Supporting Information

Drug-Triggered Self-Assembly of Linear Polymer into Nanoparticles for Simultaneous Delivery of Hydrophobic and Hydrophilic Drugs in Breast Cancer Cells

Sandeep Palvai, [†,#] Libi Anandi, [ψ,#] Sujit Sarkar,[ξ] Meera Augustus,[ψ] Sudip Roy,[ξ] Mayurika Lahiri, [ψ*] Sudipta Basu[†*]

† Department of Chemistry, Indian Institute of Science Education and Research (IISER)-Pune, Dr. Homi Bhabha Road, Pashan, Pune, Maharashtra, India, 411008. E-mail: sudipta.basu@iiserpune.ac.in

Ψ Department of Biology, Indian Institute of Science Education and Research (IISER)-Pune, Dr. Homi Bhabha Road, Pashan, Pune, Maharashtra, India, 411008. Email: mayurika.lahiri@iiserpune.ac.in

ξ CSIR-National Chemical Laboratory, Pune, Dr. Homi Bhabha Road, Pashan, Pune, Maharashtra, India, 411008

 # These authors contributed equally to this work.
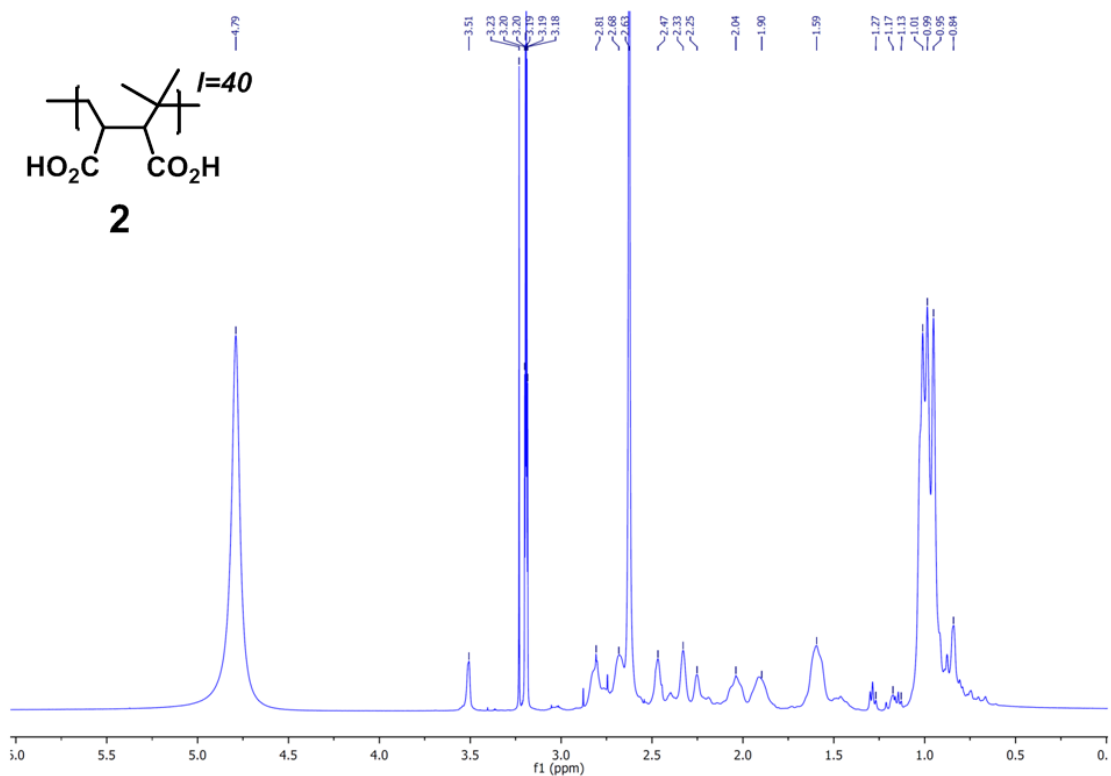
Content:

Figure S1: ^{1}H NMR spectra of Poly(isobutylene-alt-maleic acid) (PMAc) **2**.
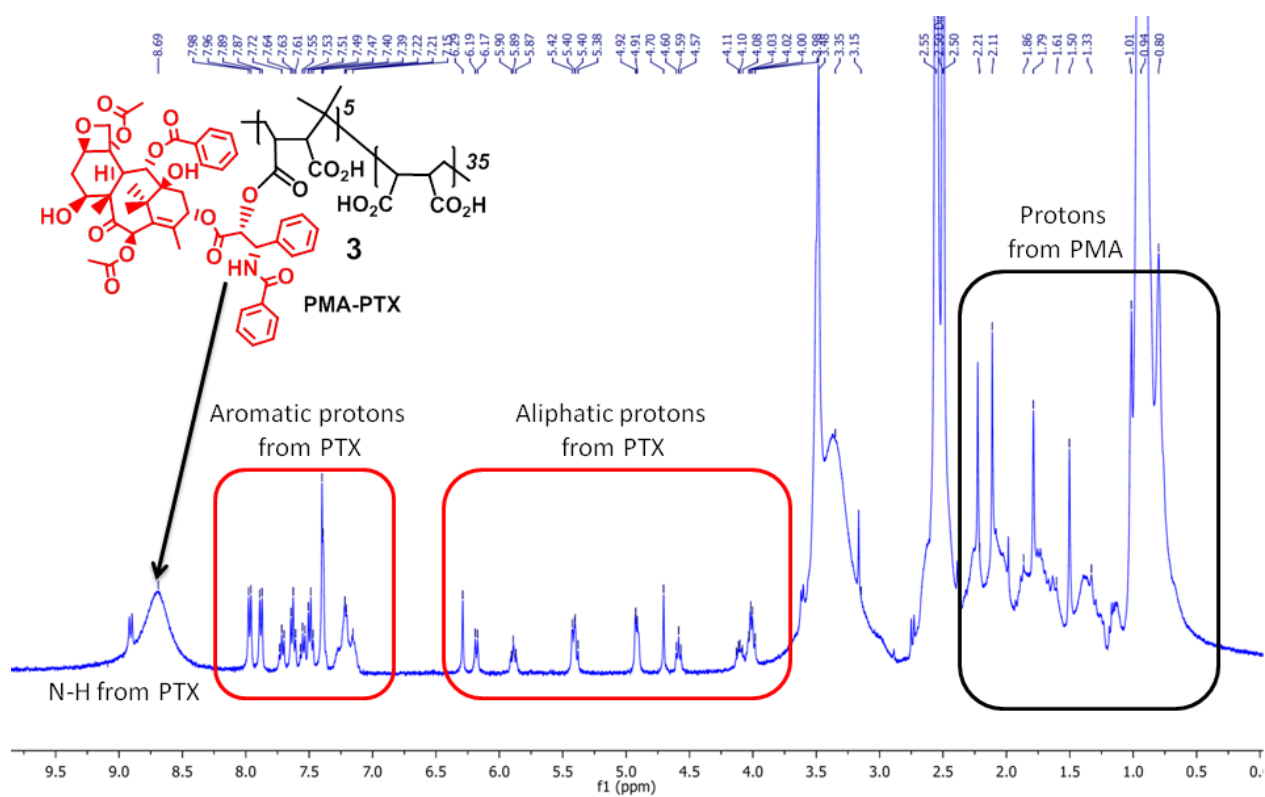

Figure S2: ^{1}H NMR spectra of PMA-PTX conjugate **3.**
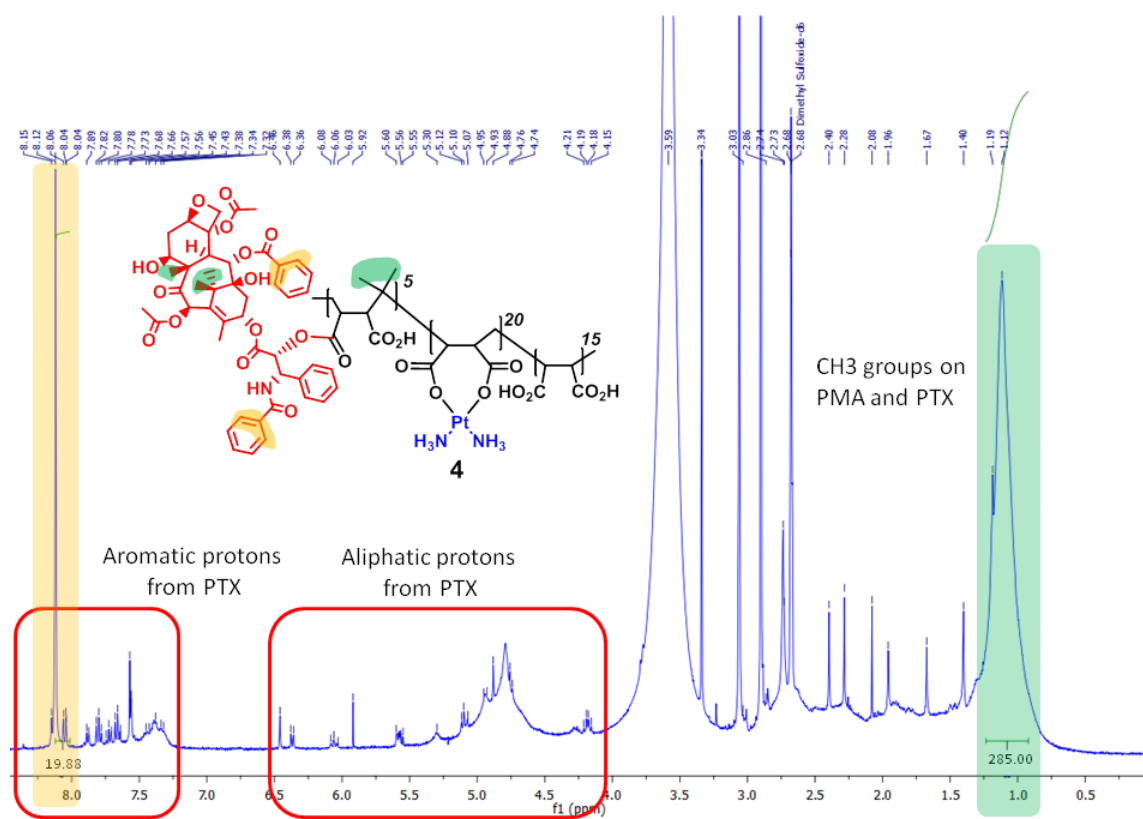
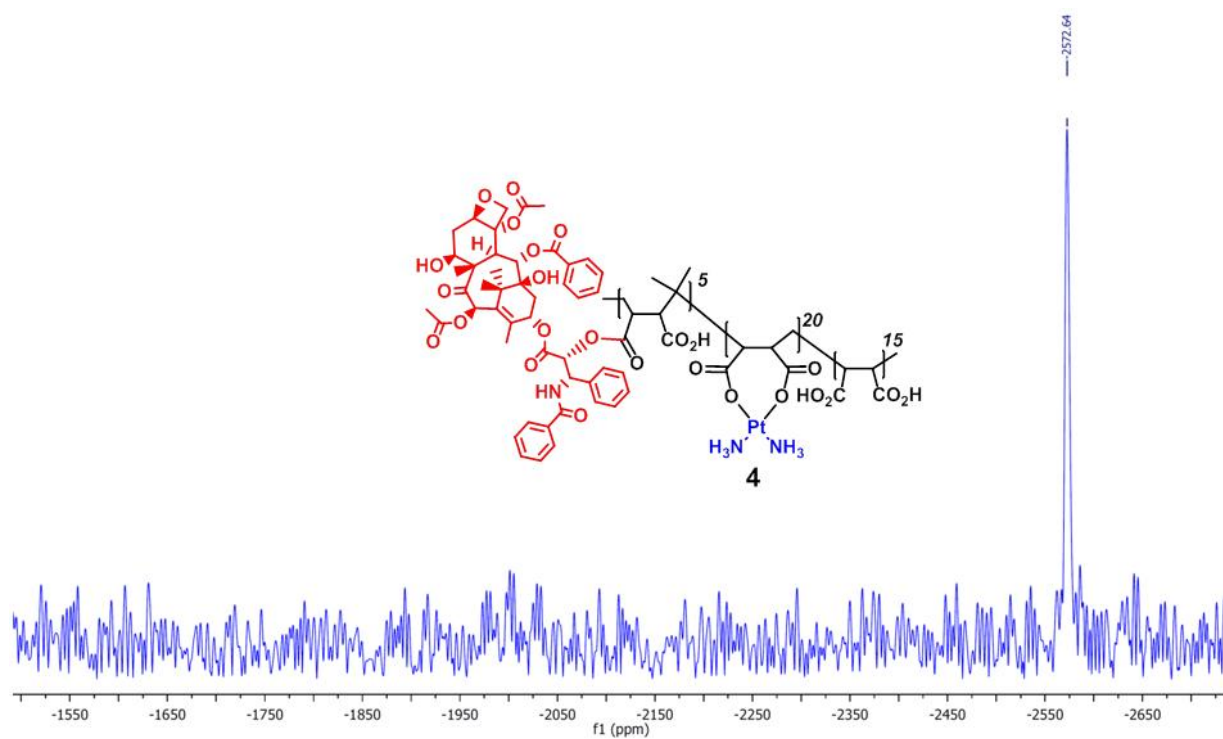**Figure S3:** ^{1}H NMR spectra of PMA-PTX-CDDP conjugate (**4**).

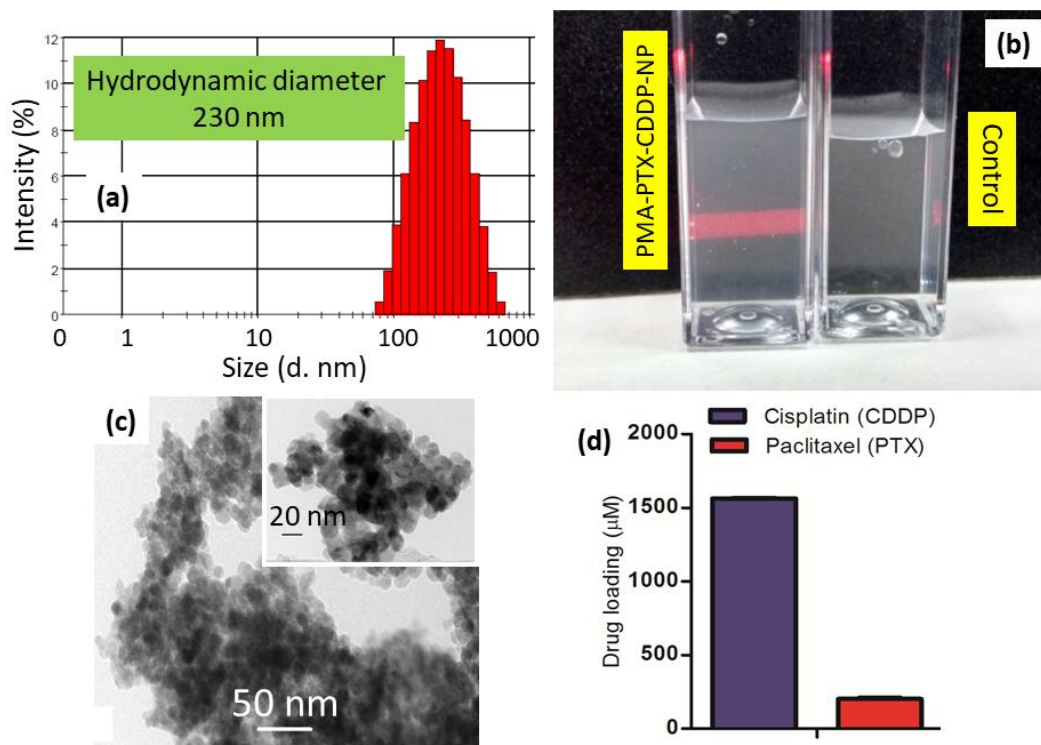**Figure S4**: [195]Pt NMR spectra of PMA-PTX-CDDP conjugate (**4**).

Figure S5: (a) Hydrodynamic diameter measurement of PMA-PTX-CDDP-NPs by dynamic light scattering (DLS). (b) Tyndall effect of PMA-PTX-CDDP-NPs. (c) TEM images of PMA-PTX-CDDP-NPs. (d) Loading of PTX and CDDP into the nanoparticle, determined by UV-Vis spectroscopy.

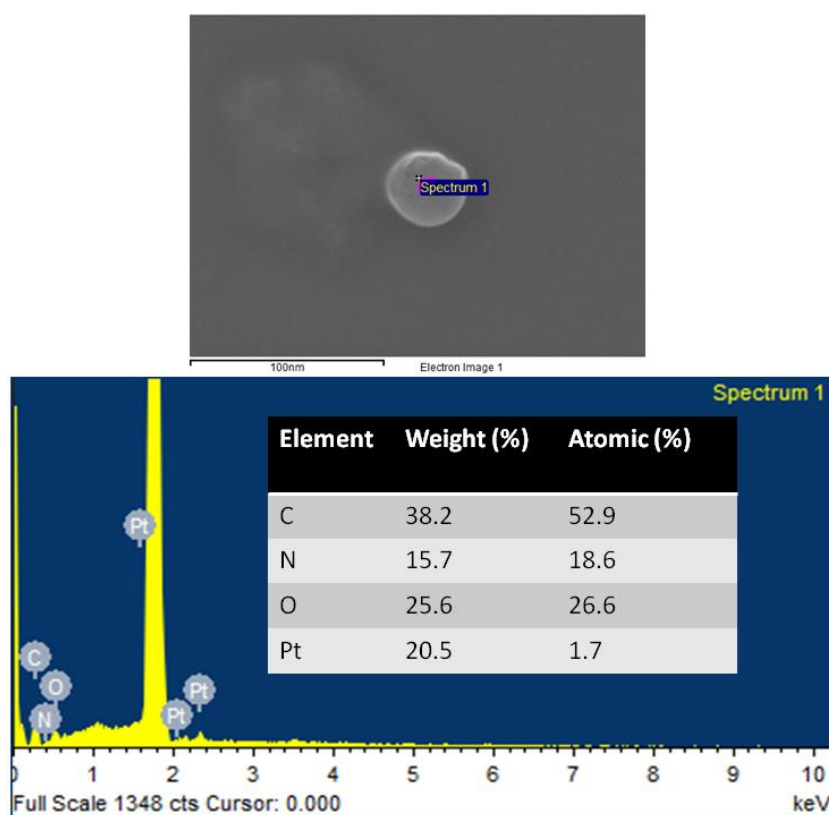| Element	Weight (%)	Atomic (%)
C	38.2	52.9
N	15.7	18.6
O	25.6	26.6
Pt	20.5	1.7

Figure S6: EDXS of PMA-PTX-CDDP-NPs to confirm the presence of cisplatin.
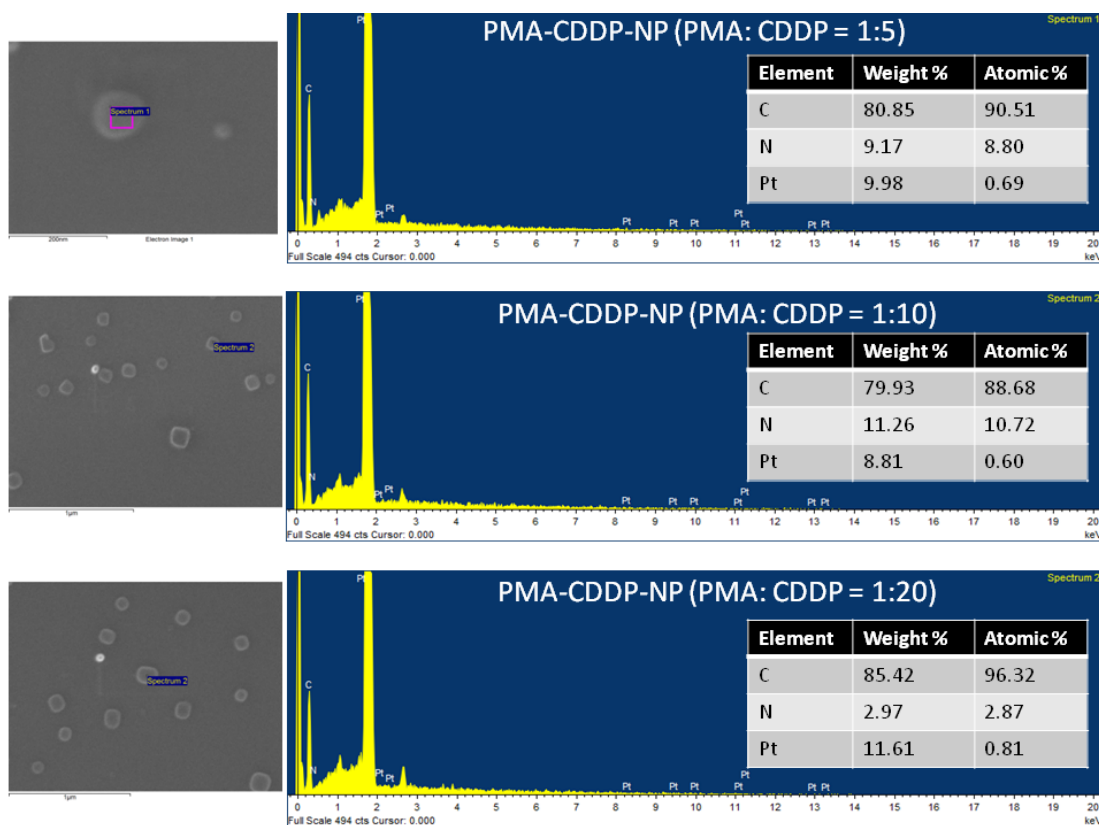
Figure S7: EDXS of PMA-CDDP-NPs at different molar ratios to show the presence of cisplatin.

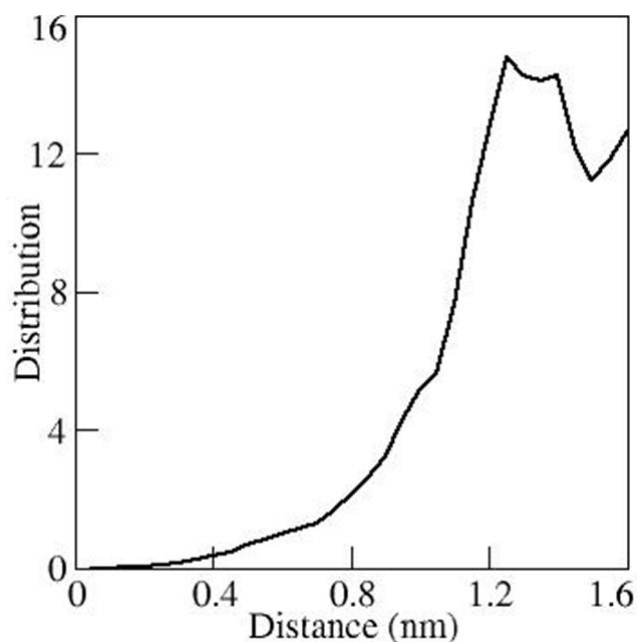**Figure S8**: Distance distribution between hydrophobic Paclitaxel residues over last 10 ns of simulation time.
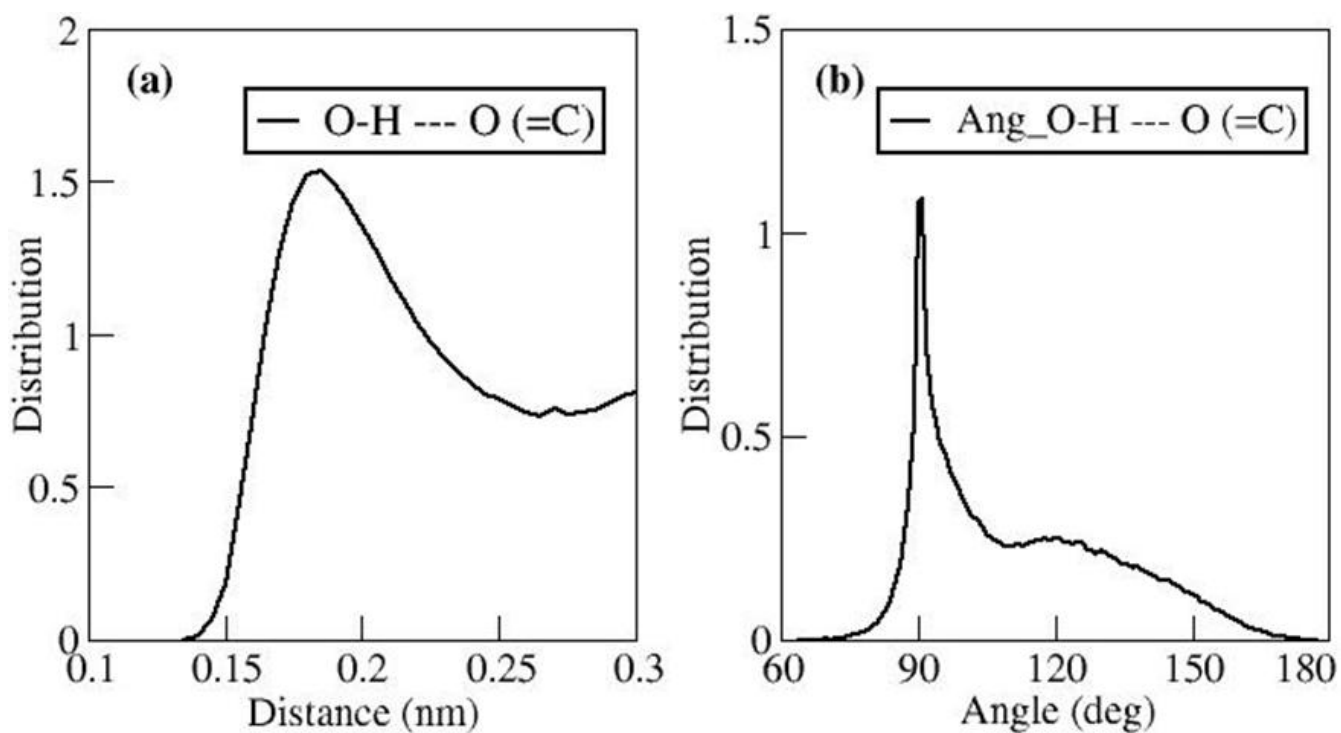
Figure S9: (a) Distance distribution between hydrogen and hydrogen bond acceptor for different possible hydrogen bonding sites. (b) Distribution hydrogen bond donor –hydrogen – hydrogen bond acceptor angles.
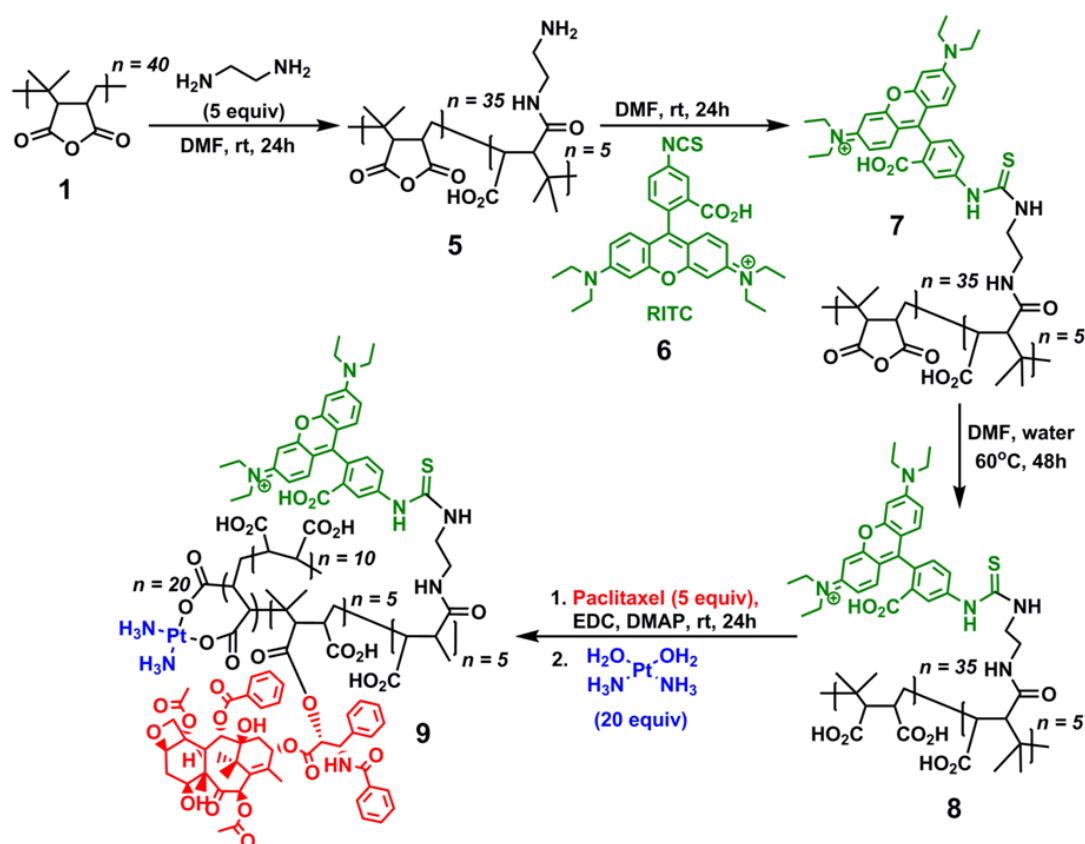

Figure S10: Synthetic scheme of PMA-RITC-PTX-CDDP conjugate (**9**).
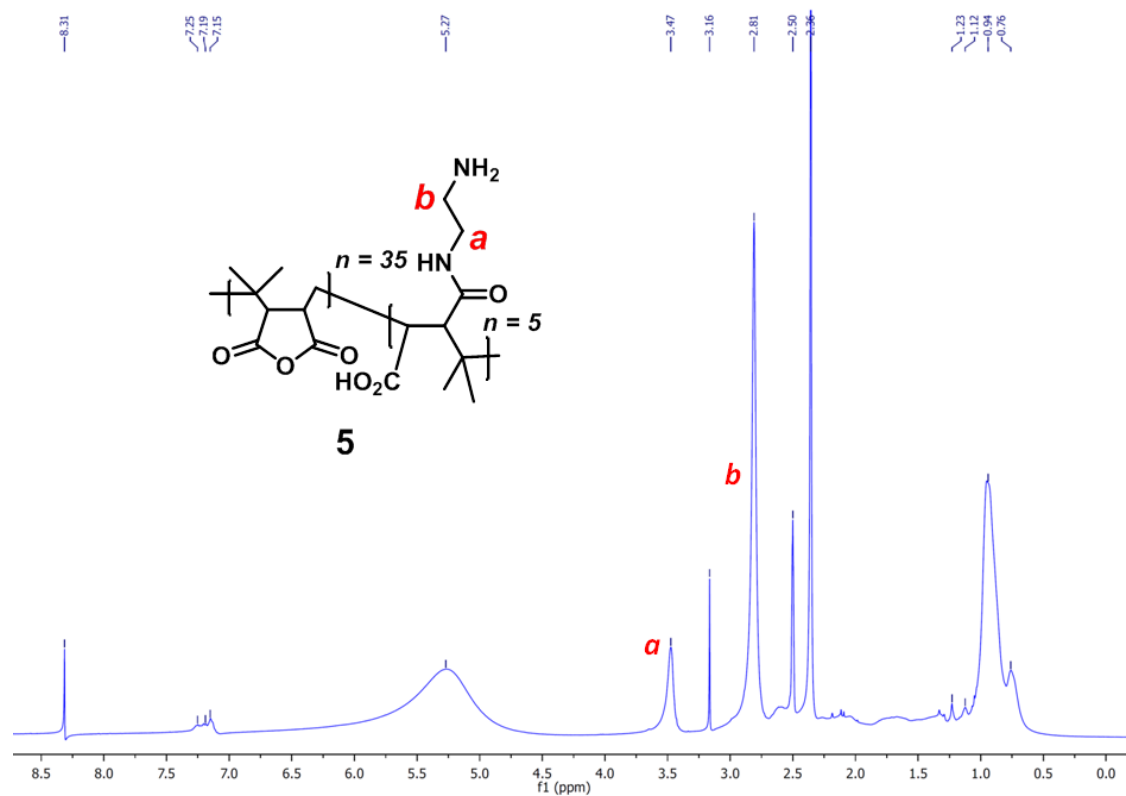
Figure S11: ^{1}H NMR spectra of PMA-ethylenediamine conjugate (**5**).

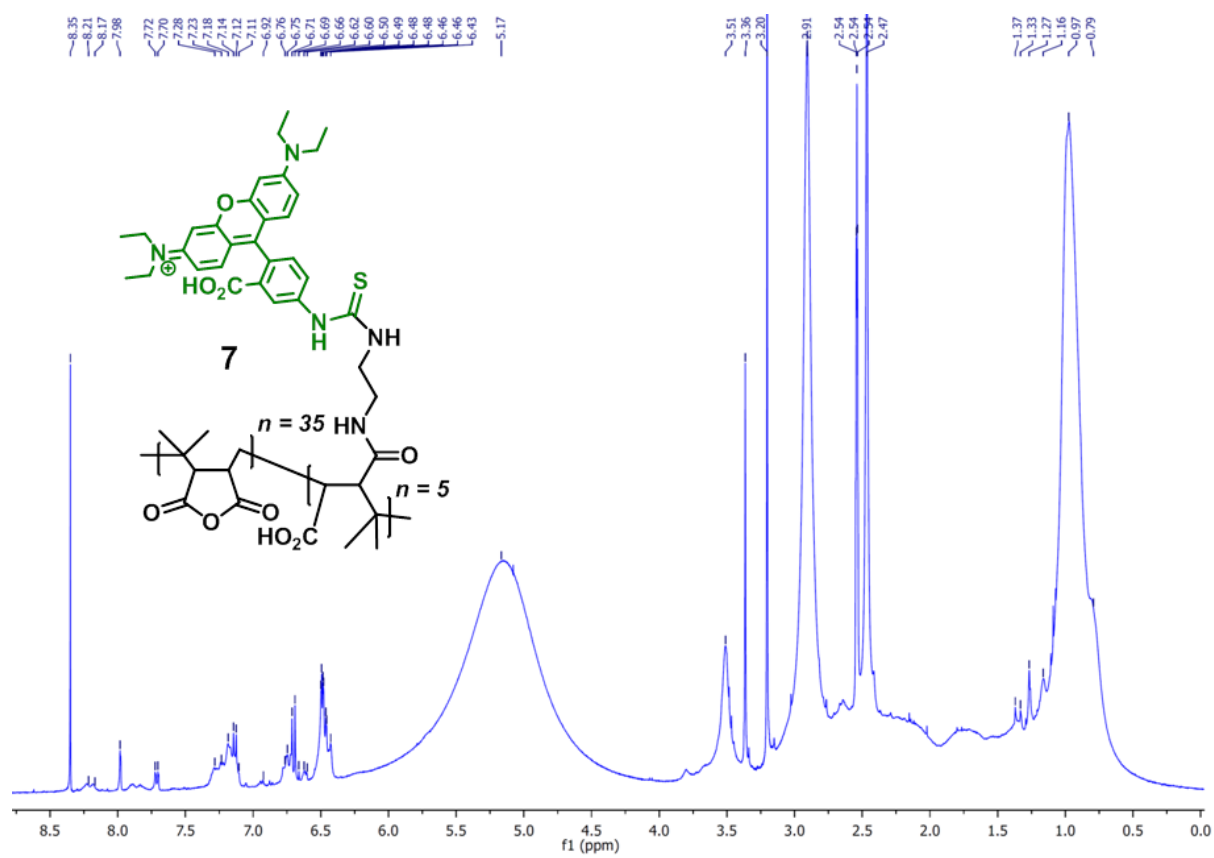**Figure S12**: ^{1}H NMR spectra of PMA-ethylenediamine-RITC conjugate (**7**).
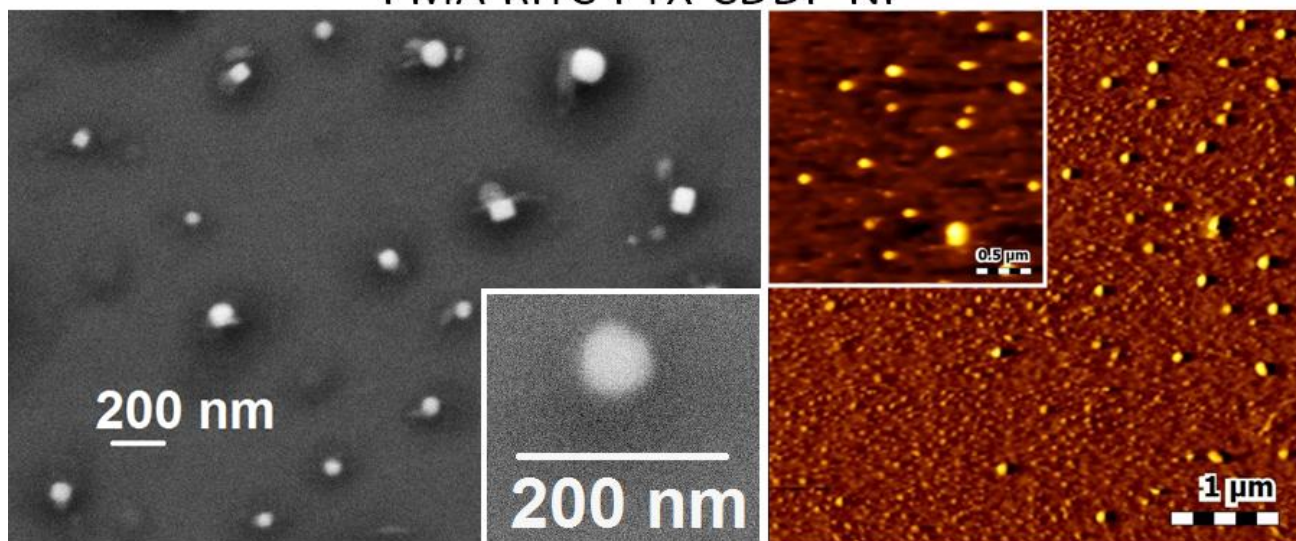
Figure S13: FESEM and AFM images of PMA-RITC-PTX-CDDP-NPs.

Element	Weight %	Atomic %
C	81.41	91.74
N	7.77	7.51
Pt	10.82	0.75
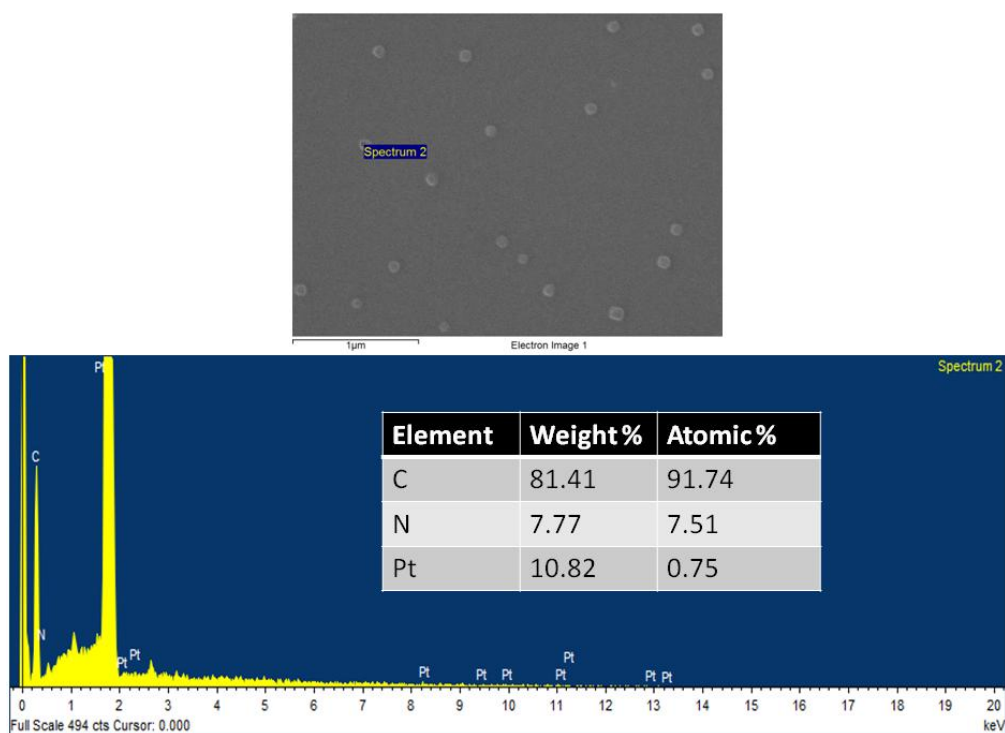
Figure S14: EDXS of PMA-RITC-PTX-CDDP-NPs to confirm the presence of cisplatin.
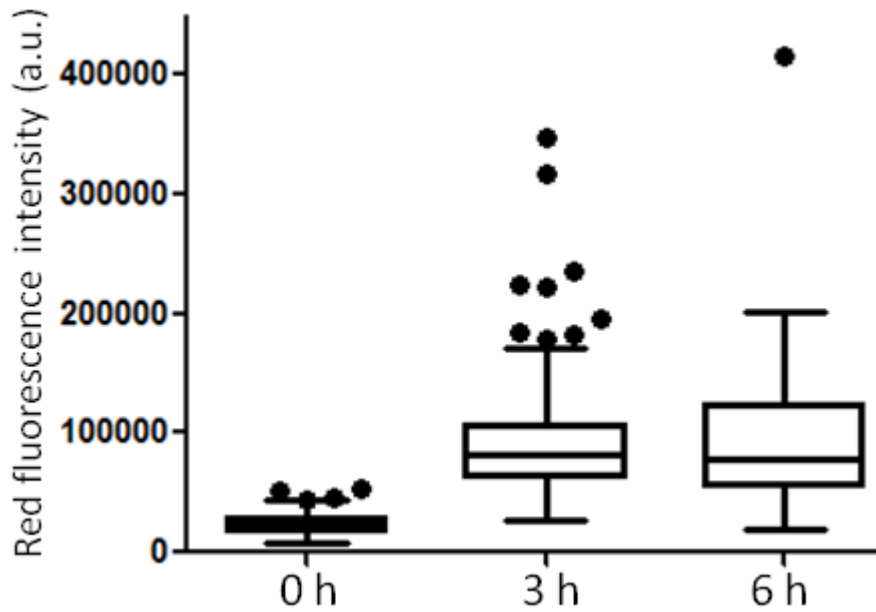
Figure S15: Quantification of red fluorescence intensity from CLSM at 0 h, 3 h and 6 h after incubation with red fluorescent PMA-RITC-PTX-CDDP-NPs in MCF7 cells.
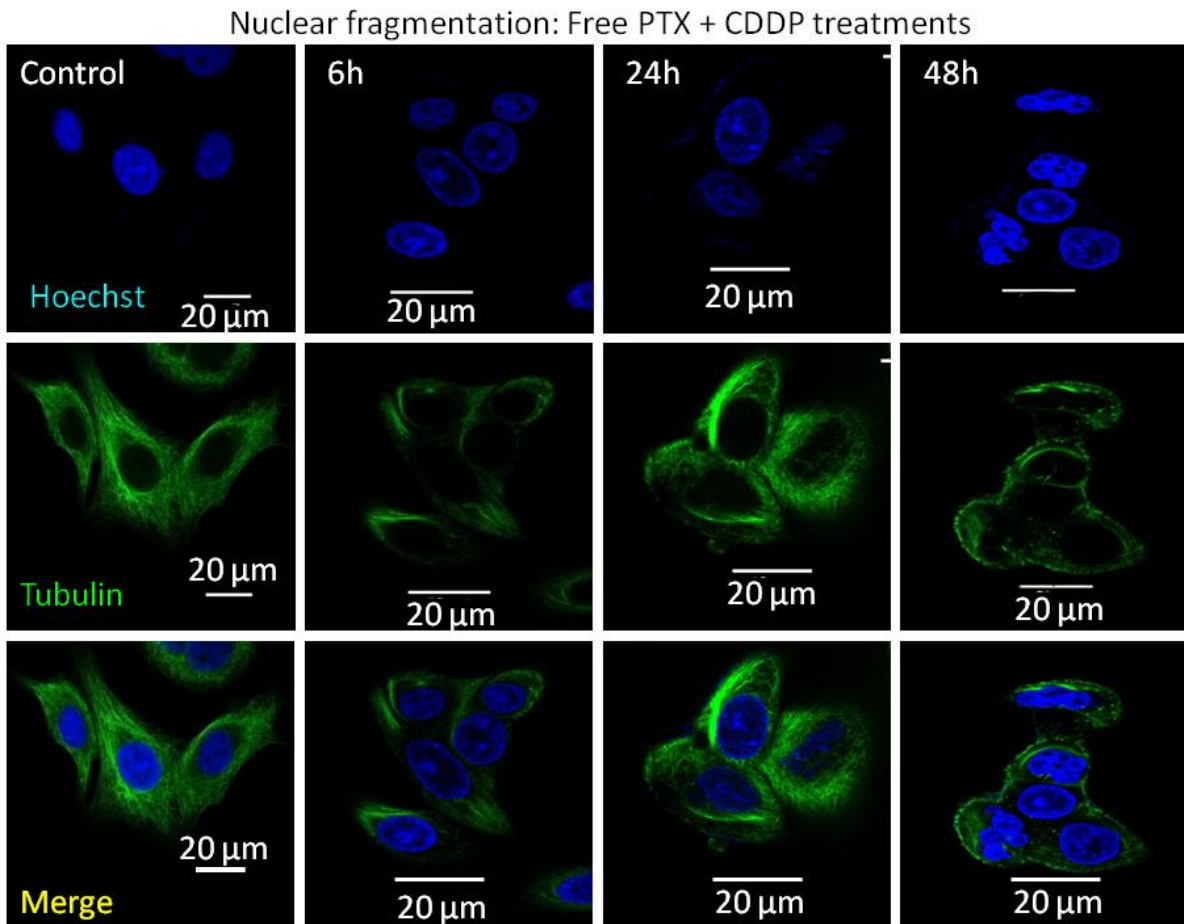
Figure S16: CLSM images of MCF7 cells treated with free PTX-CDDP cocktail at 6h, 24h and 48 h time points to visualize the nuclear fragmentation. Nucleus and tubulin were stained with Hoechst 33342 (blue) and Alexa fluor tubulin antibody (green) respectively. Scale bar = 20 μm.

Figure S17: Quantification of MCF7 cells with fragmented nuclei after treatment with PMA-PTX-CDDP-NPs and free PTX-CDDP cocktail at 6 h, 24 h and 48 h.
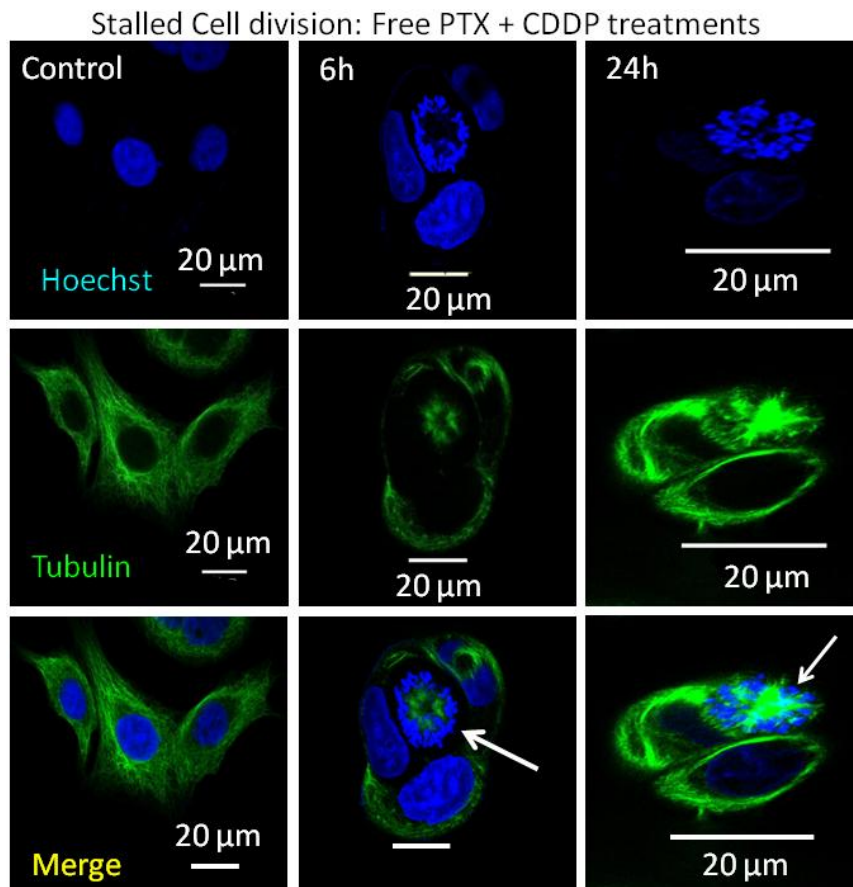
Figure S18: CLSM images of MCF7 cells treated with PTX-CDDP cocktail at 6h and 24h time points to visualize the stalled cell division. Nucleus and tubulin were stained with Hoechst 33342 (blue) and Alexa fluor tubulin antibody (green) respectively. Scale bar = 20 μm.
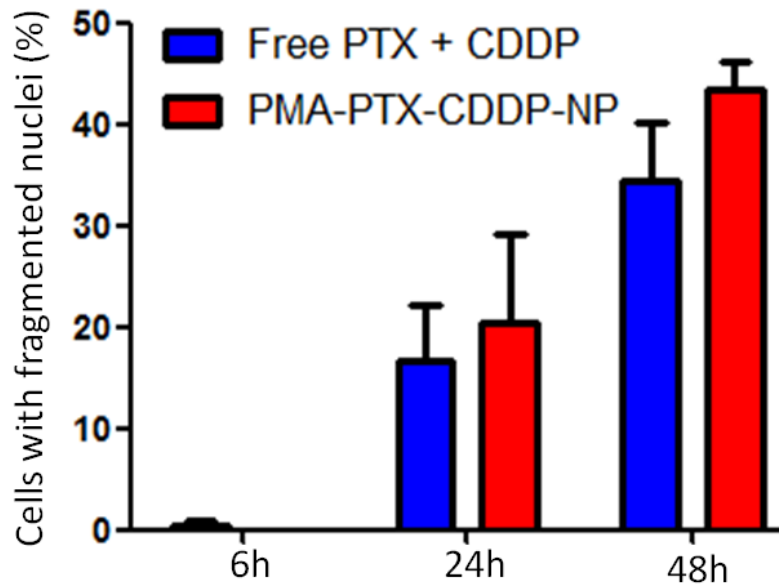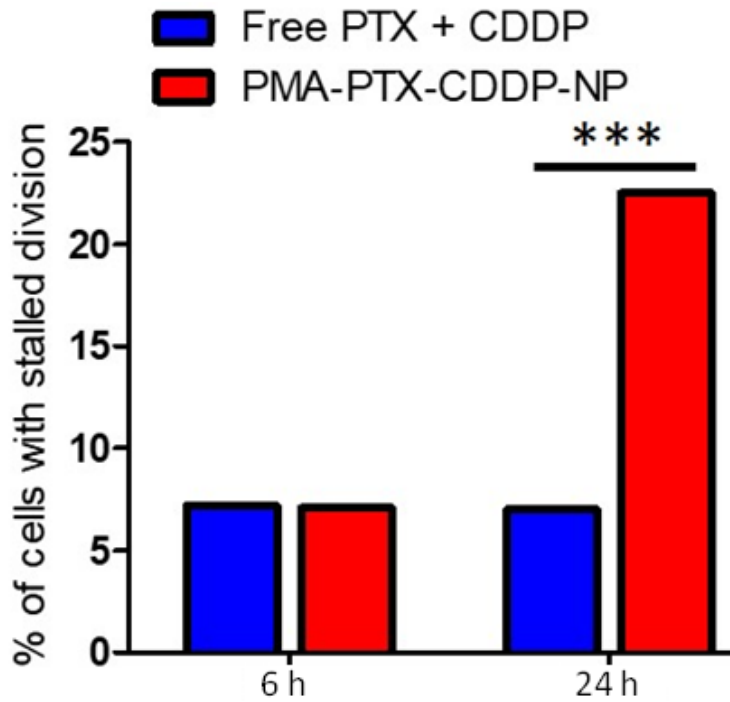
Figure S19: Quantification of MCF7 cells with stalled cell division after treatment with PMA-PTX-CDDP-NPs and free PTX-CDDP cocktail at 6h and 24h. The data represented has been pooled from three independent experiments. Statistical analysis was performed using Chi-square test, *** $p < 0.0001$.
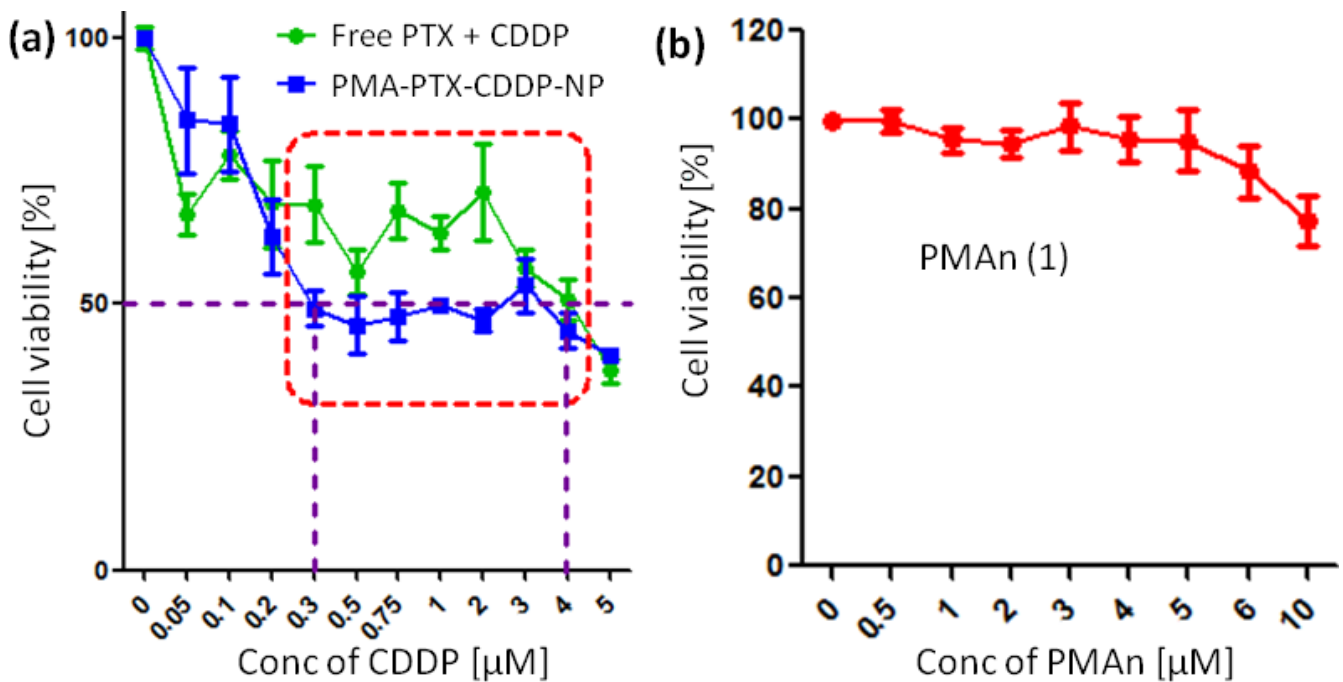

Figure S20: MTT assay of (a) PMA-PTX-CDDP-NPs and free PTX-CDDP cocktail in MCF7 cells at 24h post-incubation and (b) PMAn in a dose dependent manner.
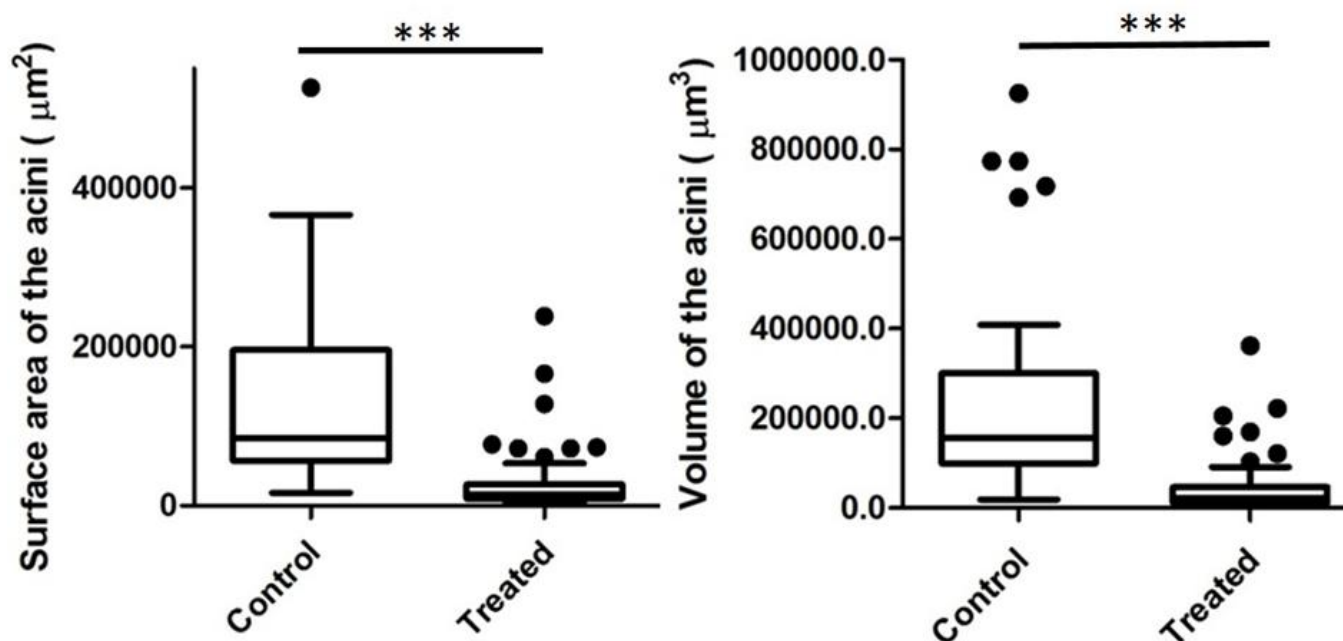
Figure S21: Quantification of the surface area and volume of the acini formed from 3D-culture of MCF7 cells after treatment with PMA-PTX-CDDP-NPs. Statistical analysis was performed using Mann Whitney test, p<0.05 has been considered as statistically significant; *** p<0.0001.

Atom	Types of atom	σ (nm)	ε (kJ mol⁻¹)	q
PT	Central PT atoms	0.4348	1.6736	0.346
COS	O atom coordinated to PT	0.303	0.502	-0.577
TOS	Another O atom coordinated to PT	0.303	0.502	-0.577
NAC	Coordinated N atom cis to COS	0.330	0.3347	-0.225
NAT	Coordinated N atom trans to COS	0.330	0.3347	-0.225
HC	H atom attached with Coordinated N	0.04	0.19246	0.180

Table S1: Atom types and their non-bonded parameters.

Angle	θ_{ijk}^0 (deg)	k_{ijk}^θ (kJ mol⁻¹ rad⁻²)	r_{ik}^0 (nm)	k_{ijk}^{UB} (kJ mol⁻¹ nm⁻²)
COS – PT - NAC	90.0	549.6	0.0	0.0
COS – PT - NAT	180.0	549.6	0.0	0.0
TOS – PT - NAC	180.0	549.6	0.0	0.0
TOS – PT - NAT	90.0	549.6	0.0	0.0

Table S2: Table showing calculated angle parameters.

Dihedral	ϕ_s (deg)	k_ϕ (kJ mol^{-1})	multiplicity
HC-NAC-PT-NAT	0.0	2.75	3
HC-NAT-PT-NAC	0.0	2.75	3

Table S3: Table showing calculated dihedral parameters.