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Preparation and Characterization of Solid Dispersions of Itraconazole by using Aerosol Solvent Extraction System for Improvement in Drug Solubility and Bioavailability

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The objective of this study was to elucidate the feasibility to improve the solubility and bioavailability of poorly water-soluble itraconazole *via* solid dispersions by using supercritical fluid (SCF). Solid dispersions of itraconazole with hydrophilic polymer, HPMC 2910, were prepared by the aerosol solvent extraction system (ASES) under different process conditions of temperature/pressure. The particle size of solid dispersions ranged from 100 to 500 nm. The equilibrium solubility increased with decrease (15 to 10 MPa) in pressure and increase (40 to 60°C) in temperature. The solid dispersions prepared at 45°C/15 MPa showed a slight increase in equilibrium solubility (approximately 27-fold increase) when compared to pure itraconazole, while those prepared at 60°C/10 MPa showed approximately 610-fold increase and no endothermic peaks corresponding to pure itraconazole were observed, indicating that itraconazole might be molecularly dispersed in HPMC 2910 in the amorphous form. The amorphous state of itraconazole was confirmed by DSC/XRD data. The pharmacokinetic parameters of the ASES-processed solid dispersions, such as T_{max}, C_{max}, and $AUC_{0\text{-}24h}$ were almost similar to Sporanox® capsule which shows high bioavailability. Hence, it was concluded that the ASES process could be a promising technique to reduce particle size and/or prepare amorphous solid dispersion of drugs in order to improve the solubility and bioavailability of poorly water-soluble drugs.

Key words: Itraconazole, Supercritical fluid, Aerosol solvent extraction system (ASES), Solubility, Solid dispersion, Poorly water-soluble drug

INTRODUCTION

Itraconazole can be administered for oral, parenteral and topical uses. However, it is administered orally because of its safety aspects and wide distribution in the body (De Beule and Van Gestel, 2001; Willems *et al.*, 2001). Although itraconazole has an excellent antifungal activity, its oral bioavailability is poor because it has a low solubility of less than 1 mg/mL in water (Woo, 2000). It is known that solubility and dissolution rate of poorly water-soluble drugs are rate-limiting factors for their oral absorption and bioavailability. Many technological methods to improve the dissolution characteristics of slightly water-soluble drugs have been reported in literature (Leuner and Dressman, 2000), such as micronization, formation of solvates, adsorbates, complexes, microspheres, or more often, solid dispersions. In the case of itraconazole, a number of studies have been taken to improve its solubility, such as forming complexes with hydroxypropyl-β-cyclodextrin (Miyake *et al.*, 1999; Stevens, 1999), solid dispersions by spray drying method (Jung *et al.*, 1999; Yoo *et al.*, 2000), preparation of pellets by fluidized-bed coating with water-soluble polymers (Paul *et al.*, 1994), or melt extrusion with water-soluble polymers (Colette *et al.*, 1997) and use of 1-methyl-pyrrolidone as a solubilizing agent (Uch *et al.*, 1999). In spite of the efforts taken for enhancing the solubility and bioavailability of itraconazole, the issue still remains a challenge.

In recent years, processing of pharmaceuticals with supercritical fluids (SCFs), especially with supercritical carbon dioxide (SC-CO_2), has received increased attention.

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SCFs have been successfully used for preparing pharmaceuticals, proteins, polymers/biopolymers, and inorganic materials in micron sized or submicron sized particles (Palakodaty and York, 1997). SCF technology has been investigated to prepare very fine powders or microspheres of many different products (explosives, polymers, pharmaceuticals, peptides, and so on) with several processes, called as 'rapid expansion of supercritical solution' (RESS, Tom and Debenedetti, 1991; Young *et al.*, 2000) and 'supercritical anti-solvent' (SAS), related processes such as SAS (Yeo *et al.*, 1994; Elvassore *et al.*, 2001), 'gas anti-solvent' (GAS) process (Moneghini *et al.*, 2001; Randolph *et al.*, 1993), 'aerosol solvent extraction system' (ASES, Bleich and Muller, 1996; Engwicht *et al.*, 1999), 'solution enhanced dispersion by supercritical fluid' (SEDS, Palakodaty *et al.*, 1998; Ghaderi *et al.*, 1999) process and so on.

In GAS or SAS process, the chosen organic solution is expanded to several fold by mixing with a dense gas in a precipitation vessel. The expansion and extraction of organic solvent into the compressed gas result in lowering solvent power of organic solvent for drug and polymer, leading to super-saturation and precipitation of drugs (solutes) in micro-and nano size. ASES process involves jet break-up of the drug/polymer solution as fine droplets into compressed carbon dioxide through an atomization nozzle (Jung and Perrut, 2001; Bustami *et al.*, 2003).

The preparation of solid dispersions often leads to the conversion of a crystalline drug into a higher energy state, e.g. the amorphous or metastable state. It is known that polymorphic transformation of the drug into amorphous or partially amorphous form of high energy state or metastable state results in higher solubility and faster dissolution of drug (Miyake *et al.*, 1999). In addition, the crystallinity and polymorphism of drug can also be controlled by changing processing parameters during the preparation of solid dispersions. The polymorph of sulfathiazole was controlled by changing temperature and pressure in SC-CO_2 during GAS and SEDS processes (Kitamura *et al.*, 1997; Kordikowski *et al.*, 2001). Hence, it is expected that the particle size/morphology and crystallinity of solid dispersion containing itraconazole could be controlled by changing temperature/pressure during ASES process, leading to improve solubility and dissolution rate.

In the present work, ASES technique has been modified to spray drug/polymer solution with carbon dioxide through a nozzle into SC-CO_2. The itraconazole solid dispersions with hydrophilic polymer were prepared to improve drug solubility and bioavailability by ASES. The prepared solid dispersions were characterized by SEM, DSC, and XRD, and the equilibrium solubility was determined in order to investigate the effect of the ASES process parameters such as temperature/pressure on the solubility, dissolution, particle size, and morphology of the solid dispersions. Finally, the pharmacokinetic values of the prepared solid dispersions were compared to a marketed product, Sporanox® capsule (Jassen Korea), which is well-known to have relatively high bioavailability (55%) after oral administration.

MATERIALS AND METHODS

Materials

Itraconazole was purchased from Recordati Co. (Spain), and hydroxypropylmethylcellulose (HPMC 2910, Pharmacoat 606) was purchased from Shinetsu Chemical Co., Ltd. (Japan). Carbon dioxide (high purity of 99.99%) was purchased from Myungsin General Gas Co., Ltd. (Korea) and ethanol was purchased from Carlo Erba (France). Acetonitrile and methanol (HPLC grade) were purchased from Tedia Co., Inc. (U.S.A.). Dichloromethane, *n*-heptane and isoamylalcohol were purchased from Daejung Chemical Co. (Korea). Triethylamine was purchased from Sigma Chemical Co. (U.S.A.). Other agents were of special reagent grade.

Preparation of solid dispersions of itraconazole with HPMC 2910

A schematic diagram of the ASES apparatus that was used in the study is presented in Fig. 1. The drug/polymer solution was prepared by dissolving itraconazole and HPMC 2910 in a solvent mixture of dichloromethane and ethanol (60:40 w/w %). Firstly, particle precipitation vessel (V = 1907.55 cm^3, I.D. = 9 cm, H = 30 cm) made up of stainless steel was pressurized by delivering carbon dioxide (CO_2) up to the desired pressure (above 7.38 MPa) by using a syringe pump (ISCO Model 260D) and then heated up to the desired temperature (above 31.06°C). When the desired pressure and temperature were reached, the drug/polymer solution was fed into the particle

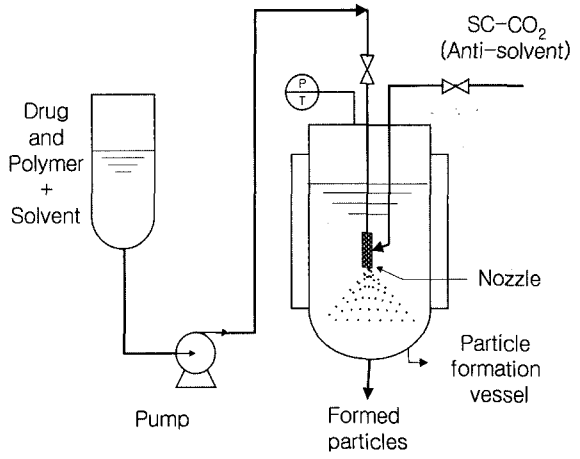

Fig. 1 Schematic diagram of the ASES apparatus

precipitation vessel by HPLC liquid pump (Nihon Seimitsu Kagaku Co. Ltd, Japan). The temperature and pressure of the precipitation vessel were 45, 60°C, and 8, 10, 12, 15 MPa, respectively. The temperature of precipitation vessel was maintained to be constant by circulating water in the water jacket throughout the experiments. The previously prepared drug/polymer solution was sprayed with CO_2 into the precipitation vessel through a gas-energizing nozzle. The flow rates of CO_2 and drug/polymer solution were controlled to be closer to 10 mL/min and about 0.3 mL/min, respectively. When the drug/polymer solution was sprayed through the nozzle, the organic solvent was rapidly extracted into SC-CO_2 leading to precipitation of very fine particles. The mixture of organic solvent and SC-CO_2, was drained out of the precipitation vessel by the back-pressure regulator (Tescom, model 26-1723-24-194). An additional CO_2 was supplied to the precipitation vessel at the same rate to eliminate residual solvents after spraying drug/polymer solution. In washing step, the flow rate and the amount of CO_2 were 20 mL/min and approximately 2,000 mL, respectively to remove residual solvents, and then the precipitation vessel was slowly depressurized to atmospheric pressure. Finally, the particles deposited on the wall and bottom of the precipitation vessel were collected.

Determination of drug content

Exact amount of particles were dissolved in the solvent mixture of dichloromethane and methanol (1:9 v/v), and then 1 mL of this solution was withdrawn. Nine mL of methanol was added to resultant. After shaking by vortex for 2 minutes, 20 μL of the sample was injected into the HPLC system (Shimadzu LC-10 AD, Kyoto, Japan) with UV-VIS detector (Shimadzu SP-10A). The chromatographic column used was μ-Bondapak C18 (3.9×150 mm, 5 μm particle size, Waters, U.S.A.) and the mobile phase was 0.05 M phosphate buffer (pH 7.0) - acetonitrile (40:60, v/v). The system was operated at ambient temperature with a flow rate of 1.0 mL/min and detection wavelength was set at 263 nm.

Particle size and distribution in solid state

The particle size and distribution were characterized by laser diffraction using Sympatec HELOS system (Sympatec GmbH, Clausthal-Zellerfeld, Germany) and RODOS dry powder disperser (at 0.5 and 4.0 bar). Lenses of 100 mm (for the drug) and 200 mm (for the carrier fractions) were employed and calculations were based on the Fraunhofer theory. From the logarithm-probability plot, geometric mean diameter (d_{geo}) was obtained by reading the size corresponding to 50% value, and geometric standard deviation (σ_{geo}) was obtained from the relation:

$$\sigma_{geo} = \frac{84.13\%\ \text{size}}{50\%\ \text{size}} = \frac{50\%\ \text{size}}{15.87\%\ \text{size}}$$

Scanning electron microscopy (SEM)

The shape and size of pure itraconazole and ASES-processed solid dispersions were observed by scanning electron microscopy (SEM, Leo 1455VP, installed in the Korea Basic Scientific Institute). Samples were sputter-coated with Au/Pd by using a vacuum evaporator and examined through a scanning electron microscope at 20 kV accelerating voltage (5000X).

Differential scanning calorimetry (DSC)

Thermal analysis was performed in a Rheometric Scientific DSC (model DLOS, U.K.). The DSC experiment was calibrated according to the user's guide by using indium. Samples (3-4 mg) were introduced in aluminum pans, sealed and heated at a rate of 10°C/min from 40 to 200°C with a nitrogen purge at 20 mL/min.

Power X-ray diffractometry (XRD)

PXRD patterns of each of the pure itraconazole and all of the solid dispersions with HPMC 2910 were recorded by a X-ray diffractometer (Generator: Rigaku, D/max-IIIC, Goniometer: θ/θ goniometer) with Ni filtered Cu-Kα line as the source of radiation, operated at 40 kV voltage and the current of 45 mA. For quantitative studies, the angular range 10-70° 2θ was scanned with a step size of 0.01° at scanning speed of 3°/min. The 2θ values and the intensities of the peaks were compared for both pure itraconazole and ASES processed solid dispersions.

Equilibrium solubility determination

The equilibrium solubility of prepared solid dispersions in pH 1.2 simulated gastric juice without pepsin was determined at 37±0.5°C. An excess amount of the particles (equivalent to 6 mg of itraconazole) was added to 6 mL of the simulated gastric juice in test tubes sealed with stoppers. The test tubes were vortexed for 5 minutes and then sonicated for 30 minutes. They were kept in a constant-temperature shaking bath maintained at 37°C for 24 h. A portion of the solution was taken and filtered through a membrane filter (0.45 μm). The solution was then diluted with HPLC mobile phase and the concentration of the drug was determined by the same HPLC method as described for the determination of drug content.

Pharmacokinetic evaluation in rats

Male Sprague-Dawley rats were obtained from Samtaco Co., Ltd. (Korea) and quarantined for 1 week before use. The rats were housed as three per cage and were maintained on a 12 h light/dark cycle at constant temperature (20-27°C) and relative humidity of 55±10%, with access to

water and Purina Laboratory Chow *ad libitum*. All rats (14-15 weeks old, approximately 300 g) were fasted over 48 h with free access to water before drug administration. Three rats were orally administered with ASES-processed solid dispersions as a test and Sporanox® pellets (from Sporanox® capsule) as a reference, respectively, in single dose of 20 mg itraconazole/kg body weight of the rat.

About 0.5 mL of blood samples were withdrawn directly from the tail vein of the rats before (0 h) and at 1, 2, 3, 4, 5.5, 7.5, and 24 h after dosing. The blood samples were centrifuged at 3000 rpm for 10 min to obtain plasma and kept frozen at -20°C until analyzed. To 200 μL of rat plasma, 50 μL of an internal standard solution (500 μg/mL econazole nitrate in methanol) and 200 μL of 1.0 M carbonate buffer (pH 7.8) were added. To this mixture, 7 mL of *n*-heptane-isoamylalcohol (9:1 v/v) was added, vortexed for 5 min and centrifuged at 3,000 rpm for 5 min. The separated organic phase was evaporated by using N_2 gas at 50°C. The residue was reconstituted with 200 μL of mobile phase (65% aqueous acetonitrile solution containing 0.05% triethylamine) and 100 μL was injected into the HPLC column (Inertsil ODS2 (250×4.6 mm, 5 mm), GL Science, Japan). Standard samples were prepared by spiking blank plasma with known amount of itraconazole and were used for the construction of calibration curves. The system was operated at ambient temperature with a flow rate of 1.2 mL/min, and detection wavelength was set at 258 nm.

All animal experiments were performed according to the "Guidelines for the Care and Use of Laboratory Animals," Chungnam National University. Statistical comparisons of the pharmacokinetic parameters and mean plasma concentration were made by using unpaired *t*-test.

Data analysis

Quantification was based on calibration curves constructed by using peak area ratio of itraconazole to internal standard (econazole nitrate) against itraconazole concentrations by using unweighted least squares linear regression. Pharmacokinetic parameters were estimated by using K-BE Test 2002 (KFDA). The differences in the pharmacokinetic parameters from ASES-processed itraconazole solid dispersion and a marketed product, Sporanox®, were evaluated by using the unpaired *t*-test. In *t*-test, a probability value of $P < 0.05$ was considered to be statistically significant.

RESULTS AND DISCUSSION

Preparation of solid dispersion

In the present study, SC-CO_2 was used as an anti-solvent to prepare itraconazole solid dispersion nanoparticles with hydrophilic polymer, HPMC 2910. It was expected that preparation of solid dispersions of water-insoluble drug with water-soluble hydrophilic polymer would lead to an increase in surface area due to reduction in particle size and decrease in crystallinity, which would be highly effective in enhancing the drug solubility. In preliminary experiments with the different ratios of drug/polymer, itraconazole solid dispersion particles could hardly be collected when drug/polymer ratio was 50: 50 w/w %, due to low yields (approximately below 15%); whereas solid dispersions could be collected with high yields at 60:40 w/w % of drug/polymer ratio. Because itraconazole and HPMC are practically insoluble in SC-CO_2, the ASES process was chosen to prepare solid dispersions of itraconazole.

In preliminary experiments, the solid dispersions of itraconazole were prepared by employing the hydrophilic carriers like HPMC 2910, polyvinylpyrrolidone (PVP) K-30, polyethylene glycol (PEG) 6000 and PEG 20000. Amongst these hydrophilic polymers, HPMC 2910 was chosen for further work because solid dispersion with HPMC 2910 showed the better solubility than other hydrophilic polymers for itraconazole. HPMCs are mixed ethers of cellulose, in which 16.5~30% of the hydroxyl groups are methylated and 4~32% are derivatized with hydroxypropyl groups. Type 2910 has an average methoxy content of 29% and a hydroxypropyl content of 10%. The molecular weight of the HPMCs ranges from about 10,000 to 1,500,000 and they are soluble in water and solvent mixtures of ethanol/methanol and dichloromethane (Harwood, 2000). Several studies with some poorly soluble drugs showed that the release rate and the bioavailability of drug could be improved through preparation of a solid dispersions in the presence of HPMC (Suzuki and Sunada, 1998; Kohri *et al.*, 1999; Okimoto *et al.*, 1997). Further works such as drug-excipient compatability are required to elucidate why HPMC is more effective than other hydrophilic polymers such as PVP or PEG for improvement in itraconazole solubility.

Itraconazole is freely soluble in dichloromethane, and HPMC is soluble in solvent mixtures of ethanol and dichloromethane. Dichloromethane and ethanol are highly miscible, and the viscosity of HPMC in this solvent mixture is known to be the lowest at the dichloromethane/ethanol ratio of 60:40 w/w % (Shin-Etsu Catalogue). Too high viscosity and concentration of drug/polymer solution could interfere with free jet-break up of drug/polymer solution into the precipitation vessel due to nozzle clogging. On the contrary, too low concentration can result in low yield of product and increase in time and amount of CO_2 needed to eliminate the whole solvent mixture. When the drug/polymer solution is sprayed through the nozzle, the organic solvents are rapidly extracted into SC-CO_2 resulting in precipitation of very fine particles. In this step,

gas-energizing nozzle as an atomizing nozzle is very effective in producing very fine sub-micron sized particles.

Particle size and distribution

To investigate the effect of pressure on particle size and distribution, we performed the experiments in the range of 8~15 MPa and at 45 and 60°C. The other factors such as the concentration, flow rate and composition of the drug/polymer solution were kept constant. As presented in Fig. 2B-2G, the size of ASES-processed solid dispersions varied between 100-500 nm. As can be seen in Fig. 2B-2G, particle size seems to decrease with an increase in pressure. However, the geometric mean diameter (d_{geo}) increased with an increase in pressure as shown in Fig. 3 (d_{geo} = 0.20, 0.25, and 0.38 μm for 10, 12, and 15 MPa of pressure, respectively). Judging from the SEM images in Fig. 2B-2G, large platy particles were observed with an increase in pressure, resulting in the increase in the geometric mean diameter. Platy particles were hardly observed in solid dispersion particles, prepared at low pressure of 10 MPa (Fig. 2B and 2E). It is postulated that the particle size would decrease with an increase in precipitation rate of solid dispersion components in the SCFs. The rate of precipitation would increase by two possibilities: one is the extract-out of organic solvent mixture into SC-CO_2; and another is decrease in solubility of solid dispersion components in SC-CO_2. As the pressure increases at a constant temperature, the

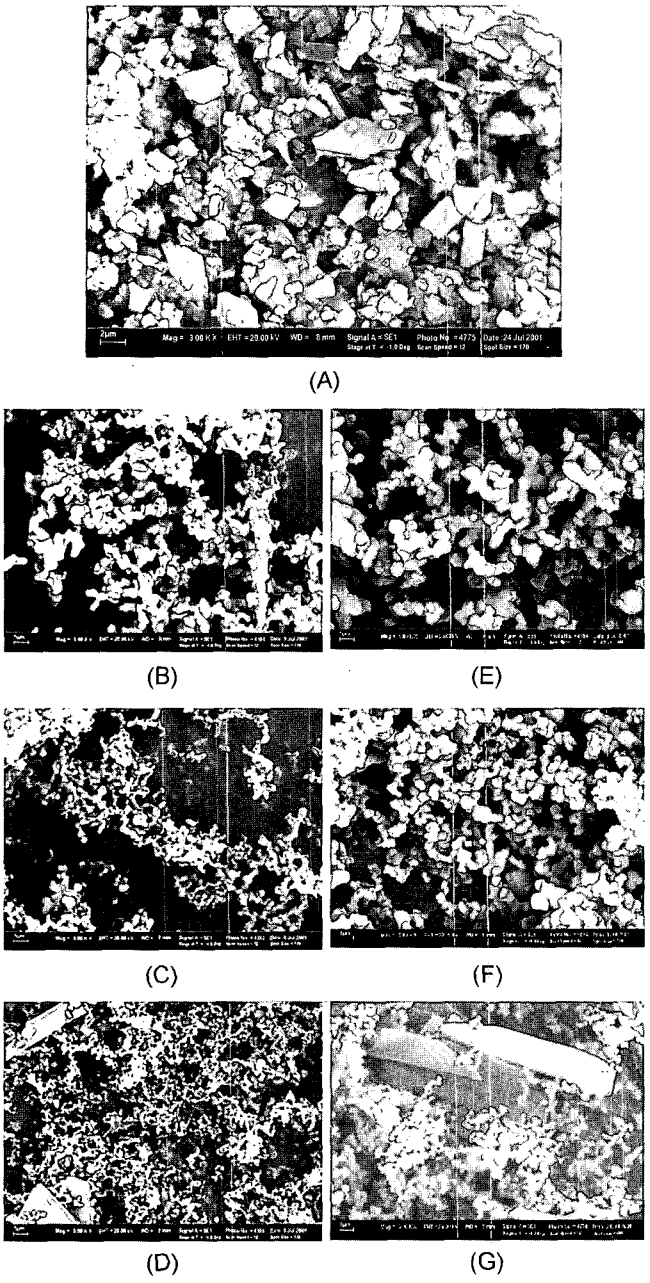

Fig. 2. SEM images of (A) Pure itraconazole (3,000×), ASES-processed solid dispersions with HPMC 2910; (B), (E) 10 MPa, (C), (F) 12 MPa, and (D), (G) 15 MPa (5,000×) at 45°C, 60°C respectively.

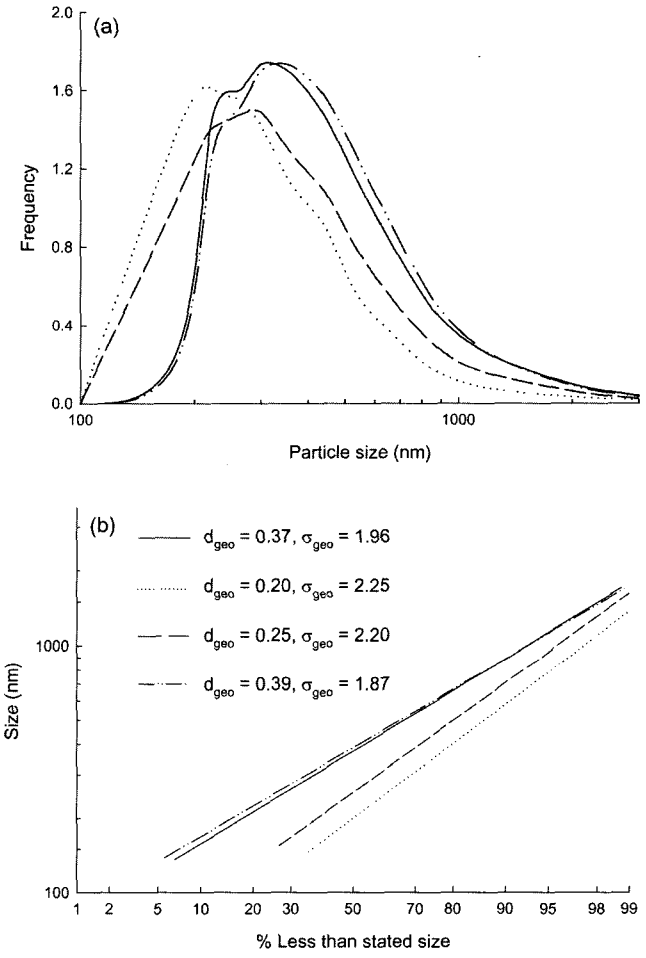

Fig. 3. Particle size distribution ((a) logarithm size-frequency distribution, (b) logarithm-probability plot) of itraconazole microparticles before and after ASES processing: —— pure itraconazole; solid dispersions at 60°C/10 MPa (·····), 60°C/12 MPa (— —), and 60°C/15 MPa (— • —).

solubility of solute components will increase resulting in the delay of precipitation rate, while the density of SC-CO_2 will increase resulting in the acceleration of precipitation rate. The density of SC-CO_2 increases about 2.5 times by increasing pressure from 8 to 15 MPa (Marr and Gamse, 2000; Martin *et al.*, 2002), and high density SC-CO_2 has a higher solvating power for organic solvents such as dichloromethane and ethanol as well as solid dispersion components. As a result, high density SC-CO_2 can rapidly extract the organic solvents and in the similar manner will also prevent the solute components from precipitating in SC-CO_2 due to their increased solubility in SC-CO_2. Consequently, super-saturation and rapid precipitation of drug can be affected by the solubility change of both organic solvents and solute components.

In comparison with particle size of solid dispersion prepared at 45 and 60°C, the solid dispersions processed at 45°C were little smaller than those processed at 60°C. It is also postulated that the smaller size of the particles could be due to the fact that the density of SC-CO_2 at 45°C is significantly higher than at 60°C (Marr and Gamse, 2003). The amount of density change of SC-CO_2 by the pressure change is smaller at 60°C than at 45°C (Marr and Gamse, 2003).

Particle morphology

To investigate the effect of the process parameters such as temperature and pressure on particle morphology, several experiments were performed in the range of 8~15 MPa at 45 and 60°C, respectively. Most of the solid dispersions were spherical in shape or somewhat aggregated. However, under the low pressure of 8 MPa, the volumetric expansion of the organic solvent was not enough to produce spherical particles. It seemed that these operating conditions could not provide a sufficient density change of SC-CO_2 for rapid precipitation of solute components.

From the SEM image (Fig. 2A), it is clear that the pure itraconazole was comprised of large irregular crystals with broad size distribution. On the contrary, ASES-processed solid dispersions prepared at 10 MPa, as presented in Fig. 2B and 2E, show a relatively regular and spherical shape with sub-micron size and narrow size distribution, but there seems to form a little bridge among the particles. However, platy particles were increasingly observed and particle morphology became heterogeneous with an increase in pressure (Fig. 2B-2D and 2E-2G). It is thought that the platy particles may have an increase in crystallinity when compared to the spherical particles, as confirmed by DSC, XRD, and solubility data. Therefore, it is clear that the morphology and particle size of drug, itraconazole, was substantially modified by the ASES process.

Thermal characteristic study from DSC

The representative DSC curves of pure itraconazole and ASES-processed solid dispersions are presented in Fig. 4. An endothermic peak of pure itraconazole was exhibited at about 168.6°C with enthalpy (ΔH) of 398 J/g, corresponding to its melting point. The endothermic peak and enthalpy (ΔH) of the prepared solid dispersions are listed in Table I. All ASES-processed solid dispersions showed a melting endothermic peak with much lower heat-flow than that of pure itraconazole and the peak was shifted from 168 to 163°C. However, the endothermic peak of solid dispersions prepared at 60°C/10 MPa was not at all observed. This indicates that itraconazole might be in the amorphous state in HPMC 2910 matrix during ASES process, which was further confirmed by Powder XRD analysis (Fig. 5). Moreover, the physical mixture of itraconazole/HPMC 2910 showed an endothermic peak corresponding to the melting point of pure itraconazole, indicating the presence of crystalline form. The solid state of drug within solid dispersions can be explained by following possibilities: itraconazole was present as very small crystals dispersed within the HPMC matrix, or it was

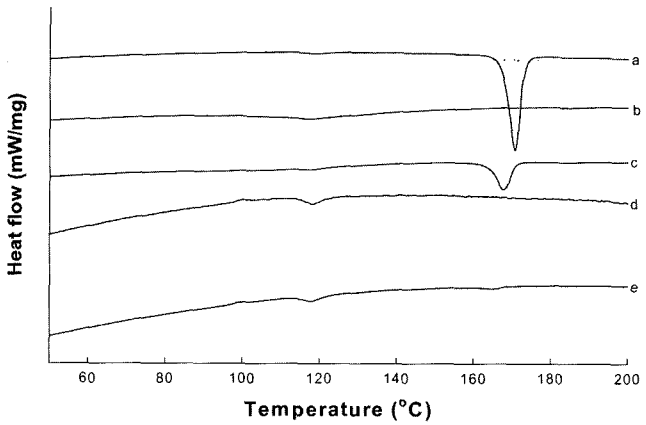

Fig. 4. Representative DSC curves of (a): pure itraconazole, (b): pure HPMC 2910, (c): physical mixture (3:2), (d), (e): solid dispersions with HPMC 2910 at 60°C/10 MPa and 60°C/12 MPa.

Table I. Thermal characteristics of pure itraconazole and ASES-processed solid dispersion with HPMC 2910 at different conditions

Processing conditions		Endothermic peak	
Temperature (°C)	Pressure (MPa)	Melting point (°C)	Enthalpy (J/g)
45	10	164.6	15.0
	12	162.2	34.8
	15	163.9	40.7
60	10	not observed	not observed
	12	163.4	30.5
	15	165.1	50.1
Physical mixture		167.8	134
Pure itraconazole		168.6	398

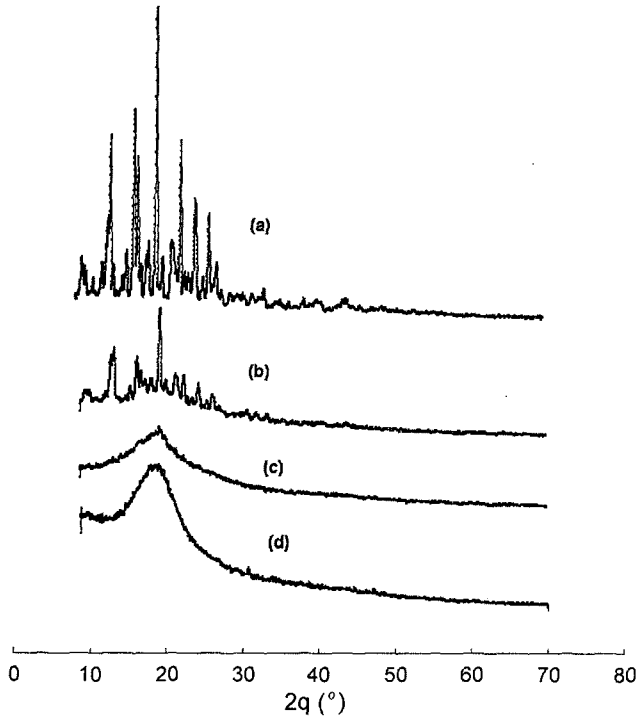

Fig. 5. X-ray diffraction patterns of (a) Pure itraconazole, (b), and (c) ASES-processed solid dispersions with HPMC 2910 at 45°C/15 MPa and 60°C/10 MPa, respectively, and (d) Pure HPMC 2910.

present as amorphous domains dispersed within the HPMC matrix, or it was present as a solid solution within the HPMC matrix. A remarkable reduction in the intensity of itraconazole endothermic peak suggests decrease in its crystallinity, which was affected by the process temperature and pressure. In the solid dispersions prepared at 60°C, itraconazole was present as amorphous domains dispersed within the HPMC matrix.

The polymorph of sulfathiazole was reported to be controlled by changing the temperature and pressure in SC-CO_2 during GAS and SEDS processes (Kitamura *et al.*, 1997; Kordikowski *et al.*, 2001). Therefore, it can be explained that the crystallization rate of drug might be controlled by changing the temperature and pressure due to the density change of SC-CO_2, resulting in decrease in crystallinity from crystalline to amorphous form.

Powder X-ray diffraction

The solid state of pure itraconazole, physical mixture and solid dispersions with HPMC 2910 were characterized by XRD (Fig. 5). The powder diffraction patterns (pdp) of pure itraconazole showed characteristic of high-intensity diffraction peaks at 2θ values of 12.05, 15.65, 19.05, and 20.35°, which corresponded to the known powder diffraction patterns of pure itraconazole. When solid dispersions prepared at 60°C/10 MPa were compared to those prepared at 45°C/15 MPa, it was interesting to note that the pdp of the dispersions prepared at 60°C/10 MPa did not exhibited any peaks corresponding to pure itraconazole. This indicates that itraconazole was present in the amorphous form (Fig. 5). While the solid dispersion prepared at 45°C/15 MPa still showed diffraction peaks corresponding to pure itraconazole, indicating the presence of crystallinity.

The formulation with pure itraconazole/HPMC 2910 prepared at 60°C/10 MPa was present in the amorphous form. This was in line with our findings from DSC thermal analysis. This result was also identical with the DSC thermogram of the sample and indicates that the amorphous state of itraconazole was formed at 60°C/10 MPa.

Equilibrium solubility

The equilibrium solubility data are presented in Table II. The results show that ASES-processed solid dispersions have much more enhanced equilibrium solubility when compared to pure itraconazole (27-610 folds). The equilibrium solubility of drug was increased with decrease (from 15 MPa to 10 MPa) in pressure and an increase (from 45°C to 60°C) in temperature. The results seemed to be consistent with those from the geometric mean diameter, the morphology, DSC, and XRD data of the particles. In general, it has been widely known that particle size and morphology such as polymorphism or amorphous form can affect the solubility and dissolution rate. In addition, many of the carriers used for solid dispersions may have some wetting properties and hence, it is reasonable to suggest that improved wetting may lead to reduced agglomeration and hence, increased surface area.

In the present study, the equilibrium solubility of ASES-processed solid dispersions increased with decrease in pressure and an increase in temperature, as presented in Table II. Particularly, the ASES solid dispersions prepared at 60°C/10 MPa had the highest solubility (122.4 ± 1.7 μg/mL). It is worth noting that the equilibrium solubility of solid dispersions might mainly be increased by decrease in crystallinity or conversion from crystalline to amorphous

Table II. Equilibrium solubility data of ASES-processed solid dispersions and pure itraconazole (n = 3)

Samples	Temperature (°C)	Pressure (MPa)	Solubility (μg/mL)
ASES-processed solid dispersions	45	10	81.5±8.5
		12	11.6±0.4
		15	5.5±1.2
	60	10	122.4±1.7
		12	16.1±0.7
		15	5.2±0.3
Pure itraconazole	-	-	0.2±0.04

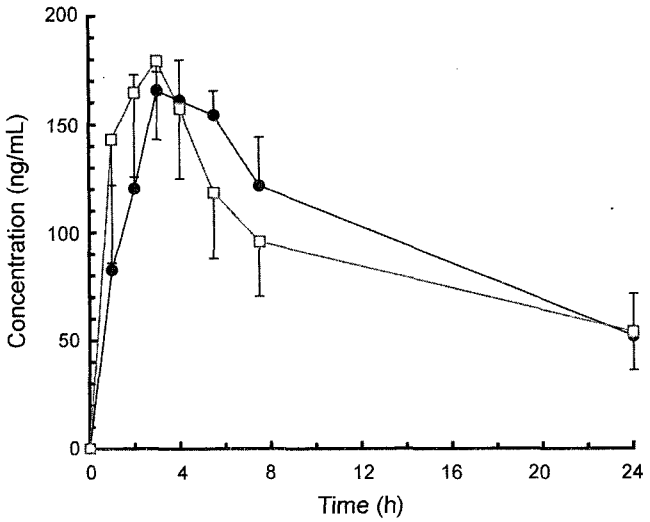

Fig. 6. Mean plasma concentration of itraconazole versus time profiles following administration of a single dose of 20 mg itraconazole/kg body weight of the rat (n = 3). Data is shown as mean±S.D., ASES-processed solid dispersions (test ●), and a marketed product, Sporanox® capsule (reference ■).

forms within the sub-micron size range (Table I, Fig. 4 and 5). Hence, it can be concluded that ASES-processed formulations show enhanced equilibrium solubility. It may be possible to obtain the optimal solubility enhancement by altering the processing parameters.

The evaluation of pharmacokinetics in rats

Fig. 6 shows the mean plasma concentration-time curves following oral administration of a marketed product, Sporanox® and ASES-processed particles, and Table III shows the pharmacokinetic parameters including $AUC_{0\text{-}24h}$, C_{max} and T_{max}. No statistically significant difference was found between test and reference ($P > 0.05$).

In summary, itraconazole solid dispersions were formed successfully at 45, 60°C and 10~15 MPa. ASES-processed solid dispersions had a relatively regular and spherical shape with a diameter range of 100-500 nm. The equilibrium solubility of ASES-processed solid dispersions at 60°C/10 MPa was dramatically increased, and it was supported from the observations like, absence of endothermic peak of itraconazole from solid dispersions, no platy particle in SEM and no crystalline diffraction peak in XRD data.

It was concluded that the solid dispersions prepared by ASES process might be a feasible technique for improving the solubility and bioavailability of itraconazole.

Table III. The pharmacokinetic parameters of ASES-processed solid dispersions and the marketed product, Sporanox® capsule after oral administration of 20 mg itraconazole/kg body weight to the rats (n = 3)

Subjects	ASES-processed solid dispersions			Commercial formulation, Sporanox capsule		
	C_{max} (ng/mL)	T_{max} (h)	$AUC_{0\text{-}24h}$ (ng·h/mL)	C_{max} (ng/mL)	T_{max} (h)	$AUC_{0\text{-}24h}$ (ng·h/mL)
1	179.6	2.0	2422	215.0	3.0	2755
2	179.7	4.0	2613	179.9	3.0	2241
3	161.1	4.0	1867	142.7	3.0	1562
Mean	173.5	3.3	2301	179.2	3.0	2186
Std	10.7	1.2	387.3	36.2	0.0	598.5

No significant differences were observed between two groups, $P > 0.05$

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Erratum

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Synthesis of 3-Alkylthio-6-Allylthiopyridazine Derivatives and Their Antihepatocarcinoma Activity

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In Fig. 2A, * (statistical significance) must be inserted in K6 as shown.