Supplementary Information for

**Compartmentalized host spaces accommodating guest aromatic molecules in a chiral way by helix-peptide-aromatic framework**

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Conclusion: We successfully demonstrate here to prepare the nanosheet having a porous and channel structure using the bolaamphiphilic peptide composed of two amphiphilic helical peptides and the ONE unit at the center of the molecule. The helical blocks associated tightly to make two layers clamping the hydrophobic ONE layer. Void spaces were accordingly generated in the hydrophobic ONE layer, because the thick helical block with a 1.5 nm diameter held one thin flat molecule of the ONE group. The void spaces can make columns of the ONE groups by accommodating the guest ONE groups in a right-handed helical arrangement. This new strategy for making molecular assemblies with porous and channel structures has a characteristic point to provide a chiral host channel which can accommodate relatively large molecules. Using helical peptides clamping an aromatic group is therefore regarded as a new framework for porous nanomaterials. Incorporated conjugated blocks were regularly arranged by host-guest interactions in a chiral way. These conjugated block columns are expected as an efficient electron transfer path having chirality.
**Materials and synthesis**

All chemicals were purchased from commercial suppliers and used without further purification. Leu-Aib peptides were synthesized by a conventional liquid-phase method. All the intermediates were identified by $^1$H NMR spectroscopy (Bruker DPX-400) and further confirmed by MALDI mass spectrometry (Bruker ultraflexIII-KE). The purity of the intermediates was checked by thin-layer chromatography and that of S29L8ONE was further confirmed by HPLC.

NMR: spectroscopy: hydrogen nuclear magnetic resonance spectroscopy

MALDI: mass spectrometry: matrix assisted laser desorption ionization mass spectrometry

HPLC: high performance liquid chromatography

HATU: 2-(7-aza-1H-benzotriazole-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate

HOAt: 1-hydroxy-7-azabenzotriazole

DIEA: $N,N$-diisopropylethylamine

DMF: $N,N$-dimethylformamide
Scheme S1. Synthetic schemes of S29L8ONE.
**Compound 2**

To a 200 mL two-necked round bottom flask were added 2-bromonaphthalene (1.00 g, 4.83 mmol), CuI (184 mg, 966 μmol), and Pd(PPh₃)₂Cl₂ (407 mg, 580 μmol), dissolved with dry THF (8.00 mL). To the mixed solution were added trimethylsilylacetylene (820 μL, 19.3 mmol), and DIEA (3.36 mL, 19.3 mmol) at 0 °C. The reaction mixture was stirred for 72 h at 60 °C. The reaction mixture was extracted with diethyl ether, and the organic layer was washed with saturated NH₄Cl aq. three times, 4 wt% KHSO₄ aq. and saturated NaHCO₃ aq. once for each. The organic layer was washed with brine, dried over anhydrous MgSO₄, filtered and concentrated under reduced pressure. The product was purified with column chromatography (CHCl₃) to afford the desired product as a brown oil. Yield: 1.30 g (quant.).

¹H NMR (400 MHz, CDCl₃, δ): 8.00–7.24 (m, 7H, aromatic), 0.28 (s, 9H, TMS).

**Compound 3**

To a 100 mL round bottom flask was added 2-(trymethlysilylacetynyl)-naphthalene (100 mg, 446 μmol), dissolved with THF (2 mL), and MeOH (2 mL) at 0 °C. To the mixed solution was added potassium carbonate (246 mg, 1.78 mmol) at 0 °C. The reaction solution was stirred for 12 h at r. t. The mixture was extracted with chloroform, and the organic layer was washed with water twice, brine once, dried over anhydrous MgSO₄, filtered and concentrated under reduced pressure. This process gave 2-acetylnaphthalene as a clear oil. Yield: 68 mg (100 %).

¹H NMR (400 MHz, CDCl₃, δ): 8.02–7.25 (m, 7H, aromatic), 3.14 (s, 1H, acetylH).
**Compound 5**

To a 300 mL round bottom flask were added 2-bromoethylamine hydrobromide (10.1 g, 49.2 mmol) and Boc₂O (10.0 g, 45.9 mmol) in a minimum amount of CHCl₃. TEA (10.0 mL, 71.6 mmol) were added dropwise at 0 °C. The reaction mixture was stirred for 17 h at r. t. The reaction mixture was washed with 4 wt% KHSO₄ aq. and saturated NaHCO₃ aq. three times for each. The organic layer was washed with brine, dried over anhydrous MgSO₄, then filtered and concentrated under reduced pressure. The product was obtained as a clear oil. Yield: 7.31 g, (67 %).

¹H NMR (400 MHz, CDCl₃, δ): 4.95 (s, 1H, BocNCH₂CH₂Br), 3.52–3.43 (t, 4H, BocNHC₃H₂CH₂Br), 1.43 (s, 9H, Boc).

**Compound 6**

To a 200 mL two-necked round bottom flask were added 2,5-dibromohydroquinone (500 mg, 1.87 mmol), K₂CO₃ (780 mg, 5.64 mmol), KI (60 mg, 36.1 mmol), and 18-crown-6 (1.50 g, 5.60 mmol), dissolved with dry DMF (25 mL), and stirred at r. t. under argon atmosphere. To the mixed solution was added compound 5 (1.30 g, 5.80 mmol), and stirred at 100 °C under argon atmosphere for 6 h. The reaction mixture was washed with 4 wt% KHSO₄ aq. and saturated NaHCO₃ aq. three times for each. The organic layer was washed with brine, dried over anhydrous MgSO₄, filtered and concentrated under reduced pressure. The product was purified with column chromatography (CHCl₃/ethyl acetate = 100/1 v/v) and washed with hexane to afford compound 6 as a white solid. Yield: 580 mg, (56 %).

¹H NMR (400 MHz, CDCl₃, δ): 7.10 (s, 2H, aromatic), 5.04 (s, 2H, BocNCH₂CH₂), 4.03–4.00 (t, 4H
BocNHCH₂CH₂), 3.56–3.54 (m, 4H, BocNHCH₂CH₂), 1.46 (s, 18H, Boc)

**Compound 7 BocONE**

2-Acetylnaphthalene (2.47 g, 16.2 mmol) in THF solution was added to a mixed solution of CuI (206 mg, 1.08 mmol), compound 6 (3 g, 5.41 mmol), and Pd(PPh₃)₂Cl₂ (760 mg, 1.08 mmol) dissolved in dry THF (10 mL). DIEA (15.1 mL, 86.6 mmol) was added dropwise to the reaction mixture at r. t. The reaction mixture was stirred for 48 h at 60 °C. The reaction mixture was concentrated under reduced pressure and dissolved in chloroform, then washed with saturated NH₄Cl aq., 4 wt% KHSO₄ aq., saturated NaHCO₃ aq. three times for each, and brine once. The organic layer was dried over anhydrous MgSO₄ for 15 min, filtered and concentrated under reduced pressure. The product was purified by recrystallization with ethyl acetate and chloroform. Then the crude product was washed with ethyl acetate and hexane to afford the desired product as a yellow solid. Yield: 2.01 g, (53 %).

¹H NMR (400 MHz, CDCl₃, δ): 8.08–7.11 (m, 16H, aromatic), 5.23 (s, 2H, NHCOO), 4.19–4.17 (t, 4H, NHCH₂CH₂), 3.64–3.63 (q, 4H, NHCH₂CH₂), 1.40 (s, 18H, Boc).

**Compound 8**

The Boc group of compound 7 (77.0 mg, 111 µmol) was deprotected by treating with 4 N HCl/1,4-dioxane (25.0 mL). The reaction mixture was removed under reduced pressure. The crude product was washed with diisopropyl ether and concentrated under reduced pressure to obtain deprotected compound 7. The deprotected compound 7 (70 mg, 123 µmol) and Boc(Leu-Aib)₄OH (448 mg, 492 µmol) were dissolved in a minimum amount of dry DMF. The mixed solution of HATU (210 mg, 553 µmol) and HOAt (75.3 mg, 553 µmol) in a minimum amount of dry DMF
and DIEA (171 μL, 983 μmol) were added in this order under argon atmosphere, and the reaction mixture was stirred for 48 h at r. t. The solvent was removed under reduced pressure. The residue was dissolved in chloroform, and the solution was washed with 4 wt% KHSO$_4$ aq. and saturated NaHCO$_3$ aq. three times for each. The organic layer was washed with brine, dried over anhydrous MgSO$_4$, then filtered and concentrated under reduced pressure. The residue was washed with diisopropyl ether three times. The residue was purified with silica gel column chromatography (CHCl$_3$/MeOH = 1/0 to 10/1 v/v) to obtain compound 8 as a yellow solid. Yield: 128 mg (46 %).

$^1$H NMR (400 MHz, CDCl$_3$, δ): 7.98–7.04 (16H, aromatic), 5.92 (s, 2H, urethane), 4.42–3.79 (16H, Leu-$^8$H, NHCHOH$_3$CH$_2$O), 1.93–1.26 (m, 90H, Aib-$^9$H, Leu-$^{10}$H, Leu-$^{11}$H, Boc), 0.92–0.78 (m, 48H, Leu-$^{12}$H).

**Compound 9 (GS29L8ONE)**

The Boc group of compound 8 (80.0 mg, 35.0 μmol) was deprotected by treating with 4 N HCl/1,4-dioxane (25.0 mL). The reaction mixture was removed under reduced pressure. The crude product was washed with diisopropyl ether and concentrated under reduced pressure. The residue was dissolved in chloroform and washed with saturated NaHCO$_3$ aq. The organic phase was dried over anhydrous Na$_2$SO$_4$ and the solvent was removed to obtain deprotected compound 8. The deprotected compound 8 was dissolved in DMF (2 mL) and was added to the solution of Sar-NCA (100 mg, 869 μmol) in DMF (3 mL) at r. t. under argon atmosphere, then the reaction mixture was stirred for 12 h. After stirring the reaction mixture, glycolic acid (54.8 mg, 720 μmol), HATU (410 mg, 1.08 mmol) and HOAt (148 mg, 1.08 mmol) in a minimum amount of dry DMF and DIEA (282 μL, 1.62 mmol) were added in this order under argon atmosphere, and the reaction mixture was stirred for 24 h at r. t. The solvent was removed under reduced
pressure, and the residue was purified by a Sephadex LH20 column with methanol as eluent twice. The residue was dissolved in milliQ water and purified with ultrafiltration seven times. Length of Sar unit was determined by MALDI-TOF-MS and NMR. Yield: 40.0 mg (34 %)

$^1$H NMR (400 MHz, CDCl$_3$/MeOH-d$_4$ 10/1 v/v, δ): 8.04–7.06 (32H, amide NH, aromatic), 4.4–3.9 (br, 132H, Leu-C$^a$H, Sar-C$^a$H$_2$, NHCH$_2$CH$_2$O), 3.1–2.9 (br, 173H, Leu-C$^a$H, Sar-C$^a$H$_2$, NHCH$_2$CH$_2$O), 1.9–1.3 (m, 72H, Aib-C$^b$H, Leu-C$^b$H, Leu-C$^7$H), 0.93–0.80 (m, 48H, Leu-C$^9$H).

MALDI-MS (m/z): [M+Na]$^+$ calcd for C$_{292}$H$_{466}$N$_{76}$O$_{80}$Na, 6340.47; found, 6347.084.
Figure S1. $^1$H-NMR spectrum of S29L8ONE in CDCl₃/MeOH d₄ (10/1 v/v).

Figure S2. MALDI-TOF mass data of S29L8ONE.
Figure S3. HPLC chromatogram of S29L8ONE monitored by PDA detector (left axis: intensity of 254 nm, right axis: intensity of 400 nm). The measurements were carried out under following conditions (column: Shodex OH pak SB-803 HQ, eluent: methanol, column temperature: 40 °C, flow rate: 1.0 mL/min). The purity of S29L8ONE was evaluated based on the peak areas and confirmed to be >95%.
**Preparation of molecular assemblies**  A mixture of S29L8ONE (0.6 mg) and BocONE (0.066 mg/eq.) in 10 μL DMF solution was injected into 1 mL MilliQ water (S29L8ONE: 9.5 × 10^{-2} mM, BocONE: 9.5 × 10^{-2} mM/eq.) under stirring at 4 °C. After 30 min stirring, the dispersions were heated at 50 °C for 1 h and further heat treatment was carried out at 90 °C for 24 h.

**Transmission Electron Microscopy (TEM) and Electron Diffraction.** The TEM images and diffraction patterns were obtained by a transmission electron microscope (JEOL, JEM-2000EXII) at an accelerating voltage of 100 kV. The molecular assemblies were mounted on a glow-discharged thin carbon coated Cu grid, and thereafter excess of the suspension was sucked by a filter paper. The samples were then stained with 2% uranyl acetate solution.

The electron diffraction diagrams were obtained in the microdiffraction mode. A small condenser aperture of 20 μm was inserted in the second condenser lens, and the first condenser lens was fully excited to obtain an electron probe of ca. 200 nm at the sample level. An apparent single crystal on the grid was searched in an extremely low dose condition with the help of an image intensifier (Gatan Pleasanton, Fiber Optics Coupled TV) to avoid irradiation damage by electrons prior to recording. Diffraction diagrams were recorded on a photographic emulsion film (Mitsubishi paper milling, MEM film), and then developed by Copinal (FUJIFILM Corp., Japan). Camera length of diffraction mode was calibrated with 111 diffraction ring of evaporated gold particles.

**Atomic Force Microscopy (AFM).** Si wafers were cleaned by 2% hydrofluoric acid, and soaked in piranha solution, then treated with 1% 3-aminopropyltriethoxysilane (APTES) toluene solution at 60 °C for 10 min. AFM measurements of self-assemblies in an aqueous solution were carried out with Bruker Multimode 8 AFM in PeakForce Fluid tapping mode using gold coated silicon nitride cantilevers (PeakForce Fluid, 0.7 N/m, Bruker) on
an APTES modified Si wafer. The molecular assembly dispersion was incubated into a fluid liquid cell on an APTES modified Si wafer for 1 h. Then, the dispersion in a fluid liquid cell was substituted by MilliQ water.
Figure S4. AFM measurements of the molecular assemblies prepared from S29L8ONE (9.5 × 10^{-2} mM) with heat treatment at 50 °C for 1 h. (A) AFM image on APTES modified silicon wafer. (B) AFM height profile (C) Molecular assembly height histogram. (D) AFM image on APTES modified silicon wafer (4 × 4 μm).
Figure S5. Negatively stained TEM image with 2% uranyl acetate of molecular assemblies prepared from S29L8ONE (9.5 × 10^{-2} mM) with heating treatment at 90 °C for 24 h.

Figure S6. AFM image and heigh profile of molecular assemblies prepared from a mixture of S29L8ONE (9.5 × 10^{-2} mM) and BocONE (1/2 mol/mol) on APTES modified silicon wafer.
UV-vis spectroscopy. The absorption spectra were recorded on a Shimadzu UV-2450PC spectrometer. The molecular assembly dispersions were diluted by a factor of 10.

Fluorescence spectroscopy. The emission spectra were measured on a Jasco FP-6600 fluorometer. The molecular assembly dispersions were diluted by a factor of 100. Emission spectra were measured at 365 nm for excited wavelength. Excitation spectra were monitored at 418.4 nm for monomer emission and at 500 nm for excimer emission.

Circular Dichroism (CD) Spectroscopy. CD spectra were measured on a CD spectropolarimeter (J-1500, JASCO, Tokyo). The molecular assembly dispersions were diluted by a factor of 5. The measurements were carried out with an optical cell of 0.1 cm optical path length.
Figure S7. Spectroscopic data of molecular assemblies; UV-vis spectra (A, E, I, M), emission spectra excited at 365 nm (B, F, J, N), excitation spectra monitored at 418.4 nm (C, G, K, O), and monitored at 500 nm (D, H, L, P). Molecular assemblies prepared from a mixture of S29L8ONE ($9.5 \times 10^{-2} \text{ mM}$) and BocONE ($9.5 \times 10^{-2} \text{ mM/eq.}$) with heat treatment at 50 °C for 1 h (A-D), with heat treatment at 90 °C for 24 h (E-H). (I-L) Spectroscopic data of BocONE in MilliQ water. The concentrations of the dispersion of BocONE in MilliQ were $9.5 \times 10^{-2} \text{ mM/eq.}$ (M-P) Summary of the data.
Reference.