THESIS APPROVAL

GRADUATE SCHOOL, KASETSART UNIVERSITY

Doctor of Philosophy (Plant Breeding)

DEGREE

Plant Breeding

FIELD

Interdisciplinary Graduate Program

PROGRAM

TITLE: High-resolution Mapping of a Brown Planthopper (BPH) Resistance Gene, *Bph3*, and Marker-assisted Selection for BPH Resistance in Rice

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THESIS

HIGH-RESOLUTION MAPPING OF A BROWN PLANTHOPPER (BPH) RESISTANCE GENE, *Bph3*, AND MARKER-ASSISTED SELECTION FOR BPH RESISTANCE IN RICE

JIRAPONG JAIRIN

A Thesis Submitted in Partial Fulfillment of
the Requirements for the Degree of
Doctor of Philosophy (Plant Breeding)
Graduate School, Kasetsart University
2008

Jirapong Jairin 2008: High-resolution Mapping of a Brown Planthopper (BPH) Resistance Gene, *Bph3*, and Marker-assisted Selection for BPH Resistance in Rice. Doctor of Philosophy (Plant Breeding), Major Field: Plant Breeding, Interdisciplinary Graduate Program. Thesis Advisor: Mr. Theerayut Toojinda, Ph.D. 148 pages.

The brown planthopper (BPH) is one of the most destructive insect pests of rice in Thailand. We performed a cluster analysis that revealed the existence of four groups corresponding to the variation of virulence against BPH resistance genes in 45 BPH populations collected in Thailand. Rice cultivars Rathu Heenati and PTB33, which carry *Bph3*, showed a broad spectrum resistance against all BPH populations. The simple sequence repeat analysis was performed to identify and localize the *Bph3* gene. Based on the linkage analysis of 208 BC_1F_2 and 333 BC_3F_2, from crosses of PTB33×RD6 and Rathu Heenati×KDML105, respectively, we were able to map the *Bph3* locus on rice chromosome 6. Physical mapping of *Bph3* was further performed using a BC_3F_3 population derived from a cross between Rathu Heenati and KDML105. According to the genome sequence database of Nipponbare, the *Bph3* locus was finally localized approximately in a 190 kb interval flanked by markers RM19291 and RM8072.

Introgression lines (ILs) with brown planthopper resistance and KDML105 grain quality characteristics were successfully developed by the integration of phenotypic and marker assisted selections in three generations of backcrossing. The linkage drag between the *Bph3* and Wx^a allele was successfully dissected and the BPH resistance gene was introgressed into the KDML105 genetic background. The improved lines were not only showed the excellent cooking and eating quality of the milled rice but they also expressed a broad spectrum resistance against BPH populations in Thailand. The ILs developed in this study will have an impact on the yield stability and sustainability in KDML105-producing areas. Additionally, the ILs can be used as genetic resources of BPH resistance to improve rice varieties with the Wx^b allele in breeding programs.

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Student's signature Thesis Advisor's signature

ACKNOWLEDGMENTS

I wish to express my appreciation and deepest gratitude to my advisor, Dr. Theerayut Toojinda, for his kindness, generous help, suggestion and criticism of my thesis research and thank him for his constructive guidance, patience and continuous encouragement throughout my study. I wish to express my sincere thanks to Associate Prof. Dr. Apichart Vanavichit for the excellent advice and strong support to finish my study.

The financial support for my research work was provided by the Rockefeller Foundation through the Kasetsart University is most gratefully acknowledged. I am also grateful to Ubon Ratchathani Rice Research Center, Rice Department for allowing me to have three years scholarship. I would like to thank the International Rice Research Institute and Phitsanulok Rice Research Center for supplied seeds and insect screening.

I am especially appreciated Dr. Poonsak Mekwatanakarn, Dr. Varapong Chamarerk and Dr. Payorm Cobelli for their supported and suggestion. I am most grateful to Mr. Sa-nguan Teangdeerith, Mrs. Phikul Leelagud, Ms. Kanlaya Sansen, and Mr. Jate Kothcharerk for valuable helping to evaluate the mapping populations, special thanks to Ms. Myint Yi, for her helping to evaluate rice grain quality. A very special word of thanks goes to several staff members and all students in the Rice Gene Discovery Unit for their kindness assisted toward me.

Finally, I wish to express special appreciation and gratitude to my family and all friends for their love and care, many support, understanding and patience throughout the preparation of the thesis.

Jirapong Jairin

September 2008

TABLE OF CONTENTS

Page

LIST OF TABLES

LIST OF TABLES (Continued)

LIST OF TABLES (Continued)

LIST OF FIGURES

LIST OF FIGURES (Continued)

LIST OF FIGURES (Continued)

LIST OF FIGURES (Continued)

HIGH-RESOLUTION MAPPING OF A BROWN PLANTHOPPER (BPH) RESISTANCE GENE, *Bph3,* AND MARKER-ASSISTED SELECTION FOR BPH RESISTANCE IN RICE

INTRODUCTION

Cultivated rice (*Oryza sativa* L.), a derivation of several Thai cultures, is the most important source of carbohydrate for Thais as well as for Asian population. Increasing rice production within the limit of paddy fields for the continued expansion of world population is a challenge to scientists. While improving yield potential genotypes can increase rice production, improving biotic stress genotypes can maintain the stability of rice yield. The widespread damage caused by insect pests constitutes the most significant factors leading to substantial and unpredictable decrease in rice yield. The brown planthopper (BPH), *Nilaparvata lugens* Stål, is one of the most serious insect pests of rice in Thailand. Continuous rice culture, extensive use of insecticides and high rate of nitrogen fertilizer application often cause outbreaks of BPH in rice fields. BPH can cause serious yield reduction by direct damaging and susceptible rice cultivars often suffer severe damage. The removal of assimilates and reduction in photosynthetic rate of leaves by BPH feeding has the greatest effect on growth and yield on rice plant. Plant death can occur if the amount of energy supplied is less than that required for tissue maintenance (Watanabe and Kitagawa, 2000; Yuan *et al*., 2005). In addition to feed on rice plant directly, BPH also causes indirect damage by transmitting viruses, which cause ragged and grassy stunt diseases (Heinrichs, 1979). One strategy for minimizing losses due to BPH is the utilization of BPH resistance genes. Consequently, breeding BPH resistant cultivar is an objective to stabilize the yield production of rice.

Developing resistant rice varieties is generally considered to be the most economic and effective way for controlling BPH. Rice plant resistance to BPH is recognized as qualitative and quantitative traits. The genetic basis of the qualitative and quantitative BPH resistance has been well studied and 21 major resistance genes

have been discovered from cultivated varieties and wild relatives. Of these genes, 17 resistance genes have been assigned to rice chromosomes (Zhang, 2007). More than half of the discovered major resistance genes could not be used against some BPH populations found in Thailand (Jairin *et al*., 2005b). Among these, *Bph1*, *bph2*, *Bph3* and *bph4* have been used extensively in Thai breeding programs. Improved rice cultivars carrying *Bph1*, *bph2* and *bph4*, however, have lost their ability against BPH in most of rice growing areas in Thailand. Only rice cultivars carrying *Bph3* have shown a higher degree and broader spectrum of resistance against the BPH. Breeding resistant cultivar with major resistance genes was highly successful, however, BPH itself also successfully adapt to feed on the resistant cultivars by changing their biotypes. The occurrence of new virulent biotypes has been a serious problem in breeding resistant rice cultivar against BPH. Identification and incorporation of new BPH resistance genes into rice cultivars is an important breeding strategy to control the damage caused by new biotypes of BPH (Jena *et al*, 2006). Therefore, selection of BPH resistance genes for improving resistant cultivars needs to be considered carefully.

Rice cultivars PTB33 and Rathu Heenati demonstrate resistance to all BPH biotypes identified at IRRI and in some field populations in Asia, including India, Philippines, Vietnam, China, Bangladesh, Laos, and Thailand (Angeles, *et al*., 1986; Jairin *et al*., 2005b; Khush, 1984; Li *et al*., 2002; Soundararajan *et al*., 2004; Velusamy *et al*., 1995). The dominant BPH resistance gene *Bph3* was first identified in cultivars Rathu Heenati (acc. no. 11730) and PTB33 (acc. no. 19325) (Ikeda, 1985; Lakshminarayana and Khush, 1977). PTB33 was found to carry two major BPH resistance genes, *bph2* and *Bph3*, and the inheritance of the digenic control of the resistance to BPH in PTB33 has been confirmed (Angeles *et al*., 1986). The gene *Bph3* was reported to be tightly linked to a recessive resistance gene, *bph4*, in cultivar Babawee (Ikeda and Kaneda, 1981; Sidhu and Khush, 1979). The study of genetic analysis by classical genetic approach of *Bph3* was shown to be closely linked to *bph4* in rice cultivar Babawee because no recombinants between these genes were observed among nearly 1,200 of F_3 progenies (Sidhu and Khush, 1979). This allelic relationship has been confirmed (Angeles *et al*., 1986). These two allelic resistance genes were

first assigned to rice chromosome 10 based on trisomic analysis (Ikeda and Kaneda, 1981). However, a recent fluorescence in situ hybridization study found that *Bph3* was physically localized on rice chromosome 4 (Yan *et al*., 2002). Furthermore, a major BPH resistance gene in cultivar Rathu Heenati was assigned to rice chromosome 4 (Sun *et al*., 2005). However, *bph4* from cultivar Babawee, which linked to *Bph3*, has been assigned to the short arm of rice chromosome 6 (Kawaguchi *et al*., 2001). According to the previous publications, there is now possibility that *Bph3* can be located on chromosome 4, 6 or 10. Our study should provide new information to confirm the location of *Bph3* on rice chromosome.

Map-based cloning represents one possible approach to isolate BPH resistance genes and elucidating the BPH resistance mechanism in rice. Recently, the publicly available rice genome sequence information has made map-based cloning in rice much more efficient to get the target genes. Three BPH resistance genes, *Bph15*, *Bph18* and *Bph19*, have been finely mapped on chromosome 3, 4 and 12, respectively (Chen *et al*., 2005; Jena *et al*., 2006; Yang *et al*., 2004). *Bph15* was finely mapped to a genomic segment of approximately 47 kb long flanked by restriction fragment length polymorphism (RFLP) markers RG1 and RG2 (Yang *et al*., 2004). The *bph19* locus was physically defined to an interval of about 60 kb flanked by simple sequence repeat (SSR) markers RM6308 and RM3134 (Chen *et al*., 2005), The *Bph18* locus was also finely localized within an 843 kb physical interval that includes three BAC clones between the sequence tagged site (STS) marker R10289S and SSR marker RM6869 (Jena *et al*., 2006). Although BPH resistance genes have been intensively discovered and studied throughout the rice genome, until recently none of the BPH resistance gene has been cloned and our current knowledge about insect resistance genes in rice plant is still limited. In this study, the construction of a high-resolution linkage map with SSR markers is a crucial step in map-based cloning of *Bph3*.

The *indica* rice cultivar KDML105 is characterized by its good eating quality with desirable fragrance and has been accepted in markets as premium jasmine rice. Additionally, the cultivar can widely adapt under rainfed lowland areas in Northeast of Thailand. Thus, KDML105 has been extensively used as a favorable quality

parental line to develop new cultivars. One limitation of this cultivar, however, is its susceptibility to brown planthopper (BPH), *Nilaparvata lugens* Stål, a major insect pest in rice-producing areas. One strategy to minimize losses due to BPH is the utilization of BPH resistance genes.

Tagging and mapping of BPH resistance genes in rice have been widely studied. To date, the number of major genes conferring BPH resistance in several cultivated and wild species has been identified and mapped with DNA markers, which facilitate marker-assisted selection (MAS) for BPH resistance in rice (Chen *et al*. 2006; Huang *et al*. 2001; Ishii *et al*. 1994; Jena *et al*., 2003; 2006; Liu *et al*., 2001; Murai *et al*., 2001; Murata *et al*., 2001; 2003b; Su *et al*., 2002; Sun *et al*. 2005; 2006; Wang *et al*., 2001; Yan *et al*. 2002; Yang *et al*., 2002; 2004). Molecular markers have been proven very useful in improving backcross breeding through precise transfer of target genomic regions. Additionally, markers allow us to estimate the genomic composition and can speed up the recipient genome recovery via background selection (Hospital, 2001). The success of using MAS to introgress BPH resistance genes from wild species into cultivated rice has been reported (Jena *et al*., 2006; Sharma *et al*., 2004). *Bph3*, one of the major BPH resistance genes, has shown a broad spectrum of resistance against BPH populations in Thailand. This cultivar has been used as a donor of BPH resistance in various conventional breeding programs. However, a poor grain quality such as high amylose content, low gel consistency, chalky endosperm and no fragrance limited the success of breeding lines. Improvement of good cooking and eating varieties via MAS has been intensively applied in rice breeding programs throughout Asian countries (Amarawathi *et al*., 2008; Liu *et al*., 2006; Toojinda *et al*., 2005; Zhang, 2007; Zhang *et al*., 2005; Zhou *et al*., 2003). According to our study, a major BPH resistance gene in Rathu Heenati was linked to the *Waxy* locus. The *Waxy* locus and the tightly linked genomic region on the short arm of chromosome 6 has been reported to control the eating and cooking quality determined by the physical and chemical properties of the starch in the endosperm especially the amylose synthesis of rice (Itoh *et al*., 2003; Lanceras *et al*., 2000; Zhou *et al*., 2003). Because the genetic dominance of the unflavored characteristics of the quality traits, especially amylose content, resistant progenies

carrying the *Waxy* allele of Rathu Heenati have high amylose content. To breed the BPH resistance using the *Bph3* allele, linkage drag will be causing a less success in low amylose rice cultivars. It is not only difficult to develop resistant line with low level of amylose content but also it will take longer time via conventional approaches to remove the linkage drag.

In this study to clarify and confirm the map position of the *Bph3* locus, we attempt to determine the *Bph3* locus on the rice linkage map using two backcross populations and simple sequence repeat (SSR) markers. We report the fine mapping of the *Bph3* locus to an approximately 190 kb target region on rice chromosome 6 using SSR markers. The SSR markers co-segregated with the *Bph3* locus were further used to determine for the presence of the *Bph3* gene in MAS. The phenotypic and marker-assisted selections were performed to break down the linkage drag and we were successful in introgression the *Bph3* allele from Rathu Heenati into KDML105 genetic background. Promising lines with good eating and cooking quality and BPH resistance are our expected result.

OBJECTIVES

1. To determine the variation of BPH populations in Thailand.

2. To identify and construct high-resolution mapping of a broad spectrum BPH resistance gene, *Bph3*, derived from rice cultivars Rathu Heenati and PTB33.

3. To develop rice introgression lines with brown planthopper resistance and KDML105 grain quality characteristics by the integration of phenotypic and marker-assisted selection.

LITERATURE REVIEW

1. Brown planthopper and its biology

The brown planthopper (BPH), *Nilaparvata lugens* Stål, (Homoptera: Delphacidae) (Figure 1) is a phloem-feeding insect pest of rice plants. BPH has become a major insect pest of rice since 1970s, with the extensive cultivation of high yielding varieties, high application rate of nitrogen fertilizer and extensive use of insecticides. BPH is widely distributed in rice growing areas throughout South and Southeast Asia. Intensive infestations have been reported from India, Sri Lanka, Bangladesh, Thailand, Myanmar, Vietnam, Cambodia, Malaysia, Philippines, Taiwan, Indonesia, Brunei, Papua New Guinea, and China. BPH is also found in East Asia (Japan, Korea), The South Pacific Islands, and Australia (Dyck and Thomas, 1979; Khush, 1979; Tanaka, 1997).

BPH is dimorphic with fully winged 'macropterous' (Figure 2a, 3a) and truncate-winged 'brachypterous' forms (Figure 2b, 3b). The macropterous is potentially migrants for colonizing new fields when the food is limited or some other environmental factors are unsuitable (Kisimoto, 1965; Pathak, 1968). Length of macropterous male is about 2.3-2.4 mm, female 2.8-3.2 mm, brachyterous male 2.0-3.1 mm, female 2.7-3.5 mm, post-tibial spur with 30-36 teeth (Okada, 1977). Genetic basis of wing polymorphism in BPH generally is presumed to be under polygenic control (Denno and Roderick, 1990; Denno *et al*., 1995). It is assumed that genes determine the level of the juvenile hormone leading to the development of wing forms (Ayoade *et al*., 1996; Bertuso and Tojo, 2002). Wing form in BPH is determined by a developmental switch that responds to environmental cues (Denno *et al*., 1995). Population density experienced during the nymphal stage, rice plant stage and nitrogen content of rice plants are the most of important environmental factors affecting wing determination in BPH (Kisimoto, 1965; Iwanaga *et al*., 1985; Syobu *et al*., 2002).

Oviposition by BPH always took place following stylet penetration into rice plants. The oviposition sequence of BPH can be divided into several main behavioral phases: forward thrusts by the apex of the ovipositor, penetration of the ovipositor with a sawing motion, release of the egg, and partial or full withdrawal of the ovipositor (Hattori and Sogawa, 2002). The eggs are usually laid as egg-groups, often in rows in the tissue of the lower part of the rice plant, mainly in sheaths. When the adult population is high, eggs are found in the upper parts of rice plants. The egg groups can be found in leaf blades and young panicles especially in a high population condition, on nitrogen limit plants or on the resistant plant (Figure 4). Some plant chemicals may effect the oviposition during stylet penetration prior to ovipositor penetration (Hattori and Sogawa, 2002). The number and ovipositor sites depend largely on the development stage of the rice plant. The egg-laying sites appear as brownish streaks. Red eye spots appear at one end of the egg before hatching. The egg stage is about 7 to 10 days in the tropics. It is also depend on the temperature. The duration of egg stage is found to be 26.7, 15.2, 8.2, 7.9, and 8.5 days at 15, 20, 25, 28, and 29°C constant, respectively. The shortest development time was at about 28°C (Mochida and Okada, 1979). The hatchability and survival rate are the highest around 25°C (Henrichs, 1994).

The newly hatched nymph is cottony white and turns purple brown (Figure 1b) within an hour and the length of the nymphs around 0.6 mm upon hatching (Feakin, 1974). BPH has five nymphal stadia, which are distinguished by shape of the mesonotum and metanotum, and body size (Figure 5). The nymphal stage is about 10 to 15 days. The development of nymph stage is about 18.2, 13.2, 12.6, 13.1, 17.0, and 18.2 days of a constant temperature of 20, 25, 29, 31, 37, and 35°C, respectively. The adult stage persisted for 16-17 days (Mochida and Okada, 1979).

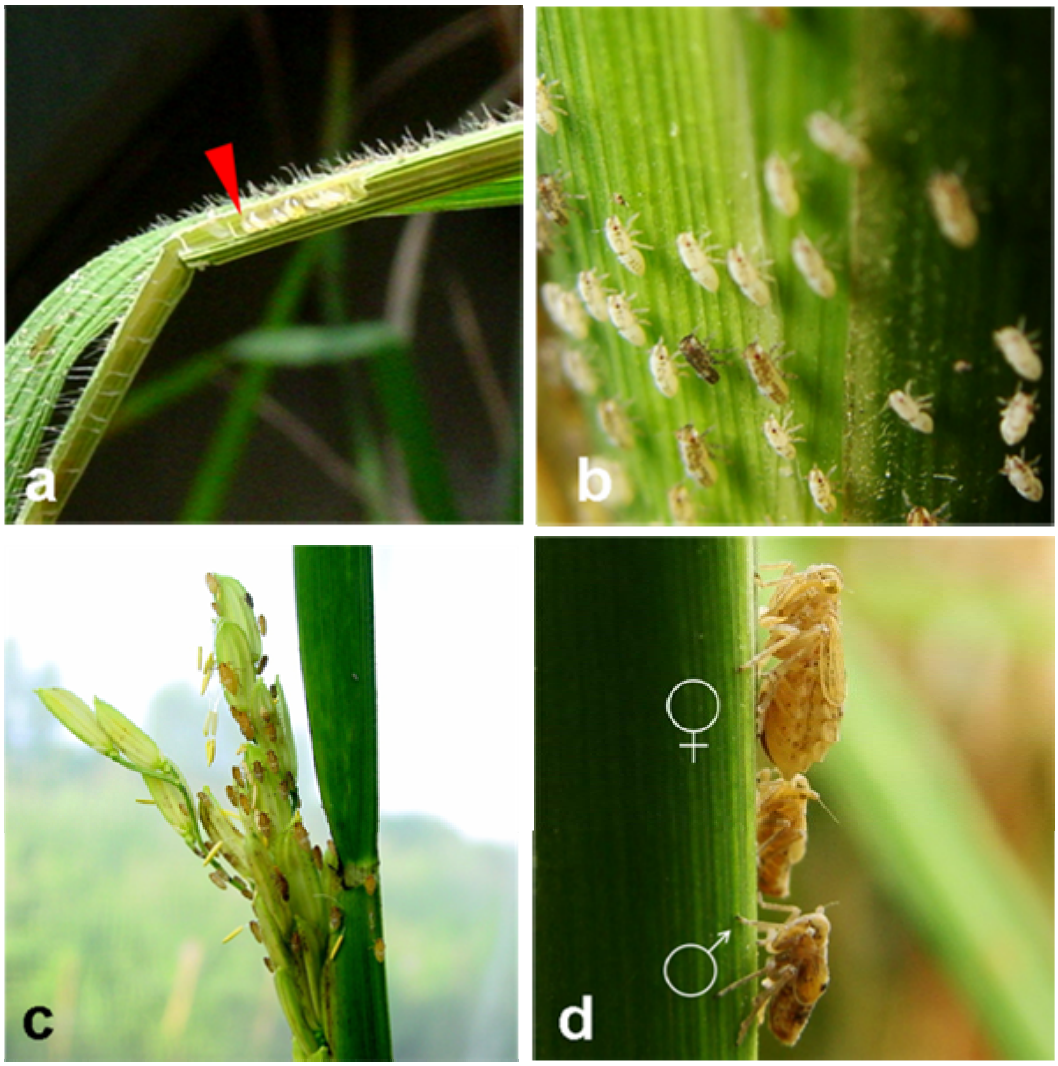

Figure 1 Brown planthopper on rice plants: (a) eggs, (b) newly hatched nymphs, (c) 4^{th}-5^{th} nymphal stages and (d) female and male adults

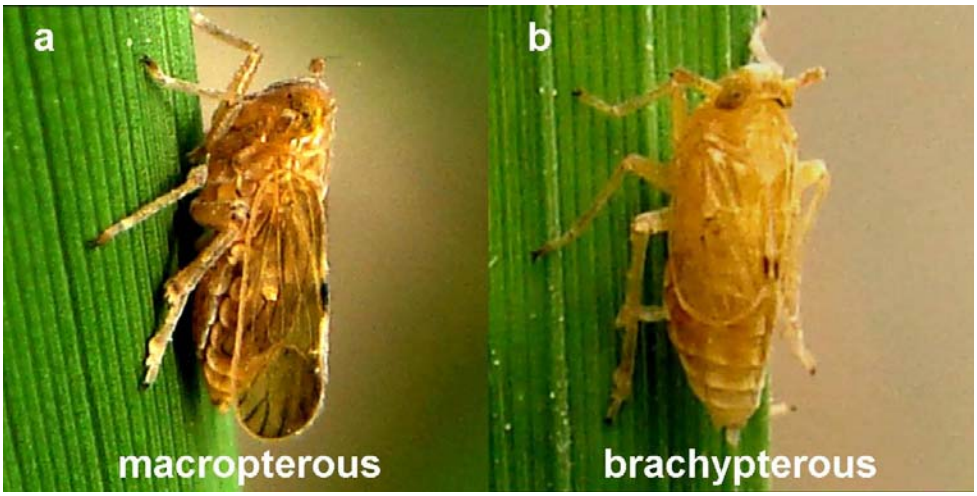

Figure 2 BPH adult female (a) marcorterus (fully-winged forms) and (b) bachyterus (truncate-winged forms)

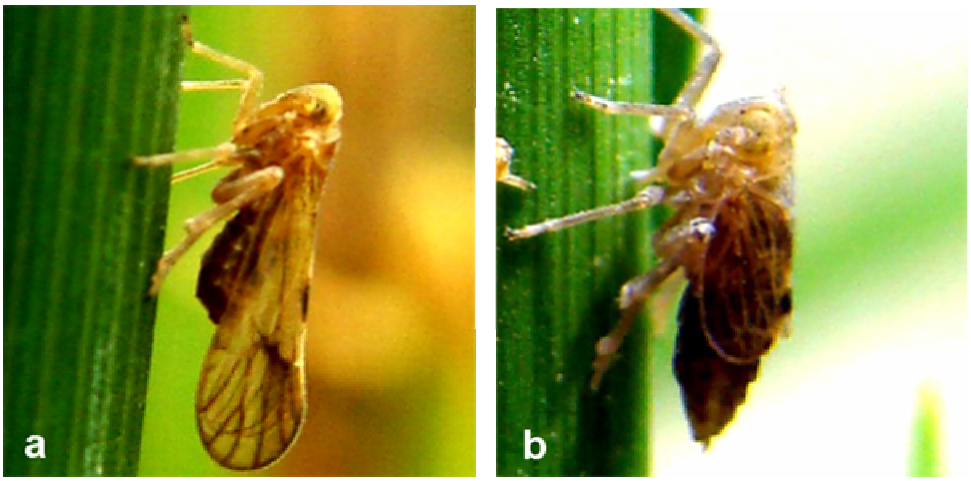

Figure 3 BPH adult male (a) marcorterus (fully-winged forms) and (b) bachyterus (truncate-winged forms)

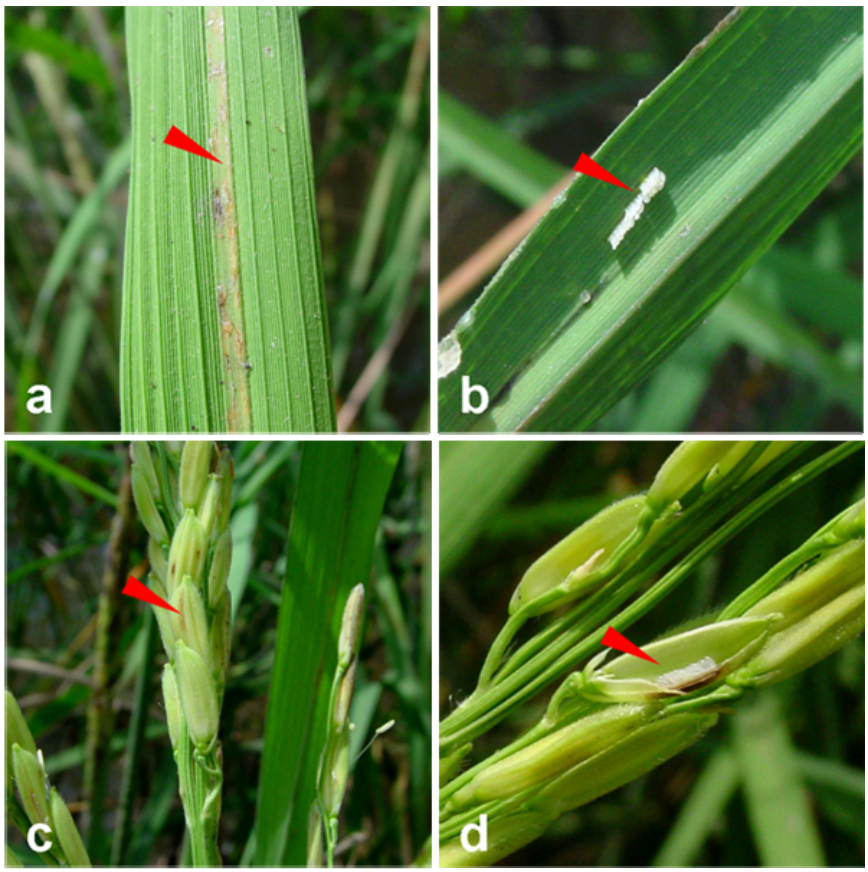

Figure 4 BPH females lay their eggs on the upper part of rice: (a,b) on the leave, and (c,d) on the panicles

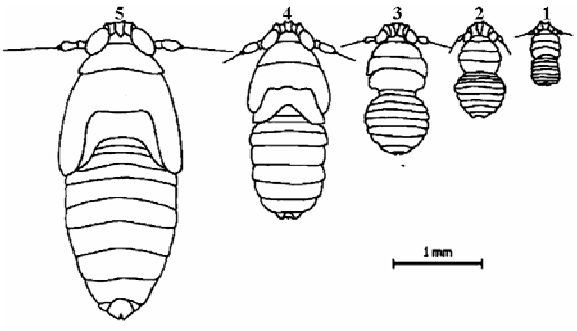

Figure 5 Five nymphal stages of BPH

Source: Mochida and Okada (1979)

2. Damage from the BPH feeding

BPH is a phloem sap-feeding insect that mostly feeds on the leaf sheath of rice plants and ingests nutrients using its piercing mouthparts or stylet (Figure 6). The feeding process of BPH can be divided into two phases including (i) probing, which is performed in parenchymal tissues, and (ii) sucking, which is done after stylets insertion into vascular bundles (Sōgawa, 1982). During feeding process, BPH salivary stylets penetrate rice plant tissues and form stylet or salivary sheaths to feed on photo assimilates translocation in the phloem sieve elements. In general, phloem sap-feeding insects secrete a watery saliva that is continuously secreted during feeding may interact with phloem proteins to prevent their coagulation (Tjallingii, 2006; Will and van Bel, 2006) or may also contain effectors that modulate plant defense responses (Hao *et al*., 2008; Miles, 1999). The stylet bundle and salivary sheath are frequently found entirely within the wall of a plant cell and it can be seen that saliva may penetrate the cell wall into an adjacent cell. Therefore, BPH feeding behavior is very damaging to the host plant, even in those penetrations which do not result in prolonged uptake of phloem sap (Spiller, 1990).

Both the nymphs and adults of BPH feed on the leaf sheaths at the basal portion of the rice plants. In most cases the BPH severely damages rice plants in the post-flowering stage. Feeding by this species can cause plant death. The typical sucking damage caused by BPH is commonly referred to as “hopperburn” (Figure 7), which has been studied and reviewed (Backus *et al*., 2005; Denno and Roderick, 1990; Sōgawa, 1982; Watanabe and Kitagawa, 2000). The first symptom of hopperburn injury appears as yellowing of the older leaf blades (chlorosis), that extends progressively, and finally the whole plant turns brown and wilts (Sōgawa, 1982). In the paddy fields, hopperburn usually appears as a browning of plants in scattered patches (Figure 7). In severe cases the patches spread rapidly on a large scale (Sōgawa, 1982). Although, until recently, mechanisms of hopperburn from the feeding of BPH is not completely understood. From the broadly studied can be suggested that combined effects of reduction of water and photoassimilate translocation, salivary composition and salivary sheath of BPH in plant tissues

probably involve and cause initiation of hopperburn symptom (Backus *et al*., 2005; Watanabe and Kitagawa, 2000). BPH feeding may reduce yield, even if the planthopper population density is not high enough to kill rice plants (Watanabe and Kitagawa, 2000). Although chlorosis is more visible, the most important symptom for yield reduction is reduced growth (stunting) and reproduction (Backus *et al*., 2005). The removal of assimilates and reduction in photosynthesis by BPH have the greatest effect on growth and yield of rice plants as compared with the disruption in the translocation of assimilates. Plant death can occur by BPH infestation, if the amount of energy supplied is less than that required for tissue maintenance (Watanabe and Kitagawa, 2000). In addition to feeding on rice plants directly, BPH also causes indirect damage by acting as vector for the viruses, which cause ragged and grassy stunt diseases (Heinrichs, 1979).

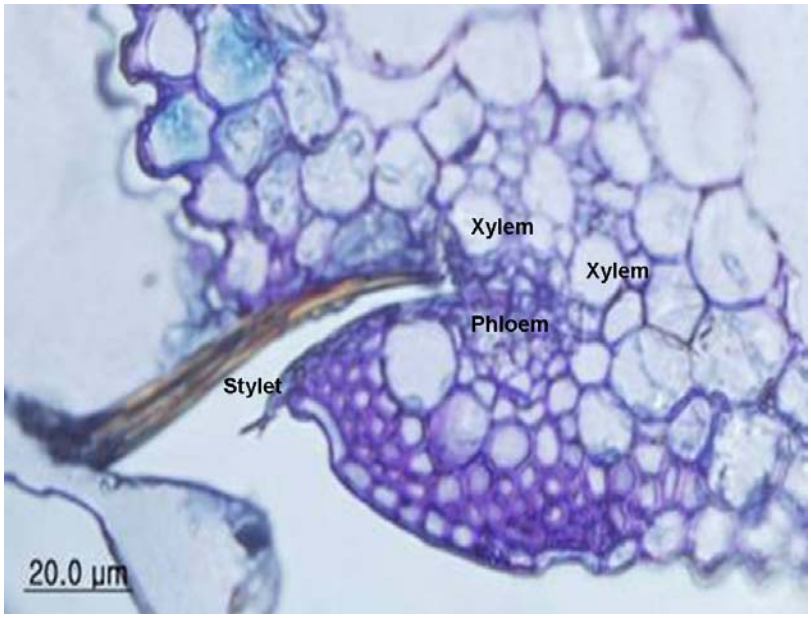

Figure 6 Cross section of rice plant vascular bundle with stylet of brown planthopper
Source: Seo *et al*. (unpublished)

Figure 7 The typical damage ‘hopperburn’ caused by BPH in rice fields

3. Symbiotic microorganisms in BPH

Phloem sap has a poor nutrition and unbalanced amino acid composition, with a high carbon/nitrogen ratio, high amount of nonessential amino acids, deficient in essential amino acids and low levels of lipids and vitamins. There is now substantial evidence that the microorganisms in phloem-feeding insects synthesize essential amino acids that are made available to the insect host supplementing of phloem sap, which is deficient in essential amino acids (Baumann *et al*., 1995; Douglas, 1989; 1998). All phloem-feeding members of the Homoptera possess symbiotic microorganisms. Planthoppers harbor yeast-like symbionts in the mycetocyte cell. The yeast-like symbiotes, harbored in BPH fat body cell, play a pivotal role in nitrogen metabolism and nitrogen recycling in the brown planthopper (Sasaki *et al*., 1996; Hongoh and Ishikawa, 1997) and the yeast-like symbiotes were transmitted to the next generation by the transovarial infection and proliferated by asexual budding (Cheng and Hou, 2001). There is also considerable evidence that the virulence of BPH populations to resistant rice varieties was related to abundance of yeast-like symbiotes (Lu *et al*., 2004).

4. Biotypes of BPH

Biotypes occur in nature as products of survival mechanism for the persistence of insect species (Nielson and Lehman, 1980). A biotype of the BPH is generally referred to as a population which has a specific ability or inability to infest and survive on rice varieties with specific genes for resistance to BPH (Sōgawa, 1981). Quantitative genetic analysis of biotype of BPH revealed that the virulence of BPH is under polygenic control (Hollander and Pathak, 1981; Tanaka, 1999). It is important to understand the biotype of BPH populations to have targeted development in breeding for BPH resistance. Four biotypes, biotype 1, 2, 3 and 4, have been designated based on the responses on BPH resistant varieties. The first BPH resistant variety was Mudgo, which identified by Pathak *et al*. (1969). It was found to be resistance to BPH population prevalent in the Southeast Asia but not in the South Asia. Thus, two biotypes of BPH existed before introduction of resistant varieties.

Biotype 3 was developed in the laboratory by rearing the insects on the resistant variety ASD7 that has the *bph2* gene for resistance (Panda and Khush, 1995). The rice varieties that have *bph2* gene were found to be susceptible to the South Asian biotype, called biotype 4 (Khush, 1992) but the varieties which have *Bph3* gene can resistance to this biotype. Therefore, the population that cannot infest any varieties with resistant genes is called biotype 1 while those populations infesting resistant varieties carrying *Bph1* and *bph2* genes were described as biotypes 2 and 3, respectively (Figure 8).

Biotypes of the BPH in Thailand have been studied since 1975 (Pongprasert and Weerapat, 1979). In order to previous studies, the results indicated that the BPH collected from the North and Northeast of Thailand were different from biotype 1, 2, 3 and 4 (Phengrat, 2000; Rithmontri *et al*., 1998; Tripop, 1996). Recently, at least 4 biotypes of BPH in Thailand have been reported (Jairin *et al*., 2007a). However until then the new biotype in Thailand was not classified and no biotype destination have been given to them.

5. Mechanisms of rice plant resistance to BPH

The concept of host plant resistance to insect pests was first proposed into three categories; antibiosis, non-preference (antixenosis) and tolerance (Painter, 1941). Antibiosis is the resistance mechanism of the plants that affect growth and survival after the insects start utilizing the plants. Antixenosis is the resistance mechanism exhibited by plants to deter colonization by insects. Tolerance is a plant ability to recover from the damage caused by the infestation with insect pests that badly damage susceptible plants (Panda and Khush, 1995). Although all mechanism of resistance can delay the time required for the insect populations to reach the economic damage level, plants with tolerance mechanism can not affect the rate of population increase of the insect while antibiosis can decrease insect populations within a few generations. Therefore, we should pay attention on these aspects when developing resistant variety against the BPH.

Various chemical substances have been reported to be present in phloem sap of rice plant that promotes BPH to stimulate or prevent sucking. Several amino acids, asparagines, glutamic acid, alanine, serine, leucine, aspartic acid, valine, were reported to be sucking stimulants (Shigematsu *et al*., 1982; Sōgawa, 1974; 1976; Sakai and Sōgawa, 1976; Sōgawa, 1982; Yoshihara *et al*., 1979). Several flavoniods and organic substances, transaconit acid, oxalic acid, tricin and 3-nitraphthalic acid have been reported to act as BPH sucking inhibitors (Bing *et al*., 2007; Zhang *et al*., 1999). Recently, Hao *et al*. (2008) found that the induced callose sealing in sieve elements plays an important role in preventing BPH from ingesting phloem sap. Although the effect of BPH feeding on physiological properties and metabolic changes in rice plants have been reported from many studies, the mechanism of rice plants resistance to BPH is still uncertain. Consequently, the mechanism of resistance is required further investigation.

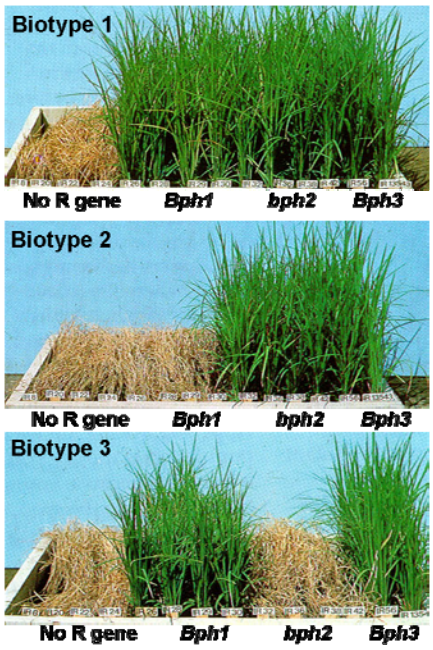

Figure 8 Resistance of rice to different biotypes of brown planthopper: biotype 1 damages varieties with no major resistance genes, biotype 2 damages varieties with the *Bph1* gene and biotype 3 damages varieties with the *bph2* gene.

Source: Pathak and Khan (1994)

6. Genetic analysis for major BPH resistance genes by classical genetic approach

Inheritance of rice resistance to BPH has been investigated since 1968 (Khush, 1979). Four resistant cultivars, Mudgo, ASD7, CO22 and MTU15, were initially analyzed. F_2 populations from the crosses of susceptible TN1 with the resistant cultivars, Mudgo, MTU15 and CO22, segregated into a ratio 3 resistant: 1 susceptible, indicating that three varieties have a dominant gene for resistance to BPH. The F_2 population from a cross of TN1×ASD7 segregated into 1 resistant: 3 susceptible, indicating that ASD7 has a recessive gene for the resistance (Athwal *et al*., 1971). The single dominant gene in Mudgo, MTU15 and CO22 was at the same locus. This locus was designated as *Bph1*. The resistance in ASD7 was controlled by a single recessive gene, designated as *bph2* (Khush, 1979). No recombination between *Bph1* and *bph2* was observed in those studies. It was indicated that these two genes are closely linked (Athwal *et al*., 1971).

Later studies, Lakshminarayana and Khush (1977) analyzed 28 resistant cultivars. Nine of the cultivars had *Bph1* and sixteen had *bph2* for resistance. Two new loci for resistance were discovered. A single dominant gene governs resistance in Rathu Heenati, a Sri Lanka rice cultivar, segregated independently of *Bph1*. The resistance gene was designated as *Bph3*. A single recessive gene in Babawee segregated independently of *bph2* and it was designated as *bph4*. Resistance in PTB21 and PTB33 is controlled by one dominant gene, *Bph3*, and one recessive gene, *bph2* (Angeles *et al*., 1986; Ikeda, 1985; Ikeda and Kaneda, 1981).

A new resistance gene for resistance to BPH biotype 4, which *Bph1* and *bph2* can not resistance, was evaluated at the Bangladesh Rice Research Institute. This gene was designated as *bph5* (Khush *et al*., 1985). Seventeen resistant cultivars, which can resistance to biotype 4 but susceptible to biotype 1, 2 and 3, were genetically analyzed using the BPH biotypes. Seven were found to have single dominant gene, which segregated independently of *bph5*. The single dominant gene was designated as *Bph6* (Kabir and Khush, 1988). The remaining ten cultivars were found to have recessive gene for resistance and eight of them were allelic to *bph5* but the recessive gene of

two cultivars were nonallelic to *bph5*. Therefore, the recessive gene was designated as *bph7* (Kabir and Khush, 1988).

Nemoto *et al* (1989) studied on two Thai cultivars, Col.5 Thailand and Col.11 Thailand, and one cultivar from Myanmar, Chin Saba and found that the cultivars carried a single recessive gene, which was allelic to each other but was not allelic to *bph2* and *bph4*. The recessive gene of these three cultivars was also nonallelic to *bph5* and *bph7,* which did not confer resistance to biotype 1, 2, and 3, but the new gene did. Therefore, this new recessive gene was different from all the other recessive genes and was designated as *bph8*. In 1988, other new resistance gene, *Bph9*, has been found in Kaharamana, Pokkali, and Balamawee (Nemoto *et al*., 1989). Recently, a new recessive gene from wild rice, *O. rufipogon*, designated as *bph14* was also identified by classical genetic approach (Li *et al*., 2002).

7. Identification and molecular mapping of major BPH resistance genes

To date, the number of major genes and several quantitative trait loci (QTL) conferring BPH resistance in several cultivated and wild species (Table 1, Figure 9) has been reported and reviewed (Alam and Cohen, 1998; Chen *et al*. 2006; Huang *et al*. 1997, 2001; Ishii *et al*. 1994; Jairin *et al*., 2005b; Jena *et al*., 2003; 2006; Jeon *et al*., 1999; Kawaguchi *et al*., 2001; Liu *et al*., 2001; Mei *et al*., 1996; Murai *et al*., 2001; Murata *et al*., 1998, 2001; Rahman *et al*., 2007; Renganayaki *et al*., 2002; Sharma *et al*. 2003a; 2003b; Soundararajan *et al*., 2004; Su *et al*., 2002; Sun *et al*. 2005; 2006; Wang *et al*., 2001; Xu *et al*., 2002; Yan *et al*. 2002; Yang *et al*., 2002; 2004). There are ten resistance genes assigned *Bph1* to *Bph9* and *bph12* were identified by classical genetic approach, of which *Bph1*, *bph2*, *Bph3*, *bph4*, *Bph6*, and *Bph9* were further identified by molecular genetic approach. Ikada and Kaneda (1981) reported the location of *Bph3* and *bph4* on rice chromosome 10 through trisomic analysis. In the same way, they also located *Bph1* and *bph2* on chromosome 4 (Ikada and Kaneda, 1983). However, the result from various previous researches' studies indicated that *Bph1* and *bph2* were located on chromosome 12 using DNA markers

analysis (Hirabayashi and Ogawa, 1995; Huang *et al*., 1997; Jeon *et al*., 1999; Murata *et al*., 1998).

RFLP markers were initially used to analyze the BPH resistance gene using a doubled haploid population derived from a cross between IR64 and Azucena (Huang *et al*., 1997). In this study the resistance gene, *Bph1*, was located on chromosome 12. Jeon *et al* (1999) reported the tagging of *Bph1* in rice cultivar Gayabyeo using Random-amplified Polymorphic DNA (RAPD) and RFLP markers. The result showed that RAPD marker RRD7 was co-segregated with *Bph1* locus on the chromosome 12 and linked with RG457, which linked with the resistance gene *Bph10* (Ishii *et al*., 1994). Hirabayashi and Ogawa (1995) were also found that *Bph1* in IR28 was located on chromosome 12 near a RFLP marker C185. The resistance gene *bph2* was reported to be recessive and closely linked to *Bph1*. Murata *et al*. (1998) reported that *bph2* was mapped at 3.5 cM from the closest RFLP marker G2140 and was considerable distance about 30 cM from *Bph1*. Recently, the resistance gene was finely mapped to locate near the isolated *OsBphi252* gene on chromosome 12 through the representational difference analysis (Park *et al*., 2008).

Yet, until recently, the location of *Bph3* on rice chromosome has not been clarified. The *Bph3* locus was reported to map on rice chromosome 4 and 10 (Ikeda and Kaneda, 1981; Kawaguchi *et al*., 2001; Sun *et al*., 2005). The resistant gene *bph4,* which was closely linked to *Bph3* (Ikeda and Kaneda, 1981; Sidhu and Khush, 1979), was mapped on chromosome 6 (Kawaguchi *et al*., 2001). Rathu Heenati was the first cultivar that has been reported to carry *Bph3*. The major resistance gene in Rathu Heenati was mapped on short arm of chromosome 4 (Sun *et al*., 2005). According to those studies, the location of *Bph3* can possibly locate on chromosome 4, 6 or 10. Thus, the location of *Bph3* on rice chromosome should be further investigated and confirmed. Murata *et al* (2001) identified a dominant gene *Bph9* using RFLP and RAPD analysis on the long arm of chromosome 12 flanked by markers G2140 and S2545. Later, Su *et al* (2006) confirmed the location of *Bph9* in the same region on chromosome 12 using SSR markers. Recently, two new QTL introgressed from a resistant *indica* cultivar, Col.5 Thailand, were detected on rice chromosome 2 and 6

with 29.4 and 46.2% of phenotypic variation explained, respectively (Sun *et al*., 2007). Two recessive resistance gene, *bph5* and *bph7* (Khush *et al*., 1985; Kabir and Khush, 1988; Nemoto *et al*., 1989) have not yet been identified the locations on the rice chromosome.

The wild species of rice have been considered to be the most important resources for BPH resistance. To date, about ten BPH resistance genes have been identified in the various wild species. Ishii *et al* (1994) first identified a resistance gene, *Bph10*, in an introgression line IR65482-4-136-2-2, which was derived from the wild species *Oryza australiensis*, on the long arm of chromosome 12 through RFLP analysis. Later, Jena *et al* (2006) mapped the resistance gene *Bph18* on the subterminal region of the long arm of chromosome 12 in an *O. australiensis*-derived line, IR65482-7-216-1-2, through SSR and sequence-tagged site (STS) analysis. The BPH resistance gene in introgression line IR54741-3-21-22, which was derived its resistance from *O. officinalis* was also mapped onto rice chromosome 11 (Jena *et al*., 2001). Yang *et al* (2002) identified the resistance gene *Bph12* on the short arm of chromosome 4 in cultivar B14, which was derived from *O. latifolia*, through SSR and RFLP. Liu *et al* (2001) identified a major dominant gene, *Bph13*(t) on the long arm of chromosome 2 in an *O. eichingeri*-derived line, ACC105159, through RFLP and SSR analyses. Renganayaki *et al* (2002) identified a resistance gene, which was designated as *Bph13*, in IR54745-2-21-12-17-6, a line with *O. officinalis*-derived resistance to BPH biotype 4, on the short arm of chromosome 3, through RAPD analysis. Huang *et al* (2001) reported that B5, a highly resistance line that derived its resistance genes from *O. officinalis*, carries two major resistance genes, *Qbp1* and *Qbp2* (later designated as *Bph14* and *Bph15*, respectively) on the long arm of chromosome 3 and the short arm of chromosome 4, respectively, through the RFLP analysis. Ren *et al* (2004) reported two minor QTL (*Qbp3* and *Qbp4*) on chromosome 2 and 9 derived from the same genetic source, B5. Hirabayashi *et al* (1998) also found two recessive genes, *bph11* and *bph12* on the long arm of chromosome 3 and the middle arm of chromosome 4, respectively, in introgression lines of *O. officinalis*, through RFLP analysis. Recently, two new dominant BPH resistance genes, *Bph20* and *Bph21*, have

been finely mapped on the rice chromosome 4 and 12, respectively. The resistance genes were introgressed into japonica cultivar 'Junambyeo' from *O. minuta*-derived resistance line IR71033-121-15 (Rahman *et al*., 2007). In addition, a BPH resistance gene from the same *O. minuta*-derived resistance line was mapped on the short arm of chromosome 6 (Jairin *et al*., 2007b).

The location of major BPH resistance genes identified from the previous studies indicated clustering of BPH resistance genes in four main regions on chromosome 3, 4, 6 and 12 (Jairin *et al*., 2007b) (Figure 10). The presence of five BPH resistance genes, *Bph1*, *bph2*, *Bph9*, *Bph10* and *Bph18* were first found to be clustered on the long arm of chromosome 12 (Chen *et al*., 2005; Jena *et al*., 2006). The cluster of BPH resistance genes on chromosome 3 has been described by Chen *et al* (2005). They found that two major resistance genes and two QTL, which derived from different four sources of BPH resistant donors, were mapped in the same region on chromosome 3 (Chen *et al*., 2005). Three resistance genes designated as *Bph12*, *Bph15* and *Bph17* from *O. officinalis*, *O. latifolia* and Rathu Heenati, respectively were mapped in the particular region on chromosome 4 (Sun *et al*., 2005; Yang *et al*., 2005; Yang *et al*., 2004). A recessive gene *bph4* from Babawee and *O. rufipogon* and two QTL from IR64 and Teqing were likewise reported in the same genomic location on short arm of chromosome 6 (Alam and Cohen, 1998; Kawaguchi *et al*., 2001; Soundararajan *et al*., 2004; Xu *et al*., 2002; Li *et al*., 2000). Although the resistance genes were located in the same region, no evidence has yet been obtained that they might share the same genomic sequence or they are different loci but tightly linked to each other or they are different alleles at the same locus. The answers of these questions could be provided in the near future when the BPH resistance genes have been cloned. Cloning of the genes would eventually lead to the elucidation of the difference and the evolution of the BPH resistance genes in rice.

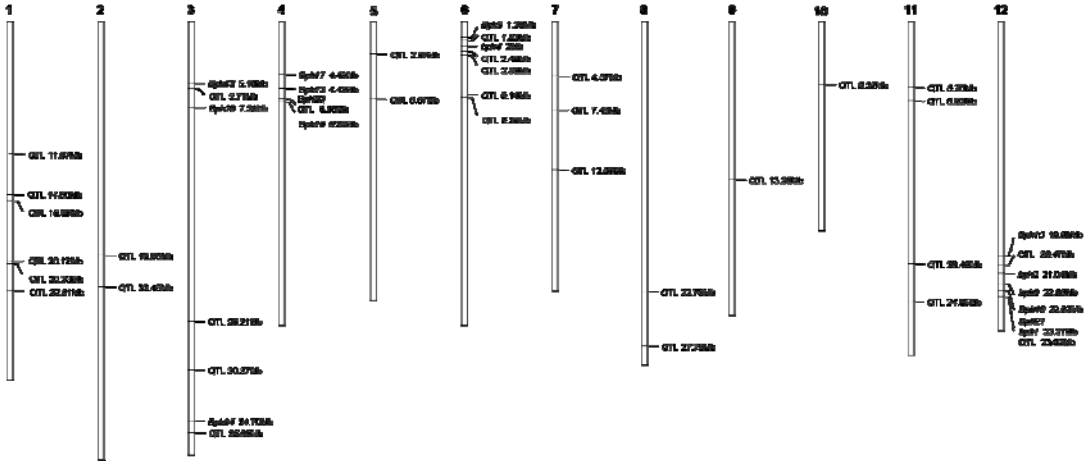

Figure 9 Location of major BPH resistance genes and QTL associated with BPH resistance throughout 12 rice chromosomes.

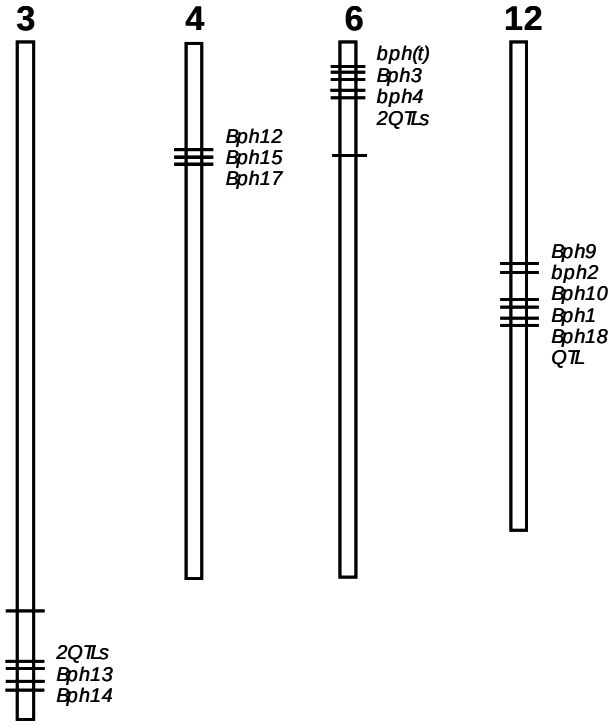

Figure 10 Locations of the BPH resistance genes and QTL clustered on rice chromosome 3, 4, 6, and 12 based on the standard genetic map of SSR markers of McCouch *et al*. (2002)

Source: Jairin *et al*. (2007b)

Table 1 A list of BPH resistance genes reported in the literatures.

BPH resistance gene	Year of identification	Source	Chr.	Reference of mapping
Bph1	1971	Mudgo	12	Hirabayashi and Ogawa, 1995; Jeon *et al*., 1999; Sharma *et al*., 2003b
bph2	1971	ASD7	12	Murata *et al*., 1998; Murai *et al*., 2001
Bph3	1977	Rathu Heenati	6	Jairin *et al*., 2007a
bph4	1977	Babawee	6	Kawaguchi *et al*., 2001
bph5	1985	ARC10550		
Bph6	1988	Swarnalata	11	Jena *et al*., 2001
bph7	1988	T12		
bph8	1989 2007	Chin Saba, Col.5 Thailand	2, 6	Sun *et al*., 2007
Bph9	1985	Pokkali, Kaharamana	12	Murata *et al*., 2001; Su *et al*., 2006
Bph10	1994	*O. australiensis*	12	Ishii *et al*., 1994
Bph11	1998	*O. officinalis*	3	Hirabayashi *et al*., 1998
Bph12	2002	*O. latifolia,* B14	4	Yang *et al*., 2002
Bph12	1998	*O. officinalis*	4	Hirabayashi *et al*., 1998
Bph13	2001	*O. eichingeri*	2	Liu *et al*., 2001
Bph14	2001	*O. officinalis*	3	Huang *et al*., 2001; Wang *et al*., 2001; Renganayaki *et al*., 2002
Bph14	2002	*O. rufipogon*		Li *et al*., 2002
Bph14	2001	B5	3	Huang *et al*., 2001
Bph15	2001	*O. officinalis,* B5	4	Huang *et al*., 2001; Wang *et al*., 2001
Bph17	2005	Rathu Heenati	4	Sun *et al*., 2005
Bph18	2006	*O. australiensis*	12	Jena *et al*., 2006
Bph19	2005	AS20-1	3	Chen *et al*., 2005
Bph20	2007	IR71033-121-15	4	Rahman *et al*., 2007
Bph21	2007	IR71033-121-15	12	Rahman *et al*., 2007
Bph(t)	2007	IR71033-121-15	6	Jairin *et al*., 2007b
Bph20	2008	ADR52	6	Marlar *et al*. unpublished
Bph21	2008	ADR52	12	Marlar *et al*. unpublished
Bph(t)	2003	*O. officinalis*	11	Jena *et al*., 2003
Bph(t)	1996	Sanguizhan	9	Mei *et al*., 1996
QTL	1998	IR64	1,2,3,4, 6,8	Alam and Cohen, 1998;
QTL	2004	IR64	1,2,6,7	Soundararajan *et al*., 2004
QTL	2000	Asominori	1,6	Yamasaki *et al*., 2000
QTL	2002	Teqing	1,3,5,8, 11	Xu *et al*., 2002
QTL	2004	*O. officinalis*	2,3,4,9	Ren *et al*., 2004
QTL	2005	Abhaya	6,10,12	Jairin *et al*., 2005b

8. Progress in cloning of BPH resistance genes

Much progress on rice plant responses to brown planthopper has been made during the last few years (Hao *et al*., 2008; Ren *et al*., 2004; Wang *et al*., 2008; Yang *et al*., 2004; Yang *et al*., 2005; Yuan *et al*., 2005). The molecular cloning of the BPH resistance genes will provide valuable information on the function of the protein products, and on the mechanism of BPH resistance in rice. The information will help breeders to maintain and manage a utilization of BPH resistance genes from a limitation of sources for BPH resistance, which will be lost their ability against new BPH biotypes. BPH resistance genes, *Bph14* and *Bph15*, on chromosome 3 and 4 in an introgression line derived from *O. officinalis* have been identified by scientists from China (Hao *et al*., 2008; Ren *et al*., 2004; Yang *et al*., 2004; Yuan *et al*., 2005). Various molecular techniques have been used to study rice responses to BPH feeding. Recently, they have reported the induced callose sealing in sieve elements of rice plants carrying *Bph14* and *Bph15* plays an important role in the inhibition of BPH (Hao *et al*., 2008). Until recently more than 20 publications have been released and BPH resistance candidate genes have been identified. In Japan, some groups of scientists tried to identify the BPH resistance gene expression by induced in responses of rice plant to BPH feeding. They targeted on *Bph1* and *bph2* genes and using technique of cDNA and ESTs to identify the candidate genes. Additionally, Park *et al* (2008) developed a modified representational difference analysis method to detect rare transcripts among those differentially expressed in a BPH resistant near-isogenic line carrying the *Bph1*. They found that an *OsBphi252* gene, which encodes a putative lipoxygenase (LOX), co-segregates with *Bph1* and may play an essential role during the BPH resistance response. Consequently, at least four resistant genes, *Bph1*, *bph2*, *Bph14* and *Bph15*, have been extensively studied to clone the resistance genes. According to the progress of all studies, BPH resistance genes will be cloned in the near future.

9. Marker-assisted backcross breeding

One of the major applications of molecular markers to rice breeding is using in maker-assisted selection (MAS) or marker-assisted backcrossing (MAB) programs. Backcross breeding is a procedure for the introgression of a target gene from a donor into the genetic background of a recipient. In backcross breeding, DNA markers can be used to control the target gene and the genetic background (Hospital, 2001). By selecting the desirable allele at markers that are closely linked to a target gene in the backcross scheme, we can use DNA markers in manipulating such traits more efficiently. In this way, linkage drag can be reduced by performing background selection at two closely linked markers flanked to the target genes (Hospital, 2001). The undesirable linkage drag in a target gene region can be removed by intensive work to select recombinants and a molecular marker tightly linked to the target gene could be useful for selecting the desired recombinants. The size of donor chromosome segments around introgressed loci and the reduction of linkage drag in MAB program have been investigated (Hospital, 2001). Several researchers have reported the efficiency of MAS for the successful transfer of genes for disease and insect resistance in rice breeding programs (Joseph *et al*., 2004; Sanchez *et al*., 2000; Sharma *et al*., 2004; Toojinda *et al*., 2005; Zhang, 2007).

In case of BPH, since a total of 21 BPH resistance genes have been identified from cultivated and wild rice species, molecular mapping of these genes has facilitated MAS of the BPH resistance genes. MAS has been successfully used in selecting for BPH resistance and pyramiding multiple genes for durable resistance against BPH (Jena *et al*., 2006; Sharma *et al*., 2004; Sun *et al*., 2006; Toojinda *et al*., 2005). Sharma *et al*. (2004) performed a molecular marker-assisted pyramiding of two BPH resistance genes, *Bph1* and *Bph2*, into a *japonica* line. Combining of more than two independent BPH resistance genes into a particular line can offer a possible means to cope with the occurrence of such virulent BPH biotypes.

MATERIALS AND METHODS

Materials

1. Plant materials

A differential set of ten BPH resistance cultivars with known resistance genes for BPH namely Mudgo [acc. no. 6663 (*Bph1*); Athwal *et al*. 1971], ASD7 [acc. no. 6303 (*bph2*); Athwal *et al*. 1971], Rathu Heenati [acc. no. 11730 (*Bph3*); Lakshminarayana and Khush 1977], PTB33 [acc. no. 19325 (*bph2* and *Bph3*); Lakshminarayana and Khush 1977], Babawee [acc. no. 8978 (*bph4*); Lakshminarayana and Khush 1977], ARC10550 [acc. no. 12507 (*bph5*); Khush *et al*. 1985], Swarnalata [acc. no. 33964 (*Bph6*); Kabir and Khush 1988], T12 [acc. no. 56989 (*bph7*); Kabir and Khush 1988], Chin Saba [acc. no. 33016 (*bph8*); Nemoto *et al*. 1989] and Pokkali [*Bph9*; Nemoto *et al*. 1989] were used to identify the variation of virulence among the BPH populations collected from rice fields in Thailand. All BPH resistance cultivars used in this study were obtained from the International Rice Research Institute (IRRI), Philippines.

2. BPH populations

Forty-five BPH populations were collected from rice fields in 31 provinces of the northeastern, northern, central and southern regions of Thailand (Appendix Table 1). The insect populations were reared on rice cultivar TN1 in a temperature-controlled room maintained at a light regime of 15/9-h light/dark and day/night temperatures of 26–28ºC. The BPH colonies were employed for BPH bioassays after four to six generations of the insects.

Methods

1. Development of mapping populations and introgression lines

In this study, rice cultivars Rathu Heenati, KDML105, PTB33 and RD6 were used as the parents for mapping populations. Rathu Heenati, a local cultivated rice from Sri Lanka, and PTB33, a local cultivar from India, were used as the donors. Both cultivars carry BPH resistance gene *Bph3*. These cultivars are resistance to BPH populations in Thailand but have poor grain quality i.e. high amylose content, chalky endosperm and no fragrance. KDML105 and RD6, Thai good grain quality cultivars, were used as recipient parents. The background information of the parents is shown in Table 2. The cross combinations that used in this study are described in Table 3 and Figure 11. To map the *Bph3* locus, 208 progenies of a BC_1F_2 population obtained from a cross between the donor parent PTB33 and the recurrent parent RD6 and 333 progenies of a BC_3F_2 population obtained from a cross between Rathu Heenati, the donor parent, and the recurrent parent KDML105 were generated.

The introgression lines were developed from the cross of Rathu Heenati×KDML105. The BC_1 generation was resulted from the backcrossing of the F_1 plants with the recurrent parent. The second and third rounds of backcrossing (BC_2 and BC_3) were derived from the cross of selected resistant BC_1 and BC_2 plants based on BPH resistant phenotype and linked markers. A total of 2,343 progenies of BC_3F_2 were obtained and used to dissect a linkage drag between *Bph3* and *Wx*-RH loci. Two BC_3F_3 individual plants were developed from the BC_3F_2 resistant plant, which showed heterozygous in the target region on chromosome 6. A total of 330 BC_3F_4 individuals derived from an individual BC_3F_3 plant that was heterozygous at the *Bph3* locus and homozygous at the *Wx*-KD locus (Wx^b) were used to confirm the location of *Bph3* on chromosome 6 and validate the BPH resistance. Fifty selected $BC_3F_{4\text{-}6}$ ILs derived from the individual plants that were homozygous for *Bph3* and Wx^b were used to determine the recurrent genetic background and evaluate the agronomic trait performance and their grain quality traits.

Table 2 Background information of parental cultivars.

Donor parent	Agronomic trait[1]				Grain quality[2]					
	FW	PH	PA	LG	AC	CK	GT	GL	GS	SC
Rathu Heenati	94	145	5	5	23.2	9	I/L	5	5	0
PTB33	93	142	5	6	24.6	9	I	5	5	0
KDML105	PS*	139	3	6	16	1	L	1	1	2
RD6	PS	140	3	5	GN**	GN	L	1	1	2

[1]FW-Days to 50% flowering; PH-Plant height (cm); PA-Phenotypic acceptability (1-excellent; 3-good; 5-fair; 7-poor; 9-unacceptable); LG-Lodging incidence (1-1-10% of plants; 3-11-20% of plants; 5-21-35% of plants; 7-36-50% of plants; 9-50-100% of plants)

[2]AC-Amylose content (%); CK-Chalkiness of endosperm (0-none; 1-less than 10%; 5-11-20%; 9-more than 20%); GT-Gelatinization temperature (H-high; H/L-high or intermediate; I-intermidiate; L-low); GL-Grain length (1-extra long; 3-long; 5-medium; 7-short); GS-Grain shape (1-slender; 3-medium; 5-bold; 9-round); SC-Scent (0-unscent; 1-lightly scented; 2-scented)

*PS=photosensitive, **GN=glutinous grain

Table 3 Populations used for mapping, MAS validity test, field performance evaluation and measuring grain quality.

Cross combinations	Generation	Application	Population size (no.)
PTB33×RD6	BC_1F_2	Mapping population	208
Rathu Heenati×KDML105	BC_3F_2	Mapping population,	333
		Braking between *Bph3* and *Waxy* locus	2,343
		Recurrent parent background analysis	50
	BC_3F_3	Finding most putative locus	28
		MAS validity test	330
	BC_3F_{4-6}	Confirmation of BPH resistance	108
		Field evaluation	50
		Grain quality traits evaluation	50

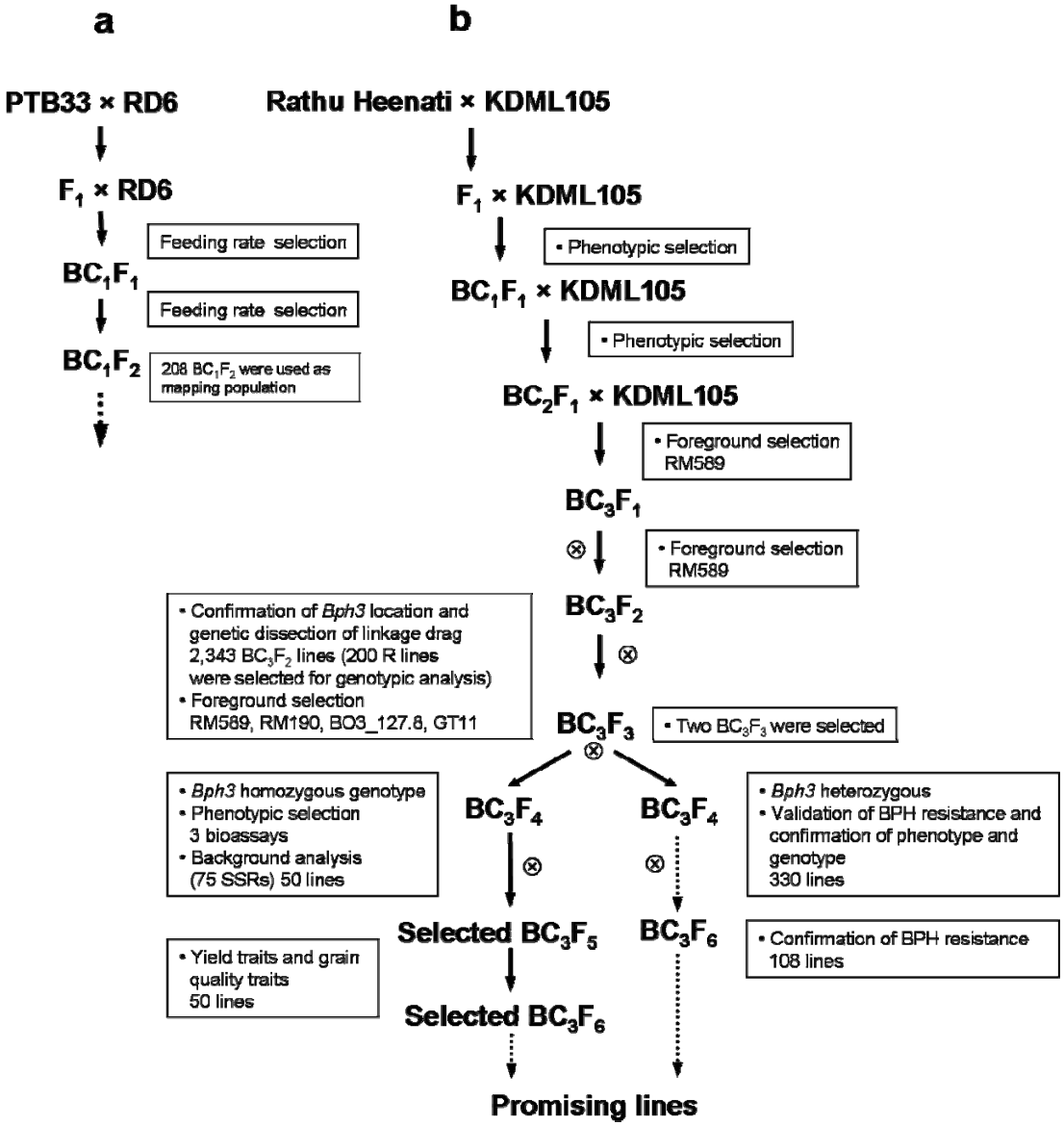

Figure 11 Scheme for the development of mapping populations, a cross of PTB33×RD6 (a) and Rathu Heenati×KDML105 (b), and BPH resistance introgression lines with details of markers used for foreground, recombinant, and background selection.

2. Bioassays for BPH resistance

Five insect bioassays namely standard seedbox screening, modified mass tiller screening, semi-field screening, antibiosis on feeding rate and antixenosis on feeding preference, were used to evaluate BPH resistance of the parents and progenies.

2.1 Standard seedbox screening (SSBS)

The SSBS was used to evaluate the BPH resistance at the seedling stage of test entries under greenhouse condition according to Heinrichs *et al*. (1985). The pre-germinated seeds of test lines were sown 5 cm apart in 20 cm rows in seedboxes. The susceptible control, TN1, was sown randomly in all the seedboxes. Seven days after sowing, the seedlings were infested with second to third nymphs of BPH at an amount approximately twenty nymphs per seedling. Damage rating on a scale of 1 (very slight damage) to 9 (all plants dead) of the test lines was done when 90% of the plants in the susceptible control row were killed according to the standard evaluation system (SES) (International Rice Research Institute, 1996).

The SSBS was also used to evaluate the BPH resistance of seedlings with and without nitrogen application. The pre-germinated seeds of test lines were sown 5 cm apart in 20 cm rows in seedboxes. The susceptible control, TN1 was sown randomly in all the seedboxes. Seven days after sowing, the seedlings were infested with second to third nymphs of BPH at an amount of twenty nymphs per seedling. Nitrogen fertilizer, Ammonium nitrate, was applied to only the seedboxs with nitrogen application 24 h before infestations.

Standard Evaluation System (SES)

Scale for standard seedbox screening test

Resistance score	**Plant status**	**Resistance level**
1	No damage or very slight damage	HR
3	First and 2nd leaves of most plants partially yellowing	R
5	Pronounced yellowing and stunting or about 10 to 25% of the plants wilting or dead and remaining plants severely stunted or dying	MR
7	More than half of the plants wilting or dead	MS
9	All plants dead	S

HR = highly resistant, R = Resistant, MR = moderately resistant,
MS = moderately susceptible, S = susceptible

2.2 Modified mass tiller screening (MMTS)

The modified mass tiller screening (MMTS) technique, which was modified from the modified seedbox screening technique (Velusamy *et al*. 1986) and the tiller seedbox screening technique (Wang *et al*. 2001), was used to evaluate the BPH resistance of the test lines under greenhouse condition. First, the seeds of the controls and test entries were separately sown in the seedling plots. The seedlings were transplanted into 7×24-m^2 plots when the seedlings had three to four tillers (approximately 20–25 days). Ten days after transplanting the seedlings were infested with third to fourth instar nymphs of the BPH at a density of ten insects per tiller. Then, we let the insects feed, mate, lay eggs and hatch freely. Until the susceptible control lines died, we evaluated the severity scores of each line individuals on a scale of 1 (very slight damage) to 9 (all plants dead) according to the SES for rice with minor modification. The remains of resistant plants were scored every ten days until the flowering stage. The MMTS was also used to evaluate the BPH resistance at the tillering stage of rice plants with and without nitrogen applications.

Standard Evaluation System (SES)

Scale for modified mass tiller screening test

Resistance score	**Plant status**	**Resistance level**
1	No damage or lower leaves slight yellowing of a few plants	HR
3	Lower leaves of most plants yellowing or wilting	R
5	Upper leaves with pronounced yellowing and some stunting or wilting	MR
7	Half of the plants wilting or with hopperburn	MS
9	All plants dead	S

HR = highly resistant, R = Resistant, MR = moderately resistant,
MS = moderately susceptible, S = susceptible

2.3 Semi-field screening (SMFS)

The SMFS was used to evaluate rice plants at the vegetative and reproductive stages in the rice field. Ten of twenty-day seedlings were transplanted (20×20 cm) in the rice field, which covered with a nylon-net. Fifteen days after transplanting, the rice plants were infested with 3^{rd}-4^{th} nymph of BPH at an amount of five insects per hill. Then, we let the insect population increasing for 1-2 generations. When all the TN1 had died, we scored the degree of damage undergone by the test seedlings. The scoring criteria were based on the SES scale for MMTS. The remains of resistant plants were scored every ten days until flowering stage.

2.4 Antibiosis on feeding rate (AFR)

Antibiosis on feeding rate bioassay was conducted to measure honeydew excretion of the BPH in a temperature-controlled room as described by Jairin *et al*. (2005a). Briefly, only one tiller from each hill of 30 days rice plant was selected for a bromocresol green-treated filter paper in a plastic cup. Plants were infested with 5

one-day-old brachypterous females of BPH which were starved for one hour prior to infestation. The feeding rate was recorded after 24 h at the 26ºC temperature. After 24 h, the filter papers were collected and the total area of blue-green spots, resulting from honeydew deposition, was measured.

2.5 Antixenosis on feeding preference (AFP)

Feeding preference or antixenosis for BPH settling on test plants was assessed by monitoring the numbers of BPH nymphs alighting on the test plants. The study was modified from the method of Heinrichs *et al*. (1985). Seeds of test varieties were sown in the pots (8 cm in diameter and 15 cm in height). At 35 days after sowing, the plants were trimmed to one tiller and rice plots were arranged in a circle of 60 cm diameter with water to a level half that of the pots. The third instar nymphs of BPH were put in approximately 30 insects per seedling. Experiments were conducted by releasing the insects at the center equidistant from the center of the plots. The number of the BPH on each variety was recorded at 24, 48, and 72 h after infestation.

3. Variation of BPH populations

The rice resistant cultivars with specific resistance genes were screened to identify the variation of virulence among BPH populations using the SSBS and the AFR methods. To determine the variation among BPH populations, the resistance or susceptibility of the differential set of ten BPH resistance cultivars was converted into binary data. Similarity matrices were calculated with Dice's coefficient and the SIMQUAL program of NTSYS-PC. Cluster analysis was carried out within the SAHN program using the UPGMA method.

4. DNA extraction and PCR amplification

Young rice leaf samples were cut and stored frozen at -80°C prior to DNA extraction. Total genomic DNA was extracted from the leaves using a rapid CTAB

DNA isolation technique following the protocol described by Chen and Ronald (1999) with minor modifications. PCR was performed in a 10 μl reaction mixture containing 10-25 ng of template DNA, 0.5 μM of each primer, 250 μM of each dNTP, 1.5 mM $MgCl_2$, 1 unit Taq polymerase and 2 μl of ×10 PCR reaction buffer. Amplification was performed for 35 cycles (1 min at 94°C, 1 min at 55°C and 2 min at 72°C) followed by 5 min at 72°C. The amplified product was electrophoresed on a 4.5% denaturing silver-stained polyacrylamide gel.

5. Determining the map location of the BPH resistance gene

Based on the results of the BPH bioassays from the MMTS and the bulked segregant analysis concept (Michelmore *et al*., 1991), we generated two groups of 15 resistant (R) and 15 susceptible (S) progenies from each of the backcross populations, BC_1F_2 and BC_3F_2, derived from the crosses of PTB33×RD6 and Rathu Heenati×KDML105, respectively. Thirty-six polymorphic SSR markers, including 13 markers covering a genetic distance of 5.4–151.1 cM on chromosome 4, 14 markers covering a genetic distance of 2.3–105.1 cM on chromosome 6 and nine markers covering a genetic distance 17.6–113.0 cM on chromosome 10 (McCouch *et al*. 1997, 2002), were selected to identify the individual progenies in the R and S groups. Seven previously reported SSR markers (RM8213, RM261, RM6487, RM401, RM190, RM469 and RM204) closely linked to *Bph3* and *bph4* (Ikeda and Kaneda 1981; Kawaguchi *et al*. 2001; Sun *et al*. 2005; Yan *et al*. 2002) were the target loci of the BPH resistance gene. A STS marker, KAM4, which completely co-segregated with *bph2* (Murai *et al*. 2001), was used to determine the *bph2* locus in the PTB33 and the R/S individuals from the BC_1F_2 progenies of the PTB33×RD6 cross.

A linkage analysis was performed using the 208 BC_1F_2 and 333 BC_3F_2 individuals and fourteen polymorphic SSR markers (Table 4). Recombination values were calculated by JOINMAP ver. 4.0 (Van Ooijen and Voorrips, 2001) with LOD scores greater than 3.0. Map distances were calculated using the Kosambi function (Kosambi, 1944). The genetic contribution to the phenotypic resistance by the

chromosome region was analyzed using MAPQTL ver. 5 at LOD threshold 3.0 (van Ooijen, 2004).

Table 4 A list of SSR and STS markers used to construct linkage maps and identify the BPH resistance gene.

Markers	**Type**	**Primer sequence**		**Size**
		Forward primer	**Reverse primer**	**(bp)**
RM3353	SSR	aatggtcgcctctctctctg	gctggcattgaccgtgtc	116
RM469	SSR	agctgaacaagccctgaaag	gacttgggcagtgtgacatg	180
RM589	SSR	atcatggtcggtggcttaac	caggttccaaccagacactg	148
RM588	SSR	gttgctctgcctcactcttg	aacgagccaacgaagcag	98
RM587	SSR	acgcgaacaaattaacagcc	ctttgctaccagtagatccagc	273
RM586	SSR	acctcgcgttattaggtaccc	gagatacgccaacgagatacc	295
RM190	SSR	gctacaaatagccacccacacc	caacacaagcagagaagtgaagc	144
RM8101	SSR	cactgacatagctaaggtctcatgtcttat	tggttaactcgctattataatgagttcg	183
RM204	SSR	gtgactgacttggtcataggg	gctagccatgctctcgtacc	174
RM8213	SSR	tgttgggtgggtaaagtagatgc	cccagtgatacaaagatgagttgg	179
RM261	SSR	gcatggccgatggtaaag	tgtataaaaccacacggcca	146
RM6487	SSR	ccgtggagaagaagctgtagacg	cttcccaacctcaacctctcg	124
RM401	SSR	gcatgagctgctctcattattgtcc	gaaacgaaccaaacgttcatcg	241
KAM4	STS	taactggtgttagtgcgaatg	aattcacggcatgtgaagccctag	300

6. Physical genetic mapping of the *Bph3* locus

Initial localization of the *Bph3* locus was based on the recent report of mapping on the short arm of rice chromosome 6. The linkage analysis was performed using 333 BC_3F_2 individuals from the cross between Rathu Heenati and KDML105. The resistance gene was located between the flanking markers RM589 and RM588. In this study, 14 additional SSR markers and two single-nucleotide polymorphisms (SNPs) markers (Table 5) covering the BPH resistance gene region were used to screen Rathu Heenati and KDML105. The SSR markers were obtained from the public database released by Gramene (http://www.gramene.org). SNP markers were designed according to the rice genome database of Nipponbare and 93-11. The polymorphic markers between Rathu Heenati and KDML105 were used to assay 28 BC_3F_3 plants for the fine genetic and physical mapping of *Bph3*. The physical location of the *Bph3* locus in the *japonica* cultivar Nipponbare was determined. A physical map spanning the resistance gene locus was constructed *in silico*, based on the contig map. The prediction of candidate resistance genes with the conserved structures in the target region anchored by tightly linked markers was then analyzed according to the sequences of Nipponbare and was based on the TIGR prediction method (http://www.tigr.org).

7. Target genes and DNA markers for MAS

All DNA markers that used to select the target loci are shown in Table 6. Two markers, RM589 and RM587, cosegregated with the *Bph3* locus, were used to select for the presence of the *Bph3* gene. One SSR marker, RM190, representing the *Waxy* locus was used to select for the presence of *waxy* allele of KDML105 (*wx*-KD) in the process of backcrossing. The SSR marker, BO3_127.8, cosegregated with rice grain aroma (Wanchana *et al*, 2005), were used to select for the fragrance allele of KDML105. The STS marker GT11 was used to identify the allele corresponding to low gelatinization temperature.

8. Genetic dissection of the target *Bph3* allele and unfavorable *Wx*-RH (Wx^a) allele

A total of 2,343 BC_3F_2 progenies from the cross of Rathu Heenati×KDML105 were used to dissect a linkage between *Bph3* and *Waxy* loci. The SSR markers closely linked to *Bph3* and *Wx*-RH loci were used to analyze BC_3F_3 progenies derived from the selected resistance BC_3F_2 lines. To dissect a linkage between *Bph3* and *Wx*-RH loci, the BC_3F_2 progenies were screened in the greenhouse for BPH resistance using MMTS. A variation in BPH resistance was observed in BC_3F_2 plants, and we classified BC_3F_2 plants into three segregation patterns: resistance, moderately resistance and susceptibility. A total of 200 resistant and moderately resistant plants were selected for genetic analysis. The progenies that show heterozygous on the *Bph3* and *Waxy* regions were selected. Only progenies that carry Rathu Heenati homozygous/heterozygous genotype at *Bph3* region and carry KDML105 homozygous genotype at *Waxy* region were selected to generate BC_3F_3. Two SSR markers, RM589 and RM190, closely linked to *Bph3* and *Waxy* loci, respectively were used to analyze BC_3F_3 progenies derived from the selected resistance BC_3F_2 lines.

9. Determination of genetic background

Based on the high-resolution rice linkage map with SSR marker (McCouch *et al*., 2002), about 120 SSR primer sets were selected and tested on the parental cultivars Rathu Heenati and KDML105. A total of 75 polymorphic SSR markers distributed throughout rice genome (approximately 5-7 markers spanning each chromosome) were then used to determine the recurrent genetic background of the 50 selected ILs from the BC_3F_4 generation.

Table 5 Microsatellite and Indel markers used to construct fine mapping of the *Bph3* locus.

Markers	Type	Primer sequence		Size (bp)
		Forward primer	Reverse primer	
RM19291	SSR	cacttgcacgtgtcctctgtacg	gtgtttcagttcaccttgcatcg	146
RM19295	SSR	gtcatggtgtctgtatggtgttgc	gtgtagattgtaggtgcatgtgagc	169
RM19296	SSR	ctagcttgacgccaaggacacc	gcacagacgcacactgatctcc	260
RM19298	SSR	tctgcatcaaatctggtgtgtagc	tcctgtagcggtcactcttacacc	299
RM8075	SSR	accaaataagcctctaatggca	gtagcaaactgatagttttgtcactaaag	200
RM19300	SSR	cctaccgcgtcattcacatgc	gacaagatcgacagccgctacg	177
RM19301	SSR	gatggagtcgaggtacgtcaagg	ggcgtcgaggtagtggtaatcg	129
RM8072	SSR	gatcactcaggtcatccattc	aatcagagaggctaaagacaataat	146
RM19308	SSR	cgagttgctttggcctatttgg	atactgacactgcaacggcaacc	182
RM19310	SSR	gcttcttcggccactgaatctgg	tgggtgggtgctcgatctgc	342
RM19311	SSR	tgcggtgctgttcacctactatcg	gcactgaagctggtgcaatcg	94
RM19312	SSR	Gcgacgtgccaagaagagacc	ttccattccaccaaagccttagc	146
RI02242	InDel	agtgagtggtaggagcagcag	caaacatggggttcacacaa	894
RI02657	InDel	ggagattcagctctccatcg	tagcttgcggttagggagac	605

Table 6 Target molecular markers for MAS.

Markers	Type	Chr	Trait	Primer sequence	
				Forward primer	Reverse primer
RM190	SSR	6	AC, GC, GT	gctacaaatagccacccacacc	caacacaagcagagaagtgaagc
BO3_127.8	SSR	8	SC	cgtggctcgacctttttaat	tcaaaccctggttacagcaa
GT11	STS	6	GT	cgagcgagggtttactgttc	ggaggaaacagcagcaactc
RM589	SSR	6	BPH	atcatggtcggtggcttaac	caggttccaaccagacactg
RM587	SSR	6	BPH	ttcccatctgcactaccataatcc	gagcagagatgtgctttgctacc

AC = amylose content; GC = Gel consistency, GT = Gelatinization temperature; SC = scent; BPH = brown planthopper resistance

10. Field evaluation of the selected introgression lines

The agronomic trait performance of the parents and progenies were evaluated in the rice field. Fifty selected introgression lines were grown in rainfed lowland field conditions during wet seasons in 2007 (June to October) at Ubon Ratchathani Rice Research Center, Thailand. Thirty-day-old seedlings were transplanted with a single plant per hill. Each of the plots consisted of five rows with 20 plants per row at the planting density of 20 cm between plants in a row, and between rows. For basal fertilizers, nitrogen, phosphorus (P_2O_5) and potassium (K_2O) were applied at 20, 20 and 20 kg ha^{-1}, respectively. For each line, five plants were sampled at heading (when >50% plants showed panicles). Individual plants were evaluated for plant height (cm) and tiller number per plant. Only forty-eight plants in the middle rows were used to determine the grain yield (g/plant) and its components. Grain weight was calculated at 14% grain moisture content.

11. Determination of the grain quality traits in the selected introgression lines

The experiments for measuring amylose content (AC), gel consistency (GC) and gelatinization temperature (GT) were conducted following the same procedures as described previously by Lanceras *et al*., (2000). AC is measured by simplified iodine colorimetric procedure (Juliano, 1971). The optical density of the amylose-iodine blue color was measured at 620 nm using a spectrophotometer. GC is measured by the length of the gel. The gel consistency values were classified as soft (61-100), medium (41-60) or hard (26-40). The GT of a grain is measured by the alkaline spreading value (ASV); a larger ASV represents more spreading in alkali, indicating that a lower GT and a smaller ASV indicates a higher GT. Alkali spreading values correspond to GT as follows: 1-2, high (74-75°C) and 6-7, low (<70°C).The percentage of grains with chalky endosperm was measured as the number of grains with opacity, counted by visual assessment, in 100 milled rice grains selected randomly from each sample. The determination of aroma was carried out by sensory method according to Wanchana *et al*., 2005. The aroma was categorized into three levels: 0 = none aromatic, 1 = mild and 2 = strong.

12. Identification of a broad-spectrum resistance against BPH populations

To monitor a broad spectrum resistance against BPH populations in Thailand, a set of 50 selected ILs were screened using SSBS against six BPH populations. Four different biotypes of BPH populations (Jairin *et al*., 2007a) were collected from Ubon Ratchathani (UBN), Nan (NAN), Kamphaeng Phet (KPP) and Phitsanulok (PSL) provinces in 2004 and two populations were collected from the outbreak fields at Det Udom (DUD), Ubon Ratchathani province and Wang Thong (WTG), Phitsanulok province in 2007. The SSBS was used to screen the introgression lines. The experiment was conducted in the greenhouse at Phitsanulok and Ubon Ratchathani Rice Research Centers, Rice Department and Rice Gene Discovery Unit, Kasetsart University. Using the SES, the rice plants were scored based on a 1-9 scale, when more than 90% of the TN1 plants were killed.

RESULTS

1. Variation of BPH populations in Thailand

In order to investigate genetic variation in the BPH populations in Thailand, we identified 45 BPH populations based on the differential set of ten resistant rice cultivars [Mudgo (*Bph1*), ASD7 (*bph2*), Rathu Heenati (*Bph3*), PTB33 (*bph2* and *Bph3*), Babawee (*bph4*), ARC10550 (*bph5*), Swarnalata (*Bph6*), T12 (*bph7*), Chin Saba (*bph8*) and Pokkali (*Bph9*)] using SSBS bioassay. The SSBS showed the differences of susceptibility of the differential set of ten resistant rice cultivars. We found that variations in virulence occurred among BPH populations against resistant cultivars carrying specific resistance genes. The result indicated that variations of virulence do exist among BPH populations from different geographic locations (Table 7). We subsequently classified the 45 BPH populations into four major groups based on SSBS and a similarity relationship of more than 0.88 (Figure 12). BPH populations in the group I were able to infest rice cultivars carrying resistance genes *Bph1*, *bph2*, *bph7*, *bph8* and *Bph9*. BPH populations in the group II were able to infest rice cultivars with *Bph1*, *bph8* and *Bph9* resistance genes. BPH populations in the group III were able to infest rice cultivars with *Bph1*, *bph2*, *bph5*, *bph8* and *Bph9* and the populations in the group IV were able to infest rice cultivars with *Bph1*, *bph5*, *bph8,* and *Bph9.*

In order to investigate the variation of the BPH populations using AFR, we also found that the variations in feeding rate, which measured by honeydew production, occurred among BPH populations when fed on the set of different resistant cultivars. The honeydew production area on filter paper treated with bromocresol green solution was presented in Table 8.

Base on the SSBS and AFR bioassays of the collected BPH populations, cultivars carrying the BPH-resistance genes, *Bph1*, *bph2*, bph5, *bph7*, *bph8* and *Bph9*, can classify as a susceptible group while cultivars with *Bph3*, *bph4*, and *Bph6* were classified as a resistant group. Only two resistant cultivars, Rathu Heenati and PTB33, both carrying *Bph3*, showed a broad-spectrum of resistance against all BPH populations used with both SSBS and AFR bioassays (Table 7-8, Figure 13-14).

According to the designation of BPH biotypes using SSBS and based on their reactions on four different resistant cultivars, Mudgo, ASD7, Rathu Heenati and Babawee, six different BPH biotypes were detected in this study (Table 9). Four biotypes were biotype 1, 2, 3 and 4 and the remaining two were new biotypes. Biotype 1 was not able to infest any cultivars whereas Biotype 2 and 3 were able to infest only cultivars with *Bph1* and *bph2* resistance gene, respectively. Biotype 4 was able to infest cultivars with both *Bph1* and *bph2* resistance genes. Two new biotypes showed different reaction from biotypes 1-4. The BPH populations were able to infest the cultivars carrying *bph4*.

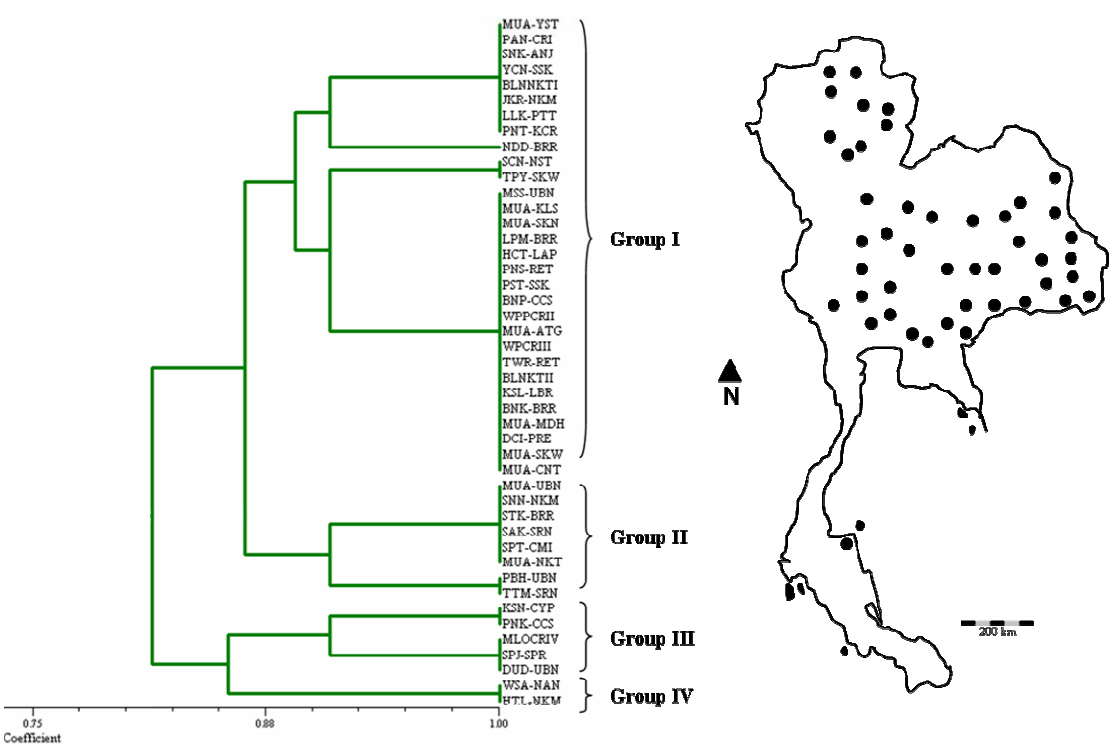

Figure 12 Cluster analysis of 45 BPH populations collected from the rice fields in Thailand (black spots in the map), based on the damage scores of the differential set of ten BPH resistant cultivars. The dendrogram was constructed using UPGMA based on Dice similarity coefficients. Scale of the dendrogram is the Dice coefficient of similarity.

Table 7 Reaction of a differential set of resistant cultivars with specific resistance genes to some BPH populations using the standard seedbox screening technique. The BPH populations were randomly selected from the total of 45 populations.

Variety	R gene[a]	Reaction of rice cultivar to brown planthopper populations[b]															
		YST	NST	KLS	UBN	CYP	NAN	CMI	ANJ	SKW	NK	ATG	BRR	SPR	CCS	SRN	SRN
TN1	None	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
Mudgo	*Bph1*	S	R	S	S	S	S	S	S	R	S	S	S	S	S	S	S
ASD7	*bph2*	S	S	S	S	S	R	S	S	S	R	S	S	S	S	S	S
Rathu	*Bph3*	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R
PTB33	*Bph3*	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R
Babawee	*bph4*	R	S	S	S	S	S	S	R	S	S	S	R	R	S	S	R
ARC10550	*bph5*	S	S	S	S	S	S	S	S	S	S	S	R	S	S	S	S
Swarnalata	*Bph6*	R	R	R	S	R	R	S	R	R	R	R	R	R	R	S	S
T12	*bph7*	S	S	S	S	R	R	S	S	S	R	S	S	R	R	S	S
Chin Saba	*bph8*	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
Pokkali	*Bph9*	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S

[a] R gene, BPH resistance gene

[b] S, Susceptible R, Resistance

Table 8 Reaction of a differential set of resistant cultivars with specific resistance genes to some BPH populations collected in Thailand using the antibiosis of feeding rate technique. The BPH populations were randomly selected from the total of 45 populations.

Variety	PANCRI	MUAPSL	DCIPRE	MUAMDH	MUAYST	SNKANR	MUANKN	WSANAN	MLACRI	MUASKN	MUAUBN
TN1	5.49±1.88	6.55±0.58	9.16±0.64	11.00±0.82	6.88±3.64	5.47±0.52	10.85±4.11	6.36±2.53	3.33±0.40	5.02±2.42	7.22±1.85
Mudgo	4.27±0.41	1.96±0.44	9.71±0.94	5.28±1.02	6.74±0.99	4.26±1.43	8.87±4.10	6.15±1.80	9.58±6.33	5.91±0.90	10.55±3.59
ASD7	5.52±1.48	3.17±0.87	13.21±2.42	10.31±1.70	7.63±1.88	7.18±1.08	7.81±2.53	3.18±0.99	5.56±2.28	2.97±0.41	9.26±3.12
Rathu Heenati	0.77±0.73	1.98±0.09	0.52±0.07	0.33±0.30	1.00±0.60	0.67±0.40	3.12±0.62	1.34±0.65	1.37±0.49	0.75±0.29	0.41±0.32
PTB33	0.37±0.13	0.32±0.22	0.08±0.03	0.05±0.00	1.06±0.81	0.22±0.24	0.51±0.12	0.71±0.0	0.16±0.23	0.78±0.55	0.16±0.23
Babawee	2.93±1.10	1.73±0.42	2.00±0.80	0.71±0.67	4.79±1.99	4.58±0.94	2.96±1.44	3.46±1.91	3.91±2.52	4.45±2.44	1.09±0.41
ARC10550	5.87±1.64	7.26±1.08	19.06±2.62	10.65±3.02	8.33±3.72	7.58±1.84	7.90±3.36	3.64±0.65	7.92±1.28	7.62±2.69	7.75±2.11
Swarnalata	1.59±0.50	1.24±0.13	0.98±0.21	3.20±1.57	0.20±0.14	0.85±0.40	4.85±1.49	1.50±0.89	0.06±0.04	0.53±0.98	11.7±5.18
T12	6.58±1.93	6.78±1-86	5.46±3.59	11.75±1.50	10.75±2.16	7.45±2.61	8.43±5.45	6.97±1.86	9.02±4.99	8.66±1.68	11.72±2.39
Chin Saba	4.50±0.25	1.55±0.26	8.40±2.23	5.52±1.72	6.35±1.58	6.78±1.58	12.53±4.28	5.83±4.11	8.60±4.60	4.08±0.84	12.43±5.21
Pokkali	5.68±1.13	2.22±1.09	3.32±1.46	13.86±2.11	9.05±0.48	6.81±0.82	9.51±2.84	5.40±2.36	6.11±3.22	5.98±2.39	3.34±0.98
Abhaya	5.85±1.36	2.38±1.23	2.01±0.47	7.65±0.24	5.63±1.35	2.03±0.75	3.98±1.34	4.08±1.65	2.55±0.72	2.16±0.82	3.64±1.36
LSD 0.05	1.54	1.24	2.87	2.13	2.80	1.79	4.42	2.77	4.36	2.31	4.00

Table 9 Reaction of resistant rice cultivars to BPH biotypes found in Thailand.

Cultivar	Reaction of BPH population					
	Biotype 1	Biotype 2	Biotype 3	Biotype 4	New biotype 1	New biotype 2
Mudgo	R	S	R	S	S	S
ASD7	R	R	S	S	R	S
Rathu Heenati	R	R	R	R	R	R
Babawee	R	R	R	R	S	S

R=resistant

S=susceptible

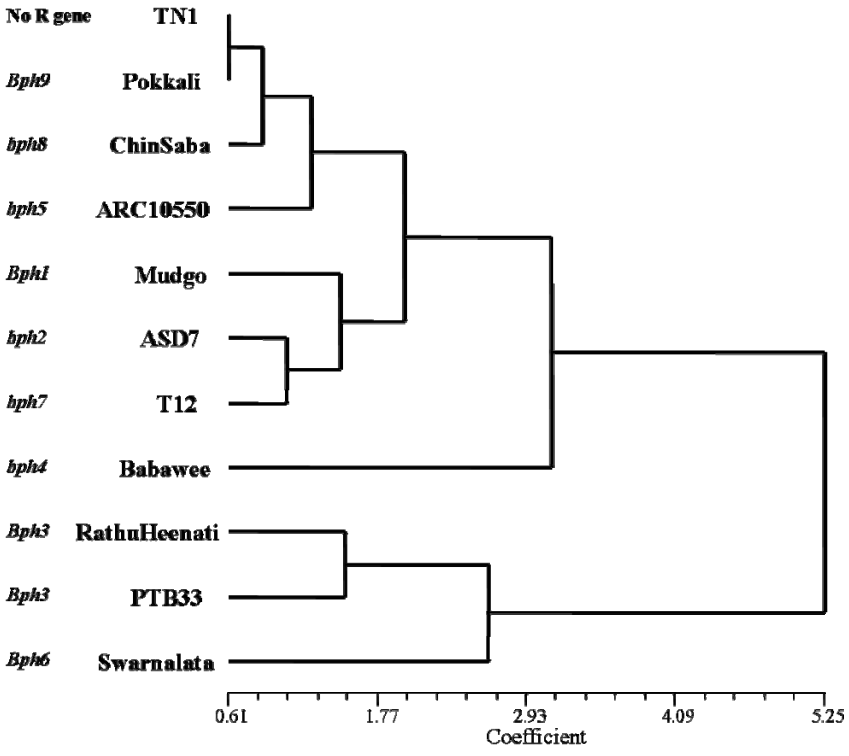

Figure 13 Cluster analysis of a differential set of rice cultivars based on data obtained from seedbox screening test of 45 BPH populations.

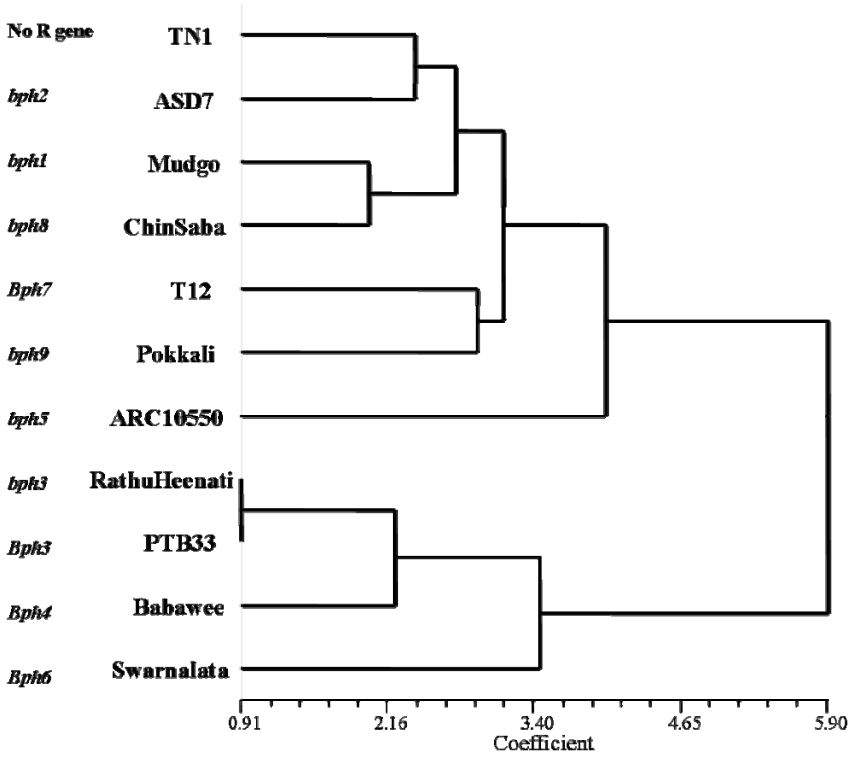

Figure 14 Cluster analysis of a differential set of rice cultivars based on data obtained from antixenosis on feeding rate of 15 BPH populations.

2. Evaluation of BPH resistance in parents

2.1 BPH resistance in the vegetative stage of rice plants

Five bioassays, SSBS, MMTS, SMFS, AFR and AFP, were used to evaluate the reaction to BPH of parental cultivars, Rathu Heenati, PTB33, KDML105 and RD6. At the seedling and tillering stages of the SSBS, MMTS and SMFS bioassays, Rathu Heenati and PTB33 expressed strong resistance to the BPH biotype in Thailand, while RD6 and KDML105 were completely susceptible to the BPH (Table 10).

Feeding rate of the BPH on the parental cultivars as indicated by area of honeydew excretion on filter papers were measured and analyzed. The areas of honeydew were varying upon different rice cultivars. The area of honeydew on Rathu Heenati, and PTB33 were significantly lower than RD6, KDML105 and TN1 (Figure 15, 16a). The results clearly showed that Rathu heenati and PTB33 were resistance to the BPH.

With a free choice among three rice varieties from the antixenosis on feeding preference test, the settling response of BPH initially was not significant different on all cultivated rice. The number of BPH on Rathu Heenati and PTB33 plants were significantly lower than TN1 plants after 24 h of infestation. The BPH began randomly landed on all rice plants and started to move to TN1 plants increasingly after 6 h of infestation. The majority of insects chose and landed on TN1 plants in the 53 h observation period (Figure 16b). The results from feeding rate and feeding preference might be indicated that Rathu Heenati and PTB33 confer resistance principally attributable to antibiosis and/or antixenosis mechanisms.

Table 10 Average damage score of the parents to BPH at vegetative stage (seedling and tillering stages) of rice plants.

Cultivar	Seedling stage by SSBS			Tillering stage by MMTS		
	7 DAI	10 DAI	14 DAI	7 DAI	15 DAI	23 DAI
Rathu Heenati	1.0 (R)	2.2 (R)	2.4 (R)	1.0 (R)	1.0 (R)	1.0 (R)
PTB33	1.0 (R)	2.4 (R)	3.5 (R)	1.0 (R)	1.0 (R)	1.0 (R)
KDML105	6.5 (MS)	8.9 (S)	9.0 (S)	5.0 (MS)	9.0 (S)	9.0 (S)
TN1	7.0 (MS)	9.0 (S)	9.0 (S)	5.0 (MS)	9.0 (S)	9.0 (S)

DAI=Days after infestation

Damage score: 1 = very slight damage, 9 = all plants dead (R = resistant; MS = moderately susceptible; S = susceptible)

SSBS, Standard seedbox screening technique

MMTS, Modified mass tiller screening technique

Figure 15 Area of honeydew excretion from BPH on filter papers treated with bromocresol green solution after released new emerging brachypterous females of BPH 24 h.

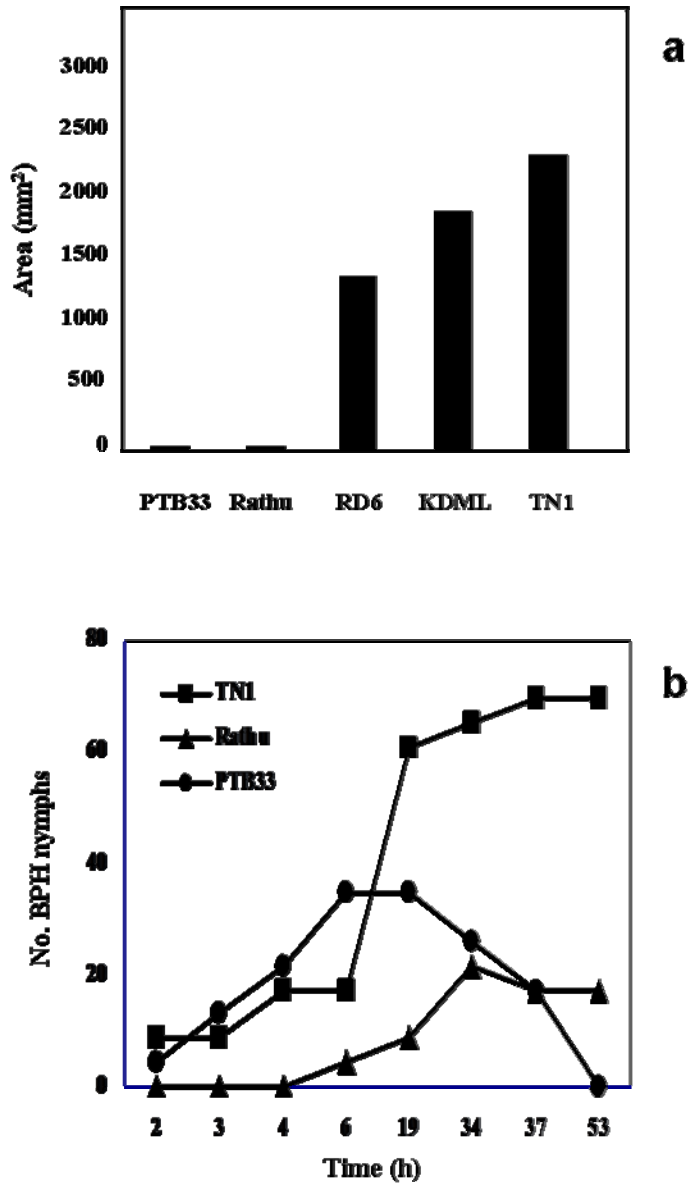

Figure 16 Antibiosis on feeding rate and antixenosis on feeding preference of BPH on the test plants **(a)** Area of honeydew excretion on filter paper (mm^2) of the parents (**b**) number of BPH nymphs alighting on the test plants.

2.2 BPH resistance in the reproductive stage of the rice plants

We attempted to evaluate the BPH resistance at the flowering stage of the parents in the rice field using SMFS. Although Rathu Heenati and PTB33 showed highly resistance to BPH in the vegetative stage (seedling to tillering stages) of the heavy BPH infestation (Figure 17a, Table 11), they showed susceptibility during the reproductive stage (flowering to grain filling stage) when the remaining BPH in the field moved to feed on the panicles and panicle necks until plants died (Figure 17b,c, Table 11). Similar to Rathu Heenati, the resistant BC_3F_2 lines from a cross between Rathu Heenati and KDML105 were also susceptible to the BPH at flowering and grain filling stages (Figure 17d). The result indicated that Rathu Heenati and ILs were susceptible to BPH at the flowering stage and the BPH can feed and grow well on panicles of the resistant plants carrying *Bph3*.

2.3 BPH resistance in vegetative stages with and without N application

The effect of nitrogen fertilizer application on the resistance of the parents was studied using SSBS and MMTS methods. At the seedling stage, Rathu Heenati and PTB33 expressed strong resistance to BPH in the seedboxes without N application. On the other hand, in the seedboxes with nitrogen application all resistant cultivars were susceptible to BPH. The resistance in Rathu Heenati and PTB33 were reduced (increasing of the damage score) in the seedboxes with nitrogen application (Table 12). At the tillering stage, Rathu Heenati and PTB33 also expressed strong resistance to BPH in the free nitrogen application, while the resistance was reduced in the plots with nitrogen application (Table 13, Figure 18). It indicated that nitrogen application has an effect on the resistance in Rathu Heenati and PTB33 at the vegetative stage.

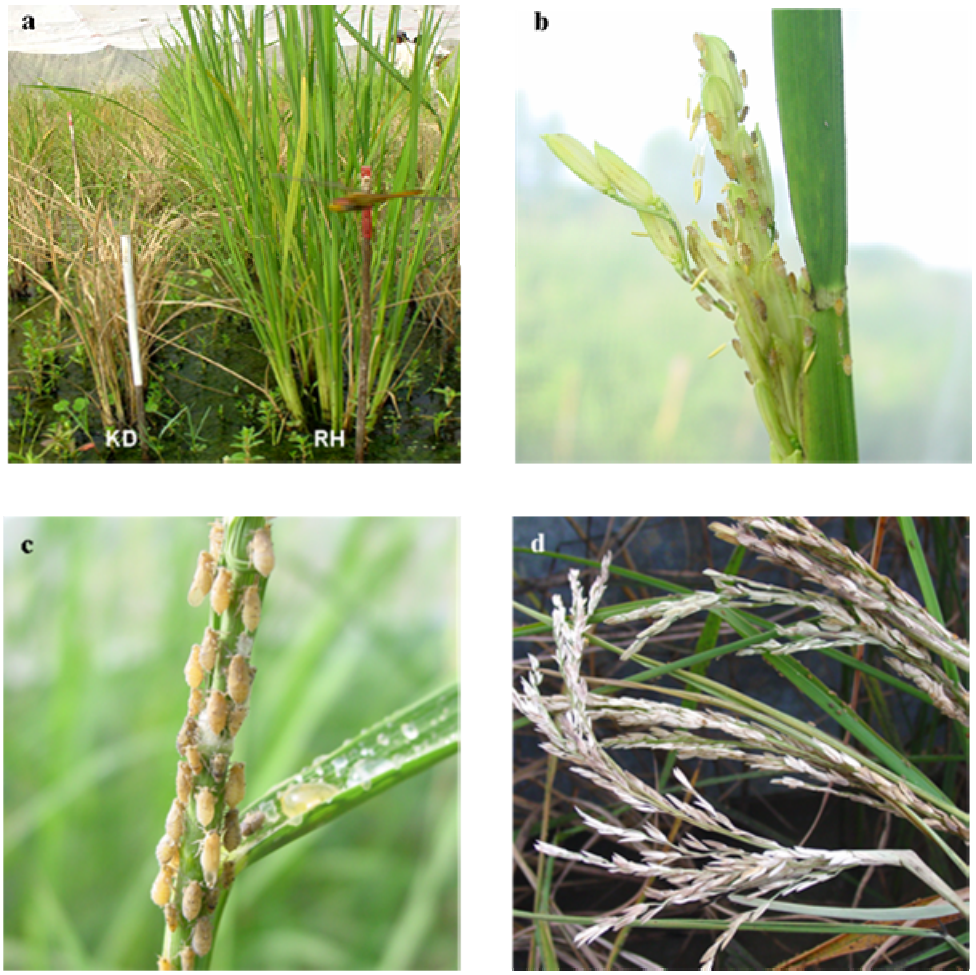

Figure 17 Evaluation of BPH resistance of Rathu Heenati in the rice field using semi-field screening **(a)** Rathu Heenati was highly resistant while KDML105 was susceptible to BPH at the vegetative stage, **(b)** BPH nymphs could feed on the panicle at the flowering stage, **(c)** BPH fed on the panicle neck at the grain filling stage, **(d)** The feeding of BPH caused the unfilled grain before the rice plant die.

Table 11 Average damage score of the parents and controls to BPH in the rice field.

Cultivar	30 DAI	40 DAI	50 DAI	Flowering stage
Rathu Heenati	1.0 (R)	1.0 (R)	1.6 (R)	9.0 (S)
PTB33	1.0 (R)	1.1 (R)	2.0 (R)	9.0 (S)
KDML105	9.0 (S)	9.0 (S)	9.0 (S)	9.0 (S)
TN1	9.0 (S)	9.0 (S)	9.0 (S)	9.0 (S)

DAI=Days after infestation, Damage score: 1=very slight damage, 9=all plants dead

Table 12 Average damage score of the parents and controls to BPH at the seedling stages of rice plants with and without nitrogen fertilizer applications.

Cultivar	With nitrogen application			No nitrogen application		
	7 DAI	10 DAI	14 DAI	7 DAI	10 DAI	14 DAI
Rathu Heenati	4.0 (MR)	5.3 (MS)	7.0 (MS)	1.0 (R)	2.2 (R)	2.4 (R)
PTB33	4.5 (MR)	6.5 (MS)	8.0 (S)	1.0 (R)	2.4 (R)	3.5 (R)
KDML105	7.5 (MS)	9.0 (S)	9.0 (S)	6.5 (MS)	8.9 (S)	9.0 (S)
TN1	7.0 (MS)	9.0 (S)	9.0 (S)	7.0 (MS)	9.0 (S)	9.0 (S)

DAI=Days after infestation; Damage score: 1=very slight damage, 9=all plants dead

Table 13 Average damage score of the parents and controls to BPH at tillering stages of rice plants with and without nitrogen fertilizer applications.

Cultivar	With nitrogen application			No nitrogen application		
	7 DAI	15 DAI	23 DAI	7 DAI	15 DAI	23 DAI
Rathu Heenati	3.0 (R)	5.0 (MS)	6.5 (MS)	1.0 (R)	1.0 (R)	1.0 (R)
PTB33	4.0 (MR)	7.0 (MS)	8.5 (S)	1.0 (R)	1.0 (R)	1.0 (R)
KDML105	5.0 (MS)	9.0 (S)	9.0 (S)	5.0 (MS)	9.0 (S)	9.0 (S)
TN1	6.0 (MS)	9.0 (S)	9.0 (S)	5.0 (MS)	9.0 (S)	9.0 (S)

DAI=Days after infestation; Damage score: 1=very slight damage, 9=all plants dead

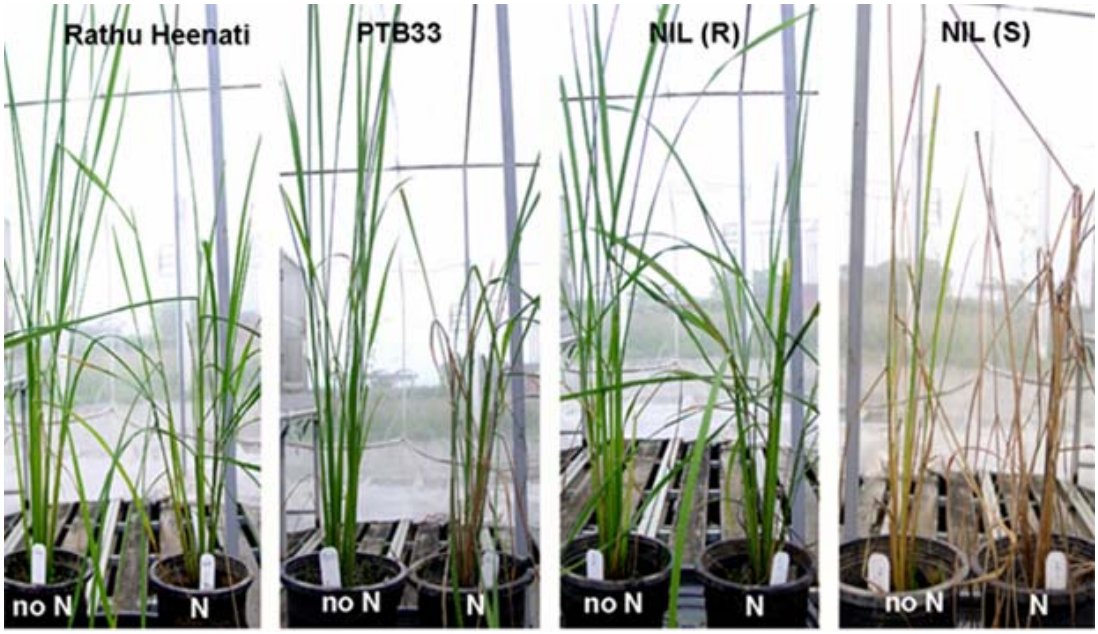

Figure 18 Effect of nitrogen applications on BPH resistance in the rice plants (15 days after infestation).

3. Identification of BPH resistance gene location

The F_1 plants of PTB33×RD6 and Rathu Heenati×KDML105 showed resistance to the BPH by SSBS bioassay, indicating that BPH resistance in PTB33 and Rathu Heenati might be controlled by a dominant gene (Table 14). Segregation of BPH resistance, which is conferred by the introgressed *Bph3* gene, was studied in 208 BC_1F_2 and 333 BC_3F_2 individuals, which derived from crosses from PTB33×RD6 and Rathu Heenati×KDML105, respectively. The resistance score of the 208 BC_1F_2 and 333 BC_3F_2 individuals that infested with the BPH population showed a continuous distribution (Figure 19). We then studied the segregation of BPH resistance in both backcross populations (Table 15) by directly assaying the phenotypes of the BC_1F_2 and BC_3F_2 individuals and found that resistant and susceptible BC_1F_2 and BC_3F_2 plants segregated in a 3:1 segregation ratio (χ^2=1.17, P>0.28; χ^2=0.03, P>0.86, respectively), which indicated the presence of a major dominance gene conferring resistance to the BPH.

To determine the map location of the BPH resistance gene, we assayed 208 BC_1F_2 individuals from the cross of PTB33×RD6 in the R and S groups with 36 polymorphic SSR markers on chromosome 4, 6, and 10 in order to determine which of the SSR markers were associated with resistance/susceptibility. This analysis showed that SSR marker RM190 on chromosome 6 was strongly associated with the resistance/susceptibility and that none of the SSR markers tested were significantly associated with BPH resistance on chromosomes 4 and 10. These results indicated that the BPH resistance gene from PTB33 was linked to RM190 on chromosome 6 (Figure 20). To confirm the location of resistance gene from Rathu Heenati, RM190 was used to identify R and S groups of the BC_3F_2 population from the cross of Rathu Heenati×KDML105. This analysis revealed that RM190 was also strongly associated with the R and S groups of the BC_3F_2. Consequently, the BPH resistance gene in PTB33 and Rathu Heenati was located in the same region on chromosome 6.

To further confirm the chromosome location of the resistance gene, we employed additional SSR markers surrounding the RM190 locus. Of 20 SSR markers tested, only six showed polymorphism between the parents. RM190 and six additional SSR markers on chromosome 6 were used to assay 208 BC_1F_2 and 333 BC_3F_2 progenies. A linkage map was constructed with LOD scores greater than 3.0 based on the segregation data. In the linkage map constructed for chromosome 6, the order of all SSR markers agreed with that of the standard SSR map (McCouch *et al*. 1997, 2002). However, the estimated distances of some markers were larger than those of the standard map. The BPH resistance locus detected from the BC_1F_2 and BC_3F_2 populations was mapped between two flanking markers RM589 and RM588 on the short arm of chromosome 6 within 0.9 and 1.4 cM of these markers, respectively (Figure 21). The tightly linked marker RM589 and RM586 explained 59.8 and 57.4% of the phenotypic variance of the BPH resistance with high LOD scores of 41.1 and 61.6 in the BC_1F_2 and BC_3F_2 populations, respectively (Table 16).

Table 14 Average damage score of the parents and F_1 populations to the BPH at vegetative stage.

Cultivar	SSBS[a]	MMTS[b]	SMFS[c]
TN1	9.0 (S)	9.0 (S)	9.0 (S)
KDML105	9.0 (S)	9.0 (S)	9.0 (S)
RD6	9.0 (S)	9.0 (S)	9.0 (S)
PTB33	2.6 (R)	1.0 (R)	1.4 (R)
Rathu Heenati	2.1 (R)	1.0 (R)	1.1 (R)
F_1 (PTB33×RD6)	3.1 (R)	-	-
F_1 (Rathu Heenati×KDML105)	3.0 (R)	-	-

[a] Standard seedbox screening

[b] Modified mass tiller screening

[c] Semi-field screening

Table 15 Segregation of BPH resistance in the recombinant F_2 and F_3 populations.

Cross combination/Generation	Number of individuals			χ^2 value	*P*
	Resistance	Susceptible	Total		
PTB33×RD6/BC_1F_2	149	59	208	1.17	0.28
Rathu Heenati×KDML105/BC_3F_2	252	81	333	0.03	0.86
Rathu Heenati×KDML105/BC_3F_3	265	93	358	0.18	0.67

[a] χ^2 values based on a 3:1 ratio

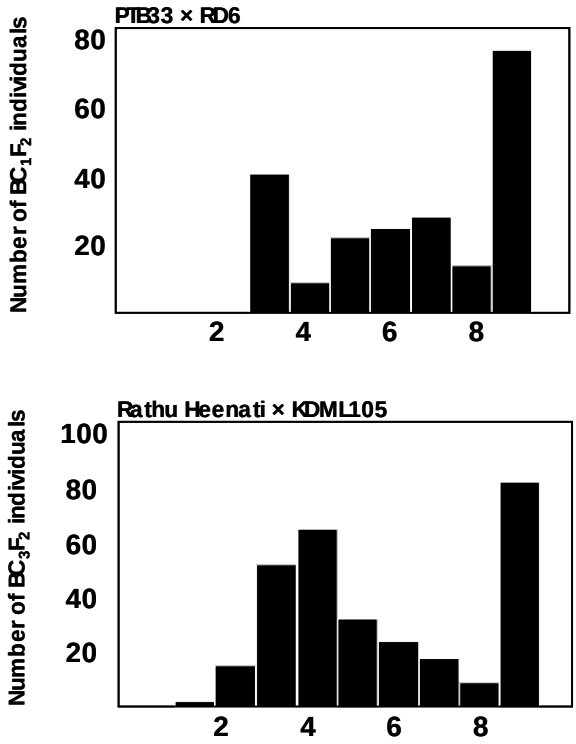

Figure 19 Frequency distribution of BPH damage rating of two mapping populations by the MMTS.

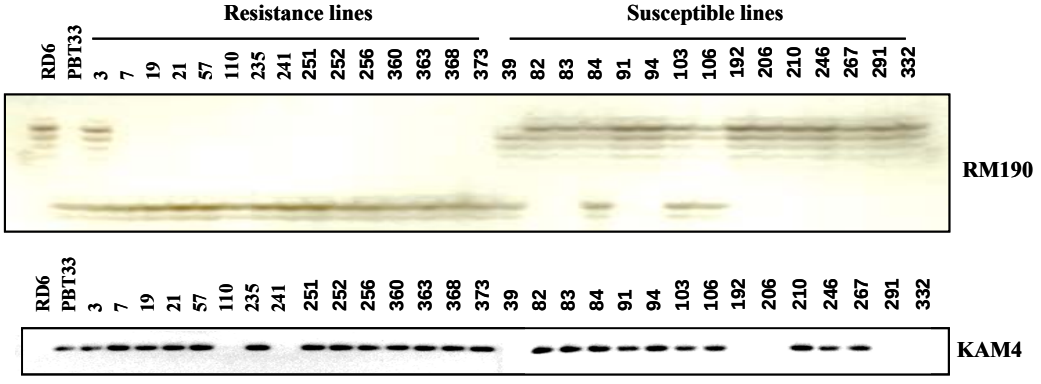

Figure 20 The SSR marker RM190 and the STS marker KAM4 linked to BPH resistance gene *Bph3* and *bph2*, respectively. The markers were identified in resistant and susceptible individual lines of the BC_1F_2 population, which were derived from a cross between PTB33 and RD6.

Table 16 The SSR markers associated to the BPH resistance genes on chromosome 6 in two different mapping populations.

Backcross population	Marker	LOD	R^2 (%)[a]	Additive[b]	*P*
BC_1F_2 (PTB33×RD6)	RM469	38.6	57.5	-2.253	0.000
	RM589	41.1	59.8	-2.259	0.000
	RM588	35.3	54.3	-2.283	0.000
BC_3F_2 (Rathu Heenati×KDML105)	RM589	57.5	54.9	-2.641	0.000
	RM586	61.6	57.4	-2.665	0.000
	RM588	61.5	57.3	-2.673	0.000

[a] Percentage of phenotypic variation explained.

[b] Additive effect of donors allele.

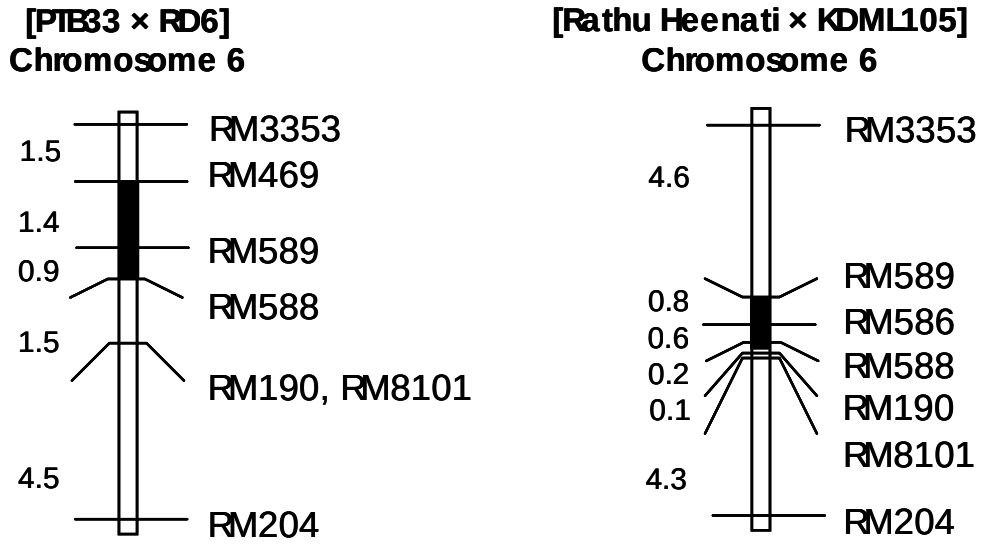

Figure 21 Linkage maps of the BPH resistance genes on the short arm of chromosome 6. Marker names are listed on the right of the chromosomes. The distance between markers is in centiMorgans. The solid bars indicate the location of the BPH resistance genes.

4. Physical mapping of the BPH resistance locus

The genetic mapping data from our previous study was used as a starting point for physical mapping of the *Bph3* locus. Using a backcross population BC_3F_2, derived from a cross between Rathu Heenati and KDML105, *Bph3* was mapped to about 1.4 cM interval between SSR markers RM588 and RM589 on the short arm of chromosome 6. In this study, we selected two BC_3F_2 plants that were heterozygous on the short arm of chromosome 6 where *Bph3* is located. A total of 330 BC_3F_3 plants derived from the selected BC_3F_2 was randomly selected and used to confirm the inheritance of BPH resistance in Rathu Heenati at the vegetative stage. Phenotypic evaluations of BPH resistance for the BC_3F_3 and the parents were conducted using the MMTS. Segregation of resistant and susceptible plants fits in a 3:1 ratio (Table 15).

The location of the *Bph3* resistance gene on the map was determined on the basis of the resistance scores of the 28 recombinant plants. MMTS was employed to distinguish resistant plants from susceptible ones among the recombinant plants. A number of 16 SSR markers located around this genomic region were selected to screen polymorphism between Rathu Heenati and KDML105. Seven SSR markers (RM19291, RM19295, RM19296, RM8072, RM8074, RM19310, and RM19311) detected polymorphisms between the two parents. These seven markers were used to narrow down the region encompassing *Bph3* locus between the two flanking markers RM589 and RM588. The resulting high-resolution map of *Bph3* showed that RM19291 and RM8072 were flanking the *Bph3* resistance gene (Figure 22, 23a). Twenty-eight plants were then identified with recombination break points between the SSR markers RM19291 and RM8072. Of these, three recombinant events were detected with marker RM19291, and five were found with marker RM8072. No recombinants were detected with the other three markers, RM19295, RM19296, and RM589. These three markers were identified to co-segregate with the *Bph3* locus. According to the genome sequence database of a *japonica* rice cultivar Nipponbare, the *Bph3* locus was finally localized to approximately 190 kb interval flanked by markers RM19291 and RM8072 (Figure 23b).

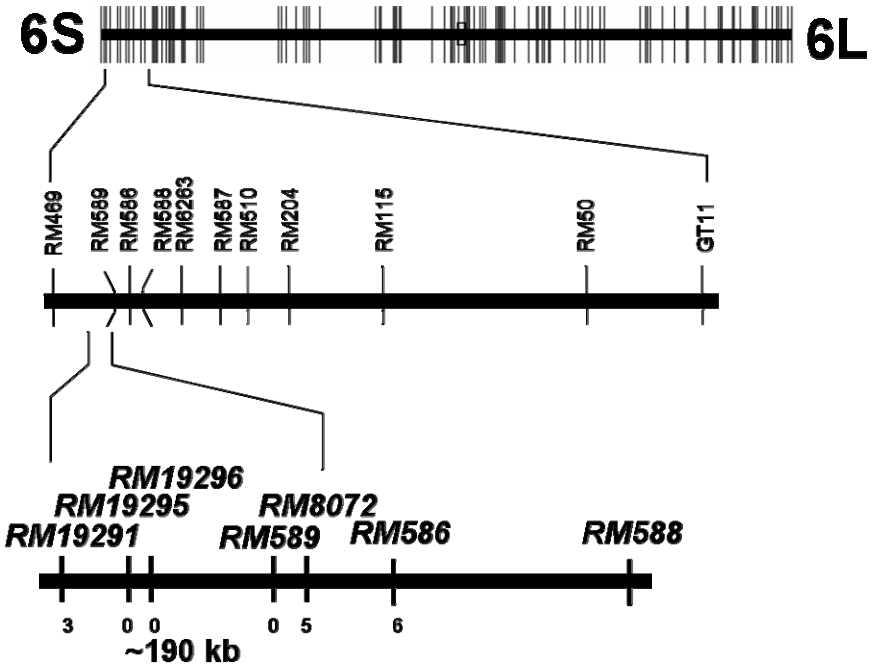

Figure 22 Fine mapping and molecular identification of *Bph3*. *Bph3* was fine mapped on the three of overlapped BAC and PAC clones of *O. sativa* cv. Nipponbare corresponding to the intervals defined by the genetic markers RM588 and RM19291.

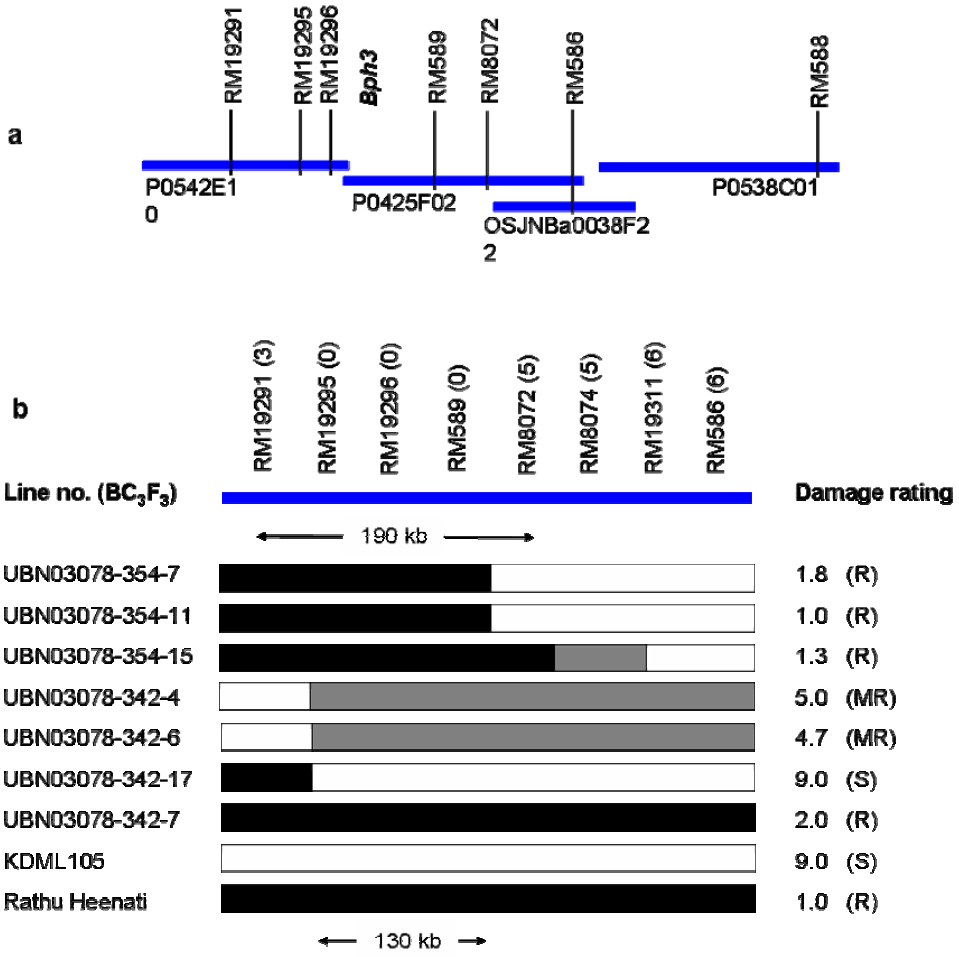

Figure 23 Physical map of the *Bph3* locus. (**a)** Physical mapping of *Bph3* locus showing four Nipponbare BAC clones interval delimited by RM19291 and RM588, (**b)** Genotypes and phenotypes of the recombinants between RM19291 and RM586. Black bars = homozygous Rathu Heenati allele; white bars = homozygous KDML105 allele; gray bars = heterozygous. The numerals in parentheses indicate the recombination events occurred at the corresponding marker loci. The BPH resistance score is on the right, R = resistant, MR = moderately resistant, and S = susceptible.

Based on the available sequence annotation database of the *japonica* rice Nipponbare (http://www.rgp.dna.afrc.go.jp; http://www.tigr.org), there are twenty-two predicted putative genes in the 190 kb target region. Of these genes, seven had unknown functions, seven were hypothetical proteins, and the functional annotation of the remaining eight genes encoded one NBS-LRR disease resistance protein (LOC_Os06g03500), two pentatricopeptides (LOC_Os06g03530 and LOC_Os06g-03570), two oligopeptide transporters (LOC_Os06g03540 and LOC_Os06g03560), one zinc finger, C_3HC_4 type family protein (LOC_Os06g03580), one transcriptional co-regulator family protein (LOC_Os06g03600), and one protein kinase family protein (LOC_Os06g03610).

5. Development of rice introgression lines (ILs) with brown planthopper resistance and KDML105 grain quality characteristics through marker-assisted selection

5.1 Marker validation for marker-assisted selection

To investigate the accuracy of the marker for MAS, a rice population consisted of 330 individuals derived from a heterozygous line of BC_3F_3 was screened for BPH resistance using MMTS method. The BPH resistance scores of the 330 lines showed a continuous distribution, ranging from a low of 1 to a high of 9 (Figure 24, Appendix Table 4). The segregation of BPH resistance in the populations by directly assaying the phenotype of the BC_3F_4 individuals and found that resistant (R), moderately resistant (MR) and susceptible (S) plants segregated in a 1:2:1 segregation ratio (χ^2=1.09, *P*=0.58) (Table 17). Genotype of all 330 BC_3F_4 plants was classified into three categories on the resistance scores as resistance, segregating and susceptibility. The corresponding BC_3F_4 plant was genotyped as RR (homozygous resistance), RS (segregating heterozygous) and SS (homozygous susceptibility), accordingly. The segregation of BC_3F_4 population showed a good fit to the expected ratio of 1:2:1 (χ^2=1.04, *P*=0.59) (Table 17). We investigated the selection efficiency based on RM589 according to the results from the genetic analysis and the phenotypic analysis by BPH resistance. Analysis using RM589 as the marker was performed on

all 330 BC_3F_4 individuals. Based on the phenotype data of BPH resistance, the selection accuracy of the marker was calculated. The results showed that RM589 had high selection accuracy of 80.6% (Figure 25). Thus RM589 could be applied to the MAS of the trait of BPH resistance.

We further confirmed the phenotype of 108 BC_3F_6 individuals derived from a heterozygous line of BC_3F_5 for BPH resistance using MMTS. The BPH resistance scores showed a continuous distribution, ranging from a low of 3 to a high of 9 (Figure 26). Among the 108 BC_3F_6 plants from the cross between Rathu Heenati and KDML105, 25 individuals were resistant, 57 were moderately resistant and the remaining 26 were susceptible, which fit a 1:2:1 segregation on the basis of χ^2 test (χ^2= 0.35, *P*=0.84), further confirming the fact that the resistance in introgression lines is conferred by a single dominant gene.

Table 17 Segregation of BPH resistance of a BC_3F_4 from a cross of Rathu Heenati×KDML105. RR = homozygous Rathu Heenati allele, RS = heterozygous, SS = homozygous KDML105 allele, R = resistant, MR = moderately resistance and S = susceptible.

Genotype/Phenotype	**RR/R**	**RS/MR**	**SS/S**	χ^2	***P***
Genotype	76	168	89	1.04	0.59
Phenotype	75	172	86	1.09	0.58

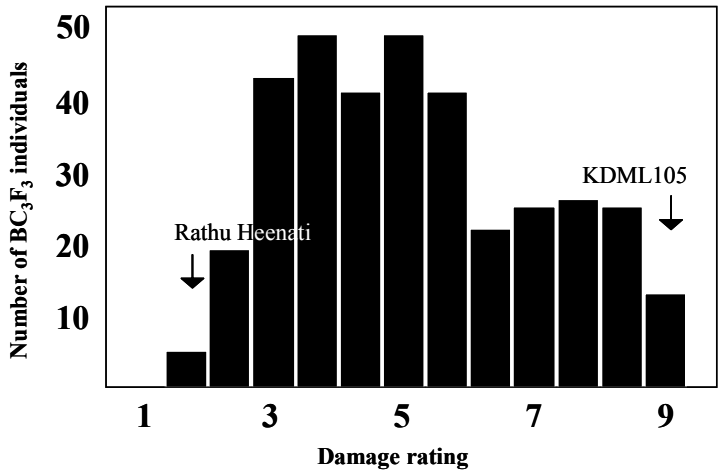

Figure 24 Frequency distribution of BPH resistance scores of BC_3F_4 from a cross Rathu Heenati×KDML105 based on the overall average of four scoring periods from the modified mass tiller screening method at the tillering stage of the rice plants. The mean scores of Rathu Heenati and KDML105 are indicated by arrows.

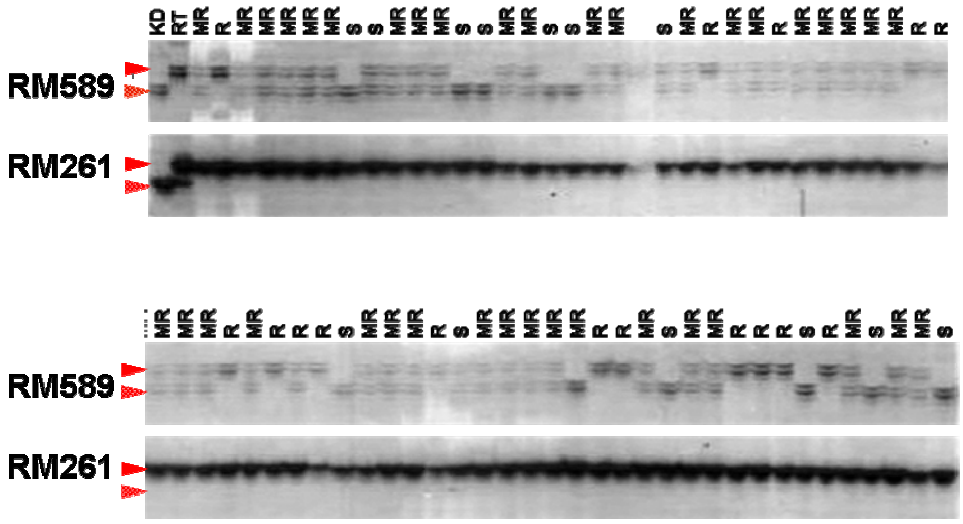

Figure 25 PCR amplification of BC_3F_4 plants with the marker RM589 on chromosome 6 linked to *Bph3* and RM261 on chromosome 4 linked to BPH resistance gene in Rathu Heenati identified by Sun *et al*., 2005.

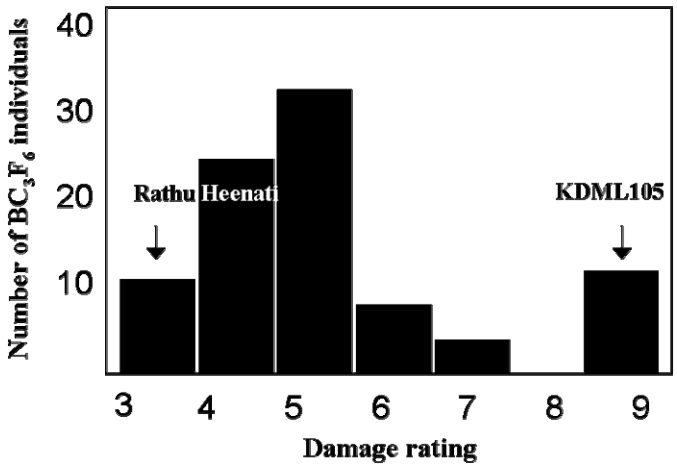

Figure 26 Frequency distribution of BPH resistance scores of a BC_3F_6 from a cross Rathu Heenati×KDML105 based on the damage scoring from the modified mass tiller screening method at the tillering stage of rice plants. The mean scores of Rathu Heenati and KDML105 are indicated by arrows.

5.2 Genetic dissection of the *Bph3* and unfavorable Wx^a allele

Two loci for BPH resistance (*Bph3*) and amylose content (Wx^a) showed co-segregated on short arm of chromosome 6 by linkage mapping of backcross inbred lines derived from the cross between Rathu Heenati and KDML105. The physical distance between two alleles was approximately 383 kb or 2.4 cM base on the SSR mapping (Figure 27). To dissect a linkage between *Bph3* and *Waxy* locus, a total of 2,343 BC_3F_2 progenies from the cross of Rathu Heenati×KDML105 was screened for BPH resistance using MMTS in the greenhouse. A variation in BPH resistance was observed in BC_3F_2 plants, and we classified BC_3F_2 plants into three segregation patterns: resistance, moderately resistance and susceptibility. Two hundred BC_3F_2 resistant and moderately resistant plants were selected for MAS. Ten BC_3F_2 progenies that carried fragrance and GT alleles and showed heterozygous or homozygous on the *Bph3* and/or *Wx* region were selected. Two SSR markers, RM589 and RM190, closely linked to *Bph3* and *Wx* loci, respectively were used to analyze BC_3F_3 progenies derived from the selected resistant BC_3F_2 lines, which showed

heterozygous at the *Bph3* region and KDML105 homozygous at the *Wx* region (Figure 27). Only two BC_3F_3 progenies with slender grains that carry Rathu Heenati homozygous and heterozygous genotypes at *Bph3* region and KDML105 homozygous genotype at *Wx* region was selected to generate BC_3F_4.

5.3 SSR based background analysis

A total of 75 polymorphic SSR markers distributed throughout 12 rice chromosomes were used for background analysis in the fifty selected ILs from the BC_3F_4 population. The presence of the genomic composition of the selected ILs using the SSR markers is shown in Appendix Table 5. The average distance between adjacent markers was ranged from 11.4 cM (chromosome 5) to 30.2 cM (chromosome 3). Among these the percent of markers homozygous for the recipient allele was ranged from 60 to 100%. The background analysis in the ILs revealed the recovery up to 91.2% of the recurrent parent alleles after three generations of backcrossing. The average parental genome recovery of the ILs was 86.9% of the KD genome while that of RH genome was 9.9% with residual heterozygosity of 3.2% (Appendix Table 6).

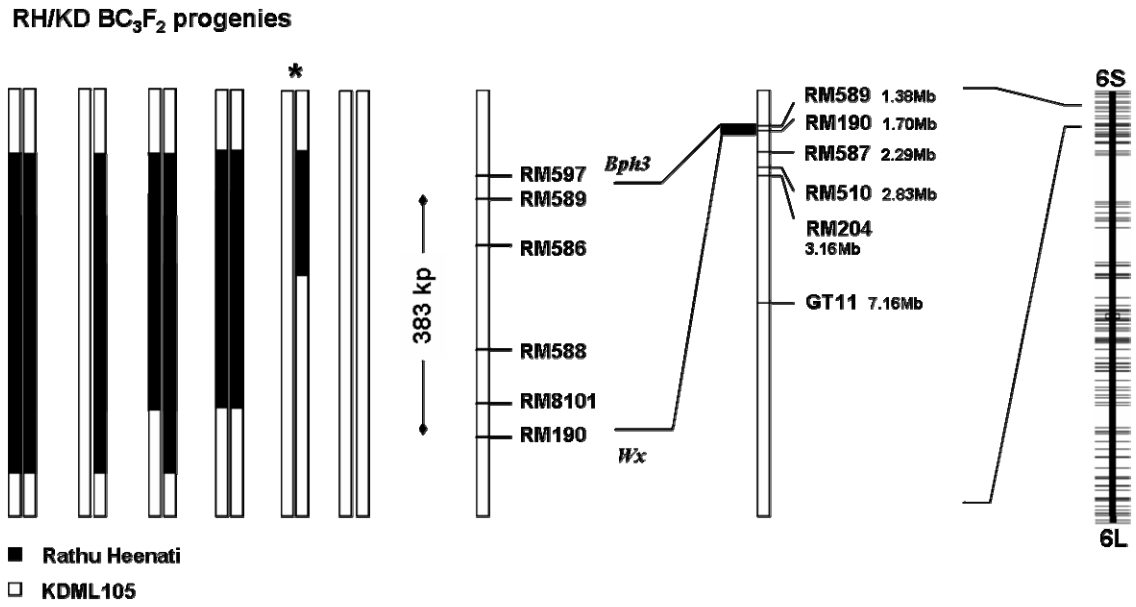

Figure 27 Fine-scale mapping of two loci, *Bph3* and *Wx*, controlling BPH resistance and amylose content, respectively. The locations of two genes, *Bph3* and *Wx* are shown in the linkage map on chromosome 6. Names of SSR markers and genes are shown on the right. Graphical genotypes of the region in six BC_3F_2 plants are shown on the left; white blocks regions derived from KDML105 and black blocks regions derived from Rathu Heenati. The progeny with an asterisk was selected to develop the BC_3F_3.

6. Evaluation of ILs for BPH resistance and other traits

6.1 Evaluation of ILs for BPH resistance

To confirm the resistance against BPH at seedling stage, six rice cultivars including PTB33, Abhaya, Rathu Heenati, IR72, TN1, KDML105, and RD6 and the mixture of seeds of selected ILs from the cross of Rathu Heenati×KDML105 were evaluated at seedling stage in the temperature-controlled room. All resistance cultivars, PTB33, Abhaya, Rathu Heenati and IR72, and the mixture of selected ILs showed highly resistance to BPH whereas all susceptible cultivars, TN1, KDML105 and RD6, were completely susceptible to the BPH (Figure 28). At the tillering stage in the greenhouse, the selections were resistance to BPH as well (Figure 29).

To confirm the level and broad spectrum of resistance against BPH populations collected in Thailand, fifty selected ILs were evaluated at the seedling stage using SSBS in the greenhouses at Rice Gene Discovery Unit, Phitsanulok Rice Research Center and Ubon Ratchathani Rice Research Center. The BPH populations were selected based on its variations from the previous study to determine a broad spectrum of resistance of the selected lines. All selected ILs carrying *Bph3* showed resistance to all BPH populations used in this study (Figure 30, Table 18). The result indicated that a broad spectrum BPH resistance gene which has been introgressed from Rathu Heenati to KDML105 was effectively against the variation of BPH populations found in Thailand.

Under free choice conditions in the antixenosis on feeding preference test, BPH avoided settling on seedlings of Babawee, PTB33, Rathu Heenati and resistant ILs, which also showed the high level of resistance to BPH in the seedbox screening. Number of BPH was decrease on resistant plants after 72 h. During this period, the number of BPH nymphs settled on the susceptible ILs and KDML105 remained significantly higher and increasing after 72 h (Figure 31).

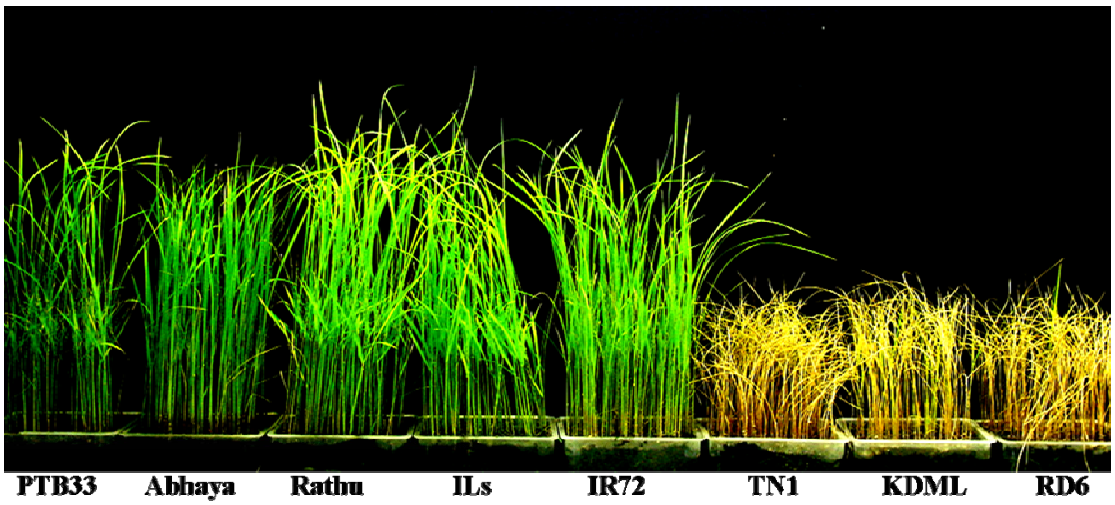

Figure 28 The levels of resistance to BPH at seedling stage in some rice cultivars and the mixture seeds of some selected introgressed lines. The screening was conducted in temperature-controlled room (26ºC).

Figure 29 The levels of resistance to BPH at tillering stage in some introgressed lines, KDML105, a susceptible recurrent cultivar, and TN1, a susceptible cultivar. The plants have been exposed to BPH feeding for two generations of the insects.

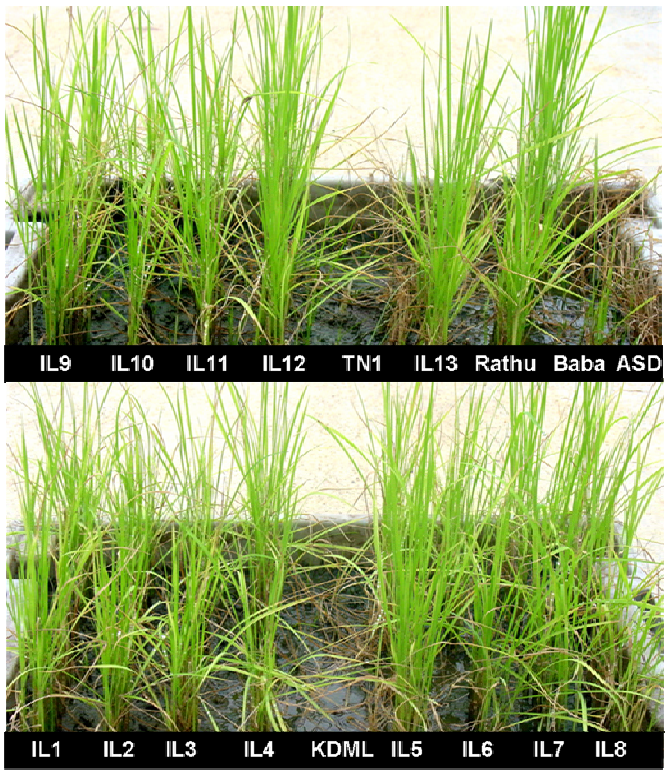

Figure 30 The levels of resistance to BPH at seedling stage in some introgressed lines, KDML105, Rathu Heenati, Babawee and ASD7.

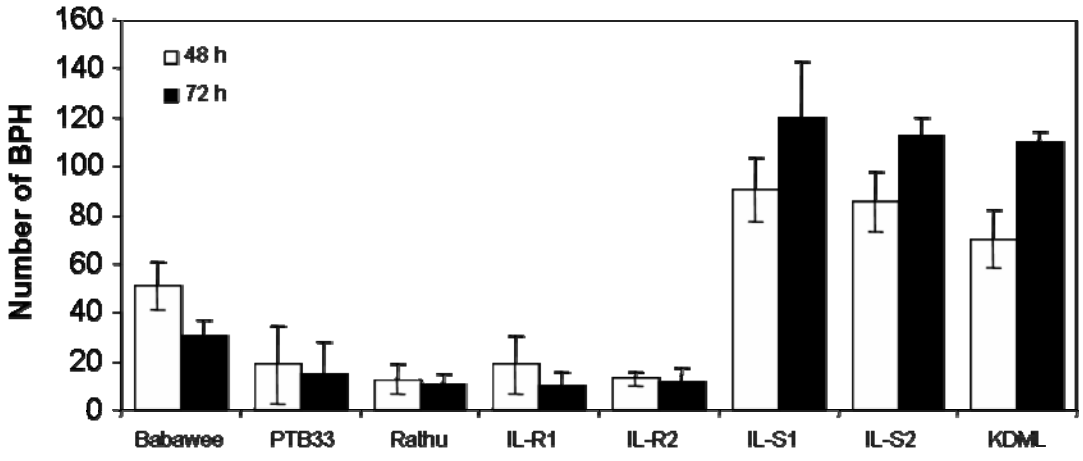

Figure 31 Number of BPH nymphs (means ± SE) settled on rice cultivars and some introgressed plants in a choice test during 48 and 72 h after infestation.

Table 18 The reaction of some introgressed lines to BPH populations collected in Thailand. The SSBS was used to evaluate the resistance.

Designation	Reaction to BPH populations*					
	UBN	**DUD**	**NAN**	**KPP**	**WTG**	**PSL**
UBN03078-80-354-20	R	R	R	MR	R	MR
UBN03078-101-342-9	R	R	R	R	R	R
UBN03078-101-342-11	R	R	R	R	R	R
UBN03078-101-342-14	R	MR	R	R	R	R
UBN03078-101-342-4-24	R	R	R	MR	R	MR
UBN03078-101-342-4-32	R	MR	R	MR	R	MR
UBN03078-101-342-4-96	R	S	S	MR	R	MR
UBN03078-101-342-4-106	R	R	MR	MR	R	MR
UBN03078-101-342-4-111	R	R	R	MR	R	MR
UBN03078-101-342-4-114	R	R	R	MR	R	MR
UBN03078-101-342-4-126	R	R	R	R	R	R
UBN03078-101-342-4-143	R	R	R	R	R	R
UBN03078-101-342-4-144	R	MR	MR	R	R	MR
UBN03078-101-342-4-148	R	R	R	R	R	R
UBN03078-101-342-4-158	R	R	R	MR	R	MR
UBN03078-101-342-6-49	R	R	R	R	R	MR
UBN03078-101-342-6-56	R	R	R	R	R	R
UBN03078-101-342-6-58	R	R	R	R	R	R
KDML105	S	S	S	S	S	S
Rathu Heenati	R	R	R	R	R	R

R = resistance; MR = moderately resistance; S = susceptible

* Four different biotypes of BPH populations (Jairin *et al*., 2007a) were collected from four provinces, Ubon Ratchathani (UBN), Nan (NAN), Kamphaeng Phet (KPP) and Phitsanulok (PSL), in 2004. Two BPH populations were collected from the outbreak fields from Det Udom (DUD), Ubon Ratchathani province and Wang Thong (WTG), Phitsanulok province in 2007.

6.2 Agronomic performance of ILs in the field trial

The agronomic performance of the selected ILs was evaluated in the rainfed lowland field in 2007. The results showed almost all of the morphological traits of ILs, including plant type, flowering date and appearance grain quality were as same as those of KDML105 (Table 19). The distribution of agronomic traits based on the phenotypes in the ILs is shown in Figure 32. The average plant height of ILs varied from 122.2 to 164.6 cm. Thirteen of the ILs had shorter plant height than KDML105 (139.0 cm). The average plant height of the ILs was found 3.8% higher than that of KDML105. The numbers of days to flowering of the selected lines were almost same as those of KDML105 (127 days). However, some selected ILs had 3-7 days delayed flowering than KDML105. The average number of panicles and grain yield per plant of the selections were 10.0 and 18.4% higher than those of KDML105, respectively. The average number of filled-grains and 1000-grain weight of the ILs were 18.9 and 6.5% higher than those of KDML105, respectively. The grain yield per plant of ILs was ranged from 17.6 to 39.0 g. All of the selected lines were awnless and white pericarp, unlike the donor Rathu Heenati, which has prominent awning and red pericarp.

The correlation of the phenotypic performance is shown in Table 5. Panicle number per plant was correlated positively with plant height ($r = 0.54$, $P < 0.001$). Grain yield per plant showed significant positive correlations with panicle number per plant, number of grain per panicle as well as with plant height ($r = 0.37$, $P < 0.01$; $r = 0.40$, $P < 0.01$; $r = 0.42$, $P < 0.01$, respectively) and was not significant with 1000-grain weight. However, 1000-grain weight was correlated positively with grain width ($r = 0.57$, $P < 0.001$). Flowering date was correlated negatively with grain yield and number of grain per panicle ($r = -0.37$, $P < 0.01$; $r = -0.39$, $P < 0.01$, respectively).

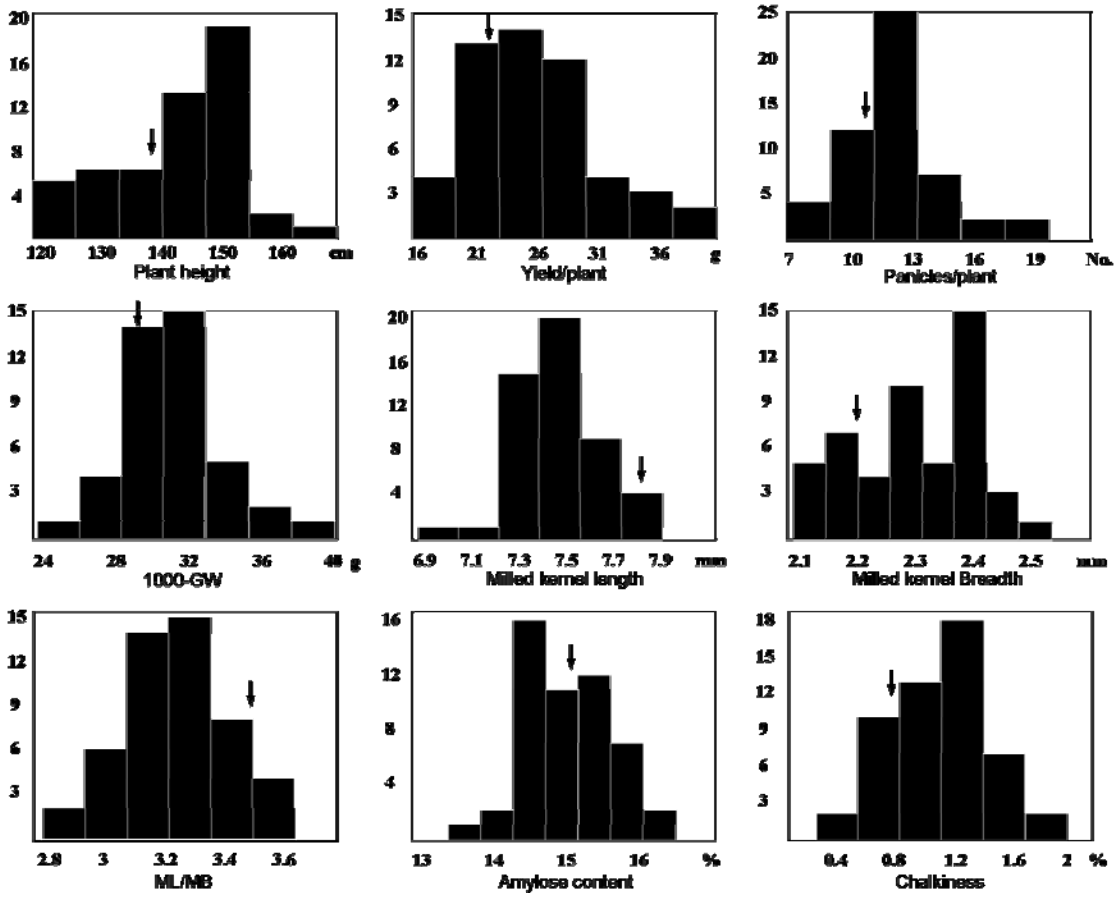

Figure 32 Frequency distribution of plant height, grain length, panicle per plant, grain yield per plant, 1000-grain weight, milled rice kernel length, milled rice kernel breadth and milled rice kernel length/breadth in BC_3F_{4-6} progeny derived from the cross Rathu Heenati×KDML105. Arrows show the mean of KDML105.

Table 19 Performance of principal agronomic traits of some selected introgressed line plants.

Designation	DH	PN	GP	NP	NU	PH	GY	GL	GB	GL/ GB	ML	MB	ML/ MB	GW
UBN03078-80-354-20	127	12.0	2.23	73.0	7.4	131.7	21.6	10.9	2.7	4.1	8.3	2.1	4.0	29.9
UBN03078-101-342-9	130	9.8	2.51	79.8	8.4	122.8	24.5	10.9	2.6	4.2	7.8	2.3	3.4	34.1
UBN03078-101-342-11	131	9.6	2.83	84.0	10.2	130.8	31.2	10.2	2.7	3.8	7.4	2.5	3.0	36.0
UBN03078-101-342-14	130	8.8	2.31	69.0	6.4	133.0	24.8	10.5	2.4	4.4	7.4	2.1	3.5	31.2
UBN03078-101-342-4-24	128	11.2	2.72	96.2	13.4	152.2	32.0	10.2	2.5	4.1	7.3	2.2	3.3	29.7
UBN03078-101-342-4-32	127	11.6	3.53	122.8	20.6	159.4	29.3	10.7	2.4	4.5	7.7	2.2	3.5	25.5
UBN03078-101-342-4-96	127	15.0	2.83	95.0	13.0	155.6	24.2	10.4	2.5	4.2	7.5	2.2	3.4	29.6
UBN03078-101-342-4-106	127	11.8	3.80	130.0	8.8	152.8	27.1	10.3	2.5	4.2	7.2	2.2	3.2	27.8
UBN03078-101-342-4-111	127	17.2	2.69	93.0	19.4	152.4	29.4	10.5	2.5	4.2	7.4	2.3	3.3	30.3
UBN03078-101-342-4-114	127	15.4	3.43	132.6	19.4	132.0	34.2	10.5	2.4	4.3	7.6	2.2	3.4	30.4
UBN03078-101-342-4-126	128	12.0	2.26	68.6	13.8	153.2	26.1	10.6	2.7	3.9	7.6	2.4	3.1	32.1
UBN03078-101-342-4-143	127	19.2	3.72	132.4	16.6	143.8	23.3	10.5	2.4	4.3	7.7	2.1	3.7	27.1
UBN03078-101-342-4-144	127	15.4	2.68	96.0	10.8	154.6	25.9	10.8	2.4	4.6	7.8	2.0	3.9	29.4
UBN03078-101-342-4-148	127	19.4	3.62	123.2	17.0	152.2	35.8	10.8	2.4	4.5	7.9	2.0	4.0	28.1
UBN03078-101-342-4-158	128	18.8	3.49	109.0	16.8	150.2	25.7	10.0	2.7	3.7	7.4	2.2	3.4	31.6
UBN03078-101-342-6-49	127	14.8	3.01	92.4	16.2	137.6	26.1	11.0	2.8	3.9	8.0	2.4	3.3	31.6
UBN03078-101-342-6-56	127	13.0	4.07	122.4	13.8	139.6	24.5	10.8	2.7	4.1	7.8	2.4	3.3	32.7
UBN03078-101-342-6-58	127	13.2	3.03	93.4	38.0	143.4	25.4	10.7	2.7	3.9	7.8	2.5	3.1	33.4
KDML105	127	13.8	2.91	102.6	11.8	139.0	22.0	11.1	2.6	4.3	7.8	2.2	3.6	29.4

DH = Days to heading; PN = Panicle number; GP = grain weight per panicle; NP = Number of grain per panicle; NU = number of unfilled grain; PH = Plant height; GY = Grain yield per plant; GL = Grain length; GB = Grain breadth; GL/GB = Grain length/grain breadth; MB = Milled rice kernel breadth; ML = Milled rice kernel length; ML/MB = Milled rice kernel breadth/Milled rice kernel length; GW = 1000-grain weight

Table 20 Correlation coefficients between panicle number (PN), plant height (PH), grain yield per plant (GY), grain weight per panicle (GP), number of grain per panicle (NP), flowering date (FD), 1000-grain weight (GW), grain breadth (GB) grain length (GL), grain length/grain breadth (GL/GB), milled rice kernel breadth (MB) and milled rice kernel length (ML) and milled rice kernel breadth/milled rice kernel length (ML/MB) of fifty selected introgression lines.

Traits	PN	PH	GY	GP	NP	NU	FD	GW	GB	GL	GL/GB	MB	ML
PH	0.54***												
GY	0.37**	0.42**											
GP	0.40**	0.41**	0.31*										
NP	0.42**	0.40**	0.40**	0.88***									
NU	0.20	0.13	0.05	0.13	0.48***								
FD	-0.31*	-0.34*	-0.37**	-0.33*	-0.39**	-0.04							
GW	0.07	0.04	-0.02	0.13	-0.05	0.09	0.13						
GB	0.23	0.16	-0.04	0.30*	0.06	0.25	0.13	0.62***					
GL	0.06	-0.07	0.08	-0.02	0.05	-0.04	-0.18	-0.29*	-0.20				
GB/GL	-0.18	-0.18	0.05	0.27	-0.04	-0.23	-0.17	-0.64***	-0.95***	0.51***			
MB	0.22	0.13	-0.05	0.25	0.04	0.29*	0.21	0.63***	0.93***	-0.35*	-0.94***		
ML	-0.01	-0.12	0.08	-0.02	0.03	-0.11	-0.27	-0.25	-0.15	0.86***	0.42**	-0.29*	
ML/MB	-0.19	-0.16	0.07	-0.21	-0.02	-0.27	-0.26	-0.61***	-0.81***	0.63***	0.93***	-0.92***	0.63***

Significant differences at ***P < 0.001, **0.01, *0.05, respectively

6.3 Grain quality traits of ILs

The quality traits of fifty selected lines were measured using seeds harvested from Ubon Ratchathani in the wet season of 2007. Almost all of the selected ILs was found to meet the KDML105 grain quality standards (Table 21). The distribution of grain and eating quality of the ILs are shown in Figure 32. The appearance character of milled rice and grain shape were measured and compared among the recipient parent and ILs. The milled rice kernel length of the selections was ranged from 6.9 to 7.9 mm. The average milled kernel of the ILs was found 4.2% shorter than that of KDML105 (7.8 mm). The average ML/MB ratio of the selections was 8.0% lower than that of KDML105 (3.55). Only three selected lines were higher than KDML105. The chalkiness of the ILs was dramatically less than that of the donor Rathu Heenati (46.88%). The percentage of chalky occurrence in rice grains of selected lines was ranged from 0.35-1.87%. Percentage chalkiness of five selected lines was observed lower than KDML105.

Grains of the fifty selected BC_3F_{4-6} lines were subjected to cooking and eating quality analysis including AC, GT, GC, and fragrance (FR). The cooking and eating quality of some selected lines are summarized in Table 6. Almost all of the selections were found to meet the KDML105 grain quality standards and had a desirable intermediate AC of 14.19-16.06% similar to those of KDML105 (15.28%). The average AC of the selected lines was 15.01%, which was approximately 1.6% lower than in KDML105. The gelatinization temperature of a grain was measured by the alkaline spreading value (ASV). The selections had the similar ASV score of 6.9–7.0 as that of KDML105 (0.7). GC was measured by the length of the gel. The average length of the gels of the selected lines was 81.2 mm, slightly higher than KDML105 (75 mm), while the average length of the gel of the donor was only 20 mm. Compared to donor, all the selections showed a decreased AC accompanied with an increase in GT and GC. Aroma is one of the most important characters of KDML105. All selected ILs were aromatic with score of 1-2. Despite of few variations of aromatic scent observed among the selections, some of ILs had similar aromatic scent as the original aroma of KDML105.

Table 21 The measurements of the grain quality traits of some selected introgression lines.

Designation	AC	CK	GT	GC	SC
UBN03078-80-354-20	15.55	0.35	7.0	52.2	2
UBN03078-101-342-9	16.06	1.13	7.0	65.0	2
UBN03078-101-342-11	15.39	1.05	6.9	60.0	1
UBN03078-101-342-14	14.95	1.01	7.0	115.0	1
UBN03078-101-342-4-24	15.61	1.40	6.9	100.0	2
UBN03078-101-342-4-32	14.58	1.37	7.0	105.0	1
UBN03078-101-342-4-96	14.36	0.59	6.9	60.0	2
UBN03078-101-342-4-106	14.96	1.09	7.0	120.0	1
UBN03078-101-342-4-111	14.63	0.76	6.9	120.0	1
UBN03078-101-342-4-114	14.39	0.47	7.0	80.0	2
UBN03078-101-342-4-126	15.25	1.02	7.0	67.5	1
UBN03078-101-342-4-143	14.19	1.35	7.0	77.5	2
UBN03078-101-342-4-144	14.56	0.61	7.0	65.0	2
UBN03078-101-342-4-148	15.73	0.90	7.0	72.5	1
UBN03078-101-342-4-158	15.58	0.97	7.0	120.0	1
UBN03078-101-342-6-49	14.43	1.28	7.0	46.5	1
UBN03078-101-342-6-56	15.39	1.87	7.0	60.0	1
UBN03078-101-342-6-58	15.22	1.29	7.0	70.0	1
KDML105	15.28	0.83	7.0	75.0	2

AC = Amylose content (%); CK = Chalkiness of endosperm (0 = none; 1 = less than 10%; 5 = 11-20%; 9 = more than 20%); GT = Gelatinization temperature (1-2, high and 6-7 low); GC = Gel consistency (80-100 = very soft; 61-80 = soft; 41-60 = medium; 36-40 = hard; less than 35 = very hard); SC = Scent (0=unscented; 1 = lightly scented; 2 = scented)

DISCUSSION

We report the localization of the major resistance gene *Bph3* to the short arm of rice chromosome 6 based on analyses utilizing SSR markers. The tightly linked SSR markers identified in this study should clarify role of the *Bph3* locus carried by the resistant donors PTB33 and Rathu Heenati. Starting from the flanking markers, we were able to locate the gene to a 190 kb segment of genomic DNA. The fragment contains twenty-two putative genes, which encode fourteen proteins (seven hypothetical and seven expressed) of unknown function, an NBS-LRR disease resistance protein, two pentatricopeptides, two oligopeptide transporters, a zinc finger protein, a transcriptional co-regulator protein, and a protein kinase protein. This result should be helpful for cloning the *Bph3* gene. The closely linked molecular markers found in this study should be also useful in maker-assisted breeding programs aimed at developing improved BPH resistance cultivars.

The resistance gene *Bph3* has been used extensively in rice breeding programs in Asia since 1980 (Khush, 1984). This is also the case in Thai breeding programs, and *Bph3* is still effective against BPH populations found in Thailand. Based on cluster analysis, we found that the field BPH populations demonstrated varying levels of virulence and, consequently, were able to classify four different groups of BPH populations based on a similarity relationship of more than 0.88. This indicated that at least four different biotypes of BPH are present in Thailand.

PTB33 and Rathu Heenati had been reported earlier to carry *Bph3* and confer a high level of resistance against BPH (Angeles *et al*., 1986; Kabir and Khush, 1988; Khush, 1984; Khush *et al*., 1985; Li *et al*., 2002; Nemoto *et al*., 1989; Sidhu and Kush, 1979). Our study also found that PTB33 and Rathu Heenati showed resistance against all of the BPH populations tested, indicating the broad-spectrum of resistance carried by these cultivars against the BPH biotypes found in Thailand. Mechanism of a broad-spectrum of resistance is considered as an importance in a breeding program. The durability of resistance genes is also important as a longer durability will slow down the appearance of virulent biotypes (Heinrichs, 1986). Based on our

unpublished data from selection experiments of the BPH fecundity, PTB33 and Rathu Heenati retain a resistance to the BPH at least 10 generations of the insects (Jairin, 2005).

Because SSR markers are co-dominant, multiallelic and available at a high density in the rice genome, which is approximately one SSR every 157 kb (McCouch *et al*., 2002), these markers can be used to scan and identify the target regions associated with interested traits. To find molecular markers tightly linked to the *Bph3* locus, we used SSR markers surrounding the target regions that had been identified in previous studies. Applying this approach, we were able to detect markers associated with the major resistance gene. Based on the SSR and linkage analysis, we assigned the major resistance gene *Bph3* to the short arm of rice chromosome 6. It should be noted that *Bph3* from PTB33 and Rathu Heenati was a major resistance gene against BPH populations, which was used in this study, since the tightly linked marker RM589 and RM586 could explain 59.8 and 57.4% of the phenotypic variance, respectively (Table 16). However, according to the frequency distribution of the damage rating of the two backcross populations, BPH resistance in PTB33 and Rathu Heenati is likely to be controlled by a major and other minor resistance gene.

Although *Bph3* has been reported to be present on chromosomes 4 and 10 (Ikeda and Kaneda, 1981; Sun *et al*., 2005; Yan *et al*., 2002), our study did not detect any selected SSR markers on those chromosomes that were associated with the R and S groups. The resistance gene *Bph3* on rice chromosome 4 assigned by Yan *et al*., (2002) was derived from *Oryza officinalis*; however, this resistance gene was later designated as a new resistance gene, *Bph15* (Yang *et al*., 2004). The resistance gene *Bph12* from *O. latifolia* was also reported in the same region on the short arm of chromosome 4 (Yang *et al*., 2002). Recently, a new major resistance gene, tentatively designated as *Bph17*, derived from Rathu Heenati has been reported in the same region of *Bph15* and *Bph12* (Sun *et al*., 2005). Our study did not detect any significant *Bph17*-tightly linked markers (RM8213, RM6487, RM401) on chromosome 4 as reported earlier by Sun *et al*., (2005). The result obtained in our study showed that the

major resistance gene carried by Rathu Heenati was tightly linked to *bph4* on chromosome 6, which has been reported earlier by Kawaguchi *et al*. (2001).

Several studies have detected major BPH resistance genes using different BPH biotypes. BPH biotypes 1 and 2 were used previously to identify *Bph3* and determine the allelic relationship between *Bph3* and *bph4* (Angeles *et al*., 1986; Heinrichs *et al*., 1985; Ikeda and Kaneda, 1981; Lakshminarayana and Khush, 1977; Sidhu and Khush, 1978). Sun *et al*. (2005) also used a mixture of biotypes 1 and 2 to identify the major resistance gene in Rathu Heenati on chromosome 4; however, the resistance gene *bph4* was assigned to chromosome 6 (Kawaguchi *et al*., 2001). We suggest that the resistance gene detected by Sun *et al*. (2005) must be a new BPH resistance gene. The present study and that of Sun *et al*. (2005) detected two different major resistance genes in Rathu Heenati, perhaps because different germplasm sources of Rathu Heenati were used. These observations may provide insights into some of the issues concerning germplasm sources of BPH resistance in mapping studies. *Bph3* was first identified in Rathu Heenati accession no. 11730 against BPH biotypes 1, 2, 3, 4 and some biotypes in Thailand (Angeles *et al*. 1986; Heinrichs *et al*. 1985; Jairin *et al*. 2005; Lakshminarayana and Khush 1977). The same accession number of Rathu Heenati provided by IRRI was also used to determine the *Bph3* locus in the present study.

PTB33 has been reported to carry two major resistance genes, *bph2* and *Bph3*. Since the BPH population used in this study was completely adapted to the *bph2* gene, we could only detect the *Bph3* locus associated with the R and S individuals. Another resistant cultivar, ASD7, carrying *bph2* was also susceptible to the BPH population collected from Ubon Ratchathani. To investigate this, we employed the PCR-based STS marker, KAM4, which showed complete co-segregation with *bph2* on rice chromosome 12 (Murai *et al*., 2001), to survey ASD7 and R and S groups of BC_1F_2 derived from the PTB33×RD6 cross. Since the KAM4 is a dominant STS marker, we found that the amplified fragment appeared only in PTB33 and ASD7 but did not appear in RD6. The amplified fragments were detected in progenies from both R and S individuals (Figure 20). This result probably indicates that the *bph2* gene in

PTB33 was not effective against the BPH population used in this study. However, the interaction between two major resistance genes in PTB33 would require further investigation.

Kawaguchi *et al*. (2001) reported that the resistance gene *bph4* from Babawee was located near restriction RFLP marker C76A on the short arm of chromosome 6. According to the standard linkage map of SSR constructed by McCouch *et al*. (2002), the tightly linked marker C76A is located in the same position as the RM190 locus. Our study indicated that the distance between RM190 and RM589, the tightly linked marker to *Bph3* locus, is about 2.4 cM. Therefore, this study clarifies the location of the broad-spectrum resistance gene *Bph3* and confirms the genetic analysis by classical genetic approach of Ikeda and Kaneda (1981) and Sidhu and Khush (1979) that *Bph3* in Rathu Heenati and PTB33 is tightly linked to the *bph4* in Babawee. These results also confirm those of Kawaguchi *et al*. (2001) using two backcross mapping populations that *Bph3* and *bph4* are localized on the short arm of rice chromosome 6.

BPH resistance in rice cultivars carrying *Bph3* was reported to govern an antixenotic reaction to BPH (Murai *et al*., 2001). Rathu Heenati has no repellent chemical against planthoppers and only has common volatiles as released by susceptible cultivars. The feeding inhibition of this cultivar occurred when the insect started to ingest phloem sap (Liu *et al*., 1994; Sexena *et al*., 1985). There were several studies confirmed that the mechanism of BPH resistance in Ratthu Heenati is associated with the phloem (Kimmins, 1989; Padgham *et al*., 1989; Padgham and Woodhead, 1988; Stevenson *et al*., 1996). In the present study, Rathu Heenati showed high resistance to BPH at the vegetative stage; only a few numbers of BPH could survive on the resistant plants. The surviving insects had light body weight, slow development and low fecundity (data not shown). On the other hand, Rathu Heenati was susceptible to BPH at the flowering and grain filling stages. BPH could feed and grow well on panicle necks and panicles of the resistant plants. This phenomenon may affect the expression of BPH resistance gene in Rathu Heenati. Further studies are needed to clarify this event especially the chemical analysis of the phloem sap from

resistant and susceptible isogenic lines (Chen *et al*., 1997). Comparison of phloem sap components by using a chemically defined diet (Fu *et al*., 2001) will also provide information to clarify the phenomenon.

The mechanism of plant resistance to phloem sap-feeding insects has been reported to involve the balance of the amino acid composition of the phloem sap (Fu *et al*., 2001; Douglas, 1993). Variation of phloem amino acid composition has been implicated in the nitrogen quality of the phloem sap for phloem feeders (Douglas, 1993; Fu *et al*., 2001; Sandstrom, 2000). It plays a major role in the performance and fitness of insects (Karley *et al*., 2002). The susceptibility of Rathu Heenati at the flowering stage observed in this study may probably involve in the nutritional quality of the phloem sap. In the rice panicles, the total nitrogen arises from remobilization of glutamine synthetase through the phloem from senescing organs (Hayakawa *et al*., 1993; Mae and Ohira, 1981; Weibull, 1988). The major forms of reduced nitrogen in the phloem sap of rice plants are glutamine and asparagine (Tabuchi *et al*., 2007). Application of a nitrogen fertilizer can dramatically increase the amount of total nitrogen and free amino acids available in the phloem sap (Yamaya *et al*., 2002), especially glutamine and asparagine (Hayashi *et al*., 1993; Tobin and Yamaya, 2001). Asparagine in the phloem sap was identified as a sucking stimulator for BPH. Difference of asparagines content in phloem sap can also be related to the host selection of BPH (Shigematsu *et al*., 1982). Therefore, the remobilization of nitrogen in rice plants can increase the total free amino acids in the phloem sap, which may affect the BPH resistance in rice plants and insect performance. Furthermore, apigenin-*C*-glycoside in phloem sap from Rathu Heenati has been reported to responsible for rice resistance to BPH (Grayer *et al*., 1994; Stevenson *et al*., 1996). This alochemical might be one of the reasons that make Rathu Heenati resistance to BPH. Currently, three possible hypotheses can explain how BPH resistance gene is involved in the phenomena: (i) a resistance gene(s) may be poorly expressed in the upper internodes of heading rice plants, (ii) the amount of the reduced nitrogen forms or nitrogenous compounds in the phloem sap may affect the expression of the BPH resistance gene, and (iii) a resistance gene(s) may involve the phloem nitrogen quality, which affects the activities of symbiotic micro-organisms in BPH. However

further studies are needed to investigate the mechanism of BPH resistance in Rathu Heenati and should elucidate which gene present in the 190 kb segment confers resistance against BPH when introduced into BPH-susceptible plants.

When the unexpected linkage drag occurred, it will endeavor to achieve a breeding goal using conventional approaches in particular when the target gene is linked with an unfavorable dominant gene. However, the goal can be obtained using molecular markers as a tool for selection. The main objective of this study was to combine KDML105 grain quality traits with BPH resistance, and it was successfully introgressed the BPH resistance gene from Rathu Heenati into KDML105 by MAS in three generations of backcrossing and dissected the linkage drag between the introgressed *Bph3* and Wx^a allele, which mainly responsible for an unfavorable characteristic of the grain quality traits, from the donor cultivar.

The quality of the rice grain is one of the primary breeding objectives of rice improvement programs. There is a strong emphasis in Thailand on increasing the quality of rice cultivars with biotic and/or abiotic tolerance. Consequently, KDML105 has been widely used as a base for the grain quality traits nationwide. KDML105 is mostly growing under rainfed lowland areas in the Northeast, the largest area for producing the best quality rice in Thailand. Almost all cultivated rice growing in these areas are susceptible to BPH. Although BPH has been considered as a minor insect pest in the rainfed areas for decades, in the recent time the BPH outbreaks have been frequently occurred in the areas. Breeding new BPH resistant cultivars with high grain quality and wide adaptability under rainfed lowland areas are, therefore, becoming necessary.

Rathu Heenati was found very effective against BPH populations in Thailand and in South East Asia. This cultivar has been considered to confer a broad spectrum and durability of resistance against BPH. However, Rathu Heenati having major disadvantages of poor grain quality as well as its appearance because of a high AC, a hard GC, a low GT, a chalky endosperm, no fragrance together with a prominent awning and a red pericarp. It had been determined that AC, GC and GT, are controlled by the *Waxy* region on chromosome 6 (He *et al*., 2006; Wang *et al*., 2007).

Unfortunately, we have found that the major BPH resistance gene in Rathu Heenati was linked to the Wx^a allele. According to the previous study, a co-segregated SSR marker with major BPH resistance gene in Rathu Heenati was located near the Wx^a approximately 380 kb based on the genome sequence of Nipponbare (http://www.gramene.org/). This is a case of linkage drag that usually occurs in breeding programs. This might be one of the reasons that we could not develop BPH resistant promising line with good cooking and eating quality using *Bph3* by conventional approaches. For example, BPH resistant cultivars or promising lines carrying *Bph3* (i.e. IR72, IR56, IR60, IR13540-56-3-2-1 and PSL2), which have been developed in various institutes, are having high amylose content in the endosperm.

To reduce the linkage drag, MAS integrate with phenotypic selection was used to select rice lines carrying recombinants heterozygous in the target region from a total of 2,343 BC_3F_2 derived from the cross between Rathu Heenati and KDML105. The number of the progenies was reduced by screening for BPH resistance before applying MAS. This is a successful example of an integrated approach to plant breeding. A small chromosome segment containing a favorable gene from the donor cultivar was introduced into elite lines. The improved lines contained a fragment less than 190 kb of the *Bph3* region from the donor parent (Jairin *et al*., 2007b). The ILs showed the same broad spectrum resistance against BPH populations in Thailand as the donor cultivar Rathu Heenati. The results have confirmed that the major broad spectrum resistance gene from Rathu Heenati has transferred to the elite lines. However, the levels of resistance of some ILs were not as high as that of Rathu Heenati. Some unidentified minor resistance genes might be lost during backcrossing. Further investigations are required for identification of the locations and effects of the other minor resistance genes.

The essential agronomic characteristics of the ILs developed in this study were almost same as those of KDML105. No significant alteration was observed in agronomic characters of the improved ILs compared to KDML105 except for the grain yield. The increase in grain yield of the ILs was probably because of increase in number of panicles per plant, number of grains per panicle and plant height but not because of 1000-grain weight.

Molecular marker-assisted selection is proved as an effective approach to improve good cooking and eating quality of the milled rice. Effectiveness of MAS for quality traits was successful in the previous studies and several advanced breeding lines/varieties have been developed (Joseph *et al*., 2004; Liu *et al*., 2005; 2006; Toojinda *et al*., 2005; Zhang *et al*., 2005; Zhang, 2007; Zhou *et al*., 2003). In this study, BPH resistant lines were successfully improved with maintaining high grain quality using MAS approach. The results revealed that the *Wx* region on chromosome 6 have major effects on the rice grain quality. The improved ILs lines can be directly developed into varieties, which will have an impact on the yield stability in KDML105-producing areas. In addition, the ILs can be served either as an immediate sources of broad spectrum and durable BPH resistance to improve good grain quality in breeding programs or as a material to combine several target genes by crossing and MAS.

CONCLUSION AND RECOMMENDATION

The main discovery of this research was the identification of the major resistance gene *Bph3* on the short arm of rice chromosome 6 near the *Waxy* locus. The tightly linked SSR markers identified in this study had clarified the role of the *Bph3* locus carried by the resistant donors PTB33 and Rathu Heenati. Starting from the flanking markers, we were able to locate the gene to a 190 kb segment of genomic DNA. The fragment contains twenty-two putative genes, which encode fourteen proteins. This result should be helpful for cloning the *Bph3* gene. The closely linked molecular markers found in this study should be also useful in maker-assisted breeding programs aimed to develop BPH resistance varieties.

Among the main objectives of rice improvement programs in Thailand, quality traits of grain have been considered as the first priority. Therefore, KDML105 has been widely used as a based for grain quality traits nationwide. KDML105 is mostly growing under rainfed lowland areas in the Northeast, which is the largest area for producing quality rice in Thailand. Almost all rice cultivars growing in the areas are susceptible to BPH. Although BPH has been considered as a minor insect pest of the rainfed areas for decades, in the recent time BPH outbreaks have been frequently occurred. Breeding new BPH resistant cultivar with good cooking and eating quality is, therefore, become necessary.

One of the main objectives of this study was to combine KDML105 quality traits with BPH resistance from the donor, Rathu Heenati. We successfully introgressed the BPH resistance gene into KDML105 by MAS and phenotypic selection in three generations of backcrossing and dissected the linkage drag between the introgressed gene and *Waxy* allele from the donor, which cause unflavored characteristics of the quality traits. All selected ILs showed broad spectrum resistance against BPH populations in Thailand. The results on agronomic performance showed that most of morphological traits of ILs, including flowering date, appearance grain quality and plant type, were same as those of KDML105. All selected ILs were found to meet the KDML105 grain quality standard. ILs developed in this research can be

directly developed into a BPH resistance variety or can serve as immediate sources of BPH resistance in KDML105 breeding programs.

Although we had successful to introgress a BPH resistance gene *Bph3* from Rathu Heenati into KDML105, all ILs and rice cultivars carry *Bph3* showed susceptible to BPH at reproductive stage. To use these materials or BPH resistance lines as a BPH resistance donor in breeding programs, we should concern about this observable fact. The further study is required to clarify this phenomenon to understand the mechanism of plant resistance to BPH at vegetative and reproductive stages. This might be deduced that screening of BPH resistance at only seedling stage of rice plant is inadequate to get a superior BPH resistance donor.

LITERATURE CITED

Alam, S.N. and M.B. Cohen. 1998. Detection and analysis of QTLs for resistance to the brown planthopper, *Nilaparvata lugens*, in a doubled-haploid rice population. **Theor. Appl. Genet.** 97: 1370-1379.

Amarawathi, Y., R. Singh, A.K. Singh, V.P. Singh, T. Mohapatra, T.R. Sharma, and N.K. Singh. 2008. Mapping of quantitative trait loci for basmati quality traits in rice (*Oryza sativa* L.). **Mol. Breeding.** 21:49-65.

Angeles, E.R., G.S. Khush and E.A. Heinrichs. 1986. Inheritance of resistance to planthoppers and leafhopper in rice. pp. 537-549. *In* International Rice Research Institute, ed. **Rice Genetics.** Los Baños, Philippines.

Athwal, D.S., M.D. Pathak, E.H. Bacalangco and C.D. Pura. 1971. Genetics of resistance to brown planthopper and green leafhopper in *Oryza sativa* L. **Crop Sci.** 11: 747-750.

Ayoade, O., S. Morooka and S. Tojo. 1996. Metamorphosis and wing formation in the brown planthopper, *Nilaparvata lugens*, after topical application of precocene II. **Arch. Insect Biochem. Physio.** 32: 485-491.

Backus, E.A., M.S. Serrano and C.M. Ranger. 2005. Mechanisms of hopperburn: an overview of insect taxonomy, behavior, and physiology. **Ann. Rev. Entomol.** 50: 125-151.

Baumann, P., C. Lai, D. Roubakhsh, N.A. Moran and M.A. Clark. 1995. Genetics, physiology, and evolutionary relationships of the genus *Buchnera* - intracellular symbionts of aphids. **Ann. Rev. Microb.** 49: 55-94.

Bertuso, A.G. and S. Tojo. 2002. The nature and titer of juvenile hormone in the brown planthopper, *Nilaparvata lugens* (Homoptera: Delphacidae) in relation

to wing morphogenesis and oocyte development. **Appl. Entomol. Zool.** 37: 117-125.

Bing, L., D. Hongxia, Z. Maoxin, X. Du and W. Jingshu. 2007. Potential resistance of tricin in rice against brown planthopper *Nilaparvata lugens* (Stål). **Acta Ecol. Sinica.** 27: 1300-1307.

Chen, D.H. and P.C. Ronald. 1999. A rapid DNA minipreparation method suitable for AFLP and others PCR applications. **Plant Mol. Biol. Rep.** 17: 53-57.

Chen, J.Q., Y. Rahbé, B. Delobel, N. Sauvion, J. Guillard and G. Febvay. 1997. Melon resistance to the aphid *Aphis gossypii*: behavioural analysis and chemical correlations with nitrogenous compounds. **Entomol. Exp. Appl.** 85: 33-44.

Chen, J.W., L. Wang, X.F. Pang and Q.H. Pan. 2006. Genetic analysis and fine mapping of a rice brown planthopper (*Nilaparvata lugens* Stål) resistance gene *bph19*(t). **Mol. Gen. Genomics.** 275: 321-329.

Cheng, D.J. and R.F. Hou. 2001. Histological observations on transovarial transmission of a yeast-like symbiote in *Nilaparvata lugens* Stål (Homoptera, Delphacidae). **Tissue and Cell**. 33: 273-279.

Denno, R.F., J. Cheng, G.K. Roderick and T.J. Perfect. 1995. Density related effects on the components of fitness and population dynamics of planthoppers, pp. 257-281. *In* R.F. Denno and T.J. Perfect, eds. **Planthoppers: Their Ecology and Management.** Chapman and Hall, New York.

Denno, R.F. and G.K. Roderick. 1990. Population biology of planthoppers. **Ann. Rev. Entomol.** 35: 489-520.

Douglas, A.E. 1989. Mycetocyte symbiosis in insects. **Biol. Rev.** 69: 409-434.

Douglas, A.E. 1993. The nutritional quality of phloem sap utilised by natural aphid populations. **Ecol. Entomol.** 18: 31-38.

Douglas, A.E. 1998. Nutritional interactions in insect-microbial symbioses: aphids and their symbiotic bacteria *Buchnera*. **Ann. Rev. Entomol.** 43: 17-37.

Dyck, V.A. and B. Thomus. 1979. The brown planthopper problem, pp. 3-17. *In* International Rice Research Institute, ed. **Brown Planthopper: Threat to Rice Production in Asia.** IRRI, Los Baños, Philippines.

Feakin, S.D. 1974. **Pest Control in Rice.** PANS Manual No.3 Center for Oversea Pest Research, London.

Fu, Q., Z. Zhang, C. Hu, F. Lai and Z. Sun. 2001. A chemically defined diet enables continuous rearing of the brown planthopper, *Nilaparvata lugens* (Stål) (Homoptera: Delphacidae). **Appl. Entomol. Zool.** 36: 111-116.

Grayer, R.J., E.M. Kimmins, E.C. Stevenson, J.B. Harbome and H.N.E Wijayagunesekera, 1994. Phenolics in rice phloem sap as sucking deterrents to the brown planthopper, (*Nilaparvata lugens*). **Acta Hort.** 381: 391-394.

Hattori, M. and K. Sogawa. 2002. Oviposition behavior of the rice brown planthopper, *Nilaparvata lugens* (Stål), and its electronic monitoring. **J. Insect Behavior.** 15: 283-293.

Hao, P., C. Liu, Y. Wang, R. Chen, M. Tang, B. Du, L. Zhu and G. He. 2008. Herbivore-induced callose deposition on the sieve plates of rice: an important mechanism for host resistance. **Plant Physiol.** 146: 1810-1820.

Hayakawa, T., T. Yamaya, T. Mae and K. Ojima. 1993. Changes in the content of two glutamate synthase proteins in spikelets of rice (*Oryza sativa*) plants during ripening. **Plant Physiol**. 101: 1257-1262.

Hayashi, H., S. Nakamura, Y. Ishiwatari, S. Mori and M. Chino. 1993. Changes in the amino acid composition and protein modification in phloem sap of rice. **Plant Soil**. 155/156: 171-174.

He, Y., Y. Han, L. Jiang, C. Xu, J. Lu, and M. Xu. 2006. Functional analysis of starch-synthesis genes in determining rice eating and cooking qualities. **Mol. Breeding.** 18: 277-290.

Heinrichs, E.A. 1979. Control of leafhopper and planthopper vectors of rice viruses, pp. 529-558. *In* K. Moramorosch and K.F. Arris, eds. **Leafhopper Vectors and Planthopper Disease Agents.** Academic Press, New York.

Heinrichs, E.A. 1986. Perspectives and directions for the continued development of insect resistant rice varieties. **Agric. Ecosyst. Environ.** 18: 9-36.

Heinrichs, E.A., F.G. Medrano and H.R. Rapusas. 1985. **Genetic Evaluation for Insect Resistance in Rice.** IRRI, Los Baños, Philippines.

Hiyashi, H. and M. Chino. 1990. Chemical composition of phloem sap from the upper most internode of the rice plant. **Plant Cell Physiol.** 31: 247-251.

Hirabayashi, H., E.R. Angeles, R. Kaji, T. Ogawa, D.S. Brar and G.S. Khush. 1998. Identification of the brown planthopper resistance gene derived from *O. officinalis* using molecular markers in rice. **Breeding Sci.** 48 (Suppl 1): 82

Hirabayashi, H. and T. Ogawa T. 1995. RFLP mapping of *Bph-1* (Brown planthopper resistance gene) in rice. **Jpn. J. Breeding.** 45: 369-371.

Hollander, J. den and P.K. Pathak. 1981. The genetics of the 'biotypes' of the rice brown planthopper, *Nilaparvata lugens*. **Entomol. Exp. Appl.** 29: 76-86.

Hongoh, Y. and H. Ishikawa. 1997. Uric acid as a nitrogen resource for the brown planthopper, *Nilaparvata lugens*: studies with synthetic diets and aposymbiotic insects. **Zool. Sci.** 14: 581-586.

Hospital, F. 2001. Size of donor chromosome segments around introgressed loci and reduction of linkage drag in marker-assisted backcross programs. **Genetics.** 158: 1363-1379.

Huang, N., A. Parco, T. Mew, G. Magpantay, S. McCouch, E. Guiderdoni, J. Xu, P. Subudhi, E.R. Angeles and G.S. Khush. 1997. RFLP mapping of isozymes, RAPD and QTLs for grain shape, brown planthopper resistance in a doubled haploid rice population. **Mol. Breeding.** 3: 105-113.

Huang, Z., L. Shu, X. Li and Q. Zhang. 2001. Identification and mapping of two brown planthopper resistance genes in rice. **Theor. Appl. Genet.** 102: 929-934.

Ikeda, R. 1985. Studies on the inheritance of resistance to the rice brown planthopper (*Nilaparvata lugens* Stål) and the breeding of resistance rice cultivars. **Bull. Nat. Agri. Res. Cent.** 3: 1-54.

Ikeda, R. and C. Kaneda. 1981. Genetic analysis of resistance to brown planthopper, *Nilaparvata lugens* Stål, in rice. **Jpn. J. Breeding.** 31: 279-285.

Ikeda, R. and C. Kaneda. 1983. Trisomic analyses of the gene *Bph-1* for resistance to the brown planthopper, *Nilaparvata lugens* Stål. **J. Breeding.** 33: 40-44.

International Rice Research Institute. 1996. **Standard Evaluation System for Rice.** Los Baños, Philippines.

Ishii, T., D.S. Brar, D.S. Multani and G.S. Khush. 1994. Molecular tagging of genes for brown planthopper resistance and earliness introgressed from *Oryza australiensis* into cultivated rice, *O. sativa*. **Genome.** 37: 217-221.

Itoh, K., H. Ozaki, K. Okada, H. Hori, Y. Takeda and T. Mitsui. 2003. Introduction of *Wx* transgene into rice *wx* mutants leads to both high- and low-amylose rice. **Plant Cell Physiol.** 44: 473-480.

Iwanaga, K., S. Tojo and T. Nagata. 1985. Immigration of the brown planthopper, *Nilaparvata lugens*, exhibiting various responses to density in relation to wing morphism. **Entomol. Exp. Appl.** 38: 101-108.

Jairin, J. 2005. **Identification of Markers Linked to Brown Planthopper Resistance Genes in Rice (*Oryza sativa* L.).** M.S. thesis, Kasetsart University.

Jairin, J., K. Phengrat, S. Teangdeerith, A. Vanavichit and T. Toojinda. 2007a. Mapping of a broad-spectrum brown planthopper resistance gene, *Bph3*, on rice chromosome 6. **Mol. Breeding.** 19: 35-44.

Jairin, J., N. Kojima and T. Nagata. 2005a. Insecticide resistance of the green rice leafhopper, *Nephotettix cincticeps*, to the systemic insecticides used for seedling-box application. **Sci. Asia.** 31: 151-158.

Jairin, J., S. Teangdeerith, P. Leelagud, K. Phengrat, A. Vanavichit and T. Toojinda. 2007b. Detection of brown planthopper resistance genes from different rice mapping populations in the same genomic location. **Sci. Asia.** 33: 347-352.

Jairin, J., S. Teangdeerith, P. Leelagud, K. Phengrat, A. Vanavichit and T. Toojinda. 2007c. Physical mapping of *Bph3*, a brown planthopper resistance locus in rice. **Mj. Int. J. Sci. Tech.** 1: 166-177.

Jairin, J., T. Toojinda, S. Tragoonrung, S. Tayapat and A. Vanavichit. 2005b. Multiple genes determining brown planthopper (*Nilaparvata lugens* Stål) resistance in backcross introgressed lines of Thai jasmine rice 'KDML105'. **Sci. Asia.** 31: 129-135.

Jena, K.K., I.C. Pasalu, Y.K. Rao, Y. Varalaxmi, K. Krishnaiah, G.S. Khush and G. Kochert. 2003. Molecular tagging of a gene for resistance to brown planthopper in rice (*Oryza sativa* L.). **Euphytica.** 129: 81-88.

Jena, K.K., J.U. Jeung, J.H. Lee, H.C. Choi and D.S. Brar. 2006. High-resolution mapping of a new brown planthopper (BPH) resistance gene, *Bph18*(t), and marker-assisted selection for BPH resistance in rice (*Oryza sativa* L.). **Theor. Appl. Genet.** 112: 288-297.

Jeon, Y.H., S.N. Ahn, H.C. Choi, T.R. Hahn and H.P. Moon. 1999. Identification of a RAPD marker linked to a brown planthopper resistance gene in rice. **Euphytica**. 107: 23-28.

Joseph, M., S. Gopalakrishnan, R.K. Sharma, V.P. Singh, A.K. Singh, N.K. Singh and T. Mohapatra. 2004. Combining bacterial blight resistance and Basmati quality characteristics by phenotypic and molecular marker-assisted selection in rice. **Mol. Breeding.** 00: 1-11.

Juliano, B.O. 1971. A simplified assay for milled rice amylose. **Cereal Sci. Today.** 16: 334-338.

Kabir, M.A. and G.S. Khush. 1988. Genetic analysis of resistance to brown planthopper in rice (*O. sativa* L.). **Plant Breeding**. 100: 54-58.

Kawaguchi, M., K. Mulata, T. Ishii, S. Takumi, N. Mori and C. Nakamura. 2001. Assigment of a brown planthopper (*Nilaparvata lugens* Stål) resistance gene *bph4* to the rice chromosome 6. **Breeding. Sci.** 51: 13-18.

Karley, A.J. and A.E. Douglas, W.E. Parker. 2002. Amino acid composition and nutritional quality of potato leaf phloem sap for aphids. **J. Exp. Biol.** 205: 3009-3018.

Kimmins, E.M. 1989. Electrical penetration graphs from *Nilaparvata lugens* on resistant and susceptible rice varieties. **Entomol. Exp. Appl.** 50: 69-79.

Kisimoto, R. 1965. Studies on polymorphism and its role in the population growth of the brown planthopper, *Nilaparvata lugens* Stål. **Rev. Appl. Entomol.** 55: 381-397.

Khush, G.S. 1979. Genetics of resistant and breeding for resistance to the brown planthopper, pp. 321-332. *In* International Rice Research Institute, ed. **Brown Planthopper: Threat to Rice Production in Asia.** IRRI, Los Baños, Philippines.

Khush, G.S. 1984. Breeding rice for resistance to insects. **Protect. Eco.** 7: 147-165.

Khush, G.S., A.N.M. Rezaul, E. Karim and R. Angeles. 1985. Genetics of resistance of rice cultivar ARC10550 to Bangladesh brown planthopper in rice (*O. sativa* L.). **Plant Breeding.** 100: 54-58.

Khush, G.S. 1992. Selecting rice for simply inherited resistance, pp. 303-340. *In* H.T. Stalker and J.P. Murphy, ed. **Plant Breeding in the 1990s.** CAB International. Wallingford.

Kimmins, F.M. 1989. Electrical penetration graphs from *Nilaparvata lugens* on resistant and susceptible rice varieties. **Entomol. Exp. Appl.** 50: 69-79.

Kisimoto, R. 1973. Leafhoppers and planthoppers, pp. 137-156. *In* A.J. Gibbs, ed. **Viruses and Invertebrates.** Elsevier, New York.

Kosambi, D.D. 1944. The estimation of map distance from recombination values. **Ann. Eugen.** 12: 172-175.

Lakshminarayana, A. and G.S. Khush. 1977. New genes for resistance to the brown planthopper in rice. **Crop Sci.** 17: 96-100.

Lanceras, J.C., Z.L. Huang, O. Naivikul, A. Vanavichit, V. Ruanjaichon and S. Tragoonrung. 2000. Mapping of genes for cooking and eating qualities in Thai Jasmine rice (KDML105). **DNA Res.** 7: 93-101.

Li, R.B., X.Y. Qin, S.M. Wei, F.K. Huang, Q. Li and S.Y. Luo. 2002. Identification and genetics of resistance against brown planthopper in a derivative of wild rice, *Oryza rufipogon* Griff. **J. Genet. Breeding.** 56: 29-36.

Liu, G., R.C. Saxena, R.M. Wilkins. 1994. Behavioral responses of the whitebacked planthopper *Sogatella furcifera* (Homoptera: Delphacidae) on rice plants whose odors have been masked. **J. lnsect Behavior.** 7: 43-353.

Liu, G.Q., H.H. Yan, Q. Fu, Q. Qian, Z.T. Zhang, W.X. Zhai and L.H. Zhu. 2001. Mapping of a new gene for brown planthopper resistance in cultivated rice introgressed from *Oryza eichingeri*. **Chinese Sci. Bull.** 46: 1459-1462.

Liu, Q., H. Yu, X. Chen, X. Cai, S. Tang, Z. Wang and M. Gu1. 2005. Field performance of transgenic *indica* hybrid rice with improved cooking and eating quality by down-regulation of *Wx* gene expression. **Mol. Breeding.** 16: 199-208.

Liu, Q., Q. Li, X. Cai, H. Wang, S. Tang, H. Yu, Z. Wang and M. Gu. 2006. Molecular marker-assisted selection for improved cooking and eating quality of two elite parents of hybrid rice. **Crop Sci.** 46: 2354–2360.

Lu, Z., X. Yu, J. Chen, X. Zheng, H. Zu, J. Zhang and L. Chen. 2004. Dynamics of yeast-like symbiote and its relationship with the virulence of brown planthopper, *Nilaparvata lugens* Stål, to resistant rice varieties. **J. Aisia-Pacipic Entomol.** 7: 317-323.

Mae, T. and K. Ohira. 1981. The remobilization of nitrogen related to leaf growth and senescence in rice plants (*Oryza sativa* L.). **Plant Cell Physiol.** 22: 1067-1074.

McCouch, S.R., X. Chen, O. Panaid, S. Temnykh, Y. Xu, Y.G. Cho, N. Huang, T. Ishii and M. Blair. 1997. Microsatellite marker development, mapping and application in rice genetics and breeding. **Plant Mol. Biol.** 35: 89-99.

McCouch, S.R., L. Teytelman, Y. Xu, K.B. Lobos, K. Clare, M. Walton, B. Fu, R. Maghirang, Z. Li, Y. Xing, Q. Zhang, I. Kono, M. Yano, R. Fjellstrom, G. DeClerck, D. Schneider, S. Cartinhour, D. Ware and L. Stein. 2002. Development and mapping of 2,240 new SSR markers for rice (*Oryza sativa* L.). **DNA Res.** 9: 199-207.

Mei, M., C. Zhuang, R. Wan, J. Wu and G. Kochert. 1996. Genetic analysis and tagging of gene for brown planthopper resistant in *indica* rice, pp. 590-595. *In*: International Rice Research Institute, ed. **Rice Genetics III.** Proceeding of the third international rice genetics symposium, IRRI, Los Baños, Philippines.

Michelmore, R.W., I. Paran, and R.V. Kesseli. 1991. Identification of markers linked to disease-resistance genes by bulked segregant analysis: A rapid method to detect markers in specific genomic regions by using segregating populations. **Proc. Natl. Acad. Sci. USA.** 88: 9828-9832.

Miles, P.W. 1999. Aphid saliva. **Biol. Rev.** 74: 41-85.

Mochida, O. and T. Okada. 1979. Taxonomy and biology of *Nilaparvata lugens* (Hom., Delphacidae), pp. 21-43. *In* International Rice Research Institute, ed. **Brown Planthopper: Threat to Rice Poduction in Asia.** IRRI, Los Baños, Philippines.

Murai, H., Z. Hashimoto, P.N. Sharma, T. Shimizu, K. Murata, S. Takumi, N. Mori, S. Kawasaki and C. Nakamura. 2001. Construction of a high-resolution linkage map of rice brown planthopper (*Nilaparvata lugens* Stål) resistance gene *bph2*. **Theor. Appl. Genet.** 103: 526-532.

Murata, K., M. Fujiwara, C. Kaneda, S. Takumi, N. Mori and C. Nakamura. 1998. RFLP mapping of a brown planthopper (*Nilaparvata lugens* Stål) resistance gene *bph2* of indica rice introgressed into a japonica breeding line 'Norin-PL4'. **Genes Genet. Syst.** 73: 359-364.

Murata, K., M. Fujiwara, H. Murai, S. Takumi, N. Mori and C. Nakamura. 2001. Mapping of a brown planthopper (*Nilaparvata lugens* Stål) resistance gene *Bph9* on the long arm of rice chromosome 12. **Cereal Res. Commun.** 29: 245-250.

Nemoto, H., R. Ikeda and C. Kaneda. 1989. New genes for resistance to brown planthopper, *Nilaparvata lugens* Stål, in rice. **Jpn. J. Breeding.** 39: 23-28.

Nielson, M.W. and W.F. Lehman. 1980. Breeding approaches in alfalfa, pp. 277-311. *In* F.G. Maxwell and P.R. Jennings, eds. **Breeding Plant Resistance to Insects.** Wiley, New York.

Okada, T. 1977. Taxonomic characters for identification of the rice brown planthopper (*Nilaparvata lugens*) and its related species in the Asian and Pacific region, pp. 1-25. *In* FFTC, ed. **The Rice Brown Planthopper.** FFTC (ASPAC), Taipei.

Padgham, D.E., S. Woodhead and H.R. Rapusas. 1989. Feeding responses of the brown planthopper to resistant and susceptible host plants. **Bull. Entomol. Res.** 79: 309-318.

Padgham, D.E. and S. Woodhead. 1988. Variety-related feeding patterns in the brown planthopper, *Nilaparvata lugens* (Stål), on its host, the rice plant. **Bull. Entomol. Res.** 78: 338-349.

Painter, R.H. 1941. The economic value and biologic significance of insect resistance in plants. **J. Econ. Entomol.** 34: 358-367.

Panda, N. and G.S. Khush. 1995. Mechanism of resistance, pp. 151-206. *In* N. Panda and G.S. Khush, eds. **Host Plant Resistance to Insect.** CAB International, Wallingford.

Park, D.S., M.Y. Song, S.K. Park, S.K. Lee, J.H. Lee, S.Y. Song, M.Y. Eun, T.R Hahn, J.K. Sohn, G. Yi, M.H. Nam, and J.S. Jeon. 2008. Molecular tagging of the *Bph1* locus for resistance to brown planthopper (*Nilaparvata lugens* Stål) through representational difference analysis. **Mol. Genet. Genomics.** 208: 163–172.

Pathak, M.D. 1968. Ecology of common insect pest of rice. **Ann. Rev. Entomol.** 13: 257-294.

Pathak, M.D., C.H. Cheng and M.E. Fortuno. 1969. Resistance to *Nephotettix impicticeps* and *Nilaparvata lugens* in rice varieties. **Nature.** 233: 502-504.

Pathak, M.D. and Z.R. Khan. 1994. **Insect pests of rice.** Inter. Rice Res. Ins. and Inter. Cen. Insect Phys. Eco. pp. 22-24.

Padgham, D.E., S. Woodhead and H.R. Rapusas. 1989. Feeding responses of the brown planthopper to resistant and susceptible host plants. **Bull. Entomol. Res.** 79: 309-318.

Padgham, D.E. and S. Woodhead. 1988. Variety-related feeding patterns in the brown planthopper, *Nilaparvata lugens* (Stål), on its host, the rice plant. **Bull. Entomol. Res.** 78: 338-349.

Pongprasert, S. and P. Weerapat. 1979. Varietal resistance to the brown planthopper in Thailand, pp. 273-283. *In* International Rice Research Institute, ed. **Brown Planthopper: Threat to Rice Production in Asia.** IRRI, Los Baños, Philippines.

Phengrat, K. 2000. **Monitoring of the biotype of rice brown planthopper (*Nilaparvata lugens* stål) in Northeastern Thailand using biological characteristic and Random Amplified Polymorphic DNA.** M.S. thesis, Khon Kaen University.

Rahman, M.L., S. Chu, W. Jiang, K.K. Jena, and H.J. Koh. 2007. Impact of brown planthopper (*Nilaparvata lugens*) resistance on other agronomic traits in an introgression line derived from *O. minuta*, p. 338. **Proceedings of the 2nd International Conference on Rice for the Future**. Bangkok, Thailand.

Ren, X., X. Wang, H. Yuan, Q. Weng, L. Zhu and G. He. 2004. Mapping quantitative trait loci and expressed sequence tags related to brown planthopper resistance in rice. **Plant Breeding.** 123: 342-348.

Renganayaki, K., A.K. Fritz, S. Sadasivam, S. Pammi, S.E. Harrington, S.R. McCouch, S.M. Kumar and A.S. Reddy. 2002. Mapping and progress toward map-based cloning of brown planthopper biotype-4 resistance gene introgressed from *Oryza officinalis* into cultivated rice, *O. sativa*. **Crop Sci.** 42: 2112-2117.

Rithmontri, T., S. Tangchupong and N. Khongrod. 1998. Biotypes of brown planthopper, *Nilaparvata lugens* Stål in Northeast region, pp. 121-135. *In* Ubon Ratchathani Rice Research Center, ed. **The 9th Ubon Ratchathani Rice Research Center Annual Meeting.** Ubon Ratchathani, Thailand.

Sakai, T. and K. Sogawa. 1976. Effects of nutrient compounds on sucking response of the brown planthopper, *Nilaparvata lugens* (Homoptera: Delphacidae). **Appl. Entomol. Zool.** 11: 82-88.

Sanchez, A.C., D.S. Brar, N. Huang, Z. Li and G.S. Khush. 2000. Sequence tagged site marker-assisted selection of three bacterial blight resistance genes in rice. **Crop Sci.** 40: 792-797.

Sandstrom, J. 2000. Nutritional quality of phloem sap in relation to host plant-alternation in the bird cherry-oat aphid. **Chemoecology.** 10: 17-24.

Sasaki, T., M. Kawamura and H. Ishikawa. 1996. Nitrogen recycling in the brown planthopper, *Nilaparvata lugens*: involvement of yeast-like endosymbionts in uric acid metabolism. **J. Insect Physiol.** 42: 125-129.

Sexena, R.C. and S.H. Okech. 1985. Role of plant volatiles in resistance of selected rice varieties to brown planthopper, *Nilaparvata lugens* (Stål) (Homoptera: Delphacidae). **J. Chem. Ecol.** 11: 1601-1616.

Sharma, P.N., A. Torii, S. Takumi, N. Mori and C. Nakamura. 2004. Marker-assisted pyramiding of brown planthopper (*Nilaparvata lugens* Stål) resistance genes *Bph1* and *bph2* on rice chromosome 12. **Hereditas.** 140: 61-69.

Sharma, P.N., K. Murata, A. Torii, S. Takumi, N. Mori and C. Nakamura. 2003a. Towards molecular cloning of resistance genes against brown planthopper (*Nilaparvata lugens* Stål) in rice: a case study of natural insect resistance genes. **Trends in Entomol.** 3: 87-96.

Sharma, P.N., Y. Ketipearachchi, K. Murata, A. Torii, S. Takumi, N. Mori and C. Nakamura. 2003b. RFLP/AFLP mapping of a brown planthopper (*Nilaparvata lugens* Stål) resistance gene *Bph1* in rice. **Euphytica.** 129: 109-117.

Shigematsu, Y., N. Murofushi, K. Ito, C. Kaneda, S. Kawabe and N. Takahashi. 1982. Sterols and asparagines in the rice plant, endogenous factors related to resistance against the brown planthopper (*Nilaparvata lugens*). **Agric. Biol. Chem.** 46: 2877-2879.

Sidhu, G.S. and G.S. Khush. 1979. Linkage relationships of some genes for disease and insect resistance and semidwarf stature in rice. **Euphytica**. 28: 233-237.

Sōgawa, K. 1974. Study on the feeding habits of the brown planthopper, *Nilaparvata lugens* (Stål) (Hemiptera: Delphacidae), Probing stimulant. **Appl. Entomol. Zool.** 9: 204-213.

Sōgawa, K. 1976. Study on the feeding habits of the brown planthopper, *Nilaparvata lugens* (Stål) (Hemiptera: Delphacidae), probing stimulatory effect of rice flavonoid. **Appl. Entomol. Zool.** 11: 160-164.

Sōgawa, K. 1981. Hybridization experiments on three biotypes of the brown planthopper, *Nilaparvata lugens* (Homoptera: Delphacidae) at the IRRI, the Philippines. **Appl. Entomol. Zool.** 16: 193-199.

Sōgawa, K. 1982. The rice brown planthopper: feeding physiology and host plant interactions. **Ann. Rev. Entomol.** 27: 49-73.

Soundararajan, R.P., P. Kadirvel, K. Gunathilagaraj and M. Maheswaran. 2004. Mapping of quantitative trait loci associated with resistance to brown planthopper in rice by means of a doubled haploid population. **Crop Sci.** 44: 2214-2220.

Spiller, N.J. 1990. An ultrastructural study of the stylet pathway of the brown planthopper *Nilaparvata lugens*. **Entomol. exp. appl.** 54: 191-193.

Stevenson, P.C., F.M. Kimmins, R.J. Grayer and S. Raveendranath. 1996. Schaftosides from rice phloem as feeding inhibitors and resistance factors to brown planthoppers, *Nilaparvata lugens*. **Entomol. Exp. Appl**. 80: 246-249.

Su, C.C., H.Q. Zhai, X.N. Cheng and J.M. Wan. 2002. Detection and analysis of QTLs for resistance to brown planthopper, *Nilaparvata lugens* (Stål), in rice (*Oryza sativa* L.), using backcross in bred lines. **Acta Gentica Sinica.** 29: 332-338.

Sun, L., C. Wang, C. Su, Y. Liu, H. Zhai and J. Wan. 2006. Mapping and marker-assisted selection of a brown planthopper resistance gene *bph2* in rice (*Oryza sativa* L.). **Acta Genetica Sinica.** 33: 717-723.

Sun, L., C. Su, C. Wang, H. Zhai and J. Wan. 2005. Mapping of a major resistance gene to the brown planthopper in the rice cultivar Rathu Heenati. **Breeding Sci.** 55: 391-396.

Sun, L, Y. Liu, L. Jiang, C. Su and J. Wan. 2007. Identification of quantitative trait loci associated with resistance to brown planthopper in the *indica* rice cultivar Col.5 Thailand. **Hereditas**. 144: 48-52.

Syobu, S., H. Mikuriya, J. Yamaguchi and M. Matsuzaki. 2002. Fluctuations and factors affecting the wing-form ratio of the brown planthopper, *Nilaparvata lugens* Stål in rice fields. **Jpn. J. Appl. Entomol. Zool.** 46: 135-143.

Tabuchi, M., T. Abiko and T. Yamaya. 2007. Assimilation of ammonium ions and reutilization of nitrogen in rice (*Oryza sativa* L.). **J. Exp. Bot.** 58: 2319-2327.

Tanaka, K. 1997. Development of resistance-breaking biotypes of the brown planthopper against resistant rice varieties. **Farming Jpn.** 31: 22-26.

Tanaka, K. 1999. Quantitative genetic analysis of biotypes of the brown planthopper *Nilaparvata lugens*: heritability of virulence to resistant rice varieties. **Entomol. Exp. Appl.** 90: 279-287.

Tjallingii, W.F. 2006. Salivary secretions by aphids interacting with proteins of phloem wound responses. **J. Exp. Bot.** 57: 739-745.

Tobin, A.K. and T. Yamaya. 2001. Cellular compartmentation of ammonium assimilation in rice and barley. **J. Exp. Bot.** 52: 591-604.

Toojinda, T., S. Tragoonrung, A. Vanavichit, J.L. Siangliw, N. Pa-In, J. Jantaboon, M. Siangliw and S. Fukai. 2005. Molecular breeding for rainfed lowland rice in the Mekong region. **Plant Prod. Sci.** 8: 330-333.

Tripop, M. 1997. **Variation of Brown Planthopper, *Nilaparvata lugens* Stål, Population from the Four Geographical Regions of Thailand.** M.S. thesis, Kasetsart University.

Van Ooijen, J.W. 2004. **MapQTL®5, Software for the Mapping of Quantitative Trait Loci in Experimental Populations**. Kyazma B.V., Wageningen, Netherlands.

Van Ooijen, J.W. and R.E. Voorrips. 2001. **JoinMap®3.0, Software for the Calculation of Genetic Linkage Maps.** Plant Research International, Wageningen, the Netherlands.

Velusamy, R., E.A. Heinrichs and F.G. Medrano. 1986. Greenhouse techniques to identify field resistance to the brown planthopper, *Nilaparvata lugens* (Stål) (Homoptera: Delphacidea), in rice cultivars. **Crop Protect.** 5: 328-333.

Velusamy, R., M. Ganesh Kumar and Y.S. Johnson Thangaraj Edward. 1995. Mechanisms of resistance to brown planthopper *Nilaparvata lugens* in wild rice (*Oryza* spp.) cultivars. **Entomol. Exp. Appl.** 74: 245-251.

Wanchana, S., W. Kamolsukyunyong, S. Ruengphayak, T. Toojinda, S. Tragoonrung and A. Vanavichit. 2005. A Rapid construction of a physical contig across a 4.5 cM region for rice grain aroma facilitates marker enrichment for positional cloning. **Sci. Asia.** 31: 299-306.

Watanabe, T. and H. Kitagawa. 2000. Photosynthesis and translocation of assimilates in rice plants following phloem feeding by the planthopper *Nilaparvata lugens* (Homoptera: Delphacidae). **J. Econ. Entomol.** 93: 1192-1198.

Wang, B., Z. Huang, L. Shu, X. Ren, X. Li and G. He. 2001. Mapping of two new brown planthopper resistance genes from wild rice. **Chinese Sci. Bull.** 46: 1092-1095.

Wang, R., W. Shen, L. Liu, L. Jiang, Y. Liu, N. Su and J. Wan. 2008. A novel lipoxygenase gene from developing rice seeds confers dual position specificity and responds to wounding and insect attack. **Plant Mol. Biol.** 66: 401-414.

Weibull, J.H.W. 1988. Free amino acids in the phloem sap from oats and barley resistant to *Rhopalosiphum padi*. **Phytochem.** 27: 2069-2072.

Will, T. and Van Bel A.J.E. 2006. Physical and chemical interactions between aphids and plants. **J. Exp. Bot.** 57: 729-737.

Xu, X.F, H.W. Mei, L.J. Luo, X.N. Cheng and Z.K. Li. 2002. RFLP-facilitated investigation of the quantitative resistance of rice to brown planthopper (*Nilaparvata lugens*). **Theor. Appl. Genet.** 104: 248-253.

Yamaya, T, M. Obara, H. Nakajima, S. Sasaki, T. Hayakawa and T. Sato. 2002. Genetic manipulation and quantitative-trait loci mapping for nitrogen recycling in rice. **J. Exp. Bot.** 53: 917-925.

Yan, H.M., R. Qin, W.W. Jin, G.C. He and Y.C. Song. 2002. Comparative physical mapping of *Bph3* with BAC-FISH in *Oryza officinalis* and *O. sativa*. **Acta Botanica Sinica.** 44: 583-587.

Yang, H., X. Ren, Q. Weng, L. Zhu and G. He. 2002. Molecular mapping and genetic analysis of a rice brown planthopper (*Nilaparvata lugens* Stål) resistance gene. **Hereditas.** 136: 39-43.

Yang, H, A. You, Z. Yang, F. Zhang, R. He, L. Zhu and G. He. 2004. High-resolution genetic mapping at the *Bph15* locus for brown planthopper resistance in rice (*Oryza sativa* L.). **Theor. Appl. Genet.** 110: 182-91.

Yoshihara, T., K. Sogawa, M.D. pathak and R. Villareal. 1979. Comparison of oxalic acid concentration in rice varieties resistant and susceptible to the brown planthopper. **Int. Rice Res. Newsl.** 4: 10-11.

Yuan, H., X. Chen, L. Zhu and G. He. 2005. Identification of genes responsive to brown planthopper *Nilaparvata lugens* Stål (Homoptera: Delphacidae) feeding in rice. **Planta.** 221: 105-112.

Zhang, G., W. Zhang, B. Lian, L. Gu, Q. Zhou and T.X. Liu. 1999. Insecticidal effects of extracts from two rice varieties to brown planthopper, *Nilaparvata lugens*. **J. Chem. Ecol.** 25: 1843-1853.

Zhang, Q. 2007. Strategies for developing green super rice. **Proc. Natl. Acad. Sci. USA.** 104, 16402–16409.

Zhang, S.L., D.H. Ni, C.X. Yi, L. Li, X.F. Wang, Z.Y. Wang and J.B. Yang. 2005. Lowering grain amylose content in backcross offsprings of *indica* rice variety 057 by molecular marker-assisted selection. **Rice Sci.** 12: 157-162.

Zhao, X.Q. and W.M. Shi. 2006. Expression analysis of the glutamine synthetase and glutamate synthase gene families in young rice (*Oryza sativa*) seedlings. **Plant Sci.** 170: 748-754.

Zhou, P.H., Y.F. Tan, Y.Q. He, C.G. Xu and Q. Zhang. 2003. Simultaneous improvement for four quality traits of Zhenshan 97, an elite parent of hybrid rice, by molecular marker-assisted selection. **Theor. Appl. Genet.** 106: 326-331.

Appendix

Appendix Table 1 Brown planthopper populations collected from rice fields in Thailand.

No.	Location		Code	Year of collection
	District	Province		
1	Muang	Yasothon	MUA-YST	2004
2	Sichon	Nakhon Si Thammarat	SCN-NST	2003
3	Muang Samsip	Ubon Ratchathani	MSS-UBN	2004
4	Muang	Kalasin	MUA-KLS	2004
5	Muang	Sakon Nakhon	MUA-SKN	2004
6	Hang Chat	Lampang	HCT-LPG	2004
7	Muang	Ubon Ratchathani	MUA-UBN	2004
8	Khon San	Chaiyaphum	KSN-CYP	2003
9	Pan	Chiang Rai	PAN-CRI	2003
10	Wiang Pa Pao	Chiang Rai	WPP-CRII	2003
11	Wiang Pa Pao	Chiang Rai	WPP-CRIII	2003
12	Muang	Mukdahan	MUA-MDH	2004
13	Wiang Sa	Nan	WSA-NAN	2003
14	San Pa Tong	Chiang Mai	SPT-CMI	2003
15	Senangkhanikhom	Amnat Charoen	SNK-ANR	2004
16	Muang	Nakhon Phanom	MUA-NPN	2004
17	Den Chai	Phrae	DCI-PRE	2003
18	Mae Lao	Chiang Rai	MLO-CRI	2003
19	Ta Phraya	Sa Kaeo	TPY-SKW	2004
20	Det Udom	Ubon Ratchathani	DUD-UBN	2004
21	Phibun Mangsahan	Ubon Ratchathani	PBH-UBN	2005
22	Huai Thalaeng	Nakhon Ratchasima	HTL-NKM	2003
23	Khok Samrong	Lop Buri	KSL-LBR	2004
24	Muang	Sa Kaeo	MUA-SKW	2004
25	Bang Len	Nakhon Pathom	BLN-NKT	2004
26	Thawatchaburi	Roi Et	TWR-RET	2005
27	Chakkarat	Nakhon Ratchasima	CKR-NRM	2004
28	Lat Lum Kaeo	Pathum Thani	LLK-PTT	2004
39	Muang	Ang Thong	MUA-ATG	2004
30	Non Din Daeng	Buri Rum	NDD-BRR	2004
31	Phanom Thuan	Kanchanaburi	PNT-KBR	2004
32	Si Prachan	Suphan Buri	SPC-SPR	2004
33	Phanom Sarakham	Chachoengsao	PNK-CCS	2004
34	Bang Nam Prieo	Chachoengsao	BNP-CCS	2004
35	Thawatchaburi	Roi Et	TWR-RET	2003
36	Satuek	Buri Rum	STK-BRR	2003
37	Sangkha	Surin	SAK-SRN	2003
38	Phon Sai	Roi Et	PNS-RET	2003
39	Ban Kruat	Buri Rum	BNK-BRR	2003
40	Sung Noen	Nakhon Ratchasima	SNN-NRM	2004
41	Tha Tum	Surin	TTM-SRN	2004
42	Lam Plai Mat	Buri Rum	LPM-BRR	2004
43	Muang	Chai Nat	MUA-CNT	2003
44	Yang Chum Noi	Si Sa Ket	YCN-SSK	2004
45	Phu Sing	Si Sa Ket	PST-SSK	2004

Appendix Table 2 Phenotype and genotype of 208 BC_1F_2 individuals from the cross of PTB33×RD6 for BPH resistance. RD=homozygous RD6; PT=homozygous PTB33; H=heterozygous.

No.	entries	score	RM190	RM588	RM589	RM469	RM3353
1	360/15-1	9	H	H	H	H	H
2	360/15-2	9	RD	RD	RD	RD	RD
3	360/15-3	9	RD	RD	RD	RD	RD
4	360/15-4	7	H	H	H	H	H
5	360/15-5	9	RD	RD	RD	RD	RD
6	360/15-6	9	H	H	H	H	H
7	360/15-7	3	PT	PT	PT	PT	PT
8	360/15-8	9	RD	RD	RD	RD	RD
9	360/15-9	9	H	H	H	H	H
10	360/15-10	7	H	H	H	H	H
11	360/15-11	9	RD	RD	RD	RD	RD
12	360/15-12	7	H	H	H	H	H
13	360/15-13	5	H	H	H	H	H
14	360/15-14	7	H	H	H	H	H
15	360/15-15	9	H	H	H	H	H
16	360/15-16	7	H	H	H	H	H
17	360/15-17	9	RD	RD	RD	RD	RD
18	360/15-18	5	PT	PT	PT	PT	PT
19	360/15-19	9	RD	RD	RD	RD	RD
20	360/15-20	9	RD	RD	RD	RD	RD
21	360/15-21	7	H	H	H	H	H
22	360/15-22	7	H	H	H	H	H
23	360/15-23	5	H	H	H	H	H
24	360/15-24	3	PT	PT	PT	PT	PT
25	360/15-25	3	PT	PT	PT	PT	PT
26	360/15-26	4	PT	PT	PT	PT	PT
27	360/15-27	7	RD	H	H	H	H
28	360/15-28	3	PT	PT	PT	PT	PT
29	360/15-29	7	H	H	H	H	H
30	360/15-30	3	PT	PT	PT	PT	PT
31	360/15-31	3	PT	PT	PT	PT	PT
32	360/15-32	5	RD	RD	RD	RD	RD
33	360/15-33	3	H	H	H	H	H
34	360/15-34	3	PT	PT	PT	PT	PT
35	360/15-35	3	PT	PT	PT	PT	PT
36	360/15-36	7	RD	RD	RD	RD	RD
37	360/15-37	7	RD	RD	RD	RD	RD
38	360/15-38	7	RD	RD	RD	RD	RD
39	360/15-39	7	H	H	H	H	H
40	360/15-40	7	PT	PT	PT	PT	PT
41	360/15-41	7	H	H	H	H	H
42	360/15-42	9	H	H	H	H	H

Appendix Table 2 (Continued)

No.	entries	score	RM190	RM588	RM589	RM469	RM3353
43	360/15-43	3	PT	PT	PT	PT	PT
44	360/15-44	9	H	H	H	H	H
45	360/15-45	7	H	H	H	H	H
46	360/15-46	9	H	H	H	H	H
47	360/15-47	9	RD	RD	RD	RD	RD
48	360/15-48	9	RD	RD	RD	RD	RD
49	360/15-49	3	PT	PT	PT	PT	PT
50	360/15-50	5	H	H	H	H	H
51	360/15-51	9	RD	RD	RD	RD	RD
52	360/15-52	3	PT	PT	PT	PT	PT
53	360/15-53	9	RD	RD	RD	RD	RD
54	360/15-54	9	RD	RD	RD	RD	RD
55	360/15-55	3	H	H	PT	PT	RD
56	360/15-56	7	H	H	H	H	H
57	360/15-57	9	RD	RD	RD	RD	RD
58	360/15-58	5	RD	RD	RD	RD	RD
59	360/15-59	5	H	H	H	H	H
60	360/15-60	7	H	H	H	H	H
61	360/15-61	7	H	H	H	H	H
62	360/15-62	3	H	H	H	H	H
63	360/15-63	3	PT	PT	PT	PT	PT
64	360/15-64	3	PT	PT	PT	PT	PT
65	360/15-65	9	H	H	H	H	H
66	360/15-66	3	PT	PT	PT	PT	PT
67	360/15-67	4	PT	PT	PT	PT	PT
68	360/15-68	5	H	H	H	H	H
69	360/15-69	7	H	H	H	H	H
70	360/15-70	3	PT	PT	PT	PT	PT
71	360/15-71	4	PT	PT	PT	PT	PT
72	360/15-72	5	H	H	H	H	H
73	360/15-73	3	PT	PT	PT	PT	PT
74	360/15-74	3	RD	RD	RD	RD	RD
75	360/15-75	5	PT	PT	PT	PT	PT
76	360/15-76	3	PT	PT	PT	PT	PT
77	360/15-77	7	PT	PT	PT	PT	H
78	360/15-78	9	RD	RD	RD	RD	RD
79	360/15-79	7	PT	PT	PT	PT	PT
80	360/15-80	9	H	H	H	H	H
81	360/15-81	9	H	H	H	H	H
82	360/15-82	9	RD	RD	RD	RD	RD
83	360/15-83	7	H	H	PT	PT	PT
84	360/15-84	5	H	H	H	H	H
85	360/15-85	3	PT	PT	PT	H	H
86	360/15-86	7	H	H	H	H	H
87	360/15-87	7	H	H	H	H	H
88	360/15-88	7	H	H	H	H	H

Appendix Table 2 (Continued)

No.	entries	score	RM190	RM588	RM589	RM469	RM3353
89	360/15-89	7	H	H	H	H	H
90	360/15-90	4	PT	PT	PT	PT	PT
91	360/15-91	3	PT	PT	PT	PT	PT
92	360/15-92	7	H	H	H	H	RD
93	360/15-93	9	RD	RD	RD	RD	RD
94	360/15-94	7	PT	PT	PT	PT	H
95	360/15-95	3	PT	PT	PT	PT	PT
96	360/15-96	4	PT	PT	PT	PT	PT
97	360/15-97	7	RD	RD	RD	RD	RD
98	360/15-98	4	PT	PT	PT	PT	PT
99	360/15-99	3	H	H	PT	PT	PT
100	360/15-100	9	RD	RD	RD	RD	RD
101	360/15-101	5	RD	RD	RD	RD	RD
102	360/15-102	7	H	H	H	H	H
103	360/15-103	3	PT	PT	PT	PT	PT
104	360/15-104	5	PT	H	H	H	H
105	360/15-105	3	PT	PT	PT	PT	PT
106	360/15-106	7	H	H	H	H	H
107	360/15-107	9	RD	RD	RD	RD	RD
108	360/15-108	7	H	H	H	PT	PT
109	360/15-109	4	H	H	H	H	H
110	360/15-110	9	RD	RD	RD	RD	RD
111	360/15-111	3	PT	PT	PT	PT	PT
112	360/15-112	3	PT	PT	PT	PT	PT
113	360/15-113	9	H	RD	RD	RD	RD
114	360/15-114	9	RD	RD	RD	RD	RD
115	360/15-115	7	H	H	H	H	RD
116	360/15-116	3	PT	PT	PT	PT	PT
117	360/15-117	7	H	H	H	H	H
118	360/15-118	7	H	H	H	H	H
119	360/15-119	7	H	H	H	H	H
120	360/15-120	3	PT	PT	PT	PT	PT
121	360/15-121	7	H	H	H	H	H
122	360/15-122	4	PT	PT	PT	PT	PT
123	360/15-123	9	H	H	H	H	H
124	360/15-124	9	H	H	H	RD	RD
125	360/15-125	9	RD	RD	RD	RD	RD
126	360/15-126	9	RD	RD	RD	RD	RD
127	360/15-127	9	RD	RD	RD	RD	RD
128	360/15-128	7	H	H	H	H	H
129	360/15-129	5	PT	PT	PT	PT	H
130	360/15-130	3	PT	PT	PT	PT	PT
131	360/15-131	9	H	H	H	H	H
132	360/15-132	5	H	H	H	H	H
133	360/15-133	7	H	H	H	H	H
134	360/15-134	7	H	H	H	H	H

Appendix Table 2 (Continued)

No.	entries	score	RM190	RM588	RM589	RM469	RM3353
135	360/15-135	9	H	H	H	H	H
136	360/15-136	7	H	H	H	H	H
137	360/15-137	9	H	H	H	H	H
138	360/15-138	3	H	H	H	H	H
139	360/15-139	3	H	H	PT	PT	PT
140	360/15-140	3	PT	H	H	H	H
141	360/15-141	3	PT	PT	PT	PT	PT
142	360/15-142	3	PT	PT	PT	PT	PT
143	360/15-143	3	PT	H	H	H	H
144	360/15-144	9	RD	RD	RD	RD	RD
145	360/15-145	9	RD	RD	RD	RD	RD
146	360/15-146	9	RD	RD	RD	H	H
147	360/15-147	9	H	H	H	H	H
148	360/15-148	3	PT	PT	PT	PT	PT
149	360/15-149	3	PT	H	H	H	H
150	360/15-150	9	RD	RD	RD	RD	RD
151	360/15-151	7	H	H	H	H	H
152	360/15-152	3	PT	PT	PT	PT	PT
153	360/15-153	7	H	H	H	H	H
154	360/15-154	5	H	H	H	H	H
155	360/15-155	9	RD	RD	RD	RD	RD
156	360/15-156	7	H	H	H	H	H
157	360/15-157	3	PT	PT	PT	PT	PT
158	360/15-158	9	H	H	H	H	H
159	360/15-159	3	PT	PT	PT	PT	PT
160	360/15-160	5	H	H	H	H	H
161	360/15-161	5	H	H	H	H	H
162	360/15-162	7	H	H	H	H	H
163	360/15-163	9	RD	RD	RD	RD	RD
164	360/15-164	7	H	H	H	H	H
165	360/15-165	9	H	H	H	H	H
166	360/15-166	3	PT	PT	PT	PT	PT
167	360/15-167	9	RD	RD	H	H	H
168	360/15-168	9	RD	RD	RD	H	H
169	360/15-169	3	PT	PT	PT	PT	PT
170	360/15-170	3	PT	PT	PT	PT	PT
171	360/15-171	3	PT	PT	PT	PT	PT
172	360/15-172	3	PT	PT	PT	PT	PT
173	360/15-173	9	RD	RD	RD	RD	RD
174	360/15-174	9	H	H	H	H	H
175	360/15-175	7	H	H	H	H	H
176	360/15-176	9	H	H	H	H	H
177	360/15-177	3	H	H	PT	PT	PT
178	360/15-178	7	H	H	H	H	H
179	360/15-179	7	H	H	H	H	H
180	360/15-180	7	H	H	H	H	H

Appendix Table 2 (Continued)

No.	entries	score	RM190	RM588	RM589	RM469	RM3353
181	360/15-181	9	H	H	H	H	H
182	360/15-182	3	PT	PT	PT	PT	PT
183	360/15-183	7	H	H	H	H	H
184	360/15-184	9	PT	PT	H	H	H
185	360/15-185	9	RD	RD	RD	RD	RD
186	360/15-186	3	PT	PT	PT	PT	PT
187	360/15-187	9	H	H	H	RD	RD
188	360/15-188	3	PT	PT	PT	PT	PT
189	360/15-189	7	H	H	H	H	H
190	360/15-190	3	PT	PT	PT	PT	PT
191	360/15-191	9	RD	RD	RD	RD	RD
192	360/15-192	3	PT	PT	PT	PT	PT
193	360/15-193	7	H	H	H	H	H
194	360/15-194	3	PT	PT	PT	PT	PT
195	360/15-195	9	RD	RD	RD	RD	RD
196	360/15-196	7	H	H	H	H	H
197	360/15-197	3	PT	PT	PT	PT	H
198	360/15-198	9	RD	RD	RD	RD	RD
199	360/15-199	3	PT	PT	PT	PT	PT
200	360/15-200	9	H	H	H	H	H
201	360/15-201	7	H	H	H	H	H
202	360/15-202	7	H	H	H	0	H
203	360/15-203	3	PT	PT	PT	PT	PT
204	360/15-204	9	RD	RD	RD	RD	RD
205	360/15-205	9	H	H	H	H	H
206	360/15-206	3	PT	PT	PT	PT	PT
207	360/15-207	5	H	H	H	H	H
208	360/15-208	9	RD	RD	RD	RD	RD

Appendix Table 3 Phenotype and genotype of 333 BC_3F_2 individuals from the cross of Rathu Heenati×KDML105 for BPH resistance.
KD=homozygous KDML105; RT=homozygous Rathu Heenati; H=heterozygous

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
1	1	4.0	H	H	H	-	H	H
2	2	4.0	KD	H	KD	KD	KD	KD
3	21	3.0	KD	KD	KD	KD	KD	KD
4	42	4.0	RT	RT	RT	RT	RT	RT
5	47	4.0	KD	KD	KD	KD	KD	KD
6	48	5.0	KD	H	KD	KD	KD	KD
7	50	3.0	RT	RT	RT	RT	RT	RT
8	51	3.0	RT	RT	RT	RT	RT	RT
9	60	2.0	H	RT	H	H	RT	RT
10	68	3.0	H	H	H	H	H	H
11	70	4.0	RT	RT	RT	RT	RT	RT
12	82	9.0	H	H	H	H	H	H
13	83	9.0	H	H	H	H	H	H
14	91	4.0	RT	RT	RT	RT	RT	RT
15	96	3.0	RT	RT	RT	RT	RT	RT
16	100	5.0	H	H	H	H	H	H
17	107	1.0	RT	RT	RT	RT	RT	RT
18	115	2.0	RT	RT	RT	RT	RT	RT
19	116	6.0	H	RT	H	H	H	H
20	119	4.0	RT	RT	RT	RT	RT	RT
21	121	3.0	RT	H	RT	RT	RT	RT
22	141	3.0	RT	RT	RT	RT	RT	RT
23	147	5.0	H	H	H	H	H	H
24	161	9.0	H	H	H	H	H	H
25	167	5.0	H	H	H	H	H	H
26	169	9.0	KD	KD	KD	KD	KD	KD
27	170	2.0	RT	RT	RT	RT	RT	RT
28	173	4.0	H	H	H	H	H	H
29	174	9.0	KD	KD	KD	KD	KD	KD
30	176	1.0	RT	RT	RT	RT	RT	RT
31	178	3.0	KD	H	KD	KD	KD	KD
32	181	9.0	KD	KD	KD	KD	KD	KD
33	186	3.0	RT	RT	RT	-	RT	RT
34	189	9.0	KD	KD	KD	KD	KD	KD
35	190	5.0	H	H	H	H	H	H
36	194	1.0	RT	RT	RT	RT	RT	RT

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
37	195	3.0	RT	RT	RT	RT	RT	RT
38	198	3.0	RT	RT	RT	RT	RT	RT
39	202	3.0	H	H	H	H	H	H
40	203	3.0	H	H	H	H	H	H
41	207	9.0	H	H	H	H	H	H
42	208	3.0	RT	RT	RT	RT	RT	RT
43	209	6.0	KD	KD	KD	KD	KD	KD
44	210	4.0	RT	RT	RT	RT	RT	RT
45	211	3.0	RT	RT	RT	RT	RT	RT
46	215	6.0	H	H	H	H	H	H
47	220	9.0	H	H	H	H	H	H
48	227	3.0	H	H	H	H	H	H
49	231	2.0	RT	RT	RT	RT	RT	RT
50	236	3.0	RT	RT	RT	RT	RT	RT
51	238	4.0	H	H	H	H	H	H
52	239	9.0	KD	KD	KD	KD	KD	KD
53	241	9.0	H	H	H	H	H	H
54	242	2.0	H	H	H	H	H	H
55	245	3.0	RT	RT	RT	RT	RT	RT
56	249	5.0	H	H	H	H	H	H
57	251	2.0	RT	H	RT	RT	RT	RT
58	255	9.0	KD	KD	KD	KD	KD	KD
59	258		KD	KD	KD	KD	KD	KD
60	259	6.0	KD	H	KD	KD	KD	KD
61	265	4.0	RT	RT	RT	RT	RT	RT
62	269	5.0	H	H	H	H	H	H
63	271	5.0	H	H	H	H	H	H
64	272	6.0	H	H	H	H	H	H
65	274	6.0	KD	KD	KD	KD	KD	KD
66	275	5.0	H	H	H	H	H	H
67	276	4.0	H	H	H	H	H	H
68	277	6.0	RT	H	RT	RT	H	RT
69	278	9.0	KD	KD	KD	KD	KD	KD
70	279	9.0	KD	KD	KD	KD	KD	KD
71	280	9.0	KD	KD	KD	KD	KD	KD
72	284	4.0	H	H	H	H	H	H
73	288	9.0	H	H	H	H	RT	H
74	289	9.0	KD	KD	KD	KD	KD	KD
75	290	6.0	H	H	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
76	291	9.0	KD	KD	KD	KD	KD	KD
77	294	9.0	KD	KD	KD	KD	KD	KD
78	298	1.0	RT	RT	RT	RT	RT	RT
79	300	9.0	KD	KD	KD	KD	KD	KD
80	302	7.0	KD	KD	KD	KD	KD	KD
81	303	3.0	RT	RT	RT	RT	RT	RT
82	304	4.0	RT	RT	RT	RT	RT	RT
83	305	3.0	H	H	H	H	H	H
84	306	4.0	H	H	H	H	H	H
85	307	4.0	RT	H	RT	RT	RT	RT
86	308	9.0	KD	KD	KD	KD	KD	KD
87	311	9.0	KD	KD	KD	KD	KD	KD
88	313	1.0	RT	RT	RT	RT	RT	RT
89	315	2.0	RT	RT	RT	RT	RT	-
90	318	9.0	KD	KD	KD	KD	KD	KD
91	320	9.0	H	H	H	H	H	H
92	321	4.0	H	H	H	H	H	H
93	322	4.0	H	RT	H	H	H	H
94	323	1.0	H	H	H	H	H	H
95	326	3.0	H	H	H	H	-	H
96	327	3.0	RT	RT	RT	RT	RT	-
97	330	4.0	RT	RT	RT	RT	RT	RT
98	331	4.0	H	H	H	H	H	H
99	333	3.0	RT	-	RT	RT	RT	RT
100	334	9.0	KD	KD	KD	KD	KD	KD
101	335	1.0	RT	RT	RT	RT	RT	RT
102	336	6.0	H	H	H	H	H	H
103	339	3.0	RT	RT	RT	RT	RT	RT
104	341	4.0	RT	RT	RT	RT	RT	RT
105	347	5.0	H	H	H	H	H	H
106	348	6.0	H	H	H	H	H	H
107	349	9.0	H	H	H	H	H	H
108	351	7.0	H	H	H	H	H	H
109	354	1.0	RT	RT	RT	RT	RT	RT
110	355	4.0	RT	RT	RT	RT	RT	RT
111	357	8.0	KD	KD	KD	KD	KD	KD
112	360	5.0	RT	RT	RT	RT	RT	RT
113	362	4.0	H	KD	H	H	H	H
114	364	4.0	H	H	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
115	365	9.0	KD	KD	KD	KD	KD	KD
116	366	1.0	RT	H	RT	RT	H	RT
117	367	5.0	H	H	H	H	H	H
118	368	5.0	H	H	H	H	H	H
119	369	4.0	H	H	H	H	H	H
120	372	9.0	H	RT	H	H	H	H
121	375	7.0	H	H	H	H	H	H
122	385	6.0	H	H	H	H	H	H
123	386	7.0	KD	KD	KD	KD	KD	KD
124	391	4.0	RT	RT	RT	RT	RT	RT
125	392	9.0	KD	H	KD	KD	KD	KD
126	393	9.0	KD	H	KD	KD	KD	KD
127	394	9.0	H	RT	H	H	H	H
128	399	9.0	H	H	H	H	H	H
129	400	8.0	H	H	H	H	H	H
130	401	4.0	RT	RT	RT	RT	RT	RT
131	402	9.0	KD	KD	KD	KD	KD	KD
132	403	3.0	RT	RT	RT	RT	RT	RT
133	404	4.0	H	H	H	H	H	H
134	410	9.0	KD	KD	KD	KD	KD	KD
135	411	9.0	H	H	H	H	H	H
136	413	6.0	H	H	H	H	H	H
137	416	9.0	KD	KD	KD	KD	KD	KD
138	417	9.0	H	H	H	H	H	H
139	418	9.0	H	H	H	H	H	H
140	419	4.0	H	H	H	H	H	H
141	420	3.0	H	H	H	H	H	H
142	421	3.0	RT	RT	RT	RT	RT	RT
143	423	6.0	H	H	H	H	H	H
144	424	7.0	H	H	H	H	H	H
145	425	9.0	KD	-	KD	KD	KD	KD
146	426	7.0	H	H	H	H	H	H
147	429	9.0	KD	KD	KD	KD	KD	KD
148	430	4.0	RT	RT	RT	RT	RT	RT
149	433	7.0	H	H	H	H	H	H
150	434	9.0	H	H	H	H	H	H
151	435	5.0	RT	RT	RT	RT	RT	RT
152	440	4.0	H	H	H	H	H	H
153	446	7.0	H	H	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
154	448	9.0	H	H	H	H	H	H
155	449	9.0	KD	KD	KD	KD	KD	KD
156	450	4.0	H	RT	RT	H	RT	RT
157	461	8.0	H	H	H	H	H	H
158	462	8.0	H	H	H	H	H	H
159	463	3.0	RT	H	RT	RT	RT	RT
160	464	9.0	H	H	H	H	H	H
161	465	9.0	H	H	H	H	H	H
162	479	4.0	RT	RT	RT	RT	RT	RT
163	480	9.0	H	H	H	H	H	H
164	484	5.0	RT	RT	RT	RT	RT	RT
165	488	9.0	KD	KD	KD	KD	KD	KD
166	489	9.0	H	H	H	H	H	H
167	497	4.0	RT	RT	RT	RT	RT	RT
168	499	6.0	H	H	H	H	H	H
169	508	9.0	H	H	H	H	H	H
170	510	5.0	H	H	H	H	H	H
171	513	3.0	RT	RT	RT	RT	RT	RT
172	514	5.0	KD	H	KD	KD	H	KD
173	516	7.0	KD	KD	KD	KD	KD	KD
174	517	5.0	RT	RT	RT	RT	RT	RT
175	518	6.0	H	H	H	H	H	H
176	519	9.0	H	H	H	H	H	H
177	520	9.0	H	H	H	H	H	H
178	523	3.0	H	H	H	H	H	H
179	524	5.0	H	H	H	H	H	H
180	525	9.0	KD	KD	KD	KD	KD	KD
181	527	4.0	RT	RT	RT	RT	RT	RT
182	530	4.0	H	H	H	H	H	H
183	531	4.0	RT	RT	RT	RT	RT	RT
184	532	4.0	H	H	H	H	H	H
185	534	4.0	H	H	H	H	H	H
186	535	9.0	KD	KD	KD	KD	KD	KD
187	540	4.0	KD	H	KD	KD	KD	KD
188	545	6.0	H	H	H	H	H	KD
189	550	6.0	KD	KD	KD	KD	KD	KD
190	554	5.0	H	H	H	H	H	H
191	561	9.0	KD	KD	KD	KD	KD	KD
192	565	9.0	KD	KD	KD	KD	KD	KD

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
193	567	3.0	RT	RT	RT	RT	RT	RT
194	568	9.0	H	KD	KD	H	KD	KD
195	569	6.0	H	H	H	H	H	H
196	572	9.0	KD	KD	KD	KD	KD	KD
197	574	9.0	KD	H	KD	KD	KD	KD
198	577	9.0	KD	KD	KD	KD	KD	KD
199	582	1.0	RT	RT	RT	RT	RT	RT
200	586	4.0	RT	RT	RT	RT	RT	RT
201	589	9.0	KD	H	KD	KD	KD	KD
202	590	6.0	H	H	H	H	H	H
203	592	3.0	H	RT	RT	H	RT	RT
204	593	3.0	RT	RT	RT	RT	RT	RT
205	595	9.0	RT	H	RT	RT	RT	RT
206	596	9.0	H	H	H	H	H	H
207	598	7.0	H	H	H	H	H	H
208	599	9.0	KD	KD	KD	KD	KD	KD
209	602	7.0	KD	H	KD	KD	KD	KD
210	603	3.0	RT	RT	RT	RT	RT	RT
211	604	3.0	RT	RT	RT	RT	RT	RT
212	608	9.0	H	H	H	H	H	H
213	610	4.0	RT	RT	RT	RT	RT	RT
214	611	5.0	RT	RT	RT	RT	RT	RT
215	615	4.0	H	H	H	H	H	H
216	619	9.0	H	H	H	H	H	H
217	621	5.0	H	H	H	H	H	H
218	622	6.0	H	H	H	H	H	H
219	625	5.0	KD	KD	KD	KD	KD	KD
220	631	9.0	KD	KD	KD	KD	KD	KD
221	638	9.0	KD	KD	KD	KD	KD	KD
222	639	3.0	RT	RT	RT	RT	RT	RT
223	640	9.0	KD	KD	KD	KD	KD	KD
224	641	3.0	RT	RT	RT	RT	RT	RT
225	645	3.0	RT	H	RT	RT	RT	RT
226	653	6.0	H	H	H	H	H	H
227	655	3.0	RT	RT	RT	RT	RT	RT
228	659	3.0	H	RT	H	H	H	H
229	660	5.0	H	H	H	H	H	H
230	673	2.0	RT	RT	RT	RT	RT	RT
231	674	7.0	H	KD	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
232	675	4.0	RT	RT	RT	RT	RT	RT
233	677	2.0	RT	H	RT	RT	RT	RT
234	681	1.0	RT	RT	RT	RT	RT	RT
235	682	1.0	RT	RT	RT	RT	RT	RT
236	684	4.0	RT	RT	RT	RT	RT	RT
237	685	9.0	KD	KD	KD	KD	KD	KD
238	687	9.0	H	KD	H	H	H	H
239	693	7.0	KD	H	H	KD	H	H
240	694	4.0	RT	RT	RT	RT	RT	RT
241	669	9.0	H	H	H	H	H	H
242	702	9.0	KD	H	KD	KD	KD	KD
243	703	9.0	KD	KD	KD	KD	KD	KD
244	705	8.0	KD	KD	KD	KD	KD	KD
245	706	9.0	H	H	H	H	H	H
246	709	9.0	H	H	H	H	H	H
247	713	8.0	H	H	H	H	H	H
248	715	9.0	H	H	H	H	H	H
249	716	9.0	KD	KD	KD	KD	RT	KD
250	717	7.0	H	H	H	H	H	H
251	718	9.0	H	H	H	H	H	H
252	720	3.0	RT	RT	RT	RT	RT	RT
253	722	9.0	KD	H	KD	KD	H	H
254	725	9.0	KD	KD	KD	KD	KD	KD
255	727	1.0	RT	RT	RT	RT	RT	RT
256	730	5.0	KD	KD	KD	KD	KD	KD
257	731	9.0	KD	KD	KD	KD	KD	KD
258	734	5.0	RT	RT	RT	RT	RT	RT
259	738	3.0	RT	RT	RT	RT	RT	RT
260	740	9.0	H	H	H	H	H	H
261	743	9.0	RT	H	RT	RT	RT	RT
262	745	3.0	RT	H	RT	RT	RT	RT
263	746	9.0	H	H	H	H	H	H
264	748	9.0	KD	KD	KD	KD	KD	KD
265	751	9.0	KD	KD	KD	KD	KD	KD
266	754	4.0	RT	RT	RT	RT	RT	RT
267	761	9.0	H	H	H	H	H	H
268	764	9.0	KD	KD	KD	KD	KD	KD
269	767	9.0	KD	KD	KD	KD	KD	KD
270	768	9.0	H	H	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
271	770	9.0	H	H	H	H	H	H
272	772	9.0	H	H	H	H	H	H
273	773	9.0	KD	KD	KD	KD	KD	KD
274	777	9.0	KD	H	KD	KD	H	KD
275	781	7.0	H	H	H	H	H	H
276	783	9.0	H	H	H	H	H	H
277	784	9.0	KD	KD	KD	KD	KD	KD
278	786	9.0	H	H	H	H	H	H
279	787	9.0	KD	KD	KD	KD	KD	KD
280	788	7.0	H	RT	H	H	RT	RT
281	789	9.0	KD	KD	KD	KD	KD	KD
282	791	5.0	RT	RT	RT	RT	RT	RT
283	104	9.0	KD	KD	KD	KD	KD	KD
284	127	9.0	KD	KD	KD	KD	KD	KD
285	138	9.0	H	H	H	H	H	H
286	163	9.0	KD	KD	KD	KD	KD	KD
287	177	9.0	KD	KD	KD	KD	KD	KD
288	183	9.0	H	H	H	H	H	H
289	191	9.0	H	H	H	H	H	H
290	193	9.0	KD	KD	KD	KD	KD	KD
291	200	9.0	KD	KD	KD	KD	KD	KD
292	205	4.0	H	H	H	H	H	H
293	216	9.0	KD	KD	KD	KD	KD	KD
294	217	3.0	RT	RT	RT	RT	RT	RT
295	223	9.0	KD	KD	KD	KD	KD	KD
296	244	6.0	H	-	H	H	H	H
297	252	3.0	H	H	H	H	H	H
298	314	4.0	H	H	H	H	H	H
299	342	4.0	KD	H	KD	KD	H	H
300	343	3.0	H	H	H	H	H	H
301	344	4.0	H	H	H	H	H	H
302	345	2.0	RT	RT	RT	RT	RT	RT
303	346	4.0	H	H	H	H	H	H
304	352	4.0	H	H	H	H	H	H
305	371	3.0	RT	RT	RT	RT	RT	RT
306	397	9.0	H	H	H	H	H	H
307	396	9.0	H	H	H	H	H	H
308	412	9.0	KD	KD	KD	KD	KD	KD
309	415	9.0	H	H	H	H	H	H

Appendix Table 3 (Continued)

No.	Entr	Score	RM8101	RM3353	RM588	RM190	RM589	RM586
310	438	9.0	H	RT	H	H	H	H
311	439	9.0	RT	H	RT	RT	RT	RT
312	442	9.0	H	H	H	H	H	H
313	443	9.0	H	H	H	H	H	H
314	445	5.0	RT	H	RT	RT	RT	RT
315	454	9.0	KD	KD	KD	KD	KD	KD
316	456	7.0	RT	RT	RT	RT	RT	RT
317	457	9.0	H	H	H	H	H	H
318	458	9.0	H	H	H	H	H	H
319	460	9.0	H	H	H	H	H	H
320	486	4.0	RT	H	RT	RT	RT	RT
321	491	6.0	H	H	H	H	H	H
322	492	5.0	H	H	H	H	H	H
323	493	4.0	RT	RT	-	RT	RT	RT
324	496	9.0	H	H	H	H	H	H
325	500	4.0	RT	RT	RT	RT	RT	RT
326	501	4.0	RT	RT	RT	RT	RT	RT
327	502	9.0	H	H	H	H	H	H
328	511	1.0	RT	RT	RT	RT	RT	RT
329	515	3.0	RT	RT	RT	RT	RT	RT
330	522	3.0	H	KD	H	H	H	H
331	528	9.0	KD	KD	KD	KD	KD	KD
332	529	9.0	-	H	H	-	H	H
333	540	4.0	H	H	H	H	H	H

Appendix Table 4 Validation of the phenotype and genotype of 330 BC_3F_5 individuals from the cross of Rathu Heenati×KDML105 for BPH resistance using modified mass tiller screening. The SSR markers linked to BPH resistance locus on chromosome 6 and 4 were used.

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
1	UBN03078-101-342-4-1	5.0	MR	H	RH
2	UBN03078-101-342-4-2	9.0	S	KD	RH
3	UBN03078-101-342-4-4	5.0	MR	H	RH
4	UBN03078-101-342-4-5	7.0	MR	H	RH
5	UBN03078-101-342-4-6	9.0	S	KD	RH
6	UBN03078-101-342-4-7	9.0	S	H	RH
7	UBN03078-101-342-4-8	9.0	S	KD	RH
8	UBN03078-101-342-4-9	5.0	MR	H	RH
9	UBN03078-101-342-4-10	9.0	S	KD	RH
10	UBN03078-101-342-4-11	9.0	S	KD	RH
11	UBN03078-101-342-4-12	3.0	R	RH	RH
12	UBN03078-101-342-4-13	5.0	MR	H	RH
13	UBN03078-101-342-4-14	3.0	R	RH	RH
14	UBN03078-101-342-4-17	9.0	S	H	RH
15	UBN03078-101-342-4-18	9.0	S	H	RH
16	UBN03078-101-342-4-19	5.0	MR	H	RH
17	UBN03078-101-342-4-21	5.0	MR	H	RH
18	UBN03078-101-342-4-22	9.0	S	H	RH
19	UBN03078-101-342-4-23	7.0	MR	H	RH
20	UBN03078-101-342-4-24	9.0	S	KD	RH
21	UBN03078-101-342-4-25	5.0	MR	H	RH
22	UBN03078-101-342-4-27	7.0	MR	H	RH
23	UBN03078-101-342-4-28	5.0	MR	H	RH
24	UBN03078-101-342-4-29	3.0	R	RH	RH
25	UBN03078-101-342-4-30	3.0	R	RH	RH
26	UBN03078-101-342-4-31	7.0	MR	H	RH
27	UBN03078-101-342-4-33	5.0	MR	H	RH
28	UBN03078-101-342-4-34	9.0	S	KD	RH
29	UBN03078-101-342-4-35	7.0	MR	H	RH
30	UBN03078-101-342-4-36	3.0	R	RH	RH
31	UBN03078-101-342-4-37	7.0	MR	H	RH
32	UBN03078-101-342-4-38	9.0	S	KD	RH
33	UBN03078-101-342-4-39	7.0	MR	H	RH
34	UBN03078-101-342-4-40	5.0	MR	H	RH
35	UBN03078-101-342-4-41	7.0	MR	H	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
36	UBN03078-101-342-4-42	7.0	MR	H	RH
37	UBN03078-101-342-4-43	9.0	S	KD	RH
38	UBN03078-101-342-4-44	5.0	MR	H	RH
39	UBN03078-101-342-4-45	5.0	MR	H	RH
40	UBN03078-101-342-4-46	9.0	S	KD	RH
41	UBN03078-101-342-4-48	7.0	MR	H	RH
42	UBN03078-101-342-4-50	5.0	MR	RH	RH
43	UBN03078-101-342-4-51	9.0	S	KD	RH
44	UBN03078-101-342-4-52	9.0	S	KD	RH
45	UBN03078-101-342-4-54	3.0	R	RH	RH
46	UBN03078-101-342-4-55	7.0	MR	H	RH
47	UBN03078-101-342-4-56	5.0	MR	H	RH
48	UBN03078-101-342-4-58	7.0	MR	H	RH
49	UBN03078-101-342-4-59	7.0	MR	H	RH
50	UBN03078-101-342-4-62	5.0	MR	H	RH
51	UBN03078-101-342-4-63	9.0	S	KD	RH
52	UBN03078-101-342-4-64	3.0	R	RH	RH
53	UBN03078-101-342-4-65	7.0	MR	KD	RH
54	UBN03078-101-342-4-66	5.0	MR	H	RH
55	UBN03078-101-342-4-69	5.0	MR	H	RH
56	UBN03078-101-342-4-70	5.0	MR	H	RH
57	UBN03078-101-342-4-71	5.0	MR	H	RH
58	UBN03078-101-342-4-72	9.0	S	KD	RH
59	UBN03078-101-342-4-73	9.0	S	KD	RH
60	UBN03078-101-342-4-74	7.0	MR	H	RH
61	UBN03078-101-342-4-75	3.0	R	RH	RH
62	UBN03078-101-342-4-76	9.0	S	KD	RH
63	UBN03078-101-342-4-78	5.0	MR	H	RH
64	UBN03078-101-342-4-79	3.0	R	RH	RH
65	UBN03078-101-342-4-81	7.0	MR	H	RH
66	UBN03078-101-342-4-82	3.0	R	RH	RH
67	UBN03078-101-342-4-87	7.0	MR	KD	RH
68	UBN03078-101-342-4-89	9.0	S	KD	RH
69	UBN03078-101-342-4-106	7.0	MR	H	RH
70	UBN03078-101-342-4-107	7.0	MR	H	RH
71	UBN03078-101-342-4-108	9.0	S	KD	RH
72	UBN03078-101-342-4-110	5.0	MR	H	RH
73	UBN03078-101-342-4-111	7.0	MR	H	RH
74	UBN03078-101-342-4-112	9.0	S	KD	RH
75	UBN03078-101-342-4-113	5.0	MR	RH	RH
76	UBN03078-101-342-4-114	5.0	MR	H	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
77	UBN03078-101-342-4-115	5.0	MR	H	RH
78	UBN03078-101-342-4-116	5.0	MR	RH	RH
79	UBN03078-101-342-4-118	9.0	S	KD	RH
80	UBN03078-101-342-4-119	7.0	MR	H	RH
81	UBN03078-101-342-4-120	7.0	MR	H	RH
82	UBN03078-101-342-4-121	9.0	S	KD	RH
83	UBN03078-101-342-4-122	9.0	S	KD	RH
84	UBN03078-101-342-4-123	9.0	S	KD	RH
85	UBN03078-101-342-4-124	9.0	S	KD	RH
86	UBN03078-101-342-4-125	5.0	MR	H	RH
87	UBN03078-101-342-4-126	7.0	MR	H	RH
88	UBN03078-101-342-4-127	7.0	MR	H	RH
89	UBN03078-101-342-4-128	5.0	MR	H	RH
90	UBN03078-101-342-4-130	9.0	S	KD	RH
91	UBN03078-101-342-4-131	5.0	MR	H	RH
92	UBN03078-101-342-4-132	9.0	S	KD	RH
93	UBN03078-101-342-4-133	5.0	MR	H	RH
94	UBN03078-101-342-4-134	5.0	MR	H	RH
95	UBN03078-101-342-4-135	9.0	S	KD	RH
96	UBN03078-101-342-4-140	3.0	R	RH	RH
97	UBN03078-101-342-4-141	5.0	MR	H	RH
98	UBN03078-101-342-4-142	5.0	MR	H	RH
99	UBN03078-101-342-4-143	5.0	MR	H	RH
100	UBN03078-101-342-4-144	5.0	MR	H	RH
101	UBN03078-101-342-4-145	5.0	MR	H	RH
102	UBN03078-101-342-4-146	5.0	MR	H	RH
103	UBN03078-101-342-4-147	7.0	MR	KD	RH
104	UBN03078-101-342-4-148	5.0	MR	H	RH
105	UBN03078-101-342-4-149	7.0	MR	KD	RH
106	UBN03078-101-342-4-150	3.0	R	RH	RH
107	UBN03078-101-342-4-151	7.0	MR	KD	RH
108	UBN03078-101-342-4-152	5.0	MR	H	RH
109	UBN03078-101-342-4-154	5.0	MR	H	RH
110	UBN03078-101-342-4-155	5.0	MR	H	RH
111	UBN03078-101-342-4-156	3.0	R	RH	RH
112	UBN03078-101-342-4-158	3.0	R	RH	RH
113	UBN03078-101-342-4-159	9.0	S	KD	RH
114	UBN03078-101-342-4-160	3.0	R	RH	RH
115	UBN03078-101-342-4-161	3.0	R	H	RH
116	UBN03078-101-342-4-162	3.0	R	RH	RH
117	UBN03078-101-342-4-163	3.0	R	RH	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
118	UBN03078-101-342-4-164	3.0	R	RH	RH
119	UBN03078-101-342-4-165	5.0	MR	H	RH
120	UBN03078-101-342-4-166	9.0	S	KD	RH
121	UBN03078-101-342-4-167	3.0	R	RH	RH
122	UBN03078-101-342-4-168	5.0	MR	H	RH
123	UBN03078-101-342-4-169	5.0	MR	H	RH
124	UBN03078-101-342-4-170	3.0	R	RH	RH
125	UBN03078-101-342-4-171	7.0	MR	H	RH
126	UBN03078-101-342-4-172	5.0	MR	RH	RH
127	UBN03078-101-342-4-173	7.0	MR	H	RH
128	UBN03078-101-342-4-174	5.0	MR	RH	RH
129	UBN03078-101-342-4-175	9.0	S	KD	RH
130	UBN03078-101-342-4-176	5.0	MR	H	RH
131	UBN03078-101-342-4-177	9.0	S	KD	RH
132	UBN03078-101-342-4-178	5.0	MR	H	RH
133	UBN03078-101-342-4-179	3.0	R	H	RH
134	UBN03078-101-342-4-180	3.0	R	H	RH
135	UBN03078-101-342-4-181	3.0	R	RH	RH
136	UBN03078-101-342-4-182	9.0	S	KD	RH
137	UBN03078-101-342-4-183	7.0	MR	KD	RH
138	UBN03078-101-342-4-184	7.0	MR	KD	RH
139	UBN03078-101-342-4-187	7.0	MR	H	RH
140	UBN03078-101-342-4-188	9.0	S	KD	RH
141	UBN03078-101-342-4-189	9.0	S	KD	RH
142	UBN03078-101-342-4-190	5.0	MR	H	RH
143	UBN03078-101-342-4-192	5.0	MR	H	RH
144	UBN03078-101-342-4-193	3.0	R	H	RH
145	UBN03078-101-342-4-194	5.0	MR	H	RH
146	UBN03078-101-342-4-195	9.0	S	KD	RH
147	UBN03078-101-342-4-196	5.0	MR	H	RH
148	UBN03078-101-342-4-197	9.0	S	KD	RH
149	UBN03078-101-342-4-198	3.0	R	H	RH
150	UBN03078-101-342-4-200	9.0	S	KD	RH
151	UBN03078-101-342-4-203	9.0	S	KD	RH
152	UBN03078-101-342-4-204	3.0	R	RH	RH
153	UBN03078-101-342-4-205	3.0	R	RH	RH
154	UBN03078-101-342-4-207	3.0	R	H	RH
155	UBN03078-101-342-4-208	3.0	R	RH	RH
156	UBN03078-101-342-4-210	9.0	S	KD	RH
157	UBN03078-101-342-4-212	9.0	S	KD	RH
158	UBN03078-101-342-4-213	5.0	MR	H	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
159	UBN03078-101-342-4-214	3.0	R	RH	RH
160	UBN03078-101-342-4-215	5.0	MR	RH	RH
161	UBN03078-101-342-4-218	9.0	S	KD	RH
162	UBN03078-101-342-4-219	5.0	MR	RH	RH
163	UBN03078-101-342-4-221	9.0	S	KD	RH
164	UBN03078-101-342-4-222	5.0	MR	H	RH
165	UBN03078-101-342-4-223	9.0	S	KD	RH
166	UBN03078-101-342-4-224	9.0	S	KD	RH
167	UBN03078-101-342-4-225	3.0	R	RH	RH
168	UBN03078-101-342-4-226	5.0	MR	H	RH
169	UBN03078-101-342-4-227	3.0	R	RH	RH
170	UBN03078-101-342-4-228	5.0	MR	H	RH
171	UBN03078-101-342-4-229	3.0	R	RH	RH
172	UBN03078-101-342-4-231	9.0	S	KD	RH
173	UBN03078-101-342-4-232	5.0	MR	H	RH
174	UBN03078-101-342-4-233	7.0	MR	H	RH
175	UBN03078-101-342-4-234	9.0	S	KD	RH
176	UBN03078-101-342-4-235	9.0	S	KD	RH
177	UBN03078-101-342-4-236	5.0	MR	H	RH
178	UBN03078-101-342-4-237	3.0	R	RH	RH
179	UBN03078-101-342-4-239	5.0	MR	H	RH
180	UBN03078-101-342-4-240	5.0	MR	H	RH
181	UBN03078-101-342-4-241	5.0	MR	H	RH
182	UBN03078-101-342-4-243	5.0	MR	H	RH
183	UBN03078-101-342-4-244	5.0	MR	H	RH
184	UBN03078-101-342-4-246	9.0	S	KD	RH
185	UBN03078-101-342-4-248	7.0	MR	H	RH
186	UBN03078-101-342-4-249	5.0	MR	H	RH
187	UBN03078-101-342-4-250	5.0	MR	H	RH
188	UBN03078-101-342-4-251	5.0	MR	H	RH
189	UBN03078-101-342-4-254	9.0	S	KD	RH
190	UBN03078-101-342-4-255	9.0	S	KD	RH
191	UBN03078-101-342-4-257	5.0	MR	H	RH
192	UBN03078-101-342-4-258	7.0	MR	H	RH
193	UBN03078-101-342-4-259	9.0	S	KD	RH
194	UBN03078-101-342-4-260	9.0	S	KD	RH
195	UBN03078-101-342-4-261	5.0	MR	H	RH
196	UBN03078-101-342-4-262	5.0	MR	H	RH
197	UBN03078-101-342-4-264	7.0	MR	H	RH
198	UBN03078-101-342-4-266	7.0	MR	H	RH
199	UBN03078-101-342-4-267	3.0	R	RH	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
200	UBN03078-101-342-4-268	7.0	MR	H	RH
201	UBN03078-101-342-4-269	5.0	MR	H	RH
202	UBN03078-101-342-4-271	5.0	MR	H	RH
203	UBN03078-101-342-4-272	5.0	MR	H	RH
204	UBN03078-101-342-4-273	5.0	MR	H	RH
205	UBN03078-101-342-4-274	5.0	MR	H	RH
206	UBN03078-101-342-4-280	7.0	MR	H	RH
207	UBN03078-101-342-4-281	5.0	MR	H	RH
208	UBN03078-101-342-4-282	3.0	R	RH	RH
209	UBN03078-101-342-4-283	3.0	R	RH	RH
210	UBN03078-101-342-4-284	5.0	MR	H	RH
211	UBN03078-101-342-4-285	5.0	MR	H	RH
212	UBN03078-101-342-4-286	5.0	MR	H	RH
213	UBN03078-101-342-4-287	5.0	MR	H	RH
214	UBN03078-101-342-4-288	3.0	R	RH	RH
215	UBN03078-101-342-4-289	5.0	MR	H	RH
216	UBN03078-101-342-4-300	3.0	R	RH	RH
217	UBN03078-101-342-4-301	5.0	MR	H	RH
218	UBN03078-101-342-6-302	3.0	R	RH	RH
219	UBN03078-101-342-6-304	9.0	S	KD	RH
220	UBN03078-101-342-6-305	5.0	MR	H	RH
221	UBN03078-101-342-6-306	5.0	MR	H	RH
222	UBN03078-101-342-6-307	5.0	MR	H	RH
223	UBN03078-101-342-6-308	3.0	R	RH	RH
224	UBN03078-101-342-6-311	7.0	MR	H	RH
225	UBN03078-101-342-6-312	7.0	MR	H	RH
226	UBN03078-101-342-6-313	7.0	MR	H	RH
227	UBN03078-101-342-6-314	5.0	MR	H	RH
228	UBN03078-101-342-6-315	5.0	MR	H	RH
229	UBN03078-101-342-6-316	9.0	S	KD	RH
230	UBN03078-101-342-6-317	3.0	R	RH	RH
231	UBN03078-101-342-6-318	3.0	R	RH	RH
232	UBN03078-101-342-6-319	5.0	MR	H	RH
233	UBN03078-101-342-6-322	9.0	S	KD	RH
234	UBN03078-101-342-6-323	5.0	MR	H	RH
235	UBN03078-101-342-6-324	5.0	MR	H	RH
236	UBN03078-101-342-6-325	3.0	R	RH	RH
237	UBN03078-101-342-6-326	3.0	R	RH	RH
238	UBN03078-101-342-6-327	3.0	R	RH	RH
239	UBN03078-101-342-6-328	9.0	S	KD	RH
240	UBN03078-101-342-6-329	3.0	R	RH	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
241	UBN03078-101-342-6-330	5.0	MR	H	RH
242	UBN03078-101-342-6-331	9.0	S	KD	RH
243	UBN03078-101-342-6-332	5.0	MR	H	RH
244	UBN03078-101-342-6-333	5.0	MR	H	RH
245	UBN03078-101-342-6-335	9.0	S	KD	RH
246	UBN03078-101-342-6-338	3.0	R	RH	RH
247	UBN03078-101-342-6-339	7.0	MR	KD	RH
248	UBN03078-101-342-6-340	9.0	S	KD	RH
249	UBN03078-101-342-6-341	5.0	MR	H	RH
250	UBN03078-101-342-6-342	9.0	S	KD	RH
251	UBN03078-101-342-6-343	9.0	S	KD	RH
252	UBN03078-101-342-6-345	5.0	MR	H	RH
253	UBN03078-101-342-6-346	3.0	R	RH	RH
254	UBN03078-101-342-6-347	7.0	MR	H	RH
255	UBN03078-101-342-6-348	7.0	MR	H	RH
256	UBN03078-101-342-6-349	7.0	MR	H	RH
257	UBN03078-101-342-6-350	5.0	MR	H	RH
258	UBN03078-101-342-6-351	7.0	MR	H	RH
259	UBN03078-101-342-6-352	7.0	MR	H	RH
260	UBN03078-101-342-6-353	7.0	MR	H	RH
261	UBN03078-101-342-6-354	9.0	S	KD	RH
262	UBN03078-101-342-6-355	9.0	S	KD	RH
263	UBN03078-101-342-6-357	5.0	MR	RH	RH
264	UBN03078-101-342-6-358	7.0	MR	H	RH
265	UBN03078-101-342-6-359	9.0	S	H	RH
266	UBN03078-101-342-6-361	9.0	S	KD	RH
267	UBN03078-101-342-6-362	3.0	R	RH	RH
268	UBN03078-101-342-6-363	5.0	MR	RH	RH
269	UBN03078-101-342-6-364	9.0	S	KD	RH
270	UBN03078-101-342-6-365	3.0	R	RH	RH
271	UBN03078-101-342-6-367	9.0	S	KD	RH
272	UBN03078-101-342-6-368	3.0	R	RH	RH
273	UBN03078-101-342-6-371	3.0	R	RH	RH
274	UBN03078-101-342-6-372	7.0	MR	H	RH
275	UBN03078-101-342-6-373	3.0	R	RH	RH
276	UBN03078-101-342-6-374	7.0	MR	H	RH
277	UBN03078-101-342-6-375	3.0	R	RH	RH
278	UBN03078-101-342-6-376	9.0	S	KD	RH
279	UBN03078-101-342-6-377	7.0	MR	H	RH
280	UBN03078-101-342-6-378	7.0	MR	H	RH
281	UBN03078-101-342-6-379	7.0	MR	H	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
282	UBN03078-101-342-6-380	3.0	R	RH	RH
283	UBN03078-101-342-6-381	7.0	MR	H	RH
284	UBN03078-101-342-6-382	3.0	R	RH	RH
285	UBN03078-101-342-6-383	7.0	MR	H	RH
286	UBN03078-101-342-6-384	5.0	MR	H	RH
287	UBN03078-101-342-6-386	3.0	R	RH	RH
288	UBN03078-101-342-6-387	3.0	R	RH	RH
289	UBN03078-101-342-6-389	3.0	R	RH	RH
290	UBN03078-101-342-6-391	7.0	MR	H	RH
291	UBN03078-101-342-6-392	3.0	R	RH	RH
292	UBN03078-101-342-6-393	3.0	R	RH	RH
293	UBN03078-101-342-6-394	9.0	S	KD	RH
294	UBN03078-101-342-6-395	3.0	R	RH	RH
295	UBN03078-101-342-6-396	5.0	MR	H	RH
296	UBN03078-101-342-6-398	5.0	MR	H	RH
297	UBN03078-101-342-6-399	9.0	S	KD	RH
298	UBN03078-101-342-6-401	5.0	MR	H	RH
299	UBN03078-101-342-4-406	5.0	MR	H	RH
300	UBN03078-101-342-4-407	9.0	S	KD	RH
301	UBN03078-101-342-4-408	9.0	S	KD	RH
302	UBN03078-101-342-4-409	3.0	R	RH	RH
303	UBN03078-101-342-4-410	3.0	R	RH	RH
304	UBN03078-101-342-4-411	9.0	S	KD	RH
305	UBN03078-101-342-4-412	9.0	S	KD	RH
306	UBN03078-101-342-4-413	7.0	MR	H	RH
307	UBN03078-101-342-4-414	9.0	S	KD	RH
308	UBN03078-101-342-4-415	3.0	R	RH	RH
309	UBN03078-101-342-4-416	3.0	R	RH	RH
310	UBN03078-101-342-4-418	3.0	R	RH	RH
311	UBN03078-101-342-4-420	7.0	MR	H	RH
312	UBN03078-101-342-4-421	5.0	MR	H	RH
313	UBN03078-101-342-4-422	9.0	S	KD	RH
314	UBN03078-101-342-4-423	3.0	R	RH	RH
315	UBN03078-101-342-4-428	5.0	MR	H	RH
316	UBN03078-101-342-4-430	3.0	R	RH	RH
317	UBN03078-101-342-4-431	9.0	S	KD	RH
318	UBN03078-101-342-4-433	7.0	MR	H	RH
319	UBN03078-101-342-4-434	9.0	S	KD	RH
320	UBN03078-101-342-4-436	5.0	MR	RH	RH
321	UBN03078-101-342-4-437	3.0	R	RH	RH
322	UBN03078-101-342-4-438	7.0	MR	H	RH

Appendix Table 4 (Continued)

ENTNO	DESIGNATION	BPH Scoring		RM589	RM261
323	UBN03078-101-342-4-440	9.0	S	KD	RH
324	UBN03078-101-342-4-441	5.0	MR	H	RH
325	UBN03078-101-342-4-442	7.0	MR	H	RH
326	UBN03078-101-342-4-443	5.0	MR	H	RH
327	UBN03078-101-342-4-444	5.0	MR	H	RH
328	UBN03078-101-342-4-446	9.0	S	KD	RH
329	UBN03078-101-342-4-448	9.0	S	KD	RH
330	UBN03078-101-342-4-449	9.0	S	KD	RH

Appendix Table 5 The 75 simple sequence repeat (SSR) and sequence tagged site (STS) markers assembled to verify genomic background of the selected introgression lines.

No	Designation	RM579	RM3627	RM140	RM1152	RM226	RM3340	RM3688	RM3515	RM6295	RM240
1	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
2	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
4	UBN03078-80-354-7	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
5	UBN03078-80-354-7	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
7	UBN03078-80-354-12	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
8	UBN03078-80-354-15	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
9	UBN03078-80-354-16	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
10	UBN03078-101-342-4-19	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
11	UBN03078-101-342-4-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
12	UBN03078-101-342-4-32	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
14	UBN03078-101-342-4-106	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
15	UBN03078-101-342-4-111	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
16	UBN03078-101-342-4-114	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
17	UBN03078-101-342-4-141	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
18	UBN03078-101-342-4-143	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
22	UBN03078-101-342-6-49	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
23	UBN03078-101-342-6-56	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
24	UBN03078-101-342-6-58	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM579	RM3627	RM140	RM1152	RM226	RM3340	RM3688	RM3515	RM6295	RM240
25	UBN03078-101-342-6-82	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
26	UBN03078-101-342-6-89	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
27	UBN03078-101-450-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
28	UBN03078-80-28-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
29	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
30	UBN03078-101-342-16	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
31	UBN03078-101-342-4-16	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
32	UBN03078-101-342-4-24	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
33	UBN03078-101-342-4-97	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
34	UBN03078-101-342-4-126	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
35	UBN03078-101-342-4-135	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
36	UBN03078-101-342-4-158	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
37	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
38	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
40	UBN03078-101-342-14	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
41	UBN03078-101-342-4-134	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
42	UBN03078-101-342-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
45	UBN03078-101-450-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
46	UBN03078-101-342-4-138	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
47	UBN03078-101-450-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
48	UBN03078-101-60-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
49	UBN03078-80-28-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
50	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM3766	RM3346	RM261	RM227	RM514	RM5548	RM8212	RM241	RM3471	RM401
1	UBN03078-80-354-11	KD	KD	KD	RT	RT	RT	KD	RT	KD	KD
2	UBN03078-80-354-20	KD	RT	KD	RT	RT	RT	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	KD	KD	RT	RT	RT	-	-	KD	KD
4	UBN03078-80-354-7	KD	KD	KD	RT	RT	RT	KD	RT	KD	KD
5	UBN03078-80-354-7	KD	KD	KD	RT	RT	RT	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	H	KD	RT	RT	RT	-	KD	KD	KD
7	UBN03078-80-354-12	KD	RT	KD	RT	RT	RT	KD	KD	KD	KD
8	UBN03078-80-354-15	KD	H	KD	RT	RT	RT	-	KD	KD	KD
9	UBN03078-80-354-16	KD	KD	KD	RT	RT	RT	KD	KD	KD	KD
10	UBN03078-101-342-4-19	KD	H	RT	RT	RT	RT	RT	KD	KD	KD
11	UBN03078-101-342-4-20	KD	KD	RT	RT	RT	RT	RT	RT	KD	KD
12	UBN03078-101-342-4-32	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
14	UBN03078-101-342-4-106	KD	H	RT	RT	RT	RT	RT	RT	KD	KD
15	UBN03078-101-342-4-111	KD	KD	RT	RT	RT	RT	RT	RT	KD	KD
16	UBN03078-101-342-4-114	KD	H	RT	RT	RT	RT	RT	KD	KD	KD
17	UBN03078-101-342-4-141	KD	RT	RT	RT	RT	RT	RT	-	KD	KD
18	UBN03078-101-342-4-143	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
22	UBN03078-101-342-6-49	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
23	UBN03078-101-342-6-56	KD	KD	RT	RT	RT	RT	RT	RT	KD	KD
24	UBN03078-101-342-6-58	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
25	UBN03078-101-342-6-82	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
26	UBN03078-101-342-6-89	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM3766	RM3346	RM261	RM227	RM514	RM5548	RM8212	RM241	RM3471	RM401
27	UBN03078-101-450-2	KD	RT	H	RT	RT	RT	H	RT	KD	KD
28	UBN03078-80-28-1	KD	KD	KD	RT	RT	RT	KD	RT	KD	KD
29	UBN03078-80-28-5	KD	RT	KD	RT	RT	RT	KD	RT	KD	KD
30	UBN03078-101-342-16	KD	KD	RT	RT	RT	RT	RT	RT	KD	KD
31	UBN03078-101-342-4-16	KD	H	RT	RT	RT	RT	RT	RT	KD	KD
32	UBN03078-101-342-4-24	KD	RT	RT	RT	RT	RT	RT	RT	KD	KD
33	UBN03078-101-342-4-97	KD	RT	RT	RT	RT	RT	RT	KD	KD	KD
34	UBN03078-101-342-4-126	KD	H	RT	RT	RT	RT	RT	KD	KD	KD
35	UBN03078-101-342-4-135	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
36	UBN03078-101-342-4-158	KD	RT	RT	RT	RT	RT	RT	KD	KD	KD
37	UBN03078-80-28-5	KD	RT	KD	RT	RT	RT	KD	RT	KD	KD
38	UBN03078-80-28-5	KD	RT	KD	RT	RT	RT	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	H	RT	RT	RT	RT	RT	KD	KD	KD
40	UBN03078-101-342-14	KD	-	RT	RT	RT	RT	RT	KD	KD	KD
41	UBN03078-101-342-4-134	KD	RT	-	RT	RT	RT	RT	KD	KD	KD
42	UBN03078-101-342-2	KD	H	H	RT	RT	RT	RT	RT	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	RT	RT	RT	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
45	UBN03078-101-450-2	KD	H	H	RT	RT	RT	H	KD	KD	KD
46	UBN03078-101-342-4-138	KD	KD	RT	RT	RT	RT	RT	KD	KD	KD
47	UBN03078-101-450-1	KD	H	H	RT	RT	RT	H	-	KD	KD
48	UBN03078-101-60-20	KD	-	RT	RT	RT	RT	H	-	KD	KD
49	UBN03078-80-28-1	KD	-	-	RT	RT	RT	KD	-	KD	KD
50	UBN03078-80-354-20	KD	H	KD	RT	RT	RT	KD	-	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM317	RM6909	MS10	RM159	RM3345	RM267	RM153	RM26	RM3353	RM1369
1	UBN03078-80-354-11	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
2	UBN03078-80-354-20	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
3	UBN03078-81-504-1	KD	KD	RT	RT	KD	KD	KD	KD	RT	H
4	UBN03078-80-354-7	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
5	UBN03078-80-354-7	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
6	UBN03078-80-354-11	KD	KD	RT	H	KD	KD	KD	KD	H	KD
7	UBN03078-80-354-12	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
8	UBN03078-80-354-15	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
9	UBN03078-80-354-16	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
10	UBN03078-101-342-4-19	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
11	UBN03078-101-342-4-20	KD	KD	RT	H	KD	KD	KD	KD	H	KD
12	UBN03078-101-342-4-32	KD	KD	RT	H	KD	KD	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
14	UBN03078-101-342-4-106	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
15	UBN03078-101-342-4-111	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
16	UBN03078-101-342-4-114	KD	KD	RT	RT	KD	KD	KD	KD	KD	KD
17	UBN03078-101-342-4-141	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
18	UBN03078-101-342-4-143	KD	KD	RT	RT	KD	KD	KD	KD	H	KD
19	UBN03078-101-342-4-144	KD	KD	RT	RT	KD	KD	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	RT	RT	KD	KD	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	RT	H	KD	KD	KD	KD	H	KD
22	UBN03078-101-342-6-49	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
23	UBN03078-101-342-6-56	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
24	UBN03078-101-342-6-58	KD	KD	RT	-	KD	KD	KD	KD	H	KD
25	UBN03078-101-342-6-82	KD	KD	RT	KD	KD	KD	KD	KD	RT	KD
26	UBN03078-101-342-6-89	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD

Appendix Table 5 (Continued)

No	Designation	RM317	RM6909	MS10	RM159	RM3345	RM267	RM153	RM26	RM3353	RM1369
27	UBN03078-101-450-2	KD	KD	RT	H	KD	KD	KD	KD	RT	H
28	UBN03078-80-28-1	KD	KD	RT	H	KD	KD	KD	KD	RT	H
29	UBN03078-80-28-5	KD	KD	RT	H	KD	KD	KD	KD	RT	H
30	UBN03078-101-342-16	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
31	UBN03078-101-342-4-16	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
32	UBN03078-101-342-4-24	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
33	UBN03078-101-342-4-97	KD	KD	RT	RT	KD	KD	KD	KD	-	KD
34	UBN03078-101-342-4-126	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
35	UBN03078-101-342-4-135	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
36	UBN03078-101-342-4-158	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
37	UBN03078-80-28-5	KD	KD	RT	RT	KD	KD	KD	KD	RT	H
38	UBN03078-80-28-5	KD	KD	RT	RT	KD	KD	KD	KD	RT	H
39	UBN03078-101-342-9	KD	KD	RT	RT	KD	KD	KD	KD	RT	KD
40	UBN03078-101-342-14	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
41	UBN03078-101-342-4-134	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
42	UBN03078-101-342-2	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
43	UBN03078-101-450-1	KD	KD	RT	H	KD	KD	KD	KD	RT	H
44	UBN03078-101-342-11	KD	KD	RT	H	KD	KD	KD	KD	RT	KD
45	UBN03078-101-450-2	KD	KD	RT	H	KD	KD	KD	KD	RT	H
46	UBN03078-101-342-4-138	KD	KD	RT	H	KD	KD	KD	KD	H	KD
47	UBN03078-101-450-1	KD	KD	RT	H	KD	KD	KD	KD	RT	H
48	UBN03078-101-60-20	KD	KD	RT	-	KD	KD	KD	KD	RT	KD
49	UBN03078-80-28-1	KD	KD	RT	KD	KD	KD	KD	KD	RT	H
50	UBN03078-80-354-20	KD	KD	RT	H	KD	KD	KD	KD	RT	KD

Appendix Table 5 (Continued)

No	Designation	RM30	RM400	RM402	RM508	GT11	RM589	RM586	RM588	RM190	RM3555
1	UBN03078-80-354-11	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
2	UBN03078-80-354-20	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
4	UBN03078-80-354-7	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
5	UBN03078-80-354-7	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
7	UBN03078-80-354-12	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
8	UBN03078-80-354-15	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
9	UBN03078-80-354-16	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
10	UBN03078-101-342-4-19	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
11	UBN03078-101-342-4-20	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
12	UBN03078-101-342-4-32	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
14	UBN03078-101-342-4-106	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
15	UBN03078-101-342-4-111	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
16	UBN03078-101-342-4-114	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
17	UBN03078-101-342-4-141	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
18	UBN03078-101-342-4-143	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
22	UBN03078-101-342-6-49	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
23	UBN03078-101-342-6-56	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
24	UBN03078-101-342-6-58	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
25	UBN03078-101-342-6-82	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
26	UBN03078-101-342-6-89	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM30	RM400	RM402	RM508	GT11	RM589	RM586	RM588	RM190	RM3555
27	UBN03078-101-450-2	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
28	UBN03078-80-28-1	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
29	UBN03078-80-28-5	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
30	UBN03078-101-342-16	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
31	UBN03078-101-342-4-16	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
32	UBN03078-101-342-4-24	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
33	UBN03078-101-342-4-97	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
34	UBN03078-101-342-4-126	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
35	UBN03078-101-342-4-135	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
36	UBN03078-101-342-4-158	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
37	UBN03078-80-28-5	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
38	UBN03078-80-28-5	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
40	UBN03078-101-342-14	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
41	UBN03078-101-342-4-134	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
42	UBN03078-101-342-2	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
45	UBN03078-101-450-2	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
46	UBN03078-101-342-4-138	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
47	UBN03078-101-450-1	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
48	UBN03078-101-60-20	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
49	UBN03078-80-28-1	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD
50	UBN03078-80-354-20	KD	KD	KD	RT	KD	RT	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM3583	RM8257	RM172	RM331	CPO4133	RM544	RM407	RM210	RM331	RM6966
1	UBN03078-80-354-11	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
2	UBN03078-80-354-20	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
4	UBN03078-80-354-7	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
5	UBN03078-80-354-7	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
7	UBN03078-80-354-12	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
8	UBN03078-80-354-15	KD	KD	KD	-	KD	KD	KD	KD	KD	KD
9	UBN03078-80-354-16	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
10	UBN03078-101-342-4-19	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
11	UBN03078-101-342-4-20	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
12	UBN03078-101-342-4-32	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
14	UBN03078-101-342-4-106	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
15	UBN03078-101-342-4-111	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
16	UBN03078-101-342-4-114	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
17	UBN03078-101-342-4-141	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
18	UBN03078-101-342-4-143	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
22	UBN03078-101-342-6-49	KD	KD	KD	KD	RT	KD	KD	KD	KD	KD
23	UBN03078-101-342-6-56	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
24	UBN03078-101-342-6-58	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
25	UBN03078-101-342-6-82	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
26	UBN03078-101-342-6-89	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM3583	RM8257	RM172	RM331	CPO4133	RM544	RM407	RM210	RM331	RM6966
27	UBN03078-101-450-2	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
28	UBN03078-80-28-1	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
29	UBN03078-80-28-5	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
30	UBN03078-101-342-16	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
31	UBN03078-101-342-4-16	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
32	UBN03078-101-342-4-24	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
33	UBN03078-101-342-4-97	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
34	UBN03078-101-342-4-126	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
35	UBN03078-101-342-4-135	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
36	UBN03078-101-342-4-158	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
37	UBN03078-80-28-5	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
38	UBN03078-80-28-5	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
40	UBN03078-101-342-14	KD	KD	KD	RT	RT	KD	KD	KD	KD	KD
41	UBN03078-101-342-4-134	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
42	UBN03078-101-342-2	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	KD	KD	RT	H	KD	KD	KD	KD	KD
45	UBN03078-101-450-2	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
46	UBN03078-101-342-4-138	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
47	UBN03078-101-450-1	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
48	UBN03078-101-60-20	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
49	UBN03078-80-28-1	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD
50	UBN03078-80-354-20	KD	KD	KD	RT	KD	KD	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	B03	RM444	RM3700	RM2190	RM242	RM3909	RM216	RM591	RM5095	RM6824	RM590
1	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
2	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	KD	KD	KD	KD	KD	RT	KD	H	KD	KD
4	UBN03078-80-354-7	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
5	UBN03078-80-354-7	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
7	UBN03078-80-354-12	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
8	UBN03078-80-354-15	KD	KD	KD	KD	KD	KD	-	KD	KD	H	KD
9	UBN03078-80-354-16	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
10	UBN03078-101-342-4-19	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
11	UBN03078-101-342-4-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
12	UBN03078-101-342-4-32	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
14	UBN03078-101-342-4-106	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
15	UBN03078-101-342-4-111	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
16	UBN03078-101-342-4-114	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
17	UBN03078-101-342-4-141	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
18	UBN03078-101-342-4-143	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
22	UBN03078-101-342-6-49	KD	KD	KD	KD	KD	KD	RT	KD	RT	KD	KD
23	UBN03078-101-342-6-56	KD	KD	KD	KD	KD	KD	KD	KD	RT	KD	KD
24	UBN03078-101-342-6-58	KD	KD	KD	KD	KD	KD	KD	KD	RT	H	KD
25	UBN03078-101-342-6-82	KD	KD	KD	KD	KD	KD	H	KD	RT	H	KD
26	UBN03078-101-342-6-89	KD	KD	KD	KD	KD	KD	KD	KD	RT	KD	KD

Appendix Table 5 (Continued)

No	Designation	B03	RM444	RM3700	RM2190	RM242	RM3909	RM216	RM591	RM5095	RM6824	RM590
27	UBN03078-101-450-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
28	UBN03078-80-28-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
29	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
30	UBN03078-101-342-16	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
31	UBN03078-101-342-4-16	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
32	UBN03078-101-342-4-24	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
33	UBN03078-101-342-4-97	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
34	UBN03078-101-342-4-126	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
35	UBN03078-101-342-4-135	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
36	UBN03078-101-342-4-158	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
37	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
38	UBN03078-80-28-5	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	KD	KD	KD	KD	KD	H	KD	-	KD	KD
40	UBN03078-101-342-14	KD	KD	KD	KD	KD	KD	RT	KD	-	KD	KD
41	UBN03078-101-342-4-134	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
42	UBN03078-101-342-2	KD	KD	KD	KD	KD	KD	H	KD	KD	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	KD	KD	KD	KD	KD	KD	KD	-	KD	KD
45	UBN03078-101-450-2	KD	KD	KD	KD	KD	KD	RT	KD	-	KD	KD
46	UBN03078-101-342-4-138	KD	KD	KD	KD	KD	KD	KD	H	KD	KD	KD
47	UBN03078-101-450-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
48	UBN03078-101-60-20	KD	KD	KD	KD	KD	KD	RT	H	-	H	KD
49	UBN03078-80-28-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	H	KD
50	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	KD	H	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM591	RM6272	RM3717	RM457	RM2191	RM224	RM1103	RM3409	RM101	RM1103	RM3226
1	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
2	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
3	UBN03078-81-504-1	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
4	UBN03078-80-354-7	KD	RT	KD	KD	KD	KD	RT	KD	KD	KD	KD
5	UBN03078-80-354-7	KD	RT	KD	KD	KD	KD	RT	KD	KD	KD	KD
6	UBN03078-80-354-11	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
7	UBN03078-80-354-12	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
8	UBN03078-80-354-15	KD	-	KD	KD	KD	KD	RT	KD	KD	KD	KD
9	UBN03078-80-354-16	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD
10	UBN03078-101-342-4-19	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
11	UBN03078-101-342-4-20	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
12	UBN03078-101-342-4-32	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
13	UBN03078-101-342-4-96	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
14	UBN03078-101-342-4-106	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
15	UBN03078-101-342-4-111	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
16	UBN03078-101-342-4-114	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
17	UBN03078-101-342-4-141	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
18	UBN03078-101-342-4-143	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
19	UBN03078-101-342-4-144	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
20	UBN03078-101-342-4-147	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
21	UBN03078-101-342-4-148	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
22	UBN03078-101-342-6-49	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
23	UBN03078-101-342-6-56	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
24	UBN03078-101-342-6-58	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
25	UBN03078-101-342-6-82	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
26	UBN03078-101-342-6-89	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD

Appendix Table 5 (Continued)

No	Designation	RM591	RM6272	RM3717	RM457	RM2191	RM224	RM1103	RM3409	RM101	RM1103	RM3226
27	UBN03078-101-450-2	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
28	UBN03078-80-28-1	KD	H	KD	KD	KD	KD	H	KD	KD	KD	KD
29	UBN03078-80-28-5	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
30	UBN03078-101-342-16	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
31	UBN03078-101-342-4-16	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
32	UBN03078-101-342-4-24	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
33	UBN03078-101-342-4-97	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
34	UBN03078-101-342-4-126	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
35	UBN03078-101-342-4-135	KD	RT	KD	KD	KD	KD	KD	KD	KD	KD	KD
36	UBN03078-101-342-4-158	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
37	UBN03078-80-28-5	KD	-	KD	KD	KD	KD	H	KD	KD	KD	KD
38	UBN03078-80-28-5	KD	RT	KD	KD	KD	KD	RT	KD	KD	KD	KD
39	UBN03078-101-342-9	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
40	UBN03078-101-342-14	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
41	UBN03078-101-342-4-134	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
42	UBN03078-101-342-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
43	UBN03078-101-450-1	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
44	UBN03078-101-342-11	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
45	UBN03078-101-450-2	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
46	UBN03078-101-342-4-138	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
47	UBN03078-101-450-1	KD	H	KD	KD	KD	KD	KD	KD	KD	KD	KD
48	UBN03078-101-60-20	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD	KD
49	UBN03078-80-28-1	KD	KD	KD	KD	KD	KD	H	KD	KD	KD	KD
50	UBN03078-80-354-20	KD	KD	KD	KD	KD	KD	RT	KD	KD	KD	KD

Appendix Table 6 Per cent recovery of recipient genome of the selected ILs.

No.	Designation	KDML105	Rathu Heenati	Heterozygous
1	UBN03078-80-354-11	88.2	10.3	1.5
2	UBN03078-80-354-20	89.7	10.3	0.0
3	UBN03078-81-504-1	86.4	10.6	3.0
4	UBN03078-80-354-7	88.2	10.3	1.5
5	UBN03078-80-354-7	89.7	8.8	1.5
6	UBN03078-80-354-11	89.6	6.0	4.5
7	UBN03078-80-354-12	88.2	8.8	2.9
8	UBN03078-80-354-15	87.7	9.2	3.1
9	UBN03078-80-354-16	89.7	7.4	2.9
10	UBN03078-101-342-4-19	86.8	8.8	4.4
11	UBN03078-101-342-4-20	85.3	11.8	2.9
12	UBN03078-101-342-4-32	88.2	8.8	2.9
13	UBN03078-101-342-4-96	89.7	8.8	1.5
14	UBN03078-101-342-4-106	83.8	11.8	4.4
15	UBN03078-101-342-4-111	88.2	11.8	0.0
16	UBN03078-101-342-4-114	88.2	8.8	2.9
17	UBN03078-101-342-4-141	83.6	14.9	1.5
18	UBN03078-101-342-4-143	86.8	10.3	2.9
19	UBN03078-101-342-4-144	91.2	8.8	0.0
20	UBN03078-101-342-4-147	88.2	11.8	0.0
21	UBN03078-101-342-4-148	89.7	7.4	2.9
22	UBN03078-101-342-6-49	83.8	13.2	2.9
23	UBN03078-101-342-6-56	83.8	13.2	2.9
24	UBN03078-101-342-6-58	85.1	10.4	4.5
25	UBN03078-101-342-6-82	83.8	11.8	4.4
26	UBN03078-101-342-6-89	86.8	13.2	0.0
27	UBN03078-101-450-2	83.8	8.8	7.4
28	UBN03078-80-28-1	86.8	7.4	5.9
29	UBN03078-80-28-5	86.8	10.3	2.9
30	UBN03078-101-342-16	83.8	14.7	1.5
31	UBN03078-101-342-4-16	83.8	13.2	2.9
32	UBN03078-101-342-4-24	85.3	13.2	1.5
33	UBN03078-101-342-4-97	86.6	10.4	3.0
34	UBN03078-101-342-4-126	86.8	8.8	4.4
35	UBN03078-101-342-4-135	86.8	11.8	1.5
36	UBN03078-101-342-4-158	85.3	11.8	2.9
37	UBN03078-80-28-5	85.1	10.4	4.5
38	UBN03078-80-28-5	86.8	11.8	1.5
39	UBN03078-101-342-9	86.6	10.4	3.0
40	UBN03078-101-342-14	84.8	12.1	3.0
41	UBN03078-101-342-4-134	86.6	9.0	4.5
42	UBN03078-101-342-2	85.3	8.8	5.9
43	UBN03078-101-450-1	91.2	5.9	2.9
44	UBN03078-101-342-11	86.6	9.0	4.5
45	UBN03078-101-450-2	85.1	7.5	7.5
46	UBN03078-101-342-4-138	86.8	7.4	5.9
47	UBN03078-101-450-1	85.1	6.0	9.0
48	UBN03078-101-60-20	85.9	9.4	4.7
49	UBN03078-80-28-1	89.2	6.2	4.6
50	UBN03078-80-354-20	88.1	7.5	4.5

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