Synthesis and evaluation of *galacto*-noeurostegine and its 2-deoxy analogue as glycosidase inhibitors

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§ Present address: Oxeltis, Cap Gamma – 1682, rue de la Valsière, CS 27384, 34189 Montpellier, France.

Content

Quantum mechanical calculation setups

Noeurostegine, benzyl protected analogue:

Conformational search: Open and closed forms of the molecule were run through a conformational search using the MCMM method implemented in the Schrödinger Suite. The search was performed using 10,000 steps in the implicit $CHCl_3$ model with no cutoff. Searches were performed using both the OPLS-2005 and MMFF force fields. Unique conformations (defined by a 0.5 Å RMSD cutoff using all heavy atoms) within 10 kJ/mol of the lowest energy structure were kept for QM geometry optimisations.

Geometry optimisation: The conformations obtained from the conformational search were DFT optimised using the pcseg-1 basis set and the wb97xd functional. The IEFPCM method was used to model the presence of $CHCl_3$. Conformations within 10 kJ/mol of the lowest energy structure were kept for MP2 single point calculations.

MP2 single point calculations: MP2 calculations were performed using the aug'-cc-pVDZ basis set and the wb97xd functional. The IEFPCM method was used to model the presence of $CHCl_3$.

Noeurostegine:

Conformational search: Charged and neutral configurations of the open and closed forms of the molecule were run through a conformational search using the MCMM method implemented in the Schrödinger Suite. The search was performed using 10,000 steps in the implicit water model with no cutoff. Searches were performed using both the OPLS-2005 and MMFF force fields. Unique conformations (defined by a 0.2 Å RMSD cutoff using all heavy atoms) within 35 kJ/mol of the lowest energy structure were kept for QM geometry optimisations.

Geometry optimisation: The conformations obtained from the conformational search were DFT optimised using the pcseg-1 basis set and the wb97xd functional. The IEFPCM method was used to model the presence of water. All conformations were kept for MP2 single point calculations.

MP2 single point calculations: MP2 calculations were performed using the aug'-cc-pVTZ basis set and the wb97xd functional. The IEFPCM method was used to model the presence of water.

Galacto-*noeurostegine, benzyl protected analogue (**28**):*

Conformational search: Open and closed forms of the molecule were run through a conformational search using the MCMM method implemented in the Schrödinger Suite. The search was performed using 10,000 steps in the implicit $CHCl_3$ model with no cutoff. Searches were performed using both the OPLS-2005 and MMFF force fields. Unique conformations (defined by a 0.5 Å RMSD cutoff using all heavy atoms) within 35 kJ/mol of the lowest energy structure were kept for QM geometry optimisations.

Geometry optimisation: The conformations obtained from the conformational search were DFT optimised using the pcseg-1 basis set and the wb97xd functional. The IEFPCM method was used to model the presence of $CHCl_3$. Conformations within 20 kJ/mol of the lowest energy structure were kept for MP2 single point calculations.

MP2 single point calculations: MP2 calculations were performed using the aug'-cc-pVDZ basis set and the wb97xd functional. The IEFPCM method was used to model the presence of $CHCl_3$.

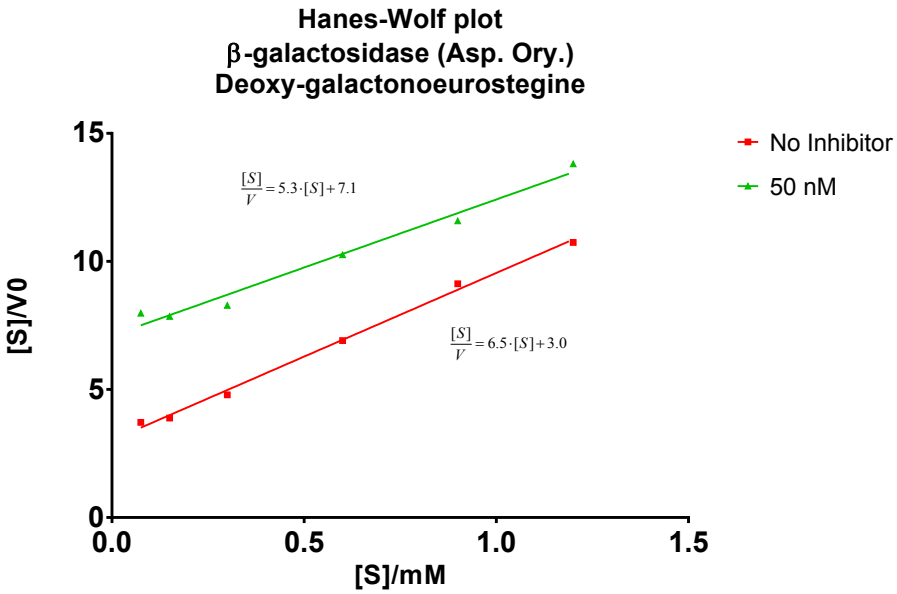
Galacto-*noeurostegine (**5**):*

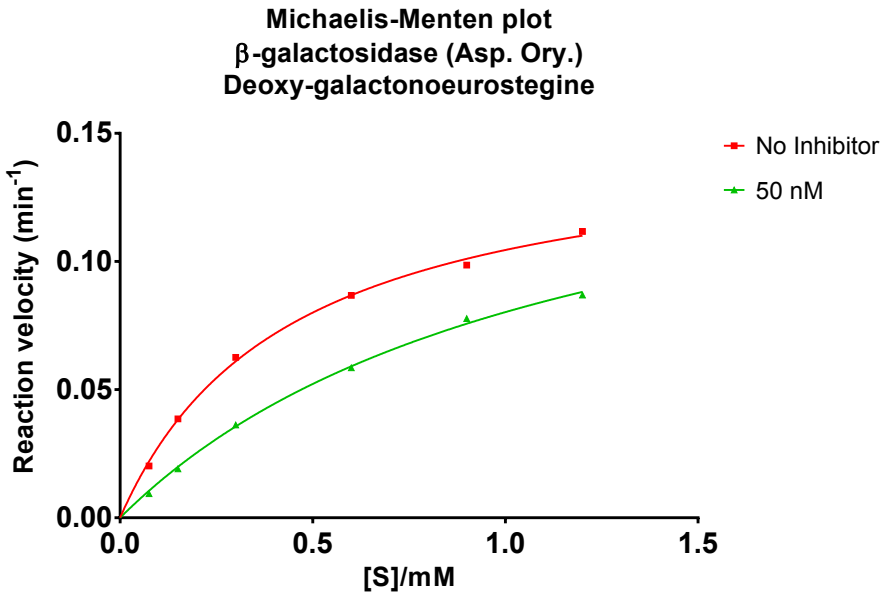
Conformational search: Charged and neutral configurations of the open and closed forms of the molecule were run through a conformational search using the MCMM method implemented in the Schrödinger Suite. The search was performed using 10,000 steps in the implicit water model with no cutoff. Searches were performed using both the OPLS-2005 and MMFF force fields. Unique conformations (defined by a 0.2 Å RMSD cutoff using all heavy atoms) within 50 kJ/mol of the lowest energy structure were kept for QM geometry optimisations.

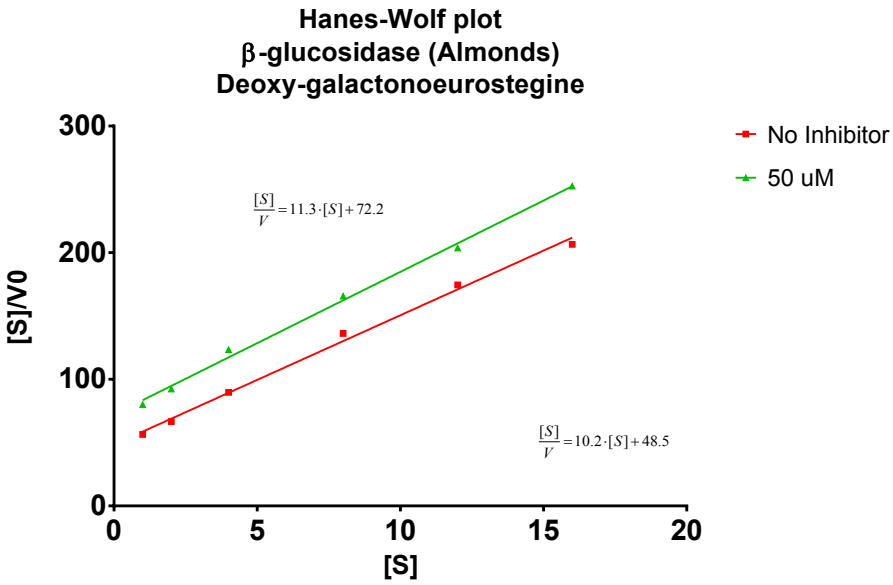
Geometry optimisation: The conformations obtained from the conformational search were DFT optimised using the pcseg-1 basis set and the wb97xd functional. The IEFPCM Method was used to model the presence of water. Conformations within 20 kJ/mol of the lowest energy structure were kept for MP2 single point calculations.

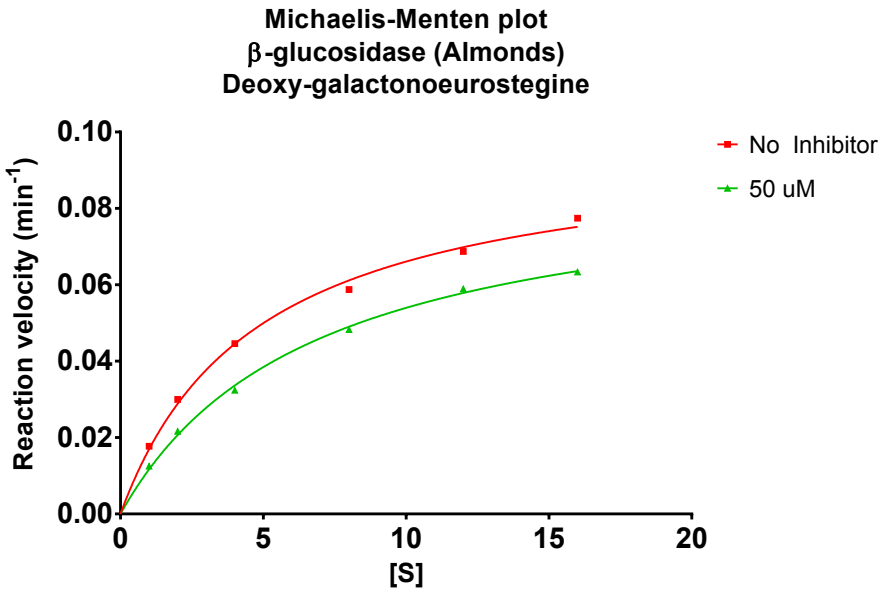
MP2 single point calculations: MP2 calculations were performed using the aug'-cc-pVTZ basis set and the wb97xd functional. The IEFPCM method was used to model the presence of water.

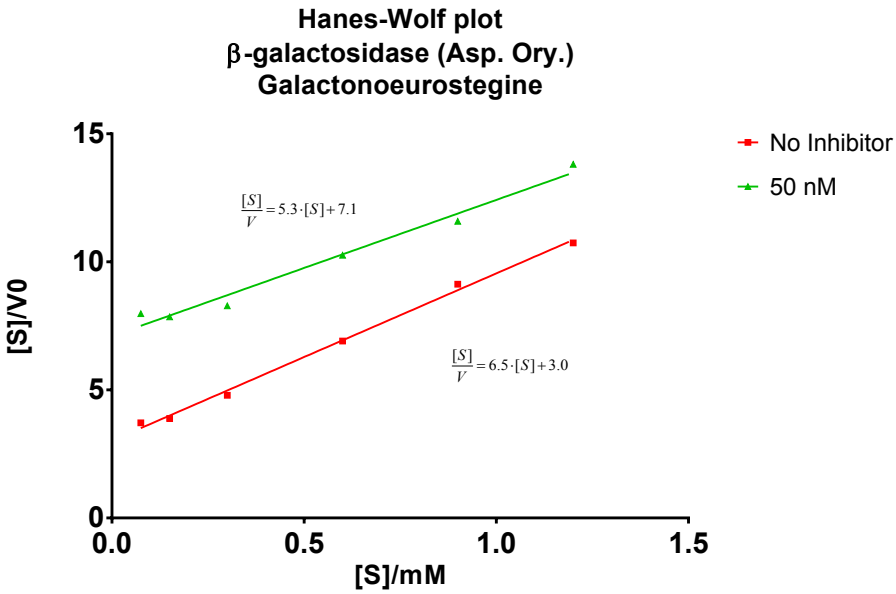
Michaelis-Menten and Hanes plots for compounds 5 and 6

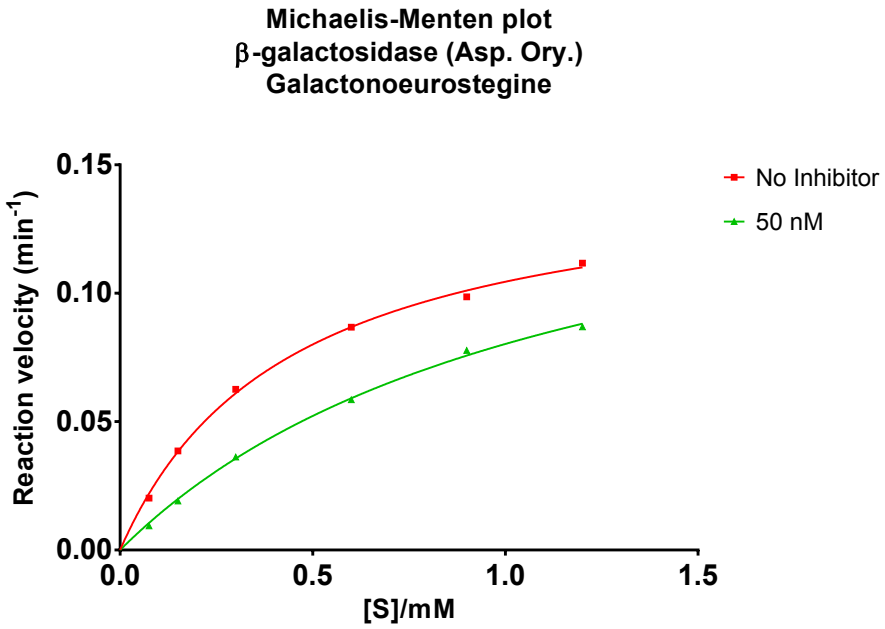


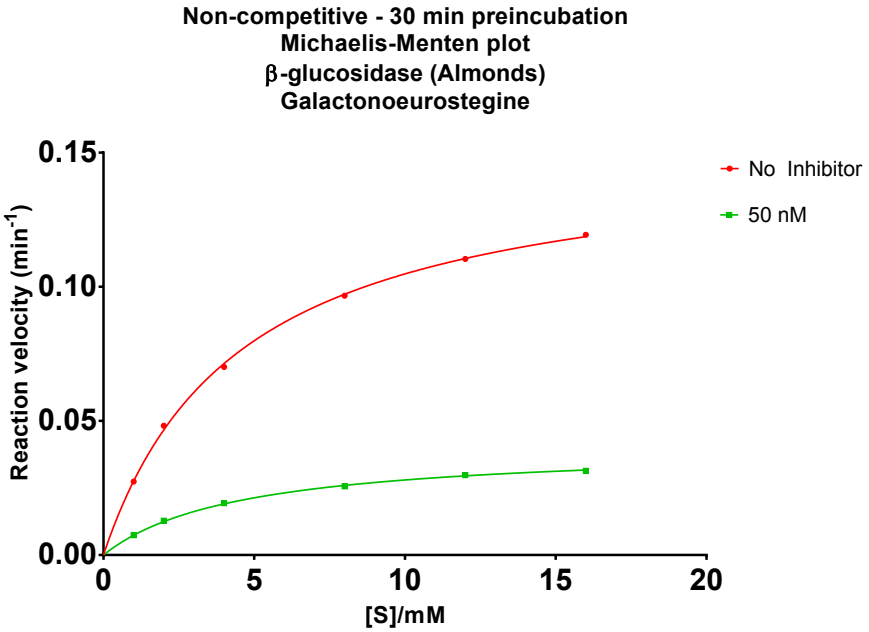


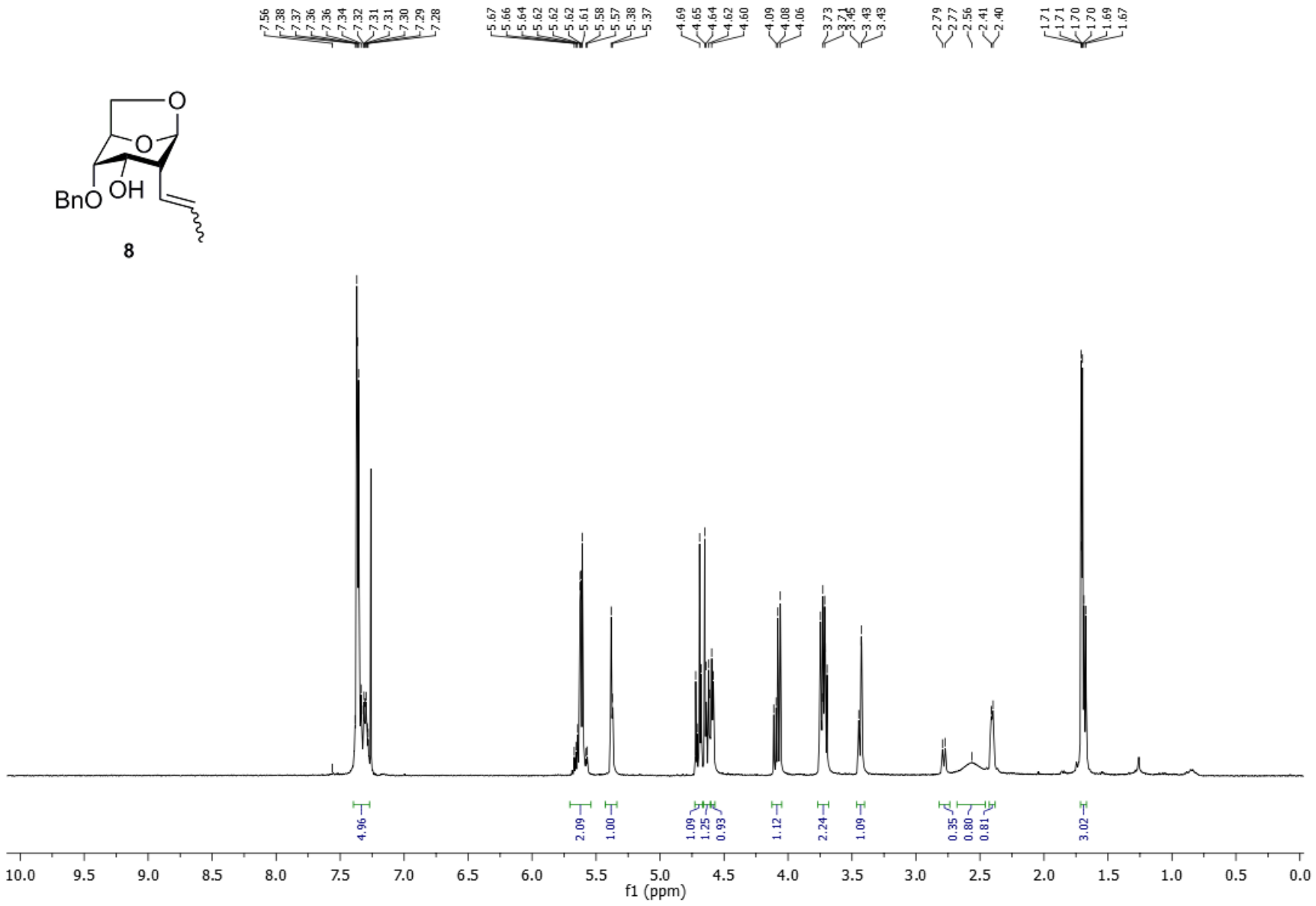

Hanes-Wolf plot
β-glucosidase (Almonds)
Deoxy-galactonoeurostegine
[S]/V0
[S]
No Inhibitor
50 uM
$\frac{[S]}{V} = 11.3 \cdot [S] + 72.2$
$\frac{[S]}{V} = 10.2 \cdot [S] + 48.5$
0
100
200
300
0
5
10
15
20


Michaelis-Menten plot
β-glucosidase (Almonds)
Deoxy-galactonoeurostegine
Reaction velocity (min-1)
[S]
No Inhibitor
50 uM
0.00
0.02
0.04
0.06
0.08
0.10
0
5
10
15
20


Hanes-Wolf plot
β-galactosidase (Asp. Ory.)
Galactonoeurostegine
[S]/V0
[S]/mM
No Inhibitor
50 nM
$\frac{[S]}{V} = 5.3 \cdot [S] + 7.1$
$\frac{[S]}{V} = 6.5 \cdot [S] + 3.0$
0
5
10
15
0.0
0.5
1.0
1.5


Michaelis-Menten plot
β-galactosidase (Asp. Ory.)
Galactonoeurostegine
Reaction velocity (min-1)
[S]/mM
No Inhibitor
50 nM
0.00
0.05
0.10
0.15
0.0
0.5
1.0
1.5


Non-competitive - 30 min preincubation
Michaelis-Menten plot
β-glucosidase (Almonds)
Galactonoeurostegine
Reaction velocity (min-1)
0.15
0.10
0.05
0.00
0
5
10
15
20
[S]/mM
No Inhibitor
50 nM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7.32
7.31
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7.30
7.29
7.28
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5.66
5.64
5.62
5.62
5.61
5.58
5.57
5.38
5.37
4.69
4.65
4.64
4.62
4.60
4.09
4.08
4.06
3.73
3.71
3.45
3.43
3.43
2.79
2.77
2.56
2.41
2.40
1.71
1.71
1.70
1.70
1.69
1.67
BnO
OH
O
O
8
4.96
2.09
1.00
1.09
1.25
0.93
1.12
2.24
1.09
0.35
0.80
0.81
3.02
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
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3.0
2.5
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1.0
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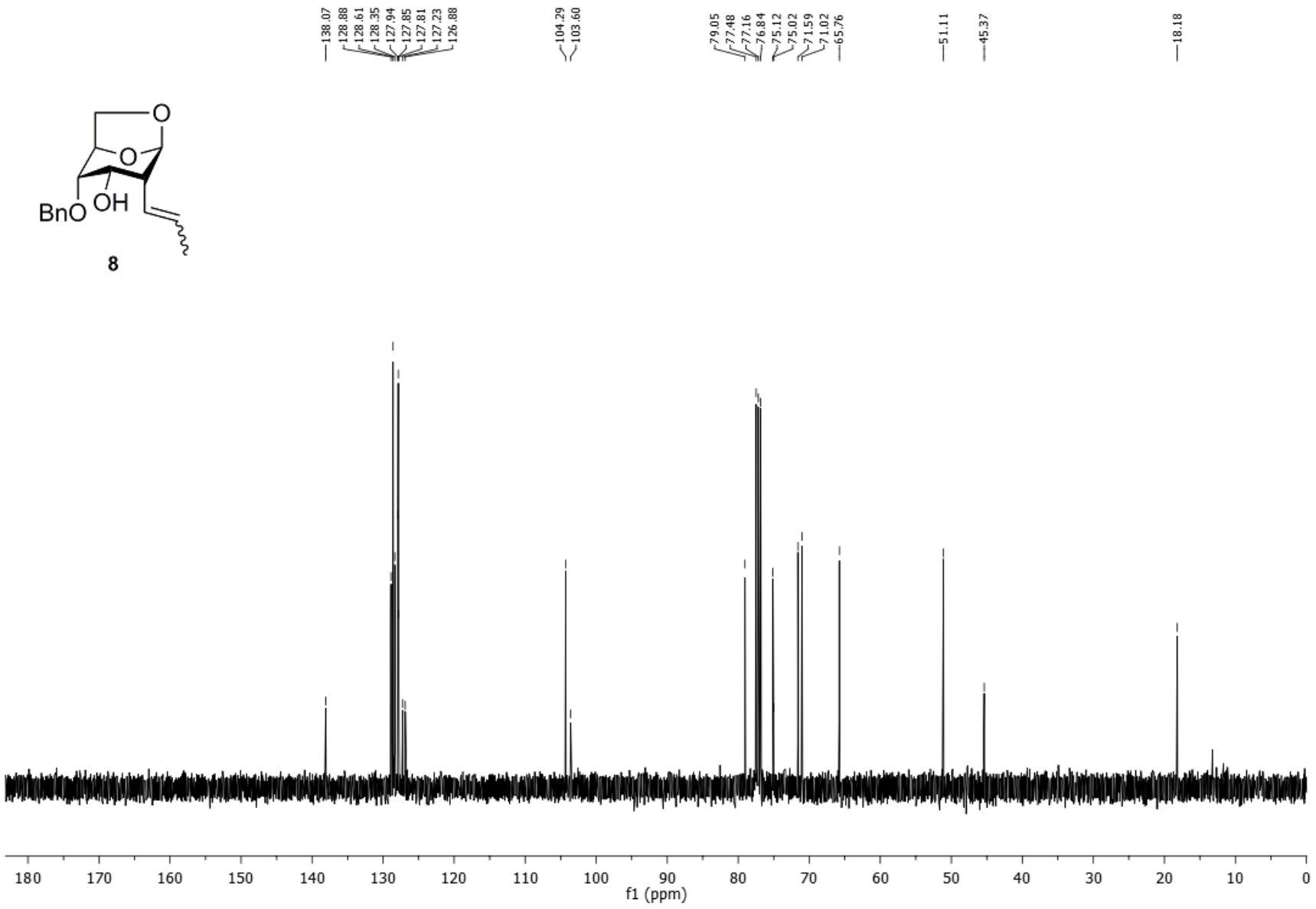
f1 (ppm)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127.81
127.23
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79.05
77.48
77.16
76.84
75.12
75.02
71.59
71.02
65.76
51.11
45.37
18.18
O
O
BnO
OH
8
180
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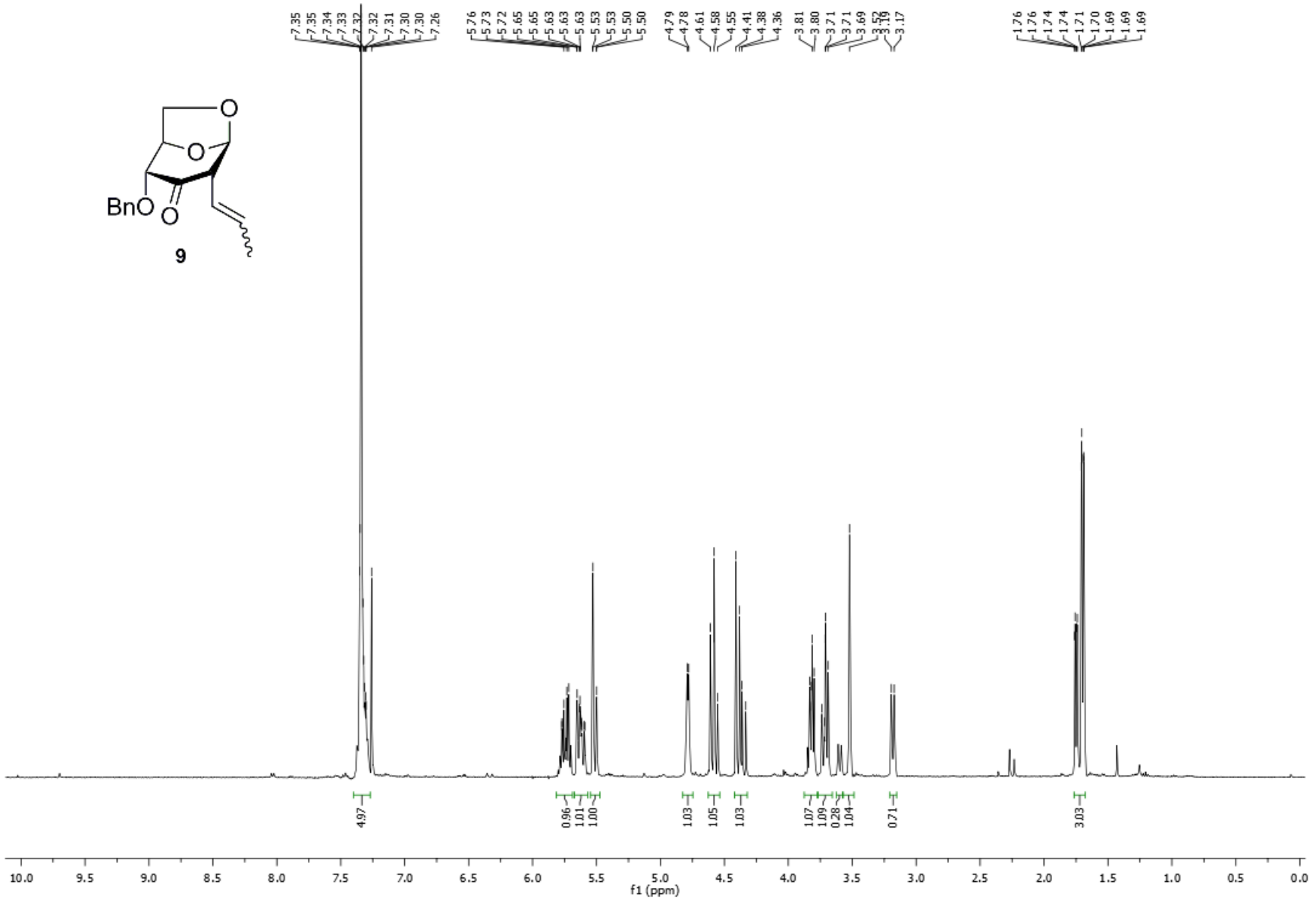
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)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O
O
O
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7.34
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7.32
7.32
7.31
7.30
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7.26
5.76
5.73
5.72
5.65
5.65
5.63
5.63
5.63
5.53
5.53
5.50
5.50
4.79
4.78
4.61
4.58
4.55
4.41
4.38
4.36
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3.69
3.52
3.17
1.76
1.76
1.74
1.74
1.71
1.70
1.69
1.69
1.69
4.97
0.96
1.01
1.00
1.03
1.05
1.03
1.07
1.09
0.28
1.04
0.71
3.03
10.0
9.5
9.0
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8.0
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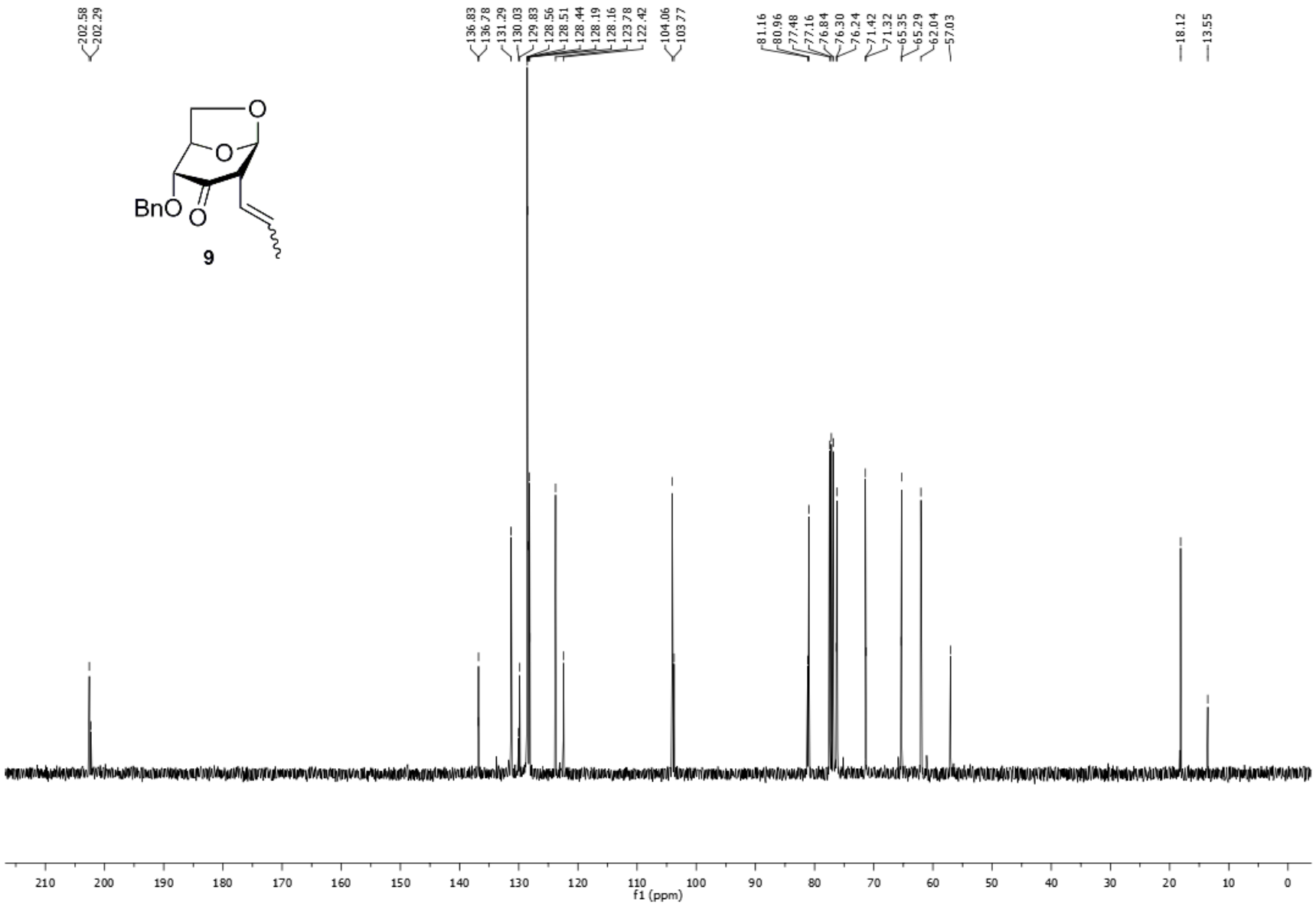
f1 (ppm)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122.42
104.06
103.77
81.16
80.96
77.48
77.16
76.84
76.30
76.24
71.42
71.32
65.35
65.29
62.04
57.03
18.12
13.55
BnO
O
O
O
9
210
200
190
180
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150
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120
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50
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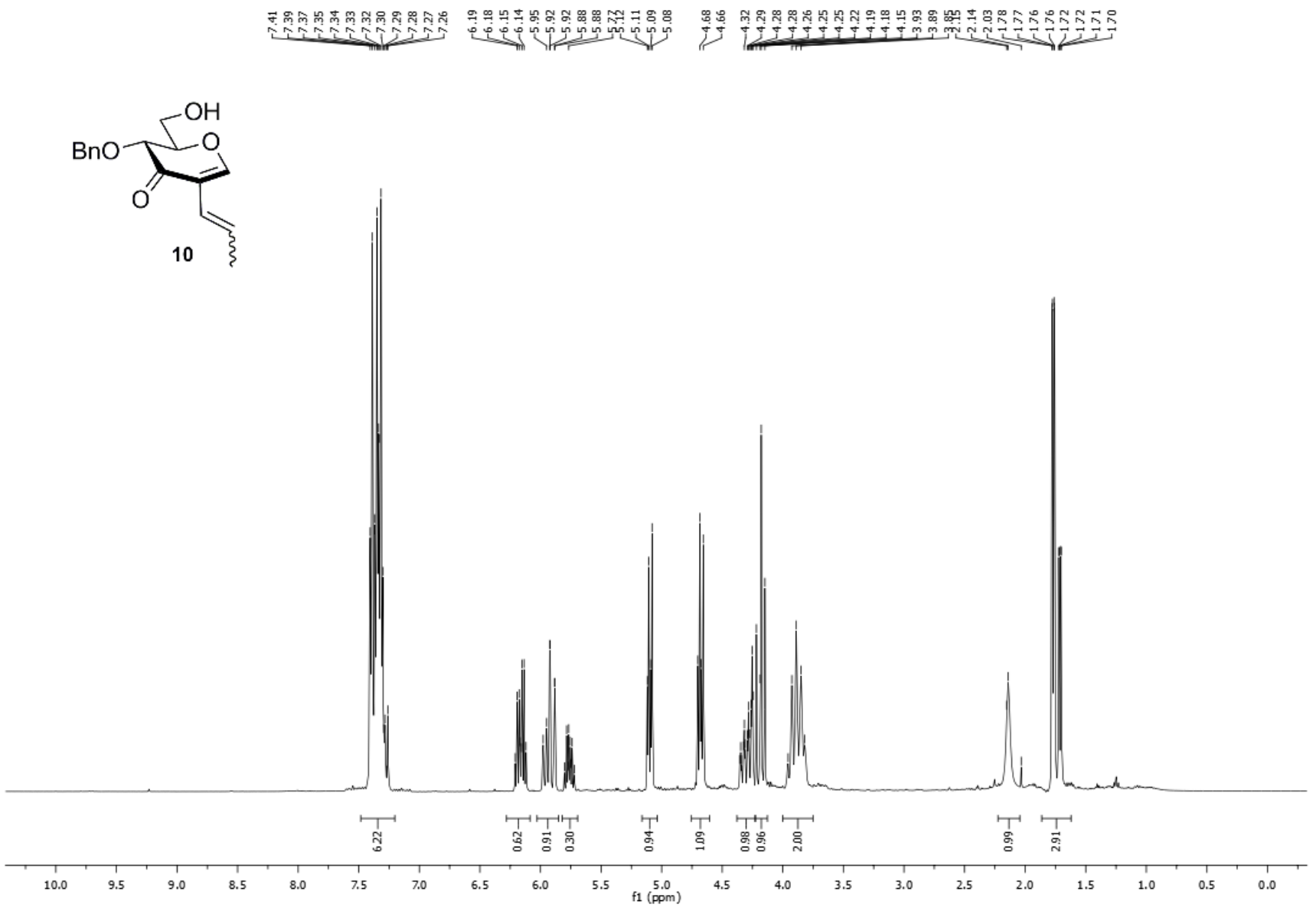
20
10
0
f1 (ppm)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6.18
6.15
6.14
5.95
5.92
5.92
5.88
5.88
5.12
5.11
5.09
5.08
4.68
4.66
4.32
4.29
4.28
4.28
4.26
4.25
4.25
4.22
4.19
4.18
4.15
3.93
3.89
3.85
2.15
2.14
2.03
1.78
1.77
1.76
1.76
1.72
1.72
1.71
1.70
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BnO
O
O
10
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0.30
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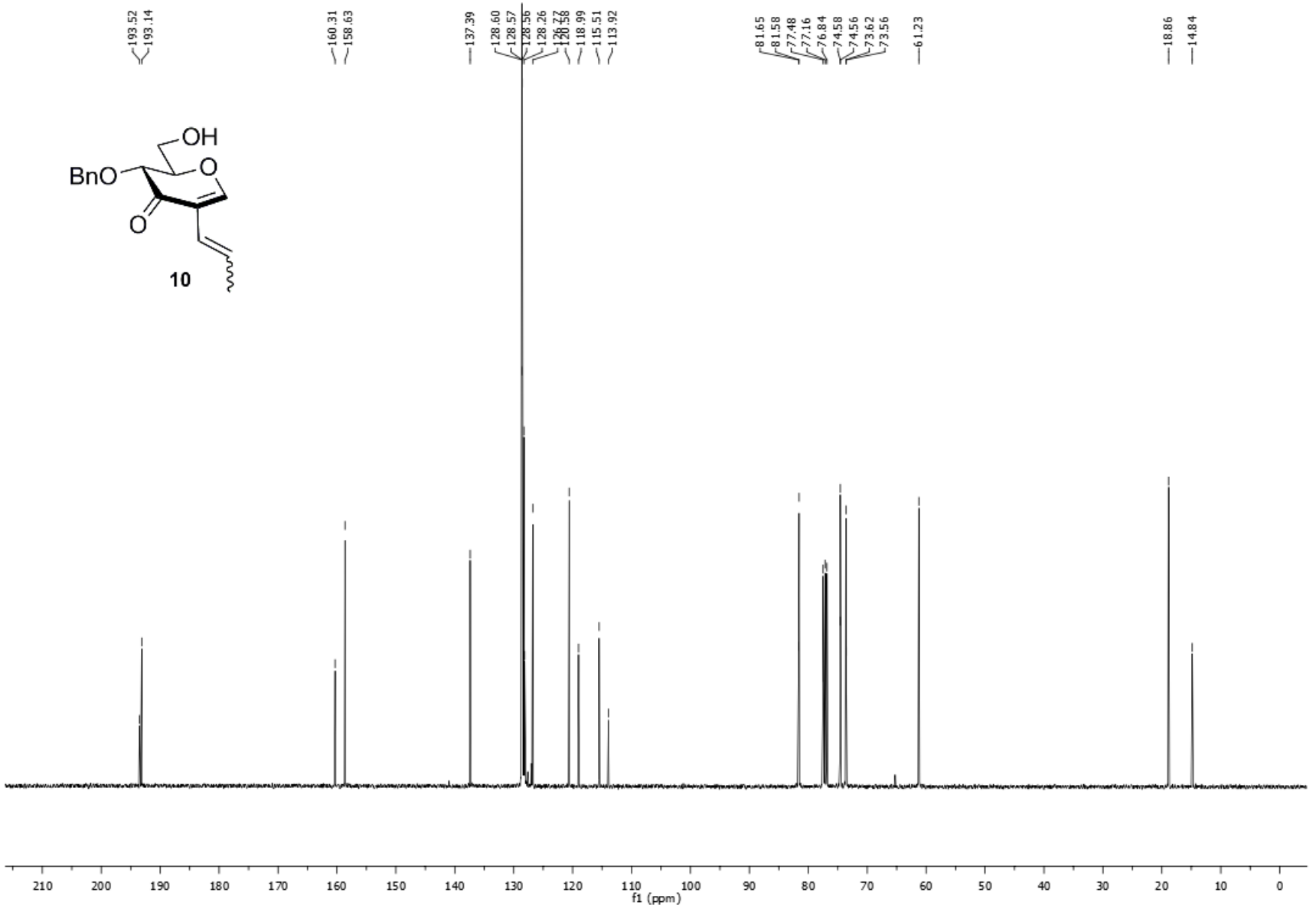
f1 (ppm)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81.58
77.48
77.16
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74.58
74.56
73.62
73.56
61.23
18.86
14.84
OH
BnO
O
O
10
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200
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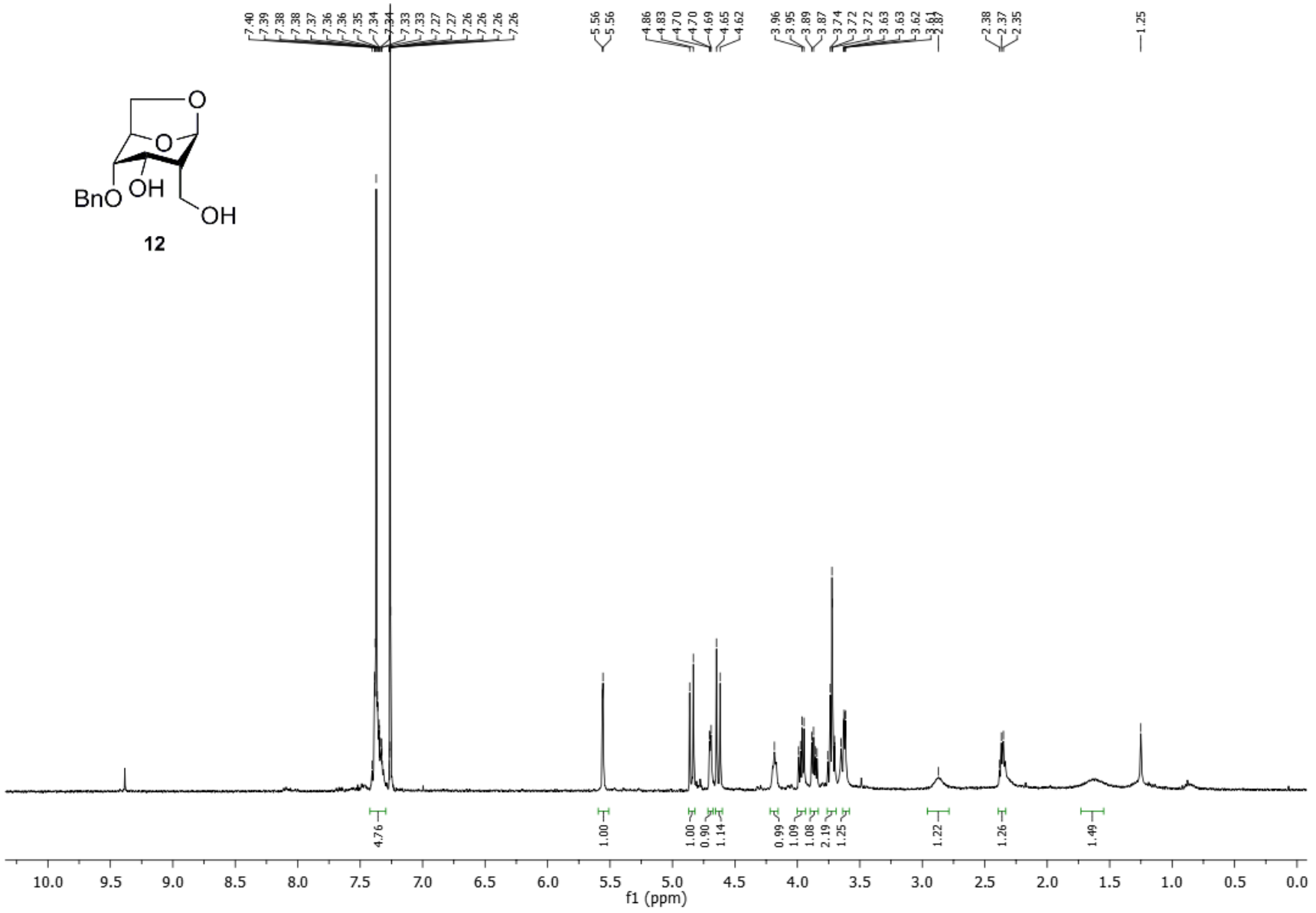
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OH
O
O
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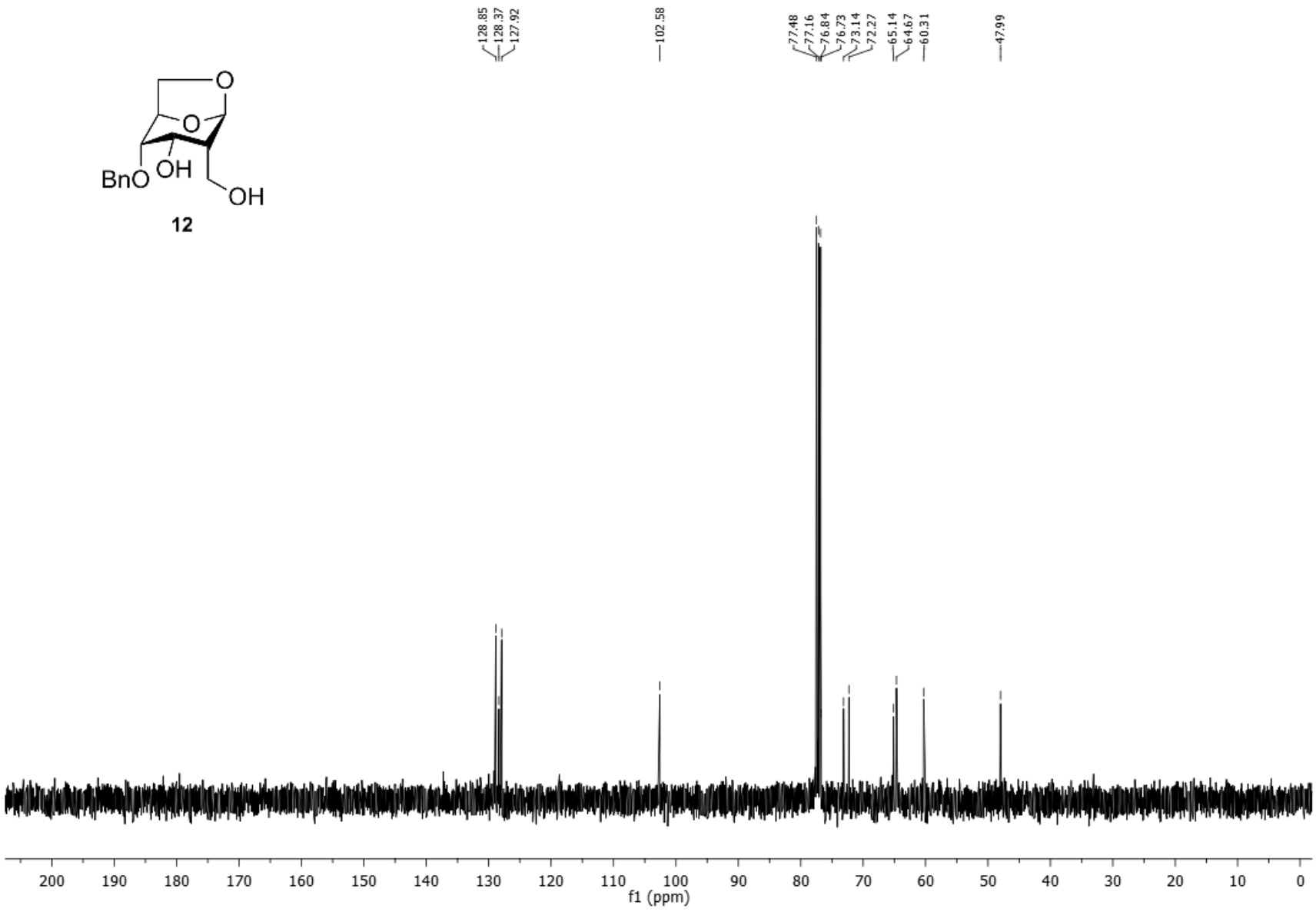
f1 (ppm)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60.31
47.99
O
O
BnO
OH
OH
12
200
190
180
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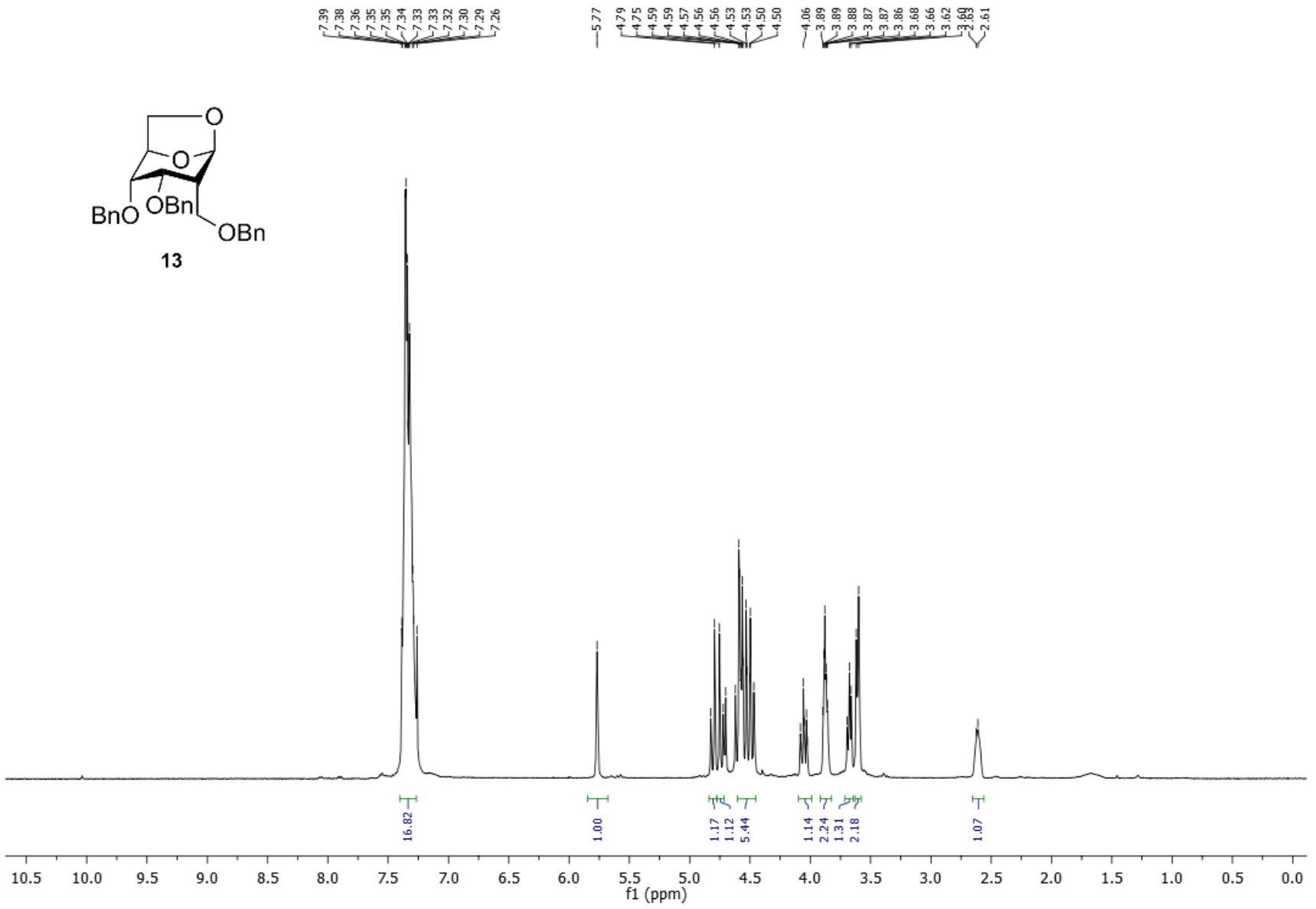
f1 (ppm)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3.59
2.61
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O
BnO
OBn
OBn
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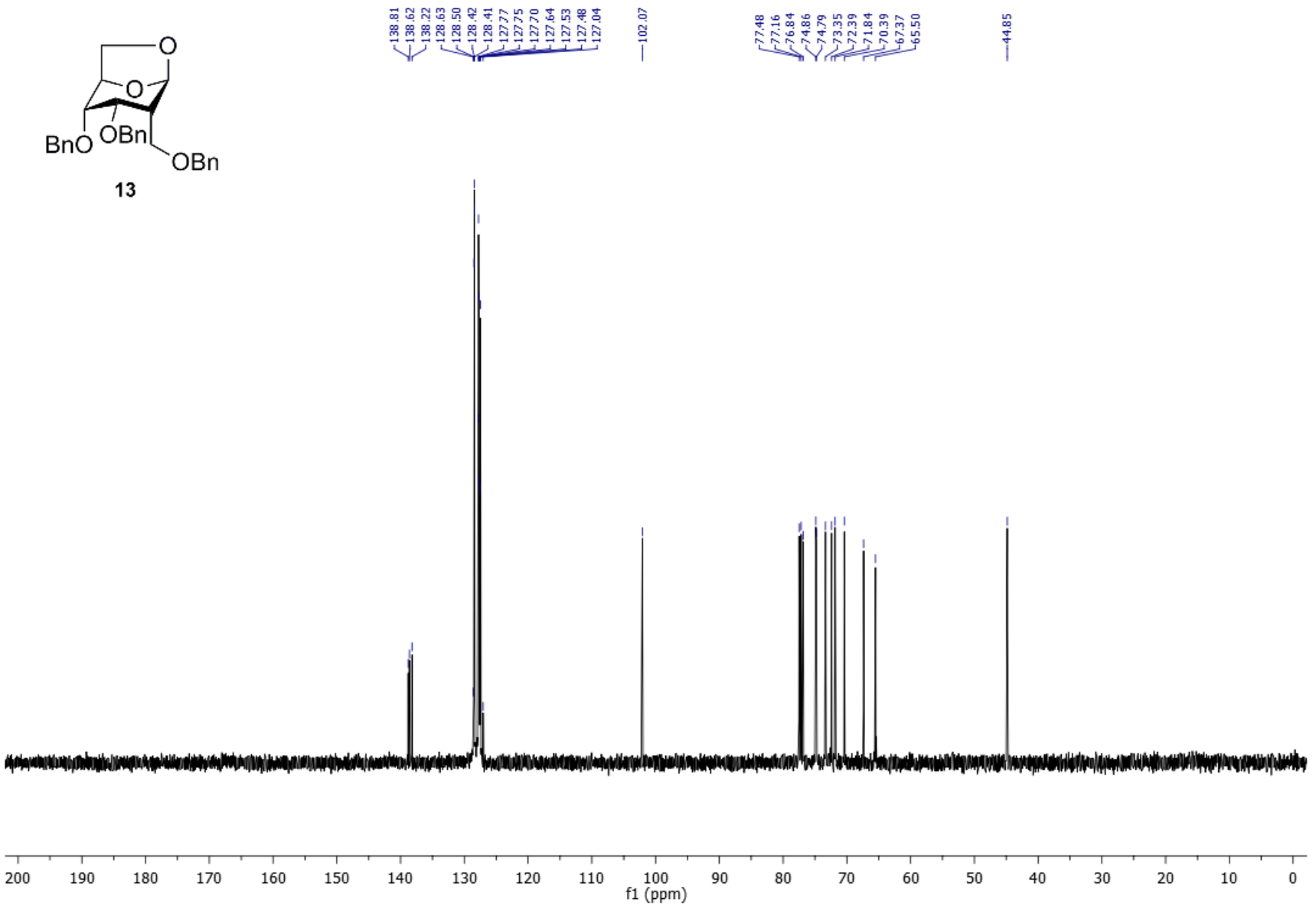
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67.37
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44.85
BnO
OBn
OBn
O
O
13
200
190
180
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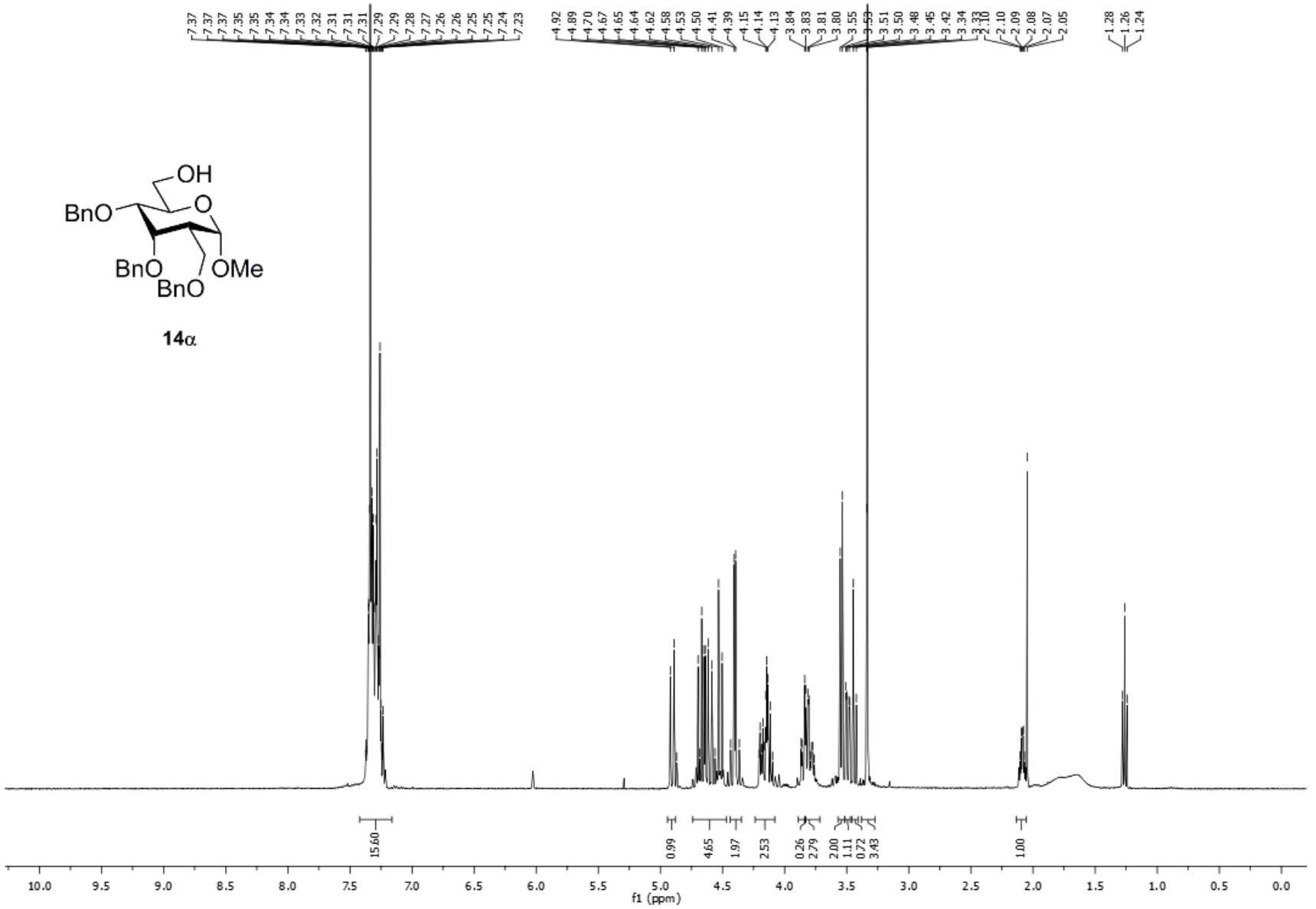
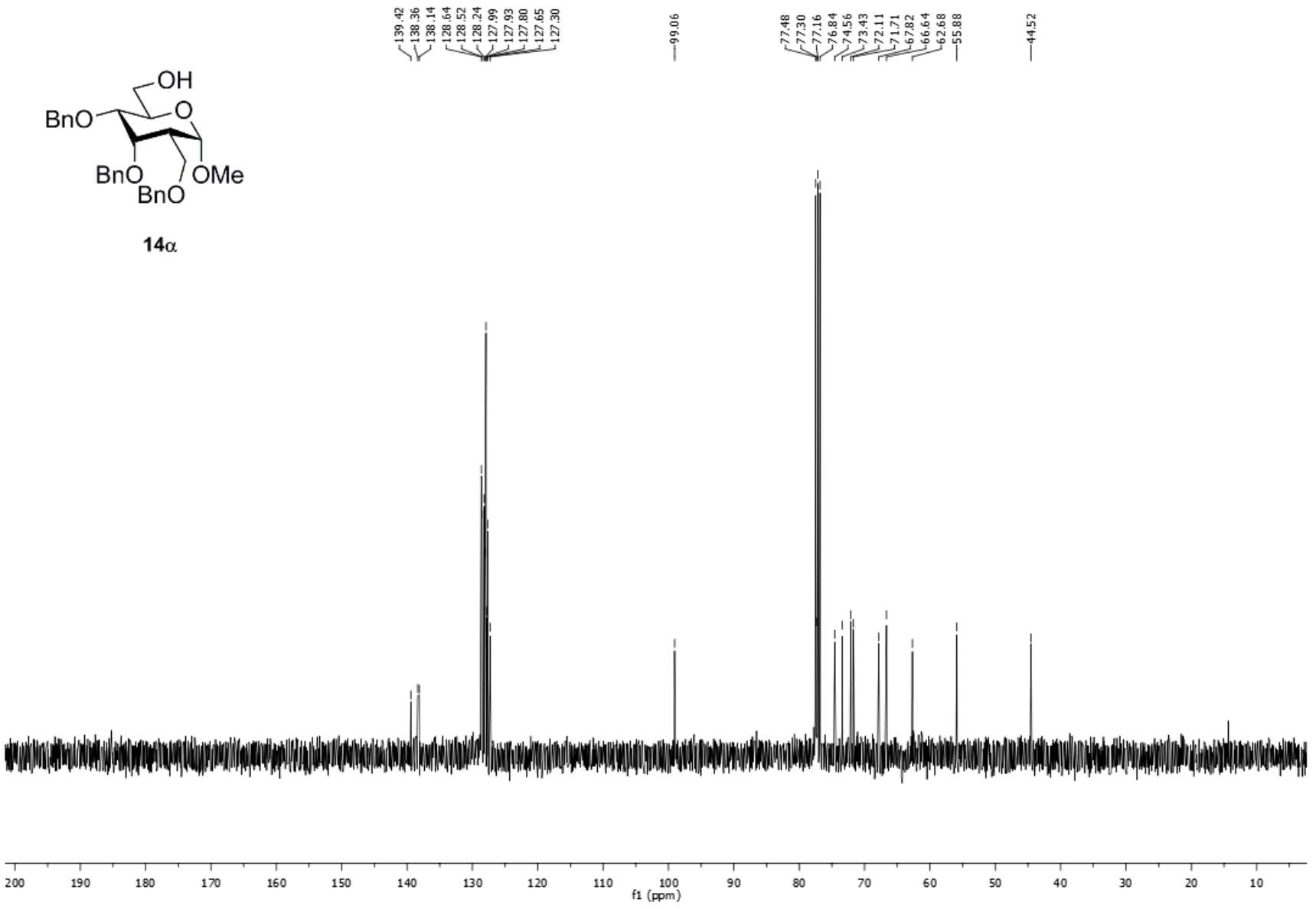

OH
O
BnO
BnO
BnO
OMe
14α
7.37 7.37 7.37 7.35 7.35 7.34 7.34 7.33 7.32 7.31 7.31 7.31 7.29 7.29 7.28 7.27 7.26 7.26 7.25 7.25 7.24 7.23
4.92 4.89 4.70 4.67 4.65 4.64 4.62 4.58 4.53 4.50 4.41 4.39 4.15 4.14 4.13 3.84 3.83 3.81 3.80 3.55 3.53 3.51 3.50 3.48 3.45 3.42 3.34 2.10 2.09 2.08 2.07 2.05
1.28 1.26 1.24
15.60 0.99 4.65 1.97 2.53 0.26 2.79 2.00 1.11 0.72 3.43 1.00
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)


OH
BnO
O
BnO
OMe
BnO
14α
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127.80
127.65
127.30
99.06
77.48
77.30
77.16
76.84
74.56
73.43
72.11
71.71
67.82
66.64
62.68
55.88
44.52
200
190
180
170
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150
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120
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100
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80
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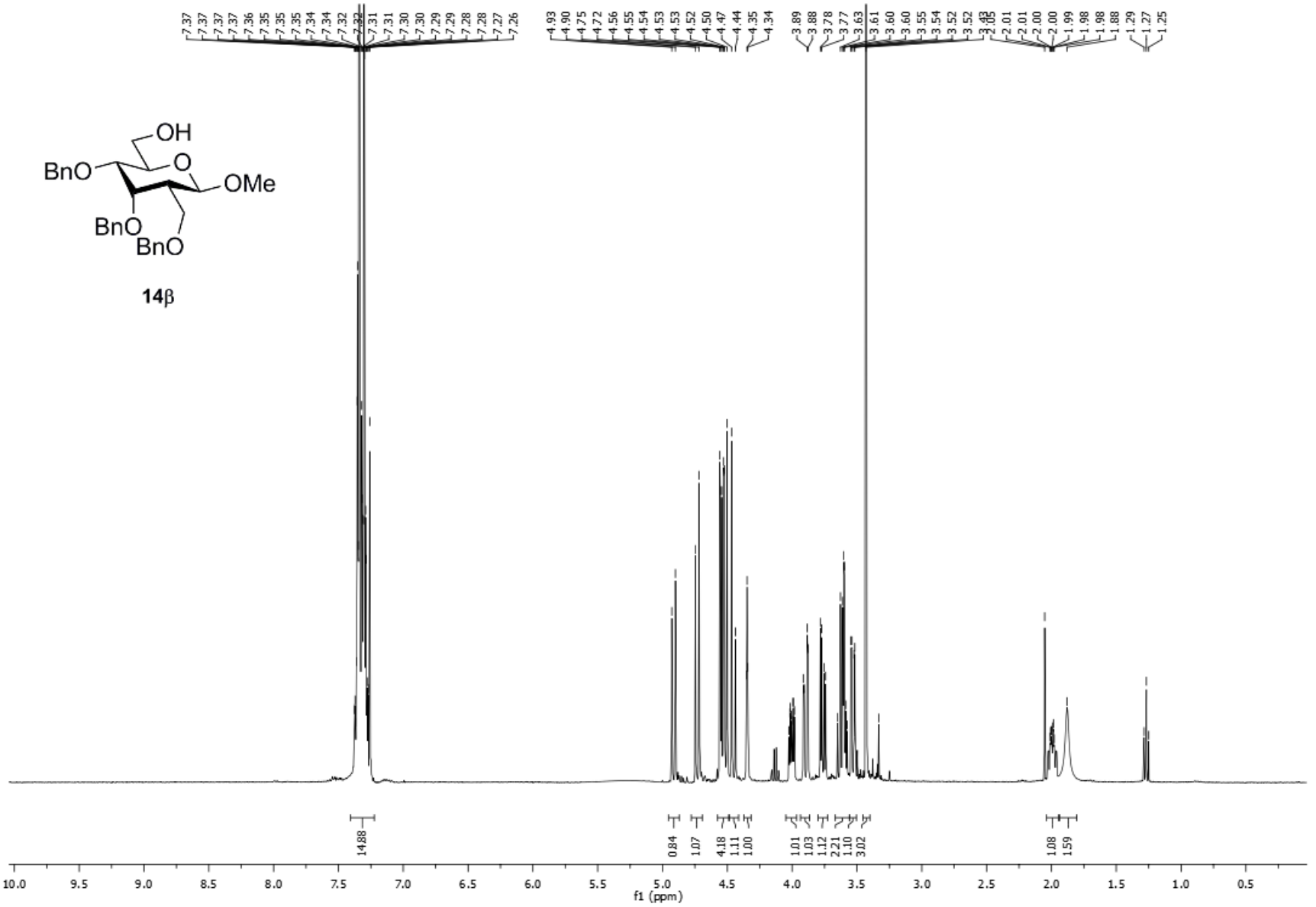
50
40
30
20
10
f1 (ppm)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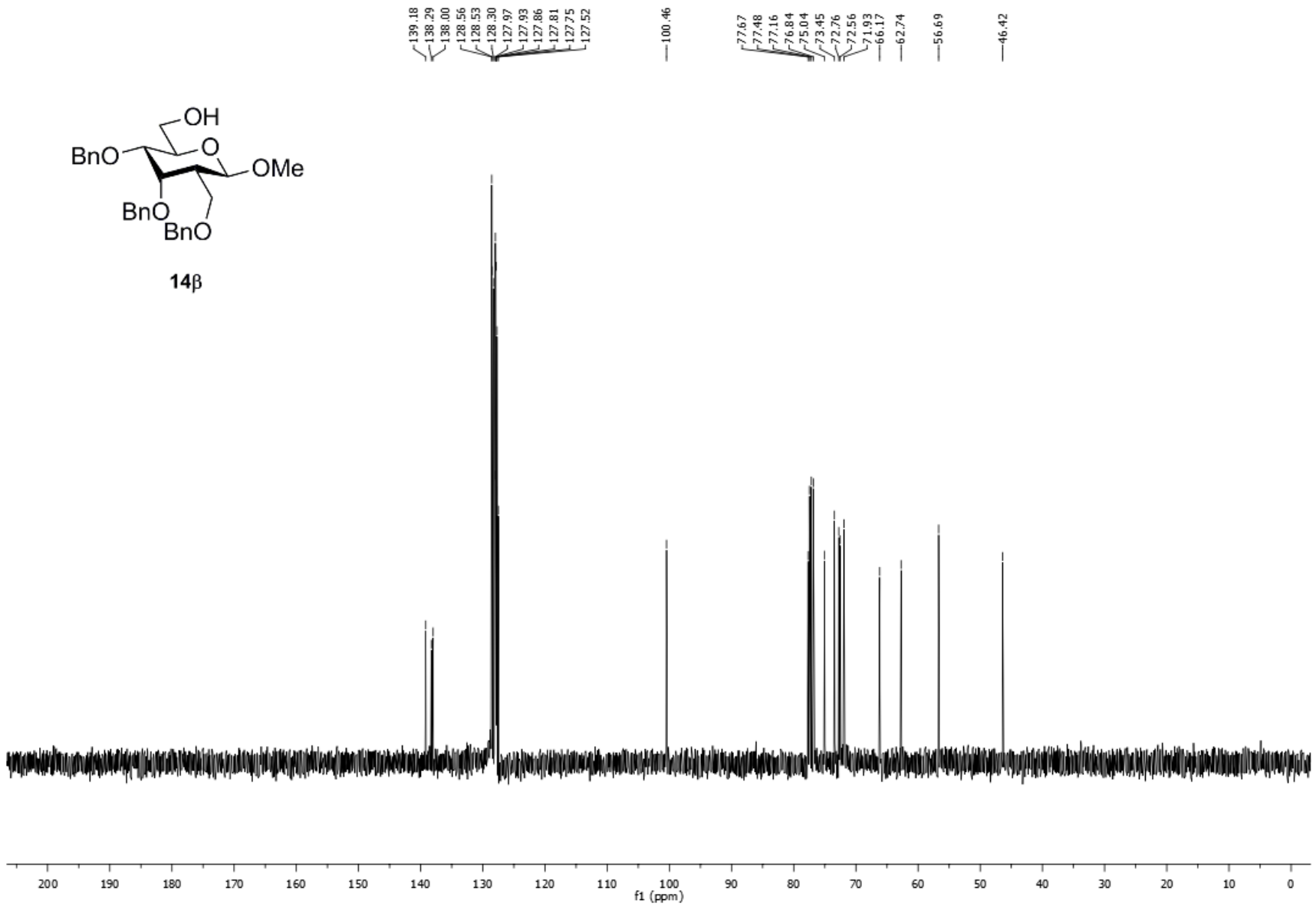
O
BnO
OMe
BnO
BnO
14β
f1 (ppm)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138.29
138.00
128.56
128.53
128.30
127.97
127.93
127.86
127.81
127.75
127.52
100.46
77.67
77.48
77.16
76.84
75.04
73.45
72.76
72.56
71.93
66.17
62.74
56.69
46.42
OH
O
BnO
OMe
BnO
BnO
14β
200
190
180
170
160
150
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120
110
100
90
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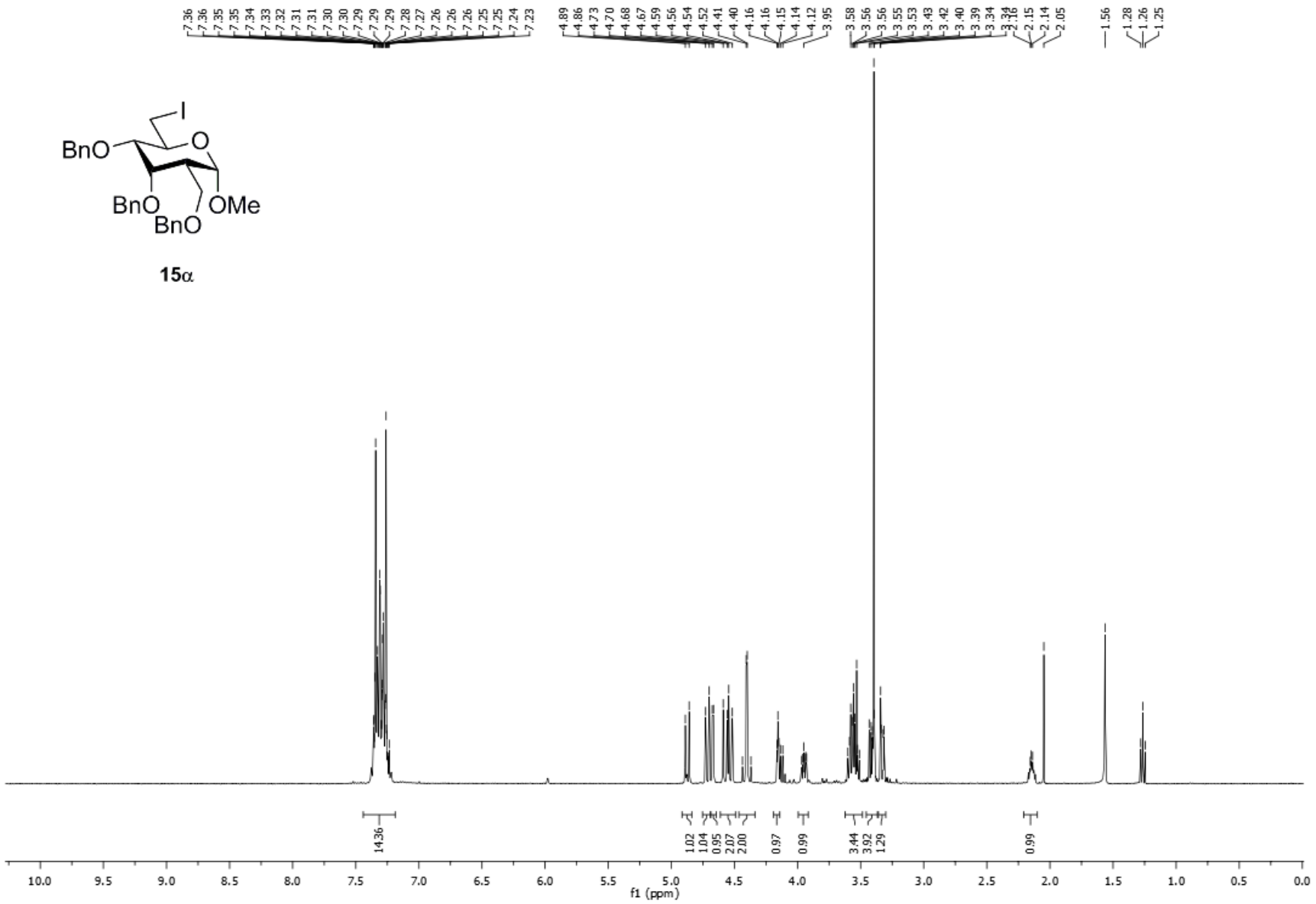
60
50
40
30
20
10
0
f1 (ppm)


I
O
BnO
BnO
BnO
OMe
15α
7.36
7.36
7.35
7.35
7.34
7.33
7.32
7.31
7.31
7.30
7.30
7.29
7.29
7.29
7.28
7.27
7.26
7.26
7.26
7.25
7.25
7.24
7.23
4.89
4.86
4.73
4.70
4.68
4.67
4.59
4.56
4.54
4.52
4.41
4.40
4.16
4.16
4.15
4.14
4.12
3.95
3.58
3.56
3.56
3.55
3.53
3.43
3.42
3.40
3.39
3.34
3.34
3.16
2.15
2.14
2.05
1.56
1.28
1.26
1.25
14.36
1.02
1.04
0.95
2.07
2.00
0.97
0.99
3.44
3.92
1.29
0.99
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
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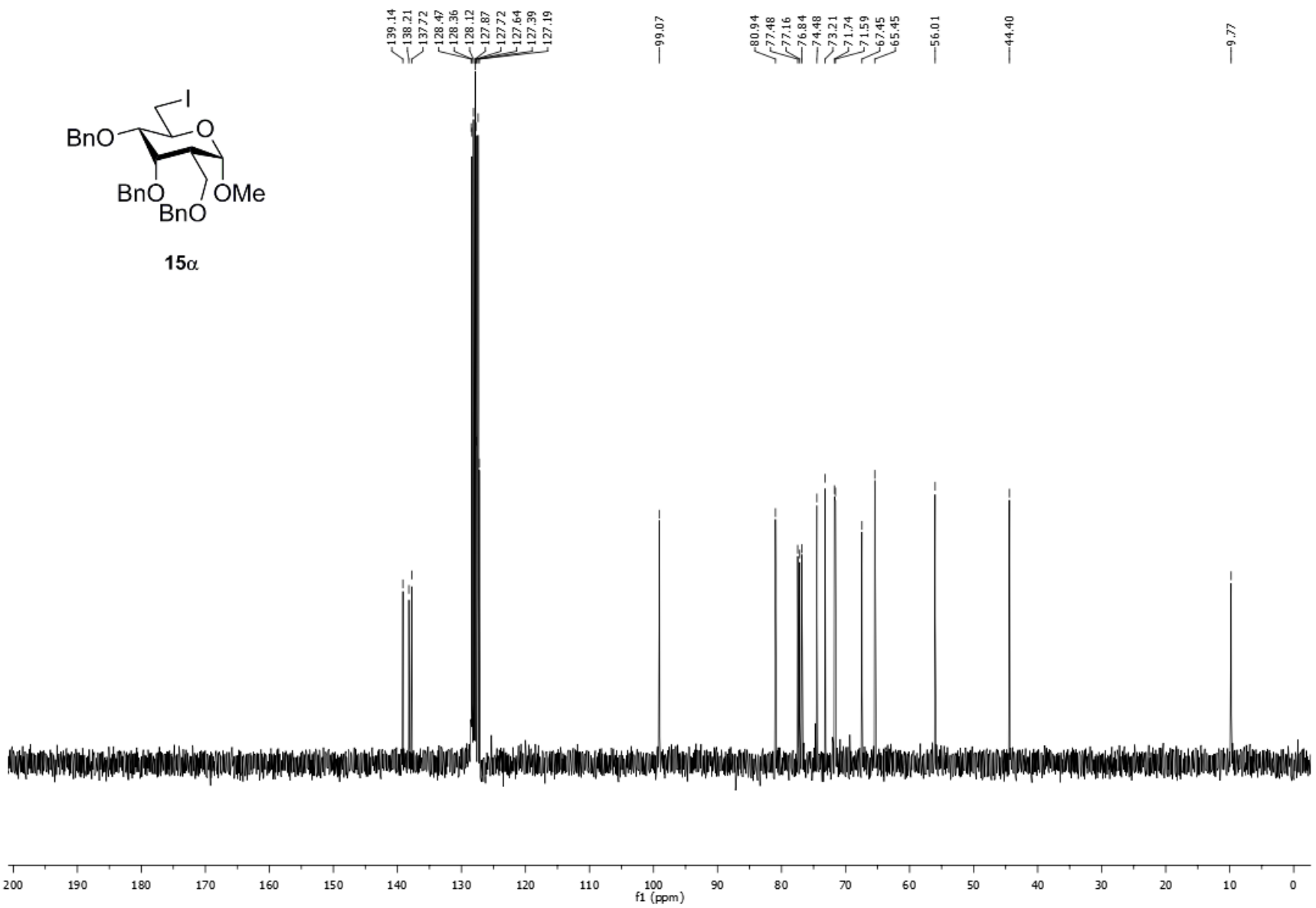
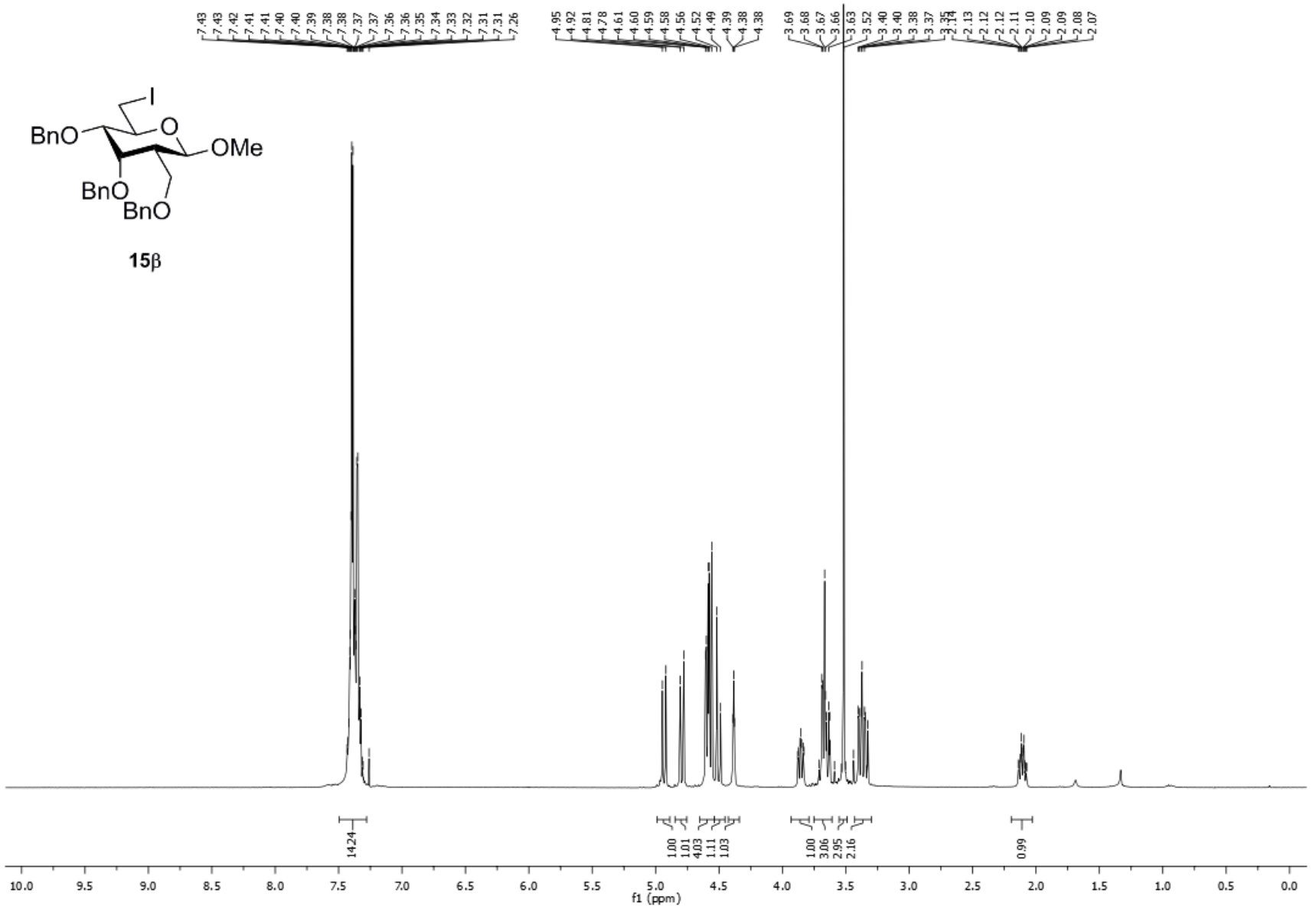
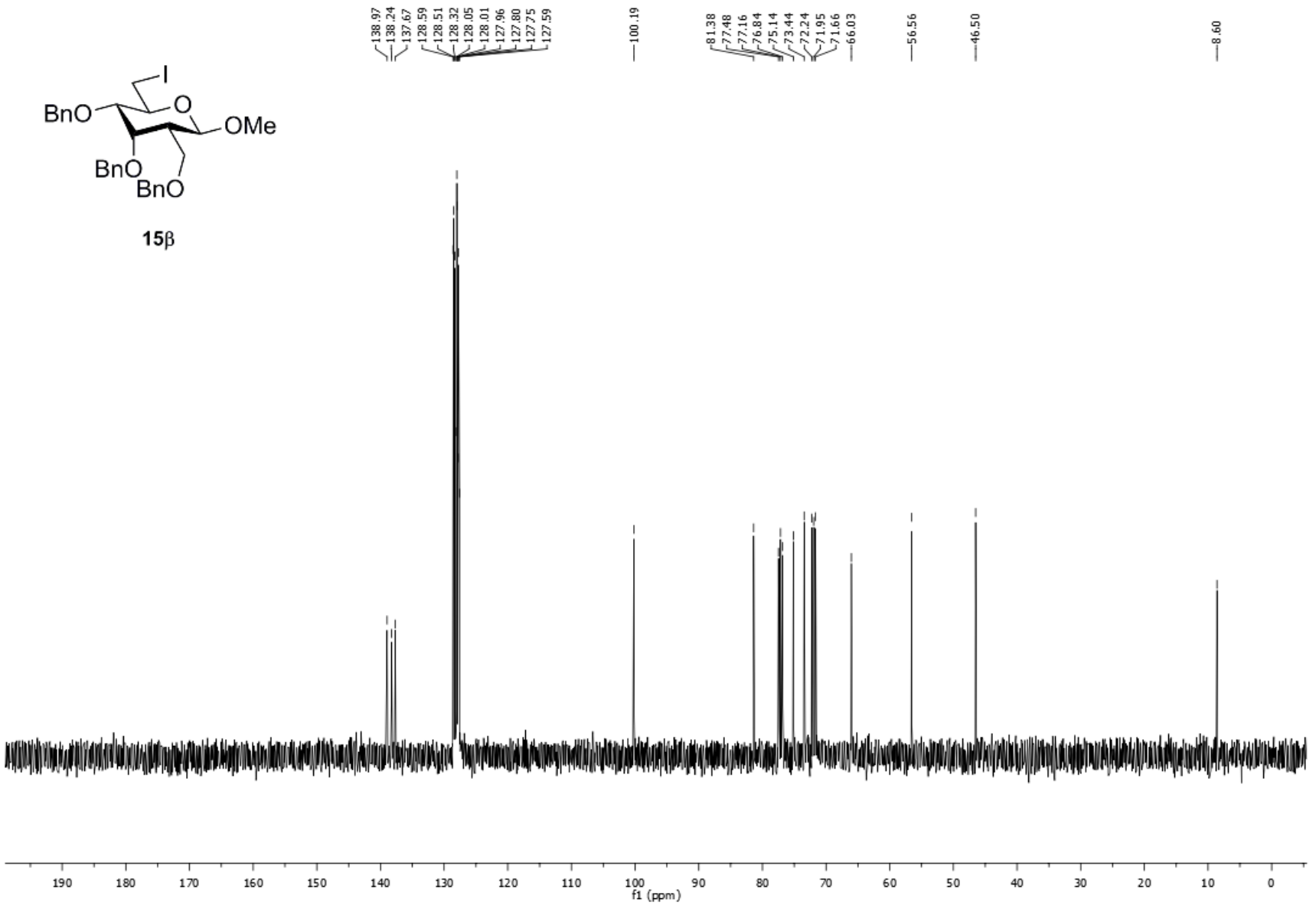
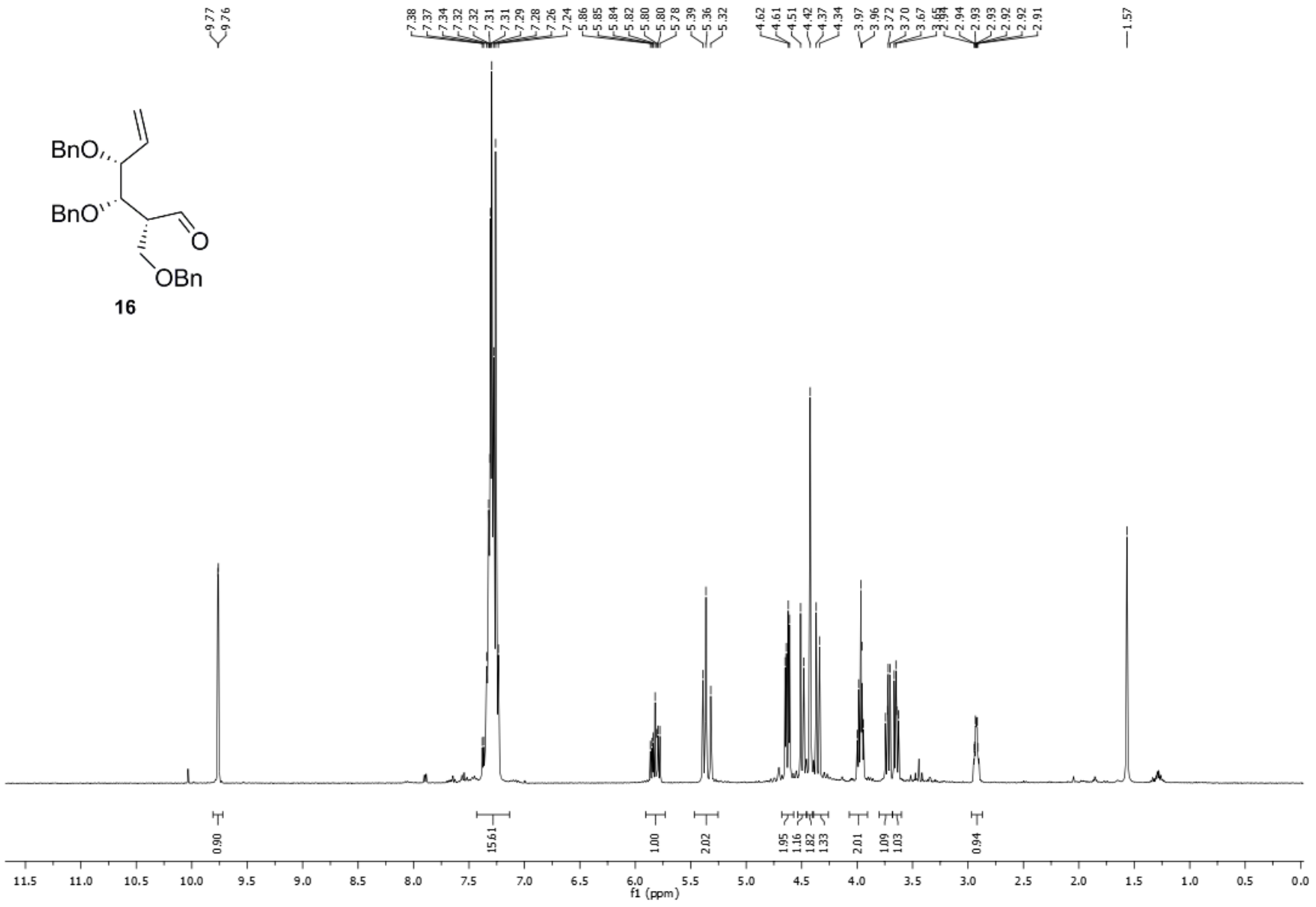
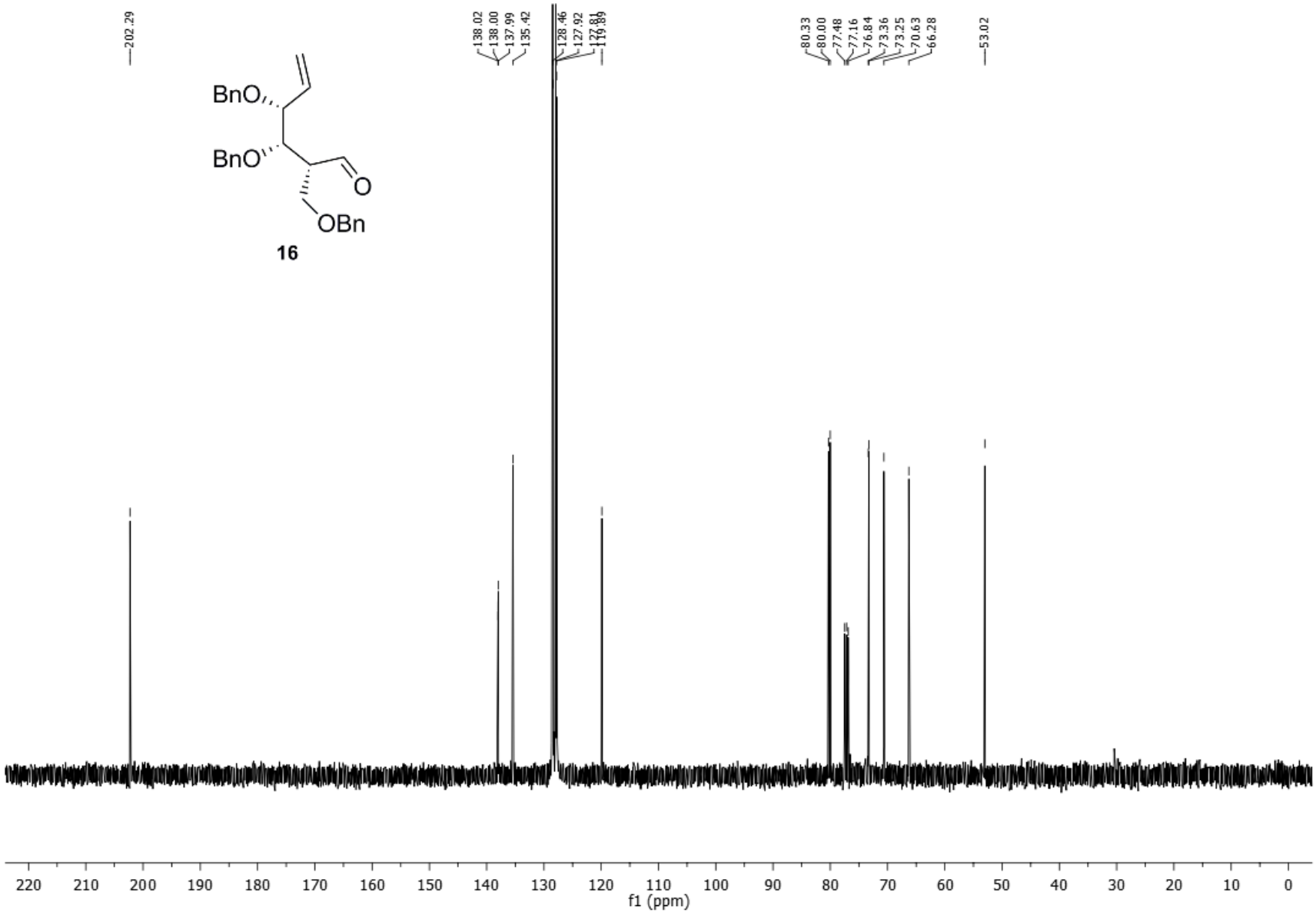
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


139.14
138.21
137.72
128.47
128.36
128.12
127.87
127.72
127.64
127.39
127.19
99.07
80.94
77.48
77.16
76.84
74.48
73.21
71.74
71.59
67.45
65.45
56.01
44.40
9.77
I
O
BnO
BnO
BnO
OMe
15α
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)


I
O
BnO
OMe
BnO
BnO
15β
7.43 7.43 7.42 7.41 7.41 7.40 7.40 7.39 7.38 7.38 7.37 7.37 7.36 7.36 7.35 7.34 7.33 7.32 7.31 7.31 7.26
4.95 4.92 4.81 4.78 4.61 4.60 4.59 4.58 4.56 4.52 4.49 4.39 4.38 4.38
3.69 3.68 3.67 3.66 3.63 3.52 3.40 3.40 3.38 3.37
2.13 2.12 2.12 2.11 2.10 2.09 2.09 2.08 2.07
14.24
1.00 1.01 4.03 1.11 1.03
1.00 3.06 2.95 2.16
0.99
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)


138.97
138.24
137.67
128.59
128.51
128.32
128.05
128.01
127.96
127.80
127.75
127.59
100.19
81.38
77.48
77.16
76.84
75.14
73.44
72.24
71.95
71.66
66.03
56.56
46.50
8.60
I
O
BnO
OMe
BnO
BnO
15β
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)


9.77
9.76
7.38
7.37
7.34
7.32
7.32
7.31
7.31
7.29
7.28
7.26
7.24
5.86
5.85
5.84
5.82
5.80
5.80
5.78
5.39
5.36
5.32
4.62
4.61
4.51
4.42
4.37
4.34
3.97
3.96
3.72
3.70
3.67
2.94
2.93
2.93
2.92
2.92
2.91
1.57
BnO
BnO
O
OBn
16
0.90
15.61
1.00
2.02
1.95
1.16
1.82
1.33
2.01
1.09
1.03
0.94
11.5
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)


202.29
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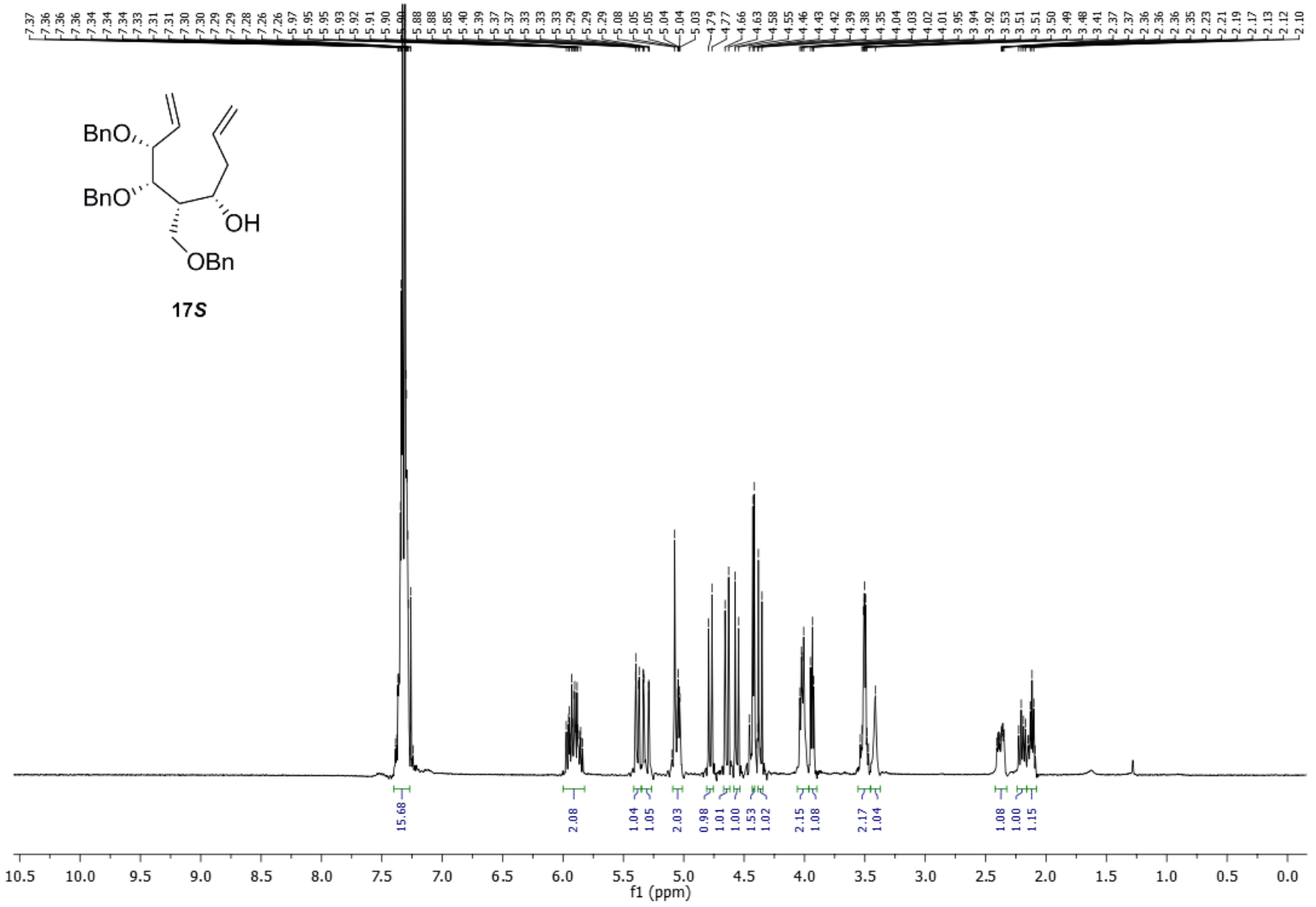
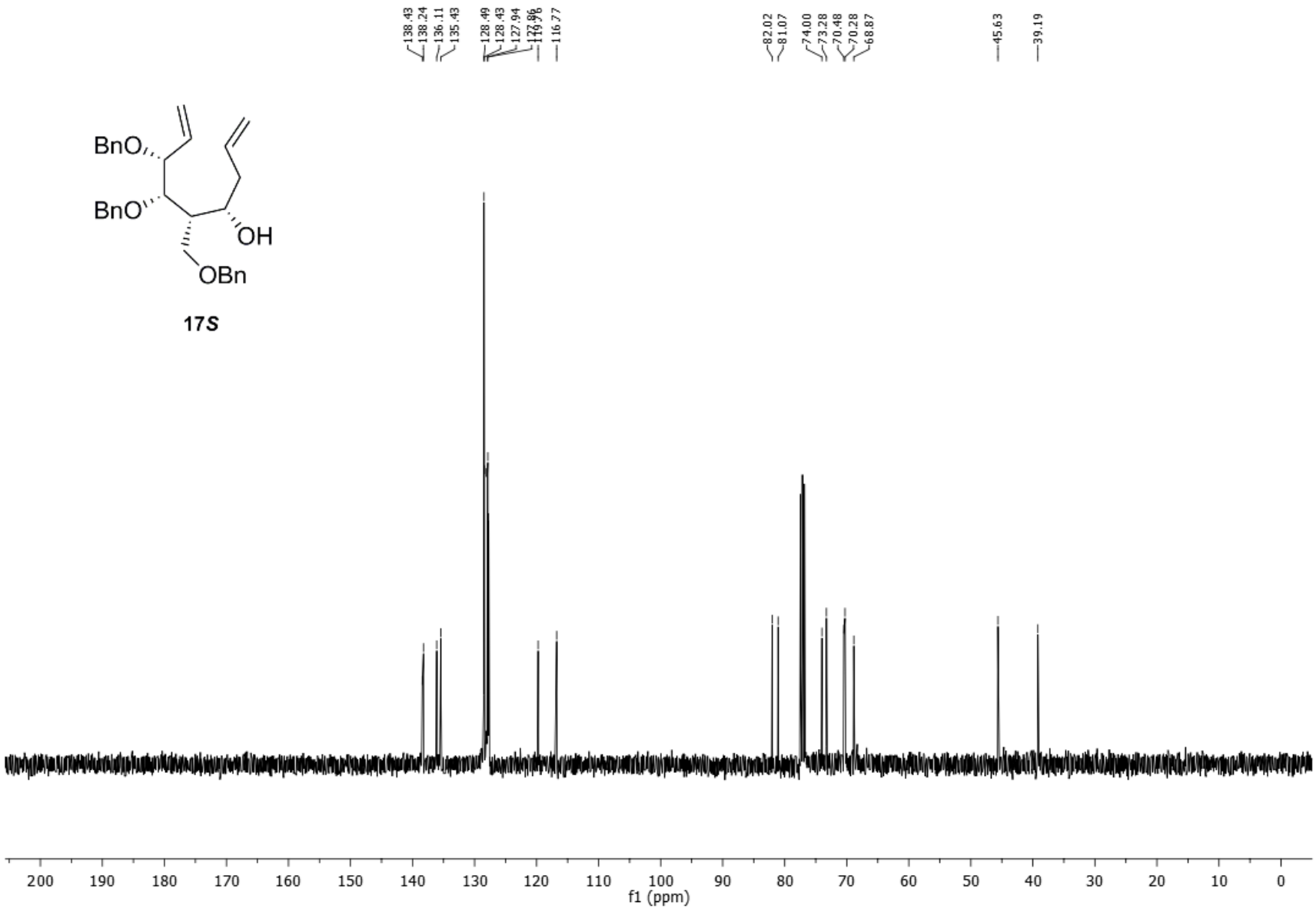
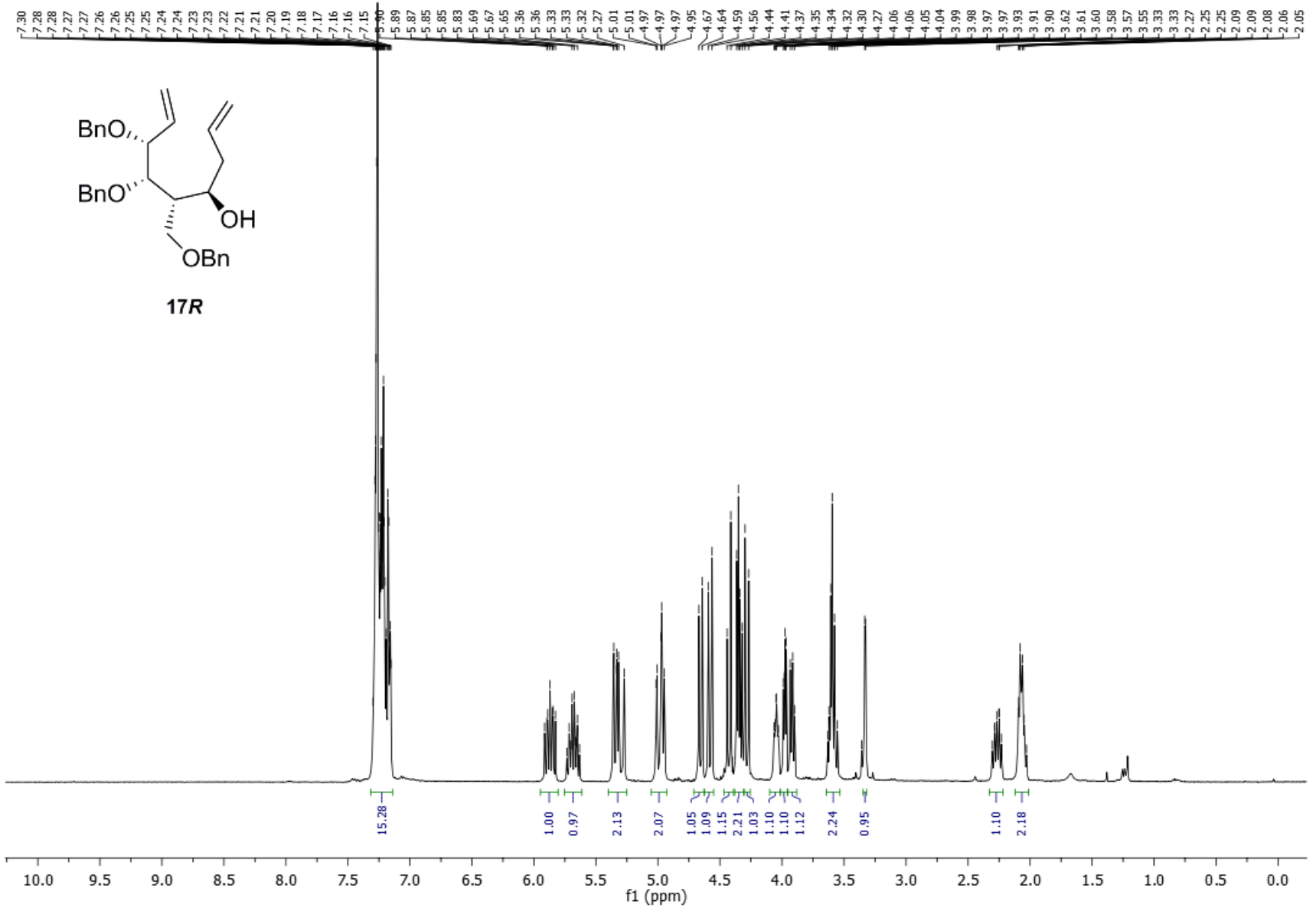
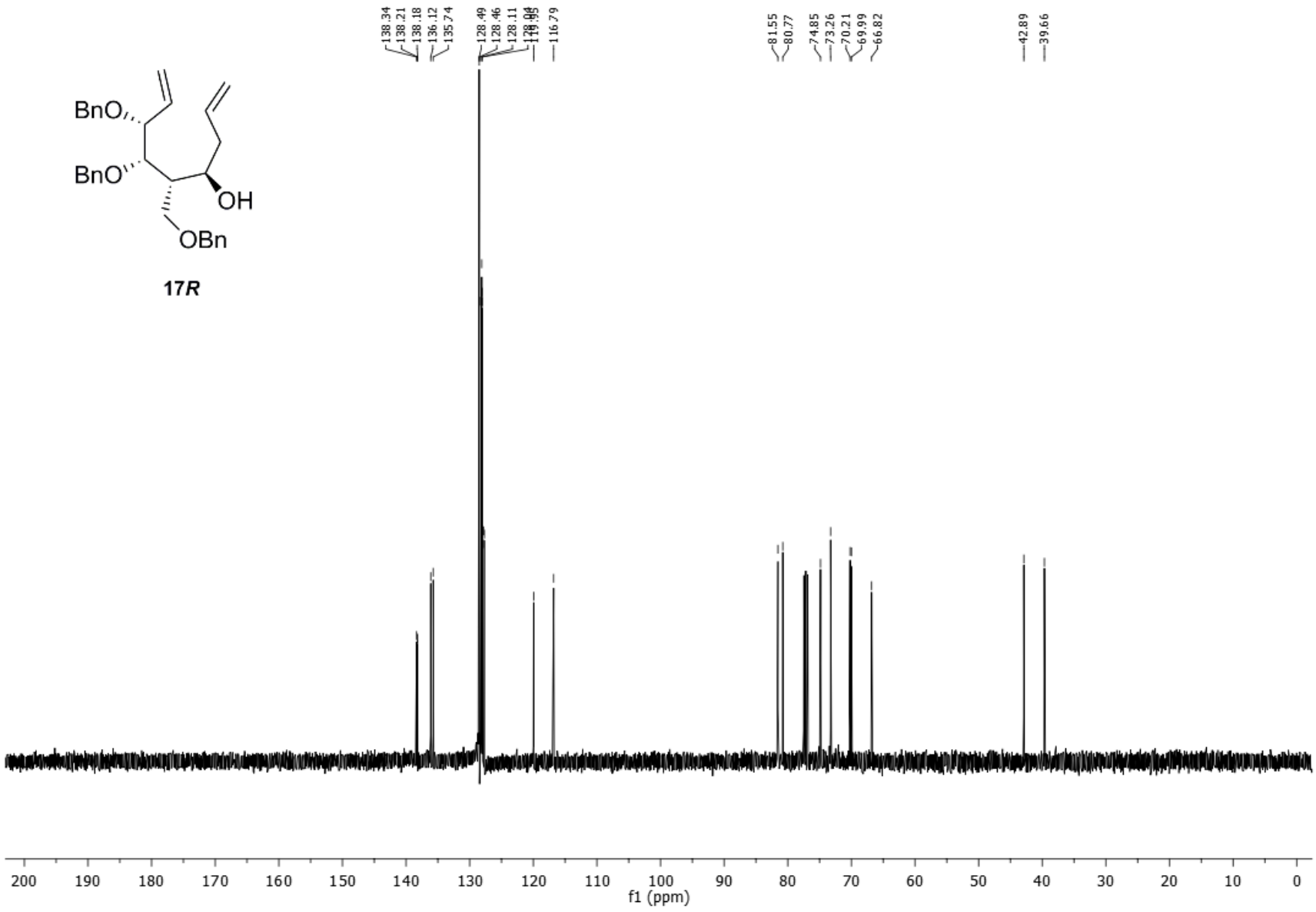
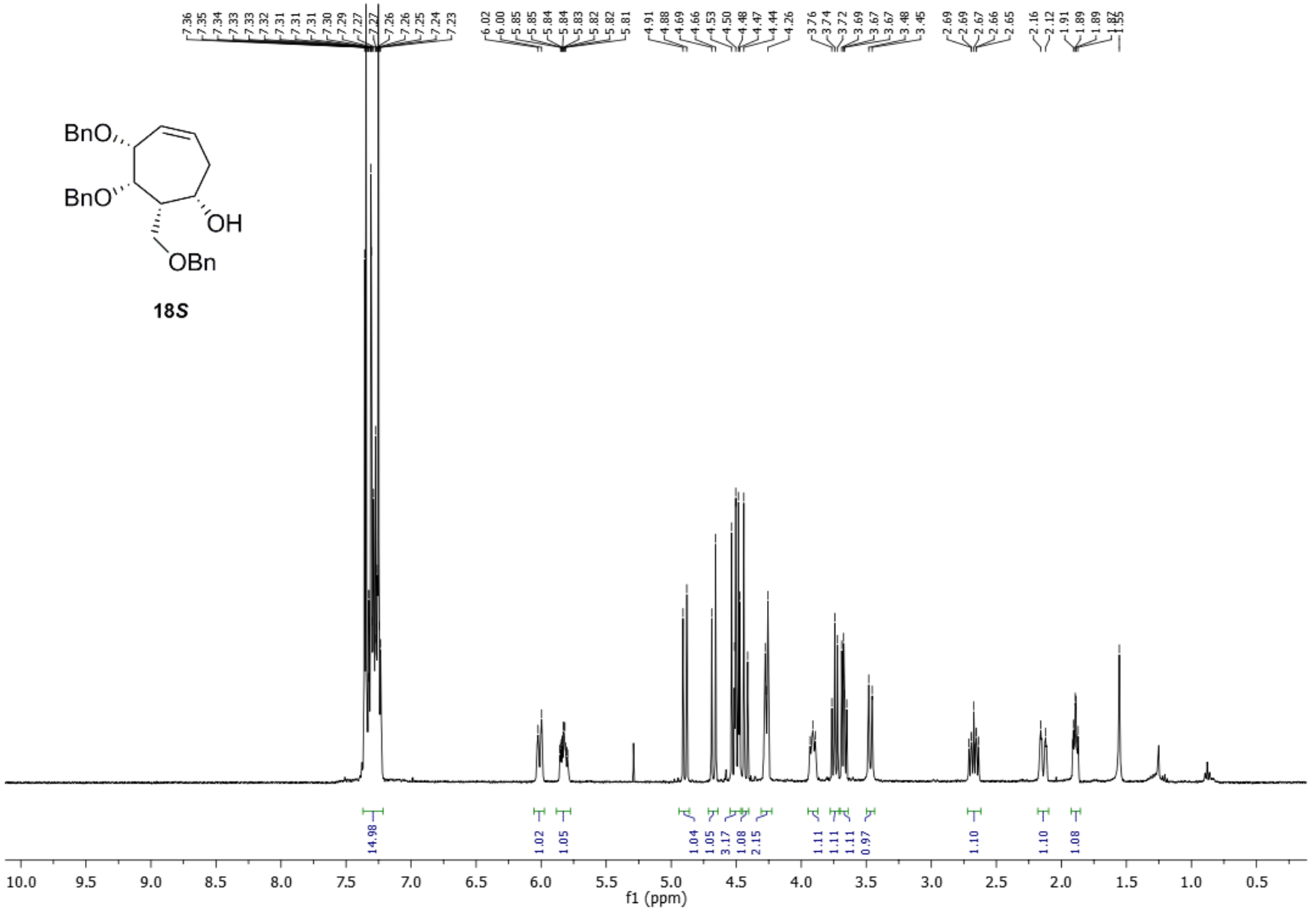
138.00
137.99
135.42
128.46
127.92
119.89
80.33
80.00
77.48
77.16
76.84
73.36
73.25
70.63
66.28
53.02
BnO
BnO
O
OBn
16
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

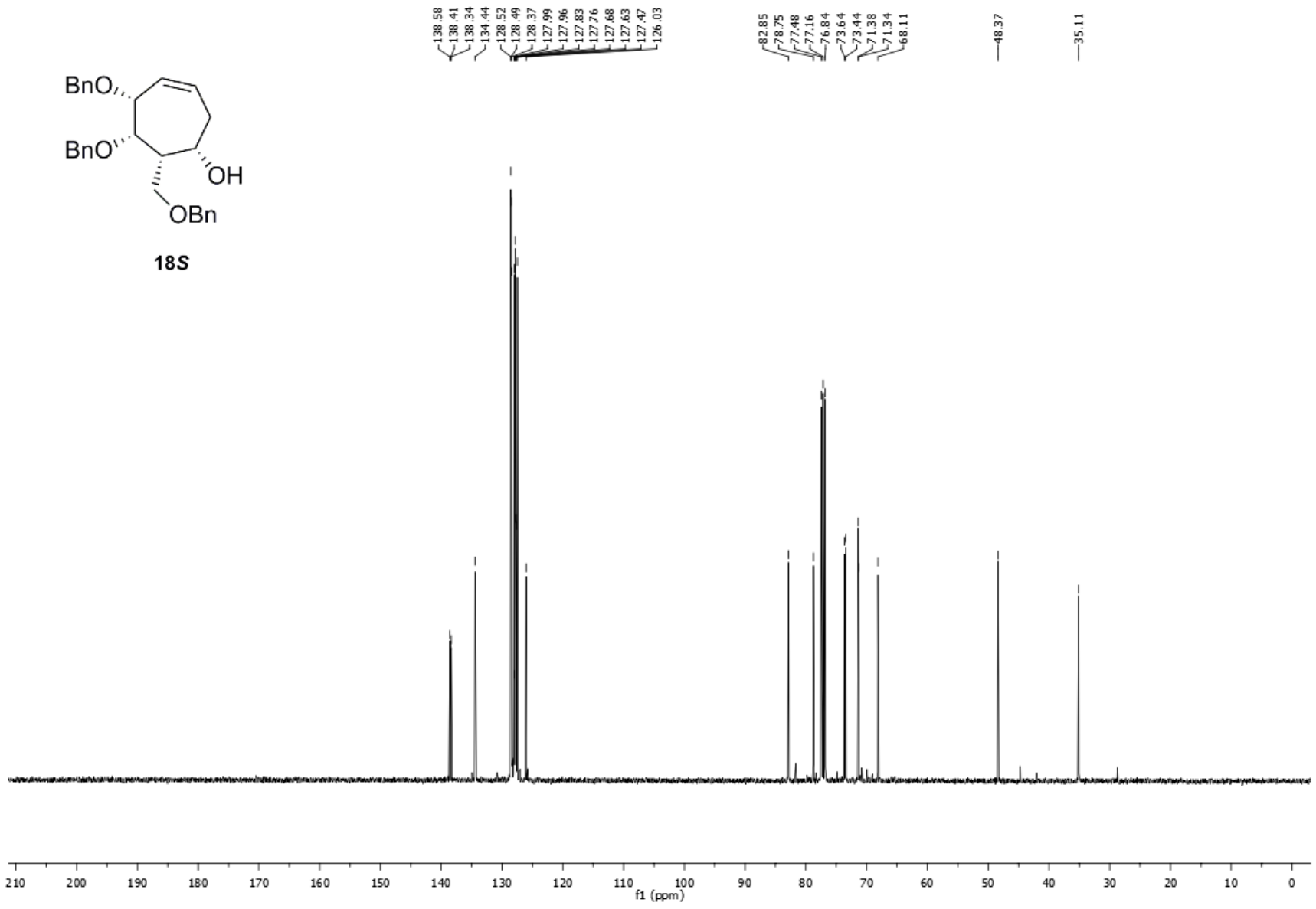

BnO
BnO
OH
OBn
17S
f1 (ppm)

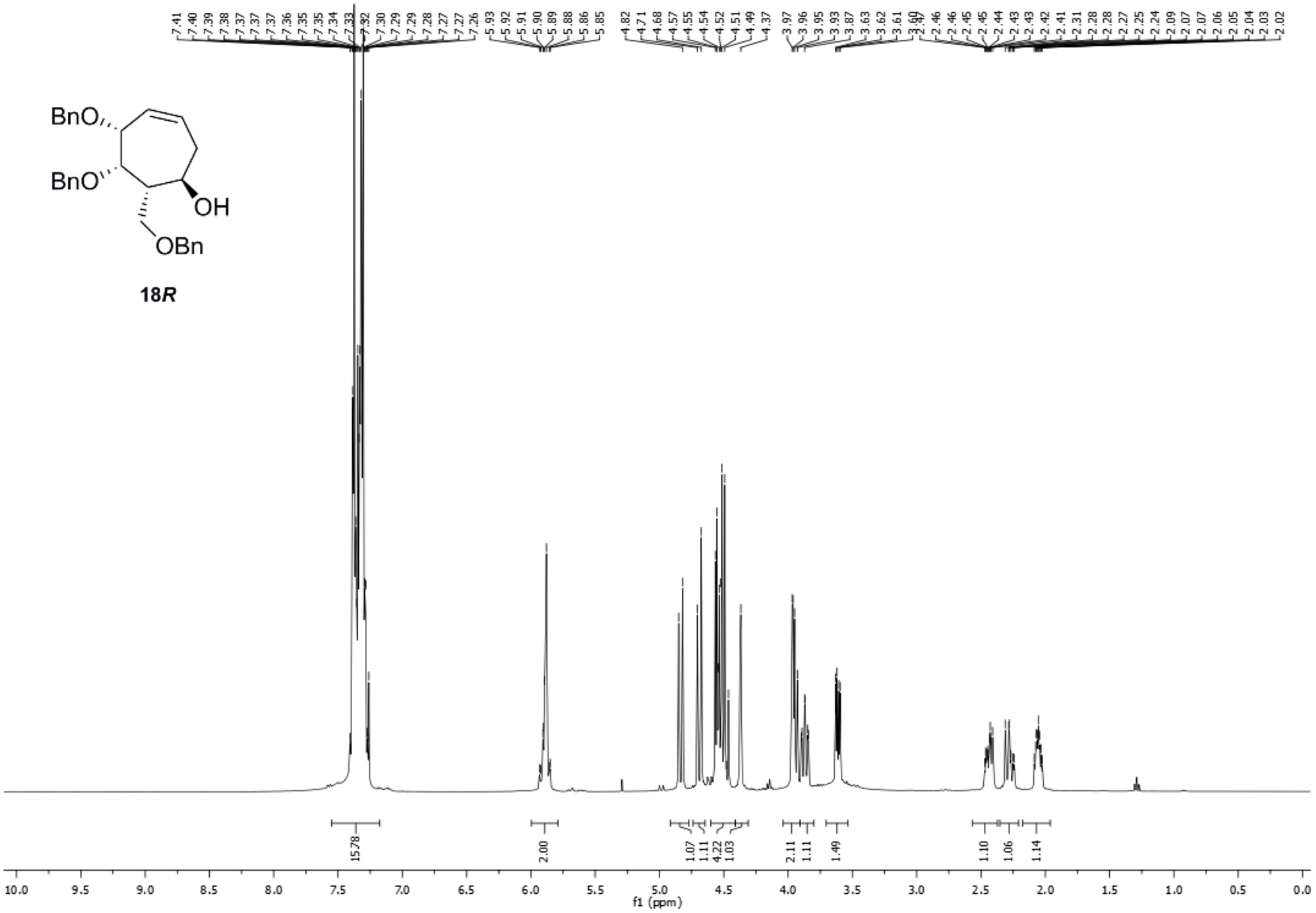

138.43
138.24
136.11
135.43
128.49
128.43
127.94
116.77
82.02
81.07
74.00
73.28
70.48
70.28
68.87
45.63
39.19
BnO
BnO
OH
OBn
17S
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

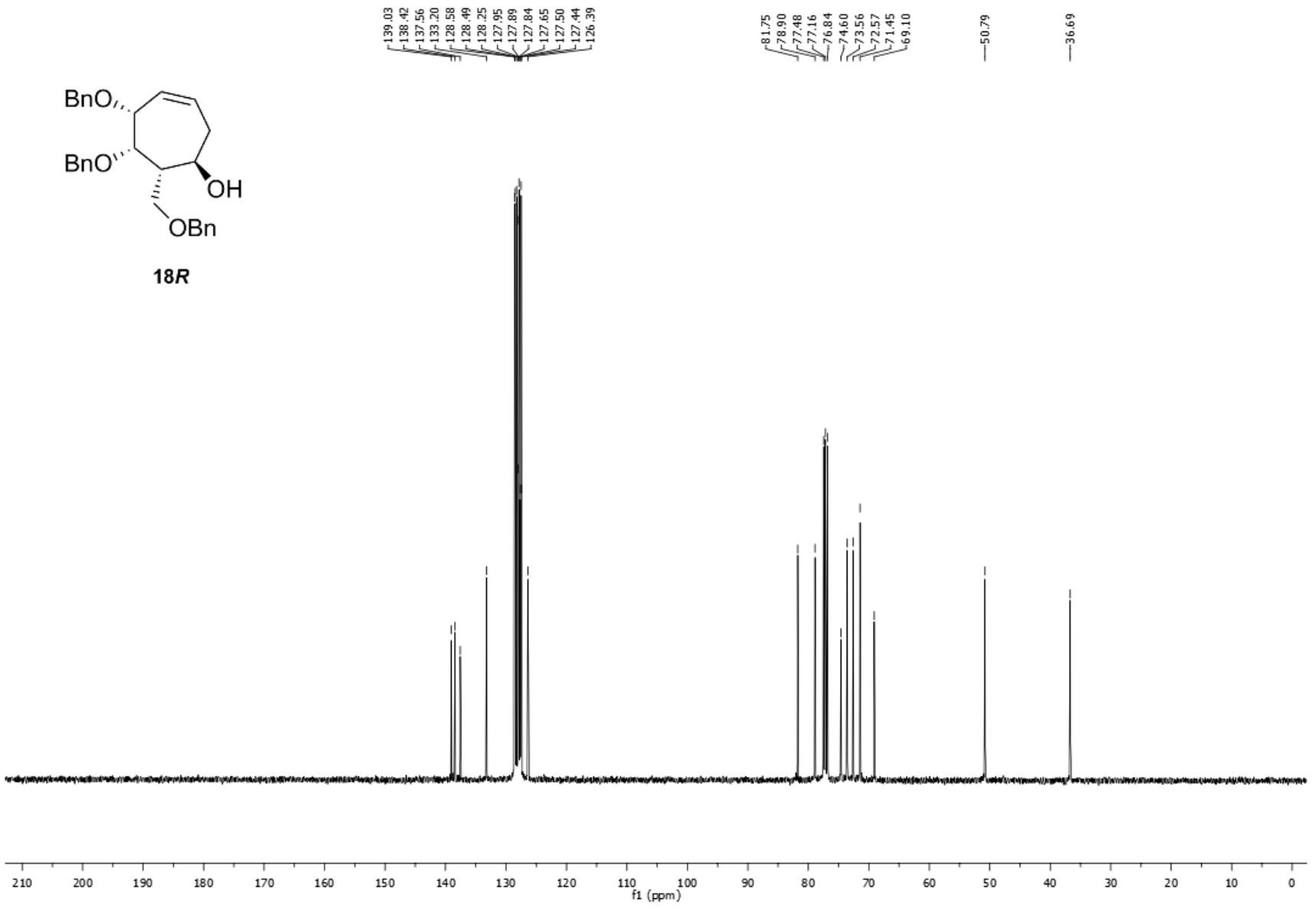

BnO
BnO
OH
OBn
17R
15.28
1.00
0.97
2.13
2.07
1.05
1.09
1.15
2.21
1.03
1.10
1.10
1.12
2.24
0.95
1.10
2.18
f1 (ppm)

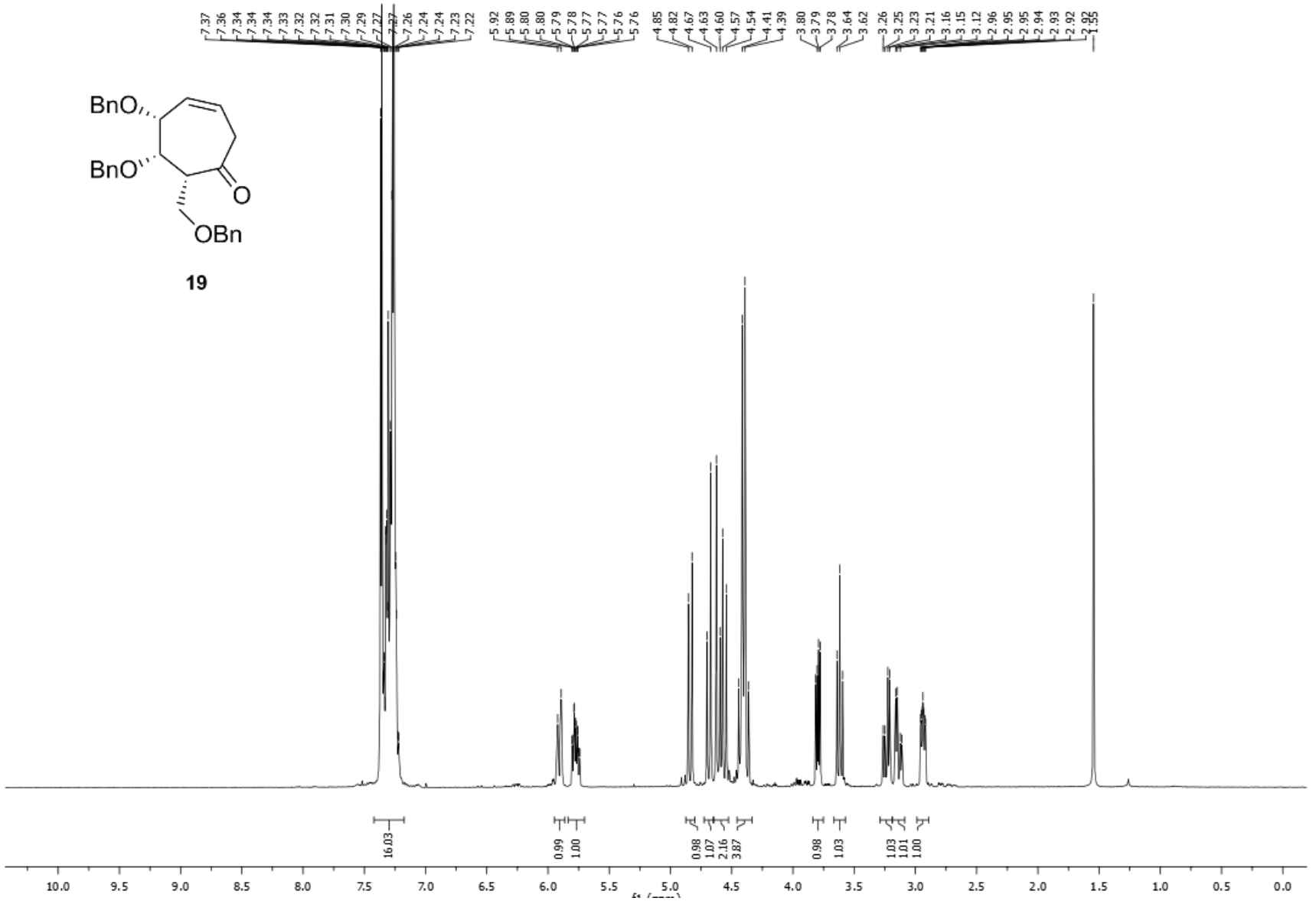

BnO
BnO
OH
OBn
17R
138.34
138.21
138.18
136.12
135.74
128.49
128.46
128.11
116.79
81.55
80.77
74.85
73.26
70.21
69.99
66.82
42.89
39.66
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

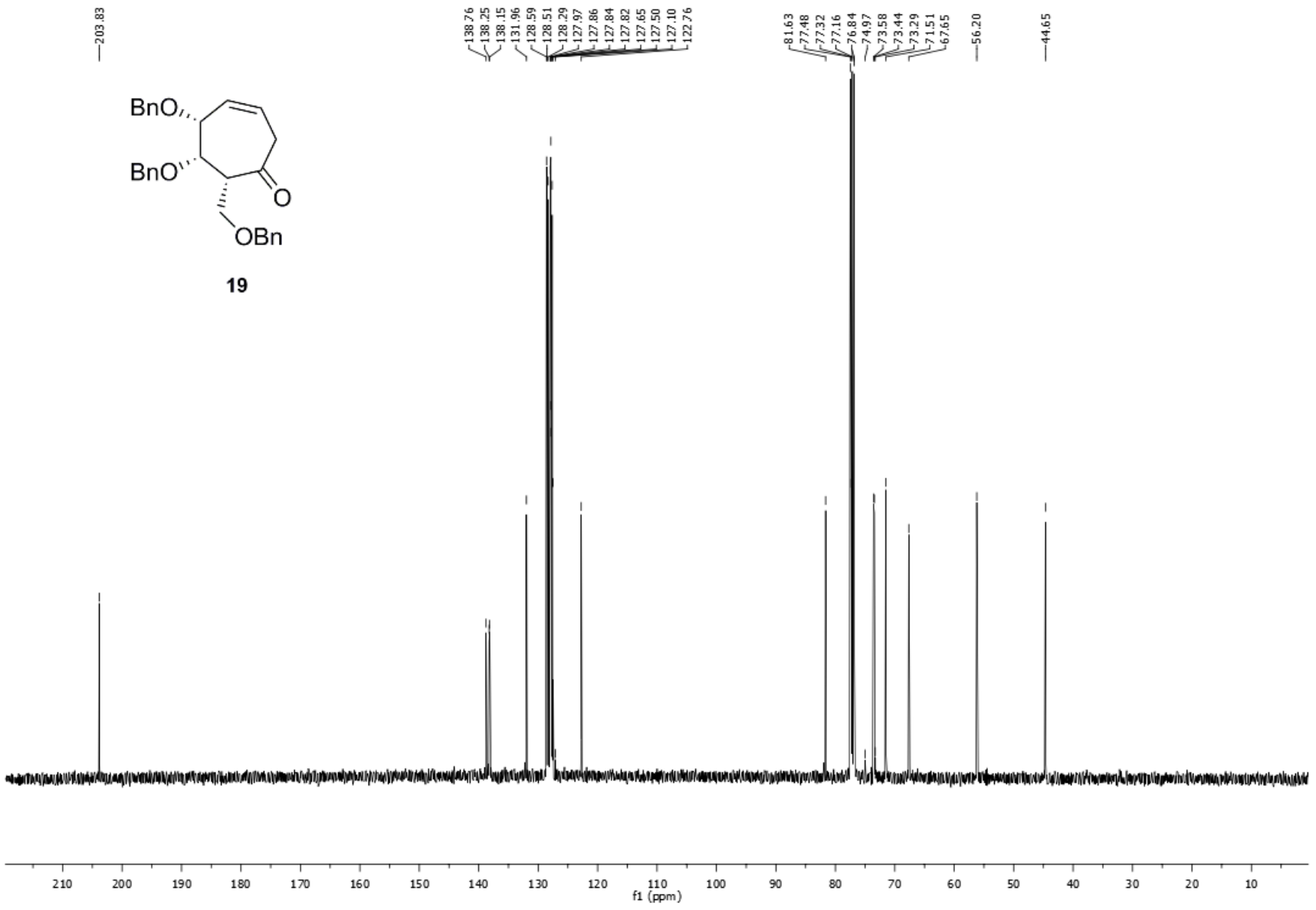

BnO
BnO
OH
OBn
18S
f1 (ppm)

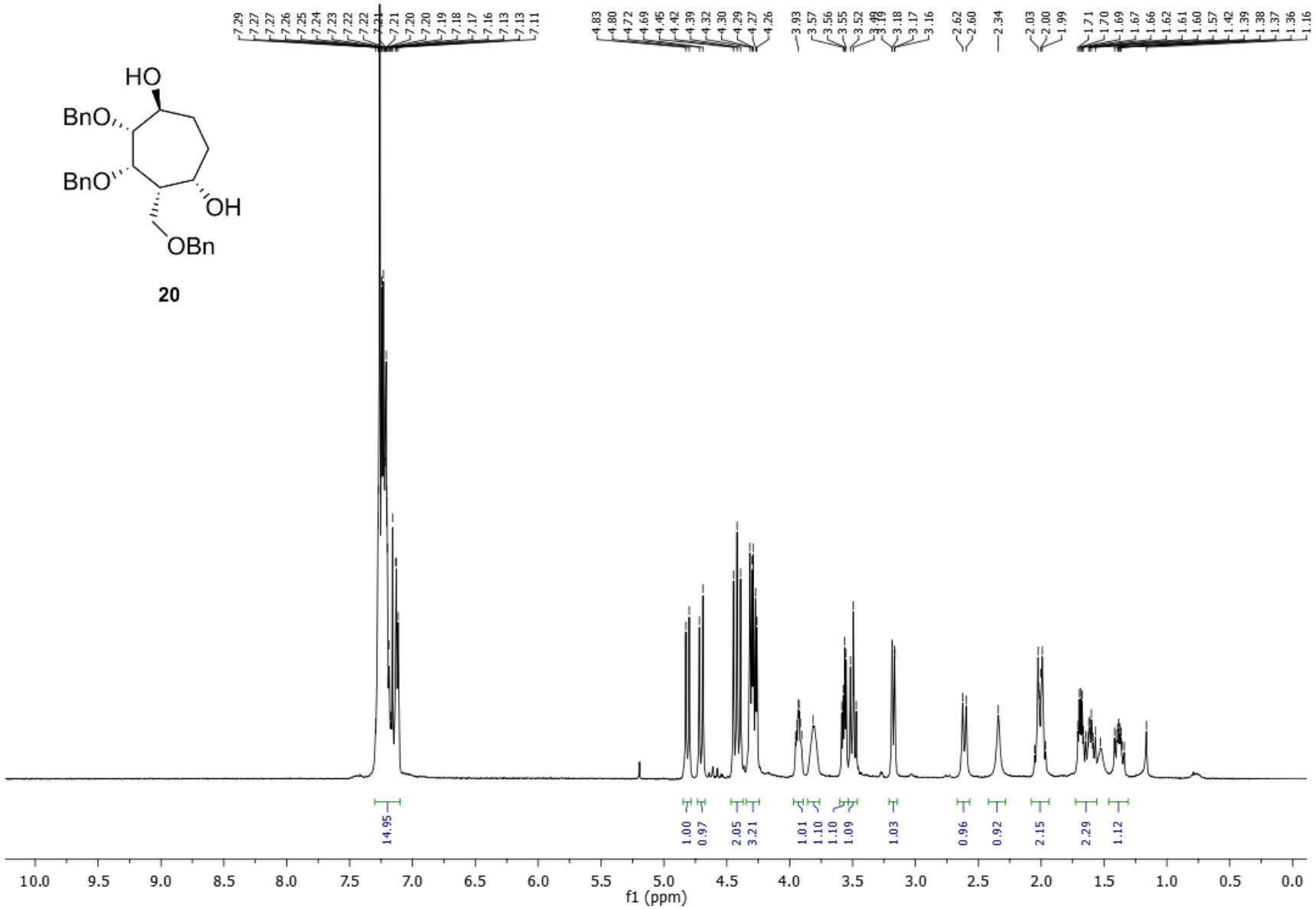

BnO
BnO
OH
OBn
18S
138.58
138.41
138.34
134.44
128.52
128.49
128.37
127.99
127.96
127.83
127.76
127.68
127.63
127.47
126.03
82.85
78.75
77.48
77.16
76.84
73.64
73.44
71.38
71.34
68.11
48.37
35.11
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

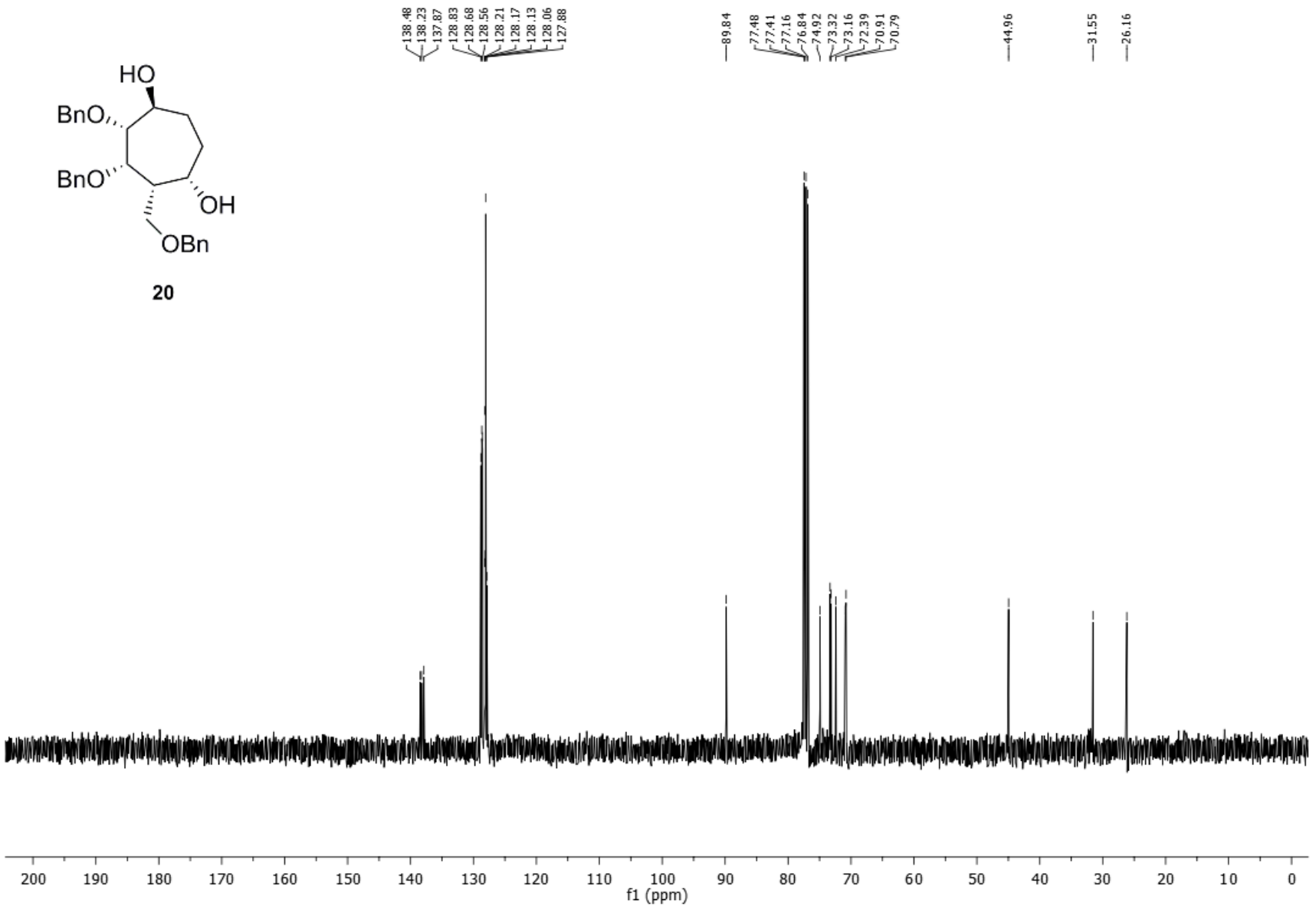

BnO
BnO
OH
OBn
18R
7.41 7.40 7.39 7.38 7.37 7.37 7.37 7.36 7.35 7.35 7.34 7.33 7.32 7.30 7.29 7.29 7.28 7.27 7.27 7.26
5.93 5.92 5.91 5.90 5.89 5.88 5.86 5.85
4.82 4.71 4.68 4.57 4.55 4.54 4.52 4.51 4.49 4.37
3.97 3.96 3.95 3.93 3.87 3.63 3.62 3.61 3.60
2.46 2.46 2.45 2.45 2.44 2.43 2.43 2.42 2.41 2.31 2.28 2.28 2.27 2.25 2.24 2.09 2.07 2.07 2.06 2.05 2.04 2.03 2.02
15.78
2.00
1.07 1.11 4.22 1.03
2.11 1.11
1.49
1.10 1.06 1.14
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)

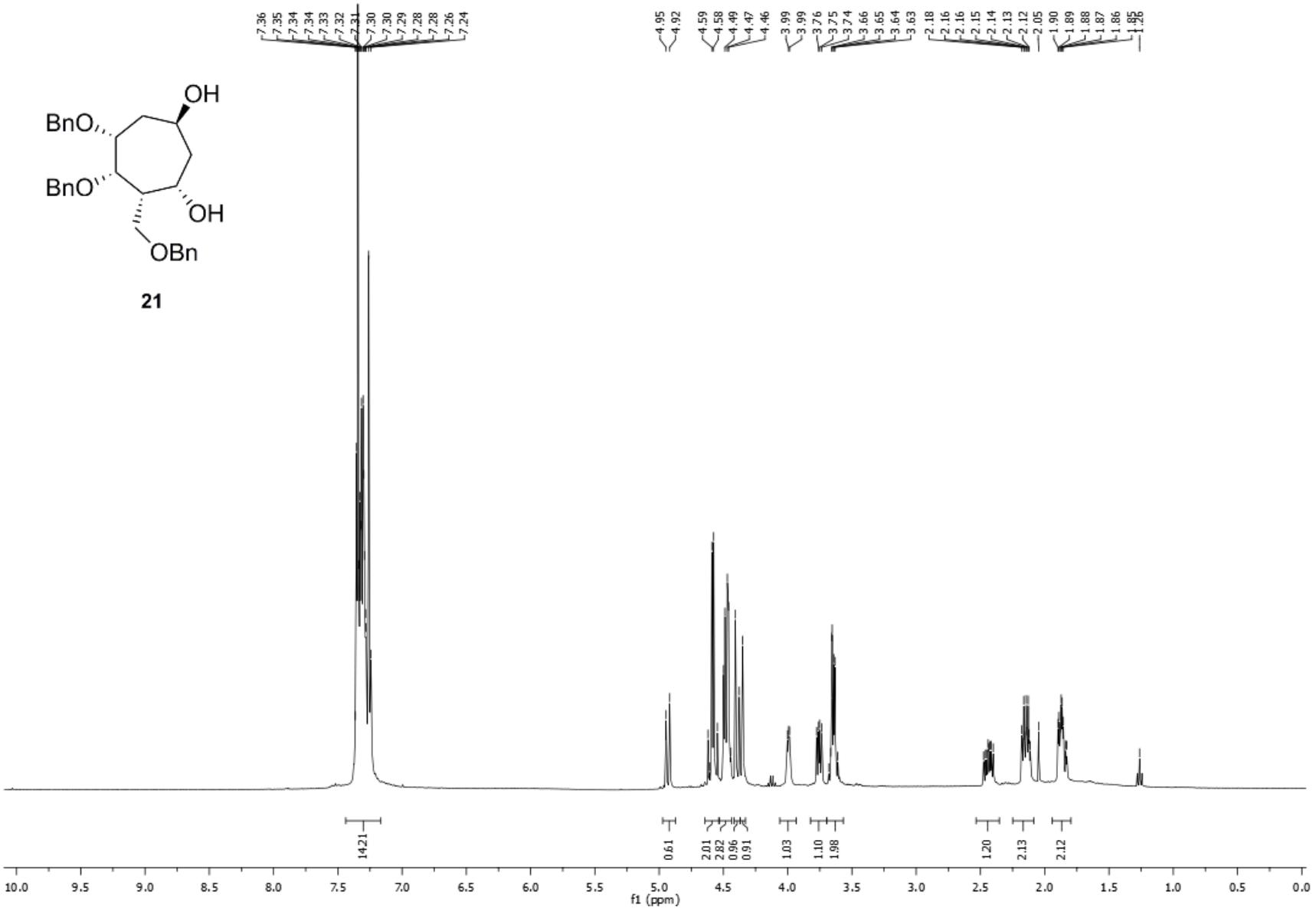

BnO
BnO
OH
OBn
18R
139.03
138.42
137.56
133.20
128.58
128.49
128.25
127.95
127.89
127.84
127.65
127.50
127.44
126.39
81.75
78.90
77.48
77.16
76.84
74.60
73.56
72.57
71.45
69.10
50.79
36.69
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

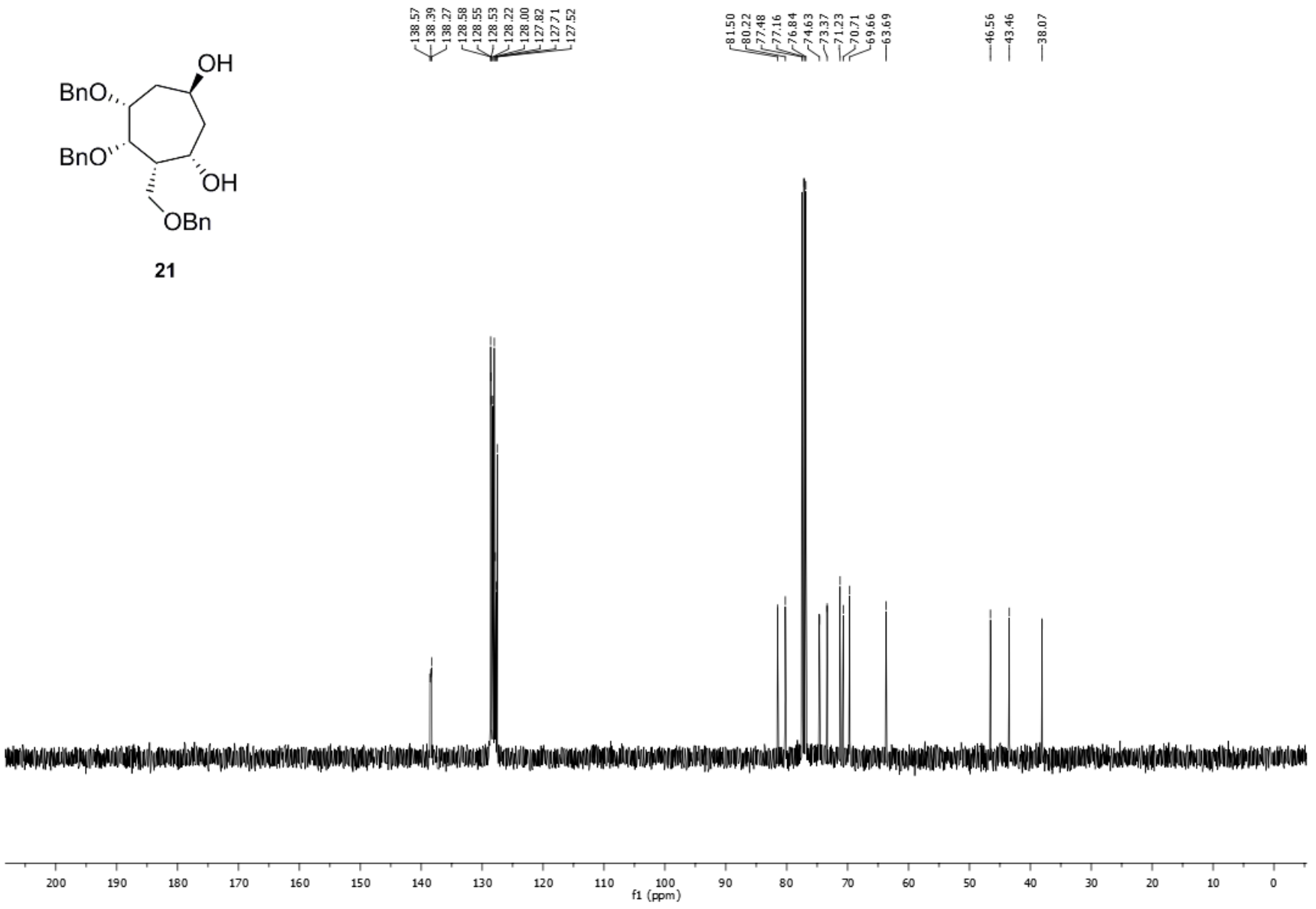

BnO
BnO
O
OBn
19
7.37 7.36 7.34 7.34 7.34 7.33 7.32 7.32 7.31 7.30 7.29 7.27 7.27 7.26 7.24 7.24 7.23 7.22
5.92 5.89 5.80 5.80 5.79 5.78 5.77 5.77 5.76 5.76
4.85 4.82 4.67 4.63 4.60 4.57 4.54 4.41 4.39
3.80 3.79 3.78 3.64 3.62
3.26 3.25 3.23 3.21 3.16 3.15 3.12 2.96 2.95 2.95 2.94 2.93 2.92 2.92 1.55
16.03
0.99 1.00
0.98 1.07 2.16 3.87
0.98 1.03
1.03 1.01 1.00
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

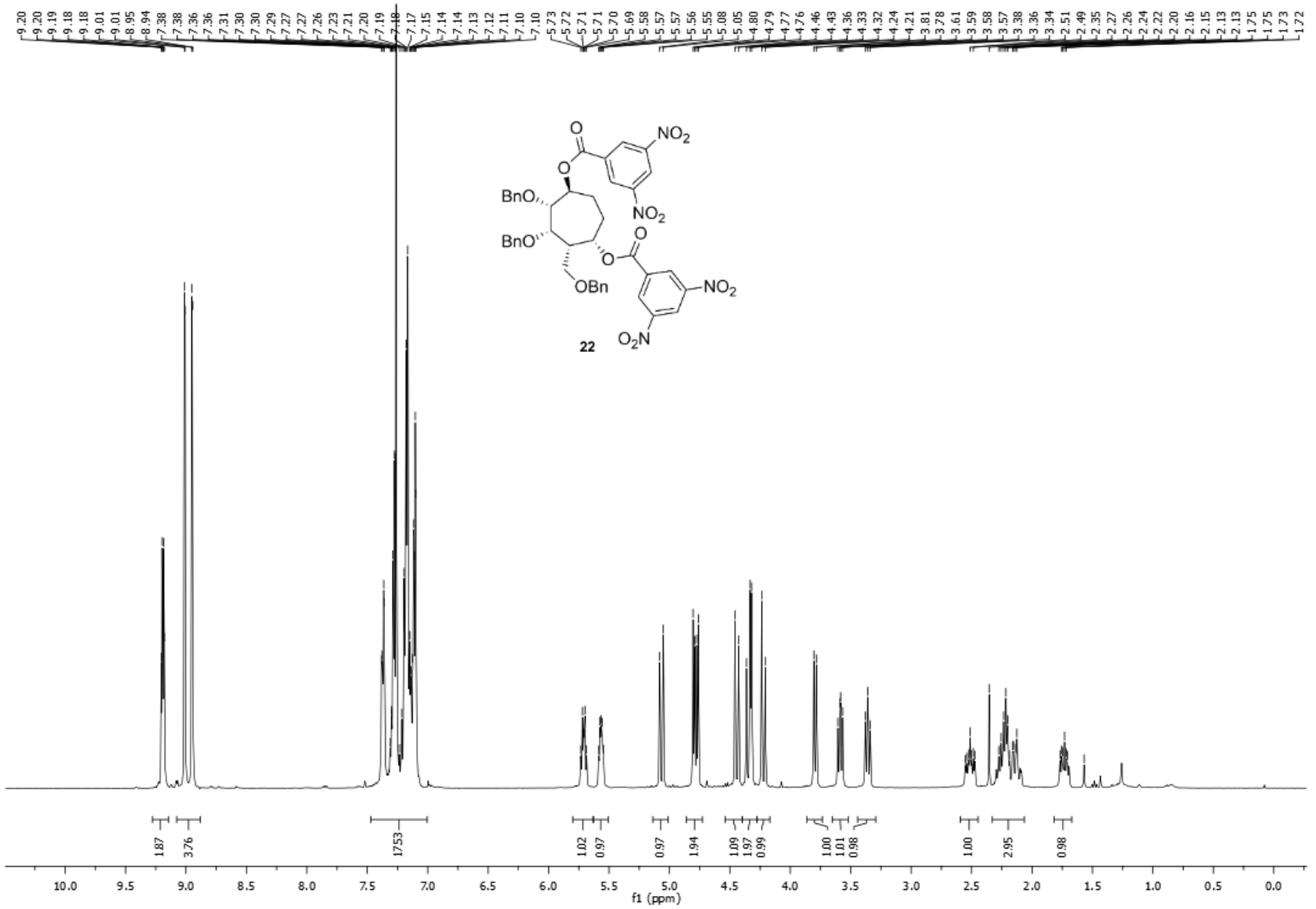

203.83
138.76
138.25
138.15
131.96
128.59
128.51
128.29
127.97
127.86
127.84
127.82
127.65
127.50
127.10
122.76
81.63
77.48
77.32
77.16
76.84
74.97
73.58
73.44
73.29
71.51
67.65
56.20
44.65
BnO
BnO
O
OBn
19
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
f1 (ppm)

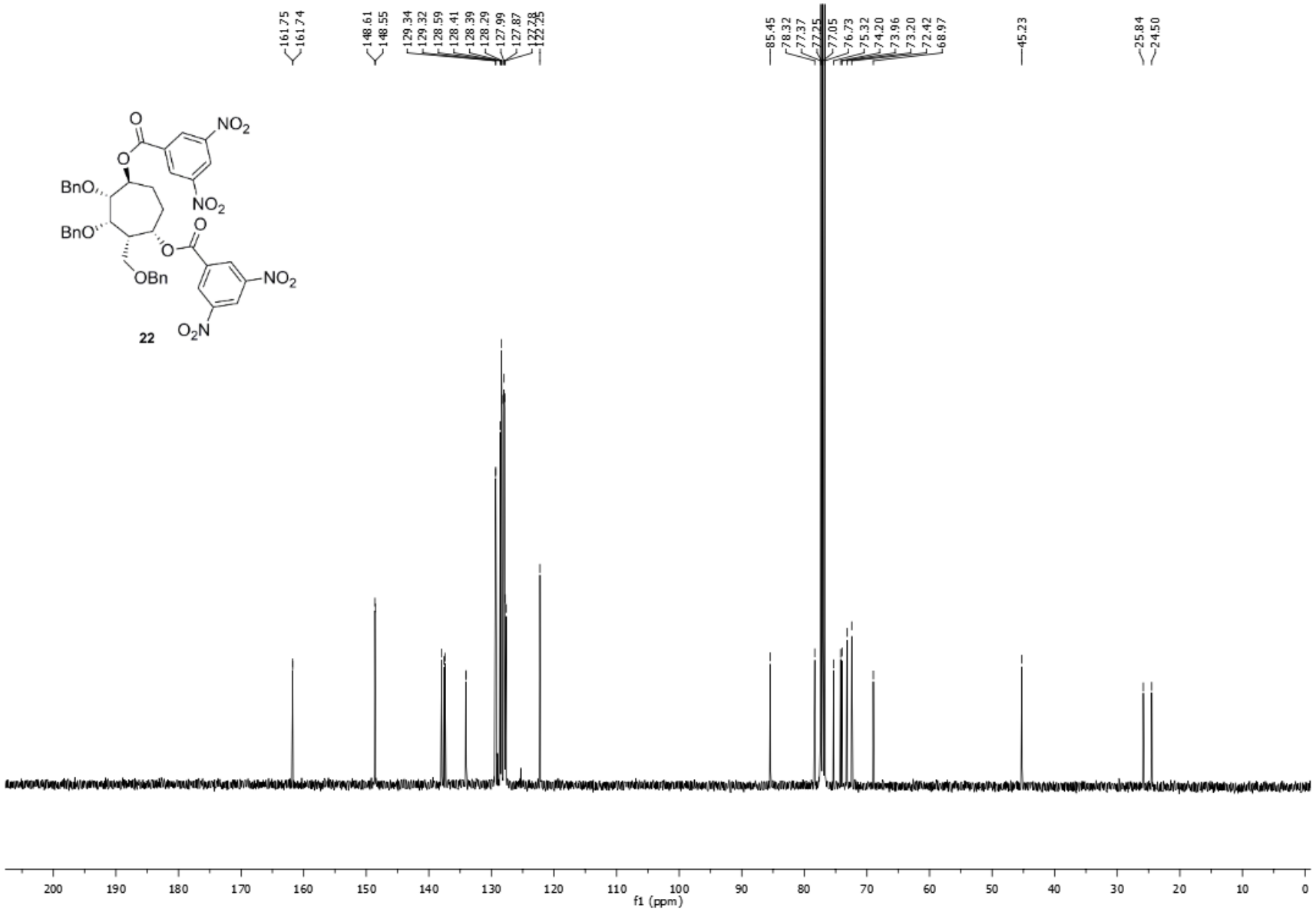

HO
BnO
BnO
OH
OBn
20
f1 (ppm)

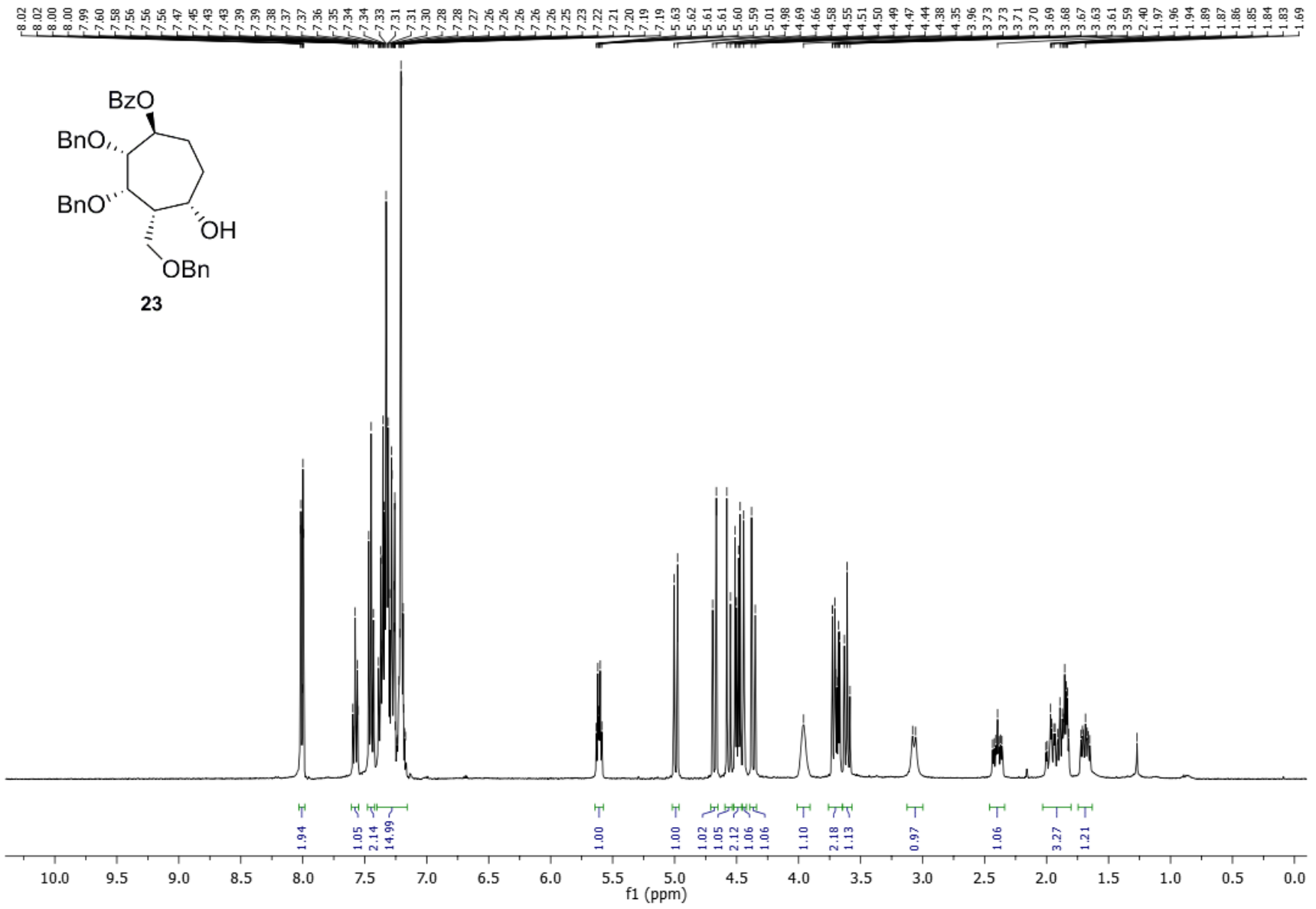

HO
BnO
BnO
OH
OBn
20
138.48
138.23
137.87
128.83
128.68
128.56
128.21
128.17
128.13
128.06
127.88
89.84
77.48
77.41
77.16
76.84
74.92
73.32
73.16
72.39
70.91
70.79
44.96
31.55
26.16
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

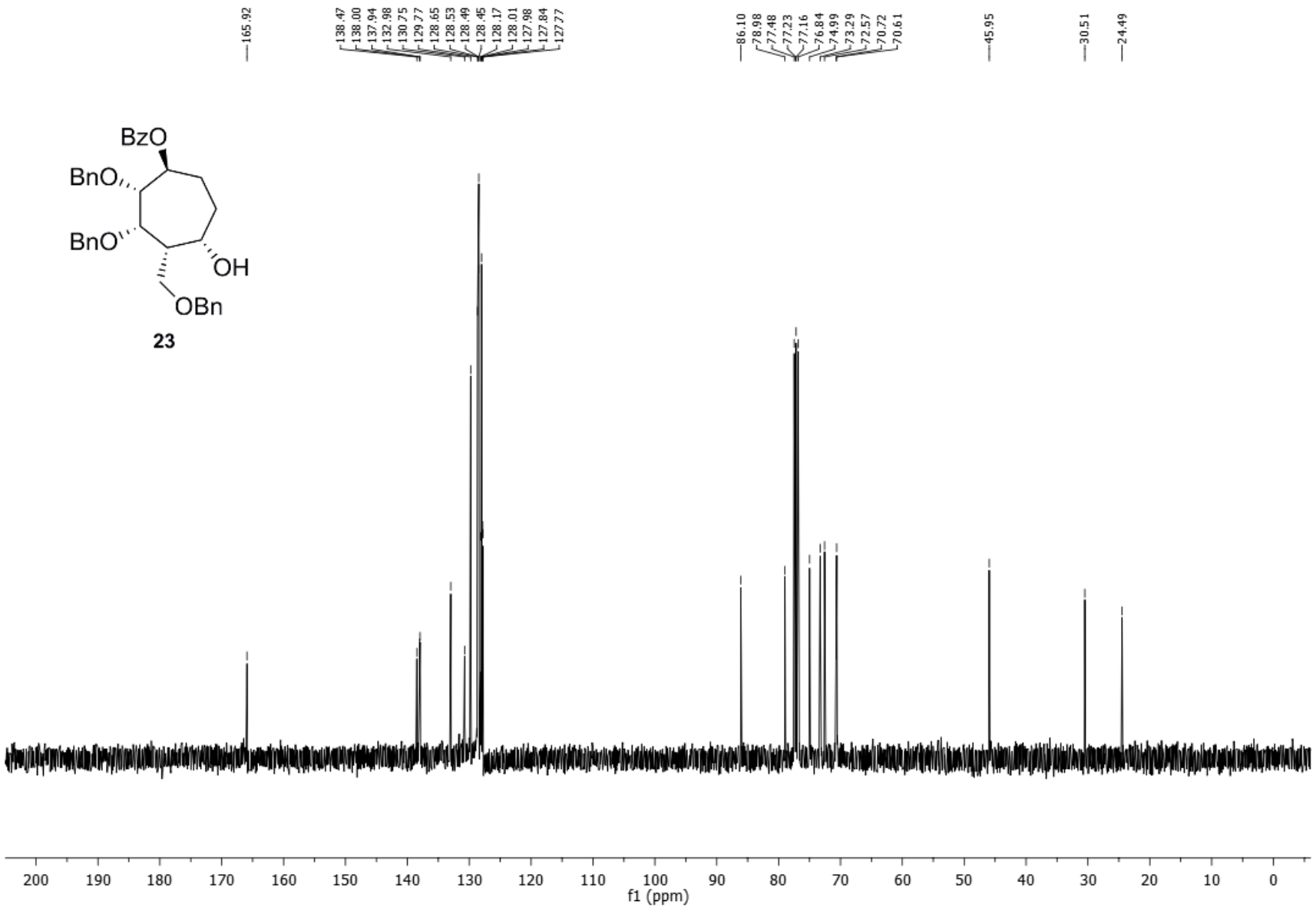

OH
BnO
BnO
OH
OBn
21
7.36
7.35
7.34
7.34
7.33
7.32
7.31
7.30
7.30
7.29
7.28
7.28
7.26
7.24
4.95
4.92
4.59
4.58
4.49
4.47
4.46
3.99
3.99
3.76
3.75
3.74
3.66
3.65
3.64
3.63
2.18
2.16
2.16
2.15
2.14
2.13
2.12
2.05
1.90
1.89
1.88
1.87
1.86
1.85
1.85
14.21
0.61
2.01
2.82
0.96
0.91
1.03
1.10
1.98
1.20
2.13
2.12
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)

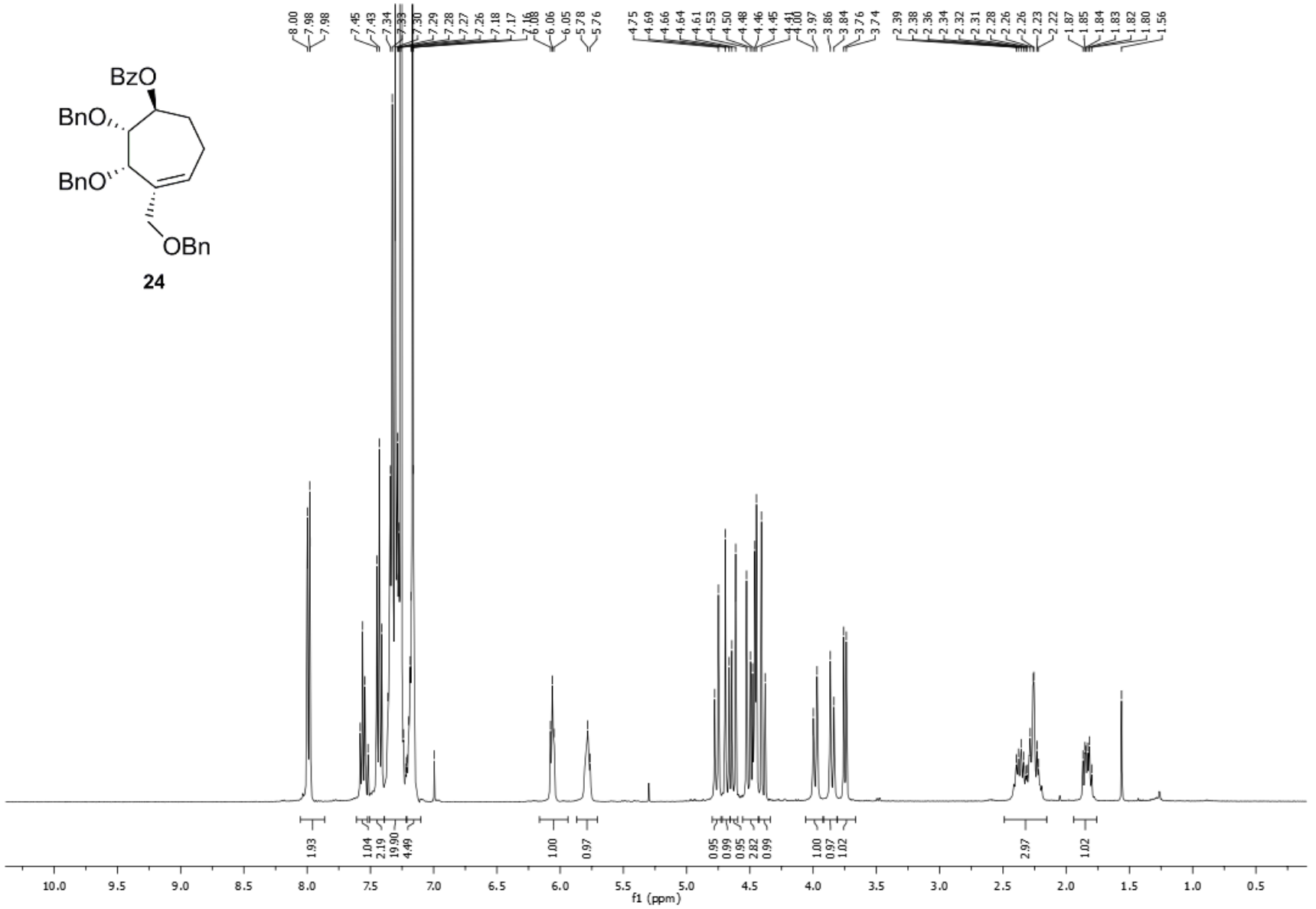

OH
BnO
BnO
OH
OBn
21
138.57
138.39
138.27
128.58
128.55
128.53
128.22
128.00
127.82
127.71
127.52
81.50
80.22
77.48
77.16
76.84
74.63
73.37
71.23
70.71
69.66
63.69
46.56
43.46
38.07
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

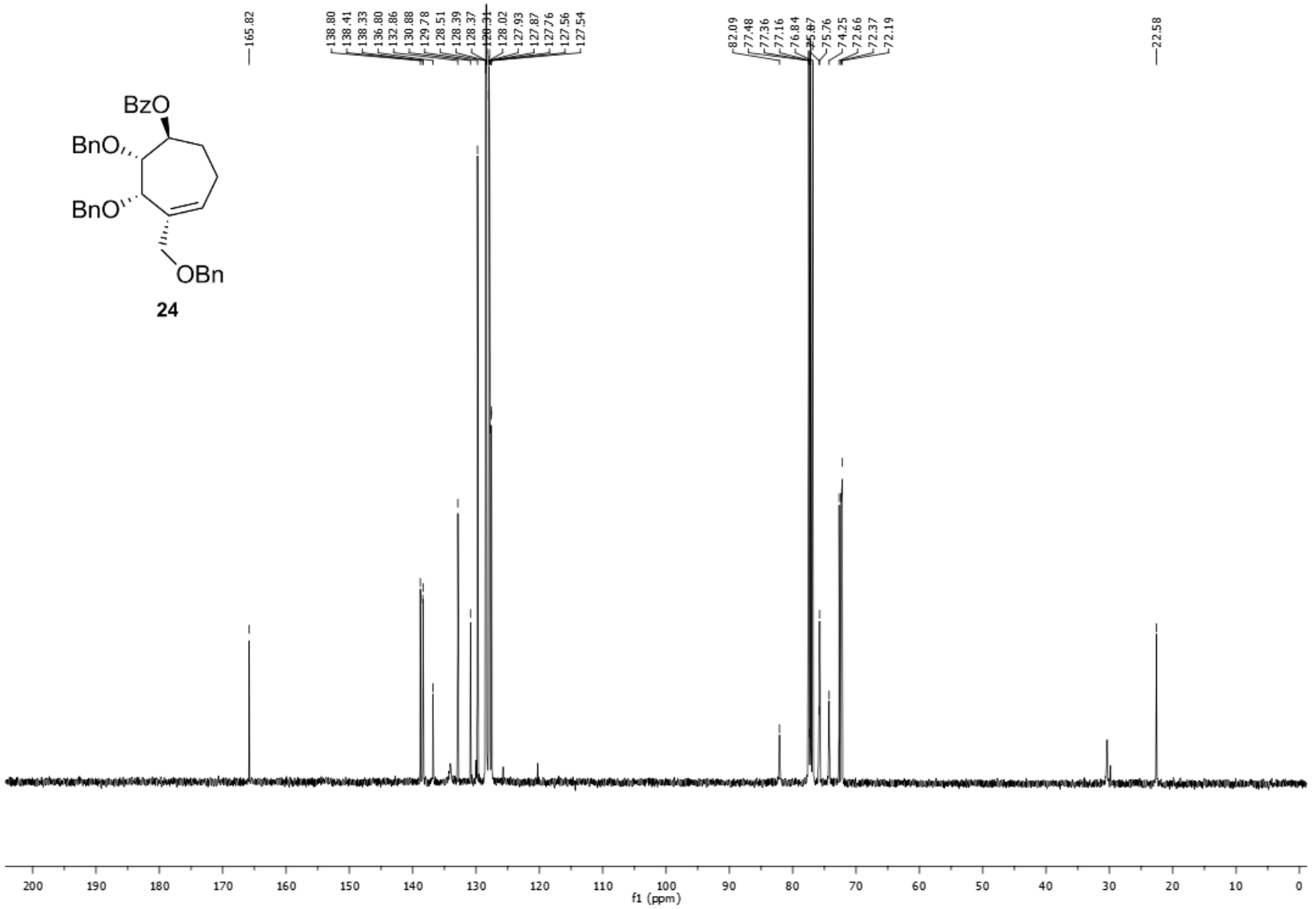

22
f1 (ppm)

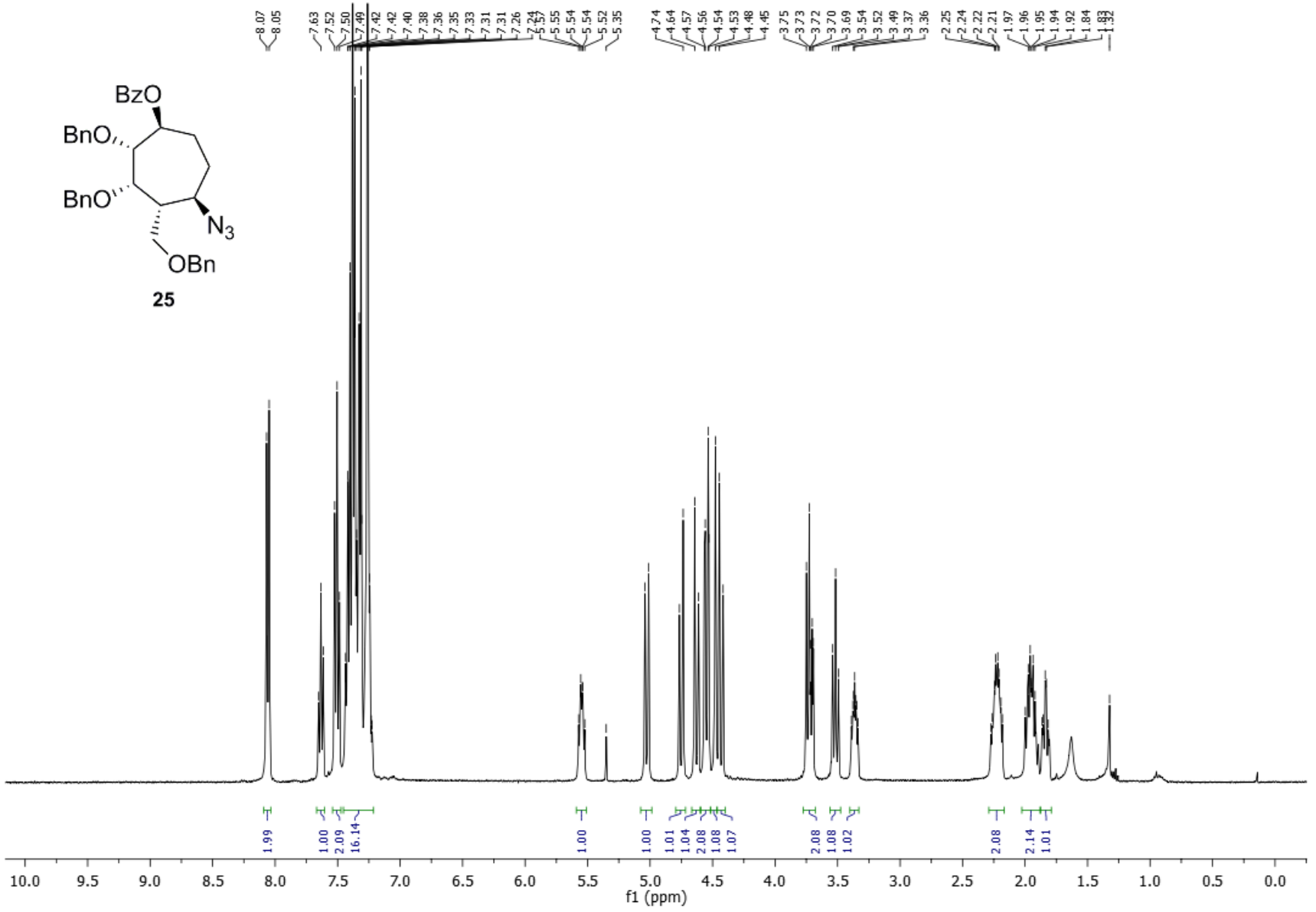

161.75
161.74
148.61
148.55
129.34
129.32
128.59
128.41
128.39
128.29
127.99
127.87
122.28
122.25
85.45
78.32
77.37
77.25
77.05
76.73
75.32
74.20
73.96
73.20
72.42
68.97
45.23
25.84
24.50
BnO
BnO
OBn
NO2
NO2
NO2
O2N
22
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

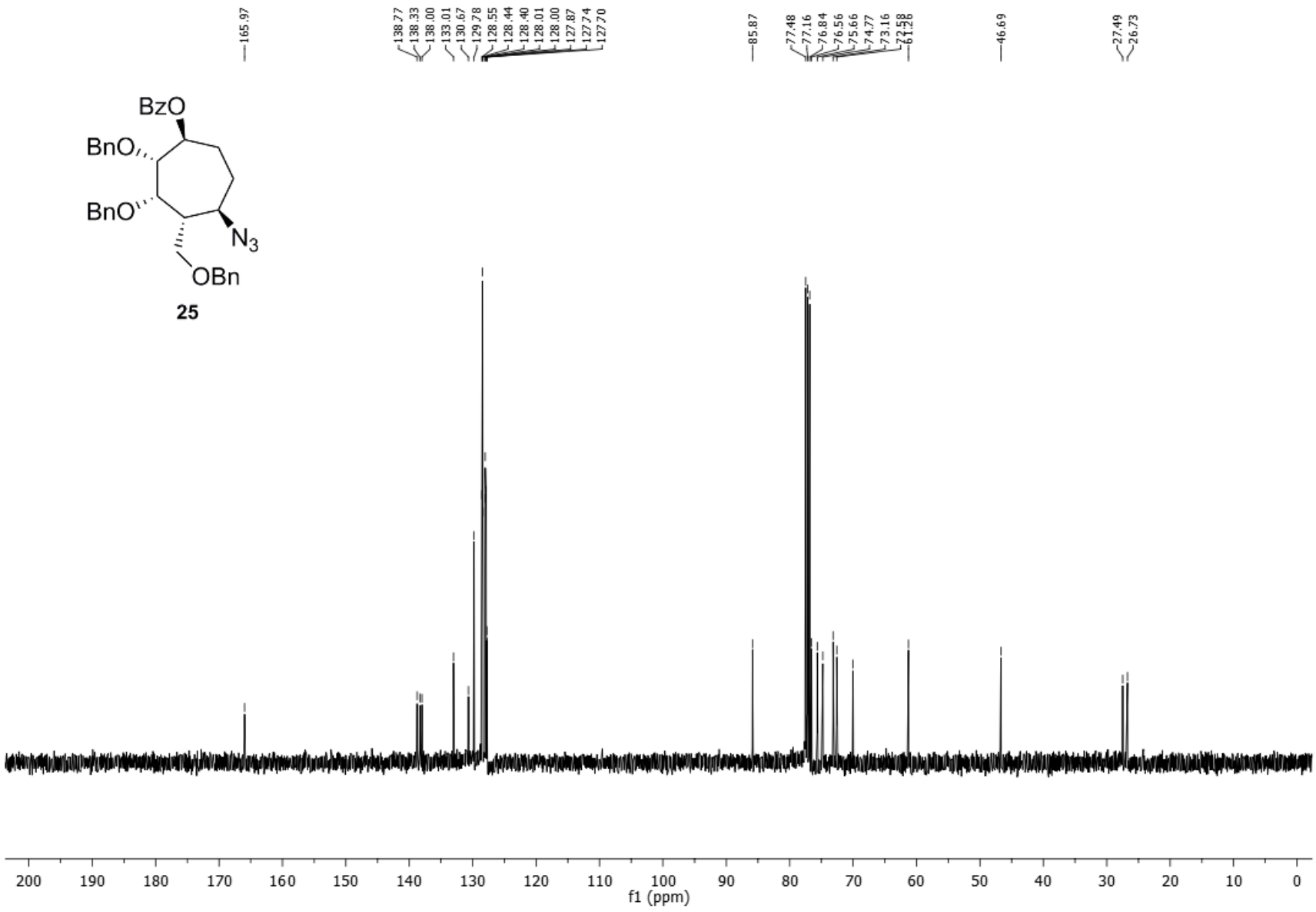

BzO
BnO
BnO
OH
OBn
23
f1 (ppm)

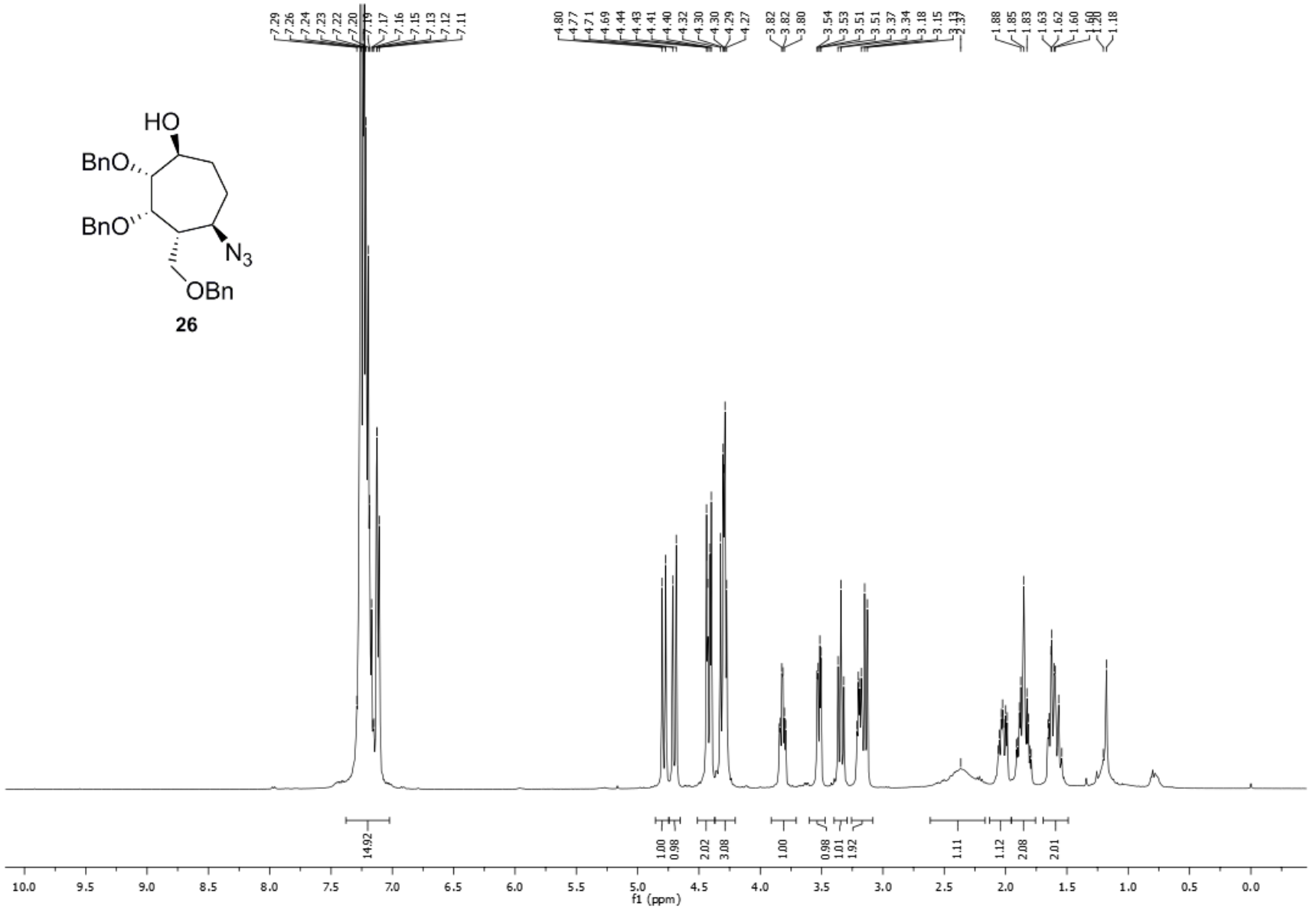

165.92
138.47
138.00
137.94
132.98
130.75
129.77
128.65
128.53
128.49
128.45
128.17
128.01
127.98
127.84
127.77
86.10
78.98
77.48
77.23
77.16
76.84
74.99
73.29
72.57
70.72
70.61
45.95
30.51
24.49
BzO
BnO
BnO
OH
OBn
23
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

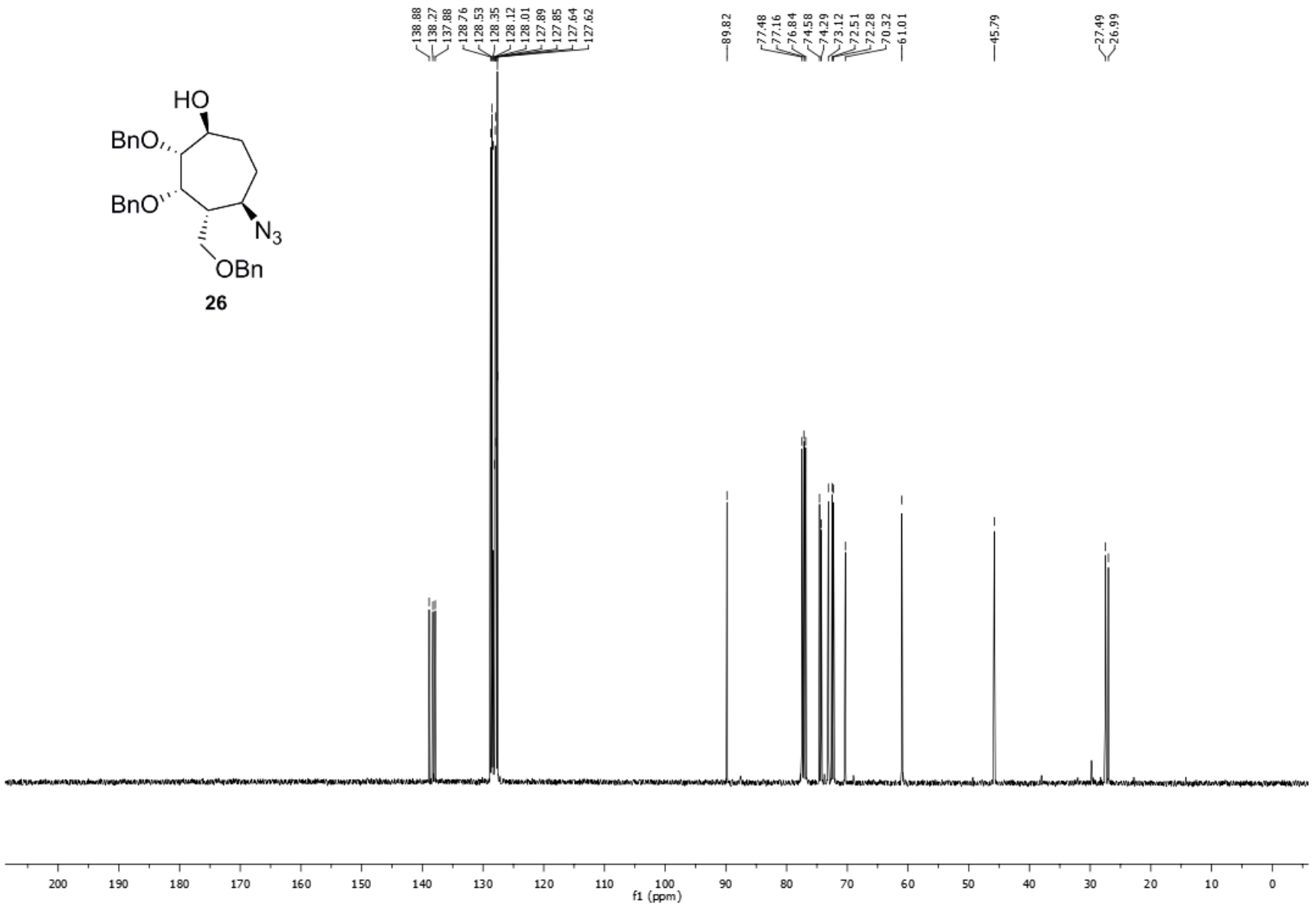

BzO
BnO
BnO
OBn
24
f1 (ppm)

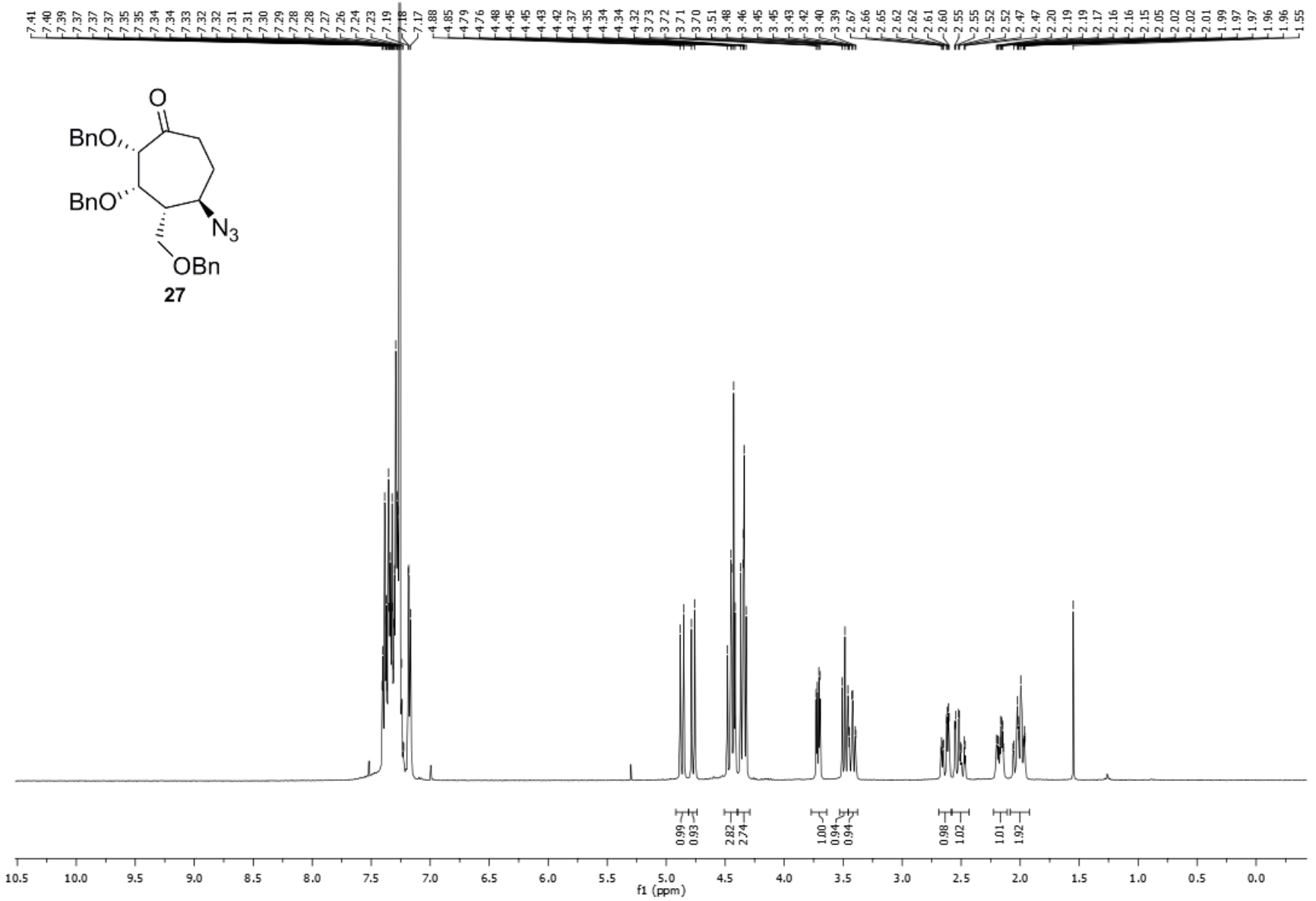

BzO
BnO
BnO
OBn
24
165.82
138.80
138.41
138.33
136.80
132.86
130.88
129.78
128.51
128.39
128.37
128.31
128.02
127.93
127.87
127.76
127.56
127.54
82.09
77.48
77.36
77.16
76.84
75.87
75.76
74.25
72.66
72.37
72.19
22.58
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

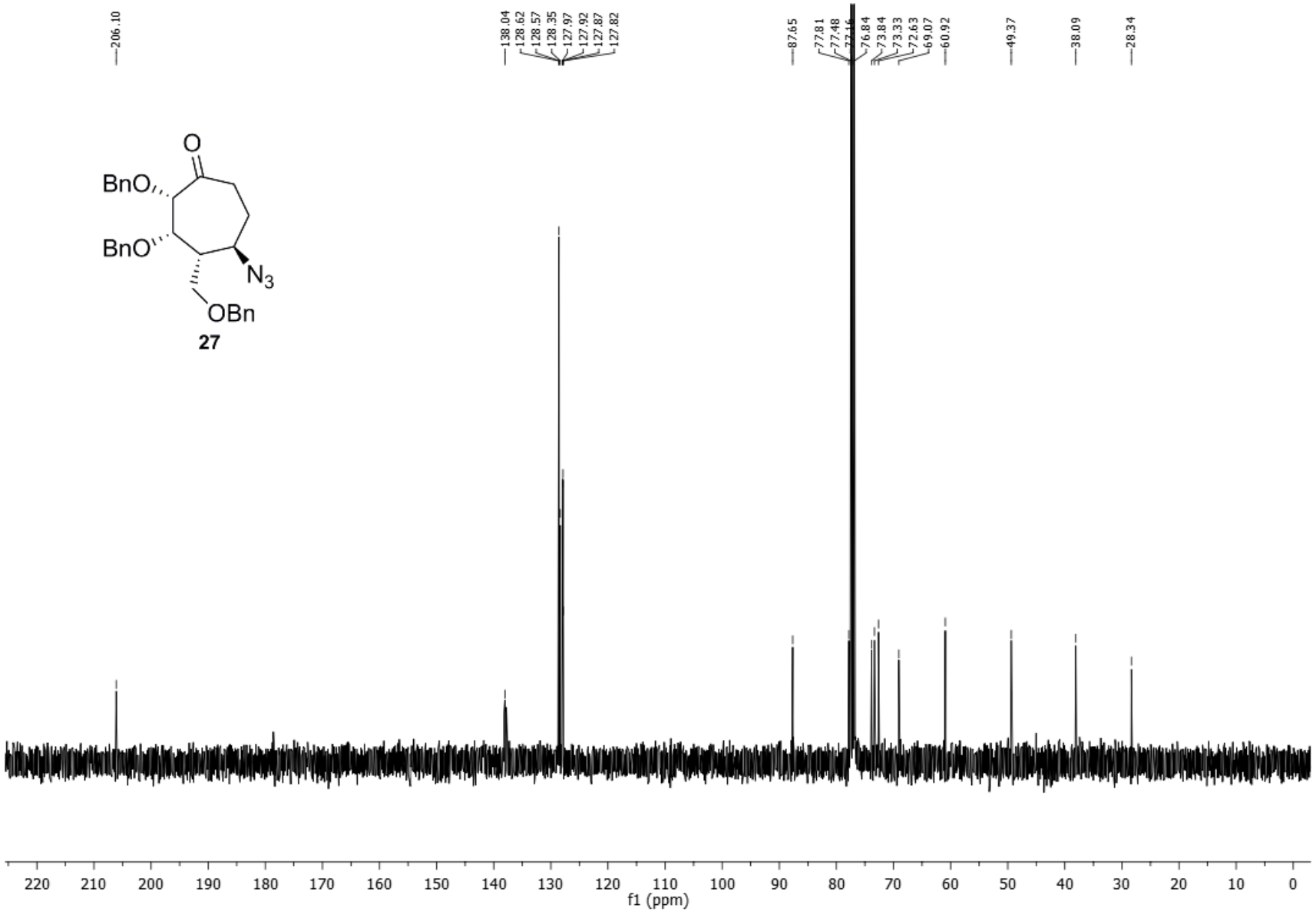

BzO
BnO
BnO
N3
OBn
25
f1 (ppm)

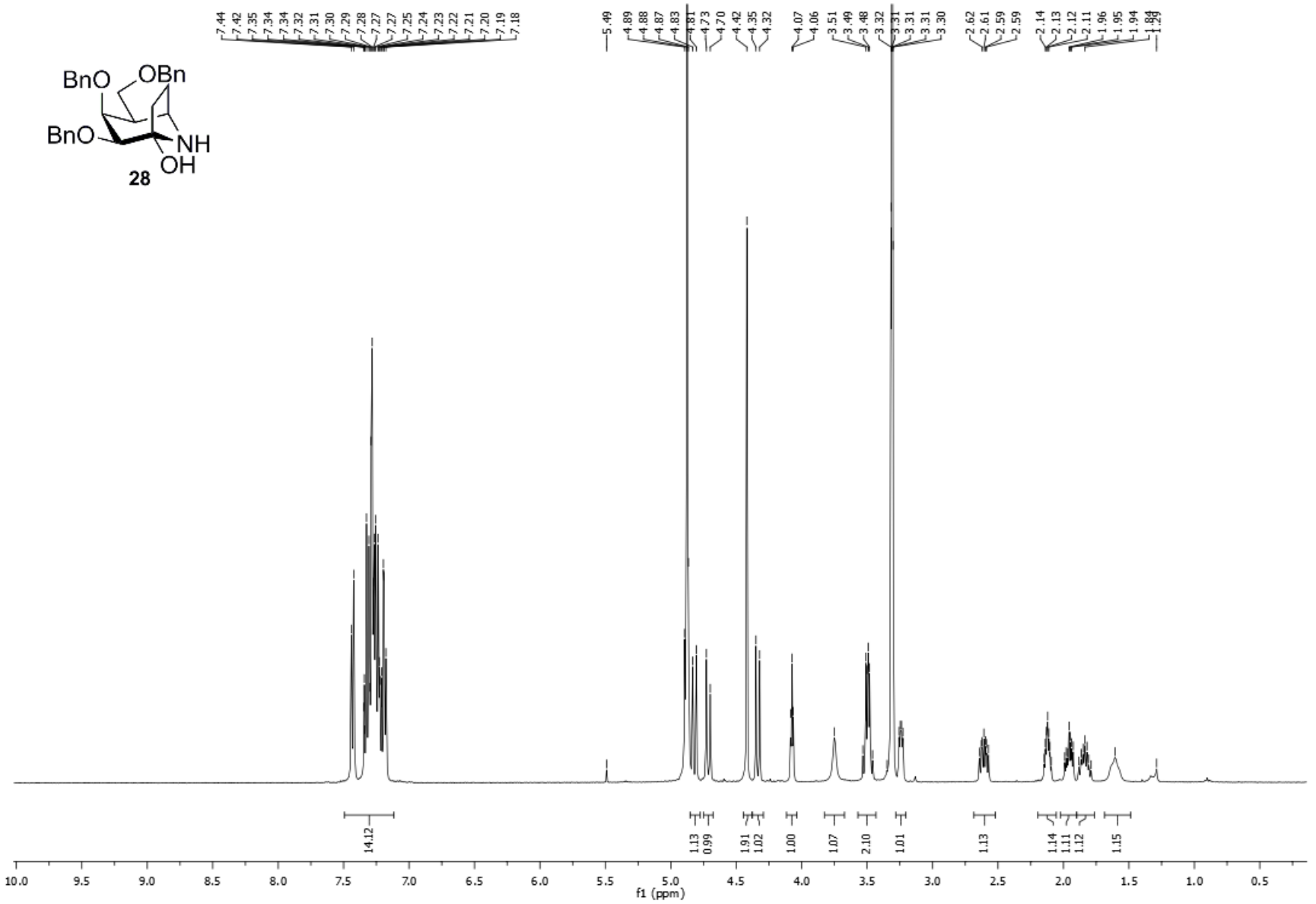

165.97
138.77
138.33
138.00
133.01
130.67
129.78
128.55
128.44
128.40
128.01
128.00
127.87
127.74
127.70
85.87
77.48
77.16
76.84
76.56
75.66
74.77
73.16
72.58
61.58
46.69
27.49
26.73
BzO
BnO
BnO
N3
OBn
25
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

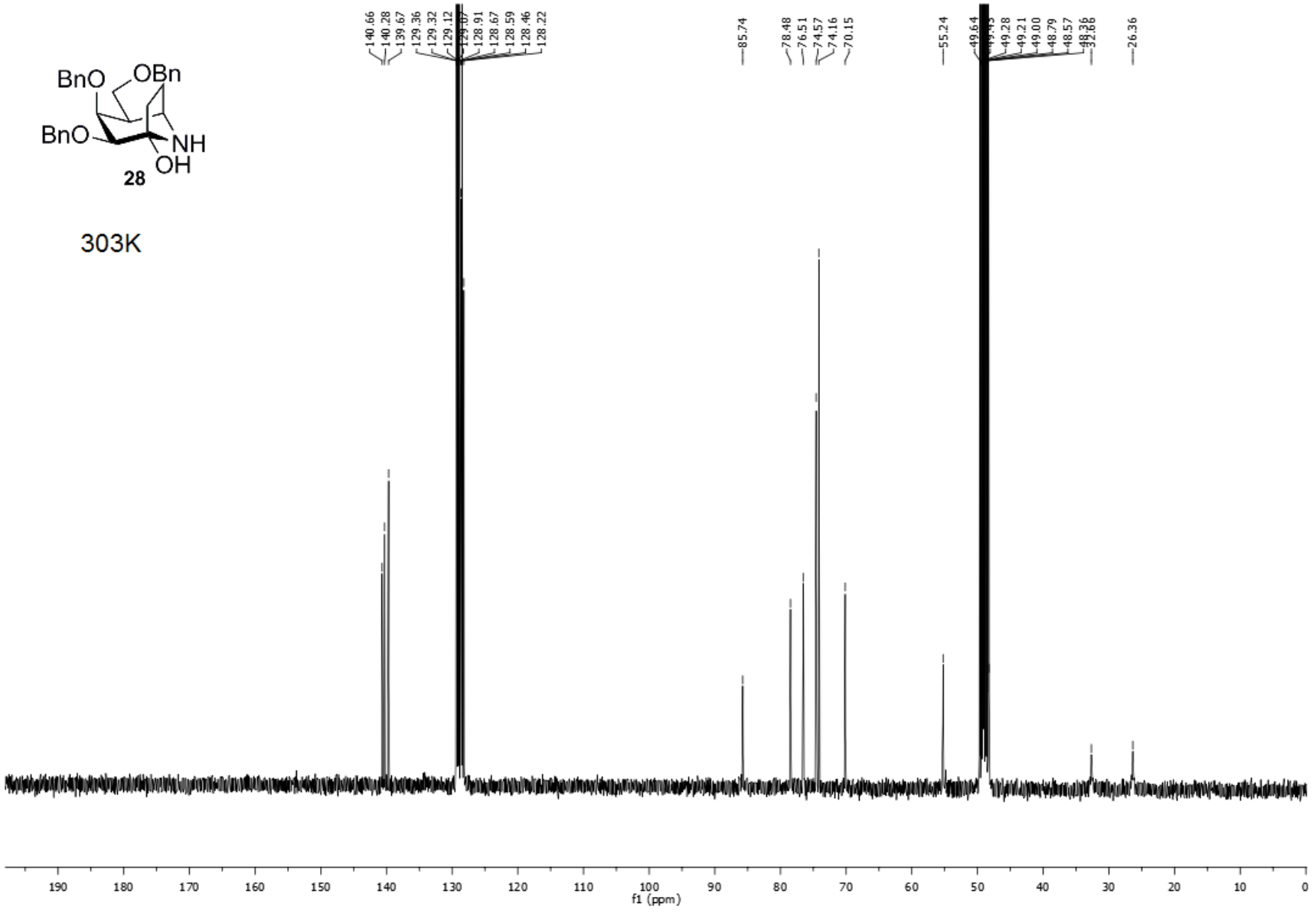

HO
BnO
BnO
N3
OBn
26
f1 (ppm)

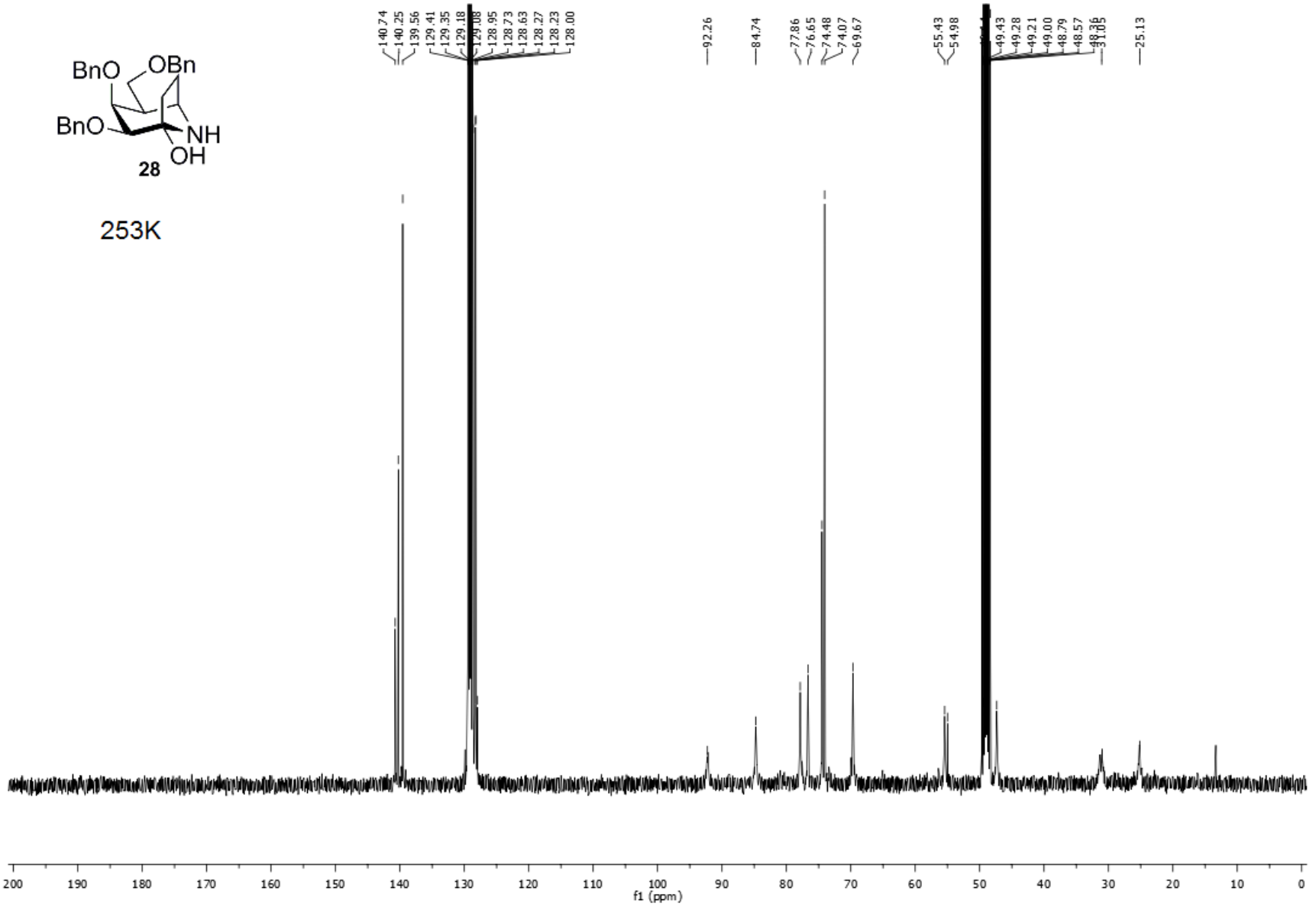

HO
BnO
BnO
N3
OBn
26
138.88
138.27
137.88
128.76
128.53
128.35
128.12
128.01
127.89
127.85
127.64
127.62
89.82
77.48
77.16
76.84
74.58
74.29
73.12
72.51
72.28
70.32
61.01
45.79
27.49
26.99
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

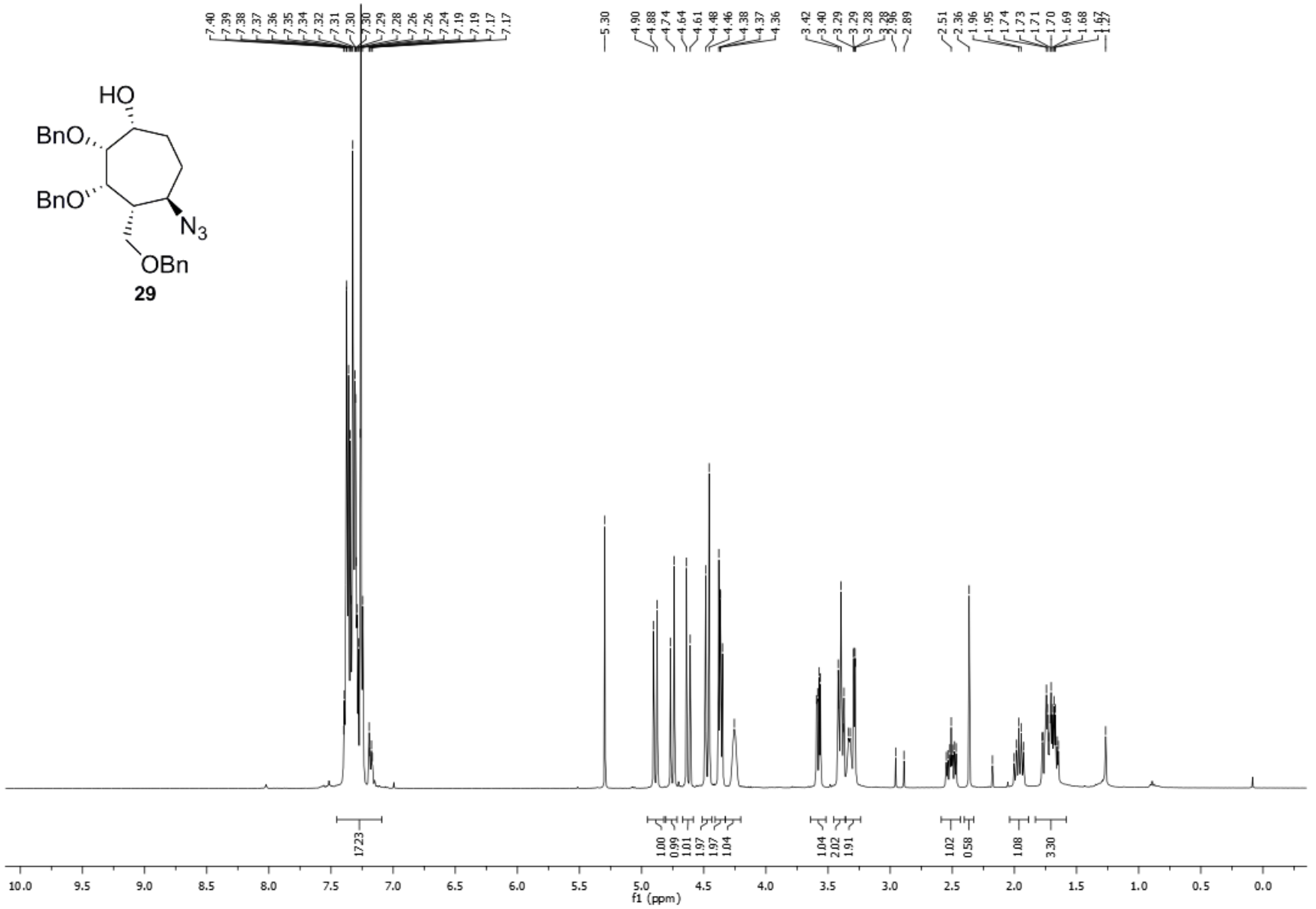

O
BnO
BnO
N3
OBn
27
f1 (ppm)

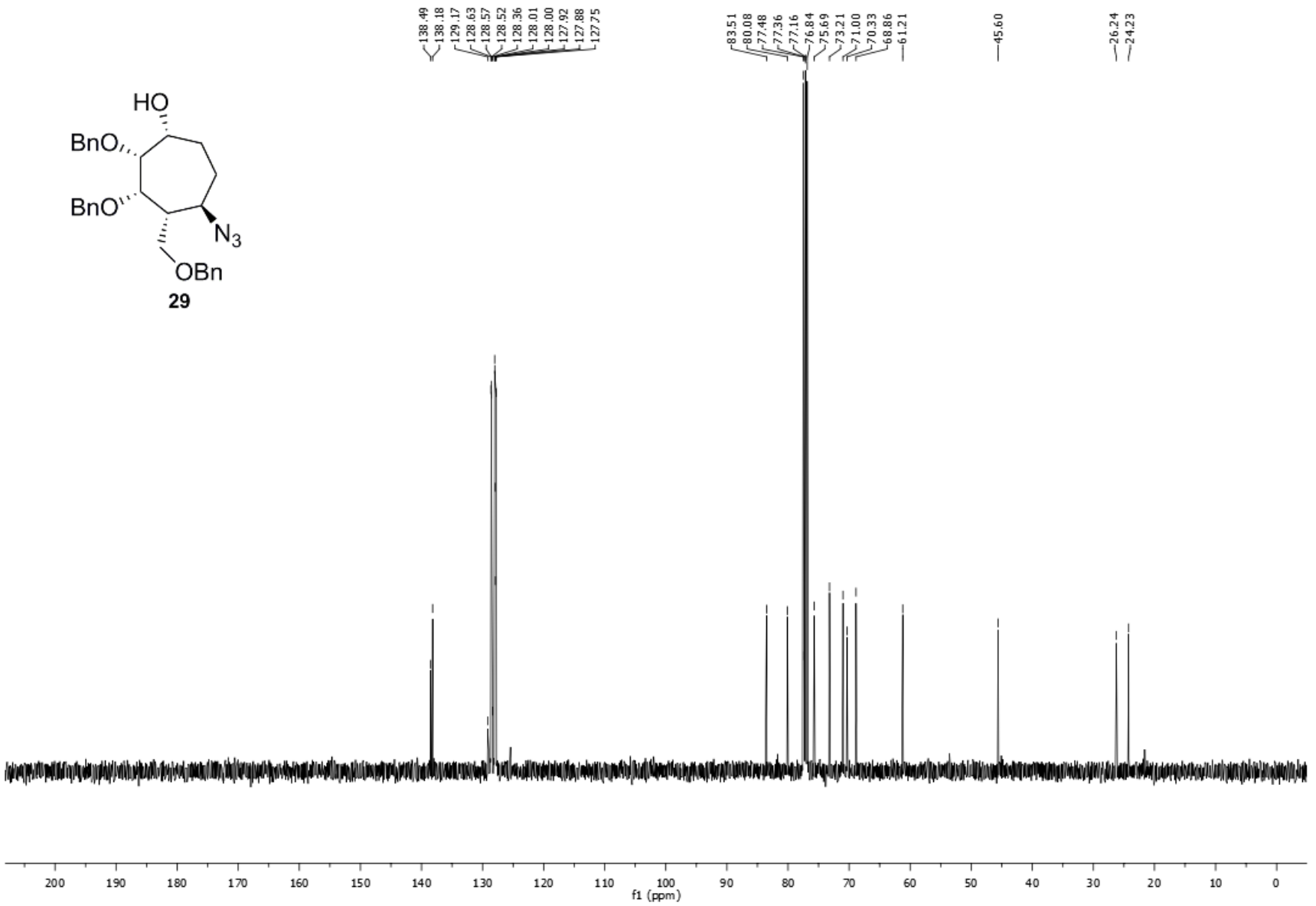

206.10
138.04
128.62
128.57
128.35
127.97
127.92
127.87
127.82
87.65
77.81
77.48
76.84
73.84
73.33
72.63
69.07
60.92
49.37
38.09
28.34
O
BnO
BnO
N3
OBn
27
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

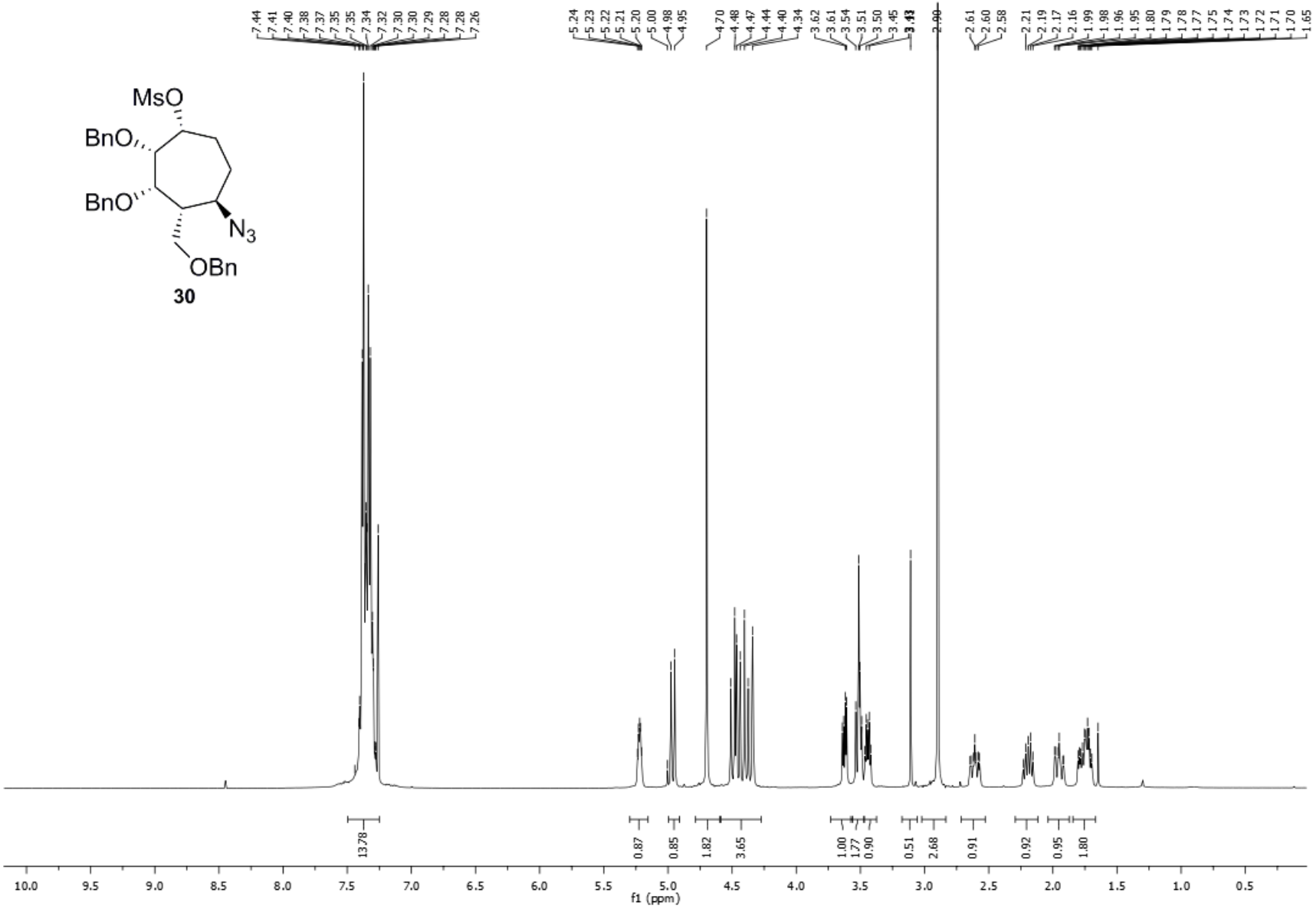

BnO
OBn
BnO
NH
OH
28
f1 (ppm)

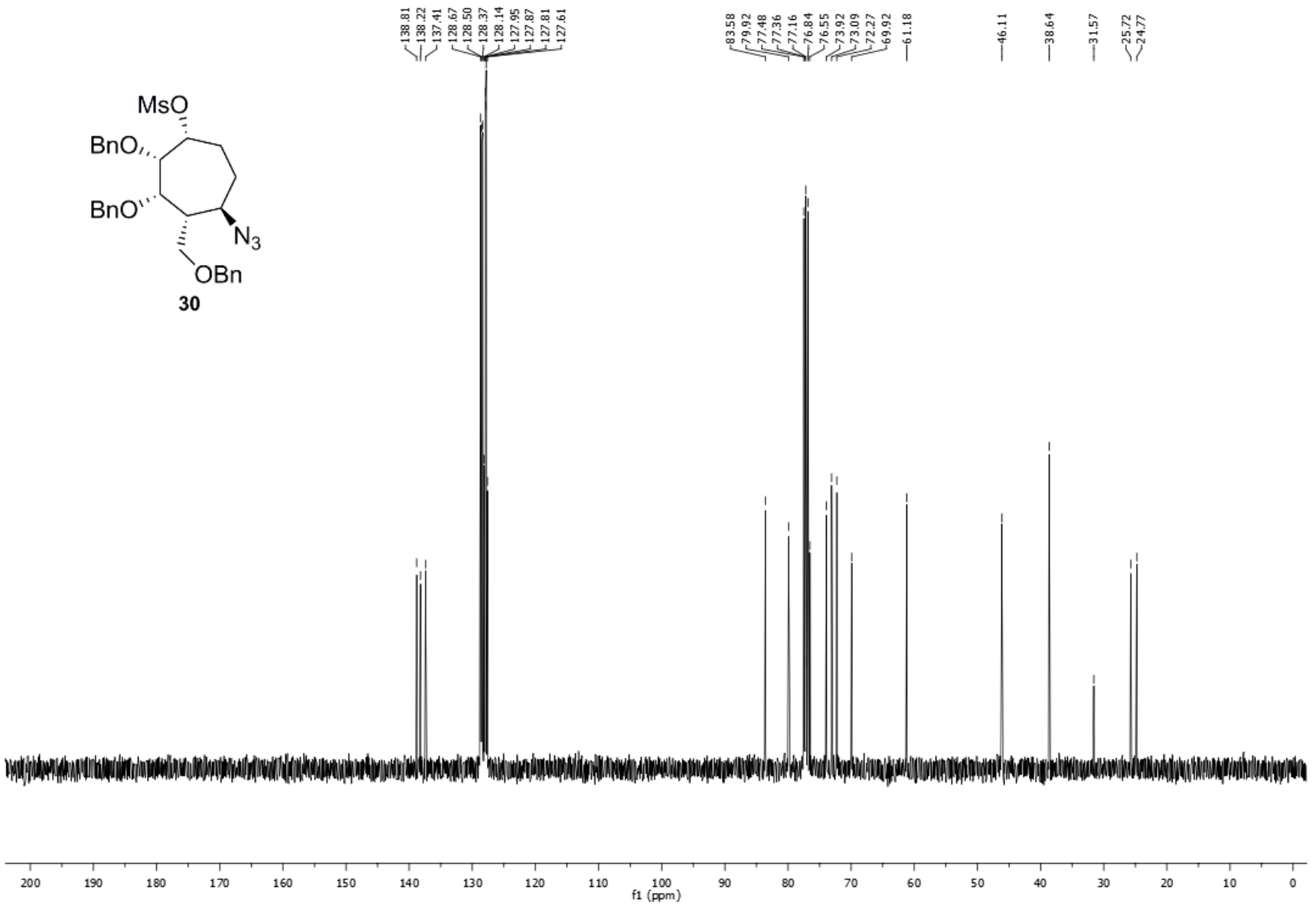

BnO
OBn
BnO
NH
OH
28
303K
140.66
140.28
139.67
129.36
129.32
129.12
128.91
128.67
128.59
128.46
128.22
85.74
78.48
76.51
74.57
74.16
70.15
55.24
49.64
49.28
49.21
49.00
48.79
48.57
26.36
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

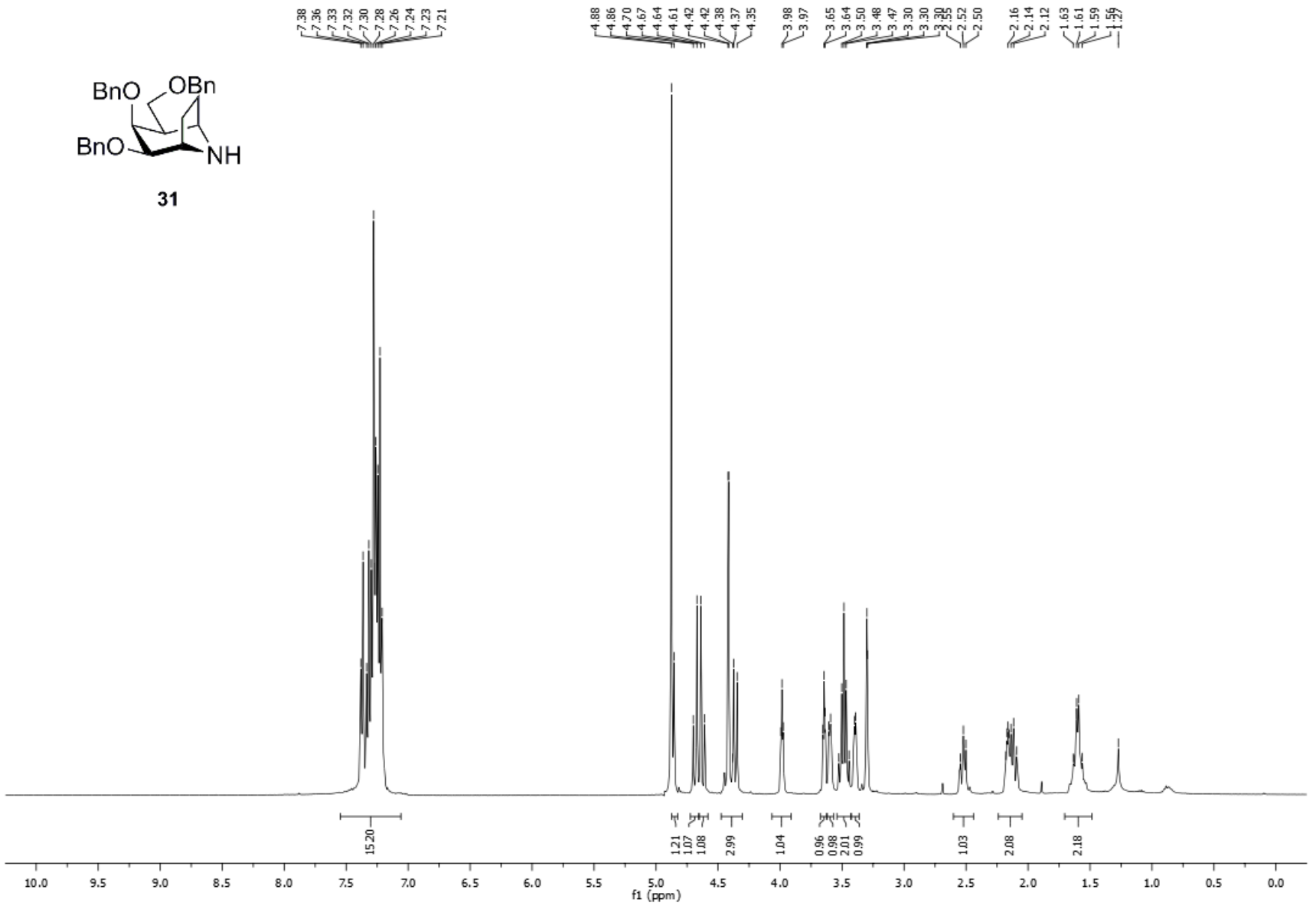

BnO
OBn
BnO
NH
OH
28
253K
140.74
140.25
139.56
129.41
129.35
129.18
129.08
128.95
128.73
128.63
128.27
128.23
128.00
92.26
84.74
77.86
76.65
74.48
74.07
69.67
55.43
54.98
49.43
49.28
49.21
49.00
48.79
48.57
25.13
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

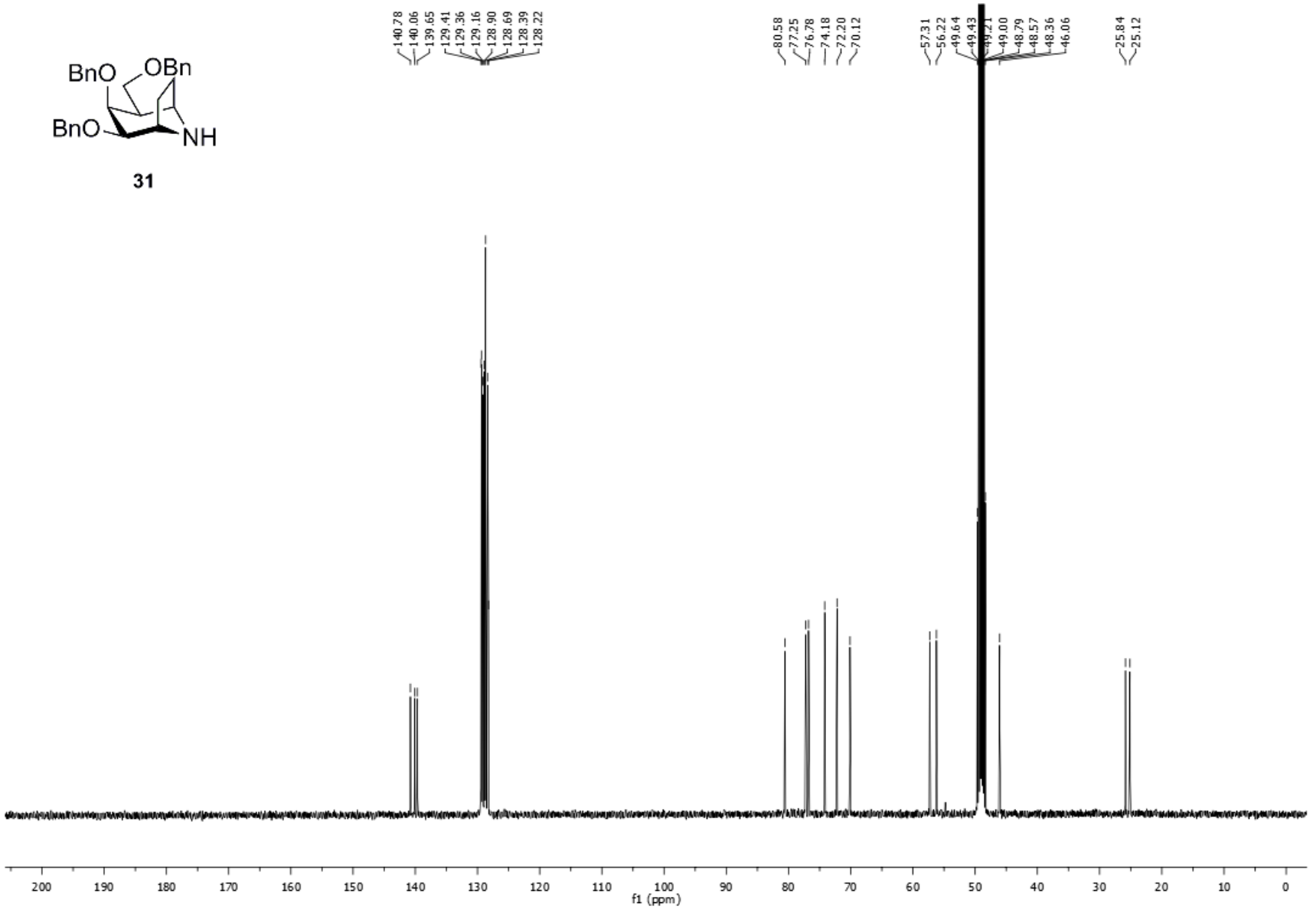

HO
BnO
BnO
N3
OBn
29
7.40 7.39 7.38 7.37 7.36 7.35 7.34 7.32 7.31 7.30 7.30 7.29 7.28 7.26 7.26 7.24 7.19 7.17 7.17
5.30
4.90 4.88 4.74 4.64 4.61 4.48 4.46 4.38 4.37 4.36
3.42 3.40 3.29 3.29 3.28 2.96 2.89
2.51 2.36 1.96 1.95 1.74 1.73 1.71 1.70 1.69 1.68
1.23
1.00 0.99 1.01 1.97 1.97 1.04
1.04 2.02 1.91
1.02 0.58
1.08
3.30
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)

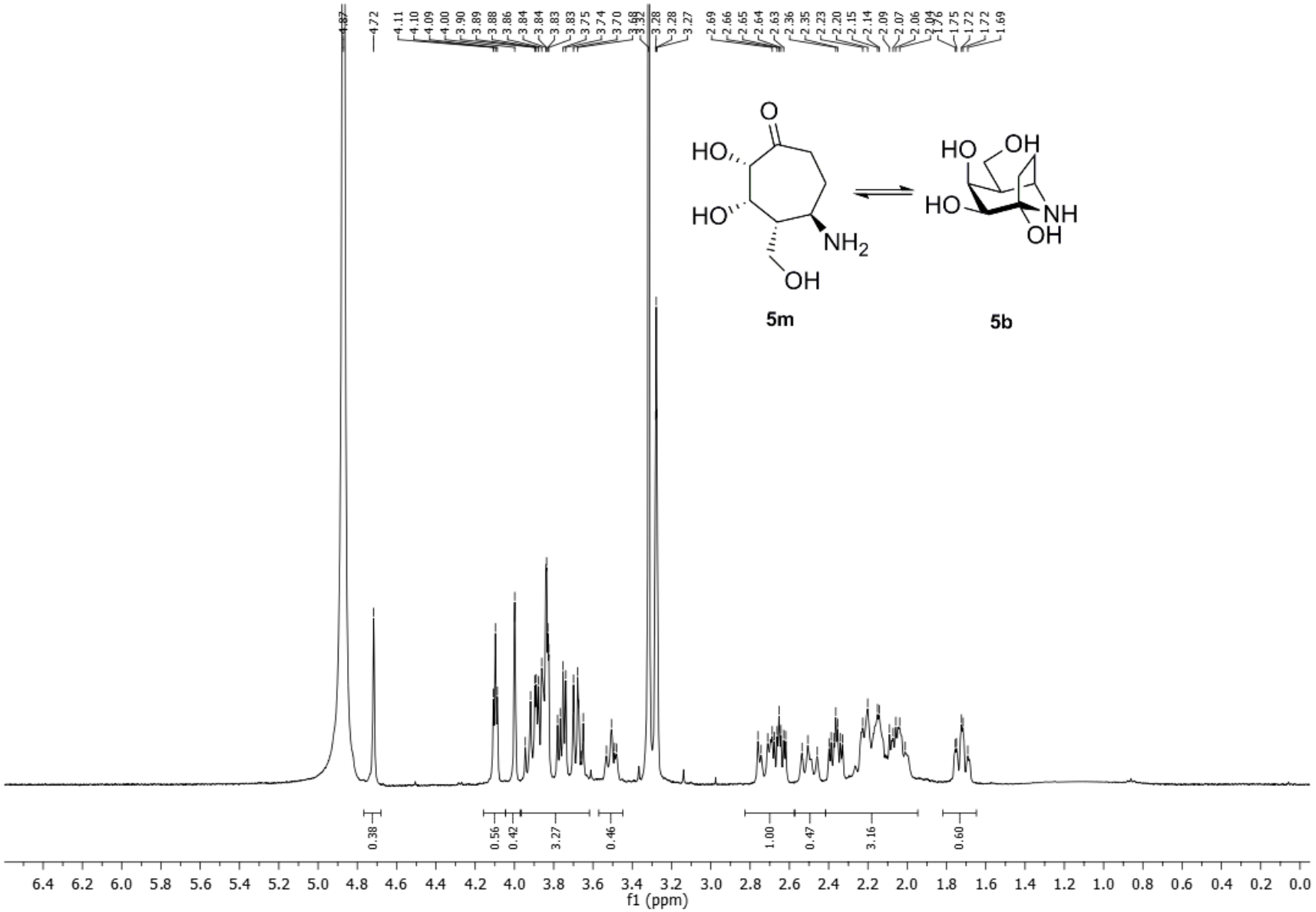

HO
BnO
BnO
N3
OBn
29
138.49
138.18
129.17
128.63
128.57
128.52
128.36
128.01
128.00
127.92
127.88
127.75
83.51
80.08
77.48
77.36
77.16
76.84
75.69
73.21
71.00
70.33
68.86
61.21
45.60
26.24
24.23
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

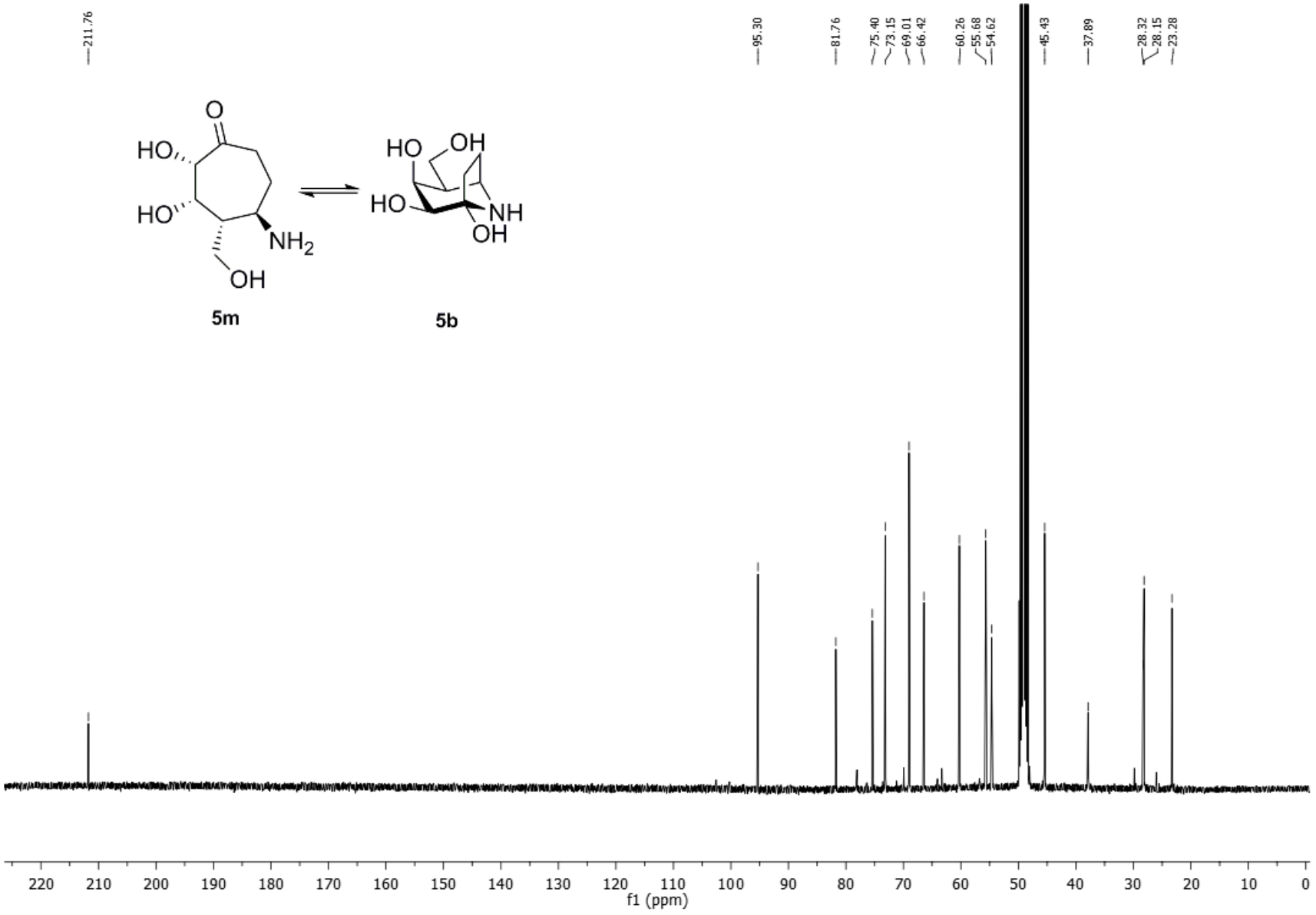

MsO
BnO
BnO
N3
OBn
30
f1 (ppm)

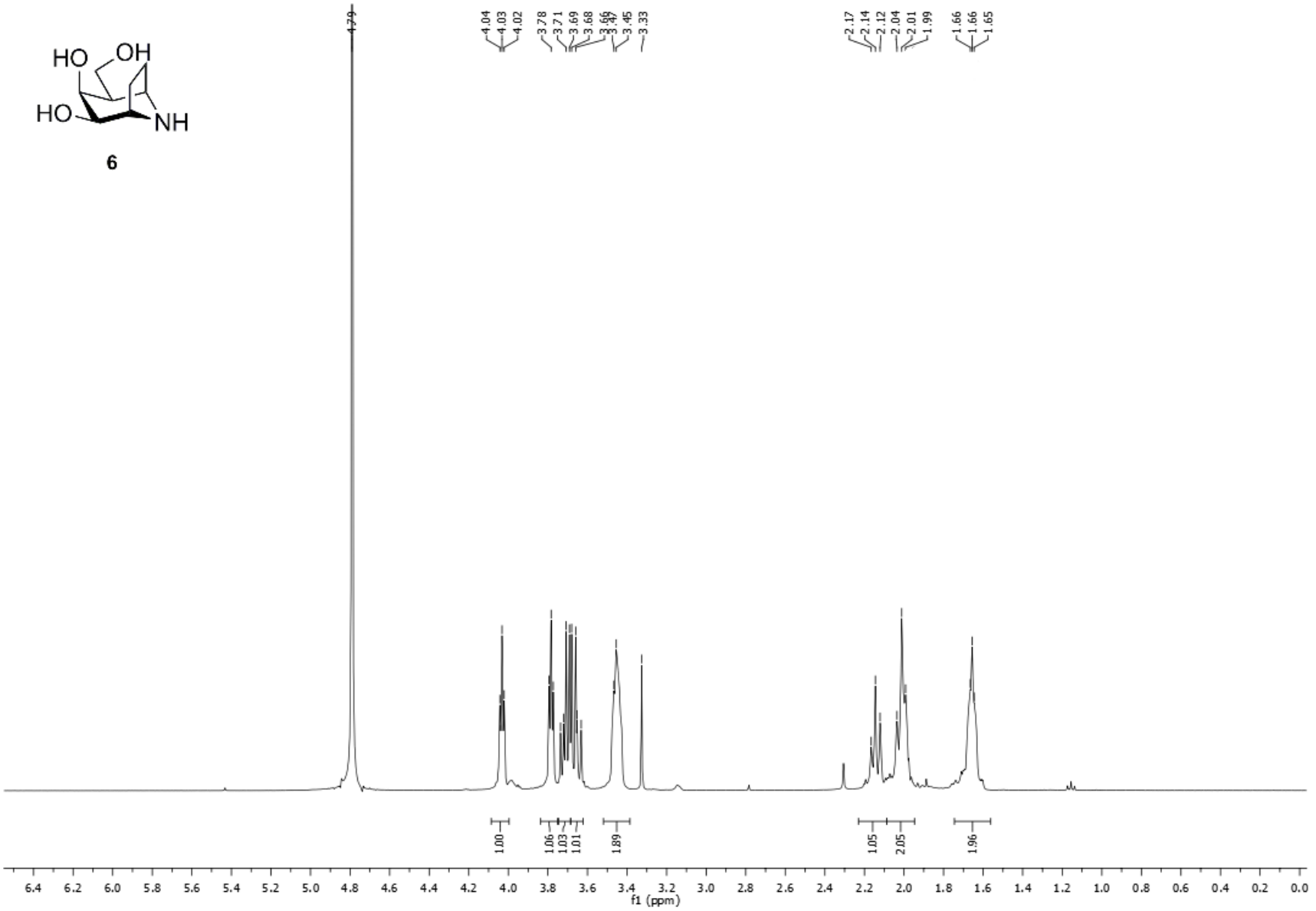

MsO
BnO
BnO
N3
OBn
30
138.81
138.22
137.41
128.67
128.50
128.37
128.14
127.95
127.87
127.81
127.61
83.58
79.92
77.48
77.36
77.16
76.84
76.55
73.92
73.09
72.27
69.92
61.18
46.11
38.64
31.57
25.72
24.77
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)


BnO
OBn
BnO
NH
31
7.38
7.36
7.33
7.32
7.30
7.28
7.26
7.24
7.23
7.21
4.88
4.86
4.70
4.67
4.64
4.61
4.42
4.42
4.38
4.37
4.35
3.98
3.97
3.65
3.64
3.50
3.48
3.47
3.30
3.30
2.52
2.50
2.16
2.14
2.12
1.63
1.61
1.59
15.20
1.21
1.07
1.08
2.99
1.04
0.96
0.98
2.01
0.99
1.03
2.08
2.18
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)


BnO
OBn
BnO
NH
31
140.78
140.06
139.65
129.41
129.36
129.16
128.90
128.69
128.39
128.22
80.58
77.25
76.78
74.18
72.20
70.12
57.31
56.22
49.64
49.43
49.21
49.00
48.79
48.57
48.36
46.06
25.84
25.12
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

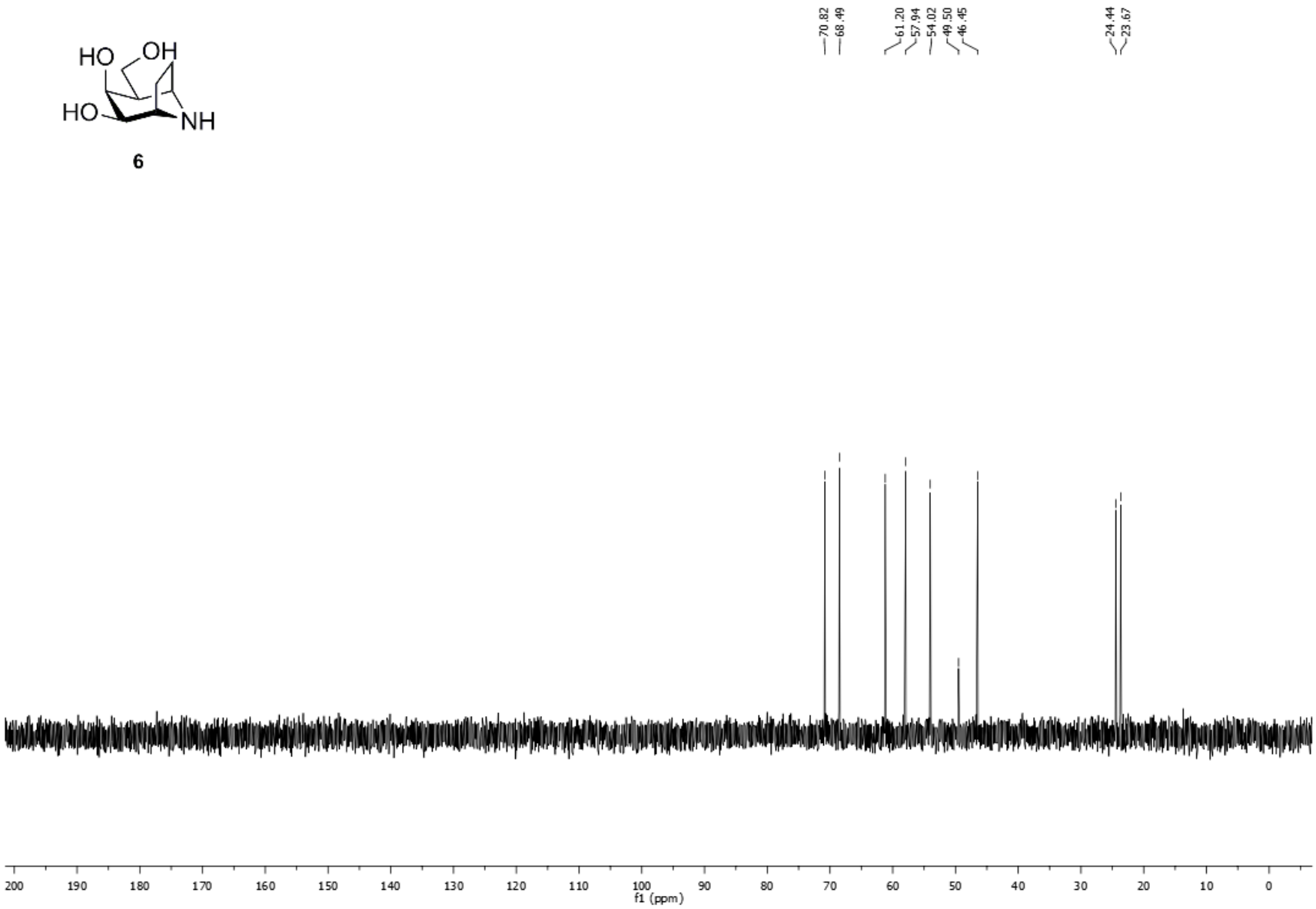

O
HO
HO
NH2
OH
HO
OH
HO
NH
OH
5m
5b
f1 (ppm)


211.76
95.30
81.76
75.40
73.15
69.01
66.42
60.26
55.68
54.62
45.43
37.89
28.32
28.15
23.28
O
HO
HO
NH2
OH
HO
OH
HO
NH
OH
5m
5b
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)


HO
OH
HO
NH
6
4.79
4.04
4.03
4.02
3.78
3.71
3.69
3.68
3.45
3.33
2.17
2.14
2.12
2.04
2.01
1.99
1.66
1.66
1.65
1.00
1.06
1.03
1.01
1.89
1.05
2.05
1.96
f1 (ppm)

HO
OH
HO
NH

6


70.82
68.49
61.20
57.94
54.02
49.50
46.45
24.44
23.67
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)