Site-Selective Bromination of Vancomycin

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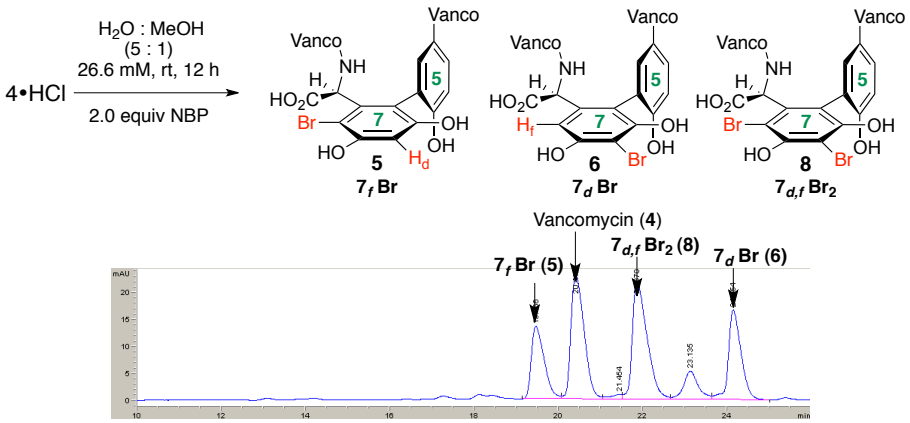
General Information

Proton NMR spectra were collected on 500 MHz spectrometers at 25 °C. Proton chemical shifts are reported in ppm (δ) relative to the solvent reference relative to TMS employed as the internal standard ($CDCl_3$, δ 7.26 ppm; DMSO δ 2.50 ppm; D_2O, δ 4.79 ppm, CD_3OD δ 3.34 ppm). Data are reported as follows: chemical shift (multiplicity [singlet (s), doublet (d), triplet (t), quartet (q), and multiplet (m)], coupling constants [Hz], integration). Infrared spectra were obtained using a FT-IR instrument utilizing a Small Orbit FT-IR ATR (Attenuated total reflectance) module. Specific rotations were determined with a Polarimeter 341 at 20 °C on the sodium D line (path length 10.0 cm). High-resolution mass spectra were obtained from institutional providers (high resolution LCMS) and the method of ionization reported herein. All solvents were either distilled or taken from a solvent purification system except H_2O, in which case DI water was used as a solvent. Vancomycin•HCl was purchased from commercial supplier (95% purity by HPLC analysis at 280 nm) and was used without further purification. No precautions were taken to exclude air unless otherwise noted. MIC determinations were performed by Micromyx, LLC (Kalamazoo, MI, USA). All other reagents were purchased from commercial sources and used without further purification. *N*-bromopthalimide (NBP) was recrystallized and was kept away from light.

Note: *Vancomycin and all the reported brominated derivatives were kept in a freezer (at −20 °C) to avoid decomposition.*

Procedure for Unselective Bromination of Vancomycin (4), Figure 2:

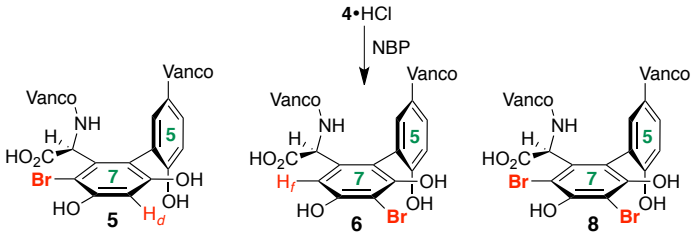


To a round-bottom flask equipped with a stir bar were added 500.0 mg (0.336 mmol, 1.0 equiv) of vancomycin•HCl (**4**). Water (10.0 mL) and MeOH (2.0 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min after which 150.0 mg (0.672 mmol, 2.0 equiv) of NBP were added. The reaction mixture was stirred at room temperature. After 20 h, 10 mL of EtOAc were added and the reaction was stirred for an additional 15 min. The organic layer was discarded and the aqueous layer was diluted with 2 mL of water. The reaction was analyzed by reverse phase HPLC (see below) and was purified by preparative reverse phase HPLC (see below).
Purification of the reaction mixture yielded, 60.0 mg (11% Yield) of 7_f-Br vancomycin (**5**), 58.0 mg (11% Yield) of 7_d-Br vancomycin (**6**), and 131.1 mg (23% Yield) of $7_{d,f}$-Br_2 vancomycin (**8**).
Each of these isomers was characterized based on change in characteristic chemical shifts in their HSQC spectra. HSQC spectra of compound **5**, **6**, and **7** were recorded in both D_2O and DMSO-d_6 and were compared with HSQC spectrum of native vancomycin (**4**). The raw spectral data are presented, including overlays with vancomycin. In the case of 7_d-Br, we also observed ROESY correlation between 7_f-H with X_7-H indicating the presence of 7_f-H, thus supporting substitution at the 7_d site of resorcinol ring. A similar ROESY correlation was absent in case of compound **5**.

HPLC Method for preparative purification: Symmetry prep C8 7µm (19 X 300 mm) column, 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

Analytical method: Symmetry prep C8 7µm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

Optimization of reaction conditions (Manuscript Table 1):



Entry	Catalyst	mol%	t (h)	% conv.	**5** : **6** : **8**
					7_f**Br** : 7_d**Br** : $7_{d,f}$**Br**$_2$
1	No catalyst	NA	12	71	1.0 : 1.0 : 1.3[b]
2a	*N,N*-dimethylacetamide	100	12	68	1.0 : 1.0 : 1.8[b]
2b	*N,N*-dimethylacetamide	7100	16	64	1.0 : 1.5 : 2.9[b]
3	Boc-Asn(Me_2)-DAla-DAla-OH (**10**)	100	2	97	1.0 : 6.8 : 2.6[b]
4	Boc-DAsn(Me_2)-DAla-DAla-OH (**11**)	100	2	90	1.0 : 4.9 : 1.8[b]
5	Boc-Gln(Me_2)-DAla-DAla-OH (**12**)	100	2	99	1.0 : 4.0 : 9.3[b]
6	Boc-DGln(Me_2)-DAla-DAla-OH (**13**)	100	2	99	1.0 : 3.0 : 3.4[b]
7	Boc-Leu-DAla-DAla-OH (**14**)	100	12	60	1.0 : 5.7 : 2.5[b]
8a	Boc-Asn(Me_2)-DAla-DAla-OH (**10**)	100	1.5	98	**1.0 : 14.6** : 2.8[c]
8b		200	1.5	99	1.0 : 19.0 : 6.0[c]
8c		50	2	97	1.0 : 3.4 : 1.1[c]
9	Boc-Leu-DAsn(Me_2)-DAla-OH (**15**)	200	12	96	1.0 : 6.5 : 2.7[c]
10	Boc-Leu-DAla-DAsn(Me_2)-OH (**16**)	200	12	85	1.0 : 3.5 : 1.0[c]
11	Boc-Asn(Me_2)-DAla-DAla-OH (**10**)	100	1.5	16	1.0 : 10.5 : 0.9[d]
12	Boc-Leu-DAla-DAsn(Me_2)-OH (**16**)	100	1.5	12	1.0 : 3.5 : 1.6[d]

a) Ratios were measured by HPLC at 280 nm wavelength. b) 2.0 equiv of NBP, 250 μL of water, 50 μL of MeOH, 8 μmol of **4**. c) 2.0 equiv of NBP, 1000 μL of water, 200 μL of MeOH, 8 μmol of **4**. d) 50 mol% NBP, 0.033 mmol of **4**.

*Note: Conversions and product ratios are uncorrected. The product ratios are highly dependent on reaction time, as each monobrominated product can be further functionalized. Thus vigorously following reaction by LC-MS, is recommended to obtained the most reproducible product distribution. Data presented in Table **1** were recorded at the indicated time. For larger scale reactions reaction progress was monitored by LCMS to obtain optimal product distribution. Conversion is defined as (100 – area % of **4**).*

Analytical method for analysis of reaction mixture: Symmetry prep C8 7μm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

General Procedure for Optimization Table:

Entry 1: To a HPLC vial equipped with a stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl. Water (250 μL) and MeOH (50 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 2a: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl and 250 μL of DMA (*Standard solution was prepared as follows: 50 μL of DMA was added to 16.8 mL of water and 250 μL of this mixture was used as a solvent for the reaction*). To this 50 μL of MeOH were added. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and were analyzed by reverse phase HPLC.

Entry 2b: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl, 250 μL of H_2O, 50 μL of MeOH and 50 μL of DMA were added. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and were analyzed by reverse phase HPLC.

Entry 3: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl and 3.2 mg (0.008 mmol, 1.0 equiv) of peptide **10**. Water (250 μL) and MeOH (50 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. An aliquot was collected at 120 min and analyzed by reversed phase HPLC.

Entry 4: The same procedure as entry 2 was employed, except peptide **11** was used.

Entry 5: The same procedure as entry 2 was employed, except peptide **12** was used.

Entry 6: The same procedure as entry 2 was employed, except peptide **13** was used.

Entry 7: The same procedure as entry 2 was employed, except peptide **14** was used.

Entry 8a: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl and 3.2 mg (0.008 mmol, 1.0 equiv) of peptide **10**. Water (1000 μL) and MeOH (250 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. An aliquot was collected at 90 min and analyzed by reverse phase HPLC.

Entry 8b: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl and 6.4 mg (0.016 mmol, 2.0 equiv) of peptide **10**. Water (1000 μL) and MeOH (250 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. An aliquot was collected at 90 min and analyzed by reverse phase HPLC.

Entry 8c: To a HPLC vial equipped with stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl and 1.6 mg (0.004 mmol, 0.5 equiv) of peptide **10.** Water (1000 μL) and MeOH (250 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. An aliquot was collected at 90 min and analyzed by reverse phase HPLC.

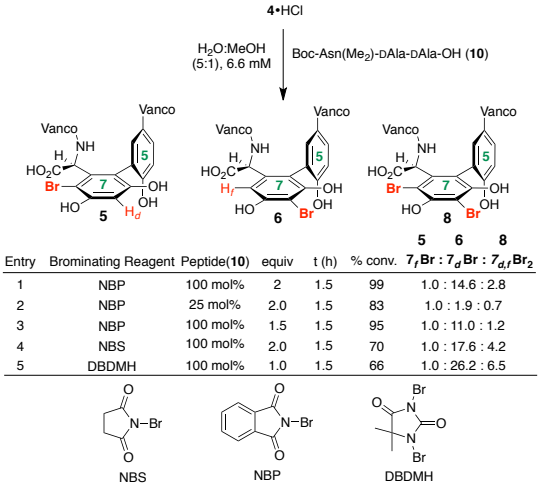
Entry 9: The same procedure as entry 2 was employed, except peptide **15** was used.

Entry 10: The same procedure as entry 2 was employed, except peptide **16** was used.

Entry 11: To a HPLC vial equipped with stir bar were added 49.0 mg (0.033 mmol, 1.0 equiv) of vancomycin•HCl and 13.2 mg (0.033 mmol, 1.0 equiv) of peptide **10**. Water (4000 μL) and MeOH (800 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 0.5 equiv) of NBP were added. An aliquot was collected at 90 min and analyzed by reverse phase HPLC.

Entry 12: To a HPLC vial equipped with stir bar were added 49.0 mg (0.033 mmol, 1.0 equiv) of vancomycin•HCl and 14.9 mg (0.033 mmol, 1.0 equiv) of peptide **16**. Water (4000 μL) and MeOH (800 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 0.5 equiv) of NBP were added. An aliquot was collected at 90 min and analyzed by reverse phase HPLC.

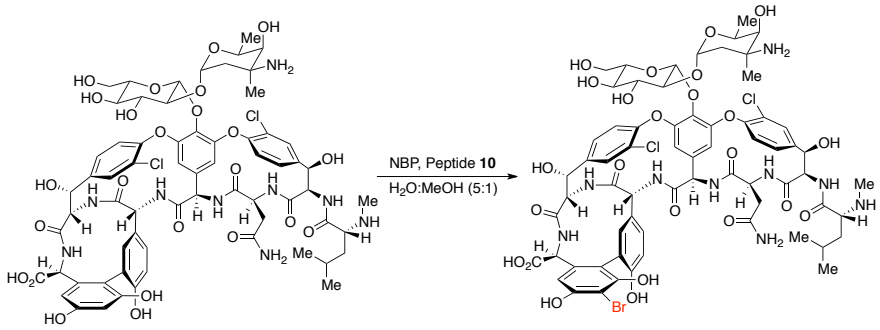
Table 2. Effect of Brominating Reagent on Product Ratio



Entry	Brominating Reagent	Peptide(**10**)	equiv	t (h)	% conv.	**5** : **6** : **8** 7_f**Br** : 7_d**Br** : $7_{d,f}$**Br**$_2$
1	NBP	100 mol%	2	1.5	99	1.0 : 14.6 : 2.8
2	NBP	25 mol%	2.0	1.5	83	1.0 : 1.9 : 0.7
3	NBP	100 mol%	1.5	1.5	95	1.0 : 11.0 : 1.2
4	NBS	100 mol%	2.0	1.5	70	1.0 : 17.6 : 4.2
5	DBDMH	100 mol%	1.0	1.5	66	1.0 : 26.2 : 6.5

The same procedure as entry 8a (Table 1) was employed, except appropriate reagents. We selected NBP because it gives conversion of vancomycin (in 90 min) compared to other brominating reagents. Moreover, the resultant phthalimide is insoluble in water and could easily be removed *via* simple wash with EtOAc, making the purification much easier on larger scale reactions.

Procedure for Bromination of Vancomycin (4) Targeting Analog 6 (Manuscript Figure 5):

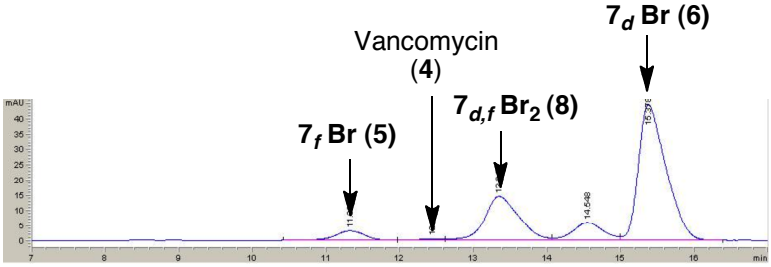


To a round bottom flask equipped with a stir bar were added 100.0 mg (0.067 mmol, 1.0 equiv) of vancomycin•HCl and 26.9 mg (0.067 mmol, 1.0 equiv) of peptide **10**. Water (8.4 mL) and MeOH (1.6 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min after which 30.7 mg (0.135 mmol, 2.0 equiv) of NBP were added. The reaction mixture was stirred at room temperature for *ca.* 90 min, until complete conversion of vancomycin was observed by LC-MS (over the period of time the reaction became cloudy). To the reaction vessel, 10 mL of EtOAc was added and reaction was stirred for an additional 15 min. The organic layer was discarded and the aqueous layer was diluted with 2 mL of water. The reaction was analyzed by reverse phase HPLC and was purified by preparative reverse phase HPLC. Purification of reaction mixture yielded, 43.1 mg (41% Yield) 7_d -Br vancomycin (**6**). ES-HRMS ($M+H^+$): Predicted 1526.3485, observed 1526.3184.

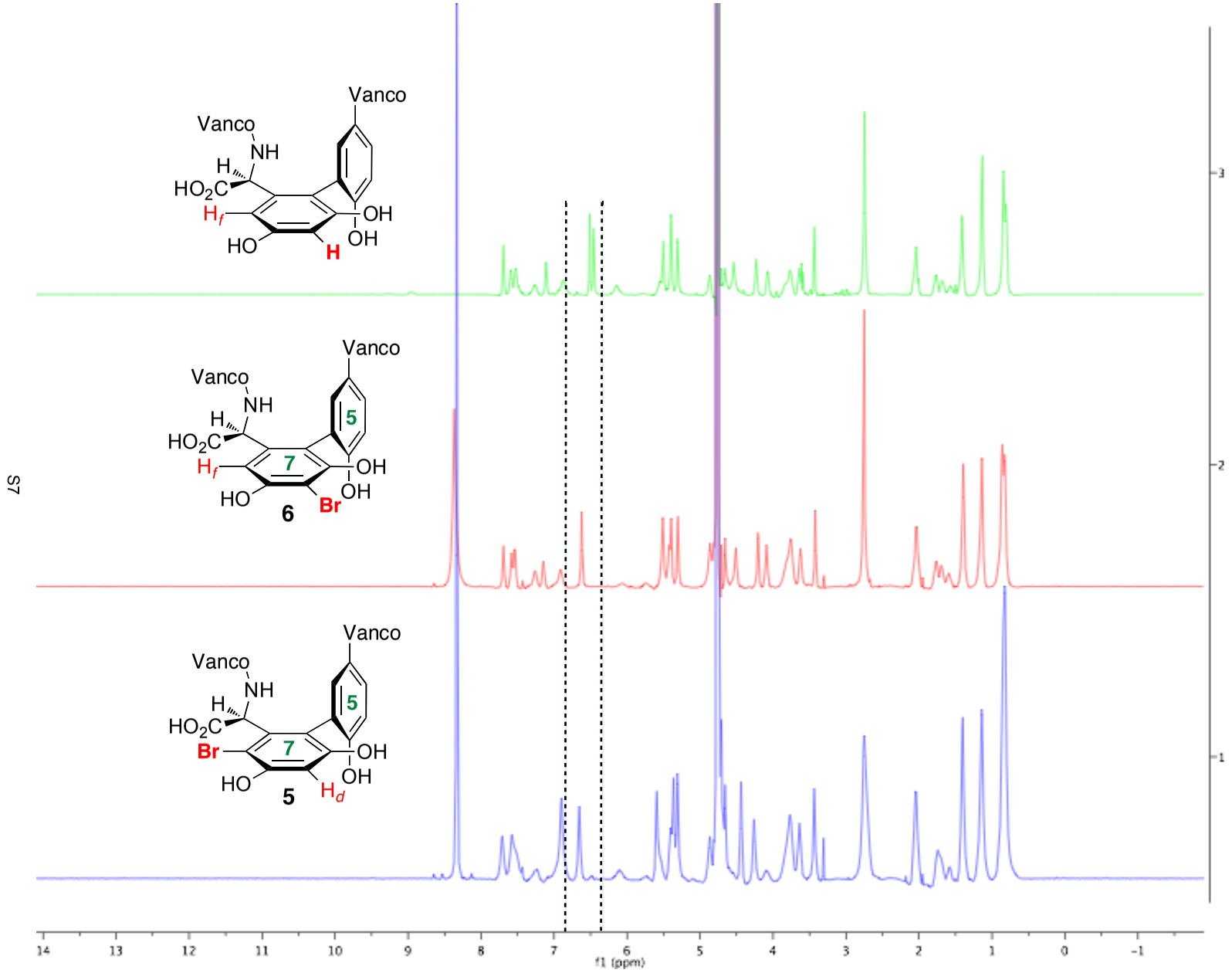
HPLC Method for preparative purification: Symmetry prep C8 7μm (19 X 300 mm) column, 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

Analytical method: Symmetry prep C8 7μm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min. *Retention time of **6** on C8 column is 15.3 min.*

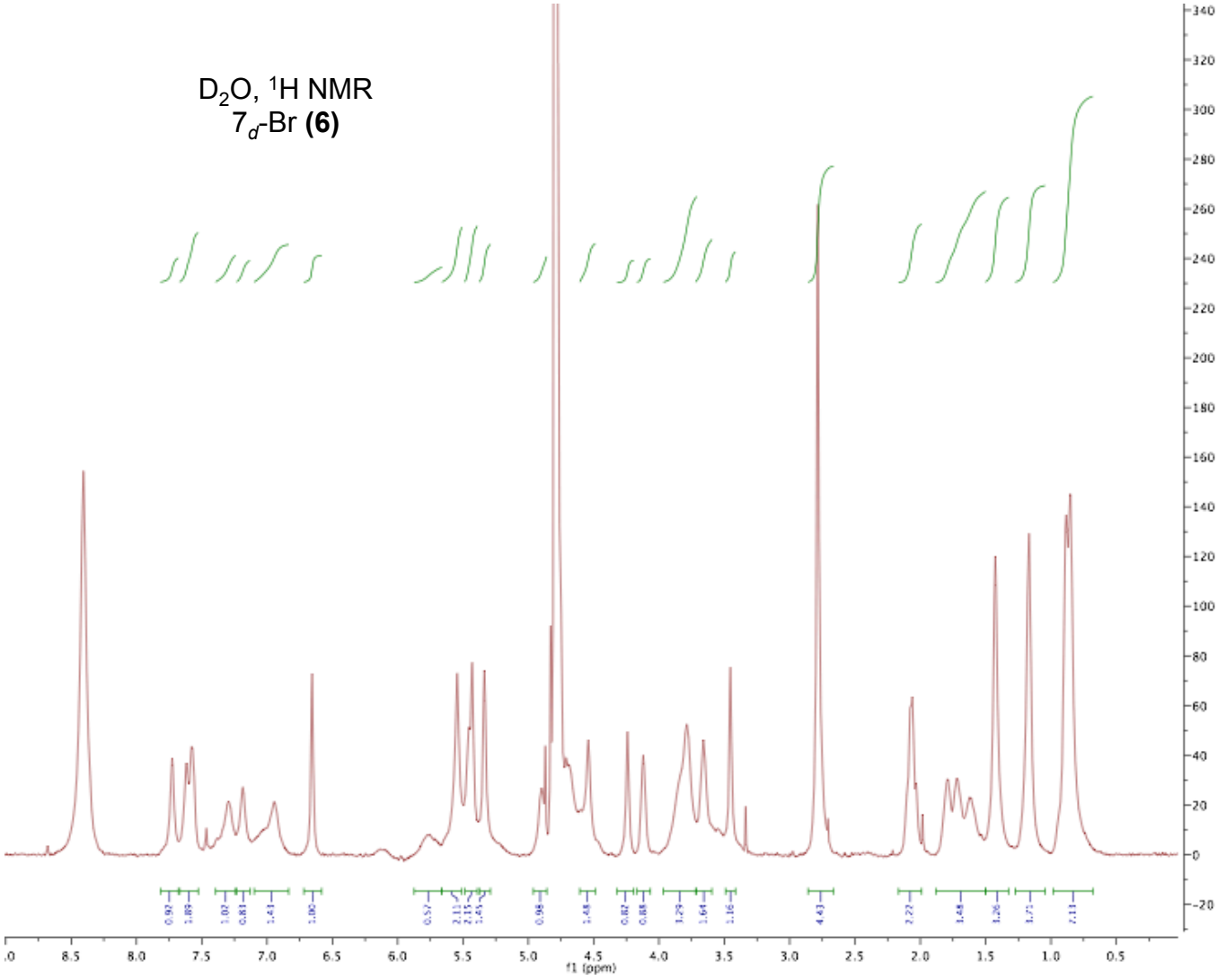
HPLC trace for the reaction mixture (at 280 nm):

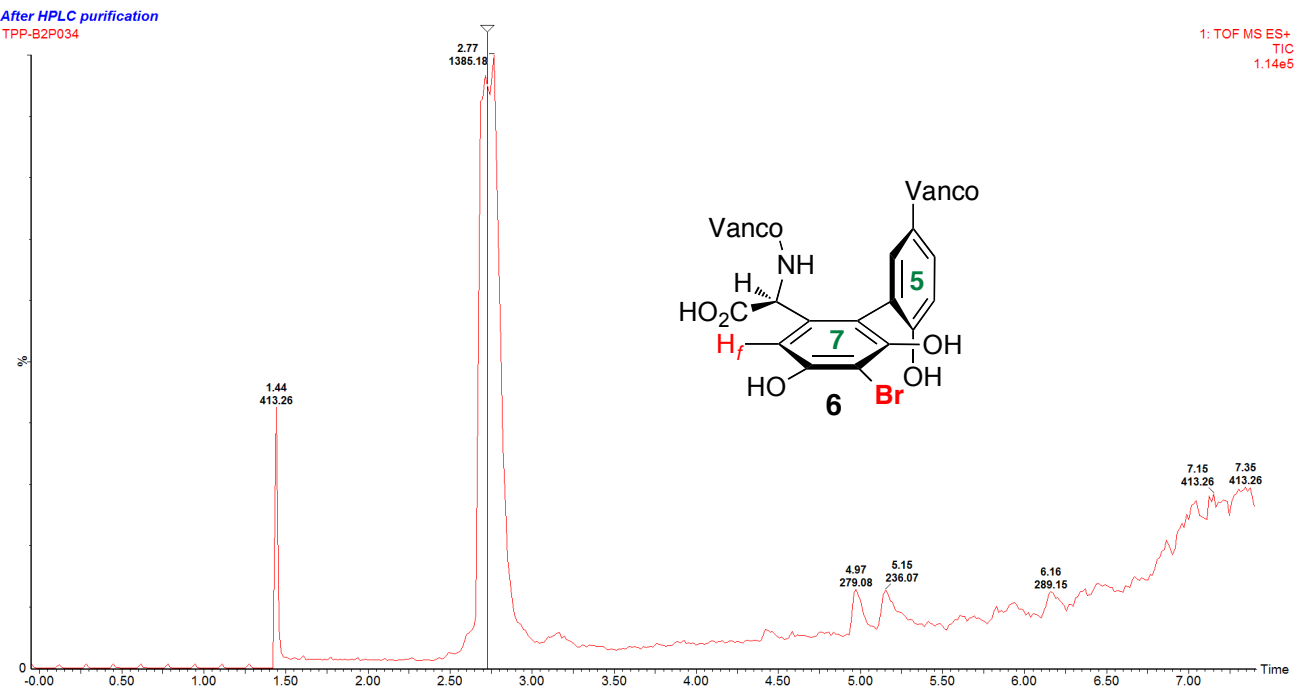


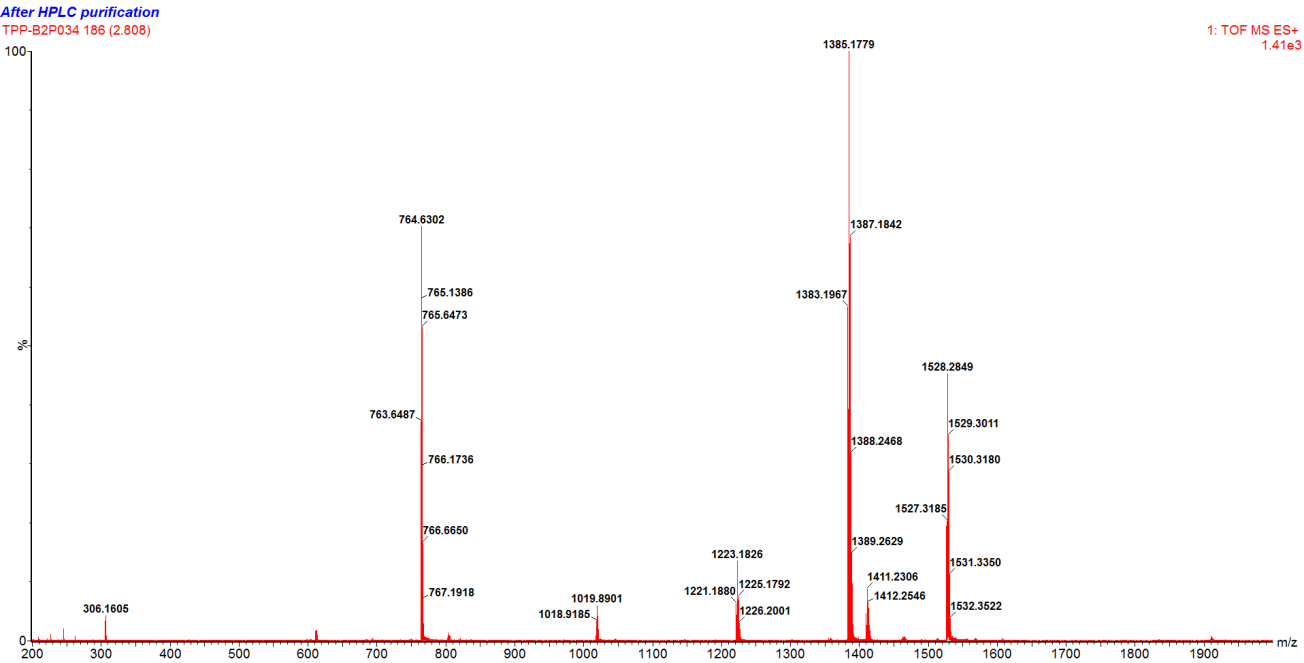
In lieu of tabulated NMR data, please see raw spectral data provided below.


Vanco
Vanco
NH
H
HO2C
Hf
OH
OH
HO
H
Vanco
Vanco
NH
H
HO2C
Hf
5
7
OH
OH
HO
Br
6
Vanco
Vanco
NH
H
HO2C
Br
5
7
OH
OH
HO
Hd
5
ST
14
13
12
11
10
9
8
7
6
5
4
3
2
1
0
-1
f1 (ppm)
3
2
1

D_2O, ^1H NMR
7_d-Br **(6)**




After HPLC purification
TPP-B2P034
1: TOF MS ES+
TIC
1.14e5
2.77
1385.18
1.44
413.26
4.97
279.08
5.15
236.07
6.16
289.15
7.15
413.26
7.35
413.26
Vanco
Vanco
NH
H
HO2C
Hf
HO
7
5
OH
OH
Br
6
%
Time


After HPLC purification
TPP-B2P034 186 (2.808)
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1385.1779
764.6302
1387.1842
765.1386
765.6473
1383.1967
763.6487
1528.2849
1529.3011
1388.2468
766.1736
1530.3180
1527.3185
766.6650
1389.2629
1223.1826
1531.3350
1411.2306
1221.1880
1225.1792
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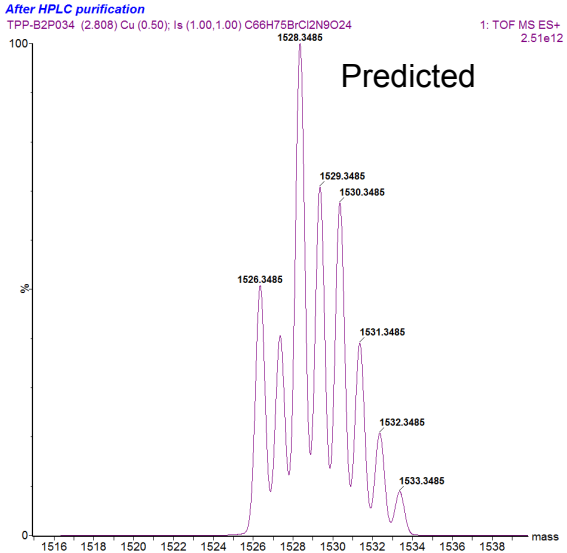
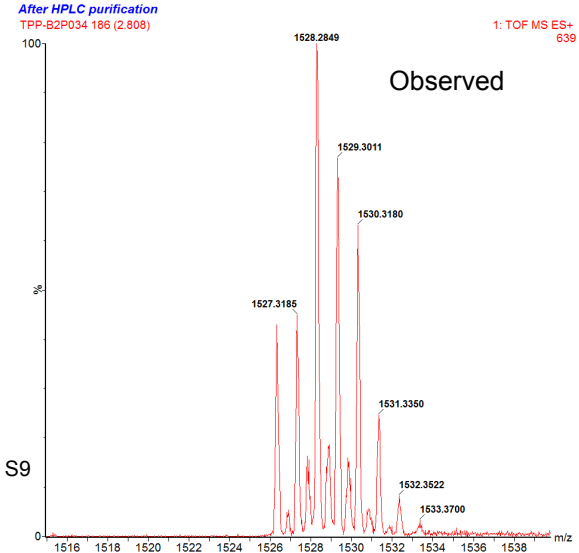
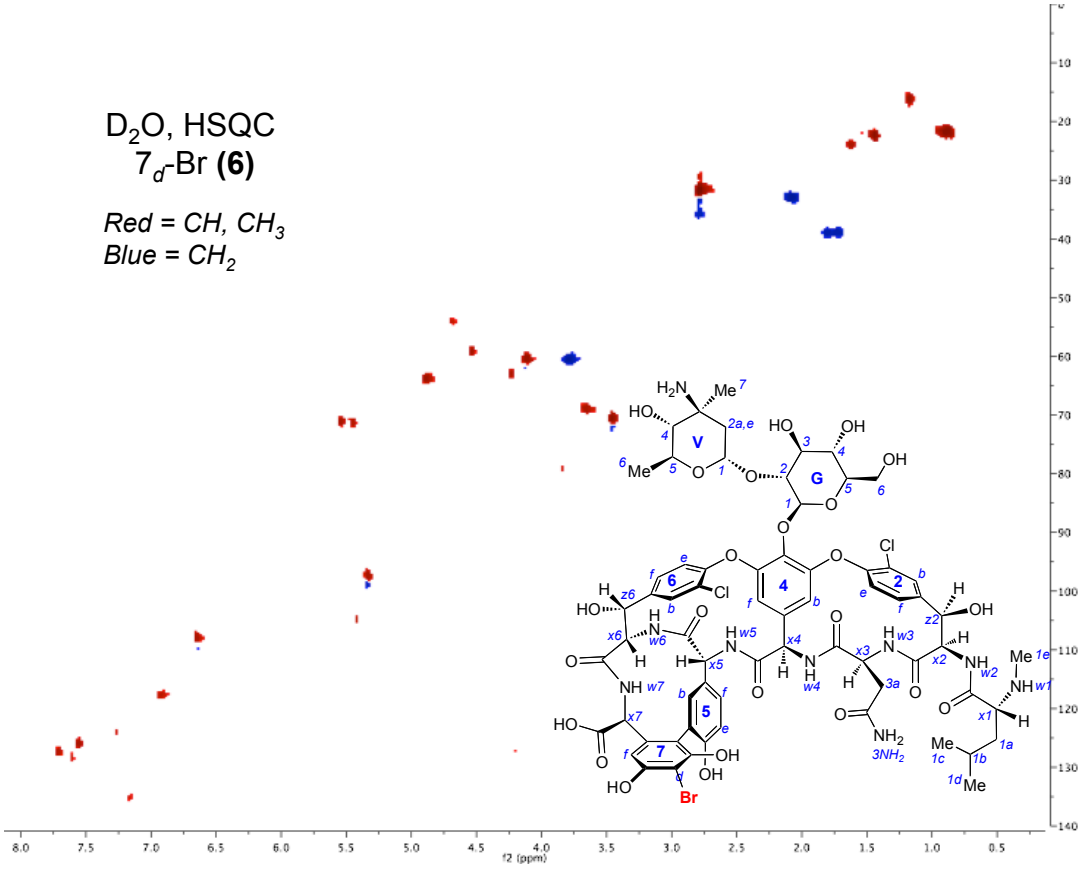
767.1918
1226.2001
1532.3522
1019.8901
1018.9185
306.1605
%
m/z


After HPLC purification
TPP-B2P034 (2.808) Cu (0.50); Is (1.00,1.00) C66H75BrCl2N9O24
1: TOF MS ES+
2.51e12
Predicted
1528.3485
1529.3485
1530.3485
1526.3485
1531.3485
1532.3485
1533.3485
%
mass


After HPLC purification
TPP-B2P034 186 (2.808)
1: TOF MS ES+
639
Observed
1528.2849
1529.3011
1530.3180
1527.3185
1531.3350
1532.3522
1533.3700
%
m/z

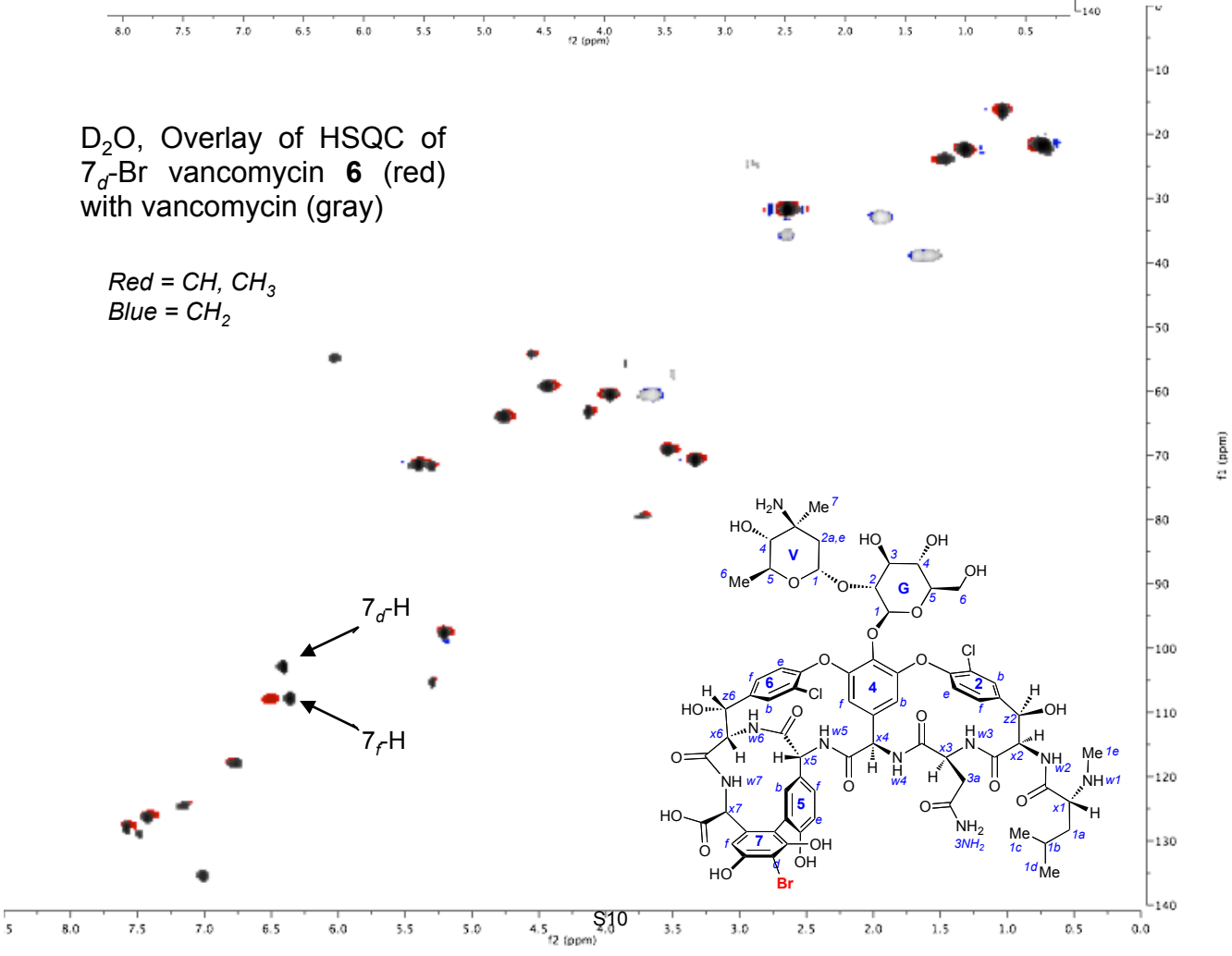
D_2O, HSQC
7_d-Br **(6)**

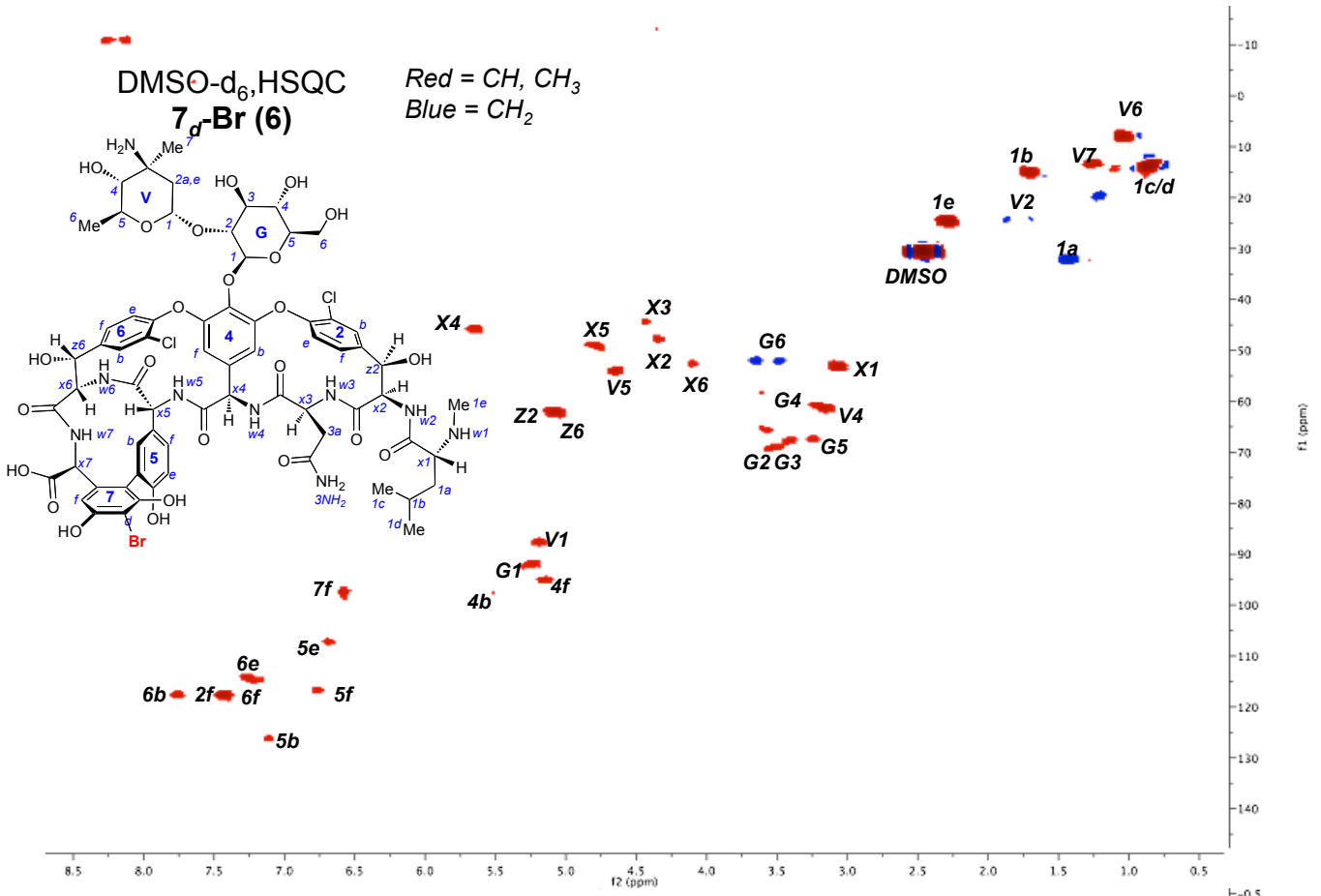
Red = CH, CH_3
Blue = CH_2

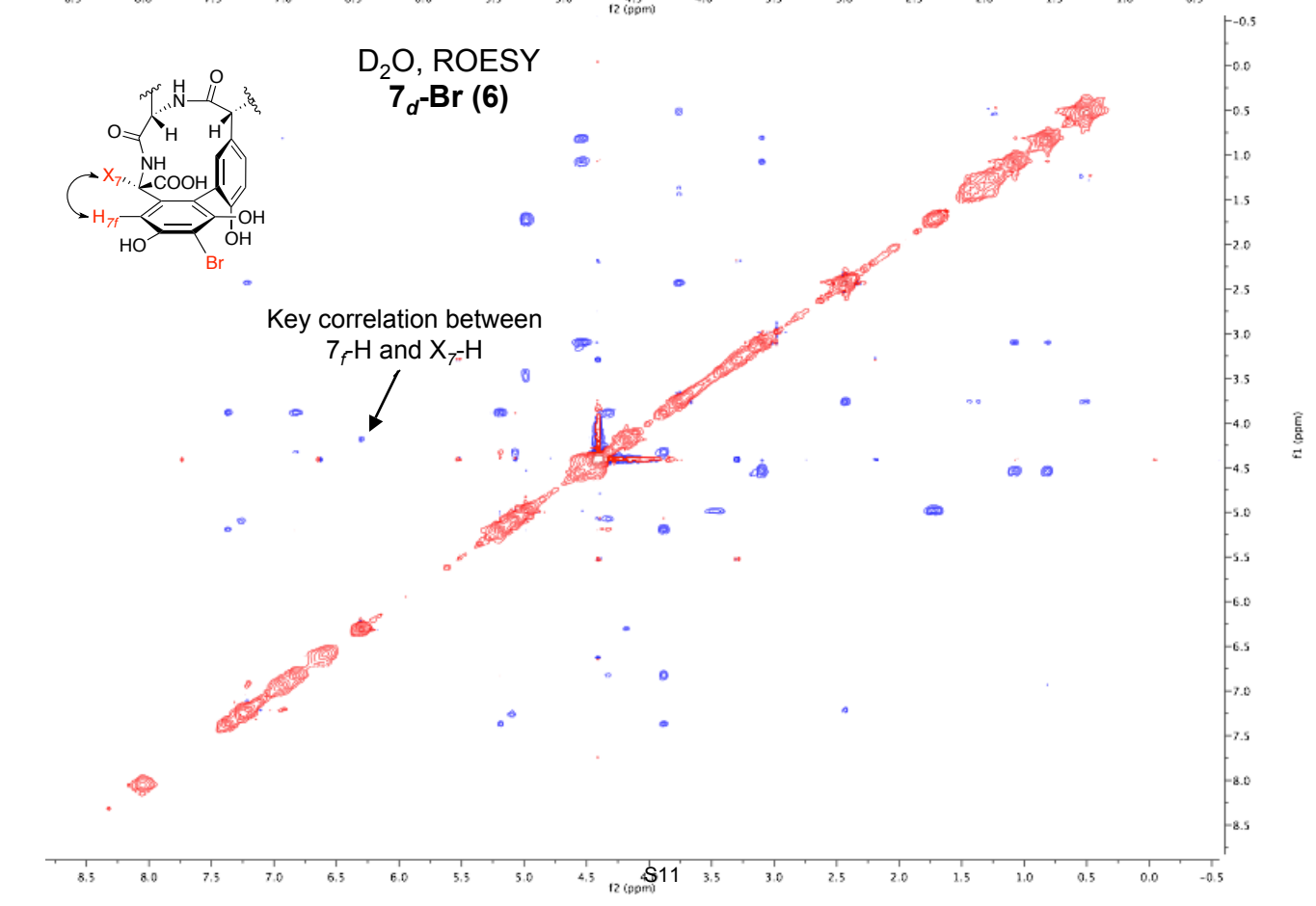


D_2O, Overlay of HSQC of 7_d-Br vancomycin **6** (red) with vancomycin (gray)

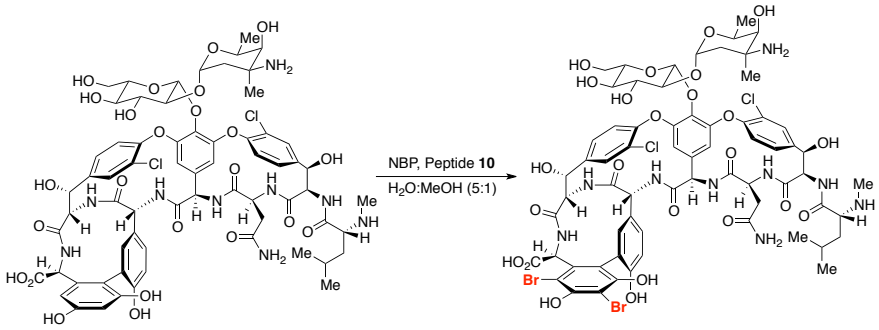
Red = CH, CH_3
Blue = CH_2




DMSO-d6,HSQC
7d-Br (6)
Red = CH, CH3
Blue = CH2
V6
1b
V7
1c/d
1e
V2
1a
DMSO
X4
X5
X3
G6
X1
X2
X6
V5
G4
V4
Z2
Z6
G5
G2 G3
V1
G1
4f
4b
7f
5e
6e
6b
2f
6f
5f
5b
f2 (ppm)
f1 (ppm)


D2O, ROESY
7d-Br (6)
X7'
H7f
Key correlation between
7f-H and X7-H
f2 (ppm)
f1 (ppm)

Procedure for Dibromination of Vancomycin (4) Targeting Analog 8 (Manuscript Figure 6):

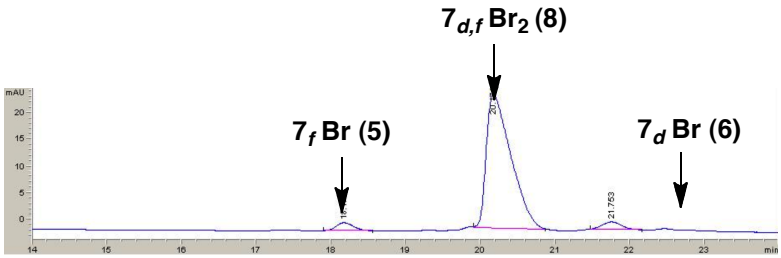


To a round bottom flask equipped with a stir bar were added 100.0 mg (0.067 mmol, 1.0 equiv) of vancomycin•HCl and 26.9 mg (0.067 mmol, 1.0 equiv) of peptide **10**. Water (2.0 mL) and MeOH (0.4 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 45.8 mg (0.201 mmol, 3.0 equiv) of NBP were added. The reaction mixture was stir at room temperature for *ca.* 60 min (over the period of time the reaction turn cloudy). To the reaction vessel, 10 mL of EtOAc was added and the reaction was stirred for an additional 15 min. The organic layer was discarded and the aqueous layer was diluted with 2 mL of water. The reaction was analyzed by reverse phase HPLC and was purified by preparative reverse phase HPLC. Purification of reaction mixture yielded 60.9 mg (55% Yield) $7_{d,f}$ –Br_2 vancomycin (**8**). ES-HRMS ($M+H^+$): Predicted 1604.2590, observed 1604.2281.

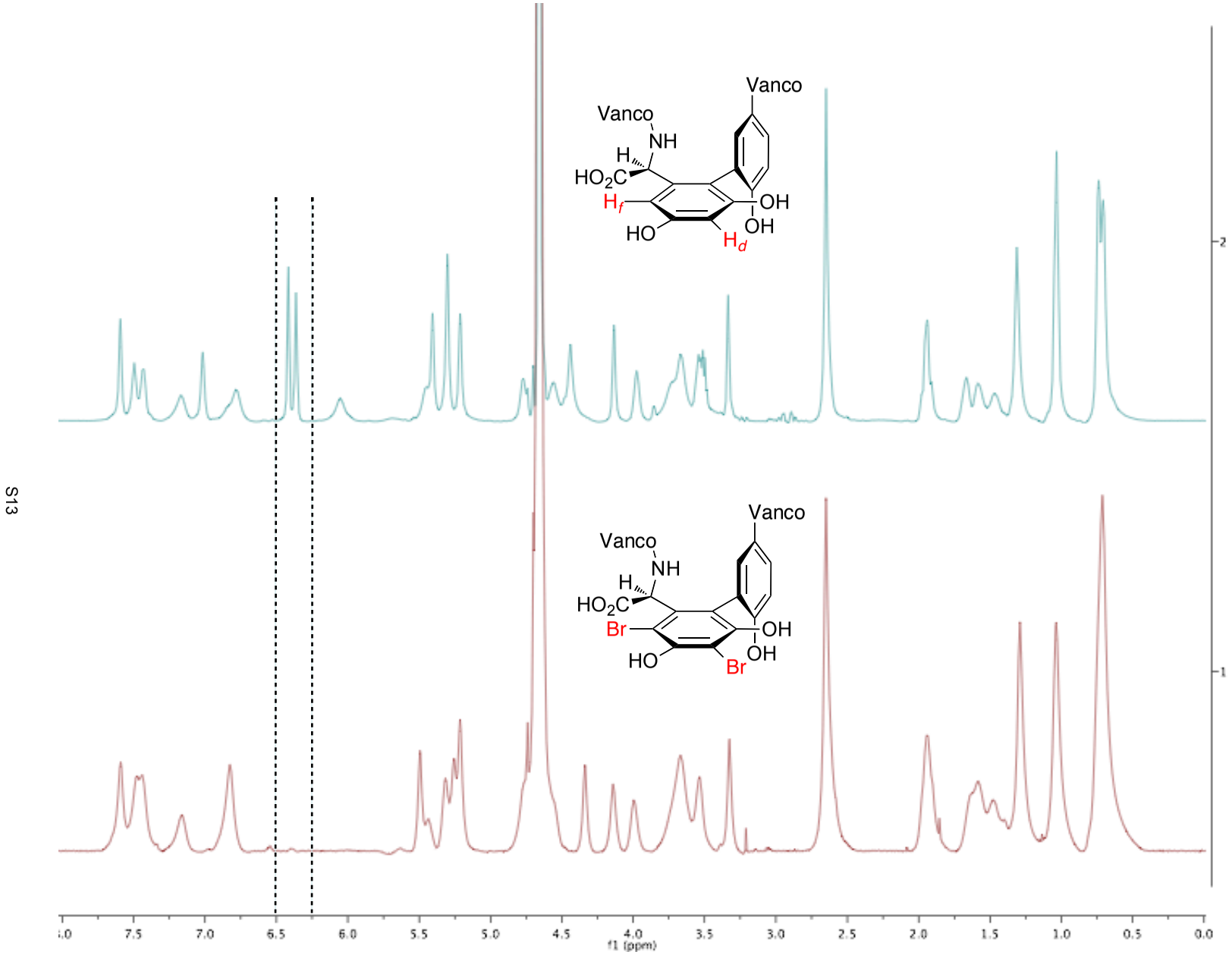
HPLC Method for preparative purification: Symmetry prep C8 7μm (19 X 300 mm) column, 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

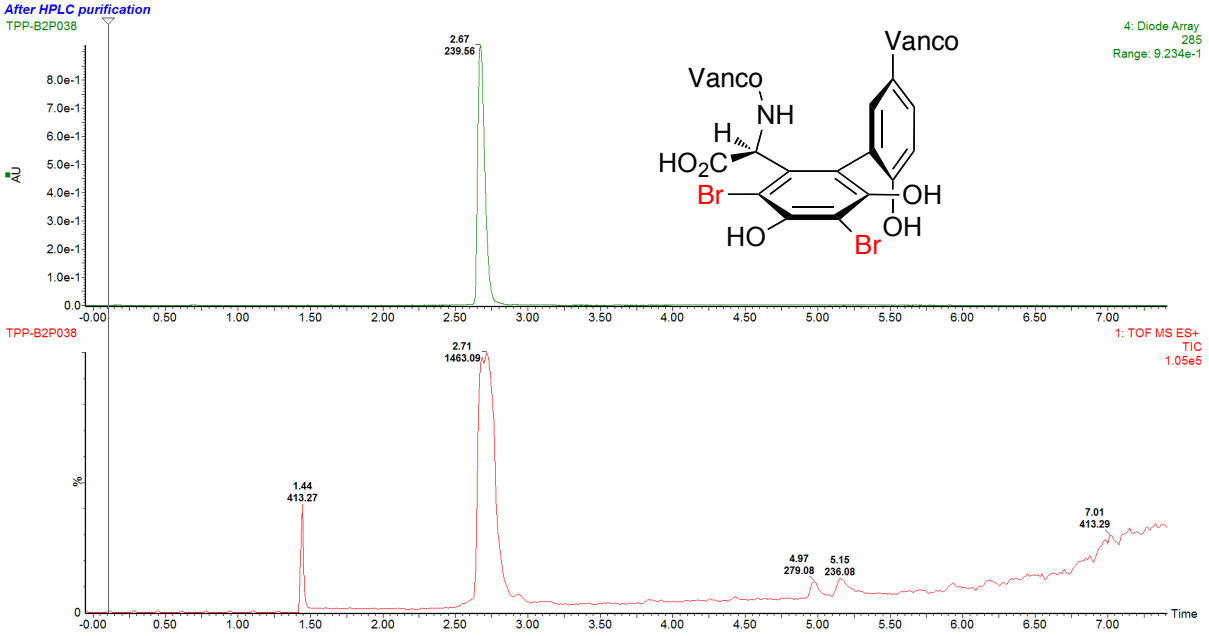
Analytical method: Symmetry prep C8 7μm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min. *Retention time of **8** on C18 column is 20.1 min and retention time on C8 column is 13.3 min.*

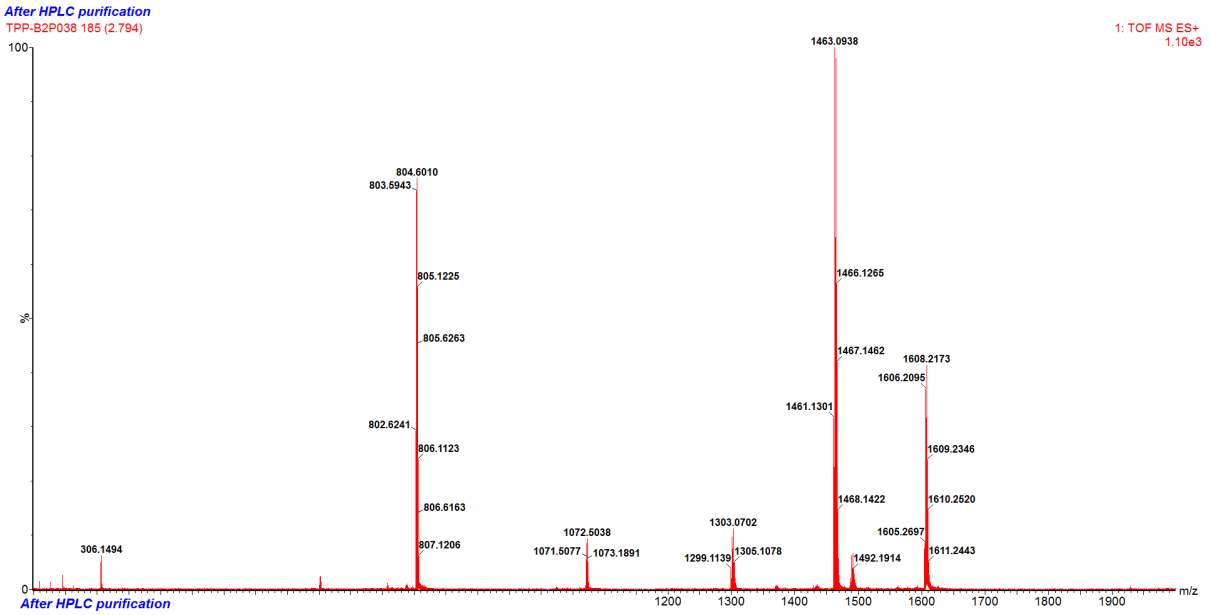
HPLC trace for the reaction mixture (at 280 nm):

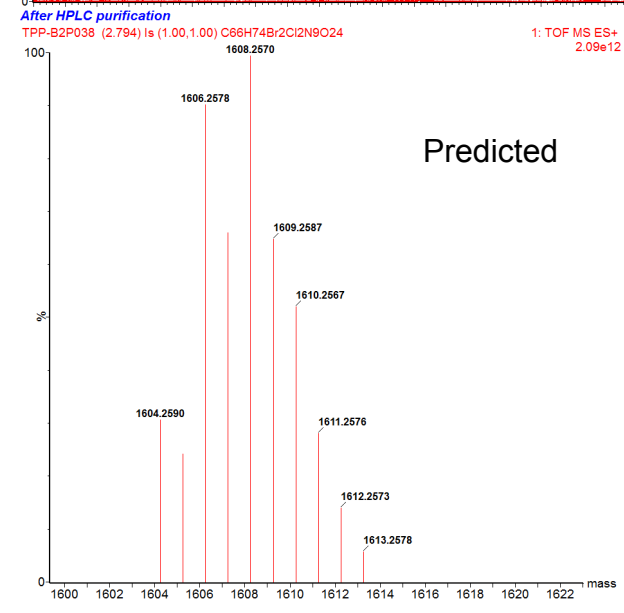


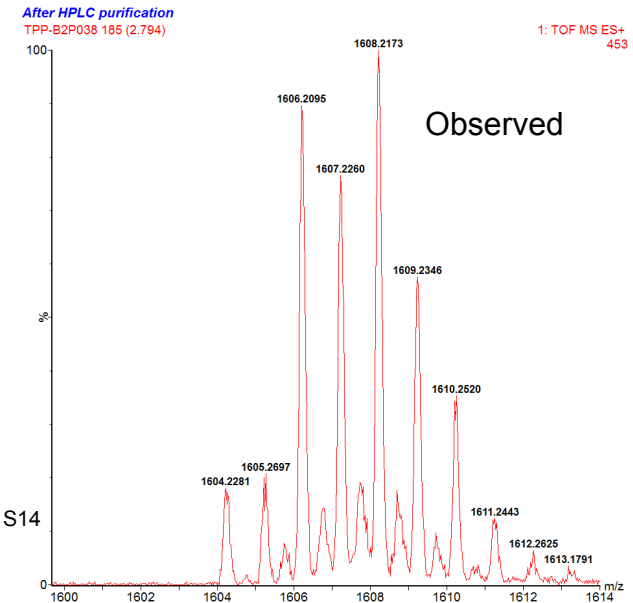
In lieu of tabulated NMR data, please see raw spectral data provided below.


Vanco
Vanco
NH
H
HO2C
Hf
OH
OH
HO
Hd
Vanco
Vanco
NH
H
HO2C
Br
OH
OH
HO
Br
2
1
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


After HPLC purification
TPP-B2P038
4: Diode Array
285
Range: 9.234e-1
2.67
239.56
AU
Vanco
Vanco
NH
H
HO2C
Br
OH
HO
OH
Br
TPP-B2P038
1: TOF MS ES+
TIC
1.05e5
2.71
1463.09
1.44
413.27
7.01
413.29
4.97
279.08
5.15
236.08
%
Time

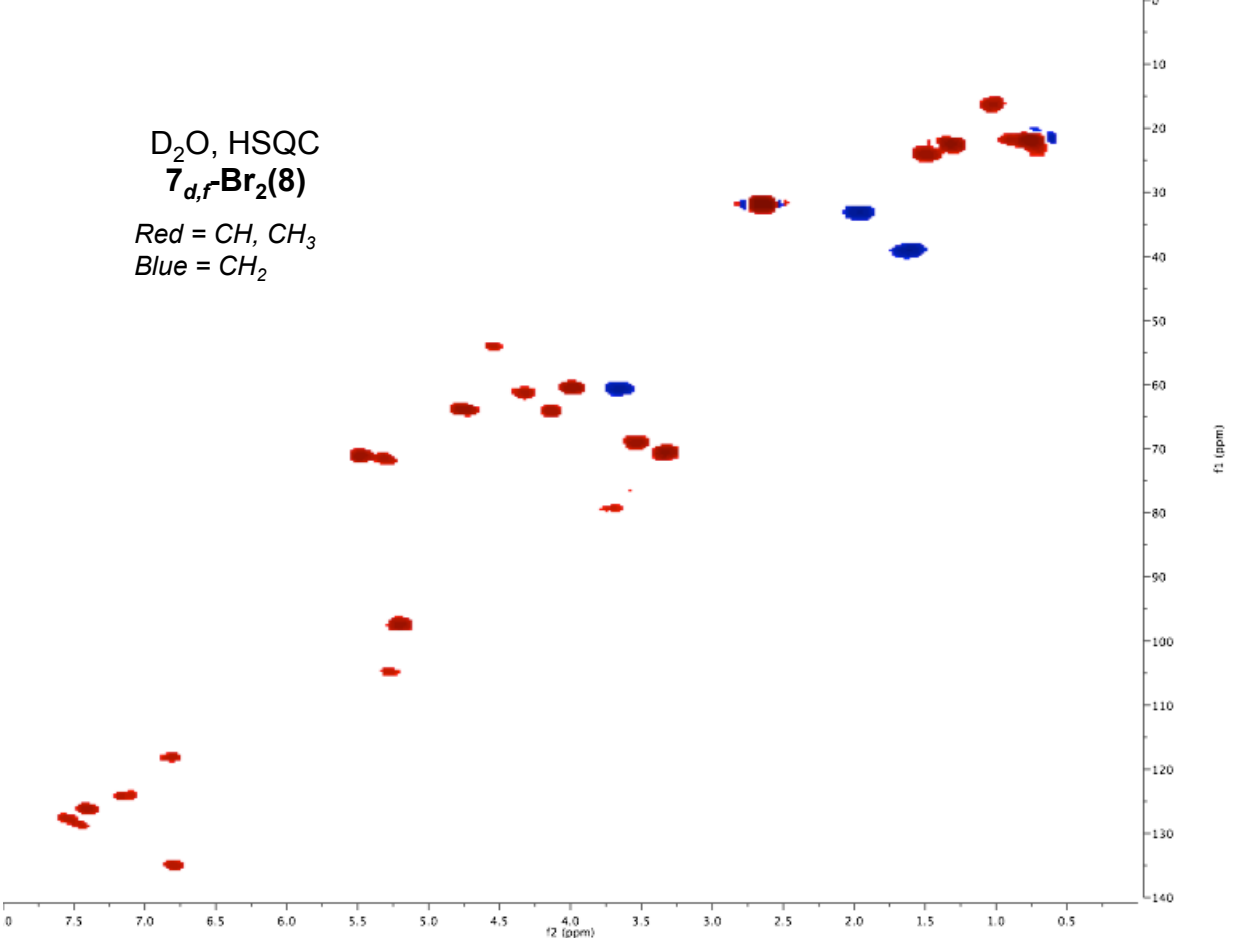

After HPLC purification
TPP-B2P038 185 (2.794)
1: TOF MS ES+
1.10e3
1463.0938
804.6010
803.5943
805.1225
805.6263
802.6241
806.1123
806.6163
807.1206
306.1494
1072.5038
1071.5077
1073.1891
1303.0702
1299.1139
1305.1078
1466.1265
1467.1462
1608.2173
1606.2095
1461.1301
1609.2346
1468.1422
1610.2520
1605.2697
1492.1914
1611.2443
%
m/z


After HPLC purification
TPP-B2P038 (2.794) Is (1.00,1.00) C66H74Br2Cl2N9O24
1: TOF MS ES+
2.09e12
Predicted
1608.2570
1606.2578
1609.2587
1610.2567
1604.2590
1611.2576
1612.2573
1613.2578
%
mass


After HPLC purification
TPP-B2P038 185 (2.794)
1: TOF MS ES+
453
Observed
1608.2173
1606.2095
1607.2260
1609.2346
1610.2520
1605.2697
1604.2281
1611.2443
1612.2625
1613.1791
%
m/z

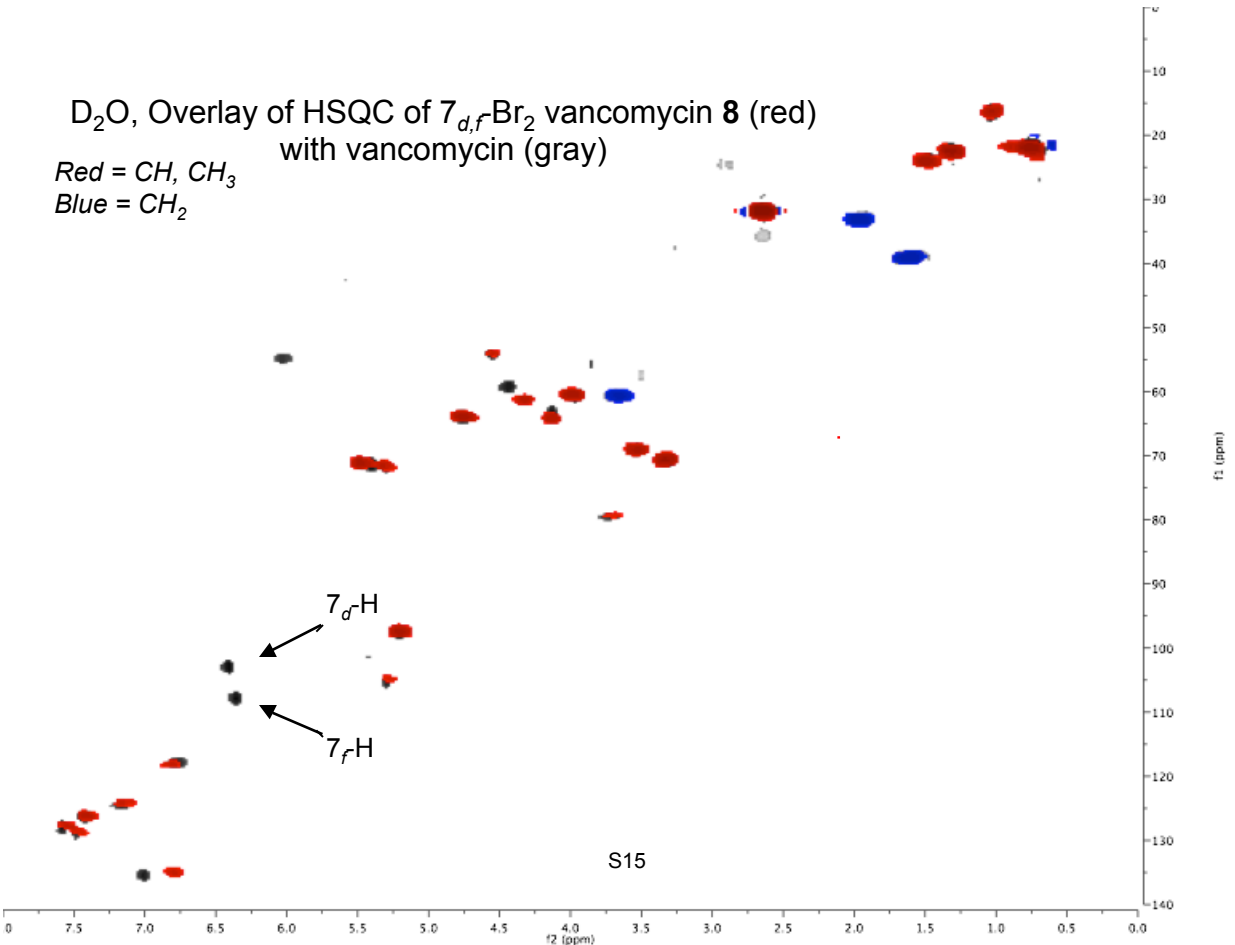
D_2O, HSQC
$7_{d,f}$-Br_2(8)

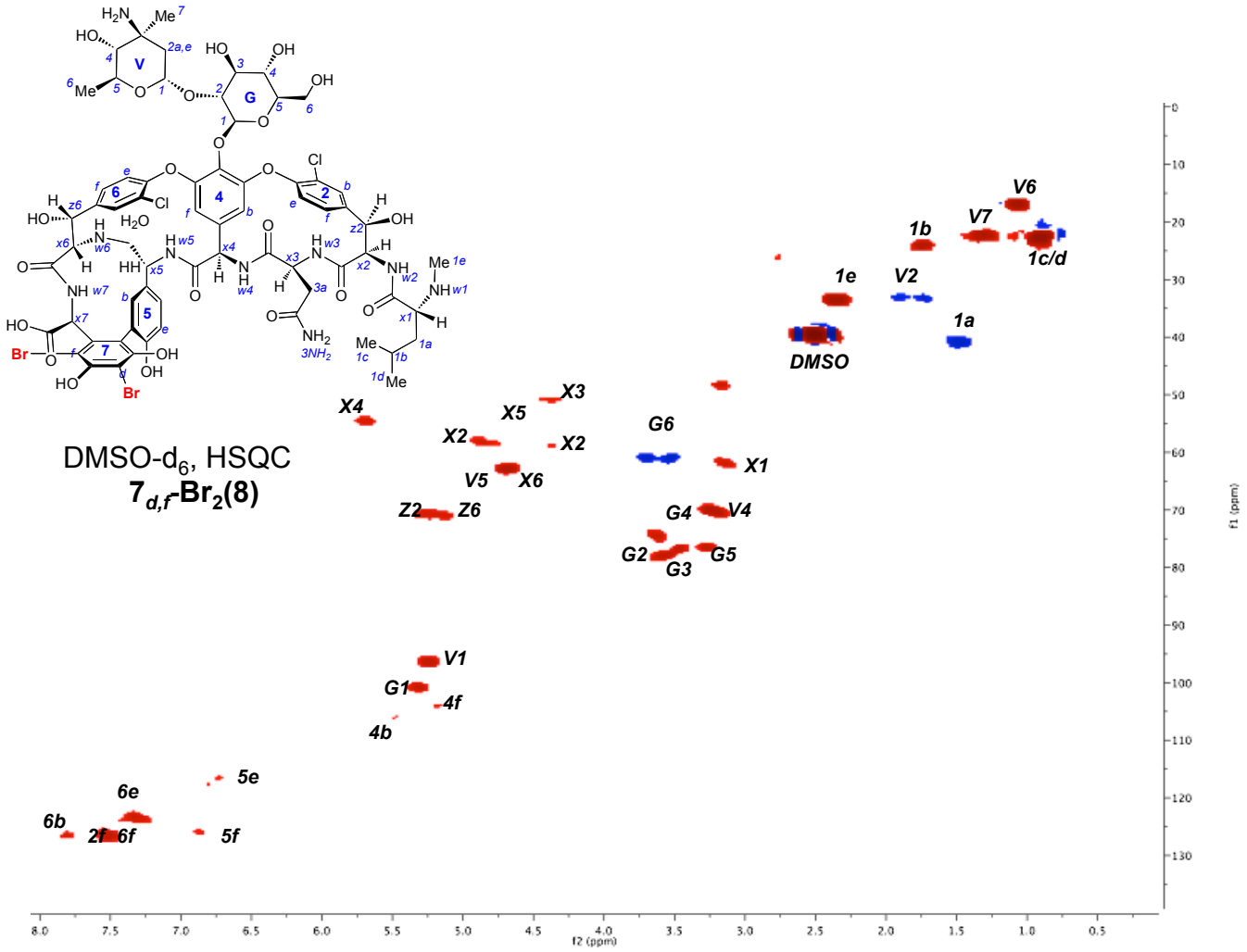
Red = CH, CH_3
Blue = CH_2



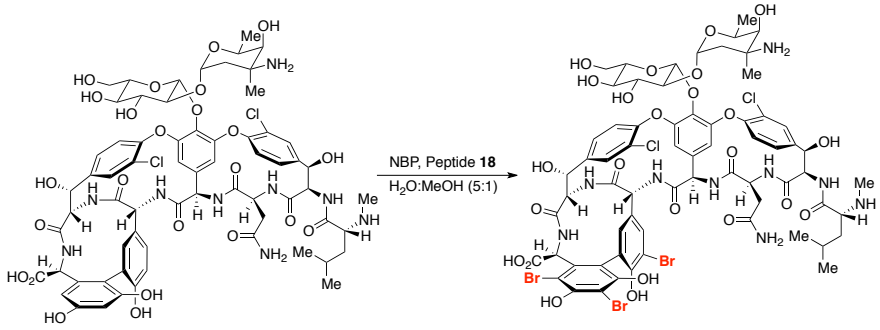
D_2O, Overlay of HSQC of $7_{d,f}$-Br_2 vancomycin **8** (red) with vancomycin (gray)

Red = CH, CH_3
Blue = CH_2




DMSO-d6, HSQC
7d,f-Br2(8)
V6
V7
1b
1c/d
1e
V2
1a
DMSO
X3
X4
X5
X2
X2
G6
X1
V5
X6
Z2
Z6
G4
V4
G2
G3
G5
V1
G1
4f
4b
5e
6e
6b
2f
6f
5f
f2 (ppm)
f1 (ppm)

Procedure for tribromination of Vancomycin (4) Targeting Analog 17 (Manuscript Figure 7):

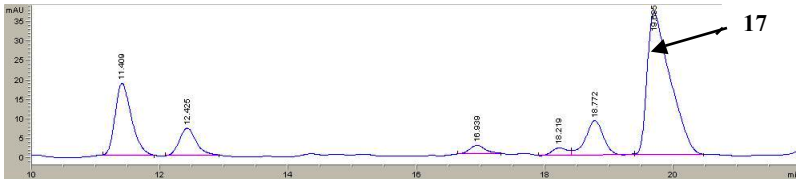


To a round bottom flask equipped with a stir bar were added 49.0 mg (0.033 mmol, 1.0 equiv) of vancomycin•HCl and 15.1 mg (0.033 mmol, 1.0 equiv) of peptide **18**. Water (1.0 mL) and MeOH (0.2 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 30.1 mg (0.132 mmol, 4.0 equiv) of NBP were added. The reaction mixture was stir at room temperature for *ca.* 120 min (over the period of time the reaction became cloudy). To the reaction vessel, 10 mL of EtOAc were added and the reaction was stirred for an additional 15 min. The organic layer was discarded and the aqueous layer was diluted with 2 mL of water. The reaction was analyzed by reverse phase HPLC and was purified by preparative reverse phase HPLC. Purification of reaction mixture yielded, 20.3 mg (35% Yield) tribrominated vancomycin (**17**). ES-HRMS ($M+H^+$): Predicted 1682.1695, observed 1682.1954.

HPLC Method for preparative purification: Symmetry prep C8 7µm column (19 X 300 mm), 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 15% MeCN over 20 min, 15 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

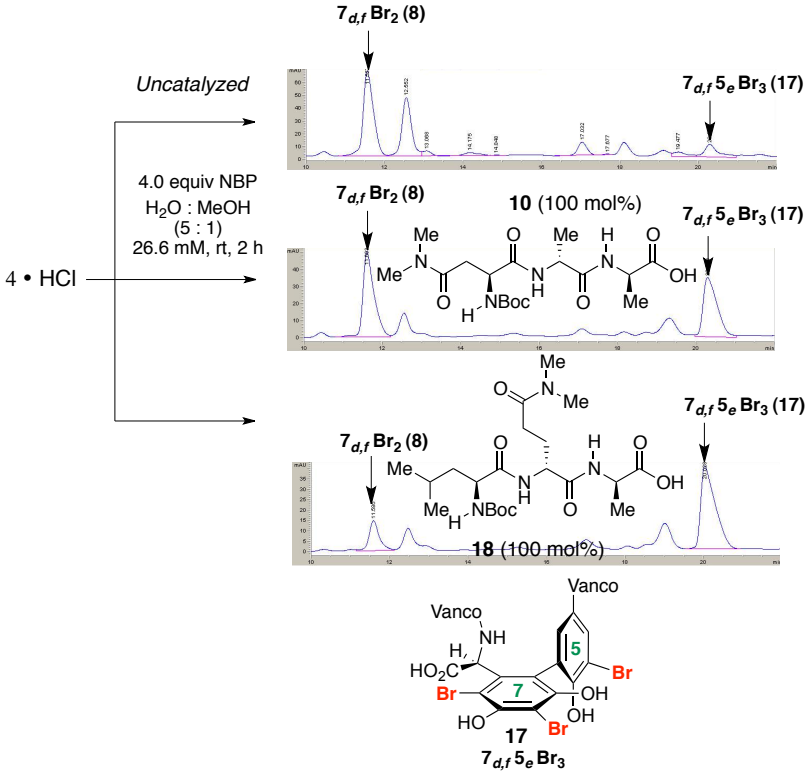
Analytical method: Symmetry prep C8 7µm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 15% MeCN over 20 min, 15 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min. Retention time of **17** on C8 column is 20.7 min.

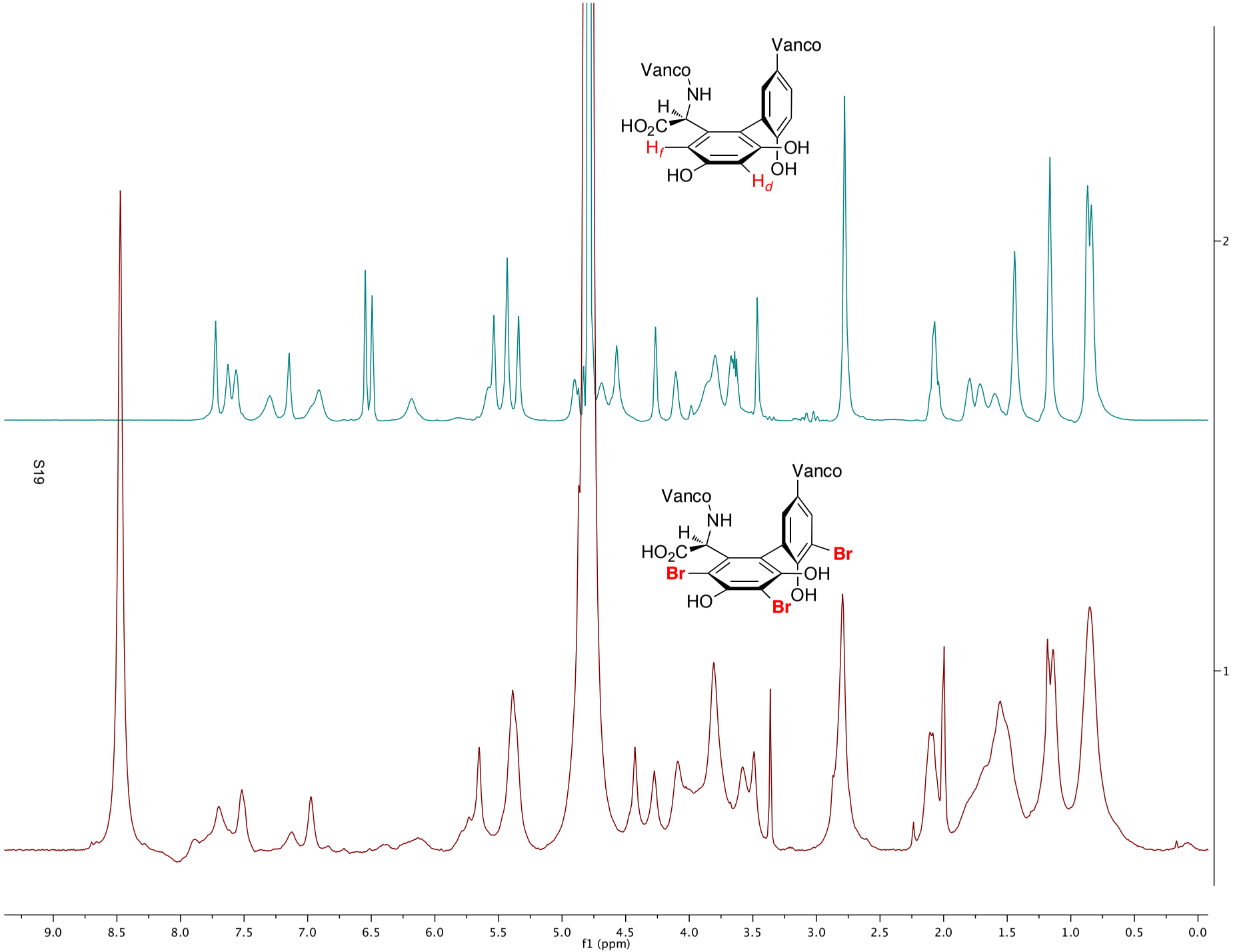
HPLC trace for the reaction mixture (at 280 nm)

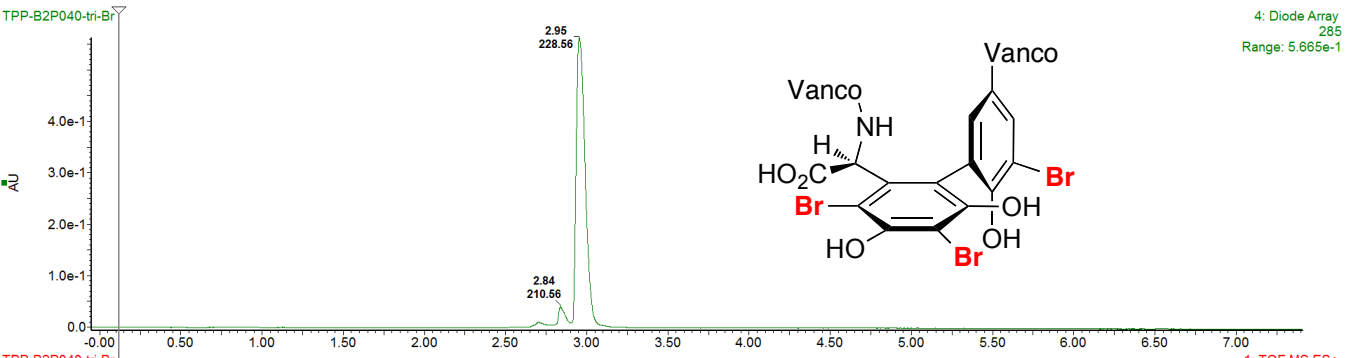


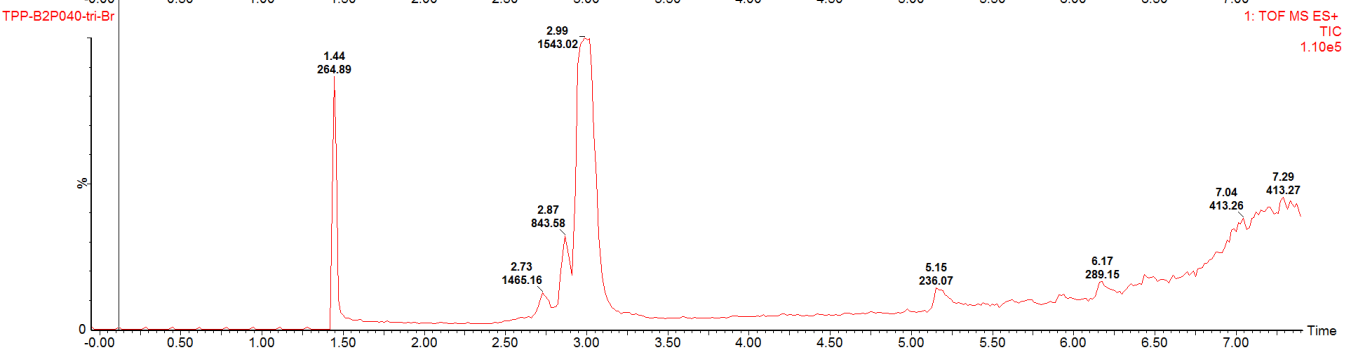
In lieu of tabulated NMR data, please see raw spectral data provided below.

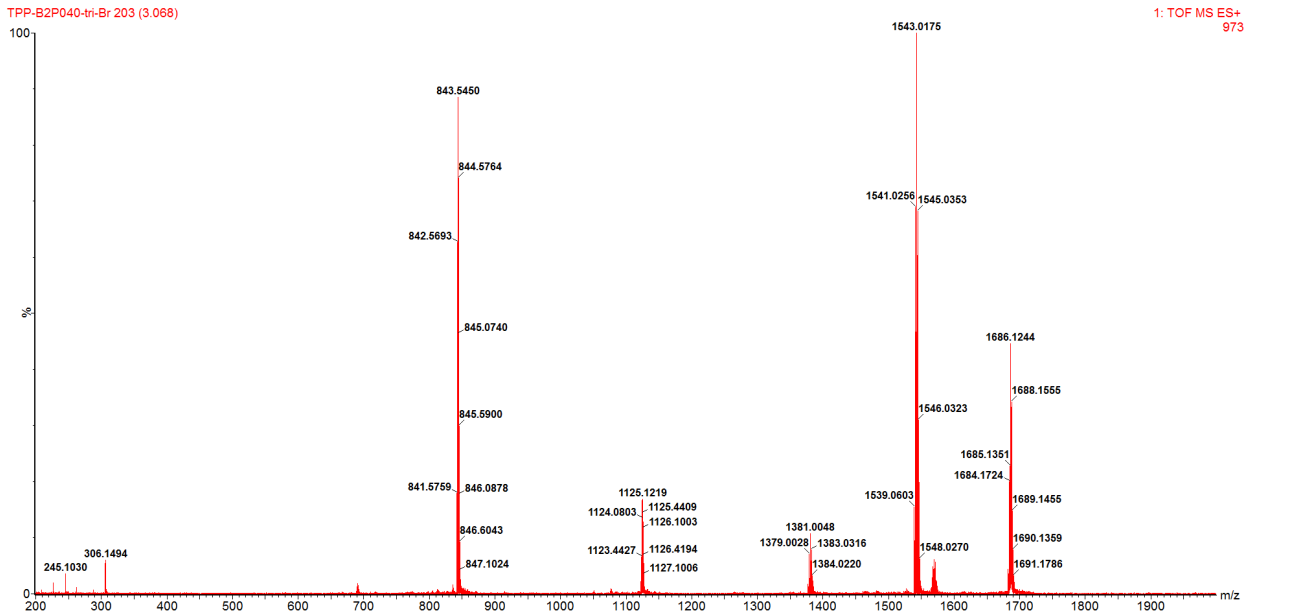
HPLC traces of uncatalyzed (top) and peptide-catalyzed (middle and bottom) tribromination of vancomycin obtained on reverse phase HPLC.

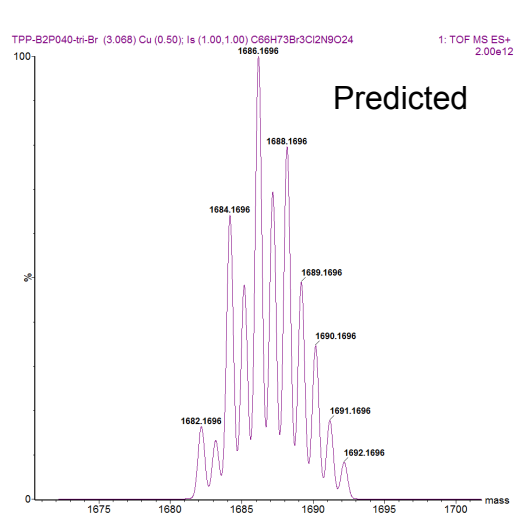


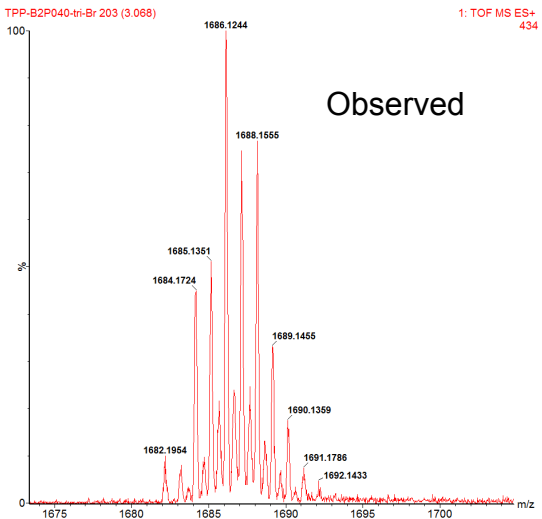

Vanco
Vanco
NH
H
HO2C
Hf
OH
HO
OH
Hd
Vanco
Vanco
NH
H
HO2C
Br
Br
OH
HO
OH
Br
2
1
S19
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


TPP-B2P040-tri-Br
4: Diode Array
285
Range: 5.665e-1
AU
2.95
228.56
2.84
210.56
Vanco
Vanco
NH
H
HO2C
Br
Br
OH
HO
OH
Br


TPP-B2P040-tri-Br
1: TOF MS ES+
TIC
1.10e5
%
1.44
264.89
2.99
1543.02
2.87
843.58
2.73
1465.16
5.15
236.07
6.17
289.15
7.04
413.26
7.29
413.27
Time

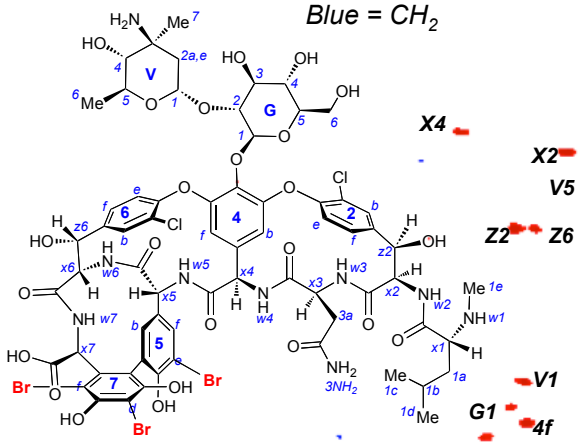

TPP-B2P040-tri-Br 203 (3.068)
1: TOF MS ES+
973
%
1543.0175
843.5450
844.5764
842.5693
1541.0256
1545.0353
845.0740
1686.1244
1688.1555
845.5900
1546.0323
1685.1351
1684.1724
841.5759
846.0878
1125.1219
1124.0803
1125.4409
1126.1003
1539.0603
1689.1455
846.6043
1381.0048
1379.0028
1383.0316
1690.1359
306.1494
245.1030
1123.4427
1126.4194
1548.0270
847.1024
1384.0220
1691.1786
1127.1006
m/z


TPP-B2P040-tri-Br (3.068) Cu (0.50); Is (1.00,1.00) C66H73Br3Cl2N9O24
1: TOF MS ES+
2.00e12
Predicted
1686.1696
1688.1696
1684.1696
1689.1696
1690.1696
1691.1696
1682.1696
1692.1696
%
mass


TPP-B2P040-tri-Br 203 (3.068)
1: TOF MS ES+
434
Observed
1686.1244
1688.1555
1685.1351
1684.1724
1689.1455
1690.1359
1682.1954
1691.1786
1692.1433
%
m/z

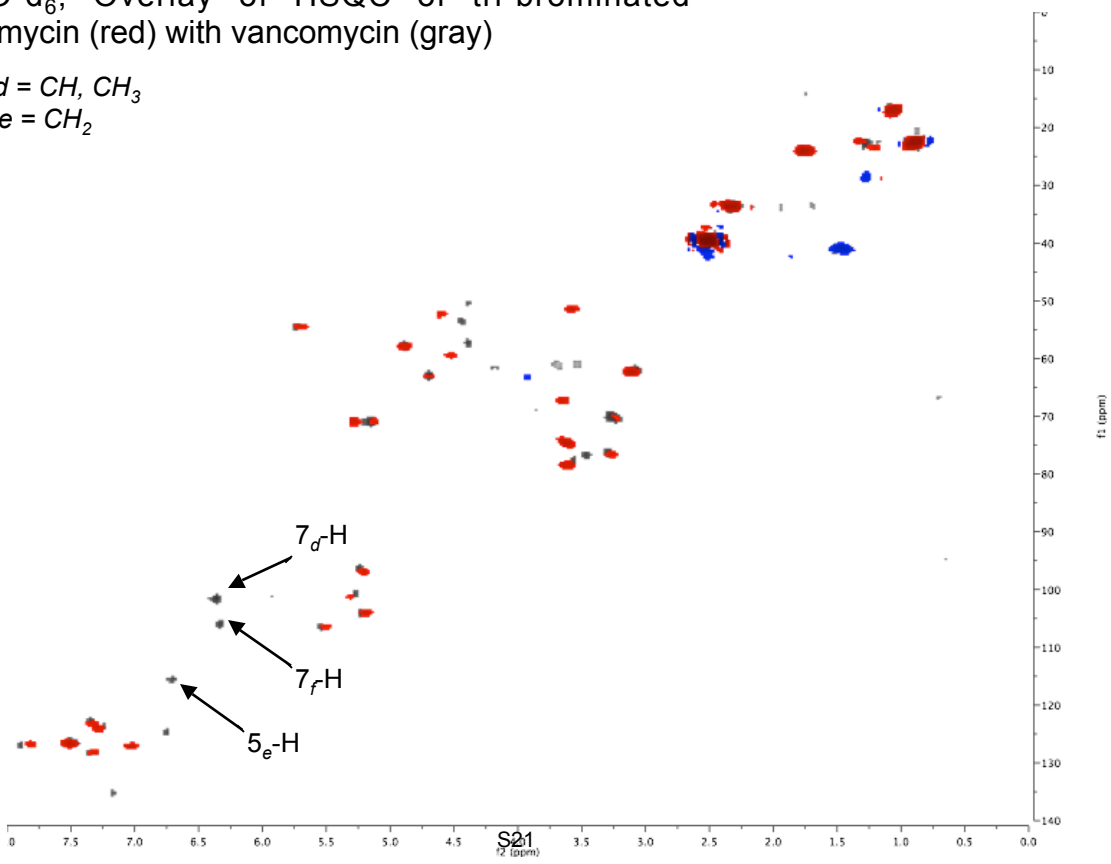
DMSO-d_6, HSQC

Red = CH, CH_3
Blue = CH_2

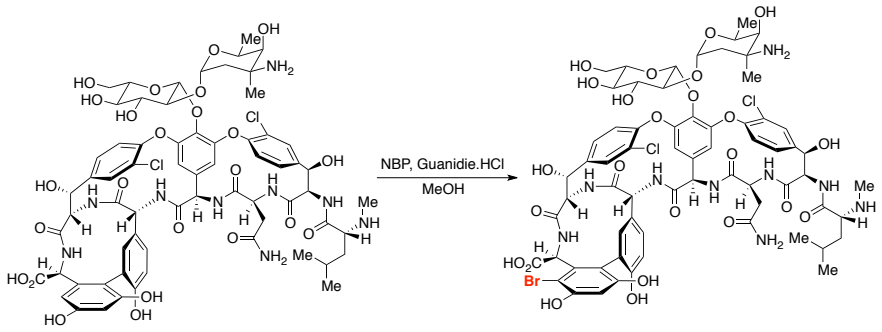


DMSO-d_6, Overlay of HSQC of tri-brominated vancomycin (red) with vancomycin (gray)

Red = CH, CH_3
Blue = CH_2



Procedure for Bromination of Vancomycin (4) Targeting Analog 5.

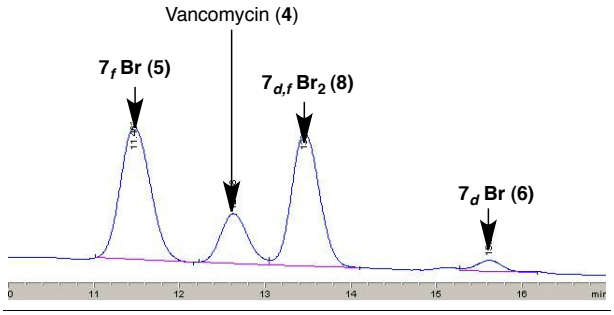


To a round bottom flask equipped with a stir bar were added 100.0 mg (0.067 mmol, 1.0 equiv) of vancomycin•HCl and 114.5 mg (1.206 mmol, 18.0 equiv) of peptide guanidine•HCl. MeOH (16.0 mL) was added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min after which 61.6 mg (0.27 mmol, 4.0 equiv) of NBP were added. The reaction mixture was stirred at room temperature for 4 h (solution remained cloudy through out the course of a reaction). To the reaction vessel, 10 mL of H_2O was added and reaction was stirred for an additional 15 min. The MeOH was removed in vacuo and reaction was analyzed by reverse phase HPLC and was purified by preparative reverse phase HPLC. Purification of reaction mixture yielded, 23.3 mg (21% Yield) 7_f-Br Vancomycin (**5**). ES-HRMS ($M+H^+$): Predicted 1526.3485, observed 1526.3184.

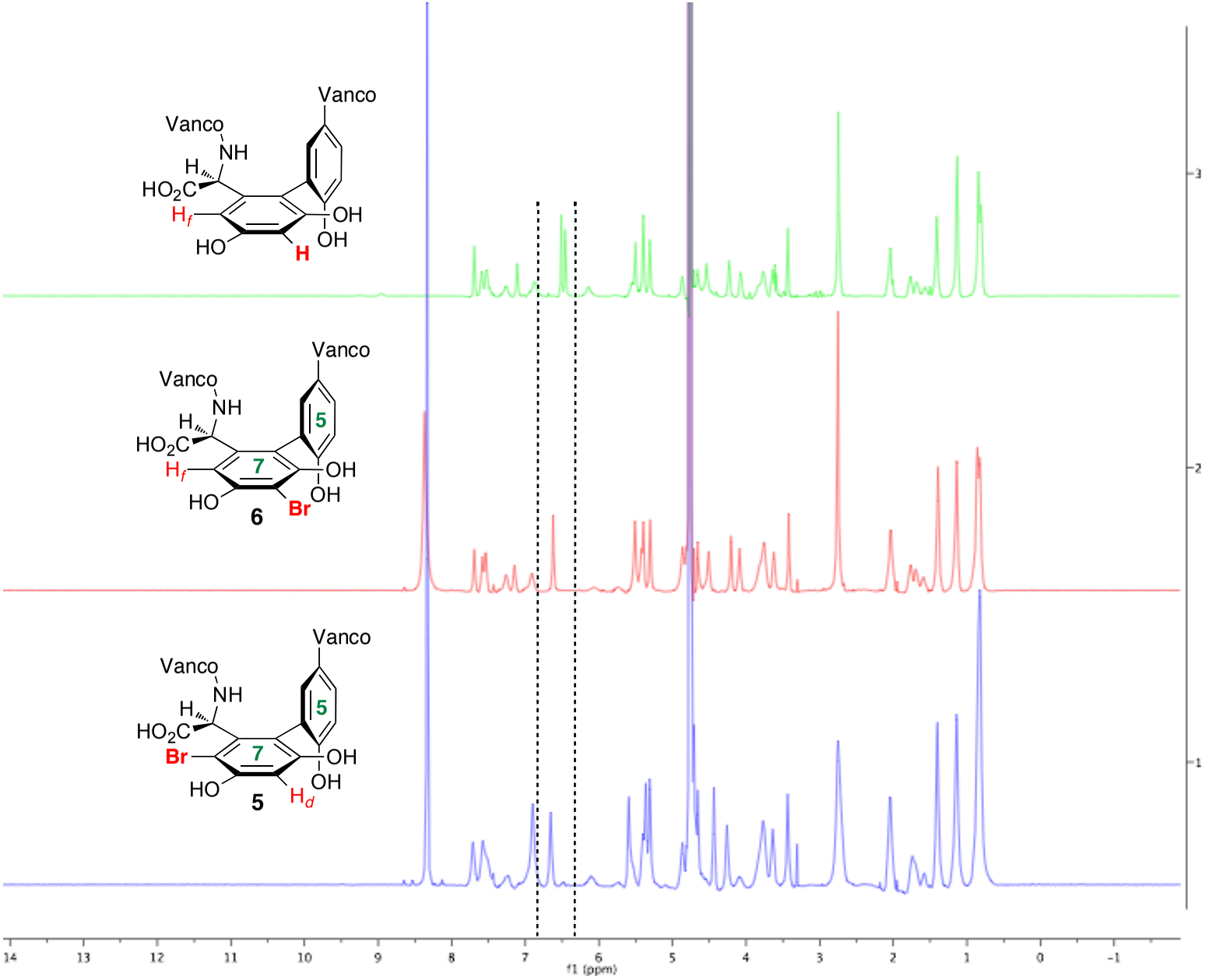
HPLC Method for preparative purification: Symmetry prep C8 7μm (19 X 300 mm) column, 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

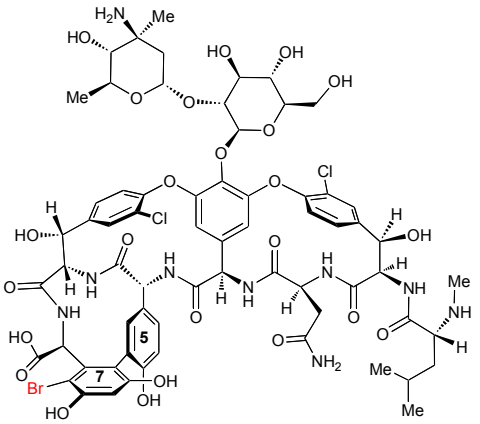
Analytical method: Symmetry prep C8 7μm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 95% MeCN over 3 min, hold at 95% MeCN for 3 min, 95% to 5% over 3 min. Retention time of **5** on C8 column is 11.6 min.

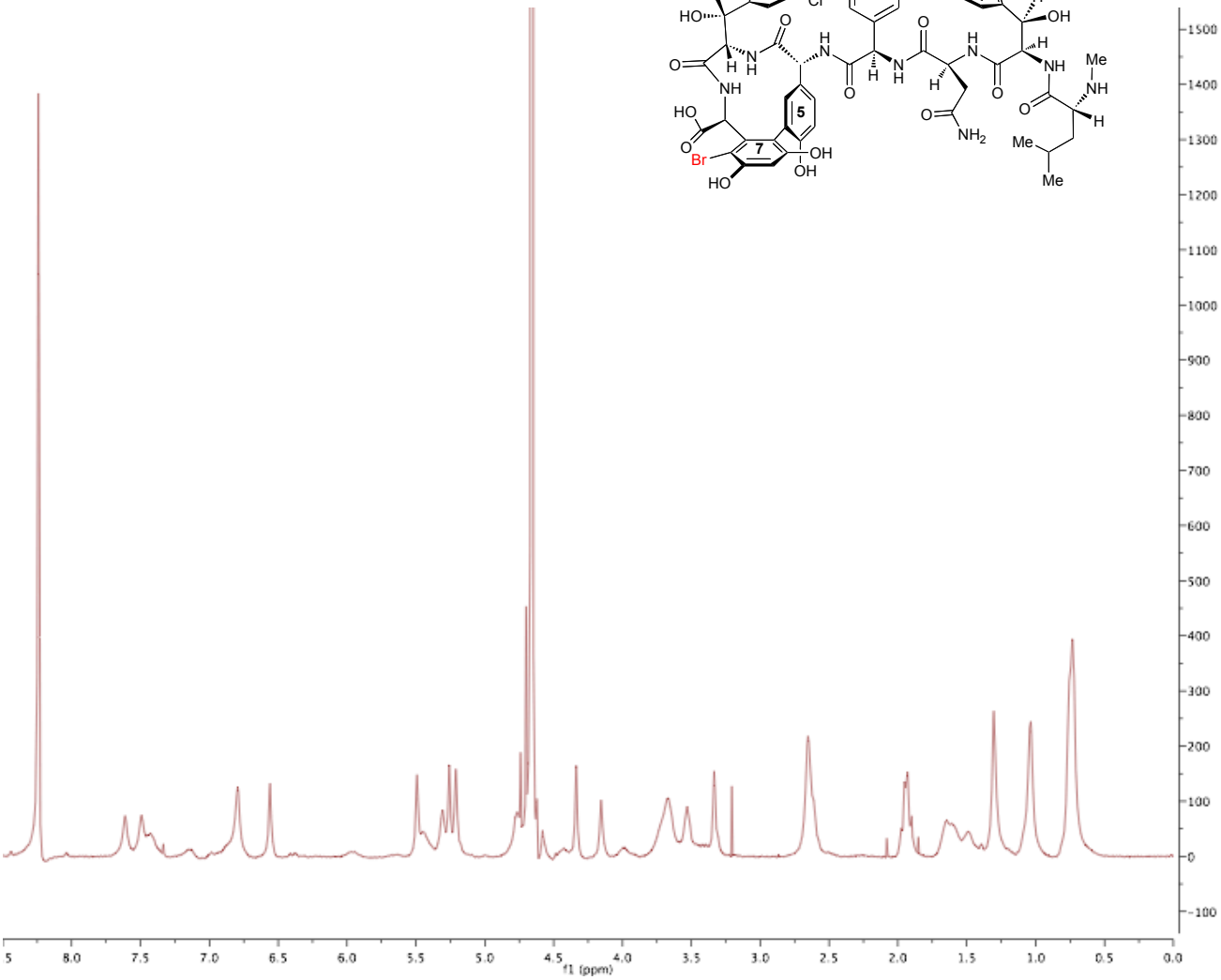
HPLC trace for the reaction mixture ***with guanidine*** (at 280 nm):

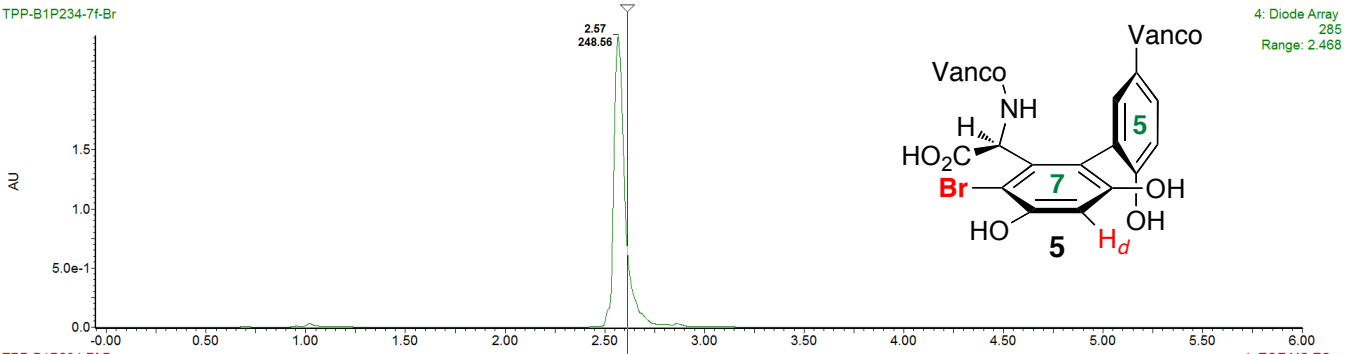


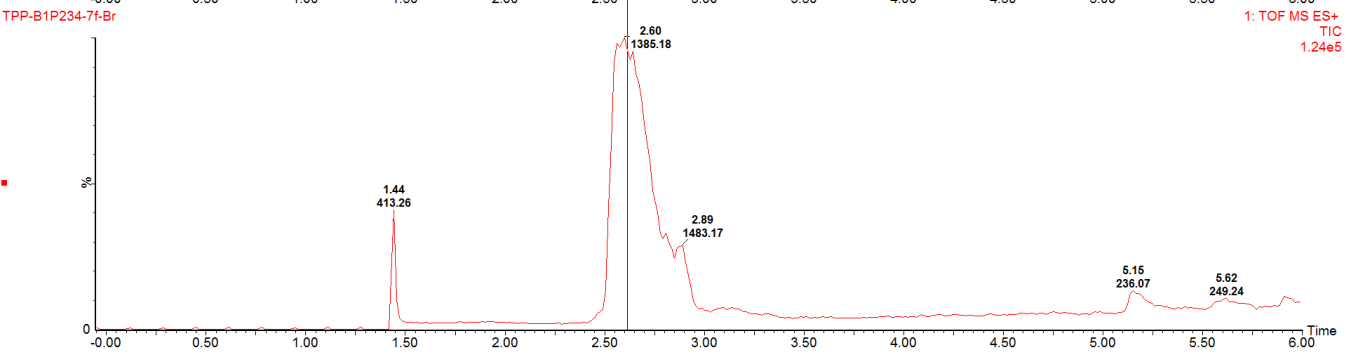
In lieu of tabulated NMR data, please see raw spectral data provided below.

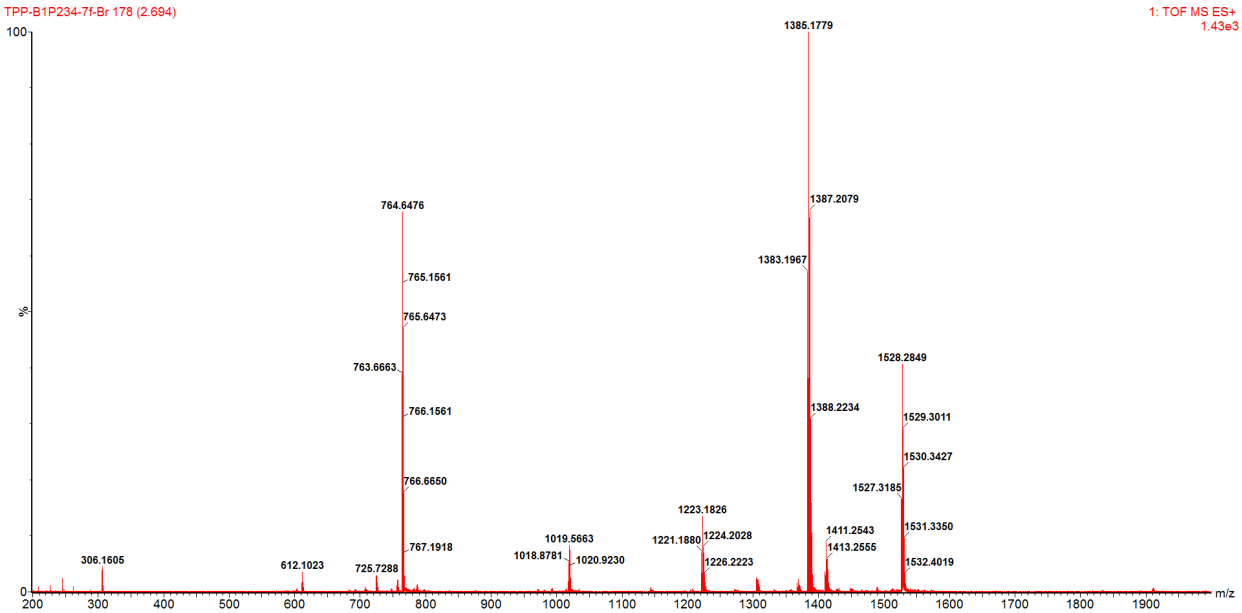

Vanco
Vanco
NH
H
HO2C
Hf
HO
OH
OH
H
Vanco
Vanco
NH
H
HO2C
Hf
5
7
HO
OH
OH
Br
6
Vanco
Vanco
NH
H
HO2C
Br
5
7
HO
OH
OH
Hd
5
f1 (ppm)

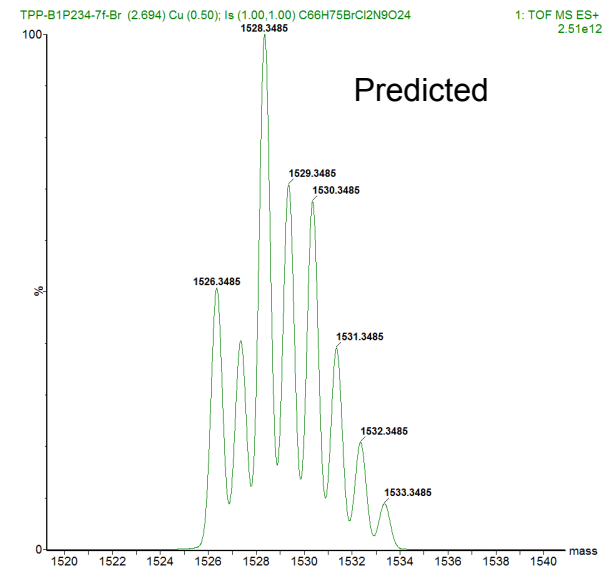

H2N
Me
HO
Me
O
HO
OH
OH
O
O
O
O
Cl
Cl
H
HO
H
N
H
O
NH
HO
O
Br
5
7
OH
OH
HO
H
N
H
O
H
N
H
O
H
N
H
O
NH2
H
OH
NH
O
Me
NH
H
Me
Me

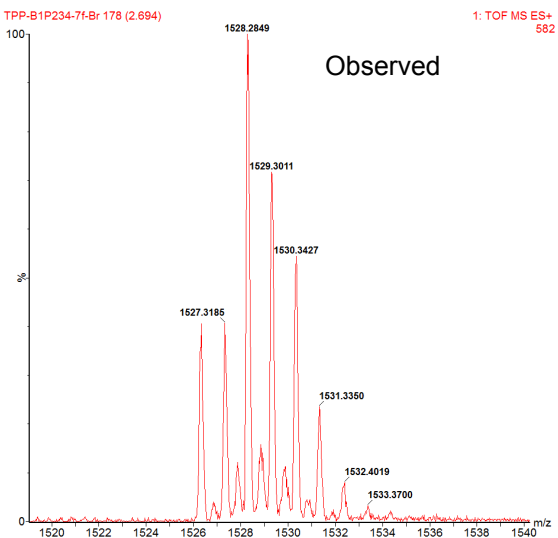

1500
1400
1300
1200
1100
1000
900
800
700
600
500
400
300
200
100
0
-100
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)


TPP-B1P234-7f-Br
4: Diode Array
285
Range: 2.468
2.57
248.56
AU
Vanco
NH
HO_2C
Br
HO
OH
OH
H_d
5
7
5


TPP-B1P234-7f-Br
1: TOF MS ES+
TIC
1.24e5
2.60
1385.18
1.44
413.26
2.89
1483.17
5.15
236.07
5.62
249.24
%
Time

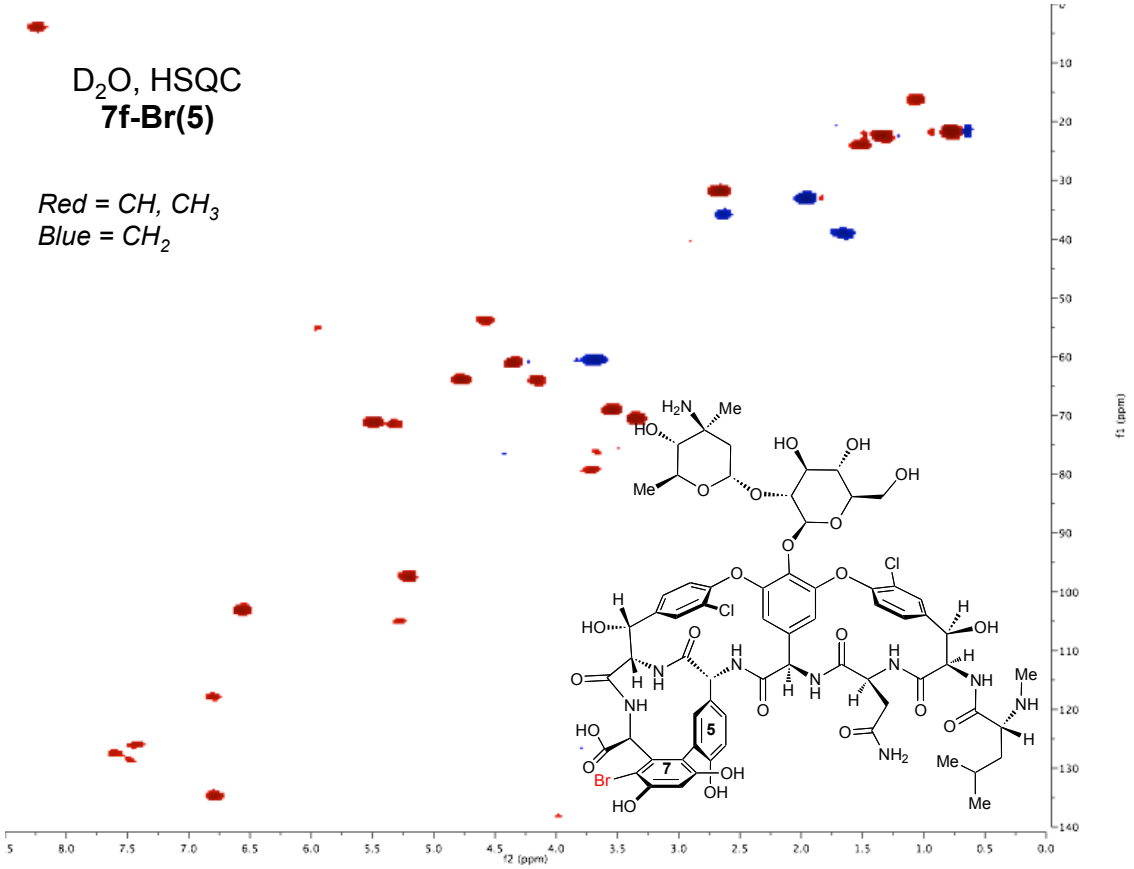

TPP-B1P234-7f-Br 178 (2.694)
1: TOF MS ES+
1.43e3
1385.1779
1387.2079
1383.1967
764.6476
765.1561
765.6473
763.6663
766.1561
766.6650
767.1918
1528.2849
1388.2234
1529.3011
1530.3427
1527.3185
1531.3350
1532.4019
1223.1826
1221.1880
1224.2028
1226.2223
1411.2543
1413.2555
1019.5663
1018.8781
1020.9230
306.1605
612.1023
725.7288
%
m/z


TPP-B1P234-7f-Br (2.694) Cu (0.50); Is (1.00,1.00) C66H75BrCl2N9O24
1: TOF MS ES+
2.51e12
Predicted
1528.3485
1529.3485
1530.3485
1526.3485
1531.3485
1532.3485
1533.3485
%
mass


TPP-B1P234-7f-Br 178 (2.694)
1: TOF MS ES+
582
Observed
1528.2849
1529.3011
1530.3427
1527.3185
1531.3350
1532.4019
1533.3700
%
m/z

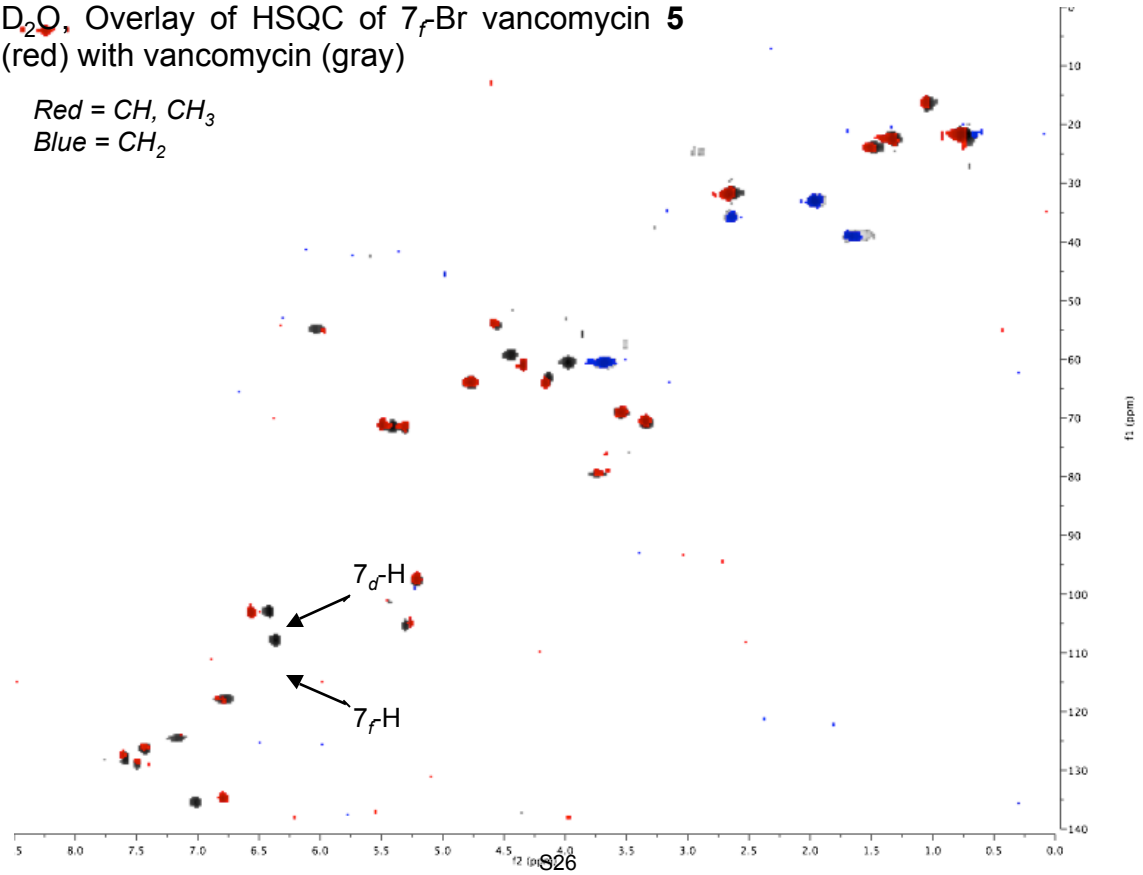
D_2O, HSQC
7f-Br(5)

Red = CH, CH_3
Blue = CH_2



D_2O, Overlay of HSQC of 7_f-Br vancomycin **5** (red) with vancomycin (gray)

Red = CH, CH_3
Blue = CH_2

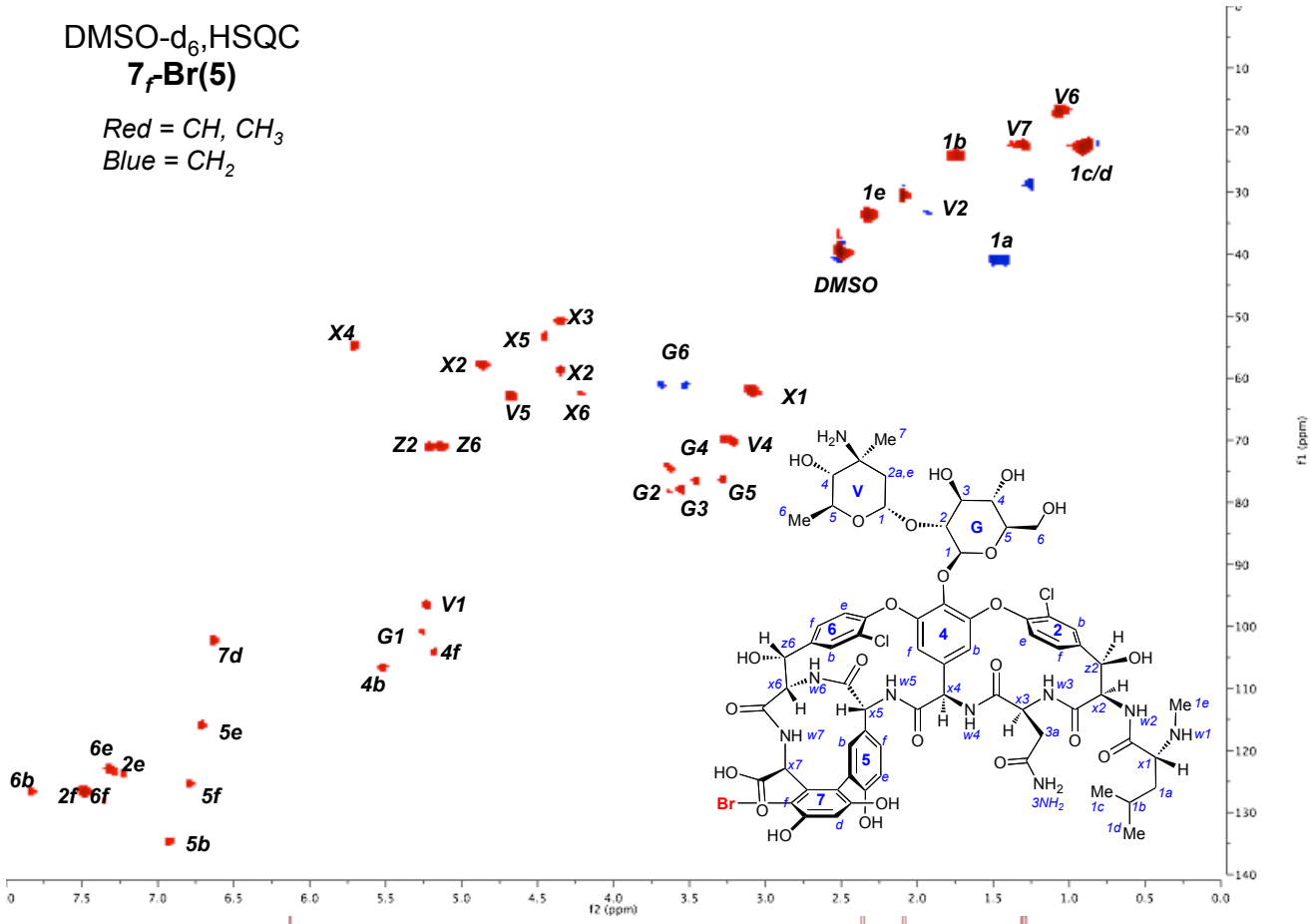


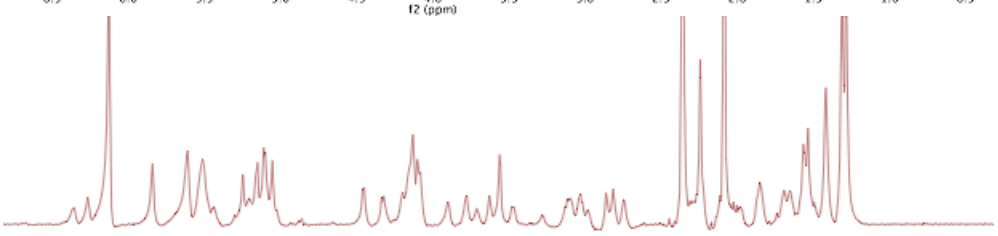
DMSO-d_6,HSQC

7_f-Br(5)

Red = CH, CH_3

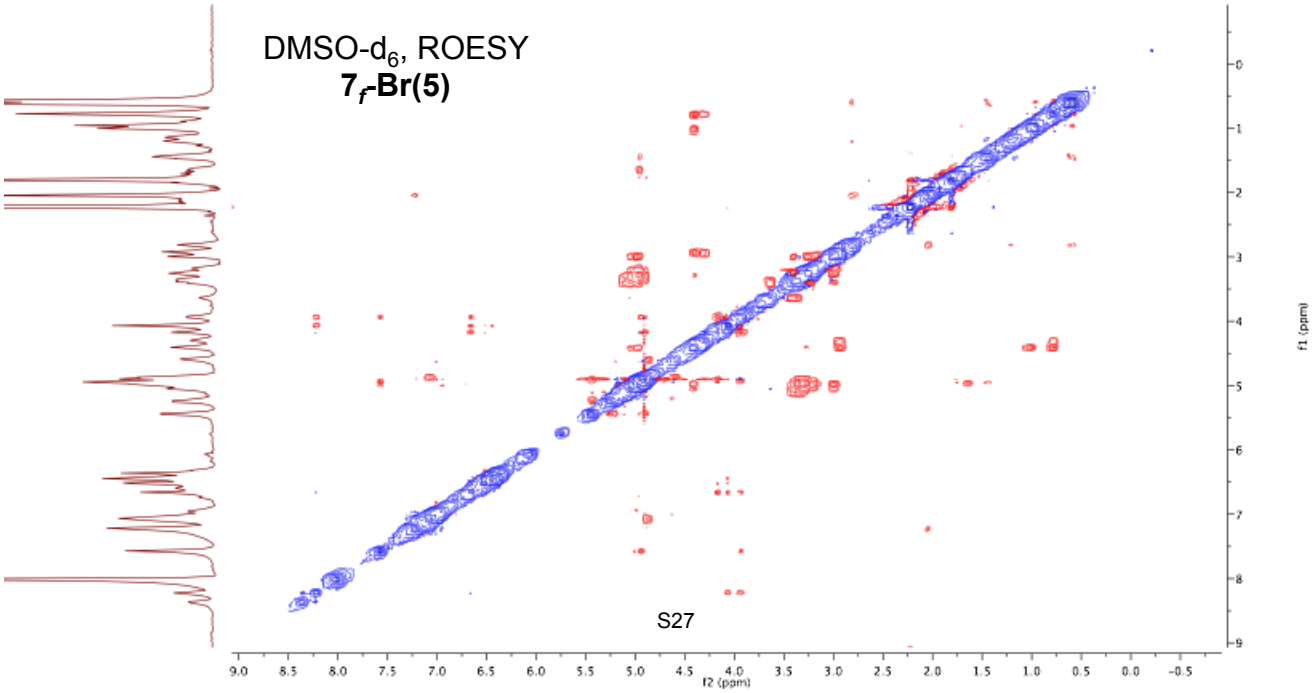
Blue = CH_2



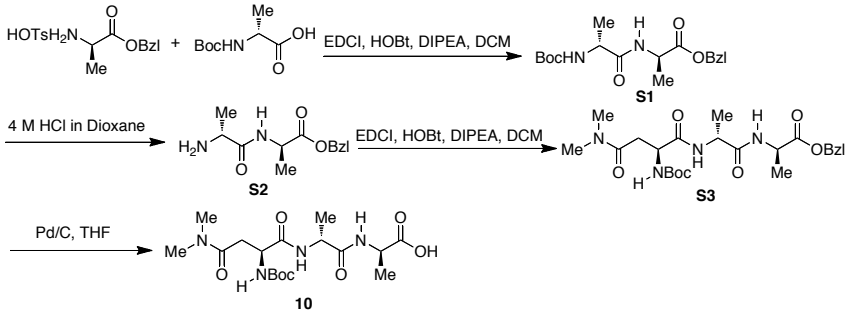


DMSO-d_6, ROESY

7_f-Br(5)



Peptide Synthesis:



Synthesis of Peptide **10**: To a dry 50 mL RBF equipped with stir bar were added 541.0 mg (2.86 mmol, 1.0 equiv.) of Boc-DAla-OH, 1.00 g (3.15 mmol, 1.1 equiv) of TsOH•H-DAla-OBzl, 819.0 mg (3.15 mmol, 1.1 equiv) of EDCI•HCl and 664.0 mg (3.15 mmol, 1.1 equiv) of HOBt•H_2O. To this, 10 mL of DCM was added and the reaction mixture was stir for *ca.*10 min. At this point, 700 μL (4.29 mmol, 1.5 equiv) of DIPEA were added. The reaction mixture was allowed to stir overnight at room temperature and reaction progress was monitored by LCMS. Upon complete conversion of starting material, 10 mL of 10% aq. citric acid solution (w/w) was added and reaction was stirred for 5 min. The reaction mixture was transferred to separatory funnel and was diluted with 70 mL of EtOAc. The aqueous layer was removed and organic layer was washed again with 10 mL of 10% aq. citric acid solution. Similarly, the organic layer was washed with 10 mL of saturated aq. $NaHCO_3$ (2 times) and 20 mL of Brine. The organic layer was dried over Na_2SO_4 and concentrated in vacuo to give a white solid (**S1**). This was treated with 4 M HCl in dioxane (5 mL) for 30 min followed by concentration in vacuo to provide HCl salt of **S2** as a white solid (700 mg). This material was carried forward without purification. In a dry 50 mL RBF equipped with stir bar were added 192.0 mg (0.77 mmol, 1.0 equiv.) of **S2**, 200.0 mg (0.77 mmol, 1.0 equiv) of Boc-Asn(Me_2)-OH, 162.0 mg (0.85 mmol, 1.1 equiv) of EDCI•HCl and 130.0 mg (0.85 mmol, 1.1 equiv) of HOBt•H_2O. To this 5 mL of DCM was added and reaction mixture was stir for *ca.*10 min. At this point, 174 μL (1.00 mmol, 1.3 equiv) of DIPEA was added. The reaction mixture was allowed to stir overnight at room temperature and the reaction progress was monitored using LCMS. Upon complete conversion of starting material, 10 mL of 10% aq. citric acid solution was added and reaction was stir for 5 min. The reaction mixture was transfer to separating funnel and was diluted with 50 mL of EtOAc. The aqueous layer was removed and organic layer was washed again with 10 mL of 10% aq. citric acid solution, 10 mL of saturated aq. $NaHCO_3$ (2 times) and 20 mL of Brine. The organic layer was dried over Na_2SO_4 and was concentrated in vacuo to give colorless oil. The crude mixture was purified by Biotage® using water and acetonitrile as eluents to give 320.0 mg of **S3** (84% Yield). Compound **S3,** 320.0 mg (0.65 mmol, 1.0 equiv) was dissolved in 10 mL of THF and carefully added to a flask containing 69.0 mg of Pd/C under N_2 atmosphere. The flask was evacuated and refilled with H_2 three times then stirred under H_2 atmosphere by using a H_2 balloon. Upon completion (by LCMS) the reaction mixture was passed through Celite® and was concentrated in vacuo to give peptide **10**.

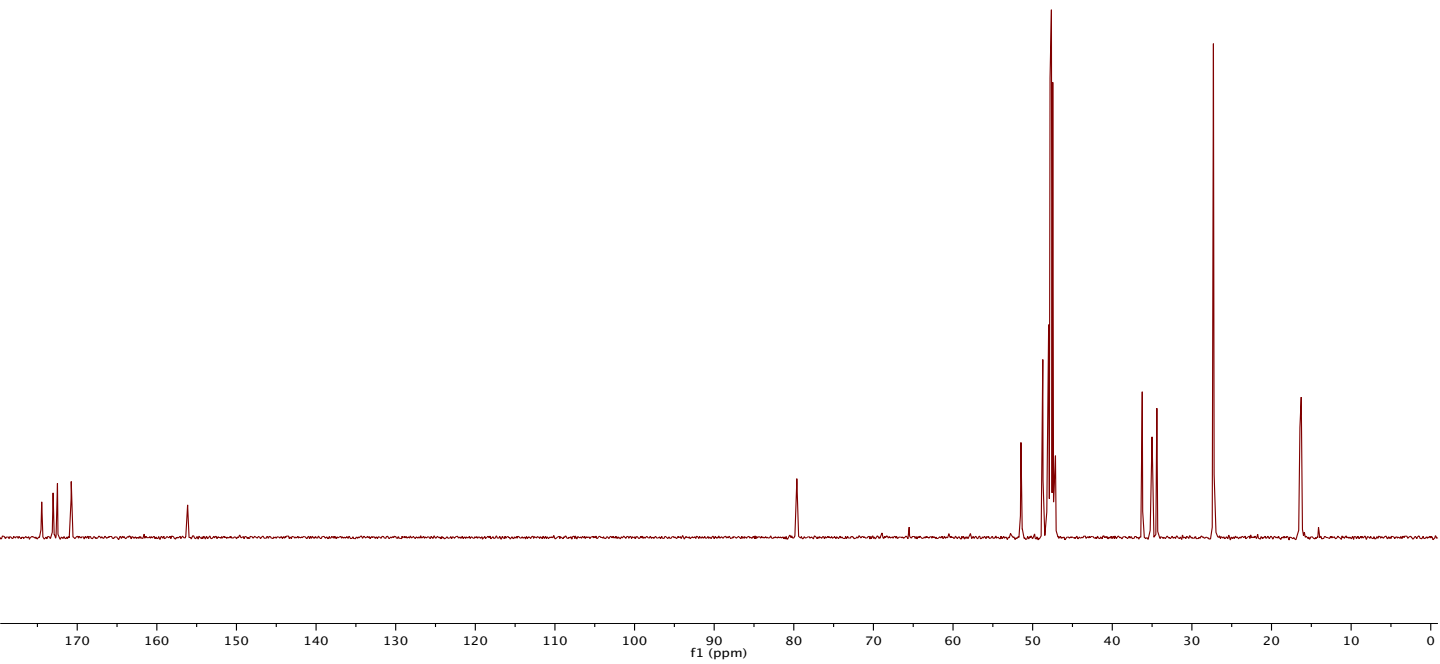
Boc-Asn(Me$_2$)-DAla-DAla-OH (10): $[\alpha]_D^{20}$ +342 (c 3.5, $CHCl_3$), **^{1}H NMR** (500 MHz, $CDCl_3$): δ 7.45 (s, 1H), 6.80 (s, 1H), 5.69 (d, *J* = 9.4 Hz, 1H), 4.41 (m, 3H), 3.29 (dd, *J* = 17.1, 3.5 Hz, 1H), 2.91 (s, 3H), 2.79 (s, 3H), 2.52 (dd, *J* = 17.1, 4.1 Hz, 1H), 1.38 (s, 15H). **^{13}C NMR** (125 MHz, $CDCl_3$): 174.2, 173.1, 172.8, 171.3, 155.7, 49.7, 48.8, 37.4, 36.3, 35.7, 28.5, 17.4, 17.0. **FT-IR**: 1521, 1635, 1646, 1700, 2934, 2979, 3313. **ESI-HRMS** ($M+H^+$): calc. 403.2124, obsrd. 403.1885.

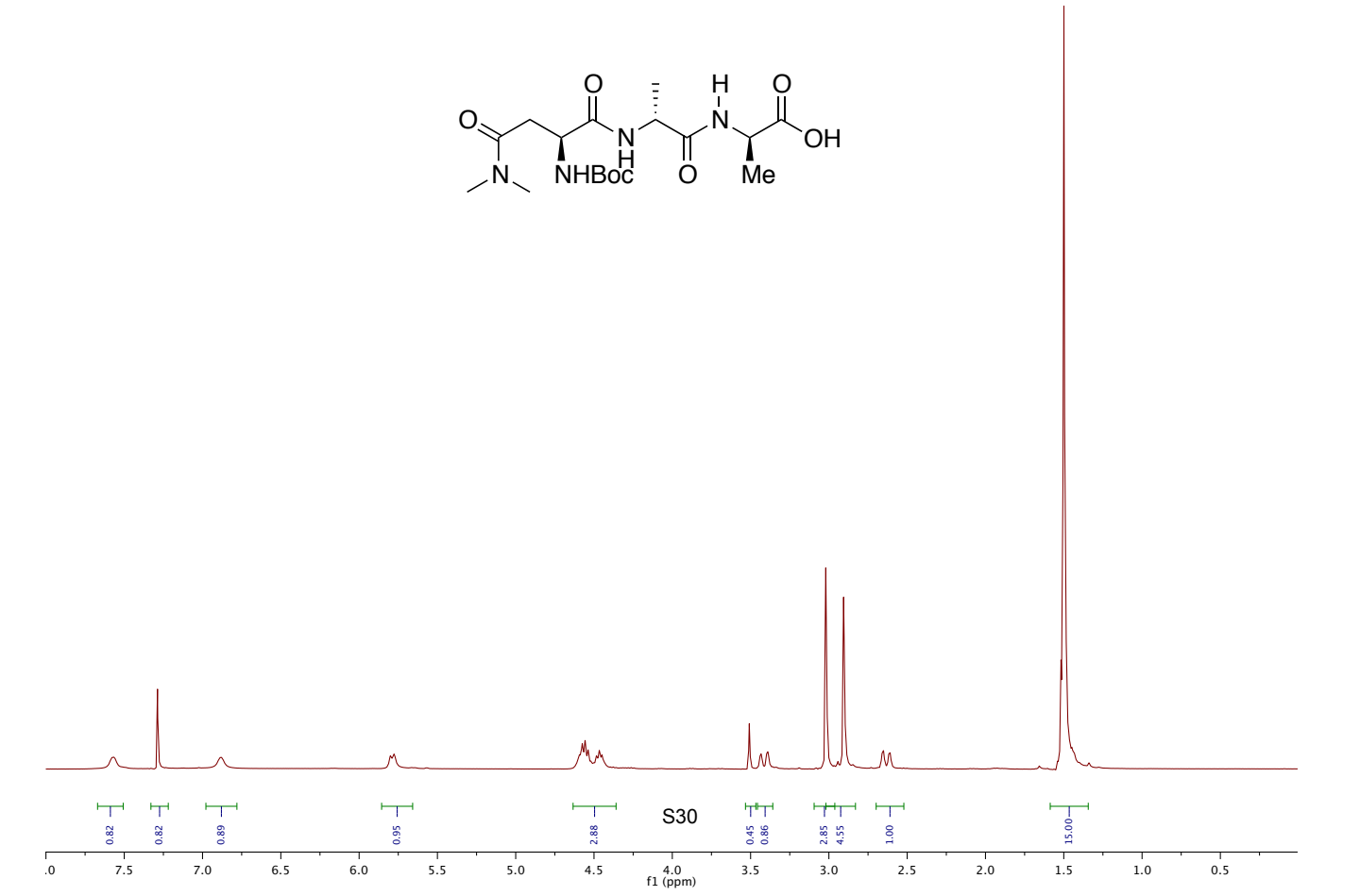
Boc-Leu-DGln(Me)$_2$-DAla-OH (18): ^{1}H NMR (500 MHz, CD_3OD) δ 4.40 (dd, *J* = 8.0, 5.6 Hz, 1H), 4.34 (q, *J* = 7.3 Hz, 1H), 4.05 - 3.96 (m, 1H), 3.28 (dt, *J* = 3.3, 1.6 Hz, 3H), 3.01 (s, 3H), 2.90 (s, 3H), 2.58 - 2.45 (m, 1H), 2.45 - 2.34 (m, 1H), 2.12 (dt, *J* = 14.1, 7.0 Hz, 1H), 1.89 (dt, *J* = 13.9, 6.9 Hz, 1H), 1.65 (dt, *J* = 13.3, 6.6 Hz, 1H), 1.50 (dd, *J* = 12.0, 6.5 Hz, 2H), 1.41 (s, 12H), 0.93 (d, *J* = 6.6 Hz, 3H), 0.90 (d, *J* = 6.6 Hz, 3H). **^{13}C NMR** (126 MHz, CD_3OD) δ 174.35, 174.21, 173.07, 171.82, 156.43, 79.23, 53.61, 52.16, 40.46, 36.28, 34.43, 28.72, 27.30, 24.56, 21.96, 20.56, 16.04. **ESI-HRMS** ($M+H^+$): calc. 459.2740, obsrd. 459.1924.

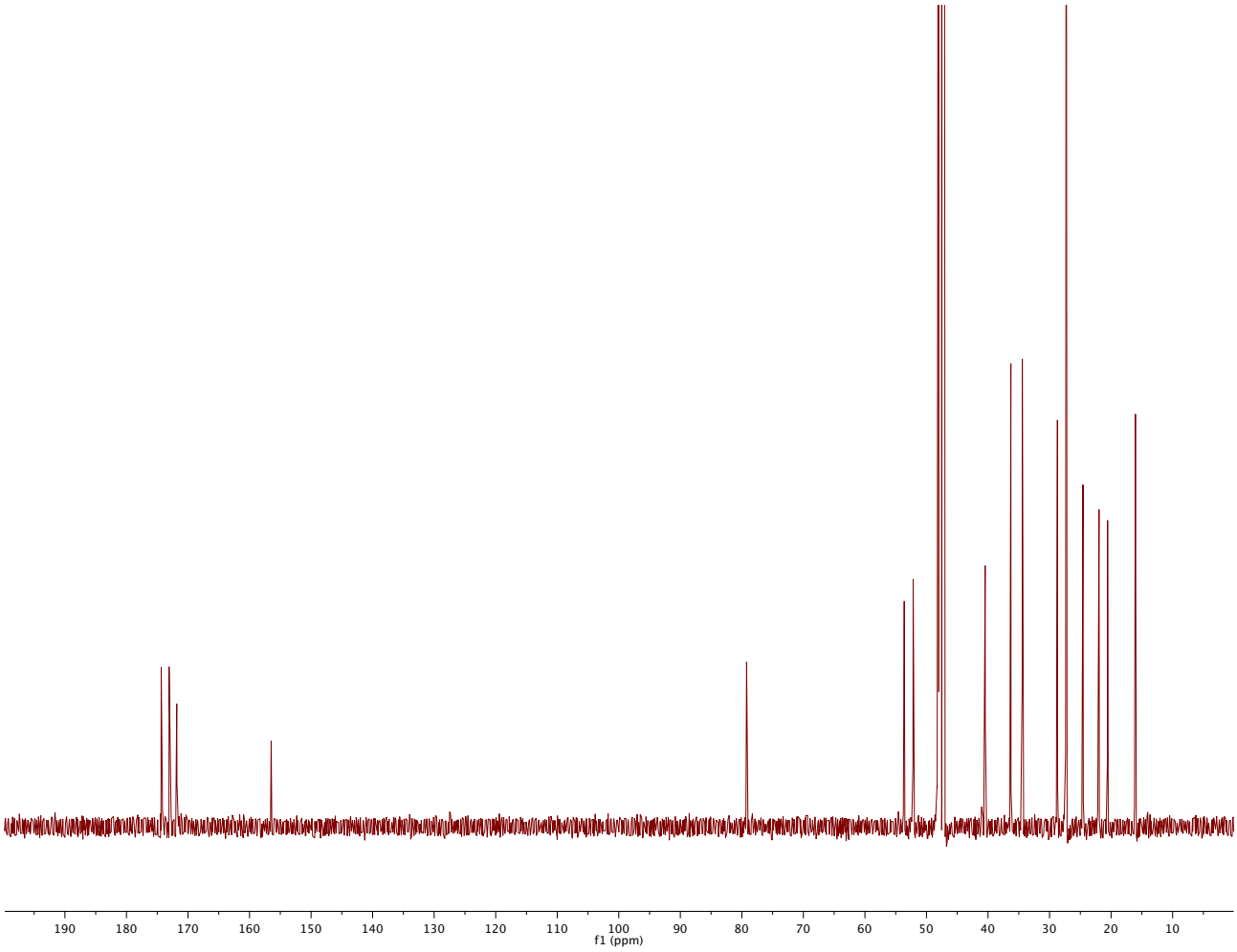
Table 4. MIC Determinations for Newly Synthesized Vancomycin Analogs.

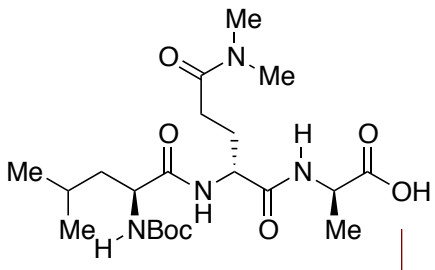
Compounds	S. aureus (standard) *S. aureus* ATCC[1] 29213 MMX[2] 100	S. aureus (MRSA) *S. aureus* ATCC[1] 43300 MMX[2] 2002	E. faecalis (standard) *E. faecalis* ATCC[1] 29212 MMX[2] 101	E. faecalis (VRE, VanB) *E. faecalis* ATCC[1] 51299 MMX[2] 202	E. faecalis (VRE, VanA) *E. faecalis* VanA MMX[2] 486
6	2, 2	4, 2	4, 4	64, 64	>64, >64
5	4, 4	4, 4	8, 8	>64, >64	>64, >64
8	4, 4	8, 4	2, 4	>64, >64	>64, >64
17	4, 4	8, 8	2, 4	64, 64	>64, >64
*Vancomycin (**4**)*	*0.5, 0.5*	*1, 1*	*2, 2*	*16, 16*	*>64, >64*

MSSA: methicillin-susceptible *S. aureus*, MRSA: methicillin-resistant *S. aureus*, VanS: vancomycin-susceptible, VanB: VanB resistance genotype/phenotype, VanA: VanA resistance genotype/phenotype
[1]American Type Culture Collection
[2]Micromyx Isolate Number
[3]CLSI acceptable limits for QC organisms (*S. aureus* ATCC 29213 and *E. faecalis* ATCC 29212) are shown in parentheses where applicable


170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

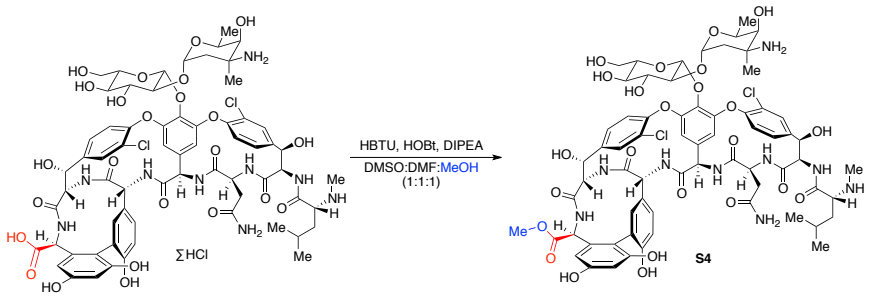

O
O
N
H
H
N
O
OH
N
NHBoc
O
Me
0.82
0.82
0.89
0.95
2.88
0.45
0.86
2.85
4.55
1.00
15.00
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
f1 (ppm)


190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
f1 (ppm)


Me
O
N
Me
O
H
O
Me
N
N
OH
H
Me
H
NBoc
O
Me

.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
f1 (ppm)

Synthesis of Vancomycin Methyl Ester:



Procedure is adapted from; Sundram, U. N.; Griffin, J. H. J. Org. Chem., ***1995****, 60, 1102.*

To a round bottom flask equipped with a stir bar were added 200.0 mg (0.134 mmol, 1.0 equiv) of vancomycin•HCl, 80.0 mg (0.206 mmol, 1.5 equiv) of HBTU and 29.0 mg (0.206 mmol, 1.5 equiv) of HOBt. DMSO (2.0 mL), DMF (2.0 mL) and MeOH (2.0 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min after which 65.0 µL (0.40 mmol, 3.0 equiv) of DIPEA were added. The reaction mixture was stirred at room temperature for 5 h (during the reaction the solution became clear and turned pale yellow). To the reaction vessel, 10 mL of H_2O was added and the reaction was stirred for an additional 15 min. Then, MeOH was removed *in vacuo* and the reaction mixture was analyzed by reverse phase HPLC and was purified by preparative reverse phase HPLC to give 32 mg of **S4**.

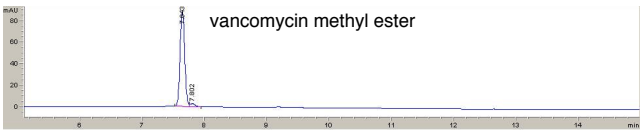
HPLC Method for preparative purification: Symmetry prep C8 7µm (19 X 300 mm) column, 24 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 50% MeCN over 3 min, 50 to 95% MeCN over 3 min hold at 95% MeCN for 3 min, 95% to 5% over 3 min.

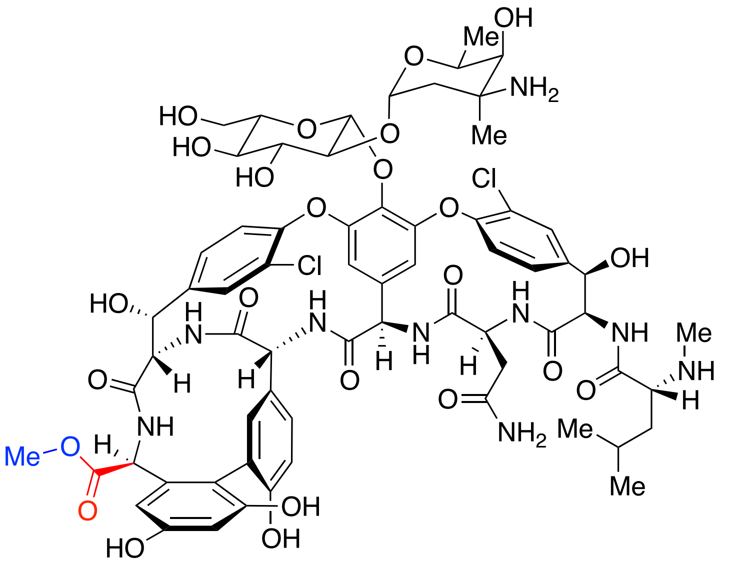
Analytical method: Symmetry prep C8 7µm column, 4 mL/min, starts at 5% MeCN (0.1% HCOOH) : 95% H_2O (0.1% HCOOH) for 3 min, 5 to 8.5% MeCN over 15 min, 8.5 to 50% MeCN over 3 min, 50 to 95% MeCN over 3 min hold at 95% MeCN for 3 min, 95% to 5% over 3 min. Retention time for desired product is 18.6 min.

The ^{1}H-NMR of the product was compared to the tabulated 1H NMR data reported in the study of Nicolaou. *Nicolaou, K. C.; Mitchell, H. J.; Jain, N. F.; Bando, T.; Hughes, R.; Winssinger, N.; Natarajan, S.; Koumbis, A. E. Chem. Eur. J.* ***1999****, 5, 2648.*

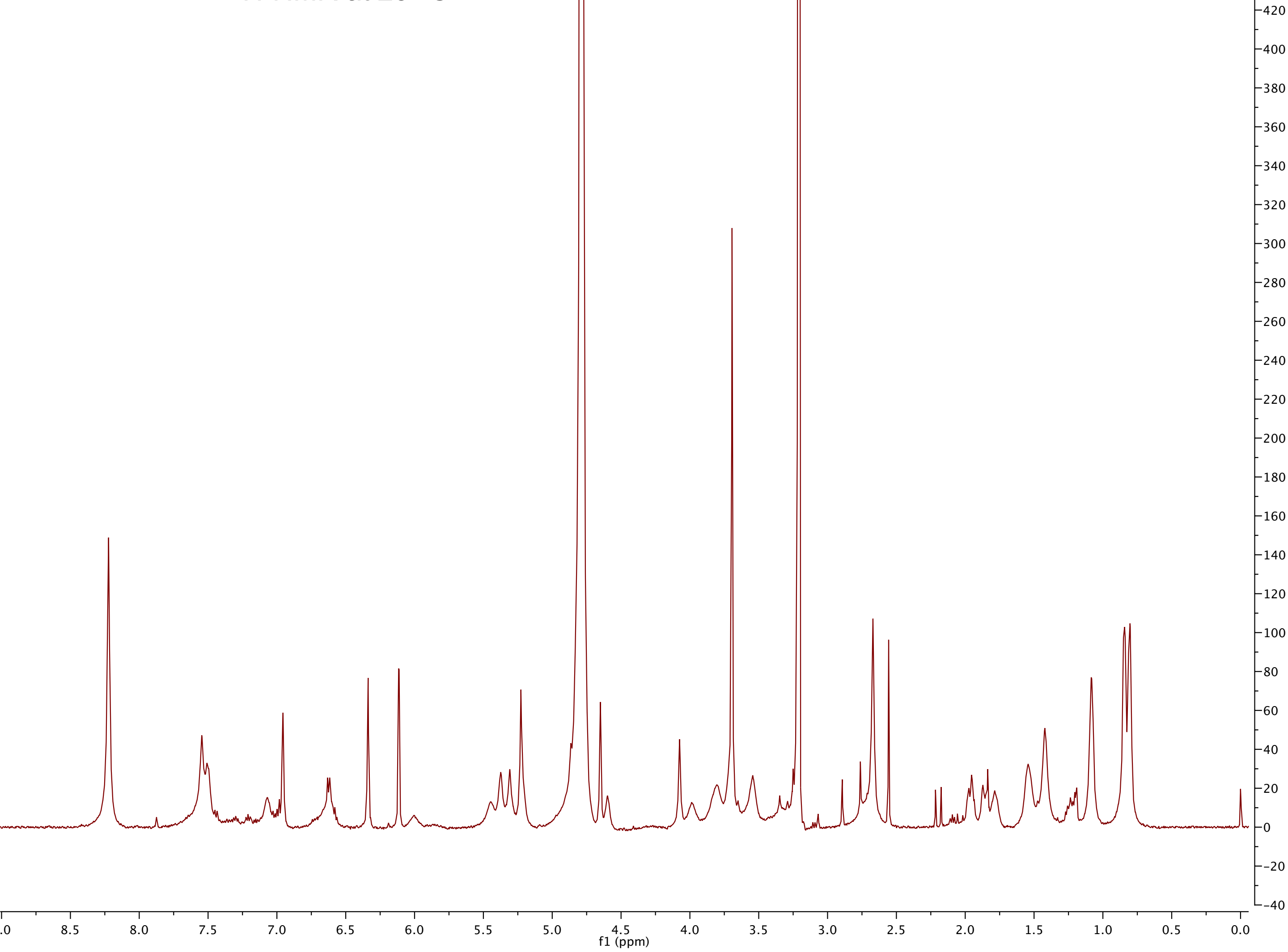
In addition, we performed additional NMR experiments to assess the assignment. See below for raw proton, NOESY, HSQC data.

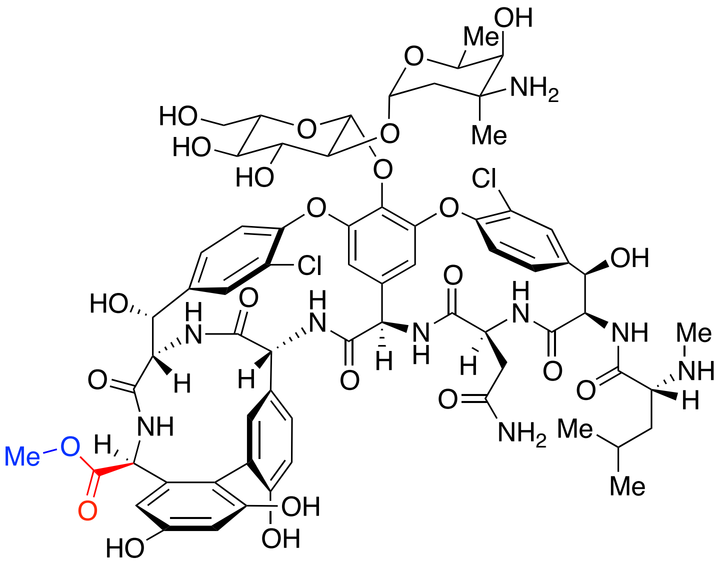
HPLC Trace:



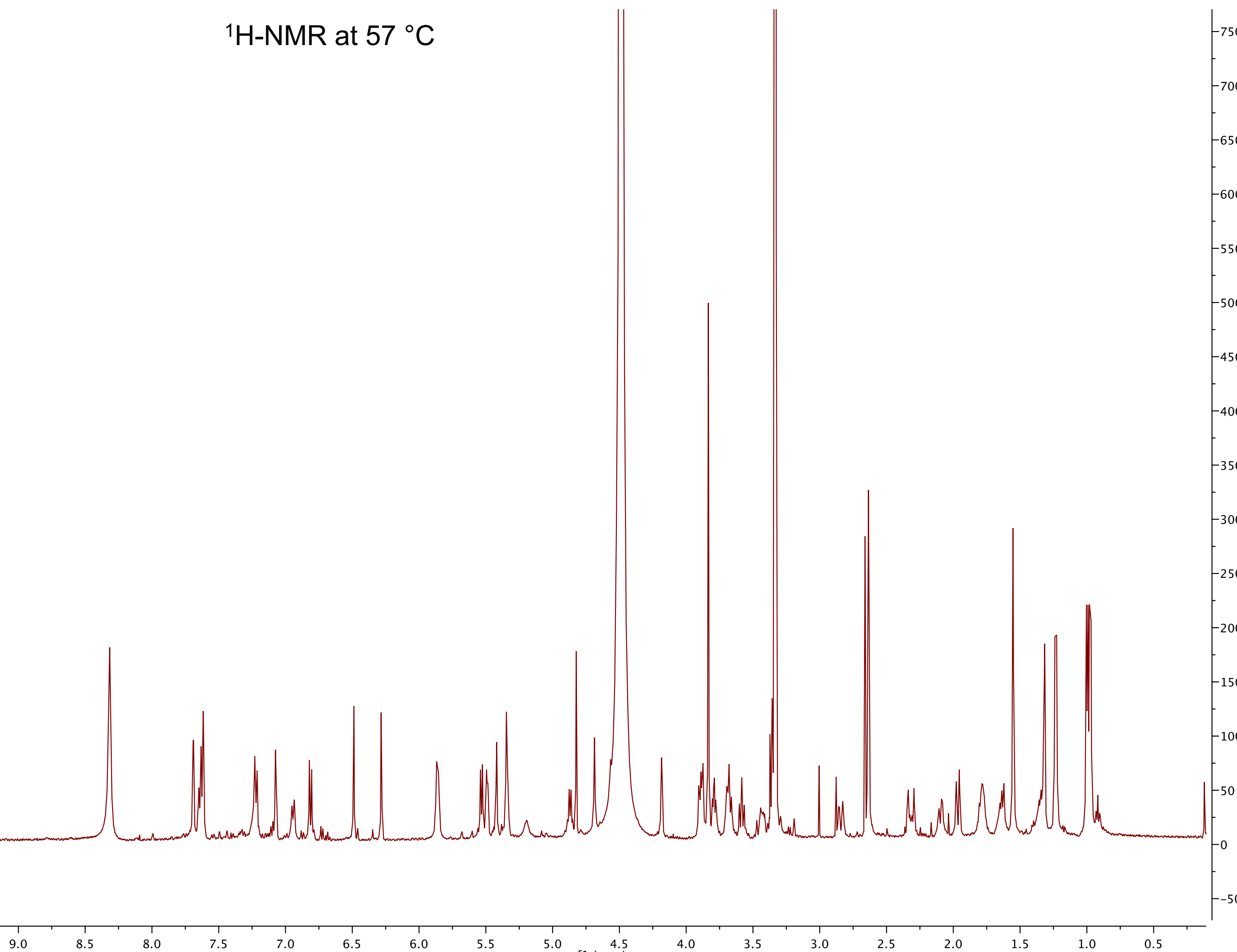


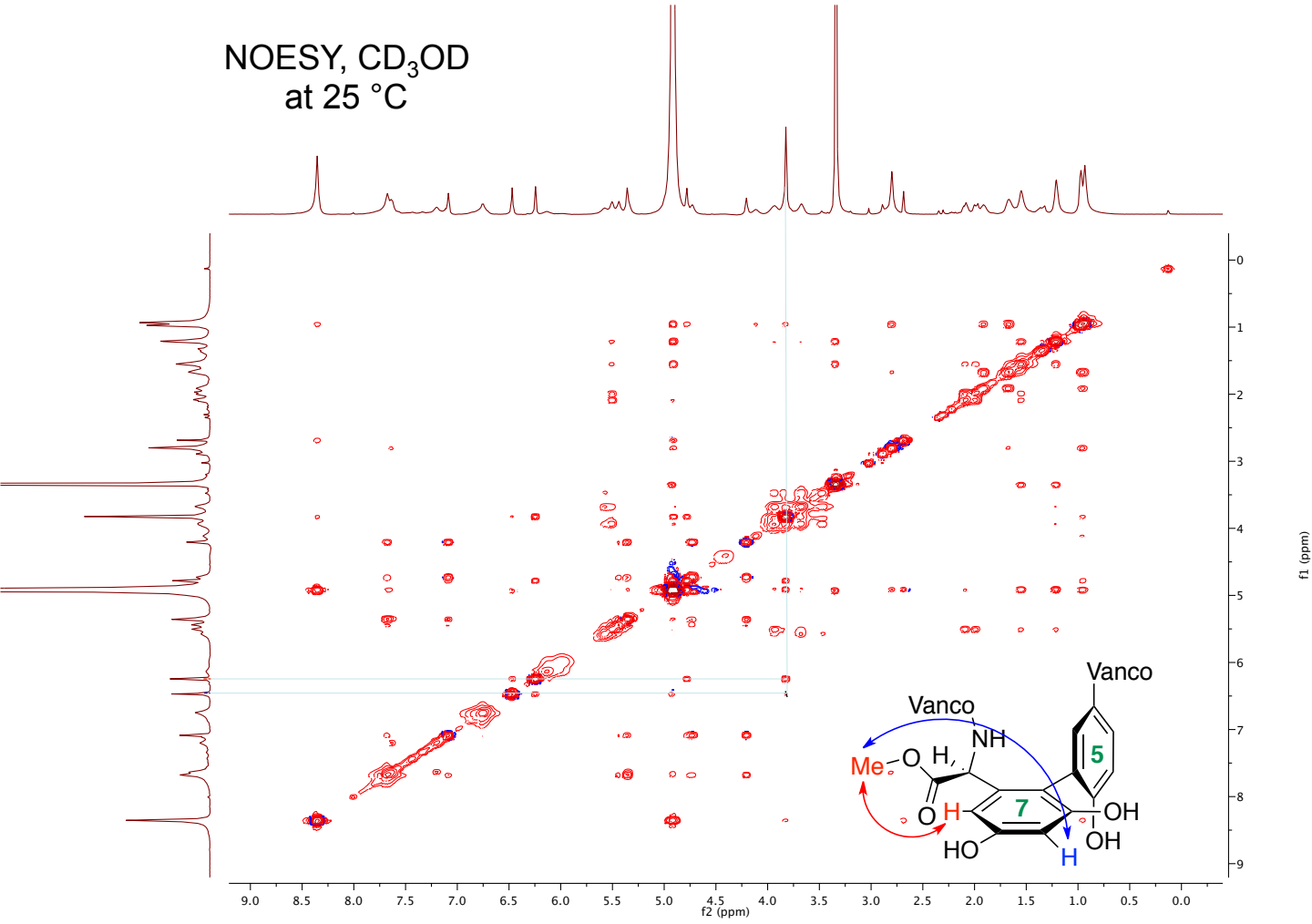
^{1}H-NMR at 25 °C

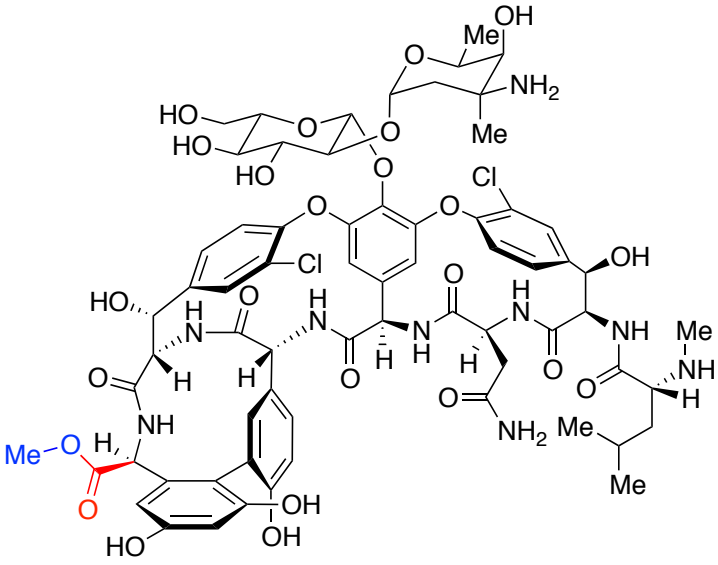

f1 (ppm)
9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
440 420 400 380 360 340 320 300 280 260 240 220 200 180 160 140 120 100 80 60 40 20 0 −20 −40

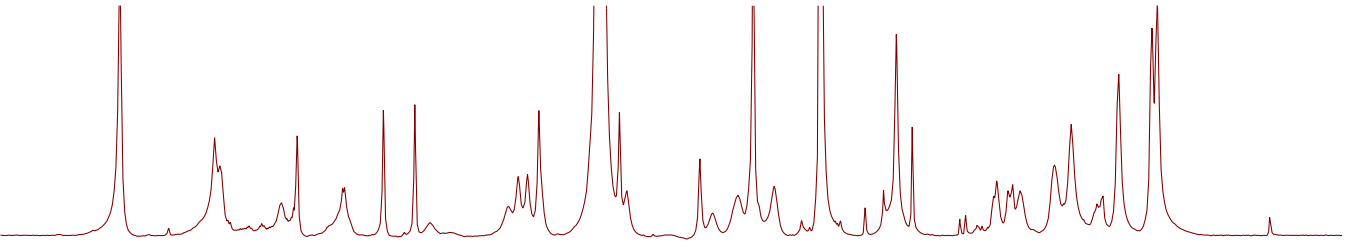

OH
Me
O
NH2
Me
HO
O
O
HO
HO
O
Cl
O
O
OH
Cl
HO
H
N
O
H
N
O
H
N
O
N
H
H
O
H
N
O
NH
Me
NH
H
O
NH
H
O
H
H
O
H
Me
O
NH2
Me
Me
Me
O
H
O
OH
HO
OH

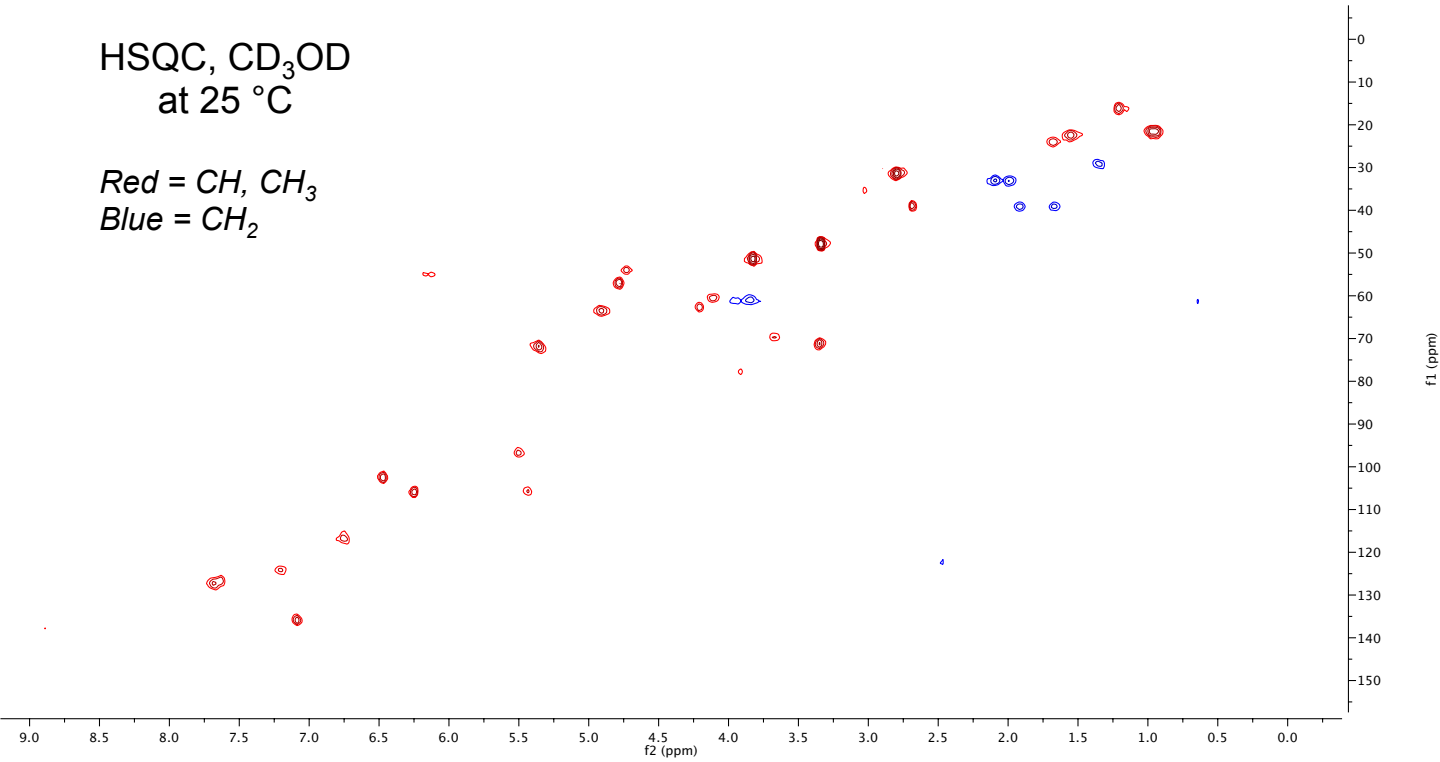
^{1}H-NMR at 57 °C


750
700
650
600
550
500
450
400
350
300
250
200
150
100
50
0
−50
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
f1 (ppm)


NOESY, CD3OD
at 25 °C
f2 (ppm)
f1 (ppm)
Vanco
NH
Me
O
H
O
H
7
5
OH
OH
HO
H

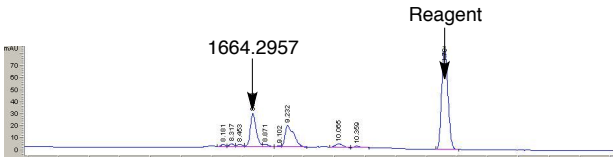




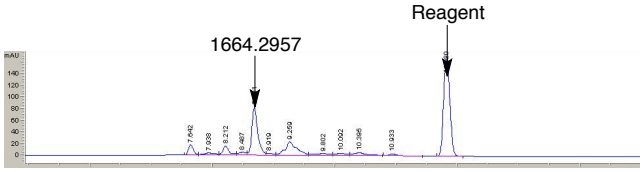

HSQC, CD3OD
at 25 °C
Red = CH, CH3
Blue = CH2
f2 (ppm)
f1 (ppm)

Bromination of Vancomycin Methyl Ester (S4):

Control: To a 2.5 dram vial equipped with a stir bar were added 12.0 mg (0.008 mmol, 1.0 equiv) of vancomycin methyl ester (**S4**). MeOH (2 mL) was added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, when 7.3 mg (0.032 mmol, 4.0 equiv) of NBP was added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.



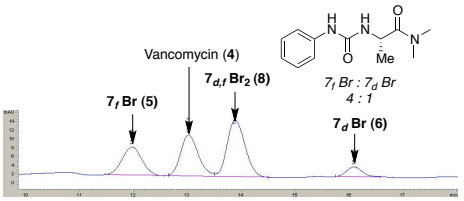
With Guanidine•HCl: To a 2.5 dram vial equipped with a stir bar were added 12.0 mg (0.008 mmol, 1.0 equiv) of vancomycin methyl ester (**S4**), 13.6 mg of guanidine•HCl (18.0 equiv) and MeOH (2 mL). The reaction mixture was allowed to stir for ca. 5 min, after which 7.3 mg (0.032 mmol, 4.0 equiv) of NBP was added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.



The HRMS and HPLC analysis of both reactions revealed that major product formed in both reactions has a mass of 1664.2957 ($M+H^+$). The identity of this material is not assigned; however, the isotope pattern of mass spectrum is consistent with that of a di-bromide containing a vancomycin methyl ester analogue with molecular formula of $C_{69}H_{81}Br_2Cl_2N_9O_{25}$. These experiments did not deliver ready access to monobromides analogous to **7_f-Br** over **7_d-Br**, as their corresponding methyl esters.

Other additives that might target carboxylic acid functionality, such as ureas, also provide some selectivity for **7_f-Br** over **7_d-Br** upon reaction with vancomycin (**4**). The raw data for these promoters are presented below.

Reaction Conditions: 0.008 mmol of **4**, 4 equiv of NBP, 3 equiv of urea, 1 mL of MeOH



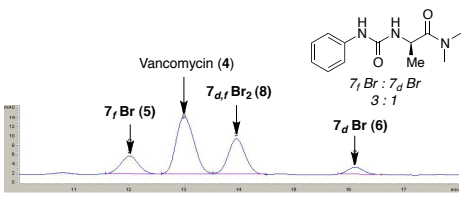
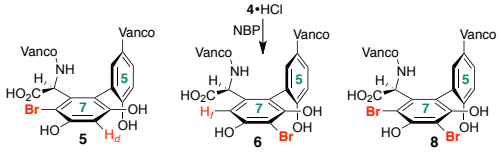


Table 4. Optimization of Reaction Conditions Aiming Analog **5** (Manuscript Table **2**)



Entry	Additive	Solvent	NBP (equiv)	t (h)	% conv.	5 : 7$_f$Br : 7$_d$Br : 7$_{d,f}$Br$_2$ 6 : 8
1	No catalyst	H_2O:MeOH (5:1)	2	12	71	1.0 : 1.0 : 1.3[b]
2	No catalyst	MeOH	2	2	17	1.0 : 1.0 : 3.0[b]
3	No catalyst	MeOH	4	1	92	14.3 : 1.0 : **53.8**[c]
4	**10** (2 equiv)	MeOH	4	1	31	0.5 : 1.0 : 1.3[b]
5	Guanidine•HCl	MeOH	4	1	85	**12.7 : 1.0 : 10.8**[c]
6	Guanidine•HCl	MeOH	4	1	92	11.8 : 1.0 : 11.4[d]

a) Ratios were measured by HPLC at 280 nm wavelength. b) 8 μmol of **4**, 6.6 mM. c) 16 μmol of **4**, 3.3 mM, 18 equiv of guanidine•HCl. d) simmilar to (c) except with 6 equiv of guanidine•HCl.

General Procedure for Table 4:

Entry 1: To a HPLC vial equipped with a stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl. Water (250 μL) and MeOH (50 μL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 2: To a HPLC vial equipped with a stir bar were added 12.5 mg (0.008 mmol, 1.0 equiv) of vancomycin•HCl. MeOH (1.2 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 3.7 mg (0.016 mmol, 2.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 3: To a HPLC vial equipped with a stir bar were added 25.0 mg (0.016 mmol, 1.0 equiv) of vancomycin•HCl. MeOH (4.0 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 14.6 mg (0.064 mmol, 4.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 4: To a HPLC vial equipped with a stir bar were added 25.0 mg (0.016 mmol, 1.0 equiv) of vancomycin•HCl and peptide **10** (2 equiv). MeOH (4.0 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 14.6 mg (0.064 mmol, 4.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 5: To a HPLC vial equipped with a stir bar were added 25.0 mg (0.016 mmol, 1.0 equiv) of vancomycin•HCl and guanidine•HCl (18 equiv). MeOH (4.0 mL) were added to the reaction flask. The reaction mixture was allowed to stir for ca. 5 min, after which 14.6 mg (0.064 mmol, 4.0 equiv) of NBP were added. Aliquots were collected at regular time intervals and analyzed by reverse phase HPLC.

Entry 6: The same procedure as entry 5 was employed, except 6 equiv of guanidine•HCl was used

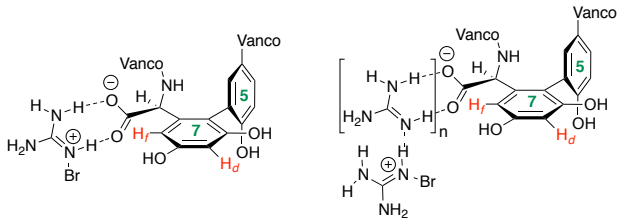


Figure 8. Speculative concept for bromide delivery by guanidine.