

Supplementary Material

Bioactivity-guided Screening of Antimicrobial Secondary Metabolites from Antarctic Cultivable Fungus *Acrostalagmus luteoalbus* CH-6 Combined with Molecular Networking

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Figure S1. Fermented cultivable fungal colonies from Fildes Peninsula, Antarctica

Figure S2. The colony morphology (left) and light microscopy (right) of *A. luteoalbus* CH-6

Figure S3. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **1**

Figure S4. ^{13}C NMR (150 MHz, CDCl_3) spectrum of compound **1**

Figure S5. HSQC (CDCl_3) spectrum of compound **1**

Figure S6. COSY (CDCl_3) spectrum of compound **1**

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Figure S17. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **1s**

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Figure S27. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **4**

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Figure S31. ^1H NMR (600 MHz, CD_3OD) spectrum of compound **6**

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Figure S33. HPLC fingerprints of separated fractions Fr.1–Fr.8 of *A. luteoalbus* CH-6

Table S1. Identified compounds by molecular networking

Table S2. OR values of compounds **4–6**

Table S3. Antimicrobial activities of the separated fractions (50 µg/mL) of *A. luteoalbus* CH-6

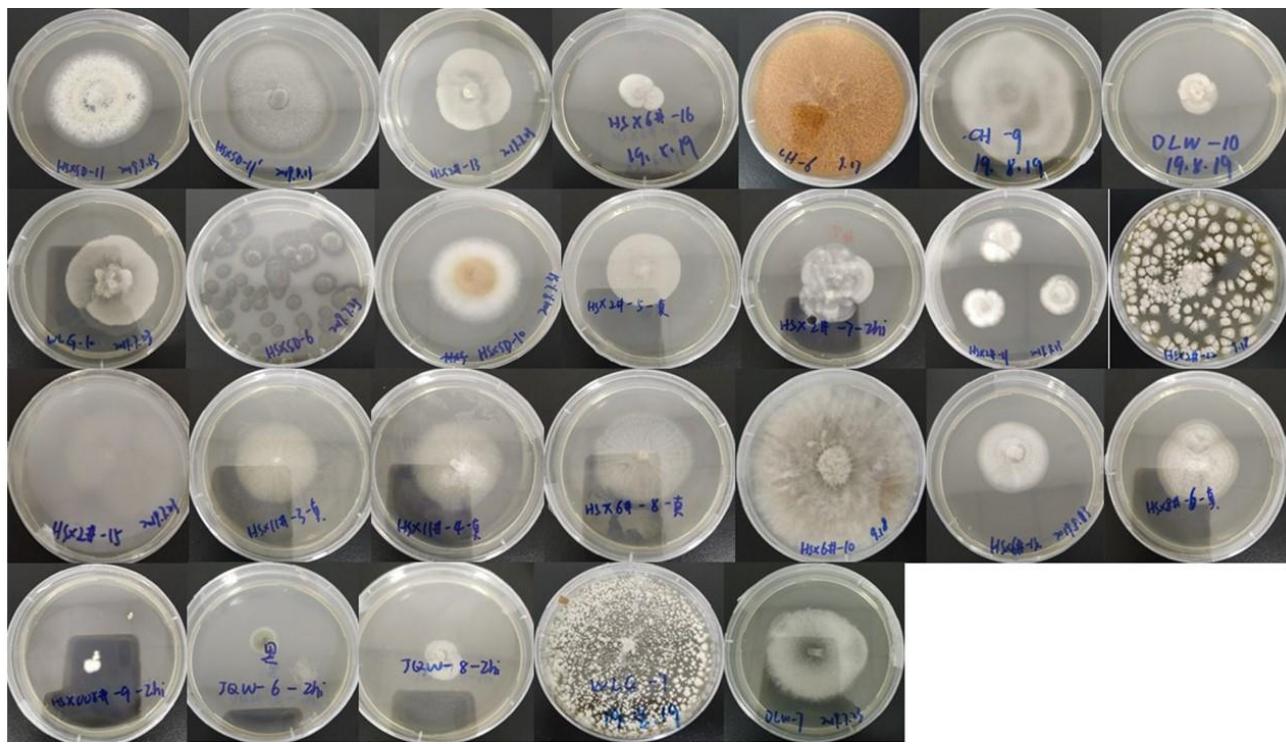
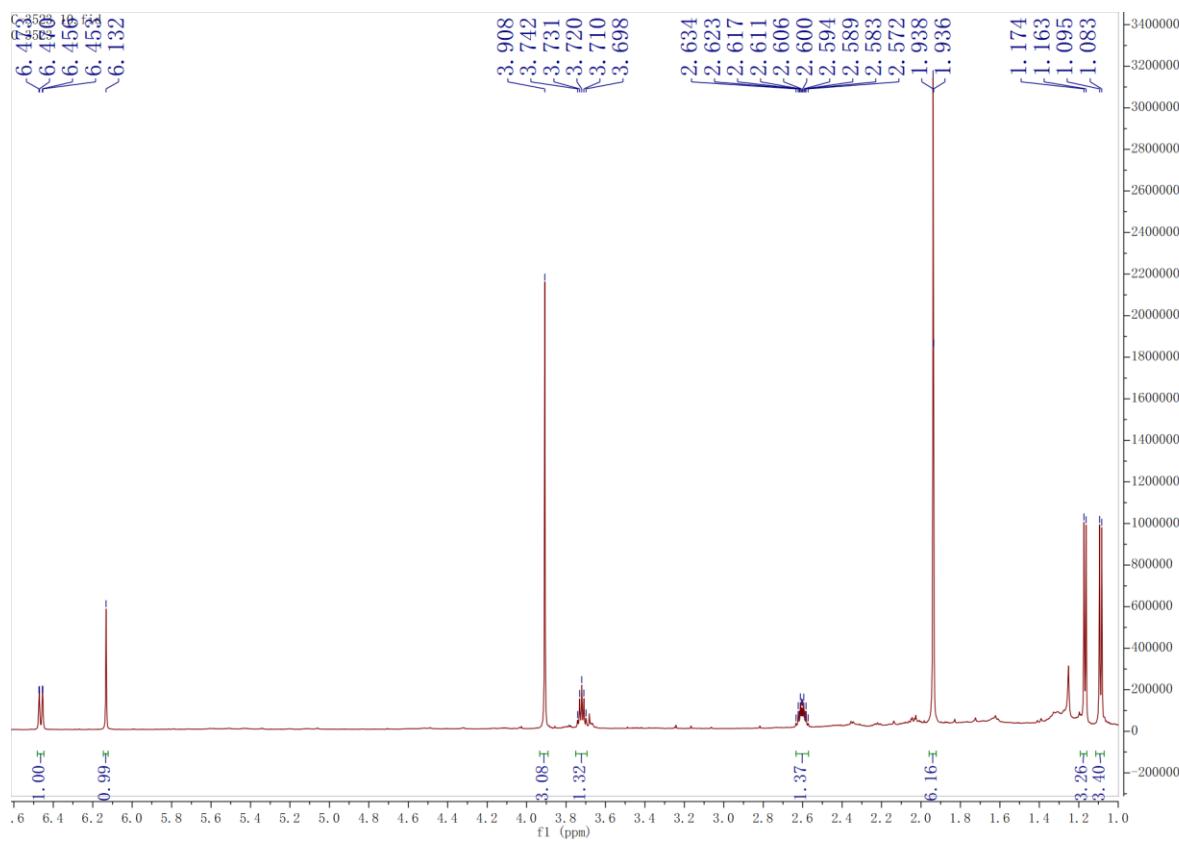
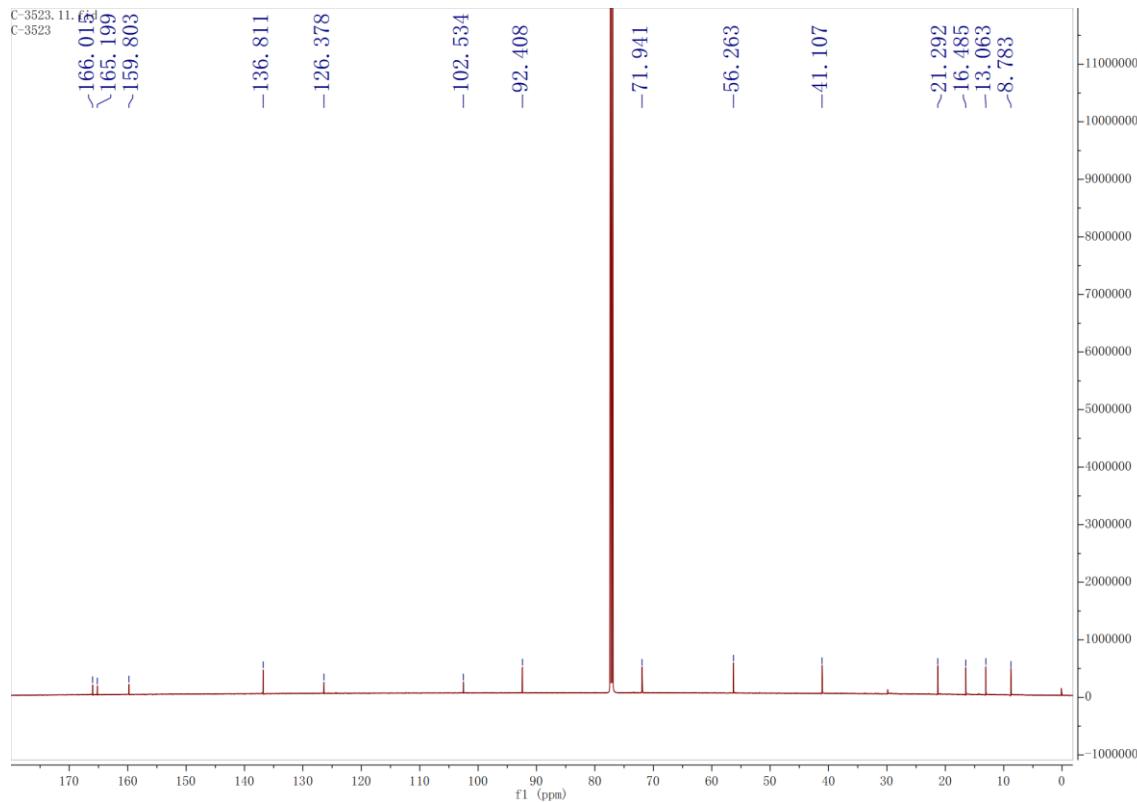


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Figure S2. The colony morphology (left) and light microscopy (right) of *A. luteoalbus* CH-6

**Figure S3.** ^1H NMR (600 MHz, CDCl_3) spectrum of compound **1****Figure S4.** ^{13}C NMR (150 MHz, CDCl_3) spectrum of compound **1**

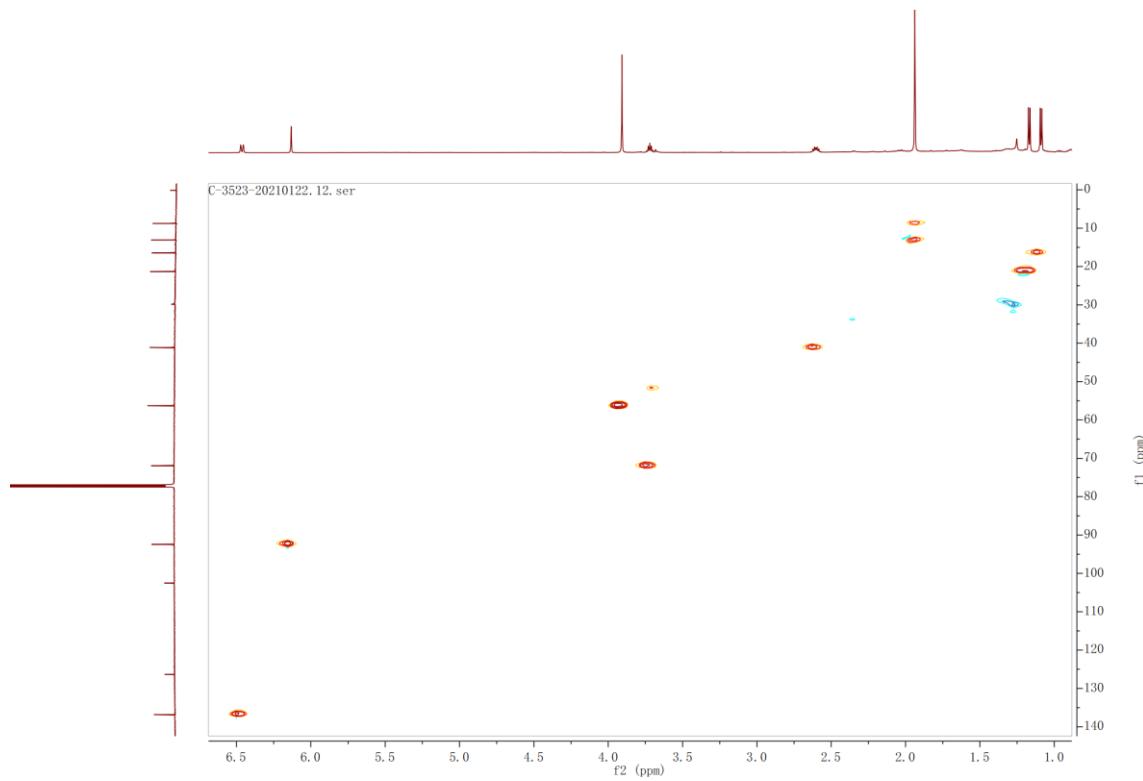


Figure S5. HSQC (CDCl_3) spectrum of compound **1**

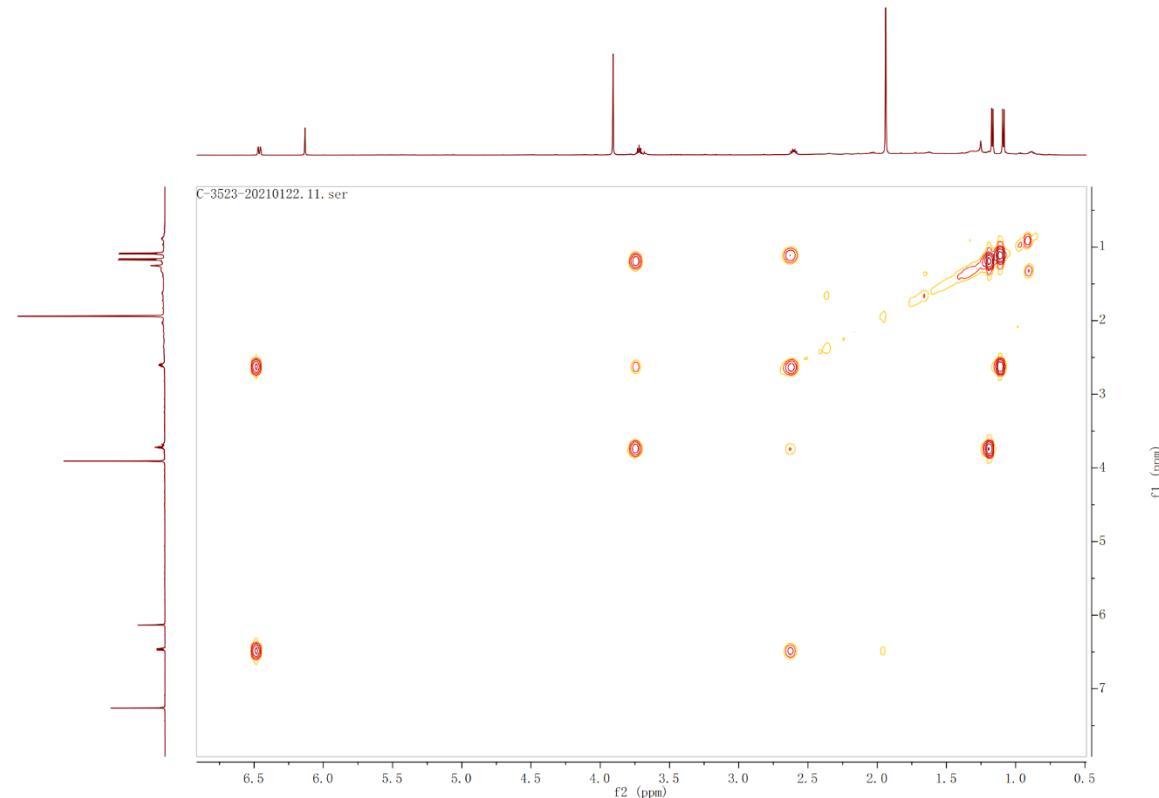


Figure S6. COSY (CDCl_3) spectrum of compound **1**

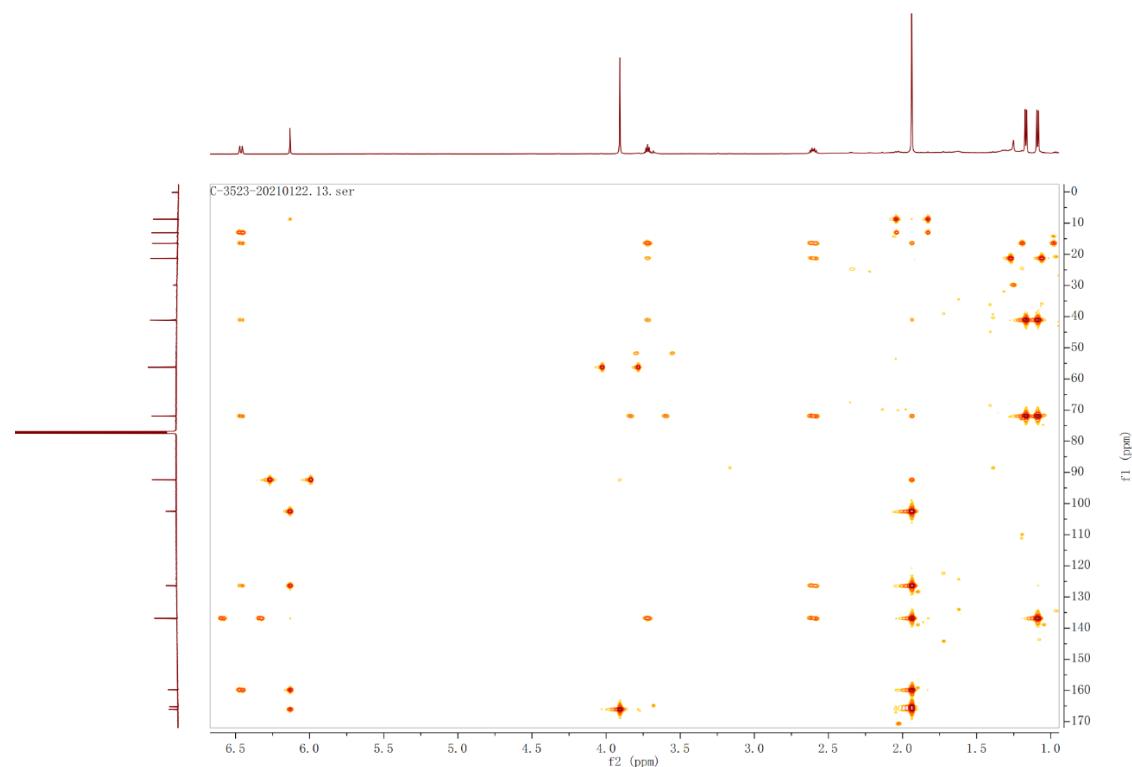


Figure S7. HMBC (CDCl_3) spectrum of compound **1**

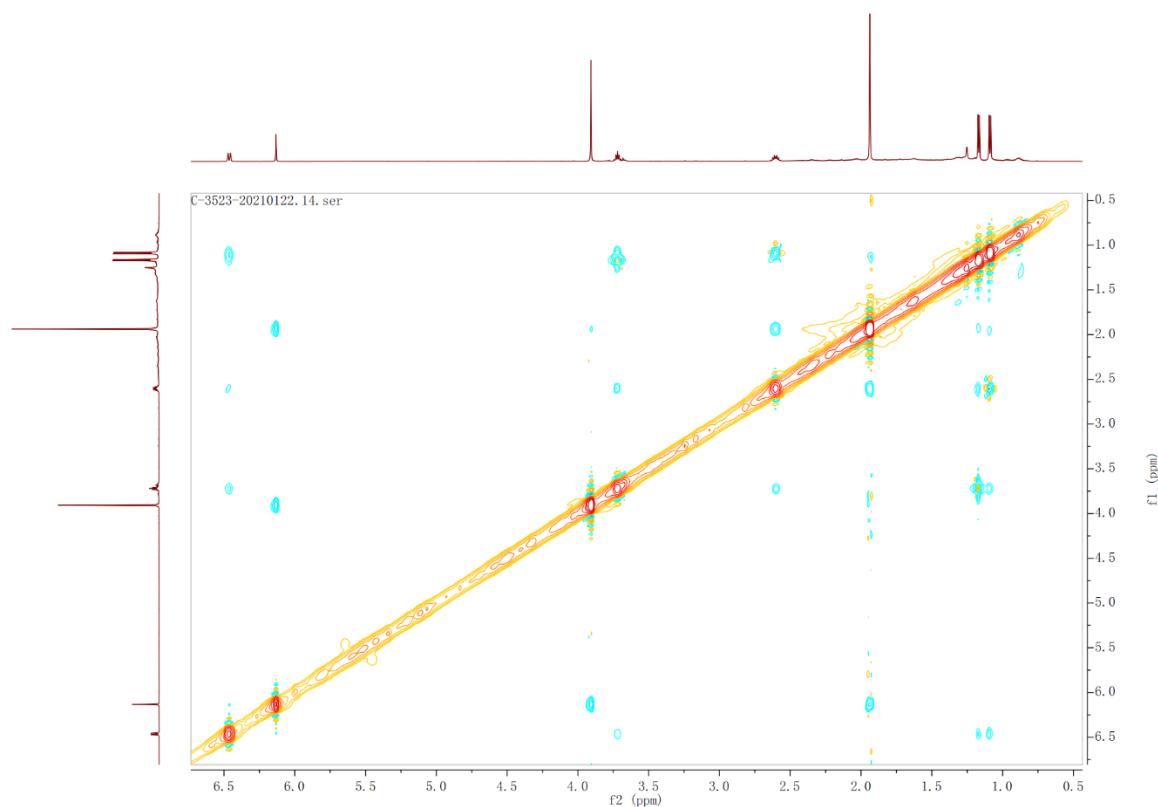


Figure S8. NOESY (CDCl_3) spectrum of compound **1**

C3523 #15 RT: 0.20 AV: 1 NL: 1.27E5
T: FTMS {1,1} + p ESI Full ms [100.00-1000.00]

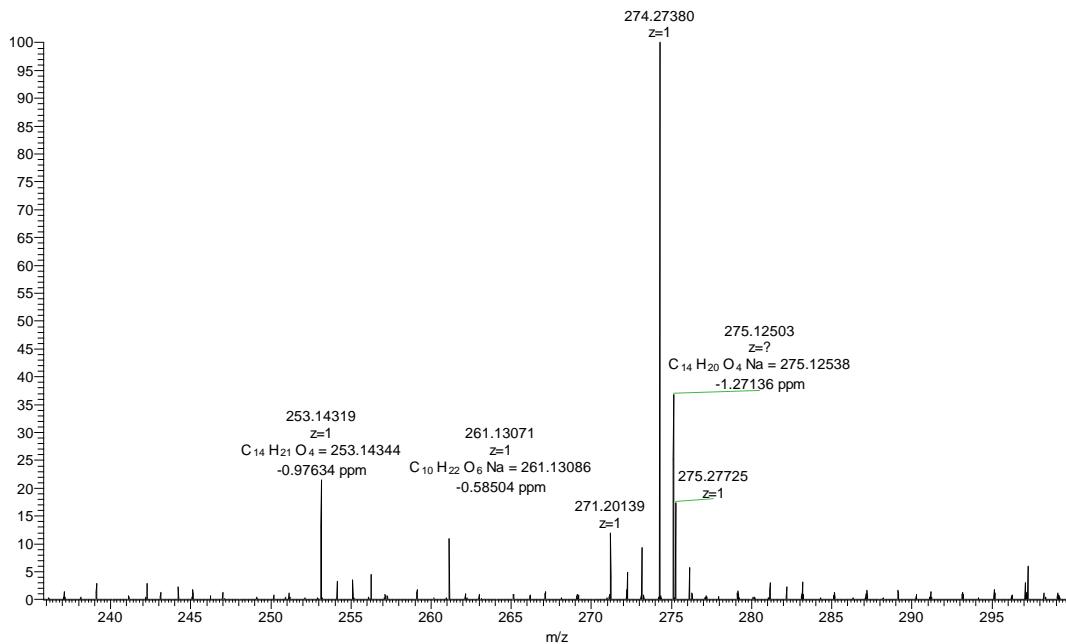


Figure S9. HR-ESI-MS spectrum of compound 1

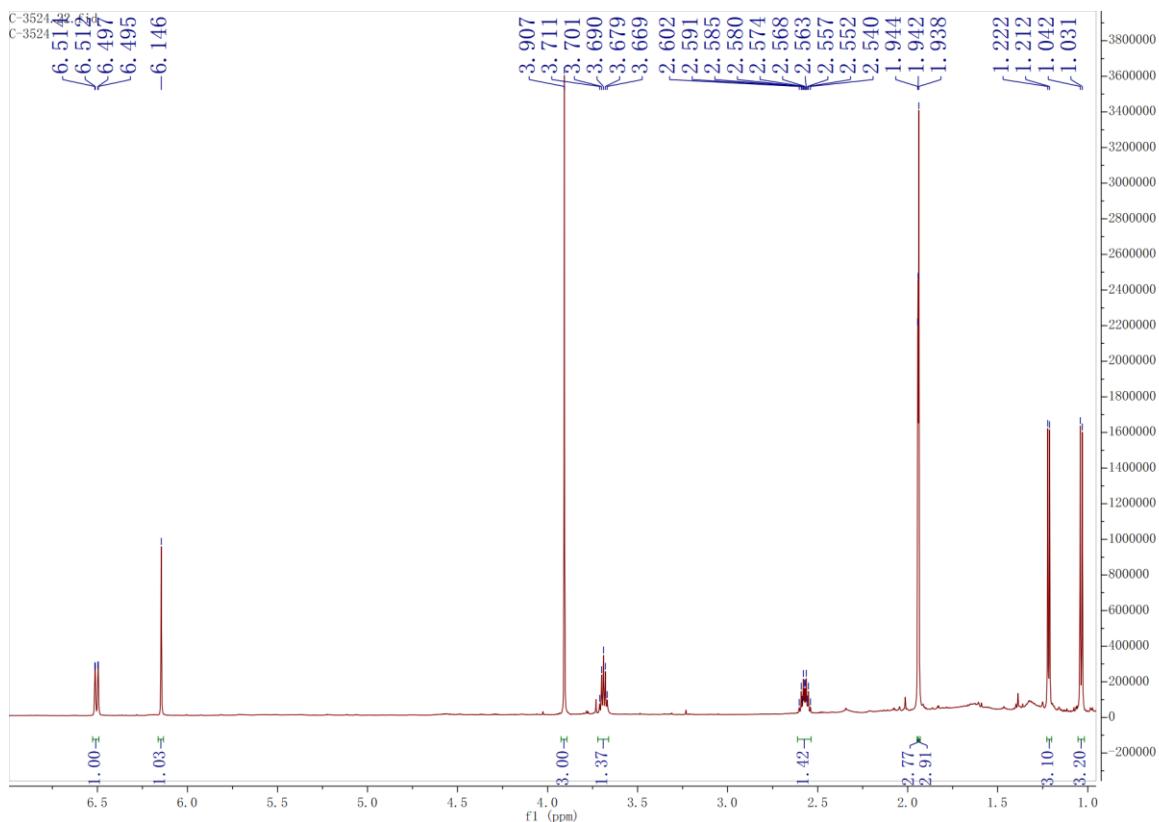
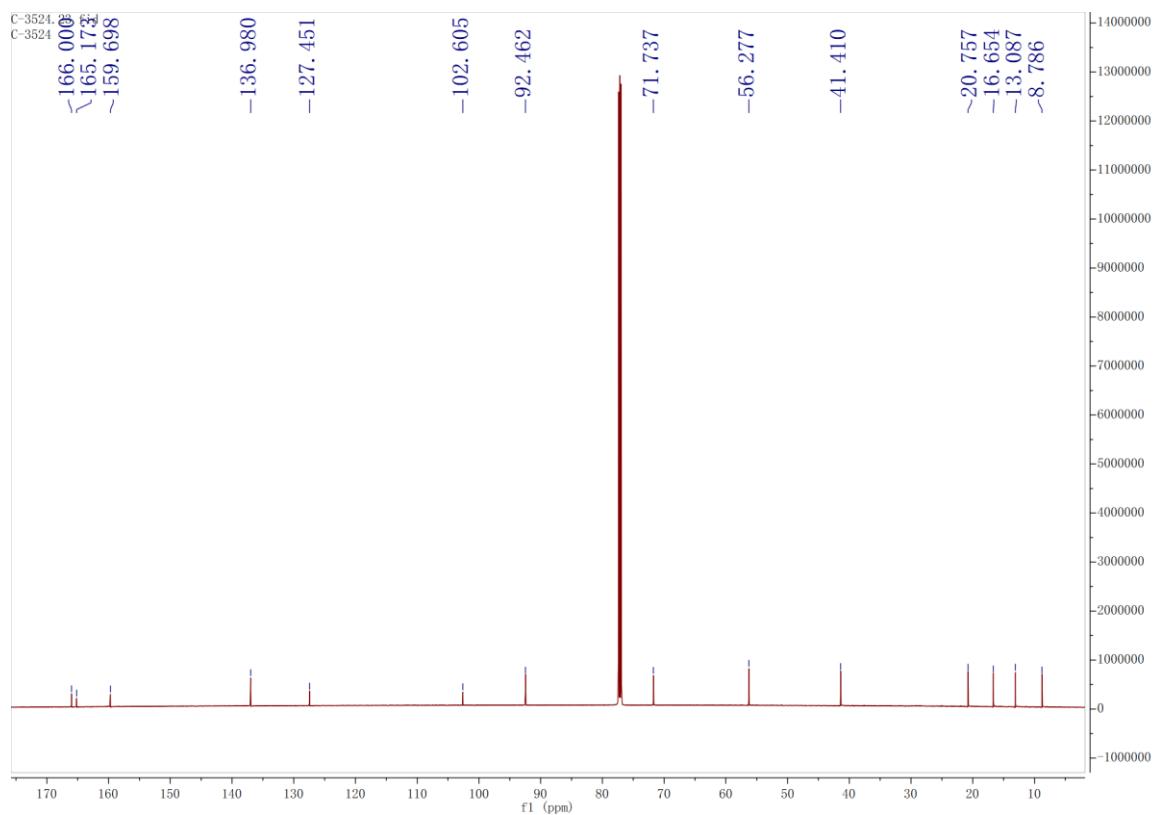
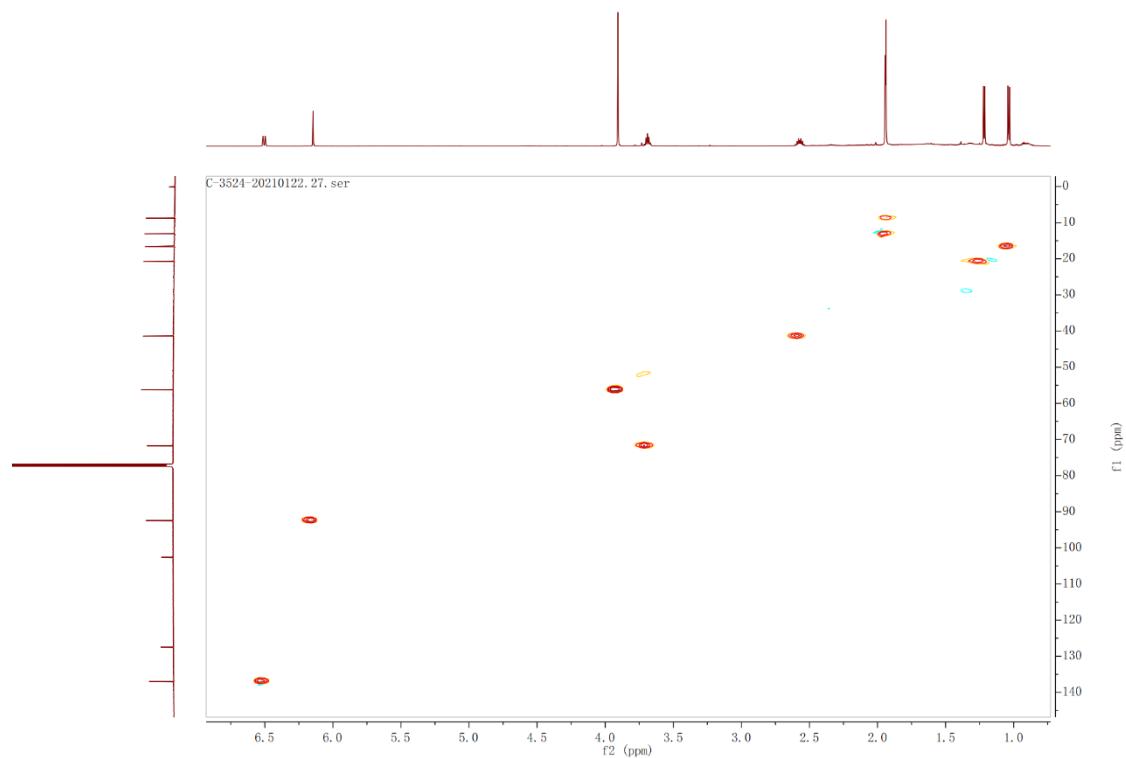


Figure S10. ^1H NMR (600 MHz, CDCl_3) spectrum of compound 2

**Figure S11.** ^{13}C NMR (150 MHz, CDCl_3) spectrum of compound 2**Figure S12.** HSQC (CDCl_3) spectrum of compound 2

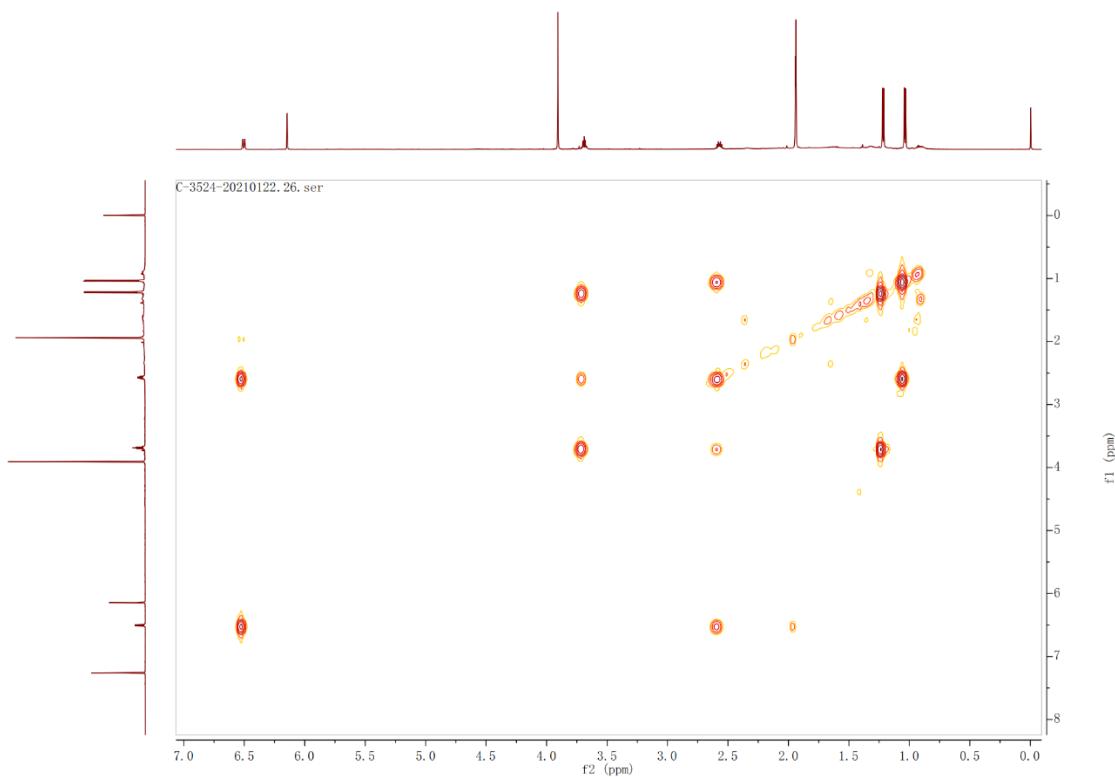


Figure S13. COSY (CDCl_3) spectrum of compound 2

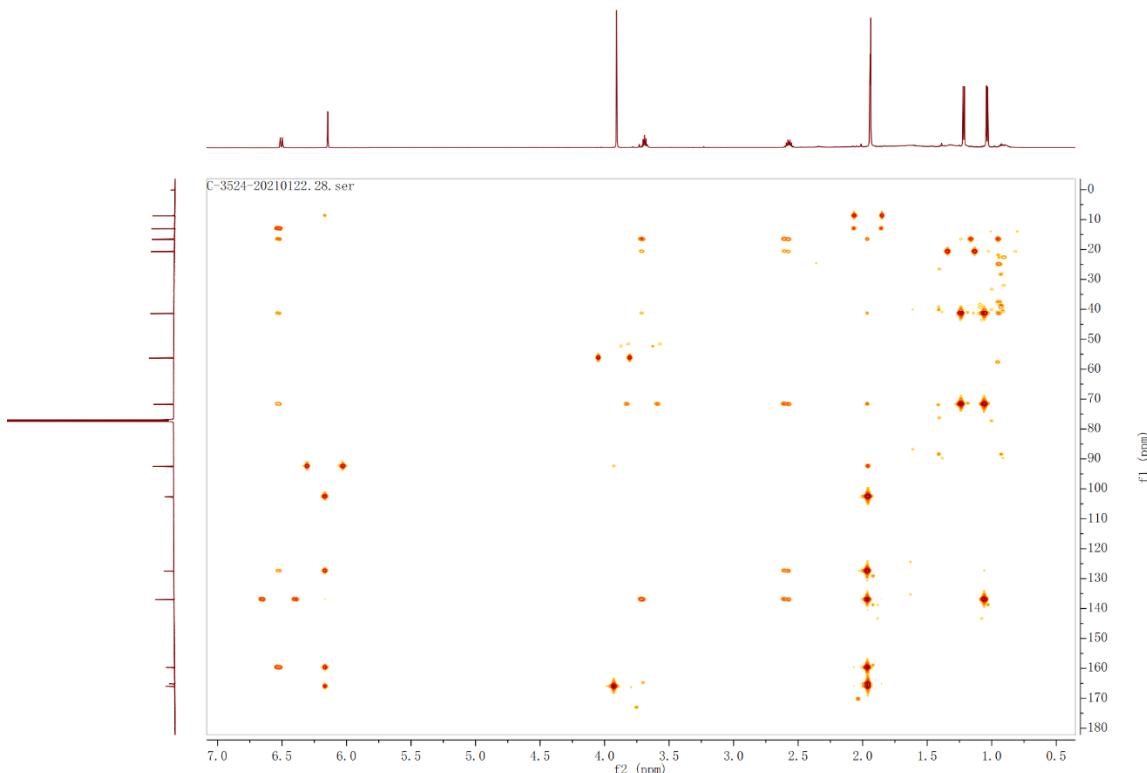
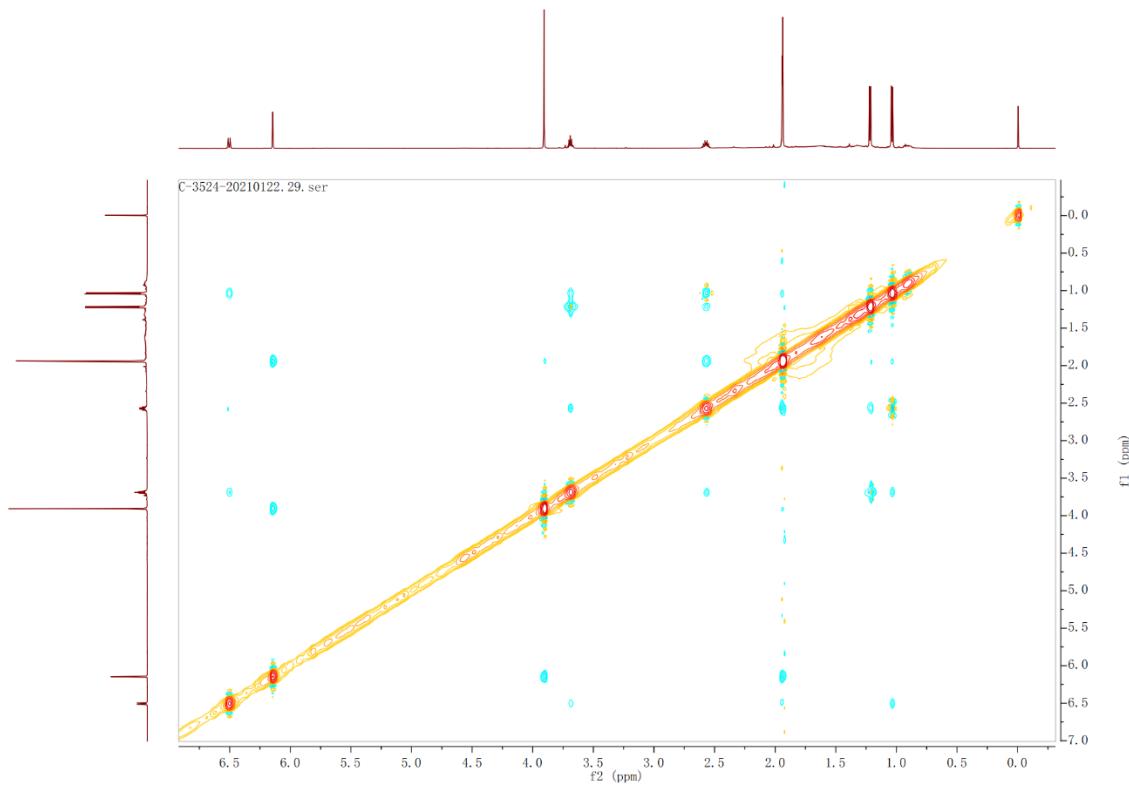
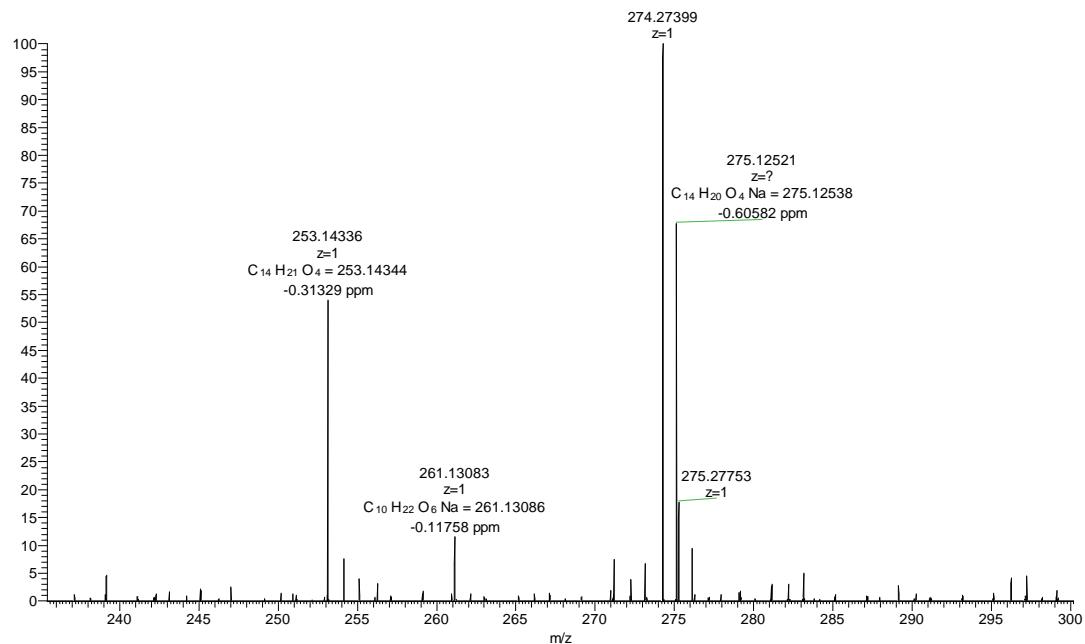


Figure S14. HMBC (CDCl_3) spectrum of compound 2

**Figure S15.** NOESY (CDCl_3) spectrum of compound **2**

C3524 #15 RT: 0.20 AV: 1 NL: 4.59E4
T: FTMS {1,1} + p ESI Full ms [100.00-1000.00]

**Figure S16.** HR-ESI-MS spectrum of compound **2**

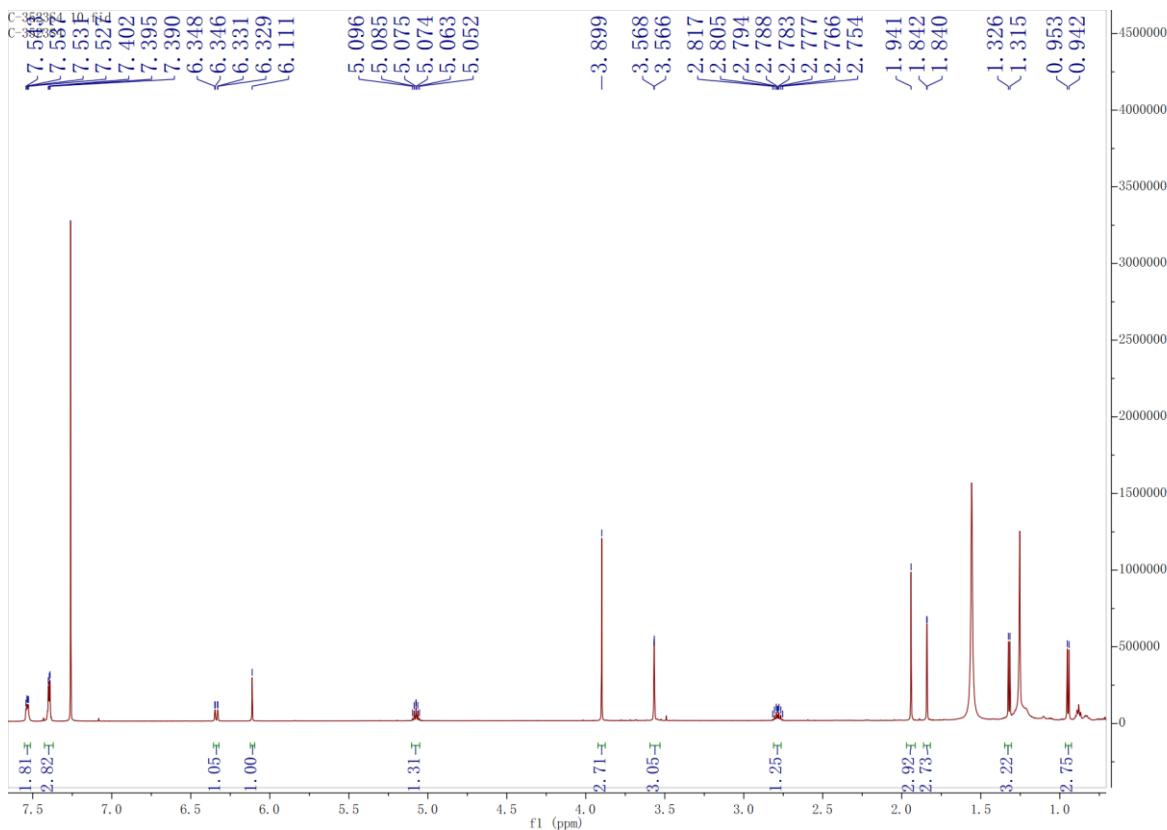


Figure S17. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **1s**

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T: FTMS {1,1} + p APCI corona Full ms [200.00-2000.00]

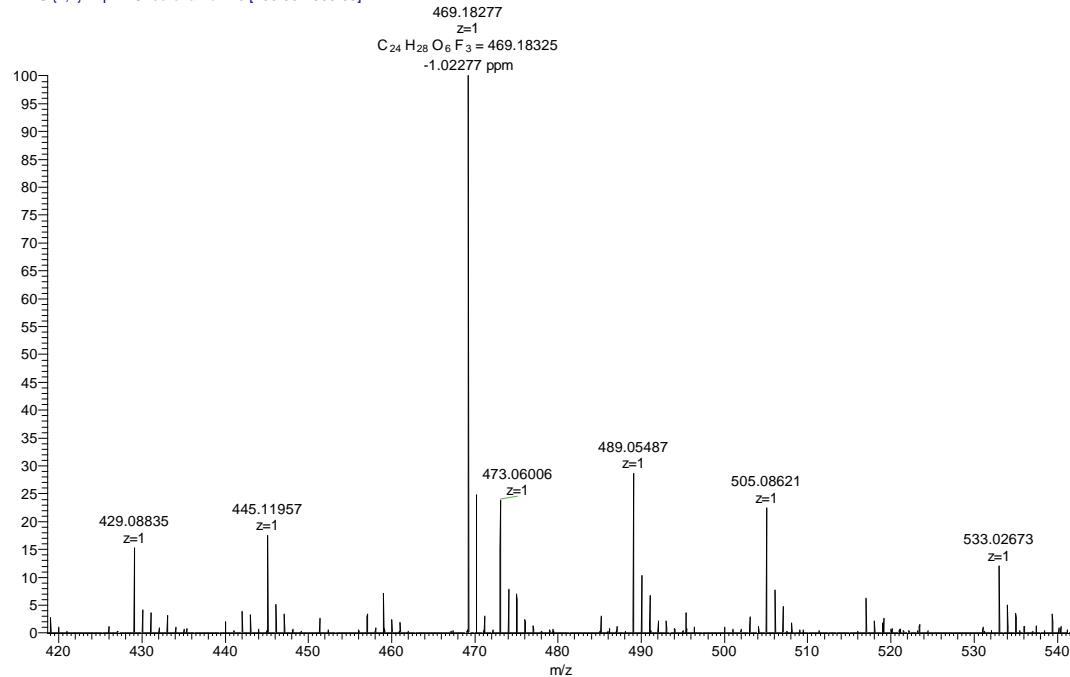
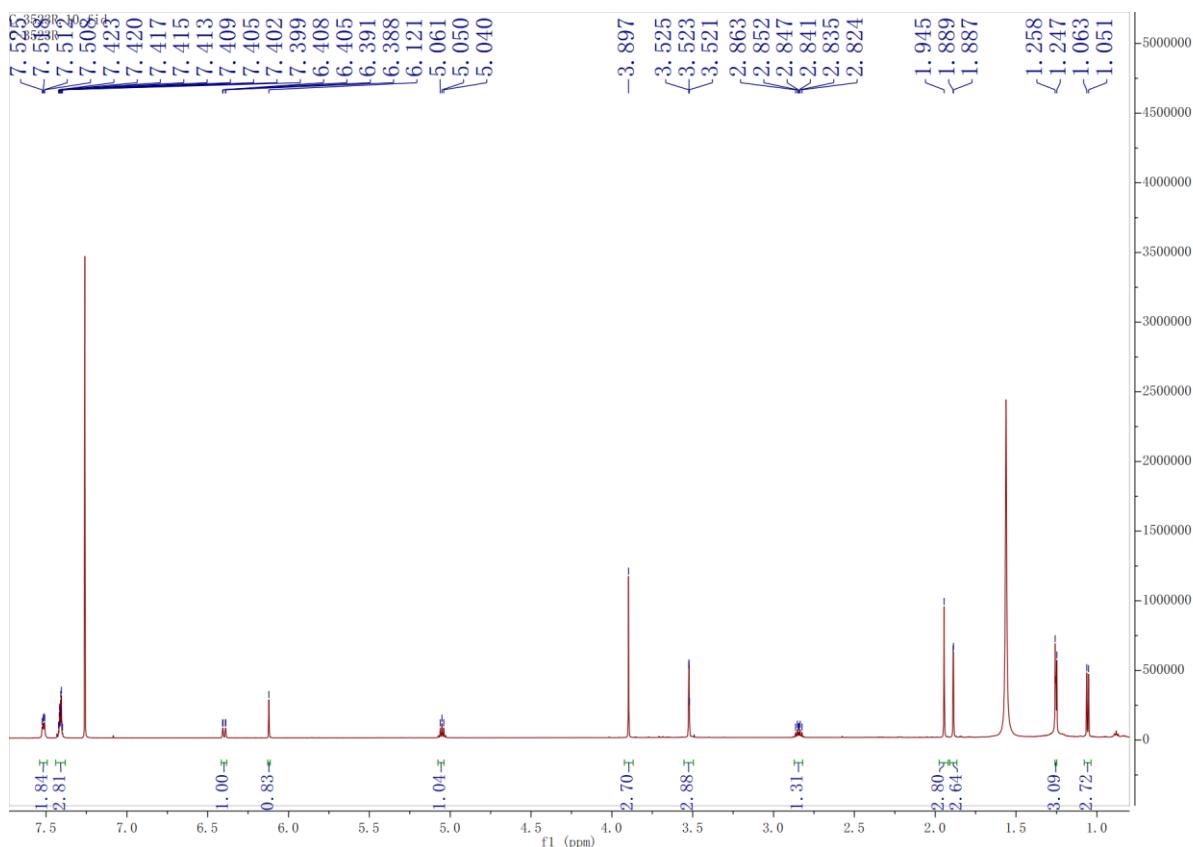
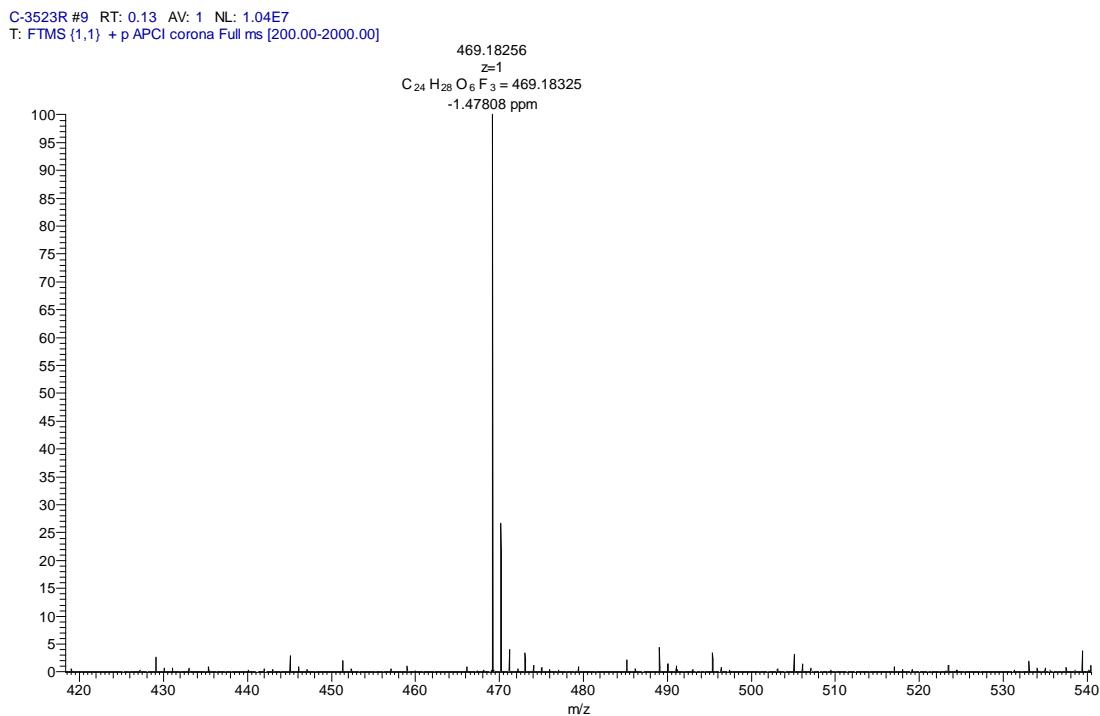


Figure S18. HR-APCI-MS spectrum of compound **1s**

**Figure S19.** ¹H NMR (600 MHz, CDCl₃) spectrum of compound 1r

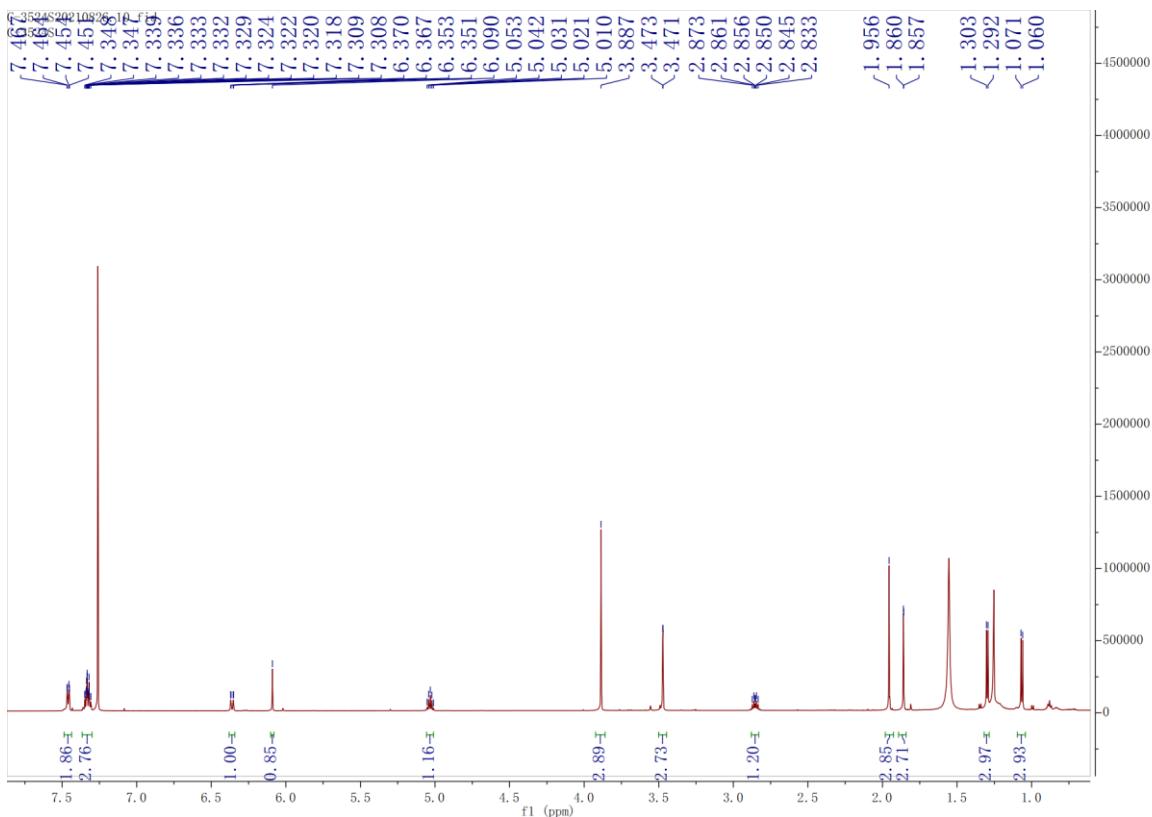


Figure S21. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **2s**

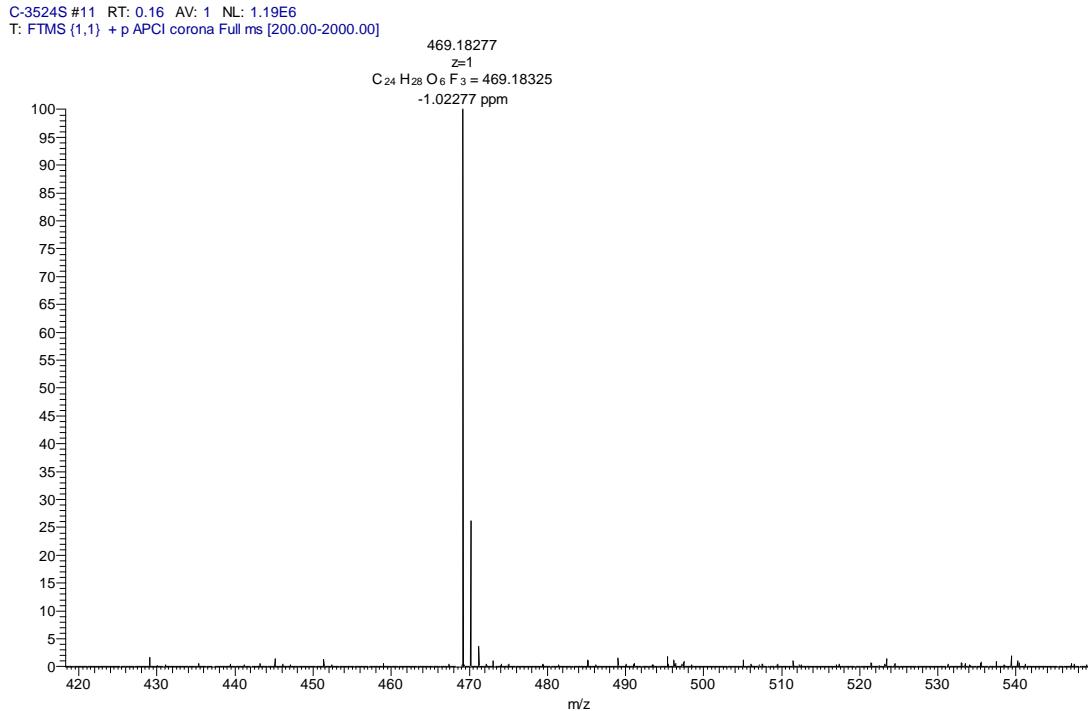


Figure S22. HR-APCI-MS spectrum of compound **2s**

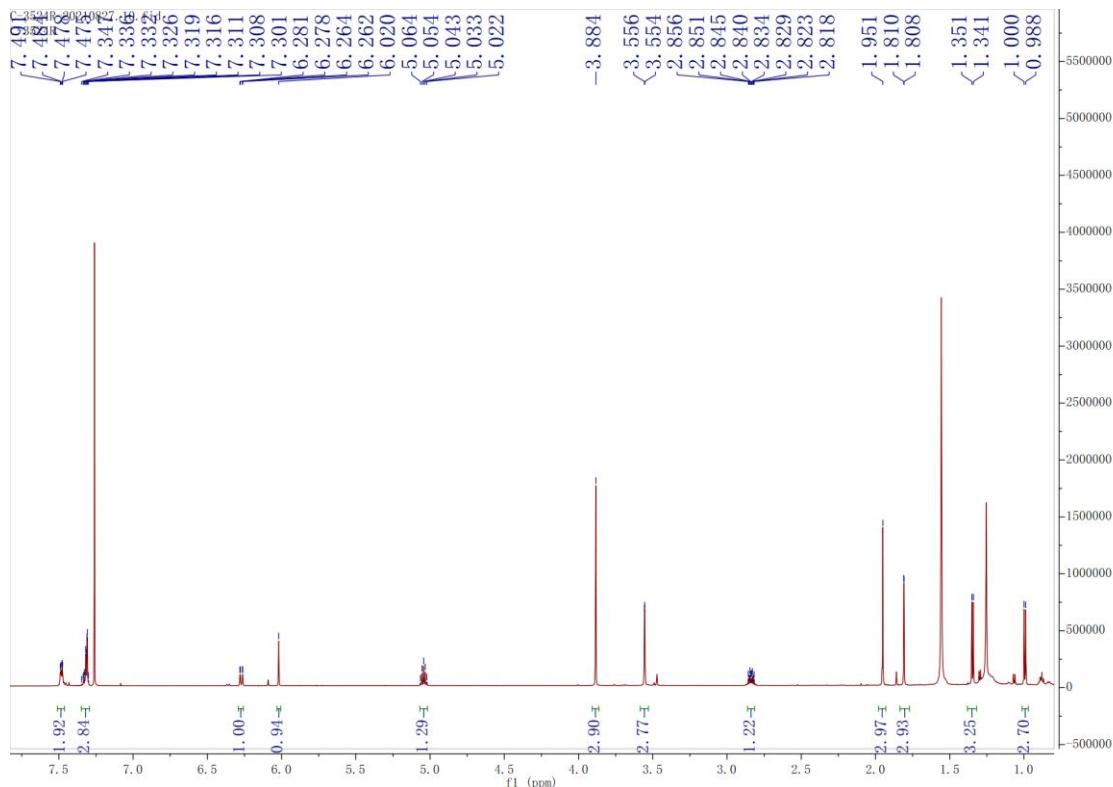


Figure S23. ^1H NMR (600 MHz, CDCl_3) spectrum of compound **2r**

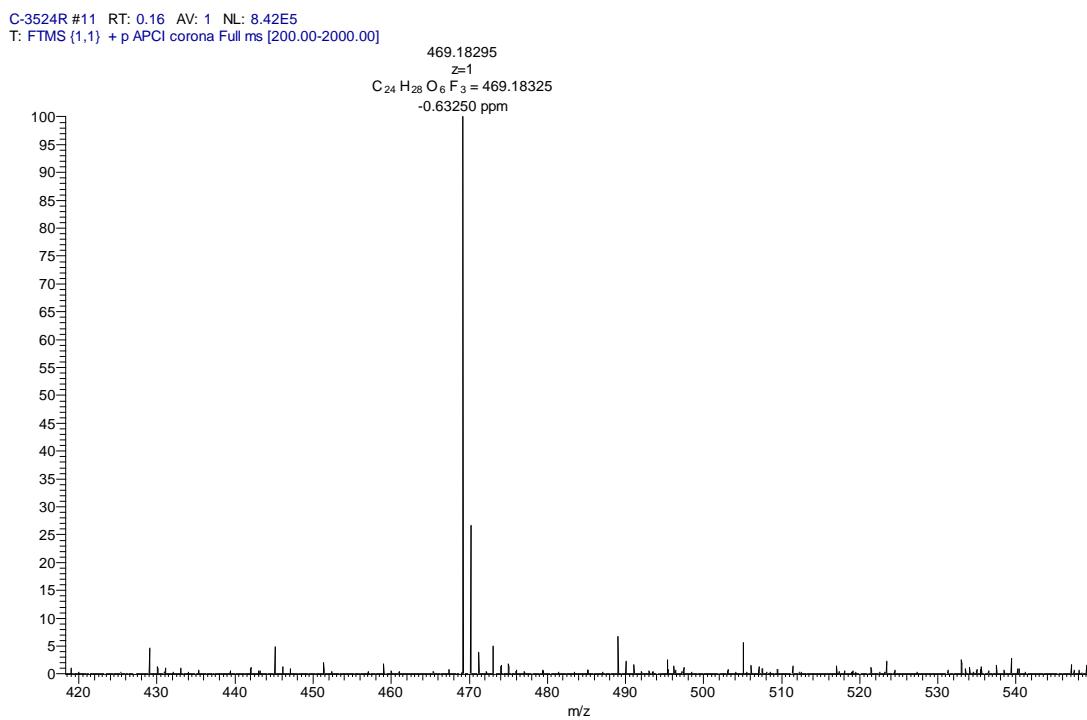


Figure S24. HR-APCI-MS spectrum of compound **2r**

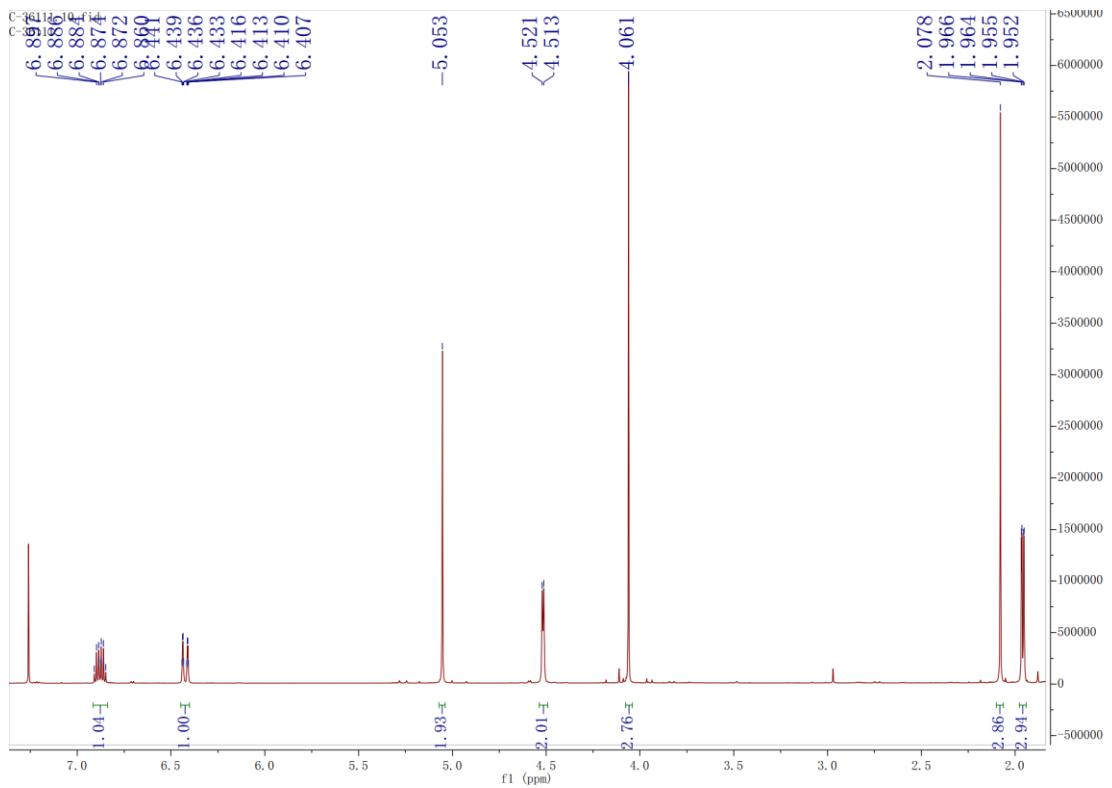


Figure S25. ^1H NMR (600 MHz, CDCl_3) spectrum of compound 3

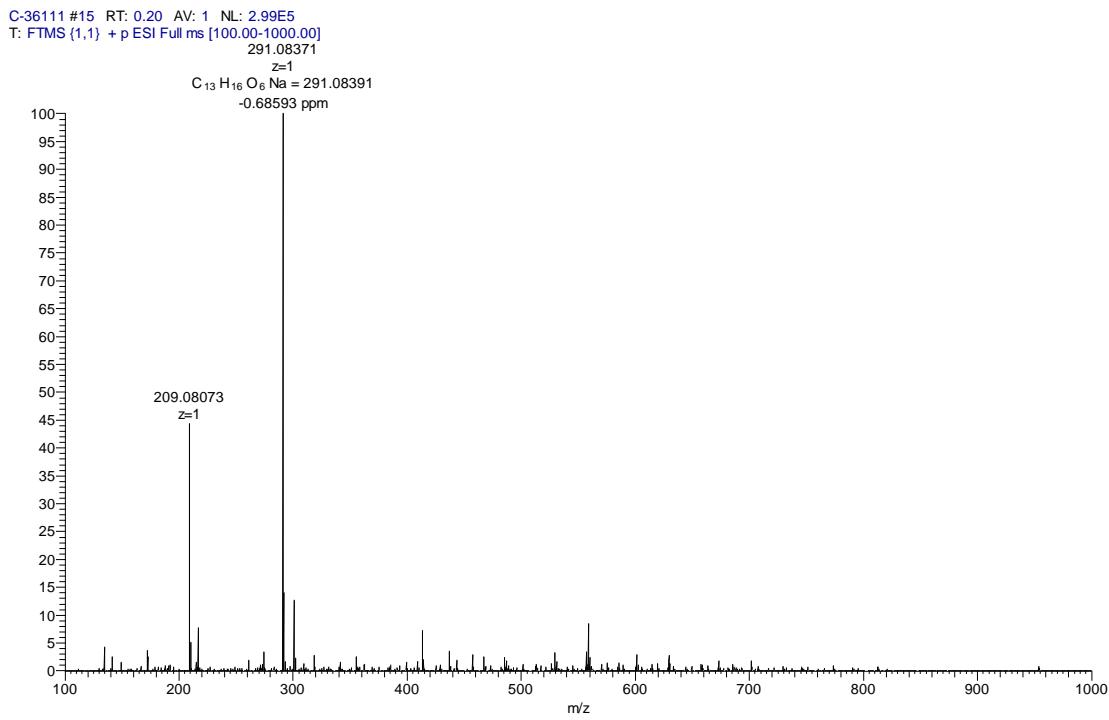


Figure S26. HR-ESI-MS spectrum of compound 3

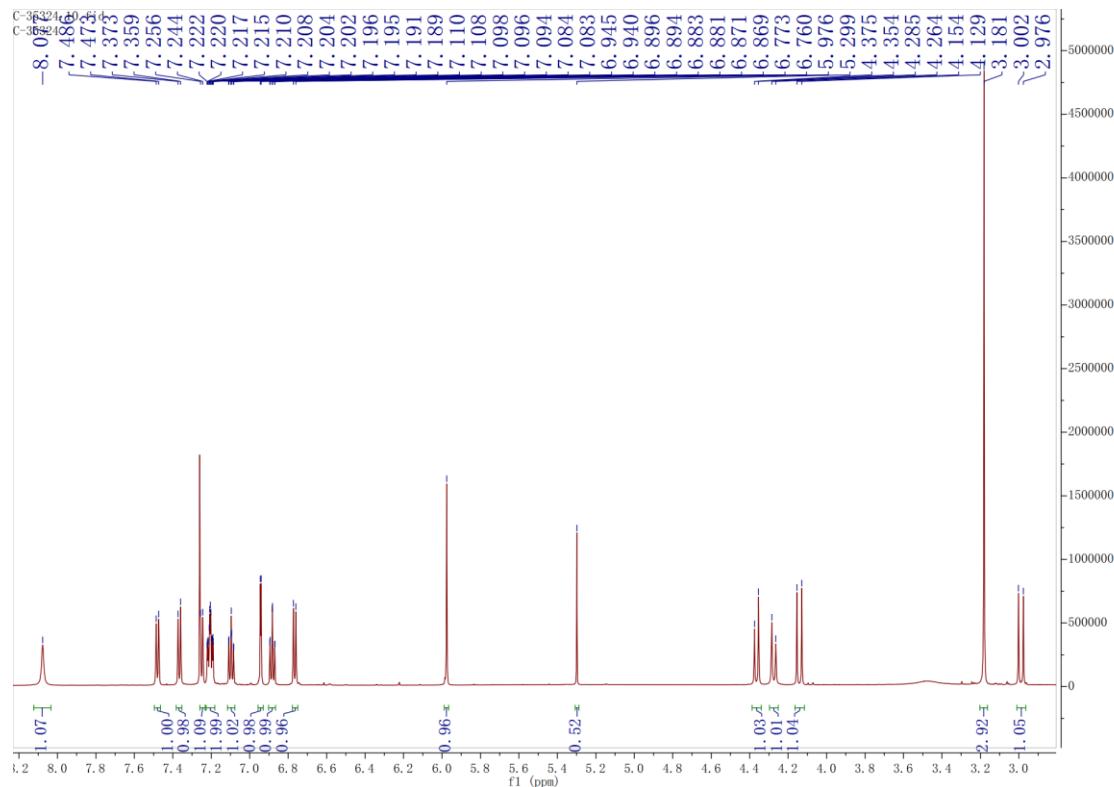


Figure S27. ^1H NMR (600 MHz, CDCl_3) spectrum of compound 4

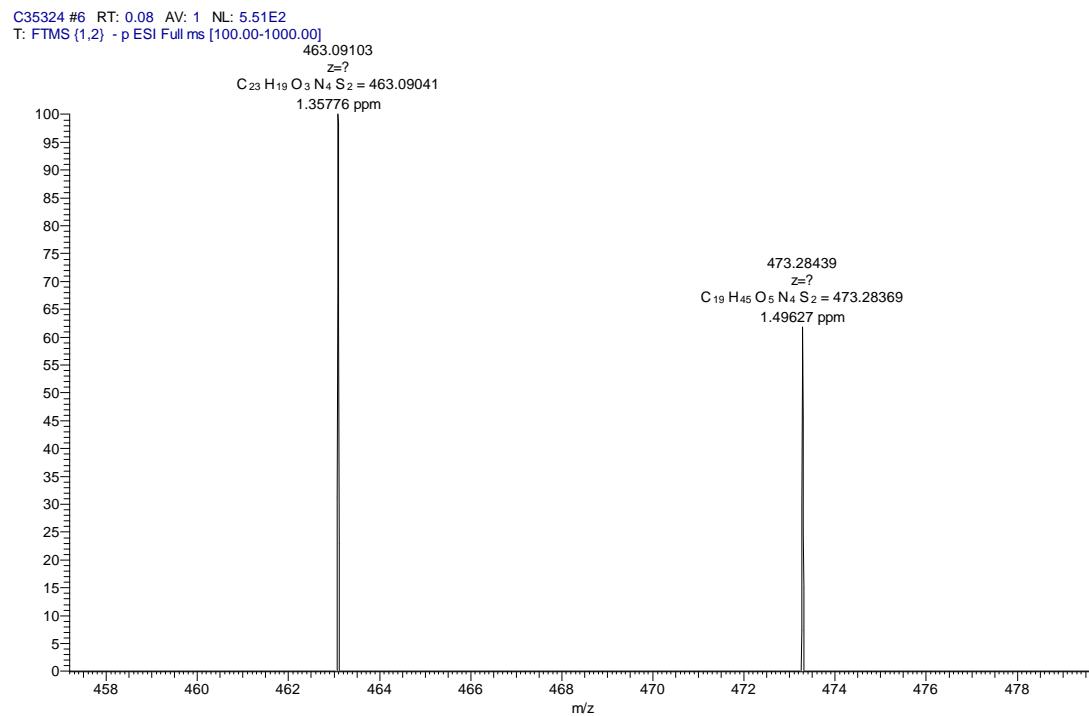


Figure S28. HR-ESI-MS spectrum of compound **4**

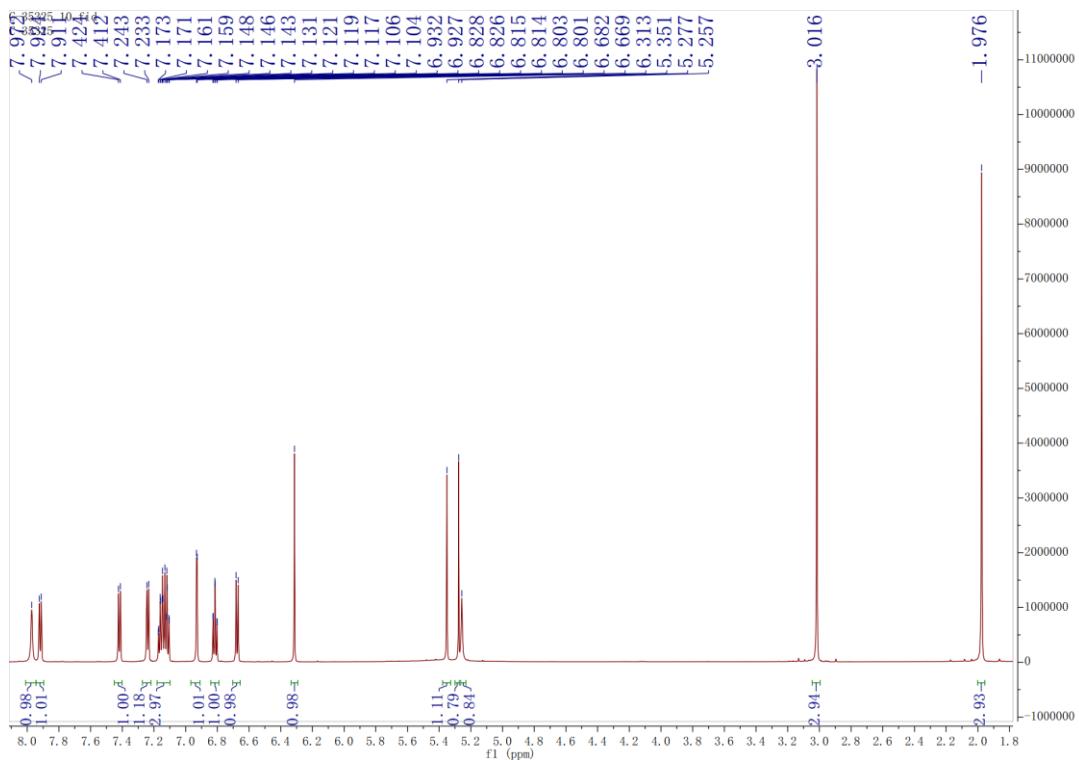


Figure S29. ^1H NMR (600 MHz, CDCl_3) spectrum of compound 5

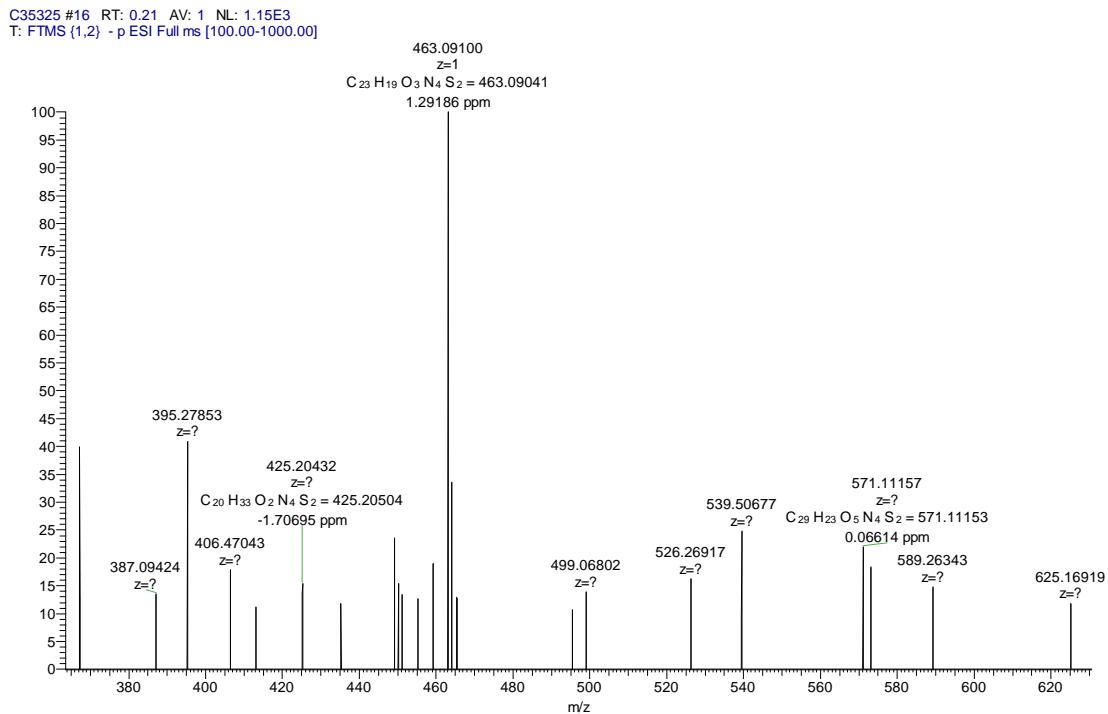
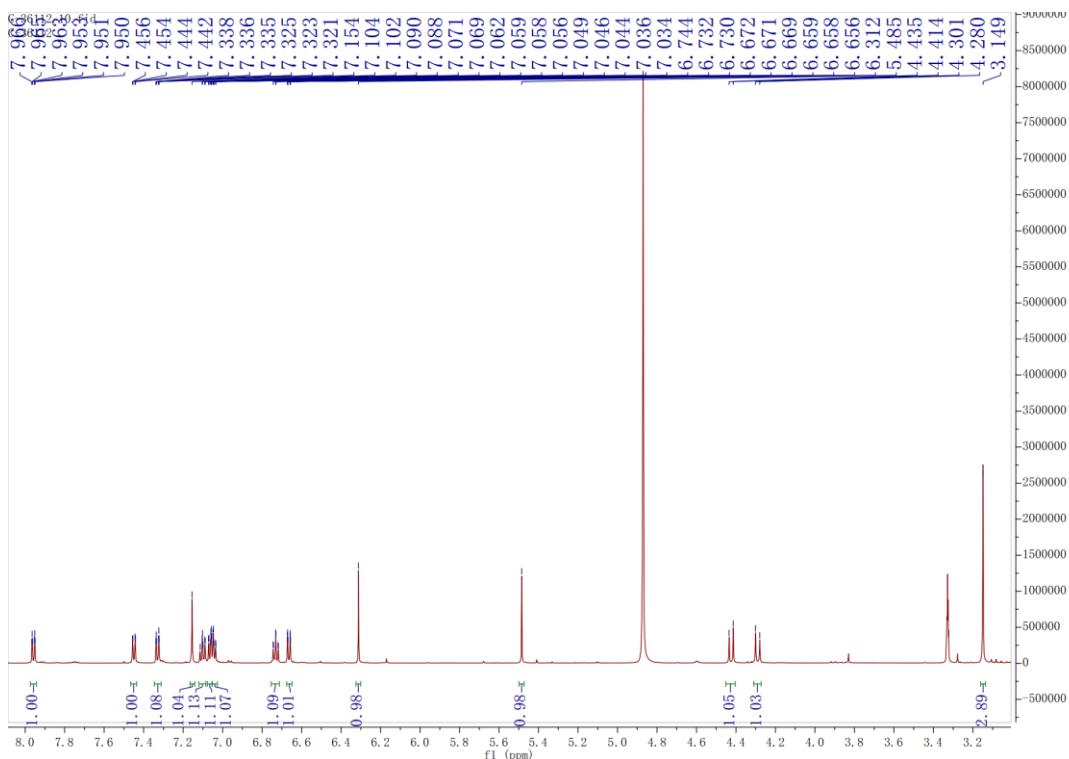
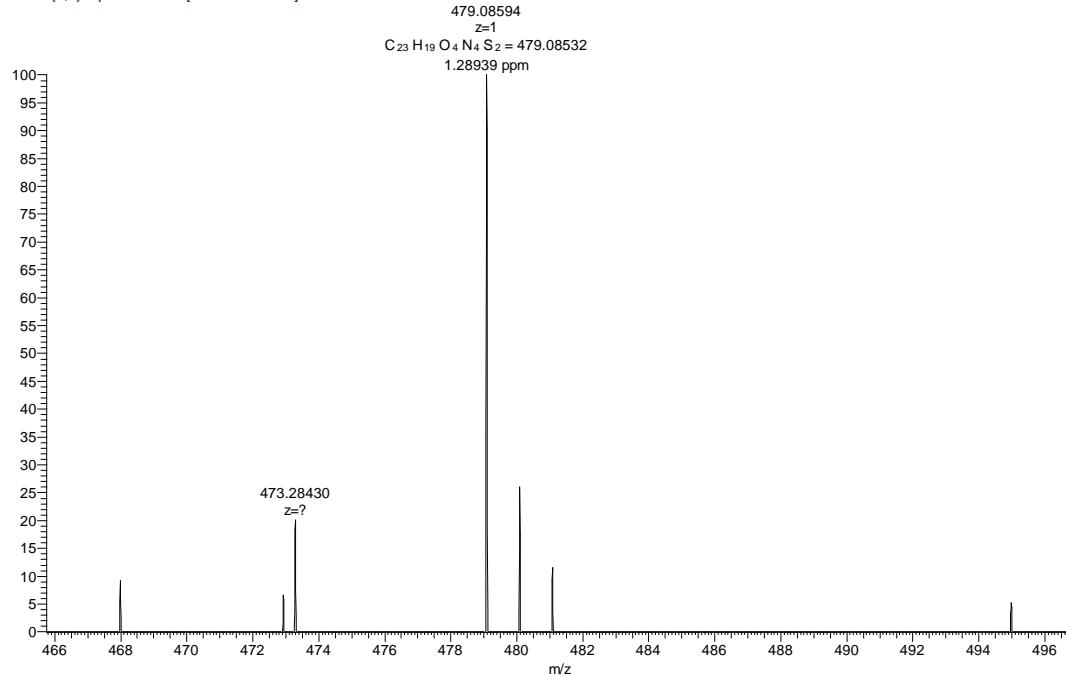


Figure S30. HR-ESI-MS spectrum of compound 5

**Figure S31.** ^1H NMR (600 MHz, CD_3OD) spectrum of compound **6**

C-36112 #10 RT: 0.13 AV: 1 NL: 2.88E3
T: FTMS (1,2) - p ESI Full ms [100.00-1000.00]

**Figure S32.** HR-ESI-MS spectrum of compound **6**

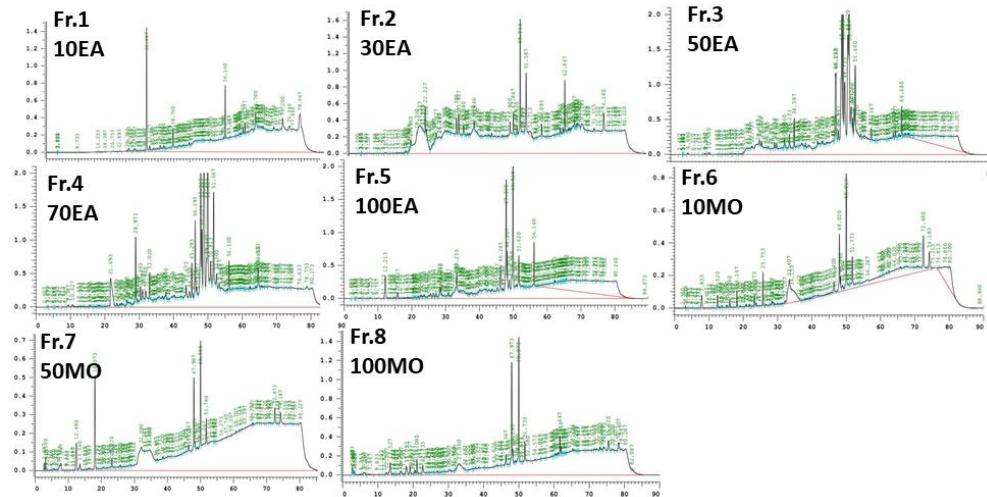


Figure S33. HPLC fingerprints of separated fractions Fr.1–Fr.8 of *A. luteoalbus* CH-6

Table S1. Identified compounds by molecular networking

Compound name	Adduct	Parent mass	CAS number	RT (min)
L-Phenylalanine, N-acetyl-Triethylcitratetriethyl 2-hydroxypropane-1,2,3-tricarboxylate	[M+H] ⁺	208.097	2018613	61.9
Dibutyl phthalate	[M+H] ⁺	277.128	77930	58.4
cyclo(D-Trp-L-Pro)	[M+Na] ⁺	284.139	509078493	63.5
L-Tyrosine	[M+H] ⁺	182.082	60184	6.1
CocamidopropylBetaine	[M+H] ⁺	343.296	N/A	53.4
1-Linoleoylglycerol	[M+H] ⁺	355.284	2258926	60.8
Monoolein	[M+H] ⁺	357.3	111035	51.0
Polanrazine B	[M] ⁺	377.32	394221002	59.4
Diethyl phthalate	[M+H] ⁺	391.285	117840	63.9
6-[3-[(3,4-dimethoxyphenyl)methyl]-4-methoxy-2-(methoxymethyl)butyl]-4-methoxy-1,3-benzodioxole	[M+H-H ₂ O] ⁺	415.211	N/A	44.5
His-Pro	[M+H-H ₂ O] ⁺	235.122	N/A	21.9
Decaethylene glycol	[M+H] ⁺	459.28	5579668	28.2
Cyclohexasiloxane, dodecamethyl	[M+NH ₄] ⁺	462.146	540976	34.2
Lyso-PC(16:0)	[M+H] ⁺	496.339	17364168	58.3
Undecaethylene glycol	[M+H] ⁺	503.311	6809707	29.4
1-(hexadecanoyloxy)-3-hydroxypropan-2-yl-octadec-9-	[M+NH ₄] ⁺	612.556	29541660	50.2

enoate					
Palmitamide	[M+H] ⁺	256.263	629549	55.1	
2,5-Piperazinedione, 3-(1 <i>H</i> -indol-3-ylmethyl)-6-methyl-	[M+H] ⁺	258.124	17079377	24.4	
cyclo(L-Tyr-L-Pro)	[M+H] ⁺	261.123	4549024	26.3	
cyclo(Phe-4-Hyp)	[M+H] ⁺	261.124	N/A	26.4	

Table S2. OR values of compounds **4–6**

Compounds	4	5	6
[α] _D ²⁰	+239 (0.1, CHCl ₃)	+561 (0.1, pyridine)	+312 (0.1, MeOH)

Table S3. Antimicrobial activities of the separated fractions (50 µg/mL) of *A. luteoalbus* CH-6

Fractions	<i>A. salmonicida</i>	<i>C. albicans</i>
1	-1.23±0.40	3.52±6.20
2	75.75±0.40	66.67±0.40
3	81.02±0.60	72.18±0.25
4	80.58±0.65	70.42±0.10
5	74.43±0.35	69.95±0.30
6	76.1±1.00	71.48±0.15
7	72.26±0.10	73.47±0.40
8	72.43±0	71.58±0.30
3.1	23.12±1.11	31.56±0.32
3.2	27.59±0.20	54.93±0.40
3.3	5.89±1.35	61.5±6.40
3.4	55.1±2.35	39.52±0.05
3.5	77.33±0.70	73.83±1.15
3.6	82.67±0.72	75.52±1.15
3.7	67.98±0.40	63.31±0.82
3.8	69±0	54.55±2.15
3.5.1	72.06±0.50	66.55±0.75
3.5.2	83.22±0.15	78.64±0.60
3.5.3	78.42±0.45	75.71±0.10
3.5.4	-0.08±1.05	43.45±9.95
3.6.1	80.58±1.75	66.59±1.75
3.6.2	85.67±1.30	76.27±1.15
3.6.3	71.25±0.15	52.18±1.45
3.6.4	23.42±1.75	8.03±0.95