Supporting Information

Membrane Active Vancomycin Analogues: A Strategy to Combat Bacterial Resistance

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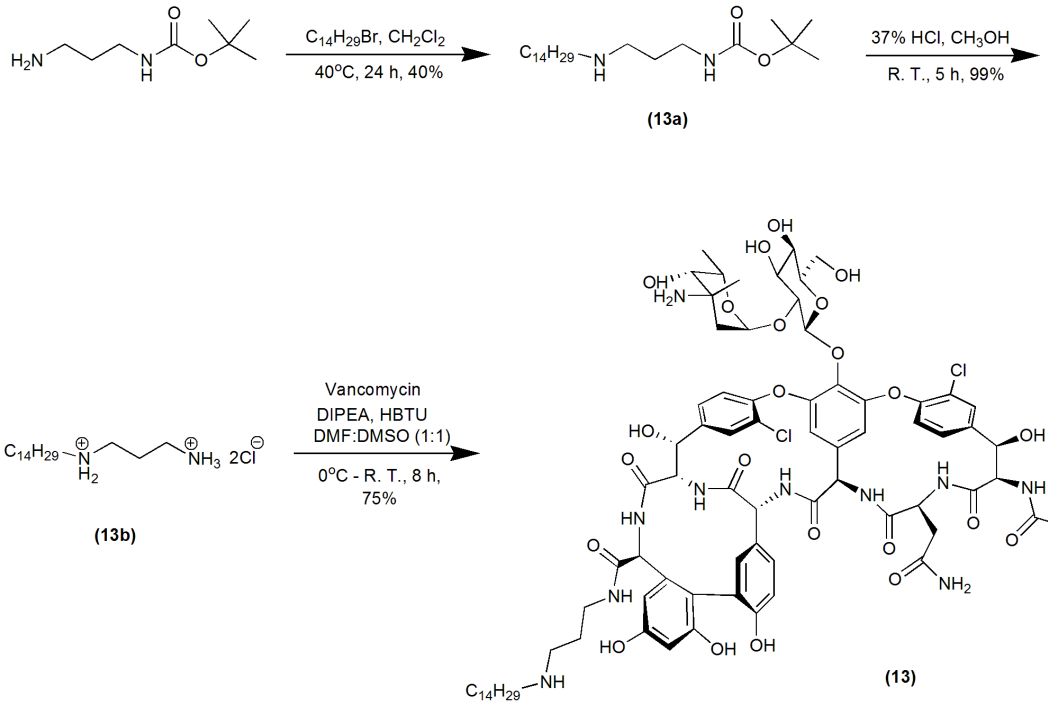
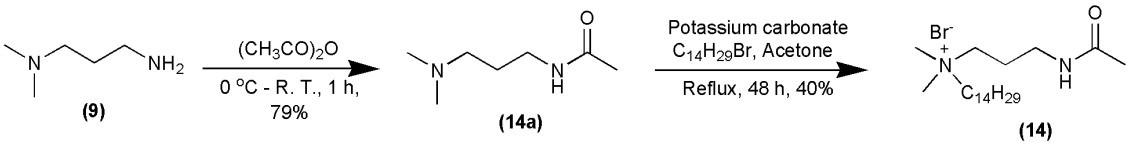
I. Supplementary schemes

Scheme S1: Synthesis of 3-(tetradecylamino)propyl-vancomycin carboxamide (control compound, **13**).



Scheme S2: Synthesis of lipophilic cationic segment (control compound, **14**).



II. Supplementary figures and Tables

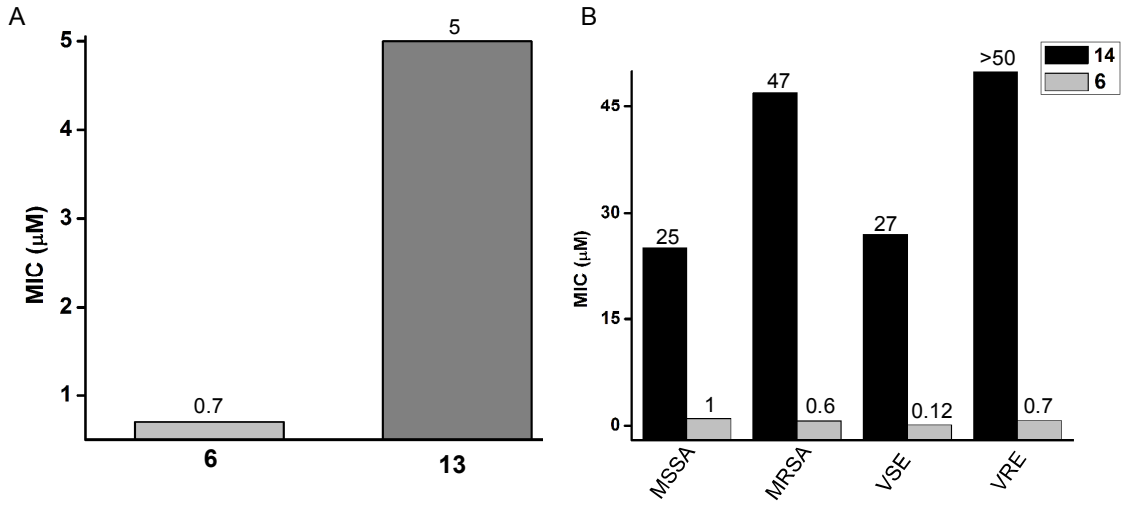


Figure S1
Antibacterial activity comparison of control compounds (**13** and **14**) with compound **6**. (A) Antibacterial activity of compounds **13** and **6** against VRE; vancomycin-resistant *E. faecium* (B) Antibacterial activity of compounds **14** and **6** against MSSA; methicillin-sensitive *S. aureus*, MRSA; methicillin-resistant *S. aureus*, VSE; vancomycin-sensitive *E. faecium* and VRE.

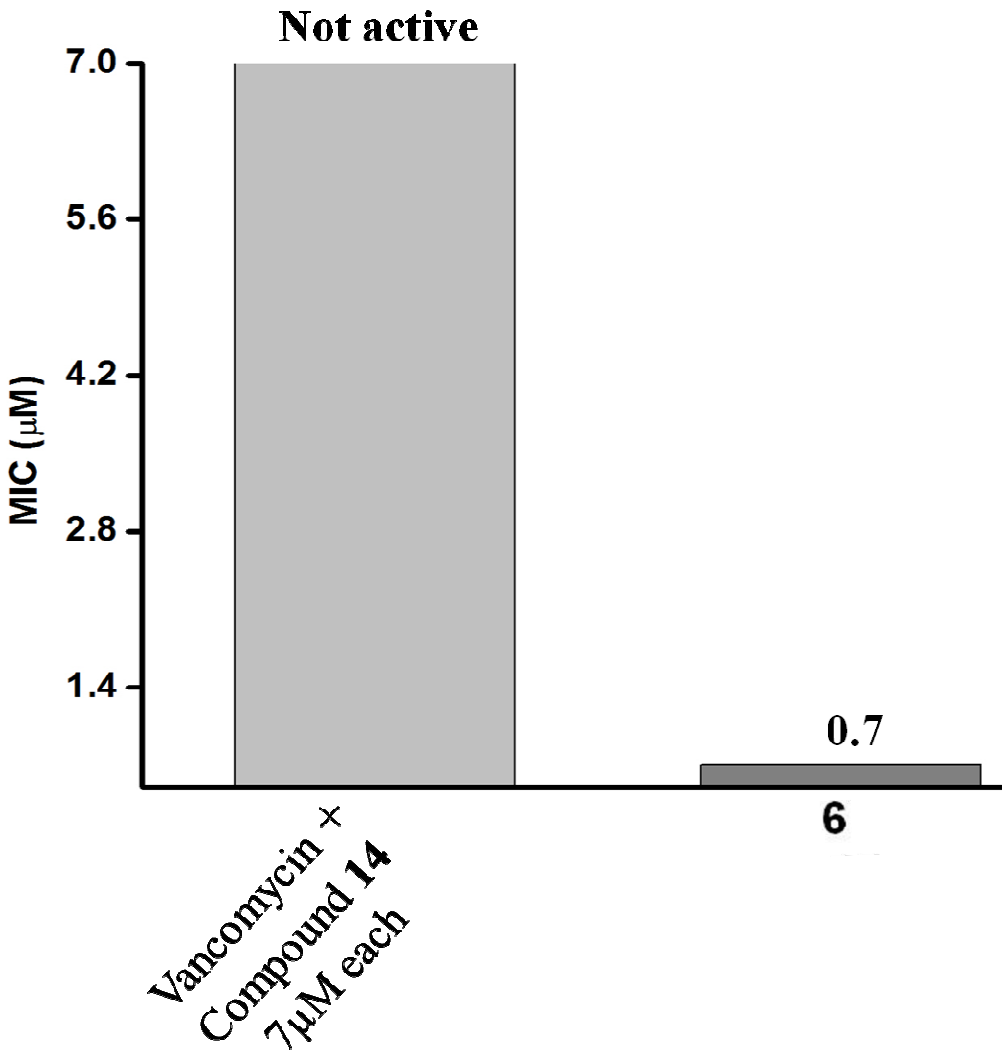


Figure S2
Antibacterial activity of compound **6** and a physical mixture of vancomycin & compound **14** (7 μM each) against vancomycin-resistant Enterococci (VRE).

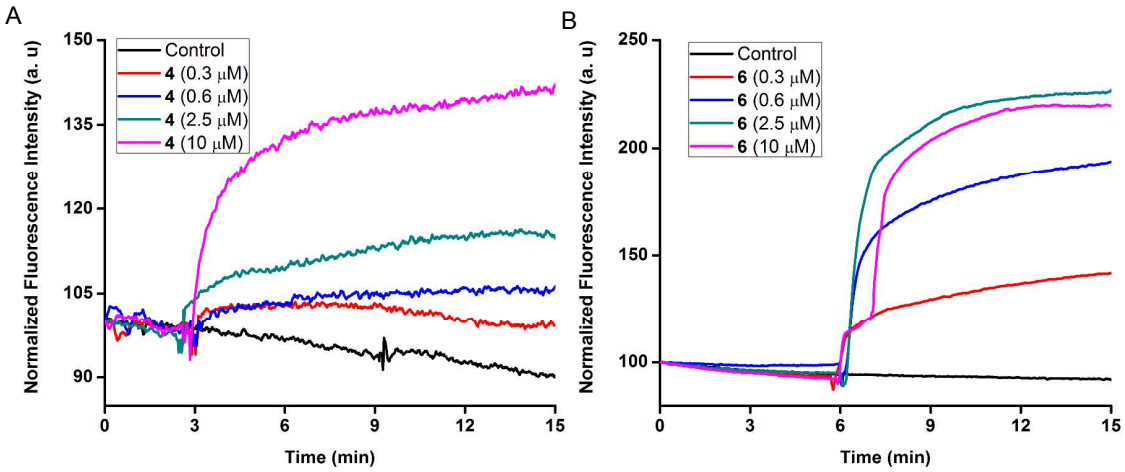


Figure S3.
Concentration dependent membrane depolarization studies of compounds **4** (A) and **6** (B) against MRSA.

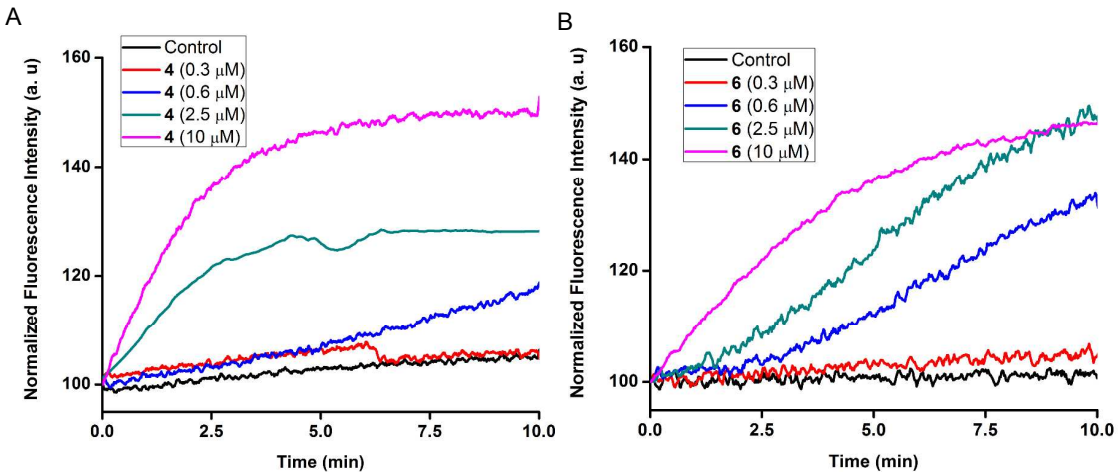


Figure S4.
Concentration dependent membrane permeabilization studies of compounds **4** (A) and **6** (B) against MRSA.

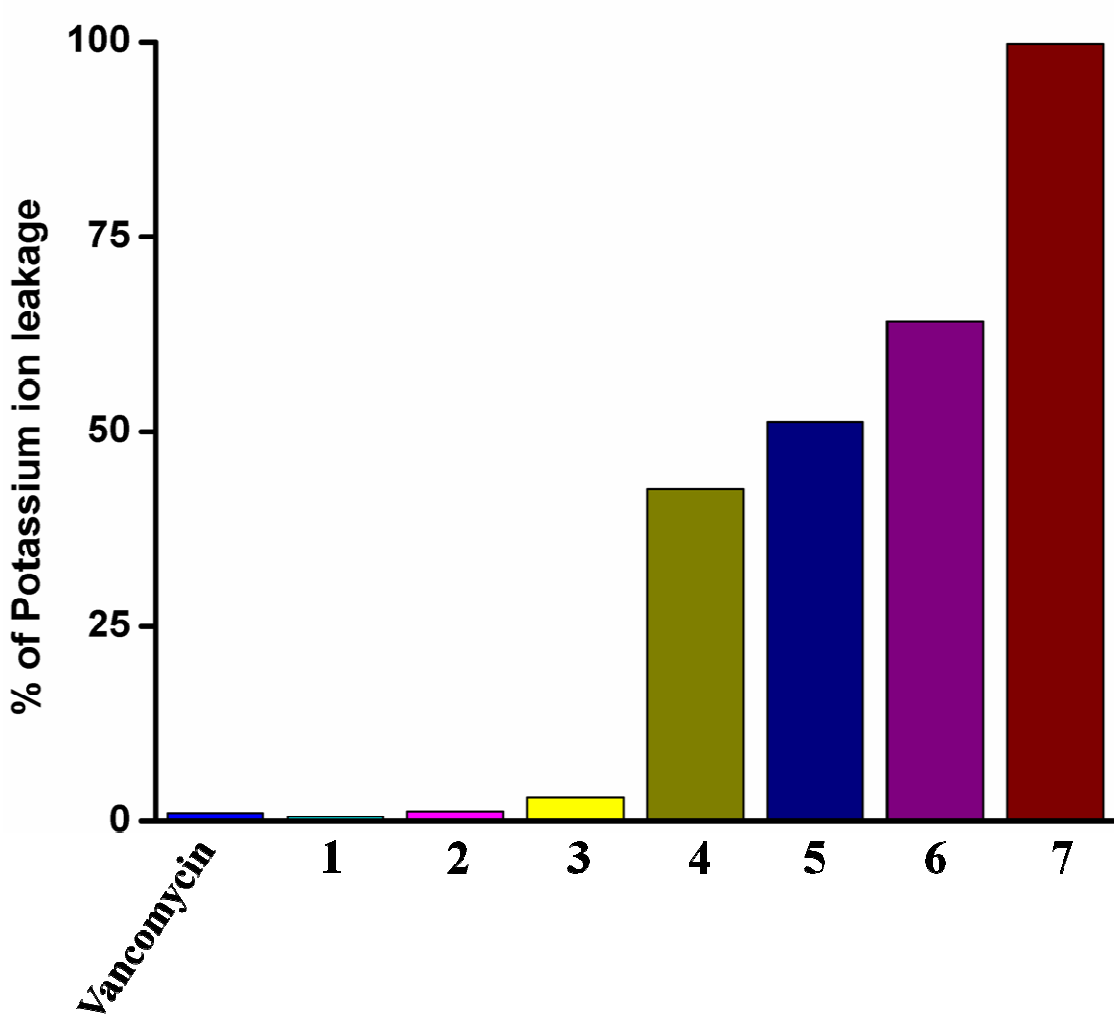


Figure S5.
Percentage of intracellular potassium ion leakage caused by lipophilic cationic vancomycin derivatives (**1**-**7**) at 10 μM against MRSA considering 100% K^+ leakage for valinomycin (10 μM) as a positive control.

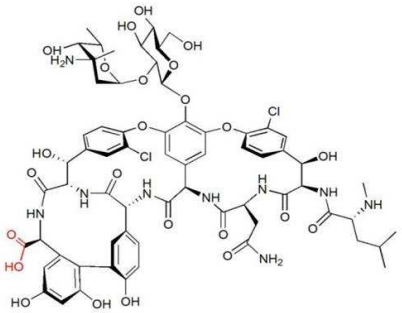
Table S1.

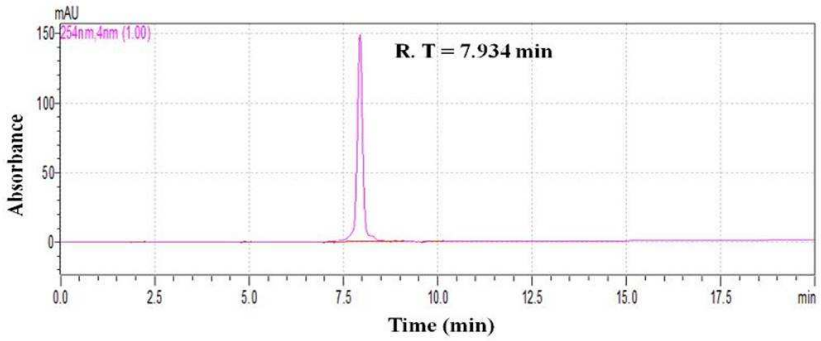
Hemolysis and Cytotoxicity data of vancomycin and compounds **4** and **6**.

	(Toxicity, μM)	
Compound	Hemolytic activity (HC_{50})	Cytotoxicity (EC_{50})
Vancomycin	100	100
4	1000	100
6	100	100

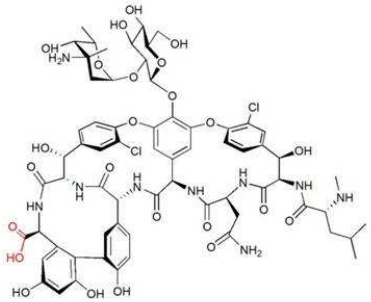
III. Characterization (HPLC chromatograms, NMR and Mass spectra of vancomycin and new vancomycin analogues

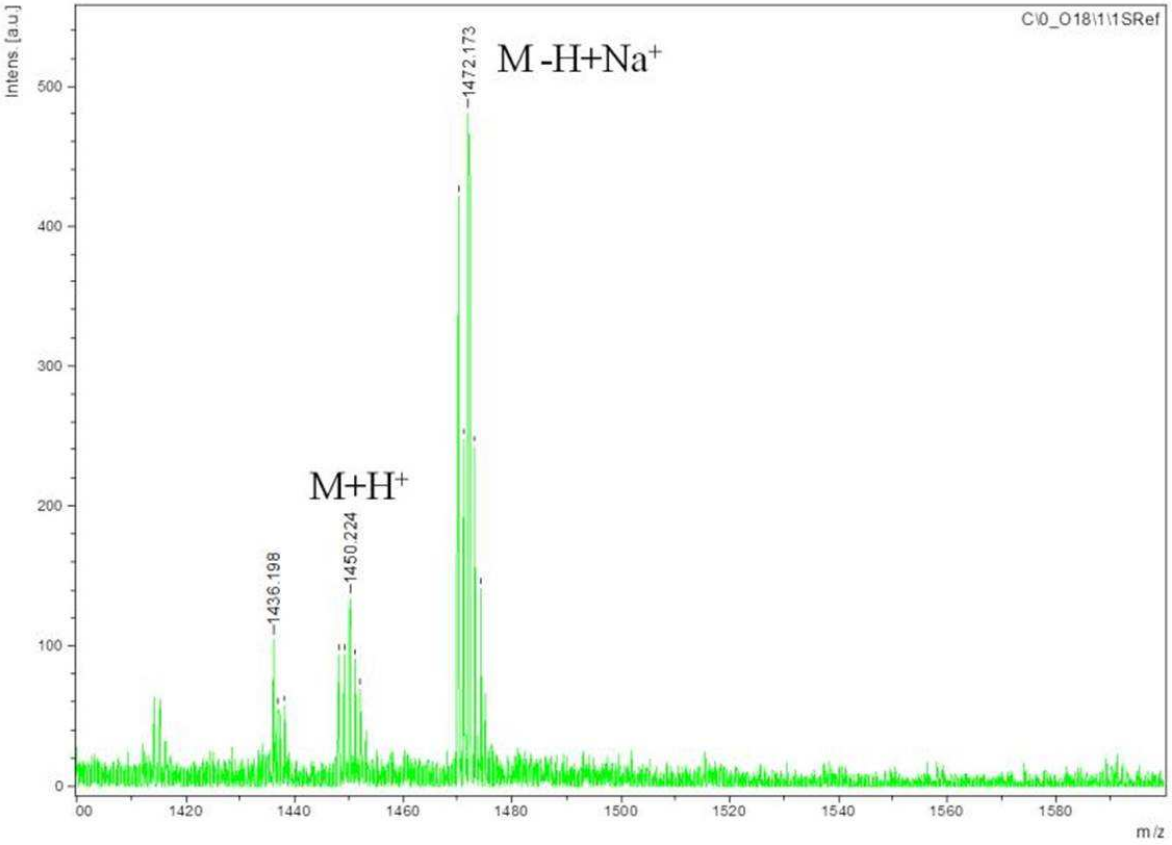
Vancomycin:



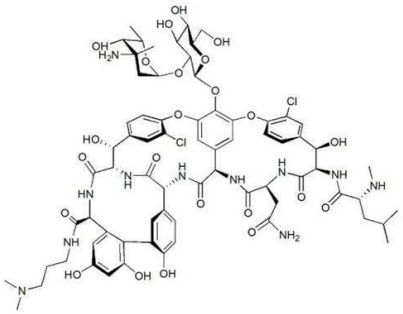


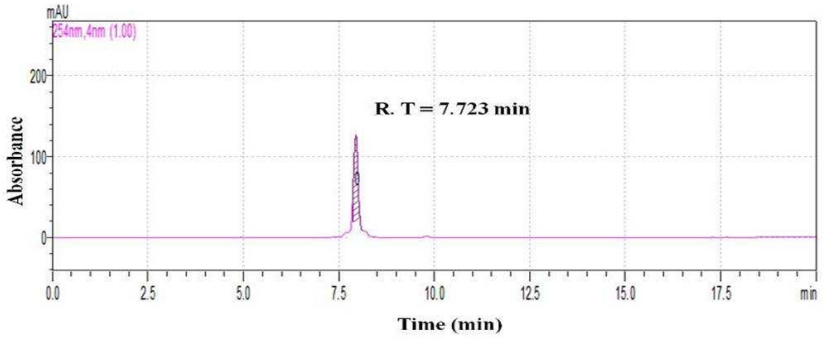
Vancomycin:



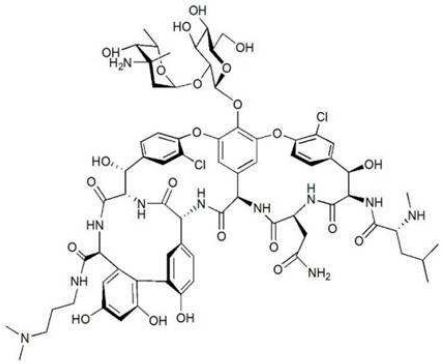

C\0_O18\1\1SRef
Intens. [a.u.]
M -H+Na+
1472.173
M+H+
1450.224
1436.198
500
400
300
200
100
0
00
1420
1440
1460
1480
1500
1520
1540
1560
1580
m/z

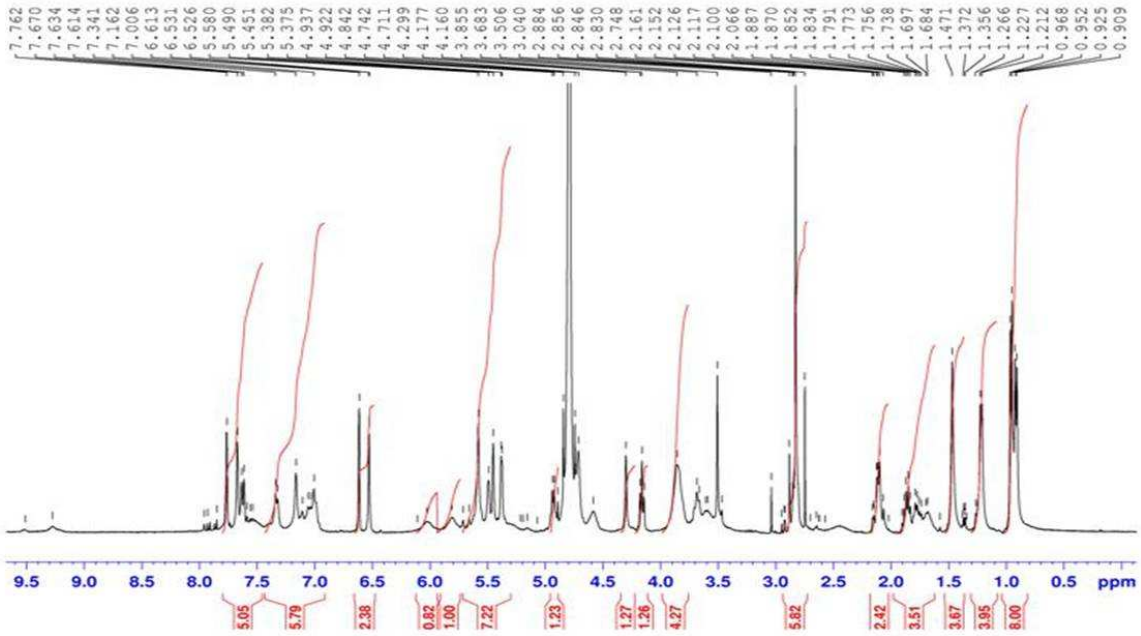
Compound **1**:



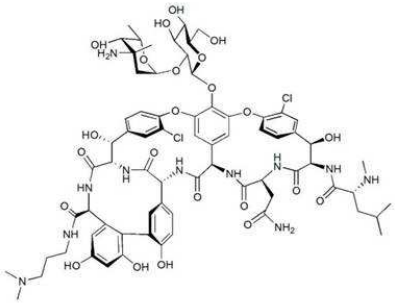

mAU
254nm,4nm (1.00)
200
100
0
Absorbance
R. T = 7.723 min
0.0
2.5
5.0
7.5
10.0
12.5
15.0
17.5
min
Time (min)

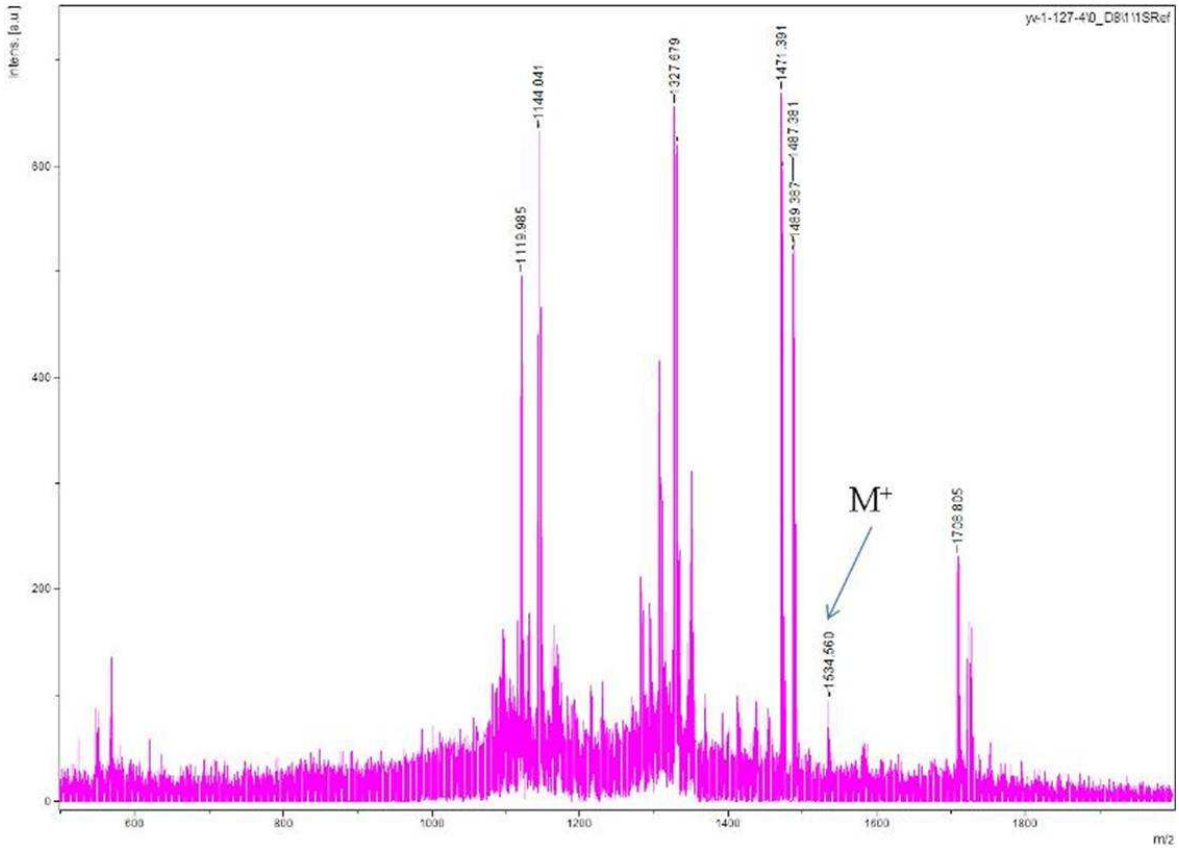
Compound **1**:



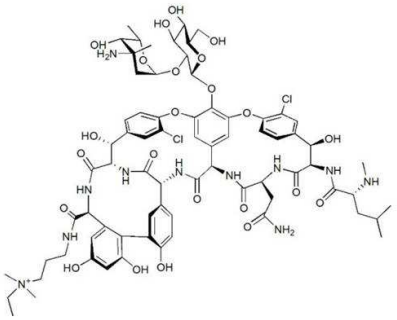

7.762
7.670
7.634
7.614
7.341
7.162
7.006
6.613
6.531
6.526
6.580
5.490
5.451
5.382
5.375
4.937
4.922
4.842
4.742
4.711
4.299
4.177
4.160
3.855
3.683
3.506
3.040
2.884
2.856
2.846
2.830
2.748
2.161
2.152
2.126
2.117
2.100
2.066
1.887
1.870
1.852
1.834
1.791
1.773
1.756
1.738
1.697
1.684
1.471
1.372
1.356
1.266
1.227
1.212
0.968
0.952
0.925
0.909
9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 ppm
5.05
5.79
2.38
0.82
1.00
7.22
1.23
1.27
1.26
4.27
5.82
2.42
3.51
3.67
3.95
8.00

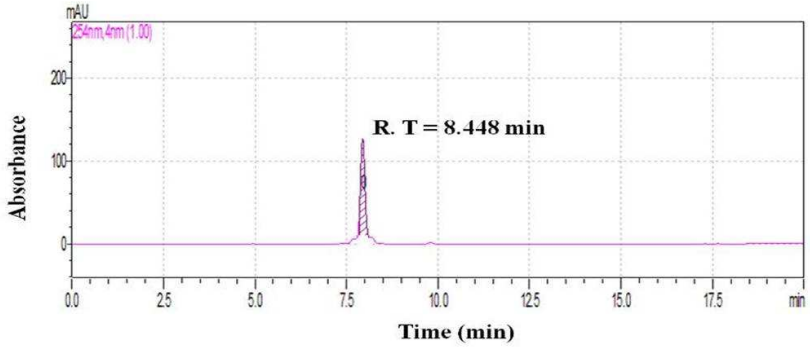
Compound **1**:



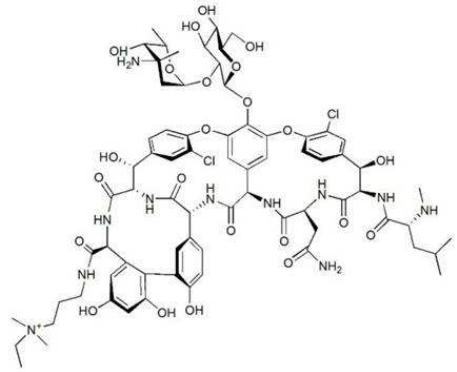

yx-1-127-410_D8\11\SRef
Intens. [a.u.]
1119.985
1144.041
1327.679
1471.391
1487.381
1534.560
1708.805
M+
m/z

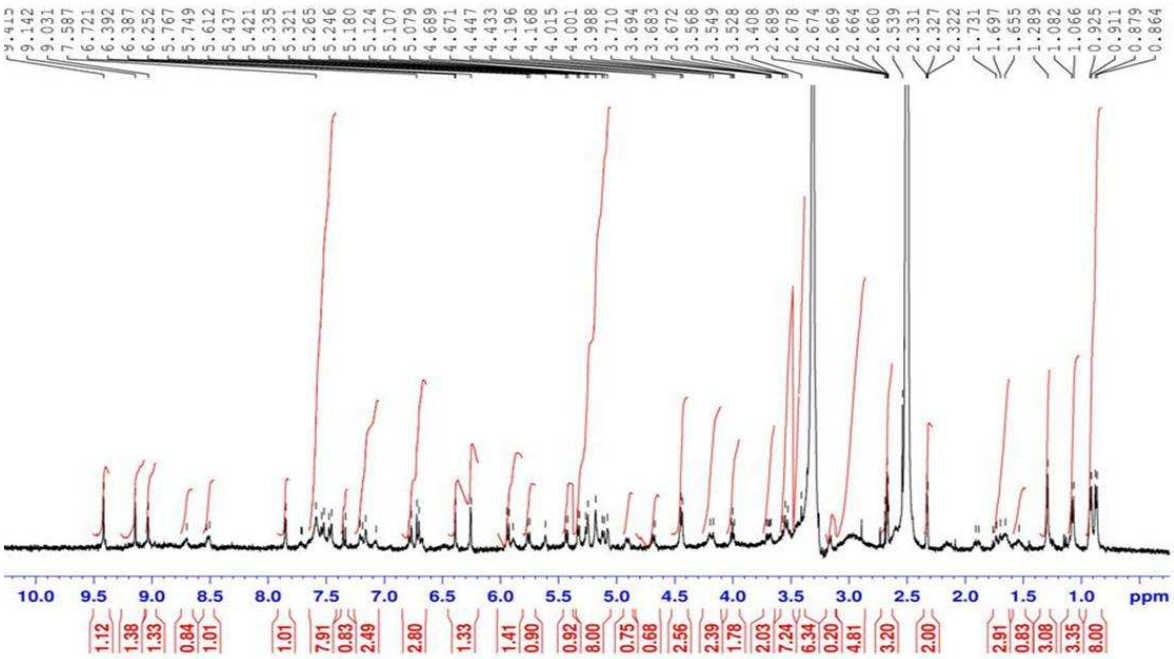
Compound **2**:



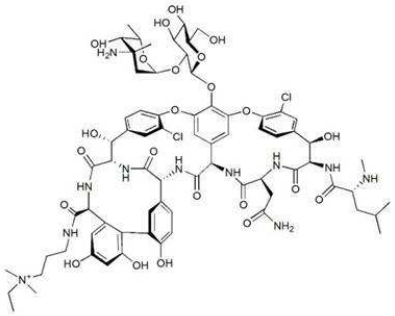

mAU
254nm,4nm (1.00)
200
100
0
Absorbance
R. T = 8.448 min
0.0
2.5
5.0
7.5
10.0
12.5
15.0
17.5
min
Time (min)

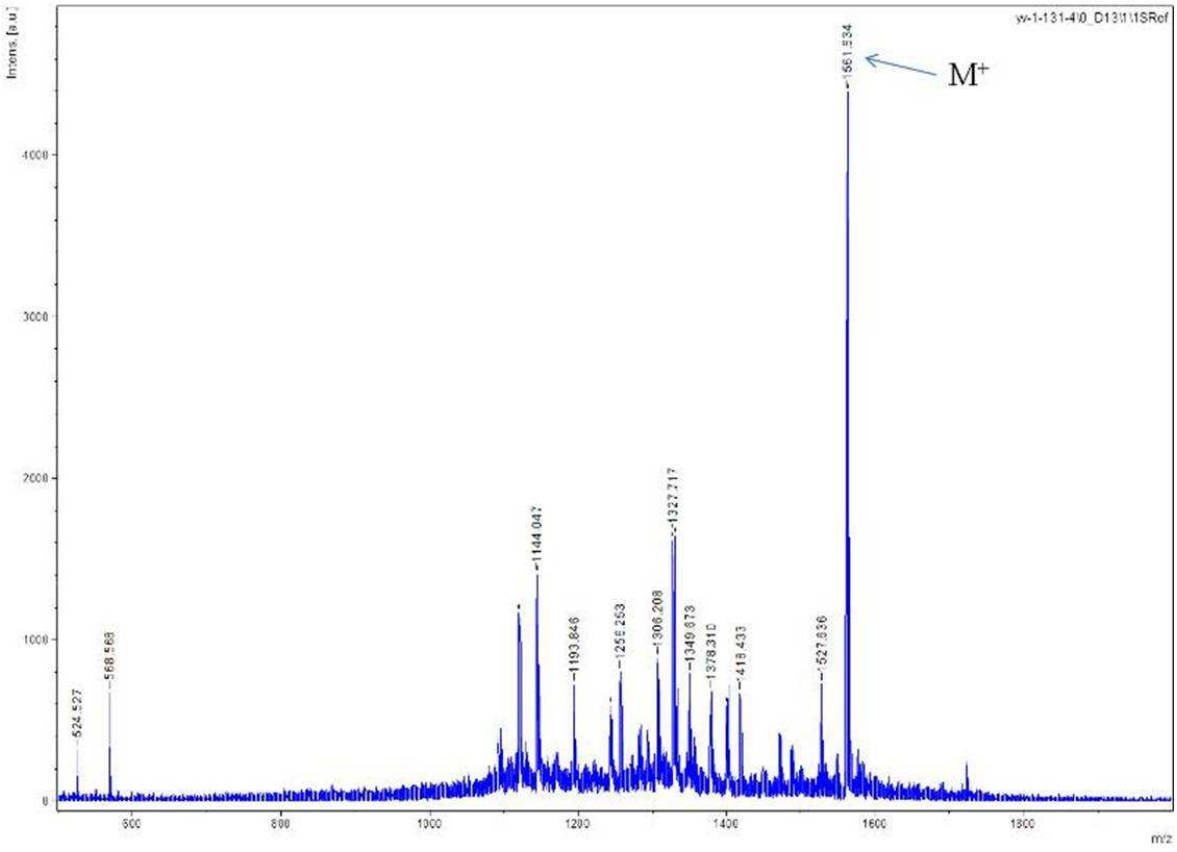
Compound **2**:



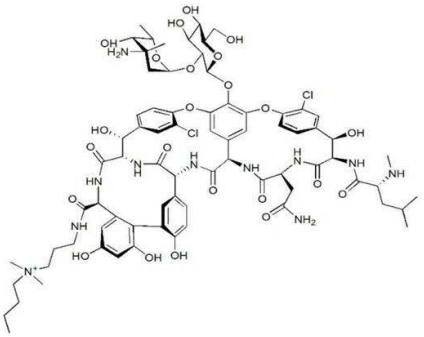

9.415
9.142
9.031
7.587
6.721
6.392
6.387
6.252
5.767
5.749
5.612
5.437
5.421
5.335
5.321
5.265
5.246
5.180
5.124
5.107
5.079
4.689
4.671
4.447
4.433
4.196
4.168
4.015
4.001
3.988
3.710
3.694
3.683
3.672
3.568
3.549
3.528
3.408
2.689
2.678
2.674
2.669
2.664
2.660
2.539
2.331
2.327
2.322
1.731
1.697
1.655
1.289
1.082
1.066
0.925
0.911
0.879
0.864
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 ppm
1.12 1.38 1.33 0.84 1.01 1.01 7.91 0.83 2.49 2.80 1.33 1.41 0.90 0.92 8.00 0.75 0.68 2.56 2.39 1.78 2.03 7.24 6.34 0.20 4.81 3.20 2.00 2.91 0.83 3.08 3.35 8.00

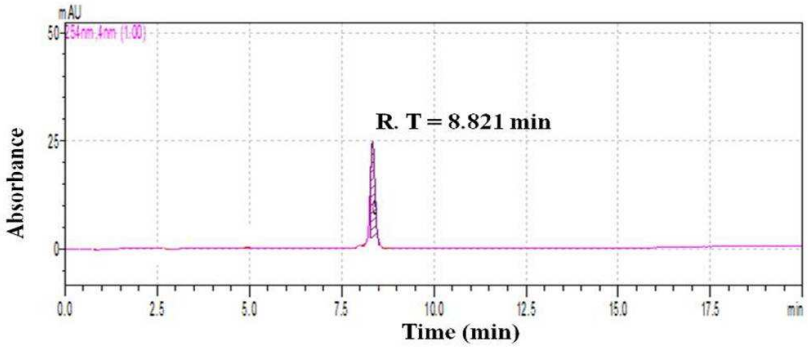
Compound **2**:



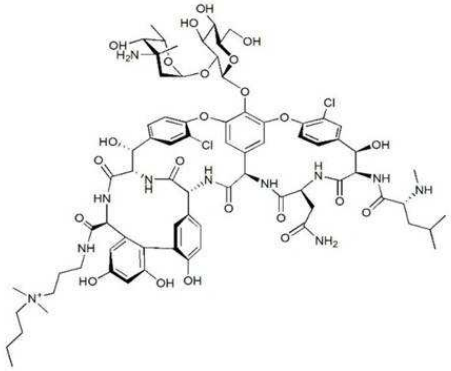

M+
1561.834
1144.047
1327.717
568.568
524.527
1193.846
1256.253
1306.208
1349.873
1378.310
1418.433
1527.836
Intens. [a.u.]
m/z

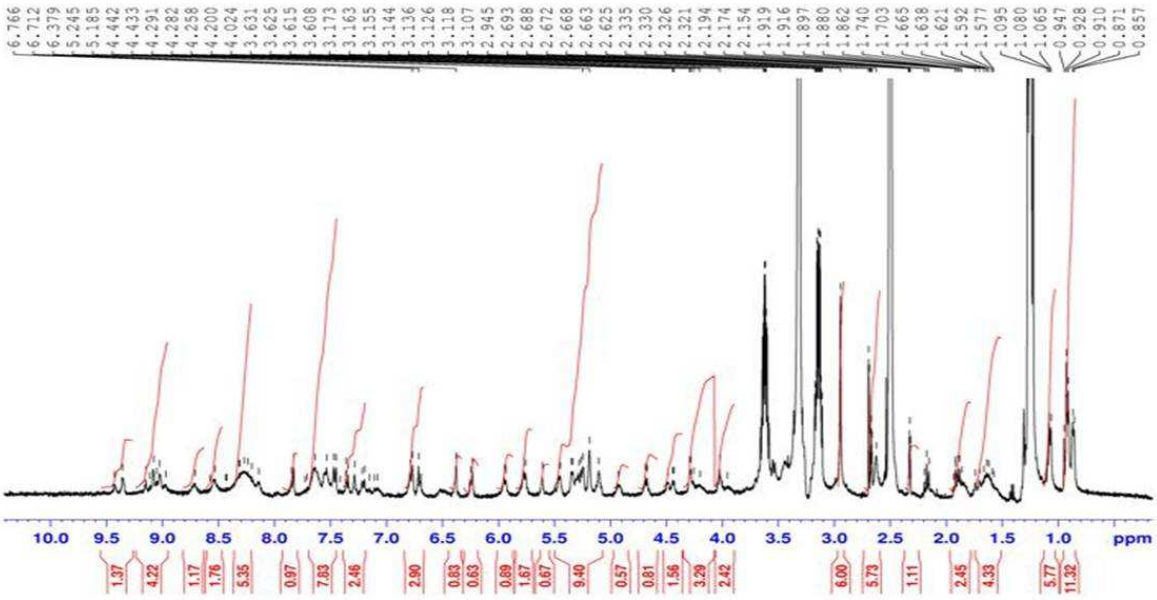
Compound **3**:



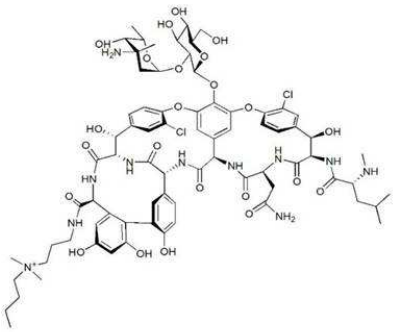

mAU
254nm,4nm (1.00)
50
25
0
R. T = 8.821 min
Absorbance
0.0
2.5
5.0
7.5
10.0
12.5
15.0
17.5
min
Time (min)

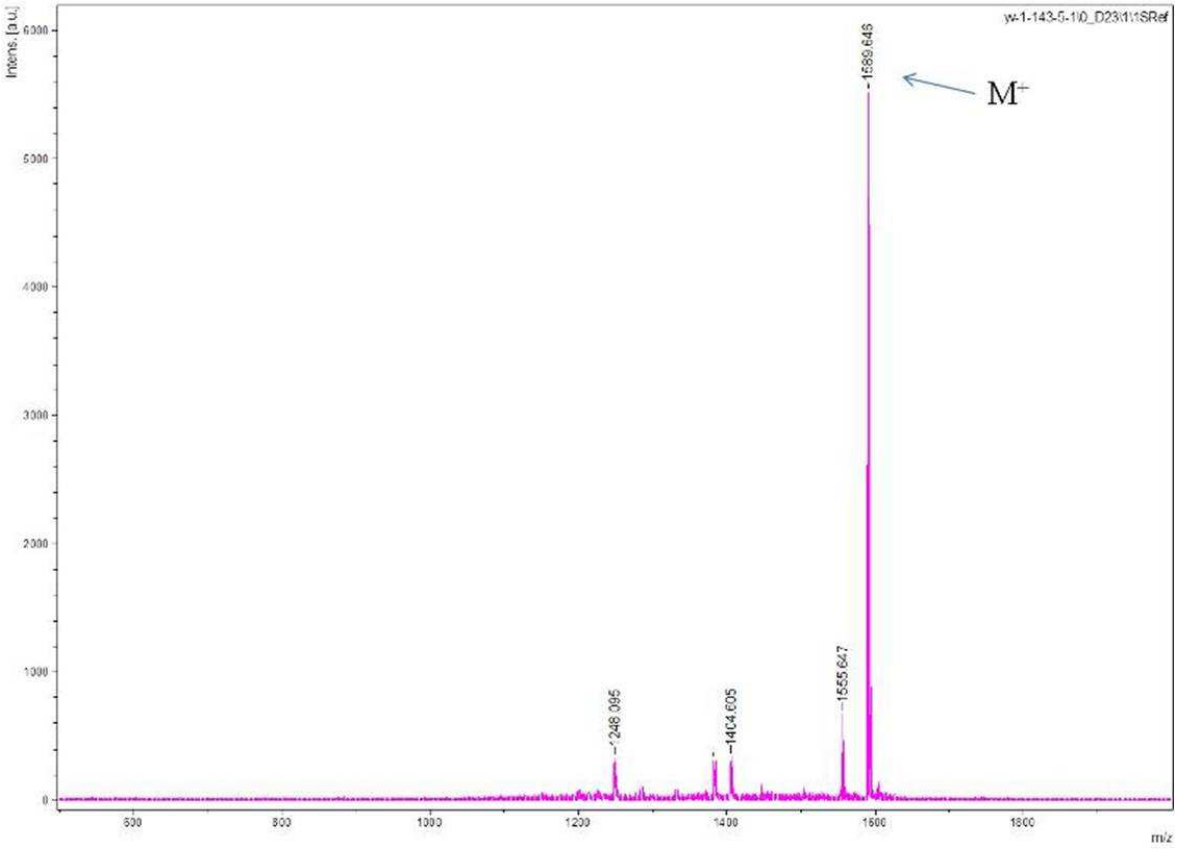
Compound **3**:



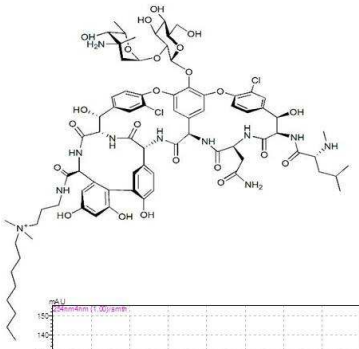

6.766
6.712
6.379
5.245
5.185
4.442
4.433
4.291
4.282
4.258
4.200
4.024
3.631
3.625
3.615
3.608
3.173
3.163
3.155
3.144
3.136
3.126
3.118
3.107
2.945
2.693
2.688
2.672
2.668
2.663
2.625
2.335
2.330
2.326
2.321
2.194
2.174
2.154
1.919
1.916
1.897
1.880
1.862
1.740
1.703
1.665
1.638
1.621
1.592
1.577
1.095
1.080
1.065
0.947
0.928
0.910
0.871
0.857
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
1.37
4.22
1.17
1.76
5.35
0.97
7.83
2.46
2.90
0.83
0.63
0.89
1.67
0.67
9.40
0.57
0.81
1.56
3.29
2.42
6.00
5.73
1.11
2.45
4.33
5.77
11.32

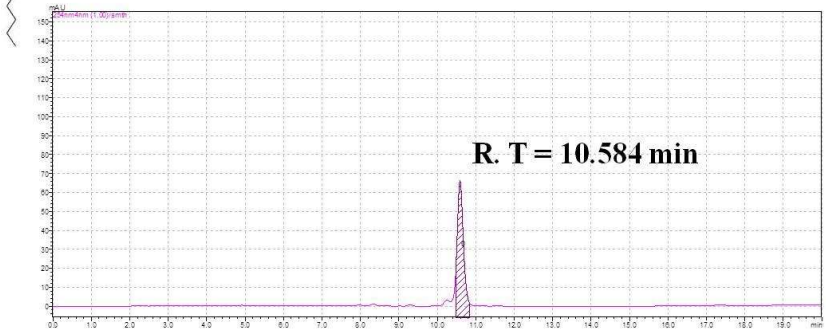
Compound **3**:



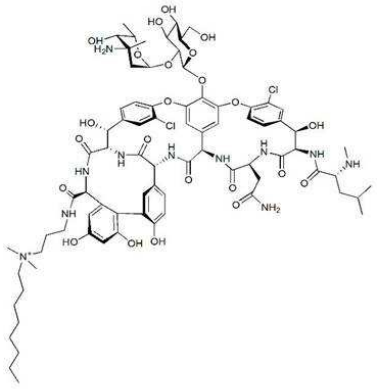

w-1-143-5-1\0_D23\1\1SRef
Intens. [a.u.]
6000
5000
4000
3000
2000
1000
0
1589.648
M+
1555.647
1404.605
1248.095
600
800
1000
1200
1400
1600
1800
m/z

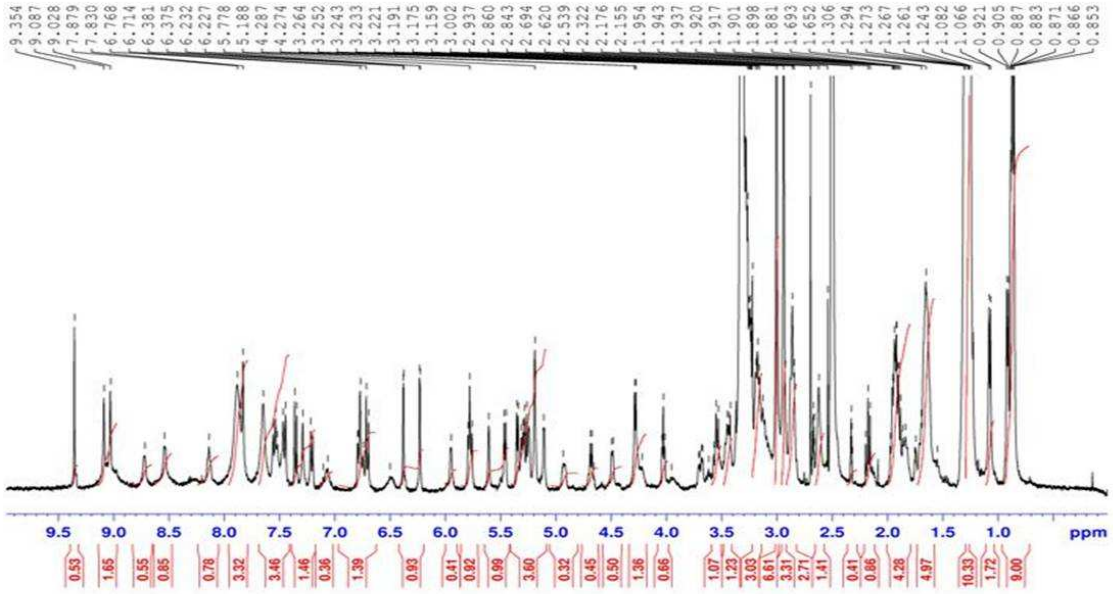
Compound **4**:



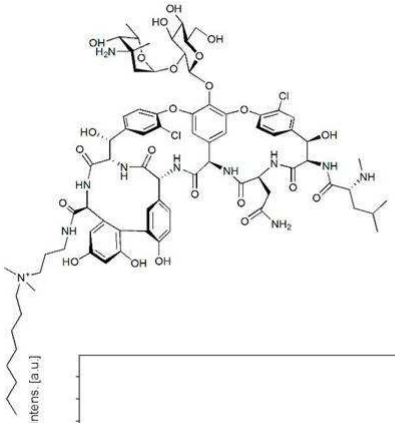


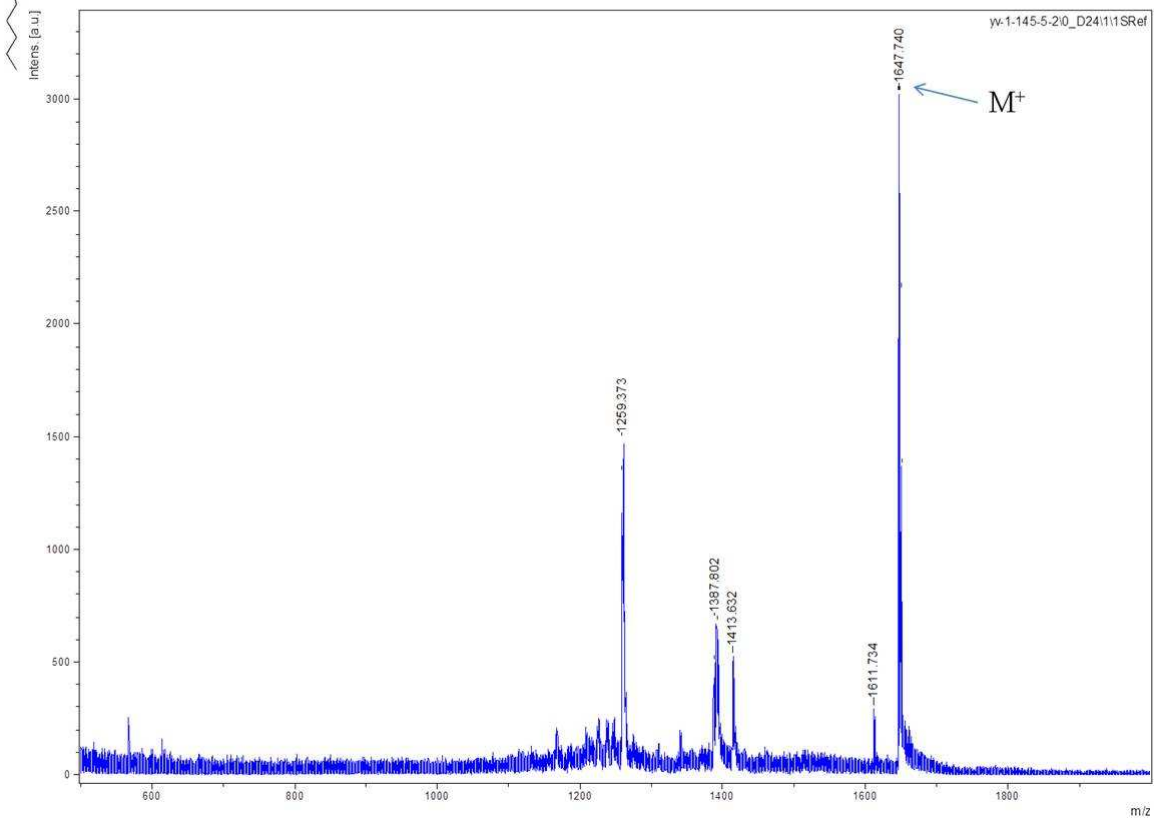
Compound **4**:



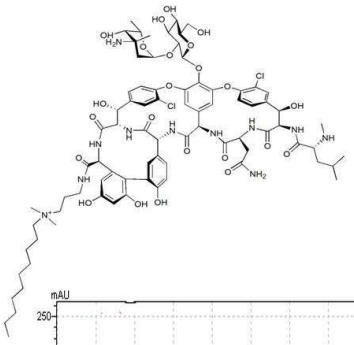

9.354
9.087
9.028
7.879
7.830
6.768
6.714
6.381
6.375
6.232
6.227
5.778
5.188
4.287
4.274
3.264
3.252
3.243
3.233
3.221
3.191
3.175
3.159
3.002
2.937
2.860
2.843
2.694
2.620
2.539
2.322
2.176
2.155
1.954
1.943
1.937
1.920
1.917
1.901
1.898
1.881
1.693
1.652
1.306
1.294
1.273
1.267
1.261
1.243
1.082
1.066
0.921
0.905
0.887
0.883
0.871
0.866
0.853
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
0.53
1.65
0.55
0.85
0.78
3.32
3.46
1.46
0.36
1.39
0.93
0.41
0.82
0.99
3.60
0.32
0.45
0.50
1.36
0.66
1.07
1.23
3.03
6.61
3.31
2.71
1.41
0.41
0.86
4.28
4.97
10.33
1.72
9.00

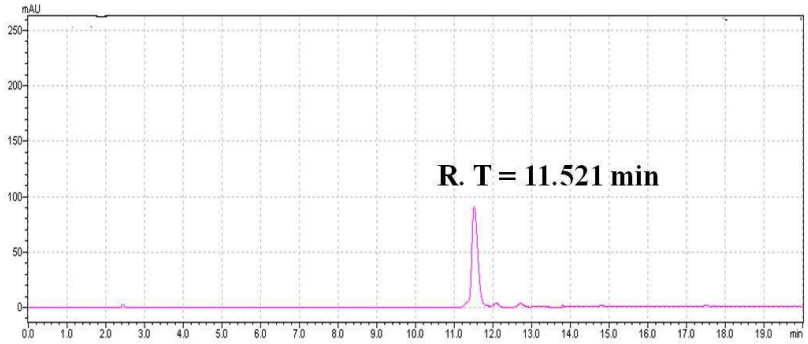
Compound **4**:



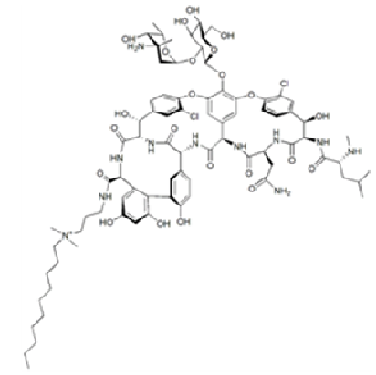

yw-1-145-5-2\0_D24\1\1SRef
Intens. [a.u.]
3000
2500
2000
1500
1000
500
0
1647.740
M+
1259.373
1387.802
1413.632
1611.734
600
800
1000
1200
1400
1600
1800
m/z

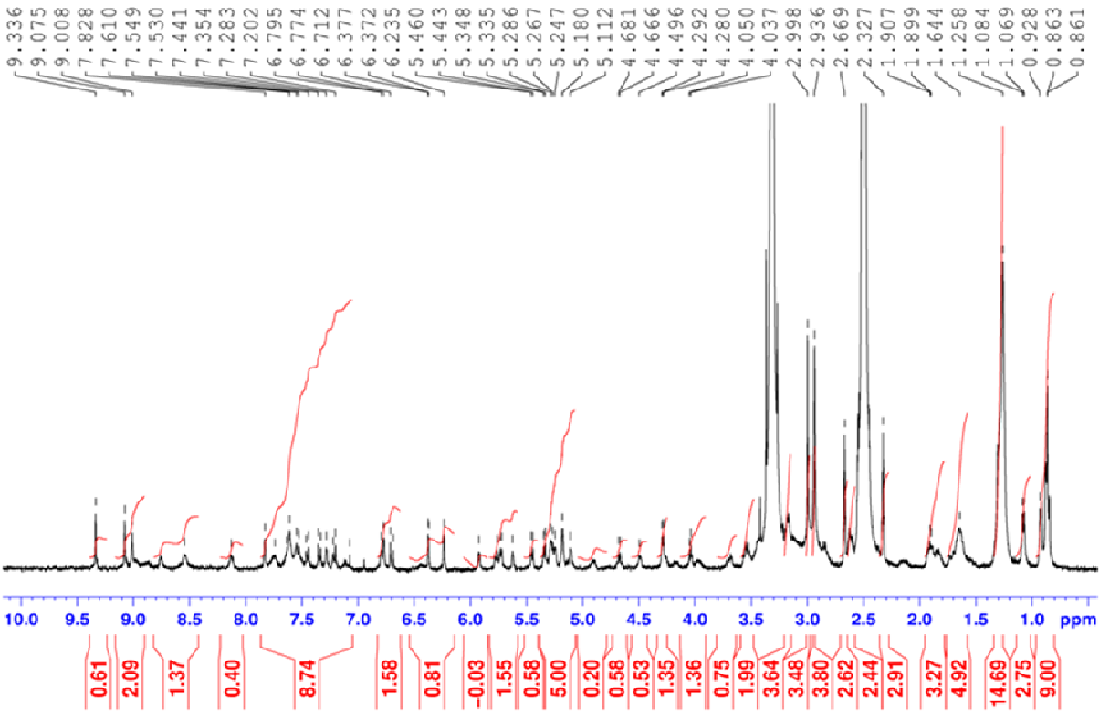
Compound **5**:



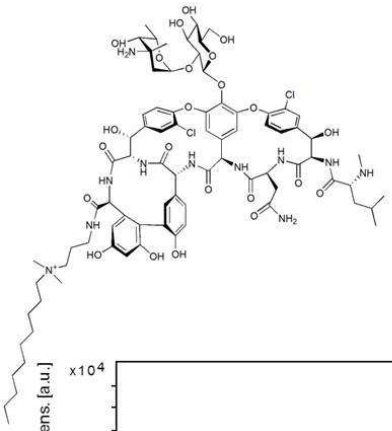

mAU
250
200
150
100
50
0
R. T = 11.521 min
0.0
1.0
2.0
3.0
4.0
5.0
6.0
7.0
8.0
9.0
10.0
11.0
12.0
13.0
14.0
15.0
16.0
17.0
18.0
19.0
min

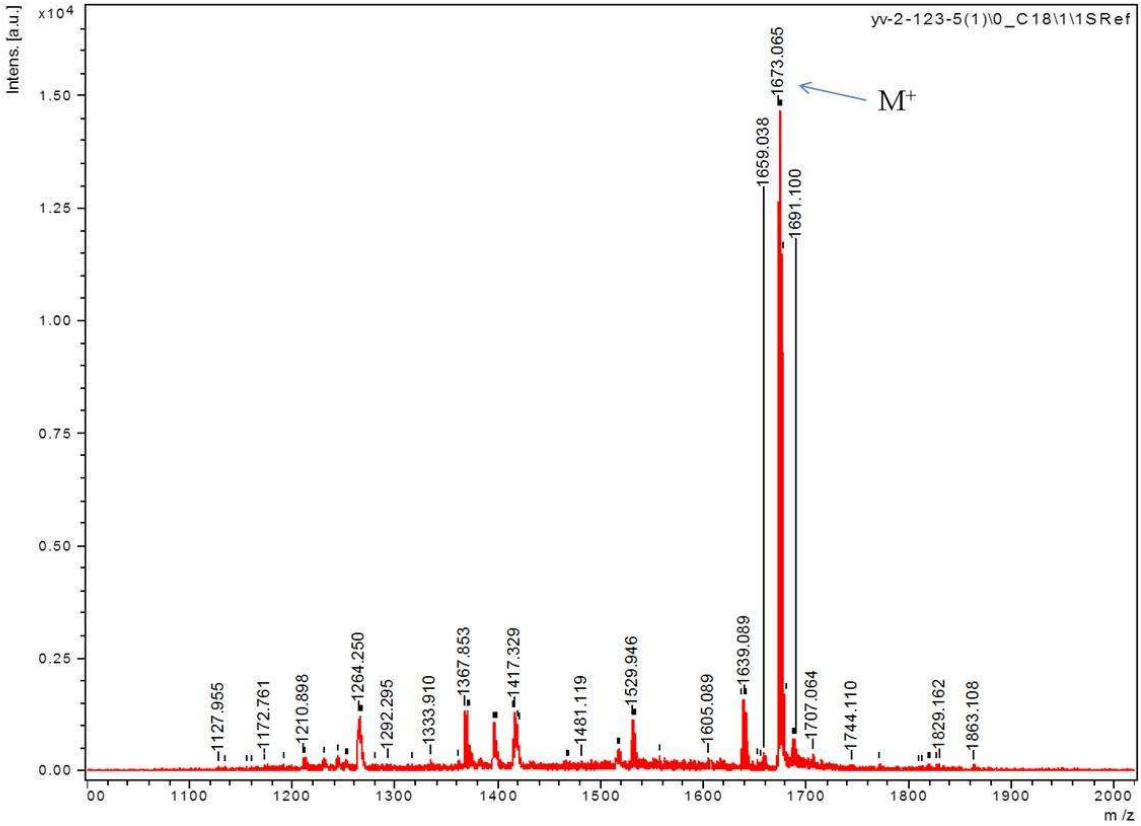
Compound **5**:



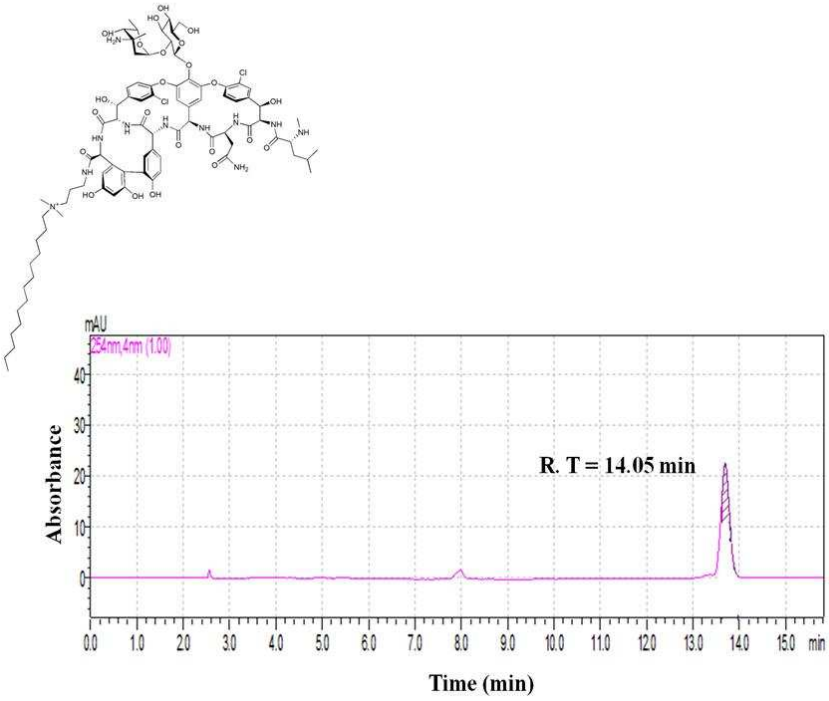

9.336
9.075
9.008
7.828
7.610
7.549
7.530
7.441
7.354
7.283
7.202
6.795
6.774
6.712
6.377
6.372
6.235
5.460
5.443
5.348
5.335
5.286
5.267
5.247
5.180
5.112
4.681
4.666
4.496
4.292
4.280
4.050
4.037
2.998
2.936
2.669
2.327
1.907
1.899
1.644
1.258
1.084
1.069
0.928
0.863
0.861
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 ppm
0.61
2.09
1.37
0.40
8.74
1.58
0.81
-0.03
1.55
0.58
5.00
0.20
0.58
0.53
1.35
1.36
0.75
1.99
3.64
3.48
3.80
2.62
2.44
2.91
3.27
4.92
14.69
2.75
9.00

Compound **5**:

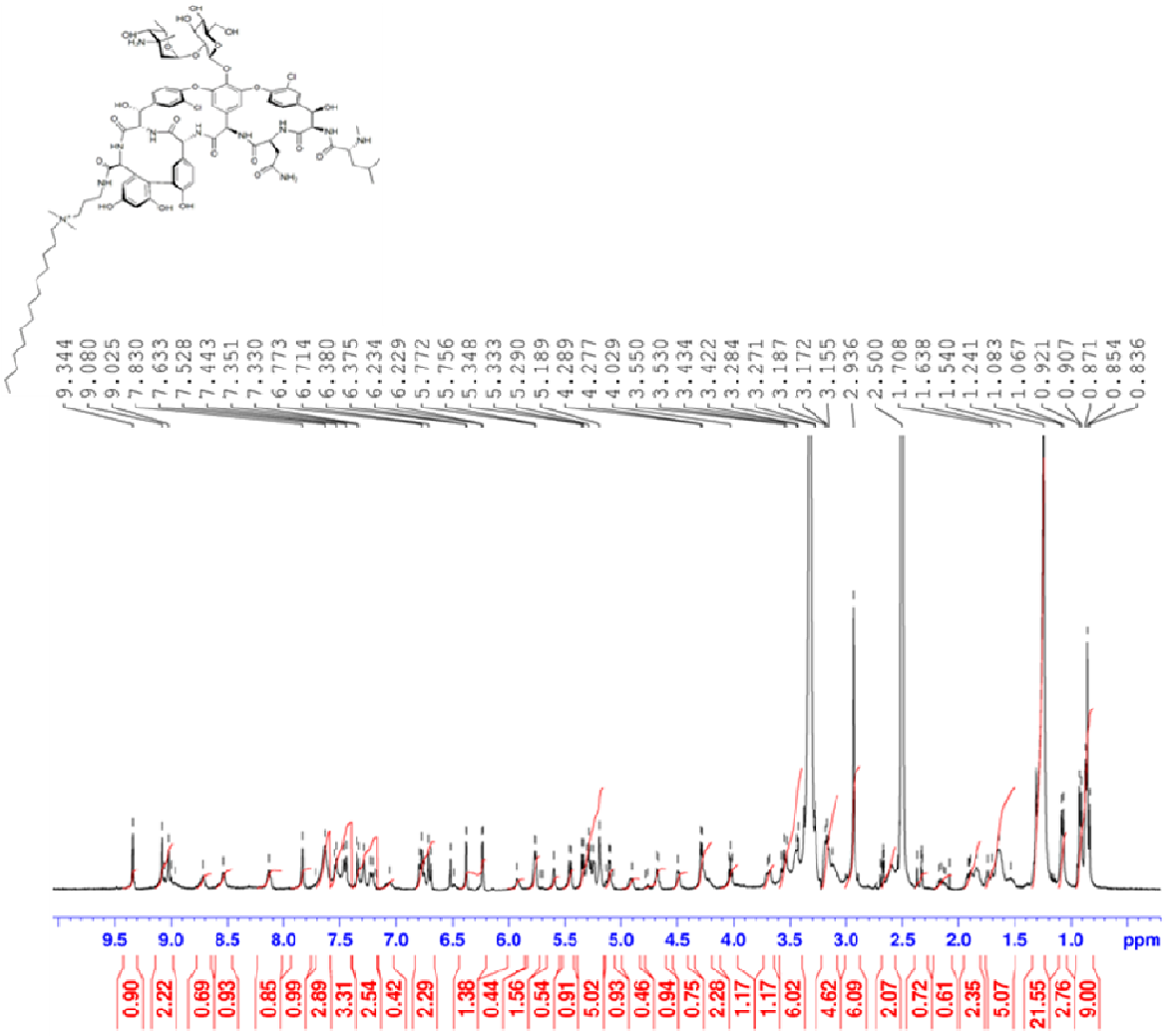



yv-2-123-5(1)\0_C18\1\1SRef
Intens. [a.u.]
x10^4
1.50
1.25
1.00
0.75
0.50
0.25
0.00
1673.065
M+
1658.038
1691.100
1127.955
1172.761
1210.898
1264.250
1292.295
1333.910
1367.853
1417.329
1481.119
1529.946
1605.089
1639.089
1707.064
1744.110
1829.162
1863.108
1100
1200
1300
1400
1500
1600
1700
1800
1900
2000
m/z

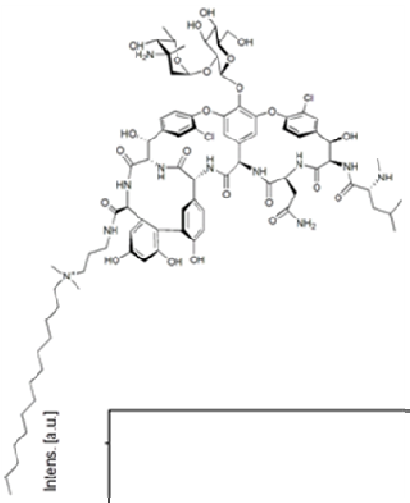
Compound **6**:

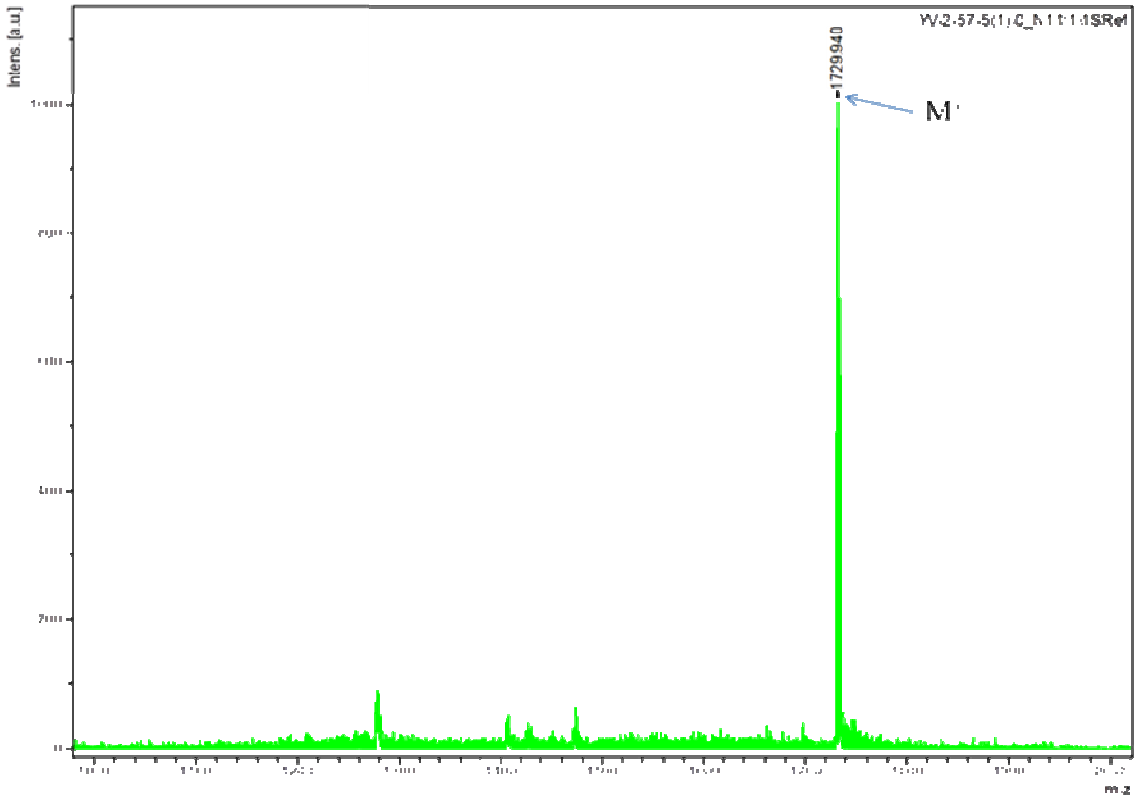


Compound **6**:

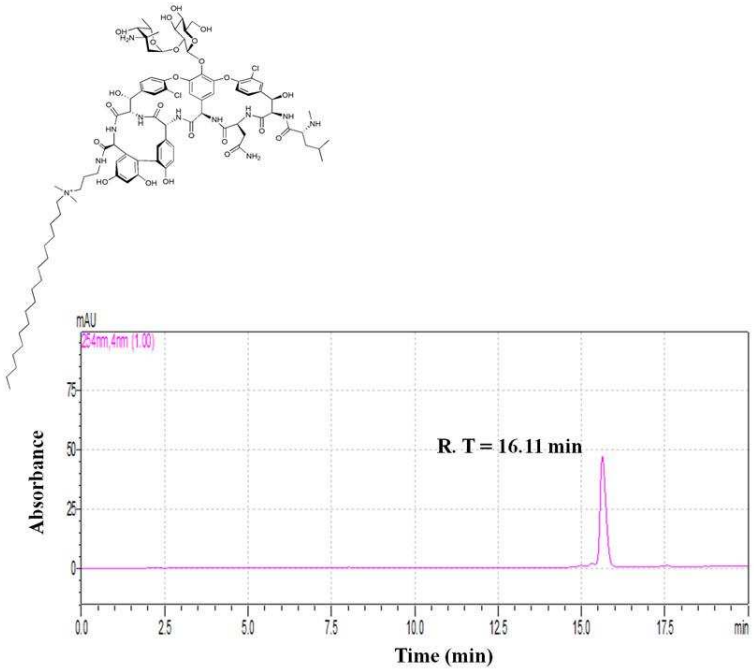

9.344
9.080
9.025
7.830
7.633
7.528
7.443
7.351
7.330
6.773
6.714
6.380
6.375
6.234
6.229
5.772
5.756
5.348
5.333
5.290
5.189
4.289
4.277
4.029
3.550
3.530
3.434
3.422
3.284
3.271
3.187
3.172
3.155
2.936
2.500
1.708
1.638
1.540
1.241
1.083
1.067
0.921
0.907
0.871
0.854
0.836
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
0.90
2.22
0.69
0.93
0.85
0.99
2.89
3.31
2.54
0.42
2.29
1.38
0.44
1.56
0.54
0.91
5.02
0.93
0.46
0.94
0.75
2.28
1.17
1.17
6.02
4.62
6.09
2.07
0.72
0.61
2.35
5.07
21.55
2.76
9.00

Compound **6**:

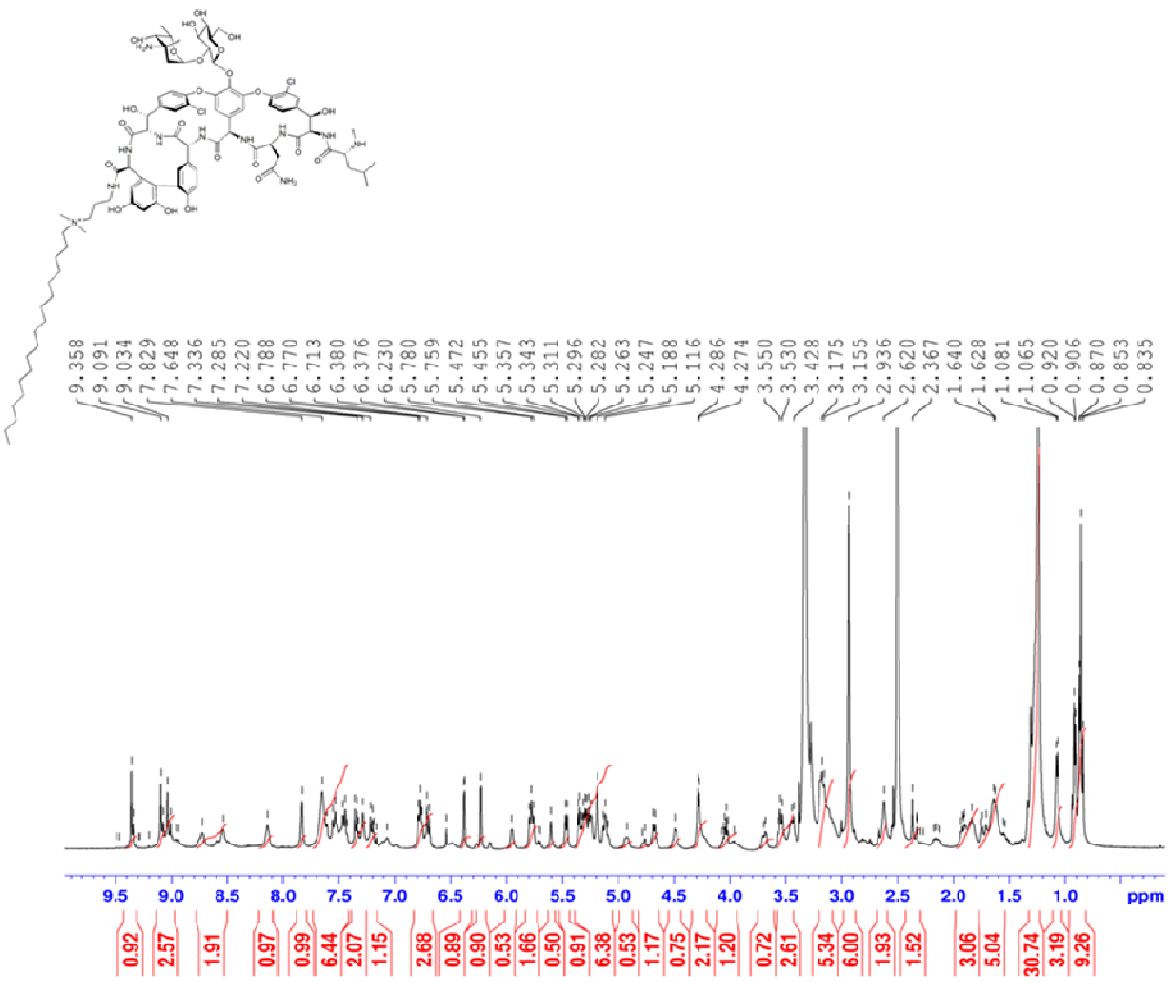




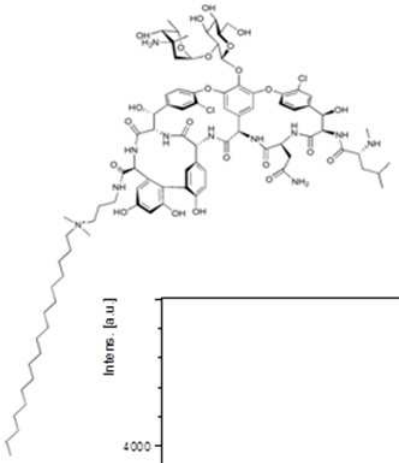
Compound **7**:

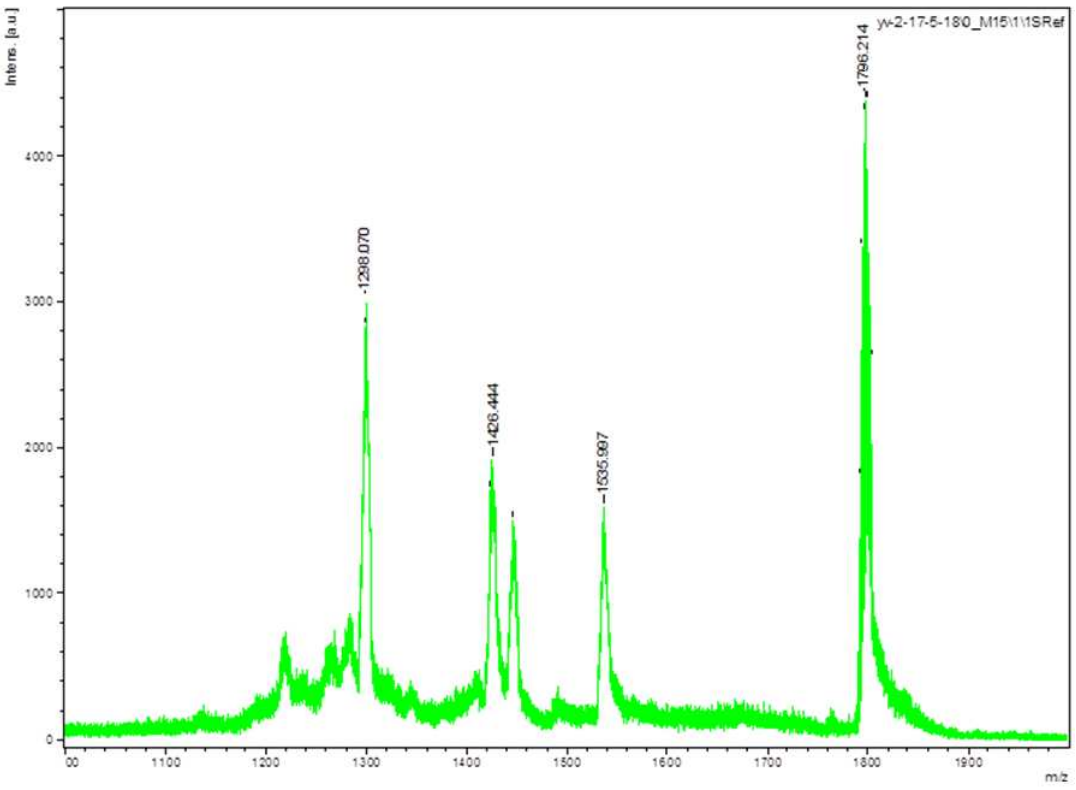

mAU
254nm,4nm (1.00)
75
50
25
0
Absorbance
R. T = 16.11 min
0.0
2.5
5.0
7.5
10.0
12.5
15.0
17.5
min
Time (min)

Compound **7**:

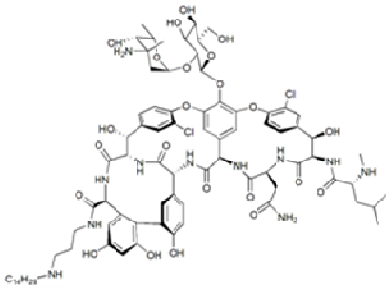

9.358
9.091
9.034
7.829
7.648
7.336
7.285
7.220
6.788
6.770
6.713
6.380
6.376
6.230
5.780
5.759
5.472
5.455
5.357
5.343
5.311
5.296
5.282
5.263
5.247
5.188
5.116
4.286
4.274
3.550
3.530
3.428
3.175
3.155
2.936
2.620
2.367
1.640
1.628
1.081
1.065
0.920
0.906
0.870
0.853
0.835
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
0.92
2.57
1.91
0.97
0.99
6.44
2.07
1.15
2.68
0.89
0.90
0.53
1.66
0.50
0.91
6.38
0.53
1.17
0.75
2.17
1.20
0.72
2.61
5.34
6.00
1.93
1.52
3.06
5.04
30.74
3.19
9.26

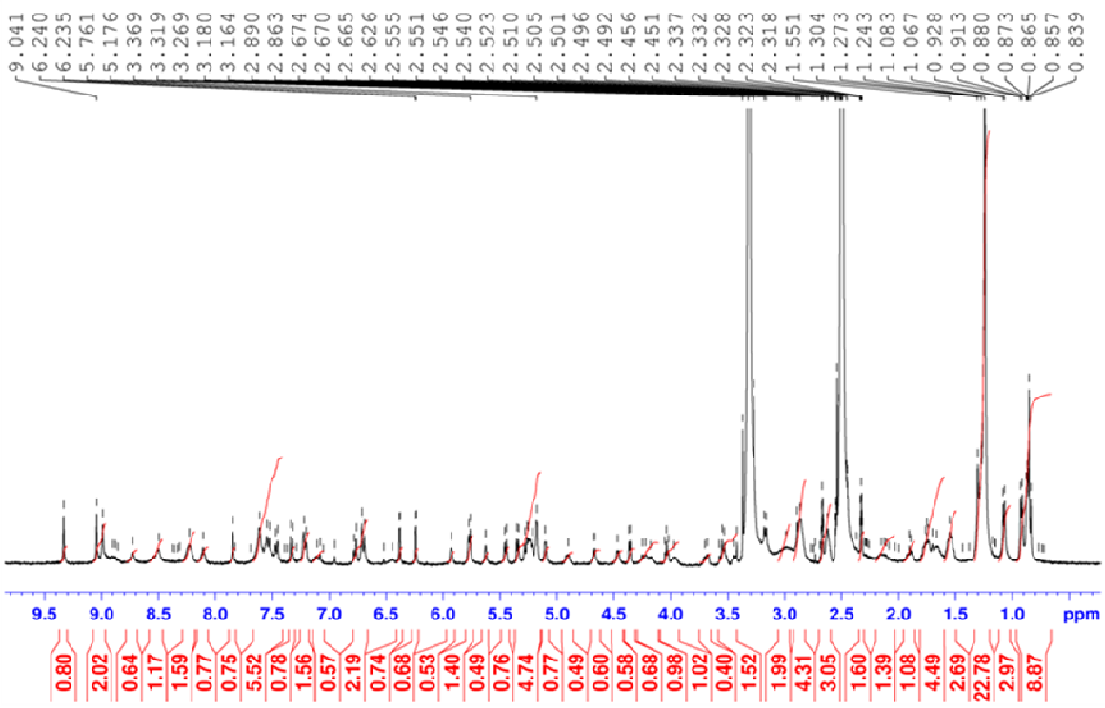
Compound **7**:




yw-2-17-5-180_M15\1\1SRef
Intens. [a.u.]
4000
3000
2000
1000
0
1298.070
1426.444
1535.997
1796.214
1100
1200
1300
1400
1500
1600
1700
1800
1900
m/z

Compound **13**:




9.041
6.240
6.235
5.761
5.176
3.369
3.319
3.269
3.180
3.164
2.890
2.863
2.674
2.670
2.665
2.626
2.555
2.551
2.546
2.540
2.523
2.510
2.505
2.501
2.496
2.492
2.456
2.451
2.337
2.332
2.328
2.323
2.318
1.551
1.304
1.273
1.243
1.083
1.067
0.928
0.913
0.880
0.873
0.865
0.857
0.839
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
0.80
2.02
0.64
1.17
1.59
0.77
0.75
5.52
0.78
1.56
0.57
2.19
0.74
0.68
0.53
1.40
0.49
0.76
4.74
0.77
0.49
0.60
0.58
0.68
0.98
1.02
0.40
1.52
1.99
4.31
3.05
1.60
1.39
1.08
4.49
2.69
22.78
2.97
8.87

Compound **13**: