DATA SUPPLEMENT

Molecular determinants of pro-arrhythmia proclivity of d- and l-sotalol via a multi-scale modeling pipeline

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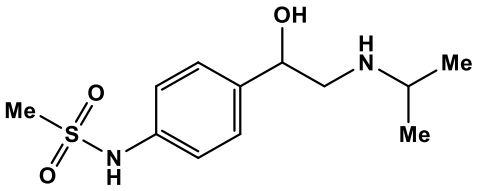
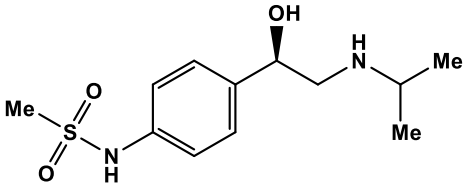
Racemic Sotalol Free Base

CAS: 3930-20-9
mp = 139.5-140.6 °C (obs.)
No reported melting point in literature
Amorphous white solid



Sotalol Free Base Enantiomers



(*R*)-*N*-(4-(1-hydroxy-2-(isopropylamino)ethyl)phenyl)methanesulfonamide

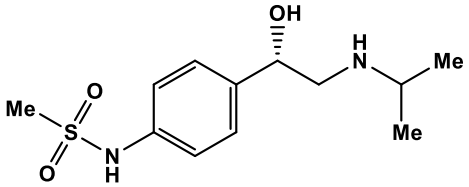
CAS: 30236-32-9

(*R*)-sotalol, (-)-sotalol, *l*-sotalol, D-sotalol

$[\alpha]_D^{25}$ = 35.3 (c = 0.31in CH_3OH)

No XRD

ee 99.6 %



(*S*)-*N*-(4-(1-hydroxy-2-(isopropylamino)ethyl)phenyl)methanesulfonamide

CAS: 30236-31-8

(*S*)-sotalol, (+)-sotalol, *d*-sotalol, L-sotalol

$[\alpha]_D^{25}$ = +17.4 (c = 0.32 in CH_3OH)

XRD confirmation of structure

ee 100 %

After resolution, enantiomeric excess (ee) of both the enantiomers was determined by chiral HPLC.

HRMS confirms the mol. wt. and molecular formula of both the enantiomers.

^{1}H- and ^{13}C-NMR spectra of free base sotalol and (*S*)- and (*R*)-sotalol
See attached spectra at the end of this file.

HRMS spectra of free base sotalol and (*S*)- and (*R*)-sotalol
See attached spectra at the end of this file.

Quantitative HPLC separation of sotalol enantiomers at increasing concentrations

Chiral HPLC resolution of sotalol enantiomers was performed with an Agilent 1100 series HPLC equipped with a Chiralpak IA 4.6 X 250 mm analytical column. Though peak broadening was observed with increasing concentration, the separation between the two enantiomers was good enough, even at 220 mM conc., to use an analytical column for quantitative separation.

Separation of sotalol enantiomers at 10 mM

```
=====================================================================
Injection Date  : 10/9/2017 1:42:56 PM          Seq. Line :   2
Sample Name     : Sotalol 10 mM                  Location : Vial 2
Acq. Operator   : Vikrant                            Inj :   1
Acq. Instrument : LC1100                       Inj Volume : 20 µl
Sequence File   : C:\CHEM32\1\DATA\VIKRANT_100917\SOTALOL1_100917.S
Method          : C:\CHEM32\1\DATA\VIKRANT METHOD\SOTALOL15MIN.M
Last changed    : 10/9/2017 10:06:23 AM by Vikrant
Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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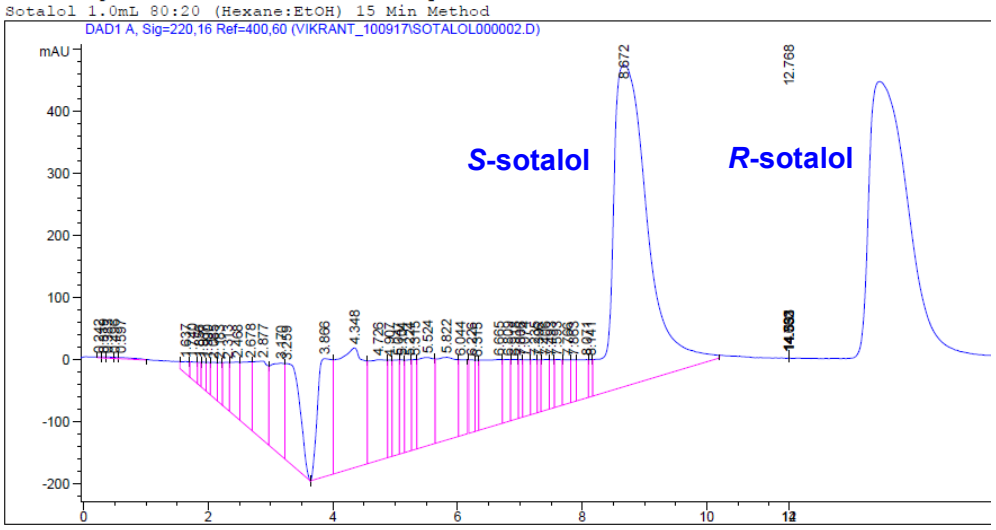

Separation of sotalol enantiomers at 100 mM

```
=====================================================================
Injection Date  : 10/12/2017 9:37:30 AM          Seq. Line :   1
Sample Name     : Sotalol 100 mM                 Location : Vial 1
Acq. Operator   : Vikrant                            Inj :   4
Acq. Instrument : LC1100                       Inj Volume : 20 µl
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Method          : C:\CHEM32\1\DATA\VIKRANT METHOD\SOTALOL15MIN.M
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Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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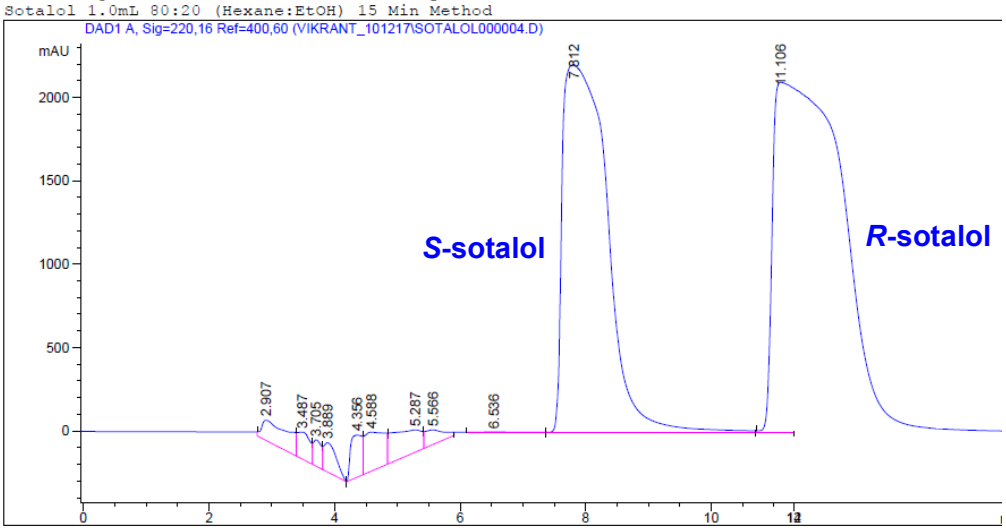

Separation of sotalol enantiomers at 160 mM

```
================================================================================
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Acq. Operator   : Vikrant                             Inj :   5
Acq. Instrument : LC1100                         Inj Volume : 20 µl
Sequence File   : C:\CHEM32\1\DATA\VIKRANT_101217\SOTALOL4_101217.S
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Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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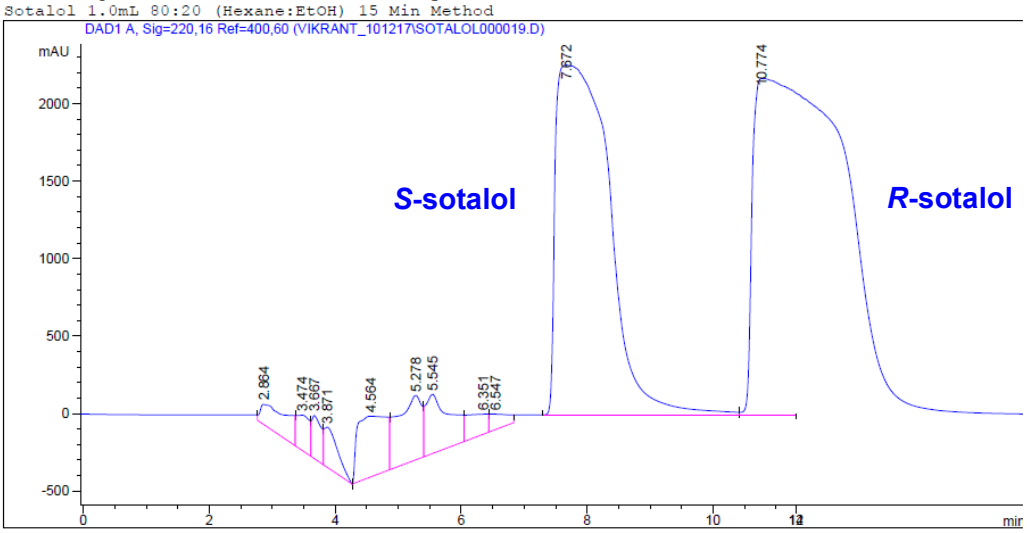

Separation of sotalol enantiomers at 220 mM

```
================================================================================
Injection Date  : 10/17/2017 4:48:04 PM          Seq. Line :   1
Sample Name     : Sotalol 220 mM                   Location : Vial 1
Acq. Operator   : Vikrant                             Inj :   2
Acq. Instrument : LC1100                         Inj Volume : 20 µl
Sequence File   : C:\CHEM32\1\DATA\VIKRANT_101217\SOTALOL9_101217.S
Method          : C:\CHEM32\1\DATA\VIKRANT METHOD\SOTALOL15MIN.M
Last changed    : 10/9/2017 10:06:23 AM by Vikrant
Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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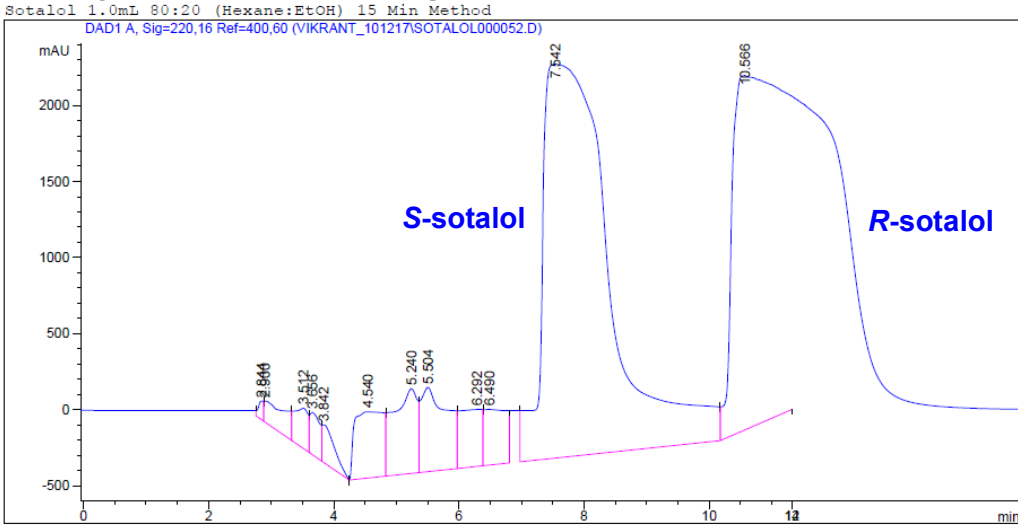

Chiral HPLC chromatograms showing successful separation of *R*- and *S*-sotalol

```
Sotalol Peak-1, Approx 3mg/mL

======================================================================
Injection Date  : 11/2/2017 12:09:52 PM          Seq. Line :   1
Sample Name     : Sotalol Peak 1                  Location : Vial 1
Acq. Operator   : Vikrant                            Inj :   1
Acq. Instrument : LC1100                       Inj Volume : 20 µl
Sequence File   : C:\CHEM32\1\DATA\VIKRANT 102117\SOTALOL EEDETERMINATION_110217.S
Method          : C:\CHEM32\1\DATA\VIKRANT METHOD\SOTALOL15MIN.M
Last changed    : 11/2/2017 12:01:53 PM by Vikrant
Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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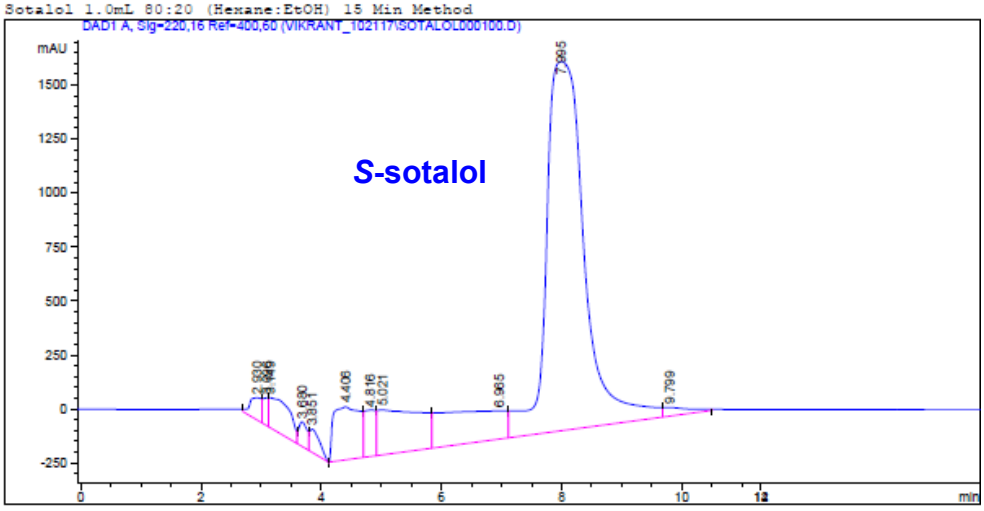


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Sotalol Peak-2, Approx 3mg/mL

======================================================================
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Sample Name     : Sotalol Peak 1                  Location : Vial 2
Acq. Operator   : Vikrant                            Inj :   1
Acq. Instrument : LC1100                       Inj Volume : 20 µl
Sequence File   : C:\CHEM32\1\DATA\VIKRANT 102117\SOTALOL EEDETERMINATION_110217.S
Method          : C:\CHEM32\1\DATA\VIKRANT METHOD\SOTALOL15MIN.M
Last changed    : 11/2/2017 12:01:53 PM by Vikrant
Sotalol 1.0mL 80:20 (Hexane:EtOH) 15 Min Method
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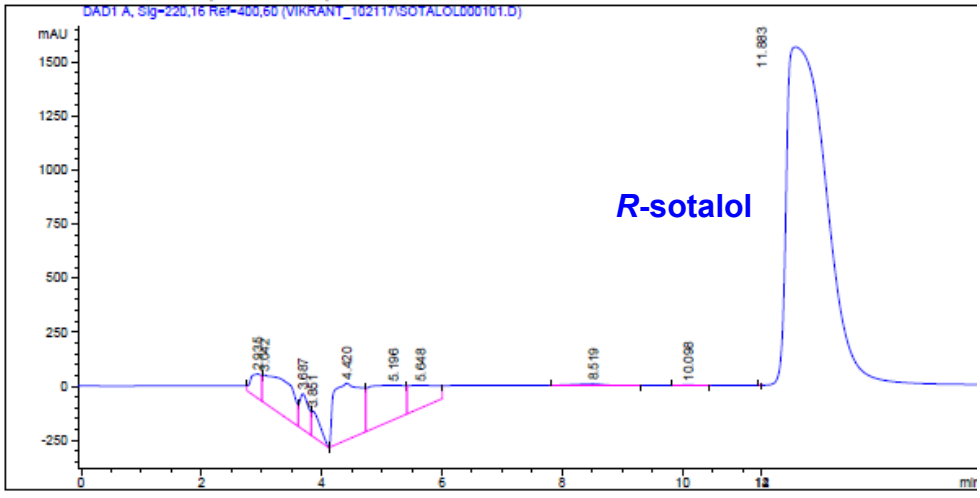
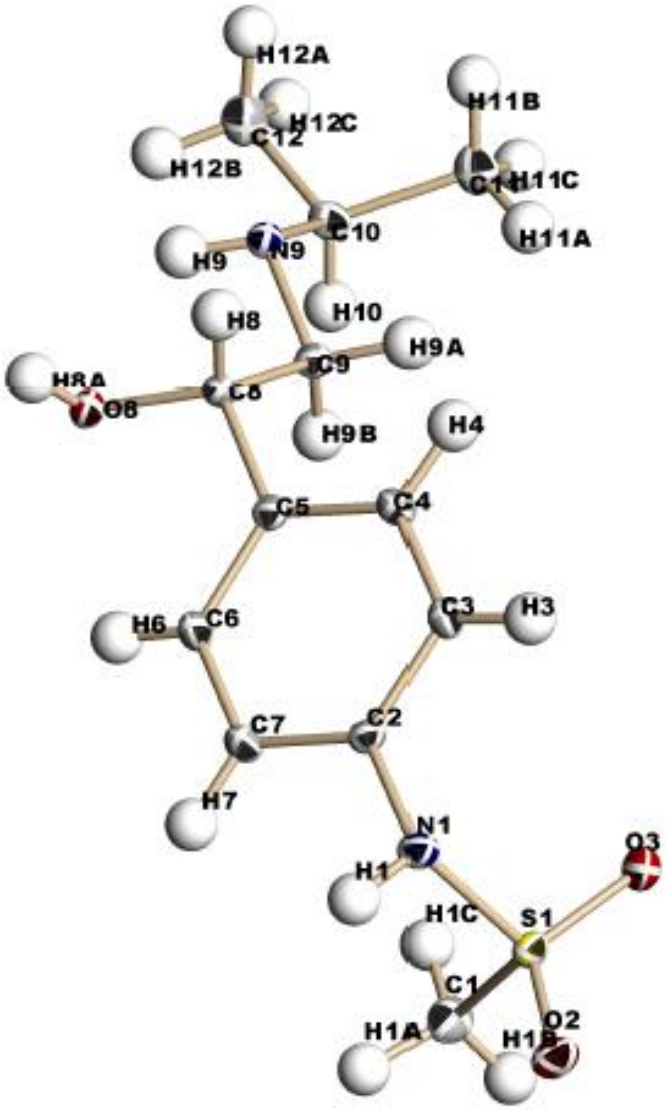


Single crystal X-ray diffraction analysis of (*S*)-sotalol



Atomic coordinates in the PDB format are provided on the next page and were also deposited to the Cambridge Structural Database with Deposition Number 2087412

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REMARK JF2692FMI_s_sotalol.pdb
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SCALE2      0.000000  0.166817  0.000000       0.000000
SCALE3      0.000000  0.000000  0.085197       0.000000
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ANISOU    1  S1          0      190    144    144     20     33    -12       S
ATOM      2  N1          0      -0.394   3.310   4.865 1.000  1.32           N
ANISOU    2  N1          0      143    175    152    -21     23    -10       N
ATOM      3  H1          0      -0.992   2.661   4.916 1.000  1.28           H
ATOM      4  C1          0       0.852   2.737   7.223 1.000  1.94           C
ANISOU    4  C1          0      278    216    199     59    -18     11       C
ATOM      5  H1A         0       0.328   1.874   7.252 1.000  2.81           H
ATOM      6  H1B         0       0.986   3.065   8.060 1.000  1.71           H
ATOM      7  H1C         0       1.682   2.639   6.734 1.000  1.92           H
ATOM      8  C2          0       0.737   3.027   4.014 1.000  1.20           C
ANISOU    8  C2          0      134    164    132    -17     19    -15       C
ATOM      9  O2          0      -1.388   4.042   6.985 1.000  1.97           O
ANISOU    9  O2          0      252    236    216     54     82     -8       O
ATOM     10  C3          0       1.312   4.055   3.279 1.000  1.37           C
ANISOU   10  C3          0      177    132    180    -10     -2      2       C
ATOM     11  H3          0       0.911   4.931   3.372 1.000  1.18           H
ATOM     12  O3          0       0.693   5.119   6.193 1.000  1.93           O
ANISOU   12  O3          0      310    181    200    -48     31    -39       O
ATOM     13  C4          0       2.388   3.787   2.442 1.000  1.33           C
ANISOU   13  C4          0      171    137    169    -25     19     10       C
ATOM     14  H4          0       2.772   4.530   1.943 1.000  2.05           H
ATOM     15  C5          0       2.897   2.490   2.331 1.000  1.16           C
ANISOU   15  C5          0      142    147    127    -19     -1    -13       C
ATOM     16  C6          0       2.310   1.476   3.069 1.000  1.52           C
ANISOU   16  C6          0      211    127    206      0     49      0       C
ATOM     17  H6          0       2.656   0.643   3.041 1.000  2.73           H
ATOM     18  C7          0       1.236   1.733   3.920 1.000  1.52           C
ANISOU   18  C7          0      194    148    199    -32     51     11       C
ATOM     19  H7          0       0.858   1.026   4.433 1.000  1.90           H
ATOM     20  C8          0       4.084   2.220   1.417 1.000  1.21           C
ANISOU   20  C8          0      159    133    140    -17     19     -2       C
ATOM     21  H8          0       3.907   2.515   0.543 1.000  1.19           H
ATOM     22  O8          0       4.403   0.836   1.344 1.000  1.31           O
ANISOU   22  O8          0      171    140    157     -1     14    -26       O
ATOM     23  H8A         0       4.025   0.505   0.681 1.000  2.37           H
ATOM     24  N9          0       6.446   2.715   0.993 1.000  1.19           N
ANISOU   24  N9          0      132    144    151    -12     18     -9       N
ATOM     25  H9          0       6.484   1.879   0.874 1.000  1.09           H
ATOM     26  C9          0       5.324   2.965   1.910 1.000  1.28           C
ANISOU   26  C9          0      148    167    141    -20     21    -11       C
ATOM     27  H9A         0       5.139   3.964   1.944 1.000  1.76           H
ATOM     28  H9B         0       5.537   2.693   2.799 1.000  1.31           H
ATOM     29  C10         0       7.761   3.128   1.543 1.000  1.42           C
ANISOU   29  C10         0      149    188    169    -18     -1     14       C
ATOM     30  H10         0       7.796   2.821   2.439 1.000  1.59           H
ATOM     31  C11         0       7.895   4.642   1.529 1.000  1.75           C
ANISOU   31  C11         0      147    207    271    -20    -17    -15       C
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ATOM     33 H11B         0       7.919   4.936   0.622 1.000  2.27           H
ATOM     34 H11C         0       8.733   4.898   1.964 1.000  2.83           H
ATOM     35  C12         0       8.856   2.473   0.714 1.000  1.95           C
ANISOU   35  C12         0      157    231    309      3     13    -43       C
ATOM     36 H12A         0       8.789   2.855  -0.197 1.000  2.23           H
ATOM     37 H12B         0       8.739   1.557   0.738 1.000  3.11           H
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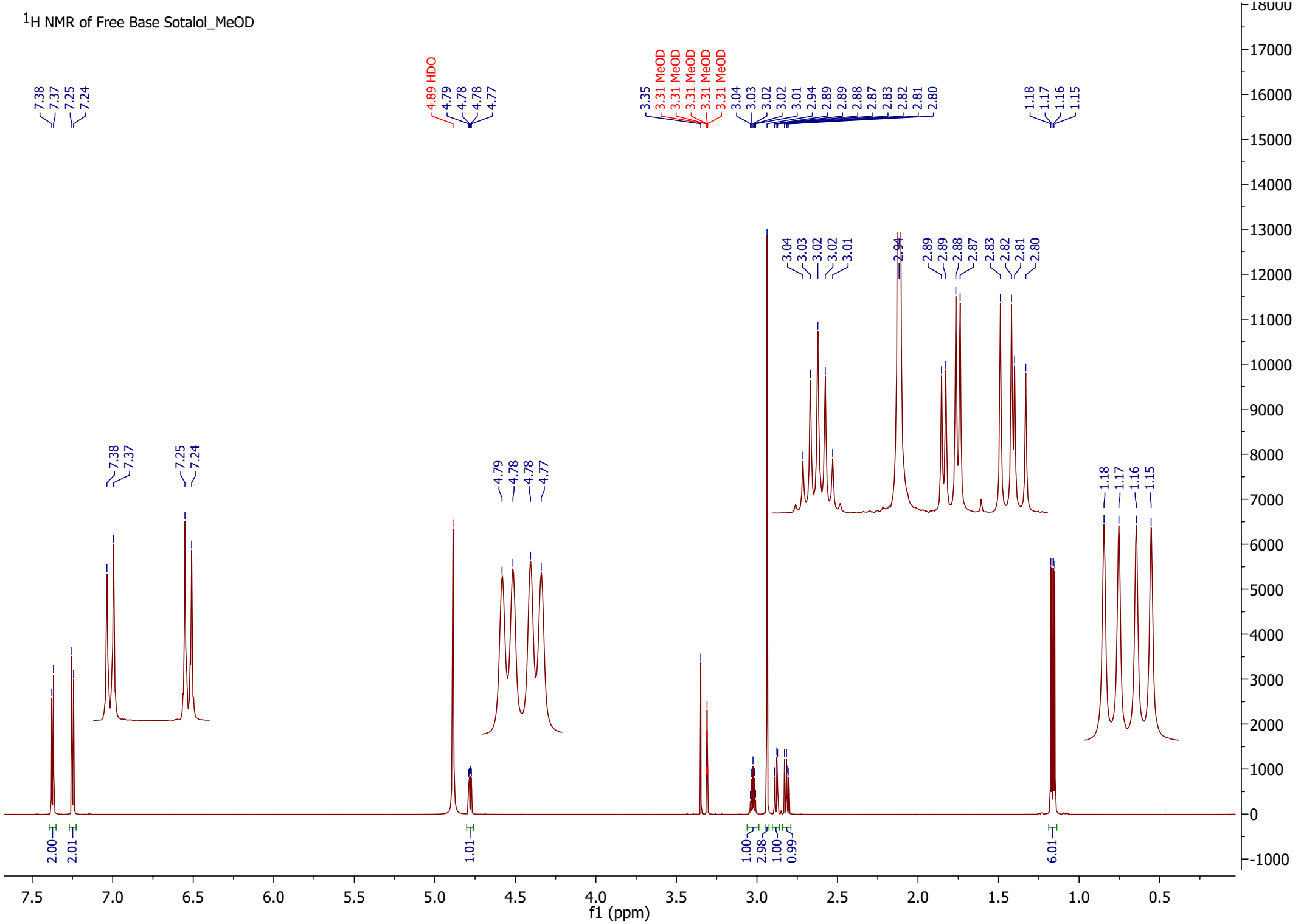
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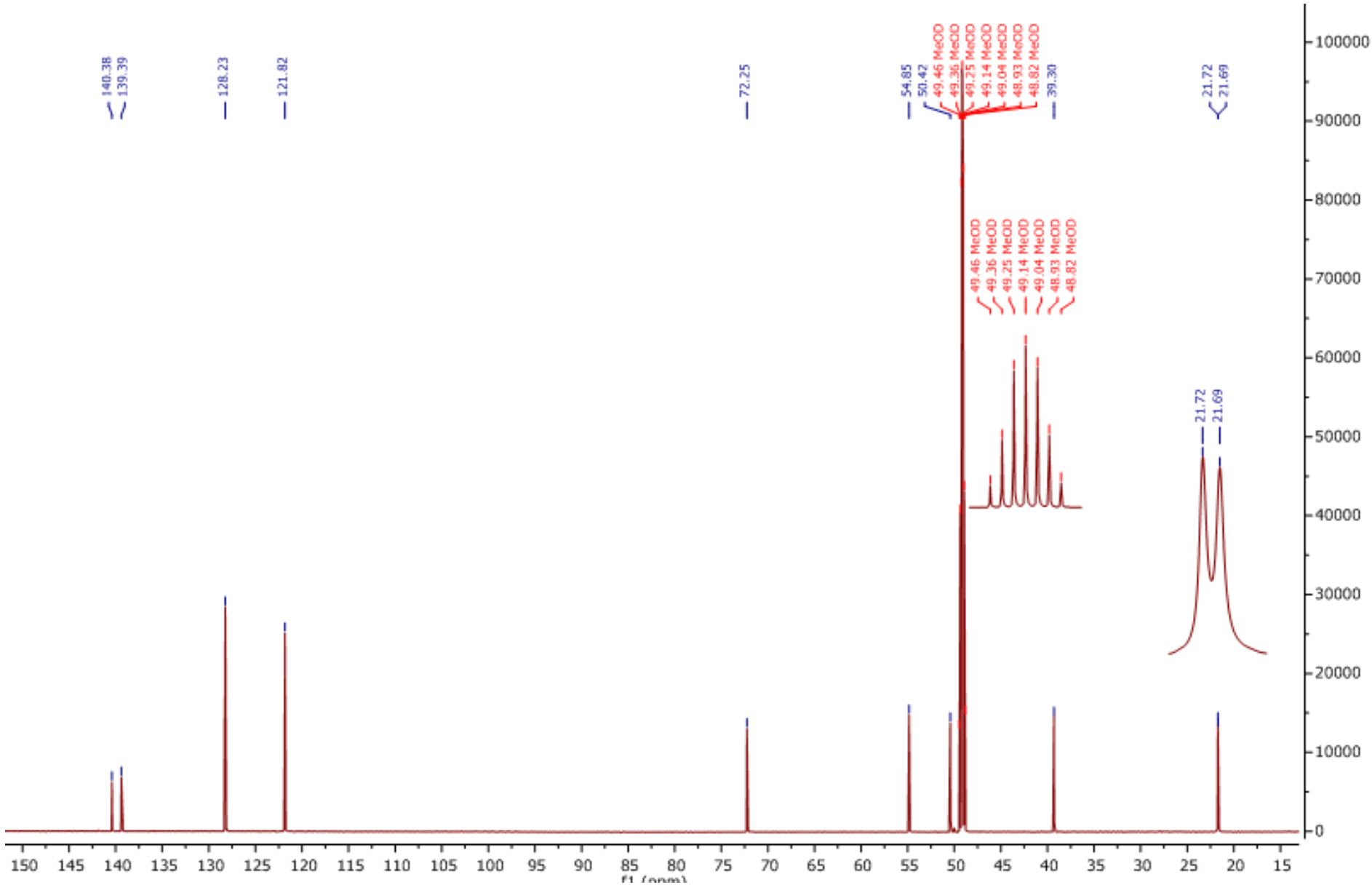
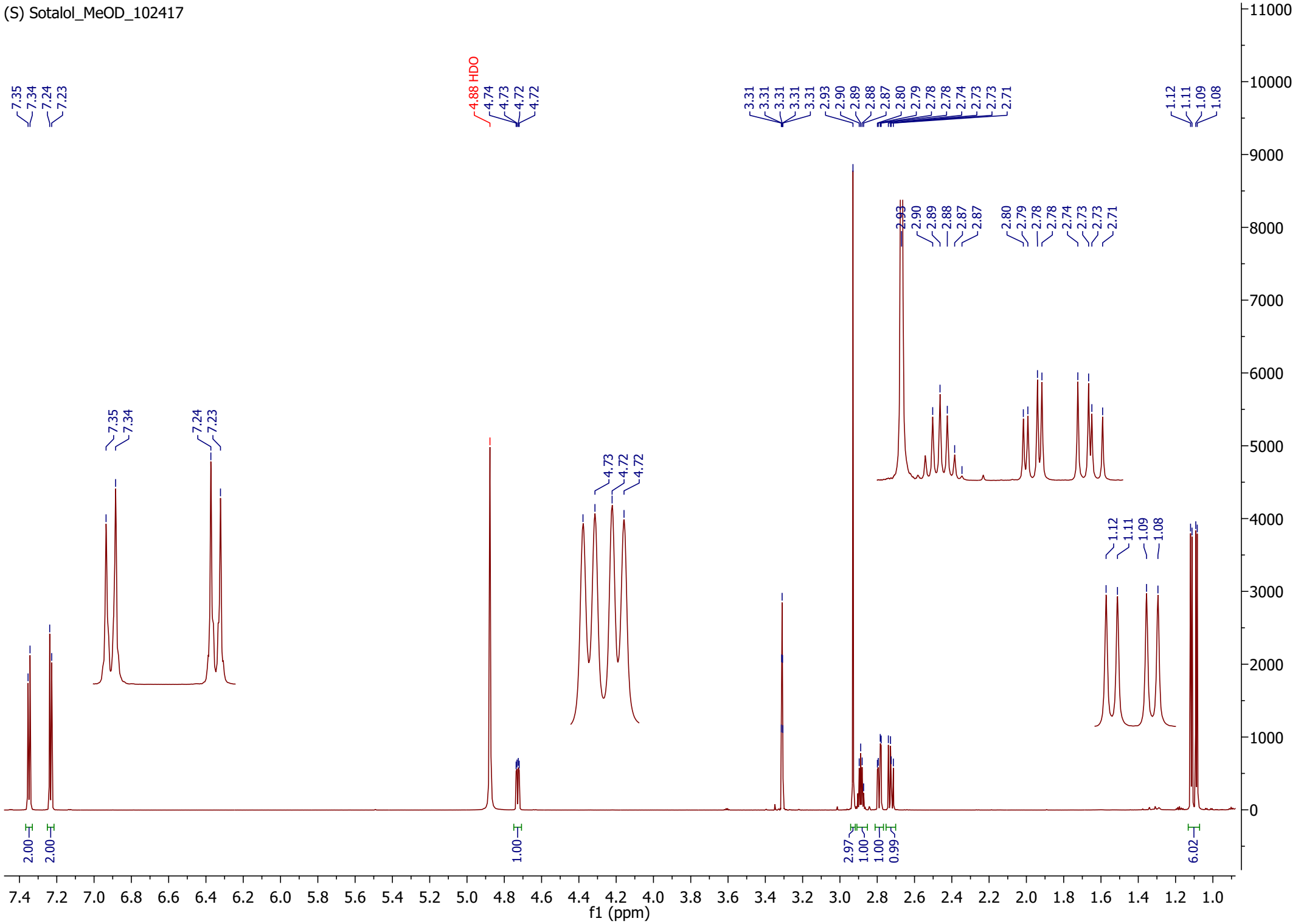
7.38
7.37
7.25
7.24
4.89 HDO
4.79
4.78
4.78
4.77
3.35
3.31 MeOD
3.31 MeOD
3.31 MeOD
3.31 MeOD
3.31 MeOD
3.04
3.03
3.02
3.02
3.01
2.94
2.89
2.89
2.88
2.87
2.83
2.82
2.81
2.80
1.18
1.17
1.16
1.15
2.00
2.01
1.01
1.00
2.98
1.00
0.99
6.01
f1 (ppm)

^{13}C NMR of Free Base Sotalol_MeOD

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7.35
7.34
7.24
7.23
4.88 HDO
4.74
4.73
4.72
4.72
3.31
3.31
3.31
3.31
3.31
2.93
2.90
2.89
2.88
2.87
2.80
2.79
2.78
2.78
2.74
2.73
2.73
2.71
1.12
1.11
1.09
1.08
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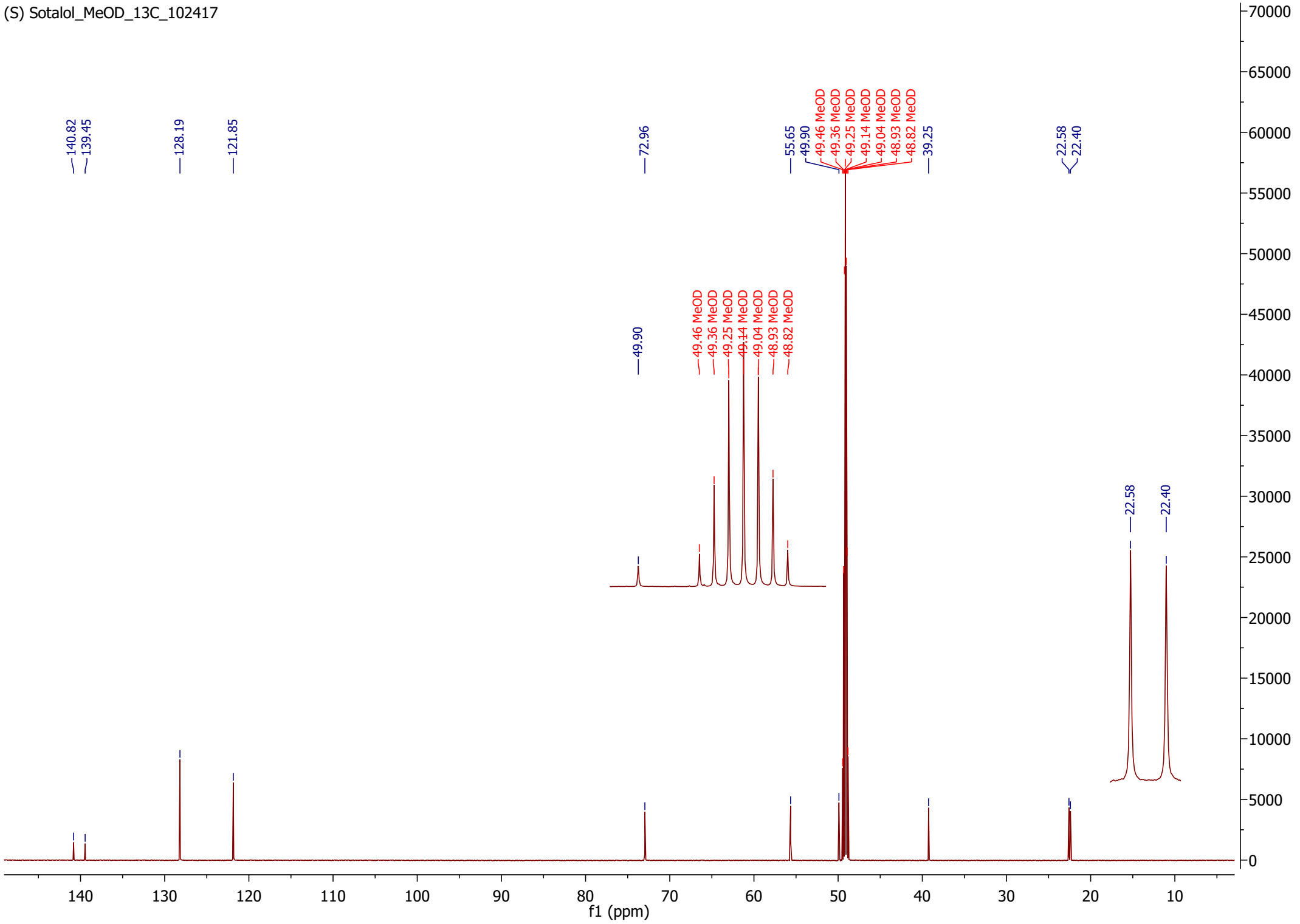
1.00
2.97
1.00
1.00
0.99
6.02
f1 (ppm)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139.45
128.19
121.85
72.96
55.65
49.90
49.46 MeOD
49.36 MeOD
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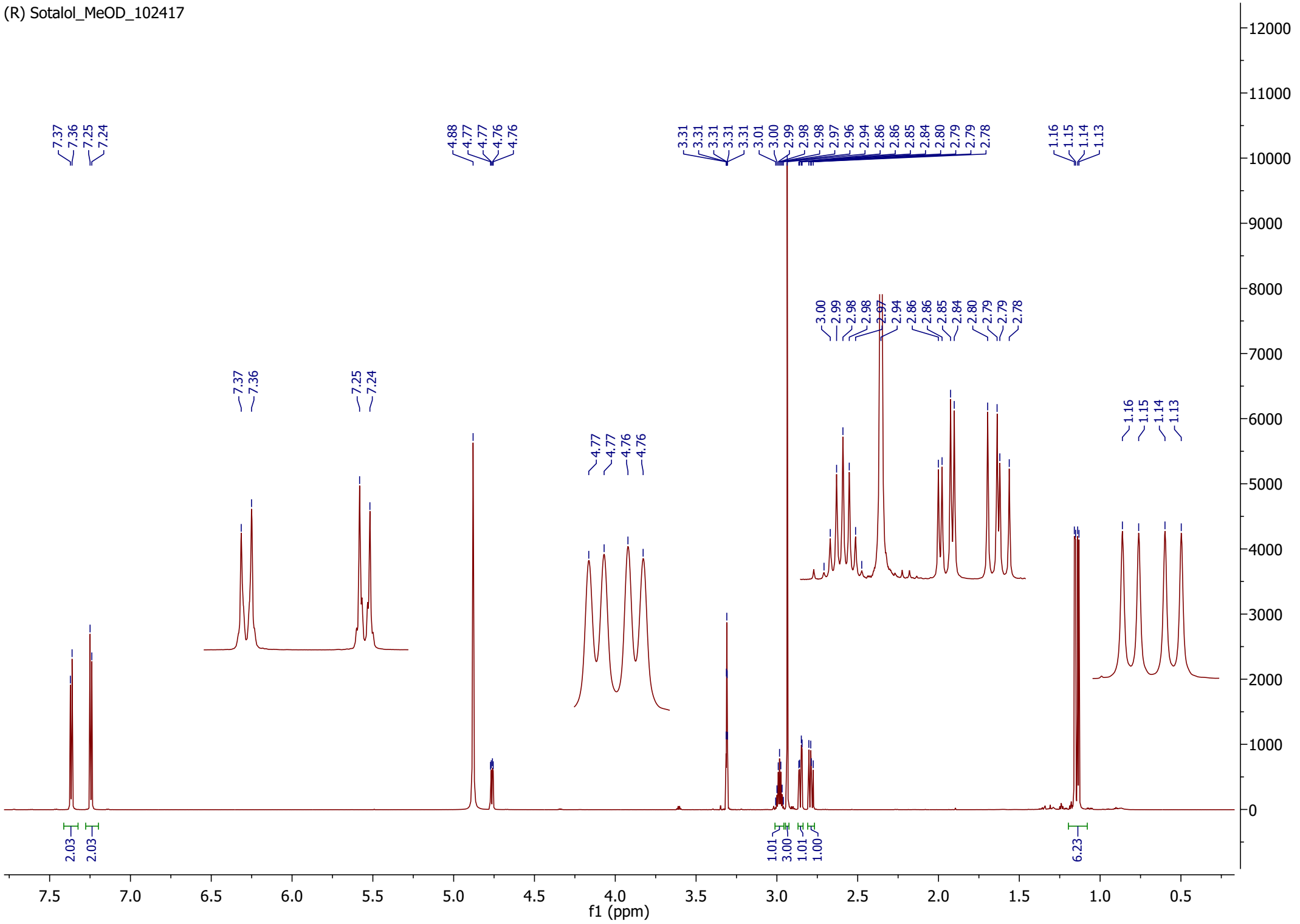
48.93 MeOD
48.82 MeOD
39.25
22.58
22.40
f1 (ppm)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7.37
7.36
7.25
7.24
4.88
4.77
4.77
4.76
4.76
3.31
3.31
3.31
3.31
3.31
3.01
3.00
2.99
2.98
2.98
2.97
2.96
2.94
2.86
2.86
2.85
2.84
2.80
2.79
2.79
2.78
1.16
1.15
1.14
1.13
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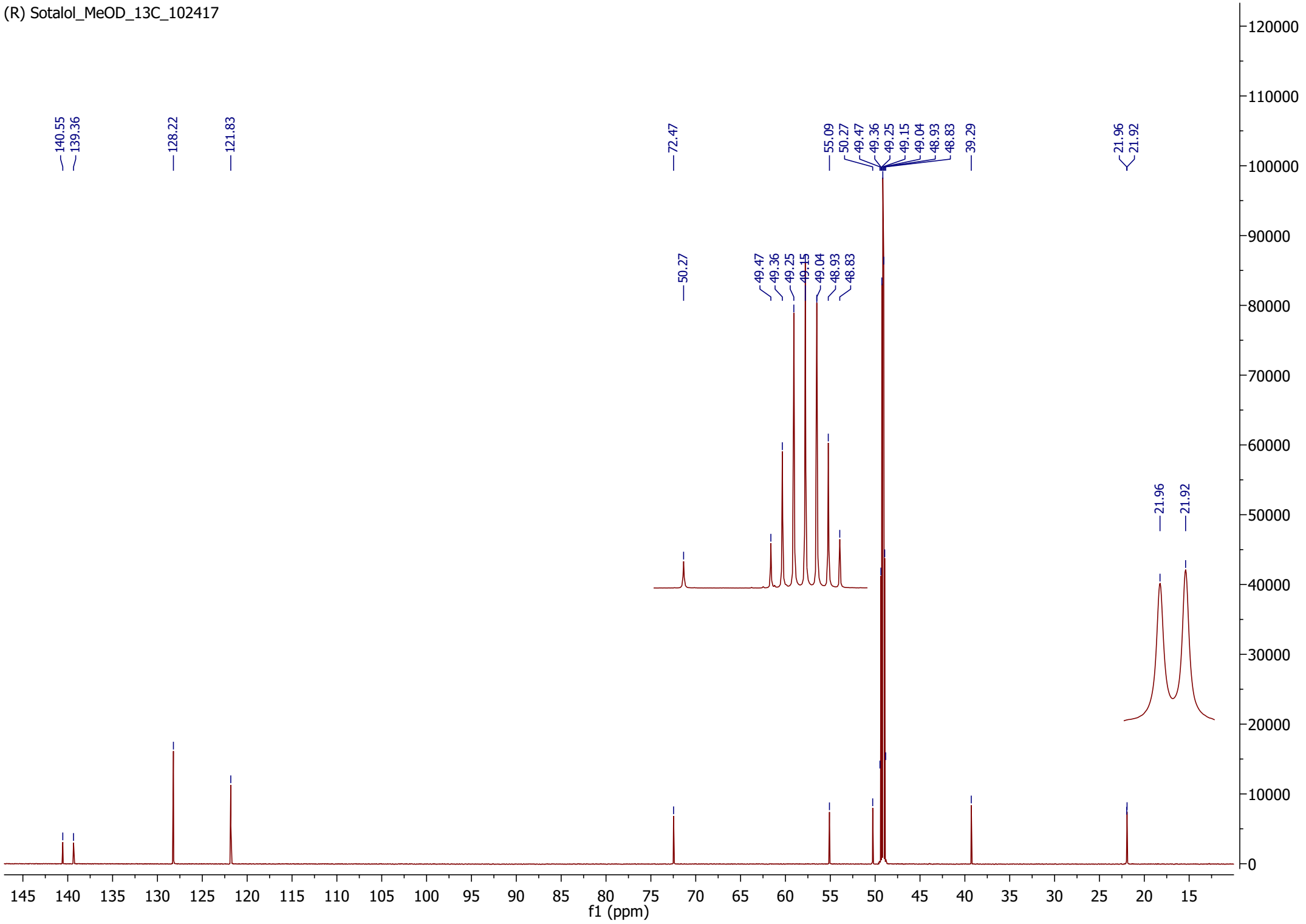
2.03
1.01
3.00
1.01
1.00
6.23
f1 (ppm)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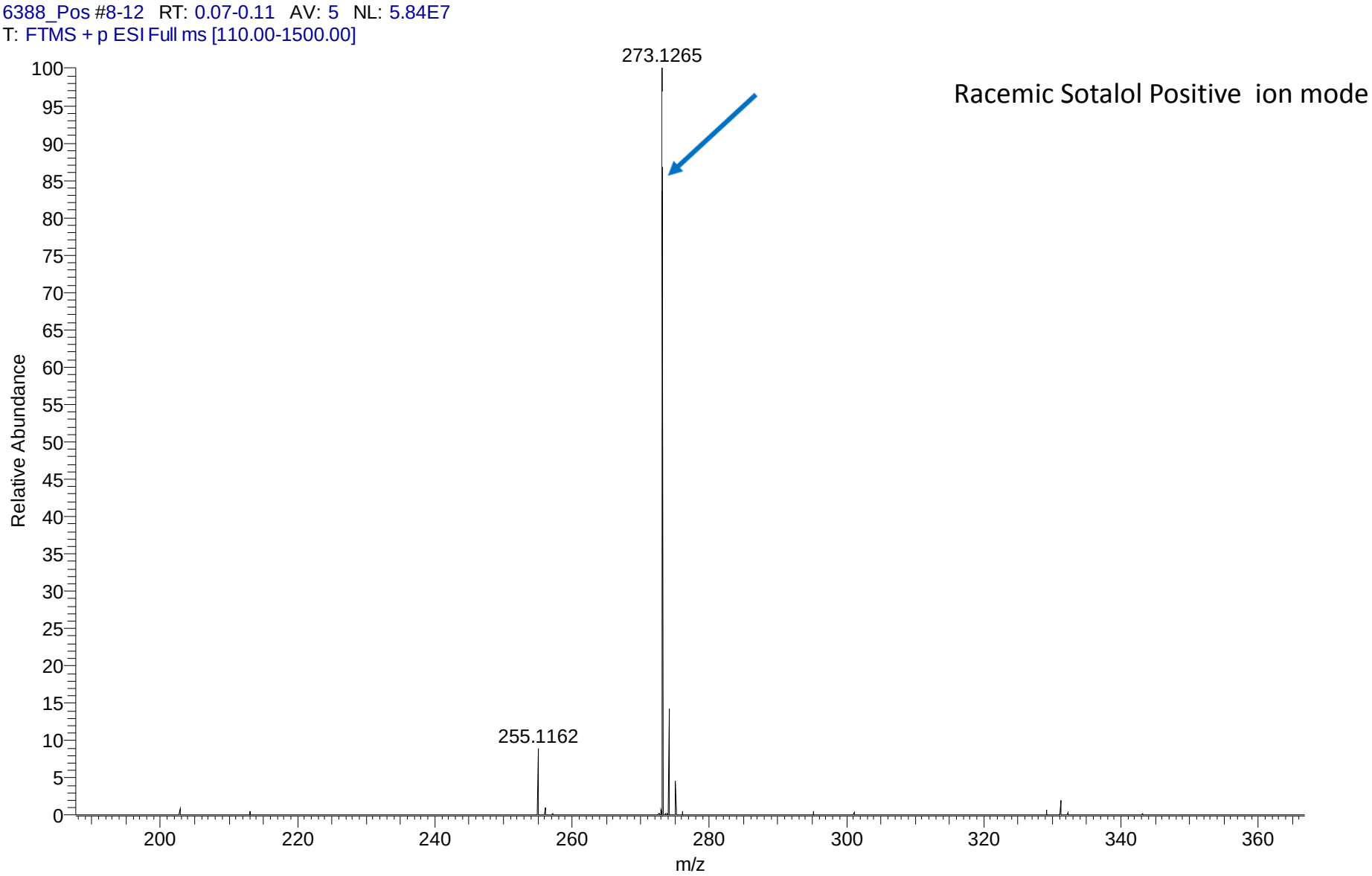
139.36
128.22
121.83
72.47
55.09
50.27
49.47
49.36
49.25
49.15
49.04
48.93
48.83
39.29
21.96
21.92
f1 (ppm)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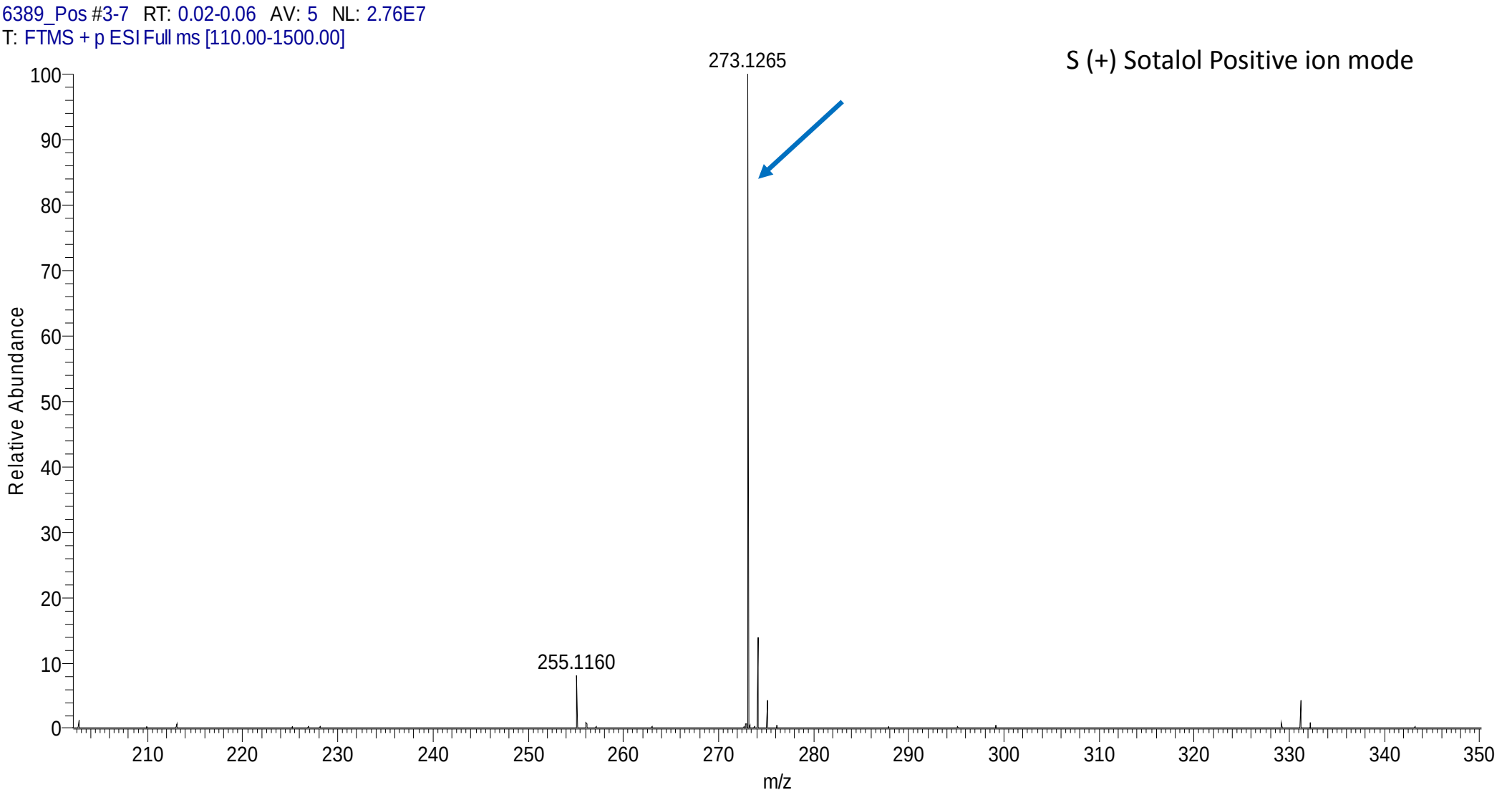
T: FTMS + p ESI Full ms [110.00-1500.00]
273.1265
255.1162
Racemic Sotalol Positive ion mode
Relative Abundance
m/z
100
95
90
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
200
220
240
260
280
300
320
340
360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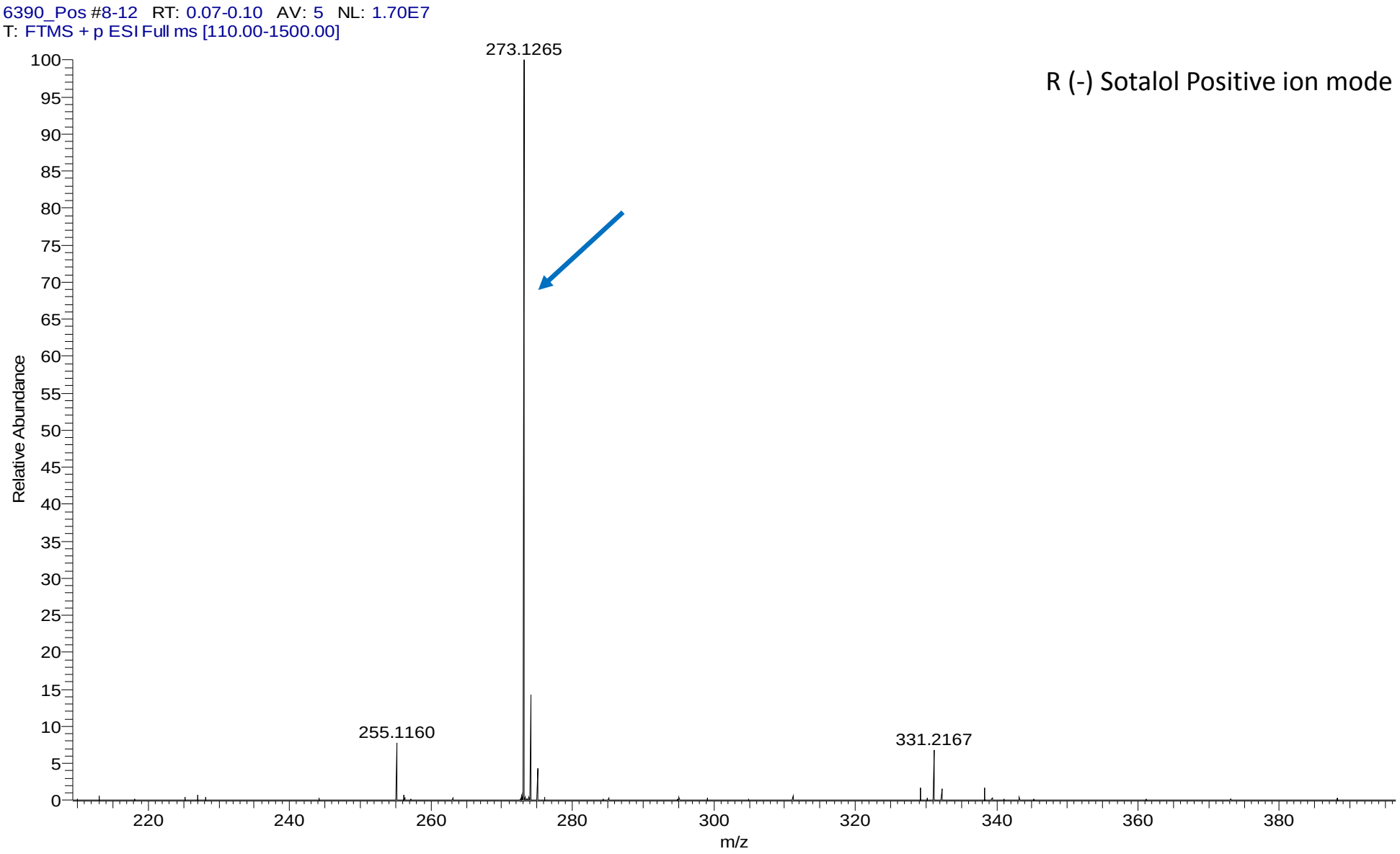
S (+) Sotalol Positive ion mode
273.1265
255.1160
Relative Abundance
m/z
100
90
80
70
60
50
40
30
20
10
0
210
220
230
240
250
260
270
280
290
300
310
320
330
340
350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273.1265
R (-) Sotalol Positive ion mode
255.1160
331.2167
Relative Abundance
m/z
100
95
90
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
220
240
260
280
300
320
340
360
380