Production of nanoliposomes using *in-house* 3D printed microfluidic chips

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BACKGROUND

Recently, 3D printing (3DP) and microfluidics (MFs) have gained a predominant role in the field of nanomedicine. These techniques are suitable for the production of innovative formulations, such as lipid nanoparticles (LNPs). 3DP enables customisation of the geometry of the device to be produced with very high precision of the internal geometry.[1] The MF technique, on the other hand, makes it possible to automate the production of nanoformulations, eliminating the batch-to-batch variability associated with classical synthetic methods.[1,2] Both techniques guarantee low production costs and fast processing times. The aim of this project was to create a constructive synergy between 3DP and MFs in order to obtain LNPs in a simple, fast and highly reproducible manner. Hence, three devices were designed and printed *in-house,* and subsequently tested for the *in-flow* production of LNPs.

MATERIALS AND METHODS

The devices designed using TinkerCad and printed using an Asiga 3DP.

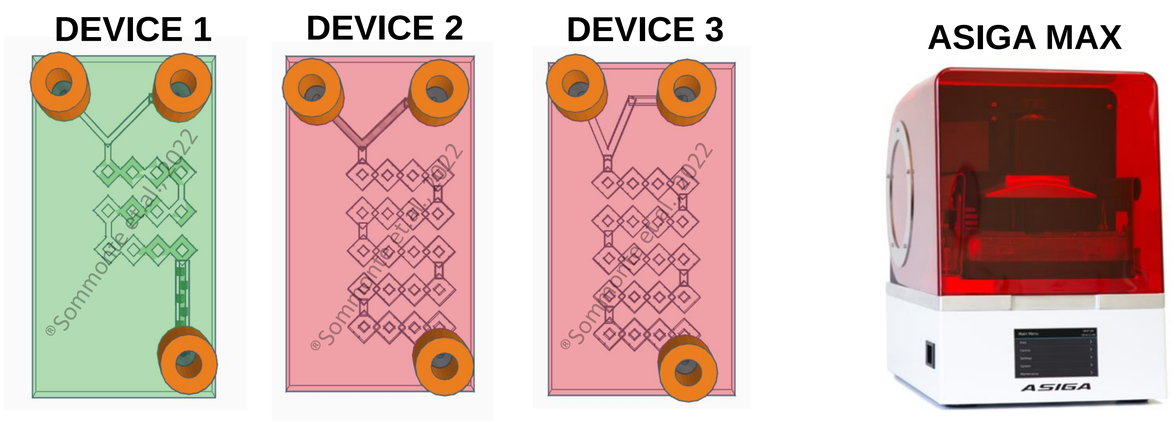
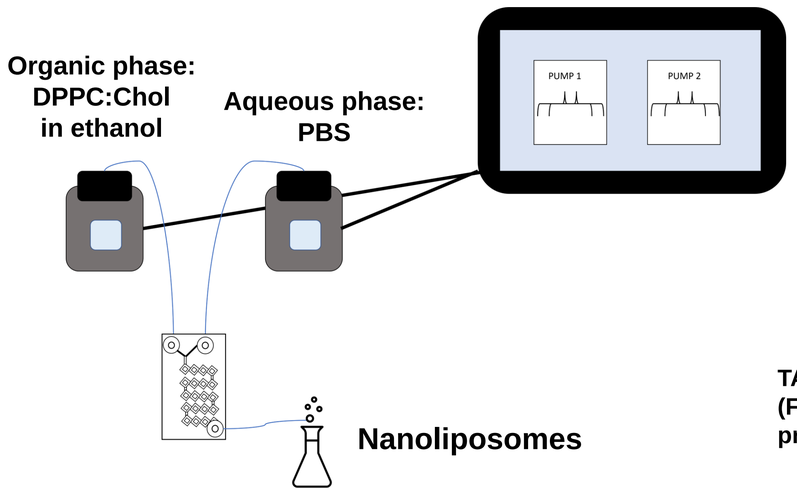


FIGURE 1: Schematic representation of each device..

Empty LNPs were produced using the microfluidic system.



FRRs
5:0.50
5:1
3:1
5:2
5:3

TABLE 1: Summary of fluid rate ratios (FRRs) used for each device in the LNP production process.

The particle size, polydispersity index (PDI), and Z-potential was measured.

RESULTS

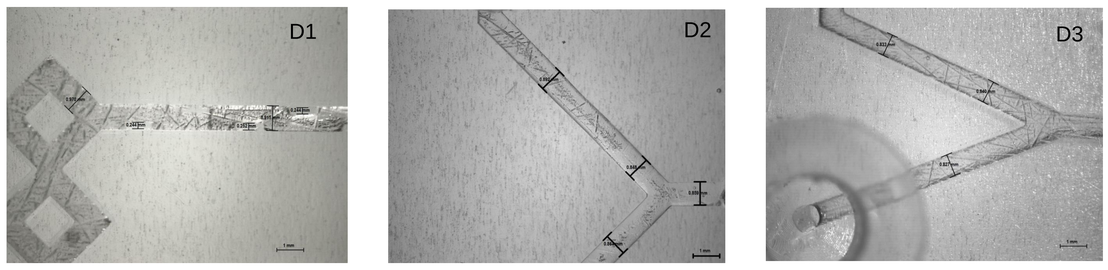


FIGURE 2: Optical microscope images (8x) of devices.

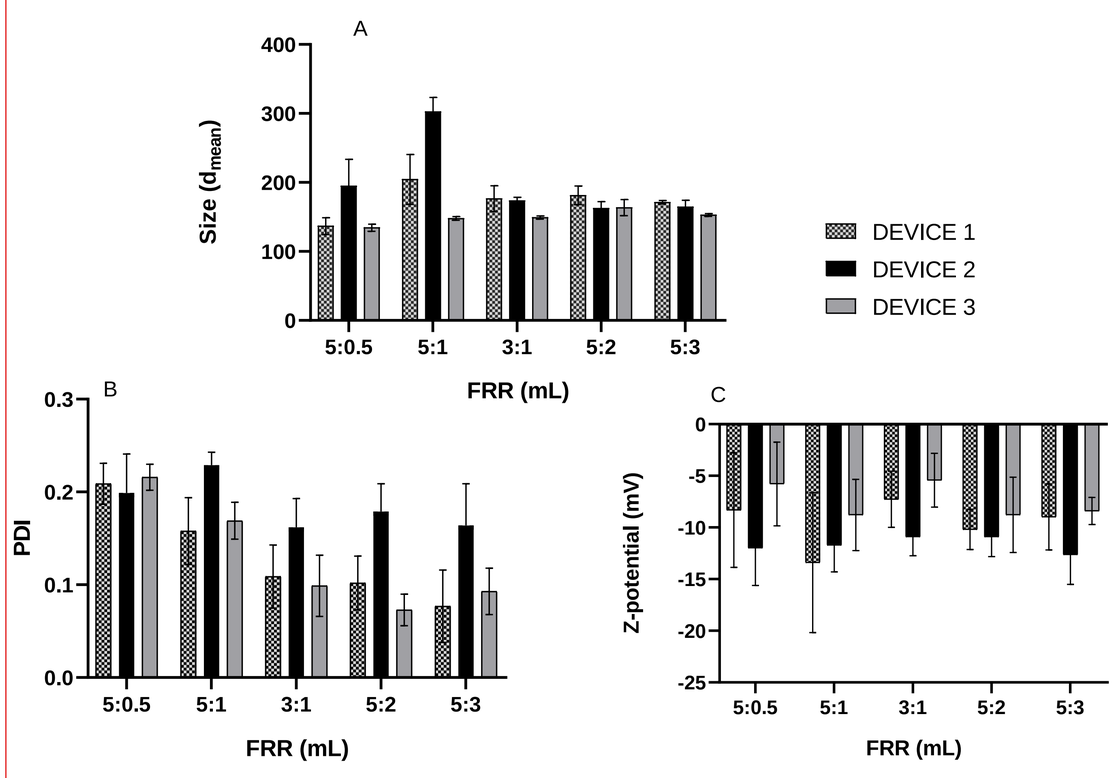


FIGURE 3: Graphical representation of LNP properties by each device and FRRs. Comparison in size (3A), Polydispersion Index (PDI, 3B) and Z-potential (3C).

DISCUSSION

- The 3D-printed chips were specially designed to improve mixing between the aqueous and lipid phase, with all devices equipped with square channels.
- DEVICE 1 possesses an internal geometry characterised by a fast-mixing mode phase and a linear channel with wedges inside, while DEVICE 2 and DEVICE 3 share the same internal geometry but a different angle of meeting point between the two phases.
- The presence of an interconnected internal geometry made it possible to create a passive chaotic advection.
- The printed devices are able to tolerate total flow rates of >10 mL min^{-1}, exceeding the mechanical resistance of many commercial ones.
- For each device various FRRs were tested. All different device geometries and operating conditions produced extremely promising results.
- The formulated liposomes were less than 200 nm in size with a PDI < 0.25. The ζ- potential is negative and stable, ranging between -14 and -8 mV.

CONCLUSIONS AND FUTURE PERSPECTIVES

This work, showed promising data and further studies are currently underway to develop a protocol suitable for drug encapsulation in nanoformulations obtained by 3D printed MF devices. The advantages of 3DP and MFs are well known in science, but the great innovation lies in the synergy between the two techniques. In this way, by producing the devices in-house and applying them by means of MFs, it is possible to decide the type of geometry to be printed with high precision and to reduce production time and costs.

References

[1] G. Ballacchino, E. Weaver , E. Mathew , R. Dorati, I. Genta, B. Conti and D. A. Lamprou. Manufacturing of 3D-Printed Microfluidic Devices for the Synthesis of Drug-Loaded Liposomal Formulations, Int. J. Mol. Sci. **2021**, 22, 8064.
[2] E. Jaradat, E. Weaver, A. Meziane, D. A. Lamprou. Microfluidics Technology for the Design and Formulation of Nanomedicine, *Nanomaterials* **2021**, *11*(12), 3440.