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CRYSTALLIZATION OF ACETAMINOPHEN IN NANO/MICRO SCALE USING SWIRL MIXING MICRO DEVICE UNDER PRESSURIZED CARBON DIOXIDE

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ABSTRACT

The fabrication of acetaminophen particles via supercritical antisolvent process with CO_2 as an antisolvent was studied. The experiments were conducted at temperatures of 35-50 ^oC and pressures of 8-15 MPa with 5-15 ml/min CO_2 flow rates. As a starting material, acetaminophen powder was dissolved in dimethyl formamide (DMF). Results of UV–vis spectrophotometry and GC-MS (gas chromatography mass spectrometry) analysis showed that there was no remaining DMF solvent in the acetaminophen particles products. It indicated that CO_2 has successfully removed DMF from acetaminophen particles products. The surface characterization by using fourier transform infrared spectroscopy (FT–IR) showed that the CO_2 solvent did not impregnate to the acetaminophen particles products. Results from scanning electron microscope (SEM) images showed that the acetaminophen particles products were successfully prepared in non-spherical shape morphologies with size less than 1 μm. Based on the result; this process seems a powerful method to modify the acetaminophen powder physically such as particle size reduction.

Keywords: supercritical antisolvent, acetaminophen, particle size, DMF, $SCCO_2$.

INTRODUCTION

Nano-and microparticles have been known to have widespread applications in pharmaceutical field and catalysis reaction. They also have been used in coloring industry to produce and improve the duration and the brightness of inks, toners and paints, and in the electric field to produce of the high-temperature superconductors (Reverchon *et al.*, 2008) [1]. Based on this application purpose, several techniques have applied to produce these particles, such as spray-drying, freeze-drying, liquid antisolvent crystallization, and ball milling processes. These techniques have several drawbacks, such as the coarse particles generation with broad particle size distribution, degradation of the product may occur due to mechanical or thermal stresses, or contamination of particles with organic solvents or other toxic substances [2]. In the spray drying process, the feed solution containing the solute is atomized via a nozzle into droplets. Due to the high surface area of these droplets and the contact with the drying gas during process, these droplets may dry rapidly. The rapid solvent removal to generate particle products may avoid the dried powder products from overheating or degrading products. However, Sosnik and Seremeta [3] reported that drug degradation is not anticipated and may occur during spray drying process. The wide size distribution of particles products and the high residual solvent in the final products when the liquid antisolvent crystallization technique was used [4]. The long–time process was also needed in this process. Therefore, different alternative techniques are being investigated. In addition, the morphology of particles may give a significant impact on the dissolution rate and can also have an intriguing effect on particle functions [5, 6]. Sen Gupta [6] reported that the particles morphology are important factors which influences on the particle behavior in blood flow in terms of cell–particle interactions, convective, and diffusive transport, lateral margination, target site, or cell binding and cellular internalization.

In the present work, supercritical carbon dioxide ($SCCO_2$) was employed as a media to produce particles from acetaminophen dissolved in dimethyl formamide (DMF) in nano- and microscales. Generally, there are two main techniques to produce particles by using supercritical fluid: rapid expansion of supercritical solutions (RESS) and supercritical antisolvent (SAS) techniques. In RESS technique, supercritical fluid was employed as a solvent while it would be as an antisolvent in SAS technique [7]. A supercritical fluid including $SCCO_2$ can be defined as any fluid which is at conditions above its critical point. It has liquid-like densities with gas–like transport properties and moderate solvent power, which can be tuned with shifts in pressure and temperature. Due to these transport properties, supercritical fluid has been employed in different fields for different applications, such as extractions, chromatography, or particle generation [8-11]. When the supercritical fluid was applied to particles formation, the organic solvent used as a solvent media can be removed completely from the particles products due to the high solubility of this solvent in supercritical fluid. In addition, the amount of organic solvent used also can be reduced. In this work, CO_2 was used as an antisolvent to produce particles from acetaminophen in DMF via swirl mixing micro device under supercritical conditions. One of the important parts in the SAS device is mixing of organic solvent and supercritical fluid [12-14]. There are many types of mixer such as Y-shaped, nozzle-type, central-collision-type, and swirl mixer that have been

developed in order to reduce the formation of large particle products and wide size distribution during particle production. Kawasaki *et al*. [13] informed that swirl mixer is one of effective mixer to fabricate small sized particles with a narrow size distribution. They applied micro swirl mixer for fabricating fine metal oxide particle by continuous supercritical water. The result showed that employing micro swirl mixer with diameter of 0.5 mm generated the average particle size of 20 nm.

Acetaminophen (also recognized as paracetamol, 4-acetaminophenol, 4-hydroxy acetanilide) is one of the most widely used medications in the world. It has been used as an antipyretic, non-opioid analgesic, and non-steroidal anti–inflammatory drug (NSAID) [15]. The chemical structure of this compound comprises of an aromatic ring substituted in para orientation by two groups: an acetamide and a hydroxyl. Due to the multiple conjugation on this compound involving the benzene ring, the hydroxyl oxygen, the amide nitrogen, and the carbonyl carbon and oxygen, acetaminophen may have a high reactivity [16]. The benzene ring is highly reactive to electrophilic aromatic substitution in oxygen, nitrogen, and the hydroxyl acidic bonds. It has high solubility in DMF; the solubility is >1000 g of acetaminophen/kg of solvent [17]. DMF is completely miscible in water and is both chemically and thermodynamically stable. This solvent was also called as the universal solvent since it can dissolve a wide variety of organic and inorganic materials. It also was applied primarily in the pharmaceutical processing [18, 19]. Several research groups have been conducted recrystallization of acetaminophen using CO_2 as an antisolvent under supercritical conditions [20-22]. Fusaro *et al*. [21] performed precipitation experiments of acetaminophen from solution in acetone using compressed CO_2 as an antisolvent. The recrystallization process was conducted in a batch system. They informed that the different operating conditions promote to different morphologies in a certain way and the specific antisolvent addition rate might be used to control the final particle size as well as the particle morphology. Rossmann *et al.* [22] also performed the $SCCO_2$ antisolvent to crystallize acetaminophen particles at pressures of 10-16 MPa and temperature of 40 ^oC. Several organic solvents such as ethanol, acetone and mixtures of ethanol and acetone were used as solvents. The acetaminophen particles morphologies are different when the varying solvent composition was applied; however the average size of the acetaminophen particles products is not much influenced. They concluded that the different strategies can be applied to tune the properties of acetaminophen particles products through the $SCCO_2$ antisolvent process.

EXPERIMENTAL SECTION

Materials

Acetaminophen ($C_8H_9NO_2$; 97.0%), dimethylformamide (C_3H_7NO; 99.5%), ethanol (C_2H_5OH; 99.5%) were obtained from Wako Pure Chemicals Industries Ltd., Japan. They were used without further purification. Carbon dioxide (CO_2: 99%) was supplied by Sogo Kariya Sanso, Inc. Japan. For preparing the acetaminophen solution, acetaminophen powder was dissolved in DMF to achieve concentrations of 7.5 mg/ml of solvent. This concentration was selected based on the previous researcher's report [20].

Methods

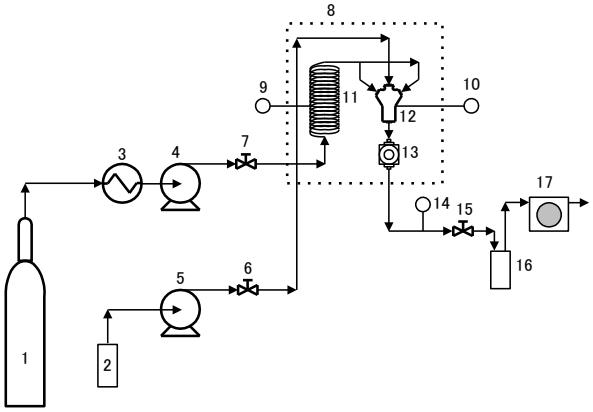

Figure-1. Experimental apparatus: 1. CO_2 cylinder; 2. Feed solution; 3. Chiller; 4, 5. High pressure pump; 6, 7. Needle valve; 8. Oven; 9, 10. Temperature monitor; 11. SUS-316 pre-heater; 12. Swirl mixer; 13. Particles collector; 14. Pressure monitor; 15. BPR; 16. Solvent trap; 17. Wet flow meter.

Figure-1 described the crystallization apparatus scheme using swirl mixing micro device under pressurized carbon dioxide. The main apparatus was high pressures pump for CO_2 and acetaminophen solution (PU–2086, Jasco, Japan; LC-6AD, Shimadzu, Japan), a heating chamber (WFO-400; EYELA, Tokyo, Japan), a swirl mixing device (4-1/16YSM-0.8-0.5-S, Sugiyama Shoji Co., Ltd., Japan) and back pressure regulator (BPR; AKICO, Tokyo, Japan). The coil preheater made of 1/8 inch stainless–steel tubing (SUS316) with 300 cm length was placed in the heating chamber to introduce the CO_2 before entering to swirl mixing micro device. K–type thermocouples were inserted in the preheater and attached in the swirl mixing micro device to monitor the temperature during experiment. To monitor the particles generation pressures, the digital pressure gauge (NS NPG-500A, Nihon Seimitsu Kagaku co., ltd, Japan) was assembled on the 1/16 inch stainless-steel tubing (SUS316) and placed between particles products collector and BPR. In this work, the particle generation from acetaminophen solution with $SCCO_2$ antisolvent was carried out at temperatures of 35-50 ^oC and pressures of 8-15 MPa. The CO_2 and the acetaminophen solution flow rates were 5-15 ml/min and 0.25 ml/min, respectively. The crystallization process can be explained briefly as follow. Initially, the power of heating chamber was switched on to heat the swirl mixing micro device including preheater to a desired temperature. Once the desired temperature was achieved, CO_2 was pumped into the crystallization apparatus system via the 1/16 inch stainless-steel capillary

tube at a desired pressure. A BPR was employed to keep a constant pressure during crystallization process. Next, the acetaminophen solution was injected into the swirl mixing micro device upon the attainment of the desired conditions. The delivery of acetaminophen solution was finished after 60 min; the fresh CO_2 was still pumped at around 60 min to remove the residual DMF solvent in the particles products. This step was needed to avoid the redissolved of particles products in DMF solvent during depressurization process. After the process was completed, the CO_2 flow was stopped and the crystallization apparatus system was slowly depressurized to atmospheric pressure. Then the particles products were collected in the bottles and stored in vacuum desiccator at room temperature. These processes were done until analysis.

Analytical methods

To identify the remaining DMF solvent in product, the acetaminophen particles products were dissolved in ethanol and analyzed by UV-vis spectrophotometry (V-550, Jasco Corporation, Japan). This analysis is simple to use and most analytes can be detected. The solution of acetaminophen particles products was placed and analyzed in a quartz cuvette with a 1 cm path length. By using a PC-driven scanning spectrophotometer operating in the fast scan mode, the allowing spectra of between 190 and 800 nm with 10 nm min-1 of bands were monitored and recorded. The acetaminophen particles products was also analyzed by using GC-MS (gas chromatography mass spectrometry) Hewlett Packard model 6890 series GC system and 5973 mass selective detector with a DB–5 MS capillary column (J&W Scientific, length 30 m, i.d. 0.25 mm, film 0.25 μm). The GC-MS carrier gas was He at a flow rate of 2 ml/min. 1 μl a solution in ethanol was injected. The injector temperature was 250 ^oC and the chromatographic exit to mass spectrometer interface temperature was 300 ^oC. The GC oven temperature was held at 45 ^oC for 1 min then programmed to increase at 5 ^oC/min to 300 ^oC. Electron ionization and positive ion mode were used. The NIST (National Institute of Standards and Technology) library of mass spectroscopy was used to identify the compounds. In order to understand the structure of acetaminophen after treatment by $SCCO_2$ antisolvent, the acetaminophen particles products collected at each operating condition were analyzed using a Spectrum Two FT-IR spectrophotometer (PerkinElmer Ltd., England). The spectra were measured in attenuated total reflectance (ATR) mode (golden single reflection ATR system, P/N 10500 series, Specac) at 4/cm resolution. The scanning wave number ranged from 4000 to 400/cm. The morphology of the acetaminophen particles products was observed using a scanning electron microscope (SEM; S-4300, Hitachi, Japan) after gold coating. The diameters of them were measured from the SEM images using image analyzer software (Image J 1.42).

RESULTS AND DISCUSSIONS

Product identification

In supercritical antisolvent, including CO_2 as a solvent, mass transfer is to be a key factor and it occurs between a droplet of organic solvent from feed solution and a compressed antisolvent to generate particles. This process was affected by the densities differences between solvent and antisolvent, viscosity, diffusivity, droplet or particle diameter, and solvent flow rate [23]. When $SCCO_2$ antisolvent was applied in pharmaceutical field application, it is able to produce various drugs formulation in nano-micro scale. Figure-2 shows the acetaminophen particles obtained (A) before and (B) after $SCCO_2$ antisolvent treatment. It was clearly observed that the original acetaminophen particles seem to have needle shape morphologies with size between 10-87 μm. Even, Biazar *et al.* [24] reported that acetaminophen particle size has been shown to follow a normal distribution with a mean particle size of 100 μm. While the particles size of acetaminophen after $SCCO_2$ antisolvent treatment was less than 1 μm with non–spherical shape morphologies. It indicates that the size of acetaminophen particle can be reduced by using CO_2 as an antisolvent under supercritical conditions.

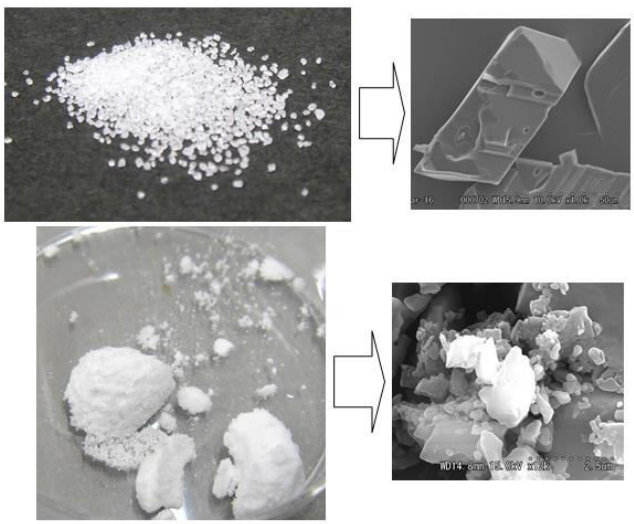

Figure-2. Acetaminophen particles before (A) and after (B) treatment by $SCCO_2$ antisolvent at pressure of 10 MPa and temperature of 40 ^oC with CO_2 flow rate 10 ml/min.

Figure-3 shows the UV–vis spectra of acetaminophen particles before and after $SCCO_2$ antisolvent treatment. Generally, the UV-vis spectrophotometry was applied to monitor chemical reaction, dissolution testing, and color determinations. This analysis is a non–destructive analytical technique and also an easy in sample preparation. Therefore, this tool was also the most frequently used technique in pharmaceutical analysis [25, 26]. Since this analysis is typically conducted on liquid solutions or suspensions, the acetaminophen particles before and after $SCCO_2$ antisolvent treatment were dissolved in ethanol with

various concentrations. The UV-vis spectrophotometry for pharmaceutical applications concern light in the wavelength range 190-800 nm, qualitatively, acetaminophen in ethanol will be identified at 249-250 ± 0.5 nm [25]. As described in Figure-3, the peak intensity at around 250 nm correspond to the acetaminophen compound in the ethanol solution was clearly detected. Conversely, the peak intensity at around 270 nm relation to the existence of DMF compound was not detected.

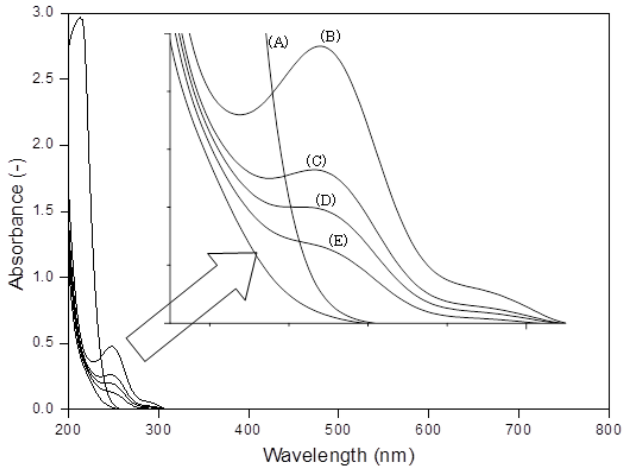

Figure-3. UV-vis spectra of acetaminophen particles products at temperature of 40 ^{o}C and pressure of 10 MPa with various CO_2 flow rates: (A) DMF in ethanol, (B) Acetaminophen, (C) 15, (D) 10, and (E) 5 ml/min, respectively.

There was no remaining DMF solvent in the acetaminophen particles products. It indicated that the DMF solvent in the acetaminophen particles products which retained in the particles products collector by placing stainless filter was successfully removed with the flowing CO_2 under supercritical fluid conditions [27]. Similar results were also obtained when the acetaminophen particles before and after $SCCO_2$ antisolvent treatment were analyzed by GC–MS. The DMF compound which used as a solvent was not detected. Figure-4 shows the GC–MS spectra of them. The GC–MS is well–known as a powerful tool to identify aromatic and aliphatic compounds. Prior to analysis using GC–MS, about 1 mg of the acetaminophen particles before and after treatment by $SCCO_2$ antisolvent were also diluted with 2 ml ethanol at room temperature. Then, these solutions were injected in the GC–MS apparatus immediately. The identities of those compounds determined through a match of mass spectra in the GC-MS computer library are reliable. As shown in Figure-4A, cyanoacetophenone (1), acetaminophen (2), and metacetamol (3) were detected clearly on the GC–MS chromatogram with retention time 3.78, 27.76, and 30.83 min, respectively. These compounds were also found in the GC–MS spectra of acetaminophen particles products after treatment by $SCCO_2$ antisolvent with the same retention time (see Figures-4B, 4C and 4D). There was no peak correspond to the existence of DMF compound. These results suggested that the DMF solvent in the collected acetaminophen particles products has been removed with the flowing CO_2 under these conditions. Kim *et al.* [27] conducted experiment for nanoparticle production from atorvastatin calcium by the $SCCO_2$ antisolvent. They reported that the various organic solvents including DMF that used as a solution solvent could be removed completely by the flowing CO_2 at temperature of 40 ^{o}C and pressure of 12 MPa.

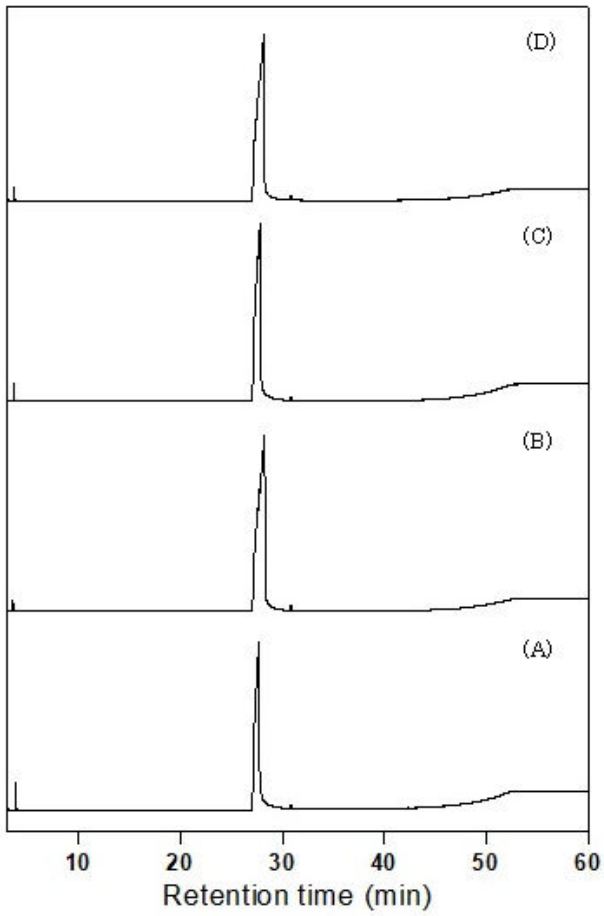

Figure-4. GC-MS chromatogram of acetaminophen particles before (A) and after treatment by $SCCO_2$ antisolvent at pressure of 10 MPa and temperature of 40 ^{o}C with CO_2 flow rates (B) 5, (C) 10, and (D) 15 ml/min, respectively.

Product characterization

In order to investigate the possibility of structural change of acetaminophen molecules after $SCCO_2$ antisolvent treatment, the acetaminophen particles products were characterized by FT-IR spectroscopy, in the wave numbers region of 4000-400/cm. Infrared spectroscopy is an analytical technique that allows to identify the unknown substances and the types of chemical bonds of the compounds in those substances content. Acetaminophen powder (without any treatment) was used directly as a control. FT–IR spectra of acetaminophen particles before and after $SCCO_2$ antisolvent treatment are

shown in Figure-5. Every molecule is consisted of many different chemical bonds, and these bonds are slightly elastic and can stretch, bend, or vibrate.

Table-1. Wave number assignments of FT–IR spectra.

Wave number [1/cm]	**Functional groups**
3500-600	O-H bonds
2800-3000	C–H Lipid region
1504	In–plane CH bending vibration from the phenyl rings
1482	Benzene
1456	CH_3 bending vibration (lipids and proteins)
1312-1317	Amide III band components of proteins collagen
1255	Amide III
1244/5	PO–2 asymmetric (phosphate I)
1206	Amide III Collagen
1180-300	Amide III band region
1145	Phosphate & oligosaccharides
1105	Carbohydrates
1030	Collagen
1020-50	Glycogen
1000-350	Region of the phosphate vibration carbohydrate residues attached to collagen and amide III vibration (in collagen)
1000-200	C–OH bonds in oligosaccharides such as mannose & galactose
1000-140	Protein amide I absorption
940	Carotenoid
892	C–C, C–O deoxyribose
600-900	CH out–of–plane bending vibrations
472-521	torsion and ring torsion of phenyl

Table-1 summarized the main regions of an infrared spectrum for single, double, or triple or bonds to hydrogen and others [28]. Every distinctive functional group on the chemical compound will absorb radiation in certain frequencies, such as the OH groups absorb strongly at 3,200-3,600/cm, the CO groups absorb strongly at 1,710/cm; and the CH_3 groups absorb strongly at 1,450 and 1,375/cm [28,29]. Therefore, some differences exist at each FT–IR spectra due to their structure properties. However, the fingerprint regions which can be applied to identify a chemical compound due to its uniqueness are 1,450 to 600/cm [29]. At these regions, there was no diversity on the FT–IR spectra of acetaminophen particles before and after $SCCO_2$ antisolvent treatment. It indicated that the acetaminophen particles products obtained by $SCCO_2$ antisolvent treatment are within a similar functional group as acetaminophen particles before CO_2 treatment. It clearly indicated that CO_2 did not impregnate to the acetaminophen particles products or there was no remaining DMF solvent in the acetaminophen particles products. At these conditions, DMF solvent able to dissolve the acetaminophen compound and at the same time be miscible with CO_2 when the low solubility of acetaminophen in CO_2 was very low [20, 30, 31]. As a result, DMF solvent can be removed completely by CO_2 leaving the acetaminophen particles as products. These results are in good agreement with the results obtained by UV–vis spectrophotometry and GC–MS which not shows the peak intensity at around 270 nm and the peak chromatogram correspond to the existence of DMF compound.

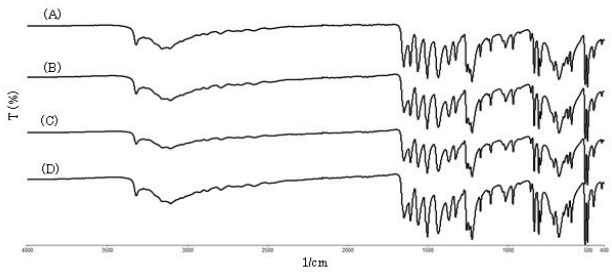

Figure-5. FT-IR spectrum of acetaminophen particles before (A) and after treatment by $SCCO_2$ antisolvent at pressure of 10 MPa and temperature of 40 ^oC with CO_2 flow rates (B) 5, (C) 10, and (D) 15 ml/min, respectively.

Effect of CO_2 flow rate

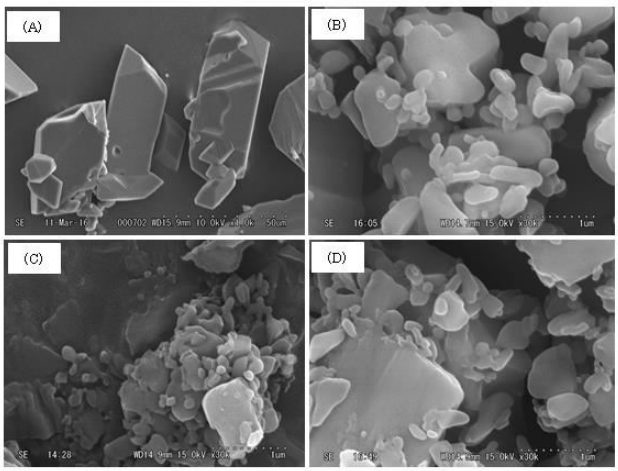

Figure-6. SEM images of (A) acetaminophen particles before and after treatment by $SCCO_2$ antisolvent at pressure of 10 MPa and temperature of 40 ^oC with CO_2 flow rate (B) 5, (C) 10, and (D) 15 ml/min, respectively.

Figure-6 shows SEM images of acetaminophen particles before and after treatment by $SCCO_2$ antisolvent at various CO_2 flow rates. Apparently, the morphology of acetaminophen particles products was almost similar and agglomeration was found during formation of acetaminophen particles at each condition. It indicated that the formation of acetaminophen particles from acetaminophen solution by using $SCCO_2$ antisolvent at various CO_2 flow rates is essentially the same process. The process consisted of mixing of solution and antisolvent, generation of supersaturation, nucleation, and growth by coagulation and condensation, followed by agglomeration [4]. Generally, the increasing flow rate in supercritical antisolvent processes may result several effects. An increase in CO_2 flow rate at a constant feed solution flow rate will decrease the size of particle products. This phenomenon was probably due to the increase in the Reynolds number affected by the CO_2 flow rate, thus turbulence increased which resulted in a better mixing between the solvents turbulence, and thus may promote to the precipitation process [32].

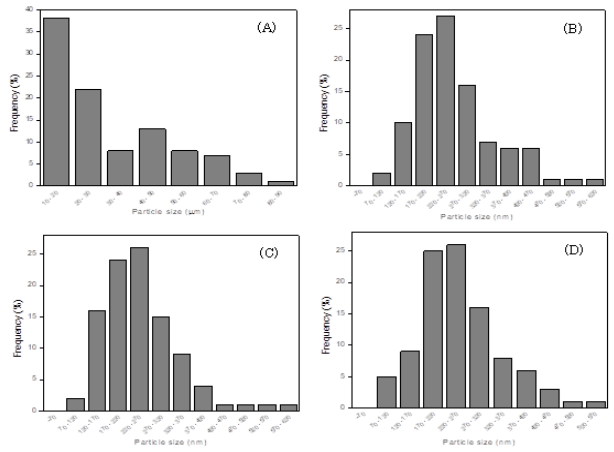

Figure-7. The particle size distribution of (A) acetaminophen particles before and after treatment by $SCCO_2$ antisolvent at pressure of 10 MPa and temperature of 40 ^oC with CO_2 flow rate (B) 5, (C) 10, and (D) 15 ml/min, respectively.

In this work, using the Image J 1.42 tool, from each image which shown in Figure-6, at least 250 different acetaminophen particles products were randomly selected, and their size were measured to generate the particle size distribution. Due to the limitation of the analytical equipment, the dynamic light scattering (DLS) analysis was not performed to characterize the acetaminophen particles products. As shown in Figure-7, although the change of CO_2 flow rate did not seem to give a high effect on the size of acetaminophen particles products, it can be seen that the higher CO_2 flow rate resulted in the narrow particle size distribution. At 5 ml/min CO_2 flow rate, the acetaminophen particles products were generated with size ranges of 70 - 620 nm. It seems that the acetaminophen particles in 170-370 nm diameters were formed dominantly. Conversely, the narrower particle size distribution was obtained when the $SCCO_2$ antisolvent was conducted at the same condition with 15 ml/min CO_2 flow rate. The acetaminophen particles size ranges were 70-570 nm and the size of particles in 170 – 320 nm diameters were produced dominantly. It seemed that the higher CO_2 flow rates might promote the mass transfer rates between CO_2 with organic solvent out of the droplets and enhanced the system turbulence thus increased the supersaturation ratio and resulted in small particles. At higher CO_2 flow rate, the composition of the bulk fluid is decreased by CO_2 flow rate which affects CO_2 dissolving in DMF as a solvent solution. As a result, the solubility of the acetaminophen in DMF will be reduced and smaller particles might be produced [12, 33, 34]. Imsanguan *et al.* [33] reported that the mean particle size decreased with increasing CO_2 flow rate in all experiments when they performed $SCCO_2$ antisolvent to precipitate andrographolide from Andrographis paniculata extracts. They explained that when CO_2 flow rate is rised, the kinetic energy of mass transfer between CO_2 with solution droplet is also increased leading high mass transfer resulting in small droplets, high super saturation, small particles. Guha *et al.* [34] also informed that at higher CO_2 flow rates, higher super saturation levels for shorter period promote to formation of more particles with a narrower size distribution. Conversely, the fluid phase produced in the $SCCO_2$ antisolvent system loaded larger quantities of the DMF solvent when the CO_2 flow rate decreases. Next, the solubilization and solute precipitation processes may occur slowly to yield the acetaminophen larger particles with broader particle size distribution. In detail, Imsanguan *et al.* [33] explained that the change in CO_2 flow rate not only influences kinetic energy of $SCCO_2$, but also the composition of fluid phase. When the ratio of $SCCO_2$ to organic solution is decreased by decreasing CO_2 flow rate, the fugacity coefficient also decreases promoting to higher solubility and a lower precipitation yield. Next, the particles formation process shifts toward the growth process and therefore larger particles would be produced. By the way, the result is, of course, good news in terms of reducing crystalline acetaminophen drug particles size. Because particles size is one of the critical parameters that determine the dissolution rate of the drug in the biological fluids. Bojnanska *et al.* [35] reported that particle size and particle size distribution have a significant effect on the bioavailability of those drugs that have poor solubility in water. They informed that the particle size distribution of the drug substance seems to be critical quality attributes affecting the dissolution rate of the drug substance released from the final peroral drug formulation. Judging from the results, it could be said that the size of acetaminophen particles has been successfully reduced from microscale to nanoscale by using $SCCO_2$ antisolvent.

Effects of pressure and temperature

It was well known that the advantages of using supercritical fluids, such as CO_2, rather than other solvents are mainly due to their physical and chemical properties being intermediate between those of liquids and gases. As

informed above, under supercritical conditions, the liquid-like density enhances the solvating power of CO_2 compared to the gaseous state, and the gas–like mass transport properties enhances the diffusion rate compared to the liquid state. They can be tuned by changing temperature and/or pressure. Next, this rapid transport property of CO_2 can be utilized as a media to produce particles in nano–micro scale. Figures 8 and 9 show the acetaminophen particles products and their size produced by using $SCCO_2$ antisolvent at various operating pressures and temperatures. Generally, the increasing operating pressure of $SCCO_2$ antisolvent will be followed by the smaller (shorter and thinner) particles products [33, 36, 37]. When the operating pressure is high, it is in favor of nucleation process to create and produce a lot of nucleus. Consequently, the particles products will be obtained with smaller size. As shown in Figure-8, it seems that the similar size of acetaminophen particles products with non-spherical shape morphologies were found at each condition. Their morphologies seem to exhibit as plate-like or needle–like shapes. However, from Figure-9, it could be seen that the particles size with ranges of 220-520 nm were obtained dominantly when the experiment was performed at pressure of 15 MPa and temperature of 35 ^oC with 10 ml/min CO_2 flow rate. In comparison, the particles size with ranges of 220-620 nm were found prominently when the experiment was performed at the same condition with the lower pressure at around 8 and 10 MPa. The higher operating pressures resulted the higher CO_2 density, and it was well known that the density of supercritical fluid had high influence on the mass transfer between organic solvent and supercritical fluid during precipitation process. The higher CO_2 density leads to the stronger the dissolving abilities of $SCCO_2$ to DMF solvent.

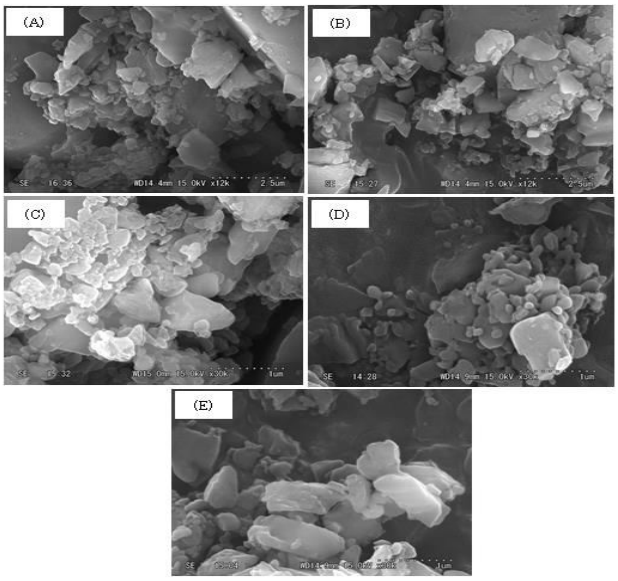

Figure-8. SEM images of acetaminophen particles after treatment by $SCCO_2$ antisolvent at various operating conditions with CO_2 flow rate 10 ml/min: (A) 35 ^oC; 8 MPa, (B) 35 ^oC; 10 MPa, (C) 35 ^oC; 15 MPa, (D) 40 ^oC; 10 MPa, and (E) 50 ^oC; 10 MPa.

Next, the rapid mass transfer between CO_2 and DMF solvent causes high super saturations for the acetaminophen and promotes in the precipitation of acetaminophen smaller particles products. Imsanguan *et al*. [33] reported that the changing the operating pressure is the main reason for changing the $SCCO_2$ density. At higher pressure, the density of CO_2 is higher to result in the difference in density between pure solvent and pure CO_2 decreases promoting to a small maximum droplet radius and short average lifetime of a droplet. These phenomenon leads to high mass transfer rate, high super saturation, fast nucleation and crystal growth rate and small crystal particles. Li *et al*. [37] conducted experiments for recrystallization and size control of puerarin using the $SCCO_2$ antisolvent process. They reported that when operating pressure was high, it was in favor of nucleation, which created a lot of nucleus and thus we obtained crystals with smaller size. On the contrary, when the operating pressure was low, the solvent was not fully expanded, more solvent existed between particles, and the time of crystal growth was prolonged and larger particles were formed. Based on the results, although there is no significant effect of the increasing operating pressure in the particle size and particle size distribution of acetaminophen particles products, it seems that the increasing operating pressure of $SCCO_2$ antisolvent may enhance the nucleation rate and particles growth of acetaminophen to form smaller sized particles [37].

Figures 8 and 9 also show that the increasing operating temperature of $SCCO_2$ antisolvent treatment at the same operating pressure and CO_2 flow rate was followed by increasing the size of acetaminophen particles products. As shown in Figure-9, the operating temperature of 35 ^oC with pressure of 10 MPa and CO_2 flow rate yielded the acetaminophen particles products with size ranges of 120-970 nm with an average particles size of 444 nm. While the acetaminophen particles products with size ranges of 70-620 nm and 120 - 920 nm with average particles sizes were 244 and 457 nm were obtained when the experiments were carried out at temperatures of 40 and 50 ^oC, respectively, with the same operating pressure and CO_2 flow rate. It seemed that the operating temperature may possess two different effects on the size of acetaminophen particles products [20, 33, 34]. It showed that the mean size of acetaminophen particles products at operating temperature of 35 ^oC was smaller than the mean size of acetaminophen particles products at operating temperature of 50 ^oC. At these conditions, the increasing operating temperature leads to the decreasing CO_2 density, as a result, the dissolving ability of CO_2 also decreases. Then super saturation will decrease and the bigger size of acetaminophen particles products will be generated. Li *et al*. [20] informed that the solubility of acetaminophen in the DMF solvent may increase with increasing operating temperature when they performed crystallization of acetaminophen micro-particle by using $SCCO_2$ antisolvent. As a result, the size of acetaminophen particles products will increase due to the super saturation will be attained slower. Li *et al*. [37] confirmed that the

bigger puerarin particles products were produced due to the super saturation is decreased when the higher operating temperature was applied on the recrystallization puerarin under $SCCO_2$ conditions. Interestingly, the mean size of acetaminophen particles products at operating temperature of 40 ^oC was smaller than the mean size of acetaminophen particles products at operating temperature of 35 ^oC. It could be explained that the increasing operating temperature resulted the decreasing of the $SCCO_2$ density. Next, the diffusivity of the $SCCO_2$ may increase and the solute solubility in $SCCO_2$ will decrease so that the supersaturation will be achieved faster. Consequently, the acetaminophen particles products size will decrease. He *et al*. [38] investigated the effect of operating temperature on the $SCCO_2$ antisolvent to micronize natural carotene. The experiments were conducted at operating temperatures of 35-50 ^oC and operating pressures of 8-20 MPa. They reported that the increasing operating temperatures had no significant effect on the size of carotene particles products. Moreover, they obtained the carotene particles products with smaller size with increasing operating temperatures. In detail Imsanguan *et al.* [33] explained that there are two competition phenomena in the particle formation as the temperature changes under $SCCO_2$ conditions. First, a decline in $SCCO_2$ density leads to the $SCCO_2$ diffusivity to give resulting in high mass transfer rates and decreasing particle product size. Second, an increase in the solute solubility will slow the super saturation attainment and an increase in the particle product size. In addition, He *et al*. [38] also informed that the effect of operating temperature depends on the employed solvent, the type of solute, and other factors. Judging from the results, it could be said that the acetaminophen particle product size seemed increase with increasing operating temperature.

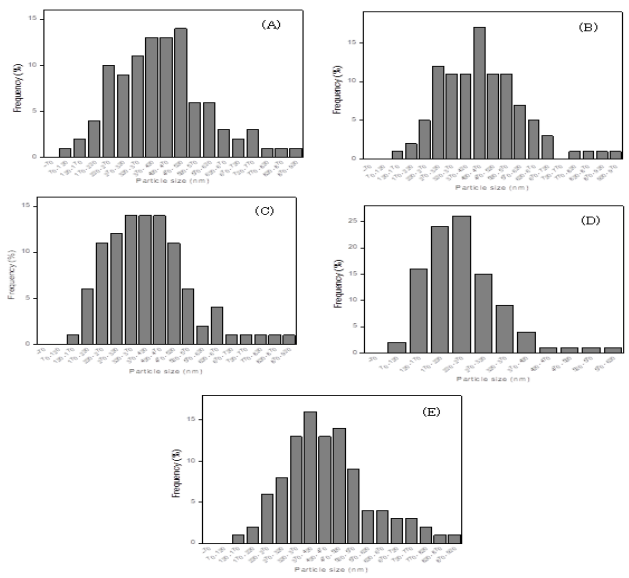

Figure-9. The particle size distribution of acetaminophen particles after treatment by $SCCO_2$ antisolvent at various operating conditions with CO_2 flow rate 10 ml/min: (A) 35 ^oC; 8 MPa, (B) 35 ^oC; 10 MPa, (C) 35 ^oC; 15 MPa, (D) 40 ^oC; 10 MPa, and (E) 50 ^oC; 10 MPa.

CONCLUSIONS

The production of acetaminophen particles via supercritical antisolvent process with CO_2 as an antisolvent has been demonstrated. The experiments were performed at temperatures of 35 – 50 ^oC and pressures of 8 -15 MPa with 5-15 ml/min CO_2 flow rates. The acetaminophen powder which dissolved in DMF was used as a starting material. The spectra of UV–vis and GC–MS analysis showed that there was no remaining DMF solvent in the acetaminophen particles products. It indicated that CO_2 has successfully removed DMF from acetaminophen particles products. The surface characterization by using FT–IR showed that the CO_2 solvent did not impregnate to the acetaminophen particles products. The SEM images showed that the acetaminophen particles products were successfully produced in non-spherical shape morphologies with size less than 1 μm. Finally, it could be said that this process seems a powerful method and to be an apt for size reduction of acetaminophen powder.

REFERENCES

[1] Reverchon E, De Marco I, Adami R, Caputo G. 2008. Expanded micro-particles by supercritical antisolvent precipitation: Interpretation of results. J Supercrit Fluids. 44: 98-108.

[2] Nunes AVM, Duarte CMM. 2011. Dense CO_2 as a Solute, Co–Solute or Co–Solvent in Particle Formation Processes: A Review. Materials. 4: 2017-2041.

[3] Sosnik A, Seremeta KP. 2015. Advantages and challenges of the spray-drying technology for the production of pure drug particles and drug-loaded polymeric carriers. Adv Colloid Interface Sci. 223: 40-54.

[4] Thorat AA, Dalvi SV. 2012. Liquid antisolvent precipitation and stabilization of nanoparticles of poorly water soluble drugs in aqueous suspensions: Recent developments and future perspective. Chem Eng J. 181-182: 1-34.

[5] Truong NP, Whittaker MR, Mak CW, Davis TP. 2015. The importance of nanoparticle shape in cancer drug delivery. Expert Opin Drug Deliv. 12: 129-142.

[6] Sen Gupta A. 2016. Role of particle size, shape, and stiffness in design of intravascular drug delivery systems: insights from computations, experiments, and nature. Wiley Interdiscip Rev Nanomed Nanobiotechnol. 8: 255-270.

[7] Reverchon E, Donsi G, Gorgoglione D. 1993. Salicylic Acid Solubilization in Supercritical CO_2 and Its Micronization by RESS. J Supercrit Fluids. 6: 241-248.

[8] Machmudah S, Zakaria, Sugeng W, Sasaki M, Goto M, Kusumoto N, Hayakawa K. 2012. Lycopene extractions from tomato peel by-product containing tomato seed using supercritical carbon dioxide. J Food Eng. 108: 290-296.

[9] Nerome H, Machmudah S, Wahyudiono, Fukuzato R, Higashiura T, Youn YS, Lee YW, Goto M. 2013. Nanoparticle formation of lycopene/⌴-cyclodextrin inclusion complex using supercritical antisolvent precipitation. J Supercrit Fluids. 83: 97-103.

[10] Goto M, Kanda H, Wahyudiono, Machmudah S. 2015. Extraction of carotenoids and lipids from algae by supercritical CO_2 and subcritical dimethyl ether. J Supercrit Fluids. 96: 245-251.

[11] Sanchez-Camargo ADP, Parada-Alfonso F, Ibanez E, Cifuentes A. 2017. On–line coupling of supercritical fluid extraction and chromatographic techniques. J Sep Sci. 40: 213-227.

[12] Baldyga J, Kubicki D, Shekunov BY, Smith KB. 2010. Mixing effects on particle formation in supercritical fluids. Chem Eng Res Des. 88: 1131-1141.

[13] Kawasaki S. Sue K, Ookawara R, Wakashima Y, Suzuki A. 2010. Development of novel micro swirl mixer for producing fine metal oxide nanoparticles by continuous supercritical hydrothermal method. J Oleo Sci. 59: 557-562.

[14] Chhouk K., Wahyudiono, Kanda H, Kawasaki S, Goto M. 2017. Micronization of curcumin with biodegradable polymer by supercritical anti–solvent using micro swirl mixer. Front Chem Sci Eng. https://doi.org/10.1007/s11705-017-1678-3.

[15] Chiam E, Weinberg L, Bellomo R. 2015. Paracetamol: a review with specific focus on the haemodynamic effects of intravenous administration. Heart, Lung and Vessels. 7: 121-132.

[16] Waring RH, Steventon GB, Mitchell SC. 2007. Molecules of Death. 2nd ed. London WC2H 9HE: Imperial College Press.

[17] Granberg RA, Rasmuson AC. 1999. Solubility of paracetamol in pure solvents. J Chem Eng Data. 44: 1391-1395.

[18] Pires LR, Guarino V, Oliveira MJ, Ribeiro CC, Barbosa MA, Ambrosio L, Pego AP. 2016. Ibuprofen-loaded poly(trimethylene carbonate-co-⌴-caprolactone) electrospun fibres for nerve regeneration. J Tissue Eng Regen Med. 10: E154-E166.

[19] Gardella L, Colonna S, Fina A, Monticelli O. 2016. A Novel Electrostimulated Drug Delivery System Based on PLLA Composites Exploiting the Multiple Functions of Graphite Nanoplatelets. ACS Appl Mater Interfaces. 8: 24909-24917.

[20] Li G, Chu J, Song ES, Row KH, Lee KH, Lee YW. 2006. Crystallization of Acetaminophen Microparticle Using Supercritical Carbon Dioxide. Korean J Chem Eng. 23: 482-487.

[21] Fusaro F, Mazzotti M, Muhrer G. 2004. Gas antisolvent recrystallization of paracetamol from acetone using compressed carbon dioxide as antisolvent. Cryst Growth Des. 4: 881-889.

[22] Rossmann M, Braeuer A, Leipertz A. 2013. Manipulating the size, the morphology and the polymorphism of acetaminophen using supercritical antisolvent (SAS) precipitation. J Supercrit Fluids. 82: 230-237.

[23] Chong GH, Spotar SY, Yunus R. 2009. Numerical Modeling of Mass Transfer for Solvent-Carbon Dioxide System at Supercritical (Miscible) Conditions. J Appl Sci. 9: 3055-3061.

[24] Biazar E, Beitollahi A, Rezayat SM, Forati T, Asefnejad A, Rahimi M, Zeinali R, Ardeshir M, Hatamjafari F, Sahebalzamani A, Heidari M. 2009. Effect of the mechanical activation on size reduction of crystalline acetaminophen drug particles. Int. J Nanomedicine. 4: 283-287.

[25] Miller R, Liggieri L. 2011. Bubble and Drop Interfaces. Leiden: Koninklijke Brill NV.

[26] Mullertz A, Perrie Y, Rades T. 2016. Analytical Techniques in the Pharmaceutical Sciences. New York: Springer Science+Business Media LLC.

[27] Kim MS, Song HS, Park HJ, Hwang SJ. 2012. Effect of solvent type on the nanoparticle formation of atorvastatin calcium by the supercritical antisolvent process. Chem Pharm Bull (Tokyo). 60: 543-547.

[28] Movasaghi Z, Rehman S, ur Rehman DI. 2008. Fourier transform infrared (FTIR) spectroscopy of biological tissues. Appl Spectrosc Rev. 43: 134-179.

[29] Dash A, Somnath S, Tolman J. 2014. Pharmaceutics: Basic Principles and Application to Pharmacy Practice. Academic Press. San Diego: Elsevier Inc.

[30] Bristow S, Shekunov BY, York P. 2001. Solubility analysis of drug compounds in supercritical carbon dioxide using static and dynamic extraction systems. Ind Eng Chem Res. 40: 1732-1739.

[31] Jodecke M, Kamps APS, Maurer G. 2012. An experimental investigation on the influence of NaCl on the solubility of CO_2 in (N, N-dimethylmethanamide + water). Fluid Phase Equilib. 334: 106-116.

[32] Sui X, Wei W, Yang L, Zu Y, Zhao C, Zhang L, Yang F, Zhang Z. 2012. Preparation, characterization and in vivo assessment of the bioavailability of glycyrrhizic acid microparticles by supercritical anti–solvent process. Int. J Pharm. 423: 471-479.

[33] Imsanguan P, Pongamphai S, Douglas S, Teppaitoon W, Douglas PL. 2010. Supercritical antisolvent precipitation of andrographolide from Andrographis paniculata extracts: effect of pressure, temperature and CO_2 flow rate. Powder Technol. 200: 246-253.

[34] Guha R, Vinjamur M, Mukhopadhyay M. 2010. Demonstration of mechanisms for coprecipitation and encapsulation by supercritical antisolvent process. Ind Eng Chem Res. 50: 1079-1088.

[35] Bojnanska E, Kalina M, Parizek L, Bartonickova E, Opravil T, Vesely M, Pekar M, Jampilek J. 2014. Determination of critical parameters of drug substance influencing dissolution: a case study. BioMed Res Int. 2014. doi:http://dx.doi.org/ 10.1155/2014/929248.

[36] Cardoso MAT, Monteiro GA, Cardoso JP, Prazeres TJV, Figueiredo JMF, Martinho JMG, Cabral JMS, Palavra AMF. 2008. Supercritical antisolvent micronization of minocycline hydrochloride. J Supercrit Fluids. 44: 238-244.

[37] Li Y, Yu Y, Wang H, Zhao F. 2016. Effect of process parameters on the recrystallization and size control of puerarin using the supercritical fluid antisolvent process. Asian J Pharm Sci. 11: 281-291.

[38] He WZ, Suo QL, Hong HL, Li GM, Zhao XH, Li CP. 2006. Supercritical antisolvent micronization of natural carotene by the SEDS process through prefilming atomization. Ind Eng Chem Res. 45: 2108-2115.