Co-precipitation of Curcumin-PVP via the Supercritical Antisolvent Process

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ABSTRACT
Curcumin is a hydrophobic compound with antimicrobial, anti-inflammatory, anticancer and antioxidant properties. However, the use of curcumin in drug formulations is limited by its poor water solubility and low bioavailability. A common strategy to improve the dissolution rate and possibly the bioavailability of hydrophobic drugs is to coprecipitate them with hydrophilic polymers. In this work, polyvinylpyrrolidone (PVP) was used to produce curcumin-PVP sub-microspheres via the Supercritical Antisolvent Process (SAS). The effect of solvent, solution concentration, solution flow rate and drug/polymer ratio was investigated. The mean particle size obtained from DMSO was between 0.308-0.324 µm. From ethanol, smaller particles with 0.269 µm mean diameter were produced. It was found that the organic solvent used has the most significant effect on particle size.

Keywords: coprecipitation, curcumin, PVP, supercritical antisolvent.

INTRODUCTION
Curcumin is a natural bioactive compound traditionally used as a spice and food colorant/additive. Over the last decades it has been proved that curcumin has numerous therapeutic properties such as anticancer, antimicrobial, antioxidant and anti-inflammatory [1]. However, its poor solubility in aqueous media leads to low oral bioavailability, which prevents the application of curcumin in drug formulations. Moreover, curcumin suffers spontaneous degradation under heat, light, and physiologic pH [2]. Recently, several curcumin formulations have been developed to address these issues, including nanoparticles, liposomes, polymeric micelles, dendrimers and hydrogels [3].

The coprecipitation of active pharmaceutical ingredients (API) with hydrophilic polymers is a common approach to protect the API against degradation while improving its dissolution properties and bioavailability. Polyvinylpyrrolidone (PVP) is a biodegradable polymer widely used in pharmaceutical applications since it has the ability to modify the crystallization kinetics of poorly water-soluble compounds and thereby produce amorphous formulations with improved dissolution profile [4]. Solid dispersions of curcumin and PVP with different molecular weights have been prepared before using conventional techniques, such as spray
drying [5] and solvent evaporation [6]. PVP has also been used as a stabilizer for curcumin nanoparticles produced via antisolvent methods followed by freeze drying [7–9]. These methods have some disadvantages such as the use of high temperature, which inhibits the processing of thermo-sensitive compounds and the requirement of an extra processing step to remove residual solvent from the formulation. Moreover, formulations with low loading efficiency and wide particle size distribution are usually produced.

Supercritical fluid-based micronization is advantageous over conventional techniques since the control of particle size can be achieved via the manipulation of the operational conditions and solvent-free formulations are obtained. Additionally, mild conditions of pressure and temperature can be used. Carbon dioxide is usually selected in supercritical fluid-based micronization processes since it is inexpensive, non-flammable, non-toxic and it has a relatively low critical pressure (73.9 bar) and critical temperature (31.1°C). Depending on the role played by the supercritical carbon dioxide (sc-CO2), it can act as solvent, co-solvent or antisolvent in relation to the solute. In a typical Supercritical Antisolvent (SAS) process, the solute is dissolved in an organic solvent and then sprayed into the precipitator through which sc-CO2 runs continuously. The instantaneous diffusion of sc-CO2 into the liquid solution promotes its supersaturation and the precipitation of the solute. The separation of solvent, antisolvent and solid is then obtained by simple depressurization.

In this work curcumin-PVP solid dispersions were produced via SAS using DMSO and ethanol as solvents. The effects of solution concentration, solution flow rate and drug/polymer ratio were studied.

**MATERIALS AND METHODS**

**Materials**

Curcumin (CUR, purity ≥ 90%) and Polyvinylpyrrolidone (PVP, Mw = 10 kg/mol) were purchased from Cayman Chemical and Sigma Aldrich, UK respectively. Dimethyl sulfoxide (DMSO, purity > 99%) was purchased from Fisher Chemical, UK, ethanol (purity = 99.97%) was purchased from VWR Chemicals and carbon dioxide (purity ≥ 99.8%) from BOC, UK. The products were used as received from the suppliers.

**SAS equipment and experimental procedure**

A diagram of the SAS process can be seen in **Figure 1**. Initially, the precipitator is pressurized until the desired pressure is achieved. The micrometric valve is then opened to allow a constant flow of CO2 and the solvent injection starts by turning the HPLC pump on. When a quasi-steady state composition of solvent and CO2 inside the precipitator is achieved, the pump is switched to the solution containing curcumin and PVP. After the desired amount of curcumin and PVP is injected into the precipitator, the pump is switched back to solvent to purge the line and then fresh CO2 is pumped through the system to remove any residual solvent. Finally, the pressure is decreased and the sample that has been precipitated inside the cellulose thimble is collected.

**Scanning electron microscopy (SEM) – Particle size**

Scanning Electron Microscopy (SEM - model Philips XL-30 FEG) was used to examine the morphology of unprocessed materials and coprecipitates. The pictures were taken at 10 kV and 10 mA. Initially, samples were placed on double-sided adhesive carbon tape and sputter coated (Polaron SC 7640) at 25 mA with platinum for 3 min. Particle size was measured via Image J analysis software. At least 200 particles of each sample were analysed and the mean value and standard deviation are reported.
RESULTS

Figure 2 shows the morphology of unprocessed materials. While curcumin has a rod-like shape, PVP particles are collapsed spheres. For both compounds, it is possible to see that particle size distribution is very wide before processing.

Figure 2. a) unprocessed curcumin; b) unprocessed PVP.

The use of SAS process for the coprecipitation of curcumin and PVP is proposed in order to obtain an amorphous formulation with improved dissolution properties. All the experiments were performed at 10 MPa, 40°C, with a CO$_2$ flow rate of 40g/min to make sure the precipitation happens in the supercritical region of the mixture CO$_2$-solvent. DMSO and ethanol were used as solvents. Solution flow rate ($f$), overall solution concentration ($C$) and
drug/polymer mass ratio (R) were varied to analyse the effect on the particle size. Table 1 presents the experimental conditions used in each experiment and the results obtained.

**Table 1.** Experimental conditions and results (f: solution flow rate; C: overall solution concentration; R: drug/polymer ratio; m.d.: mean diameter; s.d.: standard deviation).

<table>
<thead>
<tr>
<th>Exp.</th>
<th>f (ml/min)</th>
<th>C (mg/ml)</th>
<th>R</th>
<th>solvent</th>
<th>m. d. (µm)</th>
<th>s. d. (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>#1</td>
<td>0.3</td>
<td>20</td>
<td>1:3</td>
<td>DMSO</td>
<td>0.402</td>
<td>0.186</td>
</tr>
<tr>
<td>#2</td>
<td>0.3</td>
<td>10</td>
<td>1:3</td>
<td>DMSO</td>
<td>0.324</td>
<td>0.163</td>
</tr>
<tr>
<td>#3</td>
<td>0.3</td>
<td>20</td>
<td>1:6</td>
<td>DMSO</td>
<td>0.395</td>
<td>0.207</td>
</tr>
<tr>
<td>#4</td>
<td>0.6</td>
<td>20</td>
<td>1:3</td>
<td>DMSO</td>
<td>0.408</td>
<td>0.183</td>
</tr>
<tr>
<td>#5</td>
<td>0.3</td>
<td>10</td>
<td>1:3</td>
<td>Ethanol</td>
<td>0.269</td>
<td>0.114</td>
</tr>
</tbody>
</table>

**Figure 3.** Curcumin/PVP coprecipitated from different conditions specified in Table 1.
Effect of solution concentration
The effect of solution concentration was analysed by comparing experiment #1 performed at 20 mg/ml overall solution concentration and experiment #2 carried out at 10 mg/ml. In both experiments DMSO was used as the solvent, solution flow rate was 0.3 ml/min and 1:3 curcumin/PVP ratio. Figure 3 shows that spherical and well separated sub-microparticles were obtained in both experiments. The absence of the typical rod-like curcumin morphology suggests a successful coprecipitation. The decrease in solution concentration led to a decrease in the mean particle size from 0.402 µm to 0.324 µm.

Effect of drug/polymer ratio
Curcumin/PVP mass ratio was decreased from 1:3 in experiment #1 to 1:6 in experiment #3. The overall solution concentration was kept at 20 mg/ml and the DMSO solution flow rate at 0.3 ml/min. By increasing the amount of polymer in the solution, there was a negligible decrease in particle size from 0.402 µm to 0.395 µm. Therefore, in the range of conditions tested, drug/polymer ratio has little impact on particle size. Figure 3 shows that particles kept the spherical and individualized morphology in experiment #3.

Effect of solution flow rate
The effect of solution flow rate was analysed for DMSO solutions at 20 mg/ml concentration and 1:3 drug/polymer ratio. In experiment #4 the flow rate of solution was increased to 0.6 ml/min to be compared with experiment #1 performed at 0.3 ml/min. Particle size increased slightly to 0.408 µm at higher flow rates and particle size distribution became narrower. The morphology obtained is still spherical and non-coalescing (Figure 3).

Effect of solvent
Ethanol was used as solvent in experiment #5 and compared with experiment #2, both performed at 0.3 ml/min solution flow rate, 10 mg/ml solution concentration and 1:3 curcumin/PVP ratio. Particle size decreased significantly to 0.269 µm. As ethanol is less viscous than DMSO, probably the mechanism of solvent drying is faster, leading to the formation of smaller particles. The morphology of particles is reported in Figure 3.

CONCLUSION
Curcumin-PVP formulations were successfully produced and the effects of operational parameters were analysed. Experiments performed with DMSO showed similar particle size even at different concentrations, drug/polymer ratios and solution flow rates. On the other hand, the use of ethanol significantly decreased the particle size of the formulation. This indicates that the type of solvent plays a major role in the coprecipitation of curcumin-PVP via SAS. Further studies with different solvents and solvent mixtures will be conducted to understand if it is possible to tune particle size from the micrometric to the nanometric range. Release tests are going to be carried out to confirm the improvement in the dissolution properties of the coprecipitated formulation compared to raw curcumin.

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REFERENCES


