

Supporting Information

A dual pH-responsive supramolecular gelator with aggregation-induced emission properties

Marlen Externbrink, Steffen Riebe, Carsten Schmuck*, Jens Voskuhl*

Institute for Organic Chemistry, University of Duisburg-Essen, 45117 Essen (Germany)

Table of Content

1. General information	S1
2. Synthesis	S2
3. NMR experiments	S7
4. Dynamic Light Scattering	S8
5. Atomic Force Microscopy	S9
6. Molecular Modelling	S9
7. Spectroscopic Measurements	S11
8. Gelation Procedure	S12
9. Rheology	S12
10. Fluorescence Microscopy	S12
11. Quantum Yield	S13
12. NMR spectra	S13
13. IR Spectra	S16
14. Mass Spectra	S16
15. HPLC Spectra	S17
16. References	S17

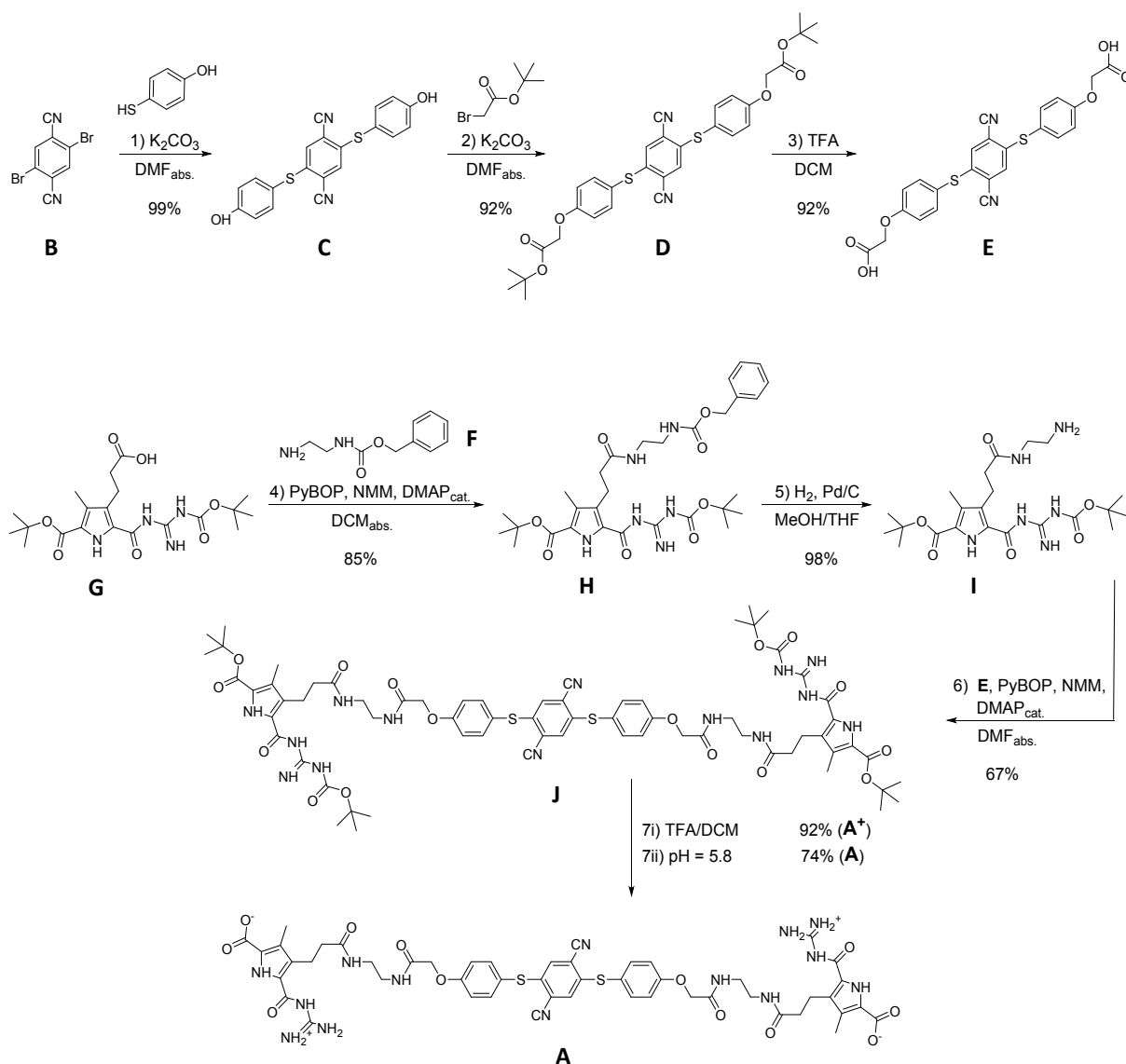
1. General information

All solvents were distilled before use. Water was purified with a TKA MicroPure ultrapure water system. All other reagents were used as obtained from commercial sources unless otherwise specified. Reactions were monitored by TLC on silica gel plates (Machery-Nagel POLYGRAM SIL G/UV254). Spots were visualised by UV light (254 nm and 366 nm). Lyophilisation was performed with a Christ Alpha 1–4 LDplus freeze dryer. The pH was determined with a pH-meter 766 Calimatic from Knick. The melting points were measured with a Büchi Melting-Point B-540 apparatus with open end glass capillary tubes. The melting points are not corrected. The NMR spectra were recorded with Bruker DMX 300, DRX 500 or AVHD 600 spectrometers. All measurements were performed at room temperature using DMSO-*d*₆ as solvent. The chemical shifts are relative to the

signals of DMSO-d₆ (δ ¹H = 2.50 ppm and δ ¹³C = 39.5 ppm). The apparent coupling constants are given in Hertz. The description of the fine structure means: s = singlet, br.s = broad singlet, d = doublet, t = triplet, m = multiplet. The IR spectra were measured on a Varian 3100 FT-IR Excalibur (compounds **C-E**) Series or a Jasco FT/IR-430 spectrometer (**A, H-J**). The low resolution ESI mass spectra were recorded with a Bruker amaZon SL and the high resolution ESI mass spectra with a Bruker maXis 4G UHR-TOF (**C-E**) or a Thermo Fischer Scientific Orbitrap LTQ XL (**A**). Analytical HPLC was performed on a Dionex HPLC apparatus that consisted of a P680 pump, an ASI-100 automated sample injector and an UVD 340U photodiode array detector with a YMC ODS-AQ column (column size: 150 x 3.0 mm, particle size: 5 μ m, pore size: 12 nm).

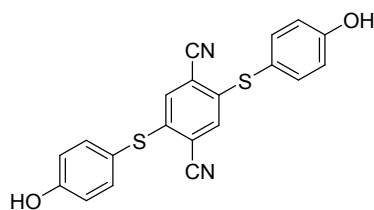
2. Synthesis

The AIE building block **E** was synthesised starting from 2,5-dibromoterephthalonitrile **B**.^[1] The synthesis of the GCP precursor **G** was previously reported by us^[2] and the linker unit Cbz-ethylenediamine **F** was prepared according to a literature procedure.^[3]



Scheme S1: Synthesis of the bis-zwitterion **A**.

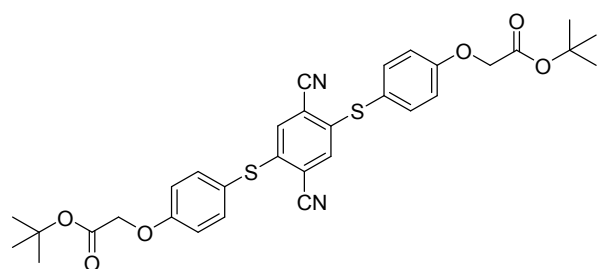
2,5-bis((4-hydroxyphenyl)thio)terephthalonitrile **C**



2,5-dibromoterephthalonitrile **B**^[1] (400 mg, 1.4 mmol, 1.0 eq), 4-hydroxythiophenol (818 mg, 6.5 mmol, 4.6 eq) and potassium carbonate (1.16 g, 8.4 mmol, 6.0 eq) were added to a 100 mL round bottom flask. The flask was evacuated and purged with argon twice. Then dry DMF (15 ml) was added and the resulting solution was stirred at 45 °C for 6 h. Upon careful quenching with 4 M HCl (50 ml) the product precipitated. The solid was separated by filtration and was washed with copious amounts of distilled water. After recrystallisation from DMF and drying in vacuo **C** (524 mg, 1.39 mmol, 99%) was obtained as a yellow-green solid. The product was synthesised according to Voskuhl et al.^[4]

Molecular Formula: C₂₀H₁₂N₂O₂S₂; **Molar Mass:** 376.45 g/mol; **Mp:** 365-370 °C (decomposition); **¹H NMR:** (300 MHz, DMSO-*d*₆) δ [ppm] = 6.90 (d, ³*J* = 8.5 Hz, 4H, Ar-*H*), 7.27 (s, 2H, Ar-*H*), 7.41 (d, ³*J* = 8.6 Hz, 4H, Ar-*H*), 10.15 (s, 2H, Ar-OH); **¹³C NMR:** (75 MHz, DMSO-*d*₆) δ [ppm] = 114.3, 115.0, 116.3, 117.1 (Ar-CH₂), 132.0, 136.4 (Ar-CH₂), 141.1, 159.2; **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3385, 3082, 3010, 2928, 2869, 2788, 2725, 2668, 2607, 2361, 2239, 2224, 1668, 1599, 1579, 1494, 1447, 1390, 1345, 1264, 1230, 1170, 1147, 1095, 1056, 1011, 881, 840, 767, 723, 709, 671, 656; **HR-MS** (neg. ESI, MeOH) *m/z* = 374.9821 ([M-H]⁻, calc.: 375.0267).

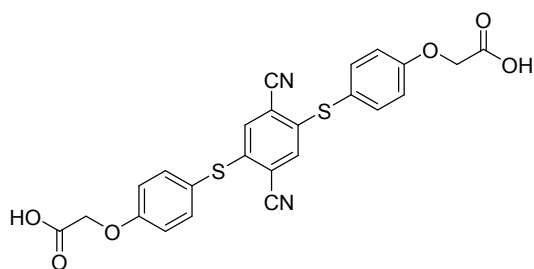
Di-*tert*-butylester **D**



C (200 mg, 0.53 mmol, 1.0 eq) and potassium carbonate (442 mg, 3.2 mmol, 6.0 eq) were added to a 100 ml round bottom flask. Dry DMF (12 ml) and *tert*-butyl bromoacetate (312 mg, 1.6 mmol, 3.0 eq) were added and the solution was stirred at 40 °C for 24 h. Then the reaction was quenched with distilled water and the product precipitated as a white solid. This solid was separated by filtration and washed with copious amounts of distilled water. Finally, the product was dried in vacuo to give **D** (297 mg, 0.49 mmol, 92%) as a white solid.

Molecular Formula: C₃₂H₃₂N₂O₆S₂; **Molar Mass:** 604.74 g/mol; **Mp:** 187 °C; **¹H NMR:** (300 MHz, DMSO-*d*₆) δ [ppm] = 1.42 (s, 18H, C(CH₃)₃), 4.75 (s, 4H, CH₂), 7.05 (d, ³*J* = 8.9 Hz, 4H, Ar-*H*), 7.38 (s, 2H, Ar-*H*), 7.52 (d, ³*J* = 8.8 Hz, 4H, Ar-*H*); **¹³C NMR:** (75 MHz, DMSO-*d*₆) δ [ppm] = 27.7 (C(CH₃)₃), 65.0 (CH₂), 81.6 (C(CH₃)₃), 115.2, 115.4, 116.5 (Ar-CH₂), 120.0, 133.1, 136.0 (Ar-CH₂), 140.8, 159.2, 167.5 (CO); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 2987, 2938, 2871, 2225, 1749, 1724, 1681, 1589, 1573, 1492, 1448, 1409, 1393, 1367, 1345, 1310, 1292, 1262, 1223, 1177, 1151, 1105, 1092, 1065, 1008, 943, 921, 884, 837, 802, 781; **HR-MS** (pos. ESI, MeOH) *m/z* = 627.1607 ([M+Na]⁺, calc.: 627.1594).

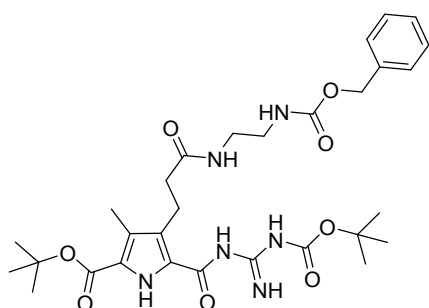
Di-carboxylic acid E



D (150 mg, 0.25 mmol) was added to a 100 ml round bottom flask and was dissolved in dichloromethane (15 ml) under stirring. Then trifluoroacetic acid (4 ml) was added and the solution was stirred at 25 °C for 6 h. The solvent was removed in vacuo and **E** was obtained (112 mg 0.23 mmol, 92%) as a white solid.

Molecular Formula: C₂₄H₁₆N₂O₆S₂; **Molar Mass:** 492.52 g/mol; **Mp:** 299 °C (decomposition); **¹H NMR:** (300 MHz, DMSO-*d*₆) δ [ppm] = 4.76 (s, 4H, OCH₂), 7.06 (d, ³J = 8.9 Hz, 4H, Ar-H), 7.41 (s, 2H, Ar-H), 7.52 (d, ³J = 8.8 Hz, 4H, Ar-H), 13.05 (br.s, 2H, COOH); **¹³C NMR:** (75 MHz, DMSO-*d*₆) δ [ppm] = 64.6 (CH₂), 115.3, 115.4, 116.5 (Ar-CH₂), 120.0, 133.3, 136.0 (Ar-CH₂), 140.9, 159.2, 169.8 (COOH); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3060, 2323, 2224, 1733, 1707, 1570, 1655, 1593, 1576, 1542, 1494, 1474, 1455, 1427, 1408, 1372, 1341, 1315, 1305, 1286, 1267, 1242, 1179, 1150, 1108, 1094, 1081, 1010, 916, 896, 833, 813, 800, 725, 712, 652; **HR-MS** (neg. ESI, MeOH) *m/z* = 491.0367 [M-H]⁻, calc.: 491.0366).

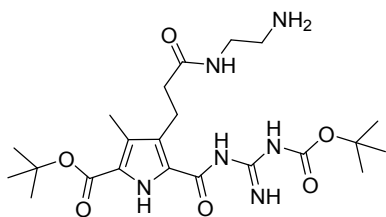
Synthesis of the GCP-derivative H



The GCP precursor **G**^[2] (4.00 g, 9.12 mmol, 1.0 eq), PyBOP (5.698 g, 11.0 mmol, 1.2 eq), NMM (7.0 ml, 63.8 mmol, 7.0 eq) and a catalytic amount of DMAP were dissolved in dry DCM (100 ml). After 10 min Cbz-ethylenediamine **F**^[3] (6.75 g, 11.0 mmol, 1.2 eq) was added dropwise and the reaction mixture was stirred at room temperature overnight. After removal of the solvent, the residue was taken up in EtOAc (400 ml) and washed with 0.5 N NaHSO₄. (3 x 150 ml) and saturated NaHCO₃ (3 x 150 ml). The organic layer was dried over MgSO₄ and evaporated under reduced pressure. The crude product was purified by column chromatography (SiO₂, EtOAc/cyclohexane = 3/2, R_f = 0.34) to give **H** (4.79 g, 7.79 mmol, 85%) as a white solid.

Molecular Formula: C₃₀H₄₂N₆O₈; **Molar Mass:** 614.70 g/mol; **Mp:** 159 °C; **¹H NMR:** (300 MHz, DMSO-*d*₆) δ [ppm] = 1.47 (s, 9H, C(CH₃)₃), 1.53 (s, 9H, C(CH₃)₃), 2.16 (s, 3H, PyCH₃), 2.23 (t, 2H, PyCH₂CH₂), 2.93 (t, 2H, PyCH₂), 2.97-3.14 (m, 4H, NHCH₂CH₂NH), 5.00 (s, 2H, Cbz-CH₂), 7.24 (t, 1H, Linker-NH), 7.26-7.43 (m, 5H, Cbz-CH), 7.81 (t, 1H, Linker-NH), 8.44, 9.43, 10.14, 10.62 (all br.s, 1H, NH); **¹³C NMR:** (75 MHz, CDCl₃) δ [ppm] = 9.8 (PyCH₃), 20.5 (PyCH₂), 27.7 (C(CH₃)₃), 28.0 (C(CH₃)₃), 36.4 (PyCH₂CH₂), 38.5, 40.0 (NHCH₂CH₂NH), 65.2 (Cbz-CH₂), 80.6 (C(CH₃)₃), 125.3 (C_q), 127.7, 128.3 (both Cbz-CH), 137.1, 156.1, 157.1, 160.2, 172.2 (all C_q); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3385, 3343, 3307, 3067, 2987, 2975, 2936, 2875, 1709, 1689, 1624, 1533, 1467, 1366, 1297, 1235, 1143, 1095, 1026, 993, 973, 839, 806, 770, 740, 700; **LR-MS** (pos. ESI, MeOH) *m/z* = 615.3 ([M+H]⁺, calc.: 615.3), 637.3 ([M+Na]⁺, calc.: 637.3).

Cbz-deprotection to I

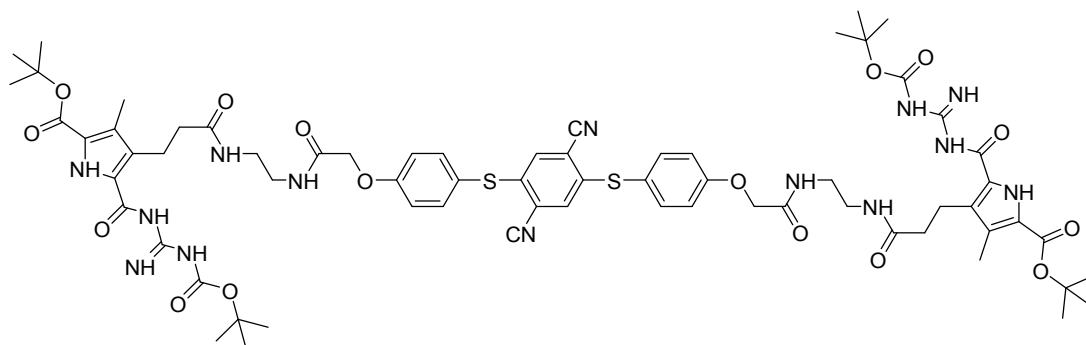


To a solution of **H** (2.00 g, 3.25 mmol) in MeOH/THF (4/1, 50 ml) was added 10% Pd/C (200 mg). The resulting suspension was stirred vigorously under a hydrogen atmosphere at room temperature. After complete consumption of the starting material (TLC analysis), the catalyst was filtered off and washed with THF. The filtrate was evaporated under reduced pressure.

The resulting oil was taken up in a small amount of MeOH and precipitated in water (75 ml). Lyophilisation gave **I** (1.53 g, 3.19 mmol, 98%) as an off-white solid.

Molecular Formula: C₂₂H₃₆N₆O₆; **Molecular Mass:** 480.57 g/mol; **Mp:** 103 °C; **¹H NMR:** (300 MHz, DMSO-*d*₆) δ [ppm] = 1.48 (s, 9H, C(CH₃)₃), 1.53 (s, 9H, C(CH₃)₃), 2.16 (s, 3H, PyCH₃), 2.25 (t, 2H, PyCH₂CH₂), 2.53 (t, 2H, NH₂CH₂), 2.94 (t, 2H, PyCH₂), 2.98-3.06 (m, 2H, CH₂NH), 7.74 (br.s, 2H, NH), 8.51 (br.s, 2H, NH); **¹³C NMR:** (75 MHz, CDCl₃) δ [ppm] = 9.8 (PyCH₃), 20.6 (PyCH₂), 27.7 (C(CH₃)₃), 28.0 (C(CH₃)₃), 36.4 (PyCH₂CH₂), 41.2, 42.2 (NH₂CH₂CH₂NH), 80.6 (C(CH₃)₃), 81.5 (C(CH₃)₃), 120.8, 125.3, 128.7, 158.0, 160.2, 172.2 (all C_q); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3458, 3383, 3278, 2976, 2934, 2866, 1725, 1691, 1627, 1531, 1454, 1393, 1268, 1297, 1238, 1142, 1100, 975, 883, 839, 805, 776, 753.

Protected zwitterion J

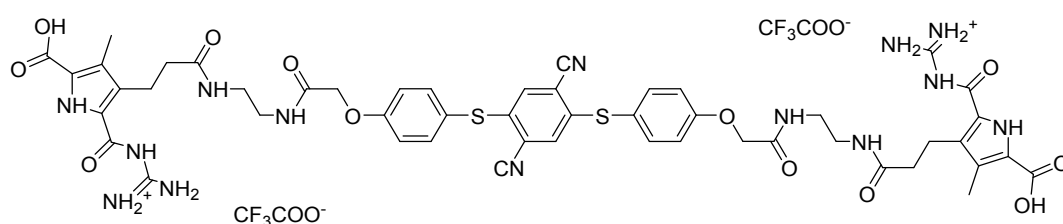


A mixture of AIE building block **E** (105 mg, 213 μmol, 1.0 eq), PyBOP (266 mg, 512 μmol, 2.4 eq), NMM (0.23 ml, 2.13 mmol, 10.0 eq) and a catalytic amount of DMAP were stirred in dry DMF (20 ml) at 50 °C for 20 min. The guanidinopyrrole **I** (246 mg, 512 μmol, 2.4 eq) was added to the solution, which was stirred for 24 h at room temperature. More PyBOP (89 mg, 171 μmol, 0.8 eq) was added and the reaction was stirred for further 24 h. Then the solvent was removed under reduced pressure. The resulting residue was taken up in a small amount of MeOH and was poured into ice-water (400 ml) under vigorous stirring. The precipitate was filtered off and dissolved in EtOAc (175 ml). The solution was washed with 0.5 N NaHSO₄ (3 x 100 ml) and saturated NaHCO₃ (3 x 100 ml). The organic layer was dried over MgSO₄ and evaporated. The crude product was purified by column chromatography (SiO₂, EtOAc/MeOH = 10/0.15, R_f = 0.40) to give **J** (202 mg, 142 μmol, 67%) as a slightly yellow solid.

Molecular Formula: C₆₈H₈₄N₁₄O₁₆S₂; **Molecular Mass:** 1417.62 g/mol; **Mp:** 136 °C; **¹H NMR** (300 MHz, DMSO-*d*₆) δ [ppm] = 1.47 (s, 18H, C(CH₃)₃), 1.52 (s, 18H, C(CH₃)₃), 2.16 (s, 6H, PyCH₃), 2.24 (t,

4H, PyCH₂CH₂), 2.93 (t, 4H, PyCH₂), 3.06-3.24 (m, 8H, NHCH₂CH₂NH), 4.53 (s, 4H, OCH₂), 7.10 (d, ³J_{H,H} = 8.9 Hz, 4H, Ar-H), 7.35 (s, 2H, Ar-H), 7.53 (d, ³J_{H,H} = 8.8 Hz, 4H, Ar-H), 7.85 (t, ³J_{H,H} = 5.3 Hz, 2H, Linker-NH), 8.19 (t, ³J_{H,H} = 5.4 Hz, 2H, Linker-NH), 8.46 (br.s, 2H, Gua-NH), 9.40, (br.s, 2H, Gua NH), 10.13 (br.s, 2H, Gua-amide-NH), 10.64 (br.s, 2H, Py-NH); ¹³C NMR (75 MHz, DMSO-d₆) δ [ppm] = 9.8 (PyCH₃), 20.5 (PyCH₂), 27.7 (C(CH₃)₃), 28.0 (C(CH₃)₃), 36.3 (PyCH₂CH₂), 38.1, 38.4 (NH₂CH₂CH₂NH), 67.0 (OCH₂), 80.6 (C(CH₃)₃), 115.2 (C_q-CN), 115.2 (CN), 116.7 (Ar-CH), 120.0, 125.3 (both C_q), 134.0 (Ar-CH), 136.0 (Ar-CH), 140.8, 156.5, 159.2, 160.2, 167.3, 172.3 (all C_q); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3391, 3083, 2979, 2932, 2229, 1726, 1628, 1591, 1532, 1491, 1452, 1393, 1368, 1293, 1238, 1144, 1103, 974, 836, 805, 775, 753; **LR-MS** (pos. ESI, MeOH) *m/z* = 1439.7 ([M+Na]⁺, calc.: 1439.6), 1417.7 ([M+H]⁺, calc.: 1417.6).

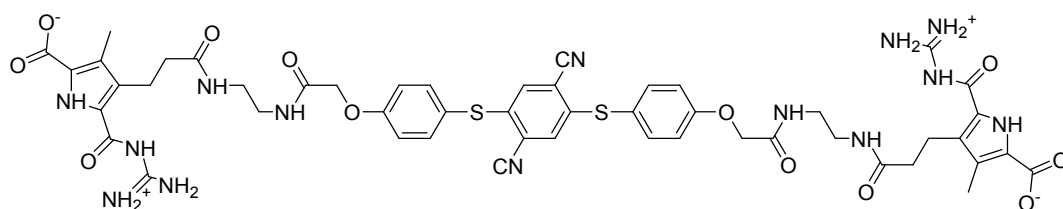
TFA-salt A⁺



J (101 mg, 71.2 μmol, 1.0 eq) was dissolved in DCM (1.5 ml) followed by the dropwise addition of TFA (0.75 ml). The reaction mixture was stirred at room temperature overnight. After removal of the solvent under reduced pressure, the residue was taken up in a small amount of MeOH and was precipitated in water. Lyophilisation gave the TFA-salt **A⁺** (87 mg, 65.3 μmol, 92%) as a slightly yellow powder.

Molecular Formula: C₅₄H₅₄F₆N₁₄O₁₆S₂; **Molecular Mass:** 1333.22 g/mol; **¹H NMR:** (300 MHz, DMSO-d₆) δ [ppm] = 2.21 (s, 6H, PyCH₃), 2.28 (t, 4H, PyCH₂CH₂), 2.90 (t, 4H, PyCH₂), 3.01-3.22 (m, 8H, NHCH₂CH₂NH), 4.53 (s, 4H, OCH₂), 7.10 (d, ³J_{H,H} = 8.9 Hz, 4H, Ar-H), 7.37 (s, 2H, Ar-H), 7.53 (d, ³J_{H,H} = 8.8 Hz, 4H, Ar-H), 7.91 (t, 2H, Linker-NH), 8.21 (t, 2H, Linker-NH), 8.33 (br.s, 8H, Gua-NH), 11.31 (br.s, 2H, Gua-amide-NH), 11.84 (br.s, 2H, Py-NH), 13.08 (br.s, 2H, COOH).

Bis-zwitterion A



The TFA-salt **A⁺** (87 mg, 65.3 μmol) was suspended in water and the pH was adjusted to 5.8 with 0.1 M NaOH and 0.1 M HCl under vigorous stirring. Then the suspension was sonicated for 15 min. These two steps were repeated until the pH remained constant at 5.8. After that the suspension was centrifuged and the supernatant was discarded whereas the remaining solid was washed thoroughly with water. After lyophilisation the bis-zwitterion **A** (53 mg, 48.0 μmol, 74%) was obtained as a slightly yellow powder.

Molecular Formula: C₅₀H₅₂N₁₄O₁₂S₂; **Molecular Mass:** 1105.17 g/mol; **Mp:** 240 °C (decomposition); **¹H NMR:** (500 MHz, DMSO-*d*₆) δ [ppm] = 2.20 (t, 4H, PyCH₂CH₂), 2.23 (s, 6H, PyCH₃), 2.91 (t, 4H, PyCH₂), 3.18 (m, 8H, NHCH₂CH₂NH), 4.53 (s, 4H, OCH₂), 6.83 (s, 2H, Ar-H), 6.97 (d, ³J_{H,H} = 8.7 Hz, 4H, Ar-H), 7.40 (d, ³J_{H,H} = 8.6 Hz, 4H, Ar-H), 7.59 (t, 2H, Linker-NH), 7.93 (br.s, 4H, Gua-NH), 8.05 (t, 2H, Linker-NH), 10.15 (br.s, 4H, Gua-NH), 12.50 (br.s, 2H, Py-NH), 14.60 (br.s, 2H, Gua-amide-NH); **¹³C NMR:** (150 MHz, DMSO-*d*₆) δ [ppm] = 9.4 (Py-CH₃), 20.9 (Py-CH₂), 35.8 (Py-CH₂CH₂), 38.1, 38.6 (NHCH₂CH₂NH), 67.0 (OCH₂), 114.8 (C_q-CN), 114.9 (CN), 116.7 (Ar-CH), 118.4, 120.2, 123.0, 129.4 (all C_q), 132.0 (Ar-CH), 134.2 (C_q), 135.9 (Ar-CH), 140.7, 156.6, 159.4, 161.1, 166.4, 167.6, 172.2 (all C_q); **FT-IR:** (ATR) $\tilde{\nu}$ [cm⁻¹] = 3309, 3192, 2921, 2853, 2741, 2227, 1714, 1652, 1591, 1540, 1490, 1443, 1335, 1271, 1244, 1224, 1176, 1155, 1122, 1090, 1052, 1008, 951, 884, 814, 767; **HR-MS:** (neg. ESI, DMSO) *m/z* = 1103.3240 ([M-H]⁻, calc.: 1103.3258), 551.1589 ([M-2H]²⁻, calc.: 551.1592).

3. NMR experiments

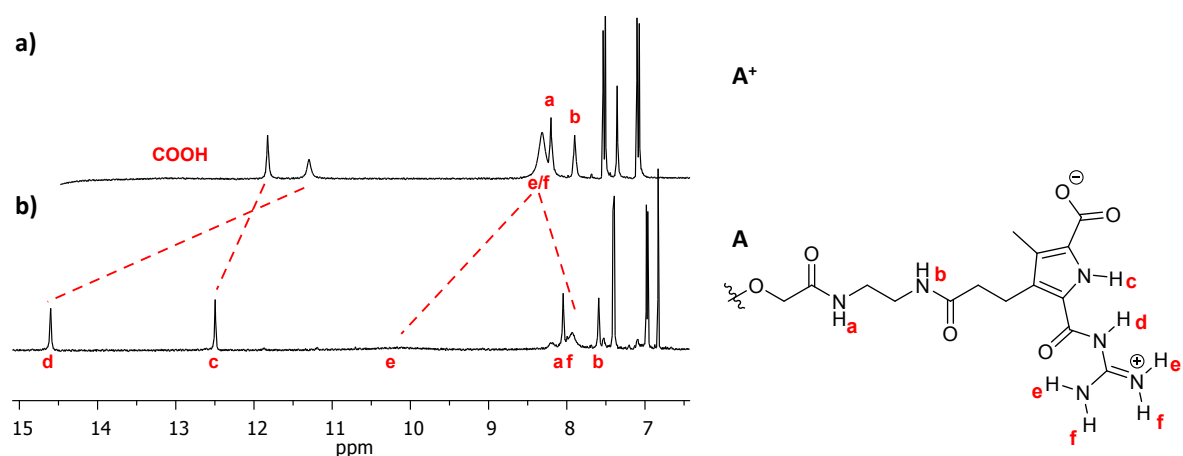


Fig. S1: Section of the ¹H NMR spectra of a) the protonated form A⁺ and b) the zwitterionic species A (c = 1 mM, DMSO-*d*₆). The self-complementary GCP zwitterions form stable dimers which can be followed by the characteristic proton shifts (dotted red line).

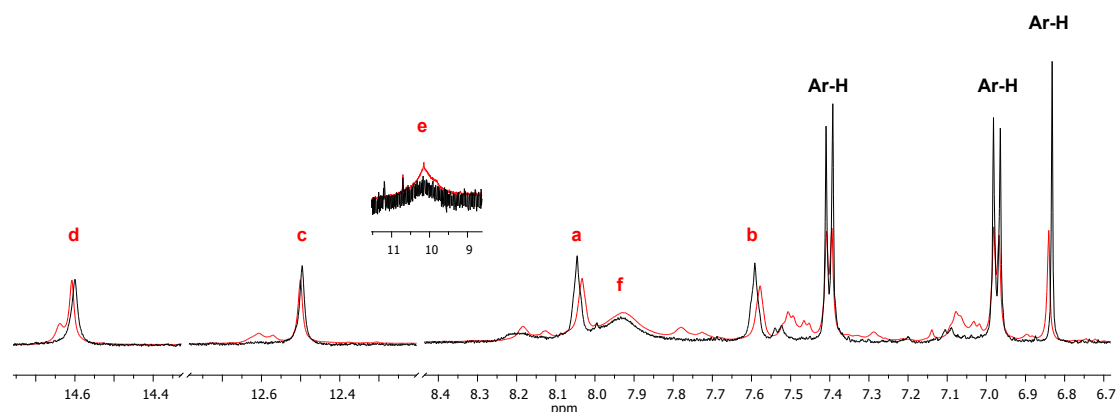


Fig. S2: Superimposition of the ¹H NMR spectra of A (in DMSO-*d*₆) at a concentration of c = 1 mM (black) and c = 12 mM (red) showing the formation of larger aggregates at higher concentrations.

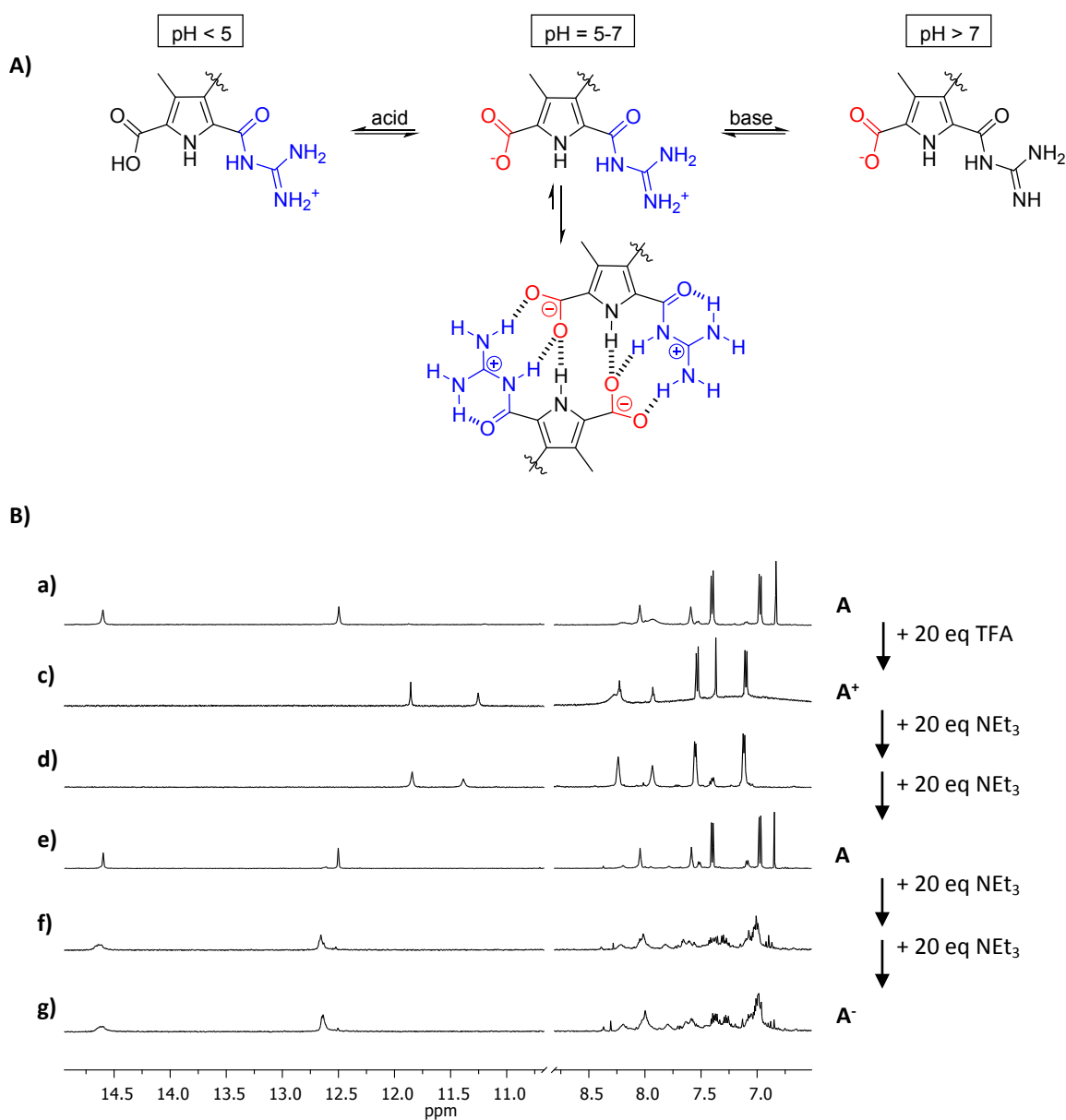


Fig. S3: A) Schematic representation of the pH-switchability of the GCP binding motif: The self-complementary GCP zwitterions exist in a pH-range around 5-7. Only the zwitterionic species form stable dimers held together by H-bond assisted ion-pairs. B) ¹H NMR spectra of a) the zwitterionic species **A** (*c* = 1 mM in DMSO-*d*₆) and b-f) the resulting ¹H NMR spectra after sequential addition of TFA and NEt₃ (20 eq acid/base per step) following the transition to the protonated form **A⁺**, back to zwitterion **A** and to the deprotonated form **A⁻**.

4. Dynamic Light Scattering (DLS)

Dynamic Light Scattering was measured on a Malvern Zetasizer Nano ZS equipped with a HeNe laser operating at a wavelength of 633 nm and a detector at an angle of 173° (non-invasive back scatter technology). Measurements were performed with sample solutions in UV cuvettes at 20 °C with 3 min equilibration time.

5. Atomic force microscopy (AFM)

Atomic force microscopy images were obtained in tapping mode using a NanoDrive Controller with an Innova Scanning Probe Microscope (Veeco) and N-type silicon cantilever (Olympus AC 160TS). Samples were prepared by spin-coating (60 rps) the solutions onto a freshly cleaved mica surface (Plano) for 10 min. The AFM data were analysed using Gwyddion-2.49 software.

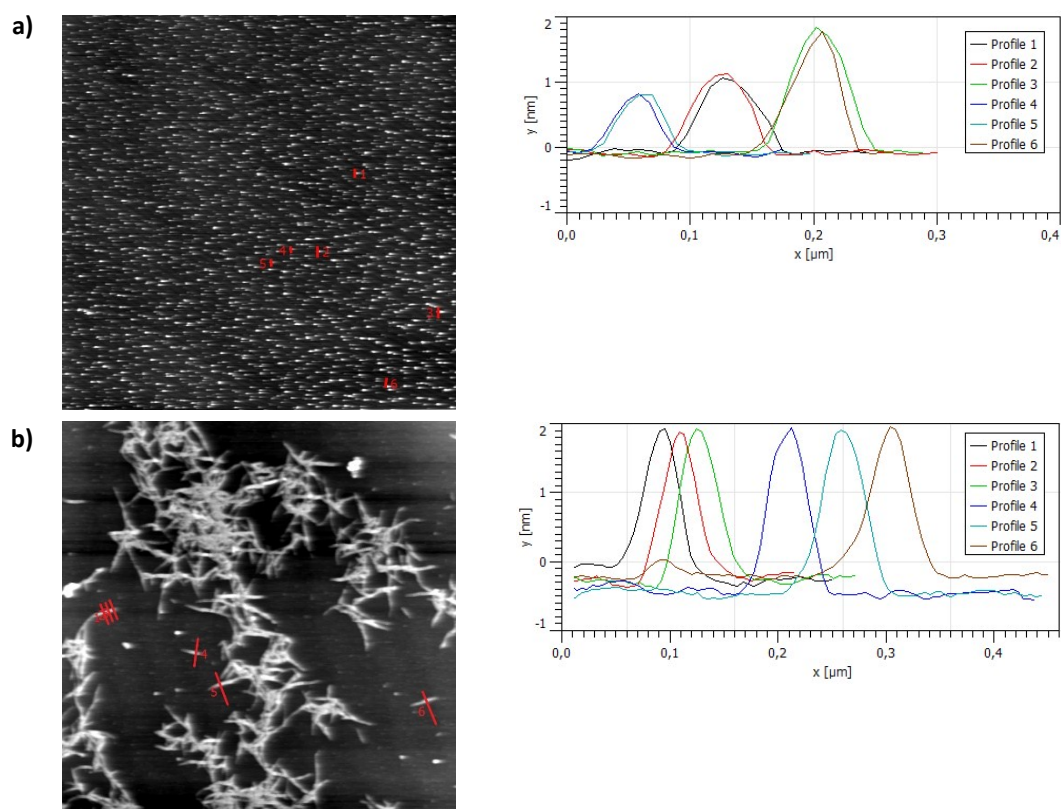


Fig. S4: AFM height images (10 μm x 10 μm) and height profiles of **A** at different concentrations in DMSO: a) ring-shaped aggregates at $c = 0.05$ mM and b) fibrillar aggregates at $c = 0.5$ mM.

6. Molecular Modelling

Force field calculations were performed using the Schrödinger Suite (release 2018-1: MacroModel v11.9, Desmond v5.3) to estimate the size of the protonated/deprotonated species $\mathbf{A}^+/\mathbf{A}^-$ (in comparison with DLS experiments) as well as to set up models for the ring-shaped and fibrillar aggregates (as seen by AFM).

The conformational search analysis was carried out with MacroModel using the mixed MCMM/LMCS method (Monte Carlo Multiple Minimum/Low-Mode Conformational Search) until the energy-minimum structure was found more than 50 times. The calculations were based on the force field OPLS_2005 (Optimised Potentials for Liquid Simulations) choosing water as solvent (GB/SA solvation model) and the energy minimisation was employed with the PRCG method (Polak-Ribiere Conjugate Gradient, convergence threshold < 0.05 kJ/(mol $\cdot\text{\AA}$)). Then the resulting minimum structures were subjected to molecular dynamics (MD) simulations (solvent: DMSO, temperature: 300 K, simulation time: ≥ 1.2 ns) in Desmond to probe their conformational stability.

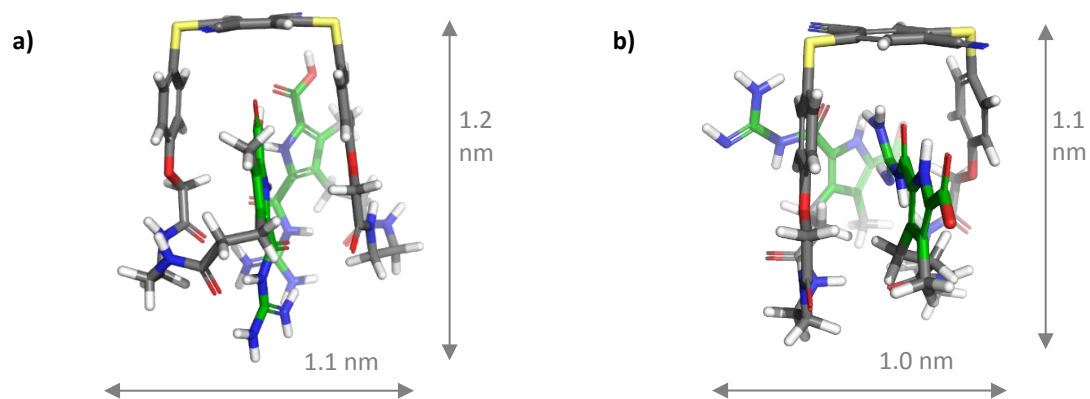


Fig. S5: Calculated structure of a) the protonated form A^+ and b) the deprotonated form A^- with a maximum length of 1.5 nm in both cases. The GCP units are marked green.

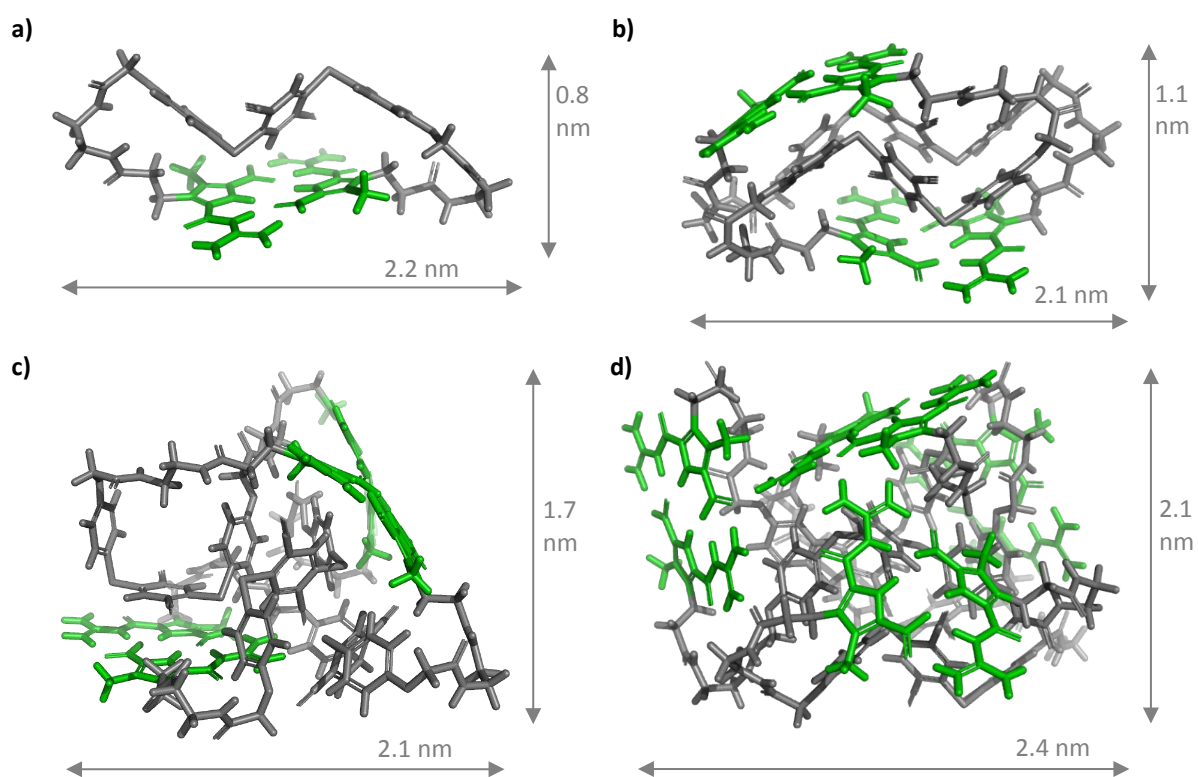


Fig. S6: Calculated structures of ring-shaped aggregates built of a) one, b) two, c) three and d) four monomers of bis-zwitterion **A**. For clarity in representation, the GCP zwitterions are coloured green and the AIE scaffolds as well as the spacer units grey.

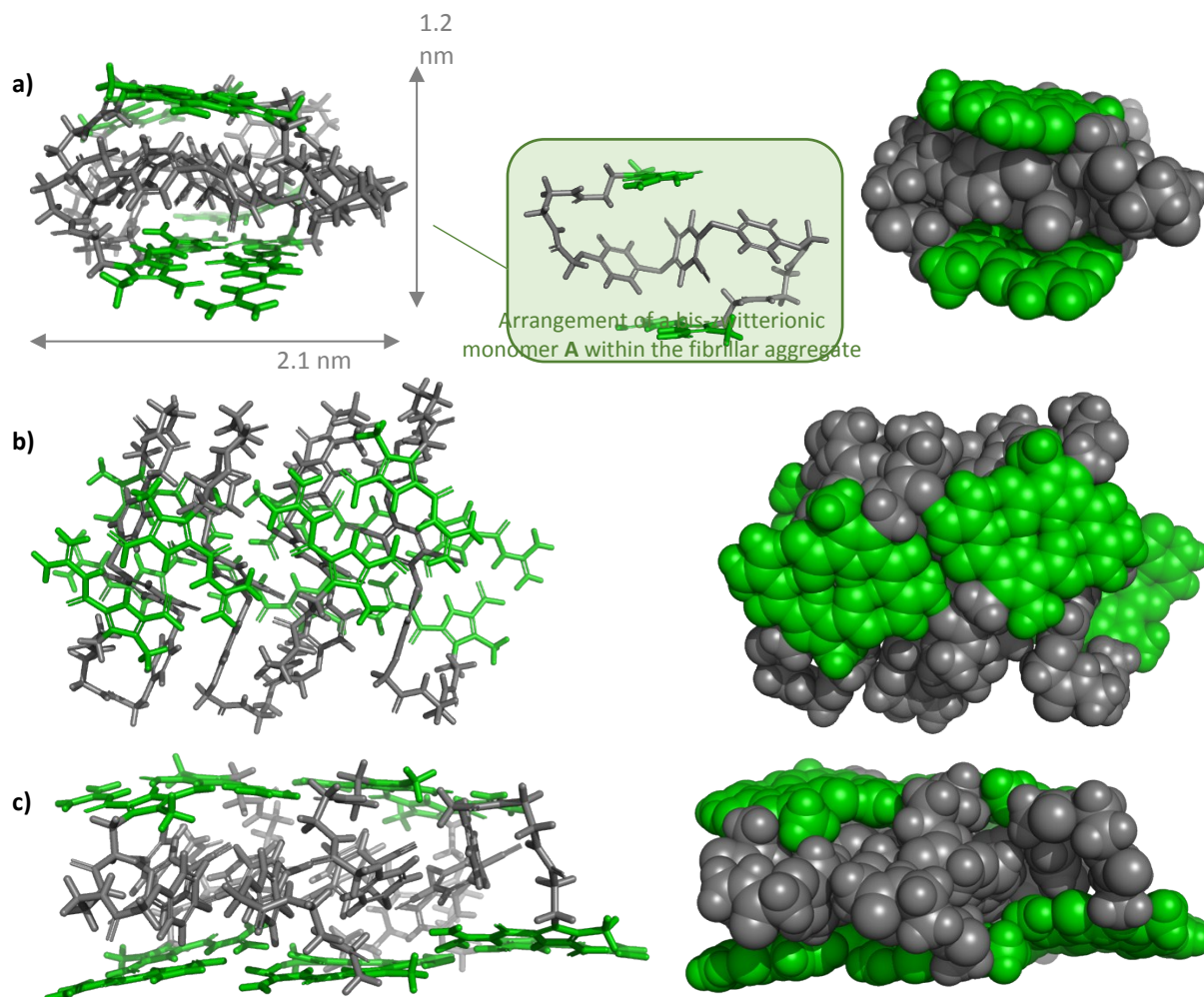


Fig. S7: Calculated structure of a short section of the fibrillar aggregates built of four monomers of **A** (stick and sphere representation): a) front view, b) top view, c) side view. The free zwitterionic ends of the tetramer were masked with simple GCP zwitterions (5-(guanidiniocarbonyl)-1*H*-pyrrole-2-carboxylate). For clarity in representation, the GCP zwitterions are coloured green and the AIE scaffolds as well as the spacer units grey.

7. Spectroscopic Measurements

Absorption spectra were measured von a Jasco V-660 and emission spectra were recorded on a Jasco FP-6500. All solvents used were of spectroscopic grade. All measurements were performed in quartz cuvettes.

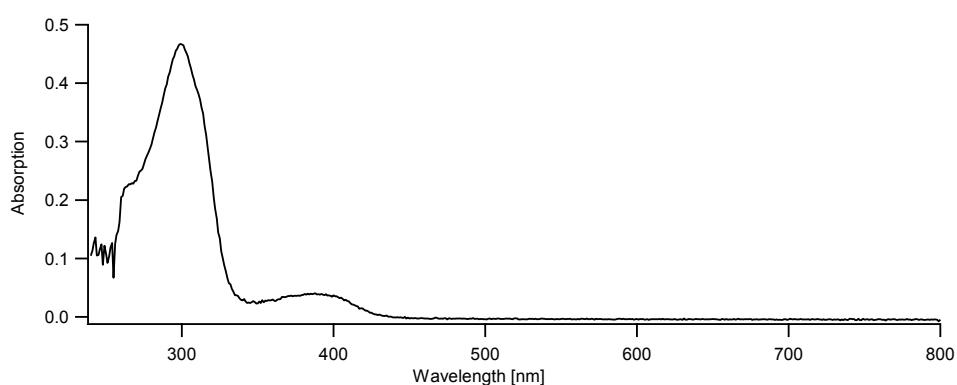


Fig. S8: Absorption spectrum of **A** ($c = 10 \mu\text{M}$, DMSO).

8. Gelation Procedure

In a glass vial, a weighed amount of **A** was taken and DMSO was added to it. The sample was sonicated in an ultrasonic bath for 10 minutes and heated at 120 °C for 3 minutes repeatedly. On keeping the sample at room temperature, the gel formation was observed within a few minutes to hours (depending on the sample concentration). The gel state was evaluated by the “stable to inversion of a test tube” method.

9. Rheology

The rheology experiments were performed on a Physica MCR 301 rheometer (Anton Paar) with a plate-plate geometry (diameter 15 mm). The temperature of the plate was controlled at 25 °C (± 0.1 °C) and the samples were loaded as hot sols. The gels were stabilised in the geometry gap (0.15 mm) for 2 h before performing the experiments. The solvent evaporation was minimised by placing a metallic cover as a solvent trap.

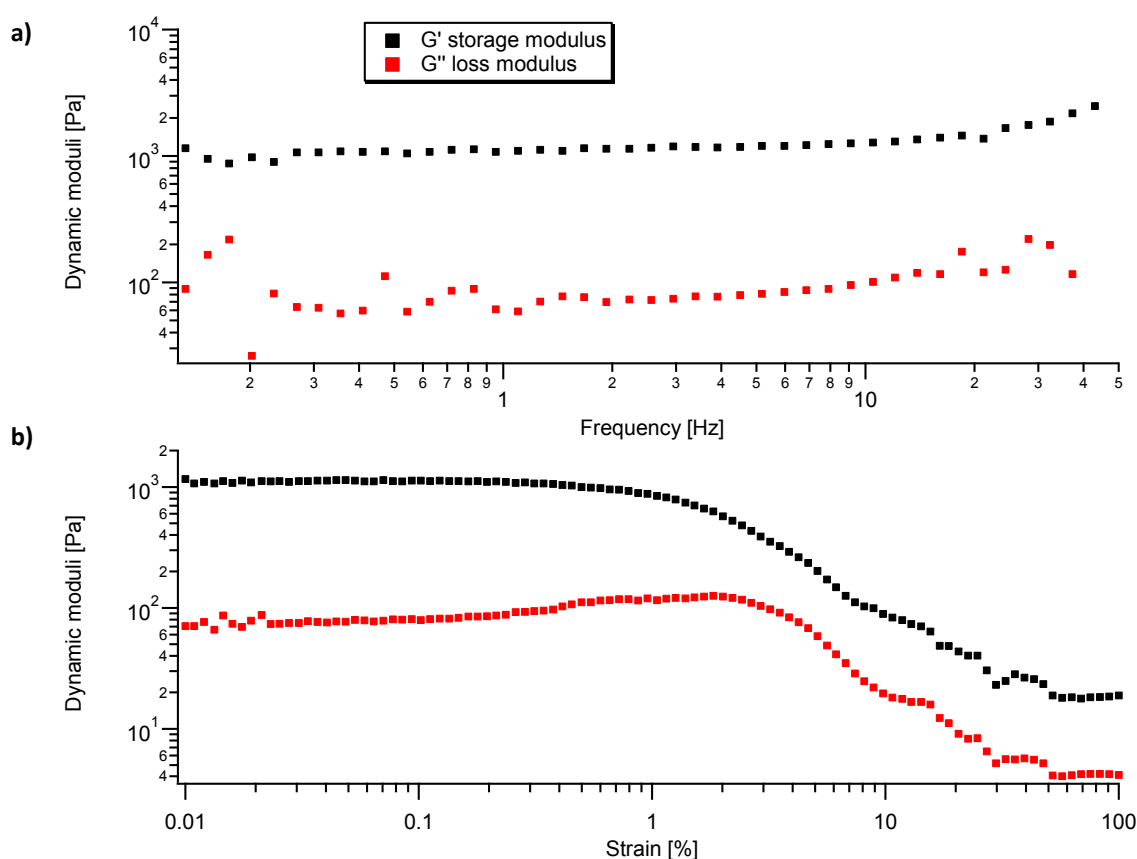


Fig. S9: Oscillatory tests of the DMSO gel of **A** ($c = 45$ mM): a) frequency sweep at a constant strain ($\gamma = 0.1$ %), b) strain sweep at a constant frequency ($f = 5$ Hz).

10. Fluorescence microscopy

Fluorescence images were taken with an Olympus BX51 microscope equipped with an Olympus DP50-CCD camera. The filter block of the microscope contained an excitation filter D360/50, a dichroic beamsplitter 400DCLP and an emission filter OG515. Therefore, the resulting fluorescence microscopy images were corrected to the color of the fluorescence light of **A** ($\lambda_{em} = 490$ nm).

11. Quantum Yield

Luminescence quantum yields were measured with a Hamamatsu Photonics absolute PL quantum yield measurement system (C9920-02) equipped with a L9799-01 CW Xenon light source (150 W), monochromator, C7473 photonic multi-channel analyser, integrating sphere, and employing U6039-05 PLQY measurement software (Hamamatsu Photonics, Ltd., Shizuoka, Japan).

12. NMR spectra

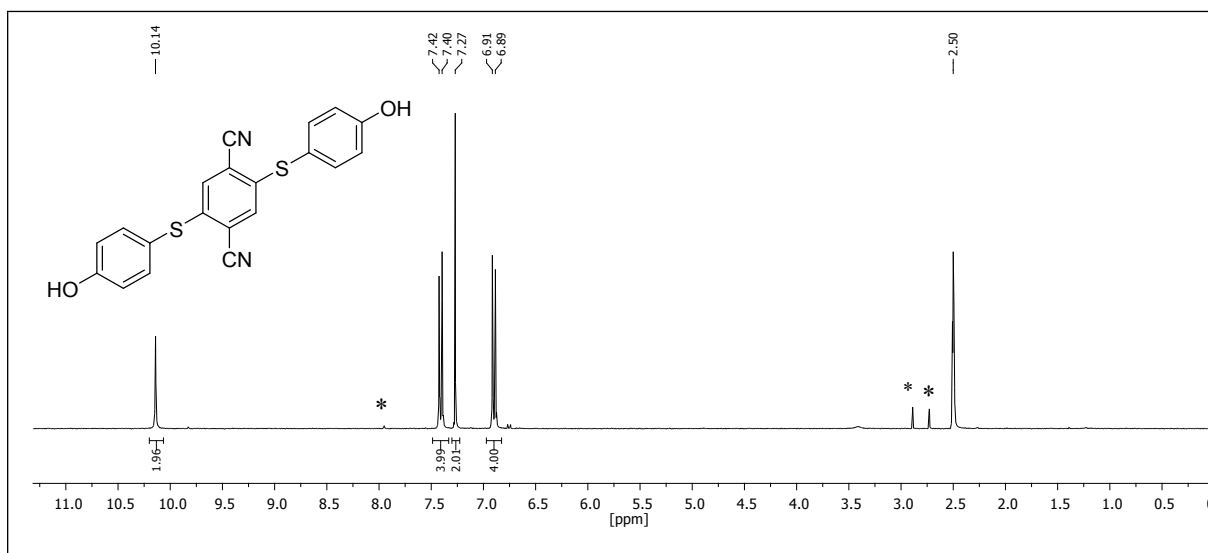


Fig. S10: ^1H NMR spectrum of **C** (300 MHz, $\text{DMSO-}d_6$), * belongs to residual DMF.

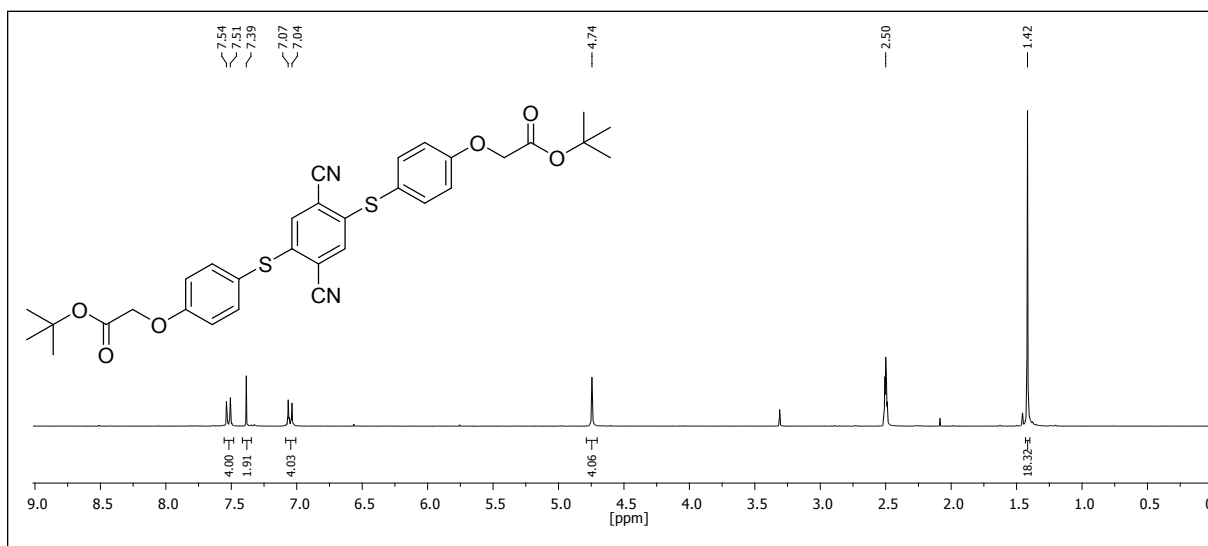


Fig. S11: ^1H NMR spectrum of **D** (300 MHz, $\text{DMSO-}d_6$).

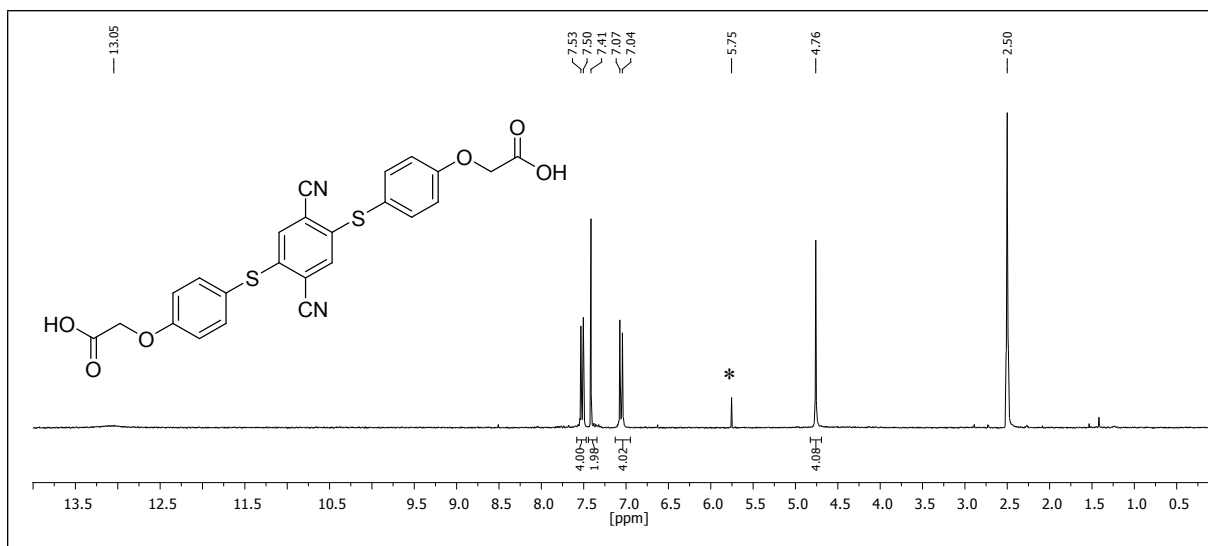


Fig. S12: ¹H NMR spectrum of **E** (300 MHz, DMSO-*d*₆), * belongs to residual DCM.

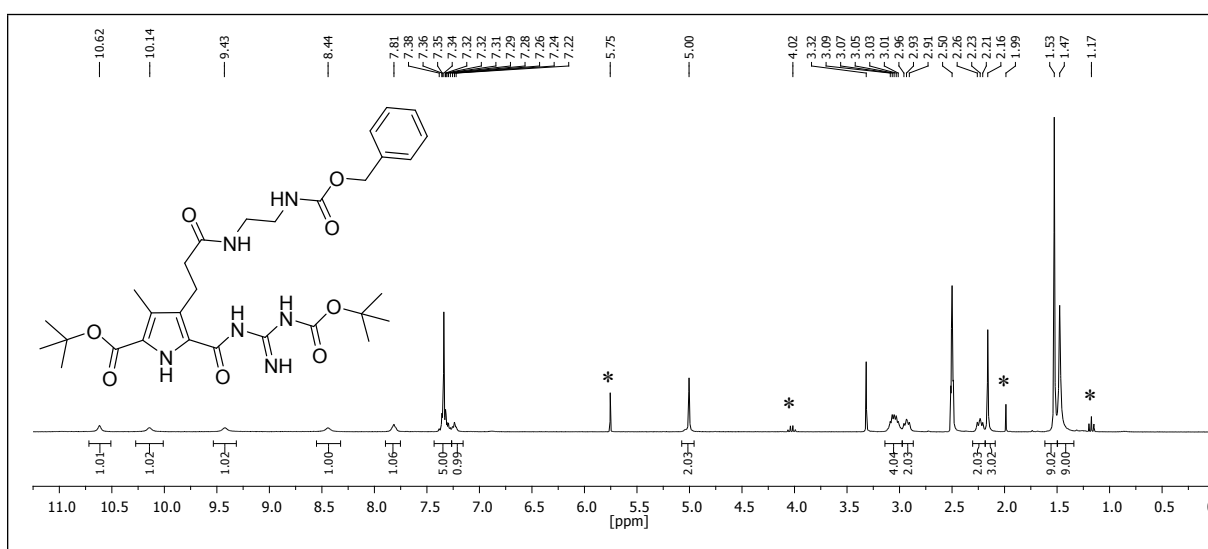


Fig. S13: ¹H NMR spectrum of **H** (300 MHz, DMSO-*d*₆), * belongs to residual EtOAc and DCM.

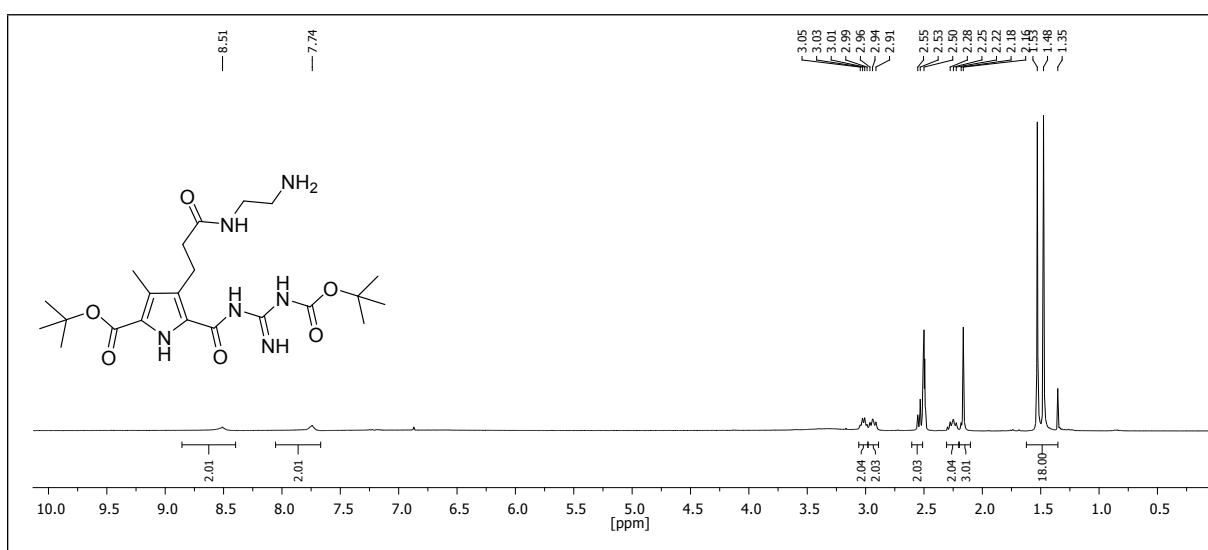


Fig. S14: ¹H NMR spectrum of **I** (300 MHz, DMSO-*d*₆).

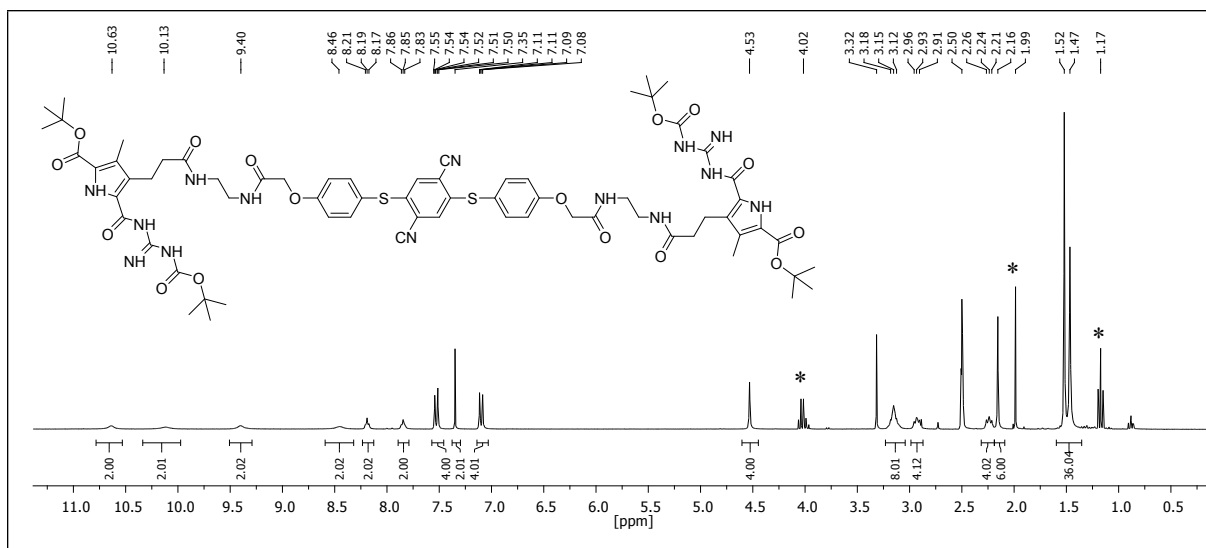


Fig. S15: ¹H NMR spectrum of J (300 MHz, DMSO-*d*₆), * belongs to residual EtOAc.

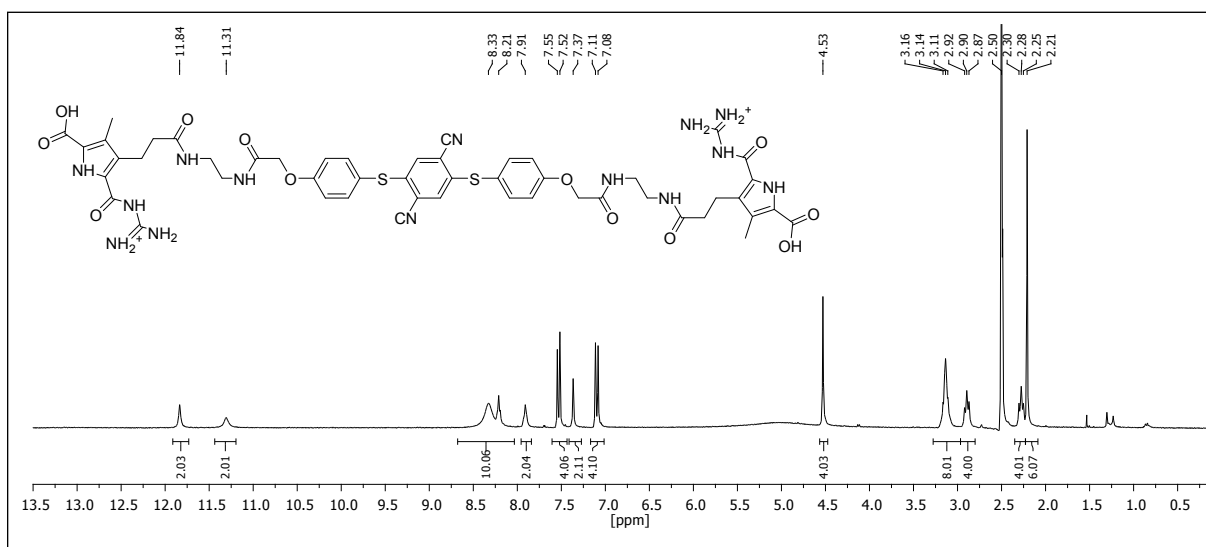


Fig. S16: ¹H NMR spectrum of A⁺ (300 MHz, DMSO-*d*₆).

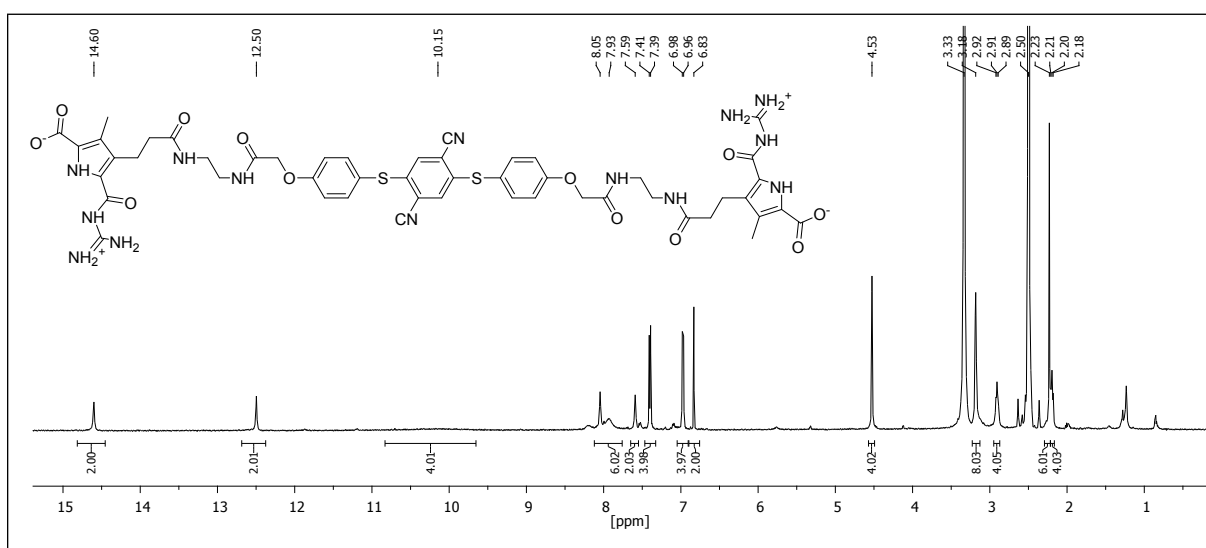


Fig. S17: ¹H NMR spectrum of A (600 MHz, DMSO-*d*₆).

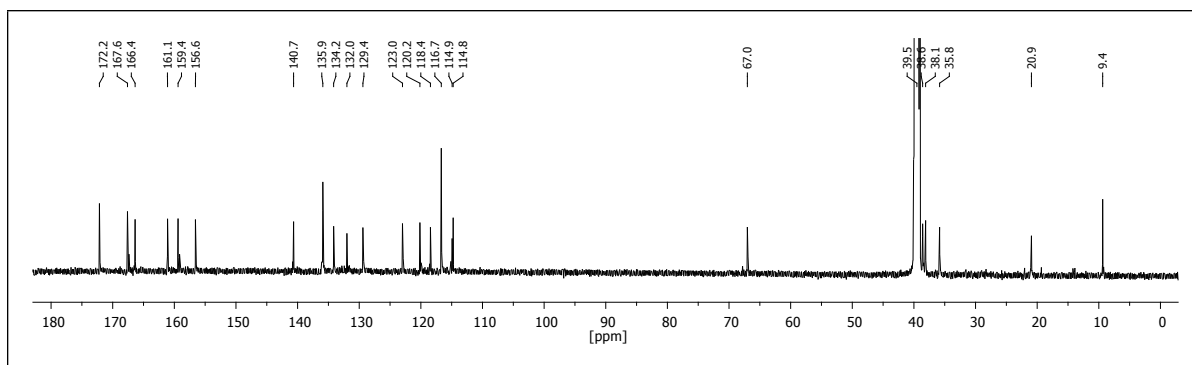


Fig. S18: ^{13}C NMR spectrum of **A** (150 MHz, $\text{DMSO-}d_6$).

13. IR-spectra

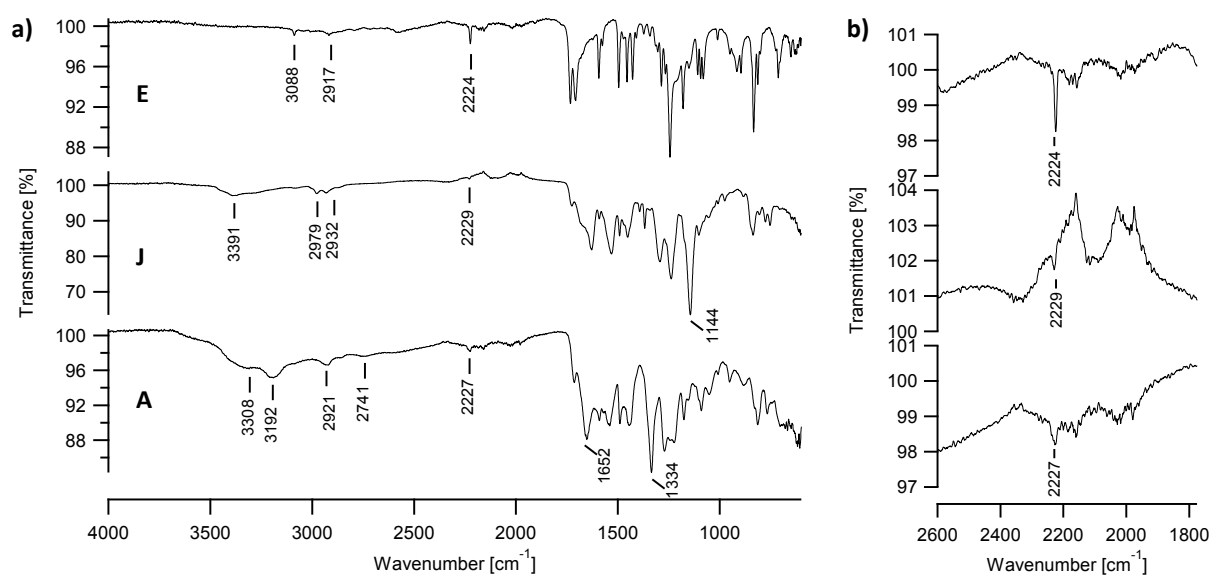


Fig. S19: Comparison of the IR spectra of **E**, **J** and **A**, (b) magnified section of the $\text{C}\equiv\text{N}$ bond stretching vibration around 2227 cm^{-1} .

14. Mass spectra

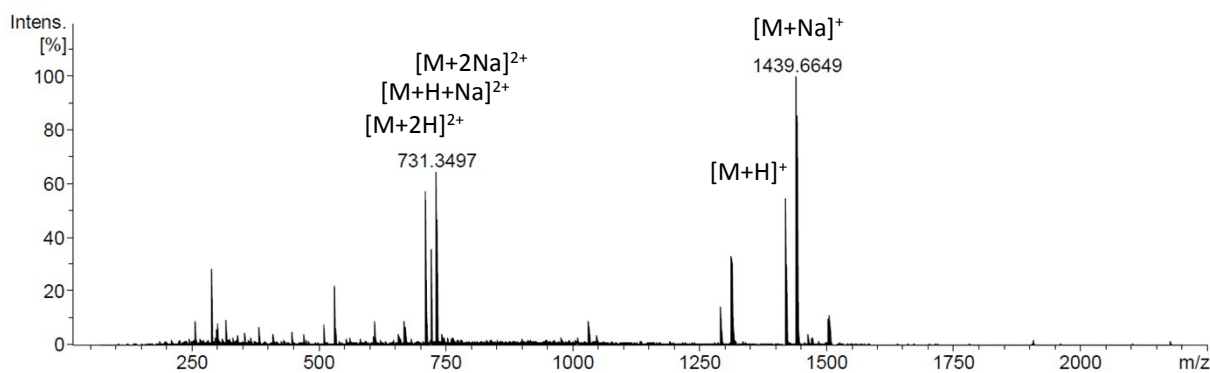


Fig. S20: LR-ESI mass spectrum of **J** (positive ion mode, MeOH).

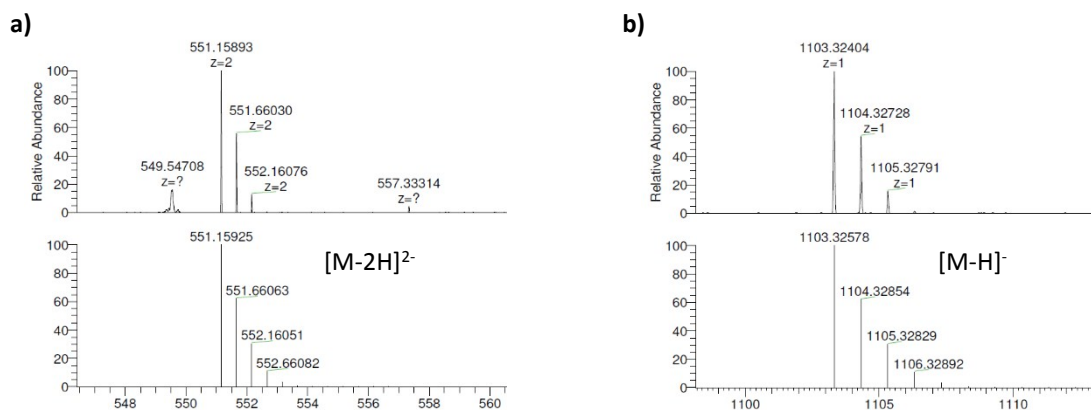


Fig. S21: HR-ESI mass spectra of **A** (negative ion mode, DMSO): comparison of the measured and the calculated mass spectra of the quasi-molecular ions a) $[M-2H]^{2-}$ and b) $[M-H]^{-}$.

15. HPLC analysis

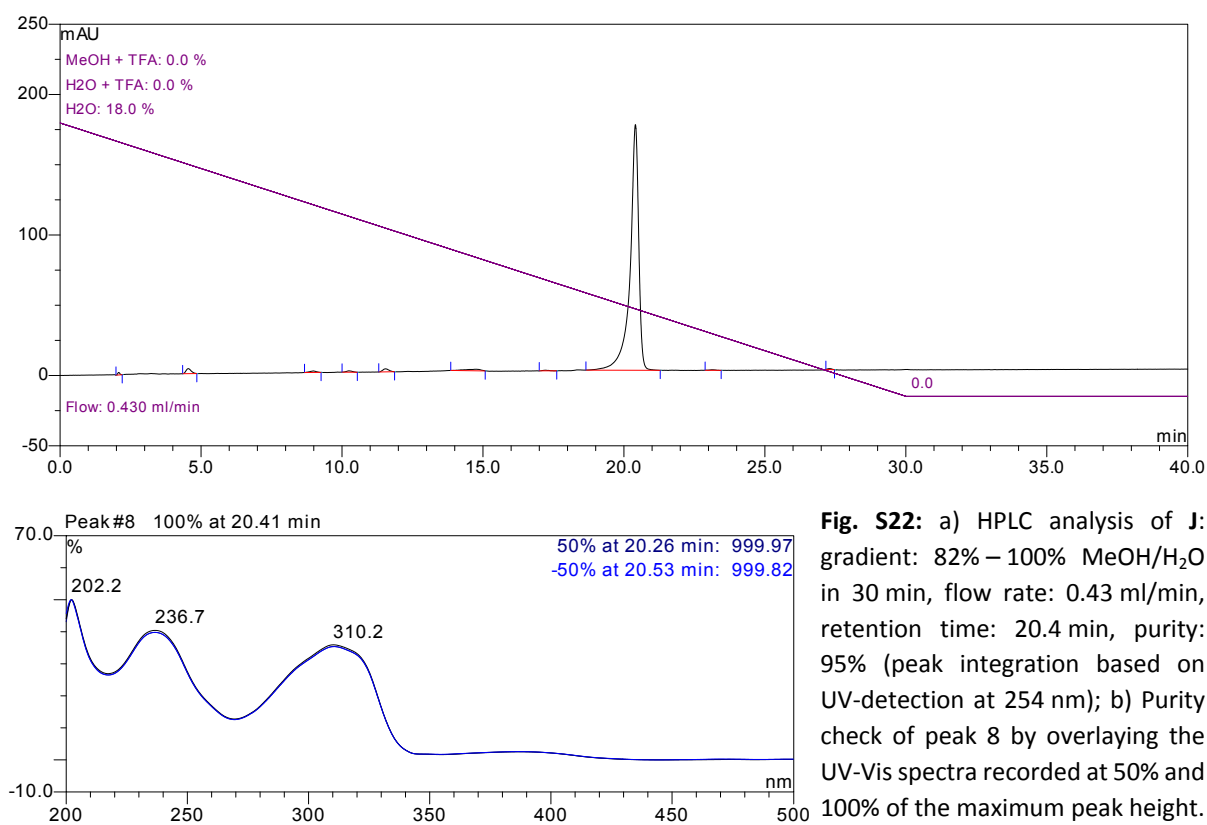


Fig. S22: a) HPLC analysis of **J**: gradient: 82% – 100% MeOH/H₂O in 30 min, flow rate: 0.43 ml/min, retention time: 20.4 min, purity: 95% (peak integration based on UV-detection at 254 nm); b) Purity check of peak 8 by overlaying the UV-Vis spectra recorded at 50% and 100% of the maximum peak height.

16. References

- [1] J-Z. Cheng, C-C. Lin, P-T. Chou, A. Chaskar, K-T. Wong, *Tetrahedron*, 2011, **67**, 734.
- [2] a) C. Schmuck, D. Rupprecht, C. Urban and N. Walden, *Synthesis*, 2006, **1**, 89; b) M. Merschky, M. Wyszogrodzka, R. Haag and C. Schmuck, *Chem. - Eur. J.*, 2010, **16**, 14242.
- [3] B. Gruber, S. Balk, S. Stadlbauer and B. König, *Angew. Chem., Int. Ed.*, 2012, **51**, 10060.
- [4] S. Riebe, C. Vallet, F. van der Vight, D. Gonzalez-Abradelo, C. Wölper, C. A. Strassert, G. Jansen, S. Knauer, J. Voskuhl, *Chem. - Eur. J.*, 2017, **23**, 13660.