

博士論文要約 (Summary)

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連合農学研究科

専攻 Course : Science of Bioresource Production

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タイトル Title	Genetic and breeding studies for brown planthopper (<i>Nilaparvata lugens</i> Stål) resistance in cultivated rice (<i>Oryza sativa</i> L. and <i>Oryza glaberrima</i> Steud.) (栽培イネ (<i>Oryza sativa</i> L.および <i>Oryza glaberrima</i> Steud.) におけるトビイロウンカ (<i>Nilaparvata lugens</i> Stål) 抵抗性の遺伝・育種学的研究)
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Chapter title: Near-isogenic lines for resistance to brown planthopper with the genetic background of Indica Group elite rice (*Oryza sativa* L.) variety ‘IR64’

キーワード Key word (Rice) (marker-assisted selection) (near-isogenic lines) (brown planthopper) (resistance genes)

Introduction and purpose

The brown planthopper (BPH), *Nilaparvata lugens* Stål, is an insect pest that severely damages rice (*Oryza sativa* L.) in Asia, causing huge yield loss. Use of resistant variety is a cost-effective and eco-friendly strategy for maintaining BPH populations below the economic injury level. However, current BPH populations have been changed to virulence against resistant varieties. In this study, to estimate effective combinations among eight BPH resistance genes (*BPH32*, *BPH17-ptb*, *BPH20*, *BPH17*, *BPH3*, *BPH25*, *BPH26* and *qBPH6*), eight near-isogenic lines with the genetic background of an Indica Group rice variety ‘IR64’ (IR64-NIL) were developed using marker-assisted selection.

Material and methods

An elite Indica Group rice variety, ‘IR64’ was used as the recurrent parent to develop NILs with BPH resistance genes, whereas eight T65 NILs (T65-*BPH32*, T65-*BPH17-ptb*, T65-*BPH20*, T65-*BPH17*, T65-*BPH3*, T65-*BPH25*, T65-*BPH26* and T65-*qBPH6*) were used as donor parents. T65-NILs were crossed with ‘IR64’ to develop F₁ plants and the F₁ plants were repeatedly backcrossed. At each generation of backcrossing, plants carrying BPH resistance genes from the donor parents (T65-NILs) were selected through MAS. Among the 384 SSR markers, 236 SSR markers distributed on 12 rice chromosomes showed polymorphism between ‘IR64’ and ‘T65’ and were further applied to detect substituted chromosomal segments from donor parents in the NILs developed. The BPH population, Koshi-2013 was to evaluate the resistance level of the NILs developed using modified seed box screening test (MSST), antibiosis test and honeydew test. We characterized days to heading (DTH), culm length (CL), panicle length (PL), flag leaf length (LL), flag leaf width (LW), and panicle number (PN) as agronomic traits of NILs with ‘IR64’.

Results

Eight NILs, IR64-*BPH32*, IR64-*BPH17-ptb*, IR64-*BPH20*, IR64-*BPH17*, IR64-*BPH3*, IR64-*BPH25*, IR64-*BPH26* and IR64-*qBPH6* were developed in this study. The genome recoveries of these NILs ranged from 89.3% to 98.8% and agronomic traits of them were similar to those of ‘IR64’. In modified seedbox screening test, resistance level of IR64-NILs was higher than that of ‘IR64’. In antibiosis test, high adult mortalities of BPH (from 56.0% to 97.0%) were observed among NILs, in comparison with that of ‘IR64’. In the honeydew test, less honeydew excretion of BPH (from 3.4 to 59.2 mm²) was observed among NILs with some exceptions, IR64-*BPH17-ptb* and IR64-*qBPH6*, in comparison with that of ‘IR64’. Among IR64-NILs, the line carrying *BPH17* showed the highest resistance level at all tests.

Conclusion and consideration

We incorporated BPH resistance genes from T65-NILs into the elite variety ‘IR64’, using marker-assisted backcross breeding to develop NILs. Among the pre-NILs developed in this study, three pre-NILs (IR64-*BPH17*, IR64-*BPH3* and IR64-*BPH26*) had significantly enhanced BPH resistance levels. The IR64-NILs are unique because each NIL has at least three BPH resistance genes. Thus, these IR64-NILs with multiple BPH resistance genes could be valuable breeding lines for enhancing resistance levels by gene pyramiding and multiline variety.

Chapter title: Development of pyramided lines carrying brown planthopper resistance genes in the genetic background of Indica Group rice (*Oryza sativa* L.) variety ‘IR64’

キーワード Key word (marker-assisted selection) (gene pyramiding) (virulent BPH) (‘IR64’)

Introduction and purpose

The development of resistant rice (*Oryza sativa* L.) varieties is a key strategy for the eco-friendly control of brown planthopper (BPH: *Nilaparvata lugens* Stål). However, BPH outbreaks occur frequently owing to the evolution of virulent strains in the field and the rapid breakdown of monogenic resistance to BPH. Therefore, to enhance BPH resistance and gauge the effectiveness of gene pyramiding against strongly virulent BPH, we developed pyramided lines (PYLs) in the genetic background of ‘IR64’ carrying BPH resistance genes.

Material and methods

Five IR64-NILs—IR64-*BPH32*, IR64-*BPH17-ptb*, IR64-*BPH20*, IR64-*BPH17*, and IR64-*BPH3* were used for the development of the PYLs. PYLs for two resistance genes were developed from IR64-NILs descended from the BC₃F₂ generation. Plants carrying BPH resistance genes were selected by marker-assisted selection (MAS) using flanking SSR markers. The BPH population, Koshi-2013 and Koshi-2020 were used to evaluate the resistance level of the NILs developed using antibiosis test, honeydew test and MSST. We compared DTH, CL, PL, LL, LW, and PN as agronomic traits of PYLs with ‘IR64’.

Results

We developed six IR64-PYLs (*BPH3+BPH17*, *BPH32+BPH17*, *BPH32+BPH20*, *BPH3+BPH17-ptb*, *BPH20+BPH3*, and *BPH17-ptb+BPH32*) through marker-assisted selection. The resistance level of most IR64-PYLs was not significantly higher than that of the corresponding NILs in antibiosis test against both BPH populations Koshi-2013 and Koshi-2020 collected in Japan. Additionally, the area of honeydew excreted ranged from 2.6 to 6.2 mm² against Koshi-2013 and 6.0 to 9.6 mm² against Koshi-2020. Furthermore, the level of BPH resistance increased in all six IR64-PYLs compared to both 'IR64' and the corresponding NILs in MSST against both BPH populations. Among them, IR64-*BPH3+BPH17* and IR64-*BPH32+BPH17* exhibited the highest resistance to BPH.

Conclusion and consideration

We developed six PYLs in 'IR64' with a total of four resistance genes. Thus, these PYLs could serve as a valuable resource for breeding programs aimed at improving resistance to virulent strains of BPH and enhancing their durability.

Chapter title: Identification and characterization of QTL for BPH resistance from African rice, *Oryza glaberrima* Steud.

キーワード Key word (*Oryza glaberrima* Steud.) (introgression lines) (brown planthopper) (quantitative trait loci) (substitution mapping)

Introduction and purpose

The brown planthopper (BPH: *Nilaparvata lugens* Stål) is a highly destructive insect pest of rice in Asia. Significant yield losses resulted due to the rapid adaptation of BPH to resistant varieties. Therefore, it is crucial to search for new resistance genes for BPH from genetic resources for proper utilization of host-plant resistance. Among rice germplasm, *O. glaberrima* Steud. is the cultivated rice endemic to West Africa and possesses several unique characteristics including weed competitiveness, tolerance to salinity conditions, tolerance to drought, luxurious wide leaves and resistance to wide range of diseases and insect pest. In this study, we used *O. glaberrima* to identify the chromosomal location of BPH resistance genes and furthermore to characterize the resistance mechanism using BPH population.

Material and methods

To identify BPH resistance accessions, a total of 121 *O. glaberrima* accessions (OGA) from the rice germplasm collection of IRRI, Los Baños, Philippines was screened for their level of resistance to BPH using antibiosis and MSST. Furthermore, *O. glaberrima* introgression lines (GILs) from African rice variety, *O. glaberrima* (IRGC 104038) originated from Senegal were used to identify loci for BPH resistance. Using genotype data and phenotype data (Damage score by MSST and honeydew area) on GILs, QTL analysis was conducted to estimate the locations of BPH resistance genes on chromosome. BPH population, Hadano-1966 was used for the screening of *O. glaberrima* accessions, evaluation of GILs and characterization of mechanism for BPH resistance QTL using antibiosis, antixenosis and honeydew test.

Results

At initial screening, OGA accessions showed a medium level of resistance including IRGC 104038 against Hadano-1966 in both antibiosis test and MSST. Furthermore, we used GILs to identify QTLs using damage scores and honeydew area. The QTL for damage score, *qBPH6*, was located between markers RM19285 and RM19288 on chromosome 6, with a PVE of 34.7%. The other QTL for honeydew area, *qHOD6*, was located between the markers RM19274 and RM19285, with a PVE of 25.2%. The *qBPH6* was delimited as approximately 1.41 Mbp between markers RM3132 and RM19359 on chromosome 6. The lines carrying *qBPH6* showed the antibiosis mechanism for BPH resistance.

Conclusion and consideration

We identified and mapped the resistance gene, *qBPH6* from *O. glaberrima* introgression lines. Thus, *qBPH6* could be effectively introgressed through MAS into the elite rice variety for durable resistance releasing as multiline.