Angewandte Chemie
Eine Zeitschrift der Gesellschaft Deutscher Chemiker

# Supporting Information



69451 Weinheim, Germany

## Supporting Information

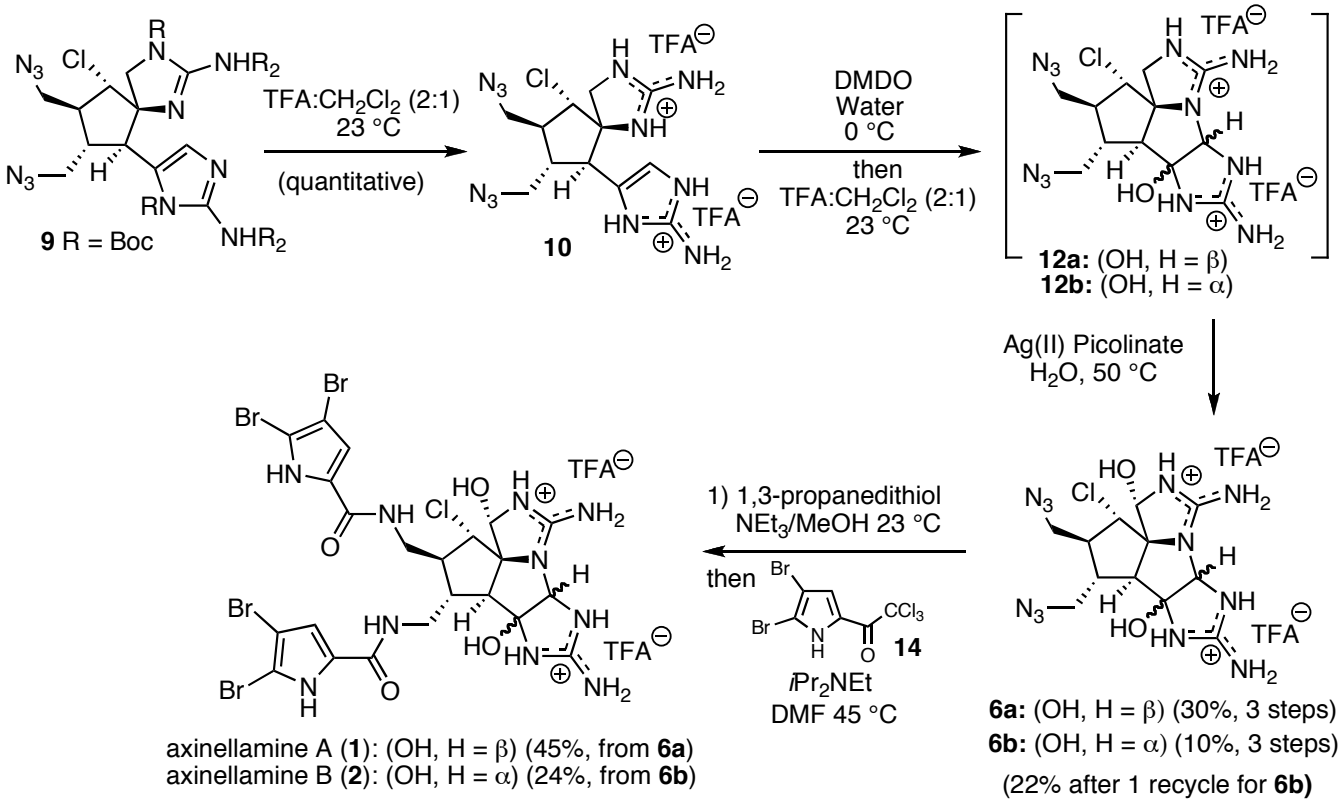
# Total Synthesis of (±)-Axinellamines A and B

Daniel P. O'Malley, Junichiro Yamaguchi, Ian S. Young,[+] Ian B. Seiple[+] and Phil S. Baran*

*Contribution from the Department of Chemistry, The Scripps Research Institute, 10650 North Torrey Pines Road, La Jolla, California 92037*

**General Procedures.** All reactions were carried out under an inert atmosphere of nitrogen or argon with dry solvents, using anhydrous conditions unless otherwise stated. Dry acetonitrile ($CH_3CN$), dichloromethane ($CH_2Cl_2$), methanol (MeOH), *N*,*N*-dimethylformamide (DMF) and triethylamine ($NEt_3$), were obtained by passing the previously degassed solvents through activated alumina columns. Reagents were purchased at the highest commercial quality and used without further purification, unless otherwise stated. Silver (II) picolinate was prepared according to the procedure of T. G. Clarke, N. A. Hampson, J. B. Lee, J. R. Morley, B. Scanlon, *Can. J. Chem*. **1969**, 47, 1649 – 1654. Yields refer to chromatographically and spectroscopically ($^1$H-NMR) homogeneous material, unless otherwise stated. Reactions were monitored by HPLC-MS on reversed phase column, using acetonitrile/water/0.1% formic acid as the mobile phase. NMR spectra were recorded on Bruker DRX-600, and calibrated using residual undeuterated solvent as an internal reference. The following abbreviations were used to explain multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, br = broad. High resolution mass spectra (HRMS) were recorded on Agilent LC/MSD TOF mass spectrometer by electrospray ionization time of flight reflectron experiments. IR experiments were recorded on a Perkin Elmer Spectrum BX FTIR spectrometer.

***Chart 1.*** Synthetic scheme for (±)-axinellamines A and B.




**9** R = Boc

**10**

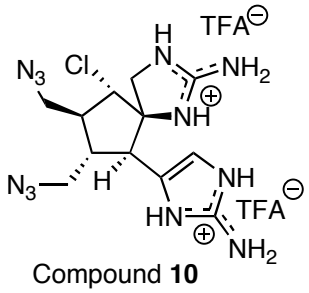
**12a:** (OH, H = β)
**12b:** (OH, H = α)

**6a:** (OH, H = β) (30%, 3 steps)
**6b:** (OH, H = α) (10%, 3 steps)
(22% after 1 recycle for **6b)**

**14**

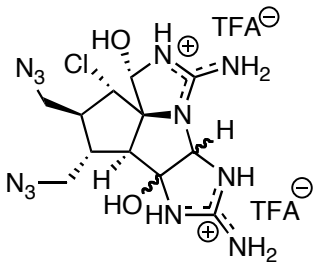
axinellamine A (**1**): (OH, H = β) (45%, from **6a**)
axinellamine B (**2**): (OH, H = α) (24%, from **6b**)

## Experimental Procedures



Compound **10**

To a solution of compound **9** (90 mg, 0.093 mmol, 1.0 equiv) in dichloromethane (1 mL) was added trifluoroacetic acid (2 mL) and the reaction stirred at 23 °C for 3 h. The solvent was then removed under reduced pressure to yield crude **10** (52 mg, 0.093 mmol, quantitative) that was of very high purity and did not require further purification. IR (neat) ν = 3183 (br), 2921, 2852, 2105, 1700, 1684, 1282, 1202, 1140 $cm^{-1}$; $^{1}$H-NMR (600 MHz, $CD_3OD$) δ 6.79 (s, 1 H), 4.21 (d, *J* = 10.2 Hz, 1 H), 4.07 (d, *J* = 10.2 Hz, 1 H), 3.80 – 3.73 (m, 2 H), 3.72 (d, *J* = 10.8 Hz, 1 H), 3.63 – 3.57 (m, 2 H), 3.23 (d, *J* = 7.8 Hz, 1 H), 2.35 – 2.32 (m, 1 H), 2.16 – 2.14 (m, 1 H); $^{13}$C-NMR (150 MHz, $CD_3OD$) δ 160.5, 149.4, 125.8, 112.6, 73.1, 65.9, 53.0, 52.6, 50.9, 47.2, 46.9, 43.0 HRMS (ESI-TOF) calcd for $C_{12}H_{18}ClN_{12}$ [M+H$^+$-2TFA] 365.1466; found 365.1465



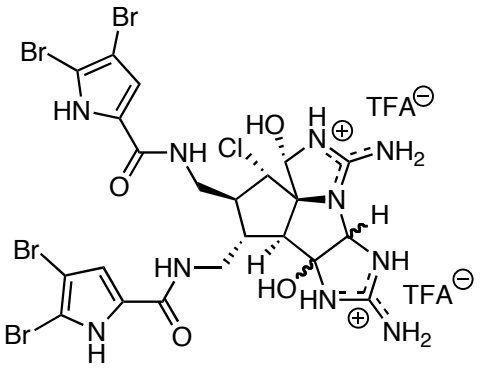
Compound **6a** OH, H = β
Compound **6b** OH, H =α

Compound **10** (15.4 mg, 0.026 mmol, 1.0 eq) was dissolved in water (1.85 mmol) and cooled to 0 °C. DMDO (1.85 mL of a ca. 0.02 M solution in acetone, 0.037 mmol, 1.4 eq) was added and the reaction stirred for 1.5 h at 0 °C. The solvent was removed under reduced pressure to yield crude diol **10** as a mixture of two major diastereomers, along with a minor amount of tetracycle **12**. The crude reaction mixture was azeotropically dried with toluene (2 x 1 mL), then dissolved in dichloromethane (1.25 mL). To this solution was added trifluoroacetic acid (2.5 mL) and the reaction stirred for 13 h at 23 °C, then the solvent removed under reduced pressure to yield crude **12** as a mixture of two major diastereomers. The crude mixture was dissolved in water (3 mL) and silver(II) picolinate (27 mg, 0.078 mmol, 3.0 eq) added and the reaction heated to 50 °C. After 2.5 and 5 h, additional portions of silver(II) picolinate (9.0 mg, 0.026 mmol, 1 eq) were added. After 16 h a final portion of silver(II) picolinate (18 mg, 0.052 mmol, 2.0 eq) was added. After an additional 3 h the reaction was filtered and the water removed under reduced pressure. The crude reaction mixture was purified by preparative HPLC (30 x 250 mm waters Atlantis C-18 column, detection at 210 nm, flow rate 42.5 mL/min, $H_2O$/MeCN with 0.1% TFA gradient: 0 min 5% MeCN, 35 min 30%, 40 min 90%, 45 min 90%) to give the major diastereomer **6a** (4.9 mg, 0.0083 mmol, 30% from **10,** rt = 21 min) and **6b** (1.6 mg, 0.0027 mmol 10% from **10**, rt = 27 min) along with unoxidized material (4.4 mg, 28%) which corresponds to the tetracycle of **6b**. This material could be recycled under the above conditions to give additional **6b** (1.9 mg, 0.0032 mmol, 12%).

Data for compound **6a** IR (neat) ν = 3170 (br), 2109, 1700, 1684, 1558, 1436, 1270, 1201, 1136, 844, 802, 723 $cm^{-1}$; $^1$H-NMR (600 MHz, $CD_3OD$) δ 5.42 (s, 1 H), 5.41 (s, 1 H), 4.23 (d, *J* = 12.0 Hz, 1 H), 3.76 (dd, *J* = 13.2, 3.0 Hz, 1 H), 3.63 – 3.57 (m, 3 H), 3.21 (d, *J* = 12.0 Hz, 1 H), 2.78 – 2.74 (m, 1 H), 2.07 – 2.03 (m, 1 H); $^{13}$C-NMR (150 MHz, $CD_3OD$) δ 158.7, 158.1, 101.8, 85.0, 81.0, 79.8, 66.0, 53.8, 51.3, 50.2, 37.7 (one signal is missing presumably due to overlap with solvent, $^{13}$C-NMR also run in $CD_3CN$ to avoid overlap); $^{13}$C-NMR (150 MHz, $CD_3CN$) δ 159.0, 157.4, 101.5, 84.4, 80.0, 79.6, 65.1, 53.5, 51.0, 49.8, 48.1, 37.0; (ESI-TOF) calcd for $C_{12}H_{18}ClN_{12}O_2$ [M+H$^+$-2TFA] 397.1364; found 397.1357.

Data for compound **6b** IR (neat) ν = 3211 (br), 2110, 1684, 1539, 1201, 1142, 800 $cm^{-1}$; $^1$H-NMR (600 MHz, $CD_3OD$) δ 5.56 (s, 1 H), 5.49 (s, 1 H), 4.04 (d, *J* = 12.0 Hz, 1 H), 3.80 (dd, *J* = 13.2, 2.4 Hz, 1 H), 3.73 (dd, *J* = 13.2, 3.6 Hz, 1 H), 3.57 (dd, *J* = 12.0, 5.4 Hz, 1 H), 3.47 (dd, *J* = 12.0, 7.8 Hz, 1 H), 3.22 (d, *J* = 3.6 Hz, 1 H), 2.49 – 2.47 (m, 1 H), 2.08 – 2.04 (m, 1 H); $^{13}$C-NMR (150 MHz, $CD_3OD$) δ 160.4,

158.9, 104.4, 85.0, 82.1, 80.3, 65.0, 54.8, 52.8, 50.3, 50.2, 38.6; HRMS (ESI-TOF) calcd for $C_{12}H_{18}ClN_{12}O_2$ [M+H$^+$-2TFA] 397.1364; found 397.1356.



axinellamine A (**1**) OH, H = β
axinellamine B (**2**) OH, H = α

**Preparation of Axinellamine A** (**1**) To a solution of **6a** (4.0 mg, 0.00641 mmol, 1.0 eq) in methanol (0.1 mL) was added 1,3-propanedithiol (10 μL, 0.098 mmol, 15.3 eq) and triethylamine (10 μL, 0.071mmol, 11.0 eq) at 23 °C and the solution stirred for 2 h (wrapped in aluminum foil). The solution was diluted with dichloromethane (1 mL) and water (1 mL) was added. The layers were vigorously mixed, and the dichloromethane layer discarded. The aqueous phase was washed two additional times with dichloromethane, and then the water removed under reduced pressure to produce the crude bis-amine (3.6 mg, 0.0058 mmol) that was of sufficient purity to be used directly for acylation. To a solution of crude bis-amine **13a** in DMF (370 μL) was added trichloroacetylpyrrole **14** (7.2 mg, 0.0192 mmol, 3.0 eq) mL). To this solution was added Hünig's base in DMF slowly sub-surface (34 μL, 10% v/v). The reaction was wrapped in foil to exclude light and heated to 45 ºC for 11 h, after which the solvent was removed under reduced pressure. The crude reaction mixture was purified by preparative HPLC (30 x 250 mm waters Atlantis C-18 column, detection at 280 nm, flow rate 42.5 mL/min, $H_2O$/MeCN with 0.1% TFA gradient: 0 min 30% MeCN, 30 min 60%, 35 min 90%, 40 min 90%, rt = 30 min) to yield pure axinellamine A (**1**) (3.1 mg, 45% from **6a**).

**Preparation of Axinellamine B (1)** To a solution of **6b** (2.4 mg, 0.004 mmol, 1.0 eq) in methanol (0.1 mL) was added 1,3-propanedithiol (8 μL, 0.080 mmol, 20.0 eq) and triethylamine (8 μL, 0.057 mmol, 14.0 eq) at 23 °C and the solution stirred for 2 h (wrapped in aluminum foil). The solution was diluted with dichloromethane (1 mL) and water (1 mL) was added. The layers were vigorously mixed, and the dichloromethane layer discarded. The aqueous phase was washed two additional times with dichloromethane, and then the water removed under reduced pressure to produce the crude bis-amine **13b** that was of sufficient purity to be used directly for acylation. A solution of trichloroacetylpyrrole **14** (4.7 mg, 0.013 mmol, 3.3 eq) in DMF (0.24 mL) was added to neat crude diamine **13b**. To this solution was added Hünig's base in DMF slowly sub-surface (22 μL, 10% v/v). The reaction was wrapped in foil to exclude light and heated to 45 ºC for 4 h, after which the solvent was removed under reduced pressure. The

crude reaction mixture was purified by preparative HPLC (30 x 250 mm waters Atlantis C-18 column, detection at 280 nm, flow rate 42.5 mL/min, $H_2O$/MeCN with 0.1% TFA gradient: 0 min 30% MeCN, 30 min 60%, 35 min 90%, 40 min 90%, rt = 28 min) to yield pure axinellamine B (**2**) (1.0 mg, 24% from **6b**).

Monitoring the above reactions (azide reduction and acylation) by LC-MS and crude $^1$H-NMR suggest that they are fairly efficient (>80%) and the diminished overall yield for the two steps sequence may be the result of the purification process on a small scale

Axinellamine A (**1**) $^1$H-NMR (600 MHz, DMSO-$d_6$) δ 12.73 (d, $J$ = 2.4 Hz, 1 H), 12.71 (d, $J$ = 2.4 Hz, 1 H), 9.88 (s, 1 H), 9.67 (s, 1 H), 9.00 (bs, 1 H), 8.94 (s, 1 H), 8.69 (bs, 1 H), 8.41 (bs, 1 H), 8.30 (t, $J$ = 6.0 Hz, 1 H), 8.18 (t, $J$ = 5.4 Hz, 1 H), 7.77 (s, 1 H), 7.41 (d, $J$ = 7.8 Hz, 1 H), 7.01 (d, $J$ = 3.0 Hz, 1 H), 6.99 (d, $J$ = 2.4 Hz, 1 H), 5.30 (d, $J$ = 1.8 Hz, 1 H), 5.26 (d, $J$ = 8.4 Hz, 1 H), 4.28 (d, $J$ = 12.0 Hz, 1 H), 3.78 – 3.74 (m, 1 H), 3.50 – 3.45 (m, 1 H), 3.25 – 3.22 (m, 1 H), 2.82 (d, $J$ = 4.2 Hz, 1 H), 2.61 – 2.58 (m, 1 H), 1.98 – 1.94 (m, 1 H). One proton signal is missing, it is assumed to be under the water signal (3.34) as reported by Quinn (*J. Org. Chem.* **1999**, *64*, 731 – 735).

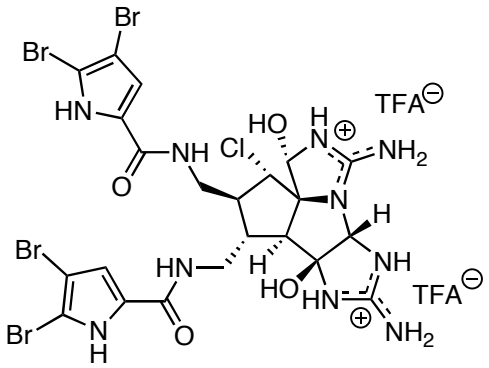
Axinellamine A (**1**) $^1$H-NMR (600 MHz, $CD_3OD$) δ 7.02 (s, 1 H), 6.90 (s, 1 H), 5.47 (s, 1 H), 5.38 (s, 1 H), 4.27 (d, $J$ = 12.0 Hz, 1 H), 3.83 (dd, $J$ = 14.4, 4.2 Hz, 1 H), 3.66 – 3.63 (m, 1 H), 3.48 – 3.43 (m, 2 H), 3.06 (d, $J$ = 5.4 Hz, 1 H), 2.82 – 2.78 (m, 1 H), 2.09 – 2.03 (m, 1 H).

Axinellamine B (**2**) $^1$H-NMR (600 MHz, DMSO-$d_6$) δ 12.69 (s, 1 H), 12.66 (s, 1 H), 10.02 (s, 1 H), 9.76 (s, 1 H), 9.05 (s, 1 H), 8.84 (s, 1 H), 8.70 (s, 1 H), 8.25 (t, $J$ = 6.0 Hz, 1 H), 7.97 (t, $J$ = 5.4 Hz, 1 H), 7.45 (s, 1 H), 7.36 (d, $J$ = 8.4 Hz, 1 H), 6.95 (d, $J$ = 1.8 Hz, 1 H), 6.92 (d, $J$ = 1.8 Hz, 1 H), 5.35 (d, $J$ = 8.4 Hz, 1 H), 5.31 (s, 1 H), 3.98 (d, $J$ = 12.0 Hz, 1 H), 3.63 – 3.60 (m, 1 H), 3.05 (d, $J$ = 4.2 Hz, 1 H), 2.25 – 2.20 (m, 1 H), 2.08 – 2.04 (m, 1 H) Based on the isolation paper (*J. Org. Chem.* **1999**, *64*, 731 – 735), two protons are under the water peak in the DMSO (3.33 and 3.34) and one proton is under the DMSO peak (2.52)

Axinellamine B (**2**) $^1$H-NMR (600 MHz, $CD_3OD$) δ 6.893 (s, 1 H), 6.886 (s, 1 H), 5.58 (s, 1 H), 5.47 (s, 1 H), 3.98 (d, $J$ = 12.0 Hz, 1 H), 3.70 (dd, $J$ = 13.8, 5.4 Hz, 1 H), 3.49 – 3.47 (m, 2 H), 3.42 (dd, $J$ = 9.0, 7.2 Hz, 1 H), 2.38 – 2.34 (m, 1 H), 2.16 – 2.13 (m, 1 H). In $CD_3OD$, one proton signal is absent, it is assumed to be under the methanol peak (3.30)

## Comparison of Spectroscopic Data

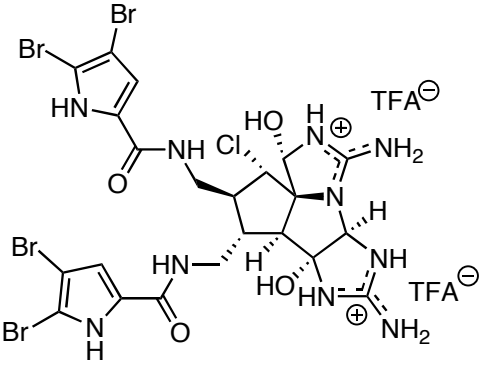
Reported Isolation: S. Urban, P. de A. Leone, A. R. Carroll, G. A. Fechner, J. Smith, J. N. A. Hooper, R. J. Quinn, *J. Org. Chem.* **1999**, *64*, 731 – 735.



**axinellamine A**

**$^1$H-NMR (DMSO-*d* $_6$)** (δ, multiplicity, *J* (Hz), inegration)

| Reported (Isolation by Quinn) | Observed (Synthetic) |
|---|---|
| 1.96, dddd, 4.2/4.2/9.6/12.0, 1 H | 1.98 - 1.94, m, 1 H |
| 2.60, ddd, 4.2/5.4/9.6, 1 H | 2.61 -2.58, m, 1 H |
| 2.84, d, 4.2, 1 H | 2.82, d, 4.2, 1 H |
| 3.24, ddd, 4.2/4.2/15.0 1H | 3.25 - 3.22, m, 1 H |
| 3.34, m, 1 H | under water signal |
| 3.47, m, 1 H | 3.50 - 3.45, m, 1 H |
| 3.74, ddd, 4.2/7.8/15.0, 1 H | 3.78 - 3.74, m, 1 H |
| 4.28, d, 12.0, 1 H | 4.28, d, 12.0, 1 H |
| 5.27, d, 7.2, 1 H | 5.26, d, 8.4, 1 H |
| 5.29, s, 1 H | 5.30, d, 1.8, 1 H |
| 6.97, s, 1 H | 6.99, d, 2.4, 1 H |
| 6.99, s, 1 H | 7.01, d, 3.0, 1 H |
| 7.39, d, 7.2, 1 H, (1-OH) | 7.41, d, 7.8, 1 H |
| 7.76, s, 1 H, (9-OH) | 7.77, s, 1 H |
| 8.14, t, 5.4, 1 H (2"-NH) | 8.18, t, 5.4, 1 H |
| 8.26, t, 5.4, 1 H (2'-NH) | 8.30, t, 6.0, 1 H |
| 8.41, bs, 2 H, (7-$NH_2$) | 8.41, bs, 1 H |
| 8.68, bs, 1 H (3-$NH_2$) | 8.69, bs, 1 H |
| 8.96, s, 1 H (6-NH) | 8.94, s, 1 H |
| 8.99, bs, 1 H (3-$NH_2$) | 9.00, bs, 1 H |
| 9.67, s, 1 H (8-NH) | 9.67, s, 1 H |
| 9.88, s, 1 H (2-NH) | 9.88, s, 1 H |
| 12.67, s, 1 H (5"-NH) | 12.71, d, 2.4, 1 H |
| 12.69, s, 1 H (5'-NH) | 12.73, d, 2.4, 1 H |

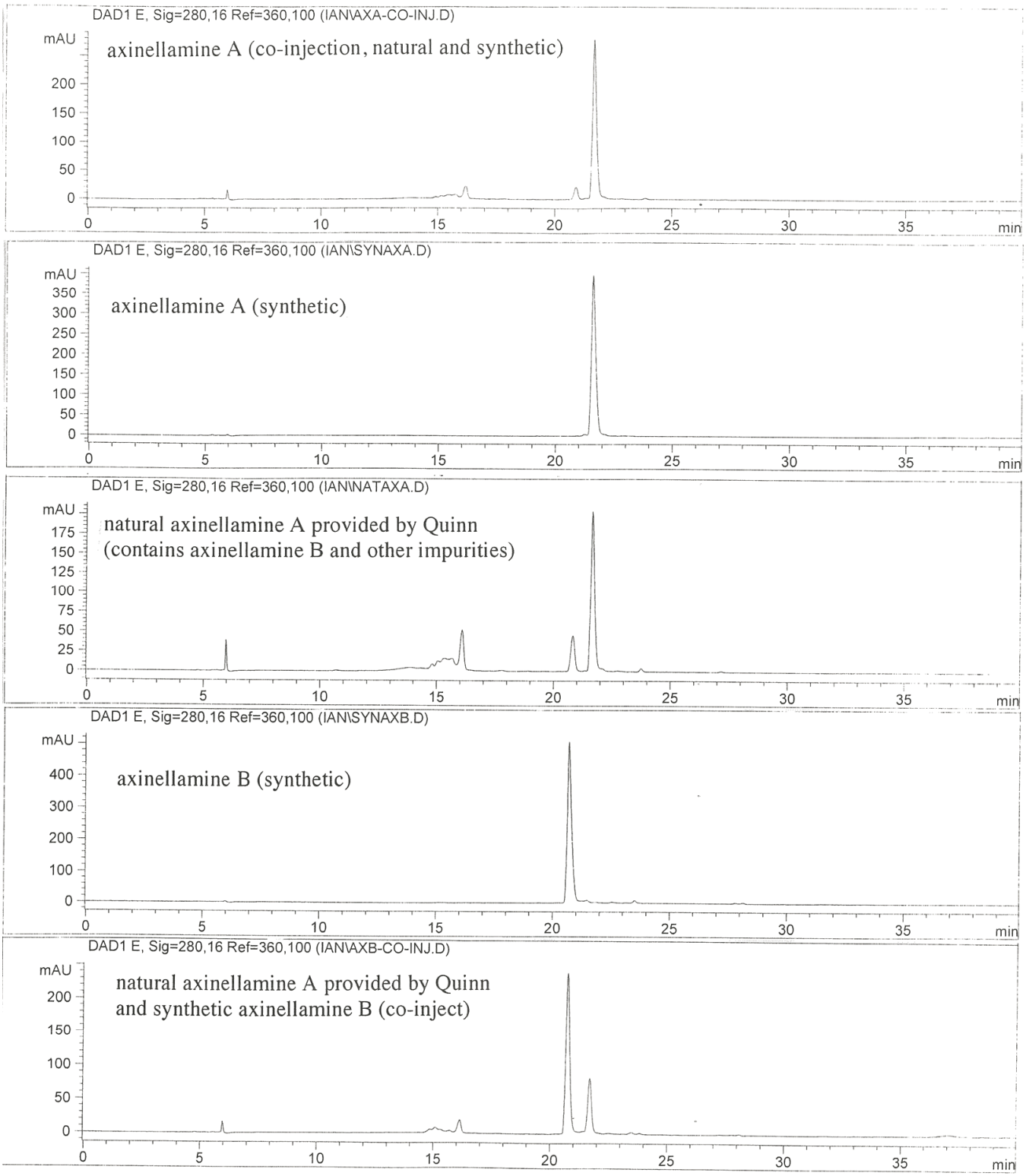


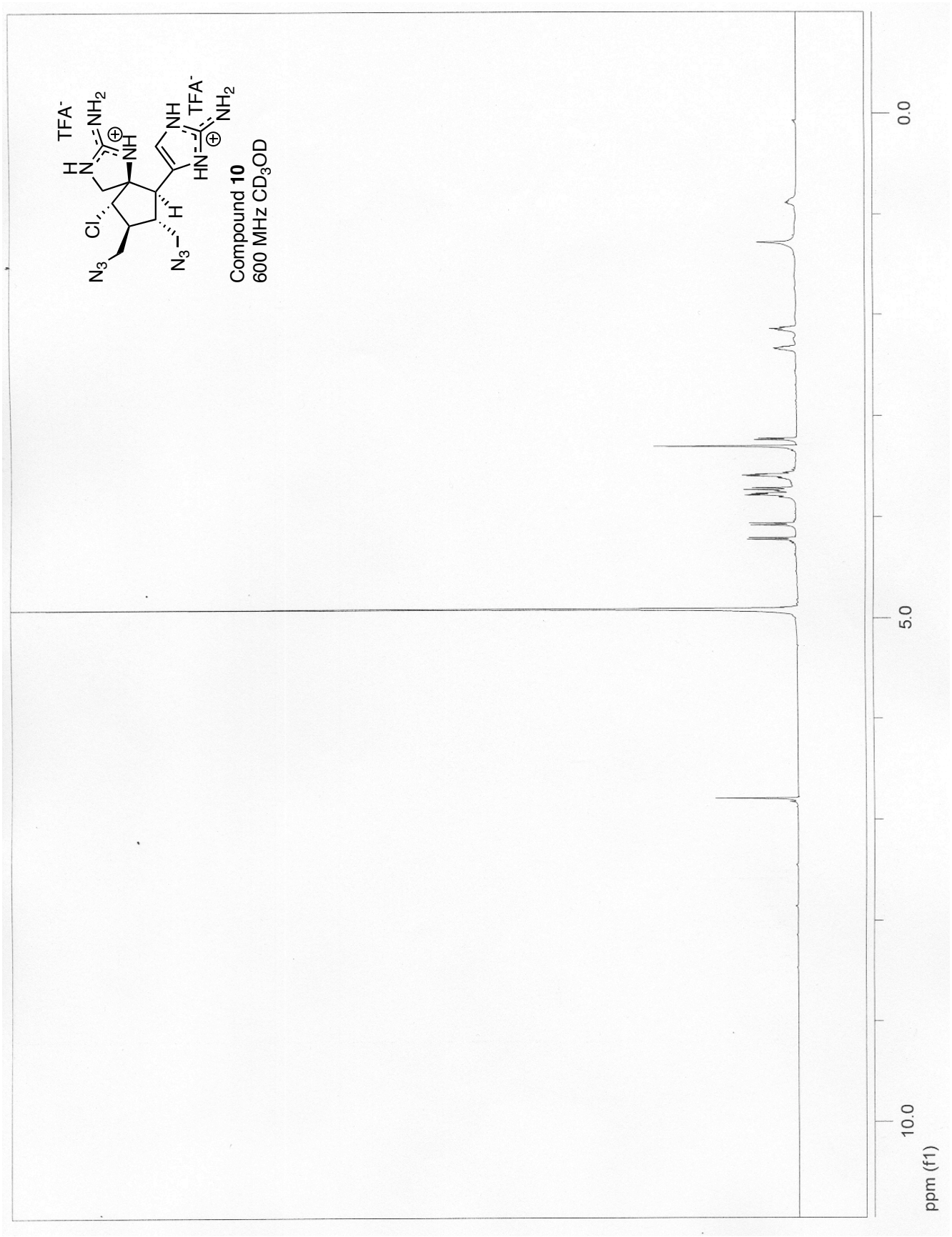
**axinellamine B**

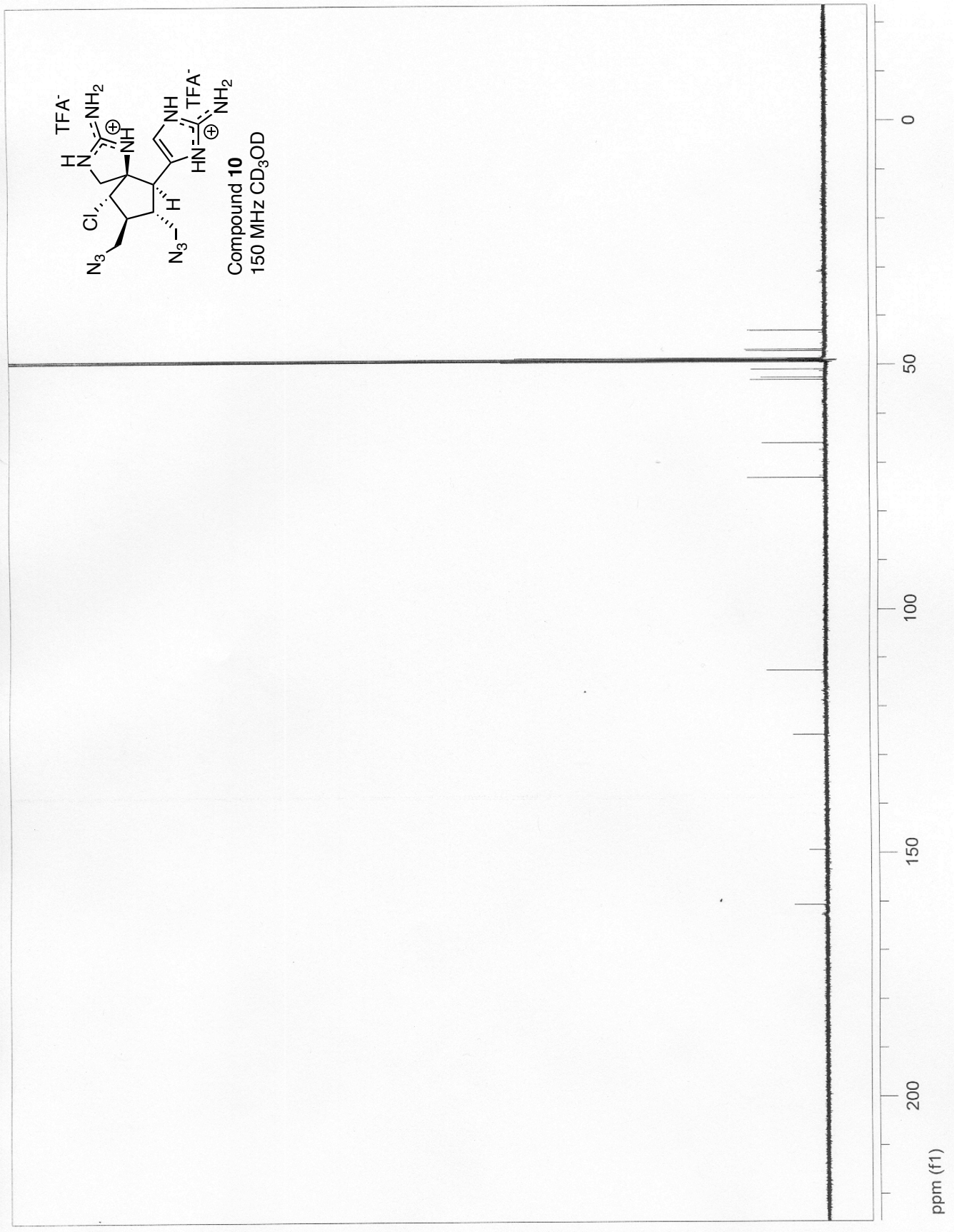
**$^1$H-NMR ( 600 MHz, DMSO-*d* $_6$)** (δ, multiplicity, *J* (Hz), inegration)

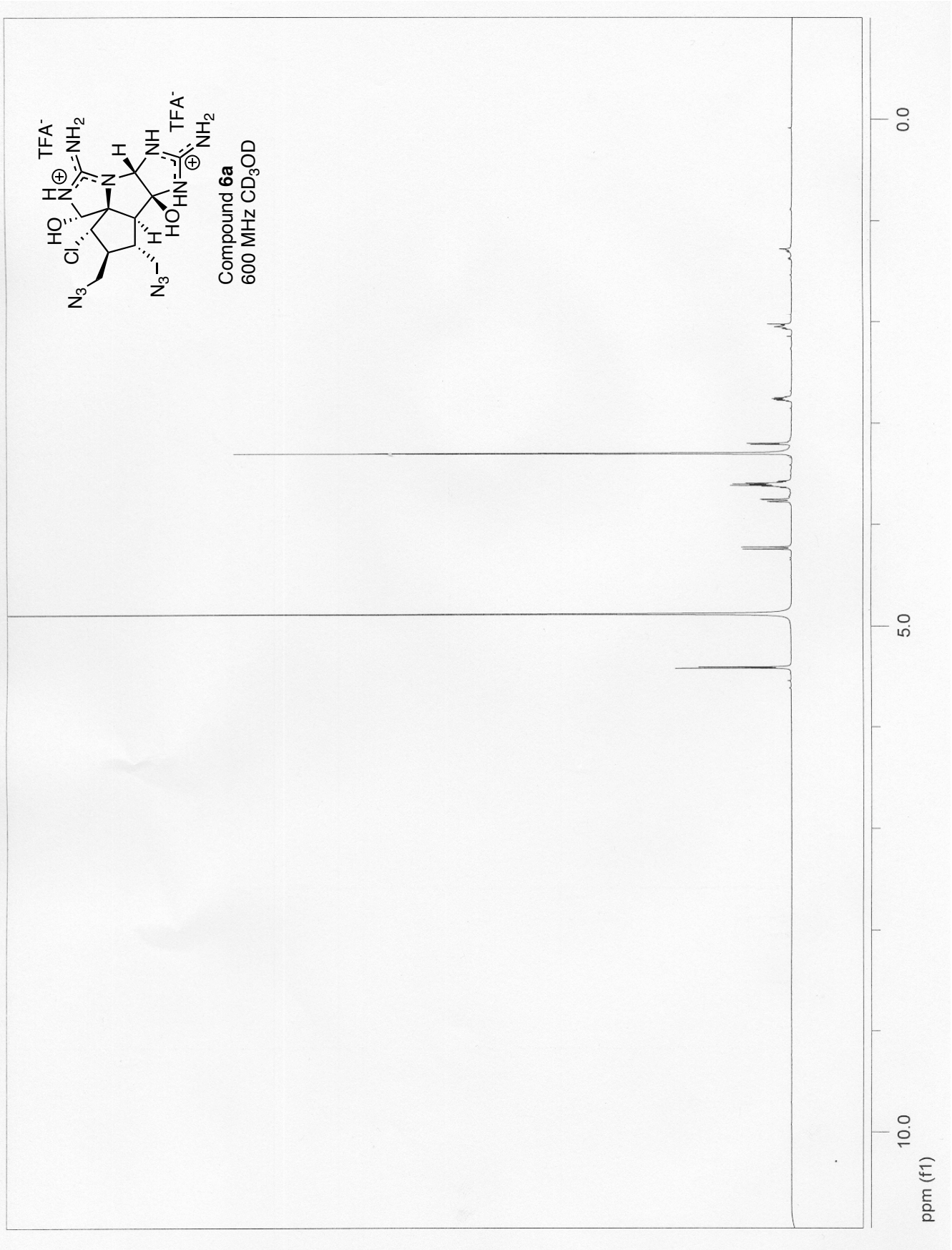
| Reported (Isolation by Quinn) | Observed (Synthetic) |
|---|---|
| 2.06, m, 1 H | 2.08 - 2.04, m, 1 H |
| 2.24, m, 1 H | 2.25 - 2.20, m, 1 H |
| 2.52, m, 1 H | under DMSO signal |
| 3.05, d, 3.6, 1 H | 3.05, d, 4.2, 1 H |
| 3.33, m, 1 H | under water signal |
| 3.34, m, 1 H | under water signal |
| 3.60, m, 1 H | 3.63 - 3.60, m, 1 H |
| 3.97, d, 12.0, 1 H | 3.98, d, 12.0, 1 H |
| 5.32, s, 1 H | 5.31, s, 1 H |
| 5.35, d, 7.0, 1 H | 5.35, d, 8.4, 1 H |
| 6.91, s, 1 H | 6.92, d, 1.8, 1 H |
| 6.93, s, 1 H | 6.95, d, 1.8, 1 H |
| 7.37, d, 7.0, 1 H (1-OH) | 7.36, d, 8.4, 1 H |
| 7.44, s, 1 H (9-OH) | 7.45, s, 1 H |
| 7.93, t, 5.4, 1 H (2"-NH) | 7.97, t, 5.4, 1 H |
| 8.22, t, 5.4, 1 H (2'-NH) | 8.25, t, 6.0, 1 H |
| visible in spectrum, not listed | 8.70, s, 1 H |
| 8.77, s, 1 H (6-NH) | 8.84, s, 1 H |
| 9.10, bs, 2 H (3-$NH_2$ and 7-$NH_2$) | 9.05, s, 1 H |
| 9.82, s, 1 H (8-NH) | 9.76, s 1 H |
| 9.85, bs, 2 H (3-$NH_2$ and 7-$NH_2$) | (not present in synthetic or isolated spectra) |
| 10.06, s, 1 H (2-NH) | 10.02, s, 1 H |
| 12.61, s, 1 H (5"-NH) | 12.66, s, 1 H |
| 12.64, s, 1 H (5'-NH) | 12.69, s, 1 H |

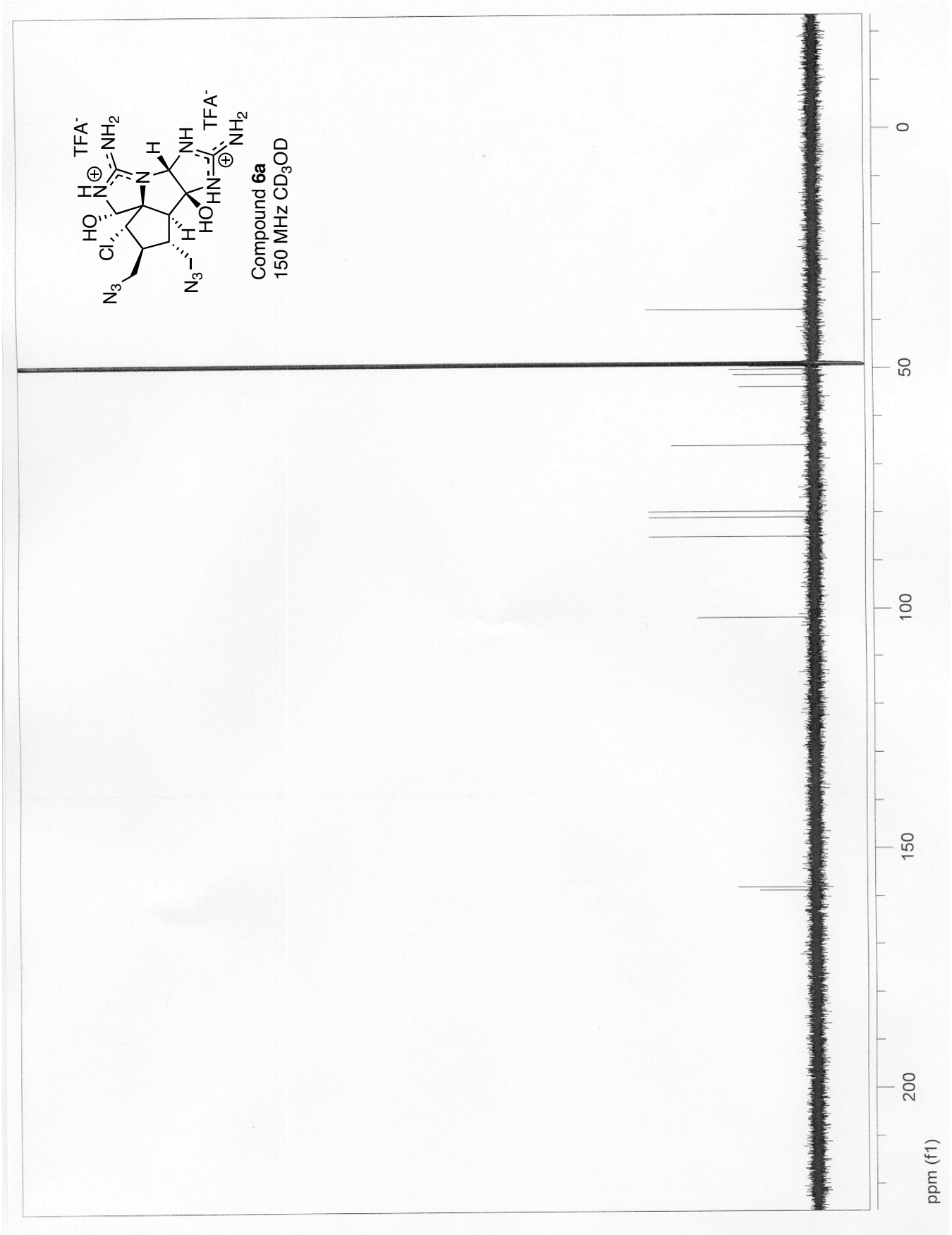
HPLC traces of natural axinellamine A (**1**) provided by Quinn and synthetic axinellamines A (**1**) and B (**2**). Co-injections of each synthetic sample with the natural material are also provided.



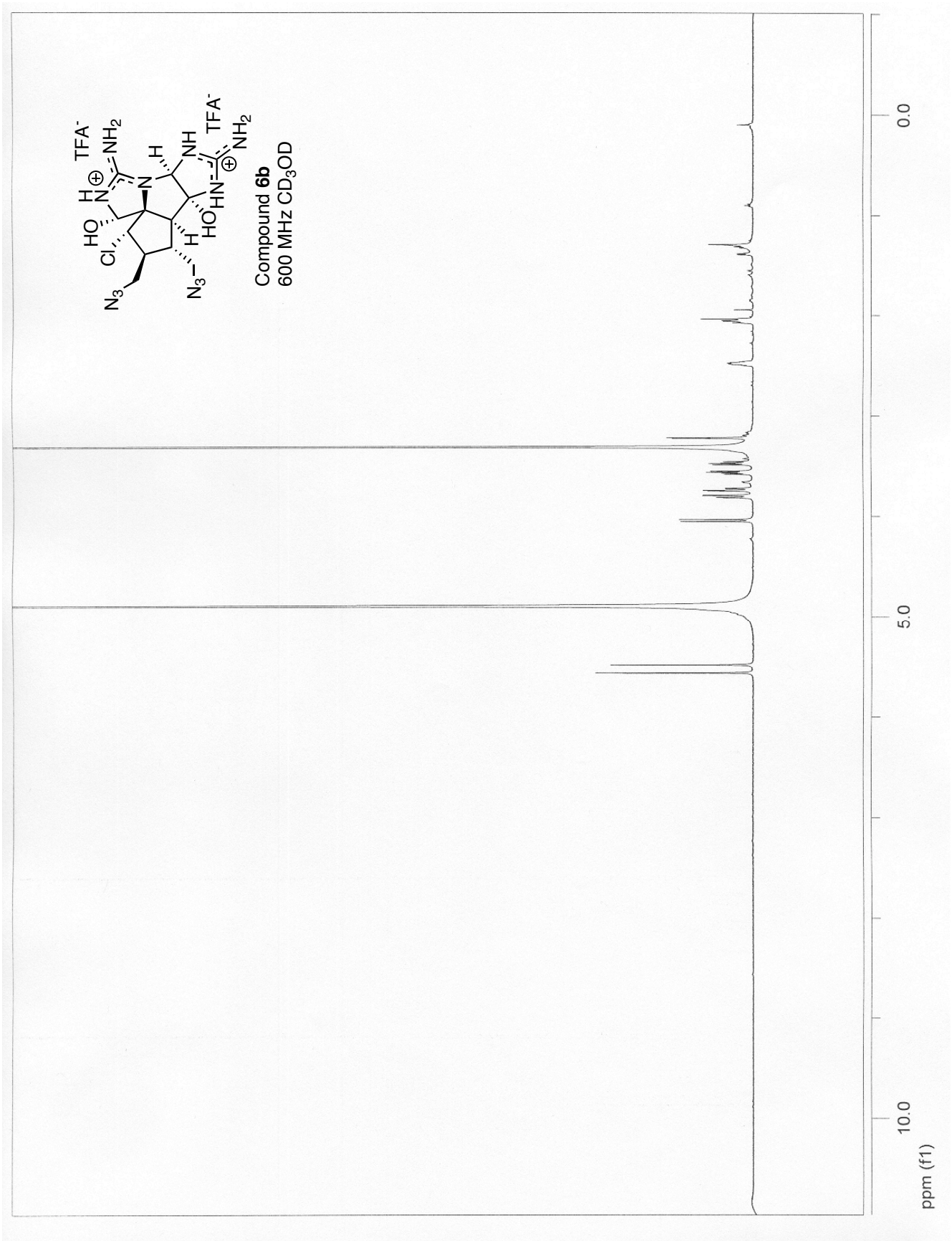

Compound 10
600 MHz CD3OD
10.0
5.0
0.0
ppm (f1)

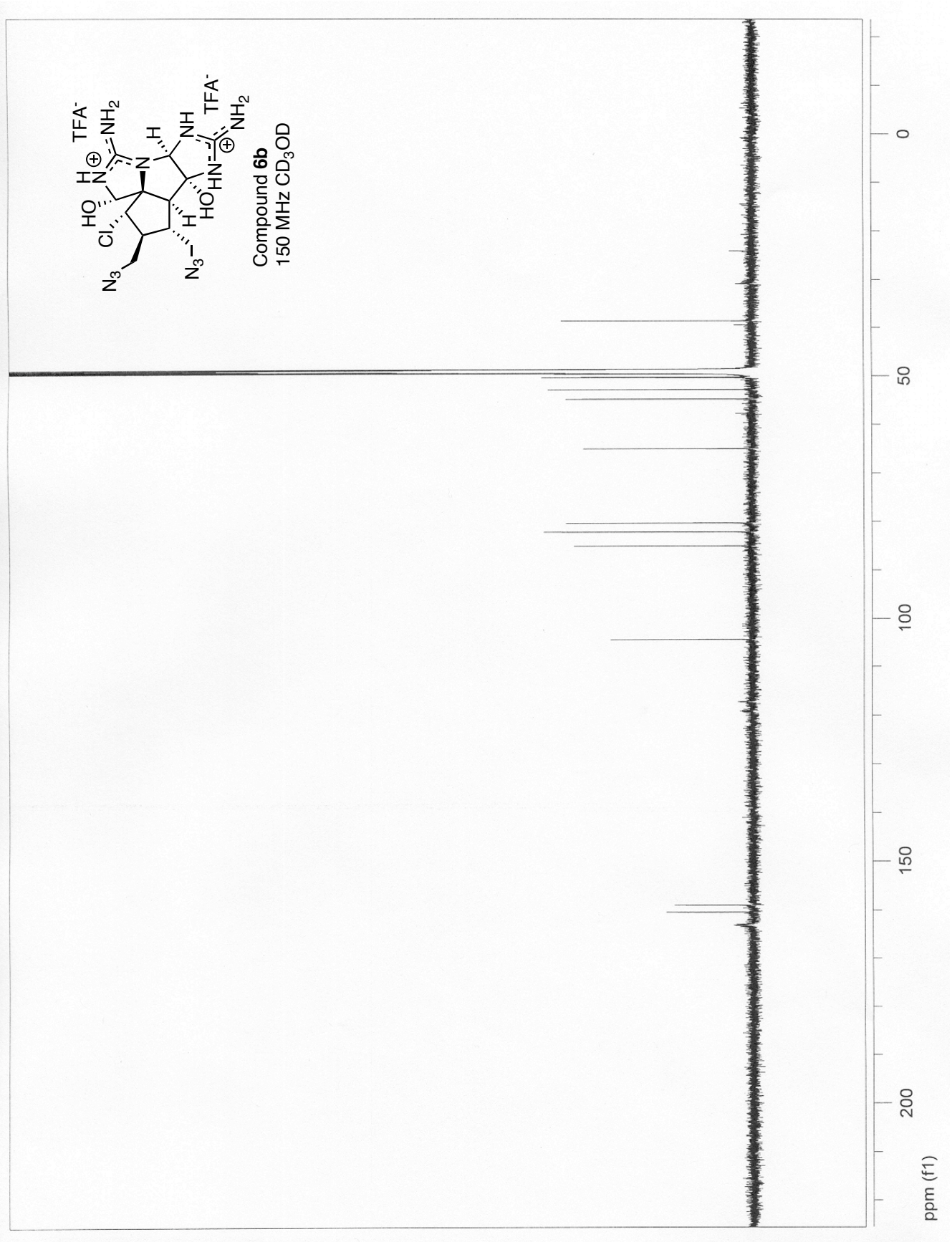

Compound 10
150 MHz CD3OD
ppm (f1)
0
50
100
150
200

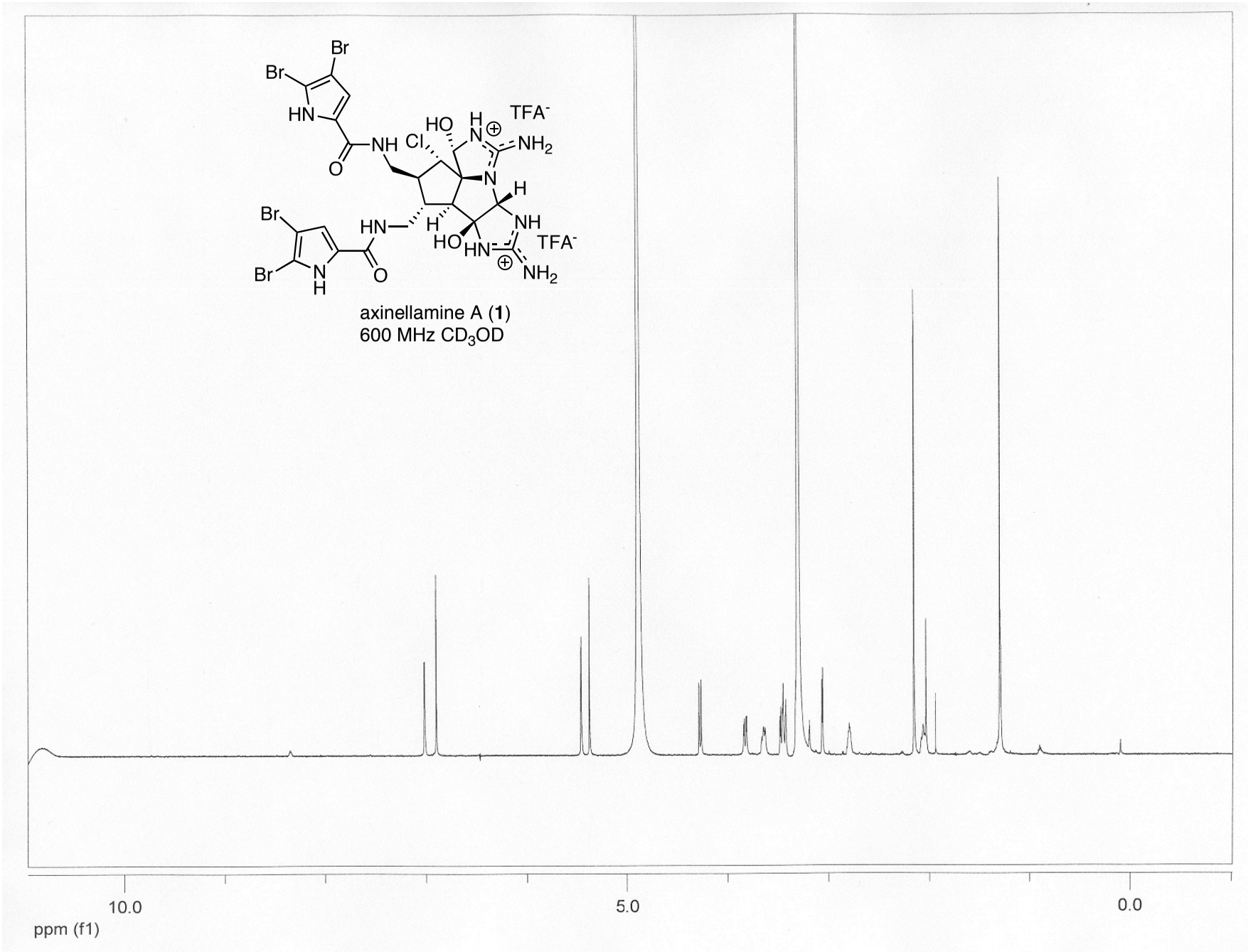

Compound 6a
600 MHz CD3OD
10.0
5.0
0.0
ppm (f1)

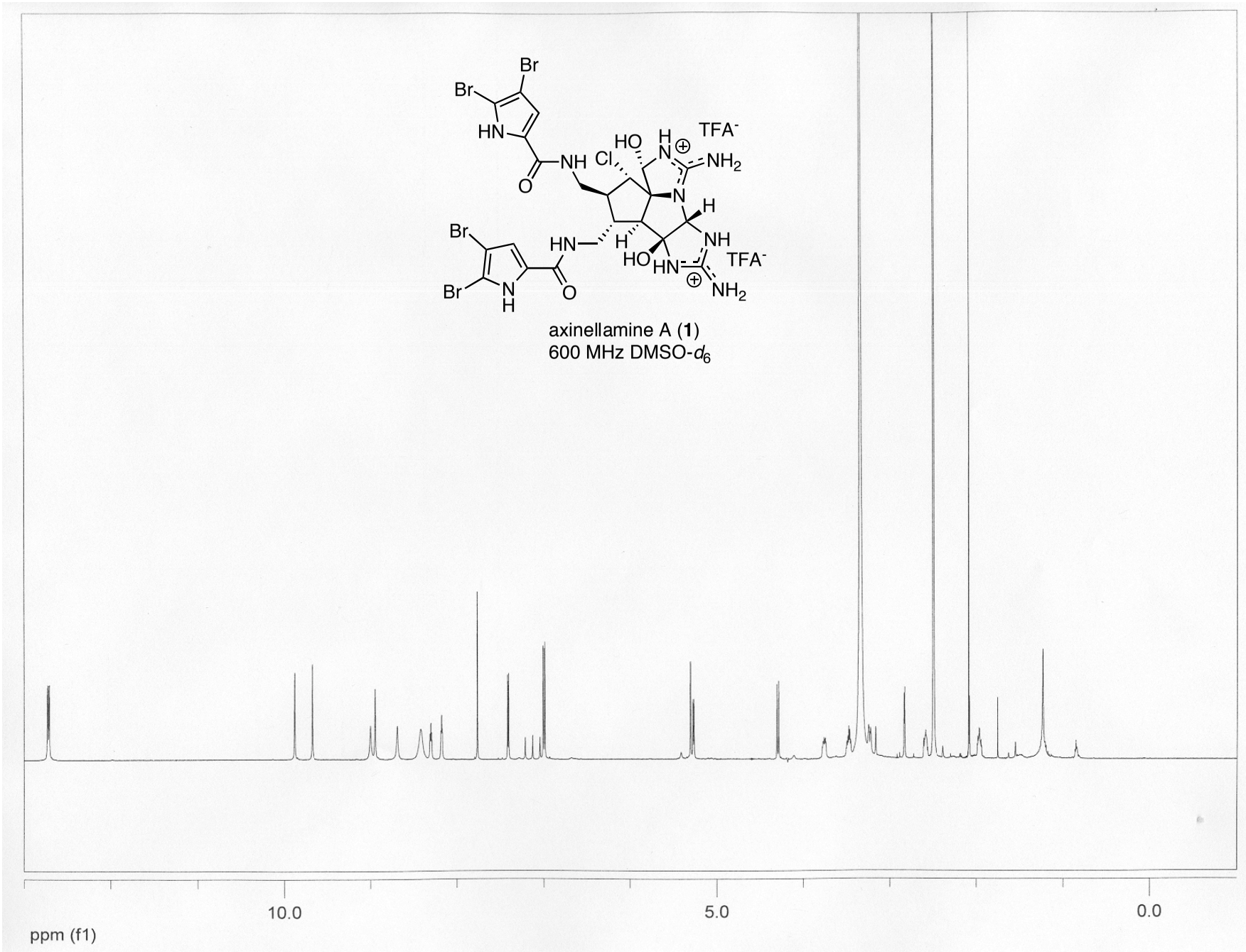

Compound 6a
150 MHz CD3OD
ppm (f1)
0
50
100
150
200

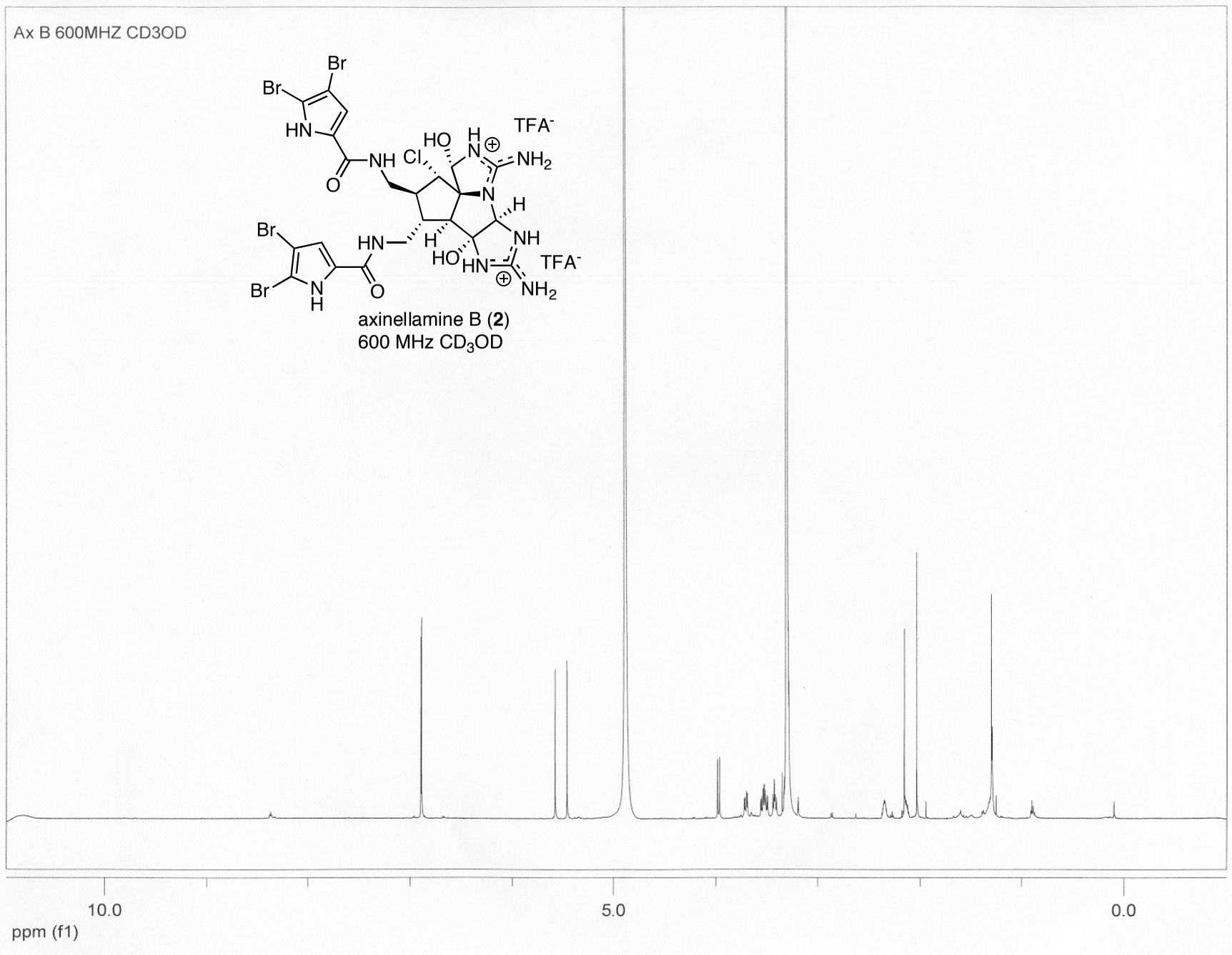

Compound 6a
150 MHz CD3CN
ppm (f1)
0
50
100
150
200

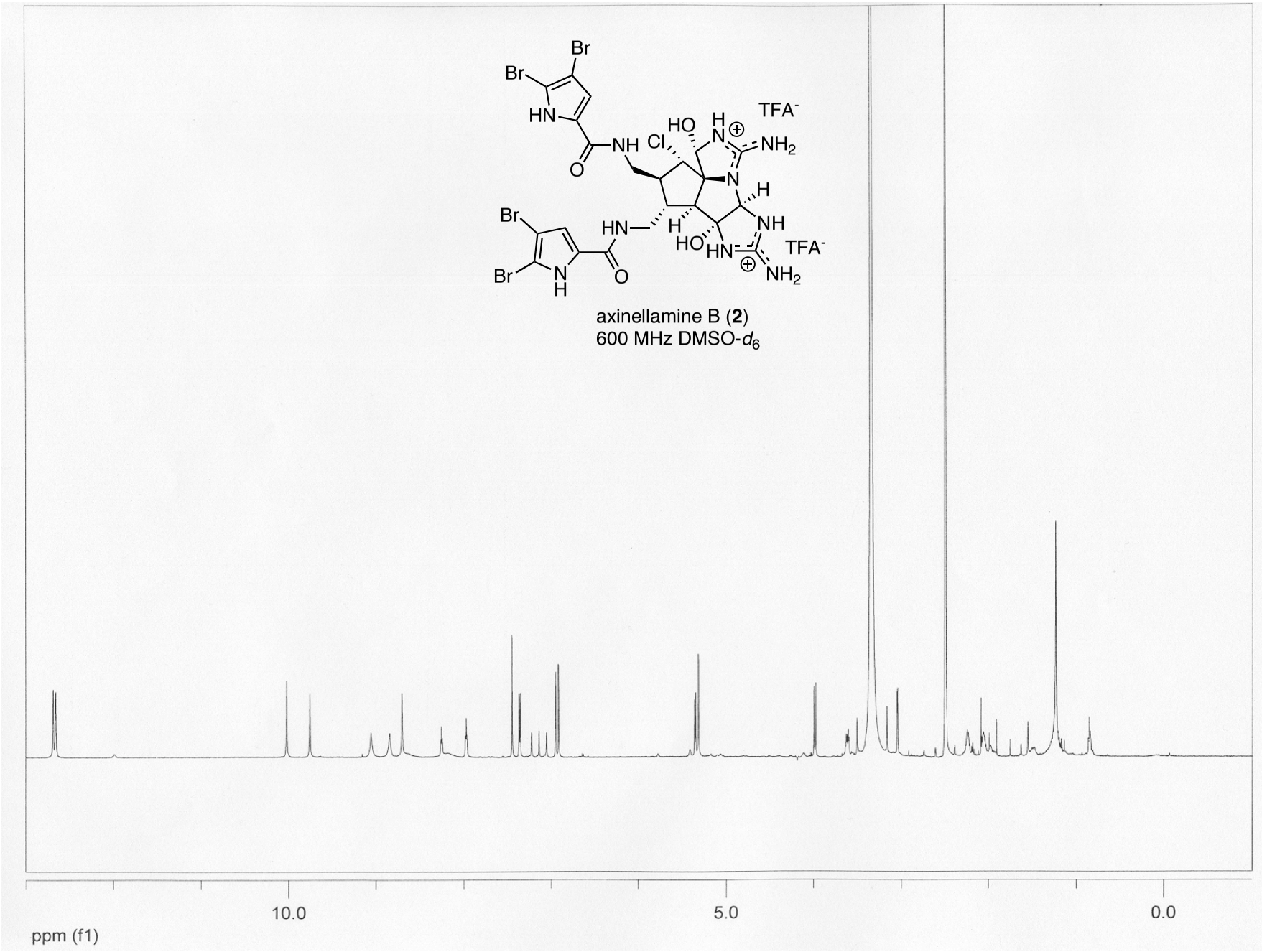

TFA-
NH2
H
N
HO
Cl
N3
NH
HO
H
N3
HN
TFA-
NH2
Compound 6b
600 MHz CD3OD
10.0
5.0
0.0
ppm (f1)


Compound 6b
150 MHz CD3OD
ppm (f1)
0
50
100
150
200


Br
Br
HN
O
NH
HO
Cl
H
N
TFA-
NH2
N
H
Br
HN
H
NH
HO
HN
TFA-
Br
N
H
O
NH2
axinellamine A (1)
600 MHz CD3OD
10.0
5.0
0.0
ppm (f1)

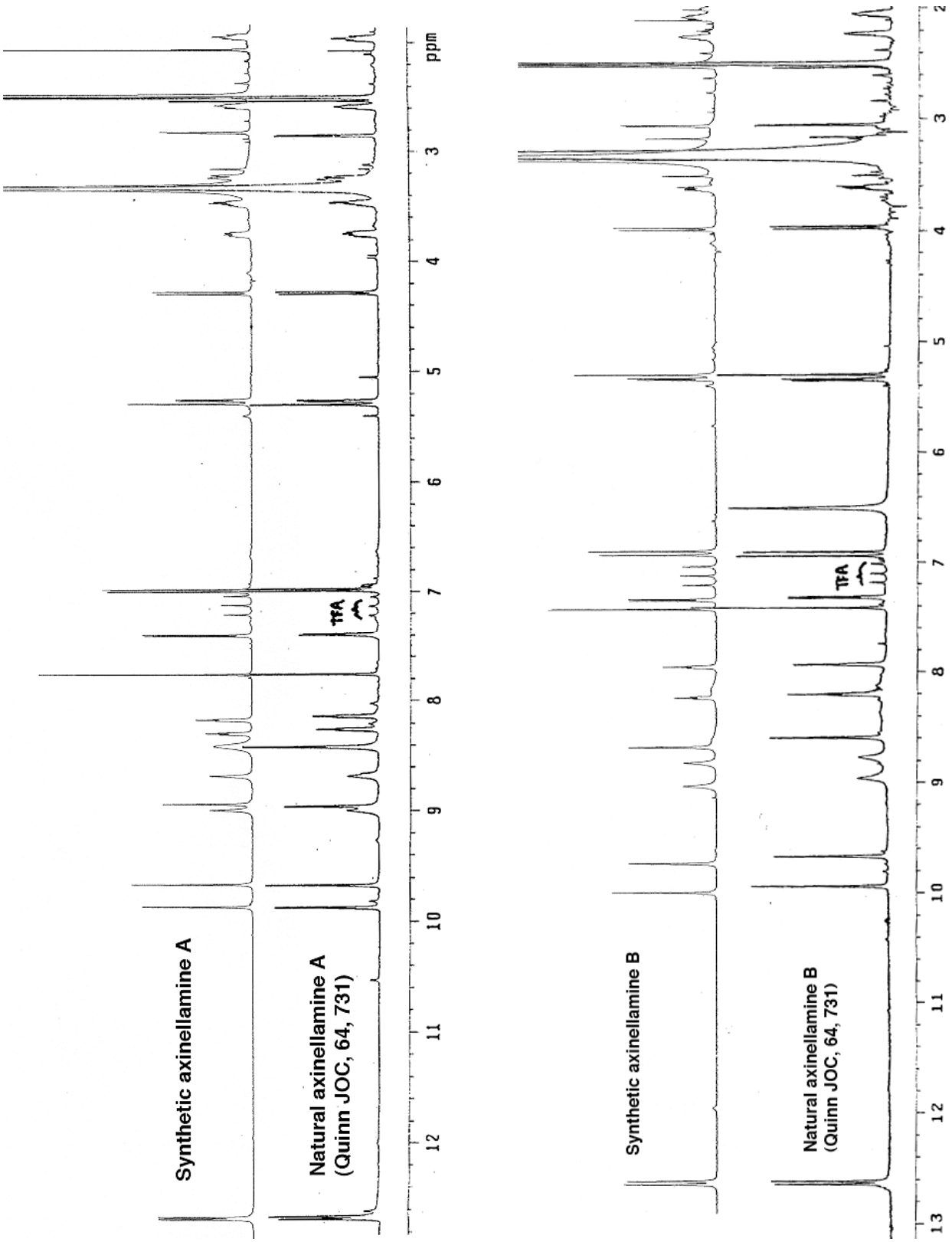

Br
Br
HN
O
NH
Cl
HO
H
N
NH2
TFA-
N
H
Br
HN
H
NH
HO
HN
TFA-
Br
N
H
O
NH2
axinellamine A (1)
600 MHz DMSO-d6
10.0
5.0
0.0
ppm (f1)


Ax B 600MHZ CD3OD
axinellamine B (2)
600 MHz CD3OD
10.0
5.0
0.0
ppm (f1)




axinellamine B (**2**)
600 MHz DMSO-$d_6$

Comparison of natural and synthetic axinellamines A (**1**) and B (**2**) ($^1$H-NMR, DMSO-$d_6$, 600 MHz)



Comparison of synthetic axinellamines A (**1**) and B (**2**) with the natural sample of “axinellamine A” provided by Quinn ($^1$H-NMR, $CD_3OD$, 600 MHz).