Electronic Supplementary Information for

Efficient "*turn-off*" fluorescence photoswitching in a highly fluorescent diarylethene single crystal

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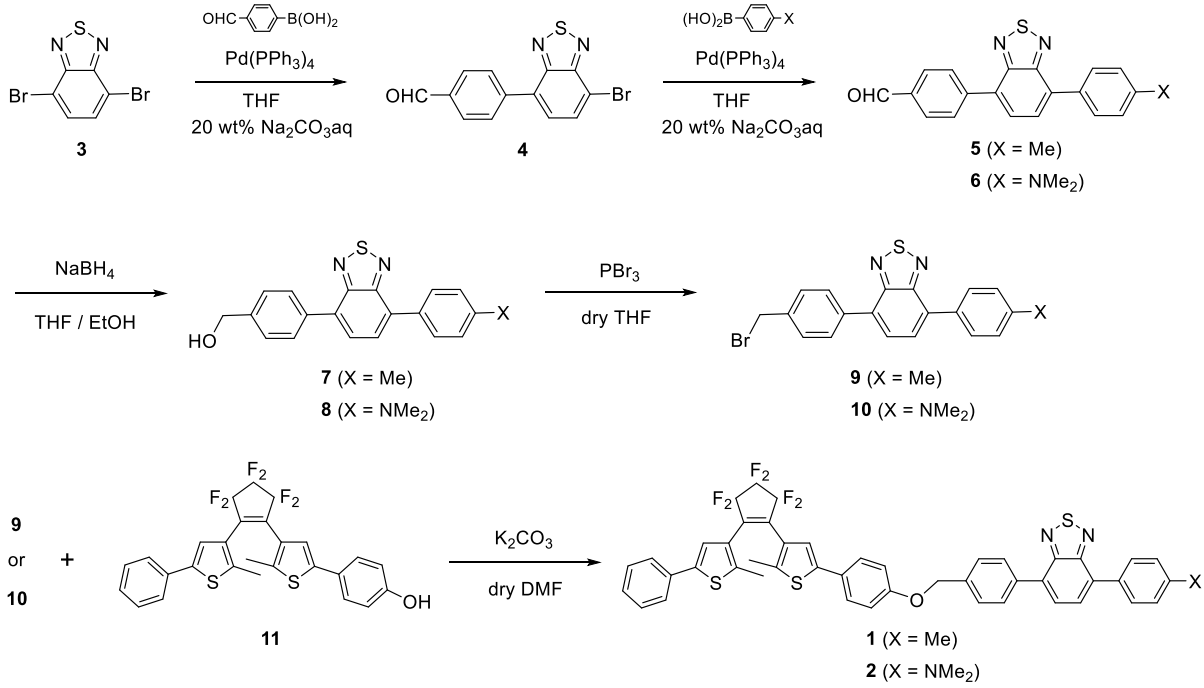
1. General

General chemicals were purchased from Tokyo Chemical Industry, Wako Pure Chemicals, or Sigma Aldrich Chemical Co., and used without further purification. ^{1}H NMR spectra were recorded on JEOL JNMEX400 spectrometer with tetramethylsilane (TMS) as the internal standard. Mass spectra were measured with a mass spectrometer (Autoflex Speed, Bruker). UV-vis absorption spectra were recorded on a Hitachi U-3310 spectrophotometer. Fluorescence spectra were measured with a Hitachi F-7000 fluorescence spectrophotometer. Fluorescence quantum yields were determined with an absolute photoluminescence quantum yield spectrometer (Hamamatsu, C9920-02). Irradiation experiments were carried out in a quartz cuvette using a Xe lamp (MAX-303, Asahi Spectra) equipped by narrow band interference filters (Semrock) or a monochromator (Horiba JobinYvon). Single crystal X-ray crystallographic analysis was conducted using a Rigaku AFC/Mercury CCD diffractometer with MoK_α radiation (λ = 0.71073 Å) monochromated by graphite. The crystal structures were solved by a direct method using SIR 92 and refined by the full-matrix least-squares method for F^2 with anisotropic displacement parameters for non-hydrogen atoms using SHELXL-2014. The isolation of the photo-generated closed-ring isomer was performed with HPLC (HITACHI ELITE LaChrom system) equipped with HITACHI L-2455 diode array detector, silica gel column (Wako, Wakosil-5SIL), using hexane/dichloromethane (70:30 in volume) as the eluent. Polarized absorption and fluorescence spectra of the single crystals were measured with a polarizing microscope (Leica, DM2500P and Nikon, LV-100) and a CCD-based PMA-11 photodetector (Hamamatsu Photonics, C7473). UV and visible irradiation to the crystals was carried out with a UV-LED irradiation system (Hamamatsu, LIGHTNINGCURE, LC-LIV3) and a 300 W xenon lamp (Asahi Spectra, MAX-303). Fluorescence photoswitching of single crystals was demonstrated using an inverted optical microscope (Nikon, Ti-E) equipped with an objective lens (Nikon, 20x, 0.45 NA) and appropriate filters (Chroma, Semrock). Fluorescence images were recorded using a digital CMOS camera (ORCA-Flash 4.0-V3.0, Hamamatsu) and the HCI Image processing system (Hamamatsu). The fluorescence signals were measured with an exposure time of 30 ms. 438-nm light from a mercury lamp was used for the excitation as well as for the photocycloreversion reaction. 390 nm UV light (100 $\mu W/cm^2$) from light emitting diode (Lumencor Light engine) was introduced into the microscope and used for the photocyclization reaction.

2. Synthesis

The synthetic route to **1** and **2** are illustrated in Scheme 1. Detailed synthetic procedures of these molecules were described below. Compounds **4**[S1] and **11**[S2] were prepared according to literature procedures.



Scheme S1 Synthetic scheme of **1** and **2**.

Synthesis of 5

Compound **4** (400 mg, 1.3 mmol) and *p*-tolylboronic acid (170 mg, 1.25 mmol) were dissolved in a mixture of THF (48 mL) and 20 wt% Na_2CO_3 aqueous solution (48 mL), and then tetrakis(triphenylphosphine)palladium(0) ($Pd(PPh_3)_4$, 125 mg, 0.11 mmol) was added. The solution was refluxed for 20 h under Ar atmosphere. After being cooled, the reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (hexane/dichloromethane = 1/3) to give 380 mg (1.2 mmol) of **5** in 92% yield as a yellow solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 2.45 (s, 3H), 7.37 (d, *J* = 8 Hz, 2H), 7.79 (d, *J* = 8 Hz, 1H), 7.84-7.90 (m, 3H), 8.05 (d, *J* = 8 Hz, 2H), 8.16 (d, *J* = 8 Hz, 2H), 10.11 (s, 1H); MS (MALDI) m/z = 330.57 $[M]^+$ (Exact Mass: 330.08); Elemental analysis: Found: C, 72.67; H, 4.31; N. 8.46. Anal. Calcd. for $C_{20}H_{14}N_2OS$: C, 72.70; H, 4.27; N, 8.48.

Synthesis of 6

Compound **4** (320 mg, 1.0 mmol) and (4-(dimethylamino)phenyl)boronic acid (200 mg, 1.2 mmol) were dissolved in a mixture of THF (38 mL) and 20 wt% Na_2CO_3 aqueous solution (38 mL), and then tetrakis(triphenylphosphine)palladium(0) ($Pd(PPh_3)_4$, 100 mg, 87 μmol) was added. The solution was refluxed for 20 h under Ar atmosphere. After being cooled, the reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (hexane/dichloromethane = 1/3) to give 300 mg (0.83 mmol) of **6** in 83% yield as an orange solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 3.05 (s, 6H), 6.89 (d, *J* = 8 Hz, 2H), 7.75 (d, *J* = 8 Hz, 1H), 7.83 (d, *J* = 8 Hz, 1H), 7.95 (d, *J* = 8 Hz, 2H), 8.04 (d, *J* = 8 Hz, 2H), 8.16 (d, *J* = 8 Hz, 2H), 10.1 (s, 1H); MS (MALDI) m/z = 359.61 $[M]^+$ (Exact Mass: 359.11); Elemental analysis: Found: C, 70.11; H, 4.78; N. 11.72. Anal. Calcd. for $C_{21}H_{17}N_3OS$: C, 70.17; H, 4.77; N, 11.69.

Synthesis of 7

Compound **5** (380 mg, 1.2 mmol) was dissolved in a mixture of THF (30 mL) and ethanol (15 mL). $NaBH_4$ (50 mg, 1.3 mmol) was added into the mixture and stirred for 10 min at room temperature. The reaction was stopped by the addition of dilute HCl aqueous solution. The reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (dichloromethane) to give 370 mg (1.2 mmol) of **7** in ~99% yield as a light yellow solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 2.45 (s, 3H), 4.80 (d, *J* = 8 Hz, 2H), 7.35 (d, *J* = 8 Hz, 2H), 7.55 (d, *J* = 8 Hz, 2H), 7.73-7.79 (m, 2H), 7.85 (d, *J* = 8 Hz, 2H), 7.96 (d, *J* = 8 Hz, 2H); MS (MALDI) m/z = 332.67 $[M]^+$ (Exact Mass: 332.10); Elemental analysis: Found: C, 72.37; H, 4.78; N. 8.54. Anal. Calcd. for $C_{20}H_{16}N_2OS$: C, 72.26; H, 4.85; N, 8.43.

Synthesis of 8

Compound **6** (300 mg, 0.83 mmol) was dissolved in a mixture of THF (25 mL) and ethanol (5 mL). $NaBH_4$ (40 mg, 1.1 mmol) was added into the mixture and stirred for 10 min at room temperature. The reaction was stopped by the addition of dilute HCl aqueous solution. The reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (dichloromethane) to give 300 mg (0.83 mmol) of **8** in ~99% yield as an orange solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 3.04 (s, 6H), 4.79 (d, *J* = 8 Hz, 2H), 6.89 (d, *J* = 8 Hz, 2H), 7.54 (d, *J* = 8 Hz, 2H), 7.69-7.77 (m, 2H), 7.9-7.98 (m, 4H); MS (MALDI) m/z = 361.76 $[M]^+$ (Exact Mass: 361.12); Elemental analysis: Found: C, 69.67; H, 5.28; N. 11.62. Anal. Calcd. for $C_{21}H_{19}N_3OS$: C, 69.78; H, 5.30; N, 11.63.

Synthesis of 9

Compound **7** (370 mg, 1.1 mmol) was dissolved in dry THF (100 mL) at 0 ºC under Ar atmosphere. 1M PBr_3 in dichloromethane solution (1.3 mL, 1.3 mmol) was slowly added into the reaction mixture and stirred for 1h at this temperature. After warming up to room temperature, the mixture was stirred for 1 h. The reaction was stopped by the addition of water. The reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (dichloromethane) to give 300 mg (0.76 mmol) of **9** in 68% yield as a light yellow solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 2.45 (s, 3H), 4.59 (s, 2H), 7.36 (d, *J* = 8 Hz, 2H), 7.57 (d, *J* = 8 Hz, 2H), 7.73-7.80 (m, 2H), 7.85 (d, *J* = 8 Hz, 2H), 7.95 (d, *J* = 8 Hz, 2H); MS (MALDI) m/z = 394.70 $[M]^+$ (Exact Mass: 394.01); Elemental analysis: Found: C, 60.69; H, 3.85; N. 7.03. Anal. Calcd. for $C_{20}H_{15}BrN_2S$: C, 60.77; H, 3.82; N, 7.09.

Synthesis of 10

Compound **8** (170 mg, 0.46 mmol) was dissolved in dry THF (50 mL) at 0 ºC under Ar atmosphere. 1M PBr_3 in dichloromethane solution (0.55 mL, 0.55 mmol) was slowly added into the reaction mixture and stirred for 1h at this temperature. After warming up to room temperature, the mixture was stirred for 1 h. The reaction was stopped by the addition of water. The reaction mixture was extracted with ethyl acetate. The organic layer was separated, washed with brine, dried over anhydrous sodium sulfate, and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (dichloromethane) to give 100 mg (0.24 mmol) of **10** in 52% yield as an orange solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 3.05 (s, 6H), 4.58 (s, 2H), 6.89 (d, *J* = 8 Hz, 2H), 7.55 (d, *J* = 8 Hz, 2H), 7.68-7.78 (m, 2H), 7.89-7.97 (m, 4H); MS (MALDI) m/z = 423.74 $[M]^+$ (Exact Mass: 423.04); Elemental analysis: Found: C, 69.65; H, 5.38; N. 11.67. Anal. Calcd. for $C_{21}H_{19}N_3OS$: C, 69.78; H, 5.30; N, 11.63.

Synthesis of 1

Compound **9** (100 mg, 0.25 mmol) was dissolved in dry DMF (9 mL), and then compound **11** (160 mg, 0.3 mmol) and K_2CO_3 (270 mg) were added into the solution. The reaction mixture was heated at 70 ºC for 3 h under Ar atmosphere. After being cooling, the reaction mixture was extracted with dichloromethane. The organic layer was separated, washed with NH_4Cl aqueous solution and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (hexane/dichloromethane = 1/1) to give 170 mg (0.2 mmol) of **1** in 78% yield as a light yellow solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 1.96 (d, *J* = 8 Hz, 6H), 2.45 (s, 3H), 5.19 (s, 2H), 7.02 (d, *J* = 8 Hz, 2H), 7.17 (s, 1H), 7.27-7.30 (m, 2H), 7.35-7.39 (m, 4H), 7.48 (d, *J* = 8 Hz, 2H), 7.54 (d, *J* = 8 Hz, 2H), 7.61 (d, *J* = 8 Hz, 2H), 7.75-7.79 (m, 2H), 7.86 (d, *J* = 8 Hz, 2H), 7.99 (d, *J* = 8 Hz, 2H); MS (MALDI) m/z = 851.04 $[M]^+$ (Exact Mass: 850.16); Elemental analysis: Found: C, 66.26; H, 3.78; N, 3.31. Anal. Calcd. for $C_{47}H_{32}F_6N_2OS_3$: C, 66.34; H, 3.79; N, 3.29.

Synthesis of 2

Compound **10** (100 mg, 0.24 mmol) was dissolved in dry DMF (9 mL), and then compound **11** (140 mg, 0.26 mmol) and K_2CO_3 (250 mg) were added into the solution. The reaction mixture was heated at 70 ºC for 4 h under Ar atmosphere. After being cooling, the reaction mixture was extracted with dichloromethane. The organic layer was separated, washed with NH_4Cl aqueous solution and filtrated. The solvent was removed by evaporation and the residue was purified by silica gel column chromatography (hexane/dichloromethane = 1/1) to give 190 mg (0.22 mmol) of **2** in 90% yield as a orange solid.

^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 1.96 (d, *J* = 8 Hz, 6H), 3.05 (s, 6H), 5.17 (s, 2H), 6.90 (d, *J* = 8 Hz, 2H), 7.02 (d, *J* = 8 Hz, 2H), 7.17 (s, 1H), 7.26-7.31 (m, 2H), 7.36-7.40 (m, 2H), 7.48 (d, *J* = 8 Hz, 2H), 7.54 (d, *J* = 8 Hz, 2H), 7.57 (d, *J* = 8 Hz, 2H), 7.71-7.76 (m, 2H), 7.94 (d, *J* = 8 Hz, 2H), 7.99 (d, *J* = 8 Hz, 2H),; MS (MALDI) m/z = 879.07 $[M]^+$ (Exact Mass: 879.18); Elemental analysis: Found: C, 65.46; H, 4.08; N, 4.77. Anal. Calcd. for $C_{48}H_{35}F_6N_3OS_3$: C, 65.51; H, 4.01; N, 4.78.

3. X-ray crystallographic analysis

Table S1 Crystallographic data for **1a**

	1a
Formula	$C_{47}H_{32}F_6N_2OS_3$
Formula weight	850.96
T/K	150
Crystal system	Triclinic
Space group	$P\bar{1}$
a/Å	11.551(2)
b/Å	12.862(3)
c/Å	15.172(3)
α/°	89.039(8)
β/°	68.064(6)
γ/°	71.586(7)
V/Å^3	1970.6(7)
Z	2
Density/g cm^{-3}	1.434
Goodness-of-fit on F^2	1.007
R_1 ($I > 2\sigma(I)$)	0.0358
wR_2 ($I > 2\sigma(I)$)	0.0890
R_1 (all data)	0.0517
wR_2 (all data)	0.0950
CCDC No.	1897359

4. Photochromism and fluorescence photoswitching of 1 in THF solution

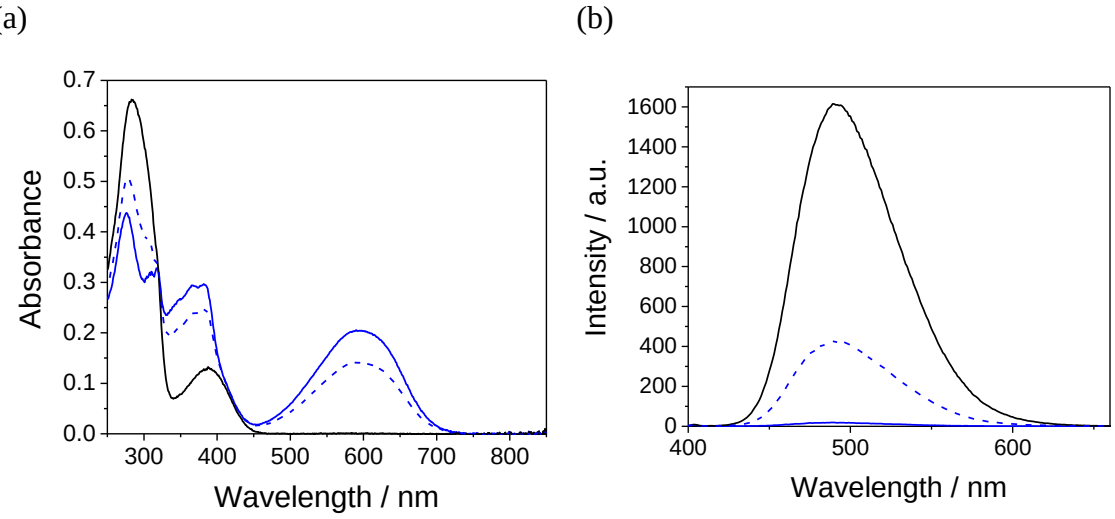


Figure S1 (a) Absorption and (b) fluorescence spectral changes of **1** in THF solution along with photocyclization and photocycloreversion reactions; the open-ring isomer (solid-black line), the closed-ring isomer (solid-blue line), and PSS under irradiation with 365 nm light (dashed-blue line).

5. Preparation of nanoparticles

Nanoparticle of **1** was prepared by the conventional reprecipitation method.[S3] The closed-ring isomer of **1** isolated by HPLC was dissolved into THF to obtain a 5.0×10^{-5} mol L^{-1} solution. 0.6 mL of this solution was quickly added into 2.4 mL of distilled water under vigorous stirring during 5 min. Final suspension of NPs was obtained in H_2O/THF mixture with a concentration of 1.0×10^{-5} mol L^{-1} and it was diluted too appropriate concentration before use.

6. Fluorescence photoswitching of 1 in a single crystal state

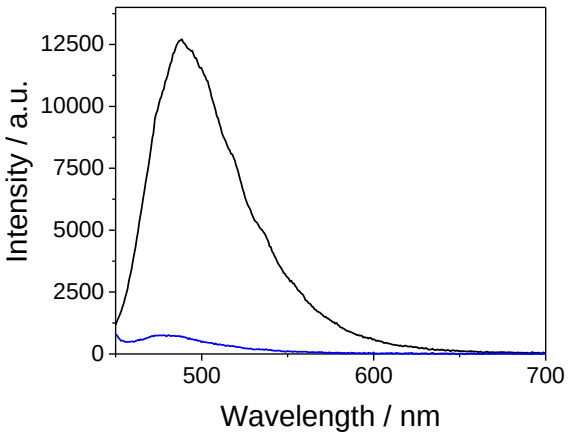


Figure S2 Fluorescence spectral changes of **1** in single crystal state; the open-ring isomer (solid-black line) and PSS under irradiation with 365 nm light (solid-blue line).

7. Fluorescence on/off photoswitching cycles of 1 in the single crystal state

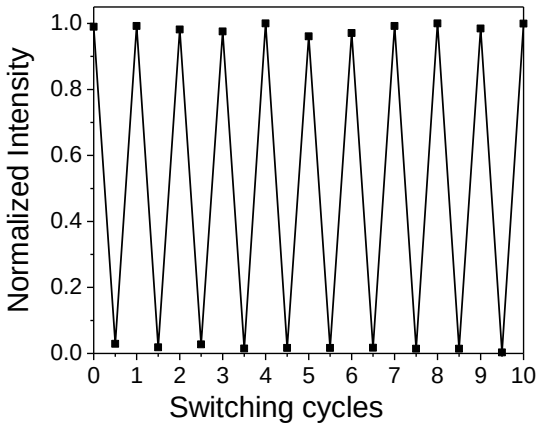


Figure S3 Peak fluorescence intensity of **1** in the single crystal state over multiple cycles of UV (365 nm) and visible (490 nm) lights irradiation.

8. Polarized absorption and fluorescence spectra of 1 in single crystal state

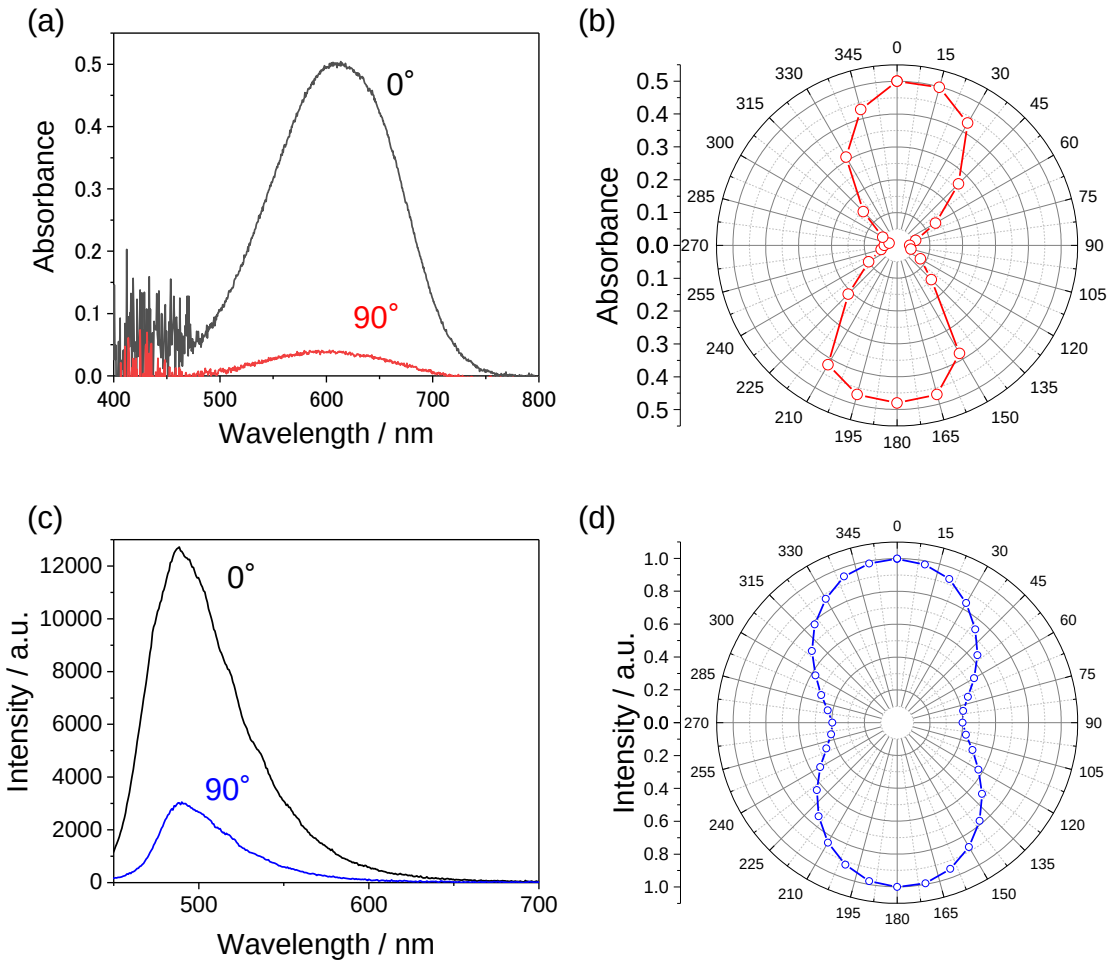


Figure S4 Polarized anisotropy of absorption and fluorescence spectra of UV-irradiated single crystal of **1**. (a) Polarized absorption spectra and (b) polar plots of the absorbance at 600 nm. (c) Polarized fluorescence spectra and (d) polar plot of the fluorescence intensity at 490 nm. The excitation wavelength used for the fluorescence detection was 438 nm.

9. References

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