A New Method for the Production of Crystalline Microparticles

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The production of crystalline-non aggregated microparticles with a narrow particle size distribution is a major challenge for the inhalation drug delivery formulation. The objective of this study was to develop a method for converting an amorphous and micronised drug produced by spray drying to crystalline form to enhance the stability and promote its inhalation performance. The feasibility of using supercritical carbon dioxide ($scCO_2$) as a conditioning agent for salbutamol sulphate that is an anti-asthmatic drug was assessed. It was found that conditioning the spray dried powder of salbutamol sulphate with neat CO_2 was not efficient for converting the amorphous form to crystalline. However, salbutamol sulphate crystals were formed after 12 hours conditioning with CO_2 modified with menthol at 150 bar and 50°C. CO_2 had negligible effect on particle size of the powder, but the aerosol performance of the drug was substantially increased after conditioning. The benign technique developed can be used for the bulk production of inhalable crystalline powders at moderate temperatures without using any organic solvent. This will be attractive for pharmaceutical processing by eliminating amorphous fractions of the powders produced by various micronisation methods.

INTRODUCTION

It has been reported that 75% of the products used in the pharmaceutical industry are in powder form [1]. Fundamental research is required to be undertaken to control the size and crystallinity of particle generation processes. Spray drying, grinding, jet milling and various forms of liquid-liquid anti-solvent precipitation are currently used for particle formation. The drawbacks of these methods include high temperatures, consumption of large amounts of organic solvents and their residues, and appearance of amorphous fraction [2, 3].

Salbutamol sulphate is a β_2–sympathomimetic for the treatment of asthmatic patients, which in powder form is broadly used for inhalation and oral formulations [4-6]. The micronised-crystalline salbutamol sulphate is currently produced by crystallisation and precipitation followed by grinding and milling [7]. Dry powder of crystalline salbutamol sulphate was produced by liquid-liquid antisolvent using high gravity packed bed followed by spray drying [3, 8]. The aerosol performance of salbutamol sulphate was remarkably enhanced. However, large amounts of organic solvent were used in this process. Amorphous form of micronsied salbutamol sulphate was produced by spray drying of aqueous solution with a poor aerosol performance [9]. The amorphous powders are not thermally stable, agglomerate easily and convert to crystalline form with a larger particle size, therefore, decrease the long term shelf of the formulation [10].

Amorphous drug is transformed to the thermodynamically stable crystalline state at ambient conditions when the glass transition temperature (T_g) is decreased to below this temperature [11]. Most of commercially available dry powder inhalation (DPI) formulations are in the form of stable crystalline forms to avoid any phase transition [12]. There are several attempts to convert amorphous drugs into crystalline form by exposing the drug to elevated humidity [13] and by co-spray drying with additives such as PEG [14]. These methods did not promote the stability and aerosol performance of the powder. Recently it was found amorphous fluticasone 17α-2 fumarate could be converted to crystalline form by conditioning spray dried powder with menthol vapour [15]. Our preliminary results demonstrated that it was not feasible to implement this method for salbutamol sulphate even after 24 hours conditioning.

Supercritical fluids have been used for the production of crystalline particles of various drugs using techniques such as rapid expansion of supercritical solution, gas antisolvent, particles from gas saturated solution [12]. However, these processes have not yet been commercially used for micronisation of pharmaceuticals [12]. Solution enhanced dispersion using supercritical carbon dioxide was used for crystallisation of salbutamol sulphate. The flake-like crystalline particles of salbutamol sulphate with average particle size of 7µm were formed from methanol solution using CO_2 as an antisolvent [16]. The powder was not suitable for inhalation drug delivery formulation.

The primary objective of this study was to produce a stable powder with a greater aerosol performance compared to the powder produced by spray drying. We investigated the conversion of amorphous form of salbutamol sulphate which was produced from spray drying of aqueous solution to crystalline form by conditioning the powder with supercritical CO_2. The effect of process variables on the degree of crystallinity and particle size was determined.

MATERIALS AND METHODS

Materials

Salbutamol sulphate (99.9% purity) was purchased from Inter-Chemical Ltd. China. Food grade carbon dioxide with 99.99% purity was purchased from British Oxygen Company (BOC) and menthol (99.9% purity) was supplied by Sigma-Aldrich. MilliQ water was used to dissolve salbutamol sulphate for the spray drying process and analysis.

Methods

The preparation of amorphous salbutamol sulphate

Amorphous salbutamol sulphate was produced using a spray drying. In each run a 10 wt% aqueous solution of salbutamol sulphate was sprayed via a nozzle with an internal diameter of 2.5 mm into a Buchi 290 mini spray drier (Buchi Laboratory-Techniques, Switzerland). The air inlet and outlet temperature were adjusted at 150°C and 103°C, respectively. A 2.4 mL/min solution was delivered into the drying chamber using a peristaltic pump and the air flow rate was adjusted using a regulator at 10 L/min. The aspiration rate and the pressure were kept at 38 mL/min and 4.14 bar, respectively. Finally, dried powder was collected from the spray drier collection pot, kept in a sealed container, and stored in a dissector at room temperature for the analysis and next stage of the process.

Powder Conditioning by the Supercritical CO_2

The schematic diagram of the experimental set up used for the conditioning of the amorphous powders by $scCO_2$ is shown in Figure 1. In each run 2.0 g of a desired solute was loaded into a custom made-stirred- high pressure- vessel (45 mL). The required amount of menthol was loaded on the top part of the vessel using a porous container. Care must be taken to avoid direct contact between the menthol and SS powder prior to dissolving menthol in CO_2. The vessel was then placed in a controlled temperature water bath. The temperature and stirrer speed were tuned using a hot plate (MSH-30D Wise Stir, Daihan Scientific Co., Ltd., Seoul, Korea). A high pressure pump (P50 Series, Thar Technologies, USA) was used for the delivery of CO_2 at high pressure to the system. Cooling water was recirculated through a jacket around the head of high pressure pump using a magnetic pump (MD-10-230GS01, Iwaki Co., Ltd., Tokyo, Japan) to condense CO_2 vapour into liquid form for pressurisation. The CO_2 was fed at a rate of 10 mL/min until the desired pressure was achieved. The system was then isolated, maintained at that desired pressure for a period of time. In this period powder was exposed to CO_2 saturated menthol phase. The stirrer was then stopped and the system was purged from menthol by passing neat CO_2 at the operating pressure and temperature for 1.5 hours at a constant flow rate of 10 mL/min. Finally, the system was depressurised; the powder was collected, placed in a closed sealed container and kept at room temperature in desiccators for future characterisations. The apparatus was also used for conditioning the amorphous powder with neat CO_2 and menthol. In the former condition no menthol was loaded into the vessel and the latter system was not pressurised by CO_2.

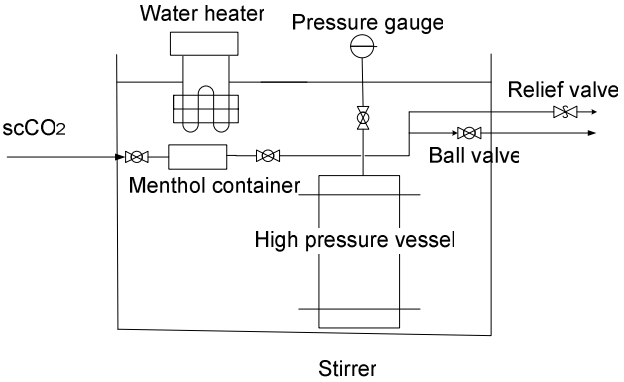


Fig. 1: Schematic diagram of the set up for conditioning $scCO_2$ modified with menthol.

Solid-state characterisation

Particle morphology

A Philips 505 Scanning Electron Microscope (Phillips, Holland) operating at 10 kV and spot size 20 was employed to determine particle morphology. Samples were coated with ~ 20nm of gold using a modified Edwards E306A coater.

Particle size distribution

Particle size distribution was measured using laser diffraction (Mastersizer S, Malvern Instrument, UK) as a dry powder. 10-30 mg of powder was charged into the dry powder dispenser and the particle size distribution was determined at 4 bar. The measurements were carried out in triplicate. Particle size analysis was based on the refractive index (RI) of salbutamol sulphate (1.553) and $RI_{imaginary}$ of salbutamol sulphate (0.100). The size distributions were expressed by the cumulative volume diameter at 10, 50 and 90%.

Particle crystallinity

Powder crystallinity was determined by X-ray diffraction (XRD, D5000, Siemens, Germany) operated at room temperature using Cu Kα radiation at 30 mA and 40 kV, with an angular increment of 0.05^{o}/s count time of 2 s. A Universal Analysis 2000 modulated differential scanning calorimetry (DSC, TA Instruments, USA) was also used to analyse the crystallinity and melting points of powders. The samples (3-7 mg) were loaded into closed aluminium pans and subjected to heat at a rate of 10^{o}C/min to a maximum temperature of 250^{o}C under a nitrogen purge.

Aerosol performance by Multi-Stage Liquid Impinger (MSLI) analysis

The methods of aerosol performance analysis are based on the Chiou et.al. technique [3]. The aerosol performance of SS powders were assessed using an Aeroliser® (Novartis Pharmaceuticals, Australia) coupled through an USP stainless steel throat to a Multi-Stage Liquid Impinger (MSLI) (Copley, United Kingdom), operating at 100 L/min. The powder (20.0±0.5 mg) was filled into hydroxypropyl methylcellulose capsule (size 3, Capsugel®, USA) and three capsules were used in each experiment. Each experiment was performed in duplicate. Particles deposited at different locations in MSLI were assayed by UV spectrophotometry (U-2000, Hitachi, Japan) at 276 nm. A calibration curve of salbutamol sulphate in water was prepared prior to conducting the aerosol performance test at the concentration range of 0.4-100 µg/mL. Aerosol performance explains how the particles were delivered deep into the lung.

RESULTS AND DISCUSSION

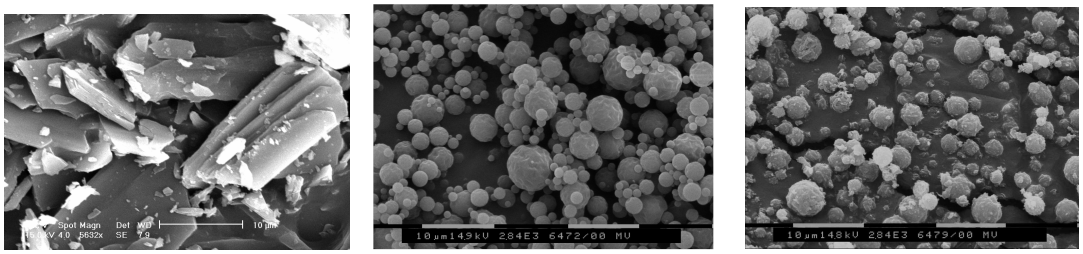
In this study we assessed the feasibility of using CO_2 modified with menthol (accepted by FDA for oral use), which has a high solubility in CO_2 (e.g. 0.68×10^{3} mole fraction at 75 bar and 40 °C)[17] for conditioning of amorphous salbutamol sulphate. The results of our previous study demonstrated that conditioning of amorphous salbutamol sulphate by menthol at 50 °C and atmospheric pressure over 24 hours and neat CO_2 at 50°C, 150 bar for 12 hours were not successful in converting the amorphous form of the drug into crystalline powder +/- stirring.

As shown in Figure 1, untreated salbutamol sulphate consists of irregular particles greater than 20 µm, not suitable for dry powder inhalation (DPI) formulation. Amorphous salbutamol sulphate was formed by spray drying with an average particle size of 6.0 µm. The surface of the microspheres was slightly corrugated during the spray drying process. Salbutamol sulphate that was conditioned by CO_2 + menthol for 12 hours at 150 bar and 50 °C using 1:1 weight ratio of menthol to salbutamol sulphate, comprised of microspheres covered on the surface by nano-scale needle-shaped crystals. The presence of this irregularity on the surface resulted in decreasing the average particle size and enhancing the aerosol performance of the powder from 32.5% (spray dried product) to 43.0% (conditioning with $scCO_2$ + menthol). Our hypothesis is as the powder was dispersed homogenously in the stirred vessel, microspheres in the vicinity of each other were integrated during the crystallisation process and formed nano-scale needle shape crystals. The glass transition temperature of salbutamol sulphate may also be depressed by the addition of CO_2 and menthol into the solid phase, leading to crystallisation.

The particle size distributions were measured for various samples of salbutamol sulphate. As shown in Table 1, average particle size of the powder processed by spray drying and conditioning by CO_2 +/- menthol were 3.1 µm and 2.6 µm, respectively.

However, there is a remarkable difference in their aerosol performance, which is attributed to the degree of crystallinity, surface morphology and agglomeration.

The results of DSC and XRD analysis confirmed that the degree of crystallinity of the salbutamol sulphate was similar to that of the raw material. The XRD spectrums of spray dried salbutamol sulphate in Figure 2(d) demonstrate the powder was in amorphous form. However, after it was conditioned by modified CO_2 the crystal structure was similar to the raw material, validating that the polymorphic form of the drug did not change after conditioning by CO_2. In the XRD profile, no peaks associated with menthol were presented in the processed powder, indicating that it was completely removed from the product.



(a) (b) (c)

Figure 1: SEM images of salbutamol sulphate (a) untreated powder. (b) spray drying (c) after conditioning with $scCO_2$ + menthol at 50^{o}C and 150 bar for 12 hrs

Table 1: Characteristics of particle size distribution of salbutamol sulphate.

Technique	**particle size distribution (µm)**		
	D(v,10%)	**D(v, 50%)**	**D(v,90%)**
Spray dried	1.102	3.050	6.205
Exposed to menthol	1.010	3.039	6.183
Exposed to CO_2	0.875	3.054	6.586
Exposed to menthol + $scCO_2$	1.058	2.558	5.176

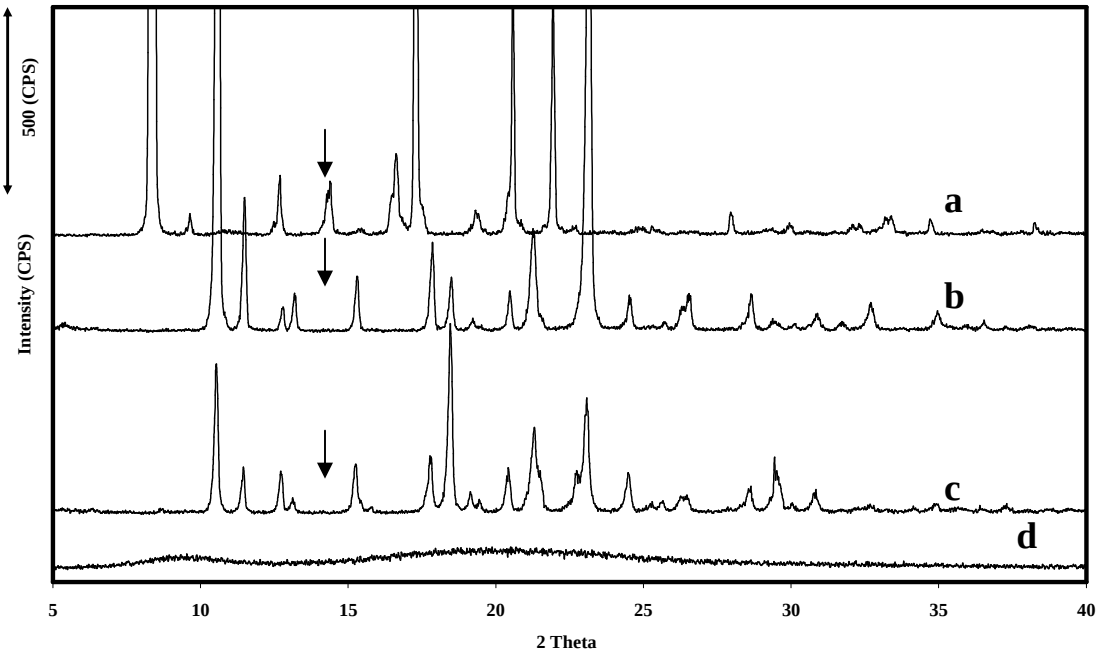


Fig. 2: XRD patterns of: (a) menthol (b) untreated salbutamol sulphate, (c) treated with $scCO_2$ + menthol at 50^{o}C and 150bar for 12 hrs. (d) spray dried of salbutamol sulphate.

CONCLUSION

This study demonstrates the potential of $scCO_2$ modified with menthol for converting amorphous form of powders to crystalline, while preserving the particle size. This new approach has great potential in pharmaceutical powder processing for various formulations where crystalline structure is critical. The process can be used to remove residue of amorphous fraction produced in a product by various techniques to enhance their stability and shelf life. The aerosol performance of salbutamol sulphate conditioned by modified CO_2 was increased, hence decreasing the dosage required for the treatment of asthma, reducing the side effects and promoting patient compliance. No organic solvent was used in the method developed for conditioning the amorphous powder. Menthol used in the process was completely removed from the product and could be recycled in a large scale production. Conditioning by modified CO_2 is a solvent free process that operates at moderate temperature and is attractive for processing fragile and heat sensitive compounds.

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