

Supplementary Materials: Ethylenediaminetetraacetic Acid Disodium Salt Acts as an Antifungal Candidate Molecule against *Fusarium graminearum* by Inhibiting DON Biosynthesis and Chitin Synthase Activity

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Table S1. Minimum inhibitory concentrations of EDTANa₂.

Isolates	MIC (mM)
<i>F. acuminatum</i>	18.75
<i>F. asiaticum</i>	9.37
<i>F. avenaceum</i>	9.37
<i>F. concentricum</i>	18.75
<i>F. culmorum</i>	18.75
<i>F. equiseti</i>	9.37
<i>F. fujikuroi</i>	9.37
<i>F. graminearum</i>	18.75
<i>F. lateritium</i> ^a	200
<i>F. oxysporum</i>	18.75
<i>F. proliferatum</i>	9.37
<i>F. solani</i>	18.75
<i>F. verticillioides</i>	18.75

^a EDTANa₂ had no fungicidal activity in the range of 4.69–150 mM. The experiment was replicated three times.

Table S2. Strains used in this study.

Strain	Species	Description
2021	<i>Fusarium asiaticum</i>	Isolated from a field experiencing Fusarium head blight epidemic in Jiangsu Province, China
PH-1	<i>Fusarium graminearum</i>	<i>Fusarium</i> reference strain PH-1
<i>F. acuminatum</i>	<i>Fusarium acuminatum</i>	Isolated from a field experiencing soybean Fusarium Root Rot in Liaoning Province, China
<i>F. avenaceum</i>	<i>Fusarium avenaceum</i>	Isolated from a root of naked barley in Gansu Province, China
<i>F. concentricum</i>	<i>Fusarium concentricum</i>	Provided by Shanghai Academy of Agricultural Sciences
<i>F. culmorum</i>	<i>Fusarium culmorum</i>	Provided by Shanghai Academy of Agricultural Sciences
<i>F. equiseti</i>	<i>Fusarium equiseti</i>	Isolated from alfalfa root rot disease suspected samples in Neimenggu Province, China
<i>F. fujikuroi</i>	<i>Fusarium fujikuroi</i>	Isolated from a field experiencing rice bakanae disease in Jiangsu Province, China

<i>F. lateritium</i>	<i>Fusarium lateritium</i>	Isolated from alfalfa root rot disease suspected samples in Neimenggu Province, China
<i>F. oxysporum</i>	<i>Fusarium oxysporum</i>	Isolated from alfalfa root rot disease suspected samples in Neimenggu Province, China
<i>F. proliferatum</i>	<i>Fusarium proliferatum</i>	Isolated from a field experiencing rice bakanae disease in Jiangsu Province, China
<i>F. solani</i>	<i>Fusarium solani</i>	Isolated from a field experiencing soybean Fusarium Root Rot in Liaoning Province, China
<i>F. verticillioides</i>	<i>Fusarium verticillioides</i>	Provided by Shanghai Academy of Agricultural Sciences

Table S3. Primers used in *TRI* genes qPCR.

Primers	Sequences
FGSG_00071 (<i>Tri1</i>) P1	GAAGATTCCCTGAAGGTCCCG
FGSG_00071 (<i>Tri1</i>) P2	TGTACCAATTCCAATCGCAGAC
FGSG_02343 (<i>Tri12</i>) P1	ATCCCCGCAGTCATTCAA
FGSG_02343 (<i>Tri12</i>) P2	CGCAATCATAGCCACTACACC
FGSG_03532 (<i>Tri8</i>) P1	ATTTACCAAGCTATCCACGACG
FGSG_03532 (<i>Tri8</i>) P2	CACTGAAAAGCCGCCTCAT
FGSG_03534 (<i>Tri3</i>) P1	ACGGTGTCAACGAACGCTT
FGSG_03534 (<i>Tri3</i>) P2	TCCAGTCGGATTGCCAGATA
FGSG_03535 (<i>Tri4</i>) P1	ATGGCCTTGCTGAGATGTACC
FGSG_03535 (<i>Tri4</i>) P2	CCAACAATACGGCGTGAGT
FGSG_03536 (<i>Tri6</i>) P1	ACCGTGCTCGGCATGAGT
FGSG_03536 (<i>Tri6</i>) P2	TCCACCCCTGCTAAAGACCCCT
FGSG_03537 (<i>Tri5</i>) P1	AGCTCACCCAGGAAACCCCT
FGSG_03537 (<i>Tri5</i>) P2	CACTCAATCGTGTCCATCACC
FGSG_03538 (<i>Tri10</i>) P1	TGGGCTCGACAAGACATT
FGSG_03538 (<i>Tri10</i>) P2	CCATCCCTCAACCAAGACA
FGSG_03540 (<i>Tri11</i>) P1	TTCACCCGACCAAACGACT
FGSG_03540 (<i>Tri11</i>) P2	TCTGCGTAGGCAAGGTTCAT
FGSG_07896 (<i>Tri101</i>) P1	CGCCAGCGAACAAAGAGGT
FGSG_07896 (<i>Tri101</i>) P2	AAAGTCGTAATCCCAGAGTCCC
TubulinP1	CGTCCAGAGCAAGAACTCATCA
TubulinP2	TGCGTCGGAACATAGCAGTAA

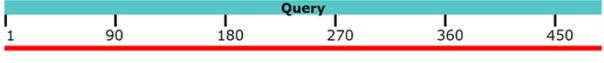
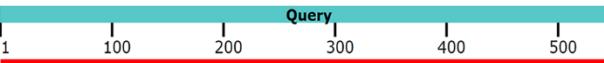
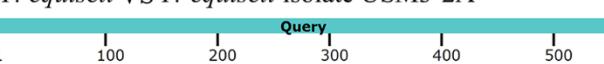
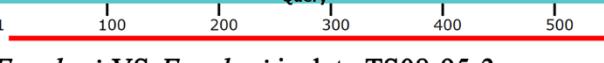
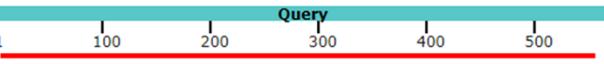
	Query Cover	E value	Per. Identity	Accession
<i>F. acuminatum</i> -VS- <i>F. acuminatum</i> isolate SYQ2	100%	0.0	100.00%	MT649858.1
				
<i>F. avenaceum</i> -VS- <i>F. avenaceum</i> strain ZMXR13	95%	0.0	99.27%	MT446118.1
				
<i>F. concentricum</i> -VS- <i>F. fujikuroi</i> isolate GX12	96%	0.0	99.63%	KJ000433.1
				
<i>F. culmonum</i> -VS- <i>F. culmonum</i> isolate F3-3-19	99%	0.0	98.73%	KX349468.1
				
<i>F. equiseti</i> -VS- <i>F. equiseti</i> isolate USM3-2A	99%	0.0	99.09%	KM111481.1
				
<i>F. fujikuroi</i> -VS- <i>F. fujikuroi</i> isolate GX12	98%	0.0	99.27%	KJ000433.1
				
<i>F. lateritium</i> -VS- <i>F. lateritium</i> strain ZMT01	97%	0.0	99.45%	MT875260.1
				
<i>F. oxysporum</i> -VS- <i>F. oxysporum</i> strain WZ-198	99%	0.0	99.27%	MN856331.1
				
<i>F. proliferatum</i> -VS- <i>F. proliferatum</i> strain BCD	97%	0.0	99.81%	KU939074.1
				
<i>F. solani</i> -VS- <i>F. solani</i> isolate TS08-95-3	95%	0.0	99.09%	AB470848.1
				
<i>F. verticillioides</i> -VS- <i>F. verticillioides</i> isolate Q2	96%	0.0	99.26%	MN698249.1
				

Figure S1. Internal transcribed spacer (ITS) sequences alignment of *Fusarium* isolates used in this study. ITS sequences of different strains were amplified using primers ITS-F/ITS-R (ITS-F: TCCGTAGGTGAAACCTGCGG, ITS-R: TCCTCCGCTTATTGATATGC). Sequences alignment was performed using NCBI Blastn (<https://blast.ncbi.nlm.nih.gov/Blast.cgi>) with default parameters.