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RESEARCH ARTICLE

OsWRKY9 is Involved in Transcriptional Regulatory Cascade Enhancing Broad-Spectrum Disease Resistance

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Abstract

Diverse pathogens, including *Fusarium fujikuroi* and *Xanthomonas oryzae* pv. *oryzae* (*Xoo*), cause significant yield losses in rice (*Oryza sativa*). The situation is expected to worsen due to rapid climate change. Thus, identifying novel genes conferring innate immunity against these pathogens is crucial for global food security. WRKY transcription factors are involved in various plant processes, including innate immunity. In rice, there are 125 OsWRKYs, with some functions reported. However, the roles of many OsWRKYs in rice immunity remain largely unknown. In this study, we investigate the role of OsWRKY9 in broad-spectrum disease resistance. *OsWRKY9* transcripts increased in response to *F. fujikuroi* and *Xoo*. The promoter of *OsWRKY9* was indirectly activated by OsWRKY65, which confers broad-spectrum resistance to *F. fujikuroi* and *Xoo*. Moreover, *OsWRKY9*-overexpressing transgenic plants exhibited enhanced resistance to both pathogens in a manner similar to transgenic plants overexpressing *OsWRKY65*. Additionally, OsWRKY9 modulated the expression of various defense-related genes regulated by OsWRKY65. These results indicate that the OsWRKY65-OsWRKY9 module enhances resistance to bakanae disease and bacterial blight.

Keywords *Fusarium fujikuroi* · Innate immunity · OsWRKY9 · OsWRKY65 · Rice · *Xanthomonas oryzae* pv. *oryzae*

Introduction

Rice (*Oryza sativa*) is one of the main cereal crops that provides food nutrient globally. However, global rice yields can be seriously damaged by bakanae disease and bacterial blight (Son et al. 2024a). *Fusarium fujikuroi* is a primary agent of bakanae disease. Although *F. fujikuroi* is classified as a hemibiotrophic pathogen, it acts as a necrotrophic pathogen in susceptible rice varieties (Matic et al. 2016; Cheng et al. 2020). To increase plant susceptibility, *F. fujikuroi* promotes gibberellin (GA) biosynthesis and activates GA signaling (Matic et al. 2016). In contrast, the jasmonic acid (JA)-dependent defense response is critical for rice immunity against *F. fujikuroi* (Matic et al. 2016; Cheng et al. 2020). However, the positive or negative regulators controlling the rice-*F. fujikuroi* interaction are largely unknown. *Xanthomonas oryzae* pv. *oryzae* (*Xoo*) is a major agent causing bacterial blight disease. Although *Xoo* is mostly considered biotrophic, this bacterial pathogen is predominantly biotrophic hemibiotrophs (De Vleesschauwer et al. 2013). *Xoo* employs various effectors to induce host susceptibility (Deb et al. 2021). For example, transcription activator-like effectors (TALEs), including PthXo1, PthXo2, PthXo3, and AvrXa7, directly activate the promoters of *SUGAR WILL EVENTUALLY BE EXPORTED TRANSPORTER 11* (*OsSWEET11*), *OsSWEET13*, and *OsSWEET14*, which are

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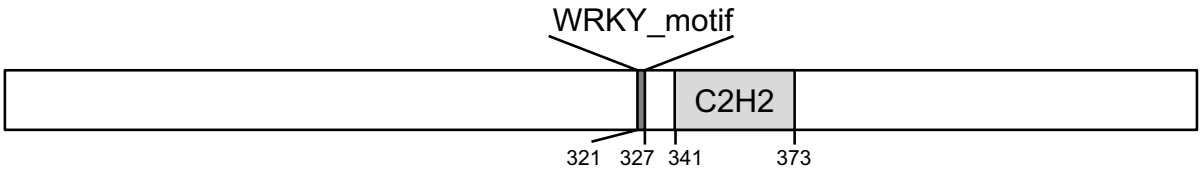

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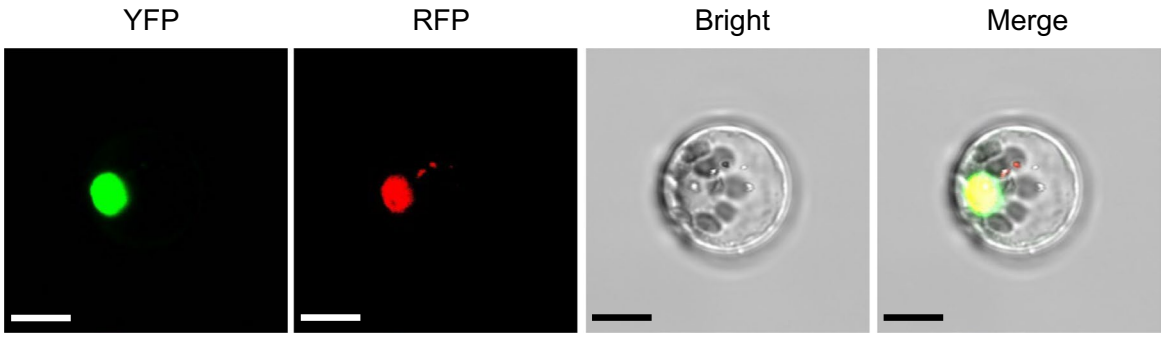

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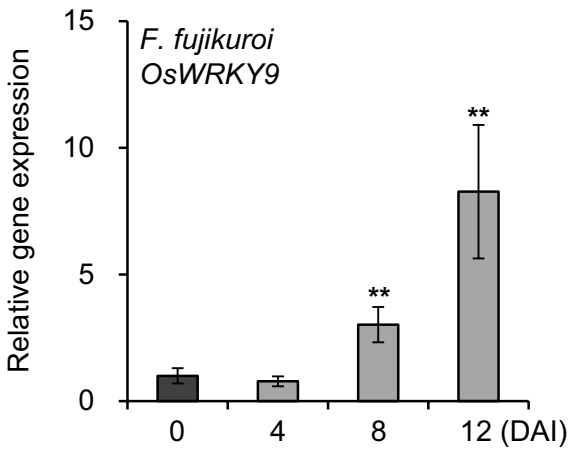

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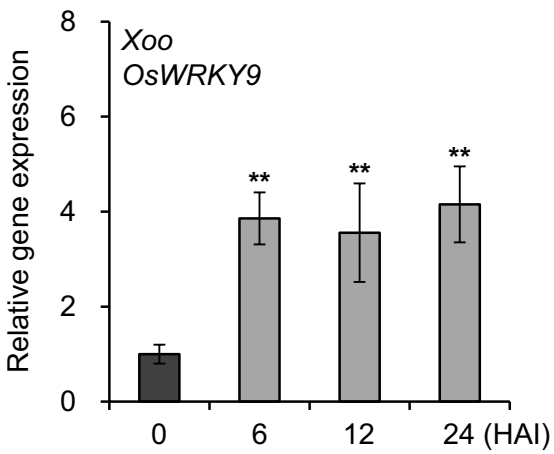


◂**Fig. 1** Subcellular localization and gene expression analysis of OsWRKY9. **A** Amino acid sequence and structural domains of OsWRKY9. The WRKY domain indicated by a solid line, the core WRKY motif by a dotted line, and the conserved zinc finger sequence by a triangle (▲). **B** Subcellular localization of OsWRKY9 in rice protoplasts. The N-terminal YFP conjugated OsWRKY9 was expressed in protoplasts with NLS-RFP and observed using confocal microscope. Scale bar: 8 μm. **C**, **D** Temporal expression analysis of *OsWRKY9*. Wild-type plants were inoculated with *F. fujikuroi* (**C**) and *Xoo* (D) for the indicated time. DAI, days after inoculation; HAI, hours after inoculation. Relative *OsWRKY9* transcription levels were quantified using RT-qPCR, with *OsActin* serving as the reference gene. Data are presented as means ± SD, with asterisks indicating values significantly different from the controls (** $P < 0.01$)

necessary for susceptibility to *Xoo* (Gupta et al. 2021). Given that mutagenesis of *OsSWEET13* and *OsSWEET14* led to enhanced resistance to various *Xoo* strains (Eom et al. 2019; Oliva et al. 2019), it has been suggested that transcription factors and signaling pathways regulating the expression of *OsSWEETs* in response to *Xoo* are crucial for bacterial blight resistance.

WRKY transcription factors play significant roles in plant immunity (Javed and Gao 2023). Transcriptome analyses suggest that various OsWRKYs are involved in rice immunity against *F. fujikuroi* and/or *Xoo* (Ji et al. 2016; Matic et al. 2016; Choi et al. 2017; Cheng et al. 2020). Indeed, OsWRKY114 enhances broad-spectrum disease resistance (BSDR) to *F. fujikuroi* and *Xoo* by regulating the expression of genes involved in defense responses and phytohormone signaling (Son et al. 2022; Song et al. 2023). Moreover, recent studies showed that OsWRKY65 induces BSDR to these pathogens by upregulating immune responses and downregulating susceptibility (S) genes, including *OsSWEETs* (Son et al. 2024b). OsWRKYs can regulate each other, and OsWRKY transcriptional regulatory cascades play key roles in disease responses. For instance, the OsWRKY45-OsWRKY13-OsWRKY42 module enhances defense responses to the fungal pathogen *Magnaporthe oryzae* (Cheng et al. 2015), and the OsWRKY80-OsWRKY4 module is involved in resistance to the fungal pathogen *Rhizoctonia solani* (Peng et al. 2016). Furthermore, transcriptional regulatory cascades comprising various OsWRKYs (e.g., OsWRKY6 and OsWRKY10) activate defense signaling against *Xoo* (Choi et al. 2015, 2020; Im et al. 2022). However, OsWRKY regulatory cascades are complex and remain largely unexplored.

Our previous study showed that OsWRKY65 induces the expression of various *OsWRKYs*, including *OsWRKY9*, *OsWRKY13*, and *OsWRKY30* (Son et al. 2024b). OsWRKY13 and OsWRKY30 function as positive regulators of defense responses to *Xoo* by transcriptionally regulating various target genes (Qiu et al. 2008; Jalmi and Sinha 2016; Wang et al. 2022). However, the biological role of OsWRKY9 remains unknown. In this study, we explored whether OsWRKY9 confers innate immunity against *F. fujikuroi* and *Xoo* and its involvement in the OsWRKY transcriptional regulatory cascade.

Materials and Methods

Plant Material and Growth Conditions

The *japonica* rice variety Ilmi was utilized in this study. Kernels were surface-sterilized with 70% ethanol and 2% sodium hypochlorite, germinated, and seedlings were grown in soil or Murashige and Skoog (MS) medium under a 16-h light/8-h dark cycle at 28 ℃.

Generation of the Transgenic Plants

OsWRKY65-overexpressing plants ($OsWRKY65^{OX}$) were previously generated and confirmed (Son et al. 2024b). *OsWRKY9*-overexpressing ($OsWRKY9^{OX}$) and *OsWRKY30*-overexpressing ($OsWRKY30^{OX}$) plants were generated previously described (Son et al. 2020). Briefly, the full-length cDNAs of *OsWRKY9* (LOC_Os01g18584) and *OsWRKY30* (LOC_Os08g38990) were amplified using specific primers (Table S1) and cloned into the pEarleygate104 vector using the Gateway system (Invitrogen, USA). The constructs were transformed into *Agrobacterium tumefaciens* strain LBA4404, which was then used for rice transformation. Transgenic plants were regenerated from calli and selected until T_3 homozygous plants were obtained.

Analysis of Gene Expression Pattern

Total RNA was extracted from the leaves of wild-type and transgenic plants using TRIzol Reagent (QIAGEN, Germany). 3 μg of total RNA was reverse-transcribed into cDNA using SuperScript III reverse transcriptase (Invitrogen, USA) following the manufacturer's protocol. Quantitative reverse-transcription polymerase chain reaction (RT-qPCR) was performed using specific primers (Table S1) as previously described (Son et al. 2024b). Gene expression levels were calculated using the comparative Ct method, with *OsActin* as the reference gene.

Subcellular Localization of OsWRKY9

Protoplasts were transiently transformed as previously described (Son et al. 2023). To determine the subcellular localization of OsWRKY9, the pEarleygate104/*OsWRKY9* construct was co-transfected with a nuclear localization signal-red fluorescent protein (NLS-RFP) construct into

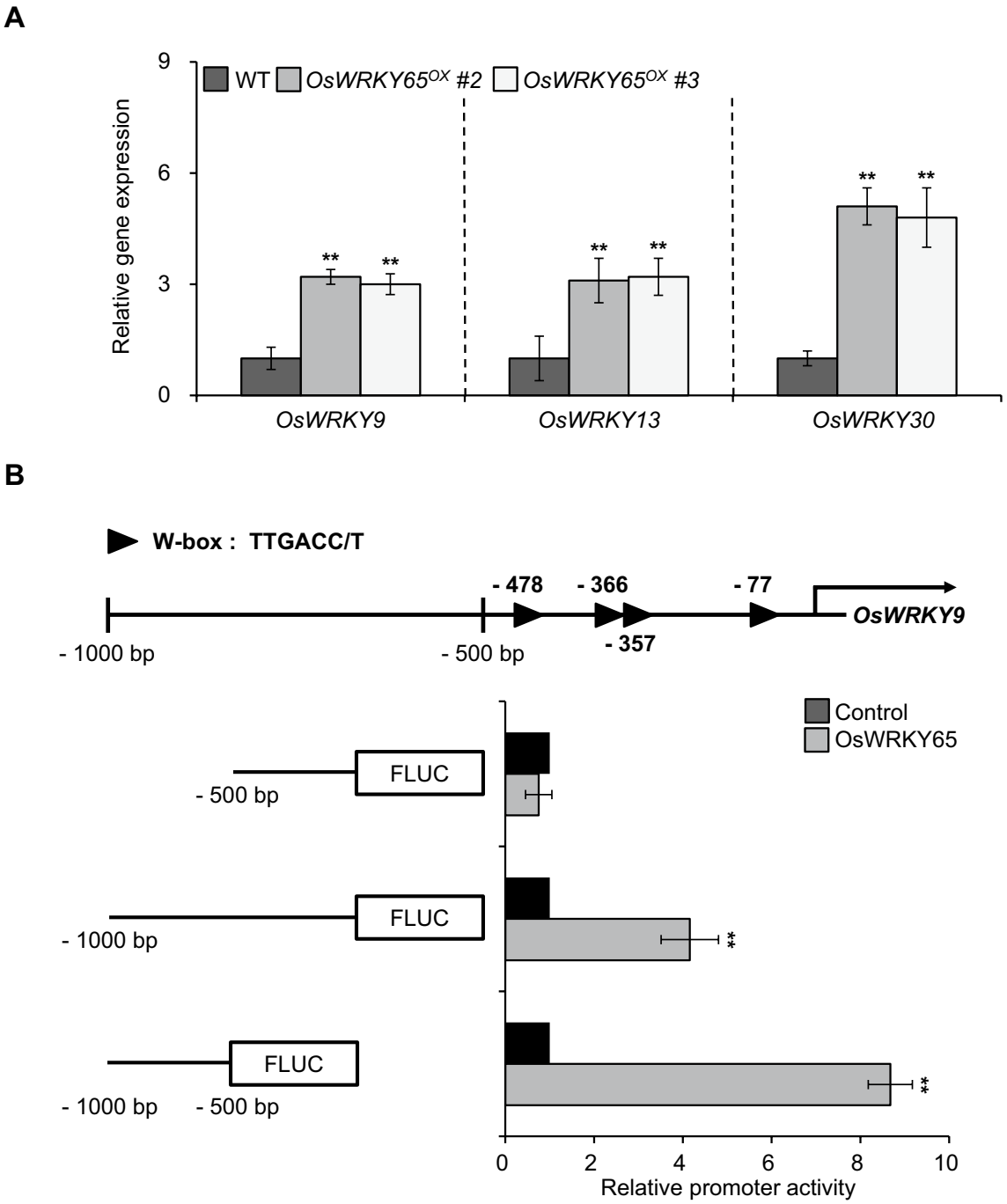


Fig. 2 Regulation of *OsWRKY9* by OsWRKY65. **A** Expression levels of *OsWRKY* genes in *OsWRKY65*^{OX}. Relative transcription levels of *OsWRKY9*, *OsWRKY13*, and *OsWRKY30* in 2-week-old *OsWRKY65*^{OX} and wild-type plants were quantified using RT-qPCR, with *OsActin* as the reference gene. Data are presented as means ± SD, with asterisks indicating values significantly different from the controls (** $P < 0.01$). **B** Promoter activity assays of truncated *OsWRKY9* promoter regions. Schematic positions and directions of the W-box motifs located within 1 kb upstream of the translation initiation site in *OsWRKY9* are shown. FLUC-conjugated truncated *OsWRKY9* promoters were co-expressed with OsWRKY65 in rice protoplasts, and relative FLUC activity was measured. RLUC activity from 35S served as the control. Data are presented as means ± SD, with asterisks indicating values significantly different from the controls (** $P < 0.01$)

protoplasts. After 10 h of incubation, fluorescence signals were observed using a Leica TCS SP8 confocal laser scanning microscope (Leica Microsystems, Germany). Yellow fluorescent protein signals were detected with excitation at 514 nm and emission at 530–570 nm, while RFP signals were detected with excitation at 543 nm and emission at 580–620 nm.

Promoter Activity Assays in Rice Protoplasts

For promoter assays, the promoter regions of *OsWRKY9* were cloned into the DJ467 vector and co-introduced into protoplasts with the effector plasmid pEarleyGate104/*OsWRKY65* and the reporter plasmids 35S:Renila luciferase (RLUC). Luciferase activities were measured using the Dual-Luciferase Reporter Assay System (Promega, USA), and promoter activity was determined based on the ratio of firefly luciferase (FLUC) to RLUC activity.

Bakanae and Bacterial Blight Disease Assays

Bakanae and bacterial blight disease assays were conducted as previously described (Son et al. 2024a, 2024b). Bakanae disease indices ranged from 0 to 3, where 0 represented no symptoms, 1 indicated blight symptoms on leaf tips, 2 indicated wilted shoots and pale green leaf color, and 3 indicated complete leaf drying. Severity of bakanae disease (disease index 2 and 3) was used to assess rice susceptibility to *F. fujikuroi* in this study. Bacterial blight disease incidence (%) was calculated as follows: (average lesion length/leaf length) × 100.

Results

OsWRKY9 Localizes to the Nucleus and Responds to *F. fujikuroi* and *Xoo*

The cDNA of the WRKY Group IIb *OsWRKY9* from the rice cultivar Ilmi comprises 1812 bp, encoding 603 amino acids with a WRKY domain containing the WRKYGQK motif and a C_2H_2 zinc finger motif (Fig. 1A). Subcellular localization was determined by transfecting pEarleygate104/*OsWRKY9* constructs expressing YFP-OsWRKY9 into rice protoplasts and observing fluorescence signals. The results demonstrated specific nuclear targeting of OsWRKY9 (Fig. 1B).

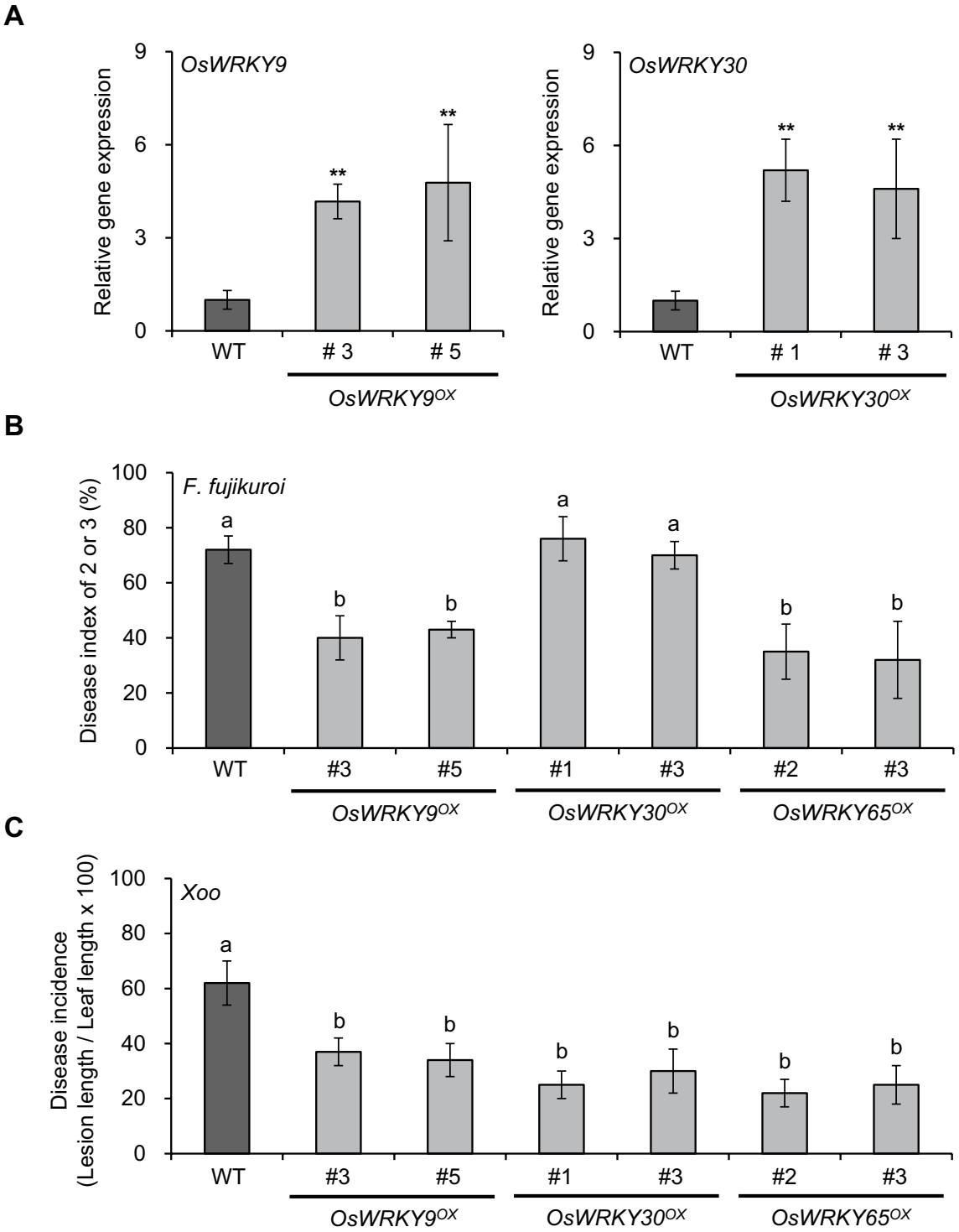
To determine whether OsWRKY9 is involved in innate immunity against *F. fujikuroi* and *Xoo*, we assessed the abundance of *OsWRKY9* transcripts following inoculation with each pathogen. Four-day-old wild-type seedlings were inoculated with *F. fujikuroi* CF283, and total RNA was extracted from shoots. The results revealed a significant upregulation of *OsWRKY9* expression in response to *F. fujikuroi* infection (Fig. 1C). Additionally, 6-week-old wild-type plants were inoculated with the compatible *Xoo* strain KACC10859 using the clip method, and leaves were harvested. RT-qPCR analysis showed an increase in *OsWRKY9* expression levels in response to *Xoo* (Fig. 1D). These findings suggest that OsWRKY9 likely plays a role in rice-pathogen interactions.

OsWRKY9 is Indirectly Induced by OsWRKY65 through Transcriptional Regulatory Mechanisms

Previous transcriptomic analysis indicated that *OsWRKY9* transcriptional levels are elevated in $OsWRKY65^{OX}$, similar to the upregulation observed for *OsWRKY13* and *OsWRKY30* (Son et al. 2024b). RT-qPCR analysis confirmed increased expression of *OsWRKY9*, *OsWRKY13*, and *OsWRKY30* in $OsWRKY65^{OX}$ lines (Fig. 2A). Since OsWRKY13 and OsWRKY30 are known to play crucial roles in rice immunity against *Xoo* (Qiu et al. 2008; Jalmi and Sinha 2016; Wang et al. 2022), these findings suggest that OsWRKY65-induced expression of *OsWRKY13* and *OsWRKY30* contributes to bacterial blight resistance. Furthermore, we hypothesized that OsWRKY9 might also function as a regulator in this transcriptional regulatory cascade. To explore the correlation between OsWRKY9 and OsWRKY65, a promoter activity assay was conducted. Analysis of the 1-kb region upstream of the *OsWRKY9* translation initiation site revealed the presence of four W-box motifs within the first 500 bp (Fig. 2B). Interestingly, OsWRKY65 induced promoter activity in the 1-kb upstream region of *OsWRKY9*, whereas it did not affect the 500 bp upstream region (Fig. 2B). Moreover, the promoter activity assay showed that the promoter region from -1000 to -500 of *OsWRKY9*, which lacks the W-box motif, is activated by OsWRKY65 (Fig. 2B). These results indicate that OsWRKY65 indirectly regulates the promoter activity of *OsWRKY9*.

OsWRKY9 Confers Resistance to Both *F. fujikuroi* and *Xoo* Pathogens in Rice

To investigate the function of OsWRKY9 in rice immunity, transgenic rice cv. Ilmi lines overexpressing *OsWRKY9* and *OsWRKY30* were generated. Among the generated lines, two lines with strong expression were selected for $OsWRKY9^{OX}$ and $OsWRKY30^{OX}$, confirmed by RT-qPCR in 2-week-old seedlings (Fig. 3A). To assess whether OsWRKY9 is involved in OsWRKY65-mediated immune responses, $OsWRKY9^{OX}$, $OsWRKY30^{OX}$, and $OsWRKY65^{OX}$ lines were infected with *F. fujikuroi* race CF283. Disease severity index scores ranged from 0 to 3, with a reduction in the sum of severe disease indices 2 and 3 observed in $OsWRKY9^{OX}$ and $OsWRKY65^{OX}$ compared to wild-type plants, whereas



OsWRKY30^{OX} showed symptoms similar to those of wild-type plants (Fig. 3B).

Furthermore, bacterial blight resistance assays revealed that *OsWRKY9*^{OX}, *OsWRKY30*^{OX}, and *OsWRKY65*^{OX} lines exhibited significantly increased resistance to the compatible *Xoo* strain KACC10859 compared to wild-type plants (Fig. 3C). Overall, these findings collectively demonstrate that OsWRKY9 confers BSDR to *F. fujikuroi* and *Xoo*, akin to the resistance conferred by OsWRKY65.

◂**Fig. 3** Disease phenotypes of *OsWRKY9*- and *OsWRKY30*-overexpressing plants. **A** Generation and verification of $OsWRKY9^{OX}$ and $OsWRKY30^{OX}$. Relative expression levels of *OsWRKY9* and *OsWRKY30* were quantified using RT-qPCR, with *OsActin* as the reference gene. Data are presented as means ± SD, with asterisks indicating values significantly different from the controls (** $P < 0.01$). **B** Bakanae disease indices of the $OsWRKY^{OX}$ lines. Four-day-old $OsWRKY9^{OX}$, $OsWRKY30^{OX}$, $OsWRKY65^{OX}$, and wild-type plants were inoculated with *F. fujikuroi* CF283 ($n = 10$). The disease index was evaluated at 6 days post-inoculation. Disease indices were categorized as follows: 0, no symptoms; 1, blight symptoms on the leaf tip; 2, wilted shoots and partial leaf drying; 3, entire leaf drying. The severity of bakanae disease (disease indices 2 and 3) was used to determine rice susceptibility to *F. fujikuroi*. Data are presented as means ± SD, with different letters indicating significant differences according to ANOVA ($P < 0.05$). **C** Bacterial blight disease resistance of the $OsWRKY^{OX}$ lines. Six-week-old $OsWRKY9^{OX}$, $OsWRKY30^{OX}$, $OsWRKY65^{OX}$, and wild-type plants were inoculated with *Xoo* strain KACC10859 ($n = 10$). The incidence of bacterial blight disease (%) was determined at 14 days post-inoculation, calculated as (average lesion length/leaf length) × 100. Data are presented as means ± SD, with different letters indicating significant differences according to ANOVA ($P < 0.05$)

OsWRKY9 Modulates the Expression of Genes Involved in the OsWRKY65 Transcriptional Regulatory Cascade

Transcriptome analysis and RT-qPCR revealed that the expression levels of key susceptibility-related genes to *F. fujikuroi* (e.g., *GA 3-OXIDASE 2* [$OsGA3_{OX}2$], $OsGA20_{OX}2$, and *JASMONATE ZIM-DOMAIN 10* [*OsJAZ10*]) and *Xoo* (e.g., *OsSWEET13*) are downregulated in $OsWRKY65^{OX}$, whereas the expressions of defense-related genes, including *12-OXOPHYTODIENOIC ACID REDUCTASE 1* (*OsOPR1*) and *PATHOGENESIS-RELATED GENE 10A* (*OsPR10a*) are upregulated (Son et al. 2024b). Therefore, to further elucidate the function of OsWRKY9 and its relationship with OsWRKY65, we examined the expression levels of these genes in $OsWRKY9^{OX}$ plants. Transcription levels of GA biosynthesis-related genes, such as $OsGA3_{OX}2$ and $OsGA20_{OX}2$, along with the JA signaling repressor *OsJAZ10*, were downregulated in 2-week-old $OsWRKY9^{OX}$ plants compared to wild-type plants, whereas transcripts of the JA biosynthesis-related gene *OsOPR1* were up-regulated (Fig. 4A). Additionally, overexpression of *OsWRKY9* decreased the expression of *OsSWEET13*, while increasing transcripts of *OsPR10a* (Fig. 4A). These findings suggest that OsWRKY9 is involved in OsWRKY65-mediated BSDR to both *F. fujikuroi* and *Xoo*. Moreover, to determine whether OsWRKY9 regulates the expression of *OsWRKY65*, we analyzed *OsWRKY65* transcripts in 2-week-old $OsWRKY9^{OX}$ and wild-type plants. The result showed that the expression level of *OsWRKY65* is not regulated by OsWRKY9 (Fig. 4B).

Discussion

Numerous studies have demonstrated that various WRKY transcription factors play critical roles in defense response-signaling pathways related to pathogen attack (Wani et al. 2021; Javed and Gao 2023). Although there are 125 OsWRKYs in the rice genome, many remain unexplored, presenting vast opportunities for future research. Previous RNA-sequencing result revealed that *OsWRKY9* expression is increased in $OsWRKY65^{OX}$, which exhibits enhanced resistance to *F. fujikuroi* and *Xoo* (Son et al. 2024b). Therefore, here, we focused on the role of OsWRKY9 in association with these pathogens.

OsWRKY9 belongs to WRKY group II and is specifically localized in the nucleus (Fig. 1A, B). In rice, eight OsWRKYs, including OsWRKY1, OsWRKY5, OsWRKY9, OsWRKY27, OsWRKY32, OsWRKY43, OsWRKY73, and OsWRKY97, belong to OsWRKYIIb (Yang et al. 2009). Among these, the expressions of *OsWRKY1*, *OsWRKY9*, and *OsWRKY43* are induced by both compatible and incompatible *Xoo* strains (Choi et al. 2017), suggesting their involvement in rice immunity. In this study, we showed that the transcription levels of *OsWRKY9* are significantly upregulated in response to not only *Xoo* but also *F. fujikuroi* (Fig. 1C, D). The initial interaction between rice and *Xoo* at the infection site determines the infection outcome, triggering rapid changes in gene expression even within minutes of *Xoo* inoculation (Kim et al. 2021). However, *F. fujikuroi* colonization progresses more slowly, with the earliest significant differential responses observed 7 days post-inoculation (Ji et al. 2016; Matic et al. 2016; Cheng et al. 2020). Indeed, the expression of *OsWRKY9* was induced within 6 h following *Xoo* inoculation (Fig. 1D), whereas it was induced after 8 days in response to *F. fujikuroi* (Fig. 1C). These results suggest that OsWRKY9 plays a crucial role in the early defense response to both *F. fujikuroi* and *Xoo*. Additionally, we determined that the expression of *OsWRKY9* is induced by OsWRKY65, and the 501–1,000 bp upstream promoter region excluding the W-box motif is necessary for its activation by OsWRKY65 (Fig. 2A, B). This suggests that OsWRKY65 indirectly activates the *OsWRKY9* promoter through a non-WRKY transcription factor. Identifying this unknown transcription factor is essential for understanding the detailed regulatory mechanism related to OsWRKY9 and OsWRKY65.

To investigate whether OsWRKY9 is involved in the OsWRKY65 transcriptional cascade and plant immunity, we generated transgenic plants expressing *OsWRKY9* and *OsWRKY30*, respectively, which are higher in $OsWRKY65^{OX}$, and analyzed their resistance to *F. fujikuroi* and *Xoo*. $OsWRKY9^{OX}$ and $OsWRKY65^{OX}$ exhibited enhanced disease resistance to both pathogens, while $OsWRKY30^{OX}$

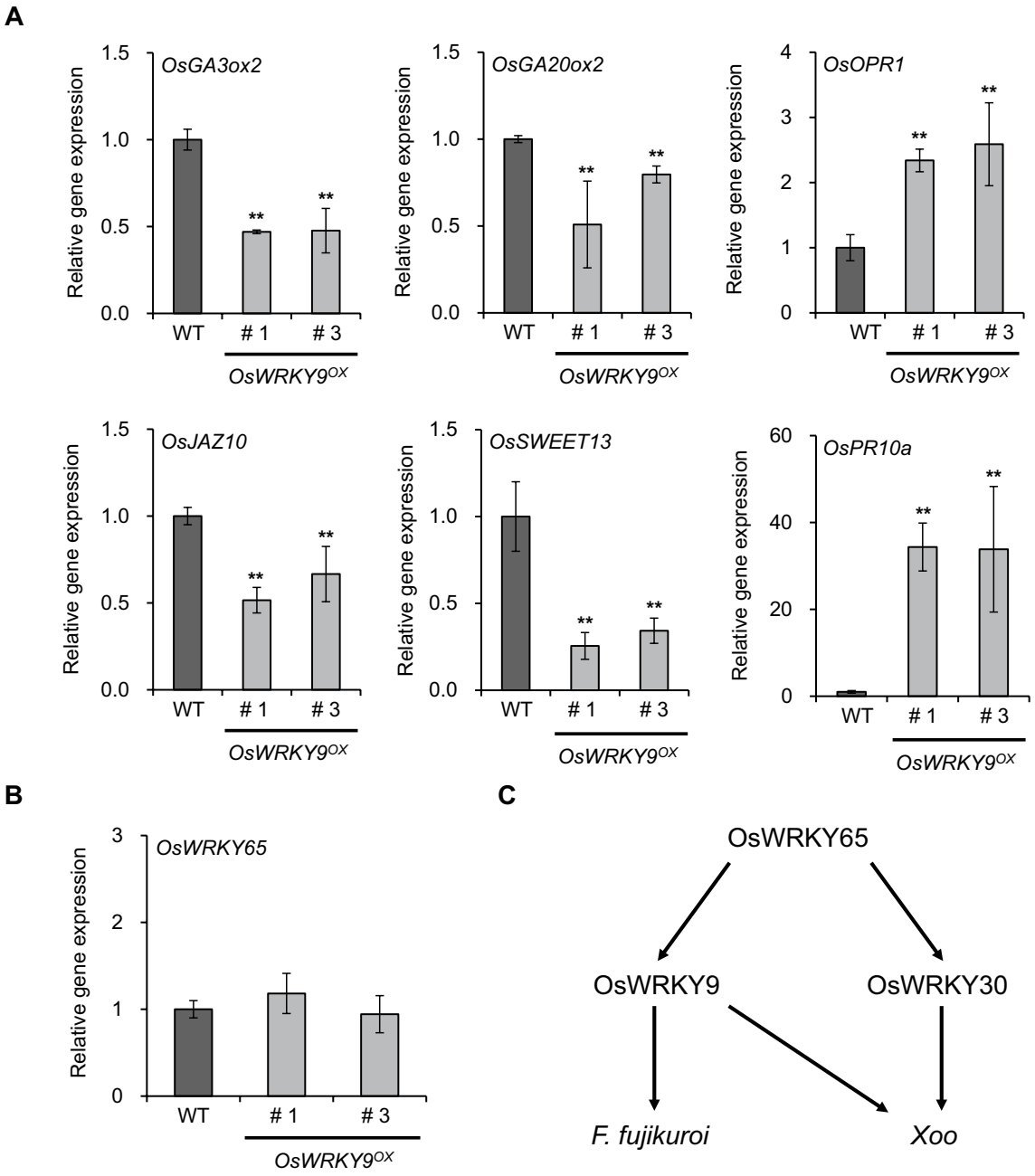


Fig. 4 Expression levels of genes regulated by OsWRKY65 in *OsWRKY9*-overexpressing plants. **A**, **B** Expression levels of susceptibility and defense-related genes in *OsWRKY9*-overexpressing plants. The relative transcription levels of $OsGA3_{OX}2$, $OsGA20_{OX}2$, *OsOPR1*, *OsJAZ10*, *OsSWEET13*, and *OsPR10a* (A) and *OsWRKY65* (B) were quantified using RT-qPCR in 2-week-old $OsWRKY9^{OX}$ and wild-type plants, with *OsActin* as the reference gene. Data are presented as means ± SD, with asterisks indicating values significantly different from the controls (** $P < 0.01$). **C** Conceptional model of the OsWRKY transcriptional regulatory cascade. OsWRKY65-induced expression of *OsWRKY9* and *OsWRKY30* enhances resistance to *F. fujikuroi* and/or *Xoo*

showed enhanced resistance only to *Xoo* (Fig. 3B, C). Previous studies have demonstrated that OsWRKY30 plays a crucial role in BSDR to *M. oryzae*, *R. solani*, *Xanthomonas oryzae* pv. *oryzicola* (*Xoc*), and *Xoo* (Peng et al. 2012; Han et al. 2013; Jalmi and Sinha 2016; Wang et al. 2022). Notably, the CCCH zinc finger transcription factor LEAF AND TILLER ANGLE INCREASED CONTROLLER (OsLIC) directly represses the expression of *OsWRKY30*, resulting in reduced resistance to *Xoc* and *Xoo* (Wang et al. 2022). However, MITOGEN-ACTIVATED PROTEIN KINASE 6 (OsMAPK6) negatively regulates OsLIC, thereby inducing *OsWRKY30*. Additionally, OsWRKY30 is implicated

in the MAPK KINASE 3-OsMAPK6 signaling conferring resistance to *Xoo* (Jalmi and Sinha 2016). Therefore, further studies are needed to determine whether OsWRKY65 and/or OsWRKY9 are associated with disease resistance to other pathogens, such as *M. oryzae*, *R. solani*, and *Xoc*, as well as with the MAPK signaling pathways. The induction of *OsSWEET* genes, including *OsSWEET13*, by various TALEs is crucial for susceptibility to *Xoo* (Xu et al. 2019). The expression level of *OsSWEET13* is significantly repressed in $OsWRKY9^{OX}$ (Fig. 4A), suggesting that *Xoo* must counteract the OsWRKY9-mediated suppression of *OsSWEET13* to induce its expression. PthXo2 and PthXo2-like TALEs are major TALEs known to induce the expression of *OsSWEET13* (Liu et al. 2024). Therefore, it is possible that the competition between the OsWRKY65-OsWRKY9 module and PthXo2 and PthXo2-like TALEs at the *OsSWEET13* promoter may determine susceptibility to *Xoo*. This hypothesis also warrants further investigation in future study.

In this study, we demonstrated that OsWRKY9 contributes to BSDR to *F. fujikuroi* and *Xoo* and proposed a transcriptional regulatory cascade conferring rice immunity (Fig. 4C). Our findings provide information on novel signaling pathways controlling immune response, potentially helping to breed rice varieties with enhanced disease resistance.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s12374-024-09439-3.

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Author Contributions SS conceptualized the study. GS, SRP, and YJ performed the experiments and data analysis. GS, JL, and D-JH generated the constructs and the transgenic plants. GS and SS wrote the manuscript. SRP, N-CP, and SS supervised the study.

Data Availability The data and materials shown in this article are available from the corresponding author SRP upon a reasonable request.

Code Availability N/A.

Declarations

Conflict of Interest The authors declare no conflicts of interests.

Ethics Approval N/A.

Consent to Participate N/A.

Consent for Publication N/A.

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