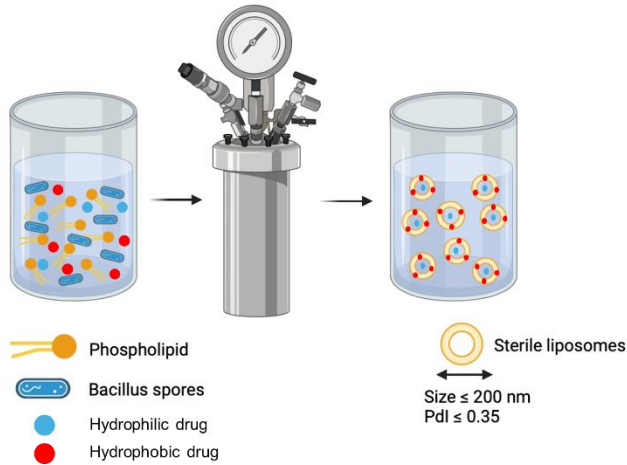


## One-step, organic solvent-free, sterile liposome production



### KEY ACHIEVEMENTS

Single-step production of sterile liposomes of different lipid composition, compatible with drug delivery:

- Size < 200 nm
- Pdl < 0.35
- SAL  $\leq 10^{-6}$
- Low (5 mM) or high (45 mM) lipid concentration
- Efficient (co)-encapsulation of hydrophobic and hydrophilic small molecules.

### KEY COMPETITIVE ADVANTAGES

- One-step, reducing production costs and facilitating scaling-up in GMP conditions.
- Solvent-free.

### ONGOING ACTIVITIES

- Efficacy of liposomes on animal models of pulmonary inflammatory diseases.
- Process transfer on a larger scale in a clean room.
- Production of liposomes and lipid nanoparticles encapsulating nucleic acids.

### KEY PUBLICATIONS

- Penoy N et al. Int J Pharm. 2024 Feb 15;651:123769
- Penoy N et al. Int J Pharm. 2022 Nov 5;627:122212

Thin film hydration, detergent removal, solvent injection, reverse phase evaporation, and emulsion are conventional liposome preparation methods. They involve complex and numerous steps and therefore scalability, reproducibility, and sterility are difficult to obtain. They also require large organic solvent volumes.

Supercritical CO<sub>2</sub> (SC CO<sub>2</sub>) is a promising alternative due to its solvent and microbicidal properties. It allows one-step liposome preparation with a better control of the physicochemical properties, improving scalability, not requiring the use of organic solvents.

Therefore, we developed a Particle from Gas Saturated Solution (PGSS) process using a Quality By Design (QbD) strategy. Using this method, we produced liposomes compatible with drug delivery with two different set of parameters (both for 30 mins at 80°C):

Lipid (mM)	concentration	Volume (mL)	Pressure (bar)
45		14	240
5		10	156

In both cases, sterility assurance level (SAL) of produced liposomes was  $< 10^{-6}$ , showing that the sterilization process is efficient.

Finally, using this method we produced liposomes containing hydrophilic budesonide and hydrophobic salbutamol with high encapsulation efficiencies, similar or higher to that obtained with the conventional method TFH (89±7% and 52±2% respectively).

### PARTNERSHIP SOUGHT

- Collaboration agreements for product and method development.
- Licencing agreements.
- GMP/GLP grade batch production on demand.