

Supporting Information

Esterase-Responsive Polymeric Prodrug-Based Tumor Targeting Nanoparticles for Improved Anti-Tumor Performance against Colon Cancer

Gang Pan^{1,#}, Yi-jie Bao^{1,#}, Jie Xu^{1,#}, Tao Liu², Cheng Liu², Yan-yan Qiu², Xiao-jing Shi¹, Hui Yu¹, Ting-ting Jia³, Xia Yuan³, Ze-ting Yuan², Pei-hao Yin^{1,4*}, and Yi-jun Cao^{1,*}

¹Department of General Surgery, Putuo Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai, 200062, China

²Centralab, Putuo Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai, 200062, China

³Department of Pharmacy, Putuo Hospital, Shanghai University of Traditional Chinese Medicine, Shanghai, 200062, China

⁴Interventional Cancer Institute of Chinese Integrative Medicine, Shanghai University of Traditional Chinese Medicine, Shanghai, 200062, China

#These authors contributed equally to this work

* To whom correspondence should be addressed. E-mail: yinpeihao1975@hotmail.com

caoyjxw@126.com

Table S1. Molecular parameters of polymers used in this study.

| Sample | M_n (g/mol) ^a | D_M ^a | M_n (g/mol) ^b |
|--|----------------------------|--------------------|----------------------------|
| P(OEGMA _{0.45} - <i>co</i> -BSMA _{0.55}) ₇₂ | 25,000 | 1.29 | 24,800 |
| P(OEGMA _{0.45} - <i>co</i> -BUF _{0.46} - <i>co</i> -NHS _{0.09}) ₇₂ | 37,700 | 1.21 | 37,700 |
| P(OEGMA _{0.45} - <i>co</i> -BUF _{0.46} - <i>co</i> -NHS _{0.062} - <i>co</i> -RGD _{0.028}) ₇₂ | 38,900 | 1.32 | 38,900 |
| P(OEGMA _{0.45} - <i>co</i> -BUF _{0.46} - <i>co</i> -NHS _{0.048} - <i>co</i> -RGD _{0.028} - <i>co</i> -Cy5 _{0.014}) ₇₂ | 39,000 | 1.31 | 39,500 |
| P(OEGMA _{0.45} - <i>co</i> -BUF _{0.46} - <i>co</i> -NHS _{0.076} - <i>co</i> -Cy5 _{0.014}) ₇₂ | 39,100 | 1.30 | 38,400 |

^a Determined by GPC using DMF as the eluent (1.0 mL/min). ^b Calculated from ¹H NMR results.

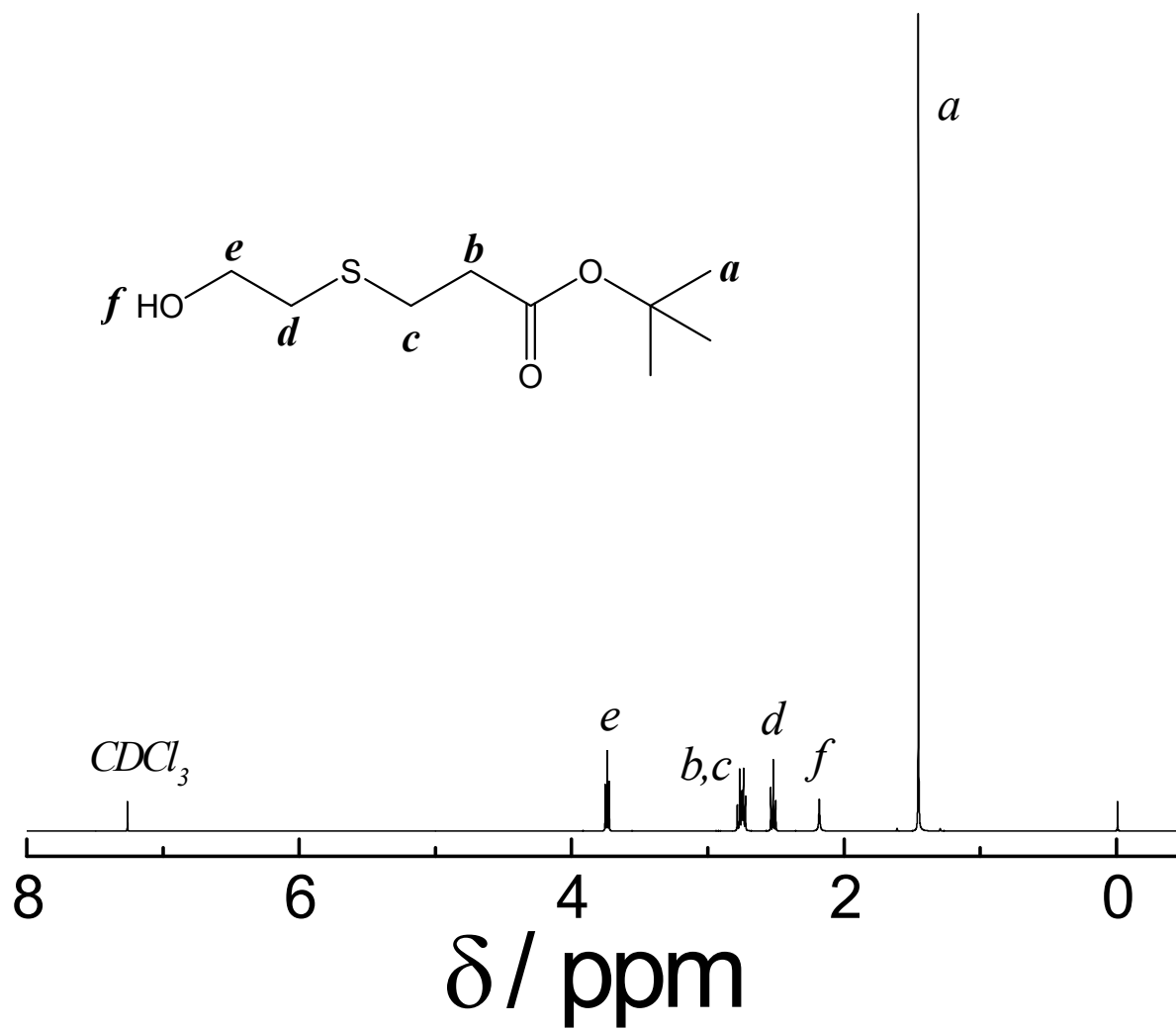


Fig. S1. ¹H NMR spectrum of *tert*-butyl-3-((2-hydroxyethyl)thio)propanoate in CDCl₃.

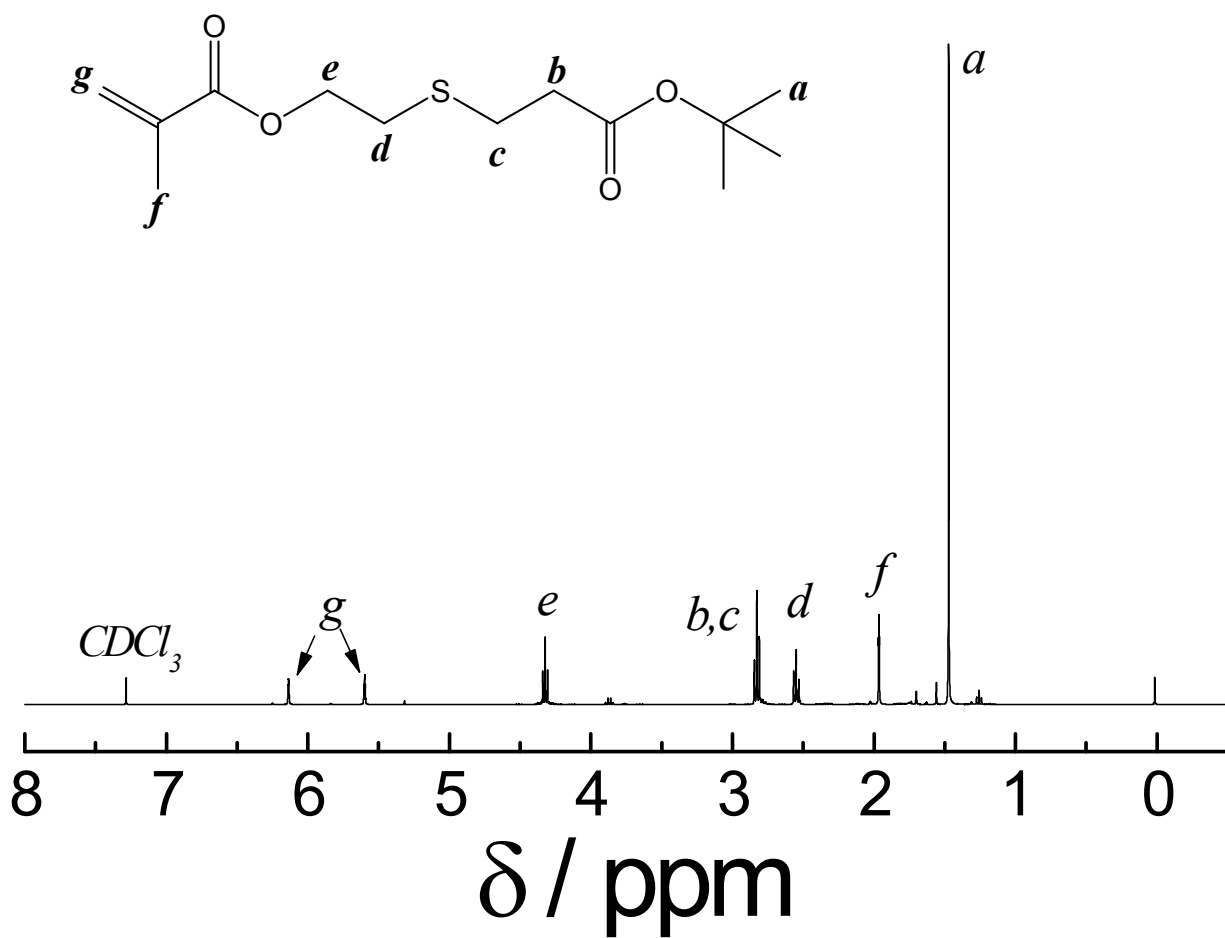


Fig. S2. ^1H NMR spectrum of 2-((3-(*tert*-butoxy)-3-oxopropyl)thio)ethyl methacrylate in CDCl_3 .

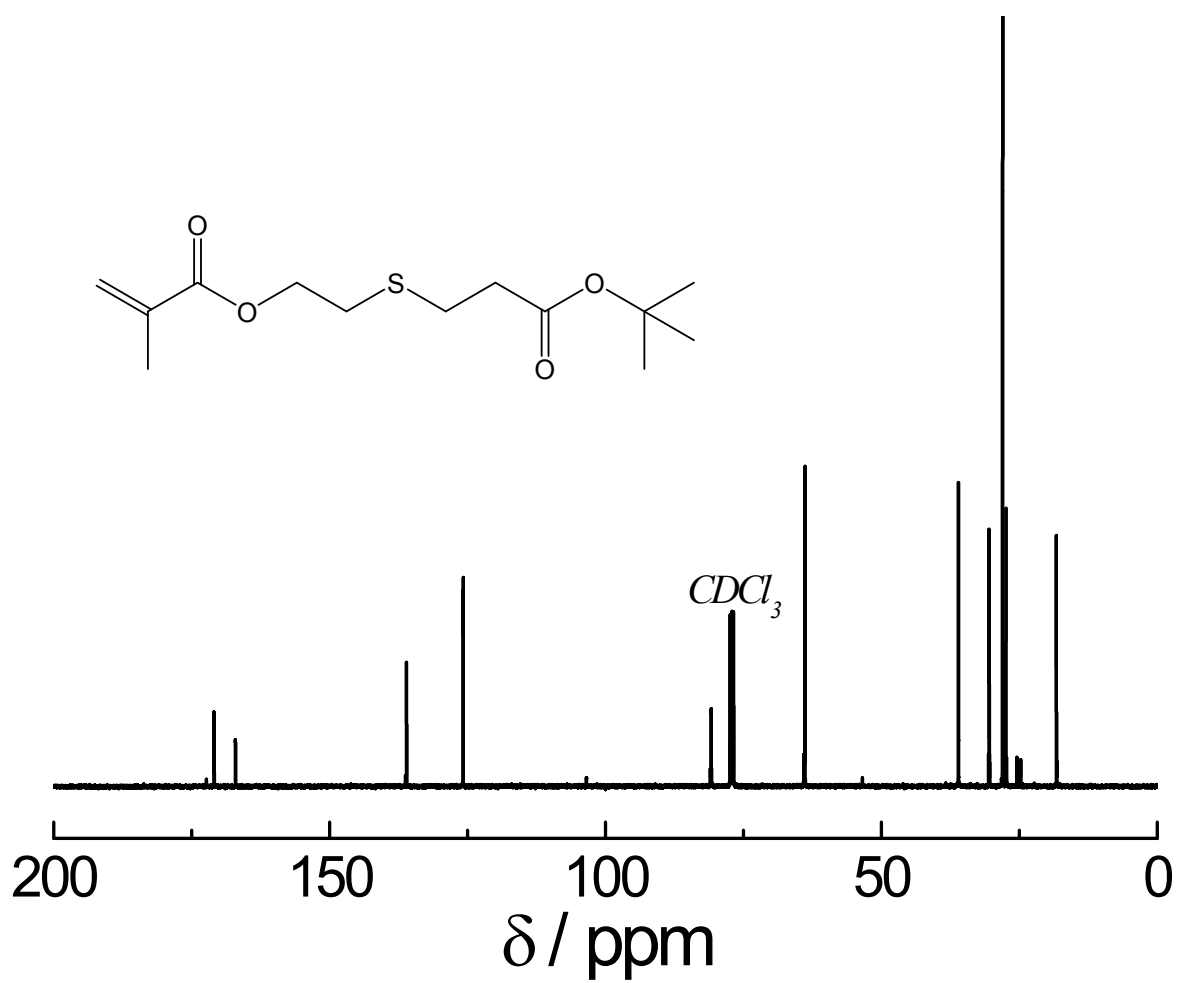


Fig. S3. ¹³C NMR spectrum of 2-((3-(*tert*-butoxy)-3-oxopropyl)thio)ethyl methacrylate in CDCl₃.

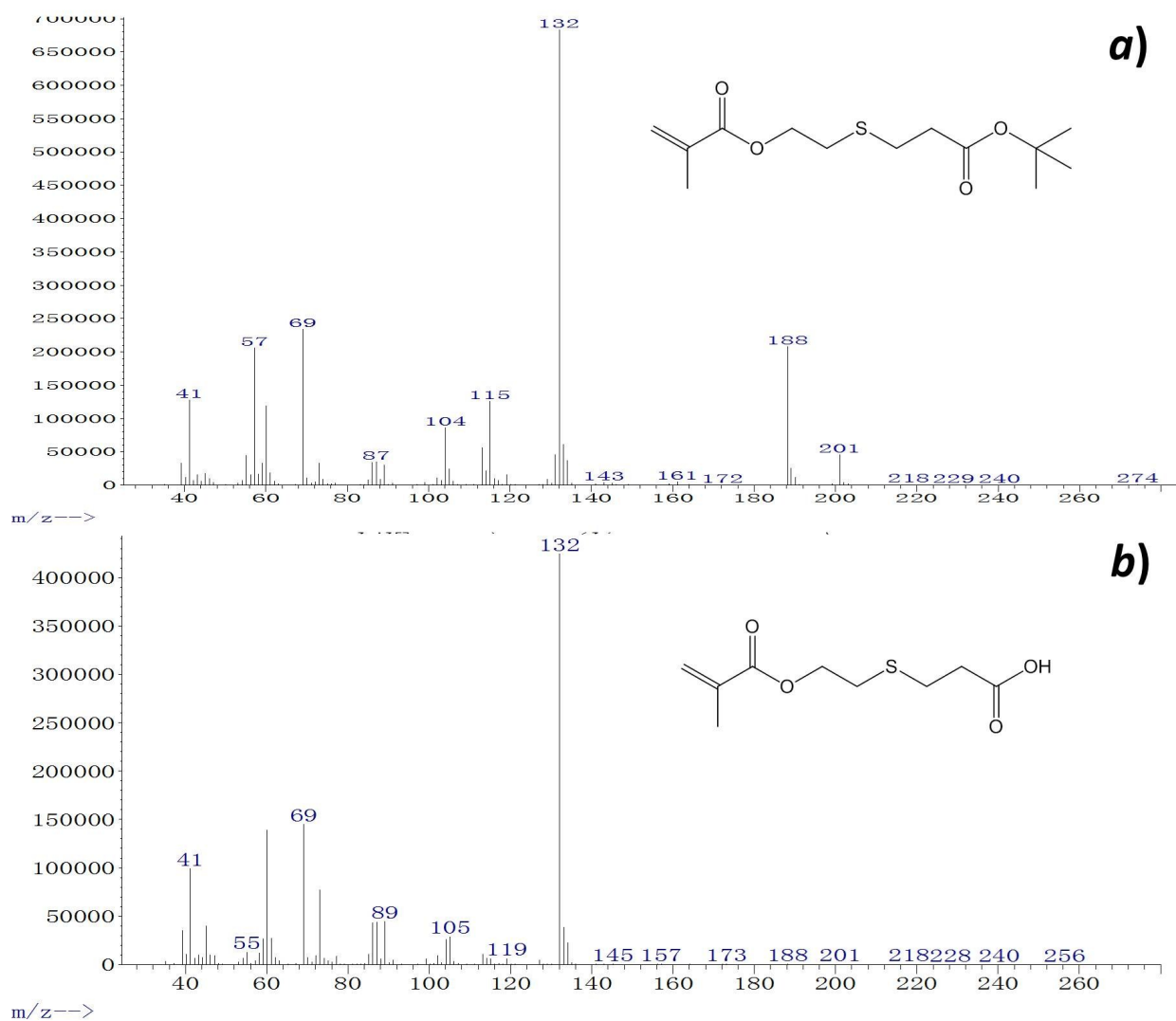


Fig. S4. ESI-Mass spectra of (a) 2-((3-(*tert*-butoxy)-3-oxopropyl)thio)ethyl methacrylate (m/z 188, calculated for $C_9H_{10}O_2S$: 188) and (b) 3-((2-(methacryloyloxy)ethyl)thio)propanoic acid (BSMA) (m/z 132, calculated for $C_5H_8O_2S$: 132).

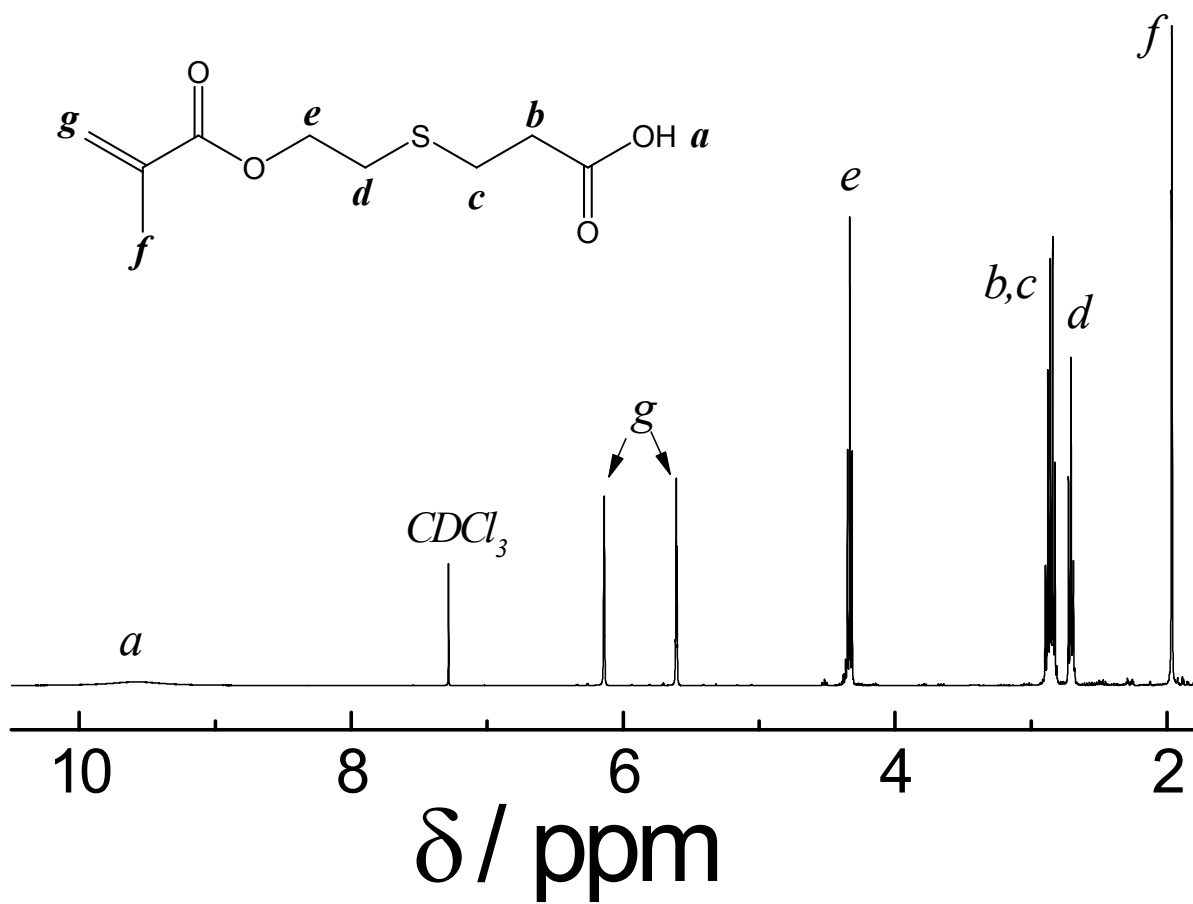


Fig. S5. ¹H NMR spectrum of BSMA in CDCl₃.

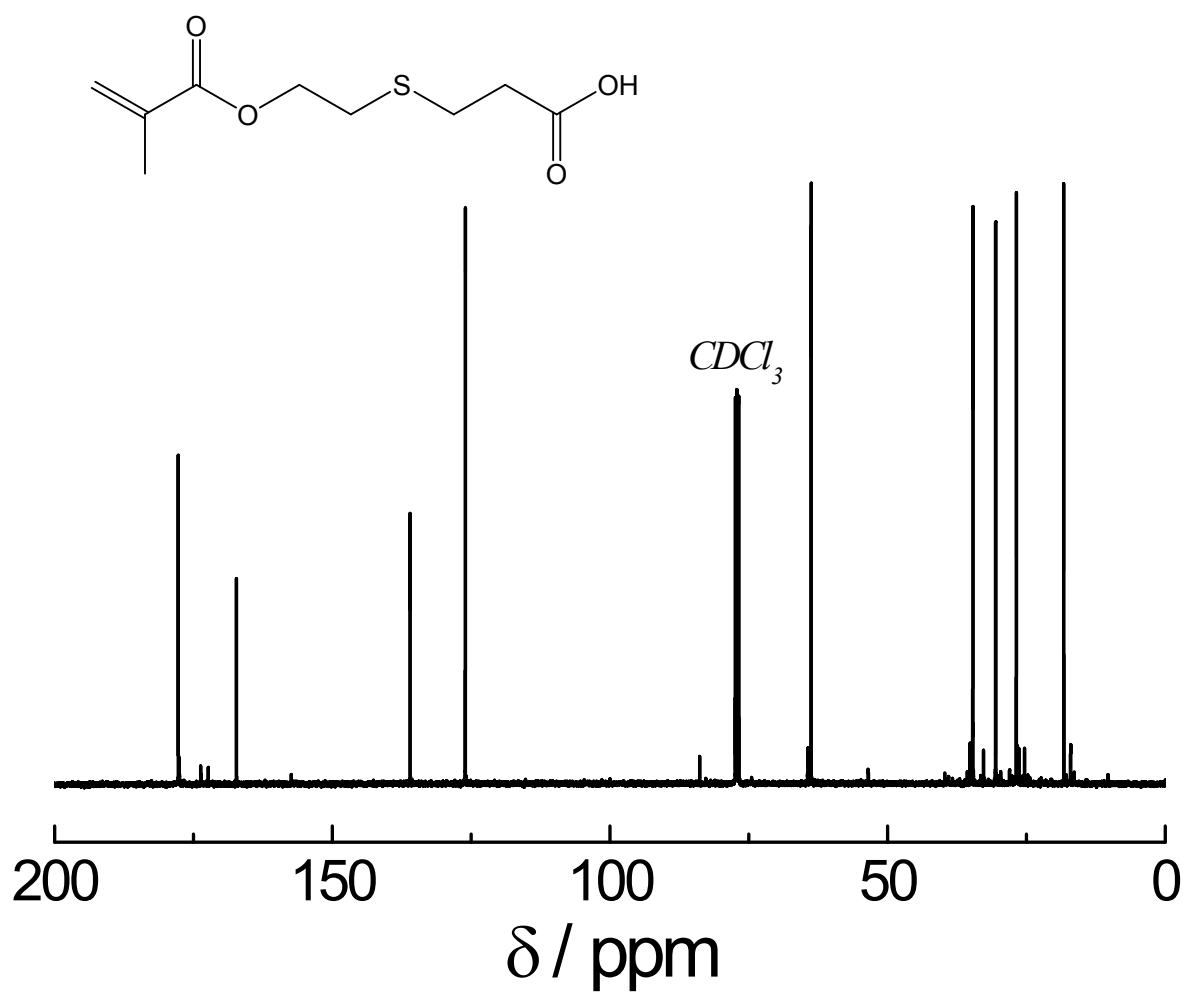


Fig. S6. ^{13}C NMR spectrum of BSMA in CDCl_3 .

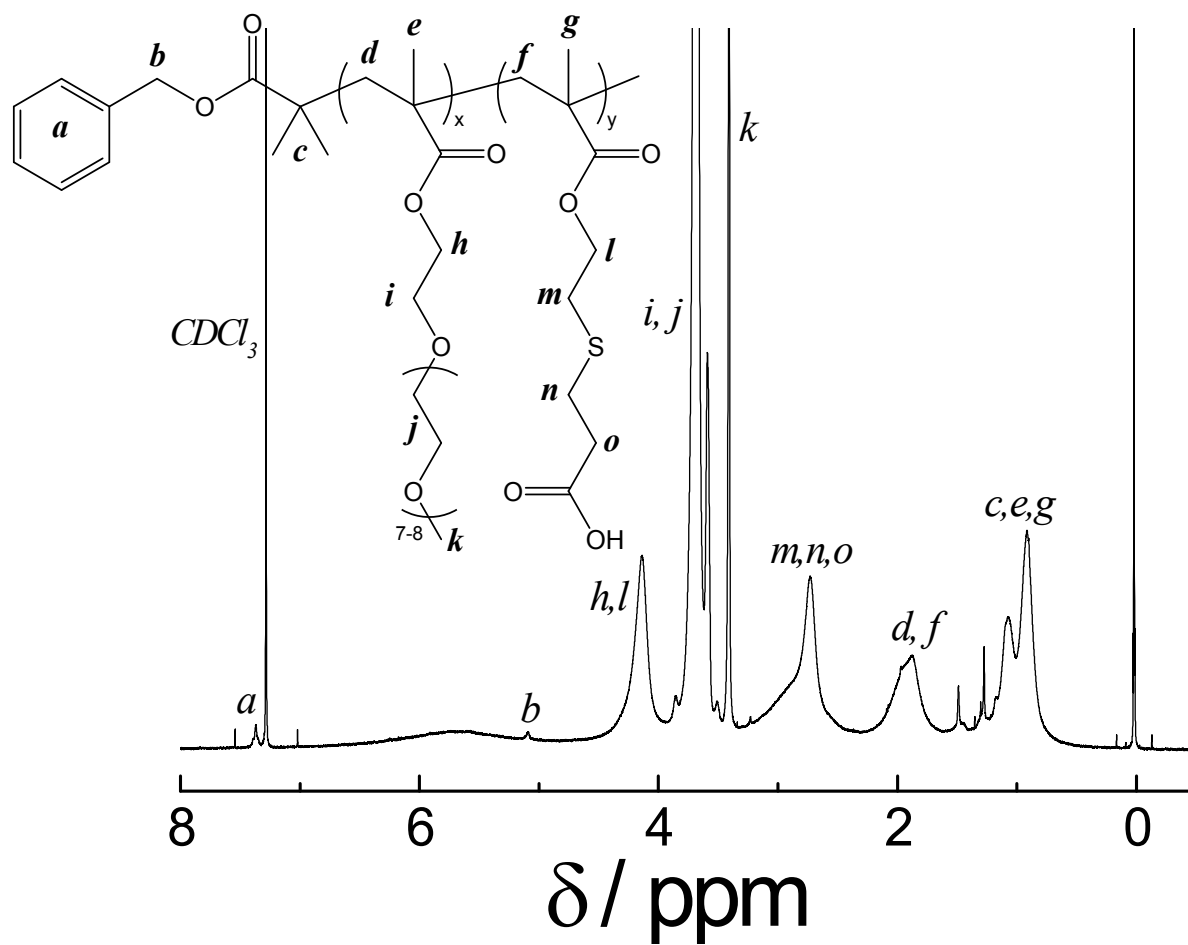


Fig. S7. ¹H NMR spectrum of P(OEGMA-co-BSMA) in CDCl₃.

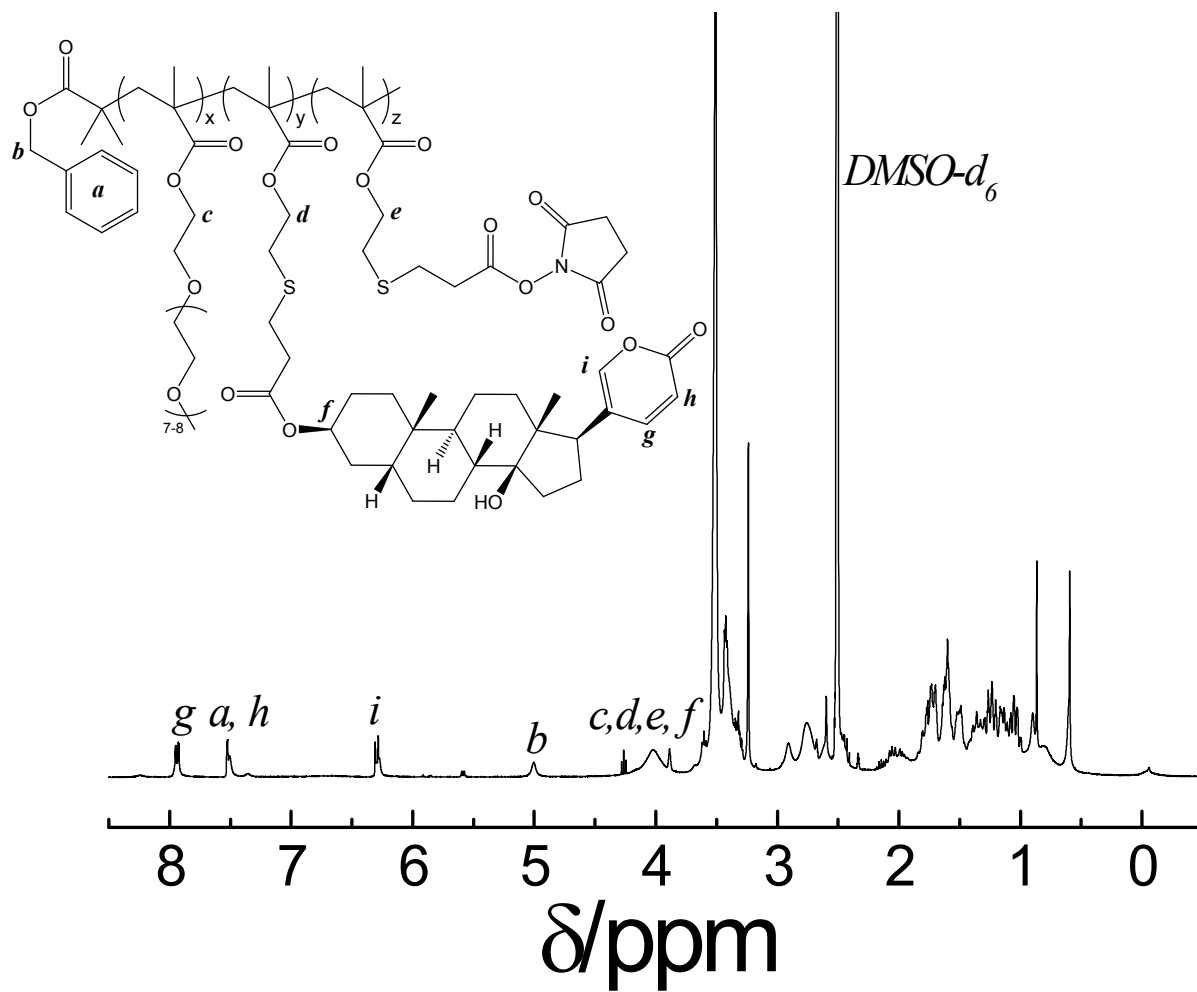


Fig. S8. ^1H NMR spectrum of P(OEGMA-co-BUF) in $\text{DMSO-}d_6$.

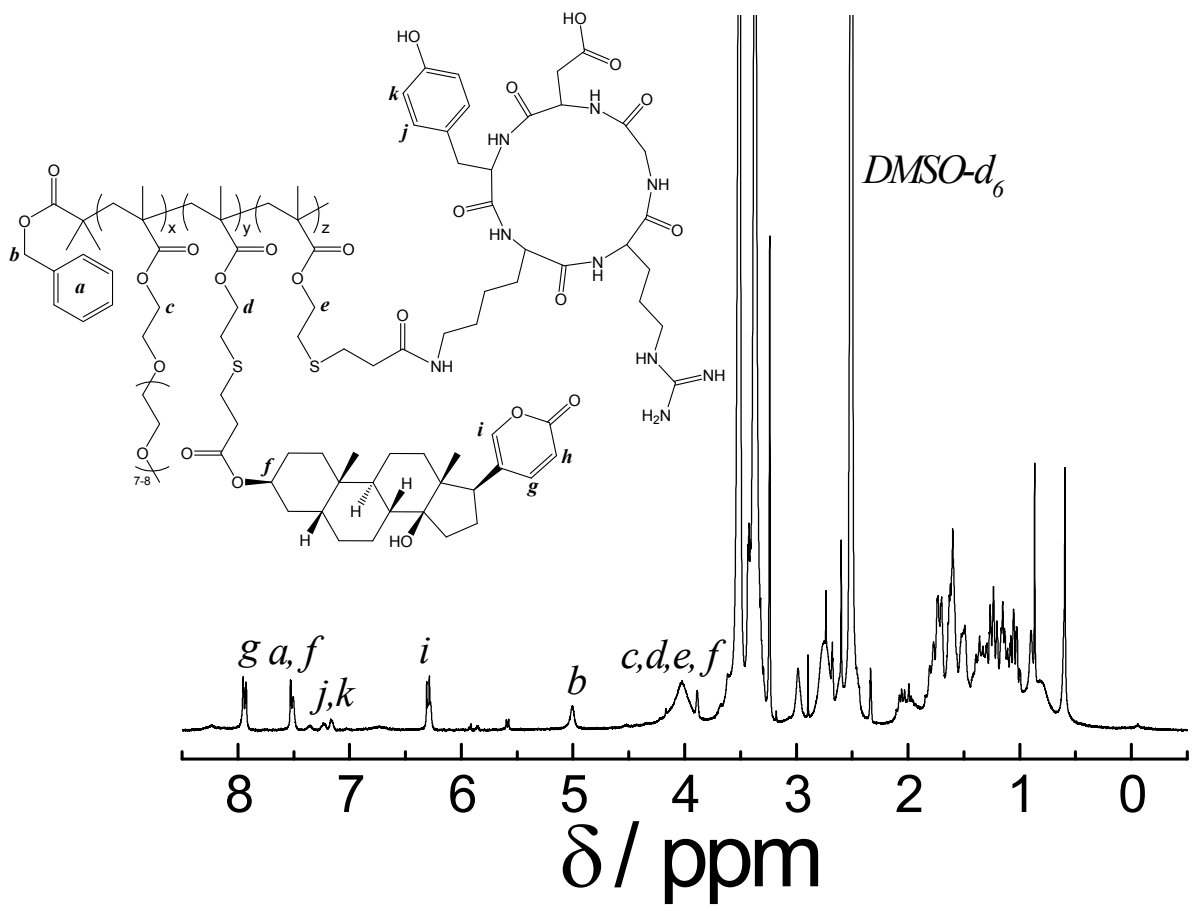


Fig. S9. ¹H NMR spectrum of P(OEGMA-co-BUF-co-RGD) in DMSO-d₆.

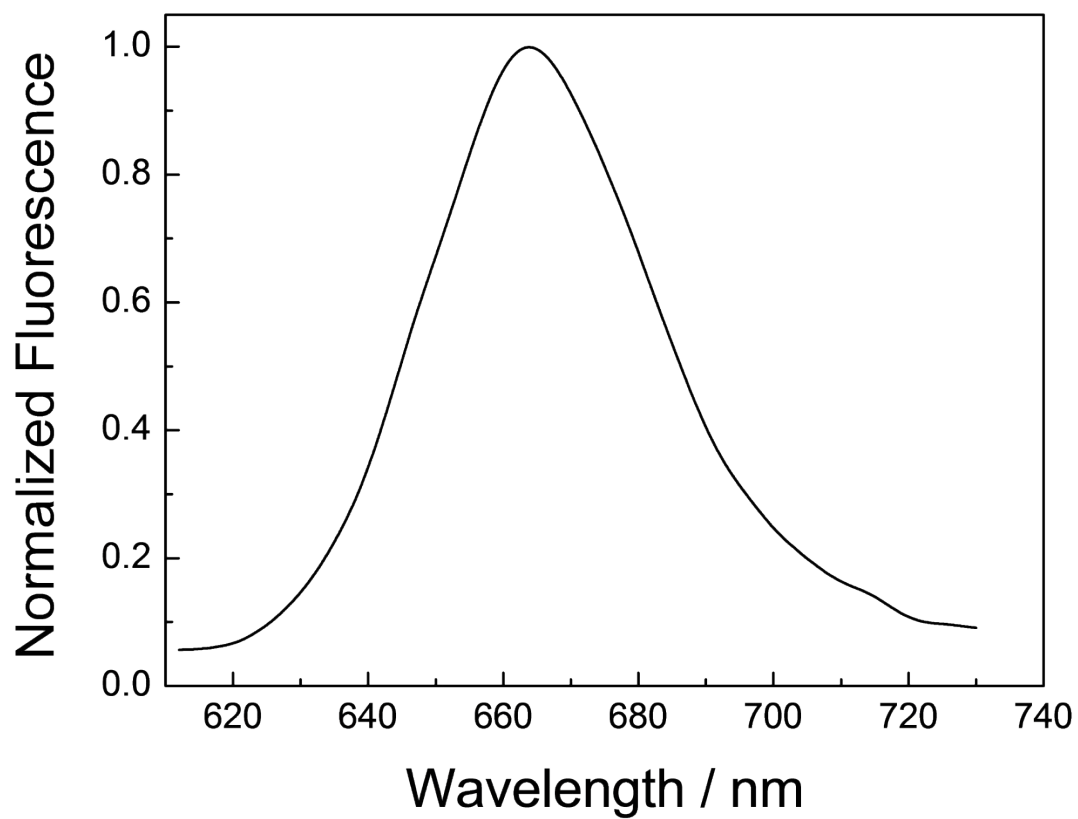


Fig. S10. Fluorescence emission spectrum recorded for the aqueous micellar solution of P(OEGMA-*co*-BUF-*co*-RGD-*co*-Cy5) ($[\text{Cy5}] = 3.0 \times 10^{-6} \text{ M}$; $\lambda_{\text{ex}} = 633 \text{ nm}$; slit widths: Ex. 5 nm, Em. 5 nm).

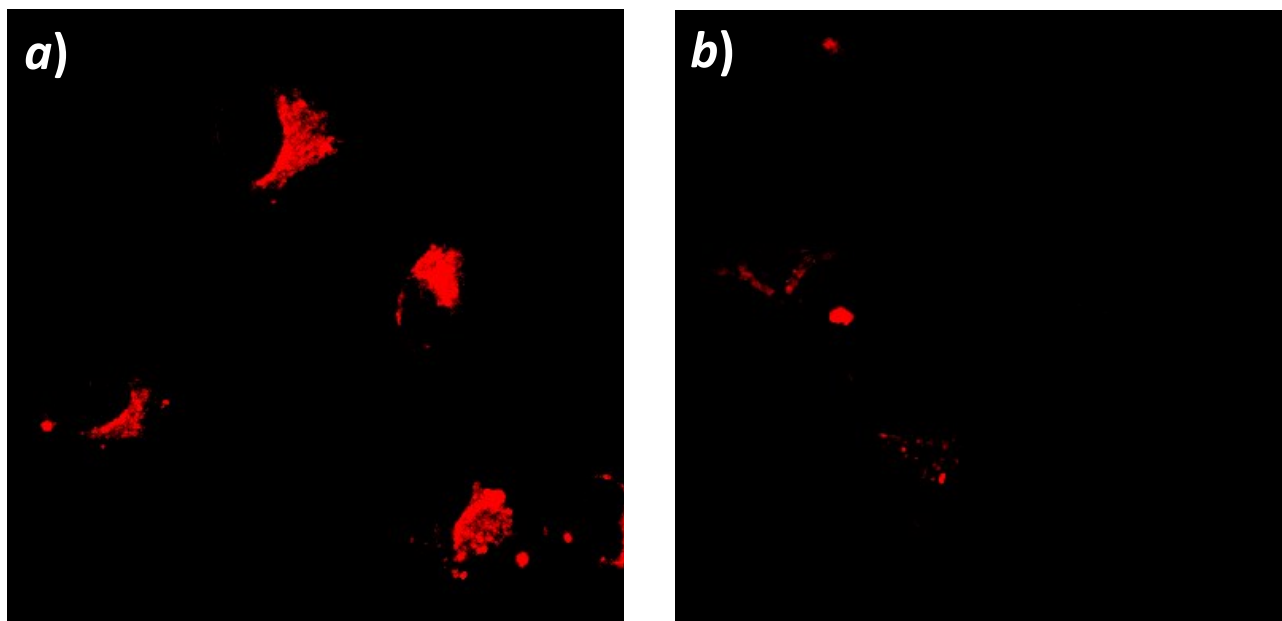


Fig. S11. Typical confocal microscopy fluorescence images recorded for LoVo cells after incubating at 37 °C with P(OEGMA-*co*-BUF-*co*-RGD-*co*-Cy5) ([Cy5] = 3.0×10^{-6} M) for 4 h (a) without and (b) with an excess of free RGD (20-fold equivalent of RGD in P(OEGMA-*co*-BUF-*co*-RGD-*co*-Cy5)) to block RGD-mediated endocytosis of nanoparticles.