DESIGN AND ASSEMBLY OF A LABORATORY-SCALE SUPERCRITICAL ANTI-SOLVENT SYSTEM (SAS AND SFEE)

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ABSTRACT

In the last years, many precipitation processes involving supercritical CO_2 were developed. CO_2 can be used in supercritical state as solvent, solute or antisolvent. Examples of techniques that use CO_2 as antisolvent are SAS (Supercritical Antisolvent) and SFEE (Supercritical Extraction of Emulsions). Although there are many articles available in the literature about the influence of operational parameters in supercritical precipitation processes, data related to the design and assembly of laboratory-scale systems are scarce. The objective of this work is to present the design and assembly of a laboratory-scale precipitation unit, able to operate the SAS and SFEE techniques, and also present details of the assembly and results for the encapsulation of beta carotene by SAS (using Poly(L-lactide) (PLLA) as coating polymer). The CO_2 compression system consists of a cooling bath (-10 ˚C), a pneumatic pump and heating bath for CO_2 to reach the operating temperature. The injection system of the organic solution consists of a HPLC pump. The organic solution and supercritical CO_2 are fed into a high pressure jacket column (internal volume of 712 mL) through a coaxial nozzle with an internal diameter equal to 127 µm. The developed system successfully produced micro particles of beta carotene with PLLA as coating agent through SAS.

INTRODUCTION

Processes involving supercritical fluids (SCF) have been used in laboratory and industrial scale and the study of these processes are widely encouraged since they can be considered clean technologies, especially when using carbon dioxide (CO_2) and water as fluids.

SCFs find application in extraction processes, particle formation, sterilization, as reaction medium, among others [1]. In recent years, numerous micronization and particle formation process involving SCFs have been developed. These processes can be classified according to the role of the SCF in the process: solvent, antisolvent, co-solvent, and propellant gas [2]. The SAS (Supercritical Anti Solvent) and SFEE (Extraction by Supercritical Fluid Emulsions) techniques belong to the category that use CO_2 as antisolvent, and have as a common feature the very low miscibility or immiscibility of the solute in the antisolvent and the complete miscibility of the solvent in the antisolvent.

Despite the abundance of articles available in the literature regarding the influence of operating parameters, polymer and solvents in different precipitation processes involving SCFs, data about the design of precipitation systems, especially with regard to the configuration of the unit and vessel, proper choice of components and difficulties that may be encountered during the process of design are scarce. Vemavarapu et al. [3] reviewed aspects of design and particle formation with SCF systems, and information from the precipitation vessel design were presented with a greater degree of detail by Chong et al. [4]. Such contributions are essential for new researchers with interest in developing its own system of particle formation. Thus, the aim of this study is to present the design and assembly of a versatile precipitation unit, capable of operate the SAS and SFEE processes, as well as presenting results from precipitation of beta carotene by SAS.

MATERIAL AND METHODS

Material

The materials and equipments used for assembly of the system are shown in Table 1. The antisolvent used in SAS processes was CO_2 (White Martins, Campinas, SP, Brazil) with 99.0 % purity. Beta carotene (Type I, synthetic, Purity ≥ 93%), used as core material, was purchased from Sigma Aldrich (Saint Louis, USA). The Poly(L-lactide) (PLLA), used as precipitation polymer by the SAS technique, was purchased from Sigma Aldrich (Saint Louis, USA). All the other solvents and chemicals were of analytical grade.

Table 1 Materials and equipment of the precipitation unit: specification, brands and maximum operating pressures.

Article	Especification	Brand	Max pressure (MPa)
CO_2 cylinder	CO_2 industrial 23 kg	White Martins	10
Stainless steel 316 tube 1/4", 1/8" e 1/16"	-	FAE S.A. /Autic Automação	55
Washers and screws for tubes 1/4", 1/8" e 1/16"	-	Detroit / Parker Corp.	55
Tees and connections 1/4", 1/8" e 1/16"	-	Detroit / Parker Corp.	55
Pneumatic pump	M111	Maximator	130
HPLC pump	Model PU 2087	Jasco	25
Odontological compressor	Model MSV6/30	Schulz S/A	0.83
Manometers	Model Z.10.B+-	Zürich LTDA	160
Thermostatic bath with cooling and circulation	Model MA-184/E	Marconi	-
Mini water bath with circulation	Model MA 126/BD	Marconi	-
CO_2 filter (0.5 μm)	SS-2F-05	Swagelok	60
Compressed air filter	B07	Norgren Express	0.7
Safety valve with pressure relief	SS-4R3A	Swagelok	43
Precipitation vessel	35 Mpa	Autic Automação	45
Needle valve coarse adjustment	Part. number 10V2071	Autoclave Engineers	130
Needle valve for fine adjustment	Part. number 8481.40.00	Autoclave Engineers	69
CO_2 flowmeter	Model PMR1 010423	Cole Parmer	-
Aluminum profiles	45 x 45mm - DS680	Jartec Automação	-
Stainless steel plate	30 cm x 71 cm	Autic Automação	-

SAS and SFEE systems

The schematic diagram of the SAS/SFEE developed equipment is shown in Figure 1. The systems that comprise the equipment are detailed as follow:

CO_2 compression system

The CO_2 compression system is composed of a cooling bath (B1) filled with ethylene glycol and an air pump (CO_2 pump) that pressurizes liquid CO_2 to the desired pressure with the aid of compressed air. Pressure regulation is done by the pressure of the compressed air supply to the pump, controlled by the air filter (F). The pump head is cooled with ethylene glycol (-10 °C) to prevent pump cavitation. A coil submerged in a heating bath (B2) is responsible to take CO_2 to the operation temperature.

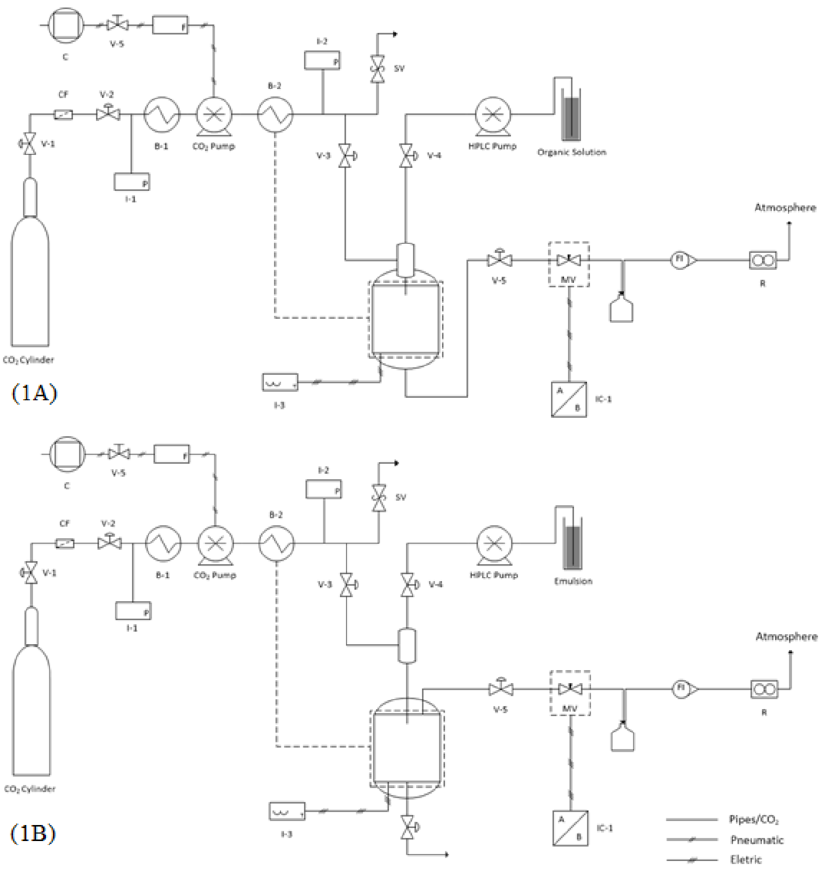

Figure 1 Experimental apparatus: (1A) SAS configuration and (1B) SFEE configuration V-1, V-2, V-3, V-4 , V-5 and V-6 – Control valves; MV – Micrometer valve; SV – Safety valve; C- Compressor; F- Compressed air filter; CF – CO_2 Filter; B1 –Cooling bath; B2 – Heating bath; I-1 e I-2 – Pressure indicators; I-3 – Temperature indicator; IC-1 – Indicators and controllers of temperature of micrometer valve, R – Rotameter; FL – Flow meter

Design of heat exchangers

Heat exchangers must be used to make CO_2 reach the desired temperature of operation. The heat exchanger consists of a coil of stainless steel submerged in a bath with temperature control. The critical parameter that must be determined is the surface area of the heat exchanger required for the operation of SAS and SFEE [4]. The calculations for determining the area of heat exchange for cooling (a) and heating (b) bathes are the following:

(a) Cooling bath

For the calculations of the coil length, the following data were considered:

Heating temperature = (T_∞) = 263.15 K;

Inlet temperature of the CO_2 = ($T_{m,in}$) = 293.15 K;
Outlet temperature of the CO_2 = ($T_{m,out}$) = 263.65 K;
Feed flow rate (maximum flow operation) $= \dot{m} = 4.68 \times 10^{-4}$ kg/s;

First, it was determined the total heat (q_{conv} = 34.53 W; 124324 J/h) required by Equation 1 and using the data presented in Table 2.

$$q_{conv} = \dot{m} c_p \left(T_{m,in} - T_{m,out} \right) \tag{1}$$

It was established the mean log of temperature differences (ΔT_{ml} =-7.21 K), using Equation 2.

$$\Delta T_{ml} = \frac{\left(\Delta T_{out} - \Delta T_{in} \right)}{\ln \left(\frac{\Delta T_{out}}{\Delta T_{in}} \right)} \tag{2}$$

Table 2 Physical chemical properties of CO_2 at different conditions.

Property	P = 6 MPa, T = 278.40 K	P = 30 MPa, T = 298.15 K
Specific heat (c_p)	2501 (J/kgK)	2868 (J/kgK)
Viscosity (μ)	0.3451 (kg/hm)	0.3961 (kg/hm)
Thermal conductivity (k)	389.5 (J/hmK)	464.4 (J/hmK)
Density (ρ)	915.5 (kg/m^3)	966.52 (kg/m^3)

The pipe used for coil has a nominal diameter of 1/8 pol. According to Kern [5], the cross-sectional area of flow within the pipe (a_t) is 3.7419 10^{-5} m^2, its outside diameter (d_e) is 1.029 10^{-2} m and internal diameter (d_i) is 6.83 10^{-3} m. To determine the overall heat transfer coefficient (U), it is necessary to calculate the mass flow (45025.25 kg/m^{2}h), by Equation 3 and from it the Reynolds number (Re) is calculated by Equation 4.

$$G = \frac{\dot{m}}{a_t} \tag{3}$$

$$\mathrm{Re} = \frac{d_i G}{\mu} \tag{4}$$

Reynolds number is 891.1, and since it is lower than 2100, one can determine the value of h_i / ϕ according to Equation 5 for laminar flow:

$$\frac{h_i}{\phi} = 1{,}86 \frac{k}{d_i} \left(\frac{d_i G c_p \mu d_i}{\mu k L} \right)^{1/3} \tag{5}$$

Where: h_i = heat transfer coefficient (W/m^{2}K); L = coil length (m); μ_p = viscosity of CO_2 at the temperature of the tube wall (Pas); φ = viscosity ratio $\left(\frac{\mu}{\mu_p} \right)^{0,14}$.

As the coil length was unknown, an initial value of 5 m was considered. The h_i / ϕ value found was 1.32 10^5 J/hm^{2}K. The next step was to determine the temperature in the pipe wall (T_p = 263.78 K), according to Equation 6, for when the hot fluid is passing inside the tube.

$$T_p = T_\infty + \frac{\frac{h_i}{\phi}}{\frac{h_i}{\phi} + h_\infty} \left(T_{m,CO_2} - T_\infty \right) \tag{6}$$

Where h_∞ = 3.0663 10^6 J/hm^{2}K (heat transfer coefficient of water) [5]. Using the values of T_p and the pressure of 6 MPa, the viscosity of CO_2 in the wall temperature (μ_p) is 0.4461 kg/mh [6]. The calculated φ was 0.9647 and h_i was 1.27 10^5 J/hm^{2}K. With *hi*, the heat transfer coefficient of external diameter of the tube (h_{io} = 84517.85 J/hm^{2}K) can be determined from Equation 7:

$$h_{io} = h_i \frac{d_i}{d_e} \tag{7}$$

Considering that there is no incrustation on the pipe, *U* was determined by Equation 8:

$$U = \frac{h_{io} h_\infty}{h_{io} + h_\infty} \tag{8}$$

Thus, the overall heat transfer coefficient was 82250.73 J/hmK. The heat exchange area and tube length were determined by Equations 9 and 10:

$$A = \frac{Q}{U \Delta T_{ml}} \tag{9}$$

$$L = \frac{A}{S_{ext}} \tag{10}$$

Where: S_{ext} = area per linear meter of pipe = 0.032309 m^2/m [5].

The values obtained for the heat transfer area and length of the coil were respectively 0.21 m^2 and 6.5 m. The value obtained for the tube length was lower than the initially set. The previous stipulated value of coil length was replaced for the calculation until the stipulated and obtained values coincide. The final value for the coil length was 6.3 m. To ensure that the CO_2 is cooled satisfactorily a length of 7.0 m was used.

(b) Heating bath

The length of the coil for the heating bath was determined in the same manner as for the cooling bath (data available in Table 2).

For the calculations, the following temperatures in the process were considered:

Heating temperature = (T_∞) = 334.15 K;

Inlet temperature of the CO_2 = ($T_{m,in}$) = 263.15 K;

Outlet temperature of the CO_2 = ($T_{m,out}$) = 333.15 K.

The calculated final length of the coil was 5.7 m. To ensure that the CO_2 is satisfactorily heated a length of 6.0 m was used.

Supercritical CO_2 flow rate measurement system

The measurement of supercritical CO_2 flow rate is a relatively complex issue due to the high operating pressure of the equipment [4]. An alternative is to measure the CO_2 flow rate after depressurization. This was achieved after decompression in micrometric valve by means of a flow meter.

Configuration of the injection system

Several nozzle configurations can be used in processes where CO_2 acts as antisolvent. According to Vemavarapu et al. [3], the nozzle configuration has direct influence on the particle morphology and the rate of solvent extraction by the SCF. Among distinct configurations available, we opted for a coaxial injection system typical of SAS system, composed of a stainless steel tee, which is connected to antisolvent and solution injection lines. Until this point, the antisolvent (CO_2) and the solution drain by separate lines, and from there to the precipitation vessel, belong to the same line, but the solution flows inside a capillary tube of 1/16¨ outside diameter and 0.02” inner diameter (0.58

mm) and the antisolvent flows out of the capillary tube and inside a tube with an outer diameter of 1/8” and inner diameter of 0.105” (2.67 mm). The assembled system has great versatility, allowing easy replacement of pipe injection of the organic solution, which enables the use of 1/16” pipes with different inner diameters.

Precipitation vessel

According to Figure 1, the difference between operating the equipment in SAS and SFEE mode is due to the output location of the antisolvent in the precipitation vessel: while in the SAS system the anti-solvent leaves the bottom of the vessel (in which a filter is coupled to retain the particles), in the configuration of SFEE, the antisolvent exits the pressure vessel from the top, which allows the suspension to remain at the bottom of the vessel during the process.

A cylindrical jacketed stainless steel vessel (volume of 712 mL: inner diameter of 7 cm and height of 18.5 cm) was used. The maximum operating pressure is 35 MPa. The geometry of the precipitation vessel was determined based on the profile of acetone droplets simulated in supercritical CO_2 (in a capillary of 100 µm inner diameter) at 318 K and 10 MPa, illustrated in Figure 2, as described by Chong et al. [4]. According to the author, the lifetime of the droplets in acetone was used as the retention time required to determine the height of the precipitation vessel.

The geometry of the vessel was determined for the following operating conditions:
Maximum feed rate of the polymer solution (*Q*) = 2.00 mL/min;
Inner diameter of the capillary (*di*) = 100 mm;
Superficial velocity (*vf*) = feed rate of the solvent (*Q*)/cross-sectional area of the capillary (*At*), according to the Equation 11.

$$vf = \frac{Q}{At} = \frac{2.00\times10^{-6}}{(\pi/4)(100\times10^{-6})^2} \approx 4.5\,\text{m/s} \qquad (11)$$

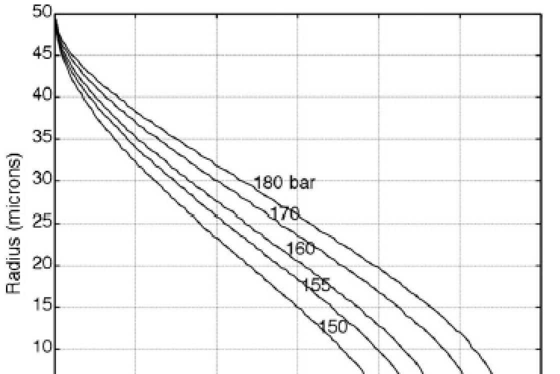

Figure 2 Evolution of the acetone droplet radius at 318 K under various pressures [4].

The length of the pressure vessel (*Lv*) required for complete removal of acetone droplet is can be calculated from the Equation 12.

$$Lv = vf \times 0.02 = 4.5 \text{ x } 0.02 = 0.09 \text{ m} = 9 \text{ mm} \qquad (12)$$

An extra length of approximately 50% was added to the precipitation vessel to ensure complete removal of solvent. Thus, the height of the precipitation vessel was set at 185 mm. The internal diameter of the precipitation chamber is 70 mm, with a ratio L_v/Di of

2.7. This ratio allows full dispersion of the particles without hitting the vessel wall to ensure precipitation and removal of the solvent.

The material used for the construction of the precipitation vessel was stainless steel 316, which is chemically inert, able to withstand high operating pressures and fully meets the requirements of the American Society of Mechanical Engineers (ASME). The construction of the pressure vessel is in accordance with the ASME Boiler and Pressure Vessel Code Section VIII, Division 1.

SAS experiments: encapsulation of betacarotene

The designed system was tested by performing the process of particle formation of beta carotene by SAS processes. The particles were obtained through the SAS process using solutions of PLLA with beta carotene in diclorometane (DCM). The feed solution was prepared by dissolving polymer and beta carotene in DCM at different concentrations, as shown in Table 3. Temperature (40 °C), PLLA solution concentration (0.5 %, w/v) and CO_2 flow rate (20.4 g/min) were kept constant in all experiments. Approximately 30 mL of feed solution was injected to the vessel. After the injection of the feed solution, the system was kept under the same conditions for 30 min, with continuous flow of CO_2. Afterwards, the vessel was depressurized and the particles were collected in petri plates, sealed and stored under refrigeration.

Table 3 Experimental conditions for SAS process

Exp	Pressure (MPa)	Flow rate of feed solution (mL/min)	Concentration of betacarotene (% m/v)
1	8	0.5	0.05
2	8	0.5	0.1
3	12	0.5	0.05
4	12	0.5	0.1
5	8	1.0	0.05
6	8	1.0	0.1
7	12	1.0	0.05
8	12	1.0	0.1
9	10	0.75	0.075
10	10	0.75	0.075
11	10	0.75	0.075

The morphology of the particles was analyzed using a scanning electron microscope equipped with a field emission gun (FESEM - FEI Quanta 650). Prior to analysis, the samples were coated with gold in a SCD 050 sputter coater (Oerlikon-Balzers, Balzers, Liechtenstein). Both equipments were available at the National Laboratory of Nanotechnology (LNNano, Campinas-SP, Brazil). Analyses of the sample surfaces were performed under vacuum, using a 5 kV acceleration voltage and a large number of images were obtained on different areas of the samples to assure the reproducibility of the results.

RESULTS AND DISCUSSION

The equipment is easy handling and good performance for particle formation by SAS was observed. In the SEM images (Figure 3) one can note particles of regular shape and homogeneous sizes, when the experiments were carried out at 10 MPa (Fig. 3.2) and 12 MPa (Fig. 3.3). An amplification on the particles surface is shown on Figure 3 (at 20000x magnitude, indicated by "c" letter), revealing their spherical geometry and porous surface.

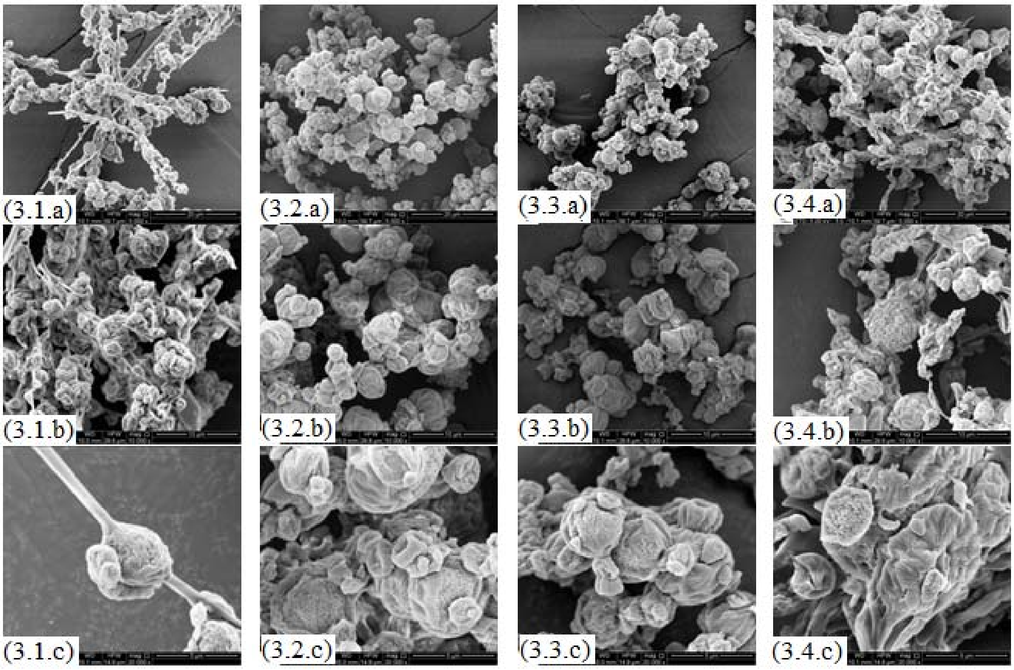

Figure 3 FESEM micrographs of the bet acarotene loaded micro particles - SAS conditions: (3.1) P = 8 MPa, Q_{sol} = 0.5 mL/min, C_{beta} = 0.05 g/100 mL; (3.2) P = 12 MPa, Q_{sol} = 0.5 mL/min, C_{beta} = 0.05 g/100 mL; (3.3) P = 10 MPa Q_{sol} = 0.75 mL/min C_{beta} = 0.075 g/100 mL and (3.4) 8 MPa, Q_{sol} = 1.0 mL/min C_{beta} = 0.01 g/100 mL and their respective amplification: (a) 5000x magnitude, (b) 10000x magnitude and (c) 20000x magnitude.

CONCLUSIONS

A SAS and SFEE coupled system was designed developed and tested using beta carotene as the model core material. The system successfully produced particles of beta carotene with PLLA as encapsulation polymer in the SAS process, with spherical geometry and porous surface.

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