Supporting Information

Oligoprolines Guide the Self-assembly of Quaterthiophenes

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1. Syntheses and analytical data of conjugates 1–5

S1.1 General materials and methods

Materials and reagents were of the highest commercial grade available, and used as received. Water used for peptide preparation and purification was Milli-Q water with resistivity of 18.2 MΩ·cm, prepared by a Sartorius Arium611VF water purification system. Solid phase peptide synthesis (**SPPS**) was performed on 200–400 mesh Rink-Amide Polystyrene AM resin from Nova-Bio-Chem. For automated peptide synthesis, a Syro I (MultiSynTech GmbH, Witten, Germany) was used.

Preparative HPLC was carried out on a Reprosil Gold column 120 C18 10 µm 150 × 16 mm. **Analytical HPLC** was performed on a Reprosil Gold column 120 C18 5 µm 150 × 4 mm. Acetonitrile (solvent A) and 0.1% TFA with 1% acetonitrile in water (solvent B) served as the mobile phases for compound purifications and analyses. Manual **gel permeation chromatography** (GPC) was performed prior to preparative recirculating GPC using Bio-Rad Bio-Beads SX1 Beads (operating range 600–14000 $gmol^{-1}$) with freshly distilled CH_2Cl_2/MeOH 9/1 as eluent. **Preparative recirculating GPC** was performed at room temperature with HPLC grade $CHCl_3$ at an elution rate of 10 mL/min through consecutive JAIGEL-2HR and JAIGEL 2.5HR columns mounted on a LC-9160 II NEXT (Japan Analytical Industry Co., Ltd.) system with coupled UV-Vis 4ch NEXT detector.

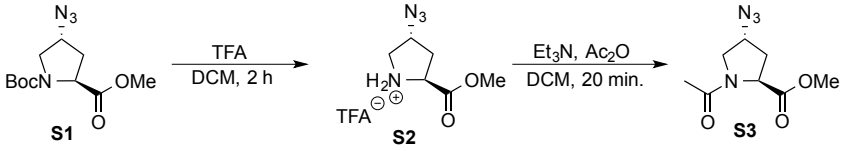
NMR spectra were recorded on a Bruker Avance III operating at 600 MHz and a Bruker Avance 400 (^{1}H NMR: 400 MHz,^{13}C NMR: 100 MHz) Chemical shifts are reported in ppm and are referenced to the solvent residual peak ($CHCl_3$ 7.26 ppm for ^{1}H, and 77.2 ppm for ^{13}C, THF-d_8 3.57 ppm and 1.71 ppm for ^{1}H, 24.4 ppm and 66.4 ppm for ^{13}C, DMSO-d_6 2.50 ppm for ^{1}H and 39.5 ppm for ^{13}C). Coupling constants are reported in Hertz (Hz). **High-resolution mass spectrometry** measurements for the oligoproline-chromophore conjugates were performed on a Bruker Daltonics SOLARIX equipped with a MALDI/ESI source and Q-TOF ion analyzer. Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) measurements for the quaterthiophene precursors were carried out on a Bruker Daltonik Reflex III mass spectrometer with the following matrices: 1,2,3-trihydroxyanthracene (dithranol), 2,5-dihydroxybenzoic acid (DHB) and 2-[3-(4-*tert*-butyl-phenyl)-2-methyl-2-propenylidene] malononitrile (DCTB).

UV-Vis spectra were recorded on an Agilent Cary 300 spectrometer. **Circular Dichroism** (CD) spectroscopic analyses were carried out with a Chirascan (Applied Biophysics Ltd, Leatherhead, UK). Quartz cuvettes with an optical path of 1.00 mm (Hellma 110-QS) or 10.00 mm (Hellma 114-QS) were used for measurements.

Transmission electron microscopy (TEM) was performed using a FEI tecnai F30 FEG instrument operating at 300 kV equipped with a 1k CCD. For sample preparation, a 200-mesh carbon coated copper grid was submitted to negative glow discharge for 40 seconds. The copper grid was then placed on 5 μL of the sample solution. After 1.5 min, the grid was lifted, excess fluid was removed with filter paper, and the grid was washed twice with water and

once with 2% uranyl acetate in water. Staining for 30 sec. with 2% uranyl acetate in water was then performed to increase the sample contrast and reveal the fine, ordered structure of the self-assembly. Longer staining times caused overstaining of specimens.

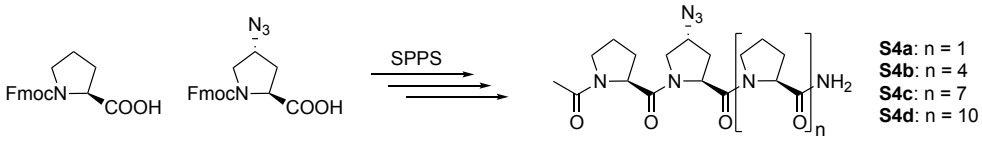
*1.2. Synthesis of acetylated azidoproline methyl ester **S3***



Scheme S1. Synthetic route toward conjugate **1**.

Boc-Azp-OMe (0.5 g, 1.8 mmol, 1 equiv.) was dissolved in 1.5 mL CH_2Cl_2 and TFA (1.5 mL, 11 equiv.) was added. After stirring the reaction mixture at room temperature for 2 hours, all volatiles were removed *in vacuo*. The resulting crude TFA-salt of H-Azp-OMe was then acetylated without further purification. H-Azp-OMe was dissolved an 10 mL of 8:1:1 CH_2Cl_2:Et_3N:Ac_2O and stirred at room temperature for 20 minutes. All volatiles were evaporated *in vacuo* and the crude product was purified using reverse phase HPLC. Acetonitrile (solvent A) and water containing 1% CH_3CN and 0.1% TFA (solvent B) were used as eluents. A flow rate of 6 mL/min, at 50°C, was used for preparative HPLC. 0.39 g (1.8 mmol, quant.) of Ac-Azp-OMe was obtained as a white solid.

1.3. Synthesis of azidoproline containing oligoprolines



Scheme S2. Synthetic route towards peptides **S4a–S4d.**

The **syntheses of peptides S4a-S4c** and amino acid Ac-(4*R*)Azp-NH_2 **S4** were performed according to protocols A–E on Rink-Amide polystyrene resin on a 102 µmol scale on a Syro I peptide synthesizer from MultiSynTech using Fmoc-Pro-OH and Fmoc-(4*R*)Azp-OH.

General protocols for Solid Phase Peptide Synthesis (SPPS)

Protocol A: Peptide couplings

Fmoc-Xaa-OH (3 equiv.) and HCTU (3 equiv.) were dissolved in DMF and added to the amino-functionalized resin. Subsequently *i*Pr_2NEt (3 equiv.) was dissolved in NMP and added to the reaction mixture. The mixture was agitated for 60 min and washed with DMF (3x).

Protocol B: Fmoc deprotections

A solution of 40% piperidine in DMF (2 mL) was added to the resin and the suspension was agitated for 5 min. The solution was removed, and the procedure was repeated with 10 min. agitation. The resin was then washed with DMF (5x).

Protocol C: N-terminal acetylation

Et_3N (30 equiv.) and Ac_2O (30 equiv.) were added to the amino-functionalized resin-bound oligoproline suspended in CH_2Cl_2. The mixture was agitated for 10 min and washed with CH_2Cl_2 (5x).

Protocol D: Cleavage from the solid support

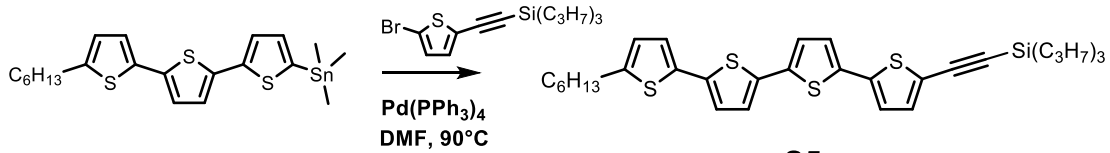
The resin was agitated for 1 h in a mixture of TFA/H_2O/DCM/TIPS (87.5:5:5:2.5) and the solution was collected by filtration. The filtrate was concentrated to a small volume under reduced pressure and the product was precipitated from cold Et_2O. The white solid was isolated by centrifugation of the suspension and decanting the supernatant. The solid was triturated with Et_2O twice and the residual white solid was dried under a stream of nitrogen, dissolved in H_2O/CH_3CN 1:1 and lyophilized to obtain the desired peptide as a white solid.

Protocol E: HPLC purifications

The crude product was purified using reverse phase HPLC. Acetonitrile (CH_3CN, solvent A) and water containing 1% CH_3CN and 0.1% TFA (solvent B) were used as eluents. A flow rate of 6 mL/min, at 50°C, was used for preparative HPLC. UV-Vis monitoring was carried out at 214 nm. After purification, pure fractions were combined and all volatiles were removed by lyophilisation.

1.4. Synthesis of 5-([triisopropylsilyl]ethynyl)-5'''-hexyl-2,2':5',2'':5'',2'''-quaterthiophene ***S5***

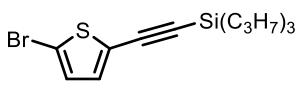
Literature known 2-bromo-5-([triisopropylsilyl]ethynyl)thiophene[1] was synthesized following a protocol for the synthesis of the 2-bromo-5-([trimethyllsilyl]ethynyl)thiophene.[3] 5''-Hexyl-[2,2':5',2''-terthien]-5-yl)trimethylstannane was prepared similarly as described in literature.[4]



S5

Scheme S3. Synthetic route towards quaterthiophene **S5**.

Synthesis of 2-bromo-5-([triisopropylsilyl]ethynyl)thiophene:



2,5-Dibromothiophene (1.0 mL, 8.87 mmol) was dissolved in 60 mL of carefully degassed diisopropylamine (DIPA). CuI (45 mg, 0.24 mmol) and bis(triphenylphosphine) palladium(II)

dichloride (150 mg, 0.21 mmol) were added. To the mixture, triisopropyl (TIPS)-acetylene (2.0 mL, 8.87 mmol) was added dropwise over 2 h. The solution was allowed to stir for 20 h at room temperature under argon atmosphere. After removal of the solvent under reduced pressure the residue was redispersed in hexane. Subsequently, the organic layer was washed with water and dried over $MgSO_4$.The solvent was removed in vacuo and the crude product was purified by column chromatography (silica, eluent: PE) to afford the TIPS-protected ethynylated thiophene as a colourless oil in 20 % yield (0.6 g, 1.75 mmol). The analytical data correspond to the literature values.[1]

^{1}H-NMR (400 MHz, $CDCl_3$): δ = 6.96 (d, 1H, *J* = 3.88 Hz), 6.90 (d, 1H, *J* = 3.88 Hz), 1.12-1.10 (m, 21H) ppm.^{13}C-NMR (100 MHz, $CDCl_3$): δ = 132.8, 129.9, 125.5, 112.8, 98.4, 97.1, 18.8, 11.4 ppm.

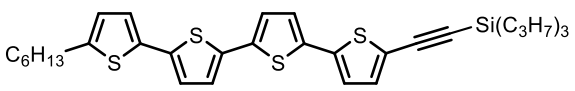
Synthesis of 5''-hexyl-[2,2':5',2''-terthien]-5-yl) trimethylstannane

n-BuLi (3.60 mL, 5.76 mmol, 1.6 M in *n*-hexane) was added dropwise to a solution of 5-hexyl-2,2′;5′,2″-terthiophene[4] (760 mg, 2.29 mmol) in anhydrous THF (30 mL) at -78°C under argon. The solution was stirred 1 h at the same temperature. Trimethyltin chloride (1.15 g, 5.76 mmol) dissolved in THF (2 mL) was added and the solution was stirred for 4 h at -78°C. The solvent was removed under reduced pressure and the residue was dissolved in H_2O (40 mL) and extracted with DCM (3x40 mL). The combined organic layers were washed with H_2O (2x40 mL) and dried over Na_2SO_4 to give the stannane (1.02g, 90%) as a yellow solid.

1H-NMR ($CDCl_3$, 400 MHz): *δ* = 7.26 (d, *J* = 2.8 Hz, 1H, H_4), 7.09 (d, *J* = 3.4 Hz, 1H, thio-H), 7.05 (d, *J* = 3.8 Hz, 1H, thio-H), 6.99 (d, *J* = 3.8 Hz, 1H, thio-H), 6.97 (d, *J* = 3.5 Hz, 1H, thio-H), 6.68 (d, *J* = 3.6 Hz, 1H, $H_{4''}$), 2.79 (t, *J* = 7.6 Hz, 2H, H_α), 1.68 (quintett, *J* = 7.6 Hz, 2H, H_β), 1.31 (m, 6H, $H_{\gamma\text{-}\varepsilon}$), 0.90 (t, *J* = 7.2 Hz, 3H, H_ζ), 0.39 (s, 9H, -$SnMe_3$) ppm.

^{13}C-NMR ($CDCl_3$, 100 MHz): *δ* = 145.62, 142.90, 137.59, 136.70, 136.05, 135.78, 134.74, 124.94, 124.86, 124.22, 123.68, 123.36, 77.48, 77.16, 76.84, 68.13, 53.58, 31.71, 30.33, 28.90, 22.72, 14.24, -8.06 ppm.

Synthesis of 5-([Triisopropylsilyl]ethynyl)-5'''-Hexyl-2,2':5',2'':5'',2'''-quaterthiophene ***S5***



S5

(5''-Hexyl-2,2':5',2''-terthien-5-yl)trimethyl stannane (700 mg, 1.41 mmol), 2-bromo-5-([triisopropylsilyl]ethynyl)-thiophene (485 mg, 1.41 mmol) and tetrakis(triphenylphosphine) palladium (81.6 mg, 0.07 mmol) were dissolved in 10 mL of DMF and stirred at 90°C under argon for 2 h. After cooling, 20 ml water and 20 ml dichloromethane were added and the layers were separated. The organic layer was dried over $MgSO_4$, filtered and the solvent was

removed. The crude product was purified by column chromatography (silica, eluent: hexanes) and 5-([triisopropylsilyl]ethynyl)-5'''-hexyl-2,2':5',2'':5'',2'''-quaterthiophene **S5** was obtained as an orange solid in 62 % yield (525 mg, 0.88 mmol).

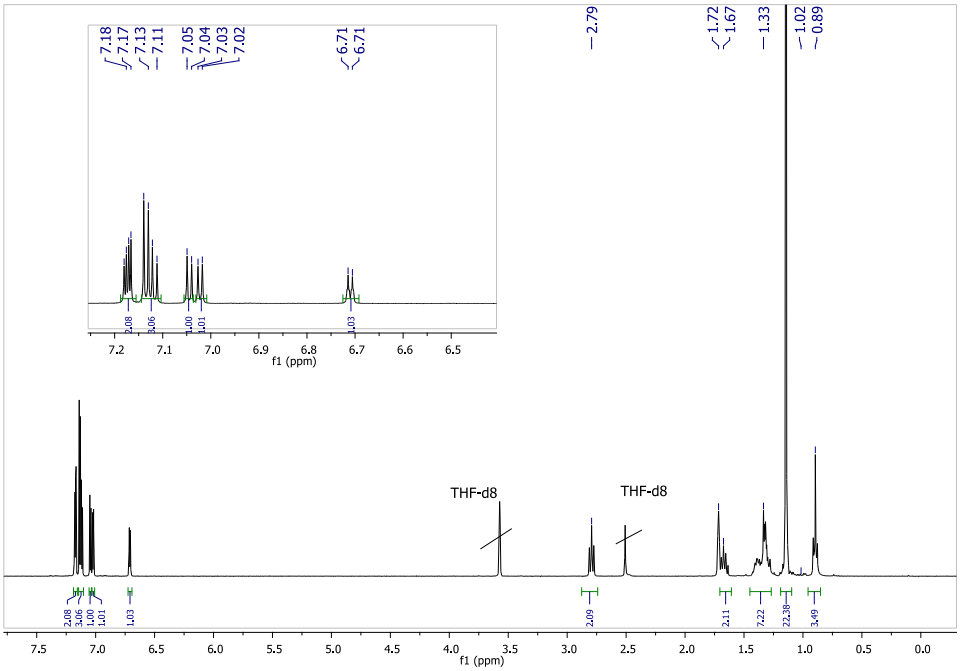
^{1}H-NMR (400 MHz, CDCl3): δ = 7.12 (d, J = 3.82 Hz, 1H), 7.08-7.03 (m, 3H), 7.00 (d, J = 3.71 Hz, 2H), 6.99 (d, J = 3.60 Hz, 1H), 6.69 (dt, J = 3.60 Hz, J = 0.89 Hz, 1H), 2.79 (t, J = 7.64 Hz, 2H), 1.68 (pseudo-quint, J = 7.52 Hz, 2H), 1.45-1.25 (m, 6H), 1.15-1.10 (m, 21H), 0.89 (t, J = 6.89 Hz, 3H) ppm.

^{1}H-NMR (400 MHz, THF-d_8): δ = 7.18 (d, J = 3.83 Hz, 1H), 7.17 (d, J = 3.85 Hz, 1H), 7.12 (m, 3H, J = 3.83 Hz), 7.04 (d, J = 3.80 Hz, 1H), 7.02 (d, J = 3.56 Hz, 1H), 6.71 (dt, J = 3.59 Hz, J = 0.89 Hz, 1H), 2.79 (t, J = 7.64 Hz, 2H), 1.67 (m, 2H), 1.45-1.25 (m, 6H), 1.15-1.10 (m, 21H), 0.89 (m, 3H) ppm.

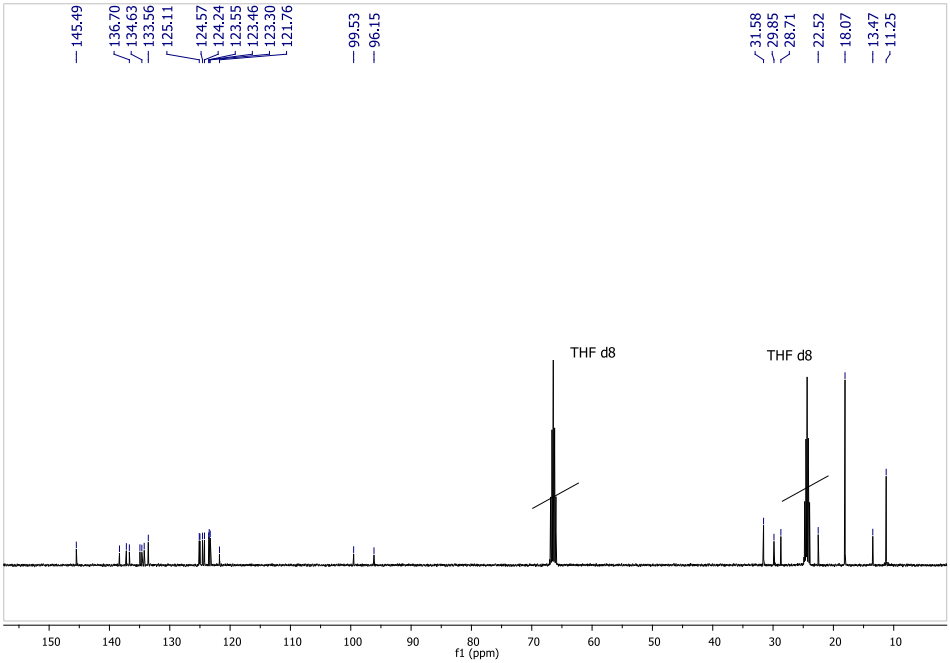
^{13}C-NMR (100 MHz, THF-d_8):145.6, 138.3, 137.2, 136.7, 134.9, 134.6, 134.2, 133.6, 125.1, 125.0, 124.6, 124.2, 123.6, 123.5, 123.3, 121.8, 99.5, 96.2, 31.6, 29.9, 28.7, 22.5, 18.1, 13.5, 11.3.

HRMS (MALDI) *m/z*: calculated $C_{33}H_{42}S_4Si$ $[M+H]^+$: 594.19386; found: 594.19392.

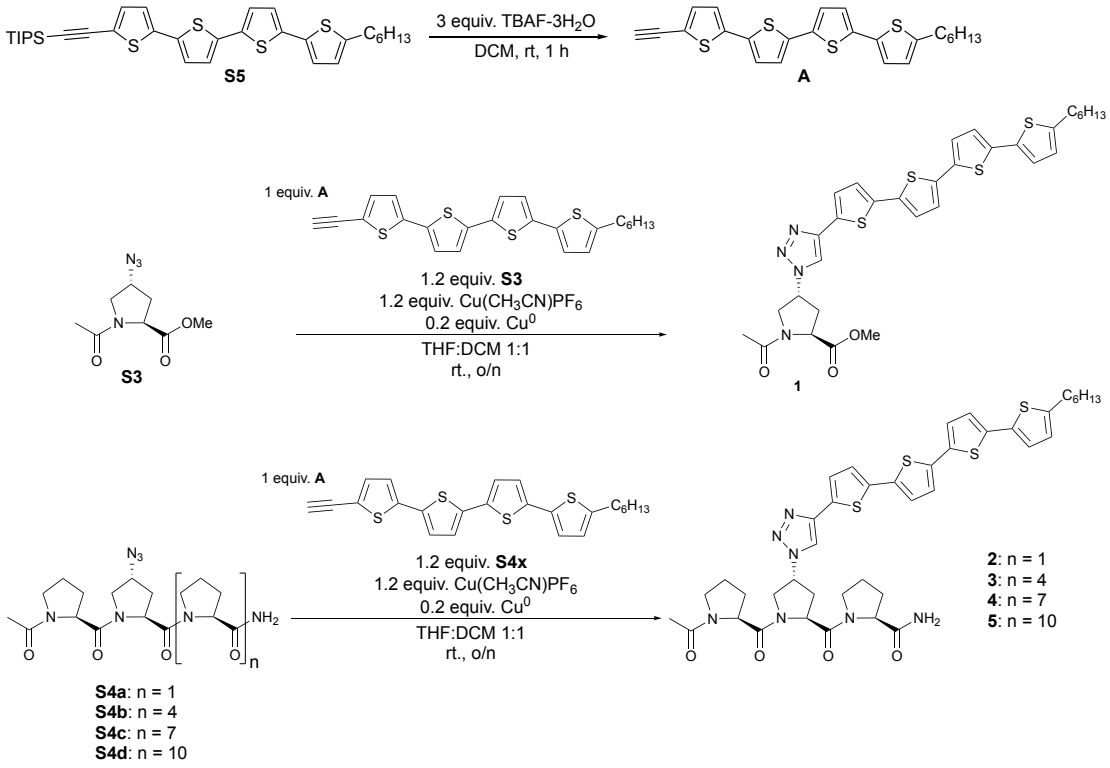
^{1}H-NMR spectrum (400 MHz, THF-d_8):



^{13}C-NMR spectrum (100 MHz, THF-d_8):



*1.5. Synthesis of oligoproline-chromophore conjugates **1–5***



Scheme S4. Synthetic route towards conjugates **1–5**.

General Methods

Protocol F: TIPS deprotection

TIPS deprotection was performed immediately before coupling the chromophore to the azide-functionalized peptides. TIPS protected hexyl quaterthiophene **S5** (1 equiv., 6.4 mg) was dissolved in CH_2Cl_2 (2 mL, 5.4 mM). TBAF-$3H_2O$ (3 equiv., 10.2 mg) was added and the reaction mixture was stirred at room temperature for 1 h. The reaction mixture was extracted with H_2O (3x). The organic solvent was then evaporated *in vacuo*. The resulting crude product **A** was used without further purification.

Protocol G: Azide-alkyne 'Click' reaction

Hexyl quaterthiophene alkyne (1 equiv.) and azidoproline functionalized mono/oligoproline (1.2 equiv., 7 mg) were added to a solution of CH_2Cl_2:THF 1:1 (2 mL, 3.5 mg/mL peptide). The reaction mixture was degassed with argon for 30 min. $Cu(CH_3CN)PF_6$ (1.2 equiv.) and Cu^0 powder (0.2 equiv.) were added and the reaction mixture was degassed for an additional 2 minutes. The reaction mixture was stirred overnight under inert atmosphere.

Protocol H: Flash column chromatography purification

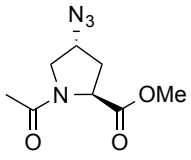
The crude products of the conjugates **1** and **2** were purified on silica (*Sigma Aldrich*, pore size 60 Å, 230 – 400 mesh particle size) using flash column chromatography. A gradient of 0%–10% MeOH in CH_2Cl_2 was used for conjugates **1** and **2** and CH_2Cl_2 for conjugate **1a** for purification. Conjugate **1a** was further purified using GPC. After purification, pure fractions were combined and all volatiles were removed *in vacuo*. The compounds were then dissolved in THF/H_2O and all volatiles were removed by lyophilisation.

Protocol I: GPC purification

The crude products of the conjugates **1a**, 6mer **3,** 9mer **4**, and 12mer **5**, were purified with manual gel permeation chromatography with freshly distilled CH_2Cl_2/MeOH 9/1 as eluent. The volatiles were removed *in vacuo*, and the products were re-purified with preparatory GPC using 0.5% Et_3N in $CHCl_3$ as eluent. The volatiles were removed from the purified products *in vacuo*. The conjugates were then dissolved in THF/H_2O and all volatiles were removed by lyophilisation.

1.6. Analytical data for azidoproline containing peptides

Analytical data for S3 (see also, Sonntag, *et al*. *J. Am. Chem. Soc.* **2006**, *128*, 14697)



HRMS (MALDI) *m/z*: calculated $C_8H_{13}N_4O_3$ $[M+H]^+$: 213.0982; found: 213.0982.

^1H NMR and ^{13}C NMR spectra show a double set of peaks due to *cis* and *trans* conformers around the tertiary amide in a ratio of 3.8:1 in d6-DMSO.

^1H NMR (600 MHz, d6-DMSO, *major conformer*) δ *ppm* = 4.48-4.42 (m, 1H, Hγ), 4.28 (t, *J* = 7.7 Hz, 1H, Hα), 3.77 (dd, *J* = 11.2, 5.3 Hz, 1H, Hδ), 3.62 (s, 3H, OCH_3, Hδ), 3.55 (ddd, *J* =

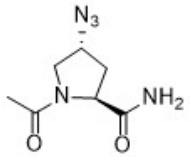
11.2, 3.3, 1.3 Hz, 1H, Hδ), 2.29 (dddd, *J* = 13.5, 8.2, 4.2, 1.3 Hz, 1H, Hβ), 2.13 (ddd, *J* = 13.5, 7.3, 5.5 Hz, 1H, Hβ), 1.98 (s, 3H, CH_3).

^{1}H NMR (600 MHz, d6-DMSO, *minor conformer*) d *ppm* = 4.72 (dd, J = 8.33, 6.2 Hz, 1H, Hα), 4.37-4.32 (m, 1H, Hγ), 3.71 (s, 3H, OCH_3, Hδ), 3.57 (dd, J = 12.4, 3.6, 1.4 Hz, 1H, Hδ), 3.45 (dd, *J* = 12.4, 5.7 Hz, 1H, Hδ), 2.42-2.32 (m, 2H, Hβ), 1.85 (s, 3H, CH_3).

^{13}C NMR (600 MHz, d6-DMSO, *major conformer*) δ *ppm* = 171.7, 168.4, 59.3, 56.7, 52.2, 51.8, 34.2, 21.8.

^{13}C NMR (600 MHz, d6-DMSO, *minor conformer*) δ *ppm* = 172.0, 168.8, 57.7, 57.6, 52.47, 50.3, 35.9, 21.3.

Analytical data for S4



HRMS (ESI) *m/z*: calculated $C_7H_{11}N_5NaO_2$ $[M+Na]^+$: 220.0805; found: 220.0805.

HPLC: R_t = 4.47 min; gradient 98% to 75% B in 20 min at 25 °C. Flow 1.0 mL min^{-1}.

^{1}H NMR and ^{13}C NMR spectra show a double set of peaks due to *cis* and *trans* conformers around the tertiary amide in a ratio of 14:1 in $CDCl_3$.

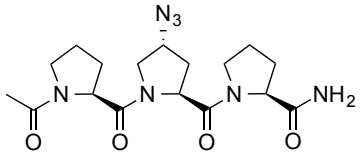
^{1}H NMR (400 MHz, $CDCl_3$, *major conformer*) δ *ppm* = 4.68 (dd, *J* = 8.2, 4.1 Hz, 1H, Hα), 4.40 – 4.33 (m, 1H, Hγ), 3.74 (dd, *J* = 10.7, 6.3 Hz, 1H, Hδ), 3.41 (dd, *J* = 10.7, 5.4 Hz, 1H, Hδ), 2.71 (ddd, *J* = 13.2, 6.1, 4.1 Hz, 1H, Hβ), 2.11 (s, 3H, CH_3), 2.08 – 1.99 (m, 1H, Hβ).

^{1}H NMR (400 MHz, $CDCl_3$, *minor conformer*) δ *ppm* = 4.38 (m, 1H, Hα), 4.21 (m, 1H, Hγ), 3.96 – 3.88 (m, 1H, Hδ), 3.65 (dd, *J* = 12.6, 5.4 Hz, 1H, Hδ), 2.56 – 2.47 (m, 1H, Hβ), 2.35 (m, 1H, Hβ), 2.11 (s, 3H, CH_3).

^{13}C NMR (400 MHz, $CDCl_3$, *major conformer*) δ *ppm* = 172.5, 170.9, 59.5, 57.9, 52.8, 32.9, 22.6.

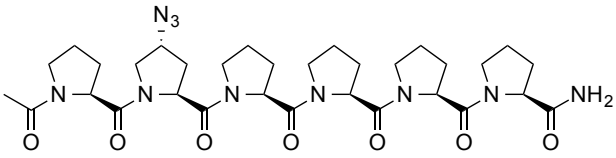
^{13}C NMR (400 MHz, $CDCl_3$, *minor conformer*) δ *ppm* = 172.5, 170.9, 60.3, 58.3, 51.6, 33.0, 21.9.

Analytical data for S4a



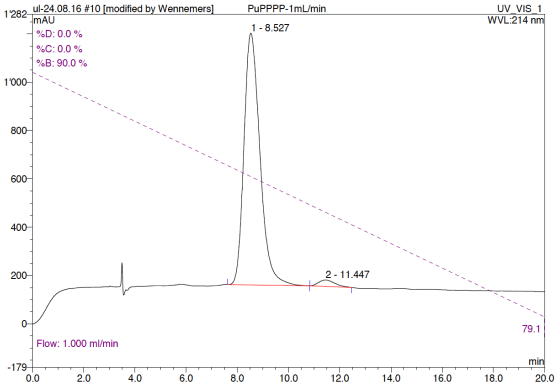
Analytical data matches previously published work.[2]

Analytical data for S4b

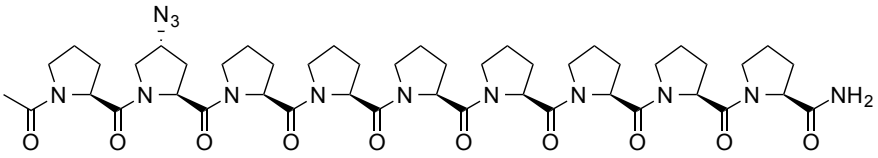


HRMS (ESI) *m/z*: calculated $C_{32}H_{47}N_{10}O_7$ $[M+H]^+$: 683.3624; found: 638.3612.

HPLC: R_t = 8.53 min; gradient 90% to 80% B in 20 min at 25 °C. Flow 1.0 mL min^{-1}.

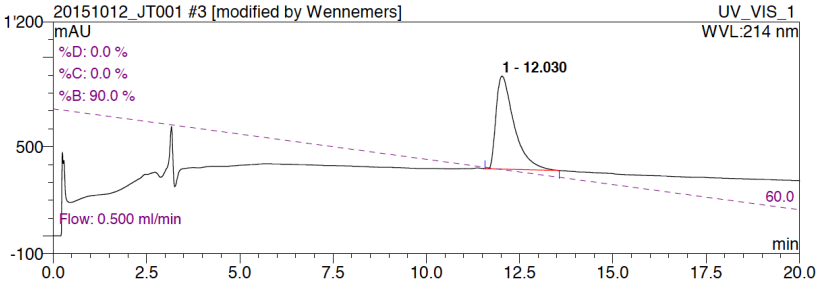


Analytical data for S4c

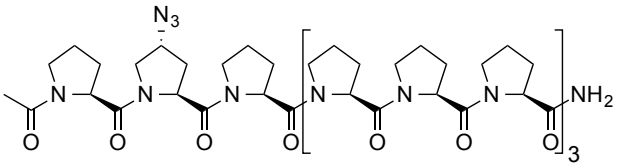


HRMS (ESI) *m/z*: calculated $C_{47}H_{68}N_{13}O_{10}$ $[M+H]^+$: 974.5207; found: 974.5198.

HPLC: R_t = 12.03 min; gradient 90% to 60% B in 20 min at 25 °C. Flow 0.5 mL min^{-1}.

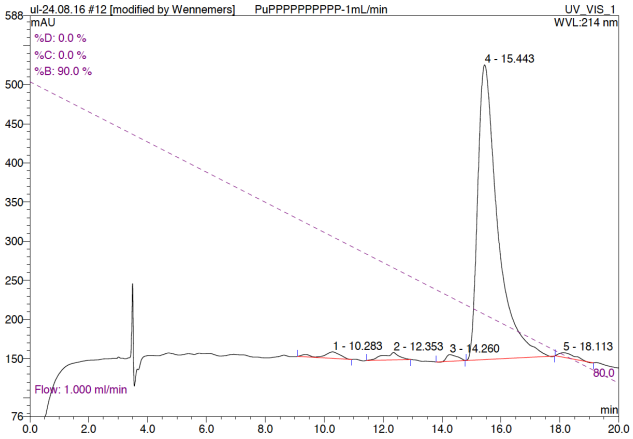


Analytical data for S4d



HRMS (ESI) *m/z*: calculated $C_{62}H_{89}N_{16}O_{13}$ $[M+H]^+$: 1265.6790; found: 1265.6793.

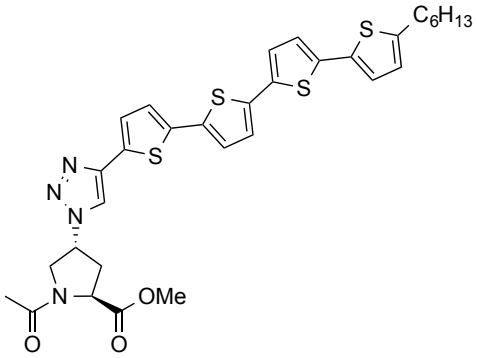
HPLC: R_t = 15.44 min; gradient 90% to 80% B in 20 min at 25 °C. Flow 1.0 mL min^{-1}.



*1.7. Analytical data for peptide-chromophore conjugates **1**–**5***

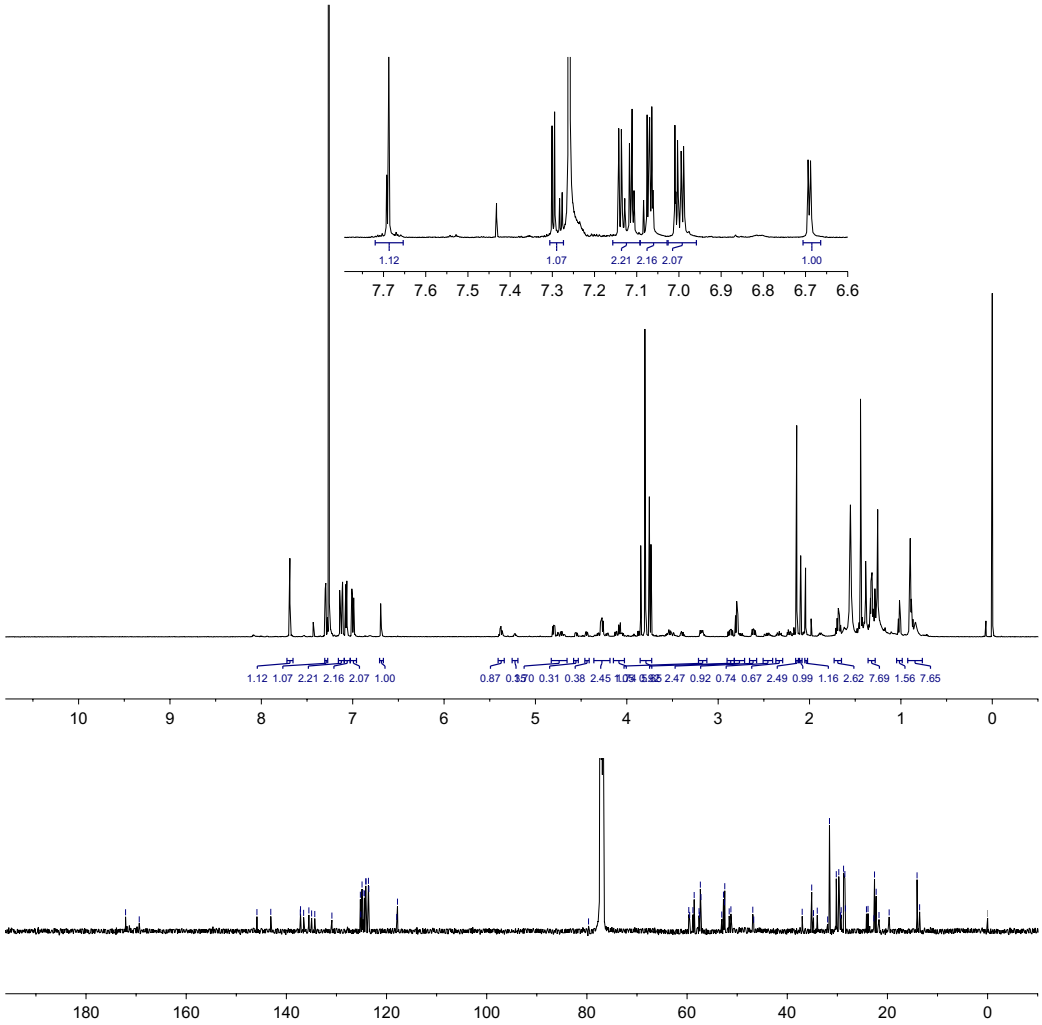
Note, the NMR spectra show a double set of peaks due to *cis* and *trans* conformers around the tertiary amide(s). The solubility of the conjugates is generally low in essentially any solvent at concentrations necessary to record NMR spectra.

Analytical data for conjugate 1

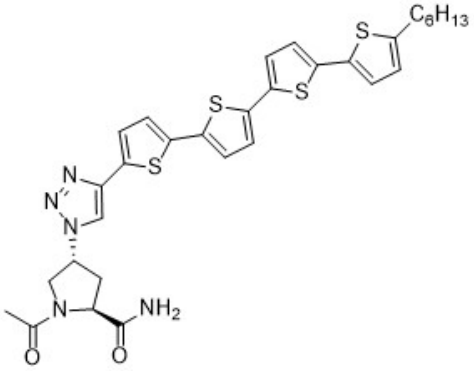


HRMS (MALDI-TOF) *m/z*: calculated $C_{32}H_{34}N_4O_3S_4$ $[M+H]^+$: 650.1508; found: 650.1507

^{1}H NMR and ^{13}C NMR spectra (600 MHz, $CDCl_3$, 297K)



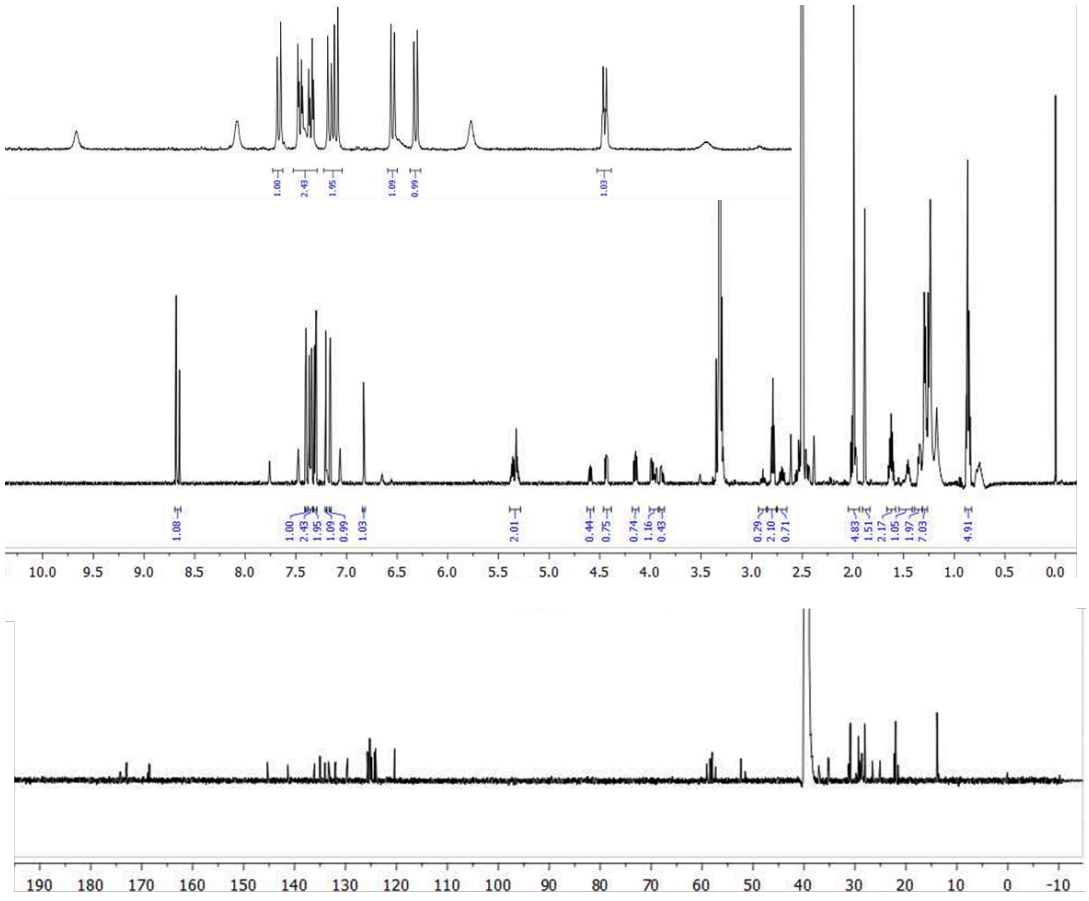
Analytical data for conjugate 1a



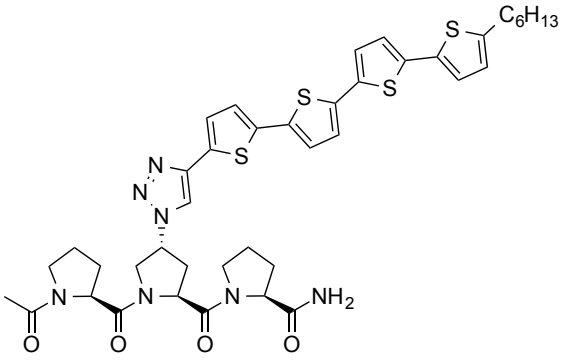
HRMS (MALDI-TOF) *m/z*: calculated $C_{31}H_{33}N_5O_2S_4$ $[M+H]^+$: 635.1512; found: 635.1513.

^{1}H NMR and ^{13}C NMR spectra (600 MHz, DMSO-d$_6$, 297K)

(Note, the solubility of this conjugate in solvents other than DMSO is too low to record NMR spectra. The solubility is even low in DMSO, the ^{13}C spectrum shown below was acquired using 20000 pulses.)

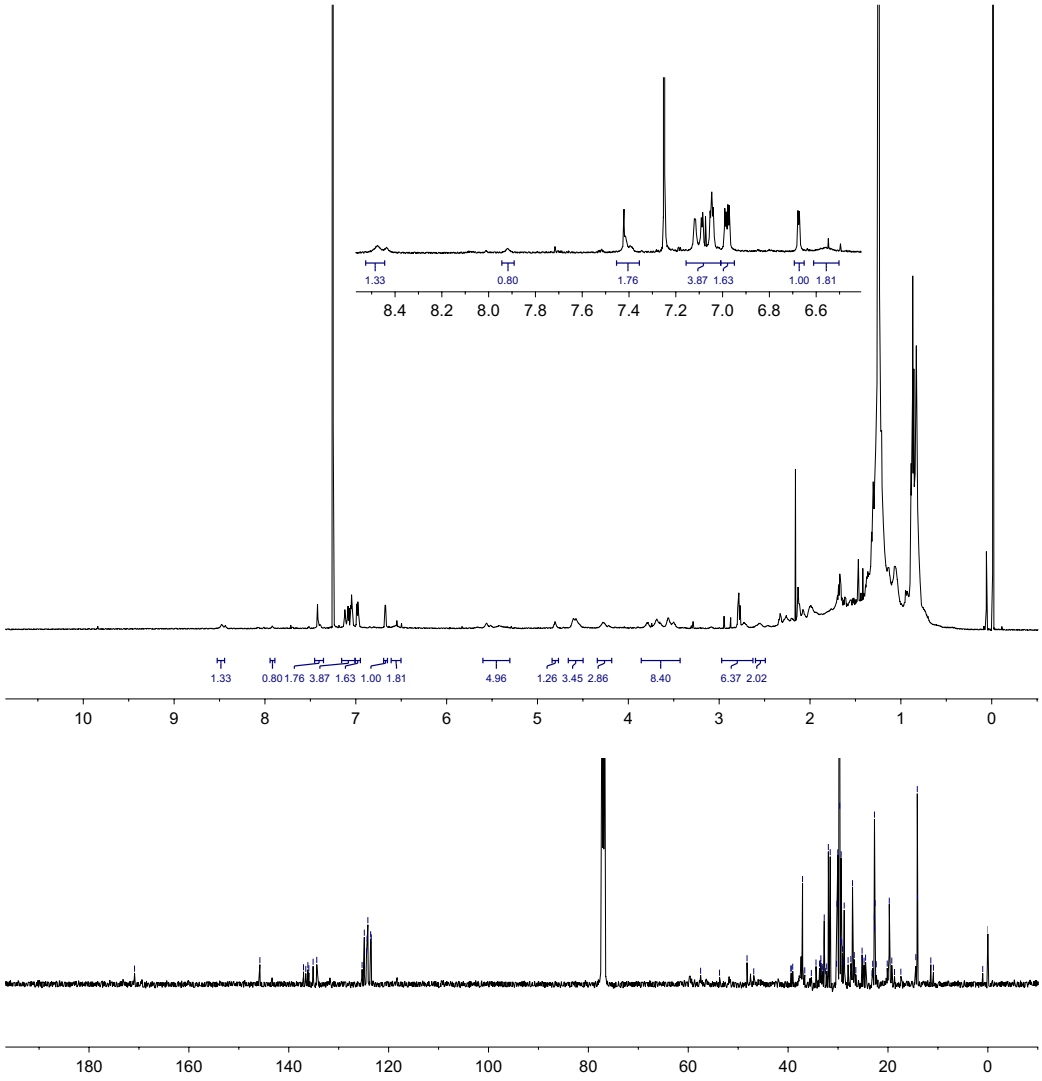


Analytical data for conjugate 2

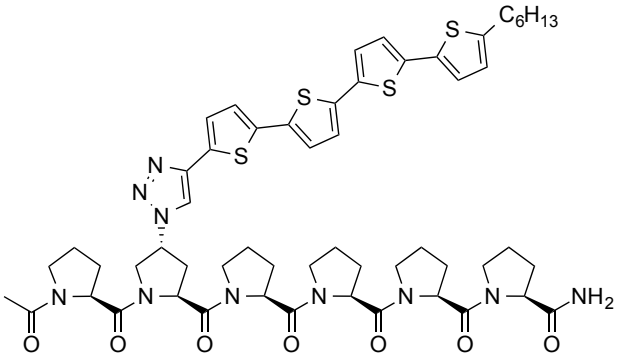


HRMS (MALDI-TOF) *m/z*: calculated $C_{41}H_{47}N_7O_4S_4$ $[M+H]^+$: 829.2567; found: 829.2565

^{1}H NMR and ^{13}C NMR spectra (600 MHz, $CDCl_3$, 297K)

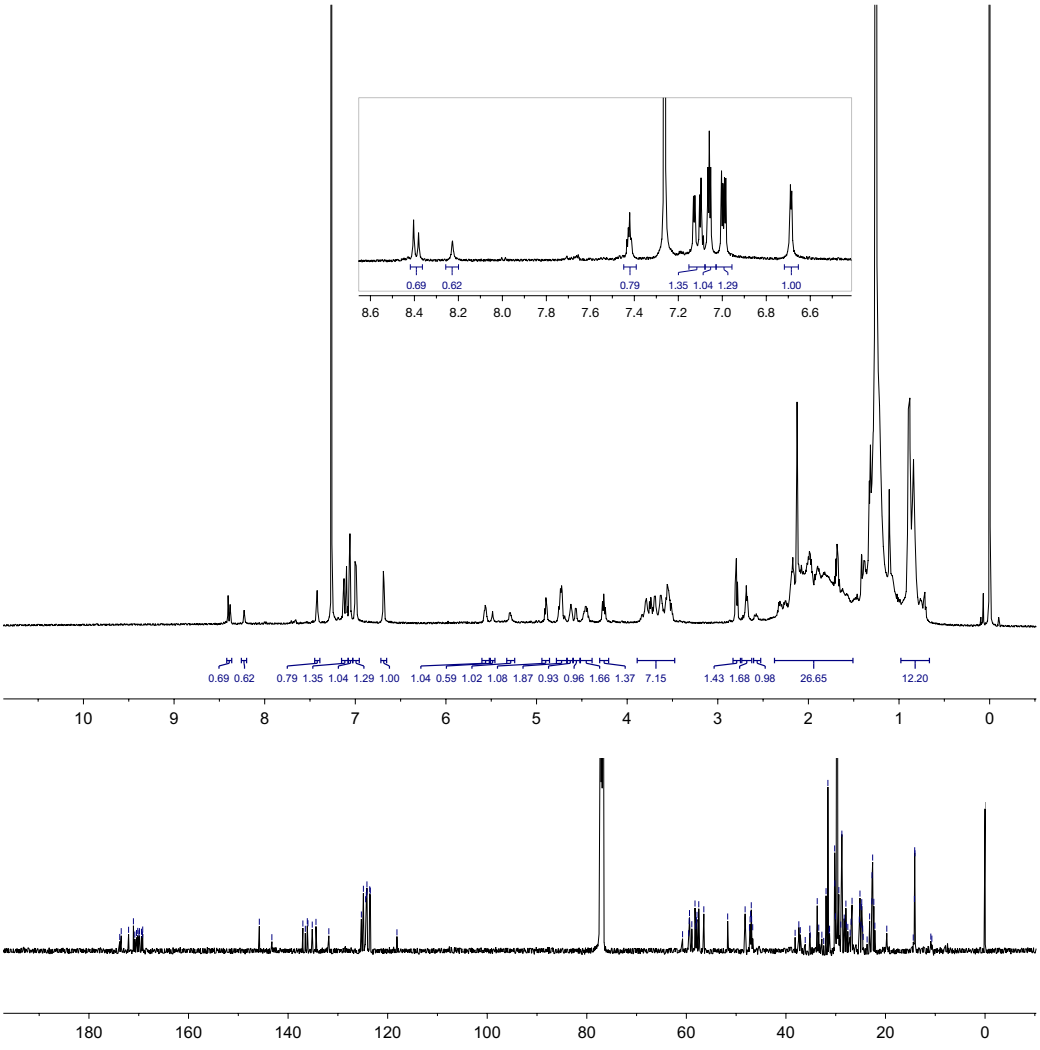


Analytical data for conjugate 3

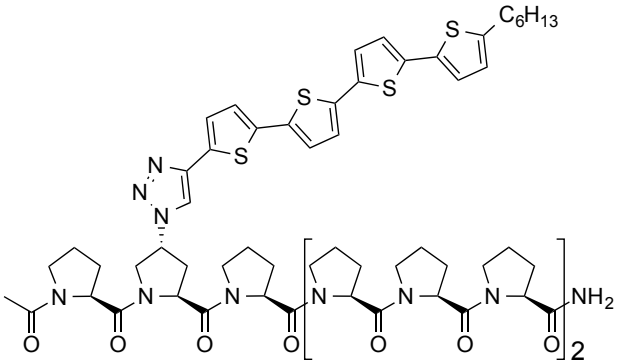


HRMS (MALDI-TOF) *m/z*: calculated $C_{56}H_{68}N_{10}O_7S_4$ $[M+H]^+$: 1120.4150; found: 1120.4128.

^{1}H NMR and ^{13}C NMR spectra (600 MHz, $CDCl_3$, 297K)

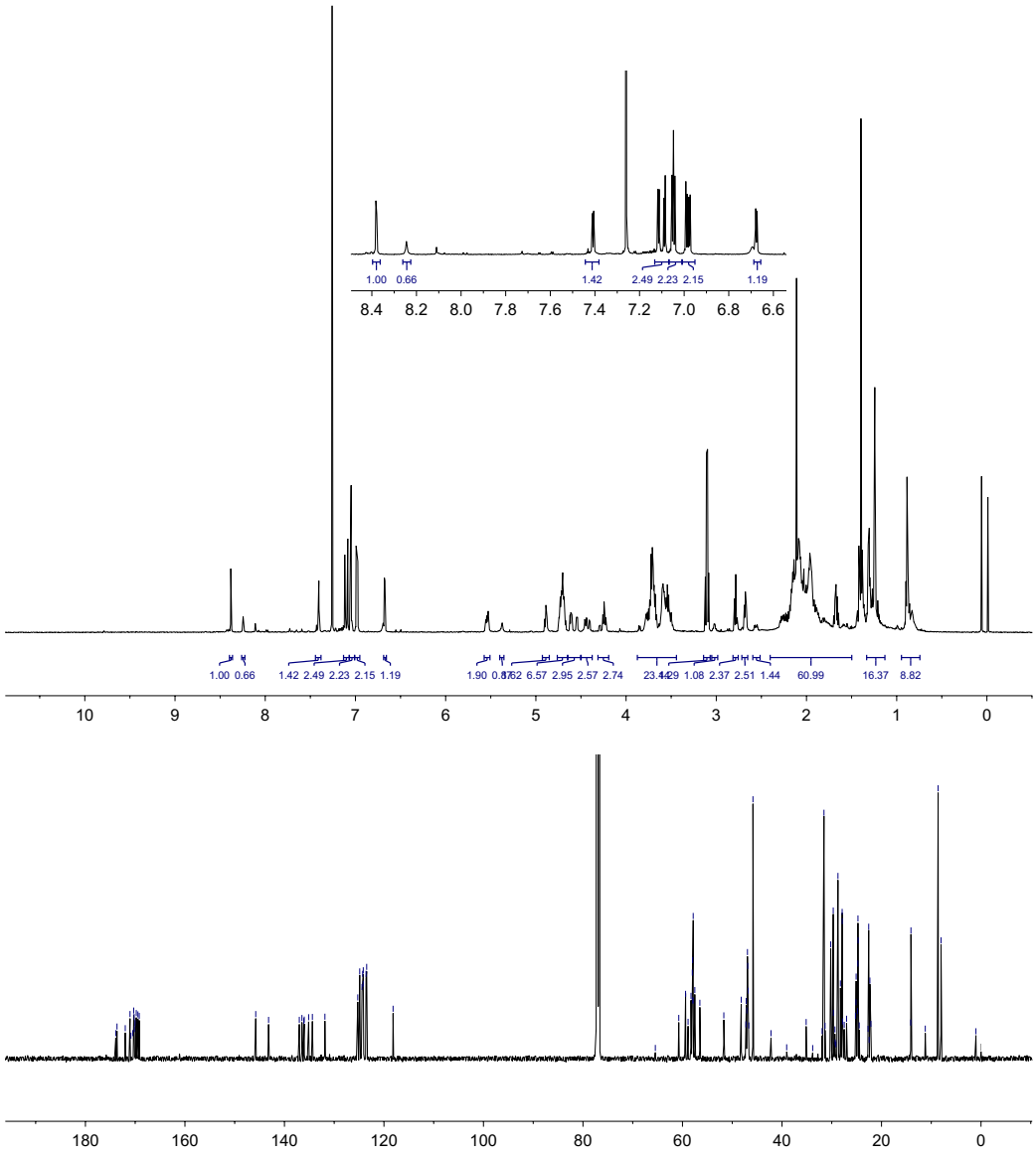


Analytical data for conjugate 4

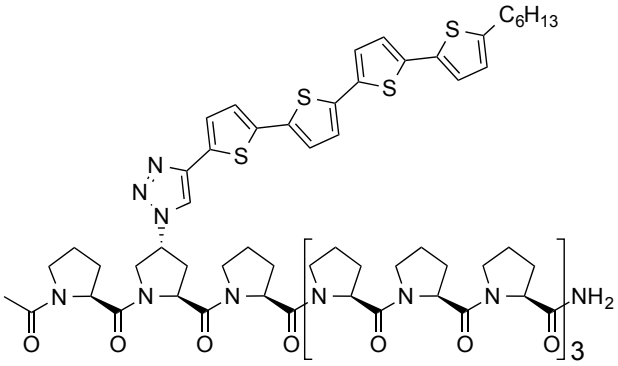


HRMS (MALDI-TOF) *m/z*: calculated $C_{71}H_{89}N_{13}O_{10}S_4$ $[M+H]^+$: 1411.5733; found: 1411.5731.

^{1}H NMR and ^{13}C NMR spectra (600 MHz, $CDCl_3$, 297K)

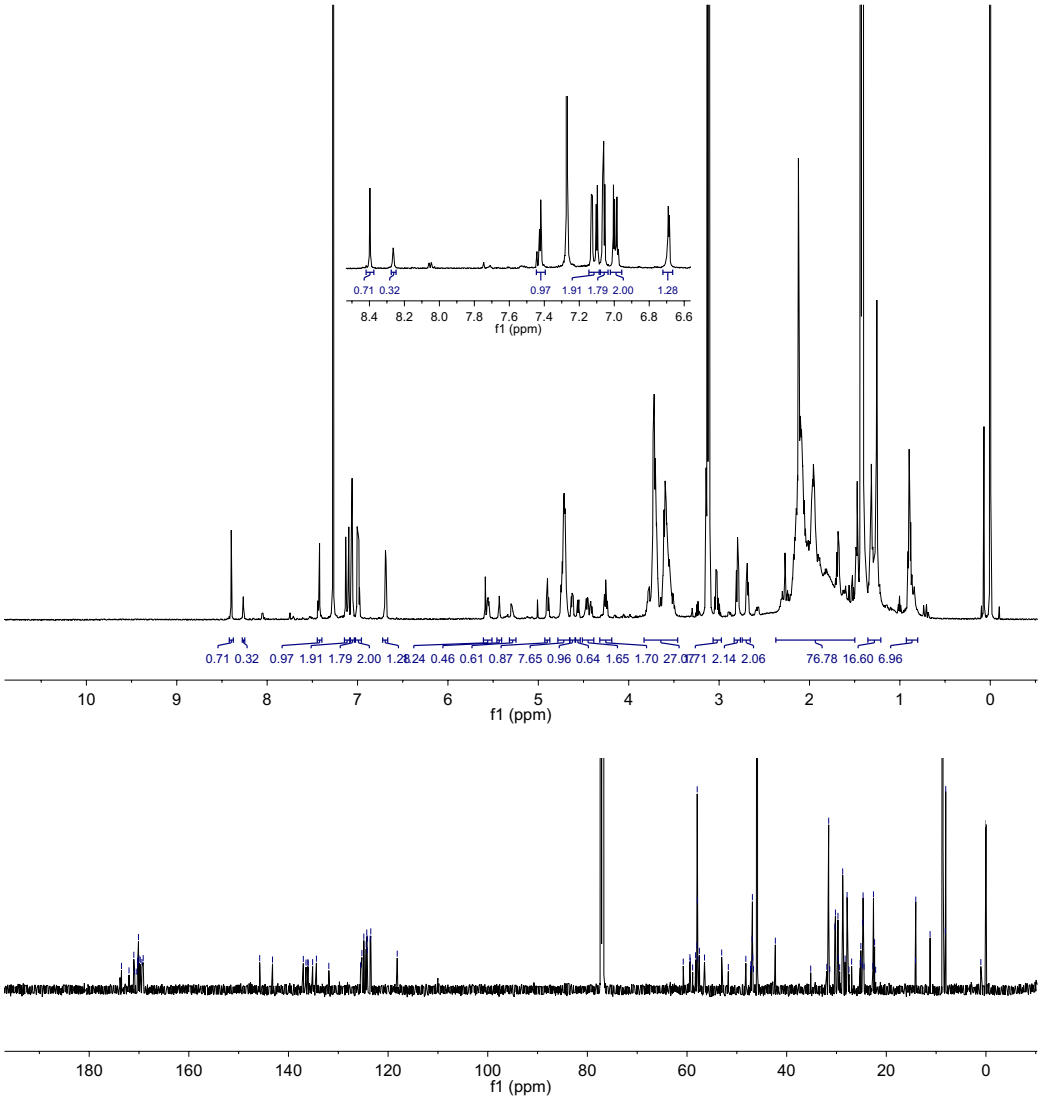


Analytical data for conjugate 5



HRMS (MALDI-TOF) *m/z*: calculated $C_{86}H_{110}N_{16}O_{13}S_4$ $[M+H]^+$: 1702.7316; found: 1702.7297.

^{1}H NMR and ^{13}C NMR spectra (600 MHz, $CDCl_3$, 297K)



2. Self-assembly of conjugates 1–5

Preparation of supramolecular assemblies

A stock solution of the oligoproline-quaterthiophene conjugate in THF (500 μ M) was used to prepare 50 µM samples in different THF:H_2O ratios: 10:90, 20:80, 30:70, 50:50, and 100:0. Solutions were annealed in a sand bath at 90 °C for 60 min and then slowly cooled down to room temperature over 120 min to allow self-assembly to proceed under thermodynamic control. Partial precipitation of self-assembled material was occasionally observed. The solutions/suspensions were studied using UV-Vis and CD spectroscopy, as well as TEM. 50 µM solutions in 100% H_2O were prepared without a stock solution, and treated with the same annealing procedure.

*Spectroscopic data of conjugates **1**–**5** in different THF:H_2O mixtures*

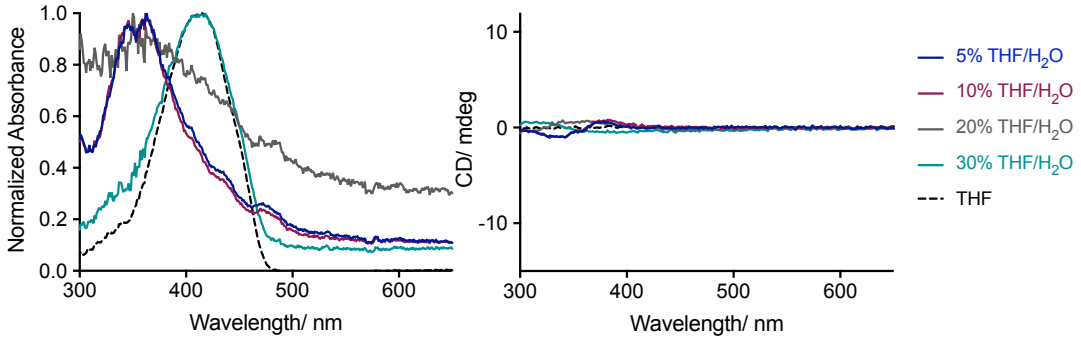


Figure S1: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of monomer **1**, 50 µM, 1 mm path length, in varying amounts of THF in water. *The CD spectra have significantly lower signal intensity than the one presented in manuscript since a cuvette with a shorter path length (1 mm) was used. The CD spectrum presented in the manuscript was recorded using a 10 mm path length cuvette.*

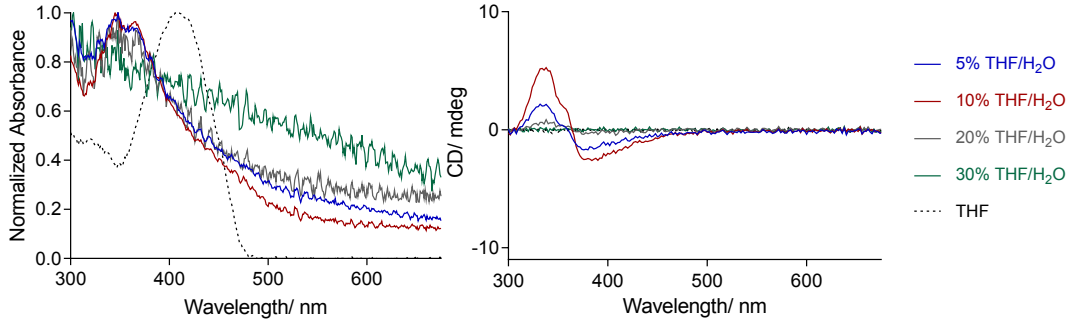


Figure S1a: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of monomer **1a**, 50 µM, 1 mm path length, in varying amounts of THF in water.

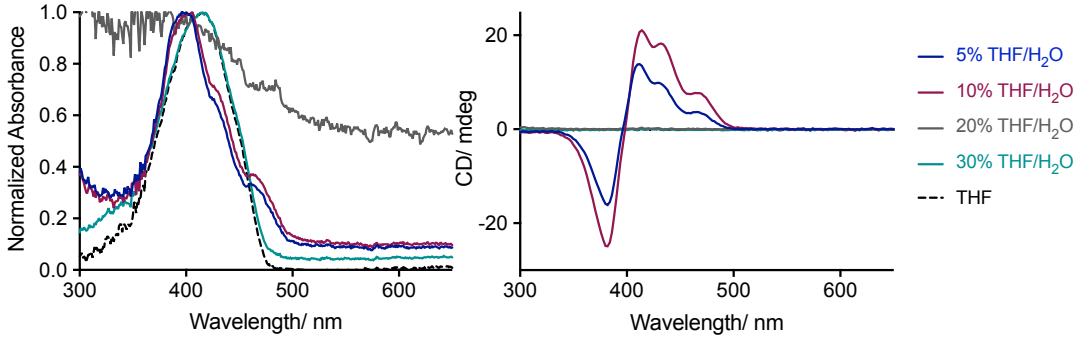


Figure S2: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of trimer **2**, 50 μM, 1 mm path length, in varying amounts of THF in water.

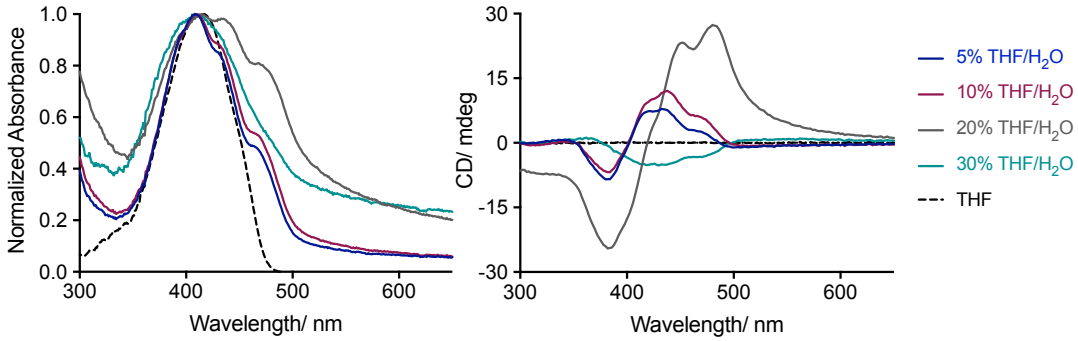


Figure S3: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of hexamer **3**, 50 μM, 1 mm path length, in varying amounts of THF in water.

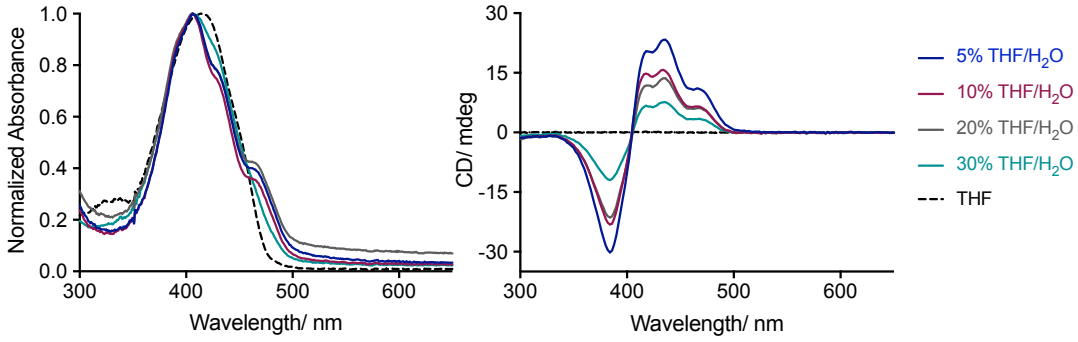


Figure S4: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of nonamer **4**, 50 μM, 1 mm path length, in varying amounts of THF in water.

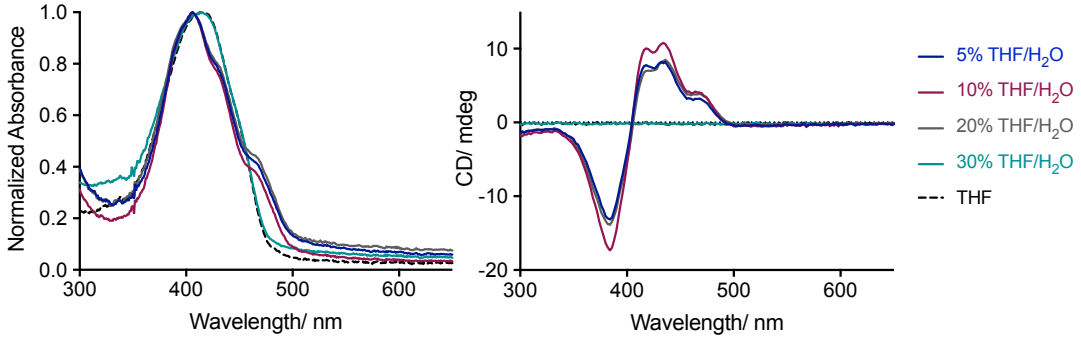


Figure S5: Normalized absorption spectra (left) and CD spectra (right) of annealed solutions of dodecamer **5**, 50 µM, 1 mm path length, in varying amounts of THF in water.

3. GIWAXS sample preparation and data

Thin films of 1 (200 µM) were prepared from the freshly annealed solutions in THF and H_2O (30:70) on heated (50 °C) Si/SiO_2 substrate. Annealing was performed overnight, and then the cap was opened and the THF was allowed to slowly evaporate. GIWAXS measurements of the thin films were performed at the beamline BL09 of the DELTA Synchrotron in Dortmund using a photon energy of 13 keV (λ = 0.9537 Å). The beam size was 1.0 mm × 0.2 mm (width × height), and samples were irradiated just below the critical angle for total reflection with respect to the incoming X-ray beam (0.1°). The scattering intensity was detected on a 2D image plate (MAR-345) with a pixel size of 150 μm (2300 × 2300 pixels), and the detector was placed 523 mm from the sample center. Scattering data are expressed as a function of the scattering vector: $q = 4\pi/\lambda \sin(\Theta)$, where Θ is a half the scattering angle and λ = 0.9537 Å is the wavelength of the incident radiation. Here qxy (qz) is a component of the scattering vector in-plane (out-of-plane) to the sample surface. All X-ray scattering measurements were performed under vacuum (~1 mbar) to reduce air scattering and beam damage to the sample. All data processing and analysis was performed by using the software package Datasqueeze (http://www.datasqueezesoftware.com).

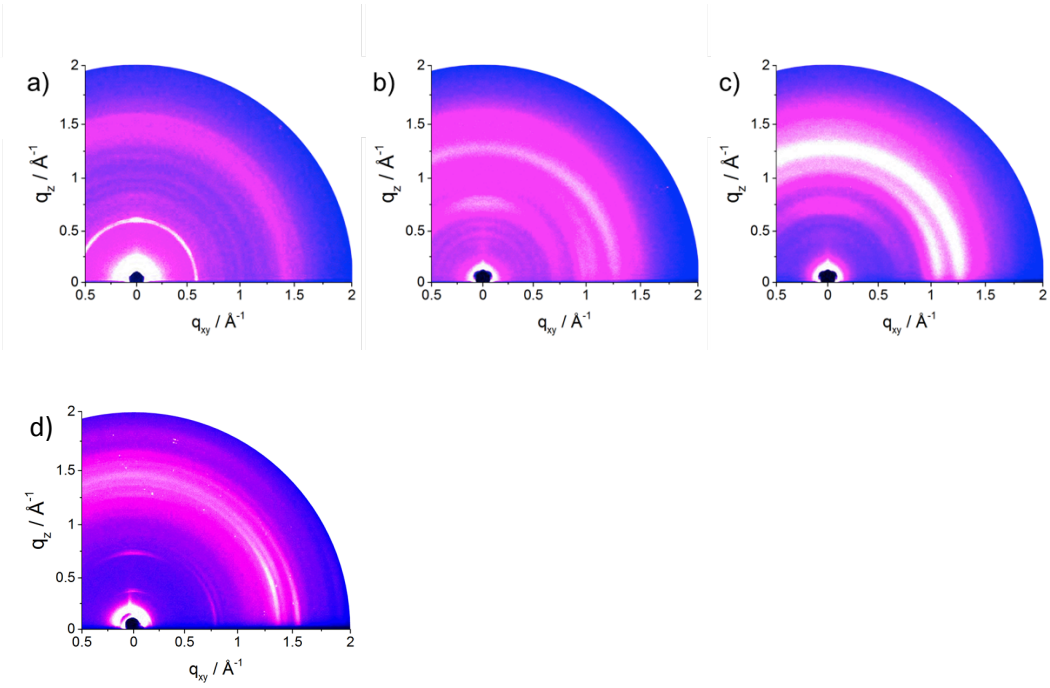


Figure S6. GIWAXS of conjugate a) **2**, b) **4**, c) **5** and d) **1a** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate.

4. AFM micrographs

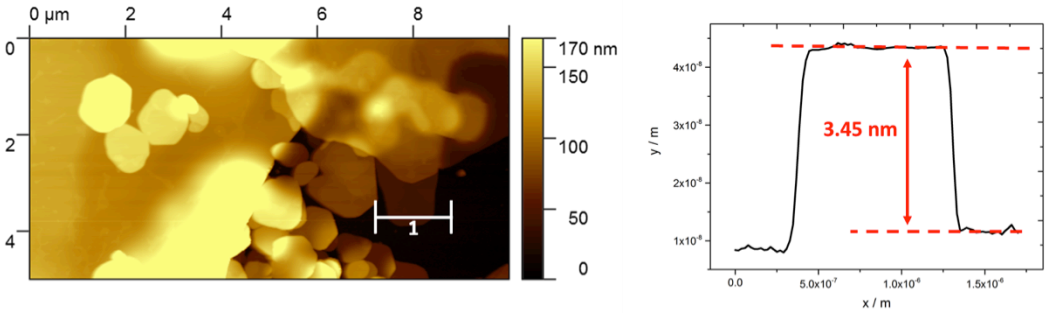


Figure S7. AFM and height profile of conjugate **1** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate

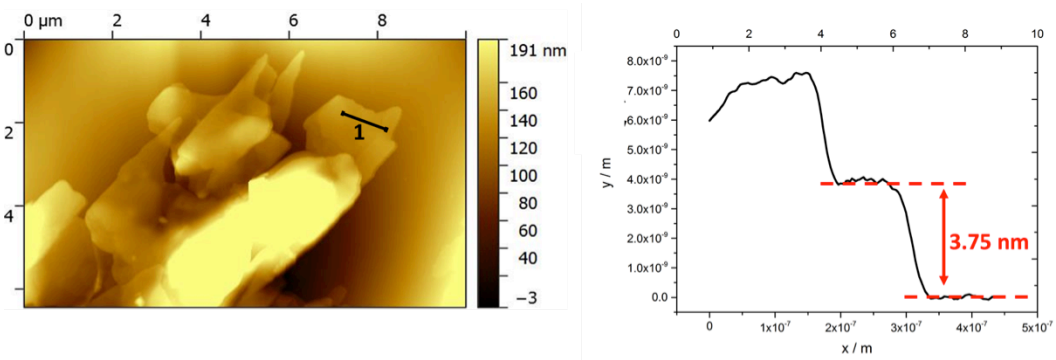


Figure S8. AFM and height profile of conjugate **2** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate

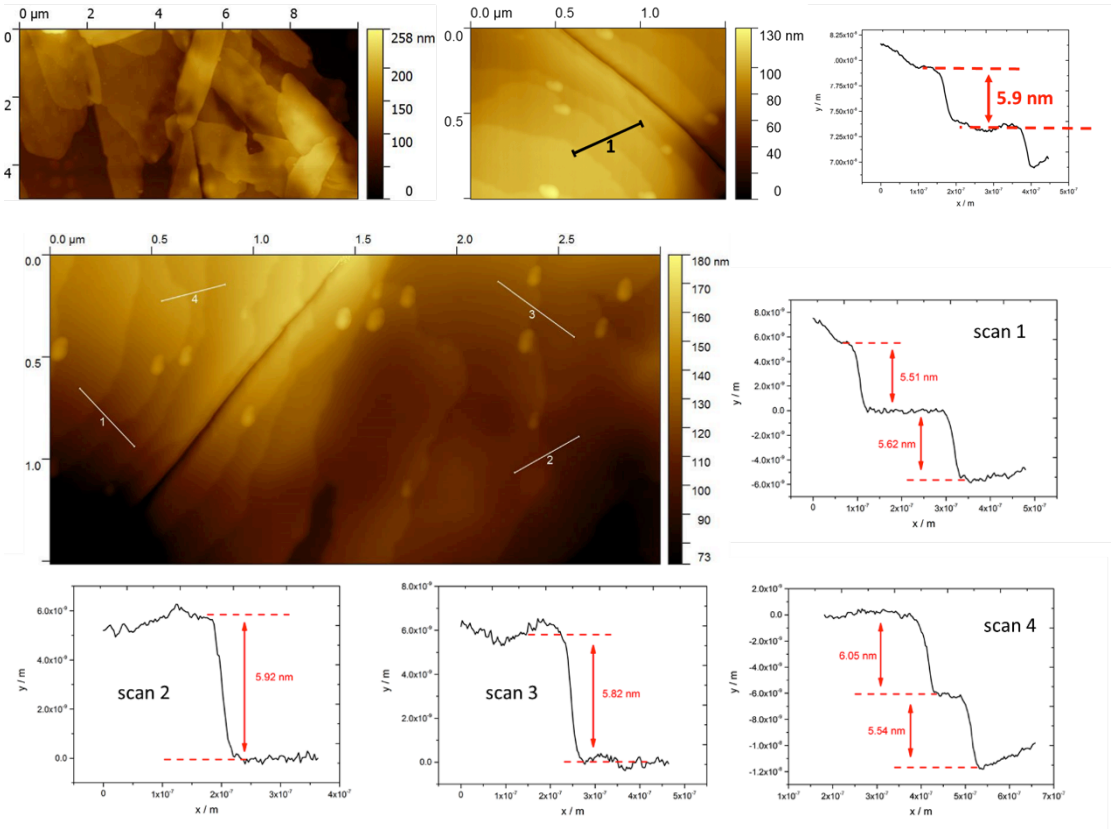


Figure S9. AFM and height profile of conjugate **3** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate.

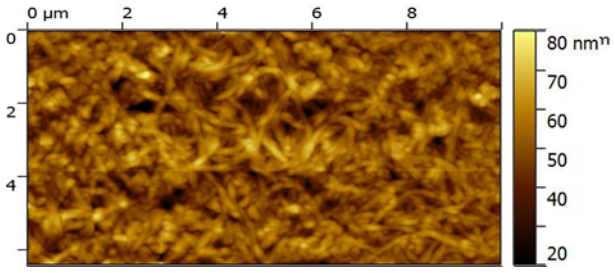


Figure S10. AFM of conjugate **4** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate.

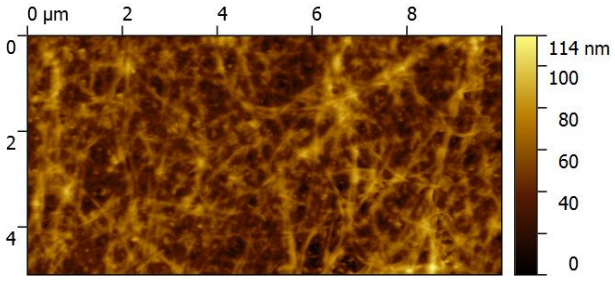


Figure S11. AFM of conjugate **5** deposited from aged THF and H_2O mixture (30:70) on Si/SiO_2 substrate.

5. Additional TEM micrographs

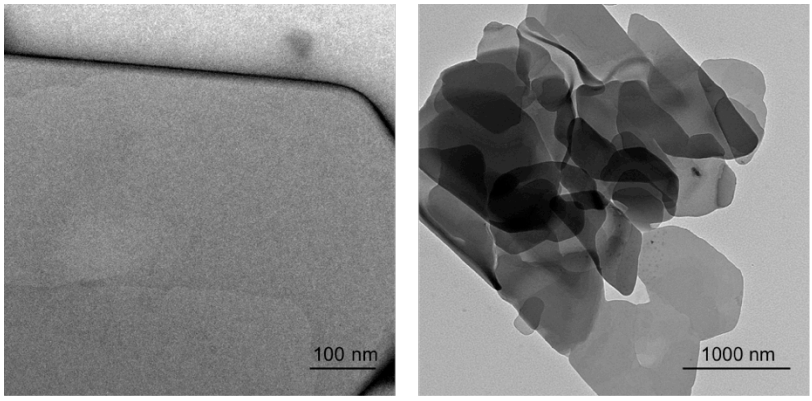


Figure S12. TEM micrographs of conjugate **1** deposited from solutions of THF:H_2O (20:80, 50µM), after annealing and stained with 2% uranyl acetate.

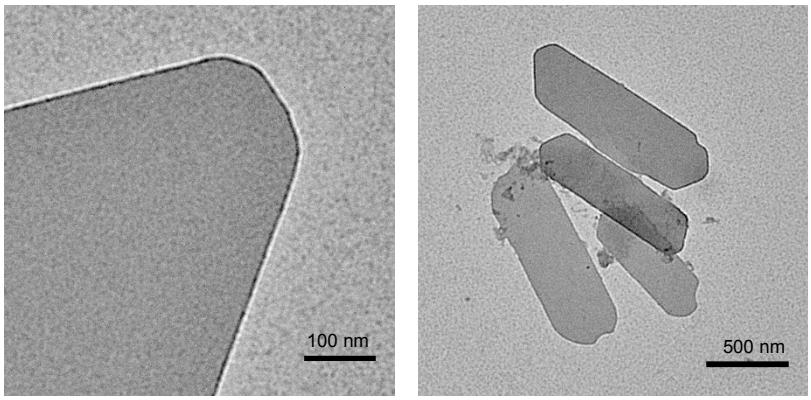


Figure S12a: TEM micrographs of conjugate **1a** deposited from solutions of THF:H_2O (20:80, 50µM), after annealing and stained with 2% uranyl acetate.

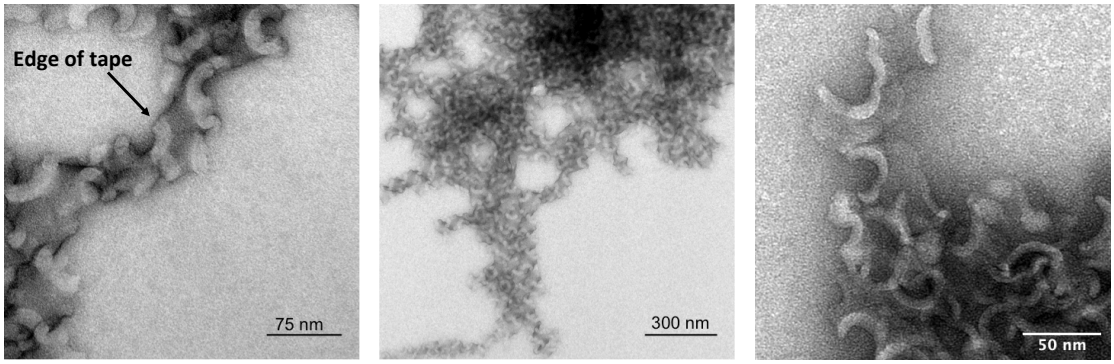


Figure S13. TEM micrographs of conjugate **2** deposited from solutions of THF:H_2O (20:80, 50μM) after annealing and stained with 2% uranyl acetate.

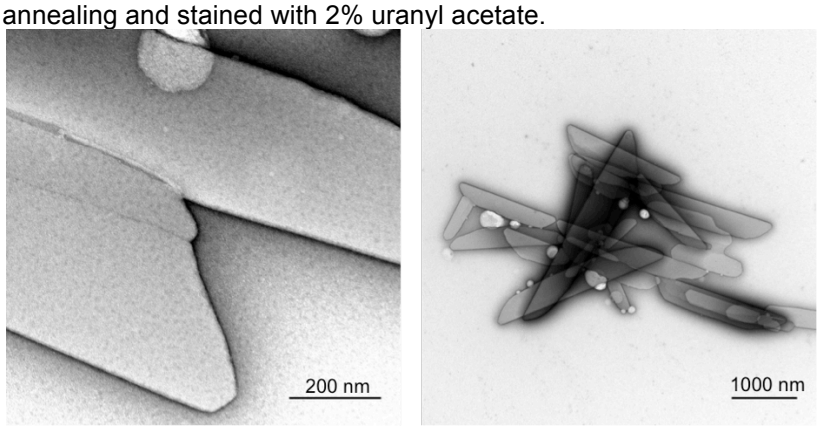


Figure S14. TEM micrographs of conjugate **3** deposited from solutions of THF:H_2O (20:80, 50μM) after annealing and stained with 2% uranyl acetate.

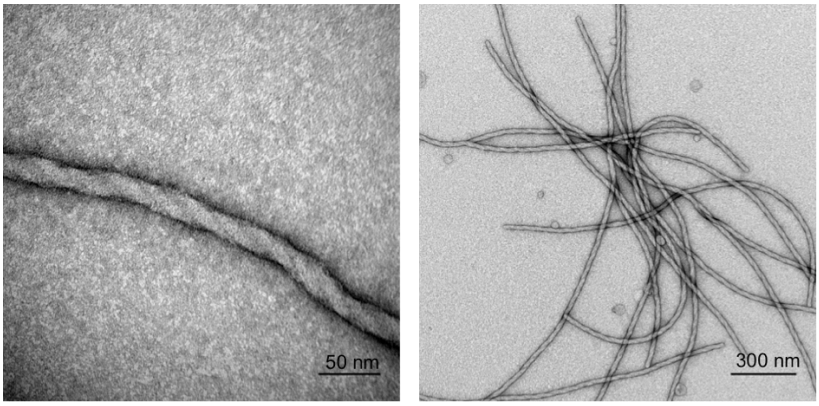


Figure S15. TEM micrographs of conjugate **4** deposited from solutions of THF:H_2O (20:80, 50μM) after annealing and stained with 2% uranyl acetate.

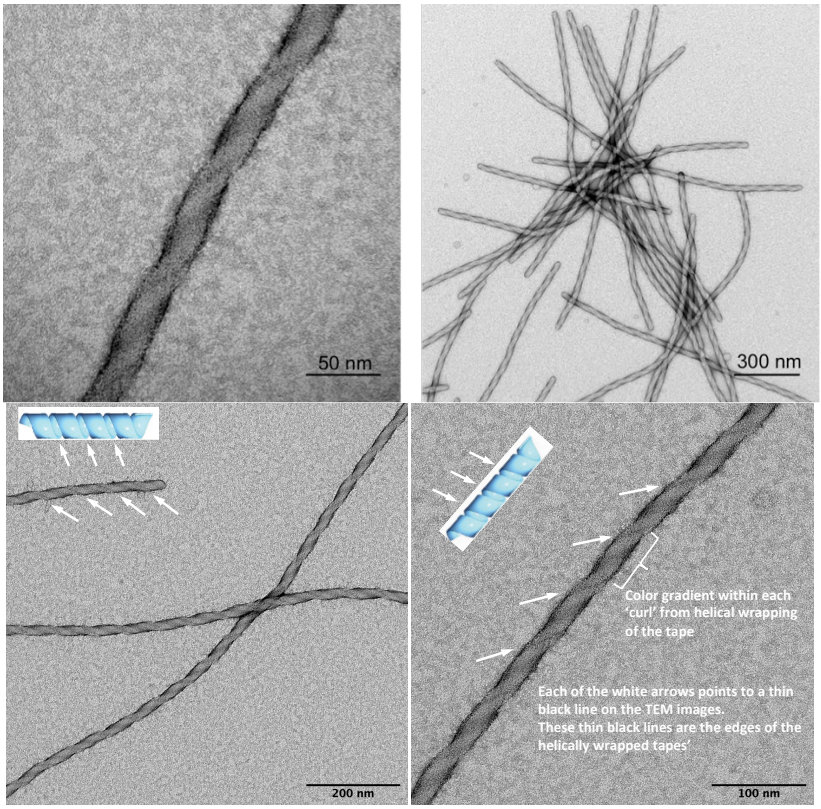


Figure S16. TEM micrographs of conjugate **5** deposited from solutions of THF:H_2O (10:90, 50μM) after annealing and stained with 2% uranyl acetate.

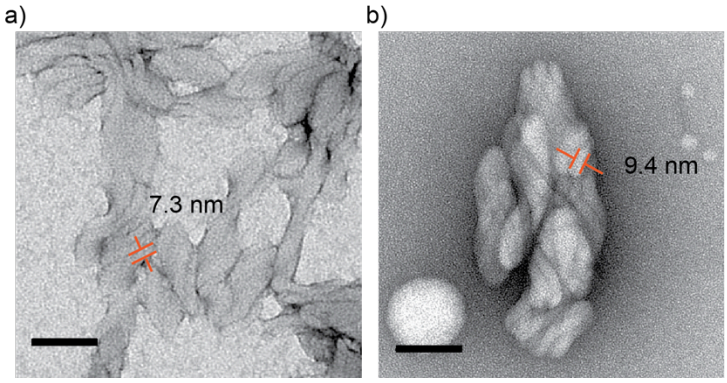


Figure S18. TEM of conjugates **4** (a) and **5** (b) deposited out of 50mM solutions in 100% H_2O and stained with 2% uranyl acetate. Scale bars represent 50 nm.

6. References

1. B. T. Holmes, P. Deb, W. T. Pennington, and T. W. Hanks, *J. Polym. Res.*, 2006, **13**, 133–144.

2. U. Lewandowska, W. Zajaczkowski, L. Chen, F. Bouillière, D. Wang, K. Koynov, W. Pisula, K. Müllen, H. Wennemers, *Angew. Chem. Int. Ed.* **2014,** *53*, 12537–12541.

3. C. Huang, C. Zhen, S. Su, K. Loh, Z. Chen; *Org. Lett.*, 2005, **7**, 391–394.

4. K. Wang, R.-Z. Liang, J. Wolf, Q. Saleem, M. Babics, P. Wucher, M. Abdelsamie, A. Amassian, M. Ryan Hansen, and P. M. Beaujuge, Adv. Funct. Mater., 2016, **26**, 7103–7114.