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Supporting Information

Cancer-targeted Reactive Oxygen Species-degradable Polymer Nanoparticles for Near Infrared Light-induced Drug Release

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List of Abbreviations (according to sequence of appearance)

ROS	Reactive oxygen species
NIR	Near-infrared
MED	Minimum effective dose
MTD	Maximum tolerated dose
MPE	Maximum permissible exposure
PPADT	Methoxy poly(ethylene glycol)
mPEG	Methoxy poly(ethylene glycol)
mPEG-*b*-PPADT	mPEG-*block*-PPADT
IC_{50}	Half maximal inhibitory concentration
TPP	Meso-tetraphenylporphyrin
SiNc	Silicon 2,3-naphthalocyanine bis(trihexylsilyloxide)
Biotin-PEG-*b*-PPADT	Biotin-PEG- *block* -PPADT
TEA	triethylamine
Mw	Molecular weight
M_n	Number average molecular weight
PDI	Polydispersity index
s-PA	Micelles encapsulating SiNc
DMA	9,10-dimethylanthracene
NBT	Nitro blue tetrazolium
sp-PA	Micelles encapsulating SiNc and paclitaxel
PBS	Phosphate-buffered saline
bsp-PA	Biotin-decorated micelles encapsulating SiNc and paclitaxel
F-PA	Micelles labelled with fluorescein
bF-PA	Biotin-decorated micelles labelled with fluorescein
RPMI	Roswell Park Memorial Institute
bs-PA	Biotin-decorated micelles encapsulating SiNc
i.v.	Intravenous
BDT	1,4-benzenedimethanethiol
DMP	2,2-dimethoxypropane
TSP	3-(trimethylsilyl)propionic acid
FBS	Fetal bovine serum

Detailed Experimental Procedures

Materials. 1,4-benzenedimethanethiol (BDT) and paclitaxel were obtained from Tokyo Chemical Industry Co., Ltd (Tokyo, Japan). 2,2-dimethoxypropane (DMP), anhydrous ethyl acetate, *p*-toluenesulfonic acid (PTSA), anhydrous toluene, triethylamine (TEA) were purchased from Sigma-Aldrich Co. (St. Louis, MO, USA). From Sunbio, Inc. (Gyeonggi-do, Korea), methoxy poly(ethylene glycol) maleimide (mPEG maleimide, MW = 5 kDa) was acquired. Tetrahydrofuran (THF) were procured from Avantor Performance Materials, Inc. (Center Valley, PA, USA), chloroform from Junsei Chemical Co., Ltd. (Tokyo, Japan), and n-hexane from Samchun Pure Chemical Co (Gyeonggi-do, Korea). From Cambridge Isotope Laboratories, Inc (Tewksbury, MA, USA), chloroform-d ($CDCl_3$) containing (trimethylsilyl)propionic acid (TSP) was obtained.

Synthesis of PPADT. PPADT was synthesized by polycondensation using procedures similar to the previously reported procedures.[1-3] In brief, DMP (983 mg, 8.0 mmol) was added into a 250 mL two-neck flask filled with stirring anhydrous toluene (50 mL). PTSA (4.57 mg, 24 μmol) was dissolved in anhydrous ethyl acetate (457 μL) and added to the reaction solution at room temperature. The flask is then attached to a distillation head. BDT (1.36 g, 8.0 mmol) was added to the stirring mixture 1 h later and the temperature was increased to 95 °C for 30 min. During this process, a second monomer solution containing PTSA (2.73 mg, 12.4 μmol) in anhydrous ethyl acetate (273 μL) and DMP (499 mg, 4.8 mmol) were dissolved in anhydrous toluene (12 mL) and stirred for 30 min at room temperature. This second monomer solution was added to the hot reaction solution dropwise at 28 μL min^{-1} over 12 h. The reaction was continued at 95 °C for another 24 h while the brown solution became darker. To obtain the brown polymer, the reaction solution was precipitated in cold *n*-hexane and vacuum dried overnight. ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 1.60-1.63 (*br*, 6H of thioketal of PPADT); 3.82-3.84 (*br*, 4H of methylene of PPADT); and 7.15-7.35 (*br*, 4H of benzene of PPADT).

Synthesis of mPEG-*b*-PPADT. The synthesis of mPEG-b-PPADT was the same as same as previously reported procedure.[1] Briefly, in a 100 mL one-neck flask, PPADT (50 mg, 7.4 μmol) and TEA (2.06 μL, 1.49 μmol) were dissolved in chloroform (10 mL) while mPEG-maleimide (5 kDa, 36.8 mg, 7.4 μmol) was dissolved in chloroform (10 mL) in a glass vial separately. The mPEG-maleimide solution was added to the PPADT solution dropwise over an hour at room temperature. The reaction continued for another 2 h before chloroform was evaporated. After which, the off-white solid was sonicated in degassed deionized water (20 mL) to extract mPEG-*b*-PPADT while PPADT precipitated. The off-white suspension was filtrated through a hydrophilic polyvinylidene fluoride membrane (pore size: 0.22 μm, GVWP04700, Merck Millipore Ltd., Co., Cork, Ireland). The collected filtrate was dialyzed against degassed deionized water using regenerated cellulose (RC) membrane for 2 days to remove unreacted mPEG-maleimide (molecular weight cut-off, MWCO = 25 kDa). The purified filtrate was lyophilized over 2 days. ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm): 1.60-1.63 (*br*, 6H of thioketal of PPADT); 3.61-3.64 (*br*, 4H of PEG); 3.81-3.83 (*br*, 4H of methylene of PPADT); and 7.23-7.29 (*br*, 4H of benzene of PPADT).

Molecular Characterization. The synthesized polymers was analyzed by gel permeable chromatography (GPC) equipped with a Styragel® HR THF 7.8 × 300 mm column to calculate their number and weight average molecular weight. THF was the eluent and the flow rate was set at 1.0 mL min^{-1}. Calibration curve was drawn from analyzed polystyrene standards and the analyzed samples were compare with it. For ^{1}H NMR analysis (NMR Varian VXR-4000 (400 MHz) spectrometer), PPADT and mPEG-*b*-PPADT were dissolved in $CDCl_3$. The chemical shifts of each chemical group were measured in part per million (δ values) against trimethylsilyl chloride (TMS) as the internal standard at 25 °C, and the coupling constants in Hertz. The functional groups in the block copolymers were identified by Fourier transform-infrared spectroscopy (FT-IR; FT/IR-6100, JASCO Analytical Instruments, MD, USA).

Supplementary Table 1. SiNc and paclitaxel loading yield and encapsulation efficiency in sp-PA and bsp-PA.

	Components	Loading yield (%)	Encapsulation efficiency (%)
sp-PA	SiNc	4.82	95.1
	Paclitaxel	1.07	37.8
bsp-PA	SiNc	4.84	95.7
	Paclitaxel	1.04	36.5

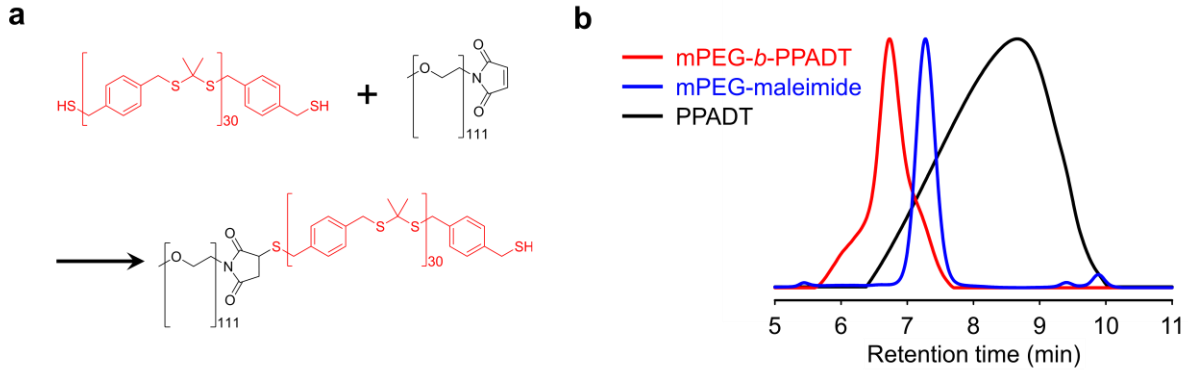


Figure S1. (a) Synthesis scheme of ROS-sensitive mPEG-b-PPADT. (b) GPC analysis of PPADT, mPEG-melaimide, and mPEG-*b*-PPADT.

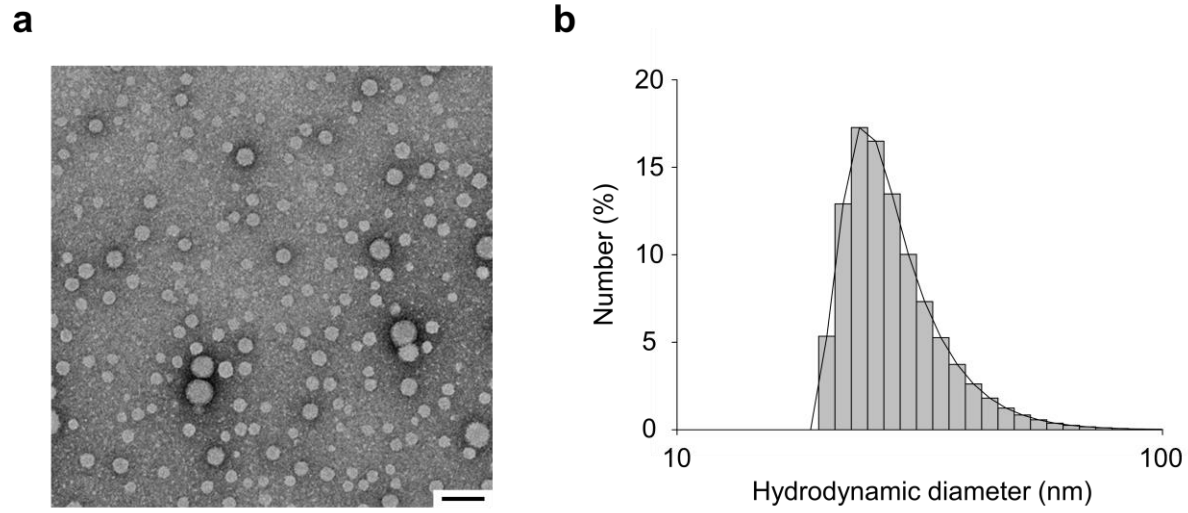


Figure S2. TEM image (scale bar = 100 nm) (a) and hydrodynamic diameter distribution (b) of s-PA.

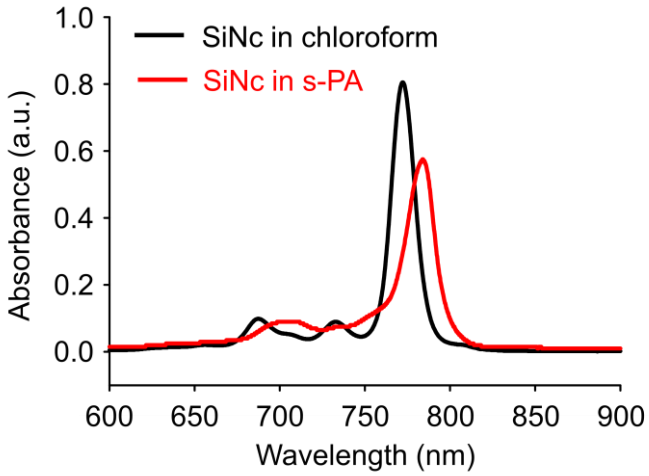


Figure S3. Absorption of SiNc (2.41 μg mL^{-1}) in chloroform (black) and s-PA (red).

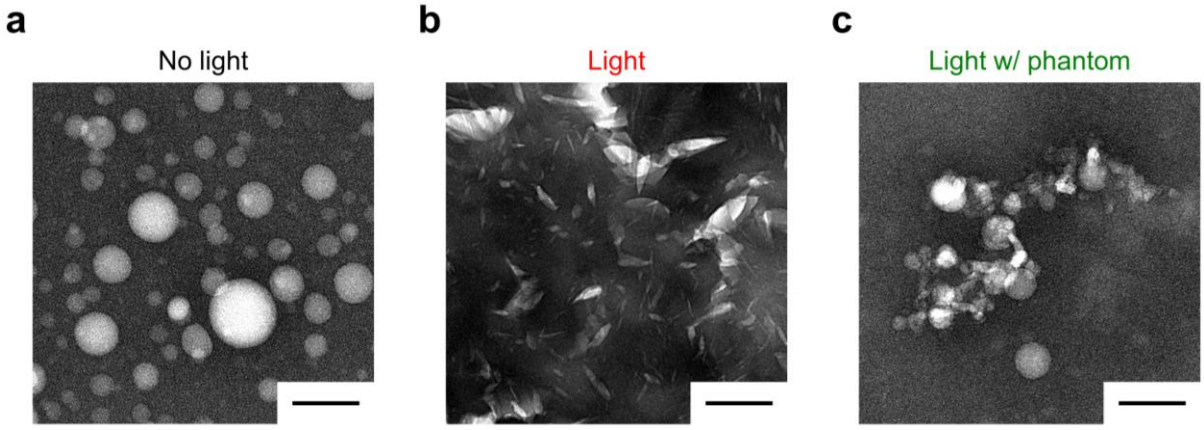


Figure S4. TEM images of s-PA without light irradiation (a), with light irradiation in the absence (b) and presence of tissue-like phantom (c) (scare bar: 100 nm).

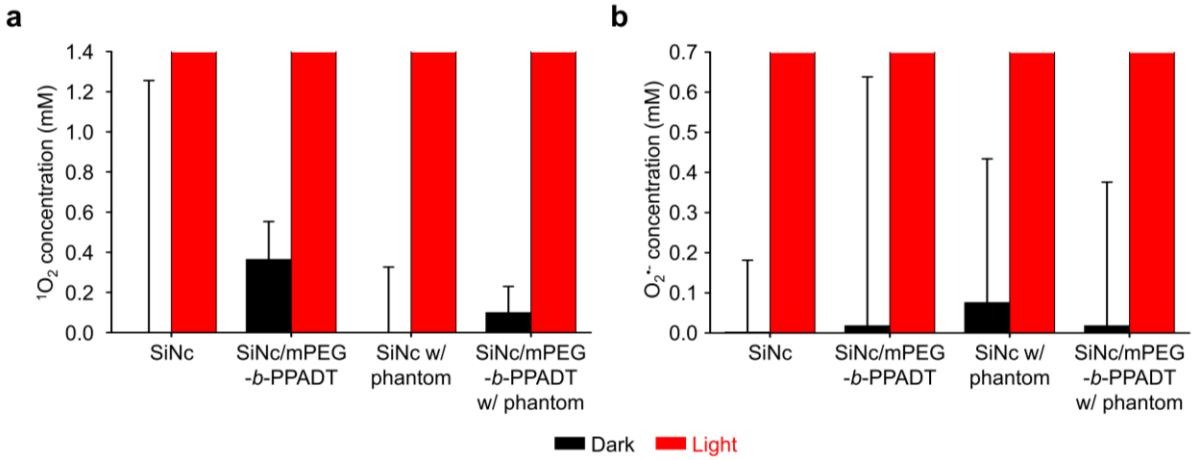


Figure S5. NIR light-induced ROS generation. Concentration of $^1O_2$ (a) and $O_2^{\bullet-}$ (b) generated by SiNc in the presence of mPEG-*b*-PPADT under light illumination (808 nm, 300 mW cm^{-2}, 20min) and in the presence of tissue-like phantoms.

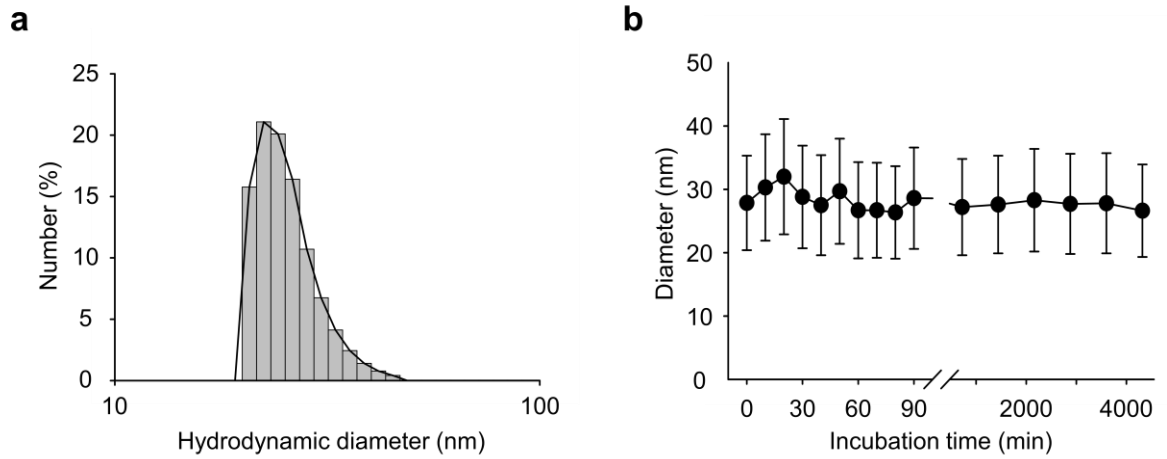
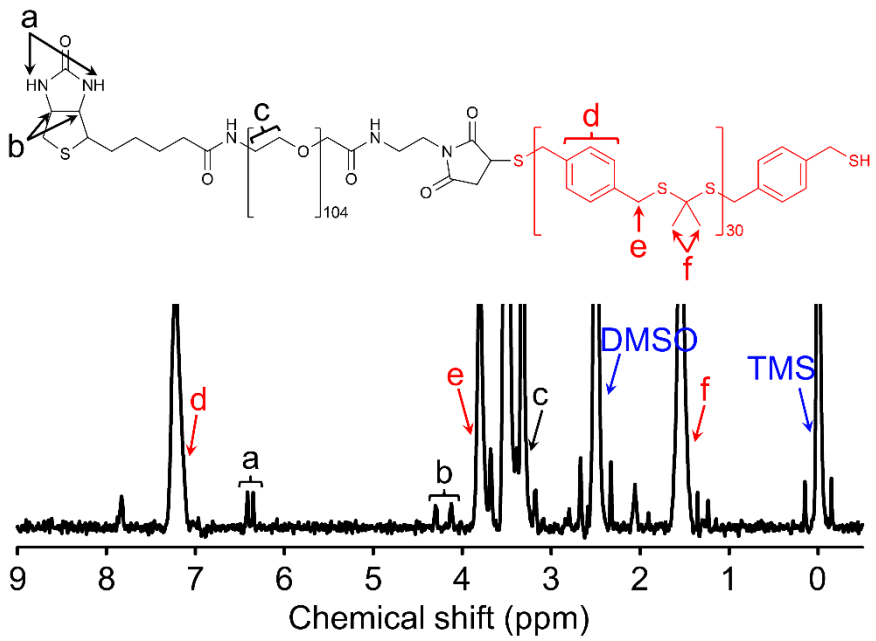


Figure S6. (a) Hydrodynamic diameter distribution of sp-PA in PBS containing 10 % FBS. (b) Dispersion stability of sp-PA in PBS containing 10% FBS at 37 °C over three days.

a



b

Peaks	d	f	b
No. of H in each functional group	4	6	2
No. repeating unit per polymer chain	30	30	104
Relative area	1	1.35	0.234
Relative no. of biotin-PEG-*b*-PPADT	1	0.898	1.41

Figure S7. (a) ^{1}H NMR spectrum of biotin-PEG-*b*-PPADT (400 MHz, DMSO-D_6) δ (ppm): 1.52-1.55 (6H of thioketal of PPADT); 3.32-3.51 (4H of PEG); 3.78-3.81 (4H of methylene of PPADT); 4.12 and 4.30 (2H of methylene of biotin); 6.35 and 6.41 (2H of urea of biotin); and 7.22-7.26 (4H of benzene of PPADT). (b) Relative areas of each peak in the ^{1}H NMR spectrum in (a).

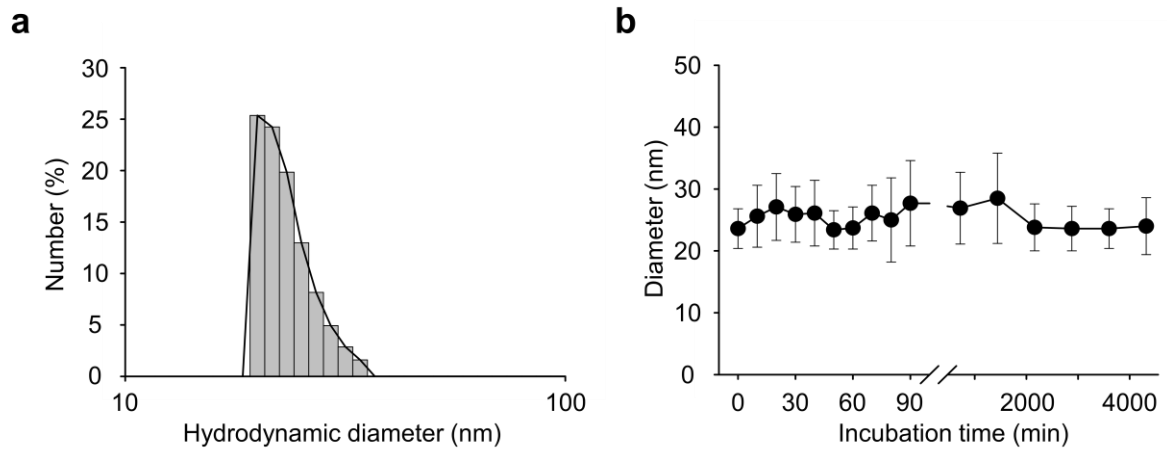


Figure S8. (a) Hydrodynamic diameter distribution of bsp-PA in PBS containing 10 % FBS. (b) Dispersion stability of bsp-PA in PBS containing 10% FBS at 37 °C over three days.

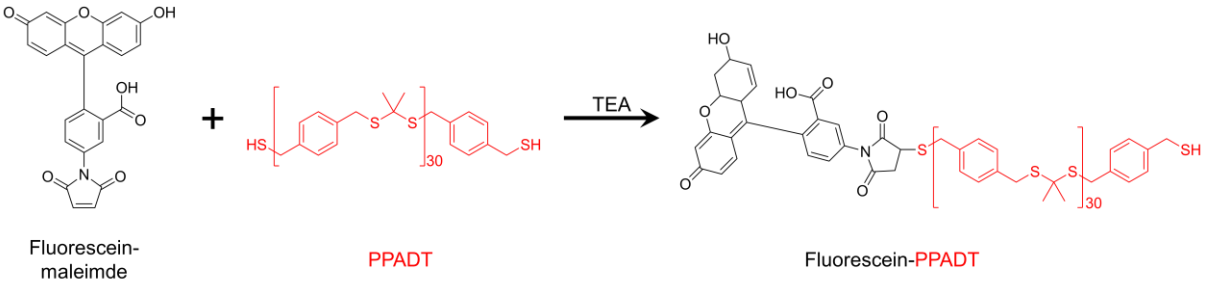


Figure S9. Synthesis scheme of fluorescein-PPADT.

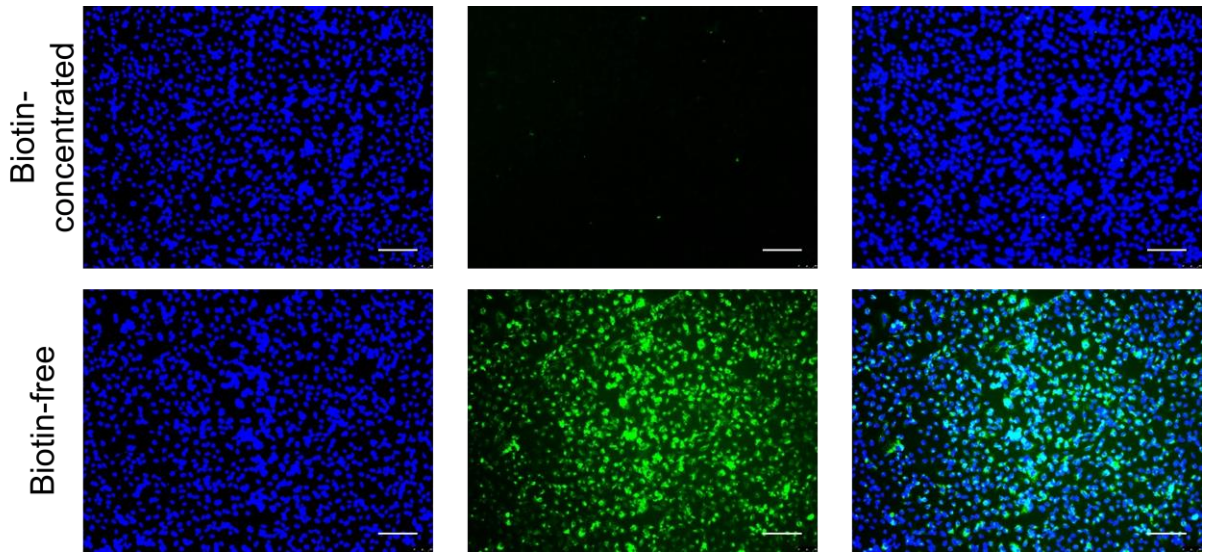


Figure S10. Fluorescence microscopic images of bF-PA in biotin-free and biotin-concentrated (8.19 mM) DMEM (blue: DAPI stained nuclei; green: fluorescein-labelled micelles; and scale bar = 150 μm).

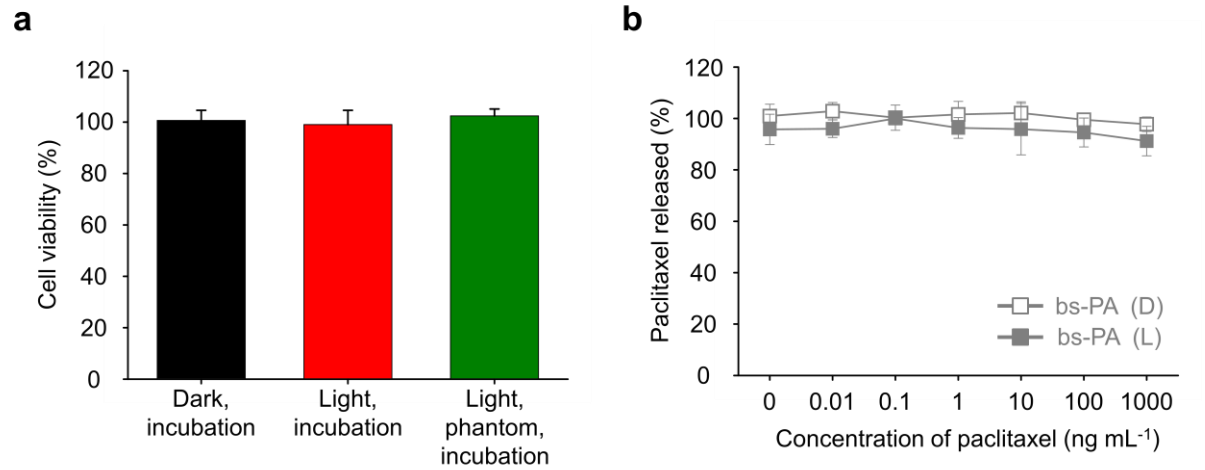


Figure S11. A549 cell viability profiles of NIR light (808 nm, 300 mW cm^{-2}, 20 min) (a) and bs-PA without and with light illumination (808 nm, 300 mW cm^{-2}, 20 min).

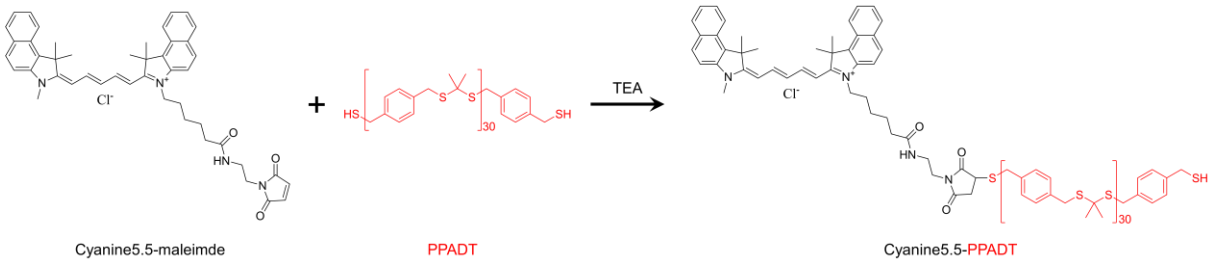


Figure S12. Synthesis scheme of cyanine5.5-PPADT.

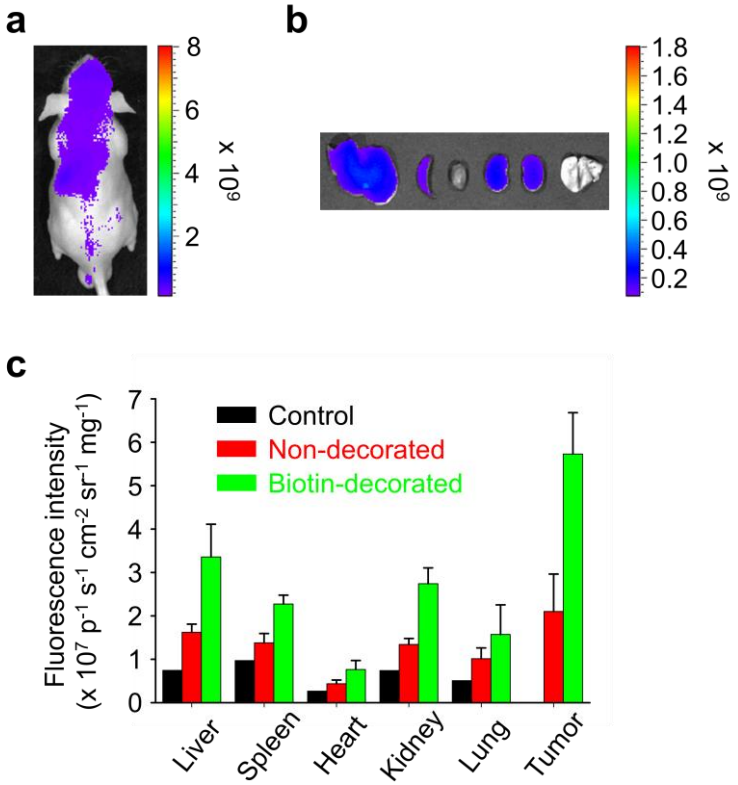


Figure S13. (a) *In vivo* cyanine 5.5 fluorescence images of live Balb/c nude mouse. (b) Representation cyanine 5.5 fluorescence images of dissected organs (liver, spleen, heart, kidneys, lungs and respectively) of mouse. (c) Comparison of cyanine 5.5 fluorescence intensity in each dissected organs and tumors of mice injected with non-decorated and biotin-decorated micelles with those of control mouse without injection.

References

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2. J. S. Kim, S. D. Jo, G. L. Seah, I. Kim and Y. S. Nam, *J. Ind. Eng. Chem.*, 2015, **21**, 1137-1142.
3. D. S. Wilson, G. Dalmasso, L. Wang, S. V. Sitaraman, D.; Merlin and N. Murthy, *Nat. Mater.*, 2010, **9**, 923-928.