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Supplementary Information for

Distinct modes of manipulation of rice auxin response factor OsARF17 by different plant RNA viruses for infection

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Materials and methods

Plant materials and treatments. Rice seeds of Wuyujing No.3 were used for this study. The SP8 transgenic plants expressing the SRBSDV P8 protein were in the rice cultivar Nipponbare (*O. sativa* L. subsp. *japonica*, cv. NIP) background, while plants overexpressing OsARF17 and the OsARF17 mutant plants used Zhonghua 11 (ZH11) seedlings as the background. RBSDV-infected plants were obtained from fields in Shandong Province, China. The SRBSDV- infected plants were gifts from Prof. Guohui Zhou and RSV-infected plants were obtained from Prof. Yijun Zhou. To check the auxin responsiveness of SP8 transgenic plants, the shoots of vigorous seedlings were immersed in 0.1 μM 1-Naphthaleneacetic acid (NAA, Sigma-Aldrich) or 0.1 μM 2, 4-Dichlorophenoxyacetic acid (2, 4-D, Sigma- Aldrich) for 10-days in darkness. Images were taken after 10 days treatment, and the primary root lengths were measured. The experiments were repeated three times with similar results. All other plants were grown in the greenhouse at 28-30 °C with a 14h-light/10h-light/dark cycle.

Total RNA extraction and qRT-PCR assays. Total RNA was extracted from rice leaves using the TRIzol reagent (Invitrogen, Carlsbad, CA, USA) according to the manufacturer's protocol. First-strand cDNA was synthesized using the fast quant RT kit (Tiangen, Beijing, China). The qRT-PCR analysis was performed using the ChamQTM SYBR qPCR Master Mix (Low ROX Premixed) by the ABI7900HT Sequence Detection System (Applied Biosystems, Carlsbad, CA, USA). The housekeeping gene OsUBQ5 (AK061988) was used to normalize the statistic (1). The mRNA expression levels were calculated by the $2^{-\Delta\Delta C(t)}$ method (2). The experiments were repeated at least three times with similar results. Each biological sample consisted of 10 to 12 pooled plants. The qRT-qPCR primers used in this study are listed in Table S1.

Western blot analysis. SRBSDV-, RBSDV- and RSV- infected rice leaves at 30 dpi were extracted with SDS lysis buffer for western blot analysis as previously described (3). Anti-P10 polyclonal antibody (provided by Prof. Jianxiang Wu) was used to test for SRBSDV/RBSDV infection and an anti-RSV-CP polyclonal antibody produced in our laboratory was used for diagnosis of RSV.

Insect vectors and virus inoculation assay. SRBSDV is mainly transmitted by the white-backed planthopper (*Sogatella furcifera,* WBPH), RBSDV and RSV are transmitted by the small brown planthopper (*Laodelphax striatellus,* SBPH), while RSMV is transmitted by leafhoppers (4). The transmission experiments were done as described previously with some minor modifications (5, 6). Briefly, SBPH carrying RBSDV or RSV were transferred to rice seedlings at the 3 to 4-leaf stage (about three viruliferous insects per seedling) for 3 days. The insects were then completely removed. SRBSDV inoculation assays were similar but used 2-3-leaf rice seedlings infested with nymphs either carrying SRBSDV or virus-free (7). For RSMV inoculation assays, leafhoppers carrying RSMV were placed on seedlings at the 3 to 4-leaf stage for 3 days. The inoculated plants were grown in the greenhouse to obverse symptoms. In each experiment, there were negative controls using the same number of virus-free insects. Plants infected with SRBSDV/RBSDV were stunted and had darkened leaves, RSV-infected plants had necrotic stripes and stunting and the leaves of RSMV-infected plants had yellow stripes and mosaic or twisting symptoms. The infected-plants were tested at 30 dpi by RT-PCR and western blotting. The specific primers used to detect (SRBSDV, RBSDV, RSV and RSMV) are listed in Table S1. The number of infected plants was determined following RT-PCR to calculate the viral incidence. The experiments were repeated at least three times with similar results.

Vector construction and plant transformation. To generate transgenic

plants over-expressing OsARF17, the full-length ORF of OsARF17 was inserted into the pCV1300 vector, driven by the 35S promoter. The recombinant plasmid was introduced into *Agrobacterium tumefaciens* strain GV3101 and transformed into rice ZH11 background as described previously (5). The primers used are listed in Table S1.

Yeast Two-Hybrid (Y2H) and Three-Hybrid (Y3H) Assays. The rice cDNA library for Y2H assays were generated by cloning cDNA synthesized from the rice mRNAs to the prey vector pGADT7. The library was screened using each of the viral proteins of SRBSDV as the baits in vector pGBKT7. Yeast transformation and screening were done as described in the manufacturer's instructions with some modification (Clontech; Mountain View, CA, USA). Firstly, these bait vectors were introduced into the AH109 yeast strain for autoactivation verification. The full-length cDNA library was transformed into AH109 yeast strain, which contained the viral proteins of pGBKT7 bait plasmid. The sequences were analyzed by BLASTn (NCBI database). The bait and prey vectors were constructed by PCR using the listed primers (Table S1). Different combinations of plasmids were transformed into yeast strain AH109. The transformants were cultivated on an SD/-Leu/-Trp (SD-L-T) medium and then transferred to SD/-Leu/-Trp-His-Ade (SD-L-T-H-Ade) selection medium for the interaction test. Yeast cells were photographed after 3 days of incubation at 30°C. All experiments were repeated three times with similar results. For Y3H assays, the full-length CDS sequences of OsIAA20 and $CTD^{OsARF17}$ (the CTD domain of OsARF17) were ligated into the pBridge vector (Clontech), to express DNA binding domain fusion proteins. *SP8* was cloned into the same pBridge vector which contained the *MET25* promoter to drive transcription. Y3H assays were based on the manufacturer's protocol (Takara, Japan) and the different combinations were co-transformed into the yeast strain AH109. The transformants were cultivated on an SD-L-T medium and then transferred to liquid SD/-Trp/-Leu media overnight until the liquid

became turbid. The yeast was diluted (1:100) in liquid SD-L-T-H-Met media and then different concentrations of Met were added. Beta-Galactosidase enzyme activity was then determined according to the protocol (Takara, Japan). Dilutions of suspended yeast (Five microliters) were plated on SD/-T/-L/-H-Ade and SD-L-T-H-Met solid media. Yeast cells were observed after 3 days incubation at 30°C. The experiments were repeated at least three times with similar results.

Co-IP. *N. benthamiana* leaves (five-week-old) were transformed by agro-infiltration with the following combinations: OsARF17-MYC/SP8-FLAG, GFP-MYC/SP8-FLAG (as a negative control), CTD-MYC/SP8-FLAG, DBD-MYC/SP8-FLAG, OsARF17-MYC/P2-FLAG, OsARF17-MYC/GFP-FLAG (as a negative control), DBD-MYC/P2-FLAG, CTD-MYC/P2-FLAG and OsARF17-MYC/M-FLAG. The vectors were constructed by ligation-independent cloning using the primers listed in Table S1. 72 hours after agro-infiltration, the leaves were ground in liquid nitrogen and extracted using IP lysis buffer (Thermo Scientific, Cat. no. 87788) with 1 g samples in a 1ml reaction solution with the addition of 10 mM DTT, a protease inhibitor cocktail (Roche, Switzerland). After maintaining them at 4°C for 10 min, the mixtures were centrifuged at 1,000g for 20 min, and the supernatant was then incubated with 20 μl Pierce^{TM} anti-c-Myc magnetic beads (Thermo Scientific, USA) or anti-FLAGM2 beads (Sigma-Aldrich, USA) for 2-3 h at 4°C. The beads had been pre-washed three times with 1×PBS. The immunoprecipitates were then washed four times with 1×PBS and resuspended in 80 μL 2×SDS-PAGE sample buffer (500 mMTris-HCl, pH=6.8, 50% glycerin, 10% SDS, 1% bromophenol blue and 2% β-mercaptoethanol). Subsequently, the protein samples were boiled at 95 °C for 10 min and separated on a 10% SDS-PAGE gel for western blotting analysis.

In vitro **pull-down assays.** The full-length SP8 protein was constructed into

pCold-TF-His vector and expressed as a recombinant His-SP8 fusion protein. The recombinant His-SP8 and His-TF (Trigger Factor) proteins were purified from *E. coli*. The OsARF17 CTD domain was constructed into pGEX6P1 and pET28a-HMT vector to form glutathione-S-transferase (GST)-tagged GST-CTD and His-CTD, respectively. The recombinant proteins were expressed in *E. coli*. Primers used for the constructs are listed in Table S1. Protein expression was induced by 0.8 mM IPTG. The supernatant containing target protein after sonication was incubated with 20 µL glutathione (GST) sepharose beads (Thermo Scientific) or Beaver Beads™ His-tag Protein Purification beads for 1 h at 4 °C, and then washed to remove the non-specifically bound proteins. For interaction pull-down assays, the purified His-SP8 and OsARF17-MYC or GFP-MYC proteins (transiently expressed in *N. benthamiana* cells) were incubated with Myc magnetic beads at 4°C for 2h. For competitive assays, 10 µg GST-CTD was mixed with 0, 2, 10 or 20 µg His-SP8 or 20 µg of His-TF was mixed with10 µg His-CTD and then incubated and immobilized onto glutathione GST sepharose beads at 4°C for 2 h. The beads were centrifuged at 5000g for 1 min and washed four times with 1× PBS. The proteins bound onto beads were separated by SDS-PAGE.

BiFC assays. For BiFC assays, the full-length SP8 and OsARF17 were cloned into either the N-terminus of YFP or the C terminus of YFP to generate nYFP-SP8 and OsARF17-cYFP vectors. Primers used are listed in Table S1. The constructs were then transiently expressed in *N. benthamiana* cells using Agrobacterium strain GV3101. After incubation for 48-72 h, the YFP fluorescence was observed using a confocal laser microscopy (Leica TCS SP10). YFP was excited with 488 nm and its emissions were detected at 500 to 542 nm. Three biological repeats were conducted with similar results.

Electrophoretic mobility shift assays. For protein purification, the full-length CDS of P2 was cloned into the pET28a vector to express MBP-tagged fusion

protein MBP-P2. Because the solubility of the DBD domain protein of OsARF17 was very low, the shortest functional domain was inserted into the pCold-TF vector to express His-tagged fusion protein His-DBD. The pET28a-P2 and pCold-DBD vectors were transferred into *E. coli* strain DE3, with the empty pET28a and pCold-TF vectors used as the corresponding negative controls. Protein expression was induced by 0.8 mM IPTG. EMSAs were performed using the Light Shift Chemiluminescent EMSA kit following the manufacturer's instructions (Thermo Scientific, Cat. no. 20148). The AuxREs probes (50 nt) in the promoter of OsGH3.5 were labeled using Digoxigenin (DIG). The biotin-labeled probes were used as the hot probes and the unlabeled probes as competition probes for assays. For interference assays, equal amounts (10 μg) His-DBD were mixed with 0, 1 or 10 μg MBP-P2, or 10 μg of MBP was used alone. The primers used are listed in Table S1.

FRET-FLIM Measurements. The full-length OsARF17 and SP8 were inserted respectively into PCV-Ven and PCV-Tur to express them fused to Yellow Fluorescent Protein (YFP) as mVenus Ven-OsARF17 or to Cyan Fluorescent Protein (CFP) as mTurquoise mTur-SP8. They were expressed in *N. benthamiana* epidermal cells and the fluorescence lifetime of CFP- (donor) was experimentally measured in the absence and presence of the YFP-(acceptor) using a confocal laser scanning microscope. FRET-FLIM measurements were conducted as previously described (8). Fluorescence lifetime of the donor was measured in the presence and absence of the acceptor. FRET efficiency (E) was calculated by comparing the lifetime of the donor in the presence (τ_{DA}) or abscence (τ_D) of the acceptor: $E=1-(\tau_{DA})/(\tau_D)$ (8). The mean lifetime data are listed in Table S2. Values shown are the means ± SD of 3 biological replicates. Significant differences were identified using Fisher's least significant difference tests. **at the top of columns indicates significant difference at $p \leq 0.01$.

Dual Luciferase Assays. For the transcriptional activity assays, the full-lengths of OsARF18, SP8, or P2 were cloned into the PCV-BD vectors as effectors and the empty PCV-BD was used as the negative control. The promoter of 5**gal* was used to drive the firefly luciferase gene (LUC) as a reporter and the Renillia luciferase (*REN*) reporter gene controlled by Cauliflower mosaic virus promoter (35S) was the reference. The BD domain can bind to the 5*gal promoter. GV3101 strains harboring the combinations of effectors and reporters were transformed into *N. benthamiana* leaves for 72 h. For interference in dual-LUC assays, the OsARF17-MYC and P2-FLAG were used as effectors, and the promoter region of *OsGH3.5* (500bp) was ligated into the pGREENII0800-LUC vector as reporter. The *Renilla* LUC (REN) gene of pGREENII0800-LUC was used as a control. Agrobacterium infiltration of tobacco was performed as above. The dual-LUC assays were performed using the Luciferase Reporter Assay System (Promega, Madison, WI) following the manufacturer's instructions. The relative luciferase activity was analyzed used LUC/REN ratios. For each assay, three biological repeats were conducted with similar results. The primers used for these constructs are listed in Table S1.

Statistical Analysis. Differences were analyzed using a one-way or two-way ANOVA with Fisher's least significant difference tests. A p-value ≤ 0.05 was considered statistically significant. All analyses were performed using ORIGIN 8 software.

Accession numbers: Sequence data from this study can be found in the rice genome annotation project database under the following accession numbers: *OsARF1,* Os01g13520; *OsARF2,* Os01g48060; *OsARF3,* Os01g54990; *OsARF4,* Os02g04810; *OsARF5,* Os02g04810; *OsARF6,* Os02g06910; *OsARF7,* Os02g35140; *OsARF8,* Os02g41800; *OsARF9,* Os04g36054; *OsARF10*, Os04g43910; *OsARF11*, Os04g56850; *OsARF12*, Os04g57610; *OsARF13*, Os04g59430; *OsARF14*, Os05g43920; *OsARF15*, Os05g48870;

OsARF16, Os06g09660; *OsARF17,* Os06g46410; *OsARF18,* Os06g47150; *OsARF19,* Os06g48950; *OsARF20,* Os07g08520; *OsARF21,* Os08g40900; *OsARF22*, Os10g33940; *OsARF23*, Os11g32110; *OsARF24*, Os12g29520; *OsARF25,* Os12g41950; *OsIAA20,* Os06g07040;

Figs. S1-S15

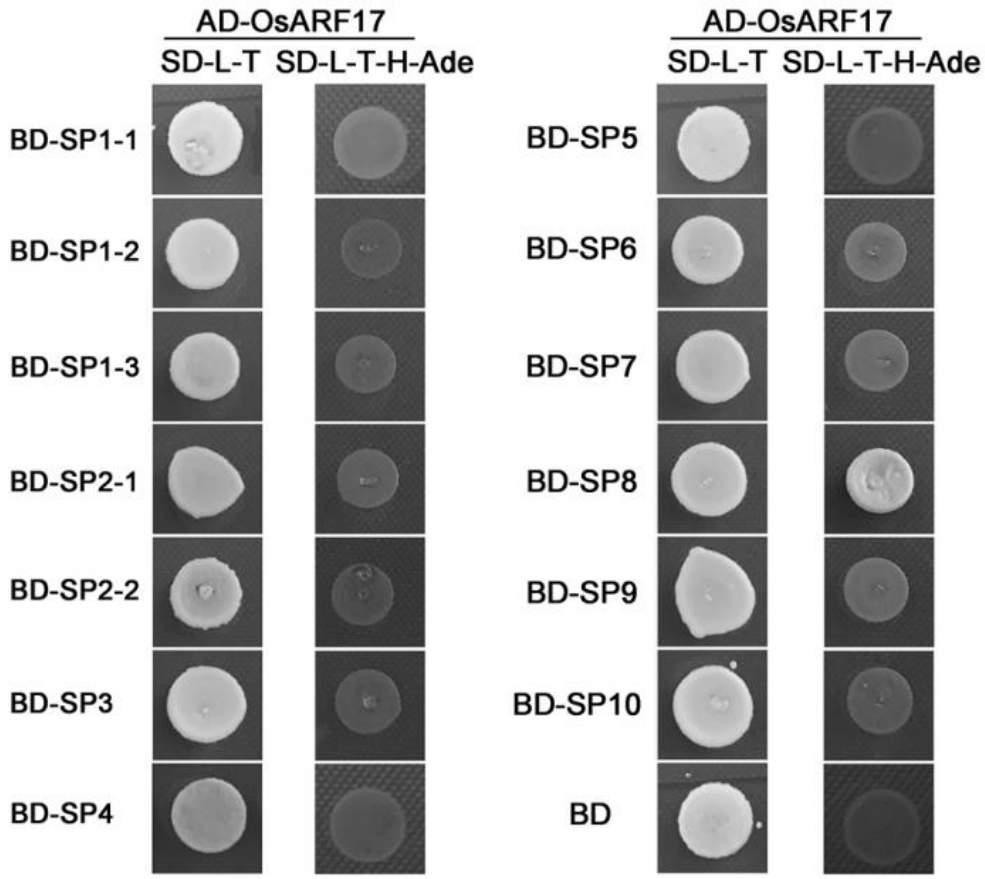

Fig. S1. Yeast two-hybrid (Y2H) assays showing that OsARF17 only interacted with SP8 and not with other SRBSDV proteins. The different combinations of constructs transformed into yeast cells were grown on selective media SD-L-T and interactions were tested with SD-L-T-H-Ade. Pictures were taken after 3 days of incubation at 30°C.

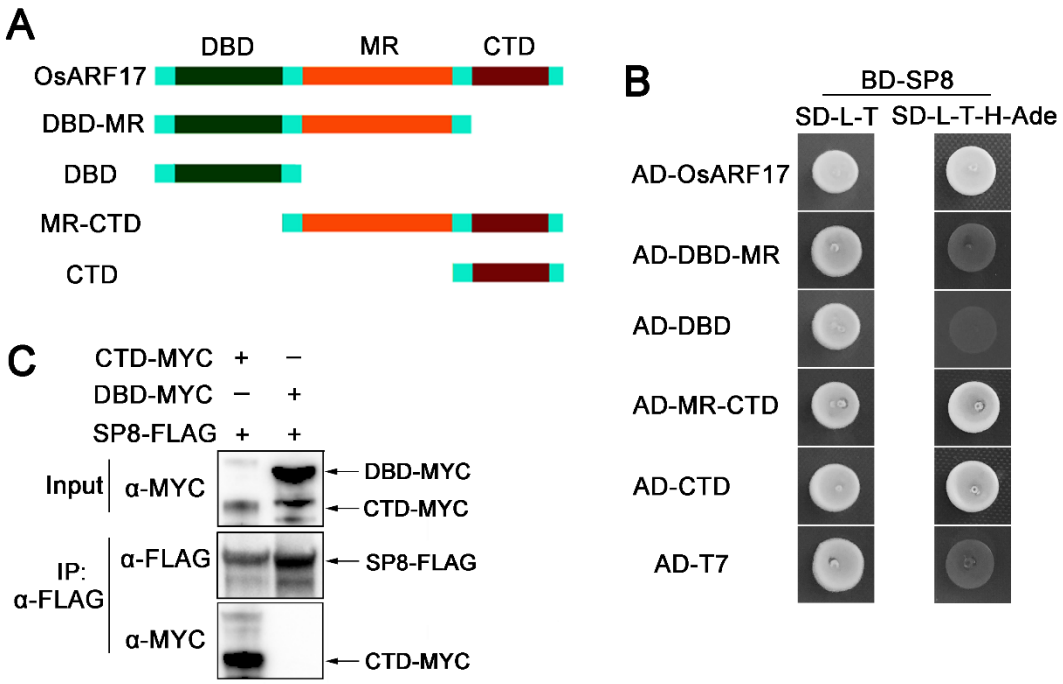

Fig. S2. SP8 protein interacts with the CTD domain of OsARF17. (A) Schematic diagram of the full-length and a series of OsARF17 truncated mutants constructed. DBD: DNA-binding domain; MR: middle region; CTD: carboxy-terminal dimerization domain. Dark green rectangles represented DBD domain. Orange rectangles represented MR domain. Dark red rectangles represented CTD domain. (*B*) The CTD domain of OsARF17 was required for binding to SP8 by Y2H assays. The different combinations of constructs transformed into yeast cells were grown on selective media SD/-Trp/-Leu (SD-L-T) and interactions were tested with SD/-Trp/-Leu/-His/-Ade (SD-L-T-H-Ade). Pictures were taken after 3 days of incubation at 30°C. (*C*) Co-IP assays confirm that the CTD domain was required for the SP8-OsARF17 interaction *in vivo*. Total proteins were extracted and immunoprecipitated using anti-FLAG magnetic beads. The coimmunoprecipitated proteins were detected by anti-MYC antibody. SP8 interacts with the CTD domain, but not with the DBD domain.

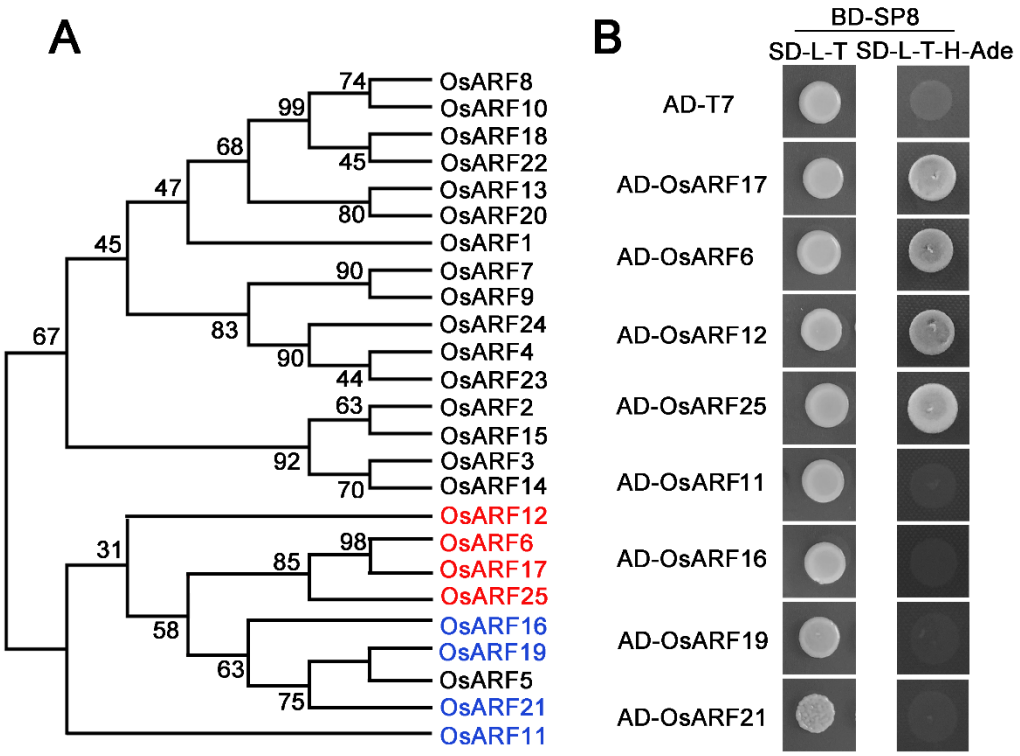

Fig. S3. (*A*) Phylogenetic analysis of the ARF proteins in the rice genome. The phylogenetic tree was drawn with Mega5.0 software. The red font and the blue font represent interaction and non-interaction with SP8 in *B*, respectively.

(*B*) Y2H assay to assess the interaction of SP8 with other OsARFs (OsARF6, OsARF12, OsARF25, OsARF11, OsARF16, OsARF19 and OsARF21). Only OsARF6, OsARF12 and OsARF25 interacted with SP8.

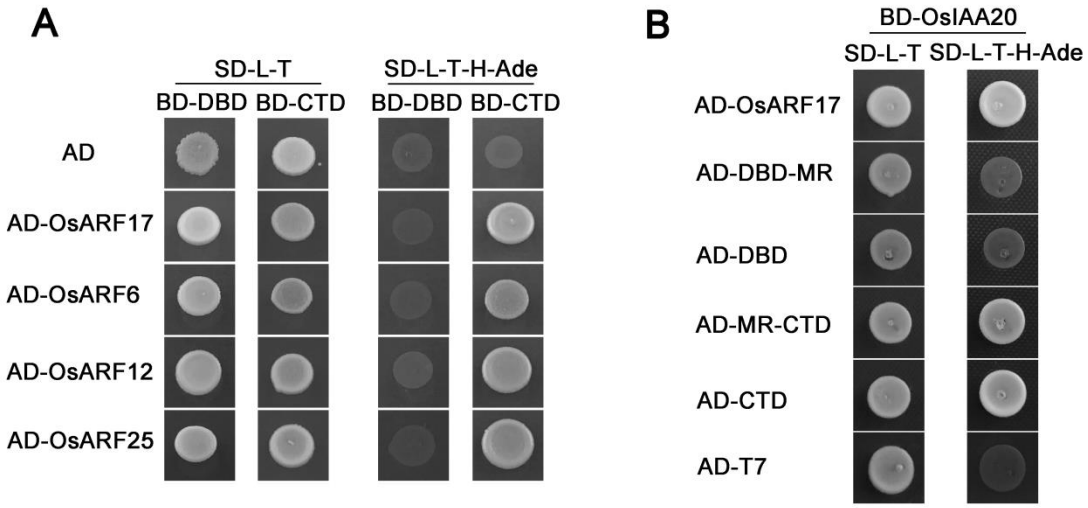

Fig.S4. (*A*) Y2H experiments confirmed that OsARF17 could form self-dimers (OsARF17) or heterodimers (OsARF6, OsARF12 and OsARF25) through its CTD domain, but not by the DBD domain. BD-DBD: DBD domain of OsARF17 cloned into pGBKT7 vector. BD-CTD: CTD domain of OsARF17 cloned into pGBKT7 vector. The different combinations of transformed yeast cells were grown on SD-L-T-H-Ade selective media to test the interaction. (*B*) Y2H assays showing that the CTD domain of OsARF17 was required for the interaction between OsARF17 and OsIAA20. The interactions were examined on SD-L-T-H-Ade selective media. Photos were taken after 3 days of incubation at 30°C.

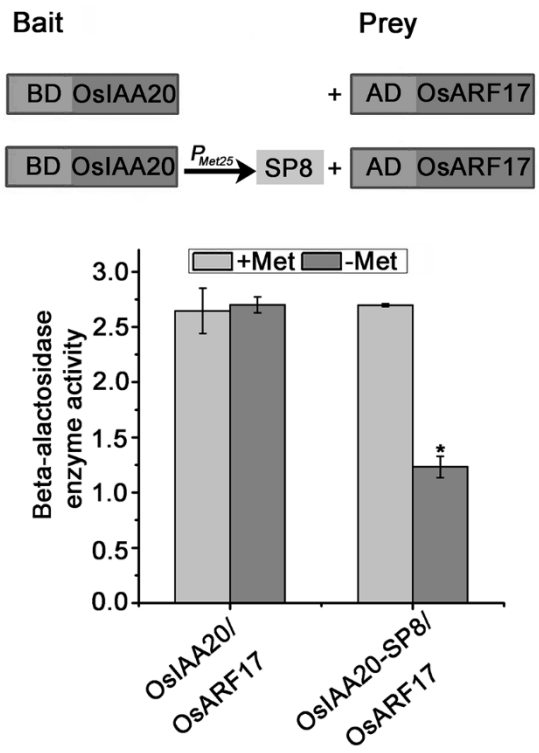

Fig.S5. The effect of SP8 on the OsARF17-OsIAA20 interaction. Upper panel**:** schematic diagrams of the bait and the prey vectors used in Y3H assays. OsIAA20 was ligated into the pBridge vector as the bait, and the expression of SP8 was driven by the methionine inducible *Met25* promoter. OsARF17 was cloned into pGADT7 vectors as prey. Lower panel: yeast cells harboring bait and prey vectors were grown on selective media SD-L-T for 3 days. The yeast cells were shaken with SD-L-T-H-Ade or SD/-Trp/-Leu/-His/-Met liquid selective media. β-Galactosidase enzyme activity was measured with or without SP8.

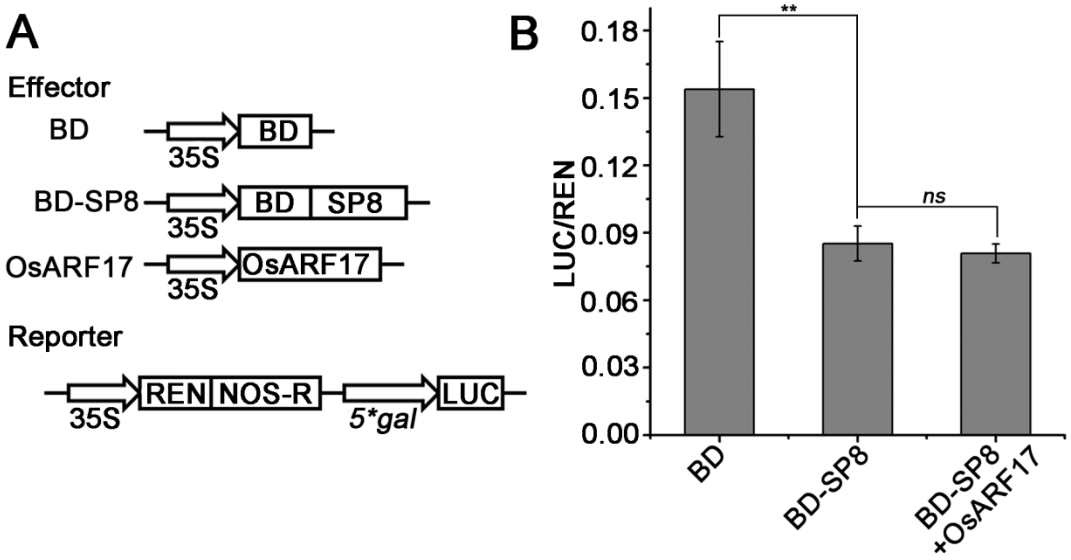

Fig.S6. OsARF17 did not affect the repressor activity of SP8. (*A*) Schematic diagrams of the effectors and reporters used in the dual-luciferase assays. The effectors: BD, SP8-BD and OsARF17 fused with MYC-tag. The reporters: 35S: REN-NOS-R/5**gal* pro:LUC plasmids. BD domain could bind the*5*gal* promoter. (*B*) The relative LUC activities were measured in *N. benthamiana* cells using the combinations shown in (*A*). The LUC activity significantly decreased in presence of GD-SP8 compared with the empty effector GD. The empty GD effector was used as a negative control. The LUC activity of GD-SP8 was not obviously different when GD-SP8 was expressed alone or with OsARF17.The LUC/REN ratio represents the relative LUC activity. Values shown are the means ± SD of 3 biological replicates. Significant differences were identified using Fisher's least significant difference tests. **at the top of columns indicates significant difference at $p \leq 0.01$. *ns*, no significant difference.

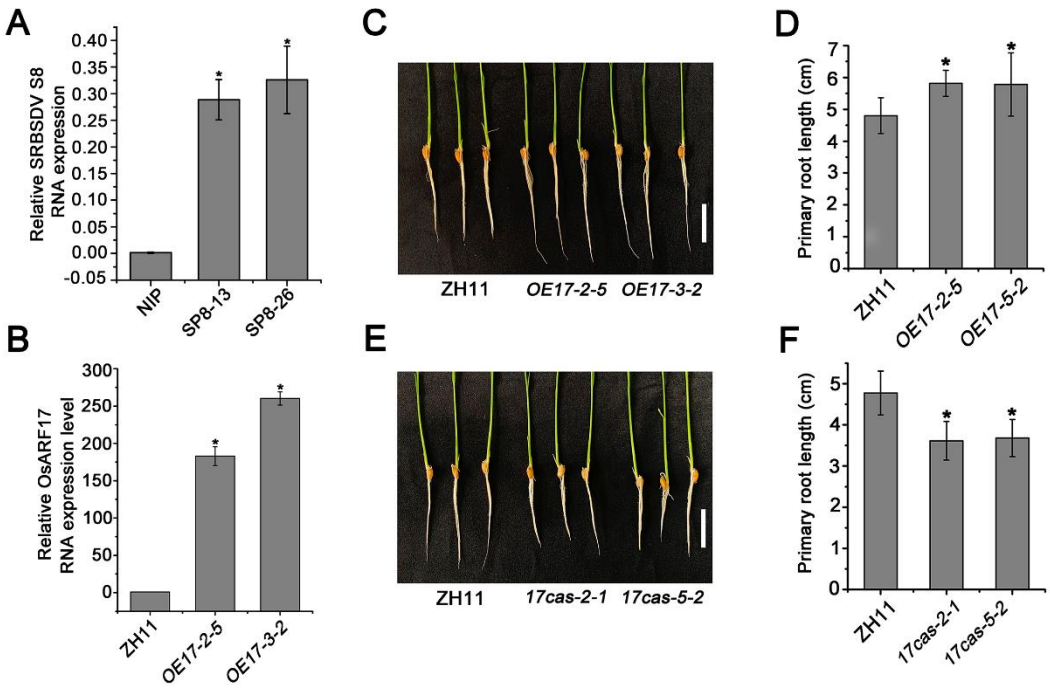

Fig.S7. Identification of *SP8* and *OE17* transgenic plants. (*A*) The relative expression levels of the SRBSDV S8 gene to housekeeping gene OsUBQ5 in control NIP, *SP8-13* and *SP8-26* plants. (*B*) The relative expression levels of OsARF17 gene in control ZH11, *OE17-2-5* and *OE17-3-2* plants. (*C*) Morphology of the primary roots of WT and transgenic seedlings overexpressing *OsARF17* after normal nutrition for 10 days. Scale bars: 2 cm. (*D*) Statistical analyses of *OE17-2-5* and *OE17-3-2* primary root length. (*E*) Morphology of WT and 17cas (*17cas-2-1* and *17cas-5-2*) mutant plants primary roots under normal nutritional solution for 10 days. Scale bars: 2 cm. (*F*) Statistical analyses of *17cas-2-1* and *17cas-5-2* primary root lengths. Values shown are the means ± SD of 3 biological replicates. Significant differences were identified using Fisher's least significant difference tests. *at the top of columns indicates significant difference at $p \leq 0.05$.

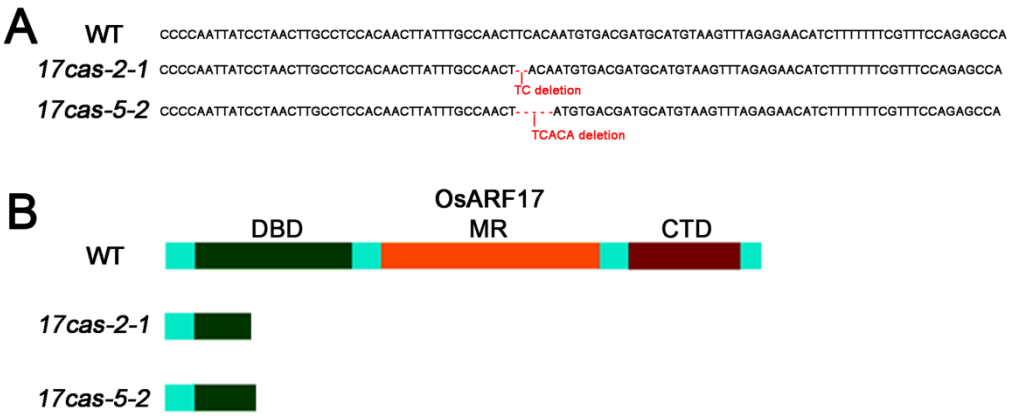

Fig.S8. The mutations of OsARF17 inserted into rice plants. (*A*) The mutants of OsARF17 with the nucleotide deletions in the T3 generation are showed by red letters. Dark green rectangles represent the DBD domain of OsARF17, orange rectangles are the MR domain and dark red rectangles are the CTD domain. (*B*) The two or five nucleotide deletions in *17cas-2-1* or *17cas-5-2* plants result in premature stop codons. Three complete domains of OsARF17 were truncated. DBD: DNA-binding domain; MR: middle region; CTD: carboxy-terminal dimerization domain.

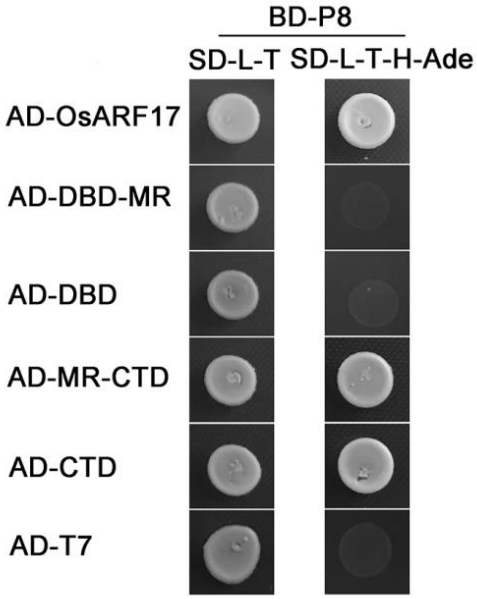

Fig.S9. Y2H assays showing that RBSDV P8 protein also interacted with OsARF17 and that the CTD domain of OsARF17 was required for the interaction. The interactions were examined with SD-L-T-H-Ade selective media. Photos were taken after 3 days of incubation at 30°C.

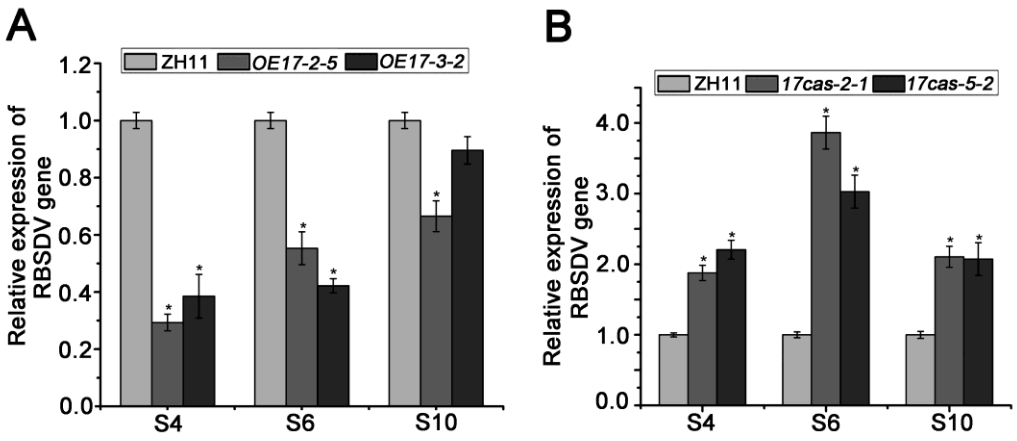

Fig.S10. Effect of OsARF17 on RBSDV infection. (*A*) qRT-PCR results showing the relative expression levels of RBSDV S4, S6 and S10 gene in virus infected *OE17-2-5* and *OE17-3-2* plants compared with the RBSDV-infected non-transformed ZH11 controls at 30 dpi. (*B*) The relative expression level of RBSDV S4, S6 and S10 genes in virus-infected *17cas-2-1* and *17cas-5-2* plants compared with RBSDV-infected ZH11 plants. Values shown are the means ± SD of 3 biological replicates. Significant differences were identified using Fisher's least significant difference tests. *at the top of columns indicates significant difference at $p \leq 0.05$.

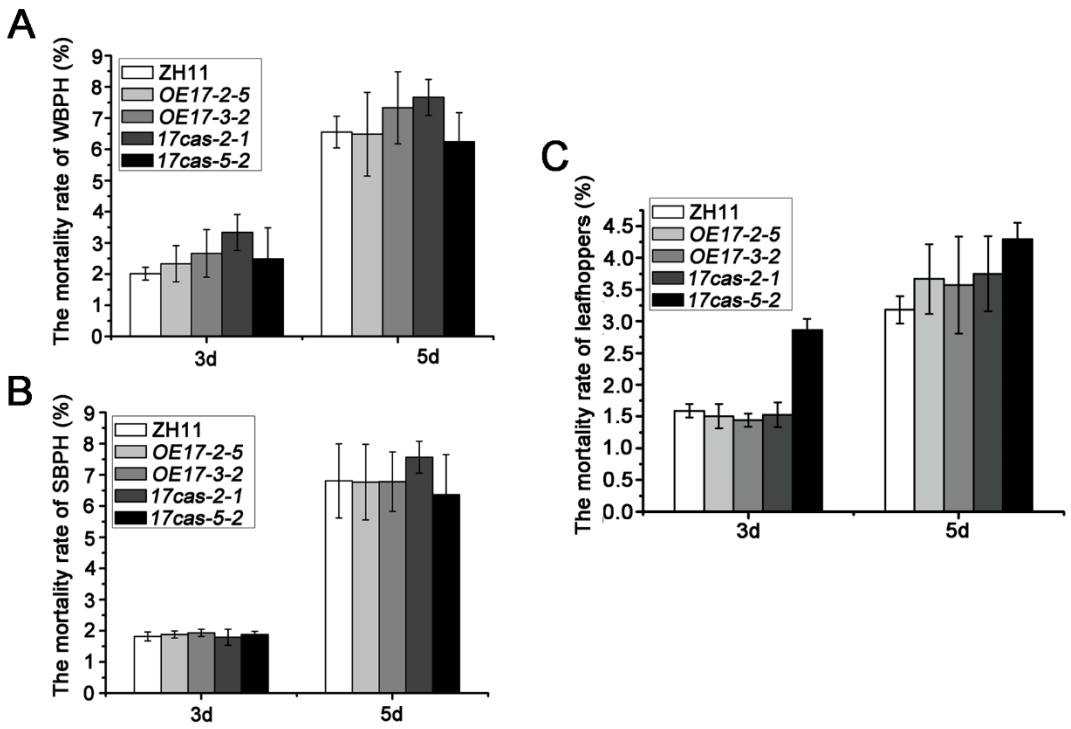

Fig.S11. The mortality rate of white-backed planthoppers (WBPH), small brown planthoppers (SBPH) or leafhoppers on control (ZH11) and transgenic plants. Ten-day-old seedlings of OsARF17-related transgenic lines were infested with virus-free WBPH (*A*), SBPH (*B*) or leafhoppers (*C*) at 6 insects per seedling for 5 days and insect survival was counted at 3 and 5 days. Each treatment had at least 60 insects. Values shown are the means ± SD of 3 biological replicates. Significant differences were identified using Fisher's least significant difference tests.

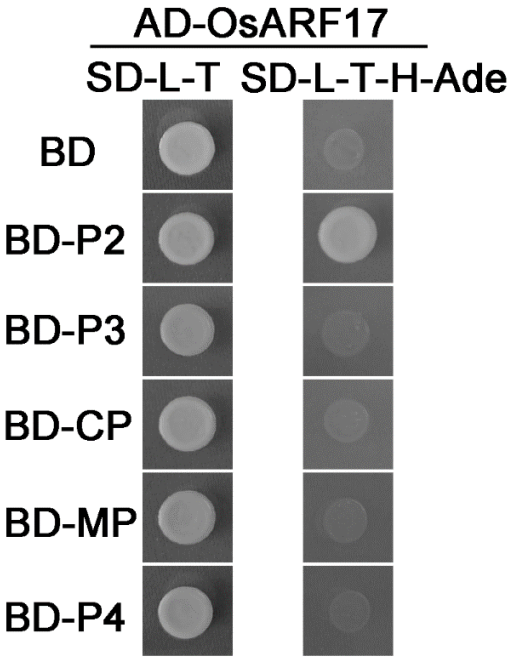

Fig.S12. Y2H assays showing that OsARF17 interacts with RSV P2, but not with other RSV proteins. The different combinations of constructs transformed into yeast cells were grown on selective media SD-L-T and interactions were tested with SD-L-T-H-Ade. Pictures were taken after 3 days of incubation at 30°C.

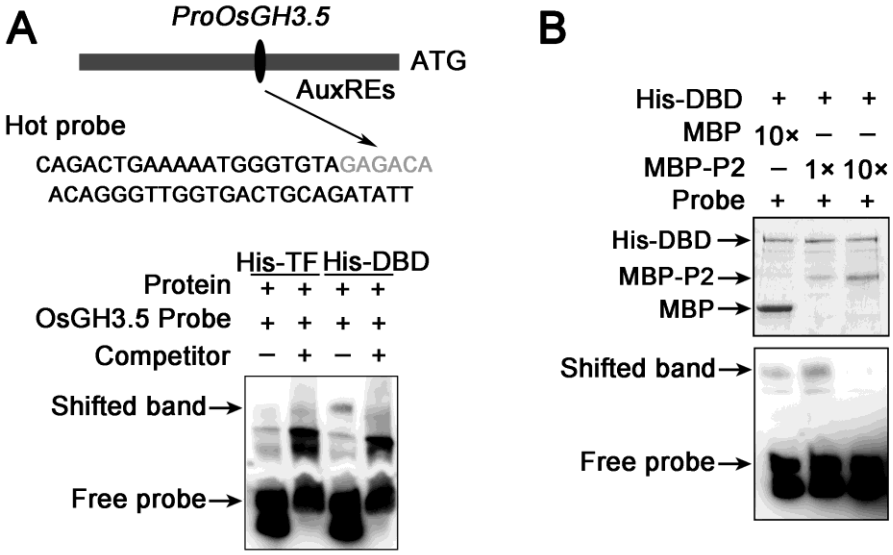

Fig.S13. P2 interferes with the ability of OsARF17 to bind to the promoter of *OsGH3.5 in vitro*. (*A*) Schematic diagrams of the OsGH3.5 promoters. The positions of AuxREs motif DNA binding sites are shown as a black ellipse. The DIG-labeled probes were used for EMSA and the grey letters indicated by the arrows are the binding sites. EMSA assays confirmed that OsARF17 directly bound the AuxREs of the*OsGH3.5* promoter. (*B*) EMSA assays showing that with increasing amounts of MBP-P2 fusion protein, the DNA binding activity of OsARF17 significantly decreased. Equal amounts of His-DBD proteins and probes were used; 10×indicate the excess amount of MBP-P2 relative to initial amount. MBP protein was the negative control.

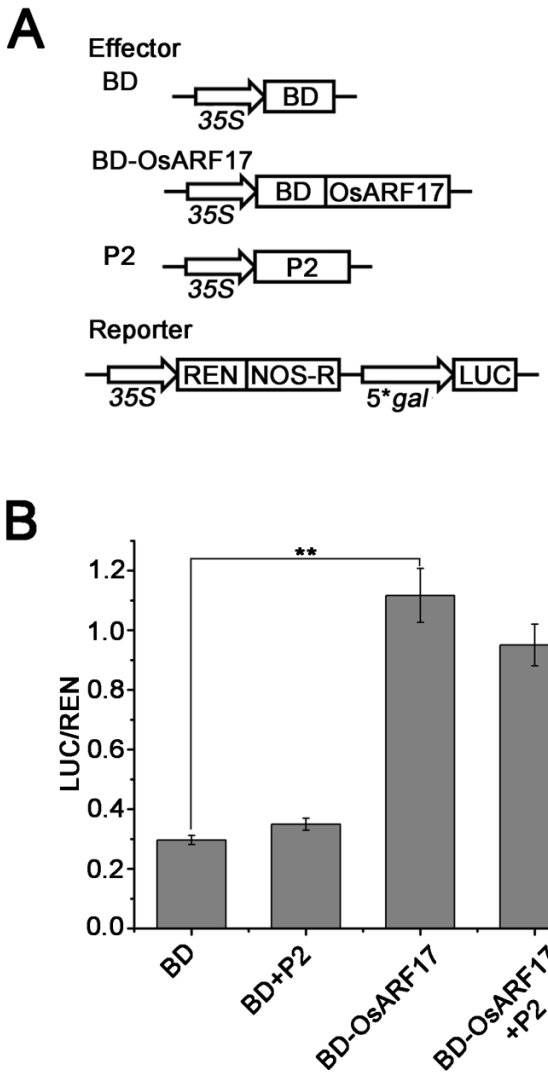

Fig.S14. P2 has no effect on the transcriptional activity of OsARF17. (*A*) Schematic diagrams of the effectors and reporters used in the dual-luciferase experiments. The effectors: BD, OsARF17-BD and P2 fused with FLAG-tag. The reporters: 35S: REN-NOS-R/5**gal* pro:LUC plasmids. BD domain could bind the *5*gal* promoter. (*B*) The relative LUC activities were measured in *N. benthamiana* cells using the combinations displayed in (*A*). The LUC/REN ratio represents the relative LUC activity. Values shown are the means ± SD of 5 biological replicates. Significant differences were identified using Fisher's least significant difference tests. **at the top of columns indicates significant difference at $p \leq 0.01$.

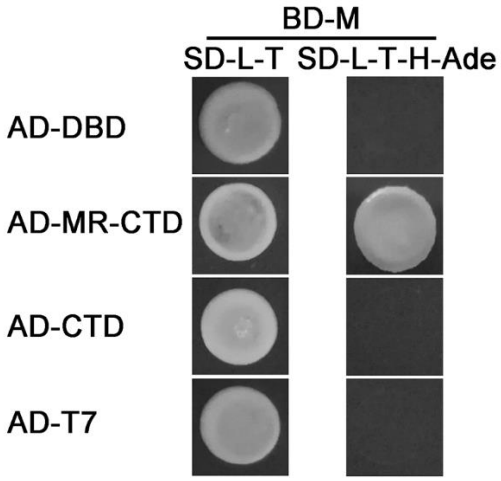

Fig.S15. RSMV M protein interacts with the MR-CTD domain of OsARF17 in Y2H assays. The different combinations of constructs transformed into yeast cells were grown on selective media SD/-Trp/-Leu (SD-L-T) and interactions were tested with SD/-Trp/-Leu/-His/-Ade (SD-L-T-H-Ade). Pictures were taken after 3 days of incubation at 30°C.

Table S1 The primers used in this study.

Gene Name	Primer Name	Sequence 5'-3'
For Yeast Two-Hybrid (Y2H) Assays		
OsARF homologues	AD-OsARF17-F	GACGTACCAGATTACGCTCATATGATGAGGCTTTCGTCGTCGT
	AD-OsARF17-R	GCAGCTCGAGCTCGATGGATCCTTAGAATTCAACTGAGCCGAC
	AD-OsARF6-F	GACGTACCAGATTACGCTCATATGATGAAGCTCTCGCCGTCG
	AD-OsARF6-R	GCAGCTCGAGCTCGATGGATCCTTAGAACTCAACTGAGCCCACC
	AD-OsARF12-F	GACGTACCAGATTACGCTCATATGATGAGCTCGTCGTCGGCG
	AD-OsARF12-R	GCAGCTCGAGCTCGATGGATCCTTAGGACAGATACCGTGGATCAT
	AD-OsARF25-F	GACGTACCAGATTACGCTCATATGATGAAGCTCTCTCCGCCG
	AD-OsARF25-R	GCAGCTCGAGCTCGATGGATCCTTAGTAGTCCAGAGGCGCTACA
	AD-OsARF11-F	GACGTACCAGATTACGCTCATATGATGGCGTCCTCGCAGGAG
	AD-OsARF11-R	GCAGCTCGAGCTCGATGGATCCTTAAGCTGCTTGAATACAGTCGT
	AD-OsARF16-F	GACGTACCAGATTACGCTCATATGATGAAGGATCAGGGATCATC
	AD-OsARF16-R	GCAGCTCGAGCTCGATGGATCCTTAACCTCTCCATGCATTTG
	AD-OsARF19-F	GACGTACCAGATTACGCTCATATGATGATGAAGCAGGCGCAGA
	AD-OsARF19-R	GCAGCTCGAGCTCGATGGATCCTTACGCAGTATTCCAATACCT
	AD-OsARF21-F	GACGTACCAGATTACGCTCATATGATGGCGAGCTCCGGCGG
	AD-OsARF21-R	GCAGCTCGAGCTCGATGGATCCTTAGTCGCCG GTTATCCT
OsARF17 truncated mutants	AD-DBD-MR-F	GACGTACCAGATTACGCTCATATGATGAGGCTTTCGTCGTCGT
	AD-DBD-MR-R	GCAGCTCGAGCTCGATGGATCCTTATTGCATTAGAAGTGACTGTGAA
	AD-DBD-F	GACGTACCAGATTACGCTCATATGATGAGGCTTTCGTCGTCGT
	AD-DBD-R	GCAGCTCGAGCTCGATGGATCCTTATACACTTGACTCTTCTGT
	AD-MR-F	GACGTACCAGATTACGCTCATATGATGACAGAAGAGTCAAGTGTA
	AD-CTD-F	GACGTACCAGATTACGCTCATATGATGTTTGGCGTTAACATAGATT
	AD-CTD-R	GCAGCTCGAGCTCGATGGATCCTTAGAATTCAACTGAGCCGACAG
	BD-DBD-F	ATCTCAGAGGAGGACCTGCATATGATGAGGCTTTCGTCGTCGT
	BD-DBD-R	GCCGCTGCAGGTCGACGGATCCTTATACACTTGACTCTTCTGT

	BD-CTD-F	ATCTCAGAGGAGGACCTGCATATGATGTTTGGCGTTAACATAGATT
	BD-CTD-R	GCCGCTGCAGGTCGACGGATCCTTAGAATTCAACTGAGCCGACAG
SRBSDV	BD-SP3-F	ATCTCAGAGGAGGACCTGCATATGATGCAAGACGTACAAAGGAC
	BD-SP3-R	GCCGCTGCAGGTCGACGGATCCCTATGTTTTATGAAGAAGAG GT
	BD-SP4-F	ATCTCAGAGGAGGACCTGCATATGATGTTGAAAG TAAATGTGCA GG
	BD-SP4-R	GCCGCTGCAGGTCGACGGATCCTCATTTTTTG GGATGAATTA AGA
	BD-SP5-1-F	ATCTCAGAGGAGGACCTGCATATGATGACATATTTGAAAGTGAAGAT
	BD-SP5-1-R	GCCGCTGCAGGTCGACGGATCCTCATCGCGCT GTAGTTGGTT
	BD-SP6-F	ATCTCAGAGGAGGACCTGCATATGATGTCTACCA ACCTCACGAA
	BD-SP6-R	GCCGCTGCAGGTCGACGGATCCTTACTCTGAA ATAAGTTGCCAG
	BD-SP7-1-F	ATCTCAGAGGAGGACCTGCATATGATGGATAGACCTGCTCGAGA
	BD-SP7-1-R	GCCGCTGCAGGTCGACGGATCCTTAAGATGATGGAGATTCAAAAA
	BD-SP8-F	ATCTCAGAGGAGGACCTGCATATGATGATCGGTACATACGATGAT
	BD-SP8-R	GCCGCTGCAGGTCGACGGATCCTTAACACAGAATACTAACGGCG
	BD-SP9-F	ATCTCAGAGGAGGACCTGCATATGATGGCAGACCTAGAGCGTA
	BD-SP9-R	GCCGCTGCAGGTCGACGGATCCTCAAACGTCCAATTTAAGTGAA
	BD-SP10-F	ATCTCAGAGGAGGACCTGCATATGATGGCTGACATAAGACTTGAC
	BD-SP10-R	GCCGCTGCAGGTCGACGGATCCTCATCTGGTGACTTTATTTAAC
RSV	BD-P2-F	ATCTCAGAGGAGGACCTGCATATGATGGCATTACTCCTCTTCAAT
	BD-P2-R	GCCGCTGCAGGTCGACGGATCCTTACATTAGAATAGGGCACT
	BD-P3-F	ATCTCAGAGGAGGACCTGCATATGATGAACGTGTTCACATCGTC
	BD-P3-R	GCCGCTGCAGGTCGACGGATCCCTACAGCACAGCTGGAGAGC
	BD-CP-F	ATCTCAGAGGAGGACCTGCATATGATGGGTACCAACAAGCCAG
	BD-CP-R	GCCGCTGCAGGTCGACGGATCCCTAGTCATCTGCACCTTCTG
	BD-MP-F	ATCTCAGAGGAGGACCTGCATATGATGGCTTTGTCTCGACTTTTG
	BD-MP-R	GCCGCTGCAGGTCGACGGATCCCTACATGATGACAGAAACTTC
RSMV	BD-N-F	ATCTCAGAGGAGGACCTGCATATGATGGCAACCGACAAGTCTTT
	BD-N-R	GCCGCTGCAGGTCGACGGATCCTTAAGCCTTGGTCTGGAAGAT
	BD-P-F	ATCTCAGAGGAGGACCTGCATATGATGAGTGTGCCAGAGGATAC

	BD-P-R	GCCGCTGCAGGTCGACGGATCCTCACAGTGCATCATCATAGTA
	BD-P3-F	ATCTCAGAGGAGGACCTGCATATGATGAAGATCATCTGCAGTACT
	BD-P3-R	GCCGCTGCAGGTCGACGGATCCTCAAGTAGCAAACTTGACATG
	BD-M-F	ATCTCAGAGGAGGACCTGCATATGATGGCCGTTCCGTGGACT
	BD-M-R	GCCGCTGCAGGTCGACGGATCCCTAACTCCAGATTATACTTCC
	BD-G-F	ATCTCAGAGGAGGACCTGCATATGATGATGAGGATTTCGGTCTTT
	BD-G-R	GCCGCTGCAGGTCGACGGATCCTTAGTCACTTTCTATAGTATATTC
	BD-P6-F	ATCTCAGAGGAGGACCTGCATATGATGGAGTTCAATTGGCCTTG
	BD-P6-R	GCCGCTGCAGGTCGACGGATCCTTAGGCATCATGCAGTACCC
RBSDV	BD-P8-F	ATCTCAGAGGAGGACCTGCATATGATGACTGGCACCCATGACGA
	BD-P8-R	GCCGCTGCAGGTCGACGGATCCTTATACAATAATCGAGGAAGCT
For Yeast Three-Hybrid (Y3H) Assays		
OsIAA20	BD-OsIAA20-F	ATCTCAGAGGAGGACCTGCATATGATGGAGCTCGAGCTCGGC
	BD-OsIAA20-R	GCCGCTGCAGGTCGACGGATCCTTACACCAGTATCTTGAGCCGT
	pBridge-BD-OsIAA20-F	GTTGACTGTATCGCCGGAATTCATGATGGAGCTCGAGCTCGGC
	pBridge-BD-OsIAA20-R	TTAGCTTGGCTGCAGGTCGACTTACACCAGTATCTTGAGCCGT
OsARF17 CTD domain	pBridge-BD-CTD-F	GTTGACTGTATCGCCGGAATTCATGTTTGGCGTTAACATAGATT
	pBridge-BD-CTD-R	TTAGCTTGGCTGCAGGTCGACTTAGAATTCAACTGAGCCGACAG
SRBSDV P8 protein	pBridge-Met-SP8-F	GAAGAAGAGAAAGGTGGCGGCCGCATGATGATTGGCACCTATGATGA
	pBridge-Met-SP8-R	CATGGGAGATCAGCCCGAAGATCTTTAGCAAAGAATAGACACAGCT
For Dual Luciferase Assays		
OsARF17	pCV-BD-OsARF17-F	GTTGACTGTATCGCCGGTCGACATGAGGCTTTCGTCGTCGT
	pCV-BD-OsARF17-R	CGATCGGGGAATTCGAGCTCTTAGAATTCAACTGAGCCGAC
SRBSDV P8 protein	pCV-BD-SP8-F	GTTGACTGTATCGCCGGTCGACATGATCGGTACATACGATGAT
	pCV-BD-SP8-R	CGATCGGGGAATTCGAGCTCTTAACACAGAATACTAACGGCG
RSV P2 protein	pCV-BD-P2-F	GTTGACTGTATCGCCGGTCGACATGGCATTACTCCTCTTCAAT
	pCV-BD-P2-R	CGATCGGGGAATTCGAGCTCTTACATTAGAATAGGGCACT
OsGH3.5 promoter	pGREEN-proOsGH3.5-F	CTTGATATCGAATTCCTGCAGCATGCAGAGAGATGCTGAAAT
	pGREEN-proOsGH3.5-R	ATGTTTTTGGCGTCTTCCATGGGCTGTGAATTGTGATGTGAAG

For construction of overexpression vector		
OE17	OE17-F	GTTCCAGATTACGCTGGATCCATGAGGCTTTCGTCGTCGT
	OE17-R	ATCGGGGAAATTCGAGCTCTTAGAATTCAACTGAGCCGAC
SP8	SP8-F	GTTCCAGATTACGCTGGATCCATGATCGGTACATACGATGAT
	SP8-R	ATCGGGGAAATTCGAGCTCTTAACACAGAATACTAACGGCG
For RT-PCR		
RBSDV	RB-F	AGCACTACAGATGTTACACAC
	RB-R	GCCTGTCCCTATCAGATTTCAAC
SRBSDV	SRB-F	CGCGTCATCTCAGACTACAG
	SRB-R	TTTGTCAGCATCTAAAGCGC
RSV	RSV-CP-1F	AGGCAATCAATGACATCTCC
	RSV-CP-1R	ATCTCTCACAAAGCCAGTGC
RSMV	RSMV-detection-F	TGCACAGACGTTAGTGAGTTAC
	RSMV-detection-R	TCCGTCTTTCATAGCCTTCAG
17cas	17cas-F	ACCGGGCTAAGTGGTTGTAG
	17cas-R	ATTTCCCACAGGGTCACAGC
For qRT-PCR		
OE17	qRT17-F	GCTTTGGGAGATTGAGCCCT
	qRT17-R	TCTCGGAGCCACATGAGAGA
SP8	qRTSP8-F	TCAGTTCTCTTCGACCCACC
	qRTSP8-R	GCCGAAACTAGCTCTCTCAC
SRBSDV	qSR-S4-F	AAAGTGAACCCGTTGCTGAC
	qSR-S4-R	TGCAACGCTAGATCCTATGC
	qSR-S6-F	ATCTGCTTTTCCCCTTCCGA
	qSR-S6-R	GATTCCGCGTTTGAAGAGTCA
	qSR-S10-F	ACAAACATGGAGCGGAGTCT
	qSR-S10-R	CGGTCTTACGCAACGATGAA
RBSDV	qRB-S4-F	ATTTGCGTTTTGCAATTTCC
	qRB-S4-R	GCTCTACGACGACCAAATCC

	qRB-S6-F	AGCGTGTTGAAAACGAGATC
	qRB-S6-R	CGCTTTGCAAATTCAACAAG
	qRB-S10-F	AACAACCGACCAACAATCAC
	qRB-S10-R	GAGCAGGAACTTCACGACAG
RSV	qRSV-CP-F	AGGCAATCAATGACATCTCC
	qRSV-CP-R	ATCTCTCACAAAGCCAGTGC
RSMV	qRSMV-N-F	AGAGGTTGGAGAGGGGAAGA
	qRSMV-N-R	TAGCCGCCCTTCTATCCTTG
For BiFC Assays		
OsARF17	OsARF17-cYFP-F	CTGTACAAGTCCGGAGTCGACATGAGGCTTTCGTCGTCGT
	OsARF17-cYFP-R	CGATCGGGGAAATTCGAGCTCGAATTCAACTGAGCCGAC
SRBSDV P8 protein	nYFP-SP8-F	ATCGAGGACTCCGGAGTCGACATGATCGGTACATACGATGAT
	nYFP-SP8-R	CGATCGGGGAAATTCGAGCTCTTAACACAGAATACTAACGGCG
For FRET-FLIM Assays		
PCV-SP8-Tur	mTur-SP8-F	GGACTCTAGACCCCTGGGATCCATGATCGGTACATACGATGAT
	mTur-SP8-R	CCTCGCCCTTGCTCACCATGTCGACACACAGAATACTAACGGCG
PCV-OsARF 17-Ven	Ven-OsARF17-F	GGACTCTAGACCCCTGGGATCCATGAGGCTTTCGTCGTCGT
	Ven-OsARF17-R	CCTCTCCCTTGCTCACCATGTCGACGAATTCAACTGAGCCGAC
For EMSA Assays		
	OsGH3.5-cis-F	CAGACTGAAAAATGGGTGTAGAGACAACAGGGTTGGTGACTGCAGAT ATT
	OsGH3.5-cis-R	AATATCTGCAGTCACCAACCCTGTTGTCTCTACACCCATTTTTCAGTCT G
For protein purification		
SRBSDV P8 protein	pColdTF-SP8-F	GTGGTATCGAAGGTAGGCATATGATGATCGGTACATACGATGAT
	pColdTF-SP8-R	ACAAGCTTGAATTCGGATCCTTAACACAGAATACTAACGGCG
OsARF17 trunted DBD domain	pColdTF-DBD-F	GTGGTATCGAAGGTAGGCATATGATGTACGCTCAGATGACGCT
	pColdTF-DBD-R	ACAAGCTTGAATTCGGATCCTTAATCACTAGATAGGACTGAA

OsARF17 CTD domain	pColdTF-CTD-F	GTGGTATCGAAGGTAGGCATATGATGTTTGGCGTTAACATAGATT
	pColdTF-CTD-R	ACAAGCTTGAATTCGGATCCTTAGAATTCAACTGAGCCGACAG
	6P1-CTD-F	TCCAGGGGCCCCTGGGATCCATGTTTGGCGTTAACATAGATT
	6P1-CTD-R	GTCAGTCACGATGCGGCCGCTTAGAATTCAACTGAGCCGACAG
RSV P2 protein	pET28a-P2-F	CTGTATTTTCAGGGCCATATGATGGCATTACTCCTCTTCAAT
	pET28a-P2-R	ACGGAGCTCGAATTCGGATCCTCACATTAGAATAGGGCACTC
For MYC or FLAG tags transient expression vectors		
MYC tagged	Lic-OsARF17-F	CgACgACAAgACCgTCACCATGAGGCTTTCGTCGTCGT
	Lic-OsARF17-R	gAggAgAagAgCCgTCgGAATTCAACTGAGCCGACAG
FLAG tagged	Lic-SP8-F	CgACgACAAgACCgTCACCATGATCGGTACATACGATGAT
	Lic-SP8-R	gAggAgAagAgCCgTCgACACAGAATACTAACGGCG
	Lic-P2-F	CgACgACAAgACCgTCACCATGGCATTACTCCTCTTCAAT
	Lic-P2-R	gAggAgAagAgCCgTCgCATTAGAATAGGGCACT
MYC tagged	Lic-DBD-F	CgACgACAAgACCgTCACCATGAGGCTTTCGTCGTCGT
	Lic-DBD-R	gAggAgAagAgCCgTCgTACACTTGACTCTTCTGT
	Lic-CTD-F	CgACgACAAgACCgTCACCATGTTTGGCGTTAACATAGATT
	Lic-CTD-R	gAggAgAagAgCCgTCgGAATTCAACTGAGCCGACAG
MYC or FLAG tagged	Lic-GFP-F	CgACgACAAgACCgTCACCCTGGACGGCGACGTAAAC
	Lic-GFP-R	gAggAgAagAgCCgTCgGTTGTGGCGGATCTTGAAGT

Table S2 FRET-FLIM assays showing that OsARF17 interacts with SP8.

Donor	Acceptor	τ	Δt	Number	E	p-value
mTur-SP8	-	3.9633	-	30	-	-
mTur-SP8	Ven	3.9563	7	32	0.18	0.94854
mTur-SP8	Ven-OsARF17	3.2242	739	34	34.7	3.83403E-4

Mean lifetime, τ, in ns. $\Delta t = \tau_D - \tau_{DA}$ (in ps). τ_{DA}, the donor in the presence of the acceptor; τ_D, the donor in the absence of the acceptor. Number: total number of measured. % FRET efficiency: $E = 1 - (\tau_{DA}/\tau_D)$. p-value significant differences were identified using Fisher's least significant difference tests.

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