Electronic Supplementary Material (ESI) for Organic Chemistry Frontiers.

Electronic Supplementary Information for

Dynamics of Mechanically Bonded Macrocycles in Radial Hetero[4]Catenane Isomers

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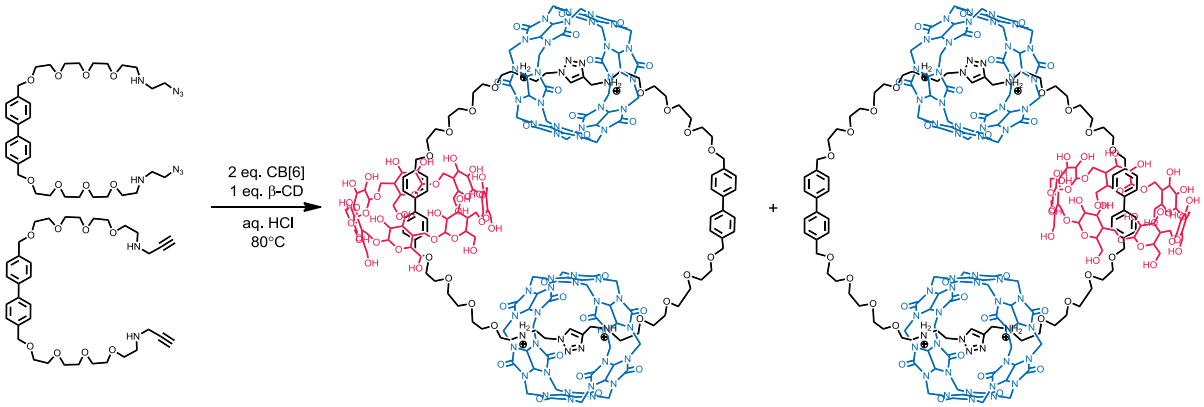
1. Synthesis

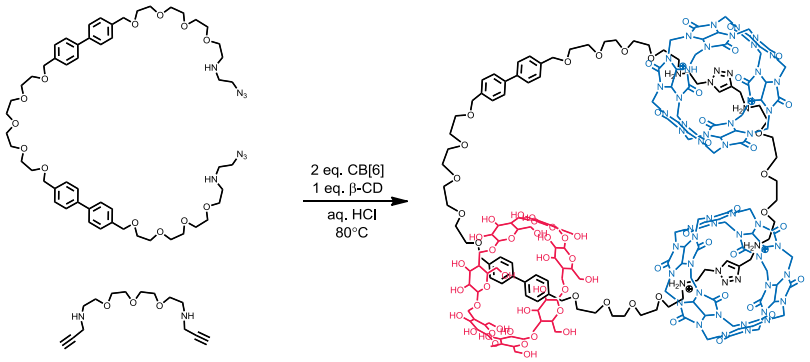
General. All reagents were purchased from commercial suppliers (Dkmchem, J & K and Aldrich) and used without further purification. Solvents for synthesis were of analytical grade (ACI Labscan and DUKSAN Pure Chemicals). MeCN and DMF were distilled over CaH_2 before use. **BP1-Azide**,[1] **BP1-Alkyne**,[1] **TEG-Alkyne**,[1] **BP2-Azide**,[1] and cucurbit[6]uril (CB[6])[2] were synthesized according to literature procedures. HPLC analyses were carried out using a Waters-Alliance e2695 system coupled to a 2489 UV/Vis detector. ESI-MS were carried out using a Waters-Acquity UPLC H-Class system coupled with a QDa MS detector. NMR spectra were recorded on Bruker DPX spectrometers with working frequencies of 500 MHz for ^1H, and 125 MHz for ^{13}C, respectively. Chemical shifts are reported in ppm and referenced to solvent residues (for ^1H: D_2O: δ = 4.79 ppm).



Scheme S1.

4C-1. A mixture of **BP1-azide** (14 mg, 0.02 mmol) and β-CD (23 mg, 0.02 mmol) in 50 mM aq. HCl (10 mL) was stirred at room temperature for 10 mins until a clear solution was obtained. A mixture of **BP1-Alkyne** (13 mg, 0.02 mmol) and CB[6] (40 mg, 0.04 mmol) in 50 mM aq. HCl (10 mL) was heated to 80 °C for 2 hr and was added to the **BP1-Azide/β-CD** solution over 1 hr. The reaction mixture was heated to 80 °C for overnight. HPLC analysis showed that *ca*. 72% of the materials in the product mixture corresponds to **4C-1**. The mixture was concentrated and the residue was re-dissolved in 1 mL of water. **4C-1** was isolated as a tetraformate salt by injecting 0.15 mL of the crude solution onto a preparative column (see details below). Isolated yield = 9 mg, 65 %. ^1H NMR (500 MHz, D_2O, 298 K) δ = 7.81 (m, 4H), 7.62

(br, 4H), 7.57 (d, *J* = 7.5 Hz, 4H), 7.49 (br, 4H), 6.48 (s, 2H), 5.75–5.64 (m, 24H), 5.43 (m, 24H), 5.02 (d, *J* = 3.4 Hz, 7H), 4.68 (m, 6H), 4.25–4.12 (m, 32H), 4.03 (m, 4H), 3.93–3.80 (m, 20H), 3.78–3.55(m, 82H), 3.54–4.49 (m, 7H). $^{13}C\{^{1}H\}$ NMR (125 MHz, D_2O, 298 K) δ = 156.5, 156.1, 156.0, 129.0, 126.4, 120.0, 118.6, 102.1, 102.1, 80.9, 73.4, 72.0, 71.9, 70.1, 69.7, 69.7, 69.5, 68.8, 64.3, 59.6, 51.5, 51.2. HRMS (ESI+) calcd. for $C_{184}H_{252}N_{58}O_{75}$ $[M+4H]^{4+}$ (*m/z*): 1118.9429, found: 1118.9629.



Scheme S2.

4C-2. A mixture of **BP2-Azide** (21 mg, 0.02 mmol) and β-CD (23 mg, 0.02 mmol) in 50 mM aq. HCl (10 mL) was stirred at room temperature for 10 mins until a clear solution was obtained. A mixture of **BP0-Alkyne** (5 mg, 0.02 mmol) and CB[6] (40 mg, 0.04 mmol) in 50 mM aq. HCl (10 mL) was heated to 80 °C for 2 hr and was added to the **BP2-Azide/β-CD** solution over 1 hr. The reaction mixture was heated to 80 °C for overnight. HPLC analysis showed that *ca*. 82% of the materials in the product mixture corresponds to **4C-2**. The mixture was concentrated and the residue was re-dissolved to 1 mL of water. **4C-2** was isolated as a tetraformate salt from 0.15 mL of the crude solution by preparative HPLC (see details below). Isolated yield = 10 mg, 75% ^{1}H NMR (500 MHz, D_2O, 298 K) δ = 7.71 (d, *J* = 7.9 Hz, 2H), 7.66 (d, *J* = 7.9 Hz, 2H), 7.62 (d, *J* = 7.8 Hz, 2H), 7.60–7.56 (m, 4H), 7.51–7.45 (m, 6H), 6.51 (s, 1H), 6.48 (s, 1H), 5.75–5.63 (m, 24H), 5.46 (d, *J* = 19.0 Hz, 24H), 5.01 (d, *J* = 3.7 Hz, 7H), 4.69 (s, 2H), 4.67 (s, 2H), 4.60 (s, 2H), 4.53 (m, 2H), 4.27–4.19 (m, 24H), 4.17 (m, 4H), 4.13 (t, J = 4.4 Hz, 4H), 4.04 (t, J = 4.7 Hz, 4H), 3.93–3.80 (m, 20H), 3.78–3.53 (m, 80H), 3.50 (m, 7H). $^{13}C\{^{1}H\}$ NMR (125 MHz, D_2O, 298 K) δ = 156.5, 156.1, 128.9, 126.4, 121.7, 120.0, 118.6, 102.1, 80.9, 73.4, 71.9, 71.8, 70.1, 69.7, 59.6, 51.4, 51.2. HRMS (ESI+) calcd. for $C_{184}H_{252}N_{58}O_{75}$ $[M+4H]^{4+}$ (*m/z*): 1118.9429, found: 1118.9430.

2. HPLC Analysis

HPLC analyses were carried out using a Waters-Alliance e2695 system coupled to a 2489 UV/Vis detector. Distilled H_2O (Watsons), MeCN (DUKSAN Pure Chemicals) and formic acid (J & K) were used as received. A C18 SunFire preparative columns (5 μm, 10 × 250 mm) was used with the gradient elution described below. UV-Vis absorbance was monitored at 254 nm.

Elution Method (flow rate = 3.0 mL/min)

time/min	H_2O (with 0.1% FA)	MeCN (with 0.1% FA)
0	95%	5%
18	72%	28%
19	0%	100%
25	0%	100%

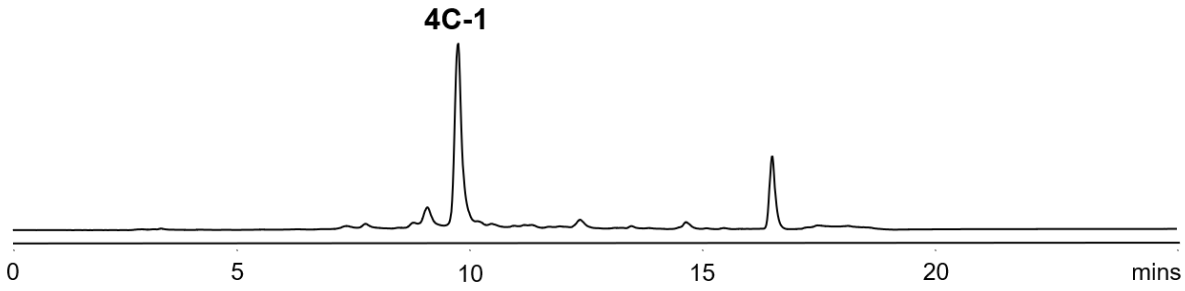


Figure S1. HPLC chromatogram of the crude product mixture of **4C-1**. The species eluted at 16.5 min is the [3]catenane with no interlocked β-CD.

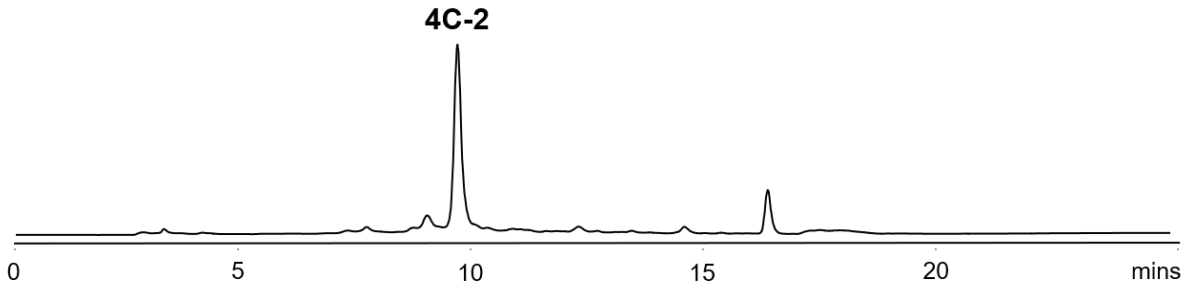


Figure S2. HPLC chromatogram of the crude product mixture of **4C-2**. The species eluted at 16.5 min is the [3]catenane with no interlocked β-CD.

3. NMR

*NMR studies of neutral forms of **4C-1** and **4C-2***

To a sample of 3.6 mM **4C-1** in D_2O was added 2 eq. of NaOH (30 µL of a 120 mM solution in D_2O). The solution was mixed well and the ^1H NMR spectrum was recorded. Additional amount of NaOH was added and ^1H NMR spectra of the [4]catenane with 4 eq. and 6 eq. of NaOH were obtained and the stacked spectra are shown in Figure S13. The neutral form of **4C-2** was studied similarly and the stacked spectra are shown in Figure S14.

*Dynamics of **4C-1** and **4C-2** by variable temperature ^1H NMR*

^1H NMR spectra were obtained from a 2 mM solution of the [4]catenanes in D_2O from 298 K to 348 K at intervals of 10 K. $\Delta G^{\ddagger}$ for the shuttling of β-CD between the two biphenylenes in **4C-2** was calculated using the Erying equation.

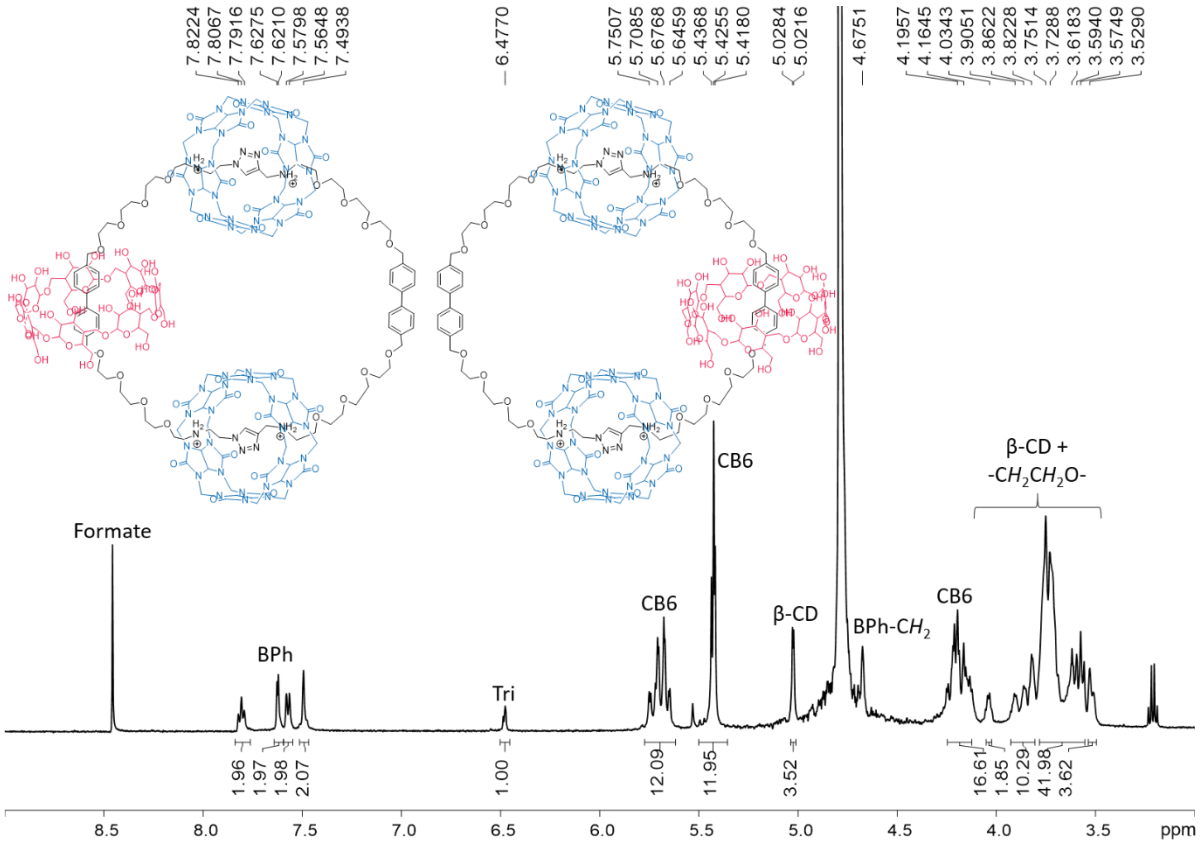


Figure S3. ^{1}H NMR (500 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-1**.

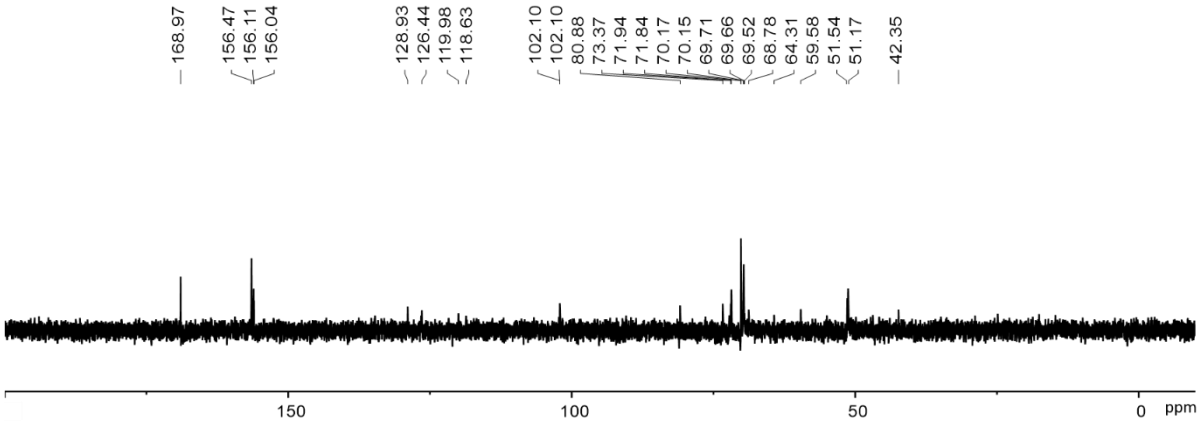


Figure S4. $^{13}C\{^{1}H\}$ NMR (125 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-1**.

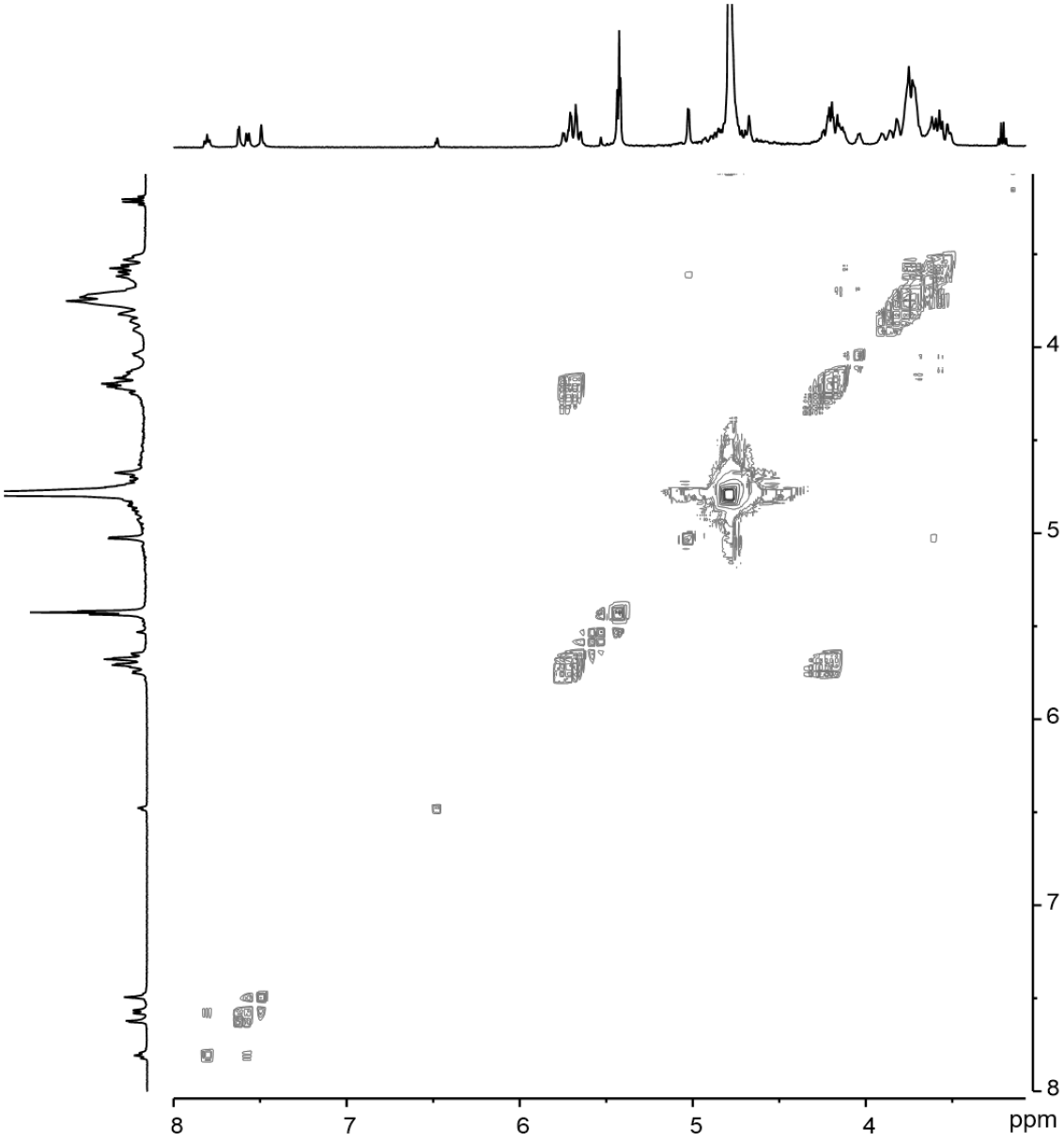


Figure S5. 2D COSY (500 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-1**.

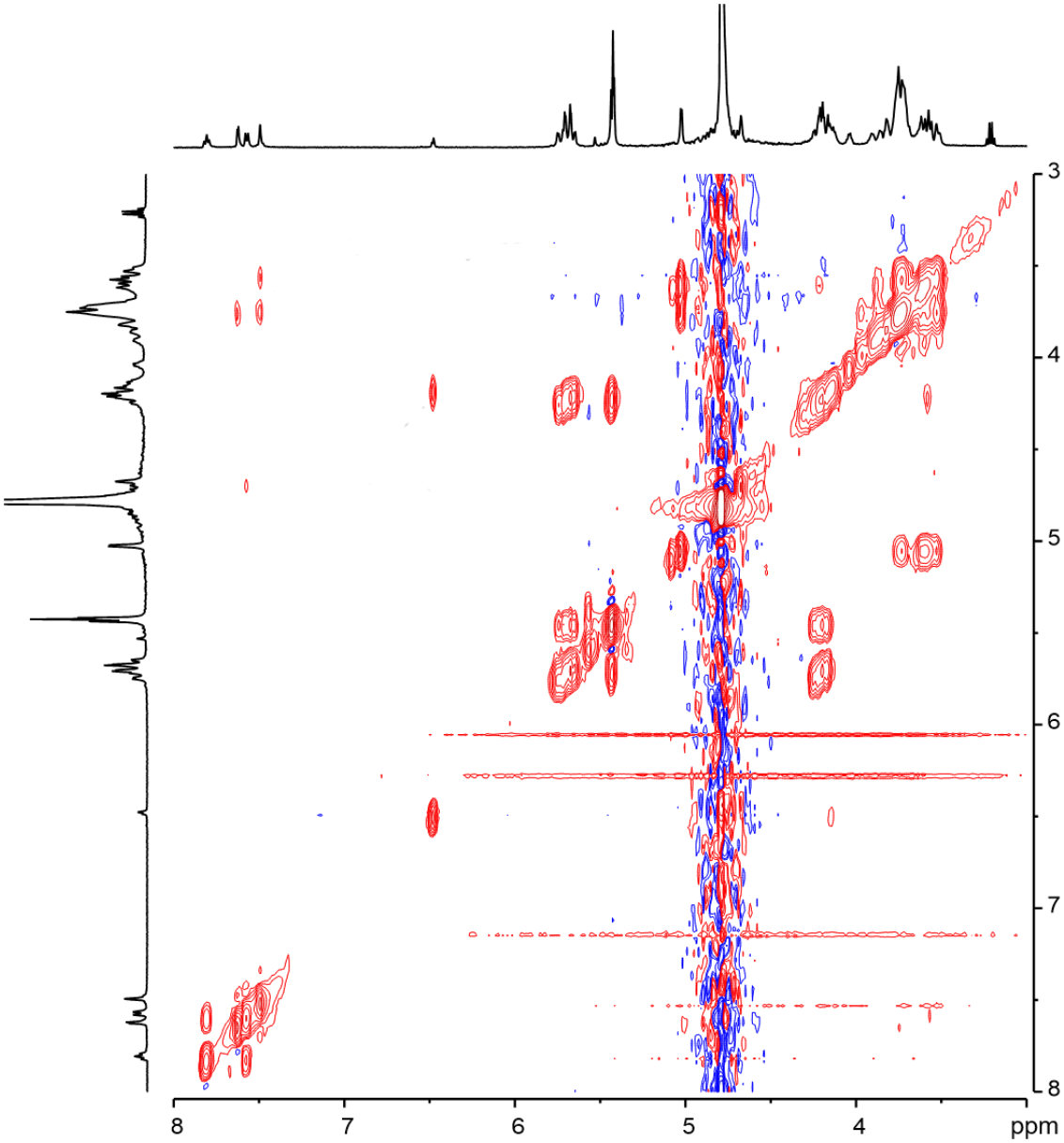


Figure S6. 2D NOESY (500 MHz, D_2O, 298 K, mixing time = 700 ms) spectrum of the formate salt of the tetra-protonated form of **4C-1**.

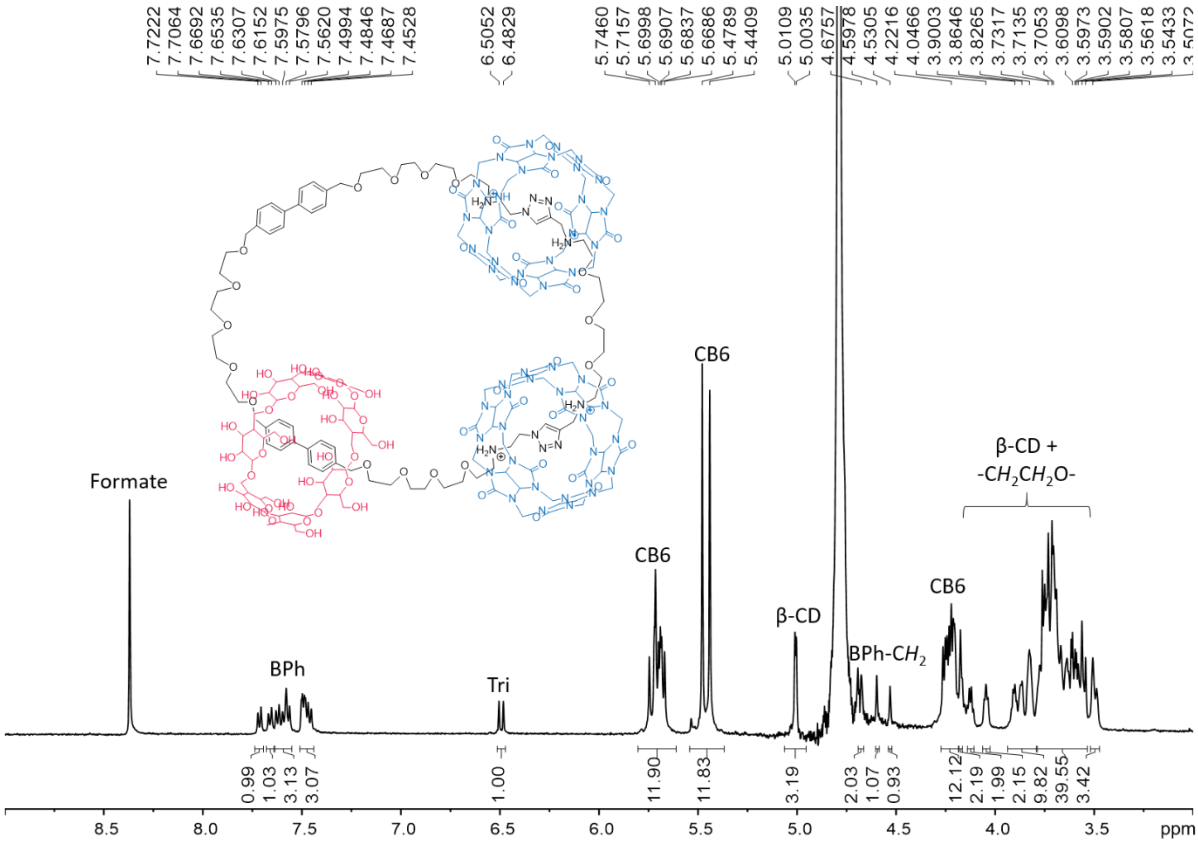


Figure S7. ^{1}H NMR (500 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-2**.

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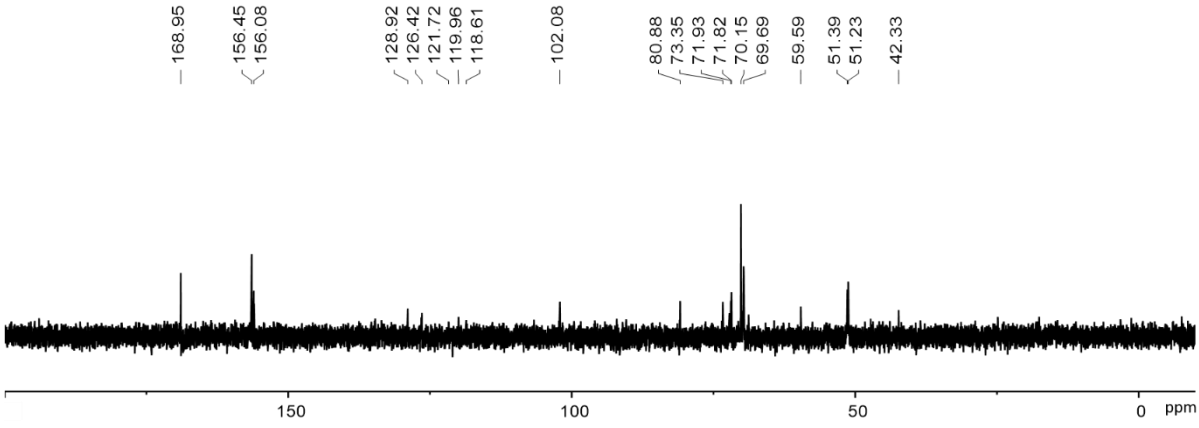

Figure S8. $^{13}C\{^{1}H\}$ NMR (125 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-2**.

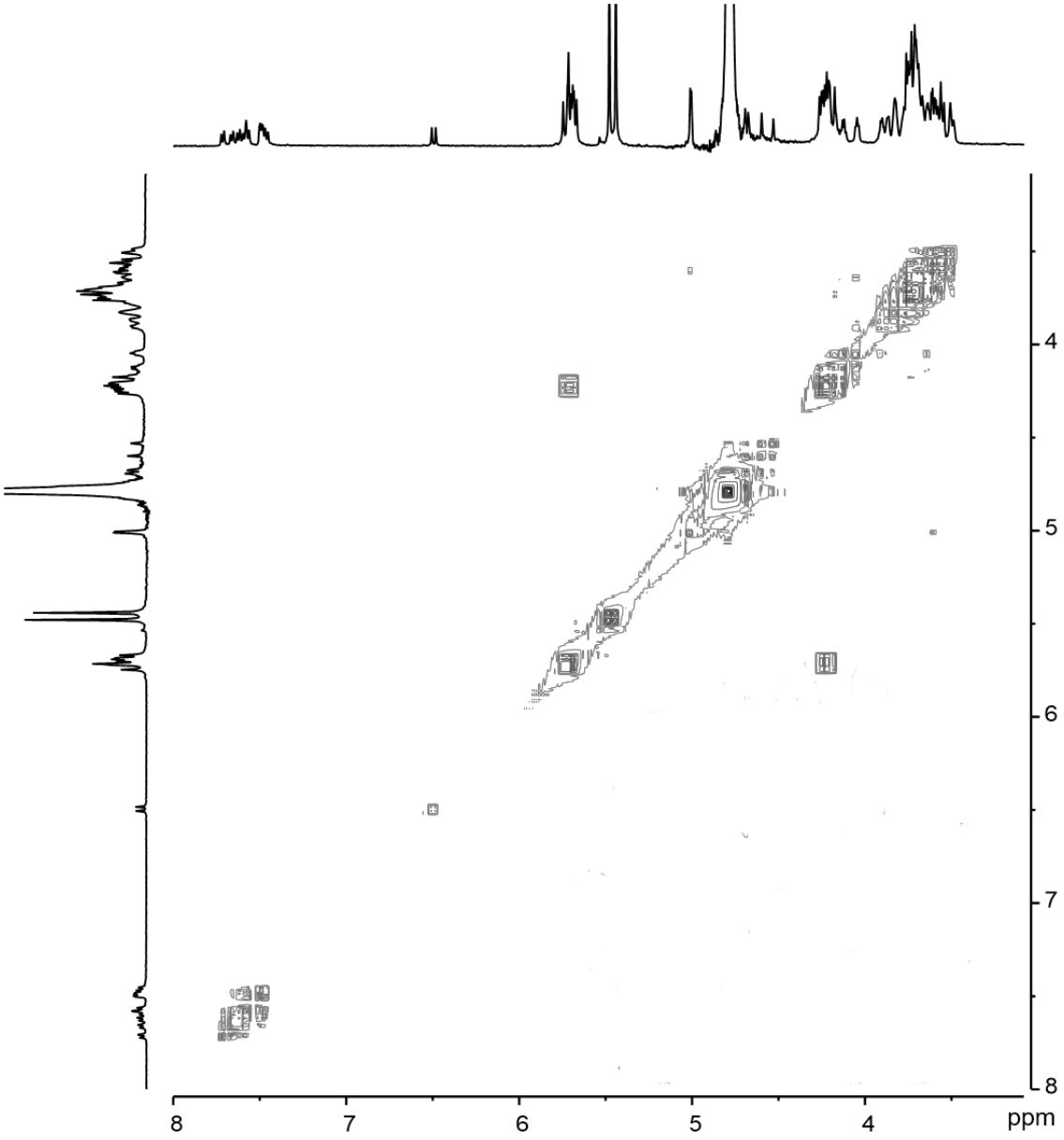

Figure S9. 2D COSY (500 MHz, D_2O, 298 K) spectrum of the formate salt of the tetra-protonated form of **4C-2**.

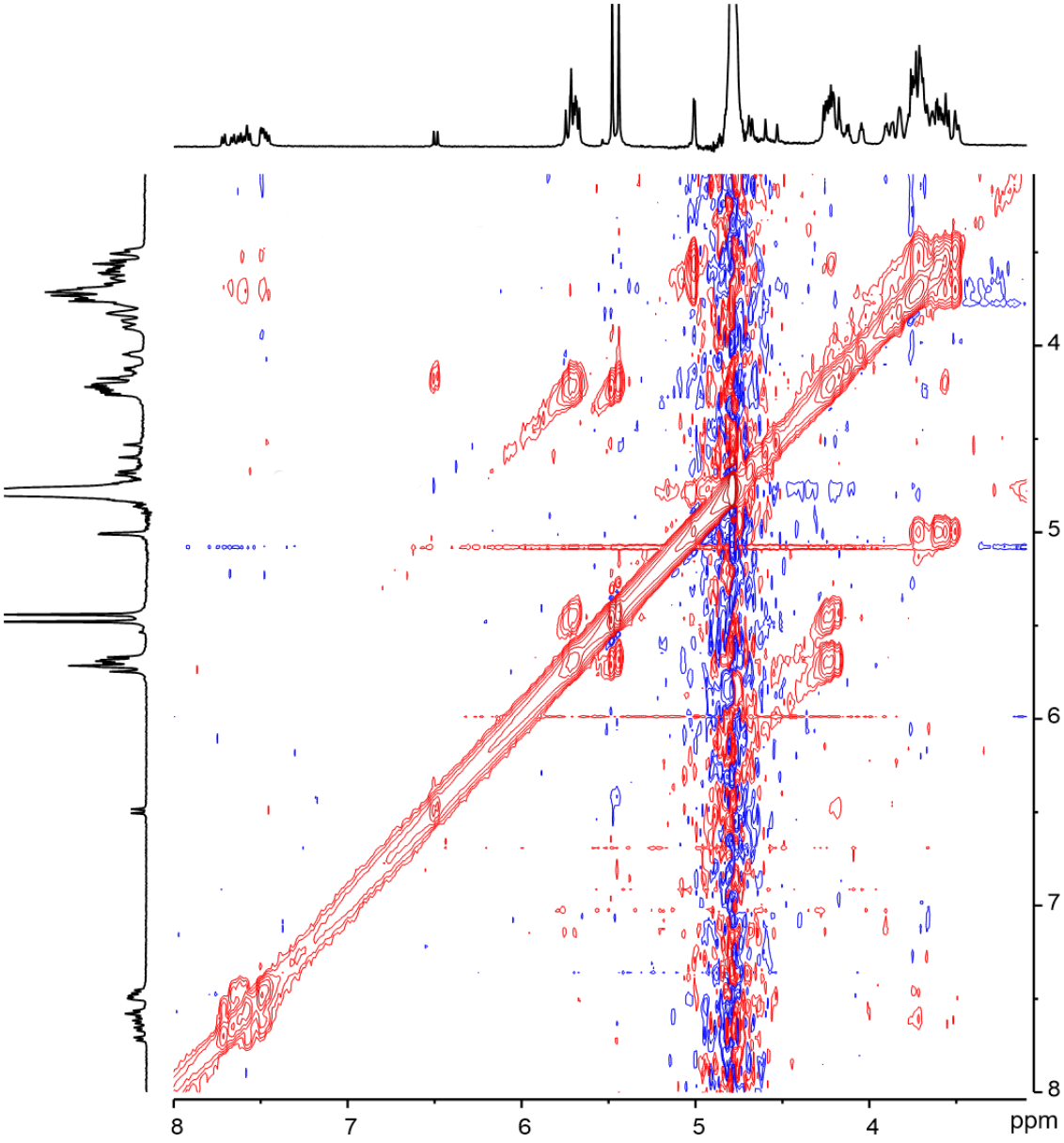

Figure S10. 2D NOESY (500 MHz, D_2O, 298 K, mixing time = 700 ms) spectrum of the formate salt of the tetra-protonated form of **4C-2**.

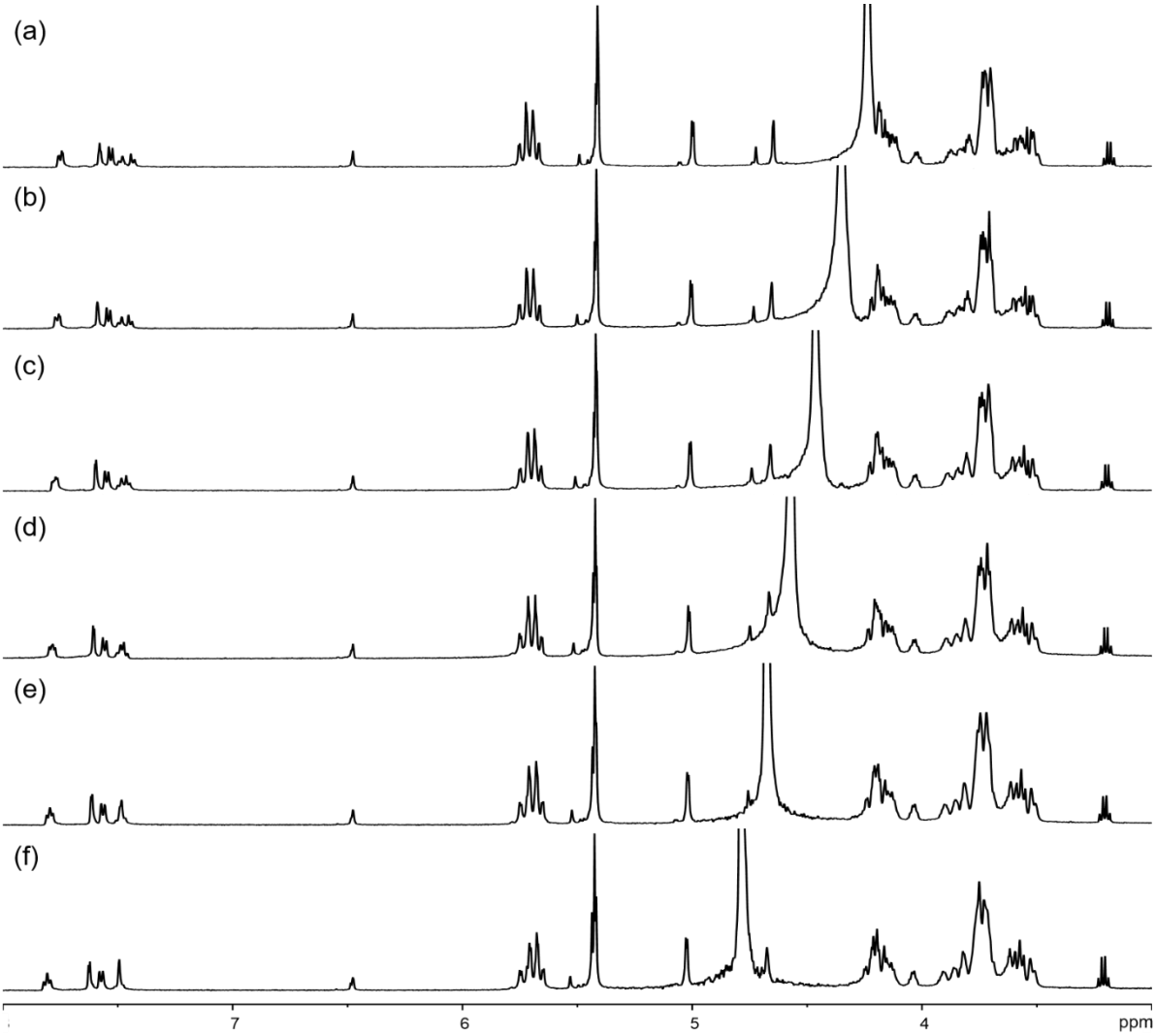

Figure S11. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-1** at (a) 348 K, (b) 338 K, (c) 328 K, (d) 318 K, (e) 308 K and (f) 298 K.

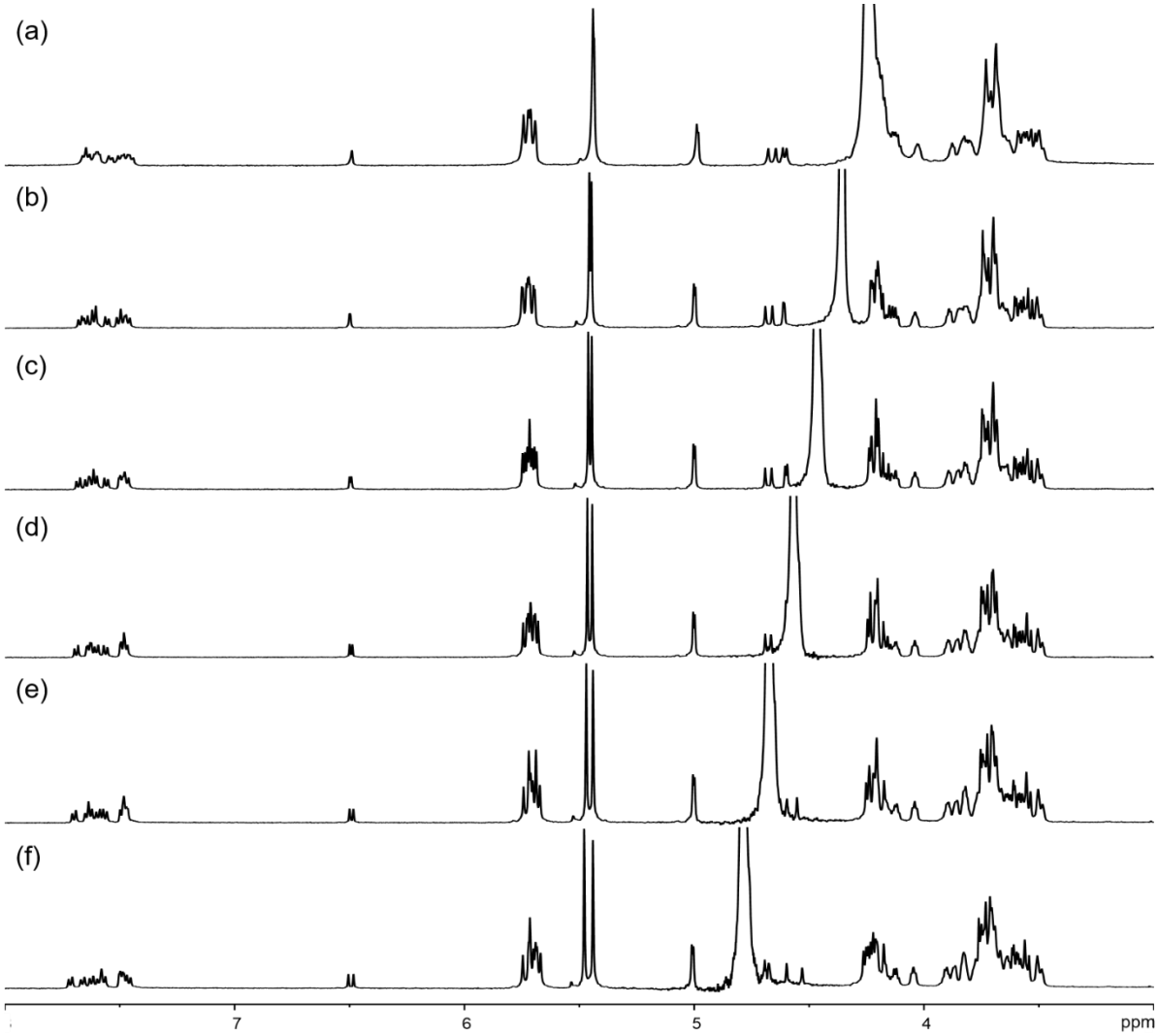

Figure S12. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-2** at (a) 348 K, (b) 338 K, (c) 328 K, (d) 318 K, (e) 308 K and (f) 298 K.

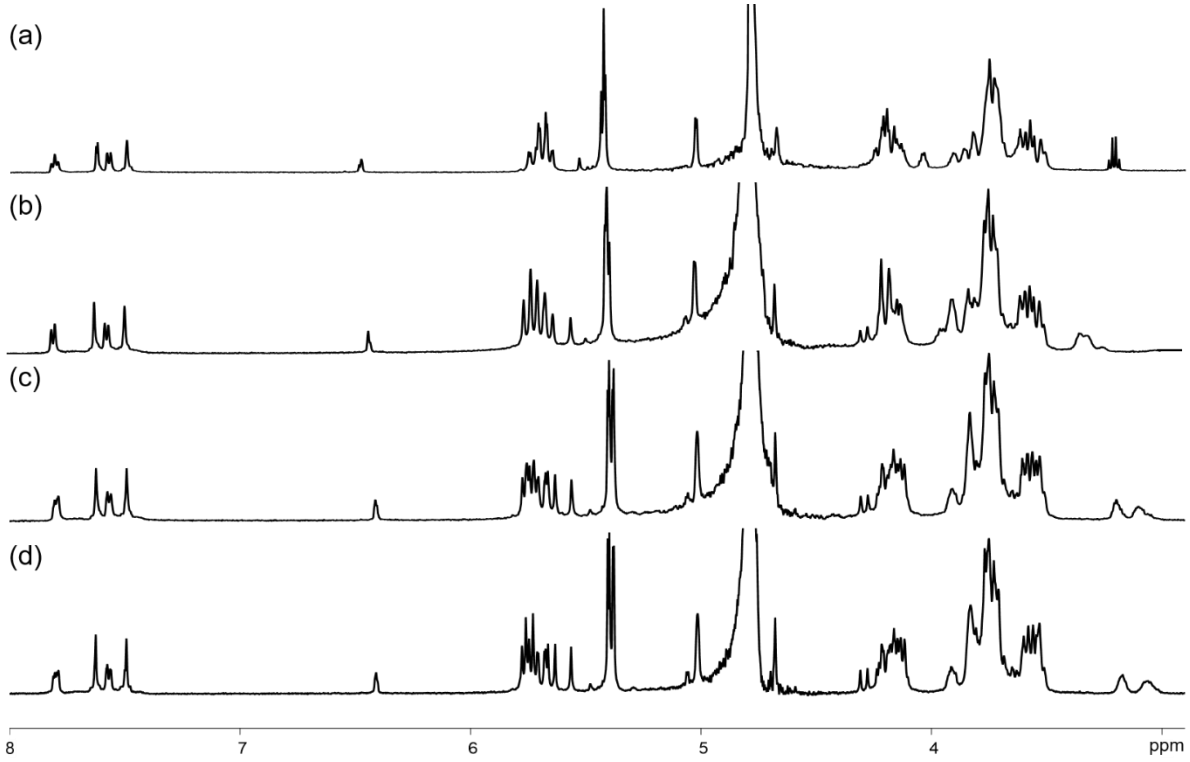

Figure S13. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-1** in the presence of (a) 0, (b) 2, (c) 4, (d) 6 eq. of NaOH.

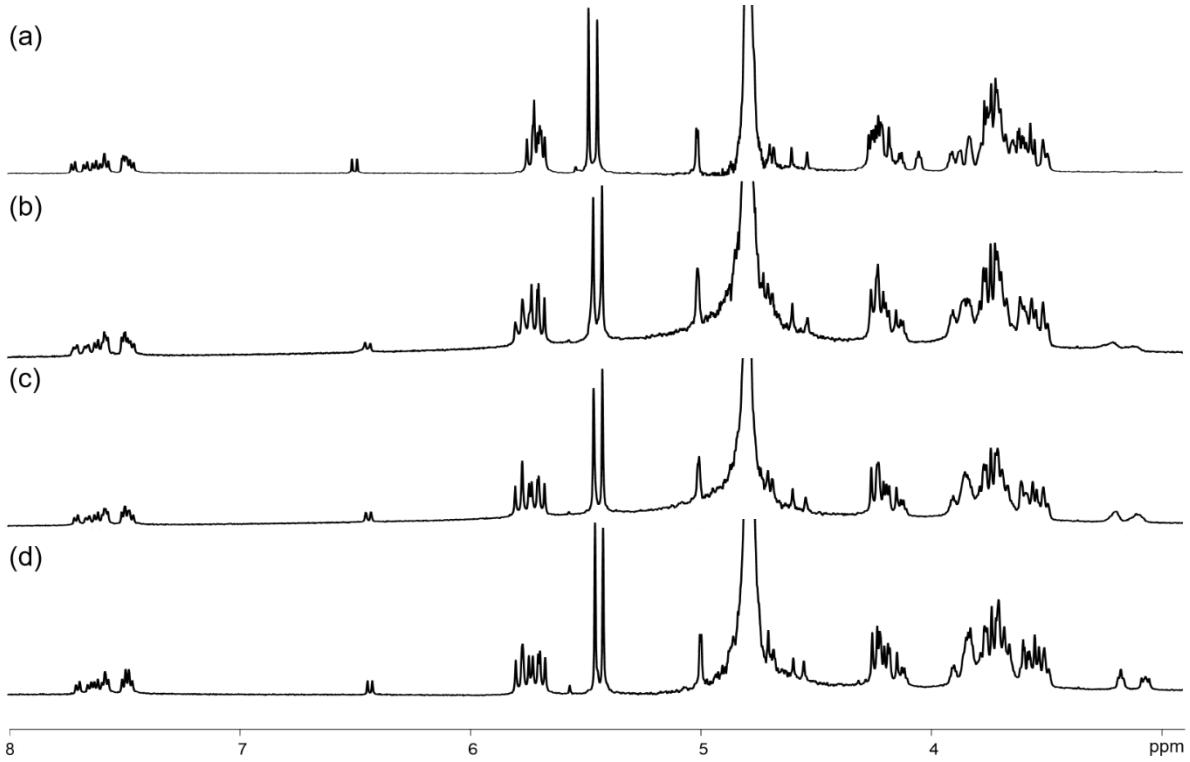

Figure S14. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-2** in the presence of (a) 0, (b) 2, (c) 4, (d) 6 eq. of NaOH.

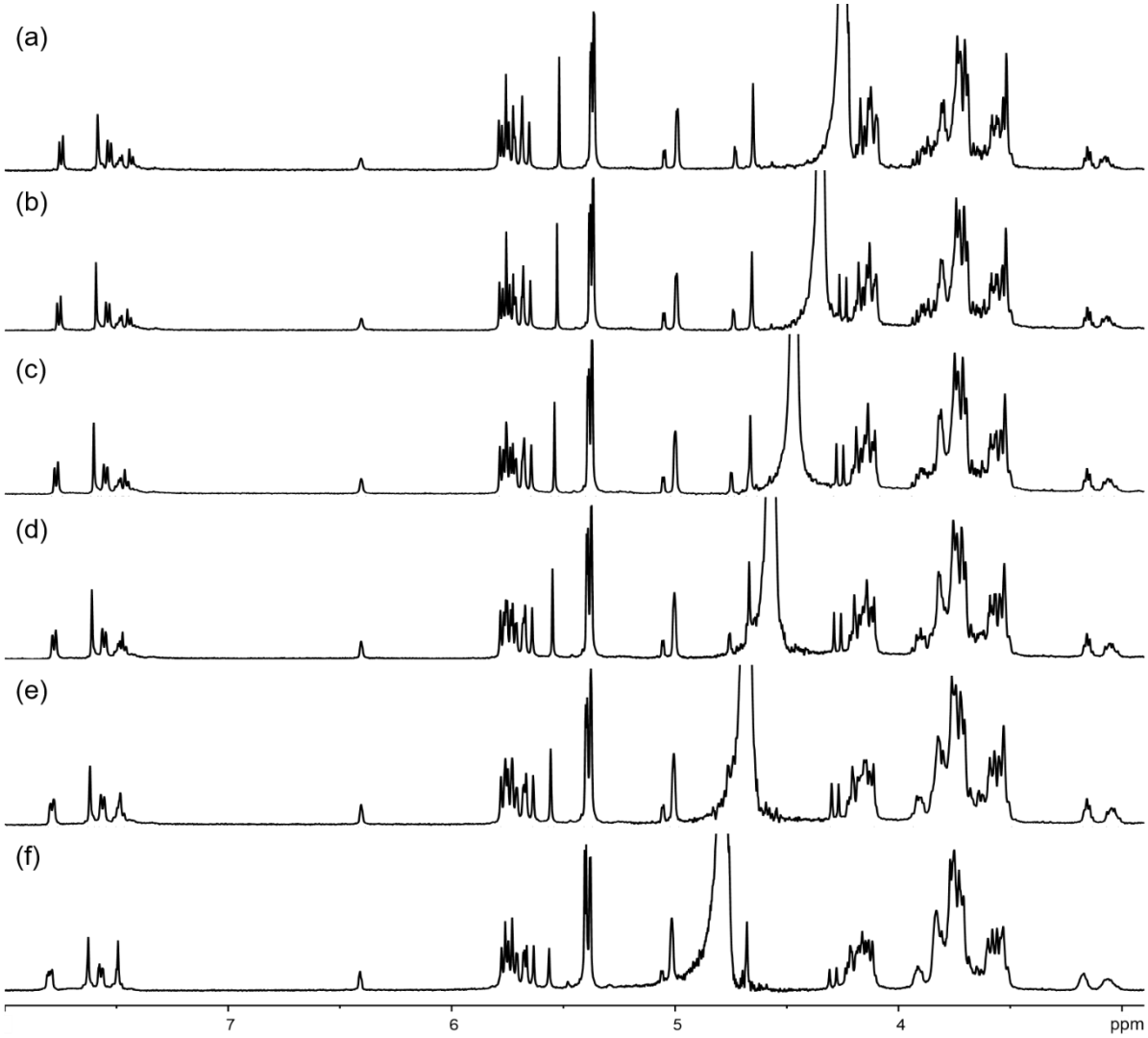

Figure S15. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-1** in the presence of 6 eq. of NaOH at (a) 348 K, (b) 338 K, (c) 328 K, (d) 318 K, (e) 308 K and (f) 298 K.

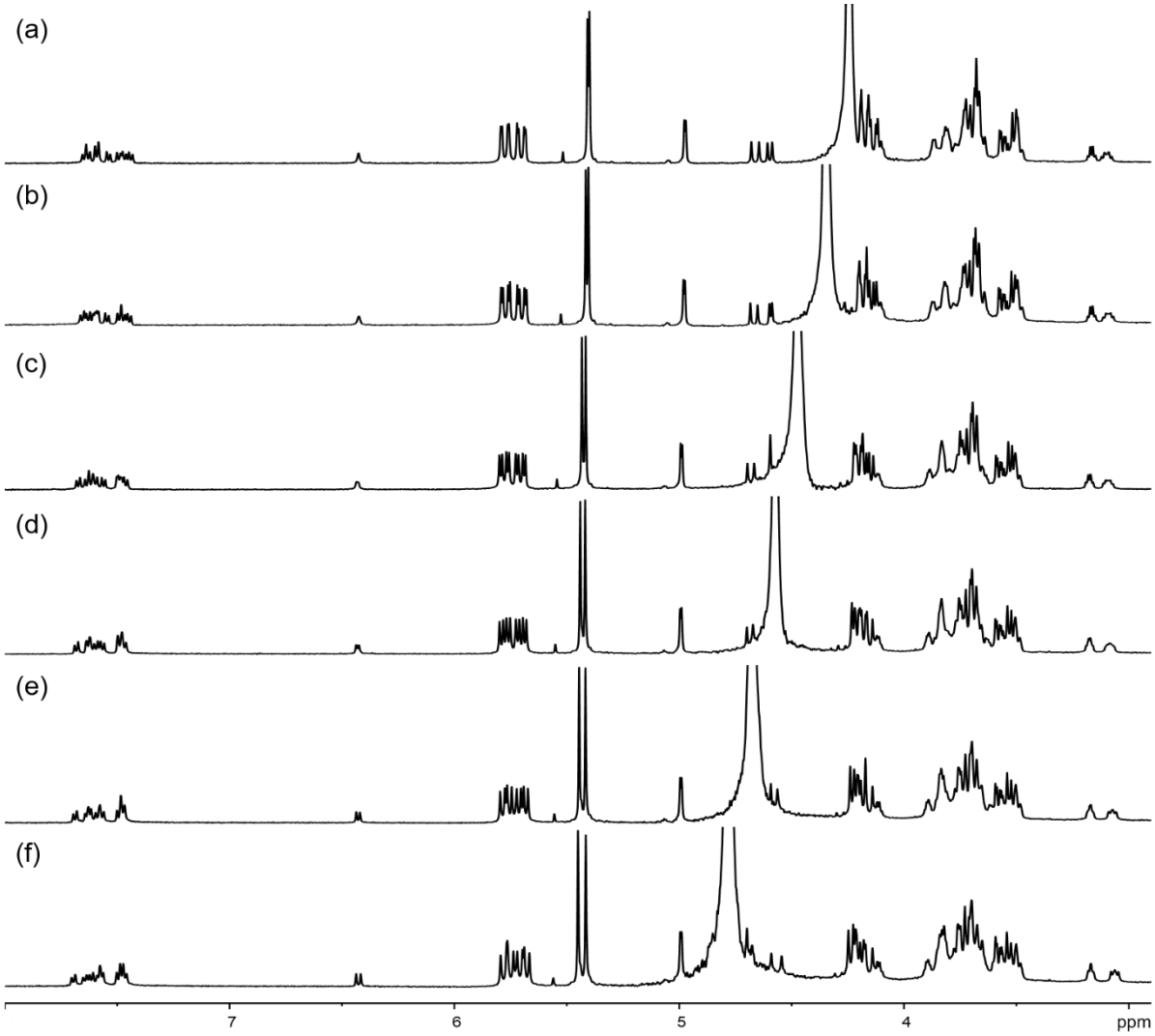

Figure S16. ^{1}H NMR (500 MHz, D_2O) spectra of the formate salt of the tetra-protonated form of **4C-2** in the presence of 6 eq. of NaOH at (a) 348 K, (b) 338 K, (c) 328 K, (d) 318 K, (e) 308 K and (f) 298 K.

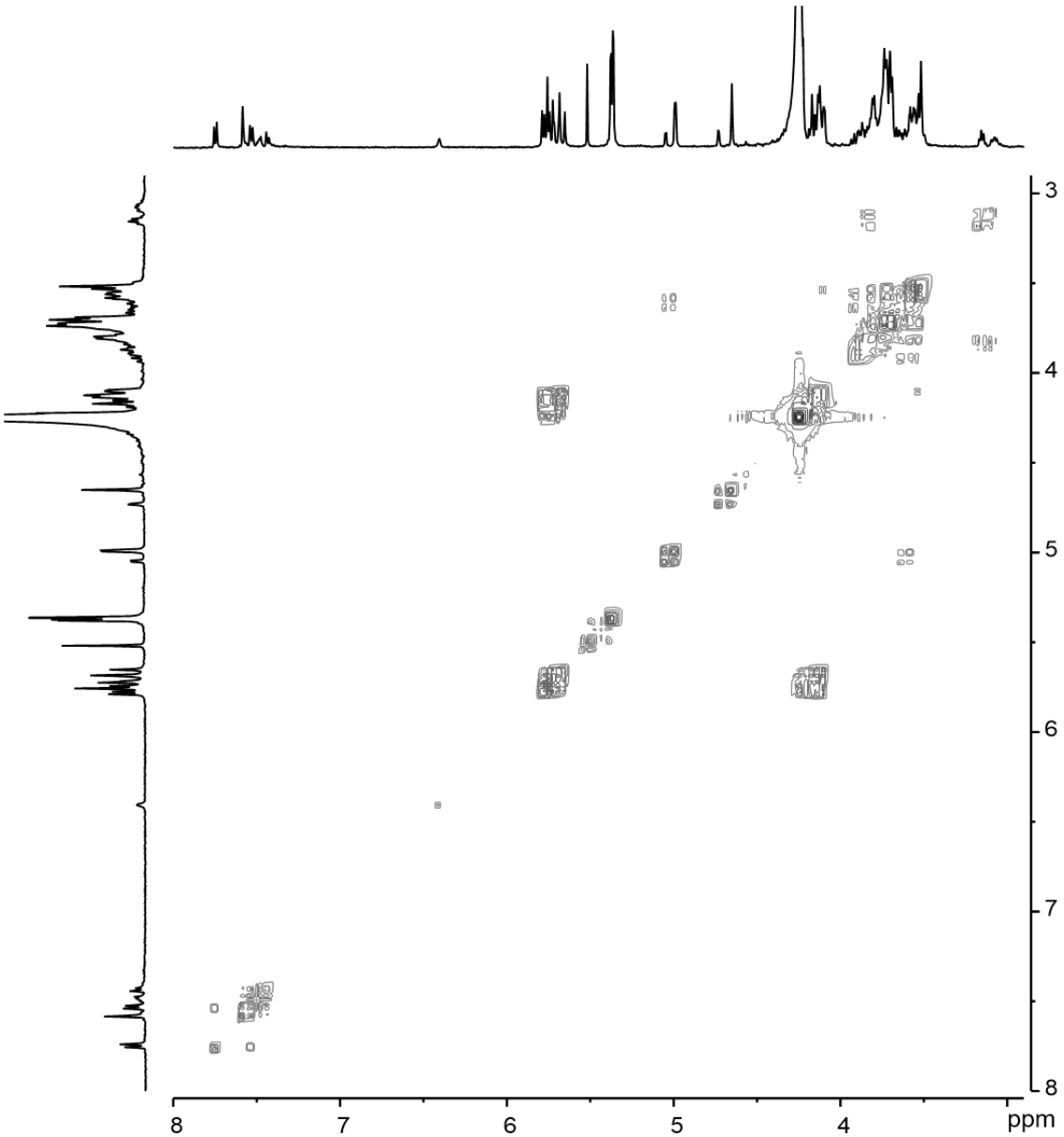

Figure S17. 2D COSY (500 MHz, D_2O, 348 K) spectrum of the formate salt of the tetra-protonated form of **4C-1** in the presence of 6 eq. of NaOH

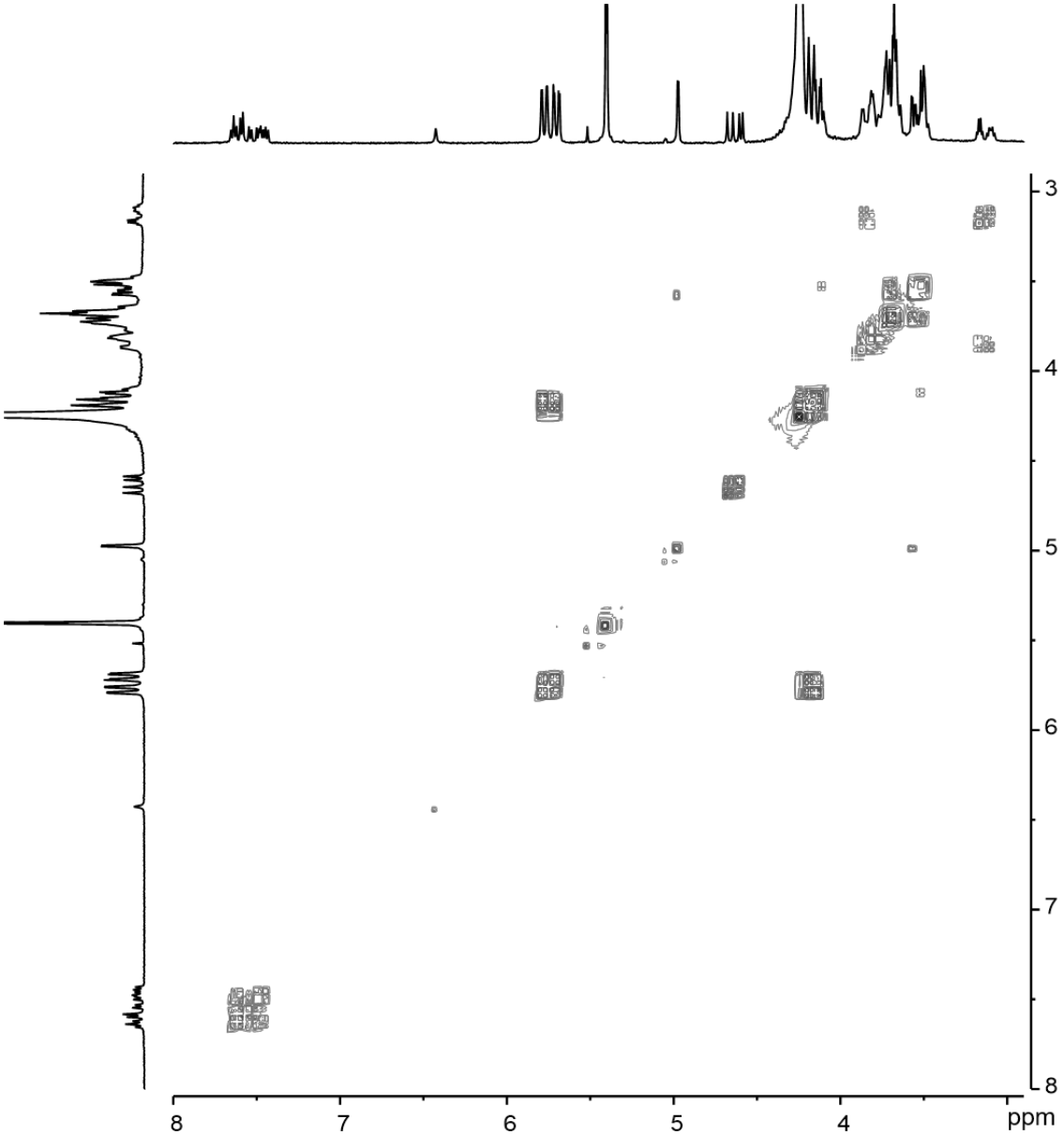

Figure S18. 2D COSY (500 MHz, D_2O, 348 K) spectrum of the formate salt of the tetra-protonated form of **4C-2** in the presence of 6 eq. of NaOH

4. ESI-MS

Mass spectrometry was performed on a Thermo Scientific LTQ FLEET mass spectrometer or a Finnigan LCQ mass spectrometer. HR-ESI-MS were carried out on a Bruker ESI Quadrupole TOF mass spectrometer. MS^2 and MS^3 experiments were carried out on a Thermo Scientific LTQ FLEET mass spectrometer. Isotopic patterns were simulated using IsoPro, version 3.1.

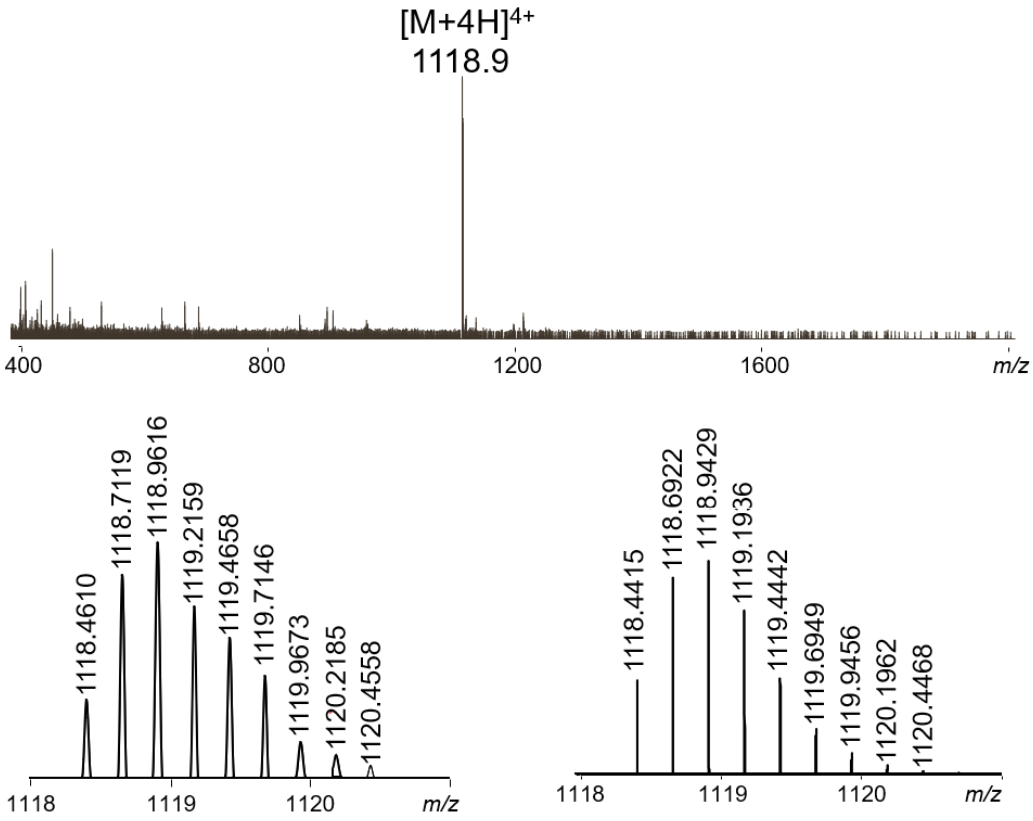

Figure S19. (a) ESI-MS spectrum (+ve) and (b) HRMS of the peak at *m/z* = 1118.9 (left: experimental; right: simulation) of **4C-1**.

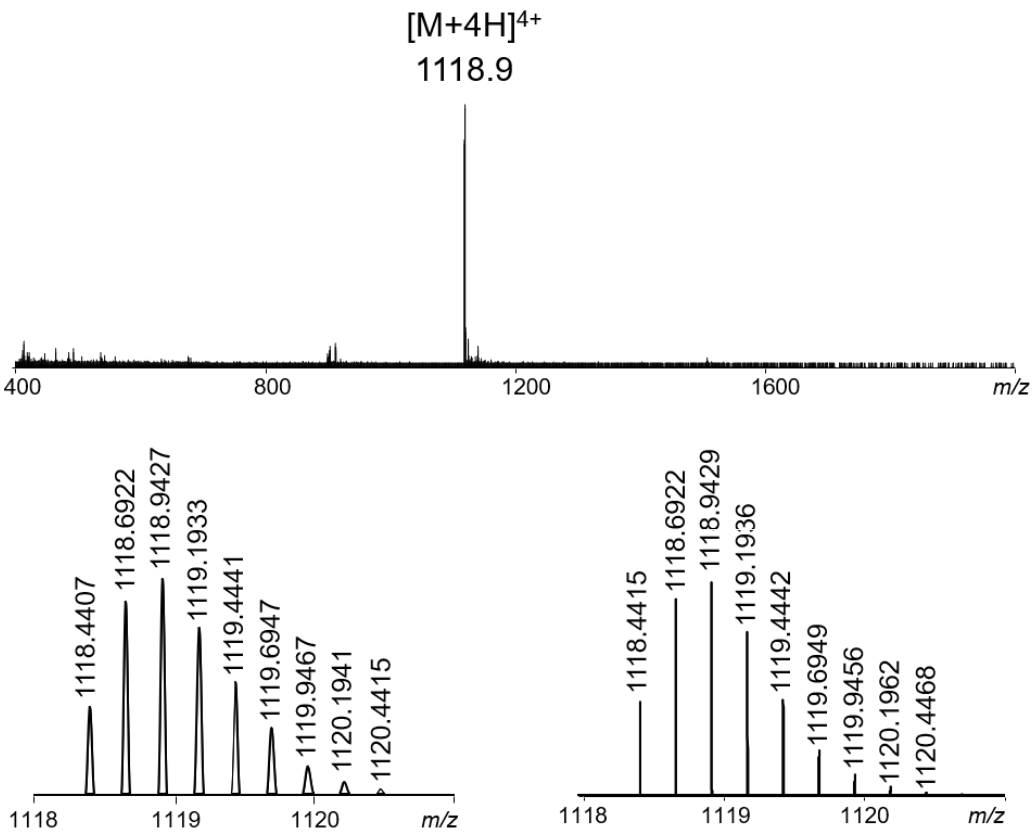

Figure S20. (a) ESI-MS spectrum (+ve) and (b) HRMS of the peak at *m/z* = 1118.9 (left: experimental; right: simulation) of **4C-2**.

5. References

1. A. W. H. Ng, C.-C. Yee, H. Y. Au-Yeung, *Angew. Chem. Int. Ed.,* **2019**, *58*, 17375-17382.

2. D. Bardelang, K. A. Udachin, D. M. Leek, J. A. Ripmeester, *CrystEngComm*, **2007**, *9*, 973-975.