Supporting Information

Two-photon Fluorophore Labeled Multi-functional Drug Carrier for Targeting Cancer Therapy, Inflammation Restraint and AIE Active Bioimaging

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1. Materials and Methods

1-Bromo-4-iodobenzene, tetrakis (triphenylphosphine) palladium (0) (Pd $(PPh_3)_4$), 4-(triphenylamino) phenylboronic acid, 2-Iodoethanol, 4-pyridylboronic acid, and lithium bis (trifluoromethanesulphonyl) imide were purchased from Adamas Reagent, Ltd (Shanghai, China). Bis(2-hydroxyethyl) disulfide, 4-dimethylaminopyridine (DMAP), dicyclohexylcarbodiimide (DCC) and ibuprofen were obtained from Chengdu Best Reagent Co., Ltd (Chengdu, China).

2.1 Preparation of Ibup-SS-MA.

Under Ar atmosphere, bis (2-hydroxyethyl) disulfide (4.00 g, 25.97 mmol) and trimethylamine (TEA, 36.07 mmol) were dissolved in THF (150 mL) in a dry flask. After cooling down in an ice bath, methacryloyl chloride (2.71 g, 25.93 mmol) was dissolved in THF (10 mL) and dropwise added into the solution under strong stirring. The reaction mixture was allowed to perform at room temperature for 24 h. The solution was filtered to remove the salt, concentrated and purified by a silica gel (EA/PE, 1/5, v/v) to obtain HO-SS-MA. Then, under Ar atmosphere, HO-SS-MA (0.90 g, 4.70 mmol), ibuprofen (0.98 g, 4.75 mmol), DCC (1.95 g, 9.47 mmol), DMAP (5.74 mg, 0.047 mmol) were dissolved in dry methylene dichloride in a dry

flask. The reaction was stirred at room temperature for 24 h. The product was purified by a silica gel (EA/PE, 1/3).

2.2 Synthesis of TPF-OH (NTf_2).

TPF-OH was firstly prepared via a series of reactions as follows: 4-(triphenylamino) phenylboronic acid (2.1 g, 7.3 mmol), 4-(4-Bromophenyl) pyridine (1.9 g, 8.1 mmol), Pd $(PPh_3)_4$ (426 mg, 0.37 mmol) and sodium carbonate (2.35 g, 22.2 mmol) were added to a 250 mL three-necked round-bottomed flask under argon (Ar) protection. A mixed solution of 100 mL Toluene, 30 mL EtOH and 10 mL H_2O were degassed and added into the flask under the protection of Ar. The mixture was then stirred at room temperature for 10 min and then conducted at 110 ^oC for 24 h. After the solution was filtered and concentrated under vacuum, the obtained solid was dissolved in DCM and filtered. After that, the solution was concentrated by rotary evaporation. The pure product TPF was obtained by a silica gel with EA and PE (1/3, V/V).

Under Ar atmosphere, TPF (2.3 g, 5.8 mmol) was dissolved in 100 mL chloroform, 2-iodoethanol (11.2 g, 65 mmol) was added into the solution under stirring and the reaction was carried out at room temperature for 72 h. The solution was washed with brine and dried with anhydrous Na_2SO_4, the resulted solution was concentrated and redissolved in a mixed solution of 30 mL DCM and 20 mL H_2O. Then bis (trifluoromethanesulphonyl) imide (2.1 g, 7.3 mmol) was added and the mixed solution was stirred at room temperature for 48 h. The solution was extracted with saline and the organic layer was dried with anhydrous Na_2SO_4 and concentrated by rotary evaporation. The pure product TPF-OH (NTf_2) was purified by a silica gel with DCM and methanol (10/1, V/V).

2.3 Synthesis of TPF-MA.

TPF-OH (1 g, 1.38 mmol) and triethylamine (0.49 mL, 2.82 mmol) were dissolved in 50 mL dry THF under Ar atmosphere. Methacryloyl chloride (0.2 mL, 2.073 mmol) dissolved in 10 mL dry THF was added dropwise into the mixed solution under ice bath. The reaction was stirred at room temperature for 24 h. The resulted solution was filtered and concentrated under vacuum. TPF-MA was obtained by recrystallization with DCM and PE.

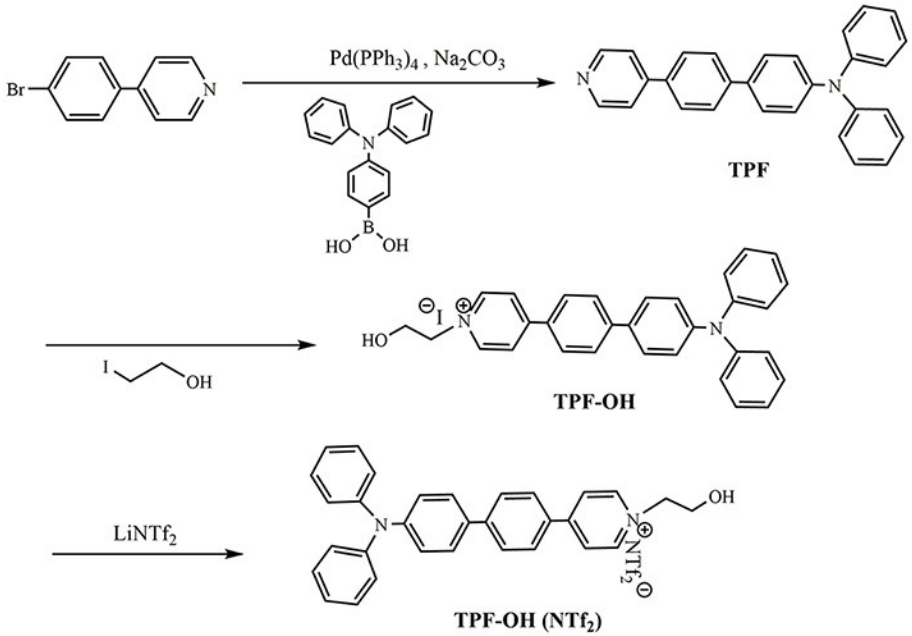

Figure S1. The synthetic route of TPF-OH (NTf_2).

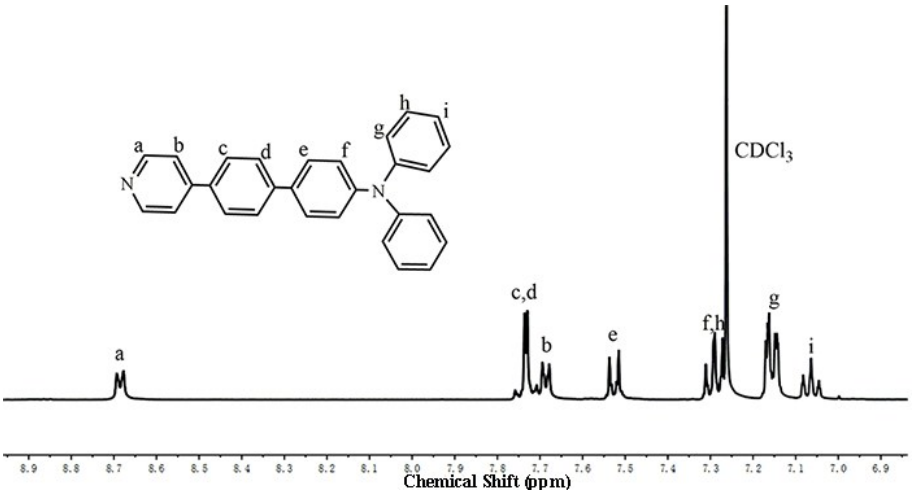

Figure S2. ^1H NMR spectrum of TPF in $CDCl_3$.

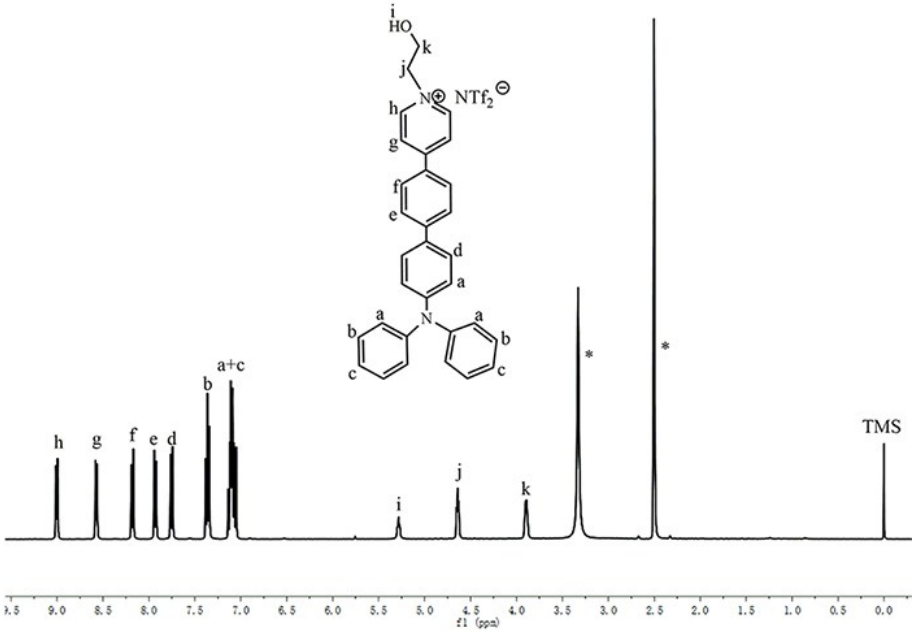

Figure S3. ^{1}H NMR spectrum of TPF-OH (NTf_2) in DMSO-*d*6.

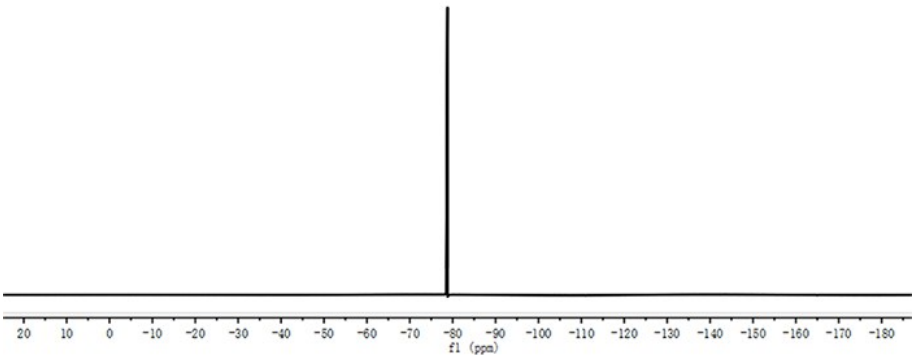

Figure S4. Fluorine spectrum of TPF-OH (NTf_2) in DMSO-*d*6.

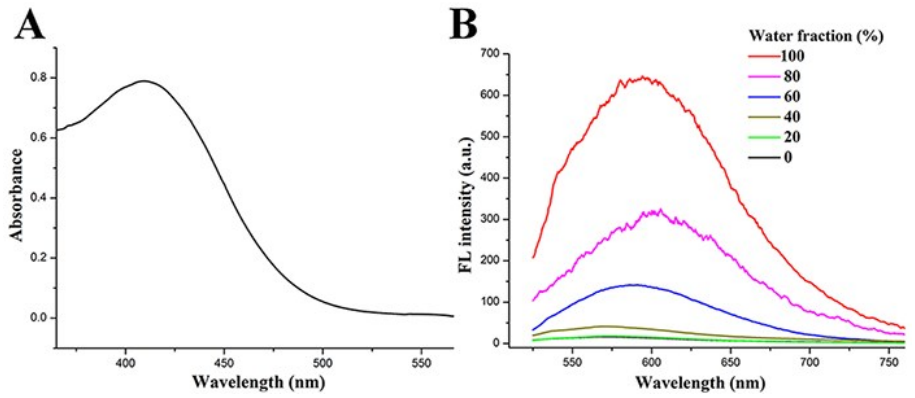

Figure S5. The UV absorption of TPF-OH (NTf_2) in THF (A) and FL

intensity of TPF-OH (NTf_2) in THF and water with different water fractions (V/V) (B).

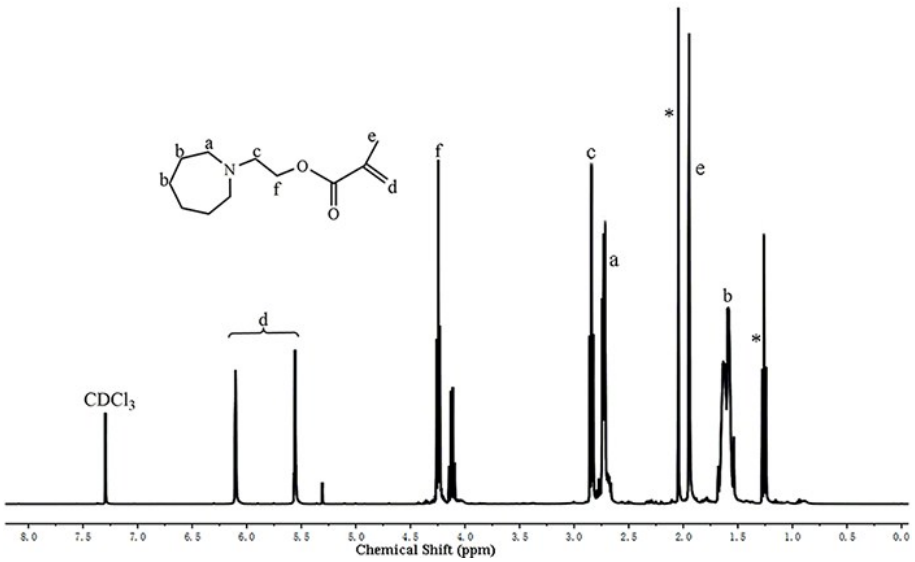

Figure S6. ^{1}H NMR spectrum of AEMA monomer in $CDCl_3$.

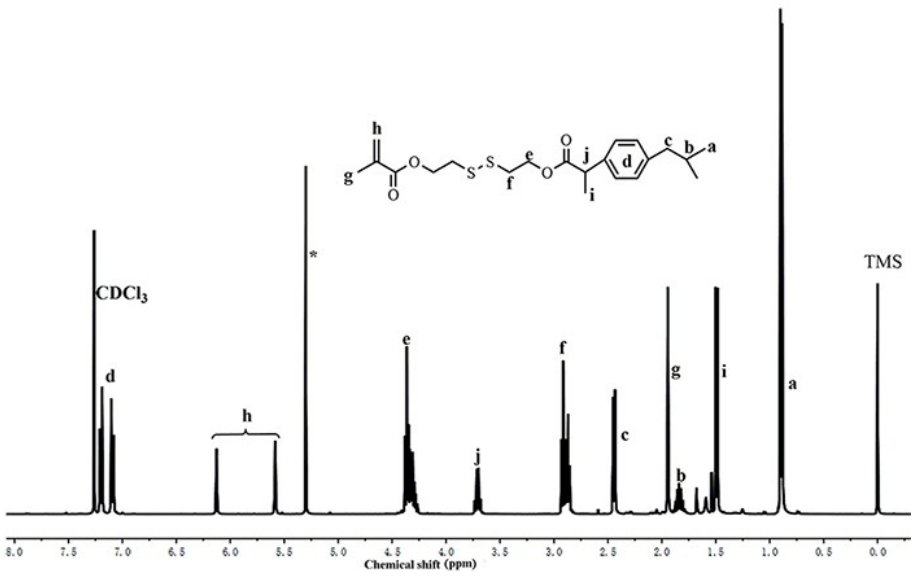

Figure S7. ^{1}H NMR spectrum of Ibup-SS-MA in $CDCl_3$.

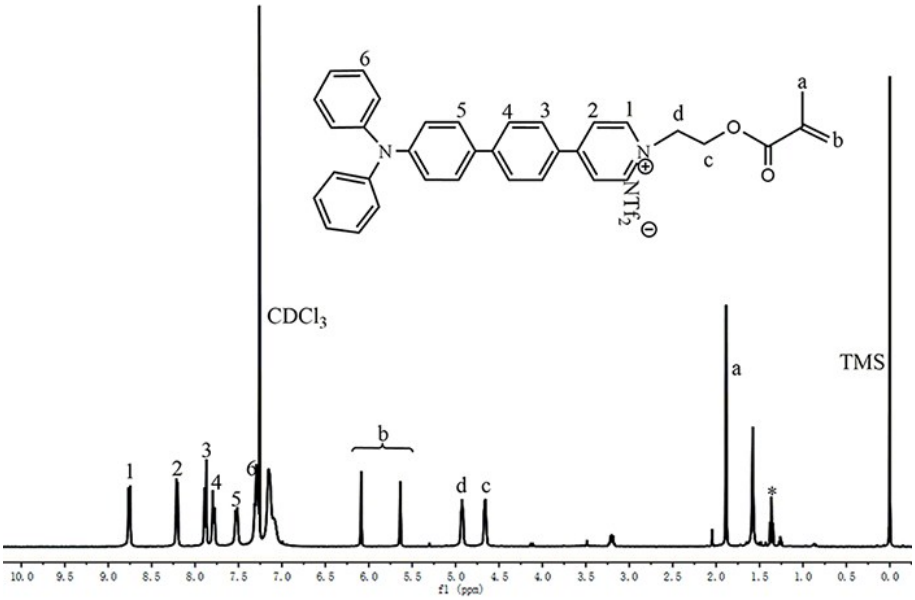

Figure S8. ^{1}H NMR spectrum of TPF-MA in $CDCl_3$.

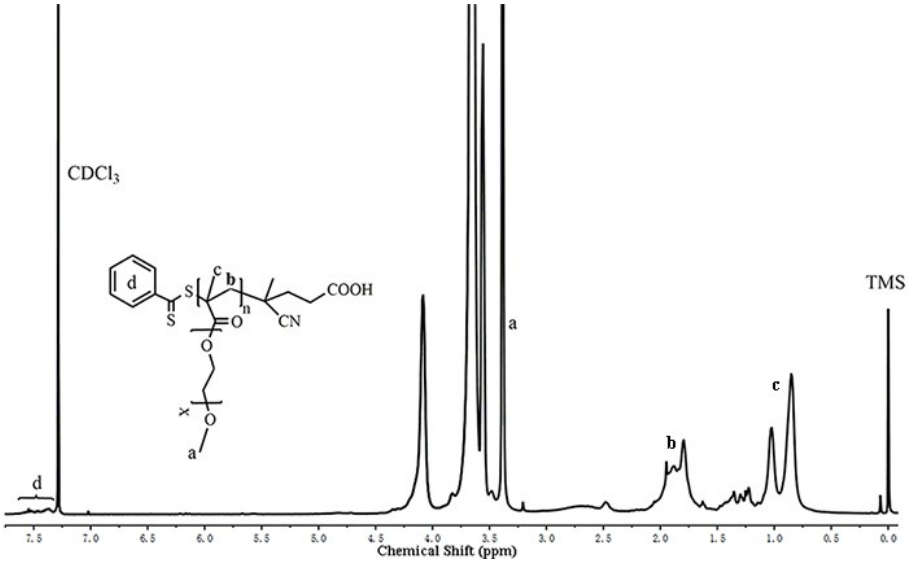

Figure S9. ^{1}H NMR spectrum of $PPEG_{300}$ in $CDCl_3$.

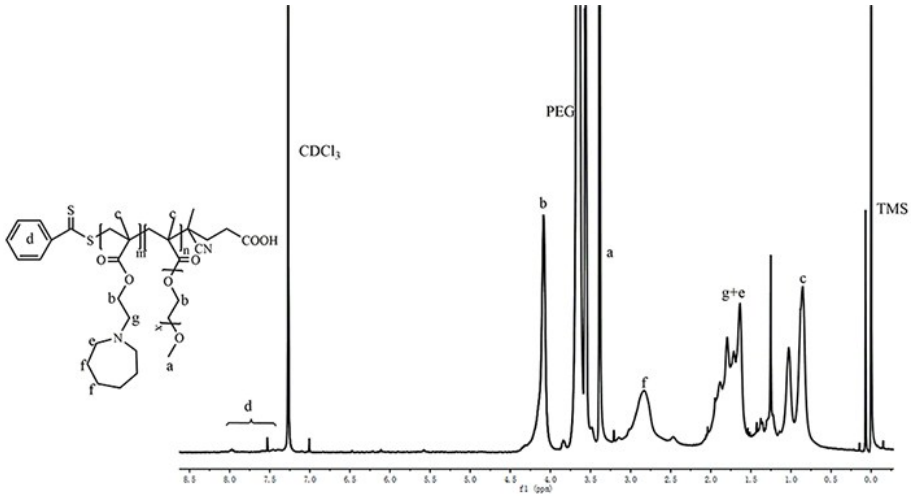

Figure S10. ^{1}H NMR spectrum of copolymer PAEMA-PPEG in $CDCl_3$.

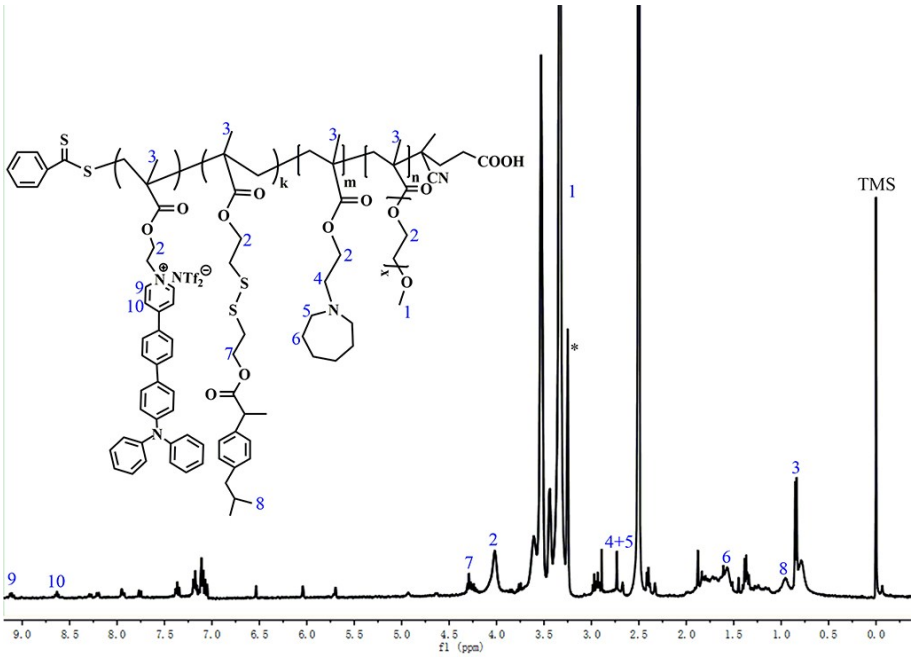

Figure S11. ^{1}H NMR spectrum of triblock copolymer P (TPF-*co*-Ibup)-PAEMA-PPEG in DMSO-d_6.

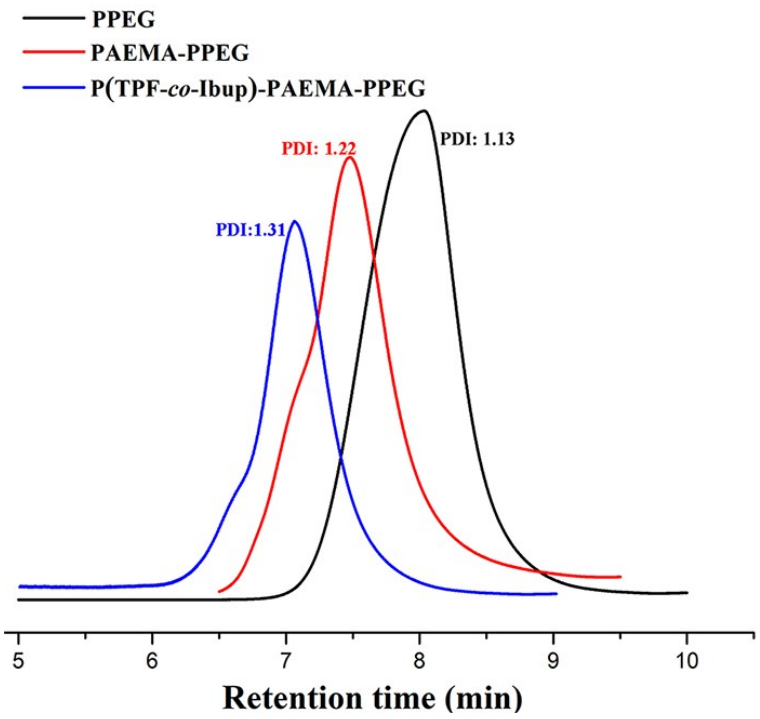

Figure S12. GPC trace of polymers with THF as eluent.

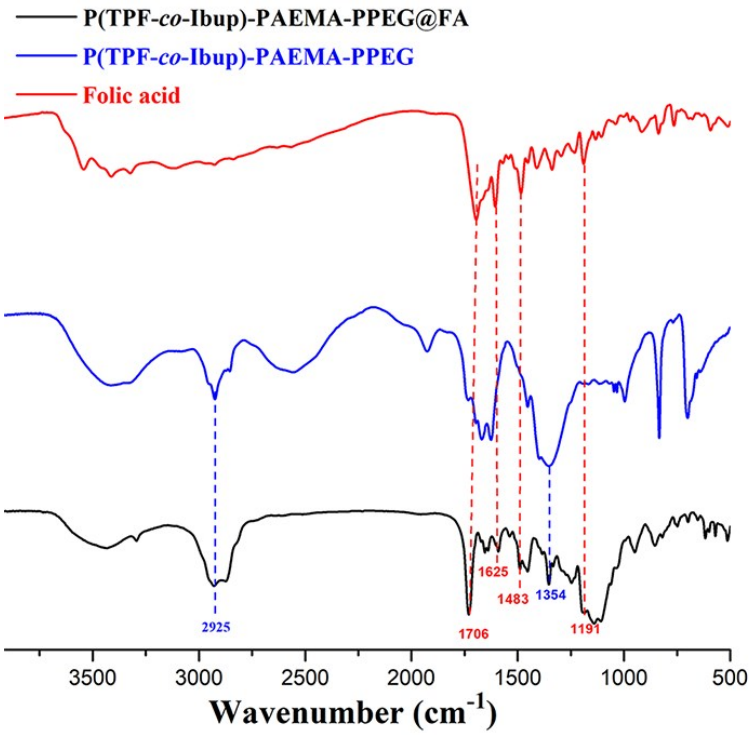

Figure S13. The infrared spectra of folic acid, P (TPF-*co*-Ibup)-PAEMA-PPEG, and P (TPF-*co*-Ibup)-PAEMA-PPEG@FA.

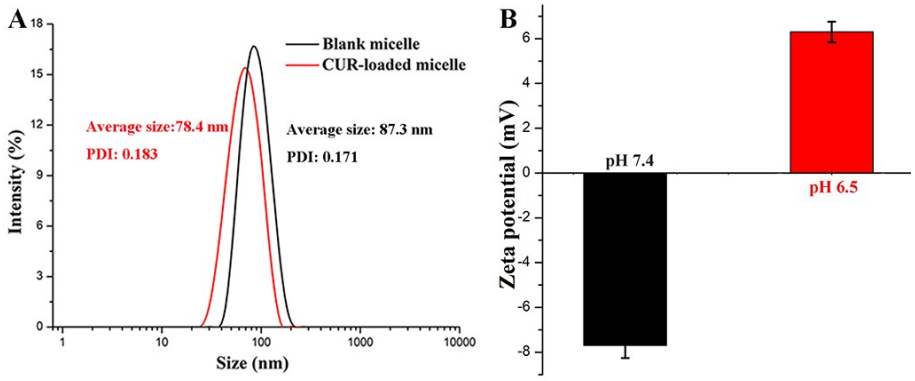

Figure S14. (A)The particle size of P(TPF-*co*-Ibup)-PAEMA-PPEG micelles and CUR-loaded micelles; (B)The zeta potential of NPs at different pH environments.

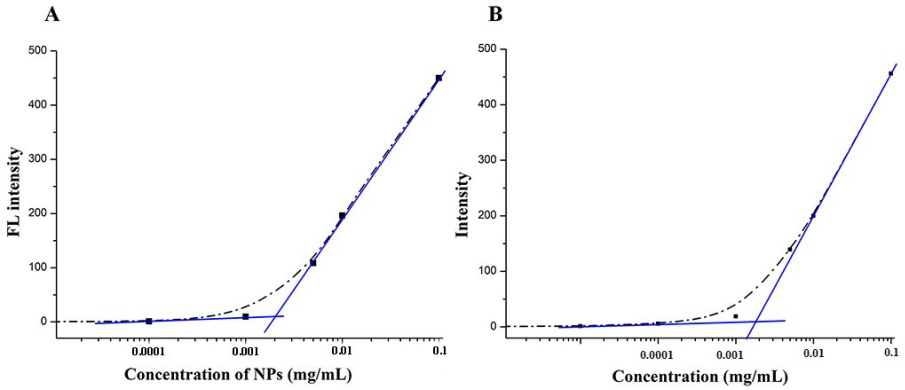

Figure S15. The CMC of NPs. (A) P (TPF-*co*-Ibup)-PAEMA-PPEG; (B) P (TPF-*co*-Ibup)-PAEMA-PPEG@FA.

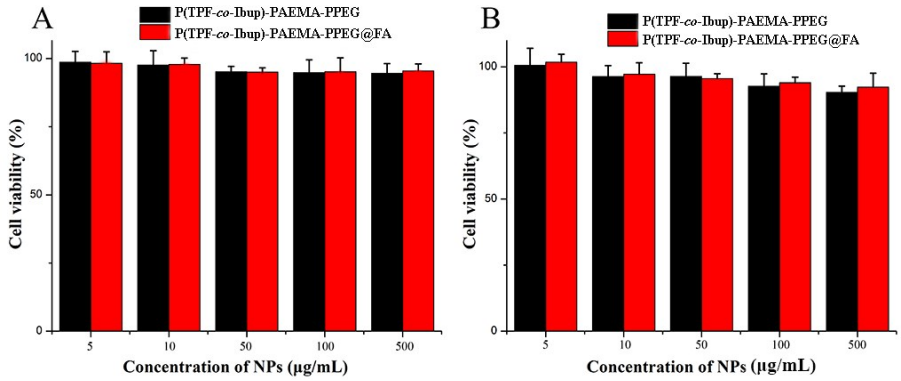

Figure S16. The cytotoxicity of the polymeric micelles against 4T1 cells for 24 h (A) and 48 h (B);

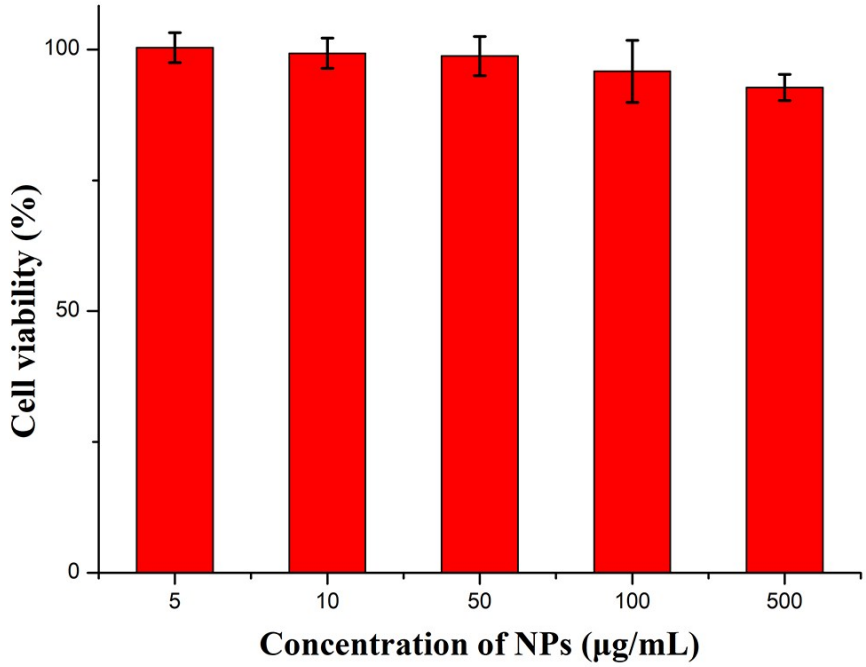

Figure S17. The cytotoxicity of the P (TPF-*co*-Ibup)-PAEMA-PPEG@FA micelles against 4T1 cells for 48 h at pH 6.5.

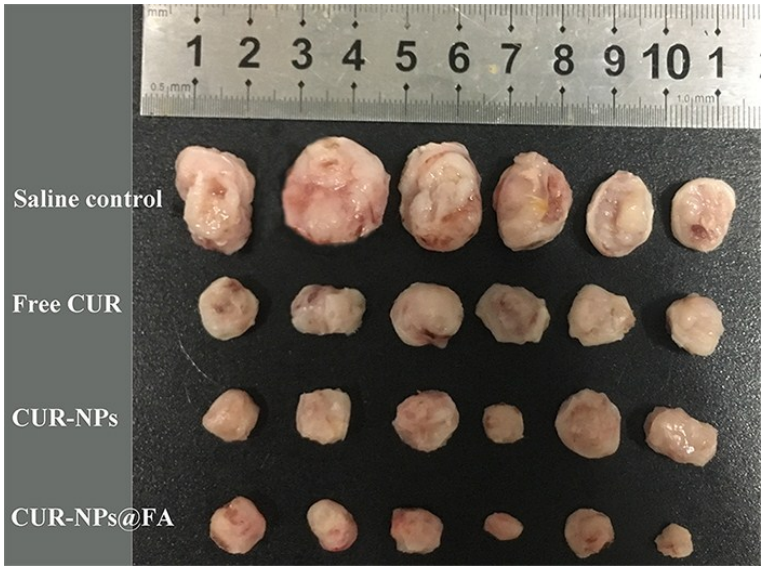

Figure S18. The photo graph of isolated tumor samples of different treatment groups.

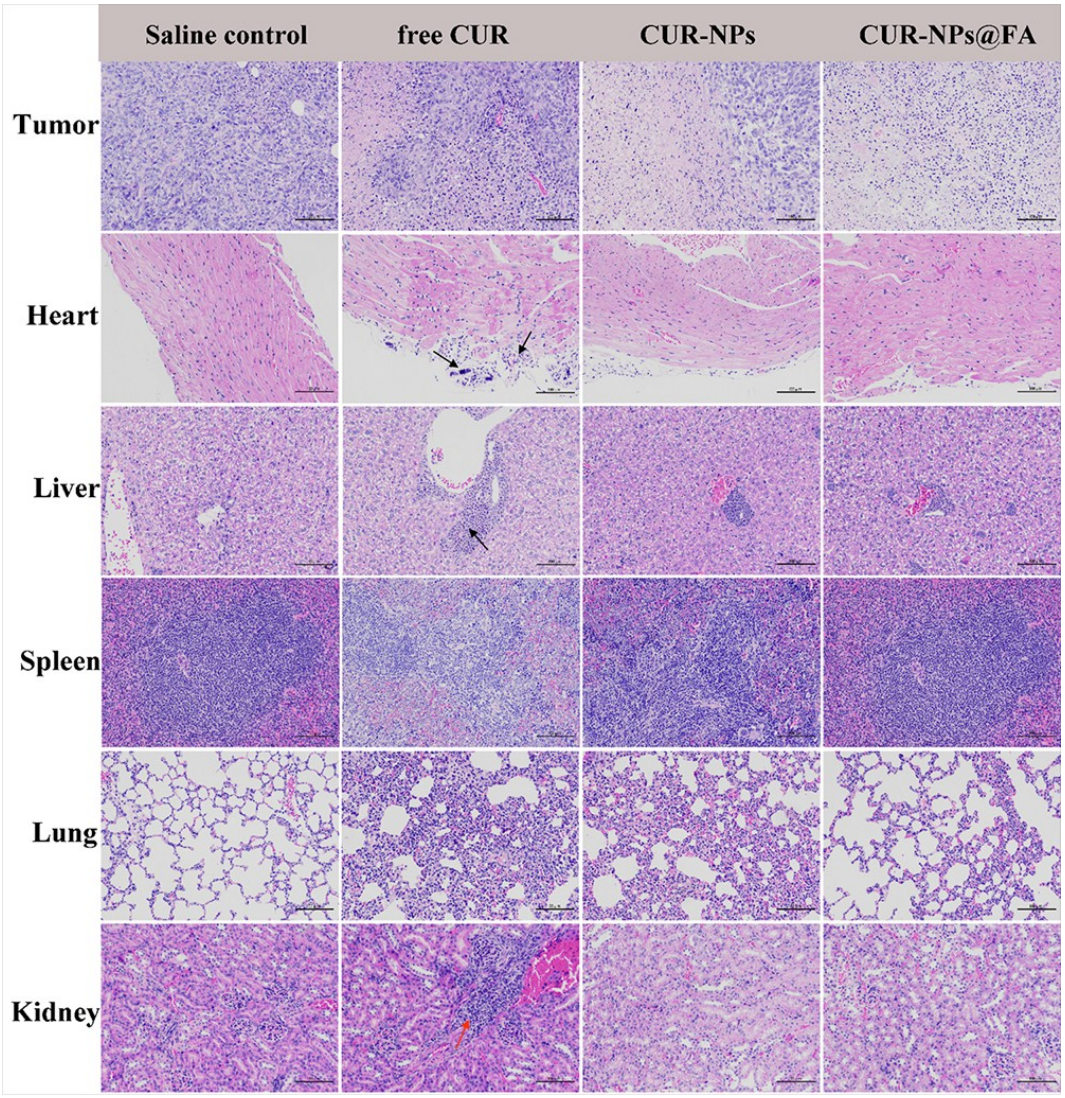

Figure S19. H&E staining assays for tumors and major organs with different treatments for 21 days (all tissues: 200×).