# Host-guest interaction of nitroxide radicals with water-soluble pillar[6]arene

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## Electronic Supplementary Information (21 pages)

*1. Materials and methods*

All reagents were commercially available and used as supplied without further purification. Compounds **WP5** [1,2,3]and **WP6** [4,5,6,7] were synthesized according to the previously described procedures. NMR spectra were recorded with a Bruker Avance DMX 400 spectrophotometer with the deuterated solvent as the lock and the residual solvent or TMS as the internal reference. EPR spectra were recorded with a Bruker X-band EPR spectrometer. The following acquisition parameters were used under aerobic conditions: microwave power, 10 mW; modulation frequency, 100 kHz; modulation amplitude, 1.0 G. The following acquisition parameters were used under anaerobic conditions: microwave power, 2.0 mW; modulation frequency, 100 kHz; modulation amplitude, 0.2 G. The following acquisition parameters were used at low temperature conditions: microwave power, 2.0 mW; modulation frequency, 100 kHz; time constant, 40.96 ms; conversion time, 20 ms; modulation amplitude, 2.0 G.

*2. EPR parameters and association constants in the **4-AT ⊂ WP6** system*

**Table S1** Hyperfine splitting constant ($\alpha_N$), g-factor and three relaxation parameters (α, β and γ) of free and complexed **4-AT**, and binding constants (*K*) of the complex GH (**4-AT**:**WP6**, 1:1) and the complex $GH_2$ (**4-AT**:**WP6**, 1:2) at room temperature. The hfsc and relaxation parameters are given in Gauss units.

| | G | GH | $GH_2$ |
|---|---|---|---|
| g | 2.00647 | 2.00661 | 2.00654 |
| $\alpha_N$ (G) | 16.93 | 16.48 | 16.96 |
| $\alpha_{H(6)}$ (G) | 0.49 | 0.56 | 0.53 |
| $\alpha$ | 0.595 | 0.624 | 0.696 |
| $\beta$ | 0.008 | 0.728 | 0.080 |
| $\gamma$ | 0.020 | 0.759 | 0.046 |
| lgK | - | 3.486 | 6.876 |

*3. pH titration experiments of **4-AT** and **4-AT ⊂ WP6***

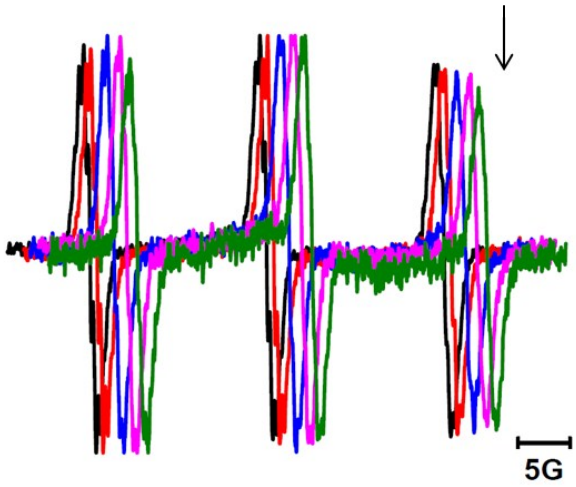



**Fig. S1** EPR spectra of **4-AT** (black) and **4-AT ⊂ WP6**(25eq) at pH of 10.5 (red); 9.5 (blue); 9.0 (magenta);8.0 (green).

*4. $^1$H NMR spectra of **4-AT** and **4-HT** in the presence of **WP5** or **WP6***

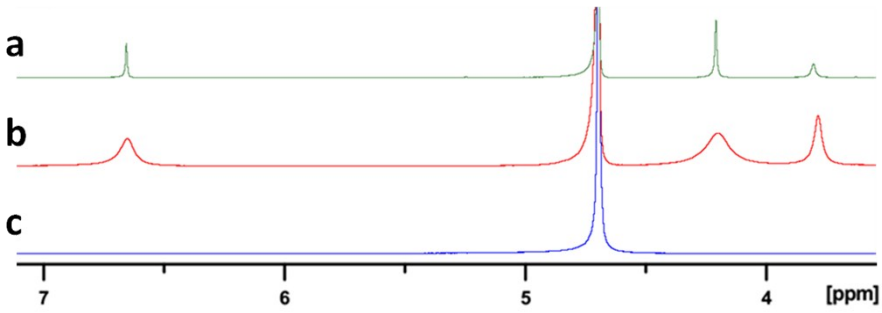



***Fig. S2*** $^{1}$H NMR spectra of (a) **WP6** (2.5 mM), (b) **4-AT** (1.0 mM) with **WP6** (2.5 mM) and (c) **4-AT** (1.0 mM) in $D_2O$.

**Table S2** Half widths of NMR peaks from **WP6** and **4-AT ⊂ WP6**

| | Peak at 6.66ppm | Peak at 4.21ppm | Peak at 3.81ppm |
|---|---|---|---|
| **WP6** | 3.09 Hz | 3.61 Hz | 7.68 Hz |
| **4-AT ⊂ WP6** | 27.51 Hz | 46.96 Hz | 15 Hz |

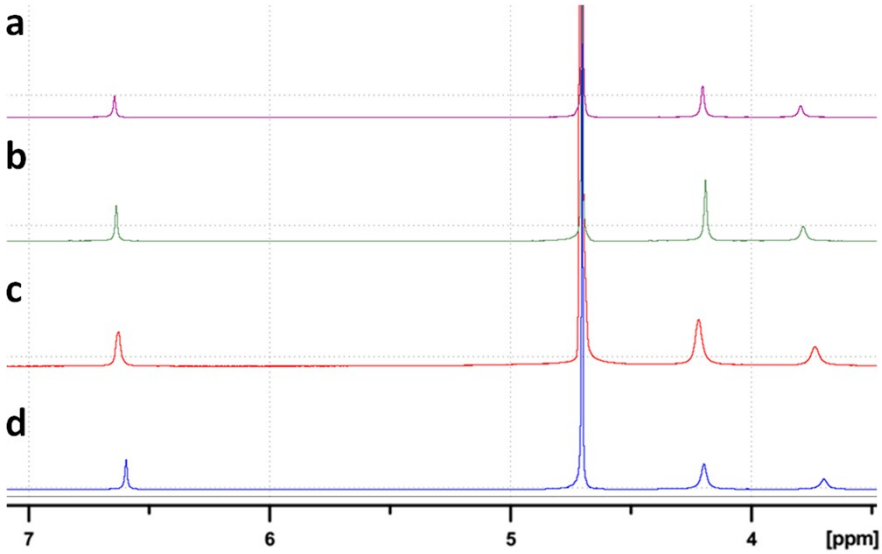
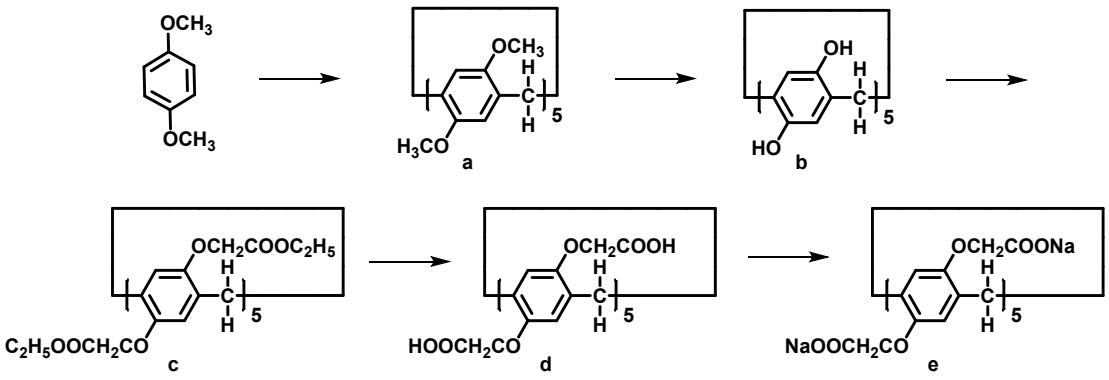



***Fig. S3*** $^{1}$H NMR（400 MHz, $D_2O$）spectra of (a) **WP5** (2.5 mM), (b) **4-AT** (1.0 mM) with **WP5** (2.5 mM), (c) **WP6** (2.5 mM) and (d) **4-HT** (1.0 mM) with **WP6** (2.5 mM).

*5. Syntheses of compounds **WP5, WP6** and **4-ATH***

*5.1. Synthesis of compound **WP5***




Synthesis of **a**: To a solution of 1,4-dimethoxybenzene (1.38 g, 10.0 mmol) and paraformaldehyde (0.300 g, 10.0 mmol) in 1,2-dichloroethane (95.0 ml) trifluoroacetic acid (5.00 ml) was added. Reaction was refluxed for 2 h. After cooling, the reaction mixture was poured into methanol. The resulting precipitate was collected. For a chromatography-free procedure the crude product was dissolved in $CH_2Cl_2$ and then methanol was added (up to 1:1 volume ratio) to yield a crystalline **a**. On the other hand, the crude product was subjected to chromatography (PE:DCM 100:0 up to 1:3) to give **a** (0.820 g, 55%). The proton NMR spectrum of **a** is shown in Fig. S4. $^{1}$H NMR ($CDCl_3$, 400 MHz, 25 °C, TMS): $\delta$ 6.78 (s, 10H, aromatic protons), 3.77 (s, 10H, methylene protons), 3.66 (s, 30H, methyl protons). The $^{13}$C NMR spectrum of **a** is shown in Fig. S5. $^{13}$C NMR ($CDCl_3$, 100 MHz, 25 °C, TMS): $\delta$ 150.8; 128.3, 114.0 (aromatic carbon), 55.7 (carbon on the methylene bridge), 29.6 (methyl-carbon).

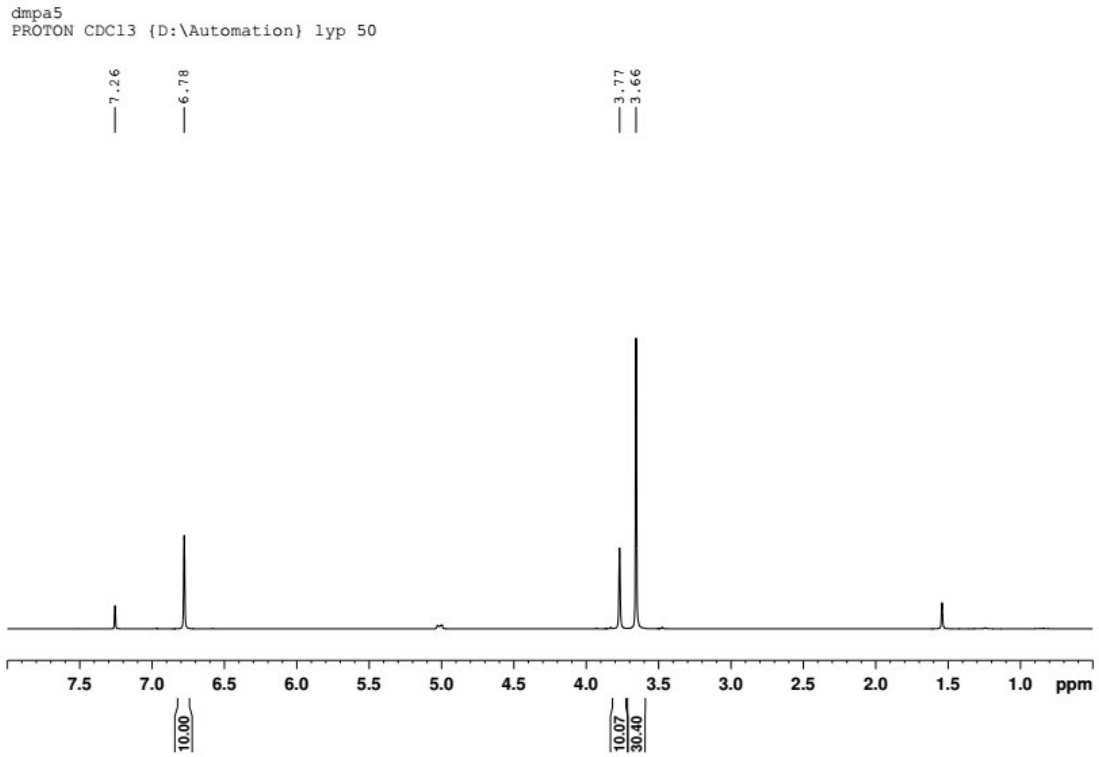



***Fig. S4*** $^{1}$H NMR spectrum (400 MHz, $CDCl_3$, 293K) of **a**

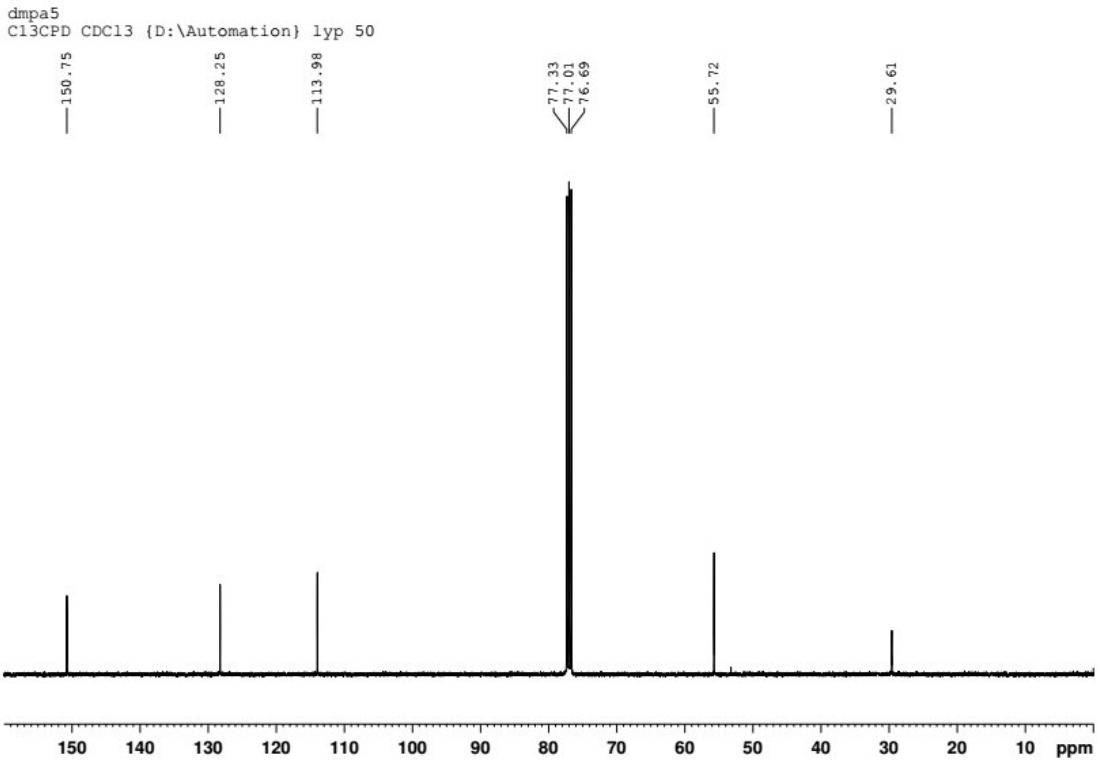



***Fig. S5*** $^{13}$C NMR spectrum (100 MHz, $CDCl_3$, 293K) of **a**

Synthesis of **b**: To a solution of Compound **a** (1.50 g, 2.00 mmol) in anhydrous $CH_2Cl_2$ (150 ml) boron tribromide $BBr_3$ (5.60 ml, 15.6 g, 62.0 mmol) was then added slowly and the resulting mixture stirred at r.t. for 72 h. Water (100 ml) was then added and the reaction mixture was continuously stirred for 24 h at r.t. The resulting white precipitate was filtered and washed with water to give **b** as a white powder (1.11 g, 91%). The proton NMR spectrum of **b** is shown in Fig. S6. $^1$H NMR (DMSO-$d_6$, 400 MHz, 25 °C, TMS): $\delta$ 7.99 (s, 10H, hydroxyl protons), 6.58 (s, 10H, aromatic protons), 3.66 (s, 10H, methylene protons). The $^{13}$C NMR spectrum of **b** is shown in Fig. S7. $^{13}$C NMR (DMSO-$d_6$, 100 MHz, 25 °C, TMS): $\delta$ 146.1 126.4, 117.3 (C of aromatic), 29.1 (C of methylene bridge).

Note: Pillar[5]arene (**b**) was synthesized as a white powder whose color slowly turns to be brown due to oxidation after exposure to air.

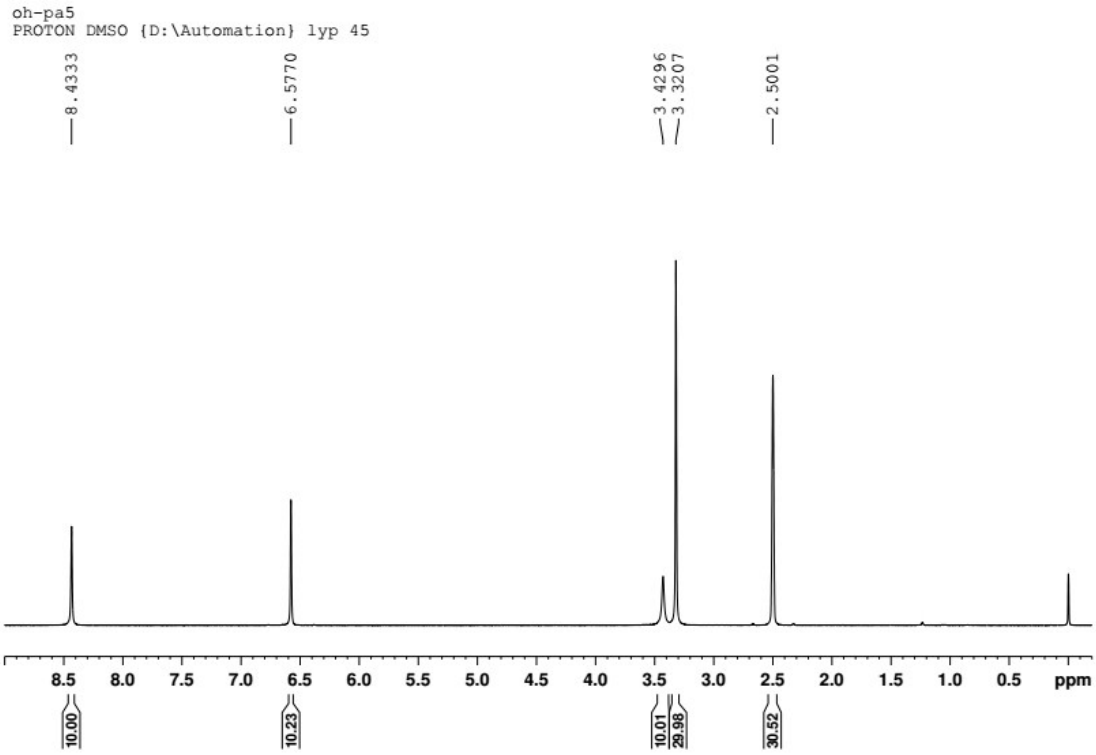



***Fig. S6*** $^1$H NMR spectra (400 MHz, DMSO-$d_6$, 293K) of **b**

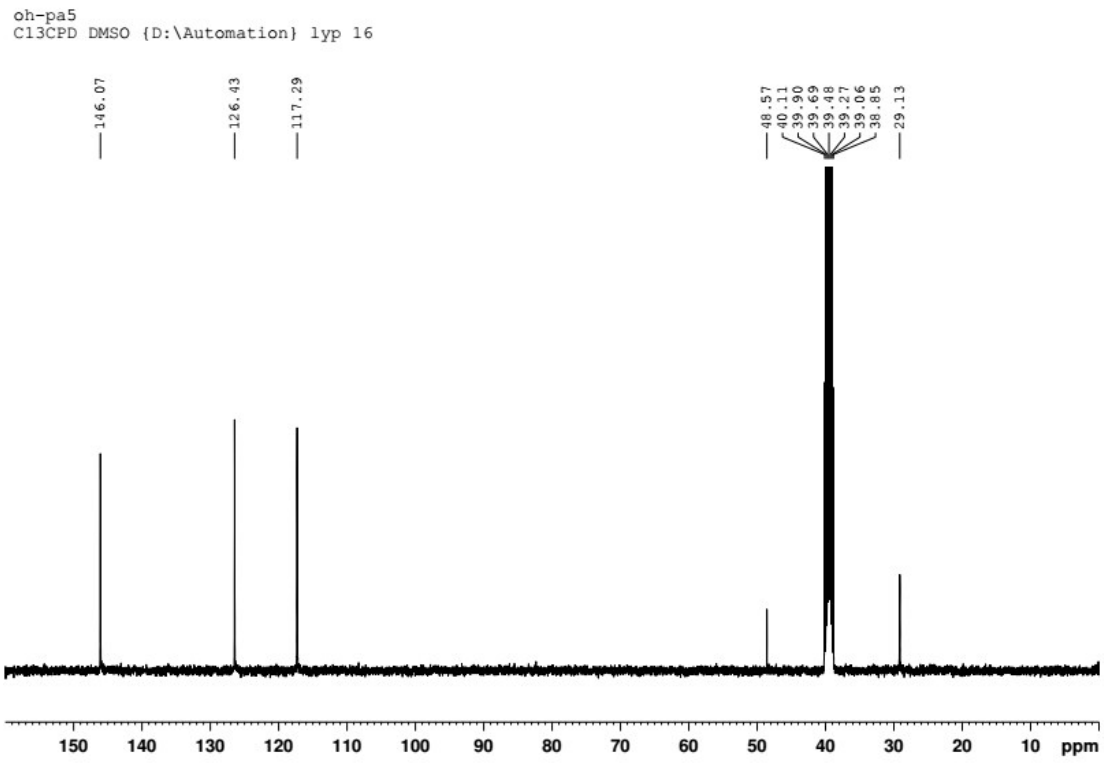



***Fig. S7*** $^{13}$C NMR spectra (100 MHz, DMSO-$d_6$, 293K) of **b**

Synthesis of **c**: To a solution of **b** (1.50 g, 2.40 mmol) in $CH_3CN$ (70.0 ml) $K_2CO_3$ (3.60 g, 26.0 mmol) was added and the mixture stirred for 45 min. at r.t. KI (35.0 mg) and excess of ethyl bromoacetate (5.00 ml, 7.50 g, 45.0 mmol) were added to the reaction mixture which was then refluxed for 20 h under nitrogen. After cooling the reaction mixture was filtered and washed with DCM (20.0 ml). The organic layers were combined and the solvent was removed under reduced pressure. The resulting oily product was subjected to the column chromatography (silica gel 200-300 size; DCM:EA, 100:0 up to 9:1). The second fraction (Rf: 0.60) was collected and recrystalized from acetone to give **c** as a white crystalline product (2.90 g, 80.0%). The proton NMR spectrum of **c** is shown in Fig. S8. $^1$H NMR ($CDCl_3$, 400 MHz, 25 °C, TMS): $\delta$ 7.05 (s, 10H, aromatic protons), 4.54 (q, 20H, $J$ = 15.7, 23.4 Hz O-methylene protons), 4.08 (m, 20H, $J$ = 7.2 Hz, methylene protons), 3.86 (s, 10H, methylene bridge protons), 0.97 (t, 30H, methyl protons). The $^{13}$C NMR spectrum of **c** is shown in Fig. S9. $^{13}$C NMR ($CDCl_3$, 100 MHz, 25 °C, TMS): $\delta$ 169.3 (C of carbonyl), 149.0, 128.7, 114.4 (C of aromatic), 65.7 (C of O-methylene), 60.9 (C of O-ethylene) 29.2 (C of methylene bridge), 13.8(C of terminal methyl).

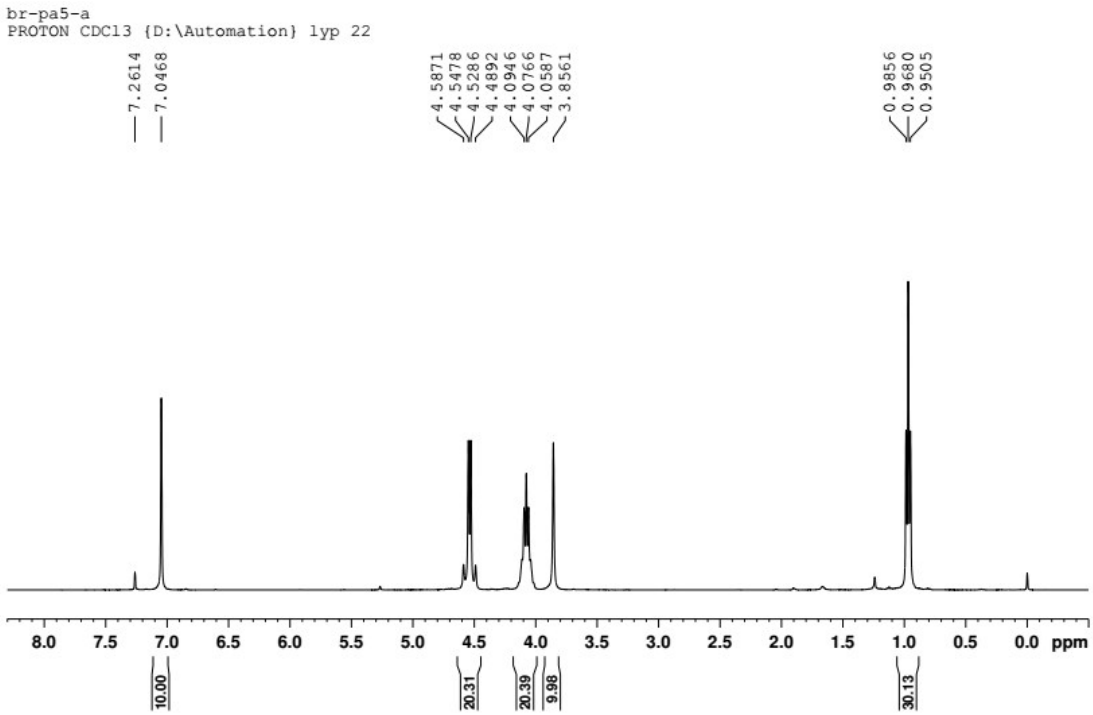



***Fig. S8*** $^{1}$H NMR spectra (400 MHz, $CDCl_3$, 293K) of **c**

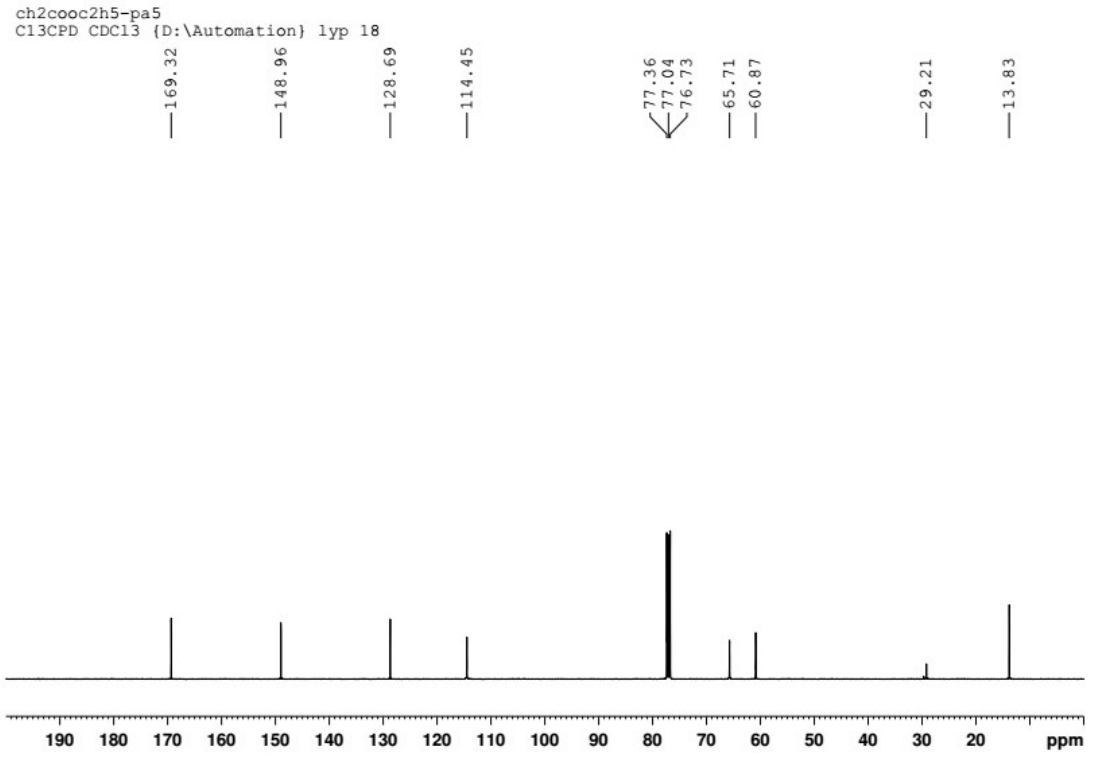



***Fig. S9*** $^{13}$C NMR spectra (100 MHz, $CDCl_3$, 293K) of **c**

Synthesis of **d**: To a solution of **c** (1.47 g, 1.00 mmol) in THF (60.0 mL) aqueous NaOH solution (30.0 mL, 20%) was added. The reaction mixture was refluxed for 15 h. After cooling to rt, the mixture was concentrated under reduced pressure. The residue was diluted into 100 mL of $H_2O$, and then acidified with HCl. The resulting precipitate was collected by filtration, washed with $H_2O$, and dried under vacuum to give **d** (1.17 g, 98.0%). The proton NMR spectrum of **d** is shown in Fig. S10. $^1$H NMR (DMSO-$d_6$, 400 MHz, 25 °C, TMS): $\delta$ 12.94 (s, 10H), 7.10 (s, 10H), 4.69 (d, $J$ = 15.9 Hz, 10H), 4.41 (d, $J$ = 15.8 Hz, 10H), 3.74 (s, 10H). The $^{13}$C NMR spectrum of **d** is shown in Fig. S11. $^{13}$C NMR (DMSO-$d_6$, 100 MHz, 25 °C, TMS): $\delta$ 170.5 (C of carbonyl), 148.4, 128.0, 114.2 (C of aromatic), 65.0 (C of O-methylene).

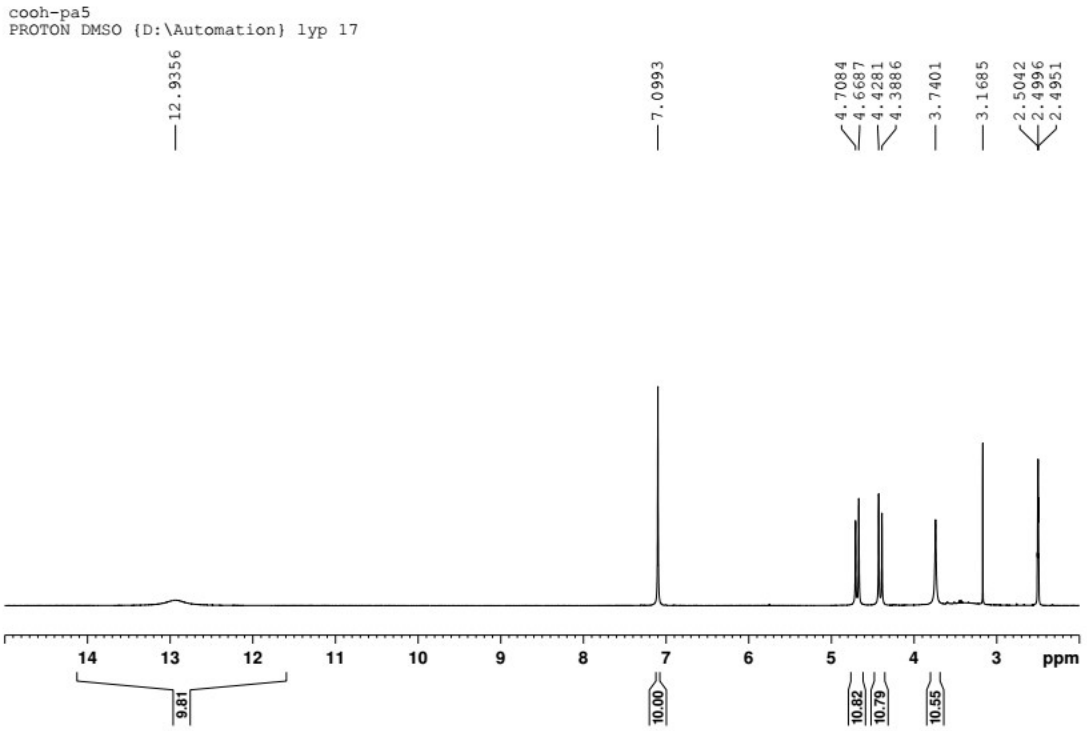



***Fig. S10*** $^1$H NMR spectra (400 MHz, DMSO-$d_6$, 293K) of **d**

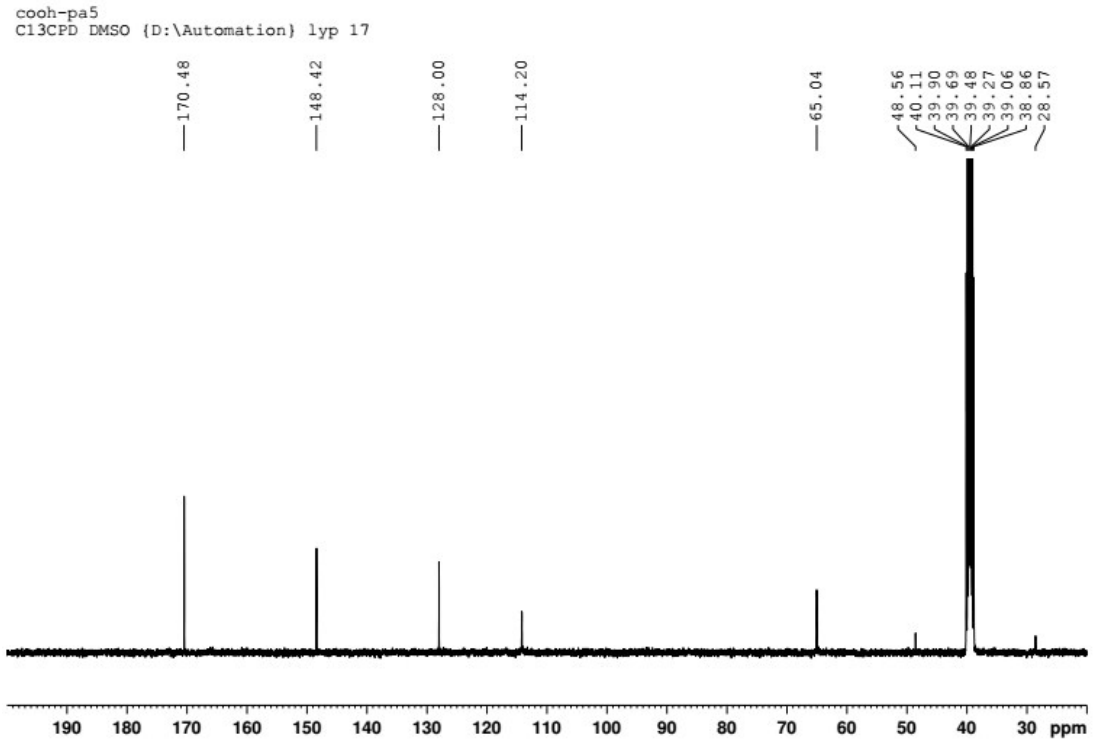



***Fig. S11*** $^{13}$C NMR spectra (100 MHz, DMSO-$d_6$, 293K) of **d**

Synthesis of **e**: To a solution of **d** (0.240 g) dispersed in deionized $H_2O$ (3.00 mL) NaOH solution (80.0 mg NaOH in 1.00 mL of deionized $H_2O$) was added dropwise into the mixture until the reaction mixture was clear. The final product **e** was obtained after drying under vacuum (0.250 g, 95.0%). The proton NMR spectrum of **e** is shown in Fig. S12. $^1$H NMR ($D_2O$, 400 MHz, 25 °C, TMS): $\delta$ 6.66 (s, 10H), 4.25 (s, 10H), 3.76 (s, 10H). The $^{13}$C NMR spectrum of **e** is shown in Fig. S13. $^{13}$C NMR ($D_2O$, 100 MHz, 25 °C, TMS): $\delta$ 177.4 (C of carbonyl), 149.3, 128.7, 114.8(C of aromatic), 67.8(C of O-methylene).

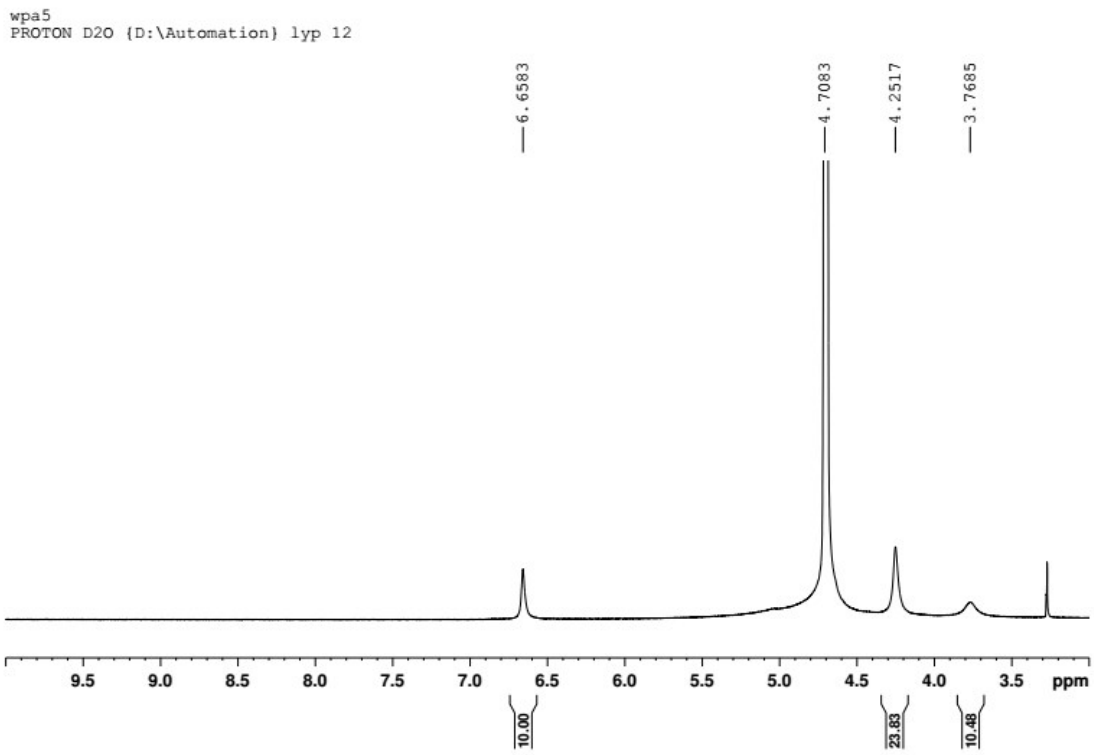



***Fig. S12*** $^{1}$H NMR spectra (400 MHz, $D_2O$, 293K) of **e**

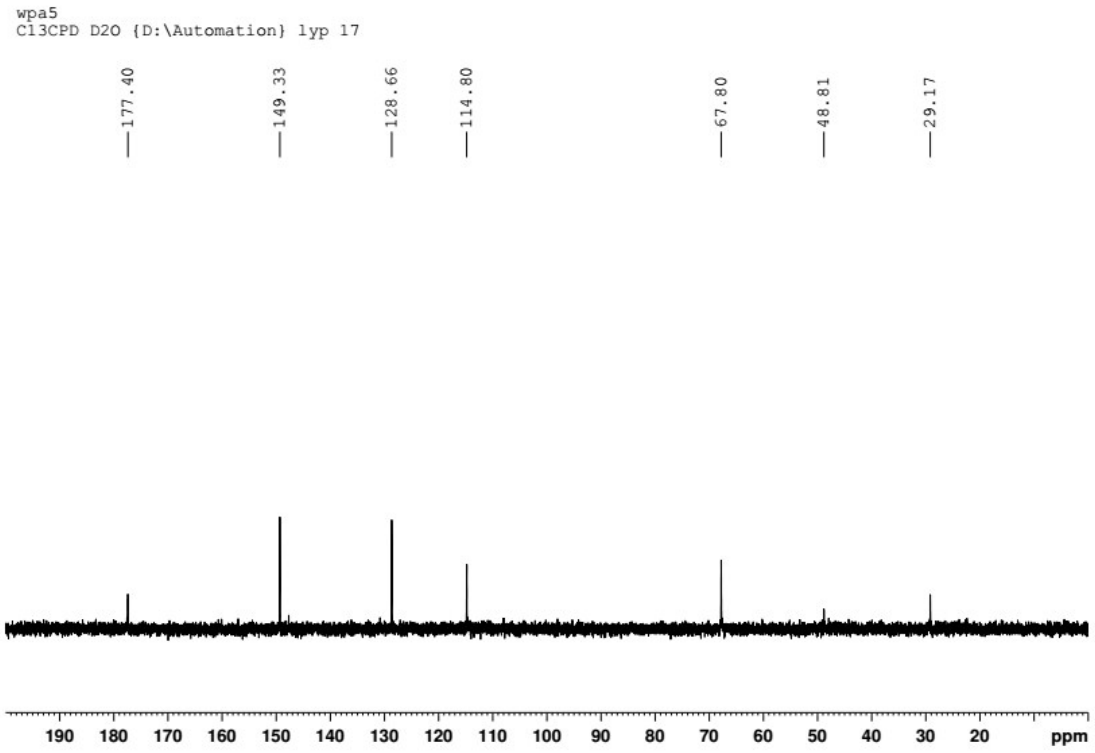
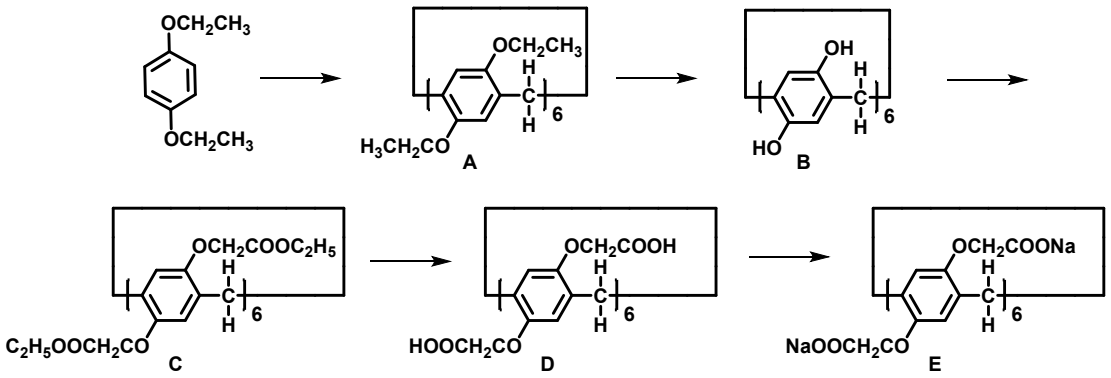



***Fig. S13*** $^{13}$C NMR (100 MHz, $D_2O$, 293K) spectra of **e**

*5.2. Synthesis of compound **WP6***




Synthesis of **A**: To the solution of 1,4-diethoxybenzene (6.00 g, 36.0 mmol) in chloroform (300 ml) was added paraformaldehyde (1.08 g, 36.0 mmol). The suspension was stirred at 25 °C for 20 min. And then, boron trifluoride diethyl etherate (4.50 ml, 36.0 mmol) was added and the resulting reaction mixture was continuously stirred at 25 °C for 20 min. Then, the reaction was quenched by water. The organic phase was separated and washed with saturated aqueous $NaHCO_3$, $H_2O$, and brine. The crude product was purified by column chromatography ($CH_2Cl_2$:petroleum ether = 4 : 1) to get **A** (0.965 g, 15%). The proton NMR spectrum of **A** is shown in Fig. S14. $^1$H NMR ($CDCl_3$, 400 MHz, 25 °C, TMS): $\delta$ 6.69 (s, 12H, aromatic protons), 3.82 (q, 24H, $J$ = 6.9 Hz, methylene protons), 3.79 (s, 12H, methylene protons), 1.28 (t, 36H, $J$ = 6.8 Hz, methyl protons). The $^{13}$C NMR spectrum of **A** is shown in Fig. S15. $^{13}$C NMR ($CDCl_3$, 100 MHz, 25 °C, TMS): $\delta$ 150.4; 127.9, 115.3 (C of aromatic), 64.0 (C of methylene bridge), 30.9 (C of methylene groups), 15.2. (C of methyl group).

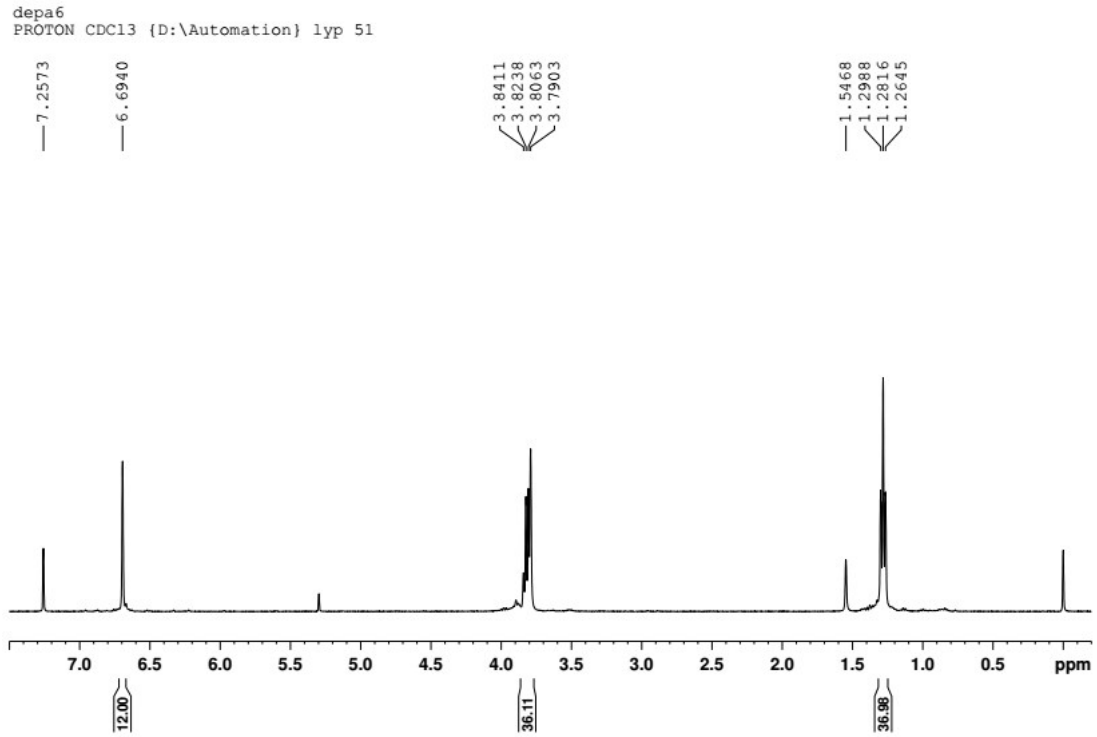



***Fig. S14*** $^{1}$H NMR spectra (400 MHz, $CDCl_3$, 293K) of **A**

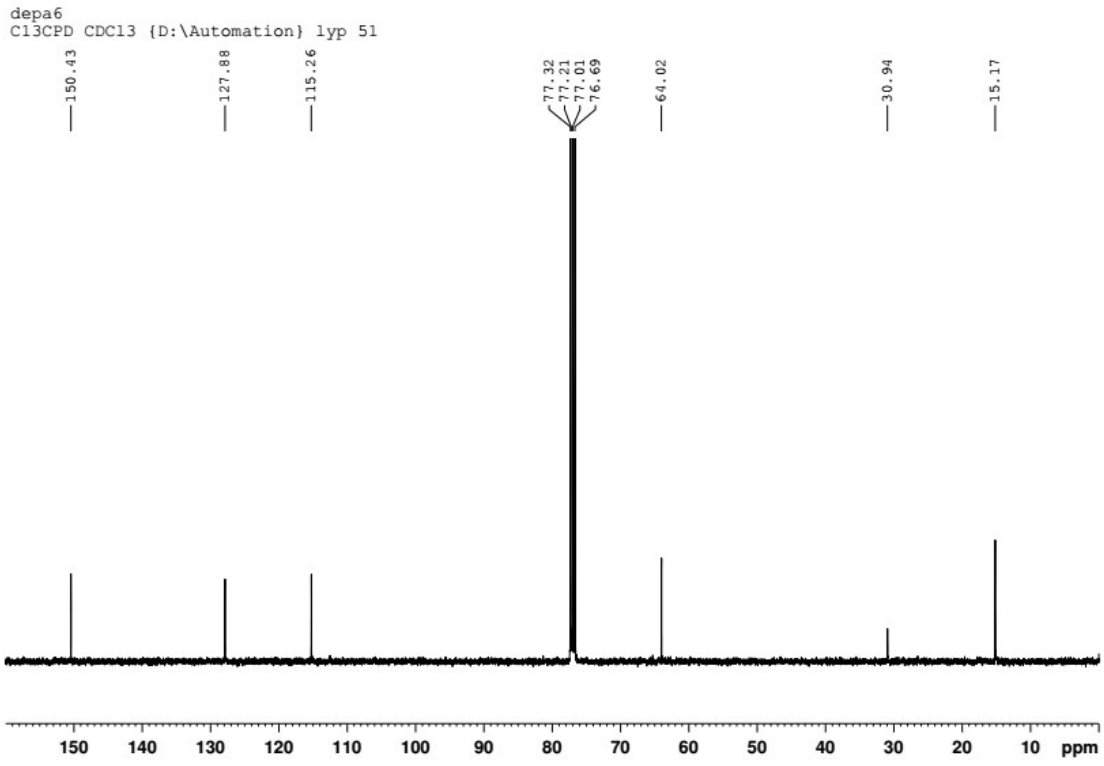



***Fig. S15*** $^{13}$C NMR spectra (100 MHz, $CDCl_3$, 293K) of **A**

Synthesis of **B**: To a solution of compound **A** (1.04 g, 0.740 mmol) in anhydrous $CH_2Cl_2$ (50.0 mL) was added boron tribromide $BBr_3$ (3.71 g, 14.8 mmol). The mixture was stirred at 25 °C for 24 h. Then water was added into the mixture. The resulting precipitate was collected and washed with $H_2O$ to give **B** (0.577 g, 81.0 %). The proton NMR spectrum of **B** is shown in Fig. S16. $^1$H NMR (DMSO-$d_6$, 400 MHz, 25 °C, TMS): $\delta$ 8.33 (s, 12H, hydroxyl protons), 6.40 (s, 12H, aromatic protons), 3.50 (s, 12H, methylene protons). The $^{13}$C NMR spectrum of **B** is shown in Fig. S17. $^{13}$C NMR (DMSO-$d_6$, 100 MHz, 25 °C, TMS): $\delta$ 146.8, 125.5, 117.0 (C of aromatic), 29.2 (C of methylene bridge).

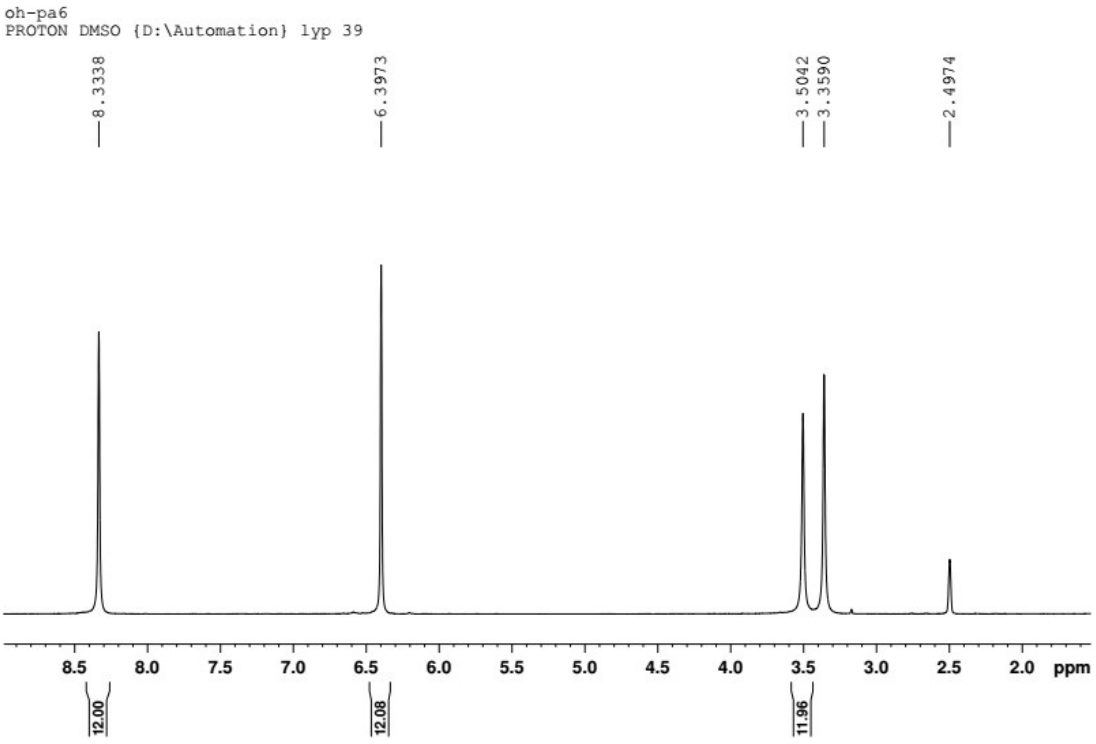



***Fig. S16*** $^1$H NMR spectra (400 MHz, DMSO-$d_6$, 293K) of **B**

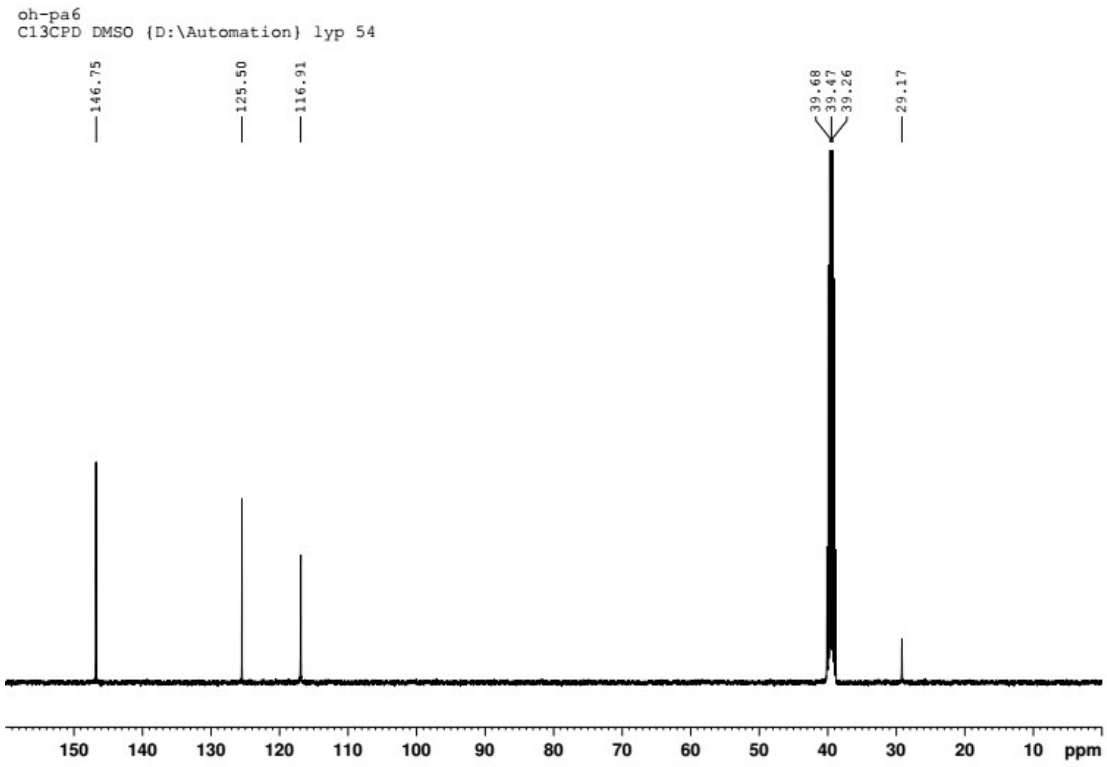



***Fig. S17*** $^{13}$C NMR spectra (100 MHz, DMSO-$d_6$, 293K) of **B**

Synthesis of **C**: To a solution of **B** (0.730 g, 1.00 mmol) in $CH_3CN$ (50.0 ml) $K_2CO_3$ (6.62 g, 48.0 mmol) was added and the mixture was stirred for 45 min at r.t. KI (35.0 mg) and excess of ethyl bromoacetate (2.59 g, 24.0 mmol) were added to the reaction mixture which was then refluxed for 20 h under nitrogen. The cooled reaction mixture was filtered and washed with DCM. The filtrate was evaporated under vacuum, and the residue was purified by crystallization in a mixture of methanol and DCM. The product of **C** was collected by filtration, washed with methanol, and dried under vacuum (0.320 g, 20.0%). The proton NMR spectrum of **C** is shown in Fig. S18. $^1$H NMR ($CDCl_3$, 400 MHz, 25 °C, TMS): $\delta$ 6.89 (s, 12H, aromatic protons), 4.49 (s, 24H, methylene protons), 4.22 (m, 24H, $J$ = 3.7 Hz, O-methylene protons), 3.89 (s, 12H, methylene bridge protons), 1.24 (t, 36H, $J$ = 7.0 Hz, methyl protons). The $^{13}$C NMR spectrum of **C** is shown in Fig. S19. $^{13}$C NMR ($CDCl_3$, 100 MHz, 25 °C, TMS): $\delta$ 169.4 (C of carbonyl), 150.0, 127.9, 115.2 (C of aromatic), 65.9 (C of O-methylene), 60.8 (C of O-ethylene) 29.2 (C of methylene bridge), 14.1 (C of terminal methyl).

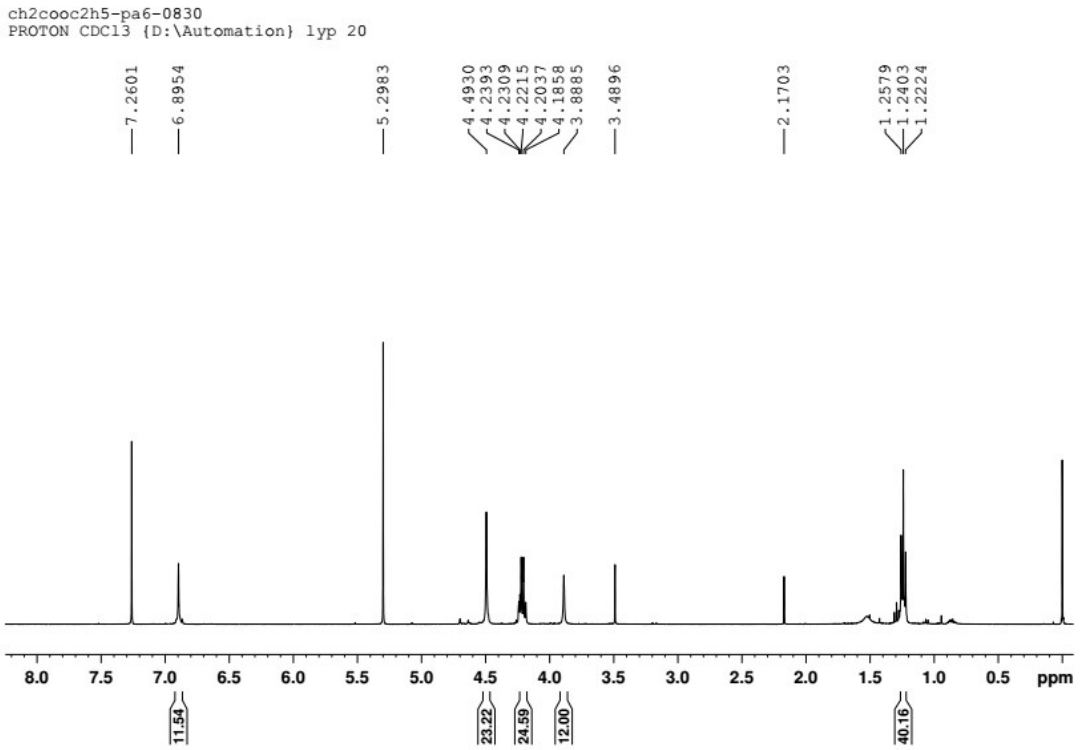



***Fig. S18*** $^{1}$H NMR spectra (400 MHz, $CDCl_3$, 293K) of **C**

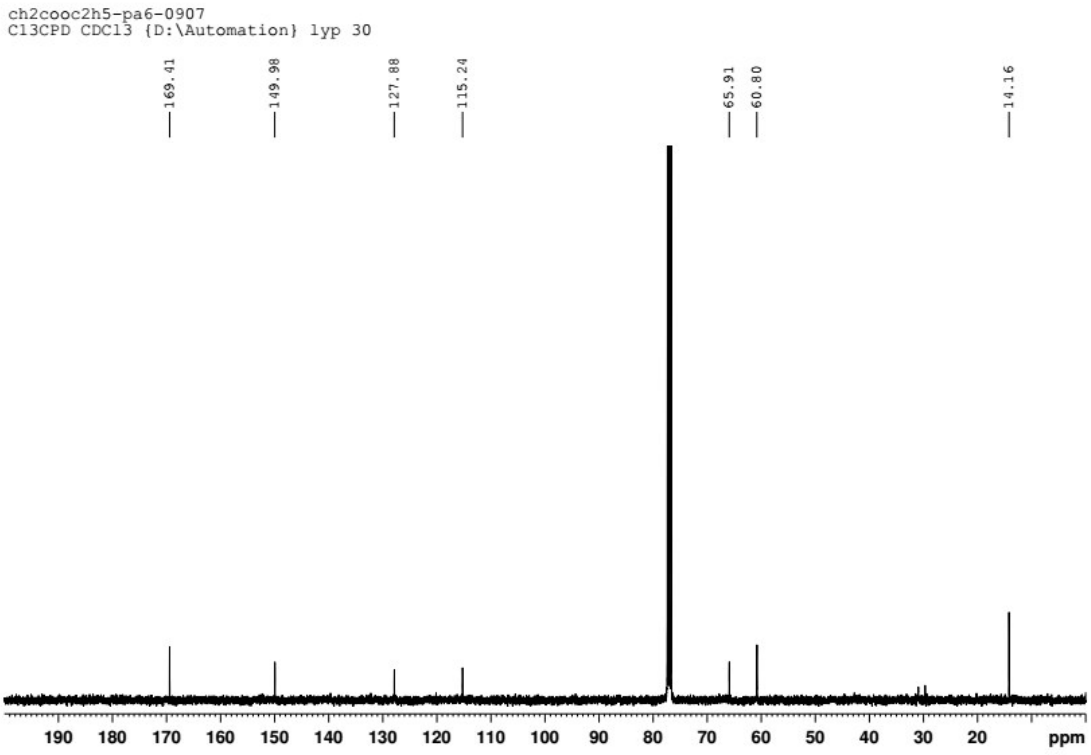



***Fig. S19*** $^{13}$C NMR spectra (100 MHz, $CDCl_3$, 293K)of **C**

Synthesis of **D**: To a solution of compound **C** (0.450 g, 0.255 mmol) in ethanol (45.0 mL) and tetrahydrofuran (45.0 mL) NaOH (0.900 g, 22.5 mmol) was added. The resulting reaction mixture was refluxed for 24 h. Then the organic solvents and most water were evaporated under vaccum. The residue was diluted with 100 mL of $H_2O$, and then acidified with HCl. The resulting precipitate was collected by filtration, washed with $H_2O$, and dried under vacuum to give **D** (0.357 g, 98.0%). The proton NMR spectrum of **D** is shown in Fig. S20. $^1$H NMR (DMSO-$d_6$, 400 MHz, 25 °C, TMS): $\delta$ 6.82 (s, 12H), 4.48 (d, 12H, $J$ = 4.8 Hz), 3.71 (s, 12H). The $^{13}$C NMR spectrum of **D** is shown in Fig. S21. $^{13}$C NMR (DMSO-$d_6$, 100 MHz, 25 °C, TMS): $\delta$ 170.6 (C of carbonyl), 149.3, 126.6, 114.6 (C of aromatic), 65.1 (C of O-methylene).

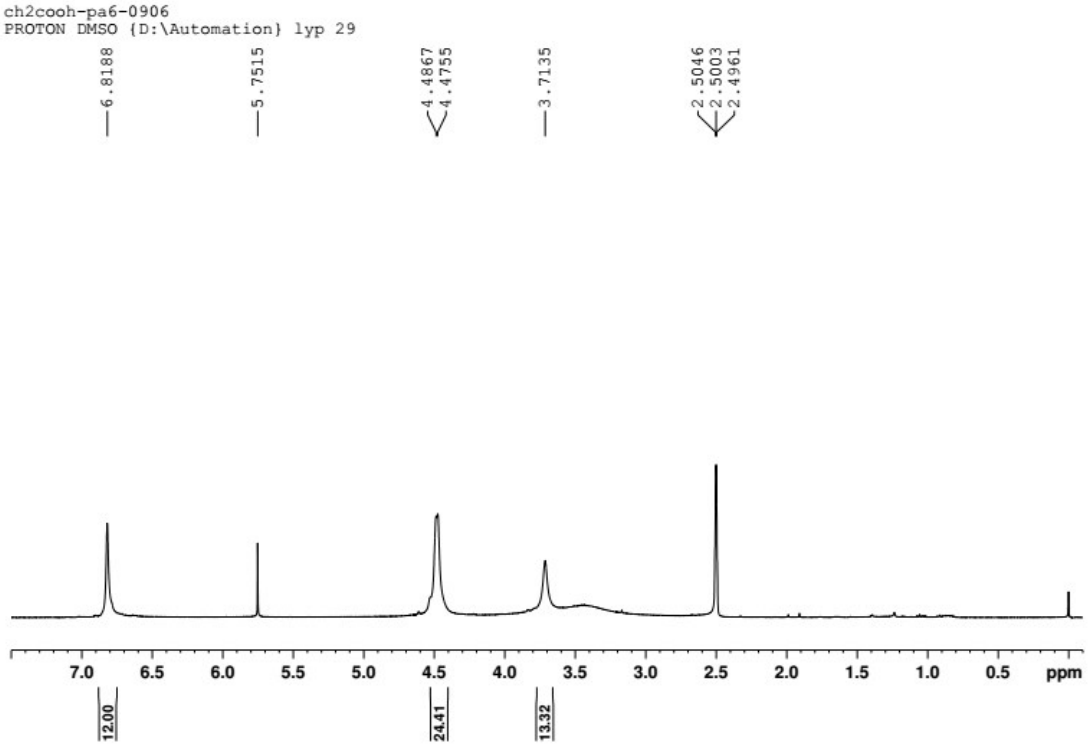



***Fig. S20*** $^1$H NMR spectra (400 MHz, DMSO-$d_6$, 293K) of **D**

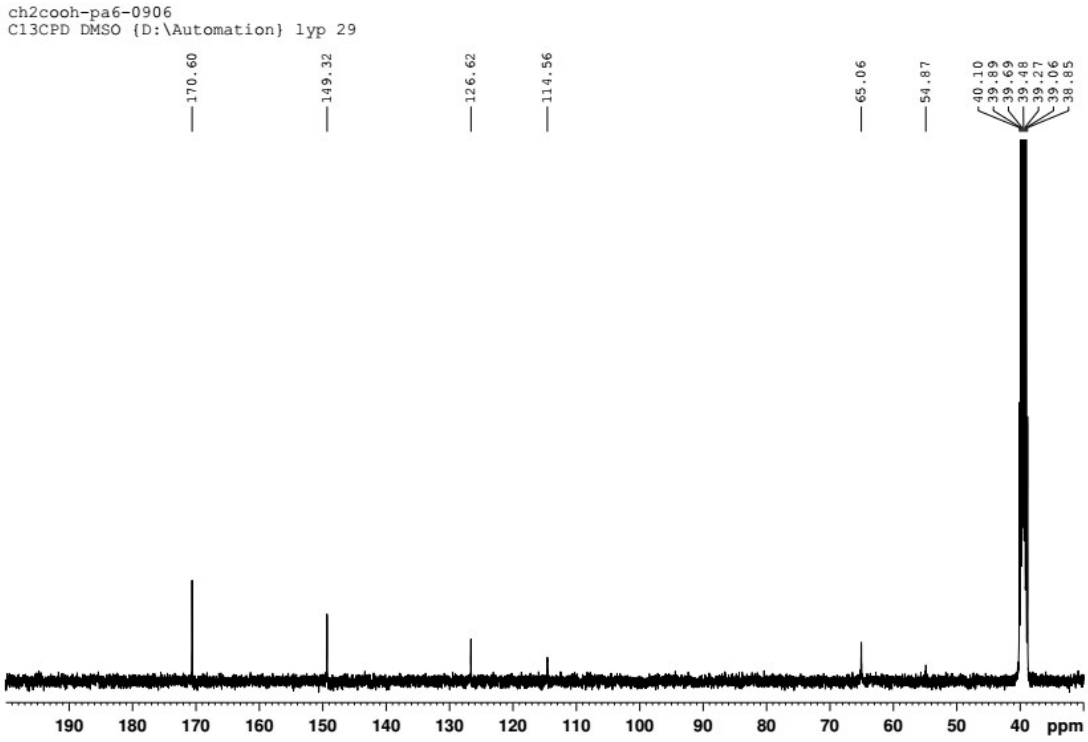



***Fig. S21*** $^{13}$C NMR spectra (100 MHz, DMSO-$d_6$, 293K)of **D**

Synthesis of **E**: To a solution of **D** (610 mg, 0.430 mmol) in $H_2O$ (30.0 mL) NaOH (17.0 mg, 0.430 mmol) were added and stirred at room temperature for 12 h. Water was removed under reduced pressure to give **E** as a white solid (0.723 g, 100%). The proton NMR spectrum of **E** is shown in Fig. S22. $^1$H NMR ($D_2O$, 400 MHz, 25 °C, TMS): $\delta$ 6.68 (s, 12H), 4.24 (s, 12H), 3.83 (s, 12H). The $^{13}$C NMR spectrum of **E** is shown in Fig. S23. $^{13}$C NMR ($D_2O$, 100 MHz, 25 °C, TMS): $\delta$ 178.1, 150.7, 128.9, 116.4, 68.8, 30.9.

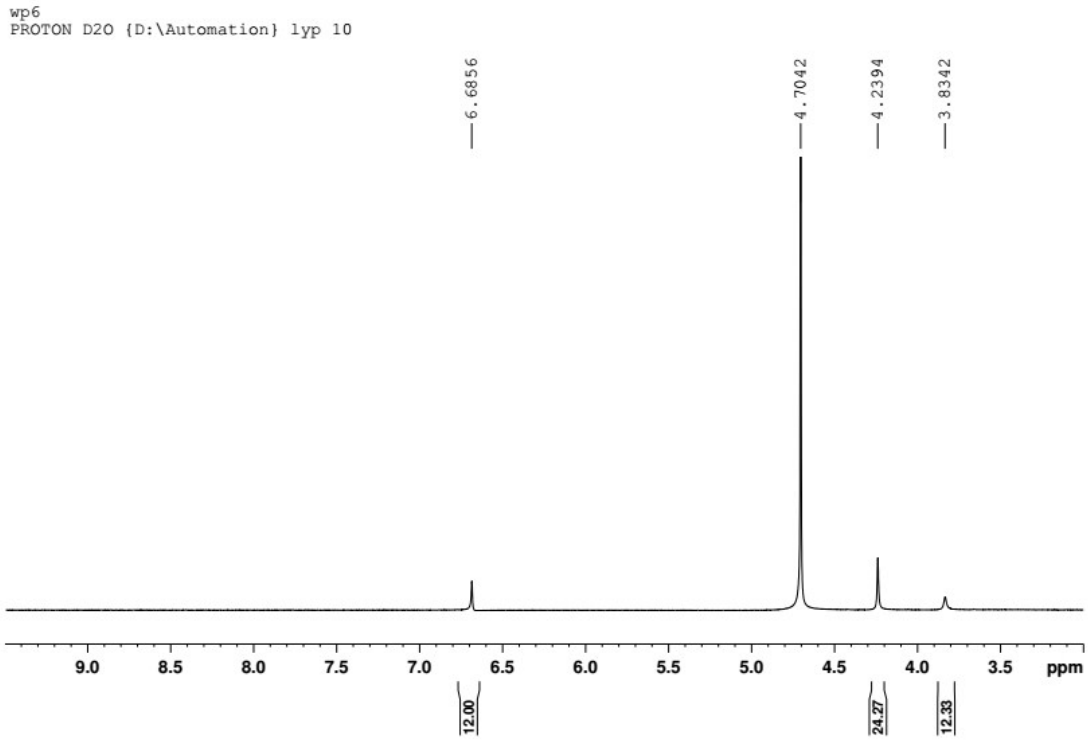



***Fig. S22*** $^{1}$H NMR spectra (400 MHz, $D_2O$, 293K) of **E**

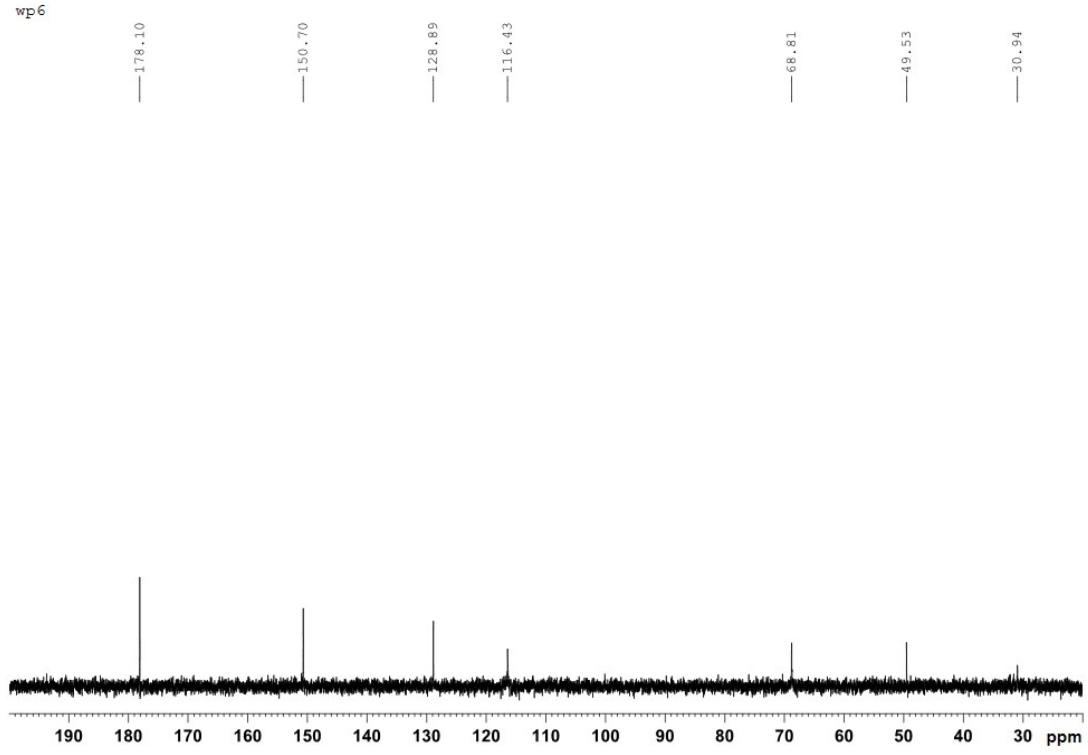
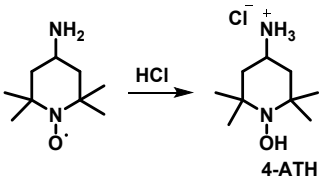



***Fig. S23*** $^{13}$C NMR spectra (400 MHz, $D_2O$, 293K) of **E**

*5.3. Synthesis of compound **4-ATH***




To a solution of **4-AT** (147 mg, 0.860 mmol) in ethanol (0.200 ml) aqueous hydrochloric acid (0.150 ml, 1.72 mmol) was added. The reaction mixture was stirred for 3h at room temperature until no EPR signal was detectable. The solvent was removed and the residue was recrystallized from 2-propanol to afford **4-ATH** as a white solid (0.536 g, 30%). The proton NMR spectrum of ***4-ATH*** is shown in Fig. S24. $^{1}$H NMR ($D_2O$, 400 MHz, 25 °C, TMS): $\delta$ 3.76 (m, 1H), 2.22 (d, 2H, $J$=13.76Hz), 1.85 (t, 2H, $J$=13.08, 13.32Hz), 1.34 (s, 12H). The $^{13}$C NMR spectrum of ***4-ATH*** is shown in Fig. S25. $^{13}$C NMR ($D_2O$, 100 MHz, 25 °C, TMS): $\delta$ 67.6, 41.2, 39.6, 27.0, 19.0.

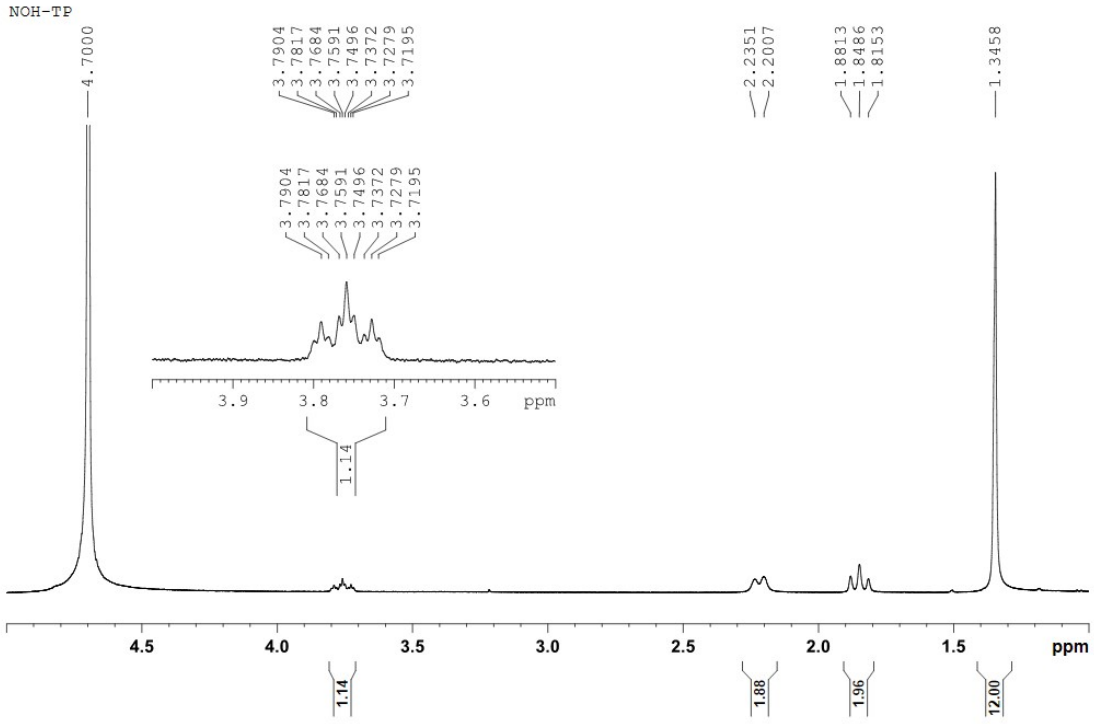



***Fig. S24*** $^{1}$H NMR spectra (400 MHz, $D_2O$, 293K) of **4-ATH**

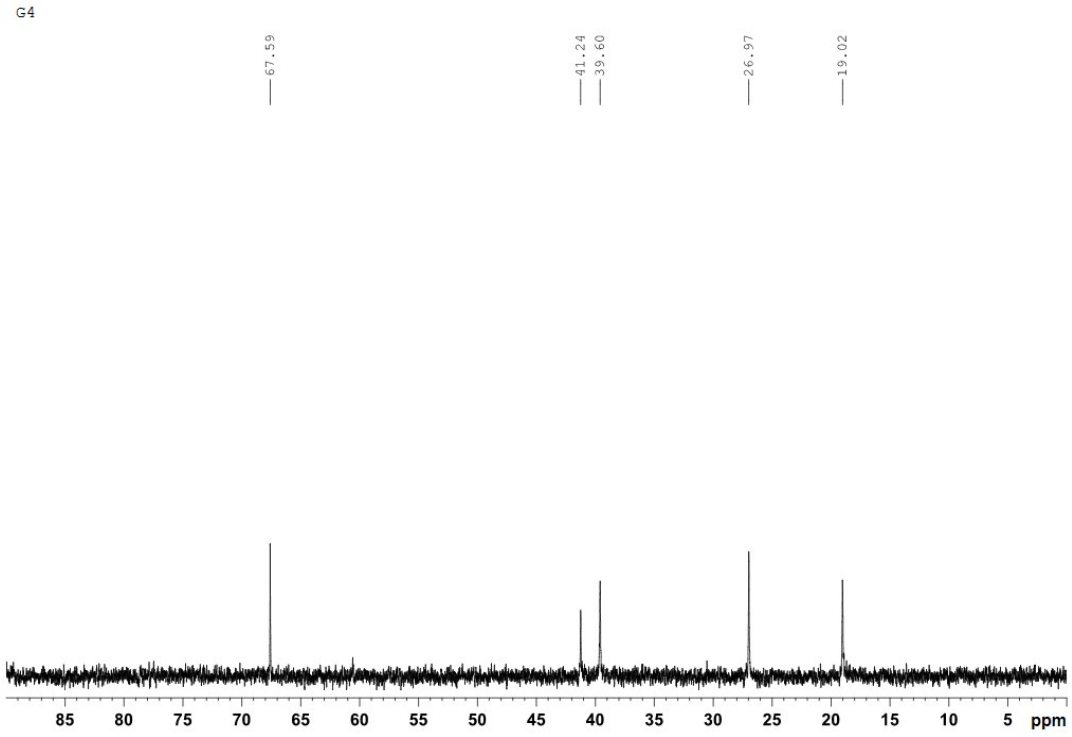



***Fig. S25*** $^{13}$C NMR spectra (100 MHz, $D_2O$, 293K) of **4-ATH**

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