CONTINUOUS REGIME MICROFLUIDIC SYSTEM FOR NANOCAPSULES GENERATION

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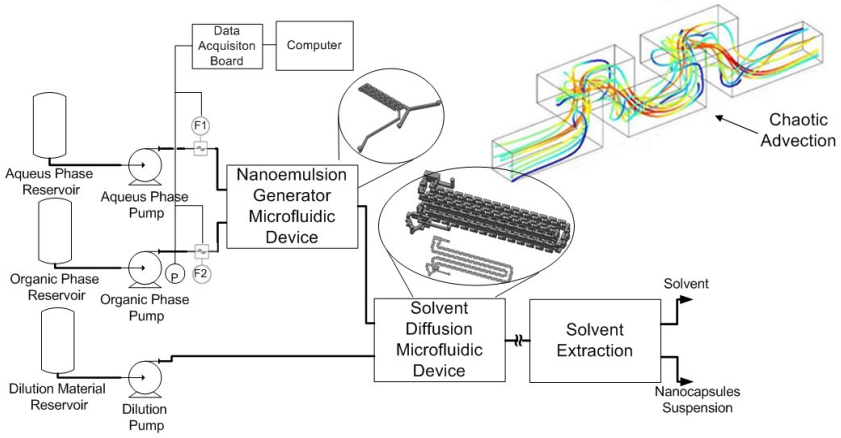
Abstract: *This research aims to develop a microfluidic system for chemical process miniaturization for nanocapsules generation. The emulsification, diffusion and solvent extraction/evaporation process was selected as target for miniaturization. LTCC was the selected substrate. Experiments showed the viability of controlling nanocapsules sizing by means of total flow rate through devices. Generated nanocapsules size varied between 790,5 nm ± 36,9 nm and 209,7 nm ± 3,5 nm with a polydispersity index (pdi) between 0,313 ± 0,016 and 0,09 ± 0,017 when flow rate varied between 90 mL/min and 293 mL/min. Other experiments were carried out up to 323 mL/min obtaining nanocapsules sizes down to 193 nm and pdi of 0,109. The flow rate working region is an order of magnitude higher than reported devices which bases its functioning in micromixers. Research results show the developed system potentiality in reducing the existing gap between laboratory and chemical process industry for microfluidic devices.*

Key-Words*: Nanocapsules; Microfluidics; LTCC; Nanotechnology; Chemical Processes Miniaturization*

Introduction: Since the late 70's several authors started to publish their articles in the nanoparticles field. Optical, chemical, magnetic, catalytic, mechanical and electrical nanoparticles properties are strongly dependant not just by its composition but also by their size and shape [1, 2].
In the pharmaceutical industry, pharmaceutical active compounds are encapsulated using nanocapsules. This practice offers several advantages, between them: the use in sustained release systems, improvement in bioavailability and biocompatibility, etc [4-8].
The emulsification - diffusion and solvent extraction/evaporation is a chemical process used for the pharmaceutical active compound encapsulation. This process makes possible the particles generation with sizes ranging from micrometer to nanometer [5, 8, 9] .
A traditional emulsification process requires high energy investment, as shear forces, for disperse phase drop size reduction and to obtain a higher system stability [10]. Conventional reactors and mixing mechanism suffers from non-homogeneous bulk shear forces distribution, leading to a poor control over the generated particles sizes and a broad size distribution. Thus, it is important to develop alternatives processes or technologies that makes possible nanocapsules generation with size control and narrow size distribution.
Microfluidic devices are built using different substrates, one of them is the Low Temperature Co-fired Ceramic (LTCC). LTCC displays interesting advantages such as the possibility to microfabricate 3D geometries, is chemically inert to most solvents, it has a low contact angle, presents low thermal coefficient of expansion, can hold out high operational temperatures, can withstand high internal pressures, allowing implementation of several chemical process with applications in stringent environments [11].
This work focuses on a construction of a continuous regime microfluidic system for pharmaceutical active compounds nanocapsules generation. This is done by miniaturizing the emulsification - diffusion and solvent extraction/evaporation chemical process through a microfluidic approach using the LTCC technology. The proposed system looks toward the generated nanocapsules size setting by controlling process variables like total flow rate through devices. It also aims the possibility to use the developed system for an industrial production line of pharmaceutical active compound nanocapsules. For this, the system should be able to work at flow rates higher than the reported in the literature.

Experimental: The unit operations chemical process involved in a conventional emulsification - diffusion and solvent extraction/evaporation process were analyzed. As a result, it was concluded that steps referred to the nanoemulsion and diffusion could be done by means of microfluidic techniques maintaining the same operation principles. The developed system layout is presented on Fig.1.



Aqueous Phase:
- Tween®20 - 1% (w/v)
- Ethyl Acetate - 2,5% (w/v)
- Deionized water - 390 ml

Organic Phase:
- Polycaprolactone PCL 10kDa - 2% (w/v)
- Pluronic®F127 NF - 2% (w/v)
- Miglyol®810 oil - 5% (w/v)
- Hydrocortisone Acetate - 0,5% (w/v)
- Ethyl Acetate - 100 ml

Dilution Material:
- Tween®20 - 1% (w/v)
- Deionized water - 500 ml

Fig 1. Continuous regime microfluidic system layout for nanocapsules generation.

The base microfluidic geometry selected for the emulsion and diffusion steps was the 3D Serpentine, Fig. 1. This geometry makes possible to obtain a similar action to the mechanical agitation. This action propitiates emulsion generation inside the microchannels when used immiscible fluids at the entrance.
The system has three main modules. The first one generates the emulsion. Two reservoirs for aqueous and organic phases, two HPLC pumps, two flow rate sensors, a pressure sensor and the 3D Serpentine micromixer for emulsion generation integrates this module. The second module uses another microfluidic geometry based in 3D Serpentine for solvent diffusion from inside the drop. This module also has a reservoir for deionized water and a pump. Finally, there is the solvent extraction module.
The Fig.1 also shows the materials used for samples preparation. The mixing of chemical components was carried out with magnetic stirring until phases become completely translucent. The preparation temperature for aqueous phase and dilution was 21 °C while for organic phase was 60 °C.
The selected technique for measuring nanocapsules sizes (Tp) and polydispersity index (pdi) was the Dynamic Light Scattering (DLS) using a Nano-ZS equipment.

Results and discussion: For system validation, the total flow rate in the emulsion generation module was swept from 10 mL/min to 65 mL/min with 5 ml/min steps while the flow rate ratio between aqueous and organic phases was four. By its time, the flow rate ratio between dilution flow rate and emulsion flow rate was kept constant at 3,5. Experiments without solvent extraction were conducted.
The Fig. 2 shows the nanocapsules sizes and pdi variation as the total system flow rate varies from 90 ml/min to 293 ml/min. It can be seen that a system were nanocapsules sizes are controlled by mean of total flow rate was obtained, which is extremely interesting for process like this. The nanocapsules have a tendency to reduce their sizes as total flow rate through devices increase. Nanocapsules with sizes varying from 790,5 nm ± 36,9 nm to 209,7 nm ± 3,5 nm with pdi between 0,313 ± 0,016 and 0,09 ± 0,017 were obtained.
A flow rate increase in a 3D Serpentine micromixer leads to chaotic advection intensification. As a result, there is an increase in the flow streamlines TORÇÃO E ENTRECRUZAMIENTO leading to an increase in the shearing forces. As consequence, there is a tendency to reduce the generated drops sizes in the emulsification stage. The same applies to the dilution stage, where the chaotic advection intensification lead to a better mixing between the emulsion and the dilution liquid, propitiating a higher solvent diffusion rate from inside the emulsion drops to the surrounding liquid.
Tests were done with the system working at total flow rate as high as 323 ml/min and nanocapsules with size of 193 nm and pdi of 0,109 were obtained. This value is an order of magnitude higher than the previously reported in the scientific literature. Higher flow rate were not tested, but the system is not limited to the previously mentioned value. Reducing nanocapsules size as total system flow rate increases is an advantage because turns possible to achieve a higher production rate. This is very interesting when an industrial production scale is desired. To increase even more the production rate, each module could have more than one microfluidic device working in a batch configuration (scale out). The fact to have a microfluidic system working at flow rates in the order of 320 ml/min facilitates the production scale out.

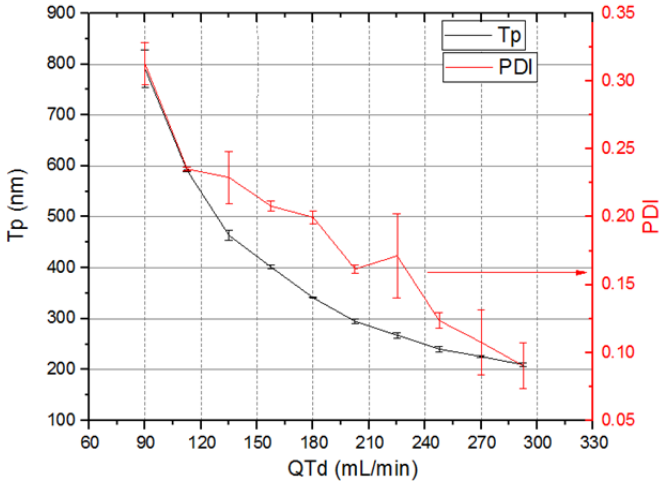


Fig 2. Nanocapsules sizes and pdi versus total flow rate through devices.

Conclusion: This work presents a continuous regime microfluidic system for pharmaceutical active compounds nanocapsules generation. Three modules integrate the system, the emulsification module, the dilution module and the solvent extraction module. The emulsification and dilution devices, manufactured with LTCC technology, make use of microfluidic topologies based on 3D Serpentine micromixers.

The developed system generates nanocapsules with mean sizes between 790,5 nm ± 4,7% and 209,7 nm ± 1,7%, with pdi between 0,313 ± 5,1% and 0,09 ± 18,9%, when a total system flow rate varies between 90 ml/min and 293 ml/min. The system can work at flow rates higher than 293 ml/min, tests with 323 ml/min obtained nanocapsules with sizes of 193 nm and pdi of 0,109.

Results show that this microfluidic system has the capability to adjust the nanocapsule size through controlling the total system flow rate.

A system like this, with the potentiality to generate nanocapsules with narrow size distribution and control over size, operating in continuous regime at flow rate in the order of 320 ml/min or higher, has not been previously reported by scientific literature which is an original contribution of this work.

This work also shows a way to reduce the existing gap between the microfluidic devices developed for a lab proof of concept and a possible use of such devices in an industrial production.

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