Supporting Information

Acyclic Cucurbit[n]uril Dendrimers

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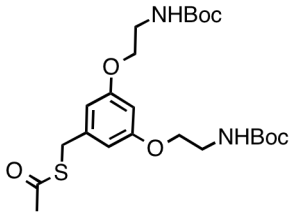
General Experimental Details.

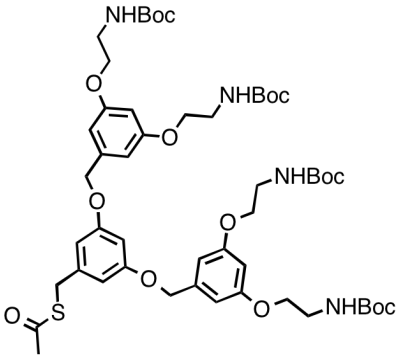
Starting materials were purchased from commercial suppliers were used without further purification. Compounds **D1-Br**[1], **D2-Br**[1], **D3-Br**[1] and **1**[2] were prepared according to the literature procedures. Melting points were measured on a Meltemp apparatus in open capillary tubes and are uncorrected. IR spectra were recorded on a JASCO FT/IR 4100 spectrometer and are reported in cm^{-1}. NMR spectra were measured on spectrometers operating at 400, 500, or 600 MHz for ^{1}H and 125 or 150 MHz for ^{13}C NMR spectra. Routine mass spectrometry was performed using a JEOL AccuTOF electrospray instrument (ESI). High resolution electrospray mass spectra were recorded on a Bruker 12T Apex IV fourier transform ion cyclotron resonance mass spectrometer. Plasmid EGFP-N1 (Clontech) was obtained from *E. coli* using the Endofree Plasmid Maxi Kit (Qiagen) according to the manufacturer's instructions. Scanning electron microscopy (SEM) was done on Hitachi SU-70 Analytical UHR. Size of particles was determined by Dyamic Light Scattering (DLS). Size and ζ potential were measured using a Malvern nanoZS apparatus using the software and instructions included with the instrument.

Synthetic procedures and characterization Data.

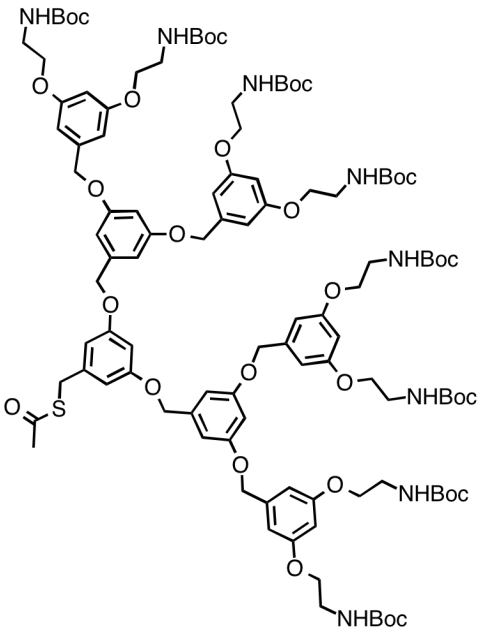


Compound D1-SAc. Thioacetic acid (90 μL, 1.26 mmol) was added to a stirred solution of **D1-Br**[1] (308 mg, 0.63 mmol) and K_2CO_3 (349 mg, 2.52 mmol) in DMF (10 mL), at room temperature, under nitrogen. After 20 h, the mixture was extracted with Et_2O, washed with water, saturated aqueous NaCl, dried (Na_2SO_4) and concentrated. Column chromatography (SiO_2, hexanes/AcOEt 75:25) gave **G1-SAc** (275 mg, 90%). Pale yellow solid. M.p. 96-98 °C. IR (ATR, cm^{-1}): 3361 (N-H), 1684 (C=O, thioester), 1597 (C=O, Boc). ^{1}H NMR (400 MHz, $CDCl_3$): 6.42 (d, J = 2 Hz, 2H), 6.31 (t, J = 2 Hz, 1H), 4.99 (t, J = 5 Hz, 2H), 4.02 (s, 2H), 3.96 (t, J = 5 Hz, 4H), 3.50 (q, J = 5 Hz, 4H), 2.34 (s, 3H), 1.44 (s, 18H). ^{13}C NMR (125 MHz, $CDCl_3$): 195.0, 160.0, 156.0, 140.2,

107.8, 100.4, 79.7, 67.4, 40.2, 33.7, 30.5, 28.5. MS (ESI, positive mode): *m/z* 485 ([M+H]$^+$, calcd. for $C_{23}H_{36}N_2O_7S\bullet H^+$: 485.2321).

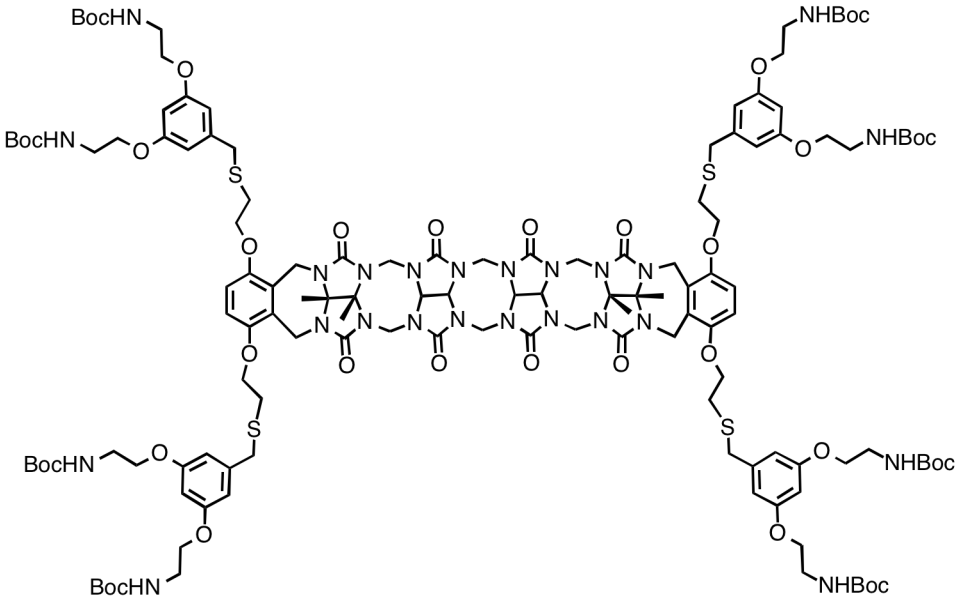


Compound D2-SAc. Thioacetic acid (70 μL, 0.98 mmol) was added to a stirred solution of **D2-Br**[1] (206 mg, 0.20 mmol) and K_2CO_3 (406 mg, 2.94 mmol) in DMF (10 mL), at room temperature under nitrogen. After 20 h, the mixture was extracted with Et_2O, washed with water, saturated aqueous NaCl solution, dried (Na_2SO_4) and concentrated. Column chromatography (SiO_2, hexanes/AcOEt 6:4) gave **D2-SAc** (151 mg, 74%). Pale yellow solid. M.p. 45-47 °C. IR (ATR, cm^{-1}): 3330 (N-H), 1687 (C=O, thioester), 1593 (C=O, Boc). ^{1}H NMR (400 MHz, $CDCl_3$): 6.55 (d, *J* = 2 Hz, 4H), 6.52 (d, *J* = 2 Hz, 2H), 6.46 (t, *J* = 2 Hz, 1H), 6.39 (t, *J* = 2 Hz, 2H), 5.00 (t, *J* = 5 Hz, 4H), 4.92 (s, 4H), 4.05 (s, 2H), 4.02 (t, *J* = 5 Hz, 8H), 3.54 (q, *J* = 5 Hz, 8H), 2.35 (s, 3H), 1.45 (s, 36H). ^{13}C NMR (125 MHz, $CDCl_3$): 195.1, 160.1 (two peaks), 156.0, 140.1, 139.5, 108.2, 106.3, 101.2, 101.1, 79.7, 70.0, 67.5, 40.2, 33.7, 30.4, 28.6. MS (ESI, positive mode): *m/z* 1037 ([M+Na]$^+$, calcd. for $C_{51}H_{74}N_4O_{15}S\bullet Na^+$: 1037.4769).



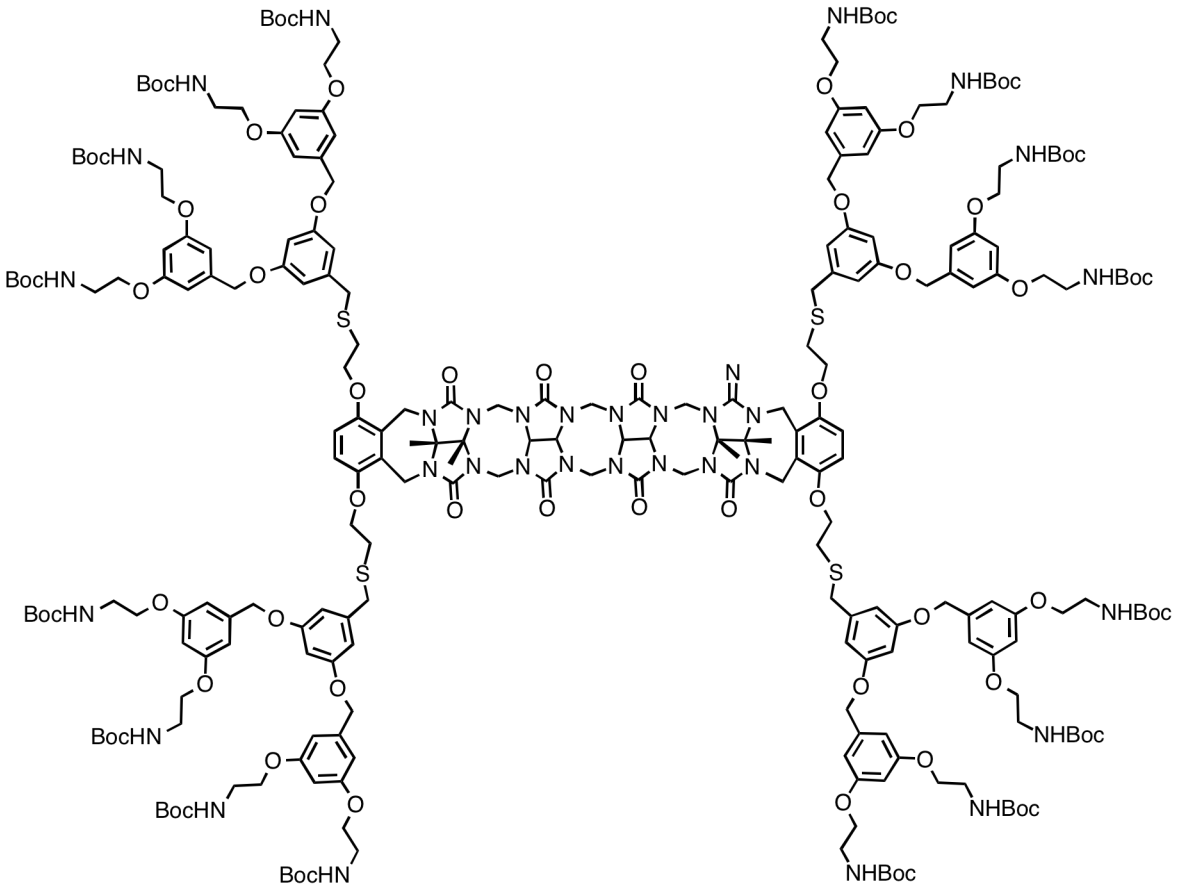
Compound D3-SAc. Thioacetic acid (70 μL, 0.98 mmol) was added to a stirred solution of **D3-Br**[1] (417 mg, 0.20 mmol) and K_2CO_3 (406 mg, 2.94 mmol) in DMF (10 mL), at room temperature, under nitrogen. After 20 h, the mixture was extracted with Et_2O, washed with water and a saturated aqueous NaCl solution, dried (Na_2SO_4) and concentrated. Column chromatography (SiO_2, hexanes/AcOEt 45:55) gave **D3-SAc** (293 mg, 71%). Pale yellow solid. M.p. 60-62 °C. IR (ATR, cm^{-1}): 3334 (N-H), 1689 (C=O, thioester), 1593 (C=O, Boc). ^{1}H NMR (400 MHz, $CDCl_3$): 6.65 (d, *J* = 2 Hz, 4H), 6.56 (d, *J* = 2 Hz, 8H), 6.54 (d, *J* = 2 Hz, 2H), 6.53 (t, *J* = 2 Hz, 2H), 6.48 (t, *J* = 2 Hz, 1H), 6.39 (t, *J* = 2 Hz, 4H), 5.04 (t, *J* = 5 Hz, 8H), 4.95 (s, 8H), 4.94 (s, 4H), 4.05 (s, 2H), 3.99 (t, *J* = 5 Hz, 16H), 3.51 (q, *J* = 5 Hz, 16H), 2.34 (s, 3H), 1.44 (s, 72H). ^{13}C NMR (125

MHz, $CDCl_3$): 195.1, 160.2, 160.1, 156.0, 140.1, 139.5, 139.4 108.2, 106.6, 106.2, 101.8, 101.3, 101.0, 79.7, 70.1 (two peaks), 67.5, 40.2, 33.7, 30.4, 28.6. MS (MALDI-TOF): *m/z* 2098 ($[M+Na]^+$, calcd. for $C_{107}H_{150}N_8O_{31}S•Na^+$: 2098.0025).

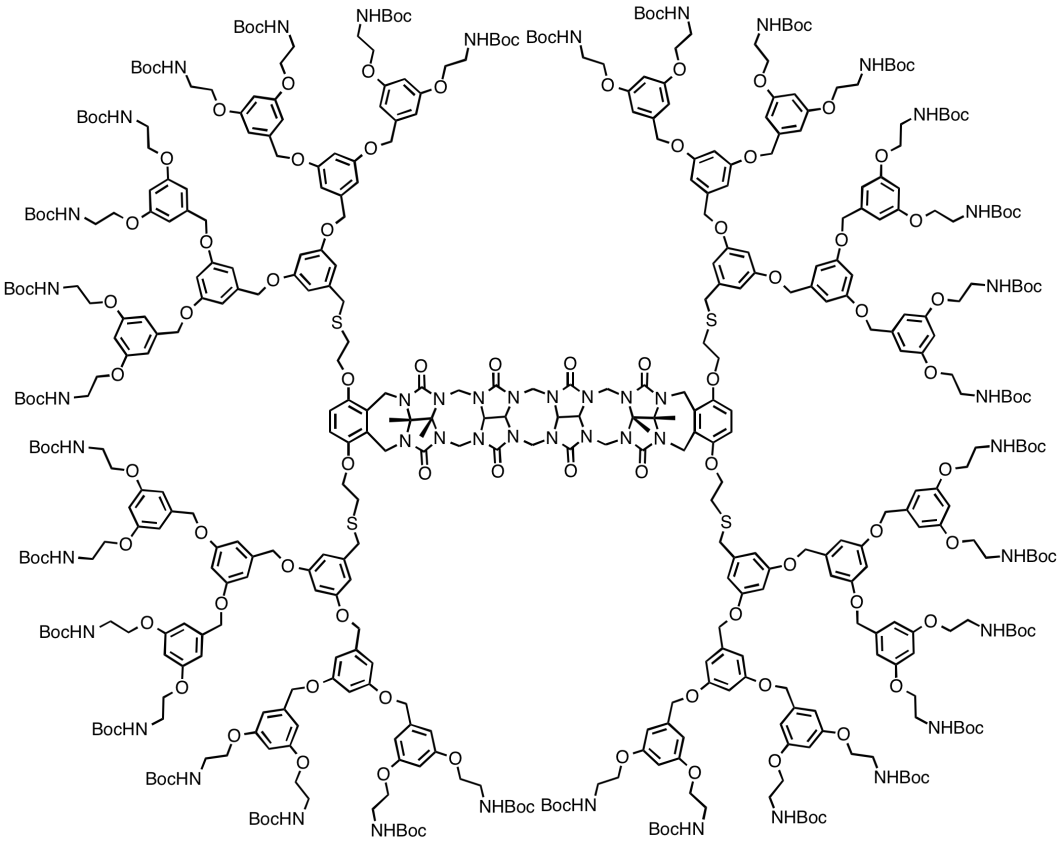


Compound G1-Boc.

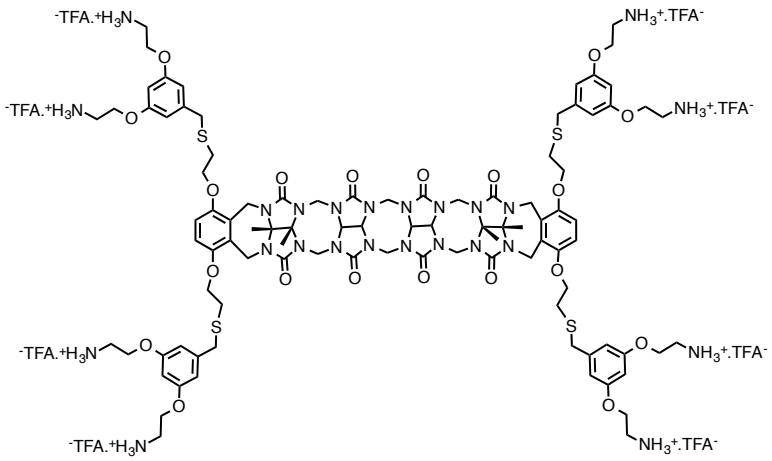
A mixture of **D1-SAc** (100 mg, 0.21 mmol), **1**[2] (57 mg, 0.04 mmol) and K_2CO_3 (569 mg, 4.12 mmol) in ethanol (5 mL) was bubbled with nitrogen for 20 minutes. Then the mixture was stirred at room temperature, under nitrogen, for 4 hours. The mixture was precipitated with water and centrifuged. The solid was dissolved in a minimum amount of EtOAc and precipitated with hexanes and centrifuged. This process was repeated until the excess of thiol was completely removed (as judged by TLC). The precipitate was dried under high vacuum to give **G1-Boc** (65 mg, 56%). White solid. M.p 160-162 °C. IR (ATR, cm^{-1}): 3338 (N-H), 1691 (C=O, ureidyl), 1593 (C=O, Boc). ^1H NMR (400 MHz, DMSO-*d6*): 6.96 (t, *J* = 5 Hz, 8H), 6.80 (s, 4H), 6.54 (d, *J* = 2 Hz, 8H), 6.35 (d, *J* = 2 Hz, 4H), 5.62-5.26 (m, 14H), 4.28-3.78 (m, 26H), 3.91 (t, *J* = 5 Hz, 16H), 3.26 (q, *J* = 5 Hz, 16H), 2.84 (t, *J* = 6 Hz, 8H), 1.68 (s, 6H), 1.64 (s, 6H), 1.37 (s, 72H). ^{13}C NMR (125 MHz, DMSO-d_6): 159.4, 155.6, 155.3, 153.9, 150.1, 141.1, 128.1, 114.7, 107.6, 99.7, 77.7, 77.3, 76.2, 71.0, 70.5, 69.8, 66.4, 66.3, 53.3, 48.4, 35.9, 34.5, 30.8, 28.2, 16.4, 15.4. MS (ESI, positive mode): *m/z* 1437 ($[M-2H+K]^{2+}$, calcd for $C_{134}H_{186}N_{24}O_{36}S_4K^{2+}$: 1437).



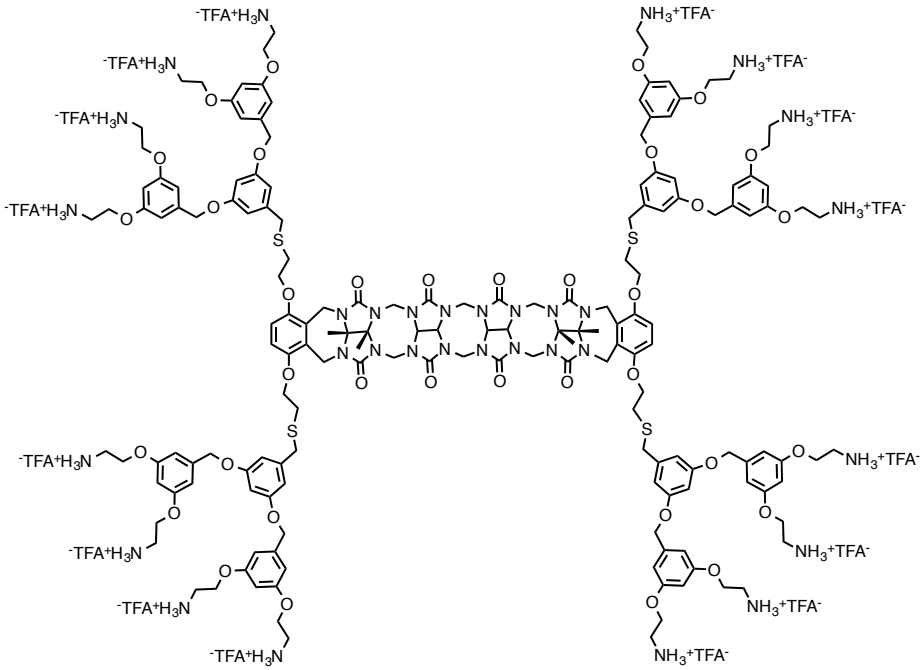
Compound G2-Boc. A mixture of **G2-SAc** (127 mg, 0.13 mmol), **1**[2] (40 mg, 0.03 mmol) and K_2CO_3 (400 mg, 2.89 mmol) in ethanol (5 mL) was bubbled with nitrogen for 20 minutes. Then the mixture was stirred at room temperature, under nitrogen, for 3 days. The mixture was precipitated with water and centrifuged. The solid was dissolved in a minimum amount of EtOAc and precipitated with hexanes and centrifuged. This process was repeated until the excess of thiol was completely removed (as judged by TLC). The precipitate was dried under high vacuum to give **G2-Boc** (89 mg, 62%). White solid. M.p. 138-140 °C. IR (ATR, cm^{-1}): 3338 (N-H), 1691 (C=O, ureidyl), 1591 (C=O, Boc). ^{1}H NMR (500 MHz, DMSO-*d6*): 6.96 (t, *J* = 5 Hz, 16H), 6.82 (s, 4H), 6.65 (m, 8H), 6.55 (m, 16H), 6.49 (m, 4H), 6.41 (m, 8H), 5.67-5.23 (m, 14H), 4.93 (s, 16H), 4.25-3.78 (m, 26H), 3.91 (t, *J* = 5 Hz, 32H), 3.25 (q, *J* = 5 Hz, 32H), 2.85 (m, 8H), 1.57 (broad s, 6H), 1.53 (broad s, 6H), 1.36 (s, 144H). ^{13}C NMR (125 MHz, DMSO-*d6*): 159.6, 159.3, 155.6, 155.3, 153.9, 150.1, 141.2, 139.3, 128.1, 114.7, 107.8, 106.2, 100.5, 100.3, 77.7, 77.3, 76.1, 70.9, 70.5, 70.0, 69.1, 66.5, 53.3, 48.3, 36.0, 34.5, 30.9, 28.2, 16.3, 15.3. MS (ESI, positive mode): *m/z* 841 (100%, $[M+8H+2K]^{6+}$, calcd for $C_{246}H_{348}N_{32}O_{68}S_4K_2^{6+}$: 841).



Compound G3-Boc. A mixture of **G3-SAc** (200 mg, 96.3 μmol), **1**[2] (30.5 mg, 21.9 μmol) and K_2CO_3 (300 mg, 2.17 mmol) in ethanol (5 mL) was bubbled with nitrogen for 20 minutes. Then the mixture was stirred at room temperature, under nitrogen, for 4 days. The mixture was precipitated with water and centrifuged. The solid was dissolved in a minimum amount of EtOAc and precipitated with hexanes and centrifuged. This process was repeated until the excess of thiol was completely removed (as judged by TLC). The precipitate was dried under high vacuum to give **G3-Boc** (144 mg, 71%). White solid. M.p. 104-106 °C. IR (ATR, cm^{-1}): 3319 (N-H), 1691 (C=O, ureidyl), 1593 (C=O, Boc). ^{1}H NMR (400 MHz, DMSO-d_6): 6.94 (t, J = 5 Hz, 32H), 6.83 (s, 4H), 6.68 (m, 16H), 6.55 (m, 52H), 6.40 (m, 16H), 5.62-5.19 (m, 14H), 4.94 (s broad, 48H), 4.26-3.80 (m, 26H), 3.90 (t, J = 5 Hz, 64H), 3.25 (q, J = 5 Hz, 64H), 2.87 (m, 8H), 1.49 (broad s, 6H), 1.46 (broad s, 6H), 1.34 (s, 288H). ^{13}C NMR (150 MHz, DMSO-*d6*): 159.6, 159.4, 158.6, 155.6, 154.9, 153.9, 150.2, 141.2, 139.2, 128.1, 114.8, 107.8, 106.6, 106.1, 101.1, 100.4, 100.3, 77.7, 77.2, 76.1, 70.9, 70.4, 70.0, 69.3, 69.1, 66.8, 66.5, 53.1, 48.3, 36.1, 34.6, 30.7, 28.2, 16.2, 15.2. MS (ESI, positive mode): *m/z* 855 ($[M+7H+5K]^{11+}$, calcd for $C_{470}H_{651}N_{48}O_{132}S_4K_5^{11+}$: 855).

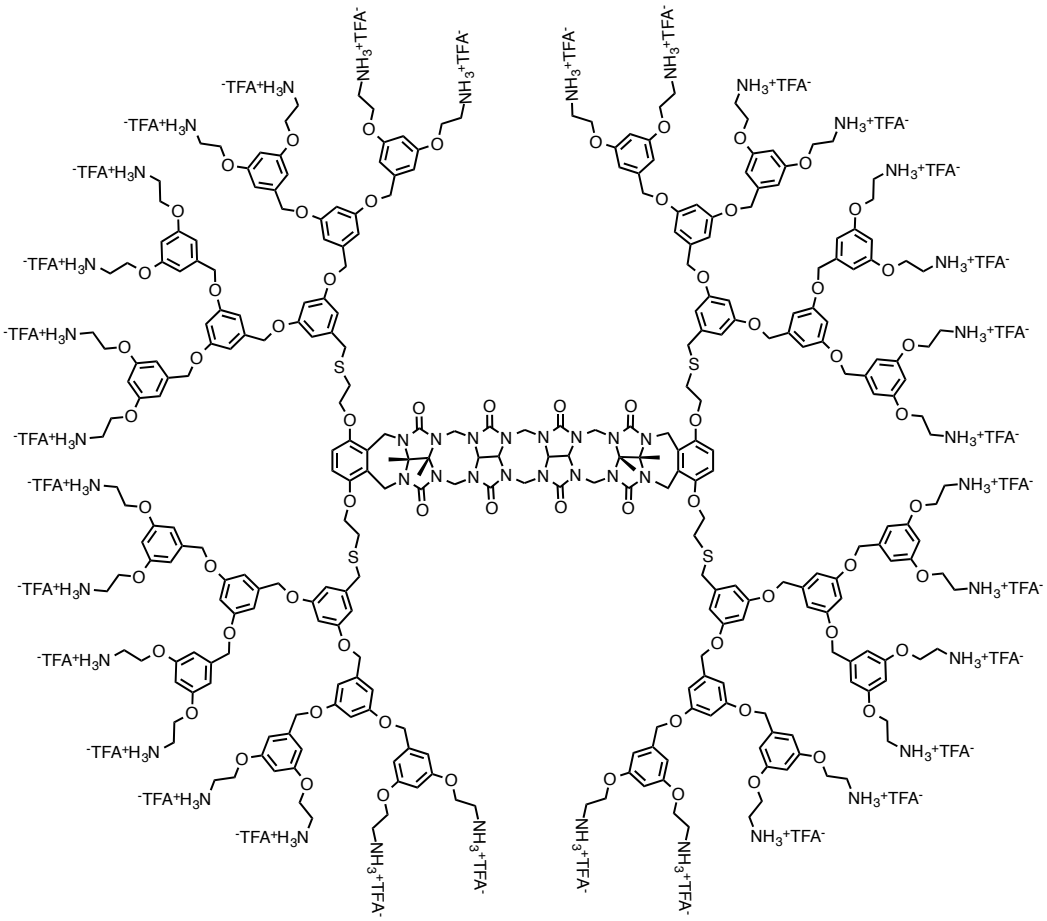


Compound G1. G1-Boc (79 mg, 27.8 μmol) was dissolved in TFA (1 mL). After 20 minutes, TFA was removed by rotary evaporation then under high vacuum to give **G1** as its TFA salt (82 mg, quantitative). Colorless solid. M.p. 108-110 °C. IR (ATR, cm^{-1}): 3439 (N-H), 1672 (C=O, ureidyl), 1593 (C=O, TFA). ^{1}H NMR (600 MHz, DMSO-*d6*, 70°C): 7.93 (broad s, amines), 6.88 (s, 4H), 6.67 (d, *J* = 2 Hz, 8H), 6.48 (t, *J* = 2 Hz, 4H), 5.63 (d, *J* = 16 Hz, 2H), 5.54 (d, *J* = 16 Hz, 4H), 5.47 (d, *J* = 9 Hz, 2H), 5.37 (d, *J* = 9 Hz, 2H), 5.30 (d, *J* = 16 Hz, 4H), 4.24-3.98 (m, 34H), 3.86 (dd, *J* = 36.5 Hz, *J* = 13.5 Hz, 8H), 3.23 (m, 16H), 2.90-2.79 (m, 8H), 1.73 (s, 6H), 1.65 (s, 6H). ^{13}C NMR (125 MHz, D_2O, dioxane as internal reference): 163.4 (q, *J* = 35.5 Hz), 159.7, 157.3, 156.4, 151.2, 142.1, 129.7, 117.1 (q, *J* = 291.5 Hz), 115.8, 109.0, 101.4, 79.3, 78.1, 72.1, 70.9, 64.8, 53.5, 49.4, 39.7, 36.6, 35.6, 31.1, 16.7, 16.0. HR-MS (ESI, positive mode): *m/z* 2038.8325 (3%, $[M+H]^+$, calcd for $C_{94}H_{125}N_{24}O_{20}S_4^+$: 2038.8413), 1019.9271 (100%, $[M+2H]^{2+}$, calcd for $C_{94}H_{126}N_{24}O_{20}S_4^{2+}$: 1019.9246), 680.2860 (85%, $[M+3H]^{3+}$, calcd for $C_{94}H_{127}N_{24}O_{20}S_4^{3+}$: 680.2857), 510.4642 (23%, $[M+4H]^{4+}$, calcd for $C_{94}H_{128}N_{24}O_{20}S_4^{4+}$: 510.4662).



Compound G2. G2-Boc (45 mg, 9 μmol) was dissolved in TFA (1 mL). After 20 minutes, TFA was removed by rotary evaporation then under high vacuum to give **G2** as its TFA salt (45 mg, quantitative).

Colorless solid. M.p. 154-156 °C. IR (ATR, cm^{-1}): 3439 (N-H), 1672 (C=O, ureidyl), 1593 (C=O, TFA). ^{1}H NMR (600 MHz, DMSO-d_6, 70 °C): 8.04 (broad s, amines), 6.93 (s, 4H), 6.73-6.63 (m, 24H), 6.59-6.50 (m, 12H), 5.75-5.25 (m, 14H), 5.03 (s, 16H), 4.24-4.06 (m, 18H), 4.18 (t, J = 5 Hz, 32H), 3.85 (s, 8H), 3.22 (t, J = 5 Hz, 32H), 2.93-2.79 (m, 8H), 1.67 (broad s, 6H), 1.60 (broad s, 6H). ^{13}C NMR (125 MHz, D_2O, dioxane as reference): 163.3 (q, J = 35.5 Hz), 160.2, 159.8, 157.4, 156.5, 150.7, 141.7, 140.1, 128.9, 117.2 (q, J = 291.5 Hz), 115.4, 109.0, 107.4, 102.0, 101.8, 79.2, 78.3, 72.3, 72.1, 70.2, 64.8, 53.7, 49.4, 39.6, 37.0, 36.0, 31.4, 16.4, 15.6. HR-MS (ESI, positive mode): m/z 1120.8316 (46%, $[M+3H]^{3+}$, calcd for $C_{166}H_{215}N_{32}O_{36}S_4^{3+}$: 1120.8303), 840.8755 (98%, $[M+4H]^{4+}$, calcd for $C_{166}H_{216}N_{32}O_{36}S_4^{4+}$: 840.8747), 672.9016 (100%, $[M+5H]^{5+}$, calcd for $C_{166}H_{217}N_{32}O_{36}S_4^{5+}$: 672.9013), 560.7522 (68%, $[M+6H]^{6+}$, calcd for $C_{166}H_{218}N_{32}O_{36}S_4^{6+}$: 560.9191), 480.9319 (12%, $[M+7H]^{7+}$, calcd for $C_{166}H_{219}N_{32}O_{36}S_4^{7+}$: 480.9318).



Compound G3. **G3-Boc** (106 mg, 11.5 μmol) was dissolved in TFA (2 mL). After 20 minutes, TFA was removed by rotary evaporation then under high vacuum to give **G3** as its TFA salt (103 mg, quantitative). Colorless solid. M.p. 134-136°C. IR (ATR, cm^{-1}): 2912 (N-H), 1668 (C=O, ureidyl), 1591 (C=O, TFA). ^{1}H NMR (600 MHz, DMSO-d_6, 70°C): 8.08 (broad s, amines), 6.92 (broad s, 4H), 6.81-6.53 (m, 84H), 5.68-5.25 (m, 14H), 5.04 (broad s, 32H), 5.00 (broad s, 16H), 4.33-4.03 (m, 18H), 4.18 (t, J = 5 Hz, 64H), 3.91-3.81 (m, 8H), 3.21 (t, J = 5 Hz, 64H), 2.92 (overlapping with HOD signal, m, 8H), 1.63 (broad s, 6H), 1.54 (broad s, 6H). ^{13}C NMR (150 MHz, D_2O, dioxane as reference): 163.4 (q, J = 35.5 Hz), 160.3, 159.8, 157.3, 156.5, 150.3, 142.3, 140.1, 128.8, 117.2 (q, J = 291.5 Hz), 115.1, 108.9, 107.3, 107.1, 102.1, 101.2, 79.3, 78.3, 72.2, 70.8, 70.2, 64.7, 53.5, 49.5, 39.6, 37.1, 35.8, 31.3, 15.9, 15.3. HR-MS (ESI, positive mode): m/z 1501.6949 (5%, $[M+4H]^{4+}$, calcd for $C_{310}H_{392}N_{48}O_{68}S_4^{4+}$: 1501.6920), 1201.5580 (23%, $[M+5H]^{5+}$, calcd for $C_{310}H_{393}N_{48}O_{68}S_4^{5+}$: 1201.5552), 1001.4657 (41%, $[M+6H]^{6+}$, calcd for $C_{310}H_{394}N_{48}O_{68}S_4^{6+}$: 1001.4640), 858.5428 (100%, $[M+7H]^{7+}$, calcd for $C_{310}H_{395}N_{48}O_{68}S_4^{7+}$: 858.5417).

References

(1) Sigwalt, D.; Holler, M.; Iehl, J.; Nierengarten, J.-F.; Nothisen, M.; Morin, E.; Remy, J.-S. *Chem. Commun.* **2011**, *47*, 4640.

(2) Zhang, B.; Zavalij, P. Y.; Isaacs, L. *Org. Biomol. Chem.* **2014**, *12*, 2413.

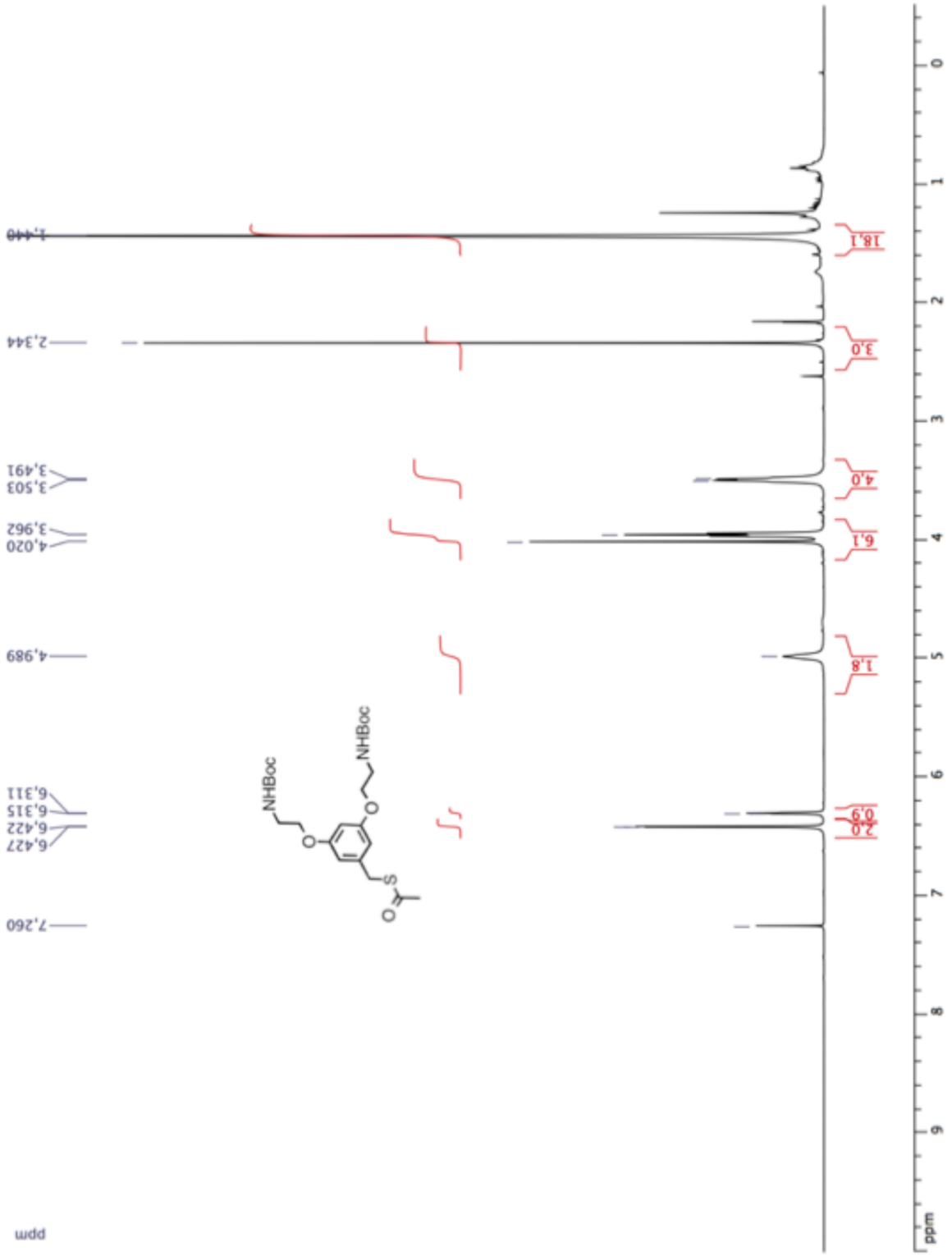


Figure S1. ^{1}H NMR recorded ($CDCl_3$, 400 MHz) for **D1-SAc**. Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

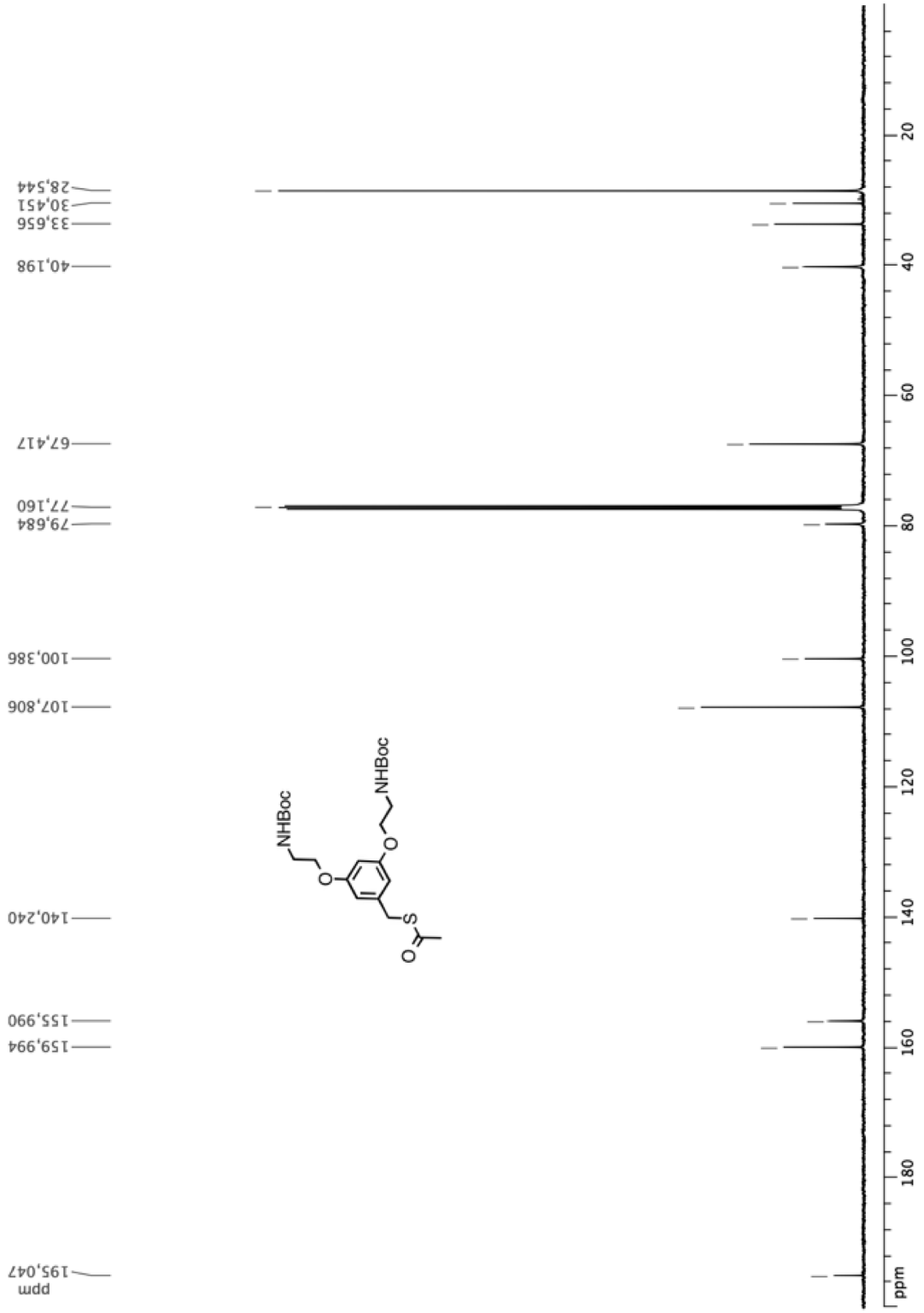


Figure S2. ^{1}H NMR recorded ($CDCl_3$, 125 MHz) for **D1-SAc**.

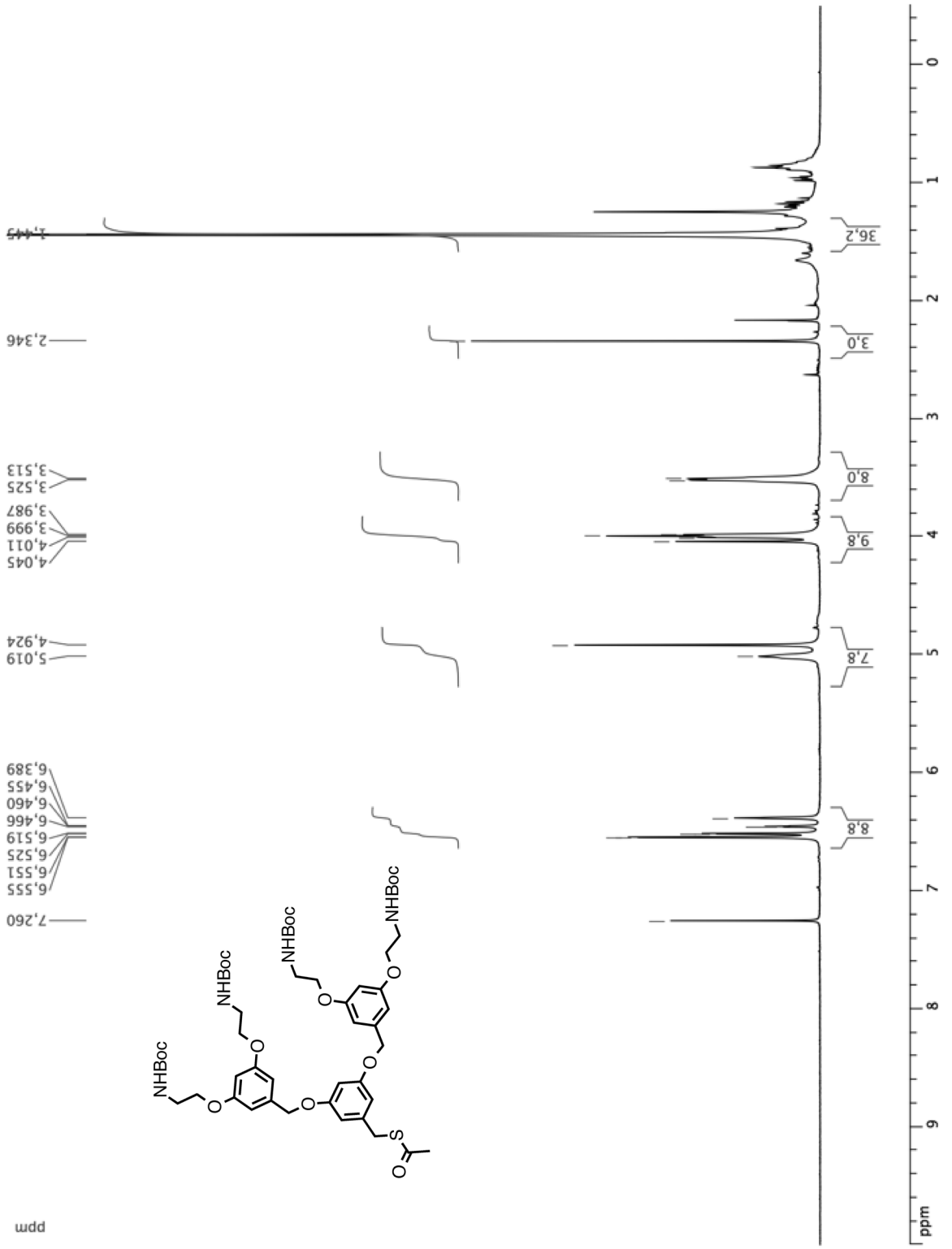


Figure S3. ^{1}H NMR recorded ($CDCl_3$, 400 MHz) for **D2-SAc**. Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

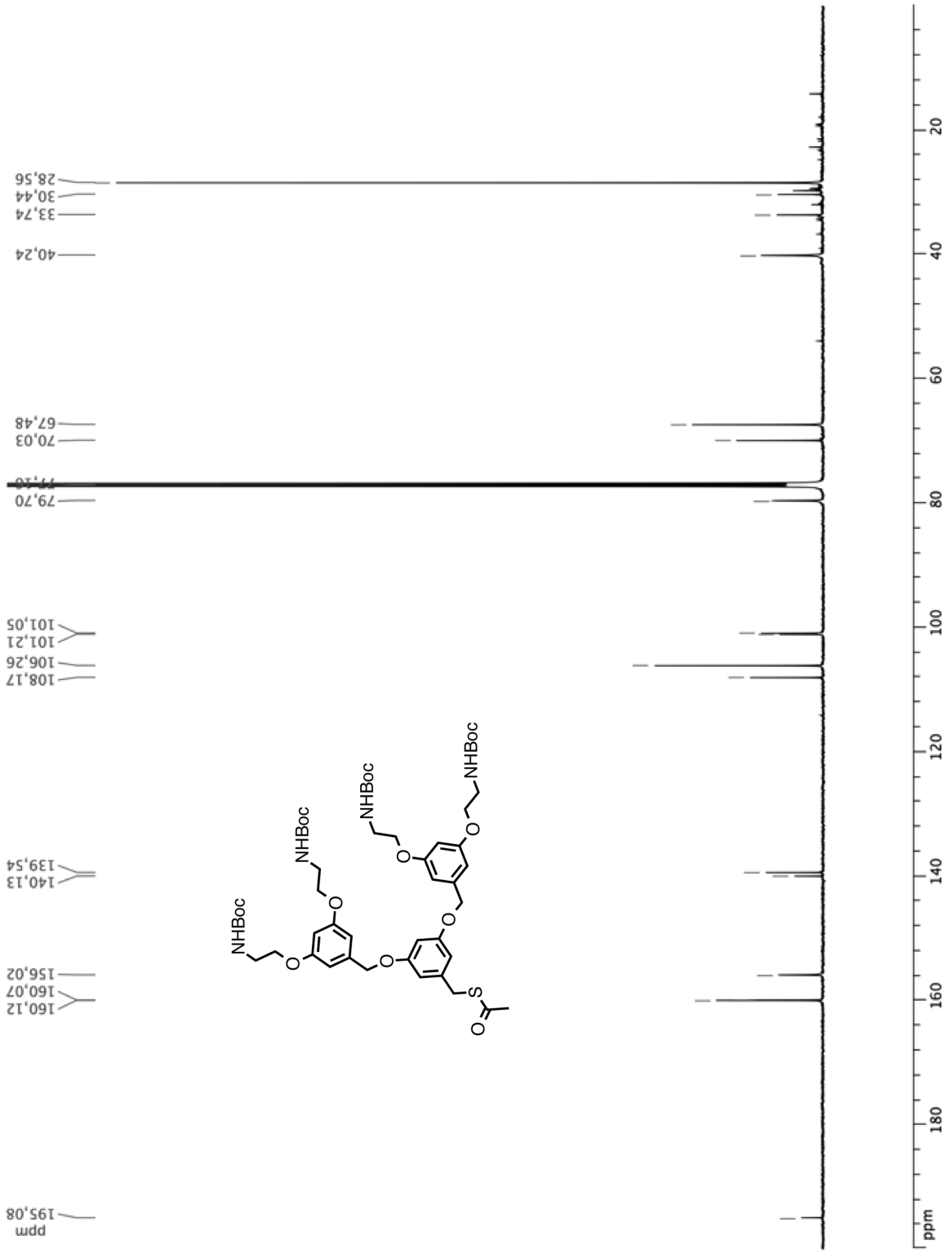


Figure S4. ^{13}C NMR recorded ($CDCl_3$, 125 MHz) for **D2-SAc**.

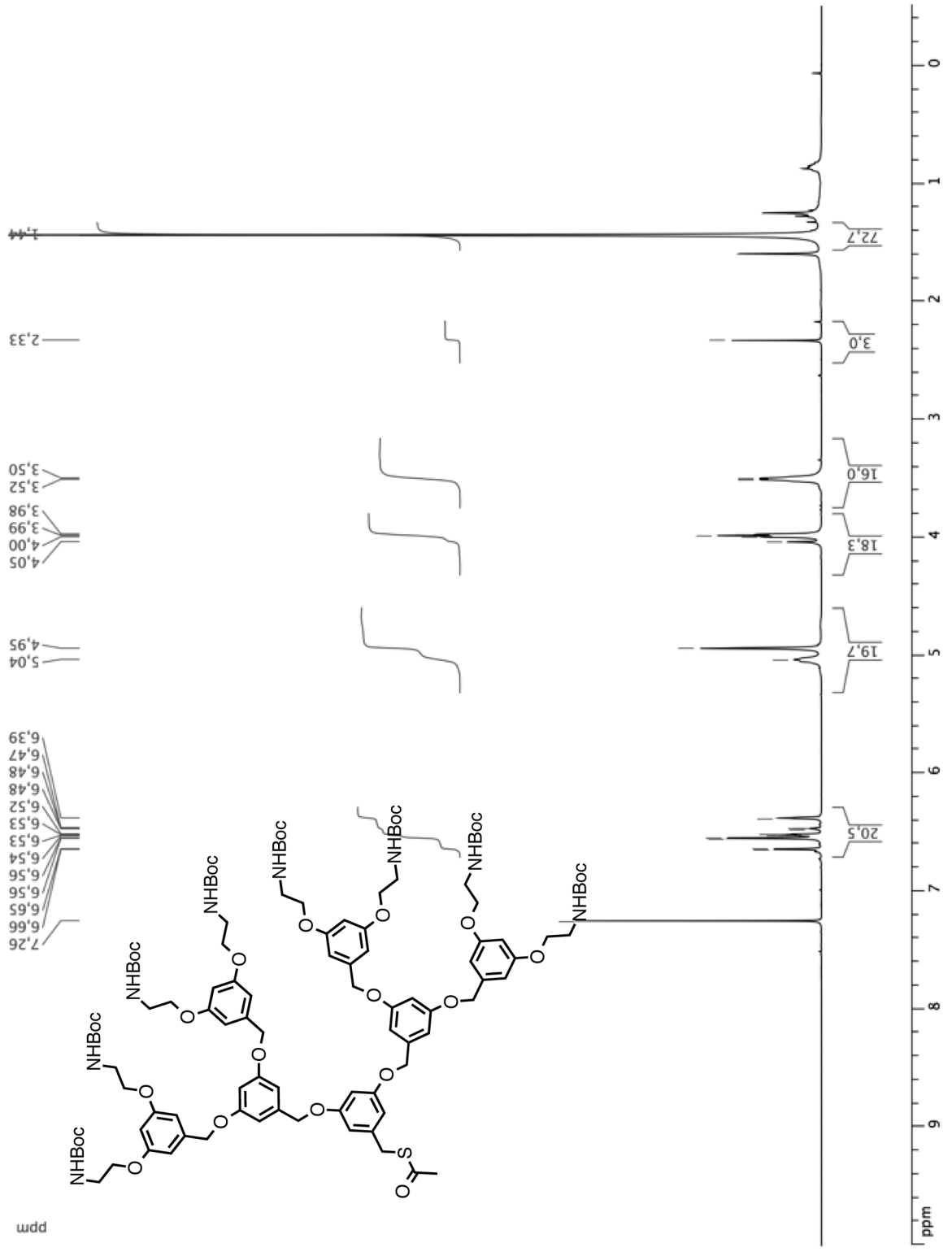


Figure S5. ^{1}H NMR recorded ($CDCl_3$, 400 MHz) for **D3-SAc.** Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

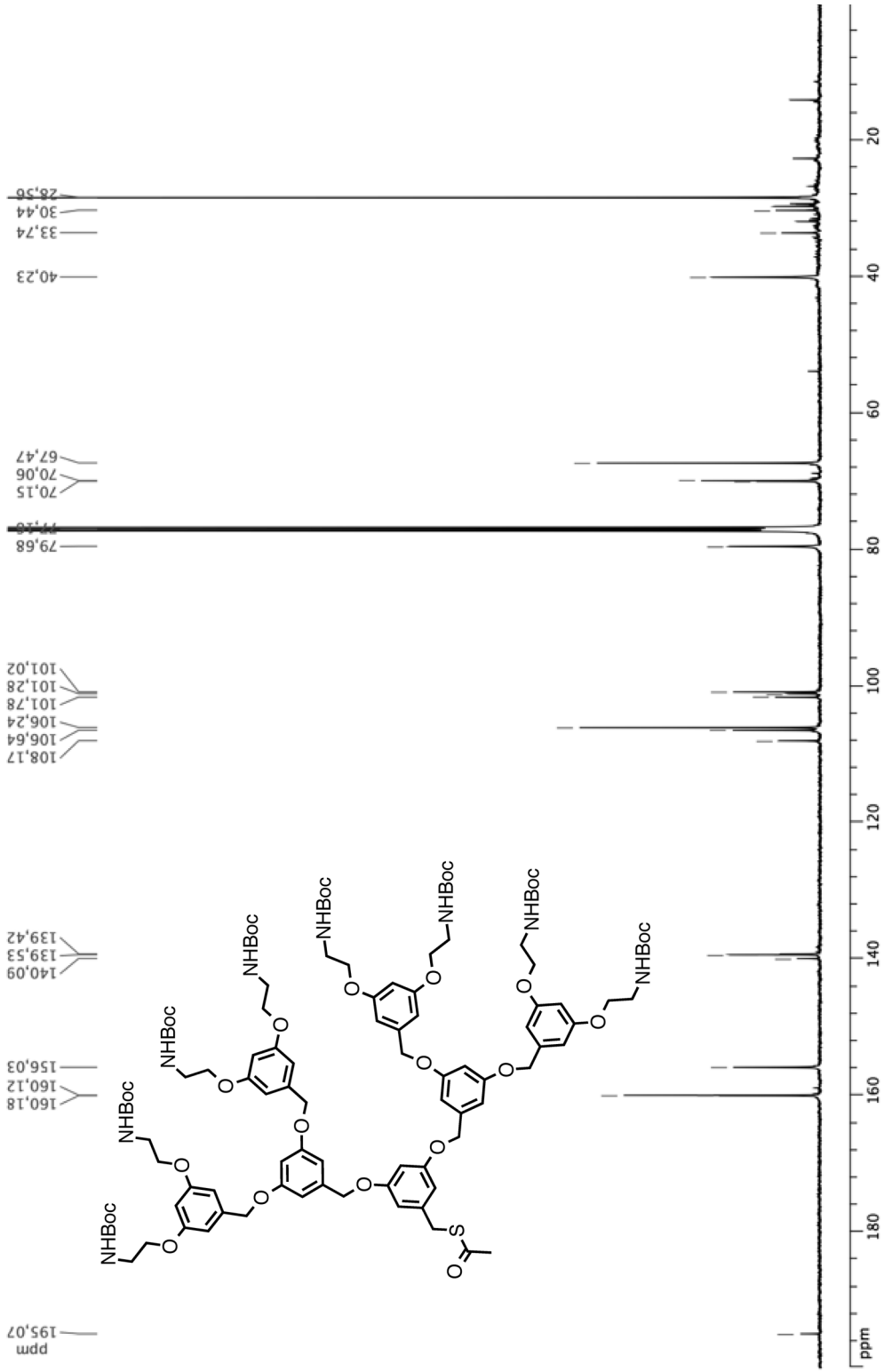


Figure S6. ^{13}C NMR recorded ($CDCl_3$, 125 MHz) for **D3-SAc.**

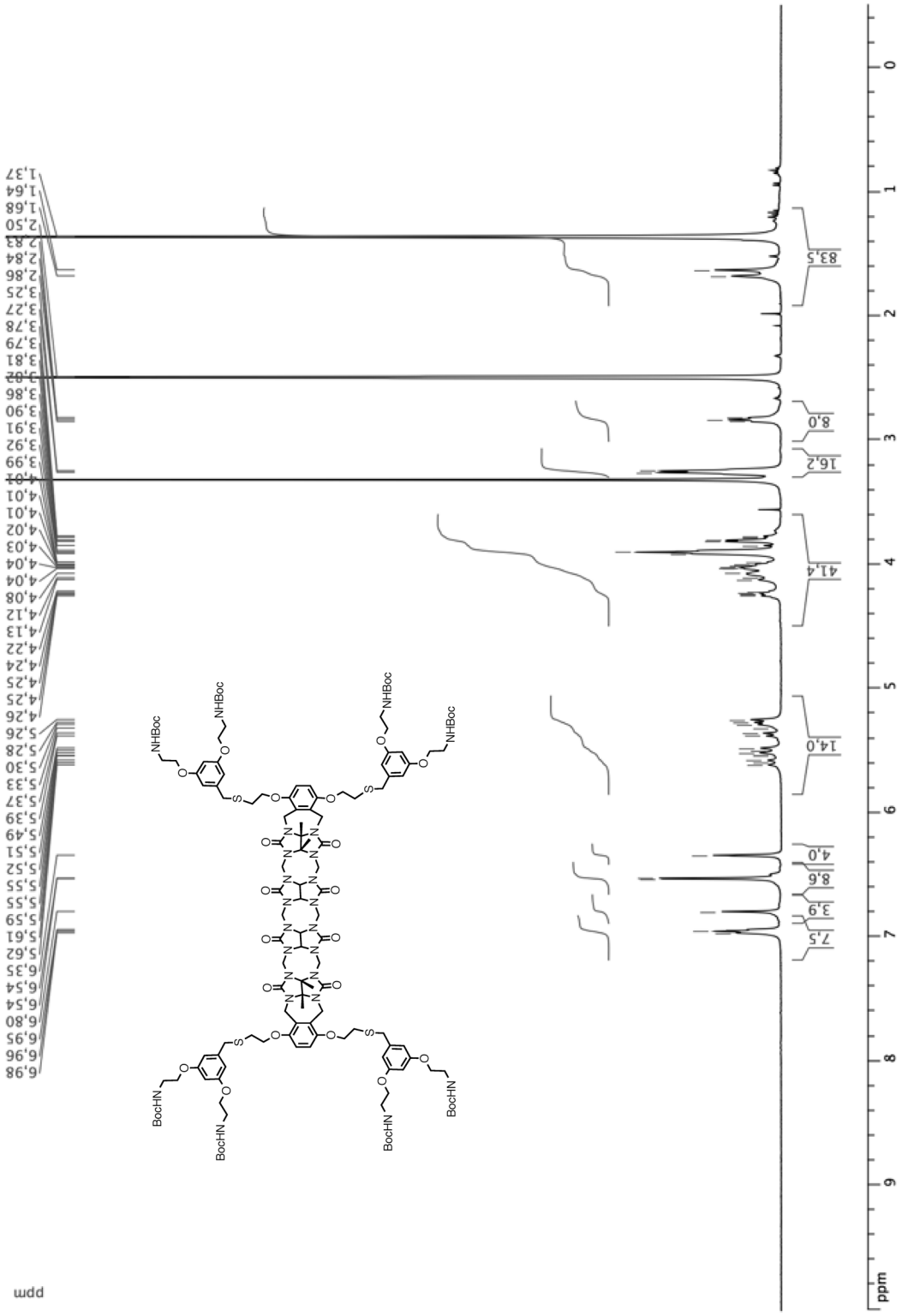


Figure S7. ^{1}H NMR recorded (DMSO-*d6*, 400 MHz) for **G1-Boc**. Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

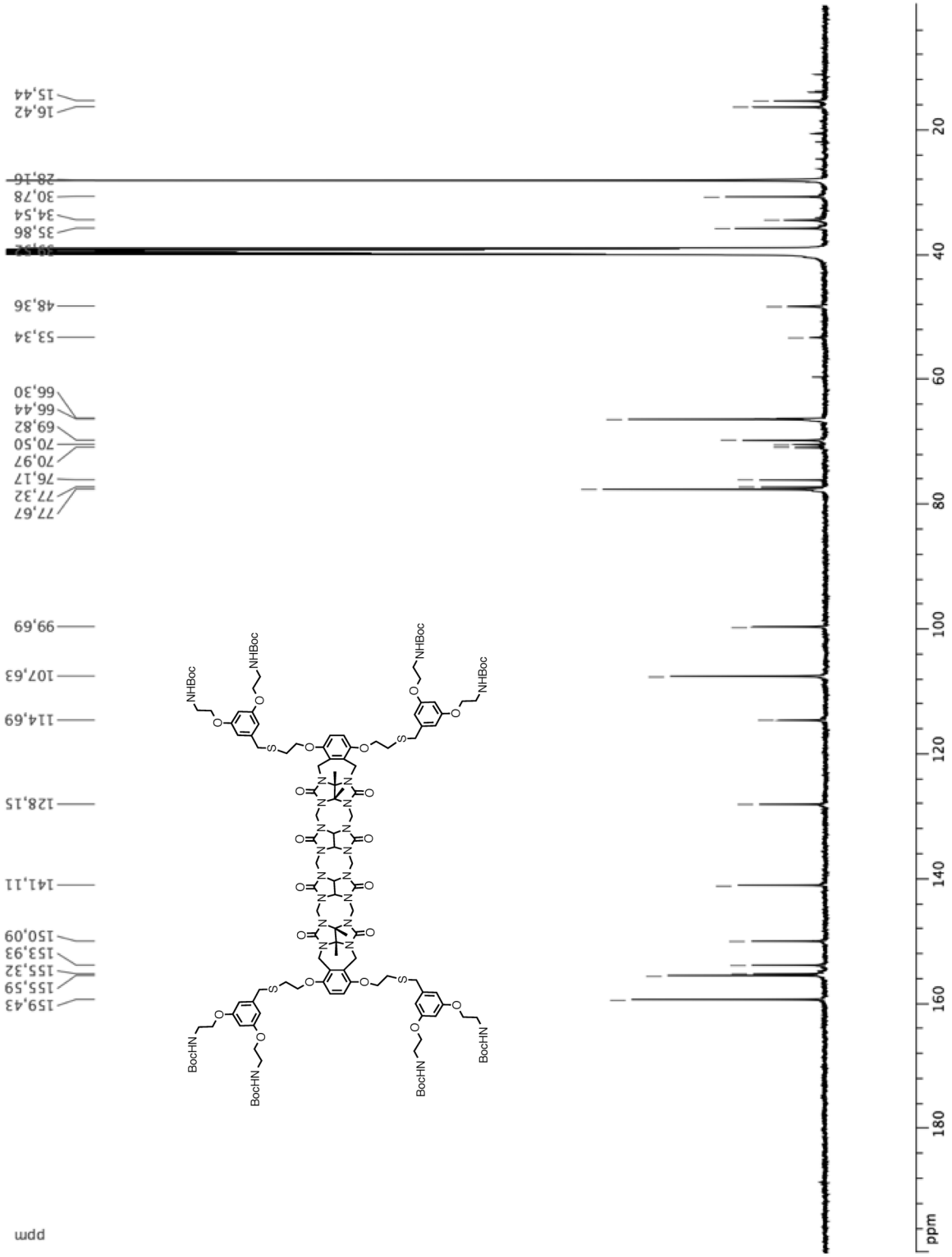


Figure S8. ^{13}C NMR recorded (DMSO-*d6*, 125 MHz) for **G1-Boc**.

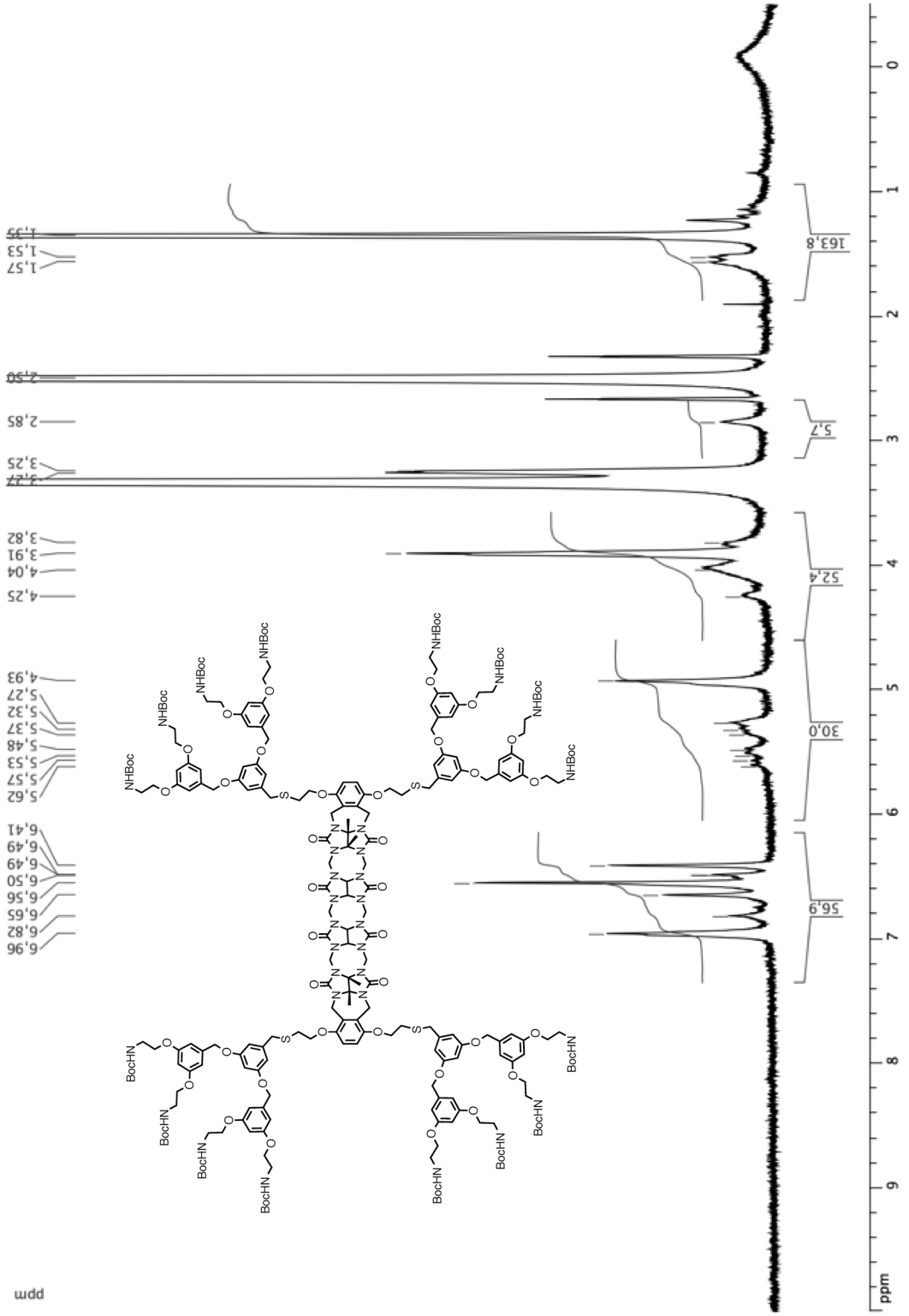


Figure S9. [1]H NMR recorded (DMSO-*d6*, 400 MHz) for **G2-Boc**. Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

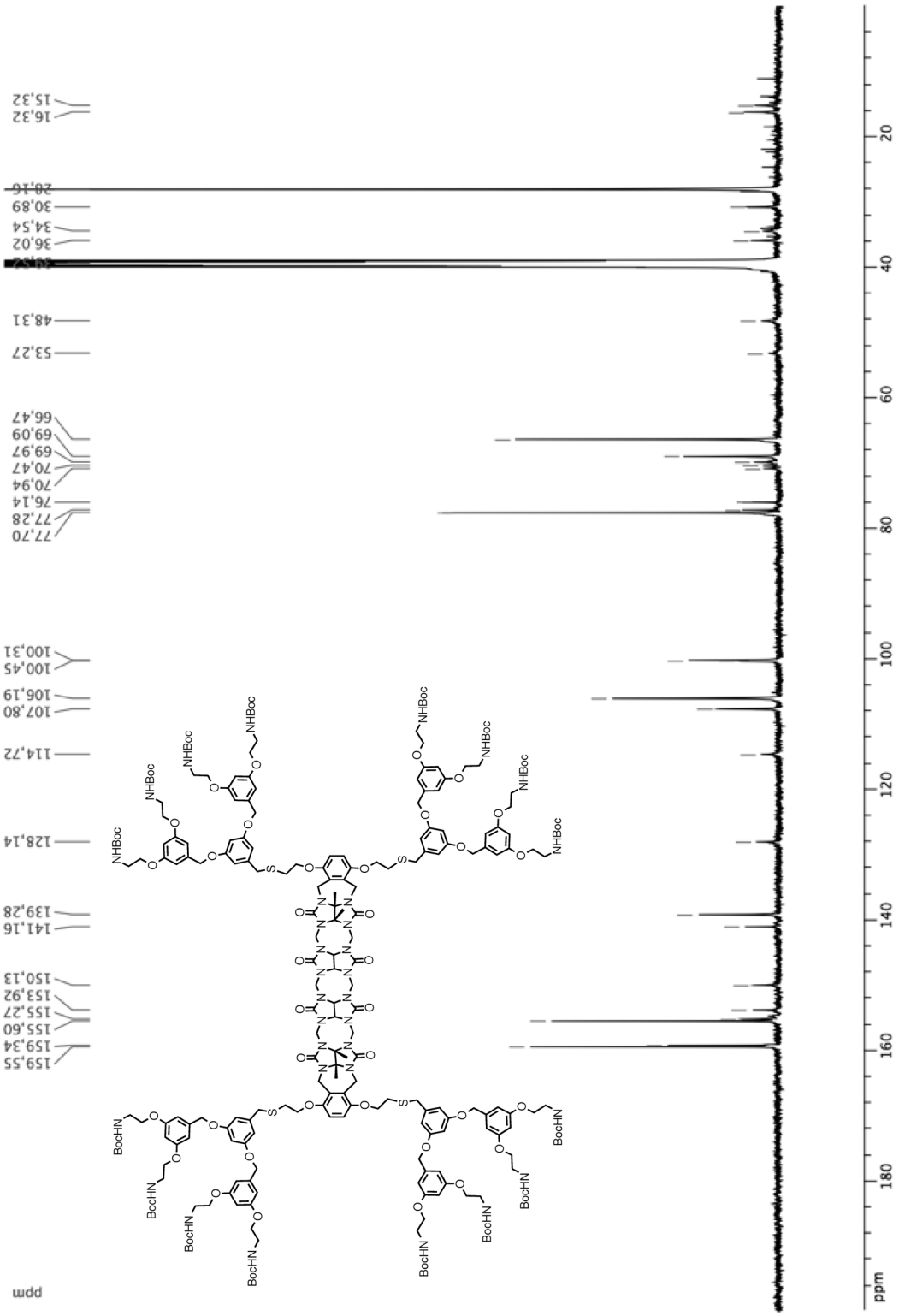


Figure S10. ^{13}C NMR recorded (DMSO-*d6*, 125 MHz) for **G2-Boc**.

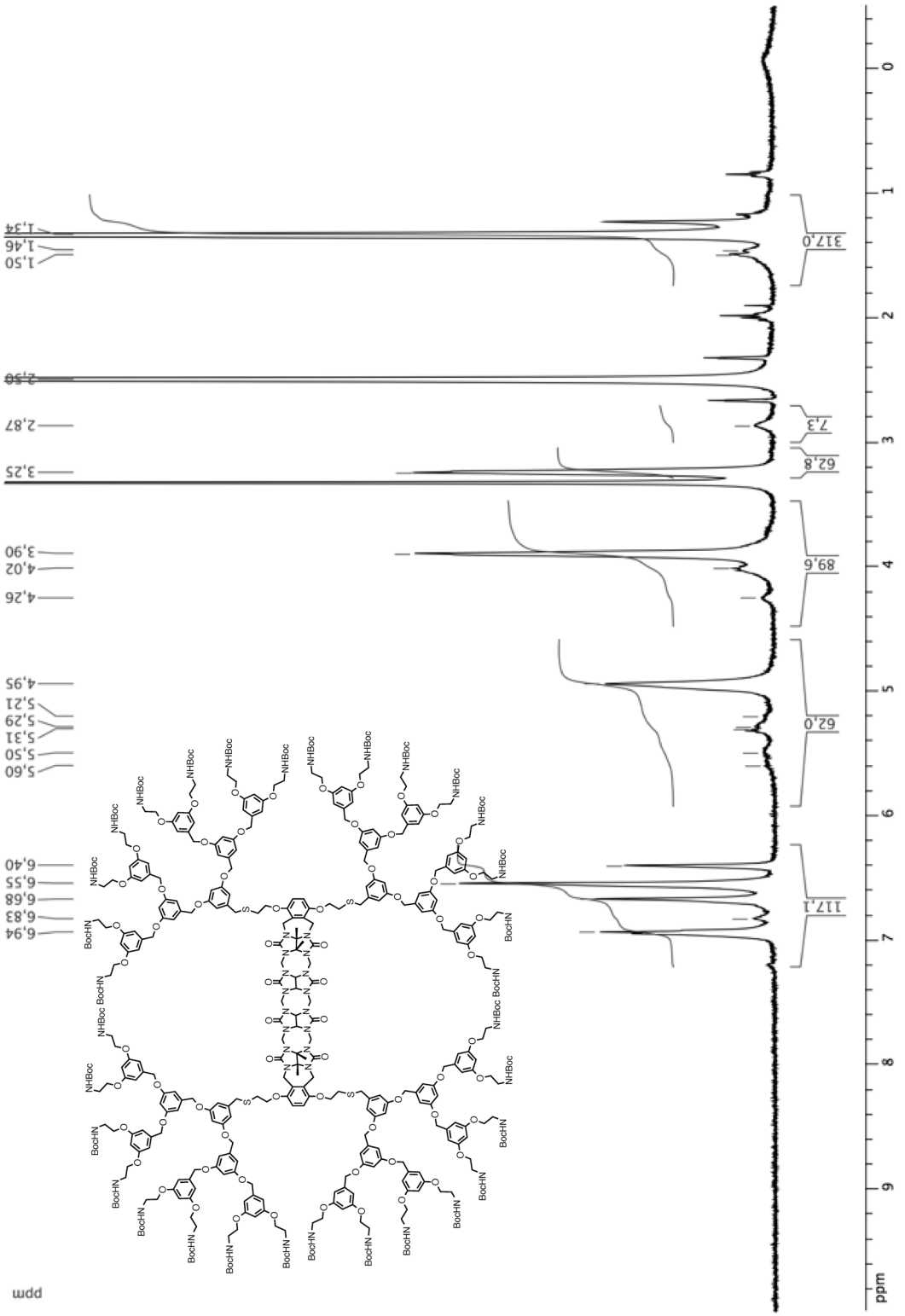


Figure S11. [1]H NMR recorded (DMSO-*d6*, 400 MHz) for **G3-Boc**. Peaks at 0.8 and 1.2 ppm arise from residual hexanes used during purification.

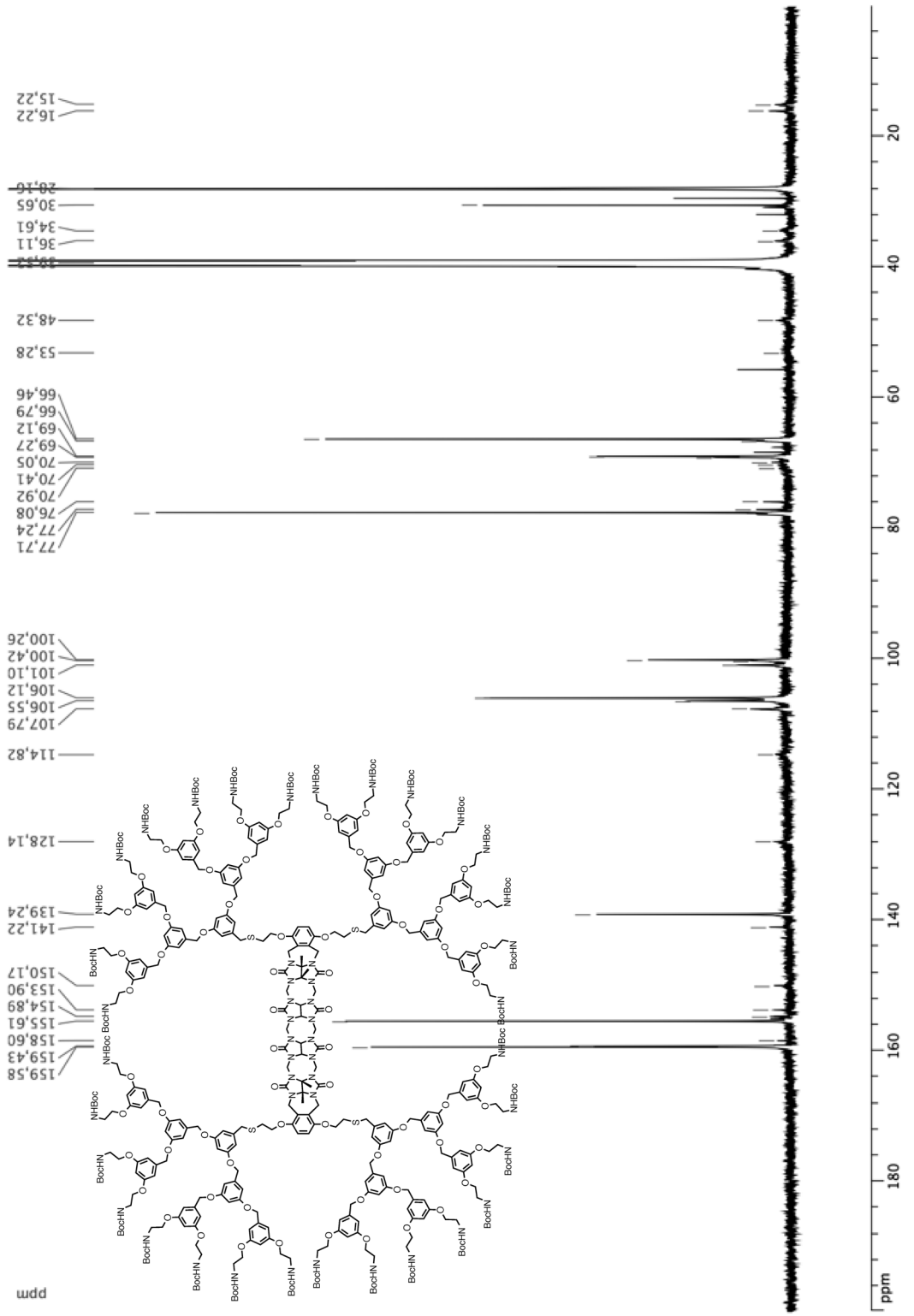


Figure S12. ^{13}C NMR recorded (DMSO-*d6*, 125 MHz) for **G3-Boc**.

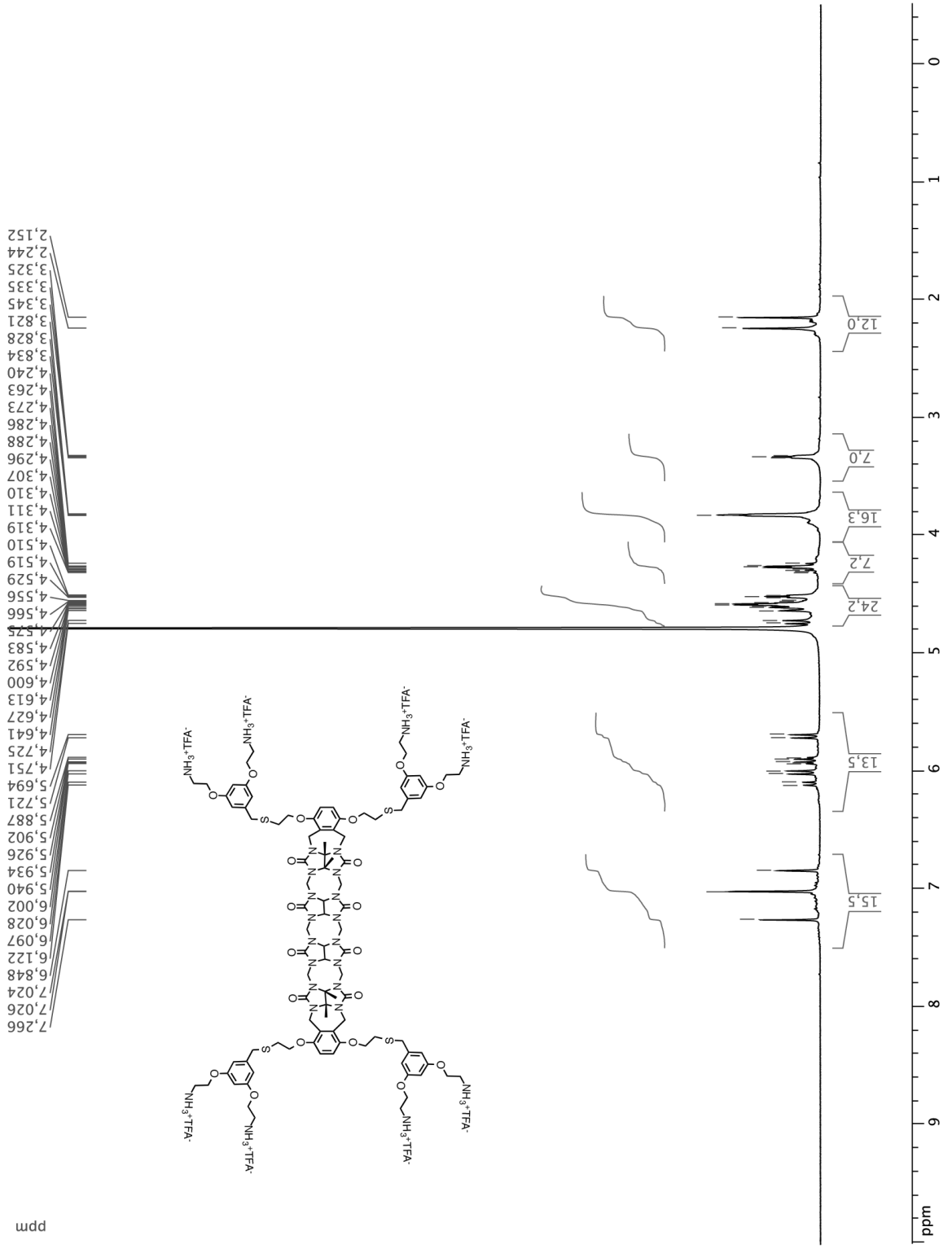


Figure S13. ^{1}H NMR spectrum recorded for **G1** (D_2O, 600 MHz, 70°C).

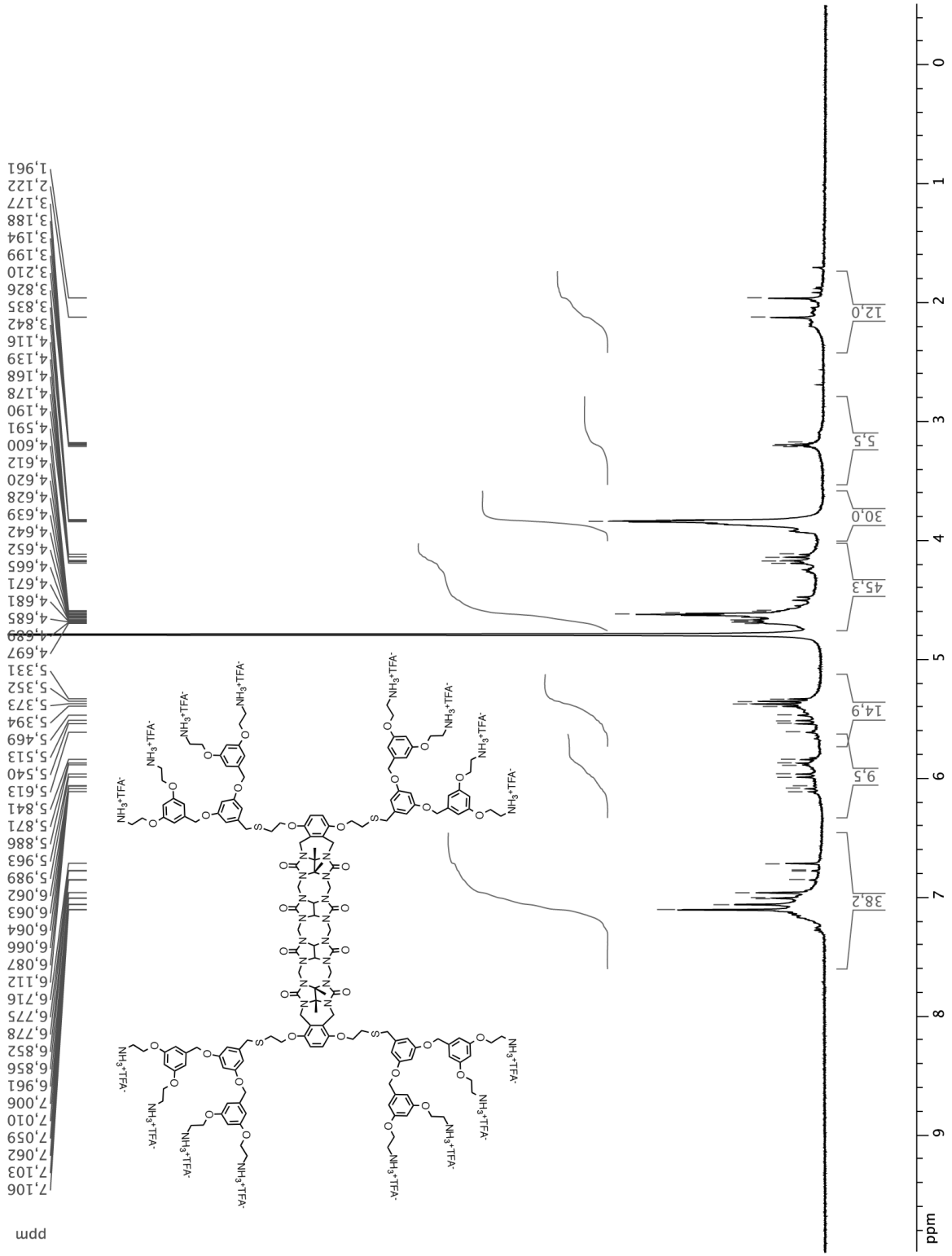


Figure S14. ^{1}H NMR spectrum recorded for **G2** (D_2O, 600 MHz, 70°C).

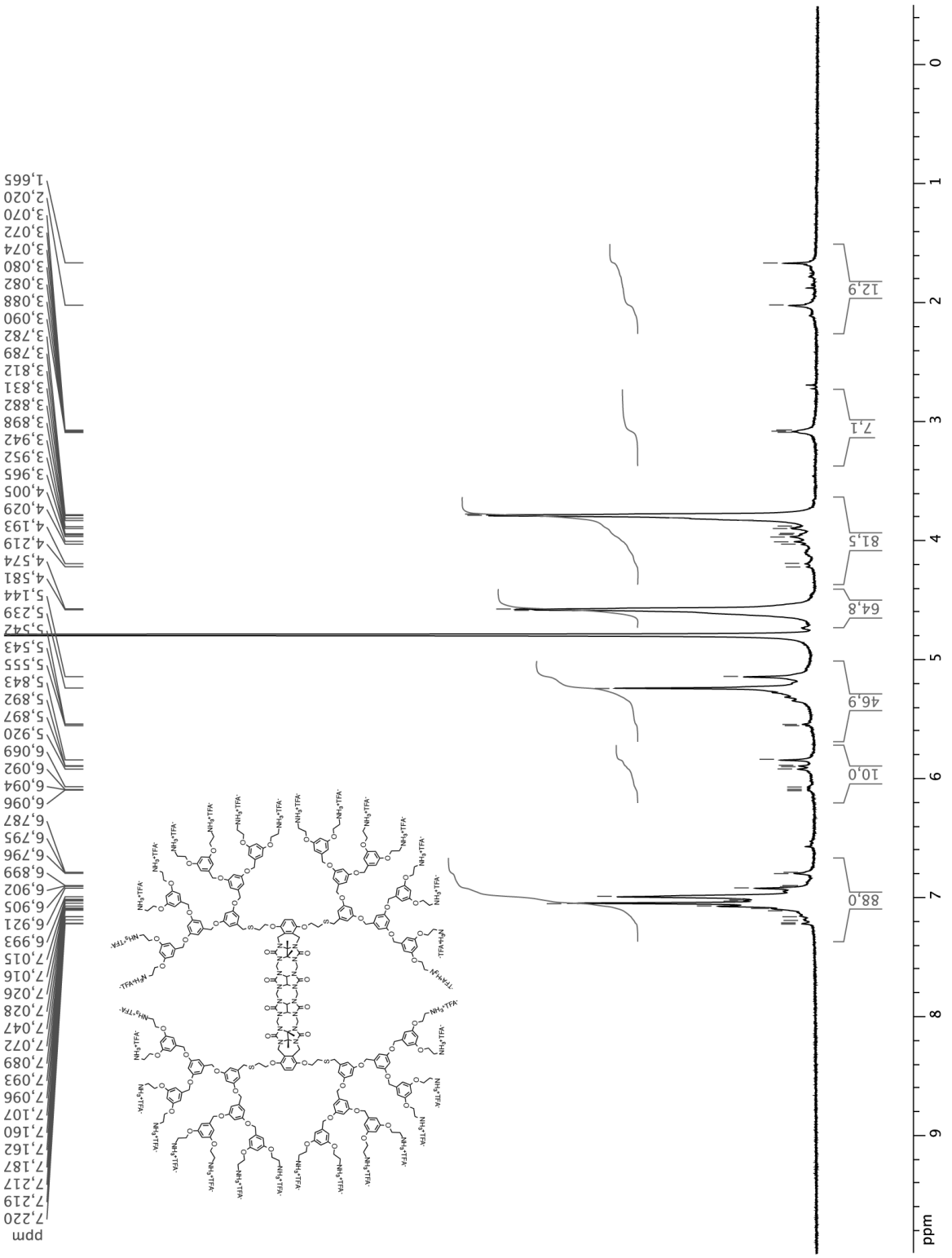


Figure S15. ^{1}H NMR spectrum recorded for **G3** (D_2O, 600 MHz, 70°C).

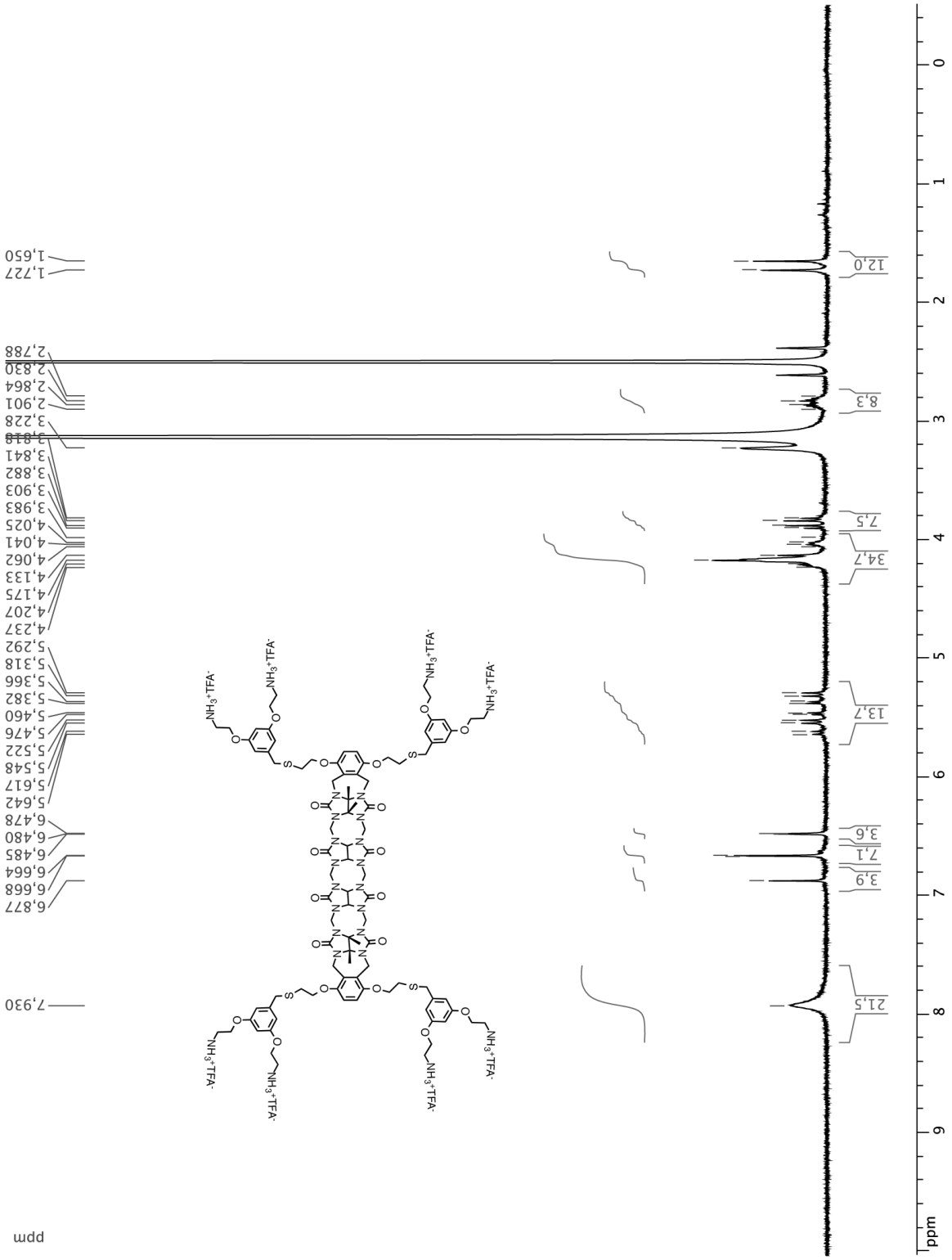


Figure S16. ^{1}H NMR spectrum recorded for **G1** (DMSO-*d6*, 600 MHz, 70°C).

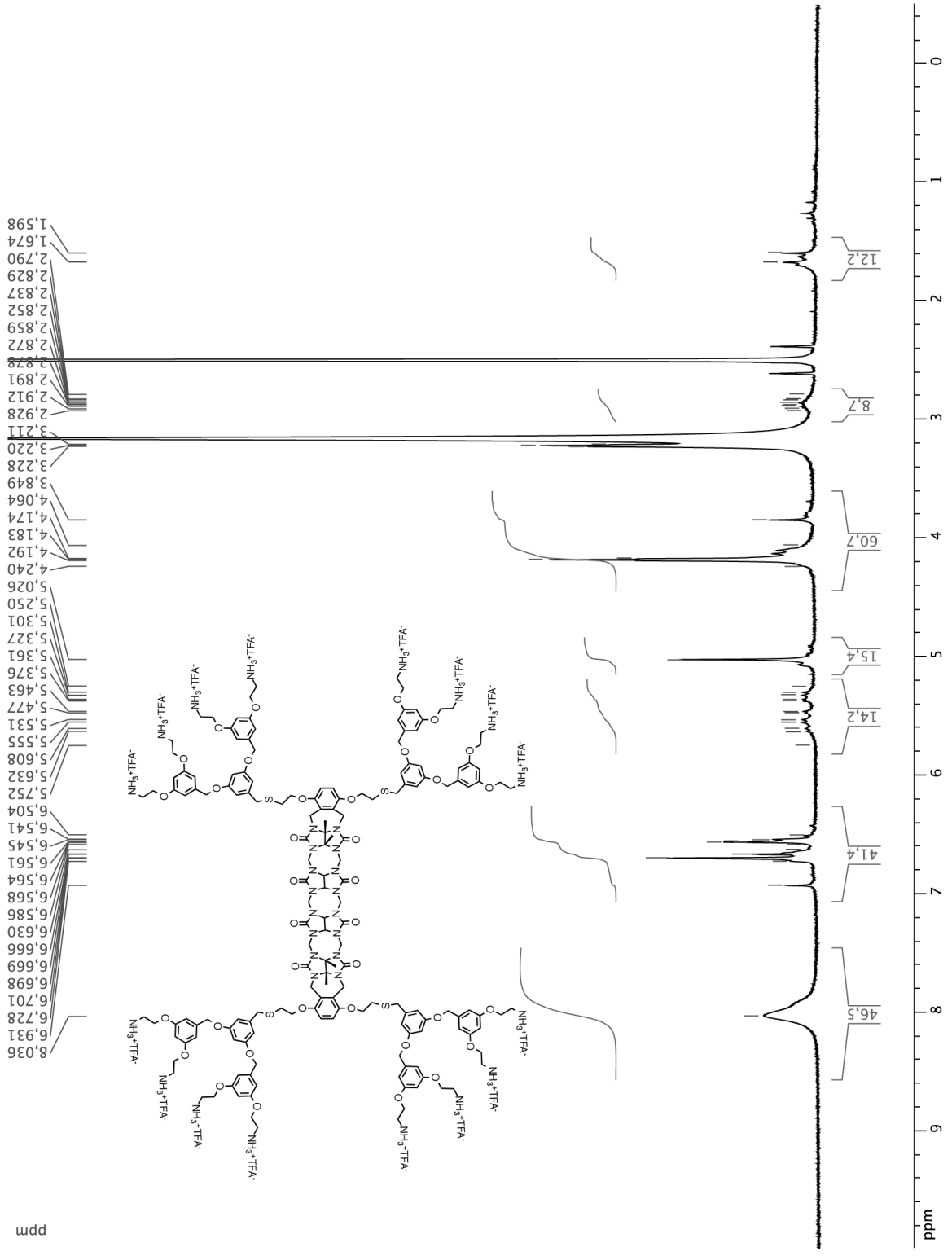


Figure S17. ^{1}H NMR spectrum recorded for **G2** (DMSO-*d6*, 600 MHz, 70°C).

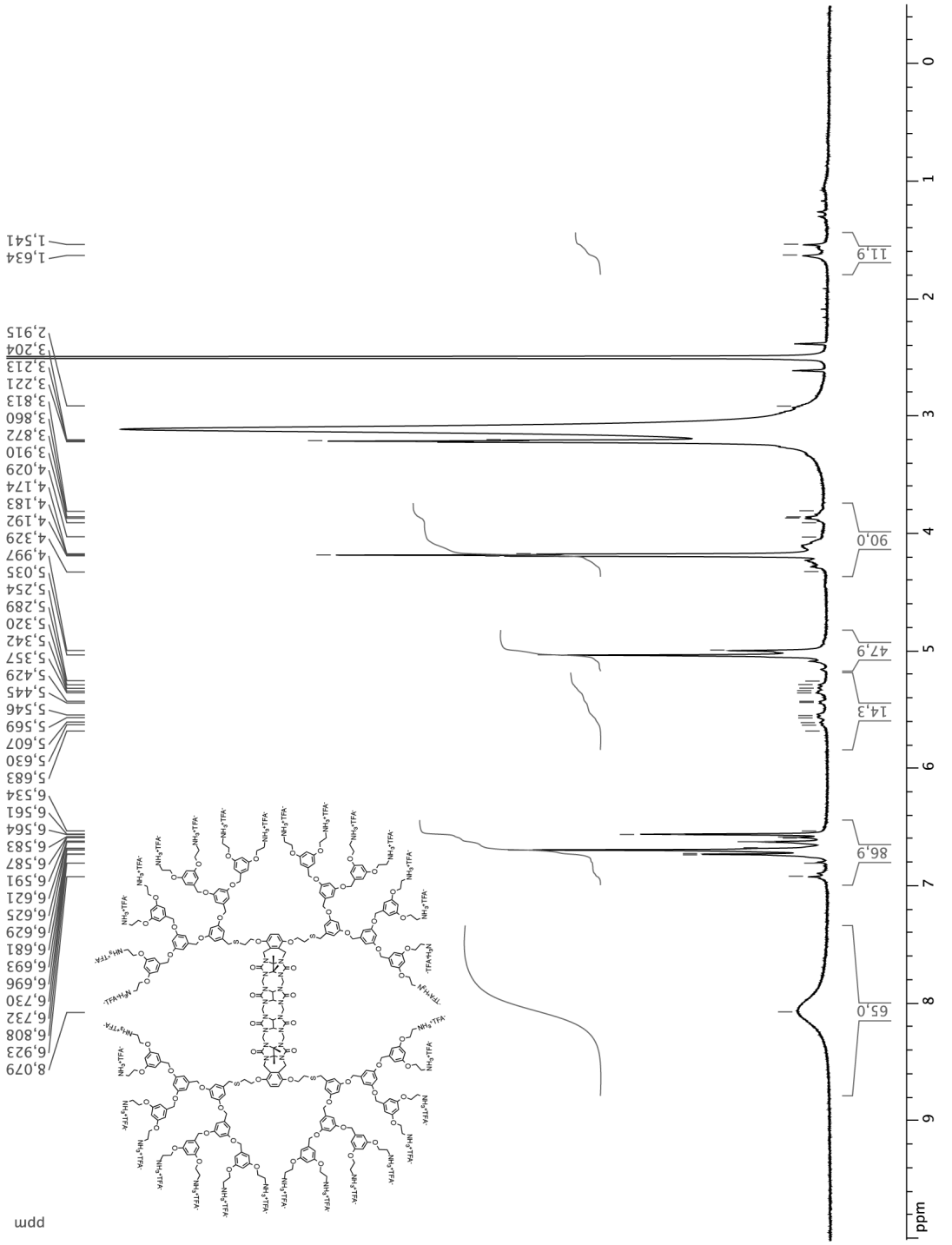


Figure S18. ^{1}H NMR spectrum recorded for **G3** (DMSO-*d6*, 600 MHz, 70°C).

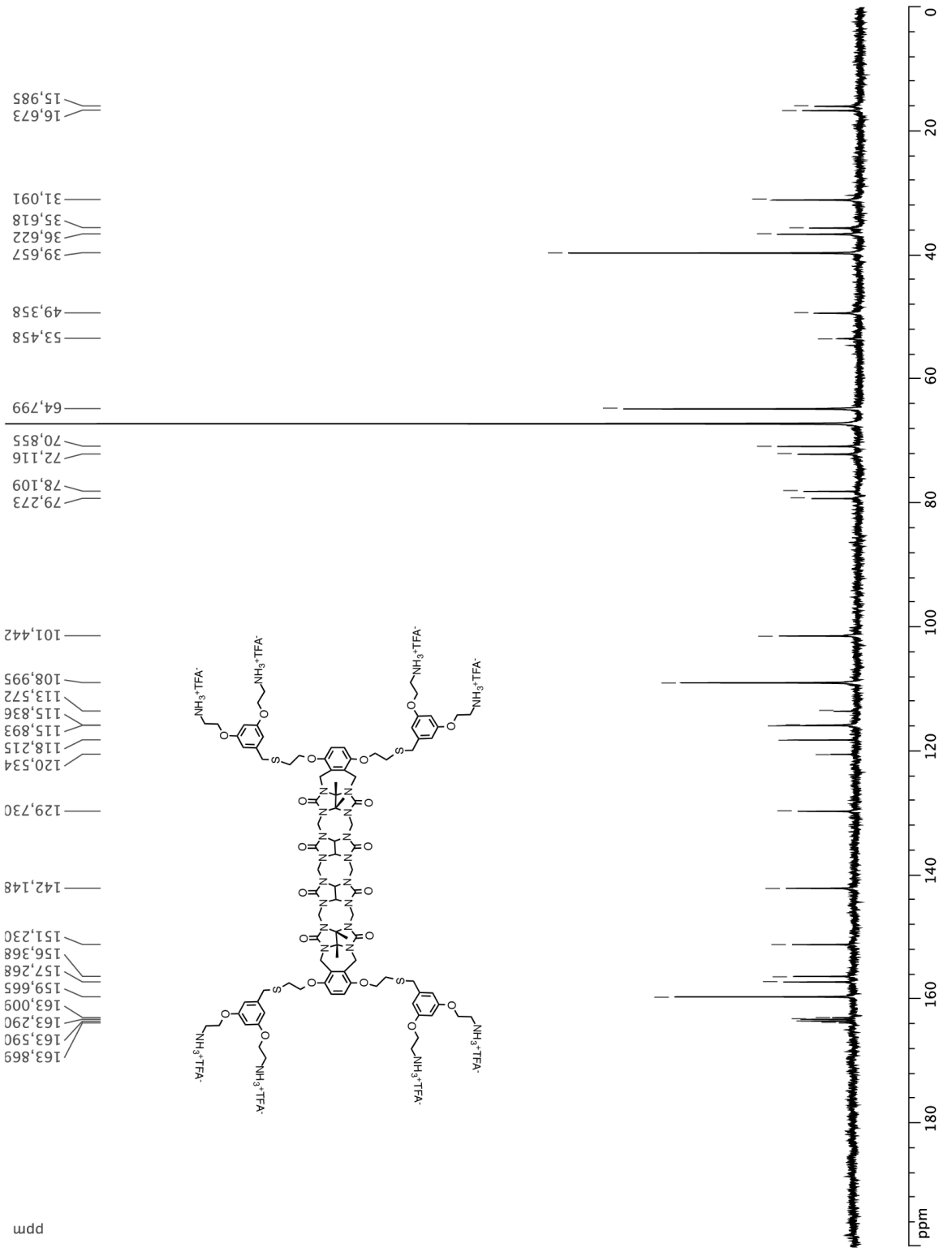


Figure S19. ^{13}C NMR spectrum recorded for **G1** (D_2O, dioxane as reference, 125 MHz).

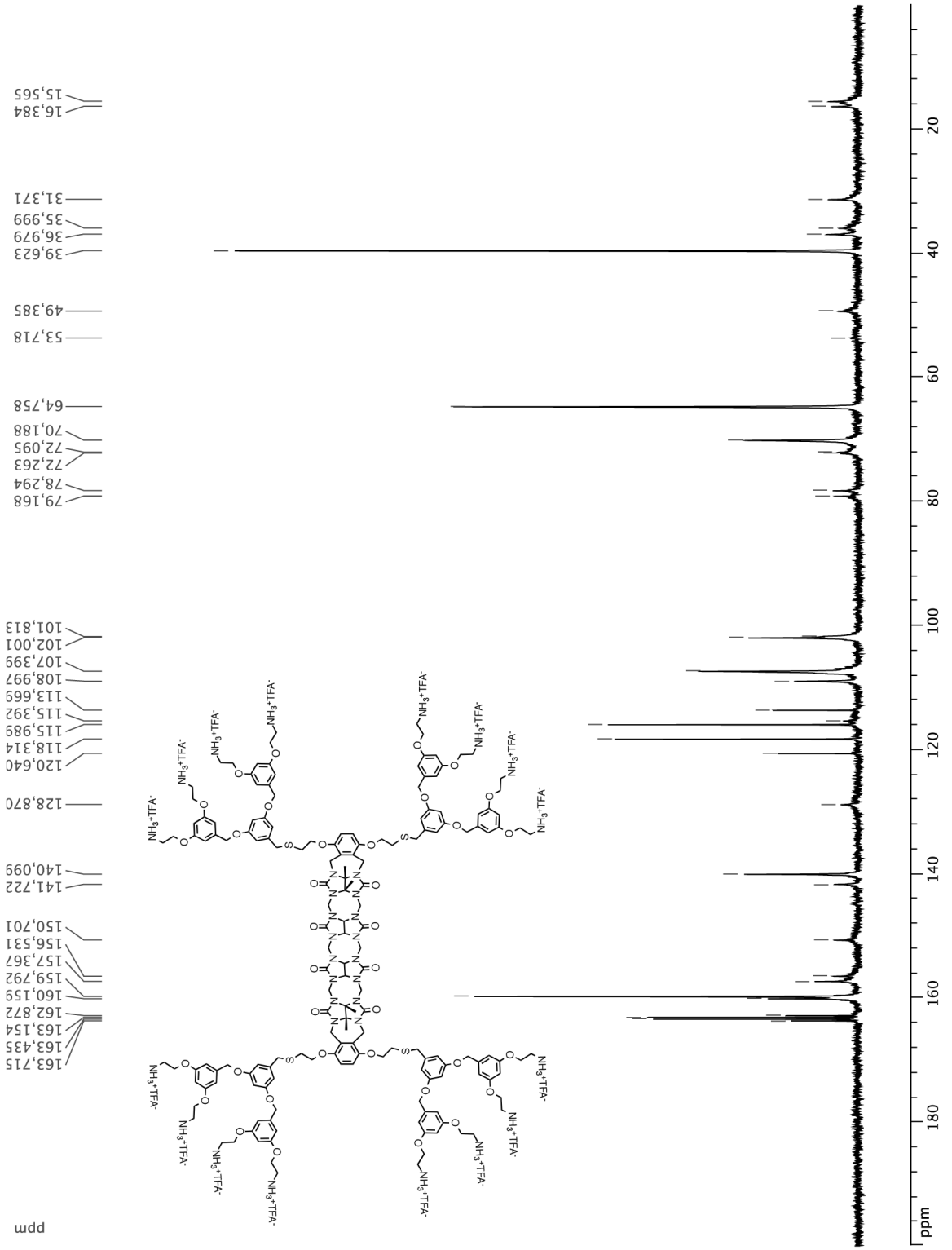


Figure S20. ^{13}C NMR spectrum recorded for **G2** (D_2O, dioxane as reference, 125 MHz).

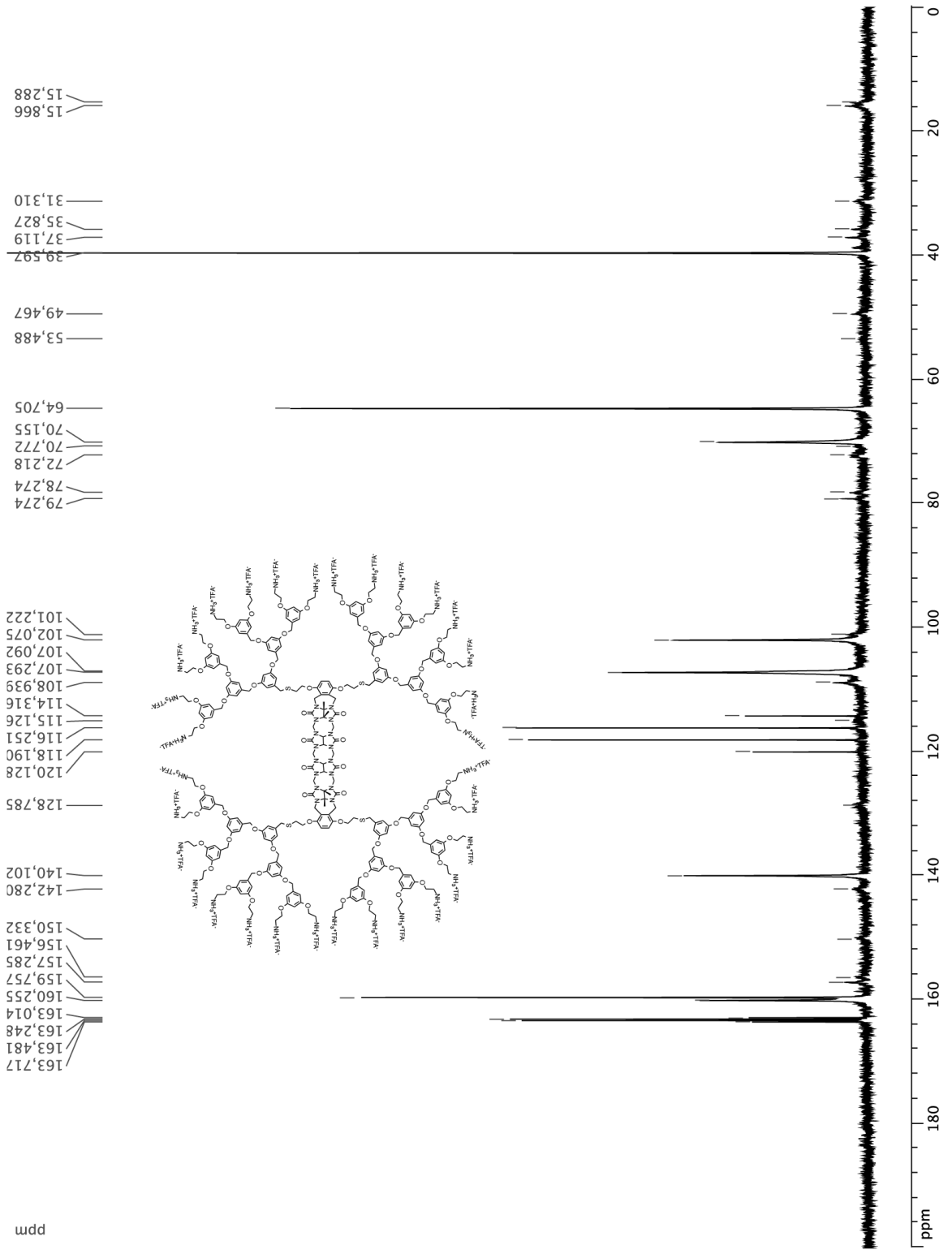


Figure S21. ^{13}C NMR spectrum recorded for **G3** (D_2O, dioxane as reference, 150 MHz).

Agarose gel electrophoresis.

Water (20 μL) containing glucose (5 %), pEGFP (0.8 μg) and increasing amounts of cationic molecules were subjected (30 min of complexation time) to electrophoresis in a 1% agarose gel containing 1mm EDTA and 40 mm Tris acetate buffer and 0.5 μg mL^{-1} ethidium bromide, for 40 min at 125 V. DNA was visualized with an UV transilluminator at 254 nm.

Scanning Electron Microscopy.

Preparation of samples. A drop of water solution of molecule or complexes (30 min complexation time) was deposited on a silica wafer and left overnight to dry.

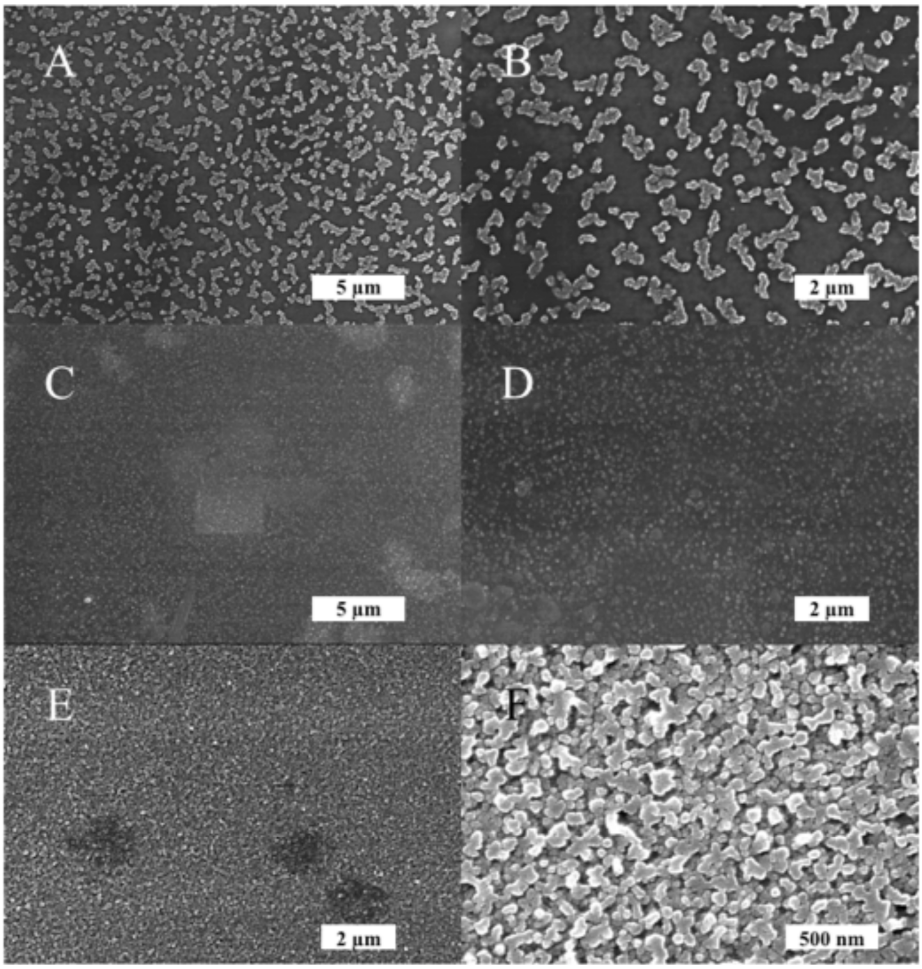


Figure S22. SEM of **G1** (A and B); **G2** (C and D); **G3** (E and F) at 90 μM of amine.

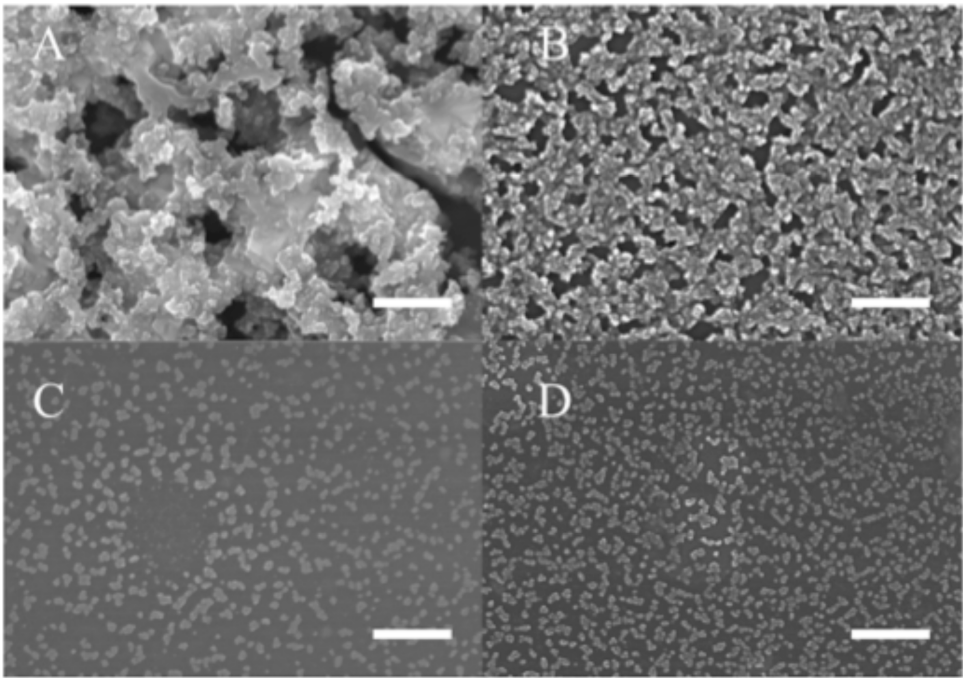


Figure S23. SEM images of DNA only (3 μg/mL) (A); **G1** (B), **G2** (C) or **G3** (D) with DNA at N/P 20 ratio. Scale bar: 2 μm.

Size determination and ζ potential.

Samples were prepared in water.

Table 1. Size of particles determined by Dynamic Light Scattering (nm).

	Molecule alone (2.4 mM of amine)	Molecule with DNA (pEGFP at N/P 20)
G1	118	75
G2	2* and 131	66
G3	2* and 100	81

* These values are at the detection limit of the apparatus and probably correspond to small amounts of monomeric dendrimer. The size of DNA solution alone present two maximums of intensity at 49 nm and 471 nm.

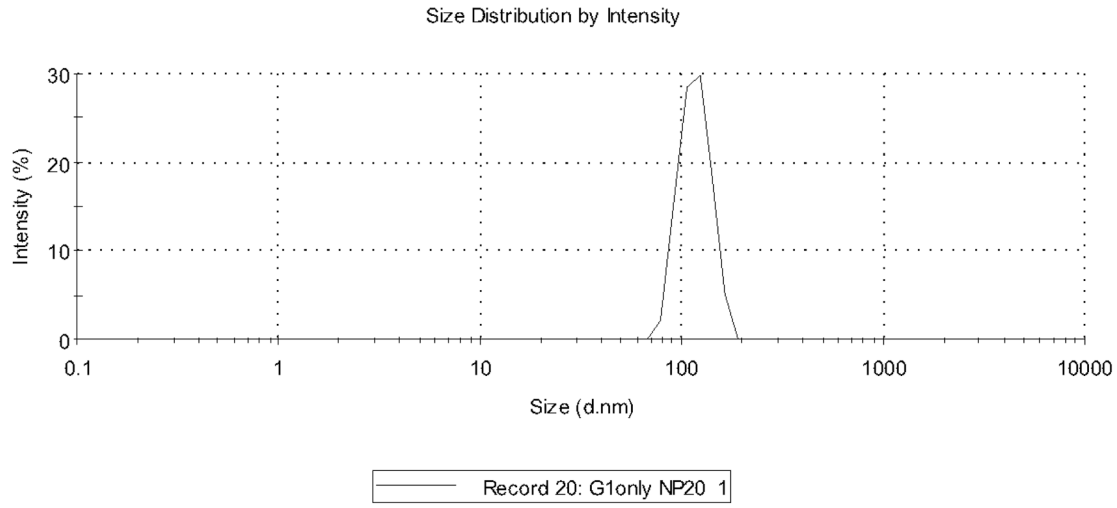


Figure S24. DLS of **G1** in water (2.4 mM of amine). Maxima at 118 nm.

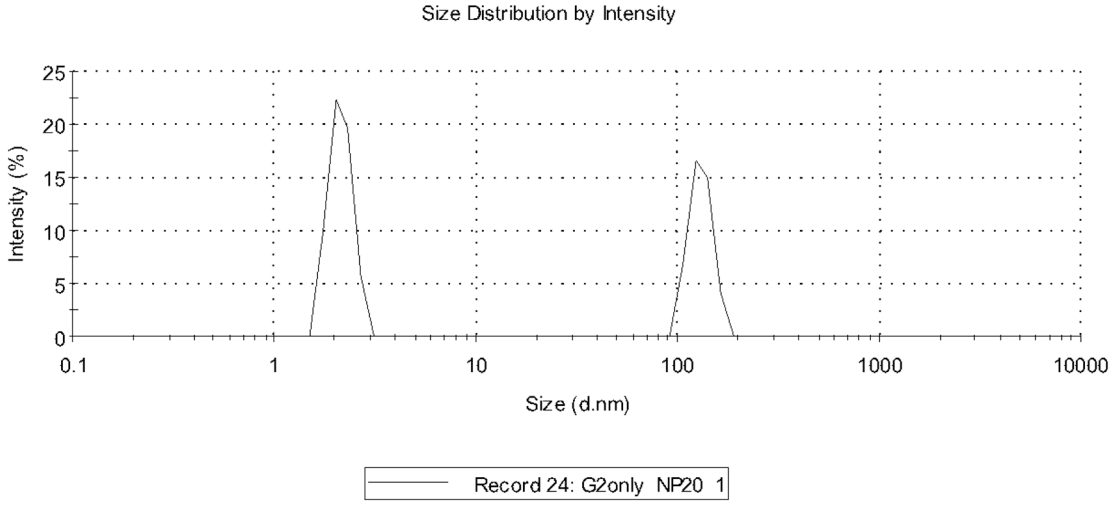


Figure S25. DLS of **G2** in water (2.4 mM of amine). Maxima at 2 nm and 131 nm.

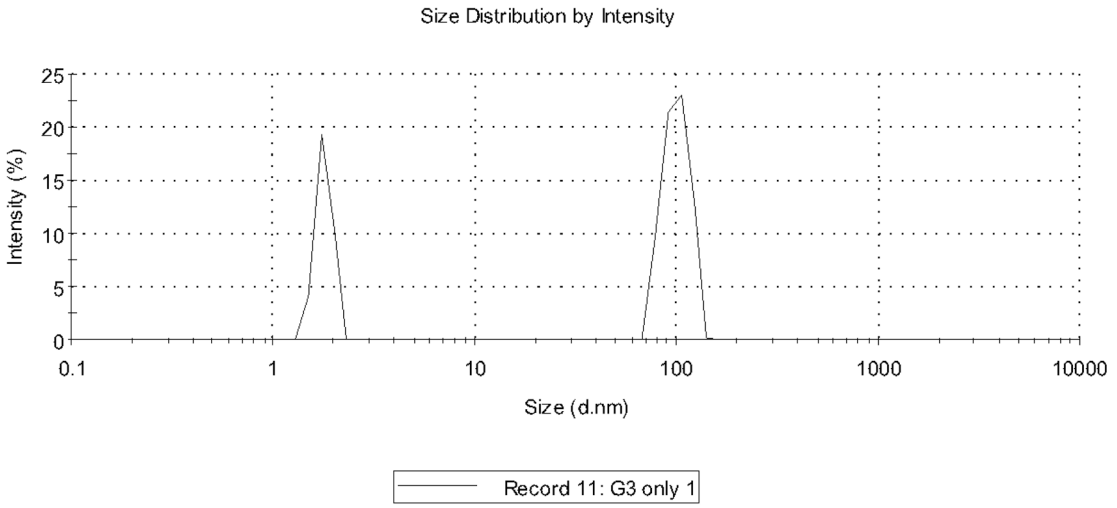


Figure S26. DLS of **G3** in water (2.4 mM of amine). Maxima at 2 nm and 100nm.

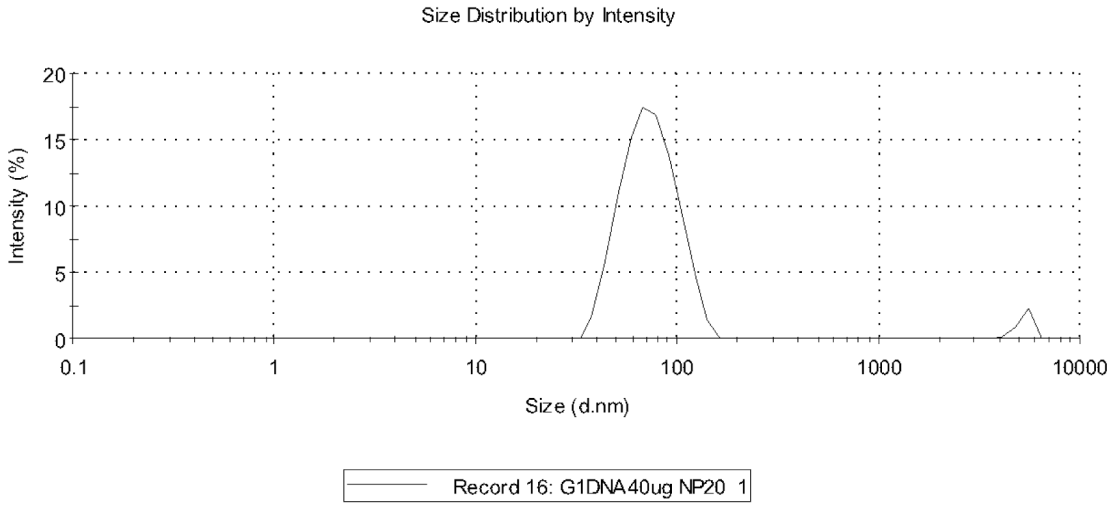


Figure S27. DLS of **G1** with DNA (40 μg/mL) at N/P ratio 20. Maxima at 75 nm.

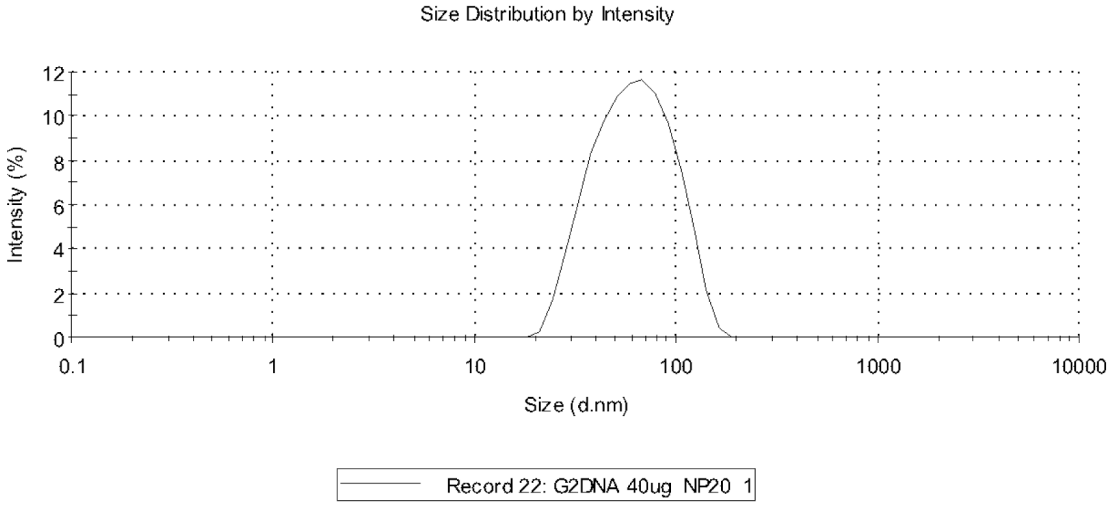


Figure 28. DLS of **G2** with DNA (40 μg/mL) at N/P ratio 20. Maxima at 66 nm.

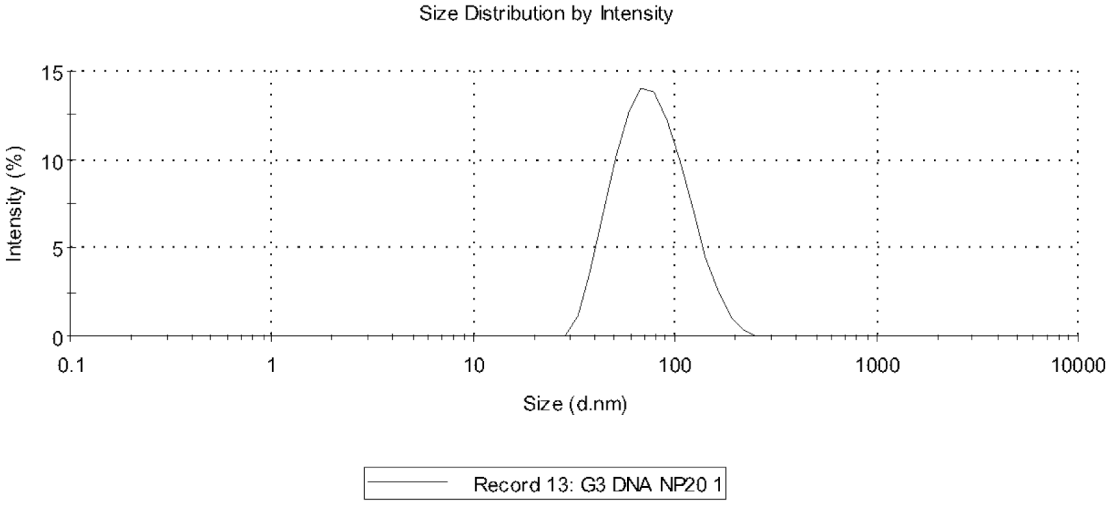


Figure S29. DLS of **G3** with DNA (40 μg/mL) at N/P ratio 20. Maxima at 81 nm.

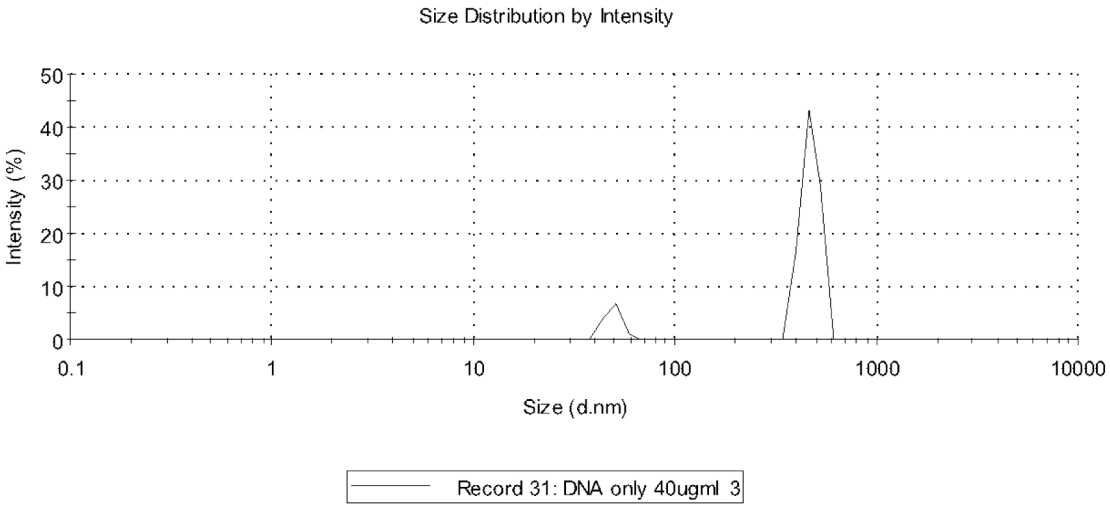


Figure S30. DLS of DNA only (40 μg/mL). Maximums at 49 nm and 471 nm.

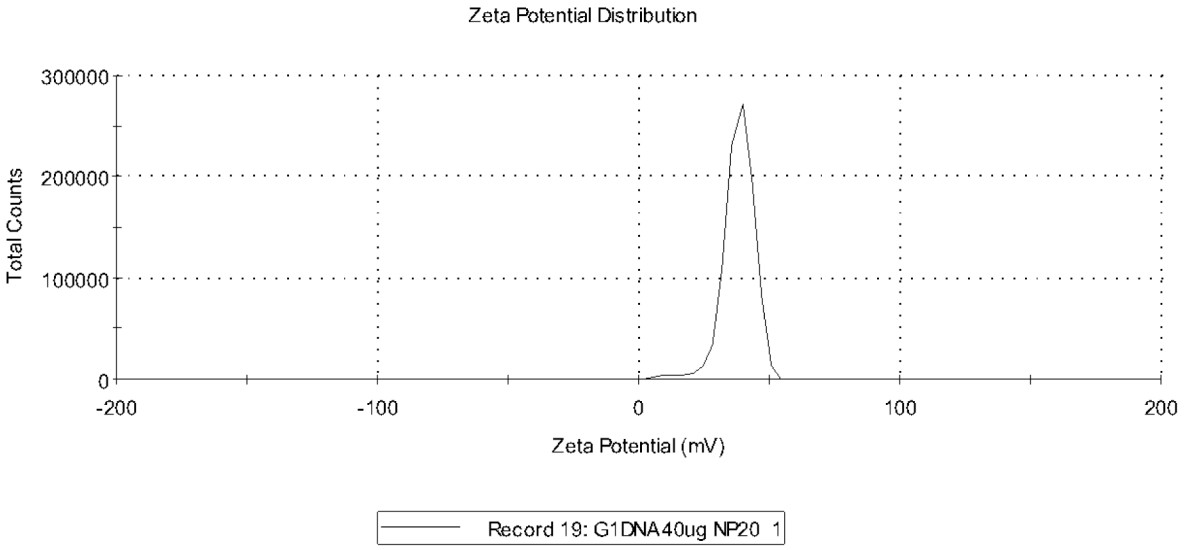


Figure S31. Zeta potential of **G1** with DNA (40 μg/mL) at N/P ratio 20. Maximum at 38 mV.

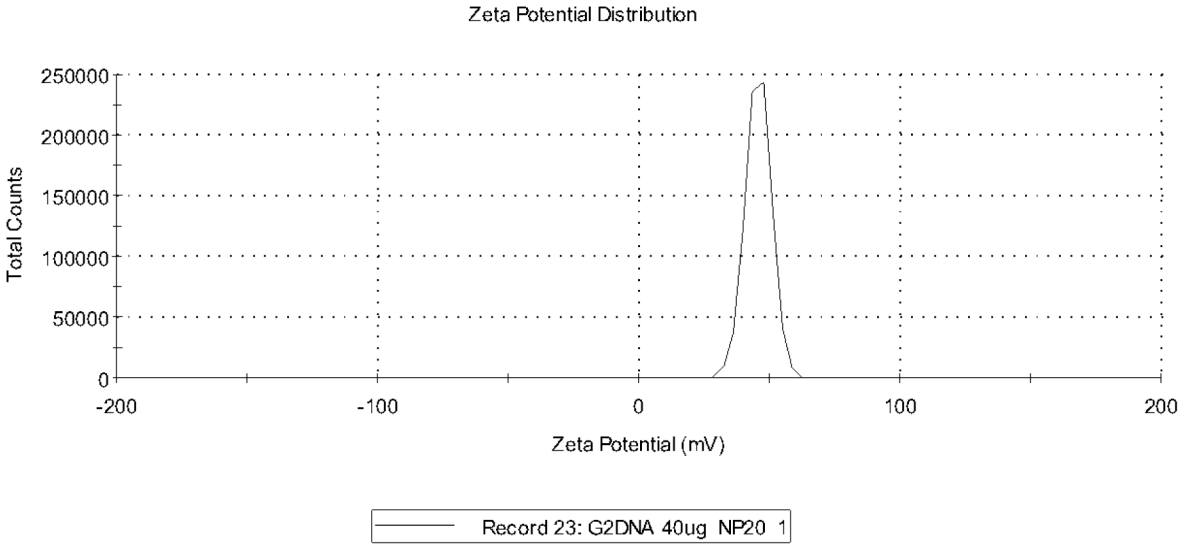


Figure S32. Zeta potential of **G2** with DNA (40 μg/mL) at N/P ratio 20. Maximum at 46 mV.

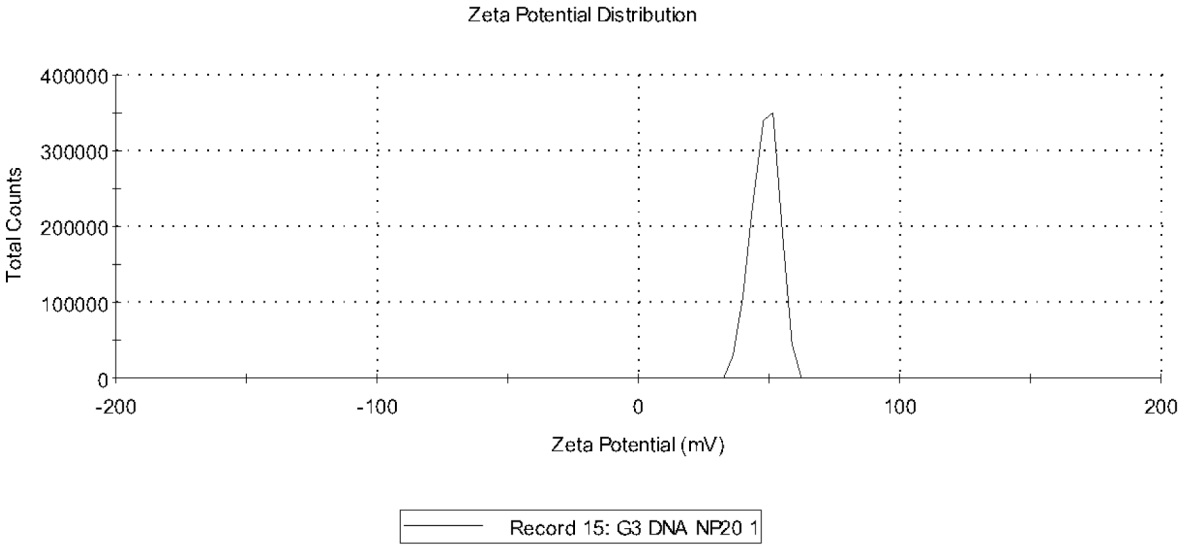


Figure S33. Zeta potential of **G3** with DNA (40 μg/mL) at N/P ratio 20. Maximum at 49 mV.

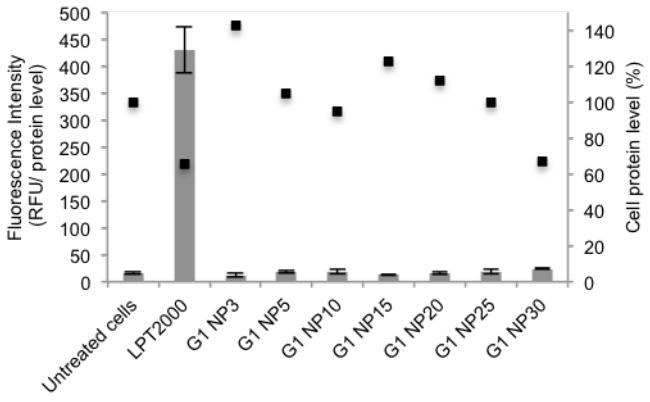
Gene Transfection Experiments

Cell Culture. HeLa cells were grown in Eagle's Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% FBS, L-glutamine (2 mM), penicillin (100 units/mL) and streptomycin (100 µg/mL). Cells were maintained at 37°C in a 5% CO_2 humidified atmosphere and all experiments were done in triplicates. The day before experiment, cells were seeded in 24-multiwell plates at 50.10^3 cells/well in fresh complete medium (1 mL).

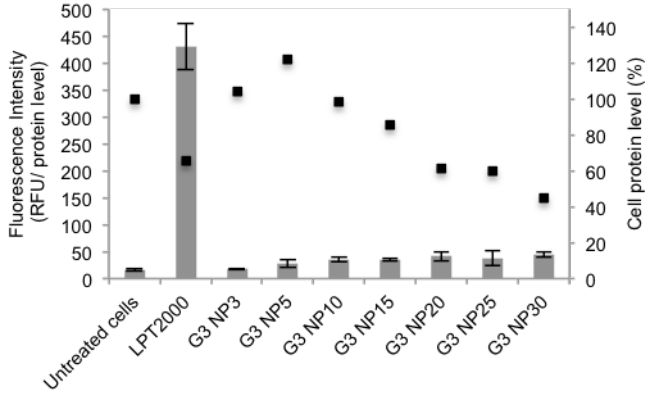
Polyplexes formation for pEGFP delivery. The procedure is for a 24-multiwell plates experiment. Typically, an aqueous solution of the G*n* (*n* = 1, 2 or 3) (volume depending on the desired N/P ratio) was diluted up to 50 µL in water containing 5% glucose. The solution was vortexed and left for 10 min. Separatly, an aqueous solution of pEGFP (corresponding to 2 µg of pEGFP) was diluted up to 50 µL in water containing 5% glucose. Then, the solution was vortexed and left for 10 min, after which the G1, G2 or G3 solution was added to the pEGFP solution, and vortexed (15 s). Finally, the polyplexes were incubated for 30 min at room temperature and added in each well by dilution with the cell medium without serum (1 mL). Four hours later, each well was completed with serum (0.1 mL). The gene expression profiles were analyzed 24 h after addition of polyplexes.

Quantification of the EGFP gene expression. EGFP gene expression was determined 24 h after delivery. The cell medium was removed and the wells were washed with PBS (1 mL) twice. Lysis buffer RIPA (0.1 mL) was added to each well and incubated at RT for 20 min. The lysat was centrifuged at 12000 rpm for 15 min. 50 µL of supernatant was transferred to a 96-multiwell plate and the fluorescence of EGFP was measured with a plate reader (λex : 485 nm, λem : 525 nm). Fluorescence intensity was normalized per mg of cell protein by using the BCA protein assay (ThermoFischerScientific). The errors bars represent standard deviation derived from

triplicate experiments. Lipofectamine 2000 is a commercial transfection reagent (Life Technologies), which was used according to manufacturer's instructions.



a)



b)

Figure S34. Gene delivery experiments of pEGFP on Hela cells at various N/P (polyplexes prepared in 5% glusoce solutions) for: a) G1, and b) G3. Fluorescence intensity (bars) and percentage of total cellular proteins (squares) are given for negative control (untreated HeLa cells), positive control (lipofectamine 2000). Means and standard deviation of triplicate experiments are given.