First Total Synthesis of (+)-Broussonetine W: Glycosidase Inhibition of Natural Product & Analogs

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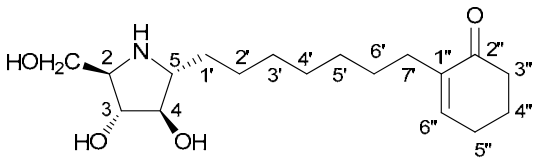
1. Comparison of ^{13}C NMR of synthetic broussonetine W (4) with natural product.

Table 1. Comparison of the synthetic broussonetine W (**4**) with natural product



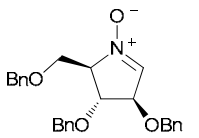
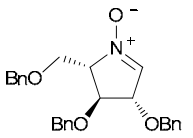
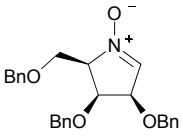
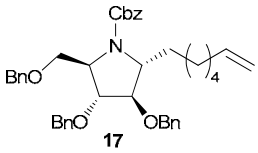
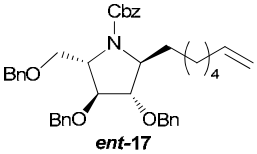
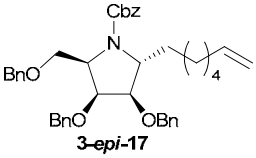
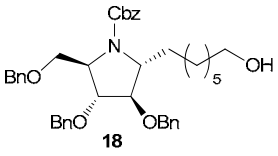
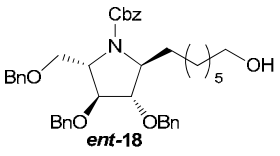
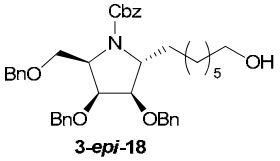
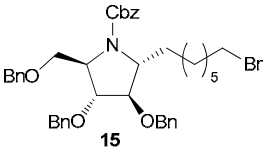
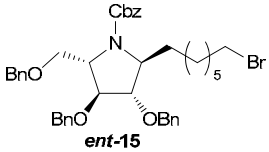
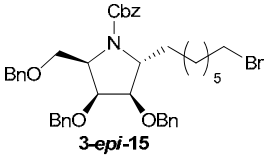
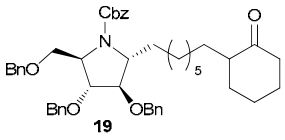
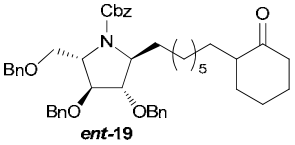
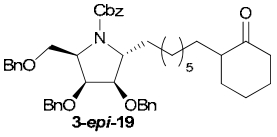
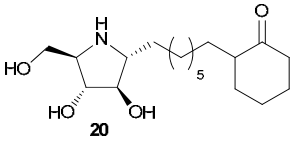
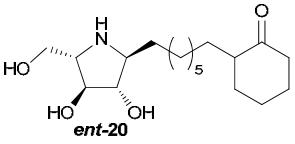
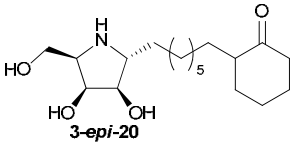
		W (lit.)[a]	W (4)[b]	
Pyrrolidine moiety	CH_2OH	63.45	62.99	-0.46
	2	64.97	65.21	0.24
	3	80.07	79.60	-0.47
	4	84.06	83.53	-0.53
	5	62.74	62.85	0.11
Octyl moiety	1'	35.43	34.80	-0.63
	2'	27.23	27.23	0.00
	3'	30.12	30.14	0.02
	4' } 5'	29.74, 29.70	29.82, 29.82	0.08, 0.12
	6'	28.98	29.09	0.11
	7'	29.96	30.09	0.13
Cyclohexenone moiety	1''	139.79	139.91	0.12
	2''	198.81	198.81	0.00
	3''	38.81	38.94	0.13
	4''	23.43	23.57	0.14
	5''	26.09	26.22	0.13
	6''	145.09	145.15	0.06

a) δ in pyridine-d_5 at 125 MHz. b) δ in pyridine-d_5 at 126 MHz.

2. Table for all the new compounds.

Experimental procedures and characterization data for all the new compounds see the general procedures in essay for the details.

Table 1. New Compounds in the total synthesis of (**4**, ***ent*-4**, **3-*epi*-4**)

	Synthesis of **4**	Synthesis of ***ent*-4**	Synthesis of **3-*epi*-4**
Cyclic Nitrones: (Starting Materials)	 **D-*arabino*-nitrone** **(10)**	 **L-*arabino*-nitrone** **(*ent*-10)**	 **D-*lyxo*-nitrone** **(3-*epi*-10)**
Step1-3: Grignard addition Reduction Protection of amino group	 **17** 87% yield in 3 steps starting from **10**	 ***ent*-17** 98% yield in 3 steps starting from ***ent*-10**	 **3-*epi*-17** 66% yield in 3 steps starting from **3-*epi*-10**
Step 4: Hydroboration-Oxidation	 **18** 73% yield derived from olefin **17**	 ***ent*-18** 93% yield derived from olefin ***ent*-17**	 **3-*epi*-18** 100% yield derived from olefin **3-*epi*-17**
Step 5: Appel reaction	 **15** 97% yield derived from alcohol **18**	 ***ent*-15** 85% yield derived from alcohol ***ent*-18**	 **3-*epi*-15** 88% yield derived from alcohol **3-*epi*-18**
Step 6: α-alkylation of imine (*N*-cyclohexylidenecyclohexanamine **14**)	 **19** 73% yield derived from bromide **15**	 ***ent*-19** 71% yield derived from bromide ***ent*-15**	 **3-*epi*-19** 60% yield derived from alcohol **3-*epi*-15**
Step 7: 10% Pd/C-catalyzed hydrogenolysis reaction	 **20** 96% yield derived from ketone **19**	 ***ent*-20** 84% yield derived from ketone ***ent*-19**	 **3-*epi*-20** 75% yield derived from ketone **3-*epi*-19**

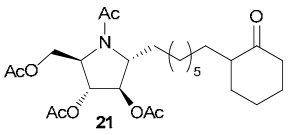
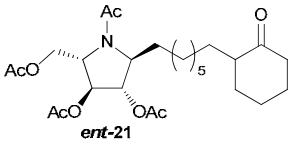
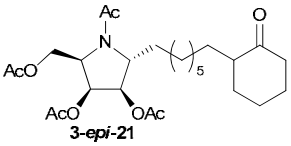
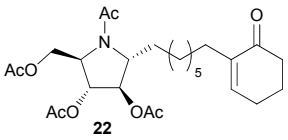
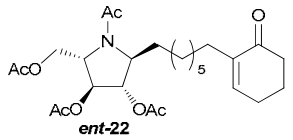
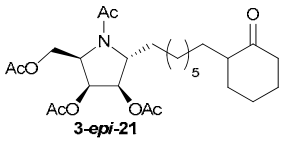
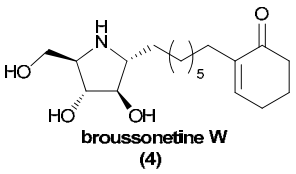
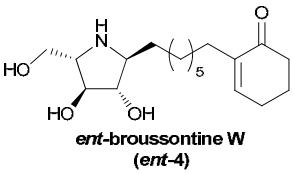
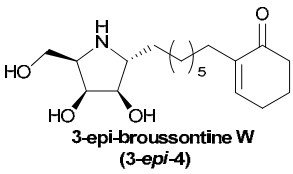
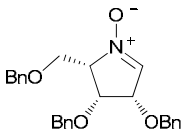
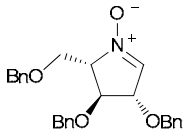
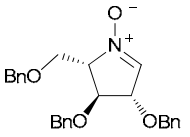
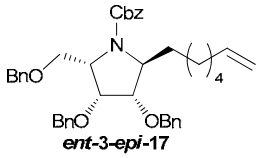
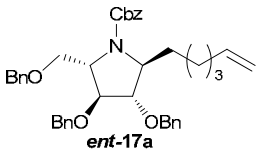
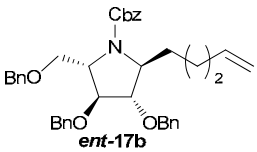
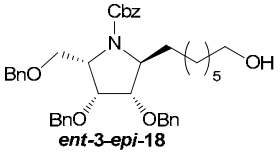
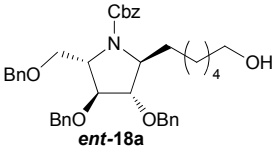
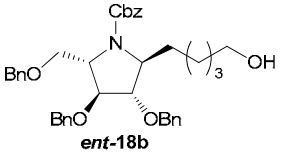
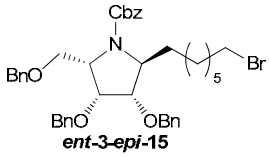
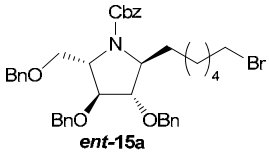
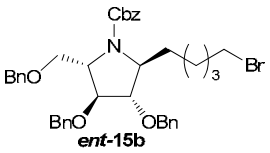
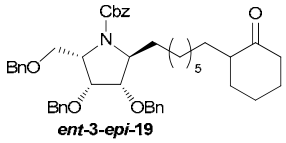
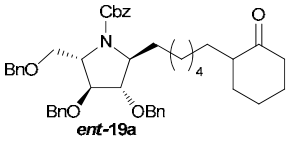
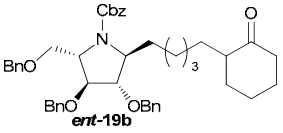
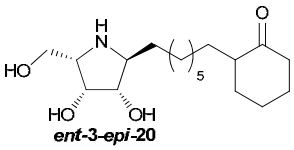
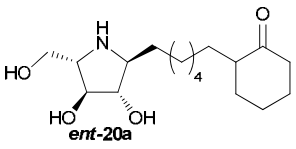
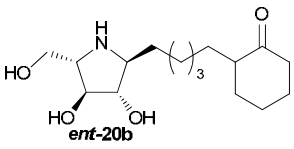
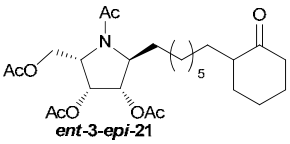
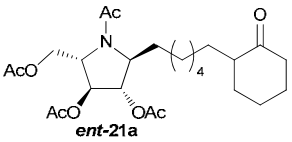
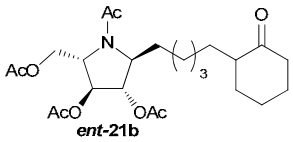
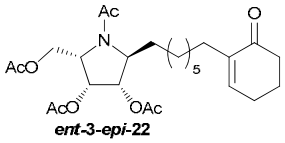
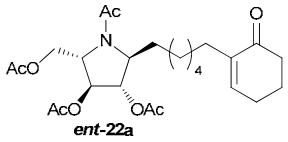
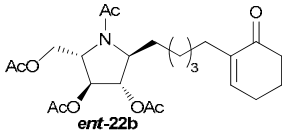
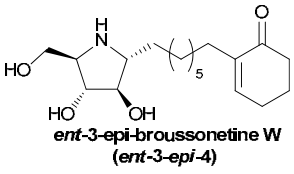
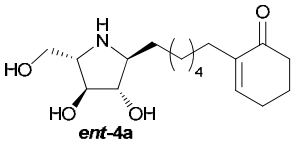
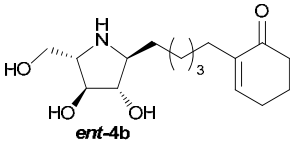
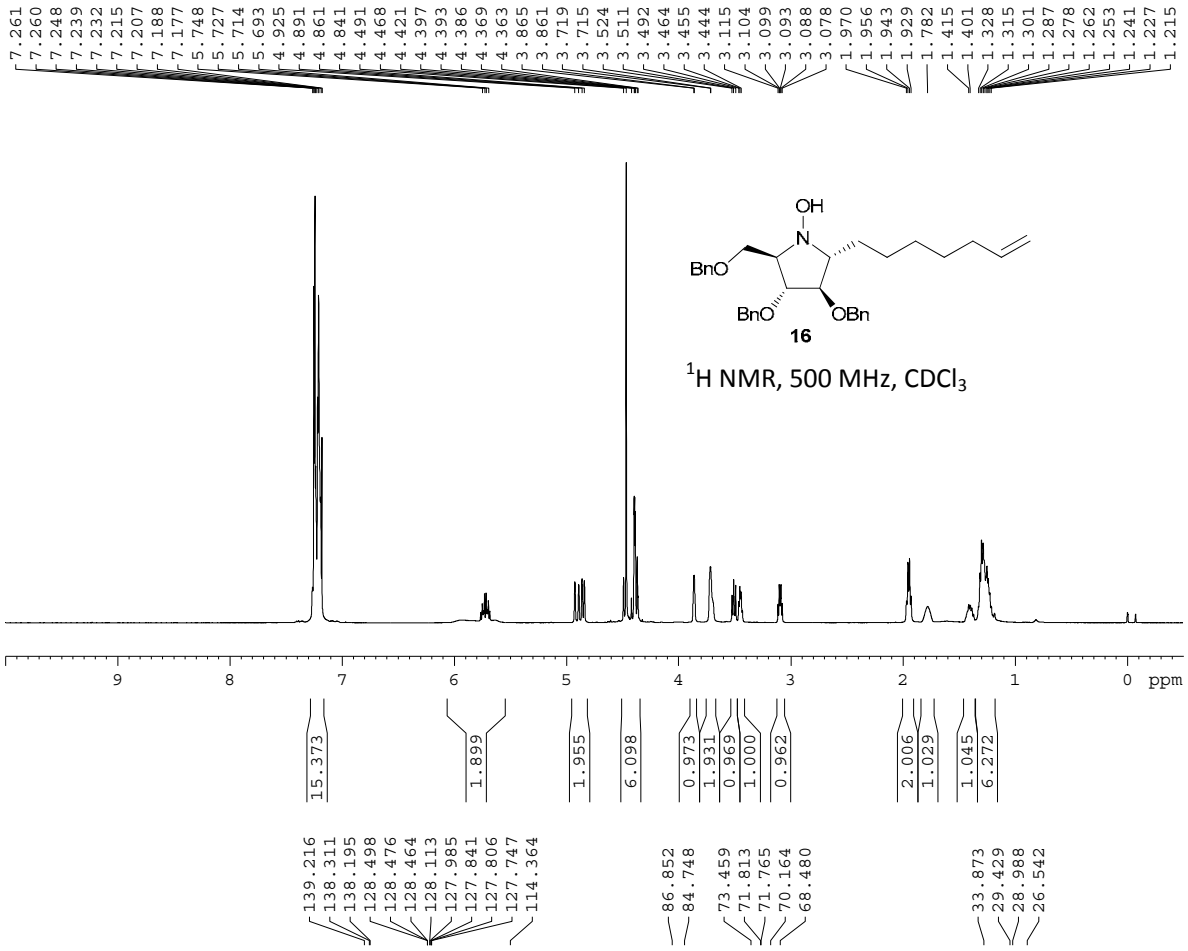
Step 8: Protection of the amino and hydroxyl groups with Ac_2O	 **21** 95% yield derived from ketone **20**	 ***ent*-21** 94% yield derived from ketone ***ent*-20**	 **3-*epi*-21** 76% yield derived from ketone **3-*epi*-20**
Step 9-10: Bromination-elimination of ketone	 **22** 83% yield in 2 steps derived from ketone **21**	 ***ent*-22** 85% yield in 2 steps derived from ketone ***ent*-21**	 **3-*epi*-21** 78% yield in 2 steps derived from ketone **3-*epi*-21**
Step 11: Removal of acetyl groups	 **broussonetine W (4)** 92% yield derived from compound **22**	 ***ent*-broussontine W (*ent*-4)** 78% yield derived from compound ***ent*-22**	 **3-epi-broussontine W (3-*epi*-4)** 89% yield derived from compound **3-*epi* -22**
Total Yield In 10 Steps:	**31%**	**29%**	**14%**

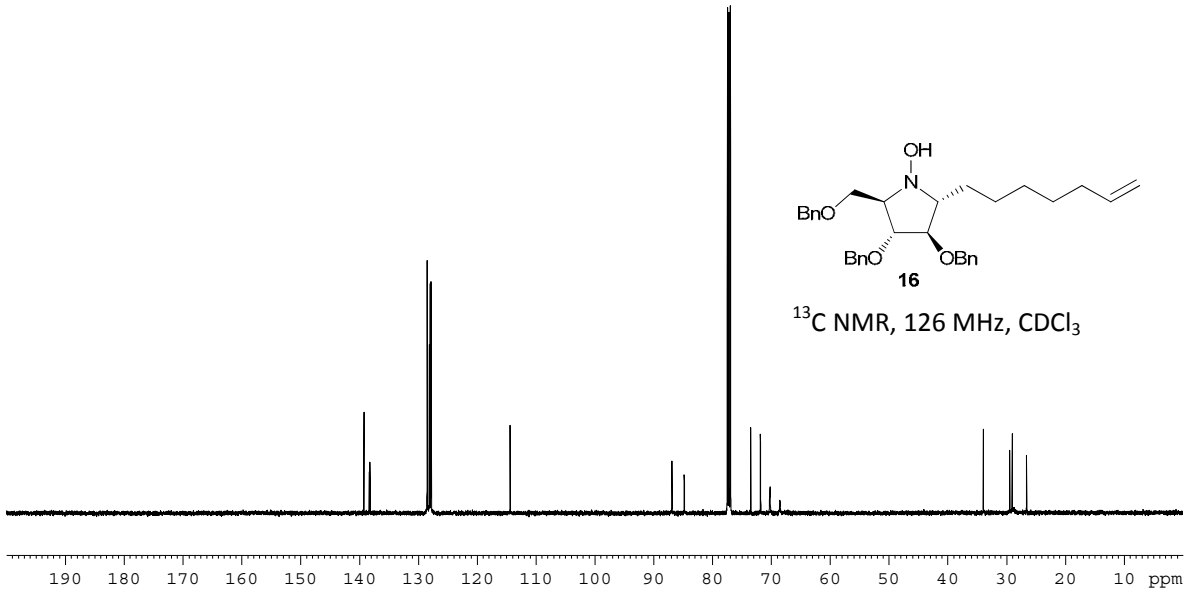
Table 2. New Compounds in the total synthesis of (***ent*-3-*epi*-4**, ***ent*-4a**, ***ent*-4b**)

	Synthesis of ***ent*-3-*epi*-4**	Synthesis of ***ent*-4a**	Synthesis of ***ent*-4b**
Cyclic Nitrones: (Starting Materials)	 **L-*lyxo*-nitrone (*ent*-3-*epi*-10)**	 **L-*arabino*-nitrone (*ent*-10)**	 **L-*arabino*-nitrone (*ent*-10)**
Step1-3: Grignard addition Reduction Protection of amino group	 ***ent*-3-*epi*-17** 76% yield in 3 steps starting from ***ent*-3-*epi*-10**	 ***ent*-17a** 84% yield in 3 steps starting from ***ent*-10**	 ***ent*-17b** 93% yield in 3 steps starting from ***ent*-10**
Step 4: Hydroboration-Oxidation	 ***ent*-3-*epi*-18** 73% yield derived from olefin ***ent*-3-*epi*-17**	 ***ent*-18a** 91% yield derived from olefin ***ent*-17a**	 ***ent*-18b** 91% yield derived from olefin ***ent*-17b**

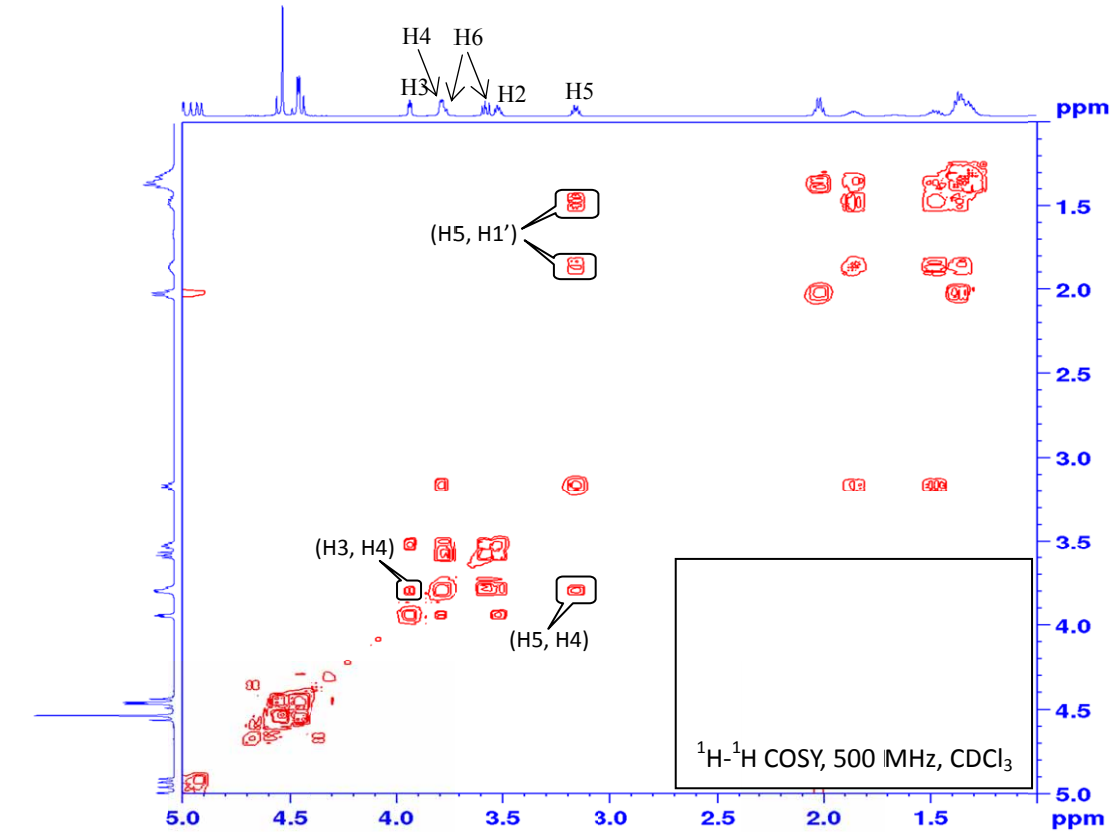
Step 5: Appel reaction	 ***ent*-3-*epi*-15** 96% yield derived from alcohol ***ent*-3-*epi*-18**	 ***ent*-15a** 92% yield derived from alcohol ***ent*-18a**	 ***ent*-15b** 93% yield derived from alcohol ***ent*-18b**
Step 6: α-alkylation of imine (*N*-cyclohexylidenecyclohexanamine **14**)	 ***ent*-3-*epi*-19** 71% yield derived from bromide ***ent*-3-*epi*-15**	 ***ent*-19a** 68% yield derived from bromide ***ent*-15a**	 ***ent*-19b** 71% yield derived from alcohol ***ent*-15b**
Step 7: 10% Pd/C-catalyzed hydrogenolysis reaction	 ***ent*-3-*epi*-20** 79% yield derived from ketone ***ent*-3-*epi*-19**	 ***ent*-20a** 82% yield derived from ketone ***ent*-19a**	 ***ent*-20b** 83% yield derived from ketone ***ent*-19b**
Step 8: Protection of the amino and hydroxyl groups with Ac_2O	 ***ent*-3-*epi*-21** 94% yield derived from ketone ***ent*-3-*epi*-20**	 ***ent*-21a** 93% yield derived from ketone ***ent*-20a**	 ***ent*-21b** 90% yield derived from ketone ***ent*-20b**
Step 9-10: Bromination-elimination of ketone	 ***ent*-3-*epi*-22** 83% yield in 2 steps derived from ketone ***ent*-3-*epi*-21**	 ***ent*-22a** 74% yield in 2 steps derived from ketone ***ent*-21a**	 ***ent*-22b** 74% yield in 2 steps derived from ketone ***ent*-21b**
Step 11: Removal of acetyl groups	 ***ent*-3-epi-broussonetine W (*ent*-3-*epi*-4)** 77% yield derived from compound ***ent*-3-*epi*-22**	 ***ent*-4a** 86% yield derived from compound ***ent*-22a**	 ***ent*-4b** 74% yield derived from compound ***ent*-22b**
Total Yield In 10 Steps:	**18%**	**23%**	**23%**

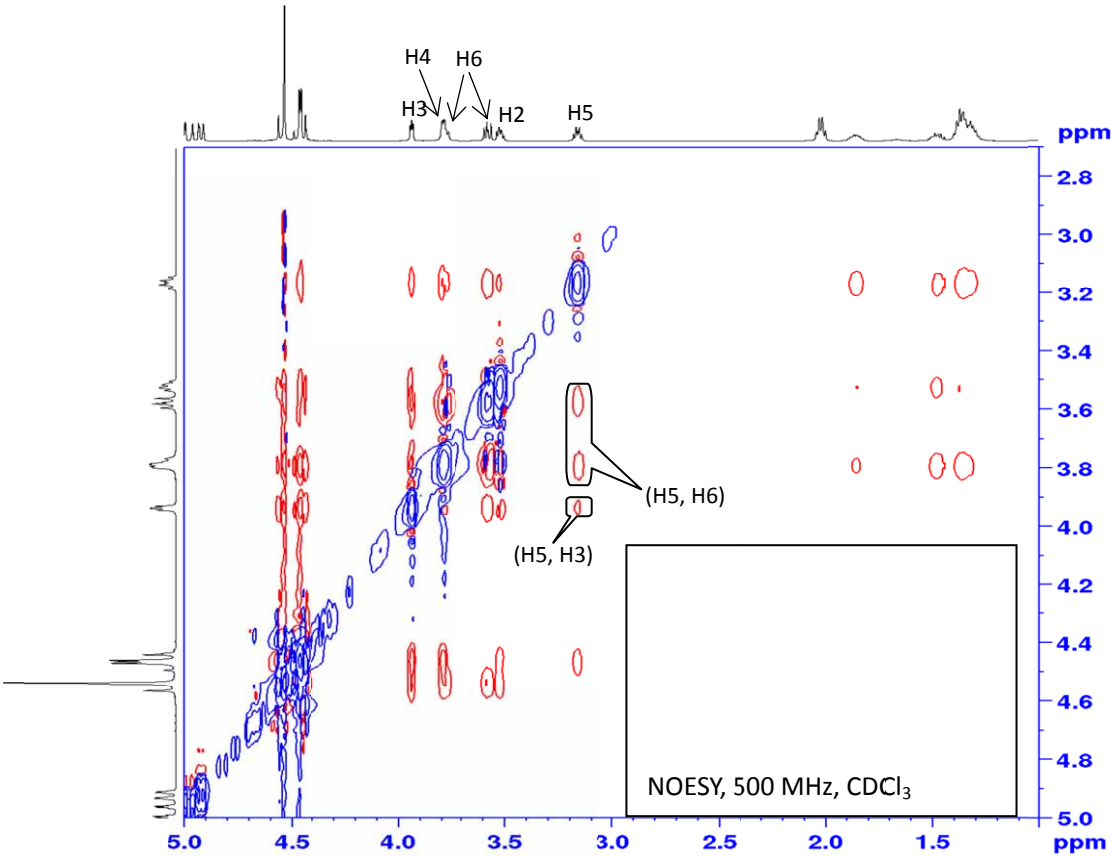
3. Copies of ^{1}H , ^{13}C and 2D NMR spectra of new compounds.

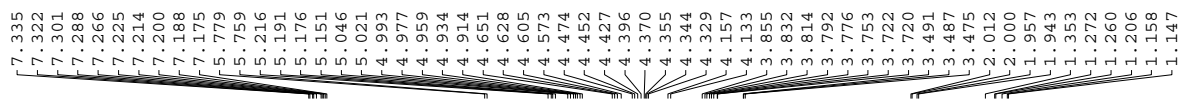


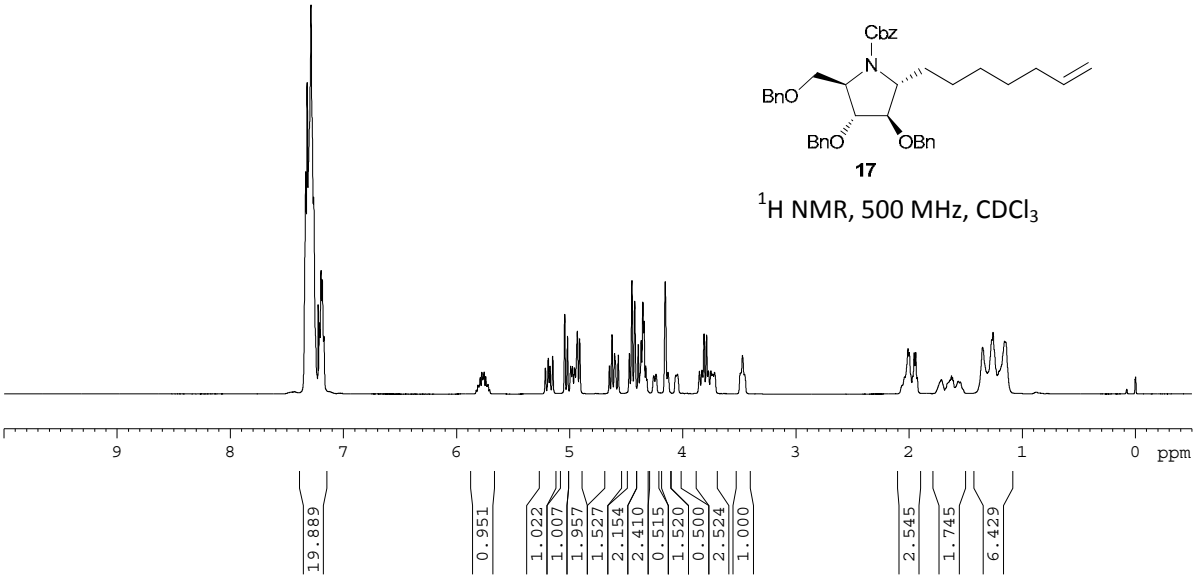


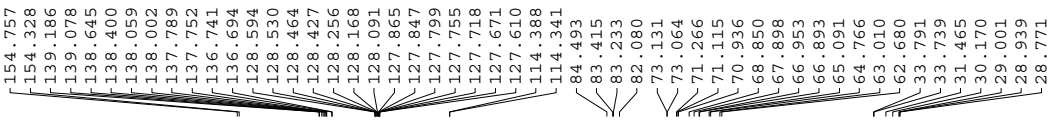
COSY of compound **16**

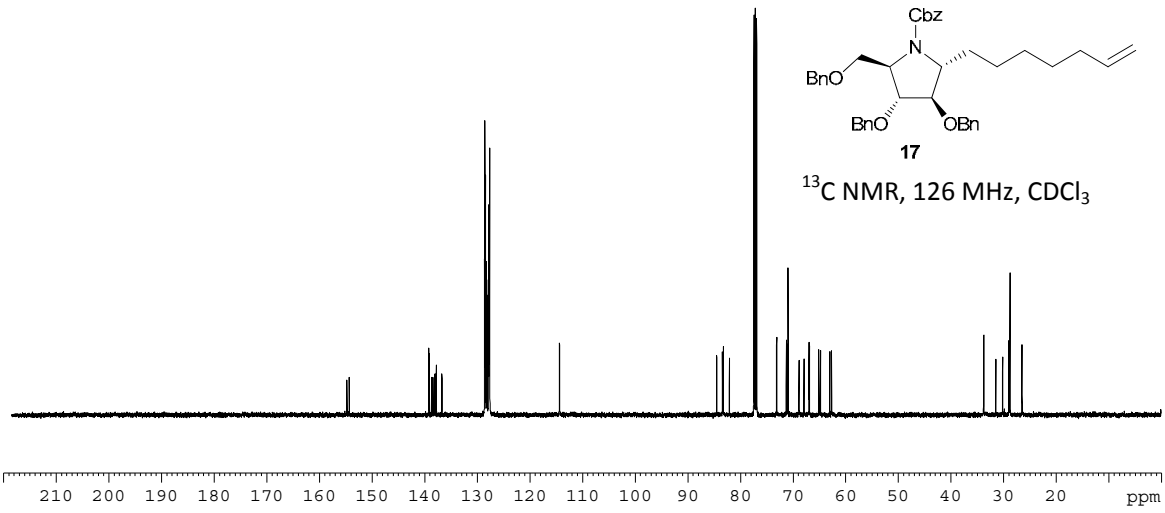


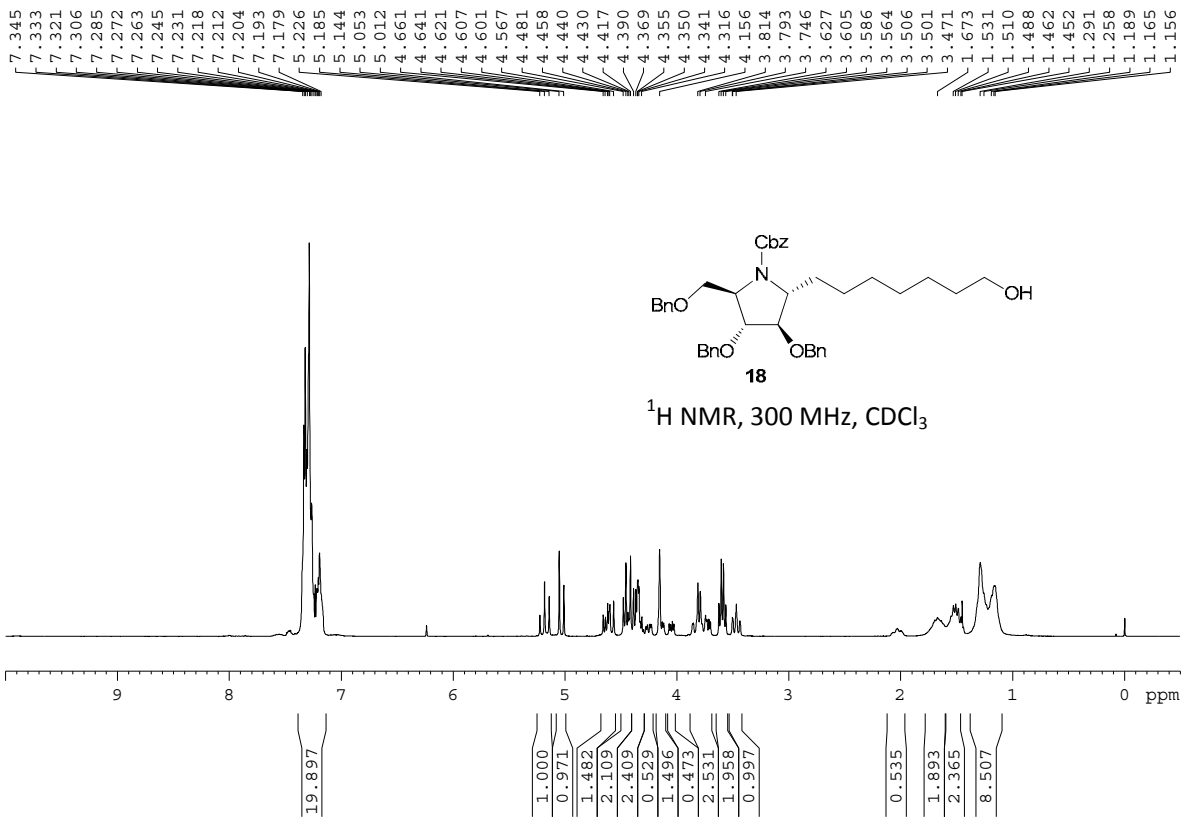


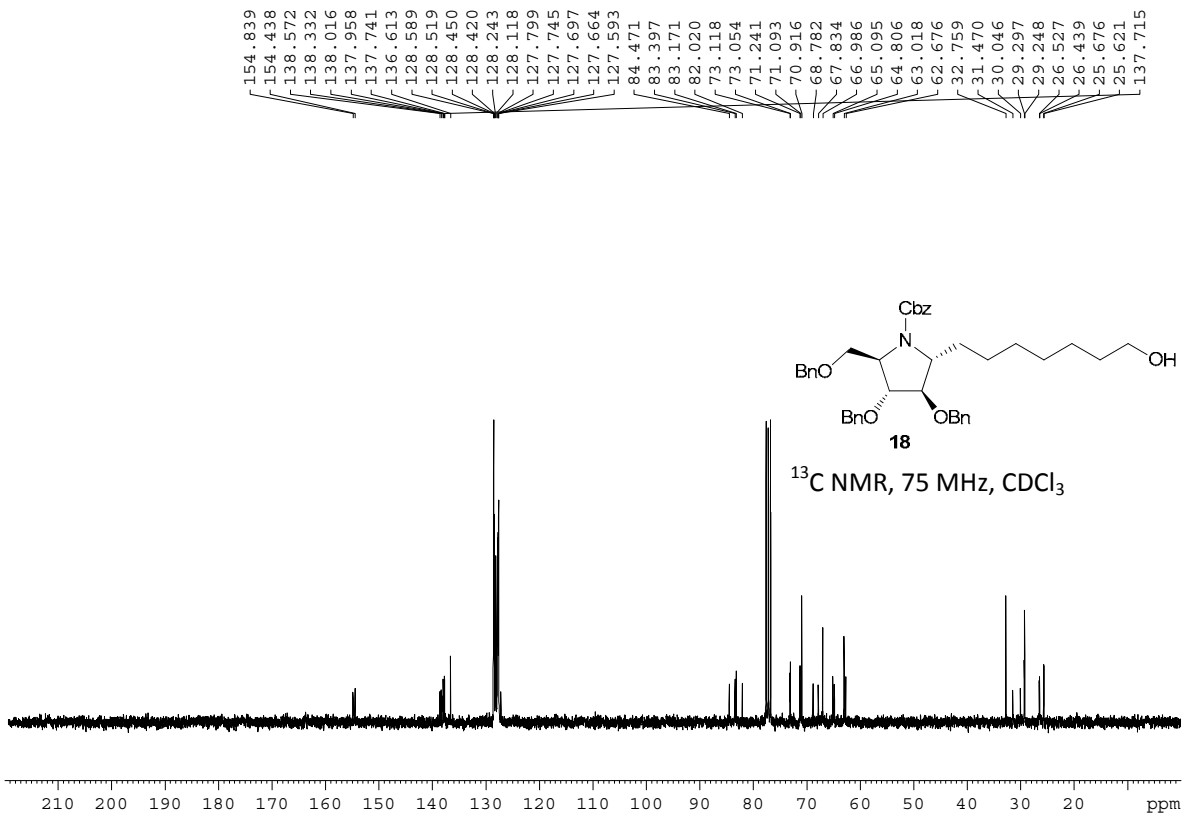

7.335
7.322
7.301
7.288
7.266
7.225
7.214
7.200
7.188
7.175
5.779
5.759
5.216
5.191
5.176
5.151
5.046
5.021
4.993
4.977
4.959
4.934
4.914
4.651
4.628
4.605
4.573
4.474
4.452
4.427
4.396
4.370
4.355
4.344
4.329
4.157
4.133
3.855
3.832
3.814
3.792
3.776
3.753
3.722
3.720
3.491
3.487
3.475
2.012
2.000
1.957
1.943
1.353
1.272
1.260
1.206
1.158
1.147

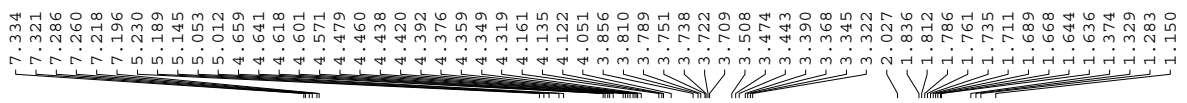

Cbz
N
BnO
BnO
OBn
17
^{1}H NMR, 500 MHz, CDCl$_3$
9
8
7
6
5
4
3
2
1
0
ppm
19.889
0.951
1.022
1.007
1.957
1.527
2.154
2.410
0.515
1.520
0.500
2.524
1.000
2.545
1.745
6.429

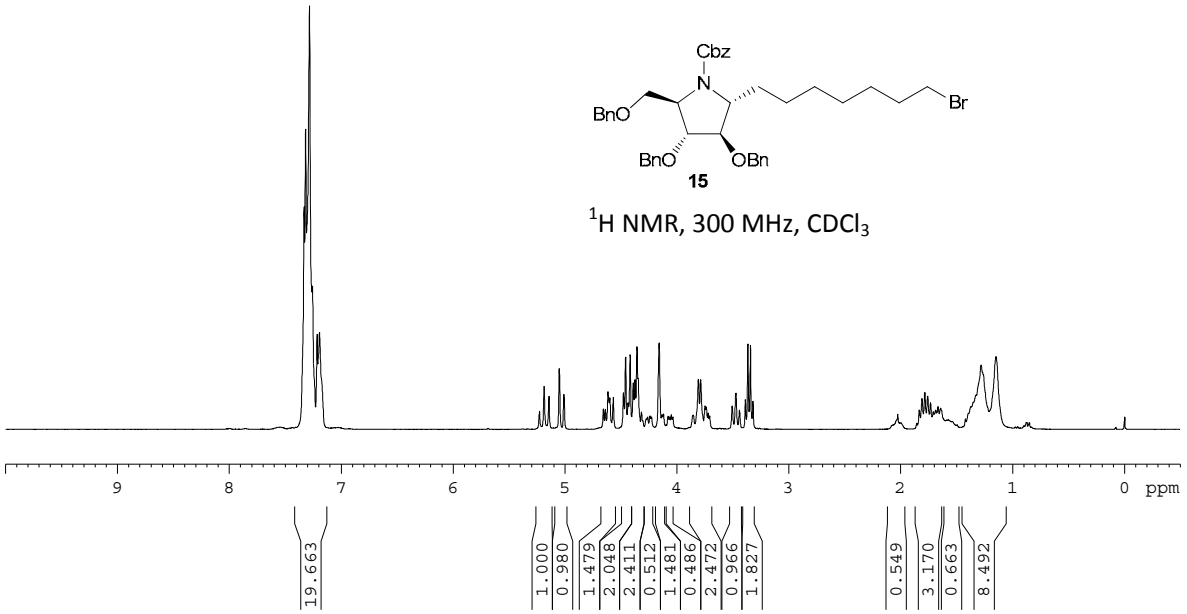

154.757
154.328
139.186
139.078
138.645
138.400
138.059
138.002
137.789
137.752
136.741
136.694
128.594
128.530
128.464
128.427
128.256
128.168
128.091
127.865
127.847
127.799
127.755
127.718
127.671
127.610
114.388
114.341
84.493
83.415
83.233
82.080
73.131
73.064
71.266
71.115
70.936
68.850
67.898
66.953
66.893
65.091
64.766
63.010
62.680
33.791
33.739
31.465
30.170
29.001
28.939
28.771


Cbz
N
BnO
BnO
OBn
17
^{13}C NMR, 126 MHz, CDCl$_3$
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

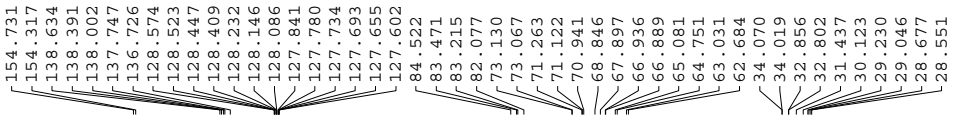

7.345
7.333
7.321
7.306
7.285
7.272
7.263
7.245
7.231
7.218
7.212
7.204
7.193
7.179
5.226
5.185
5.144
5.053
5.012
4.661
4.641
4.621
4.607
4.601
4.567
4.481
4.458
4.440
4.430
4.417
4.390
4.369
4.355
4.350
4.341
4.316
4.156
3.814
3.793
3.746
3.627
3.605
3.586
3.564
3.506
3.501
3.471
1.673
1.531
1.510
1.488
1.462
1.452
1.291
1.258
1.189
1.165
1.156
Cbz
N
BnO
OH
BnO
OBn
18
1H NMR, 300 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0 ppm
19.897
1.000
0.971
1.482
2.109
2.409
0.529
1.496
0.473
2.531
1.958
0.997
0.535
1.893
2.365
8.507

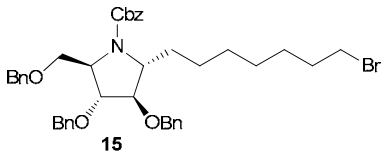

154.839
154.438
138.572
138.332
138.016
137.958
137.741
136.613
128.589
128.519
128.450
128.420
128.243
128.118
127.799
127.745
127.697
127.664
127.593
84.471
83.397
83.171
82.020
73.118
73.054
71.241
71.093
70.916
68.782
67.834
66.986
65.095
64.806
63.018
62.676
32.759
31.470
30.046
29.297
29.248
26.527
26.439
25.676
25.621
137.715
Cbz
N
BnO
OH
BnO
OBn
18
13C NMR, 75 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm



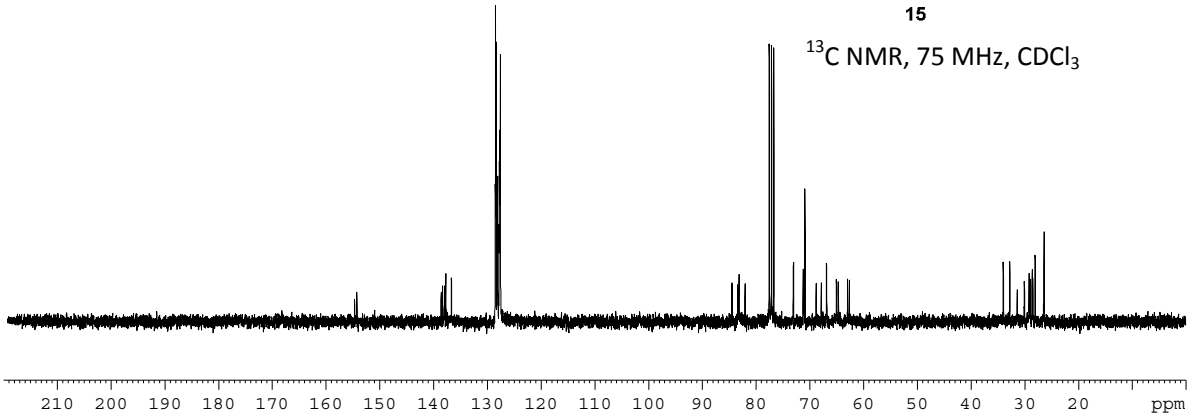


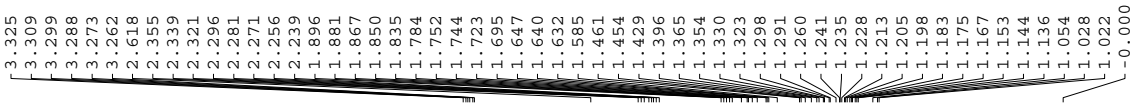
^{1}H NMR, 300 MHz, $CDCl_3$

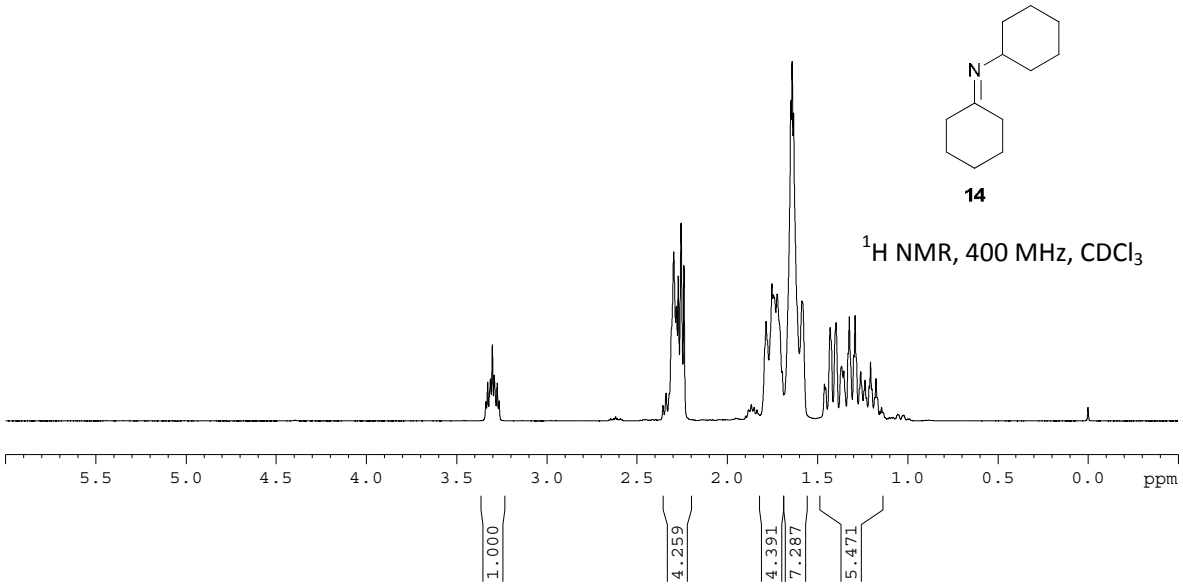




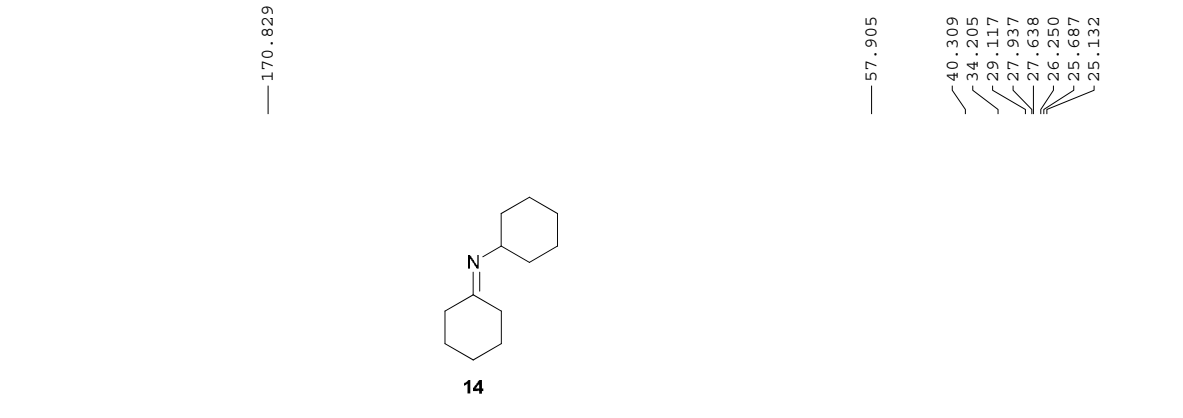
^{13}C NMR, 75 MHz, $CDCl_3$



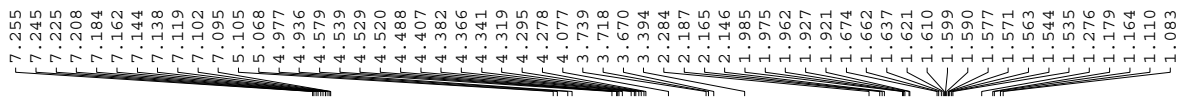


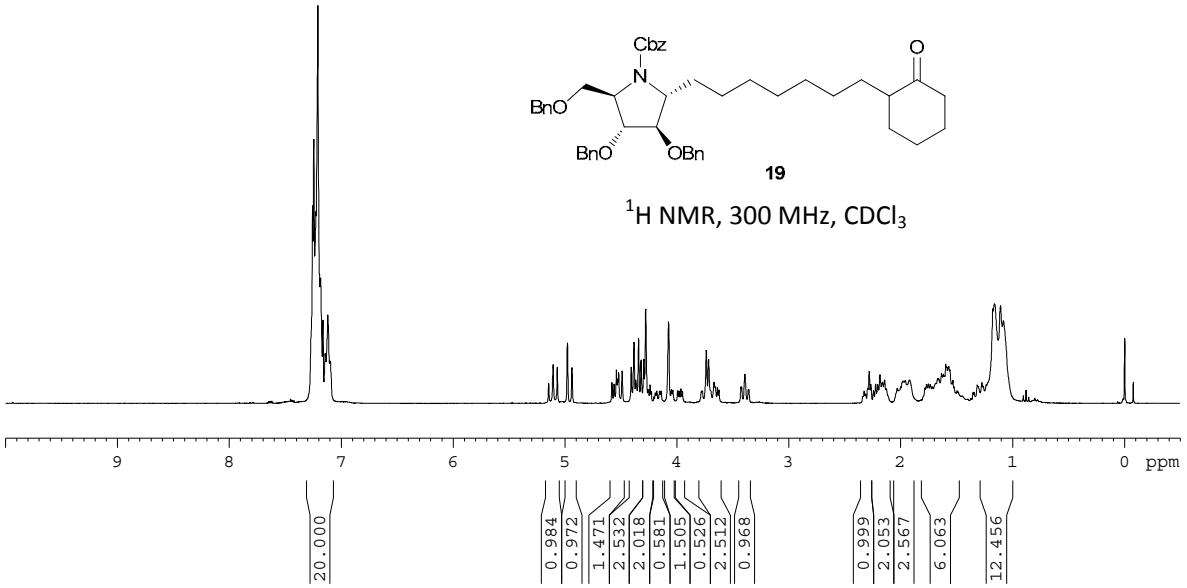


^{1}H NMR, 400 MHz, $CDCl_3$



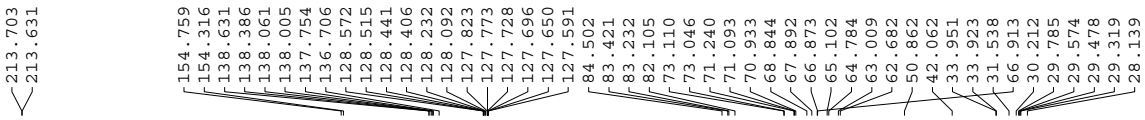
^{13}C NMR, 101 MHz, $CDCl_3$

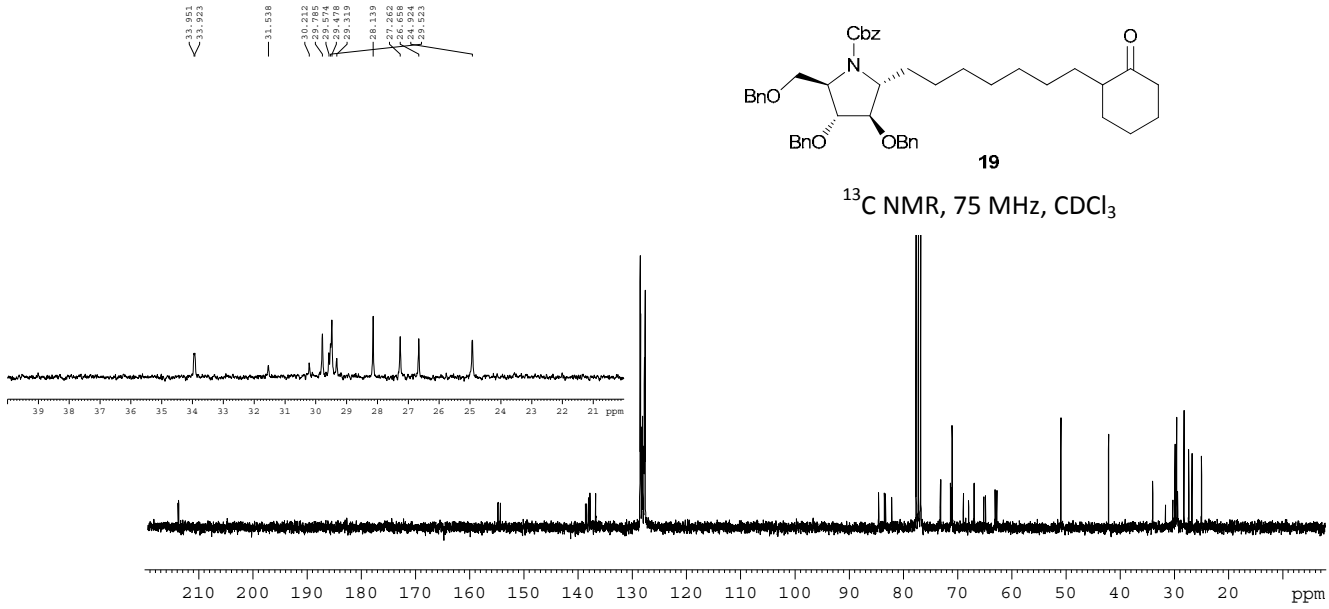




19

^{1}H NMR, 300 MHz, $CDCl_3$



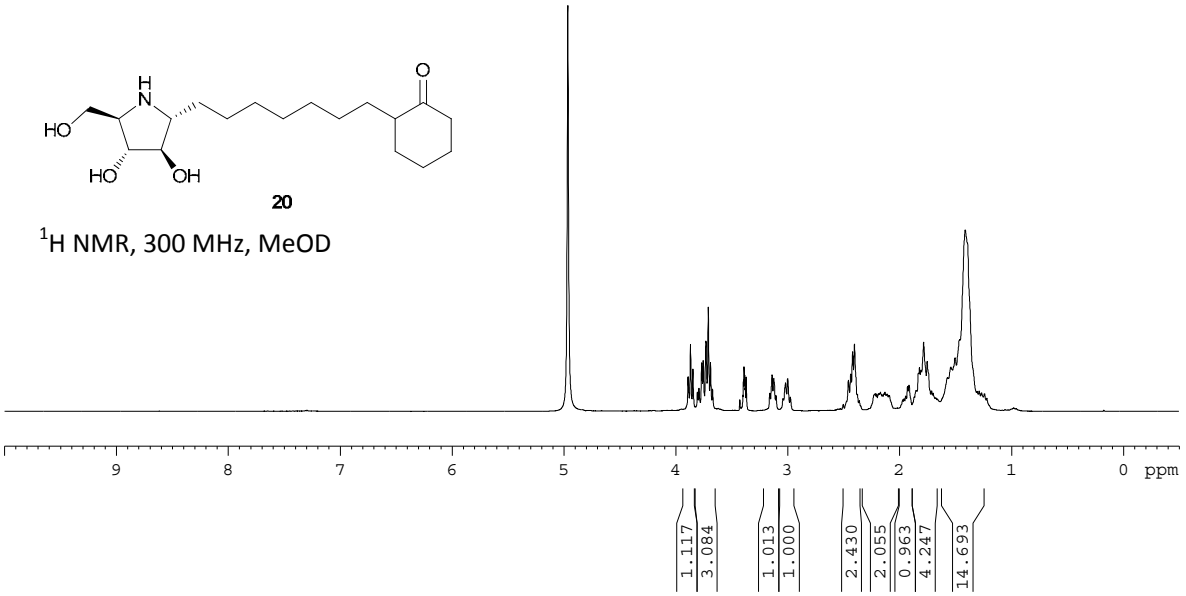


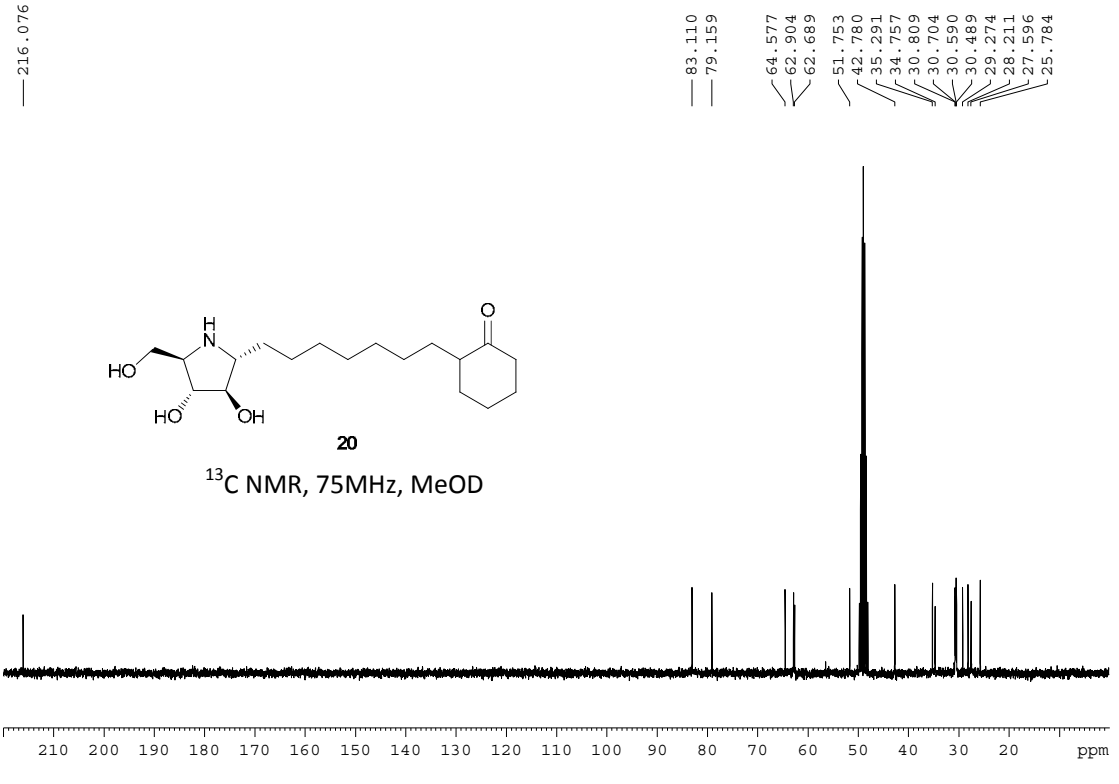
19

^{13}C NMR, 75 MHz, $CDCl_3$

20

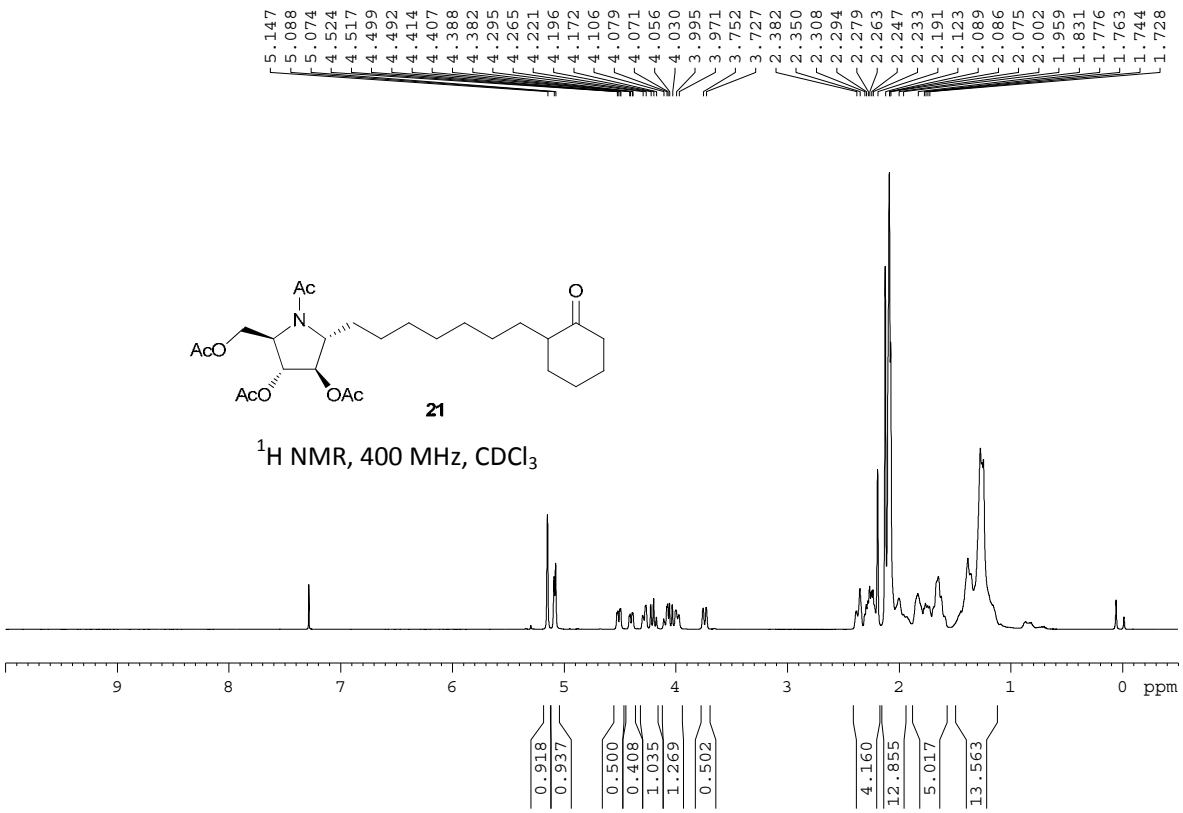
^{1}H NMR, 300 MHz, MeOD

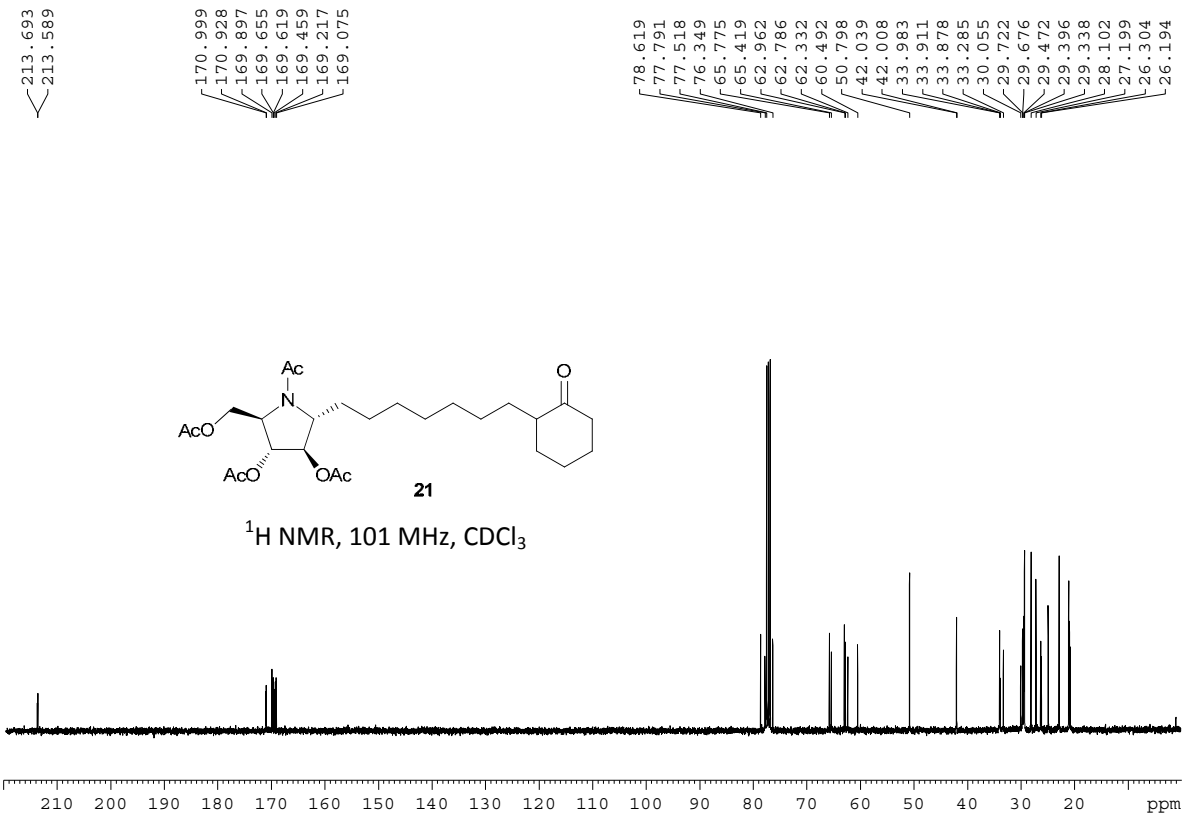


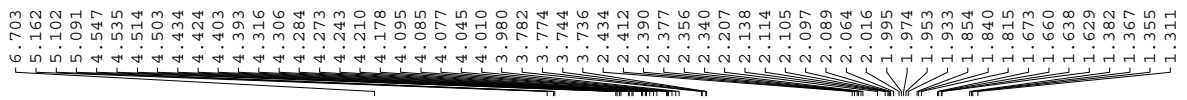


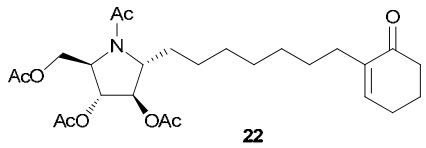
20

^{13}C NMR, 75MHz, MeOD


5.147
5.088
5.074
4.524
4.517
4.499
4.492
4.414
4.407
4.388
4.382
4.295
4.265
4.221
4.196
4.172
4.106
4.079
4.071
4.056
4.030
3.995
3.971
3.752
3.727
2.382
2.350
2.308
2.294
2.279
2.263
2.247
2.233
2.191
2.123
2.089
2.086
2.075
2.002
1.959
1.831
1.776
1.763
1.744
1.728
Ac
N
AcO
AcO
OAc
O
21
1H NMR, 400 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0 ppm
0.918
0.937
0.500
0.408
1.035
1.269
0.502
4.160
12.855
5.017
13.563

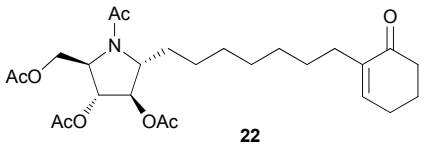

213.693
213.589
170.999
170.928
169.897
169.655
169.619
169.459
169.217
169.075
78.619
77.791
77.518
76.349
65.775
65.419
62.962
62.786
62.332
60.492
50.798
42.039
42.008
33.983
33.911
33.878
33.285
30.055
29.722
29.676
29.472
29.396
29.338
28.102
27.199
26.304
26.194
Ac
N
AcO
AcO
OAc
O
21
1H NMR, 101 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm





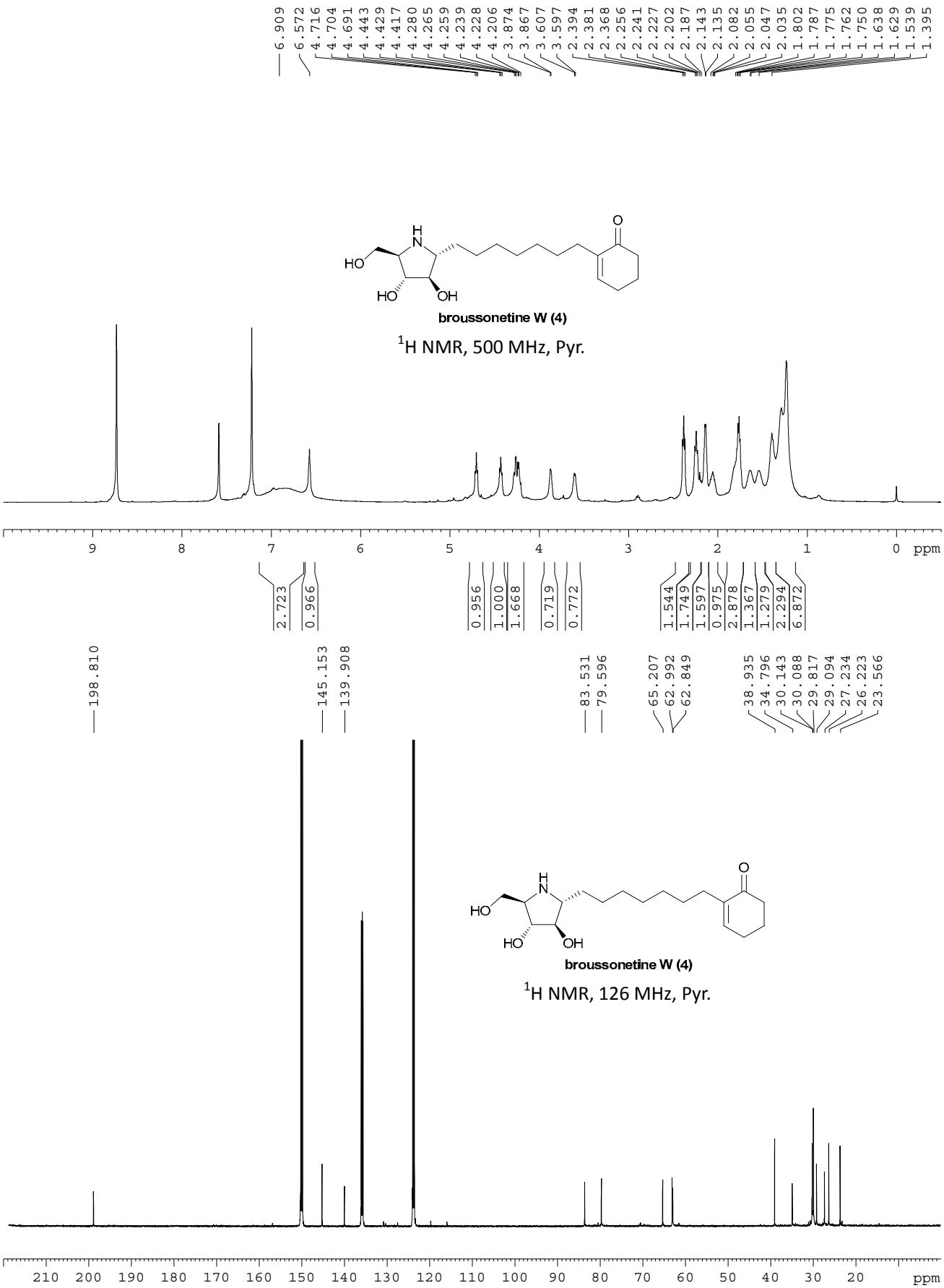
22

^{1}H NMR, 300 MHz, $CDCl_3$

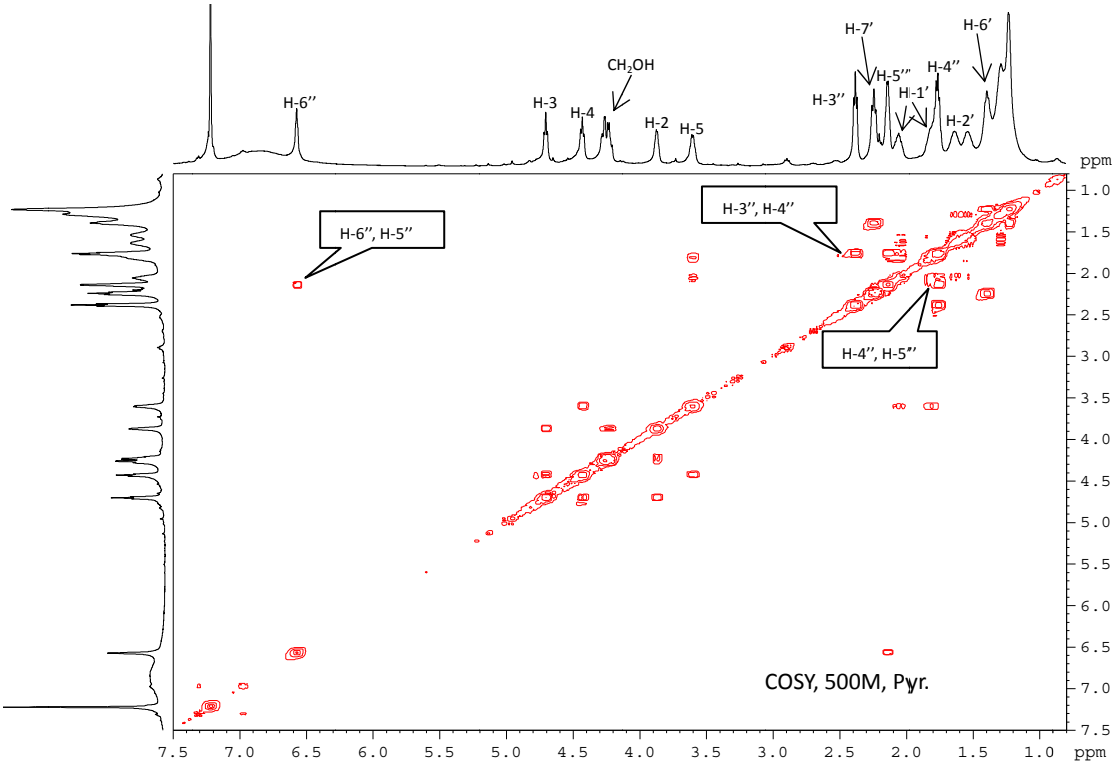


22

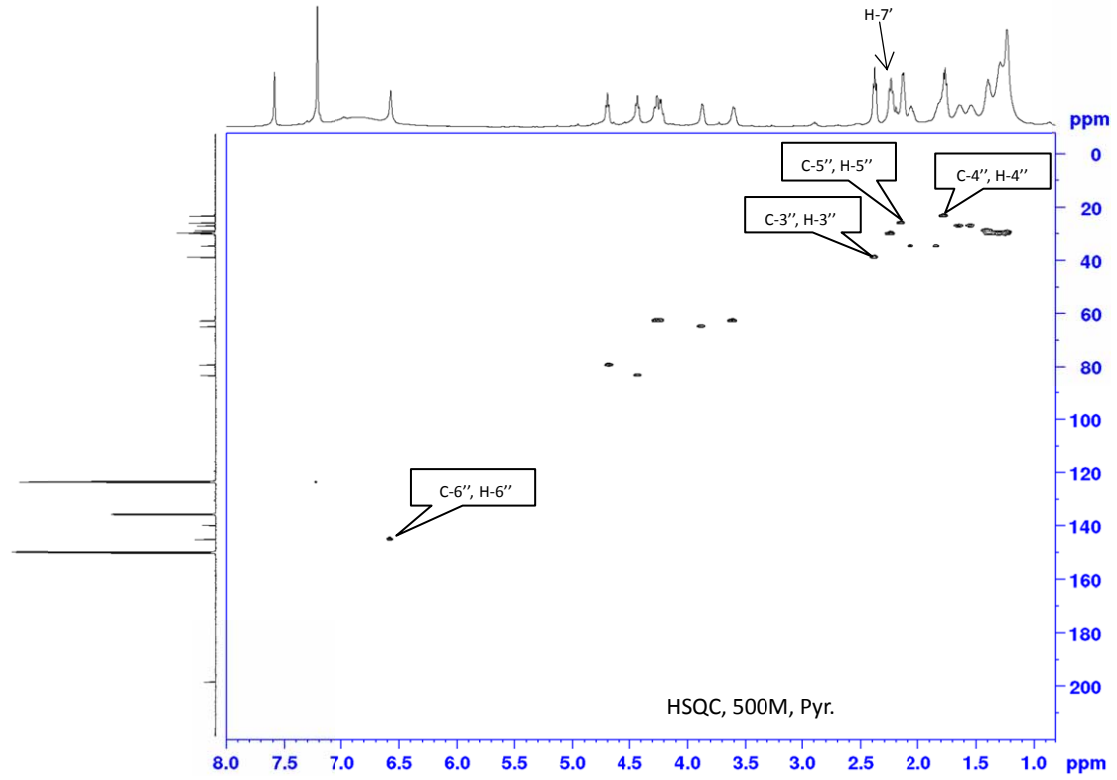
^{13}C NMR, 75 MHz, $CDCl_3$


6.909
6.572
4.716
4.704
4.691
4.443
4.429
4.417
4.280
4.265
4.259
4.239
4.228
4.206
3.874
3.867
3.607
3.597
2.394
2.381
2.368
2.256
2.241
2.227
2.202
2.187
2.143
2.135
2.082
2.055
2.047
2.035
1.802
1.787
1.775
1.762
1.750
1.638
1.629
1.539
1.395
HO
HO
OH
broussonetine W (4)
1H NMR, 500 MHz, Pyr.
9
8
7
6
5
4
3
2
1
0
ppm
2.723
0.966
0.956
1.000
1.668
0.719
0.772
1.544
1.749
1.597
0.975
2.878
1.367
1.279
2.294
6.872
198.810
145.153
139.908
83.531
79.596
65.207
62.992
62.849
38.935
34.796
30.143
30.088
29.817
29.094
27.234
26.223
23.566
HO
HO
OH
broussonetine W (4)
1H NMR, 126 MHz, Pyr.
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

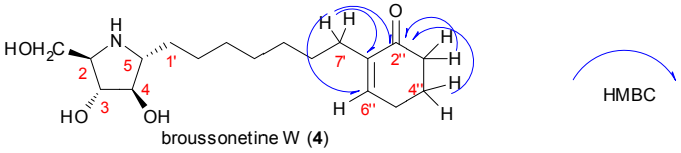
COSY of the synthetic broussonetine W (**4**)



HSQC of the synthetic broussontine W (**4**)

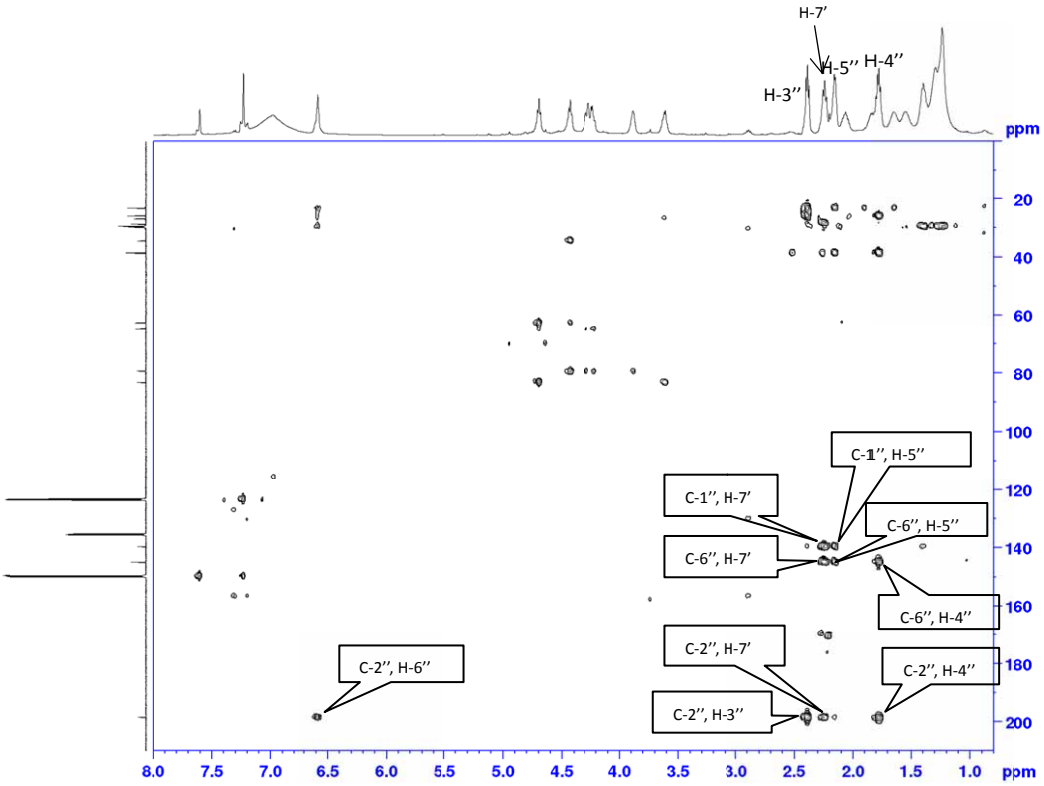


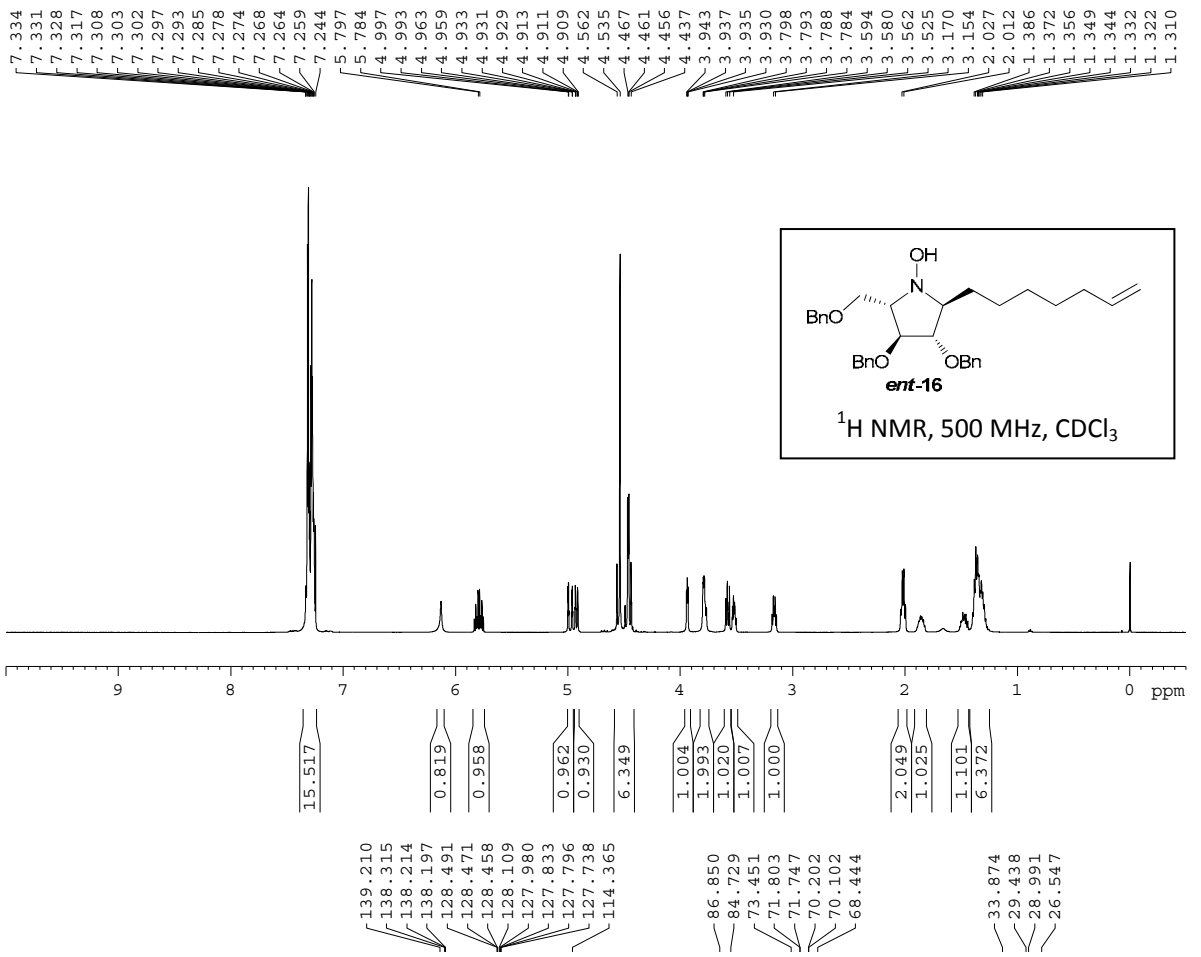
HMBC of the synthetic broussonetine W (**4**)

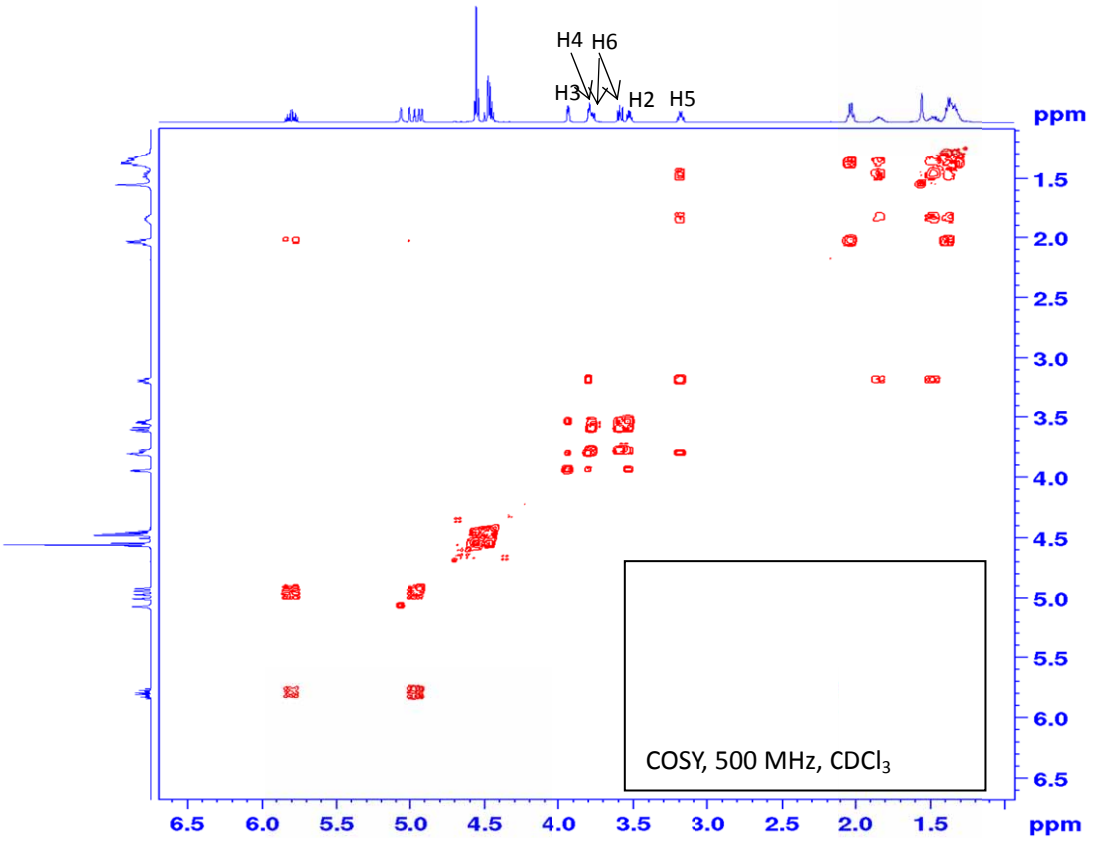


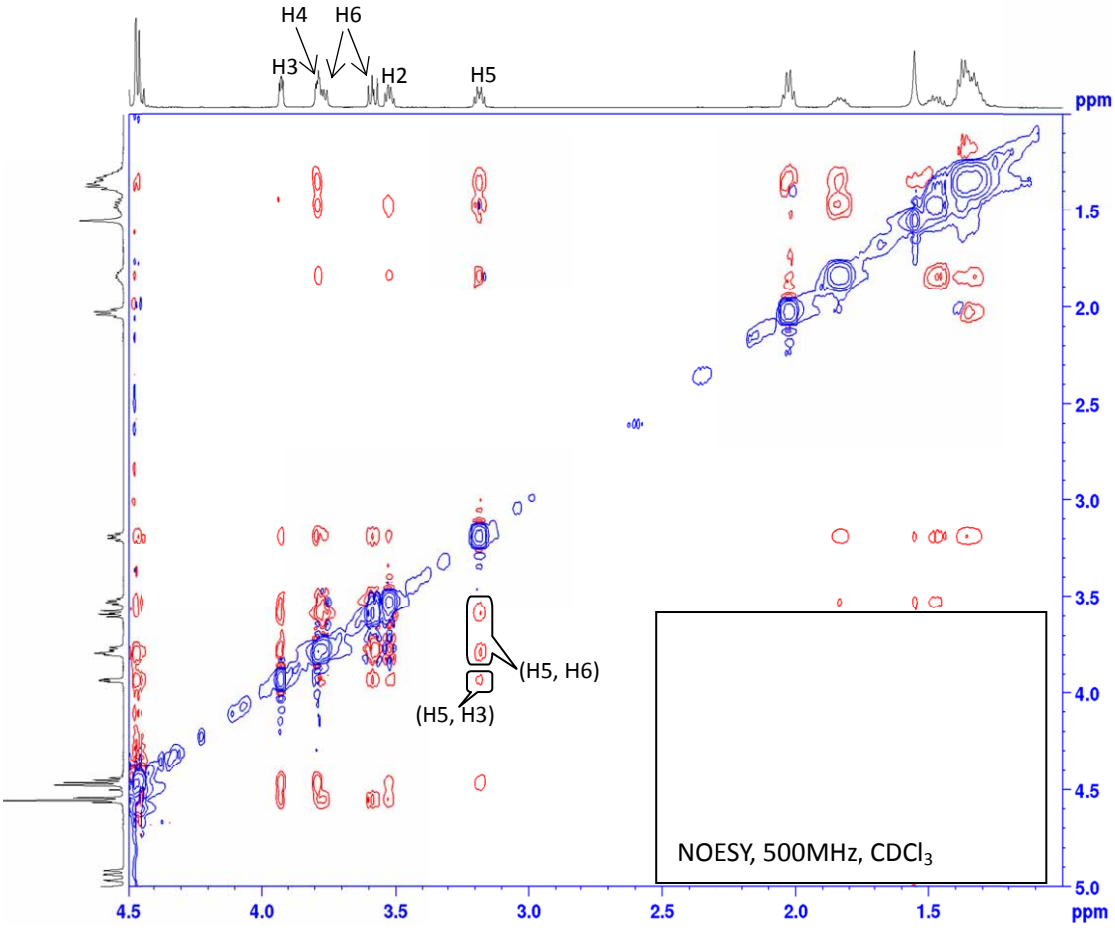
broussonetine W (**4**)

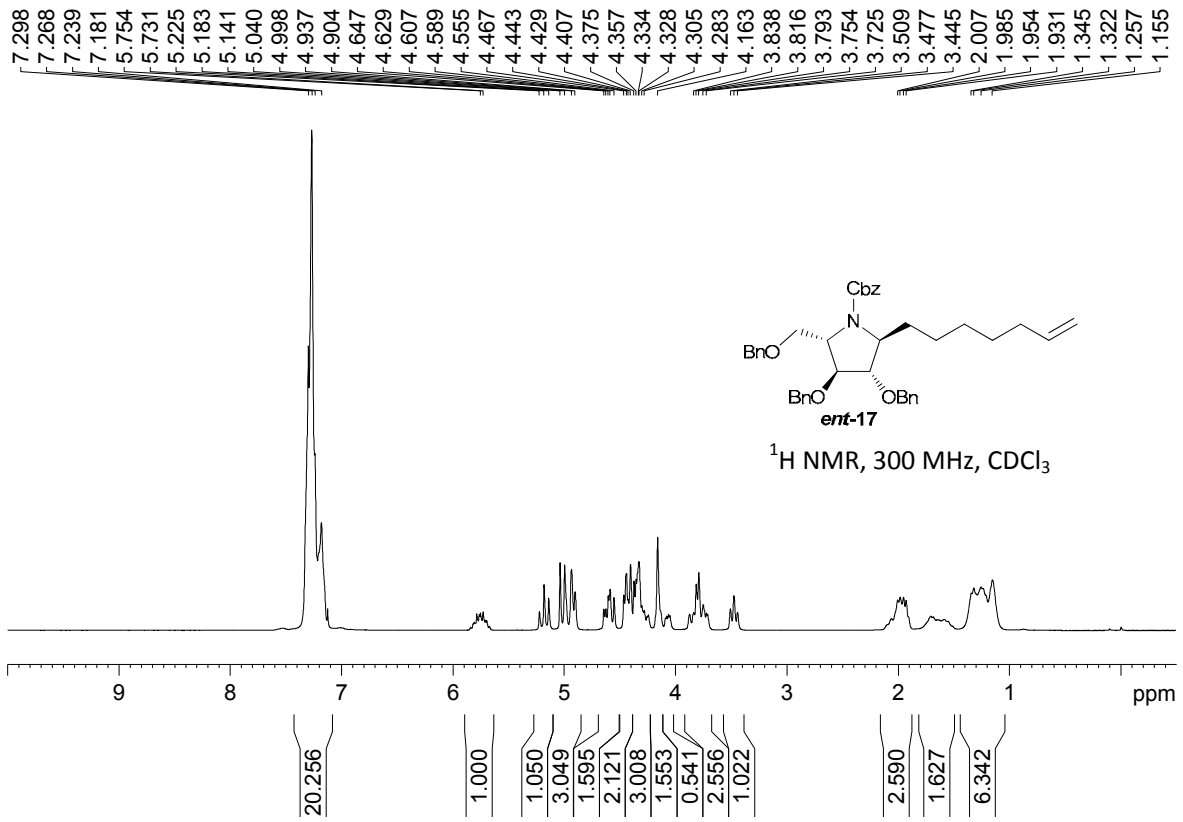
HMBC, 500M, Pyr.

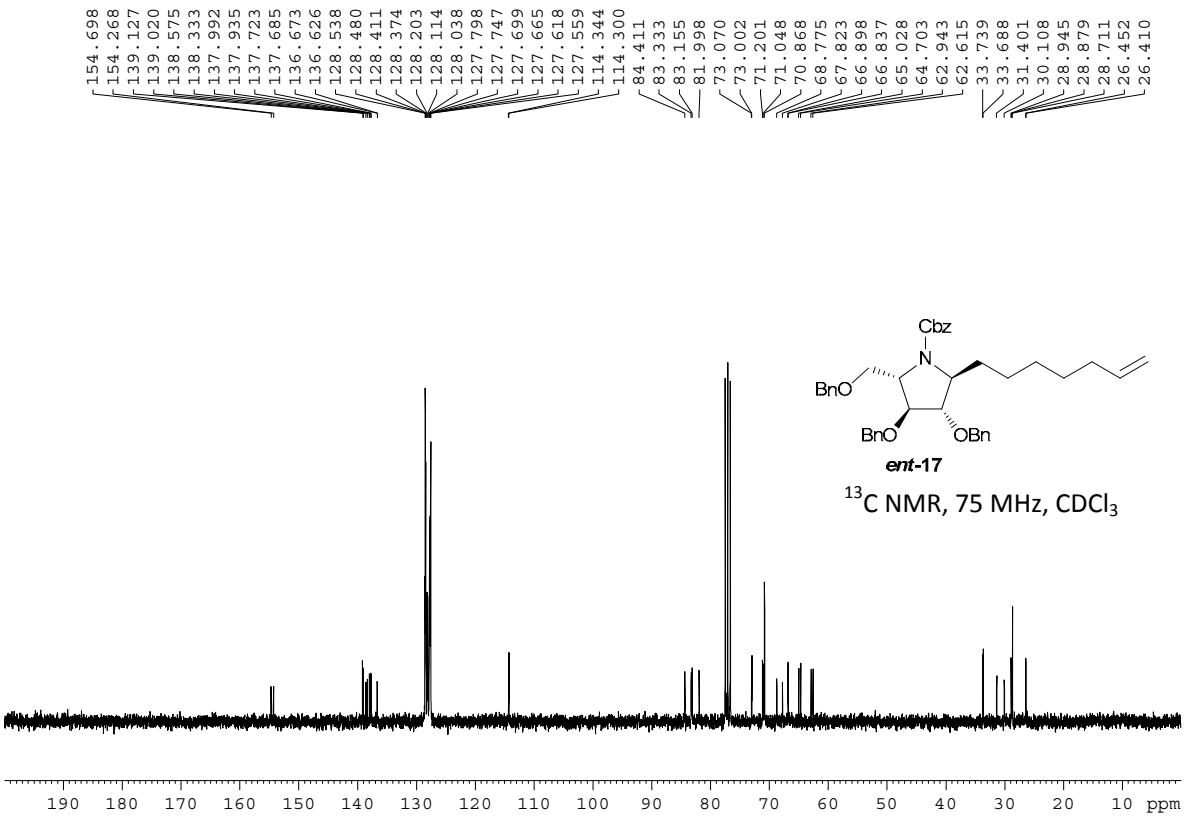


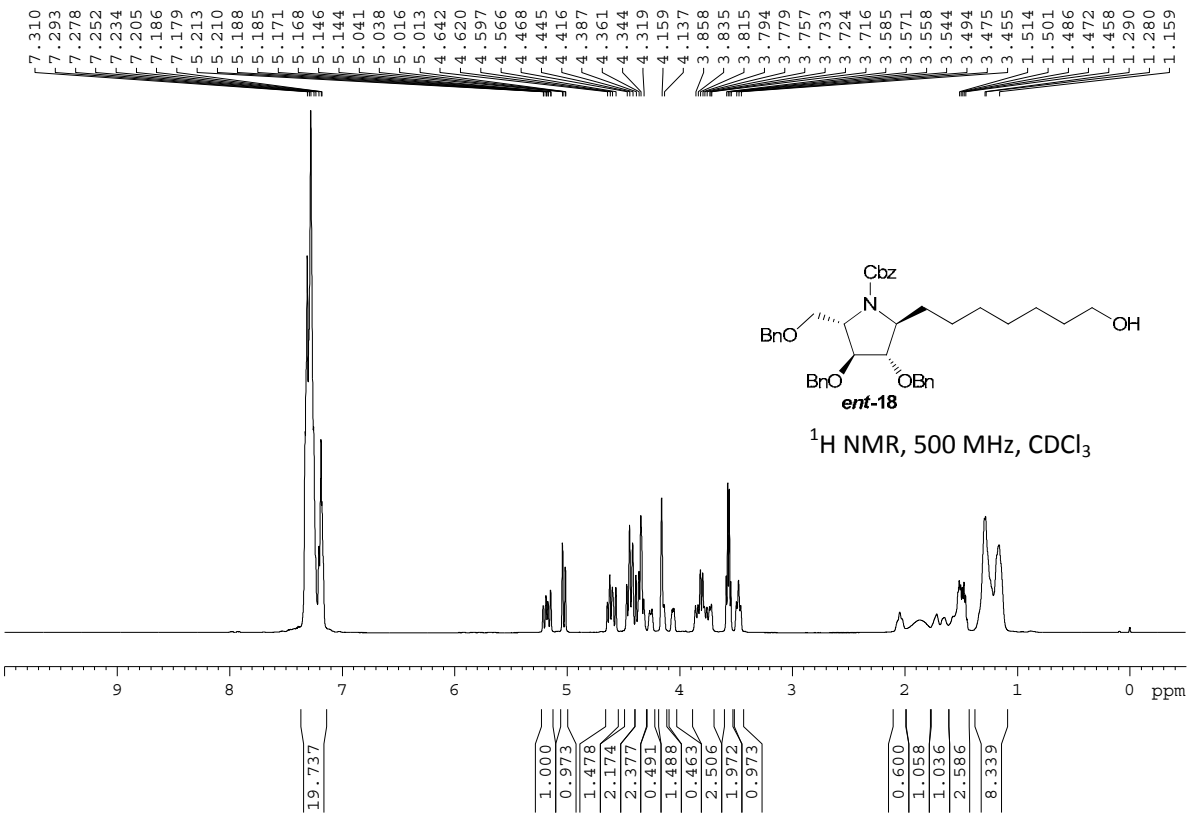

7.334
7.331
7.328
7.317
7.308
7.303
7.302
7.297
7.293
7.285
7.278
7.274
7.268
7.264
7.259
7.244
5.797
5.784
4.997
4.993
4.963
4.959
4.933
4.931
4.929
4.913
4.911
4.909
4.562
4.535
4.467
4.461
4.456
4.437
3.943
3.937
3.935
3.930
3.798
3.793
3.788
3.784
3.594
3.580
3.562
3.525
3.170
3.154
2.027
2.012
1.386
1.372
1.356
1.349
1.344
1.332
1.322
1.310
OH
N
BnO
BnO
OBn
ent-16
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
15.517
0.819
0.958
0.962
0.930
6.349
1.004
1.993
1.020
1.007
1.000
2.049
1.025
1.101
6.372
139.210
138.315
138.214
138.197
128.491
128.471
128.458
128.109
127.980
127.833
127.796
127.738
114.365
86.850
84.729
73.451
71.803
71.747
70.202
70.102
68.444
33.874
29.438
28.991
26.547
OH
N
BnO
BnO
OBn
ent-16
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

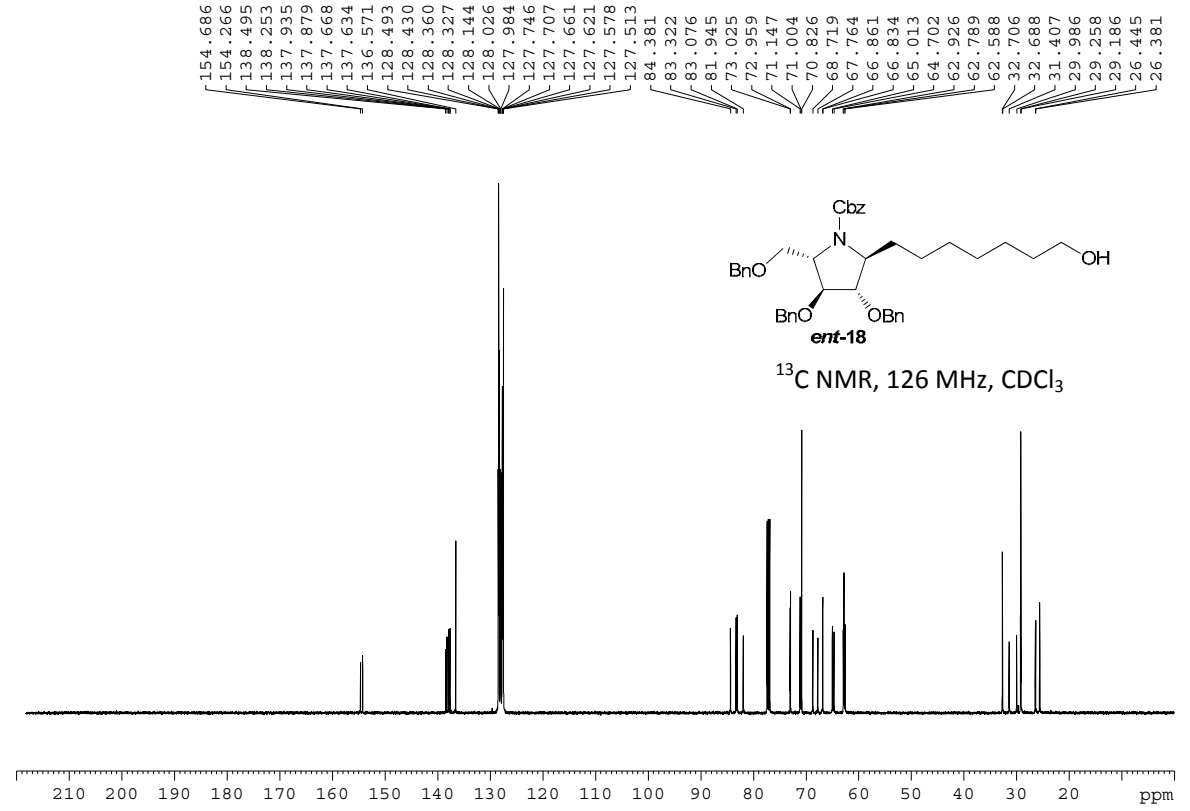

H4
H6
H3
H2
H5
ppm
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
5.5
6.0
6.5
COSY, 500 MHz, CDCl3

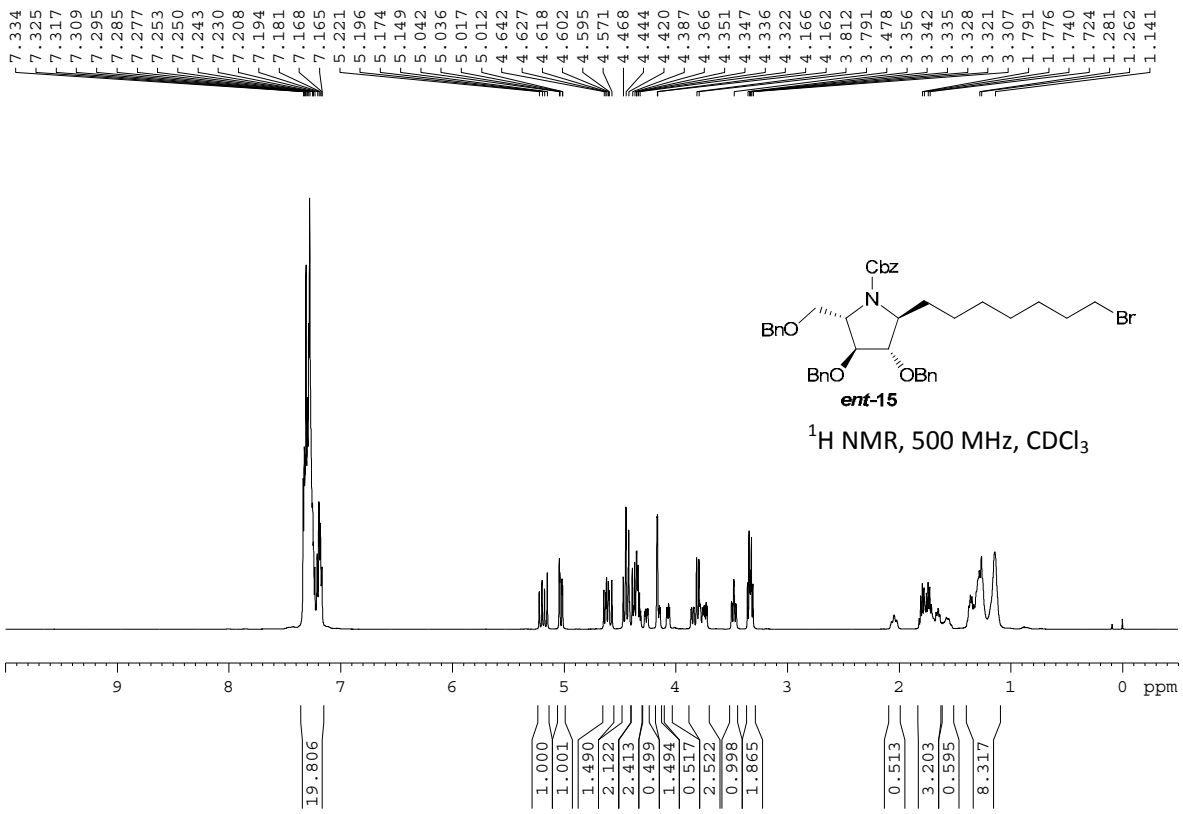

H4
H6
H3
H2
H5
ppm
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
(H5, H6)
(H5, H3)
NOESY, 500MHz, CDCl3


7.298
7.268
7.239
7.181
5.754
5.731
5.225
5.183
5.141
5.040
4.998
4.937
4.904
4.647
4.629
4.607
4.589
4.555
4.467
4.443
4.429
4.407
4.375
4.357
4.334
4.328
4.305
4.283
4.163
3.838
3.816
3.793
3.754
3.725
3.509
3.477
3.445
2.007
1.985
1.954
1.931
1.345
1.322
1.257
1.155
Cbz
N
BnO
BnO
OBn
ent-17
1H NMR, 300 MHz, CDCl3
9
8
7
6
5
4
3
2
1
ppm
20.256
1.000
1.050
3.049
1.595
2.121
3.008
1.553
0.541
2.556
1.022
2.590
1.627
6.342

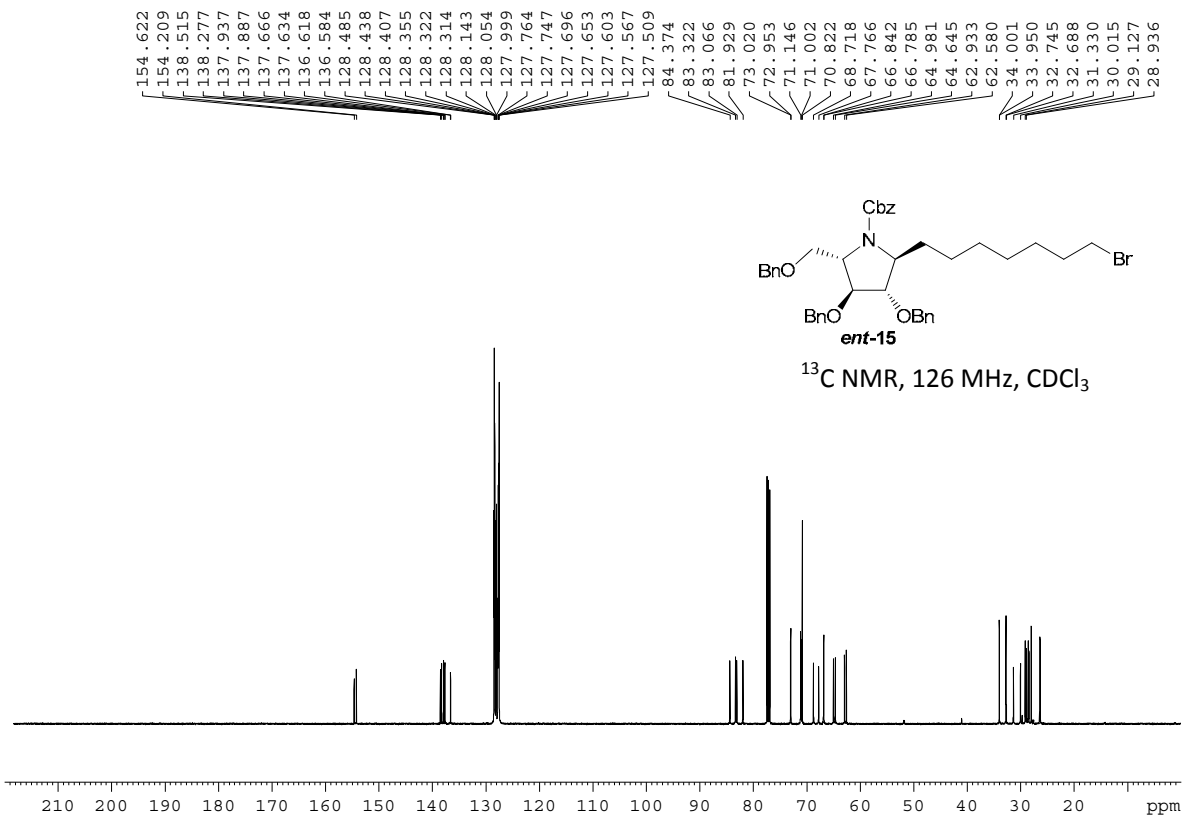

154.698
154.268
139.127
139.020
138.575
138.333
137.992
137.935
137.723
137.685
136.673
136.626
128.538
128.480
128.411
128.374
128.203
128.114
128.038
127.798
127.747
127.699
127.665
127.618
127.559
114.344
114.300
84.411
83.333
83.155
81.998
73.070
73.002
71.201
71.048
70.868
68.775
67.823
66.898
66.837
65.028
64.703
62.943
62.615
33.739
33.688
31.401
30.108
28.945
28.879
28.711
26.452
26.410
Cbz
N
BnO
BnO
OBn
ent-17
13C NMR, 75 MHz, CDCl3
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm


7.310
7.293
7.278
7.252
7.234
7.205
7.186
7.179
5.213
5.210
5.188
5.185
5.171
5.168
5.146
5.144
5.041
5.038
5.016
5.013
4.642
4.620
4.597
4.566
4.468
4.445
4.416
4.387
4.361
4.344
4.319
4.159
4.137
3.858
3.835
3.815
3.794
3.779
3.757
3.733
3.724
3.716
3.585
3.571
3.558
3.544
3.494
3.475
3.455
1.514
1.501
1.486
1.472
1.458
1.290
1.280
1.159
Cbz
N
BnO
OH
BnO
OBn
ent-18
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
19.737
1.000
0.973
1.478
2.174
2.377
0.491
1.488
0.463
2.506
1.972
0.973
0.600
1.058
1.036
2.586
8.339

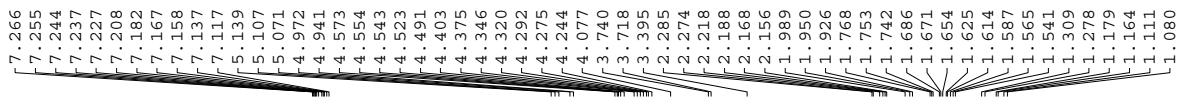

154.686
154.266
138.495
138.253
137.935
137.879
137.668
137.634
136.571
128.493
128.430
128.360
128.327
128.144
128.026
127.984
127.746
127.707
127.661
127.621
127.578
127.513
84.381
83.322
83.076
81.945
73.025
72.959
71.147
71.004
70.826
68.719
67.764
66.861
66.834
65.013
64.702
62.926
62.789
62.588
32.706
32.688
31.407
29.986
29.258
29.186
26.445
26.381
Cbz
N
BnO
OH
BnO
OBn
ent-18
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

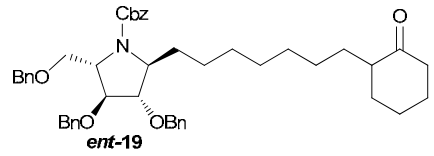


^{1}H NMR, 500 MHz, $CDCl_3$



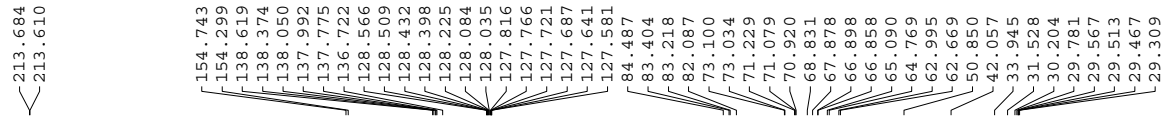
^{13}C NMR, 126 MHz, $CDCl_3$

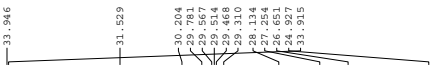


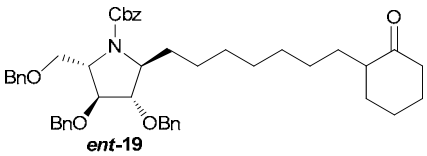


***ent*-19**

^{1}H NMR, 400 MHz, $CDCl_3$

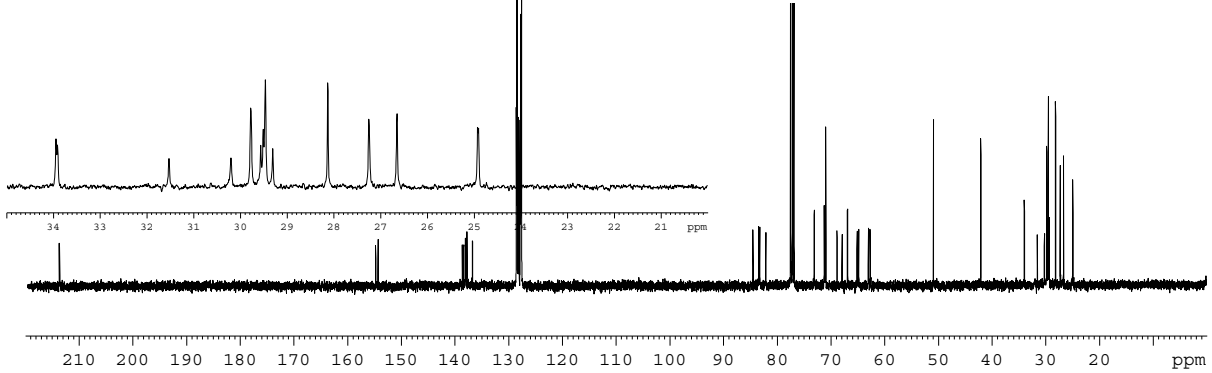


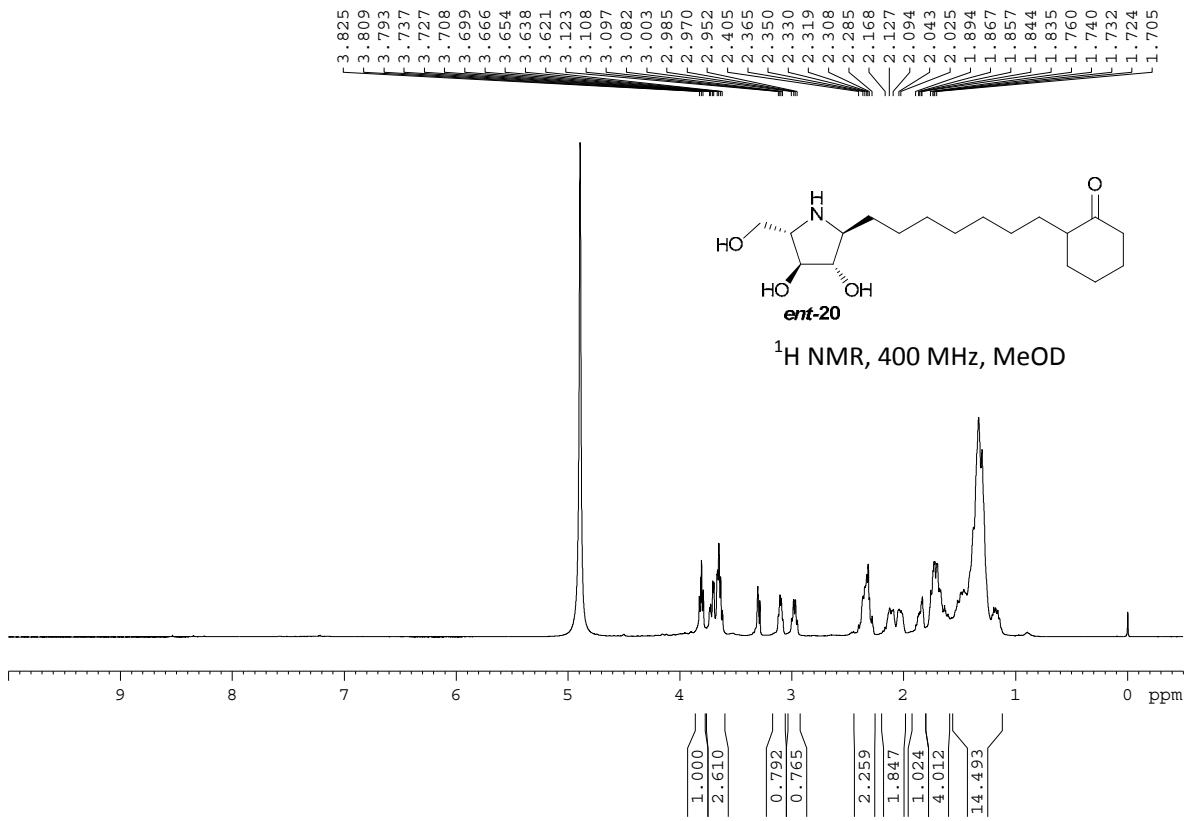




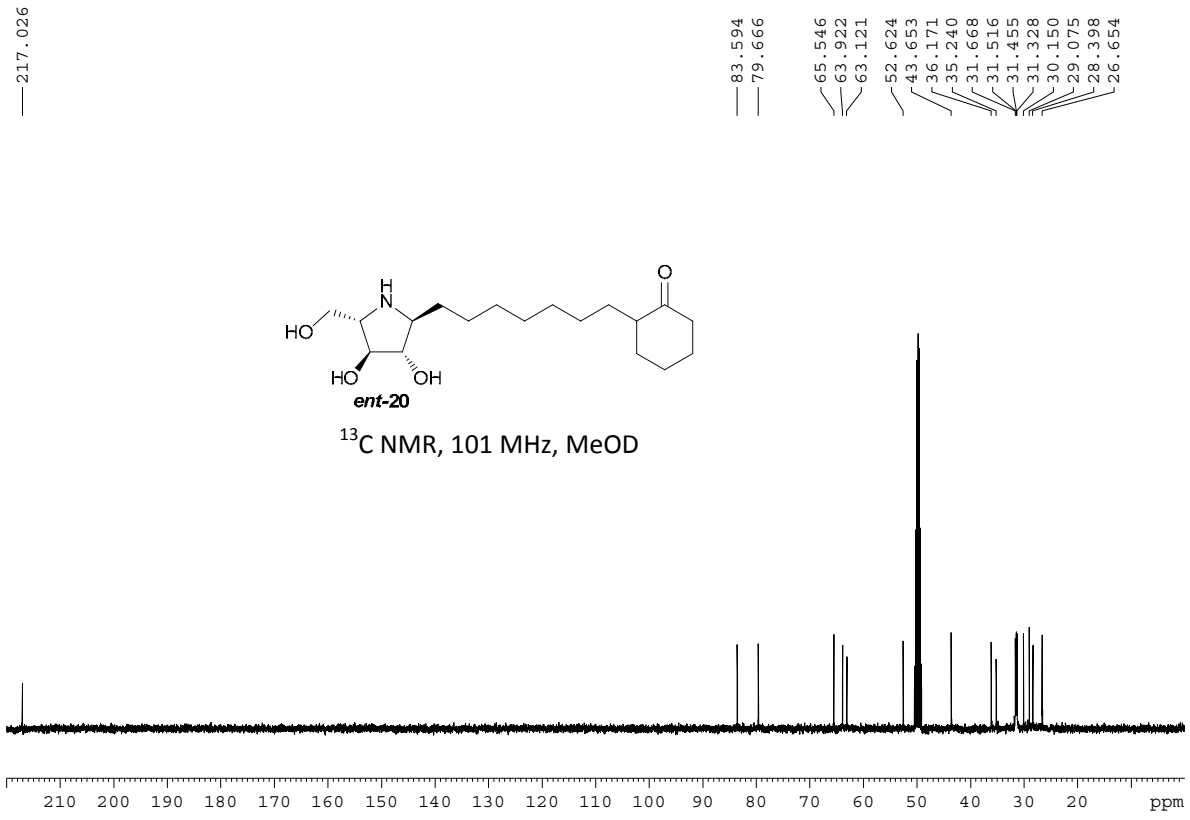
***ent*-19**

^{13}C NMR, 101 MHz, $CDCl_3$

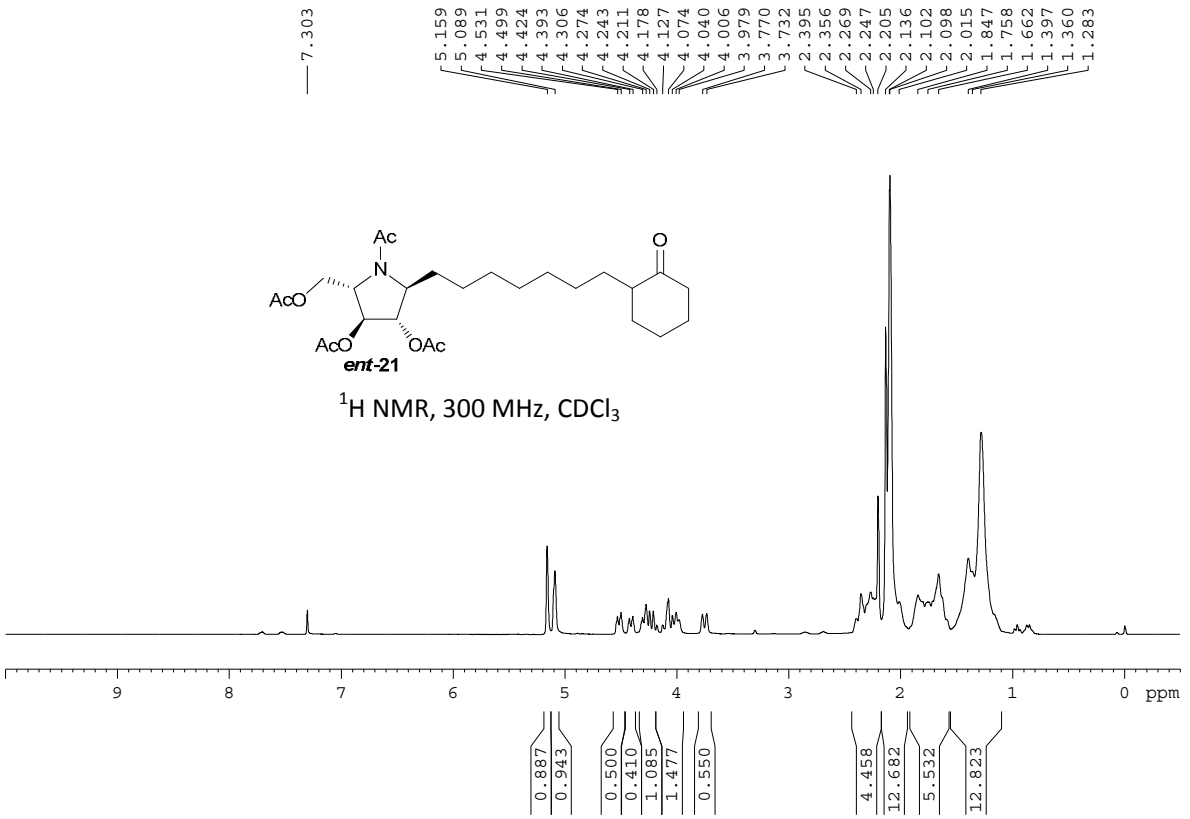




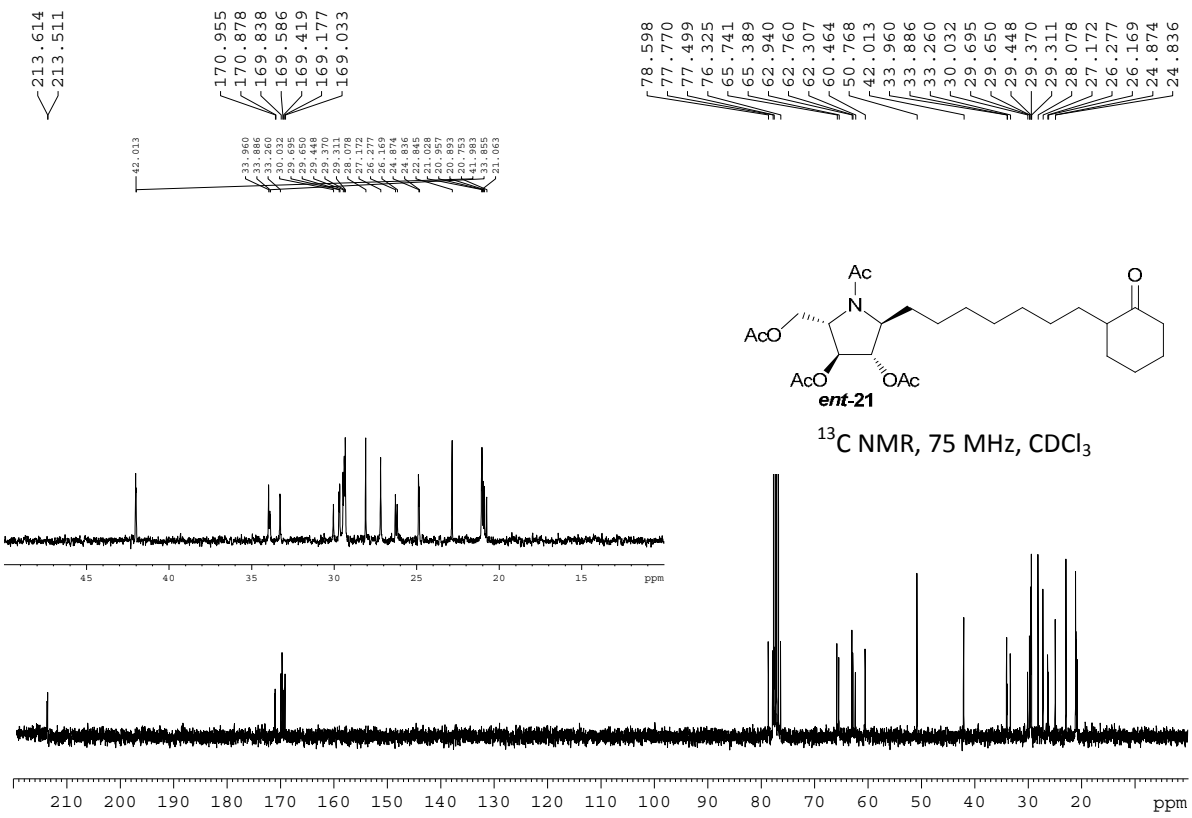
^{1}H NMR, 400 MHz, MeOD



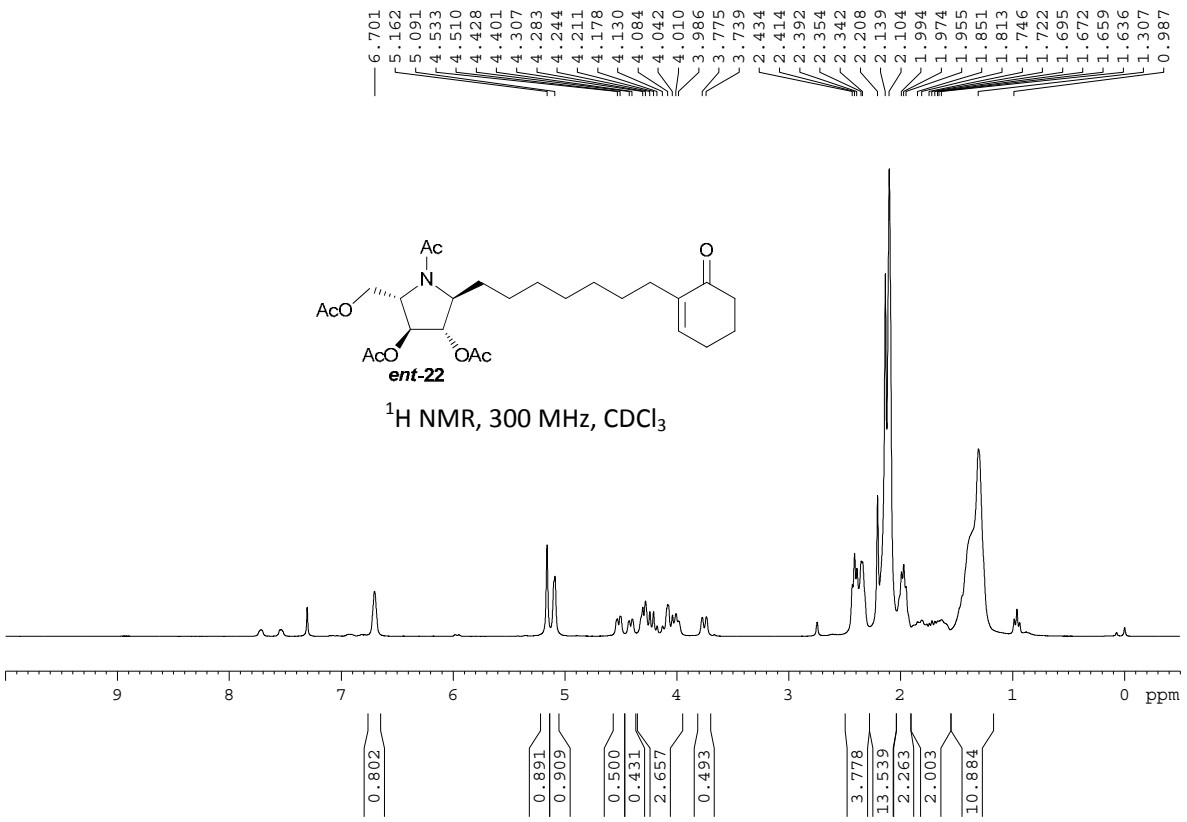
^{13}C NMR, 101 MHz, MeOD



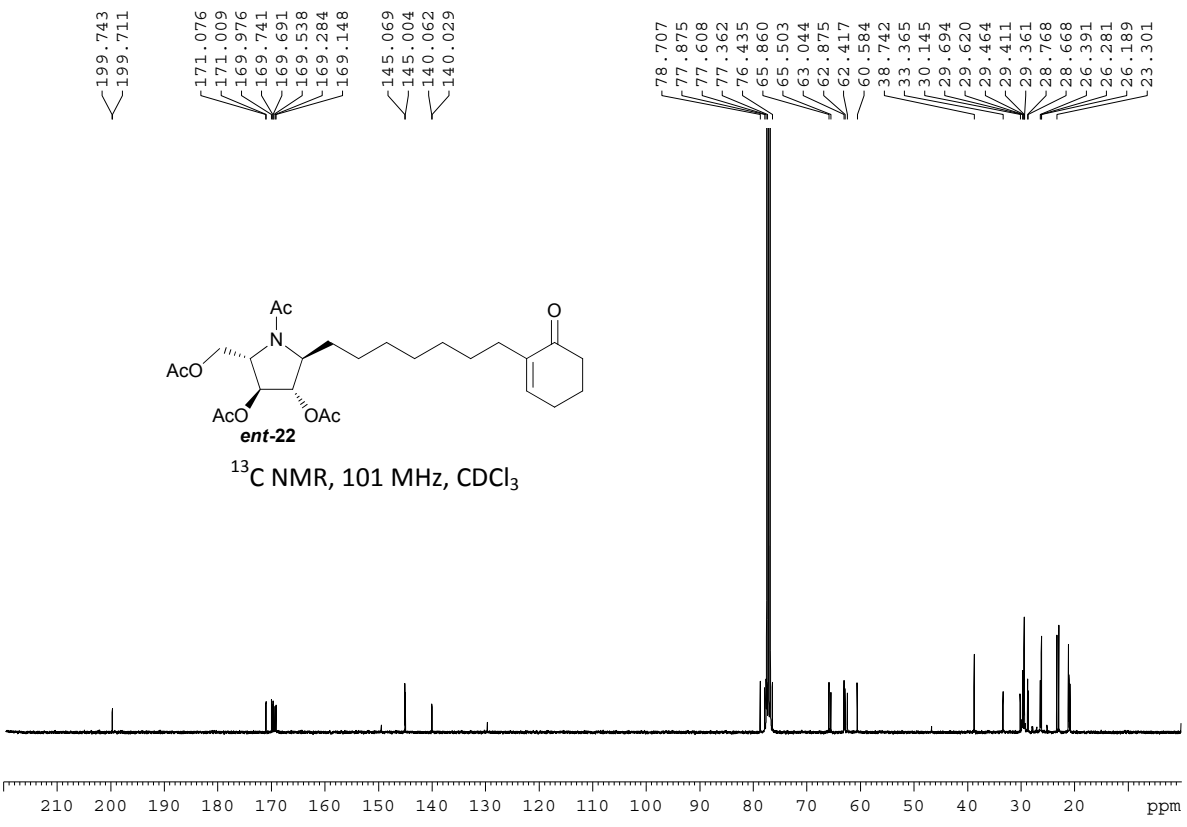
^{1}H NMR, 300 MHz, $CDCl_3$



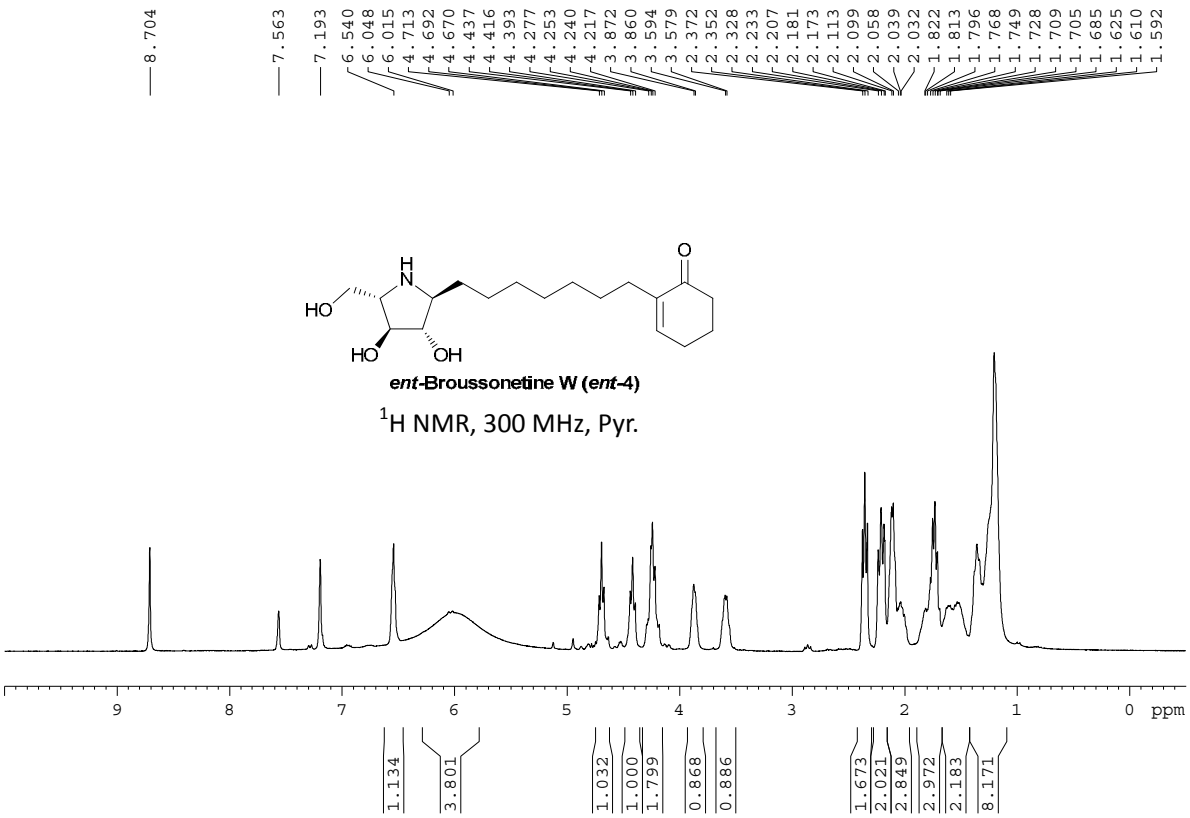
^{13}C NMR, 75 MHz, $CDCl_3$

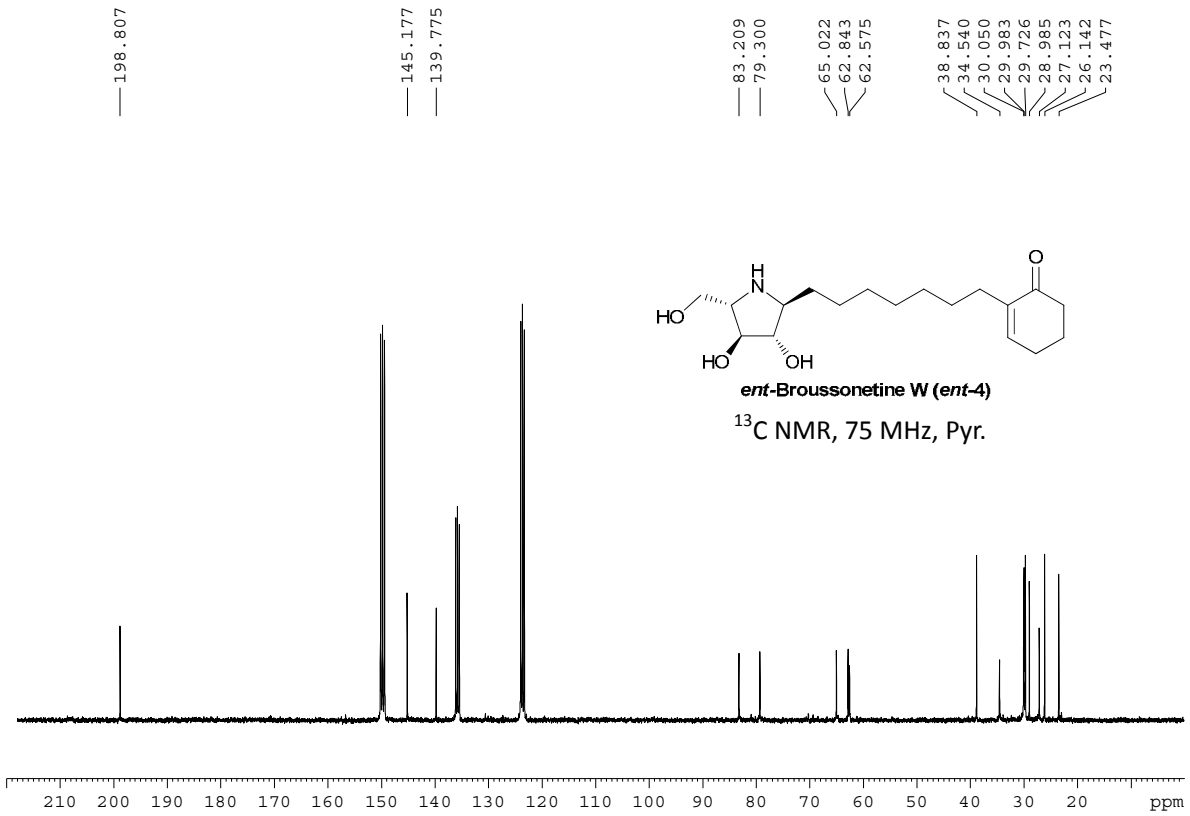


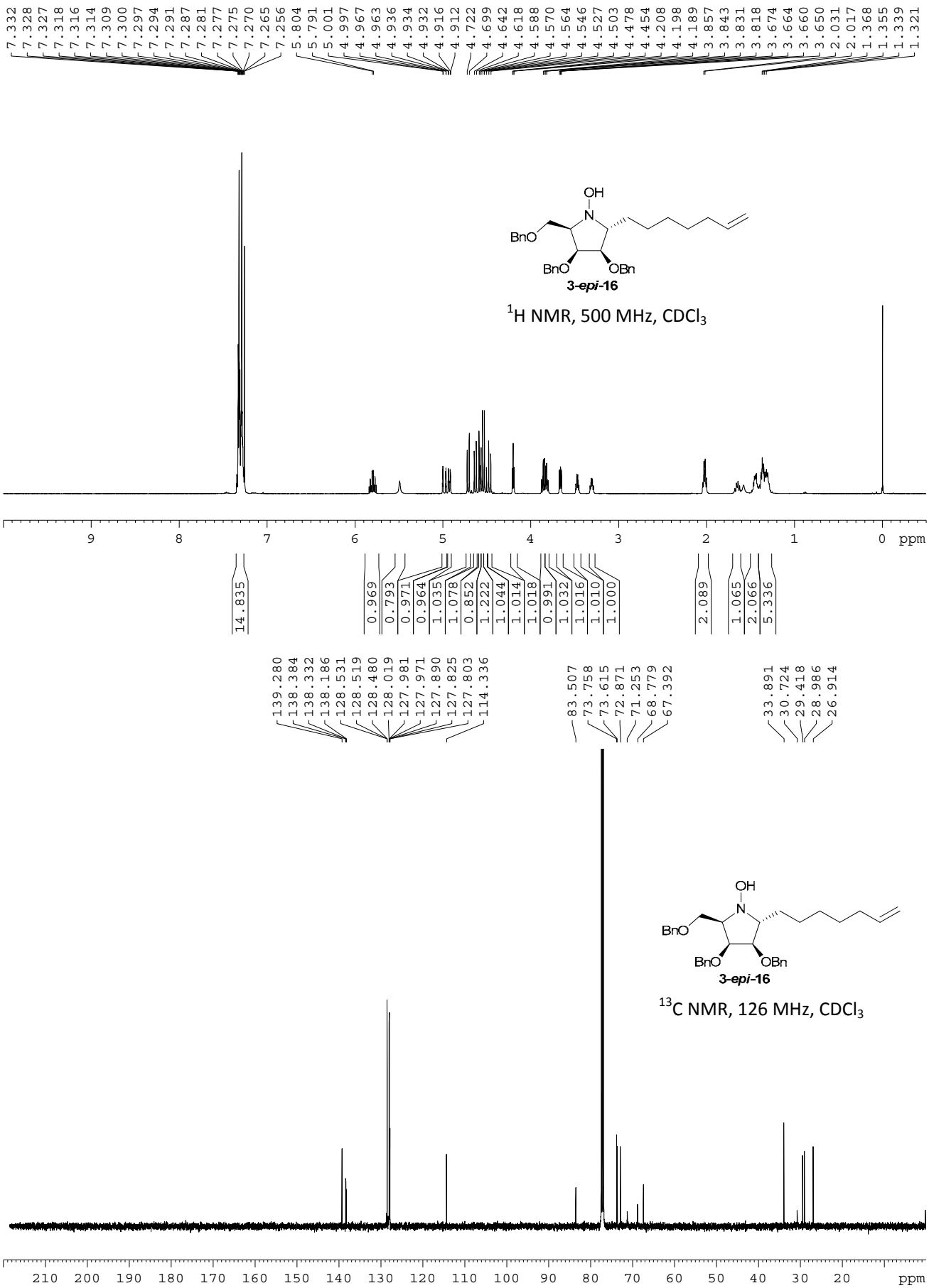
^{1}H NMR, 300 MHz, $CDCl_3$

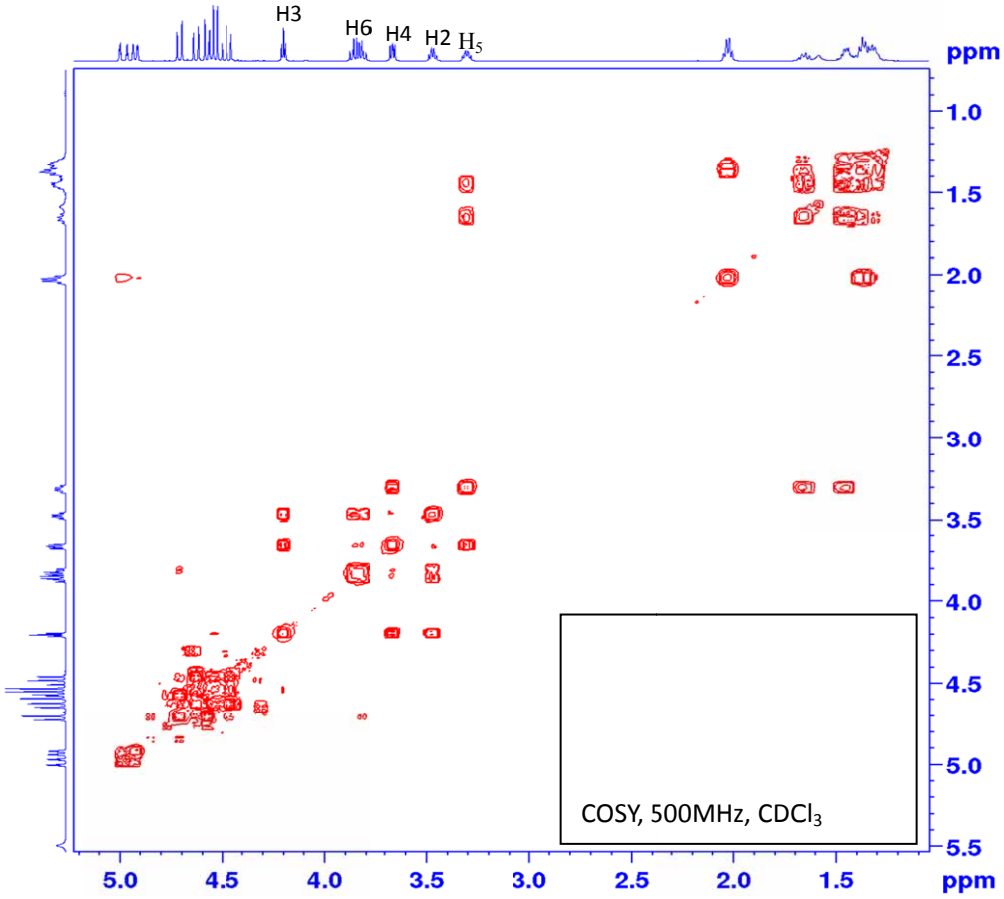


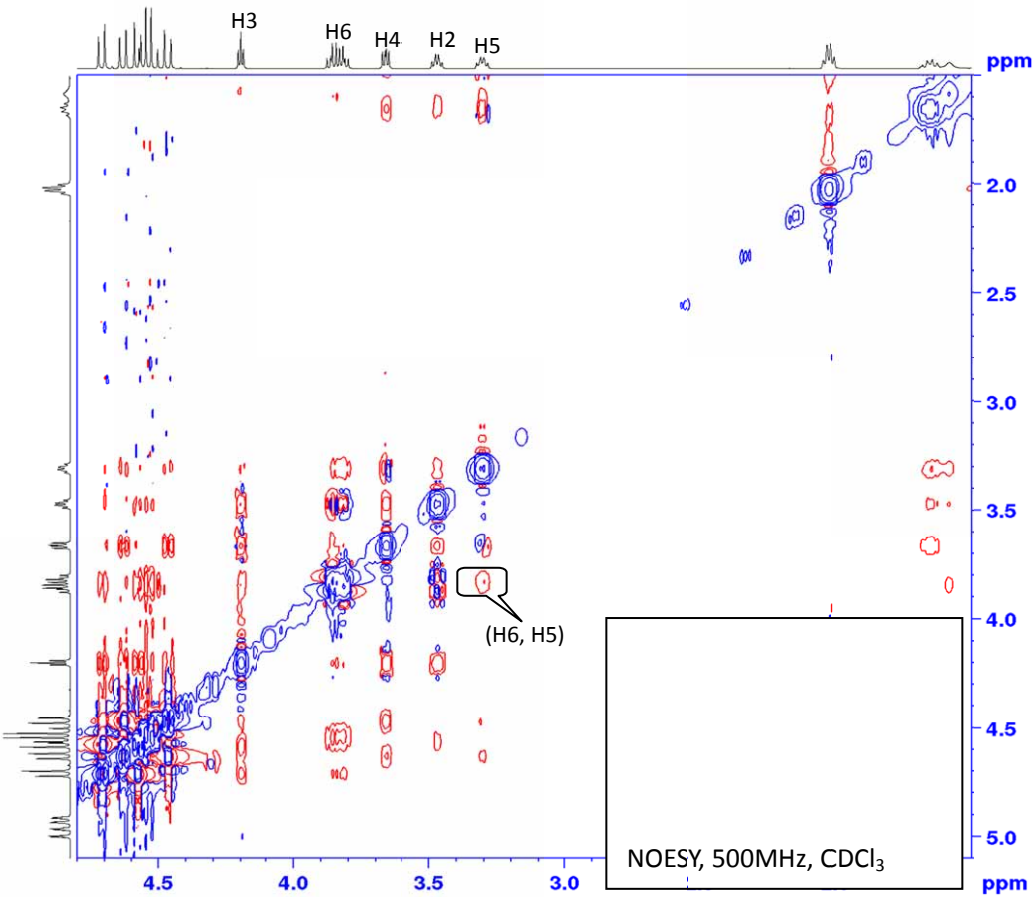
^{13}C NMR, 101 MHz, $CDCl_3$

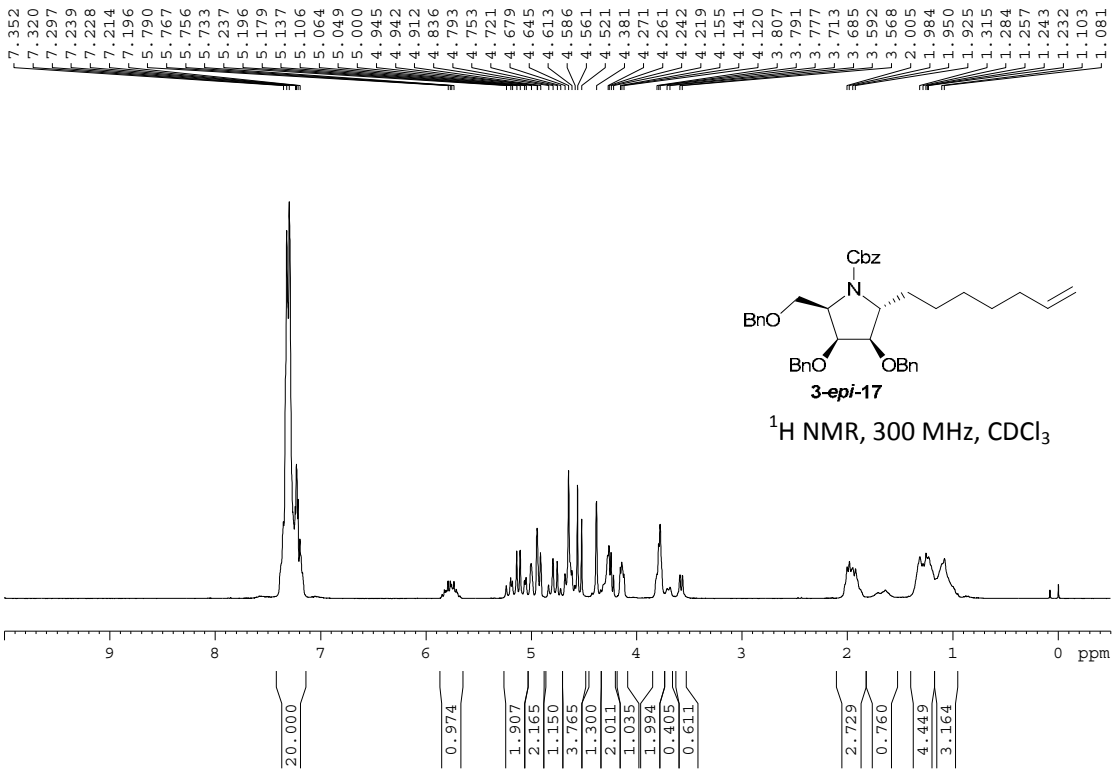

8.704
7.563
7.193
6.540
6.048
6.015
4.713
4.692
4.670
4.437
4.416
4.393
4.277
4.253
4.240
4.217
3.872
3.860
3.594
3.579
2.372
2.352
2.328
2.233
2.207
2.181
2.173
2.113
2.099
2.058
2.039
2.032
1.822
1.813
1.796
1.768
1.749
1.728
1.709
1.705
1.685
1.625
1.610
1.592
HO
H
N
HO
OH
O
ent-Broussonetine W (ent-4)
1H NMR, 300 MHz, Pyr.
9
8
7
6
5
4
3
2
1
0
ppm
1.134
3.801
1.032
1.000
1.799
0.868
0.886
1.673
2.021
2.849
2.972
2.183
8.171

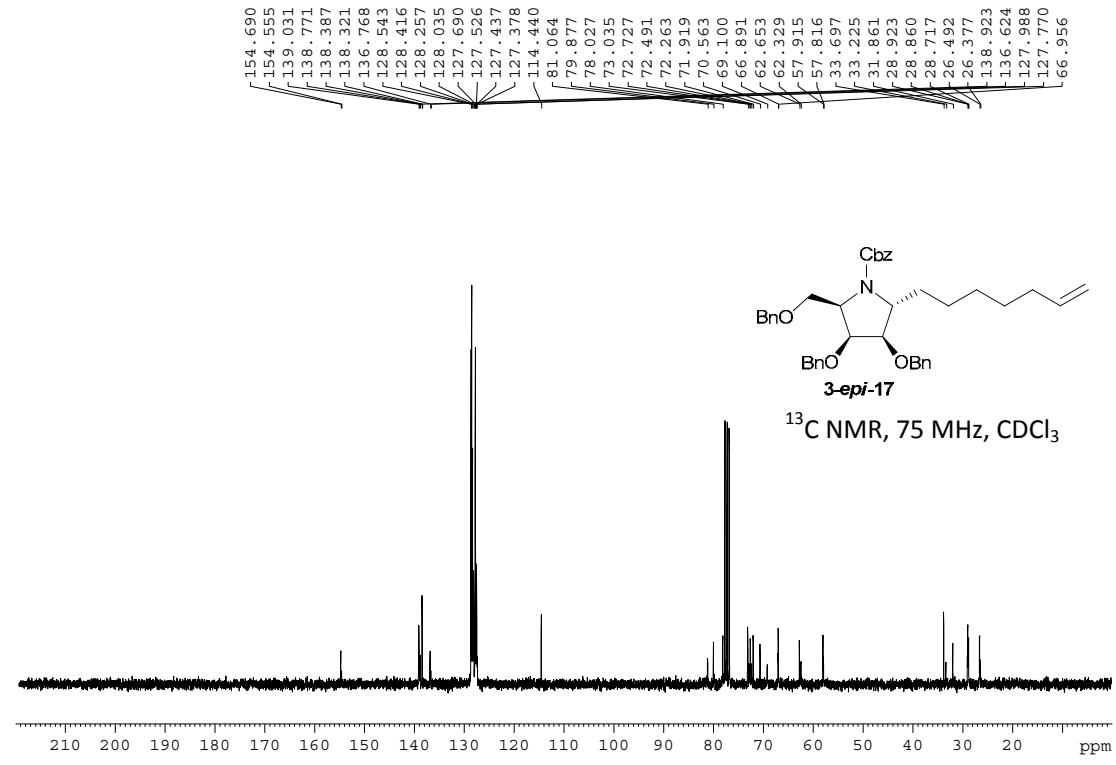

198.807
145.177
139.775
83.209
79.300
65.022
62.843
62.575
38.837
34.540
30.050
29.983
29.726
28.985
27.123
26.142
23.477
HO
H
N
HO
OH
O
ent-Broussonetine W (ent-4)
13C NMR, 75 MHz, Pyr.
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

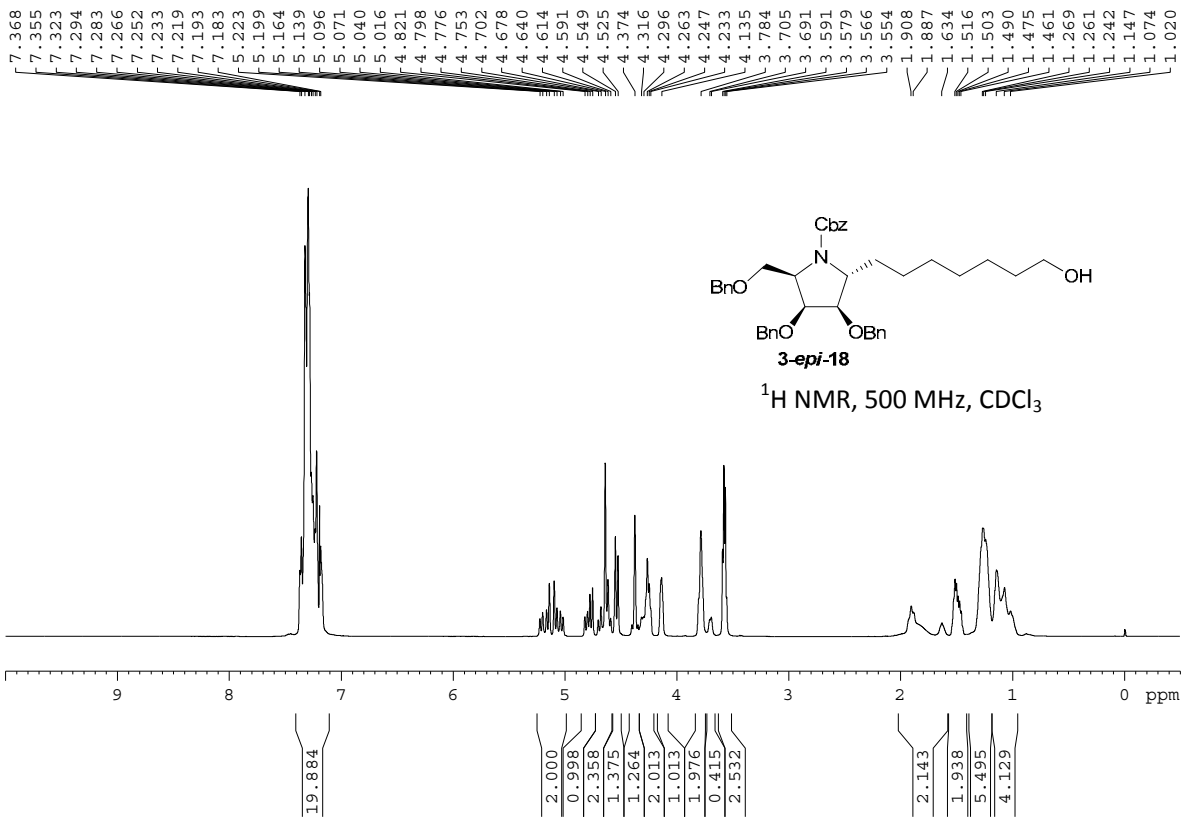

7.332 7.328 7.327 7.318 7.316 7.314 7.309 7.300 7.297 7.294 7.291 7.287 7.281 7.277 7.275 7.270 7.265 7.256 5.804 5.791 5.001 4.997 4.967 4.963 4.936 4.934 4.932 4.916 4.912 4.722 4.699 4.642 4.618 4.588 4.570 4.564 4.546 4.527 4.503 4.478 4.454 4.208 4.198 4.189 3.857 3.843 3.831 3.818 3.674 3.664 3.660 3.650 2.031 2.017 1.368 1.355 1.339 1.321
OH
N
BnO
BnO
OBn
3-epi-16
1H NMR, 500 MHz, CDCl3
9 8 7 6 5 4 3 2 1 0 ppm
14.835 0.969 0.793 0.971 0.964 1.035 1.078 0.852 1.222 1.044 1.014 1.018 0.991 1.032 1.016 1.010 1.000 2.089 1.065 2.066 5.336
139.280 138.384 138.332 138.186 128.531 128.519 128.480 128.019 127.981 127.971 127.890 127.825 127.803 114.336 83.507 73.758 73.615 72.871 71.253 68.779 67.392 33.891 30.724 29.418 28.986 26.914
OH
N
BnO
BnO
OBn
3-epi-16
13C NMR, 126 MHz, CDCl3
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm


H3
H6
H4
H2
H5
ppm
1.0
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
ppm
COSY, 500MHz, CDCl3

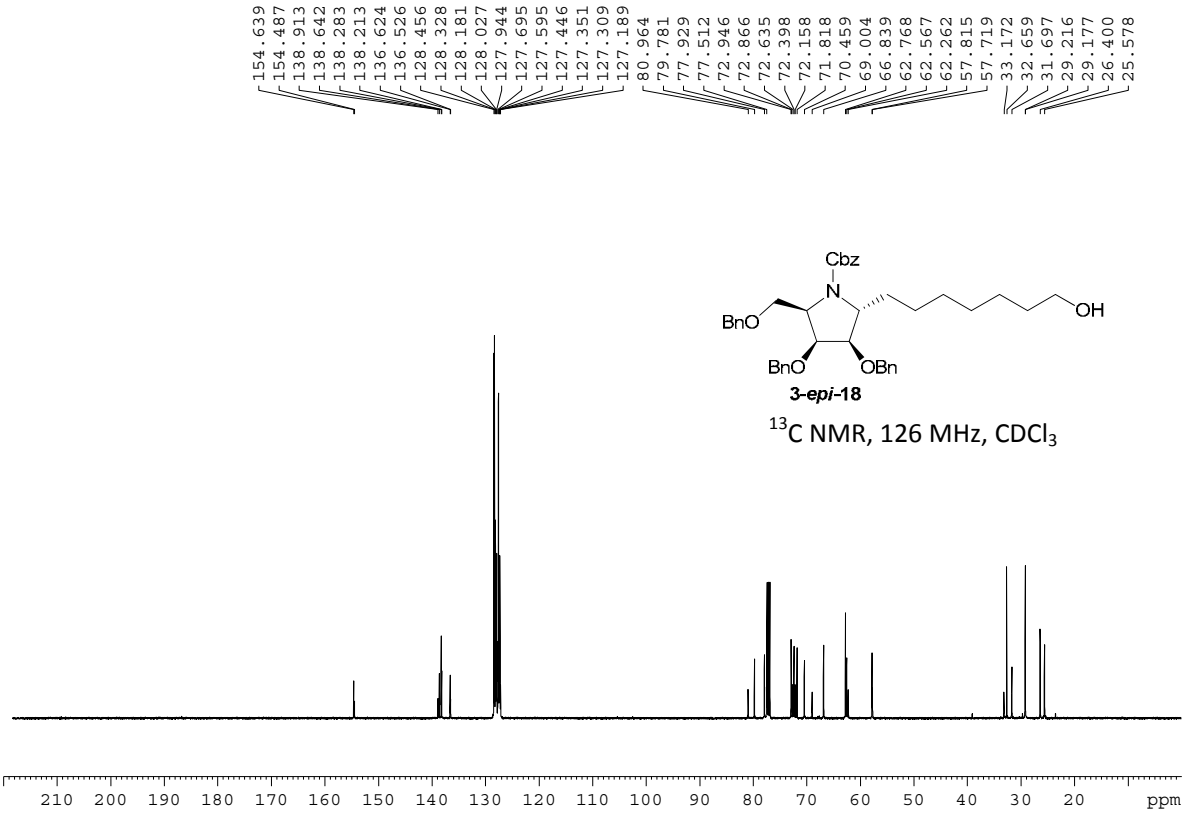

H3
H6
H4
H2
H5
ppm
2.0
2.5
3.0
3.5
4.0
4.5
5.0
4.5
4.0
3.5
3.0
ppm
(H6, H5)
NOESY, 500MHz, CDCl3


7.352
7.320
7.297
7.239
7.228
7.214
7.196
5.790
5.767
5.756
5.733
5.237
5.196
5.179
5.137
5.106
5.064
5.049
5.000
4.945
4.942
4.912
4.836
4.793
4.753
4.721
4.679
4.645
4.613
4.586
4.561
4.521
4.381
4.271
4.261
4.242
4.219
4.155
4.141
4.120
3.807
3.791
3.777
3.713
3.685
3.592
3.568
2.005
1.984
1.950
1.925
1.315
1.284
1.257
1.243
1.232
1.103
1.081
Cbz
N
BnO
BnO
OBn
3-epi-17
1H NMR, 300 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
20.000
0.974
1.907
2.165
1.150
3.765
1.300
2.011
1.035
1.994
0.405
0.611
2.729
0.760
4.449
3.164

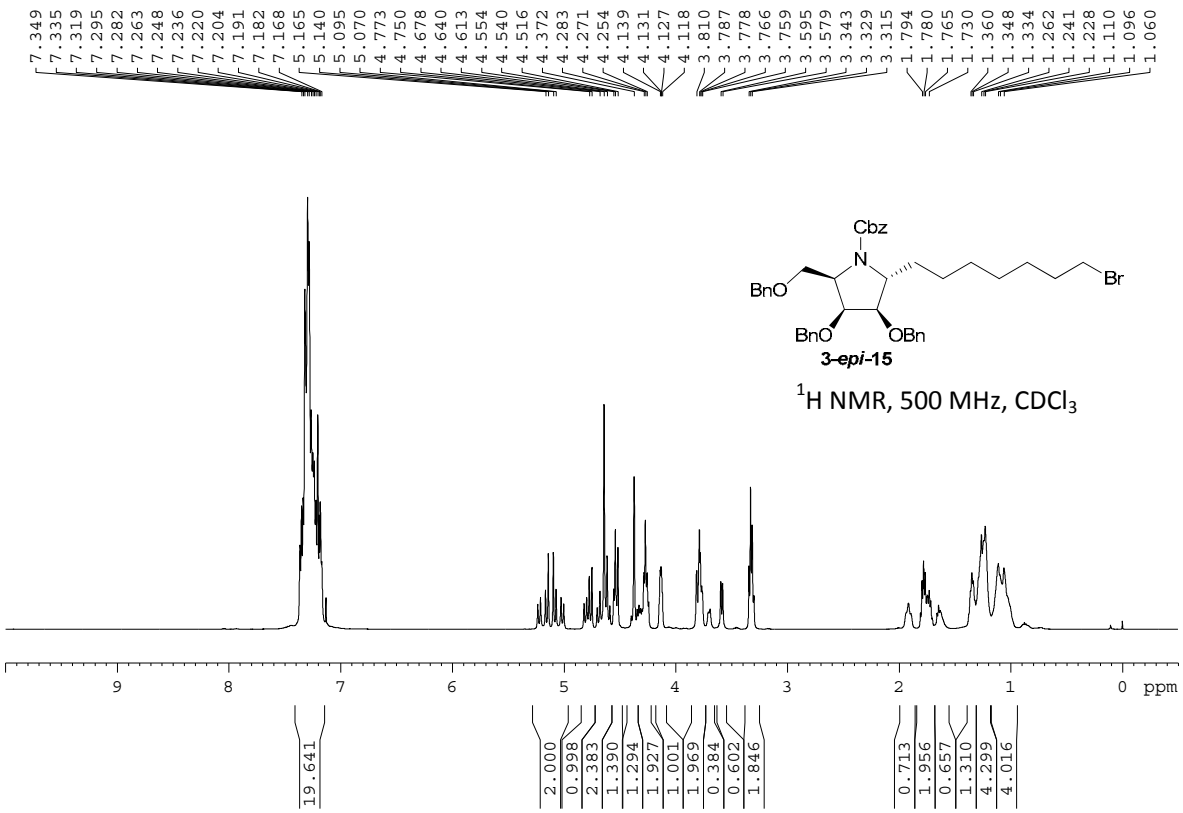

154.690
154.555
139.031
138.771
138.387
138.321
136.768
128.543
128.416
128.257
128.035
127.690
127.526
127.437
127.378
114.440
81.064
79.877
78.027
73.035
72.727
72.491
72.263
71.919
70.563
69.100
66.891
62.653
62.329
57.915
57.816
33.697
33.225
31.861
28.923
28.860
28.717
26.492
26.377
138.923
136.624
127.988
127.770
66.956
Cbz
N
BnO
BnO
OBn
3-epi-17
13C NMR, 75 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

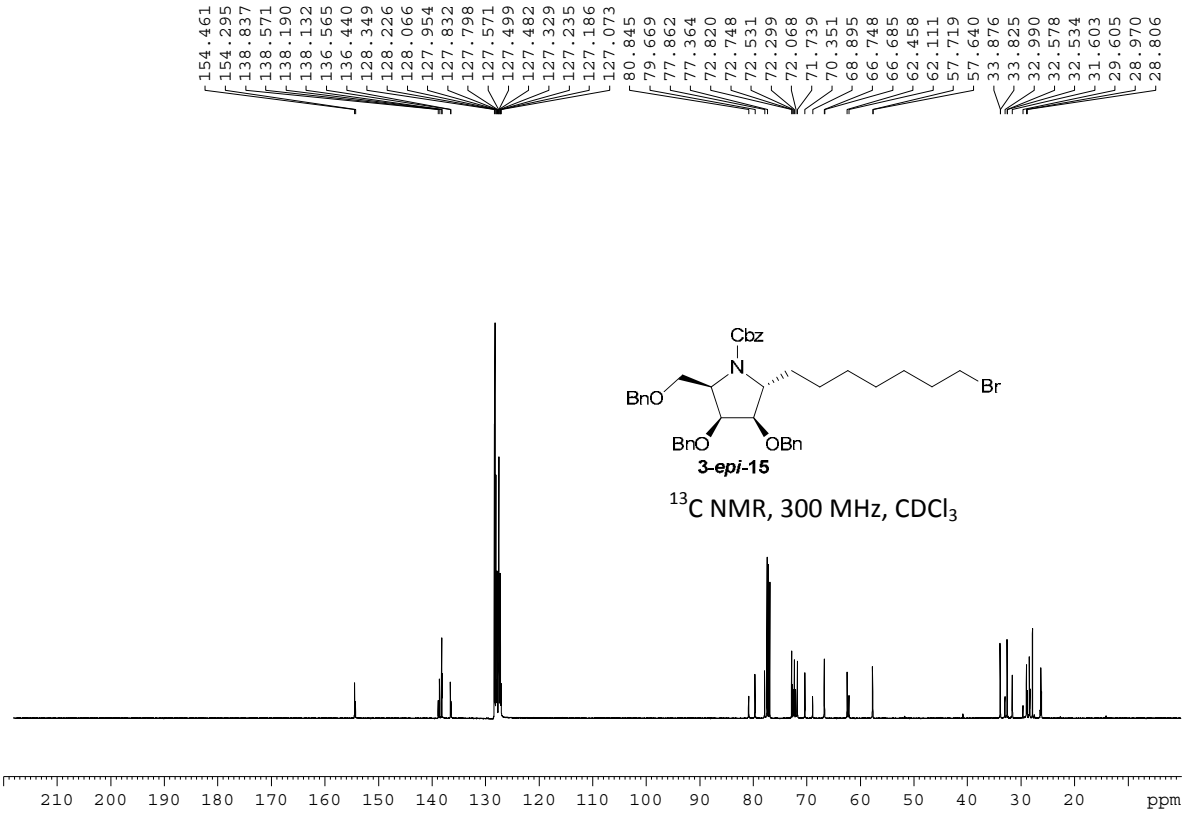


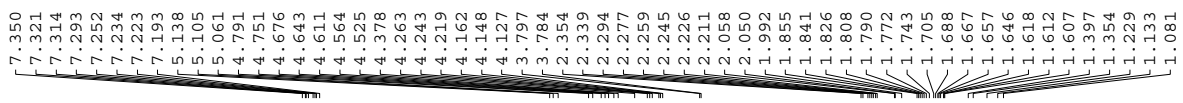
^{1}H NMR, 500 MHz, $CDCl_3$

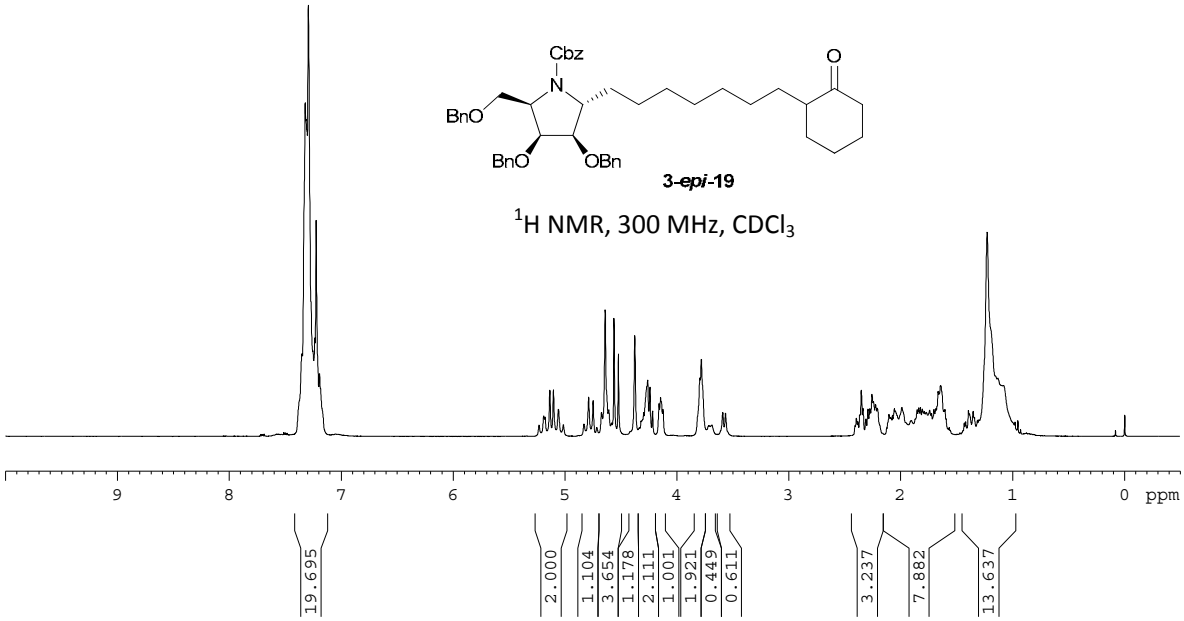


^{13}C NMR, 126 MHz, $CDCl_3$

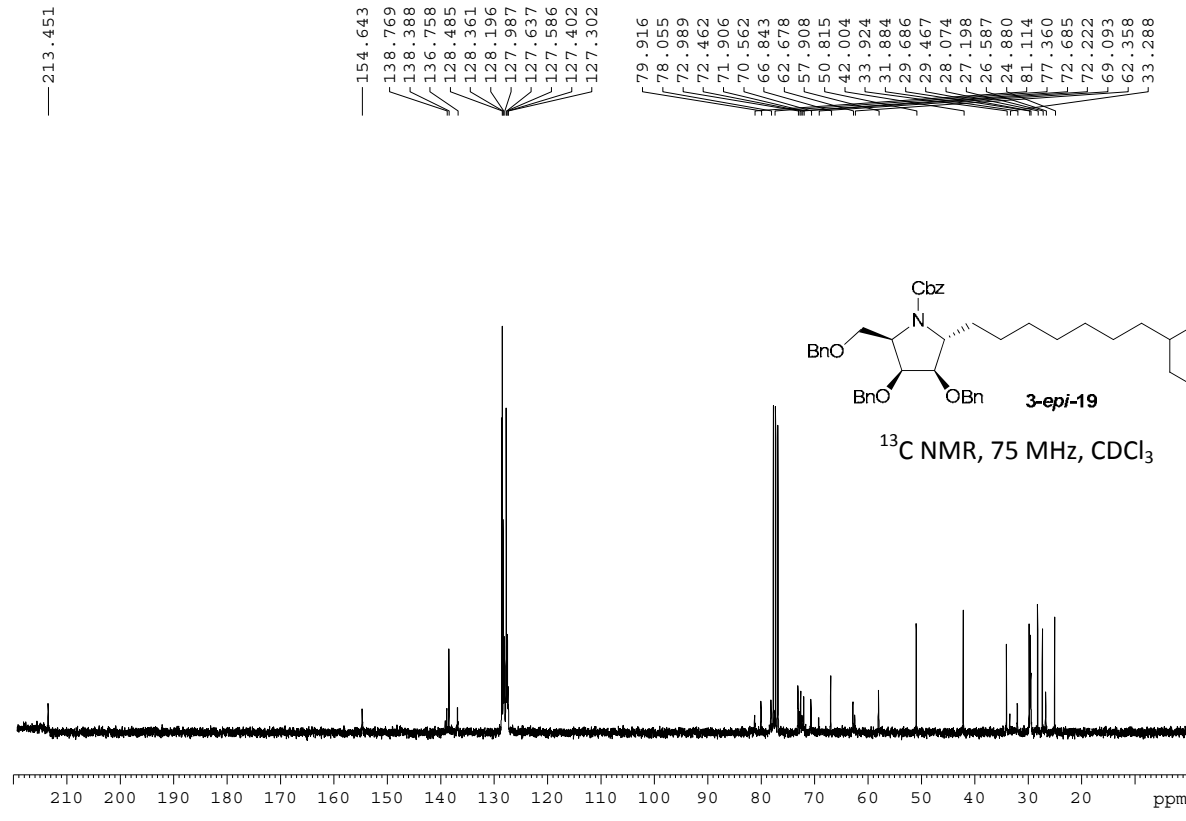

7.349 7.335 7.319 7.295 7.282 7.263 7.248 7.236 7.220 7.204 7.191 7.182 7.168 5.165 5.140 5.095 5.070 4.773 4.750 4.678 4.640 4.613 4.554 4.540 4.516 4.372 4.283 4.271 4.254 4.139 4.131 4.127 4.118 3.810 3.787 3.778 3.766 3.759 3.595 3.579 3.343 3.329 3.315 1.794 1.780 1.765 1.730 1.360 1.348 1.334 1.262 1.241 1.228 1.110 1.096 1.060
Cbz
N
BnO
Br
BnO
OBn
3-epi-15
1H NMR, 500 MHz, CDCl3
9 8 7 6 5 4 3 2 1 0 ppm
19.641
2.000 0.998 2.383 1.390 1.294 1.927 1.001 1.969 0.384 0.602 1.846
0.713 1.956 0.657 1.310 4.299 4.016


154.461 154.295 138.837 138.571 138.190 138.132 136.565 136.440 128.349 128.226 128.066 127.954 127.832 127.798 127.571 127.499 127.482 127.329 127.235 127.186 127.073 80.845 79.669 77.862 77.364 72.820 72.748 72.531 72.299 72.068 71.739 70.351 68.895 66.748 66.685 62.458 62.111 57.719 57.640 33.876 33.825 32.990 32.578 32.534 31.603 29.605 28.970 28.806
Cbz
N
BnO
Br
BnO
OBn
3-epi-15
13C NMR, 300 MHz, CDCl3
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm

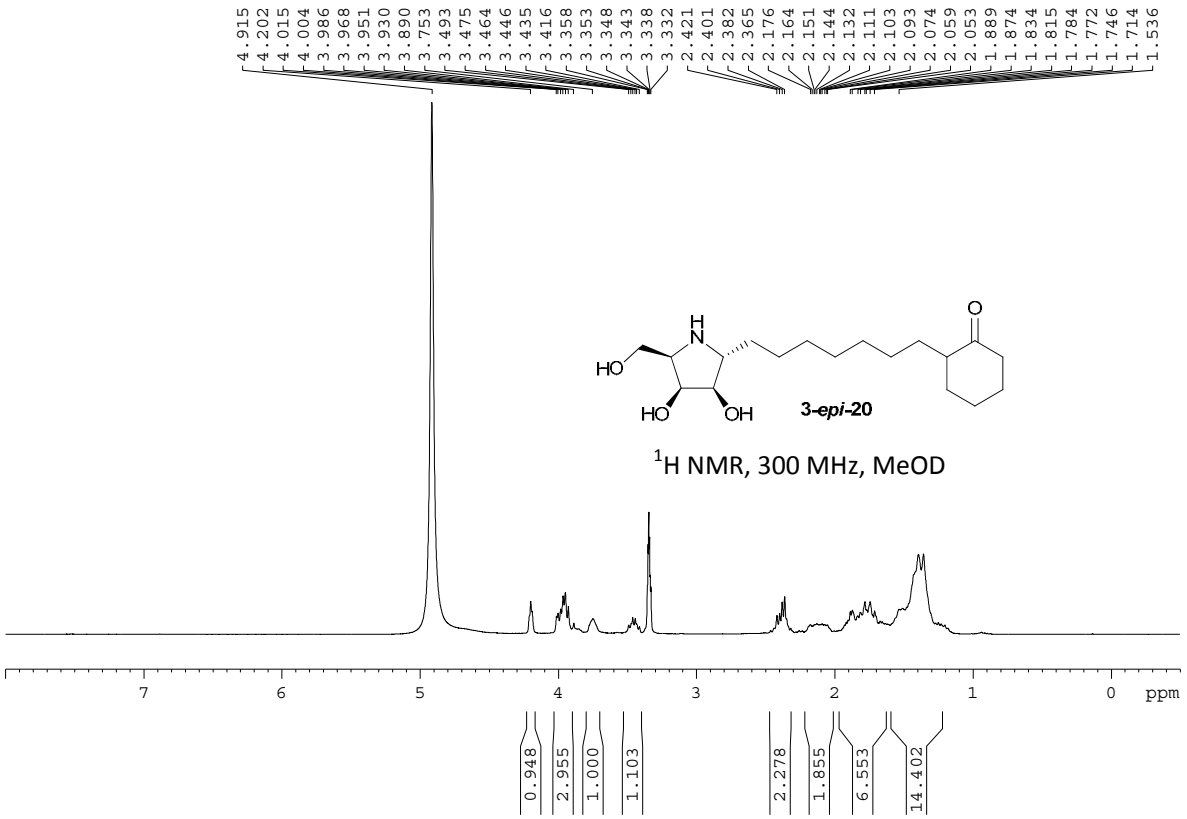




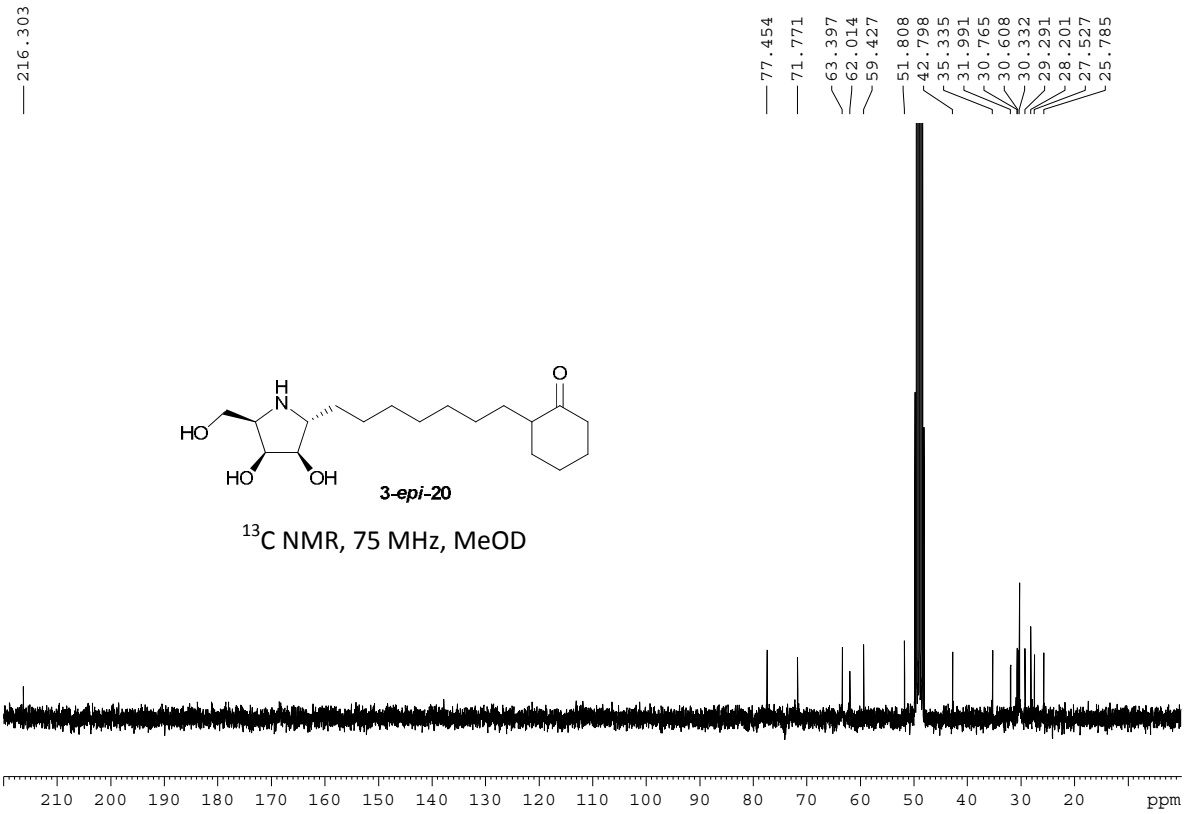
^{1}H NMR, 300 MHz, $CDCl_3$



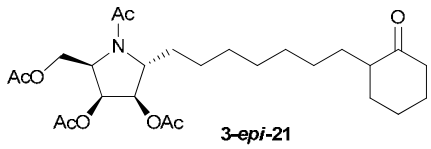
^{13}C NMR, 75 MHz, $CDCl_3$



^{1}H NMR, 300 MHz, MeOD

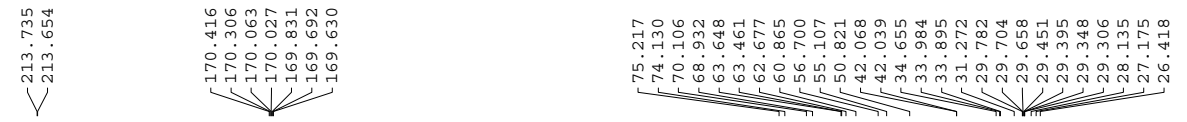


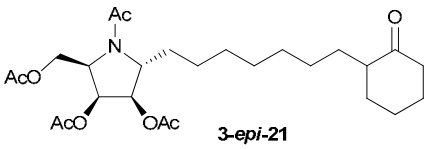
^{13}C NMR, 75 MHz, MeOD



3-*epi*-21

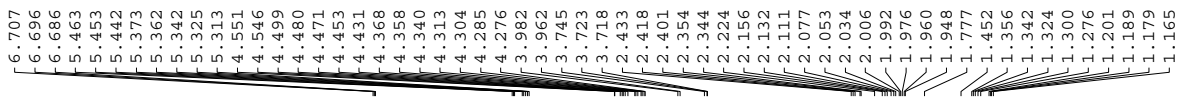
^{1}H NMR, 500 MHz, $CDCl_3$

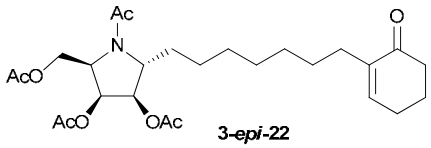




3-*epi*-21

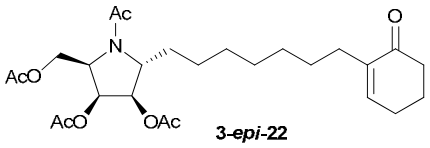
^{13}C NMR, 126 MHz, $CDCl_3$





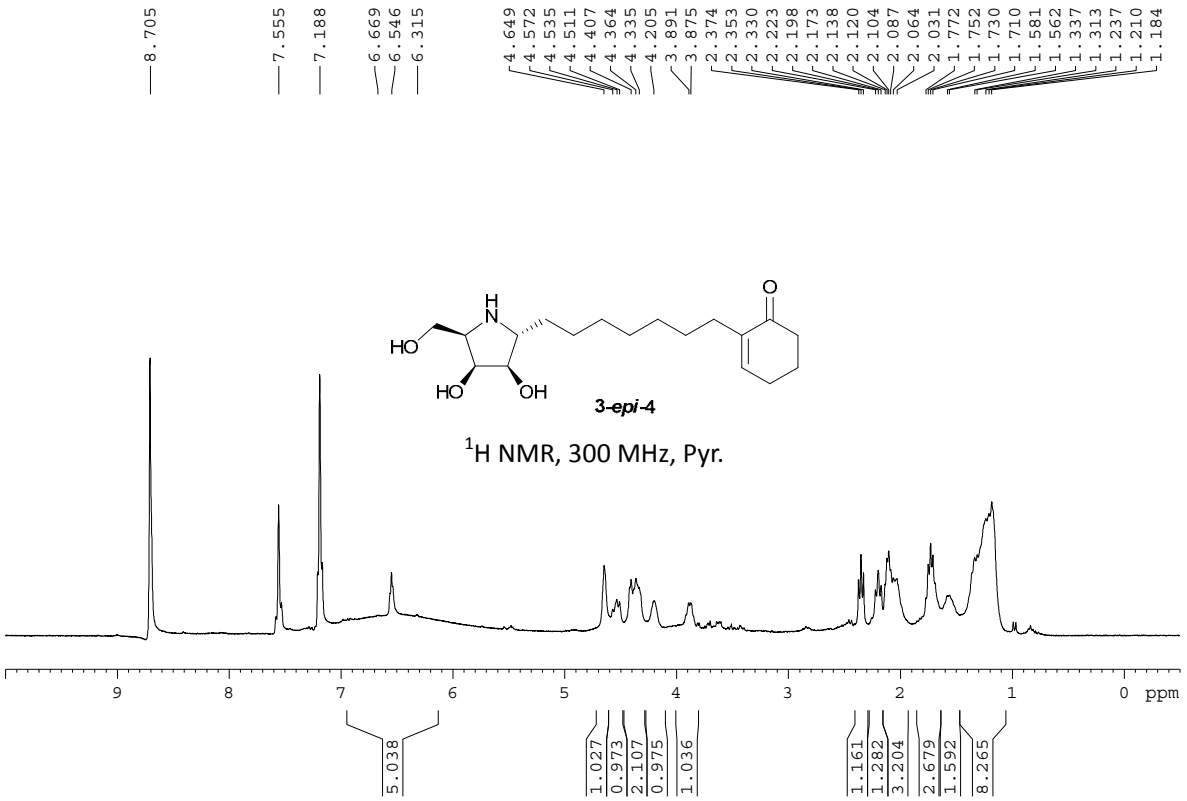
3-*epi*-22

^{1}H NMR, 400 MHz, $CDCl_3$

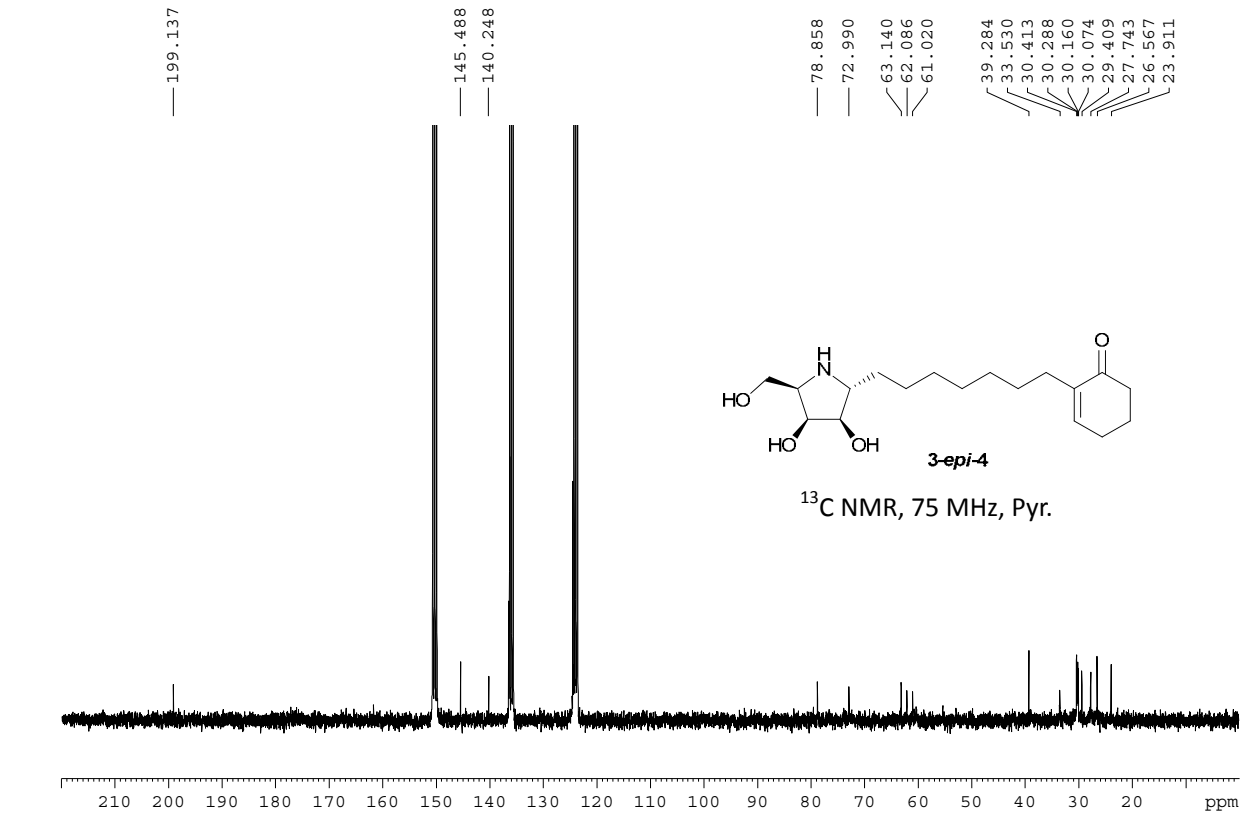


3-*epi*-22

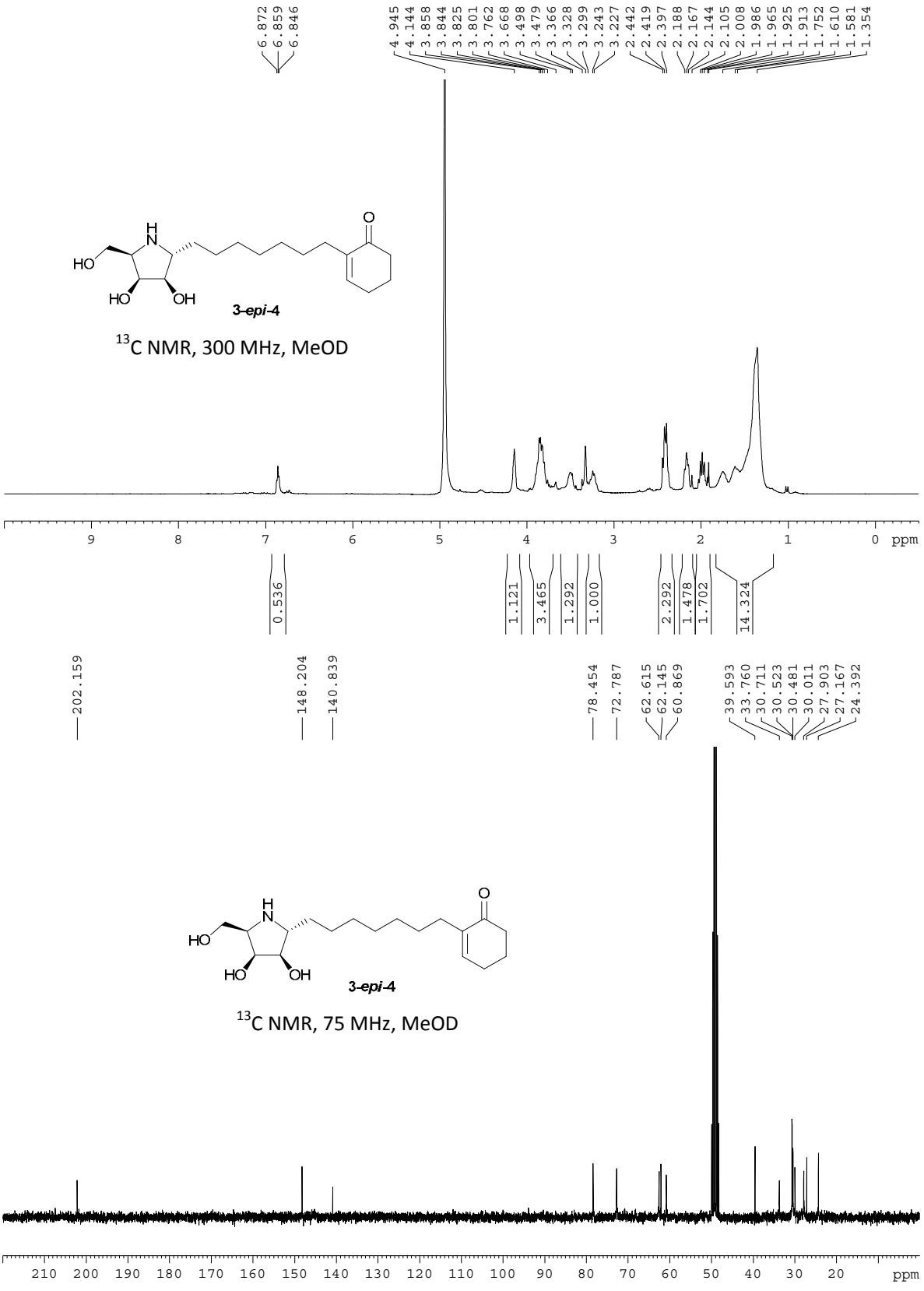
^{13}C NMR, 101 MHz, $CDCl_3$

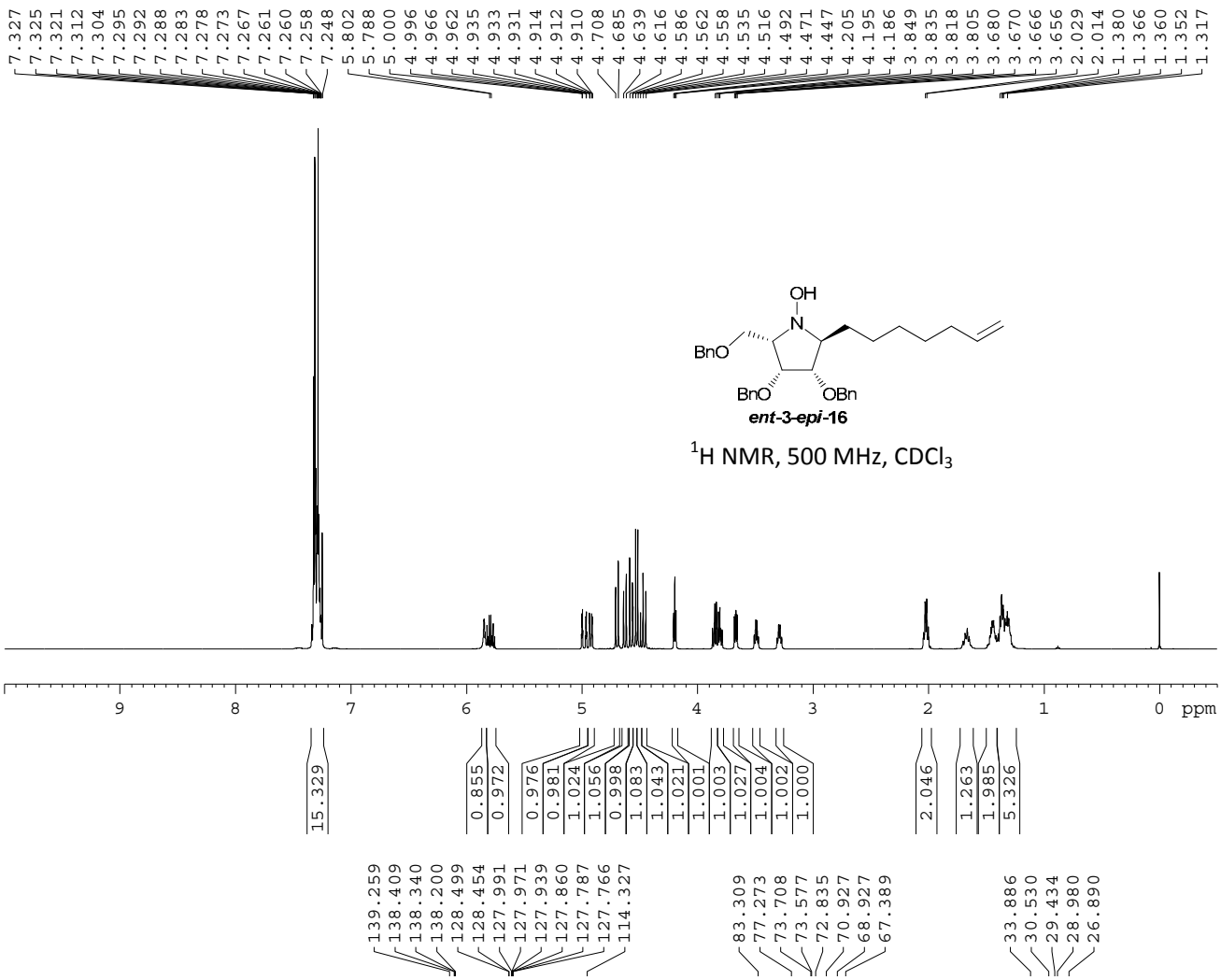


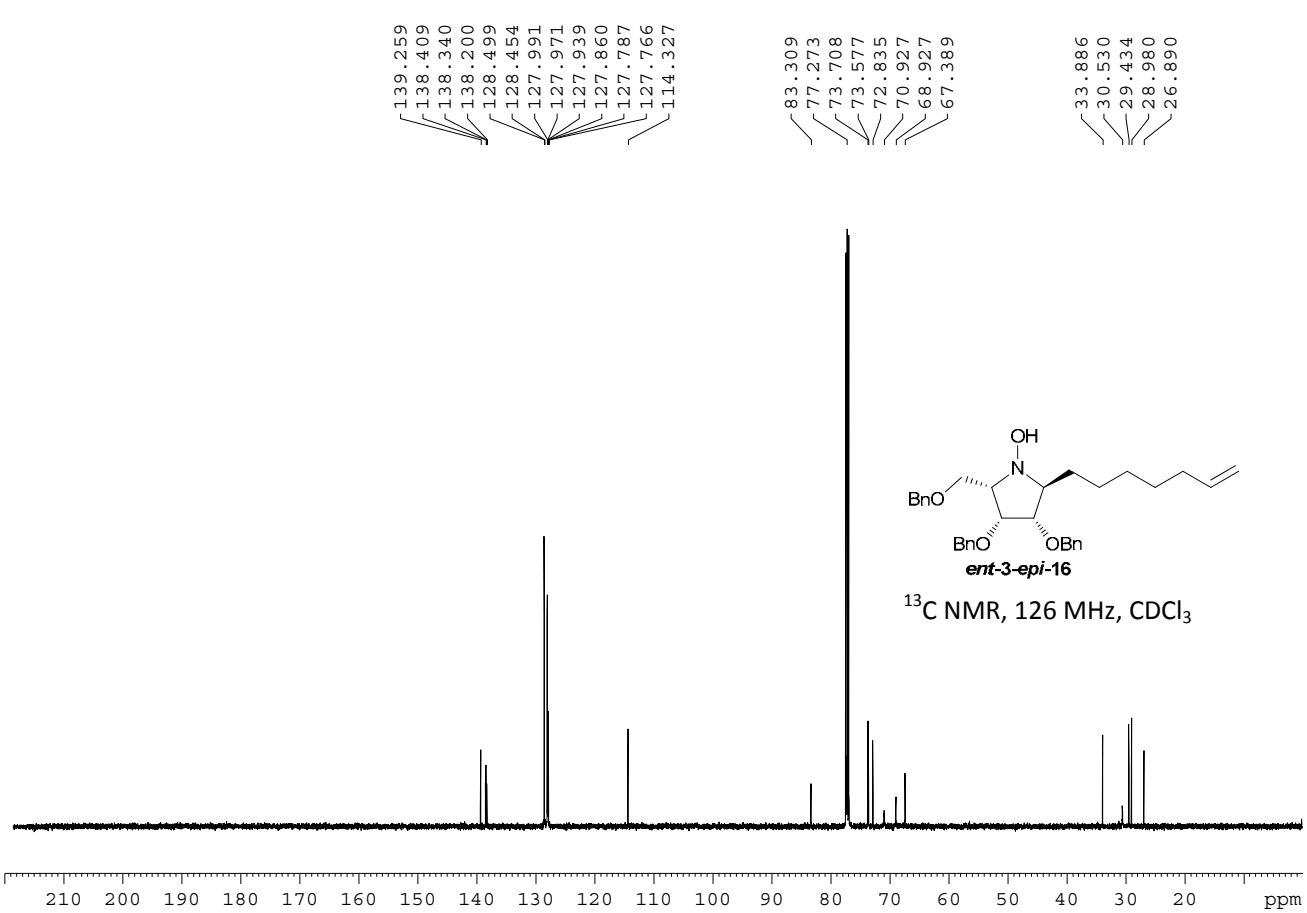
^{1}H NMR, 300 MHz, Pyr.

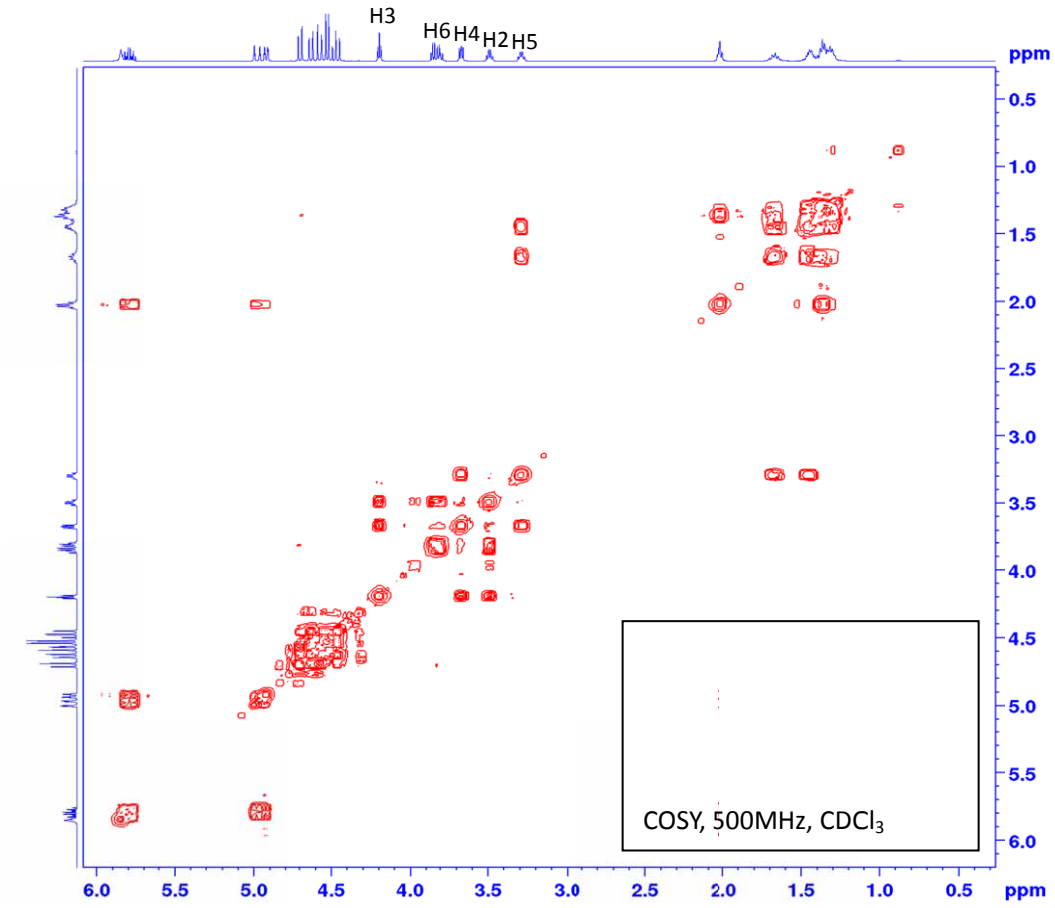


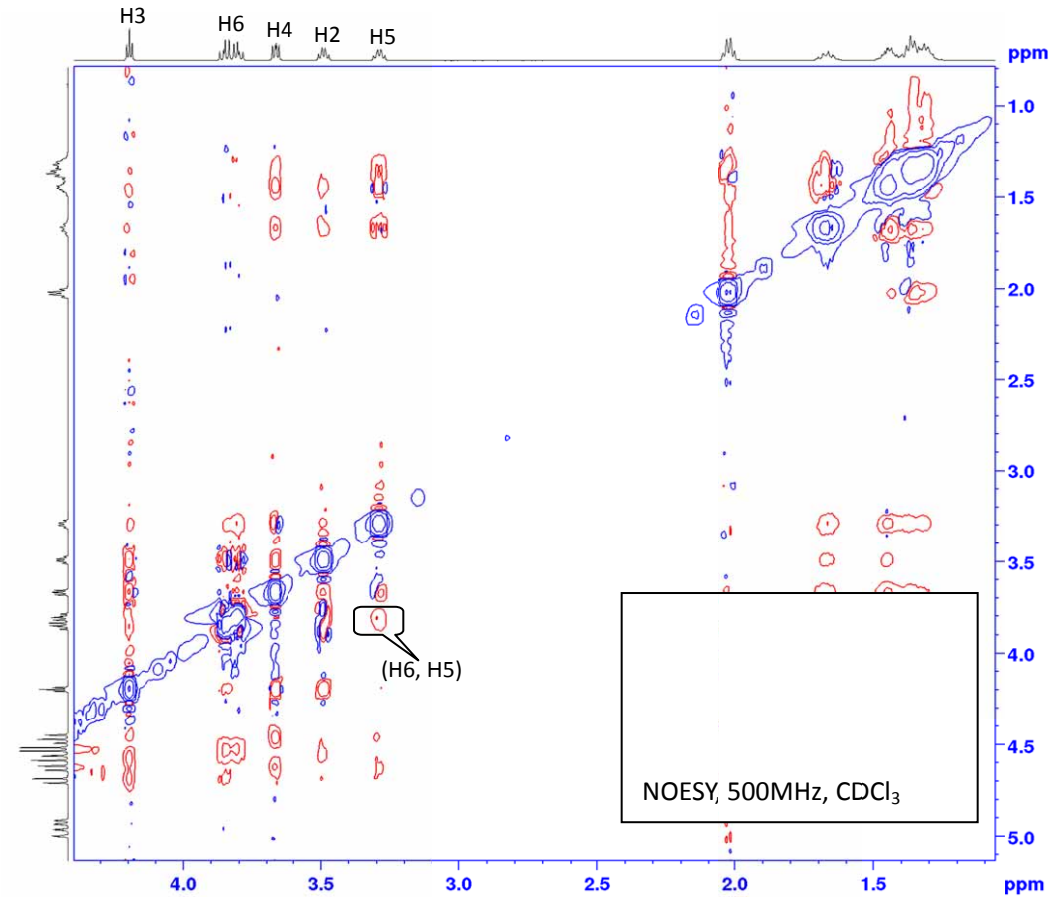
^{13}C NMR, 75 MHz, Pyr.

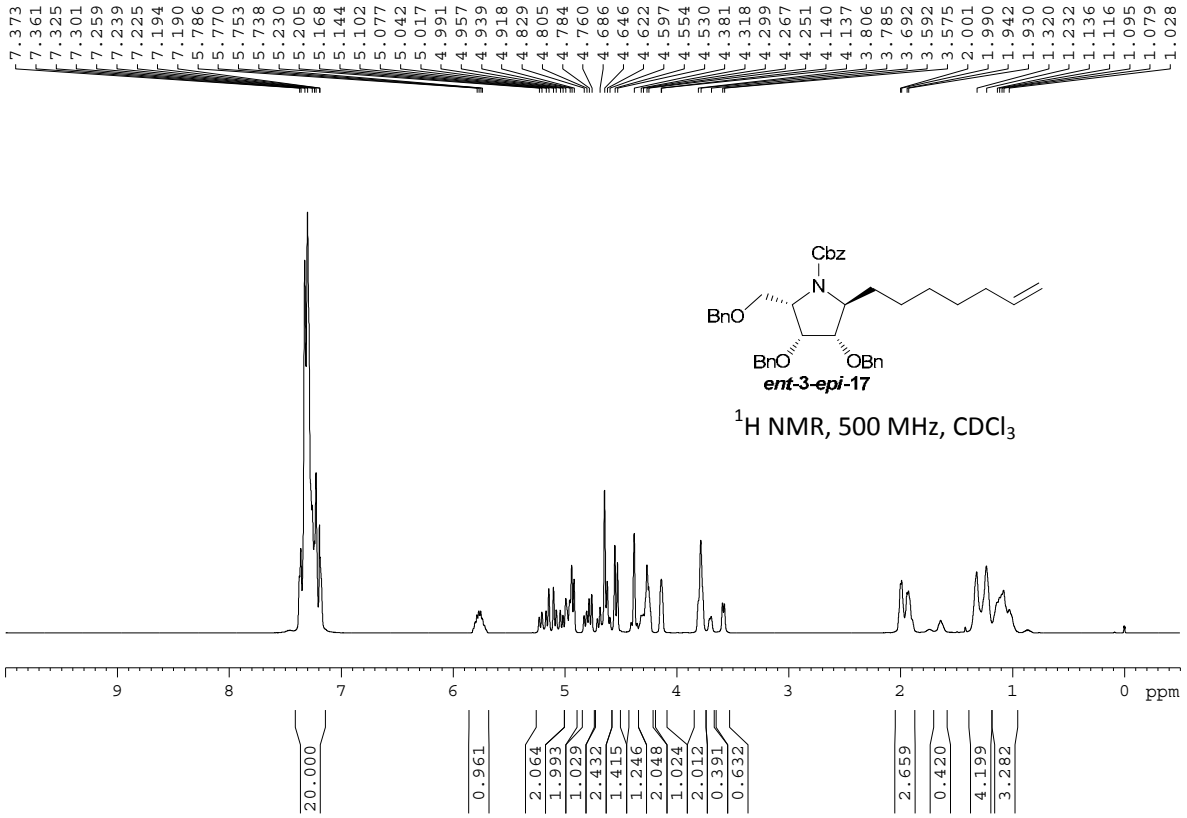

6.872
6.859
6.846
4.945
4.144
3.858
3.844
3.825
3.801
3.762
3.668
3.498
3.479
3.366
3.328
3.299
3.243
3.227
2.442
2.419
2.397
2.188
2.167
2.144
2.105
2.008
1.986
1.965
1.925
1.913
1.752
1.610
1.581
1.354
HO
HO
OH
H
N
O
3-epi-4
13C NMR, 300 MHz, MeOD
9
8
7
6
5
4
3
2
1
0
ppm
0.536
1.121
3.465
1.292
1.000
2.292
1.478
1.702
14.324
202.159
148.204
140.839
78.454
72.787
62.615
62.145
60.869
39.593
33.760
30.711
30.523
30.481
30.011
27.903
27.167
24.392
HO
HO
OH
H
N
O
3-epi-4
13C NMR, 75 MHz, MeOD
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


7.327
7.325
7.321
7.312
7.304
7.295
7.292
7.288
7.283
7.278
7.273
7.267
7.261
7.260
7.258
7.248
5.802
5.788
5.000
4.996
4.966
4.962
4.935
4.933
4.931
4.914
4.912
4.910
4.708
4.685
4.639
4.616
4.586
4.562
4.558
4.535
4.516
4.492
4.471
4.447
4.205
4.195
4.186
3.849
3.835
3.818
3.805
3.680
3.670
3.666
3.656
2.029
2.014
1.380
1.366
1.360
1.352
1.317
OH
N
BnO
BnO
OBn
ent-3-epi-16
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
15.329
0.855
0.972
0.976
0.981
1.024
1.056
0.998
1.083
1.043
1.021
1.001
1.003
1.027
1.004
1.002
1.000
2.046
1.263
1.985
5.326

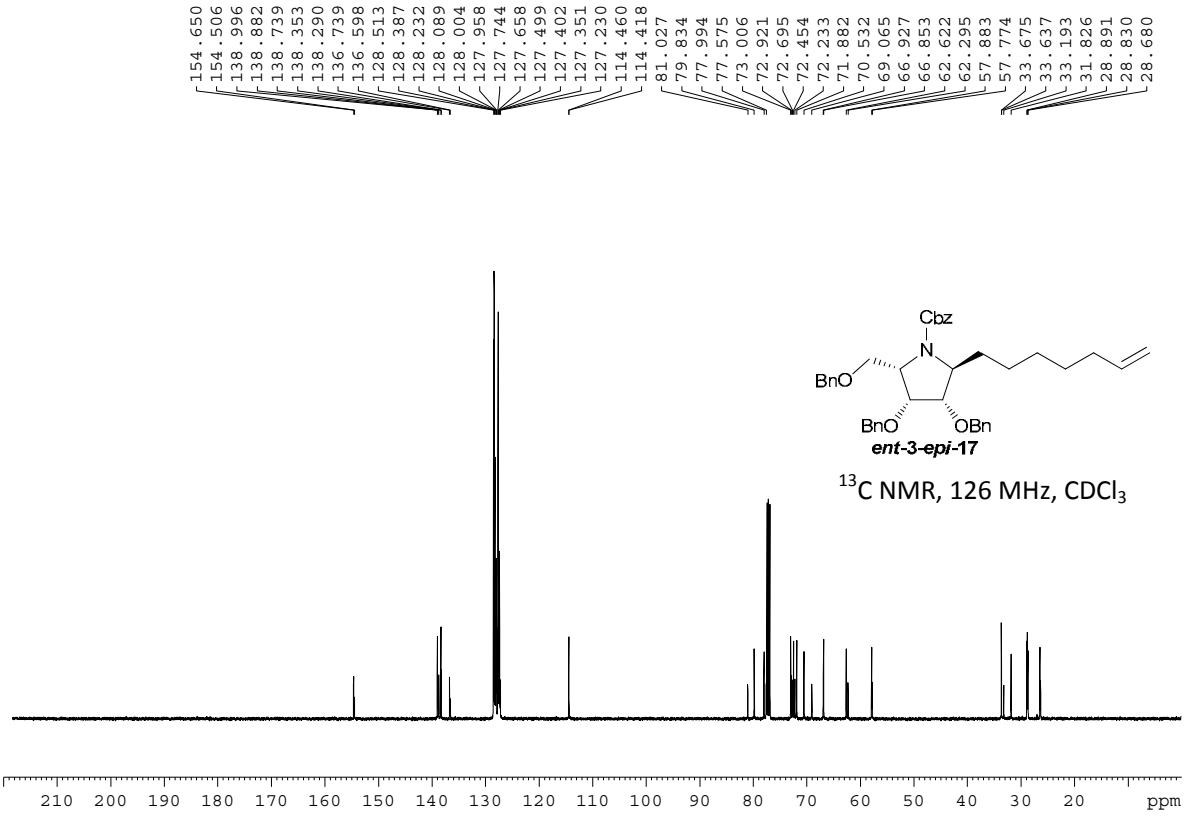

139.259
138.409
138.340
138.200
128.499
128.454
127.991
127.971
127.939
127.860
127.787
127.766
114.327
83.309
77.273
73.708
73.577
72.835
70.927
68.927
67.389
33.886
30.530
29.434
28.980
26.890
OH
N
BnO
BnO
OBn
ent-3-epi-16
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


H3
H6
H4
H2
H5
ppm
0.5
1.0
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
5.5
6.0
COSY, 500MHz, CDCl3

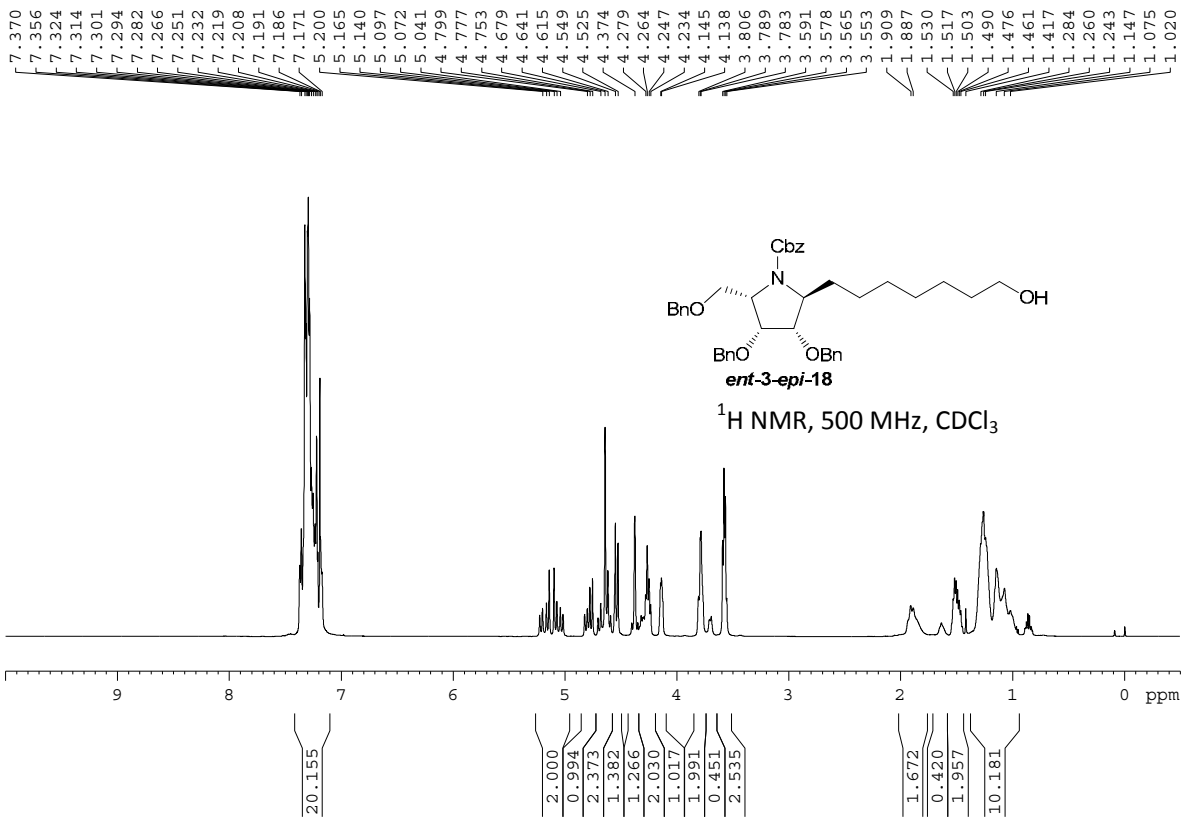

H3
H6
H4
H2
H5
ppm
1.0
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
(H6, H5)
NOESY, 500MHz, CDCl3

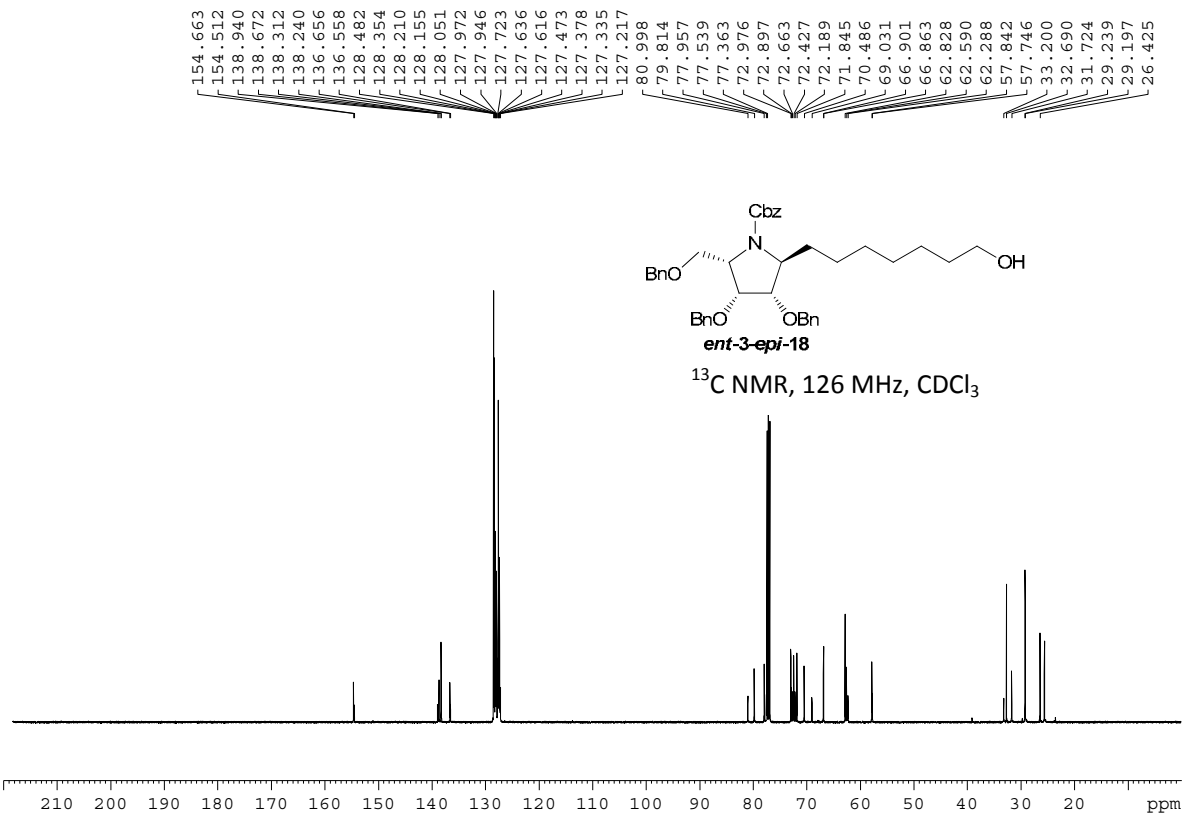


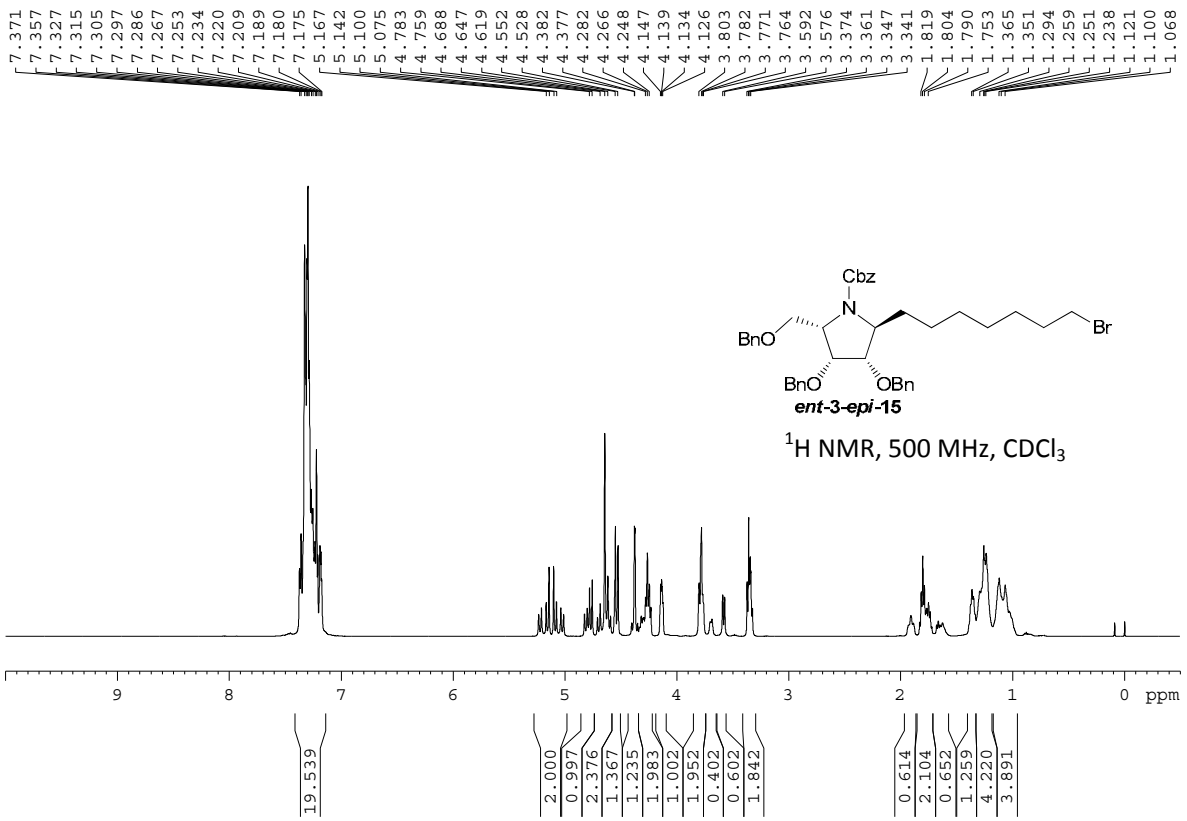
^{1}H NMR, 500 MHz, $CDCl_3$

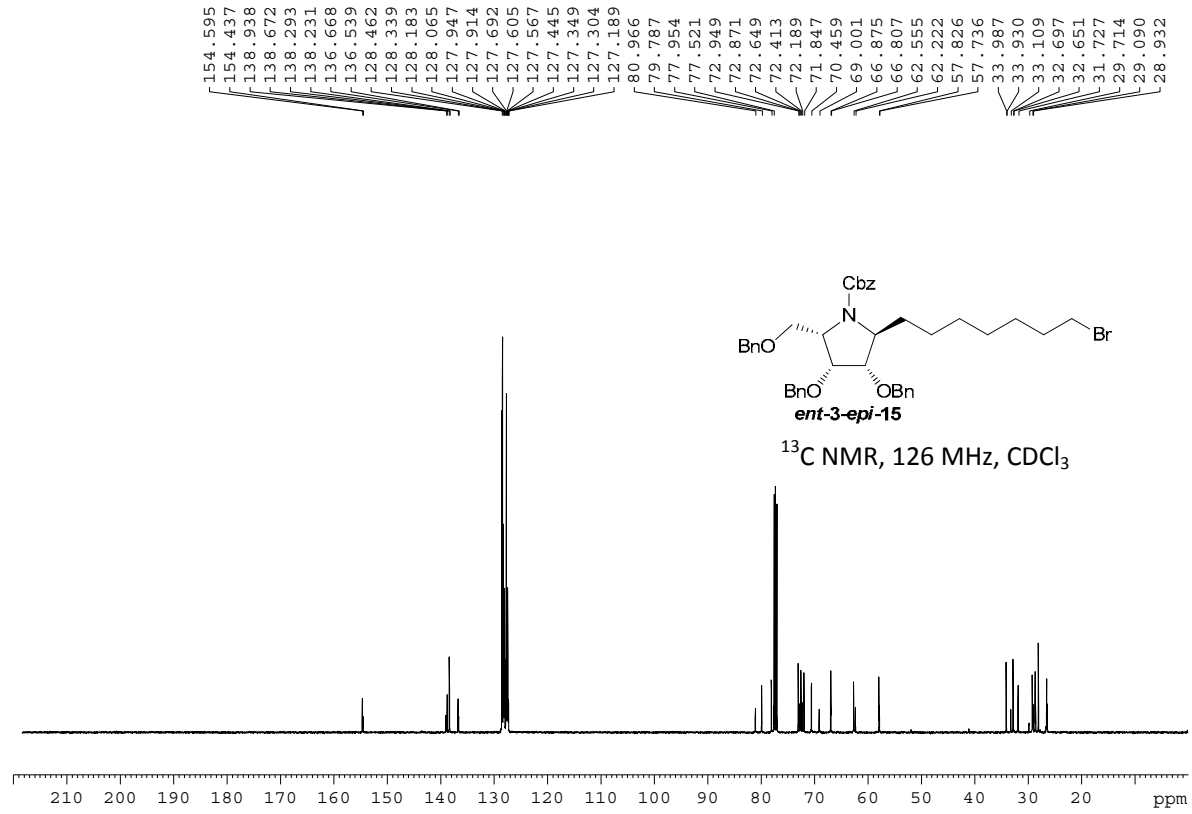


^{13}C NMR, 126 MHz, $CDCl_3$


7.370
7.356
7.324
7.314
7.301
7.294
7.282
7.266
7.251
7.232
7.219
7.208
7.191
7.186
7.171
5.200
5.165
5.140
5.097
5.072
5.041
4.799
4.777
4.753
4.679
4.641
4.615
4.549
4.525
4.374
4.279
4.264
4.247
4.234
4.145
4.138
3.806
3.789
3.783
3.591
3.578
3.565
3.553
1.909
1.887
1.530
1.517
1.503
1.490
1.476
1.461
1.417
1.284
1.260
1.243
1.147
1.075
1.020
Cbz
N
BnO
OH
BnO
OBn
ent-3-epi-18
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
20.155
2.000
0.994
2.373
1.382
1.266
2.030
1.017
1.991
0.451
2.535
1.672
0.420
1.957
10.181


154.663
154.512
138.940
138.672
138.312
138.240
136.656
136.558
128.482
128.354
128.210
128.155
128.051
127.972
127.946
127.723
127.636
127.616
127.473
127.378
127.335
127.217
80.998
79.814
77.957
77.539
77.363
72.976
72.897
72.663
72.427
72.189
71.845
70.486
69.031
66.901
66.863
62.828
62.590
62.288
57.842
57.746
33.200
32.690
31.724
29.239
29.197
26.425
Cbz
N
BnO
OH
BnO
OBn
ent-3-epi-18
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


7.371
7.357
7.327
7.315
7.305
7.297
7.286
7.267
7.253
7.234
7.220
7.209
7.189
7.180
7.175
5.167
5.142
5.100
5.075
4.783
4.759
4.688
4.647
4.619
4.552
4.528
4.382
4.377
4.282
4.266
4.248
4.147
4.139
4.134
4.126
3.803
3.782
3.771
3.764
3.592
3.576
3.374
3.361
3.347
3.341
1.819
1.804
1.790
1.753
1.365
1.351
1.294
1.259
1.251
1.238
1.121
1.100
1.068
Cbz
N
BnO
Br
BnO
OBn
ent-3-epi-15
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0 ppm
19.539
2.000
0.997
2.376
1.367
1.235
1.983
1.002
1.952
0.402
0.602
1.842
0.614
2.104
0.652
1.259
4.220
3.891


154.595
154.437
138.938
138.672
138.293
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136.668
136.539
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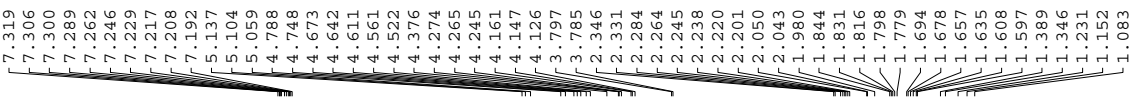
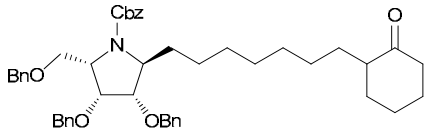
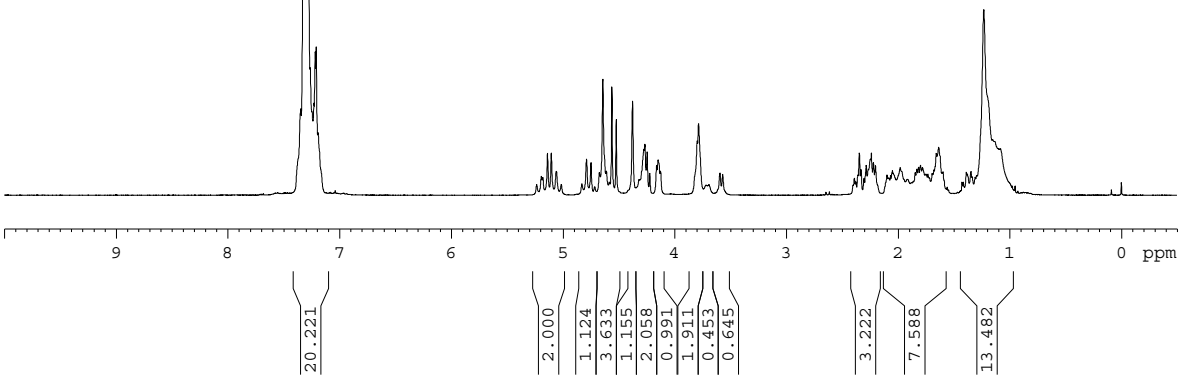
128.339
128.183
128.065
127.947
127.914
127.692
127.605
127.567
127.445
127.349
127.304
127.189
80.966
79.787
77.954
77.521
72.949
72.871
72.649
72.413
72.189
71.847
70.459
69.001
66.875
66.807
62.555
62.222
57.826
57.736
33.987
33.930
33.109
32.697
32.651
31.727
29.714
29.090
28.932
Cbz
N
BnO
Br
BnO
OBn
ent-3-epi-15
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

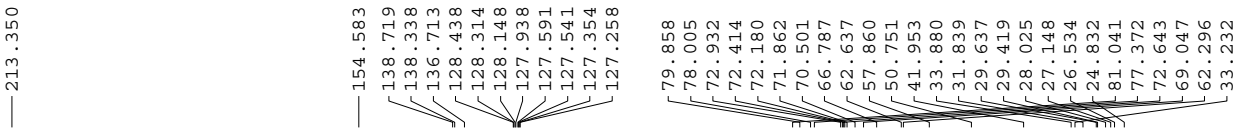


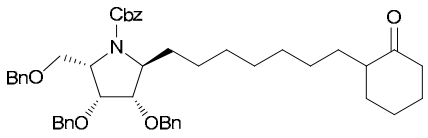


***ent*-3-*epi*-19**

^{1}H NMR, 300 MHz, $CDCl_3$

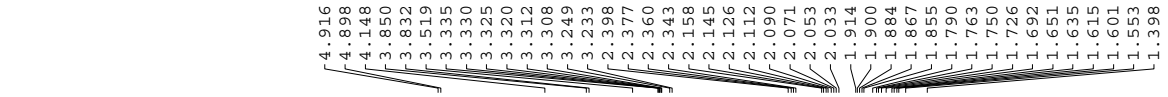


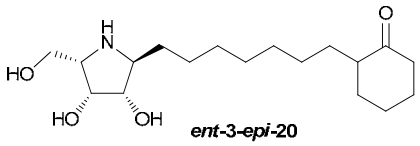




***ent*-3-*epi*-19**

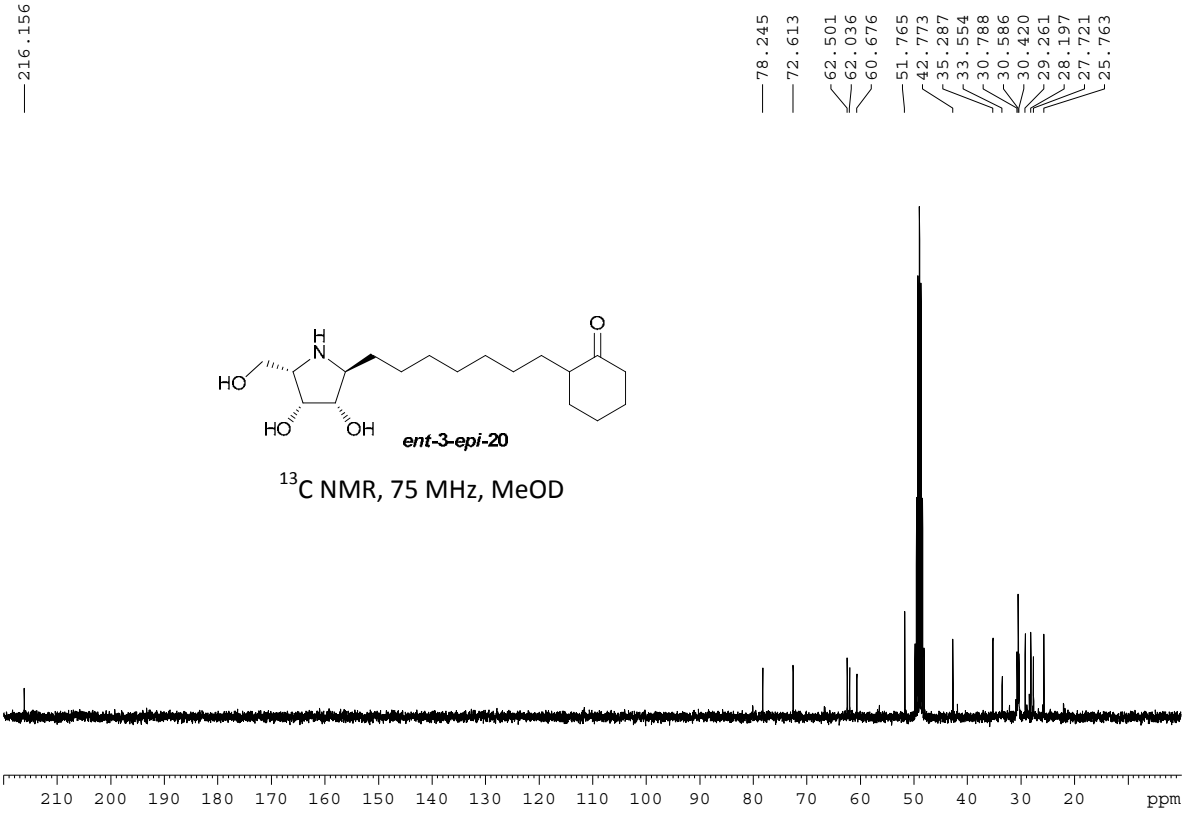
^{13}C NMR, 75 MHz, $CDCl_3$





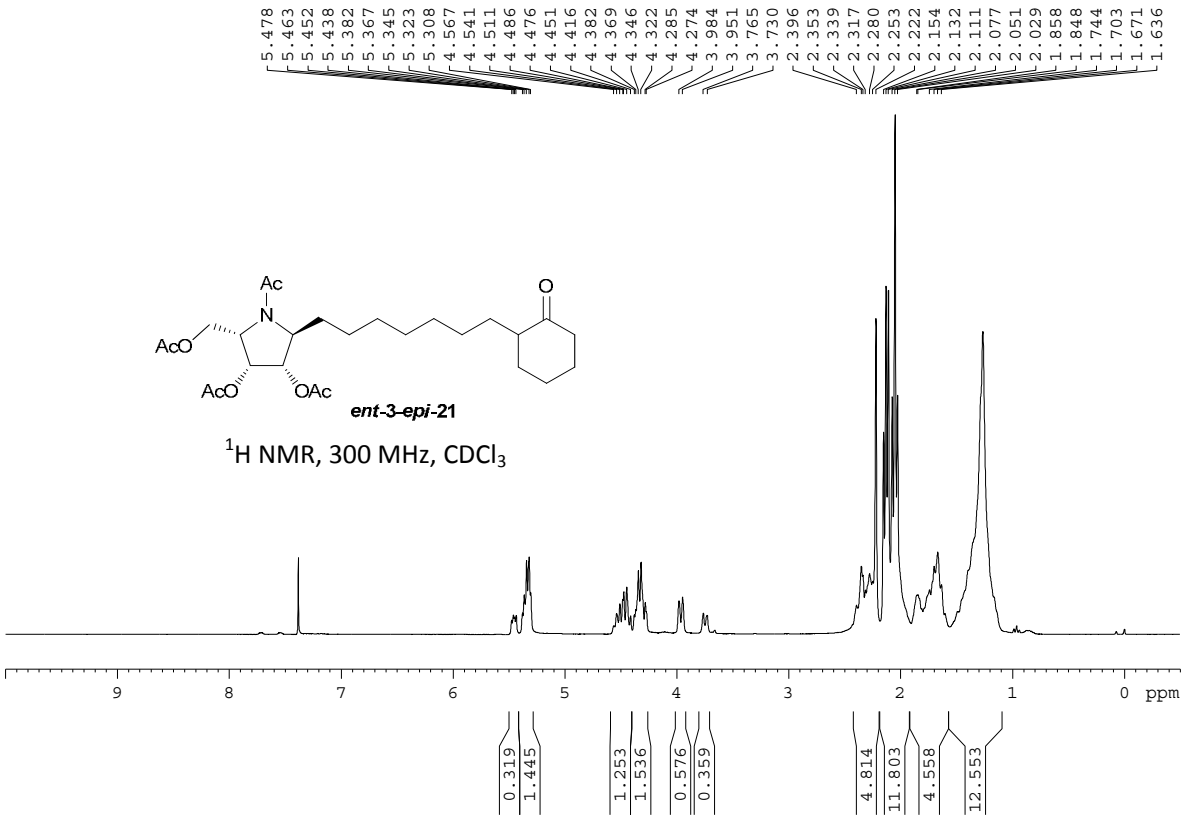
ent-3-*epi*-20

^{1}H NMR, 300 MHz, MeOD



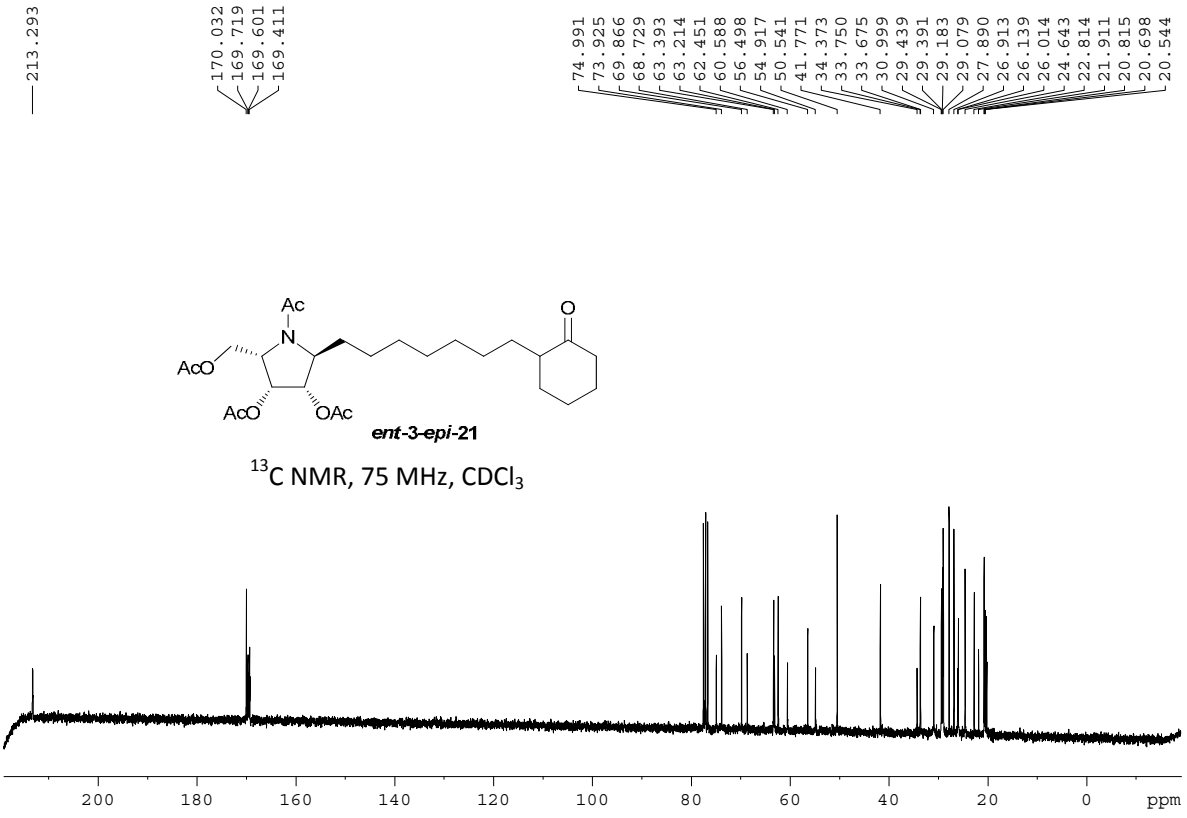
ent-3-*epi*-20

^{13}C NMR, 75 MHz, MeOD



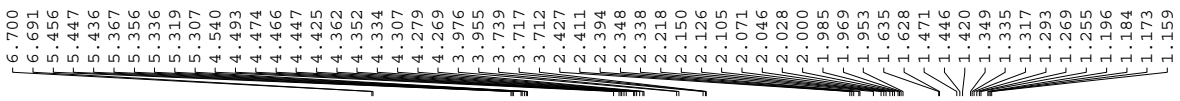
***ent*-3-*epi*-21**

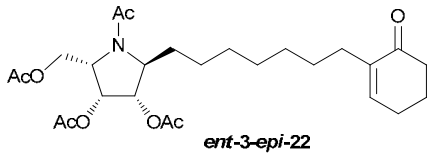
^{1}H NMR, 300 MHz, $CDCl_3$



***ent*-3-*epi*-21**

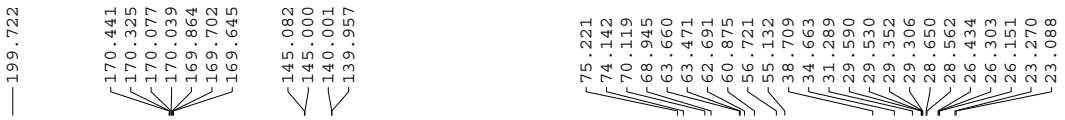
^{13}C NMR, 75 MHz, $CDCl_3$

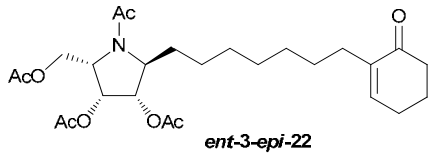




***ent*-3-*epi*-22**

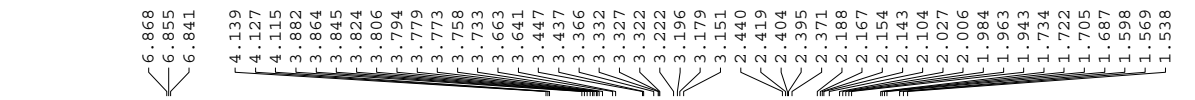
^{1}H NMR, 400 MHz, $CDCl_3$

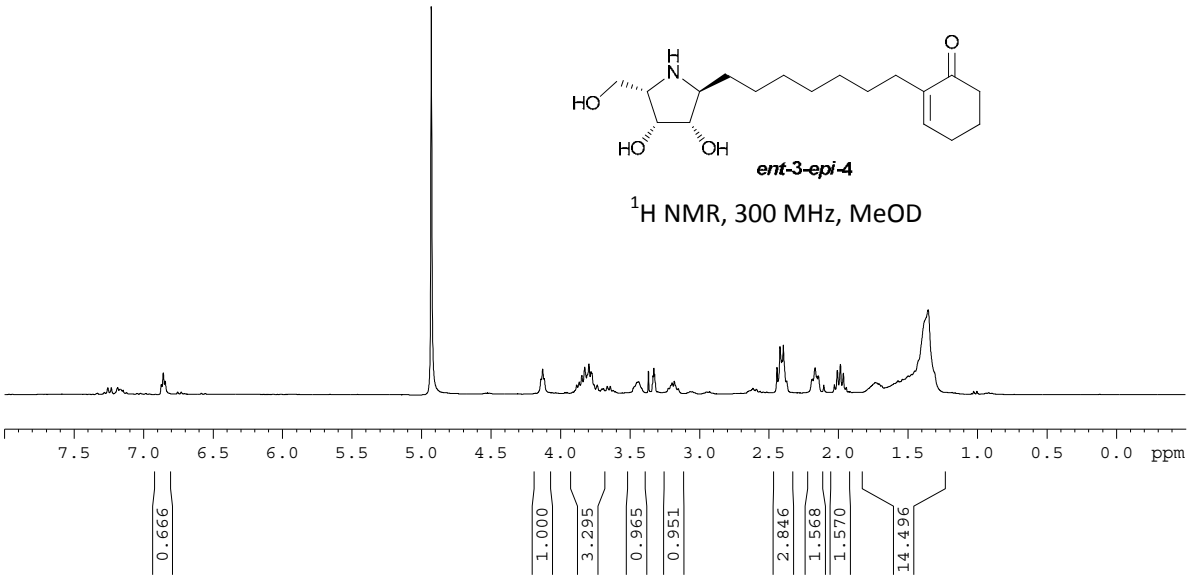




***ent*-3-*epi*-22**

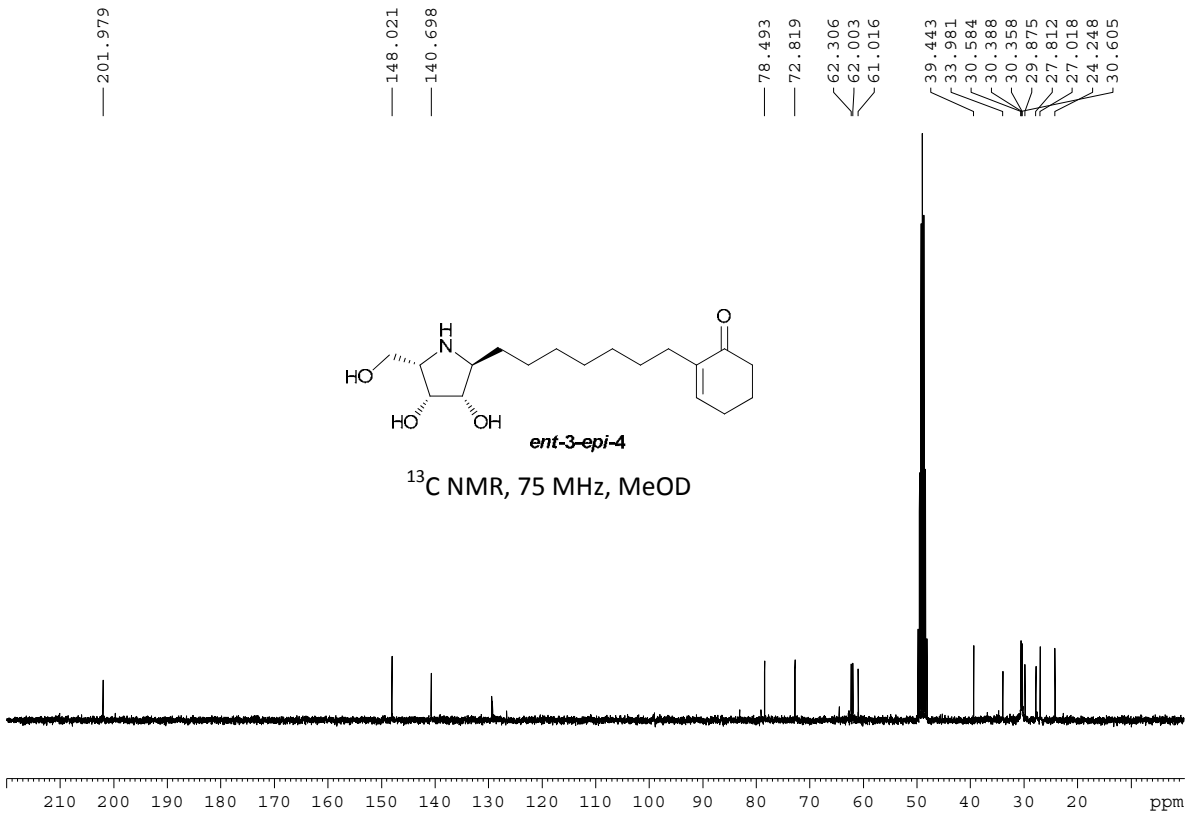
^{13}C NMR, 101 MHz, $CDCl_3$





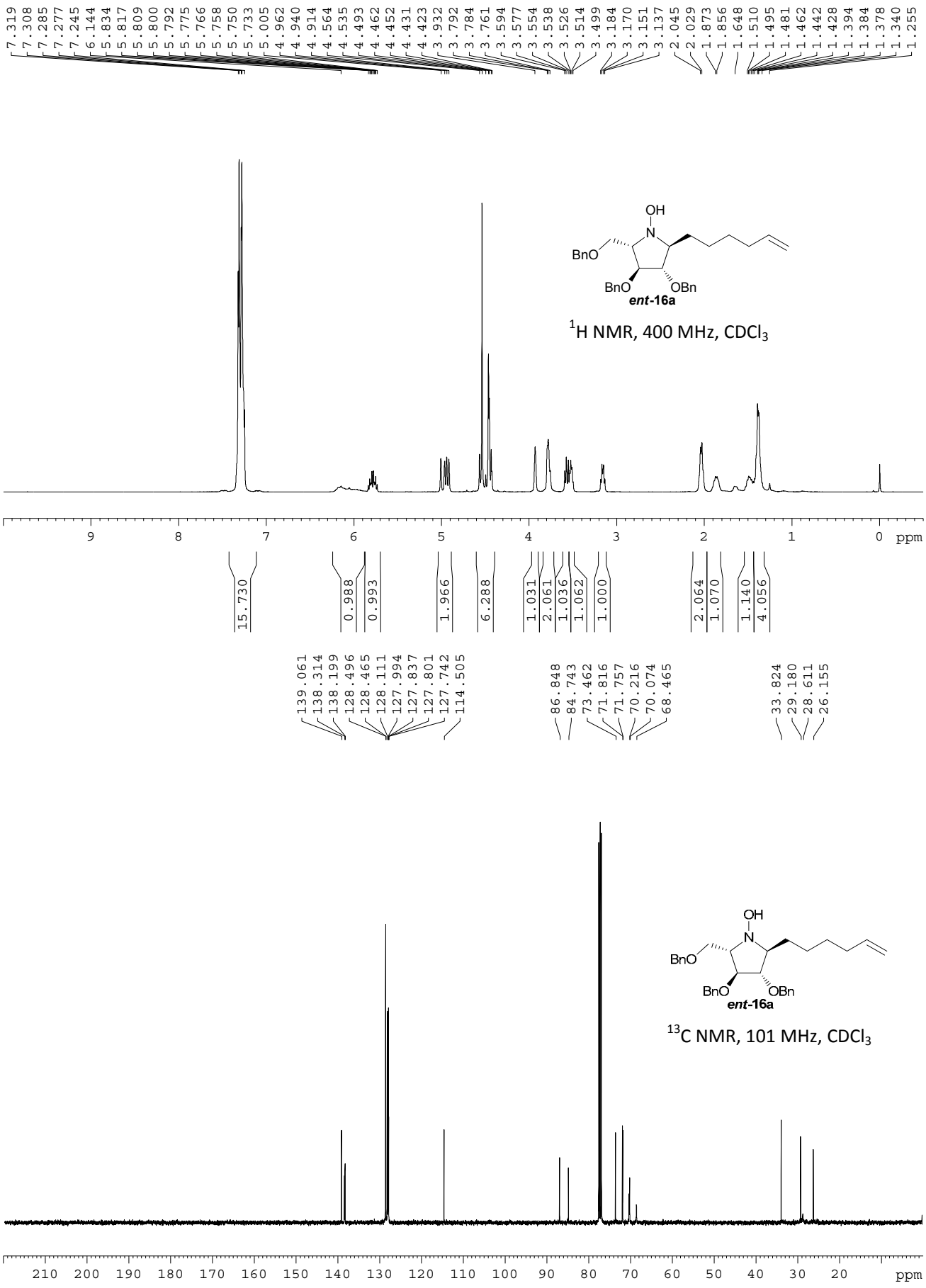
***ent*-3-*epi*-4**

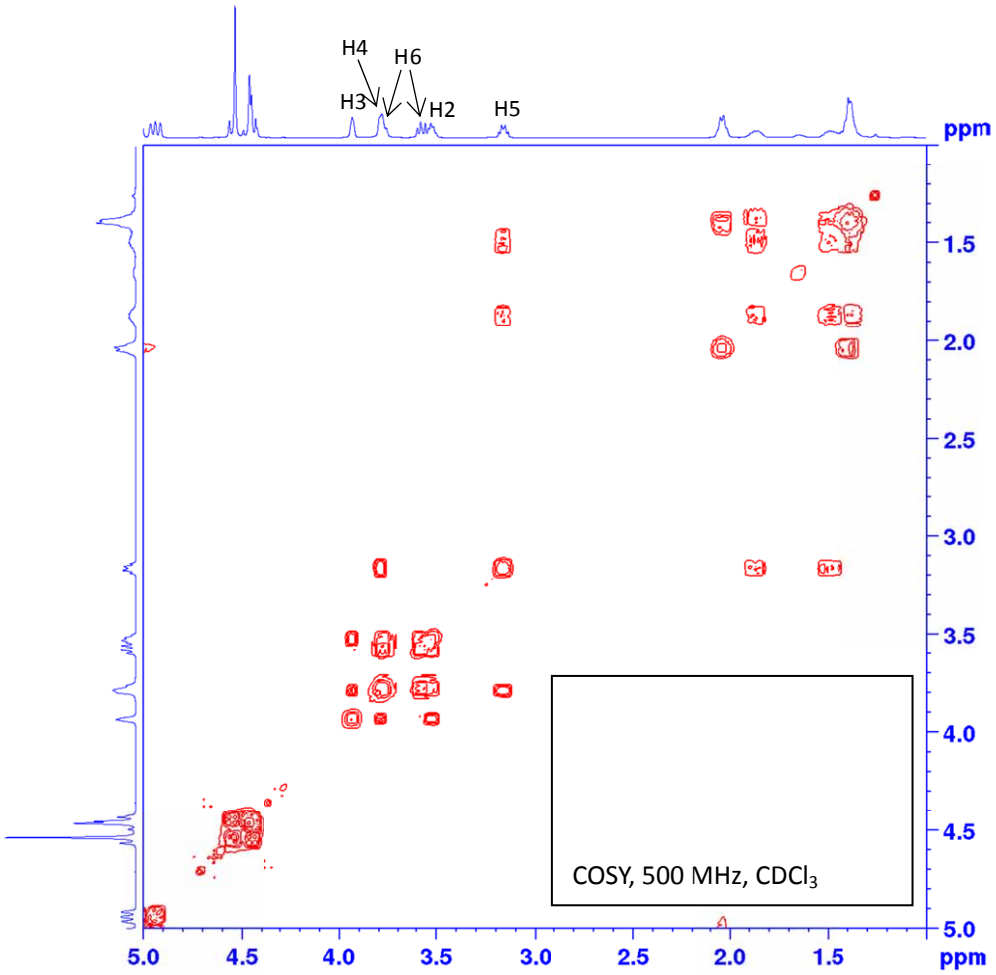
^{1}H NMR, 300 MHz, MeOD

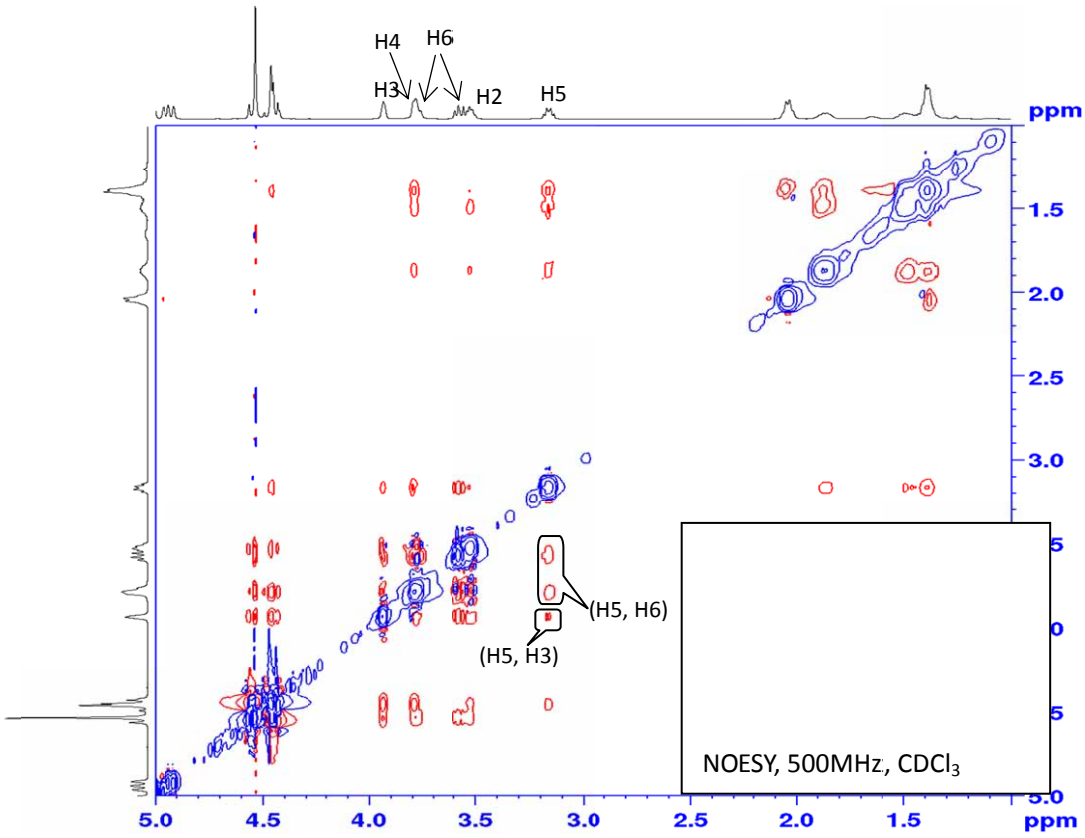


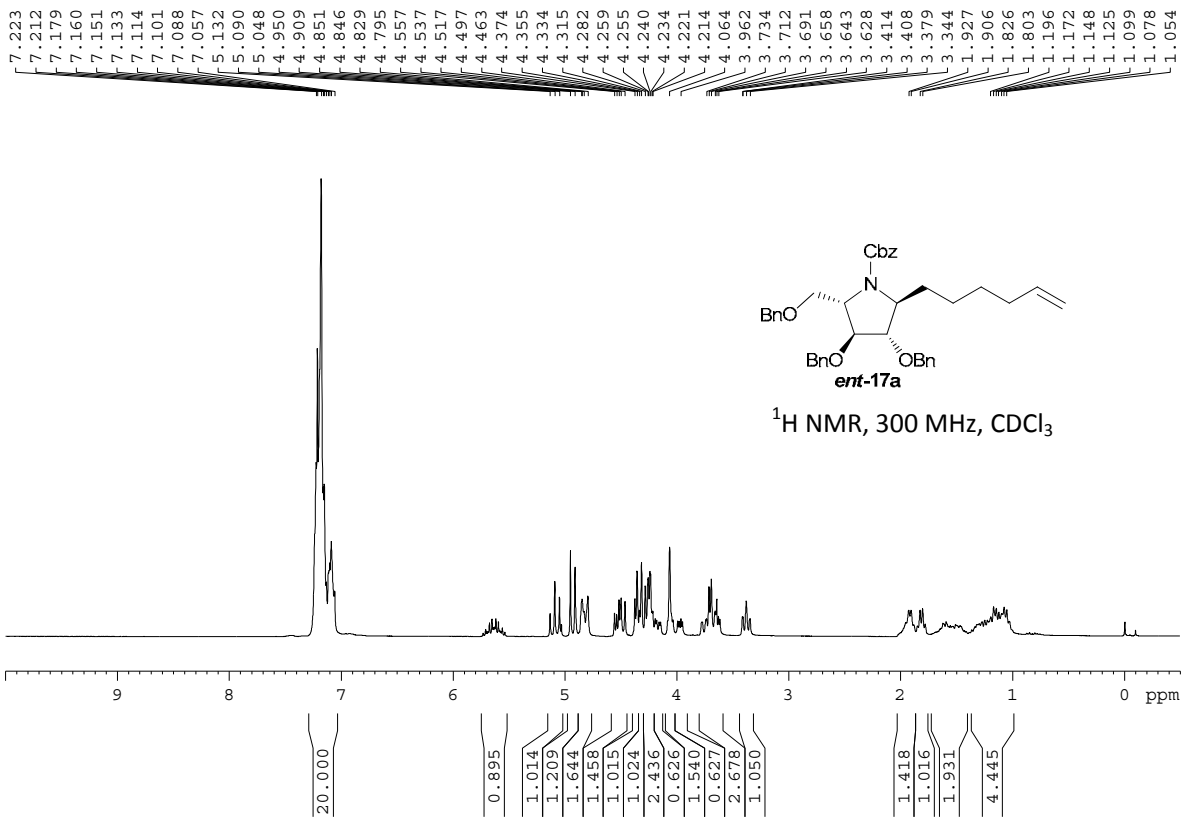
***ent*-3-*epi*-4**

^{13}C NMR, 75 MHz, MeOD

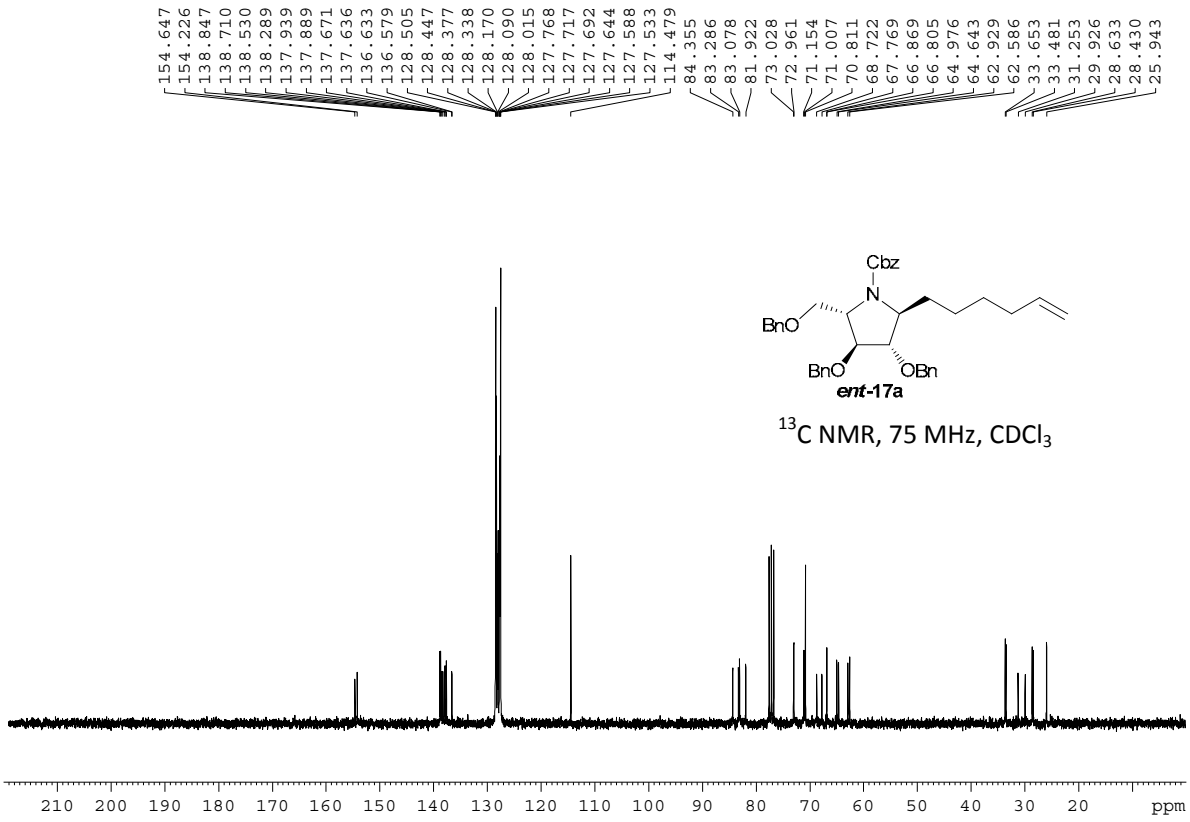

7.319
7.308
7.285
7.277
7.245
6.144
5.834
5.817
5.809
5.800
5.792
5.775
5.766
5.758
5.750
5.733
5.005
4.962
4.940
4.914
4.564
4.535
4.493
4.462
4.452
4.431
4.423
3.932
3.792
3.784
3.761
3.594
3.577
3.554
3.538
3.526
3.514
3.499
3.184
3.170
3.151
3.137
2.045
2.029
1.873
1.856
1.648
1.510
1.495
1.481
1.462
1.442
1.428
1.394
1.384
1.378
1.340
1.255
OH
N
BnO
BnO
OBn
ent-16a
1H NMR, 400 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
15.730
0.988
0.993
1.966
6.288
1.031
2.061
1.036
1.062
1.000
2.064
1.070
1.140
4.056
139.061
138.314
138.199
128.496
128.465
128.111
127.994
127.837
127.801
127.742
114.505
86.848
84.743
73.462
71.816
71.757
70.216
70.074
68.465
33.824
29.180
28.611
26.155
OH
N
BnO
BnO
OBn
ent-16a
13C NMR, 101 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


H4
H6
H3
H2
H5
ppm
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
COSY, 500 MHz, CDCl3

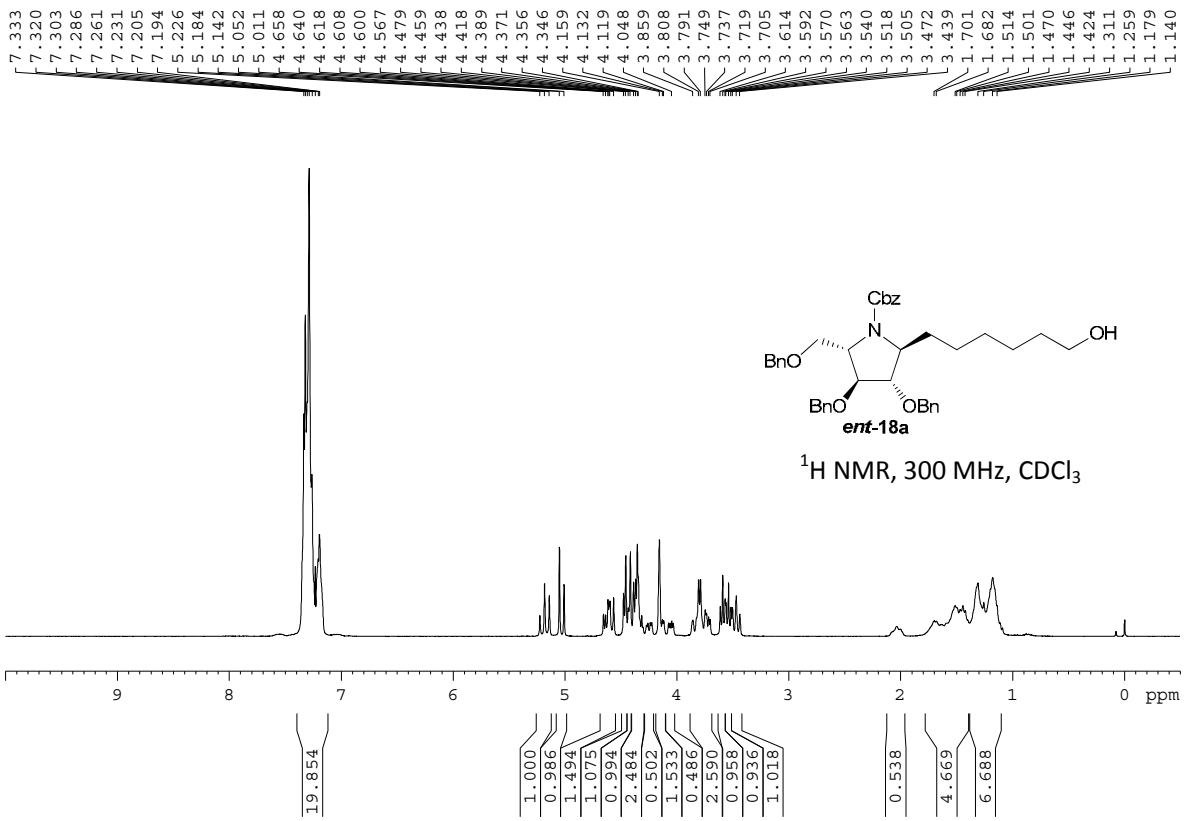

H4
H6
H3
H2
H5
ppm
(H5, H6)
(H5, H3)
NOESY, 500MHz, CDCl3

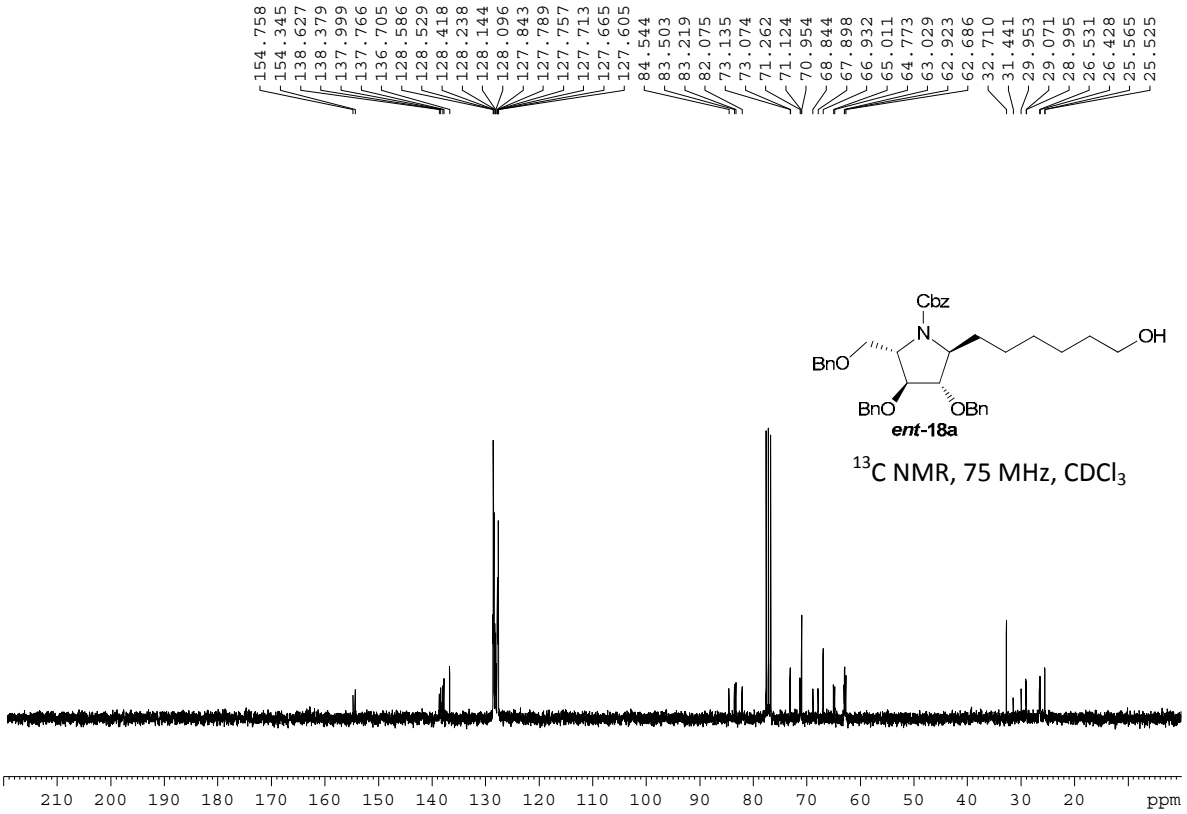


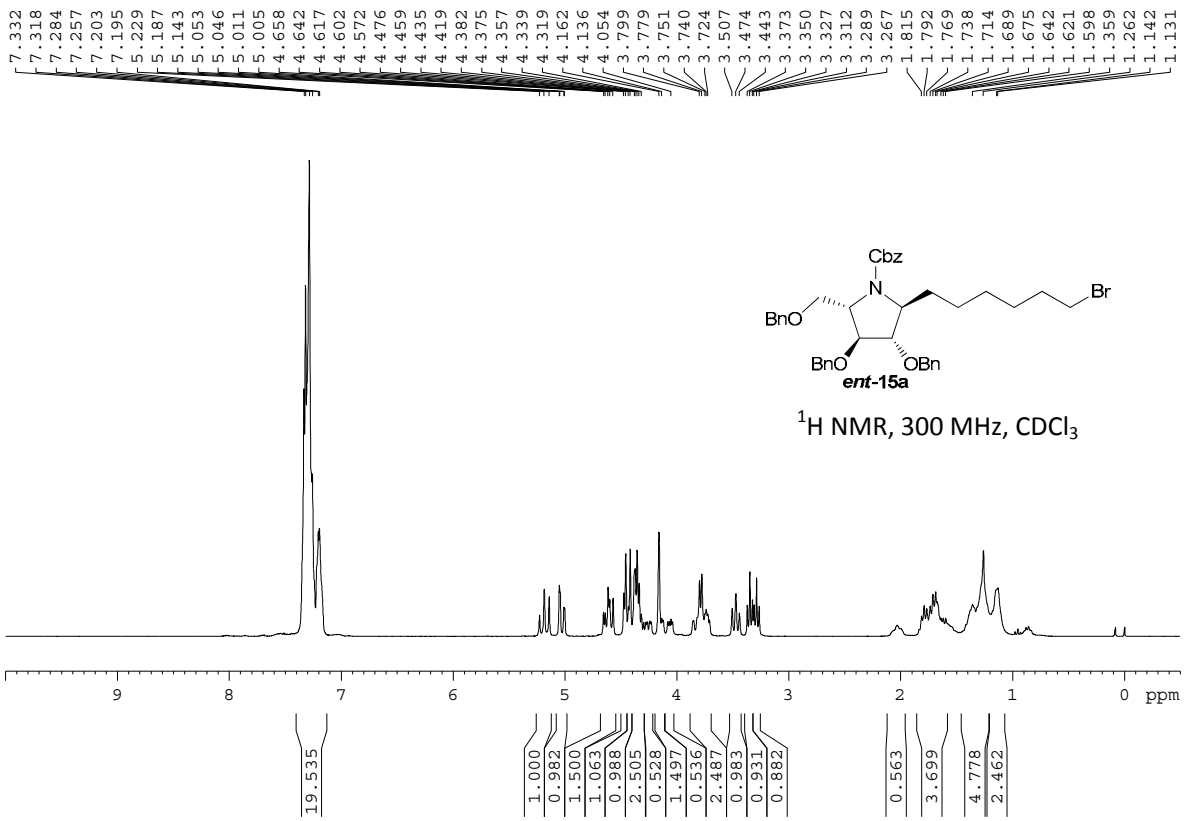
^{1}H NMR, 300 MHz, $CDCl_3$



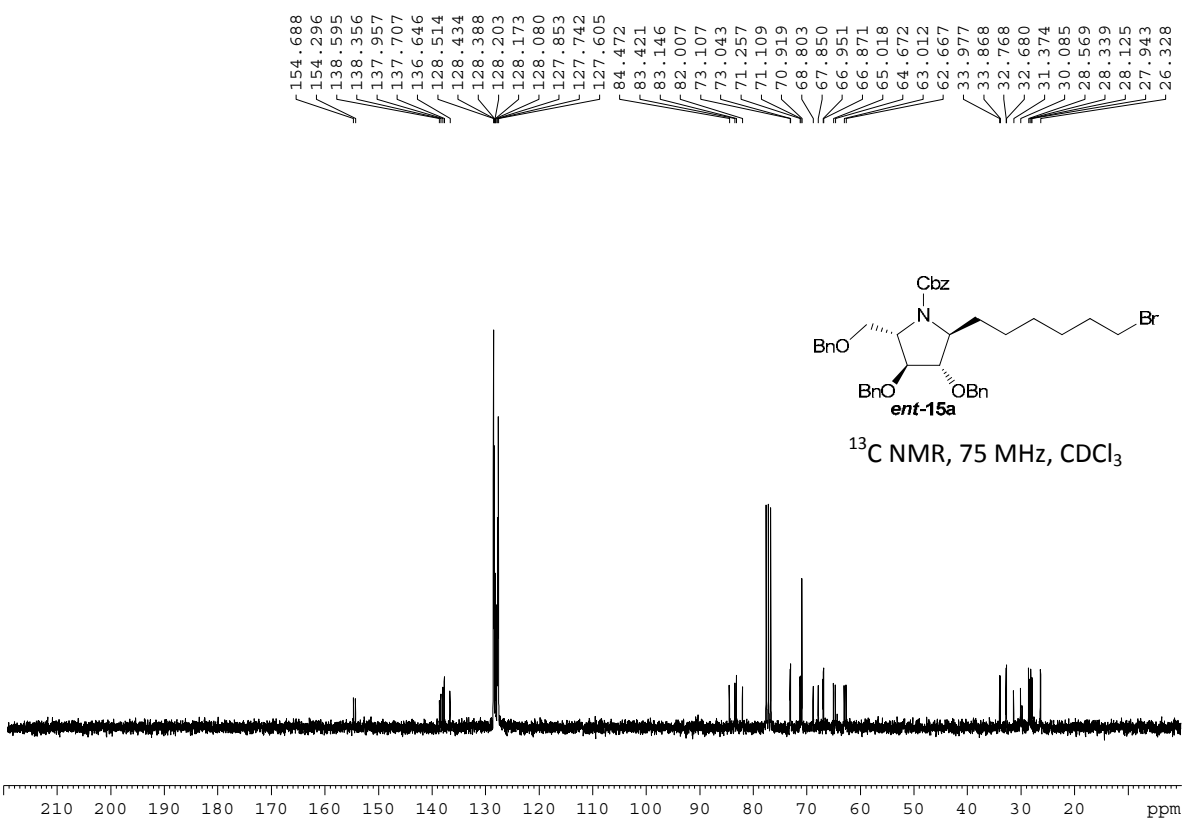
^{13}C NMR, 75 MHz, $CDCl_3$


7.333 7.320 7.303 7.286 7.261 7.231 7.205 7.194 5.226 5.184 5.142 5.052 5.011 4.658 4.640 4.618 4.608 4.600 4.567 4.479 4.459 4.438 4.418 4.389 4.371 4.356 4.346 4.159 4.132 4.119 4.048 3.859 3.808 3.791 3.749 3.737 3.719 3.705 3.614 3.592 3.570 3.563 3.540 3.518 3.505 3.472 3.439 1.701 1.682 1.514 1.501 1.470 1.446 1.424 1.311 1.259 1.179 1.140
Cbz
N
OH
BnO
BnO
OBn
ent-18a
1H NMR, 300 MHz, CDCl3
9 8 7 6 5 4 3 2 1 0 ppm
19.854
1.000 0.986 1.494 1.075 0.994 2.484 0.502 1.533 0.486 2.590 0.958 0.936 1.018
0.538 4.669 6.688

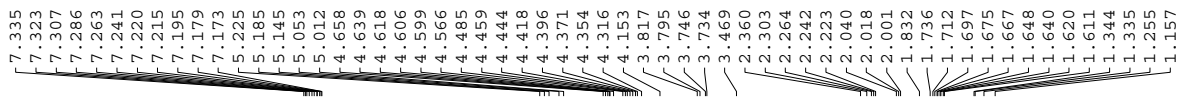

154.758 154.345 138.627 138.379 137.999 137.766 136.705 128.586 128.529 128.418 128.238 128.144 128.096 127.843 127.789 127.757 127.713 127.665 127.605
84.544 83.503 83.219 82.075 73.135 73.074 71.262 71.124 70.954 68.844 67.898 66.932 65.011 64.773 63.029 62.923 62.686
32.710 31.441 29.953 29.071 28.995 26.531 26.428 25.565 25.525
Cbz
N
OH
BnO
BnO
OBn
ent-18a
13C NMR, 75 MHz, CDCl3
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 ppm

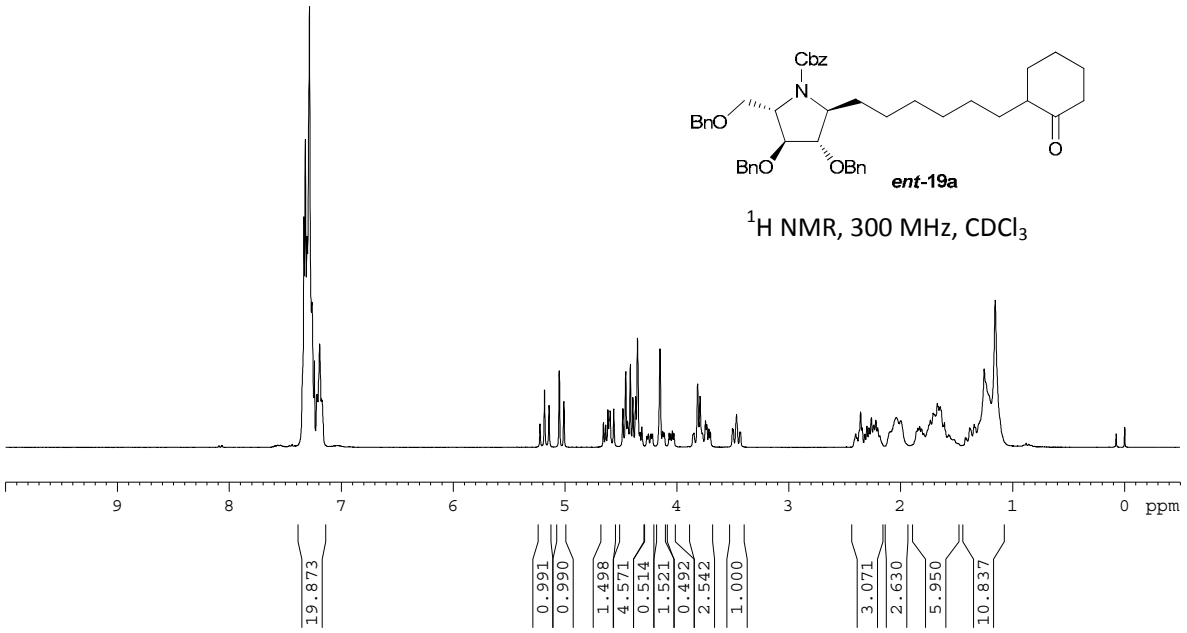


^{1}H NMR, 300 MHz, $CDCl_3$



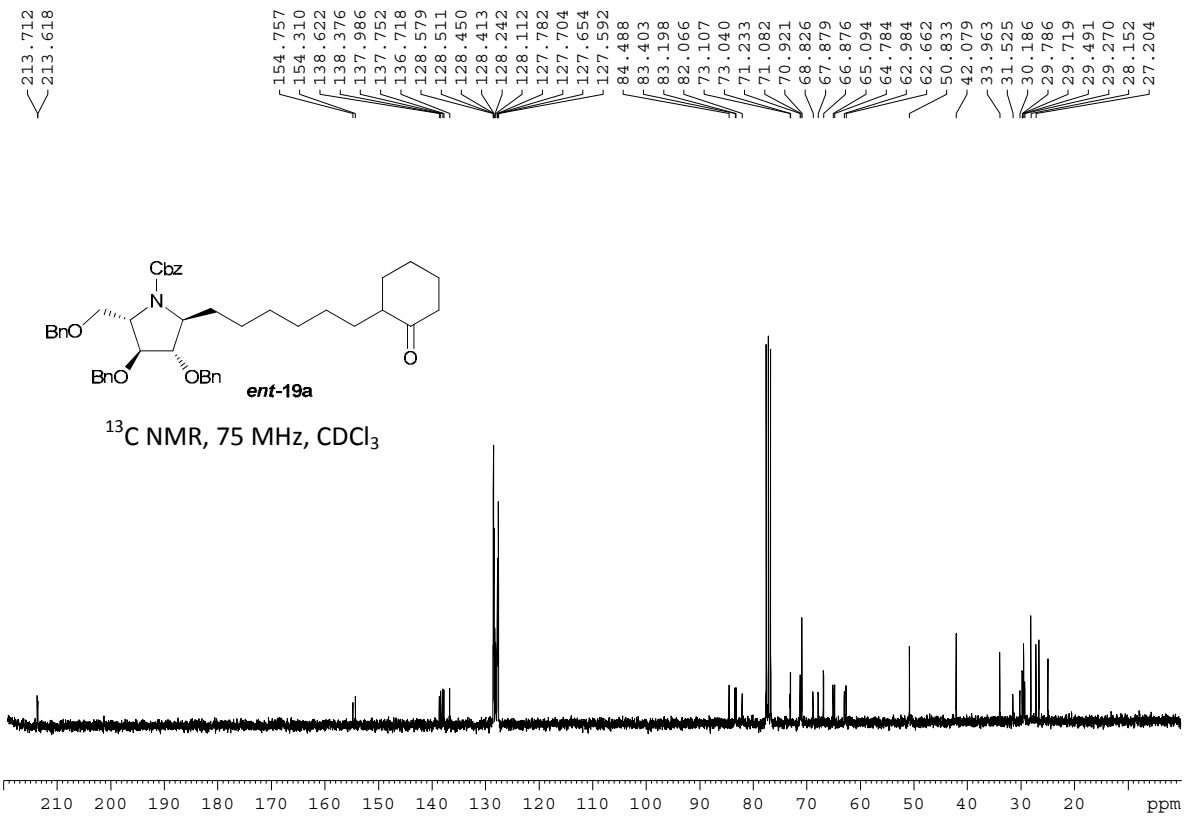
^{13}C NMR, 75 MHz, $CDCl_3$





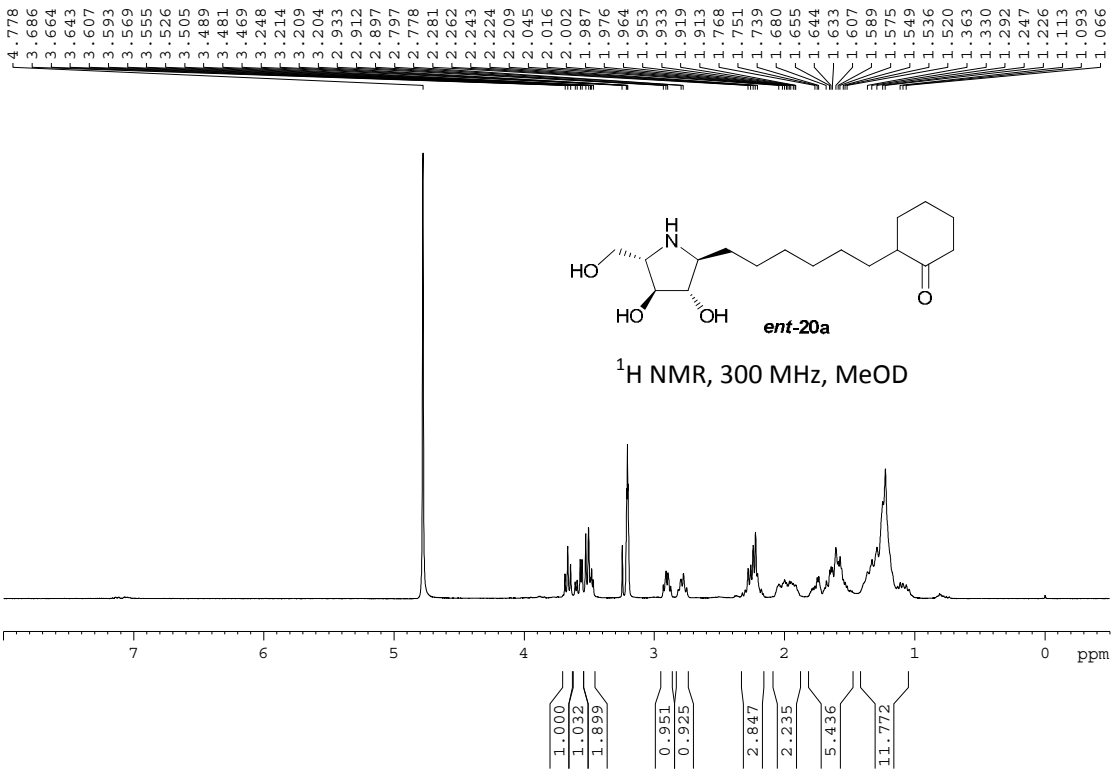
***ent*-19a**

^{1}H NMR, 300 MHz, $CDCl_3$

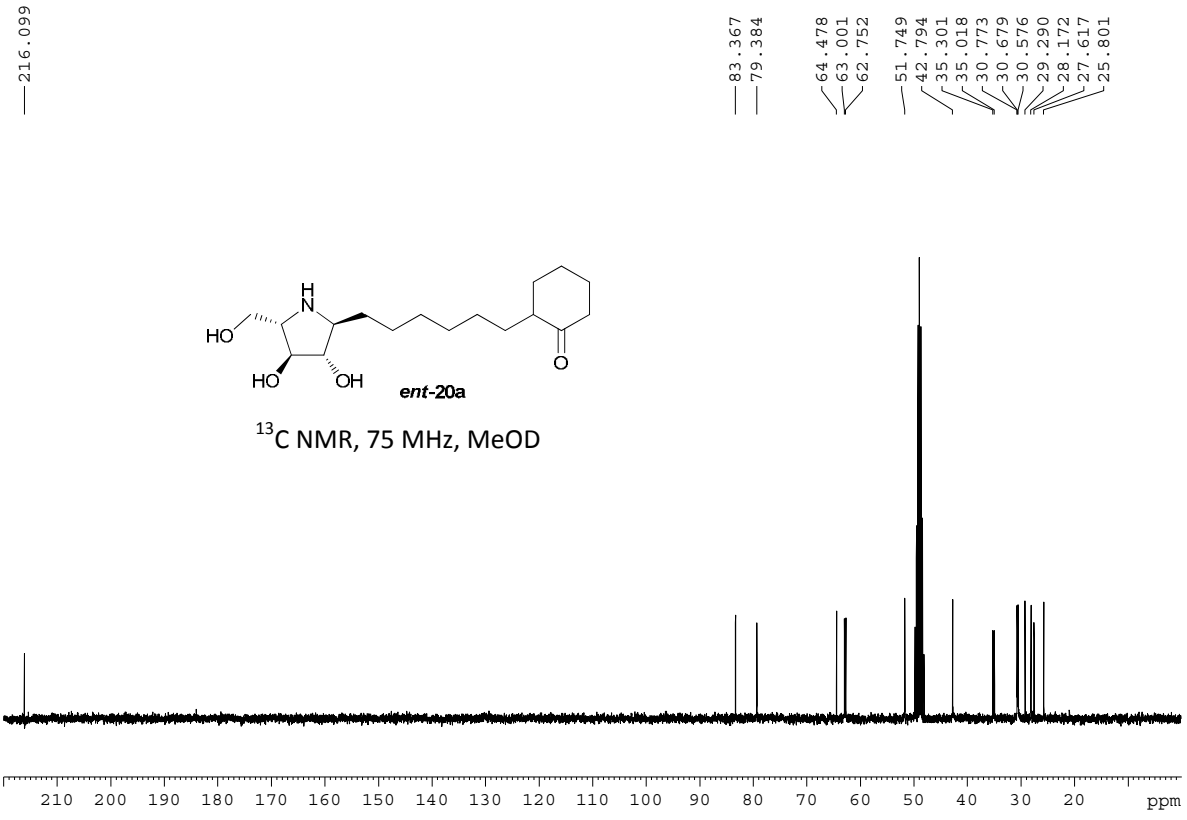


***ent*-19a**

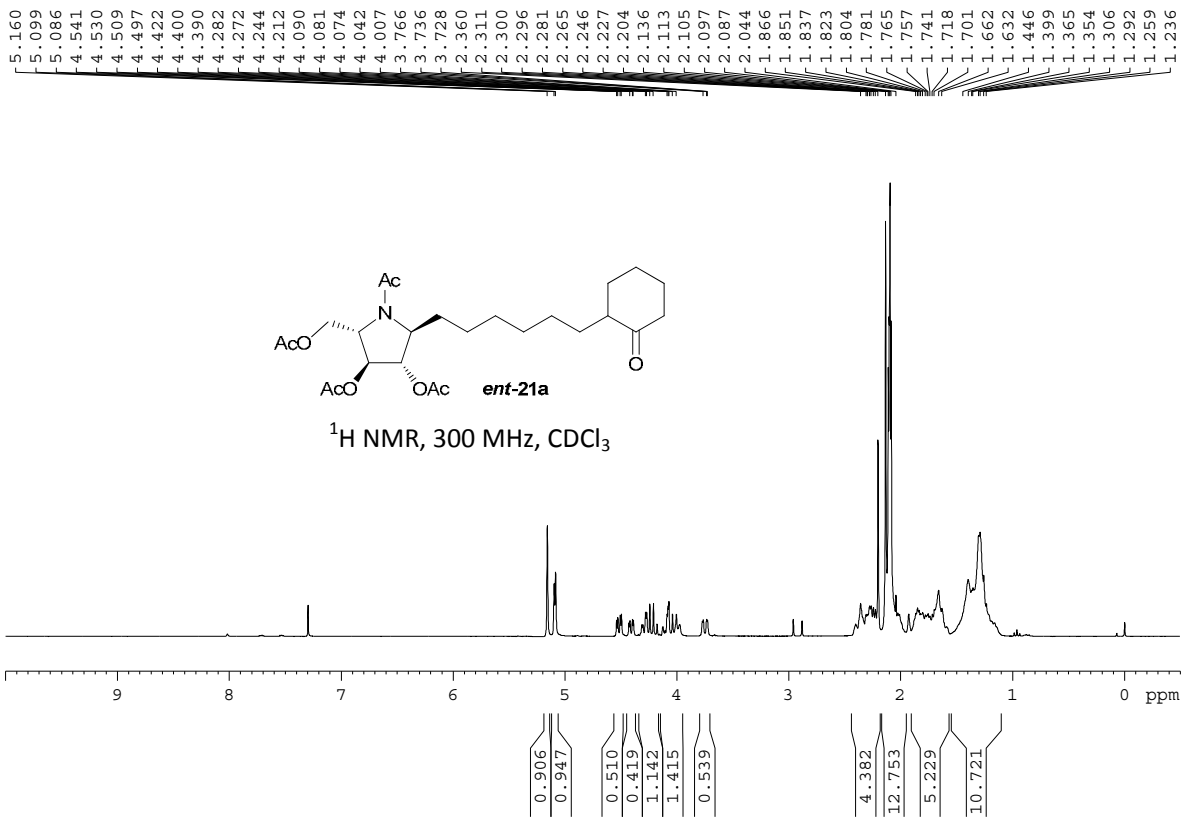
^{13}C NMR, 75 MHz, $CDCl_3$

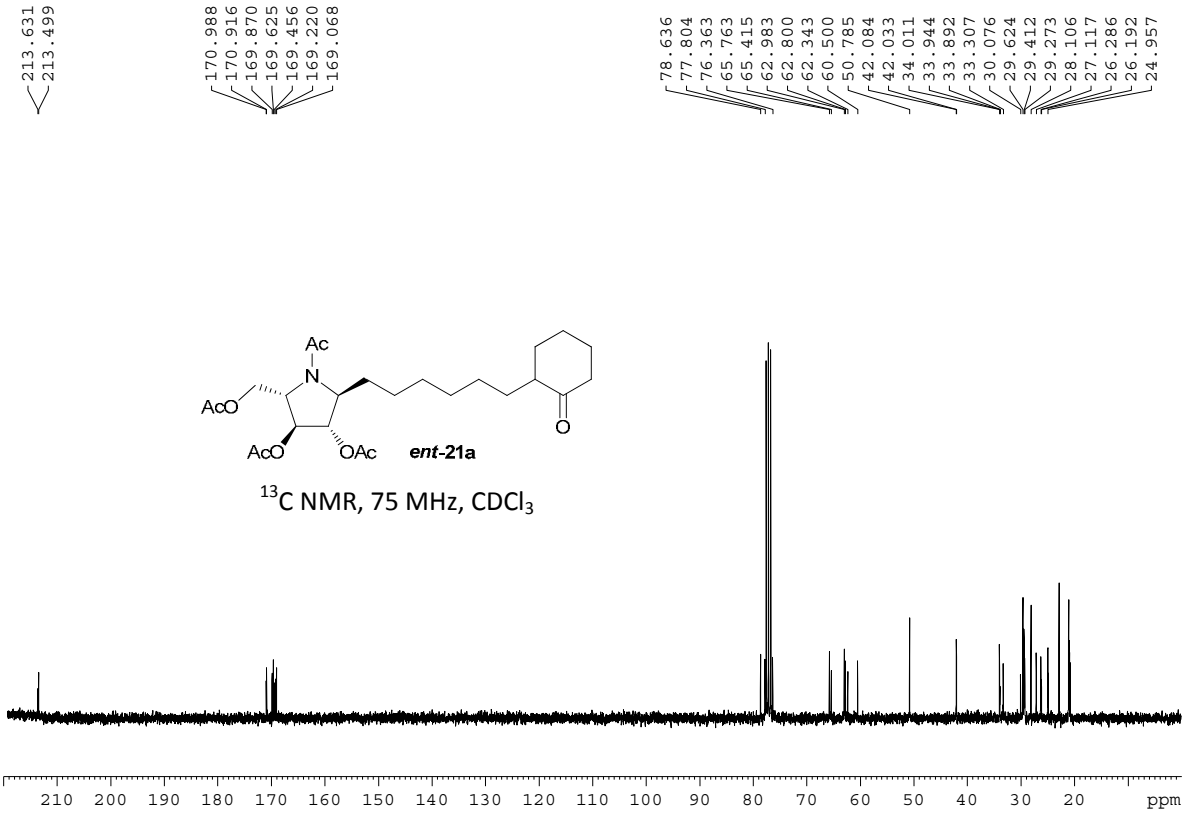


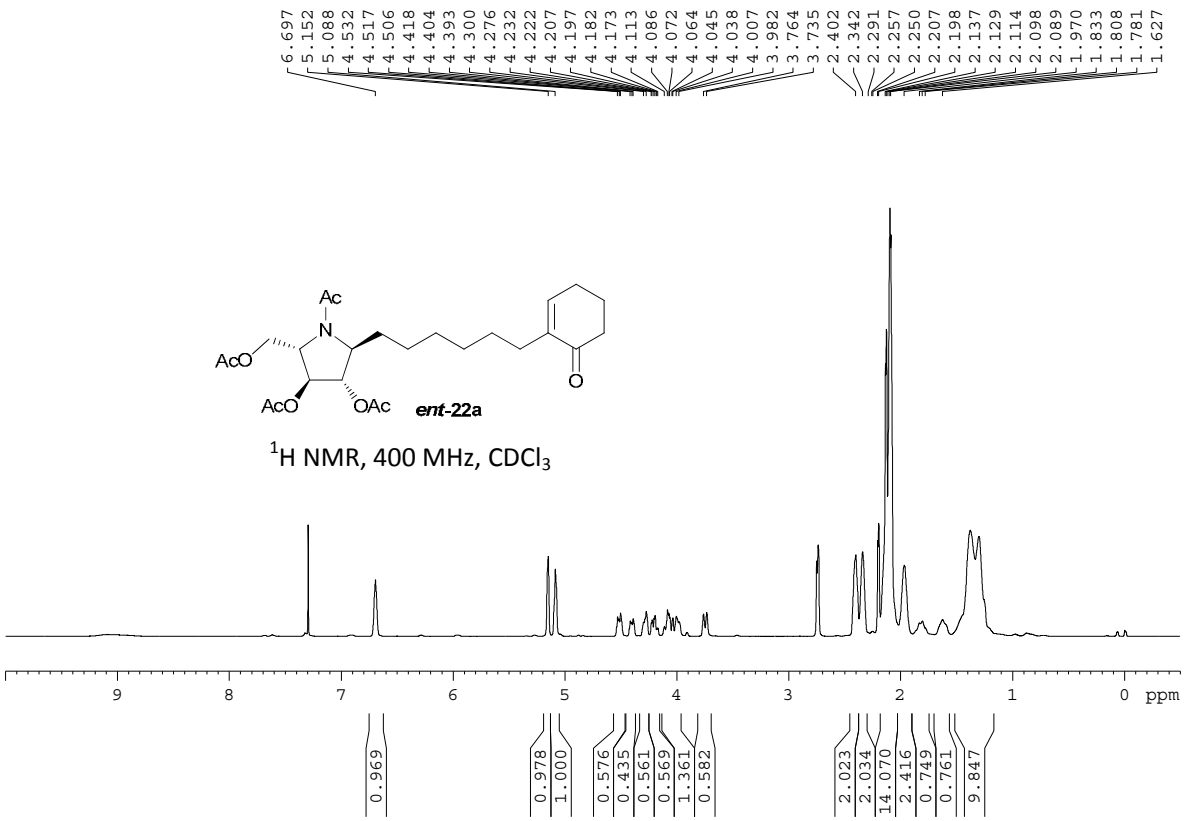
^{1}H NMR, 300 MHz, MeOD

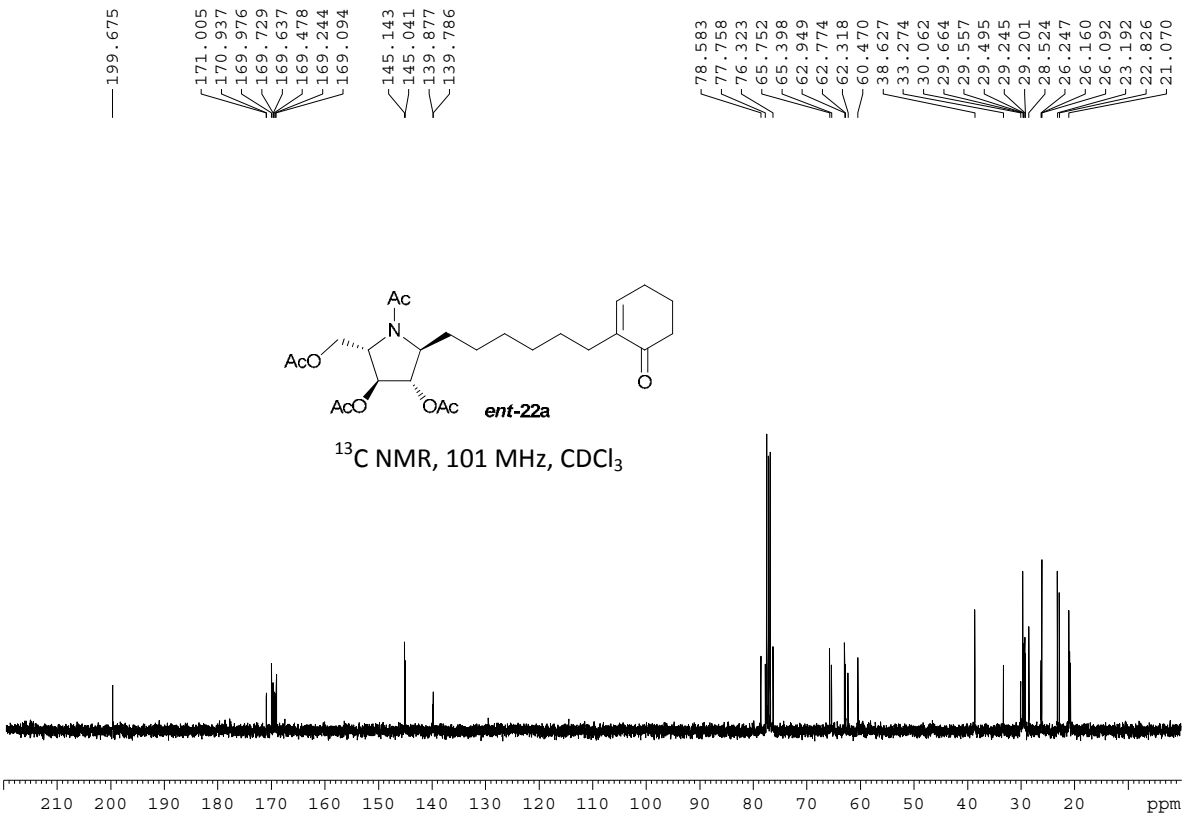


^{13}C NMR, 75 MHz, MeOD


5.160
5.099
5.086
4.541
4.530
4.509
4.497
4.422
4.400
4.390
4.282
4.272
4.244
4.212
4.090
4.081
4.074
4.042
4.007
3.766
3.736
3.728
2.360
2.311
2.300
2.296
2.281
2.265
2.246
2.227
2.204
2.136
2.113
2.105
2.097
2.087
2.044
1.866
1.851
1.837
1.823
1.804
1.781
1.765
1.757
1.741
1.718
1.701
1.662
1.632
1.446
1.399
1.365
1.354
1.306
1.292
1.259
1.236
Ac
N
AcO
AcO
OAc
O
ent-21a
1H NMR, 300 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
0.906
0.947
0.510
0.419
1.142
1.415
0.539
4.382
12.753
5.229
10.721


213.631
213.499
170.988
170.916
169.870
169.625
169.456
169.220
169.068
78.636
77.804
76.363
65.763
65.415
62.983
62.800
62.343
60.500
50.785
42.084
42.033
34.011
33.944
33.892
33.307
30.076
29.624
29.412
29.273
28.106
27.117
26.286
26.192
24.957
Ac
N
AcO
AcO
OAc
O
ent-21a
13C NMR, 75 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm


6.697
5.152
5.088
4.532
4.517
4.506
4.418
4.404
4.393
4.300
4.276
4.232
4.222
4.207
4.197
4.182
4.173
4.113
4.086
4.072
4.064
4.045
4.038
4.007
3.982
3.764
3.735
2.402
2.342
2.291
2.257
2.250
2.207
2.198
2.137
2.129
2.114
2.098
2.089
1.970
1.833
1.808
1.781
1.627
Ac
N
AcO
AcO
OAc
O
ent-22a
1H NMR, 400 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0 ppm
0.969
0.978
1.000
0.576
0.435
0.561
0.569
1.361
0.582
2.023
2.034
14.070
2.416
0.749
0.761
9.847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145.041
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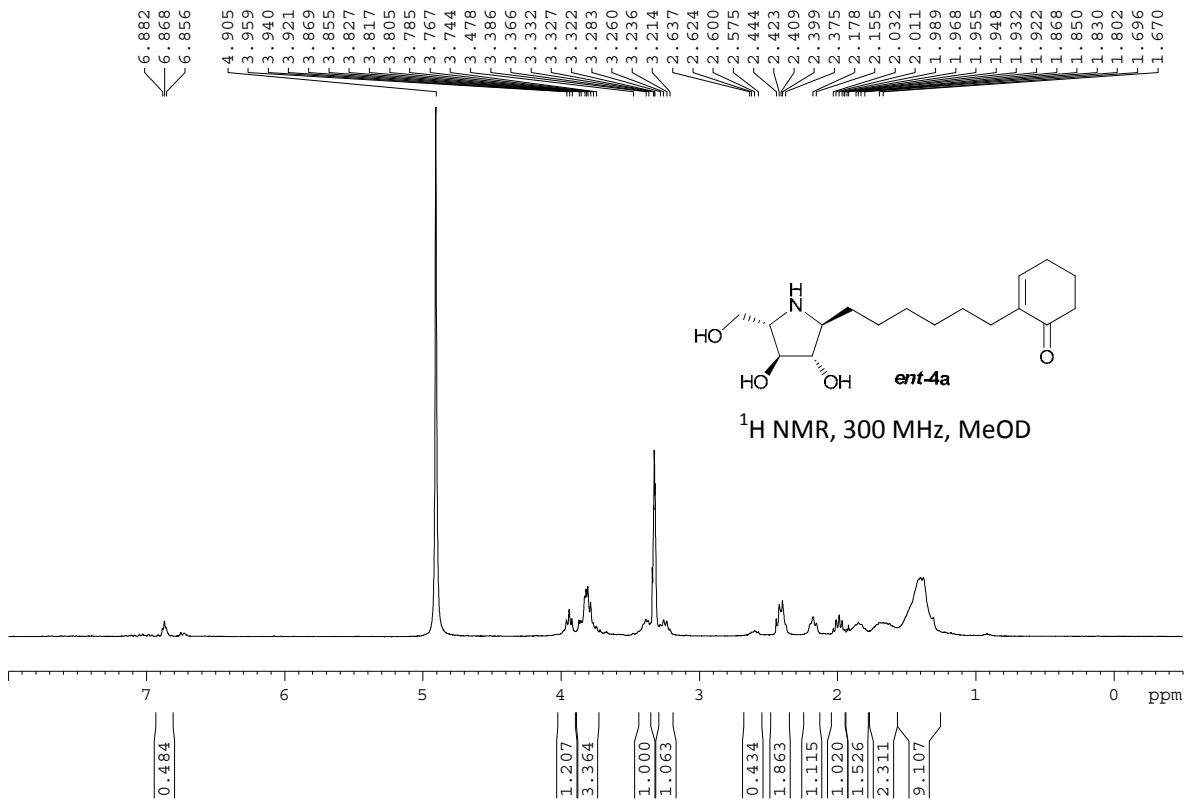
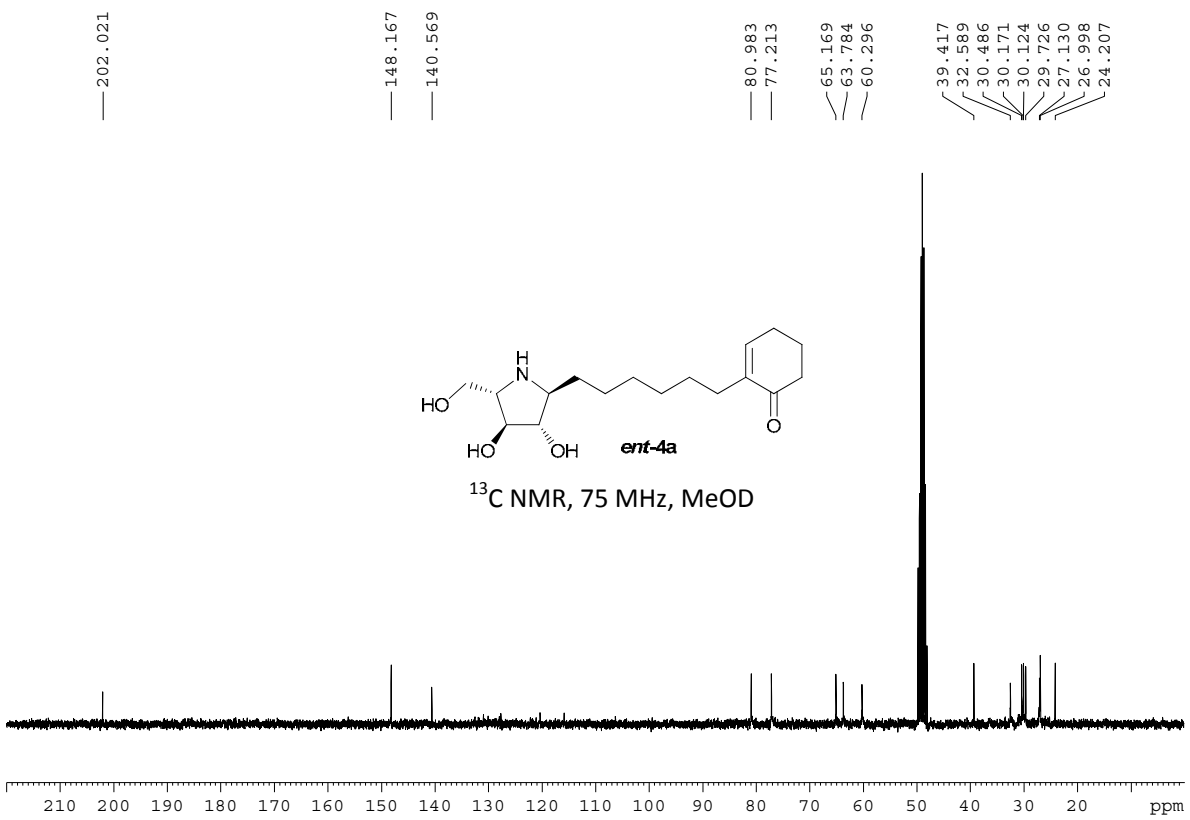
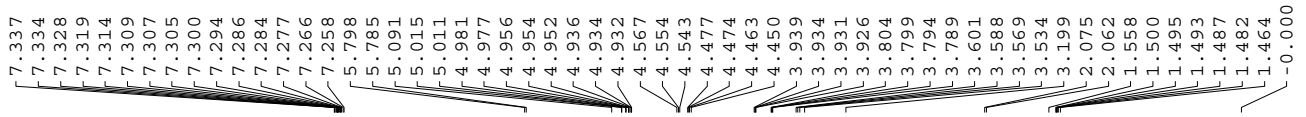
139.786
78.583
77.758
76.323
65.752
65.398
62.949
62.774
62.318
60.470
38.627
33.274
30.062
29.664
29.557
29.495
29.245
29.201
28.524
26.247
26.160
26.092
23.192
22.826
21.070
Ac
N
AcO
AcO
OAc
O
ent-22a
13C NMR, 101 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

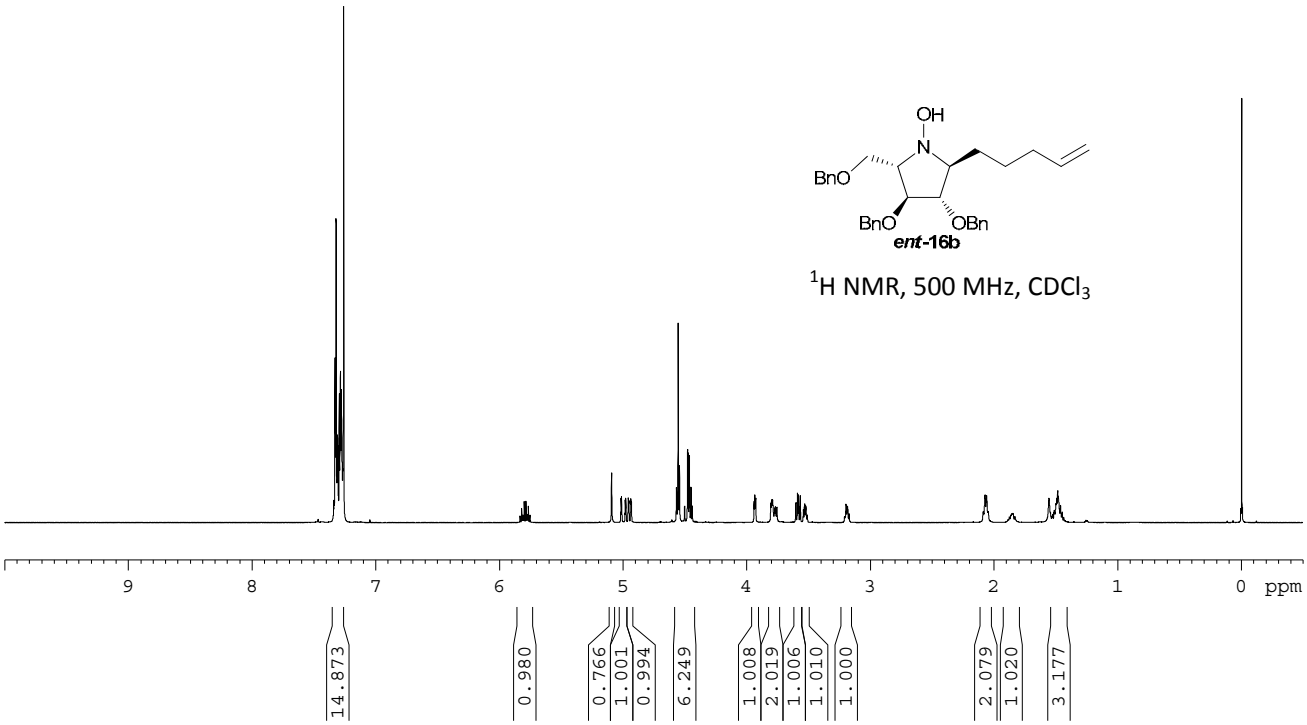


^{1}H NMR, 300 MHz, MeOD

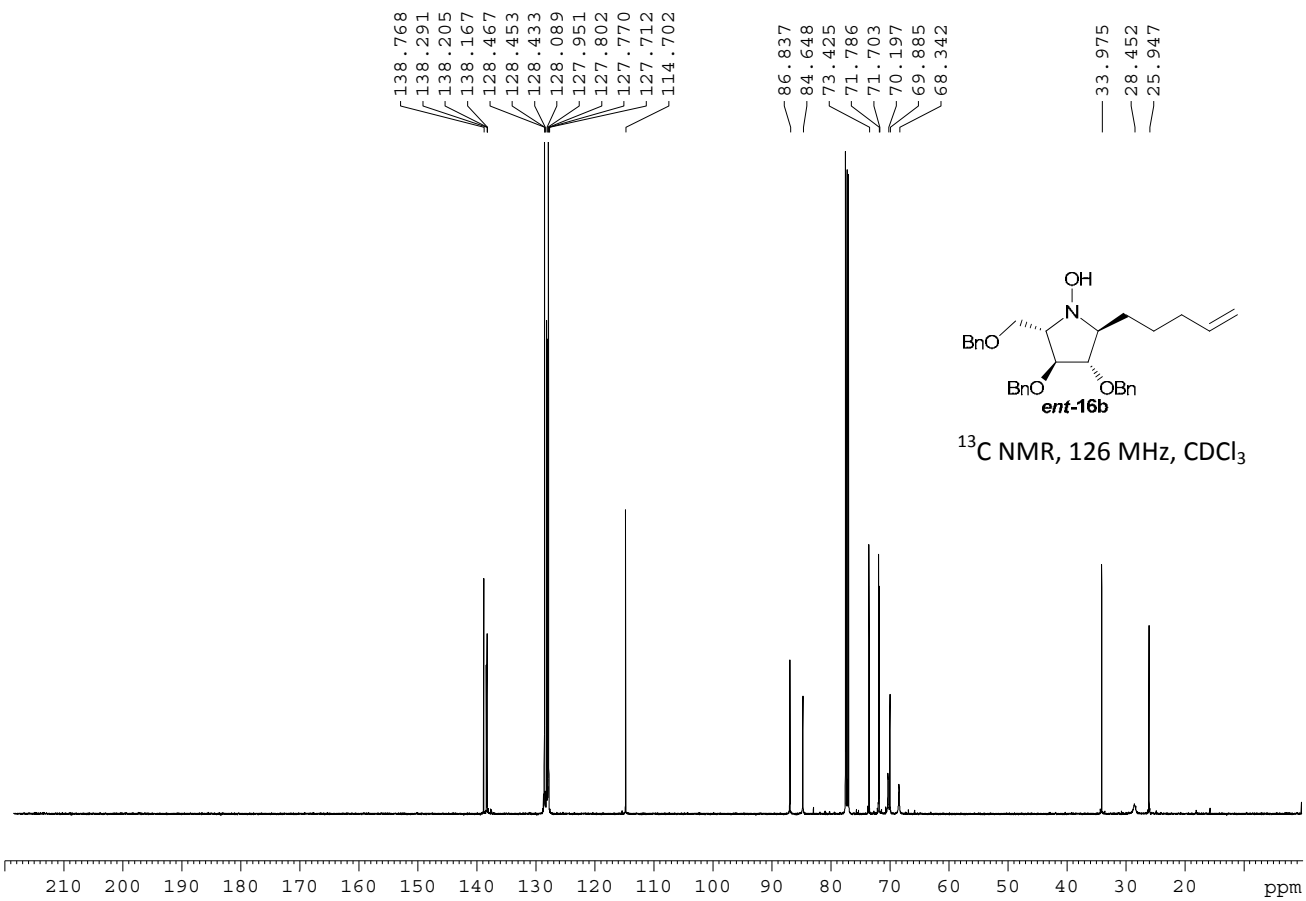


^{13}C NMR, 75 MHz, MeOD

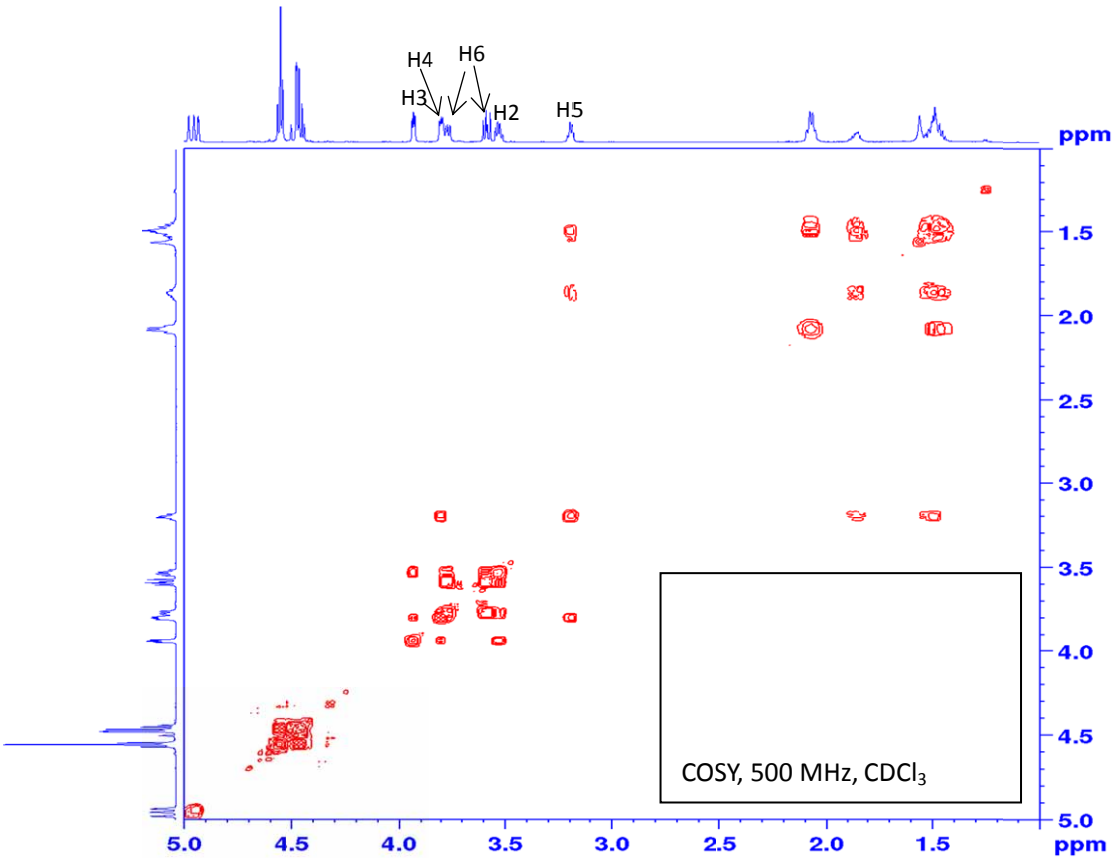


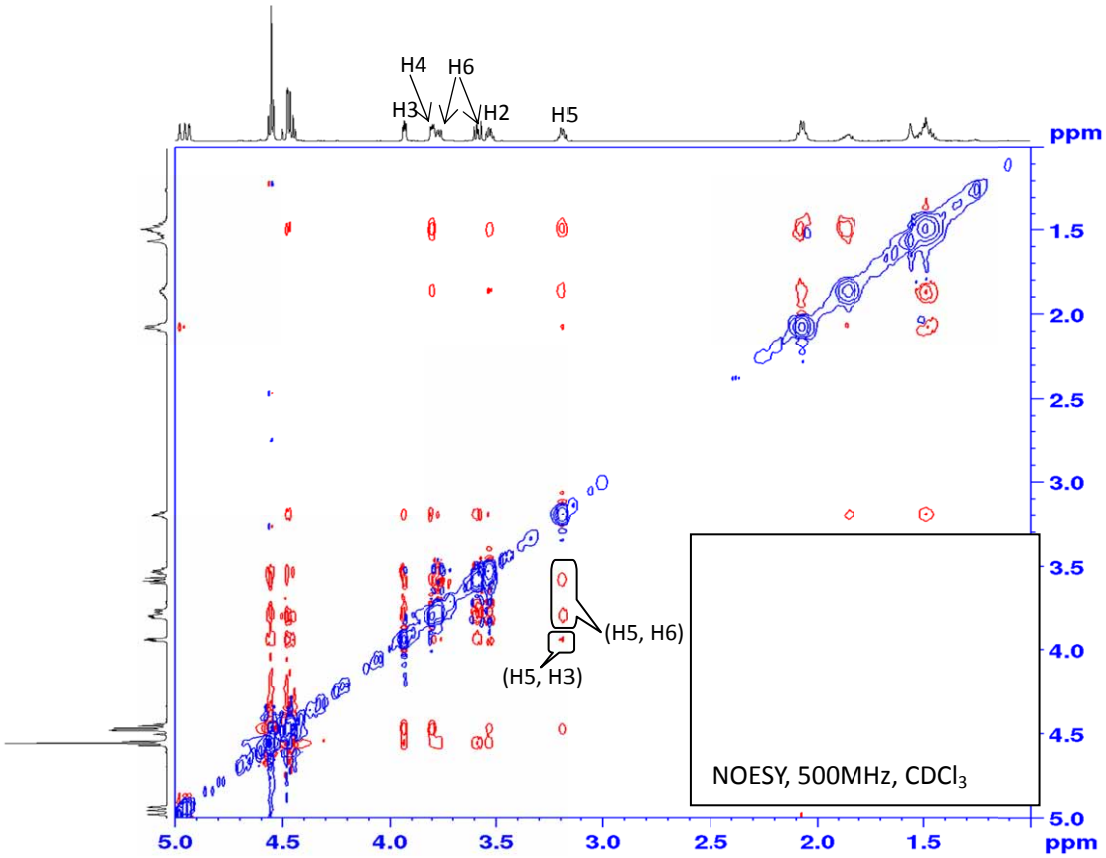


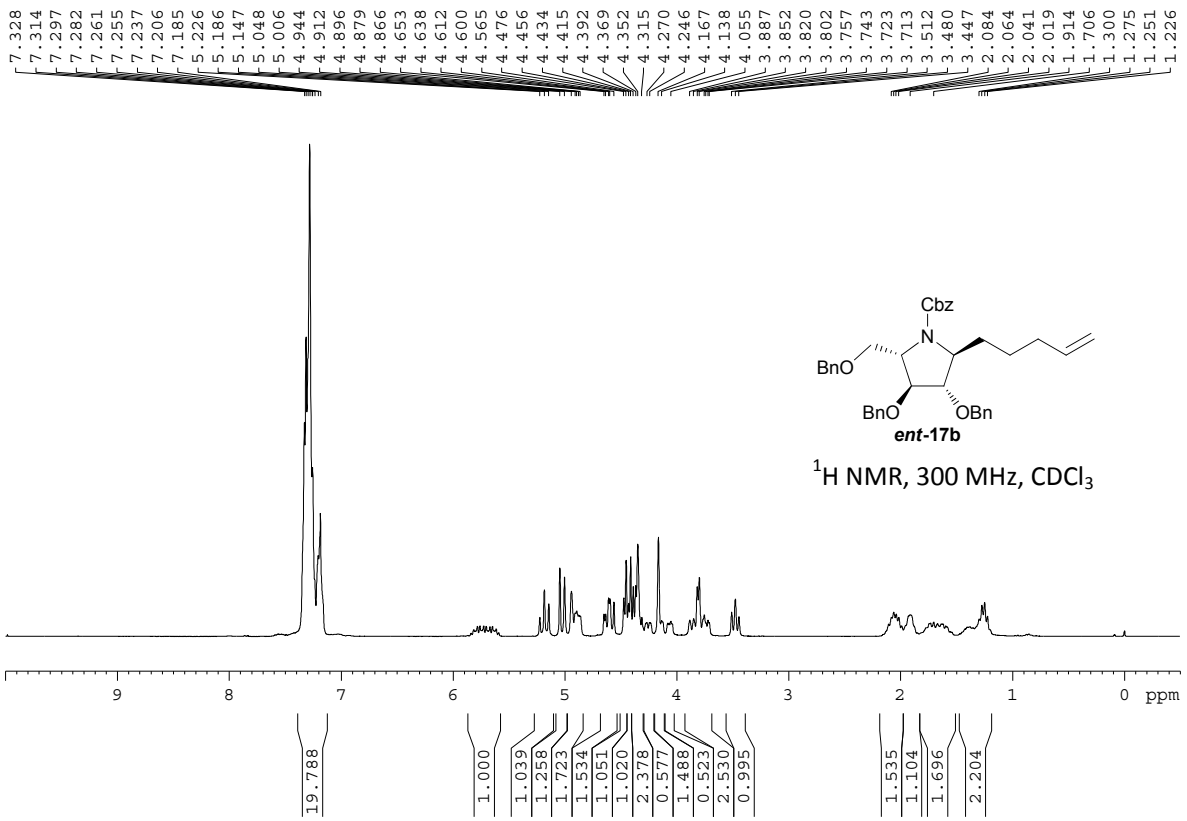
^{1}H NMR, 500 MHz, $CDCl_3$



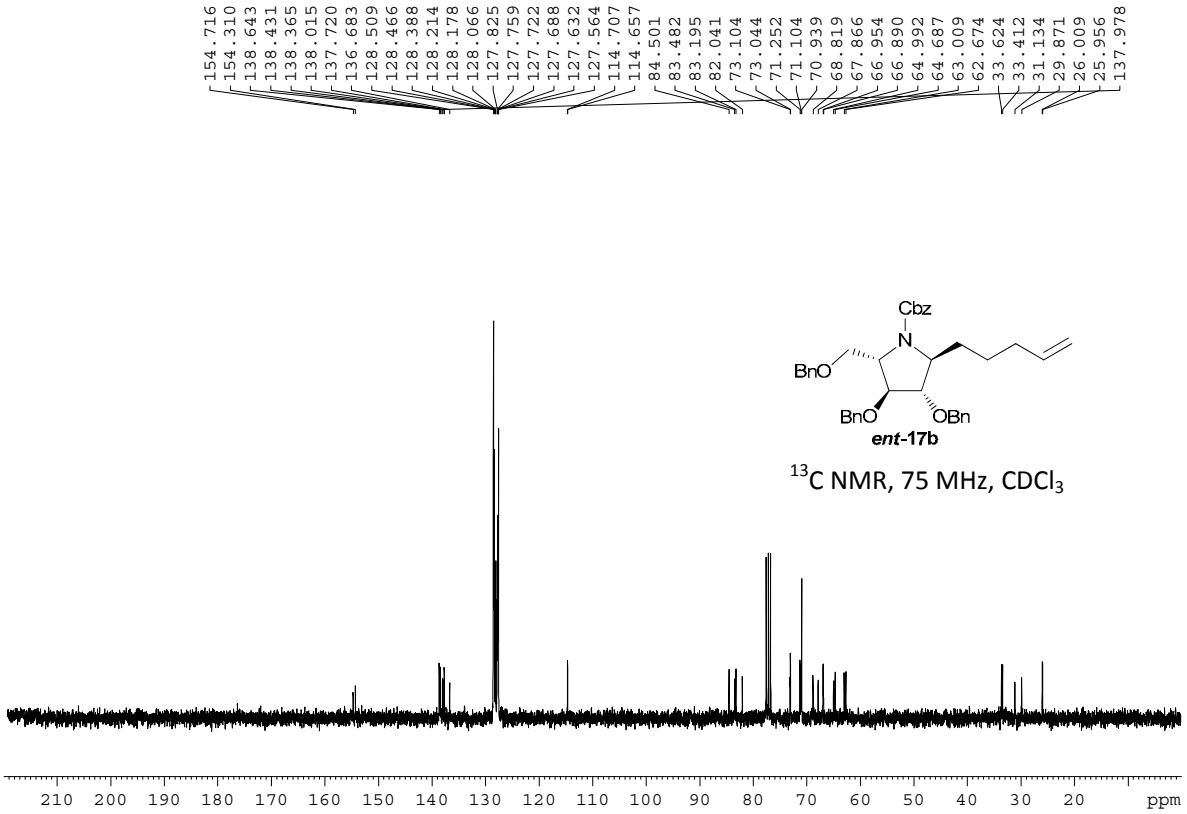
^{13}C NMR, 126 MHz, $CDCl_3$


H4
H6
H3
H2
H5
ppm
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
ppm
COSY, 500 MHz, CDCl3

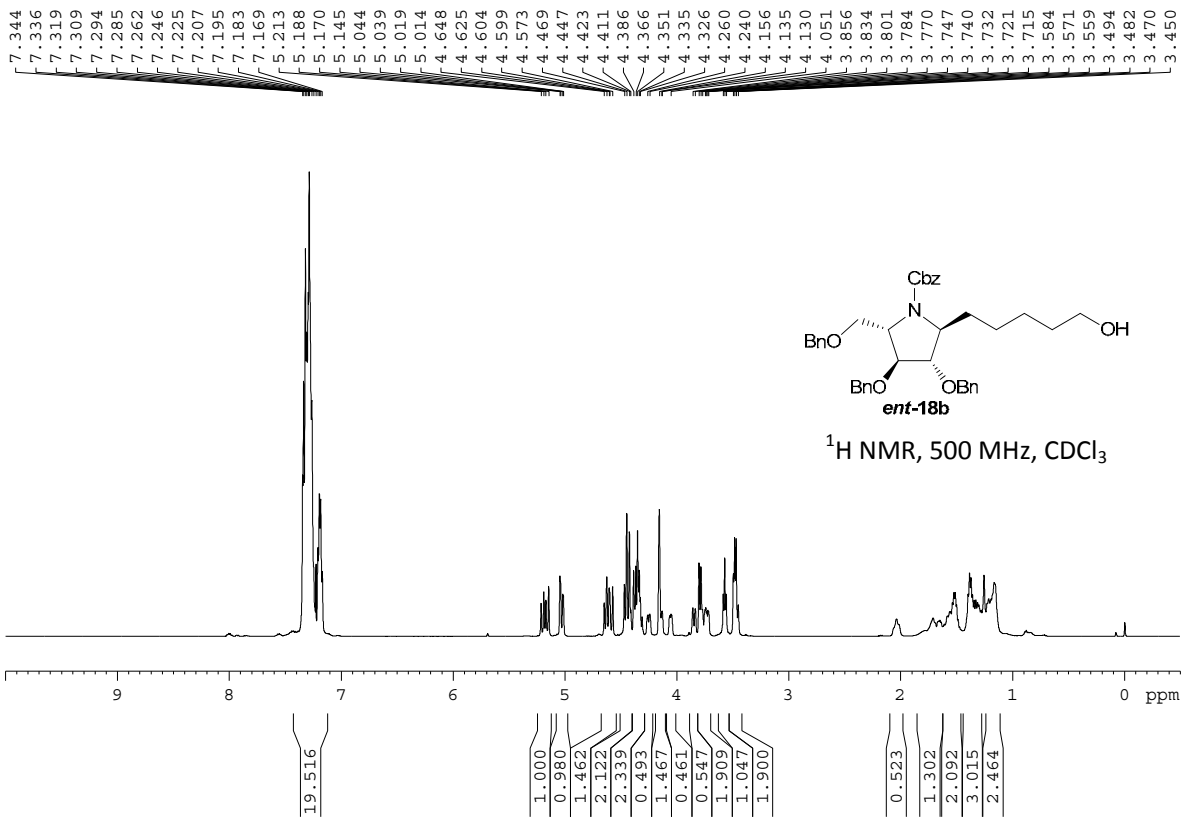

H4
H6
H3
H2
H5
ppm
1.5
2.0
2.5
3.0
3.5
4.0
4.5
5.0
(H5, H6)
(H5, H3)
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
ppm
NOESY, 500MHz, CDCl3



^{1}H NMR, 300 MHz, $CDCl_3$



^{13}C NMR, 75 MHz, $CDCl_3$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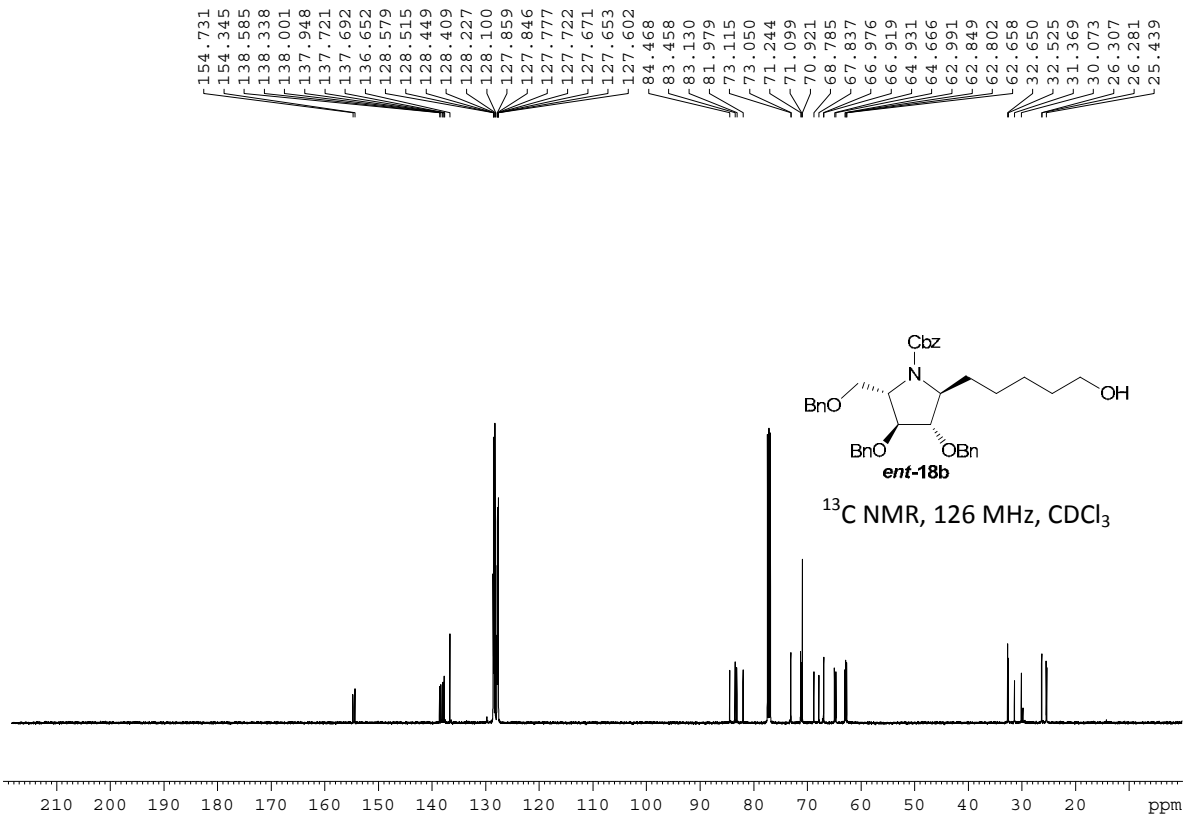
7.309
7.294
7.285
7.262
7.246
7.225
7.207
7.195
7.183
7.169
5.213
5.188
5.170
5.145
5.044
5.039
5.019
5.014
4.648
4.625
4.604
4.599
4.573
4.469
4.447
4.423
4.411
4.386
4.366
4.351
4.335
4.326
4.260
4.240
4.156
4.135
4.130
4.051
3.856
3.834
3.801
3.784
3.770
3.747
3.740
3.732
3.721
3.715
3.584
3.571
3.559
3.494
3.482
3.470
3.450
Cbz
N
BnO
OH
BnO
OBn
ent-18b
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0
ppm
19.516
1.000
0.980
1.462
2.122
2.339
0.493
1.467
0.461
0.547
1.909
1.047
1.900
0.523
1.302
2.092
3.015
2.464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128.515
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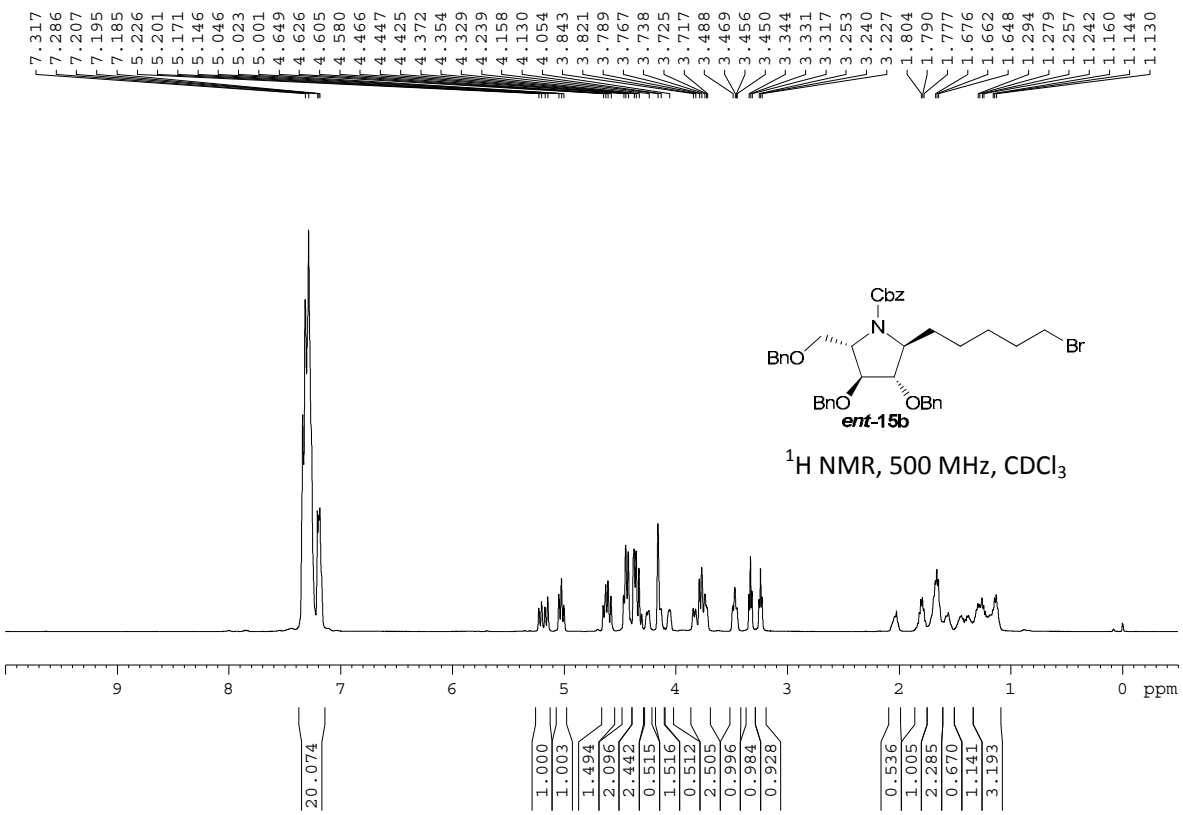
128.227
128.100
127.859
127.846
127.777
127.722
127.671
127.653
127.602
84.468
83.458
83.130
81.979
73.115
73.050
71.244
71.099
70.921
68.785
67.837
66.976
66.919
64.931
64.666
62.991
62.849
62.802
62.658
32.650
32.525
31.369
30.073
26.307
26.281
25.439
Cbz
N
BnO
OH
BnO
OBn
ent-18b
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm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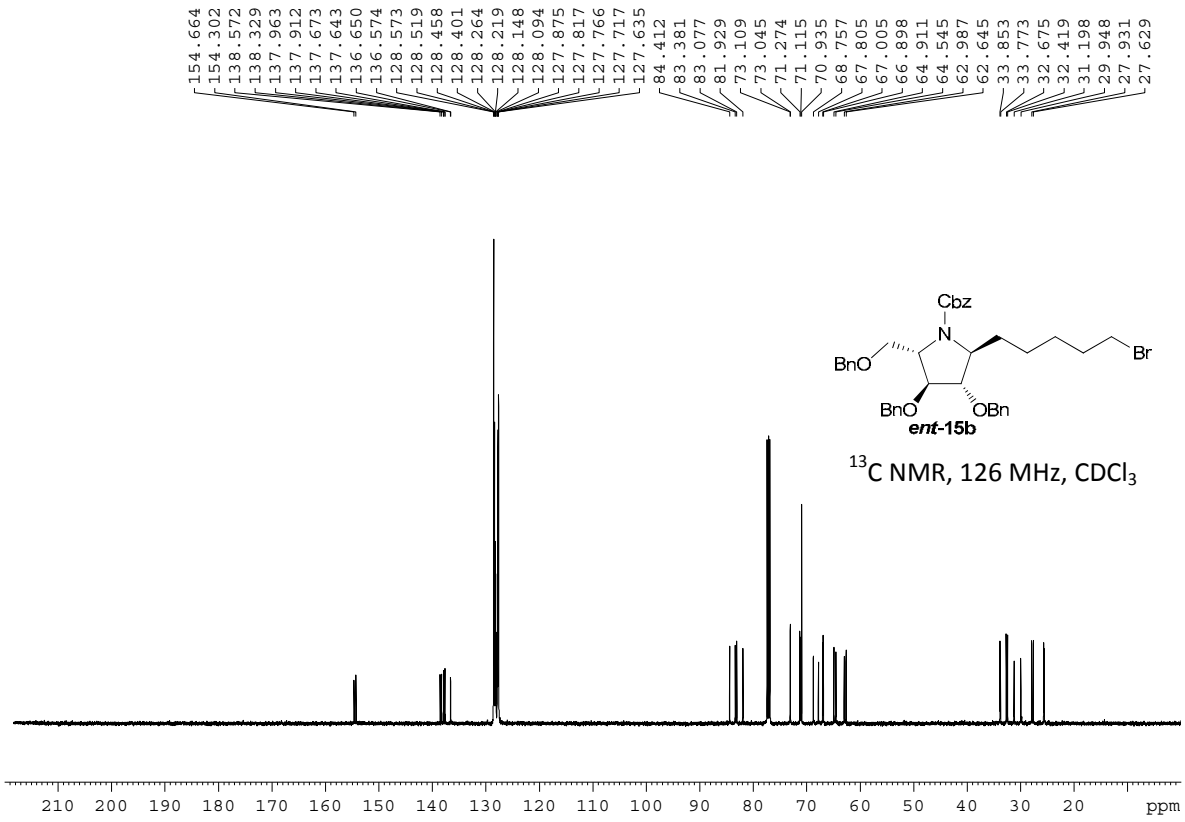
7.286
7.207
7.195
7.185
5.226
5.201
5.171
5.146
5.046
5.023
5.001
4.649
4.626
4.605
4.580
4.466
4.447
4.425
4.372
4.354
4.329
4.239
4.158
4.130
4.054
3.843
3.821
3.789
3.767
3.738
3.725
3.717
3.488
3.469
3.456
3.450
3.344
3.331
3.317
3.253
3.240
3.227
1.804
1.790
1.777
1.676
1.662
1.648
1.294
1.279
1.257
1.242
1.160
1.144
1.130
Cbz
N
BnO
Br
BnO
OBn
ent-15b
1H NMR, 500 MHz, CDCl3
9
8
7
6
5
4
3
2
1
0 ppm
20.074
1.000
1.003
1.494
2.096
2.442
0.515
1.516
0.512
2.505
0.996
0.984
0.928
0.536
1.005
2.285
0.670
1.141
3.193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137.912
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137.643
136.650
136.574
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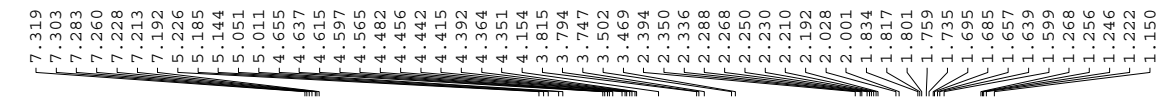
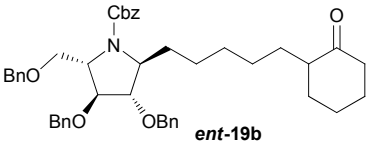
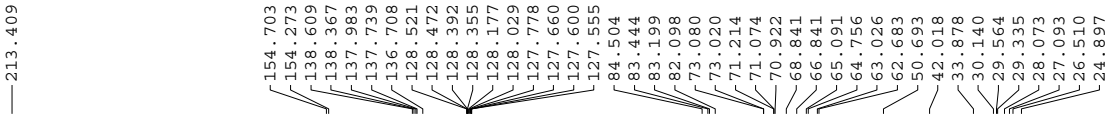
128.519
128.458
128.401
128.264
128.219
128.148
128.094
127.875
127.817
127.766
127.717
127.635
84.412
83.381
83.077
81.929
73.109
73.045
71.274
71.115
70.935
68.757
67.805
67.005
66.898
64.911
64.545
62.987
62.645
33.853
33.773
32.675
32.419
31.198
29.948
27.931
27.629
Cbz
N
BnO
Br
BnO
OBn
ent-15b
13C NMR, 126 MHz, CDCl3
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
ppm

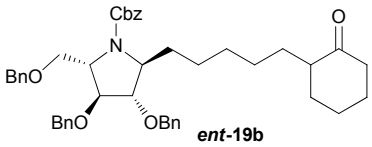




***ent*-19b**

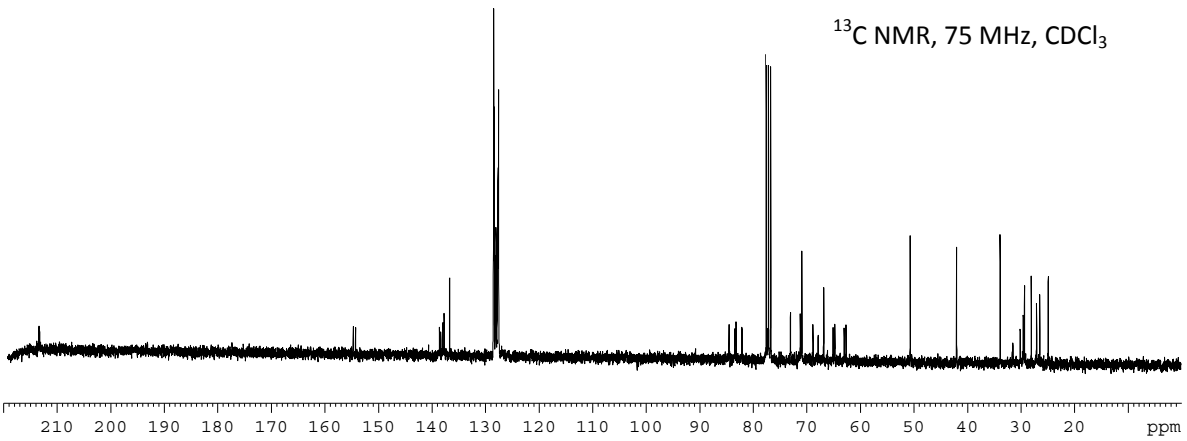
^{1}H NMR, 300 MHz, $CDCl_3$

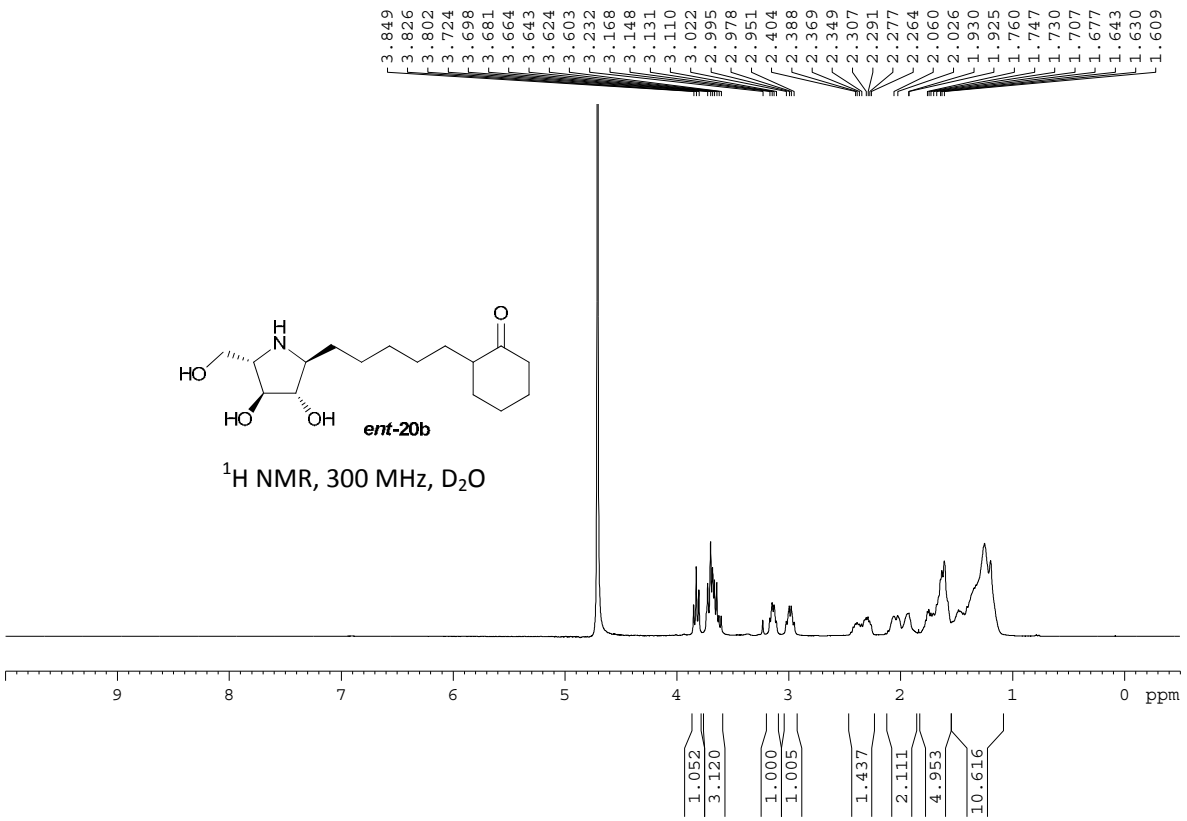




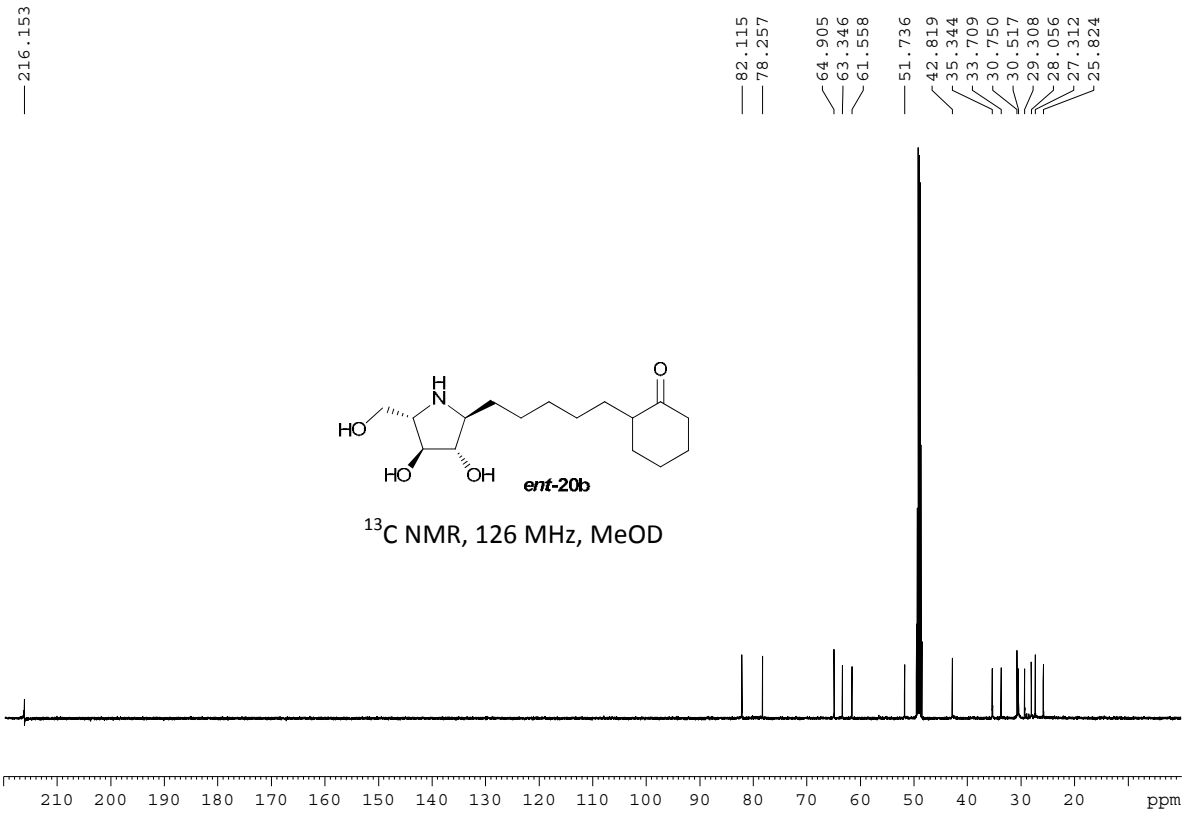
***ent*-19b**

^{13}C NMR, 75 MHz, $CDCl_3$

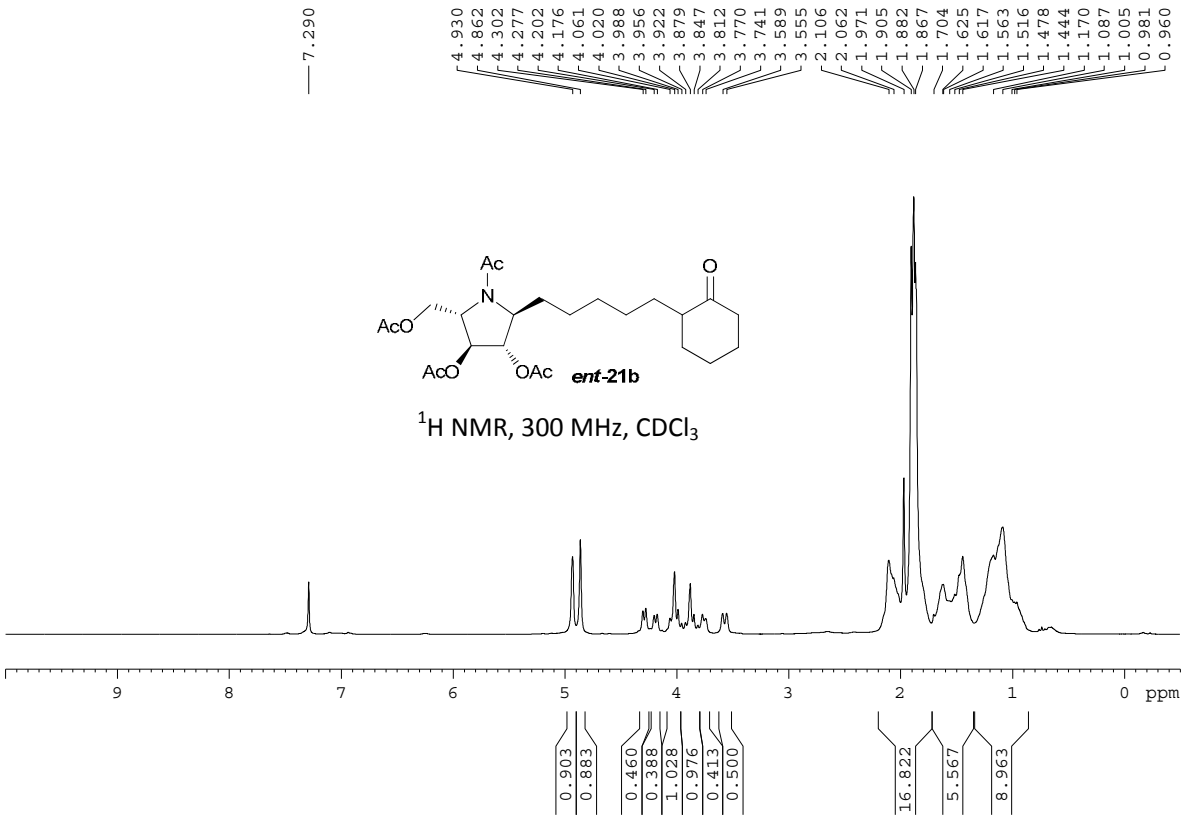




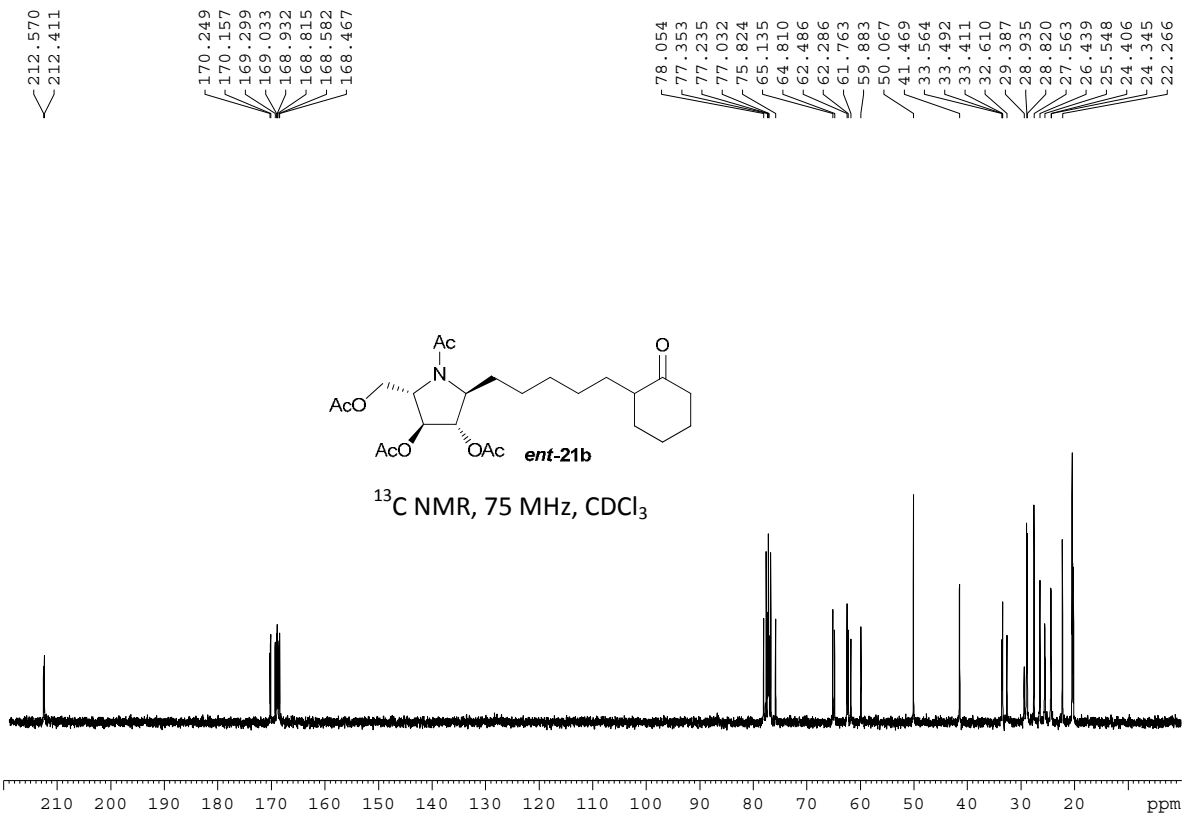
^{1}H NMR, 300 MHz, D_2O



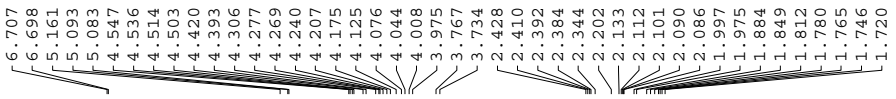
^{13}C NMR, 126 MHz, MeOD

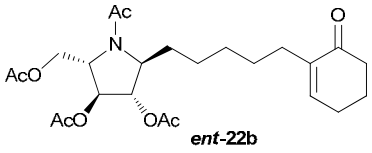


^{1}H NMR, 300 MHz, $CDCl_3$



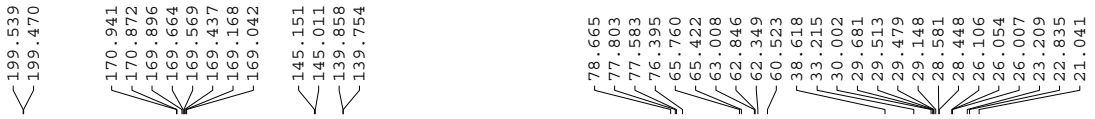
^{13}C NMR, 75 MHz, $CDCl_3$

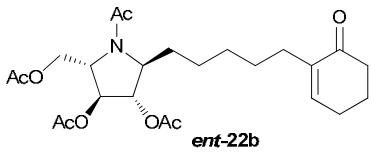




***ent*-22b**

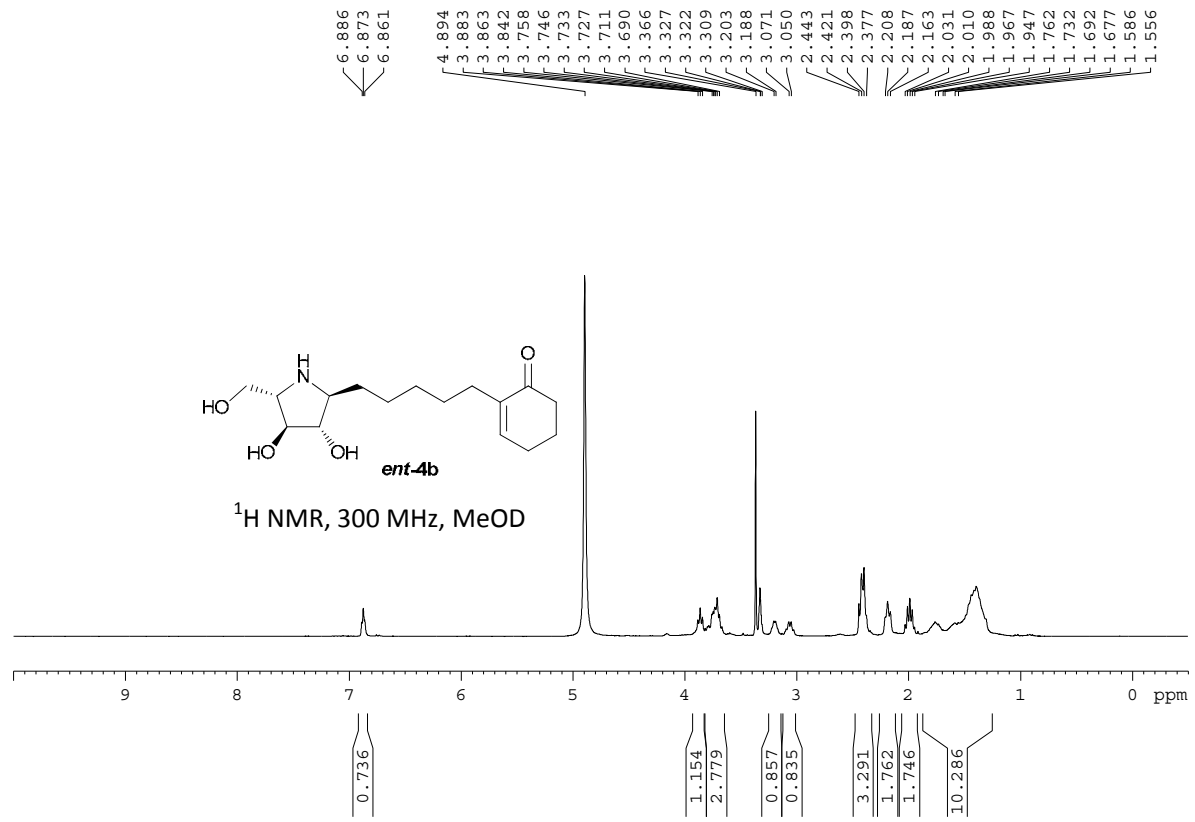
^{1}H NMR, 300 MHz, $CDCl_3$



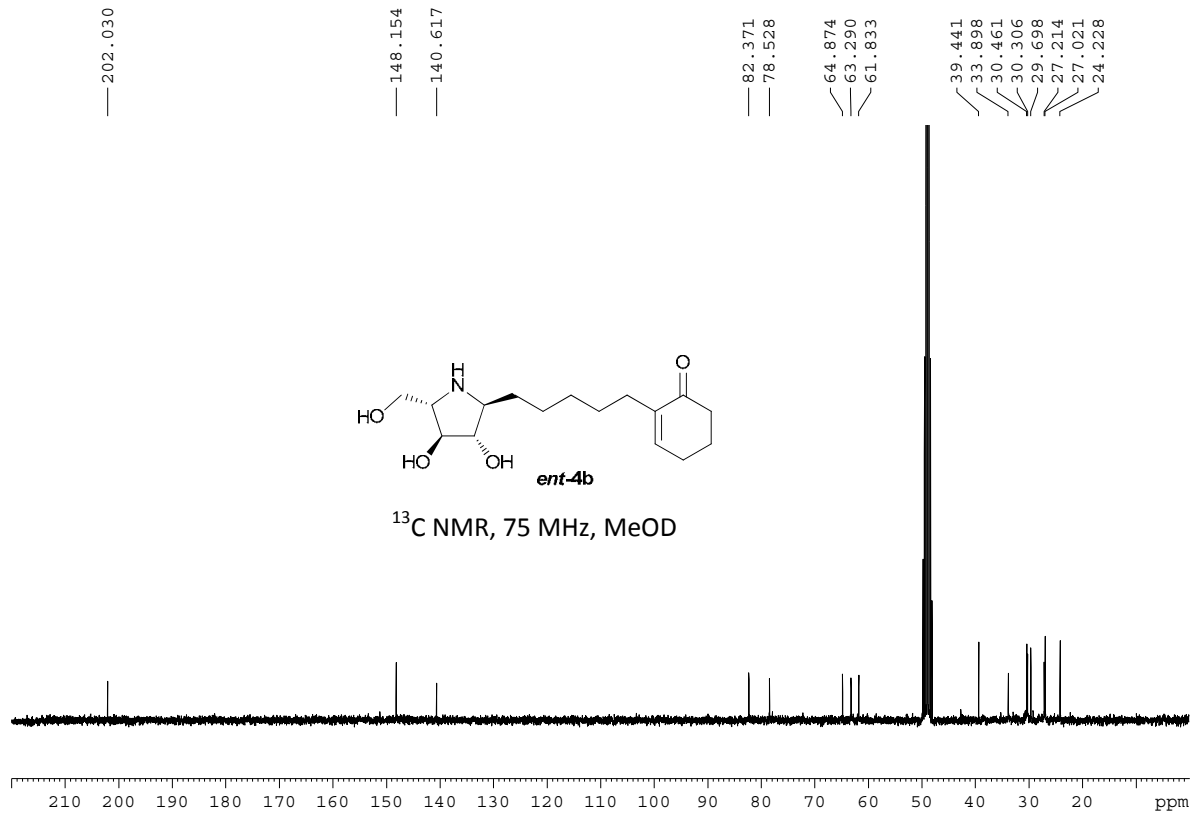


***ent*-22b**

^{13}C NMR, 75 MHz, $CDCl_3$



^{1}H NMR, 300 MHz, MeOD

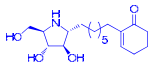
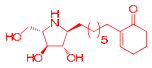
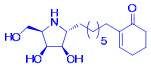
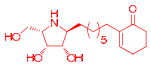
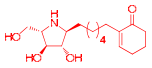
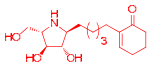


^{13}C NMR, 75 MHz, MeOD

4. Results of the bioassay

The enzymes α-glucosidase (from yeast; from rice), β-glucosidase (from almond; from bovine liver), *α*-galactosidase (from coffee beans), β-galactosidase (from bovine liver), *α*-mannosidase (from jack bean), β-mannosidase (from snail), *α*-L-fucosidase (from bovine kidney), *α,α*-trehalase (from porcine kidney), amyloglucosidase (from *A. niger*), α-L-rhamnosidase (from *Pdecumbens*), *β*-glucuronidases (from bovine liver; from *E. coli*), *p*-nitrophenyl glycosides, and various disaccharides were purchased from Sigma-Aldrich Co. Brush border membranes were prepared from the rat small intestine according to the method of Kessler et al, (Kessler et al 1978)[1] and were assayed at pH 6.8 for rat intestinal maltase using maltose. ER *α*-glucosidase II was prepared from rat liver according to the method of Hino et al.[2] For rat intestinal glucosidases and porcine kitrehalase activities, the reaction mixture (0.2 mL) contained 25 mM substrate and the appropriate amount of enzyme, and the incubations were performed for 10 min at 37 ˚C. The reaction was stopped by heating at 100 ˚C for 3 min. After centrifugation (600 g; 10 min), 0.035 mL of the resulting reaction mixture were added to 2.1 mL of the Glucose CII-test Wako (Wako Pure Chemical Ind., Osaka, Japan). The absorbance at 505 nm was measured to determine the amount of the released D-glucose. Other glycosidase activities were determined using an appropriate *p*-nitrophenyl glycoside as substrate at the optimum pH of each enzyme. The reaction mixture (0.2 mL) contained 2 mM of the substrate and the appropriate amount of enzyme. The reaction was stopped by adding 0.4 mL of 400 mM Na_2CO_3. The released *p*-nitrophenol was measured spectrometrically at 400 nm. The assay results are summarized in the following Table 3 and Table 4.

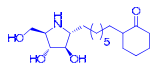
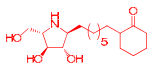
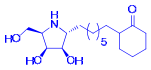
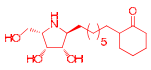
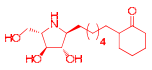
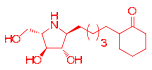
Table 3. Concentrations of Broussonetine W and Its Analogues (**4**, ***ent*-4**, **3-*epi*-4**, ***ent*-3-*epi*-4**, ***ent*-4a**, ***ent*-4b**) Giving 50% Inhibition of Various Glycosidases.

	IC_{50} (μM)					
						
enzyme	**4**	***ent*-4**	**3-*epi*-4**	***ent*-3-*epi*-4**	***ent*-4a**	***ent*-4b**
α-glucosidase						
yeast	22	407	NI[a] (23.7%)[b]	310	758	622
rice	119	0.73	63	24	1.1	1.3
rat intestinal maltase	67	0.047	NI (43.9%)	3.5	0.19	0.2
intestinal isomaltase	NI (29.3%)	1.5	NI (0%)	114	3.0	5.1
rat intestinal sucrase	216	0.20	111	3.4	0.25	0.28
ER α-glucosidase II	NI (43.5%)	7.5	NI (13.2%)	200	15	21
β-glucosidase						
almond	9.8	NI (39.5%)	NI (31.8%)	92	NI (27.2%)	NI (13.4%)
bovine liver	0.12	180	44	0.99	59	263
α-galactosidase						
coffee bean	NI (19.6%)	NI (4.2%)	NI (46.9%)	NI (15.8%)	NI (1.1%)	NI (19.1%)
β-galactosidase						
bovine liver	0.03	127	11	0.18	11	105
α-mannosidase						
jack bean	NI (15.1%)	NI (0%)	385	NI (5.4%)	NI (0%)	NI (6.5%)
β-mannosidase						
snail	282	NI (0%)	NI (7.7%)	NI (0%)	NI (0%)	NI (1.2%)
α-L-fucosidase						
bovine kidney	NI (16.8%)	NI (0%)	NI (12.3%)	20.4	NI (0%)	NI (43.3%)
α, α-trehalase						
porcine kidney	NI (0.8%)	NI (8.3%)	NI (2.2%)	NI (3.3%)	NI (16.4%)	NI (10.0%)
amyloglucosidase						
Aspergilusniger	30	NI (0%)	NI (2.8%)	448	NI (16.2%)	NI (0%)
α-L-rhamnosidase						
Penicillium decumbens	NI (35.1%)	174	NI (47.3%)	166	591	1000
β-glucuronidase						
E.coli	3.3	38	14	83.3	25	60
bovine liver	NI (46.1%)	379	27	NI (40.2%)	NI (46.7%)	186

[a] NI: No inhibition (less than 50% inhibition at 1000 μM).

[b] ( ): inhibition % at 1000 μM.

Table 4. Concentrations of Broussonetine W's Analogues (**20**, ***ent*-20**, **3-*epi*-20**, ***ent*-3-*epi*-20**, ***ent*-20a**, ***ent*-20b**) Giving 50% Inhibition of Various Glycosidases

	IC_{50} (μM)					
						
enzyme	**20**	***ent*-20**	**3-*epi*-20**	***ent*-3-*epi*-20**	***ent*-20a**	***ent*-20b**
α-glucosidase						
yeast	80	NI[a] (32.7%)[b]	NI (44.0%)	NI (16.7%)	NI (46.8%)	NI (39.1%)
rice	70	1.7	7.9	33	1.4	3.7
rat intestinal maltase	32	0.15	1.6	13	0.11	0.14
intestinal isomaltase	663	3.7	40.3	366	2.1	3.8
rat intestinal sucrase	80	0.36	3.1	18	0.14	0.17
ER α-glucosidase II	NI (46.9%)	14	157	458	12	25
β-glucosidase						
almond	43	NI (37.2%)	NI (25.4%)	NI (35.5%)	NI (35.8%)	NI (39.0%)
bovine liver	0.10	14	27	40	237	372
α-galactosidase						
coffee bean	NI (13.8%)	NI (14.6%)	NI (29.4%)	NI (13.5%)	NI (0%)	NI (0.7%)
β-galactosidase						
bovine liver	0.046	8.5	35	14	42	129
α-mannosidase						
jack bean	NI (33.7%)	NI (0%)	245	NI (2.4%)	NI (1.2%)	NI (0%)
β-mannosidase						
snail	224	NI (15.2%)	NI (0%)	NI (0%)	NI (0.9%)	NI (0%)
α-L-fucosidase						
bovine kidney	NI (25.2%)	NI (0.7%)	NI (18.9%)	28	NI (35.7%)	NI (7.6%)
α, α-trehalase						
porcine kidney	NI (0.8%)	NI (3.3%)	NI (0%)	NI (0%)	NI (10.0%)	NI (13.2%)
amyloglucosidase						
Aspergilusniger	41	NI (28.3%)	NI (7.7%)	NI (8.6%)	NI (4.6%)	NI (1.5%)
α-L-rhamnosidase						
Penicillium decumbens	NI (31.4%)	152	895	443	330	410
β-glucuronidase						
E.coli	9.5	79	21	45	428	435
bovine liver	NI (39.7%)	NI (45.2%)	NI (39.0%)	NI (12.3%)	NI (31.9%)	597

[a] NI: No inhibition (less than 50% inhibition at 1000 μM).

[b] ( ): inhibition % at 1000 μM

(1) Kessler, M.; Acuto, O.; Strelli, C.; Murer, H.; Semenza, G. A. *Biochem. Biophys. Acta.* **1978**, *506*, 136.

(2) Hino, Y., Rothman, J. E. *Biochemistry* **1985**, *24*, 800-805.