

Supplementary information:

PEG-lipid based micelles cause cholesterol efflux in Niemann Pick Type C-1 disease based lysosomal storage disorder

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Fig. S1 Characterization and efficacy of different liposomal nanocarriers with increasing PEG-lipid content.

Nanocarriers composed of differing ratios of HSPC and DSPE-PEG_{2k} were assembled in the presence of HPβCD. A. TEM images of 95 %, 50 % and 25 % HSPC (Scale bar represents 200 nm) of these materials were measured for size and morphology characterization. B. DLS of these set of liposomes was measured to characterize size and high-performance liquid chromatography (HPLC)-tandem mass spectrometry was performed to measure encapsulation efficiency of HPβCD. C. Nanocarriers containing HPβCD (0.5 mM) were exposed on *Npc1*^{-/-} for 24 hours, cells were fixed and filipin staining was performed to visualize reduction in lysosomal cholesterol.

Fig. S2 Effects of DSPE-PEG, HPBCD and their combination on cellular toxicity.

Npc1^{-/-} MEFs cells were treated with DSPE-PEG_{2k} micelles (0-500 μM) and HPβCD (0-10 mM). Cells were fixed and stained with deep red nuclear mask. The cell number was calculated as a function of different treatment groups. The combinations were relatively non-toxic, a significant decrease in cell number was only observed at the highest combination concentration.

Fig. S3 DSPE-PEG shows enhanced cholesterol clearance with longer PEG chain lengths.

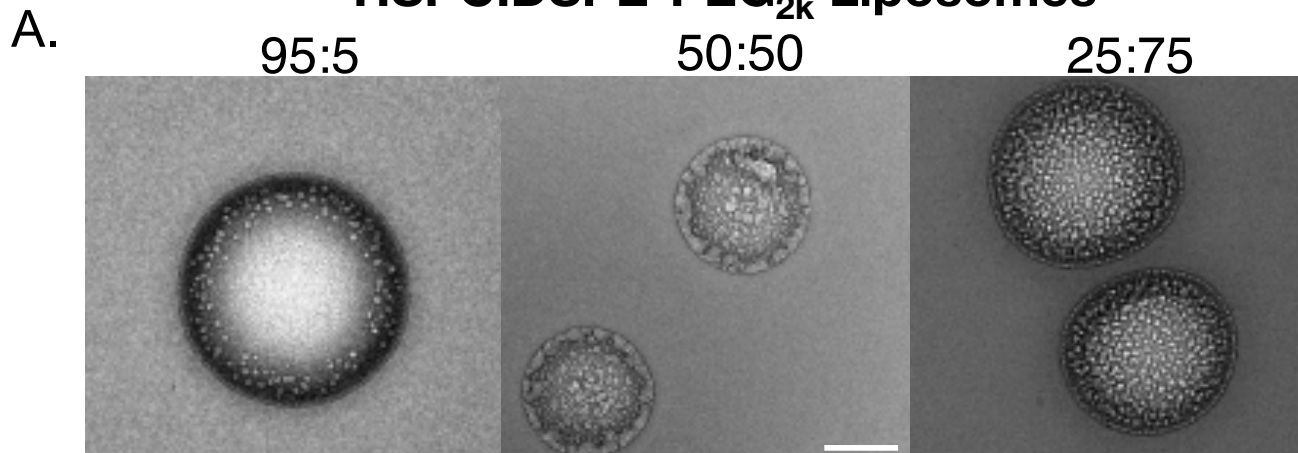
MEF *Npc1*^{-/-} cells were treated for 24 hour with 10 μM of DSPE-PEG₃₅₀, DSPE-PEG_{1k}, DSPE-PEG_{2k}, DSPE-PEG_{5k}, DSPE-PEG_{10k}, DSPE-PEG_{20k}, DSPE-PEG_{30k} and DSPE or PEG_{2k} (unconjugated), 16 images per well (n=3) were collected and representative images are shown.

Fig. S4 Effects of Pluronic block copolymer on cholesterol clearance.

We exposed two concentrations (1.5 mg/ml and 0.15 mg/ml) of Pluronic block copolymers-L64, P84, P103 and F127 on *Npc1* deficient cells for 24 hours. Cells were fixed and stained for filipin and imaged using the EVOS microscope. (n=5 images were captured).

Fig. S1

HSPC:DSPE-PEG_{2k} Liposomes



B.

Liposomal Composition	Average Size (DLS)	Loading Efficiency
95 % HSPC 5 % DSPE-PEG _{2k}	255 nm	22.9 %
50 % HSPC 50 % DSPE-PEG _{2k}	15.69 nm	35.7 %
25 % HSPC 75 % DSPE-PEG _{2k}	13.54 nm	30.9 %
100 % DSPE-PEG _{2k}	11.7 nm	10.4 %

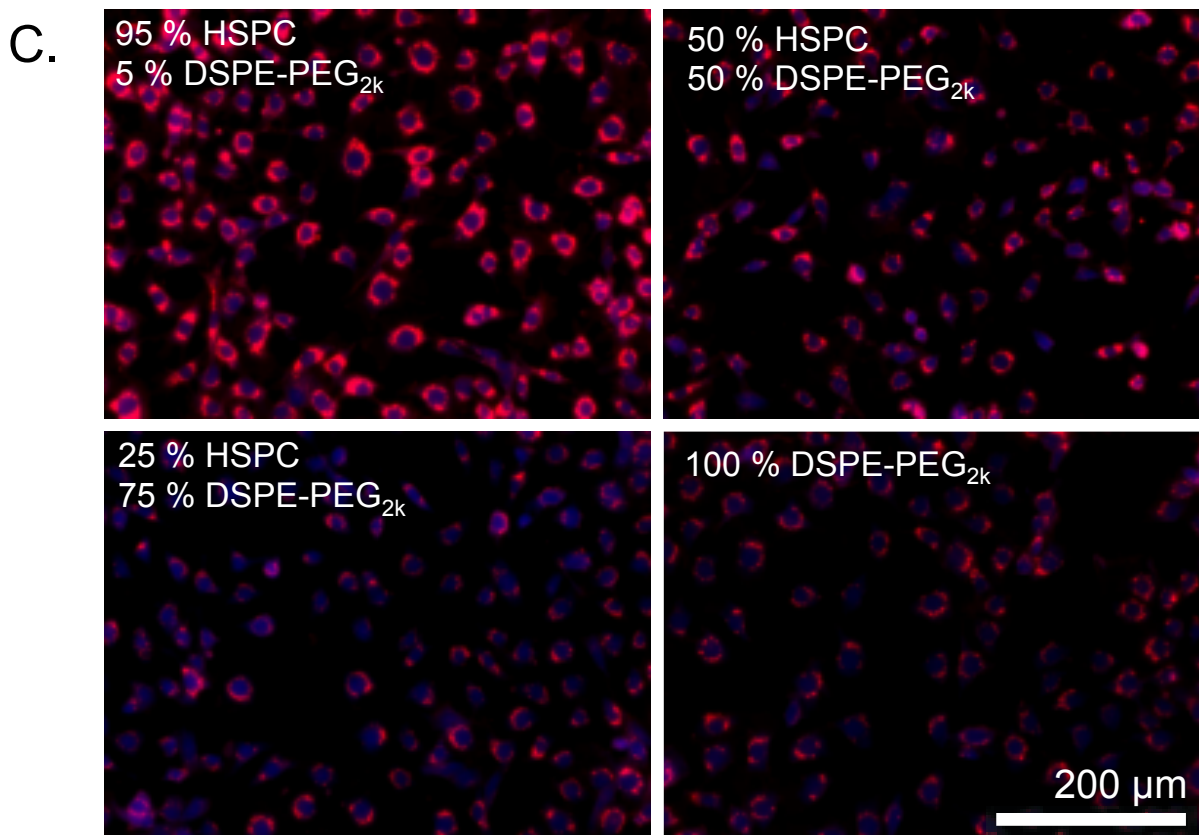


Fig. S2

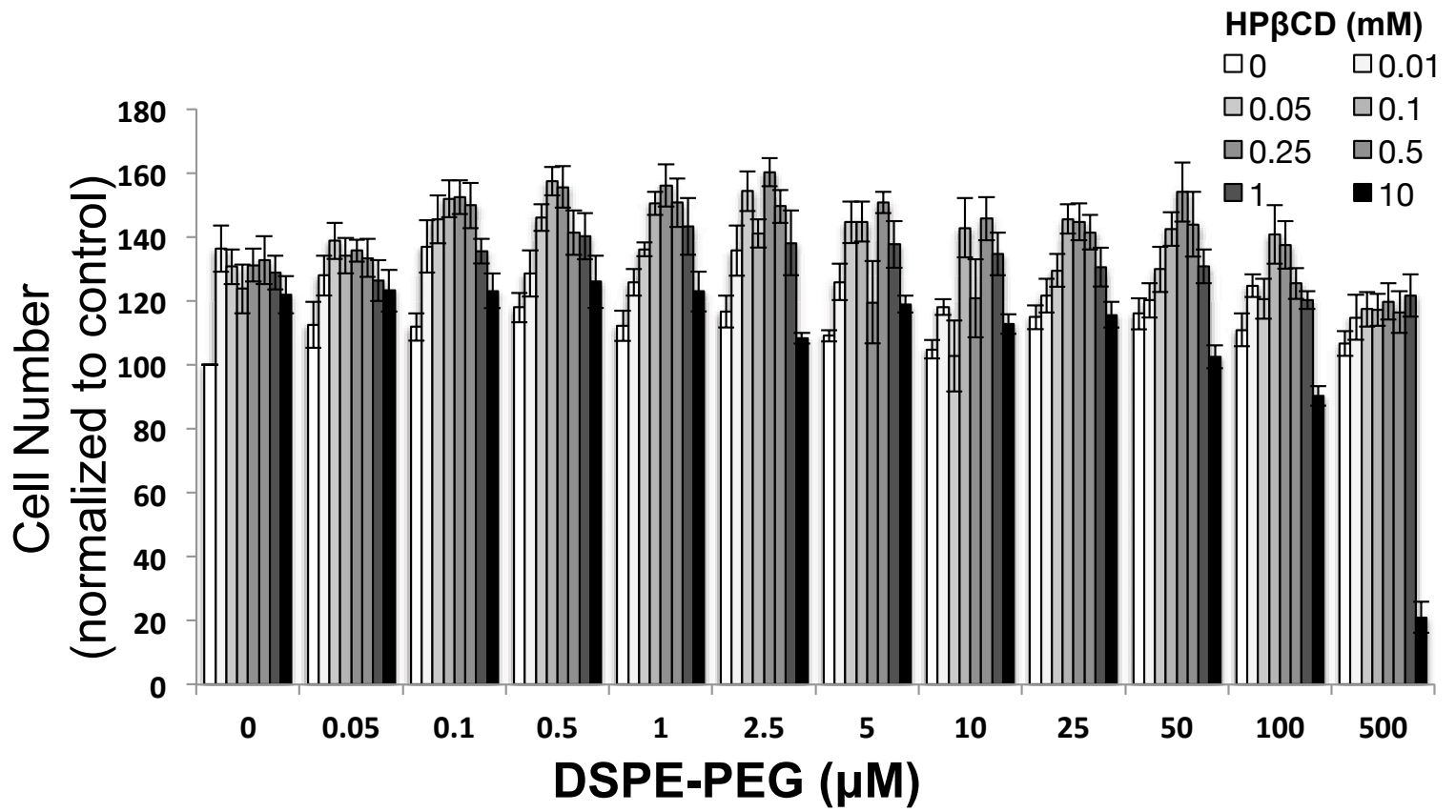


Fig. S3

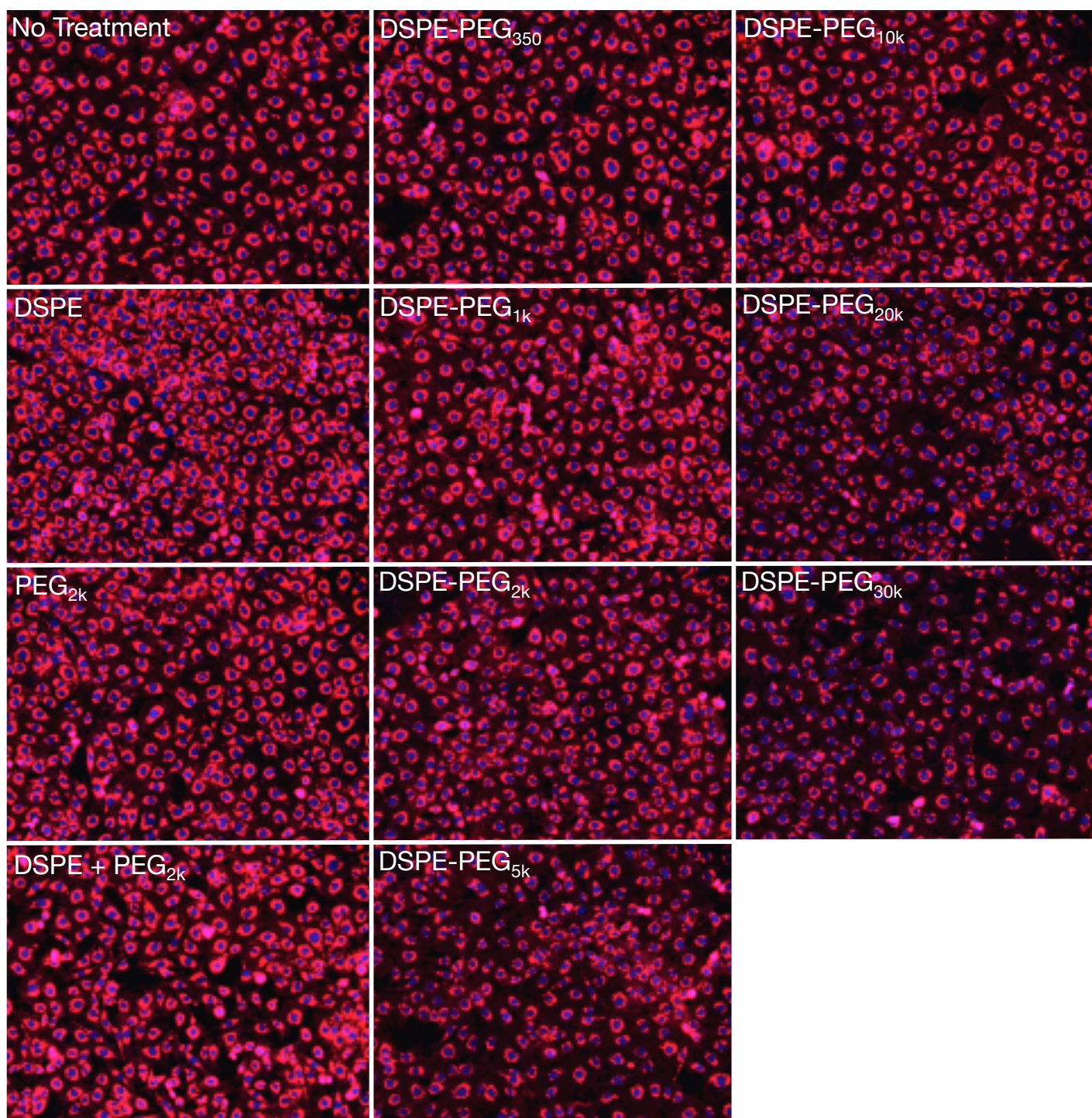


Fig. S4

