INTRODUCTION

Design and fabrication of pharmaceutical particulate systems is still largely an art as opposed to a fundamental science. However, a more systematic design and manufacture of particulate systems including nanoparticles is being enabled by the application of novel technologies, such as supercritical fluid (SCF) technology, which is the focus of this chapter (1). A fluid is supercritical when it is compressed beyond its critical pressure \(P_c\) and heated beyond its critical temperature \(T_c\). SCF technology has emerged as an important technique for particle manufacturing. In many industrial applications, it is poised to replace the conventional recrystallization and
milling processes, mainly because of the quality and the purity of the final particles and environmental benefits. There are a variety of SCFs available as listed in Table 1.

<table>
<thead>
<tr>
<th>SCF</th>
<th>$T_c$ (°C)</th>
<th>$P_c$ (bar)</th>
<th>Safety hazard</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethylene</td>
<td>9.3</td>
<td>50.3</td>
<td>Flammable gas</td>
</tr>
<tr>
<td>Trifluoromethane (fluoroform)</td>
<td>25.9</td>
<td>47.5</td>
<td></td>
</tr>
<tr>
<td>Chlorotrifluoromethane</td>
<td>28.9</td>
<td>39.2</td>
<td></td>
</tr>
<tr>
<td>Ethane</td>
<td>32.3</td>
<td>48.8</td>
<td>Flammable gas</td>
</tr>
<tr>
<td>Carbon dioxide</td>
<td>31.1</td>
<td>73.7</td>
<td></td>
</tr>
<tr>
<td>Dinitrogen monoxide (laughing gas)</td>
<td>36.5</td>
<td>72.6</td>
<td>Not combustible but enhances combustion of other substances</td>
</tr>
<tr>
<td>Sulfur hexafluoride</td>
<td>45.5</td>
<td>37.6</td>
<td></td>
</tr>
<tr>
<td>Chlorodifluoromethane (HCFC 22; R 22)</td>
<td>96.4</td>
<td>49.1</td>
<td>Combustible under specific conditions</td>
</tr>
<tr>
<td>Propane</td>
<td>96.8</td>
<td>43.0</td>
<td>Extremely flammable</td>
</tr>
<tr>
<td>Ammonia</td>
<td>132.4</td>
<td>112.7</td>
<td>Flammable and toxic</td>
</tr>
<tr>
<td>Dimethyl ether (wood ether)</td>
<td>126.8</td>
<td>52.4</td>
<td>Extremely flammable</td>
</tr>
<tr>
<td>Trichlorofluoromethane (CFC 11, R 11)</td>
<td>198.0</td>
<td>44.1</td>
<td></td>
</tr>
<tr>
<td>Isopropanol</td>
<td>235.2</td>
<td>47.6</td>
<td>Highly flammable</td>
</tr>
<tr>
<td>Cyclohexane</td>
<td>280.3</td>
<td>40.7</td>
<td>Highly flammable</td>
</tr>
<tr>
<td>Toluene</td>
<td>318.6</td>
<td>41.1</td>
<td>Highly flammable</td>
</tr>
<tr>
<td>Water</td>
<td>374.0</td>
<td>220.5</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviation: SCF, supercritical fluid.

Supercritical CO$_2$

Out of the fluids listed in Table 1, carbon dioxide is the SCF of choice because it is nonflammable, nontoxic, inexpensive, and has mild critical temperature. Hence, much of the attention has been given to supercritical carbon dioxide for pharmaceutical particle formation.

No amount of compression can liquefy the SCF. In fact, pressure can be used to continuously change the density from...
gas-like conditions to liquid-like conditions. Near the critical region, small changes in the pressure can give rise to large changes in the density. Figure 1 shows how density of carbon dioxide is varied by pressure at different temperatures.

In addition to density, diffusivity of the SCFs is higher than that of liquid solvents, and can be easily varied. For typical conditions, diffusivity in SCFs is of the order of $10^{-3}$ cm$^2$/sec as compared to $10^{-1}$ for gases and $10^{-5}$ for liquids. Typical viscosity of SCFs is of the order of $10^{-4}$ g/cm/sec, similar to that of gases, and about 100-fold lower than that of liquids. High diffusivity and low viscosity provide rapid equilibration of the fluid.

**SOLUBILITY IN SUPERCRITICAL CO$_2$**

Carbon dioxide (O=C=O) is a nonpolar molecule with a small polarity due to the quadrupole moment. Hence, nonpolar or light molecules (e.g., menthol, methanol, acetone, toluene, and hexanes) easily dissolve in CO$_2$, whereas the polar or
heavy molecules (e.g., griseofulvin, paclitaxel, tetracycline, and dexamethasone phosphate) have a very poor solubility. For example, solubility of menthol in CO\textsubscript{2} is as high as 5 mol\% (Fig. 2), whereas the solubility of griseofulvin in CO\textsubscript{2} is only about 18 ppm (Fig. 3). Solubilities of other pharmaceutical compounds are shown in Figures 4–6. A comprehensive compilation of solubility data in supercritical CO\textsubscript{2} is given in a recent book by Gupta and Shim (6).

Three important factors that govern drug solubility in supercritical CO\textsubscript{2} are the vapor pressure of drug, drug–CO\textsubscript{2} interaction, and density of CO\textsubscript{2}. Drug vapor pressure is a function of temperature (\(T\)), and CO\textsubscript{2} density is a function of pressure (\(P\)) and \(T\). (Fig. 7). Mendez–Santiago and Teja (8) observed that the solubility (\(y_2\) \(\mu\)mol/mol) can be correlated using the following equation:

\[
y_2 = \frac{10^6}{P} \exp \left( A \frac{B \rho_1}{T} + C \right)
\]

where \(P\) is in bars, \(T\) is in Kelvin, \(\rho_1\) is CO\textsubscript{2} density in moles per milliliter. Constants \(A\), \(B\), and \(C\) are listed in Table 2.

**Figure 2** Solubility of menthol in CO\textsubscript{2}. *Abbreviation: CO\textsubscript{2}, carbon dioxide. Source: Ref. 2.*
**Figure 3** Solubility of griseofulvin in CO₂. *Source:* From Ref. 2.

**Figure 4** Solubility of nicotinic acid in CO₂. *Source:* From Ref. 4.
Figure 5  Solubility of chloramphenicol in CO₂. Source: From Ref. 5.

Figure 6  Solubility of salicylic acid in CO₂. Source: From Ref. 3.
for selected drugs. Density of pure CO₂ can be obtained from NIST Standard Reference Database (http://webbook.nist.gov/chemistry/) at the desired \( T \) and \( P \). Alternatively, the following empirical expression can be used (9):

\[
\rho_1 = \frac{1}{44} \exp \left( -27.091 + 0.609\sqrt{T} + \frac{3966.170}{T} - 3.445P + 0.401\sqrt{P} \right)
\]

Figure 7  Solubility of \( \alpha \)-tocopherol in CO₂ at 333 K. Source: From Ref. 7.

Rapid Expansion of Supercritical Solution for Particle Formation

From the previous section it is evident that the solubility of pharmaceutical compounds is highly dependent on CO₂ pressure. As the pressure is reduced, solubility decreases because of a reduction in the CO₂ density, which is closely related to its solubility power (8–11). At a high pressure, the drug can be dissolved in CO₂ and if the pressure is reduced to ambient, the drug precipitates out as fine particles. The depressurization
Table 2  Values of the Constants for Equation (1)

<table>
<thead>
<tr>
<th>Drug</th>
<th>A</th>
<th>B</th>
<th>C</th>
</tr>
</thead>
<tbody>
<tr>
<td>7-Azaindole</td>
<td>-8.412</td>
<td>87.110</td>
<td>20.66</td>
</tr>
<tr>
<td>Behenic acid</td>
<td>-4.473</td>
<td>61.240</td>
<td>6.80</td>
</tr>
<tr>
<td>Biphenyl</td>
<td>-10.200</td>
<td>132.800</td>
<td>25.75</td>
</tr>
<tr>
<td>Brassylc acid</td>
<td>-10.860</td>
<td>146.100</td>
<td>21.01</td>
</tr>
<tr>
<td>Capsaicin</td>
<td>-7.172</td>
<td>70.830</td>
<td>19.54</td>
</tr>
<tr>
<td>Cholecalciferol</td>
<td>-9.784</td>
<td>172.500</td>
<td>18.42</td>
</tr>
<tr>
<td>Diphenylamine</td>
<td>-18.720</td>
<td>397.100</td>
<td>33.40</td>
</tr>
<tr>
<td>Eicosanoic acid</td>
<td>-15.990</td>
<td>161.600</td>
<td>36.97</td>
</tr>
<tr>
<td>1-Eicosanol</td>
<td>-14.530</td>
<td>122.500</td>
<td>36.15</td>
</tr>
<tr>
<td>Endrin</td>
<td>-9.912</td>
<td>167.800</td>
<td>20.29</td>
</tr>
<tr>
<td>Ergocalciferol</td>
<td>-1.092</td>
<td>173.500</td>
<td>21.51</td>
</tr>
<tr>
<td>Flavone</td>
<td>-11.430</td>
<td>110.100</td>
<td>27.38</td>
</tr>
<tr>
<td>D(-)-Fructose</td>
<td>-871.2</td>
<td>10.740</td>
<td>-4.29</td>
</tr>
<tr>
<td>3-Hydroxyflavone</td>
<td>-9.746</td>
<td>81.530</td>
<td>21.31</td>
</tr>
<tr>
<td>Ketoprofen</td>
<td>-12.090</td>
<td>186.100</td>
<td>24.72</td>
</tr>
<tr>
<td>Medroxyprogesterone acetate</td>
<td>-10.270</td>
<td>186.100</td>
<td>17.77</td>
</tr>
<tr>
<td>Methoxychlor</td>
<td>-12.670</td>
<td>184.100</td>
<td>27.38</td>
</tr>
<tr>
<td>Monocrotaline</td>
<td>-10.440</td>
<td>8.057</td>
<td>20.28</td>
</tr>
<tr>
<td>Mystiric acid</td>
<td>-17.250</td>
<td>173.100</td>
<td>44.84</td>
</tr>
<tr>
<td>Naproxen</td>
<td>-9.723</td>
<td>122.900</td>
<td>18.11</td>
</tr>
<tr>
<td>Narasin</td>
<td>-8.529</td>
<td>124.900</td>
<td>13.86</td>
</tr>
<tr>
<td>Nifedipine</td>
<td>-10.020</td>
<td>168.500</td>
<td>15.92</td>
</tr>
<tr>
<td>Nimesulide</td>
<td>-13.820</td>
<td>186.900</td>
<td>28.14</td>
</tr>
<tr>
<td>Nitrendipine</td>
<td>-9.546</td>
<td>151.400</td>
<td>15.91</td>
</tr>
<tr>
<td>Octacosane</td>
<td>-19.860</td>
<td>123.000</td>
<td>52.55</td>
</tr>
<tr>
<td>1-Octadecanol</td>
<td>-17.290</td>
<td>141.000</td>
<td>45.32</td>
</tr>
<tr>
<td>Palmityl behenate</td>
<td>-8.378</td>
<td>59.180</td>
<td>18.44</td>
</tr>
<tr>
<td>Penicillin V</td>
<td>-6.459</td>
<td>73.730</td>
<td>13.29</td>
</tr>
<tr>
<td>Phenylacetic acid</td>
<td>-13.730</td>
<td>14.450</td>
<td>35.78</td>
</tr>
<tr>
<td>Piroxicam</td>
<td>-10.560</td>
<td>18.130</td>
<td>17.57</td>
</tr>
<tr>
<td>Progesterone</td>
<td>-12.090</td>
<td>21.040</td>
<td>23.43</td>
</tr>
<tr>
<td>t-Retinol</td>
<td>-8.717</td>
<td>168.900</td>
<td>16.60</td>
</tr>
<tr>
<td>Salinomycin</td>
<td>-18.890</td>
<td>185.500</td>
<td>42.05</td>
</tr>
<tr>
<td>Stigmasterol</td>
<td>-13.010</td>
<td>169.000</td>
<td>25.23</td>
</tr>
<tr>
<td>Testosterone</td>
<td>-14.330</td>
<td>238.300</td>
<td>26.42</td>
</tr>
<tr>
<td>Theobromine</td>
<td>-7.443</td>
<td>114.000</td>
<td>8.31</td>
</tr>
<tr>
<td>Theophyline</td>
<td>-6.957</td>
<td>94</td>
<td>760</td>
</tr>
<tr>
<td>Triaccontane</td>
<td>-22.965</td>
<td>199.800</td>
<td>57.22</td>
</tr>
<tr>
<td>Triocetylphosphine oxide</td>
<td>-9.378</td>
<td>211.900</td>
<td>17.65</td>
</tr>
<tr>
<td>Vanillin</td>
<td>-7.334</td>
<td>136.500</td>
<td>14.53</td>
</tr>
</tbody>
</table>

Source: From Ref. 8.
can be done very fast; so fast that CO₂ comes out of the nozzle at the speed of sound. The fast depressurization results in a very fast rate of precipitation providing small drug particles. This process is termed as rapid expansion of supercritical solution (RESS) and has been tested for a wide variety of drugs. A schematic of the RESS process is shown in Figure 8.

The bulk drug is solubilized in CO₂ in a high-pressure chamber. The solution is then passed through a nozzle to rapidly reduce the pressure. In some applications, the nozzle is also heated to avoid clogging due to freezing of CO₂ by sudden expansion. The precipitated drug particles are collected in an ambient pressure bag filter. The morphology of the resulting particles (crystalline or amorphous) depends on the molecular structure of the drug and RESS process conditions (solubilization temperature, expansion temperature, pressure drop across nozzle, nozzle geometry, impact distance of the jet against collection surface, etc.).

Most of the drug particles produced by RESS, have been in the 1–5 µm-size range. The rapid expansion of supercritical CO₂ does produce nuclei 5–10 nm in diameter, but these nuclei grow because of coagulation and condensation to produce the final micrometer-size particle. The micronized drugs include 2–5 µm aspirin, 3–5 µm caffeine, 2–3 µm cholesterol, 2 µm ibuprofen, 1–3 µm nifedipine, 2–5 µm progesterone, 1–5 µm salicylic acid, 2–5 µm testosterone, 4–12 µm theophylline, and 1–2 µm α-tocopherol (3,12–19).

**Figure 8** Schematic of RESS process. **Abbreviation:** RESS, rapid expansion of supercritical solution.
For a few drugs, nanoparticles have also been obtained using RESS. These nanonized drugs include 100 nm lidocaine, 200 nm griseofulvin, 200 nm β-sitosterol (20,21). Recently, by expanding the drug CO₂ mixture in a liquid medium containing stabilizers, Pathak et al. (22) have obtained small nanoparticles of ibuprofen and naproxen.

As the obtained particles are free of organic solvents and the high-pressure part of the equipment is not too expensive, theoretically RESS process is very useful. Unfortunately, for most drugs, nanoparticles are not obtained. Instead, oriented-fused particles are obtained (Fig. 9).

Another major drawback of the RESS process is the low solubility of most drugs in supercritical carbon dioxide. For example, solubility of griseofulin is only 18 ppm. Hence, to obtain 18 mol of griseofulvin, one needs to use one million mol of CO₂ (i.e., 1 g griseofulvin particles from about 7 kg CO₂). The worst part is the collection problem. For the earlier example, 1 g of powder would be dispersed in 3573 L of gaseous CO₂ requiring efficient filtration.

Addition of cosolvents, such as methanol, acetone, or ethanol, can enhance the drug solubility to some extent.

---

**Figure 9** Scanning electron micrograph of griseofulvin particles obtained from RESS process (solubilization in CO₂ was done at 196 bar, 40°C). *Abbreviation:* RESS, rapid expansion of supercritical solution.
But, the presence of such a cosolvent in the expansion chamber is not desired, as it will lead to solubilization of the particles in the cosolvent.

**RESS WITH SOLID COSOLVENT FOR NANOPARTICLE FORMATION**

Recently, Thakur and Gupta (2,23) have addressed both the challenges of RESS (low solubility and growth by coagulation) by utilizing a cosolvent that is solid at the nozzle exit conditions. The solid cosolvent (SC) enhances the solubility in supercritical carbon dioxide and provides a barrier for coagulation in the expansion chamber. The SC is later removed from the solute particles by lyophilization (sublimation). The new process is termed as RESS–SC.

In RESS, all the nuclei or small particles of solute are surrounded by the same kind of particles as in Figure 10(A). But in the RESS–SC process, nuclei or small particles of the solute are surrounded by excess SC particles. This reduces the probability of solute particle growth by coagulation. The

**Figure 10** (A) Magnified view of the RESS nozzle. (B) Schematic of RESS–SC process. Circles represent drug particles, whereas stars represent solid–cosolvent particles. **Abbreviations:** RESS, rapid expansion of supercritical solution; RESS–SC, rapid expansion of supercritical solution solid cosolvent.
RESS–SC concept is depicted in Figure 10(B). The lyophilization step shown in the figure is carried out separately after the expansion.

The choice of a proper SC is the key for successful RESS-SC. Various requirements for the selection of the SC are:

- good solubility in supercritical CO₂,
- solid at nozzle exit condition (5–30°C),
- good vapor pressure for easy removal by sublimation,
- should be nonreactive with drugs or CO₂, and
- inexpensive.

Menthol is a solid compound (melting point, 42°C) that satisfies the requirements mentioned earlier. It has appreciable solubility in CO₂ (Fig. 2) and can easily sublime under vacuum. Menthol naturally occurs in mint-flavored plants, and is widely used in antipruritic agents, mouthwashes, nasal sprays, food, etc. Because of its wide use in food and pharmaceuticals, menthol does not seem to possess harmful effects and its use as a cosolvent with supercritical carbon dioxide still carries the benign benefit of the technology. The following are two examples of the RESS-SC process using menthol solid cosolvent.

Griseofulvin Nanoparticles

Using menthol cosolvent, griseofulvin solubility can be enhanced by up to 28-fold, as shown in Table T3. The nanoparticles obtained from the RESS–SC process are in the size range of 50–250 nm (Fig. 11), which is about 10-fold smaller than in RESS. In addition, due to the solubility enhancement, the CO₂ requirement is about 28-fold lower.

Aminobenzoic Acid Nanoparticles

By using menthol cosolvent, the solubility of 2-aminobenzoic acid can be enhanced by up to 100-fold as shown in Figure 12 (23). The RESS–SC process produced ~80 nm size nanoparticles, which is significantly smaller than the ~610 nm size nanoparticles obtained from the RESS process. Menthol is
**Table 3** Solubility of Griseofulvin in Supercritical CO₂ with Menthol Cosolvent

<table>
<thead>
<tr>
<th>P (bar)</th>
<th>T (°C)</th>
<th>Menthol amount (µmol/mol)</th>
<th>Griseofulvin solubility (µmol/mol)</th>
<th>Enhancement factor&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>96</td>
<td>40</td>
<td>21,000</td>
<td>27</td>
<td>28</td>
</tr>
<tr>
<td>117</td>
<td>40</td>
<td>25,000</td>
<td>71</td>
<td>–</td>
</tr>
<tr>
<td>130</td>
<td>40</td>
<td>37,000</td>
<td>133</td>
<td>20</td>
</tr>
<tr>
<td>198</td>
<td>40</td>
<td>42,000</td>
<td>217</td>
<td>15</td>
</tr>
<tr>
<td>239</td>
<td>40</td>
<td>60,000</td>
<td>266</td>
<td>15</td>
</tr>
<tr>
<td>96</td>
<td>50</td>
<td>5,000</td>
<td>2</td>
<td>15</td>
</tr>
<tr>
<td>130</td>
<td>50</td>
<td>24,000</td>
<td>43</td>
<td>12</td>
</tr>
<tr>
<td>164</td>
<td>50</td>
<td>34,000</td>
<td>110</td>
<td>15</td>
</tr>
</tbody>
</table>

<sup>a</sup>Ratio of griseofulvin solubility in menthol/CO₂ to that in pure CO₂.

**Abbreviation:** CO₂, carbon dioxide.
Before the invention of the RESS–SC process, the low-solubility aspect of supercritical CO₂ was utilized to produce particles by its antisolvent action. The drug is dissolved in an organic solvent, and then the solution is injected into supercritical carbon dioxide. The SCF, due to its high diffusivity, rapidly extracts the solvent precipitating the drug particles. A schematic of the supercritical antisolvent (SAS) concept is shown in Figure 14.

The SAS process has been proposed with numerous acronyms (SAA, SEDS, GAS, ASES, etc.) in the literature, but the basic concepts remain the same. Typically, 50–200 μm nozzles have been utilized in SAS. When the injection of the drug solution is complete, a washing step is carried out to remove the organic solvent so as to prevent it from condensing during
the depressurizing step. For this purpose, the feed of supercritical CO₂ is maintained to carry out the residual solvent. Once all the residual solvent is removed, the vessel pressure is reduced to atmospheric pressure, and the solid particles are collected on a filter at the bottom of the vessel. A review of SAS-based processes is provided by Jung and Perrut and by Charbit et al. (24,25). A polymer can be coprecipitated along with the drug to obtain controlled release formulation (26,27).

Figure 13 2-Aminobenzoic acid particles from (A) RESS and (B) RESS–SC processes. Abbreviations: RESS, rapid expansion of supercritical solution; RESS–SC, rapid expansion of supercritical solution solid cosolvent.

Figure 14 Schematic of SAS process. Abbreviation: SAS, supercritical antisolvent.
The particle size and morphology depends on the nozzle geometry, solution velocity, CO₂ pressure, and the type of organic solvent used. The SAS process provides mostly 15 μm drug particles. Examples include 10–40 μm acetaminophen from ethanol, 1–10 μm ascorbic acid and aspirin from ethanol, 1.2–2 μm budesonide from methylene chloride, 0.5–20 μm camptothecin from dimethyl sulfoxide, 1–5 μm chlorpheniramine maleate from methylene chloride, 1.7 μm fluticasone-17-propionate from methylene chloride, 14 μm ibuprofen from methanol, 1–5 μm indomethacine from methylene chloride, 1–10 μm insulin from hexafluoro isopropanol, 1–5 μm insulin from dimethyl sulfoxide, 0.5–5 μm insulin from ethanol, 1–5 μm lysozyme from dimethyl sulfoxide (Winters #115), 1–10 μm paracetamol and saccharose from ethanol, 2–20 μm sulfathiazole from acetone and methanol, and 1.5 μm trypsin from ethanol (27–38).

A few SAS studies have produced nanoparticles. These are listed in Table 4, along with the process conditions used.

In SAS, the inability to form small nanoparticles and to have a narrow size distribution can be attributed to particle growth after nuclei formation. The main phenomenon in

### Table 4 Drug Nanoparticles from SAS-Based Precipitation Processes

<table>
<thead>
<tr>
<th>Drug</th>
<th>Solvent</th>
<th>( P ) (bar)</th>
<th>( T ) (K)</th>
<th>Particle size (nm)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Albumin</td>
<td>Water/ethanol</td>
<td>50–500</td>
<td>39</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>N-Methylpyrrolidone</td>
<td>150</td>
<td>313</td>
<td>300–1200</td>
<td>40</td>
</tr>
<tr>
<td>Gentamicin/PLA</td>
<td>Methylene chloride</td>
<td>85</td>
<td>308</td>
<td>200–1000</td>
<td>41</td>
</tr>
<tr>
<td>Hydrocortisone</td>
<td>Dimethyl sulfoxide</td>
<td>100</td>
<td>308</td>
<td>500–1000</td>
<td>29</td>
</tr>
<tr>
<td>Ibuprofen</td>
<td>Dimethyl sulfoxide</td>
<td>100</td>
<td>308</td>
<td>500–1000</td>
<td>29</td>
</tr>
<tr>
<td>Naloxone/l-PLA</td>
<td>Methylene chloride</td>
<td>85</td>
<td>308</td>
<td>200–1000</td>
<td>41</td>
</tr>
<tr>
<td>Insulin</td>
<td>Water/ethanol</td>
<td>50–500</td>
<td>39</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Naltrexen/l-PLA</td>
<td>Methylene chloride</td>
<td>85</td>
<td>308</td>
<td>200–1000</td>
<td>41</td>
</tr>
<tr>
<td>Nicotinic acid</td>
<td>Ethanol</td>
<td>400–750</td>
<td>42</td>
<td></td>
<td></td>
</tr>
<tr>
<td>RhDNase</td>
<td>Ethanol</td>
<td>50–500</td>
<td>39</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salbutamol</td>
<td>Methanol/acetone</td>
<td>100</td>
<td>333</td>
<td>500</td>
<td>42</td>
</tr>
</tbody>
</table>

*Abbreviation: SAS, supercritical antisolvent.*
RESS is the high rate of pressure reduction, where in SAS, it is the high diffusivity of supercritical CO$_2$. The antisolvent action (mixing or mass transfer of solvent and antisolvent) needs to be even faster than SAS, in order to produce smaller particles of $<300$ nm in size.

**SA WITH ENHANCED MASS (EM) TRANSFER (SAS-EM) PROCESS FOR NANOPARTICLE FORMATION**

A significant improvement in the SAS process is introduced by Gupta and Chattopadhyay leading to nanoparticles of controllable size that are up to an order of magnitude smaller than those resulting from the conventional SAS process, and have a narrower size distribution (43). Like the SAS, this process, SAS–EM, utilizes supercritical carbon dioxide as the antisolvent, but in this case the solution jet is deflected by a surface vibrating at an ultrasonic frequency that atomizes the jet into much smaller droplets. Furthermore, the ultrasound field generated by the vibrating surface enhances mass transfer and prevents agglomeration through increased mixing. The particle size is controlled by varying the vibration intensity of the deflecting surface, which in turn is easily adjusted by changing the power supplied to the attached ultrasound transducer. The SAS–EM process is shown in Figure F15.

The SAS–EM process has been demonstrated by the formation of tetracycline, griseofulvin, lysozyme, and dexamethasone phosphate nanoparticles (44–46). The size is easily varied from 100 to 1000 nm by the power supply knob on the ultrasonic processor. These results are summarized in Table 5.

SAS–EM has been scaled up by Thar Technologies (www.thartech.com) for production at pilot scale (Fig. 16). This unit can produce up to 1 kg nanoparticle/day. It has one precipitation vessel and two separate collection vessels. One collection vessel can be used to collect the nanoparticles, while the other can be used to remove the nanoparticles for final use. The system is fully automated and can provide nanoparticles.
continuously. The ultrasound power supply is controlled by a computer, which in turn controls the nanoparticle size.

**FUNDAMENTALS GOVERNING PARTICLE FORMATION WITH RESS AND SAS**

Both SAS and RESS are complex processes involving the interaction of jet hydrodynamics, phase equilibrium, nucleation and growth (48,49). In SAS, additional complexity arises because of droplet formation, and mass transfer into and out of the droplets. In both cases, a high supersaturation is achieved, which results in rapid precipitation of the dissolved drug. In RESS, a sudden change in the fluid pressure causes
rapid precipitation, whereas in SAS the sudden diffusion of 
CO\(_2\) into a drug solution causes drug precipitation. For RESS, 
the nanoparticle population balance equation accounting for 
particle nucleation and growth dynamics is as follows (50).

\[
\frac{\partial n}{\partial t} = J(\nu^*) \delta(\nu - \nu^*) - \frac{\partial (Gg n)}{\partial \nu} \\
+ \frac{1}{2} \int_0^{\nu} \beta(\nu - \tilde{\nu}, \tilde{\nu}) n(\nu - \tilde{\nu}, t)n(\tilde{\nu}, t)d\tilde{\nu} \nonumber \\
\times \int_0^{\infty} \beta(\nu, \tilde{\nu}) n(\tilde{\nu}, t)d\tilde{\nu} 
\] (3)

to obtain the number concentration of the particles from nuclea-
tion, condensation, coagulation, and decoagulation. Where \(n\) 
is the number concentration, \(t\) is the time, \(J\) is the nucleation
rate, $\delta$ is the delta function, $v$ is the nanoparticle volume, $G_g$ is the condensation rate, and $\beta$ is the coagulation function.

Nucleation rate, $J$, is obtained from supersaturation (51)

$$J = 2N_2 \frac{Py_2}{\sqrt{2\pi m_2 kTL}} \frac{1}{kT} \left[ \frac{\sigma (v_2^2)}{kT} \exp \left\{ - \frac{16\pi}{3} \left( \frac{\sigma (v_2^2)^{2/3}}{kT} \right) \right\} \right]$$

where $y_2$ is the actual drug mole fraction in CO$_2$ phase; $y_2^{eq}$ is the equilibrium drug mole fraction over a flat surface (i.e., solubility); $S$ is the supersaturation ratio, $y_2/y_2^{eq}$; $k$ is the

Figure 16  SAS–EM commercial unit by Thar Technologies, Inc. Abbreviation: SAS–EM, supercritical antisolvent with enhanced mass transfer.
Boltzmann constant; $N_2$ is the number concentration of the solute in the fluid phase; and $P$ is the pressure. The equilibrium solubility can be obtained from Equation (1) as discussed earlier. It will be a function of pressure, temperature, and cosolvent if present.

Particles grow by the condensation of solute from the fluid phase onto the particle surface. The net rate of a single molecule condensation onto a spherical particle is given by (52),

\[ G_g = G_g = 2 \pi d_p D \left[ N_2 - N_2^{eq}(g) \right] \]

where $d_p$ is the diameter of spherical particles containing $g$ molecules and $D$ is the diffusion coefficient for the solute molecule in the fluid phase.

The particle size and concentration can also change by coagulation and decoagulation. For coagulation of two particles (1 and 2), rate of coagulation ($J'$) can be expressed as (53)

\[ J' = K_{12} N_1 N_2 \]

where $N_1$ and $N_2$ are the number concentrations of the coagulating particles and $K_{12}$ is the effective coagulation coefficient given as

\[ K_{12} = \left[ \frac{2kT}{3\mu} \left( \frac{D_{p1} + D_{p2}}{D_{p1}D_{p2}} \right)^2 \right] + \left[ \frac{du}{dy} \left( \frac{D_{p1} + D_{p2}}{6} \right)^3 \right] + \left( \frac{\pi \varepsilon_k}{120n} \right)^{1/2} \left( \frac{D_{p1} + D_{p2}}{3} \right) \]

which is the sum of Brownian, laminar shear, and turbulent coefficients. And

\[ N_i(r, t) = N_i(0) \left[ 1 - \frac{D_{p1} + D_{p2}}{2r} \text{erfc} \left( \frac{2r - (D_{p1} + D_{p2})}{4\sqrt{D_{12}t}} \right) \right] \]

where $du/dy$ is the velocity gradient in the case of laminar flow; $\varepsilon_k$ is the rate of dissipation of kinetic energy per unit mass; $\nu$ is the kinematic viscosity of the fluid; $r$ is the distance of the particle from the center of the fixed particle; and $D_{12}$ is the effective diffusion coefficient.
OTHER APPLICATIONS OF SCFs FOR PARTICLE ENGINEERING

SCFs can be applied to a variety of other applications where nano- and microdimensions of the drug material in excipient are important for drug release (54). These include the following.

**Porous Particles and Polymer Foams**

Since a fast removal of dissolved CO₂ can be achieved by rapid depressurization, this behavior can be used to create foams, especially that of poly(lactide–co–glycolide) (PLGA) polymer, because CO₂ has a good solubility in this approved polymer. Hile et al. (55) prepared PLGA foam capable of sustained release of basic fibroblast growth factor for tissue engineering applications. To prepare the foam, a water-in-oil microemulsion consisting of an aqueous protein phase (typical reverse micelle domain size of 5–10 nm) and an organic polymer solution was prepared. The microemulsion was filled in molds and then placed in a pressure vessel. Now, the pressure vessel was pressurized with supercritical CO₂, to extract the organic phase, causing the polymer to precipitate onto the protein droplets. Now the vessel is purged with more CO₂ to remove the solvent from the system. Finally, the vessel is depressurized in 10–12 sec causing rapid removal of the CO₂ that was dissolved in the polymer, making a porous foamy structure.

Koushik and Kompella (56) employed an SCF pressure-quench technique to create porous peptide (deslorelin) encapsulating PLGA particles (Fig. 17). On SC CO₂ treatment (1200 psi, 33°C for 30 min) of deslorelin, PLGA particles prepared using emulsion–solvent evaporation, the mean particle size of the deslorelin PLGA microparticles increased from 2.2 to 13.8 μm, the mean porosity increased from 39% to 92.38%, the mean bulk density reduced from 0.7 to 0.082 g/cm³, mass spectrometry indicated structural integrity of released deslorelin, the circular dichroism spectrum indicated stabilization of β-turn conformation of the peptide, and the scanning elec-
Electron microscopy confirmed increased particle size and pore formation. Further, the deslorelin release was sustained during the seven-day study period and the residual solvent content was reduced from 4500 ppm to below the detection limit (<25 ppm).

**Liposomes**

Liposomes, in which nanodomains of drug are stabilized using lipids, are useful drug carriers for both small and macromolecular drugs. Unfortunately, the conventional methods of making liposomes require large amounts of organic solvents and have difficulty with scale-up for hydrophilic drugs. Lipids actually have some solubility in supercritical CO₂, and this behavior has been used to form liposomes without using organic solvents. For example, Fredereksen et al. (52) dissolved a phospholipid (1-palmitoyl-2-oleoylphosphatidylcholine) and cholesterol in supercritical CO₂ using 7% ethanol cosolvent. The mixture is expanded into an aqueous state containing fluorescein isothiocyanate (FITC)–dextran at low pressure. Because of the sudden reduction in the solubility of the phospholipid and the cholesterol at the nozzle tip, liposome-encapsulating FITC–dextran was formed. The process yielded 200-nm-size liposomes (termed as critical fluid liposomes) with 20% encapsulation efficiency. The main benefit of this process is the significantly reduced use of organic solvent. Later, Castor and Chu (57) prepared liposomes

---

**Figure 17** Supercritical-fluid pressure-quench technique to create porous microparticles. Abbreviation: CO₂, carbon dioxide. Source: From Ref. 56.
containing hydrophobic drugs, such as paclitaxel, camptothecins, doxorubicin, vincristine, and cisplatin. These formulations including 150–250-nm paclitaxel liposomes are claimed to be more effective against tumors in animals compared to commercial formulations.

Inclusion Complexes

Inclusion compounds, such as inclusion of poorly water-soluble drugs in cyclodextrin, are useful in enhancing bioavailability. Basically, the lipophilic drug is included in the lipophilic interior of the cyclodextrin molecule. The exterior of the cyclodextrin molecule is hydrophilic, and hence the whole complex can be dissolved in water. Inclusion can be achieved when both the drug and the cyclodextrin molecules are in a dissolved state, i.e., have a higher molecular mobility as compared to the solid forms. In conventional technique, both are dissolved in an organic solvent and then the solvent is removed. Unfortunately, the concentration of the residual solvent is high in the final product (58).

Supercritical CO$_2$ processes allow preparation of drug–cyclodextrin inclusion complexes without the use of organic solvents. This is because the interaction of supercritical CO$_2$ with solid cyclodextrin makes the cyclodextrin molecules more fluid. This interesting plasticizing effect of supercritical CO$_2$ has been well known for organic polymers, for which the glass transition or melting can be achieved at a lower temperature with SC CO$_2$. To make inclusion compounds, the physical solid mixture of the drug and cyclodextrin is exposed to supercritical CO$_2$, and then rapidly CO$_2$ is removed by depressurization.

Bandi et al. (59) prepared budesonide and indomethacin hydroxypropyl–cyclodextrin (HPBCD) complexes using an organic solvent-free SCF process (59,60). The process involved the exposure of drug–HPBCD mixtures to supercritical carbon dioxide. The ability of the SCF process to form complexes was assessed by determining drug dissolution using a high-performance liquid chromatography assay, crystallinity using powder x-ray diffraction (PXRD) and differential scanning calorimetry.
calorimetry, and drug–excipient interactions using Fourier transform infrared spectroscopy (FTIR). The SC CO₂ process did not alter the dissolution rate of pure drugs but resulted in two- and threefold higher dissolution rates for budesonide and indomethacin–HPBCD mixtures, respectively. SCF-processed mixtures exhibited a disappearance of the crystalline peaks of the drugs (PXRD), a partial or a complete absence of the melting endotherm of the drugs (DSC), and a shift in the C=O stretching of the carboxyl groups of the drugs (FTIR), consistent with the loss of drug crystallinity and the formation of intermolecular bonds with HPBCD. Thus, budesonide and indomethacin–HPBCD complexes with an enhanced dissolution rate can be formed using a single-step, organic solvent-free SC CO₂ process. Similar inclusion complexes were also reported for piroxicam using a supercritical CO₂ process (61).

Solid Dispersions

In many delivery applications, molecularly intimate mixtures (i.e., solid dispersion) of drug with excipients, such polymers are needed. An organic solvent, which can dissolve both, does bring the two in intimate contact while in solution. Unfortunately, when the solvent is removed by evaporation or by addition of a liquid antisolvent, the drug and the polymer phases precipitate out or separate. Hence, the dispersion of the two is poor in the solid state. Supercritical CO₂ antisolvent induces the precipitation about 100-fold faster than the liquid antisolvent, not allowing enough time for the drug and the polymer domains to separate out. Thus, supercritical CO₂ precipitation can provide a more dispersed solid mixture. Supercritical CO₂-based precipitation is superior to the liquid-based precipitation or the milling process. For example, a solid dispersion of carbamazepine in polyethylene glycol (PEG)-4000, produced by CO₂ method, increased the rate and the extent of dissolution of carbamazepine (62). In this method, a solution of carbamazepine and PEG4000 in acetone was loaded in a pressure vessel, in which supercritical CO₂ was added from the bottom to obtain solvent-free particles.
SAFETY AND HEALTH ISSUES

When dealing with supercritical carbon dioxide, there are two safety and health issues that are to be kept in mind when designing and operating the extractor: (i) the high pressure involved requires that personnel is protected from the plant by proper isolating walls and (ii) if carbon dioxide is released in the closed atmosphere it can lead to asphyxiation, as it can replace the oxygen in the surroundings.

CONCLUSIONS

For particle formation, SCF technology offers two processes: (i) RESS for drugs that are soluble in supercritical CO₂ and (ii) SAS for drugs that are poorly soluble in supercritical CO₂. In RESS, a sudden change in the fluid pressure causes rapid precipitation, whereas in SAS the sudden diffusion of CO₂ into a drug solution causes drug precipitation. Conventionally, both the technologies have produced microparticles in the 1–5-μm-size range. With enhancement in mixing, SAS-EM process produces nanoparticles of controllable size. With the reduction in particle coagulation, the RESS–SC process produces nanoparticles with a high yield. The RESS–SC equipment is expected to be cheaper than SAS–EM, because the residence time of the drug in the high-pressure chamber is lower in the former. The particle formation techniques can also be employed for the preparation of liposomes and solid dispersions of drugs and solubility enhancing carriers. In addition, SCF exposure or pressure-quench techniques can be employed to form porous structures or inclusion complexes and to remove residual solvents in pharmaceutical particulate systems.

REFERENCES


56. Koushik K, Kompella UB. Preparation of large porous deslor- 
elin-PLGA microparticles with reduced residual solvent and 
cellular uptake using a supercritical CO₂ process. Pharm Res 

57. Castor TP, Chu L. Methods and apparatus for making li-po-
somes containing hydrophobic drugs. U.S. Patent 5,776,486, 
1998.

58. Lin SY, Kao YH. Solid particulates of drug-b-cyclodextrin 
inclusion complexes directly prepared by a spray-drying tech-

59. Bandi N, Wei W, Roberts CB, Kotra LP, Kompella, UB. 
Preparation of budesonide- and indomethacin-hydroxypropyl-
β-cyclodextrin (HPβCD) complexes using an organic-solvent-
free, single-step supercritical fluid process. Eur J Pharm Sci 
2004; 23(2):159–168.

60. Mayo A, Kompella UB. Supercritical fluid technology in 
pharmaceutical research. In: James S, ed. Encyclopedia of 
Pharmaceutical Technology. New York: Marcek Dekker Inc., 

Application of supercritical carbon dioxide for the preparation 
of a piroxicam-beta-cyclodextrin inclusion compound. Pharm 

62. Moneghini M, Kikic I, Voinovich D, Perissutti B, Filipovic-Grcic 
J. Processing of carbamazepine-PEG 4000 solid dispersions 
with supercritical carbon dioxide: preparation, characteriza-
tion, and in vitro dissolution. Int J Pharm 2001; 222(1): 
129–138.