

Supporting Information: Pheophorbide A and Paclitaxel biore-sponsive nanoparticles as double-punch platform for cancer therapy

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Table S1. Optimization studies on PheoA@PTX₂S nanoparticles using different PheoA/PTX₂S ratio and different concentrations of PheoA in DMSO. 2.

[PTX ₂ S] (mg/mL _{DMSO})	[PheoA] (mg/mL _{DMSO})	% PheoA (W _{PheoA} /W _{PTX₂S})	Hydrodynamic Diameter (nm)	PDI
10	0.5	15	147 ± 1	0.17 ± 0.02
		20	157 ± 1	0.14 ± 0.01
		25	142 ± 6	0.15 ± 0.01
	1.5	30	61 ± 2	0.12 ± 0.03
		35	93 ± 3	0.11 ± 0.01
		40	78 ± 2	0.10 ± 0.02

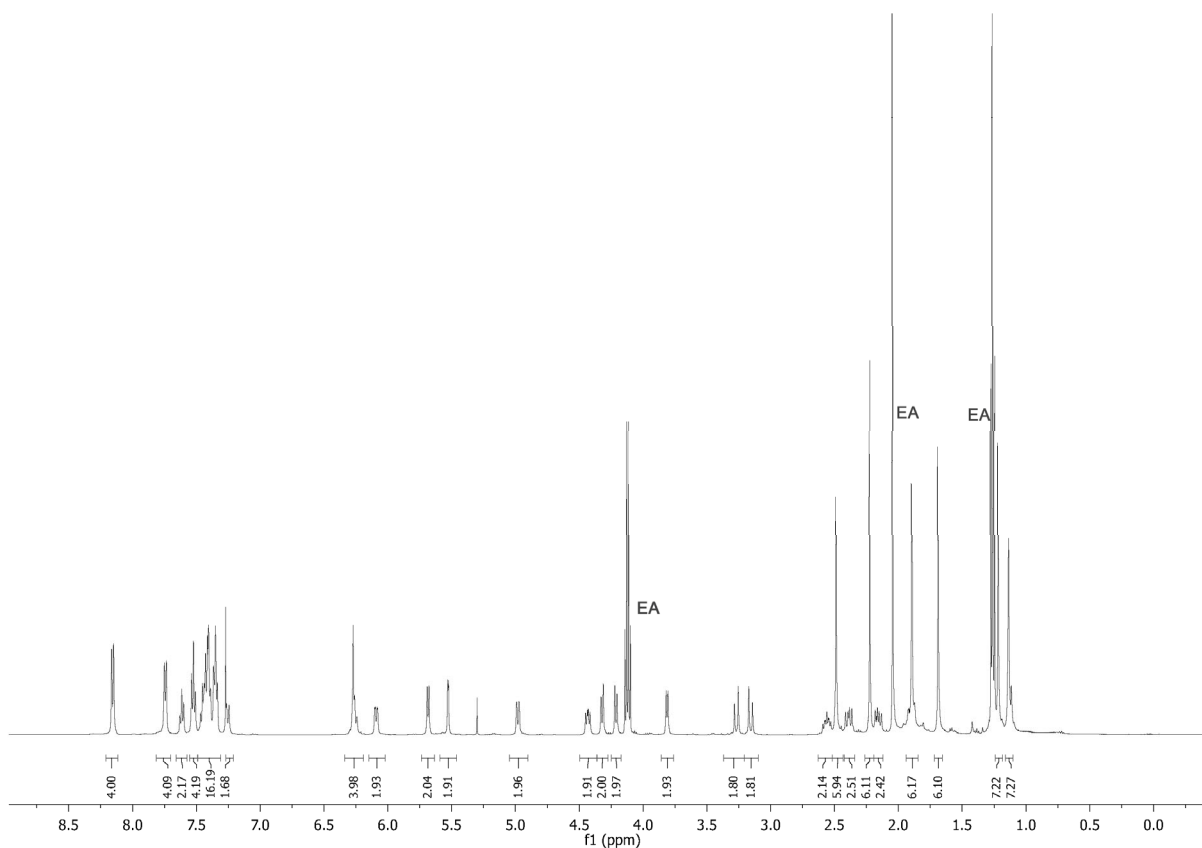


Figure S1. ¹H-NMR of PTX₂S in CDCl₃ recorded on 500 MHz Varian spectrometer. 2.

Figure S2. PheoA@PTX₂S nanoparticles disassembly studies under different conditions: **a)** in the presence of 10 mM GSH at 37 °C for 24 h and **b)** in the presence of different H₂O₂ concentrations and plus light irradiation. 3.

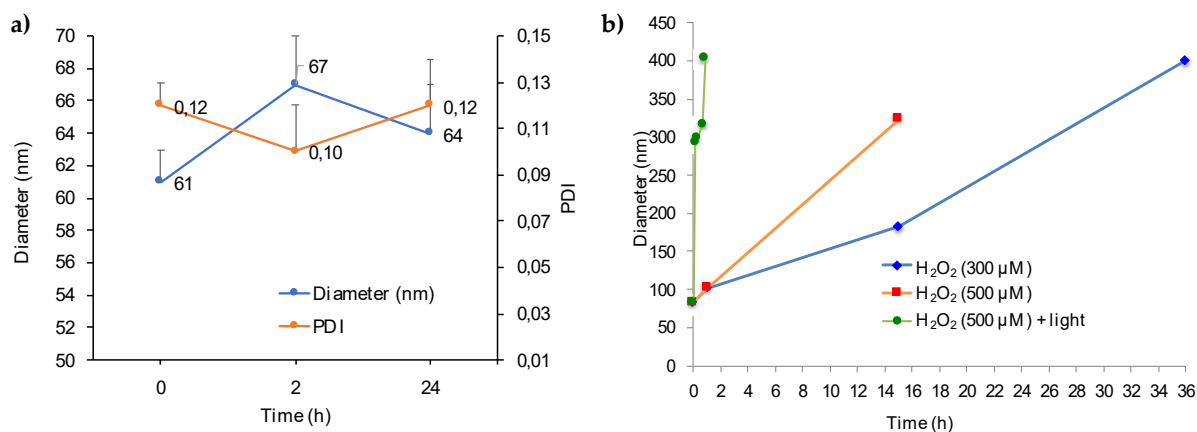


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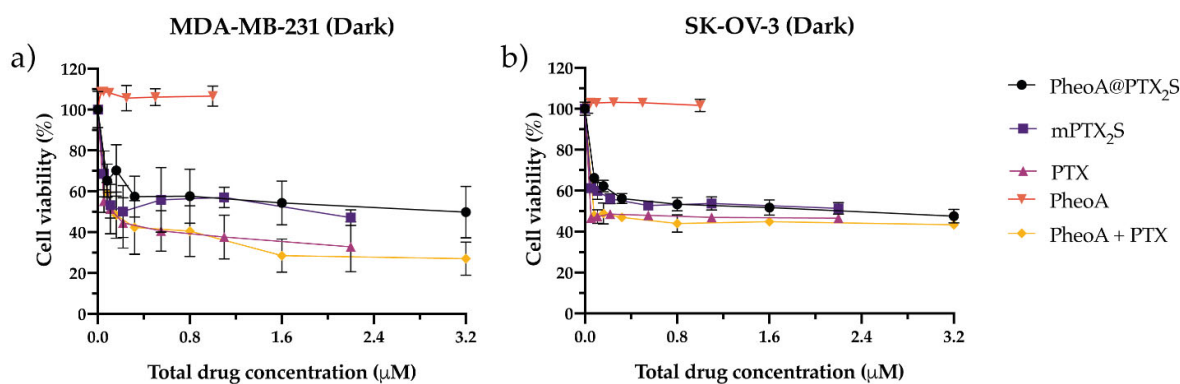


Figure S3. Dark cytotoxicity in vitro. Dose-response curves of MDA-MB-231 **a)** or SK-OV-3 **b)** cells incubated in the dark for 24 h with single drugs or their combination delivered free or in nanoparticles and released for additional 24 h in drug-free medium before cell viability measurement with MTS assay. Total drug concentration is referred to PTX + PheoA concentration. Data are expressed as mean percentage \pm SD of at least three independent experiments, carried out in triplicate.

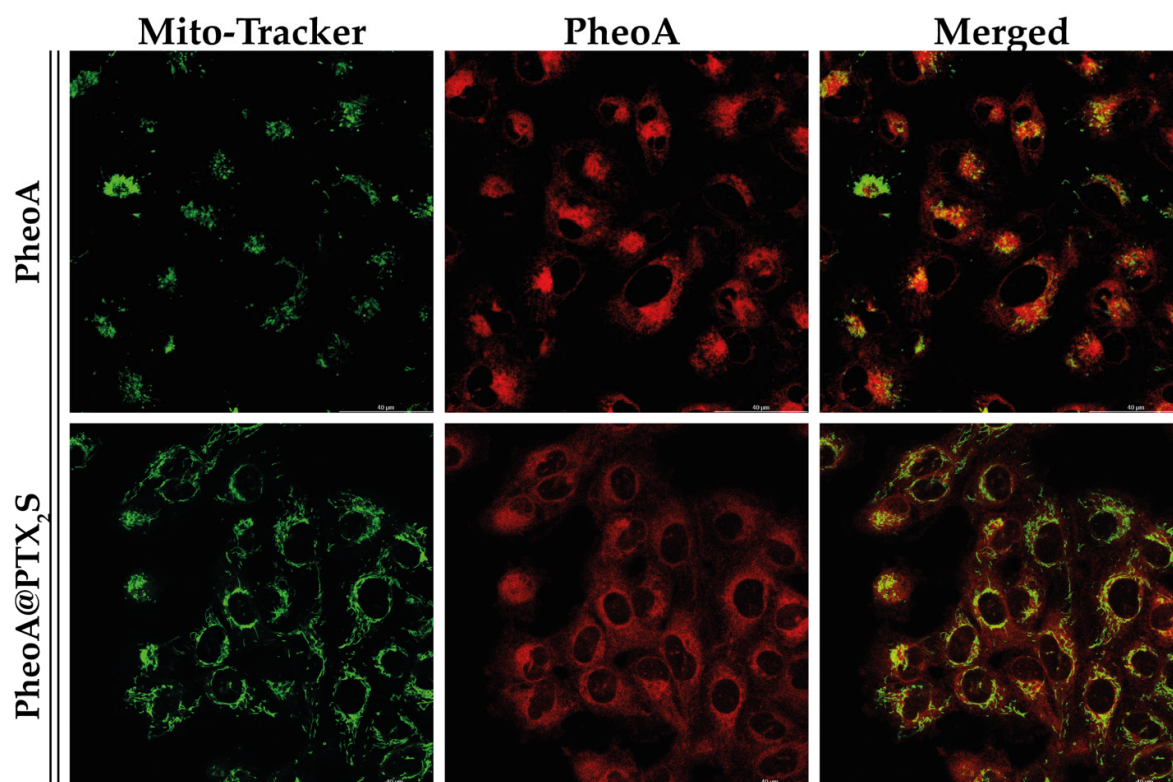


Figure S4. In vitro intracellular localization studies. Confocal microscopy images of MDA-MB-231 cells showing only slight co-localization between the red fluorescence of PheoA (delivered in the standard solvent or loaded in PheoA@PTX₂S) and the green fluorescence of MitoTracker used as specific probe for mitochondria. Scale bars: 40 μ m.

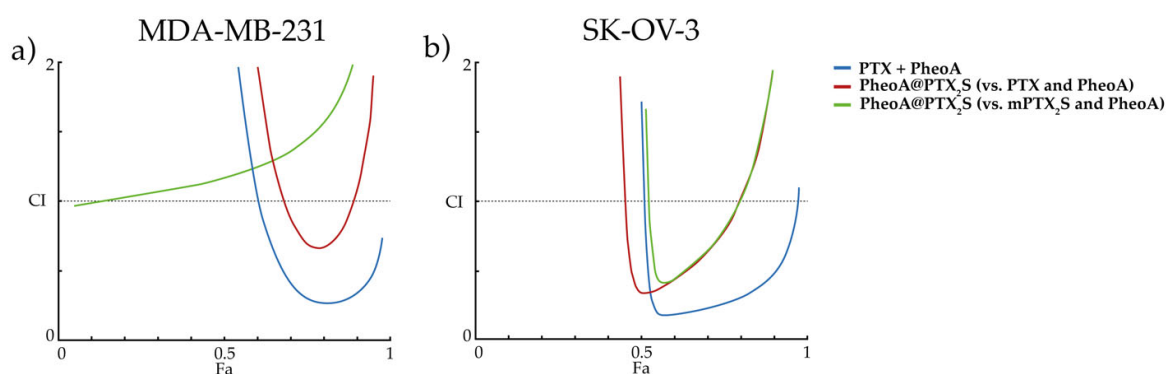


Figure S5. Combination index analysis. Plots of Combination Index vs. Fa (Fa-CI plot) relative to MDA-MB-231 **a)** and SK-OV-3 **b)** cells treated with the combination of PTX and PheoA. Data reported in Fig. 5 of the main text were analyzed with the Compusyn software and Fa-CI plots derived.