PriMiSpheres

6 months internship proposal

Subject: Sustained release of therapeutic protein: formulation of porous polymer microspheres using the prilling process coupled with temperature-induced phase separation. (PriMiSpheres project)

Keywords: Extended protein release; Prilling; Porous microspheres

Context: Knowledge of protein structure and function has led to their use in human medicine. The development of delivery systems, such as microspheres, to enhance their action is a major challenge, given the fragility of these molecules. Continuous and complete release of the encapsulated protein relies in particular on control of the properties of the carrier system (diameter, porosity, hydrophilic/lipophilic balance of the biocompatible polymer, …) [1].
In this context, the "PriMiSpheres" project aims to develop an original process for formulating porous microspheres (~50µm) of biodegradable polymer (PLLA or PLGA) for continuous protein release, by coupling several processes already studied separately at the MINT laboratory.

The proposed process comprises five steps: *(i)* Preparation of an organic phase comprising the active molecule, the polymer of interest (PLLA or PLGA) and a solvent; *(ii)* Extrusion of this phase through a vibrating nozzle to form monodisperse droplets (Prilling process); *(iii)* Reception of the droplets in a "cold" environment (temperature below the solidification temperature of the solvent) to induce phase separation within the solidified droplets (TIPS or Thermal Induced Phase Separation process); (iv) Extraction of the solvent by immersion in an aqueous or ethanolic solution, while preserving the porous structure of the polymer; (v) Extraction of the residual solvent by supercritical CO2 (CO_2sc) to obtain a microsphere powder. Finally, the environmental impact of the produced drugs will be evaluated whether through the assessment of their carbon impact (Ecovamed database [7]) or their pollutant impact (hazard score [8]).

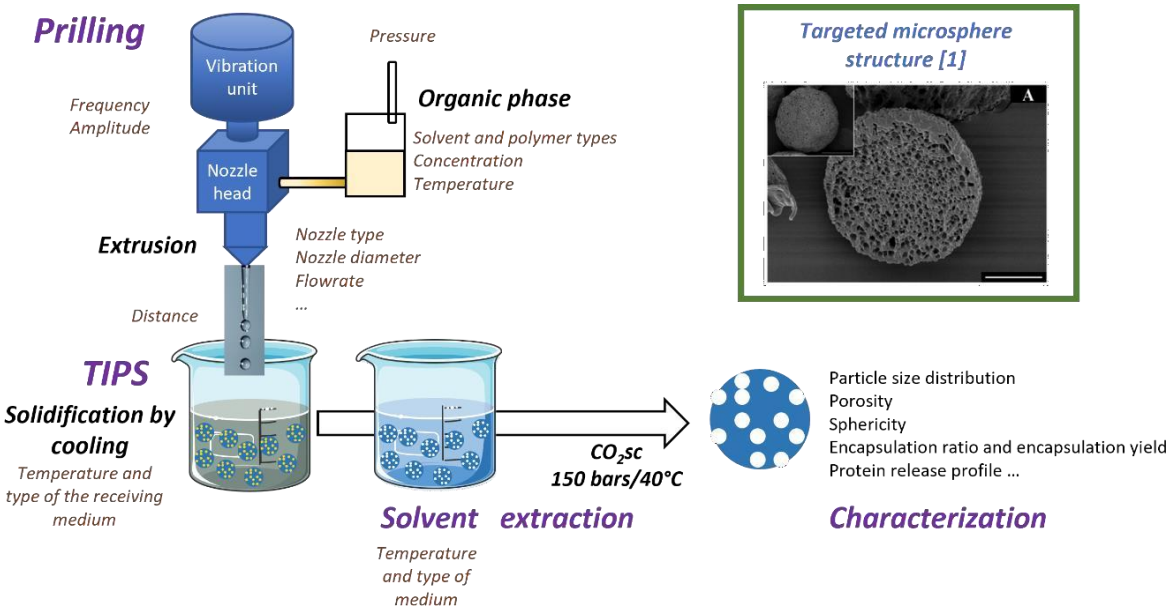

Figure 1: graphical abstract of the planned process.

A prilling process has already been developed by MINT for the production of PLGA microparticles (30 to 100 µm) with controlled size and sphericity (VT Tran [2], F. Violet [3], and T. Nguyen-Pham [4] theses), but the particles obtained did not have a porous structure. The TIPS process coupled with CO_2sc drying was developed by MINT as part of S. Gay's thesis [5] to develop low-density microcellular PLLA matrices with modifiable structural properties. It has been demonstrated that the CO_2sc drying process reduces environmental impacts by a factor of four on average compared to freeze-drying [6]

Internship objectives: within the scope of the PriMiSpheres project, the aim of this internship will be to focus on the first step of this innovative process development, which is the organic phase and process phenomenology characterization. The dynamic viscosity and density of the organic phase will be characterized. Phase diagrams will be drawn up for different polymer/solvent pairs, in the presence or absence of surfactants. The optimum conditions for jet formation (length, size and break-up) will be determined using a high-speed camera. Finally, the rheological and physicochemical properties of the organic phase/receiving medium interface will be determined.

For this purpose, different critical parameters will be optimized, for example:
- For the organic phase: the choice of polymer solvent (glycofurol or a class III solvent such as DMSO), the possible addition of a surfactant and/or an anti-solvent to modulate particle pore size.
- For prilling: nozzle type and diameter, vibration frequency and amplitude, flow rate, nozzle-receiving medium distance, etc.
- For phase separation and solvent extraction: the nature and temperature of the receiving medium, with the aim of preserving the spherical shape and porosity of droplets after impact with the surface of the medium, etc.

The choices made will have to take into account the feasibility of the process on an industrial scale in the context of good manufacturing practice.
In addition to process development, the environmental and societal impact of the process and the finished product will have to be considered, for example by assessing the hazard score (formerly known as the PBT index for Persistence, Bioaccumulation and Toxicity).

Profile: The candidate should hold a Master 2 or engineering degree (process engineering, chemical engineering, physics, etc.), have a satisfactory level of English and be organized and rigorous. She/he must also show a strong interest in the healthcare field; a specialization or double degree in pharmaceutical sciences would be a plus. You will be required to draft reports, write scientific publications and give oral presentations on your research work. Written and spoken fluency (English and French) would be appreciated.

The person recruited will have access to the prilling (Spherisator®) and CO2sc pilots already present in the laboratory, as well as to the equipment needed to characterize the microspheres obtained, some of which is available in the laboratory, on the platforms of the University of Angers or through collaborations or services (SEM, XRD, UPLC, etc.).

Sources:

[1] M.Q. Le, J.C. Gimel, X. Garric, T.Q. Nguyen-Pham, C. Paniagua, J. Riou, M.C. Venier-Julienne, Modulation of protein release from penta-block copolymer microspheres, 2020, European Journal of Pharmaceutics and Biopharmaceutics, 152, 175-182

[2] V.T. Tran, J.P. Benoît, M.C. Venier-Julienne, Why and how to prepare biodegradable, monodispersed, polymeric microparticles in the field of pharmacy?, 2011, International Journal of Pharmaceutics , 407 (1), 1-11

[3] M.Q. Le, F. Violet, C. Paniagua, X. Garric, M.C. Venier-Julienne, Penta-block copolymer microspheres: Impact of polymer characteristics and process parameters on protein release, 2018, International Journal of Pharmaceutics, 535 (1), 428-437

[4] T.Q. Nguyen-Pham, L. Benyahia, G. Bastiat, J. Riou, M.C. Venier-Julienne, Behavior of poly(d,l-lactic-co-glycolic acid) (PLGA)-based droplets falling into a complex extraction medium simulating the prilling process, 2020, Journal of Colloid and Interface Science, 561, 838-848

[5] S. Gay, B. Calvignac, L. Ouanssi Kamtcheu, T. Beuvier, E. Boller, M. Sauceau, S. Del Confetto, F. Boury, A. Gibaud, Synchrotron X-ray *In Situ* Tomography of Thermally Induced Phase Separation of Polylactic Acid in 1,4-Dioxane Solution, 2018, Crystal Growth & Design 18 (12), 7496-7503

[6] S. Gay, G. Lefebvre, M. Bonnin, B. Nottelet, F. Boury, A. Gibaud, B. Calvignac, PLA scaffolds production from Thermally Induced Phase Separation: Effect of process parameters and development of an environmentally improved route assisted by supercritical carbon dioxide, 2018, The Journal of Supercritical Fluids, 136, 123-35

[7] Ecovamed, https://www.ecovamed.com

[8] B. Dupont, S. Faure. Le hazard score, un outil pour réduire l'impact environnemental des prescriptions. Actualités Pharmaceutiques, 2020, 59, 27-32.