

Supporting Information

Multi pH-sensitive polymer-drug conjugate mixed micelles for efficient co-delivery of doxorubicin and curcumin to synergistically suppress tumor metastasis

Yuanhang Zhou, Chuhan Zhou, Yang Zou, Yao Jin, Shidi Han, Qi Liu, Xinping Hu, Leqi Wang, Yining Ma, and Yan Liu*

*Beijing Key Laboratory of Molecular Pharmaceutics and New Drug Delivery Systems,
School of Pharmaceutical Sciences, Peking University, Beijing 100191, China*

*Corresponding author. Phone: 86 10 82801508; e-mail: yanliu@bjmu.edu.cn

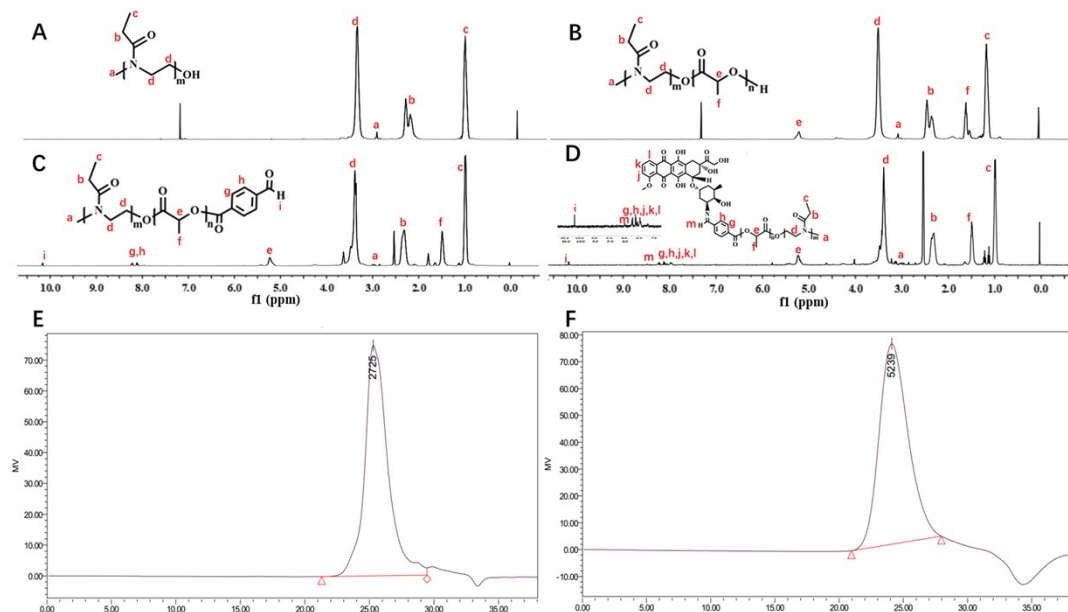


Figure S1. ^1H NMR spectra of PEOz-OH (A), PEOz-PLA (B), PEOz-PLA-CHO (C), PEOz-PLA-imido-DOX (D), GPC curves of PEOz-OH (E) and PEOz-PLA (F).

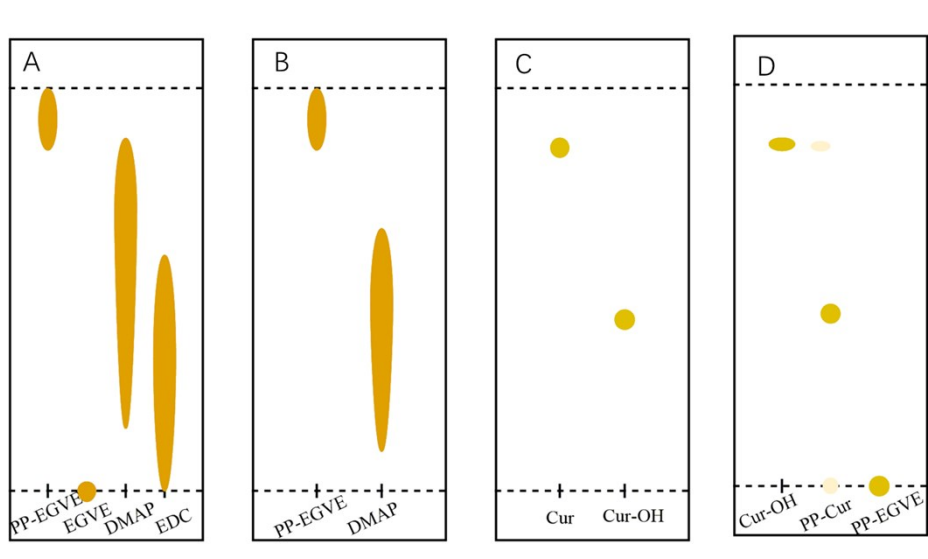


Figure S2. (A) The TLC results of PEOz-PLA-EGVE (PP-EGVE), EGVE, DMAP and EDC. Iodine was used as color reagent. (B) The TLC results of PEOz-PLA-EGVE (PP-EGVE) and DMAP. Iodine was used as color reagent. (C) The TLC results of Cur and Cur-OH under natural light. (D) The TLC results of Cur-OH, PEOz-PLA-ace-Cur (PP-Cur) and PEOz-PLA-EGVE (PP-EGVE). Iodine was used as color reagent.

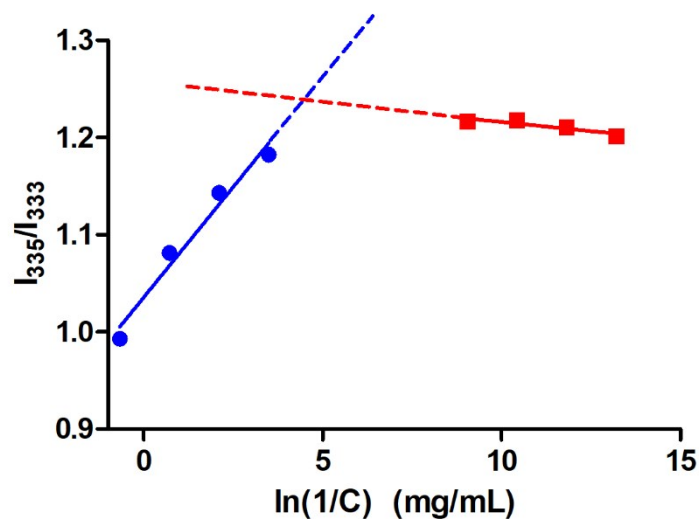


Figure S3. Plot of I_{333}/I_{335} vs. $\ln(1/C)$ for the mixture of PEOz-PLA-ace-Cur and PEOz-PLA-imi-DOX in 5:1 mass ratio of Cur to DOX.

Table S1. Physicochemical characteristics of various micelles (n=3).

Micelles	Diameter (nm)	PDI□	LC (%)		Cur:DOX (w/w)
			DOX	Cur	
PP-DOX-PMs	24.31±2.04	0.28±0.02	6.27	--	--
PP-Cur-PMs	143.17±0.05	0.17±0.01	--	9.52	--
PP-Cur1/PP-DOX1-Mix-PMs	40.31±2.18	0.83±0.08	3.48	4.09	1.17
PP-Cur3/PP-DOX1-Mix-PMs	69.67±2.06	0.58±0.02	1.94	6.25	3.23
PP-Cur5/PP-DOX1-Mix-PMs	104.64±0.94	0.15±0.01	1.60	7.96	4.96
PP-Cur10/PP-DOX1-Mix-PMs	113.33±1.94	0.18±0.01	0.93	8.52	9.16
PP-Cur25/PP-DOX1-Mix-PMs	128.53±0.29	0.14±0.01	0.35	9.05	25.92
Cur5+DOX1/PP-PMs	22.97±1.31	0.27±0.03	1.54	8.33	5.41

Table S2. IC₅₀ and CI values of free drugs and various micelle formulations against MDA-MB-231 cells by SRB assay (*n*=6).

□ Formulation	IC ₅₀ (μg/mL)		CI
	DOX	Cur	
DOX·HCl	0.26±0.11	-	-
Cur	-	5.91±1.49	-
PP-DOX-PMs	0.32±0.07 ^a	-	-
PP-Cur-PMs	-	5.81±0.78 ^b	-
PP-Cur1/PP-DOX1-Mix-PMs	0.17±0.01 ^c	0.22±0.02 ^f	0.57
PP-Cur3/PP-DOX1-Mix-PMs	0.17±0.01 ^c	0.53±0.02 ^f	0.61
PP-Cur5/PP-DOX1-Mix-PMs	0.13±0.04 ^d	0.66±0.04 ^f	0.53
PP-Cur10/PP-DOX1-Mix-PMs	0.16±0.02 ^c	1.45±0.20 ^f	0.75
PP-Cur25/PP-DOX1-Mix-PMs	0.19±0.05 ^c	5.01±1.28 ^c	1.47

a: *p*>0.05 vs. DOX·HCl.

b: *p*>0.05 vs. Cur.

c: *p*<0.05 vs. PP-DOX-PMs.

d: *p*<0.001 vs. PP-DOX-PMs.

e: *p*<0.05 vs. PP-Cur-PMs.

f: *p*<0.001 vs. PP-Cur-PMs.

g: *p*>0.05 vs. PP-Cur-PMs.

Table S3. Blood routine index of Balb/c nude mice after treatment with various formulations

(n=3).

Blood routine index	Saline	PP- DOX-PMs	Cur5+DOX1/PP-PMs	PP-Cur5/PP-DOX1-Mix-PMs
GR	3.67±0.41	4.07±1.72	5.43±0.83	8.27±3.81
GR%	40.6±3.26	51±5.63	64.37±11.3	68.27±6.38**
HCT	61.33±7.45	49.13±2.81	52.63±6.97	54.57±7.16
HGB	217.67±32.87	183±12.57	197.67±23.8	198.33±26.61
LY	4.87±0.92	3.43±0.73	2.7±0.99	3.07±0.39**
LY%	53.13±3.71	45.67±6.13	31.17±10.19*	28.13±5.56
MCH	15.4±0.4	16.17±0.33	15.7±1.48	17.35±0.75
MCHC	353.33±11.61	372.67±9.88	376±13.74	363.33±2.05
MCV	44.05±0.25	43.37±0.26	41.63±2.9	47.7±2.5
MO	0.57±0.05	0.27±0.09*	0.4±0.16	0.4±0.08
MO%	6.27±0.46	3.33±0.82*	4.47±1.88	3.6±1.07
MPV	5.23±0.12	5.03±0.12	4.9±0.22	4.97±0.09
PCT	0.31±0.01	0.57±0.08**	0.45±0.07*	0.39±0.07
PDW	13.8±0.14	13.47±0.31	12.47±0.57*	13.7±0.71
PLT	583.67±36.65	1122.33±141.88**	926.33±177.6	784.67±146.34
RBC	13.15±1.65	11.33±0.61	12.63±1.34	10.77±1.83
RDW	13.17±0.74	12.93±0.17	14.13±1.83	12.8±0.08
WBC	9.1±1.28	7.77±2.45	8.53±0.4	11.73±4.26

* $p < 0.05$, ** $p < 0.001$, vs saline.

Abbreviation:

GR, granulocyte; HCT, hematocrit; HGB, hemoglobin; LY, lymphocyte; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; MCV,

mean corpuscular volume; MO, monocyte; MPV, mean platelet volume; PCT, plateletcrit; PDW, platelet distribution width; PLT, platelet count; RBC, red blood cell; RDW, red blood cell volume distribution width; WBC, white blood cell.