



Lipid nanoparticles for the treatment of inflammatory lung cancer in 3D and organ-on-chip models



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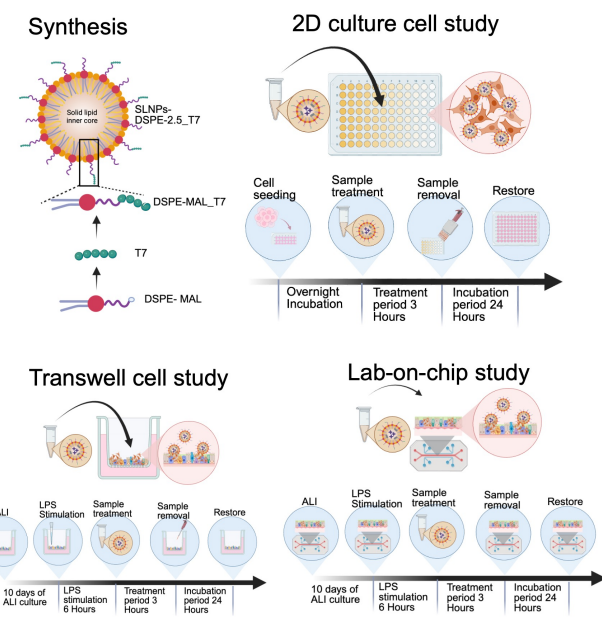
Abstract

Inflamed or high-inflammation lung cancer is a major driver of mortality, often correlating with advanced disease stages, poor prognosis, and reduced survival. Despite recent advances in cancer medicine, metastatic disease remains the main challenge for effective therapies. Nanoparticles such as solid lipid nanoparticles (SLNPs) offer many advantages in drug delivery due to their intrinsic properties, including improved bioavailability and the potential for selective targeting. To validate their potential, we use the 3D organ lab-on-a-chip to reduce reliance on preclinical models. The lab-on-a-chip is an advanced system for evaluating drug response under simulated human disease conditions. It simulates *in vivo* tumor microenvironment, aiding in the understanding of SLNPs' performance and tumor responses. Here, we designed SLNPs that are effectively targeted at cancer and inflamed epithelium.

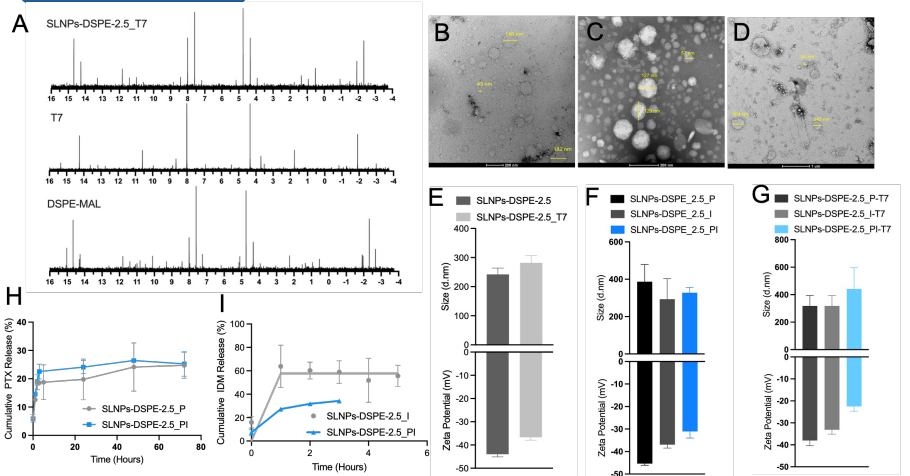
Introduction

Lung cancer comprises a heterogeneous group of malignancies that together constitute the leading cause of cancer-related mortality worldwide, with an estimated 1.8 million deaths annually [1]. The etiology of lung cancer is multifactorial, with tobacco smoking remaining the predominant risk factor, responsible for an estimated 85–90% of cases [2]. In these never-smokers, alternative risk factors such as exposure to radon gas, environmental air pollution, occupational carcinogens (e. g., asbestos, arsenic, silica), and genetic predisposition play a more prominent role [3]. Nanotechnology and novel drug delivery systems, such as inhalable extracellular vesicles-based carriers for chemotherapeutics like paclitaxel, are being explored to enhance tumor-specific drug delivery while minimizing systemic toxicity. In the context of minimizing reliance on preclinical models, lab-on-chip models are a valuable alternative. Importantly, because these vascularized lab-on-a-chip models contain an endothelium-lined channel that supports dynamic fluid flow, potential therapeutics can be tested in a pharmacologically relevant context in which drugs pass through a mucosal barrier that mimics the physiological condition.

Methods

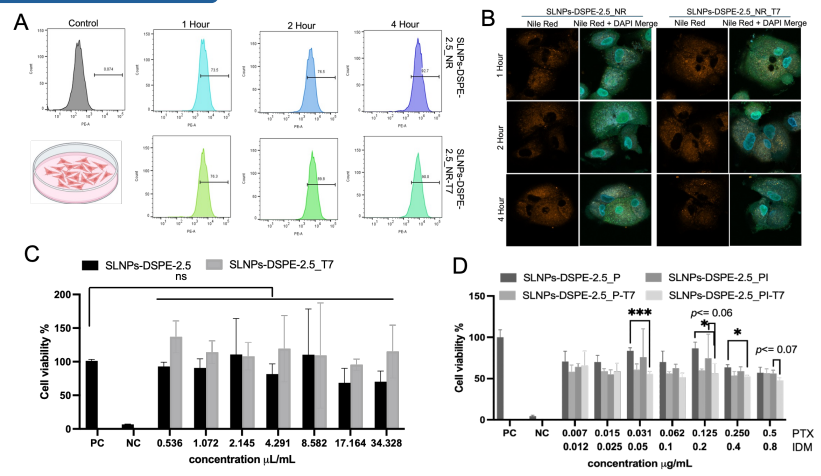


Synthesis



Physicochemical characteristics. A) NMR analysis of T7 conjugation for SLNPs-DSPE-2.5_T7. TEM images B) SLNPs-DSPE-2.5_PI, C) SLNPs-DSPE-2.5_T7, and D) SLNPs-DSPE-2.5_PI-T7. E-G) Dynamic light scattering (DLS) size and surface zeta potential. The cumulative drug release study H) describes the release of PTX from single-loaded SLNPs-DSPE-2.5_P and dual-loaded SLNPs-DSPE-2.5_PI. I) describes the release of IDM from single-loaded SLNPs-DSPE-2.5_IDM and dual-loaded SLNPs-DSPE-2.5_PI.

2D culture cell study



Assessing uptake of SLNPs-DSPE_Nr by the PE channel at different time intervals. A) Histogram analysis of FACS. B) Confocal analysis of GFP-expressed A549. Cell viability from the crystal violet assay. C) Describes the cell viability of the controls (SLNPs-DSPE-2.5 and SLNPs-DSPE-2.5_T7). F) Cell viability of PTX against GFP-expressing A549. Statistical analysis through One-way ANOVA and t-test, * p < 0.03, ** p < 0.01, *** p < 0.001. Positive control (PC) and negative control (NC).

Conclusion

SLNPs-DSPE-2.5_T7 is a transferrin-targeted SLNP that is highly accumulated at cancerous and inflamed epithelium. SLNPs-DSPE-2.5_T7 carry a combination of therapeutic payload of PTX and IDM. Further, we tested its therapeutic potential in inflamed HBE/A549 using a transwell/Lab-on-chip. By using a mucosal administration approach with a human NSCLC cell line within the lab-on-chip, we were able to recapitulate organ microenvironment-specific cancer behaviors, including rampant growth in the lung microenvironment, as well as tumor responses to therapy *in vitro*. These studies demonstrate the engineering of *in vitro* human orthotopic cancer models with clinical relevance.

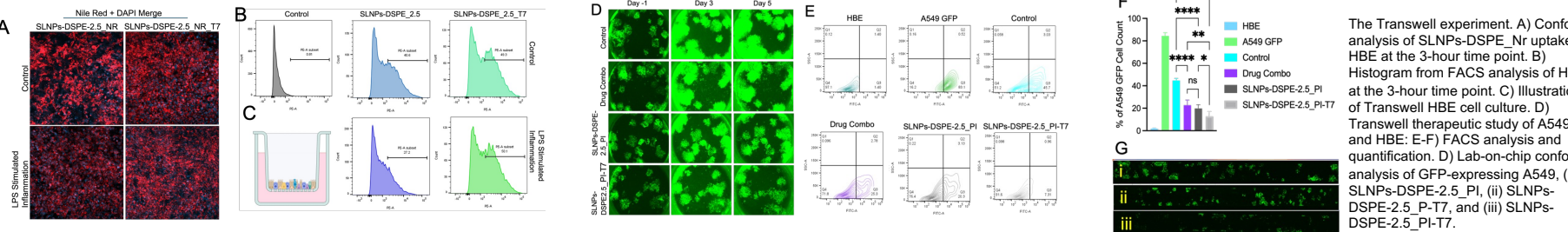
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Transwell/Lab-on-chip cell study



The Transwell experiment. A) Confocal analysis of SLNPs-DSPE_Nr uptake in HBE at the 3-hour time point. B) Histogram from FACS analysis of HBE at the 3-hour time point. C) Illustration of Transwell HBE cell culture. D) Transwell therapeutic study of A549 and HBE. E-F) FACS analysis and quantification. G) Lab-on-chip confocal analysis of GFP-expressing A549, (i) SLNPs-DSPE-2.5_PI, (ii) SLNPs-DSPE-2.5_P-T7, and (iii) SLNPs-DSPE-2.5_PI-T7.