Supporting Information for:

Self-assembled Nanodiamond Supraparticles for Anticancer Chemotherapy

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Supporting Figures

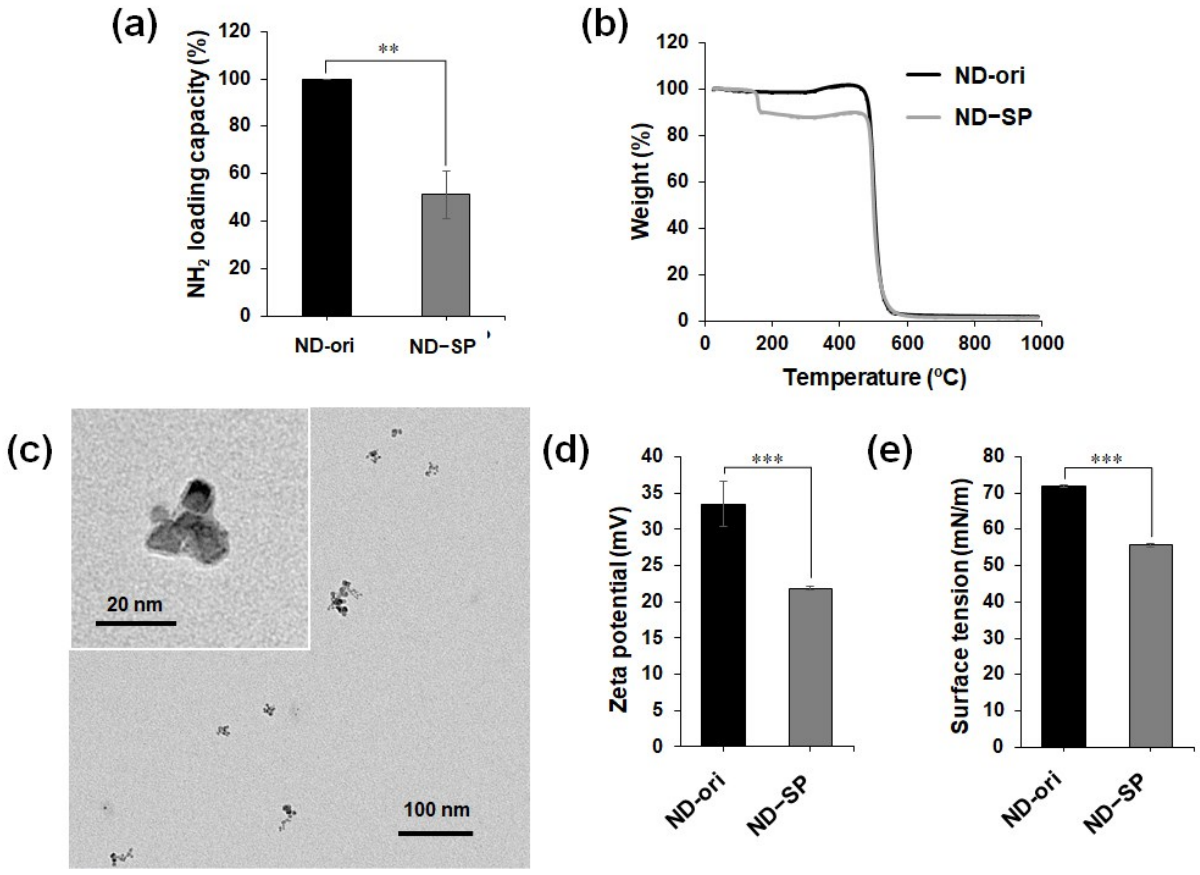

Figure S1. Synthesis and characterization of ND−SPs. a) Comparison of NH_2 loading capacity between ND-ori and ND−SPs. ND-ori was set as 100 (n = 3). b) TGA of ND-ori and ND-SPs. c) Representative TEM image of ND-ori. d) Zeta potential of ND-ori and ND−SPs. Sample concentration: 3.2 mg ml^{-1} (n = 3). e) Surface tension of ND-ori and ND−SP at 25°C. Samples concentration: 3.2 mg ml^{-1} (n = 3).

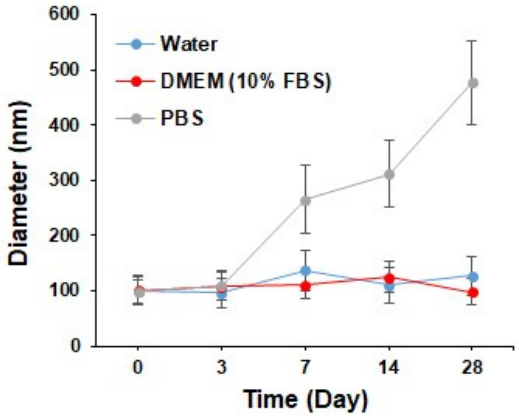

Figure S2. Colloid stability test of ND−SPs in different media for 28 days by DLS.

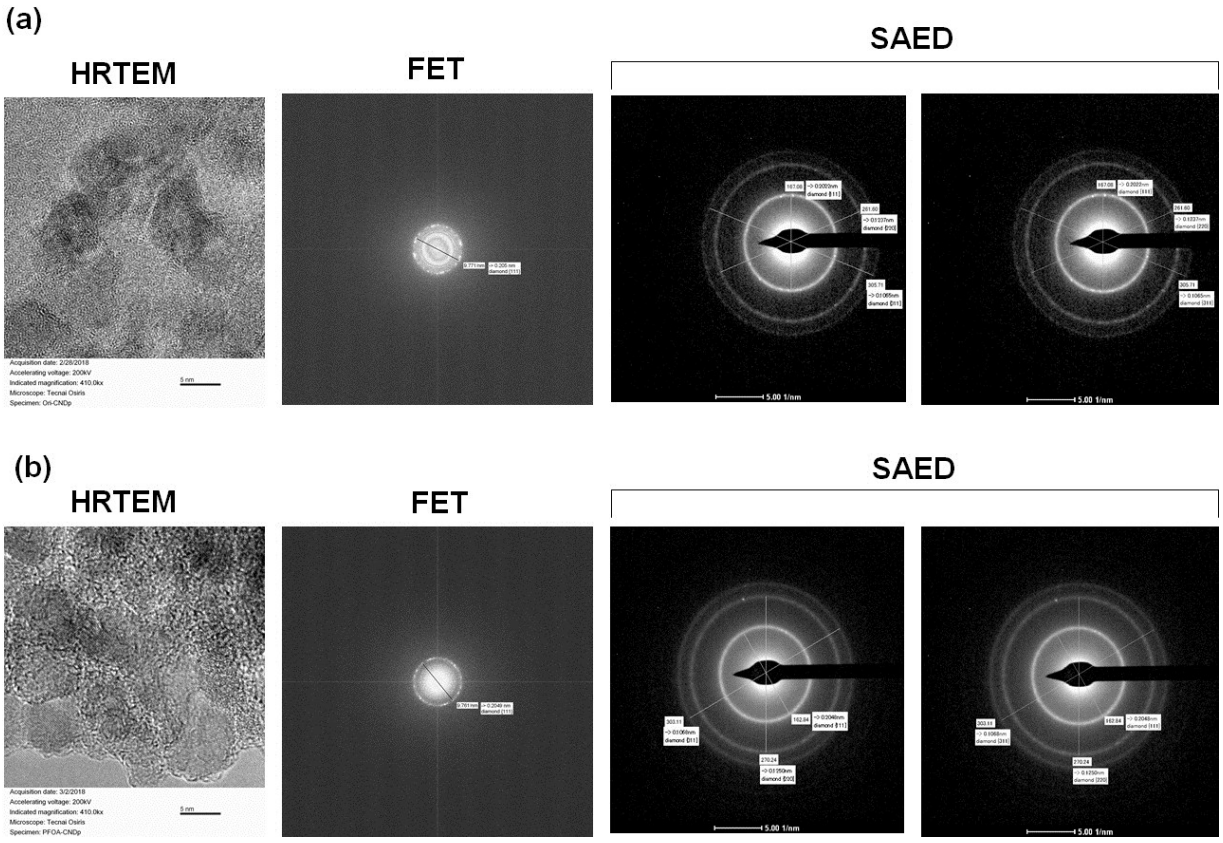

Figure S3. Characterization of crystal structures of a) ND-ori and b) ND−SP by HRTEM, field-effect transistor (FET) analyzing and selected area (electron) diffraction observations.

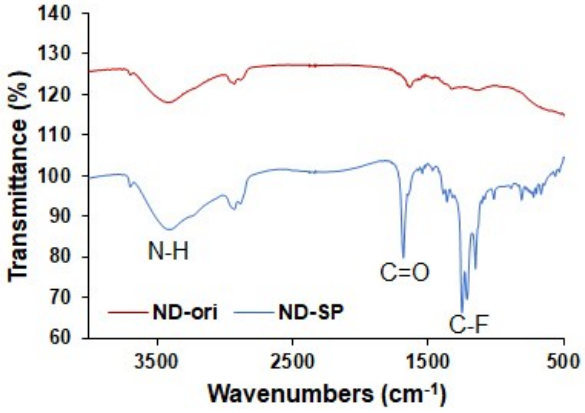

Figure S4. FTIR spectra of ND-ori and ND−SP.

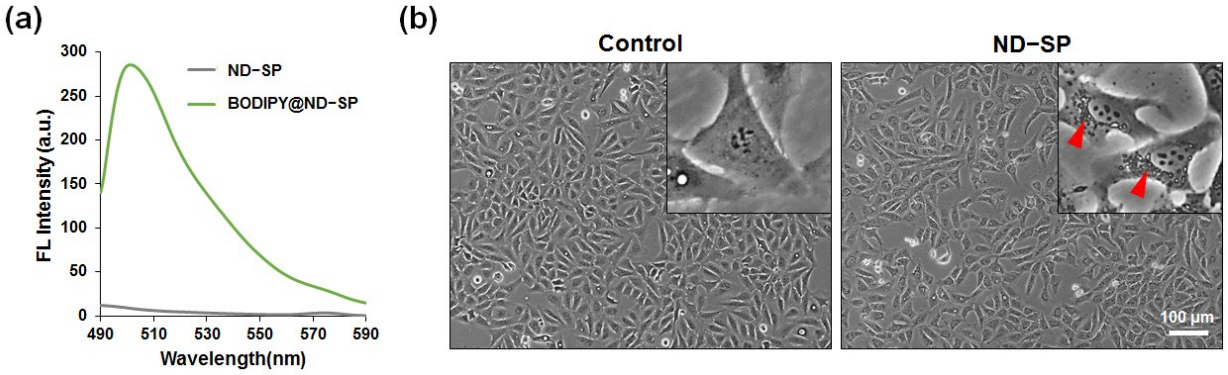

Figure S5. Internalization and subcellular distribution of ND−SPs. a) Fluorescence (FL) intensity of ND-SPs before (gray curve) and after (green curve) BOBIPY incorporation. b) Phase contrast images of U2OS cells incubated with (right) or without (left) ND−SPs (32 μg ml^{-1}) for 12 h. Red arrows in the high magnification image indicate ND−SP aggregates in the perinuclear area.

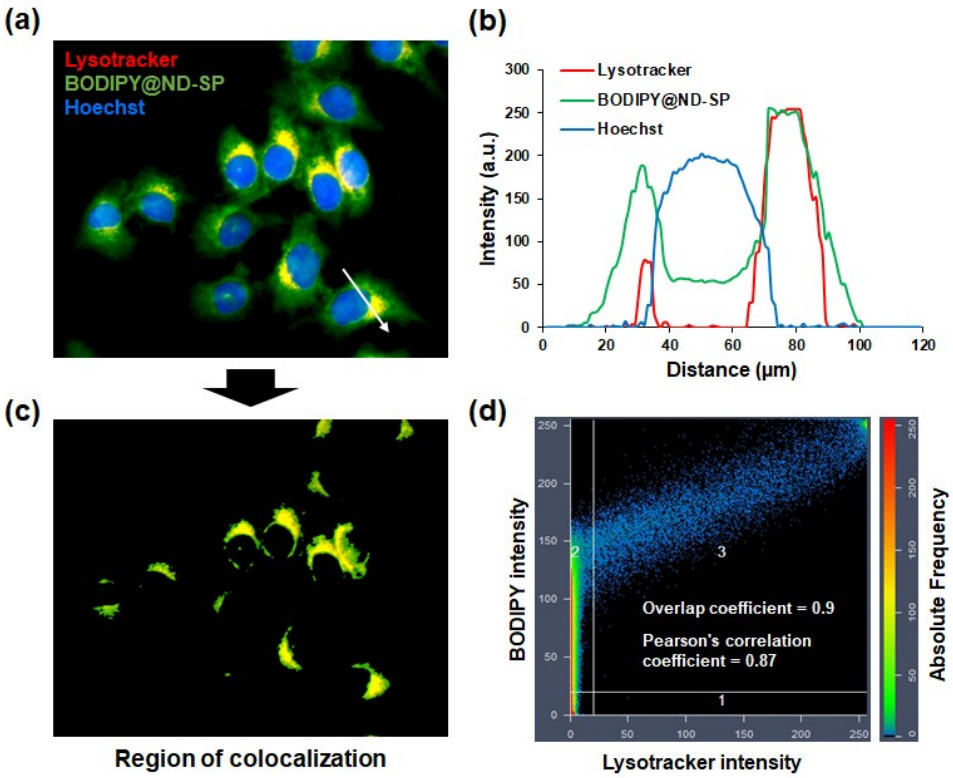

Figure S6. a) Fluorescence merged image of HeLa cells (corresponding to Figure 2d). White arrow represents the location for characterization of fluorescence intensity profile. b) Fluorescence intensity profile across the white arrowed line in Figure S2a. c) Colocalization region of Lysotracker and BODIPY@ND−SP. d) Scatter plots of fluorescence intensities of green (BODIPY@ND−SP) and red (lysotracker) by construct.

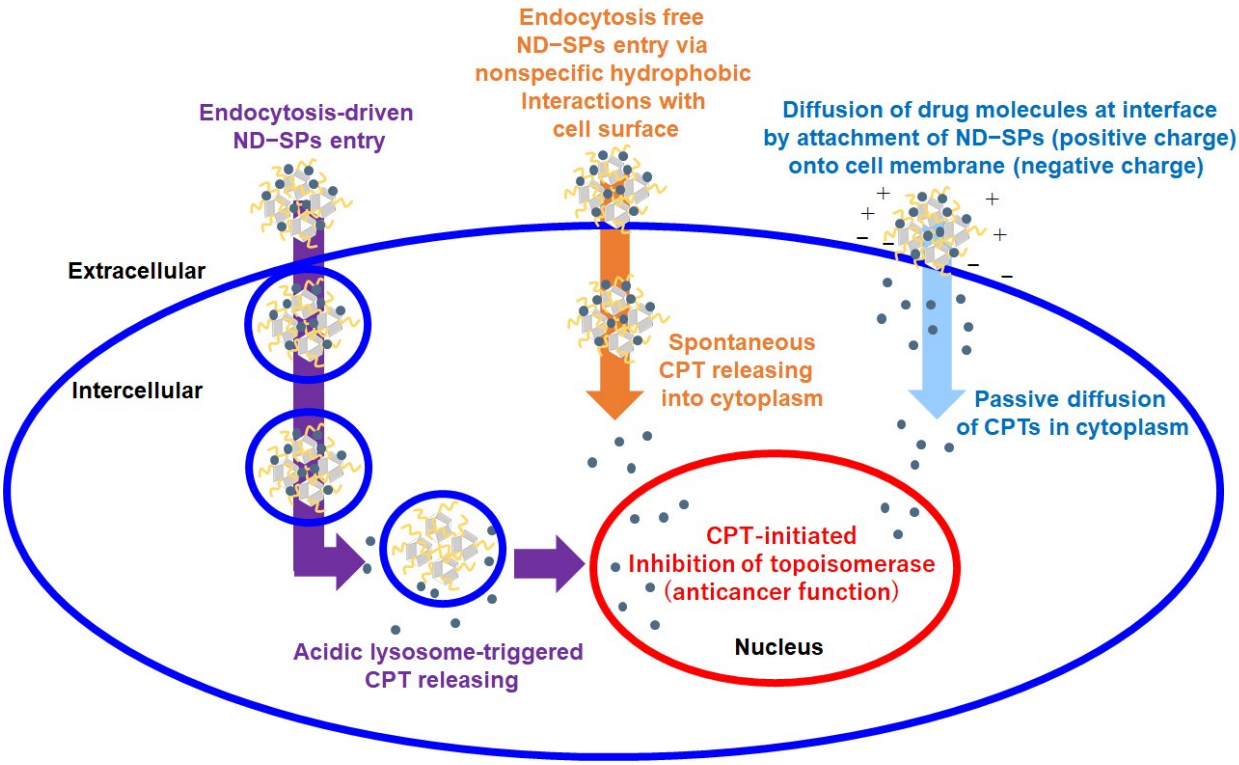

Figure S7. Schematic illustration of potential pathways of ND−SPs internalized by cancer cells.

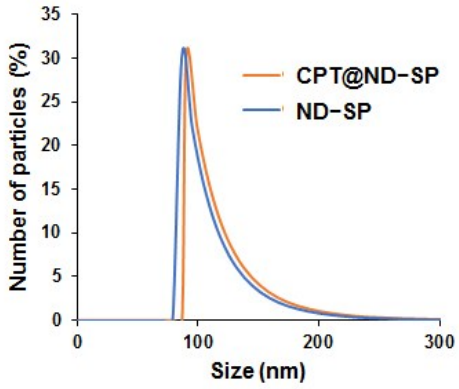

Figure S8. DLS analyses of ND−SP and CPT@ND−SP size distributions.

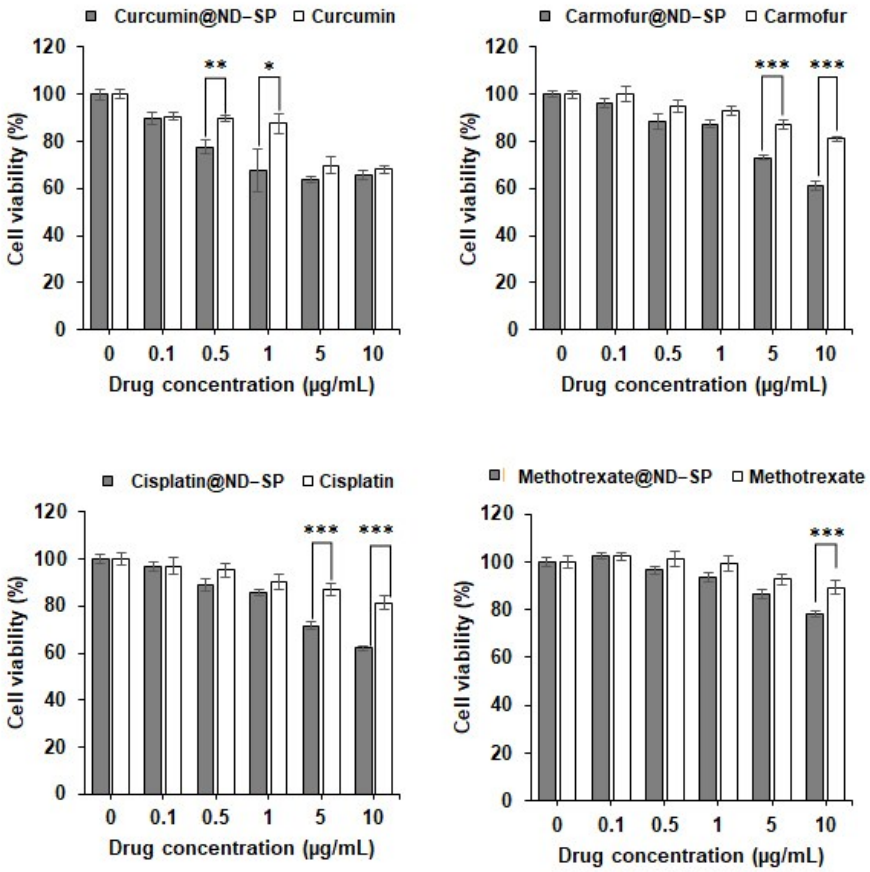

Figure S9. *In vitro* anticancer effect of ND−SPs as a drug delivery carrier. Cytotoxicity evaluation of ND−SP with or without incorporation of various anticancer drugs. U2OS cell viability was tested after 24 h of treatment.

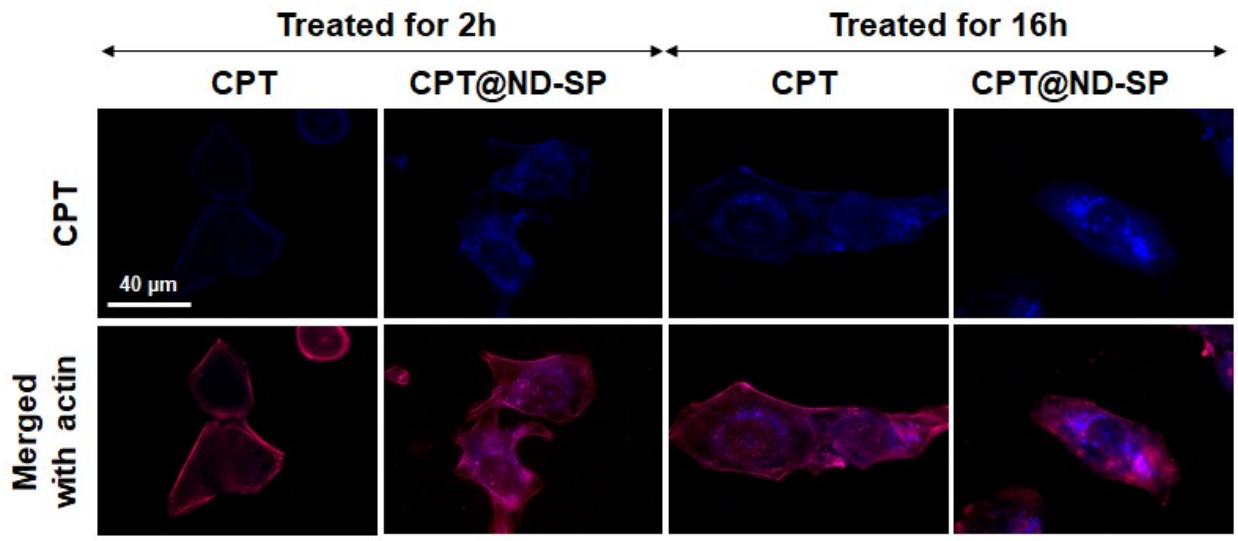

Figure S10. Cellular uptake of free CPT and CPT@ND−SP. U2OS cells were treated with an equivalent dose of CPT (20 μg ml^{-1}, blue color) for 2 and 16 h. Cytoskeleton (actin) was stained with phallodin (red color) to indicate presence of cells.

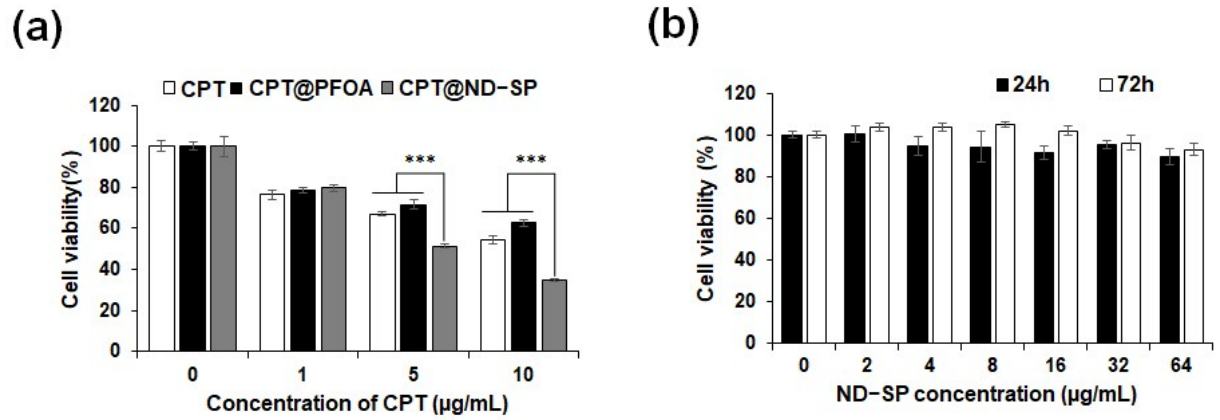

Figure S11. *In vitro* anticancer effect of ND−SPs as a drug delivery carrier. a) Viability of U2OS cells treated with CPT, CPT@PFOA, and CPT@ND−SPs for 24 h. b) Cytotoxicity evaluation of ND−SP in TIG-3 human normal fibroblast cells.

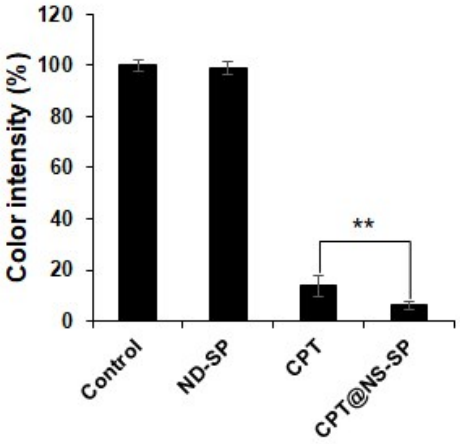

Figure S12. Quantitation of crystal violet staining of U2OS cells with or without ND−SPs (32 μg ml^{-1}), CPT (10 μg ml^{-1}), or CPT@ND−SP (CPT, 10 μg ml^{-1}; ND, 32 μg ml^{-1}) treatment for 48 h.

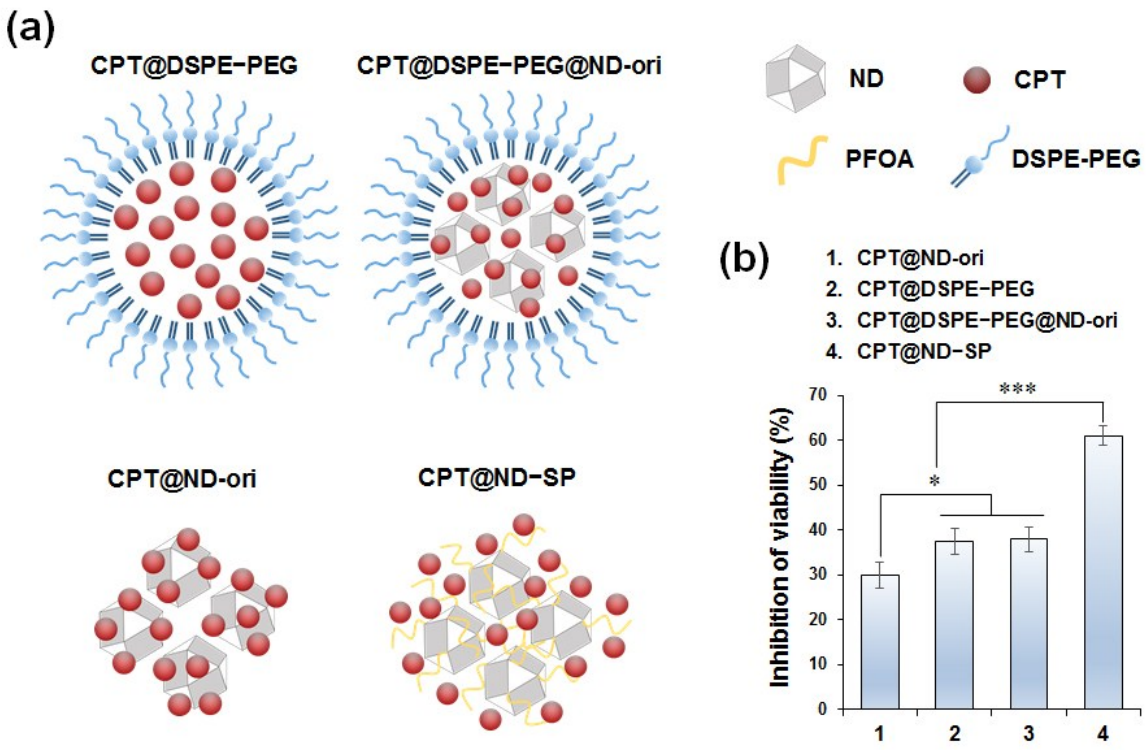

Figure S13. *In vitro* anticancer effect of ND−SPs as a drug delivery carrier. a) Schematic illustration of CPT-loaded DSPE-PEG phospholipids, ND-ori, and ND−SP nanoclusters. b) Inhibitory effect of nanocomplexes shown in a) on cell viability. U2OS cancer cells were treated with the nanocomplexes for 24 h or with an equivalent dose of CPT (10 μg ml^{-1}).

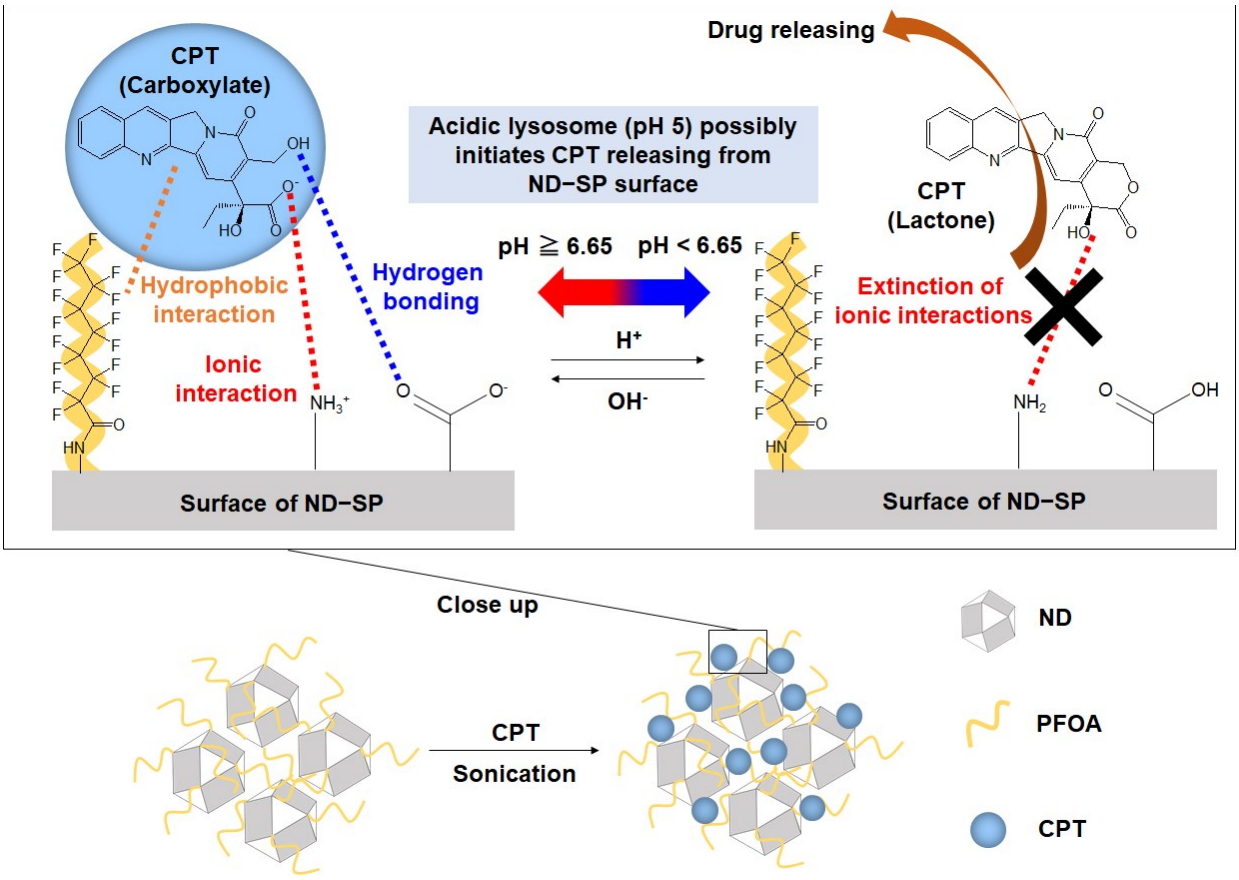

Figure S14. Schematic illustration of interactions between CPT and the surface of ND−SP, and the potential releasing mechanism of CPT molecules from ND−SPs by pH changing.

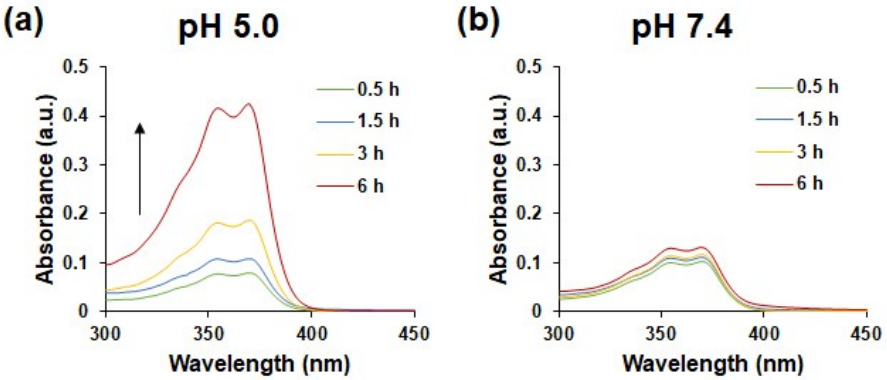

Figure S15. UV−vis absorption spectra of released CPT from ND−SPs in PBS at pH 5.0 (a) and 7.4 (b).

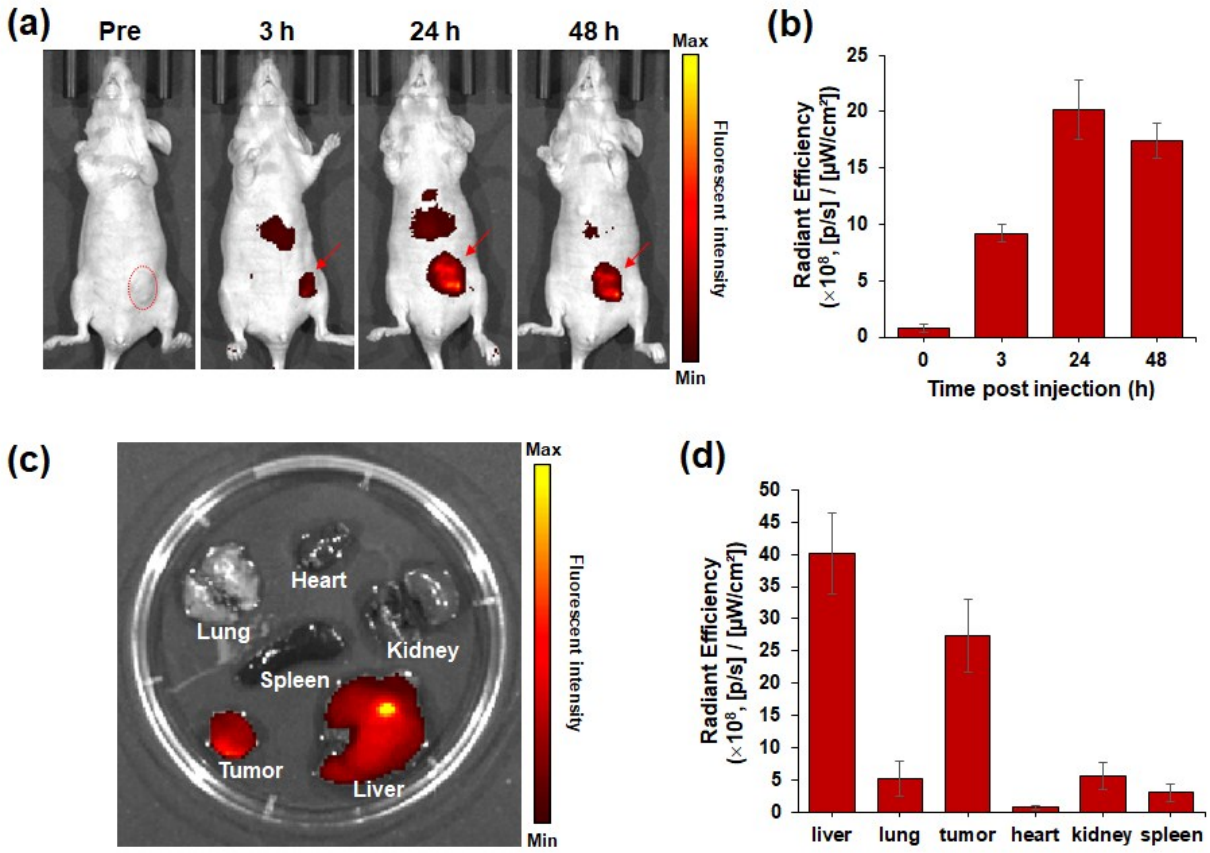

Figure S16. Biological distribution of ND−SPs. (a) Fluorescence imaging of HT-29 tumor-bearing mice after intravenous injection of ND-SPs (3 mg ml^{-1}, 200 μl). Red dashed circle represents the location of a solid tumor. Red arrows display the location of ND-SPs where they are targeted in tumor. (b) Corresponding radiant efficiency of tumor at different time post injection. (c) *Ex vivo* image and quantitative radiant efficiency (d) of tumor and major organs at 24h post injection.

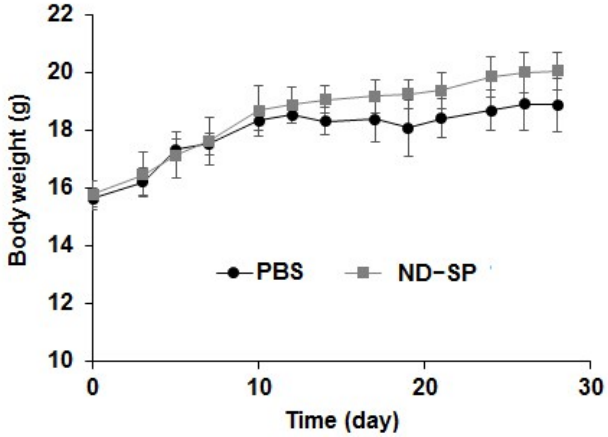

Figure S17. Average body weight of mice orally administered ND−SP (35.6 mg kg^{-1}) or PBS (vehicle control).

Table S1. CBCs and biochemical parameters of mice injected with ND−SP or PBS for 28 days

Measured value	Entry	Unit	PBS (n = 5)	ND−SP (n = 5)	P value
CBC	WBC	$\times 10^2\ \mu l^{-1}$	91 ± 21	89 ± 14	> 0.05
	RBC	$\times 10^4\ \mu l^{-1}$	978 ± 31	967 ± 20	> 0.05
	PLT	$\times 10^4\ \mu l^{-1}$	70 ± 4	71 ± 2	> 0.05
Biochemical parameters	CRP	$\mu g\ ml^{-1}$	1.6 ± 0.1	1.6 ± 0.2	> 0.05
	TP	$g\ dl^{-1}$	4.2 ± 0.2	4.1 ± 0.1	> 0.05
	ALB	$g\ dl^{-1}$	2.8 ± 0.1	2.6 ± 0.1	> 0.05
	BUN	$mg\ dl^{-1}$	15 ± 1	16 ± 1	> 0.05
	CRE	$mg\ dl^{-1}$	0.13 ± 0.01	0.13 ± 0.01	> 0.05
	AST	$IU\ l^{-1}$	45 ± 6	52 ± 4	> 0.05
	LDH	$IU\ l^{-1}$	183 ± 28	221 ± 55	> 0.05
	AMY	$IU\ l^{-1}$	1778 ± 207	1661 ± 128	> 0.05
	CK	$IU\ l^{-1}$	218 ± 98	233 ± 55	> 0.05

Results represent mean ± standard deviation of five experiments. Statistical analyses were performed with the Student's t test.

Abbreviations: ALB, albumin; AMY, amylase; AST, aspartate aminotransferase; BUN, blood urea nitrogen; CK, creatine kinase; CRE, creatinine; CRP, C-reactive protein; LDH, lactate dehydrogenase; PLT, platelet; RBC, red blood cell; TP, total protein; WBC, white blood cell.