

## Supporting Information

Molecular engineering to red-shift the absorption band of  
AIE photosensitizers and improve their ROS generation  
ability

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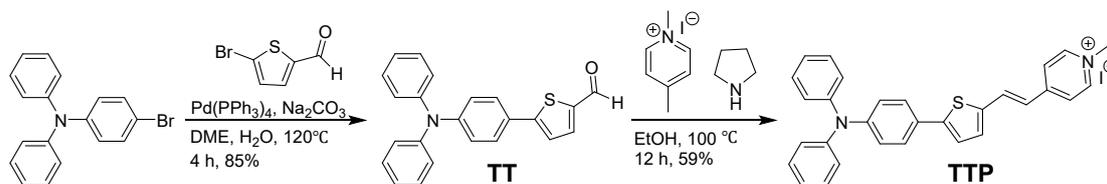
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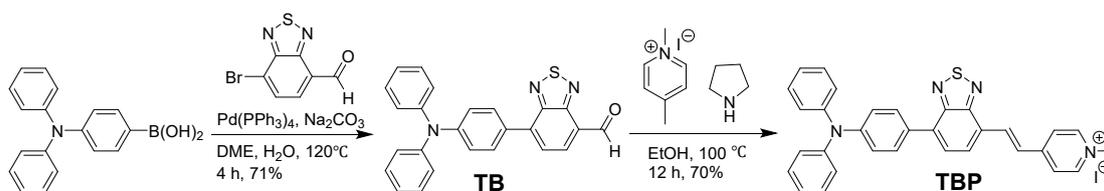
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## Synthesis and characterization of TTP, TBP, TBCP, and TBQ:



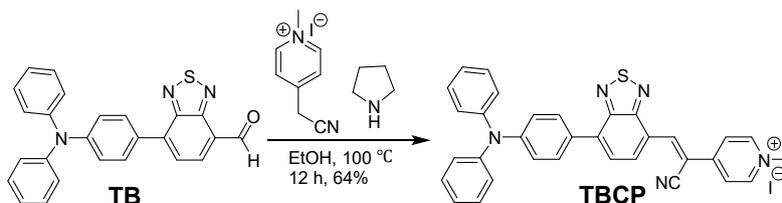
**Synthesis of TT:** A solution of (4-(diphenylamino)phenyl)boronic acid (4.00 g, 13.8 mmol), 5-bromo-2-thiophenecarboxaldehyde (2.40 g, 12.6 mmol), Pd(PPh<sub>3</sub>)<sub>4</sub> (480 mg, 0.416 mmol) and Na<sub>2</sub>CO<sub>3</sub> (2.66 g, 25.1 mmol) in H<sub>2</sub>O (20 mL) and DME (50 mL) was stirred at 120 °C for 4 h. After being cooled to room temperature, the reaction mixture was added water (30 mL), and the resulting mixture was extracted with ethyl acetate. The extracts were combined, washed with H<sub>2</sub>O and brine, dried over Na<sub>2</sub>SO<sub>4</sub> and filtered, the resulting crude product was purified by flash chromatography (silica, ethyl acetate/hexanes = 1/9) to give **TT** (3.74 g, 85%) as a yellow solid: <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 9.85 (s, 1H), 7.71 (d, *J* = 4.0 Hz, 1H), 7.53 (d, *J* = 8.4 Hz, 2H), 7.32 – 7.26 (m, 5H), 7.15 – 7.05 (m, 8H).

**Synthesis of TTP:** A solution of **TT** (100 mg, 0.28 mmol), 1,4-dimethylpyridin-1-ium iodide (96 mg, 0.42 mmol) and pyrrolidine (2.1 mg, 0.030 mmol) in EtOH (3 mL) was stirred at 100 °C overnight. After being cooled to room temperature, the volatile components of the mixture were removed under reduced pressure, and the resulting crude product was purified by flash chromatography (Al<sub>2</sub>O<sub>3</sub>, MeOH/CH<sub>2</sub>Cl<sub>2</sub> = 1/4) to give **TTP** (95 mg, 59%) as a red solid: <sup>1</sup>H NMR (400 MHz, DMSO-d<sub>6</sub>) δ 8.79 (d, *J* = 6.4 Hz, 2H), 8.20 (d, *J* = 16.0 Hz, 1H), 8.15 (d, *J* = 6.4 Hz, 2H), 7.63 (d, *J* = 8.4 Hz, 2H), 7.51 – 7.48 (m, 2H), 7.37 – 7.33 (m, 5H), 7.13 – 7.06 (m, 6H), 6.98 (d, *J* = 8.8 Hz, 2H), 4.20 (s, 3H); <sup>13</sup>C NMR (100 MHz, DMSO) δ 152.2, 147.8, 147.2, 146.6, 144.8, 138.7, 133.8, 129.8, 126.8, 126.3, 124.8, 124.2, 123.9, 122.9, 122.2, 121.2, 46.8; HRMS (ESI) *m/e* calcd for C<sub>30</sub>H<sub>25</sub>N<sub>2</sub>S (M-I)<sup>+</sup> 445.1733, found 445.1738.

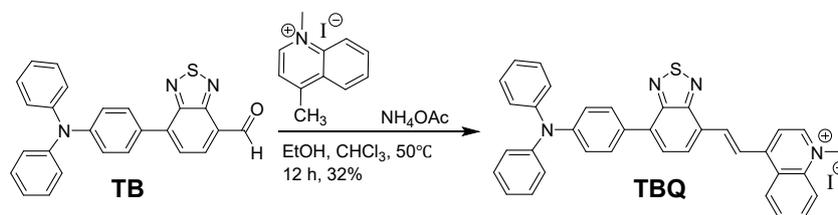


**Synthesis of TB:** A solution of (4-(diphenylamino)phenyl)boronic acid (1.10 g, 3.82 mmol), 7-bromobenzo[c][1,2,5]thiadiazole-4-carbaldehyde (840 mg, 3.46 mmol), Pd(PPh<sub>3</sub>)<sub>4</sub> (120 mg, 0.104 mmol) and Na<sub>2</sub>CO<sub>3</sub> (872 mg, 10.4 mmol) in H<sub>2</sub>O (5 mL) and DME (15 mL) was stirred at 120 °C for 4 h. After being cooled to room temperature, the reaction mixture was added water (10 mL), and the resulting mixture was extracted with ethyl acetate. The extracts were combined, washed with H<sub>2</sub>O and brine, dried over Na<sub>2</sub>SO<sub>4</sub> and filtered. After removal of volatile components from the filtrate, the resulting crude product was purified by flash chromatography (silica, ethyl acetate/hexanes = 1/9) to give **TB** (1.13 g, 71%) as a dark solid: <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 10.75 (s, 1H), 8.29 (d, *J* = 7.2 Hz, 1H), 7.95 (d, *J* = 8.8 Hz, 2H), 7.86 (d, *J* = 7.6 Hz, 1H), 7.34 (t, *J* = 7.6 Hz, 4H), 7.21 (d, *J* = 7.6 Hz, 6H), 7.13 (t, *J* = 7.2 Hz, 2H).

**Synthesis of TBP:** A solution of **TB** (82 mg, 0.20 mmol), 1,4-dimethylpyridin-1-ium iodide (47 mg, 0.20 mmol) and pyrrolidine (1.4 mg, 0.020 mmol) in EtOH (3 mL) was stirred at 100 °C overnight. After being cooled to room temperature, the volatile components of the mixture were removed under reduced pressure, and the resulting crude product was purified by flash chromatography (Al<sub>2</sub>O<sub>3</sub>, MeOH/CH<sub>2</sub>Cl<sub>2</sub> = 1/9) to give **TBP** (80 mg, 70%) as a red solid: <sup>1</sup>H NMR (400 MHz, DMSO-d<sub>6</sub>) δ 8.91 (d, *J* = 5.6 Hz, 2H), 8.38 – 8.34 (m, 3H), 8.14 (d, *J* = 6.4 Hz, 1H), 8.04 – 7.99 (m, 3H), 7.40 – 7.36 (m, 5H), 7.14 – 7.09 (m, 8H), 4.28 (s, 3H); <sup>13</sup>C NMR (100 MHz, DMSO) δ 153.3, 153.1, 152.4, 148.0, 146.7, 145.2, 136.4, 134.3, 132.6, 130.4, 129.8, 129.6, 127.1, 127.0, 126.1, 124.8, 123.9, 123.8, 121.7, 47.0; HRMS (ESI) *m/e* calcd for C<sub>32</sub>H<sub>25</sub>N<sub>4</sub>S (M-I)<sup>+</sup> 497.1794, found 497.1795.



**Synthesis of TBCP:** A solution of **TB** (150 mg, 0.37 mmol), 4-(cyanomethyl)-1-methylpyridin-1-ium iodide (99 mg, 0.38 mmol) and pyrrolidine (2.6 mg, 0.037 mmol) in EtOH (8 mL) was stirred at 100 °C overnight. After being cooled to room temperature, the volatile components of the mixture were removed under reduced pressure, and the resulting crude product was purified by flash chromatography ( $\text{Al}_2\text{O}_3$ ,  $\text{MeOH}/\text{CH}_2\text{Cl}_2 = 1/9$ ) to give **TBCP** (87 mg, 64%) as a dark purple solid:  $^1\text{H}$  NMR (400 MHz,  $\text{DMSO-d}_6$ )  $\delta$  9.17 (s, 1H), 9.08 (d,  $J = 6.8$  Hz, 2H), 8.76 (d,  $J = 7.6$  Hz, 1H), 8.54 (d,  $J = 7.2$  Hz, 2H), 8.18 (d,  $J = 8.0$  Hz, 1H), 8.12 (d,  $J = 8.8$  Hz, 2H), 7.42 – 7.38 (m, 4H), 7.18 – 7.14 (m, 6H), 7.12 (d,  $J = 8.8$  Hz, 2H), 4.38 (s, 3H);  $^{13}\text{C}$  NMR (100 MHz,  $\text{DMSO}$ )  $\delta$  154.2, 152.3, 148.6, 148.4, 146.5, 146.1, 144.7, 136.9, 130.7, 130.5, 129.8, 128.8, 126.6, 125.1, 124.2, 123.5, 123.4, 121.2, 116.1, 107.4, 47.5; HRMS (ESI)  $m/e$  calcd for  $\text{C}_{33}\text{H}_{24}\text{N}_5\text{S}$  ( $\text{M-I}$ ) $^+$  522.1747, found 522.1757.



**Synthesis of TBQ:** A solution of **TB** (100 mg, 0.25 mmol), 1,4-dimethylquinolin-1-ium iodide (71 mg, 0.25 mmol) and  $\text{NH}_4\text{OAc}$  (22 mg, 0.29 mmol) in  $\text{EtOH}/\text{CHCl}_3 = 1/4$  (5 mL) was stirred at 50 °C overnight. After being cooled to room temperature, the volatile components of the mixture were removed under reduced pressure, and the resulting crude product was purified by flash chromatography ( $\text{Al}_2\text{O}_3$ ,  $\text{MeOH}/\text{CH}_2\text{Cl}_2 = 1/9$ ) to give **TBQ** (53 mg, 32%) as a purple solid:  $^1\text{H}$  NMR (400 MHz,  $\text{DMSO-d}_6$ )  $\delta$  9.43 (d,  $J = 6.4$  Hz, 1H), 9.18 (d,  $J = 15.2$  Hz, 1H), 8.92 (d,  $J = 8.0$  Hz, 1H), 8.64 (d,  $J = 6.8$  Hz, 1H), 8.58 (d,  $J = 16.4$  Hz, 1H), 8.52 (d,  $J = 8.8$  Hz, 1H), 8.39 (d,  $J = 7.2$  Hz, 1H), 8.34 (t,  $J = 7.2$  Hz, 1H), 8.16 (t,  $J = 8.0$  Hz, 1H), 8.08 – 8.04 (m, 3H), 7.41 (t,  $J =$

8.0 Hz, 4H), 7.16–7.11 (m, 8H), 4.59 (s, 3H); <sup>13</sup>C NMR (100 MHz, DMSO-d<sub>6</sub>) δ 153.3, 153.1, 152.0, 148.3, 148.1, 146.7, 138.7, 137.9, 135.1, 134.5, 132.7, 130.5, 129.8, 129.7, 129.4, 127.0, 126.4, 126.3, 125.7, 124.9, 124.0, 123.3, 121.6, 119.6, 116.4, 44.9; HRMS (ESI) m/e calcd for C<sub>36</sub>H<sub>27</sub>N<sub>4</sub>S (M-I)<sup>+</sup> 547.1951, found 547.1960.

**Fluorescence quantum yield measurement:** Fluorescence quantum yields (relative values) of AIE-PSs (10 μM) in DMSO containing 95% toluene were measured by detecting the fluorescence (λ<sub>ex</sub> = 500 nm) and calculated according to the following expression:

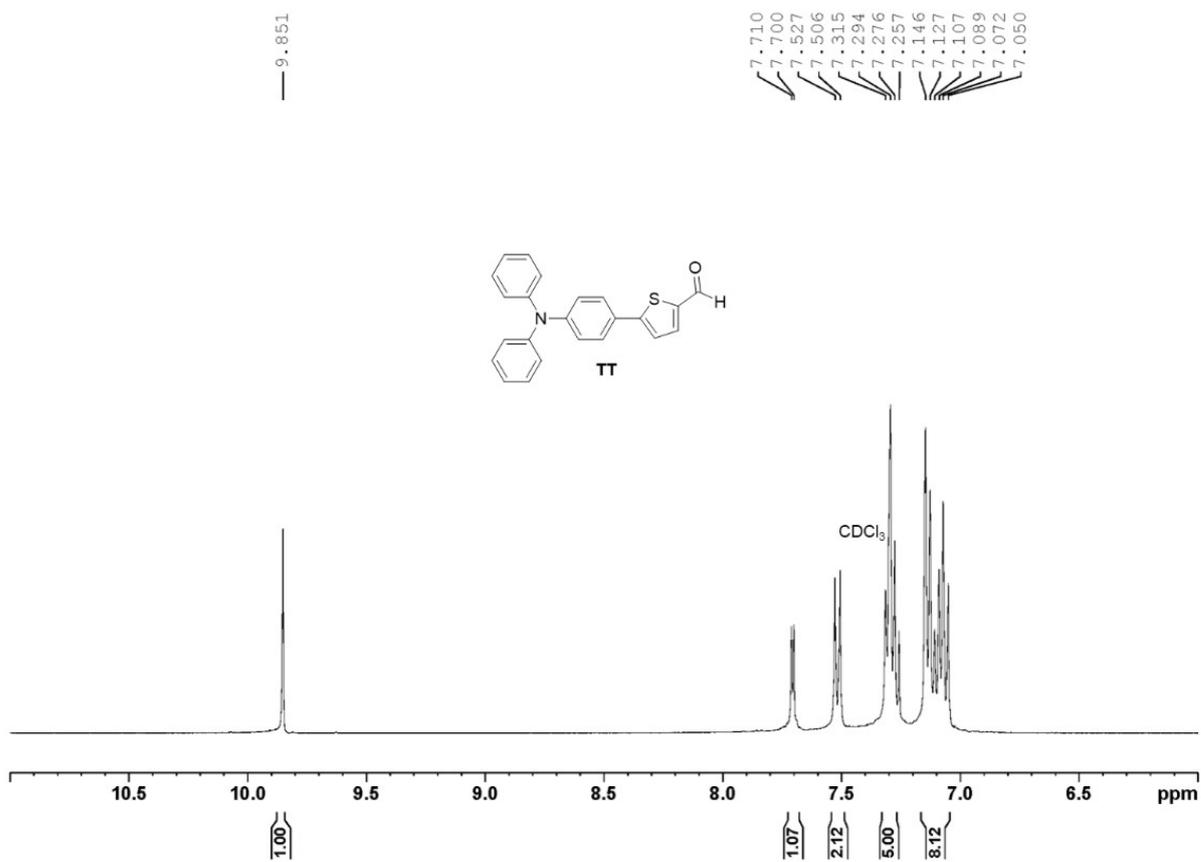
$$\phi_{fS} = \frac{F_S}{F_R} \times \frac{(1 - 10^{-A_R})}{(1 - 10^{-A_S})} \times \phi_{fR}$$

where the subscripts R and S refer to the reference and the sample, respectively, Φ<sub>f</sub> is the fluorescence quantum yield, and Φ<sub>fR</sub> is the fluorescence quantum yield of TTPy (Φ<sub>f</sub> = 0.089) which was measured by a calibrated integrating sphere in DMSO containing 95% toluene, *F* is the integrated fluorescence intensity under fluorescence emission spectrum, *A* is the value of OD<sub>500</sub>.

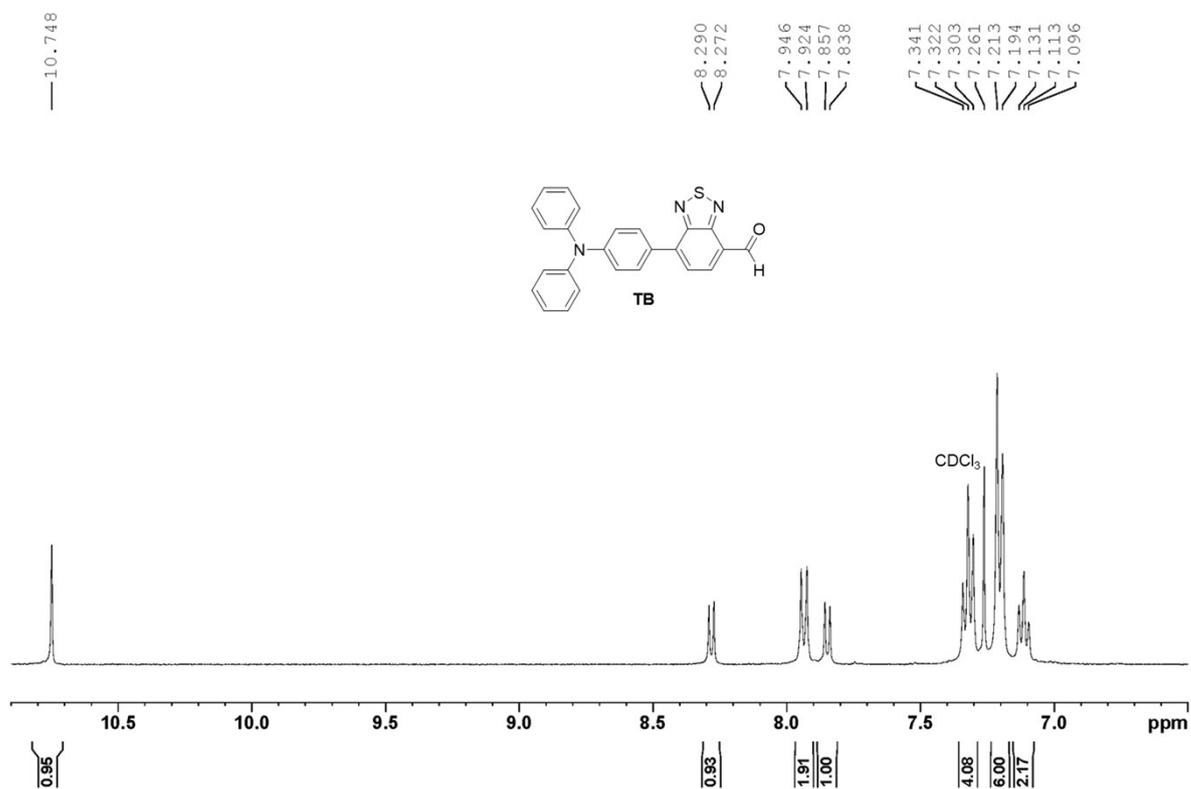
**Singlet oxygen (<sup>1</sup>O<sub>2</sub>) detection and quantum yield measurement:** The <sup>1</sup>O<sub>2</sub> generation was studied using 9,10-anthracenediy-bis(methylene)dimalonic acid (ABDA) as an indicator, as the absorbance of ABDA will decrease upon reaction with <sup>1</sup>O<sub>2</sub>. Typically, a mixture of ABDA (100 μM) and AIE-PSs (10 μM) in water with 1% DMSO was exposed to green (517 ± 5 nm, 9 mW/cm<sup>2</sup>) or red (605 ± 5 nm, 9 mW/cm<sup>2</sup>) light. The decomposition of ABDA was monitored by recording the absorbance decrease at 378 nm every 5 s. <sup>1</sup>O<sub>2</sub> quantum yields of AIE-PSs were calculated by the equation:

$$\phi_{\Delta AIE-PSs} = \frac{K_{AIE-PSs} \times A_{RB}}{K_{RB} \times A_{AIE-PSs}} \times \phi_{\Delta RB}$$

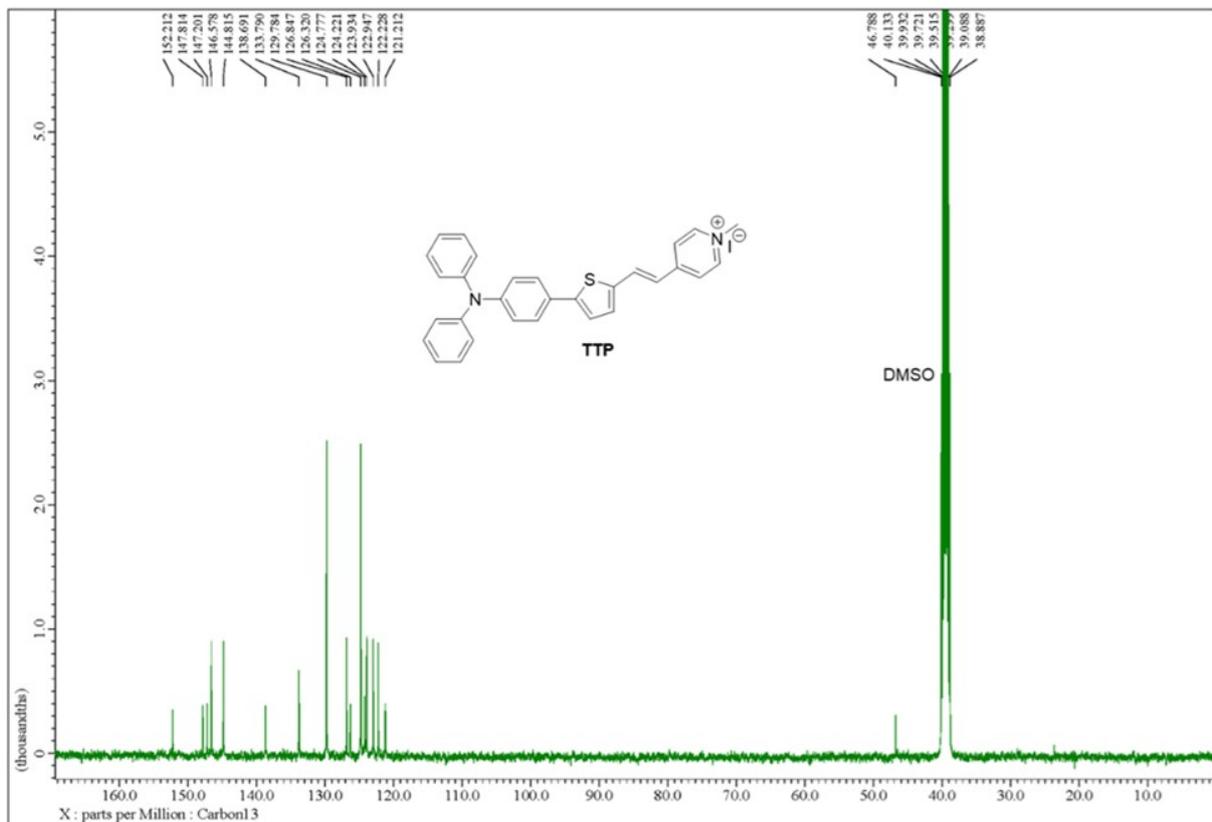
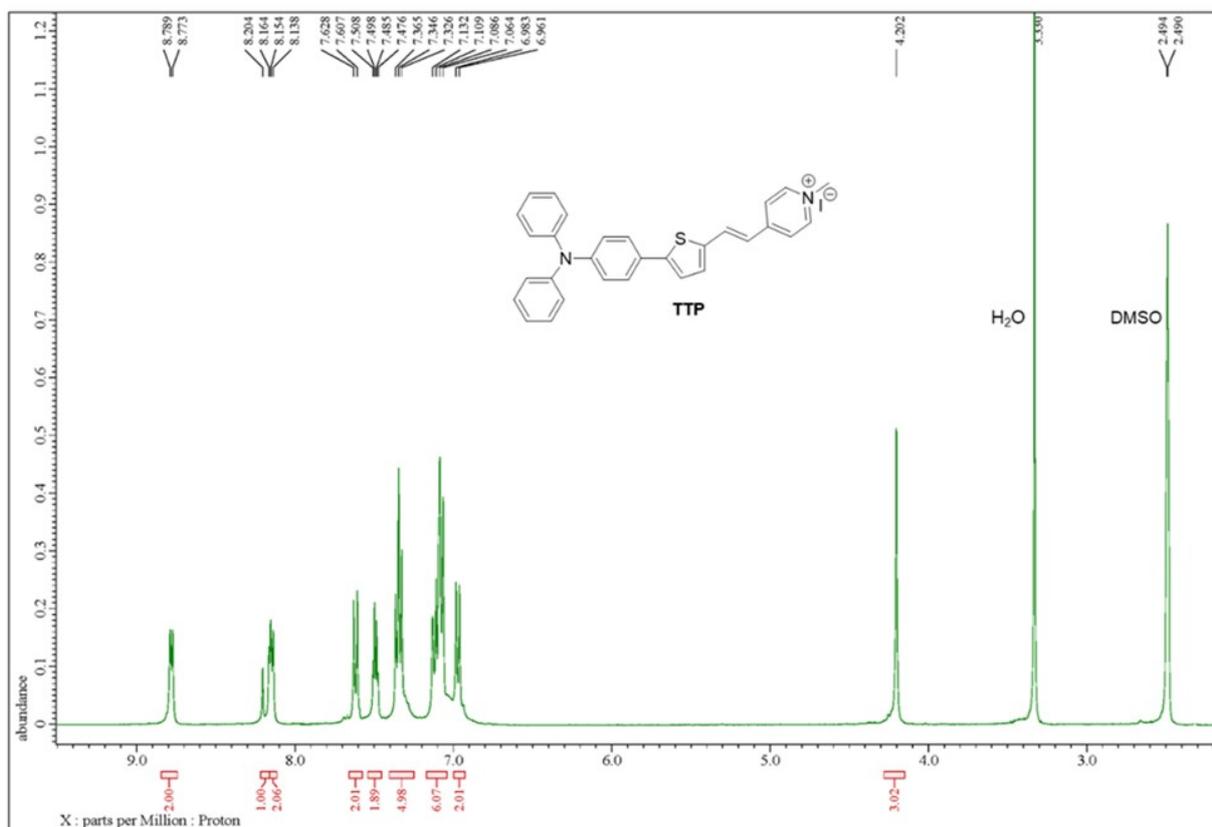
Φ<sub>ΔRB</sub> is the singlet oxygen quantum yield of Rose Bengal in water (Φ<sub>Δ</sub> = 0.75), *A* is the value of OD<sub>517</sub>, and *K* is the slope of the descent curve of ABDA absorbance decrease.



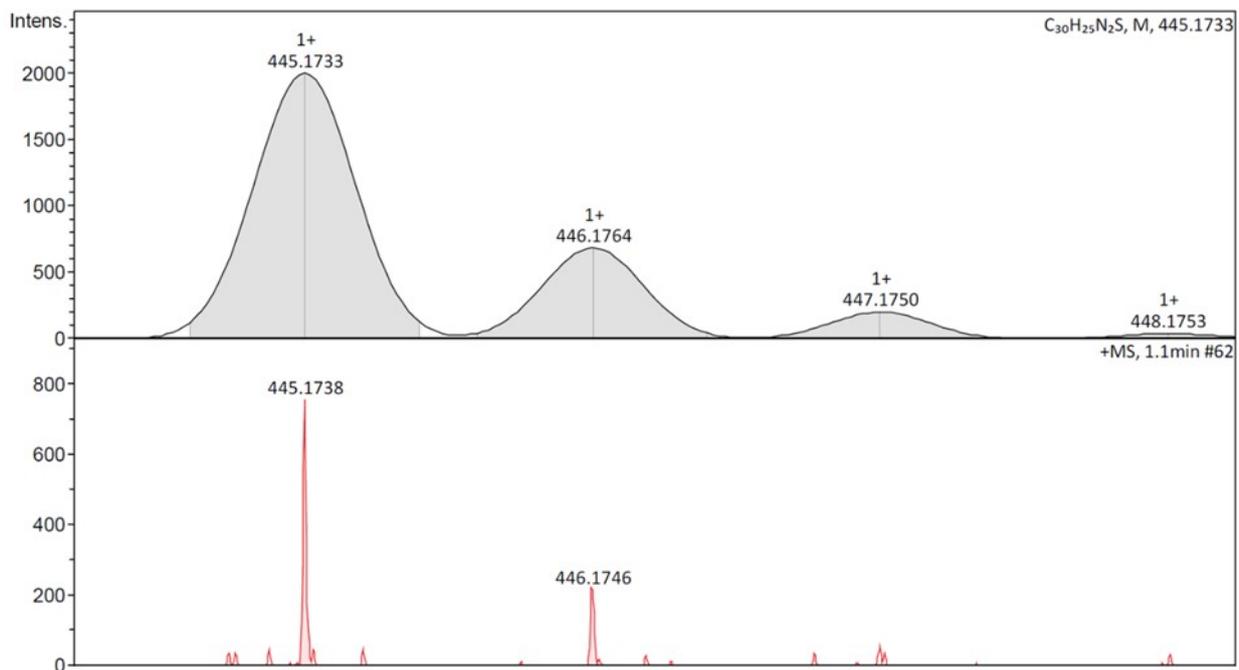
**Figure S1.**  $^1\text{H}$  NMR spectrum of **TT**.



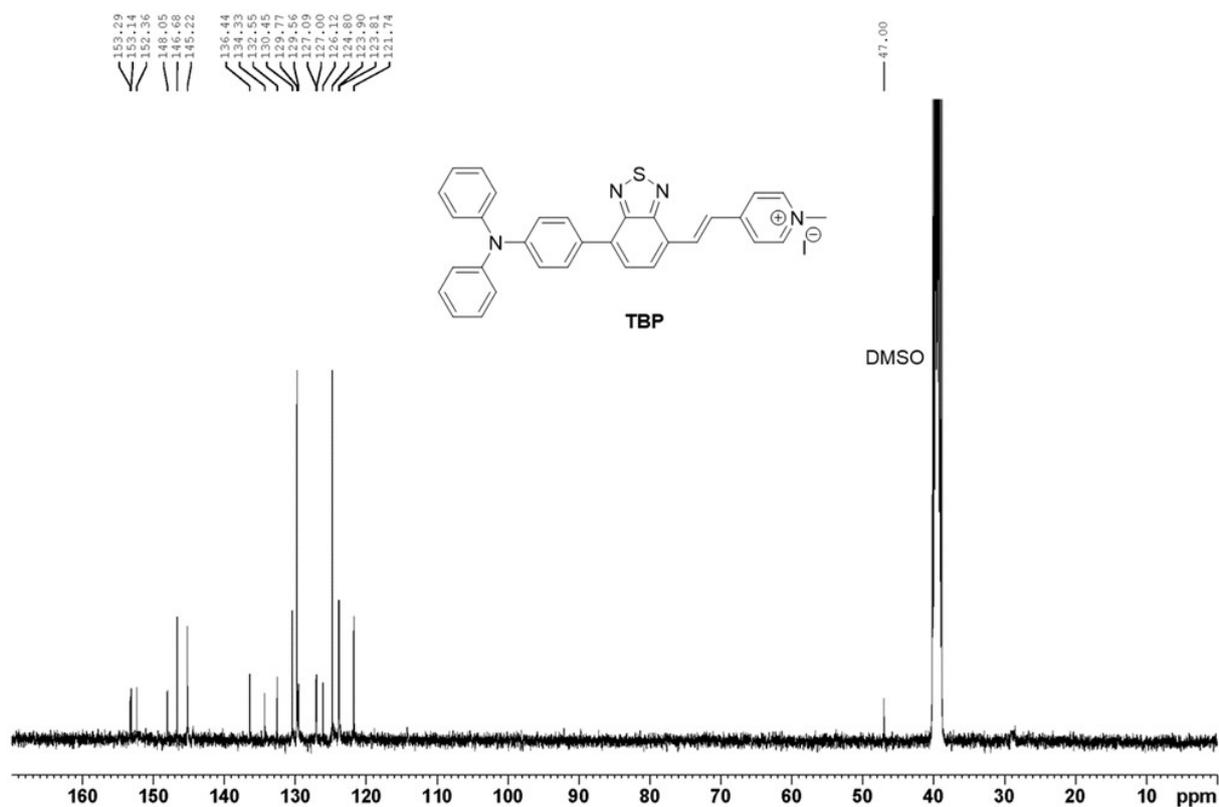
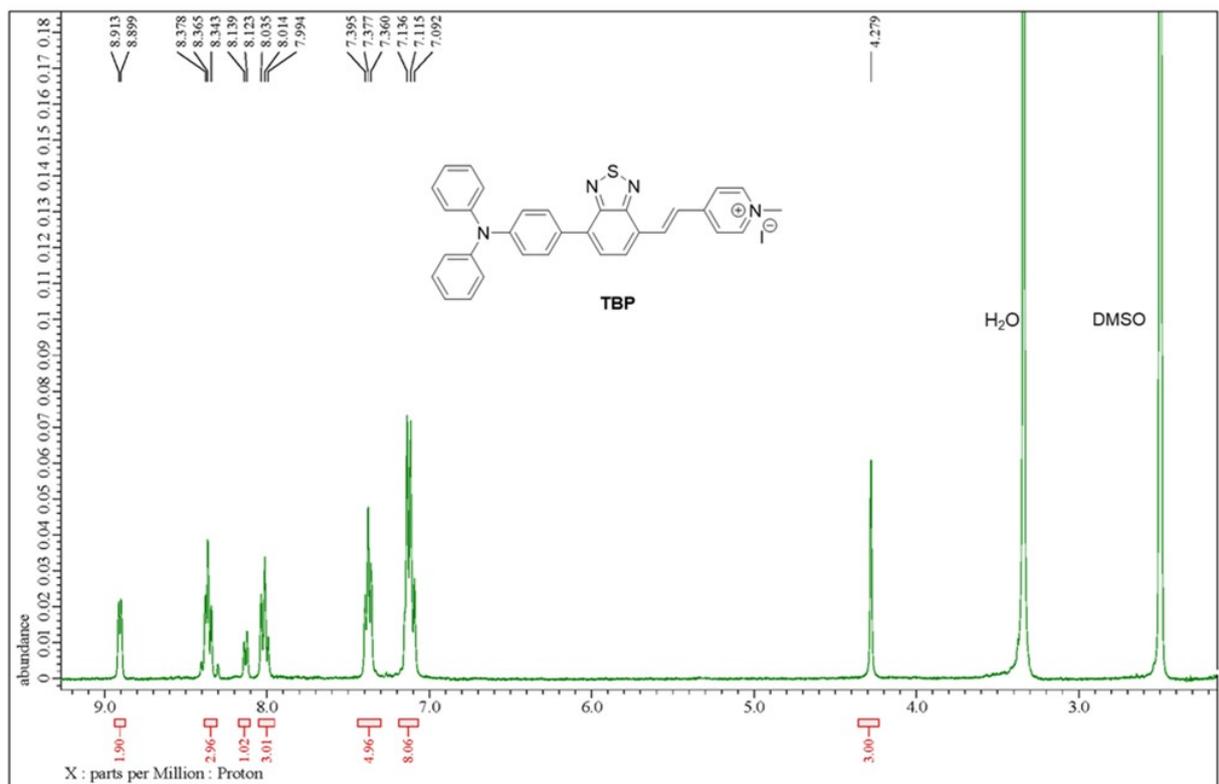
**Figure S2.**  $^1\text{H}$  NMR spectrum of TB.



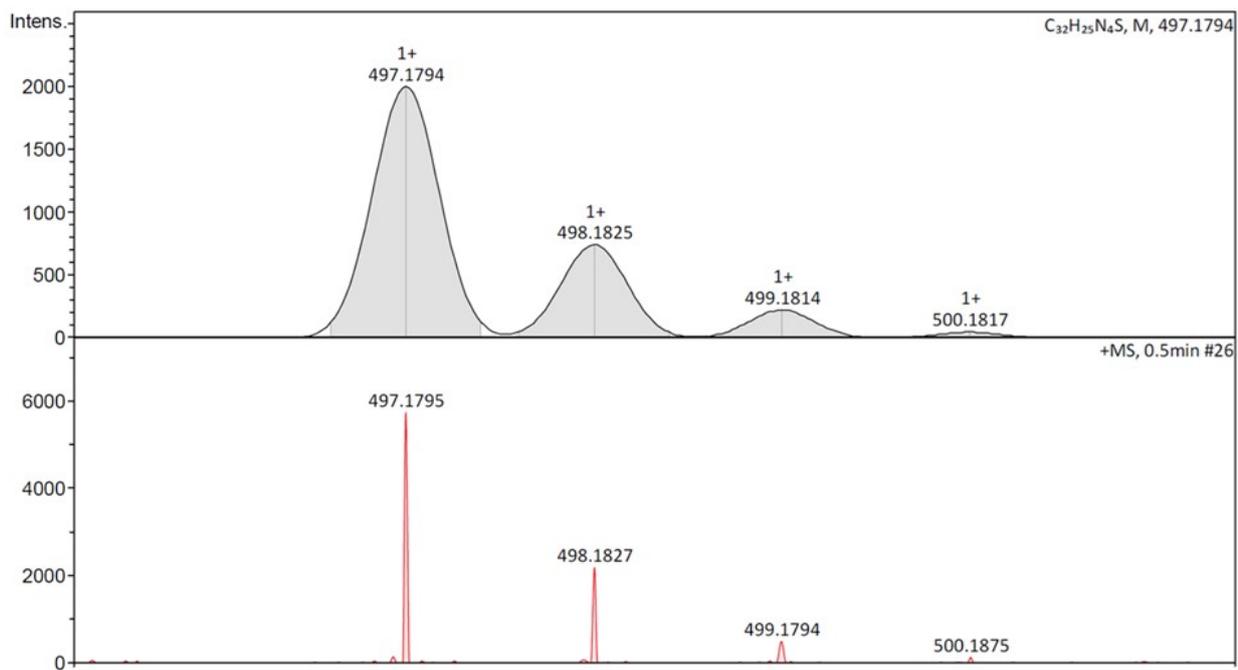
**Figure S3.**  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectra of TTP.



**Figure S4.** HRMS spectrum of TTP.



**Figure S5.** <sup>1</sup>H NMR and <sup>13</sup>C NMR spectra of TBP.



**Figure S6.** HRMS spectrum of TBP.

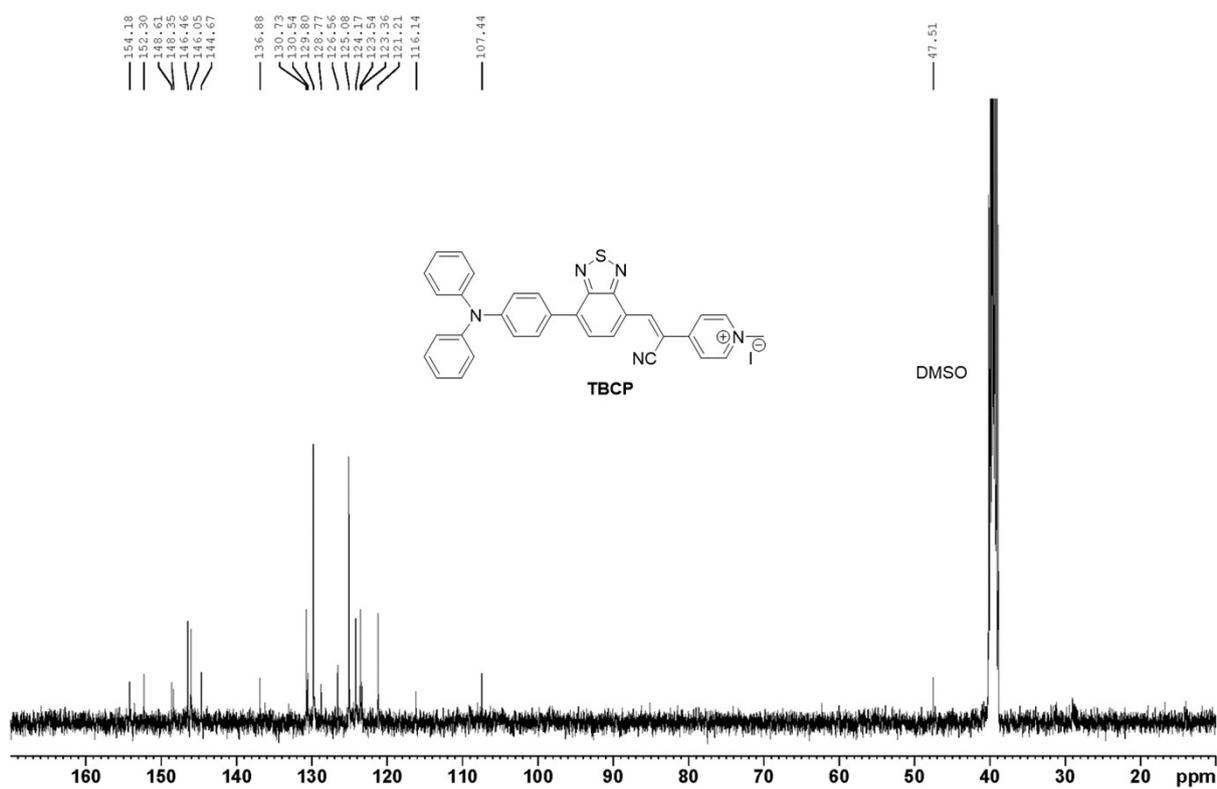
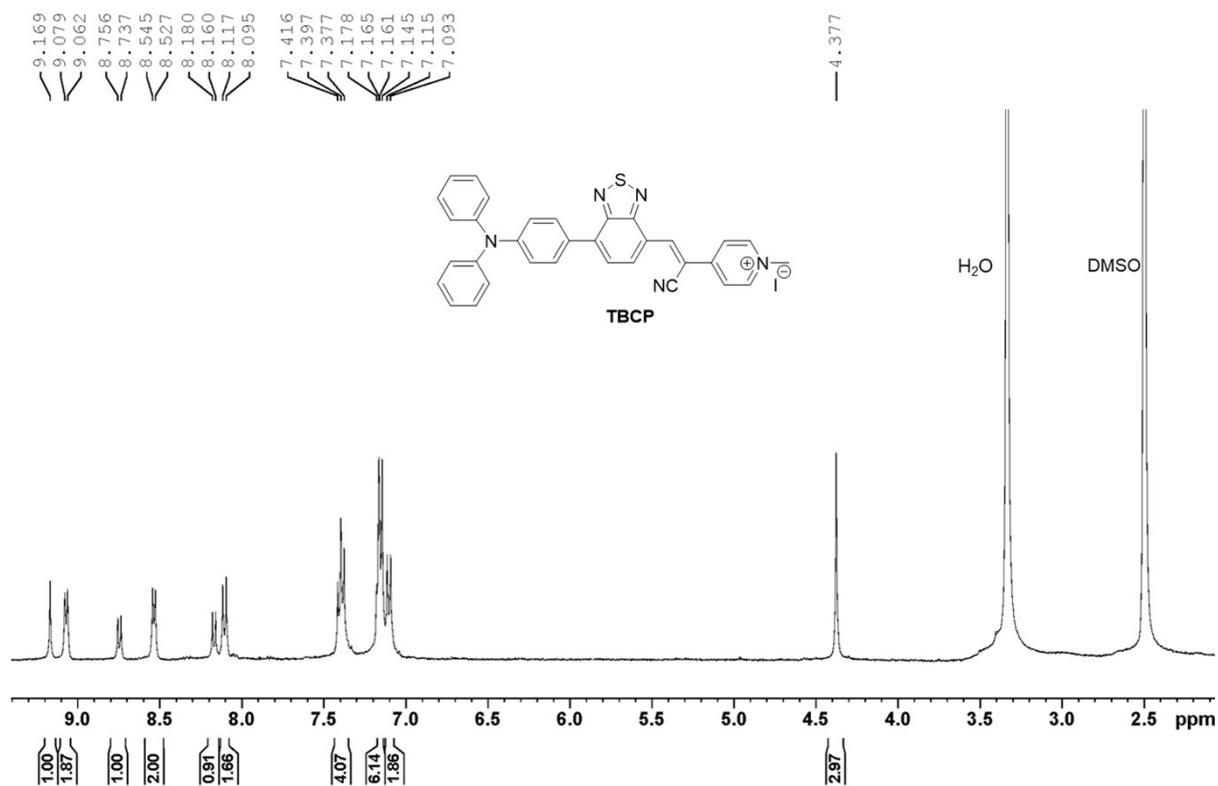
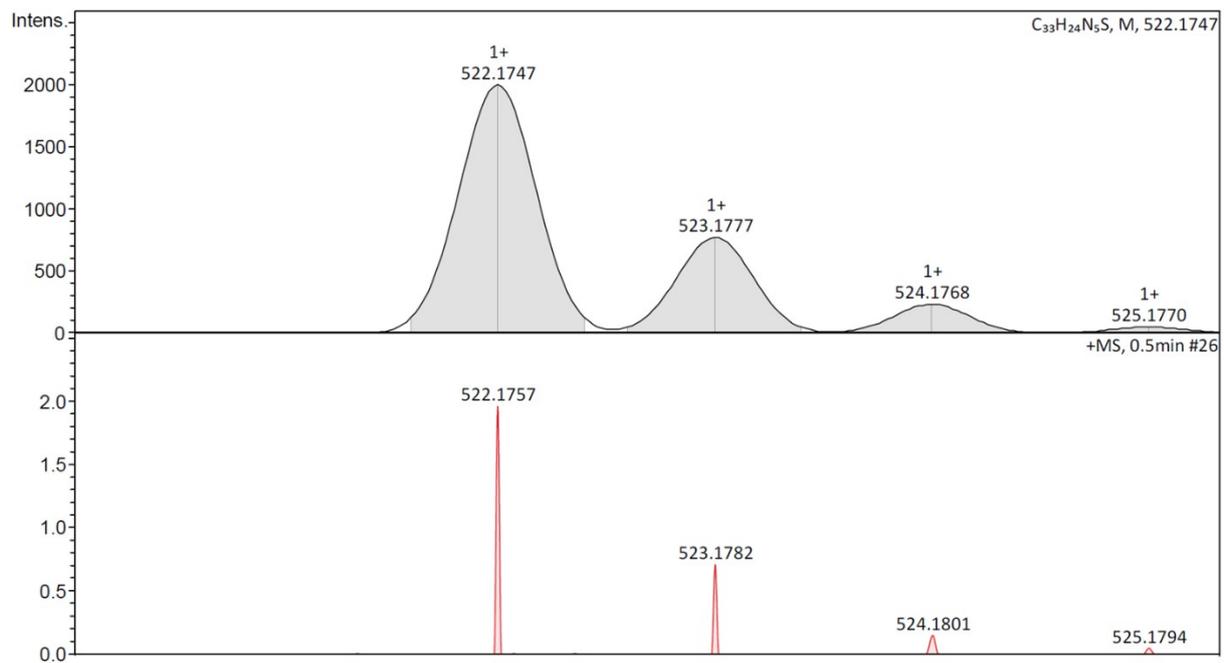


Figure S7. <sup>1</sup>H NMR and <sup>13</sup>C NMR spectra of TBCP.



**Figure S8.** HRMS spectrum of **TBCP**.

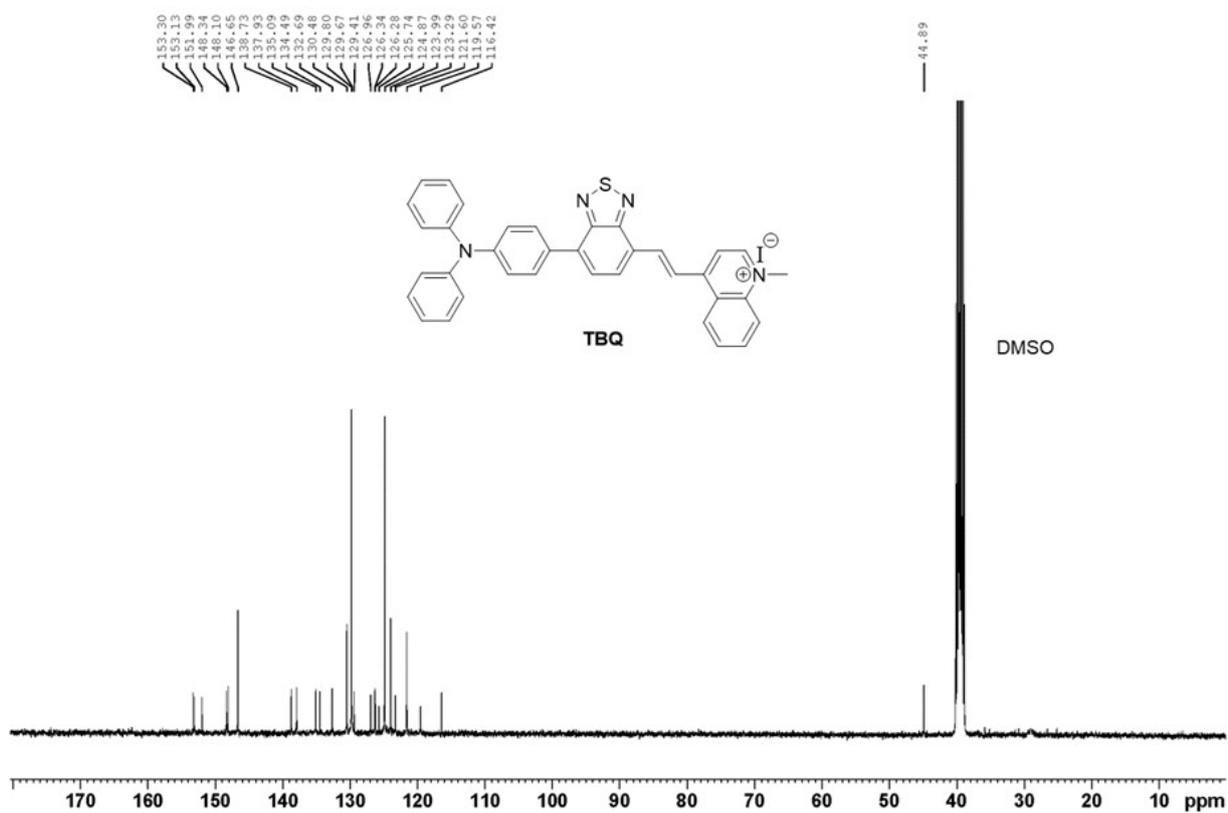
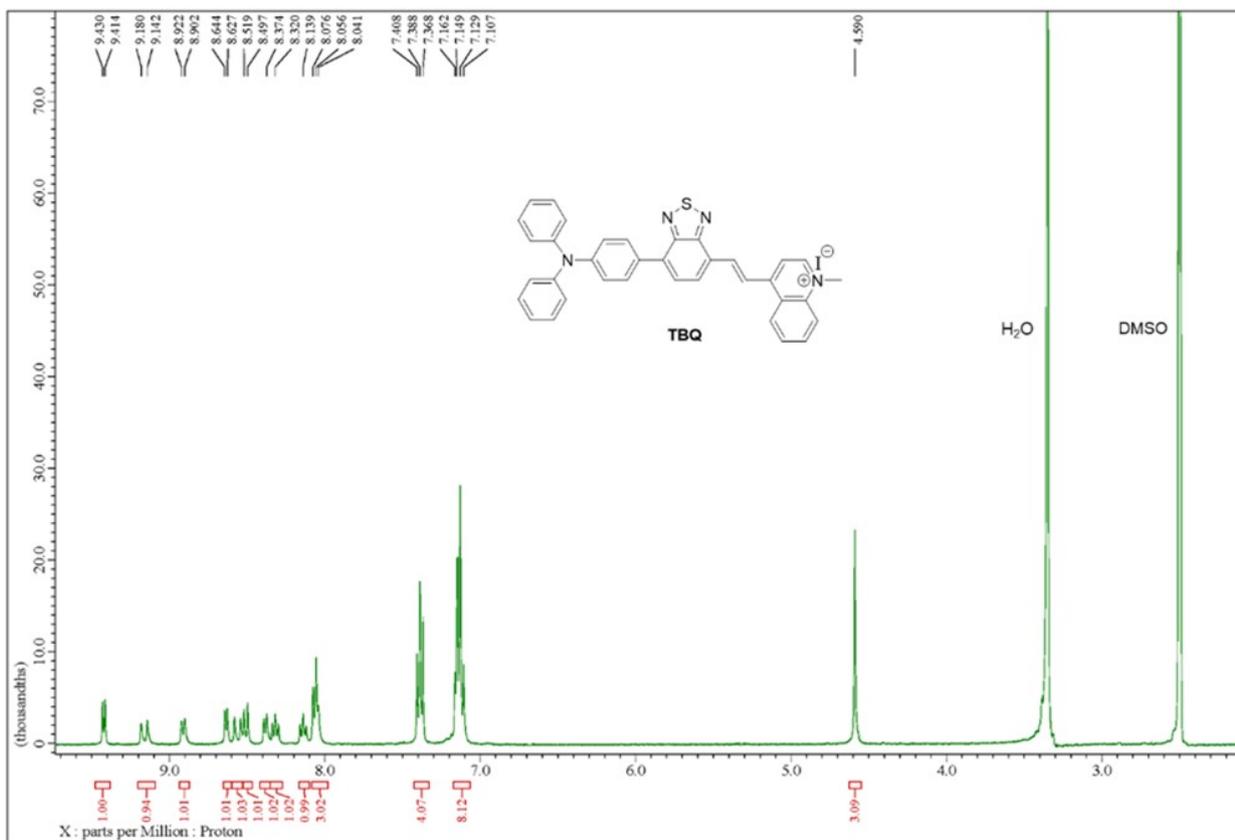
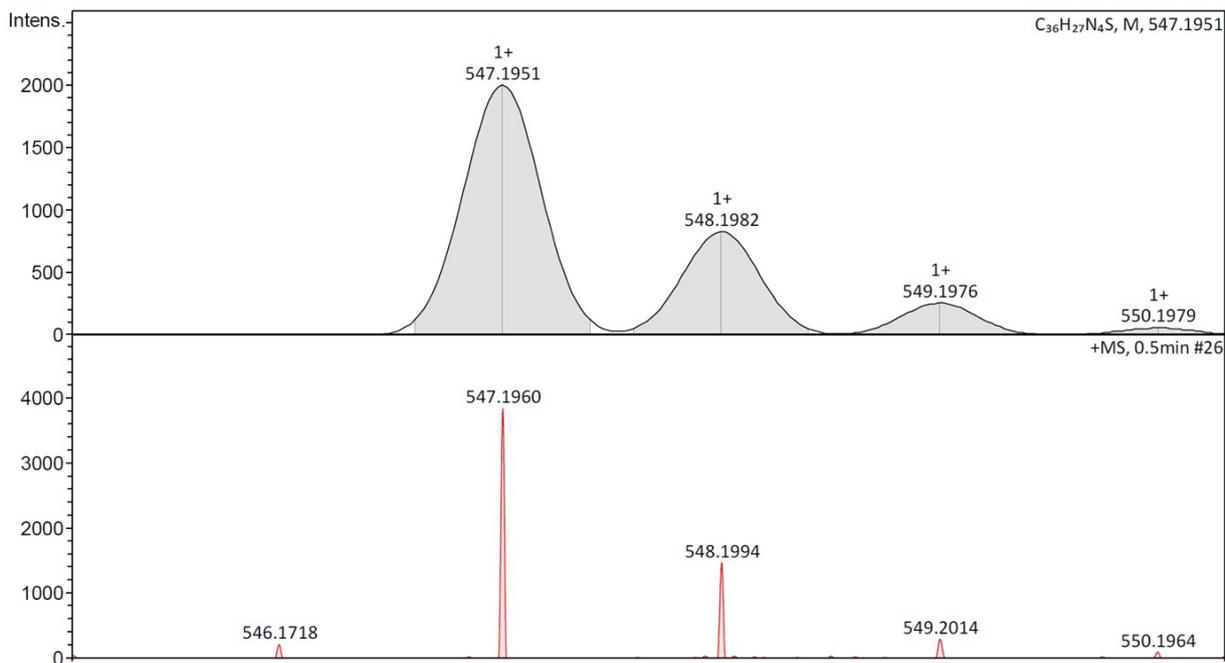
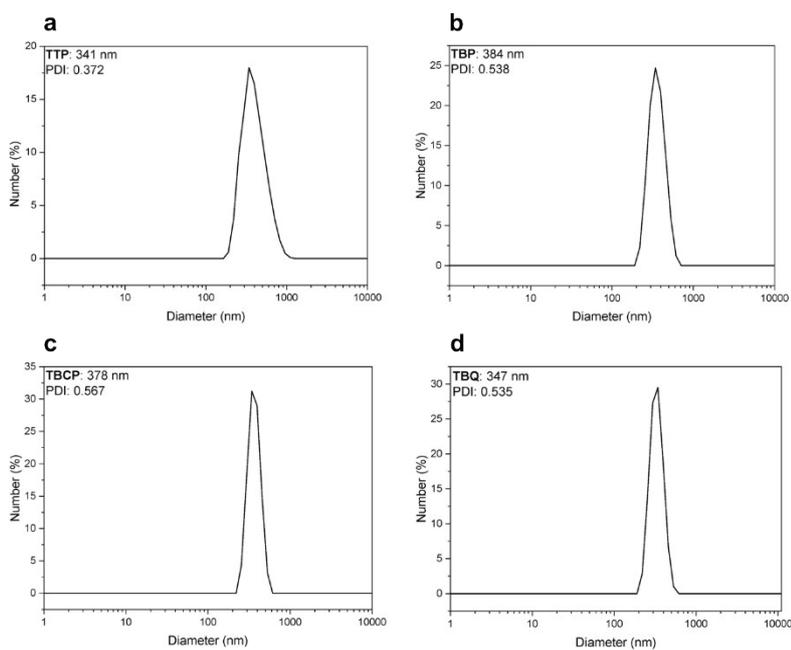


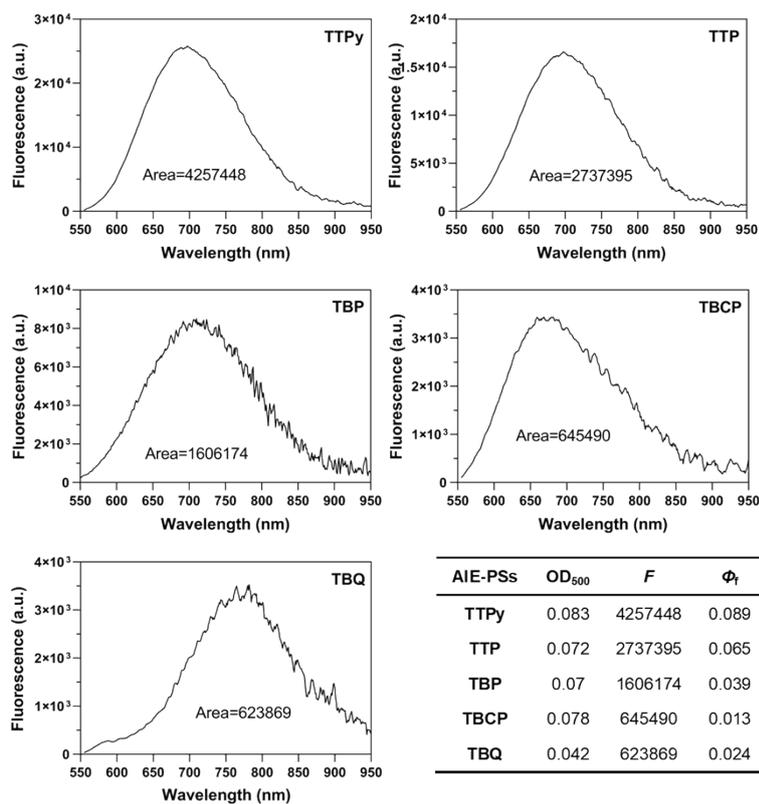
Figure S9. <sup>1</sup>H NMR and <sup>13</sup>C NMR spectra of TBQ.



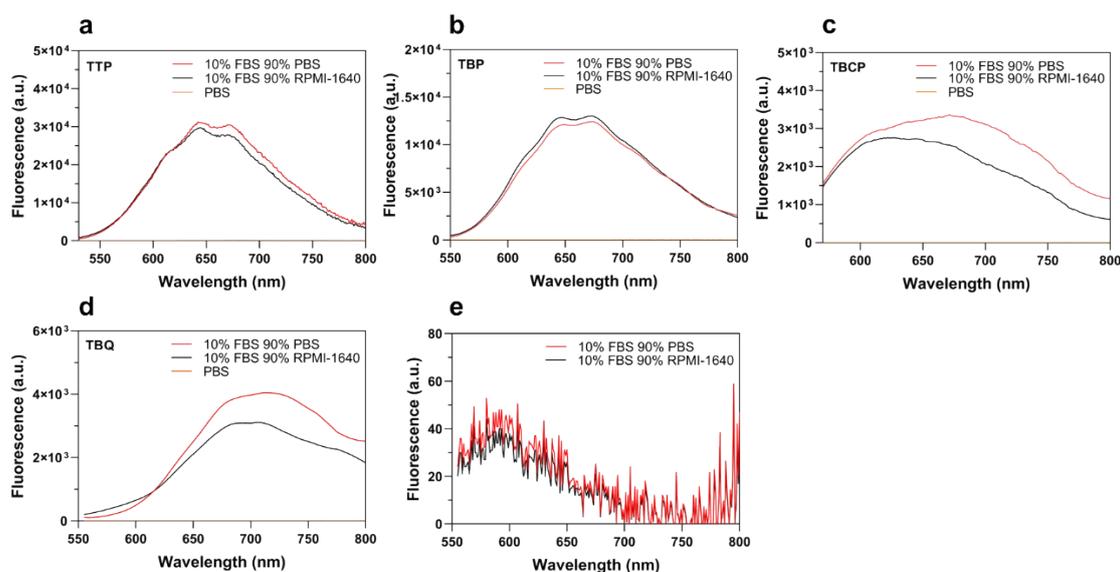
**Figure S10.** HRMS spectrum of TBQ.



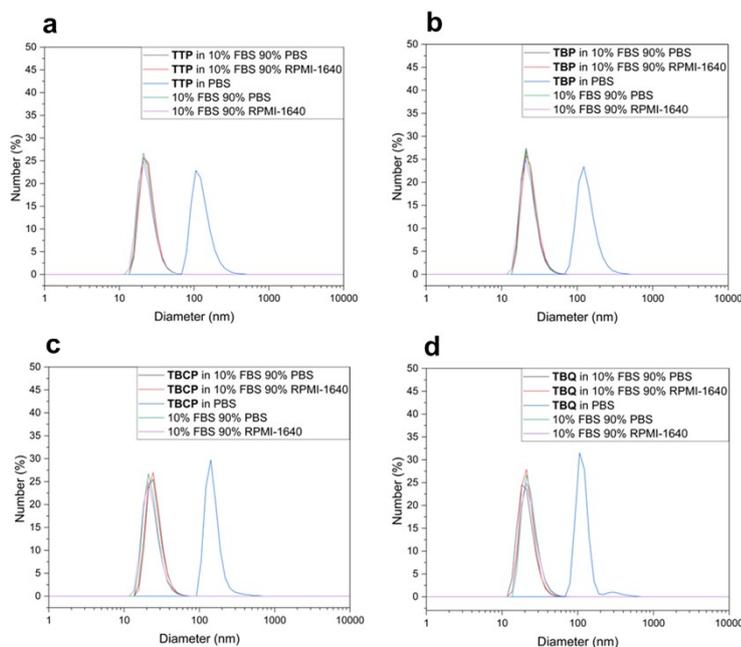
**Figure S11.** Size distributions of (a) TTP, (b) TBP, (c) TBCP, and (d) TBQ (10  $\mu$ M) in DMSO containing 95% toluene.



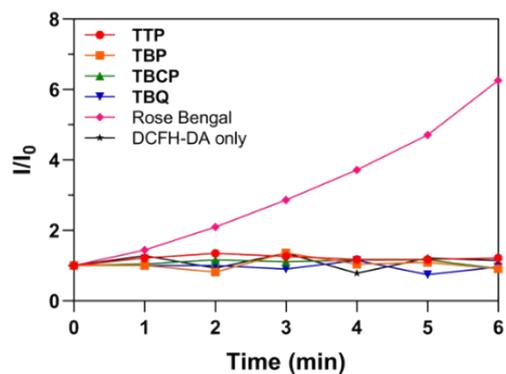
**Figure S12.** The measurement of fluorescence quantum yields ( $\Phi_f$ ) of AIE-PSs in DMSO containing 95% toluene ( $\lambda_{ex} = 500$  nm). *F* is the integrated fluorescence intensity under fluorescence emission spectrum.



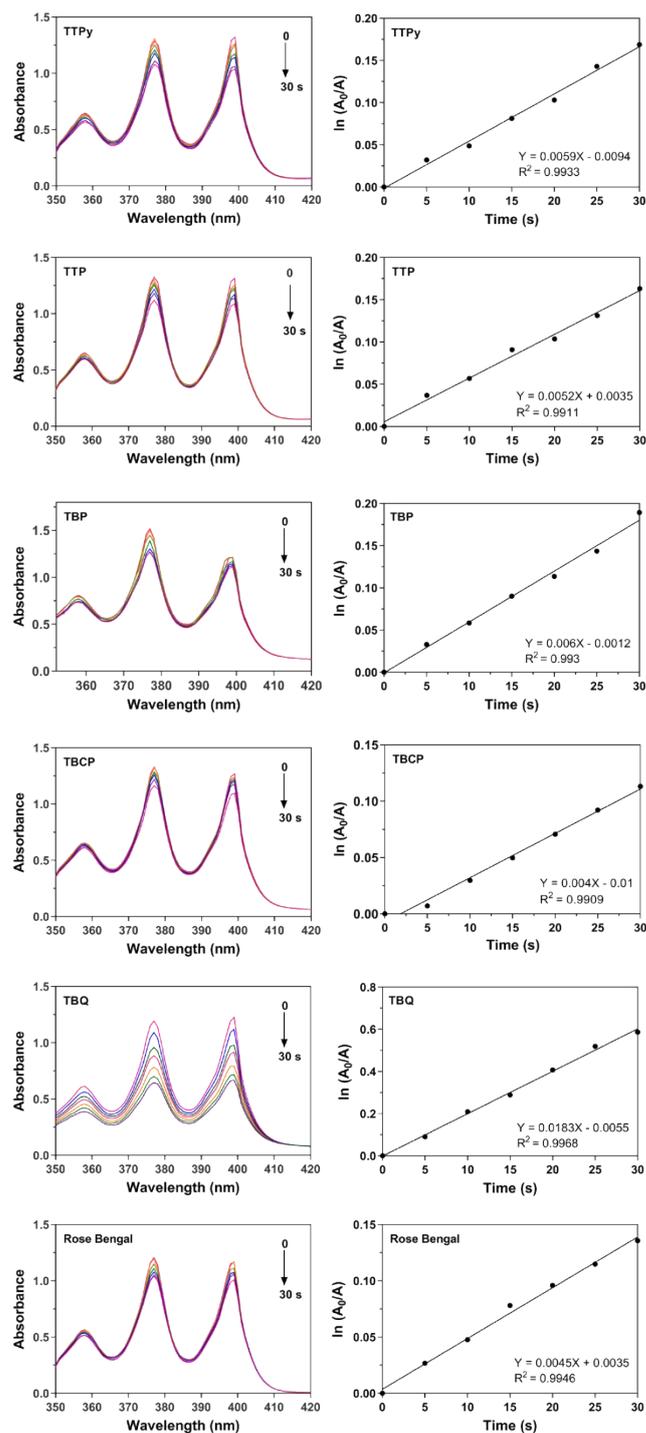
**Figure S13.** The photoluminescence spectra of (a) TTP, (b) TBP, (c) TBCP, and (d) TBQ (10  $\mu$ M) in 10% fetal bovine serum (FBS)/90% phosphate buffer saline (PBS), 10% FBS/90% PRMI-1640, or 100% PBS. (e) The photoluminescence spectra of background solvents containing 10% FBS/90% PBS, or 10% FBS/90% PRMI-1640.  $\lambda_{\text{ex}} = 500$  nm.



**Figure S14.** Size distributions of (a) TTP, (b) TBP, (c) TBCP, and (d) TBQ (10  $\mu$ M) in 10% FBS/90% PBS, 10% FBS/90% PRMI-1640, or 100% PBS.

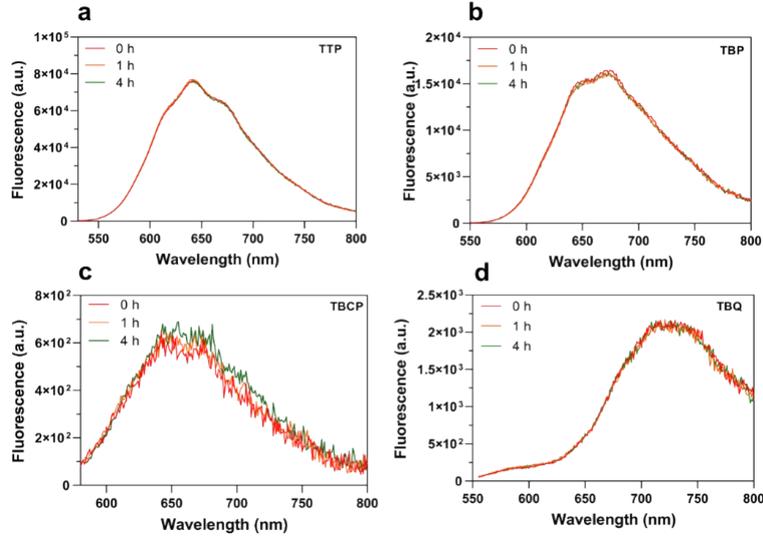


**Figure S15.** ROS generation abilities of AIE-PSs and Rose Bengal (10  $\mu\text{M}$  in water containing 50% methanol) under green light irradiation using DCFH-DA (100  $\mu\text{M}$ ) as an indicator.  $I_0$  and  $I$  are the fluorescence intensities of DCF at 520 nm, excited at 480 nm.

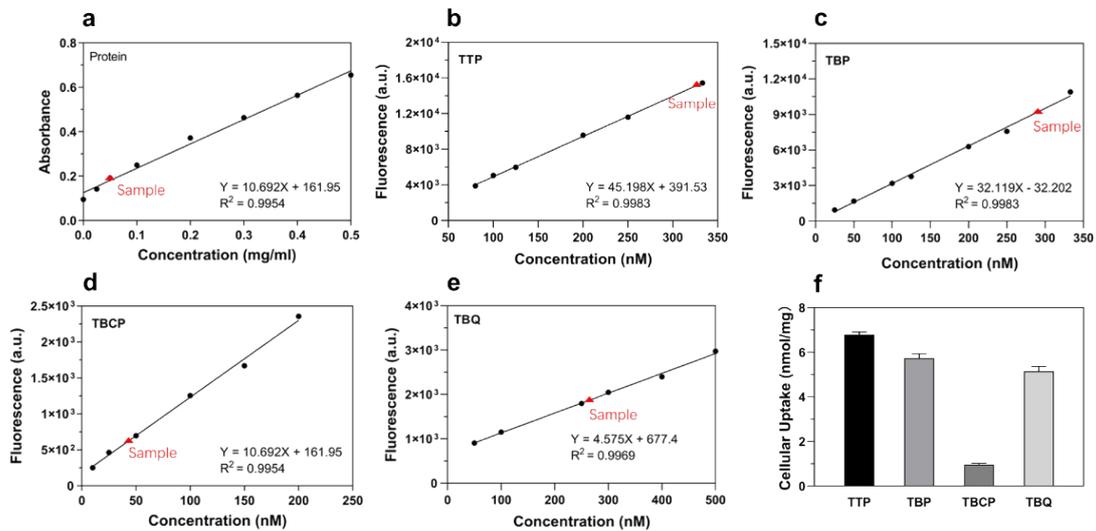


AIE-PSs	TTPy	TTP	TBP	TBQP	TBQ	Rose Bengal
OD <sub>517</sub>	0.121	0.115	0.116	0.165	0.250	0.075
K <sub>AIE-PSs</sub>	0.0059	0.0052	0.0060	0.0040	0.0183	0.0045
Φ <sub>Δ</sub>	0.609	0.565	0.646	0.303	0.886	0.75

**Figure S16.** The measurement of singlet oxygen quantum yields ( $\Phi_{\Delta}$ ) of AIE-PSs in water containing 1% DMSO.  $K$  is the slope of the descent curve of ABDA absorbance decrease.



**Figure S17.** The stability of AIE-PSs (10  $\mu$ M) in the lysate (water with 1% sodium dodecyl sulfate and 0.1 M NaOH) measured by tracking their fluorescence change at 0, 1, and 4 h after mixing. (a) **TTP** ( $\lambda_{\text{ex}} = 478$  nm), (b) **TBP** ( $\lambda_{\text{ex}} = 500$  nm), (c) **TBCP** ( $\lambda_{\text{ex}} = 540$  nm), and (d) **TBQ** ( $\lambda_{\text{ex}} = 525$  nm).



**Figure S18.** (a) The standard absorption curve of proteins ( $\lambda_{\text{abs}} = 562$  nm) detected using BCA protein assay kit. The standard fluorescence curves of (b) **TTP** ( $\lambda_{\text{ex}} = 478$  nm,  $\lambda_{\text{em}} = 650$  nm), (c) **TBP** ( $\lambda_{\text{ex}} = 500$  nm,  $\lambda_{\text{em}} = 670$  nm), (d) **TBCP** ( $\lambda_{\text{ex}} = 540$  nm,  $\lambda_{\text{em}} = 640$  nm), and (e) **TBQ** ( $\lambda_{\text{ex}} = 525$  nm,  $\lambda_{\text{em}} = 720$  nm) in the lysate (water with 1% sodium dodecyl sulfate and 0.1 M of NaOH). (f) The calculated cellular uptakes of **TTP**, **TBP**, **TBQ**, and **TBCP**,  $n = 3$ .