

Electronic Journal of Plant Breeding



Research Article

Molecular diversity assessment of rice genotypes for brown planthopper resistance using microsatellite markers

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Abstract

The brown planthopper (*Nilaparvata lugens* (Stål)) is a major insect pest that is primarily present in Asia causing a significant impact on rice crop. Seventy-three rice genotypes were evaluated for molecular diversity based on known BPH resistance loci. A total number of 108 alleles were detected by 39 polymorphic markers with an average of 2.37 alleles per locus. The allele frequency which is useful in estimating the frequency of alleles, ranged from 0.054 to 0.9726, while the expected heterozygosity varied between 0.054 (RM494) and 0.688 (RM231) with an average of 0.431. The Polymorphic Information Content (PIC) values provided an estimation of the marker's discriminating power by ranged from 0.066 (RM496) to 0.621 (RM231) suggesting moderate level of polymorphism for the selected BPH specific SSR markers among the germplasm lines. Accounting the major allele frequency, heterozygosity and PIC content, the marker RM231 associated with *QBph3.1* and regarded as the most informative one along with RM19291, RM335, RM469, RM518, RM8213, RM228, RH078 and RM589 for dissecting the molecular diversity of germplasm lines with respect to BPH resistance. Based on the marker data, the genotypes were classified into seven clusters that distinguished the lines clustering as resistant and susceptible separately. The information about the genetic diversity of these lines will be extremely useful for proper selection of parents related to BPH reaction, especially for gene mapping and for marker assisted selection.

Key words

Rice, Brown planthopper resistance, Molecular diversity, Microsatellite markers

INTRODUCTION

Being the Asia's most economical and culturally important food crop, Rice (*Oryza sativa* L.) is regarded as one of the most important crops in the world. More than three billion people around the world depend on rice for 20-80 per cent of their dietary intake. Approximately 52 per cent of the global production of rice is lost annually owing to the damage caused by biotic stress factors, of which 25 per cent is attributed to the attack of insect pests (Brookes and Barfoot, 2003). Among the biotic stresses, Brown

Planthopper (BPH), *Nilaparvata lugens* (Stål) is considered as the most destructive pest, causing a significant yield loss in most of the rice cultivating areas of Asia. The pest is a monophagous and non-traditional insect that has advanced from non-significance to prominence, posing a threat to rice production. BPH sucks the phloem sap with its piercing-sucking mouthparts and affects the growth and vigor of rice plants leading to hopperburn symptoms (Backus *et al.*, 2005). The pest also serves as a vector for

viral diseases including Grassy stunt and Ragged stunt in most of the rice cultivars of India as well as Asia (Jena and Kim, 2010; Sarao *et al.*, 2016). Chemical method of BPH control is not only costly, but also harmful to one's health, polluting the ecosystem, and disrupting the natural balance of BPH predators that hold the pest population (Jhansi Lakshmi *et al.*, 2010a and b; Mohanapriya *et al.*, 2019). This can also lead to the development of insecticide-resistance in the pest (Mohan *et al.*, 2019). Host plant resistance as a part of integrated pest management is the most cost-effective and reliable way to control BPH (Soundararajan and Jeyaprakash, 2019).

The genetic studies of BPH resistance in rice are increasing since the discovery of molecular markers. Till date, 38 genetic loci for BPH resistance have been identified in rice species using markers assigned to different rice chromosomes (Balachiranjeev *et al.*, 2019). Because of their high polymorphic nature and transferability, SSR markers have proven to be very effective tools in the study of BPH genes, genetic diversity, and relatedness of most crop species among the PCR-based markers (Ishii *et al.*, 1994). The present study was undertaken to assess the genetic variability among different rice germplasm

lines using microsatellite (SSR) markers linked to BPH resistance genes in order to unravel the molecular diversity among the lines.

MATERIALS AND METHODS

The materials used in the study comprised of seventy-three genotypes of rice (**Table 1**) which includes MAGIC *indica* lines (27), BPH gene differentials (30), popular rice varieties (3), wild species (5), germplasm lines (7) and landraces (1). The materials were collected from ICAR-Indian Institute of Rice Research (IIRR), Hyderabad, Institute of Biotechnology, Professor Jayashankar Telangana State Agricultural University (PJ TSAU), Hyderabad and Agricultural Research Institute (ARI), Rajendranagar, Hyderabad.

Genomic DNA was extracted from young and healthy leaves of 73 genotypes using the CTAB method given by Murray and Thompson (1980). A total of 53 microsatellite markers reportedly linked to BPH resistance (**Table 2**) on chromosomes 1, 2, 3, 4, 6, 10 and 12 were used to characterize and assess genetic diversity among the germplasm lines having varied response to brown planthopper stress. Polymerase chain reaction (PCR)

Table 1. List of 73 genotypes used in the study

| S.No. | Genotype | S.No. | Genotype | S.No. | Genotype |
|-------|------------------|-------|------------------------|-------|-----------|
| 1 | Mudgo | 26 | Sinasivappu | 51 | M1 |
| 2 | IR 64 | 27 | Balamwee | 52 | M131 |
| 3 | ASD 7 | 28 | IR 62 | 53 | M189 |
| 4 | Milyang 63 | 29 | RathuHeenati accession | 54 | M190 |
| 5 | RathuHeenati | 30 | IR 65482-136-2-2 | 55 | M227 |
| 6 | Babawee | 31 | M4 | 56 | M240 |
| 7 | ARC 10550 | 32 | M201 | 57 | RPV1160 |
| 8 | Swarnalatha | 33 | M229 | 58 | RPV1355 |
| 9 | T12 | 34 | M284 | 59 | RPV1189 |
| 10 | Chinsaba | 35 | M286 | 60 | IET23993 |
| 11 | Pokkali | 36 | M312 | 61 | CG171 |
| 12 | IR 65482-7-216 | 37 | M344 | 62 | PH190 |
| 13 | IR 71033-121-15 | 38 | M359 | 63 | CG180 |
| 14 | MUT NS1 | 39 | M61 | 64 | CG156 |
| 15 | OM 4498 | 40 | M80 | 65 | CG211 |
| 16 | RP 2068-18-3-5 | 41 | M88 | 66 | BM71 |
| 17 | MO1 | 42 | M123 | 67 | KNM118 |
| 18 | MTU 1010 | 43 | M179 | 68 | BPT5204 |
| 19 | RP BIO 4918-230S | 44 | M182 | 69 | PTB33 |
| 20 | IR 26 | 45 | M187 | 70 | TN1 |
| 21 | IR 40 | 46 | M276 | 71 | IM6 |
| 22 | IR 66 | 47 | M278 | 72 | RNR 15048 |
| 23 | IR 72 | 48 | M293 | 73 | 10-3 |
| 24 | Utrirajappan | 49 | M362 | | |
| 25 | Ndiang Marie | 50 | M289 | | |

Table 2. SSR markers specific for BPH resistance genes used for molecular diversity analysis

| S.No. | Gene | Marker | Chromosome | Forward | Reverse |
|-------|------------------------------------|---------|------------|---------------------------|---------------------------|
| 1 | <i>Bph33(t)</i> | RM488 | 1 | CAGCTAGGGTTTTGAGGCTG | TAGCAACAACCAGCGTATGC |
| 2 | <i>Bph33(t)</i> | RM212 | 1 | CCACTTTCAGCTACTACCAG | CACCCATTTGTCTCTCATTATG |
| 3 | <i>Bph33(t)</i> | RM11522 | 1 | TAAGTGCAGTGCTCAACAAAGG | CTAGGTACCCGATTAAGATTCACC |
| 4 | <i>Bph13(t)</i> | RM250 | 2 | GGTTCAAACCAAGCTGATCA | GATGAAGGCCTTCCACGCAG |
| 5 | <i>Bph13(t)</i> | RM240 | 2 | CCTTAATGGGTAGTGTGCAC | TGTAACCATTCTTCCATCC |
| 6 | <i>Qbph3</i> | RM313 | 3 | TGCTACAAGTGTCTTCAGGAC | GCTCACCTTTTGTGTTCAC |
| 7 | <i>Qbph3</i> | RM7 | 3 | TTGCCATGAAGTCTCTCG | CCTCCCATCATTTTCGTTGT |
| 8 | <i>QBph3</i> | RM2453 | 3 | TAGGTGTTTCAGGAGTAAAGA | AAACCAGTATTGCTTACAAG |
| 9 | <i>Qbph3.1</i> | RM231 | 3 | CCAGATTATTTCTGAGGTC | CACTTGCATAGTTCTGCATTG |
| 10 | <i>Bph3</i> | RM307 | 4 | GTAATACCGACCTACCGTTCAC | CTGCTATGCATGAAGTCTC |
| 11 | <i>Bph3</i> | RH784 | 4 | TTCTTGGCTGATTGGGAGTAT | ATTTGGAATGGAGCAGAGTGA |
| 12 | <i>Bph3</i> | RH078 | 4 | GCCGATTTGGTCTAACTTCATT | CAACCAGCAAGGAAACAATAAA |
| 13 | <i>Bph3</i> | RHD9 | 4 | GTCCATCCGAAGGTGAAAGT | CCGAACATCGAGGAATACAA |
| 14 | <i>Bph3</i> | WH2 | 4 | CCCACCACACCAGAGATAAA | ACACAACACCCGCATACAA |
| 15 | <i>Bph3</i> | RHC10 | 4 | CAATACGGGAGATTGGAGT | TTGGGAAGCATACGAGTGA |
| 16 | <i>Bph3</i> | W1 | 4 | TCCTAATCAGCCAATAATCA | GCAATCTAGTGCACGAACATA |
| 17 | <i>Bph3, Bph17(t)</i> | RM5953 | 4 | AACTTTCTGTGATGGTATC | ATCCTTGTCTAGAATTGACA |
| 18 | <i>Bph17(t)</i> | RM518 | 4 | CTCTTACTCACTACCATGG | ATCCATCTGGAGCAAGCAAC |
| 19 | <i>Bph17(t)</i> | RM206 | 4 | CCCATGCGTTTAACTATTCT | CGTTCATCGATCCGTATGG |
| 20 | <i>Bph17(t)</i> | RM8213 | 4 | AGCCCAGTGATACAAAGATG | GCGAGGAGATACCAAGAAAG |
| 21 | <i>Bph17(t)</i> | MS10 | 4 | CAATACGAGAAGCCCCTCAC | CTGAAGGAACACGCGGTAGT |
| 22 | <i>Qbph4, Bph17(t)</i> | RM335 | 4 | GTACACACCCACATCGAGAAGC | TCCATGGATATACGAGGAGATGC |
| 23 | <i>Qbph4</i> | RM551 | 4 | AGCCCAGACTAGCATGATTG | GAAGGCGAGAAGGATCACAG |
| 24 | <i>Bph3</i> | RM584 | 6 | TATGTGGTTGGCTTGCCTAGTGG | TGCCCATATGGTCTGGATGTGC |
| 25 | <i>Bph3</i> | RM225 | 6 | TGCCCATATGGTCTGGATG | GAAAGTGGATCAGGAAGGC |
| 26 | <i>Bph3</i> | RM508 | 6 | GGATAGATCATGTGTGGGGG | ACCCGTGAACCACAAAGAAC |
| 27 | <i>Bph3, Bph32</i> | RM588 | 6 | TCTTGCTGTGCTGTAGTGTACG | GCAGGACATAAATACTAGGCATGG |
| 28 | <i>Bph3, Qbph6</i> | RM8101 | 6 | GTGTAGTTACGACCAATGATACGC | TATAATGAGTTTCGAGCCGATCC |
| 29 | <i>Bph3, Bph32</i> | RM589 | 6 | GTGGCTTAACCACATGAGAACTACC | TCACATCATTAGGTGGCAATCG |
| 30 | <i>Bph3, Bph17(t)</i> | RM190 | 6 | GCTACAAATAGCCACCCACACC | CAACACAAGCAGAGAAGTGAAGC |
| 31 | <i>Bph3, bph4, Qbph4, Bph17(t)</i> | RM401 | 6 | TGGAACAGATAGGGTGTAAAGG | CCGTTCAACAACACTATACAAGC |
| 32 | <i>Bph3, bph4, Bph32</i> | RM19291 | 6 | CACTTGCACGTGCTCTGTACG | GTGTTTCAGTTCACCTTGCATCG |
| 33 | <i>Bph3, Qbph6</i> | RM469 | 6 | AGCTGAACAAGCCCTGAAAG | GACTTGGGCAGTGTGACATG |
| 34 | <i>bph4</i> | RM217 | 6 | ATCGCAGCAATGCCTCGT | GGGTGTGAACAAGACAC |
| 35 | <i>Bph32</i> | RM8072 | 6 | GATCACTCAGGTATCCATTC | AATCAGAGAGGCTAAAGACAATAAT |
| 36 | <i>Bph32</i> | RM6775 | 6 | GCAGATCAAGTATGCCCTGCC | TCGCTAGATAGGGGATGTGG |
| 37 | <i>Bph20 (t), Bph29</i> | RM435 | 6 | CTGGTTAATTACGTGCATGTCTGG | GGCATGTCATGTCTTGGTCTCC |
| 38 | <i>Qbph6</i> | RM510 | 6 | AACCCGATTAGTTTCTCGCC | TGAGGACGACGAGCAGATTC |
| 39 | <i>Qbph6</i> | RM6818 | 6 | GTCGCATTTCGTCTCCACC | ACCATTTCCAGATGACTCGG |
| 40 | <i>Qbph6</i> | RM8215 | 6 | GTTCTCCCTTCATGACACAG | TAGAGACTTTATATTTGGTGTGC |
| 41 | <i>Qbph6</i> | RM587 | 6 | ACGCGAACAAATTAACAGCC | CTTTGCTACCAGTAGATCCAGC |
| 42 | <i>Qbph6</i> | RM314 | 6 | CTAGCAGGAACCTTTTCAGG | AACATTCCACACACACACGC |
| 43 | <i>Qbph10</i> | RM484 | 10 | TCTCCCTCCTCACCATTGTC | TGCTGCCCTCTCTCTCTCTC |
| 44 | <i>Qbph10</i> | RM496 | 10 | GACATGCGAACAACGACATC | GCTGCGGCGCTGTATAC |
| 45 | <i>Qbph10</i> | RM216 | 10 | GCATGGCCGATGGTAAAG | TGTATAAAACCACACGGCCA |
| 46 | <i>Qbph10</i> | RM333 | 10 | GTACGACTACGAGTGCACCAA | GTCTTCGCGATCACTCGC |
| 47 | <i>Qbph10</i> | RM228 | 10 | CTGGCCATTAGTCTTGG | GCTTGCGGCTCTGCTTAC |
| 48 | <i>Qbph10</i> | RM406 | 10 | GAGGGAGAAAGGTGGACATG | TGTGCTCCTTGGGAAGAAAG |
| 49 | <i>Qbph10</i> | RM494 | 10 | GGGAGGGGATCGAGATAGAC | TTTAACCTTCTTCCGCTCC |
| 50 | <i>bph7</i> | RM3448 | 12 | CTTCTCCTTCTCCTCCTC | CACGTGACACGTACACCCTC |
| 51 | <i>bph18</i> | RM273 | 12 | GAAGCCGTCGTGAAGTTACC | GTTTCTACCTGATCGCGAC |
| 52 | <i>bph18</i> | RM6506 | 12 | GTCCTCAGGTGATTCGCC | GTCGCTCGAAGCCAATTAAG |
| 53 | <i>bph18</i> | RM3331 | 12 | CCTCTCCATGAGCTAATGC | AGGAGGAGCGGATTTCTCTC |

was performed in 10 µl volume using Eppendorf gradient thermocycler. The reaction mixture contained 2 µl template DNA, 0.5 µl each forward and reverse primers, 4 µl of TAKARA master mix and 3 µl of double distilled water. Thermal cycler was programmed to 1 cycle of 5 min at 94°C, 35 cycles of denaturation at 95°C for 1 min, 45s for annealing temperature (55 to 60°C depending on the marker used) and 40s at 72°C for primer elongation. Finally, 1 cycle of 7 min at 72°C was used for final extension. The reproducibility of the amplification products was checked twice for each primer. SSR analysis was carried out by 3% Agarose gel electrophoresis and the DNA fragments were visualized under UV-trans illuminator and documented using a documentation system (GELSTAN) which was stored for further scoring and permanent records.

The sizes of the amplified fragments were estimated with the help of Alpha image software by Gel documentation system using 100 bp DNA ladder (XcelGEN) as size standard. Only clear and unambiguous SSR markers were scored for the presence or absence of the corresponding band among the germplasm lines. The score '1' and '0' indicates the presence and absence of the bands, respectively. The binary data matrix of SSR markers from the 73 germplasm lines was subjected to cluster analysis and a Neighbor joining tree with bootstrap values was constructed using the un-weighted pair group method with arithmetic mean (UPGMA) using DARwin 6.0.21 software (Perrier *et al.*, 2003). To estimate the discriminatory power of a marker, the Polymorphic Information Content (PIC) for each SSR marker along with allelic frequency and observed heterozygosity were calculated using CERVUS 3.0.7 software (Kalinowski *et al.*, 2007).

RESULTS AND DISCUSSION

Assessing the genetic diversity of genotypes is essential for efficiently preserving, characterizing and exploiting biodiversity. Fifty-three microsatellite markers that were

linked with BPH resistance having co-dominant and high repeatability were selected to evaluate the diversity of the rice germplasm resources (Table 2). Out of the 53 markers used, 39 SSR markers showed polymorphism by revealing 108 alleles (Table 3). The number of alleles per locus varied from a minimum of 2 (RM190, RM435, RM250, RM508, RM518, RM216, RM206, RM314, RM3331, RM494, RM212, RM488, RM7, RM510, RM8215, RM508 and RM11522) to 5 (RM8213), with an average of 2.76 alleles per locus. Significant differences in allelic diversity among BPH specific microsatellite loci with a mean of 2.26 alleles was reported by Supari *et al.*(2019), while characterizing Malaysian rice cultivars using SSR markers and 2.37 alleles by VijayaLakshmi *et al.*(2010), while assessing rice genotypes for brown planthopper resistance.

Gene diversity or expected heterozygosity (H), is a common statistic for assessing genetic variation within the population. In the present study, the expected heterozygosity (Table 3 and Fig. 1) differed among the markers ranging from 0.054 (RM494) to 0.688 (RM231) with an average of 0.431. The lesser mean value of the heterozygosity indicated that majority of the germplasm lines showed a single allele in their respective microsatellite profiles. Allele frequency, also known as gene frequency, is the relative frequency of an allele at a particular locus in a population which is expressed as fraction or per cent (Gillespie *et al.*, 2004). The mean major allele frequency was 0.64 with the marker RM494 exhibiting the highest major allele frequency of 0.9726 followed by RM496 (0.965). The lowest values for major allele frequency were observed for the markers RM307 (0.054) and RM231 (0.373).

To determine the information of each marker and its discriminatory potential, the Polymorphic Information Content (PIC) was estimated for each locus. The

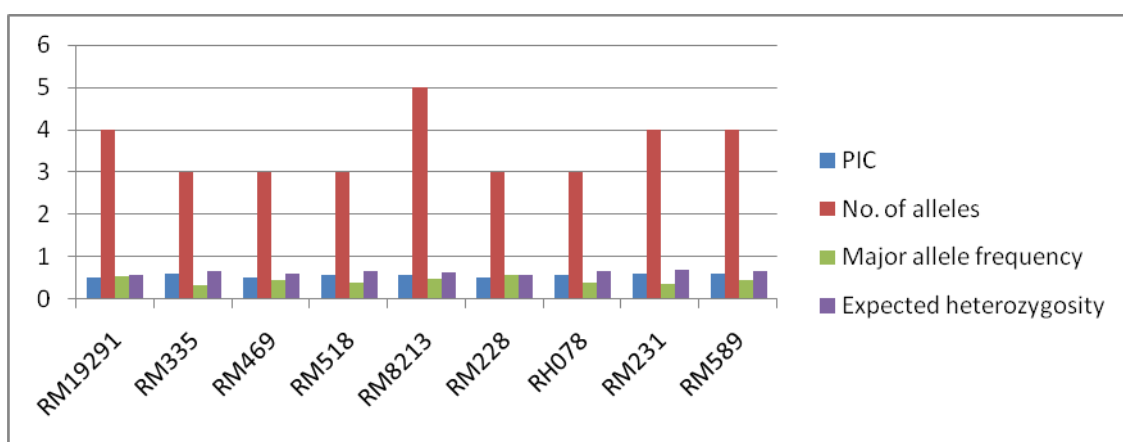


Fig. 1. Histogram representing the PIC, Number of alleles, allele frequency and expected heterozygosity of the most informative markers identified in the study

Table 3. Total number of alleles, Major allele frequency, heterozygosity and polymorphism information content (PIC) value of polymorphic SSR markers linked with BPH resistance assayed in 73 rice genotypes

| S.No | Marker | Number of alleles | Expected heterozygosity | PIC | Allele frequency |
|------|---------|-------------------|-------------------------|-------|------------------|
| 1 | RM190 | 2 | 0.495 | 0.37 | 0.569 |
| 2 | RM435 | 2 | 0.197 | 0.176 | 0.8904 |
| 3 | RM250 | 2 | 0.253 | 0.219 | 0.8529 |
| 4 | RM508 | 2 | 0.424 | 0.332 | 0.6985 |
| 5 | RM584 | 2 | 0.479 | 0.362 | 0.6119 |
| 6 | RM216 | 2 | 0.479 | 0.362 | 0.6119 |
| 7 | RM206 | 2 | 0.498 | 0.372 | 0.553 |
| 8 | RM314 | 2 | 0.277 | 0.237 | 0.8356 |
| 9 | RM3331 | 2 | 0.492 | 0.369 | 0.5764 |
| 10 | RM494 | 2 | 0.054 | 0.052 | 0.9726 |
| 11 | RM212 | 2 | 0.503 | 0.375 | 0.5205 |
| 12 | RM488 | 2 | 0.487 | 0.367 | 0.589 |
| 13 | RM7 | 2 | 0.465 | 0.355 | 0.6389 |
| 14 | RM510 | 2 | 0.482 | 0.364 | 0.6027 |
| 15 | RM8215 | 2 | 0.258 | 0.223 | 0.8493 |
| 16 | RM508 | 2 | 0.388 | 0.311 | 0.7397 |
| 17 | RM11522 | 2 | 0.283 | 0.242 | 0.8308 |
| 18 | RM588 | 3 | 0.247 | 0.231 | 0.863 |
| 19 | RM335 | 3 | 0.672 | 0.593 | 0.3361 |
| 20 | RM225 | 3 | 0.367 | 0.333 | 0.7823 |
| 21 | RM469 | 3 | 0.611 | 0.524 | 0.4571 |
| 22 | RM518 | 3 | 0.665 | 0.586 | 0.4 |
| 23 | RM496 | 3 | 0.067 | 0.066 | 0.9658 |
| 24 | RM3448 | 3 | 0.486 | 0.43 | 0.6781 |
| 25 | RM484 | 3 | 0.386 | 0.328 | 0.7534 |
| 26 | RM6775 | 3 | 0.106 | 0.102 | 0.9452 |
| 27 | RM217 | 3 | 0.531 | 0.47 | 0.6357 |
| 28 | RM587 | 3 | 0.508 | 0.39 | 0.5616 |
| 29 | RM228 | 3 | 0.585 | 0.512 | 0.5616 |
| 30 | RM240 | 3 | 0.217 | 0.202 | 0.8803 |
| 31 | MS10 | 3 | 0.542 | 0.453 | 0.5822 |
| 32 | RH078 | 3 | 0.666 | 0.587 | 0.3904 |
| 33 | RM401 | 4 | 0.341 | 0.316 | 0.8 |
| 34 | RM19291 | 4 | 0.585 | 0.509 | 0.558 |
| 35 | RM589 | 4 | 0.66 | 0.591 | 0.4615 |
| 36 | RM231 | 4 | 0.688 | 0.621 | 0.3732 |
| 37 | RM307 | 4 | 0.502 | 0.46 | 0.0542 |
| 38 | RM333 | 4 | 0.239 | 0.226 | 0.8696 |
| 39 | RM8213 | 5 | 0.636 | 0.566 | 0.4922 |

PIC value of a marker for detecting polymorphism is determined by the number of observable alleles and the frequency distribution of those alleles, thereby providing an estimation of the marker's discriminating power (Nagy *et al.*, 2012). Usually, markers with PIC value of more than 0.5 are considered to be informative markers for

genetic studies for measuring the polymorphism for a marker locus (DeWoody *et al.*, 1995). In the present study, the polymorphism information content (PIC) values ranged from 0.066 (RM496) to 0.621 (RM231) suggesting moderate level of polymorphism for the selected SSR markers specific to BPH among the rice genotypes

Table 4. Clustering of germplasm lines based on molecular diversity

| Cluster | Number of genotypes | Germplasm lines |
|---------|---------------------|---|
| I A | 7 | Mudgo, IR64, IR72, ASD7, Milayang63, Rathuheenati, Utrirajappan |
| I B | 9 | Sinnasivappu, IR62, Balamwee, Ndiang Marie, OM4498, IR65482-136-2-2, RP 2068-18-3-5, M4, M201 |
| II | 7 | M286, M227, M240, TN1, KNM118, BPT5204, RNR15048 |
| III A | 2 | PH190, CG171 |
| III B | 4 | M187, M182, M278, M276 |
| IVA | 4 | BM71, M179, 10-3, IM6 |
| IV B | 4 | M359, M80, M61, M88 |
| IV C | 3 | M289, M362, M293 |
| V A | 3 | M1, M131, RPV 1355 |
| V B | 3 | M284, M189, M190 |
| V C | 4 | RPV1189, CG211, CG156, CG180 |
| VI A | 9 | RP BIO 4918-230S, MTU1010, M229, M344, M312, RathuHeenati accession, IR40, IET23993, PTB33 |
| VI B | 4 | Pokkali, Chinsaba, IR71033, IR-65485-7-216 |
| VI C | 8 | T12, Babawee, IR26, Swarnalatha, ARC10550, MO1, RPV1160, MUTNS1 |
| VII | 2 | M123, IR 66 |

the highly resistant checks of Rathuheenati and PTB33 clustered in I and VI, respectively. Similarly, moderately resistant lines were grouped in cluster V and susceptible lines in Cluster III B and VA. Wild species PH190, CG171 alongwith CG211, CG156, and CG180 have separately formed clusters III(A) and V(C) respectively indicating the power of the chosen markers in distinguishing the wild species from the rest. On the whole, the results indicated the presence of considerable diversity in the germplasm lines studied. From the present study, the most diverse genotypes namely, PTB33, CG180, CG171 and IR64 can be selected and utilized for BPH resistance breeding programmes.

Having gene information for BPH target loci that are derived from markers can help breeders to use the germplasm more efficiently. The current study provided an insight of the genetic diversity of 73 rice genotypes having different reactions to the pest. The selected SSR markers were able to clearly distinguish the germplasm lines into clusters which was in agreement with the phenotypic reaction to the pest insect. Considering the major allele frequency, heterozygosity and PIC content, the marker RM231 linked to *QBph3.1* has been regarded as the most informative for dissecting the molecular diversity of germplasm lines with respect to BPH resistance. The study also identified the markers, RM19291 (*Bph3*, *bph4*, *Bph32*), RM335 (*Bph17(t)*), RM469 (*Bph3*, *Qbph6*), RM518 (*Bph17(t)*), RM8213 (*Bph17(t)*), RM228 (*Qbph10*), RH078 (*Bph3*), and RM589 (*Bph3*) to be useful in molecular diversity studies.

ACKNOWLEDGEMENT

The authors greatly acknowledge the support provided by ICAR-Indian Institute of Rice Research, Hyderabad, PJTSAU and ARI, Rajendranagar, Hyderabad for providing the resources and facilities for conducting the experiment.

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