Supporting Information

An AIE photosensitizer with unquenched fluorescence based on nitrobenzoic acid for tumor-targeting and image-guided photodynamic therapy

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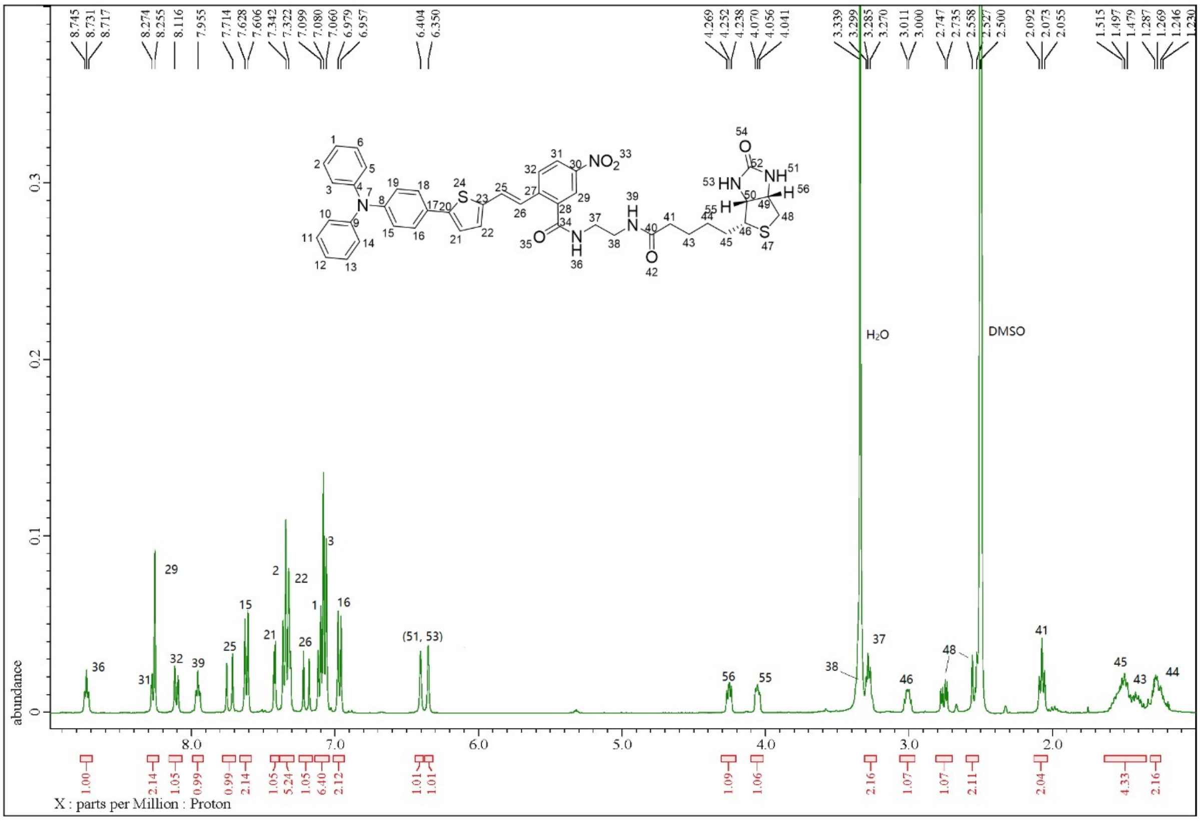
Synthesis of TTVBA: A mixture of 5-(4-(diphenylamino)phenyl)thiophene-2-carbaldehyde [1] (355 mg, 1.0 mmol), methyl 2-methyl-5-nitrobenzoate (293 mg, 1.5 mmol) and K_2CO_3 (414 mg, 3.0 mmol) in DMSO (10 mL) was stirred at 80 ^oC for 24 h and cooled to room temperature. Acetic acid aqueous solution (0.2 M, 40 mL) was added, and the resulting mixture was extracted with ethyl acetate. The extracts were combined, washed with H_2O and brine, dried over Na_2SO_4 and filtered. After removal of volatile components from the filtrate, the resulting crude product was purified by flash chromatography (silica, MeOH/CH_2Cl_2 = 1/9) to give **TTVBA** (440 mg, 85%) as a red solid, the ^1H NMR data is consistent with the reported.[2]

Synthesis of Biotin-TTVBA: To a solution of **TTVBA** (100 mg, 0.19 mmol), *N*-(2-aminoethyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide (67 mg, 0.23 mmol) and HBTU (110 mg, 0.29 mmol) in DMF (8 mL) was added DIEA (75 mg, 0.58 mmol), and the mixture was stirred at room temperature for 30 min. Water (30 mL) was added, and the resulting mixture was extracted with ethyl acetate. The extracts were combined, washed with H_2O and brine, dried over Na_2SO_4 and filtered. After removal of volatile components from the filtrate, the resulting crude product was purified by flash chromatography (silica, MeOH/CH_2Cl_2 = 1/9) to give **Biotin-TTVBA** (114 mg, 75%) as a red solid: ^1H NMR (400 MHz, DMSO-*d6*) δ 8.74 (t, J = 5.6 Hz, 1H), 8.27-8.25 (m, 2H), 8.12 (d, J = 8.8 Hz, 1H), 7.97 (t, J = 5.6 Hz, 1H), 7.75 (d, J = 16.0 Hz, 1H), 7.63 (d, J = 8.8 Hz, 2H), 7.42 (d, J = 3.6 Hz, 1H), 7.36-7.31 (m, 5H), 7.22 (d, J = 16.0 Hz, 1H), 7.12-7.06 (m, 6H), 6.98 (d, J = 8.8 Hz, 2H), 6.40 (s, 1H), 6.35 (s, 1H), 4.27-4.24 (m, 1H), 4.07-4.04 (m, 1H), 3.30-3.26 (m, 2H), 3.03-2.98 (m, 1H), 2.78 (dd, J = 12.4, 5.2 Hz, 1H), 2.56-2.53 (m, 2 H), 2.09 (t, J = 7.6 Hz, 2H), 1.60-1.36 (m, 4H), 1.30-1.23 (m, 2H); ^{13}C NMR (100 MHz, DMSO-*d6*) δ172.4, 166.8, 162.7, 147.3, 146.7, 145.2, 144.3, 141.3, 140.1, 136.3, 129.7, 128.0, 127.0, 126.6, 126.1, 124.5, 124.4, 123.7, 123.2, 122.6, 122.4, 61.0, 59.2, 55.4, 39.3, 38.2, 35.3, 28.2, 28.0, 25.2; HRMS (ESI) m/e calculated for $C_{43}H_{42}N_6O_5S_2Na$ $(M+Na)^+$ 809.2550, found 809.2527.



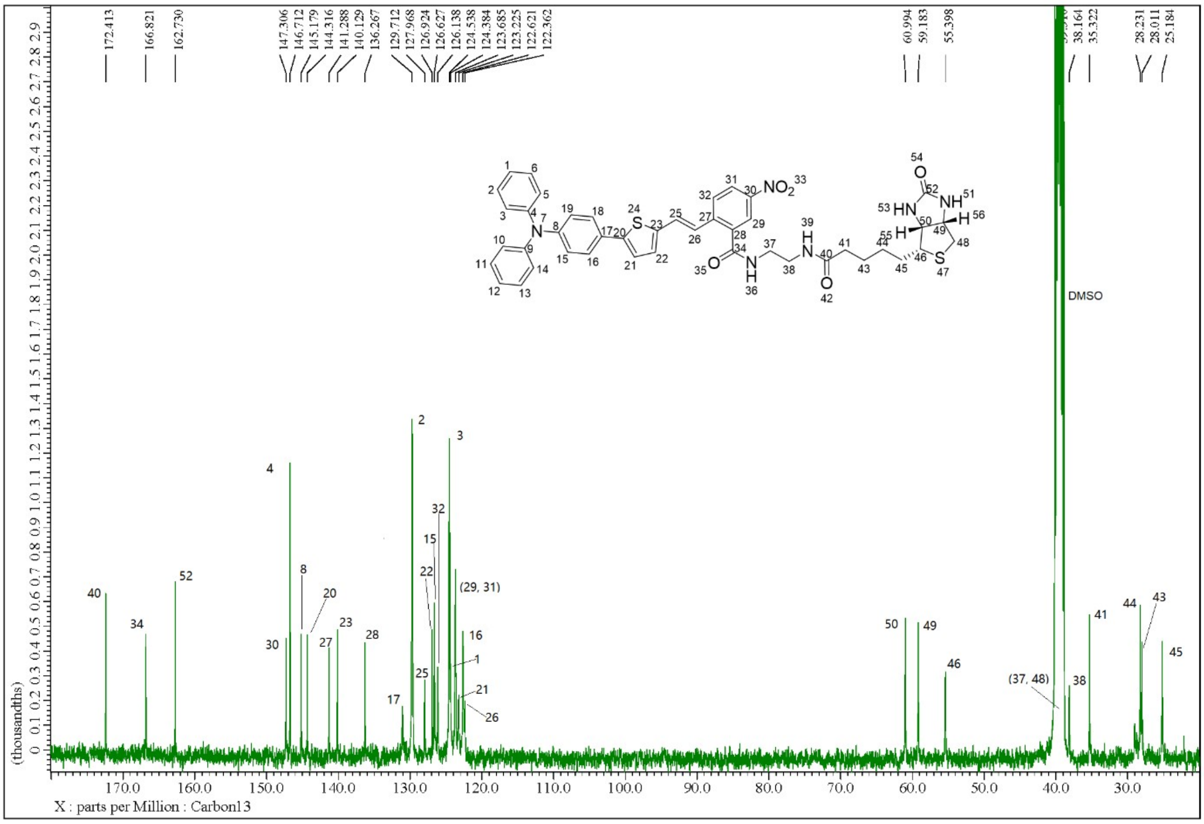


Fig. S1. ^{1}H NMR and ^{13}C NMR spectra of **Biotin-TTVBA**.

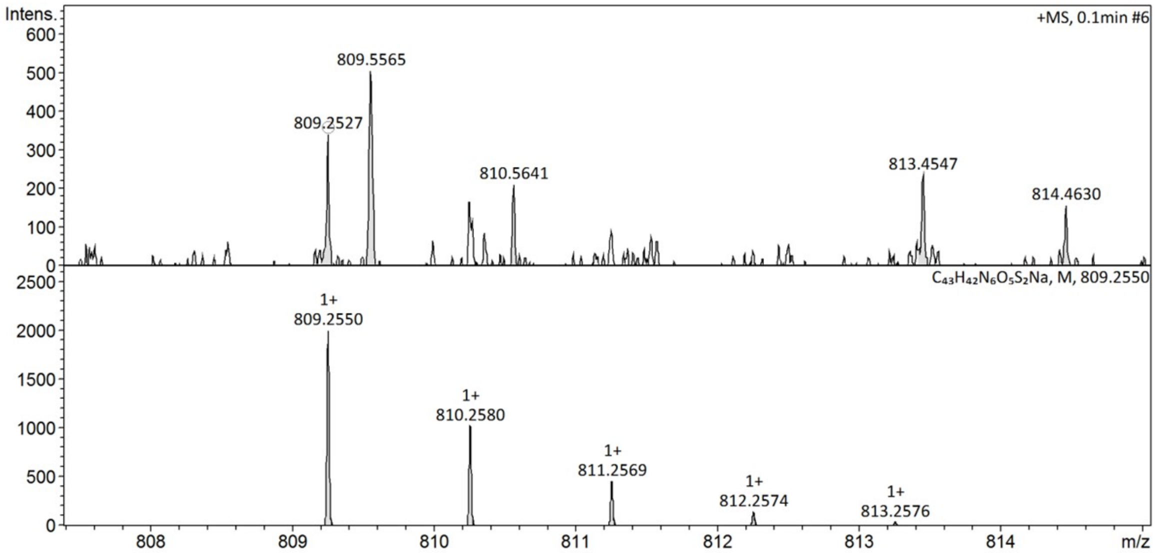
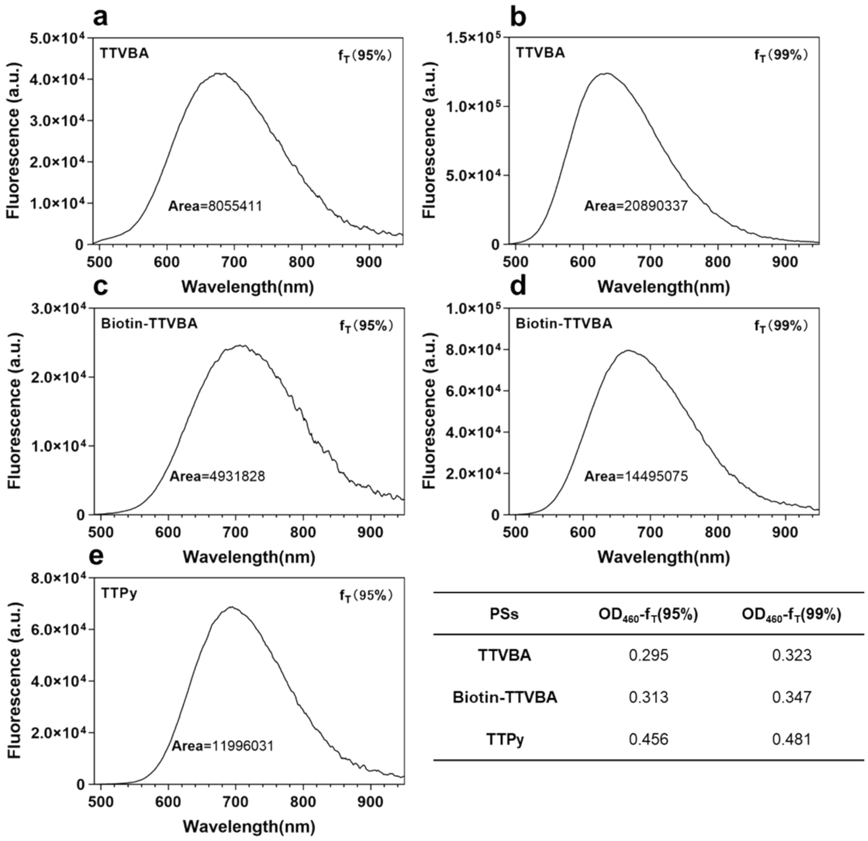


Fig. S2. HRMS spectrum of **Biotin-TTVBA**.



PSs	OD_{460}-f_T(95%)	OD_{460}-f_T(99%)
TTVBA	0.295	0.323
Biotin-TTVBA	0.313	0.347
TTPy	0.456	0.481

Fig. S3. The measurements of fluorescence quantum yield of (**a**) (**b**) **TTVBA** and (**c**) (**d**)**Biotin-TTVBA** (10 μM) in DMSO with 95% or 99% toluene (λ_{ex} = 460 nm), with (**e**) **TTPy** (Φ = 0.089) in DMSO with 95% toluene as a reference.

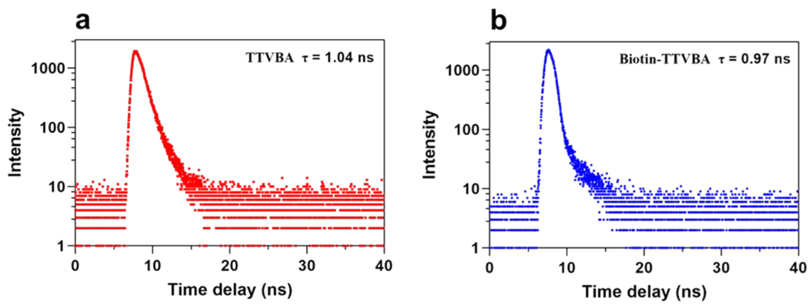
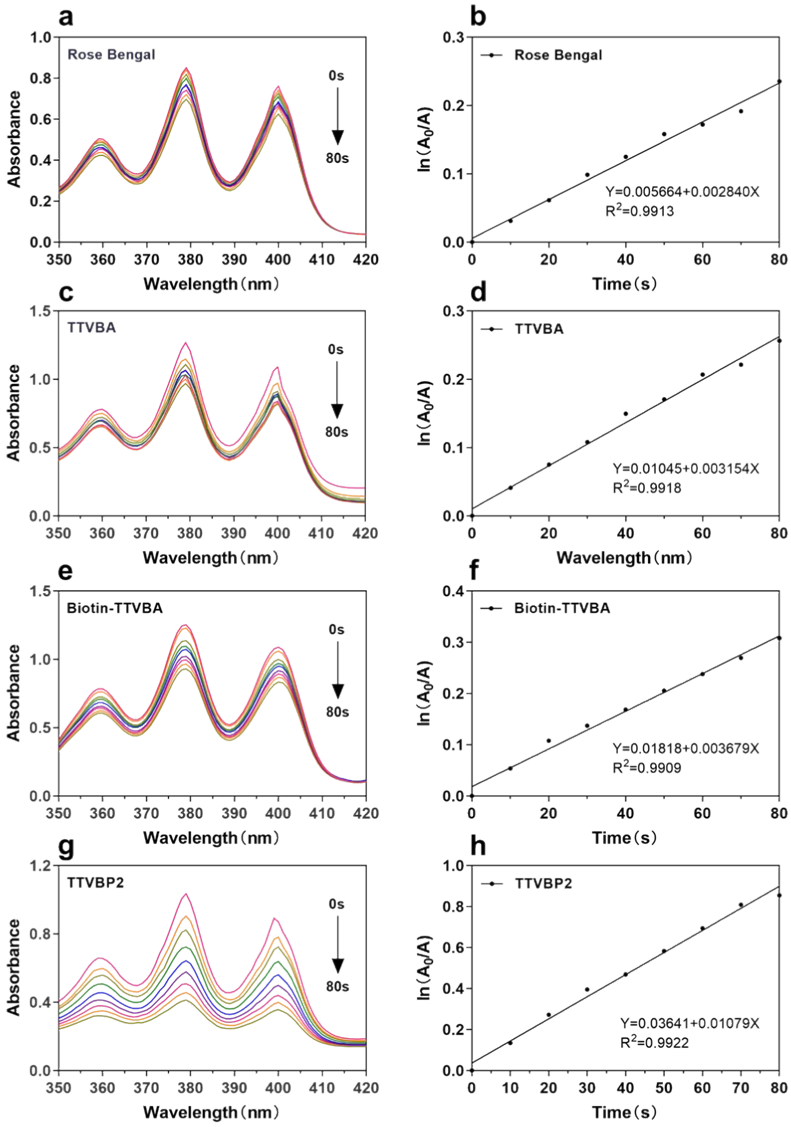
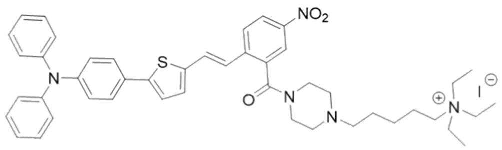


Fig. S4. Fluorescence decay curves of (**a**) **TTVBA** and (**b**) **Biotin-TTVBA** (10 μM) in DMSO with 99% toluene.





TTVBP2

PSs	Rose Bengal	TTVBA	Biotin-TTVBA	TTVBP2
OD_{415}	0.0227	0.1320	0.1429	0.1841

Fig. S5. The measurements of the singlet oxygen quantum yield. Photodegradation of ABDA with (**a**) Rose Bengal, (**c**) **TTVBA**, (**e**) **Biotin-TTVBA** and (**g**) **TTVBP2** under blue light irradiation (415 ± 5 nm, 20 mW/cm^2). The decomposition rate constants of ABDA by (**b**) Rose Bengal, (**d**) **TTVBA**, (**f**) **Biotin-TTVBA** and (**h**) **TTVBP2**.

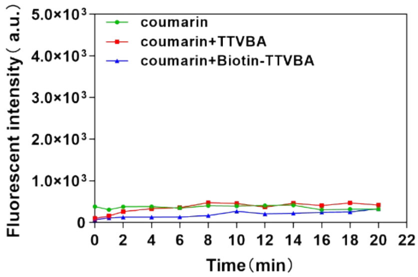


Fig. S6. Analysis of hydroxyl radicals generated by **TTVBA** or **Biotin-TTVBA** in PBS under white light (400 – 700 nm, 20 mW/cm^2) irradiation using coumarin (λ_{ex} = 360 nm, λ_{em} = 450 nm) as a specific probe.

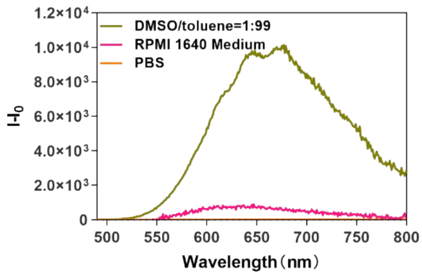


Fig. S7. Photoluminescence spectra of **Biotin-TTVBA** (10 µM) in different solvents (λ_{ex} = 458 nm).

References

1. D. P. Hagberg, X. Jiang, E. Gabrielsson, et al., *Journal of Materials Chemistry*, 2009, **19**, 7232-7238.
2. H. Wang, X. Pan, Y. Wang, et al., *Science China Materials*, 2021, **64**, 2601-2612.