi function. Phenotypic mean data of leave blast severity on July 23 for partial resistance were used to identify QTLs by employing MapMaker/QTL V 1.1 (Lincoln et al. 1993). The threshold of the LOD score used for declaring the presence of the QTL was 3.0. Single and interval marker analyses were performed using QGene 3.06z.

In the first trial of this study to detect QTLs, the putative location was identified on chromosome 1, and then 40 markers previously mapped on the targeted chromosome 1 region were additionally surveyed. Polymorphic markers were used for segregation analysis of the 190 F_2 plants.

Results

Field evaluation of blast resistance in F_3 families

Partial resistance to blast can only be evaluated and detected without effective complete resistance to the disease. After the final assessment of disease severity in the upland trial field, we isolated blast strains from leaf blast lesions in the field and identified their races. All of the strains belonged to the race 007.0 that can attack both parental cultivars of the tested F_3 families and that was the race used to make inoculum in the trial.

The average scores of the leaf blast severity of Danghang-Shali (9.9 or 94.9% for diseased leaf area, DLA) and Hokkai 188 (6.1 or 43.7% for DLA) at the final assessment was statistically significant difference at $P<0.01$ (Fig. 2). The frequency distribution of the leaf blast severity in the F_3 families was almost bimodal. The range of the distribution at the final assessment was from 5.0 to 10.0, and its boundary score was 9.0 (Fig. 2). The F_3 families were divided into 2 groups: "r" (partially resistant) and "s" (susceptible) based on the boundary score 9.0 and bounds of leaf blast severity scores of the parental cultivars (Fig. 2). The segregation ratio for the numbers of "r" and "s" families fitted the expected 3:1 ratio (Table 1). These results suggest that the partial resistance to leaf blast in Hokkai 188 is mainly controlled by a dominant gene.

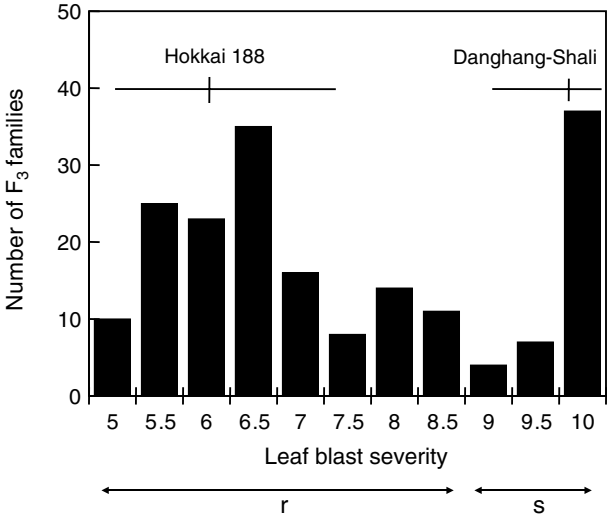

Fig. 2 Frequency distribution of leaf blast severity in F_3 families from the cross between Danghang-Shali and Hokkai 188. *Vertical* and *horizontal lines* indicate the averages and range of disease severity scores of the parental cultivars, respectively. The F_3 families were divided into "r" (partially resistant) and "s" (susceptible) based on the boundary score 9.0 and bounds of leaf blast severity scores of the parental cultivars

Table 1 Segregation of partial resistance to leaf blast in F_3 families from the cross between Danghang-Shali and Hokkai 188

Number of F_3 families			Expected ratio	X^2	P
r	s	Total			
142	48	190	3:1	0.007	0.9–0.95

r Partially resistant rice families

s Susceptible families with a low level of partial resistance

Levels of the partial resistance in the F_3 families were decided from those of the parental cultivars

Scores of leaf blast severity of r families were 9.0 and more and those of s families were less than 9.0

Parental polymorphism and genetic linkage map

One thousand three hundred sixty-four SSR markers distributed throughout the 12 rice chromosomes were used to detect polymorphism between Danghang-Shali and Hokkai 188. Out of these, 172 showed polymorphism between the two parents. The percentage of polymorphism detected using 3% agarose gels was 12.6%. One hundred forty-seven co-dominant and clear polymorphic markers were chosen for mapping and analysis in the 126 F_2 individuals. A genetic linkage map consisting of the 147 SSR markers was constructed. The map covered 1710.4 cM in length on 12 chromosomes using the Kosambi function with an average distance of 11.6 cM between adjacent markers. The map orientation was based on the IRMI SSR linkage map of rice given by McCouch et al. (2002). The linkage order of almost all markers coincided with the map reported by McCouch et al. (2002).

QTL analysis for partial resistance to leaf blast

Two putative QTLs were detected on chromosomes 1 and 8 based on interval mapping (Fig. 3). One QTL, strongly associated with partial resistance to blast, was mapped on the long-arm of chromosome 1 and located in the region of markers RM1216 and RM5501. The phenotypic variation explained by this QTL was 69.4%. The resistance allele of this QTL was contributed by Hokkai 188. The high level of the LOD score (Fig. 3) strongly suggested the presence of a major resistance gene in Hokkai 188. According to the current blast resistance gene nomenclature (Nagato and

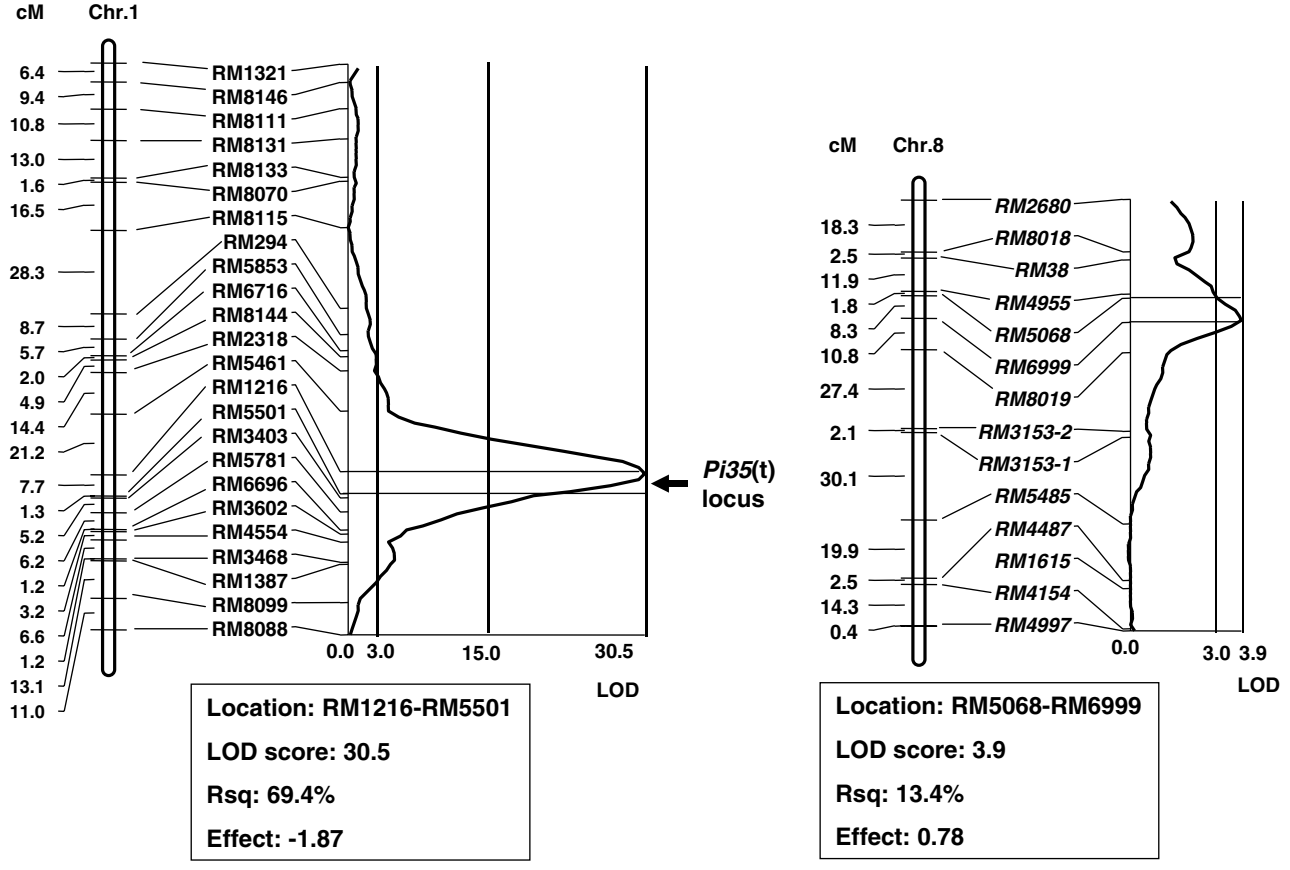

Fig. 3 QTL likelihood maps for partial resistance to leaf blast of rice chromosome 1 and 8 using 126 individuals of F_2 population derived from a cross between Danghang-Shali and Hokkai 188

Yoshimura 1998) we designate this newly identified gene as *Pi35*(t) because there are no reports for major genes conferring partial resistance to blast on chromosome 1. The second QTL was identified on chromosome 8 in the vicinity of RM5068 and RM6999 and explained 13.4% of the variation using MAPMAKER/QTL. The allele responsible for the resistance of this locus came from Danghang-Shali.

Mapping of *Pi35*(t) using 190 F_2 progeny

The purpose of this study was first to identify the chromosomal region controlling partial resistance to leaf blast in Hokkai 188. We have identified the major gene, *Pi35*(t), and mapped it on the long arm of chromosome 1 between the markers RM1216-RM5501 using a subset of the 126 F_2 individuals. To saturate the region we have surveyed 40 additional SSR markers that are located within or nearby the *Pi35*(t) region. A linkage map with 13 markers and a total length of 47.6 cM around the *Pi35*(t) region was constructed based on the genotype data obtained from the full set of 190 F_2 individuals (Fig. 4). The average distance between markers in this region was 3.6 cM. This linkage map has 10 new markers and a higher density than that presented in Fig. 3 of this study. The gene *Pi35*(t) was then identified on a 3.5 cM interval between SSR markers RM1216 and RM1003. The amount of phenotypic variation explained by *Pi35*(t) was 70.8% with the LOD score of 45.1. This map position of *Pi35*(t) corresponded well to the previously identified one from this study using a subset of the 126 F_2 individuals.

Discussion

Segregation data on leaf blast severity in the F_3 families from a cross between Danghang-Shali and Hokkai 188 in the upland nursery showed that partial resistance to leaf blast in Hokkai 188 was mainly controlled by a

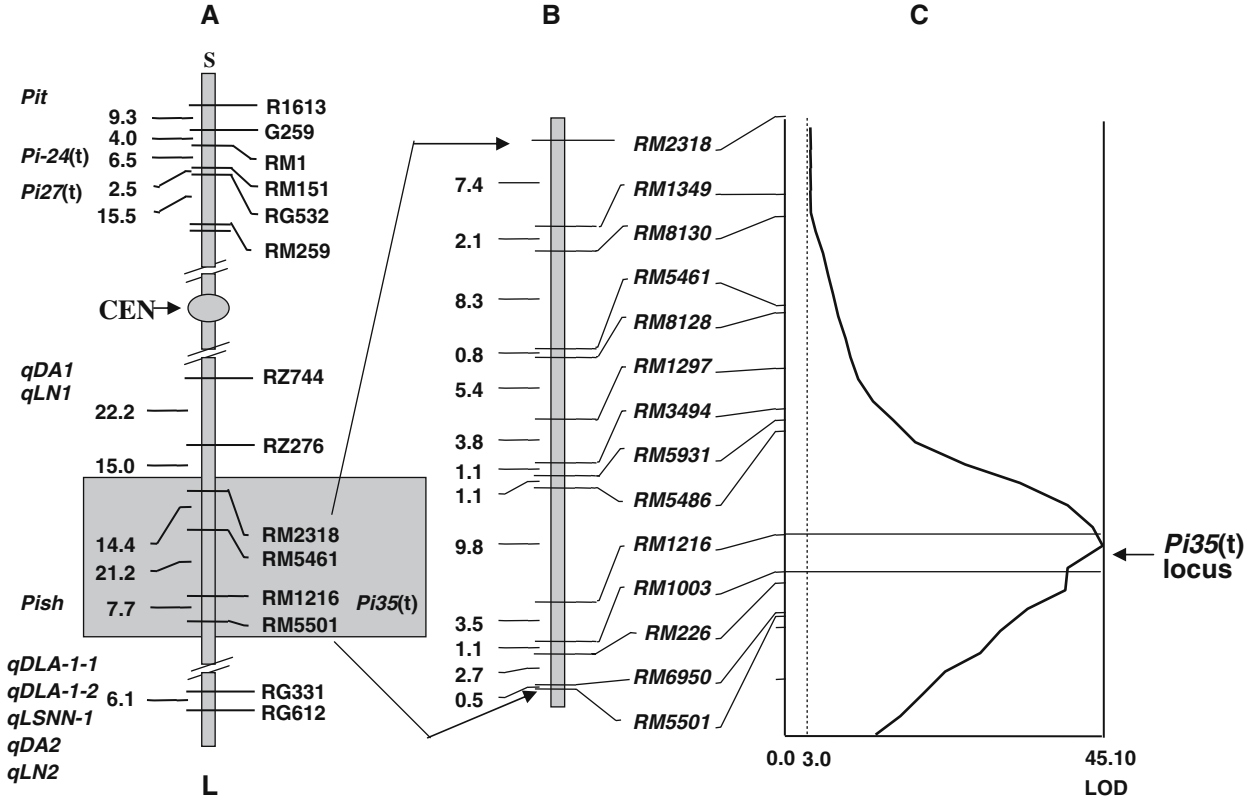

Fig. 4 Mapping of the region containing *Pi35*(t) by adding SSR markers and its comparison with other genes conferring resistance to blast on rice chromosome 1. **a** The locations of several genes and QTLs from Kaji et al.(1997); Sallaud et al. (2003); Zhu et al. (2004); Araki et al. (2003); Wang et al. (1994); Prashanth et al. (2000) and *Pi35*(t) from this study. The marker locations and map distances (*cM*) were identified and estimated from the standard genetic maps JRGP RFLP 2000 and IRMI integrate SSR 2003 (Gramene). **b** A chromosomal segment constructed from 190 F_2 individuals (64 individuals were added to the first population) using MapMaker version 2.0. **c** QTL likelihood map for *Pi35*(t)

dominant gene. This observation was confirmed by QTL analyses, and we designated the gene as *Pi35*(t). These results are consistent with the presumption of Mikami et al. (1990).

Hokkai 188 was developed from a Chinese cultivar Reishiko that has an extremely high level of partial resistance to blast (Koyama 1952; Watanabe 1980, Fig. 1). The origin of *Pi35*(t) identified in this study is thought to be from Reishiko because among rice cultivars used for the development of Hokkai 188 (except for unchecked Tohokukei 3), Reishiko was only identical with Hokkai 188 in regions near a marker designed from PAC sequences close (less than 1.9 cM; details not shown) to *Pi35*(t). The origins of genes *pi21* and *Pi34*(t) conferring partial resistance to blast are considered to be from upland rice cultivars (Fukuoka and Okuno 2001; Zenbayashi et al. 2002). However, Koyama (1952) and Watanabe (1980) did not indicate that Reishiko was an upland rice cultivar. This suggests that the origin of *Pi35*(t) is probably from a lowland rice cultivar in China.

Over the past ten years, molecular markers have facilitated the identification of chromosomal regions associated with many complex traits in rice (McCouch and Doerge 1995). The QTL approach using DNA markers has been employed to detect genomic regions controlling different complex traits including partial resistance to blast (Wang et al. 1994; Chen et al. 2000; Fukuoka and Okuno 2001; Tabien et al. 2002; Zenbayashi et al. 2002).

In this study, a total of 147 SSR markers were mapped from a survey of 1364 markers established using the 126 F_2 individuals from the cross between Danghang-Shali and Hokkai 188. Fortunately, a major QTL named *Pi35*(t) was identified on the long-arm of chromosome 1 and accounted for 69.4% of the phenotypic variation (Fig. 3). Therefore, the whole genome survey was not needed and research could be focused on the chromosome 1 region of *Pi35*(t) using an expanded F_2 population of 190 individuals (Fig. 4).

Several studies have identified major genes controlling partial resistance to blast in rice cultivars and mapped them on rice chromosome. Fujii et al. (2000) identified a major gene *Pb1* conferring partial resistance to panicle blast and mapped it on chromosome 11, and Zenbayashi et al. (2002) found a major QTL for partial blast resistance, which was located near the *Pb1* location reported by Fujii et al. (2000). Moreover, Shinoda et al. (1971) found a linkage between a major gene *Pif* for partial resistance to blast and the complete blast resistance gene *Pik* on chromosome 11. All of them have been located on chromosome 11.

Concerning chromosome 1 where *Pi35*(t) was mapped, two QTLs for partial resistance to blast in the *japonica* cultivar Moroberekan were detected in the vicinity of the markers RZ744 and RG612 on the chromosome, and a QTL with the largest effect was mapped near RZ744 on it in Moroberekan (Wang et al. 1994). Meanwhile, Prashanth et al. (2000) found three QTLs on chromosome 1 for partial resistance to leaf blast in the *indica* cultivar IR64. *Pi35*(t) identified in this study is located near the QTLs on chromosome 1 reported by Wang et al. (1994) and Prashanth et al. (2000) for partial resistance to blast. However, a comparison of the *Pi35*(t) location in this population with the reported QTLs for partial resistance on chromosome 1 did not show any coincidence. Moreover, compared to the previously identified QTLs in this chromosome region, *Pi35*(t) exhibited a major effect with a high contribution to phenotypic variation and close linkage to the SSR markers RM1216 and RM1003 (Table 2). Several complete blast resistance genes *Pi-t*, *Pi-24*(t), *Pi-27*(t) and *Pish* have also been mapped on chromosome 1 by Kaji et al. (1997), Sallaud et al. (2003), Zhu et al. (2004) and Araki et al. (2003),

Table 2 Different genes conferring blast resistance on rice chromosome 1 and their comparison with *Pi35*(*t*)

Trait	Chr#	Gene	Locus	References
Leaf blast (CR)	1	*Pi-t*	0 cM to R1613	Kaji et al. (1997)
Leaf blast (CR)	1	*Pi-24*(*t*)	Near RG532	Sallaud et al. (2003)
Leaf blast (CR)	1	*Pi-27*(*t*)	RM151-RM259	Zhu et al. (2004)
Leaf blast (CR)	1	*Pish*	RM212-OSR3	Araki et al. (2003)
Leaf blast (PR)	1	*qLN1, qDA1*	RZ744-RZ276	Wang et al. (1994)
Leaf blast (PR)	1	*qLN2, qDA2*	RG612-RG140	Wang et al. (1994)
Leaf blast (PR)	1	*Pi-35*(*t*)	RM1216-RM1003	This study
Leaf blast (PR)	1	*qDLA-1-1*	RG381-RG331	Prashanth et al. (2000)
Leaf blast (PR)	1	*qDLA-1-2*	RZ19-RG331	Prashanth et al. (2000)
Leaf blast (PR)	1	*qLSNN-1*	RZ730-RG331	Prashanth et al. (2000)

CR Complete resistance

PR Partial resistance

respectively (Table 2, Fig. 4). The locations of these genes did not coincide with *Pi35*(t) except for *Pish*, which was mapped at 7.2 cM from left boundary marker RM212 and 15.2 cM from right boundary marker OSR3 (RM226) (Fig. 4). In this study RM226 was mapped at 1.1 cM next to RM1003, which is linked to *Pi35*(t). Even though the positions of *Pish* and *Pi35*(t) identified in this study are closely linked, the blast race 007.0, which was used in this study for evaluation of partial resistance to leaf blast in the F_3 families from the cross between Danghang-Shali and Hokkai 188, can attack *Pish* as most of blast races distributed in Japan can do it (Imbe and Matsumoto 1985). This means that detection of *Pish* was impossible in this study and *Pish* is different from *Pi35*(t). Moreover, their respective resistance types and origins are different (Koyama 1952; Watanabe 1980; Imbe and Matsumoto 1985, Table 2). We have reviewed other loci of rice blast resistance genes mapped on chromosome 1. However, we could not find major genes conferring partial resistance to blast on this chromosome. Hence, we conclude that *Pi35*(t) is a new gene.

This study showed that partial resistance to leaf blast in the cultivar Hokkai 188 is controlled by a major gene. The breakdown of this resistance has not been reported for many years (Mikami et al. 1990). Therefore, *Pi35*(t) is considered to be a durable resistance gene in rice breeding and blast disease management. However, further studies on durability of the *Pi35*(t) resistance gene are necessary. A study to detect more flanking markers and fine mapping of the *Pi35*(t) locus is currently in progress. We are undertaking to select recombinant lines carrying *Pi35*(t) to facilitate physical mapping and positional cloning of the gene and to pyramid blast resistance genes including *Pi35*(t) into the elite cultivars through a marker-assisted selection strategy (Huang et al. 1997).

Acknowledgments We would like to thank Dr. Yano, M. (National Institute for Agricultural Science—NIAS, Japan) for providing the SSR primers for parental survey. We gratefully acknowledge technical assistance of Dr. Kawasaki, S. (NIAS, Japan). We also thank Dr. Fukuoka, M. (NIAS, Japan), Dr. Nguyen, B. (Texas Tech University, USA) and Dr. Takeuchi, Y. (National Institute of Crop Science, Japan) for their helpful advices in the data analyses. This work was partially supported by the grant from Japan Society for Promotion of Science.

References

Araki E, Yanoria MJT, Ebron LA, Mercado-Escueta D, Takai T, Fukuta Y (2003) Mapping of a rice blast resistance gene *Pish*. Breeding research, 5 (Suppl. 2), 333

Asaga K (1981) A procedure for evaluating field resistance to blast in rice varieties. J Cent Agric Stn 35:51–138

Bonman JM (1992) Durable resistance to rice blast disease-environmental influences. Euphytica 63:115–123

Chen DH, Nelson RJ, Wang GL, Inukai T, Mackill DJ, Ronald PC (2000) Characterization of blast resistance in the durable resistance rice cultivar Moroberekan. In: Tharreau D, Lebrun MH, Talbot NJ, Notteghem JL (eds) Advances in Rice Blast Research. Kluwer Academic Publishers, Dordrecht, The Netherlands, pp 17–27

Ezuka A (1972) Field resistance of rice varieties to rice blast disease. Rev Plant Prot Res 5:1–21

Fujii K, Hayano-Saito Y, Saito K, Sugiura N, Hayashi N, Tsuji T, Izawa T, Iwasaki M (2000) Identification of a RFLP marker tightly linked to the panicle blast resistance gene, *Pb1*, in rice. Breed Sci 50:183–188

Fukuoka S, Okuno K (2001) QTL analysis and mapping of *pi21*, a recessive gene for field resistance to rice blast in Japanese upland rice. Theor Appl Genet 103:185–190

Huang N, Angeles ER, Domingo J, Magpantay G, Singh S, Zhang G, Kumaravadivel N, Bennett J, Khush GS (1997) Pyramiding of bacterial blight resistance genes in rice marker assisted selection using RFLP and PCR. Theor Appl Genet 95:313–320

Imbe T, Matsumoto S (1985) Inheritance of resistance of rice varieties to the blast fungus strains virulent to the variety "Reiho". Jpn J Breed 35:332–339

Kaji R, Ogawa T, Nishimura M (1997) RFLP mapping of a blast resistance gene, *Pit*, in rice. Breed Sci (Suppl. 1) 47:37

Kiyosawa S (1981) Gene analysis for blast resistance. Oryza 18:196–203

Kiyosawa S (1982) Genetic and epidemiological modeling of breakdown of plant disease resistance. Annu Rev Phytopathol 20:93–117

Koyama T (1952) On the breeding of highly resistant varieties to rice blast by the hybridization between Japanese varieties and foreign variety in Japanese-type in rice. Breed Sci 2:25–30

Lincoln SE, Daly MJ, Lander ES (1993) Mapping genes controlling quantitative traits using MapMaker/QTL version 1.1: a tutorial and reference manual. A Whitehead Institute for Biomedical Research Technical Report, 2nd edn. Whitehead Institute, Cambridge, Massachusetts

McCouch SR, Doerge RW (1995) QTL mapping in rice. Trends Genet 11:482–487

McCouch SR, Teytelman L, Xu Y, Lobos KB, Clare K, Walton M, Fu B, Maghirang R, Li Z, Xing Y, Zhang Q, Kono I, Yano M, Fjellstrom R, DeClerck G, Schneider D, Cartinhour S, Ware D, Stein L (2002) Developing and mapping of 2240 new SSR markers for rice (Oryza sativa L.). DNA Res 9(6):199–207

Mikami T, Kawamura Y, Horisue N (1990) Estimation of resistant genes and field resistance to leaf blast of a rice cultivar 'Fukei 138'. Rep. Tohoku Br., Crop Sci. Soc. Japan 33:87–88

Miyamoto M, Yano M, Hirasawa H (2001) Mapping of quantitative trait loci conferring blast field resistance in the Japanese upland rice variety Kahei. Breed Sci 51:257–261

Murray MG, Thompson WK (1980) Rapid isolation of high-molecular-weight plant DNA. Nucleic Acids Res 8:4321–4325

Nagato Y, Yoshimura A (1998) Report of the committee on gene symbolization, nomenclature and linkage groups. RGN 15:13–74

Naito H, Iwano M, Fujita Y, Ashizawa T (1999) Distribution of pathogenic races of rice blast fungus in Japan in 1994. Misc Pub Natl Agric Res Cent 39:1–92

Ou SH (1985) Blast. In: Rice disease, 2nd edn. The Cambrian News Ltd., UK, pp 109–201

Prashanth GB, Shailaja Hittalmani, Srinivasachary Y, Shashidhar and Shashidhar HE (2000) Identification of DNA markers linked to partial resistance for blast disease in rice across four locations. In: Tharreau D, Lebrun MH, Talbot NJ, Notteghem JL (eds) Advances in Rice Blast Research. Kluwer Academic Publishers, Dordrecht, The Netherlands, pp 34–42

Rossman AY, Howard RJ, Valent B (1992) *Pyricularia grisea* the correct name for the rice blast fungus. Mycologia 82(4):509–512

Sallaud C, Lorieux M, Roumen E, Tharreau D, Berruyer R, Svestasrani P, Garsmeur O, Guesquiere A, Notteghem JL (2003) Identification of five new blast resistant variety IR64 using a QTL mapping strategy. Theor Appl Genet 106:794–803

Shinoda H, Toriyama K, Yunoki T, Ezuka A, Sakurai Y (1971) Studies on the varietal resistance of rice to blast 6. Linkage relationship of blast resistance genes. Bull Chugoku Agric Exp Sta A 20:1–25

Tabien RE, Li Z, Paterson AH, Marchetti JW, Stanel Pinson SRM (2002) Mapping QTLs for field resistance to the rice blast pathogen and evaluating their individual and combined utility in improved varieties. Theor Appl Genet 105:313–324

Toriyama K, Yunoki T, Shinoda H (1968) Breeding rice varieties for resistance to blast. II. Inheritance of high field resistance of Chugoku No. 31 (Suppl. 1). Jpn J Breed 18:145–146

Wang GL, Mackill DJ, Bonman M, McCouch SR, Champoux MC, Nelson RJ (1994) RFLP mapping of genes conferring complete and partial resistance to blast in a durable resistance rice cultivar. Genetics 136:1421–1434

Watanabe S (1980) Rice breeding using foreign rice cultivars. In: Yamazaki Y, Kosaka T (eds) Rice blast disease and resistance breeding. Hakuyusha, Tokyo, pp 34–46

Zenbayashi K, Ashizawa T, Tani T, Koizumi S (2002) Mapping of the QTL (quantitative trait locus) conferring partial resistance to leaf blast in rice cultivar Chubu 32. Theor Appl Genet 104:547–552

Zhu M, Wang L, Pan QH (2004) Identification and characterization of a new blast resistance gene located on chromosome 1 through linkage and differential analysis. Phytopathology 94(5):515–518