Supporting Information

Ring-shaped self-assembly of naphthalene-linked chlorophyll dimer

Tatsuma Ishii[1], Shogo Matsubara[1,2]*, Hitoshi Tamiaki[1]*

[1] Graduate School of Life Sciences, Ritsumeikan University, Kusatsu, Shiga, 525-8577, Japan
[2] Graduate School of Engineering, Nagoya Institute of Technology, Nagoya, Aichi, 466-8555, Japan

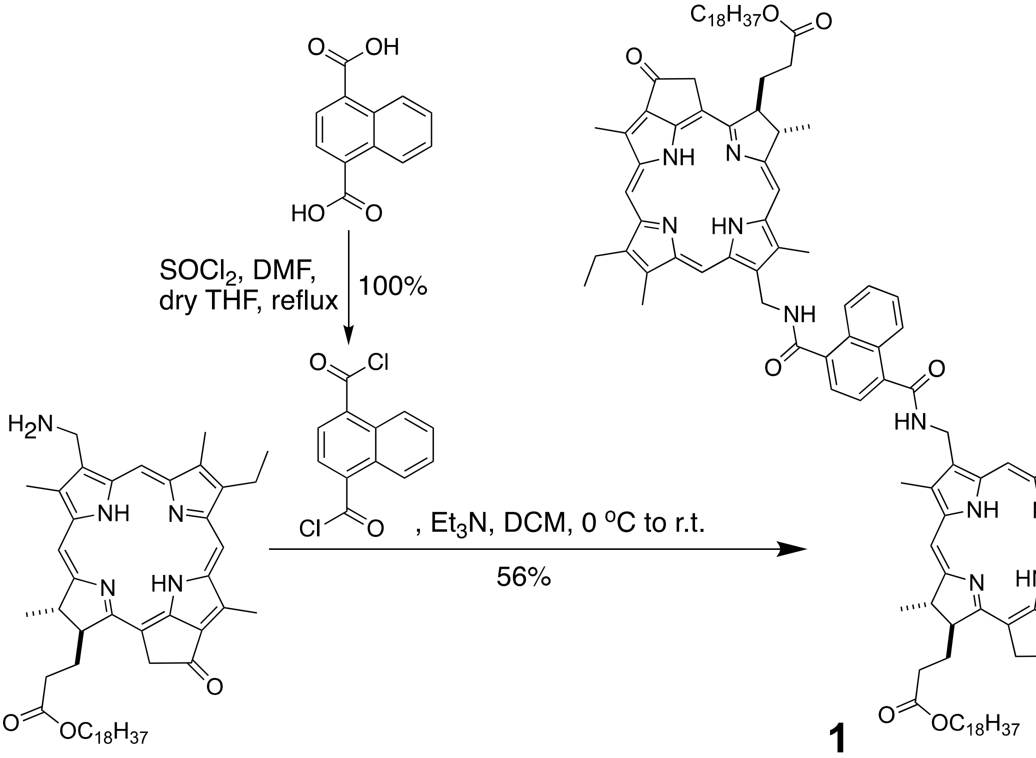
Corresponding Authors
E-mail: matsubara.shogo@nitech.ac.jp (S. Matsubara)
tamiaki@fc.ritsumei.ac.jp (H. Tamiaki)

1. Experimental

1-1. General

All the reactions were performed in the dark under nitrogen or argon atmosphere and monitored by thin layer chromatography and UV-vis absorption spectroscopy. UV-vis absorption and CD spectra were measured in solution with a 10 mm quartz cell at room temperature using a Hitachi U-3500 spectrophotometer, a Jasco J-720W spectropolarimeter, respectively. FT-IR spectra were recorded on a Shimadzu IRAffinity-1 spectrometer using a KBr cell. ^{1}H NMR spectrum was recorded in $CDCl_3$ using a JEOL ECA-600 (600 MHz) spectrometer. $CHCl_3$ ($\delta_H = 7.26$ ppm) was used as internal standards. High-resolution mass spectra (HRMS) were recorded on a Bruker micrOTOF II spectrometer; Electrospray ionization (ESI) and positive mode in a $CHCl_3$ solution. AFM measurements were performed by a Bruker AXS Multimode 8 AFM system in tapping mode in air at room temperature on an HOPG substrate, and a Bruker silicon cantilever RTESP-300 was used. Dynamic light scattering (DLS) measurements were performed by a Malvern Zetasizer Nano ZS instrument.

1-2. Synthesis of chlorophyll dimer 1



A mixture of 1,4-naphthalenedicarboxylic acid (40.3 mg, 186 μmol), $SOCl_2$ (100 μL, 1.38 mmol), and a catalytic amount of *N,N*-dimethyl formamide (DMF) in 10 mL of dry tetrahydrofuran (THF) was refluxed for 3 h. After cooling down to room temperature, $SOCl_2$, DMF, and THF were removed *in vacuo* to give 1,4-naphthalenedicarboxyl dichloride (46.5 mg, 184 μmol, 100%). The product (4.7 mg, 18.6 μmol) was dissolved in 10 mL of dichloromethane (DCM), to which were added to stearyl 3-aminomethyl-pyropheophorbide-*a*[S1] (31.1 mg, 39.4 μmol) and Et_3N (26.4 μL, 189 μmol) at 0 °C. After stirring for 1 day at room temperature, the reaction mixture was quenched by addition of a water, extracted by DCM, washed with water, and dried over Na_2SO_4. After evaporating the solvent, the residue was purified by silica gel chromatography to give **1** as a black solid (18.3 mg, 10.4 μmol, 56%); mp >300 °C; UV-vis ($CHCl_3$) λ_{max} = 666 (relative intensity, 0.54), 609 (0.08), 538 (0.09), 508 (0.10), 412 (1.00), 400 (0.82), 381 (0.60), 319 nm (0.23); ^1H NMR ($CDCl_3$, 50 °C, 600 MHz) δ_H/ppm = 9.22 (2H, s, 5-H×2), 9.17 (2H, s, 10-H×2), 8.38 (2H, s, 20-H×2), 8.32/8.31 (2H, d, *J* = 6 Hz, 5,8-H of naphthalene), 7.338/7.343 (2H, d, *J* = 6 Hz, 6,7-H of naphthalene), 7.29 (2H, s, 2,3-H of naphthalene), 6.73 (2H, br, 3^1-NH×2), 5.50 (4H, br, 3-CH_2×2), 5.18, 5.03 (each 2H, d, *J* = 18 Hz, 13^1-CH_2×2), 4.33 (2H, br-q, *J* = 7 Hz, 18-H×2), 4.13 (2H, br-d, *J* = 9 Hz, 17-H×2), 4.04–3.94 (4H, m, 17^2-$COOCH_2$×2), 3.44 (4H, q, *J* = 8 Hz, 8-CH_2×2), 3.39 (6H, s, 12-CH_3×2), 3.21 (6H, s, 2-CH_3×2), 3.02 (6H, s, 7-CH_3×2), 2.59–2.50, 2.49–2.42, 2.26–2.16, 2.11–2.00 (each 2H, m, 17-CH_2CH_2×2), 1.70 (6H, d, *J* = 7 Hz, 18-CH_3×2), 1.51 (6H, t, *J* = 8 Hz, 8^1-CH_3×2), 1.30–1.19 (60H, m, 17^2-$COOC_2(CH_2)_{15}$×2), 0.87 (6H, t, *J* = 7 Hz, 17^2-$COOC_{17}CH_3$×2), −2.02 (2H, br-s, NH×2) [The proton signals of the 17^2-$COOCCH_2$ and the other NH in macrocycle were overlapped with proton peaks of water and tetramethylsilane, respectively.]; ^{13}C NMR ($CDCl_3$, 50 °C, 151 MHz) δ_C/ppm = 195.6, 173.0, 171.2, 168.5, 154.9, 151.0, 148.9, 145.0, 142.8, 136.6, 133.7, 133.5, 130.8, 130.5, 128.6, 127.6, 127.3, 125.7, 123.8, 106.4, 104.0, 100.8, 100.7, 97.9, 96.5, 93.0, 64.9, 51.9, 49.9, 48.0, 34.8, 31.9, 31.3, 29.8, 29.7, 29.63, 29.60, 29.55, 29.47, 29.41, 29.33, 29.19, 28.6, 25.9, 23.0, 22.7, 19.5, 19.4, 17.2, 14.0, 11.8, 11.13, 11.10 [Three ^{13}C peaks in the 17^2-octadecyl moiety were overlapped with the others.]; HRMS (ESI) found: *m/z* = 1761.1429 and 881.0752, calcd for $C_{112}H_{146}N_{10}O_8$: $[M+H]^+$, 1761.1432, $[M+H_2]^{2+}$, 881.0752.

1-3. Preparation of the samples for UV-vis absorption and CD spectroscopies

Twenty nmol of chlorophyll dimer **1** was dissolved in $CHCl_3$ (2000 or 100 μL) to give low- or high-concentrated solution, respectively. The low-concentrated $CHCl_3$ solution (10 μM) was used as the sample of monomeric **1** for UV-vis absorption and CD spectroscopies. The high-concentrated $CHCl_3$ solution of **1** (100 μL) was diluted with 1900 μL of hexane to give the sample of assembled **1** in 5%(v/v) $CHCl_3$–hexane (10 μM).

1-4. Preparation of the samples for FT-IR spectroscopy

Similarly as mentioned in section 1-3, 1 mM solution and dispersion of **1** in $CHCl_3$ and 5%(v/v) $CHCl_3$–hexane, respectively, were prepared. The $CHCl_3$ solution and 5%(v/v) $CHCl_3$–hexane dispersion were injected to a sealed liquid cell with KBr windows to give the samples of monomeric and assembled **1** for FT-IR spectroscopy, respectively.

1-5. Preparation of the AFM samples

Similarly as mentioned in section 1-3, 1, 10, and 100 μM dispersion of assembled **1** in 5%(v/v) $CHCl_3$–hexane were prepared. The 5%(v/v) $CHCl_3$–hexane dispersions were casted on an HOPG substrate and spin-coated using a MIKASA Opticoat MS-B 100 (2000 rpm) to give the AFM sample.

2. Spectral data of 1

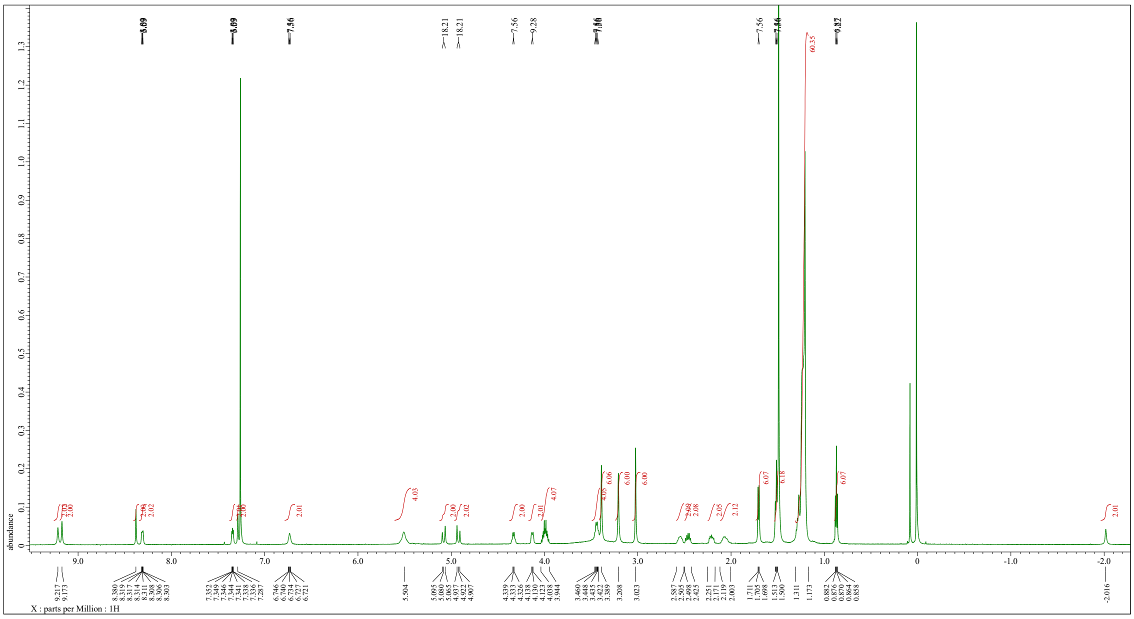


Figure S1. ^{1}H NMR spectrum of **1** in CDCl$_3$ at 50 ^{o}C.

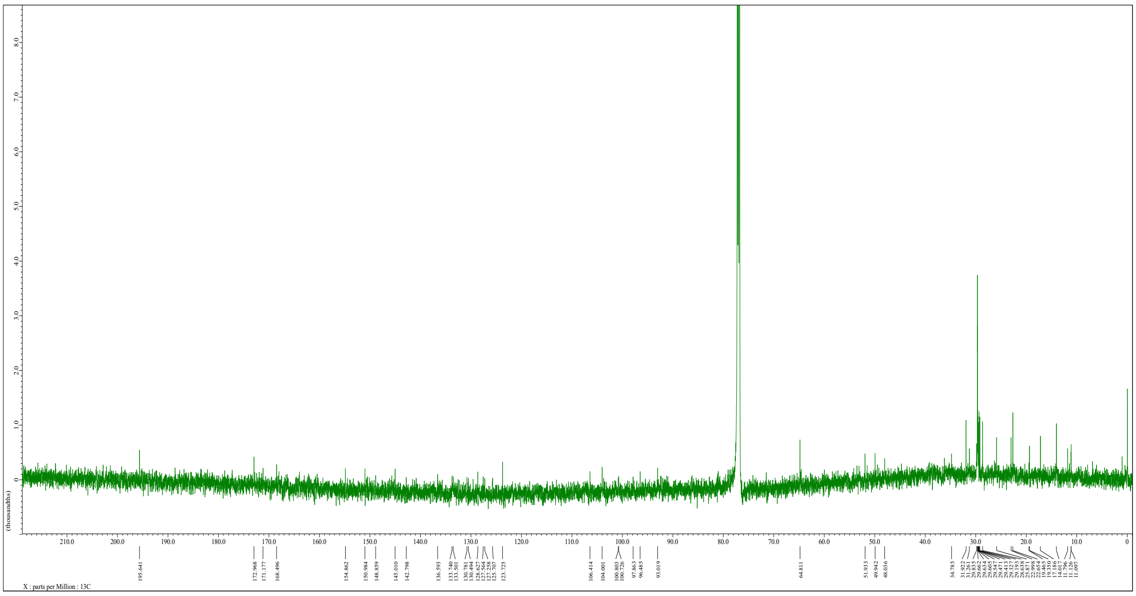


Figure S2. ^{13}C NMR spectrum of **1** in CDCl$_3$ at 50 ^{o}C.

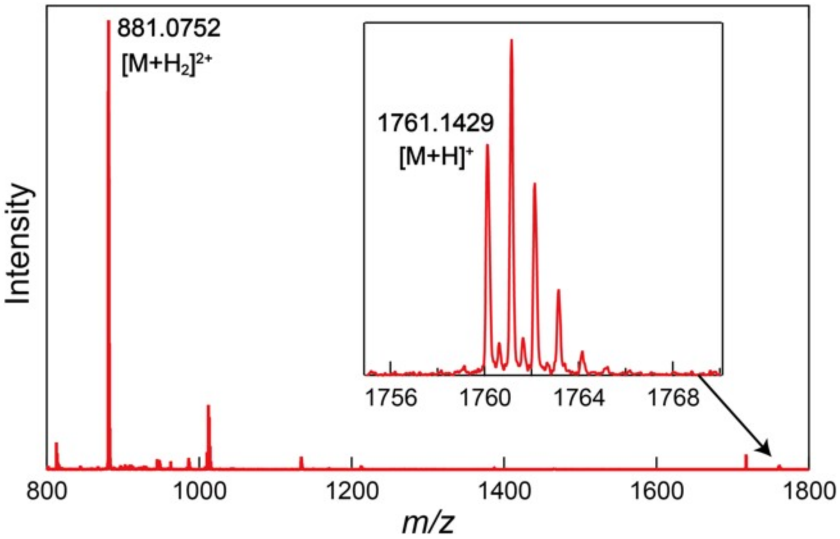


Figure S3. ESI-TOF mass spectrum of **1**.

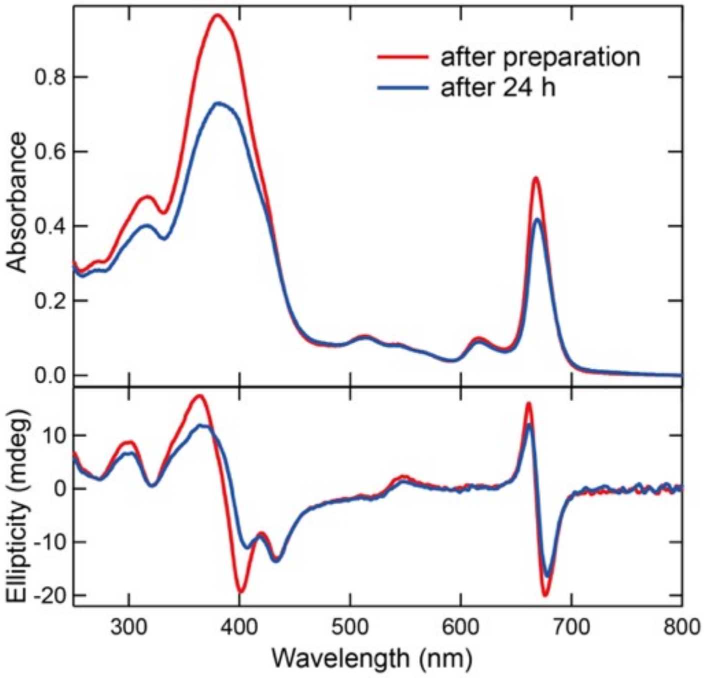


Figure S4. UV-vis absorption (upper) and CD spectra (lower) of **1** (10 µM) in 5%(v/v) $CHCl_3$–hexane just after preparation (red) and after standing for 24 h (blue).

3. Size of the nanorings

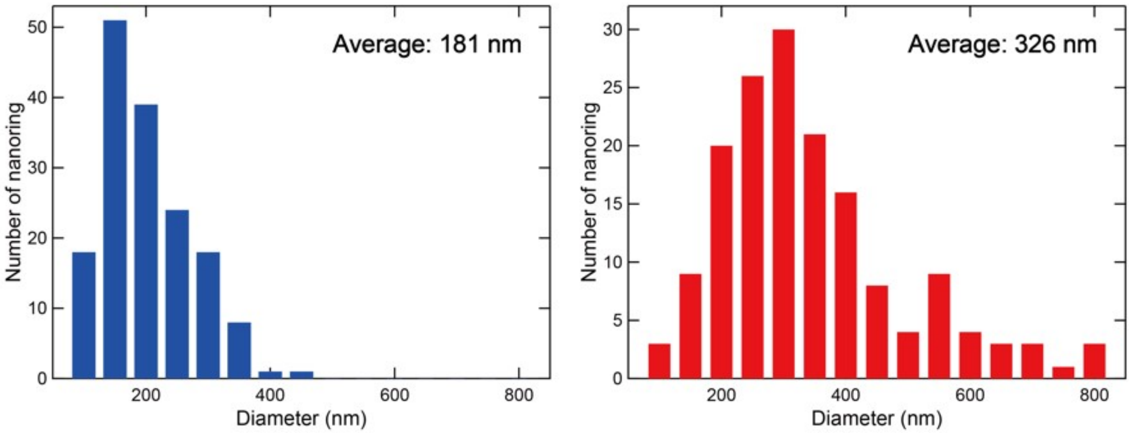


Figure S5. The distribution histograms of the short (left, blue) and long axes of the nanorings (right, red) evalated by AFM images.

Table S1. The numbers of nanofibers and nanorings counted from each two AFM images (5 × 5 μm) of assembled dimer **1** after standing the sample for 0, 3, 6, 12, and 24 h and the number ratios of nanorings over nanostructures observed.

Sample standing hour	Number of nanofibers	Number of nanorings	Ratio of nanorings over nanostructures
0	113	3	3%
3	133	22	14%
6	113	29	20%
12	56	44	44%
24	45	64	59%

4. Additional AFM images

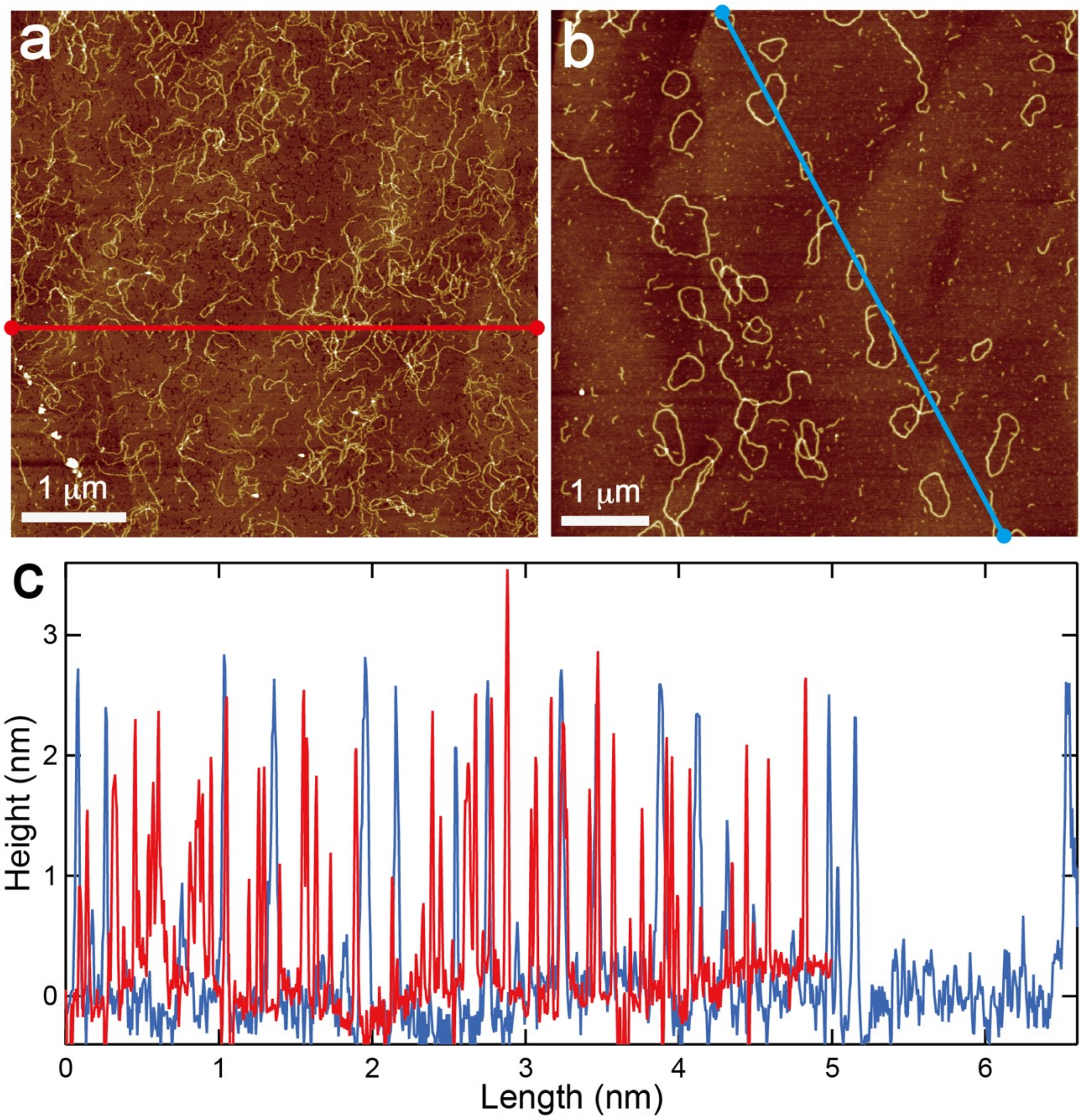
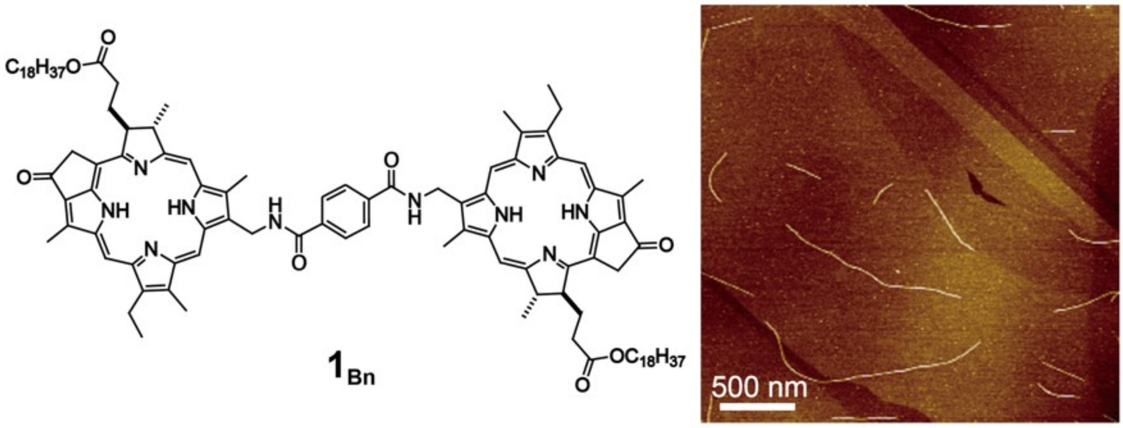


Figure S6. AFM images of nanofibers (a) and nanorings (b). The concentration of **1** in AFM samples was 10 μM. All the white bars show a 1 μm length. (c) Height profiles along the red-line in Figure S6a and blue-line in Figure S6b.



$\mathbf{1}_{Bn}$

Figure S7. Chemical structure of benzene-liked chlorophyll dimer $\mathbf{1}_{\mathbf{Bn}}$ (left) and AFM image of assembled $\mathbf{1}_{\mathbf{Bn}}$ on an HOPG substrate (right).

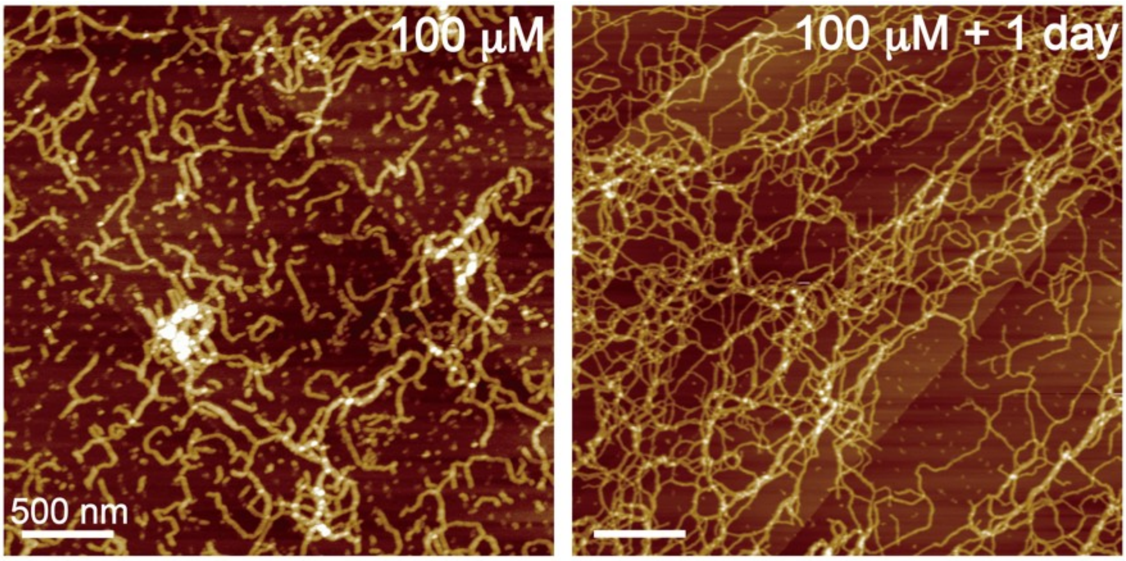


Figure S8. AFM images of assembled dimer **1** (100 μM) after standing the suspension for 0 h (left) and 1 day (right). All the bars show a 500 nm length.

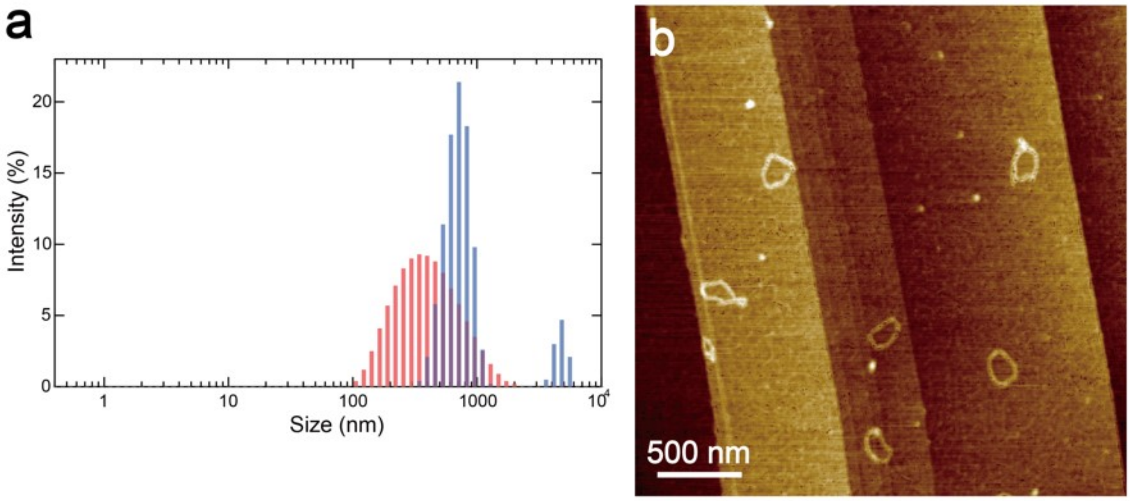


Figure S9. (a) DLS size distributions of self-aggregates of **1** (10 μM) before (blue) and after filtration using a membrane filter with a 0.45 μm pore (red). (b) AFM image of 1-day stood dispersion of assembled **1** (10 μM) after filtration by using a membrane filer with a 0.45 μm pore.

4. Reference

S1. S. Matsubara, H. Tamiaki, Supramolecular Nanofibers Constructed by Hydrogen Bonding of Chlorophyll Dimer. *Chem. Lett.*, 2021, **50**, 999-1001.