Supporting Information

Hyaluronic acid functional amphipathic and redox-responsive polymer particles for co-delivery of doxorubicin and cyclopamine to eradicate breast cancer cells and cancer stem cells

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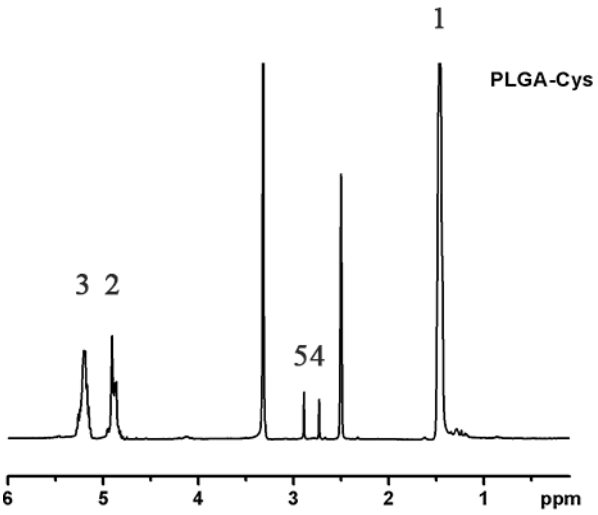

Figure S1. ^{1}H NMR spectrum of PLGA-Cys.

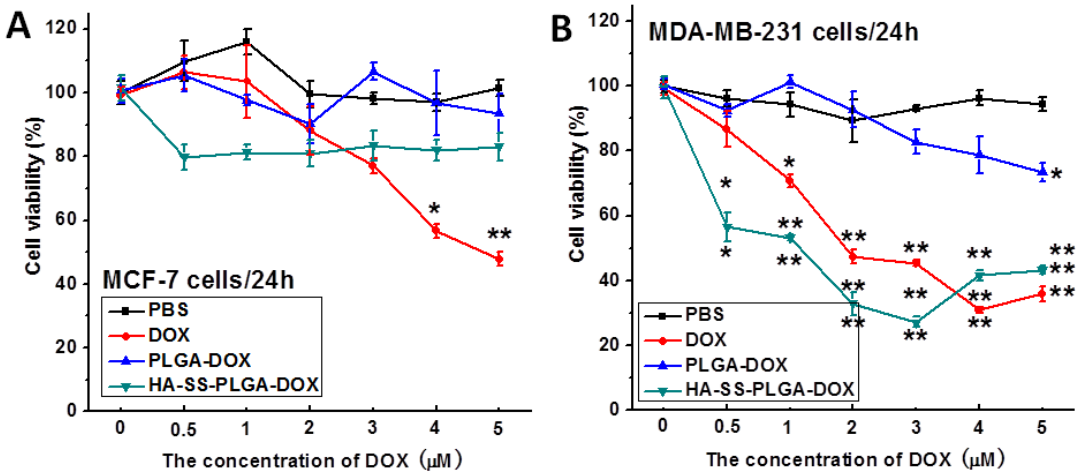

Figure S2. ***In vitro*** **cytotoxicity after 24h treatment.** The cytotoxicity of DOX, PLGA-DOX and HA-SS-PLGA-DOX towards breast cancer cells was evaluated by CCK-8 assay. (A) MCF-7 cells. (B) MDA-MB-231 cells. *$p < 0.05$ and **$p < 0.01$ compared to the PBS control group (one-way ANOVA, Tukey's post-hoc test).

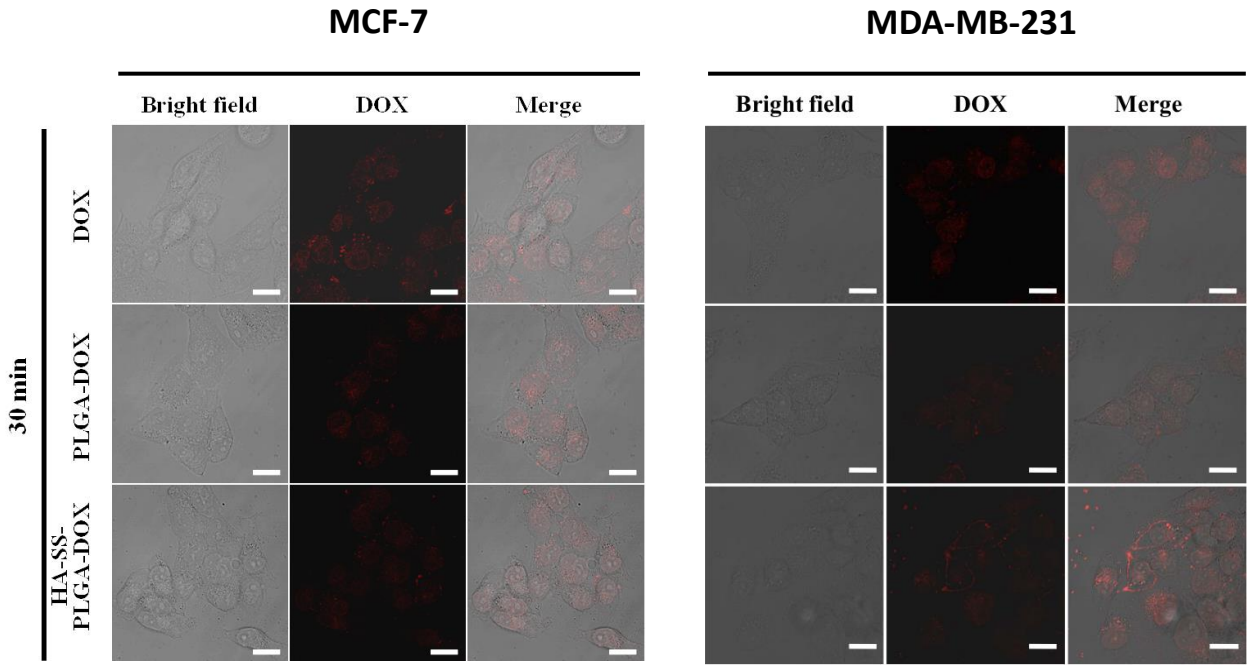

Figure S3. The cell uptake of particles by confocal microscopic study. The MCF-7 and MDA-MB-231 cells were treated with free DOX, PLGA-DOX, HA-SS-PLGA-DOX for 30 min. The red fluorescence of DOX was observed by a confocal microscope.

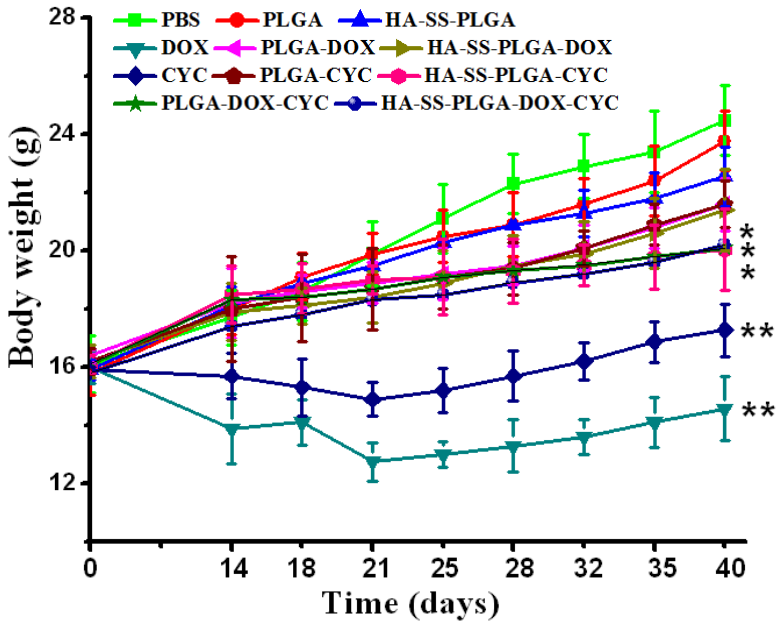

Figure S4. The body weights of mice. Data are expressed as mean ± SD. At the end of treatment, the average body weight between groups was compared by Student's t test. *$p < 0.05$ and **$p < 0.01$, vs. PBS group.

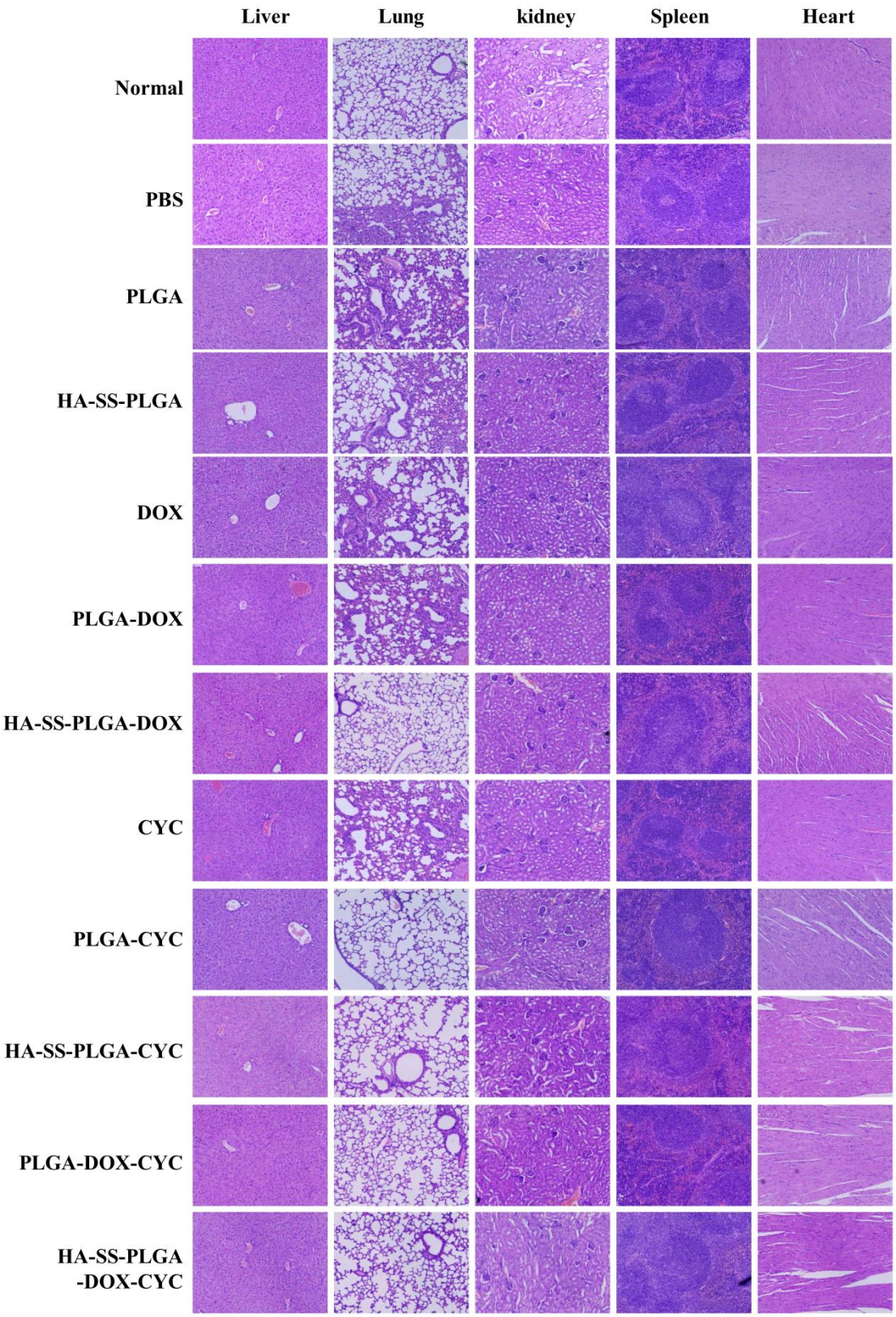

Figure S5. Histological examinations of liver, spleen, lung and kidney.

Table S1. The effects of formulation parameters on drug loading content and encapsulation efficiency

Sample number	PVA&F68 (W/V %)	Copolymer : DOX : CYC (mass ratio)	EE（%）of DOX&CYC	Size(nm) (By DLS)	PDI (By DLS)	Zeta Potential (mv)
1	0.5&0	50:1:2	65.3±2.1&43.3±2.1	327.4±3.2	0.116±0.016	-40.2±5.2
2	1&0	50:1:2	72.2±1.2&48.5±1.1	298.4±2.4	0.176±0.021	-42.1±4.5
3	2&0	50:1:2	76.1±1.9&66.0±1.4	270.6±4.5	0.181±0.006	-36.2±7.1
4	1&1	50:1:2	70.6±0.9&58.2±1.2	245.3±6.1	0.091±0.012	-38.5±2.3
5	1&1	100:1:2	80.3±0.6&71.2±2.4	264.2±2.1	0.114±0.042	-41.1±7.4
6	1&1	200:1:2	85.1±1.1&75.1±1.3	283.1±7.1	0.211±0.012	-32.8±4.7
7	1&1	50:0:2	----- & 68.5±2.5	223.1±4.3	0.164±0.024	-32.1±5.2
8	1&1	50:1:0	77.2±2.4 & ----	210.1±1.2	0.087±0.047	-34.5±2.4

Table S2. The body weights and coefficients of various tissues to body weight at the end of treatment

		Organ coefficients (mg/g)			
Group	**Body weight (g)**	**Liver**	**Spleen**	**Lung**	**Kidneys**
PBS	24.48 ± 1.20	71.88 ± 8.16	7.12 ± 0.97	7.11 ± 0.98	14.99 ± 4.09
PLGA	23.78 ± 1.02	72.12 ± 9.65	6.97 ± 0.63	7.09 ± 1.31	15.16 ± 5.11
HA-SS-PLGA	22.55 ± 1.01	71.09 ± 7.67	6.88 ± 0.59	7.12 ± 0.77	15.21 ± 3.98
DOX	14.57 ± 1.11**	74.88 ± 6.98	7.54 ± 0.33	7.32 ± 0.56	15.48 ± 3.12
PLGA-DOX	21.60 ± 0.94	69.65 ± 7.22	7.28 ± 1.28	6.99 ± 0.87	14.87 ± 4.61
HA-SS-PLGA-DOX	21.42 ± 1.45	71.32 ± 6.45	6.86 ± 0.73	7.10 ± 0.59	15.23 ± 5.32
CYC	17.28 ± 0.98**	73.34 ± 9.51	7.37 ± 1.14	7.24 ± 1.23	15.34 ± 5.43
PLGA-CYC	21.61 ± 0.87	70.11 ± 10.43	7.20 ± 0.63	6.89 ± 0.96	15.32 ± 3.46
HA-SS-PLGA-CYC	20.03 ± 1.41*	69.62 ± 6.47	7.04 ± 0.77	7.21 ± 1.72	15.21 ± 4.13
PLGA-DOX-CYC	20.10 ± 1.62*	73.12 ± 5.41	6.91 ± 0.83	7.17 ± 1.14	15.44 ± 4.32
HA-SS-PLGA-DOX-CYC	20.19 ± 1.08*	68.95 ± 8.34	7.10 ± 0.29	7.14 ± 1.21	15.35 ± 4.09

*** and ** represent significant difference compared to the control group, *$p < 0.05$ and **$p < 0.05$.**

Table S3. Complete blood counts

Group	WBC (10^9/L)	RBC (10^{12}/L)	HGB (g/L)	PLT (10^9/L)	NEUT (%)	LY (%)	MONO (%)
Normal	3.22 ± 0.28	10.61 ± 1.31	157.76 ± 9.74	894.34 ± 52.11	1.28 ± 0.07	85.47 ± 7.26	3.51 ± 0.99
PBS	4.23 ± 0.54	11.02 ± 2.21	160.65 ± 9.28	842.98 ± 49.67	2.08 ± 0.03	90.52 ± 7.84	3.90 ± 1.11
PLGA	3.97 ± 0.26	9.84 ± 1.45	153.97 ± 6.18	910.75 ± 57.31	1.91 ± 0.11	90.10 ± 8.95	3.55 ± 0.65
HA-SS-PLGA	3.84 ± 0.61	10.06 ± 2.07	151.98 ± 7.09	891.56 ± 46.78	1.57 ± 0.31	91.23 ± 8.31	4.07 ± 1.23
DOX	3.52 ± 0.76	9.88 ± 1.98	151.36 ± 9.77	735.57 ± 32.46*	1.51 ± 0.14	92.10 ± 9.11*	1.81 ± 0.33*
PLGA-DOX	3.47 ± 0.76	9.99 ± 2.31	149.53 ± 10.16	791.09 ± 41.98	2.11 ± 0.24	91.53 ± 7.27	3.60 ± 1.63
HA-SS-PLGA-DOX	3.31 ± 0.57	9.96 ± 1.78	158.09 ± 10.31	906.22 ± 54.13	1.65 ± 0.41	90.06 ± 8.03	3.78 ± 0.36
CYC	3.23 ± 0.56	10.11 ± 1.69	151.57 ± 7.08	769.53 ± 65.32*	1.83 ± 0.65	90.43 ± 7.22	1.27 ± 0.87*
PLGA-CYC	3.07 ± 0.41	9.90 ± 2.17	152.75 ± 9.87	896.11 ± 45.78	1.78 ± 0.97	88.95 ± 8.05	4.08 ± 0.07
HA-SS-PLGA-CYC	3.49 ± 0.77	10.03 ± 1.99	159.31 ± 10.22	900.56 ± 51.43	2.09 ± 0.19	91.32 ± 6.45	3.82 ± 0.95
PLGA-DOX-CYC	3.74 ± 0.24	9.81 ± 2.06	152.87 ± 9.34	861.75 ± 53.13	1.93 ± 0.31	88.97 ± 3.96	3.64 ± 1.02
HA-SS-PLGA-DOX-CYC	3.44 ± 0.64	9.95 ± 1.98	156.43 ± 10.54	891.09 ± 45.65	2.07 ± 0.54	92.43 ± 7.54	4.33 ± 1.65

*** represents significant difference compared to the normal control group, *$p < 0.05$.**

Table S4. Blood biochemical analyses

Group	BUN (mmol/L)	CR (mmol/L)	ALT(U/L)	AST(U/L)	LDH(U/L)	CK(U/L)
Normal	11.71 ± 1.23	12.09 ± 2.13	60.77 ± 9.88	127.38 ± 10.94	597.65 ± 77.35	673.77 ± 54.12
PBS	12.22 ± 0.97	11.34 ± 1.76	58.54 ± 10.05	166.19 ± 9.65*	1943.09 ± 324.98**	1270.52 ± 34.87**
PLGA	11.93 ± 2.43	11.79 ± 1.98	53.77 ± 7.64	153.87 ± 8.21	991.26 ± 76.56**	1025.43 ± 85.12**
HA-SS-PLGA	11.21 ± 2.08	10.87 ± 2.01	58.34 ± 6.15	131.44 ± 6.94	1064.34 ± 127.43**	877.45 ± 43.09*
DOX	7.65 ± 1.94*	7.49 ± 1.46*	43.67 ± 6.23*	137.81 ± 5.09	1144.58 ± 109.87**	687.45 ± 34.21
PLGA-DOX	7.89 ± 0.69*	8.03 ± 2.41*	44.81 ± 9.76*	132.09 ± 6.83	721.73 ± 43.71*	598.35 ± 19.65
HA-SS-PLGA-DOX	10.95 ± 2.06	11.12 ± 1.97	62.54 ± 5.16	129.65 ± 7.87	607.67 ± 10.76	631.56 ± 26.77
CYC	7.18 ± 1.94*	6.62 ± 0.67*	50.31 ± 4.76*	133.45 ± 6.15	599.98 ± 12.78	909.54 ± 43.67*
PLGA-CYC	10.23 ± 1.87	7.26 ± 2.01*	56.43 ± 3.95	130.26 ± 4.22	578.53 ± 63.84	694.39 ± 32.08
HA-SS-PLGA-CYC	11.67 ± 1.94	11.89 ± 1.09	58.93 ± 7.25	129.27 ± 8.41	626.87 ± 51.65	701.65 ± 31.93
PLGA-DOX-CYC	10.99 ± 0.52	13.17 ± 3.07	68.42 ± 6.22	134.12 ± 7.87	641.04 ± 91.82	688.63 ± 46.01
HA-SS-PLGA-DOX-CYC	11.06 ± 2.11	11.98 ± 2.07	63.74 ± 5.24	132.76 ± 8.43	609.45 ± 87.31	704.87 ± 54.22

*** and ** represent significant difference compared to the normal control group, *$p < 0.05$ and **$p < 0.05$.**