



# Membrane Fluidity as a New Means to Selectively Target Cancer Cells with Fusogenic Lipid Carriers

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## SUPPORTING INFORMATION

# Membrane Fluidity as a New Mean to Selectively Target Cancer Cells with Fusogenic Lipid Carriers

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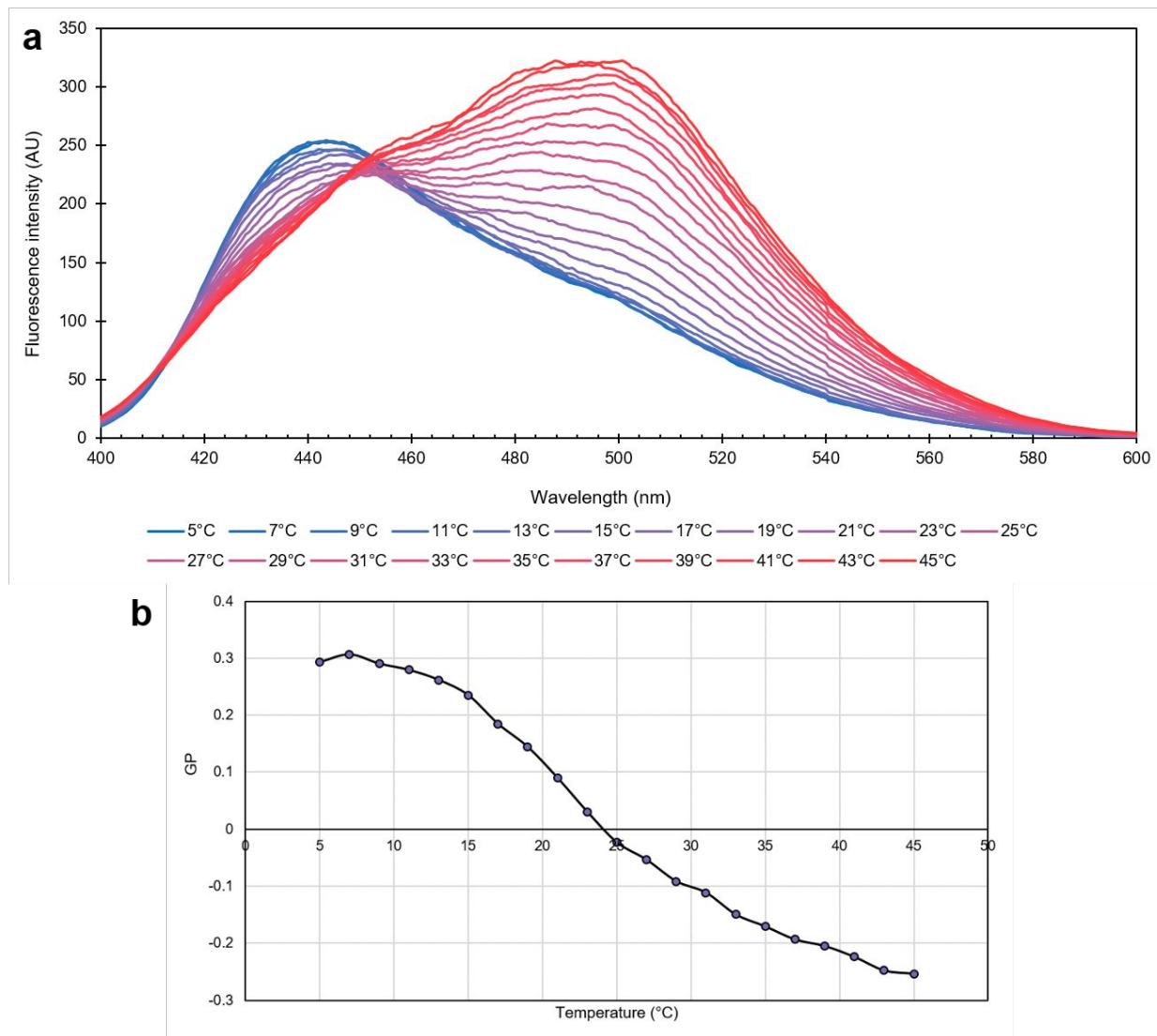
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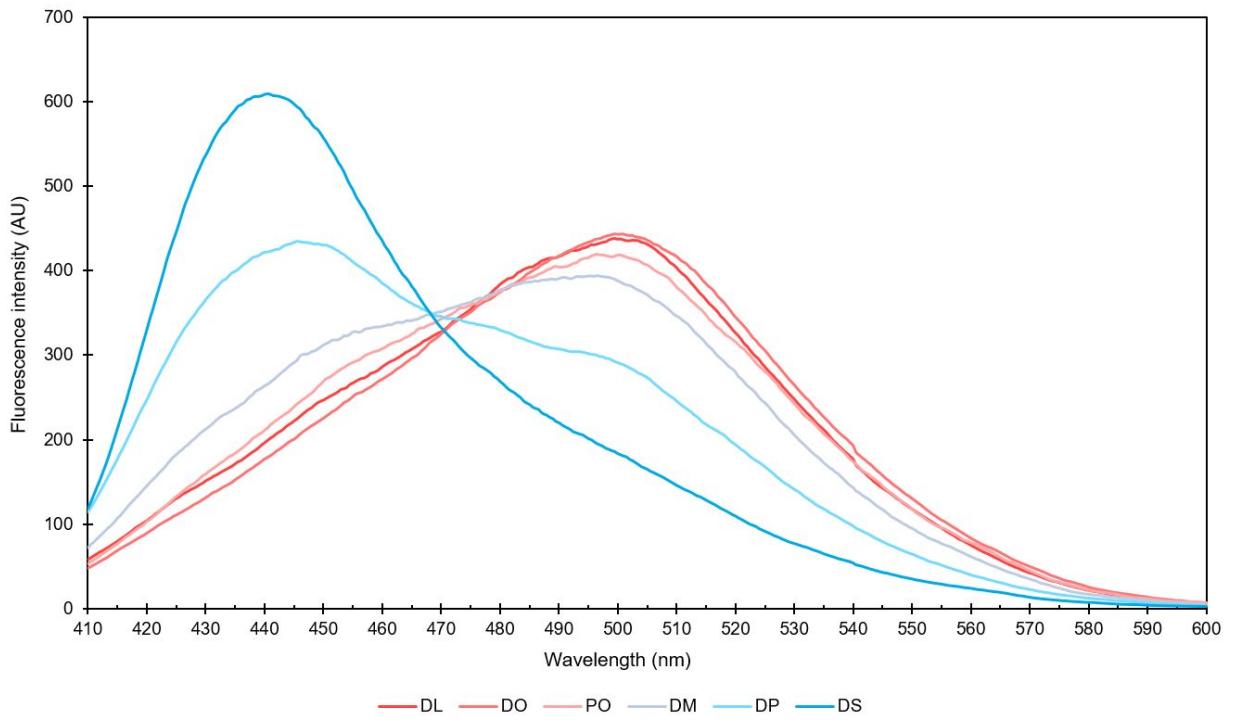
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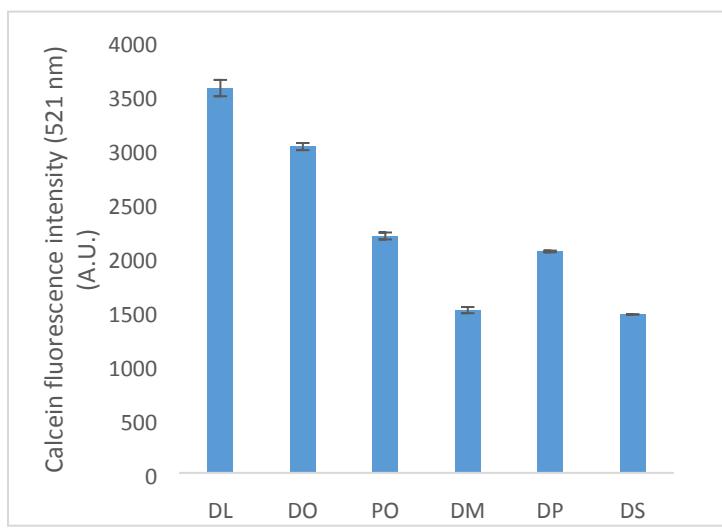


**Figure S1. Dioll accurately reports the membrane fluidity state of liposome membranes.**

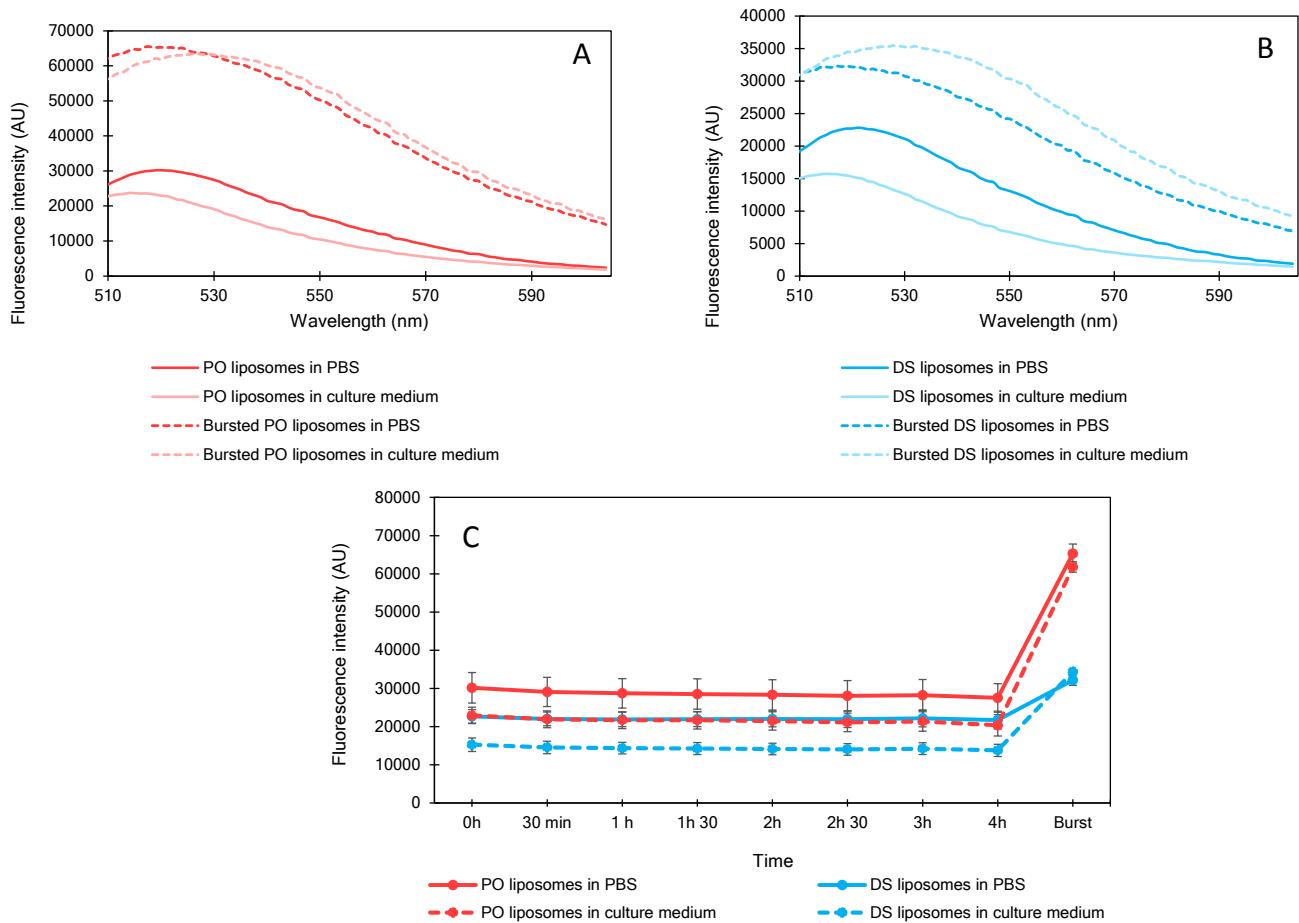
- a) Fluorescence spectra of DMPC liposomes incubated with Dioll for 15 min, from 5°C to 45°C.
- b) GP measurements, showing an inflection at 24°C, the usual phase-transition temperature of DMPC.



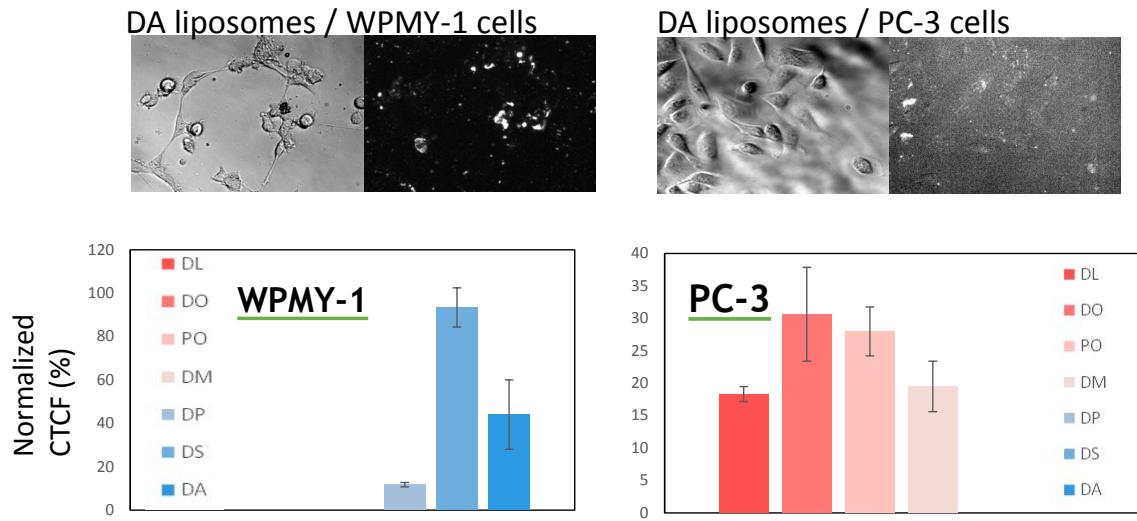
**Figure S2. Dioll fluorescence spectra** in liposomes with varying phase-transition temperatures reports differences in membrane fluidity states of liposomes made of 80% PC and 20% DOPE.



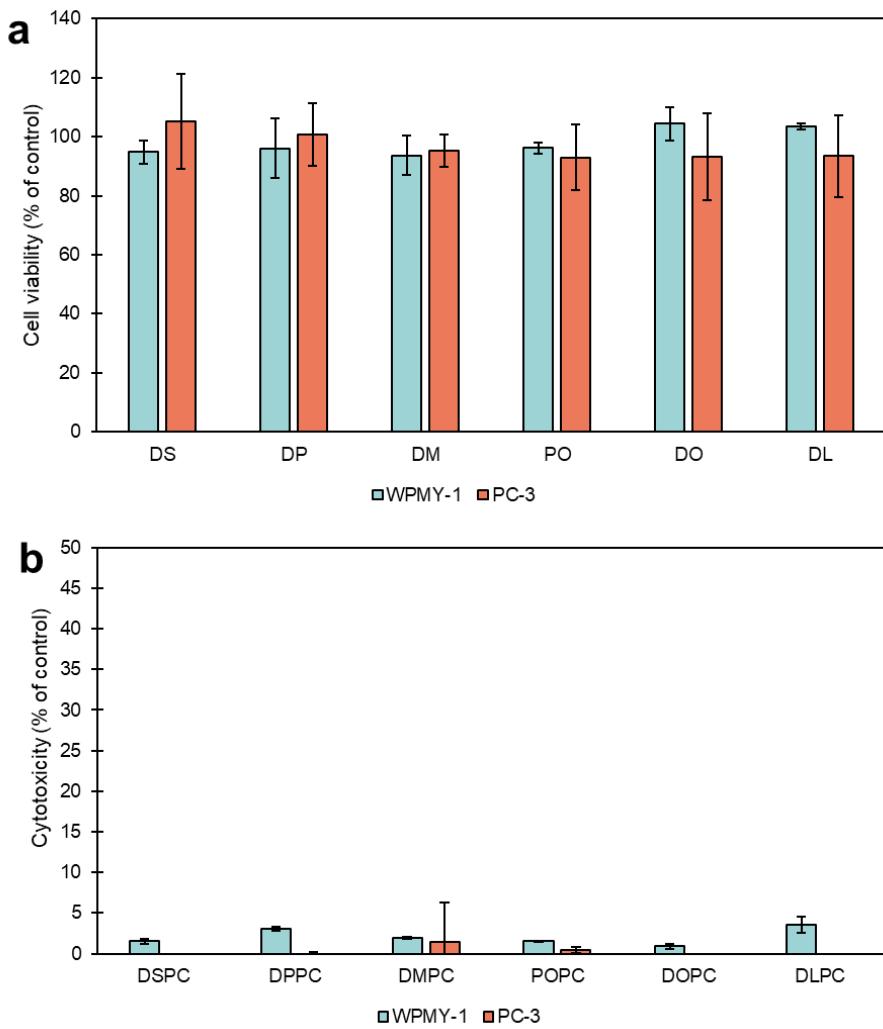
**Figure S3. Calcein load in liposomes.** Calcein fluorescence intensity at 521 nm (488 nm excitation) as a function of liposome preparation for a liposome concentration of 100 µg/ml. Calcein was loaded in the liposomes at a concentration of 5 µM. Free calcein was removed with size exclusion chromatography.



**Figure S4: Liposome stability in cell culture medium.** PO (A) and DS (B) liposomes were loaded with 60 mM calcein and diluted either in PBS or in FBS-containing DMEM medium. Fluorescence spectra were recorded after excitation at 488 nm before (full lines) and after (dotted lines) liposome burst with 10  $\mu$ l HCl 1M. The fluorescence intensity increased after liposome burst indicated release from quenching. C. Evolution of calcein fluorescence (measured at maximum emission wavelength, 521 nm) with time in PBS (full lines) or in FBS 10 % cell culture medium. Fluorescence was low and constant for several hours, both in PBS and in cell culture medium, but drastically increased after liposome burst indicating that FBS-containing media did not affect liposome integrity.



**Figure S5: DA liposomes (composed of 78 % of DAPC (C20:0), 20 % DOPE and 2 % NBD-PE) interact with WPMY-1 but not PC-3 cells.** Top: fluorescence microscopy images of WPMY-1 (left) or PC-3 cells incubated with DA liposomes. Bottom: Fluorescence intensity (CTCF) quantified on fluorescence images and normalized over the maximum fluorescence intensity value. Left: WPMY-1, right: PC-3 cells incubated with DL, DO, PO, DM, DP, DS and DA liposomes.



**Figure S6. Liposomes do not affect cell viability and show no cytotoxic effect.** a) Cell viability quantification, assessed on plated cells with the MTT assay 2h30 after liposome treatment. Results expressed as a percentage of cell viability relative to an untreated control corresponding to 100% viability. b) Cytotoxicity quantification, assessed on the same cells than (a) with the LDH assay. Results expressed as a percentage of cell death relative to an untreated control corresponding to 0% toxicity and a control treated with 10% Triton X-100 corresponding to 100% toxicity.