Manufacture of ultrafine drug particles via the Rapid Expansion of Supercritical Solution (RESS) Process using Taguchi approach

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Abstract

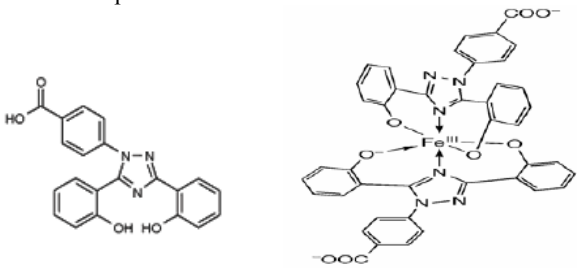
The poor water solubility of many drugs is a challenge in pharmaceutical research. Recently, there have been great interests in finding environmentally freindly methods producing fine particles of pharmaceutical products for applications in pharmaceutical engineering. A promising method to improve the bioavailability of pharmaceutical agents is the Rapid Expansion of Supercritical Solutions. Deferasirox (DFS), a tridentate chelator, requires two molecules for iron (III) coordination. The bioavailability (the percentage of the drug absorbed compared to its initial dosage) is limited by this insolubility. . The RESS experiments were carried out in a broad temperature range (35-55 ^{o}C) and pressures at 150–200 bar. The effects of extraction pressure, extraction temperature, nozzle diameter, and spraying distance based on Taguchi design on the shape and size of the particles formed are discussed. Our results show that extraction pressure and extraction temperature can significantly affect the morphology and size of the precipitated particles. For the Taguchi design and subsequent analysis, the software named as Qualitek-4 was used.

Key Words: Nanoparticles, Iron Chelator, Deferasirox, Rapid Expansion of Supercritical Solution, Taguchi Method

1 Introduction

In recent years, significant effort has been devoted to develop drug formulation and delivery systems for issues such as targeting and controlled release [1–3]. In fact, the solubility is a serious limitation in drug development and related requirements for bioavailability and normal absorption pattern. According to the established statistics, about a third of the drugs listed in the United States Pharmacopeia are poorly water-soluble or insoluble and more than 40% of new drug development has failed because of poor biopharmaceutical properties [4-5]. The nanosizing of drug particles has been identified as a potentially effective and broadly applicable approach.For example, smaller-diameter particles correspond to a faster dissolution rate, thus potentially higher activity and easier absorption. Other distinct advantages include tissue or cell specific targeting of drugs, longer circulating capacity in the blood, higher stability against enzymatic degradation, and the reduction of unwanted side effects [6-7]. Several conventional techniques are used for reduction of particle size such as crushing, grinding, milling, spray drying, freeze-drying and recrystallization of the solute particles from solutions using liquid antisolvents. But all these techniques have got several disadvantages. Some substances are unstable under milling conditions, and in recrystallization process the product is contaminated with solvent. In addition, there are thermal and chemical degradation of products due to high temperatures, high-energy requirements, large amount of solvent use, solvent-disposal problems, and broad particle size distributions. Due to their several drawbacks, the use of supercritical fluids has increased rapidly over the last few years and several processes for particle formation have been studied [8].Supercritical fluid processing techniques have been applied to the particle formation in drug formulation [9-10]. The methods of fine particles formation using supercritical fluids are: rapid expansion of supercritical solutions (RESS), anti-solvent processes (gas anti-solvent (GAS), supercritical anti-solvent (SAS), aerosol solvent extraction system (ASES), solution enhanced dispersion by supercritical fluids (SEDS)) and particles from gas saturated solutions/suspensions (PGSS) [11]. The RESS process consists of extraction and precipitation unit. A substance is solubilised in a supercritical fluid (SCF) at the extraction unit, than the supercritical solution is suddenly depressurised in a nozzle causing fast nucleation and fine particle formation. Due to the rapid expansion of supercritical solution through a nozzle, the large decrease in density, and hence decreasing the SCF solvating power. The solute becomes supersaturated and then precipitaded. The driving force of the nucleation process is supersaturation. Higher supersaturation leads to increase the nucleation rate and tends to decrease the particle size. Advantages of RESS process are that nano or microparticles are produced, providing a solvent-free product and controllable particle size. The morphology and size distribution of the precipitated material is related to pre-expansion and expansion conditions, extraction parameters, spray distance and nozzle design [12]. Carbon dioxide is commonly used as a supercritical fluid because it is non-toxic, non-flammable, and cheap. It has a low critical temperature and pressure (T_c = 31.1 $^{\circ}$C and P_c = 73.8 bar) that allow for low temperature processing. From a pharmaceutical point of view, supercritical carbon dioxide has several advantages, including being solvent-free, and being able to be used in a single-stage process and at moderate processing temperatures. Hereditary hemochromatosis (HH) is an autosomal recessive disorder characterized by progressive iron overload through increased intestinal absorption. Phlebotomy, the preferred treatment, can prevent or reverse some complications of iron overload, such as hepatic damage; however, compliance is variable and some patients are poor candidates because of underlying medical disorders and/or poor venous access. Thus, if an oral iron chelator such as deferasirox (Exjade) proves to be tolerable and effective, HH patients will have an alternative treatment option. Iron-chelation therapy is essential in iron-overloaded patients.

Iron chelation is a key feature in the management of transfusion dependent anaemias— such as b-thalassaemia major, b-thalassaemia intermedia, sickle-cell disease and myelodysplastic syndrome—to prevent end-organ damage and improve survival. Exjade is supplied as a tablet that is dispersed in water or juice. Deferasirox (Exjade, ICL670, Novartis Oncology) belongs to a novel class of tridentate iron chelators, the N-substituted bis-hydroxyphenyl triazole. Two molecules of deferasirox are needed to form a soluble complex with one Fe3+ ion. The active is a white to slightly yellow and not hygroscopic powder. It has a good permeability and it is practically insoluble in water and in acid medium, the solubility increasing with pH. Therefore, the particle size is likely to be important to the rate and possibly to the extent of absorption [13-15]. Poor aqueous solubility represents a major hurdle in achieving adequate oral bioavailability for a large percentage of drug compounds in drug development nowadays. Nanosizing refers to the reduction of the active pharmaceutical ingredient (API) particle size down to the sub-micron range, with the final particle size typically being 100–200 nm. The reduction of particle size leads to a significant increase in the dissolution rate of the API, which in turn can lead to substantial increases in bioavailability [16]. Several authors have reviewed the applications of RESS on the preparation of fine and ultra fine particles. Formations of anthracene fine particles by Nagahama et al [17].Krober et al in 2000 have reported an investigation of RESS for synthesis of small organic particle [18]. To our knowledge, no research paper is available on micronization of deferasirox by supercritical carbon dioxide. The experiments were carried out to investigate the effect of extraction temperature (308–328 K) and pressure (15–20MPa), spray distance (1-5 cm), nozzle diameter (500-1200 μm) on the size and morphology of the precipitated deferasirox particles.



(a) (b)

Fig.1.(a)Molecular structure of Deferasirox(b)Function of Deferasirox

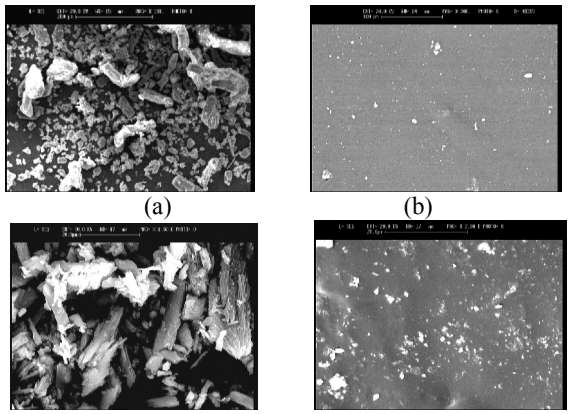
2 Experimental

2.1. Materials

The solute used during this study, Deferasirox, was prepared from Pharmaceutical Arasto company and Carbon dioxide (99.9 %< purity) was purchased from Abughadareh Gas Chemical Company.

2.2. Particle characterization

Precipitated deferasirox particles were analyzed their size by scanning electron microscopy (SEM) (S360-CAMBRIDGE).Before the SEM analysis, the process either oroginal samples must be coated by a sputter-coater (SC-7640-Polaron) with Pd-Pt under the presence of argon (99.9% < purity) at the room temperature for a period of 100 s under an accelerating voltage of 20 KV. The SEM images of original and precipitated deferasirox are given in Fig.2.



(a) (b)

(c) (d)

Fig.2.The SEM images of (a,c) Unprocessed Deferasirox Particles(b)Processed Deferasirox

2.3. Experimental technique

The RESS pilot plant is shown in Fig. 3. At first, the gaseous CO2 from a cylinder capsule was passed through a filter and then entered into a refrigerator to make liquid CO2.The liquid CO2 was then pumped by a reciprocating high pressure pump into a surge tank The surge tank dampened the pressure fluctuations produced by operation of the pump. at the outlet of the surge tank a bourdon gauge in the range of 0-250 bar was placed. The pressurized CO2 then entered into an extraction vessel. It should mention that the surge tank and the extraction vessel are surrounded by a regulating hot water jacket. The basket which is packed by sample and glass wool was placed into an extraction vessel. For each condition the extractor vessel was held for 2 hours to ensure equilibrium has been obtained. The equilibrated solution was then expanded by a preheated fine needle valve into a nozzle. The precipitated deferasirox particles were collected on the stub and analyzed by a SEM to monitor the particle size and its morphology. A new type of nozzle(Fig.4) were designed and fabricated to achieve ultrafine nanosize particles.

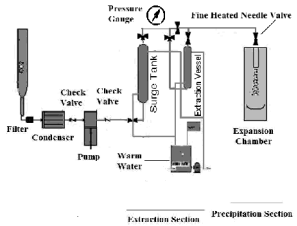


Fig3. The schematic diagram of the experimental apparatus for the RESS process

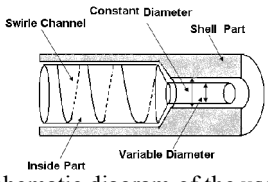


Fig.4. Schematic diagram of the used nozzle

3 Results and discussions

In this work, the influence of RESS parameters such as extraction temperature (308-328 K), extraction pressure (150-200 bar), spraying distance (1-5cm) and effective nozzle diameter (500-1200 µm) were investigated on the mean particle size of the micronized deferasirox particles which are expressed below. In addition, the Taguchi method was used to arrange the experimental conditions of micronization of deferasirox particles based on L-9 array as shown in Table 1. Anova table for deferasirox based on Qualitek-4 software is drafted below (Table2).The measurements of average particle size of deferasirox are done by SigmaScan Pro Image analysis.

Run number	P	T	SD	ND	Average Size(µm)
1	150	35	1	500	0.47
2	150	45	3	900	0.69
3	150	55	5	1200	1.64
4	175	35	3	1200	0.82
5	175	45	5	500	0.40
6	175	55	1	900	0.94
7	200	35	5	900	1.40
8	200	45	1	1200	1.00
9	200	55	3	500	1.25

Table1. Taguchi L-9 array

For Taguchi design the statistical software namely Qualitek-4 was applied. (fig5). (Dark Blue=T, Green=P, Blue=ND, Red=SD).

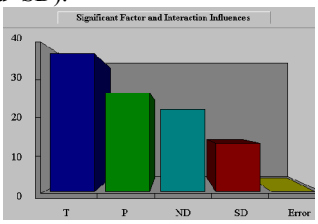


Fig.5.Influence of paparameters

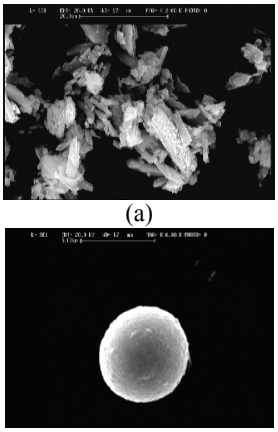
Factor	DOF	Sum of Square	Variance	Percent (%)
P	2	.372	.186	26.829
T	2	.52	.26	37.513
SD	2	.182	.091	13.171
ND	2	.312	.156	22.478
ERROR	-	-	-	-
TOTAL	8	1.388		100%

Table2.Anova Table for Deferasirox

The results of analysis show that the optimum pressure, temperature, nozzle diameter and spraying distance are 175 bar, 318 K , 500 µm and 1 cm, respectively.

3.1 Effect of extraction pressure

Generally, the variation of the extraction pressure brings about a change in the concentration of Deferasirox.. The pressure studied here varies from 150 to 200 bar. An increase in extraction pressure from 150 bar to 175 is observed to induce decrease in the average particle size. Similar results were reported by A.Z. Hezave, F. Esmaeilzadeh [19]. The reason for this is that an increase in the solute solubility results in higher supersaturations in the fluid upon expansion. According to the classical theory of nucleation, higher supersaturation causes higher nucleation rate and the particle volume is inversely proportional to the nucleation rate, our above results appear to agree with simple theoretical predictions. Similar results have also been reported for other organic solutes. But the finest average particle size and the smallest particle size distribution are observed at 175bar.In other words a further increase of extraction pressure till 200 bar yields a marked increase of the particle size, which may suggest a decoupling of the two processes of nucleation and growth. Perhaps at high deferasirox concentrations (i.e., high extraction pressure), the particle growth may be dominant or a particle may include several nuclei during the growth process. Therefore large particles seem to be readily produced and a broad particle size distribution may be obtained. Similar results have been reported for formation of aspirin by Z.Huang et al [20].As can be seen in Fig.6. , the morphology of the original particles was changed from irregular shape to spherical state at high pressure.



(a)

(b)

Fig.6.The SEM images of (a) original deferasirox (b) Processed deferasirox

3.2 Effect of extraction temperature

The extraction temperature for deferasirox in this study ranges from 308 to 328 °K. increasing the extraction temperature leads to decrease in the density of CO2 and concurrent increase in the solute's vapour pressure. The decrease of the solvent density causes a decrease of the solvent strength. On the other hand, a concurrent increases in the solute's vapour pressure leads to an increase in the deferasirox solubility. An increase of extraction temperature from 308 to 318K leads to the increase of the supersaturation and nucleation rate as a result of increased solute concentration. The increase of nucleation rate leads to decrease in the deferasirox particle growth time, and consequently the smaller particle was obtained. However, continuously increasing temperature (at 328 K) caused to increase of the average particle size. Because increasing the temperature leads to increase the deferasirox concentration. During the expansion, high deferasirox concentration brings about the increase of the particle size, as a consequence of coagulation among particles. In the literature, similar results have also been reported in the literature [21].

3.3 Effect of nozzle diameter

It was expected that the nozzle diameter and its dimension in the RESS process highly affected the particle formation. In this study, the effect of the effective nozzle diameter was investigated (500-1200 μm). The results of the experiments show the average particles sizes can be reduced by lowering the diameter of nozzle. In other words, the particle size increases with increasing the nozzle diameter. Several studies have been reported that the change of the nozzle diameter can play an important role for processing materials [22].

3.4 Effect of spraying distance

The spraying distance can have a pronounced effect on the characterization of the particles, since the nucleation and growth process continues in post-expansion region. In this study, three spraying distances (1, 3, 5 cm) from the tip of the nozzle were tested. The results show only a little change in the spraying distance from 1 cm to 5cm causes an increase in the average particle size. Similar results were reported by Yildiz et al [23].

4 Conclusions

The rapid expansion of supercritical solutions (RESS) was successfully used to produce deferasirox submicron particles. Deferasirox was micronized and a great size reduction of deferasirox particles in comparison with the original one was observed. The obtained results show a good performance of these used nozzles in comparison with the capillary nozzle used. We conclude the extraction temperature and extraction pressure has a main role in the particle size and the morphology of deferasirox particles. However by increasing the nozzle diameter and spraying distance, the particle size increases. In addition, the optimum Pressure, temperature, nozzle diameter and spraying distance for micronization of deferasirox particles are 175 bar, 318 K , 500 μm and 1 cm, respectively.

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