Supporting Information

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SI Results

Protein Sequence Comparison of Stpk-V with Its Putative Homologues from the Susceptible Wheat Variety and Other Species. The putative protein sequences of Stpk-A, Stpk-B, and Stpk-D were obtained as described in *SI Materials and Methods*. When comparing the protein sequence of Stpk-V with its three wheat orthologous genes, it was found that the sizes of the four sequences were identical, and their homology was greater than 97%. There were seven amino acids specific to Stpk-V that were not located in the conserved kinase domain. The polymorphisms were located in the N terminus and the C terminus, and this could change the structure of the protein (Fig. S3). The relationship of these polymorphisms with the function of the genes will be further investigated by point mutational analysis.

To identify the orthologous genes or homologous genes from other species, the protein sequence of Stpk-V was used as the query to search the National Center for Biotechnology Information protein database, and the nucleotide sequence of *Stpk-V* was used to search the genomic sequence of the model species. The orthologous genes in *Brachypodium distachyon* (BRADI3G04940), *Oryza sativa* (LOC_Os02g06930), *Zea mays* (GRMZM2G169020), *Sorghum bicolor* (EEF38111.1), *Arabidopsis thaliana* (AT1G16670), *Populus trichocarpa* (EEF12427.1), and *Vitis vinifera* (CBI21796.3), and the homologue in *Ricinus communis* (EEF38111.1), were identified. Stpk-V shared the highest similarity (79%) with LOC_Os02g06930.2 from rice at the protein level. The multiple sequence analysis also showed that the N and C termini are more diverse, whereas the middle part is relatively conserved. It is interesting that the orthologous genes of *Stpk-V* have alternative splicing forms in *B. distachyon, O. sativa*, and *Z. mays* (one splicing form contains the FPEVTNGVLLLQ sequence and the other lacks it). However, in dicotyledons such as *A. thaliana, P. trichocarpa*, *V. vinifera*, and *R. communis*, all homologues lack the FPEVTNGVLLLQ sequence. Stpk-V contains the FPEVTNGVLLLQ sequence, and we did not identify its alternative splicing forms in *H. villosa* and T6VS·6AL (Fig. S4*A*). The phylogenetic analysis of the 12 sequences demonstrated that Stpk-V is closer to the sequences from the *Poaceae* groups (Fig. S4*B*).

Comparison of the Putative Promoter of the Stpk-V and Its Orthologous Genes in the Susceptible Yangmai158 Line. There were polymorphisms between *Stpk-V* and its orthologous genes at the promoter region: 35 specific SNPs and one 78-bp insertion in *pStpk-V* (Fig. S5). In *pStpk-D* and *pStpk-A*(*B*), the number 1, 9, 11, 14, and 15 SNPs resulted in the change of binding sites of the W-box protein, MYB-like protein, and homeobox protein. The *pStpk-D* and *pStpk-A*(*B*) also lack one homeobox protein binding site in the missing 78-bp region. The effect of the polymorphisms in the putative promoter region on the expression of the genes remains to be studied in the future.

SI Materials and Methods

GeneChip Microarray. The Barley1 GeneChip (part no. 900515; Affymetrix) (1) was chosen to screen the *Bgt* induced genes in *H. villosa* because barley is a relative of *H. villosa*. GeneChip microarray experiments were conducted in the National Engineering Center for Biochip in Shanghai, China, according to the Affymetrix technical manual. After hybridization, the GeneChips were scanned in the Agilent 2500A GeneArray scanner, and Affymetrix MAS5.0 software was used to conduct the image analysis. The User-Defined parameter was selected under the Normalization menu, and the Normalization Value was set as 1. All Probe Sets was selected under the Scaling menu, and the Target Signal was set as 500. Only when the signal ratio value was more than 2 was the corresponding probe regarded as upregulated.

Semiquantitative RT-PCR. The *Stpk-V* expression level was analyzed by semiquantitative RT-PCR under different abiotic and biotic stresses in *H. villosa*, T6VS·6AL, and transgenic plants. When the seedings had one fully expanded and one newly developed leaves, the seedlings were treated with different stresses: for *Bgt* inoculation, the leaves were inoculated with a mixture of strains; for heat stress, the seedlings were treated under 40 °C for 1 h; for salt stress, the roots were immersed in 300 mmol/L NaCl; for hormone and H_2O_2 treatments, the leaves were sprayed with 5 mmol/L SA, 1 mmol/L JA, or 7 mmol/L H_2O_2. For *Fusarium graminearum* inoculation, the spikes were inoculated with spores by a single floret injection. RNA was isolated using TRIzol reagent, and first-strand synthesis cDNA was performed on 2 μg of total RNA using a reverse transcription system according to the manufacturer's manual (Invitrogen). The primers used for *Stpk-V* were designed according to the probe Contig17515 (*CINAU15* forward, 5′-AGATCCAACACCAGTTCAAG-3′ and *CINAU15* reverse, 5′-ATGTTAAGGAGGCTTGTGTC-3′), and the primers for the housekeeping gene *Tubulin* were (forward) 5′-AGAACACTGTTGTAAGGCTCAAC-3′ and (reverse) 5′-GAGCTTTACTGCCTCGAACATGG-3′. Amplification was performed at 94 °C for 3 min; 27 cycles of 94 °C for 30 s, 55 to 58 °C for 30 s, and 72 °C for 2 min; followed by 10 min at 72 °C. PCR products were electrophoresed on 1% agarose gels.

RACE. RACE was conducted to obtain the full-length cDNA of *Stpk-V* from the cDNA of *H. villosa* inoculated for 24 h. 3′-RACE was conducted using the 3′-RACE System for Rapid Amplification of cDNA Ends kit (Invitrogen). *Stpk-V*-3′ (5′-GCAGGGAGATTTGGTGAAA-3′) was used to perform the first-round PCR and *Stpk-V*-3′-nest (5′-CCTGCAGGTTCCTGAAAGTTGG-3′) was used to conduct the nest PCR. 5′-RACE was conducted using the 5′-Full RACE kit (Takara). *Stpk-V*-5′P (5′-CTGCAACCAACTTCAGGTCA-3′; the first C was phosphorylated) was used as the primer to transcribe RNA. The primers *Stpk-V*-5′F (5′-TAGCAGGCGAAAAGGATGTT-3′) and *Stpk-V*-5′R (5′-ACATGGTGGGTCGATGTCTT-3′) were used to perform the first-round PCR, and *Stpk-V*-5′F-nest (5′-CTCGGAGAAGATCAGCAAGC-3′) and *Stpk-V*-5′R -nest (5′-ATGGTGGGTCGATGTCTTGT-3′) were used to conduct the nest PCR.

FISH and Sequential GISH. FISH using 30-kb TAC as the probe in del.6VS-1 and T6VS·6AL was conducted as described (2). The hybridization solution contained 50% deionized formamide, 2× SSC, 10% dextran sulfate, 100 ng/μL of sheared salmon testis DNA, 1 ng/μL of labeled TAC probe, and 300 ng/μL sheared blocking genomic DNA of Chinese Spring and *H. villosa*. Twenty microliters of the denatured hybridization mixture was applied to each slide and hybridization was performed in a humid chamber at 37 °C overnight. Posthybridization washes were in 2× SSC and 1× PBS solution at room temperature for 10 min each. Digoxigenin-labeled probe was detected with FITC-conjugated antidigoxigenin antibody. Chromosomes were counterstained with DAPI (Vector Laboratories). After TAC-FISH, the hybridization signals were removed for the following GISH, which was performed using gDNA of *H. villosa* as the probe. Chromosome

and signal images were captured using a CCD camera attached to an epifluorescence microscope (BX61; Olympus). The gray-scale images were then separately pseudocolored and merged by using IPLab Spectrum version 3.1 software. Fraction lengths of hybridization sites were calculated as the distance from the centromere to the hybridization signal relative to the total length of the chromosome arm. More than 10 chromosomes were measured for each FL calculation by using IPLab Spectrum version 3.1 software.

Vector Construction and Stpk-V Subcellular Localization. Gateway technology was used to construct the vector for Stpk-V subcellular localization according to the manufacturer's protocol (Invitrogen) The *Stpk-V* gene was amplified using the *att*B site containing the following primers: forward, 5′-GGGGACAAGTTTGTACAA-AAAAGCAGGCTTCATGGGTTGTTCTCCTTTCTTT-3′; and reverse, 5′-GGGGACCACTTTGTACAAGAAAGCTGGGTC-CTCACACTCTGATATTGCGGT-3′. A BP recombination reaction between the *att*B site flanking *Stpk-V* PCR fragment and an *att*P-containing donor vector pDonor201.1 was conducted to generate an entry vector pDNOR201.1-*Stpk-V*. Subsequently, an LR recombination reaction between the *att*L-containing entry clone pDNOR201.1-*Stpk-V* and an *att*R-containing destination vector pUbi-GW-C-term*YFP* was conducted to generate an expression vector pUbi-*Stpk-V-YFP*. Thus, the C terminus of the Stpk-V was fused with the YFP protein. The destination vector pUbi-GW-C-term*YFP* was provided by Prof. Qianhua Shen (Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Beijing, China). The single-cell transient expression assay was performed following the instructions of the PDS 1000/He Particle Delivery System (Bio-Rad) as described earlier. After bombardment, the leaf was keep in the dark and at room temperature for at least 36 h, and then was observed under the a confocal microscope (TCS-SP2; Leica). The Stpk-V subcellular localization was determined based on the distribution of the YFP signal.

Vector Construction and Wheat Transformation. The ORF fragment of the *Stpk-V* was amplified using the forward *Sma*I-containing primer (5′-TCCCCCGGGATGGGTTGTTCTCCTTTCTT-3′) and the reverse *Sac*I-containing primer (5′-ACCGAGCTCTCACT-CACACTCTGATATTG -3′). The recombinant vector pAHC: *Stpk-V* was constructed using the pAHC 25 vector as the start vector (3). pAHC 25 was cut with *Sma*I and *Sac*I enzyme to remove the GUS gene, and then the PCR product of *Stpk-V* cut with the same two enzymes was inserted to pAHC 25. Therefore, the recombinant pAHC:*Stpk-V* was constructed by replacing the *GUS* gene with the ORF of *Stpk-V*. In the pAHC:*Stpk-V*, the *Stpk-V* gene was placed under the control of the ubiquitin (*Ubi*) promoter and was followed by the NOS terminator sequence. The herbicide tolerance gene (*bar*) was used as a selectable marker gene. pAHC: *Stpk-V* transgenic wheat plants were produced by particle bombardment of calli cultured from immature embryos of susceptible variety Yangmai158 as described (4). When screening the positive transgenic genes, Southern blotting analysis, BASTA resistance evaluation, and target genes detection were conducted in different generations. The procedure of Southern blotting procedure was according to the method by Qi et al. (5) using the *bar* gene as the probe in the T_0 generation plant. Evaluation of BASTA resistance was conducted in T_1 generation plants. The genes, including the *bar* gene, *Ubi*-promoter gene, and target gene *Stpk-V*, were detected in T_0 to T_3 generation plants. The primers for the genes were as follows: *bar*, 5′-CGAGACAAGCACGGTCAACTTC-3′ and 5′-AAACCCACGTCATGCCAGTTC-3′; *Ubi*-promoter, 5′-CCAC-ATCATCACAACCAAGC-3′ and 5′-ACCCAGATCTCCCCCA-AATC-3′; and *Stpk-V*, 5′-ATTTCTGATTTCGGTTTAGC-3′ and 5′-ACTTCCGTGTCACTTGTCC-3′.

Functional Analysis of *Stpk-V* by VIGS. A 280-bp fragment amplified with primers (forward, 5′-TAAGCTAGCGGCGGCTAAGGA-ATGGA-3′; reverse, 5′-CTAGCTAGCCCTCGGGCGATACC-AA-3′) was reversely inserted into the RNAγgammab to form the recombinant vector RNAγgammab:*Stpk-V*as for gene silencing. The 280-bp fragment used for VIGS was from 188 to 467 nt in the ORF, and the corresponding amino acid sequence was from 63 to 155 aa. The second fully expanded leaves of the seedlings were infected with virus BSMV:*Stpk-V*as (the γ strand was RNAγgammab:*Stpk-V*as), using BSMV:*Ta-PDS*as (the γ strand was RNAγgammab:*Ta-PDS*as) and BSMV:*GFP* as the control. The fifth leaves were used to evaluate the resistance to *Bgt* and to check the expression level of *Stpk-V* by real-time PCR. Real-time RT-PCR was operated on a iCycleriQ MultiColor Real Time PCR Detection System (BioRad) using SYBRGreen dye. Three independent RT-PCR runs were performed. To ensure only endogenous gene transcripts and not those from recombinant virus were amplified, primers (5′-ATTTCTGATTTCGG-TTTAGC-3′ and 5′-ACTTCCGTGTCACTTGTCC-3′) for real-time RT-PCR were designed from the region that is not present in the fragments used for eliciting silencing.

Isolation of *Stpk-A*, *Stpk-B*, and *Stpk-D* from Yangmai158. Three orthologous genes of *Stpk-V*, *Stpk-A* (JF439306), *Stpk-B* (JF439308), and *Stpk-D* (JF439307), were isolated from the A, B, and D genomes by PCR using the DNA of susceptible Yangmai158 as template and the primers covering the whole ORF of *Stpk-V* (5′-TATGATGGCAGCTGTTTAAG-3′ and 5′-TATT-TGCAACCAACTTCAGG-3′). Amplification was performed at 94 °C for 3 min; 30 cycles of 94 °C for 30 s, 55 °C for 30 s, and 72 °C for 3 min; followed by 10 min at 72 °C. PCR products were extracted from the agarose and then inserted into the pGEM-T Easy vector (Promega). The plasmids DNA of the positive clones were used as templates individually in the next round of PCR, and this time the primers *CINAU15-F and CINAU15-R* were used. The *Stpk-A*, *Stpk-B*, and *Stpk-D* were discriminated based on the amplified fragment size produced from each clone in the second round of PCR. The corresponding cDNA sequences of the three genes were not obtained by RT-PCR. The putative protein sequences of three wheat genes were obtained by matching the cDNA sequence of *Stpk-V* to the *gDNA* sequences of *Stpk-A*, *Stpk-B* to *Stpk-D*, and then the putative exons in the three sequences were spliced. The sequence alignment was conducted by DNAMAN software (Lynnon BioSoft, Quebec City, Canada).

Isolation of the Putative Promoter from *Stpk-V* and Its Orthologous Genes in Yangmai158. The putative promoter was isolated by PCR using the *Stpk-V*–containing subclone and the DNA of Yangmai158 as the template, respectively. The primers were 5′-AG-AGACTGAAATCGGCTTCG-3′ and 5′-CAGCTGCCATCAT-ATCCTGA-3′, which could amplify the 1,202-bp sequence upstream of the transcription start site of the *Stpk-V*. The amplified sequence from *Stpk-V* was designated as *pStpk-V*. Two types of sequences were isolated from Yangmai158: one was designated as *pStpk-D* (JF439310) and the other one was named as *pStpk-A*(*B*) (JF439309), which was proposed to be from the A or the B genome. The sequence alignment was conducted by DNAMAN software. The *cis*-elements in the putative promoter was analyzed online by MatInspector software (http://www.genomatix.de).

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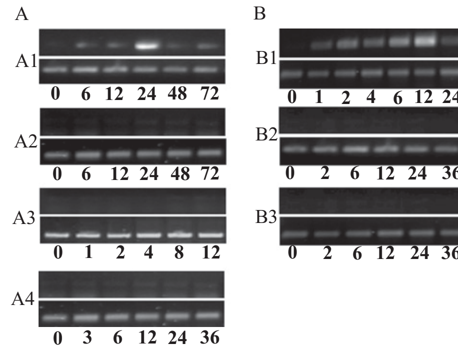

Fig. S1. RT-PCR analysis of the *Stpk-V* expression pattern in resistant T6VS·6AL. (*A*) *Stpk-V* expression pattern in response to biotic or abiotic stresses. *Stpk-V* was induced by *Bgt* inoculation (*1*), but not by *F. graminearum* inoculation (*2*) or heat and salt treatment (*3* and *4*). (*B*) *Stpk-V* expression pattern in T6VS·6AL by treatment with H_2O_2 (*1*), SA (*2*), and JA (*3*). *Stpk-V* was induced only by H_2O_2. *Tubulin* was used as the control. Numbers under each figure indicate hours after stress treatment.

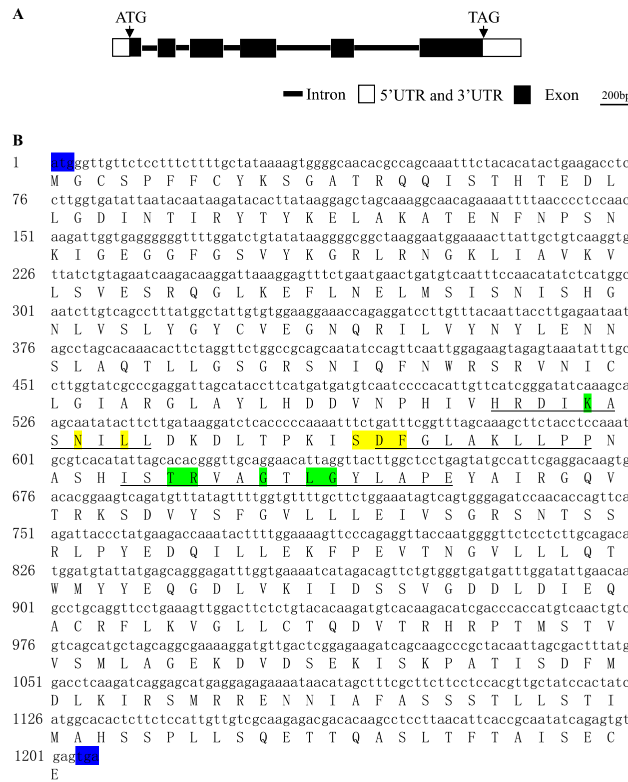

Fig. S2. The structure and the ORF sequence of *Stpk-V.* (*A*) The gene has six exons and five introns. (*B*) The nucleotide and the putative amino acid sequence of *Stpk-V*. The "ATG" and "TGA" labeled in blue are the start and stop codons, respectively. The putative catalytic loop and activation loop are underlined. The ATP binding pocket is labeled in yellow and the substrate binding pocket in green.

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Stpk-A   MGCSPFFCYKSGATRQQISTHTEDLPGDINTIRYTYKELARATENFNPSNKIGEGGFGSVYKGRLRNGKLIAVKVLSVESR   81
Stpk-D   MGCSPFFCYKSGATRQQISTHTEDLPGDINTIRYTYRELARATENFNPSNKIGEGGFGSVYKGRPRNGKLIAVKVLSVESR   81
Stpk-B   MGCSPFFCYKSGATRQQISTHTEDLPGDINTIRYTYKELARATENFNPSNKIGEGGFGSVYKGRLRNGKLIAVKVLSVESR   81
Stpk-V   MGCSPFFCYKSGATRQQISTHTEDLLGDINTIRYTYKELAKATENFNPSNKIGEGGFGSVYKGRLRNGKLIAVKVLSVESR   81
                                  *              *

Stpk-A   QGLKEFLNELMSISNISHGNLVSLYGYCVEGNQRILVYNYLENNSLAQTLLGSGRSNIQFNWRSRVNICLGIARGLAYLHD   162
Stpk-D   QGLKEFLNELMSISNISHGNLVSLYGYCVEGNQRILVYNYLENNSLAQTLLGSGRSNIQFNWRSRVNICLGIARGLAYLHD   162
Stpk-B   QGLKEFLNELMSISNISHGNLVSLYGYCVEGNQRILVYNYLENNSLAQTLLGSGRSNIQFNWRSRVNICLGIARGLAYLHD   162
Stpk-V   QGLKEFLNELMSISNISHGNLVSLYGYCVEGNQRILVYNYLENNSLAQTLLGSGRSNIQFNWRSRVNICLGIARGLAYLHD   162

Stpk-A   DVNPHIVHRDIKASNILLDKDLTPKISDFGLAKLLPPNATHISTRVAGTLGYLAPEYAIRGQVTRKSDVYSFGVLLLEIVS   243
Stpk-D   DVNPHIVHRDIKASNILLDKDLTPKISDFGLAKLLPPNASHISTRVAGTLGYLAPEYAIRGQVTRKSDVYSFGVLLLEIVS   243
Stpk-B   DVNPHIVHRDIKASNILLDKDLTPKISDFGLAKLLPPNASHISTRVAGTLGYLAPEYAIRGQVTRKSDVYSFGVLLLEIVS   243
Stpk-V   DVNPHIVHRDIKASNILLDKDLTPKISDFGLAKLLPPNASHISTRVAGTLGYLAPEYAIRGQVTRKSDVYSFGVLLLEIVS   243

Stpk-A   GRSNTSPRLPYEDQILLEKFPEVTNGVLLLQTWMYYEQGDLAKIIDSSAGDDLDIEQACRFLKVGLLCTQDVTRHRPTMST   324
Stpk-D   GRSNTSSRLPYEDQILLEKFPEVTNGVLLLQTWMYYEQGDLAKIIDSSAGDDMDIEQACRFLKVGLLCTQDVTRHRPTMST   324
Stpk-B   GRSNTSSRLPYEDQILLEKFPEVTNGVLLLQTWMYYEQGDLAKIIDSSAGDDLDIEQACRFLKVGLLCTQDVTRHRPTMST   324
Stpk-V   GRSNTSSRLPYEDQILLEKFPEVTNGVLLLQTWMYYEQGDLVKIIDSSVGDDLDIEQACRFLKVGLLCTQDVTRHRPTMST   324
                                                  *      *

Stpk-A   VVSMLTGEKDVDSEKISKPATISDFMDLKIRSMRRENNIAFASSSTLLSTIMAHSSPLLSQETTQASITFTTISERE...   401
Stpk-D   VVSMLTGEKDVDSEKISKPATISDFMDLKIRSMRRENNIAFASSSTLLSTIMAHSSPLLSQETTQASITFTAISERE...   401
Stpk-B   VVSMLTGEKDVDSEKISKPATISDFMDLKIRSMRRENNIAFASSSTLLSTIMAHSSPLLSQETTQASITFTAISERE...   401
Stpk-V   VVSMLAGEKDVDSEKISKPATISDFMDLKIRSMRRENNIAFASSSTLLSTIMAHSSPLLSQETTQASLTFTAISECE...   401
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Fig. S3. The amino acid sequence comparison of the protein encoded by *Stpk-V* with those encoded by its orthologous genes (*Stpk-A*, *Stpk-B*, and *Stpk-D*) from susceptible wheat variety Yangmai158. There are 13 polymorphisms, and seven of them are specific to Stpk-V, which are mostly located in the N and C termini. Asterisks represent polymorphisms specific to Stpk-V.

Fig. S4. Multiple sequence alignment and phylogenetic tree analysis between Stpk-V with 11 proteins from eight other species, including *B. distachyon* (BRADI3G04940.1, BRADI3G04940.2), *O. sativa* (LOC_Os02g06930.1, LOC_Os02g06930.2), *Z. mays* (GRMZM2G169020.1, GRMZM2G169020.2), *S. bicolor* (EEF38111.1), *A. thaliana* (AT1G16670), *P. trichocarpa* (EEF12427.1), and *V. vinifera* (CBI21796.3), and the homologue in the *R. communis* (EEF38111.1). (*A*) The multiple sequence alignment of the 12 sequences. The totally conserved residues are labeled in dark blue, whereas the residues with a lower degree of conservation are labeled in light blue or pink. The alternative splicing sequence FPEVTNGVLLLQ is underlined. (*B*) The phylogenetic tree of the 12 sequences, indicating Stpk-V is closer to the sequences from the *Poaceae* groups.

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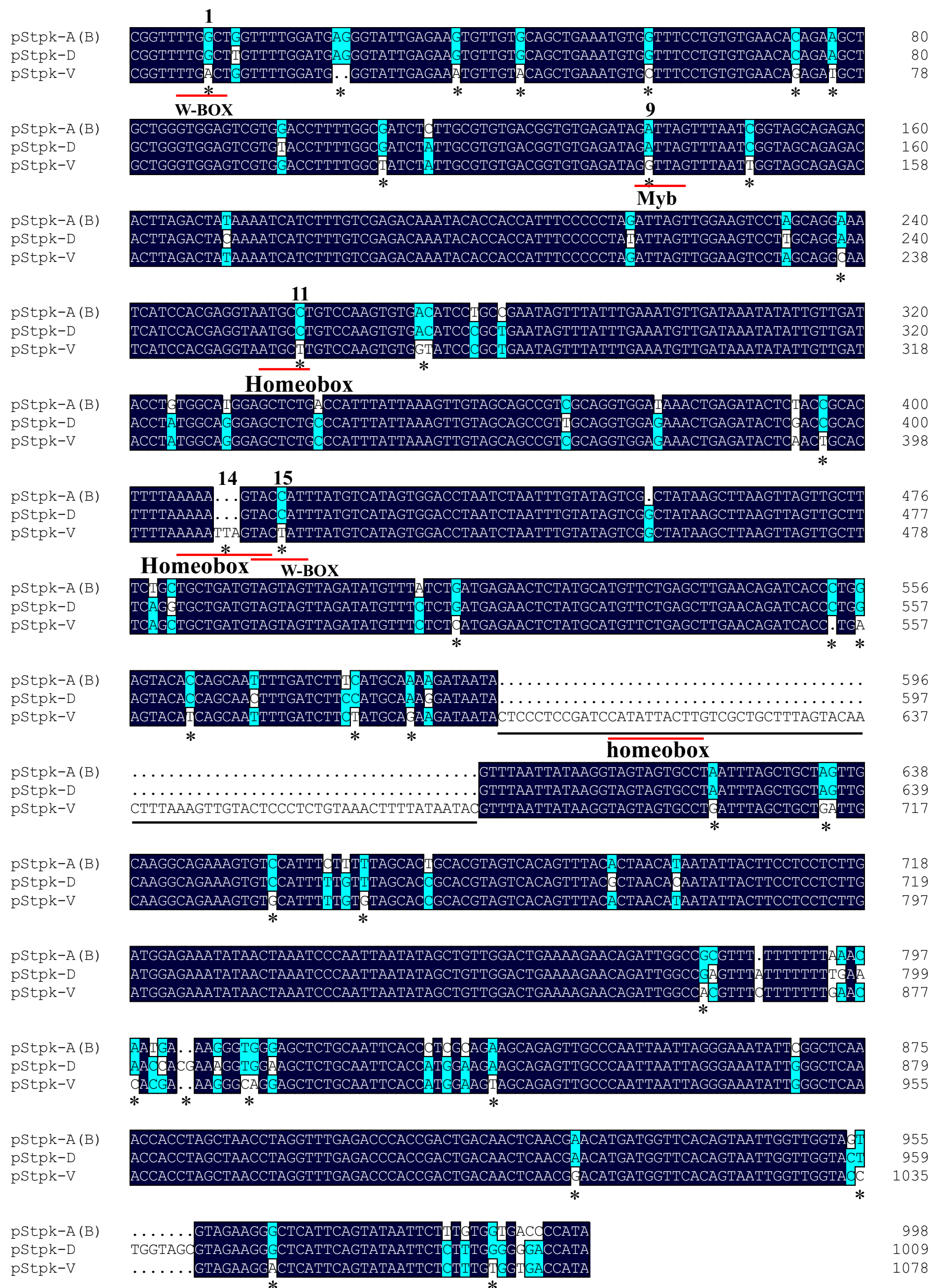

Fig. S5. Comparison of the putative promoter of *Stpk-V* with that of its orthologous genes. There are 35 specific SNPs and one 78-bp insertion in the *pStpk-V* sequence. In the *pStpk-D* and *pStpk-A*(*B*), the numbers 1, 9, 11, 14, and 15 SNPs resulted in the change of binding sites of the W-box protein, MYB-like protein, and the homeobox protein. The *pStpk-D* and *pStpk-A*(*B*) sequence also lack one homeobox protein binding site only found in the 78-bp insertion of *pStpk-V*. The specific SNPs of *pStpk-V* are labeled with asterisks, and the insertion sequences are underlined with a black line. The aforementioned five SNPs are highlighted with the numbers above each site, and the corresponding putative binding transcription factors are placed under the *cis*-elements indicated by the red line.

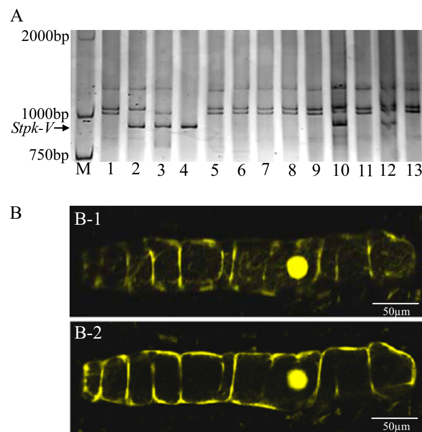

Fig. S6. Mapping and subcellular localization of *Stpk-V*. (*A*) *Stpk-V* mapping by PCR analysis using different cytogenetic stocks. *Stpk-V* was amplified from T6VS·6AL (*2*), *T. durum*–*H. villosa* amphiploid (*3*), *H. villosa* (*4*), DA6V (*10*), and del6VS-1 (*12*), but not from Yangmai158 (*1*), DA1V (*5*), DA2V (*6*), DA3V (*7*), DA4V (*8*), DA5V (*9*), DA7V (*11*) and del6VS-2 (*13*). "M" represents the DL2000 marker. The arrow indicates *Stpk-V*. (*B*) Subcellular localization of Stpk-V by the transient expression assay in the epidermal cell of *H. villosa* with the fusion protein Stpk-V-YFP. B-1 and B-2 are two images scanned at different layers of the same cell. The confocal images showed that Stpk-V is localized in the membrane, cytoplasm and nucleus. (B-1) The Stpk-V-YFP signal was observed mostly in the nucleus and cytoplasm. (B-2) The Stpk-V-YFP signal was observed mostly in the nucleus and membrane. The images were scanned using a confocal microscope (TCS-SP5; Leica). (Scale bar, 50 μm.)

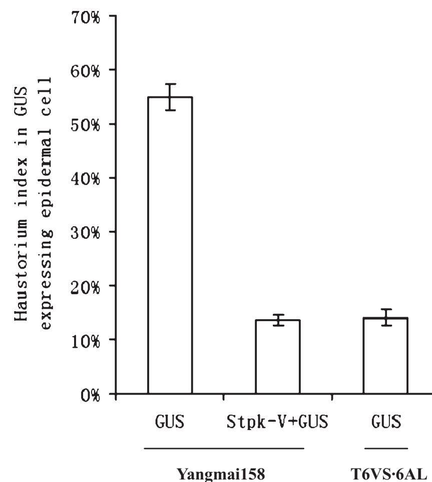

Fig. S7. Functional analysis of *Stpk-V* in the transient expression assay. The graph shows the haustorium index in the *Stpk-V+GUS*–cotransformed cells, in comparison with that in the *GUS*-transformed cells of the susceptible control Yangmai158 and resistant control T6VS·6AL.

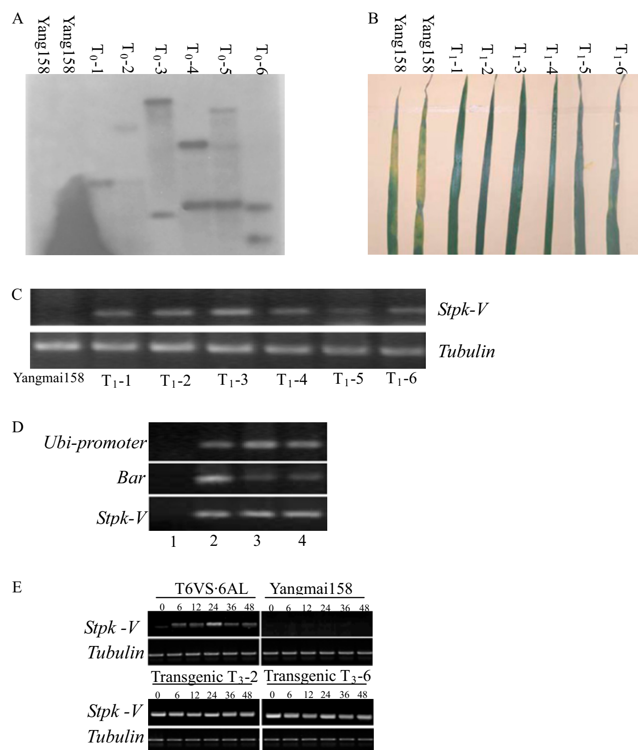

Fig. S8. Validation of positive transformation of plants with pAHC:*Stpk-V* by Southern blotting, BASTA resistance evaluation, gene expression analysis, and detection of transgenes. (*A*) Southern blotting using the *bar* gene as the probe in six positive T_0 plants showed that the gene had been inserted into the genome successfully. (*B*) Positive T_1 plants showed resistance to BASTA. (*C*) *Stpk-V* expressed constitutively in six individual BASTA resistant T_1 plants. (*D*) *Ubi-promoter*, selective marker gene *bar*, and target *Stpk-V* could be amplified from two representative T_3-generation transgenic lines (*3* and *4*) and the positive control pAHC:*Stpk-V* plasmid (*2*), but not from the negative control Yangmai158 (*1*). (*E*) Expression patterns of *Stpk-V* in resistant T6VS·6AL, susceptible Yangmai158, and two T_3 resistant transgenic lines (T_3-2 and T_3-6) after *Bgt* inoculation. *Stpk-V* was induced by *Bgt* in the resistant T6VS·6AL, undetectable in the susceptible Yangmai158, but constitutively expressed in the resistant transgenic plants. *Tubulin* was used as the control. Numbers over each figure indicate hours after *Bgt* inoculation.

Table S1. Resistance grade of homozygous T_3 transgenic plants to different individual strains of *Bgt*

	Resistance grade to *Bgt*			
Strain of *Bgt*	T_3-2	T_3-6	T6VS·6AL	Yangmai158
413	0	0	0	4
611	0	0	0	4
411	0	0	0	4
11	0/0;	0/0;	0/0;	4/3
311	0	0	0	4
711	0	0	0	4
18	0	0	0	4
401	0	0	0	4