# Solvent-free nanoparticles for encapsulation of water- soluble compounds

CARLOTTA MATTIODA, XXXVII Cycle DIMEAS, Dipartimento di Ingegneria Meccanica e Aerospaziale, Politecnico di Torino

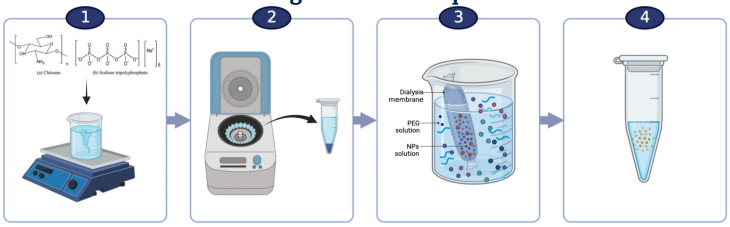
## INTRODUCTION

The increasing societal concerns regarding sustainable practices with lower environmental impact have put pressure on the pharmaceutical industry[1] to focus on greener formulation and synthesis techniques. For instance, the use of **safer solvents** is identified as a clear priority towards sustainable chemical processes. In this endeavour, the introduction of innovative, **solvent-free techniques** is a key step towards greener production of pharmaceuticals[2]. Nanoparticles (NPs) have attracted increasing attention as drug delivery systems, since they can protect the active principle, enhance its transport, and reduce the undesired effects[3].

**This PhD project aims at the development of innovative synthesis techniques to prepare NPs, that will replace organic solvents with water-based solutions.**

## MATERIAL AND METHODS

### Ionic gelation technique



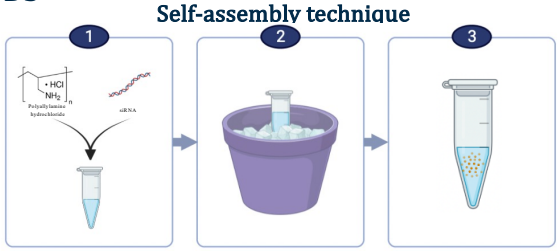

Mix chitosan (CS) and sodium tripolyphosphate (TPP) and stir for 60 min at RT

Centrifuge for 5 min at 6000 rpm and collect the supernatant

Dialysis of the supernatant to concentrate the NPs solution

Collect the NPs

### Self-assembly technique



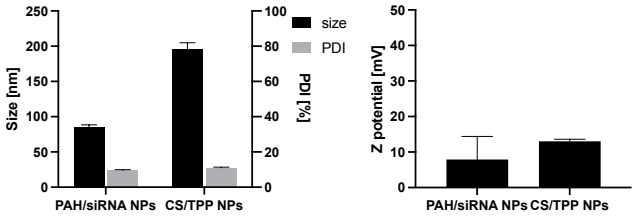

Mix Polyallylamine Hydrochloride (PAH) and siRNA (N/P=2) in water

Let the NPs solution stabilize for 30 min at 4°C

Collect the NPs

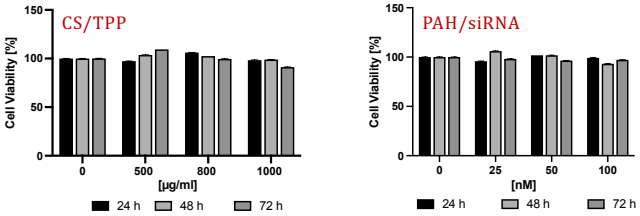
## RESULTS

### NPs characterization



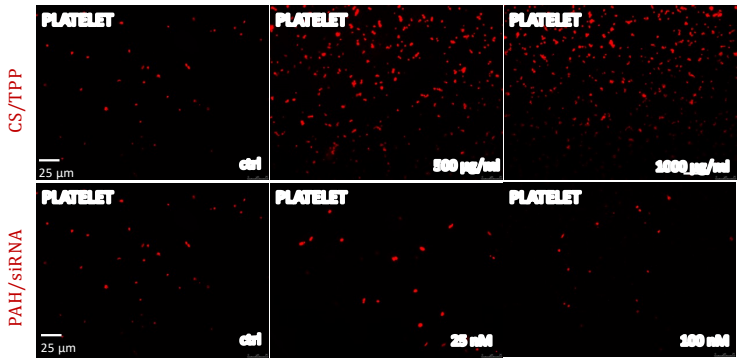

CS/TPP NPs have a size in the 200 nm range, a low polydispersity index (0,25) and positive surface charge (+13 mV).
PAH/siRNA NPs have a size in the 90 nm range, a low polydispersity index (0,24) and positive surface charge (+9 mV).

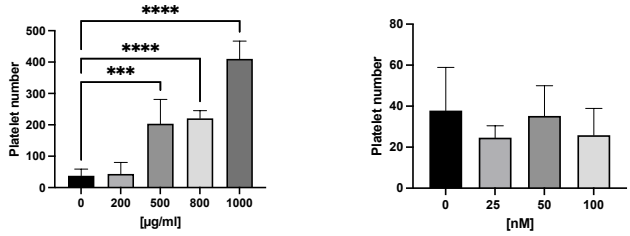
### Viability tests




Cell viability tests were performed on human fibroblasts (Hff-1) treated with empty CS/TPP NPs and PAH/siRNA NPs.
Results show **no evident signs of toxicity for the bare NPs** at any of the tested concentrations.

### Platelet activation tests




Platelet activation was measured by counting the number of adherent platelets after treatment with NPs . The results show that CS/TPP NPs cause high platelet activation at concentrations above 200 µg/ml.
PAH/siRNA NPs did not show signs of platelet activation.

## CONCLUSIONS

Two **solvent-free NPs** synthesis techniques were optimized. **Non-toxic** NPs with **homogenous size distribution and positive surface charge** were obtained with CS and PAH. PAH/siRNA NPs did not trigger platelet activation, supporting their further application in platelet-assisted drug delivery. Future work will focus on: i) surface modification of PAH/siRNA NPs with platelet anchoring ligand to favour reinternalization within platelets (**in cooperation with CSTS healthcare**); ii) development of a metastatic melanoma in vitro model (**in collaboration with University of Newcastle**); iii) drug loading with therapeutic antibodies and siRNAs for melanoma treatment.

## HARD SKILLS (66/100)

- **From science to business: how to get technology out of laboratories -** The course summarizes the main concepts for the technology transfer
- **Advanced therapies (nanomedicine, gene and cell therapy) in surgery -** The course describes the synthesis and characterization of nanostructured systems
- **Research and Nanomedicine 2022, V Edition -** Nano-therapeutics systems and regenerative medicine workshop
- **Approcci innovativi in ambito cosmetico, in ottemperanza con la regolamentazione Europea -** The course is about alternative methods to *in vivo* experiments

## SOFT SKILLS (30/40)

- **Navigating the hiring process: CV, tests, interview**
- **Research integrity**
- **Responsible research and innovation, the impact on social challenges**
- **Thinking out of the box**
- **Time management**
- **Writing Scientific Papers in English**

REFERENCES
1) Milanesi, M. et Al. Journal of Cleaner Production 261 (2020).
2) Pena-Pereira, F. et Al. Green Chemistry 17 (2015).
3) Wilczewska, A. Z. at Al. Pharmacological Reports 64 (2012)

Tutors: Gianluca Ciardelli, Jacek Tuszynski, Clara Mattu

PhD Day 2022
25th October 2022, Dental School, Torino