Electronic supporting information

Assessing therapeutic potential of magnetic mesoporous nanoassemblies for chemo-resistant tumors

Lina Pradhan [a, d, ‡], B. Thakur [b, ‡], R. Srivastava [c], P. Ray [b, *] , D. Bahadur [d, *]

[a] *Centre for Research in Nanotechnology and Sciences, IIT Bombay, Mumbai, 400076,India.*

[b] *Tata Memorial Centre, Kharghar, Navi Mumbai, 410210, India.*

[c] *Department of Biosciences and Bioengineering, IIT Bombay, Mumbai, 400076, India.*

[d] *Department of Metallurgical Engineering and Materials Science, IIT Bombay, Mumbai, 400076 India.*

'‡' These authors contributed equally.

*Corresponding authors. Tel.: +91 22 2740 5119 (P. Ray). Tel.: +91 22 2576 7632 (D. Bahadur).

E-mail addresses: pray@actrec.gov.in (P. Ray), dhirenb@iitb.ac.in (D. Bahadur).

1. Results and discussion

TEM analysis

The TEM images of MMNA and LMMNA are seen as spherical stucture with an average diameter of about ~73 and 92 nm (size distribution is shown in our pevious study), respectively (ESI Figure S1). ESI Figure S1A shows the aggregate of smaller-sized Fe_3O_4 nanoparticles, and a single MMNA is magnified in ESI Figure S1B. As shown in ESI Figure S 1C, the lipid coating on the surface of the MMNA slightly increased the size of the MMNA. ESI Figure S1D shows a magnified image of a single LMMNA and a thin layer of lipid over the MMNA markedasred lines with yellow arrows. This result suggests that the lipid-coated MMNA were successfully synthesized.

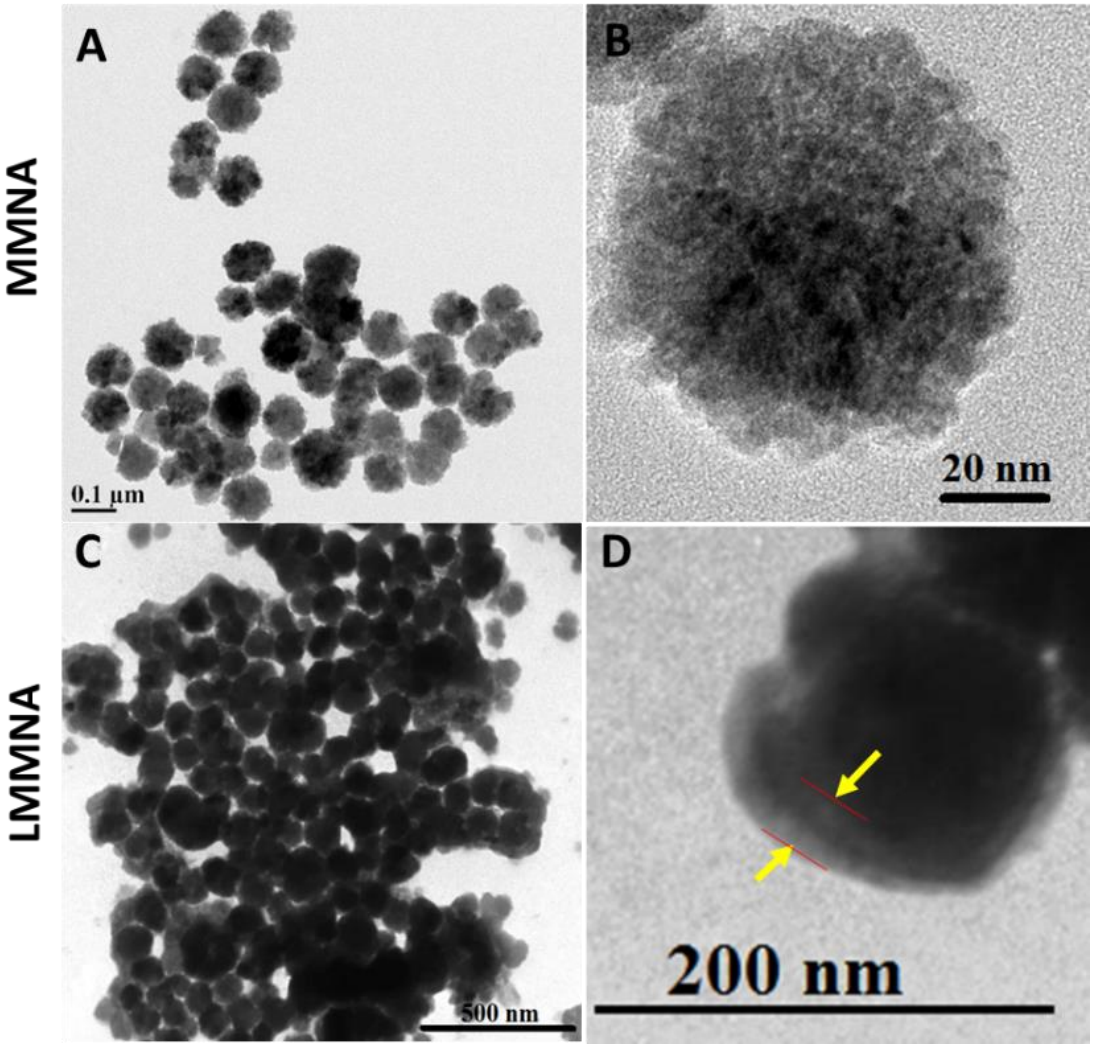

Figure S1. TEM images of (A) MMNA, (B) Magnified image of single MMNA, (C) Lipid coated MMNA (LMMNA) and (D) Magnified image of single LMMNA and red lines with yellow arrow marked for thin lipid layer on the surface of MMNA.

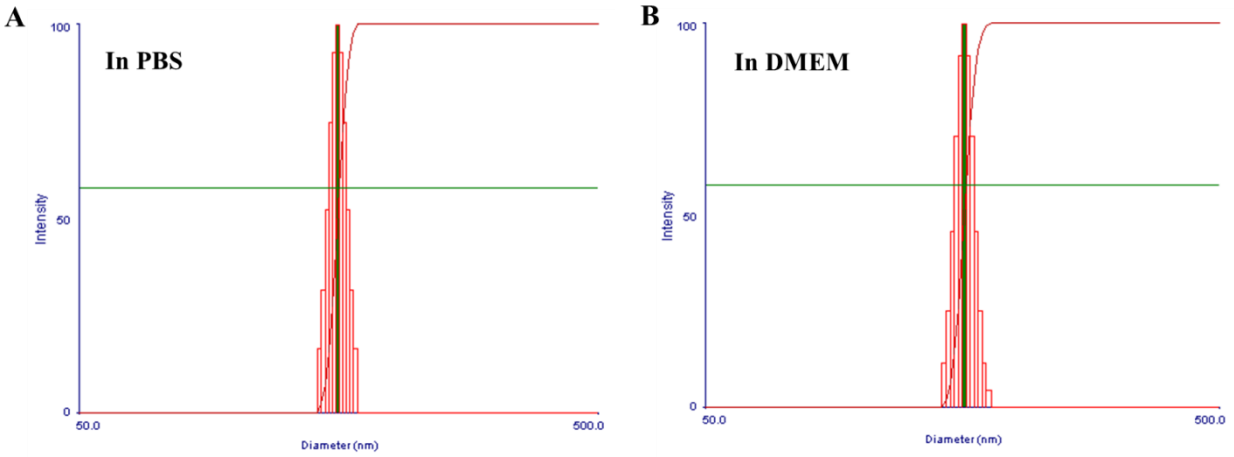

Figure S2. DLS measurement of LMMNA in PBS and DMEM showing mean hydrodynamic diameter of (A) ~157 nm in PBS and (B) ~159 nm in 10% FBS supplemented DMEM, respectively.

Loading efficiency and loading capacity of DOX:TXL-LMMNA

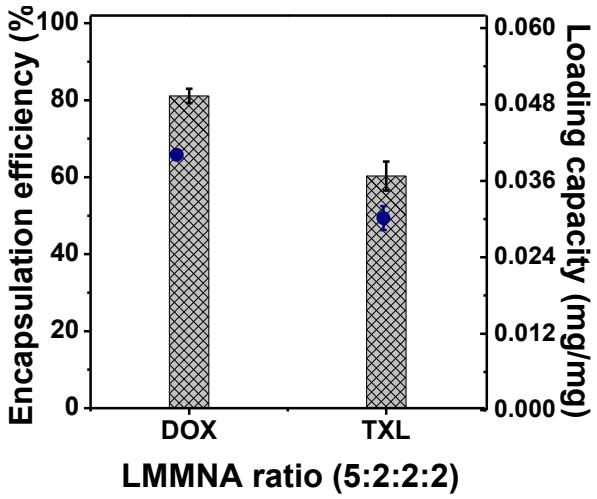

Figure S3. Encapsulation efficiency (%) with loading capacity (mg/mg) of DOX and TXL in LMMNA (5:2:2:2 ratio). All experiments were performed in triplicate.

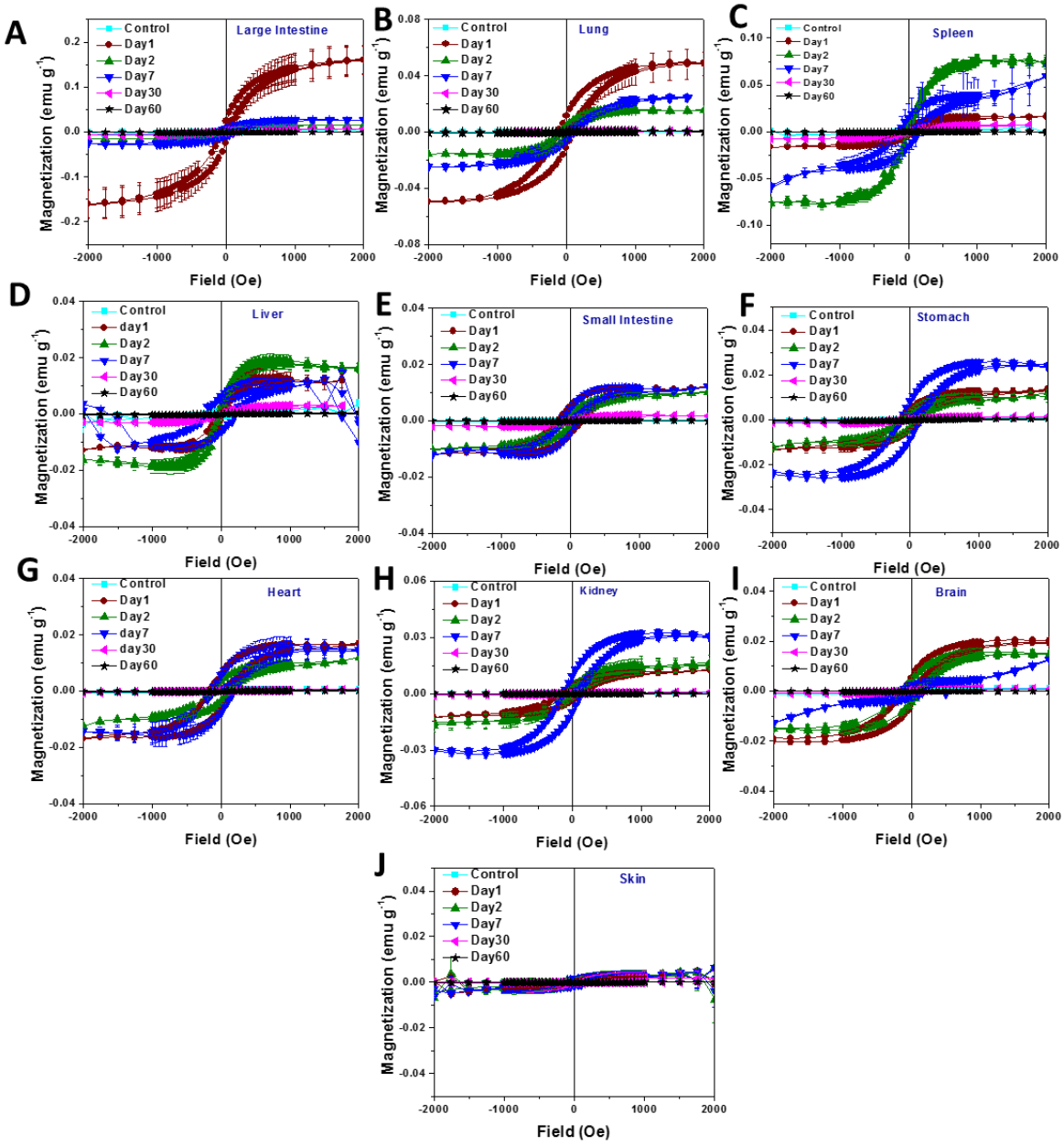

Figure S4. Magnetic hysteresis curves of (A) large intestine, (B) lung, (C) spleen, (D) liver, (E) small intestine, (F) stomach, (G) heart, (H) kidney, (I) brain and (J) skin of non-tumored nude mice post administration of DOX:TXL-LMMNA.

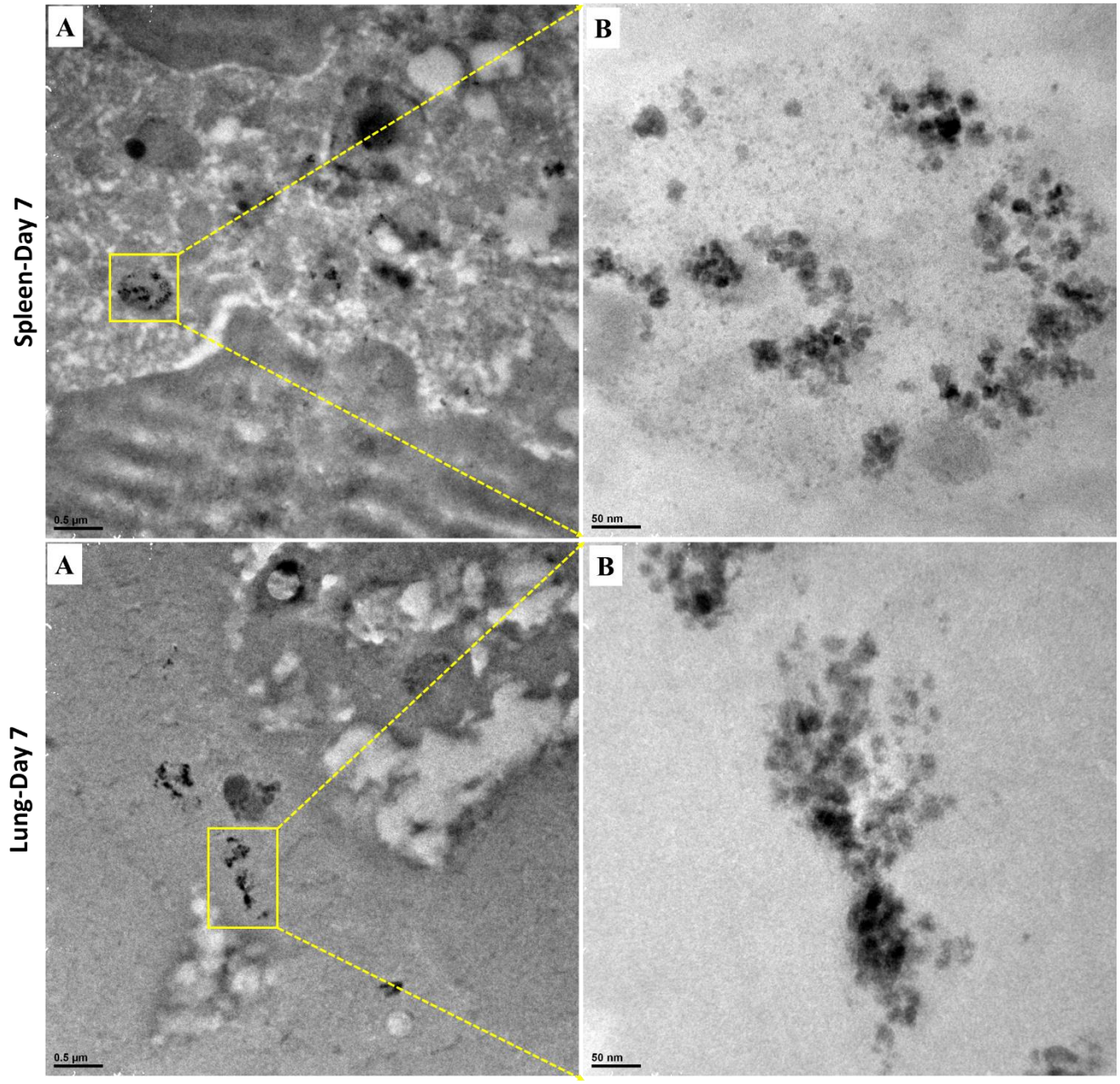

Figure S5. TEM images of uptake and accumulation of nanoparticles in spleen and lung tissues after Day 7 treatment. Column (A) both tissues show the localization of the LMMNA into tissue and column (B) magnified image of the area indicated by the yellow box in column (A), showing the morphology and accumulation of LMMNA.

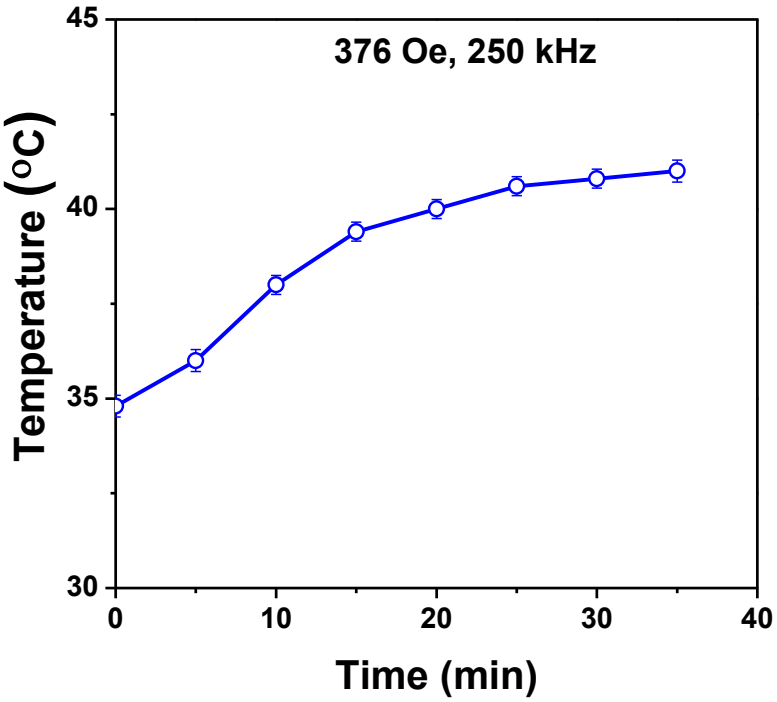

Figure S6. Thermal profile of tumor surface after intratumoral administration of DOX:TXL-LMMNA (1 mg) under ACMF.

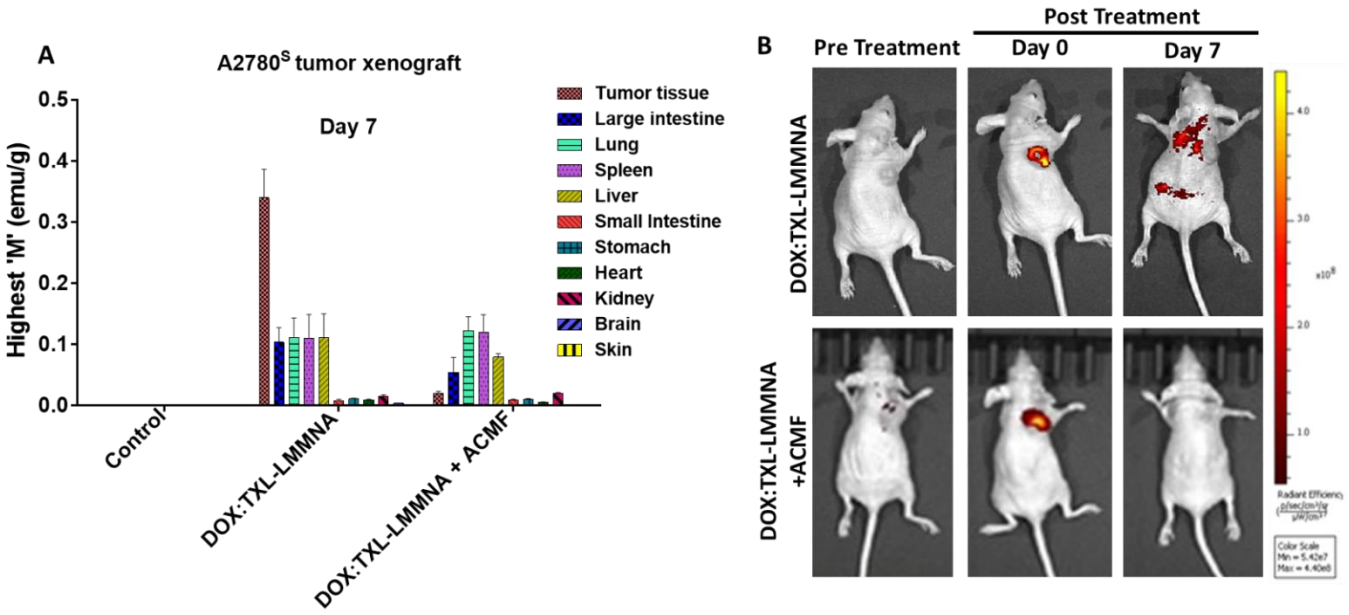

Figure S7. *In vivo* biodistribution using 50 mg/kg of DOX:TXL-LMMNA in the absence/presence of ACMF at 376 Oe, 250 kHz frequency in A2780^{S} tumor xenografts NUDE mice. (A) Highest magnetization values of vital organ tissues at Day 7 (B) Representative fluorescence images of A2780^{S} tumor xenografts.

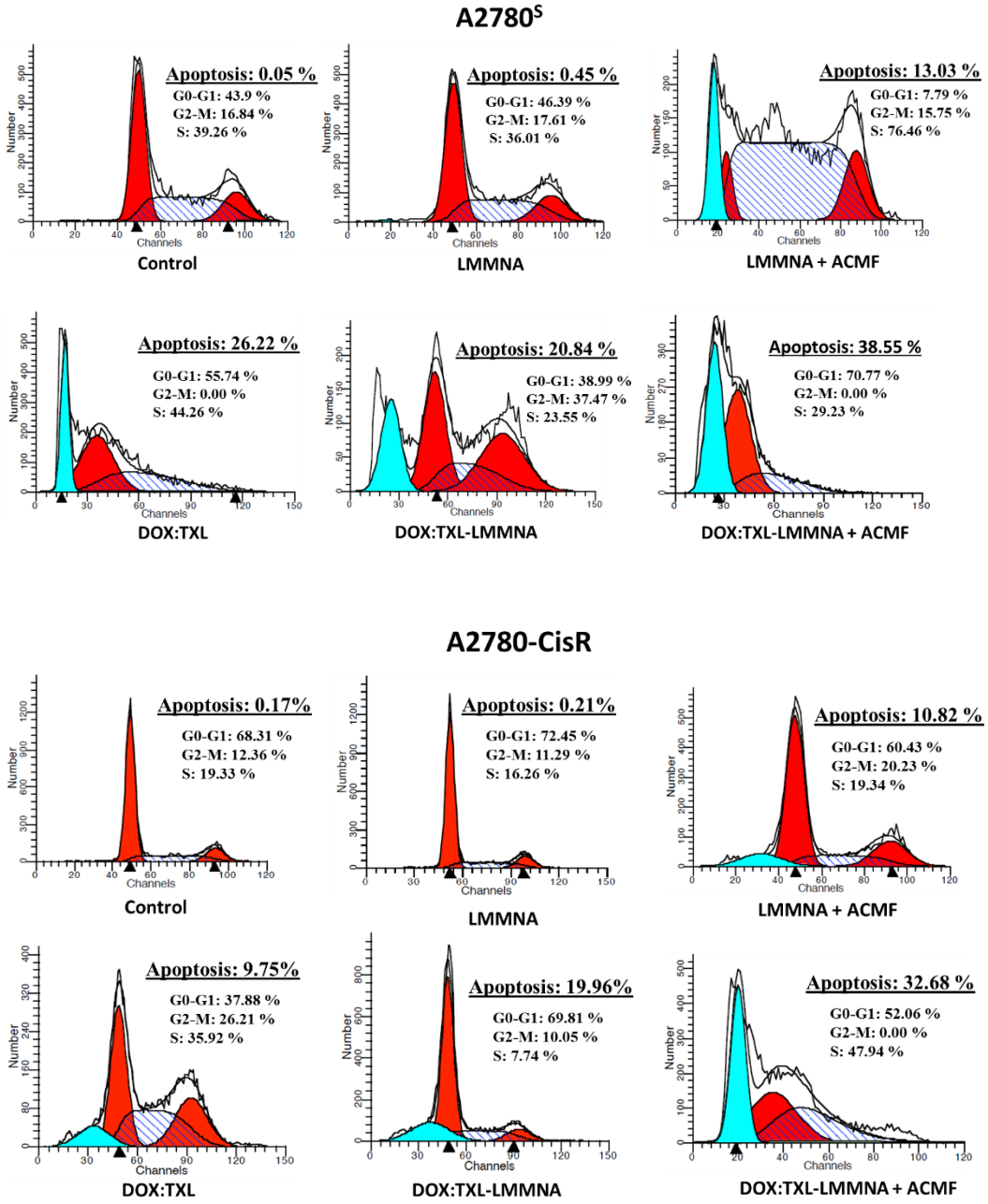

Figure S8. The cell cycle distribution and apoptotic population percentages by PI staining on $A2780^S$ and A2780-CisR cell lines. The histograms show the untreated as a control, treated with LMMNA, DOX:TXL and DOX:TXL-LMMNA in the absence and presence of ACMF. In all cases, G1/G2 phase is arrested with LMMNA+ACMF, only DOX:TXL, DOX:TXL-LMMNA and DOX:TXL-LMMNA+ACMF, respectively.

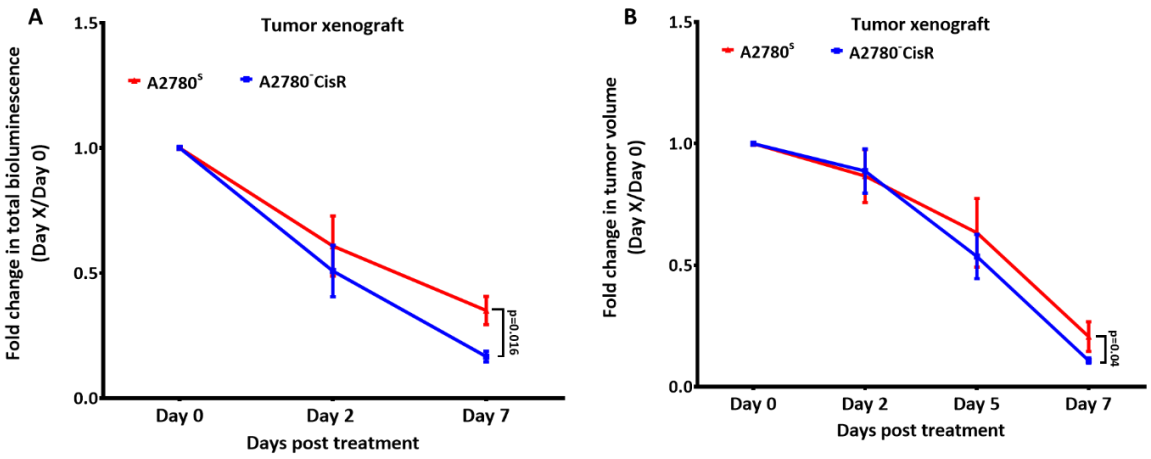

Figure S9. Comparison between efficacy of DOX:TXL-LMMNA in the presence of ACMF on $A2780^{S}$ and A2780-CisR tumor xenografts. A. Fold change (normalized with Day 0) in bioluminescence signal (A) and tumor volume (B) between sensitive and resistant tumor xenograft.

Table S1: Number of mice used in biodistribution study

Experiment Condition	Untreated	Days post DOX:TXL-LMMNA adminstration					Total
Number of mice		Day 1	Day 2	Day 7	Day 30	Day 60	
	5	5	5	5	5	5	30

Table S2: Number of mice used for estimating *In vivo* therapeutic efficacy of DOX:TXL-LMMNA on A2780 S tumor xenografts

Experiment Condition	Untreated	DOX:TXL-LMMNA	DOX:TXL-LMMNA+ACMF	Total
Number of mice	5	5	5	15

A total of additional 10 mice, treated with DOX:TXL-LMMNA+ACMF where kept for 6 month followup study.

Table S3: Number of mice used for estimating *In vivo* therapeutic efficacy of DOX:TXL-LMMNA on A2780-CisR tumor xenografts

Experiment Condition	Cisplatin+ACMF	DOX:TXL+ACMF	LMMNA+ACMF	DOX:TXL-LMMNA+ACMF	Total
Number of mice	5	5	5	5	20