Supplementary Materials: The Transcription Factor OsWRKY45 Negatively Modulates the Resistance of Rice to the Brown Planthopper *Nilaparvata lugens*

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1     ATGACGTCATCGATGTCGCCGGCGCCGGCGCCGGCGTACGCGCAGGTGATGGAGGACATG
1     M  T  S  S  M  S  P  A  P  A  P  A  Y  A  Q  V  M  E  D  M
61    GAGAAGGGGAAGGAGCTGGCGGCGCAGCTGCAGGGGCTCCTCCGCGACTCGCCGGAGGCC
21    E  K  G  K  E  L  A  A  Q  L  Q  G  L  L  R  D  S  P  E  A
121   GGCCGCTTCGTCGACCAGATTCTCCACACCTTCTCCCGGGCGATGCGGGCGCTCGACAAG
41    G  R  F  V  D  Q  I  L  H  T  F  S  R  A  M  R  A  L  D  K
181   GCGGCGGTCTCCGCCGCCGGAGGAGAAGGGTCGGAGGTGCAGAGCGAGGTCACCTGCGGG
61    A  A  V  S  A  A  G  G  E  G  S  E  V  Q  S  E  V  T  C  G
241   GGCGGGGCCAGCGCCGGCGGGAAGAGGAAAGCCCCCGCCGCCGACCGGAAGGCCAACTGC
81    G  G  A  S  A  G  G  K  R  K  A  P  A  A  D  R  K  A  N  C
301   CGCAGGAGGACGCAGCAATCGTCCGGGAATTCGGTGGTCGTCAAGAACCTCGACGACGGC
101   R  R  R  T  Q  Q  S  S  G  N  S  V  V  V  K  N  L  D  D  G
361   CAGGCATGGCGCAAGTACGGGCAGAAGGAGATCCAAAACTCCAAGCACCCAAAGGCCTAC
121   Q  A  W  R  K  Y  G  Q  K  E  I  Q  N  S  K  H  P  K  A  Y
421   TTCCGGTGCACGCACAAGTACGACCAGCTGTGCACGGCGCAGCGGCAGGTGCAGCGCTGC
141   F  R  C  T  H  K  Y  D  Q  L  C  T  A  Q  R  Q  V  Q  R  C
481   GACGACGACCCGGCGAGCTACAGGGTCACCTACATCGGCGAGCACACCTGCCGGGACCCG
161   D  D  D  P  A  S  Y  R  V  T  Y  I  G  E  H  T  C  R  D  P
541   GCCACCGCCCCCATCATCGCGGCGCACGTCATCCACCAGGTCGCCGCCGGCGACAACGAC
181   A  T  A  P  I  I  A  A  H  V  I  H  Q  V  A  A  G  D  N  D
601   GACGGCTGCGGCGGCCTCCAAGCGGGGTCCCGCCTCATCAGCTTCGTCGCCGCGCCGGCG
201   D  G  C  G  G  L  Q  A  G  S  R  L  I  S  F  V  A  A  P  A
661   GCGCCAGTAGACGCTGCCGCGGCGCCGACGACCAGCACGATCACCACGGTCACCGCGCCG
221   A  P  V  D  A  A  A  A  P  T  T  S  T  I  T  T  V  T  A  P
721   GGCCCGCTGCTGCAGCCGCTCAAGGTGGAGGGCGGCGTCGGCTCGTCCGACCAGGAGGAG
241   G  P  L  L  Q  P  L  K  V  E  G  G  V  G  S  S  D  Q  E  E
781   GTGCTGAGCAGCCTCACGCCCGGCAGCTCCGCGGCGCGCGGCGGCGGCGGCGGCGGCGGA
261   V  L  S  S  L  T  P  G  S  S  A  A  R  G  G  G  G  G  G  G
841   GTCGCGGGTCCCTTCGGGCCGGACCAGGGCGATGTCACGTCCTCCCTGCACTGGAGCTAC
281   V  A  G  P  F  G  P  D  Q  G  D  V  T  S  S  L  H  W  S  Y
901   GACGCCGTCGCCGGCATGGAGTTCTTCAAGAACGACGAGGTTGTCTTCGATCTGGACGAC
301   D  A  V  A  G  M  E  F  F  K  N  D  E  V  V  F  D  L  D  D
961   ATTATGGGTTTGAGCTTTTGA
321   I  M  G  L  S  F  *
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Figure S1. Sequences of nucleotides and deduced amino acids of *OsWRKY45*. The stop codon is marked by an asterisk.

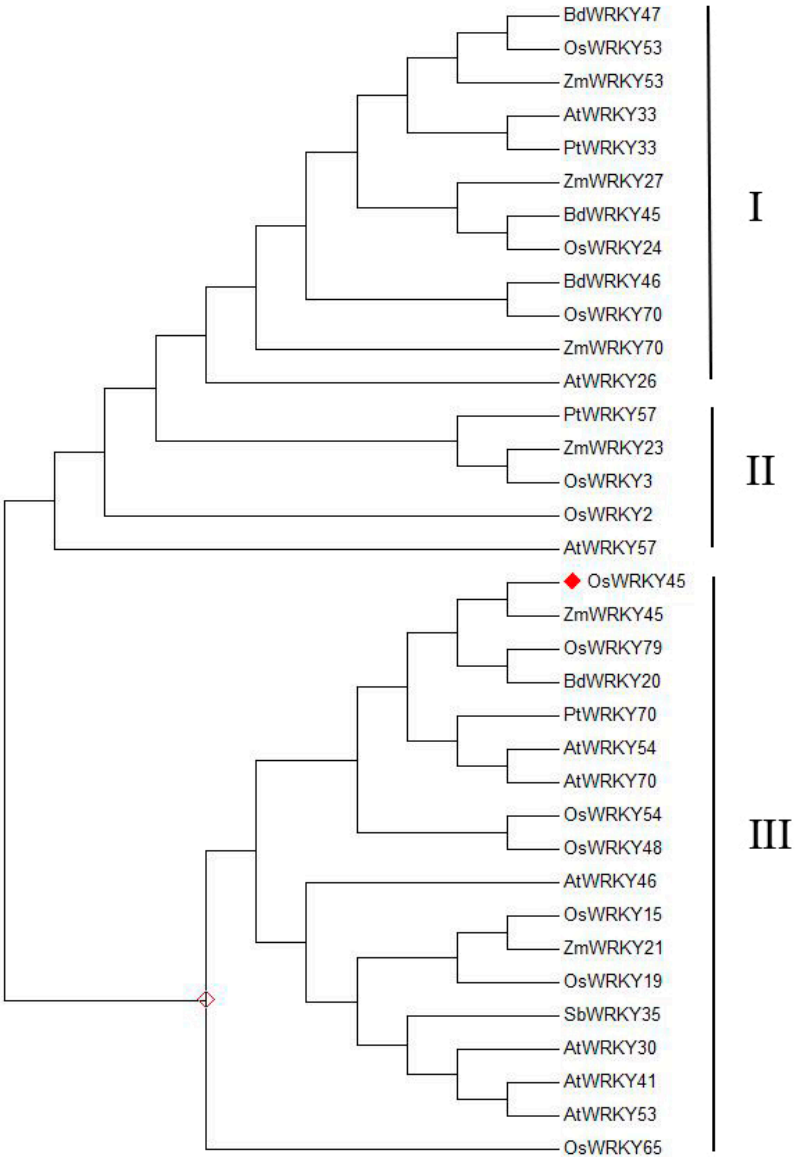

Figure S2. Phylogenetic analysis of WRKY TFs from different species. The unrooted tree was constructed with a neighbor-joining method on the basis of the alignment of protein sequences and confirmation of the tree topology by bootstrap analysis (1000 replicates) were performed with MEGA software (default settings except the replicates of the bootstrap value). Species acronyms are included before the protein name: At, *Arabidopsis thaliana*; Bd: *Brachypodium distachyon*; Pt: *Populus trichocarpa*; Sb: *Sorghum bicolor*; Ta: *Triticum aestivum*; Zm: *Zea mays*. Sequence data in the phylogenic tree can be found in the GenBank/EMBL data libraries under accession number: AtWRKY26 (At5g07100); AtWRKY30 (At5g24110); AtWRKY33 (At2g38470); AtWRKY41 (At4g11070.1); AtWRKY46 (At2g46400); AtWRKY53 (At4g23810); AtWRKY54 (At2g40750); AtWRKY57 (AT1G69310); AtWRKY70 (At3g56400); BdWRKY20 (Bradi1g63220.1); BdWRKY45 (Bradi2g53760.1); BdWRKY46 (Bradi2g22440.1); BdWRKY47 (Bradi2g00280.1); OsWRKY2 (Os10g42850); OsWRKY3 (Os03g55080); OsWRKY15 (Os01g46800.1); OsWRKY19 (Os05g49620.1); OsWRKY24 (Os01g61080); OsWRKY45 (Os05g25770.1); OsWRKY48 (Os05g40060); OsWRKY53 (Os05g27730); OsWRKY54 (Os05g40080); OsWRKY65 (Os12g02470.1); OsWRKY70 (Os05g39720); OsWRKY79 (Os03g21710.1); PtWRKY33 (Potri.013G153400.1); PtWRKY57 (Potri.008G094000.1); PtWRKY70 (Potri.013G090300.1); SbWRKY35 (Sobic.010G045700.1); ZmWRKY21 (GRMZM2G408462_T01); ZmWRKY23 (GRMZM2G018721_T01); ZmWRKY27 (GRMZM2G036703_T01); ZmWRKY45 (GRMZM2G004060_T01); ZmWRKY53 (GRMZM2G012724_T01); ZmWRKY70 (GRMZM2G012724_P01). I, II and III represent group I, group II and group III of WRKY TFs, respectively. The red diamond indicates the gene *OsWRKY45* that is characterized in this paper.

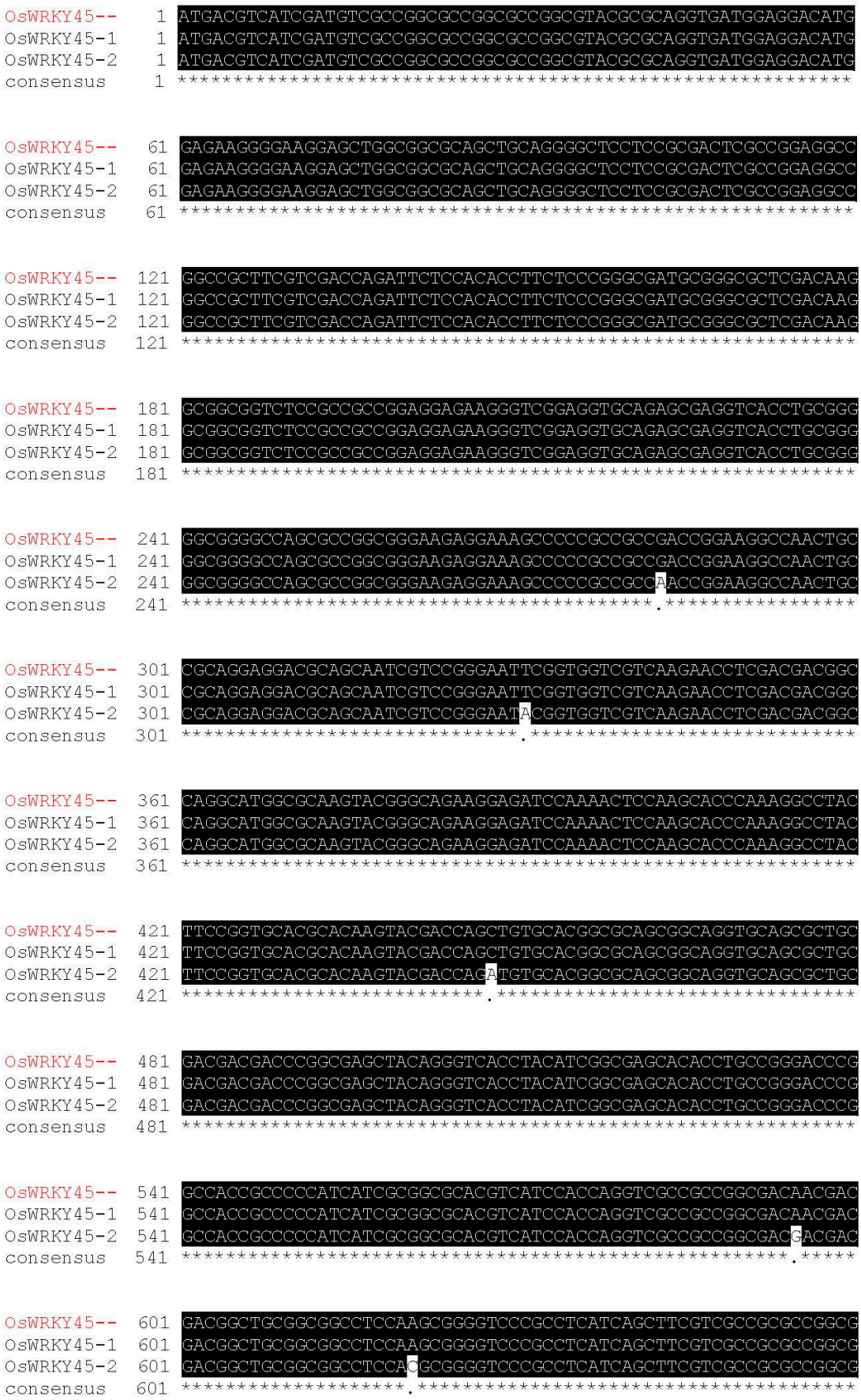

Figure S3. *Cont.*

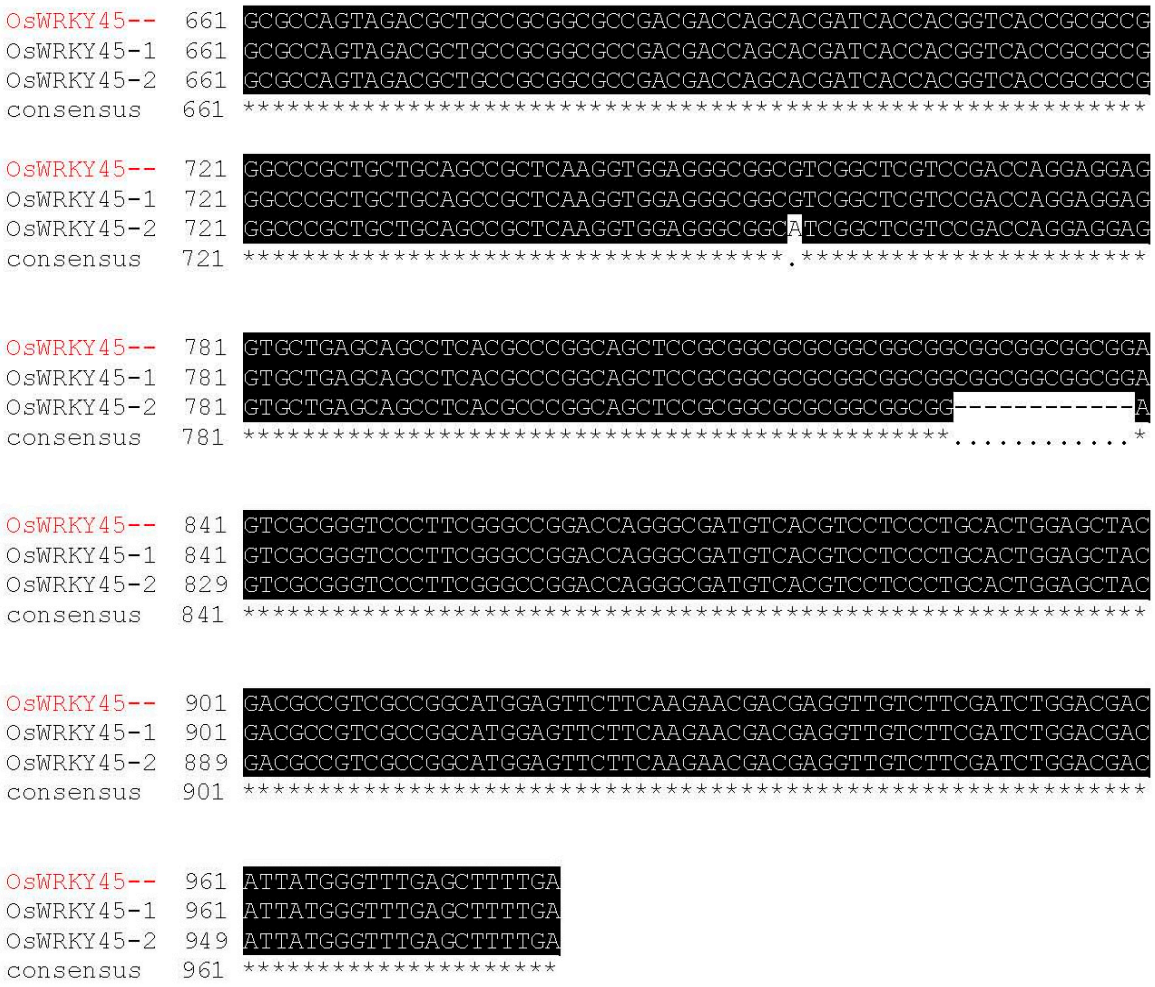

Figure S3. cDNA sequence alignment of *OsWRKY45* cloned in this article with *japonica*-derived *WRKY45* (*OsWRKY45*-1) and *indica*-derived*WRKY45* (*OsWRKY45*-2). The asterisk indicates the bases that are identical in the three genes; the dot indicates the bases that are identical in two of the three genes. Non-shaded spaces represent inconsistent bases in the three genes.

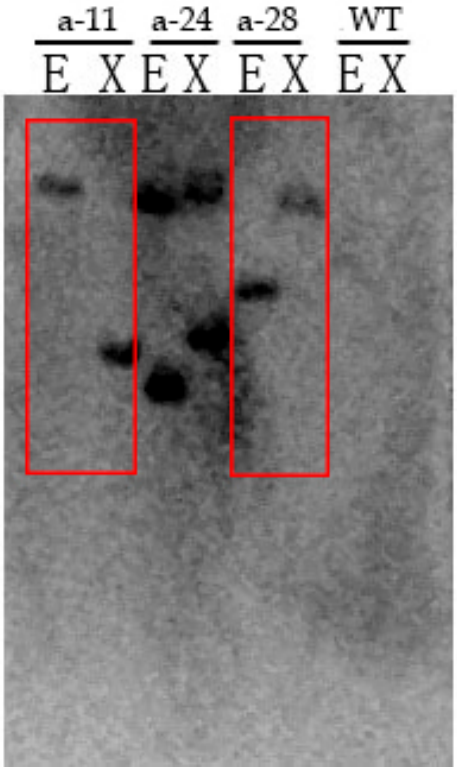

Figure S4. DNA gel-blot analysis of as-*wrky*. Genomic DNA was digested with *Eco*R I (E) or *Xba* I (X). The blot was hybridized with a probe specific for reporter gene *gus*. Two transgenic lines (as-11 and as-28) have a single insertion of the transgene.

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OsWRKY45      ATG--------------------------ACGTCATCGATGTCGCCGGCGC--------
OsWRKY15      ------------------------------------ATGGCCG---------------
OsWRKY48      ATGGCGCTGATTGCCACTGGTGCTACCGCTACTGCTACTGCGGCGCCGGTGGCTAGCCCG
OsWRKY54      ATGGCGCTCACTATGGTTAGTCTCGCCGC-----------CATCGCCGG---GAAGCC--
                                                    . **

OsWRKY45      -------CGGCGCCGGCGTACGCGCAGGTGATGGAGG---------ACATGGAGAAGGGG
OsWRKY15      ----------CCGGAGCGGACGAGCGGTGCCGCGCGCTGGTGTCCGGCCTGCTGTCGTCG
OsWRKY48      GCTGCTTCTTCCATGGCGTCCG---AGCTGATGGCGCAGGGG------CGGGAGTCCGCC
OsWRKY54      --------CACCGCGGCGTCGGCTCAGCTCGTCGCCGAGGGG------AGGGAGTCGGCG
                        *   .*** . *   .*      *.             . * :*:.

OsWRKY45      AAG----GAGCTGGCGGCGCAGCTGCAGGGGCTCCTCCGCGACTCGCCGGAGGCC-----
OsWRKY15      ATCGACCGGTCTATAAGCATCGCGAG----------------------GTCGT---GCTG
OsWRKY48      ----GCCGTCCTGGAAGCCCTGCTCCATG-GCGCGTC-GC-TCCCGCCGGCGCACGGCGG
OsWRKY54      ----GCCCGCCTGTATGCCCTGCTGGTCG-GCTCGTC-----------GGCGCTCCACGG
                        **. . **   **                        * .*

OsWRKY45      ----GGCCGCTTCGTCGACCAGATTCTCCACACCTTCTCCCGGGCGATGCGGGCGCTCGA
OsWRKY15      CACGGAGGCCGCCGCCGCCGGG----------------------CG---------CCTGA
OsWRKY48      TGCCCACGCCCTCGCCGCCGAGATCCTTCGCTGCTGCGACCGCGCG---------CTCGC
OsWRKY54      CCCTGTGGGCCTCGCCGAGCAGATCCTCCTCTGCTTCGACCGCGCG---------CTCGC
                       *  ** **.  .*                     **         *  *.

OsWRKY45      CAAGGC---GGCGGTCTCCGCCGCCGGAGGA----------------------GAAGGGT
OsWRKY15      C--CCAGCAGGCCGGCGCCG-CGCCTGAGTCGCCGCCGTCCGCCGACGGTAGCGCCGGCT
OsWRKY48      CGCGTTGCGCGCCGGCGGCGATGCCGAA-TC---GTCGTCAGC-----------------
OsWRKY54      CAAGCTGCACGGCGTCAACCTCGCCGGA-GC---TGAGGACGACGACGACGCTGCCGGCA
              *         *  * *  *   *** .*  .

OsWRKY45      C-----------GGAGGTGCAGAGCGAGGTCA--CCTG-CGGGGGCGGG-----------
OsWRKY15      CGGACCTCGGC--------G----CCGACT--------CCCGGTGCCGCGCCAATG-----
OsWRKY48      -------CGACACGAAGCGCAAGCCGGCAACCGCCCAGCCGTCGACGCGCCGGAGGAG--
OsWRKY54      ACGGCAACGGCAGGAAGCGCAAGCCGGGG----------CGTGGTCGTGGCTTGACTGCC
                         *     **.           **  * ** *

OsWRKY45      --GCCAGCGCCGGCGG----GAAGAGGAAAGCCCCCGCCGCCGACCGGAAGGCCAACTGC
OsWRKY15      ----CGGCGGGGCCGTGCAAGAAGAGGAAGA-------C------------GCT-----G
OsWRKY48      --AAGAGCGACGGCGAGCGGCG----------------------------GCGGAGCGG
OsWRKY54      GCTTCAGCTGCAGCGAGCTCCAAAAGGATGA-------G------------GGTGAGCAA
                   .**   . **      .                            *

OsWRKY45      CGCAGGAGGACG------CAGCAATCGTCCG---GGAATTCGGTGGTCGTCAAGAACCTC
OsWRKY15      CCCA------AGTGGAGCAAGCAGGTGAAGGTGAGGTCCGTGCAGGACGTCGGCCCCCTC
OsWRKY48      C---GGCGGCGGCGGAGCCGGCGA----------GGGTGGAGAAG-GCGCGG--ACGTCG
OsWRKY54      TGCTGGAGGAAATGGA----GCAA----------GGATTGAGAGG-AAAGCG--ACAATG
                        .       **..          **     *  *  ..  .  ..

OsWRKY45      GACGACGGCCAGGCATGGCGCAAGTACGGGCAGAAGGAGATCCAAAACTCCAAGCACCCA
OsWRKY15      GACGACGGGTTCAGCTGGAGGAAGTACGGGCAGAAGGACATCCTCGGCGCCAAGTACCCA
OsWRKY48      GAGGACGGATTCCTGTGGAGGAAGTACGGGCAGAAGGAGATCAAGAACAGCAAGCACCCG
OsWRKY54      GATGACAAATTTCTGTGGAGGAAGTACGGGCAGAAGGAGATCAAGAACAGCAAGCACCCG
              ** ***..  :    ***.* ***************** ***.: ..*  **** ****.

OsWRKY45      AAGGCCTACTTCCGGTGCACGCACAAGTACGACCAGCTGTGCACGGCGCAGCGGCAGGTG
OsWRKY15      AGAGCCTACTTCCGGTGCACGCACCGGCACACGCAGGGCTGCCACGCCAGCAAGCAGGTG
OsWRKY48      AGGCTCTACTACCGGTGCAGCTACAAGGACGACCATGGCTGCACGGCGACGAAGCAGGTC
OsWRKY54      AGGTTTTACTACCGGTGCAGCTACAAGGACGACCATGGCTGCACGGCGACGAAGCAAGTC
              *..   ****:********   **..* **.. **    ***.. ** .  ..***.**
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Figure S5. *Cont.*

```
OsWRKY45      CAGCGCT---------------GCGACGACGACCCGGCGAGCTACAGGGTCACCTACATC
OsWRKY15      CAGCG----------CGCGGACGGCG------ACCCGCTGCTCTTCGACGTCGTGTACCAC
OsWRKY48      CAGCAGTCGGAGGAA---------G------ACCCTTCCCTCTACGTCATCACCTACTTC
OsWRKY54      CAGCAGTCGGAGACGGCGGACGACGACACTGCCTCGCCCGTCTACATCATCACCTACTTC
              ****.                  *      .* *      **:*.  .**.  *** :*

OsWRKY45      GGCGAGCACACCTGCCGGGA---CCC---GGCCA---CCGCCCCCATCATCG--------
OsWRKY15      GGCGACCACACCTGCGCGCATGGCGTGCGCTCCGCCGCCGCCGCCATCGACGGCCAGGCC
OsWRKY48      GGCGACCACACCTGCAGCT---GCCAGACGGCCGCCGCCGCCGCCATGGACGACGACGA-
OsWRKY54      GGCGAGCACACCTGCCG------CCATGGGGACGACGCCGCCGCCATGGTCGTCGACGG-
              ***** *********        *       .*.   ***** **** .:**

OsWRKY45      -CGGCGCACGTCATC------CACCAGGTCGCCGCCGGCGA-------CAACGACGACGG
OsWRKY15      GCGGCGAGCGCGGAGCAAAAGCATCA-GCCGACGCCGCCGCAGGAGCAGAACGCCGTCTC
OsWRKY48      -CGACGA----TGA---AAACTCGCA-G------------C------ATTTCGTCATCAA
OsWRKY54      -CGGCGA----GGA---GGAGGATCA-GCTCTCGCCGGCGC------AGATGGTCATCAG
               **.**.     .:        . ** *            .        :: * *.:*

OsWRKY45      CTGCGG---CGGCCTCCAAGCGGGGTCCCGCCTCATCAGCTTCGTCGCCGCGCCGGCGGC
OsWRKY15      GGTCGCGTTC-------------------------------------ACCTCCATG-----
OsWRKY48      CTTCGGGCC-GGCCACCGCG-----AGCCG---CAGC-GGTTCG---CCGCCTCT-----
OsWRKY54      CTTCGCTTCCAGCAACG------------G---CGGC-GATGCTTCTGTGTCCTG-----
                 **

OsWRKY45      GCCAGTAGACGCTGCCGCGGCGCCGACGACCAGCACGA-----TCACCACGGTCACCGCG
OsWRKY15      GCCG-TCGTCAACGCATCGACGTCGTCGCCCTT----------------------------
OsWRKY48      GCTCTACGAC-----GACGGCGACGACGGCGATGTCTGGAGGGAGACGGCGGCCACCCCA
OsWRKY54      GCCGTGC--------TCCGGCGACGACGCCCAGAA--------------------------
              **    .          **.** **:** * :

OsWRKY45      CCGGGCCCGCTGCTGCAGCCGCTCAAGGTGG---AGGGCGGCGTCGGCTCGTCCGACCAG
OsWRKY15      ------------------------CGTGTCC---CCGGCGATGTCGG-----ACTGCCAG
OsWRKY48      CCGTCGTCGAGGCAG-------TCCAGGTGCTCGCCGGAGGGA---G-----ACGGGGAG
OsWRKY54      -CAACAGCGAGACATCGC--ATGAATCGAGCCCGCCGGAGGCGCCGG-----CCGGCGAG
                                .  *:     . **.*. .   *     .* .  **

OsWRKY45      GAGGAGGTGCTGAGCAGCCTCACGCCCGGCAGCTCCGCGGCGCGCGGCGGCGGCGGCGGC
OsWRKY15      ATTA-----------------------------GCTACGAGCTGGGCGGTGGCAGCATG
OsWRKY48      GAATCCGGGGTG----------------------AAGATGAGCAAGGAAG---------A
OsWRKY54      GAAGAACGTCTG----------------------CGGCCGTGCAC--------------A
              .:                                  . * **

OsWRKY45      GGCGGAG----TCGCGGGTCCCTTCGGGCCGGACCAGGGCGATGTCACGTCCTCCCTGCA
OsWRKY15      GCCGGAGTCCGCAACGTGC-CCGACGT----------G---GAGCT--TGCCT-------
OsWRKY48      GCCAGTGG---ACTCGTGT-CCG--GGGCCGTCAG-CG---GTGAGTTCGCCGGCCGACG
OsWRKY54      GCCGCCG------GCGTGT-CTGACGAACCGATCA-TG---GAGTCAACGCCG-CCGGCG
              * *.  *       ** *  *    *           *   .:*      **

OsWRKY45      CTGGAGCTACGACGCCGTCGCCGGCATGGAGTTCTTCAAGAACGACGAGGTTGTCTTCGA
OsWRKY15      --------CCAAGACGAACTCCTCCATGGGAGACGACATGG------AGTTCATGTTCTC
OsWRKY48      TCG---TTTCTTGTTCGTCTCCGGCAATGGAGCCCG--AT------------------CT
OsWRKY54      CCGGAGCTTCTTGC--------GGATCTGAAGCCCA--TG---------GATGGGTGCCT
                      * :           .:  *..  *    :

OsWRKY45      TCTGGACGACATT-------------------------------------------------
OsWRKY15      GCTGGACTCCGATTTCTTGGA-------------------------------CACCTA---
OsWRKY48      GCTGGGCTGCTTGAACTGGGATGATGACTTTGGAGACAGCTCGTTCGTCGACGCCGATGA
OsWRKY54      CCTTGACGGCGAGAG------------------------CTTGTTTGGCAT---GGATGA
               ** *.*  * :
```

Figure S5. *Cont.*

```
OsWRKY45      --------------ATGGGTTTGAGCTTTTGA----------------------------
OsWRKY15      -------------CAAGTACTCCAGCTATTTCTAA------------------------
OsWRKY48      GTTCATGAATTTCGATGAGATTGATCTGTTTCAAATCTACTCCTAG--------------
OsWRKY54      ACTCGTCTACTTTCATGAGC----------TCTCAGCTGCTCTCGGCCTACTCGATCGAG
                           *:* .            .

OsWRKY45      --------------------
OsWRKY15      --------------------
OsWRKY48      --------------------
OsWRKY54      ACTGGGGTGCGCCAGTATAG
```

Figure S5. cDNA sequence alignment of *OsWRKY45*, *OsWRKY15*, *OsWRKY48* and *OsWRKY15*. Sequence used for *OsWRKY45* antisense transformation is underlined in red. The asterisk under sequences indicates the bases that are identical in the four genes; the colon indicates the bases that have strongly similar properties among sequences; the dot indicates the bases that have weakly similar properties among sequences.

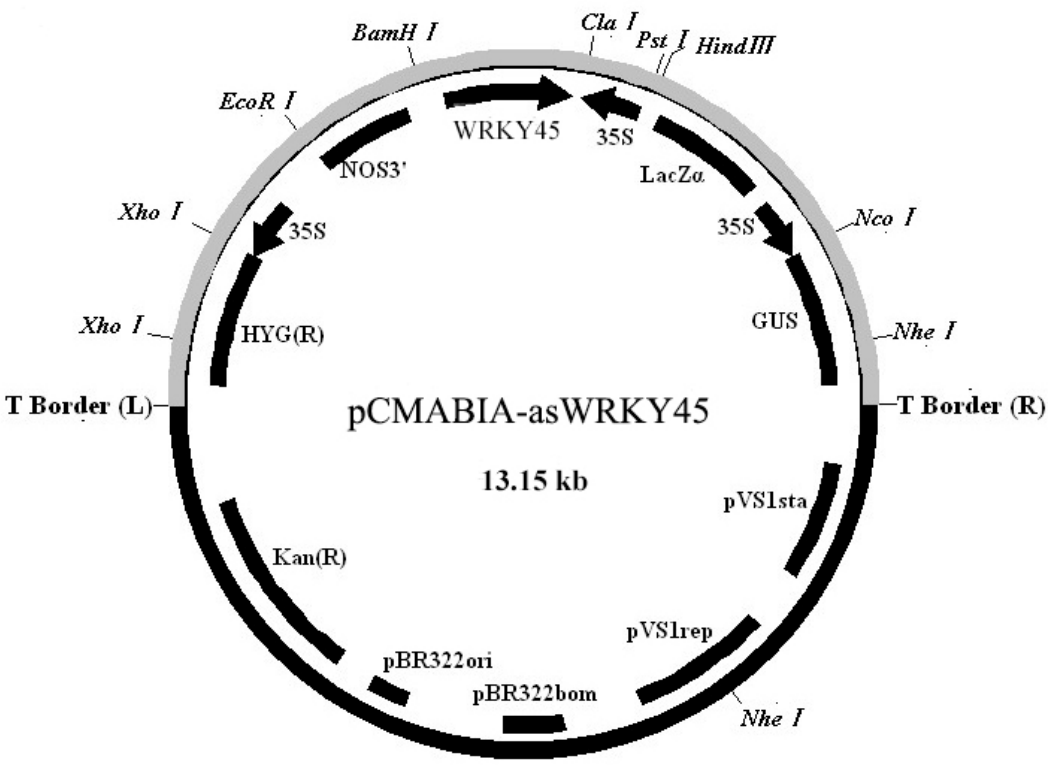

Figure S6. Transformation vectors were used in this study.

Table S1. Primers and probes used for qRT-PCR of target genes.

Gene Name	TIGR ID	Forward Primers(5′–3′)	Reverse Primers (3′–5′)	Probe
ACTIN	Os03g50885	TGGACAGGTTATCACCATTGGT	CCGCAGCTTCCATTCCTATG	CGTTTCCGCTGCCCTGAGGTCC-BHQ1
MPK6	Os06g06090	CGCACGCTCAGGGAGATC	GGTATGATATCCCTTATGGCAACAA	CTCCGCCACATGGACCACGAGAA-BHQ1
MPK3	Os03g17700	CGACTTCGAGCAGAAGGCTCTA	GTTCATCTCGATCGCTTCGTT	ACGAGGACCAAATGAAGCAGCTGAT-BHQ1
WRKY45	Os05g25770	TCGGTGGTCGTCAAGAACC	AAGTAGGCCTTTGGGTGCTT	CAAGTACGGGCAGAAGGAGATCCAA-BHQ1
WRKY53	Os05g27730	AACGGCTGCTCCATGAAGAA	TTGTGTGCGCCCTTGTAGAC	CTCGCCGACGGCCGCATC-BHQ1
WRKY13	Os01g54600	GCGCAAGTACGGCCAGAA	CCTTGGAGCTACTGCACCTGTA	CCCATCAAGGGCTCTCCCTACCCA-BHQ1
OsACS2	Os04g48850	CACCCCGAGGCATCCAT	ATTGGCGATCCTCTTGAACTG	TGCACACCGGAGGGCGTCT-BHQ1
ERF3	Os01g58420	GTTCGCTTTCCTTTCAGAGGATA	GCAGCCTGCTCATAGAAAAAGTAA	CCGTTGCACGTCCAGCAACGC-BHQ3