Applicability of the Rapid Expansion of Supercritical Solutions with a Solid Cosolvent (RESS-SC) Technique to the Production of Theophylline Nanoparticles

Hirohisa UCHIDA [1*], Masamichi NISHIJIMA [1], Kyohei SANO [1], Kohei DEMOTO [1], Junichi SAKABE [2], Yusuke SHIMOYAMA [2]

[1] Department of Chemistry and Material Engineering, Shinshu University, 17-1 Wakasato-4, Nagano-city, Nagano 380-8533, Japan
[2] Department of Chemical Engineering, Tokyo Institute of Technology, 2-12-1 Ookayama, Meguro-ku, Tokyo 152-8550, Japan
* Corresponding author, e-mail : uchida@shinshu-u.ac.jp ; Fax : (+81)-26-269-5402

ABSTRACT

Rapid expansion of supercritical solutions (RESS) technique using supercritical carbon dioxide (sc-CO_2) has been expected as an effective and environmentally friendly nanoparticle design method. However, due to extremely low solubility of polar drugs in sc-CO_2, the RESS technique has limited commercial applicability. In order to enhance the solubility of drugs in sc-CO_2, a liquid cosolvent is generally added to sc-CO_2. The addition of a liquid cosolvent to sc-CO_2, however, often have a bad influence on the properties such as particle sizes, morphology, crystal structure and purity of particles produced by the RESS technique. As a modified RESS technique to overcome these problems, RESS with solid cosolvent (RESS-SC) in which a solid cosolvent such as L-menthol is added to the solutions was proposed by Thakur and Gupta (Ind. Eng. Chem. Res., 44, 7380 (2005)).

In this work, the production of theophylline nanoparticles via the RESS-SC technique using sc-CO_2 with L-menthol or vanillin as a solid cosolvent has been examined. Moreover, the effects of the particle collection conditions such as collection temperatures and spray distances on the theophylline nanoparticles produced were investigated in order to elucidate crystal growth mechanism of the present process. The sc-CO_2 saturated with the solute and the solid cosolvent was expanded rapidly through a drilled capillary nozzle ($D = L = 50$ μm) into a collection chamber with a jacket to control the collection temperature so that it becomes a given value. The particles produced were collected on a membrane filter (pore size: 100 nm) set at the outlet of the collection chamber. The experimental temperature and pressure are 313.2 K and 22.0 MPa, respectively. The particle collection temperatures are 265.2, 283.2 and 303.2 K and the spray distances are 3, 5 and 7 cm. A liquid phase appeared at the pre-expansion section in the case of using L-menthol as a solid cosolvent and this suggests that it is unsuitable for a solid cosolvent in the present conditions. In the case of using vanillin as a solid solvent, on the other hand, we have successfully produced nano-sized particles of around 85 nm, which are about one-third as large as those produced by the conventional RESS technique. The particle collection conditions such as the collection temperatures and the spray distances have negligible effects on the particle sizes. Namely, the effects of crystal growth in the collection vessel on the crystallized particles would be very small. This result suggests that the crystal growth of theophylline particles crystallized in the collection vessel may be blocked by the crystallized vanillin particles those surrounds theophylline particles.

INTRODUCTION

In the pharmaceutical industry, the particle sizes of pharmaceutical substances are of importance for their bioavailability. The bioavailability can be improved by reducing the particle size of the pharmaceutical substances. The development of methods for the preparation of nanoparticles has received a great deal of attention. Various micronization

techniques using supercritical fluids have recently been proposed [1-3]. In particular, recently new micronization techniques using supercritical fluids such as rapid expansion of supercritical solutions (RESS), supercritical antisolvent recrystallization (SAS), and particles from gas saturated solutions (PGSS) processes have been proposed and the techniques have been paid much attention and have been expected as effective and environmentally friendly particle design methods [1-3]. The most well-known micronization technique based on supercritical technique would be the RESS technique using supercritical carbon dioxide (sc-CO_2). A solute is dissolved in a sc-CO_2 and the solution expands rapidly through a nozzle. Due to this strong and rapid pressure, and temperature drop, the solute dissolved becomes insoluble in a low-pressure gas and thus a high supersaturation is created in the spray jet. Fast nucleation and growth of crystalline particles occur. Consequently, micro- or nano-sized particles can be produced because of the very high supersaturation and a very small growth time. After the process, the CO_2 is in the gaseous phase so that solvent free and dry products can be achieved and an additional wash and drying process is not necessary. The RESS technique has been paid much attention and has been expected as an effective and environmentally friendly particle design method. We successfully produced nano-sized particles of racemic ibuprofen (mean particle size: about 200 nm) and theophylline (mean particle size: about 270 nm) with the narrow particle size distribution and proposed the strategy for nanoparticle design in our previous work [2-4]. However, due to extremely low solubility of polar drugs in sc-CO_2, the RESS technique has limited commercial applicability. In order to enhance the solubility of the drugs in sc-CO_2, a liquid cosolvent is generally added to sc-CO_2. The addition of the liquid cosolvent to sc-CO_2, however, often have a bad influence on the properties such as particle sizes, morphology, crystal structure and purity of particles produced by the RESS technique. As a modified RESS technique to overcome these problems, RESS with solid cosolvent (RESS-SC) in which a solid cosolvent such as L-menthol is added to the solutions was proposed by Thakur and Gupta [5].

This work has aimed to examine the effects of the particle collection conditions such as collection temperatures and spray distances on the theophylline nanoparticles production by RESS-SC technique using sc-CO_2 with L-menthol or vanillin as a solid cosolvent. In this work, the production of nanoparticles of theophylline as a model compound has been performed. Theophylline is one of the xanthine derivatives and is bronchodilator to treat asthma. The choice of a proper solid cosolvent is a key element in the RESS-SC process. Requirements for the selection of the solid cosolvent are as follows: it should (i) have good solubility in sc-CO_2, (ii) not become a liquid phase during the process, (iii) have a sufficiently high vapor pressure so that the solid cosolvent can be removed from a crystallized product only by sublimation, (iv) be nonreactive with the drug and CO_2, (v) be nontoxic, (vi) have an affinity with the drug and (vii) not form a solid solution with the drug. We examined the effects of a solid cosolvent such as L-menthol or vanillin because the substances may satisfy the above requirements. They have also widely been used in several industries such as food and pharmaceutics.

EXPERIMENTAL

Materials

Theophylline (supplied by Wako Pure Chem. Ind. Ltd.; its purity is greater than 99%) was used as a solute. L-menthol (supplied by Tokyo Chemical Industry Co., Ltd.; its purity is greater than 99%) and vanillin (Sigma-Aldrich; its purity is greater than 99%) were used as solid cosolvents. High-purity CO_2 (its purity is greater than 99.99%, supplied by Showa Denko Gas Products Co., Ltd.) as received was used as a solvent.

Apparatus and Procedures

The apparatus is basically a flow-type and consists of a section of preparing a supercritical solution saturated with a solute, that of the production of solid particles, and that of the collection of particles produced. The apparatus is shown schematically in **Figure 1**. From the CO_2 cylinder (1), CO_2 was supplied and was liquefied through the cooling unit (4). The liquefied CO_2 was sent to the preheater (6) by the feed pump (5) (Nihon Seimitsu Kagaku Co., Ltd., NP-KX-500). When the CO_2 passed through the preheater in a thermostated air bath (10) at an experimental temperature which was controlled within ±0.1 K, it became a sc-CO_2. We used an equilibrium cell (9) (Taiatsu Techno Co., SC-CO_2 Compact Reactor), which was made of SUS316, and inner diameter, height, and volume of which were 40 mm, 65 mm, and 80 cm^3, respectively. The mixture of a solid solute and a solid cosolvent was packed into the cell with glass beads to prevent channeling. The molar ratio of the solid solvent to the solid solute in the mixture is about 1:1. The cell was placed in the thermostated air bath. Valve V3 was closed, and valves V2 and V4 were opened to admit sc-CO_2 into the equilibrium cell. When sc-CO_2 passed through the equilibrium cell, the sc-CO_2 was in contact with the solid solute and the solid cosolvent under an equilibrium pressure. The equilibrium pressure was measured by the pressure transducer (PI) (Setra Systems. Inc., Model C280E) with accuracy ±0.038 MPa. The sc-CO_2 saturated with the solid solute and the solid cosolvent was expanded rapidly through a drilled capillary nozzle (13) (D = L= 50 μm) into the collection chamber (14) with a jacket to control temperature after passing through the heated tube (pre-expansion section) at a selected pre-expansion temperature. The particles produced were collected on a membrane filter (15) (pore size: 100 nm) set at the outlet of the collection chamber and the membrane filter was sucked by a vacuum pump (17) (ULVAC KIKO, Inc., Direct Drive Oil-Sealed Rotary Vacuum Pump GLD-051) to keep the pressure in the collection chamber at atmospheric pressure. The distance between the expansion nozzle and the membrane filter (spray distance) was constant at a selected distance. Before analyzing the produced particles to examine the size, the morphology and the crystal structure, the particles were dried in a vacuum dryer (temperature: 313.2 K, pressure: 0.006 MPa, time: 24 h) to remove the solid cosolvent using sublimation.

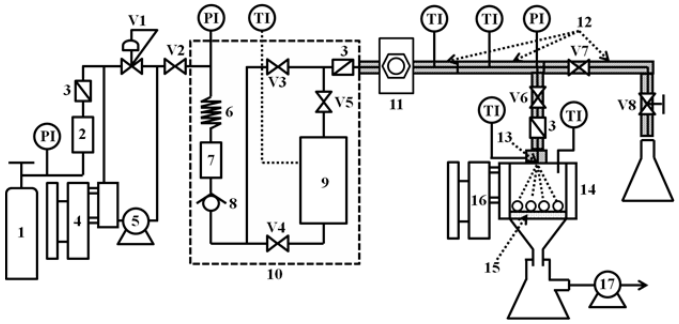

Figure 1. Schematic diagram of experimental apparatus
(1) CO_2 cylinder, (2) dryer, (3) filter, (4) cooling unit, (5) feed pump, (6) preheater, (7) buffer, (8) stopper, (9) equilibrium cell, (10) thermostated air bath, (11) view cell, (12) heater (13) expansion nozzle, (14) collection chamber with a jacket to control temperature, (15) membrane filter, (16) jacket temperature controller, (17) vacuum pump, (PI) pressure indicator, (TI) temperature indicator, (V1) back-pressure regulator, (V2–V7) stop valves, (V8) needle valve to control the flow rate.

The morphology of original and particles produced was observed with a scanning electron microscope (SEM) (Keyence Co., VE-9800). The SEM samples were prepared by the covering with a thin layer of platinum and palladium using a sputter coater. Particle sizes and the size distribution were determined by an image analysis of photomicrographs that is the counting at least 800 particles from the photomicrographs. The particle size was determined by the diameter based on the Ferret diameter. The crystal structure of the particles were analyzed with a powder X-ray diffractometer (XRD) (Rigaku Co., Miniflex II).

The temperature and pressure of the section of preparing the supercritical solution of the solid solute and the solid cosolvent; the equilibrium temperature and pressure, were 313.2 K,

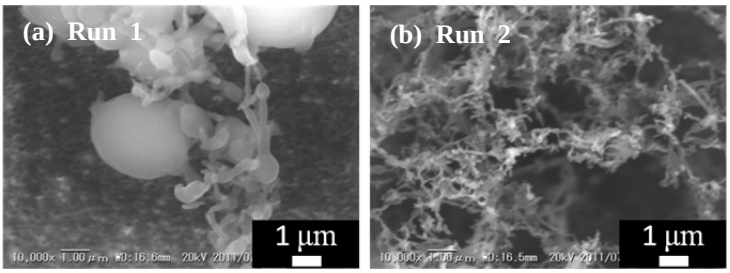

Figure 2 SEM photographs of theophylline particles produced by the RESS-SC process using L-menthol at the collection temperature 265.2 K and the spray distance 3 cm; (a) Run 1, (b) Run 2.

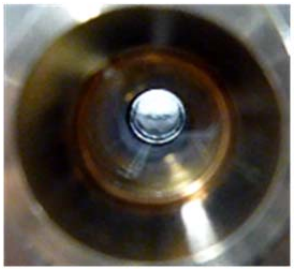

Figure 3 Picture of the gas-liquid two-phase fluid of CO_2 + L-menthol as observed through the view cell.

Table 1 Experimental Conditions and Results of the Mean Particle Size of the theophylline Particles Produced by the RESS-SC process (solid cosolvent: Vanillin).

Collection Temp. (K)	Spray distance (cm)	Mean particle size (nm)	CV (%)
265.2	3	85	21
283.2	3	87	20
303.2	3	90	19
265.2	5	89	19
265.2	7	88	21

Original sample ; mean particle size = 71 μm, CV = 65%

and 22.0 MPa, respectively. The temperatures of the pre-expansion section and the nozzle were constant by 338.2 K and 343.2 K respectively. The spray distances were 3, 5 and 7 cm and the collection temperature were 265.2, 283.2 and 303.2 K to examine the effect of the parameters on the production of particles by the RESS-SC technique.

RESULTS AND DISCUSSION

(1) Solid cosolvent: L-menthol

The experimental results are shown in **Figures 2 and 3**. Figure 2 shows the lack of reproducibility of crystal properties such as morphology and size of the theophylline particles produced by the RESS-SC technique using L-menthol. This problem was caused by the appearance of a liquid phase at the pre-expansion section and in the nozzle as shown in Figure 3 and this suggests that L-menthol is unsuitable for a solid cosolvent in the present conditions.

(2) Solid cosolvent: vanillin

The experimental results are shown in **Table 1** and **Figures 4-6**. The mean size of theophylline particles of the original sample was 71 μm as shown in Figure 4 and the particle size distribution (CV: 65%) was also shown in **Figure 5**. Table 1 shows the effect of the collection temperatures and the spray distances on the theophylline nanoparticles production using the RESS-SC process. The particle size of the primary particles was very small (the mean size: about 85 nm) with the narrow particle size distribution (CV: about 21%) as shown in Figures 4 and 5, which was 800 times smaller than that of the original sample. Moreover, the sizes of the theophylline particles micronized by the RESS-SC process using vanillin were more than three times smaller than those of the theophylline particles obtained by the RESS process reported by Sakabe *et al.* (the mean size: about 270 nm) [4]. As can be seen from Figure 6, XRD analysis showed that the crystal structure of the theophylline particles did not change after the micronization using the present RESS-SC process. Table 1 shows that the collection temperatures and the spray distances have negligible effects on the particle sizes. Namely, the effects of crystal growth in the collection vessel on the crystallized theophylline particles would be very small. On the other hand, as for the micronization of theophylline using the RESS process reported by Sakabe *et al*, the collection temperatures have negligible

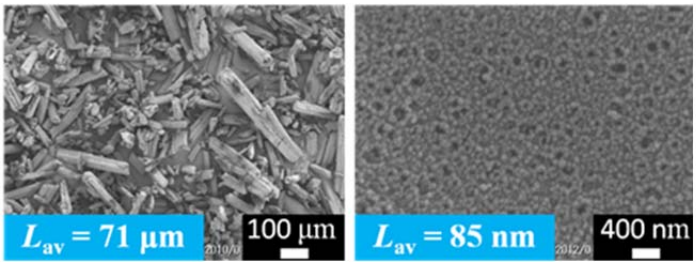

Figure 4 SEM photographs of theophylline particles; left: original, right: produced by the RESS-SC process at collection temperature 265.2 K and spray distance 3 cm (solid cosolvent: vanillin).

effects on the particle sizes, though the mean sizes of the micronized particles increased with increasing of spray distances [6]. In other words, the mean sizes of the micronized particles increased with increasing of residence time. In this work, not only the collection temperatures but also the residence time that is proportionate to the spray distances have negligible effects on the particle sizes. This result suggests that the crystal growth of theophylline particles crystallized in the collection vessel may be blocked by the crystallized vanillin particles those surrounds theophylline particles.

CONCLUSION

We showed that L-menthol is unsuitable for a solid cosolvent in the present conditions. On the other hand, the production of nanoparticles (mean size: 85 nm) of theophylline with narrow particle size distributions was successfully performed by the RESS-SC technique using sc-CO_2 with vanillin as a solid cosolvent. The particle sizes were about one-third as large as those produced by the conventional RESS technique. The RESS-SC technique will allow the formation of nano-sized particles, thus confirming the possibility of processing pharmaceutical materials with this technique.

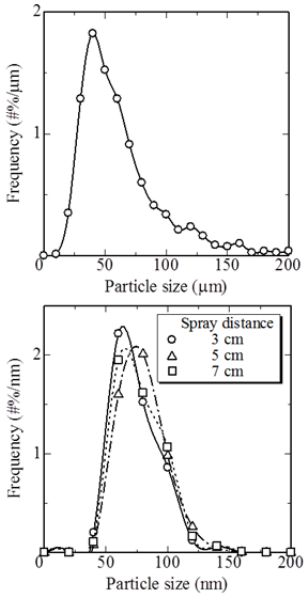

Figure 5 Particle size distributions of theophylline; upper: original, lower: produced by the RESS-SC process at the collection temperature 265.2 K (solid cosolvent: vanillin).

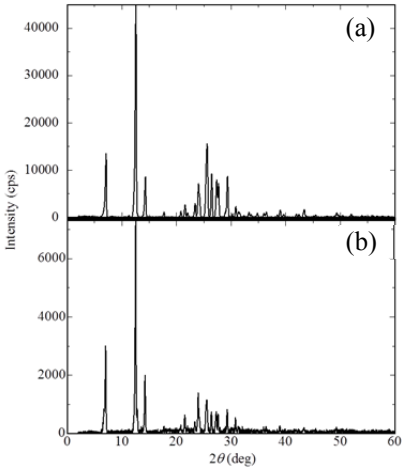

Figure 6 XRD patterns of theophylline (a) original sample, and (b) particles produced by the RESS-SC process at collection temperature 265.2 K and spray distance 3 cm (solid cosolvent: vanillin).

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