Research Paper

Breeding and characterization of the world’s first practical rice variety with resistance to brown spot (Bipolaris oryzae) bred using marker-assisted selection

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Brown spot (BS) caused by Bipolaris oryzae is a serious disease of rice and decreases grain yield. Breeding for BS resistance is an economical solution but has not hitherto been achieved. To develop a practical BS-resistant variety, we introduced a chromosomal segment including a quantitative trait locus (QTL) for BS resistance, qBSfR11, derived from the BS-resistant local resource ‘Tadukan’, into the genetic background of the high-yielding but susceptible ‘Mienoyume’. Resistance is controlled by a single recessive gene in a 1.3-Mbp region. We named this gene bsr1 (brown spot resistance 1). The near-isogenic line bsr1-NIL had a greater yield with larger grain width than Mienoyume but similar other agronomic traits in fields where BS was mild; it had a significantly lower BS disease score and a 28.8% higher yield in fields where BS was more severe, and it showed resistance to multiple isolates of BS fungus. It showed stable resistance to BS and had excellent agricultural traits in the presence of BS. We developed the bsr1-NIL with resistance to BS and applied it for variety registration to Ministry of Agriculture, Forestry and Fisheries in Japan as ‘Mienoyume BSL’. This is the first report for the BS resistant rice variety bred using marker-assisted selection.

Key Words: Oryza sativa L., disease resistance, brown spot, qBSfR11, bsr1, near-isogenic line, breeding variety.

Introduction

Brown spot (BS) is a fungal disease that is caused by Bipolaris oryzae and infects various parts of rice plants. The incidence of grain yield losses by BS and the use of countermeasures to BS (e.g., silicon fertilization) in the USA and India have been reported (Barnwal et al. 2013, Datnoff et al. 1991). The rate of yield reduction is up to 20% (Chakrabarti 2001, Kamal and Mia 2009, Ou 1985). It is highly possible that BS will become a more serious disease under global warming because its optimal temperature range for growth of the pathogen is relatively high (Mizobuchi et al. 2016).

In Japan in 2019, BS infected 159,482 ha, the third-largest area after sheath blight (581,367 ha) and rice blast (451,197 ha) (JPPA 2021). During 1975–2019, the area peaked in 1984 (384,836 ha) and has since decreased, but it is gradually increasing again (JPPA 2020). In Niigata prefecture, a decrease in the application of fungicides because of expansion of the use of rice-blast-resistant lines (Ishizaki et al. 2005) is presumed to be a reason for the spread of BS (Yamaguchi et al. 2007). As many rice-blast-resistant varieties have now been developed, we need to pay more attention to BS.

Some local genetic resources such as ‘Tadukan’ (Ohata and Kubo 1974, Yoshii and Matsumoto 1951), ‘CH45’ (Misra 1985), ‘Dawn’ (Eruotor 1986), and ‘Tetep’ (Eruotor 1986, Ohata and Kubo 1974, Yoshii and Matsumoto 1951) are resistant to BS, and some quantitative trait loci (QTLs) for resistance have been detected (Mizobuchi et al. 2016). We detected a major BS resistance QTL, qBSfR11, on chromosome (Chr.) 11 by field resistance tests with recombinant inbred lines derived from crosses between the resistant ‘Tadukan’ and the susceptible ‘Hinohikari’ (Sato et al. 2015). The Tadukan allele at the QTL also conferred BS resistance in the ‘Koshihikari’ background (Sato et al. 2015). Two other QTLs were detected near qBSfR11: BSq11.2v, derived from IR62266 (Katara et al. 2010), and...
Breeding of NILs with BS resistance. We bred ‘Mienoyume BSL’, which is with the shortest Tadukan segment among the BS-resistant we evaluated the BS resistance of 52 NILs (19 BC2017a). Because Mienoyume has high yield and good grain appearance, we developed near-isogenic lines (NILs) of it with BS resistance. We bred ‘Mienoyume BSL’, which is the world’s first practical variety with resistance to BS bred using marker-assisted selection (MAS). We discuss the stability of resistance to multiple BS strains.

Materials and Methods

Breeding of NILs

Fig. 1 shows the breeding scheme used for the development of the NILs. The donor parent (R307-48-9) was a NIL developed by Sato et al. (2015), in which the major resistance QTL (qBSfR11) on Chr. 11, derived from indica ‘Tadukan’ (resistant), had been introduced into the genetic background of ‘Koshihikari’ (susceptible). qBSfR11 was transferred into the Mienoyume background by sequential backcrossing method. During backcrossing or selfing from 2014 to 2015, promising individuals or lines were selected by MAS with simple sequence repeat (SSR) markers (McCouch et al. 2002) based on the target region on Chr. 11. In 2016, six SSR markers at the qBSfR11 locus were used to verify the size of the substituted segments. In 2017, we evaluated the BS resistance of 52 NILs (19 BC2F3, 33 BC2F4), divided into 12 groups based on their generations and genotypes, by field evaluation testing described later (Fig. 2A). The whole genome of six resistant NILs (one in group-9, two in group-10, three in group-11) was surveyed by using 243 single-nucleotide polymorphism (SNP) markers distributed evenly across the 12 chromosomes (Nagasaki et al. 2010, Supplemental Table 1). A BS-resistant NIL, named bsr1-NIL, was selected as a promising line. In 2018, three more SSR markers (RM27159, RM27163, and RM27244), located downstream of the six SSR markers used in 2016, were used to determine the genotype of bsr1-NIL in the qBSfR11 region and to delimit the chromosomal location of the BS-resistance gene in a group-11 NIL with the shortest Tadukan segment among the BS-resistant lines.

Field trials

Field trials were conducted in the paddy fields at Mie Prefecture Agricultural Research Institute (Mie, Japan) in Matsusaka (34°63’N, 136°48’E) and Iga (34°70’N, 136°13’E).

BS resistance was evaluated in Iga on a scale of 0 (no incidence) to 9 (severe) according to the procedure of Matsumoto et al. (2016) by using B. oryzae strain Iga-2 (Acc. No. 245177, MAFF Genebank) with two or three replications. In 2016, the inheritance mode of BS resistance was evaluated by using 153 individuals in the BC2F3 generation derived from one BC2F1 individual that had been confirmed to be heterozygous at the qBSfR11 locus by using SSR marker RM27073 (McCouch et al. 2002). We investigated their RM27073 genotype and BS resistance by field evaluation testing.

Other tests of agronomic traits of bsr1-NIL were conducted in Iga and Matsusaka, with two replications in 2018 and three replications in 2019 and 2020. bsr1-NIL and Mienoyume were transplanted on 14 or 15 May in Matsusaka and on 10 or 11 May in Iga at four seedlings per hill in 120 hills and six rows per replication, with a spacing of 30 cm × 15 cm in 2018 and 2019 and 30 cm × 18 cm in 2020. Nitrogen fertilizer was applied at 4.8 g N m−2 at transplanting and 4.0 g m−2 at heading in Matsusaka, and at 5.6 and 3.4 g m−2, respectively, in Iga. Major agronomic traits (days to heading, culm length, panicle length, brown rice yield, panicle number, 1000-grain weight, brown rice protein content, grain appearance, grain shape) were measured each year. Days to heading was calculated as days from transplanting to heading. From the results of these trials, bsr1-NIL was confirmed as a candidate for a practical BS-resistant variety for its high yield.

In 2020, to evaluate the effect of qBSfR11 on agronomic traits, we grew bsr1-NIL and Mienoyume in a part of the test field where BS was more severe after heading. Seedlings were transplanted on 28 May at four seedlings per hill in 80 hills and four rows per replication (three replications), with a spacing of 30 cm × 15 cm. Slow-release N fertilizer was applied at 7.5 g N m−2 at transplanting. Spreader plants (Mienoyume inoculated with Iga-2 strain) were planted around the plots but not within them. Plant height, stem number, and leaf greenness (SPAD value) were measured as indicators of crop growth at the panicle formation stage, when BS had not yet occurred. SPAD values were measured with a SPAD-502Plus chlorophyll meter (Konika Minolta, Inc., Tokyo, Japan). Yield, yield components (panicle number, spikelet number per panicle, percentage of filled spikelets, and 1000-grain weight), brown rice protein content, and grain appearance were measured at maturity.

Inoculation test using multiple BS strains

In 2020, bsr1-NIL, Mienoyume (susceptible), and Tadukan (resistant) were grown in 5.5 cm × 15.0 cm × 9.5 cm containers filled with sterilized soil (Clean No. 2, Ibiko Corporation, Gifu, Japan) inside a greenhouse of Mie Prefecture Agricultural Research Institute. Five seeds of each were sown in a row, at four rows per container. The isolates of BS fungus used were B. oryzae T. AOKI AR0126 (isolated...
in Okinawa prefecture; MAFF Genebank Acc. No. 235499) and F-1 (isolated in Ehime prefecture; Acc. No. 305067). Inoculation of fungus and evaluation of disease symptoms followed the methods of Sato et al. (2008).

DNA isolation and marker analyses

Total DNA was extracted from the leaves by using the CTAB method (Murray and Thompson 1980). PCR and electrophoresis for SSR analyses followed the method of Sato et al. (2015), but with Taq enzyme from GoTaq Green Master Mix (Promega, Madison, WI, USA), 55°C annealing.

Fig. 1. Breeding scheme for development of Mienoyume NILs.
temperature, and 3.0% gel concentration. All experimental procedures for the SNP analysis followed the method of Sato et al. (2015).

**Results**

**Graphical representation of NIL genotypes**

Fig. 2A shows the graphical genotypes at the *qBSfR11* region (between RM1219 and RM2191-1) and the phenotypes (BS disease scores and heading dates) of 52 NILs (BC$_5$F$_3$ 19 lines, BC$_4$F$_4$ 33 lines) that had been confirmed to be homozygous for either the Tadukan allele or the Mienoyume allele between RM1219 and RM2191-1 by SSR analysis in 2016. The genotype of the donor parent (R307-48-9) was the same as that of Tadukan (*qBSfR11* donor), and its BS disease score was significantly lower than that of Mienoyume. In both the BC$_5$F$_3$ and BC$_4$F$_4$ generations, the groups with Mienoyume segments from RM27073 to RM2191-1 (groups-1, 2, 3, 5, 6, 7, and 8) had the same disease scores as Mienoyume. In contrast, the groups with Tadukan segments there (groups-4, 9, 10, 11, and 12) had lower disease scores than Mienoyume.

The heading dates of all 52 NILs were the same as that of Mienoyume. SSR analysis downstream of RM2191-1 in Fig. 2A: Graphical genotypes in the *qBSfR11* region on Chr. 11 by SSR analyses in (A) 2016 and (B) 2018. □ Homozygous for Mienoyume; ■ homozygous for Tadukan. Numbers in parentheses beside SSR markers indicate their physical map positions (kbp) on Chr. 11 in IRGSP v. 1.0. A: 52 NILs in BC$_5$F$_3$ or BC$_4$F$_4$ generation with their BS disease scores (means ± SD) and heading dates in 2017. **Significant difference from Mienoyume at 1% in (1) BC$_5$F$_3$ and (2) BC$_4$F$_4$ generations (except in groups with one line) by Dunnett’s test. There was no significant difference in heading dates between NILs (both generations) and Mienoyume at 5% by Dunnett’s test. B: NIL of group-11.
2018 showed that a NIL of group-11 had a 1.3-Mbp Tadukan segment from RM27073 to RM27159 (Fig. 2B).

Inheritance mode of BS resistance

Fig. 3 shows the frequency distribution of BS disease scores in 153 BC$_2$F$_2$ individuals, based on the genotypes of SSR marker RM27073 at the qBSfR11 locus. Disease scores of 0 to 4 were considered to indicate resistance and those of 4.5 to 9 to indicate susceptibility. The BC$_2$F$_2$ individuals segregated in a 1:3 ratio of resistant: susceptible (Table 1), confirming that the resistance to BS is controlled by a single recessive gene at the qBSfR11 locus. We named this gene bsr1 (brown spot resistance 1).

Genetic backgrounds and agronomic traits of bsr1-NIL

We selected a resistant NIL (bsr1-NIL) in the BC$_2$F$_4$ generation in 2017. bsr1-NIL had the group-9 genotype (Fig. 2A). It had R307-48-9 segments on Chr. 11 (3.5 Mbp from aa 11004652 to aa 11007953; Fig. 4C). On all other chromosomes except for Chr. 11, it was homozygous for Mienoyume segments. The other five lines tested for the Tadukan segment from RM27073 to RM27159 (Fig. 2B). Inheritance mode of BS resistance.

In both yield-trial paddy fields where BS was less severe than in the BS resistance test field, there was no significant difference in BS disease score between bsr1-NIL and Mienoyume (Table 2). However, some traits were significantly different. In Matsusaka, brown rice yield and grain width of bsr1-NIL were 34 g m$^{-2}$ higher and 0.06 mm larger, respectively, than those of Mienoyume. In Iga, grain width of bsr1-NIL was 0.06 mm larger than that of Mienoyume. In a part of the BS resistance test field where BS was more severe, the BS disease score of bsr1-NIL was 3.0 lower than that of Mienoyume (Table 3, Fig. 4A, 4B). There were no significant differences in growth characteristics at the panicle formation stage between bsr1-NIL and Mienoyume. On the other hand, brown rice yield and percentage of filled spikelets of bsr1-NIL were respectively 106 g m$^{-2}$ and 12.3% higher than those of Mienoyume. The protein content of brown rice of bsr1-NIL was 1.3% lower than that of Mienoyume. In addition, comparing agronomic traits of bsr1-NIL and Mienoyume in BS resistance test field (severe conditions) and yield-trial field (mild conditions), in ‘severe conditions’, both of them had lower brown rice yield and percentage of filled spikelets and higher protein content of brown rice than in ‘mild conditions’, and those degree of decrease or increase was smaller in bsr1-NIL than in Mienoyume. 1000-grain weight and grain width were also lower in ‘severe conditions’, but those degree of decrease was same in bsr1-NIL and Mienoyume. This showed that bsr1-NIL had larger 1000-grain weight with larger grain width than Mienoyume regardless of the BS severity. The same result was also found in Table 2. Panicle number and spikelet number per panicle were different between two fields, but this result is presumed not to be due to BS because they are the yield components determined before the heading stage when BS began to be severe in this study. These results suggest that BS reduced the ripening of rice and decreased the brown rice yield, and qBSfR11 introduced into bsr1-NIL had the effect of reducing the decrease in brown rice yield by suppressing the decrease in percentage of filled spikelets by BS.

Resistance of bsr1-NIL to other isolates of BS fungus

At the seedling stage, the disease score of bsr1-NIL was significantly lower than that of Mienoyume following artificial inoculation of B. oryzae T. AOKI AR0126 and F-1 (Table 4). Thus, bsr1-NIL showed resistance to multiple isolates of BS fungus.

Table 1. The segregation of BC$_2$F$_2$ individuals as resistant or susceptible to BS

| Generation | Number of individuals | Resistant type | Susceptible type | $\chi^2$-value (1:3) | p-value |
|------------|------------------------|----------------|-----------------|---------------------|---------|
| BC$_2$F$_2$ | 36                     | 117            |                 | 0.18                | 0.67    |

Discussion

Characteristics of BS-resistance QTL, qBSfR11

Mienoyume is highly susceptible to BS and is more susceptible than Koshihikari (Matsumoto et al. 2017a). Here, we developed NILs with resistance to BS by using qBSfR11, derived from Tadukan, which had been identified as a major QTL responsible for resistance to BS (Sato et al. 2015). qBSfR11 had been previously confirmed to confer BS resistance in the Koshihikari genetic background (Sato...
et al. 2015). Here, it conferred resistance in the Mienoyume genetic background also (Table 3, Fig. 4). This result strongly indicates that qBSfR11 is effective at conferring resistance to BS.

The genotype of a resistant NIL in group-11 showed that qBSfR11 was located around the 1.3-Mbp interval RM27073 to RM27159 (Fig. 2B). The candidate genomic region was narrowed from the donor parent R307-48-9. Annotation of the ‘Nipponbare’ sequence in RAP-DB shows 107 genes predicted within this interval (Sakai et al. 2013). It is hard to predict which of the genes might be related to BS resistance because there have been no reports of genes associated with BS resistance and because the morphological and physiological functions of qBSfR11 are not yet known. Thus, further delimitation of the candidate genomic region of qBSfR11 will be necessary to identify the gene corresponding to qBSfR11.

The distribution of BS disease scores in 153 BC1F2 individuals suggested that resistance to BS is controlled by a single recessive gene (Fig. 3). We named this gene bsr1 (brown spot resistance 1). Mwendo et al. (2017) reported that resistance to BS was controlled by one or two dominant genes, whereas Adair (1941) reported the involvement of several recessive genes. These present and previous studies show that there are different genes for BS resistance, with different modes of inheritance. Goel et al. (2006) suggested that pyramiding QTLs for BS resistance would be effective because the resistance in four lines of wild rice Oryza nivara showed quantitative inheritance. In future work, qBSfR11 should be an effective QTL for pyramiding to enhance BS resistance.

Characteristics of bsr1-NIL

By marker-assisted selection of foreground and background and BS resistance, bsr1-NIL was selected as a candidate for a practical variety with resistance to BS.

Bipolaris oryzae is genetically diverse in Bangladesh (Kamal and Mia 2009), the Philippines (Burgos et al. 2013), India (Archana et al. 2014), and Iran (Ahmadpour et al. 2018). Inoculation of seedlings of 80 rice varieties with...
Table 2. Agronomic traits of *bsr1*-NIL and Mienoyume in 3 years (2018–2020)

| Test site | Line or variety | BS disease score (0–5) | Days to heading (days) | Heading date | Ripening date | Culm length (cm) | Panicle length (cm) | 1000-grain number (g m⁻³) | 1000-grain weight (g) | Protein content of brown rice (%) | Grain appearance (%) |
|-----------|----------------|------------------------|------------------------|--------------|---------------|------------------|---------------------|--------------------------|---------------------|-----------------------------|---------------------|
|           |                |                        |                        |              |               |                  |                     |                          |                     |                            |                     |
| Matsuoka  | *bsr1*-NIL    | 0.0 ± 0.1              | 81.3 ± 2.5             | 8.04         | 9.06          | 72.1 ± 5.4       | 20.8 ± 0.5          | 639 ± 12                 | 4006.6 ± 47.2        | 23.0 ± 0.8                   | 7.0 ± 0.5               |
|           | Mienoyume     | 0.8 ± 1.1              | 81.3 ± 2.5             | 8.04         | 9.06          | 73.4 ± 6.2       | 20.8 ± 0.5          | 605 ± 14                 | 3884 ± 39.3           | 22.5 ± 0.8                   | 7.2 ± 0.6               |
|           | *test*        |                        |                        |              |               |                  |                     |                          |                     |                            |                     |
| Iga       | *bsr1*-NIL    | 0.4 ± 0.5              | 86.3 ± 5.2             | 8.04         | 9.11          | 72.0 ± 7.1       | 20.5 ± 0.7          | 670 ± 30                 | 3985 ± 21.4           | 23.9 ± 0.3                   | 6.6 ± 0.1               |
|           | Mienoyume     | 2.1 ± 1.4              | 86.3 ± 5.2             | 8.04         | 9.11          | 74.4 ± 6.9       | 20.6 ± 0.6          | 635 ± 25                 | 3993 ± 13.0           | 23.4 ± 0.1                   | 6.9 ± 0.0               |
|           | *-test*       |                        |                        |              |               |                  |                     |                          |                     |                            |                     |

Values of each agronomic trait are shown as means ± SD over 3 years. BS disease score was ranked on a scale of 0 (no incidence) to 5 (severe) by visual survey at maturity, different from the method of Matsumoto *et al.* (2016). Yield and 1000-grain weight were calculated from filled grains screened through a 1.85-mm-mesh sieve, at a moisture content of 15%. Protein content of brown rice is expressed on a dry-weight basis of filled grains evaluated by near-infrared reflectance spectroscopy (6500HON, Nireco, Tokyo, Japan). Grain appearance is the ratio of perfect grains to filled grains, evaluated by a grain rice quality inspector (RN500, Kett, Tokyo, Japan). Length and width of 1000 filled grains were measured with a Satake Rice Analyzer (RGQ110B, Satake, Hiroshima, Japan). *Significant at 5%. p < 0.10 is indicated.

Table 3. Agronomic traits of *bsr1*-NIL and Mienoyume in fields with different degree of BS in 2020

| Test site (Degree of BS) | Line or variety | BS disease score (0–9) | Heading date | Ripening date | Plant height (cm) | Stem number (m⁻²) | SPAD value | Brown rice yield (g m⁻³) | Yield comparis (°%) | Panicle number (m⁻³) | Spikelet number per panicle (%) | Percentage of filled spikelets (%) | 1000-grain weight (g) | Protein content of brown rice (%) | Grain appearance (%) | Grains shape |
|-------------------------|----------------|------------------------|--------------|---------------|------------------|-------------------|-------------|-------------------------|-------------------|---------------------|---------------------------------|-----------------------------------|-------------------|---------------------------------|----------------------|-------------|
| Yield-trial field (Mild) | *bsr1*-NIL    | 1.0 ± 0.0              | 8.12         | 9.09          | 79.9 ± 2.1      | 5659 ± 41.1      | 36.1 ± 2.5   | 474 ± 42                 | 128.8             | 3527 ± 2.6          | 87.3 ± 3.2                      | 67.1 ± 6.4                       | 27.7 ± 0.2        | 7.7 ± 0.1                        | 91.1 ± 1.4           | 48.1 ± 0.1 |
| Mienoyume               | 3.0 ± 0.0      | **                      | 8.12         | 9.09          | 81.8 ± 1.1      | 6007 ± 60.0     | 36.8 ± 0.8   | 368 ± 18                 | 100.0             | 3836 ± 18.5         | 82.1 ± 10.2                     | 54.8 ± 3.1                       | 22.3 ± 0.0        | 9.0 ± 0.6                        | 87.8 ± 1.7           | 5.06 ± 0.06 |
| *-test*                 |                |                        |              |               |                  |                   |             |                        |                   |                     |                                 |                                     |                   |                                 |                      |             |
| Mienoyume               | 3.0 ± 0.0      | **                      | 8.12         | 9.09          | 79.9 ± 2.1      | 5659 ± 41.1      | 36.1 ± 2.5   | 662 ± 37                 | 100.0             | 3701 ± 4.9          | 83.6 ± 2.1                      | 89.0 ± 1.9                       | 24.0 ± 0.0        | 6.8 ± 0.1                        | 90.8 ± 0.4           | 5.02 ± 0.02 |
| *-test*                 |                |                        |              |               |                  |                   |             |                        |                   |                     |                                 |                                     |                   |                                 |                      |             |

Values of each agronomic trait are shown as means ± SD of three replications in 2020. Both fields were set up in Iga. Yield and 1000-grain weight were calculated from filled grains screened through a 1.85-mm-mesh sieve, at a moisture content of 15%. Brown rice yields were compared as that of *bsr1*-NIL divided by that of Mienoyume. Percentage of ripened spikelets was calculated as number of filled spikelets divided by total number of spikelets. Protein content of brown rice is expressed on a dry-weight basis of filled grains evaluated by near-infrared reflectance spectroscopy (6500HON, Nireco, Tokyo, Japan). Grain appearance is the ratio of perfect grains to filled grains, evaluated by a rice grain quality inspector (RN500, Kett, Tokyo, Japan). Length and width of 1000 filled grains were measured with a Satake Rice Analyzer (RGQ110B, Satake, Hiroshima, Japan). Significant at *5%, **1%, ***0.1%. p < 0.10 is indicated.
The impact of BS infection to agricultural traits and the effect of qBSfR11 in improving rice yield and quality

In the BS resistance test field, bsr1-NIL had 28.8% higher yield than Mienoyume (Table 3). We inferred that the reason was the higher percentage of filled spikelets and larger 1000-grain weight. There were no significant differences in growth characteristics between bsr1-NIL and Mienoyume at the panicle formation stage, when BS was mild. BS became more severe after heading, and so is likely to affect ripening. In addition, it was presumed that the higher percentage of filled spikelets was the effect of qBSfR11 of suppressing BS and the larger 1000-grain weight was the effect of new genes related to grain width in the qBSfR11 region. The protein content of brown rice of bsr1-NIL was significantly lower than that of Mienoyume. Vidhyasekaran and Ramadoss (1973) reported that severe infection reduced both yield (~20% to 40%) and quality (i.e., increased protein content), as here. Dallagnol et al. (2014) reported that BS reduced yield by reducing grain number per panicle, 1000-grain weight, and the percentage of filled grains. Aluko (1975) reported that severe infection reduced grain number per panicle and individual grain weight, resulting in a yield loss of 30% to 43%, compared with only 12% under moderate infection. The BS pathogen attacks the rice plant from seedling to milk stage (Sunder et al. 2014). The degree of yield loss and contributing factors are thought to vary depending on the degree and timing of BS infection. If BS is serious at an earlier stage than here, there is a high possibility that BS will affect not only ripening, but also yield components such as panicle number, and damage will be greater. Because BS resistance QTL, qBS11, which detected in the same region as qBSfR11 (Sato et al. 2015), has resistance to BS at the seedling stage of rice (Sato et al. 2008), bsr1-NIL is expected to have resistance even if BS occurs at an earlier stage than here. On the other hand, bsr1-NIL had a lower yield and a higher protein content of brown rice (lower quality) in BS severe conditions than in mild conditions, although its yield decrease and protein content increase were smaller than those of Mienoyume (Table 3). As the resistance type of bsr1-NIL with qBSfR11 is not true resistance but field resistance, pyramiding of QTLs is required for further enhancement of BS resistance.

First practical BS resistant variety bred using MAS

We submitted bsr1-NIL for variety registration with the Ministry of Agriculture, Forestry and Fisheries in Japan as ‘Mienoyume BSL’ (where BSL = Brown Spot resistance Line). This is the world’s first practical BS resistant variety bred using MAS.

Author Contribution Statement

KM designed the research and wrote the manuscript; KM and YO mainly performed the experiments and analyzed data; TY, TO, YH performed phenotypic examinations of...
NILs; SS selected individuals with MAS for foreground; RM and HS selected individuals and lines with MAS for foreground and background, and oversaw and improved manuscript.

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