Supplementary Information

Micellar Nanoformulation of Lipophilized Bortezomib: High Drug Loading, Improved Tolerability and Targeted Treatment of Triple Negative Breast Cancer

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Synthesis and characterization of cRGD-PEG-P(TMC-co-DTC) and PEG-P(TMC-co-DTC)

cRGD-functionalized PEG-P(TMC-*co*-DTC) diblock copolymer was prepared by statistical copolymerization of TMC and DTC using squaric acid ethyl ester end-functionalized PEG (SAE-PEG-OH, M_n=7.5 kg/mol) as an initiator followed by coupling with cRGD-NH_2 (**Scheme S1**). SAE-PEG-P(TMC-*co*-DTC) was obtained with a controlled M_n of 7.5-(3.8-1.8) kg/mol and an M_w/M_n of 1.18 (**Table S1**). ^{1}H NMR spectrum displayed characteristic peaks of SAE moiety at δ 4.65 and 1.34 (**Fig. S4A**), indicating that SAE groups were intact during reaction and workup. Further reaction with cRGD resulted in complete disappearance of signals of SAE groups (**Fig. S4B**). Instead, phenyl protons assignable to cRGD moieties were detected at δ 7.22. BCA protein assays revealed that cRGD-PEG-P(TMC-*co*-DTC) had a high cRGD functionality of 98.6%. In a similar way, PEG-P(TMC-*co*-DTC) was synthesized with an M_n of 5.0-(3.7-1.8) kg/mol and an M_w/M_n of

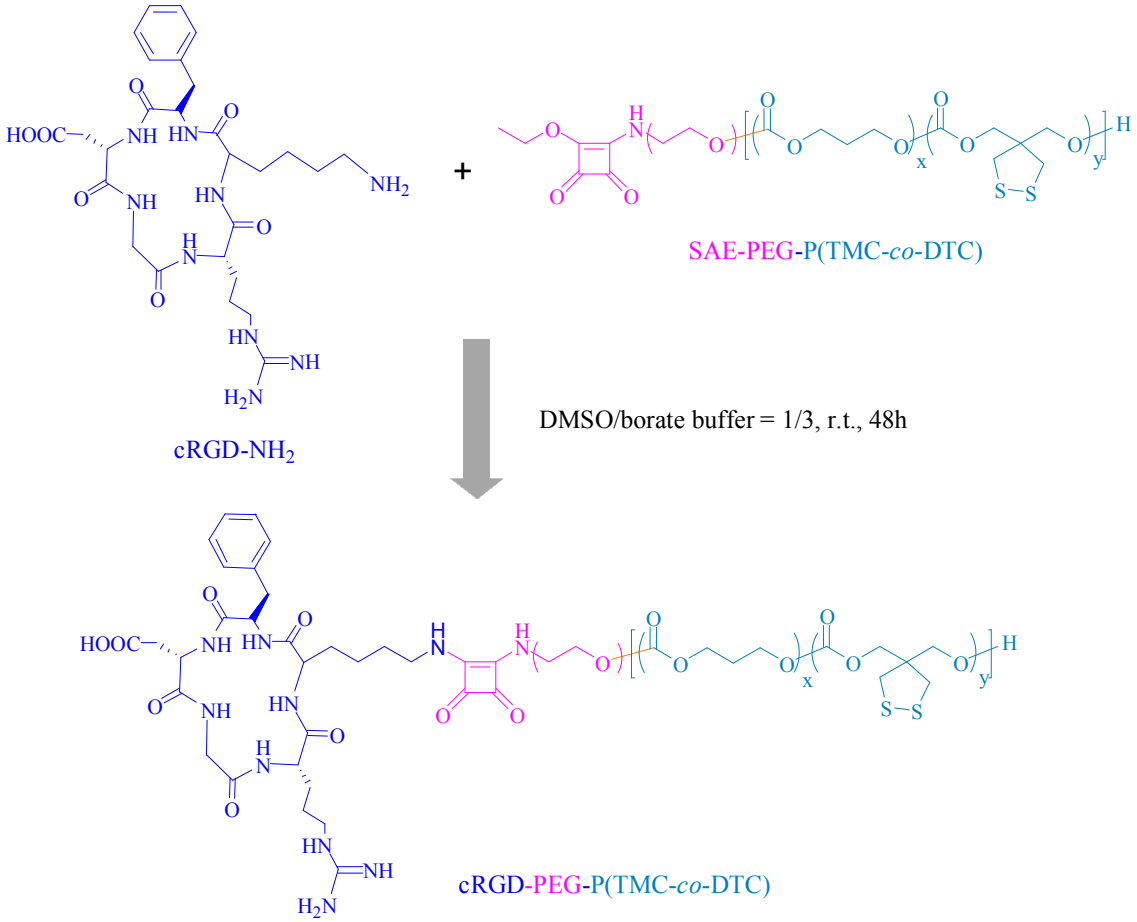
1.12 using MeO-PEG-OH (M_n=5.0 kg/mol) as an initiator (**Fig. S4C, Table S1**)

Table S1. Characterization of cRGD-PEG-P(TMC-*co*-DTC) and PEG-P(TMC-*co*-DTC).

copolymer	M_n (kg/mol)		M_w/M_n [b]	Yield (%)
	design	determined[a]		
PEG-P(TMC-DTC)	5.0-(4.0-2.0)	5.0-(3.7-1.8)	1.12	88.6
cRGD-PEG-P(TMC-DTC)	7.5-(4.0-2.0)	7.5-(3.8-1.8)	1.18	84.2

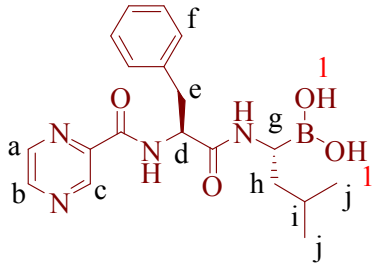
[a.] Determined by ^{1}H NMR.

[b.] Determined by gel permeation chromatography (GPC).



Scheme S1. Synthesis of cRGD-PEG-P(TMC-*co*-DTC).

A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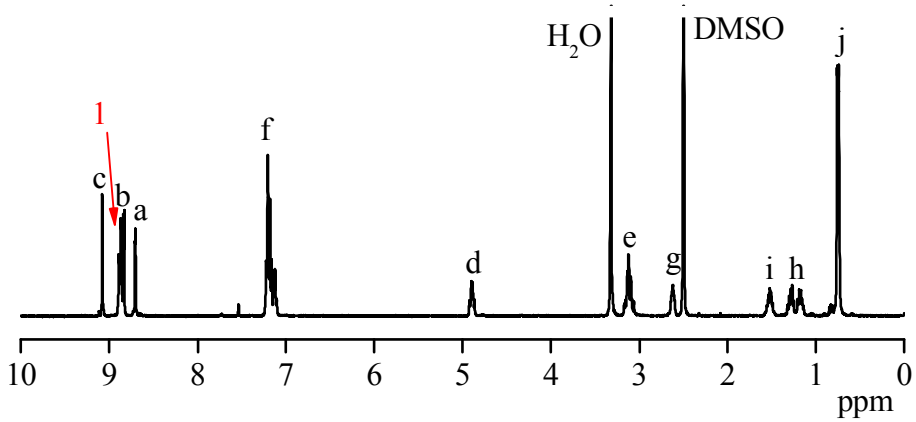
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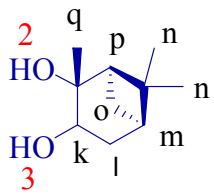
B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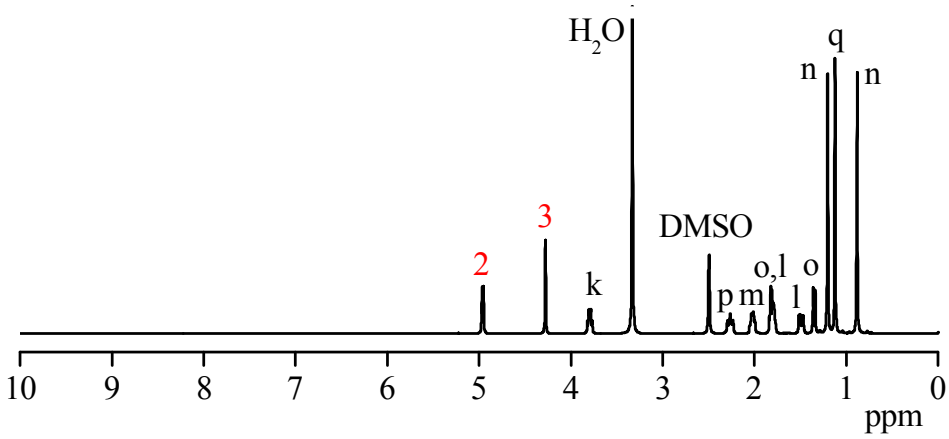
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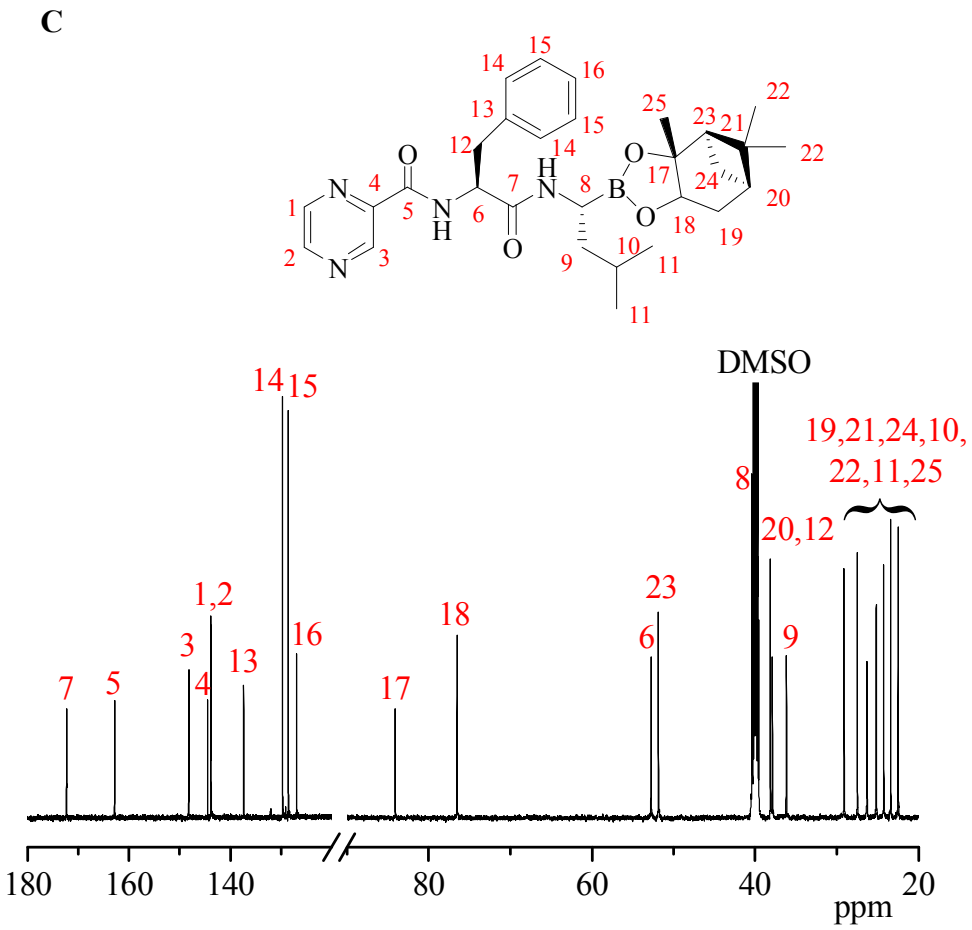


Figure S1. ^{1}H NMR spectra (400 MHz, DMSO-d_6) of BTZ (A) and pinanediol (B). (C) ^{13}C NMR spectrum (150 MHz, DMSO-d_6) of BP.

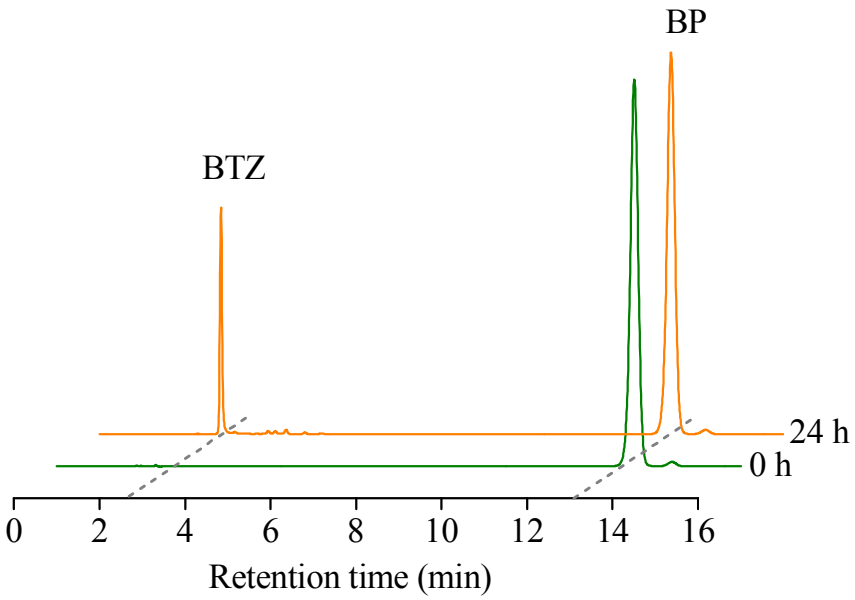


Figure S2. Reverse-phase HPLC chromatograms of BP and its hydrolytic products obtained in DMSO/PBS (pH 7.4, 10 mM) (1/9, v/v) at 37 °C and pH 7.4.

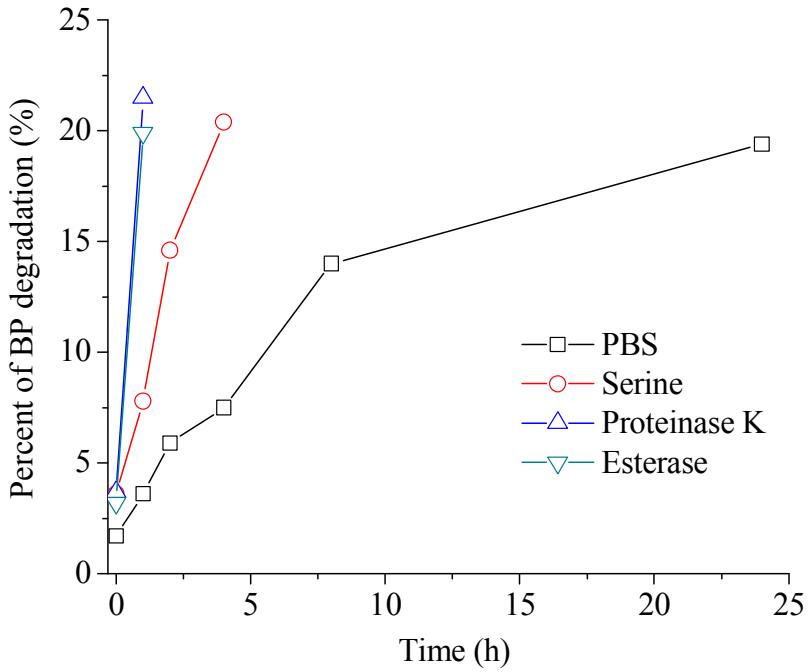
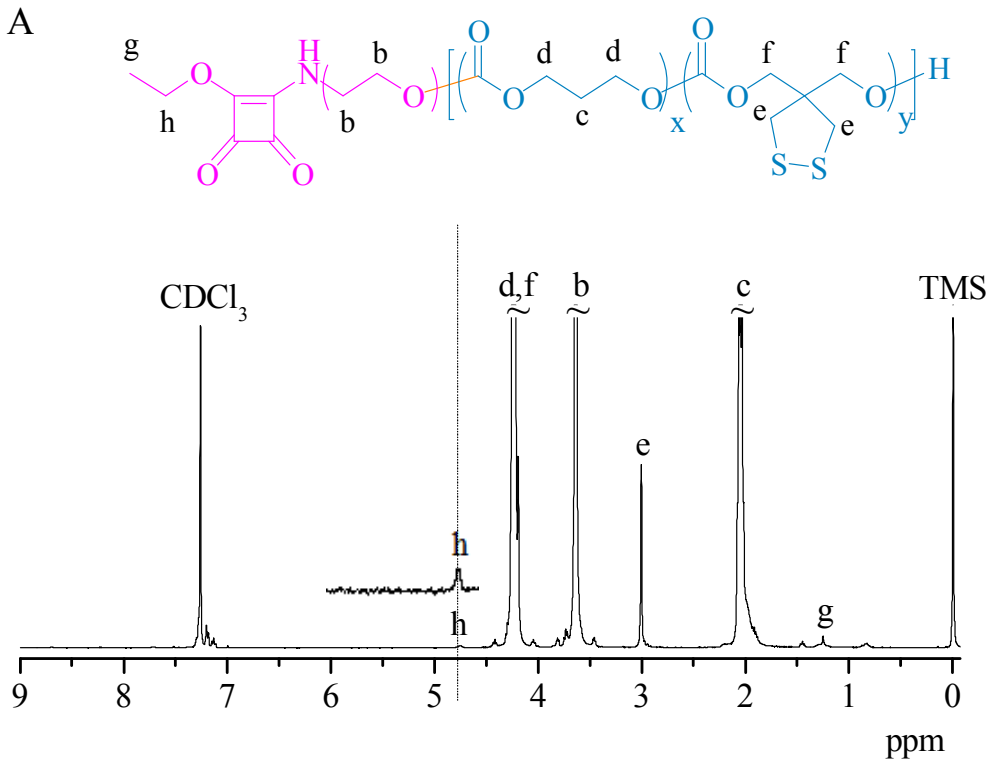


Figure S3. Degradation kinetics of BP in PBS (pH 7.4, 10 mM) or PBS containing serine (5 equiv. with respect to borate ester), proteinase K (15 IU/mL), or esterase (500 IU/mL).



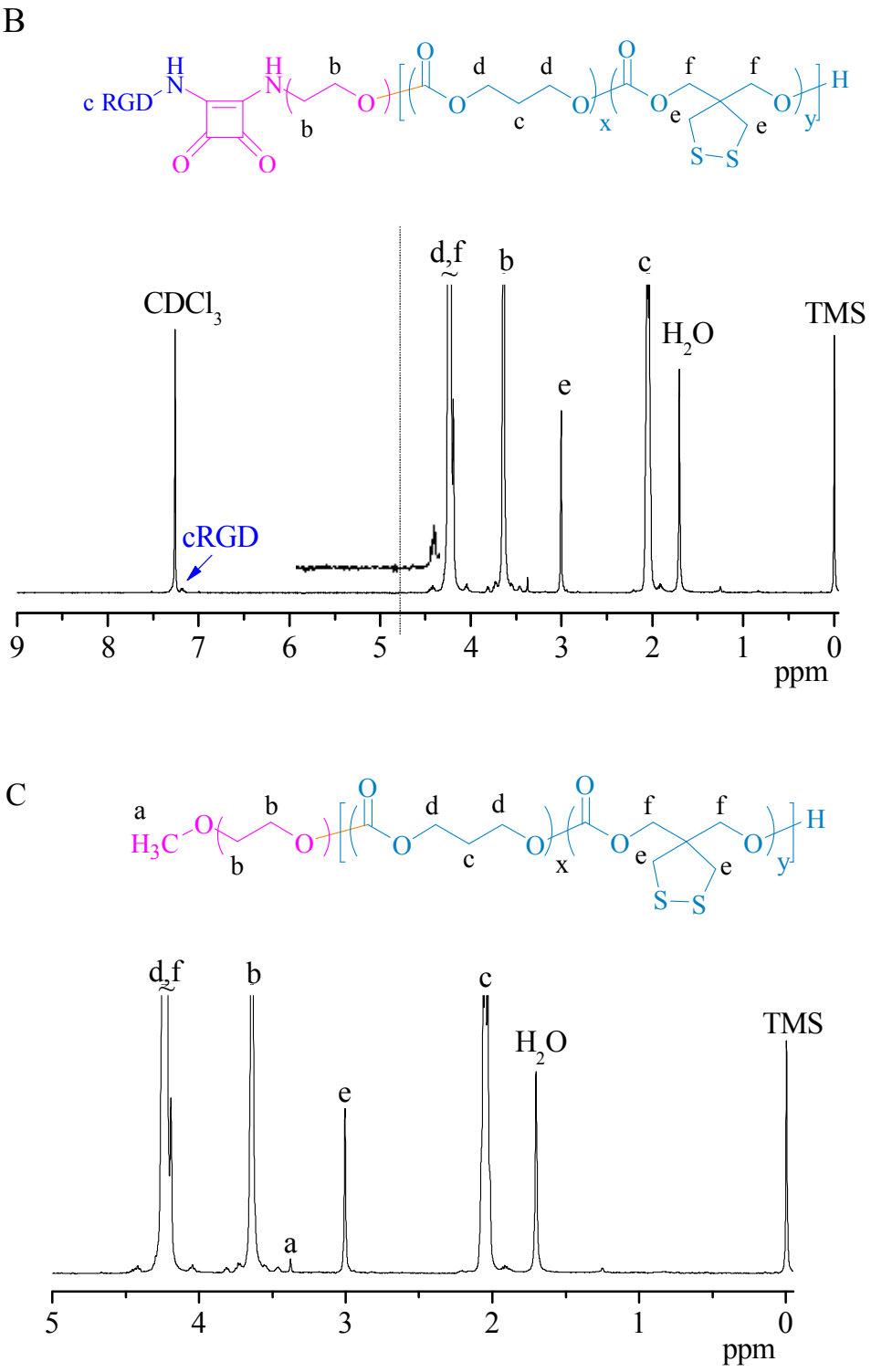


Figure S4. ^{1}H NMR spectra (600 MHz, $CDCl_3$) of SAE-PEG-P(TMC-*co*-DTC) (A), cRGD-PEG-P(TMC-*co*-DTC) (B), and MeO-PEG-P(TMC-*co*-DTC) (C).