Theor Appl Genet (2012) 125:1033–1046
DOI 10.1007/s00122-012-1893-8

ORIGINAL PAPER

Fine mapping and identification of candidate rice genes associated with *qSTV11*^{SG}, a major QTL for rice stripe disease resistance

Tackmin Kwon · Jong-Hee Lee · Soo-Kwon Park · Un-Ha Hwang · Jun-Hyun Cho · Do-Yeon Kwak · Yeong-Nam Youn · Un-Sang Yeo · You-Chun Song · Jaesung Nam · Hang-Won Kang · Min-Hee Nam · Dong-Soo Park

Received: 2 January 2012 / Accepted: 11 May 2012 / Published online: 1 July 2012

Abstract Rice stripe disease, caused by rice stripe virus (RSV) is a serious constraint to rice production in subtropical regions of East Asia. We performed fine mapping of a RSV resistance QTL on chromosome 11, *qSTV11*^{SG}, using near-isogenic lines (NILs, BC_6F_4) derived from a cross between the highly resistant variety, Shingwang, and the highly susceptible variety, Ilpum, using 11 insertion and deletion (InDel) markers. *qSTV11*^{SG} was localized to a 150-kb region between InDel 11 (17.86 Mbp) and InDel 5 (18.01 Mbp). Among the two markers in this region, InDel 7 is diagnostic of RSV resistance in 55 Korean *japonica* and *indica* rice varieties. InDel 7 could also distinguish the allele type of Nagdong, Shingwang, Mudgo, and Pe-bi-hun from Zenith harboring the *Stv-b*^{i} allele. As a result, *qSTV11*^{SG} is likely to be the *Stv-b*^{i} allele. There were 21 genes in the 150-kb region harboring the *qSTV11*^{SG} locus. Three of these genes, LOC_Os11g31430, LOC_Os11g31450, and LOC_Os11g 31470, were exclusively expressed in the susceptible variety. These expression profiles were consistent with the quantitative nature along with incomplete dominance of RSV resistance. Sequencing of these genes showed that there were several amino acid substitutions between susceptible and resistant varieties. Putative functions of these candidate genes for *qSTV11*^{SG} are discussed.

Communicated by M. Wissuwa.

Electronic supplementary material The online version of this article (doi:10.1007/s00122-012-1893-8) contains supplementary material, which is available to authorized users.

T. Kwon · J.-H. Lee · S.-K. Park · U.-H. Hwang · J.-H. Cho · D.-Y. Kwak · Y.-N. Youn · U.-S. Yeo · Y.-C. Song · H.-W. Kang · M.-H. Nam · D.-S. Park (✉)
National Institute of Crop Science, Milyang 627-803, Korea
e-mail: parkds9709@korea.kr

J. Nam
Dong-A University, 37 Nakdong-Daero, Busan, Korea

Introduction

Rice stripe disease is a severe type of virus disease in temperate and subtropical regions of East Asia. Rice stripe virus (RSV) was estimated to affect over 0.5 Mha in Korea and Japan (Chung 1974; Toriyama 1995) in the 1960s. In Korea, the RSV infection area decreased to 0.1 ~ 2 % with pesticide applications and the wide use of resistant varieties In the 2000s (Lee et al. 2008). In China, RSV has resulted in severe yield losses, where approximately 0.6 Mha per year of rice were infected by RSV from 2000 to 2003, which rose to 1 Mha in 2004. Consequently, rice yield has been reduced by 30–40 % in heavily infected fields. Rice stripe virus has become one of the major rice diseases in China (Xie et al. 2005; Zhang et al. 2007; Zhao et al. 2010).

Typical symptoms of RSV include chlorosis and weakness of newly emerged leaves. Early disease symptoms include white or yellow spots on leaves, which gradually become a white or yellow stripe (Takahashi et al. 1991). Subsequently, the diseased leaves become yellow and necrotic. Finally, plant growth terminates and the plants begin to die. Rice stripe virus is transmitted by the small brown planthopper (SBPH; *Laodelphax striatellus*) (Ou 1972; Nault 1994). The RSV genome consists of four single-stranded RNA segments. Viral mRNAs transcribed from viral RNA or viral cRNA by RNA-dependent RNA polymerase are released into the cytoplasm. Then, a 59-capped short ribonucleotide leader cleaved from the host mRNA is added to the viral mRNAs by cap-snatching. The 59-capped RSV RNA is transcribed efficiently in host

cells (Falk and Tsai 1998; Shimizu et al. 1996). Although extensive functional analysis of the RSV genome has been conducted, the interaction between RSV and rice plants, which may clarify the mechanisms behind the appearance of disease symptoms, is unclear (Lu et al. 2009; Xiong et al. 2008, 2009).

Resistant cultivars are the most economic and effective way to control RSV. Two loci, *Stv-a* and *Stv-b*, have been reported to exhibit resistance to RSV in Japanese upland rice (Washio et al. 1968a). *Stv-a* is linked with the glutinous endosperm (wx) and lies on chromosome 6, whereas *Stv-b* is located on chromosome 11. An *indica* rice variety, Zenith, harbors *Stv-a* and *Stv-b* (Hayano-Saito et al. 2000a; Washino et al. 1968). Some *indica* varieties including Modan and Mudgo have a different resistance gene, *Stv-b*^{i}, which is allelic with *Stv-b* and incompletely dominant (Hayano-Saito et al. 2000a; Washio et al. 1968b, c). The resistance gene in *Kanto PL 3*, which is derived from the *indica* rice Pe-bi-hun, is closely linked to the *Stv-b* locus (Ikeda and Kaneda 1982).

Many studies have identified the quantitative trait loci (QTL) underlying RSV resistance. Hayano-Saito et al. (1998) mapped *Stv-b*^{i} from Modan to a 1.8-cM interval between XNpb220 and XNpb257 on chromosome 11. The *Stv-b*^{i} gene in Modan was finely mapped to a 286-kb region covered by two overlapping BAC clones (Hayano-Saito et al. 2000b). In the Korean *indica* variety Milyang 23, one QTL for RSV resistance has been detected in the interval between markers XNpb202 and C1172 on chromosome 11 (Maeda et al. 1999), which was reported to be allelic with *Stv-b*^{i}. Maeda et al. (2004) also mapped a QTL between R728 and G257 in this region from the Japanese upland variety Kanto 72. Ding et al. (2005) detected a QTL between markers XNpb202 and C1172 from the *indica* variety DV85. New genes that confer RSV resistance were mapped on chromosome 2, 3, and 7, respectively, to escape the risk of single gene resistance being overcome by changes in the virus population or the risk of losing the function of resistance (Ding et al. 2004; Maeda et al. 2004; Wu et al. 2009).

Beginning in the 1960s, *Stv-b*^{i} was introduced by backcrossing from the *indica* paddy variety Modan to the *japonica* paddy variety Norin no. 8 in Japan. *Stv-b*^{i} was subsequently used for breeding *japonica* rice varieties in Korea and Japan.

In the current study, we employed near-isogenic lines (NILs) generated by crossing a resistant *indica* cultivar with a susceptible *japonica* variety to identify RSV resistance gene from the resistant rice variety Shingwang by fine mapping using new sequences with an insertion/deletion (InDel) markers. A genome-wide rice polymorphism database developed by Shen et al. (2004) contains >400,000 InDel polymorphisms. These sequences with an InDel can be used to generate PCR markers that are able to distinguish between the *indica* variety 93-11 and the *japonica* variety Nipponbare (Shen et al. 2004). They are co-dominant markers that yield two possible alleles depending on the presence or absence of the insertion sequence. Expression levels of the genes detected in the interval between selected markers were analyzed by semi-quantitative RT-PCR. Furthermore, InDel analysis demonstrated that the RSV marker co-segregated with resistance. This molecular marker may be useful for rice breeding to improve RSV resistance via marker-assisted selection.

Materials and methods

Plant materials

We developed NILs (BC_6F_4) generated from five backcrosses of Shingwang, a resistant *indica* cultivar, with the genetic background of the susceptible *japonica* variety, Ilpum.

Evaluation of RSV resistance

Forty seeds were grown in plastic pots (25 × 45 cm in length), and 2-week-old seedlings were subsequently placed inside clear plastic cages with the SBPH with three biological replications. We placed a resistant and susceptible variety as control in each cage. Second or third instar SBPH nymphs were maintained in these cages at a density of approximately seven insects per seedling over a period of 3 days during the feeding experiments. The proportion of viruliferous SBPH was approximately 38 % using an ELISA assay. SBPHs were scattered daily to prevent them from remaining in one area. After 3 days, the insects were removed and plants were transplanted in a plastic house at the National Institute of Crop Science (Milyang, Korea). The reaction to RSV was evaluated using the method of Wu et al. (2011) by calculating the proportion of healthy plants (the percentage of healthy plants in a given plot) at 1 month after transplanting. Plants with yellow and white stripe disease lesions and more severe symptoms were considered susceptible plants. In contrast, plants without typical disease lesions or those that exhibited slight symptoms or disappeared,were considered resistant plants.

Genotype analysis of the BC_5F_5 resistant line (parental line of 460 BC_6F_3 NILs)

SSR markers evenly distributed in the entire rice chromosome were selected from the Gramene database (http://www.gramene.org), and the polymorphic markers were

used for frame mapping of a BC_5F_5 resistant line. To identify the introgression region from the donor parent, Shingwang, YR24982-9-1 (BC_5F_5 resistant line) was genotyped using 205 polymorphic SSR markers covering the 12 rice chromosomes (Supplementary Table A). For PCR, the temperature cycling conditions were 4 min at 94 °C, followed by 40 cycles of 94 °C for 30 s; 55 or 60 °C for 30 s; and 72 °C for 1 min, and a final extension at 72 °C for 7 min. The amplification products were separated using either a 4 % denatured polyacrylamide gel or a 3 % agarose gel and visualized by silver staining or EtBr, respectively.

Selection SSR markers and development of InDel markers for fine mapping

The *qSTV11*^{SG} region was previously detected Kwak et al. (2011) using $BC_5F_{2:3}$ NILs of Shingwang and *Ilpum*. SSR markers distributed in the *qSTV11*^{SG} region were selected from the Gramene database (http://www.gramene.org) for fine mapping. Additional InDel markers were designed by analyzing the sequence differences between Nipponbare (Gramene database; http://www.gramene.org) and 93-11 (BGI-RIS; http://rice.genomics.org.cn) based on the introgression region of YR24982-9-1 from the donor parent, Shingwang, in chromosome 11 to produce PCR products of 250–350 bp with fragment size differences in the range 15–25 bp that are suitable for detection with agarose gels. Amplification reactions were performed using 20 μl PCR reaction mixture that contained 50 μM of dNTPs, 10 pmol of each primer, 1× PCR buffer, 0.5 U of *Taq* DNA polymerase (Solgent, Daejeon, Korea), and 20 ng of DNA template. Thermal cycling was carried out in a PTC-200 thermal cycler (MJ Research Inc., Waltham, MA, USA) programmed for 2 min at 95 °C, followed by 35 cycles of 95 °C for 20 s, 55 °C or 60°C for 40 s and 72 °C for 30 s, and 5 min at 72 °C for a final extension. The amplified DNA products were loaded onto a 4 % denatured polyacrylamide gel in 0.5× TBE buffer, then visualized by silver staining (Xu et al. 2002) for SSR marker or a 3 % agarose gel, and visualized with EtBr.

Genotype and phenotype analyses of resistant and susceptible rice varieties using selected InDel markers

For the genotype and phenotype analysis of the 55 Korean resistant and susceptible rice varieties, we compared the reaction to RSV and allele type of InDel markers designed in this study with a dominant marker, ST10, developed by Hayano-Saito et al. (2000b). The RSV Reaction of the 55 Korean rice varieties was analyzed previously by Kwak et al. (2011).

Semi-quantitative RT-PCR

Total RNA was isolated from the culms of fully emerged third or fourth leaves of Ilpum, NIL, and Shingwang 0, 3, 6, 10, and 15 days after SBPH feeding. First strand cDNA synthesis and PCR amplification from total RNA using the SuperScript III One-Step RT-PCR system (Invitrogen, Carlsbad, CA, USA) were performed according to the manufacturer's instructions. Gene-specific primers were designed to produce DNA fragments of each gene in the *qSTV11*^{SG} locus as well as the *actin* gene (Table 1).

Results

Development of the RSV resistant NILs

RSV-resistant NILs were developed by conducting four backcrosses between Shingwang, a resistant *indica* cultivar as the donor of the *qSTV11*^{SG} locus, and Ilpum, *a* susceptible *japonica* cultivar as the recurrent parent. We selected a resistant line by bioassay in each generation, which was then backcrossed with the recurrent parent. We selected YR24982-9-1 as a representative genotype in the BC_5F_5 NIL population as it maintained a high level of resistance to RSV in all bioassay experiments. The growth and development of YR24982-9-1 was indistinguishable from that of Ilpum for most characteristics. Of the 205 polymorphic markers covering the 12 rice chromosomes used to survey introgression in the resistant BC_5F_5 line, 11 markers showed the donor parent Shingwang allele on chromosome 7 (RM418, RM21652, RM21636), chromosome 11 (RM26748, RM26795, RM229, and RM206), and chromosome 12 (RM20, RM453, RM247, and RM3813), suggesting that 94.6 % of the YR24982-9-1 genome was recovered from the recurrent parent (Fig. 1). The introgression region detected on chromosome 11 appeared to co-localize with the *Stv-b*^{i} loci proposed by Hayano-Saito et al. (2000b), Maeda et al. (1999, 2004), and Ding et al. (2005).

RSV resistance of NIL, YR24982-9-1

The reactions to RSV of YR24982-9-1 and Ilpum are shown in Table 2 and Fig. 2. Reaction tests in the greenhouse indicated that the proportion of healthy plants of the resistant line, YR24982-9-1, was 86 % with no growth termination; 33 % of the tested plants showed no symptoms, and 53 % showed white or yellow stripes without a growth disorder. The proportion of healthy plants of the susceptible variety, Ilpum, was 26, and 74 % of the tested plants showed yellow and necrotic leaves with a growth disorder or growth termination.

Table 1 Primers used in the semi-quantitative RT-PCR analyses

Primer	Forward primer	Reverse primer
Os11g31410	CGTCTCTGCGATCGTTGTTT	TTGCAACTGCAATCTGCTCC
Os11g31420	TGGTGTGGGAAGCTGGATTA	TGGTGATTCCTTAATCCAGCC
Os11g31430	TTAACCCTCGAGCCGAAGTC	ACCAATTTCATCCACTTTGTGC
Os11g31440	AGGGAGGGGAAACGGAGTAG	GCTTCAAGTGCAGACCCAAC
Os11g31450	GGAACCTGGATTGTGGGAAT	CGCAAAATGGAGATTGCCTA
Os11g31460	TGAAGGAACCTAGCGCAATG	GGGCTAGCGCTCCAAGTAGT
Os11g31470	TTCCGTGTGGATGATGATTCTA	CTGGCTCTTGAATGATGTCATAG
Os11g31480	CCCATCACGGTTGCTTACAG	TTTTACGAGCTCGGCAAGTG
Os11g31490	CACATCGACCTACCGGTCAC	AGCCTCTCGTCGCTATCCTC
Os11g31500	CTTGCGAACCCTGTTCTTGA	CTTCAGCAAGAACTCGACCG
Os11g31510	CTTCATCGTCGAACCAAGACA	GAAGGTTGATGTATCCTTCTCCA
Os11g31520	GCTTTGGGAAAGGGATGAAC	TTCTGCCAAATGTGCTCCAT
Os11g31530	CCTGACTGGCACCATACCAC	TCAAATTCCCGACAAGGACA
Os11g31540	CTTGATCTCCAGGGCAACCT	ACCCAGTTGATCCGACAGTG
Os11g31550	CTCAGGGCCTCTGATTCCTC	TTGACATTGCCCAGAGAAGC
Os11g31560	TCGAACAACCTGCTCACTGG	TTTTCGGCATGTCATGGACT
Os11g31570	AGTGCAGTGTGCTGCCTCTC	ATCCAGATGCTCCTTGAGGC
Os11g31590	CGGCTACAGGAAGCTGTTCA	GAACACGCCGAGCATCTTG
Os11g31600	ACCAAATGAGGTTCTTGCCC	TGCAGCTCCTTTGGTGTCTT
Os11g31610	GAGGAAGAAGAGGTGGCCG	GAATCCAGCGGATCTTCTCC
Os11g31620	TTTTGGGGCAATCGTACAGT	AATTCAGGTGCATCACCCCT
Actin	GGAACTGGTATGGTCAAGGC	AGTCTCATGGATACCCGCAG

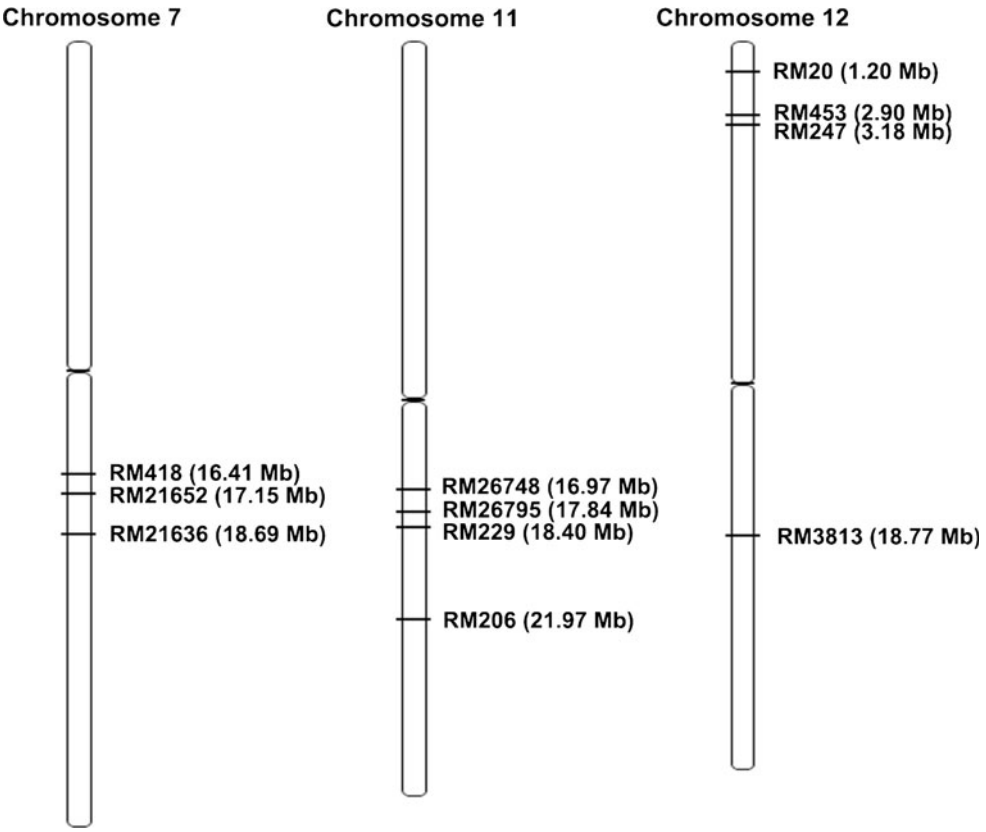


Fig. 1 Frame map of the resistant NIL, YR24982-9-1, showing the introgression region of the donor parent Shingwang in the Ilpum background in the complete chromosome using 205 SSR markers. Introgressed SSR markers and their physical location in each chromosome are indicated

Table 2 Reaction to RSV on resistant YR24982-9-1 and susceptible Ilpum after an artificial RSV inoculation

Variety	No./(Proportion) of healthy plants (%)		No./(Proportion) non-healthy plants (%)	
	No symptom	White or yellow stripe	Yellow and necrotic	Growth termination
YR24982-9-1	17 (33 %)	27 (53 %)	7 (14 %)	0 (0 %)
Ilpum	9 (10 %)	5 (16 %)	16 (52 %)	7 (22 %)

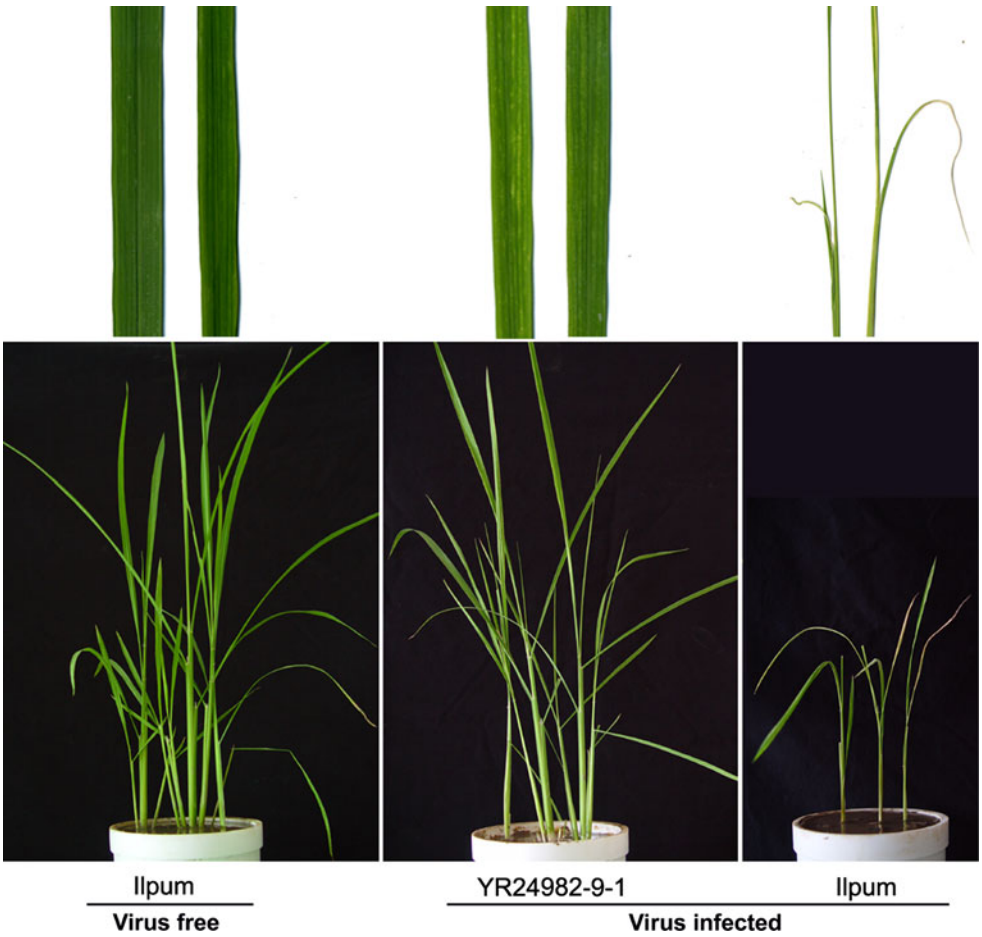


Fig. 2 Typical symptoms of stripe disease on the resistant NIL, YR24982-9-1, and susceptible variety, Ilpum

Genotype and phenotype analysis of 460 BC_6 NILs

We analyzed the genotypes of 460 BC_6F_2 NILs with polymorphic markers based on the introgression region of YR24982-9-1 from a donor parent, Shingwang, in chromosome 11 to select a segment introgression line for fine mapping of *qSTV11*^{SG}. The polymorphic markers, InDel 1 to InDel 6, and RM6897, were designed based on the introgression region of YR24982-9-1 from a donor parent, Shingwang, in chromosome 11 (Table 3).

The genotypes of 460 BC_6F_2 NILs with polymorphic markers are illustrated in Fig. 3. Chromosome segment introgression lines that were heterozygous in a different region were selected based on the genotypes of each line to induce further recombination in the given chromosomal region.

The genotypes of plants from 20 selected $BC_6F_{2:3}$ NILs were analyzed using 14 polymorphic markers as shown in Table 3. Finally, we established 10 homozygous chromosome segment introgression BC_6F_4 lines targeted to the region between InDel 1 and InDel 2. The reaction to RSV of the selected BC_6F_4 lines was denoted as the proportion of healthy plants (Table 4).

Bioassay was conducted with three biological replications. The reaction to RSV was determined by a statistical analysis of the proportion of healthy plants. The average

Table 3 Polymorphic markers used for fine mapping of *qSTV11*^{SG}

Primer ID	Forward primer (5′–3′)	Reverse primer (5′–3′)
RM6897	ATATCCGATGTGACACGCAG	AGGATAAATTGGGTGGGGAC
RM6680	GGAGGACGTTGATATGTCTCG	TGCGACACAGTTACAAAGCC
RM1355	GGAGGACGTTGATATGTCTCG	TGCGACACAGTTACAAAGCC
InDel 1	CAGCAGCCCTATGGAAACAA	TGGAGGGGAATAGGATTTGG
InDel 2	ACCGACGGTGATGTAGGGAT	ACTCCACATGGAACACCACG
InDel 3	CAATTTAGATCGATGGCCGA	ATGTGGGCAGATGGCACTTA
InDel 4	CTCAAGCTCGACAAGGTCTG	CCTCCGTCGTCTGCCTTTTT
InDel 5	AAAGGAATTAAATTGTACCACAAACA	CATGGAGTTGAGTTGGTTAATGC
InDel 6	CAGGATGAGGCTACAATTTTCAA	GTCTGGATCCACTTCATACCA
InDel 7	AAATTCCAATGCCCAAAACC	TCCTCATGGAGCTCATCCAA
InDel 8	GGCAAATTTGGAAAGGTGGT	TCAACAGCGCCATACATTAAAA
InDel 9	CGCCGATGGCAATAAATAAA	TGGACCATATCTCTGCGTGG
InDel 10	CTGTTCCGTTGCTGCTCTGT	GGTGAATCTATCGCCTGCAA
InDel 11	AAAATATCGGGGCCATCAAT	TGATCAAGCTGGACCAAGTTCT

proportion of healthy plants on YR24982-9-1 and Ilpum was 82.1 and 11.8 %, respectively, which was similar to the previous result (Table 2). According to Duncan's new multiple range test, the proportion of healthy plants on the resistant NIL, YR24982-9-1, belonged to Group ab. Varieties/lines classified to Group a and Group ab were regarded as resistant, and Group c was considered susceptible.

The physical map with the additional InDel markers, InDel 7 to InDel 11, and the genotypes of the ten tested recombinants are illustrated in Fig. 4. The 11 InDel markers and three SSR markers were used to characterize the introgression region of ten chromosome segment introgression BC_6F_4 lines, including five resistant lines (1-158-5, 2-128-8, 2-194-2, 2-42-2, and 2-43-3) and five susceptible lines (2-171-7, 1-196-3, 1-196-9, 1-186-12, and 2-43-3). Considering the graphical genotypes and the resistance phenotypes of the recombinants, it is clear that the locus conferring resistance to RSV was an approximate 150 kb region on the physical map between InDel 11 (17.86 Mbp) and InDel 5 (18.01 Mbp) and was tightly linked to InDel 7 and InDel 8.

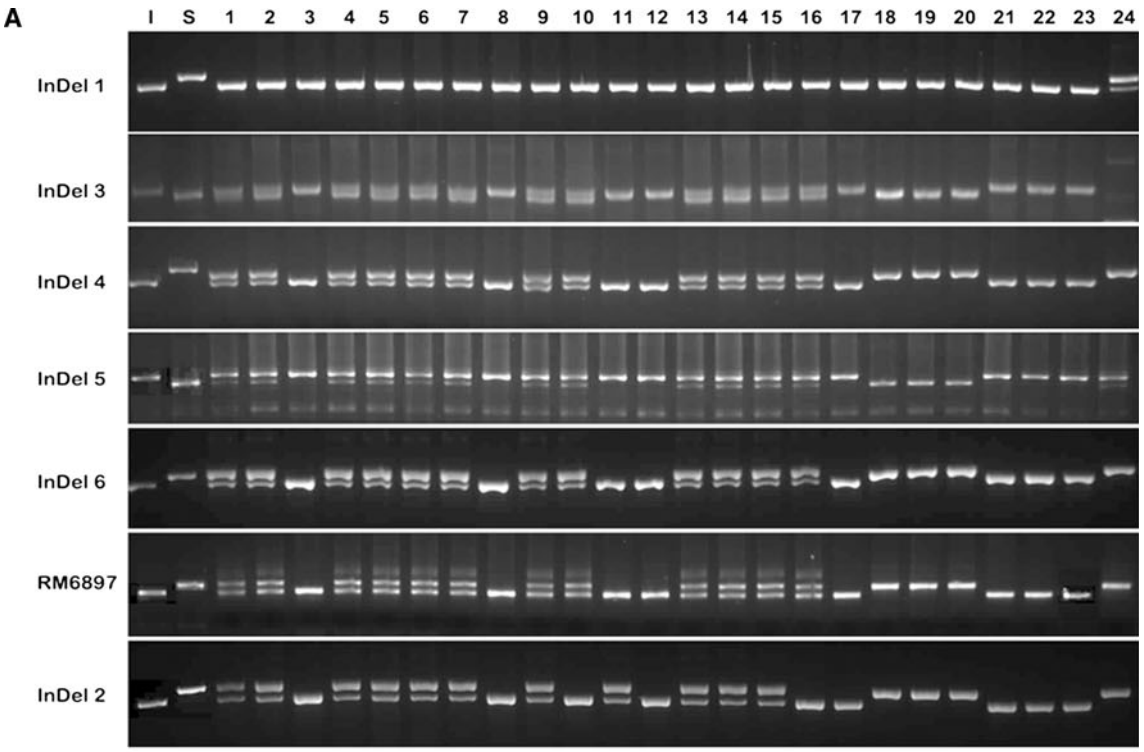
Genotype and phenotype analysis of resistant and susceptible rice varieties using new InDel markers

To select an InDel marker for marker-assisted selection of stripe disease resistance in a rice breeding program, we compared the reaction with RSV and genotype of InDel 7 and InDel 8, of the 55 Korean rice varieties (Fig. 5). The presence of InDel 7 was perfectly correlated with RSV resistance in all *japonica* and *indica* rice tested. InDel 8 was also closely linked to *qSTV11*^{SG} in NILs, but it did not amplify in the resistant *japonica* rice varieties, and did not show a consistent allele type in the susceptible *japonica* or resistant *indica* rice varieties. All *japonica* varieties used in this study could also be discriminated by the ST10 marker, whereas this marker could not identify some of the resistant *indica* varieties, including Shingwang. These results suggest that a newly developed marker set of InDel 7 can be used in PCR-assisted selection of stripe disease resistance of *Stv-b*^{i} in both *indica* and *japonica* rice breeding programs.

Gene prediction in the critical region of chromosome 11 and their expression level during RSV infection

The physical distance of the 150-kb interval between the markers, InDel 11 and InDel 5 encompassed three contigs, AC136150, AC136491, and AC133710 by aligning marker sequences with the complete genome sequence of the *japonica* cultivar, Nipponbare, using Gramene (http://www.gramene.org/Multi/blastview). Sequence analysis of the 150-kb region harboring the *qSTV11*^{SG} locus on the Gramene database indicated that this region contained 21 candidate genes (Table 5). This region included five transposon proteins (LOC_Os11g31410, LOC_Os11g31460, LOC_Os11g31490, LOC_Os11g31510, and LOC_Os11g31520), two ATP-binding proteins (LOC_Os11g31480, and LOC_Os11g31500), and four putative brassinosteroid insensitive 1 associated receptor kinase 1 precursors (LOC_Os11g31530,

Fig. 3 Genotype analysis of 460 BC_6F_2 NILs with polymorphic markers designed based on the introgression region of YR24982-9-1 from a donor parent, Shingwang, in chromosome 11. **a** Typical band pattern of BC_6F_2 NILs (I; Ilpum, S; Shingwang. **b** Genotype of selected NILs for fine mapping followed by generation advance (NIL; YR24982-9-1). *Black* and *gray bars* indicate chromosome regions derived from the donor parent (Shingwang) and the background parent (Ilpum), respectively. NIL Indicates YR24982-9-1. *Underlined italics* indicate the position of ST10 marker designed by Hayano-Saito et al. (2000b) ▶


A
I S 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24
InDel 1
InDel 3
InDel 4
InDel 5
InDel 6
RM6897
InDel 2

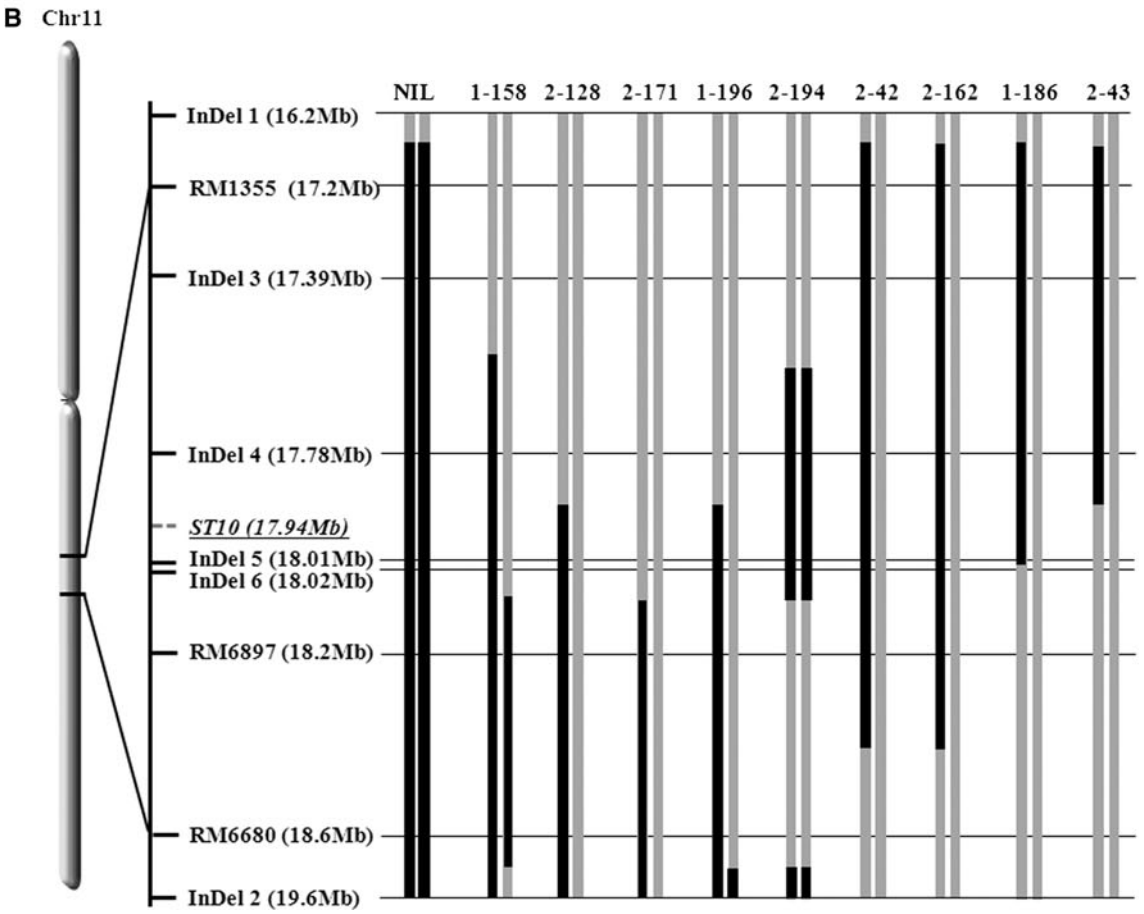

B Chr11
NIL 1-158 2-128 2-171 1-196 2-194 2-42 2-162 1-186 2-43
InDel 1 (16.2Mb)
RM1355 (17.2Mb)
InDel 3 (17.39Mb)
InDel 4 (17.78Mb)
ST10 (17.94Mb)
InDel 5 (18.01Mb)
InDel 6 (18.02Mb)
RM6897 (18.2Mb)
RM6680 (18.6Mb)
InDel 2 (19.6Mb)

Table 4 Proportion of healthy plants in the selected BC_6F_4 lines

Variety/lines	Proportion of healthy plant (%)[A]				
	Rep1	Rep2	Rep3	Average[B]	Reaction to RSV
Shingwang	96.8	100	89.6	95.4[a]	R
YR24982-9-1	84.2	80.0	82.0	82.1[ab]	R
Ilpum	8.3	10.6	16.6	11.8[c]	S
1-158-5	72.7	85.7	NT	79.2[ab]	R
2-128-8	90.9	90.9	100	93.9[a]	R
2-171-7	0	0	12.5	4.1[c]	S
1-196-3	0	0	25.0	8.3[c]	S
1-196-9	0	20.0	18.1	12.7[c]	S
2-194-11	88.8	81.8	100	90.2[ab]	R
2-42-2	80.0	100	77.7	85.9[ab]	R
2-162-3	88.8	90.0	90.0	89.6[ab]	R
1-186-12	0	16.6	25.0	13.8[c]	S
2-43-3	0	12.5	0	4.1[c]	S

[A] Bioassay on RSV was conducted in three biological replications. Each line was tested with 10 ~ 12 plants per replication

[B] Values with different letters are significantly different at $P = 0.05$

NT not tested

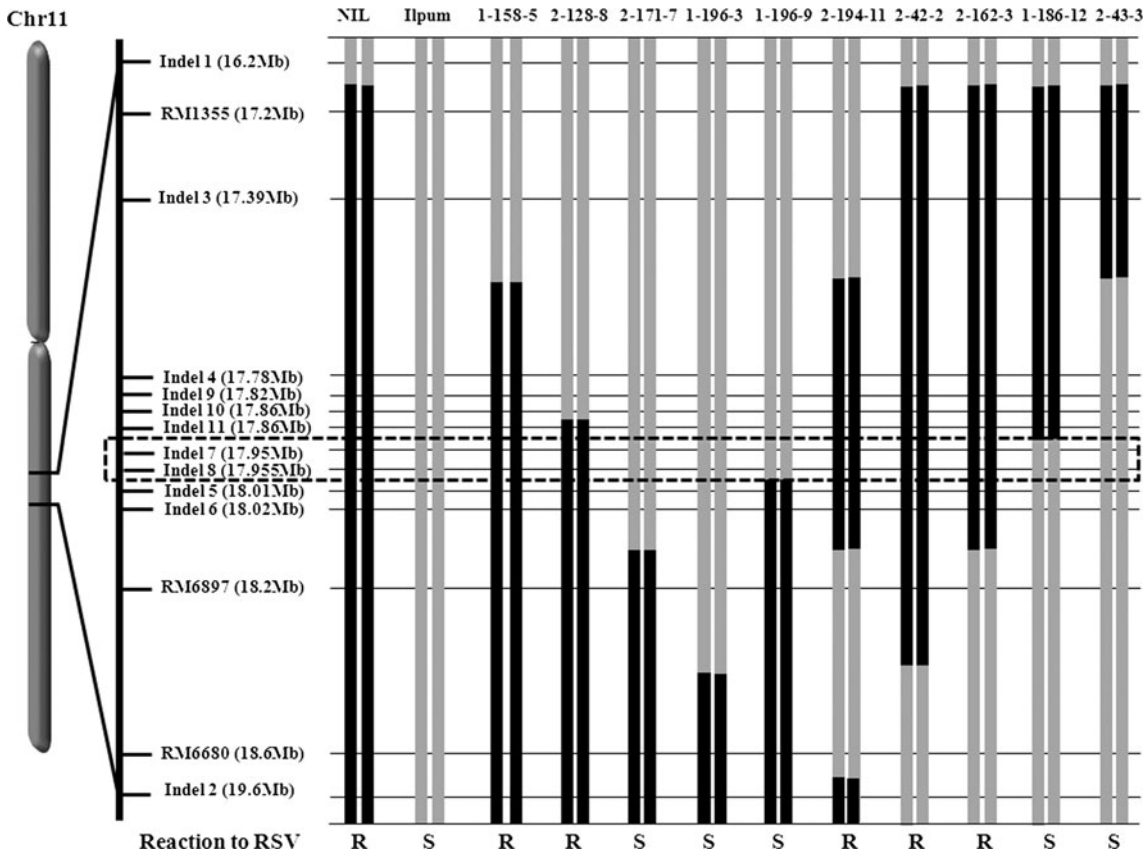


Fig. 4 The identification of $qSTV11^{SG}$ using chromosome segment introgression lines. The *dotted box* represents the positions of $qSTV11^{SG}$. *Black* and *gray bars* indicate chromosome regions derived from the donor parent (Shingwang) and the background parent (Ilpum), respectively. *NIL* indicates YR24982-9-1

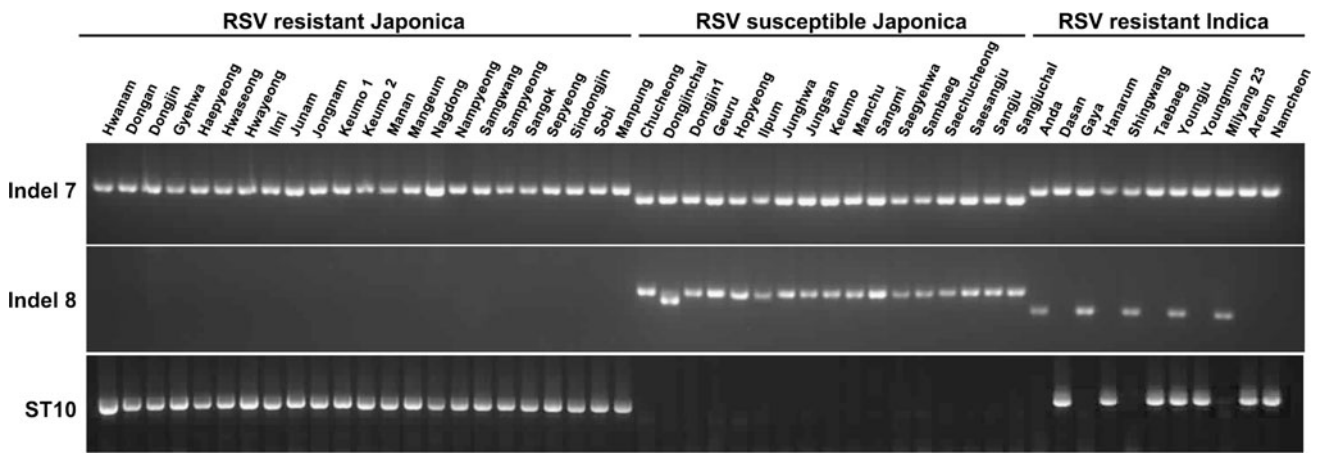


Fig. 5 Genotype and phenotype analysis of resistant and susceptible rice varieties using new InDel markers. Reaction to RSV on each variety was clearly identified during breeding procedures for each variety

Table 5 Genes in the $qSTV11^{SG}$ region and their putative function

ID	Locus identifier	Physical location (bp)[a]	Putative function
1	LOC_Os11g31410	17,863,981–17,869,844	Retrotransposon protein, putative, Ty1-copia subclass
2	LOC_Os11g31420	17,871,580–17,876,696	Expressed protein
3	LOC_Os11g31430	17,884,332–17,898,358	Expressed protein
4	LOC_Os11g31440	17,904,271–17,906,116	Hypothetical protein
5	LOC_Os11g31450	17,906,620–17,912,711	Expressed protein (kinase domain)
6	LOC_Os11g31460	17,914,035–17,915,224	Retrotransposon protein, putative, Ty3-gypsy subclass
7	LOC_Os11g31470	17,915,879–17,924,311	Expressed protein
8	LOC_Os11g31480	17,925,339–17,933,364	ATP-binding protein, putative, expressed
9	LOC_Os11g31490	17,934,262–17,936,688	Retrotransposon protein, putative
10	LOC_Os11g31500	17,943,595–17,950,401	ATP-binding protein, putative
11	LOC_Os11g31510	17,950,905–17,952,134	Transposon protein, putative, Pong sub-class
12	LOC_Os11g31520	17,952,883–17,954,366	Transposon protein, putative, Pong sub-class
13	LOC_Os11g31530	17,960,078–17,962,052	BRASSINOSTEROID INSENSITIVE 1-associated receptor kinase 1 precursor, putative
14	LOC_Os11g31540	17,964,319–17,966,417	BRASSINOSTEROID INSENSITIVE 1-associated receptor kinase 1 precursor, putative
15	LOC_Os11g31550	17,969,410–17,972,055	BRASSINOSTEROID INSENSITIVE 1-associated receptor kinase 1 precursor, putative
16	LOC_Os11g31560	17,977,420–17,979,509	BRASSINOSTEROID INSENSITIVE 1-associated receptor kinase 1 precursor, putative
17	LOC_Os11g31570	17,982,152–17,985,770	Expressed protein
18	LOC_Os11g31590	17,998,079–17,999,506	Armadillo/beta-catenin-like repeat
19	LOC_Os11g31600	18,003,663–18,004,326	Hypothetical protein
20	LOC_Os11g31610	18,007,790–18,008,149	Hypothetical protein
21	LOC_Os11g31620	18,019,166–18,024,771	OsFBL55 - F-box domain and LRR containing protein, expressed

LOC_Os11g31540, LOC_Os11g31550, LOC_Os11g31560). We analyzed the expression level of the 21 candidate genes of the RSV resistant varieties, Shingwang, and YR24982-9-1, and the susceptible variety, Ilpum (Fig. 6). Our RT-PCR data showed that 12 of 21 genes in the 150-kb region harboring the $qSTV11^{SG}$ locus were not expressed in either resistant or susceptible varieties. Four genes including LOC_Os11g31480, LOC_Os11g31540, LOC_Os11g31540, and LOC_Os11g31620 showed a similar expression pattern between resistant and susceptible varieties. Two putative transposon proteins, LOC_Os11g31510 and LOC_Os11g31520, were expressed in Shingwang, whereas they were not expressed in the resistant NIL and YR24982-9-1 or the susceptible variety, Ilpum. LOC_Os11g31430, LOC_Os11g31450, and LOC_Os11g31470 were classified as expressed proteins (Table 5) and exclusively expressed in the susceptible variety, Ilpum. We

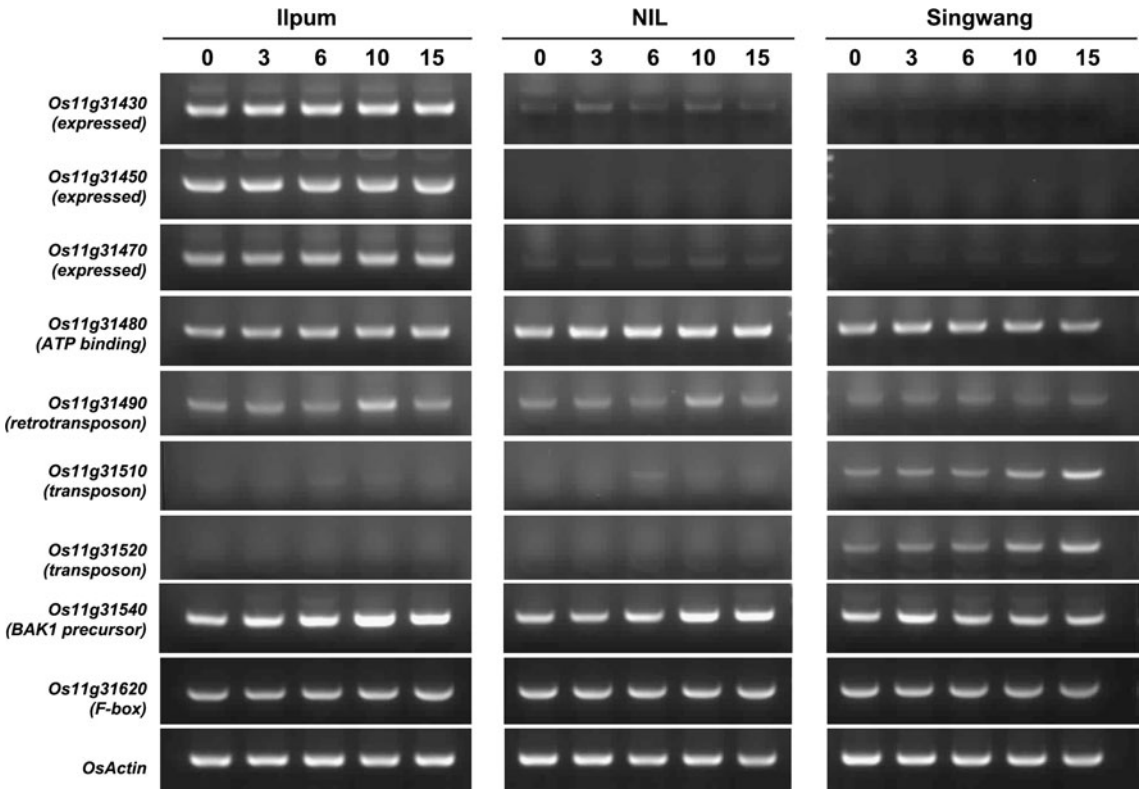


Fig. 6 Semi-quantitative RT-PCR analysis of genes in the $qSTV11^{SG}$ region on the resistant (Shingwang and NIL; YR24982-9-1) and susceptible rice varieties in response to RSV. Total RNA was extracted from rice seedlings after different feeding time periods of RSV infected SBPH. *Lane 1* control, *Lane 2* 3 days, *Lane 3* 6 days, *Lane 4* 10 days, *Lane 5* 15 days after feeding. The number of reaction cycles for each cDNA was 32 cycles except for the following 3 cDNAs: *Os11g31480*; 35 cycles, *Os11g31480*; 35 cycles, and *actin*; 28 cycles. Primers for the rice *actin1* gene were used as an internal reference control

isolated the coding region of the three genes from Ilpum, Shingwang, and YR24982-1 by PCR, respectively. Sequence analysis of the isolated genes revealed that there were several base substitutions between susceptible (Nipponbare and Ilpum) and resistant (Shingwang and YR24982-1) varieties (Fig. 7). The LOC_Os11g31430 sequence in the resistant varieties had a single-nucleotide changes that converted Cys^{149} to Arg. Furthermore, LOC_Os11g31450 and LOC_Os11g31470 in the resistant varieties deviated from susceptible varieties by converting four (Ile^{25} to Lys, Met^{156} to Thr, Leu^{258} to Phe, and Met^{377} to Thr) and two (Arg^{561} to Ser and $Stop^{860}$ to Arg) amino acids, respectively.

Discussion

In this study, we performed fine mapping of the $qSTV11^{SG}$ locus related to RSV resistance in Shingwang using $BC_6F_{2:3}$ NILs with newly developed InDel markers. Our results revealed that the $qSTV11^{SG}$ locus was located at a 150-kb region between the markers, InDel 11 and InDel 5. This region coincided with previous studies on resistance QTL in rice. Hayano-Saito et al. (2000b) mapped the *Stv-b*^{i} gene in Modan to a 286-kb region between the two markers, XNpb220 and XNpb257. Maeda et al. (1999) also reported a QTL in the interval between markers XNpb202 and C1172 from *Milyang23*. Recently, Wu et al. (2011) suggested that $qSTV11^{TQ}$ is *Stv-b*^{i} or its multi-alleles in the 55.7-kb region between the two markers, CAPs3 and CAPs2.

Varieties with a single resistance gene are at an increased risk of their resistance being overcome by new virus strains. Thus, scientists have attempted to discover various genetic resources for resistance to RSV in rice-breeding programs. Three genes have been commonly utilized in rice breeding, *Stv-a*, *Stv-b*, and *Stv-b*^{i}. *Stv-b* and *Stv-b*^{i} are allelic and located on chromosome 11. Japanese upland varieties including Kuroka, and an *indica* variety, Zenith, are known to harbor two pairs of complementary dominant genes, *Stv-a* and *Stv-b* (Washio et al. 1968a). St. no. 1 is the first resistant *japonica* variety developed in Japan harboring *Stv-b*^{i} from the *indica* variety, Modan. The first RSV-resistant *japonica* rice variety in Korea was Nagdong (Chung et al. 1975), and the *Stv-b*^{i} gene of Nagdong was introduced from a Japanese variety, Mineyutaka, derived from St. no. 1. The *Stv-b*^{i} gene has

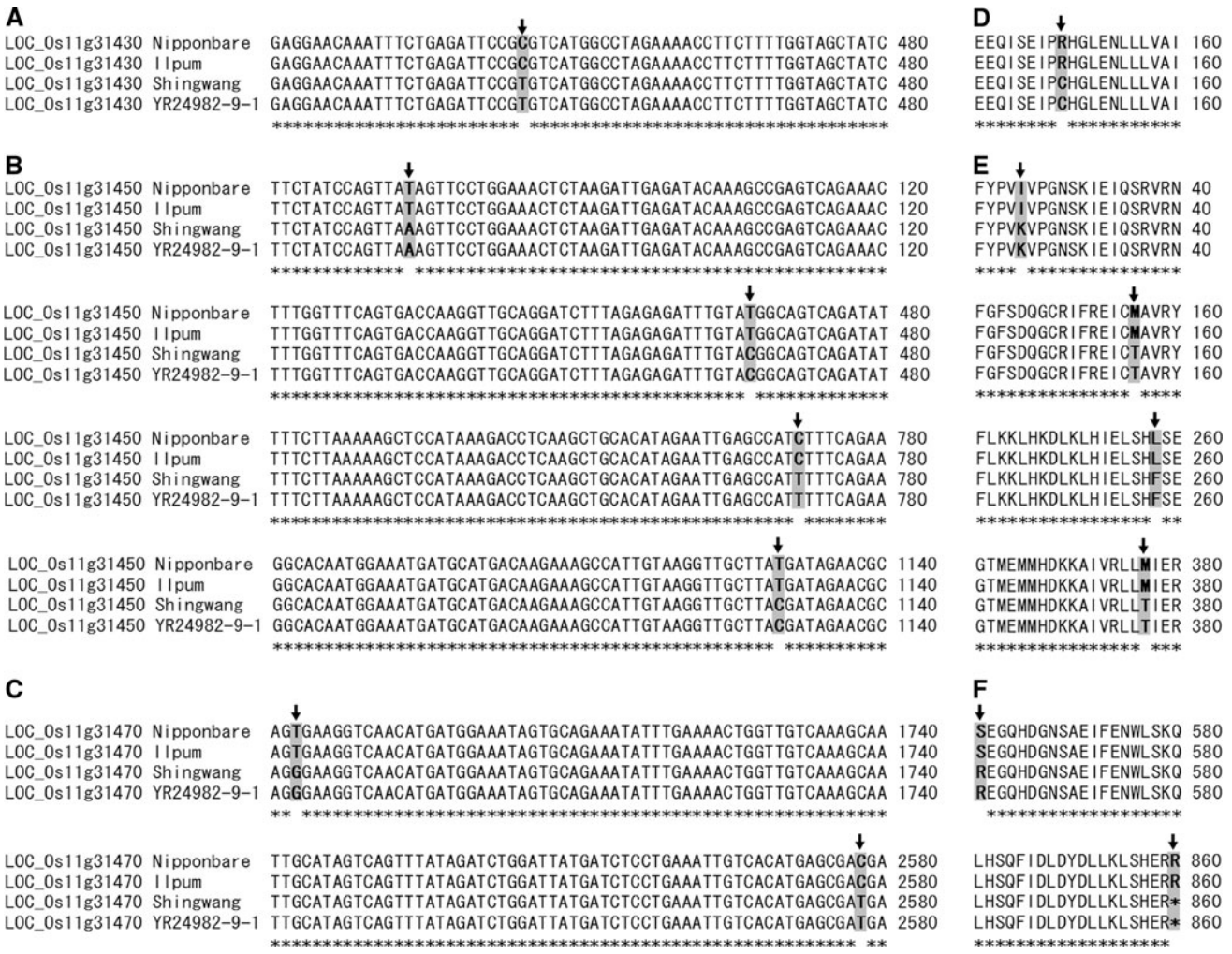


Fig. 7 Multiple sequence alignment of LOC_Os11g31430, LOC_Os11g31450 and LOC_Os11g31470 from the Nipponbare, Ilpum, Shingwang, and YR24982-9-1. **a–c** Alignment of the cDNA sequence of each candidate genes. **d–f** Alignment of the predicted amino acid sequence of the candidate proteins. *Arrows* represent single-base or amino acid substitutions

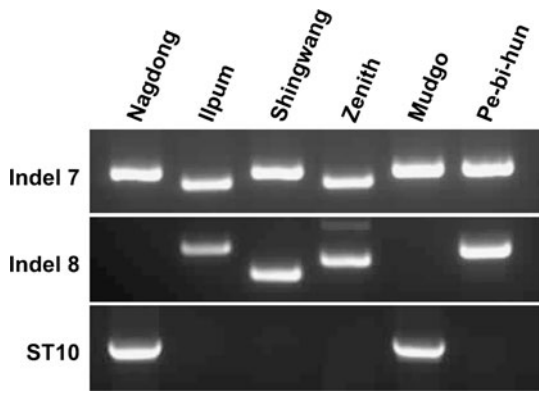


Fig. 8 Genotype analysis of rice varieties resistant to RSV using selected InDel markers

provided stable resistance to RSV since it was first introgressed into *japonica* paddy rice varieties. The *Stv-a* locus is linked to the glutinous endosperm (*wx*) loci on chromosome 6 (Hayano-Saito et al. 1998). Many QTLs have been identified near the *Stv-b* locus from resistant varieties including Kanto PL 3 (Ikeda and Kaneda 1982), Teqing (Wu et al. 2011), Milyang 23 (Maeda et al. 1999), and DV85 (Ding et al. 2005), but it is unclear whether they are *Stv-b*^{i}, *Stv-b* or a new allele as the actual sequence of the resistant gene has not been identified. Hayano-Saito et al. (1998) reported a *Stv-b*^{i} specific STS marker, ST10 and it can be used for PCR-assisted selection for stripe disease resistance of the *Stv-b*^{i} gene.

The allele type of InDel 7 could distinguish most of the RSV-resistant *japonica* and *indica* rice varieties in this study (Fig. 5). InDel 7 primer set was designed with an insertion/deletion of LOC_Os11g31500 (Table 5), which was found to target the same location as InDel 3 among ten markers developed by Wu et al. (2011). We analyzed whether a new InDel 7 marker could discriminate *Stv-b*^{i} by adapting it to rice varieties harboring the *Stv-b*^{i} allele

including Nagdong and Mudgo, and varieties harboring the *Stv-b* allele including Zenith (Hayano-Saito et al. 2000a; Washino et al. 1968). Both InDel 7 and ST10 could classify Nagdong and Mudgo as the same allele type (Fig. 8). Additionally, InDel 7 further differentiated Shingwang harboring *qSTV11*^{SG} and Pe-bi-hun harboring a resistance gene closely linked to the *Stv-b* locus (Ikeda and Kaneda 1982) from Zenith (*Stv-b*) and Ilpum (susceptible). By combining the results observed in Figs. 5 and 8, it is possible to infer that InDel 7 was able to distinguish *Stv-b*^{i} in both *indica* and *japonica* rice, whereas ST10 only did so in *japonica* rice varieties. These results also suggest that the resistance gene, *qSTV11*^{SG}, is likely to be *Stv-b*^{i}. The newly developed InDel 7 marker can be used in PCR-assisted selection of stripe disease resistance of *Stv-b*^{i} in both *indica* and *japonica* rice-breeding programs.

Expression analysis of 21 candidate genes in a 150 kb region harboring the *qSTV11*^{SG} locus revealed that three genes, LOC_Os11g31430, LOC_Os11g31450, and LOC_Os11g31470 were exclusively expressed in the susceptible variety, Ilpum, but not in the resistant varieties, Shingwang and resistant NIL, YR2498-9-1. It is unclear whether the differences in expression patterns of these genes between susceptible and resistant varieties resulted from altered stability of mRNAs caused by sequence variations found in the resistant variety or a difference in promoter efficiency. Nevertheless, expression profiles of these three genes were consistent with their quantitative nature along with incomplete dominance, which is clearly different from a single dominant resistance gene-mediated resistance exhibiting complete (qualitative) resistance in several crops (reviewed by Maule et al. 2007). Therefore, we hypothesize that *qSTV11*^{SG} might be a necessary plant factor that is required by the virus to complete the RSV infection cycle and to induce disease phenotypes in rice. Consistent with this hypothesis, recessive resistance genes mutated in host components for a step of the virus life cycle are more prevalent for resistance to viruses with an extremely limited genome size compared with fungal or bacterial pathogens. Some plant eIF4E proteins interact with the genome-linked viral proteins (Vpg) of *Potyvirus* spp., and this interaction is required for *Potyvirus* spp. to complete their infection processes (Schaad et al. 2000; Kang et al. 2005; Miyoshi et al. 2006; German-Retana et al. 2008). Disruption of the interaction between eIF4E and Vpg of *Potyvirus* spp. has been suggested as an underlying mechanism for recessive virus resistance associated with eIF4E (Léonard et al. 2000; Yeam et al. 2007). In rice, two recessive resistant genes to rice tungro virus and rice yellow mottle virus, respectively, have been identified as eIF4G genes (Lee et al. 2010; Albar et al. 2006). A rice mutant with a disrupted gene for a NAC-domain (a domain characterized by NAM, ATAF1, 2, and CUC2 transcription factors) (Olsen et al. 2004) protein is resistant to the *Rice dwarf virus* (RDV), suggesting that the gene is required for the RDV replication (Yoshii et al. 2004).

Among the three candidate genes for *qSTV11*^{SG}, LOC_Os11g31430 and LOC_Os11g31470 are closely related to each other (70 % identity) and are likely rice orthologs of the arabidopsis *HASTY* (*HST*) gene. The arabidopsis *HST* gene is the ortholog of mammalian exportin-5 (Exp5) which is the nuclear export receptor (Bollman et al. 2003) for exporting small RNAs including pre-miRNAs, mature miRNAs, and tRNAs into the cytoplasm (Calado et al. 2002; Bohnsack et al. 2004; Lund et al. 2004; Park et al. 2005). Unlike RDV-infected rice plants, RSV infection enhances the accumulation of some rice miRNA*s (star miRNA is expressed at lower levels compared to miRNA in the opposite arm of a hairpin) as well as induces novel phase miRNAs from miRNA precursors (Du et al. 2011). Moreover, the viral NS3 protein of RSV suppresses RNA interference (RNAi) signals of host defense mechanism (Xiong et al. 2009). The RHBV NS3 also interferes with the host RNAi pathway through binding to siRNAs as well as to miRNAs (Hemmes et al. 2007). These data indicate that RSV infection can alter RNAi machinery by manipulating host small RNA profiles. Consequently, we speculated that LOC_Os11g31430 and LOC_Os11g31470 may be involved in exporting miRNA*s, which are then degraded. Additionally, new phased miRNAs induced during RSV infection inhibit the general RNAi defense system of the plant or expression of virus-induced symptoms, although their nuclear RNA exporting function needs to be demonstrated empirically. More interestingly, among six HST/EXP5 orthologs in the rice genome, five genes clustered between 17884332 and 18239523 in chromosome 11 in which *qSTV11*^{SG} was mapped in this study. Further studies on the isolation of the candidate resistance genes in the 150-kb region will shed light on the molecular mechanism for RSV resistance.

Acknowledgments This study was supported by grants from the Rural Development Administration (PJ0086852012).

References

Albar L, Bangratz-Reyser M, Hébrard E, Ndjiondjop MN, Jones M, Ghesquière A (2006) Mutations in the eIF (iso)4G translation initiation factor confer high resistance of rice to Rice yellow mottle virus. Plant J 47:417–426

Bohnsack MT, Czaplinski K, Gorlich D (2004) Exportin 5 is a RanGTP-dependent dsRNA-binding protein that mediates nuclear export of pre-miRNAs. RNA 10:185–191

Bollman KM, Aukerman MJ, Park MY, Hunter C, Berardini TZ, Poethig RS (2003) HASTY, the Arabidopsis ortholog of exportin 5/MSN5, regulates phase change and morphogenesis. Development 130:1493–1504

Calado A, Treichel N, Muller EC, Otto A, Kutay U (2002) Exportin-5-mediated nuclear export of eukaryotic elongation factor 1A and tRNA. EMBO J 21:6216–6224

Chung BJ (1974) Studies on the occurrence, host range, transmission, and control of rice stripe disease in Korea. Kor J Plant Prot 13(4):181–204

Chung KY, Park RK, Chung KS, Lee SK, Jeon BT, Jin YD (1975) A new high yielding rice variety resistant to rice stripe virus 'Milyang 15 (Nagdong)'. Res Rept NYAES 17(c):17–24. (in Korean)

Ding XL, Jiang L, Liu SJ, Wan JM (2004) QTL analysis for rice stripe disease resistance gene using recombinant inbred lines derived from crossing of Kinmaze and DV85. Acta Genetica Sin 31:287–292

Ding XL, Jiang L, Zhang YX, Sun DZ, Zhai HQ, Wan JM (2005) Detection and analysis of QTL for resistance to stripe disease in rice, using backcross inbred lines. Acta Agron Sin 31(8):1041–1046 (in Chinese with English Abstract)

Du P, Wu J, Zhang J, Zhao S, Zheng H, Gao G, Wei L, Li Y (2011) Viral infection induces expression of novel phased microRNAs from conserved cellular microRNA precursors. PLoS Pathog 7:e1002176

Falk BW, Tsai JH (1998) Biology and molecular biology of viruses in the genus Tenuivirus. Annu Rev Phytopathol 36:139–163

German-Retana S, Walter J, Doublet B, Roudet-Tavert G, Nicaise V, Lecampion C, Houvenaghel MC, Robigalia C, Michon T, LeGall O (2008) Mutational analysis of plant cap-binding protein eIF4E reveals key amino acids involved in biochemical functions and *Potyvirus* infection. J Virol 82:7601–7612

Hayano-Saito Y, Tsuji T, Fujii K, Saito K, Iwasaki M, Saito A (1998) Localization of the rice stripe disease resistance gene, Stvb-i, by graphical genotyping and linkage analyses with molecular markers. Theor Appl Genet 96:1044–1049

Hayano-Saito Y, Saito K, Hujii K, Touyama T, Tsuji T, Sugiura N, Izawa T, Iwasaki M (2000a) SCAR marker for selection of the rice stripe resistance gene *Stvb-i*. Breed Res 2(2):67–72 (in Japanese with English Abstract)

Hayano-Saito Y, Saito K, Nakamura S, Kawasaki S, Iwasaki M (2000b) Fine physical mapping of the rice stripe resistance gene locus, Stvb-i. Theor Appl Genet 101:59–63

Hemmes H, Lakatos L, Goldbach R, Burgyan J, Prins M (2007) The NS3 protein of Rice hoja blanca tenuivirus suppresses RNA silencing in plant and insect hosts by efficiently binding both siRNAs and miRNAs. RNA 13:1079–1089

Ikeda R, Kaneda C (1982) Genetic relationships of brown planthopper resistance to dwarf and stripe disease resistance in rice. Jpn J Breed 32:177–185

Kang BC, Yeam I, Frantz JD, Murphy JF, Jahn M (2005) The *pvr1* locus in *Capsicum* encodes a translation initiation factor eIF4E that interacts with *Tobacco etch virus* VPg. Plant J 42:392–405

Kwak DY, Lee BC, Choi I, Yeo US, Cho JH, Lee JY, Song YC, Yun YN, Park DS, Kang HW, Nam MH, Lee JH (2011) Identification of a major QTL, $qSTV11^{SG}$, associated with resistance to rice stripe virus disease originated from Shingwangbyeo in rice (*Oryza Sativa* L.). Korean J Breed Sci 43(5):396–401

Lee BC, Yoon YN, Hong SJ, Hong YK, Kwak DY, Lee JH, Yeo US, Kang HW, Hwang HG (2008) Analysis on the Occurrence of Rice stripe virus. Res Plant Dis 14(3):210–213

Lee JH, Muhsin M, Atienza GA, Kwak DY, Kim SM, De Leon TB, Angeles ER, Coloquio E, Kondoh H, Satoh K, Cabunagan RC, Cabauatan PQ, Kikuchi S, Leung H, Choi IR (2010) Single nucleotide polymorphisms in a gene for Translation Initiation Factor (eIF4G) of rice (*Oryza sativa*) associated with resistance to rice tungro spherical virus. MPMI 23(1):29–38

Léonard S, Plante D, Wittmann S, Daigneault N, Fortin MC, Laliberetéé JF (2000) Complex formation between *Potyvirus* Vpg and translation eukaryotic initiation factor 4E correlates with virus infectivity. J Virol 74:7730–7737

Lu L, Du Z, Qin M, Wang P, Lan H, Niu X, Jia D, Xie L, Lin Q et al (2009) Pc4, a putative movement protein of rice stripe virus, interacts with a type I DnaJ protein and a small Hsp of rice. Virus Genes 38:320–327

Lund E, Guttinger S, Calado A, Dahlberg JE, Kutay U (2004) Nuclear export of microRNA precursors. Science 303:95–98

Maeda H, Nemoto H, Yagi T, Fukuta Y (1999) QTL analysis for rice stripe disease resistance using recombinant inbred lines (RILs) derived from crossing between Milyang and Akihikari. In: China. Association of Agricultural Science, China National Rice Research Institute, China National Hybrid Rice Research and Development Center, China Foundation for Agricultural Science and Education. Prospects of Rice Genetics and Breeding for the 21st Century: Paper Collection of International Rice genetics and Breeding Symposium. China Agricultural Science and Technology Press, Beijing, pp 53–57

Maeda H, Sugisawa T, Nemoto H, Sunohara Y (2004) QTL analysis for rice stripe resistance in the Japanese upland rice Kanto 72. Breed Sci 54:19–26

Maule AJ, Caranta C, Boulton MI (2007) Sources of natural resistance to plant viruses: status and prospects. Mol Plant Pathol 8:223–231

Miyoshi H, Suehiro N, Tomoo K, Muto S, Takahashi T, Tsukamoto T, Ohmori T, Natsuaki T (2006) Binding analyses for the interaction between plant virus genome-linked protein (VPg) and plant translational initiation factors. Biochemie 88:329–340

Nault LR (1994) Transmission biology, vector specificity and evolution of planthopper-transmitted plant viruses. In: Denno RE, Perfect TJ (eds) Planthopper, their ecology and management. Chapman and Hall, New York, pp 429–448

Olsen AN, Ernst HA, Leggio LL, Skriver K (2004) NAC transcription factors: structurally distinct, functionally diverse. Trends Plant Sci 10:1360–1385

Ou SH (1972) Virus diseases. In: Diseases Rice (ed) pp. Commonwealth Mycological Institute, Surrey, pp 14–22

Park MY, Wu G, Gonzalez-Sulser A, Vaucheret H, Scott Poethig R (2005) Nuclear processing and export of microRNAs in Arabidopsis. PNAS 102:3691–3696

Schaad MC, Anderberg RJ, Carrington JC (2000) Strain-specific interaction of the tobacco etch virus NIa protein with the translation initiation factor eIF4E in the yeast two hybrid system. Virology 234:84–92

Shen YJ, Jiang H, Jin JP, Zhang ZB, He YY, Wang G, Wang C, Qian L, Li X, Yu QB, Liu HJ, Chen DH, Gao JH, Huang H, Shi TL, Yang ZN (2004) Development of genome-wide DNA polymorphism database for map-based cloning of rice genes. Plant Physiol 135:1198–1205

Shimizu T, Toriyama S, Takahashi M, Akutsu K, Yoneyama K (1996) Non-viral sequences at the 59 termini of mRNAs derived from virus-sense and virus-complementary sequences of the ambisense RNA segments of rice stripe tenuivirus. J Gen Virol 77:541–546

Takahashi Y, Omura T, Shohara K, Tsuchizaki T (1991) Comparison of four serological methods for practical detection of ten viruses of rice in plants and insects. Plant Dis 75:458–461

Toriyama S (1995) Viruses and molecular biology of *Tenuivirus*. In: Singh RP, Singh US, Kohmoto K (eds) Pathogenesis and Host Specificity in Plant Diseases, Viruses and Viroids, vol 3. Pergamon, Oxford, p 211–223

Washino O, Toriyama K, Ezuka A, Sakurai Y (1968) Studies on the breeding of rice varieties resistance to stripe disease. III. Genetic studies on resistance to stripe in foreign varieties. Jpn J Breed 17:167–172

Washio O, Ezuka A, Sakurai Y, Toriyama K (1968a) Studies on the breeding of rice varieties resistant to rice stripe disease. II.

Genetic study on resistance to stripe disease in Japanese upland rice. Jpn J Breed 18:96–101

Washio O, Ezuka A, Sakurai Y, Toriyama K (1968b) Studies on the breeding of rice varieties resistant to rice stripe disease. III. Genetic studies on resistance to stripe in foreign varieties. Jpn J Breed 18:167–172

Washio O, Ezuka A, Sakurai Y, Toriyama K (1968c) Testing method for, genetics of and breeding for resistance to rice stripe disease. Bull Chugoku Natl Agric Exp Stn A16:39–197

Wu SJ, Zhong H, Zhou Y, Zuo H, Zhou LH, Zhu JY, Ji CQ, Gu SL, Gu MH, Liang GH (2009) Identification of QTLs for the resistance to rice stripe virus in the *indica* rice variety Dular. Euphytica 165:557–565

Wu X, Zuo S, Chen Z, Zhang Y, Zhu J, Ma N, Tang J, Chu C, Pan X (2011) Fine mapping of $qSTV11^{TQ}$, a major gene conferring resistance to rice stripe disease. Theor Appl Genet 122:915–923

Xie RK, Mao BH, Wang YD, Zhao Y, Zhen YL (2005) The incidence and control of rice stripe in *indica* hybrid rice. Hybrid Rice 20:48–49

Xiong R, Wu J, Zhou Y, Zhou X (2008) Identification of a movement protein of the tenuivirus rice stripe virus. J Virol 82:12304–12311

Xiong R, Wu J, Zhou Y, Zhou X (2009) Characterization and subcellular localization of an RNA silencing suppressor encoded by rice stripe tenuivirus. Virology 387:29–40

Xu SB, Tao YF, Yang ZQ, Chu JY (2002) A simple and rapid method used for silver staining and gel preservation. Hereditas 24:336–338

Yeam I, Cavatorta JR, Ripoll DR, Kang BC, Jahn MM (2007) Functional dissection of naturally occurring amino acid substitutions in eIF4E that confers recessive *Potyvirus* resistance in plants. Plant Cell 19:2913–2928

Yoshii M, Nishikiori M, Tomita K, Yoshioka N, Kozuka R, Naito S, Ishikawa M (2004) The *Arabidopsis Cucumovirus Multiplication 1* and *2* loci encode translation initiation factors 4E and 4G. J Virol 78:6102–6111

Zhang SX, Li L, Wang XF, Zhou GH (2007) Transmission of rice stripe virus acquired form frozen infected leaves by the small brown planthopper (Laodelphax striatellus Fallen). J Virol Methods 146:359–362

Zhao F, Cai Z, Hu T, Yao H, Wang L, Dong N, Wang B, Ru Z, Zhai W (2010) Genetic analysis and molecular mapping of a novel gene conferring resistance to rice stripe virus. Plant Mol Biol Rep 28:512–518