Supporting Information

Compatibility study

The isothermal stress testing was performed to evaluate the compatibility of MTX & ACL with all excitements including PCL. Briefly, 20 mg of MTX and ACL was dissolved in 3 ml DMF and ethanol, respectively and mixed together with melted all lipids & polymer mixture. The mixture was centrifuged at 10,000 rpm for 10 min, and clear supernatant was taken in the glass vials and stored at 600˚C/RH 75% for 4 weeks. The samples were quantitatively analyzed for residual MTX & ACL content by HPLC and FTIR spectroscopy methods

Solubility study

The solubility of MTX & ACL was determined in Gelcuire and PCL as reported earlier [28]. Briefly, Gelucire and PCL were heated at 10°C above their melting point in temperature regulated water bath, and liquid lipids & PCL taken in culture tubes and kept on water bath with continuous stirring. The small quantity of MTX & ACL was then gradually added separately until drug saturation is achieved. The estimated amount of supernatant was quickly transferred into chloroform methanol mixture. The mixture was suitably diluted with mobile phase, and filtered through 0.22µ filter to estimate the drug content by HPLC in triplicates.

SI Table 1: HPLC condition for ACL and Methotrexate

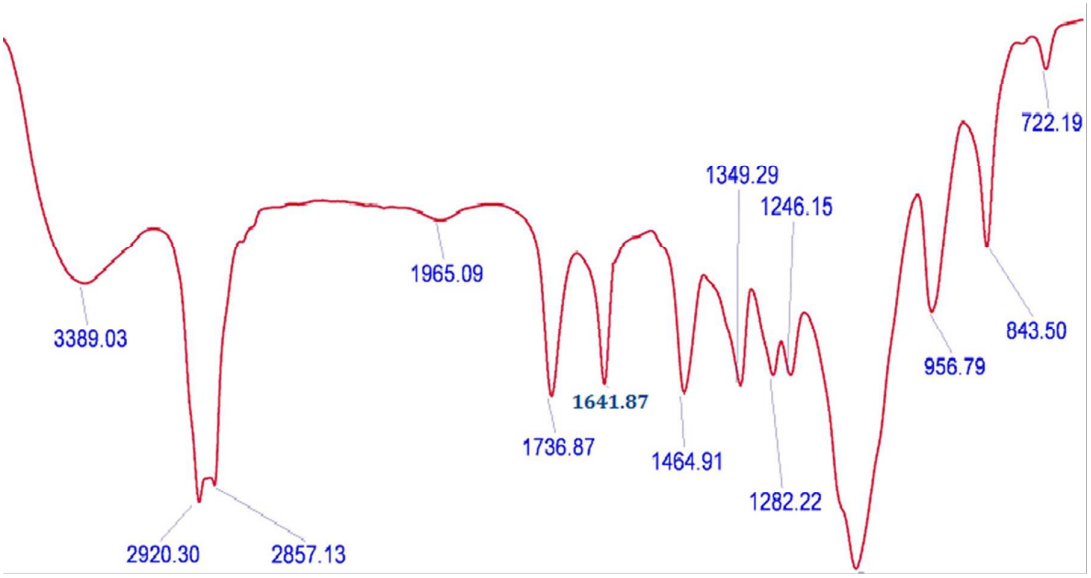
Particular(s)	Methotrexate	ACL
Instrumentation	Shimadzu LC-2010 C_{HT} equipped with a system controller, quaternary gradient pump, solvent delivery module, online degasser, auto-samples with cooler, auto injector (injection volume ranging between 5 – 100 µL) and UV-Vis detector was used for analysis	
Column	RP C18 (10µm, 4.6 mm×250 mm, Waters)	RP; Thermohypersil BDS C18 ; Column length: (250x4.6 mm) ; 5µ
Mobile phase	10 mM KH_2PO_4 buffer (pH=3.0), Methanol, ACN (70:20:10)	Methanol: 0.02% orthophosphoric acid pH(3.0) (70:30 v/v)
Diluent	Mobile phase	Mobile phase
λ_{max} (nm)	303	275
Flow rate (mL/min)	0.6	1
Run time (min)	20	20
Column oven temperature (°C)	30	30
Injection volume (µL)	10	20 µL
Mode	Isocratic	Isocratic
Retention time (Mints)	7.5± 0.4	8.4 ± 0.5

SI Table 2: Dosing schedule of ACL & DMBA.

Groups	Treatment and dosing
Control	Control DMBA treatment 50 mg/ kg/week, orally administered for three consecutive weeks
Prophylactic and anticancer activity	Free ACL and LPHNPs-ACL (equivalent to 10 mg/kg free ACL) was administered twice a week from day one before DMBA administration till 13th weeks

Compatibility study

All samples were quantified for initial & residual content of MTX and ACL through HPLC method. There were hardly any significant changes seen in the initial and final drugs content, and therefore suggests the compatibility of MTX & ACL with their excipients.



SI Figure 1: FTIR of fucose anchored LPHNPs-(MTX+ACL)

SI Table 3: Data of compatibility study showing initial & final content of MTX & ACL

	Initial recovery (%)	Recovery after 4 weeks (%)
MTX	96.89±1.07	95.38±2.16
ACL	96.12±2.17	95.13±2.22

SI Table 4: Solubility of ACL & MTX

Solid Lipids/PCL	Solubility (mg/gm)	
	ACL	MTX
Gelucire® 48/16	375.06±11.8.37	187.76±13.26
PCL	115±10.56	263±13.26
Gelucire® 48/16+Ethanol (1:1)	435.06±13.81	137.76±13.26
PCL+DMF(1:1)	155±10.56	393±13.26

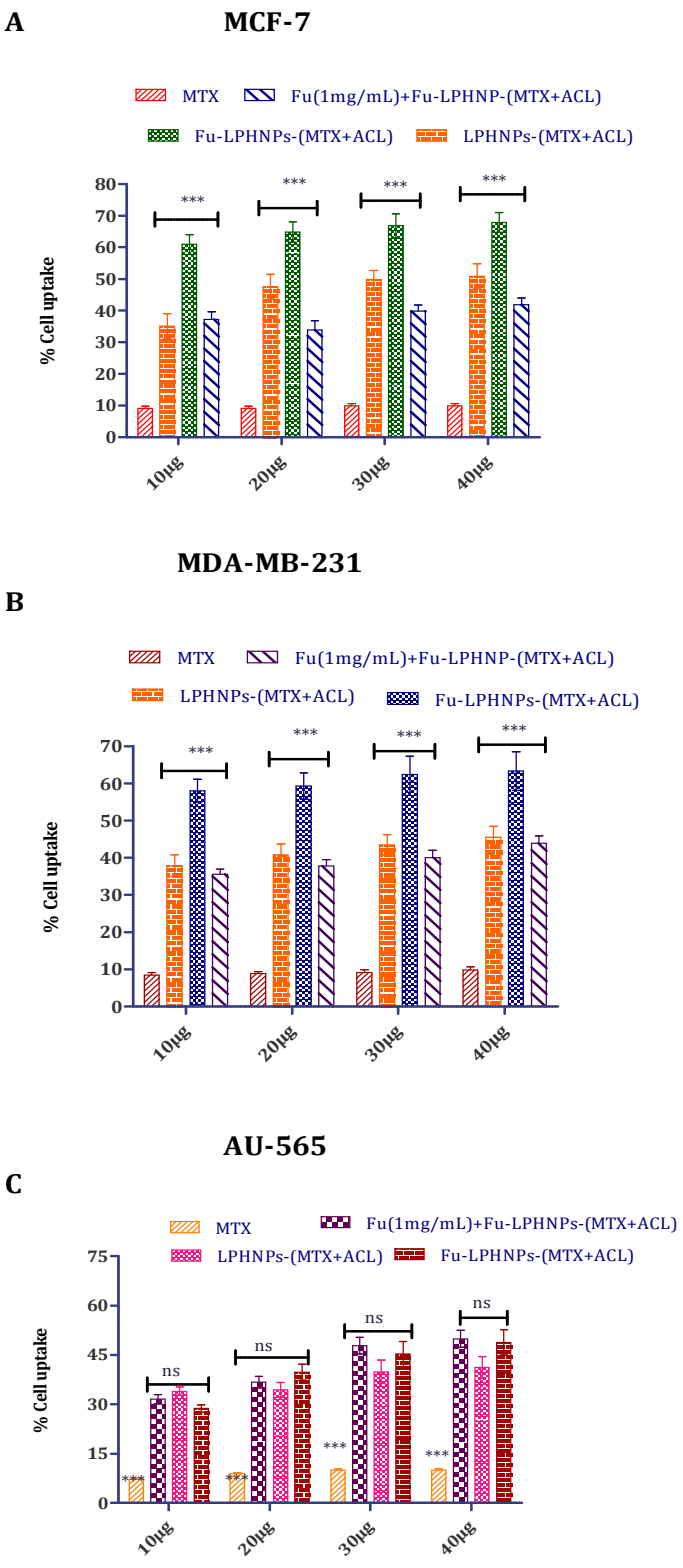


Figure 2: Quantitative cell uptake analysis is presented with concentration-dependence of drugs in, (A) MCF (B) MDA-MB-231 and (C) AU-565 cells