FgHAT2 is involved in regulating vegetative growth, conidiation, DNA damage repair, DON production and virulence in *Fusarium graminearum*

Supporting information

Appendix A. Primers used in this study.

Appendix A. PCR primers used in this study.

Primer	Sequence (5'-3')	Relevant characteristics
HPH-F	CTTGCCTGGAGCTAGTGGA	Amplification of HPH gene cassette
HPH-R	TGTTCGGTCGGCATCTACT	
Tri6-RT-F	ATGATTTACATGGAGGACGA	qRT-PCT analysis of *TRI6* expression
Tri6-RT-R	TCAACCCTTGTGTATCCGC	
Tri12-RT-F	TTCCACAGTCATCTTTCCCCA	qRT-PCT analysis of *TRI12* expression
Tri12-RT-R	TCAAGTACGTCCTTATCCGCT	
2AD-F	GCCATGGAGGCCAGTGAATTCATGGACGTCGACATG	Amplification of the full length cDNA sequence of *FgHAT2* for fusion with pGADT7 plasmid
2AD-R	ATGCCCACCCGGGTGGAATTCTCATGTATCTTGCGC	
2BK-F	ATGGCCATGGAGGCCGAATTCATGGACGTCGACATG	Amplification of the full length cDNA sequence of *FgHAT2* for fusion with pGBKT7 plasmid
2BK-R	TCGACGGATCCCCGGGAATTCTCATGTATCTTGCGC	
1AD-F	GCCATGGAGGCCAGTGAATTCATGGAAGACACCACG	Amplification of the full length cDNA sequence of *FgHAT1* for fusion with pGADT7 plasmid
1AD-R	ATGCCCACCCGGGTGGAATTCTCAGGCATCTTCCAC	
1BK-F	ATGGCCATGGAGGCCGAATTCATGGAAGACACCACG	Amplification of the full length cDNA sequence of *FgHAT1* for fusion with pGBKT7 plasmid
1BK-R	TCGACGGATCCCCGGGAATTCTCAGGCATCTTCCAC	
2-UF	GATCTGAAATAGGAGTTGAT	Amplification of upstream fragment of *FgHAT2* for the gene deletion
2-UR	TCCACTAGCTCCAGCCAAGGCCATAATGAATTTTATATG	
2-DF	AGTAGATGCCGACCGAACAATACAGGGAGATTTGAGACC	Amplification of downstream fragment of *FgHAT2* for the gene deletion
2-DR	GACTGGCGTGCTTGAGACTG	
2-NF	TCTTCGTGTACTGGGAGGGT	Amplification of the fragment, including full length ORF of *FgHAT2* and its upstream and downstream sequences
2-NR	GAGAGTTTAAGTCTTTTGTG	
2YW-F	ATGGCAAGTCTGGTGATGTC	For identification of *FgHAT2* gene deletion mutants (presence or not)
2YW-R	GACACGGAACCAATAAAGTT	
2DX-F	ATCTTCCCACTTCCCCTGTG	For identification of *FgHAT2* gene deletion mutants (size difference)
2DX-R	ACCACATCAAACAATAGAGG	

1-UF	GTATCAATCCCCAGCAGTAT	Amplification of upstream fragment of *FgHAT1* for the gene deletion
1-UR	TCCACTAGCTCCAGCCAAGAAACTCGCGATGCGCTCCGT	
1-DF	AGTAGATGCCGACCGAACAATCAATGTTTTTAATTACGC	Amplification of downstream fragment of *FgHAT1* for the gene deletion
1-DR	CAGTGTCTGTGCTGGTATCA	
1-NF	ACTGCGAACTGAGTACATGA	Amplification of the fragment, including full length ORF of *FgHAT1* and its upstream and downstream sequences
1-NR	GTCGTCTACCTGCCAACCCG	
1YW-F	CCGGCACAATCAACGAACTT	For identification of *FgHAT1* gene deletion mutants (presence or not)
1YW-R	GTGGGAGGCTGGAAGTAGTA	
1DX-F	GCCATATCCCTCTTGTTGCT	For identification of *FgHAT1* gene deletion mutants (size difference)
1DX-R	CGAGAAGGTCTGCACATCCG	
2Com-F	GGTACCCGGGATCCTCTAGAGAATATGCGAATAAACATTT	Amplification of fragment including full length ORF of *FgHAT2* and its native promoter and terminator DNA sequences for the complementation
2Com-R	TGCCTGCAGGTCGACTCTAGACAGTAATCGTAAAGTGTTCT	
2-JF	ATTGTGGGTAAGGACGACGG	For identification of *FgHAT2* complemented transformants
2-JR	CATGTGGTCGGGGTAGCG	
1/2-UF	GATCTGAAATAGGAGTTGAT	Amplification of upstream fragment of *FgHAT2* for gene double knockout
1/2-UR	CAAAATAAGCATTGATGTGTTGACCTCCATTTTATATGGCCAAGTTGT	
1/2-DF	CTATCGCCTTCTTGACGAGTTCTTCTGACCTCAGAACTTGCTGTCCTG	Amplification of downstream fragment of *FgHAT2* for gene double knockout
1/2-DR	GACTGGCGTGCTTGAGACTG	
2GFP-F	GACCTCGAGGGGGGGCCCAAGCCCGTCCAAATGTTA	Amplification of the native promoter region and full length ORF of *FgHAT2* for fusion with GFP tag
2GFP-R	TCCTCGCCCTTGCTCACCATTGTATCTTGCGCTAG	
1GFP-F	GACCTCGAGGGGGGGCCCATGACATCCATGCCAACA	Amplification of the native promoter region and full length ORF of *FgHAT1* for fusion with GFP tag
1GFP-R	TCCTCGCCCTTGCTCACCATGGCATCTTCCACTCGAGCCT	
NEO-F	GGAGGTCAACACATCAATGCT	For identification of FgHAT1FgHAT2 gene deletion mutants (presence or not)
NEO-R	TCAGAAGAACTCGTCAAGAAG	

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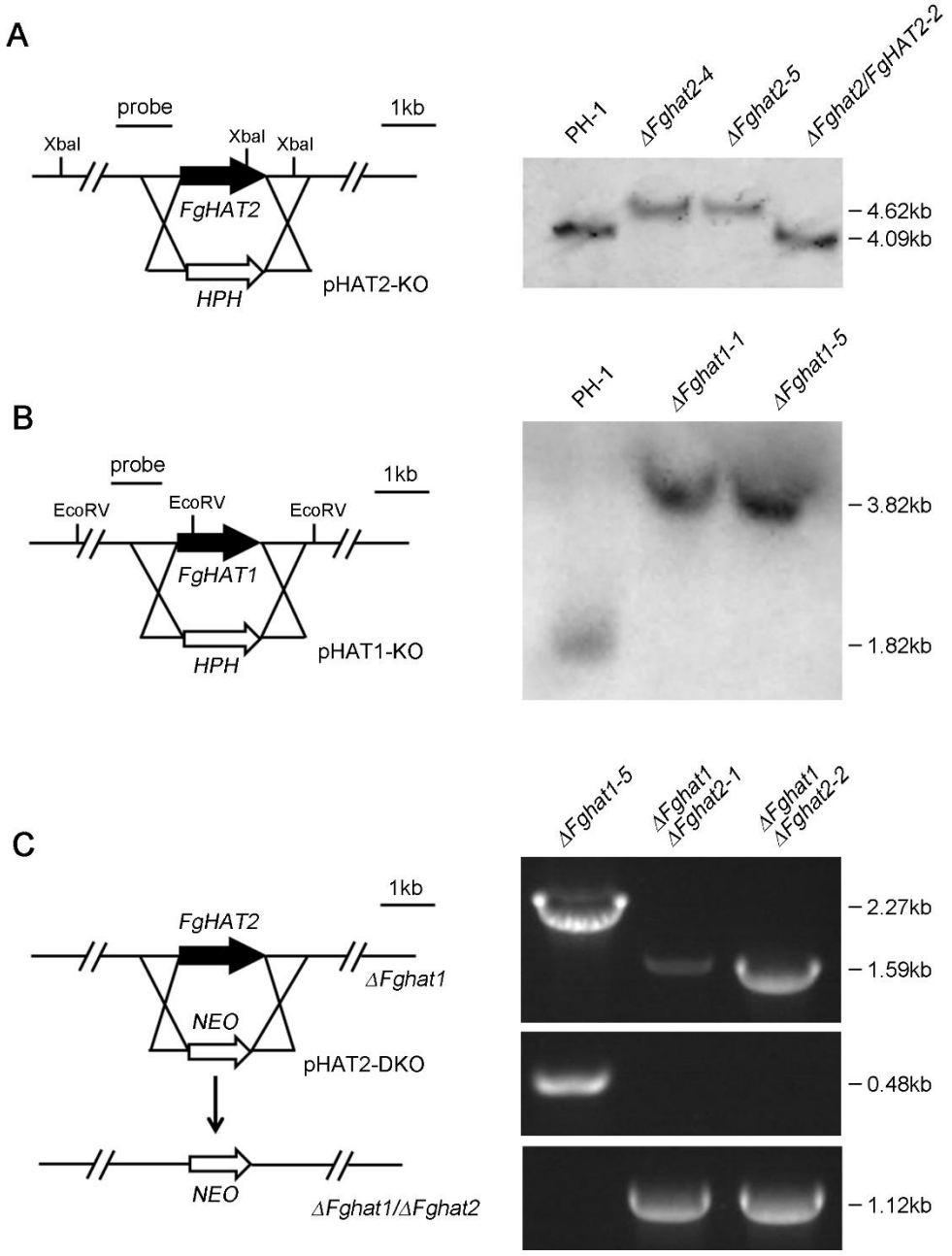
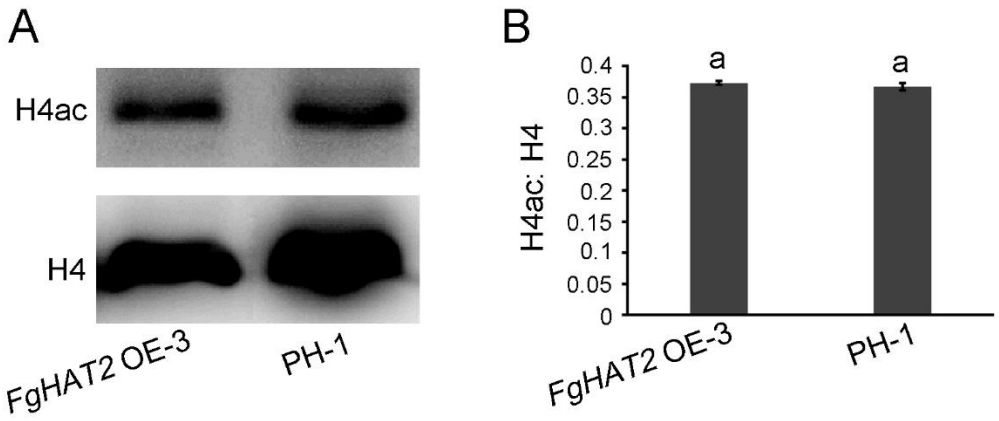
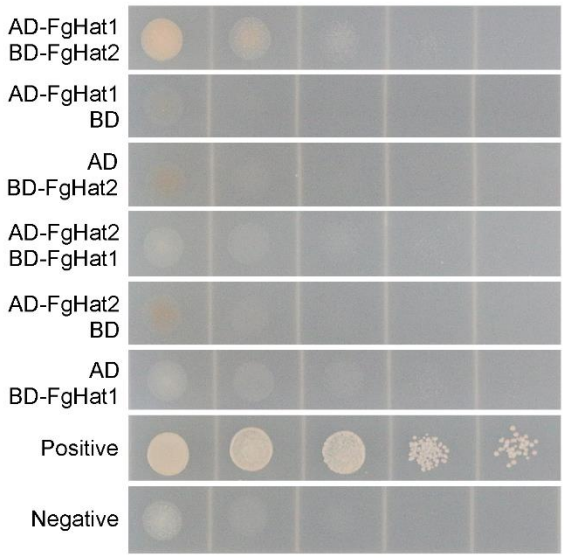


Appendix B. Targeted gene replacement of *FgHAT1* and *FgHAT2*. (A) Strategy for targeted gene deletion of *FgHAT2* (left) and confirmation of *ΔFghat2* mutants by Southern blot analysis (right). Genomic DNA was digested with Xba Ⅰ and probed with a ~1.0 kb fragment amplified with the primers 2-UF/2-UR. **(B)** Strategy for targeted gene deletion of

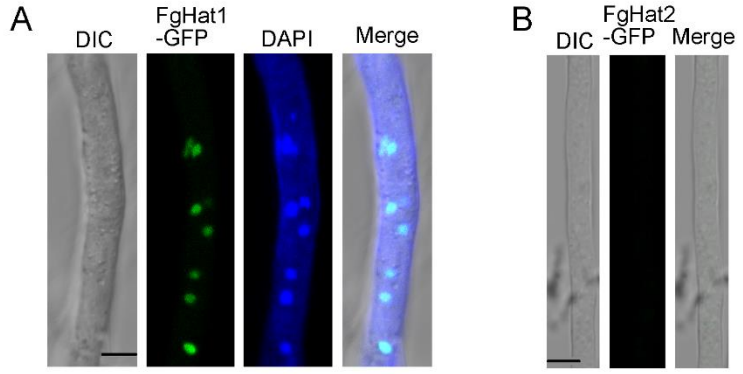
FgHAT1 (left) and confirmation of *ΔFghat1* mutants by Southern blot analysis (right). Genomic DNA was digested with EcoRⅤ and probed with a ~1.0 kb fragment amplified with the primers 1-UF/1-UR. **(C)** Generation of *ΔFghat1ΔFghat2* mutants and confirmation of the double gene mutants by PCR analysis. Top panel, the electrophoresis band pictures of PCR products with primers 2DX-F/2DX-R to confirm *FgHAT1FgHAT2* gene deletion mutants (size difference). Middle panel, the electrophoresis band pictures of PCR products with primers 2YW-F/2YW-R to confirm the presence of *FgHAT2* or not. Bottom panel, the electrophoresis band pictures of PCR products with primers NEO-F/NEO-R to confirm the presence of *NEO* or not.



Appendix C. Overexpression of *FgHAT2* do not affect overall acetylation level of histone H4. (A) Western blot analysis of overall acetylation level histone H4 in PH-1 and the strain overexpressing of *FgHAT2* gene. The antibody H4ac was used for detection of overall acetylation levels of histone H4. Antibody H4 was used as a loading reference. **(B)** Quantification of Western blot signals in (A). The same small letters indicate no significant difference of acetylation level of histone H4.



Appendix D. FgHat1 does not interact with FgHat2. The interactions between FgHat1 and FgHat2 was investigated using yeast two-hybrid analysis. Growth of yeast transformants was determined on SD-Trp-Leu-His-Ade medium. Plasmids pGBKT7-Lam and pGADT7-T were used as a negative control, and pGBKT7-53T and pGADT7-T were used as a positive control.



Appendix E. The subcellular localization of FgHat1 and FgHat2. (A) FgHat1-GFP mainly localizes to the nucleus. Nuclei were stained with DAPI. **(B)** Fluorescence of FgHat2-GFP was unobservable. Bar = 5 μm.