Microfluidic droplet-based synthesis of polymeric nanoparticles as controlled drug delivery systems

Emanuele Mauri [a], **Emanuele Limiti** [a], **Sara M. Giannitelli** [a], **Pamela Mozetic** [b], **Marcella Trombetta** [a], **Alberto Rainer** [a,b]

[a] Department of Engineering, Campus Bio-Medico di Roma, via Alvaro del Portillo 21, 00128 Rome, Italy

[b] Institute of nanotechnology (NANOTEC), National Research Council, via Monteroni, 73100, Lecce, Italy

e.mauri@unicampus.it

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Introduction

- The design of polymer nanoparticles (NPs) plays a leading role in the definition of innovative strategies for nanomedicine[1]
- Several batch techniques for NP synthesis have been proposed to combine different polymers through the orthogonal activation of their inner chemical groups and produce engineered nanocarriers for controlled drug delivery purposes.
- However, the clinical translation of NPs is still challenging due to significant limitations occurring in the conventional reaction routes

Conventional batch synthesis

- scarce control over reaction parameters
- limited NP monodispersity
- limited reproducibility
- difficult control over NP physicochemical properties
- use of large volumes of chemicals and therapeutics

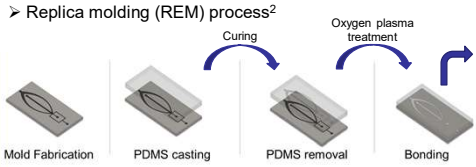
→ **MICROFLUIDIC PLATFORMS**

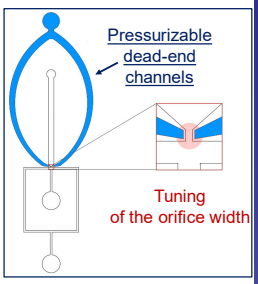
- Scalable yields
- High throughput
- Efficient tunable pharmacokinetics

Materials & Methods

1. CHIP FABRICATION

- Replica molding (REM) process[2]

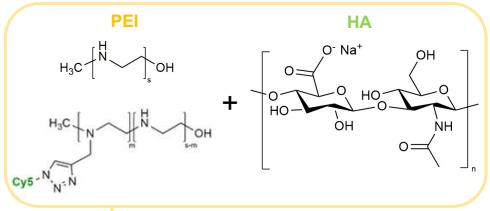


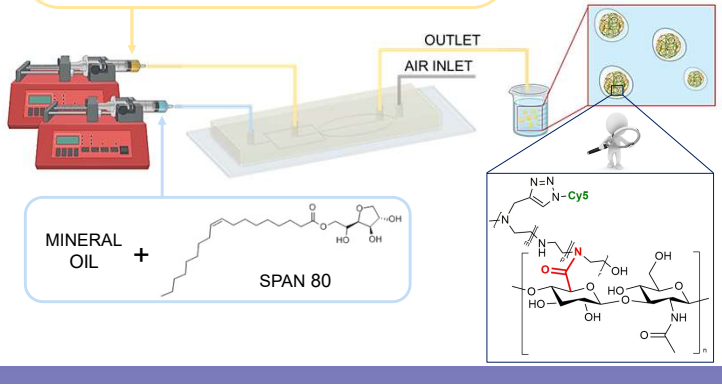


- Chip endowed with a valve-based flow focusing geometry

2. NPs SYNTHESIS

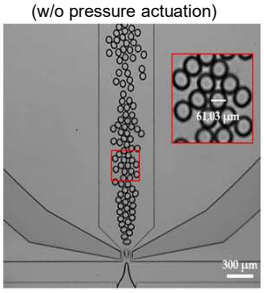
- Microfluidic droplet-based generation[3] with pressure microactuation



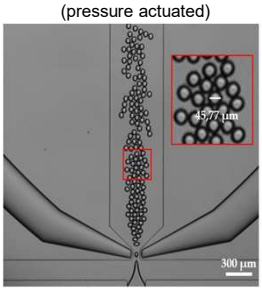


Results: NPs size & drug release

CHIP 0 BAR (w/o pressure actuation)

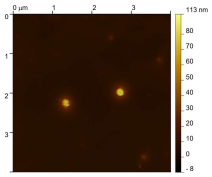


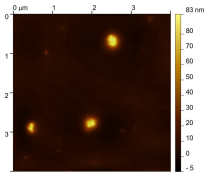
CHIP 2 BAR (pressure actuated)



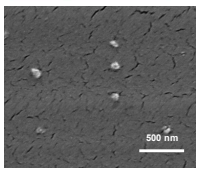
DLS analysis

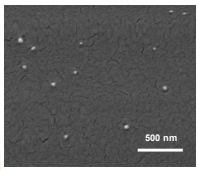
	Diameter (nm)	PDI (-)	z-potential (mV)
NPs @ chip 0 bar	188.3	0.011	- 2.22
NPs @ chip 2 bar	92.4	0.022	- 2.13





AFM analysis





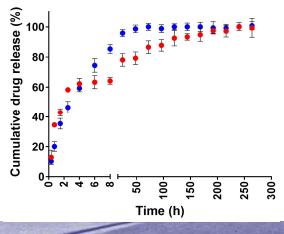
SEM analysis (dehydrated specimens)
NPs @ chip 0 bar → 92 nm
NPs @ chip 2 bar → 58 nm

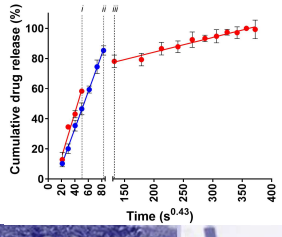
NPs present swelling behaviour: **NANO-HYDROGELS**

***In vitro* drug release profiles**

Different drug release kinetics
NPs @ chip 0 bar → up to 3 days
NPs @ chip 2 bar → up to 10 days

Different diffusion regimes with different slopes: the transition and duration of the regimes depend on NPs size and drug-polymer interactions (*i.e., drug adsorption or aliphatic-aromatic stacking*)

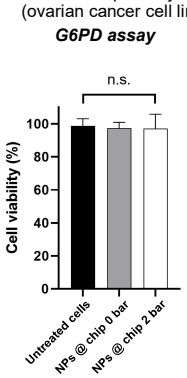


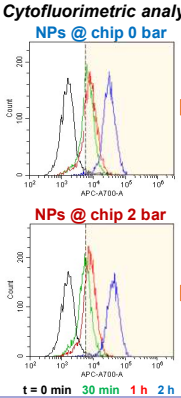


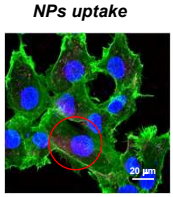
Results: biocompatibility

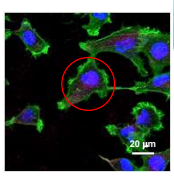
- The biocompatibility and internalization of NPs was evaluated using **OVCA-433 cell line** (ovarian cancer cell line)

G6PD assay ***Cytofluorimetric analysis*** ***NPs uptake***









Efficient NPs internalization already after 30 min (> 50%) without toxic effects

Conclusion

- The proposed pressure-actuated chip enables the active tuning of the flow focusing geometry, modulating the diameter of the microdroplets and, as a result, the NP size.
- The NPs are characterized by a **very low polydispersity index** (PDI), **homogeneous morphology** and **high lot-to-lot reproducibility**, which highlight the potential of the developed microfluidic strategy to provide highly controllable and scalable production yields. Moreover, the NPs do not generate toxic effects in cellular environment, suggesting their promising application as nanocarriers for drug delivery, in several healthcare scenarios.
- The pressure-actuated microfluidic platforms ensure the continuous in-flow production of NPs, promoting a possible scale-up that could support the progress toward the clinical translation of technology.

References

[1] E. Mauri et al., *Gels* 2021, 7, 36

[2] M. Gori et al., *Biotechnol. Bioeng.*, 2021, 118, 142-152

[3] S. Sohrabi et al., *RSC Adv.*, 2020,10, 27560-27574