Protein Phosphatase 2A Catalytic Subunit PP2A-1 Enhances Rice Resistance to Sheath Blight Disease

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Rice (Oryza sativa) production is damaged to a great extent by sheath blight disease (ShB). However, the defense mechanism in rice against this disease is largely unknown. Previous transcriptome analysis identified a significantly induced eukaryotic protein phosphatase 2A catalytic subunit 1 (PP2A-1) after the inoculation of Rhizoctonia solani. Five genes encoding PP2A exist in rice genome, and these five genes are ubiquitously expressed in different tissues and stages. Inoculation of R. solani showed that the genome edited pp2a-1 mutants using the CRISPR/Cas9 were more susceptible to ShB than the wild-type control, but other PP2A gene mutants exhibited similar response to ShB compared to wild-type plants. In parallel, PP2A-1 expression level was higher in the activation tagging line, and PP2A-1 overexpression inhibited plant height and promoted the resistance to ShB. PP2A-1-GFP was localized in the cytoplasm and nucleus. In addition, R. solani-dependent induction kinetics of pathogen-related genes PBZ1 and PR1b was lower in pp2a-1 mutants but higher in PP2A-1 activation line compared to those in the wild-type. In conclusion, our analysis shows that PP2A-1 is a member of protein phosphatase, which regulates rice resistance to ShB. This result broadens the understanding of the defense mechanism against ShB and provides a potential target for rice breeding for disease resistance.

Keywords: PP2A-1, sheath blight, resistance, enhance, rice

INTRODUCTION

R. solani is the causative agent of ShB in rice (Savary et al., 1995; Suryadi et al., 2013), and which damages rice during the entire growth period, and predominantly targets the leaves, sheaths, and panicles, eventually resulting in the withering and lodging of the entire plant. A severe form of ShB can lower the rice produce by ∼50% (Savary et al., 2000). The rapid variation, wide host range, and high survival ability of the pathogen can make the disease control more challenging (Taheri and Tarighi, 2011; Yellareddygari et al., 2014; Singh et al., 2019). Currently, there is a dearth of ShB-resistant cultivars, therefore, the strategies to quell ShB involve the use of fungicides (Savary et al., 2000). However, fungicides directly affect the living environment of other microorganisms and increase the cost of cultivation. Thus, to develop ShB resistance in rice, it is necessary to isolate resistant cultivars and understand their underlying defense mechanisms against ShB.
Extensive studies have been performed to investigate the mechanism of rice defense against ShB. Overexpression of chitinase, β-1,3-glucanase, or OsPGIP1 (polygalacturonase-inhibiting protein) (Shah et al., 2009; Mao et al., 2014; Zhu et al., 2019), OsACS2 (key enzyme in ethylene synthesis) (Helliwell et al., 2013), OsGSTU5 (tau class glutathione-S-transferase 5) (Tiwari et al., 2020), and Os2H16 (Li et al., 2013, 2018) were found to promote rice resistance to ShB. In addition, BSR2 (broad-spectrum resistance 2) (Maeda et al., 2019) or a transcription factor complex including LPA1 (indeterminate domain 14, IDD14) and IDD13 (Sun et al., 2019, 2020) were reported to positively regulate rice resistance to ShB while SWEET11 (sugar will eventually be exported transporter 11) (Gao et al., 2018) exhibited a negative regulation. The transcription factor OsWRKYs also plays an important role in resistance to sheath blight (Peng et al., 2012, 2016; Wang et al., 2015; Jimmy and Babu, 2019; Yuan et al., 2020). In addition, salicylic acid-dependent immunity showed a positive regulation in ShB resistance in rice and Brachypodium distachyon (Kouzai et al., 2018).

Protein phosphatase also plays an important role in plant defense response. The protein phosphatases (PPs) with a vast array of structures and functions are mainly categorized as serine/threonine (Ser/Thr) PPs and protein tyrosine phosphatases (PTPs). PP1, PP2A, PP2B, and PP2C account for the sub-divisions of the protein tyrosine phosphatase group. The PP2A complex comprises three subunits: A, B, and C with scaffolding, regulatory, and catalytic roles, respectively (Yu et al., 2005; Durian et al., 2016). The role of PP2A protein in plant abiotic stress signal transduction has been confirmed. For instance, drought and elevated salinity induce high levels of OsPP2A-1 and OsPP2A-3, the closely associated genes coding for the C-subunit of PP2A (Yu et al., 2003). In Arabidopsis, the growth of roots and shoots is augmented by PP2A-C5 overexpression in the presence of several salts indicating the vital function of protein in growth to combat salinity (Hu et al., 2017). AtPP2A is involved in acclimation to light as well as when responding to pathogens, both based on the regulation of ROS (Rahikainen et al., 2016; Máté et al., 2019). Exposure of wheat to R. cerealis or hydrogen peroxide showed elevated TaPP2A-4B and TaPP2A-4D RNA levels revealing the involvement of PP2A in the biotic stress response. Silencing of TaPP2A in wheat boosted the expression of ROS-scavenging and pathogenesis-related (PR) RNA molecules (Zhu et al., 2018). Resistance to Botrytis cinerea and leaf senescence in Arabidopsis involves the role of PP2A-Bγ. The swift induction of the gene coding for the heterotrimERIC PP2A catalytic subunit, LePP2A-1 was observed when resistant tomato plants were challenged with Pseudomonas syringae pv. tomato (a virulent strain) (He et al., 2004). A mutation which was isolated from rice blast fungus was inserted into the promoter region of MoPPG1, a ser/thr-PP2A catalytic subunit (PP2Ac) gene, which made the mutant defective in the growth of vegetative mycelium and could not cause disease (Du et al., 2013). Fusarium graminearum contains three kinds of PP2A (FgPP2A, FgSit4, and FgPgl), which play a key role in the growth, development, and pathogenicity of fungi (Liu et al., 2018). Our recent transcriptomic study showed the sensitivity of PP2A-1 expression to R. solani infection (Yuan et al., 2020). However, PP2A function in rice defense to ShB is unknown.

In this study, PP2A-1 was significantly induced following R. solani inoculation. Further bioinformatics, genetic, and molecular analyses were performed to identify the function of PP2A family members in rice defense to ShB. Our results broaden the knowledge of the underlying ShB defense mechanisms and provide a potential target for resistant breeding in rice.

MATERIALS AND METHODS

Plant and Fungal Materials

Four rice lines/cultivars, including Japonica rice cultivar Dongjin (DJ), Zhonghua11 (ZH11), pp2a-1 CRISPR/Cas-9 genome editing mutants in ZH11 background, and PP2A-1 activation tagging line (PP2A-1 OX) in DJ background were used in this study. All the rice lines used in this study were grown in a greenhouse in natural light. The type strain used in this study was R. solani AG1-IA.

Construction of the CRISPR/Cas9 Plasmids

The human codon-optimized hSpCas9 (Cong et al., 2013) was linked to the maize ubiquitin promoter (UBI) in an intermediate plasmid followed by its insertion into a binary pCAMBIA1300 vector (Cambia, Australia) harboring the HPT (hygromycin B phosphotransferase) gene. A point mutation kit (Transgen, China) was used to eliminate the original BsaI site in the backbone of pCAMBIA1300. A OsU6 promoter fragment (Feng et al., 2013), ccdB, a gene for negative selection flanked by two BsaI sites, and a pX260-derived sgRNA (Cong et al., 2013) were inserted employing an In-Fusion cloning kit (Takara, Japan) into this vector to produce the CRISPR/Cas9 binary vector pBGK032 (Figure 1). The vector was maintained in Escherichia coli strain DB3.1.

The targeting specificity and the selection of the 23-bp targeting sequences (including PAM) was done employing a BLAST search (http://blast.ncbi.nlm.nih.gov/Blast.cgi) against the rice genome (Hsu et al., 2013). The designed targeting sequences were annealed to synthesize the oligo adaptors. The vector pBGK032 was restriction digested using BsaI and purified employing a DNA purification kit (Tiangen, China). This was ligated with 0.05 mM of oligo adaptor (10 μL) resulting in CRISPR/Cas9 plasmids, which were directly transformed in competent E. coli cells.

Transformation of Plants and Mutation Detection

Agrobacterium tumefaciens strain EHA105 was transformed with the CRISPR/Cas9 plasmids followed by rice transformation following an earlier published protocol (Nishimura et al., 2006). The genomic DNA from these transformants was extracted, and PCR was conducted employing primer pairs flanking the designed target site. The Degenerate Sequence Decoding approach was applied to directly sequence and identified the PCR products (300–500 bp) method (Ma et al., 2015).
RNA Extractions
The total cellular RNA was extracted from the 1-month-old plant sheath, roots, leaves, or 3-month-old flower tissues. For analysis of *R. solani*-mediated gene expression, Trizol reagent (Invitrogen, China) was used to isolate the total RNA from 0.1 g of rice leaves, roots, leaf sheath, and flowers. Then the product was purified using the RNeasy mini kit (Promega, China) and RNase-Free DNase I (Promega, China) following manufacturer's instructions. The cDNAs were synthesized with M-MLV Reverse Transcriptase (Promega) kit following manufacturer's instructions.

Sequence Analysis of PP2A
The PP2A amino acid sequences were isolated from *Arabidopsis*, rice, tobacco, and tomato to perform phylogenetic analysis. MEGA7 software was used for multiple sequence alignment of the original sequences. The comparison results were edited by GeneDoc to export the multi-sequence alignment results graph. MEGA7 software was used for phylogenetic tree construction using the nearest neighbor-joining method (Kumar et al., 2016).

cDNA Synthesis and qRT–PCR
Reverse transcription using 2 µg of each purified RNA sample was done using a Prime Script TMRT Reagent Kit with gDNA Eraser (TaKaRa, China) in accordance with the provided instructions. qRT-PCR was performed on the ABI 7500 RT-PCR system (Applied Biosystems, United States). The composition of the mix was: 10 µL 2× SYBR Premix Ex Taq, 0.4 µL 50× ROX Reference Dye II, 0.4 µM of each primer, and 5 µL of the cDNA template (50-fold dilution) in a net volume of 20 µL. The conditions were: 95°C for 30 s; 95°C for 5 s, 58°C for 15 s, and 72°C for 34 s for 40 cycles. The 2^((−ΔΔC(T)) approach was employed to estimate the expression levels of target gene(s) relative (Livak and Schmittgen, 2001). *Ubiquitin* was used as an internal reference. Table 1 presents the primers in this study.

Inoculation With *R. solani* and Scoring Response of Rice Plants
Rice plants were grown in the glasshouse for 1 month prior to inoculation with the pathogen *R. solani* AG1-IA. The second leaf of the main tiller was cut into 10-cm slices, placed on wet
TABLE 1 | Sequence of the primers used in this study.

| Primer      | Sequence                                      |
|-------------|-----------------------------------------------|
| Ubiquitin F | CACCGTTCAACACACATCCAG                       |
| Ubiquitin R | TGAAGACCCCTGACTGGGAAG                       |
| PP2A-1 F    | CACGGTGTTGCGGCTGGTCTGGT                      |
| PP2A-1 R    | CGCGTTGTGTCCGGCTCTATTTT                      |
| PP2A-2 F    | GCTAGACCTACGATGCTTCTGGT                     |
| PP2A-2 R    | TACACTGCTGCTTCCTGCTTTG                      |
| PP2A-3 F    | CTCTCATCGCAAGGACACTAA                       |
| PP2A-3 R    | TGTGTGCTGTCACATTGGCGAGGAAG                   |
| PP2A-4 F    | CGAAGAAGTCTGGTACCCATATTCC                   |
| PP2A-4 R    | ATCAAGTTGTTCCTGGTCACATC                     |
| PP2A-5 F    | TAGCTGGGTCTGAAACTGTTATG                     |
| PP2A-5 R    | AAATACGGGGCTTCTGGTGTGAC                     |
| PBZ1 F      | CCGTCGCGAATGGCTCTAA                        |
| PBZ1 R      | CTCAAACCGACGGAGATTTG                       |
| PR1b F      | CGCTGCTCATACAGTGAACTA                      |
| PR1b R      | ACCTGAAAAGGAGAAAAGCAGGG                     |
| PP2A-1 GFP F| CATTAGGTGGCGGTGCTGGGCTGC                   |
| PP2A-1 GFP R| AGATCTCATAAAGGTCTGGGTCTGGCG                |

filter paper and stored in a culture dish (36 × 36 × 2.5 cm). In a completely randomized design, five leaves were placed in each plate, with a total of three replicate plates for each treatment. The fungal plug (7 mm in diameter) was cut from the Potato Dextrose Agar (PDA) plate with R. solani and placed on the back of the leaf. The leaves were cultured for 72 h at 25°C under continuous light, and the moisture of the filter paper was maintained with sterile water (Gao et al., 2018). Measurement from 0 (no lesion) to 9 (lesions occupying 90–100% of the leaf surface) was done after visual observation. Scores from one to eight represented 10–80% diseased leaf area (Prasad and Eizenga, 2008).

Rice plants that were cultured in a greenhouse for 1 month prior to the tillering stage were used for inoculation. The sheath of the first leaf of the main stem was inoculated with R. solani AG1-IA. The PDA fungal plug was inoculated into rice leaf, sprayed with sterile water, and the severity of the disease was determined after 24, 48, and 72 h.

Construction of PP2A-1-GFP Plasmid and Its Subcellular Localization

PP2A-1 ORF region was amplified by PCR and moved into pCAMBIA1302 vector to create PP2A-1-GFP plasmid. The Agrobacterium-mediated transient expression approach was followed to introduce the fusion proteins into Nicotiana benthamiana (Kim et al., 2009). The location of the protein was monitored via GFP fluorescence with a confocal microscope (SP5; Leica, Solms, Germany).

Statistical Analysis

The significant differences between different groups were analyzed using Microsoft Excel to compute the mean, standard deviation, and the Student’s t-test. Dunnett’s test was done employing the SPSS 19.0 statistical software.

RESULTS

Inoculation of R. solani Significantly Induced PP2A-1 Expression

Our previous transcriptome analysis identified that PP2A-1 expression was induced by the inoculation of R. solani AG1-IA (Yuan et al., 2020). Rice genome harbors five PP2A isoforms, and a phylogenetic analysis of PP2A proteins from Arabidopsis, rice, tobacco, and tomato revealed that OsPP2A-1 clustered with OsPP2A-3, NtPP2A-2, AtPP2A-1, AtPP2A-2, and AtPP2A-5, all of which belong to Subfamily II, while OsPP2A-5, OsPP2A-4, NtPP2A-2, AtPP2A-1, AtPP2A-2, AtPP2A-5, and LePP2A-5 belong to Subfamily I (Figure 1A). The homologous sequence alignment of PP2As showed that OsPP2A-1 and OsPP2A-3 shared 98% similarity. The red box indicates a highly conserved region between the five PP2As (Figure 1B). qRT-PCR of the 5 PP2A genes was done for the verification of the transcriptome data post-inoculation with R. solani after 0, 24, 48, and 72 h. The results indicated that only PP2A-1 expression was induced by R. solani infection, and PP2A-1 expression was the highest 72 h after inoculation, while the other four PP2A genes did not respond to R. solani. PBZ1, a marker gene was used for evaluating pathogen infection, its expression was down-regulated at 24 h after inoculation, while it was up-regulated after 48 h and 72 h of inoculation (Figure 1C; Table 2). In addition, tissue-specific expression of PP2As was examined by qRT-PCR. All PP2As were expressed in root, leaf sheath, leaf, and flower tissues, while PP2A-1 was expressed highly in root and flower, indicating that PP2As were ubiquitously expressed in different tissues and developmental stages (Figure 1D).

pp2a-1 Mutants Are Susceptible to ShB

To analyze the function of PP2A genes in rice defense to ShB, Crispr/Cas9 induced genome editing mutants for PP2A were generated. The PP2A genes consist of multiple exons and introns in the genome (Figure 2A). The sequencing of PP2A genome editing mutants revealed that pp2a-1 mutants have a genomic lesion in the first exon with 1 or 2-bp insertions (pp2a-1-1, pp2a-1-2) and the pp2a-2 mutant has a 1-bp deletion in the 11th
exon. The **pp2a-3**, **pp2a-4**, and **pp2a-5** mutants contained edited sequences in the first exon with a 1-bp insertion, 2-bp insertion, and 1-bp deletion, respectively (**Figure 2A**). After inoculation with *R. solani* AG1-IA, **pp2a-1** genome editing mutants in ZH11 background were more susceptible than ZH11 plants, showing obvious chlorosis (**Figure 2B**), while other **pp2a** genome editing mutants in DJ background had no obvious disease grade differences compared with that in wild-type plants (DJ).
FIGURE 4 | PBZ1 and PR1b expression patterns in wild-type, pp2a-1, and PP2A-1 overexpression plants. (A) PBZ1 and (B) PR1b expression after R. solani inoculation. The PBZ1 and PR1b gene expression patterns were evaluated after 0 and 48 h following R. solani inoculation. ZH11 is the control of pp2a-1-1 and pp2a-1-2, while DJ is the control of PP2A-1 OX plants. Normalization of expression was done with reference to the internal control ubiquitin. The experiments were done in triplicate. Significant changes are illustrated by various letters (P < 0.05).

PP2A-1 Overexpression Enhanced Rice Resistance to ShB

Since pp2a-1 mutants were susceptible to ShB, the PP2A-1 overexpression plants were further examined in response to ShB. We isolated a PP2A-1 activation tagging line (PP2A-1 OX) in a T-DNA insertional library (Jeong et al., 2002). In the activation tagging line, T-DNA was inserted in the promoter region in which four copies of the 35S promoter activated the PP2A-1 expression (Figure 3A). The qRT-PCR results indicated that PP2A-1 expression was significantly higher in hetero- and homozygous PP2A-1 activation tagging plants than wild type and the PP2A-1 expression level was significantly higher in homozygous compared to heterozygous plants (Figure 3B). PP2A-1 OX lines displayed a semi-dwarf phenotype, with homozygous plants significantly shorter than heterozygous and wild-type plants (Figure 3C). Next, we selected the homozygous PP2A-1 OX to inoculate R. solani AG1-1A. The lesion area of PP2A-1 homozygous overexpression plants was smaller than that of DJ (wild-type) after 48 h of inoculation (Figure 3D). The lesion coverage on leaves was 48.1 and 30.8%, respectively (Figure 3E), indicating that overexpression of PP2A-1 enhanced rice resistance to ShB. In addition, PP2A-1-GFP and free GFP were expressed in tobacco leaves, and PP2A-1-GFP signal was detected in the cytoplasm and nucleus (Figure 3F).

PP2A-1 Positively Regulates Defense Gene Expression

PP2A-1 OX plants were less susceptible while pp2a-1 mutants were more susceptible to ShB compared to the wild-type control. The expression patterns of defense genes PBZ1 and PR1b in wild-type, pp2a-1, and PP2A-1 OX plants were examined following R. solani inoculation. qPCR results showed that there was no significant difference in the expression levels of PBZ1 and PR1b among wild-type, genome editing mutants, and overexpression lines with no R. solani inoculation. However, the expression level of PBZ1 in pp2a-1 mutants was significantly lower than that in control ZH11, while it was higher in PP2A-1 OX plants than that in wild-type DJ after 48 h of inoculation (Figure 4A). The expression pattern of PR1b was similar to that of PBZ1 at 48 h of inoculation, which showed lower and higher induction kinetics.
in pp2a-1 mutants and PP2A-1 OX compared to that in wild-type plants, respectively (Figure 4B).

DISCUSSION

Rice ShB caused by R. solani, bacterial leaf blight caused by Xanthomonas oryzae, and rice blast caused by Magnaporthe oryzae are three major diseases that significantly affect rice yield in China (Suryadi et al., 2013). The use of resistant varieties has been the primary means of disease control. However, due to the lack of resistant varieties and that sclerotium has a strong reproductive capacity, ShB control is challenging. Therefore, it is necessary to screen disease resistance genes and understand their resistance mechanism. Previous studies showed that PP2A regulates the development of lateral and primary roots, seed germination, and abiotic stress response against high concentration of sugar, salt, and drought (Yu et al., 2003; País et al., 2009; Liu et al., 2014; Hu et al., 2017). PP2A also plays important roles in biotic stress resistance. For example, AtPP2A is involved in regulation of PTI (pathogen-associated molecular pattern-triggered immunity) upon response to P. syringae pv. tomato (Pto) DC3000 infection; LePP2A gene was rapidly induced by inoculating with the model strain P. syringae pv. tomato; TaPP2A-4B and TaPP2A-4D may negatively regulate wheat defense response to R. cerealis infection by triggering the generation of ROS and PTI-mediated induction of PR genes (He et al., 2004; Segonzac et al., 2014; Durian et al., 2016; Zhu et al., 2018), suggesting that PP2A may be a key regulator of PAMP induced immunity. In rice, the induction of okadaic acid-dependent AMY3 and RCht2 (rice chitinase) transcription are regulated via the PP2A signal transduction pathway (Luan et al., 1993; Kim et al., 1998). However, the role of PP2A in rice disease resistance still remains unclear.

Our results indicate that the protein phosphatase 2A catalytic subunit OsPP2A-1 enhances resistance to sheath blight disease in rice. In our previous transcriptome analysis, OsPP2A-1 was significantly induced by R. solani inoculation (Yuan et al., 2020). The CRISPR/Cas9-mediated genome editing lines revealed higher susceptibility of pp2a-1 mutants to ShB, compared to wild-type control and other PP2A mutants (Figure 2). While PP2A-1 OX lines displayed a semi-dwarf phenotype, homozygous plants were significantly shorter than heterozygous and wild-type plants (Figure 3C). Inoculation of R. solani AG1-IA demonstrated that the PP2A-1 OX lines were less susceptible to ShB. The PP2As are ubiquitously expressed in different stages and tissues, and PP2A-1-GFP was localized at the cytosol and nucleus in tobacco leaves. The PP2A-1 expression level was significantly higher in the activation tagging line, with higher PP2A-1 expression inhibiting plant height while promoting ShB resistance.

The vital involvement of PP2A in responding to pathogens by plants has been demonstrated in recent studies (He et al., 2004; Zhu et al., 2018). For example, in A. thaliana, RLKs FLS2 (flagellin sensing receptor2) recognizes and EFR (EF-Tu receptor) is capable of recognizing the EF- Tu (elongation factor), both are PAMPs (pathogen-associated molecular pattern) of bacterial pathogens. The autophosphorylation and functioning of BAK1 (BR11-associated kinase 1) is limited by PP2A-holoenzymne (Segonzac et al., 2014). PP2A-c4 and PP2A-a1 gene knockout mutants display a stronger resistance to virulent P. syringae pv. tomato DC3000 (Segonzac et al., 2014). The BSMV-VIGS (barley stripe mosaic virus–induced gene silencing) approach was applied to augment R. cerealis resistance in wheat attributed to TaPP2A-4C and TaPP2A-4D knock-down, suggesting the negative regulation of TaPP2A to wheat sharp eyespot. In many species, PP2A appears to function as a negative regulator, while OsPP2A-1 was found to positively regulate resistance to R. solani in this study. It seemed that the same gene may play different functions in response to infection of different types of pathogens. For example, WRKY transcription factors were more resistant to the hemibiotrophic bacterial pathogen P. syringae, but more susceptible to necrotrophic fungal pathogen B. cinerea in Arabidopsis (Xu et al., 2006), implying that PP2A-1 might play diverse functions when experiencing different stimuli. In our study, we confirmed that the CRISPR/Cas9-induced pp2a-1 gene editing mutants more susceptible to R. solani, while the other pp2as were similar to wild-type plants in response to R. solani infection. It may be valuable to dissect the associated molecular mechanism in the future research.

As mentioned earlier, PP2As comprises three subunits (A, B, and C). In the A. thaliana genome, these subunits are encoded by five genes of subunit C, three genes of subunit A, and 17 genes of subunit B (Farkas et al., 2007) to establish a minimum of 255 novel forms of the molecule. Immunity in plants is influenced by subunits A and B (with scaffolding and regulatory functioning, respectively). Resistance to P. syringae pv. tomato was augmented due to a subunit B-B’/0 deficient mutation (Kataya et al., 2015). AtPP2A-B’γ enhances the negatively regulated defense against Myzus persicae (green peach aphid) and B. cinerea (a necrotrophic fungus) (Trotta et al., 2011; Rasool et al., 2014). PR protein phosphorylation (PR1, PR2–PR5) is augmented by mutations in subunit AtPP2A-B’γ (Trotta et al., 2011). The constitutive expression of PR1a, PR1b, and PR5 was induced by NbNPP4-1 and NbNPP4-2 silencing in N. benthamiana (He et al., 2004). PR2 levels were up-regulated by TaPP2A silencing (Zhu et al., 2018). PBZ1, a PR10 family protein accumulates in rice tissues which are in the process of cell mortality (Huang et al., 2016; Moselhy et al., 2016). In this study, PBZ1 and PR1b genes were up-regulated by R. solani infection, suggesting that PBZ1 and PR1 play a role in ShB resistance in rice. The results indicated that R. solani-induced PBZ1 and PR1b expressions are under control of PP2A-1, suggesting that the expression of PP2A-1 might be through the activation of PR genes to promote rice defense.

Taken together, our findings suggest that the protein phosphatase 2A catalytic subunit, PP2A-1, regulates the defense response in rice to R. solani infection. This study revealed a new function of the rice PP2A in immune response, which provided a potential target for breeding ShB-resistant lines.
DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding authors.

AUTHOR CONTRIBUTIONS

QL, QM, and YX conceived and designed the studies and wrote the manuscript. QL, JC, VK, ZL, and DY collected and analyzed the data. All authors have read and approved the final version of the manuscript.

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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