Supporting Information

Charge-Reversal Polymeric Nanomodulators for Ferroptosis-Enhanced Photodynamic Therapy

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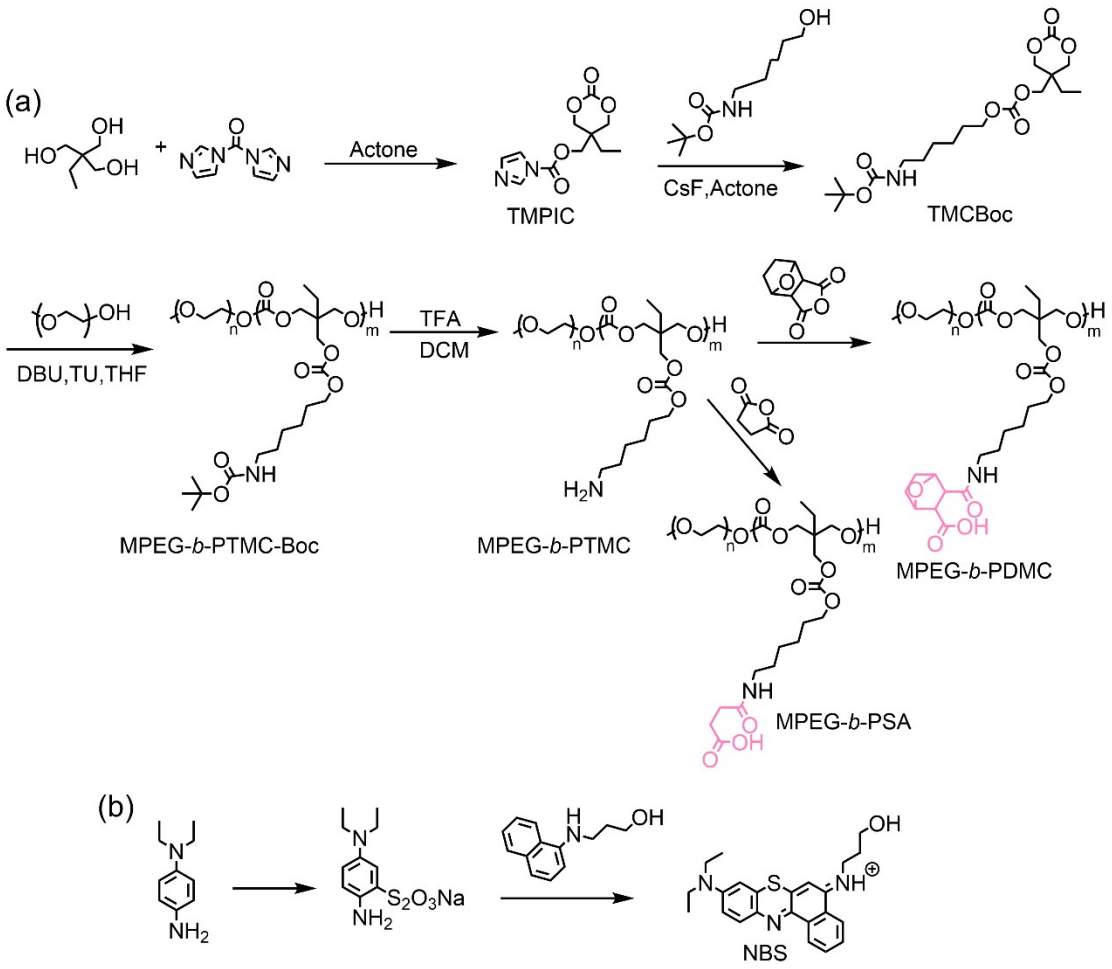
1. Materials

The 1-aminonaphthalene (99%), N,N-diethyl-p-phenylenediamine (99%), 1,1,1-trimethylolpropane (98%), 1,1'-carbonyldiimidazole (CDI) (97%), 6-(Boc-amino)-1-hexanol (95%) were obtained from J&K Chemical LTD. 1,8-diazabicyclo [5.4.0] undec-7-ene (DBU) (99%), and polyethylene glycol monomethyl ether (MPEG, M_n = 5.0 kg/mol) were purchased from Aladdin (Shanghai, China). The TU catalyzer and TMPIC were synthesized according to our previous work.[1] The MTT and calcein-AM/propidium iodide (PI) detection kit were purchased from Beyotime Biotechnology Co. All other reagents were widely available commercially. The 4T1, MCF-7, and HepG2 cells were purchased from the Institute of Basic Medical Sciences (IBMS) of the Chinese Academy of Medical Sciences.

2. Methods

^{1}H NMR and ^{13}C NMR spectra were performed with Bruker Avance III 500 spectrometers. Mass spectrometric (MS) data was carried out using LTQ Orbitrap XL instruments. Absorption and emission spectra were performed with a UV Vis spectrophotometer (Lambda 750S) and a VARIAN CARY Eclipse fluorescence spectrophotometer (Serial No. FL0812 M018), respectively. The weight average molecular weight and distribution were determined using the Waters 1515 HPLC system with tetrahydrofuran (THF) as the eluent. Transmission electron microscopy (TEM) images were measured on HT7700 EXALENS. The diameter of the nanoparticles was determined by dynamic light scattering (DLS) on Malvern Zetasizer Nano ZS90 (Malvern, UK). The light source was 660 nm LED (GH-BT-22W, Shenzhen Guangyuanhong Technology). The output power of the laser was measured by a power meter (LP100/TS15, Changchun New Industries Optoelectronics Technology). Con focal laser scanning microscope (CLSM) images were performed on an Olympus FV3000 confocal laser scanning microscope.



Scheme S1. The synthesis process of MPEG-*b*-PDMC and NBS.

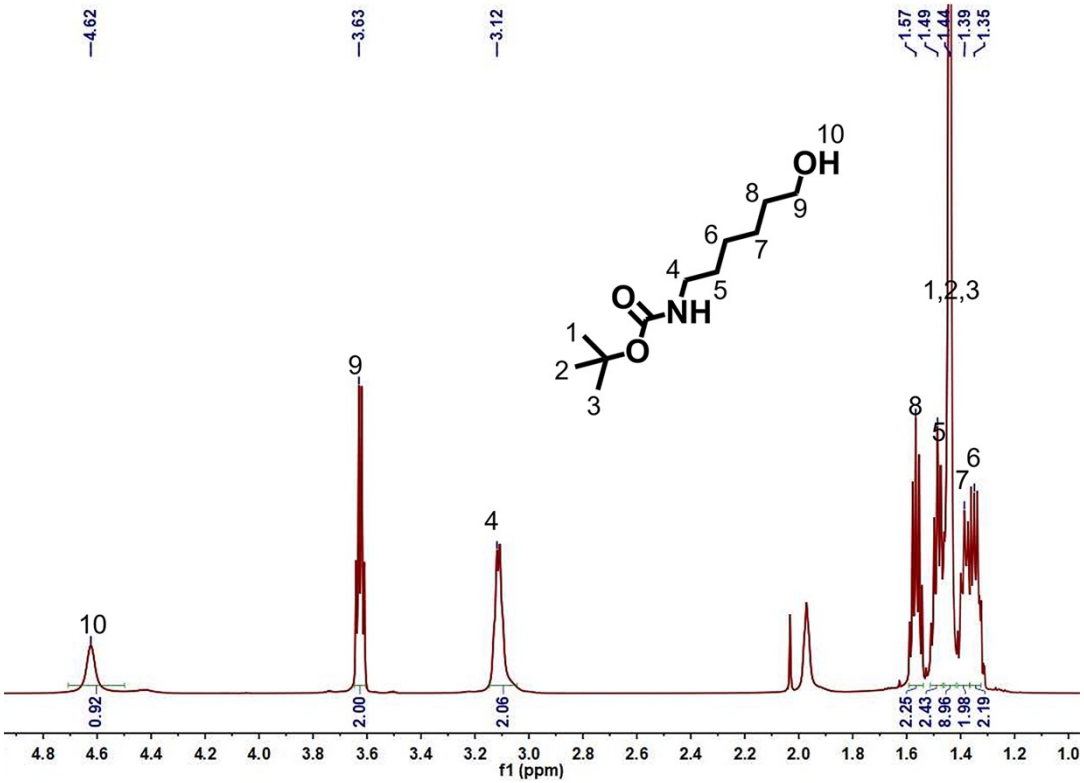


Figure S1. ^{1}H NMR spectrum of HO-Boc (500 MHz, $CDCl_3$).

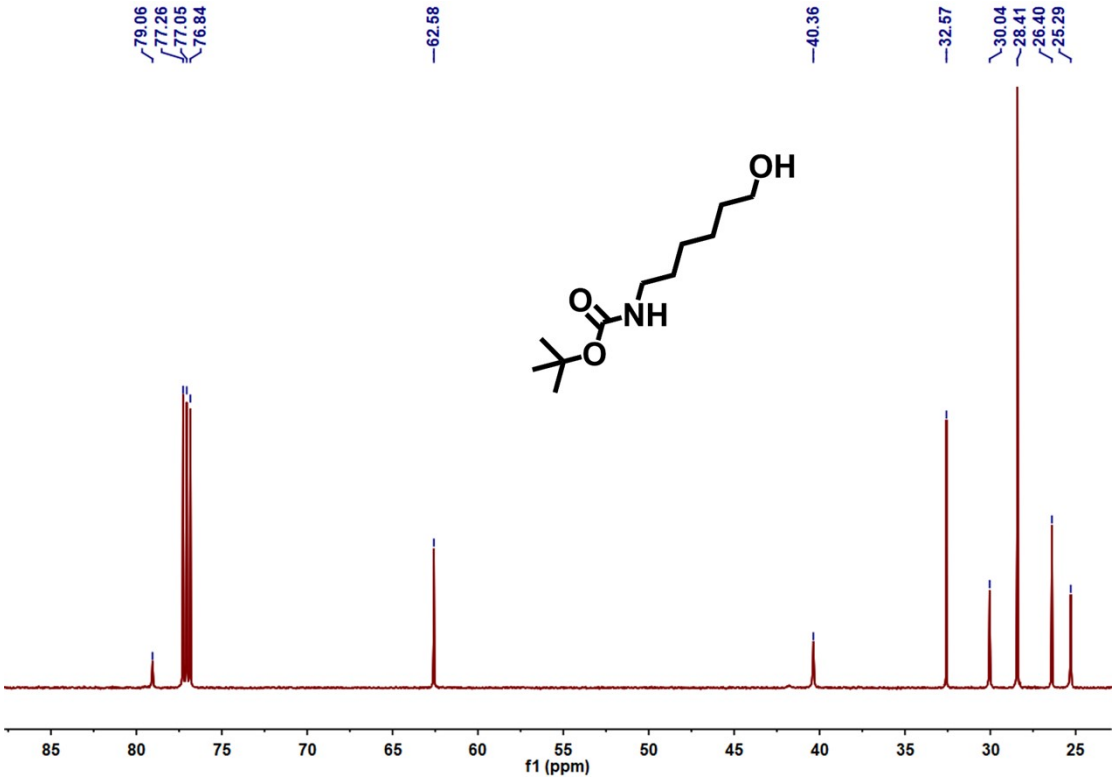
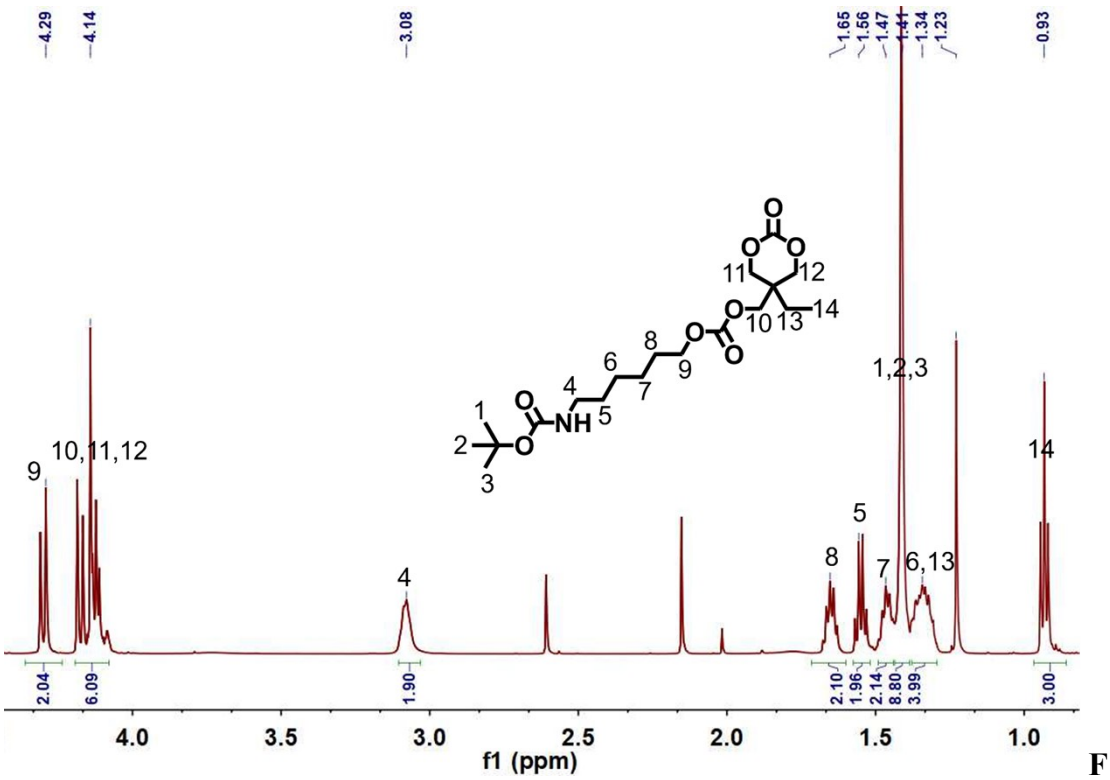


Figure S2. ^{13}C NMR spectrum of HO-Boc (125 MHz, $CDCl_3$).



igure S3. ^{1}H NMR spectrum of TMC-Boc (500 MHz, $CDCl_3$).

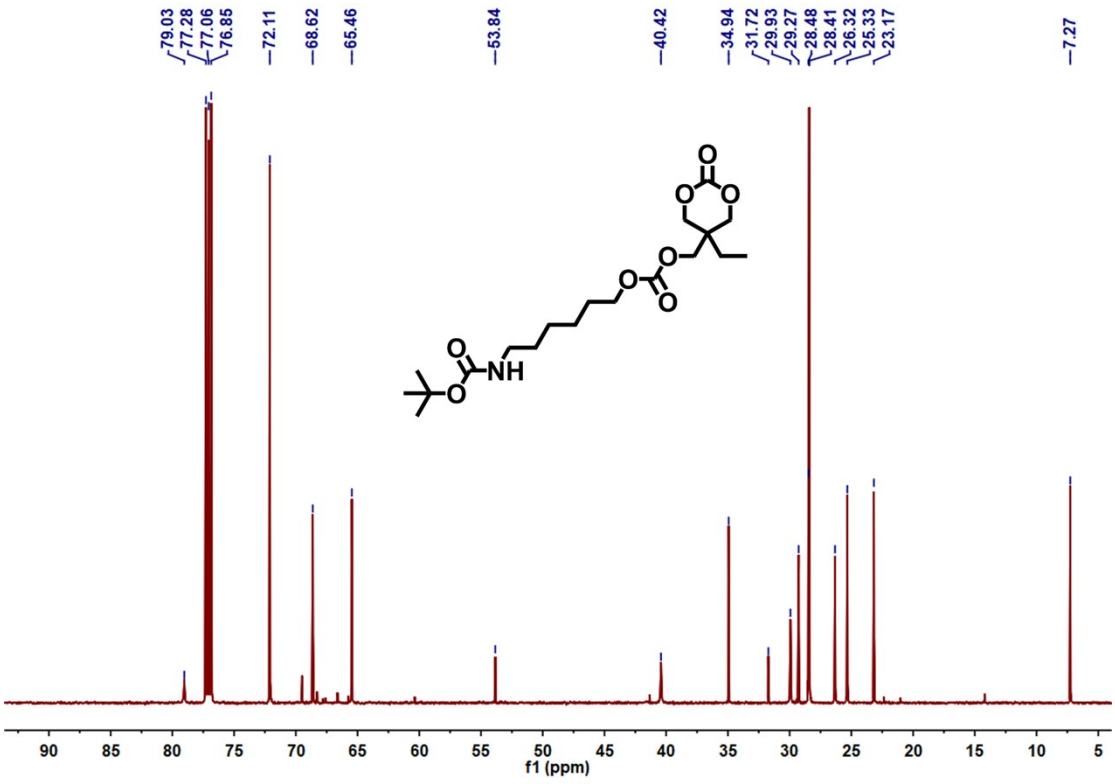


Figure S4. ^{13}C NMR spectrum of TMC-Boc (125 MHz, CDCl3).

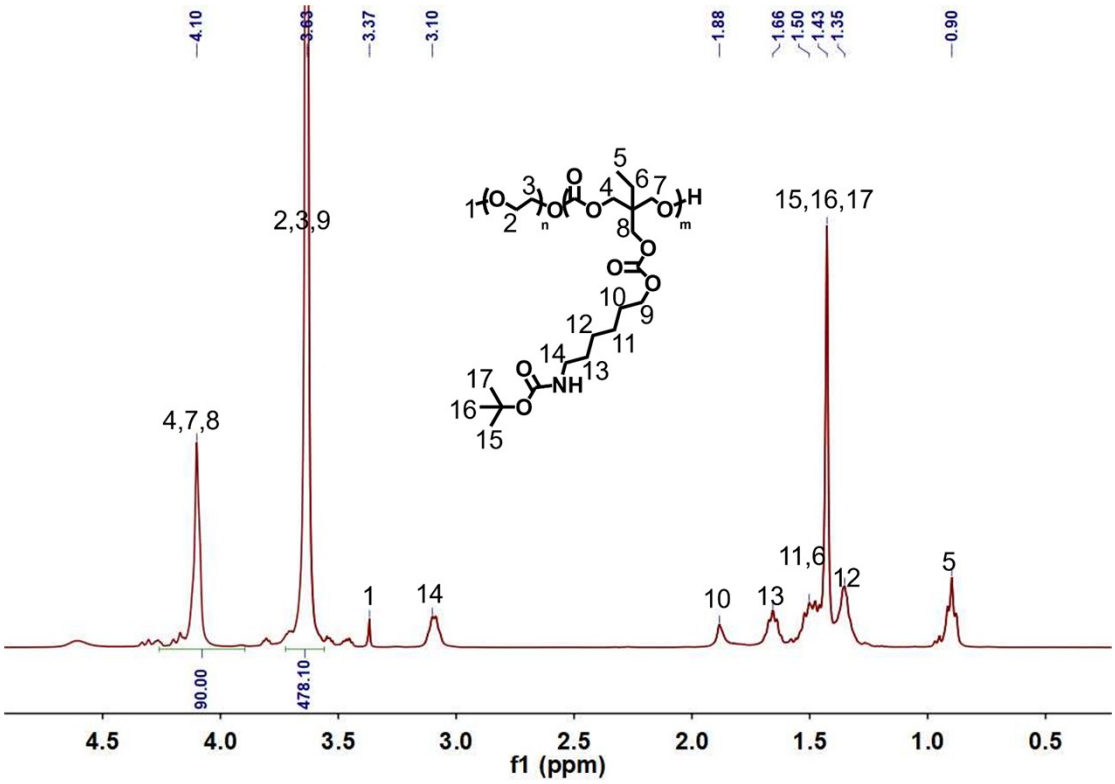


Figure S5. ^{1}H NMR spectrum of MPEG-*b*-PTMAC-Boc (500 MHz, $CDCl_3$).

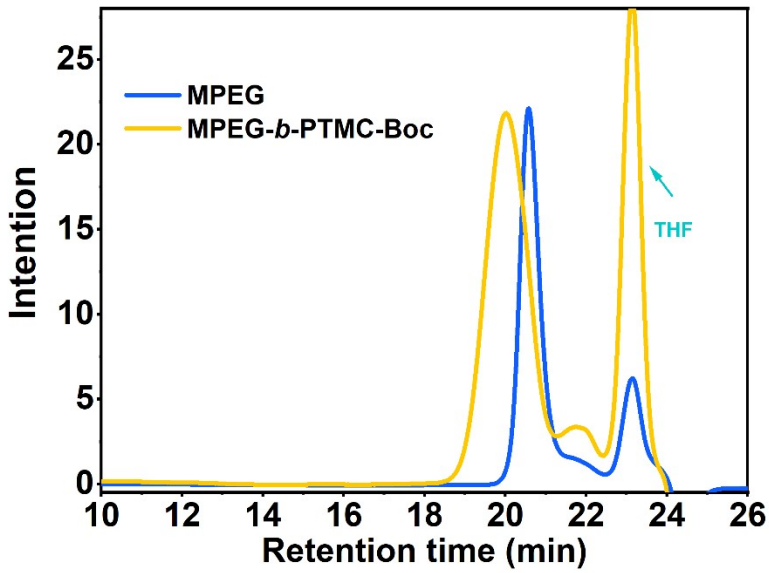


Figure S6. GPC traces for MPEG and MPEG-*b*-PTMC-Boc. A clear peak shift to the higher molecular weight of MPEG-*b*-PTMC-Boc demonstrated successful polymerization.

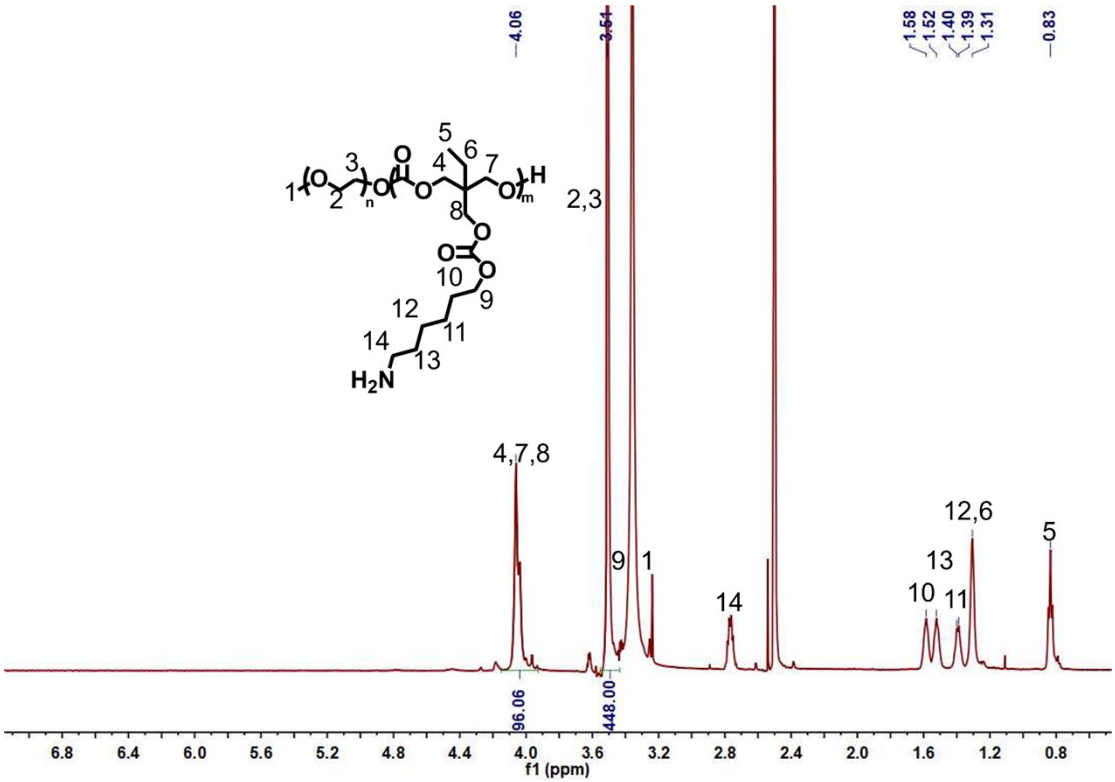


Figure S7. ^{1}H NMR spectrum of MPEG-*b*-PTMC (500 MHz, $(CD_3)_2SO$).

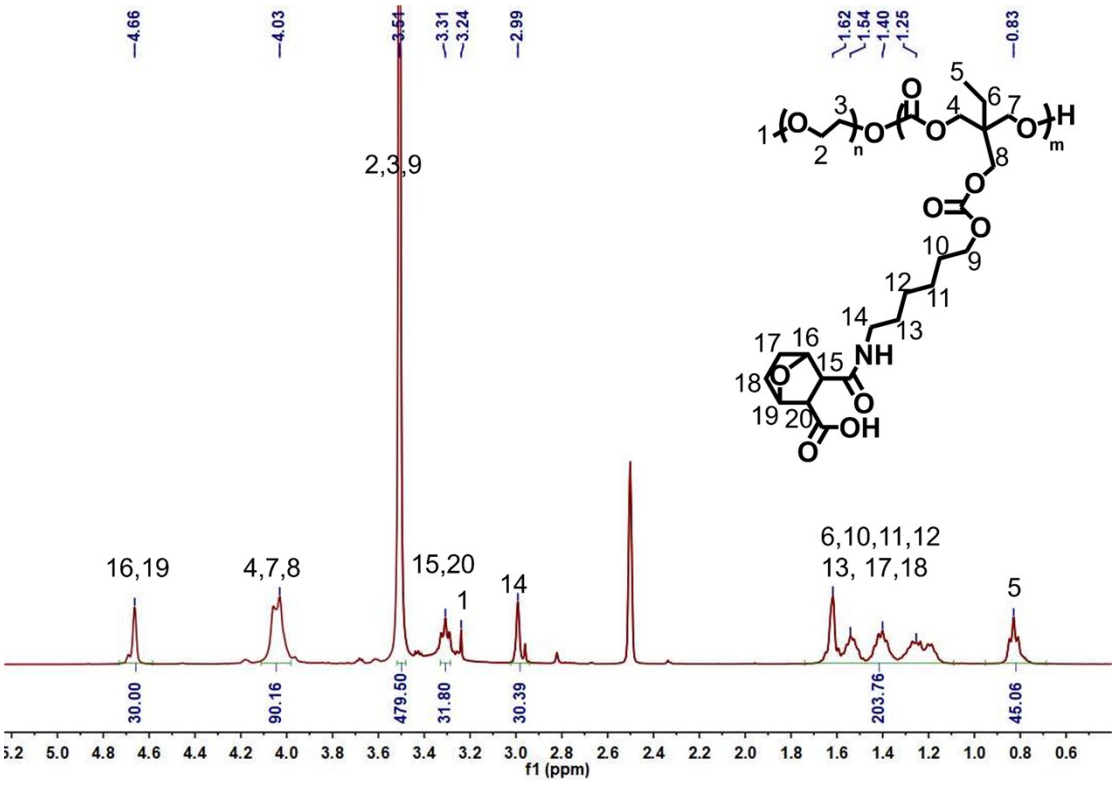


Figure S8. ^{1}H NMR spectrum of MPEG-*b*-PDMC (500 MHz, $(CD_3)_2SO$). The MPEG

contained 112 repeating units of -(CH_2-CH_2-O)-, which corresponds to 448 protons on each MPEG-*b*-PDMC chain. Accordingly, the number of DMC was calculated as 15 in each polymer chain, and thus the molecular weight of MPEG-*b*-PDMC is about 12 kg mol^{-1}. The drug loading content of DMC = [186.05 * 15] / [5000 + 471.5 * 15] * 100= 23.1%.

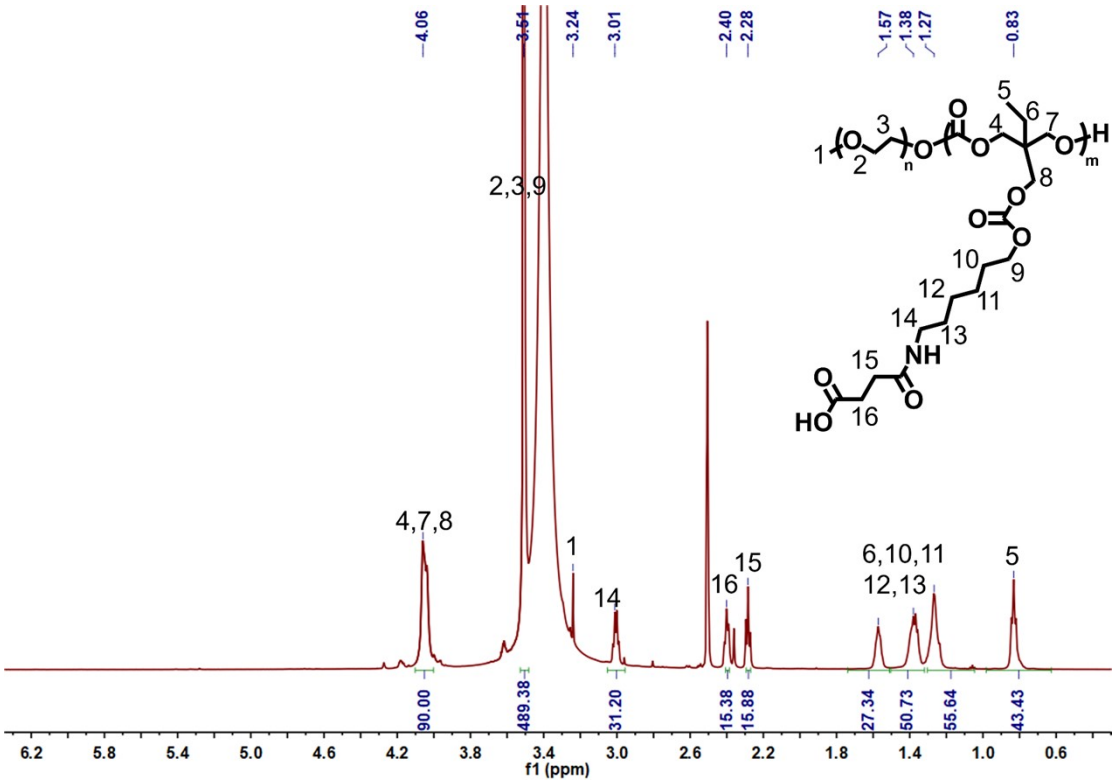


Figure S9. ^{1}H NMR spectrum of MPEG-*b*-PSA (500 MHz, $(CD_3)_2SO$).

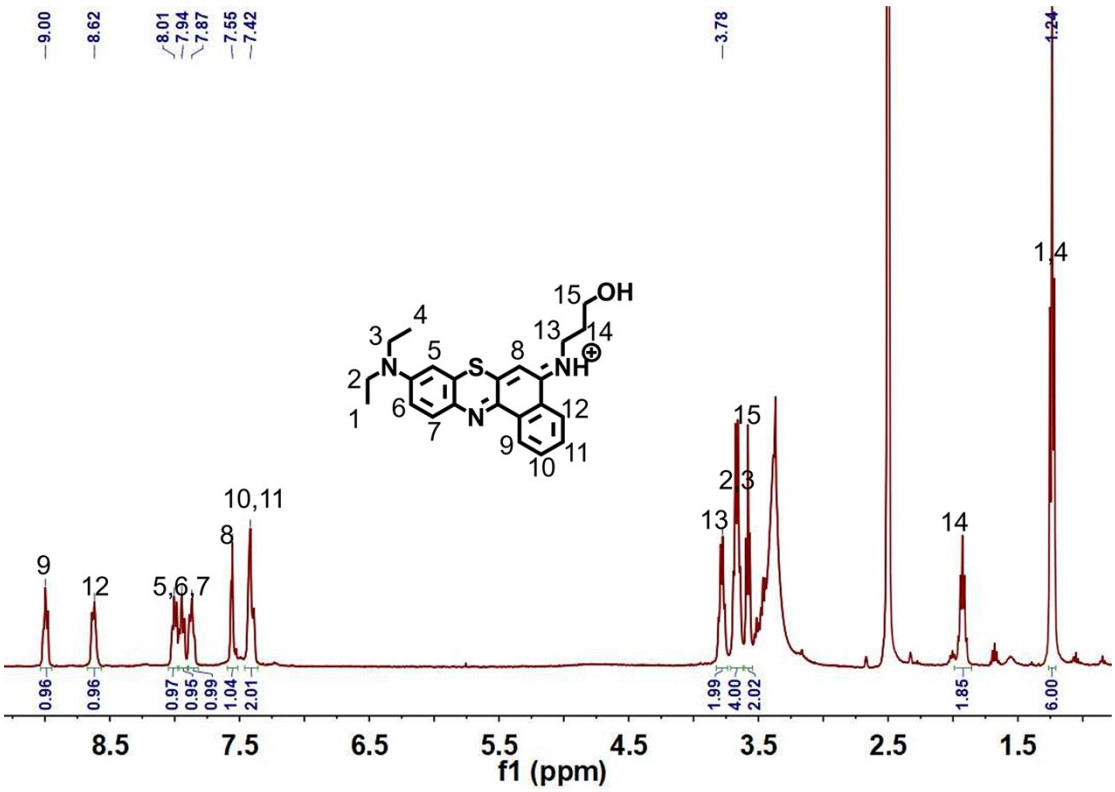


Figure S10. ^{1}H NMR spectrum of NBS (500 MHz, $(CD_3)_2SO$).

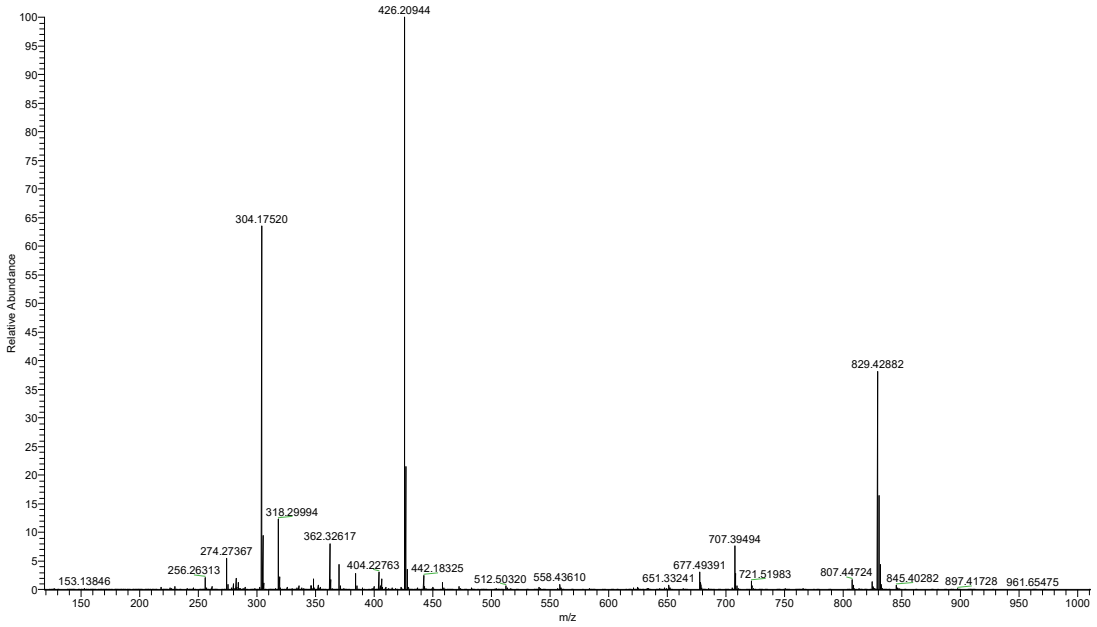


Figure S11. ESI-MS spectrum of TMC-Boc.

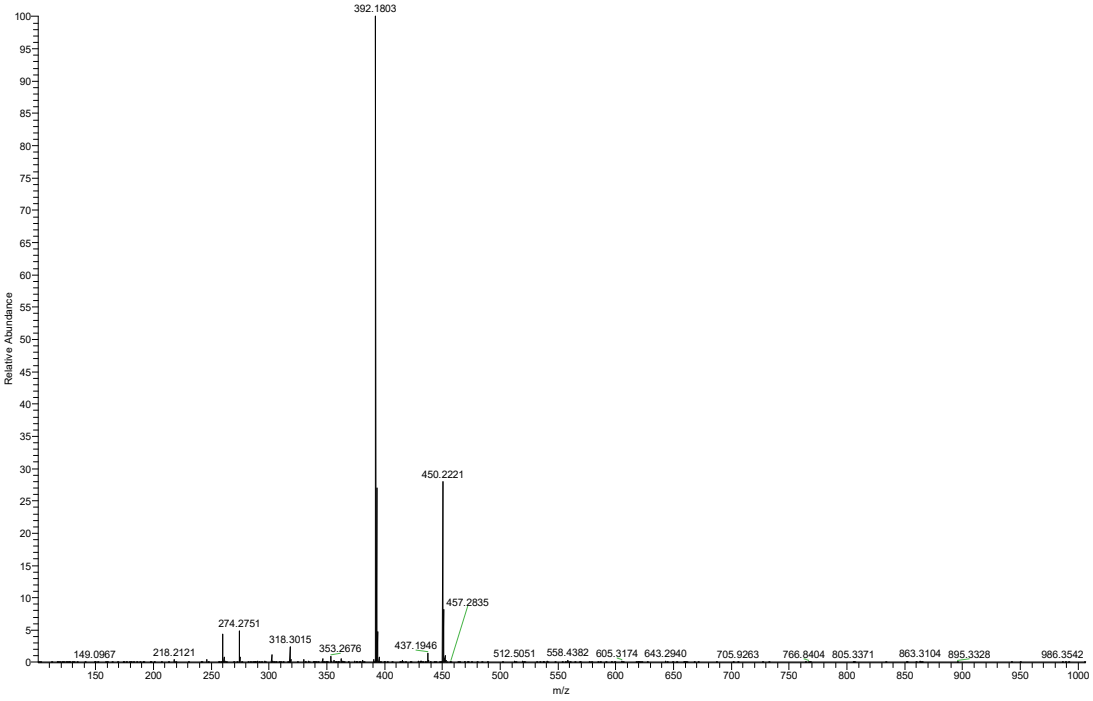


Figure S12. ESI-MS spectrum of NBS.

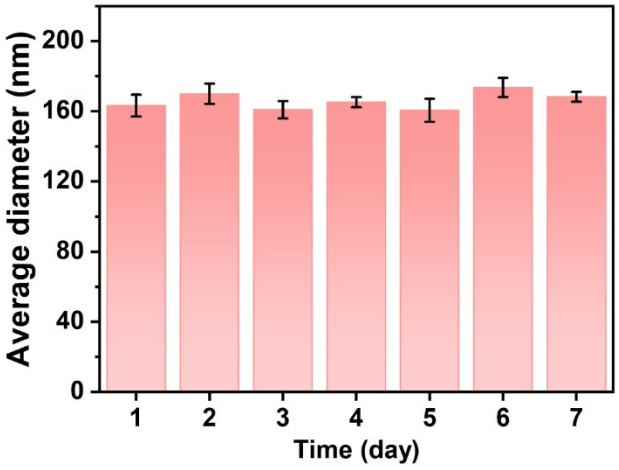


Figure S13. The particle size of NBS-PDMC NPs changes with time in PBS.

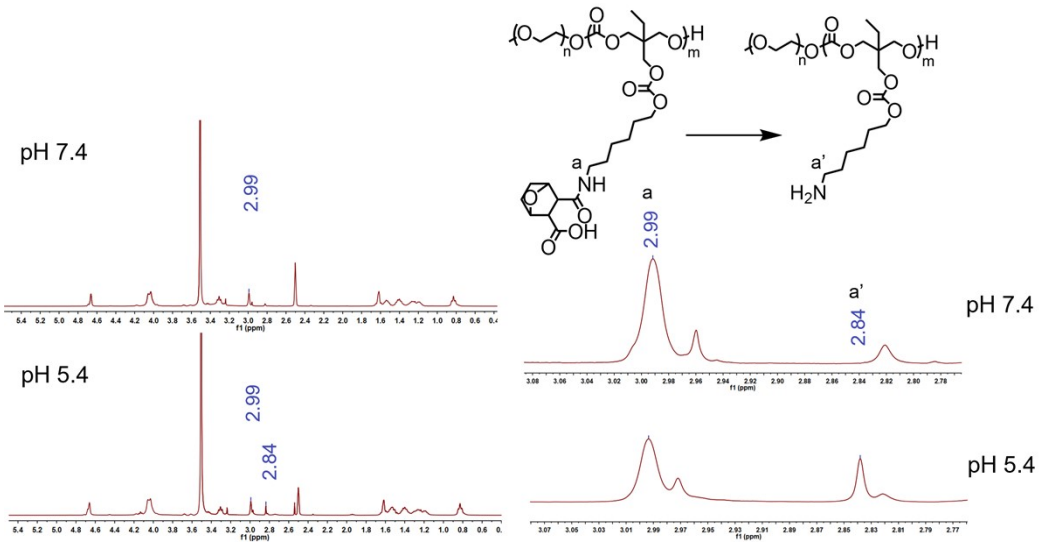


Figure S14. The hydrolysis of MPEG-b-PDMC at 5.4 for 4 h was detected by ^{1}H NMR spectroscopy (500 MHz, $(CD_3)_2SO$).

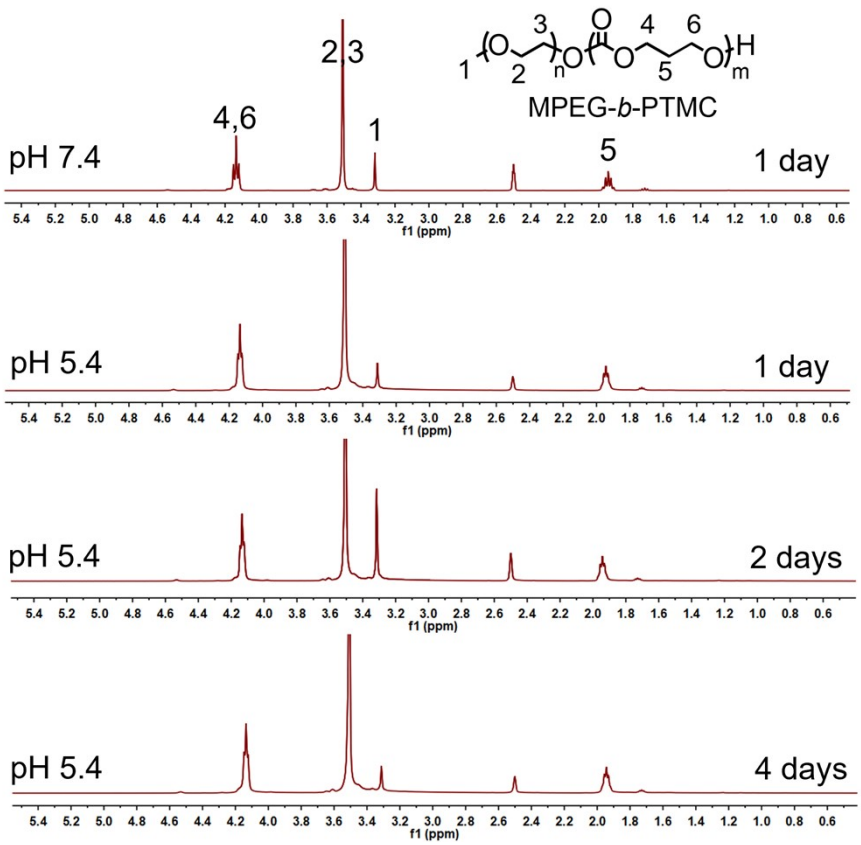


Figure S15. The stability of MPEG-PTMC was monitored by ^{1}H NMR spectroscopy at pH 5.4 with different times (500 MHz, $(CD_3)_2SO$).

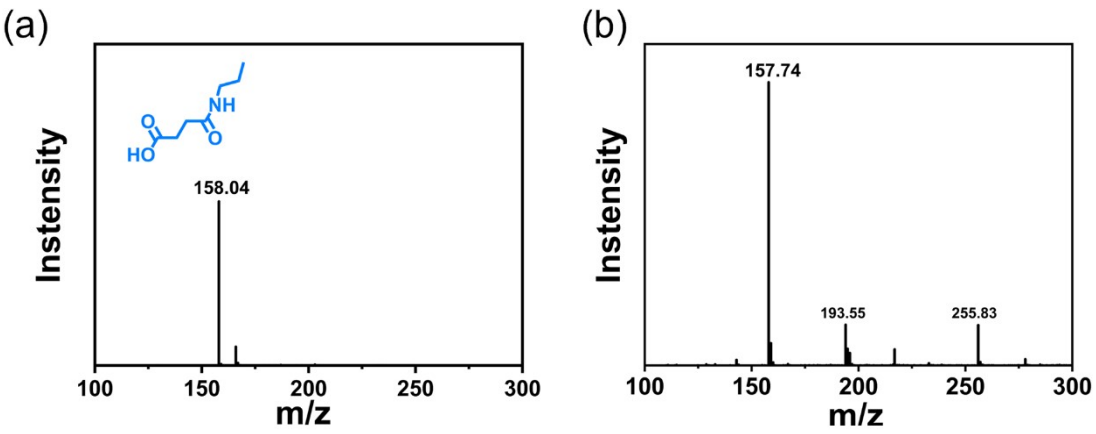


Figure S16. The MS of the SA-linked propylamine after 12 h incubation at a) pH 7.4 and b) pH 5.4 conditions.

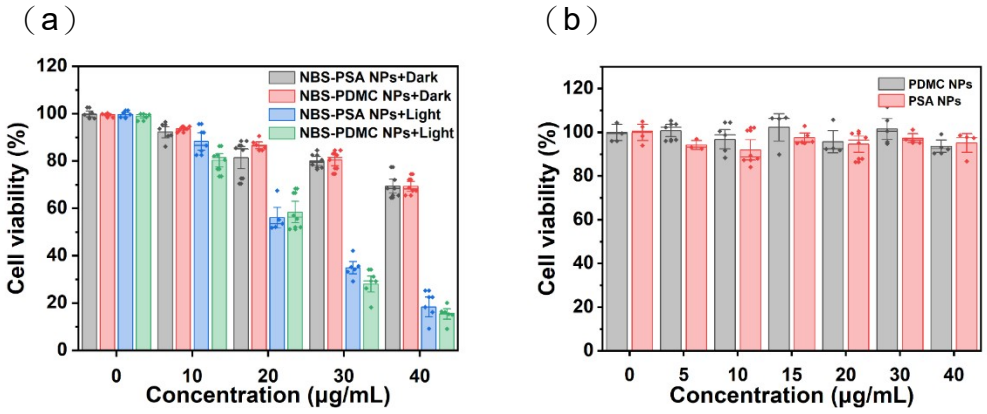


Figure S17. a) The cytotoxicity of NBS-PSA NPs and NBS-PDMC NPs against HepG2 cells was determined with light irradiation (660 nm, 10 mW cm^{-2}, 4 min) and dark. b) The cytotoxicity of PDMC NPs and PSA NPs against HepG2 cells.

Table S1. The half-maximal inhibitory concentration (IC_{50}) was treated with NBS-PSA NPs and NBS-PDMC NPs under light irradiation (660 nm, 10 mW cm^{-2}, 4 min). The concentration unit for fractions was μg/mL.

Cell	4T1	MCF-7	Hepg2
NBS-PSA NPs	24.42	24.39	23.15
NBS-PDMC NPs	19.08	18.97	21.37

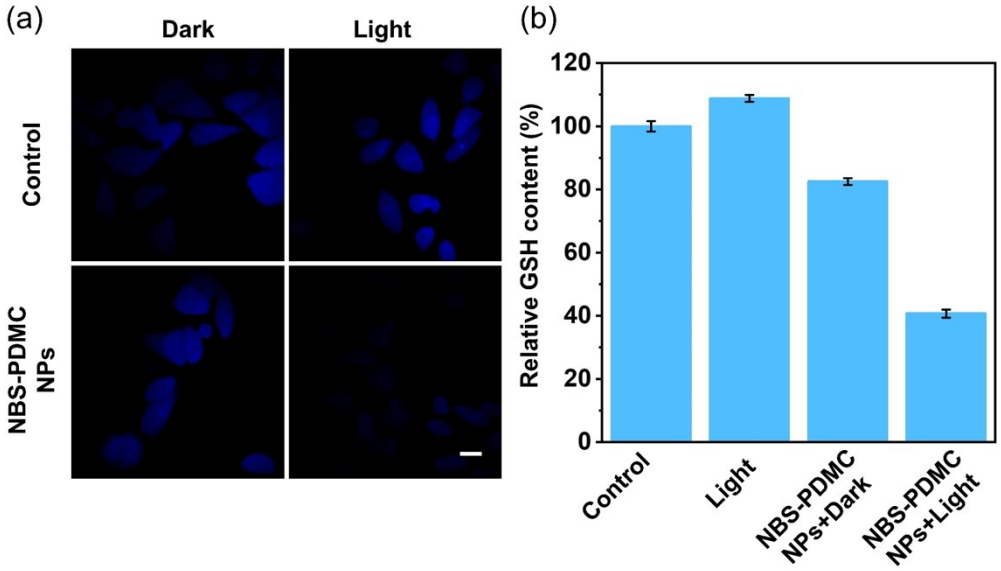


Figure S18. a) Fluorescence image of GSH level in MCF-7 cells with different treatments. Scale bars: 20 μm. b) The relative GSH content in MCF-7 with different treatments.

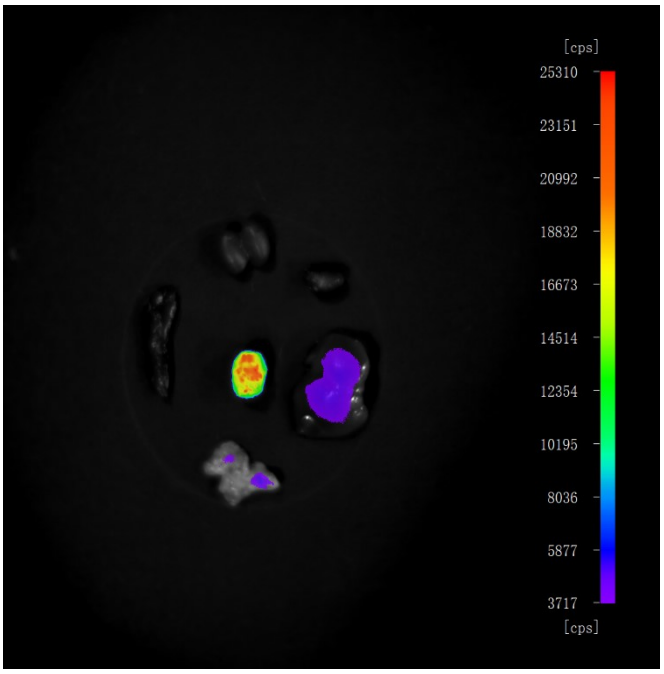


Figure S19. The fluorescence image of major organs and tumors excised from mice injected with NBS-PDMC NPs.

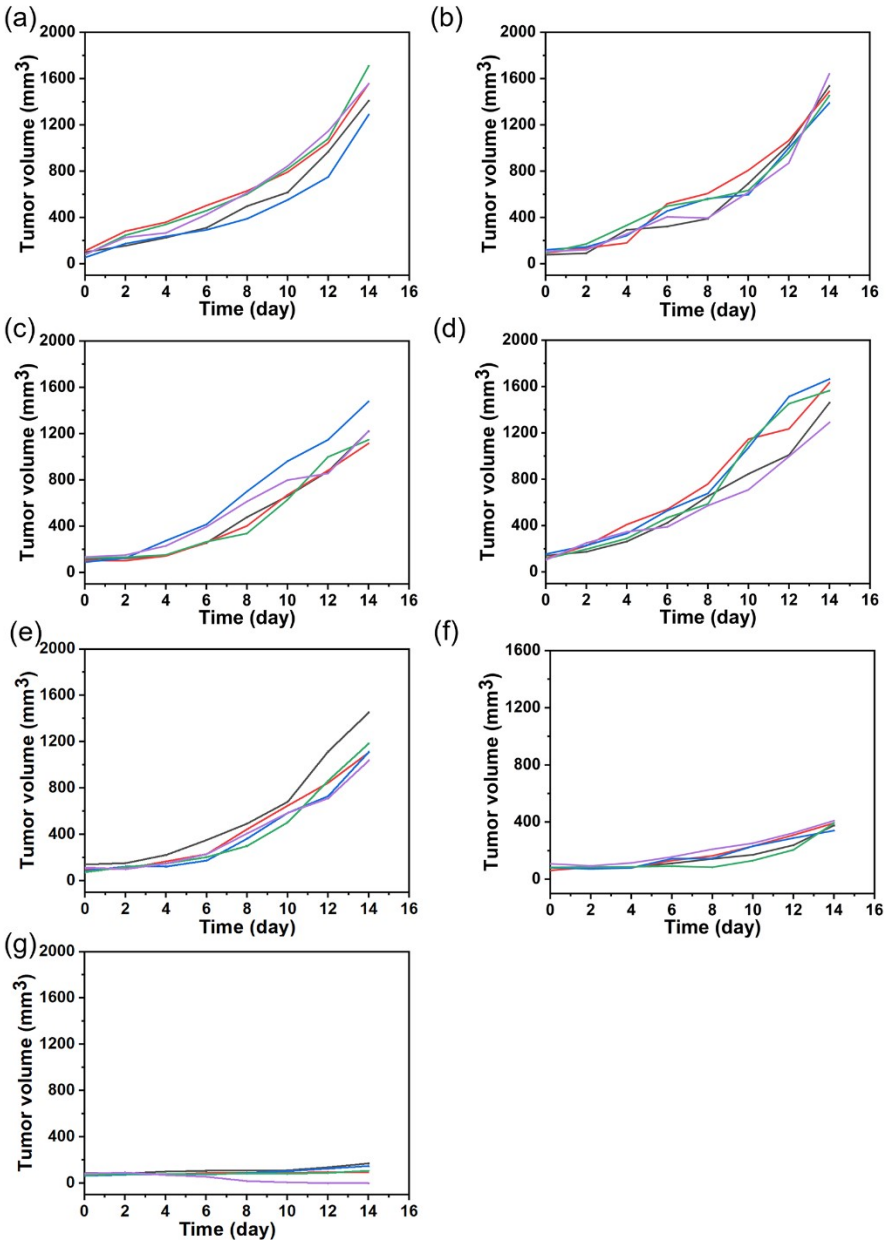


Figure S20. The tumor volume changes of the a) Control group; b) Light group; c) PDMC NPs+Light group; d) NBS-PSA NPs group; e) NBS-PDMC NPs group; f) NBS-PSA NPs+Light group; g) NBS-PDMC NPs+Light group.

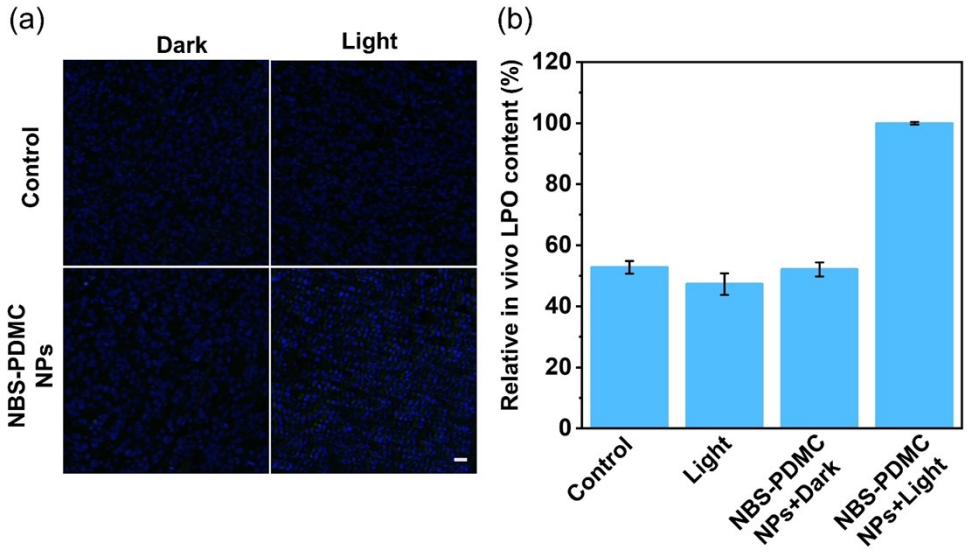


Figure S21. a) Fluorescence image of LPO level with different treatments *in vivo*. Scale bars: 20 μm. b) The relative LPO content *in vivo*.

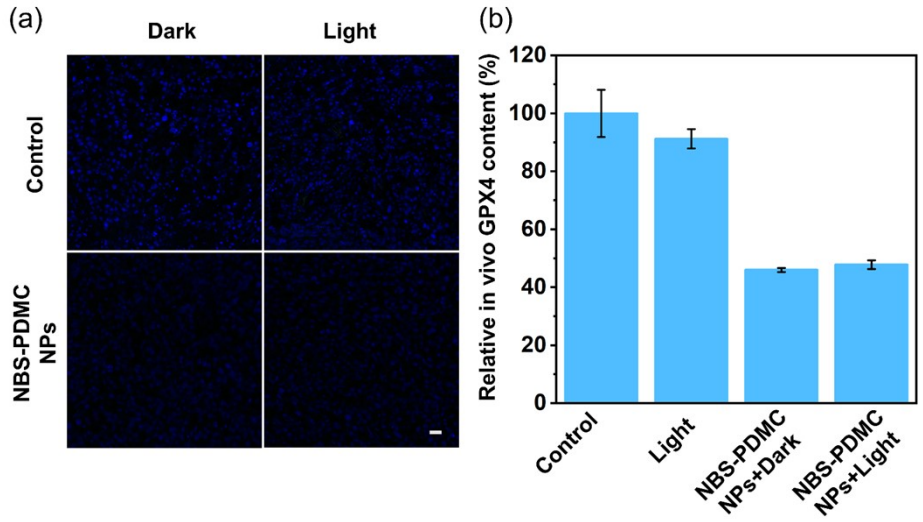


Figure S22. a) Fluorescence image of GPX4 level with different treatments *in vivo*. Scale bars: 20 μm. b) The relative GPX4 content *in vivo*.

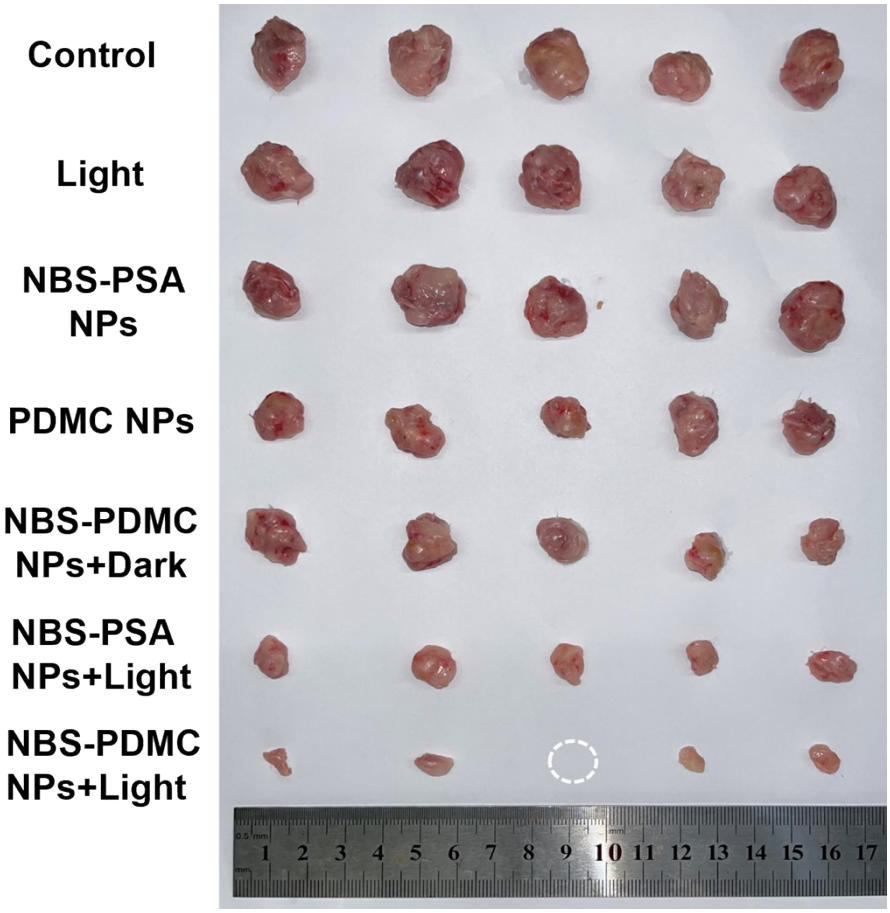


Figure S23. Photographic of excised tumors in mice subjected to different treatments.

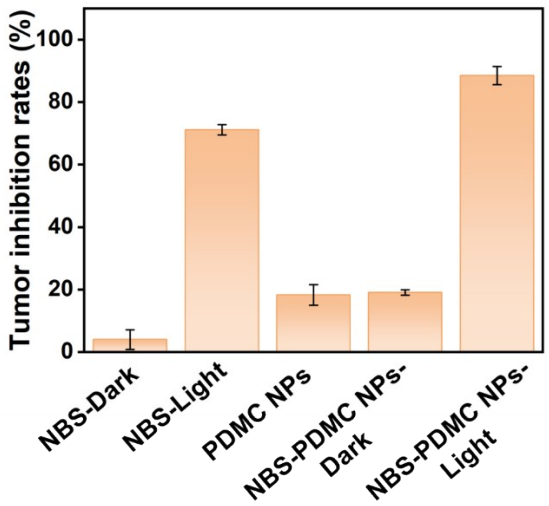


Figure S24. Tumor inhibition rates in different treatment groups.

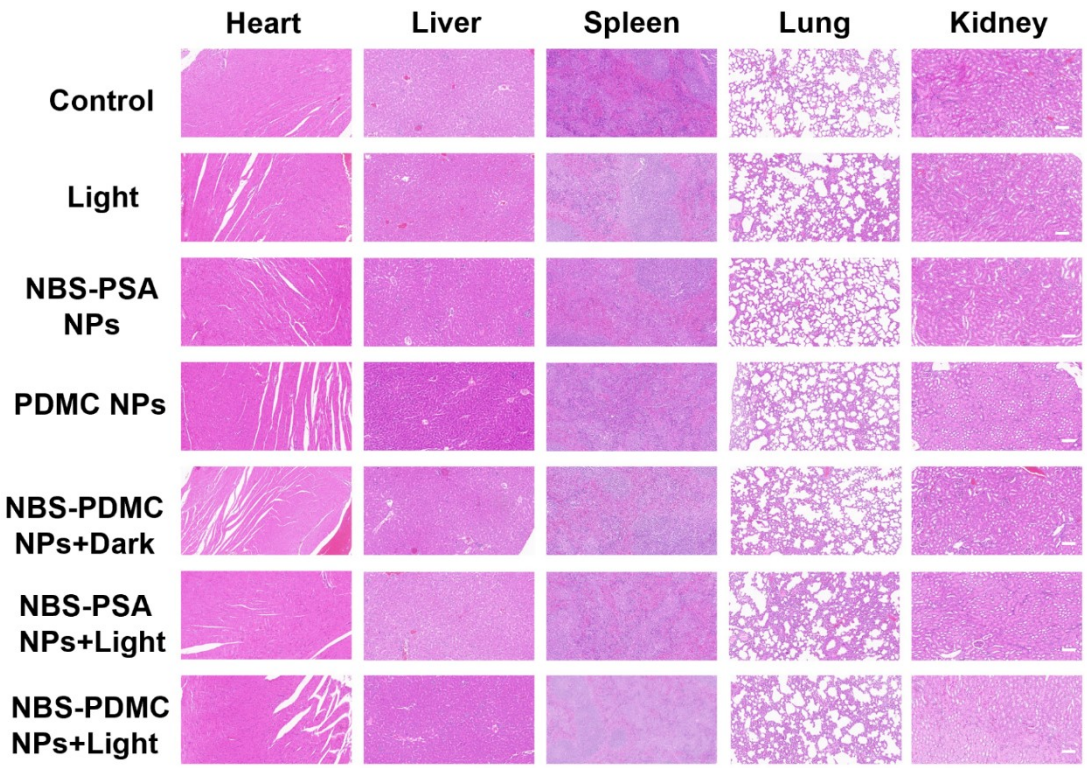


Figure S25. Images of H&E staining of major organs in each group with different treatments. tissues collected from mice in the different groups. Scale bars: 100 μm.

	Control	Light	PDMC NPs	NBS-PSA NPs	Reference range	Units
RBC	9.03	7.77	8.71	8.74	6.36-9.42	10^12/L
PLT	408	410	482	517	450-1590	10^9/L
MCHC	309	333	312	316	302-353	g/L
RDW_CV	12.2	11.3	11.4	12.0	11-17	%

	NBS-PSA NPs+Light	NBS-PDMC NPs	NBS-PDMC NPs+Light	Reference range	Units
RBC	8.79	8.86	8.47	6.36-9.42	10^12/L
PLT	433	557	528	450-1590	10^9/L
MCHC	320	333	322	302-353	g/L
RDW_CV	11.7	14.9	12.7	11-17	%

Figure S26. Routine blood analysis in mice subjected to different treatments: RBC; PLT; MCHC; and RDW_CV.

1. M. He, G. He, P. Wang, S. Jiang, Z. Jiao, D. Xi, P. Miao, X. Leng, Z. Wei, Y. Li, Y. Yang, R. Wang, J. Du, J. Fan, W. Sun and X. Peng, *Adv. Sci.*, 2021, **8**, 2103334.