Supporting Information

Zwitterionic Block Copolymer Prodrug Micelles for pH Responsive Drug Delivery and Hypoxia-Specific Chemotherapy

Jianyang Zhao,[1,2] *Yi-Yang Peng,*[3] *Diana Diaz-Dussan,*[3] *Jacinta White,*[2] *Wei Duan,*[4] *Lingxue Kong,*[1*] *Ravin Narain,*[3*] *and Xiaojuan Hao*[2*]

[1] Institute for Frontier Materials, Deakin University, Geelong, Victoria 3216, Australia.

[2] Manufacturing, CSIRO, Research Way, Clayton, Victoria 3168, Australia.[3] Department of Chemical and Materials Engineering, University of Alberta, 116 Street and 85th Avenue, Edmonton T6G 2G6, Alberta, Canada.

[4] School of Medicine, Deakin University, Geelong, Vic 3216, Australia.

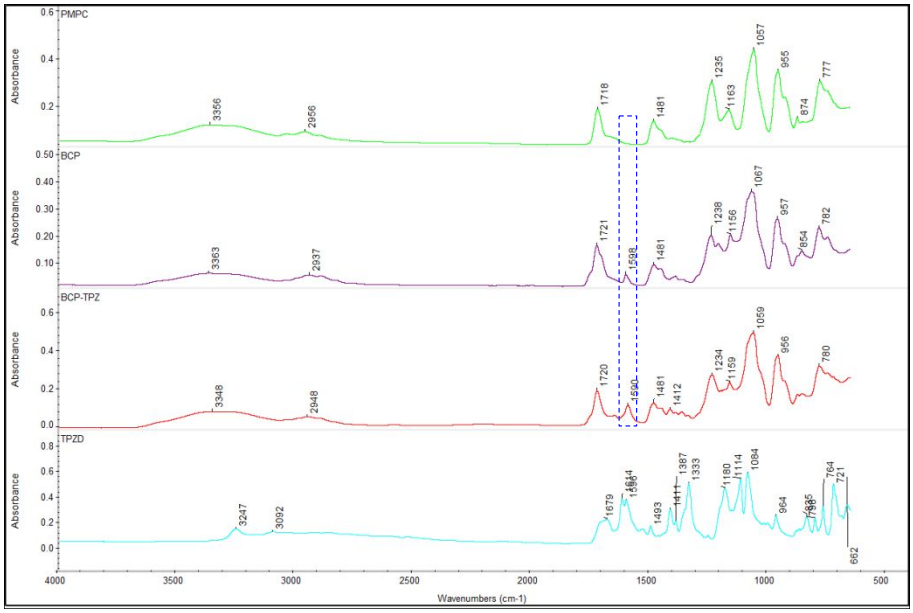

Figure S1. FT-IR spectra of PMPC, BCP, BCP-TPZ, and TPZD.

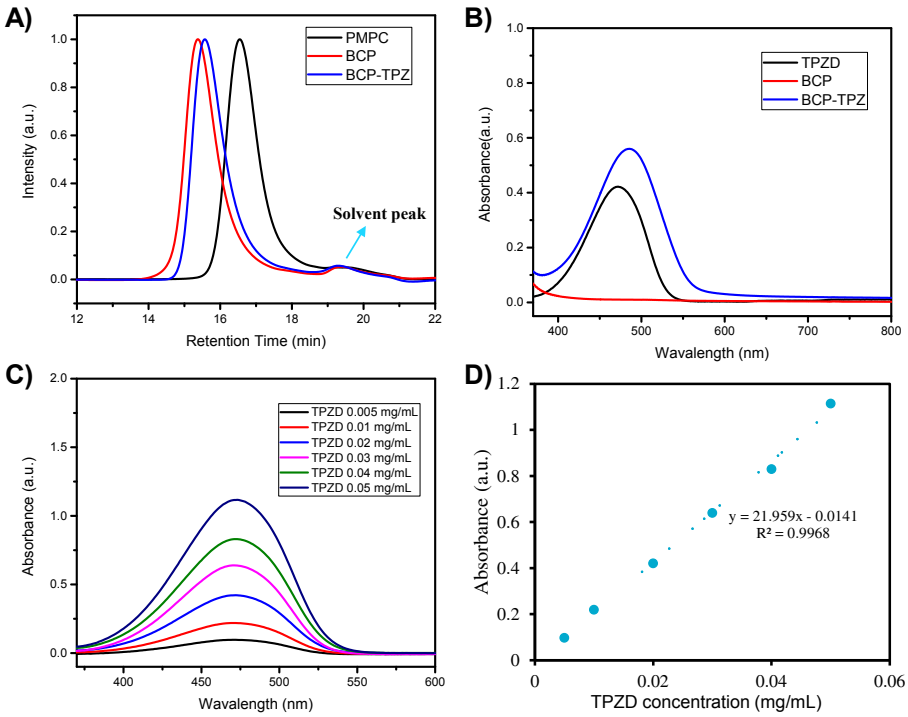

Figure S2. (A) GPC curves of PMPC, BCP, and BCP-TPZ. (B) UV-VIS spectra of PMPC, BCP, and BCP-TPZ. (C) UV-VIS spectra of TPZD in PBS with different concentrations. (D) Standard calibration curve and calculation formula of TPZD.

Table S1. Composition and characterization of PMPC, BCP, and BCP-TPZ.

Polymer	*Mn* (g/mol)			Size (nm)/*D*					
	^{1}H NMR[a]	GPC[b]	PDI	0.5 mg/mL[c]	0.5 mg/mL[d]	1 mg/mL[c]	1 mg/mL[d]	2 mg/mL[c]	2 mg/mL[d]
$Poly(MPC)_{47}$	13860	7500	1.50	-	-	-	-	-	-
$P(MPC)_{47}$-*b*-$(DEGMA_{36}$-*co*-$FPMA_{25})$, BCP	26260	12850	1.87	38/0.11	38/0.13	34/0.057	36/0.047	36/0.046	39/0.061
$P(MPC)_{47}$-*b*-$(DEGMA_{36}$-*co*-$[(FPMA$-*g*-$TPZD)_{25}]$, BCP-TPZ	31790	18260	1.83	51/0.17	54/0.17	59/0.17	57/0.18	58/0.19	59/0.18

[a] Calculated from ^{1}H NMR signal integration. [b] Obtained from aqueous GPC using 0.2 M $NaNO_3$, 0.01M Na_2PO_4 in Milli-Q water with 200 ppm Na_3N (pH = 8) as an eluent. [c] DLS tested at 25 °C. [d] DLS tested at 37 °C.

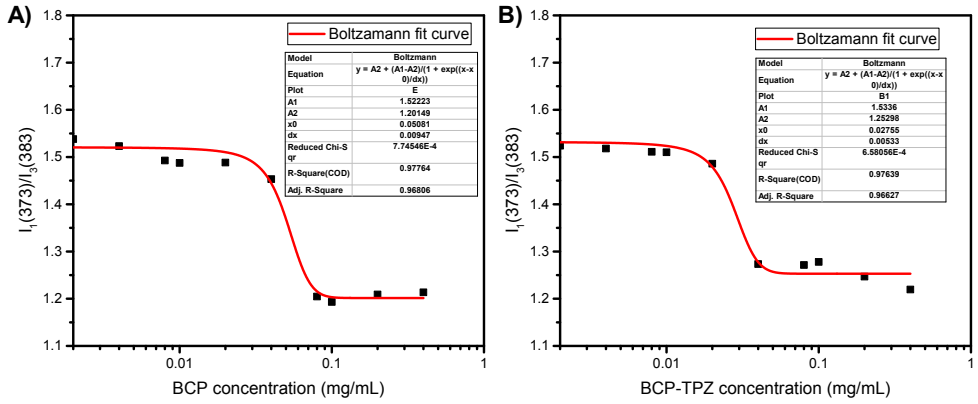

Figure S3. Boltz fitting of pyrene 1:3 ratio (I_1/I_3) versus concentration of copolymers in aqueous solution: (A) BCP and (B) BCP-TPZ.

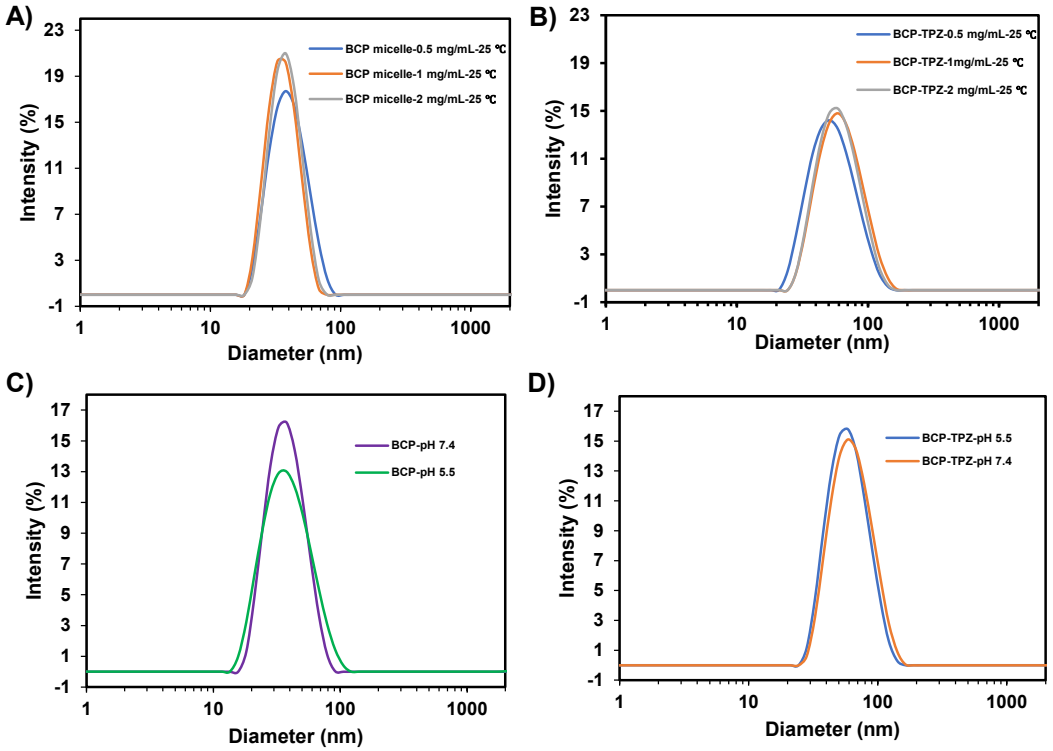

Figure S4. (A) Hydrodynamic size distribution of (A) BCP micelles and (B) BCP-TPZ micelles at different concentrations at 25 ℃. (C) Hydrodynamic size distribution comparison of BCP micelles

under pH 7.4 and 5.5 at 37 ℃. (D) Hydrodynamic size distribution comparison of BCP-TPZ micelles under pH 7.4 and 5.5 at 37 ℃.

Table S2. Zeta potentials of BCP and BCP-TPZ micelles under pH 7.4 and 5.5.

Micelles[a]	Zeta Potentials (mV)	
	pH 7.4	pH 5.5
BCP	-23.67 ± 2.00	-23.40 ± 2.43
BCP-TPZ	-25.87 ± 1.85	-27.50 ± 0.20

[a]BCP and BCP-TPZ micells prepared in deionized water at 0.5 mg/mL.

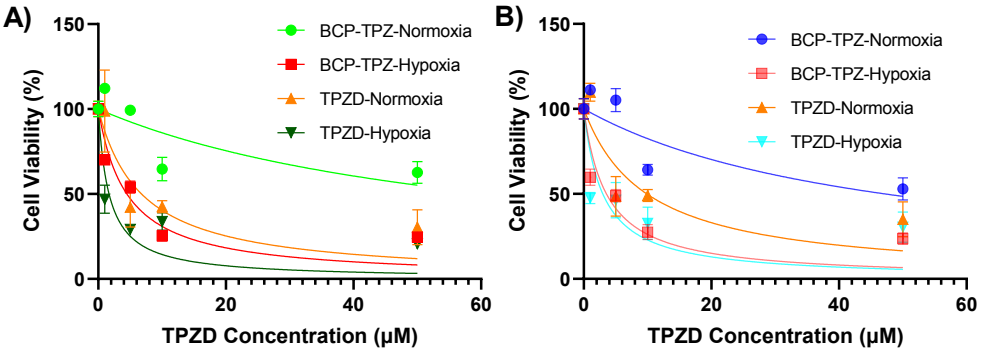

Figure S5. Dose-response curves of different formulations against (A) HeLa cells under normoxic and hypoxic conditions; (B) HepG2 cells under normoxic and hypoxic conditions. Data shown are means ± S.D., n=5.

Table S3. Cytotoxicity of BCP-TPZ and TPZD against HeLa and HepG2 cells under normoxic/hypoxic conditions.

Compound	Cytotoxicity (IC50, μM)					
	HeLa-Normoxia	HeLa-Hypoxia	HCR[a]	HepG2-Normoxia	HepG2-Hypoxia	HCR[a]
BCP-TPZ	61.6	4.5	**13.7**	47.2	3.5	**13.5**
TPZD	6.8	1.7	**4.0**	9.9	3.0	**3.3**

[a]Hypoxia cytotoxicity ratio, HCR = IC_{50} (normoxia)/IC_{50} (hypoxia).

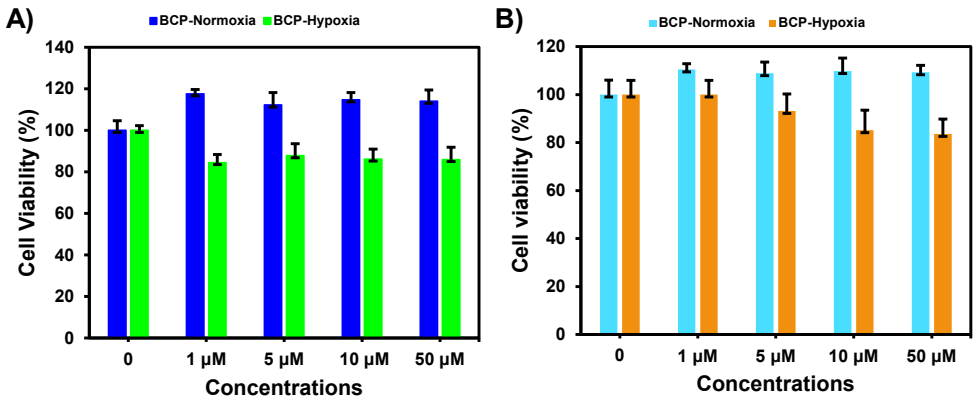

Figure S6. Cytotoxicity evaluation of BCP copolymer under normoxic and hypoxic condition against (A) HeLa cells and (B) HepG2 cells. Data shown are means ± S.D., n = 5.