Structural elucidation, total synthesis and cytotoxic activity of effphenol A

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Contents

Abbreviations

Ac	Acetyl
HOAc	Acetic acid
ACN	Acetonitrile
DCM	Dichloromethane
THF	Tetrahydrofuran
TFA	Trifluoroacetic acid
PTSA	*p*-Toluenesulfonic acid
PPTS	Pyridinium *p*-toluenesulfonate
MSA	Methylsulphonic acid
HRMS	High resolution mass spectroscopy
NMR	Nuclear magnetic resonance
HPLC	High performance liquid chromatography
rt	Room temperature
ESI	Electron spray ionization
TLC	Thin layer chromatography

1. Experimental Section

1.1 General experimental procedures

HRESIMS were collected on a Thermo MAT95XP (Thermo Fisher Scientific, Bremen, Germany). NMR spectra were acquired by a Bruker Avance-600 spectrometer (Bruker, Fällanden, Switzerland). Circular dichroism (CD) spectra were afforded under N_2 gas by a Jasco 820 spectropolarimeter (Jasco Corporation, Kyoto, Japan). Optical rotations were obtained by an Anton Paar MCP-500 spectropolarimeter (Anton Paar, Graz, Austria). UV spectra were acquired using a Shimadzu UV-2600 spectrophotometer (Shimadzu, Kyoto, Japan). IR data were done with a Shimadzu IR Affinity-1 spectrometer (Shimadzu, Kyoto, Japan). Preparative HPLC was performed using a YMC-pack ODS-A column (250 × 20 mm, 5 μm, 12 nm, YMC Co., Ltd, Kyoto, Japan). A YMC-pack ODS-A/AQ column (250 × 10 mm, 5 μm, 12 nm, YMC CO., Ltd, Kyoto, Japan) was used for semipreparative HPLC separation and the CHIRALPAK IC column (250 × 10 mm, 5 μm) column for chiral semipreparative HPLC separation. Silica gel (100-200 mesh and 200-300 mesh, Qingdao Marine Chemical Inc., Qingdao, China), C_{18} reversed-phase silica gel (40–63 μm, Merck, Darmstadt, Germany) and Sephadex LH-20 gel (Pharmacia Fine Chemical Co. Ltd., Uppsala, Sweden) were used in the chromatography processes. Fractions were monitored by TLC and spots were detected on heated TLC plates (silica gel GF_{254} plates, Qingdao Marine Chemical Inc., Qingdao, China) with 10% H_2SO_4 in EtOH under UV light.

1.2 Fungal material and identification

The strain FS524 used in this work was isolated from a sediment sample, which was collected at the depth of 1428 m in the South China Sea (110°59′04″ E, 18°00′47″ N) in June 2017. The sequence data for this strain have been submitted to the GenBank under accession No. MN545626. By using BLAST (nucleotide sequence comparison program) to search the GenBank database, FS524 has 99.8% similarity to *Trichobotrys effuse* DFFSCS021 (Accession No. JX156367). And the strain was

preserved at the Guangdong Provincial Key Laboratory of Microbial Culture Collection and Application, Guangdong Institute of Microbiology.

1.3 Fermentation, extraction and isolation

The marine fungus T. *effuse* FS524 was cultured on potato dextrose agar (PDA) at 28℃ for 7 days to prepare the seed culture, and then inoculated into flasks (3 L) containing 9 g sea salts, 250 g of rices and 300 mL of waters. After that, all flasks were incubated at 28 °C for one month and the fermented rice substrate was extracted repeatedly with EtOAc. After the evaporation of the solvent, a dark brown solid (67.3) was obtained. The crude extract was fractionated by silica gel column chromatography (100-200 mesh) with two gradient systems of increasing polarity (petroleum ether/EtOAc, 30:1 → 1:1; CH_2Cl_2/CH_3OH, 10:1 → 1:1) to furnish nine fractions (A-I).

Fraction G (11.1 g) was subjected to C-18 reversed-phase silica gel CC (gradient elution with MeOH-H_2O, 30:70 → 100:0) to afford seven subfractions (G1-G7). Compound **1** (10.6 mg, t_R = 33.5 min) was obtained from G5 followed by semi-preparative HPLC (MeCN-H_2O, 50:50, 2 mL/min) and semi-preparative HPLC equipped with a chiral column (MeCN-H_2O, 45:55, 2 mL/min), respectively.

Effphenol A (**1**): dark red crystal. UV (MeOH) λ_{max} (log ε): 207 (4.58), 319 (4.08) nm. IR ν_{max}: 3361, 1614, 1435, 1236, 1099, 835 cm^{-1}. ^1H (600 MHz) and ^{13}C (150 MHz) NMR spectral data, see Table 1. HRESIMS: *m*/*z* 451.1739 $[M + H]^+$ (calcd for $C_{26}H_{27}O_7$, 451.1751).

1.4 X-ray crystallographic analysis of compound **1**

The single-crystal X-ray diffraction data for compound **1** was collected on Agilent Xcalibur Nova single-crystal diffractometer using CuKα radiation at 99.9 K. The crystal structure was refined by full-matrix least-squares calculation. Crystallographic Data have been deposited at the Cambridge Crystallographic Data Center with the deposition number of CDCC 1974672 for **1**. Copies of these data can be obtained free

of charge via www.ccdc.cam.au.ck/conts/retrieving.html.

Table S1. X-ray crystallographic data for **1**

Empirical formula	$C_{26}H_{28}O_8$
Formula weight	468.48
Temperature/K	99.9(9)
Crystal system	triclinic
Space group	P-1
a/Å	8.3229(2)
b/Å	10.5201(2)
c/Å	13.8336(3)
α/°	88.816(2)
β/°	73.989(2)
γ/°	76.976(2)
Volume/Å^3	1133.19(4)
Z	2
ρ_{calc} g/cm^3	1.373
μ/mm^{-1}	0.845
F(000)	496.0
Crystal size/mm^3	0.07 × 0.05 × 0.04
Radiation	CuKα (λ = 1.54184)
2Θ range for data collection/°	8.636 to 124.066
Index ranges	$-9 \le h \le 9$, $-11 \le k \le 11$, $-15 \le l \le 11$
Reflections collected	9494
Independent reflections	3468 [R_{int} = 0.0239, R_{sigma} = 0.0286]
Data/restraints/parameters	3468/0/321
Goodness-of-fit on F^2	0.931
Final R indexes [I>=2σ (I)]	R_1 = 0.0353, wR_2 = 0.0914
Final R indexes [all data]	R_1 = 0.0414, wR_2 = 0.0952
Largest diff. peak/hole / e Å^{-3}	0.20/-0.21

1.5 Cytotoxic activity assay

Compound **1** was evaluated for their cytotoxic activity against SF-268, MCF-7, HepG-2, A549 cell lines by using the SRB method[1]. The cells (180 μL) with a density of 3×10^4 cells/mL of media on 96-well plate were put under 37 °C at 5% CO_2 condition and incubated for 24 h. Then, 20 μL of various concentrations of compounds were added and further incubated for 72 h. After that, the cell monolayers were fixed by 50% (wt/v) trichloroacetic acid (50 μL) and stained for 30 min by 0.4% (wt/v) SRB, which was dissolved in 1% acetic acid. The unbound dye was removed by washing repeatedly with 1% acetic acid, and then dissolved into the protein-bound dye in 10 mM Tris base solution (200 μL) for OD determination at 570 nm using a microplate reader. Cisplatin was used as a positive control possessing potent cytotoxic activity. All data were obtained in triplicate, and the IC_{50} values were calculated by the SigmaPlot 10.0 software (Systat Software Inc., San Jose, California, America) with the use of a non-linear curve-fitting method. The SF-268, MCF-7, HepG-2, A549 cell lines were purchased by the Chinese Academy of Sciences Cell Bank.

References

[1] P. Skehan, R. Storeng, D. Scudiero, A. Monks, J. McMahon, D. Vistica, J. T. Warren, H. Bokesch, S. Kenney and M. R. Boyd, *J. Natl. Cancer. Inst.*, 1990, **82**, 1107–1112.

2. Experimental Procedures

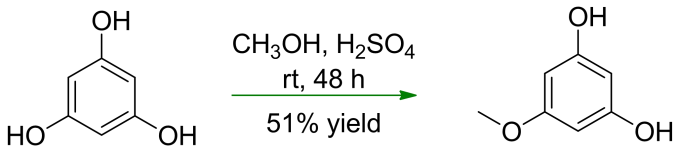
2.1 General experimental procedures

All reactions were conducted in a nitrogen atmosphere with dry solvents under anhydrous conditions, unless otherwise noted. Reagents were purchased at high commercial quality, and used without further purification. Thin-layer chromatography (TLC) was conducted with 0.25 mm Tsingdao silica gel plates (60F-254) and visualized by exposure to UV light (254 nm) or stained with potassium permanganate. Silica gel (ZCX-II, 200-300 mesh) used for flash column chromatography was purchased from Qing Dao Hai Yang Chemical Industry Co. of China. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Brüker Advance 500 (^{1}H: 500 MHz, ^{13}C: 125 MHz). Chemical shifts reported in parts per million relative to $CDCl_3$ (^{1}H NMR: 7.27

ppm, ^{13}C NMR: 77.00 ppm), CD_3COCD_3-d_6 (^1H NMR: 2.05 ppm, ^{13}C NMR: 30.0, 206.3 ppm), CD_3OD-d_4 (^1H NMR: 3.33 ppm, ^{13}C NMR: 47.5 ppm), and DMSO-d_6 (^1H NMR: 2.50 ppm, ^{13}C NMR: 39.5 ppm). Mass spectrometric data were obtained using ABI-Q Star Elite high resolution mass spectrometer. Anhydrous THF was distilled from sodium-benzophenone until a deep blue color persisted, CH_2Cl_2 (DCM) was distilled from calcium hydride. Yields referred to chromatographically purified products unless otherwise stated. The following abbreviations were used to explain the multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, brs = broad singlet.

2.2 The synthetic procedures

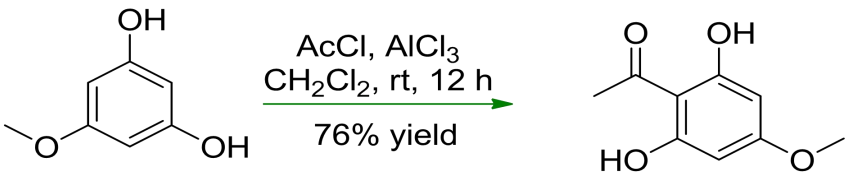
*1.4.1 The Synthesis of 5-Methoxybenzene-1,3-diol (**9**)*



8 **9**

Phloroglucinol (1.26 g, 10 mmol) was slowly dissolved in anhydrous methanol (20 mL) at room temperature. To this clear solution, concentrated H_2SO_4 (2 mL) was added carefully, and the mixture was stirred at room temperature for 12 h. Then, the crude mixture was quenched with water (50 mL), extracted with $CHCl_3$ (5 × 60 mL), washed with brine, and concentrated *in vacuum*. The crude product was purified by flash column chromatography (silica gel, hexane : EtOAc = 2 : l) to provide **9** (714 mg, 5.1 mmol, 51% yield) as yellow rod-like crystals. **9**: ^1H NMR (500 MHz, $CDCl_3$): δ_H 5.90 (s, 3H), 3.70 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 161.6, 158.7, 95.1, 92.7, 54.0. The known compound **9** was prepared according to the literature (*Chem. Comm*. 2019, 55(66), 9837-9840), and the NMR data were also agree with the reported ones.

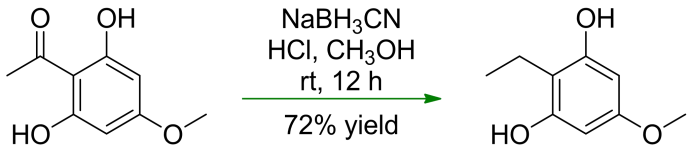
*1.4.2 The Synthesis of 1-(2,6-Dihydroxy-4-methoxyphenyl)ethanone (**10**)*



9 **10**

To a solution of phloroglucinol **9** (560 mg, 4 mmol) in CH_2Cl_2 (15 mL) was added $AlCl_3$ (1.06 g, 8 mmol) at 0 °C. The mixture was stirred at 0 °C for 15 mins, and the AcCl (460 mg, 6 mmol) was added in one portion. The mixture was stirred at room temperature under nitrogen atmosphere for 12 h. After most of the start material was consumed, the mixture was poured into the ice water (25 mL). Then, the mixture was extracted with EtOAc (3 × 25 mL), washed with brine, and concentrated *in vacuum*. The crude product was purified by flash chromatography (silica gel, hexane : EtOAc = 5 : l) to provide the product 1-(2,6-dihydroxy-4-methoxyphenyl) ethanone **10** (550 mg, 3.04 mmol, 76% yield) as a yellow-brown oil. **10**: ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 5.93 (s, 2H), 3.79 (s, 3H), 2.64 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 203.5, 166.1, 164.1, 104.8, 92.7, 54.4, 31.3. The NMR data of the known compound **10** was agree with those of reported data in the previous literature (*Eur. J. Med. Chem.* 2010, 45, 4258-4269).

*1.4.3 The Synthesis of 2-Ethyl-5-methoxybenzene-1,3-diol (**4**)*

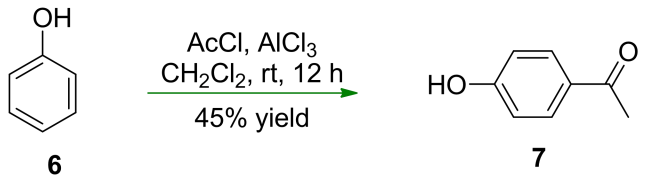


10 **4**

Acylphloroglucinol **10** (364 mg, 2.0 mmol) was dissolved in methanol (6 mL), and concentrated hydrochloric acid (0.1 mL) was then added. Then, $NaBH_3CN$ (628 mg, 10.0 mmol) was added slowly, and the resulting mixture was stirred for 1 h at room temperature. After most start material was disappeared, acetone (0.5 mL) was added to quench the reaction. The resulting reaction mixture was extracted with EtOAc (3 × 25 mL), washed with brine, and concentrated *in vacuum*. The crude product was further purified by 15 cm long flash chromatography (silica gel, hexane :

EtOAc = 3 : l) to afford the desired product **4** (249 mg, 1.48 mmol, 72% yield) as a yellowish solid. **4**: ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 6.03 (s, 2H), 4.77 (s, 2H), 3.75 (s, 3H), 2.60 (q, J = 7.6 Hz, 2H), 1.17 (t, J = 7.6 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$): δ_H 158.8, 154.9, 109.1, 94.4, 55.3, 16.0, 13.9. The NMR data of the known compound **4** was agree with those of reported data in the previous literature (*Chem. Ber*. 1984, 117(11), 3270-3279.)

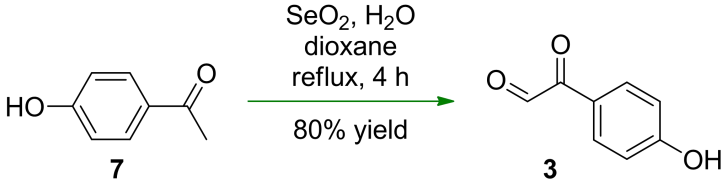
1.4.4 The Synthesis of 2-Ethyl-5-methoxybenzene-1,3-diol (7)



6 **7**

A 50 mL flame-dried flask was charged with 10 mL CH_2Cl_2, phenol **6** (940 mg, 10 mmol) and $AlCl_3$ (2.66 g, 20 mmol) were added. After stirring for 15 mins at 0 ^{o}C, AcCl (870 mg, 11 mmol) was added dropwised at 0 ^{o}C over 3 mins. The mixture was stirred vigorously for 12 h at room temperature. Then, the mixture was poured into ice water (20 mL) and extracted with $CHCl_3$ (3 × 20 mL). The combined organic phases were washed with brine (30 mL), dried over Na_2SO_4, and filtered. Removal of solvent by rotary evaporation and purification by flash column chromatography (silica gel, hexane : EtOAc = 10 : l) afforded **7** (612 mg, 4.5 mmol, 45% yield) as a white powder. **7**: ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 7.93 (m, 2H), 6.92 (m, 2H), 6.11 (brs, 1H), 2.58 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 197.2, 160.3, 131.0, 130.4, 115.3, 26.3. The NMR data of the known compound **7** was agree with those of reported data in the previous literature (*J. Chin. Chern. Soc.*, 2000, 47(2), 381-388.)

*1.4.5 The Synthesis of 2-(4-Hydroxyphenyl)-2-oxoacetaldehyde (**3**)*

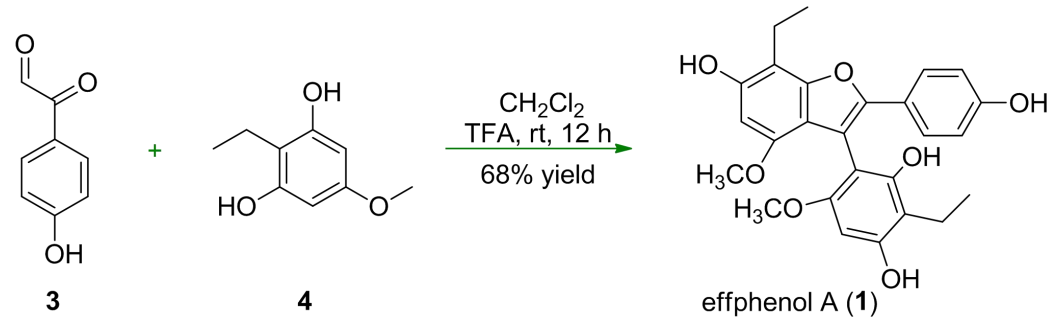


7 **3**

SeO_2 (1.65 g, 15 mmol), 1,4-dioxane (9 mL) and water (1 mL) were added to a

20 mL three-necked bottle and fitted it with an condenser. The mixture was heated to 50 °C and stirred until the solid dissolved. It was followed by addition of **7** (1.36 g, 10 mmol) and the reaction was maintained at reflux temperature. After the reaction was complete, as monitored by TLC, the mixture was filtered through a pad of celite. The filtrate was evaporated to afford a crude product, and it was further purified by flash column chromatography (silica gel, hexane : EtOAc = 1 : l) afforded the crude product **3** as a yellow solid, which contained about 10% impurity and could be used in next step without further purification. Compound **3** was a known compound, and it was prepared according to the previous literature (*Tetrahedron*, 2016, 72(43), 6843-6853)

*1.4.5 The Synthesis of Effphenol A (**1**)*



The mixture of **3** (121 mg, 0.8 mmol), phloroglucinol **4** (420 mg, 2.5 mmol), TFA (0.3 mL) in THF (3 mL) was stirred at room temperature for 12 h. The reaction was quenched with 10 mL of water, extracted with ethyl acetate (3 × 10 mL). The combined organic phase was dried over anhydrous sodium sulfate, filtered and concentrated, the residue was purified by column chromatography on silica gel (hexane : EtOAc = 2 : l) to give 244 mg of effphenol A (**1**), yield 68%. ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 7.53 (d, J = 8.1 Hz, 2H), 6.76 (d, J = 8.7 Hz, 2H), 6.22 (s, 1H), 6.14 (s, 1H), 5.25 (s, 1H), 4.91 (s, 3H), 3.63 (s, 3H), 3.60 (s, 3H), 2.92 (d, J = 7.2 Hz, 2H), 2.67 (q, J = 7.5 Hz, 2H), 1.36 (t, J = 7.6 Hz, 3H), 1.17 (t, J = 7.5 Hz, 4H); ^{13}C NMR (125 MHz, $CDCl_3$): δ_H 156.8, 155.3, 154.5, 154.4, 152.9, 152.5, 151.4, 150.0, 127.0, 124.0, 115.4, 113.2, 109.0, 106.2, 104.7, 102.0, 95.2, 92.2, 55.9, 55.7, 16.6, 16.5, 14.1, 14.0.

The Comparison of ^{1}H and ^{13}C NMR Data of Natural and Synthetic Effphenol A (1)

The Comparison of ^{1}H NMR Data of Effphenol A (1)

Position	^{1}H chemical shift/δ ppm (Natural sample, 600 MHz)[a]	^{1}H Chemical shift /δ ppm (Synthetic sample, 500 MHz)[b]	$\Delta\delta$/ppm[c]
H-1	-	-	-
H-2	6.31 (s)	6.31	0.00
H-3	-	-	-
H-4	-	-	-
H-5	-	-	-
H-6	-	-	-
H-7	3.56 (s)	3.56 (s)	0.00
H-8	2.90 (m)	2.91 (m)	+0.01
H-9	1.30 (t, 7.5)	1.30 (t, 7.5)	0.00
H-10	-	-	-
H-11	-	-	-
H-12	-	-	-
H-13	7.50 (d, 8.8)	7.50 (d, 8.8)	0.00
H-14	6.76 (d, 8.8)	6.76 (d, 8.7)	0.00
H-15	-	-	-
H-16	6.76 (d, 8.8)	6.76 (d, 8.7)	0.00
H-17	7.50 (d, 8.8)	7.50 (d, 8.8)	0.00
H-18	-	-	-
H-19	-	-	-
H-20	-	-	-
H-21	-	-	-
H-22	6.20 (s)	6.20 (s)	0.00
H-23	-	-	-
H-24	2.67 (q, 7.3)	2.67 (q, 7.3)	0.00
H-25	1.11 (t, 7.5)	1.11 (t, 7.5)	0.00
H-26	3.53 (s)	3.53 (s)	0.00

[a]The ^{1}H NMR data were recorded on a Bruker Avance 600 spectrometer in CD_3COCD_3 and referenced against residual CH_3COCH_3 in CD_3COCD_3 as 2.05 ppm.

[b]The ^{1}H NMR data were recorded on a Bruker Avance 500 spectrometer in CD_3COCD_3 and referenced against residual CH_3COCH_3 in CD_3COCD_3 as 7.27 ppm.

[c]$\Delta\delta$/ppm refers the relative difference of each signal between the synthetic and natural samples.

The Comparison of ^{1}H and ^{13}C NMR Data of Natural and Synthetic Effphenol A (1)

The Comparison of ^{13}C NMR Data of Effphenol A (1)

Position	^{13}C chemical shift/δ ppm (Natural sample, 150 MHz)[a]	^{13}C Chemical shift /δ ppm (Synthetic sample, 125 MHz)[b]	$\Delta\delta$/ppm[c]
C-1	153.5	153.5	0.00
C-2	95.8	95.9	+0.01
C-3	153.5	153.5	0.00
C-4	106.7	106.8	+0.01
C-5	154.9	154.9	0.00
C-6	114.3	114.4	+0.01
C-7	56.0	56.1	+0.01
C-8	17.3	17.3	0.00
C-9	14.6	14.6	0.00
C-10	--*	--*	-
C-11	150.3	150.3	0.00
C-12	124.5	124.5	0.00
C-13	127.5	127.5	0.00
C-14	116.0	116.0	0.00
C-15	157.9	157.9	0.00
C-16	116.0	116.0	0.00
C-17	127.5	127.5	0.00
C-18	102.2	102.3	+0.01
C-19	155.3	155.4	+0.01
C-20	110.4	110.5	+0.01
C-21	156.6	156.6	0.00
C-22	92.3	92.4	+0.01
C-23	157.8	157.8	0.00
C-24	17.2	17.2	0.00
C-25	14.6	14.6	0.00
C-26	55.6	55.6	0.00

[a]The ^{13}C NMR data were recorded on a Bruker Avance 600 spectrometer in CD_3COCD_3 and referenced against residual CH_3COCH_3 in CD_3COCD_3 as 206.2 ppm.

[b]The ^{13}C NMR data were recorded on a Bruker Avance 500 spectrometer in CD_3COCD_3 and referenced against residual CH_3COCH_3 in CD_3COCD_3 as 206.2 ppm.

[c]$\Delta\delta$/ppm refers the relative difference of each signal between the synthetic and natural samples.

*not detected.

2. NMR, HRESIMS, CD, UV and IR spectrum of compound 1

Figure S1. ^{1}H NMR spectrum (600 MHz, CD_3COCD_3) of **1**

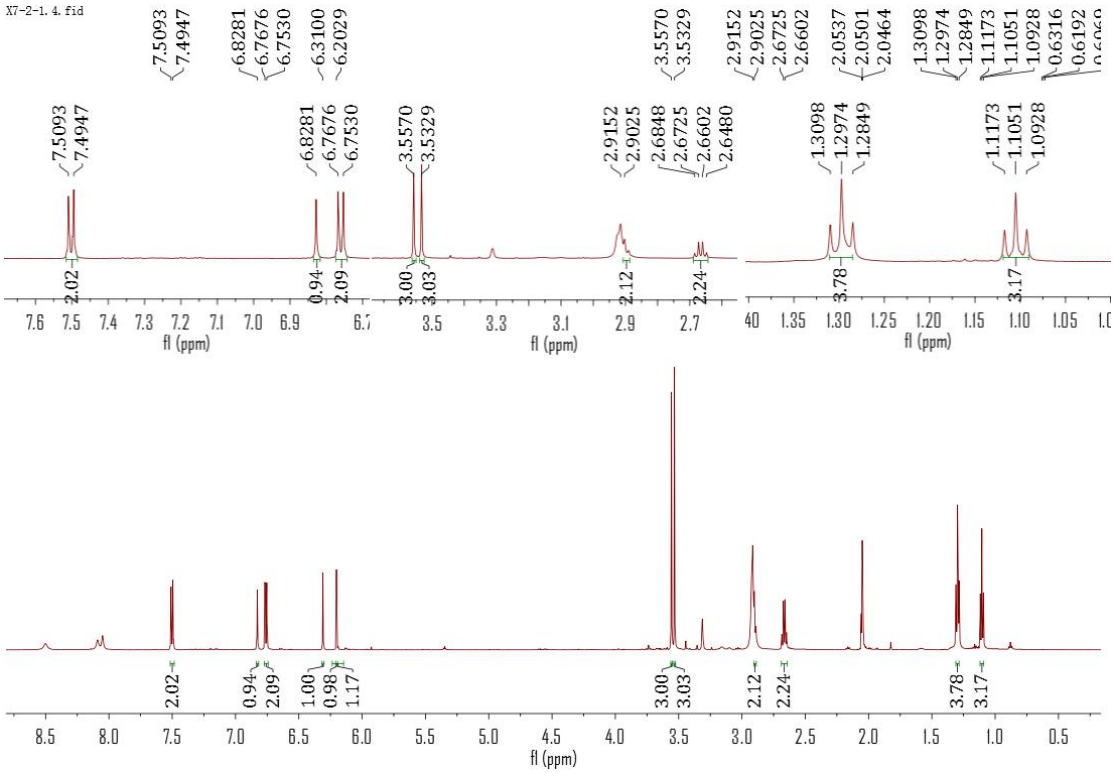


Figure S2. ^{13}C NMR spectrum (150 MHz, CD_3COCD_3) of **1**

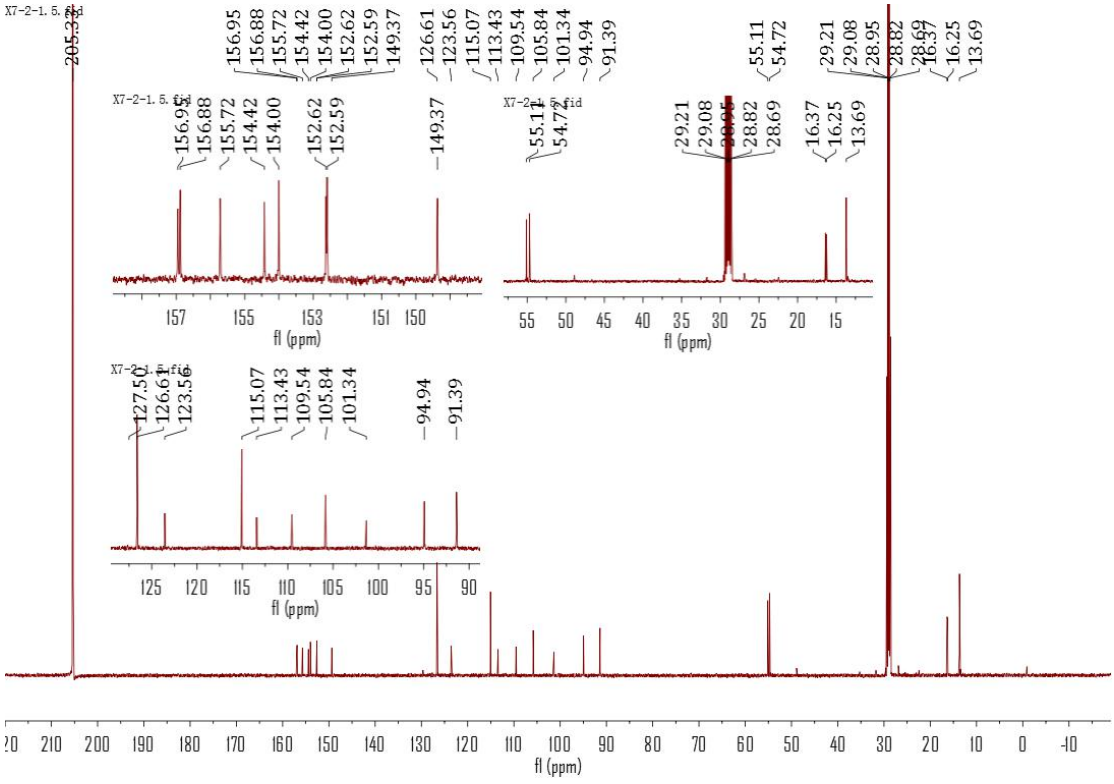


Figure S3. ^{1}H-^{1}H COSY spectrum of **1** in CD_3COCD_3

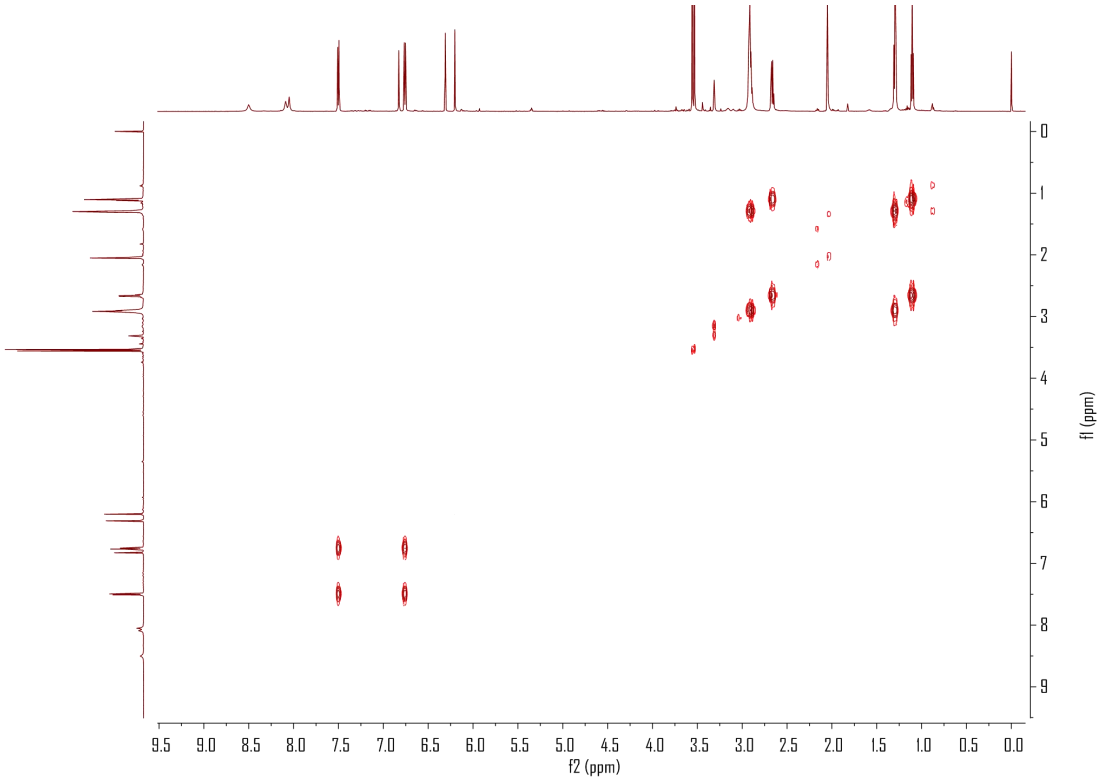


Figure S4. HSQC spectrum of **1** in CD_3COCD_3

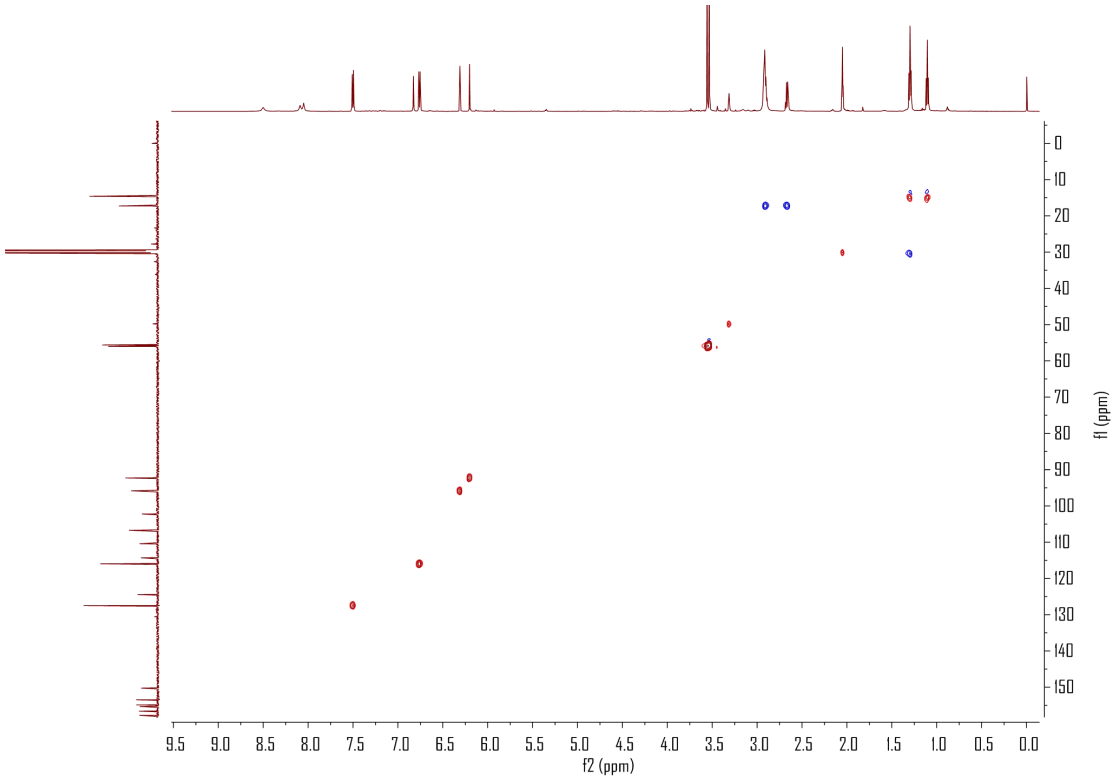


Figure S5. HMBC spectrum of **1** in CD_3COCD_3

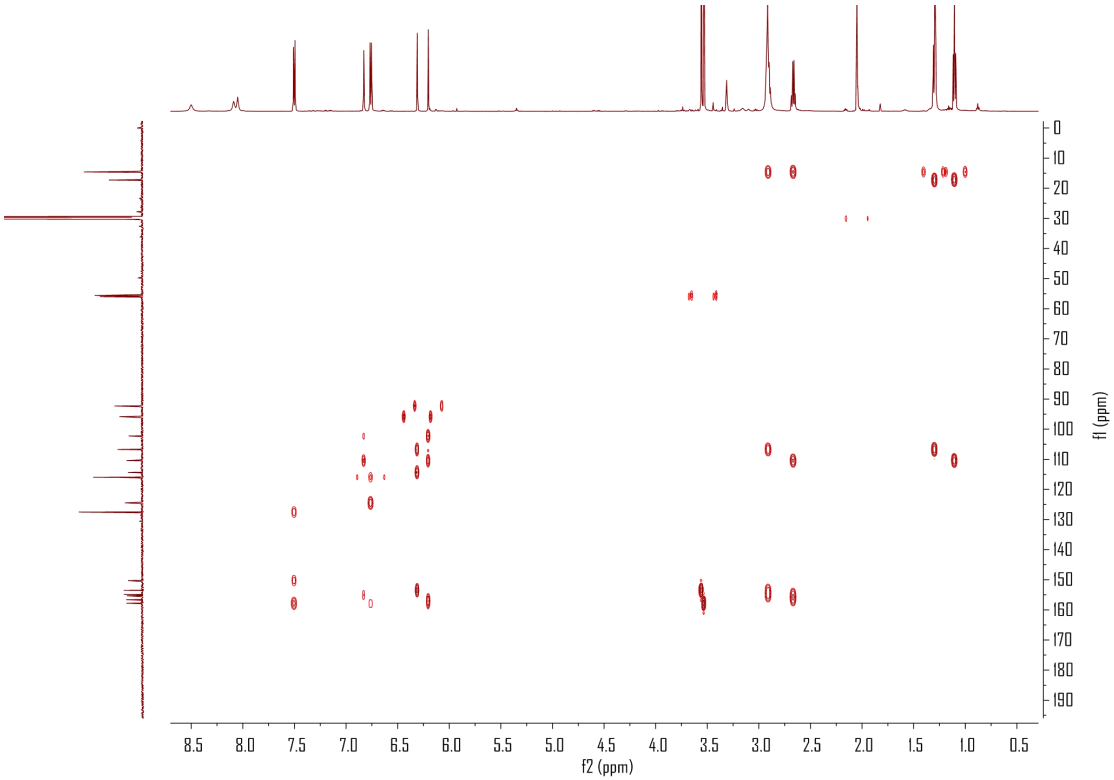
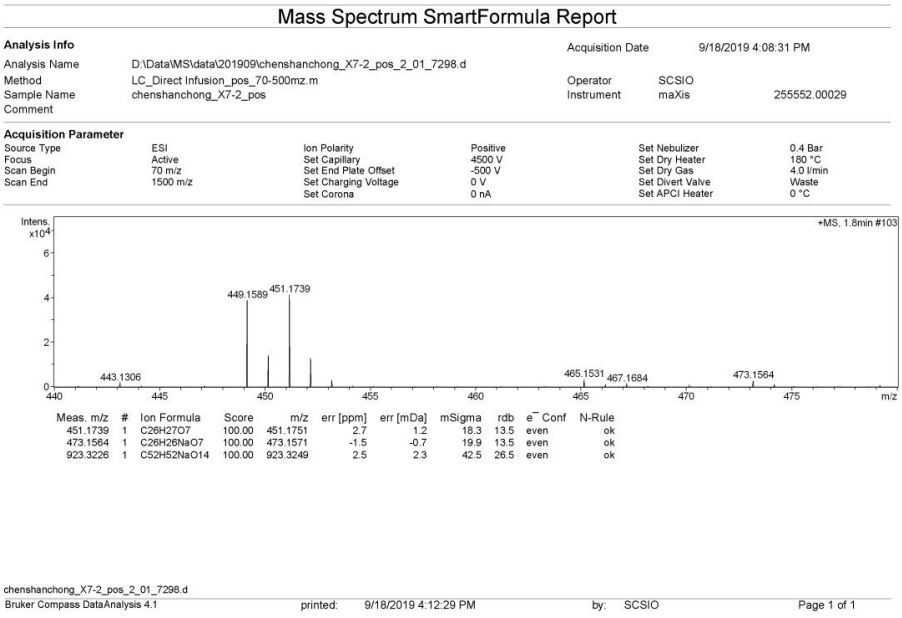


Figure S6. HRESIMS spectrum of **1**



Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name	D:\Data\MS\data\201909\chenshanchong_X7-2_pos_2_01_7298.d	Acquisition Date	9/18/2019 4:08:31 PM
Method	LC_Direct Infusion_pos_70-500mz.m	Operator	SCSIO
Sample Name	chenshanchong_X7-2_pos	Instrument	maXis 255552.00029
Comment			

Acquisition Parameter

Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Active	Set Capillary	4500 V	Set Dry Heater	180 °C
Scan Begin	70 m/z	Set End Plate Offset	-500 V	Set Dry Gas	4.0 l/min
Scan End	1500 m/z	Set Charging Voltage	0 V	Set Divert Valve	Waste
		Set Corona	0 nA	Set APCI Heater	0 °C

Meas. m/z	#	Ion Formula	Score	m/z	err [ppm]	err [mDa]	mSigma	rdb	e⁻ Conf	N-Rule
451.1739	1	C26H27O7	100.00	451.1751	2.7	1.2	18.3	13.5	even	ok
473.1564	1	C26H26NaO7	100.00	473.1571	-1.5	-0.7	19.9	13.5	even	ok
923.3226	1	C52H52NaO14	100.00	923.3249	2.5	2.3	42.5	26.5	even	ok

chenshanchong_X7-2_pos_2_01_7298.d
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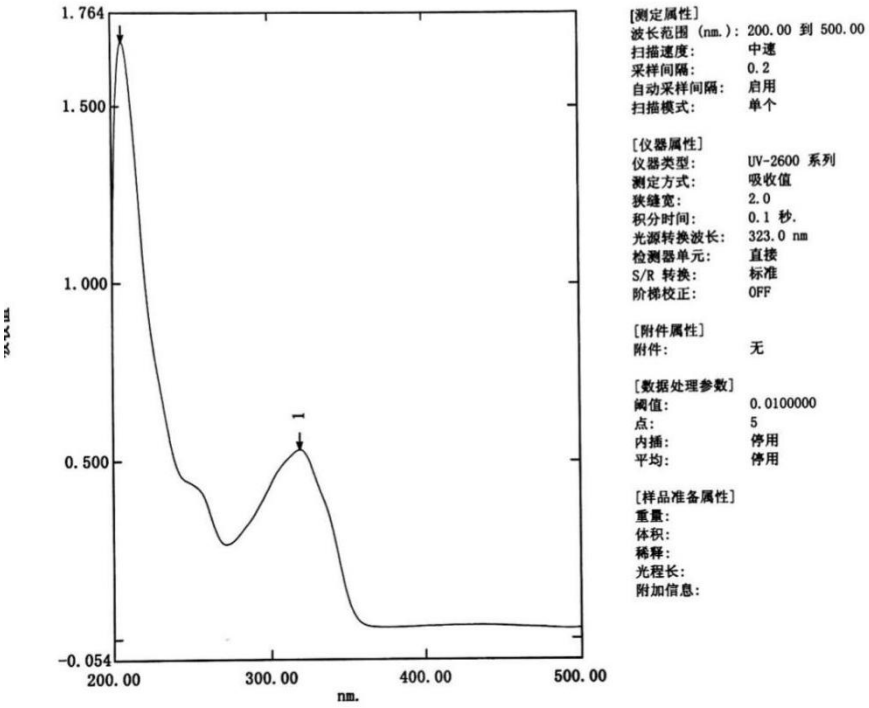
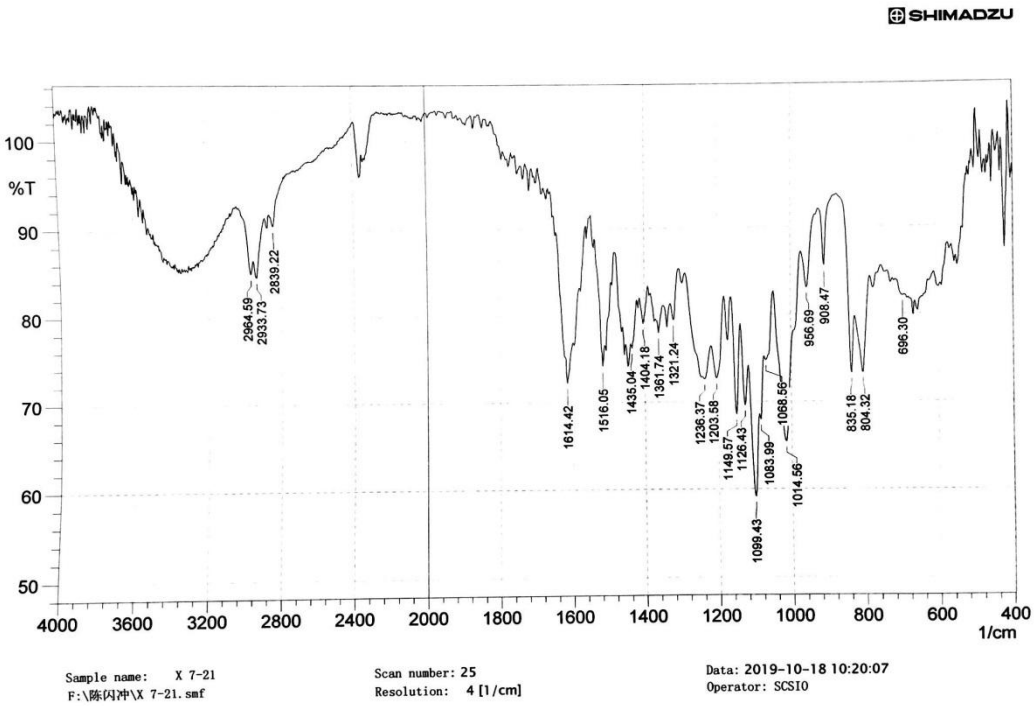
Figure S7. UV spectrum of **1**.



No.	P/V	波长(nm)	吸收值	描述
1	●	319.00	0.533	
2	●	206.80	1.681	

Figure S8. IR spectrum of **1**



5. NMR spectra for synthesis compounds

Figure S1. ^{1}H NMR spectrum (500 MHz, CD_3OD) of **9**

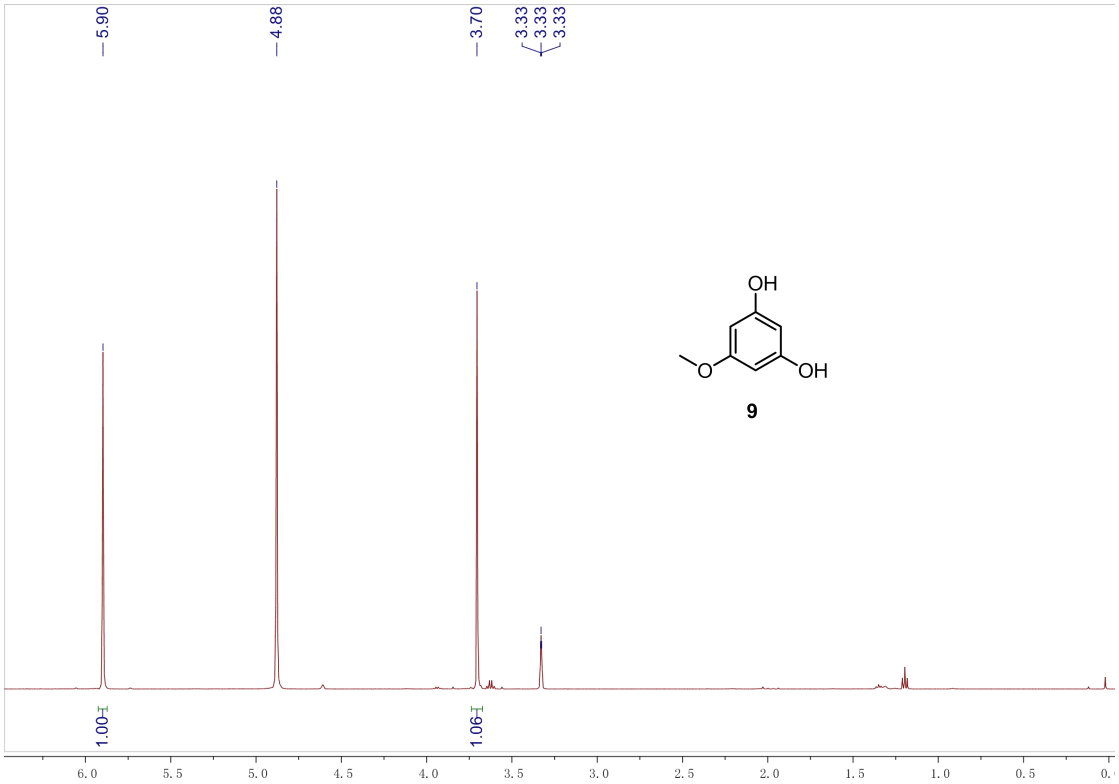


Figure S2. ^{13}C NMR spectrum (125 MHz, CD_3OD) of **9**

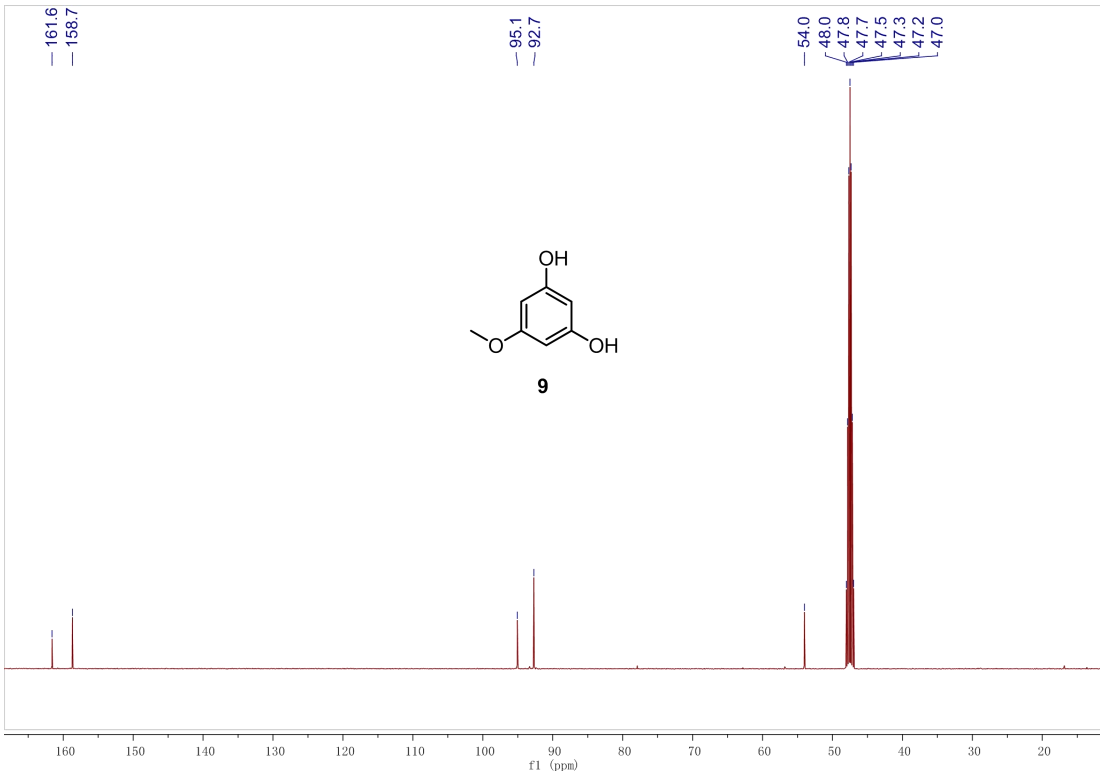


Figure S3. ^{1}H NMR spectrum (500 MHz, CD_3OD) of **10**

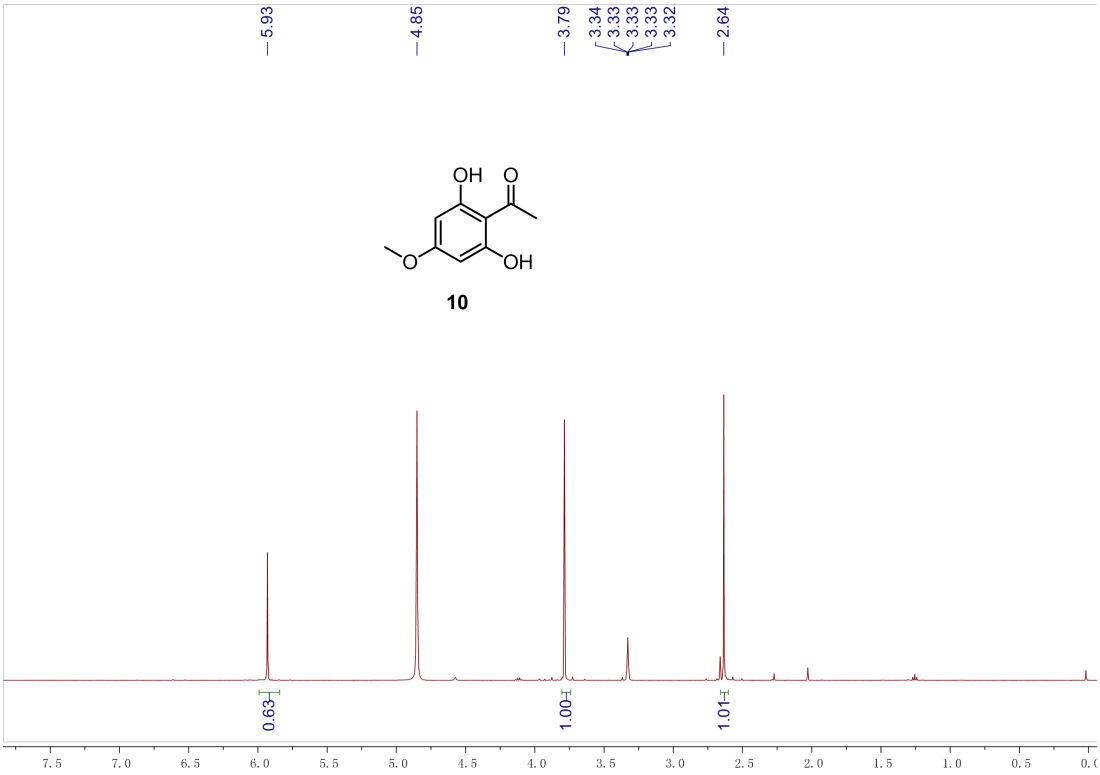


Figure S4. ^{13}C NMR spectrum (125 MHz, CD_3OD) of **10**

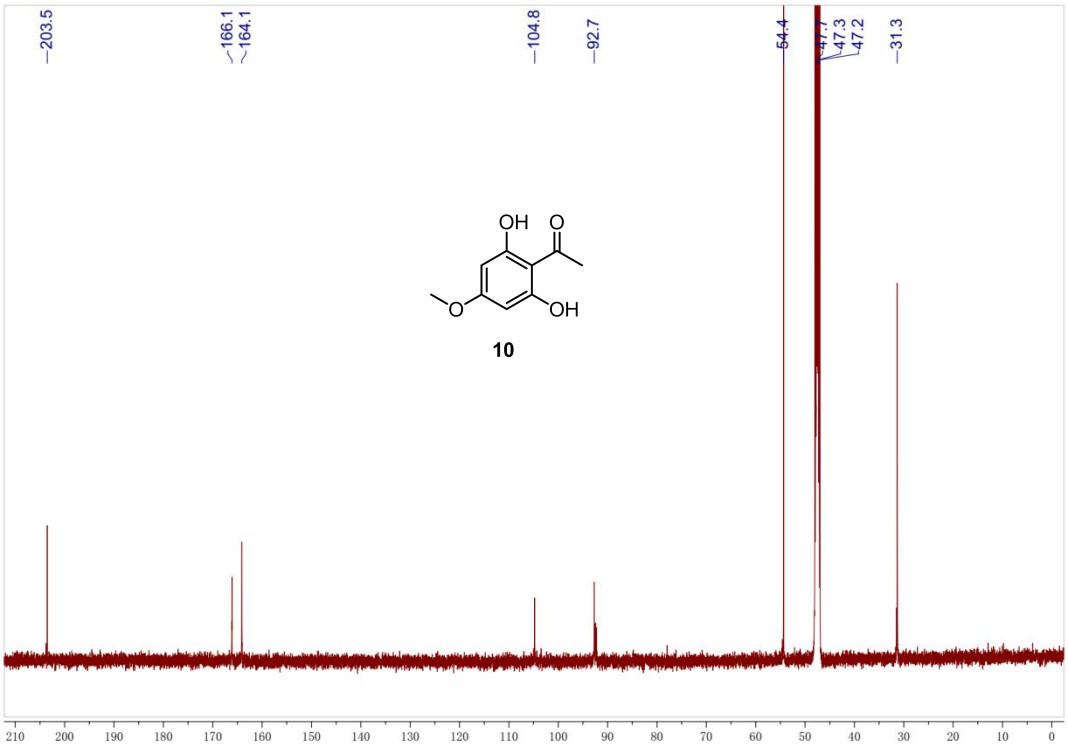


Figure S5. ^{1}H NMR spectrum (500 MHz, $CDCl_3$) of **4**

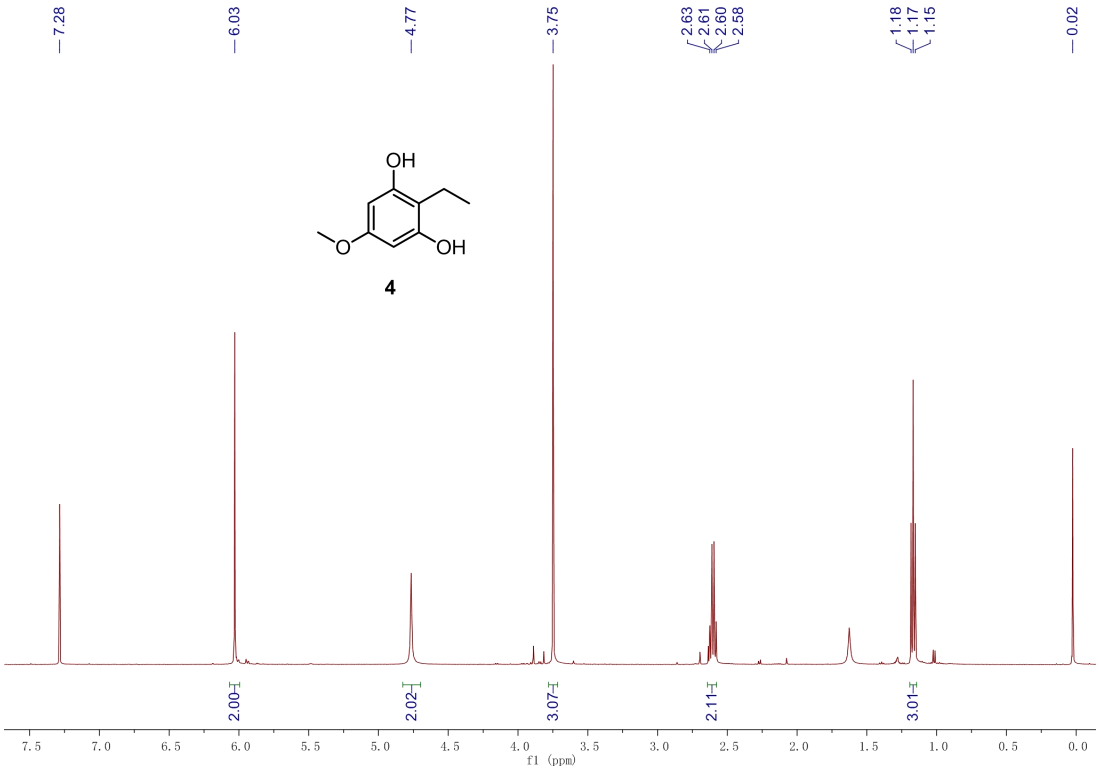


Figure S6. ^{13}C NMR spectrum (125 MHz, $CDCl_3$) of **4**

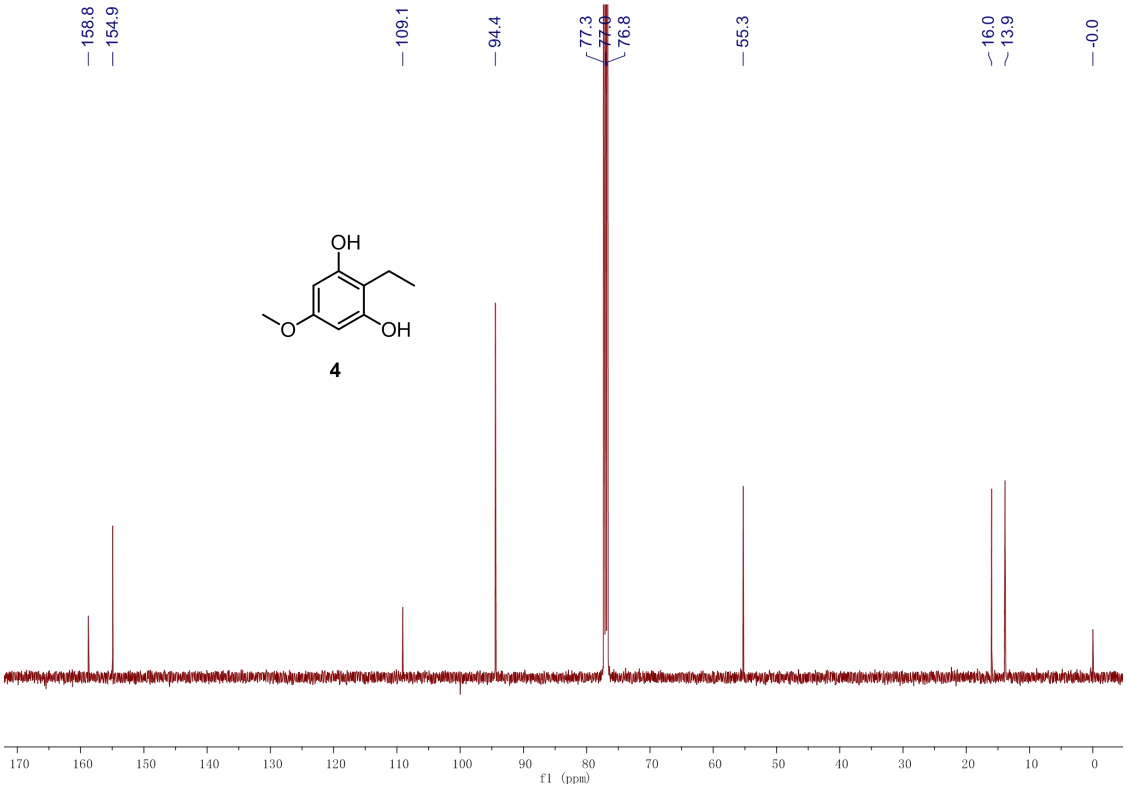


Figure S7. ^{1}H NMR spectrum (500 MHz, $CDCl_3$) of **3**

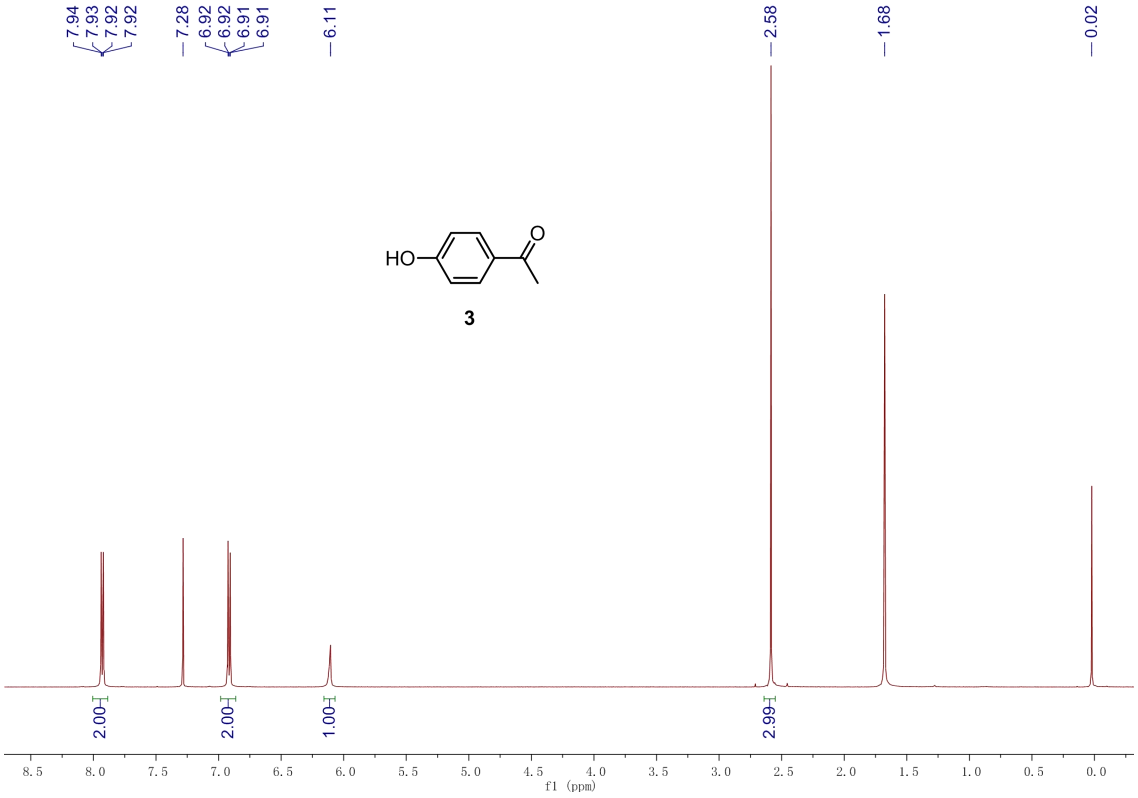


Figure S8. ^{13}C NMR spectrum (500 MHz, $CDCl_3$) of **3**

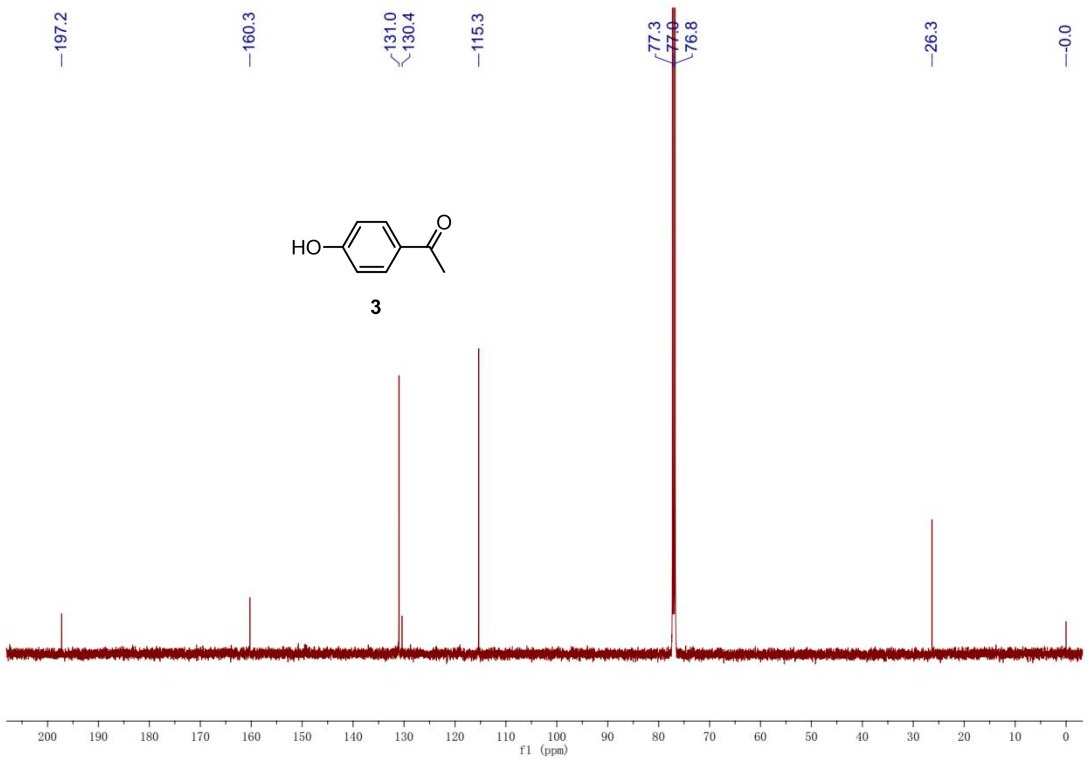


Figure S9. ^{1}H NMR spectrum (500 MHz, $CDCl_3$) of the synthetic**1**.

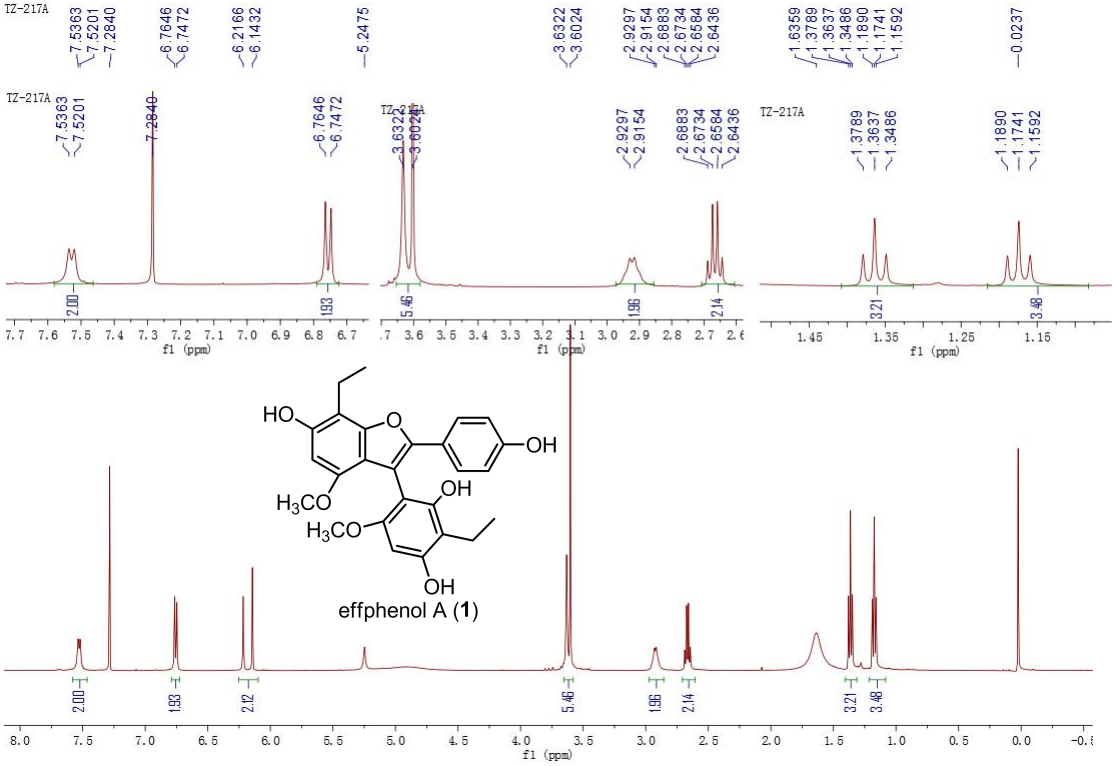


Figure S10. ^{13}C NMR spectrum (125 MHz, $CDCl_3$) of the synthetic **1**.

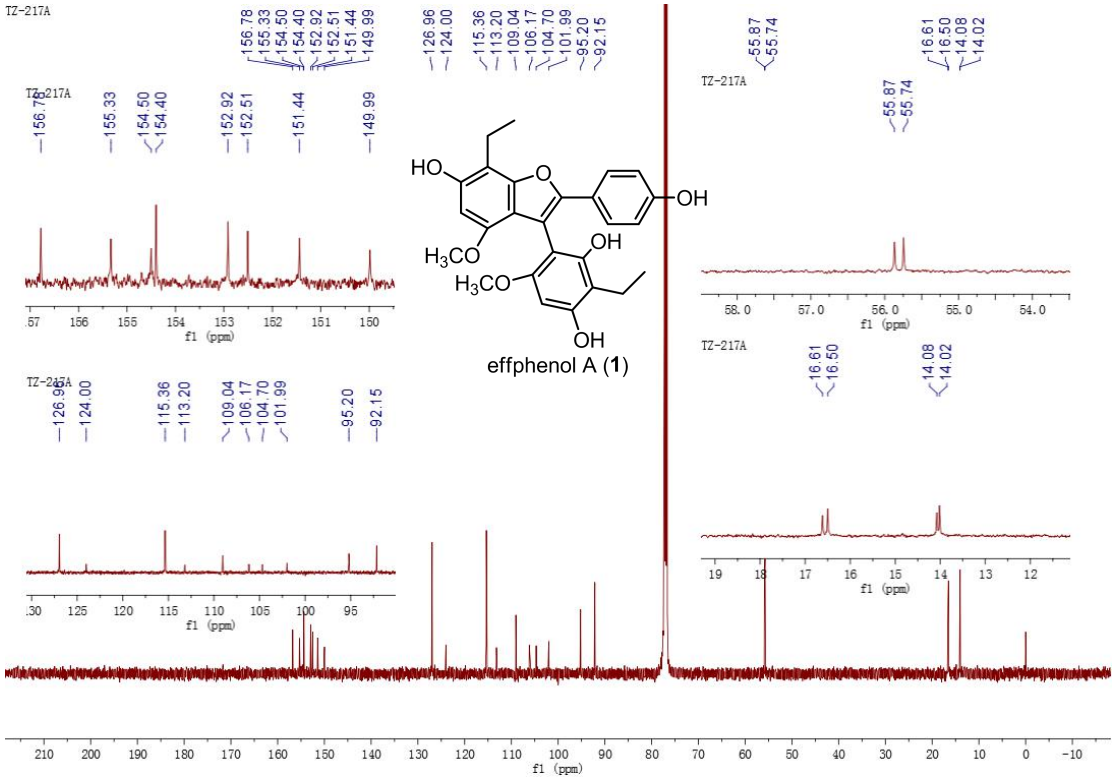


Figure S11. ^{1}H NMR spectrum (500 MHz, CD_3COCD_3) of the synthetic **1**.

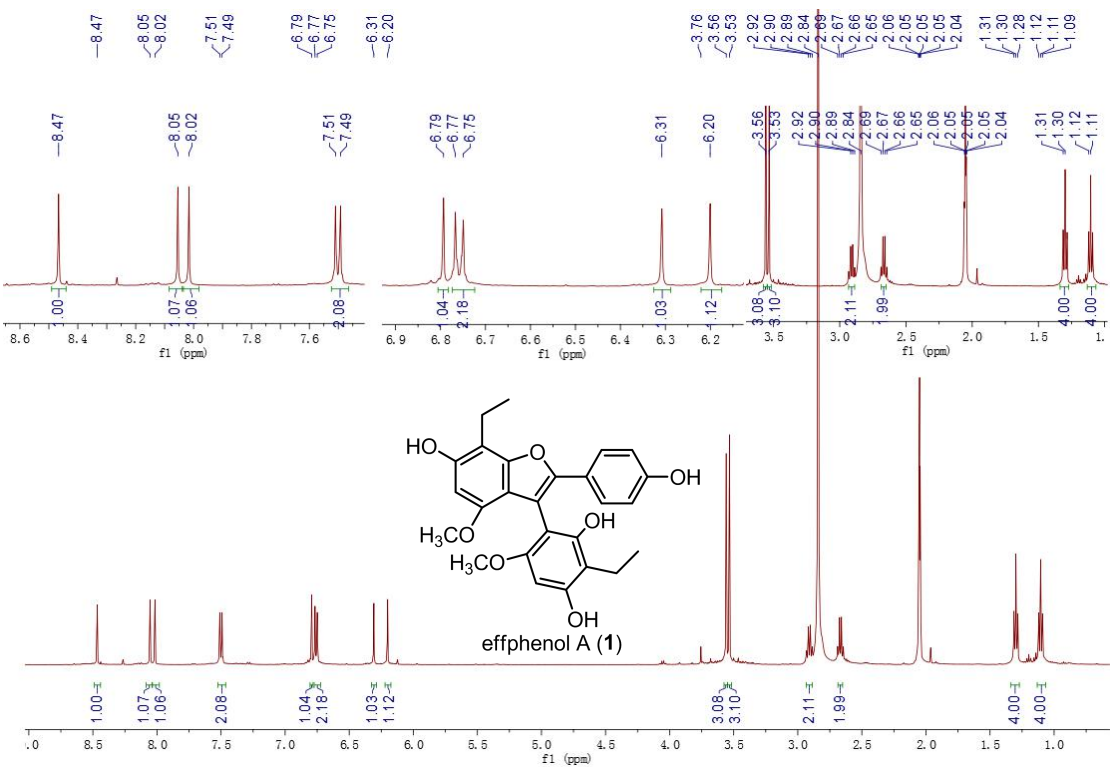


Figure S12. ^{13}C NMR spectrum (500 MHz, CD_3COCD_3) of the synthetic **1**.

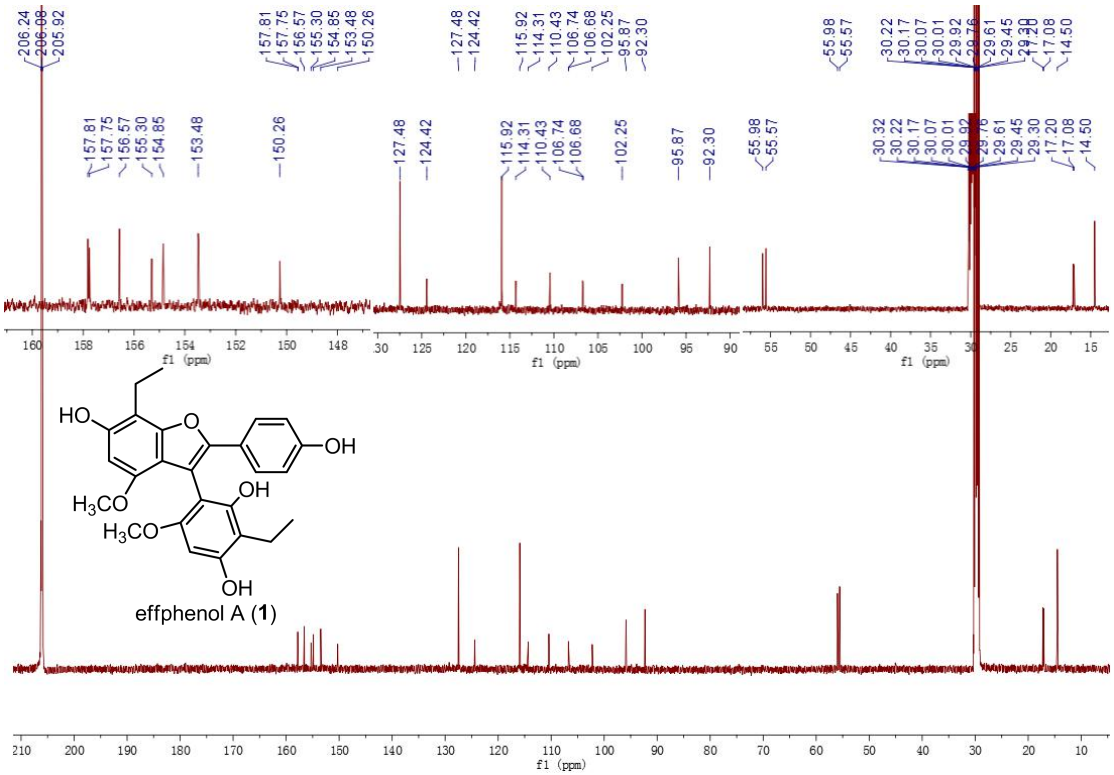


Figure S13. The comparison of ^{1}H NMR spectra of the natural product **1** and synthetic **1**.

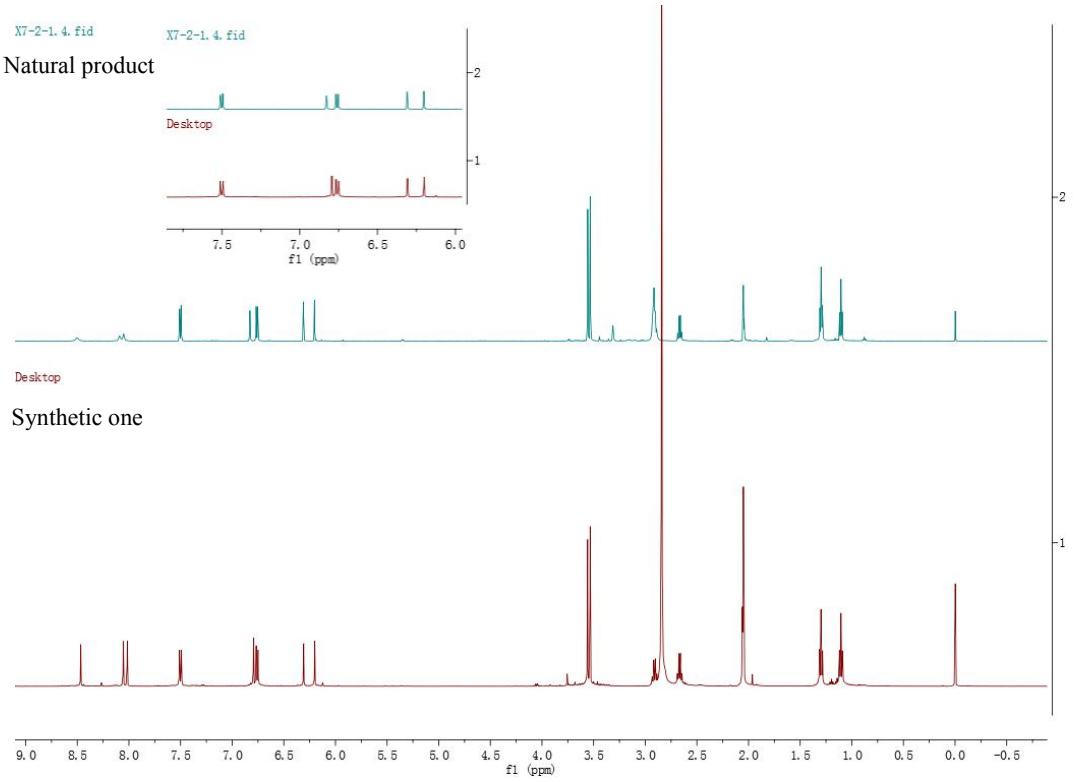


Figure S14. The comparison of ^{13}C NMR spectra of the natural product **1** and synthetic **1**.