SUPPORTING INFORMATION

Synthesis of a Tricyclic Pterolobirin H Analogue: Evaluation of Anticancer and Anti-inflammatory Activities and Molecular Docking Investigations

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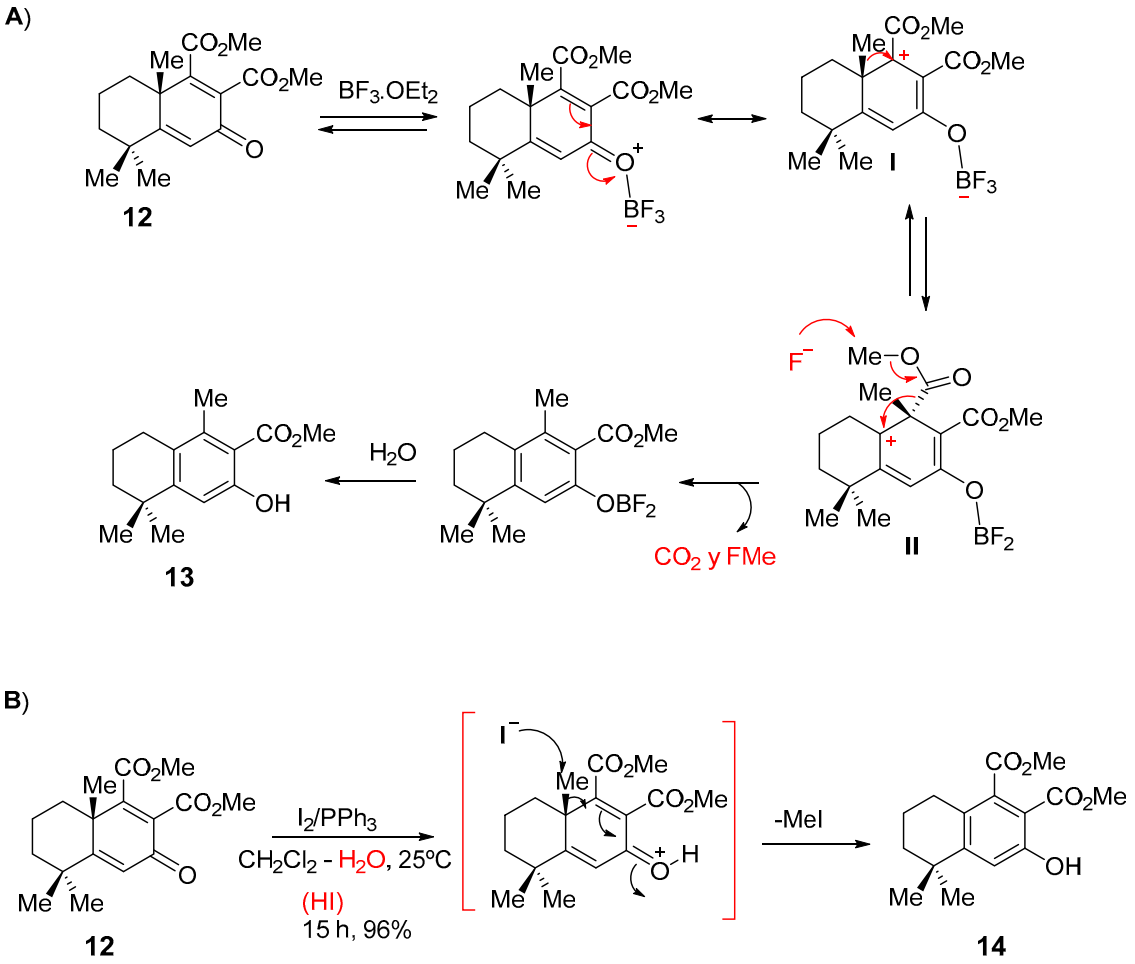


Figure S1. Proposed Mechanisms for the obtention of phenols **13** (A) and **14** (B) from dienone **12**.

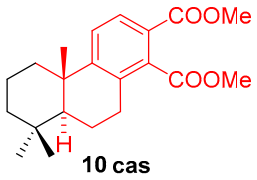
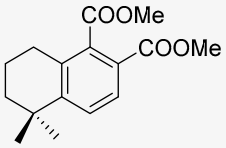
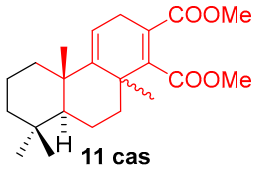
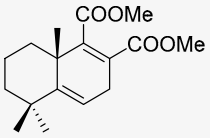
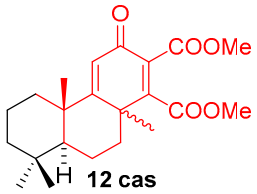
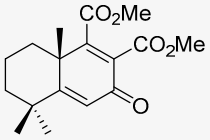
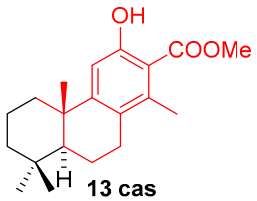
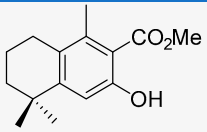
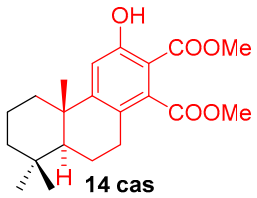
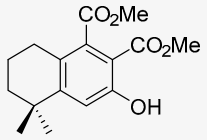
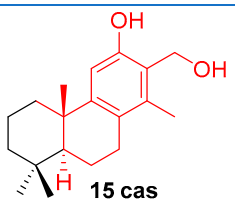
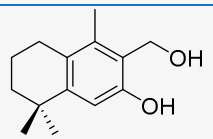
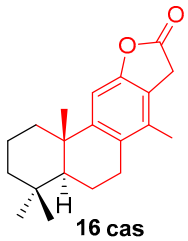
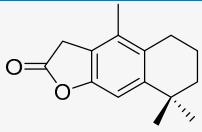
Compound	Simplified analogue
 10 cas	 10
 11 cas	 11
 12 cas	 12
 13 cas	 13
 14 cas	 14
 15 cas	 15
 16 cas Pterolobirin H (**3**)	 Simplified analogue of pterolobirin H -**16**

Table S1. Pterolobirin H (**3**), its intermediates from our previous report [1] and the simplified analogues.

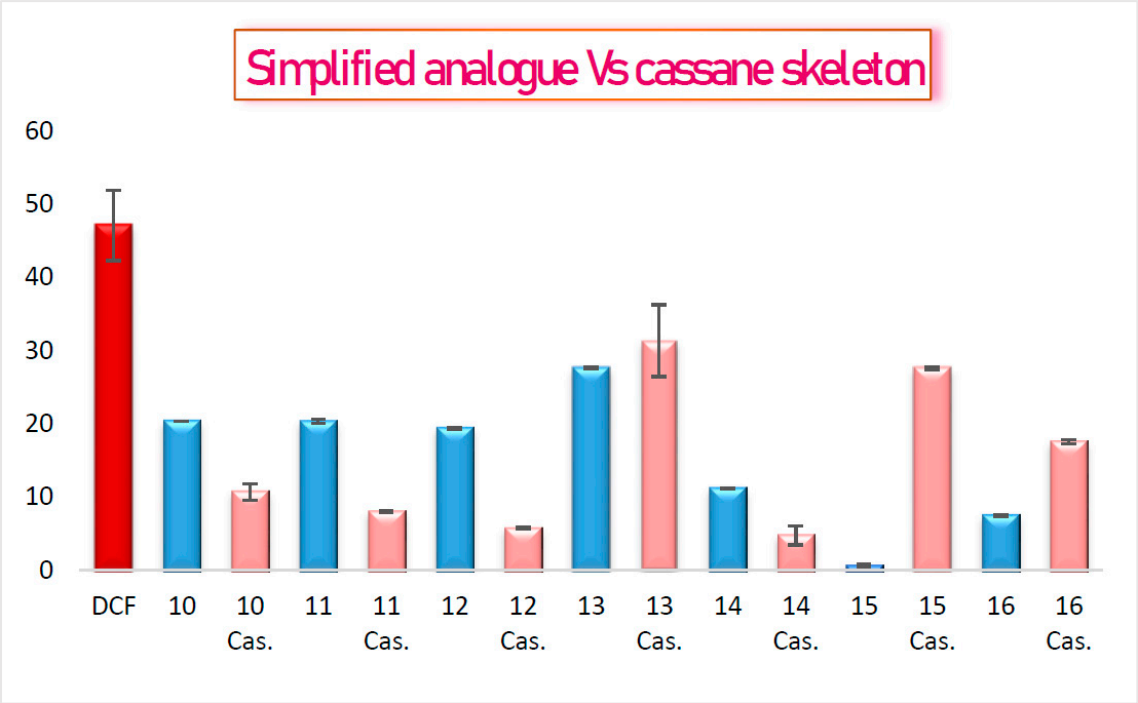


Figure S2. NO release-inhibitory effects $IC_{50\ NO}$ (µg/mL) concentrations for DCF, simplified analogues (**10-16**), and correspondent cassane diterpenoids (**10 cas -16 cas/ pterolobirin H (3)**). The data represent the mean ± SD of at least two independent experiments performed in triplicate.

Simplified analogue	IC_{50NO} (μg/mL)	Cassane skeleton	IC_{50NO} (μg/mL)
10	20.37 ± 0.01	**10 Cas.**	10.71 ± 1.14
11	20.34 ± 0.30	**11 Cas.**	7.98 ± 0.10
1**2**	19.44 ± 0.12	**12 Cas.**	5.71 ± 0.14
13	27.60 ± 0.13	**13 Cas.**	31.37 ± 4.90
14	11.18 ± 0.09	**14 Cas.**	4.74 ± 1.27
15	**0.62 ± 8.21**	**15 Cas.**	**27.57 ± 0.20**
16	**7.49 ± 0.15**	**16 Cas. (pterolobirin H (3))**	**17.57 ± 0.31**

Table S2. The $IC_{50\ NO}$ values of cassanes **10** cas–**16** cas, and their simplified analogues **10**–**16**.

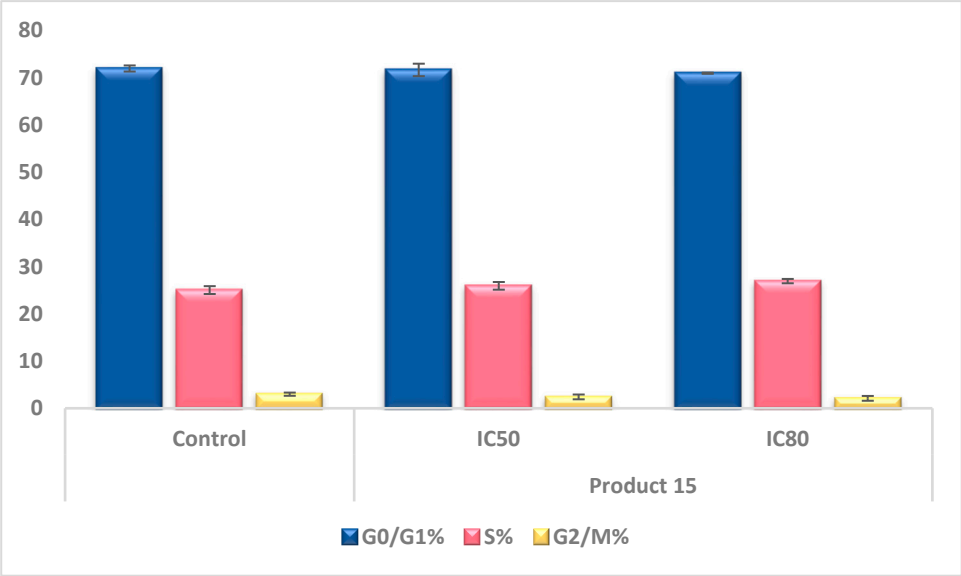


Figure S3. Top: Percentages of B16-F10 skin-melanoma cells in each cell-cycle phase, after 72 h treatment with compound **15**, at IC_{50} and IC_{80} concentrations. G0/G1 phase (Blue), S phase (pink), and G2/M phase (peach). Control (untreated cells); samples (cells treated with compounds **15**). Values represent means ± S. D. of at least three independent experiments performed in duplicate.

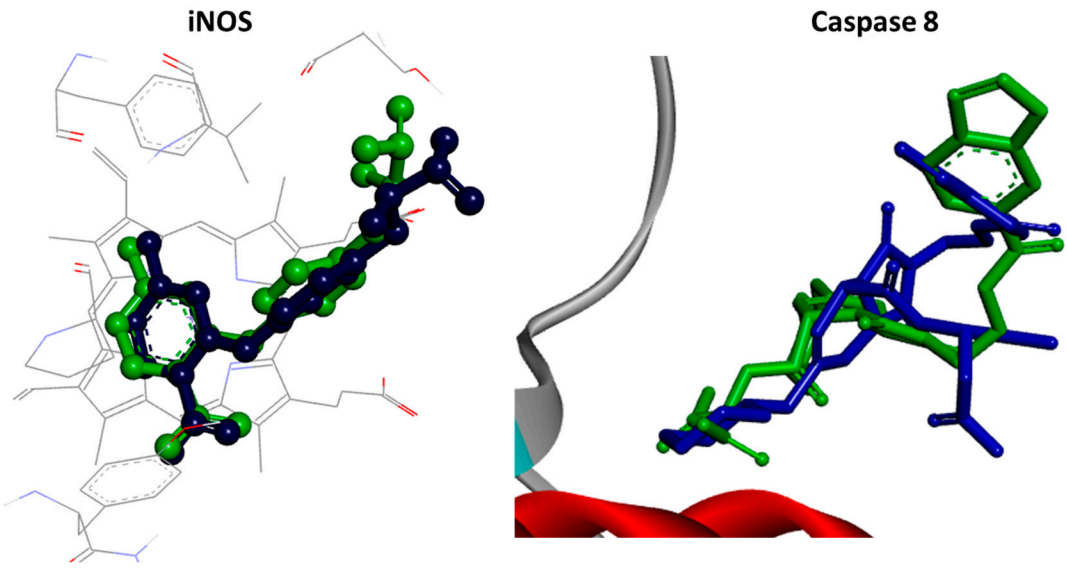


Figure S4. Poses of cognate YWO ligand on 4UX6 crystal structure (blue) and redocked (green).

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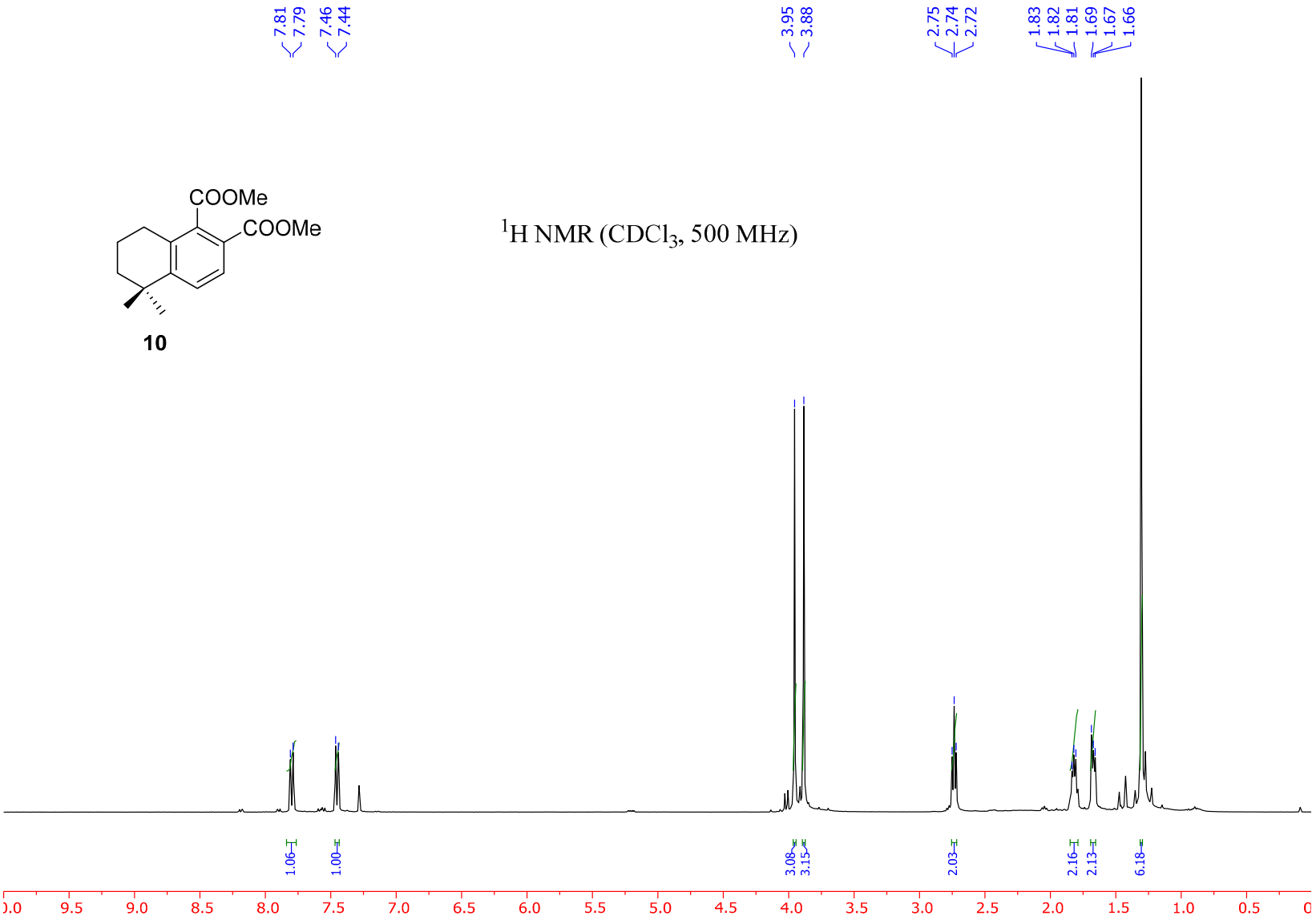
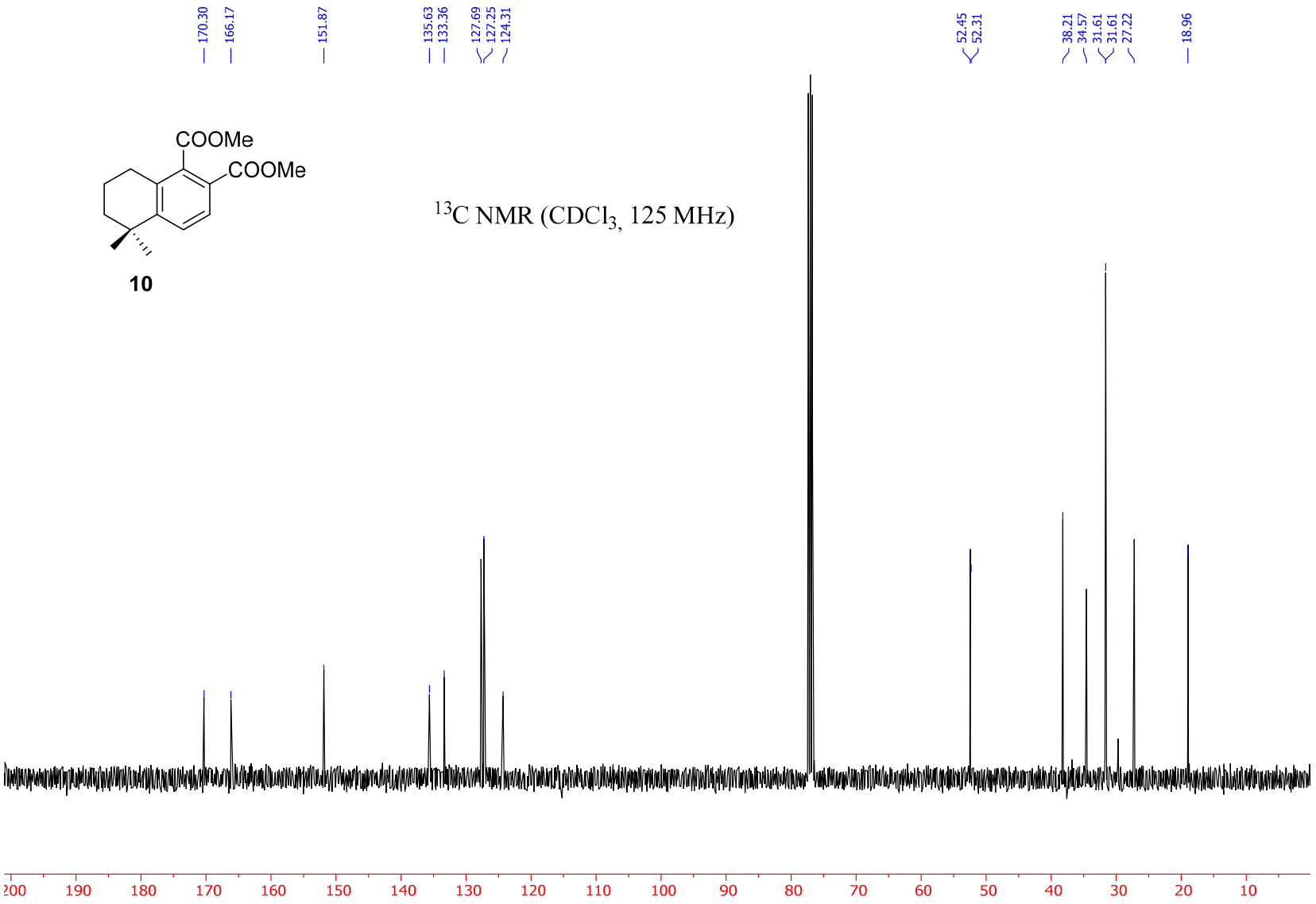
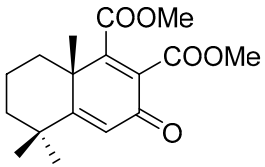

7.81
7.79
7.46
7.44
3.95
3.88
2.75
2.74
2.72
1.83
1.82
1.81
1.69
1.67
1.66
COOMe
COOMe
10
^{1}H NMR ($CDCl_3$, 500 MHz)
1.06
1.00
3.08
3.15
2.03
2.16
2.13
6.18
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5


170.30
166.17
151.87
135.63
133.36
127.69
127.25
124.31
52.45
52.31
38.21
34.57
31.61
31.61
27.22
18.96
COOMe
COOMe
10
^{13}C NMR ($CDCl_3$, 125 MHz)
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10

^{1}H NMR ($CDCl_3$, 500 MHz)



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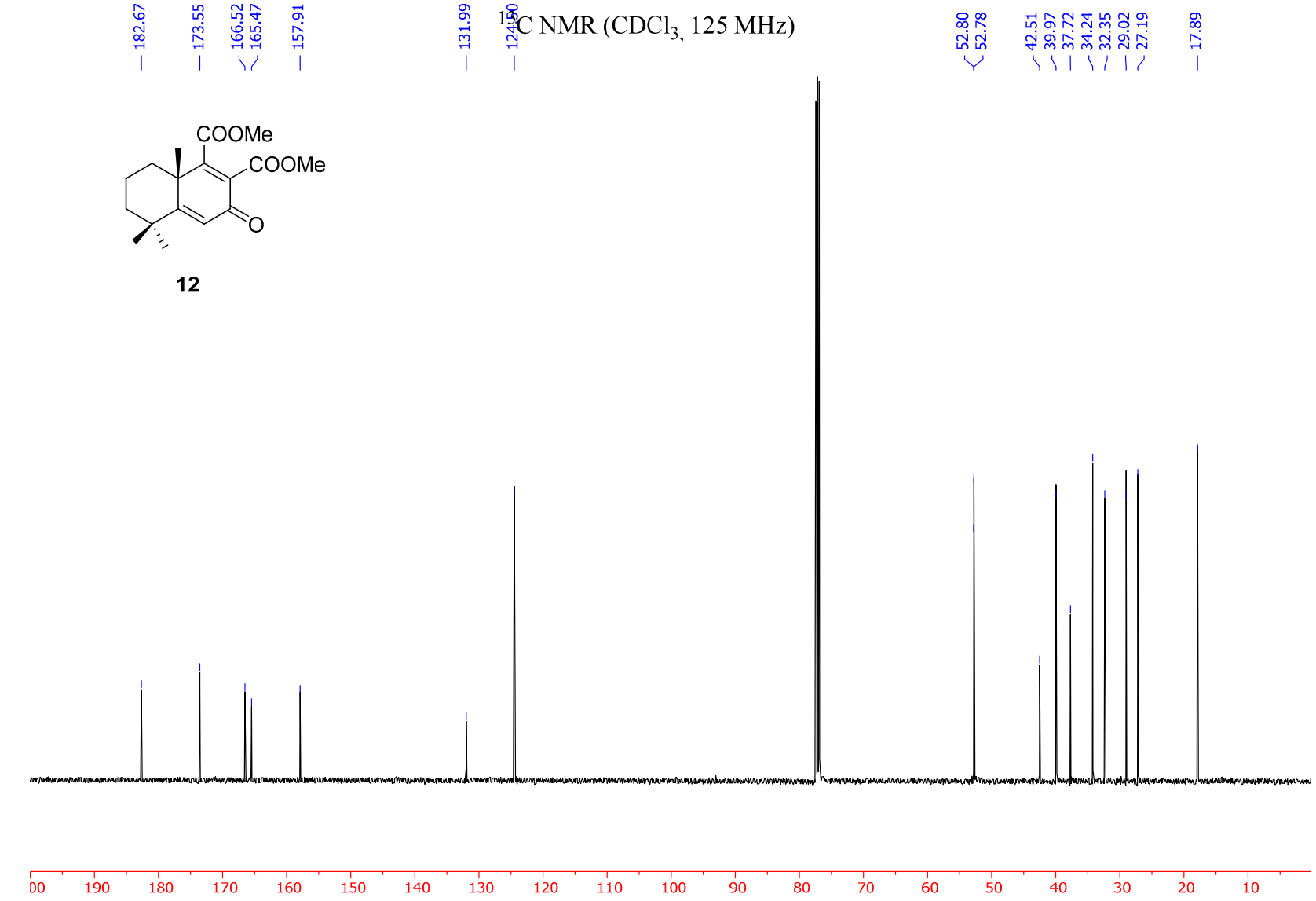
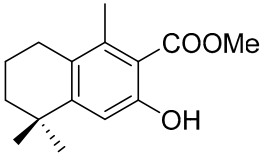

182.67
173.55
166.52
165.47
157.91
131.99
124.50
52.80
52.78
42.51
39.97
37.72
34.24
32.35
29.02
27.19
17.89
COOMe
COOMe
O
12
^{13}C NMR ($CDCl_3$, 125 MHz)
00
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10

^{1}H NMR ($CDCl_3$, 500 MHz)



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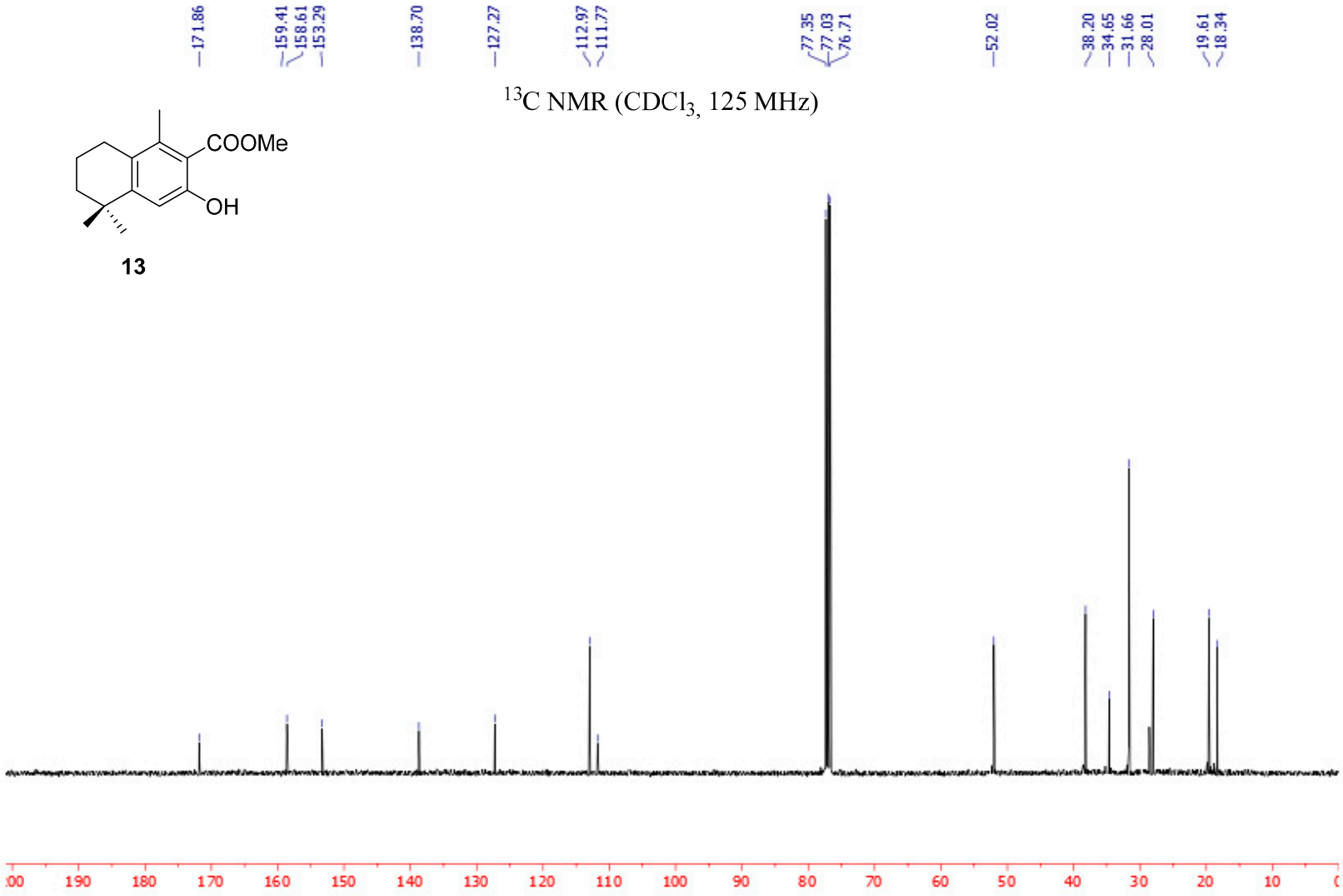
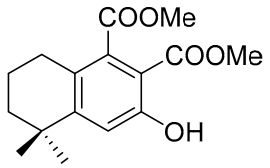

171.86
159.41
158.61
153.29
138.70
127.27
112.97
111.77
77.35
77.03
76.71
52.02
38.20
34.65
31.66
28.01
19.61
18.34
COOMe
OH
13
^{13}C NMR ($CDCl_3$, 125 MHz)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10

^{1}H NMR ($CDCl_3$, 500 MHz)



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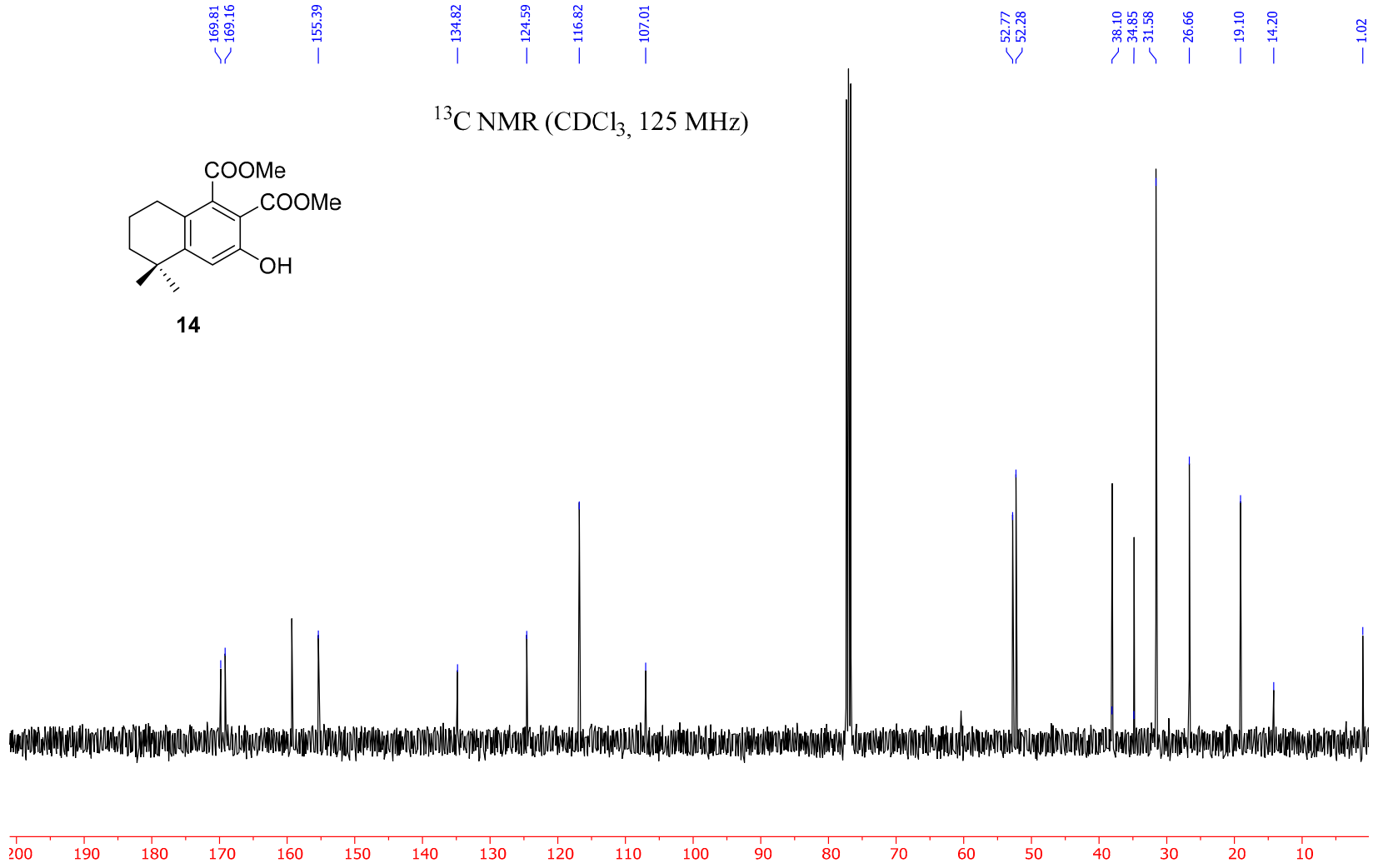
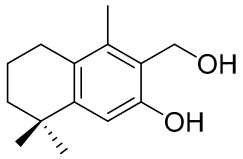

169.81
169.16
155.39
134.82
124.59
116.82
107.01
52.77
52.28
38.10
34.85
31.58
26.66
19.10
14.20
1.02
^{13}C NMR ($CDCl_3$, 125 MHz)
COOMe
COOMe
OH
14
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10

^{1}H NMR (CDCl$_3$, 500 MHz)



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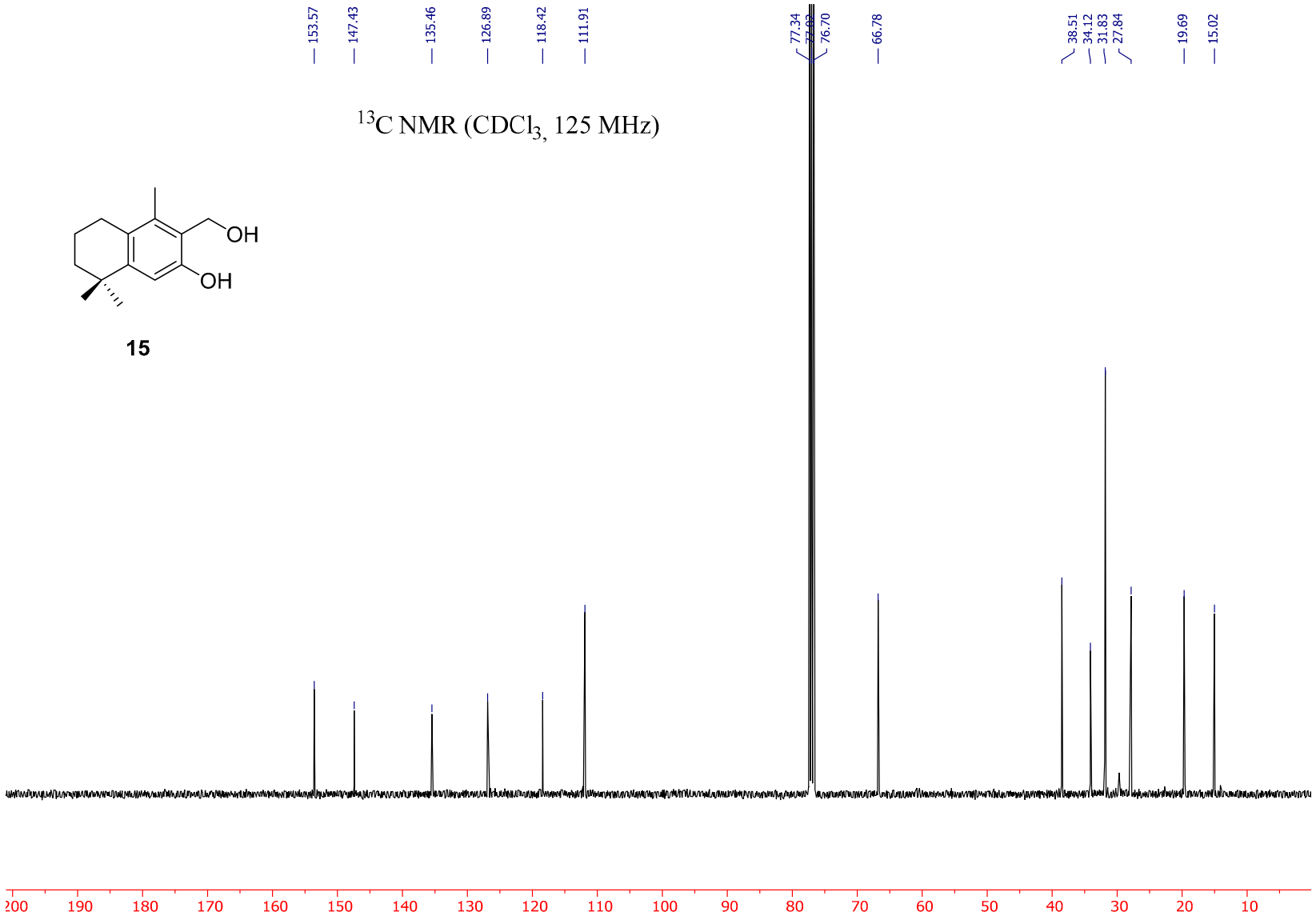
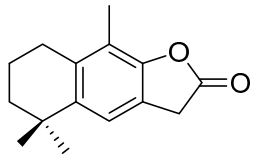


^{13}C NMR ($CDCl_3$, 125 MHz)

15

^{1}H NMR ($CDCl_3$, 500 MHz)



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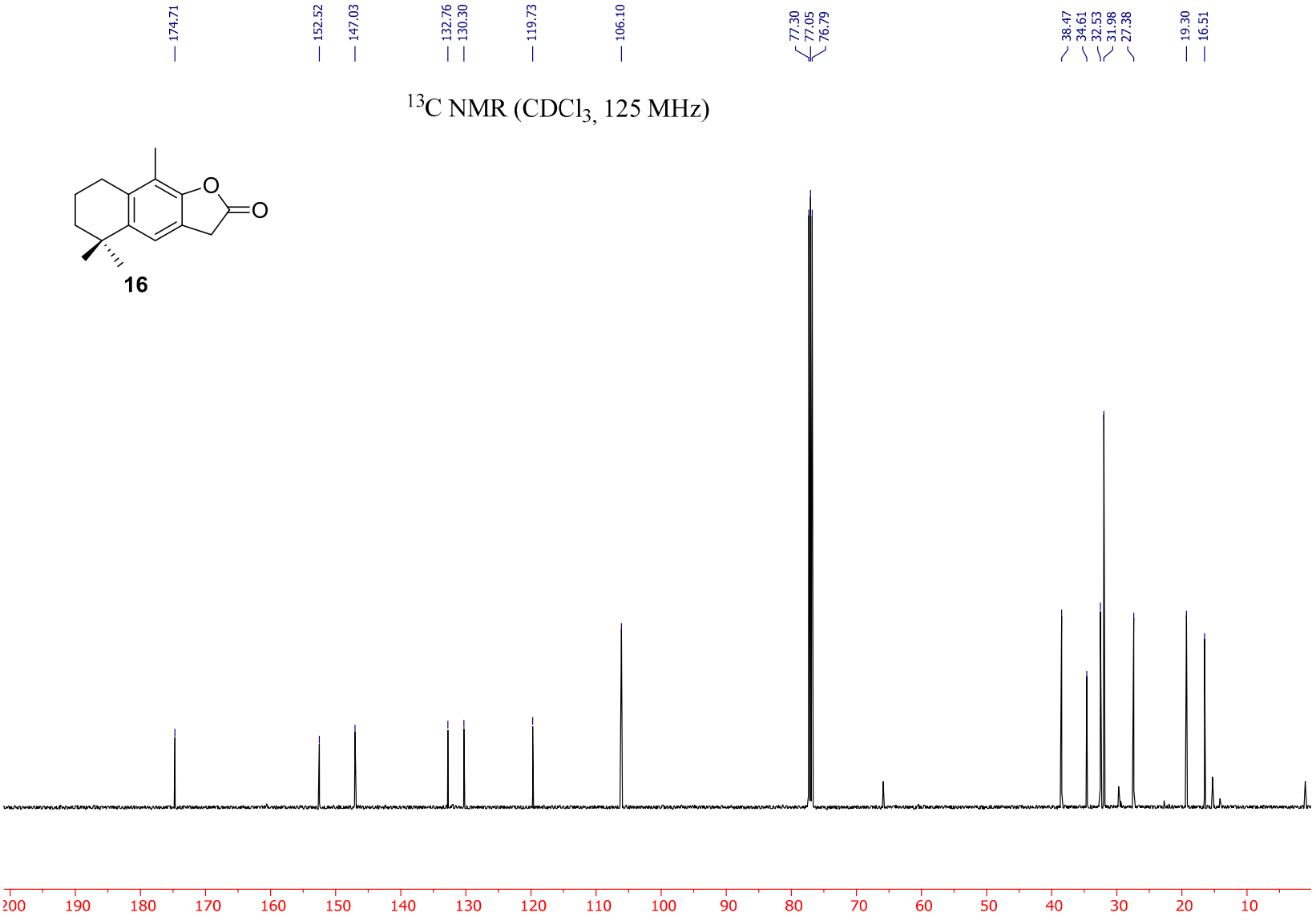
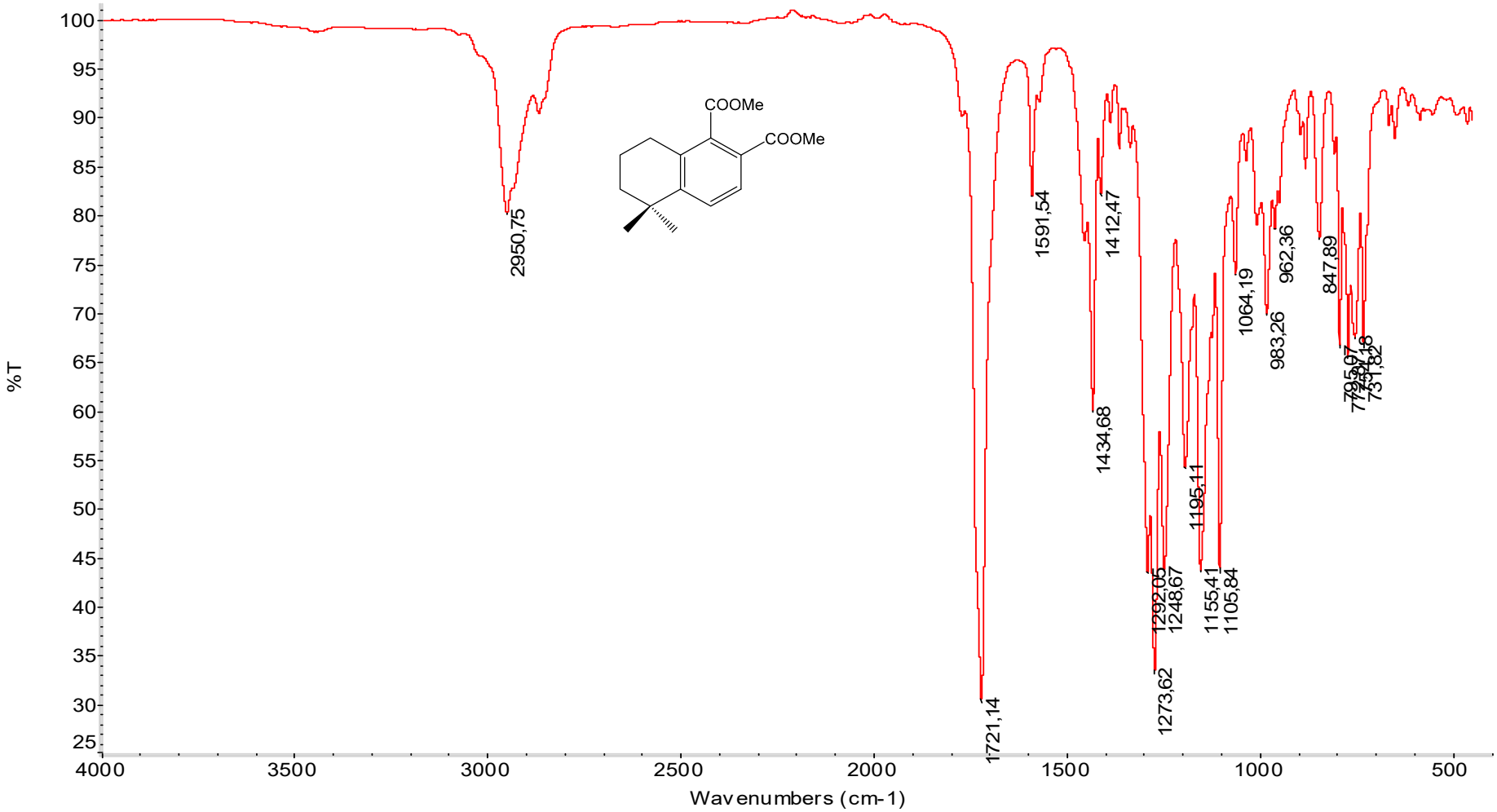
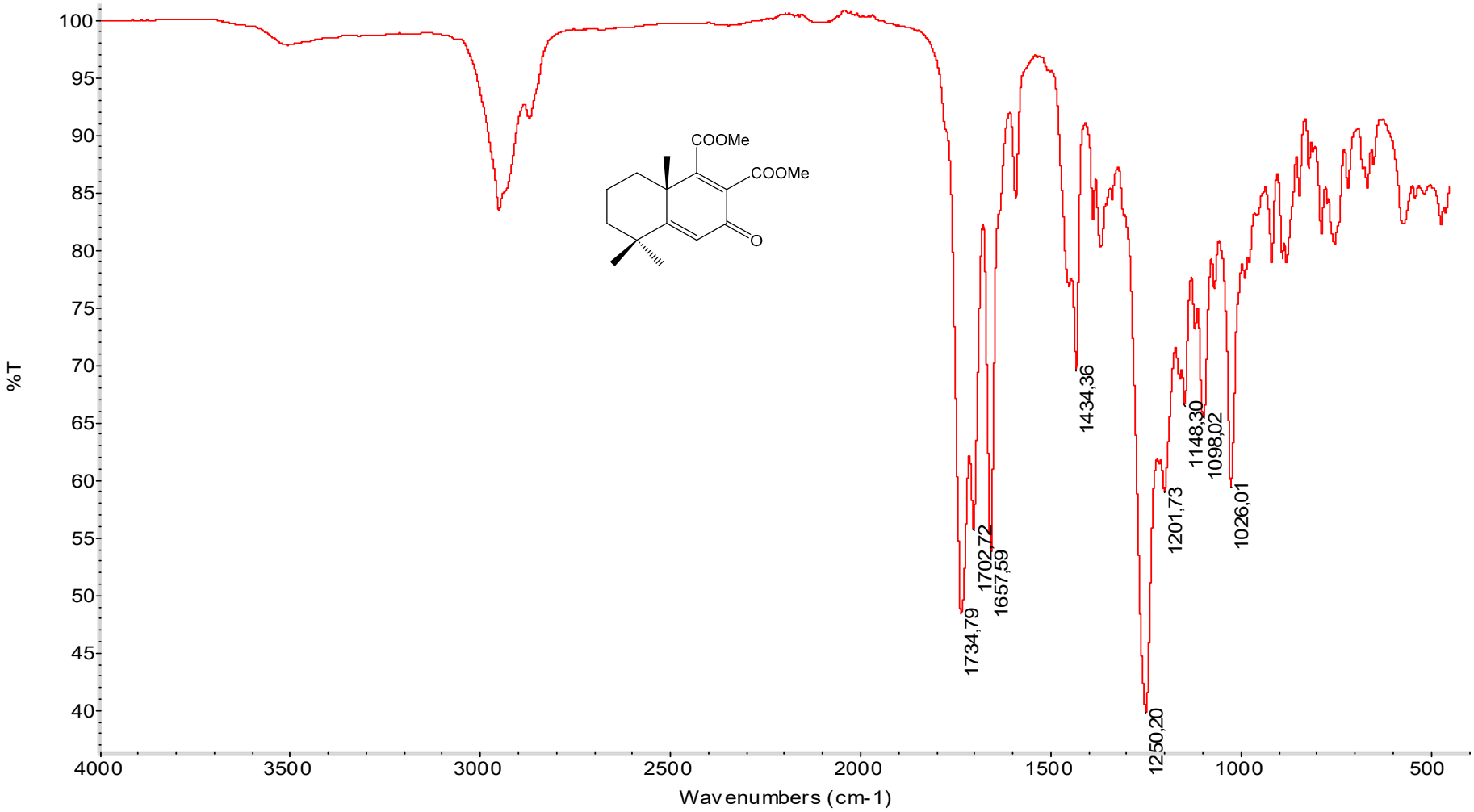
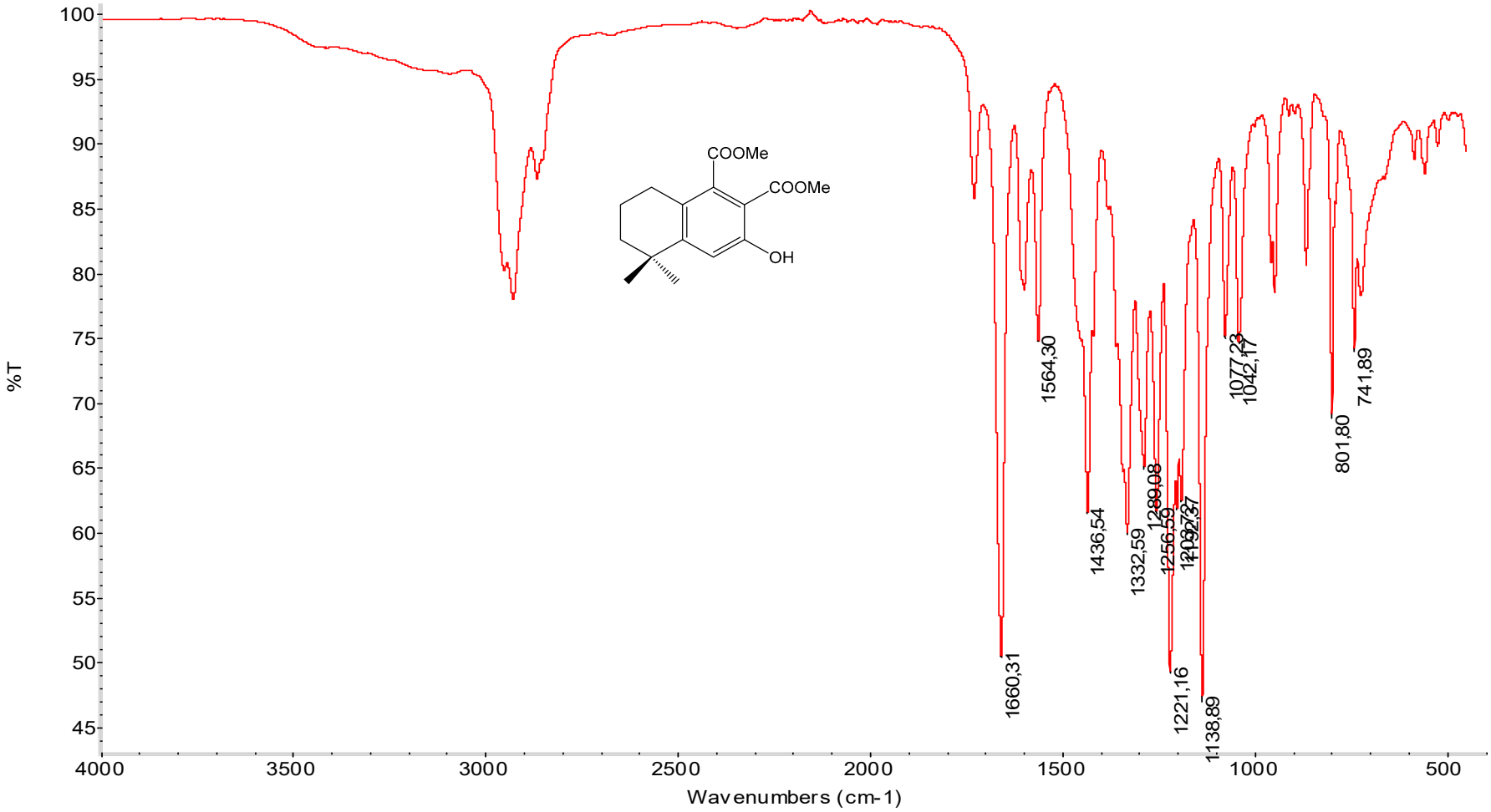


^{13}C NMR ($CDCl_3$, 125 MHz)

Infrared spectra of Compounds 10, 12, 13, 14, 15, and 16




100
95
90
85
80
75
70
65
60
55
50
45
40
%T
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
COOMe
COOMe
O
1734,79
1702,72
1657,59
1434,36
1250,20
1201,73
1148,30
1098,02
1026,01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95
90
85
80
75
70
65
60
55
50
45
%T
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
COOMe
COOMe
OH
1660,31
1564,30
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1332,59
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1221,16
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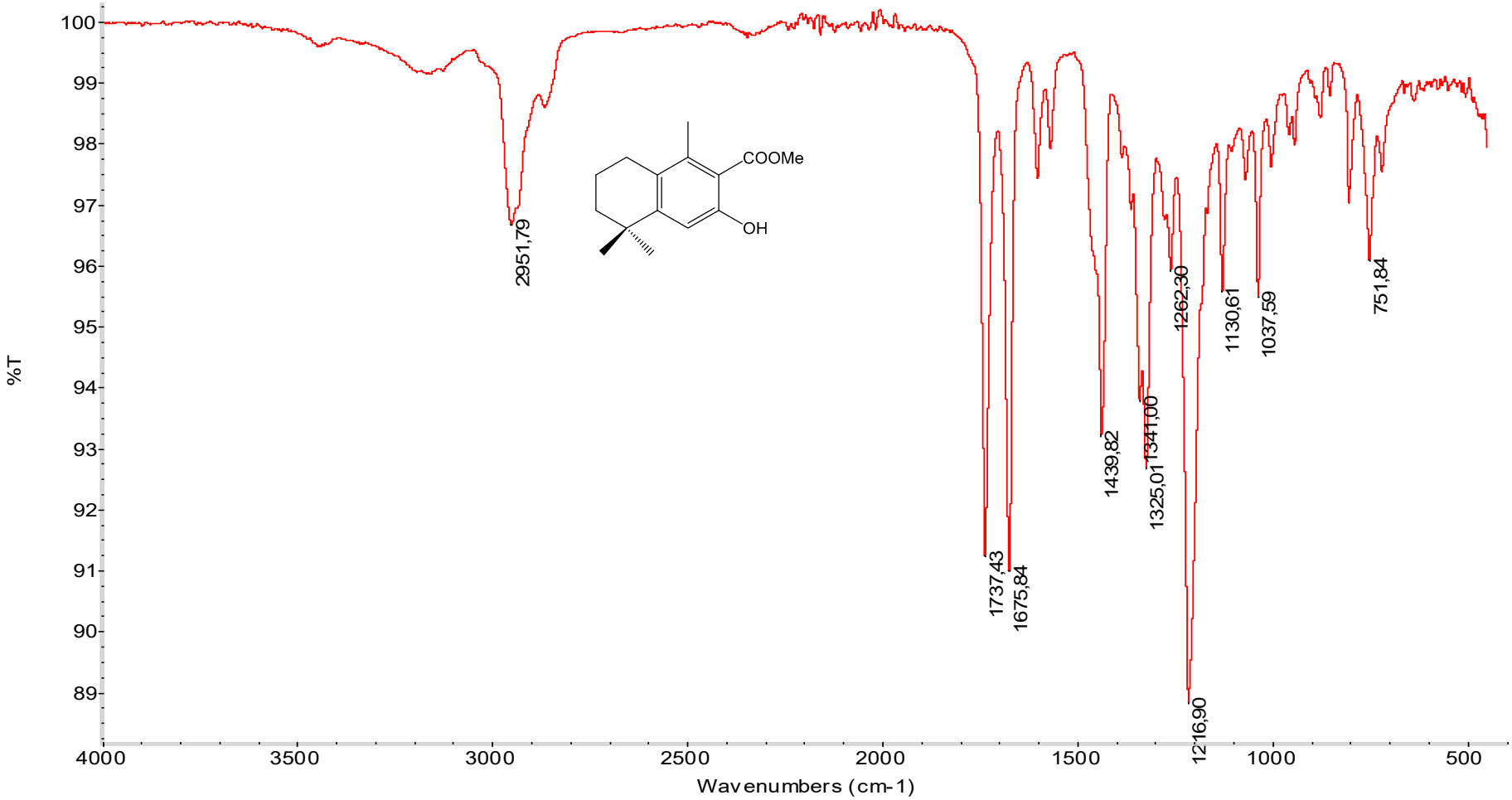
1077,23
1042,17
801,80
741,89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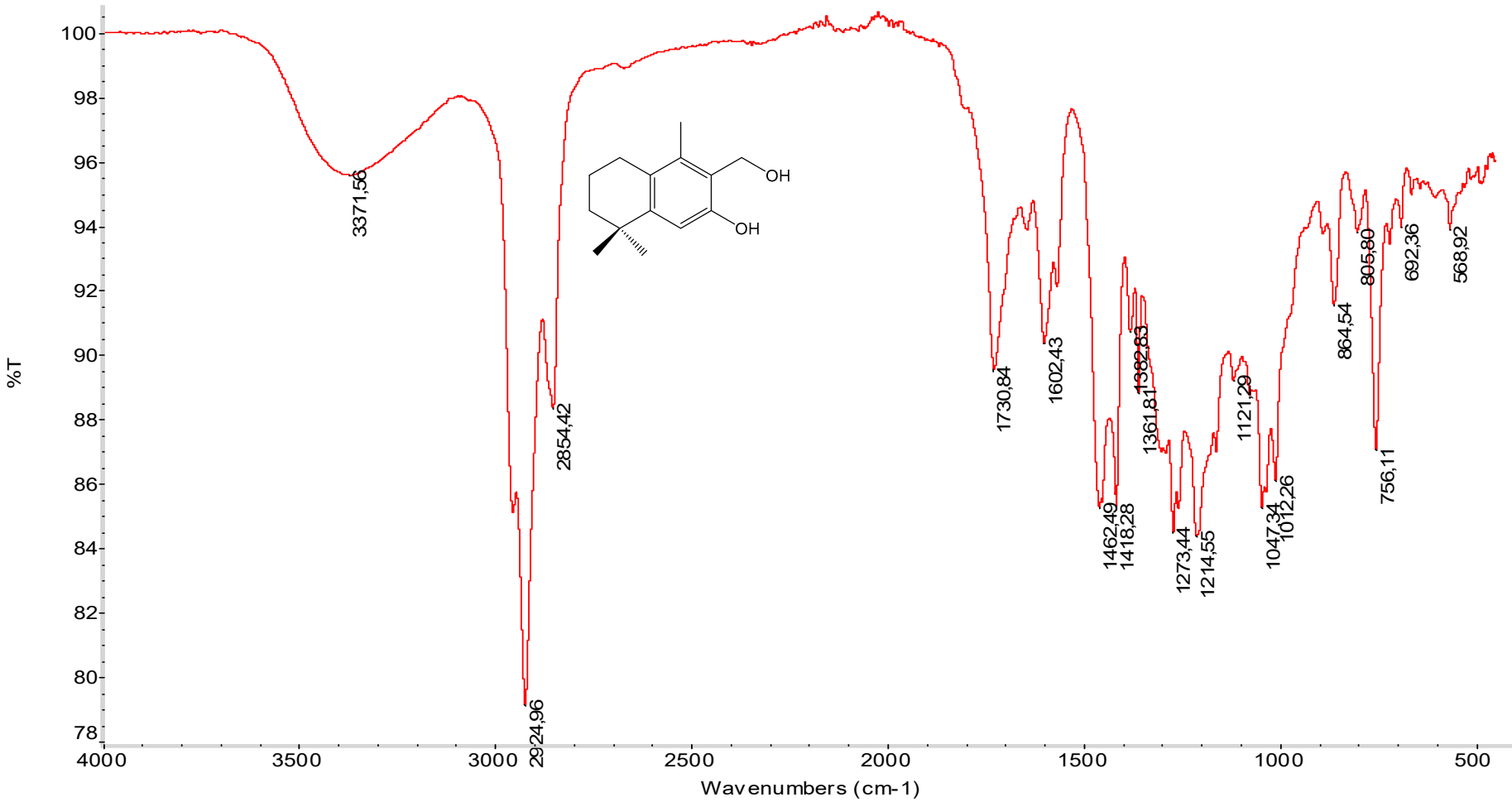
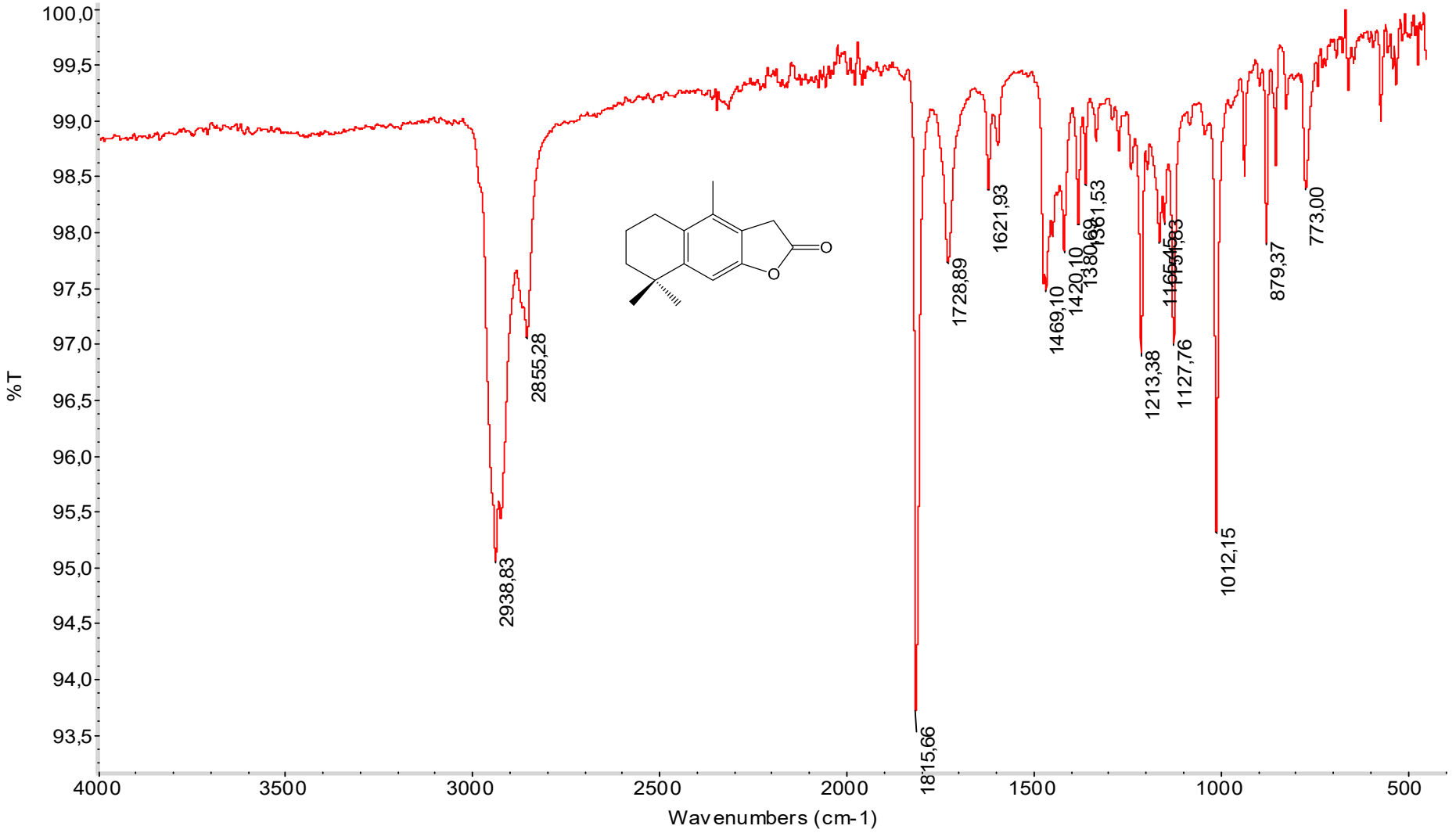
99
98
97
96
95
94
93
92
91
90
89
%T
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
COOMe
OH
2951,79
1737,43
1675,84
1439,82
1341,00
1325,01
1262,30
1216,90
1130,61
1037,59
751,84


%T
100
98
96
94
92
90
88
86
84
82
80
78
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
3371,56
2924,96
2854,42
1730,84
1602,43
1462,49
1418,28
1382,83
1361,81
1273,44
1214,55
1121,29
1047,34
1012,26
864,54
805,80
756,11
692,36
568,92
OH
OH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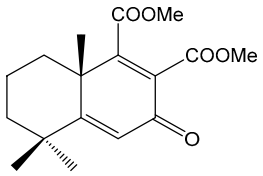
96,0
95,5
95,0
94,5
94,0
93,5
%T
4000
3500
3000
2500
2000
1500
1000
500
Wavenumbers (cm-1)
2938,83
2855,28
1815,66
1728,89
1621,93
1469,10
1420,10
1380,68
1381,53
1213,38
1127,76
1012,15
879,37
773,00
O
O

HRMS spectra of compounds 12,13, 14, 15, and 16

Elemental Composition Report



Single Mass Analysis
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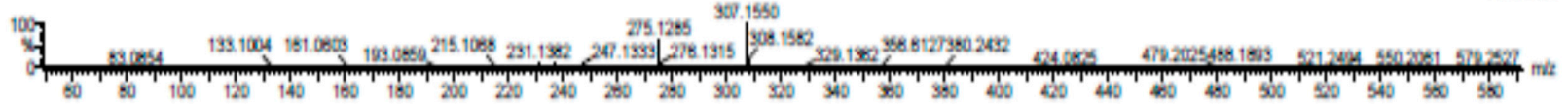
Element prediction: Off
Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Even Electron Ions
202 formula(e) evaluated with 5 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-50 H: 0-100 O: 0-25 23Na: 0-1

20_12_16 1 1264 (5.878)

1: TOF MS ES+
7.08e+006



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Maximum:		20.0	25.0	50.0				
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
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	307.1521	2.9	9.4	3.5	752.7	4.020	1.80	C15 H24 O5 23Na
	307.1580	-3.0	-9.8	-5.5	756.3	7.633	0.05	C8 H28 O10 23Na
	307.1604	-5.4	-17.6	-2.5	756.1	7.465	0.06	C10 H27 O10
	307.1487	6.3	20.5	15.5	758.5	9.871	0.01	C24 H19

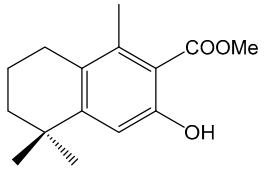
Elemental Composition Report

Single Mass Analysis

Tolerance = 25.0 PPM / DBE: min = -20.0, max = 50.0

Element prediction: Off

Number of isotope peaks used for i-FIT = 3



Monoisotopic Mass, Even Electron Ions

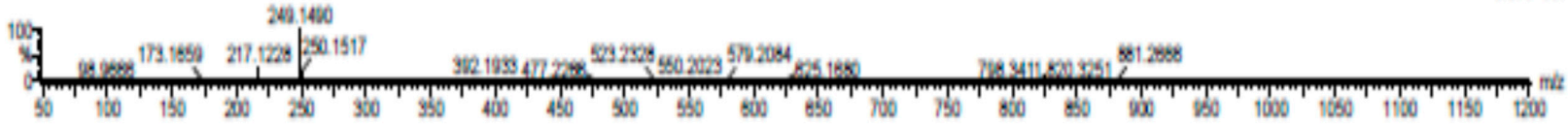
146 formula(e) evaluated with 4 results within limits (all results (up to 1000) for each mass)

Elements Used:

C: 0-50 H: 0-100 O: 0-25 23Na: 0-1

20_12_18 2 1538 (7.139)

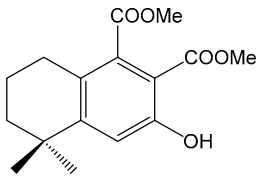
1: TOF MS ES+
3.87e+007



Minimum:				-20.0				
Maximum:		20.0	25.0	50.0				
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
249.1490	249.1491	-0.1	-0.4	5.5	968.6	0.005	99.48	C15 H21 O3
	249.1467	2.3	9.2	2.5	973.9	5.260	0.52	C13 H22 O3 23Na
	249.1525	-3.5	-14.0	-6.5	982.3	13.653	0.00	C6 H26 O8 23Na
	249.1549	-5.9	-23.7	-3.5	981.1	12.501	0.00	C8 H25 O8

Single Mass Analysis

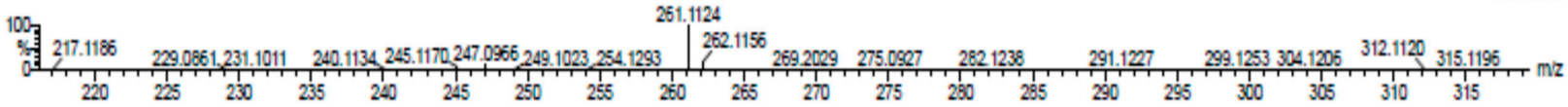
Tolerance = 25.0 PPM / DBE: min = -20.0, max = 50.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 3



Monoisotopic Mass, Even Electron Ions
211 formula(e) evaluated with 7 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-50 H: 0-100 O: 0-25 Na: 0-1

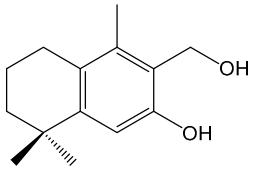
20_12_16 4 REP 1492 (6.937)

1: TOF MS ES+
3.30e+007



Minimum:				-20.0				
Maximum:		20.0	25.0	50.0				
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
315.1196	315.1197	-0.1	-0.3	-12.5	685.7	12.835	0.00	H27 O18
	315.1208	-1.2	-3.8	6.5	673.3	0.405	66.69	C16 H20 O5 Na
	315.1174	2.2	7.0	18.5	682.3	9.463	0.01	C25 H15
	315.1232	-3.6	-11.4	9.5	674.0	1.106	33.10	C18 H19 O5
	315.1150	4.6	14.6	15.5	681.5	8.656	0.02	C23 H16 Na
	315.1139	5.7	18.1	-3.5	680.0	7.188	0.08	C7 H23 O13
	315.1267	-7.1	-22.5	-2.5	679.7	6.824	0.11	C9 H24 O10 Na

Elemental Composition Report



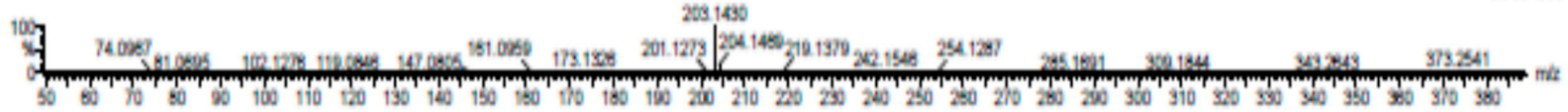
Single Mass Analysis

Tolerance = 25.0 PPM / DBE: min = -20.0, max = 50.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Even Electron Ions
128 formula(e) evaluated with 3 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-50 H: 0-100 O: 0-25 Na: 0-1

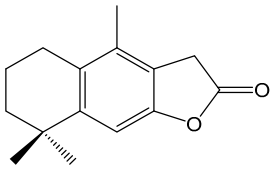
20_12_16 5 2145 (9.975)

1: TOF MS ES+
6.70e+006



Minimum:				-20.0				
Maximum:		20.0	25.0	50.0				
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
219.1379	219.1385	-0.6	-2.7	5.5	387.9	0.003	99.68	C14 H19 O2
	219.1361	1.8	8.2	2.5	393.6	5.741	0.32	C12 H20 O2 Na
	219.1420	-4.1	-18.7	-6.5	404.5	16.651	0.00	C5 H24 O7 Na

Elemental Composition Report



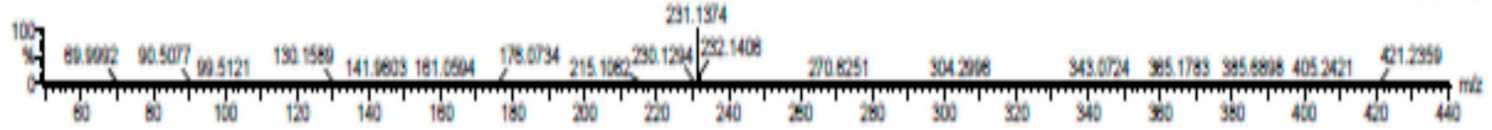
Single Mass Analysis

Tolerance = 25.0 PPM / DBE: min = -20.0, max = 50.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Even Electron Ions
138 formula(e) evaluated with 3 results within limits (all results (up to 1000) for each mass)
Elements Used:
C: 0-50 H: 0-100 O: 0-25 Na: 0-1

20_12_18 7 1616 (7.509)

1: TOF MS ES+
3.17e+006



Minimum:				-20.0				
Maximum:		20.0	25.0	50.0				
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
231.1374	231.1385	-1.1	-4.8	6.5	664.1	0.003	99.70	C15 H19 O2
	231.1361	1.3	5.6	3.5	669.9	5.812	0.30	C13 H20 O2 Na
	231.1420	-4.6	-19.9	-5.5	680.4	16.324	0.00	C6 H24 O7 Na