Review

Design of PLGA-based depot delivery systems for biopharmaceuticals prepared by spray drying

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\textbf{A R T I C L E  I N F O}

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\textbf{A B S T R A C T}

Currently, most of the approved protein and peptide-based medicines are delivered via conventional parenteral injection (intramuscular, subcutaneous or intravenous). A frequent dosing regimen is often necessary because of their short plasma half-lives, causing poor patient compliance (e.g. pain, abscess, etc.), side effects owing to typical peak-valley plasma concentration time profiles, and increased costs. Among many sustained-release formulations poly lactic-co-glycolic acid (PLGA)-based depot microparticle systems may represent one of the most promising approaches to provide protein and peptide drugs with a steady pharmacokinetic/pharmacodynamic profile maintained for a long period. However, the development of PLGA-based microparticle systems is still impeded by lack of easy, fast, effective manufacturing technologies. The aim of this paper is to review recent advances in spray drying, a one-step, continuous microencapsulation process, for manufacturing of PLGA-based depot microparticle systems with a focus on the recent efforts on understanding of the role of nozzle design in the microencapsulation of proteins/peptides, and the effect of critical solvent properties and process parameters on the critical quality attributes of the spray-dried microparticles.

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1. Introduction

As a result of the development of recombinant DNA technology in the past two decades, over 200 biopharmaceutical products are on the market with an annual revenue of nearly US \$ 167 billion
(Rader, 2013; Walsh, 2010). There are more and more new medicines based on biotechnology rather than organic chemistry. This is because biopharmaceuticals are often less toxic, more specific and have a more predictable in vivo action than small-molecule drugs (Frokjaer and Otzen, 2005). In spite of this, several challenges confront pharmaceutical scientists for the efficacious and safe delivery of proteins and peptides, such as (i) their intrinsic physicochemical instability, (ii) their poor permeability through biological membranes, (iii) their short plasma half-lives (Frokjaer and Otzen, 2005; Moeller and Jorgensen, 2008). Hence, most of the approved protein and peptide-based medicines are delivered via a needle (intramuscularly, subcutaneous or intravenous) because of the assured bioavailability. However, a frequent dosing regimen is often necessary because of their short plasma half-lives, which further causes poor patient compliance (e.g. pain, abscess, etc.), unwanted side effects, and increased costs.

Vast efforts have been focused on the development of novel strategies for extending the duration of proteins and peptides in vivo, including delivery of biopharmaceuticals via sustained-release drug delivery systems (Cun et al., 2015; Shi and Li, 2005; Yang et al., 2012a), and modification of proteins and peptides via chemical approaches such as PEGylation, acetylation, and protein fusion (Beals and Shanafelt, 2006; Yang and Frokjaer, 2009). To date, peptides/proteins with chemical modification are typically limited to weekly dosing, in contrast, it is possible to obtain the one to three months of pharmacological effect with polymer based sustained-release systems (Schwendeman et al., 2014). Among them, poly lactic-co-glycolic acid (PLGA)-based particulate delivery systems may represent one of the most promising approaches. PLGA, a copolymer that is synthesized through random ring-opening co-polymerization of two different monomers, the cyclic dimers (1,4-dioxan-2,5-diones) of glycolic acid and lactic acid, has been approved by the US FDA and European Medicine Agency (EMA) (Wischke and Schwendeman, 2008). It is now commercially available with different molecular weights and copolymer compositions and the properties of the polymer can be tailored by varying the molecular weight, L/G ratio, PLA stereoregion, end-group functionalization, and PEGylation. The general trends are summarized in Table 1. To date, PLGA-based particle systems have been investigated to deliver various biopharmaceutical agents (e.g. leuprolide, octreotide, sCT, rGH, insulin, rhEPO, DGR, VEGF, BDNF, rhEGF, INF-α2b, vaccines, siRNA, etc.) via injection routes (i.e. intravenous, subcutaneous or intramuscular injection) (Bertram et al., 2010; Cun et al., 2010; Desai and Schwendeman, 2013; Geng et al., 2008; Han et al., 2001; He et al., 2006; Jensen et al., 2010; Park and Na, 2009; Takada et al., 2003; Ungaro et al., 2009; Wang et al., 2004; Zhang et al., 2008). In the last two decades, several products based on PLGA depots have been brought to the market (Table 2) (Kumar and Palmieri, 2010; Kumar et al., 2006; Ye et al., 2010). Besides systemic therapy, PLGA depots have also been investigated to deliver the drugs for local/site-specific therapy, e.g. intra-ocular and intra-articular drug delivery, and several PLGA-based products for intra-ocular and intra-articular drug delivery have entered into the clinical trials (Evans et al., 2014; Kuno and Fujii, 2011). In addition, PLGA microparticle formulations have also been intensively investigated for lung administration (Aguiar et al., 2004; Edwards et al., 1997; Jensen et al., 2010; Kawashima et al., 1999; Yang et al., 2012a,b). However, no PLGA-based sustained pulmonary delivery system has yet advanced to the clinical trial stage.

The development of PLGA-based particulate delivery systems is still impeded by a variety of challenges, including initial burst release, incomplete protein release, and poor protein stability during the production process, storage, and drug release process. The total body of literature in the area is immense and the reader is referred to the excellent review articles (Fredenberg et al., 2011; Giteau et al., 2008; Schwendeman et al., 2014; Weert et al., 2000; Wischke and Schwendeman, 2008). In addition, for the protein and peptide drugs, the lack of easy, fast, effective microencapsulation technologies represents one of the major hurdles for commercialization of proteins-loaded PLGA particulate delivery systems (Kumar and Palmieri, 2010). The complexity of the production process can not only increase the risk of protein denaturation, but also make the sterilization of products difficult, often eventually resulting in the commercial failure of the products (Desai and Schwendeman, 2013; Shi and Li, 2005). For example, the first FDA approved injectable protein depot (Nutropin Depot, produced by Cryogenic spray-drying technology) was finally discontinued by the manufacturers due to the complexity of the manufacturing process (Desai and Schwendeman, 2013). Currently, there are a few production technologies available for preparation of PLGA based depot delivery systems, which have been reviewed elsewhere (Wan et al., 2013b; Ye et al., 2010; Yeo et al., 2001). In this review we will focus on the spray drying process to design PLGA-based depot delivery systems for proteins and peptides. Especially, we will discuss the role of nozzle design and solvent properties in the spray drying process, which influence the PLGA microparticle formation process and critical quality attributes of the product.

2. Current production technologies of PLGA-based particle products

A variety of microencapsulation methods have been developed over past decades, including emulsion methods (Cui et al., 2005; Cun et al., 2008), phase separation (coacervation) (Nihant et al., 1995; Nihant et al., 1994), spray drying (Mok and Park, 2008; Wan et al., 2014a,b), spray freeze drying (SFD) (Bi et al., 2008; Saluja et al., 2010), electrospray (ES) drying (Bohr et al., 2015, 2012) and supercritical fluid technology (SCF) (Casettari et al., 2011; Salmaso et al., 2009). The features of the current widely used microencapsulation technologies are summarized in Table 3.

Nowadays, emulsion methods, coacervation and spray drying have generally reached large scale production and have been utilized to produce the marketed PLGA-based particle products (Kumar and Palmieri, 2010). However, manufacturers still face many problems, such as the significant unanticipated costs in scale-up production, the residual organic solvent and sterilization (Schwendeman et al., 2014). Spray drying, well known as a one-

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Table 1: Key parameters and corresponding effects on PLGA properties (adapted from (Mader, 2011)).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Influence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Molecular weight (Mn)</td>
<td>High Mn results in a longer degradation time</td>
</tr>
<tr>
<td>L/G ratio</td>
<td>Polymers with one monomer degrade more slowly</td>
</tr>
<tr>
<td>Stereochemistry</td>
<td>Degradation times: PLA &gt; PGA &gt; PLGAS0:50</td>
</tr>
<tr>
<td>Blockage of acidic endgroups</td>
<td>L-PLA: semicrystalline</td>
</tr>
<tr>
<td>PEGylation</td>
<td>α,β-PLA: amorphous</td>
</tr>
<tr>
<td></td>
<td>Polymers with free-COOH group are more hydrophilic</td>
</tr>
<tr>
<td></td>
<td>Increase in hydrophilicity</td>
</tr>
</tbody>
</table>
step, continuous production process, has attracted lots of interests owing to its advantages such as possibility of scaling-up production with automatic controlling, cost-effectiveness, commercial availability of different process layouts, and suitability for different types of feeds (e.g., solutions, suspensions, or emulsions).

3. The spray drying process for production of PLGA-based depot delivery systems

Spray drying is the process of transforming a liquid feed into a dried powder by spraying the feed into a hot drying medium (air, inert gas such as nitrogen), which can be divided into three steps: (i) atomization of the feed into small droplets via an atomizer; (ii) drying of the droplets upon contact with the drying gas and particle formation; and (iii) separation of the dry particles from the drying medium. As a scalable continuous process technology, it covers production rates from milligram quantities to tons per hour. The original purpose of introducing spray drying into pharmaceutical industry was just to obtain the dried products by the removal of solvent from feedstock (Snyder, 2012). With the expanding requirements for the precise control of solid-dosage particle properties in pharmaceutical industry, its applications are shifting to the category of ‘particle engineering’ in recent literature due to its ability to manipulate the individual particle attributes at a uniquely high level of precision through a combination of formulation and process parameters (Baldinger et al., 2011; Lebrun et al., 2012; Maltesen et al., 2008; Paudel et al., 2013; Vehring, 2008). The particle engineering potential of the spray drying process is summarized in Table 4.

3.1. Atomization of the feed into small droplets

In the spray drying process, the bulk fluid is usually broken into individual droplets through an atomizer. Numerous different atomizing devices have been developed, and rotary atomizers, two-fluid nozzles and pressure nozzle are the most widely used in industry. In recent years, a variety of novel nozzles has been developed for the various purposes. For example, the piezoelectric mesh-nozzle (piezoelectric driven spray head) is able to generate fine droplets in the size range of 300 nm to 5 μm (Lee et al., 2011).

Table 2
PLGA-based microparticles available on the market and their production technologies.

<table>
<thead>
<tr>
<th>API</th>
<th>Polymer</th>
<th>Commercial name</th>
<th>Technology</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td>Somatropin (rDNA orgin)</td>
<td>PLGA</td>
<td>Nutropin Depot&lt;sup&gt;®&lt;/sup&gt;</td>
<td>Alkermes ProLease&lt;sup&gt;®&lt;/sup&gt; (Cryogenic spray-drying)</td>
<td>Alkermese &amp;Genentech</td>
</tr>
<tr>
<td>Luprolide acetate</td>
<td>PLA or PLGA</td>
<td>Lupron Depot&lt;sup&gt;®&lt;/sup&gt;</td>
<td>Double emulsion (water in oil in water)</td>
<td>TAP</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Trenantone&lt;sup&gt;®&lt;/sup&gt;</td>
<td></td>
<td>Takeda</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Enantone Gyn&lt;sup&gt;®&lt;/sup&gt;</td>
<td></td>
<td>Takeda</td>
</tr>
<tr>
<td>Goserelin acetate</td>
<td>PLA</td>
<td>Zoladex&lt;sup&gt;®&lt;/sup&gt;</td>
<td>N/A</td>
<td>AstraZeneca</td>
</tr>
<tr>
<td>Octreotide acetate</td>
<td>PLGA-glucose</td>
<td>Sandostatin&lt;sup&gt;®&lt;/sup&gt; LAR</td>
<td>Phase separation</td>
<td>Novartis</td>
</tr>
<tr>
<td>Triptorelin pamoate</td>
<td>PLGA</td>
<td>Trelstar&lt;sup&gt;®&lt;/sup&gt; Depot</td>
<td>Phase separation</td>
<td>Pfizer</td>
</tr>
<tr>
<td></td>
<td>PLGA</td>
<td>Decapeptyl&lt;sup&gt;®&lt;/sup&gt; SR</td>
<td>Phase separation</td>
<td>Ipsen-Beaufour</td>
</tr>
<tr>
<td>Lanreotide</td>
<td>PLGA</td>
<td>Somatuline&lt;sup&gt;®&lt;/sup&gt; LA</td>
<td>Phase separation</td>
<td>Ipsen-Beaufour</td>
</tr>
<tr>
<td>Buserelin</td>
<td>PLGA</td>
<td>Suprecur&lt;sup&gt;®&lt;/sup&gt; MP</td>
<td>N/A</td>
<td>Aventis</td>
</tr>
</tbody>
</table>

Table 3
Summary of the features of selected particle engineering technologies.

<table>
<thead>
<tr>
<th>Technology</th>
<th>Pros.</th>
<th>Cons.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Emulsion methods</td>
<td>Controlled particle size with relatively narrow distribution; widely used for the delivery of small molecular drugs and small peptides; feasibility for scale-up production</td>
<td>Relatively low encapsulation efficiency; the use of toxic organic solvents; incomplete protein release, etc.</td>
</tr>
<tr>
<td>Phase separation (coacervation)</td>
<td>Relatively high encapsulation efficiency; effective control of the particle size with narrow size distribution</td>
<td>Particle aggregation, difficult scale-up production; residual solvents</td>
</tr>
<tr>
<td>Spray drying</td>
<td>Fast, continuous, one-step process; relatively easy to scale-up and cost-effective; ability to control physical properties (size, density, surface property, etc.) and be fully automated; relatively high drug entrapment efficiency</td>
<td>Potential protein denaturation due to the thermal stress, atomization shear stress and interfacial stress (air-liquid or water-oil) in the spray drying process</td>
</tr>
<tr>
<td>Spray freeze drying</td>
<td>Porous spherical particles with extremely low density; avoiding denaturation due to the thermal stress</td>
<td>Particles are relatively fragile; potential protein denaturation due to the stresses associated with freezing and air-liquid interface generated during atomization, time consuming, and safety issue</td>
</tr>
<tr>
<td>Electrospray</td>
<td>Particle size within hundreds of micrometers down to several nanometers; controlled particle size with nearly monodisperse distribution</td>
<td>Low output due to the application of the cone jet electrospray</td>
</tr>
<tr>
<td>Supercritical fluid technology</td>
<td>Small particles with a narrow size distribution; avoiding the use of flammable, toxic solvents; rapid removal of the SCF and solvent without the need for an extensive drying step</td>
<td>Non-uniform mixing and long time scales due to gradual introduction of the drug solution; limited solvation power of SCs; complex process</td>
</tr>
</tbody>
</table>
Table 4
Particle engineering via the spray drying process and their pharmaceutical application.

<table>
<thead>
<tr>
<th>Engineered particle properties</th>
<th>Pharmaceutical Function</th>
<th>Representative references</th>
</tr>
</thead>
<tbody>
<tr>
<td>Precise size control (‘tunable’ particle-size distributions with sub-micron precision)</td>
<td>Pulmonary delivery application requires precise particle size control; Improve bioavailability of BCS II drugs by enhanced dissolution rate through increased API-specific surface area (reduced particle size);</td>
<td>Chougule et al. (2008); Jensen et al. (2010); Maltesen et al. (2008)</td>
</tr>
<tr>
<td>Density modulation</td>
<td>Pulmonary delivery application requires precise particle density control;</td>
<td>Maltesen et al. (2008); Tsapis et al. (2002); Vanbever et al. (1999)</td>
</tr>
<tr>
<td>Surface roughness (smooth vs wrinkled surfaces)</td>
<td>Improve the fluidity and dispersability of the powder products;</td>
<td>Kwok et al. (2011); Wang et al. (2015)</td>
</tr>
<tr>
<td>Solid-state modulation</td>
<td>To improve the dissolution rate of poorly water soluble APIs by transferring them from crystalline form to amorphous form; Improved product bioavailability utilizing polymer solid dispersions to stabilize active amorphous forms and enhance API dissolution rates compared with crystalline structures;</td>
<td>Fouad et al. (2011); Paudel and Mooter (2012); Paudel et al. (2013)</td>
</tr>
<tr>
<td>Coating/Microencapsulation</td>
<td>Controlled release of API from solid dispersions of polymer matrices and encapsulation at the particle level; Taste masking of API via individual particle coating;</td>
<td>Kondo et al. (2014); Legako and Dunford (2010); Meeus et al. (2015a,b); Pabari et al. (2012)</td>
</tr>
</tbody>
</table>

BCS, biopharmaceutics classification system.

In addition, the co-axial ultrasonic atomizer and the 3-fluid nozzle have been developed and applied for microencapsulation (Wan et al., 2014b; Wen et al., 2013; Yeo and Park, 2004). Their features are summarized in Table 5. In addition to the atomizer design, the operating conditions and the rheology of bulk fluids also play very important roles in controlling the atomization characteristics (e.g. droplet size and distribution). For the rotary atomizer, the channel design, the disc angular velocity and the feed flow rate control the mean droplet size. For example, the disc diameter is usually 15–20 cm and a rotary atomizer is normally used when the desired droplet size falls within the range 20–150 μm which requires disc edge velocities of 180 and 75 m/s, respectively (Masters, 1991). As to two-fluid nozzles, gas flow properties such as velocity and density also influence the droplet size (Sloth, 2007). Furthermore, it should be noted that the gas applied can also influence atomization as a result of its density or oxygen content (Aftel et al., 1996).

Additionally, the rheological properties of the bulk fluid can influence the droplet size distribution. For example, the mean droplet size increases with an increase in the viscosity of the bulk fluid. Usually, the viscosity of bulk fluids should be sufficiently low for pumping to provide a consistent mass flow to the atomizer. When the high viscous bulk fluid is spraying, one has to take into account the blockage of the feeding line (Snyder, 2012).

3.2. Droplet drying and particle formation

Once the droplets enter the drying chamber, the droplets drying/particles formation take place upon contacting with the drying gas, which is driven by the difference between the vapor pressure of the solvents and their partial pressure in the gas phase. The kinetics of the droplet drying/particle formation highly influence the physicochemical properties of the dried particles, thus, it is a prerequisite for rational particle engineering to

Table 5
The typical features of the atomizers commonly used in literature.

<table>
<thead>
<tr>
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</tr>
</thead>
<tbody>
<tr>
<td>Rotary atomizer</td>
<td>The bulk fluid is flung off a rotating disk and broken into droplets at the edge of disc due to the centrifugal force</td>
<td>Rarely blocks; enables spray generation from very different solutions and suspensions; relatively narrow droplet size distribution</td>
<td>Mechanically complex and requires regular maintenance of the moving parts</td>
</tr>
<tr>
<td>Two-fluid nozzle</td>
<td>After contacting with a high velocity gas, the liquid jet is atomized into droplets by the high frictional forces between the gas and the liquid jet</td>
<td>Smaller droplet sizes; narrow droplet size distribution, enables production of droplets with a diameter in the range of a few microns up to about 300 μm</td>
<td>Utilized in smaller scale spray dryers due to the limited drying residence time</td>
</tr>
<tr>
<td>Pressure nozzle</td>
<td>Pressurized liquid is used to atomize through a restricting orifice or via a tangentially fed swirl chamber</td>
<td>Mechanically simple</td>
<td>Wide size distribution; easily blocks</td>
</tr>
<tr>
<td>Ultrasonic atomizer</td>
<td>Ejection of droplets from a liquid film formed on a surface with vibration induced by the passage of ultrasonic energy</td>
<td>Uniform, controllable particle size and distribution,</td>
<td>Harsh operating conditions for biomacromolecules, cannot handle high flow rates</td>
</tr>
<tr>
<td>Piezoelectric mesh-nozzle</td>
<td>Ultrafine droplets are generated by a vibrating mesh: when the piezoelectric actuator is driven at an ultrasonic frequency, the mesh will vibrate upwards and downwards, injecting precisely sized droplets from the holes and generating the aerosols</td>
<td>Precise control over the particle size and distribution</td>
<td>Droplet coalescence; harsh operating conditions for biomacromolecules</td>
</tr>
</tbody>
</table>
understand the droplets drying/particles formation process. The process in the classic drying theory is divided into four phases (Fig. 1):

(i) the droplet firstly undergoes rapid heating but without mass change;
(ii) the droplet experiences rapid mass losses and shrinks, but without a change in the droplet surface temperature (usually called ‘constant rate’ period): it is usually modeled based on the $D^2$-law, which has been thoroughly reviewed elsewhere (Vehring, 2008; Vehring et al., 2007). Based on the model, the Peclet number $(Pe) = \frac{k}{BD}$, where $k$ is the drying rate and $D$ is the diffusion coefficient of the solutes “Ⅹ” has been widely used to predict the particle formation process and the particle properties (Pajander et al., 2014; Tsapis et al., 2002; Vehring et al., 2007). In addition, in this constant rate period, the liquid droplet will experience a temperature close to the thermodynamic wet-bulb temperature that is significantly lower than the local drying gas temperature, which provides the important theory for spray drying of thermally labile materials (e.g. biomacromolecules);
(iii) crust starts forming with a sharp increase in the temperature of particle surface, but also with a decrease in the mass transfer rate (usually called ‘falling rate’ period). The solidification of the crust shell creates an internal droplet diffusion-controlled mass transfer process, which reduces the rate of solvent escape from the inner core to the surface;
(iv) crust thickens towards the droplet center until a particle has formed, in which, the particle temperature may rise to the local gas field temperature once evaporation has ceased.

It should be noted that the process of droplet drying and particle formation can be accomplished within a time range from milliseconds to a few seconds according to the different process parameters (e.g. temperature) and formulation parameters (e.g. solvent type).

3.3. Separation of the dry particles from the drying medium

The commonly used approaches to separate the dry powder from the drying medium include cyclone separation and baghouse filtration. Each of them has its own advantages and disadvantages for efficient recovery of spray dried products, which are summarized in Table 6. Recently, the electrostatic precipitator has been developed and applied in Nano Spray Dryer B-90 (Buchi®) to collect the fine particles (Lee et al., 2011).

4. Role of critical solvent properties in preparing PLGA microparticles via spray drying

In general, the solvents, with different solvent properties such as solvent power and solvent volatility, affect the characteristics of spray-dried particles mainly through changing the characteristics of feed solutions and drying kinetics. Herein, the role of co-solvent in preparing the PLGA microparticles via spray drying will be discussed with a focus on the recent understanding of how the critical solvent properties influence the microencapsulation process and the outputs quality.

4.1. Effect of solvent properties on the drying kinetics of droplets and the particle formation

By using the single droplet levitation technique, Schiffter (2005) found that the evaporation of a binary solvent system presented two different stages: (i) a fast evaporation stage, which is determined by the component with a higher vapor pressure at the droplet surface and the droplet radius decreases fast with time; (ii) a slow evaporation stage: with the evaporation proceeding, most of the component with a higher vapor pressure has evaporated and its mass flux turns little, thereby the evaporation process is determined by the evaporation of the component with a slower evaporation rate. It is worth to know that, in theory, some co-solvent mixtures may form an azetropes that may speed up the drying process and influence the particle formation. However, from a practical point of view, it is unlikely to happen in the spray drying process because the formation of azetropes has to be at a fixed pressure (Chen et al., 1995), which is not easy to achieve in the complex and dynamic spray drying process. But, the researchers should bear this issue in mind during selecting the solvent pairs and analyzing the drying kinetics of co-solvent systems. Wulsten et al. investigated the drying kinetics of dichloromethane (DCM)/ethanol (EtOH) binary solvent mixtures (with different compositions) using the droplet levitation technique and found that the drying profiles of DCM:EtOH = 90:10 and 75:25 showed biphasic plots, whereas at the lower weight fraction of DCM (DCM:EtOH = 25:75) the profile was more linear (Wulsten et al., 2009). The observations suggested that the occurrence of a biphasic drying profile was highly dependent on the composition of less-volatile and more-volatile components in the binary solvent system. Further, it can be speculated that the composition of co-solvent systems, the solubility of solutes in the solvent systems, the viscosity of the feed, and the polymer chain architecture in the drying droplets will kinetically shift, which further affects the precipitation process of solutes (particle formation). For examples, in the case study of itraconazole/PVP or HPMC, Wulsten et al. (2009) found that the polymeric component determined the drying rate, whereas the drug determined the particle morphology. The dual functionality of the two solutes could be related to their different precipitation processes due to the different solubilities of the two components in the binary solvent mixtures (DCM and EtOH).

In our previous study, a model system based on monitoring the weight loss of the solvents and the solutions using thermogravimetric analysis (TGA) under controlled temperature, fixed evaporation surface area and constant nitrogen purge was applied to study the drying kinetics of binary solvent systems composed of acetone (ACE) and methanol (MeOH) (Wan et al., 2013a). The evaporation profiles (Fig. 2) showed that the evaporation of the co-solvent systems presented three distinct drying stages, including a fast evaporation stage with slow decline of evaporation rates, a fast

Fig. 1. The schematic representation of development in mass and temperature for the drying of a single droplet inside a spray dryer.
The typical features of cyclone separation and baghouse filtration.

<table>
<thead>
<tr>
<th>Separation approach</th>
<th>Fundamentals</th>
<th>Pros.</th>
<th>Cons.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cyclone (inertial separation method)</td>
<td>Utilize the velocity lag between gas media and dried powder induced by their difference in density.</td>
<td>• Mechanically simple; • Potential of high collection efficiency in a continuous process; • Amenable to cleaning in place</td>
<td>• Collection efficiency is dependent on the hardware dimensional size and particle diameter, which must be addressed for scale-up</td>
</tr>
<tr>
<td>Baghouse filtration</td>
<td>Extract the particles from the gas stream via either a torturous path-depth filter or a size-exclusion membrane filter.</td>
<td>• Relatively higher collection efficiency; • Consistent collection performance over a range of manufacturing scales</td>
<td>• Powder compaction; • Hardware complexity; • Potential of product contamination; • Difficult to clean</td>
</tr>
</tbody>
</table>

A decline of evaporation rate stage, and a slow evaporation stage with slow decline of evaporation rate, which resulted from the different volatilities of ACE and MeOH. Compared to the pure solvent systems, the drying kinetic profiles of PLGA solutions showed an extended drying time, which is because of the affinity between the solvent molecules and the PLGA molecules. However, no obvious extension of the drying time was observed with an increase in the amount of MeOH from 10% to 31% (molar ratio), which is likely because of the interplay between the reduced viscosity and the lowering of the volatility of the solution with an

\[ \text{Fig. 2. Estimation of the evaporation rates of the pure solvents (A) and the feed solutions (B). Results denote average values (n = 3). Reprinted from Pharm. Res., 30 (2013) 1065–1076, Wan, F., et al., Critical solvent properties affecting the particle formation process and characteristics of celecoxib-loaded PLGA microparticles via spray-drying. Copyright 2013, with permission from Springer.} \]
increase in the content of methanol. In addition, it should be noted that the evaporation profiles of the PLGA solutions containing 25% and 31% (molar ratio) MeOH exhibited two-phase decrease in the evaporation rate at the initial stage (Fig. 2B). It may be attributed to the anti-solvent precipitation of the PLGA during the drying process.

Hence, the particle formation process in the binary solvent system composed of ACE and MeOH can be speculated to be as follows (illustrated in Fig. 3): the molar ratio between ACE and MeOH in the solution would keep decreasing upon evaporation due to the lower evaporation rate of MeOH than that of ACE, which would lead to the anti-solvent precipitation of PLGA in a shorter time once the MeOH content (poor solvent for PLGA, Table 7) in solution reaches a certain point (in the case of ACE:MeOH = 69:31 and 75:25 (molar ratio); Fig. 3 upper row). PLGA precipitation would further form the shell around the droplets and thicken/shrink toward the core of droplet upon further evaporation, whereas the drug would migrate to the surface along with the evaporation of residual solvent because of its good solubility in both ACE and MeOH, eventually determining the drug distribution in the resulting microparticles.

4.2. Effect of solvent properties on the characteristics of PLGA microparticles

The effect of solvent on the characteristics of spray-dried particles (such as particle size and distribution, porosity, morphology, physical stability, drug release behavior, etc.) has been widely reported (Rain et al., 1999; Bohr et al., 2014; Paudel and Mooter, 2012; Rabbanil Seville, 2005; Wu et al., 2011; Wulsten et al., 2009; Zhou et al., 2001). Indeed, the solvents affect the characteristics of spray-dried particles mainly through changing the properties of feed solutions and the drying kinetics, which depend on the critical solvent properties (e.g. solvent power, solvent volatility, etc.). For systems composed of a single solvent, the aforementioned mechanical models can be used to predict the characteristics of spray-dried particles (e.g. size, density, surface chemical composition, etc.) (Vehring, 2008; Vehring et al., 2007). In contrast, when multiple solvents (usually binary solvents mixture) are applied, the droplets drying and particle formation process becomes much more complex because, as discussed in section 4.1, the distinct evaporation rates of the solvents, interplaying with the different solvent-solute interactions, may lead to a different precipitation process of the solutes. For example, in a study of levitated single-droplet drying of PVPcoVA/itraconazole (Wulsten et al., 2009), the particle surface changed from smooth to rough (more structured) when increasing the fraction of EtOH, which may result from changing the precipitation process of itraconazole. Furthermore, it was found that drug-polymer miscibility was mainly affected by solvent power (both to polymer and drug) and the evaporation rate of the feed solution. If the interplay between solvent power (both to polymer and drug) and the evaporation rate of the feed solution enhances the drug-polymer interaction, it contributes to a favorably drug-polymer miscibility and vice versa (Paudel and Mooter, 2012).

In our previous study, it was observed that the particle size was decreased with an increase in the proportion of poor solvent in the solvent system (Wan et al., 2013a), which can be attributed to the fact that the decrease in the viscosity of the PLGA solution resulted in a smaller droplet size. Furthermore, the decrease in drying rate of the co-solvent systems with an increase in the proportion of less-volatile component (in this case MeOH) led to a prolonged time for solidification and shrinkage of microspheres, eventually resulting in a smaller particle size with a compact inner structure (Vehring et al., 2007). In addition, a high drug surface enrichment was observed with an increase in the proportion of poor solvent in the solvent system. This is because that PLGA precipitates and forms a shell in a shorter time owing to the anti-solvent effect (Fig. 3), whereas the migration of the drug to the surface along with the evaporation of residual solvent lead to a higher drug surface enrichment. As a result, spray-dried PLGA microparticles prepared from a poor solvent with a low volatility exhibited severe burst release compared to those prepared from a good solvent with high volatility. In Fig. 4, a comparison between the drug surface enrichment and drug initial burst release of spray-dried PLGA microparticles produced using different solvent systems is illustrated. As we can see, the initial burst release is not correlated to the drug surface enrichment. One possibility could be that the distinct PLGA molecular behaviors in the feed solutions would result in a different connectivity and diffusivity of the spray-dried polymer matrix, eventually influencing the drug release behavior (Wan et al., 2013c).

It is well known that the solvent power has a great impact on the molecular conformation of the polymer, for example, in a good solvent, the polymer molecules tend to swell and expand, whereas they will adopt a compact conformation in a poor solvent.

![Fig. 3. Schematic description of PLGA precipitation processes in different solvent systems (adapted from (Wan et al., 2013a)).](image-url)
Table 7
Physical properties of solvents and PLGA.

<table>
<thead>
<tr>
<th>Solubility parameters&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Evaporate rate&lt;sup&gt;b&lt;/sup&gt; (BuAc = 1)</th>
<th>Boiling point (°C)</th>
<th>Interfacial tension&lt;sup&gt;c&lt;/sup&gt; (mN/m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetone</td>
<td>15.5</td>
<td>20.0</td>
<td>5.6</td>
</tr>
<tr>
<td>Methanol</td>
<td>15.1</td>
<td>22.3</td>
<td>4.1</td>
</tr>
<tr>
<td>Dichloromethane</td>
<td>18.2</td>
<td>20.3</td>
<td>27.5</td>
</tr>
<tr>
<td>Acetonitrile</td>
<td>15.3</td>
<td>6.1</td>
<td>2</td>
</tr>
<tr>
<td>PLGA (85:15)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>16.0</td>
<td>14.1</td>
<td>21.7</td>
</tr>
<tr>
<td>PLGA (75:25)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>16.1</td>
<td>11.7</td>
<td>22.1</td>
</tr>
<tr>
<td>PLGA (50:50)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>16.4</td>
<td>12.3</td>
<td>23.1</td>
</tr>
</tbody>
</table>

<sup>a</sup> Solubility parameters of organic solvent in MPa/12 at 25°C from Hansen Solubility Parameters: A User’s Handbook, 2nd ed.
<sup>b</sup> Relative to the evaporation rate of Butyl Acetate (BuAc = 1).
<sup>c</sup> Data generated by 2.5% (w/v) PLGA solution (Wan et al., 2014b).
<sup>d</sup> Solubility parameters of PLGA were calculated according to the L.G ratio (Schenderlein et al., 2004).

(Baldursdóttir et al., 2003; Elias, 1997). Based on the analysis of the polymer behavior in the feed solutions, combined with the drying kinetics of the binary solvent system discussed above, it is reasonable to speculate that in the case that ACE:MeOH = 69:31 (molar ratio) is used, PLGA molecules become increasingly more compact with an increase of the MeOH content in the droplets, which eventually results in the solidification of PLGA in the form of ‘aggregates’ which lack polymer chain entanglements (Fig. 5, upper row); in contrast, when a good solvent system is applied, PLGA molecules may form more tapped chain entanglements in the droplet with increasing PLGA concentration on the surface of the droplets with evaporation of solvent(s), which would result in a tight network among PLGA molecules (Fig. 5, bottom row). This is supported by $^1H$ → $^{13}C$ cross-polarization magic-angle spinning (CP/MAS) solid state NMR experiments (To be published).

The effect of solvent power on the connectivity and the diffusivity of the spray-dried PLGA matrix has also been shown in another study. As seen from Fig. 6, the burst release of lysozyme from the PLGA microparticles formulations prepared using ACE (M50F10ACE, herein M50 means the mass ratio of PLGA to lysozyme is 50:1; F10 means the feeding rate ratio of outer fluid to inner fluid is 10:1; ACE means the organic solvent is acetone) and DCM (M50F4DCM) deviated, though both of them exhibited a similar lysozyme surface enrichment and particle size. As DCM is a better solvent for PLGA as compared to ACE, the connectivity and the diffusivity of the PLGA matrices prepared by using DCM are much denser and have a better ability to resist the collapse of the PLGA matrices during the release process. A similar effect of solvent power on the microparticle characteristics was also observed in an electrospraying process for the preparation of PLGA microparticles (Bohr et al., 2015).

5. Novel nozzle design for microencapsulation in the spray drying process

The emergence of novel derivatives of the traditional spray drying process is driven by the combined need for effective, scalable microencapsulation methods for protein based drugs and the continuous manufacturing complied with the concept of quality by design (QbD). As the apparent limitation of conventional spray drying with a 2-fluid nozzle for encapsulating hydrophilic macromolecules into hydrophobic polymer microparticles due to their inverse solubility, many attempts have been made to prepare protein-loaded PLGA microparticles by spray drying of emulsions (w/o) or nanoparticle suspension (s/o) (Cleland and Jones, 1996; Jensen et al., 2010; Shi and Hickey, 2010; Yang et al., 2012b). Additionally, Mok et al. (2008) utilized PEG to improve the protein solubility in organic solvent via non-covalent interactions between proteins and PEG. Subsequently, the protein-loaded PLGA microparticles were prepared by spray drying a PLGA methylene chloride.

![Comparison of drug surface enrichment and drug initial burst release (in 15 min.) from spray-dried PLGA microparticles with different solvent systems](adapted from Wan et al., 2013a))
solution containing the PEG-protein complexes. Although the aforementioned approaches have shown promising results for microencapsulation of protein drugs within PLGA microparticles, they also increase the risk of protein denaturation and make the production process much more complex, which may result in the commercial failure of the products (e.g., Nutropin Depot) (Wu and Jin, 2008). In addition, from an industrial production point of view, the parenteral or inhalable microparticles formulations are in general manufactured by an aseptic process in order to assure sterility of products (Kumar and Palmieri, 2010), however, the multiple steps increase the difficulty and the cost of the sterilization of the products.

Huge efforts have also been made to overcome the complexity of the manufacturing process. Recently, a relatively simple microencapsulation method based on the phenomenon of polymer self-healing was developed in Steven P. Schwendeman’s group (Desai and Schwendeman, 2013). For further information on the active self-healing encapsulation method, the interested reader is referred to a recent review article which discusses the approach in detail (Schwendeman et al., 2014). In the current paper, we will discuss the efforts to modify the conventional spray drying process using newly-designed atomizers (nozzles). A common characteristic of some newly-designed atomizers (nozzles) is the presence of two separate channels for liquid feeding, thereby providing the possibility to eliminate the complicated pre-preparation process (such as preparation of nanosuspensions, nano-scale complexes, w/o/w emulsion etc.) and enable continuous manufacturing. However, the newly-designed construct is not the ‘universal key’: the critical factors/parameters have to be identified and optimized to ensure the quality of the final product (e.g., microencapsulation efficiency).

Fig. 5. Schematic description of the impact of solvent properties on the PLGA molecular conformation in spray-dried PLGA microparticles (adapted from Wan et al., 2013a,c)).

5.1. Co-axial ultrasonic atomizer

The co-axial ultrasonic atomizer (Sono-Tek, Milton, NY, USA) was investigated to generate the reservoir-type microcapsules based on the interfacial solvent exchange method (Yeo et al., 2003; Yeo et al., 2004; Yeo and Park, 2004). In this method, reservoir-type microcapsules can be generated via: (i) midair collision between the drops of individual liquids fed from two separate channels, followed by encapsulation of the aqueous drops by the polymer drops due to the surface tension gradient existing between the two liquid drops; (ii) the formation of a 'transient emulsion' prior to atomization on the atomizing surface by absorption of the underlying vibration energy (Yeo and Park, 2004).

The quality of the polymer film formed on the aqueous surface, which determines the microencapsulation efficiency, highly depends on the spread of the polymer organic solution on the aqueous surface and subsequent phase separation of the water insoluble polymer (Yeo et al., 2004). Therefore, careful selection of a proper solvent is highly important. First of all, the interfacial tension of the organic solvent must be low enough to make the polymer solution spread easily over the aqueous surface. In addition, in order to cause the instant phase separation of polymer film, the organic solvents should be miscible with water to a certain degree (Yeo et al., 2003, 2004). For example, ethyl acetate, with 8.0% (w/w) of water solubility and 23.8 dyn/cm of surface tension, was found to be one of the optimal solvents for PLGA (lactic acid/glycolic acid = 50:50, intrinsic viscosity = 0.58 dl/g, Birmingham Polymers) (Yeo et al., 2003).

In addition to the physical properties of the organic solvent, the flow rate ratio of aqueous solution to polymer solution also plays an important role in determining the microencapsulation efficiency (Fig. 7). For example, the microencapsulation efficiency was observed to decrease from 53.95% to 26.8% as the flow rate ratio of aqueous solution to polymer solution increased from (0.125 ml/min)/(3 ml/min) to (0.5 ml/min)/(3 ml/min) (Yeo and Park, 2004). This is because that the higher flow rate ratio of aqueous solution to polymer solution tends to form an oil-in-water transient emulsion instead of a water-in-oil emulsion, preventing the internalization of the aqueous phase into the polymeric organic phase.

Furthermore, the concentration of the polymer organic solution, the molecular weight and the monomer ratio also have a potential impact on the microencapsulation efficiency, though it has not yet been experimentally observed. Because, in theory, the viscosity of the polymer solution is influenced by these factors, which will eventually affect the spreading capability of the polymer organic solution on the aqueous phase.

5.2. Co-axial electrospray

Electrospray is another liquid atomization-based particle generation process (Peltonen et al., 2010). In electrospray, the...
liquid feed is atomized via the electrical shear stress generated by applying a high voltage of several kilovolts to the microcapillary nozzle. It can usually be operated in several modes, but the most widely used mode is the ‘cone-jet’ mode (also referred as Taylor-cone, the electrostatic force is equal to the surface tension of the liquid) because it is stable and can produce mono-dispersed droplets. Once the Taylor cone is formed at the tip of the capillary nozzle, the liquid feed is broken into small droplets upon increasing the charge. The highly charged droplets travel along the electric field towards a counter-electrode, which results in particle formation as the solvents evaporate. The variety of process parameters, such as conductivity, flow rate, surface tension, viscosity, as so on, affects the physicochemical properties of the resulting particles. For more detailed information about the production of fine particles via electrospay technique, the reader is referred to several review articles (Jaworek, 2007; Jaworek and Sobczyk, 2008; Peltonen et al., 2010).

Loscertales et al. were the first to demonstrate the production of microcapsule droplets via co-axial electrospay (Loscertales et al., 2002). They observed that the size of the microcapsule droplets is influenced by the process parameters (such as feeding liquid flow rates), the physical properties of the liquid feeds and the interaction between the inner and outer liquids during the spray. In another study, Mei and Chen further identified the criteria that predict the formation of core-shell structured droplets: (i) the ratio of the charge relaxation lengths of the inner and outer jets (r) is less than 500, (ii) the ratio of the inertial breakup lengths of the inner and outer jets is less than 0.015 (Mei and Chen, 2007). To date, co-axial electrospray has shown attractive advantages in the fabrication of protein-loaded biodegradable polymer microparticles (e.g. relatively high encapsulation effectiveness, near mono-disperse particle size distribution, etc.) (Lee et al., 2010; Xie et al., 2008; Xie and Wang, 2007), though industrial-scale processing has not yet been reached. However, in a recent study, compact multiplexed electrospray (with a high number of sources per unit area) has been developed, which can dramatically increase the throughput and reduce the cost per electrospray source (Deng et al., 2009).

5.3. Three-fluid nozzle

The three-fluid nozzle possesses two channels for liquid feed and one channel for atomization gas (Wan et al., 2013b), which makes it versatile for various solvents. Spray drying with a 3-fluid nozzle has been used to investigate the microencapsulation of fish oil, small molecular drugs and nanoparticles (Kondo et al., 2014; Legako and Dunford, 2010; Pabari et al., 2012; Tokárová et al., 2013). In our recent studies, we investigated the potential of using a spray dryer equipped with a 3-fluid nozzle to microencapsulate protein drugs (lysozyme and bovine serum albumin (BSA)) into PLGA microparticles in a one-step process (Wan et al., 2014a; Wan et al., 2014b). Our studies indicated that the microencapsulation efficiency is significantly influenced by the organic solvents used in the outer fluid, the feeding rate ratio of outer fluid to inner fluid and the viscosity of the inner feed solution. To better understand the roles of these factors in the microencapsulation process, it is necessary to elaborate on the droplet formation and drying process.

Basically, a 3-fluid nozzle is a pneumatic nozzle like a 2-fluid nozzle. The atomization mechanism of a pneumatic nozzle is that high frictional force over liquid surfaces created by high velocity gas causes disintegration of the liquid into spray droplets. Although the liquid disintegration process has not yet been fully understood, the entire process is likely to occur in two phases.
The complexity of the spray patterns produced via the pneumatic nozzles provides the possibility of encapsulating aqueous droplets into polymer organic droplets via ‘midair collision’ and ‘transient emulsion’ introduced above. Hence, the factors influencing the microencapsulation efficiency in the co-axial ultrasonic atomizer (e.g. interfacial tension of organic solvents, immiscibility of organic solvents with water, feeding rate ratio) also have an impact on the formation efficiency of coated droplets via a 3-fluid nozzle. Further, once the microcapsule droplets form and enter the drying chamber, the distinct drying rates of solvents, the composition of the entire solvent system and the migration of solutes will influence the distribution of solutes in the resulting spray-dried particles, eventually resulting in distinct microencapsulation efficiencies (Fig. 9).

For example, in our previous study, compared to acetoneitrile (ACN) and ACE, the microencapsulation efficiency of lysozyme significantly increased when DCM was used as the organic solvent (Wan et al., 2014b). The explanation should be sought in its lower interfacial tension and immiscibility with water. Additionally, DCM evaporates faster, a PLGA shell forms and gets thicker and denser in less time, which may hinder migration of lysozyme molecules across the PLGA shell to the droplet surface with the evaporation of water, eventually decreasing the surface enrichment of lysozyme (Vehring et al., 2007; Wan et al., 2013a).

The impact of the diffusion of proteins inside a microcapsule droplet upon drying of a binary solvent system can be understood from another study (Wan et al., 2014a). In the study, hyaluronic acid (HA), a hydrophilic polymer with high viscosity, was incorporated into the inner feed solution (aqueous protein solution) to suppress the diffusion of BSA upon drying. We found that the spatial distribution of BSA in the PLGA matrix, confirmed qualitatively and quantitatively by using confocal laser scanning microscope (CLSM) and X-ray photon spectroscopy (XPS), was changed from a surface-enriched pattern to a core-enriched pattern when HA was introduced into the PLGA matrix, irrespective of the organic solvents applied. This is because the addition of HA into the inner feed solution, without any influence on the surface tension, dramatically increased the viscosity of aqueous phase and resulted in the entanglements formed among polymer molecules in the drying process, thereby hindering the migration of BSA from the inner phase to the outer phase (Surendrakumar et al., 2003).

6. Conclusion

Spray drying, a scalable, one-step production process, has shown the prospect in simplifying the manufacturing process of proteins-loaded PLGA microparticles when equipped with novel atomizers. The novel atomizers (nozzles) have the ability to eliminate the complicated pre-preparation process (such as preparation of nanosuspensions, nano-scale complexes, etc.) and enable continuous manufacturing by feeding the liquid feeds via two separate channels. Nevertheless, the product quality is also highly influenced by the formulation, process parameters and solvent compositions. Especially solvent constitutes a major part of the feed in the spray drying process. The microparticle formation process and the final microparticle characteristics can be tuned via a rational selection of solvent compositions to adjust the drying kinetics of a feed and the precipitation rate of solute(s).

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References


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