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European Journal of Pharmaceutics and Biopharmaceutics 45 (1998) 67–74

European Journal of Pharmaceutics and Biopharmaceutics

Research article

Size controlled production of biodegradable microparticles with supercritical gases

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Received 28 May 1997; accepted 17 June 1997

Abstract

Polymer microparticles were produced by means of the aerosol solvent extraction system. A solution of 3% w/w poly(L-lactic acid) in dichloromethane was sprayed into supercritical or near critical carbon dioxide gas phase. The mean particle size by volume moderately depended on the nozzle diameter and the spraying pressure used. When the polymer solution was saturated with carbon dioxide up to 5.0 MPa, the particle size and morphology of the particles were unchanged and the product was less agglomerated. Microparticles with mean diameters from 6 to 50 μm were achieved by decreasing the carbon dioxide density from 690 to 250 kg/m^3. The surface structure of the particles sprayed in low density carbon dioxide showed cracks and holes. All other particles were non-porous with a smooth surface. © 1998 Elsevier Science B.V.

Keywords: Aerosol solvent extraction system; Supercritical carbon dioxide; Biodegradable polymers; Microparticles

1. Introduction

Various microencapsulation techniques exist to produce drug containing polymer particles. Conventional processes including solvent evaporation, spray drying or organic phase separation techniques have several disadvantages such as considerable amounts of residual organic solvent in the microparticles and low encapsulation efficiencies [1].

Gases at ambient conditions show low solubilities for non volatile substances. The solubility is increased rapidly near the critical pressure and temperature, as was reported at the end of the last century [2]. One of the most common gases used is carbon dioxide which is non toxic, non flammable, environmentally acceptable, and available in large quantities and in a pure state. The solvent power of fluid gases can be compared with liquids but the mass transfer properties are gas-like [3]. Due to the low critical temperature of 31°C, carbon dioxide is especially appropriate for the treatment of thermal-labile substances [4].

Following our publication of the aerosol solvent extraction system (ASES) process, several different techniques using supercritical carbon dioxide for microparticle production were launched [5]. Rapid expansion of supercritical solutions (RESS) was the first method investigated [6,7]. The polymer is placed into a high pressure vessel where it is dissolved in the supercritical carbon dioxide. In the second step, the pressure is reduced, which decreases the solubility of the polymer in carbon dioxide suddenly and the polymer is then precipitated [8,9]. This change causes a high super saturation of solute leading to a high nucleation rate resulting in small and uniform particles. In principle with RESS it is possible to produce microparticles with any type of polymer with sufficient solubility in the supercritical gas phase. However, in general, polymers

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PII S0939-6411(97)00124-0

have a very low solubility in supercritical carbon dioxide (< 1 wt.%) [10] except the low molecular weight fractions [11,12]. Furthermore, a polymer with high solubility in the supercritical gas will lead to sponge-like structured microparticles, due to rapid expansion during the sudden pressure reduction. Therefore a non uniform product will be obtained. To avoid the expansion of the carbon dioxide which causes the generated microparticles to be redissolved, the supercritical solution has to be heated before the expansion, to ambient conditions. In the literature [13,14] temperatures exceeding 100°C are mentioned, indicating that the RESS process is not suitable for use with thermal-labile substances. The expenditure on equipment is increased if drugs are to be incorporated into a polymeric matrix. The drug and the polymer must be dissolved in separate high pressure vessels at different temperatures, pressures and/or carbon dioxide flow rates to adjust the concentration of the drug in the polymer matrix.

An additional method, the gas antisolvent (GAS) crystallization was described by Gallagher et al. [15]. The authors dissolved the polymer in an organic solvent, which was miscible with the supercritical gas. The supercritical gas was then added to the organic solution. The liquid phase expanded and the solubility of solute decreased during this dilution procedure until the polymer precipitated to form particles. The particle size can be adjusted by pressure and temperature as well as by the rate of gas phase addition [15]. The precipitation time for particle formation was 6–8 orders of magnitude lower in the GAS crystallization than for the RESS process (10^{-5}–10^{-8} s) [16].

The precipitation with a compressed antisolvent (PCA) process uses an organic polymer solution which is sprayed directly into a high pressure carbon dioxide atmosphere [17–21]. The carbon dioxide acts as a non solvent for the polymer and as a solvent for the organic liquid. With the PCA apparatus only very small quantities of product may be produced. ASES is the scale up method of the PCA process to industrial production conditions. By depressurizing the supercritical gas phase, the organic liquid condenses and can be separated from the carbon dioxide and the purified gas can be pumped up to supercritical conditions once more [22–25].

In this versatile method for microparticle production, numerous parameters, influencing the particle formation, can be readily adjusted, e.g. carbon dioxide pressure and temperature, spraying conditions and polymer concentration. Furthermore, polymer solutions containing suspended drugs could be handled. Particle formation occurs directly in the supercritical gas phase, causing the precipitation to be sensitive to production parameters. Therefore the aim of this study was to examine the influence of the nozzle geometry and spraying conditions as well as the carbon dioxide extraction properties in the near critical range on the properties of the microparticles.

2. Materials and methods

2.1. Materials

A hydrophobic poly(L-lactic acid) with a mean molecular weight of 7000 Da was used (FhG-IAP, Teltow, Germany). The chain ends of the polymer were modified by esterification with oleyl alcohol. Some experiments were carried out with a commercially available poly(L-lactic acid) with a mean molecular weight of 102 000 Da (Resomer® L206, Boehringer Ingelheim, Germany). Dichloromethane in analytical grade (Merck, Darmstadt, Germany) was used to dissolve the polymers. The carbon dioxide used was of high purity grade (99.97%), supplied by Messer Grießheim (Frankfurt, Germany).

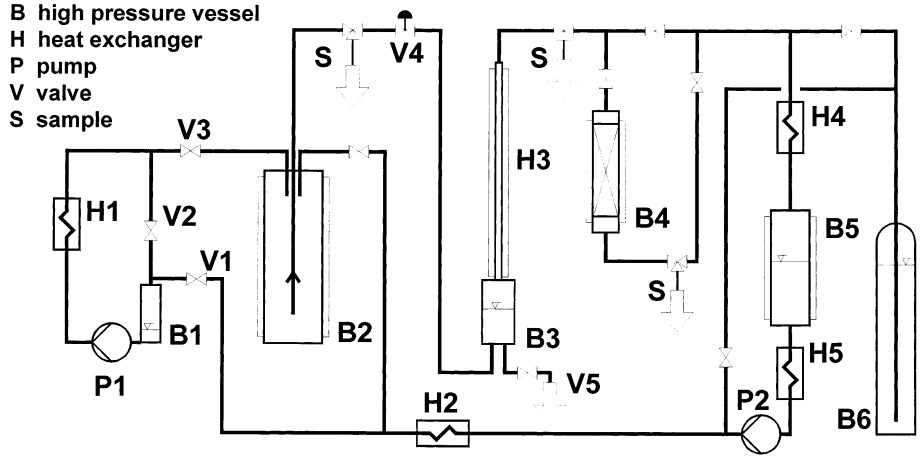

Fig. 1. Schematic of the aerosol solvent extraction system.

2.2. Microparticle production

The basis of the ASES process was described in detail earlier [23,24,26]. In this study, a newly developed apparatus was used which enabled microparticle production on a large scale (about 200 g per batch) and which was equipped with an improved system for pumping the polymer/drug solution into the high pressure column. After achieving steady state conditions in the equipment (Fig. 1), the solution containing 3% w/w polymer in 200 ml dichloromethane was sprayed through a nozzle into the high pressure vessel (B2) having a volume of 50 l. The pressure and temperature of the carbon dioxide were fixed at 8.5 MPa and 40°C, respectively, for most of the experiments. The carbon dioxide density was changed for one test series. During spraying and 30 min after this process, the high pressure vessel rested under static carbon dioxide conditions. No gas exchange occured to allow sedimentation of the formed microparticles. After this period, temperature controlled (heat exchanger H2) carbon dioxide was pumped by an adjustable diaphragm pump (P2) into the vessel at a flow rate of 40 kg/h. The dichloromethane contaminated carbon dioxide was expanded by valve V4 to subcritical conditions (4 MPa). The carbon dioxide/dichloromethane mixture was collected in the separation vessel (B3). The dichloromethane was enriched in the liquid phase of the separator due to thermodynamic equilibrium. The liquid was removed in intervals of 30 min (V5). The gas left the separation vessel through cooled vertical tubes, where residual dichloromethane condensed and rinsed back into the separator. The purified gas was directed through an adsorber unit to remove further traces of dichloromethane. The carbon dioxide was then liquefied in the condenser (H4) and stored in the liquid carbon dioxide storage vessel (B5) before it was pumped into the column again. The production was finished by backflushing the carbon dioxide into the storage vessel (B5) with a nearly constant flow rate of 15 kg/h.

The polymer solution could be delivered in different ways into the high pressure vessel. The standard method was the injection with an adjustable piston diaphragm pump (Fig. 2a) resulting in highly pulsatile solution flow. Most of the experiments were carried out using this application system. To deliver the organic solution without any pulsation, a piston unit was constructed (Fig. 2b) and placed into the solution storage vessel (B1). Carbon dioxide was used as the pressurizing fluid on the upstream side of the piston to inject the solution into the high pressure vessel. By pumping the organic solution in a closed loop (Fig. 2c) and dosing carbon dioxide directly to it, the solution was saturated with carbon dioxide gas until a constant pressure was reached. The solution storage vessel and the tubing were kept at a constant temperature of 40°C, due to the dependency of the amount of carbon dioxide diffusion into the liquid phase on the applied temperature [15,27].

2.3. Analytical methods

2.3.1. Particle size measurement

The particle size distribution by volume of the product was determined by laser diffraction (HELOS, Sympatec, Clausthal, Germany). The particles were suspended in an aqueous solution of Tween 80 (0.01% w/w). The particle size distribution was measured before and after 90 s of sonication. A previous series of tests have shown that the deagglomeration process was complete after 90 s [24]. The ratio of mean particle size ($\times$ 50) before and after sonication is called the index of agglomeration. The index of polydispersity, which is calculated from the quotient of the mean particle size by number and the mean particle size by volume, represents the width of the size distribution.

2.3.2. Particle morphology

To characterize the particle morphology, photographs were taken using a scanning electron microscope

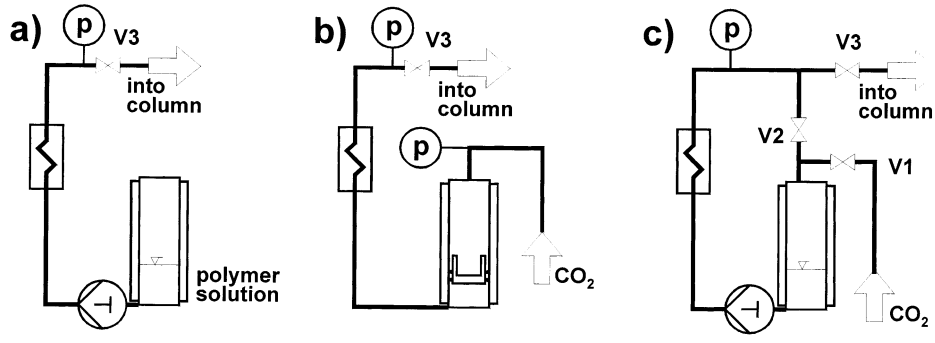

Fig. 2. Techniques for feeding the polymer solution into the high pressure vessel.

(Philips XL 20, Eindhoven, Netherlands). The particles were prepared on a conductive adhesive tape (Leittals, Plano, Marburg, Germany) and fixed on an aluminum sample tube. The samples were coated with gold for 3 min using a current of 50 mA under an Argon atmosphere at 5×10^{-2} Pa using a Sputter Coater SCD 005 (Balzas Union, Balzas, Liechtenstein).

2.3.3. Surface measurement

The total surface of the microparticles was determined by using a BET gas adsorption method (Gemini 2360, Micrometrics, Norcross) applying a five point measurement technique. The samples were prepared by rinsing nitrogen for 24 h through the powder at 40°C. The outer surface was calculated from the laser diffraction measurements. Assuming spherical microparticles, the specific outer surface by mass S_m could be calculated if the density of the particles r_P is given. By relating the surface of a spherical particle $S_P = \pi d^2$ to its mass $m_P = V_P \rho_P$ an equation for the specific surface of one particle $S_{m,P}$ follows:

$$S_{m,P} = \frac{6}{x \rho_P}$$

Under the assumption that all particles of a distribution have the same density, the outer surface of a particle collective could be calculated from particle size distribution function by volume $d\,Q_3$, which was obtained by the laser diffraction measurements (index i represents the classes of each particle size).

$$S_m = \frac{6}{\rho_P} \sum_{i=0}^{N} \frac{d\,Q_{3j}}{x_i}$$

The material density was obtained with a gas pycnometer (Accupyc 1330, Norcross, Georgia, USA).

3. Results and discussion

3.1. Spraying conditions

3.1.1. Spraying pressure

In previous experiments [23–26], the polymer solution was injected into the supercritical carbon dioxide with very little pressure difference. Presuming that the polymer solution atomizes into droplets at the outlet of the nozzle and furthermore that one microparticle originates from one droplet by 'evaporation' of the dichloromethane, the particle size could be influenced by the injection pressure. In a series of tests, the pressure difference between the inlet of the nozzle and extraction column was raised to 6.0 MPa by adjusting the volume flow of the solution.

The atomization of an injected solution without pulsation led to a decrease in particle size with increasing spraying pressure (Fig. 3a). As the spraying pressure was increased, an increasing amount of energy was brought into the system which resulted in a higher specific surface of the atomized liquid, and thus the particle size decreased. In contrast, when the polymer solution was applied by the pulsating diaphragm pump, the particle size increased with increasing spraying pressure. This effect may be due to eddies in the outlet region of the nozzle. The polymer/dichloromethane droplets in this area could collide and flow together. With an increasing solution flow resulting in increasing spraying pressure, the coalescence increases due to the higher velocities of the atomized droplets. The application of the polymer solution without pulsation exhibited a lower agglomeration tendency of the product (Fig. 3b) and led to a decrease in polydispersity (Fig. 3c). Both effects could be attributed to the occurrence of eddies in the proximity of the nozzle.

3.1.2. Carbon dioxide saturation of the spraying solution

Liquids are able to dissolve high amounts of gases at

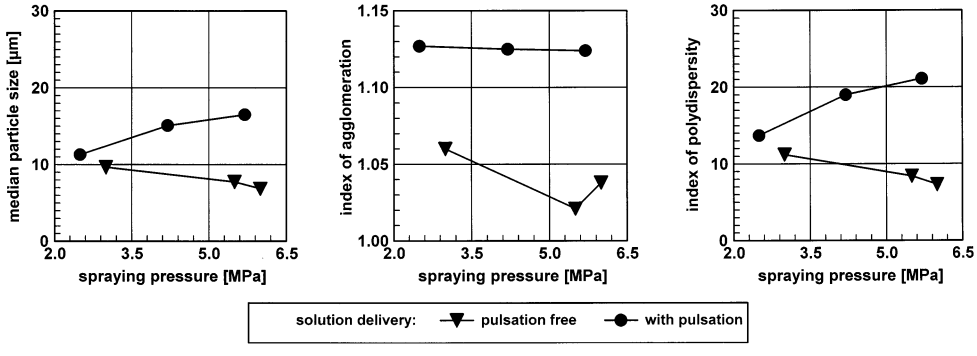

Fig. 3. Effect of spraying pressure and pulsation of the polymer solution on the (a) mean particle size, (b) agglomeration, and (c) polydispersity of the product ($\rho_{CO_2} = 438$ kg/m^3, $d_N = 0.3$ mm).

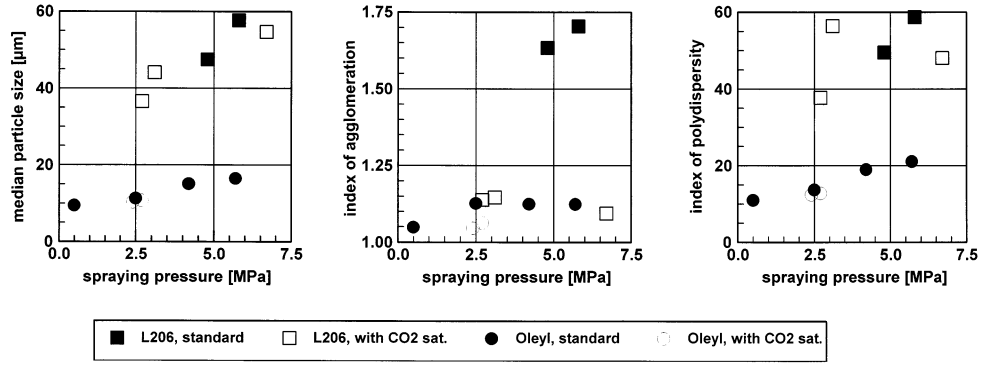

Fig. 4. Effect of spraying pressure and carbon dioxide saturation of the polymer solution with 5 MPa on (a) mean particle size, (b) agglomeration tendency, and (c) polydispersity for two polymers ($\rho_{CO_2} = 438$ kg/m^3, $d_N = 0.3$ mm, solution delivery with pulsation).

elevated pressures. Gallagher et al. [15] measured in a system consisting of carbon dioxide/dimethylformamide, a volume expansion of liquid up to 250%. In a mixture of 90% ethanol, 10% water and carbon dioxide at 45°C and 5.0 MPa, the volume expansion was 30%. The volume expansion increased to 55% at a temperature of 35°C [27].

In all experiments, a polymer solution of 200 ml was used. The application of a carbon dioxide pressure of 5.0 MPa directly on this polymer solution led to a volume expansion which could not be measured in the feeding vessel itself (Fig. 2c). However, the spraying time of this solution was extended by 94%, which indicated that the fluid phase consisted of about 50% carbon dioxide. The dissolved carbon dioxide reduced the viscosity of the liquid, resulting in a decrease in spraying pressure with increasing carbon dioxide saturation of the polymer solution. The reduction of particle size by saturation of the polymer solution with carbon dioxide was independent of the applied saturation pressure. The size reduction, which could be reached by this method of feeding the polymer solution to the extraction vessel was small (from about 15 μm without, to about 10 μm with carbon dioxide saturation at 5.0 MPa).

The effect of increased spraying pressure on the particle size depended more on the nature of the polymer used than on its saturation with carbon dioxide in the feeding vessel. Fig. 4a shows the long chain polymer (Resomer L206) forming larger particles than the oleyl derivative with a lower molecular weight. Particle formation in both cases is independent of the carbon dioxide concentration in the polymer solution. The agglomeration tendency of the product decreased with carbon dioxide saturation of the polymer solution (Fig. 4b). The particle formation from a carbon dioxide saturated solution might be faster due to a lower amount of carbon dioxide diffusing into the droplets, leading to a faster hardening of the surface and avoiding agglomeration of the particles. The polydispersity of the product was not affected by carbon dioxide saturation (Fig. 4c).

The porosity of the particles could be calculated by comparison of the total surface area and outer surface area. If the total surface area is in the same order as the outer one, the particles are free from pores. The BET measurement is unable to detect gas bubbles inside of the particles. However, the density of the polymeric material did not change after utilizing the ASES process, so that gas bubbles inside of the particles can be excluded. The total surface area of the particles modified with oleyl alcohol was 20% larger than the outer surface area. The difference between the total outer surface areas of the particles for the Resomer L206 was 50% (Fig. 5). These results could be explained by particles which were not perfectly spherical or adhesive. Even the saturation of the polymer solution with carbon dioxide was not found to affect the porosity of the particles.

3.1.3. Nozzle geometry

After studying solution delivery, the influence of the nozzle geometry was examined. Unitary nozzles with opening diameters of 0.1, 0.3, 0.5 and 0.8 mm were used.

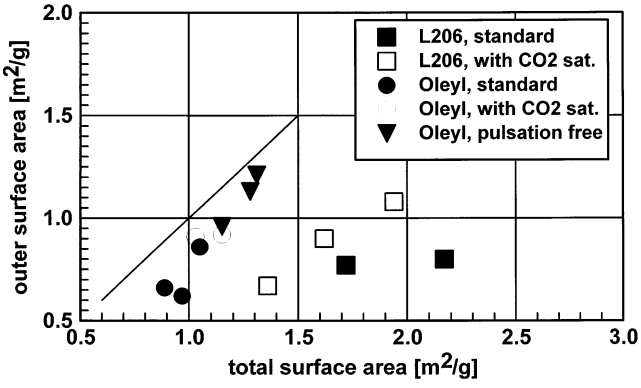

Fig. 5. Comparison of total and outer surface area of the product.

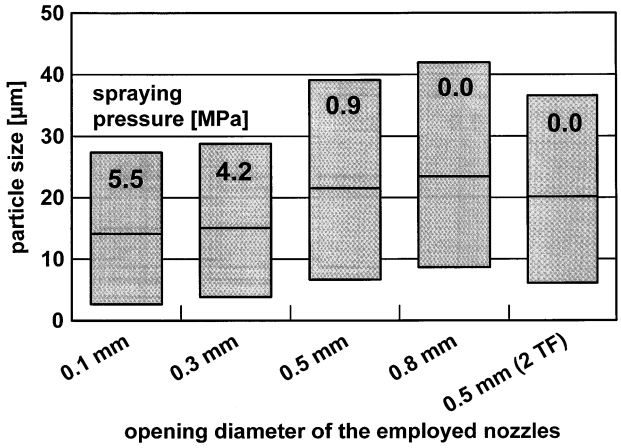

Fig. 6. Applied nozzles in the spraying process (the numbers in the bars represent the spraying pressure, 2 TF, twin-fluid nozzle; $\rho_{CO_2} = 438$ kg/m^3, solution delivery with pulsation, bar begins with the $\times 10$ quantile and ends with the $\times 90$ quantile, the line in the middle represents the median particle size).

Additionally a twin-fluid nozzle with carbon dioxide as the atomizing gas (40 kg CO_2/h, 0.5 mm opening diameter) was employed. The comparability of the nozzles was limited due to the adjustable spraying pressure. Even using the maximum pump rate of the solution resulted in spraying pressures below 1 MPa for nozzles with an opening diameter $d_N > 0.5$ mm. Utilizing the 0.1 mm nozzle, a high spraying pressure (5.5 MPa) was measured at the lowest adjustable pump rate. Considering that the particle size increased with increasing spraying pressure (solution application with pulsation), then the choice of the nozzle diameter had a slight influence on the particle size (Fig. 6). However, the twin fluid nozzle did not reduce the particle size in comparison to the corresponding unitary nozzle. Nozzle geometry had no influence on agglomeration tendency and polydispersity of the product.

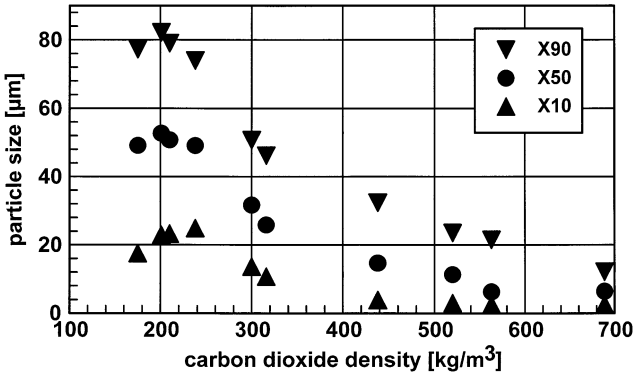

Fig. 7. Particle size versus carbon dioxide density ($\rho_{CO_2} = 438$ kg/m^3,

3.2. Carbon dioxide extraction conditions

3.2.1. Carbon dioxide density

Dixon et al. [17] found a decrease in particle size for the mixture of polystyrene in toluene solution and carbon dioxide if the density of the extracting gas was raised. The authors presumed the rapid atomization was controlled by inertial and interfacial forces. With increasing carbon dioxide density, the inertial force increases and the interfacial force decreases due to the higher miscibility of carbon dioxide and toluene. Hence

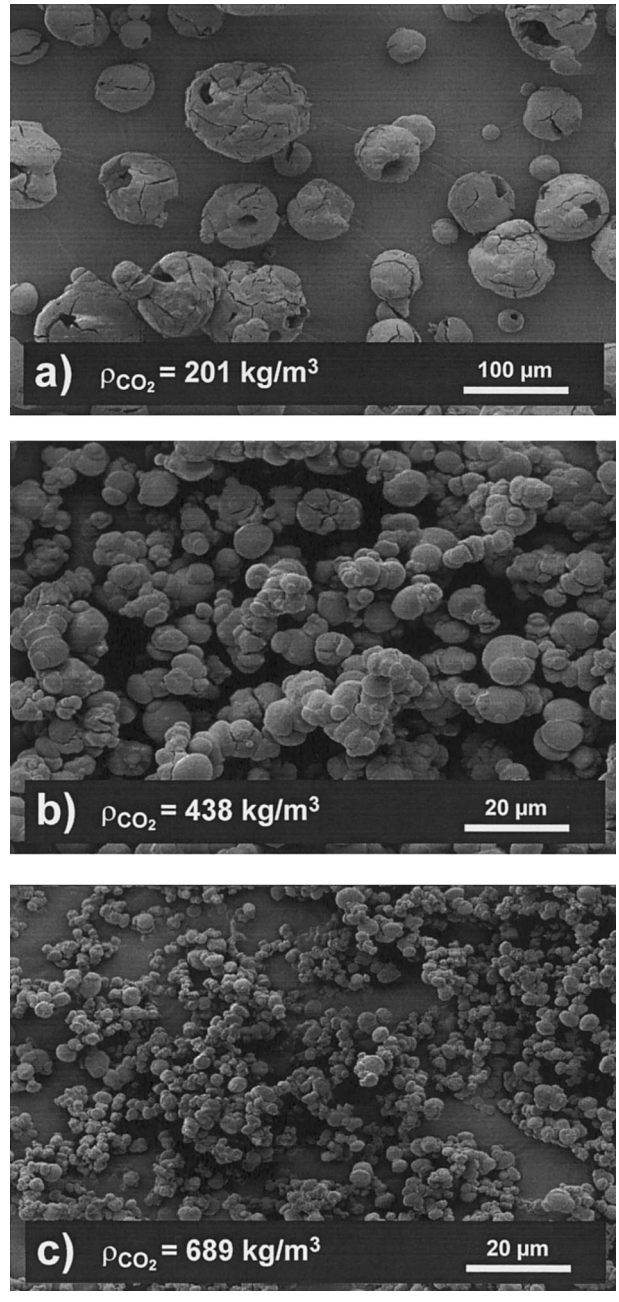

Fig. 8. SEM Photographs of microparticles produced with different carbon dioxide densities.

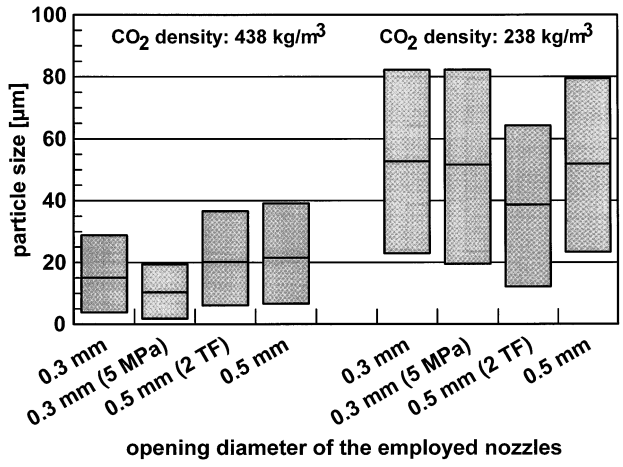

Fig. 9. Comparison of different nozzles at two extraction conditions (below the bars the opening diameter of the nozzle is given, 2 TF, twin-fluid nozzle; 5 MPa, polymer solution was saturated with 5.0 MPa carbon dioxide pressure).

it follows that the liquid jet disintegrates more rapidly and into smaller droplets if the carbon dioxide density is raised [17,28]. An investigation of L-PLA, which was sprayed from dichloromethane solution into carbon dioxide [19], resulted in increased particle sizes with increasing carbon dioxide density at constant temperature. The authors attributed the effect to mass transport mechanisms rather than to jet breakup and hydrodynamics. They observed the smallest particles at production conditions very near to the critical point of carbon dioxide, where rapid mass transfer was expected to result in high supersaturation ratios, high nucleation rates and short times for the nuclei to grow [19]. Bleich et al. [23,24] did not find a dependency of carbon dioxide density on particle size if the polymer Resomer L206 was sprayed from a dichloromethane solution into supercritical carbon dioxide at a range of density from 500 to 810 kg/m^3.

In the present study the extraction pressure was varied from 7.0 to 12.5 MPa. In all cases, the temperature was below 45°C to insure that the glass transition temperature of the polymer was higher than the carbon dioxide temperature to avoid highly agglomerated products. The temperature could not be lowered below 30°C due to technical limitations of the apparatus. The results showed a distinct dependence between particle size and carbon dioxide ex1traction density (Fig. 7). Microparticles manufactured at a carbon dioxide density of 560 kg/m^3 had a mean particle size of 6 μm. According to Bleich et al. [24], a further increase indensity did not effect the particle size. The particle size increased if the density was lowered below 560 kg/m^3. However, at a carbon dioxide extraction density lower than 250 kg/m^3, the particles showed a constant size in the range of 50 μm. Below a density of 180 kg/m^3, no particles could be formed. The solubility of dichloromethane in the carbon dioxide was too low to dissolve the injected liquid. Thus, conglutinated film-like polymer structures were found on the bottom of the column. The morphology of the particles sprayed in moderately compressed (438 kg/m^3) and highly compressed (689 kg/m^3) carbon dioxide was nearly equal (Fig. 8b and c). Particles produced at a low carbon dioxide density (201 kg/m^3) showed cracks and holes (Fig. 8a). The optical appearance of this product was similar to spray dried formulations.

3.2.2. Nozzle geometry

Further experiments were carried out with different nozzles at a carbon dioxide density of 238 kg/m^3 (p = 7.5 MPa, T = 40°C). The saturation of the polymer solution with 5.0 MPa of carbon dioxide and the employment of a 0.5 mm diameter nozzle did not influence the particle size in comparison to the 0.3 mm nozzle (Fig. 9). Smaller particles were produced by using the twin-fluid nozzle. At the lower carbon dioxide densities, a spray drying mechanism for particle formation was found. The high shear stress in the twin-fluid nozzle was responsible for the decrease in particle size. However, at a carbon dioxide density of 438 kg/m^3 the particle formation followed a precipitation process, because the shear stress in the twin-fluid nozzle did not influence the particle size.

4. Conclusions

ASES is a suitable and promising process to produce biodegradable polymeric microparticles. The median particle size can readily be adjusted in the range 6–50 μm by varying the process parameters. The saturation of the polymer solution with carbon dioxide up to 5.0 MPa did not affect particle size and morphology, and the agglomeration of the product decreased. The carbon dioxide density showed a strong influence on particle size. Particles produced at carbon dioxide densities
higher than 560 kg/m^3 had a mean diameter of 6 μm. Decreasing the carbon dioxide density led to an increase in particle size until a density of 250 kg/m^3 was reached. Between 180 and 250 kg/m^3, the produced particles had a mean diameter of 50 μm. These particles displayed cracks and holes. All other batches consisted of non porous microparticles with a smooth surface. The observed effects indicated that the particle formation could be associated with a precipitation process at high carbon dioxide densities and with a spray drying process at low carbon dioxide densities.

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