

Synthesis of Fullerene Glycoconjugates through Sulfide Connection in Aqueous Media

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Supporting Information

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Experimental details and spectral data	S1-S5
NMR and mass spectra	S6-S15

Spectral Measurement. IR spectra were obtained on an ASI Applied Systems REACT IR1000 equipped with an attenuated total reflection (ATR) instrument. NMR spectra were obtained on JEOL ECX-400 and ECA-500 spectrometers (^1H : 400 and 500 MHz; ^{13}C : 100 and 125 MHz). ^1H NMR spectra in CDCl_3 were referenced internally to tetramethylsilane as a standard, and spectra in CD_3OD and $\text{DMSO}-d_6$ were internally referenced to solvent resonance. Mass spectra were obtained on JEOL JMS-T100LC (AccuTOF; APCI/ESI-TOF MS) and Shimadzu MALDI4 (MALDI-TOF MS) instruments.

Synthesis of Penta-thiol 2: The fullerene cyclopentadiene bearing tetrahydropyranyl protected thiophenol moieties was synthesized according to the literature method.¹ The protected compound (261 mg, 0.155 mmol) was dissolved in *o*-dichlorobenzene (155 mL) under nitrogen, and 2-mercaptoethanol (78 mL) and trifluoroacetic acid (78 mL) were sequentially added to the solution. On addition of trifluoroacetic acid, the temperature of the reaction mixture slightly rose. The reaction mixture was then heated at 40 °C for 1 h, and the reaction mixture was poured into water (200 mL). The reaction mixture was diluted with chlorobenzene (100 mL) and was washed with water (200 mL \times 3) and a diluted aqueous

(1) Sawamura, M.; Iikura, H.; Nakamura, E. *J. Am. Chem. Soc.* **1996**, *118*, 12850; Nakamura, E.; Sawamura, M. *Pure Appl. Chem.* **2001**, *73*, 355.

solution of sodium hydrogencarbonate (0.24 M, 100 mL). The organic layer was concentrated to 10 mL, and hexane (100 mL) was added in one portion to afford orange precipitates. The precipitate was filtrated, and dried under reduced pressure to afford 187 mg of **2** (0.148 mmol, 95%).

THP-Protected Penta-thiol (mixture of diastereomers): IR (powder) 2935, 2846, 2360, 2342, 1590, 1490, 1187, 1077, 1034, 1003, 810 cm^{-1} ; ^1H NMR (400 MHz, CDCl_3) δ 1.56–1.72 (m, 15H), 1.75–1.94 (m, 10H), 1.95–2.08 (m, 5H), 3.50–3.64 (m, 5H), 4.07–4.21 (m, 5H), 5.13–5.18 (m, 3H), 5.18–5.24 (m, 3H), 7.23–7.28 (m, 2H), 7.30 (dd, J = 1.6, 8.4 Hz, 4H), 7.42–7.50 (m, 10H), 7.66 (d, J = 8.0 Hz, 4H); ^{13}C NMR (100 MHz, CDCl_3) δ 21.69, 21.79, 25.49, 25.53, 31.51, 58.41, 58.55, 60.56, 62.75, 64.57, 64.71, 64.76, 84.93, 85.01, 85.05, 85.12, 127.97, 128.23, 128.37, 130.91, 131.11, 131.15, 131.22, 134.60, 134.70, 134.72, 135.13, 137.79, 137.82, 137.92, 143.00, 143.47, 143.71, 143.91, 143.99, 144.09, 144.13, 144.18, 145.11, 145.36, 145.55, 145.59, 145.66, 146.67, 146.87, 146.96, 147.36, 147.53, 147.88, 148.04, 148.18, 148.46, 148.50, 148.55, 151.15, 151.19, 151.76, 152.33, 155.73, 155.76; HRMS (APCI-TOF, negative) m/z calcd for $\text{C}_{115}\text{H}_{66}\text{O}_5\text{S}_5$ $[\text{M}-\text{H}]^-$ 1685.3436, found: 1685.3363.

Penta-thiol 2: IR (powder) 3049, 2935, 2564, 2360, 2337, 1783, 1590, 1490, 1102, 1015, 830, 749 cm^{-1} ; ^1H NMR (500 MHz, $\text{CDCl}_3/\text{CS}_2$ 3/7) δ 3.41 (s, 1H), 3.46 (s, 2H), 3.52 (s, 2H), 5.13 (s, 1H), 7.08 (d, J = 8.0 Hz, 2H), 7.12 (d, J = 8.0 Hz, 4H), 7.21 (d, J = 8.0 Hz, 2H), 7.23-7.26 (overlapped with CHCl_3) 7.41 (d, J = 8.4 Hz, 4H), 7.61 (d, J = 8.0 Hz, 4H); HRMS (APCI-TOF, negative) m/z calcd for $\text{C}_{90}\text{H}_{26}\text{S}_5$ $[\text{M}-\text{H}]^-$ 1265.0560, found: 1265.0531.

Benzylated Penta-thiol 4a: Under nitrogen, penta-thiol **2** (25.0 mg, 19.7 μmol) was suspended in degassed THF (9 mL), and a degassed aqueous solution of sodium hydroxide (0.10 M, 1.2 mL) and benzyl bromide (11.8 μL , 99.0 μmol) were added. Degassed 1 M HCl (1 mL) was added to the reaction mixture after 2 h, and the mixture was stirred for an additional 1 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain benzylated penta-thiol **4a** which weighed 32.5 mg (18.9 μmol , 96% yield). IR (powder) 2963, 1489, 1452, 1396, 1260, 1091, 1069, 1014, 798, 756, 693, 668 cm^{-1} ; ^1H NMR (500 MHz, CDCl_3) δ 4.05 (s, 2H), 4.07 (s, 4H), 4.12 (s, 4H), 5.12 (s, 1H), 7.03 (d, J = 8.5 Hz, 2H), 7.08 (d, J = 8.5 Hz, 4H), 7.21-7.33 (overlapped m, 31H), 7.40 (d, J = 8.5 Hz, 4H),

7.56 (d, $J = 8.5$ Hz, 4H); ^{13}C NMR (100 MHz, CDCl_3) δ 38.53 (CH_2), 38.66 (CH_2), 38.68 (CH_2), 58.32, 58.44, 60.43, 62.78 (CH), 127.33 (CH), 127.34 (CH), 127.40 (CH), 127.95 (CH), 128.38 (CH), 128.40 (CH), 128.57 (CH), 128.59 (CH), 128.62 (CH), 128.71 (CH), 128.73 (CH), 128.75 (CH), 128.78 (CH), 129.52 (CH), 129.81 (CH), 135.84, 136.06, 136.25, 137.03, 137.19, 137.20, 137.42, 137.46, 143.17, 143.30, 143.51, 144.05, 144.14, 144.22, 144.24, 144.26, 144.35, 145.18, 145.49, 145.68, 145.77, 146.85, 147.04, 147.14, 147.40, 147.71, 148.06, 148.10, 148.22, 148.36, 148.64, 148.72, 148.76, 151.12, 151.90, 152.38, 155.72; HRMS (APCI-TOF, negative) m/z calcd for $\text{C}_{125}\text{H}_{56}\text{S}_5$ $[\text{M-H}]^-$ 1715.2907, found: 1715.2895.

2-Hydroxyethylated Penta-thiol 4b: Under nitrogen, penta-thiol **2** (20.0 mg, $15.8\ \mu\text{mol}$) was suspended in degassed THF (5 mL), and a degassed aqueous solution of sodium hydroxide (0.1 M, 0.95 mL) and 2-bromoethanol ($5.6\ \mu\text{L}$, $79\ \mu\text{mol}$) were added. Degassed 1 M HCl (0.5 mL) was added to the reaction mixture after 3 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain 2-hydroxyethylated penta-thiol **4b** which weighed 20.2 mg ($13.7\ \mu\text{mol}$, 87% yield). ^1H NMR spectrum was measured with solvent suppression by dante presaturation method. IR (powder) 3571-3249, 2943, 1616, 1489, 1462, 1398, 1092, 1045, 1013, 810, 767, $673\ \text{cm}^{-1}$; ^1H NMR (500 MHz, $\text{CD}_3\text{OD}/\text{CS}_2$ 3/7) δ 3.03 (t, $J = 6.5$ Hz, 2H), 3.06 (t, $J = 7.0$ Hz, 4H), 3.12 (t, $J = 7.0$ Hz, 4H), 3.67 (t, $J = 6.5$ Hz, 2H), 3.70 (t, $J = 7.0$ Hz, 4H), 3.74 (t, $J = 7.0$ Hz, 4H), 5.31 (s, 1H), 7.14 (d, $J = 8.5$ Hz, 2H), 7.21 (d, $J = 8.5$ Hz, 4H), 7.27 (d, $J = 8.5$ Hz, 2H), 7.39 (d, $J = 8.5$ Hz, 4H), 7.52 (d, $J = 8.5$ Hz, 4H), 7.76 (d, $J = 8.5$ Hz, 4H); HRMS (APCI-TOF, negative) m/z calcd for $\text{C}_{100}\text{H}_{46}\text{O}_5\text{S}_5$ $[\text{M-H}]^-$ 1485.1871, found: 1485.1880.

Hydroxycarbonylmethylated Penta-thiol 4c: Under nitrogen, penta-thiol **2** (127 mg, $100\ \mu\text{mol}$) was suspended in degassed THF (30 mL), and a degassed aqueous solution of sodium hydroxide (200 mM, 25 mL) and sodium iodoacetate (104 mg, $500\ \mu\text{mol}$) in water (25 mL) were added. Degassed 1M HCl (5 mL) was added to the reaction mixture after 5.5 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain hydroxycarbonylmethylated penta-thiol **4c** which weighed 125 mg ($84\ \mu\text{mol}$, 84% yield). IR (powder) 3450-3290, 1714, 1594, 1489, 1398, 1142, 1087, 1055, 1034, 1007, 885, 809, $667\ \text{cm}^{-1}$; ^1H NMR (400 MHz, $\text{CD}_3\text{OD}/\text{CS}_2$ 7/3) δ 3.65 (s, 2H), 3.68 (s, 4H),

3.72 (s, 4H), 5.30 (s, 1H), 7.21 (d, $J = 8.4$ Hz, 2H), 7.27 (d, $J = 8.4$ Hz, 6H), 7.43 (d, $J = 8.4$ Hz, 4H), 7.51 (d, $J = 8.4$ Hz, 4H), 7.75 (d, $J = 8.4$ Hz, 4H); HRMS (APCI-TOF, negative) m/z calcd for $C_{100}H_{36}O_{10}S_5$ $[M-H]^-$ 1555.0834, found: 1555.0857.

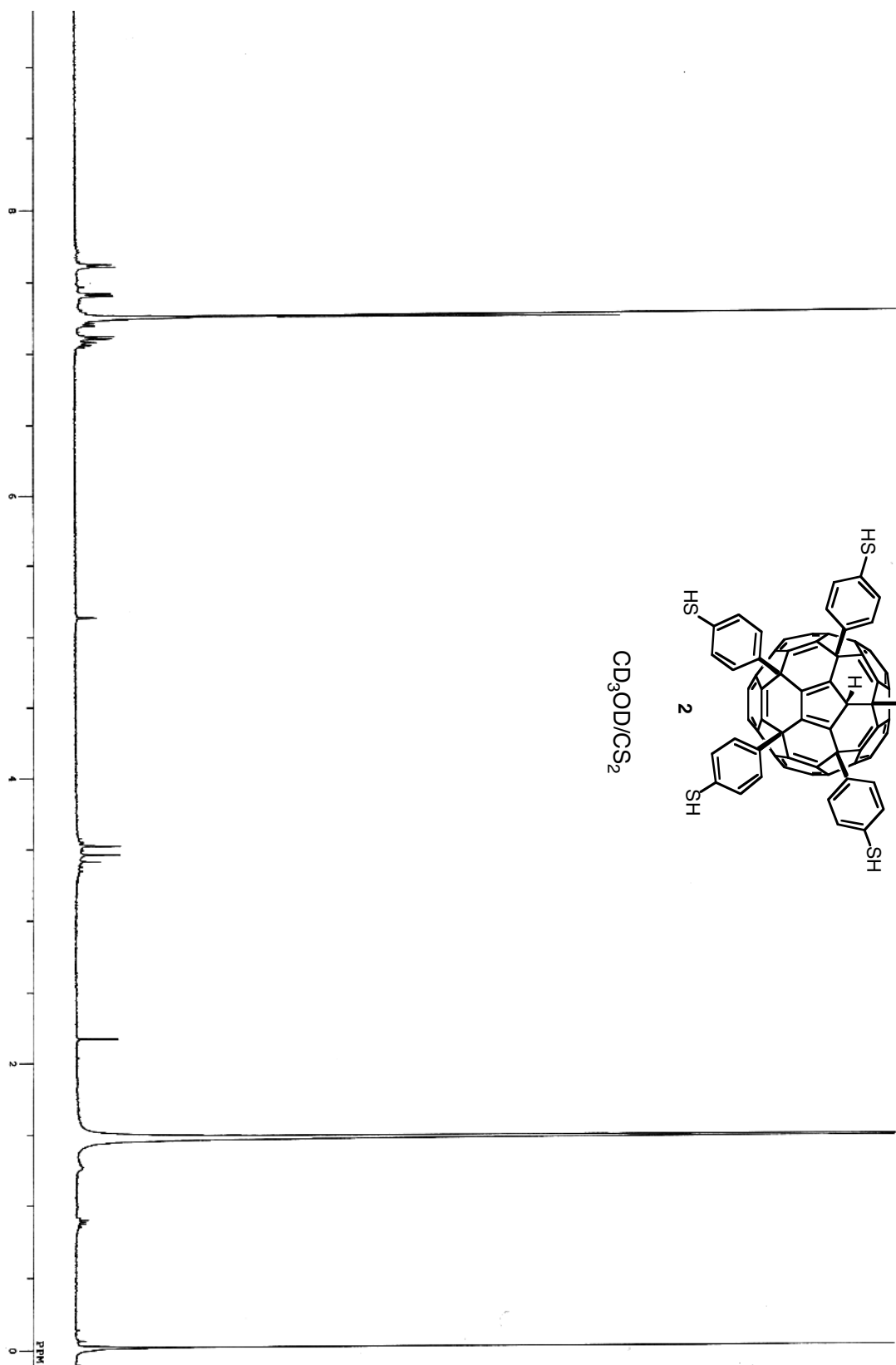
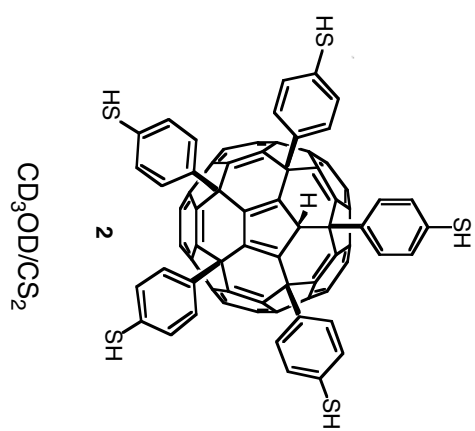
Mannoside 4d: The electrophile, 2-bromoethyl α -D-mannoside, was prepared according to the literature method.² Under nitrogen, penta-thiol **2** (20.0 mg, 15.8 μ mol) was suspended in degassed THF (5 mL), and a degassed aqueous solution of sodium hydroxide (0.10 M, 1.0 mL) and 2-bromoethyl α -D-mannoside (22.8 mg, 79.4 μ mol) in water (1.6 mL) were added. Degassed 1 M HCl (0.1 mL) and ethanol (1 mL) were added to the reaction mixture after 9 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain mannoside **4d** which weighed 26.5 mg (11.5 μ mol, 73% yield). 1H NMR spectrum in NaOH/D₂O/DMSO- d_6 was measured under a solvent suppression mode by the dante presaturation method at 30 °C. MS/MS analysis of the compound exhibited some low-MW signals due to the fragmentation. IR (powder) 3338, 2929, 1420, 1277, 1131, 1088, 1048, 972, 806, 677 cm^{-1} ; 1H NMR (500 MHz, DMSO- d_6 (0.6 mL) + 0.5 M NaOH/D₂O (40 μ L)) δ 3.07 (dt, $J = 11$, 6.5 Hz, 5H), 3.12 (dt, $J = 11$, 6.5 Hz, 5H), 3.37-3.40 (m, 10H), 3.41-3.47 (m, 10H), 3.54 (dt, $J = 11$, 6.5 Hz, 5H), 3.57-3.61 (m, 10H), 3.72 (dt, $J = 11$, 6.5 Hz, 5H), 4.64 (br s, 5H), 7.14 (br d, $J = 8.0$ Hz, 10H), 7.72 (br d, $J = 8.0$ Hz, 10H); MS (ESI-TOF, negative) m/z calcd for $C_{130}H_{96}O_{30}S_5$ $[M-H]^-$ 2296; $[M-2H]^{2-}$ 1148, found: 2296; 1148, MS (MALDI-TOF, positive) m/z calcd for $C_{130}H_{96}O_{30}S_5$ $[M+Na]^+$ 2319, found 2318.

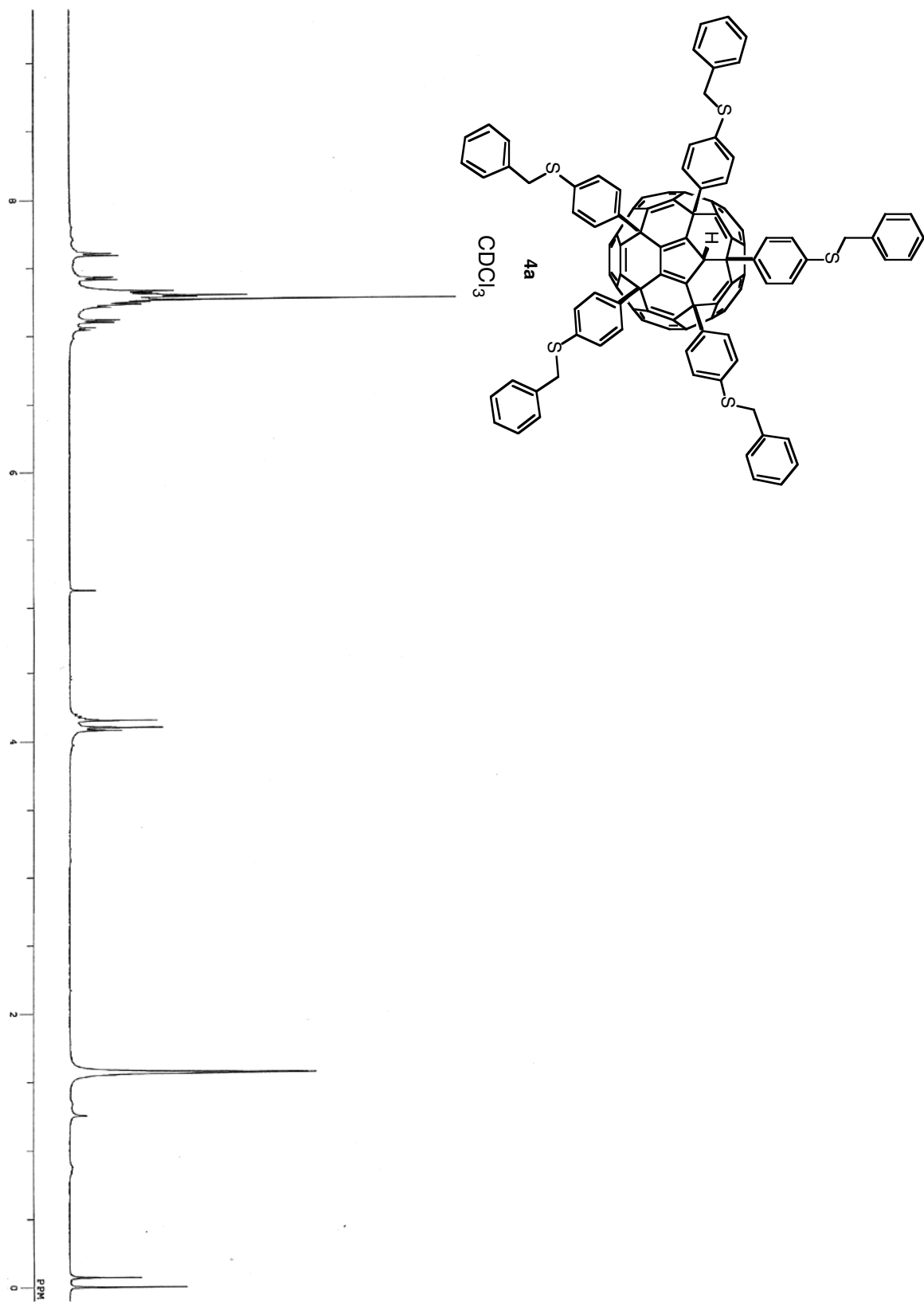
Glucoside 4e: Under nitrogen, penta-thiol **2** (20.0 mg, 15.8 μ mol) was suspended in degassed THF (5 mL), and a degassed aqueous solution of sodium hydroxide (0.10 M, 1.0 mL) and 2-bromoethyl β -D-glucoside (22.8 mg, 79.4 μ mol) in water (1.6 mL) were added. Degassed 1 M HCl (0.1 mL) and ethanol (1 mL) were added to the reaction mixture after 9 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain glucoside **4e** which weighed 28.7 mg (12.5 μ mol, 79% yield). Addition of NaOH/D₂O to a solution of **4e** in DMSO- d_6 drastically reduced the signal intensities likely

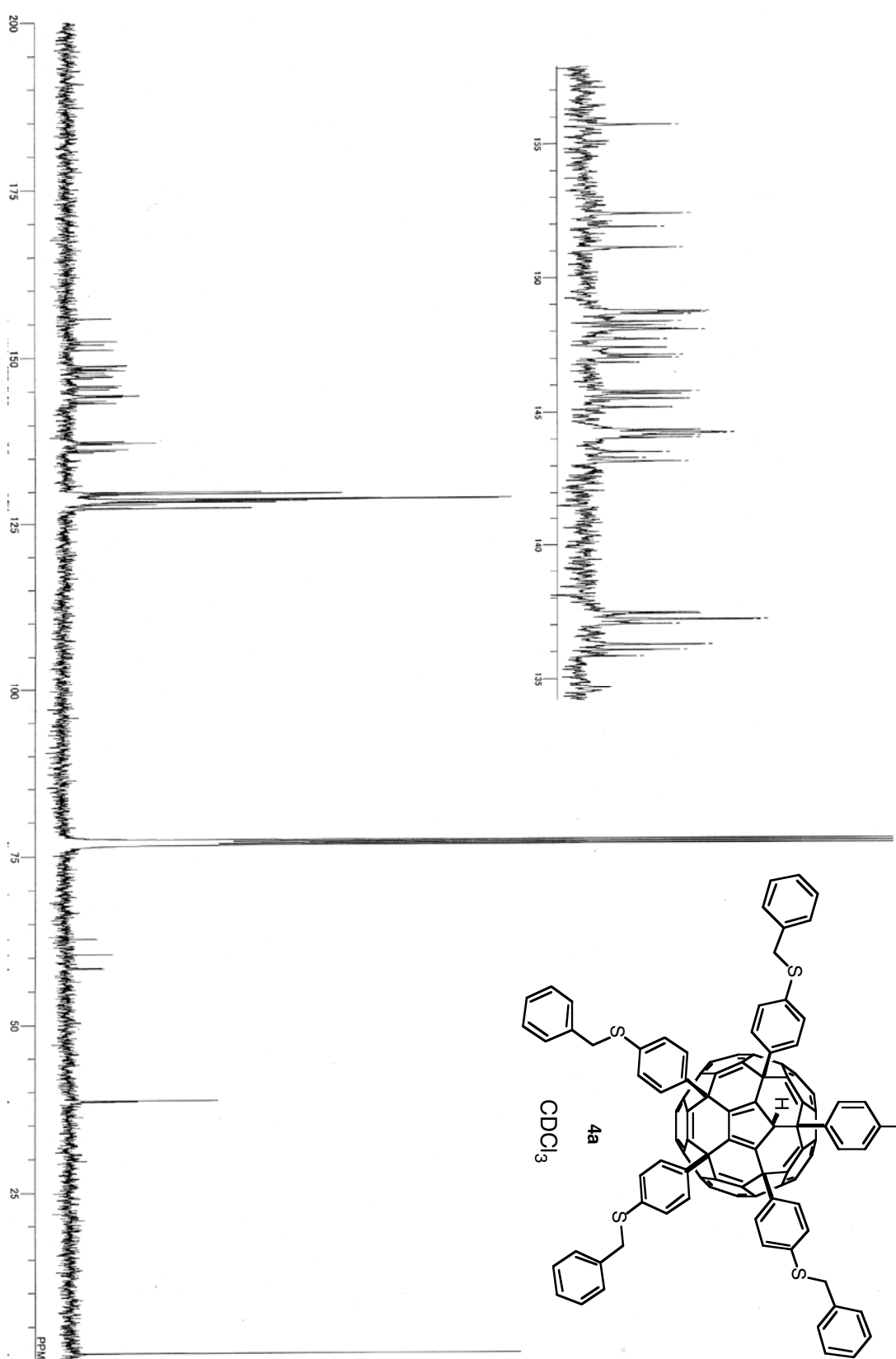
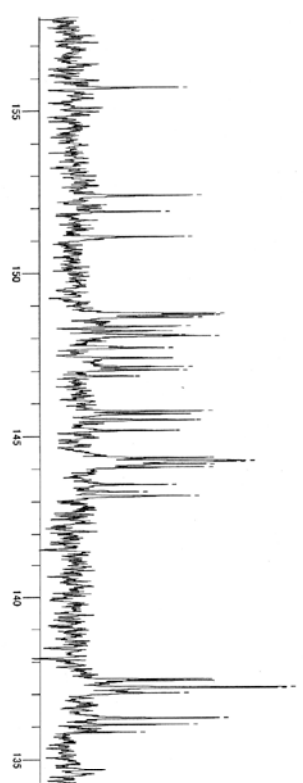
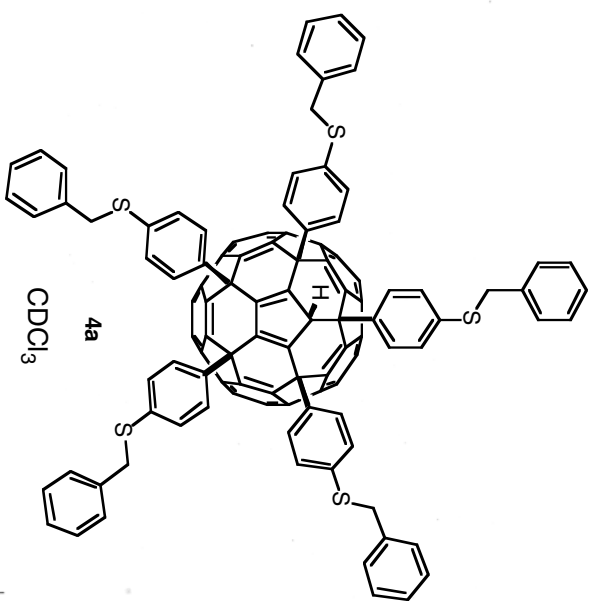
(2) Dahmén, J.; Frejd, T.; Magnusson, G.; Noori, G.; Carlström, A. -S. *Carbohydr. Res.* **1984**, 125, 237; Davis, B. G.; Maughan, M. A. T.; Green, M. P.; Ullman, A.; Jones, J. B. *Tetrahedron: Asymmetry* **2000**, 11, 245.

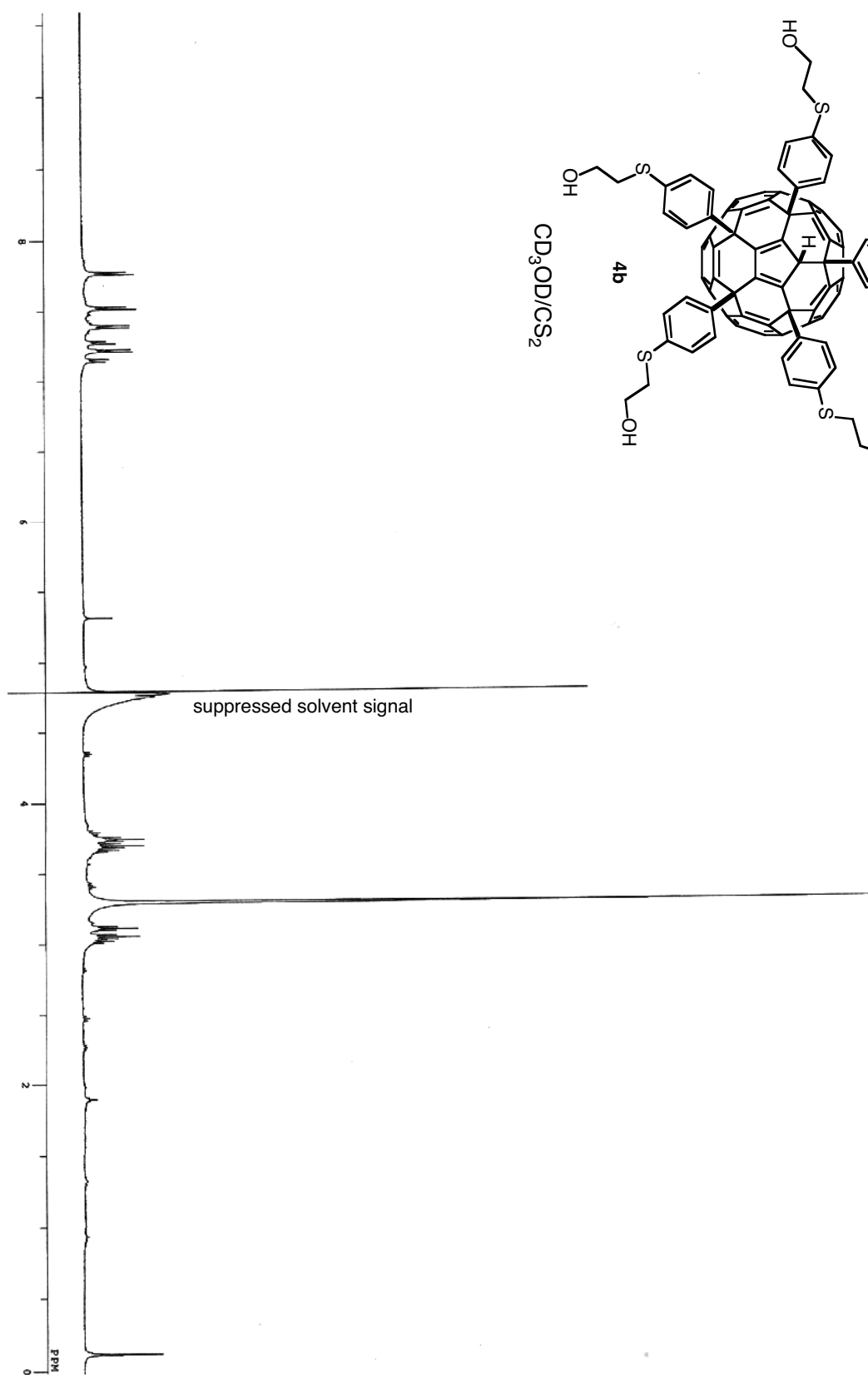
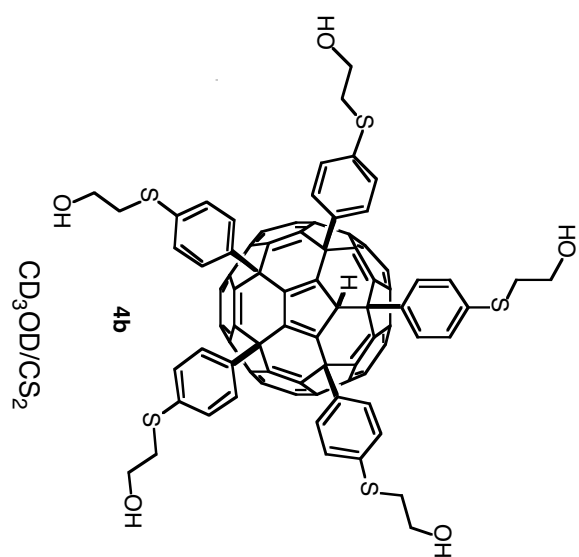
due to aggregation of the compound. The ^1H NMR spectrum in $\text{DMSO-}d_6$ shows three sets of doublets (2:2:1 in intensities) due to three non-equivalent para-substituted benzene moieties attached to the fullerene core. This observation confirms that **4e** has quasi C_5 symmetry possessing five sulfide linkages. IR (powder) 3338, 2879, 1592, 1443, 1355, 1281, 1160, 1071, 1013, 895, 673 cm^{-1} ; ^1H NMR (500 MHz, $\text{DMSO-}d_6$) δ 2.90-3.05 (overlapped m), 3.60-3.70 (overlapped m), 3.71-3.87 (overlapped m), 4.14 (d, $J = 7.5$ Hz, 1H), 4.18 (d, $J = 7.5$ Hz, 2H), 4.20 (d, $J = 7.5$ Hz, 2H), 5.80 (s, 1H), 7.12 (d, $J = 8.0$ Hz, 2H), 7.20 (d, $J = 8.0$ Hz, 2H), 7.23 (d, $J = 8.0$ Hz, 4H), 7.38 (d, $J = 8.0$ Hz, 4H), 7.71 (d, $J = 8.0$ Hz, 4H), 7.89 (d, $J = 8.0$ Hz, 4H); ^1H NMR (500 MHz, $\text{DMSO-}d_6$ (0.6 mL) + 0.5 M NaOH/ D_2O (40 μL)) δ 2.84-3.13 (overlapped br m), 3.42-3.48 (overlapped br m), 3.67-3.69 (overlapped br m), 4.03 (d, $J = 7.5$ Hz, 5H), 6.12 (d, $J = 8.5$ Hz, 10H), 6.85 (d, $J = 8.5$ Hz, 10H); MS (ESI-TOF, negative) m/z calcd for $\text{C}_{130}\text{H}_{96}\text{O}_{30}\text{S}_5$ $[\text{M-H}]^-$ 2296; $[\text{M-2H}]^{2-}$ 1148, found: 2296; 1148.

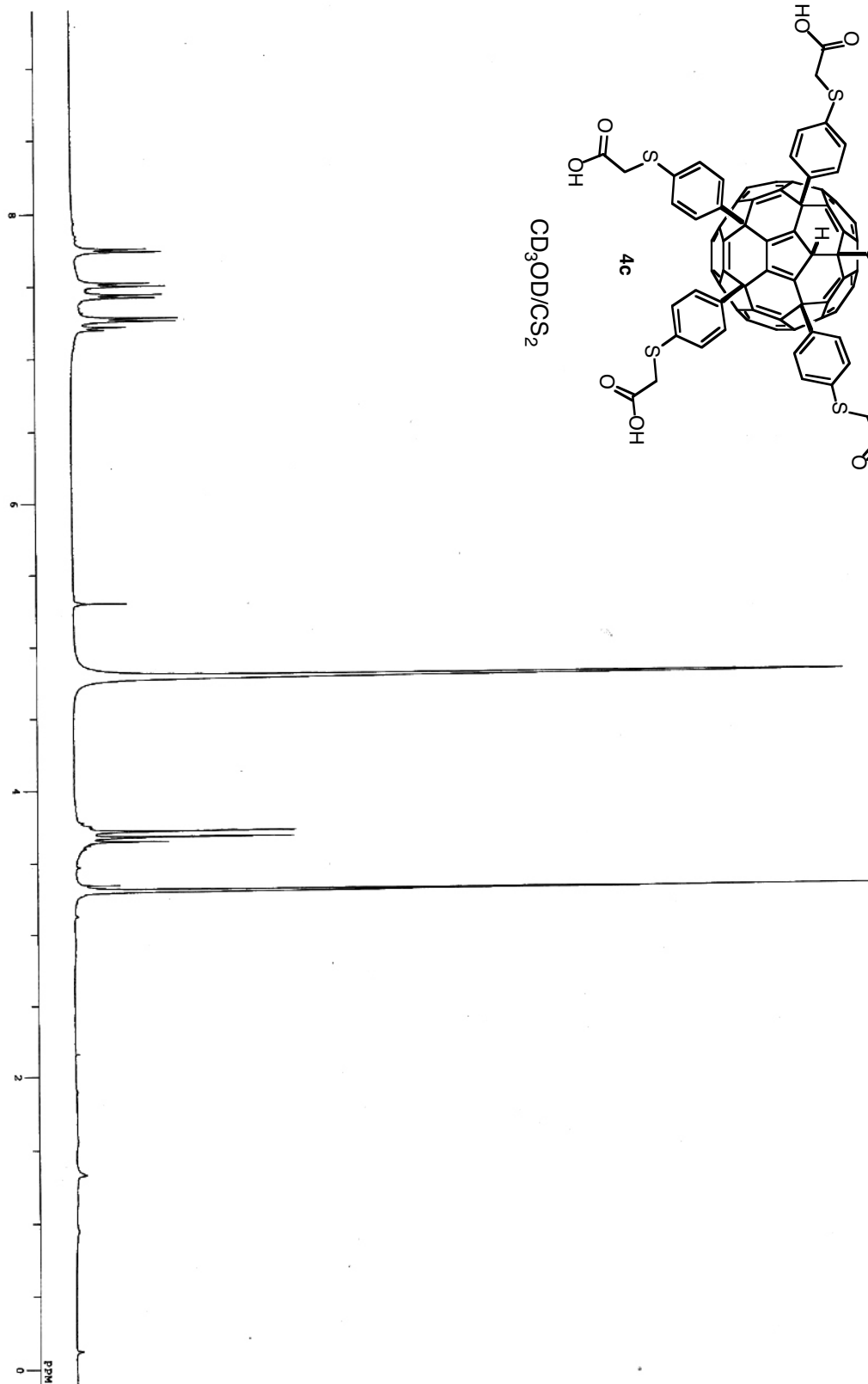
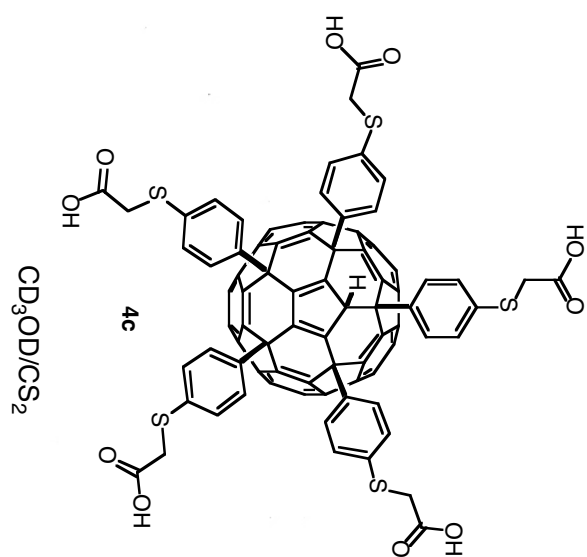
Galactoside 4f: Under nitrogen, penta-thiol **2** (20.0 mg, 15.8 μmol) was suspended in degassed THF (5 mL), and a degassed aqueous solution of sodium hydroxide (0.10 M, 1.0 mL) and 2-bromoethyl β -D-galactoside (22.8 mg, 79.4 μmol) in water (1.6 mL) were added. Degassed 1 M HCl (0.1 mL) and ethanol (1 mL) were added to the reaction mixture after 24 h to obtain orange precipitates. The precipitates were filtrated, washed with water and dried under reduced pressure to obtain galactoside **4f** which weighed 31.6 mg (13.7 μmol , 87% yield). IR (powder) 3424-3211, 2876, 1489, 1062, 1013, 885, 764, 676 cm^{-1} ; ^1H NMR (500 MHz, $\text{DMSO-}d_6$ (0.6 mL) + 0.5 M NaOH/ D_2O (40 μL)) δ 3.10 (dt, $J = 9.5, 7.5$ Hz, 5H), 3.13 (dt, $J = 9.5, 7.5$ Hz, 5H), 3.15-3.30 (m, 15H), 3.34 (t, $J = 6.5$ Hz, 5H), 3.58-3.61 (m, 10H), 3.64 (dt, $J = 9.5, 7.5$ Hz, 5H), 3.82 (dt, $J = 9.5, 7.5$ Hz, 5H), 4.13 (d, $J = 6.5$ Hz, 5H), 7.17 (d, $J = 8.0$ Hz, 10H), 7.76 (d, $J = 8.0$ Hz, 10H); MS (ESI-TOF, positive) m/z calcd for $\text{C}_{130}\text{H}_{96}\text{O}_{30}\text{S}_5$ $[\text{M+Na}]^+$ 2319; $[\text{M+2Na}]^{2+}$ 1172, found: 2319; 1172.

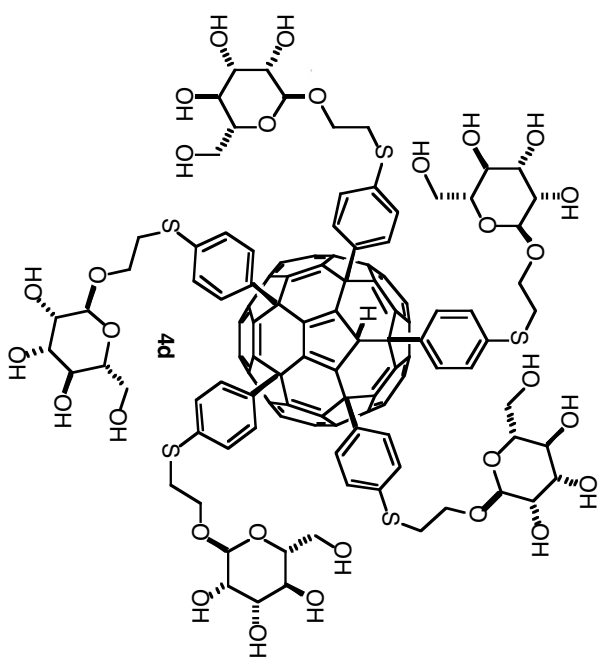




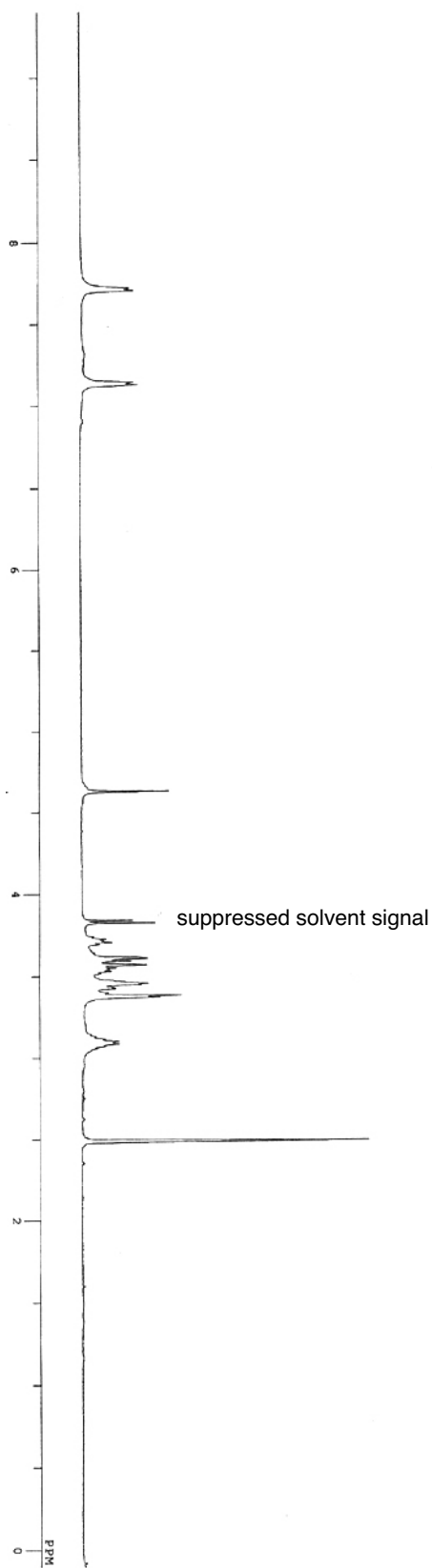






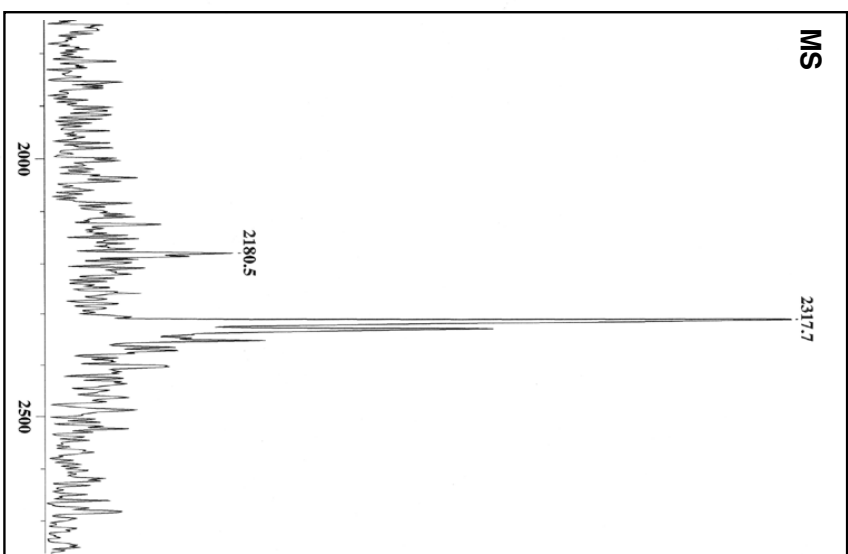
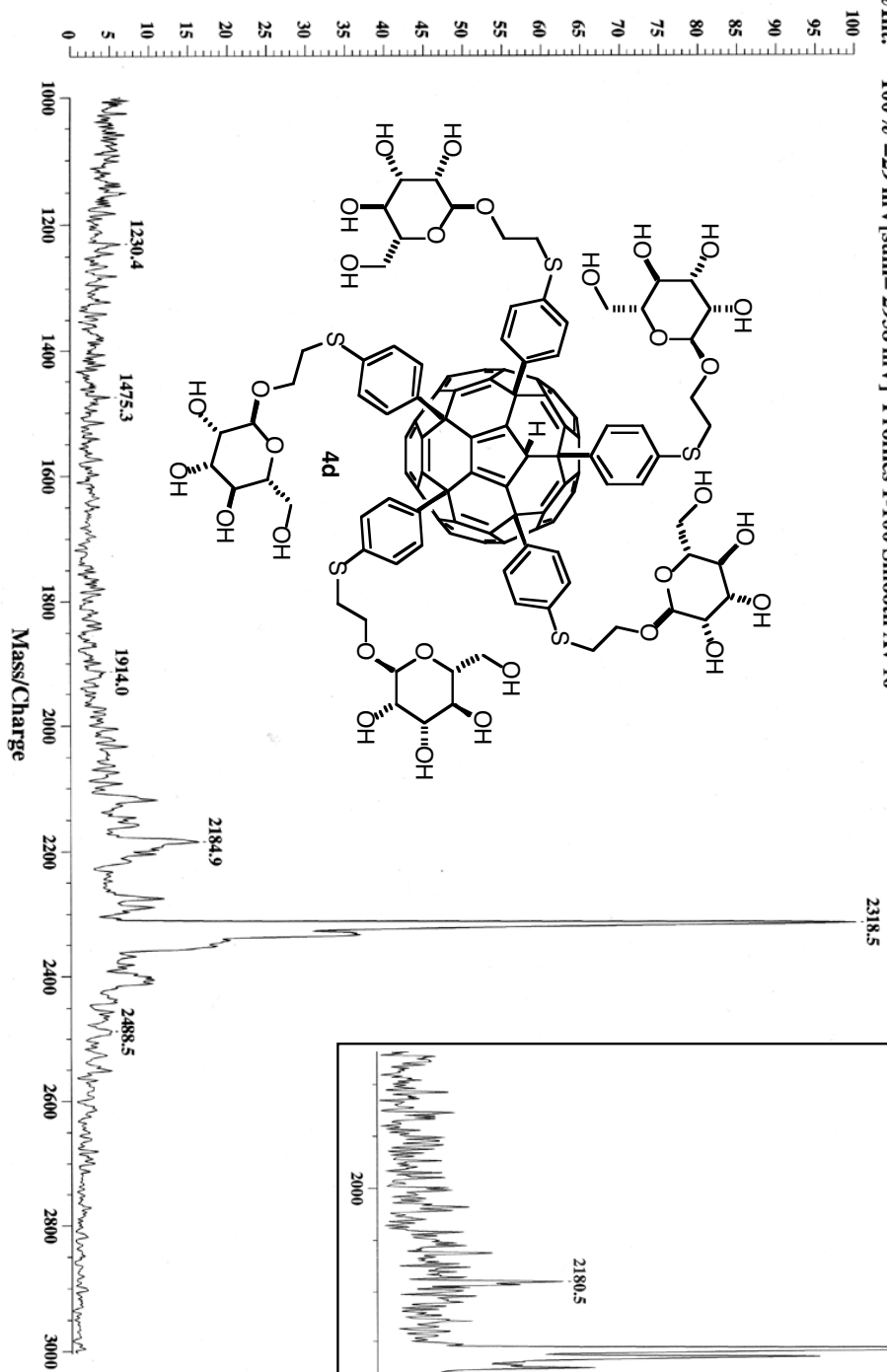


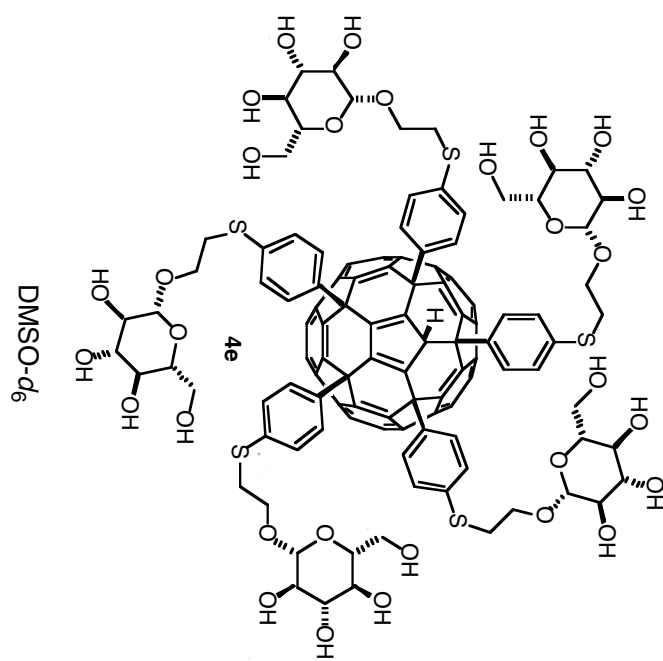
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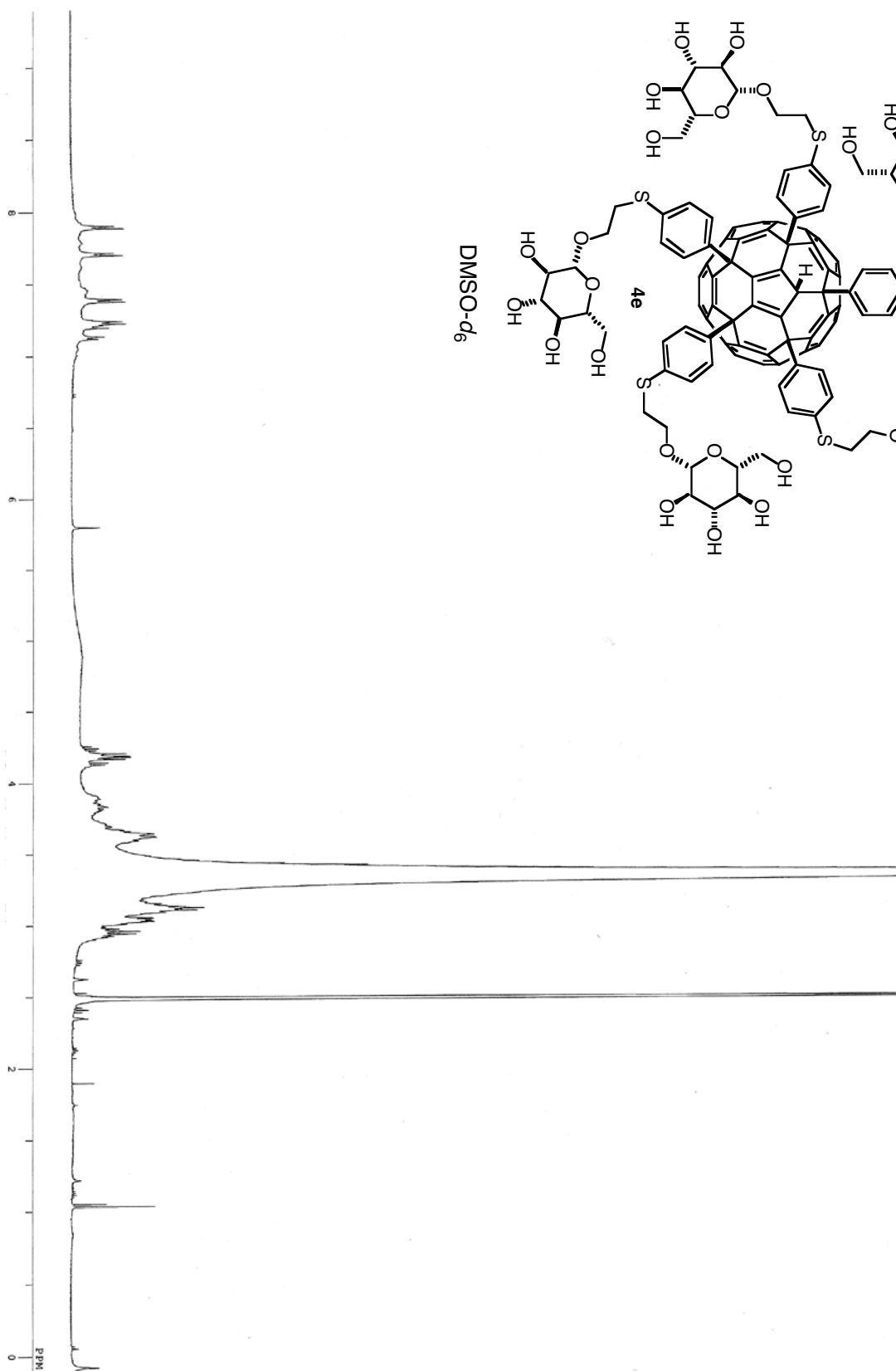
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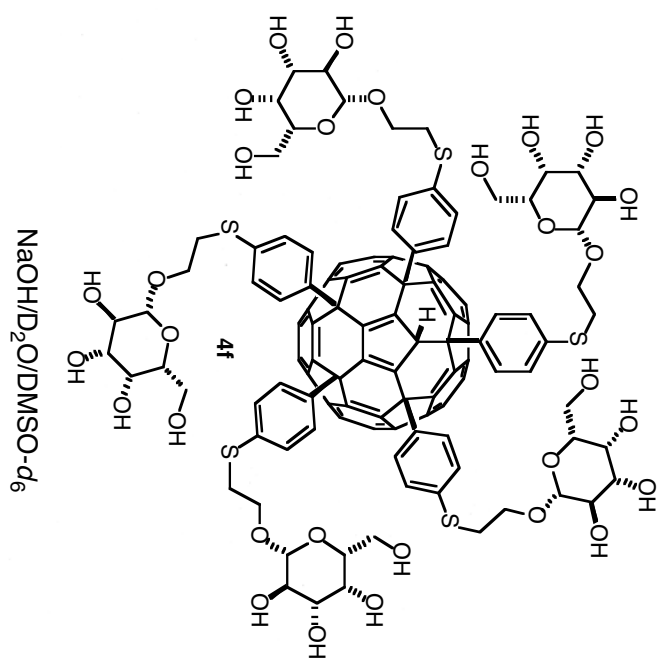




DMSO- d_6



NaOH/D₂O/DMSO-*d*₆



NaOH/D₂O/DMSO-*d*₆

