Supporting Information

Variation of formal hydrogen bonding networks within electronically delocalized pi-conjugated oligopeptide nanostructures

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General synthetic considerations

N-Methylpyrrolidone (NMP), O-(Benzotriazol-1-yl)-N,N,N',N'-tetramethyluronium hexafluorophosphate (HBTU), Wang resins, and Fmoc-protected amino acids were obtained from Advanced ChemTech. Benzotriazol-1-yl-oxytrypyrrolidinophosphonium hexafluorophosphate (PyBOP) was obtained from Oakwood Chemical. All other chemicals and reagents were received from Alpha Aesar or Sigma Aldrich and used as received. 4,4'-(1E,1'E)-1,4-phenylenebis(ethene-2,1-diyl))dibenzoic acid was prepared using literature procedures. 1

NMR Spectroscopy: 1H NMR spectra were collected using a Bruker Avance 400 MHz FT-NMR spectrometer and processed with Bruker Topspin V 1.3 or 2.1. Chemical shifts are reported in parts per million relative to residual protio solvent: DMSO (2.50 ppm), water (4.79 ppm).

Attenuated total reflection-FTIR: Data obtained on dry peptides using a Thermo Nicolet NEXUS 670 FTIR.

Electrospray Ionization Mass-Spectrometry (ESI-MS): ESI data were collected on a Thermo Finnigan LCQ Deca ion trap mass spectrometer with electrospray ionization. Samples were prepared in a 1:1 methanol/water with 1% ammonium hydroxide solution and were run in negative mode.

Reverse Phase High Performance Liquid Chromatography (RP-HPLC): was performed on an Agilent Technologies 1100 Series Quaternary LC System and an Agilent Technologies PrepStar SD-1 instrument and fitted with a Phenomonex Luna 5 mm C8 column with ammonium formate aqueous buffer (pH 8) and acetonitrile used as the mobile phase. Analytical traces were run with a linear gradient starting at 10% MeCN and ending at 40% MeCN over the course of 40
- 50 minutes.

**UV-Vis and Photoluminescence (PL):** UV-Vis spectra were collected using a Varian Cary 50 Bio UV-Vis spectrophotometer. PL data were collected using a PTI Photon Technology International Fluorometer with an Ushio Xenon short arc lamp. Micromolar concentration samples were prepared using Millipore water and the pH was adjusted by the addition of 1 M HCl or 1M KOH. PL spectra were obtained using an excitation wavelength corresponding to the $\lambda_{\text{max}}$ of absorption.

**Circular Dichroism (CD):** CD spectra were acquired using a Jasco J-810 spectropolarimeter. Acidic and basic samples were prepared by adding 1M HCl or 1M KOH, respectively, to a stock solution of the peptide in Millipore water.

**Transmission Electron Microscopy (TEM):** TEM images were acquired on a Philips EM 420 transmission electron microscope equipped with an SIS Megaview III CCD digital camera with an operating voltage of 100 kV. 200 or 300 mesh Formvar carbon coated copper grids were purchased from Electron Microscopy Sciences. Grids were prepared as follows: A stock solution of a 0.1 mg/mL peptide in Millipore water was exposed to concentrated HCl vapor for 1 min. 10 $\mu$L of this solution was pipetted onto a grid. The grid was incubated for 8 minutes. The grid was then dipped sequentially into water then into a solution of 2% uranyl acetate stain and allowed to dry in air.

**Atomic Force Microscopy:** Solid peptide was dissolved in deionized water and brought to a 0.1 mg/ml concentration. The sample was sonicated for ~5 min, and then placed into a conc. HCl chamber for two hours. 10 $\mu$L of this sample was spotted directly onto freshly cleaved mica and allowed to dry for at least one hour prior to imaging.
**Molecular Dynamics Simulations**: Molecular dynamics simulations were conducted using the GROMACS 4.6 simulation suite.\(^9\) Peptide configurations were generated using the GlycoBioChem PRODRG2 Server (http://davapc1.bioch.dundee.ac.uk/cgi-bin/prodrg)\(^{10}\) with all residues and termini in their charge-neutral states. Initial structures of the isolated peptides were relaxed by performing energy minimization to remove any forces exceeding 10 kJ/mol.nm, then equilibrating the resulting structure by conducting 100 ps of Langevin dynamics simulations at 298 K implementing a generalized-Born implicit solvent model.\(^{11}\) Pairs of peptides were immersed in a 10×10×10 nm cubic simulation box and solvated by explicit water molecules at a density of 1 g/cm\(^3\). Peptides were modeled using the CHARMM27 force field,\(^{12}\) and water modeled explicitly using the TIP3P model.\(^{13}\) Simulations were conducted in the NPT ensemble at 298 K and 1 bar, employing a Nosé-Hoover thermostat\(^{14}\) and Parrinello-Rahman barostat.\(^{15}\) The equations of motion were integrated using a leap-frog algorithm with a 2 fs time step, and bond lengths fixed to improve efficiency. Electrostatic interactions were treated using Particle Mesh Ewald (PME) with a real-space cutoff of 1.0 nm and a 0.12 nm Fourier grid spacing,\(^{16}\) and Lennard-Jones interactions shifted smoothly to zero at a 1.0 nm.

**Umbrella sampling of dimer formation.** Umbrella sampling\(^{17}\) was implemented within GROMACS to efficiently sample homodimer formation pathways by restraining the inter-molecular distance between the peptide centers of mass. The umbrella sampling technique is an accelerated simulation technique that introduces artificial restraining potentials to constrain the simulation to remain in the vicinity of a prescribed inter-molecular separation, resulting in good sampling of this region of the dimerization pathway. Sampling of the entire pathway is achieved by conducting a number of independent simulations restrained to different inter-molecular separations with overlaps in the range of distances sampled in neighboring windows. The
weighted histogram analysis method (WHAM)\textsuperscript{18} was used to combine the inter-molecular distances sampled in each biased umbrella run and compute the unbiased free energy profile (potential of mean force) along the dimerization pathway\textsuperscript{19}.

To generate initial configurations for each umbrella sampling window, we performed a non-equilibrium pulling simulation in which a pair of peptides were initialized in a parallel configuration in which the center of mass distance between the peptides was set to 4.5 nm and then applied an harmonic restraining potential in the center of mass distance with a force constant of 1000 kJ/mol nm\textsuperscript{2} to close the distance between the peptides to 0.02 nm over the course of a 1 ns simulation. Initial frames for each umbrella window were harvested from this trajectory by identifying a system configuration possessing an inter-peptide separation close to the target distance for the umbrella window. Since the non-symmetric peptides possess an inherent directionality in their primary structure (i.e., N-terminal to C-terminal sense), we performed two pulling runs, one in which the peptides were placed in a parallel configuration, and the other in an anti-parallel orientation.

Pairs of peptides were restrained at inter-molecular separations between 0.0 nm and 4.62 nm in increments of 0.14 nm using a harmonic restraining potential with a force constant of 1000 kJ/mol nm\textsuperscript{2}, and simulations in each of the thirty three umbrella windows were conducted for 2.5 ns. The first 600 ps of each run were discarded for equilibration, beyond which time the inter-molecular spacing, dimer structure, and system energy attained steady-state values. The inter-molecular distances were recorded over the production portion of each umbrella run, and used to solve for the free energy profile along the dimerization pathway parameterized by the peptide the inter-molecular spacing by solving the WHAM equations using the g\_wham solver distributed with GROMACS\textsuperscript{20}. Free energy profiles were solved to a tolerance of \(1 \times 10^{-6}\) in the probability
profile, and uncertainties in the profiles estimated by conducting 50 rounds of bootstrap resampling. For each peptide pair, a plateau in the free energy profile was observed beyond intermolecular distances of 4.0 nm, validating that the maximum umbrella window of 4.62 nm was sufficiently large such that the free energy was no longer a function of distance within the error bars of the curves. Physically, solvent screening of the inter-peptide interaction was strong enough such that the peptide pair became effectively non-interacting at these separations. For the symmetric peptides, we report a single PMF curve, whereas for the non-symmetric peptides we report two PMF profiles corresponding to parallel and antiparallel peptide orientations. Importantly, the 2.5 ns umbrella sampling windows were sufficiently long for the inter-peptide distance to equilibrate to the artificial restraining potential – as verified by the attainment of a steady-state value – but sufficiently short that rotations from parallel to antiparallel (or vice versa) orientations were not observed.

**Interaction energy decomposition.** To probe the role of the various energetic contributions as a function of peptide separation along the dimerization pathways, we split the system into two subsystems: peptides and solvent. We then computed the energetic interactions – intramolecular bends and torsions, intramolecular and intermolecular van der Waals interactions, intramolecular and intermolecular Coulomb interactions – of each subsystem with itself, and between the subsystems. Treatment of the long-range electrostatics using Particle Mesh Ewald does not admit a straightforward pairwise partitioning of the reciprocal space Coulombic interactions between different subsystems, so we reanalyzed the frames from our simulation trajectories using a reaction field treatment of electrostatics with a cutoff of 1.0 nm and an infinite dielectric constant material beyond the cutoff.
**Dimer relative twist angles.** Twist angles between peptides in the dimer, \( \Delta \theta_{\text{dimer}} \), were computed by forming the cross product between the vectors, \( \vec{v}_1 \) and \( \vec{v}_2 \), linking the terminal aromatic carbon atoms in the OPV linker region of each peptide: 
\[
\vec{v}_1 \times \vec{v}_2 = |\vec{v}_1||\vec{v}_2|\sin(\Delta \theta_{\text{dimer}}) \hat{n},
\]
where \( \hat{n} \) is the unit normal to the plane of the vectors \( \vec{v}_1 \) and \( \vec{v}_2 \). Since the peptides in the homodimer are identical, we have adopted a convention in which we label the peptides 1 and 2 such that the relative twist angle is non-negative.

**Hydrogen bonds.** To determine the presence or absence of a hydrogen bond, a geometric criterion was adopted wherein a hydrogen bond was considered made if the H-donor-acceptor angle was less than 30°, and the donor-acceptor distance less than 0.35 nm. All calculations were performed using the g_hbond application within GROMACS.\(^9\)
Synthetic Procedures

**Diethyl 4-nitrobenzylphosphonate.** Prepared according to literature reports.\(^1\)

![Diethyl 4-nitrobenzylphosphonate](image)

**(E)-methyl 4-(4-nitrostyrlyl)benzoate.** Prepared according to literature reports.\(^2\)

![**(E)-methyl 4-(4-nitrostyrlyl)benzoate](image)

**(E)-methyl 4-(4-aminostyrlyl)benzoate.** A solution of *(E)-methyl 4-(4-nitrostyrlyl)benzoate* 3 (2.01 g, 7.06 mmol) in EtOAc (100 ml) was heated to 75°C under nitrogen with stirring. Stannous Chloride dihydrate (7.95 g, 35.3 mmol) was added and the mixture was allowed to stir overnight. The reaction mixture was allowed to cool RT followed by the addition of aqueous NaHCO\(_3\) until the solution was pH 8. This suspension was filtered through a pad of celite and washed with EtOAc. The filtrate was washed with H\(_2\)O (2x) and brine (2x) and dried over MgSO\(_4\), filtered and the solvent was removed under reduced pressure to give (5.97 mmol 85%) of a yellow/orange solid. \(^1\)H-NMR (400 MHz, d-DMSO) \(\delta\): 7.90 (d, 2H, \(J=8.4\)), 7.62 (d, 2H, \(J=8.8\)), 7.32 (d, 2H, \(J=8.8\)), 7.24, (d, 1H, \(J=16.4\)), 6.96 (d, 1H, \(J=16.4\)), 6.57 (d, 2H, \(J=8.4\)), 5.43
(s, 2H), 3.84 (s, 3H); $^{13}$C-NMR (100 MHz, d-DMSO) δ: 166.0, 149.4, 143.0, 132.1, 129.5, 128.2, 126.9, 125.7, 124.1, 121.3, 113.8, 51.9; HRMS (EI/CI) m/z calculated for (C$_{16}$H$_{15}$NO$_2$) 253.1103, found 253.1101.

**$(E)$-4-(4-aminostyryl)benzoic acid.** To a suspension of $(E)$-methyl 4-(4-aminostyryl)benzoate (1.79 g, 7.06 mmol) in 1:1 EtOH/H$_2$O (120 ml), KOH (1.98 g, 35.3 mmol) was added. This mixture was refluxed overnight, then allowed to cool to RT. The volume was reduced approximately 50%, then 1M HCl was added until the solution was acidic. The resulting ppt was filtered and collected. Resulting in 1.42 g (5.93 mmol, 84 %) of an light brown solid. $^1$H-NMR (400 MHz, d-DMSO) δ: 7.77 (d, 2H, $J$ = 8.0), 7.35 (d, 2H, $J$ = 8.4), 7.27 (d, 2H, $J$ = 8.4), 7.03, (d, 1H, $J$ = 16.4), 6.87 (d, 1H, $J$ = 16.4), 6.55 (d, 2H, $J$ = 8.4), 5.30 (s, 2H); $^{13}$C-NMR (100 MHz, d-DMSO) δ: 168.5, 148.6, 140.2, 137.1, 129.3, 128.6, 127.5, 124.9, 124.4, 123.0, 113.9; HRMS (EI/CI) m/z calculated for (C$_{15}$H$_{13}$NO$_2$) 239.0946, found 239.0946.

**$(E)$-4-(4-((((9H-fluoren-9-yl)methoxy)carbonyl)amino)styryl)benzoic acid.** To a suspension of $(E)$-4-(4-aminostyryl)benzoic acid (0.822 g, 3.44 mmol) in 1:1 H$_2$O/MeCN (90 ml) Sodium Bicarb (0.144 g, 1.72 mmol) was added followed by Fmoc-OSu (1.12g, 3.33 mmol). This was allowed to stir overnight at RT. The reaction mixture was then diluted with water and the product
was collected by filtration to give 1.22 g (2.65 mmol, 77%) of a light tan solid. \(^1\)H-NMR (400 MHz, d-DMSO) \(\delta\): 9.84 (s, 1H), 7.92 (m, 4H), 7.76 (d, 2H, \(J= 7.6\)), 7.67, (d, 2H, \(J= 8.4\)), 7.56-7.41 (m, 6H), 7.38-7.31 (m, 3H), 7.20 (d, 1H, \(J= 16.4\)), 4.51 (d, 2H, \(J= 6.4\)), 4.32 (t, 1H, \(J= 6.6\)); \(^{13}\)C-NMR (100 MHz, d-DMSO) \(\delta\): 167.1, 153.3, 143.6, 141.7, 140.8, 130.9, 130.6, 129.6, 127.7, 127.4, 127.1, 126.2, 125.7, 125.1, 120.2, 118.3, 65.7, 46.6; HRMS (EI/CI) \(m/z\) calculated for \((C_{30}H_{23}NO_4)\) 461.1627, found 461.1621.

**Methyl 4-((diethoxyphosphoryl)methyl)benzoate.** To a solution of methyl 4-methylbenzoate (2.01 g, 13.3 mmol) and 1,2-Dichloroethane (100 ml) was added benzyol peroxide (catalytic amount) followed by NBS (2.84 g, 16.0 mmol) and the resulting mixture was heated to reflux while being stirred under nitrogen. After three hours, another catalytic amount of benzyol peroxide was added and the mixture was reflux for an additional three hours. The reaction mixture was allowed to cool to RT, the succinimide was precipitated out with the addition of \(\text{Et}_2\text{O}\) (20 ml) and was washed with water (2x) and brine (2x), dried over \(\text{MgSO}_4\), filtered and the solvent was removed under reduced pressure to give a light yellow clear oil. Triethyl phosphite (2.28 ml, 13.3 mmol) was added to the oil which was stirred under nitrogen at 160°C for three hours. Excess triethyl phosphite was distilled off and the product was purified by column chromatography (EtOAc) to give 2.28 g (7.97 mmol, 60%) of a light yellow clear oil. Characterization data matches that found in the literature.\(^2\)
(E)-dimethyl 4,4’-(ethene-1,2-diyl)dibenzoate. Was prepared using general procedure for the Wittig-Horner reaction: Method A. A solution of 8 (2.37 g, 8.30 mmol) and methyl 4-formylbenzoate (1.36 g, 8.30 mmol) in THF (27 ml) was added to a suspension of NaOMe (0.897 g, 16.6 mmol) in THF (55.3 ml). The reaction was stirred under nitrogen for four hours and was then neutralized with 1M HCl. The resulting mixture was extracted with dichloromethane and the collected organics filtered through a pad of silica, then washed with water (2x) and brine (2x), dried over MgSO₄, filtered and the solvent was removed under reduced pressure to yield 1.10 g (3.73 mmol, 45%) of a white crystalline solid. Characterization data matched that found in the literature.

(E)-4,4’-(ethene-1,2-diyl)dibenzoic acid. To a suspension of 9 (1.10 g, 3.70 mmol) in 3:1 EtOH/H₂O (14 ml), KOH (1.04 g, 18.5 mmol) was added and the suspension was refluxed under nitrogen for 2 days. The reaction was allowed to cool to RT before a drop of conc. HCl was added. The resulting precipitate was filtered to give 0.793 g (2.96 mmol, 80%) of a white crystalline solid. Characterization data matched that found in the literature.
Dimethyl 4,4′-((1\textit{E},1′\textit{E})-1,4-phenylenebis(ethene-2,1-diyl))dibenzoate. A solution of 8 (2.02 g, 7.05 mmol) and terephthaldehyde (0.462 g, 3.44 mmol) in THF (12 ml) was added to a suspension of NaOMe (1.12 g, 20.6 mmol) in THF (70 ml). The reaction was stirred under nitrogen for four hours and was then neutralized with 1M HCl. The resulting suspension was filtered, the collected solid was washed with THF, EtOH, and H\textsubscript{2}O, then allowed to dry to give 1.15 g (2.88 mmol, 84%) of a light yellow solid. Characterization data matched that found in the literature.\textsuperscript{5}

4,4′-((1\textit{E},1′\textit{E})-1,4-phenylenebis(ethene-2,1-diyl))dibenzonic acid. A suspension of 11 (0.501 mg, 1.25 mmol) in 3:1 EtOH/H\textsubscript{2}O (16 ml), KOH (0.352 g, 6.27 mmol) was added and the suspension was refluxed under nitrogen for 2 days. The reaction mixture was allowed to cool and the solvent was removed under reduced pressure and the resulting carboxylate salt was taken up in hot H\textsubscript{2}O (80 ml) and acidified with conc. HCl, and filtered. The resulting solid was washed with water and allowed to dry to give 0.425 g (1.15 mmol, 92%) of a yellow solid. Due to the extremely low solubility of this compound it was not possible to obtain NMR data. HRMS (EI/CI) \textit{m/z} calculated for (C\textsubscript{24}H\textsubscript{18}O\textsubscript{4}) 370.1205, found 370.1206. UV (H\textsubscript{2}O): \textit{\lambda}_{\text{max}} = 366 \text{ nm}. 

S12
**p-Divinylbenzene.** Prepared according to literature reports.  

\[
\text{OHC} \quad \text{CHO} \quad \text{CH}_2\text{(PPh}_3\text{)Br} \quad \text{K}_2\text{CO}_3 \quad \text{Dioxane reflux}
\]

\[
\text{HO} \quad \text{NH}
\]

**N-Succinimidyl-4-iodobenzoate.** Prepared according to literature reports.  

\[
\text{O} \quad \text{O} \quad \text{O} \quad \text{DCC} \quad \text{Dioxane}
\]

\[
\text{OH} \quad \text{N} \quad \text{O} \quad \text{I}
\]

\[
\text{HO} \quad \text{O} \quad \text{NH}_2 \quad \text{I} \quad \text{NH}_2 \quad \text{Pd(OAc)}_2 \quad \text{NBu}_3 \quad \text{P(o-Tolyl)}_3 \quad \text{DMF 80°C}
\]

**\((E)-4-(4-vinylstyryl)aniline.**** 4-idoaniline (684 mg, 3.12 mmol), Pd(OAc)$_2$ (8.4 mg, 0.037 mmol), tri(o-tolyl)phosphine (95.0 mg, 0.312 mmol) were added to a flame dried schlenk tube and was evacuated and backfilled with N$_2$ (3x). A solution of 1,4-divinylbenzene (1.22 g, 9.37 mmol), tributylamine (1.07 ml, 5.52 mmol) and DMF (30 ml) was degassed with a purge of N$_2$ and then cannulated onto the solid reagents. This reaction mixture was then degassed using three freeze-pump-thaw cycles and the reaction mixture was then allowed to stir under N$_2$ at 80°C for 12 h. The crude mixture was extracted with Et$_2$O, then washed with water (2x) and brine (2x), dried over MgSO$_4$, and the solvent was removed under reduced pressure. The crude mixture was then dissolved in a minimum amount of 1:1 Hexane:EtOAc and then precipitated by the addition of hexane (100 ml) and the precipitate was filtered and washed with hexane to give a light brown solid (0.332 g, 48%). $^1$H NMR (400 MHz, Chloroform-$d$) $\delta$: 7.43 (d, $J$ = 8.0 Hz, 2H), 7.38 (d, $J$ = 8.1 Hz, 2H), 7.33 (d, $J$ = 8.0 Hz, 2H), 7.03 (d, $J$ = 16.3 Hz, 1H), 6.90 (d, $J$ = 16.2 Hz, 1H), 6.73 (d, $J$ = 11.0 Hz, 1H), 6.68 (d, $J$ = 7.6 Hz, 2H), 5.74 (d, $J$ = 17.5 Hz, 1H), 5.22 (d, $J$ = 11.1 Hz, 1H), 3.75 (s, 2H). HRMS (EI/Cl) $m/z$ calculated for (C$_{16}$H$_{15}$N)$_2$221.12045, found 221.1204. 

S13
**N-succinimidyl-4-((E)-4-((E)-4-aminostyryl)styryl)benzoate.**  
(E)-4-(4-vinylstyrlylaniline (0.323 g, 1.46 mmol), N-succinimidyl-4-iodobenzoate (0.504 g, 1.46 mmol), Pd(OAc)$_2$ (3.9 mg, 0.017 mmol), tri(ortho-tol)phosphate (44.4 mg, 0.146 mmol) were added to a flame dried schlenk tube and was evacuated and backfilled with N$_2$ (3x). A solution of tributylamine (0.504 ml, 2.16 mmol) and DMF (20 ml) was degassed with a purge of N$_2$ and then cannulated onto the solid reagents. This reaction mixture was then degassed using three freeze-pump-thaw cycles and the reaction mixture was then allowed to stir under N$_2$ at 80°C for 24 h. The crude reaction mixture was poured into 100 ml of H$_2$O and filtered. The crude material was washed with hexane and THF and allowed to dry to give a brown solid (0.639g, 97%). $^1$H-NMR (400 MHz, d-DMSO) δ: 8.08 (d, 2H, $J = 8.0$ Hz), 7.86 (d, 2H, $J = 8.0$ Hz), 7.65-25 (m, 8H), 7.14 (d, 2H, $J = 16.0$), 6.92 (d, 2H, $J = 16.0$ Hz), 6.57 (d, 2H, $J = 8.0$ Hz), 5.37 (s, br, 2H), 2.91 (s, 4H).

**General Dimerization procedure.** A solution of Diacid chromophore (0.3 eq) and PyBOP (0.6 eq) was dissolved in a 2:1 solution of NMP/DCM, once dissolved DIPEA (7 eq) was added and the solution was stirred for one min. This solution was added to Pre-substituted free amine Wang resin (1.0 eq) after it was allowed to swell in DCM for 10 min. The resulting mixture was placed on a rotisserie overnight. The reaction mixture was then filtered and the resin was washed with NMP, DCM, and MeOH and that was repeated three times. A second coupling solution of Diacid chromophore (0.2 eq) and PyBOP (0.4) in 2:1 NMP/DCM followed by DIPEA (7 eq) was added to the resin and placed on the rotisserie overnight. This resin was washed as previously described.
and a blank coupling cycle with PyBOP (0.4 eq) with DIPEA (7 eq) in 2:1 NMP/DCM was allowed to react for one hour and the resin was washed as previously described. The resin was cleaved in a 1:1 solution of DCM/Cleavage cocktail (95:2.5:2.5 TFA/TIPS/H₂O) for two hours. This solution was filtered out and the resin was washed with DCM, the filtrate was evaporated until 50% of the total volume remained, then cold Et₂O was added. The precipitated peptide was centrifuged and the remaining solution was decanted off. This was repeated again with cold Et₂O, and then twice with MeCN. The final centrifuged peptide was lyophilized to give final product.

DFAG-OPV2 non-symmetric peptide (1). The general peptide synthesis was used for all amino acids except for the Fmoc-protected stilbene and the Gly after the stilbene. The Stilbene was coupled on using HATU rather then HBTU and a solution of 2:1 NMP/DCM to do the couplings in. The Gly after the stilbene was coupled using triphosgene and 2,4,6-trimethylpyridine according to a published procedure. Final peptide was obtained as a off-white powder (0.036 g, 0.036 mMol, 38%). MS (ESI -) m/z calculated for (C₅₁H₅₇N₉O₁₄)⁻ : 1018.40, found 1018.17; m/z calculated for (C₅₁H₅₇N₉O₁₄)²⁻/2 : 508.70, found 508.23; m/z calculated for (C₅₁H₅₇N₉O₁₄)³⁻/3 : 338.80, found 338.11. ¹H NMR (400 MHz, D₂O) δ: 10.6 (s, 1H), 9.45 (m, 1H), 8.76 (d, 1H, J=9.1 Hz), 8.39 (br s, 1H), 8.22 (br s, 1H), 8.05 (br s, 1H), 7.91 (br s, 2H), 7.74 (d, 2H, J=8.2 Hz), 7.66 (d, 2H, J= 8.7 Hz), 7.53 (d, 2H, J= 9.2 Hz), 7.14-7.35 (m, 14H), 4.43 (br s, 1H), 4.33 (m, 1H), 4.27 (m, 2H), 4.04 (br s, 1H), 3.86 (m, 2H), 3.67 (m, 1H), 3.18 (m, 2H), 3.07 (m, 2H),
2.85 (m, 3H), 2.67 (m, 1H), 2.42 (m, 2H), 2.39 (m, 2H), 2.33 (m, 1H), 2.25 (dd, 2H, \(J=15.6, 7.8\) Hz), 1.28 (d, 3H, \(J=7.2\) Hz), 1.17 (m, 3H).

**DFAG-OPV2 symmetric peptide (2).** General dimerization procedure previously discussed was used. Final peptide was obtained as a off-white powder (0.021 g, 0.020 mMol, 43%). MS (ESI -) \(m/z\) calculated for \((C_{52}H_{56}N_{8}O_{16})^2/2 : 523.19\), found 522.75; \(m/z\) calculated for \((C_{52}H_{56}N_{8}O_{16})^3/3 : 348.46\), found 347.75; \(m/z\) calculated for \((C_{52}H_{56}N_{8}O_{16})^4/4 : 319.10\), found 318.48. \(^1\)H NMR (400 MHz, D\(_2\)O) \(\delta\): 7.84 (d, 4H, \(J=9.2\) Hz), 7.74 (d, 4H, \(J=8.8\) Hz), 7.41 (s, 2H), 7.31 (m, 5H), 7.25 (m, 6H), 4.68 (dd, 2H, \(J=11.2, 4.4\)) 4.41 (dd, 2H, \(J=8.8, 5.0\) Hz), 4.27 (q, 2H, \(J=6.0\) Hz), 4.06 (q, 4H, \(J=17.0\) Hz), 3.27 (dd, 2H, \(J=14.0, 5.4\) Hz), 2.95 (dd, 2H, \(J=14.4, 10\) Hz), 2.65 (qd, 6H, \(J=16.0, 6\) Hz), 1.22 (d, 6H, \(J=7.5\) Hz).

**DFAG-OPV3 non-symmetric peptide (3).** The general peptide synthesis was used for all amino acids except for the Gly after the OPV. The NHS activated ester of the OPV was coupled on the deprotected glycine by dissolving the OPV (133 mg, 0.302 mmol) in NMP (~30 ml) and diisopropylethylamine (0.176 ml) this solution was added to the deprotected resin and gently agitated for 12 h. The resin was then washed with a copious amount of NMP until the filtrate was no longer fluorescent followed by a washing procedures as follows: 3x NMP, 3x DMF, 2x isopropanol, 2x water, 2x (2x THF, 2x isopropanol), 2x acetonitrile, 2x diethyl ether, 2x hexanes. The Gly after the stilbene was coupled using triphosgene and 2,4,6-trimethylpyridine using
published procedure. Final peptide was obtained as a off-white powder (0.045 g, 40%). $^1$H NMR (400 MHz, $d$-DMSO) $\delta$: 9.97 (s, 1H), 8.70 (m, 1H), 8.29 (m, 1H), 8.13 (m, 1H), 7.99 (m, 1H), 7.90 (d, 2H, $J = 8.4$ Hz), 7.75-7.55 (m, 8H), 7.40 (d, 1H, $J = 16.4$ Hz), 7.32 (d, 1H, $J = 16.1$ Hz), 7.27-7.15 (m, 10 H), 4.54 (m, 2H), 4.45(m, 2H), 4.29 (m, 1H), 4.20 (m, 1H), 3.96-3.82 (m, 2H), 3.65 (q, 2H, $J = 4.2$ Hz), 3.09 (m, 2H), 2.83 (dd, 2H, $J = 14.1$, 9.6 Hz), 2.33 (m, 2H), 1.29 (d, 3H, $J = 7.1$ Hz), 1.19 (d, 3H, $J = 7.1$ Hz). MS (ESI -) $m/z$ calculated for (C$_{59}$H$_{63}$N$_9$O$_{14}$)$^{1-}$: 1120.45, found 1120.60; $m/z$ calculated for (C$_{59}$H$_{63}$N$_9$O$_{14}$)$^{2-}$/2 : 559.73, found 560.10. $^1$H NMR (400 MHz, D$_2$O) $\delta$: 9.96 (s, 1H), 8.7 (m, 1H), 8.29 (m, 1H), 8.13 (m, 1H), 7.99 (m, 1H), 7.9 (d, 2H, $J= 8.4$ Hz), 7.65 (m, 10H), 7.36 (q, 2H, $J=15.2$), 7.14-7.27 (m, 12H), 6.49 (br s, 1H), 4.55 (m, 1H), 4.45 (m, 1H), 4.29 (m, 1H), 4.13 (m, 1H), 3.89 (m, 1H), 3.66 (dd, 1H, $J= 11.2$, 4.0 Hz), 3.09 (m, 2H), 2.84 (dd, 2H, $J=11.6$, 8.8 Hz), 2.31-2.52 (m, 4H), 1.29 (d, 3H, $J= 6.8$ Hz), 1.20 (d, 3H, $J=6.8$ Hz).

**DFAG-OPV3 symmetric peptide (4)** was synthesized according to the general on-resin dimerization method using the OPV3 diacid, and is described in a prior report.
Figure S1. AFM image of DFAG-OPV2 Symmetric on mica substrate showing the formation of 1-D nanostructures.

Figure S2. AFM image of DFAG-OPV2 non-symmetric on mica substrate showing the formation of 1-D nanostructures.
Figure S3. HPLC trace of DFAG symmetric OPV2.

Figure S4. HPLC trace of DFAG non-symmetric OPV2.

Figure S5. HPLC trace of DFAG non-symmetric OPV3.
Figure S6. Attenuated total reflectance IR spectra of DFAG-OPV2 symmetric in the solid state.

Figure S7. Attenuated total reflectance IR spectra of DFAG-OPV2 non-symmetric in the solid state.
Figure S8. Attenuated total reflectance IR spectra of DFAG-OPV3 non-symmetric in the solid state.
Figure S9. $^1$H (top, 400 MHz, CDCl$_3$) and $^{13}$C (bottom, 100 MHz, CDCl$_3$) NMR of Diethyl 4-nitrobenzylphosphonate.
Figure S10. $^1$H (top, 400 MHz, $d_6$-DMSO) and $^{13}$C (bottom, 100 MHz, $d_6$-DMSO) NMR of (E)-methyl 4-(4-nitrostyryl)benzoate.
Figure S11. $^1$H (top, 400 MHz, $d_6$-DMSO) and $^{13}$C (bottom, 100 MHz, $d_6$-DMSO) NMR of (E)-methyl 4-(4-aminostyryl)benzoate.
Figure S12. $^1$H (top, 400 MHz, $d_6$-DMSO) and $^{13}$C (bottom, 100 MHz, $d_6$-DMSO) NMR of $(E)$-4-(4-(((9H-fluoren-9-yl)methoxy)carbonyl)amino)styryl)benzoic acid.
Figure S13. $^1$H (top, 400 MHz, CDCl$_3$) and $^{13}$C (bottom, 100 MHz, CDCl$_3$) NMR of Methyl 4-((diethoxyphosphoryl)methyl)benzoate.
Figure S14. $^1$H (top, 400 MHz, $d_6$-DMSO) NMR of (E)-dimethyl 4,4'-(ethene-1,2-diyl)dibenzolate.
Figure S15. $^1$H (top, 400 MHz, $d_6$-DMSO) and $^{13}$C (bottom, 100 MHz, $d_6$-DMSO) NMR of (E)-4,4'-(ethene-1,2-diyl)dibenzoic acid.
Figure S16. $^1$H (top, 400 MHz, $d_6$-DMSO) of N-succinimidyl-4-((E)-4-((E)-4-aminostyryl)styryl)benzoate.
Figure S17. $^1$H (400 MHz, D$_2$O) NMR of DFAG-OPV2 symmetric.

Figure S18. $^1$H (400 MHz, D$_2$O) NMR of DFAG-OPV2 non-symmetric.
Figure S19. $^1$H (400 MHz, D$_2$O) NMR of DFAG-OPV3 non-symmetric.
Figure S20. ESI of DFAG-OPV2 symmetric.

Figure S21. ESI of DFAG-OPV2 non-symmetric.
Figure S22. ESI of DFAG-OPV3 non-symmetric.
Figure S23. Representative molecular snapshots of peptide homodimers extracted from the configurational ensemble residing at the minimum of the PMF for (a) OPV2 symmetric, (b) OPV2 non-symmetric (parallel), (c) OPV2 non-symmetric (antiparallel), (d) OPV3 symmetric, (e) OPV3 non-symmetric (parallel), and (f) OPV3 non-symmetric (antiparallel) peptides.
Figure S24. HPLC traces for symmetric OPV2 samples. The original solid material was reconstituted and shows the presence of two species. The eluent corresponding to 4.37 min was collected. Half of this eluent sample was kept in the dark prior to HPLC re-analysis (b). The other half was irradiated with a 254 nm handheld lamp for 10 min prior to HPLC re-analysis (c). The peak at ca. 6.5 min in (a) and (c) is attributed to the *cis* isomer.
Figure S25: UV-vis spectra for nanostructures prepared in acidic solution from symmetric OPV2 (top) and nonsymmetric OPV2 (bottom) following continual irradiation and periodic UV-vis acquisition. The arrows indicate the trends with continued irradiation. The resulting spectral signatures at ca. 330 nm and 250 nm that evolve are characteristic of *cis*-stilbene, and the oxidized 6-pi electron photocyclication product leading to phenanthrene.
References


