Hoogsteen triplexes stabilized through ethynyl-linked pyrene-indole synthesized by high-temperature Sonogashira coupling

Imrich Géci,*[a,c] Maha I. Fatthalla,[b,c,d] Maike Heintz,[c] Per T. Jørgensen[c] and Erik B. Pedersen[c]

[a] Department of Medical and Clinical Biophysics, Faculty of Medicine, P. J. Šafárik University in Košice, Trieda SNP 1, 04011 Košice, Slovakia.

[b] Department of Chemistry, Faculty of Science, Helwan University, 11795 Ain Helwan, Cairo, Egypt.

[c] Department of Physics, Chemistry and Pharmacy, University of Southern Denmark, Campusvej 55, DK-5230, Odense M, Denmark.

[d] Universite Paris-Sud, CNRS, Faculte de Pharmacie, 5 rue J.-B. Clement, Chatenay-Malabry, 92296 France

E-mail: imrich.geci@upjs.sk

Electronic Supplementary Information

Contents

Numerical listing of NMR spectra:

(S)-4-tosyloxy-1,2-O-isopropylidene-1,2-butanediol (3).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.80 – 7.78 (m, 2H, Ar), 7.36 – 7.34 (m, 2H, Ar), 4.20 – 4.08 (m, 3H, CH_2OTs + CH_2CHO), 4.01 (dd, *J* = 8.1, 6.0 Hz, 1H, CH*H*O), 3.51 (dd, *J* = 8.1, 6.9 Hz, 1H, C*H*HO), 2.45 (s, 3H, CH_3), 1.93 – 1.85 (m, 2H, CH_2CH_2N), 1.34 (d, *J* = 0.7 Hz, 3H, CH_3), 1.29 (d, *J* = 0.8 Hz, 3H, CH_3). The ^{1}H-NMR data is consistent with the literature data.[1]

^{13}C NMR (101 MHz, $CDCl_3$) δ 144.85, 132.92, 129.88, 127.89 (OTs), 108.99 ($C(CH_3)_2$), 72.28 ($CHOC(CH_3)_2$), 69.06 (CH_2O), 67.43 (CH_2OTs), 33.13 (CH_2CH_2N), 26.83 (CH_3), 25.53 (CH_3), 21.62 (CH_3).

(S)-1-(2-(2,2-dimethyl-1,3-dioxolan-4-yl)ethyl)-5-iodo-1H-indole (5).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.93 (d, *J* = 1.6 Hz, 1H, Ar), 7.41 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar), 7.12 (d, *J* = 8.7 Hz, 1H, Ar), 7.05 (d, *J* = 3.1 Hz, 1H, Ar), 6.39 (dd, *J* = 3.2, 0.9 Hz, 1H, Ar), 4.22 (m, 2H, CH_2N), 3.94 – 3.88 (m, 2H, CH_2O), 3.45 – 3.41 (m, 1H, CH_2CHO), 2.01 – 1.92 (m, 2H, CH_2CH_2N), 1.44 (s, 3H, CH_3), 1.32 (s, 3H, CH_3).

^{13}C NMR (101 MHz, $CDCl_3$) δ 135.10, 131.21, 129.83, 128.79, 111.50, 109.22, 100.75 (Ar + $C(CH_3)_2$), 82.97 (C-I), 72.93 ($CHOC(CH_3)_2$), 69.10 (CH_2O), 43.20 (CH_2CH_2N), 34.38 (CH_2CH_2N), 27.18 (CH_3), 25.68 (CH_3).

EI-MS: *m/z* = 371 (1) [M^+], 356 (26), 296 (18), 256 (98), 169 (25), 129 (60), 102 (16), 43 (73).

(S)-4-(5-iodo-1H-indol-1-yl)butane-1,2-diol (6).

^{1}H NMR (400 MHz, DMSO) δ 7.91 (dd, *J* = 1.6, 0.6 Hz, 1H, Ar), 7.40 – 7.33 (m, 3H, Ar), 6.40 (dd, *J* = 3.1, 0.7 Hz, 1H, Ar), 4.73 (d, *J* = 4.7 Hz, 1H, CHO*H*), 4.55 – 4.27 (t, *J* = 5.5, 1H, CH_2OH), 4.25 (m, 2H, CH_2N), 3.33 – 3.18 (m, 3H, CH_2OH+ C*H*OH), 1.94 (m, 1H, CH*H*CH_2N), 1.64 (m, 1H, CH*H*CH_2N).

^{13}C NMR (101 MHz, DMSO) δ 134.61, 130.72, 129.65, 128.64, 128.62, 112.24, 99.66 (Ar), 82.50 (C-I), 68.33 (CHOH), 65.71 (CH_2OH), 42.40 (CH_2CH_2N), 33.94 (CH_2CH_2N).

EI-MS: *m/z* = 331 (1) [M^+], 256 (75), 243 (15), 129 (73), 102 (31), 32 (87).

(S)-1-(bis(4-methoxyphenyl)(phenyl)methoxy)-4-(5-iodo-1H-indol-1-yl) butan-2-ol (7).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.94 (d, *J* = 1.6 Hz, 1H, Ar), 7.42 – 7.35 (m, 3H, Ar), 7.29 – 7.26 (m, 2H, Ar), 7.24 (d, *J* = 1.8 Hz, 2H, Ar), 7.22 – 7.16 (m, 2H, Ar), 7.12 (d, *J* = 8.6 Hz, 1H, Ar), 7.04 (d, *J* = 3.1 Hz, 1H, Ar), 6.84 – 6.79 (m, 5H, Ar), 6.38 (dd, *J* = 3.1, 0.8 Hz, 1H, Ar), 4.29 – 4.18 (m, 2H, CH_2N), 3.80 (d, *J* = 6.6 Hz, 6H, OCH_3), 3.78 (m, 1H, C*H*OH), 3.08 (dd, *J* = 9.4, 3.4 Hz, 1H, C*H*HODMT), 2.99 (dd, *J* = 9.4, 7.3 Hz, 1H, CH*H*ODMT), 2.36 (d, *J* = 3.8 Hz, 1H, CHO*H*), 1.87 – 1.78 (m, 2H, C*H*$_2$CH$_2$N).

^{13}C NMR (101 MHz, $CDCl_3$) δ 158.54, 144.55, 139.45, 135.72, 135.01, 131.11, 129.96, 129.65, 129.12, 128.79, 128.04, 127.86, 113.16, 111.44, 100.48 (Ar), 86.22 (Ar_3C), 82.73 (C-I), 67.86 (CH_2ODMT), 67.25 (CHOH), 55.23 (2×O-CH_3), 42.58 (CH_2*C*H$_2$N), 33.60 (*C*H$_2$CH$_2$N).

ESI-MS (TOF): m/z calcd. for $C_{33}H_{32}NO_4I$ [M^+] 633.1376, found 633.1395.

(S)-1-(bis(4-methoxyphenyl)(phenyl)methoxy)-4-(5-(2-(pyren-1-yl)ethynyl)-1H-indol-1-yl)butan-2-ol (9).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.75 (d, *J* = 9.1 Hz, 1H, Ar), 8.23 – 8.14 (m, 4H, Ar), 8.12 (d, *J* = 8.0 Hz, 1H, Ar), 8.09 – 8.00 (m, 4H, Ar), 7.56 (dd, *J* = 8.5, 1.5 Hz, 1H, Ar), 7.40 – 7.37 (m, 3H, Ar), 7.29 – 7.21 (m, 7H, Ar), 7.15 (d, *J* = 3.1 Hz, 1H, Ar), 6.83 – 6.80 (m, 4H, Ar), 6.53 (d, *J* = 3.1 Hz, 1H, Ar), 4.30 (q, *J* = 6.3, 5.9 Hz, 2H, CH_2N), 3.78 (s, 6H, OCH_3), 3.69 (m, 1H, C*H*OH), 3.11 (dd, *J* = 9.5, 3.4 Hz, 1H, C*H*HODMT), 3.03 (dd, *J* = 9.4, 7.3 Hz, 1H, CH*H*ODMT), 2.55 (m, *J* = 114.9 Hz, 1H, CHO*H*) 1.91 (m, 2H, C*H*$_2$CH$_2$N).

^{13}C NMR (101 MHz, $CDCl_3$) δ 158.56, 144.61, 135.77, 131.72, 131.35, 131.21, 130.81, 130.00, 129.45, 129.12, 128.60, 128.08, 127.90, 127.80, 127.32, 126.93, 126.15, 125.87, 125.42, 125.35, 125.21, 124.98, 124.60, 124.57, 124.47, 114.04, 113.19, 109.69, 101.64, 97.18 (Ar), 90.03 (C≡C), 89.49 (C≡C), 86.26 (Ar_3C), 67.33 (CH_2ODMT), 66.66 (CHOH), 55.24 (2×O-CH_3), 42.70 (CH_2*C*H$_2$N), 33.72 (*C*H$_2$CH$_2$N).

ESI-MS (TOF): m/z calcd. for $C_{51}H_{41}NO_3\,Na^+$ [MNa^+] 754.2928, found 754.2937.

(S)-1-(bis(4-methoxyphenyl)(phenyl)methoxy)-4-(5-(2-(pyren-1-yl)ethynyl)-1H-indol-1-yl)butan-2-yl) 2cyanoethyldiisopropylphosphoramidite (10).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.75 (dd, *J* = 9.1, 1.1 Hz, 1H , Ar), 8.21 (dd, *J* = 11.9, 8.7 Hz, 4H , Ar), 8.14 (d, *J* = 8.0 Hz, 1H , Ar), 8.10 – 8.02 (m, 4H , Ar), 7.52 (dt, *J* = 8.5, 1.7 Hz, 1H , Ar), 7.44 (dt, *J* = 8.1, 1.6 Hz, 2H , Ar), 7.30 (m, 6H , Ar), 7.26 – 7.18 (m, 2H , Ar), 7.14 (dd, *J* = 9.2, 3.1 Hz, 1H , Ar), 6.82 (ddd, *J* = 8.7, 6.8, 1.7 Hz, 4H , Ar), 6.52 (dd, *J* = 7.1, 3.1 Hz, 1H , Ar), 4.21 (m, 2H, CH_2N), 4.09 – 3.98 (m, 1H, (OC*H*$_2$$CH_2CN$), 3.89 – 3.82 (m, 1H, (OC*H*$_2$$CH_2CN$), 3.79 (dd, *J* = 2.7, 1.2 Hz, 6H, OCH_3), 3.76 – 3.70 (m, 1H, C*H*OP), 3.64 (m, 2H, C*H*$_2$ODMT), 3.24 (m, 1H, O CH_2C*H*$_2$CN), 3.09 (m, 1H, OCH_2C*H*$_2$CN), 2.60 (t, *J* = 6.4 Hz, 1H, (NC*H*$(CH_3)_2$), 2.39 (t, *J* = 6.5 Hz, 1H, (NC*H*$(CH_3)_2$), 2.35 – 2.24 (m, 1H, C*H*$_2$$CH_2N$), 2.23 – 2.12 (m, 1H, C*H*$_2$$CH_2N$), 1.22 – 1.18 (m, 9H, 3XCH_3), 1.14 (d, *J* = 6.8 Hz, 3H, CH_3).

^{13}C NMR (101 MHz, $CDCl_3$) δ 158.67, 158.63, 144.95, 144.93, 136.25, 136.13, 135.80, 131.86, 131.50, 131.37, 130.96, 130.25, 130.23, 130.17, 129.59, 129.18, 129.02, 128.85, 128.37, 128.32, 128.21, 127.96, 127.47, 126.98, 126.91, 126.30, 126.02, 125.56, 125.50, 125.35, 125.27, 125.16, 125.12, 124.76, 124.71, 124.62, 119.01, 118.98, 117.84, 117.66, 114.21, 114.12, 113.26, 109.97, 109.90, 101.72, 101.61, 97.41, 97.35 (Ar), 86.46 (C≡C), 86.42 (C≡C), 86.25 ($OCPh_3$), 71.56 (CHOP), 71.50 (CHOP), 65.73 (CH_2ODMT), 65.56 (CH_2ODMT), 58.17(O*C*H_2CH_2CN), 58.14 (O*C*H_2CH_2CN), 55.39 (2xOCH_3), 43.48 (N*C*H$(CH_3)_2$), 43.41 (N*C*H$(CH_3)_2$), 43.36 (N*C*H$(CH_3)_2$), 43.28 (N*C*H$(CH_3)_2$), 42.94 (CH_2*C*H_2N), 42.84 (CH_2*C*H_2N), 34.15 (*C*H_2CH_2N),, 33.87 (*C*H_2CH_2N),, 24.96 (CH_3), 24.92 (CH_3), 24.88 (CH_3), 24.85 (CH_3), 24.79 (CH_3), 24.72 (CH_3), 23.71 (CH_3), 23.10 (CH_3), 19.21 (*C*H_2CN), 19.17 (*C*H_2CN).

^{31}P NMR (CDCl3, 162 MHz): δ = 148.97, 148.44 in a 5:6 ratio.

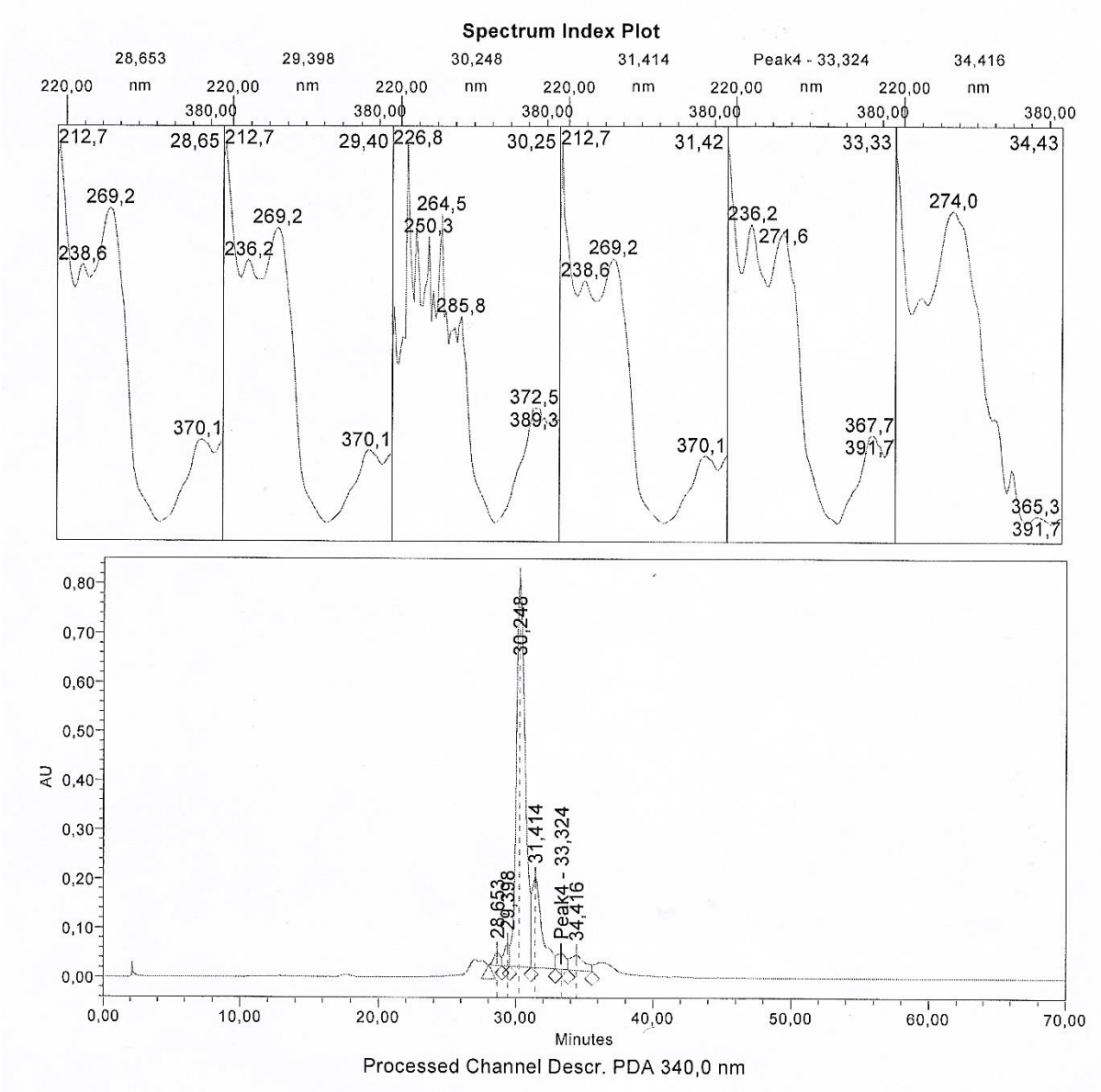
ESI-MS (TOF): m/z calcd. for $C_{60}H_{58}N_3O_5P$ Na^+ [MNa^+] 954.4012, found 954.4048.

HPLC chromatograms of oligonucleotides:

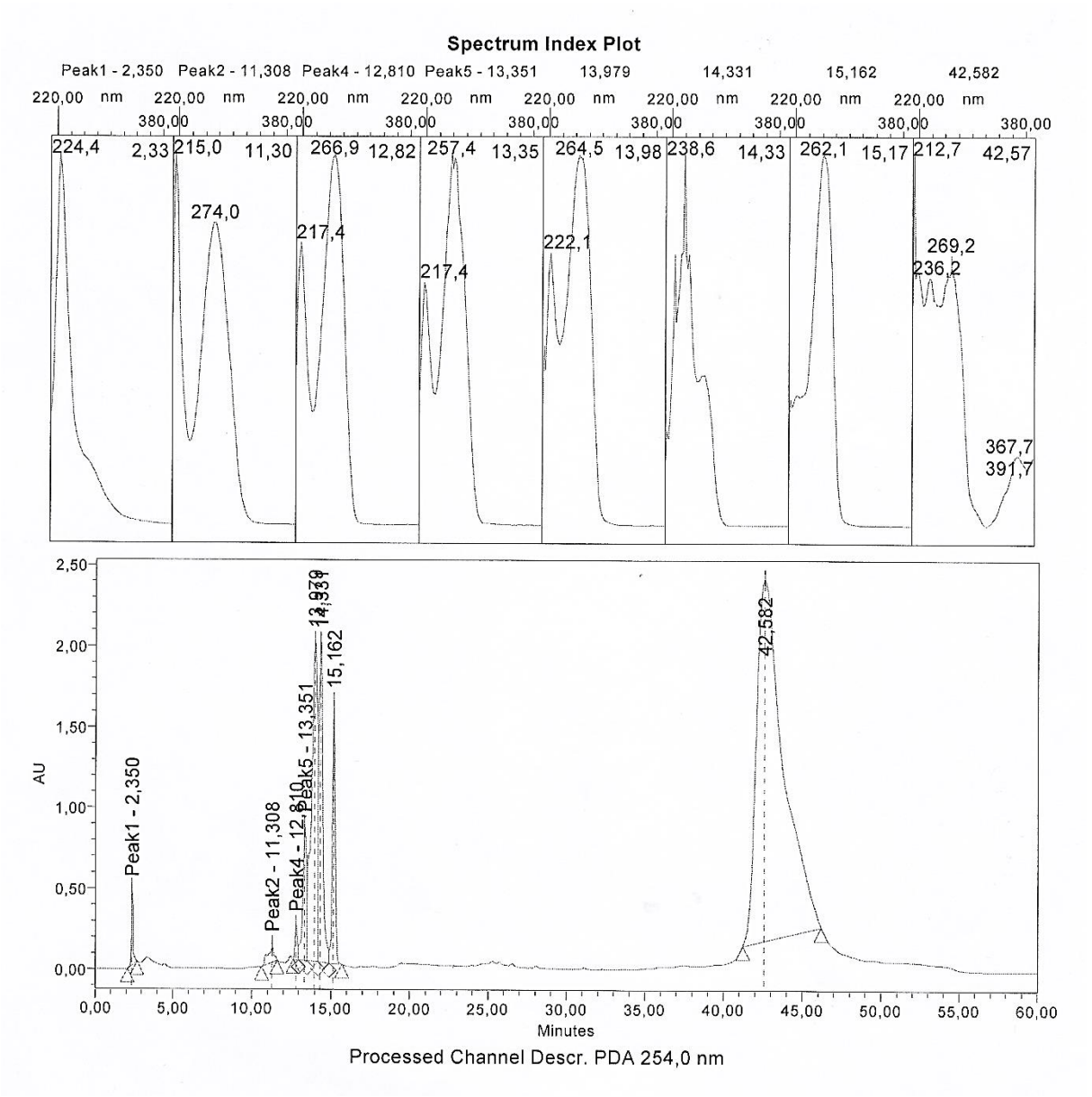
DMT-on oligonucleotides (ONs) were purified using a reverse-phase semipreparative HPLC (Waters Xterra MS C_{18} column) with a Waters Delta Prep 4000 Preparative Chromatography System and using following buffers: Buffer A [0.05m triethyl ammonium acetate in H_2O (pH 7.0)] and Buffer B (75% CH_3CN in H_2O). Gradients: 2 min 100% A, linear gradient to 70% B in 40 min, linear gradient to 100% B in 7 min, 100% B in 3 min and then 100% A in 10 min). Flow 2.5 $mLmin^{-1}$.

ON2 5'-CCCCTT**Y**TCTTTTTT, retention time 30,25 min

ON3 5'-CCCCTT**Y**$^{l\text{-}ind}$TCTTTTTT, retention time 31,41 min



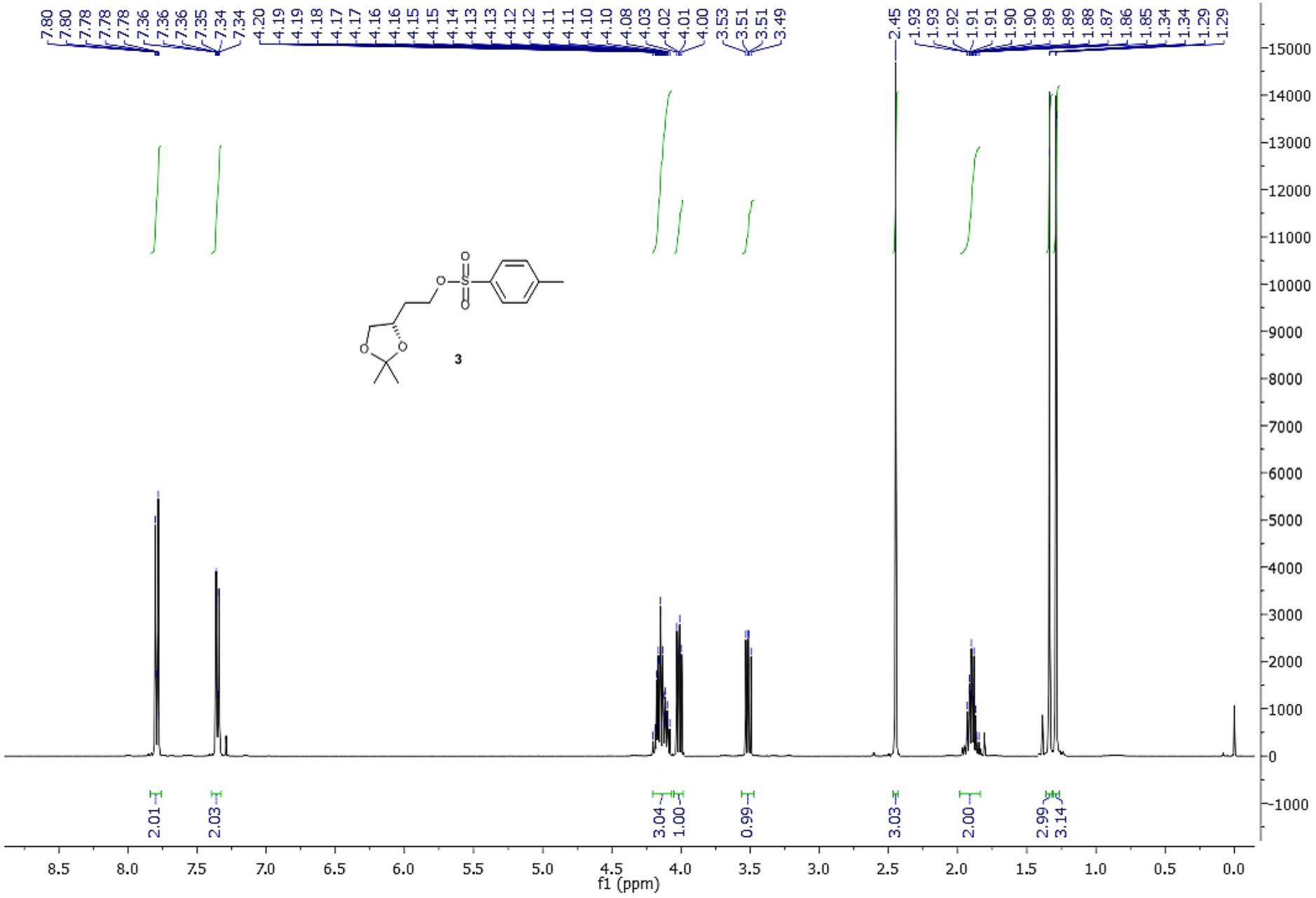
ON4 5’-**Y**CCCCTTTCTTTTTT, retention time 42,58 min



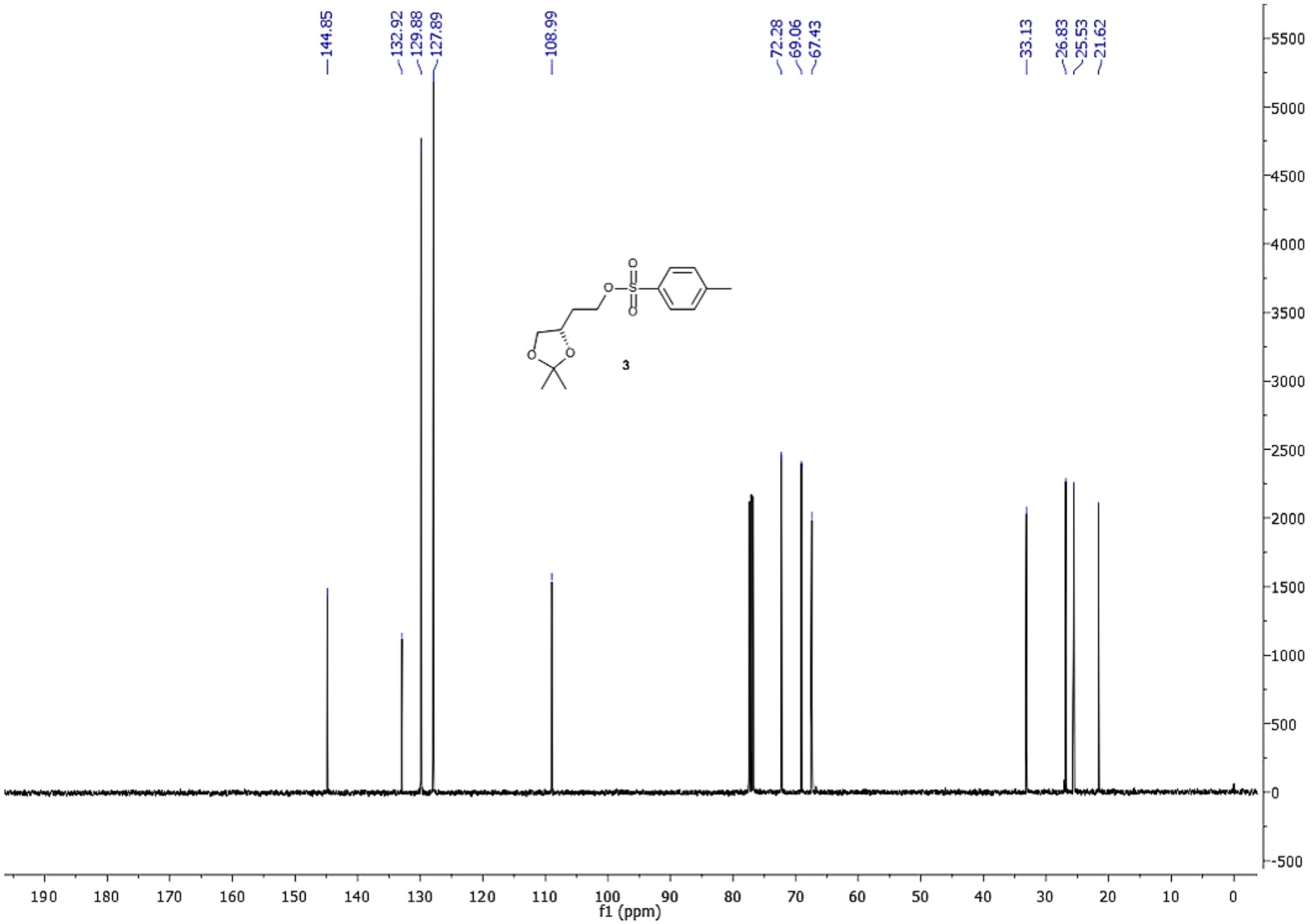
References:

1. L. Börjesson and C. J. Welch, Tetrahedron 1992, 48, 6325.

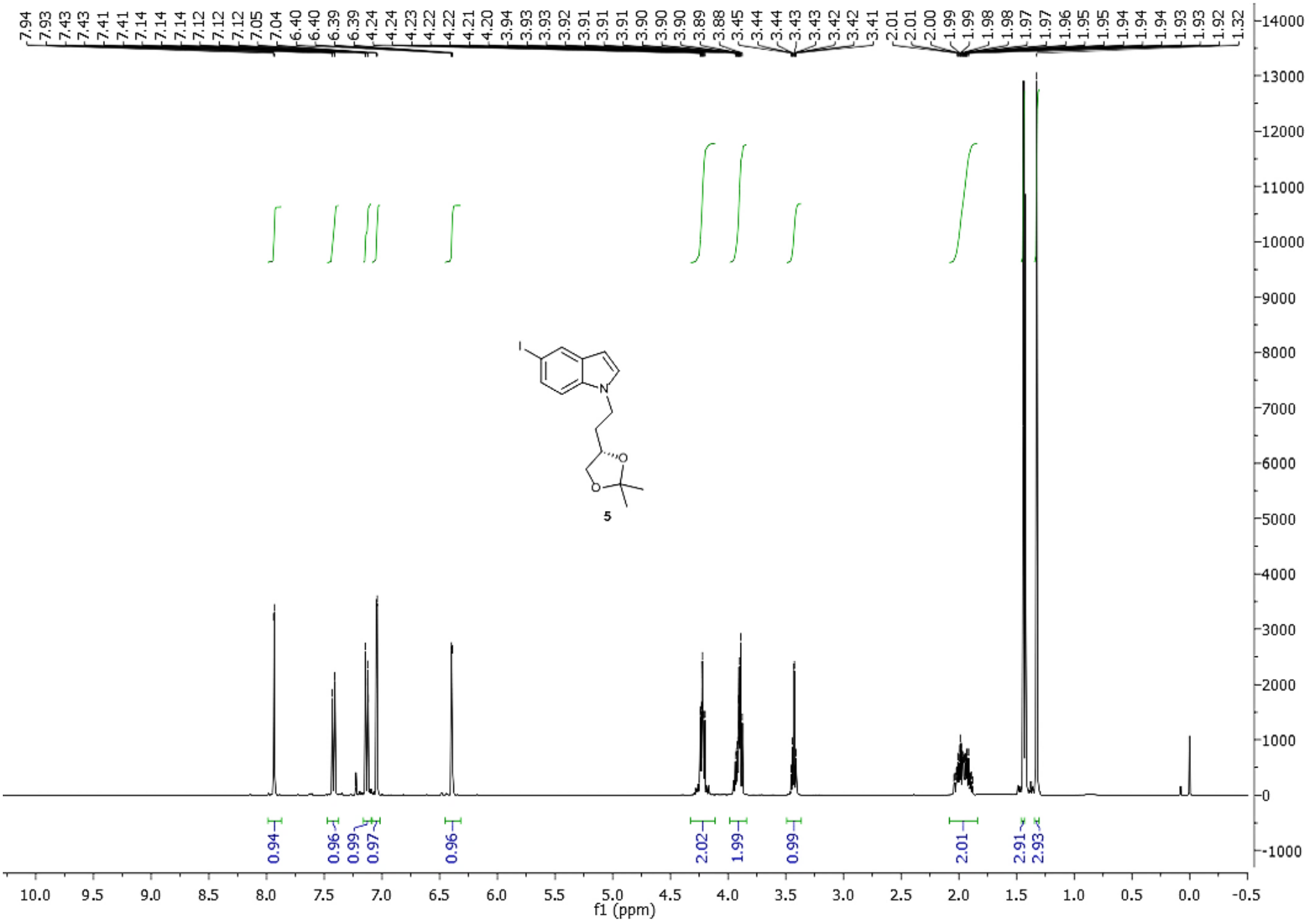
^{1}H-NMR:


7.80 7.80 7.78 7.78 7.78 7.36 7.36 7.36 7.35 7.34 7.34
4.20 4.19 4.19 4.18 4.17 4.17 4.16 4.16 4.15 4.15 4.14 4.13 4.13 4.12 4.12 4.11 4.11 4.10 4.10 4.08 4.03 4.02 4.01 4.00
3.53 3.51 3.51 3.49
2.45
1.93 1.93 1.92 1.91 1.91 1.90 1.90 1.89 1.89 1.88 1.87 1.86 1.85 1.34 1.34 1.29 1.29
3
2.01 2.03 3.04 1.00 0.99 3.03 2.00 2.99 3.14
f1 (ppm)

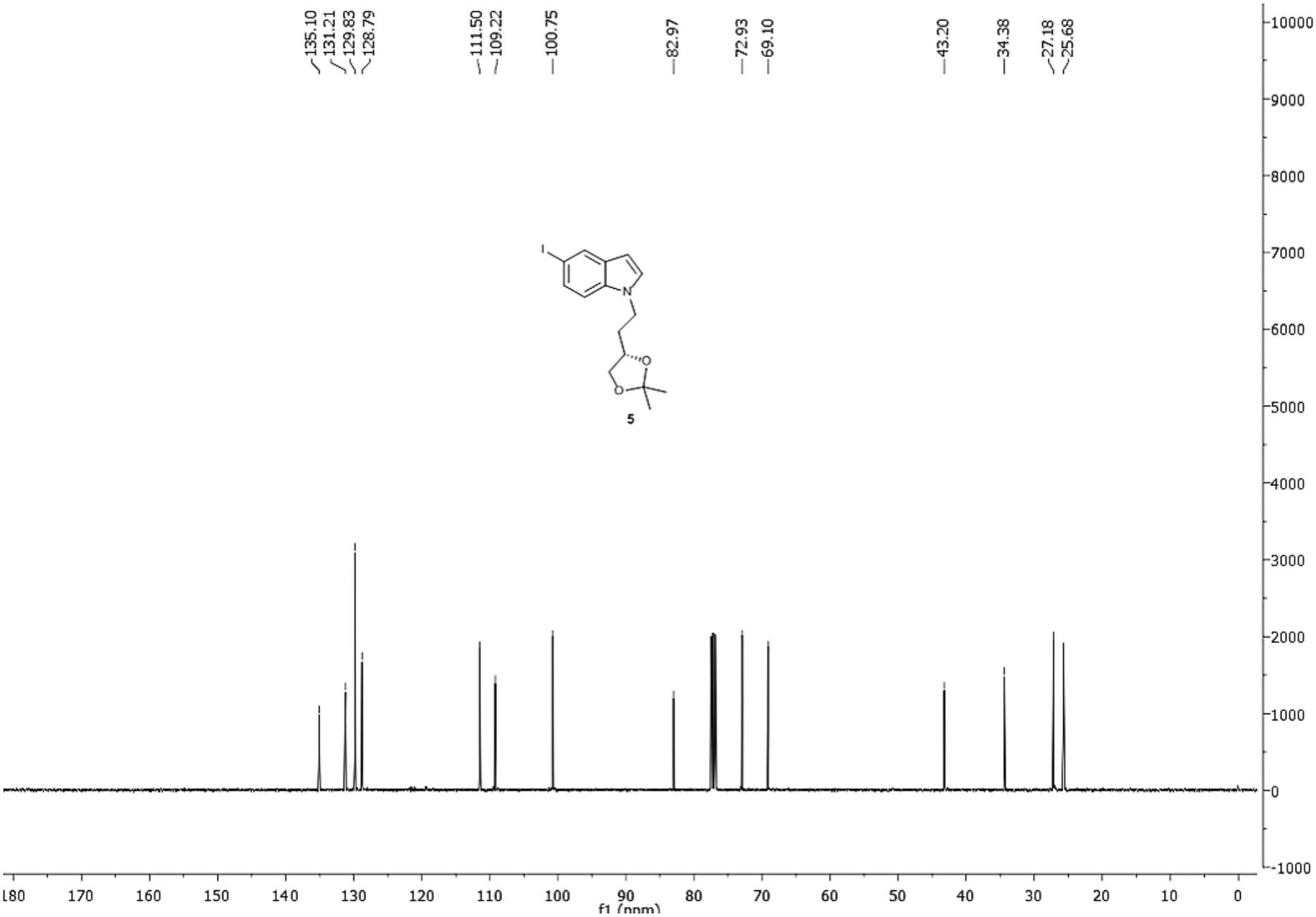
^{13}C-NMR:


144.85
132.92
129.88
127.89
108.99
72.28
69.06
67.43
33.13
26.83
25.53
21.62
3
f1 (ppm)

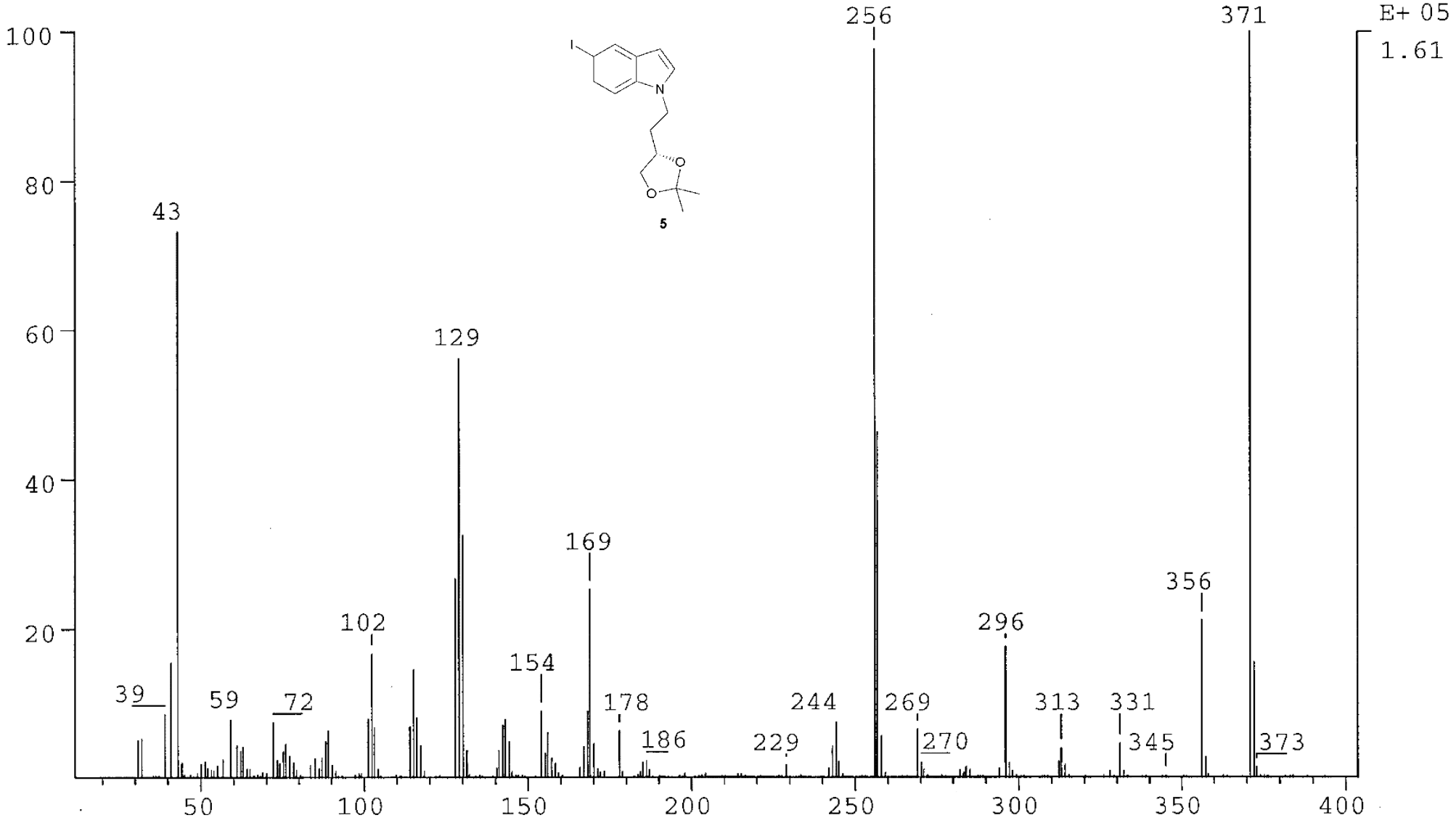
^{1}H-NMR:


7.94 7.93 7.43 7.43 7.41 7.41 7.14 7.14 7.14 7.12 7.12 7.12 7.05 7.04 6.40 6.40 6.39 6.39 4.24 4.24 4.23 4.22 4.22 4.21 4.20 3.94 3.93 3.93 3.92 3.91 3.91 3.91 3.90 3.90 3.90 3.89 3.88 3.45 3.44 3.44 3.43 3.43 3.42 3.42 3.41 2.01 2.01 2.00 1.99 1.99 1.98 1.98 1.97 1.97 1.96 1.95 1.95 1.94 1.94 1.94 1.93 1.93 1.92 1.32
I
N
O
O
5
0.94 0.96 0.99 0.97 0.96 2.02 1.99 0.99 2.01 2.91 2.93
f1 (ppm)

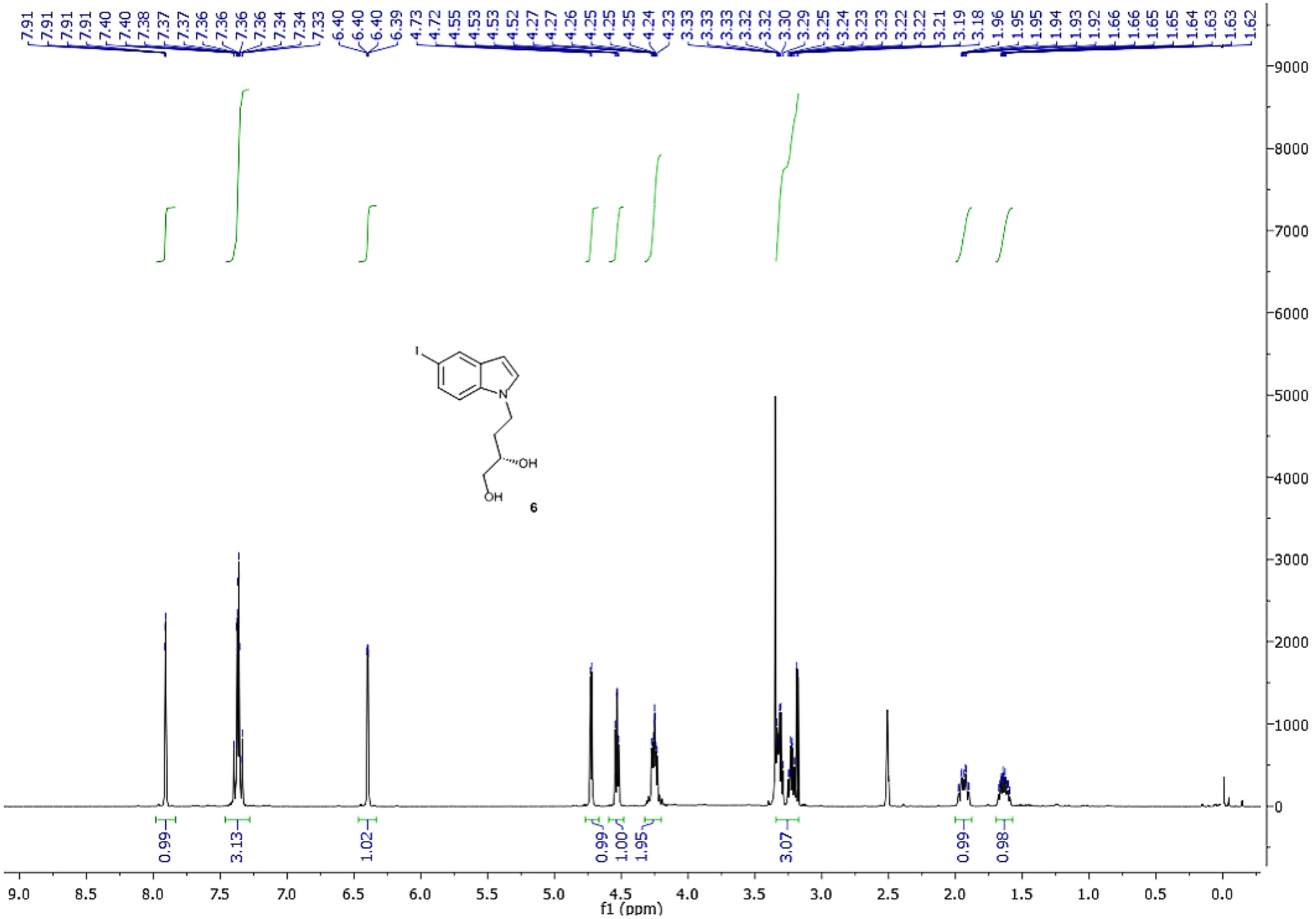
^{13}C-NMR:


135.10
131.21
129.83
128.79
111.50
109.22
100.75
82.97
72.93
69.10
43.20
34.38
27.18
25.68
I
N
O
O
5
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
-1000
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

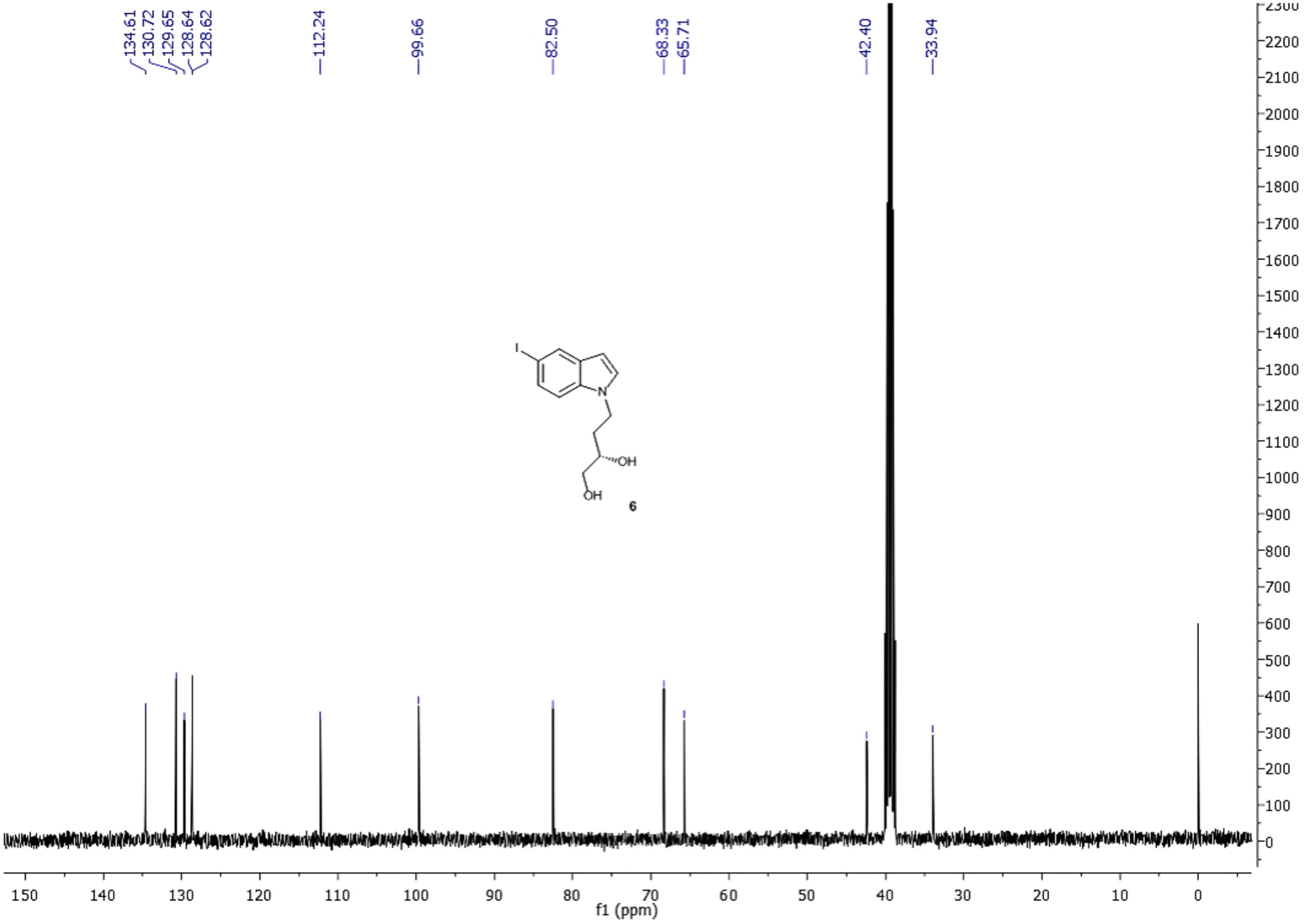
EI-MS:


E+ 05
1.61
100
80
60
40
20
43
39
59
72
102
129
154
169
178
186
229
244
256
269
270
296
313
331
345
356
371
373
50
100
150
200
250
300
350
400
5

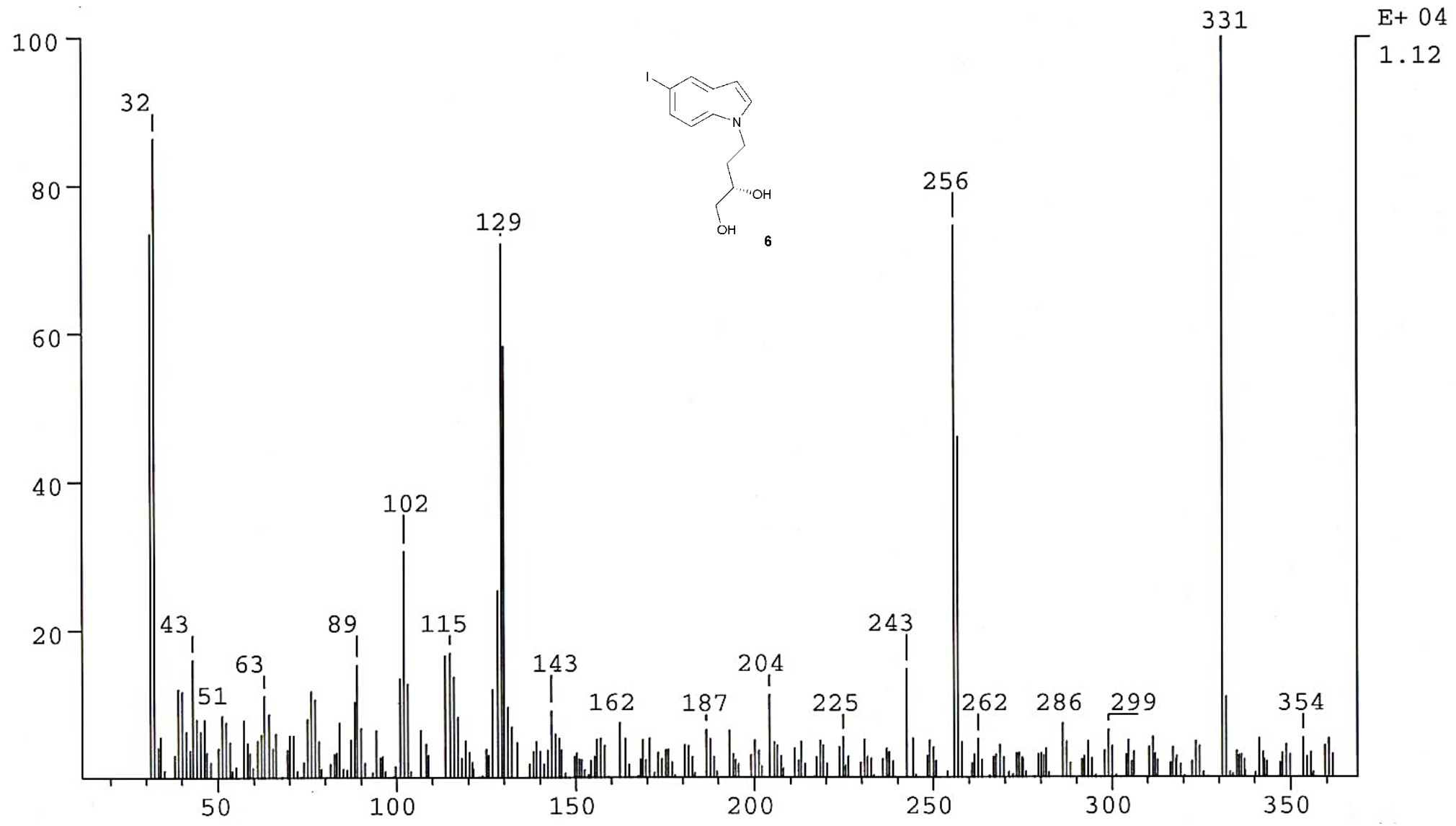
^{1}H-NMR:


7.91 7.91 7.91 7.91 7.40 7.40 7.38 7.37 7.37 7.36 7.36 7.36 7.36 7.34 7.34 7.33
6.40 6.40 6.40 6.39
4.73 4.72 4.55 4.53 4.53 4.52 4.27 4.27 4.26 4.25 4.25 4.25 4.24 4.23
3.33 3.33 3.33 3.32 3.32 3.30 3.29 3.25 3.24 3.23 3.23 3.22 3.22 3.21
3.19 3.18 1.96 1.95 1.95 1.94 1.93 1.92 1.66 1.66 1.65 1.65 1.64 1.63 1.63 1.62
I
N
OH
OH
6
0.99
3.13
1.02
0.99
1.00
1.95
3.07
0.99
0.98
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
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 0.0
f1 (ppm)

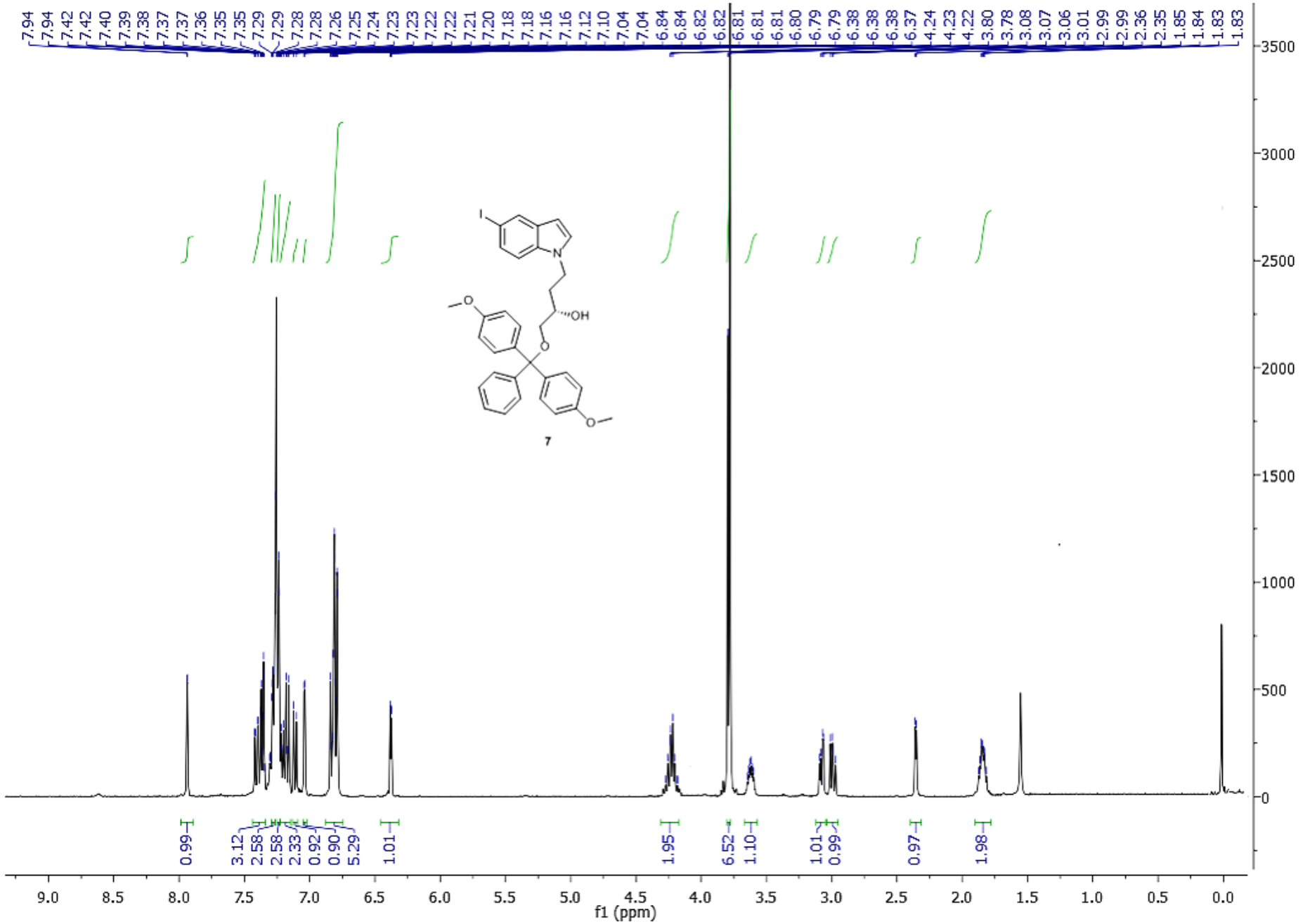
^{13}C-NMR:


134.61
130.72
129.65
128.64
128.62
112.24
99.66
82.50
68.33
65.71
42.40
33.94
I
N
OH
OH
6
f1 (ppm)

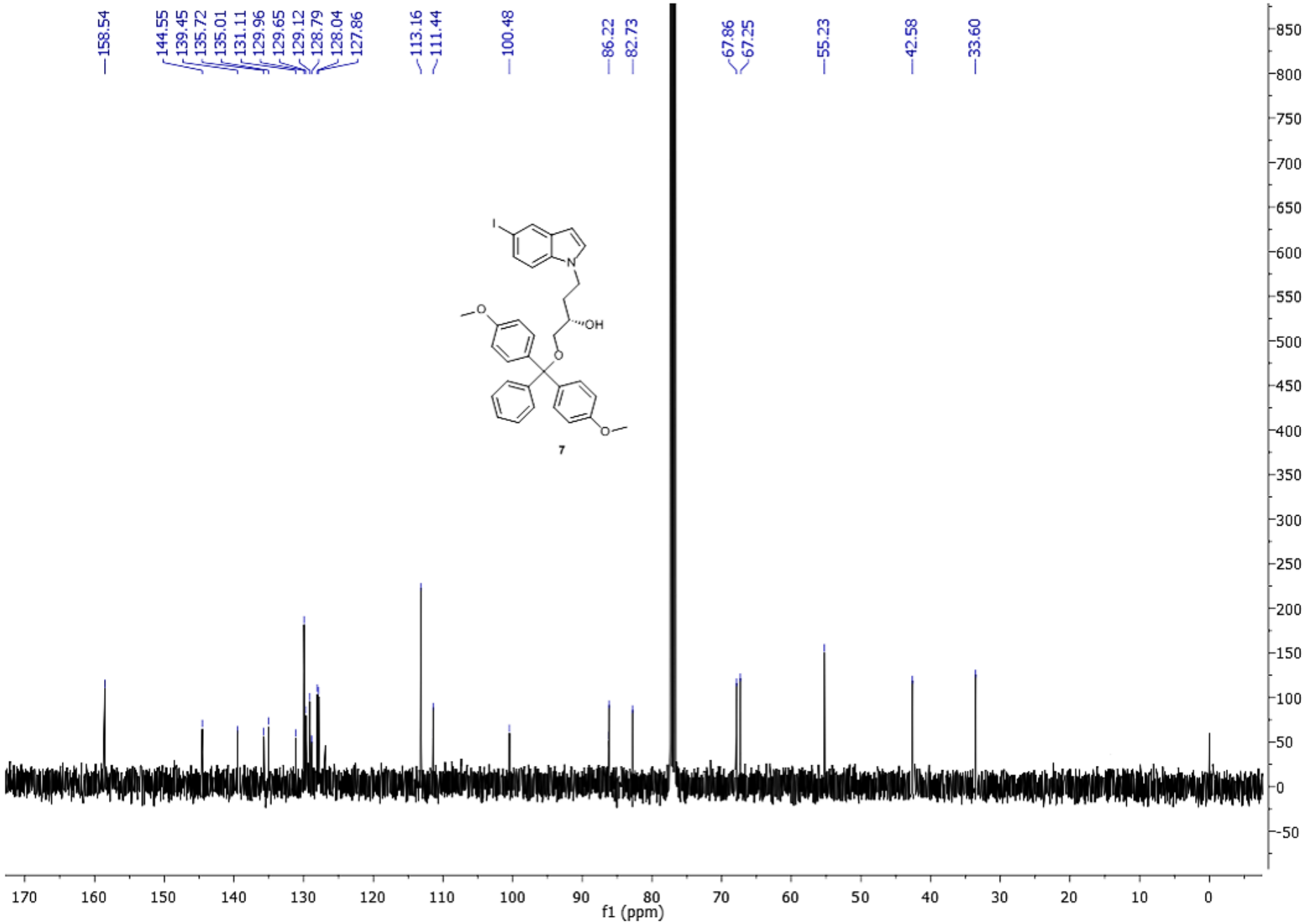
EI-MS:


E+ 04
1.12
100
80
60
40
20
32
43
51
63
89
102
115
129
143
162
187
204
225
243
256
262
286
299
331
354
50
100
150
200
250
300
350
I
N
OH
OH
6

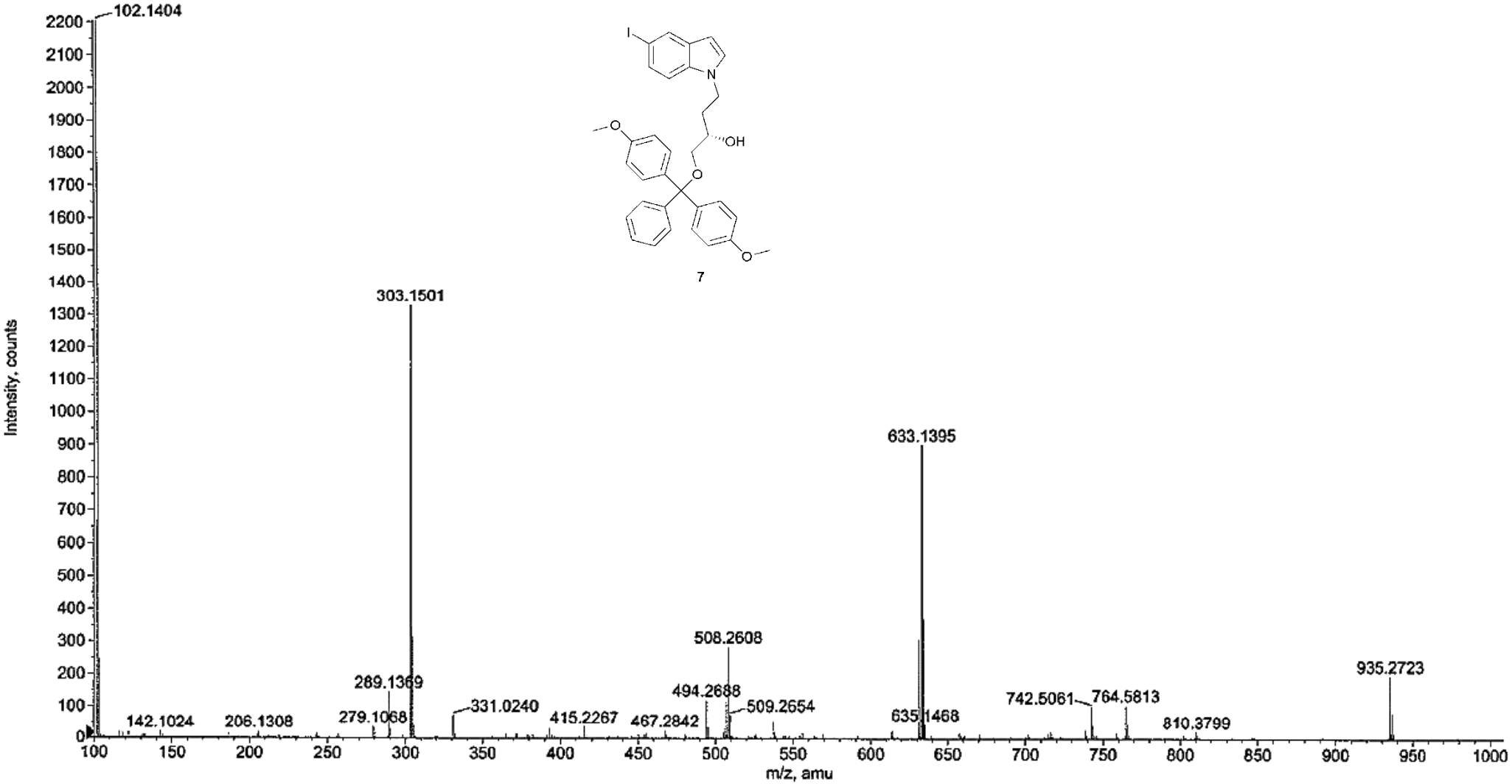
^{1}H-NMR:


7.94 7.94 7.42 7.42 7.40 7.39 7.38 7.37 7.37 7.36 7.35 7.35 7.29 7.29 7.28 7.28 7.26 7.25 7.24 7.23 7.23 7.22 7.22 7.21 7.20 7.18 7.18 7.16 7.16 7.12 7.10 7.04 7.04 6.84 6.84 6.82 6.82 6.81 6.81 6.81 6.80 6.79 6.79 6.38 6.38 6.38 6.37 4.24 4.23 4.22 3.80 3.78 3.08 3.07 3.06 3.01 2.99 2.99 2.36 2.35 1.85 1.84 1.83 1.83
OH
7
0.99 3.12 2.58 2.58 2.33 0.92 0.90 5.29 1.01 1.95 6.52 1.10 1.01 0.99 0.97 1.98
f1 (ppm)

^{13}C-NMR:

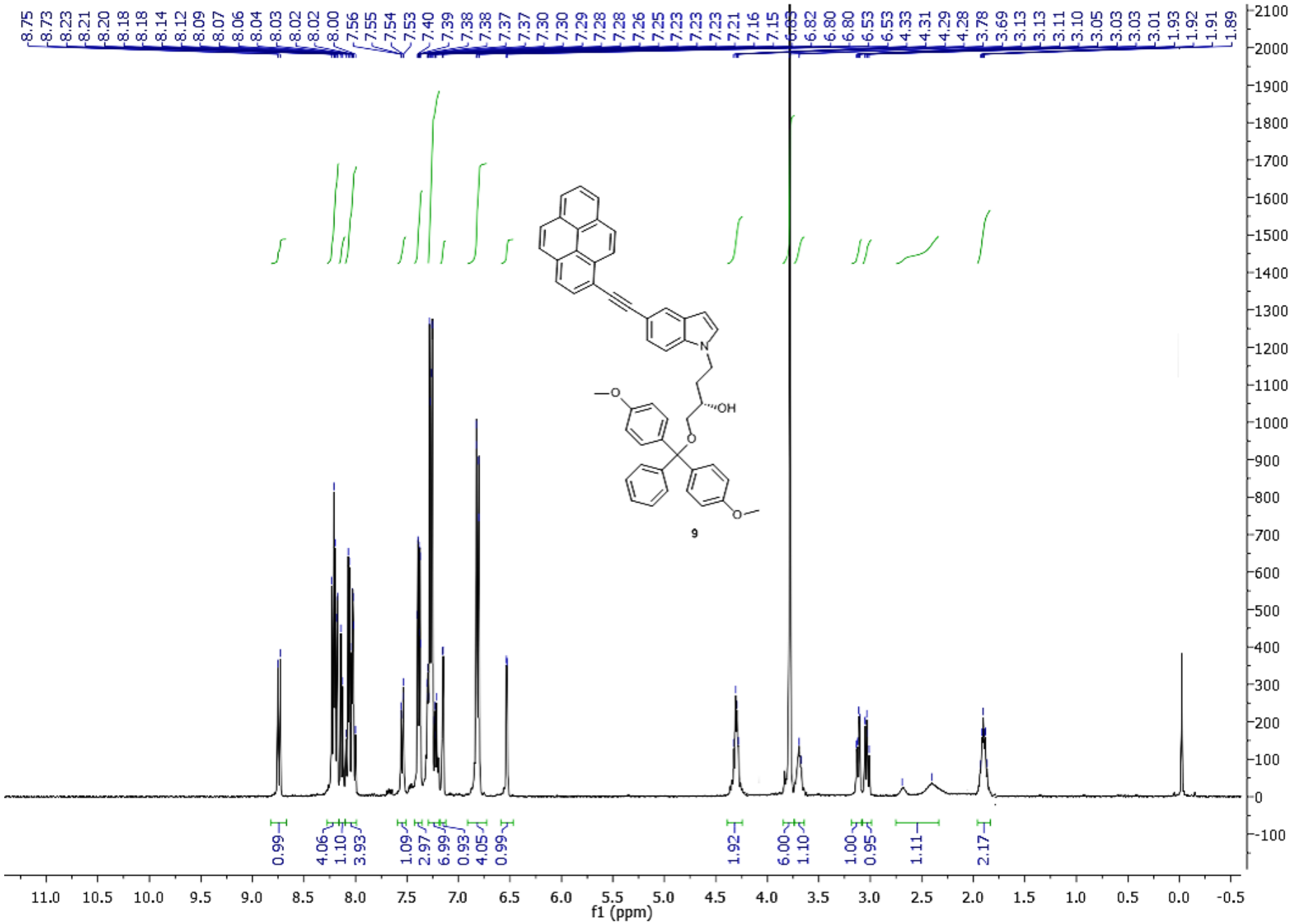

158.54
144.55
139.45
135.72
135.01
131.11
129.96
129.65
129.12
128.79
128.04
127.86
113.16
111.44
100.48
86.22
82.73
67.86
67.25
55.23
42.58
33.60
I
N
OH
O
O
O
7
f1 (ppm)

ESI-MS (TOF):

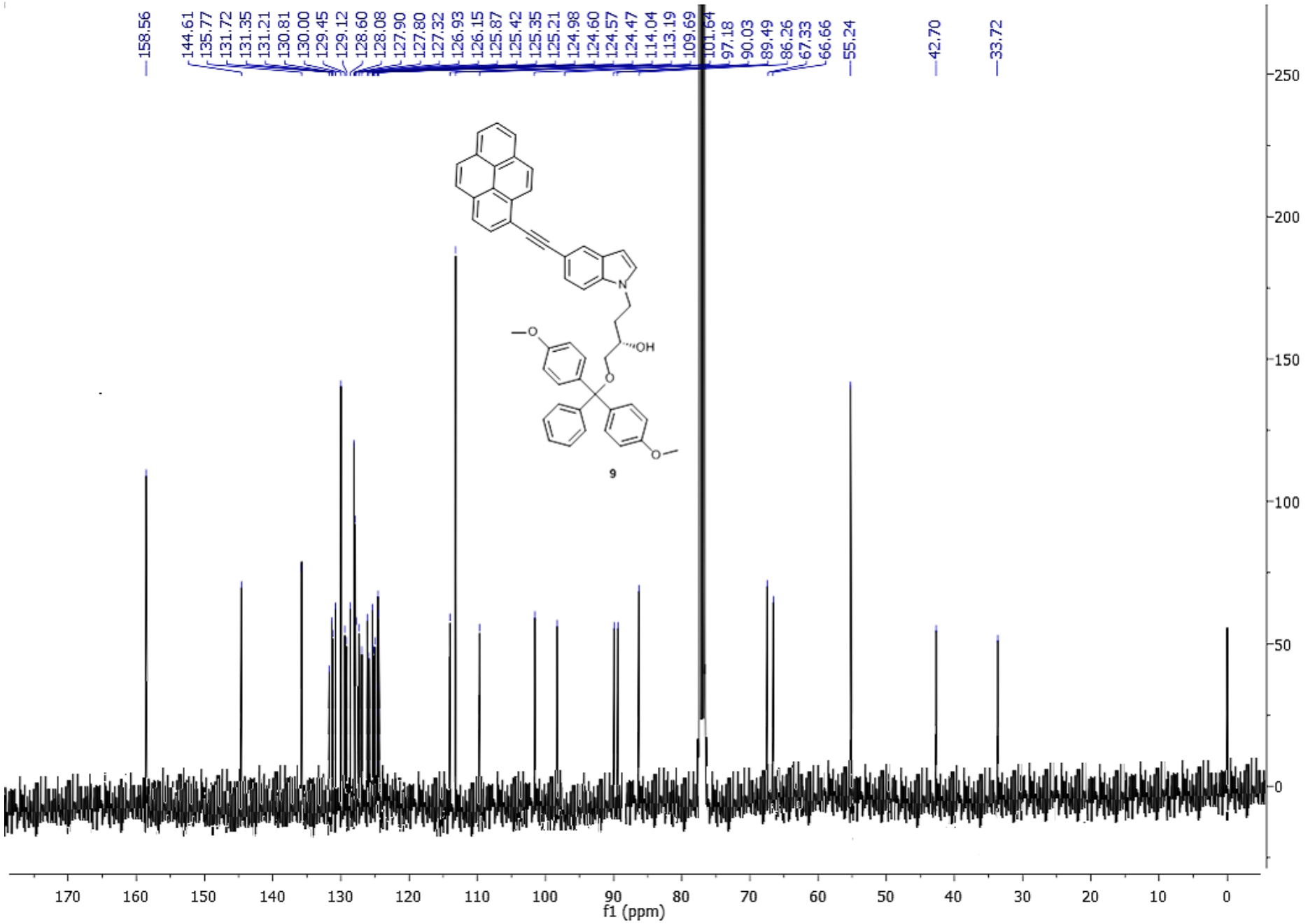

102.1404
303.1501
633.1395
508.2608
935.2723
289.1369
494.2688
509.2654
742.5061
764.5813
331.0240
142.1024
206.1308
279.1068
415.2267
467.2842
635.1468
810.3799
Intensity, counts
m/z, amu
I
N
OH
O
O
O

7

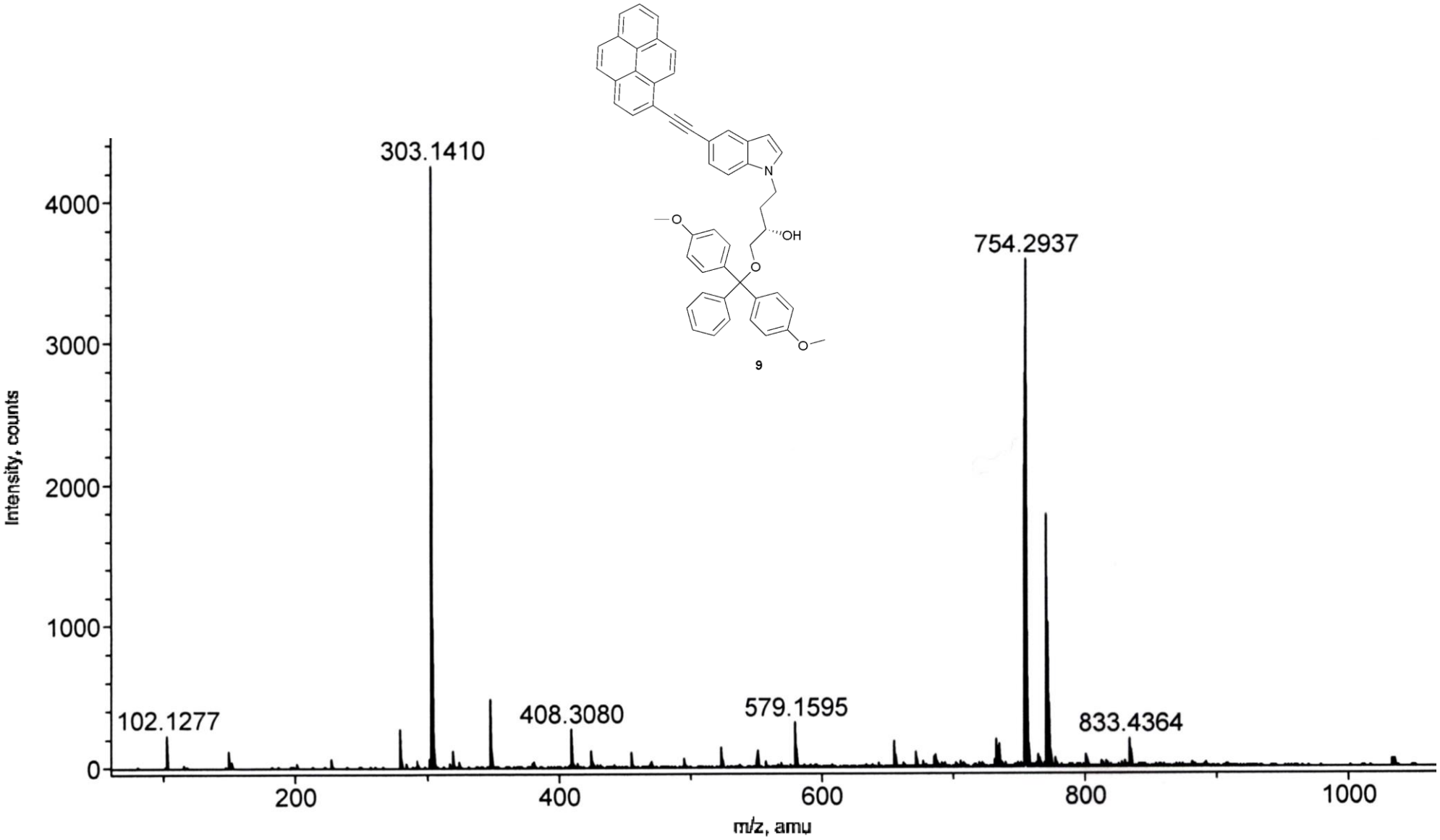
^{1}H-NMR:


8.75 8.73 8.23 8.21 8.20 8.18 8.18 8.14 8.12 8.09 8.07 8.06 8.04 8.03 8.02 8.02 8.00 7.56 7.55 7.54 7.53 7.40 7.39 7.38 7.38 7.37 7.37 7.30 7.30 7.29 7.28 7.28 7.26 7.25 7.23 7.23 7.23 7.21 7.16 7.15 6.83 6.82 6.80 6.80 6.53 6.53 4.33 4.31 4.29 4.28 3.78 3.69 3.13 3.13 3.11 3.10 3.05 3.03 3.03 3.01 1.93 1.92 1.91 1.89
OH
O
9
0.99 4.06 1.10 3.93 1.09 2.97 6.99 0.93 4.05 0.99 1.92 6.00 1.10 1.00 0.95 1.11 2.17
f1 (ppm)

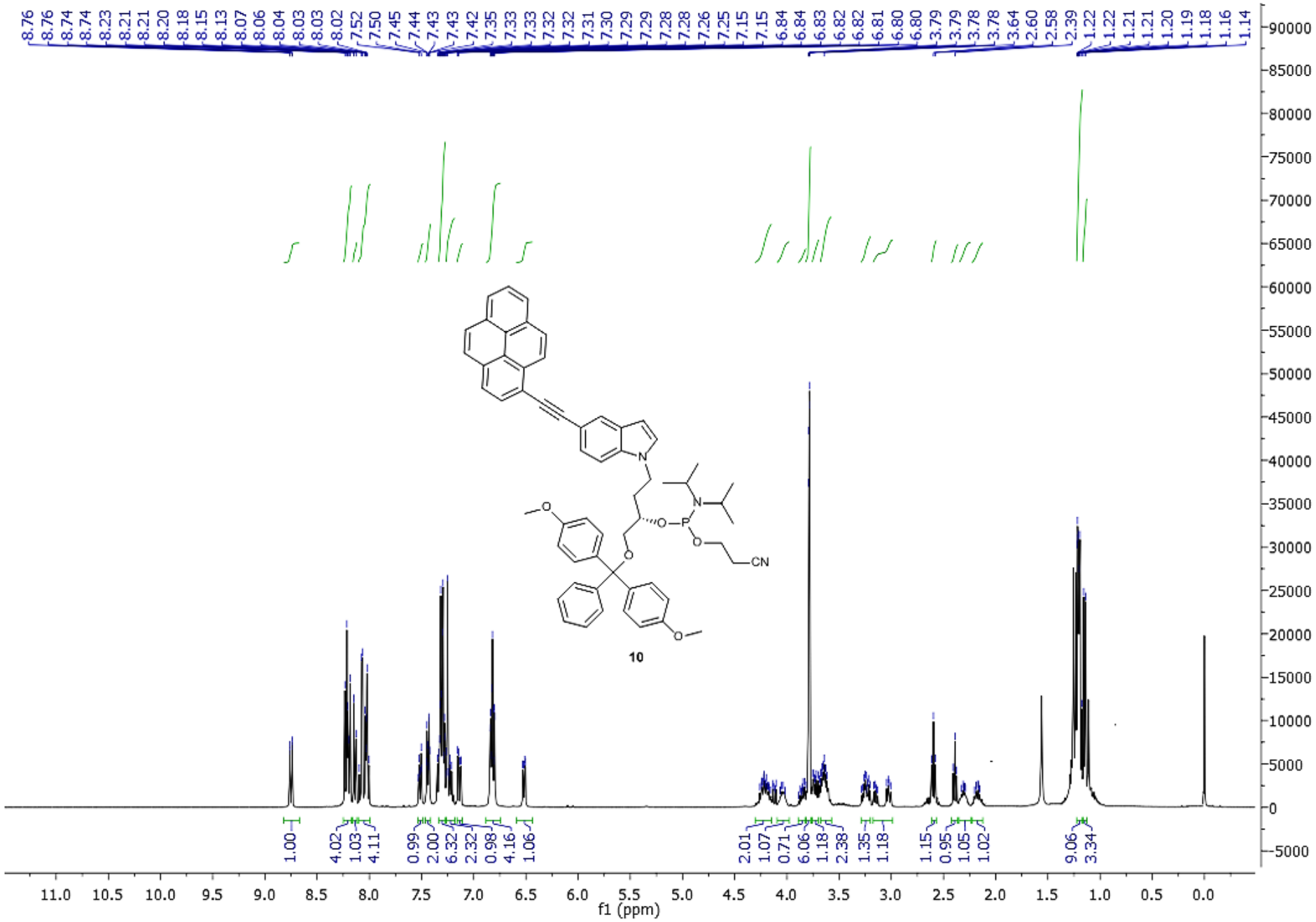
^{13}C-NMR:


158.56
144.61
135.77
131.72
131.35
131.21
130.81
130.00
129.45
129.12
128.60
128.08
127.90
127.80
127.32
126.93
126.15
125.87
125.42
125.35
125.21
124.98
124.60
124.57
124.47
114.04
113.19
109.69
101.64
97.18
90.03
89.49
86.26
67.33
66.66
55.24
42.70
33.72
OH
9
250
200
150
100
50
0
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

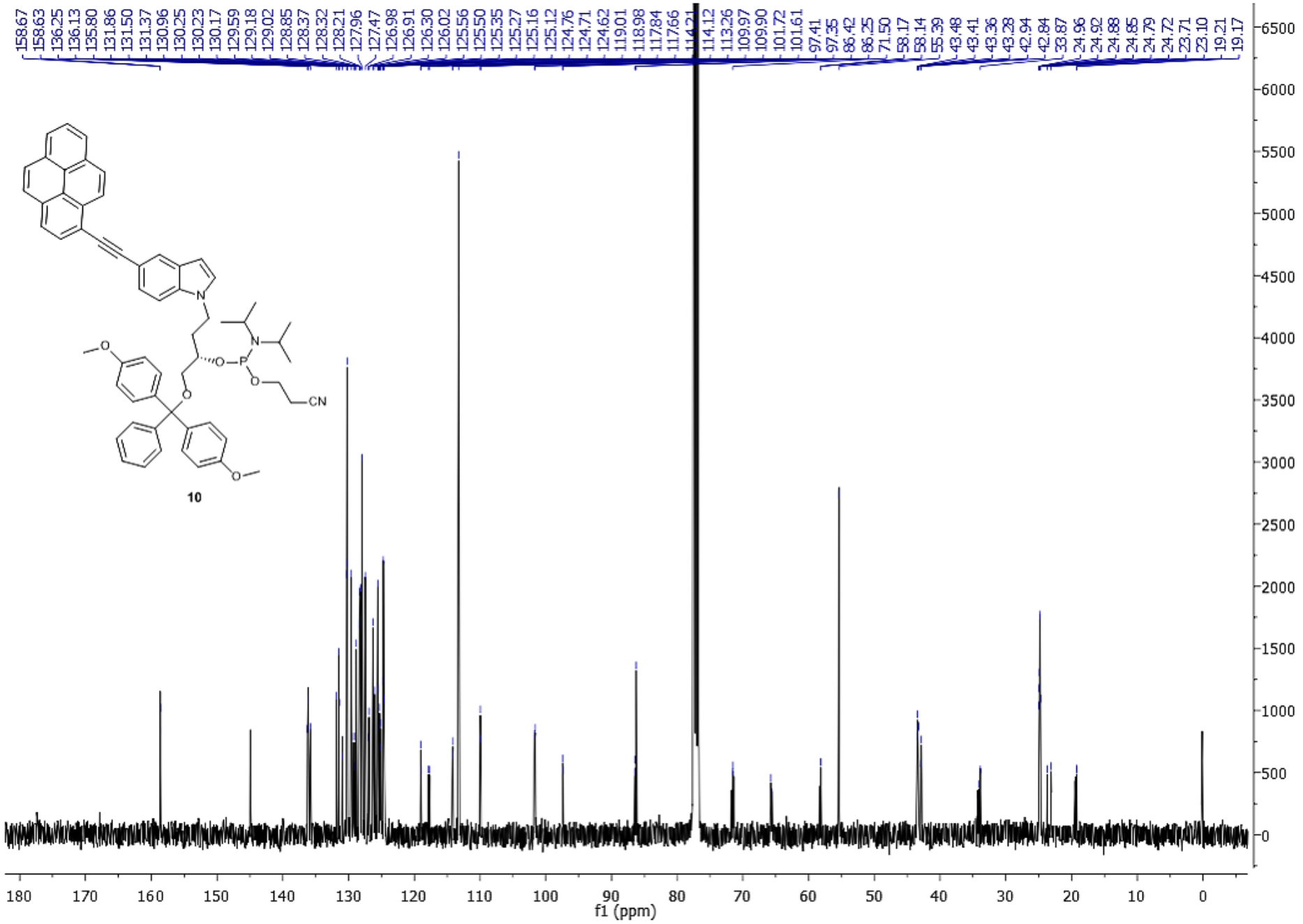
ESI-MS (TOF):


303.1410
754.2937
102.1277
408.3080
579.1595
833.4364
Intensity, counts
m/z, amu
9

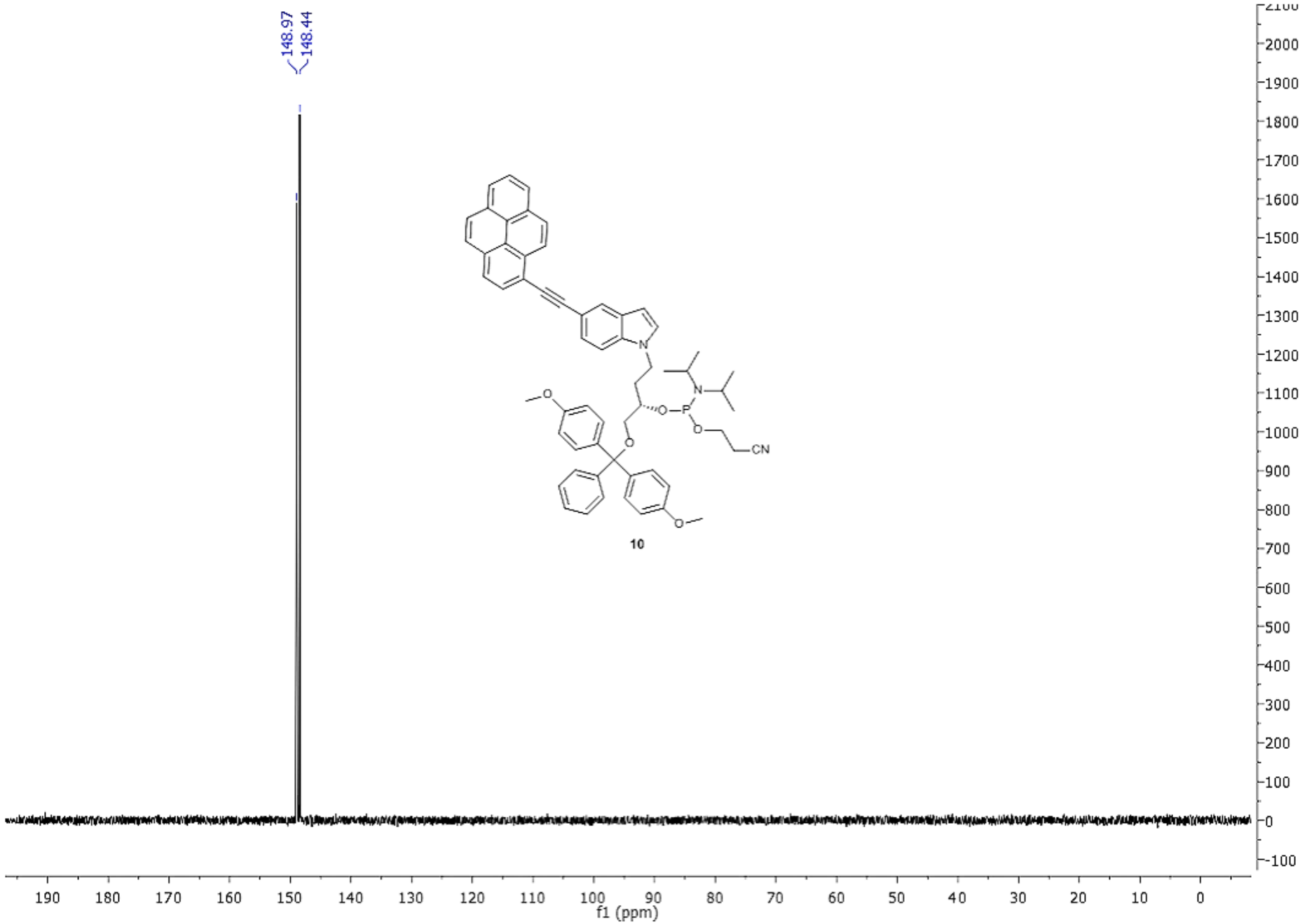
^{1}H-NMR:


8.76 8.76 8.74 8.74 8.23 8.21 8.21 8.20 8.18 8.15 8.13 8.07 8.06 8.04 8.03 8.03 8.02 7.52 7.50 7.45 7.44 7.43 7.43 7.42 7.35 7.33 7.33 7.32 7.32 7.31 7.30 7.29 7.29 7.28 7.28 7.26 7.25 7.15 7.15 6.84 6.84 6.83 6.82 6.82 6.81 6.80 6.80 3.79 3.79 3.78 3.78 3.64 2.60 2.58 2.39 1.22 1.22 1.21 1.21 1.20 1.19 1.18 1.16 1.14
90000 85000 80000 75000 70000 65000 60000 55000 50000 45000 40000 35000 30000 25000 20000 15000 10000 5000 0 -5000
1.00 4.02 1.03 4.11 0.99 2.00 6.32 2.32 0.98 4.16 1.06 2.01 1.07 0.71 6.06 1.18 2.38 1.35 1.18 1.15 0.95 1.05 1.02 9.06 3.34
11.0 10.5 10.0 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 0.0
f1 (ppm)
CN
10

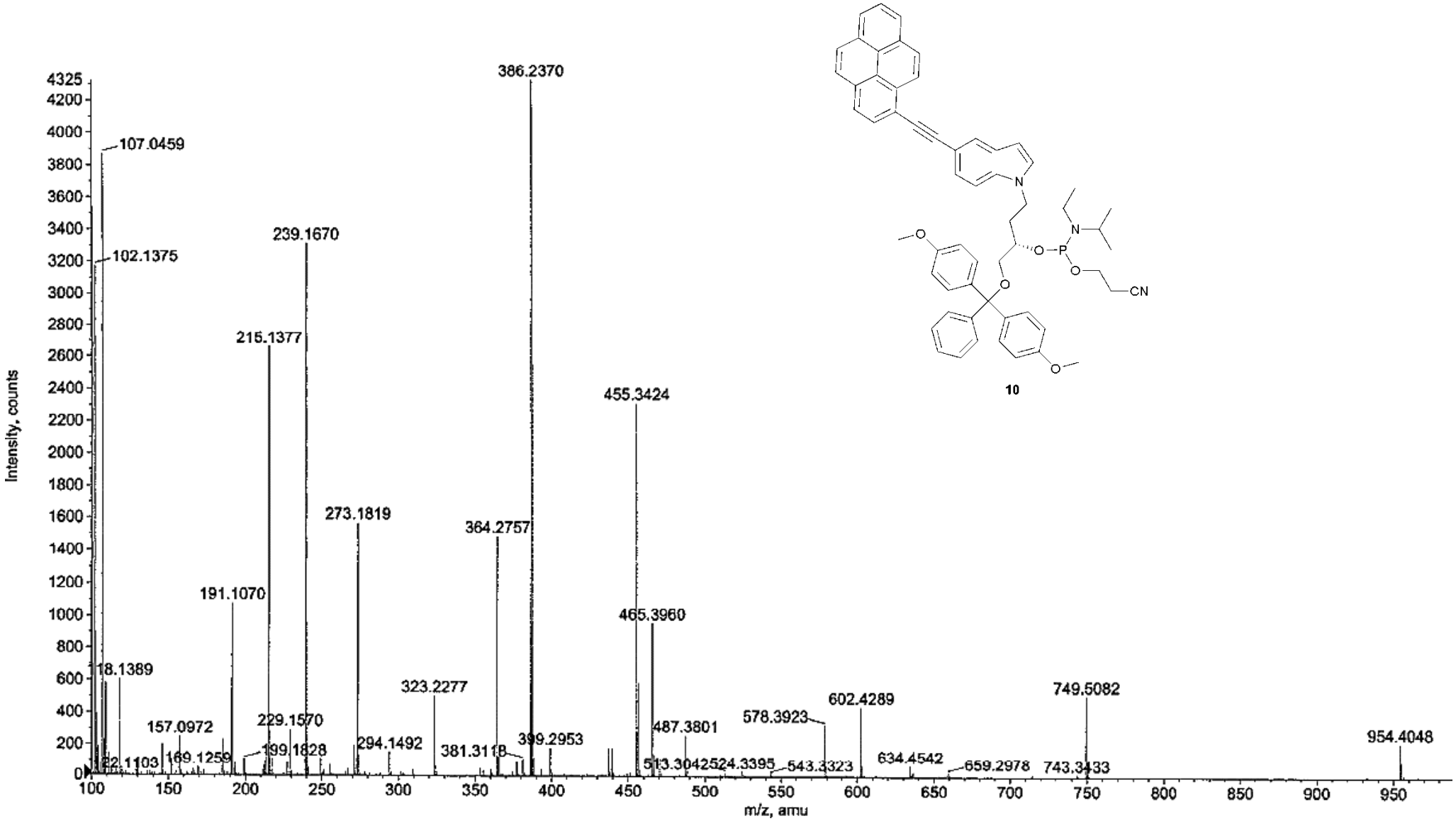
^{13}C-NMR:


158.67
158.63
136.25
136.13
135.80
131.86
131.50
131.37
130.96
130.25
130.23
130.17
129.59
129.18
129.02
128.85
128.37
128.32
128.21
127.96
127.47
126.98
126.91
126.30
126.02
125.56
125.50
125.35
125.27
125.16
125.12
124.76
124.71
124.62
119.01
118.98
117.84
117.66
114.21
114.12
113.26
109.97
109.90
101.72
101.61
97.41
97.35
86.42
86.25
71.50
58.17
58.14
55.39
43.48
43.41
43.36
43.28
42.94
42.84
33.87
24.96
24.92
24.88
24.85
24.79
24.72
23.71
23.10
19.21
19.17
CN
10
f1 (ppm)

^{31}P-NMR:



ESI-MS (TOF):


386.2370
107.0459
102.1375
239.1670
215.1377
455.3424
273.1819
364.2757
191.1070
465.3960
118.1389
323.2277
602.4289
749.5082
578.3923
157.0972
229.1570
487.3801
954.4048
294.1492
399.2953
381.3118
199.1828
169.1259
634.4542
659.2978
743.3433
543.3323
524.3395
513.3042
182.1103
Intensity, counts
m/z, amu
10