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Supplemental information

Unusual (2*R*,6*R*)-bicyclo[3.1.1]heptane ring construction in fungal α-*trans*-bergamotene biosynthesis

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Supplementary Figures

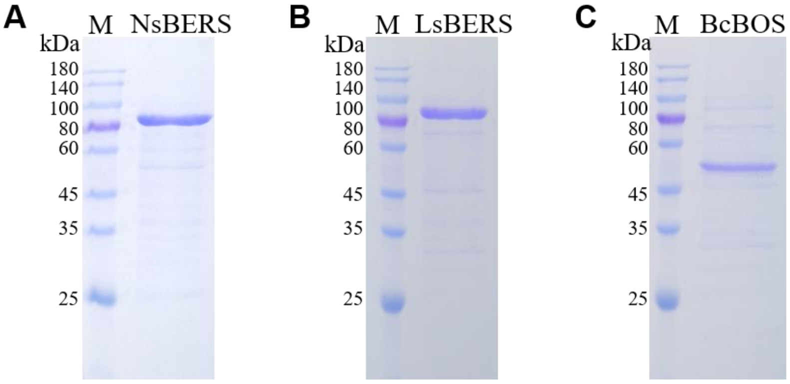


Figure S1. SDS-PAGE of purified STSs by affinity chromatography, related to Figure 2, Figure 4 and Figure S2.

(A) MBP-tagged NsBERS (91 kDa); (B) MBP-tagged LsBERS (92 kDa); (C) BcBOS (53 kDa).

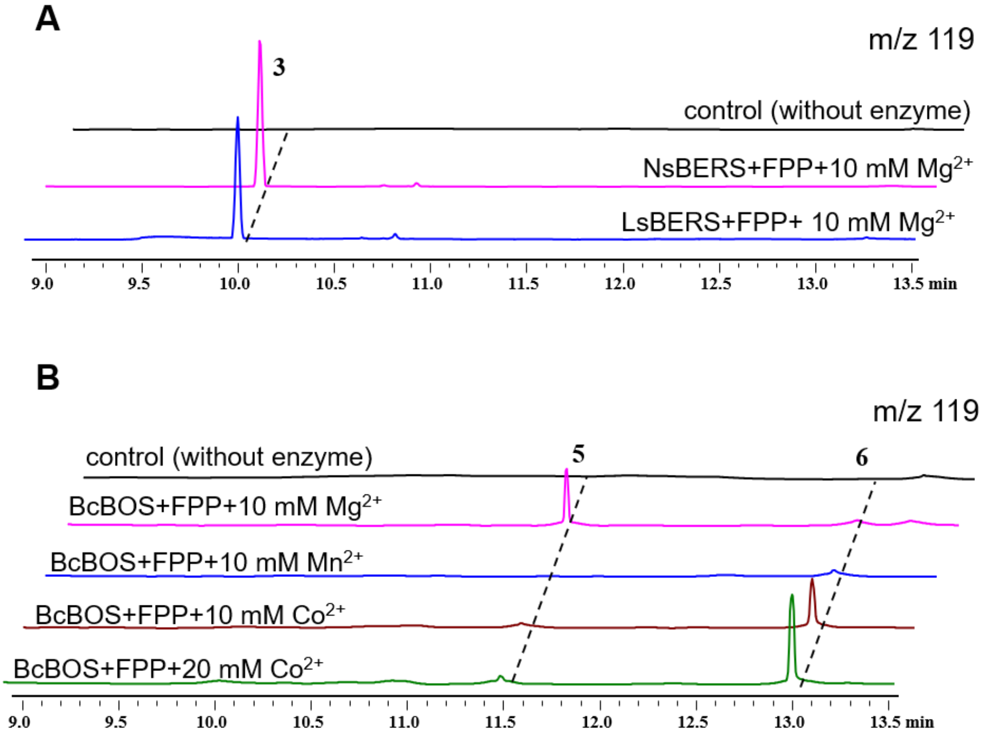


Figure S2. ***In vitro*** **characterization of NsBERS, LsBERS and BcBOS, related to Figure 2, Figure 4 and Figure S1.**

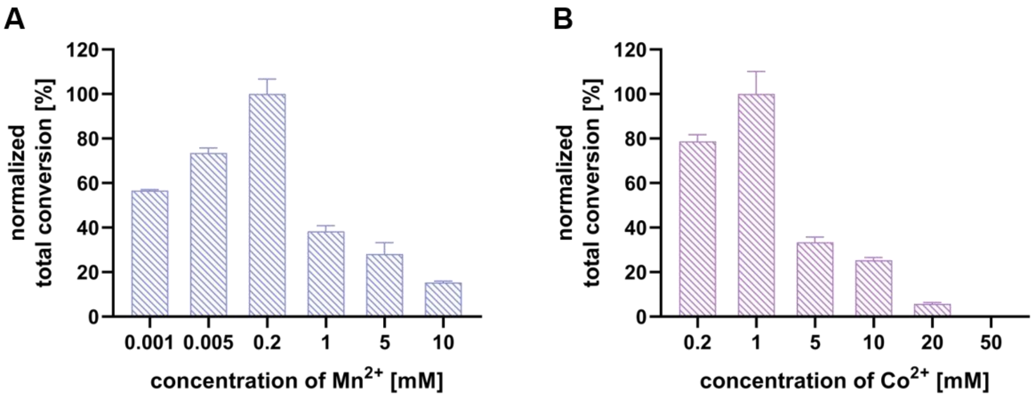


Figure S3. The optimal concentrations of divalent metal ions assay of NsBERS, related to Figure 2, Figure S1 and Figure S2.

The corresponding peak of **3** was integrated in each sample and normalized with the average conversion of optimal concentrations, respectively.

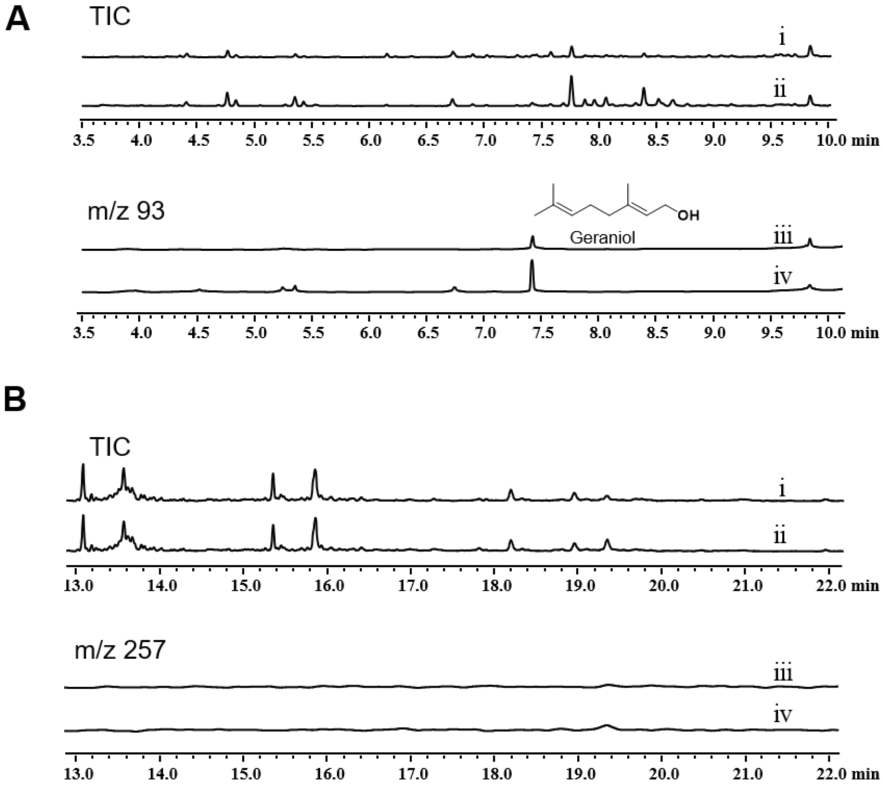
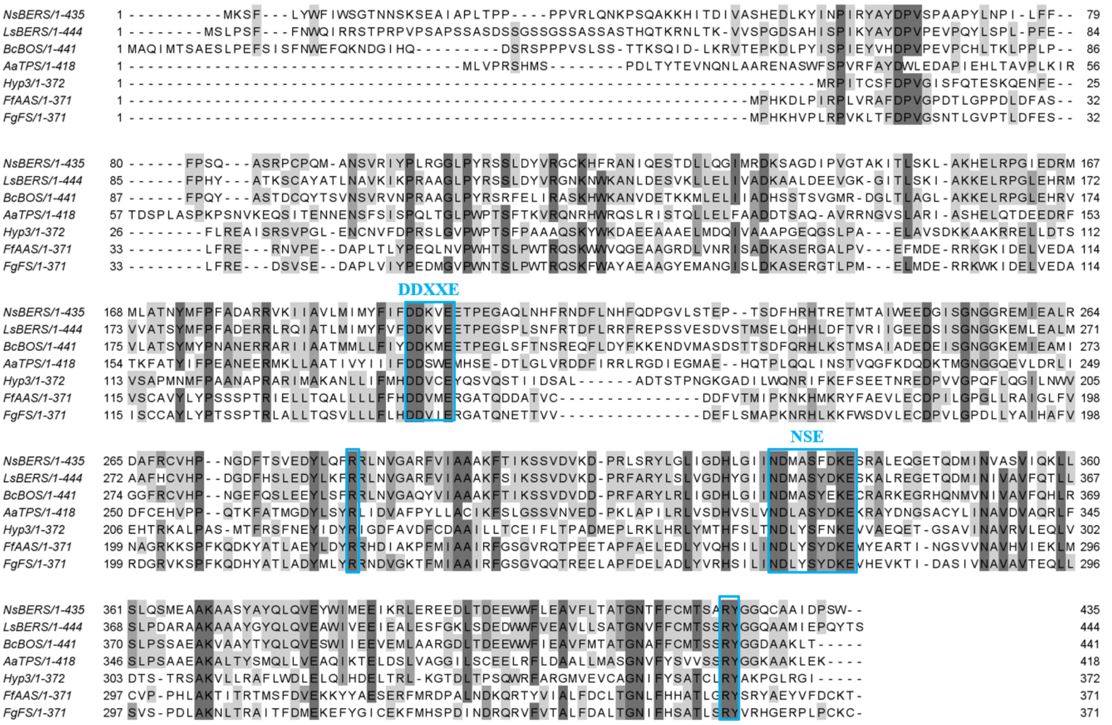


Figure S4. Substrate promiscuity assay of NsBERS, related to Figure 2 and Figure S1.

(A) GC–MS analysis of *n*-hexane layers from *in vitro* biochemical reactions of GPP without (i, iii) or with NsBERS (ii, iv); (B) GC–MS analysis of *n*-dodecane layers from *in vitro* biochemical reactions of GGPP without (i, iii) or with NsBERS (ii, iv).



```
NsBERS/1-435   1 ----------MKSF---LYWFIWSGTNNSKSEAIAPLTPP----PPVRLQNKPSQAKKHITDIVASHEDLKYINPIRYAYDPVSPAAPYLNPI-LFF--  79
LsBERS/1-444   1 --------MSLPSF---FNWQIRRSTPRPVPSAPSSASDSSGSSGSSASSASTHQTKRNLTK-VVSPGDSAHISPIKYAYDPVPEVPQYLSPL-PFE--  84
BcBOS/1-441    1 MAQIMTSAESLPEFFSISFNWEFQKNDGIHQ---------DSRSPPPVSLSS-TTKSQID-LKRVTEPKDLPYISPIEYVHDPVPEVPCHLTKLPPLP--  86
AaTPS/1-418    1 -----------------------------------MLVPRSHMS--------PDLTYTEVNQNLAARENASWFSPVRFAYDWLEDAPIEHLTAVPLKIR  56
Hyp3/1-372     1 ------------------------------------------------------------------------MRPITCSFDPVGISFQTESKQENFE--  25
FfAAS/1-371    1 -----------------------------------------------------------------MPHKDLPIRPLVRAFDPVGPDTLGPPDLDFAS--  32
FgFS/1-371     1 -----------------------------------------------------------------MPHKHVPLRPVKLTFDPVGSNTLGVPTLDFES--  32

NsBERS/1-435  80 ------FPSQ---ASRPCPQM-ANSVRIYPLRGGLPYRSSLDYVRGCKHFRANIQESTDLLQGIMRDKSAGDIPVGTAKITLSKL-AKHELRPGIEDRM 167
LsBERS/1-444  85 ------FPHY---ATKSCAYATLNAVKIKPRAAGLPYRSSLDYVRGNKNWKANLDESVKLLELIVADKAALDEEVGK-GITLSKI-AKKELRPGLEHRM 172
BcBOS/1-441   87 ------FPQY---ASTDCQYTSVNSVRVNPRAAGLPYRSRFELIRASKHWKANVDETKKMLELIIADHSSTSVGMR-DGLTLAGL-AKKELRPGLEHRV 174
AaTPS/1-418   57 TDSPLASPKPSNVKEQSITENNENSFSISPQLTGLPWPTSFTKVRQNRHWRQSLRISTQLLELFAADDTSAQ-AVRRNGVSLARI-ASHELQTDEEDRF 153
Hyp3/1-372    26 --------FLREAISRSVPGL-ENCNVFDPRSLGVPWPTSFPAAAQSKYWKDAEEAAAELMDQIVAAPGEQGSLPA---ELAVSDKKAAKRRELLDTS 112
FfAAS/1-371   33 --------LFRE---RNVPE--DAPLTLYPEQLNVPWHTSLPWTRQSKWWVQGEAAGRDLVNRISADKASERGALPV---EFMDE-RRKGKIDELVEDA 114
FgFS/1-371    33 --------LFRE---DSVSE--DAPLVIYPEDMGVPWNTSLPWTRQSKFWAYAEAAGYEMANGISLDKASERGTLPM---ELMDE-RRKWKIDELVEDA 114

NsBERS/1-435 168 MLATNYMFPFADARRVKIIAVLMIMYFIFDDKVEETPEGAQLNHFRNDFLNHFQDPGVLSTEP--TSDFHRHTRETMTAIWEEDGISGNGGREMIEALR 264
LsBERS/1-444 173 VVATSYMFPFADERRLRQIATLMIMYFVFDDKVEETPEGSPLSNFRTDFLRRFREPSSVESDVSTMSELQHHLDFTVRIIGEEDSISGNGGKEMLEALM 271
BcBOS/1-441  175 VLATSYMYPNANERRARIIAATMMLLFIYDDKMEETPEGLSFTNSREQFLDYFKKENDVMASDSTTSDFQRHLKSTMSAIADEDEISGNGGKEMIEAMI 273
AaTPS/1-418  154 TKFATYIFPEANEERMKLLAATIVYIIIFDDSWEMHSE-DTLGLVRDDFIRRLRGDIEGMAE--HQTPLQQLINSTVQGFKDQDKTMGNGGQEVLDRLI 249
Hyp3/1-372   113 VSAPMNMFPAANAPRARIMAKANLLIFMHDDVCEYQSVQSTIIDSAL------ADTSTPNGKGADILWQNRIFKEFSEETNREDPVVGPQFLQGILNWV 205
FfAAS/1-371  115 VSCAVYLYPSSSPTRIELLTQALLLFFHDDVMERGATQDDATVC---------------DDFVTMIPKNKHMKRYFAEVLECDPILGPGLLRAIGLFV 198
FgFS/1-371   115 ISCCAYLYPTSSPTRLALLTQSVLLLFLHDDVIERGATQNETTVV---------------DEFLSMAPKNRHLKKFWSDVLECDPVLGPDLLYAIHAFV 198

NsBERS/1-435 265 DAFRCVHP--NGDFTSVEDYLQFRRLNVGARFVIAAAKFTIKSSVDVKD-PRLSRYLGLIGDHLGIINDMASFDKESRALEQGETQDMINVASVIQKLL 360
LsBERS/1-444 272 AAFHCVHP--DGDFHSLEDYLKFRRLNVGARFVIAAAKFSIKSSVDVKD-PRFARYLSLVGDHYGIINDMASYDKESKALREGETQDMINIVAVFQTLL 367
BcBOS/1-441  274 GGFRCVHP--NGEFQSLEEYLSFRRLNVGAQYVIAAAKFTIKSSVDVDD-PRFARYLRLIGDHLGIINDMASYEKECRARKEGRHQNMVNIVAVFQHLR 369
AaTPS/1-418  250 DFCEHVPP--QTKFATMGDYLSYRLIDVAFPYLLACIKFSLGSSVNVED-PKLAPILRLVSDHVSLVNDLASYDKEKRAYDNGSACYLINAVDVAQRLF 345
Hyp3/1-372   206 EHTRKALPAS-MTFRSFNEYIDYRIGDFAVDFCDAAILLTCEIFLTPADMEPLRKLHRLYMTHFSLTNDLYSFNKEVVAEQET-GSAVINAVRVLEQLV 302
FfAAS/1-371  199 NAGRKKSPFKQDKYATLAEYLDYRRHDIAKPFMIAAIRFGSGVRQTPEETAPFAELEDLYVQHSILINDLYSYDKEMYEARTI-NGSVVNAVHVIEKLM 296
FgFS/1-371   199 RDGRVKSPFKQDHYATLADYMLYRRNDVGKTFMIAAIRFGSGVQQTREELAPFDELADLYVRHSILINDLYSYDKEVHEVKTI-DASIVNAVAVTEQLL 296

NsBERS/1-435 361 SLQSMEAAKAASYAYQLQVEYWIMEEIKRLEREEDLTDEEWWFLEAVFLTATGNTFFCMTSARYGGQCAAIDPSW--                       435
LsBERS/1-444 368 SLPDARAAKAAAYGYQLQVEAWIVEEIEALESFGKLSDEDWWFVEAVLLSATGNVFFCMTSSRYGGQAAMIEPQYTS                       444
BcBOS/1-441  370 SLPSSAEAKVAAYTYQLQVESWIIEEVEMLAARGDLTDEEWWFIEAVFMTATGNAFFCMTSSRYGGDAAKLT-----                       441
AaTPS/1-418  346 SLPSAAEAKALTYSMQLLVEAQIKTELDSLVAGGILSCEELRFLDAALLMASGNVFYSVVSSRYGGKAAKLEK----                       418
Hyp3/1-372   303 DTS-TRSAKVLLRAFLWDLELQIHDELTRL-KGTDLTPSQWRFARGMVEVCAGNIFYSATCLRYAKPGLRGI-----                       372
FfAAS/1-371  297 CVP-PHLAKTITRTMSFDVEKKYYAESERFMRDPALNDKQRTYVIALFDCLTGNLFHHATLGRYSRYAEYVFDCKT-                       371
FgFS/1-371   297 SVS-PDLAKNLTRAITFDMEKEFYGICEKFMHSPDINDRQRVFVTALFDALTGNIFHSATLSRYVRHGERPLPCKC-                       371
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Figure S5. Multiple sequence alignment of characterized TPSs in Clade III, related to Figure 2 and Figure 3.

The multiple sequence alignment was generated using Clustal Omega and edited with Jalview.

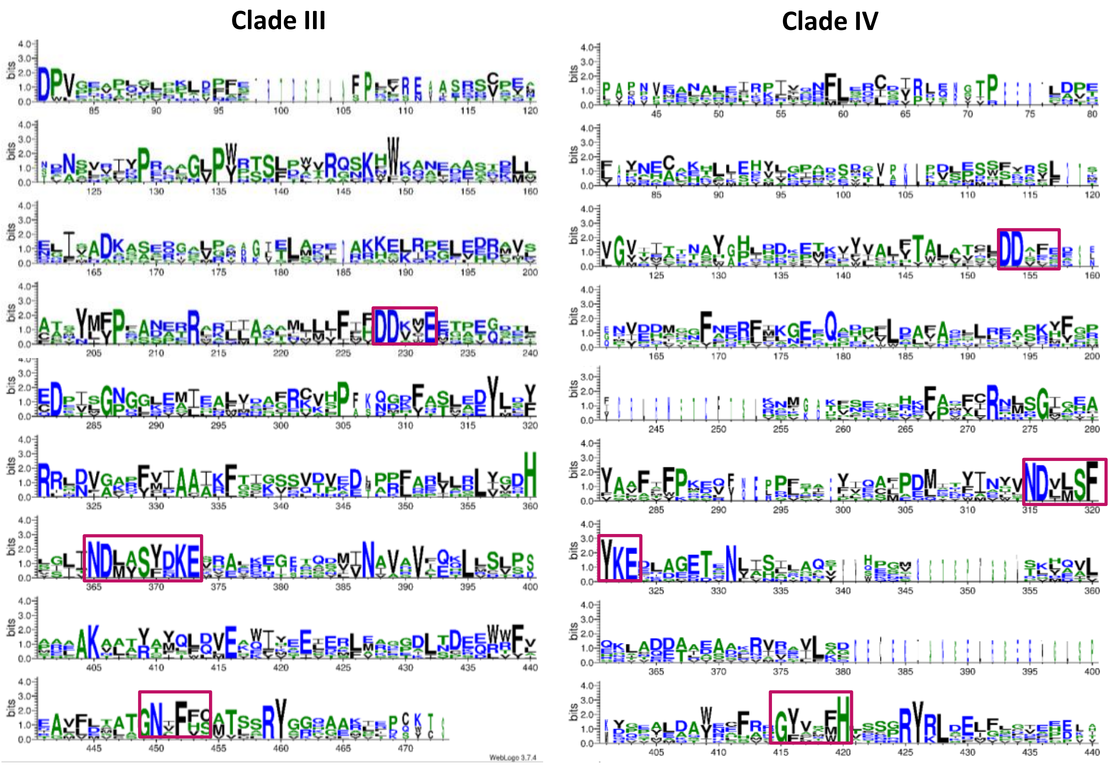


Figure S6. Conserved sites analysis of identified STSs in Clade III and Clade IV, related to Figure 3.

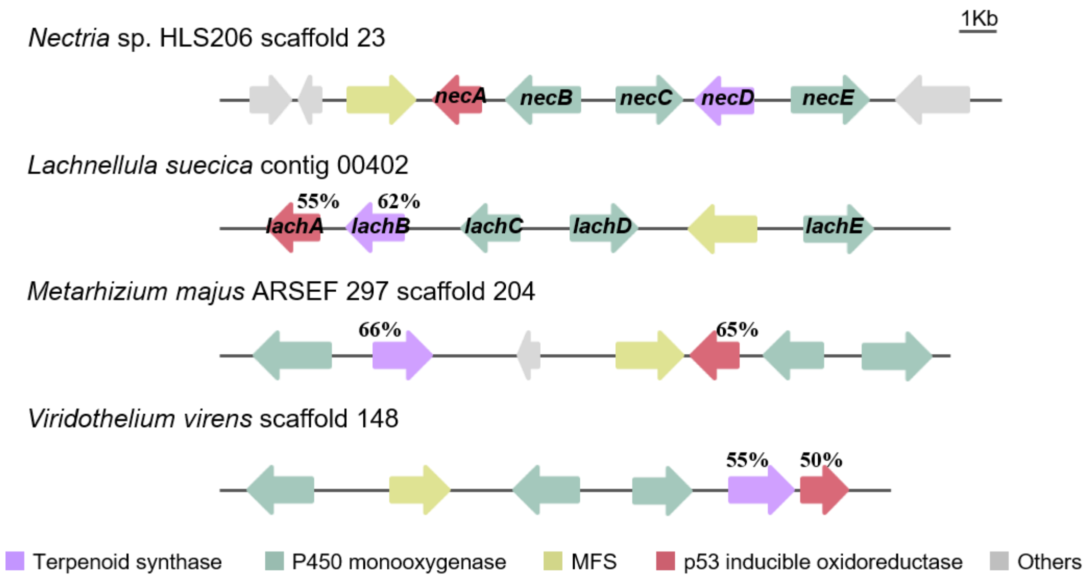
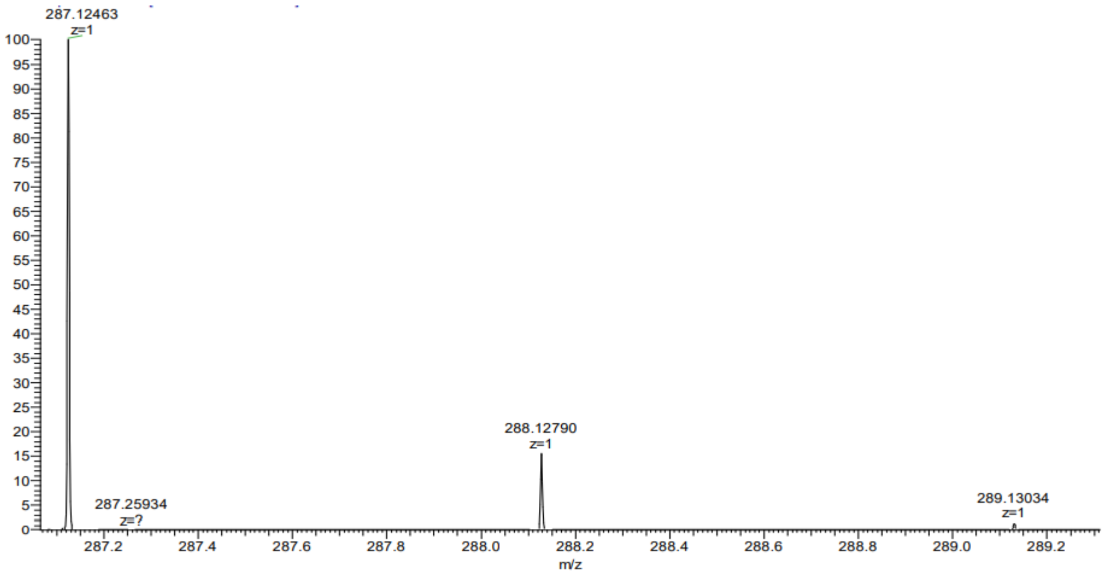


Figure S7. Homologous gene clusters of *nec* cluster, related to Figure 2.
The PIG3 variants, NsBERS-like enzymes and cytochrome P450s monooxygenases co-localized in many phytopathogen BGCs.



m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition	
287.12463	287.12538	-2.61	5.5	C15 H20 O4 Na	M+Na

Figure S8. HRESIMS spectrum of 1, related to Figure 1.

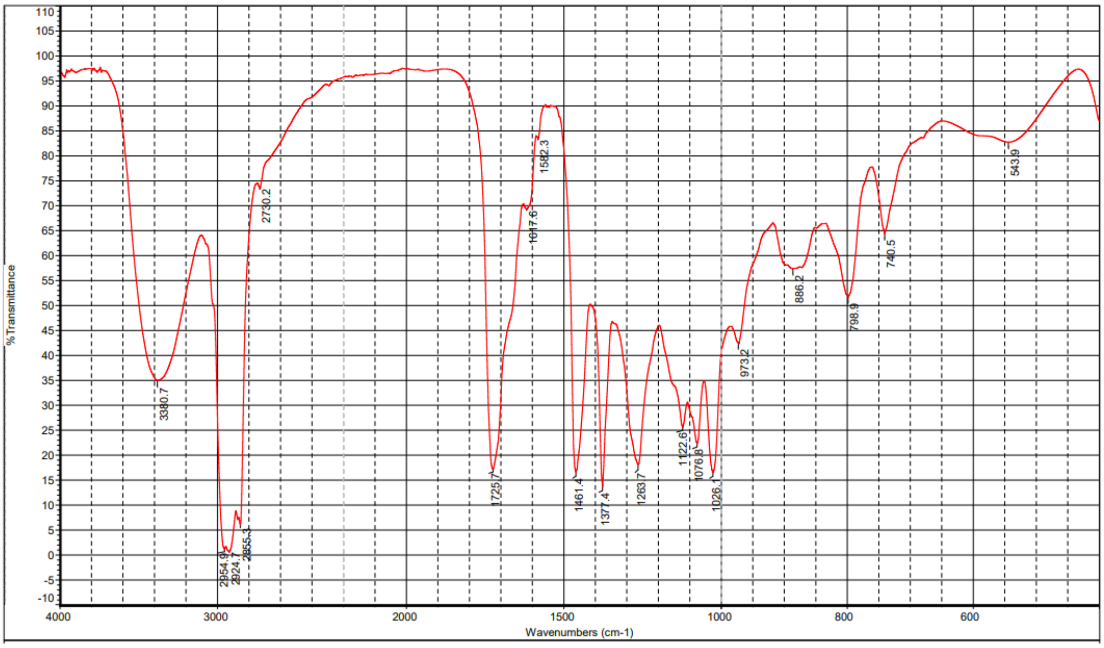


Figure S9. IR spectrum of 1, related to Figure 1.

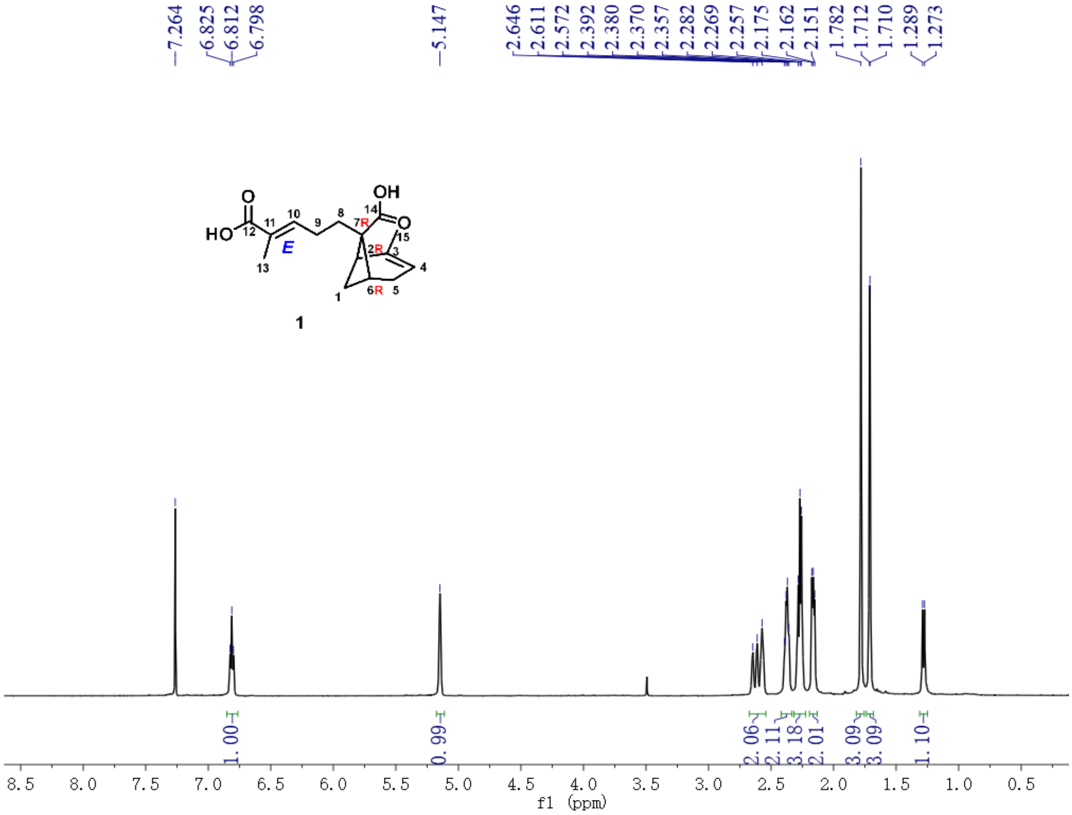


Figure S10. ^{1}H NMR spectrum (500 MHz) of 1 in $CDCl_3$, related to Figure 1 and Table 1.

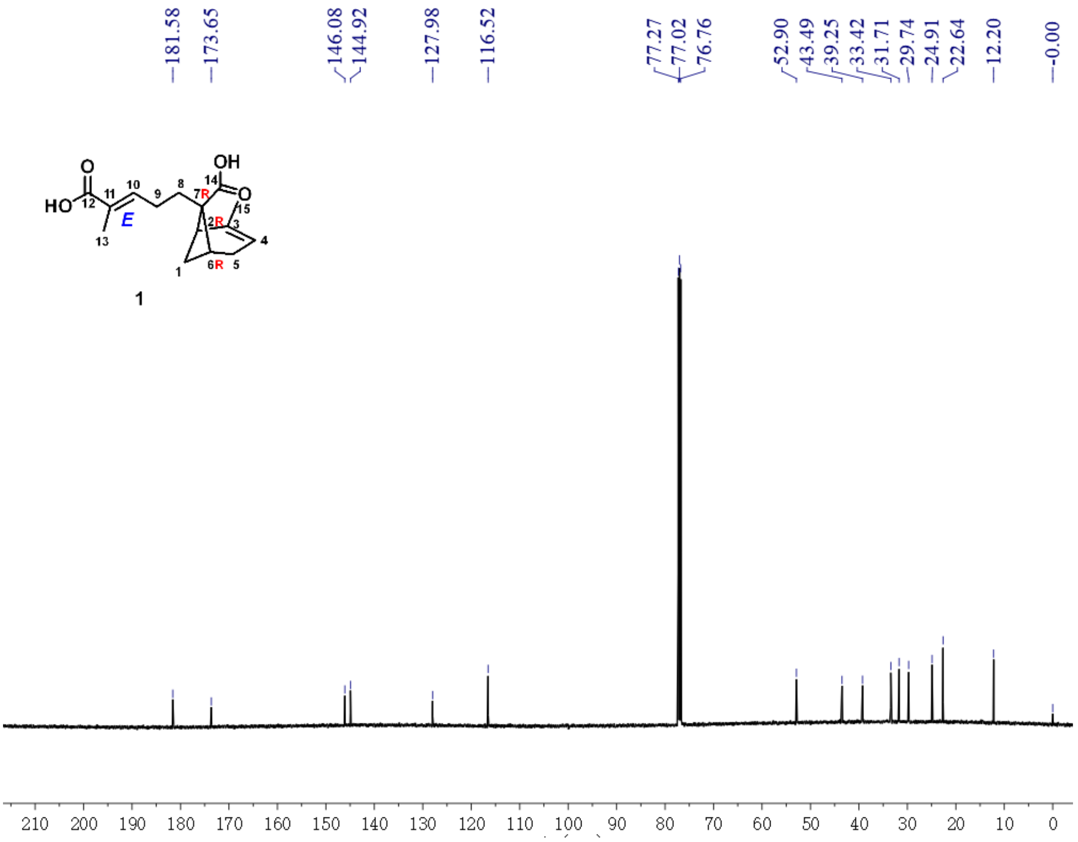


Figure S11. ^{13}C NMR spectrum (125 MHz) of 1 in $CDCl_3$, related to Figure 1 and Table 1.

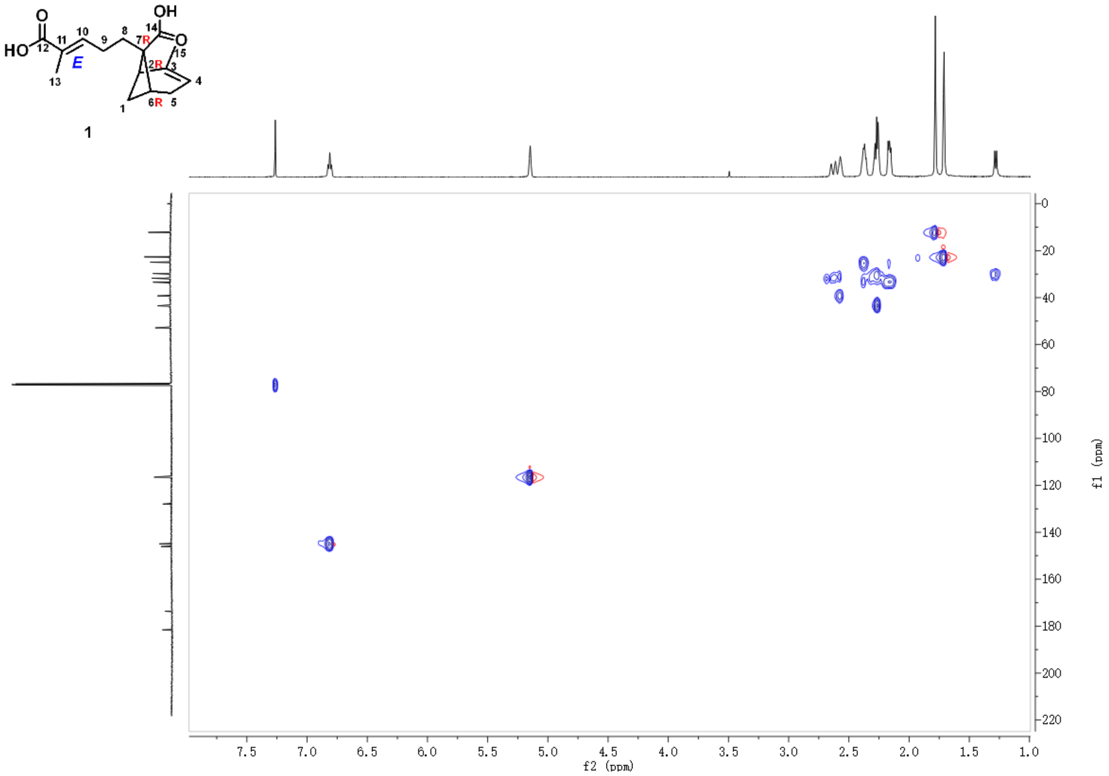


Figure S12. HSQC spectrum of 1 in $CDCl_3$, related to Figure 1.

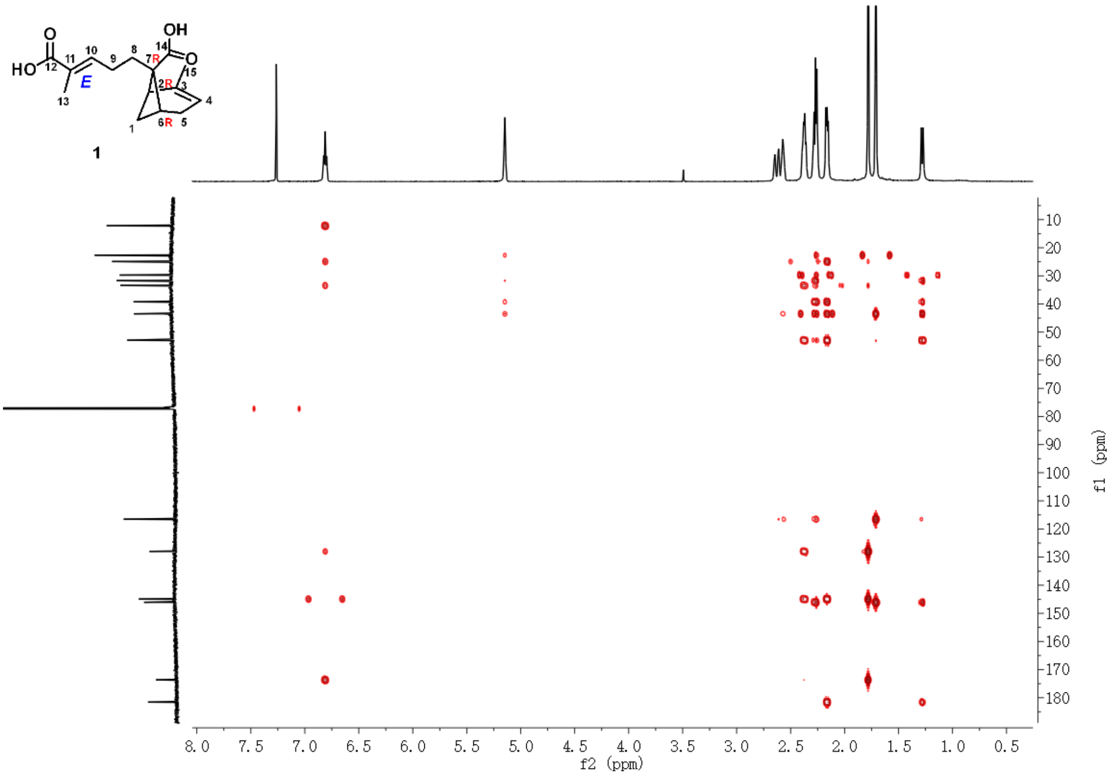


Figure S13. HMBC spectrum of 1 in CDCl$_3$, related to Figure 1.

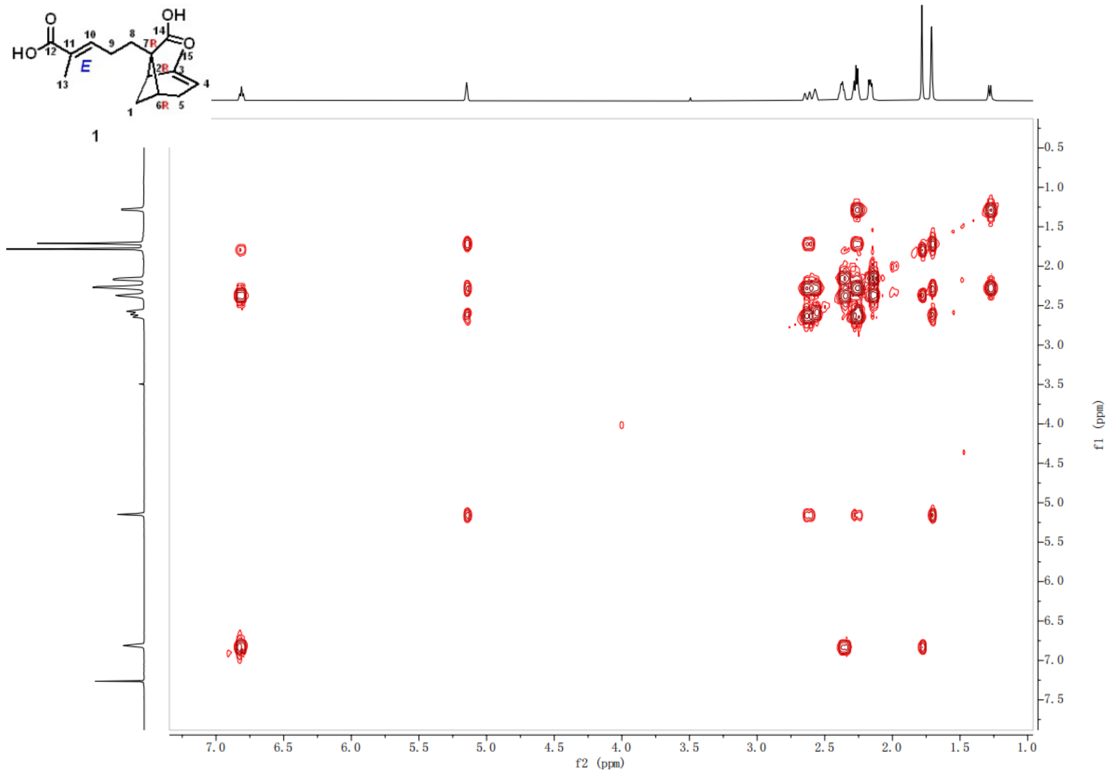


Figure S14 .^{1}H-^{1}H COSY spectrum of 1 in CDCl$_3$, related to Figure 1.

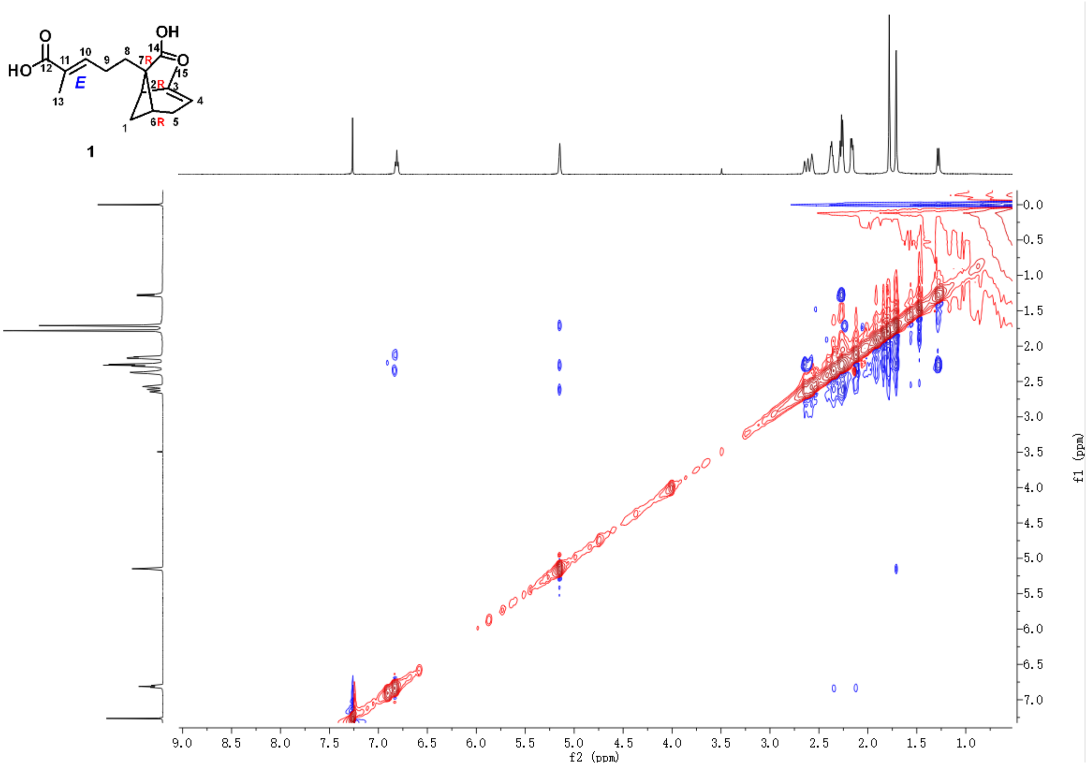


Figure S15. ROESY spectrum of 1 in $CDCl_3$, related to Figure 1.

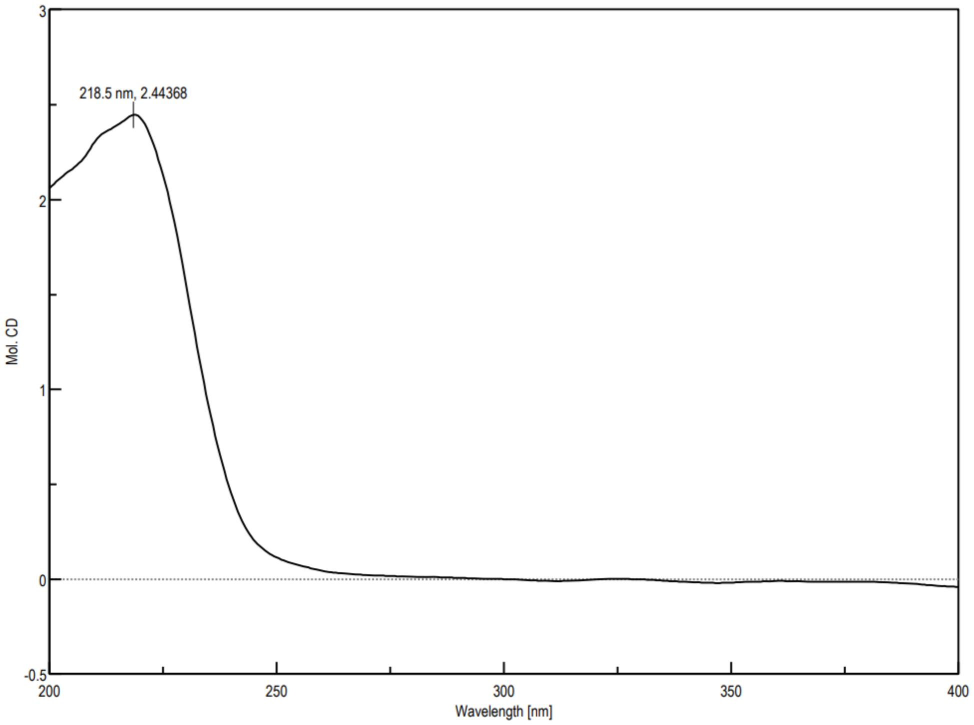
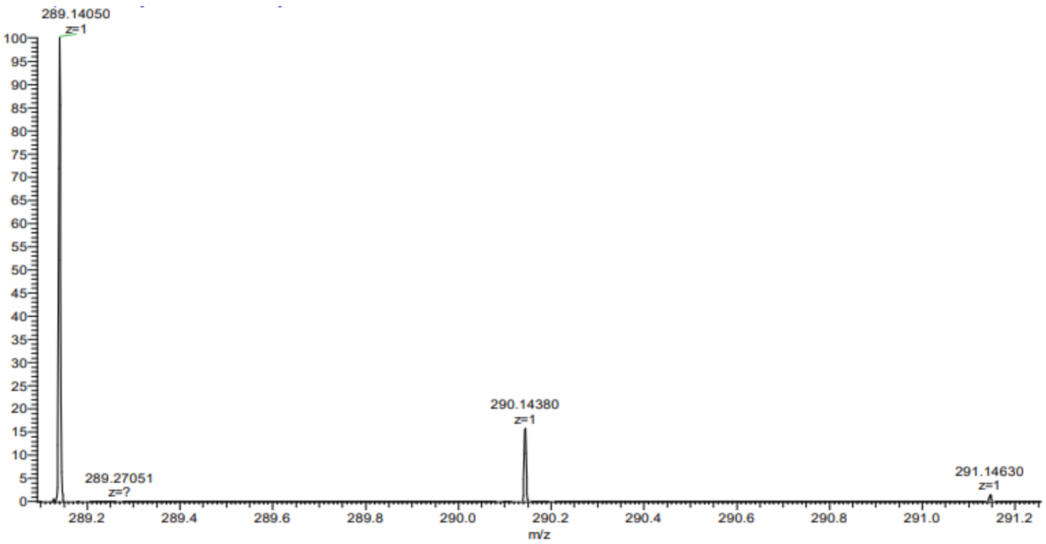


Figure S16. CD spectrum of 1 in CH_3OH, related to Figure 1.



m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition	
289.1405	289.14103	-1.83	4.5	C15 H22 O4 Na	M+Na

Figure S17. HRESIMS spectrum of 2, related to Figure 1.

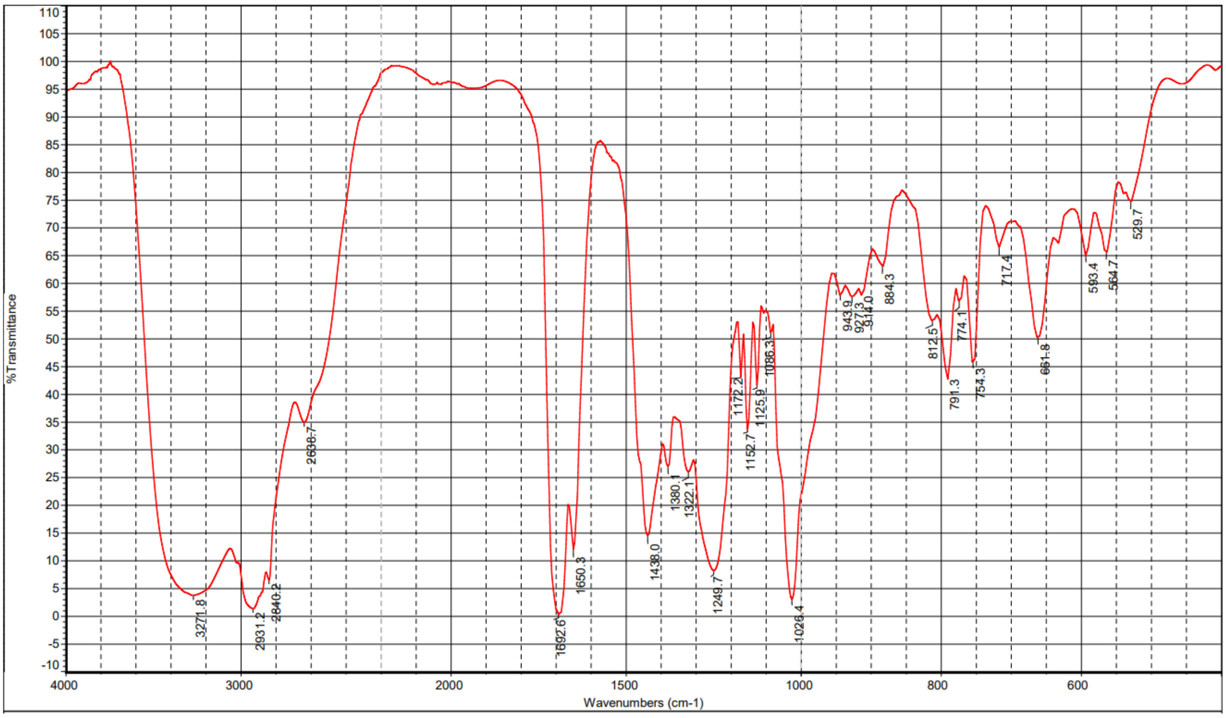


Figure S18. IR spectrum of 2, related to Figure 1.

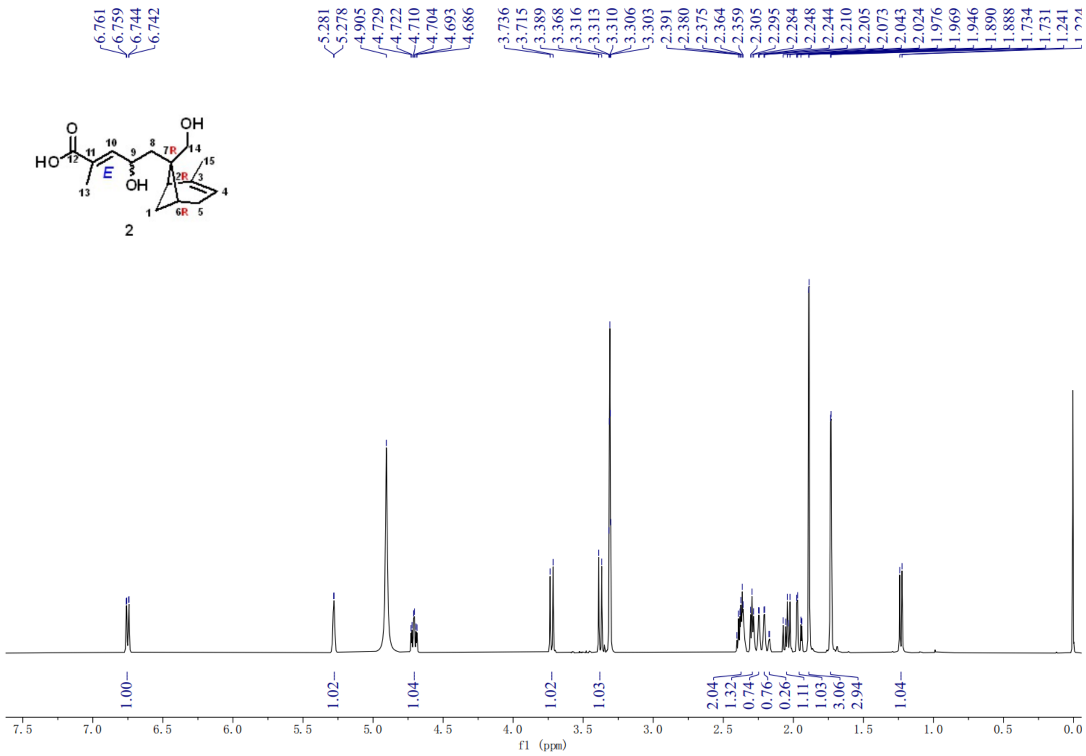


Figure S19. ^{1}H NMR spectrum (500 MHz) of 2 in CD_3OD, related to Figure 1 and Table 1.

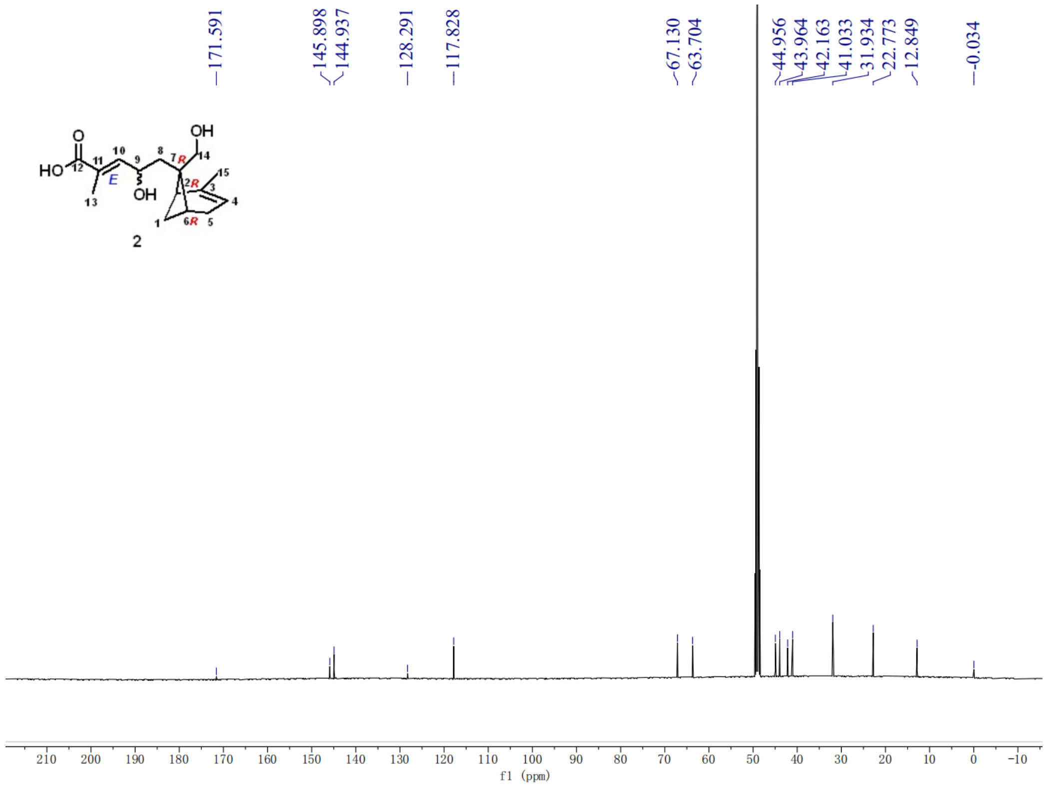


Figure S20. ^{13}C NMR spectrum (125 MHz) of 2 in CD_3OD, related to Figure 1 and Table 1.

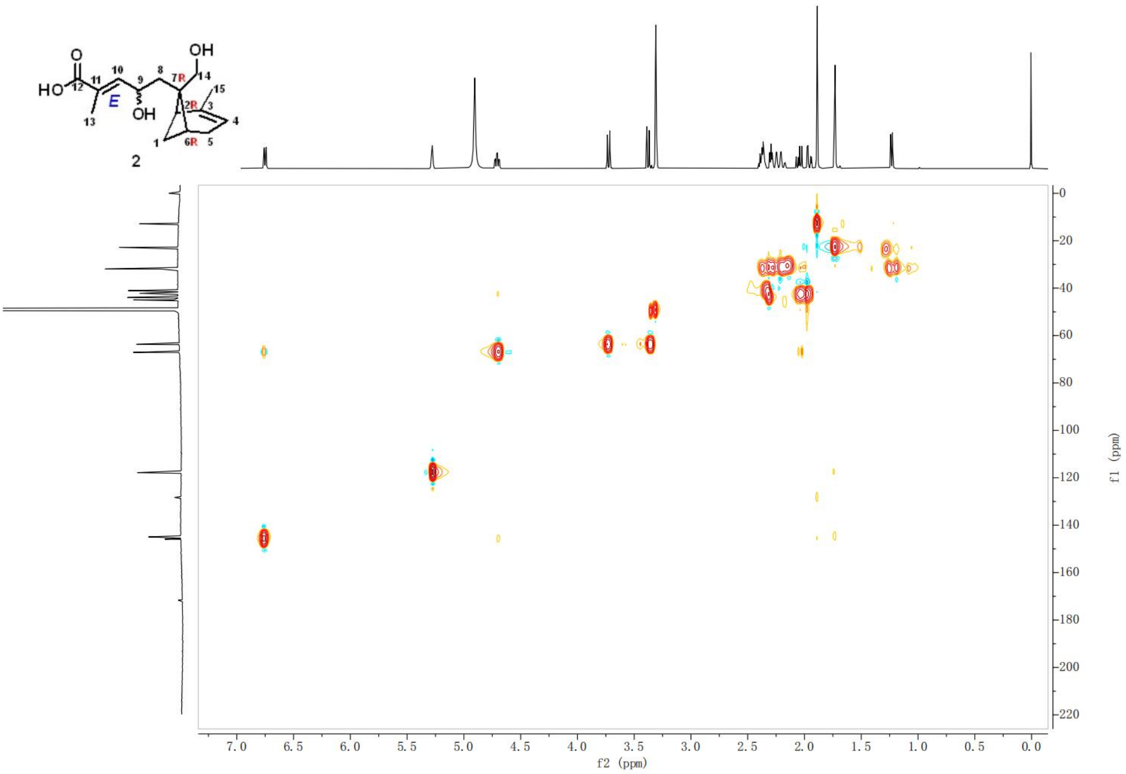


Figure S21. HSQC spectrum of 2 in CD$_3$OD, related to Figure 1.

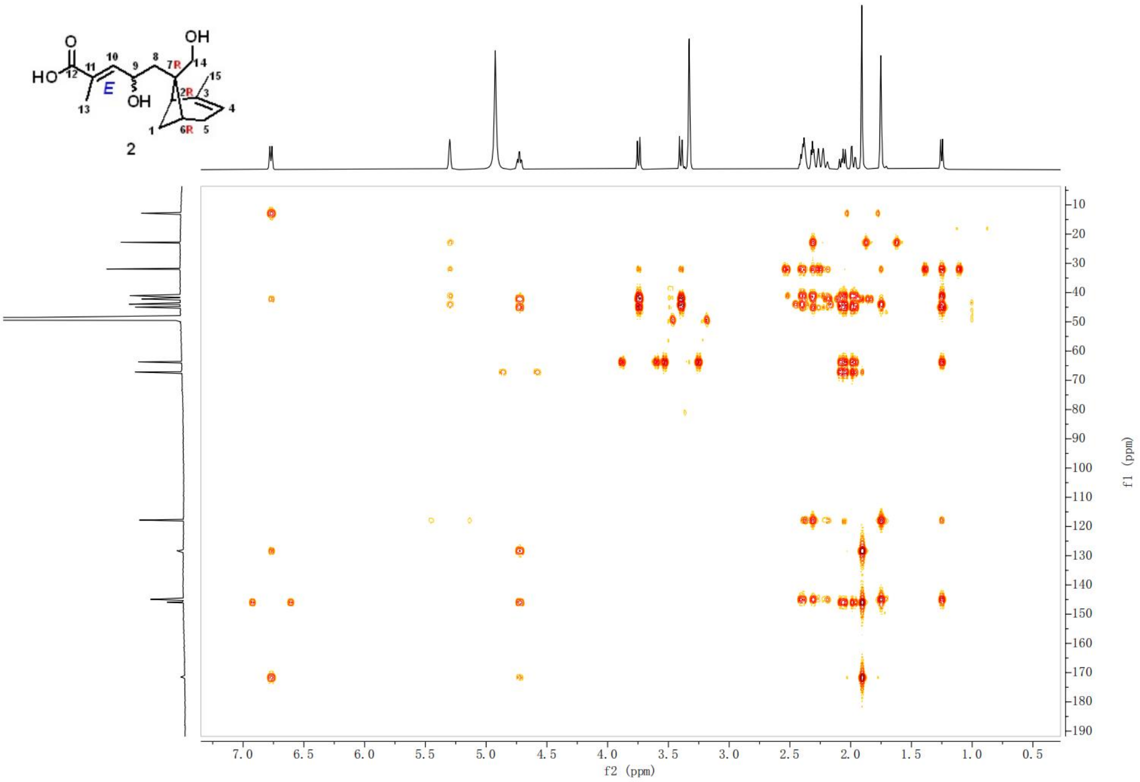


Figure S22. HMBC spectrum of 2 in CD$_3$OD, related to Figure 1.

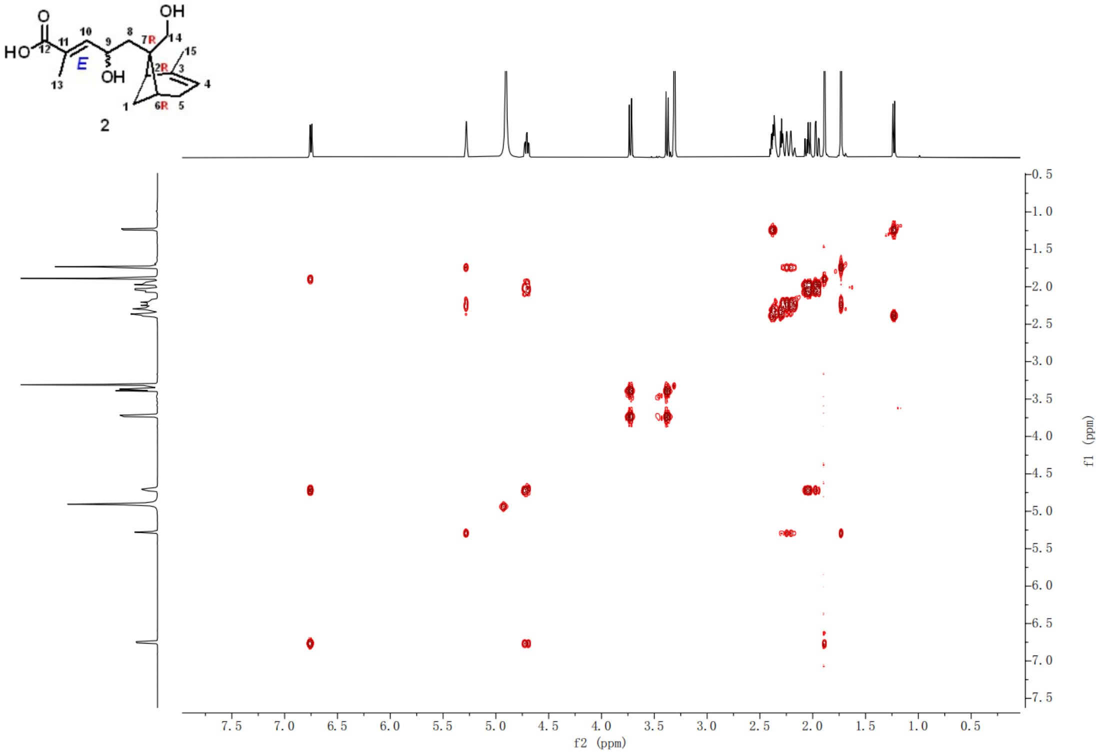


Figure S23. ^1H-^1H COSY spectrum of 2 in CD_3OD, related to Figure 1.

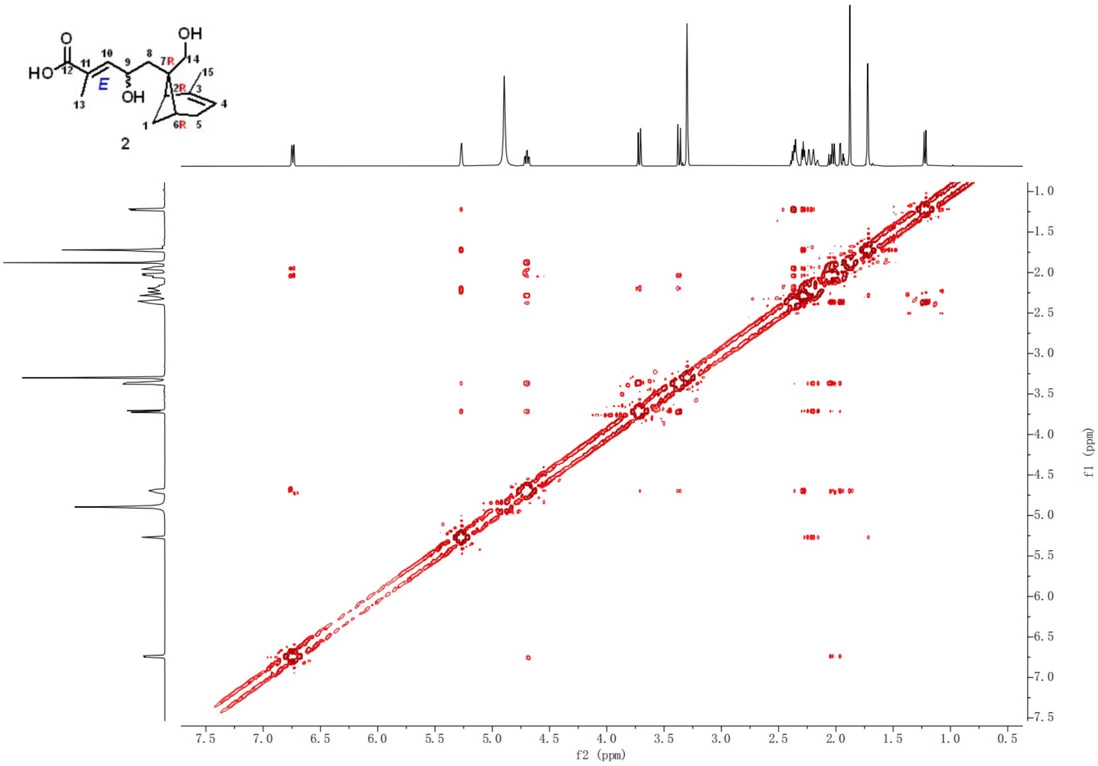


Figure S24. ROESY spectrum of 2 in CD_3OD, related to Figure 1.

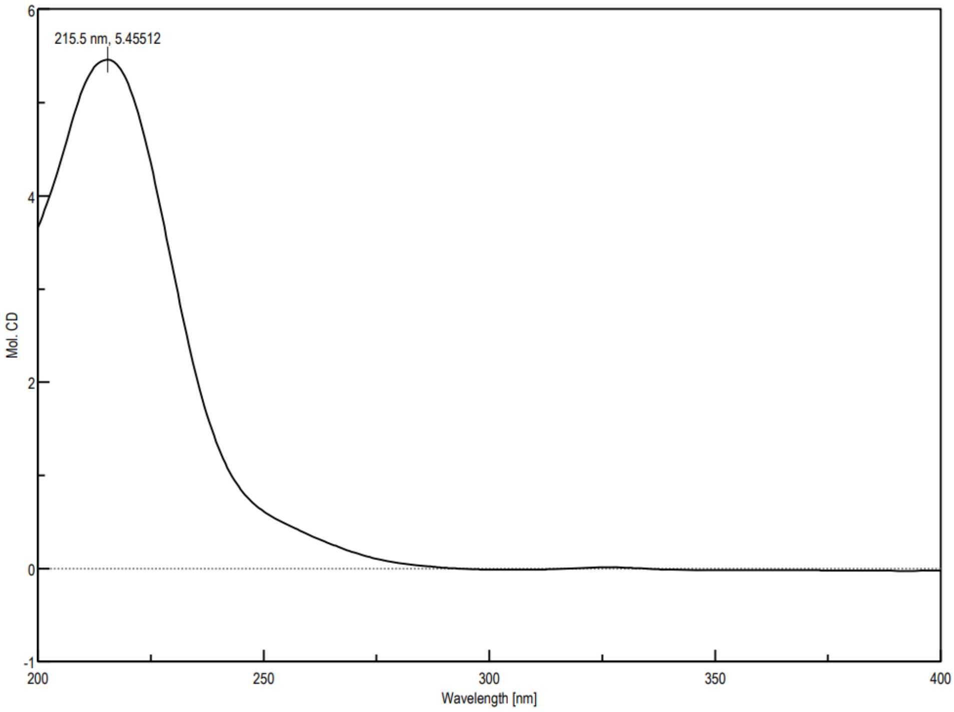
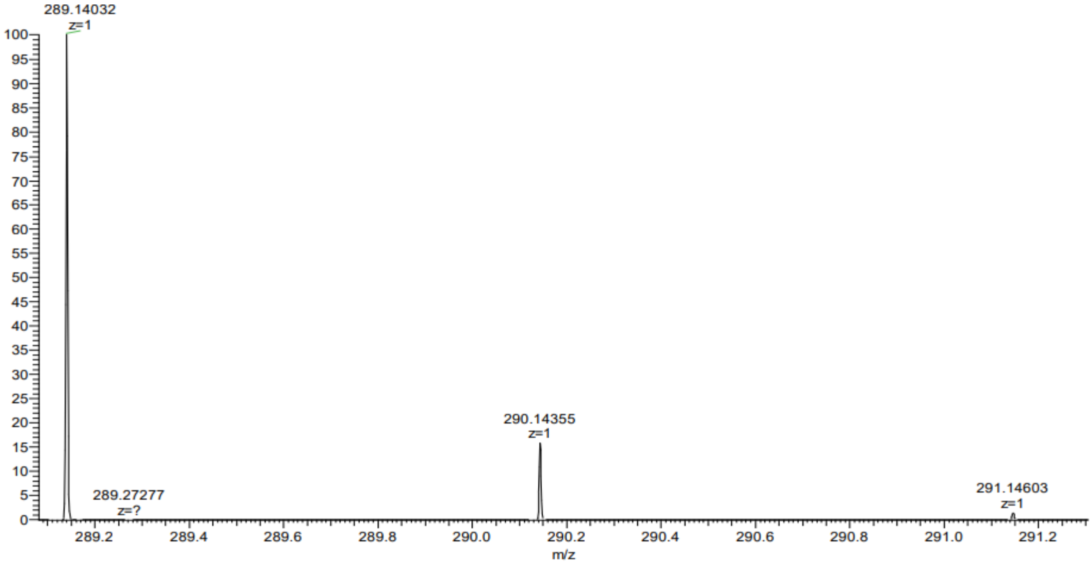
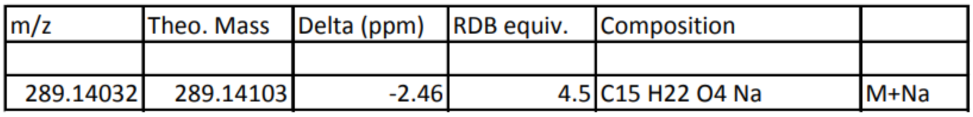


Figure S25. CD spectrum of 2 in CH_3OH, related to Figure 1.





m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition	
289.14032	289.14103	-2.46	4.5	C15 H22 O4 Na	M+Na

Figure S26. HRESIMS spectrum of 2', related to Figure 1.

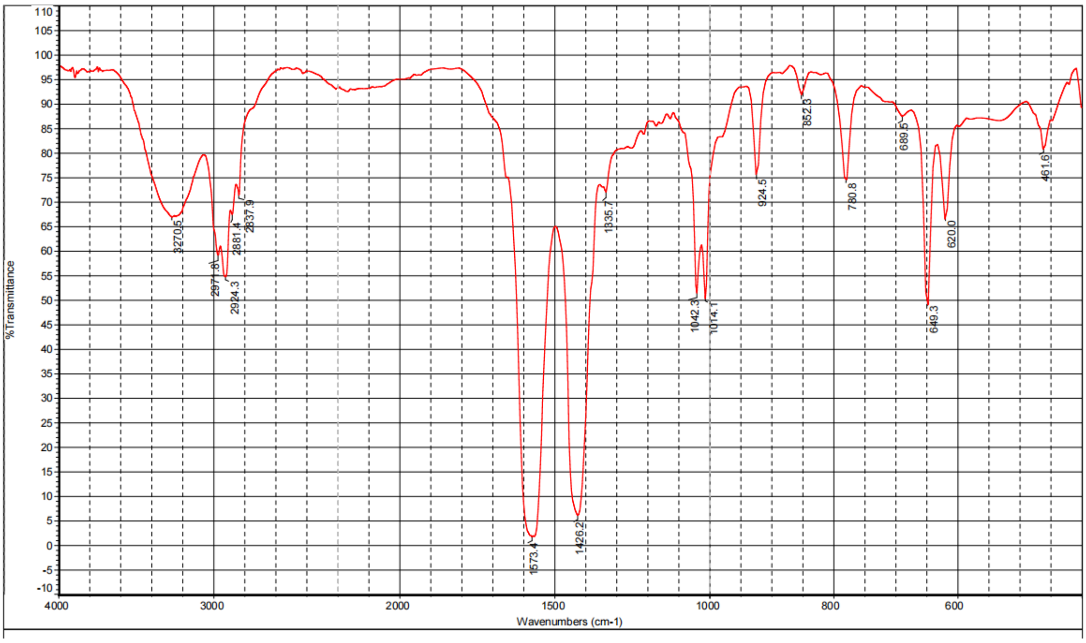


Figure S27. IR spectrum of 2', related to Figure 1.

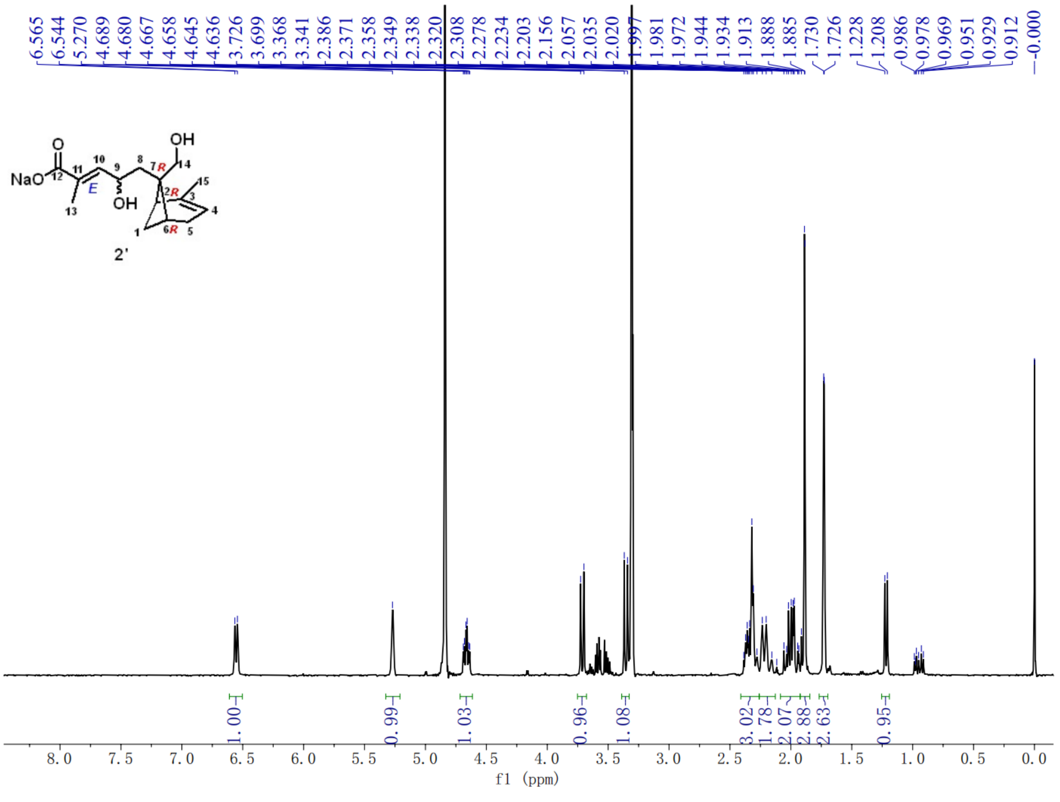


Figure S28. ^{1}H NMR spectrum (500 MHz) of 2' in CD_3OD, related to Figure 1 and Table 1.

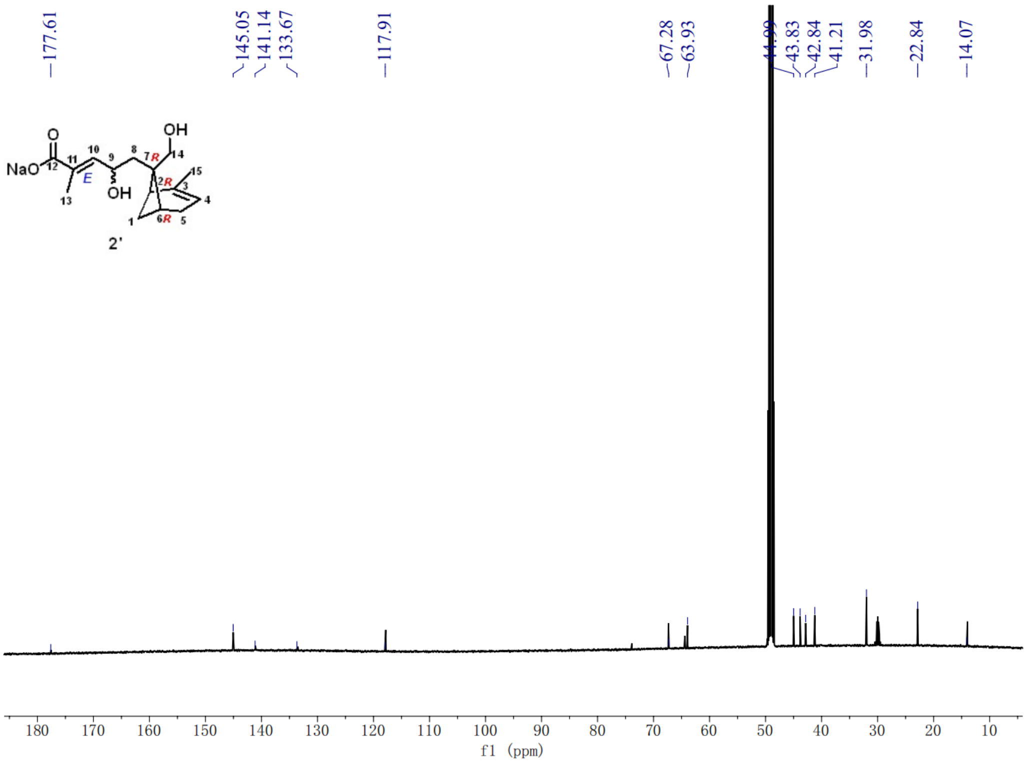


Figure S29. ^{13}C NMR spectrum (125 MHz) of 2' in CD_3OD, related to Figure 1 and Table 1.

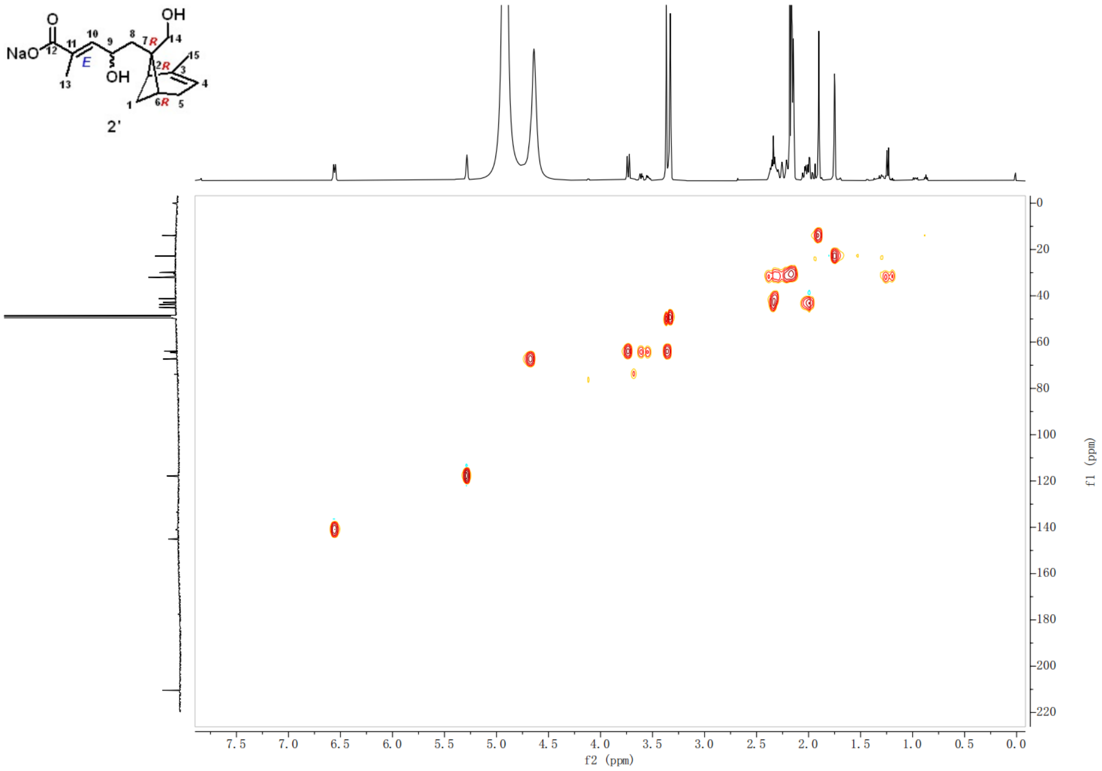


Figure S30. HSQC spectrum of 2' in CD_3OD, related to Figure 1.

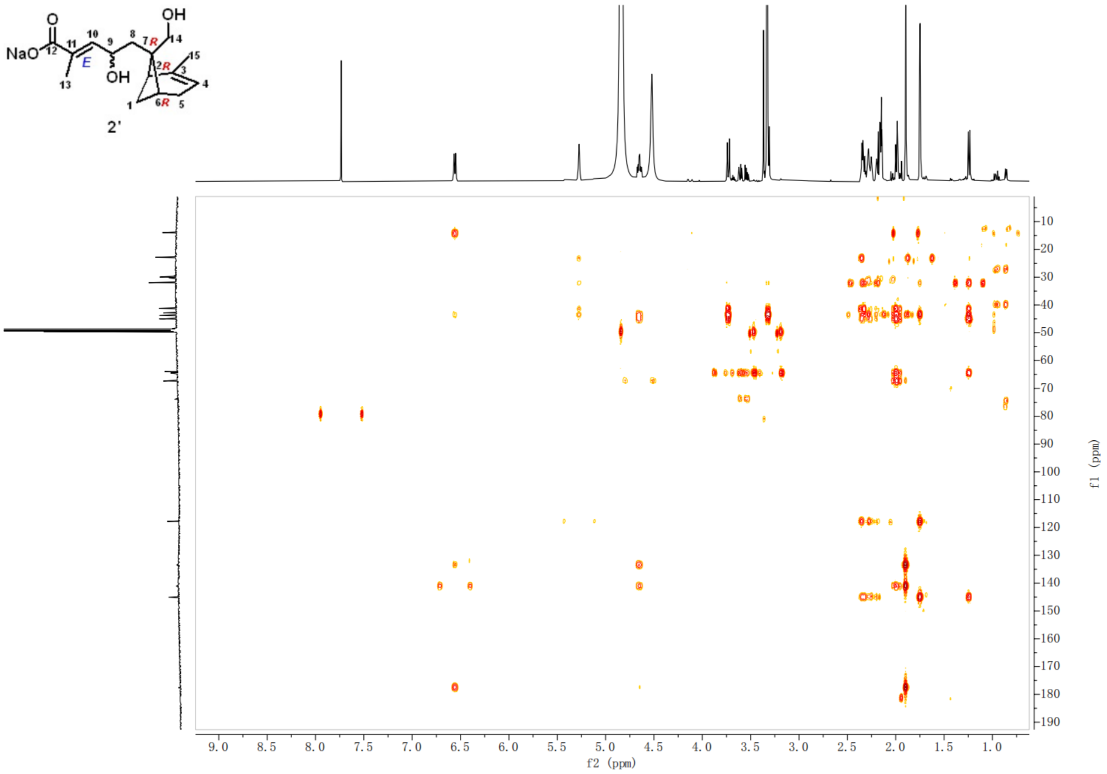


Figure S31. HMBC spectrum of 2' in CD_3OD, related to Figure 1.

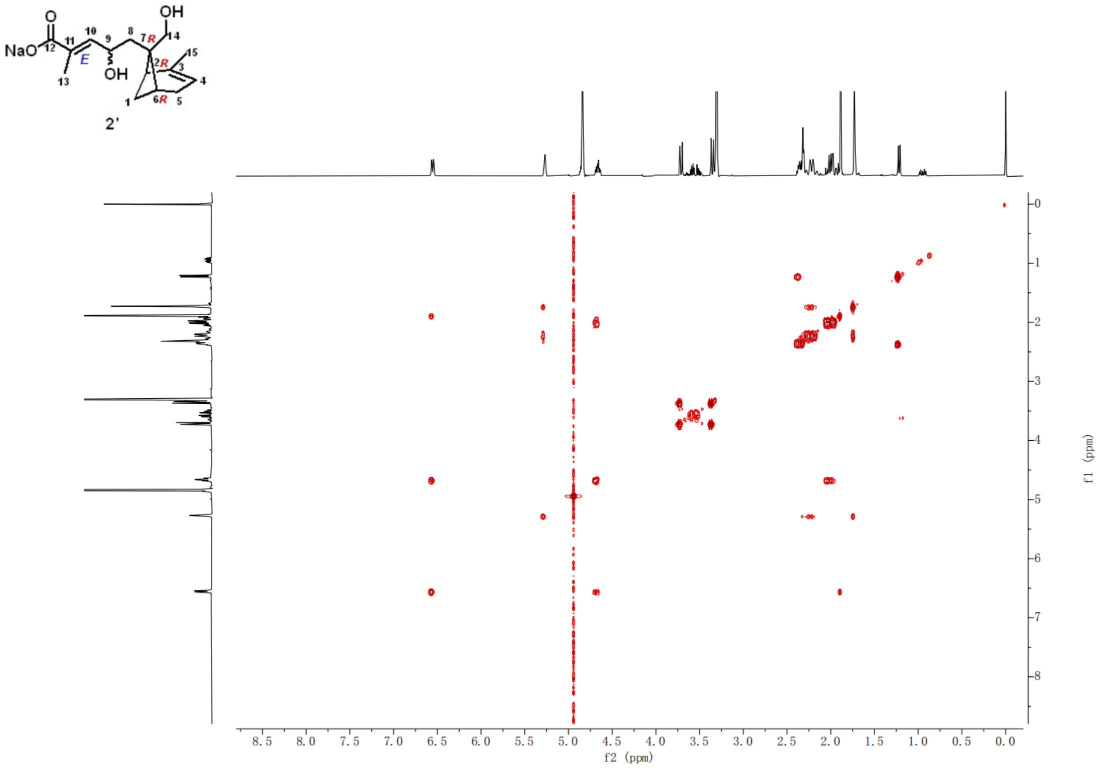


Figure S32. ^1H-^1H COSY spectrum of 2' in CD_3OD, related to Figure 1.

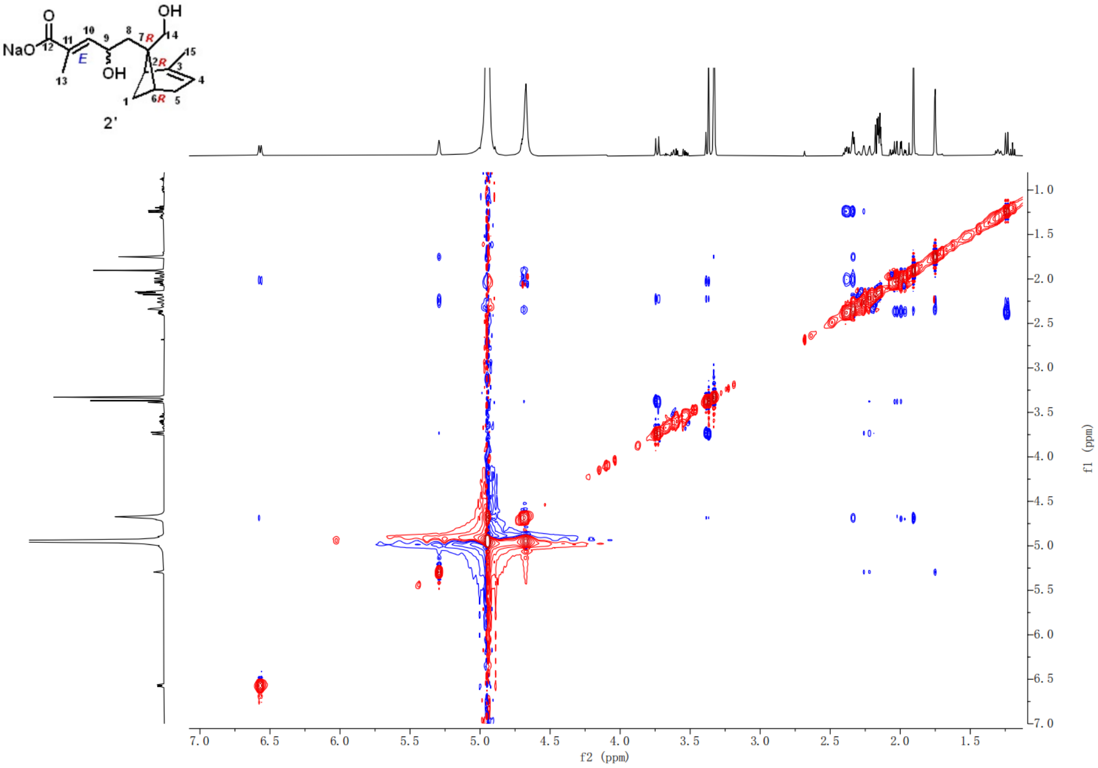


Figure S33. ROESY spectrum of 2' in CD_3OD, related to Figure 1.

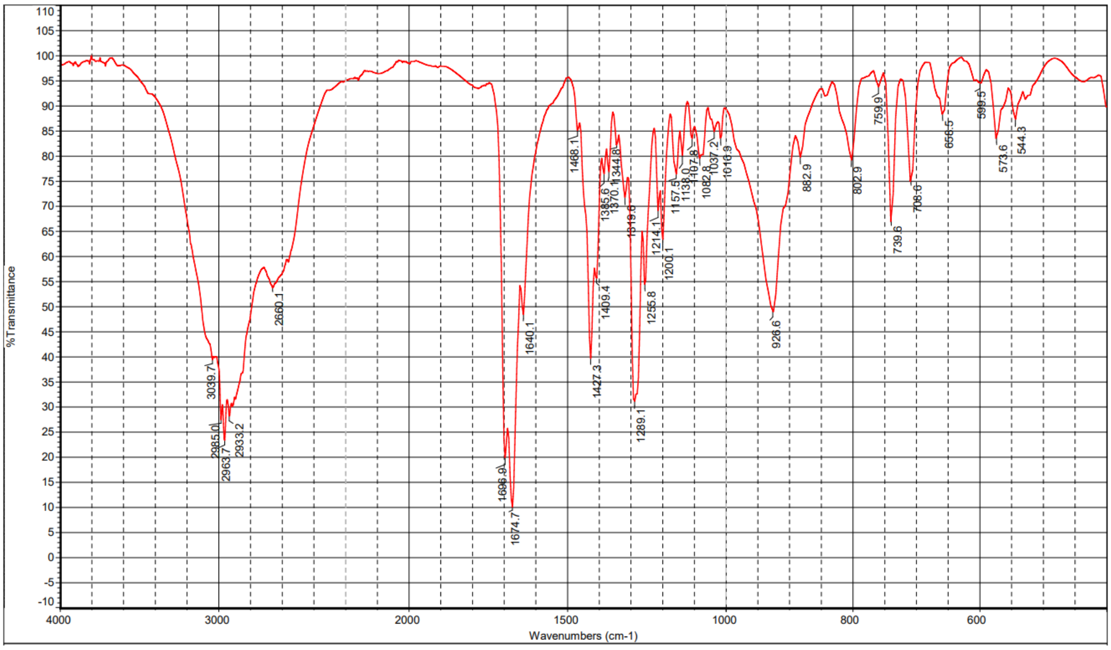


Figure S34. IR spectrum of 3, related to Figure 2.

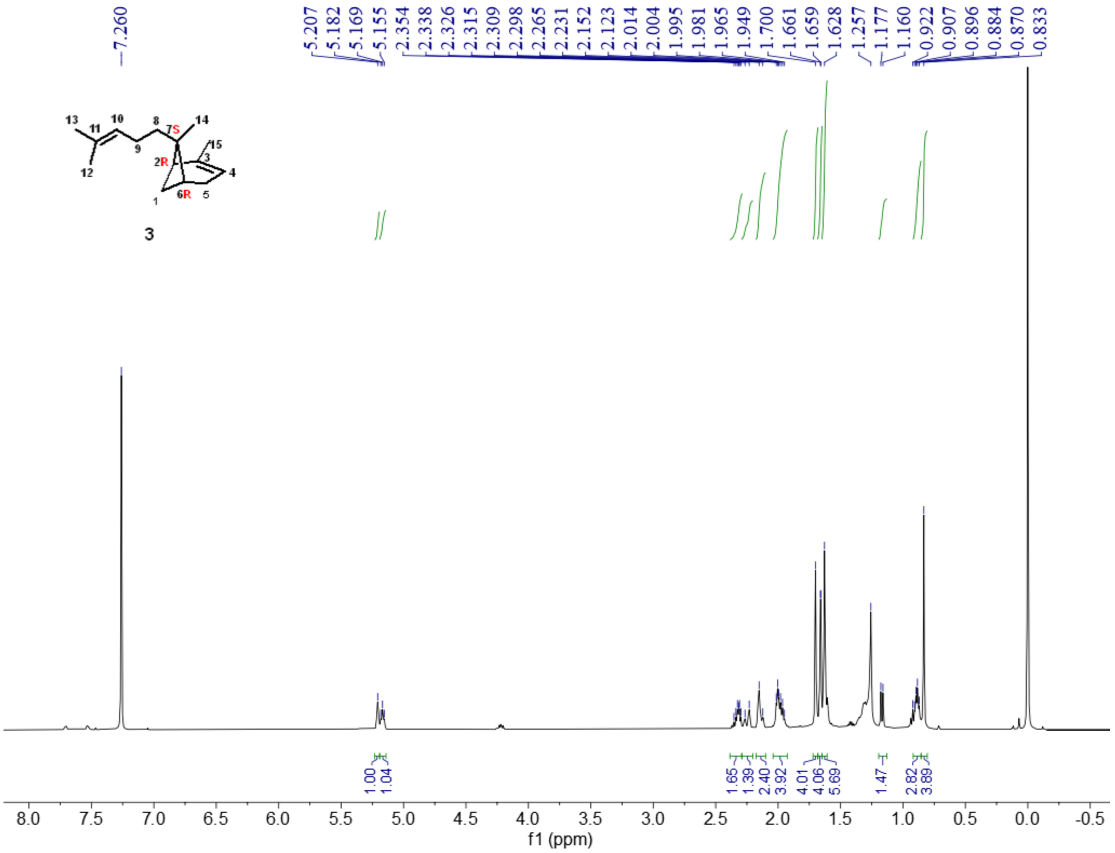


Figure S35. ^{1}H NMR spectrum (500 MHz) of 3 in $CDCl_3$, related to Figure 2 and Table S2.

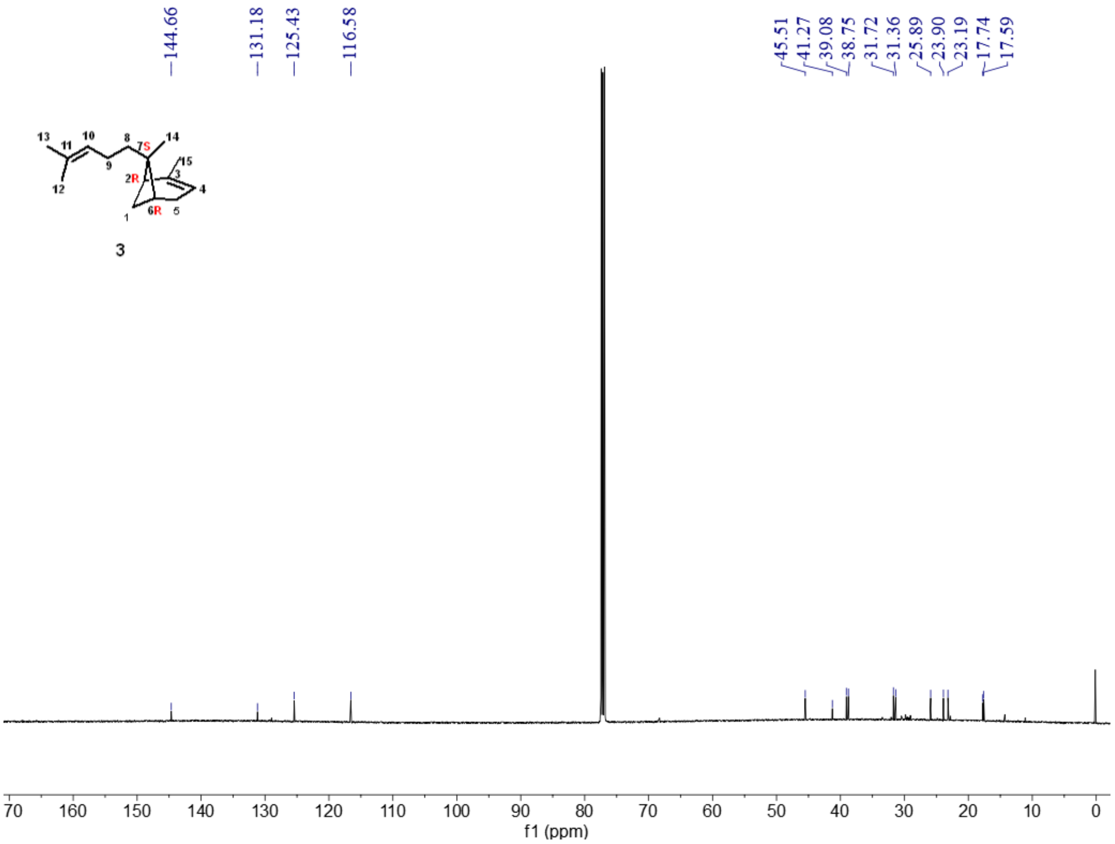


Figure S36. ^{13}C NMR spectrum (125 MHz) of 3 in $CDCl_3$, related to Figure 2 and Table S2.

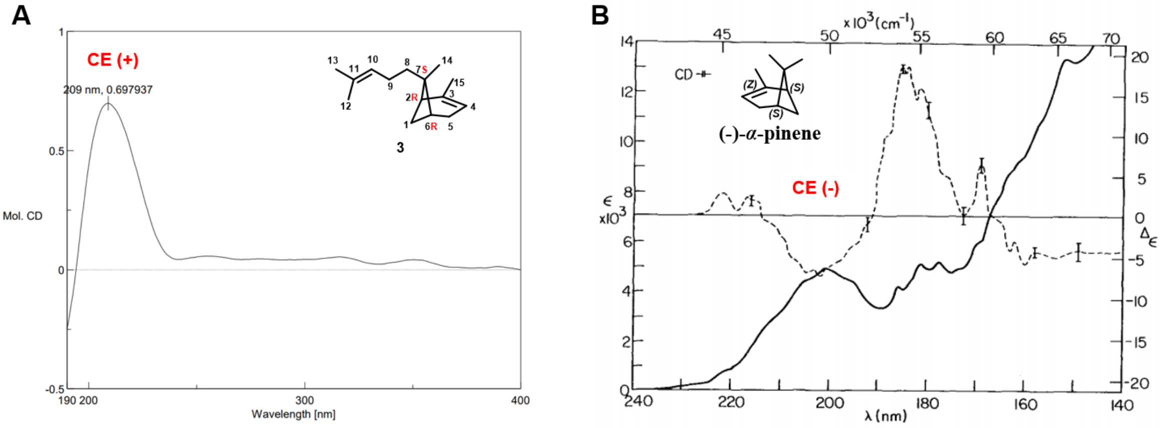


Figure S37. Comparison CD spectrum of 3 with that of (–)-*α*-pinene, related to Figure 2.

(A) CD spectrum of **3** in *n*-hexane, 209nm, CE (+);

(B) The absorption and CD spectra of (–)-*α*-pinene in ref [The Journal of Chemical Physics 59(3):1092-1098], Fig2, Table 2, 202nm, CE (–).

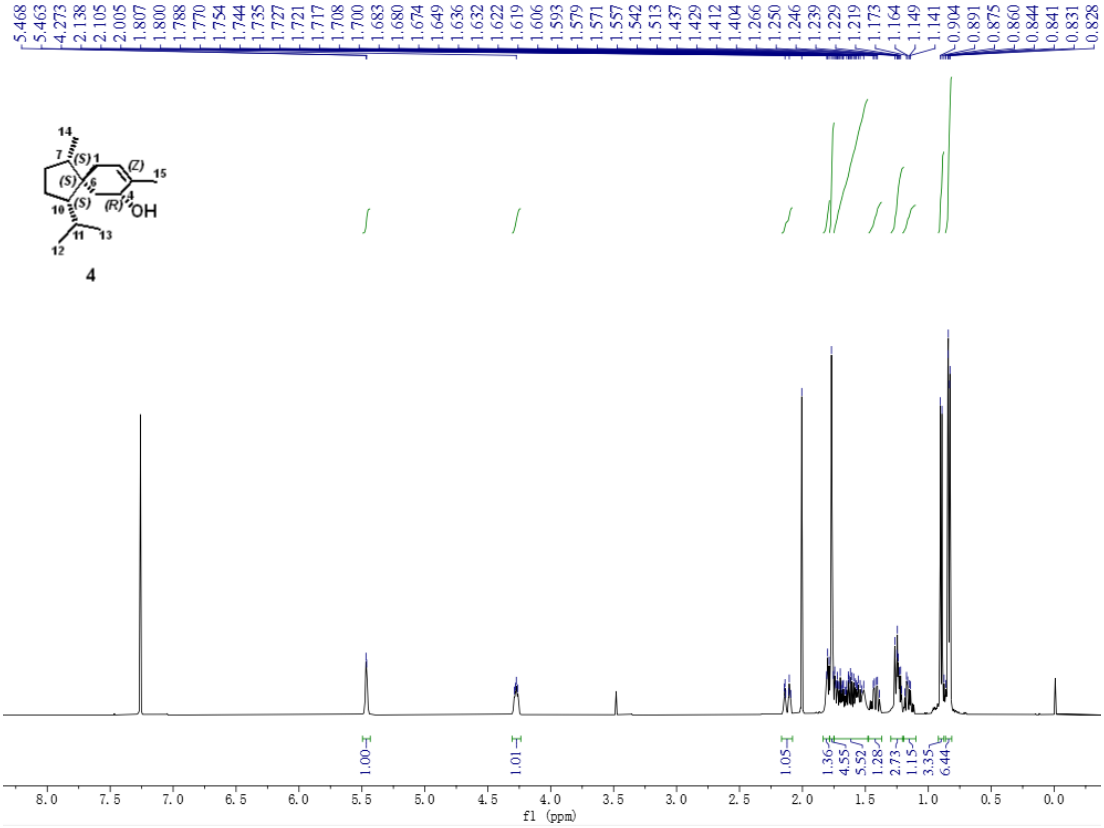


Figure S38. ^{1}H NMR spectrum (500 MHz) of 4 in $CDCl_3$, related to Figure 2 and Table S3.

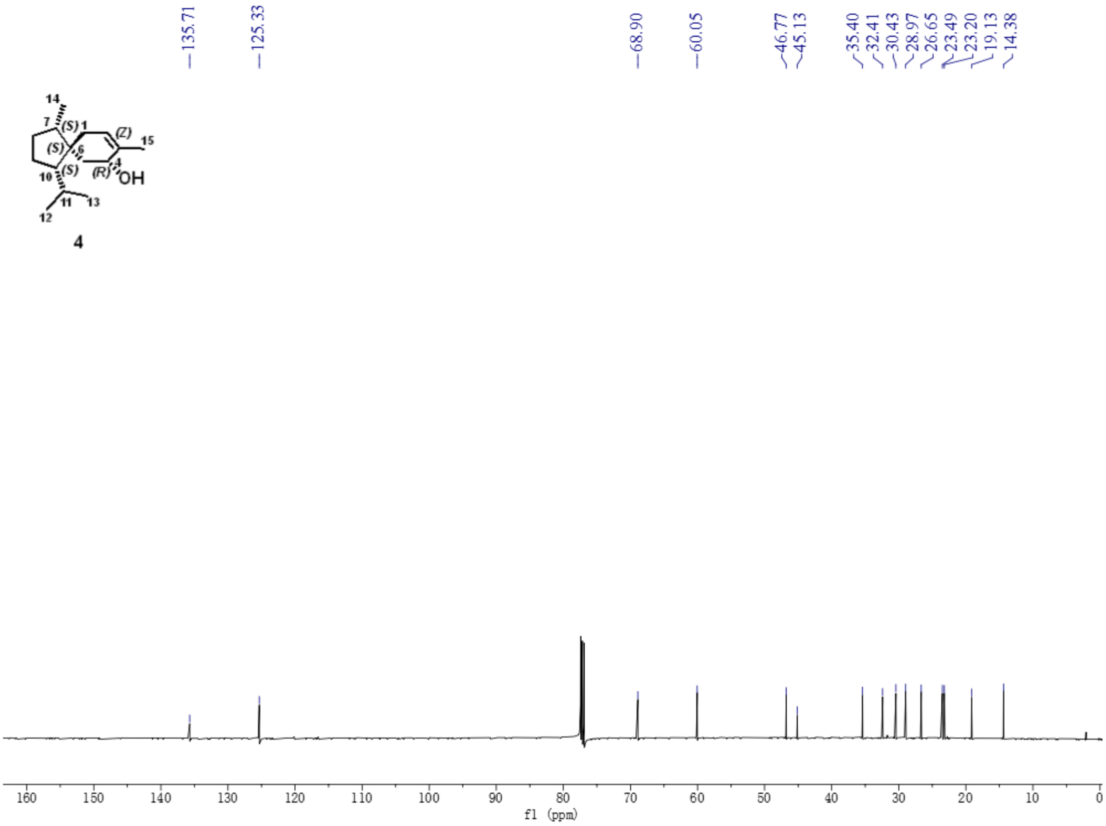


Figure S39. ^{13}C NMR spectrum (125 MHz) of 4 in $CDCl_3$, related to Figure 2 and Table S3.

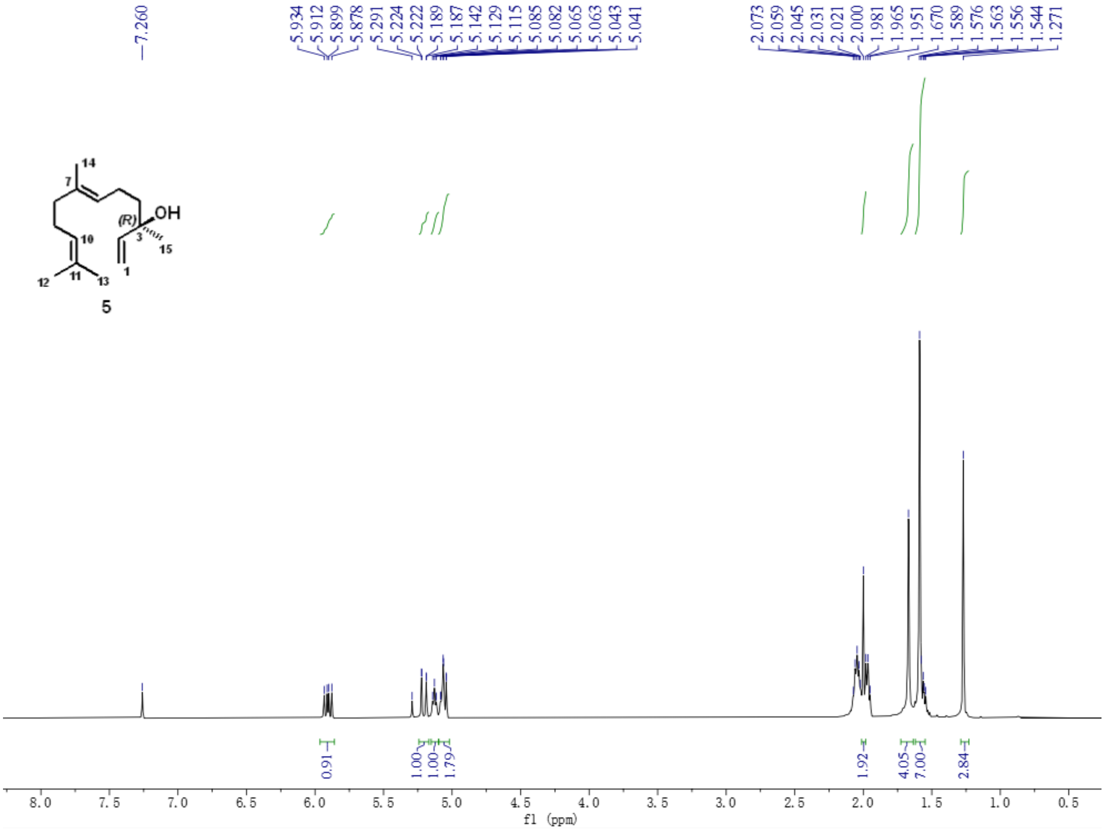


Figure S40. ^{1}H NMR spectrum (500 MHz) of 5 in $CDCl_3$, related to Figure 4 and Table S3.

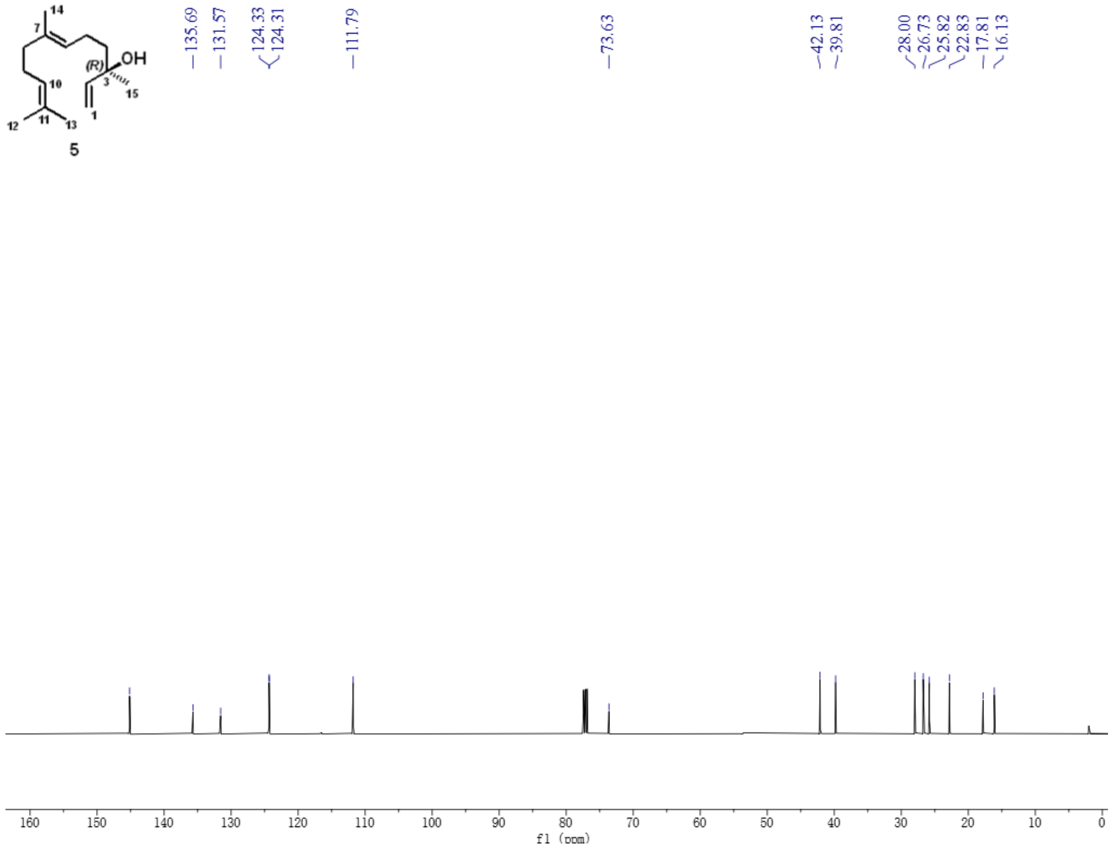


Figure S41. ^{13}C NMR spectrum (125 MHz) of 5 in CDCl$_3$, related to Figure 4 and Table S3.

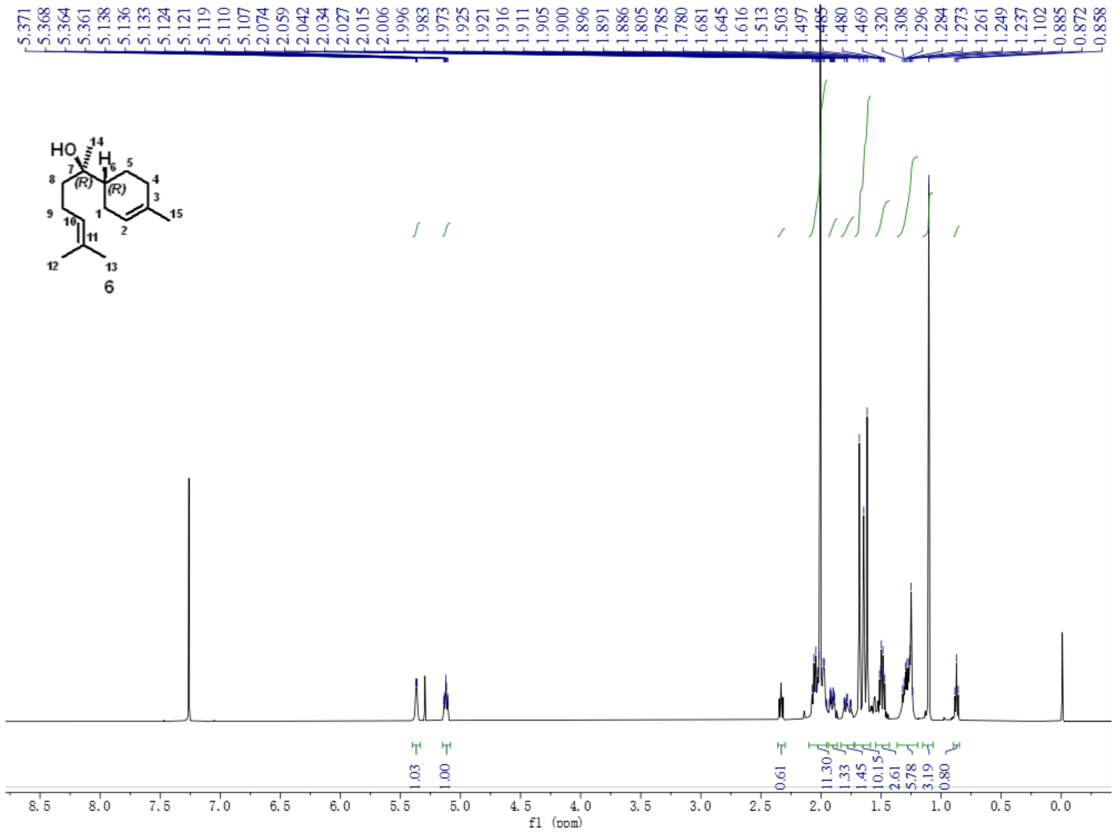


Figure S42. ^{1}H NMR spectrum (500 MHz) of 6 in CDCl$_3$, related to Figure 4 and Table S3.

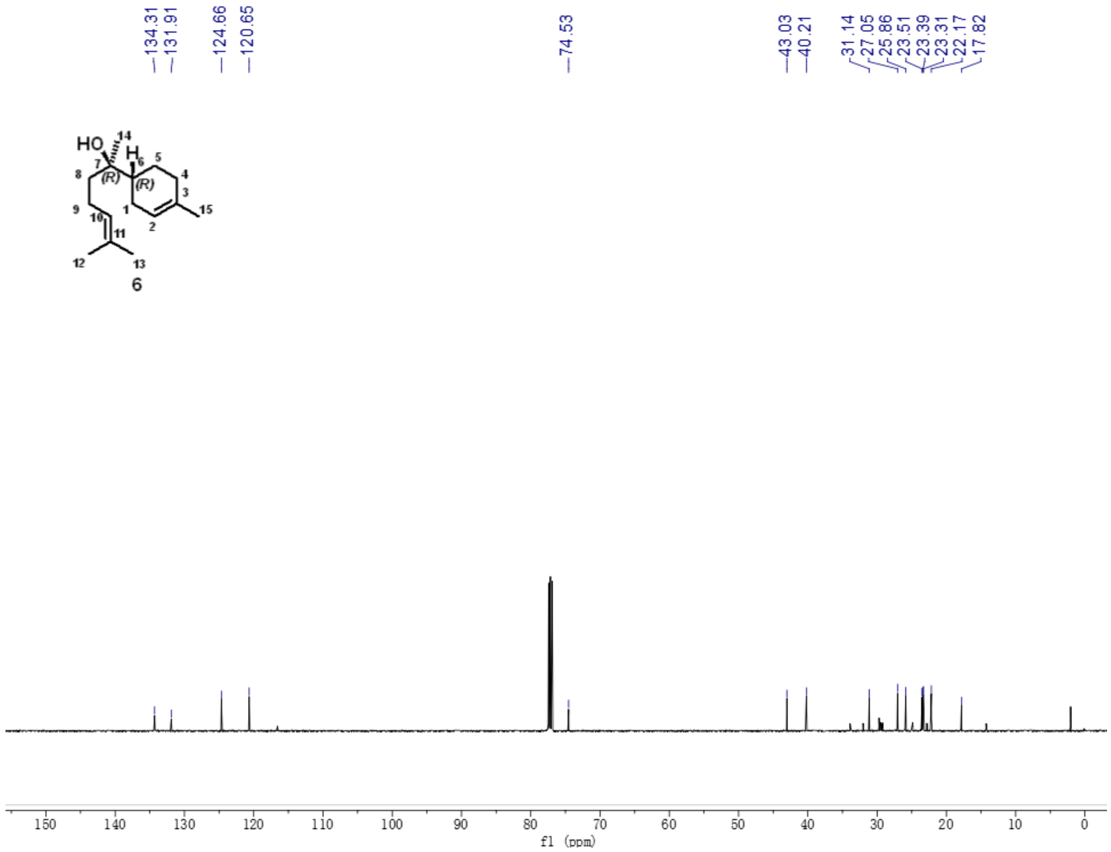


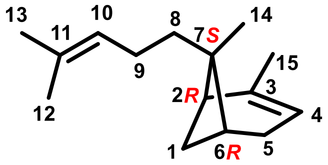
Figure S43. ^{13}C NMR spectrum (125 MHz) of 6 in $CDCl_3$, related to Figure 4 and Table S3.

Supplementary Tables

Table S1. Crystal data and structure refinement for 1, related to Figure 1.

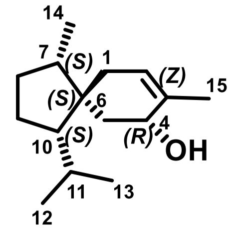
Identification code	206_3_auto
Empirical formula	$C_{15}H_{20}O_4$
Formula weight	264.31
Temperature/K	99.9(3)
Crystal system	triclinic
Space group	P1
a/Å	7.22070 (10)
b/Å	7.6941 (2)
c/Å	13.1723 (3)
α/°	93.095 (2)
β/°	99.137 (2)
γ/°	94.224 (2)
Volume/Å3	718.96 (3)
Z	2
ρ_{calc}g/cm3	1.221
μ/mm^{-1}	0.717
F(000)	284.0
Crystal size/mm3	0.11 × 0.09 × 0.03
Radiation	Cu Kα (λ = 1.54184)
2Θ range for data collection/°	6.812 to 148.786
Index ranges	$-8 \leqslant h \leqslant 8, -9 \leqslant k \leqslant 9, -16 \leqslant l \leqslant 15$
Reflections collected	26108
Independent reflections	5336 [R_{int} = 0.0238, R_{sigma} = 0.0159]
Data/restraints/parameters	5336/3/352
Goodness-of-fit on F2	1.074
Final R indexes [I>=2σ (I)]	R1 = 0.0325, wR2 = 0.0889
Final R indexes [all data]	R1 = 0.0334, wR2 = 0.0895
Largest diff. peak/hole / e Å^{-3}	0.27/-0.15
Flack parameter	0.01(6)

Table S2. The ^{1}H NMR (500 MHz) and ^{13}C NMR (125 MHz) data of 3 in $CDCl_3$, related to Figure 2, Figures S35–37.

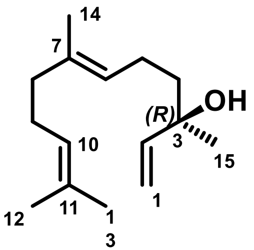


No	δ_H mult. (*J* in Hz)	δ_C
1a	1.17, d (8.5)	31.7
1b	2.31, m	
2	2.01, m	45.5
3		144.7
4	5.20, brs	116.6
5	2.13, m, 2.25, m	31.4
6	2.15, m	39.1
7		41.3
8	1.66, m (2H)	38.8
9	1.98, m (2H)	23.89
10	5.17, t (6.0)	125.4
11		131.2
12	1.63, s (3H)	17.7
13	1.70, s (3H)	25.9
14	0.83, s (3H)	17.6
15	1.66, s (3H)	23.2

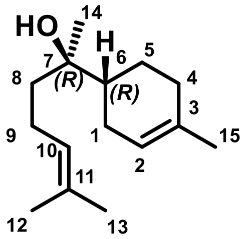
Table S3. The ^{13}C NMR (125 MHz) data of 4–6 in $CDCl_3$, related to Figure 2 and Figure 4.



4



5



6

No	4	5	6
1	35.4	111.8	27.0
2	125.3	145.2	120.6
3	135.7	73.6	134.3
4	68.9	42.1	31.1
5	32.4	26.7	22.2
6	45.1	124.3	43.0
7	46.8	135.7	74.5
8	29.0	39.8	40.2
9	26.7	28.0	23.3
10	60.0	124.4	124.7
11	30.4	131.6	131.9
12	23.5	16.1	17.8
13	23.2	22.8	25.9
14	14.4	17.8	23.5
15	19.1	25.8	23.4

Table S4. Homologues and conserved domains of the proteins in the *nec* cluster, related to Figure 2.

Protein	Size (aa)	homology (swissprot)	Identities/ Positives	Conserved domains
NecA	358	Quinone oxidoreductase PIG3	36/52 %	PIG3 p53-inducible quinone oxidoreductase
NecB	525	Tryprostatin B 6-hydroxylase	43/61 %	CYP67-like (hydroxylation)
NecC	494	CYP450 monooxygenase AMT3	38/60 %	CYP60B-like (hydroxylation)
NecD (NsBERS)	435	Terpene cyclase aneC	24/44 %	Non-plant terpene cyclases
NecE	520	*Ent*-kaurene oxidase	29/51 %	CYP503A1-like (three successive oxidations / demethylation)

Table S5. Predicted functions and conserved domains of the proteins in the *lach* cluster, related to Figure 2.

Protein	Accession number	Size(aa)	Predicted functions	Conserved domains
LachA	TVY81922.1	355	NAD(P)H quinone oxidoreductase	PIG3 p53-inducible quinone oxidoreductase
LachB (LsBERS)	TVY81921.1	444	Terpene Synthase	Non-plant terpene cyclases
LachC	-	442	Cytochrome P450	CYP503A1-like (three successive oxidations / demethylation)
LachD	TVY81920.1	537	Cytochrome P450	CYP67-like (hydroxylation)
LachE	TVY81918.1	384	Cytochrome P450	CYP503A1-like (three successive oxidations / demethylation)

Table S6. Homology search of NsBERS in NCBI, related to Figure 3.

Description	Strain	coverage	Identity	Accession
BcSTC4, sesquiterpene cyclase	*Metarhizium majus* ARSEF 297	91%	65.95%	KID93366.1
BcSTC4, sesquiterpene cyclase	*Metarhizium guizhouense* ARSEF 977	91%	65.78%	KID81896.1
hypothetical protein LSUE1_G003745	*Lachnellula suecica*	91%	62.09%	TVY81921.1
hypothetical protein EAE99_001473	*Botrytis elliptica*	92%	56.62%	KAF7939668.1
putative sesquiterpene cyclase protein	*Botrytis cinerea* BcDW1	93%	54.74%	EMR81374.1
BcSTC4, similar to sesquiterpene cyclase	*Botrytis cinerea* T4	93%	54.74%	CCD49915.2
hypothetical protein GJ744_011298	*Endocarpon pusillum*	91%	56.78%	KAF7513032.1
Bcstc4	*Botrytis cinerea* B05.10	93%	54.50%	XP_001546971.2
uncharacterized protein EAE98_003830	*Botrytis deweyae*	92%	55.88%	XP_038811923.1
hypothetical protein EV356DRAFT_562544	*Viridothelium virens*	89%	55.36%	KAF2228525.1
hypothetical protein COCHEDRAFT_1083047	*Bipolaris maydis* C5	88%	55.93%	EMD94346.1
hypothetical protein COCC4DRAFT_58253	*Bipolaris maydis* ATCC 48331	88%	55.78%	XP_014081267.1
hypothetical protein COCMIDRAFT_37699	*Bipolaris oryzae* ATCC 44560	84%	56.60%	XP_007689029.1
uncharacterized protein EAF02_006039	*Botrytis sinoallii*	98%	53.01%	XP_038758304.1
hypothetical protein BELL_0648g00110	*Botrytis elliptica*	92%	54.41%	TGO70911.1
hypothetical protein EAF03_005474	*Botryotinia squamosa*	92%	54.17%	KAF7866352.1
hypothetical protein BGAL_0106g00160	*Botrytis galanthina*	87%	52.58%	THV51576.1
hypothetical protein IFR05_015310	*Cadophora* sp. M221	99%	45.49%	KAG4429207.1
hypothetical protein KY384_006558	*Bacidia gigantensis*	88%	48.97%	KAG8528869.1
putative sesquiterpene cyclase protein	*Botrytis fragariae*	64%	60.92%	XP_037188555.1
hypothetical protein BOTCAL_0446g00010	*Botryotinia calthae*	79%	52.11%	TEY39953.1

Table S7. Primers used for this study, related to STAR Methods.

Primer name	Primer sequence (5’→3’)
pYTU-Nec04-F	CTTCATCCCCAGCATCATTACACCTCAGCAATGGCTTCCGTCAGCACC
pYTU-Nec04-R	GGGATCCACTAGTTCTAGAGCGGCCGCCTCAGCTCGGGGGTCGGGAATAATTC
pYTU-NecD-F	CTTCATCCCCAGCATCATTACACCTCAGCAATGAAAAGCTTCCTGTACTGG
pYTU-NecD-R	GGGATCCACTAGTTCTAGAGCGGCCGCCCCTGACGATTCCATCGAG
pYTU-Nec26-F	CTTCATCCCCAGCATCATTACACCTCAGCAATGTATAACACGGGGAGCC
pYTU-Nec26-R	GGGATCCACTAGTTCTAGAGCGGCCGCCGAGCAACATGCAAGGGAGAC
U-F	GAAGTGGTTGGTCGGGGTTC
U-R	CTGACTTCAACACAGTGG
pESC-URA-NsBERS-F	CAAGGAGAAAAAACCCCGGATCCATGAAAAGCTTCCTGTACTGG
pESC-URA-NsBERS-R	GAAATCAACTTCTGTTCCATGTCGACTTACCACGAGGGGTCAATAG
pET28a-MBP-NsBERS-F	GCCATCATCATCATCATCACGGATCCATGAAAAGCTTCCTGTACTGG
pET28a-MBP-NsBERS-R	GTGGTGGTGCTCGAGTGCGGCCGCTTACCACGAGGGGTCAATAG
pET28a-MBP-LsBERS-F	GCCATCATCATCATCATCACGGATCCATGAGTCTGCCCAGCTTC
pET28a-MBP-LsBERS-R	GTGGTGGTGCTCGAGTGCGGCCGCTCAACTAGTGTACTGTGG
pET28a-BcBOS-F	CAGCAAATGGGTCGCGGATCCATGGCTCAAATTATGACTTCAGCTG
pET28a-BcBOS-R	GTGGTGGTGCTCGAGTGCGGCCGCCTACGTCAGCTTGGCAG

Table S8. Plasmids used for this study, related to STAR Methods.

Plasmid name	**Genes**	**Aim**
pYTU-Nec04	*nec04* from gDNA	*A. nidulans* overexpression
pYTU-NecD	*necD* from gDNA	*A. nidulans* overexpression
pYTU-Nec26	*nec26* from gDNA	*A. nidulans* overexpression
pESC-URA-NsBERS	*necD* from cDNA	*S. cerevisiae* overexpression
pESC-URA-LsBERS	*lachB* from synthesized sequence	*S. cerevisiae* overexpression
pESC-URA-BcBOS	*bcbos* from synthesized sequence	*S. cerevisiae* overexpression
pET28a-MBP-NsBERS	*necD* from cDNA	*E. coli* overexpression
pET28a-MBP-LsBERS	*lachB* from synthesized sequence	*E. coli* overexpression
pET28a-BcBOS	*bcbos* from synthesized sequence	*E. coli* overexpression