

**Poly(HPMA)-chlorambucil conjugate nanoparticle: facilely  
fabrication, acidic responsive release and in vitro anti-cancer activity**

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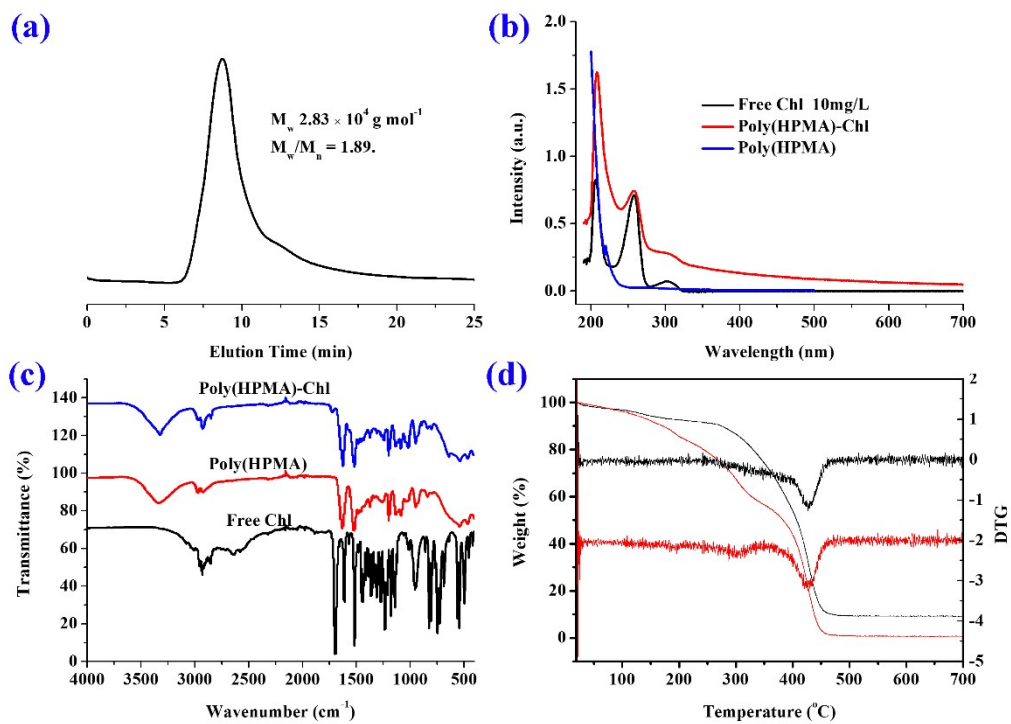


Fig. S1 (a) SEC curve of the Poly(HPMA)-Chl conjugate, UV-Vis (b), IR spectrum (c) and TGA curves of free Chl, Poly(HPMA) and Poly(HPMA)-Chl conjugate.

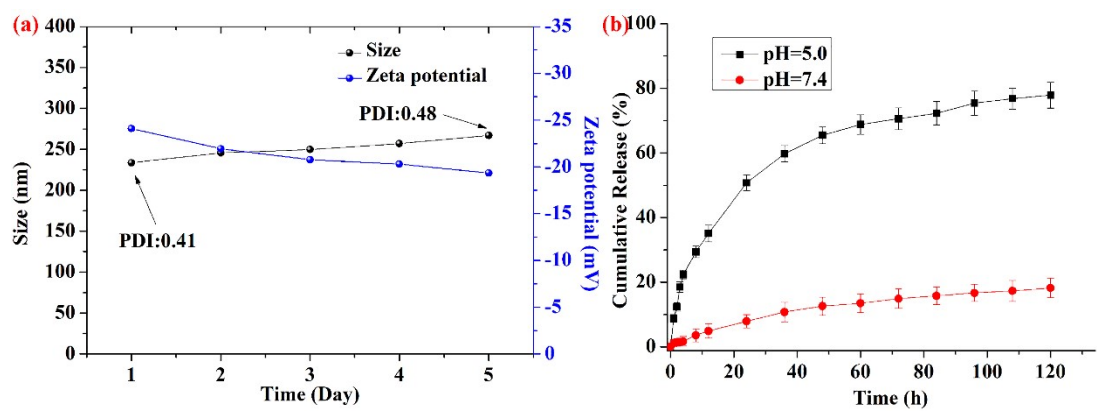


Fig. S2 (a)DLS and Zeta potential of the Poly(HPMA)-Chl conjugate for a period of five days, (b) In vitro Chl release of poly(HPMA)-Chl conjugate in PBS with different pH.

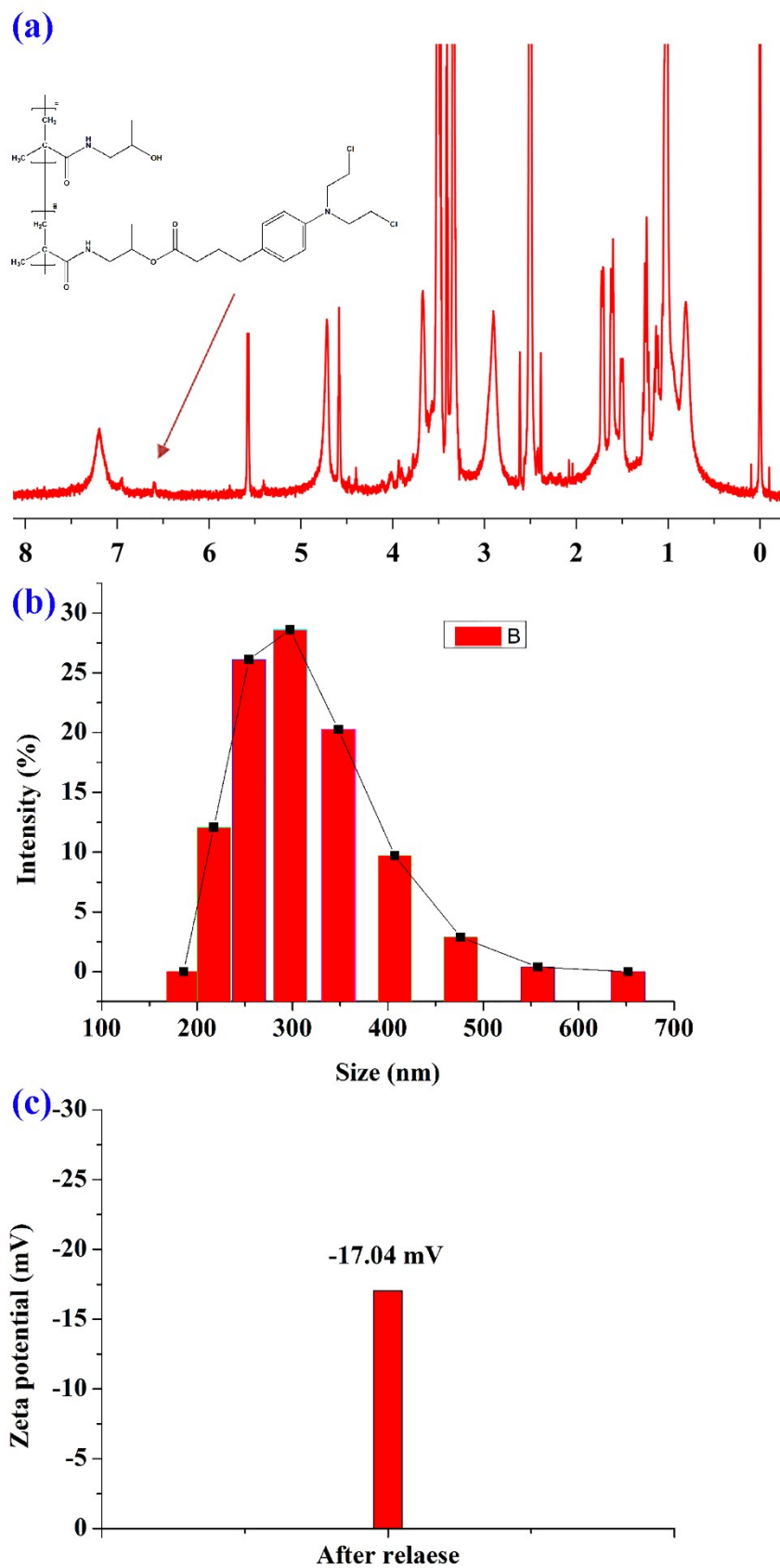


Fig. S3 (a) the  $^1\text{H}$  NMR spectrum, (b) DLS and (c) Zeta potential of the residue product after release of Chl.

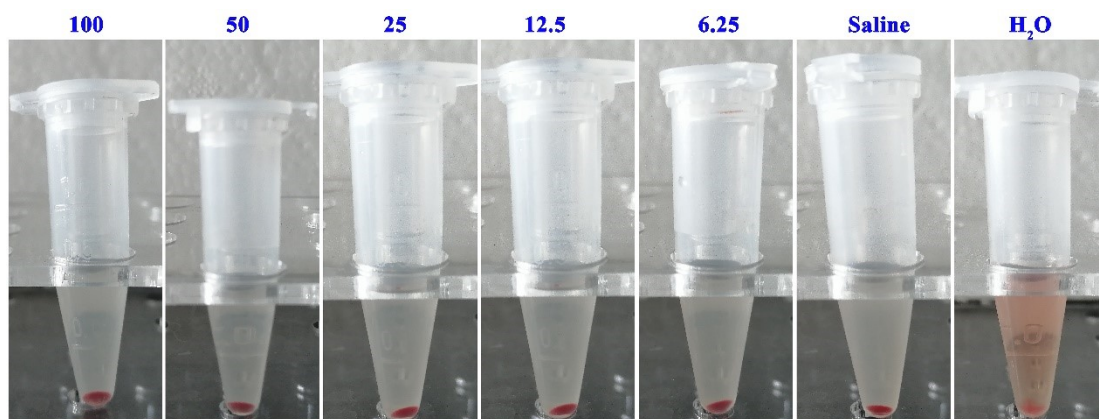


Fig. S4 Erythrocyte hemolysis resulted of Poly(HPMA)-Chl conjugate.

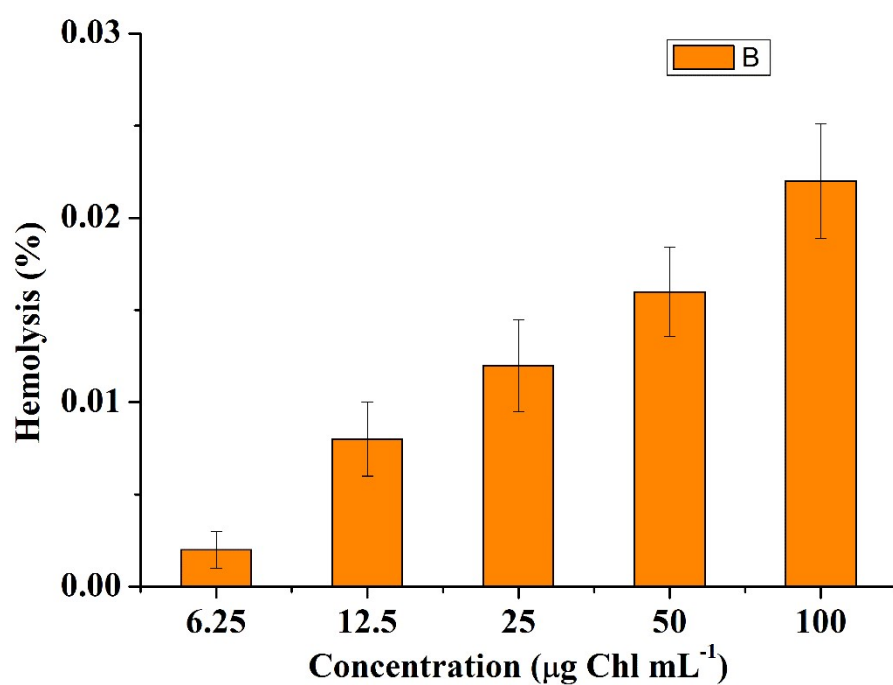


Fig. S5 Hemolysis of the Poly(HPMA)-Chl towards RBCs

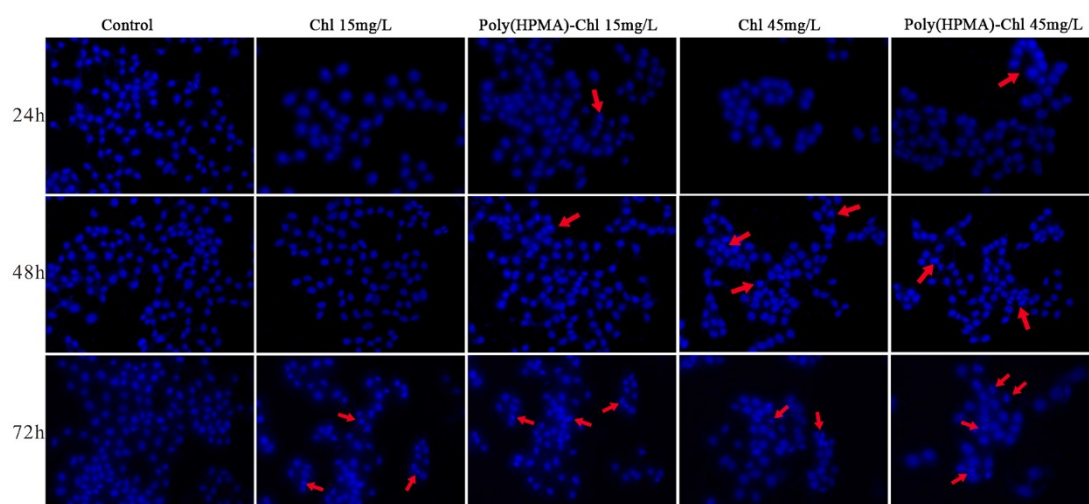


Fig. S6 Fluorescence images of HepG-2 cells incubated with different concentration free Chl and Poly(HPMA)-Chl for 24, 48 and 72 h.

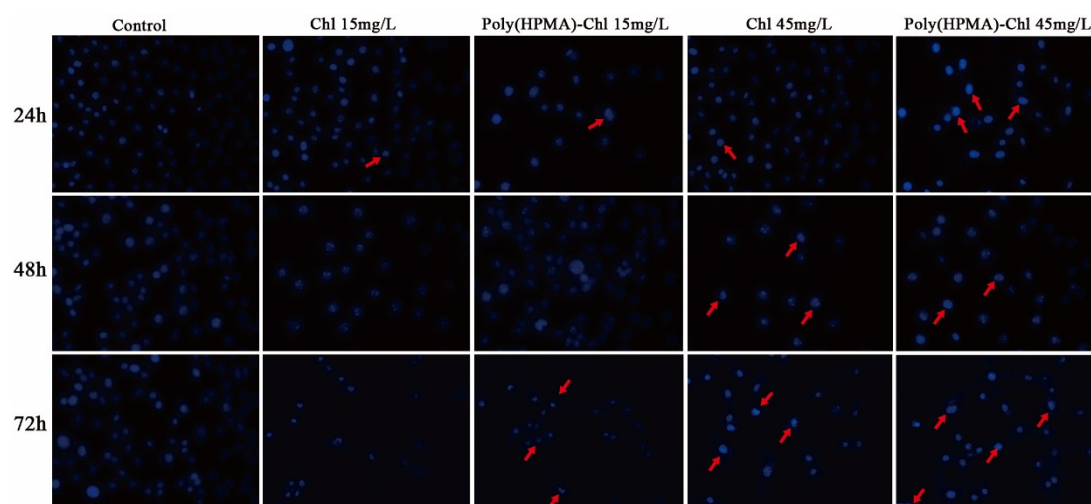


Fig. S7 Fluorescence images of MCF-7 cells incubated with different concentration free Chl and Poly(HPMA)-Chl for 24, 48 and 72 h.

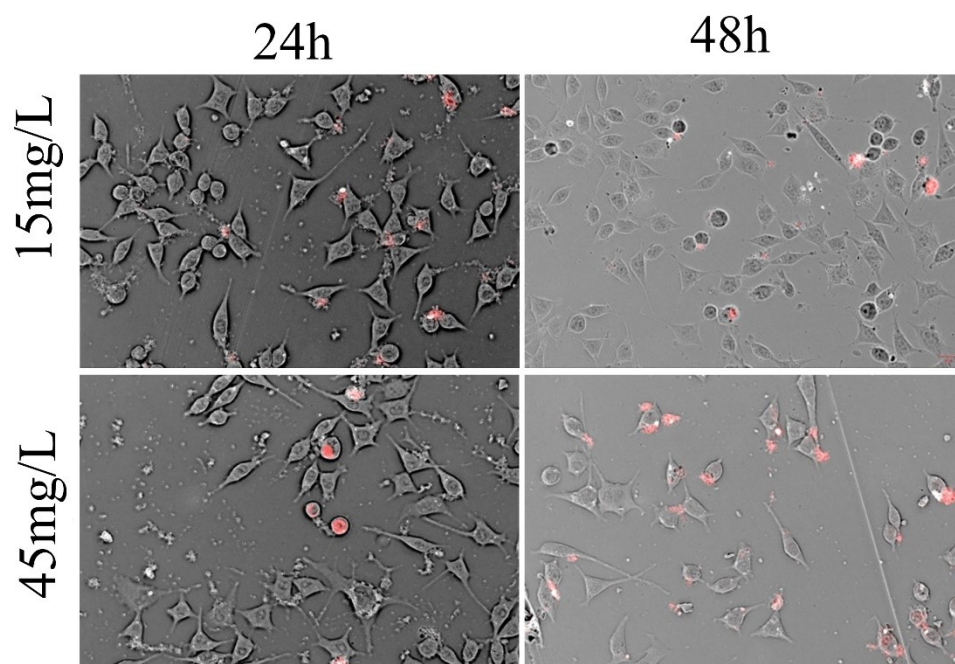


Fig. S8 the cellular uptake of the RhB labelled poly(HPMA)-Chl.



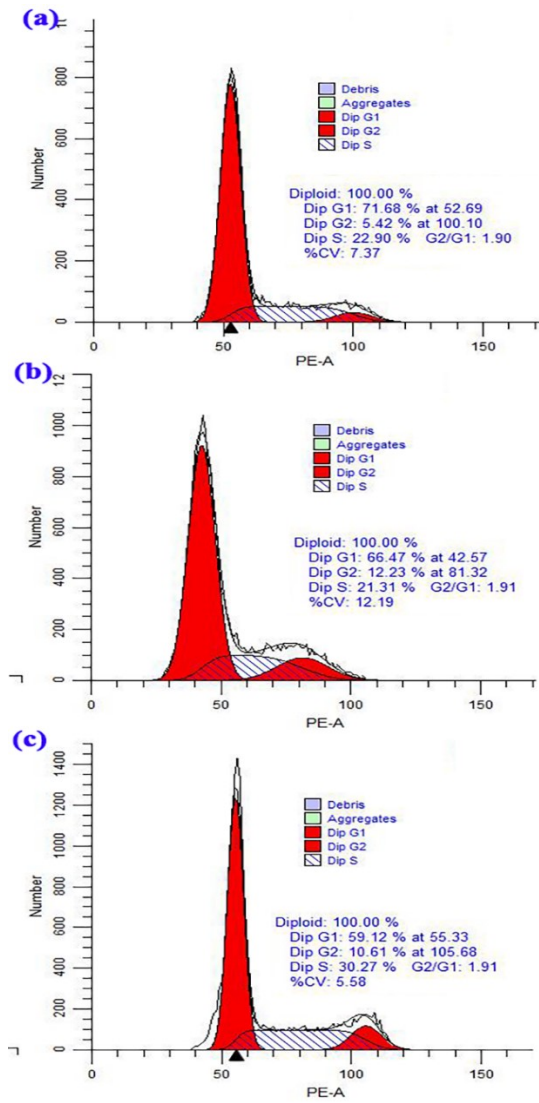


Fig. S9 The cycle data of MCF-7 cells incubated with Poly(HPMA)-Chl conjugate at different concentration (a) control, (b) 15 mg/L and (c) 45 mg/L for 48 h.

Table S1 The comparisons of the cell cycle

	Sub-G1	G0/G1	S	G2/M	Refs
PPAHC( $C_{\text{CHL}} = 60 \mu\text{g/mL}$ )	35.7%	35.5%	20.1%	12.2%	1
Platinum(IV)-chlorambucil complex (20 $\mu\text{M}$ )	-	28.05%	33.10%	38.85%	2
Chlorambucil Gemcitabine Conjugate(10 $\mu\text{M}$ )		47.8%	29.5%	22.7%	3
Mito-Chlor (2 $\mu\text{M}$ )		30%	10%	60%	4
ImImIm-(S) Chlg-PyPyPyPy-bDp	7%	48%	20%	24%	5
Poly(HPMA)-Chl 45mgChl/L	-	59.12%	30.27%	10.61%	This work

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