Supplementary Material

Deletion of the Histone Deacetylase HdaA in Endophytic Fungus *Penicillium chrysogenum* Fes1701 Induces the Complex Response of Multiple Bioactive Secondary Metabolite Production and Relevant Gene Cluster Expression

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Table S1. Primers used in this study.

Primer Name	Template Gene [a]	Sequence 5'-3'	Function
PchdaA-F	Pc21g14570		*hdaA*
PchdaA-R	Pc21g14570		cloning
PchdaA-TF	Pc21g14570	TGACTGGTCTGACGAGGAGG	*hdaA*
PchdaA-TR	Pc21g14570	GCGACTGCAGAGTCAGTCGG	transcription
Actin F	Actin	CTCTGCCCCACGCTATCTCG	analysis
Actin R	Actin	ACGATGGAAGGACCGCTCTC	
ΔHdaA-P1	Pc21g14570 (upstream)	CCTTCTTCGCTGAGCGTGACCCCGCTGCCATCCCCTGGGG	*hdaA*
ΔHdaA-P2	Pc21g14570 (upstream)	TCAATATCATCTTCTGTCGACATGCAAGGGAAAGCCACGG	deletion
ΔHdaA-P3	BleoR	CCGTGGCTTTCCCTTGCATGTCGACAGAAGATGATATTGA	
ΔHdaA-P4	BleoR	GGAATCAAGAAATCAAGGAAAAGAAGGATTACCTCTAAAC	
ΔHdaA-P5	Pc21g14570 (downstream)	GTTTAGAGGTAATCCTTCTTTTCCTTGATTTCTTGATTCC	
ΔHdaA-P6	Pc21g14570 (downstream)	ATTTTCTCCTTAGTTTGTTAGGTAGATAGTAGAAATGGCC	
ΔHdaA-VP1	Pc21g14570 (upstream)	CACACTAAAAGCTCACCGCC	diagnostic
ΔHdaA-VP2	BleoR	AAGAAGGATTACCTCTAAAC	PCR for
ΔHdaA-VP3	Pc21g14570	GGAAAACCTGGCCTTGATGG	*Δhda*A
ΔHdaA-VP4	Pc21g14570	TGGCCCGTACAAAGGATTGC	
ΔHdaA-VP5	BleoR	TCGACAGAAGATGATATTGA	
ΔHdaA-VP6	Pc21g14570 (downstream)	TATGGTTGGTGATGTGTCAC	
q12570F	Pc21g12570	AGGGAAATGAATCCAGGTGGC	qPCR of the
q12570R	Pc21g12570	TAGATGCCGCTTGTTCGGACC	chrysogine
q12590F	Pc21g12590	TGTGGAGCTCTACGAGGCTG	gene cluster
q12590R	Pc21g12590	GCTGGCAGGGCTCGTCGGTC	
q12600F	Pc21g12600	CGCCGGTGAGACTTTGATCG	
q12600R	Pc21g12600	TAAGCGTCTAATTTTCATCGC	
q12610F	Pc21g12610	TGCATGCAGCTCCATACGAGC	
q12610R	Pc21g12610	ATAGGTGGAAACAGCTCAGAC	
q12620F	Pc21g12620	TTCGCTGGCTAACTGGTCTCG	
q12620R	Pc21g12620	ATGTGGTAGACGAATTGGAGC	
q12630F	Pc21g12630	GAGCCAACTCTGTTGTCTACG	
q12630R	Pc21g12630	AGGGCAATTTGCCTCATTCTG	
q15420F	Pc21g15420	GTGTCGCTGGCCCTCCATTGG	qPCR of the
q15420R	Pc21g15420	GGAGAACACCAGTGAGCACG	roquefortine/
q15430F	Pc21g15430	TGAGATGAGTCCCGGCGAGGC	meleagrin
q15430R	Pc21g15430	TCCGTTGCGATAACCAAGTCC	gene cluster
q15440F	Pc21g15440	CAACAGCGGCCTCAACAACGG	
q15440R	Pc21g15440	CTTACTGGCCATGTGAAGCAG	

q15450F	Pc21g15450	GCTCATCAAAGATGCACTACG	
q15450R	Pc21g15450	GACACATGGTTATGCAGCGAG	
q15460F	Pc21g15460	GCGACATCGTCGGACTGAGAG	
q15460R	Pc21g15460	CAATGGAGTCGCGCCACCTGG	
q15470F	Pc21g15470	CTTGCGGTGGTTACCAGAGTC	
q15470R	Pc21g15470	ACTTTCATCCTCGTACCAACG	
q15480F	Pc21g15480	GGATAGTCTCTTGGTGGATGC	
q15480R	Pc21g15480	GAGAATGTGAACCGTAGCCG	

a. The template genes in strain 1701 were represented with the homologous genes in *P. chrysogenum* Wis 54-1255.

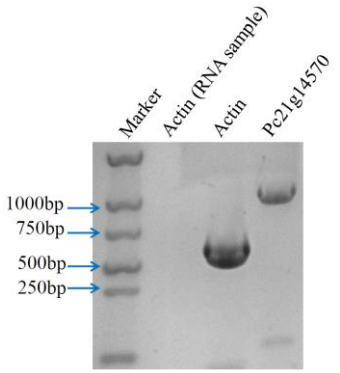

Figure S1 Transcription analysis of potential *hdaA* regulator gene (Pc21g14570) by reverse-transcription PCR. Actin gene of RNA sample and reverse-transcription sample were used as negative and positive controls, respectively.

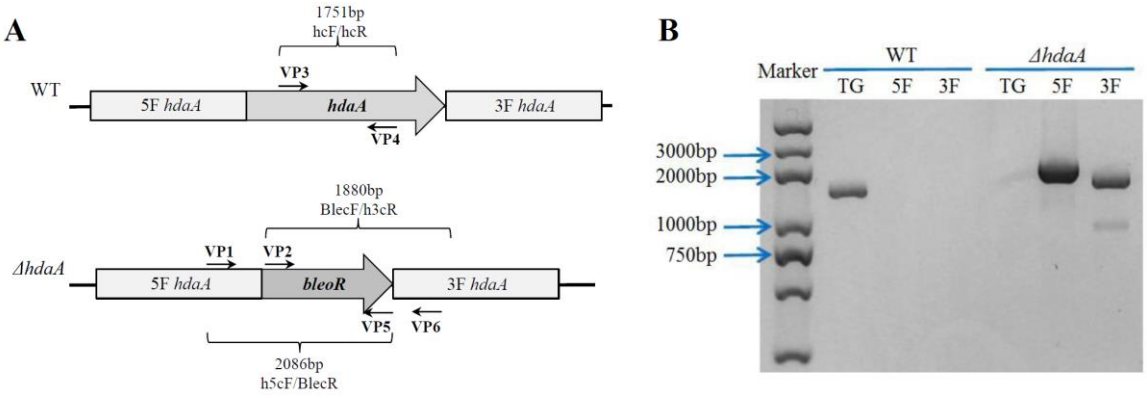

Figure S2 PCR verification of *ΔhdaA* strain. (A) Schematic illustration of diagnostic PCR. Three pairs of primers were used for PCR verification of mutant genotype. The 1.7-kb fragment can only be amplified from the wild-type strain using primers VP3-VP4, but should not appear in *ΔhdaA* mutants. The 2.1-kb and 1.9-kb fragments can be amplified from correct *ΔhdaA* mutants using primers VP1-VP5 and VP2-VP6, respectively, but was absent in the wild-type strain. (B) Genotypic verification of mutant by PCR. Note: Lane TG, using primers VP3-VP4; Lane 5F, using primers VP1-VP5; Lane 3F, using primers VP2-VP6.

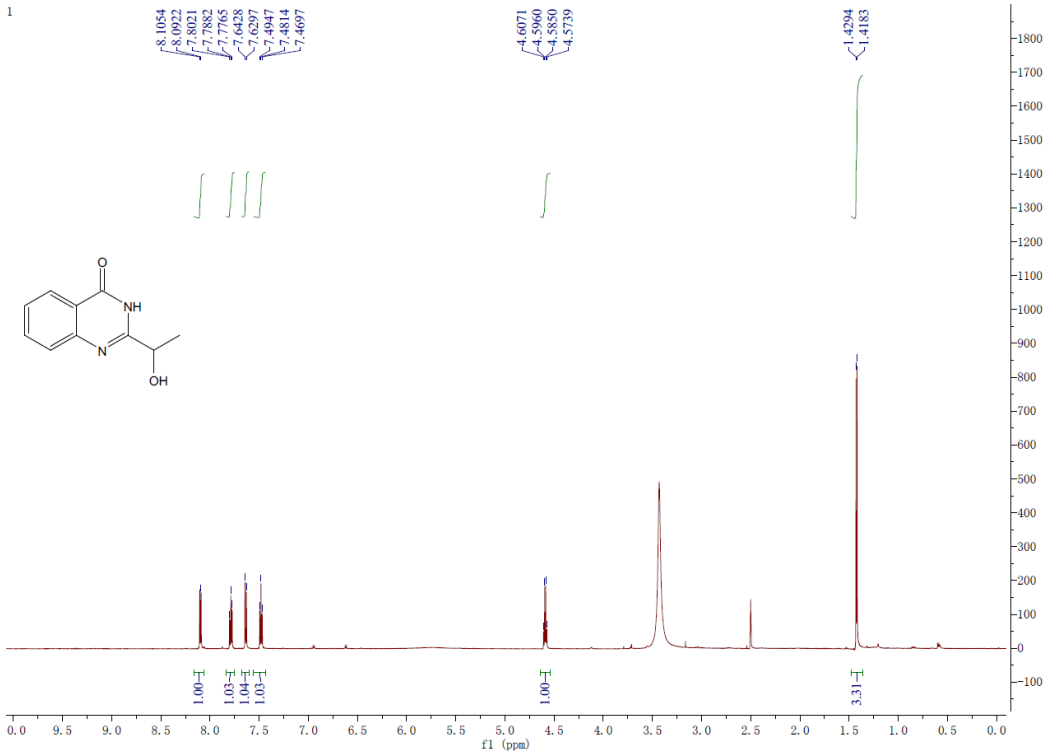

Figure S3 ^{1}H-NMR spectrum of compound **1**.

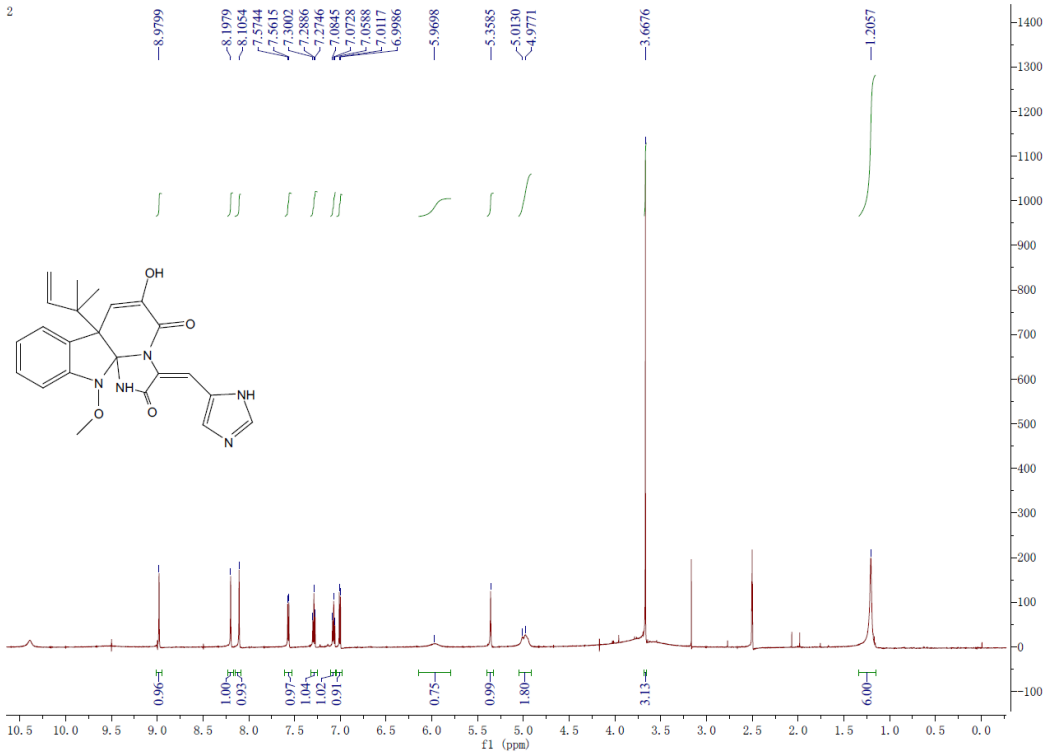

Figure S4 ^{1}H-NMR spectrum of compound **2**.

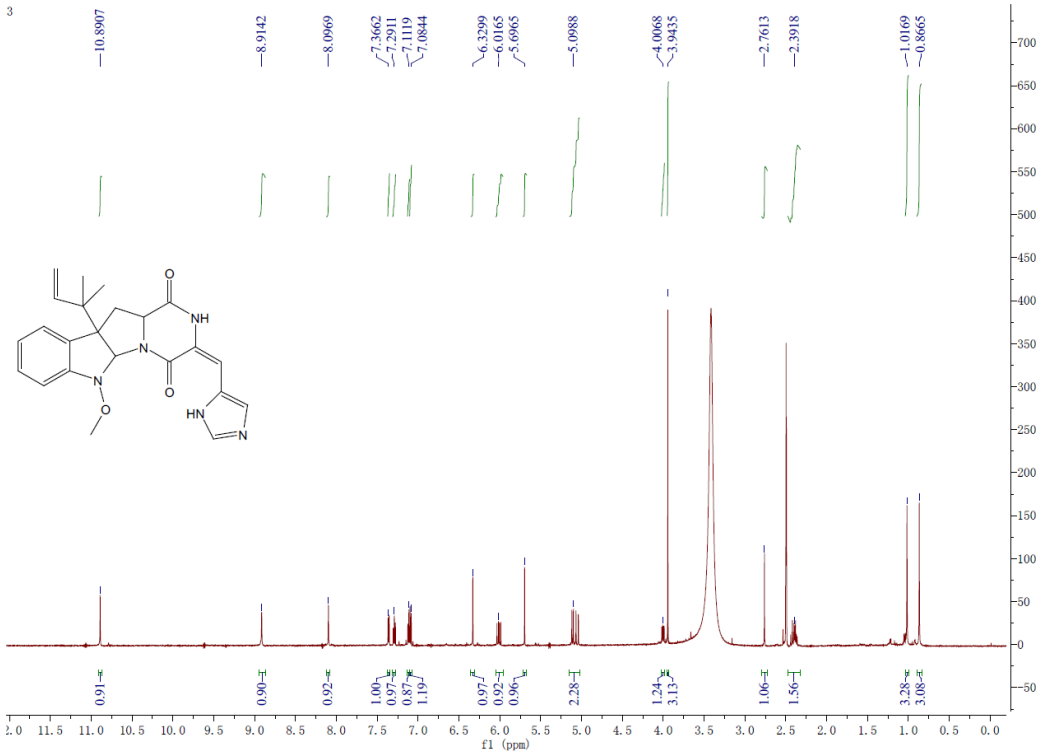

Figure S5 ^{1}H-NMR spectrum of compound **3**.

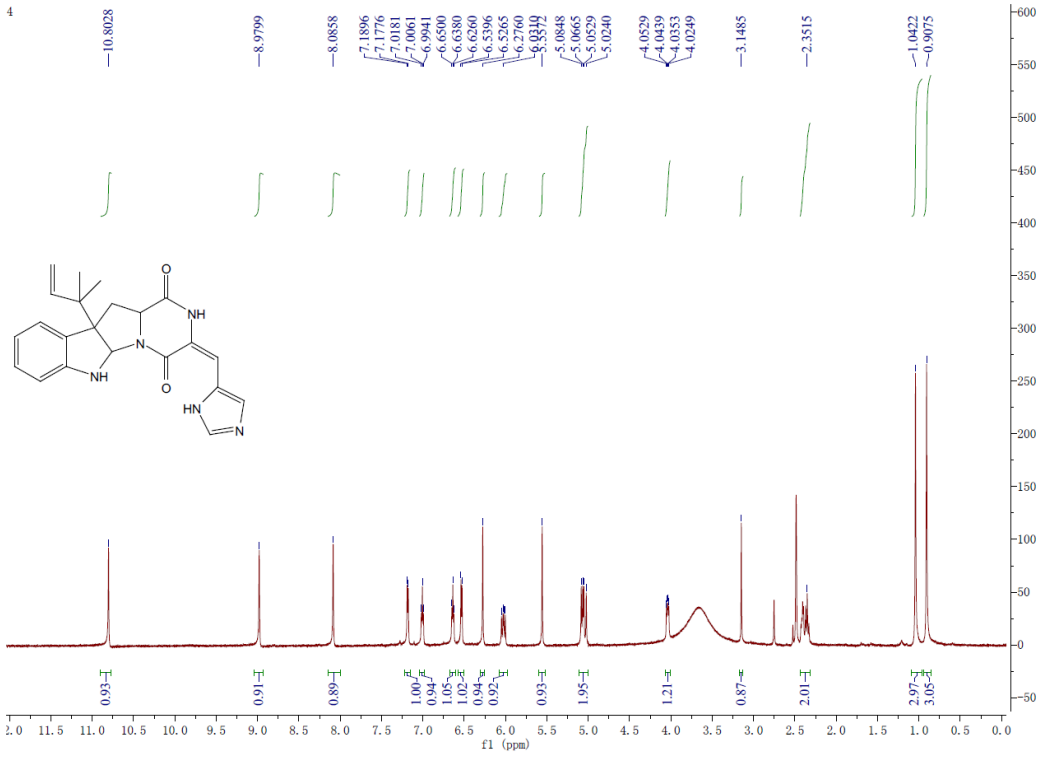

Figure S6 ^{1}H-NMR spectrum of compound **4**.

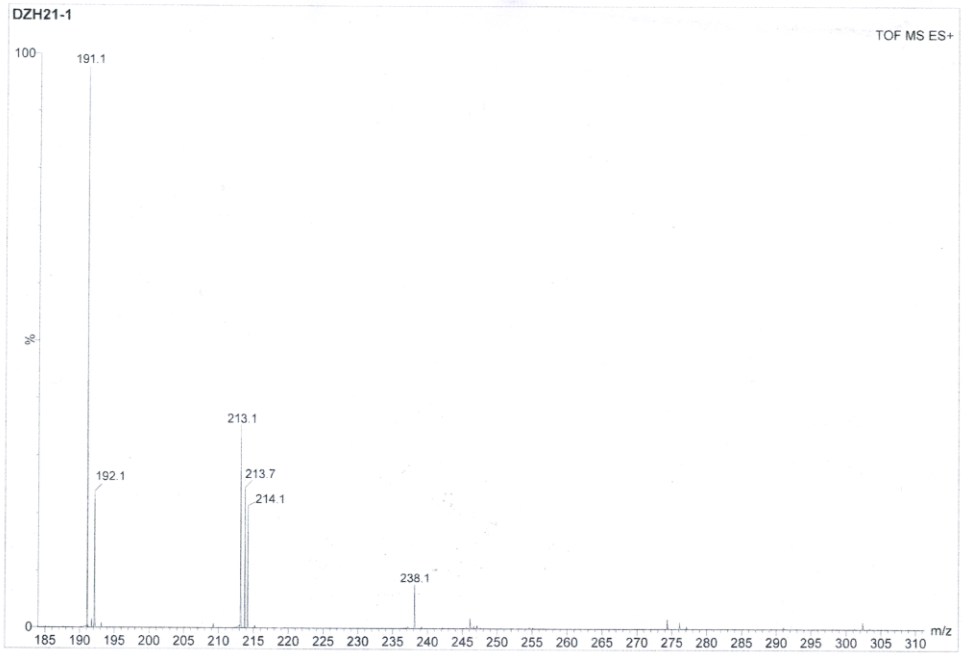

Figure S7 MS spectrum of compound **1**.

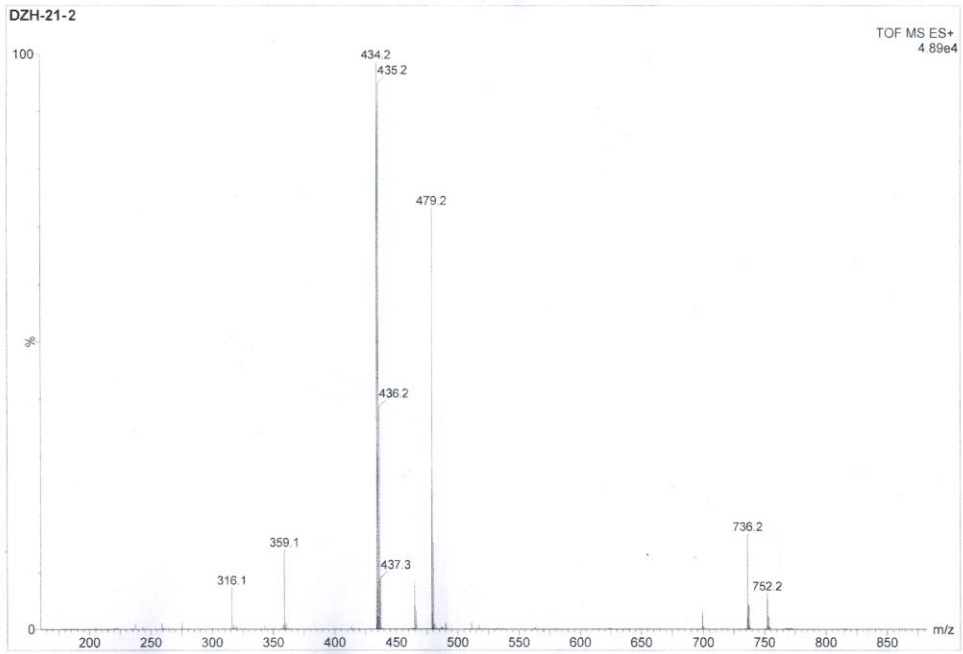

Figure S8 MS spectrum of compound **2**.

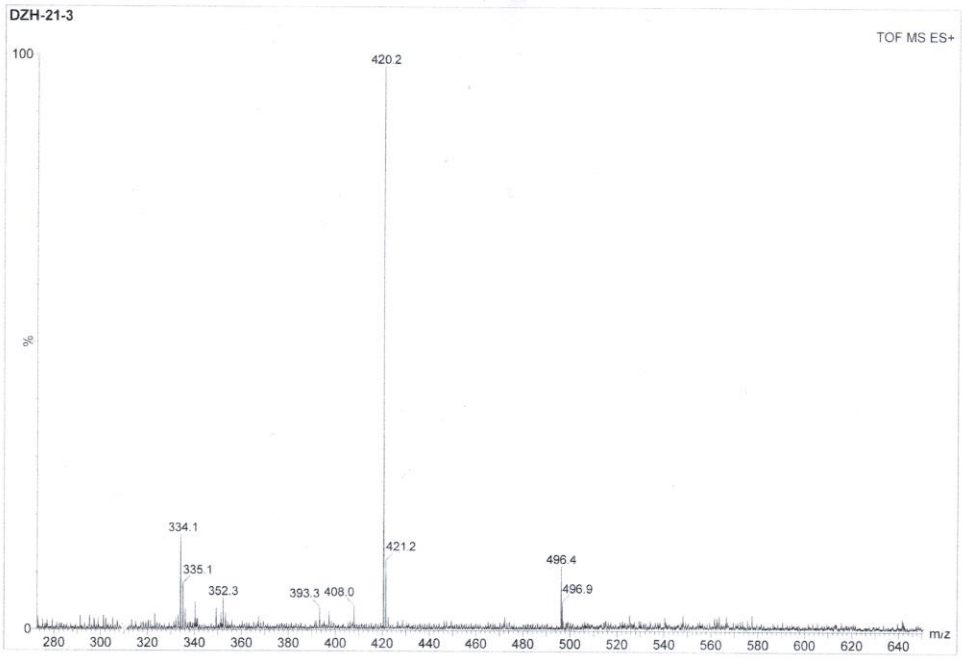

Figure S9 MS spectrum of compound **3**.

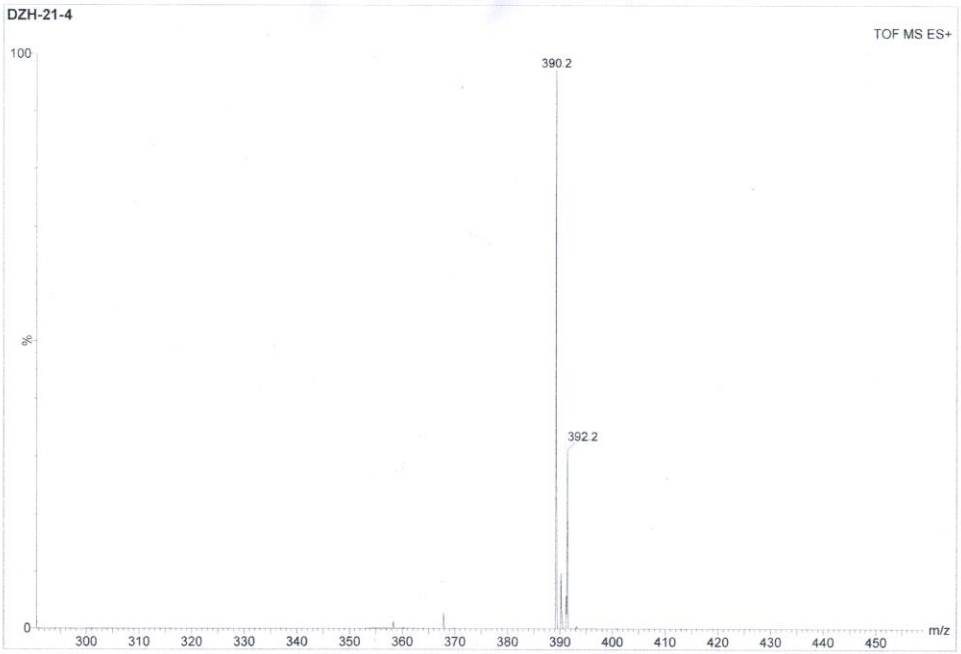

Figure S10 MS spectrum of compound **4**.