Micronization of Lipid Particles with Different Methods

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ABSTRACT

Among the several methods widely described in literature for particle micronization, this study focuses in the application of five different techniques to produce microparticles of Precirol® ATO 5, a polyglycolized glyceride (GRAS - Generally Recognized As Safe), used as a drug carrier in delivery systems.
The methods investigated include solvent evaporation, spraying techniques (spray-drying and spray-chilling), and processes with supercritical fluids (SAS- supercritical anti-solvent and PGSS® – particles from gas saturated solutions).
Different experimental conditions were tested, for the several methods, in order to optimize the process variables for this system. The particles obtained were characterized by scanning electron microscopy (SEM) and by laser diffraction spectrometry.

INTRODUCTION

Microparticles have been extremely important in the development of various clinically useful drug delivery systems [1]. The most common technique to reduce the size of particles is mechanical comminution, which includes operations like crushing, grinding and milling. These do not represent the ideal micronization process since both the properties of the compound and the surface properties of the particles might be altered in an uncontrolled manner [2]. Among the several methods widely described in literature, this study focuses in the application of different techniques that can overcome the previously mentioned problems, to produce microparticles of Precirol® ATO 5, a GRAS polyglycolized glyceride, used as a drug carrier in delivery systems [3,4]. The precipitation methods studied include conventional solvent evaporation, spraying techniques (spray-drying and spray-chilling), and processes with supercritical fluids (SAS- supercritical anti-solvent and PGSS – particles from gas saturated solutions).
For the conventional solvent evaporation method, an organic solution of the compound is prepared and further emulsified in an aqueous continuous phase to form discrete droplets. The microspheres are formed upon evaporation of the organic phase at the water/air interface, after diffusion into the aqueous phase. As the evaporation occurs, the microspheres harden and become free flowing after appropriate filtration and drying [5].
The spraying techniques have been extensively used in food and pharmaceutical industries, and are widely described in literature [6-8]. Spray drying involves the evaporation of a solvent from an atomized liquid feed, by mixing it with a drying medium, e.g. hot air. Spray chilling on its turn, uses the same principle of atomization, but this time the air stream is a cooling medium, since the feed is a molten matrix and the atomized droplets of this feed will solidify into fine particles that can thus be separated. These spraying techniques are based in fact in the same principles as the supercritical atomization methods, supercritical anti-solvent (SAS) and particles form gas saturated solutions (PGSS). Therefore SAS can be considered a spray drying process at high pressure, in the same way as PGSS can be considered as a spray chilling high pressure method. The SAS process uses the anti-solvent effect of supercritical CO_2 to precipitate the substrate(s) initially dissolved in an organic solvent. The principle of the process is to decrease the solvent power of the liquid by addition of an anti-solvent in which the solute is

insoluble. The PGSS process uses the effect of the supercritical fluid as a solute. It consists in solubilizing the supercritical fluid in melted or liquid-suspended substance(s) leading to a so called gas saturated solution/suspension that is further expanded through a nozzle with formation of solid particles or droplets [9].
In this work, the possibility of producing small lipid particles of Precirol was explored through these five precipitation processes. Different experimental conditions were tested in order to optimize the process variables for this system.

MATERIALS AND METHODS

Materials

Precirol® ATO 5, was kindly provided by Gattefossé. According to the supplier it is mainly composed of palmitic and stearic acids (C_{16} and C_{18}). Dichloromethane, DCM, CAS [78-09-2], 99.9% purity was purchased from Vaz Pereira, Portugal. Carbon dioxide (99.5%, industrial grade) was obtained from Air Liquide (France). Polyvinyl alcohol (PVA, molec. wt. 30000-70000) was purchased from Sigma Aldrich. All products were used with no further purification.

Solvent evaporation

Microspheres were prepared using different amounts of Precirol dissolved in 3 mL of DCM (concentrations between 25 and 100 mg/mL). This organic solution was then emulsified in an aqueous solution of PVA (0,25 to 1% concentration and with volumes form 100 to 800 mL)) under constant stirring with a two-baffled rotator for one hour or using an ultraturrax during 5 min (speed between 1200 to 20500 rpm). In the last case the resulting O/W emulsion was stirred with a magnetic stirrer for 1 hour at maximum speed. The hardened microparticles were then washed four times with distilled water, collected by centrifugation at 12000 rpm for 20 minutes and lyophilized.

Supercritical anti-solvent (SAS)

The experiments were carried out in a SAS apparatus that has been described elsewhere [10]. Briefly, the apparatus (Figure 1c) works in a continuous co-current mode and it consists of a precipitator in which the anti-solvent and the liquid solution are separately fed to the top of the chamber and are continuously discharged from the bottom. The precipitator is a cylindrical vessel with an inner volume of 500 cm^3. The liquid DCM solution (concentrations between 25-100mg/mL) is delivered into the chamber through a stainless steel 300 μm nozzle. The carbon dioxide is heated, up to the temperature of the experiment, before entering the precipitator. A filter of sintered steel with 0.1 μm porosity is placed at the bottom of the vessel to collect the particles produced. The solvents are separated and recovered from a second vessel. Experiments were performed in a pressure range between 8-10 MPa, temperatures of 308-318K and liquid flows between 0,5 - 1 mL/min.

Particles form gas-saturated solutions (PGSS)

This technique known as PGSS - Particles from Gas Saturated Solutions, was first described by Weidner *et al* [9]. In the apparatus used (Figure 1d), described elsewhere [11] the carbon dioxide is fed to a high pressure stirred reactor containing the product, and the operating conditions, in order to have the product melted under carbon dioxide atmosphere, were adjusted according to previous fundamental studies [12]. After a certain stirring equilibration time, the mixture is then depressurized through a valve and a nozzle. The solid particles are finally collected and separated from carbon dioxide in a vessel of about 10L volume. The parameters varied were pressure (from 11 to 19MPa), nozzle size (from 300 to 600 μm) and temperature (between 324 and 333 K).

Spray-drying

These experiments were performed using a Büchi Mini Spray Dryer B-290 (Flawil, Switzertland) schematically represented in Figure 1a. This system operates according to a co-current air and product stream, through a two-fluid nozzle with a 0.7 mm diameter inner tip and

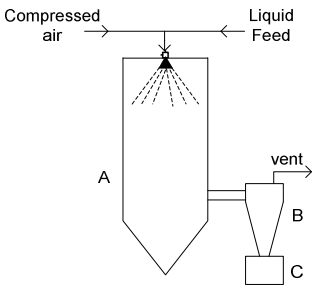
a 1.4mm diameter cap-orifice. Compressed air is used to disperse the liquid body into fine droplets which are subsequently dried in the cylinder. Liquid feed volumetric flow rate (2,5 mL/min), and drying air volumetric flow rate (40 m^3/h) were kept constant. Temperature was varied between 338 and 348 K and the concentration of Precirol in the DCM solution varied between 20 – 100 mg/mL.

Spray-chilling

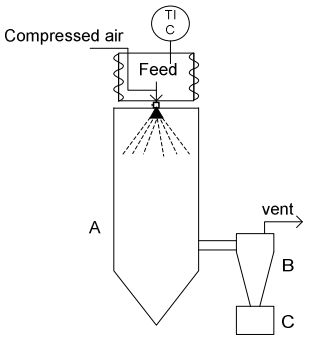
For this method the same apparatus was used, only with an adequate accessory, to allow the melting of the substance (Figure 1b). The temperature in this chamber was set at 160ºC, with silicone oil, to ensure the compound was liquefied. This liquid was then atomized to the spraying cylinder through the same two fluid nozzle, with an atomizing air volumetric flow rate of 355 dm^3/h and a drying air volumetric flow rate of 40m^3/h. Experimental limitations made possible a single test where a Precirol feed was melted at 433 K.

Particle characterization

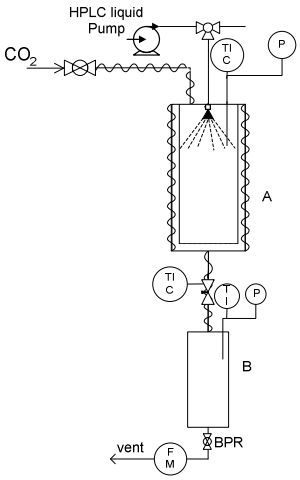
The morphology of the particles was analysed and imaged by scanning electron microscopy (SEM) after being fixed by mutual conductive adhesive tape on aluminium stubs and covered with gold palladium using a sputter coating. The particle size and size distribution of the prepared microparticles were measured by Laser diffraction spectrometry (Coulter LS 130, Coulter Electronics). The dried powder samples were suspended in deionised water with a surfactant solution and sonicated for 1 minute with an ultra-sound probe (500 W, before measurement.



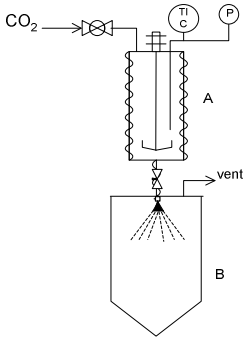
a) Spray-drying : A – drying chamber, B – separative cyclone, C – particle collector



b) Spray-chilling : A – drying chamber, B – separative cyclone, C – particle collector



c) SAS**:** A – precipitation vessel, B – separation vessel, P-pressure indicator, TIC – temperature control, FM – flowmeter, BPR – back pressure regulator



d) PGSS: Apparatus for PGSS experiments. A – high-pressure stirred vessel, B – collector, P – pressure indicator, TIC – temperature control

Figure 1: Schematic diagrams of the apparatus used in the different precipitation methods

RESULTS

A free flowing powder of Precirol was successfully obtained in all experiments performed. Representative scanning electron micrographs of the external surfaces of Precirol particles are shown in Figure 2.

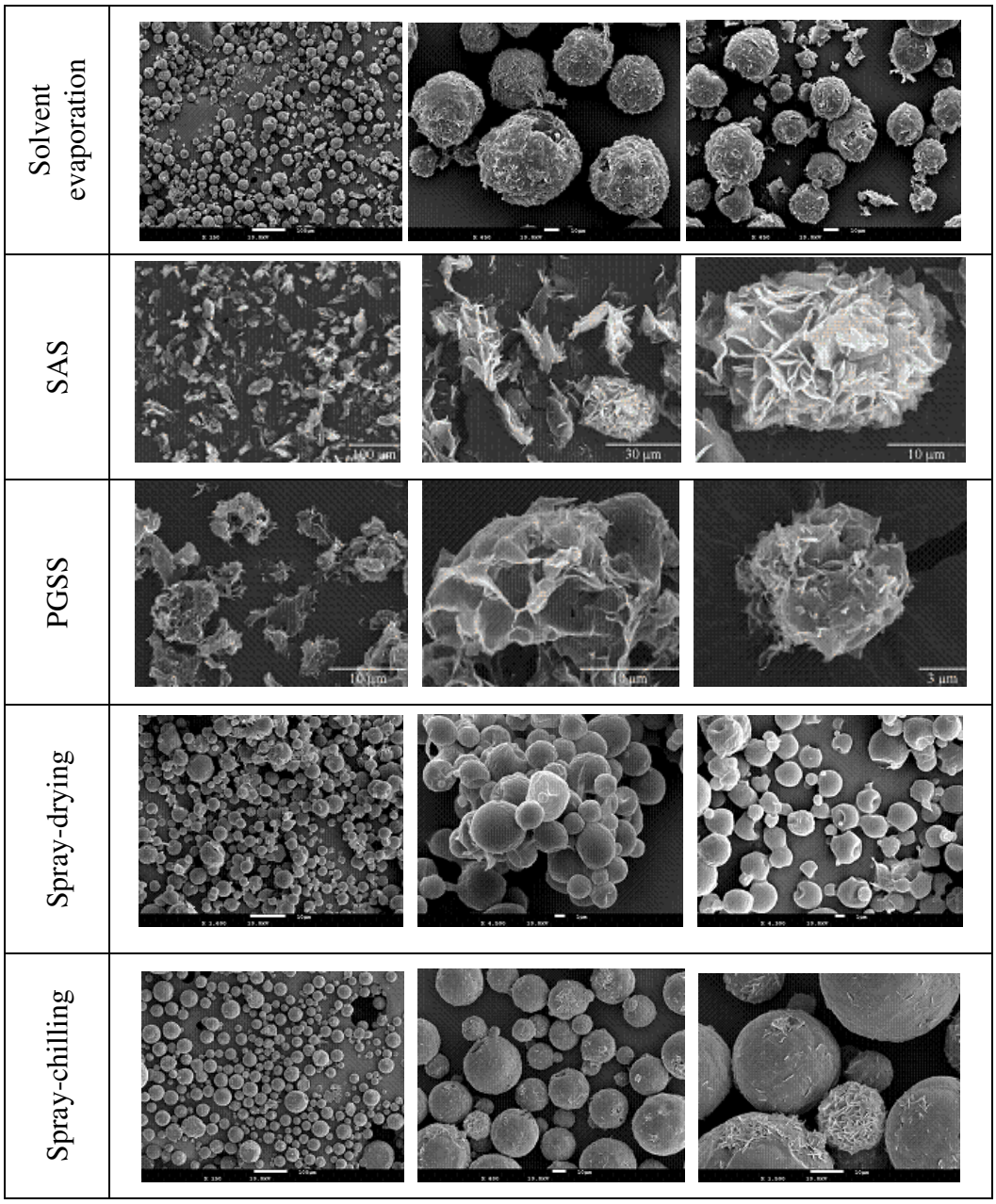


Figure 2: Scanning electron micrographs of the particles obtained in the different methods

Particles prepared from solvent-evaporation and drying techniques were spherical and presented very smooth surfaces while particles prepared using supercritical fluid technology had a laminar surface. Furthermore, when PGSS was used the particles obtained appear to be more porous than the ones prepared by SAS.

If the particle size is taken into consideration (Figure 3), it is possible to conclude that solvent-evaporation and spray chilling produced particles with bigger sizes, while for the other techniques the particle size is smaller and in the same range of magnitude. The different

experiments using spray-drying gave particles with average particle size between 5,7-15,2 μm, and the experiments using supercritical fluids produced particles between 3,9-11,1 μm and 3,8 – 9,2 μm, for SAS and PGSS respectively.

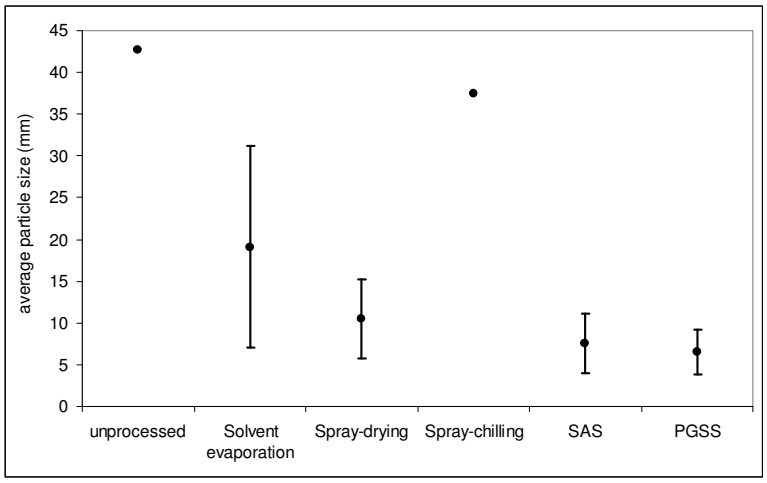


Figure 3: Average particle size range (d50, volume%) of the particles obtained in the different experiments of methods studied

In terms of operating conditions, spray–chilling, even without the use of pressure or organic solvents, requires extremely high temperatures. Solvent evaporation, on its turn, is performed at room temperature but involves the use of large amounts of solvents which, contrary to spray-drying and SAS (both one step processes), requires further drying steps. In both supercritical based processes (SAS and PGSS), the disadvantage of pressure requirement is overcome by the moderate temperatures needed.

CONCLUSION

In this study the feasibility of several techniques to process solid lipid particles, namely of Precirol has been investigated.It is possible to conclude that interesting morphologies and shapes can be obtained with different methods. The choice of the processing technique can therefore be made as a function of the final application of the product, and its desired morphology, or depending on the operating conditions needed.

ACKNOWLEDGMENTS

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