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Research paper

New spray congealing atomizer for the microencapsulation of highly concentrated solid and liquid substances

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Abstract

A new pneumatic atomizer for spray congealing, called wide pneumatic nozzle (WPN), was developed. To evaluate its performance, microparticles containing highly concentrated either solid drug (Propafenone hydrochloride, PRF) or liquid nutraceutical (Vitamin E, VE) have been prepared and characterized. The results showed that the spray congealing nozzle enabled the production of spherical and not aggregated microparticles with high yields (95% w/w) and relatively narrow size distributions; moreover, increasing the viscosity of the suspension from 50 to 500 mPa s, the particle size increased. The loading of the drug was high for microspheres (50% for PRF and 30% for VE) and the encapsulation efficiency was good for all formulations. The drug release was easily modified according to the nature of the used excipients, as both lipophilic (carnauba wax, cetearyl and stearyl alcohols) and hydrophilic (PEG 4000) carriers were employed. Moreover the results evidenced that it was possible to encapsulate actives (VE) that are in a liquid form and to enhance their availability. In conclusion the developed spray congealing nozzle was able to nebulize very viscous systems that are usually not processed by conventional apparatus and to produce microspheres with high and uniform drug content.
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Keywords: Spray congealing; Air pressure nozzle; Microparticles; Propafenone hydrochloride; Vitamin E acetate

1. Introduction

In recent years, microencapsulation has been used in several fields such as pharmaceutical, veterinary, cosmetic and agricultural as well as in food and food additives [1]. The most popular technologies used to prepare microparticles are coacervation, solvent evaporation, fluid bed coating, extrusion coating, spray drying and spray congealing [2]. Actually, the spray congealing technique (alternatively called spray chilling or spray cooling) is gaining considerable attention, especially from the view point of safety and of rapidity. In fact the spray congealing technique atomizing a solution or a dispersion of the drug into a molten carrier, which melts at a relatively low temperature (45–75 °C), overcomes the problem of residual solvents and the atomized droplets quickly solidify due to their exposure to an ambient air flow. Recent developments in spray congealing apparatus include the use of congealing chambers with integrated bag filters for cooling and separation of product in one unit. This design is very compact and thus space-saving [3]. In the last decade, several papers focused the attention on the possibility of producing solid lipid microparticles (SLMs) by spray congealing [4] to form sustained-release matrices [5–8] and to mask the bitter taste of some antibiotics [9–11]. Recently a spray congealing process for the preparation of protein loaded SLM was developed, showing that this technology is also a promising approach for the microencapsulation of proteins sensible to degradation, as for instance insulin [12].

The performance of the spray congealing process strictly depends on the atomization efficiency of the molten mixture, which may be sprayed using different types of devices, traditionally divided into rotary (or centrifugal) atomizers,

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airless nozzles and air (or two-fluid) nozzles [13]. In centrifugal atomizers, the molten mixture is dropped onto a high-speed rotating disc; the rotation spreads and sprays the fluid and the droplets solidify in a cool air stream. Rotary spray congealing has the advantages of being amenable to a wide variety of feed rates and viscosities, yielding a relatively homogeneous particle size distribution [14]. However, the spray pattern is wide and short, so requiring wide cooling chambers [5,14,15]. Airless atomizers are only seldom employed for spray congealing because to atomize viscous fluids they require very high pressures (70 bar) and high flow rates, and consequently they yield inhomogeneous droplet size distributions [13]. On the contrary air atomization is widely applied to the process, even though the atomization air must be heated to avoid excessively rapid solidification of the mixture [13]. Usually pneumatic nozzles atomize the melted mixture into a carbon-dioxide ice bath for the cooling and the obtained microparticles have to be dried for several hours [6,7,11,12]. The equipment aided with a pneumatic spraying system is narrower, longer and overall smaller than that employed with rotary atomizers. In previous works [16–21], we have proposed the use of a new atomizer for the spray congealing process; this apparatus employs ultrasonic energy (US) rather than centrifugal, pressure or kinetic energy used in traditional atomizers to nebulize the liquid and the absence of an orifice avoids obstructions. The results showed that the spray congealing method assisted by ultrasounds easily yields spherical microparticles with a good encapsulation efficiency and size distribution. The cooling of the droplets is quick due to the slow fall at room temperature, without requiring a subsequent drying step. Moreover, by selecting the lipophilic/hydrophilic character of the carrier, the microspheres can either control the release of short half-life drugs as verapamil hydrochloride [17] and theophylline [18] or enhance the dissolution rate of poorly soluble drug as carbamazepine [19], diclofenac [20] and praziquantel [21]. However, besides the above mentioned advantages, all available atomizers used for spray congealing could present some common drawbacks as the difficulty to atomize highly viscous fluids and the difficulty to achieve a drug loading exceeding 20% w/w. Generally, these sprays led to a broad size distribution (50–600 μm) with a prevalence of small or large microparticles as a function of the viscosity of the molten mass.

The main purpose of this research was to propose a novel technological solution to the tasks of atomizing highly viscous systems for spray congealing with high yields and a relatively narrow size distribution. A new spray congealing nozzle was developed and described. To show the possibilities of the new device, microparticles containing 50% w/w of a solid drug (Propafenone hydrochloride) were investigated. Secondly, the study aimed to evaluate the ability of the atomizer to encapsulate a high amount of a liquid nutraceutical (Vitamin E). The effect of the spray process on the microparticle characteristics (morphology, particle size, flowability, drug loading and release profiles) was evaluated. Furthermore, the physicochemical properties of the microspheres were examined using DSC.

2. Materials and methods

2.1. Materials

Propafenone hydrochloride (PRF) was kindly provided by Procos S.p.a. (NO, Italy). Cetearyl alcohol (an aliphatic alcohol consisting mainly of stearyl and cetyl alcohols, HLB 4.7, conforming to EP 5) was used as carrier for the preparation of PRF-loaded SLM and it was purchased from ACEF Spa (Piacenza, Italy), while different silicon dioxides (Aerosil® R812 and R972), kindly supplied by Degussa, and soya lecithin (Carlo Erba Reagenti, Italy) were used as thickening and suspending agents.

Vitamin E (α-tocopheryl acetate) (VE) was supplied by DMS Nutritional Products (Basel, Switzerland). Both lipids as stearyl alcohol and carnauba wax (Carlo Erba Reagenti, Italy) and hydrophilic low melting point polymers as PEG 4000 (Carlo Erba Reagenti, Italy) were used to prepare VE-loaded microparticles. Different silicon dioxides (Aerosil® 90 and R812) were also employed as release modifiers to enhance the availability of Vitamin E acetate.

Milli-RX20 water (Millipore, Molsheim, France) was used throughout.

2.2. Methods

2.2.1. Description of the atomizer

The new pneumatic nozzle has been identified with the acronym WPN (Wide Pneumatic Nozzle) because of the wide (4.5 mm) orifice opening (the inner diameter is usually around 1 mm). The scheme is shown in Fig. 1. WPN is a two-fluid atomizer which acts in an unusual configuration: the air enters in radial direction, while the fluid enters axially and it proceeds along a rectilinear path. The air cross the fluid with an inclination of 45° and then the mixture is sprayed. In practice, the atomizer is a particular kind of venturimeter that takes advantage of the Venturi effect.

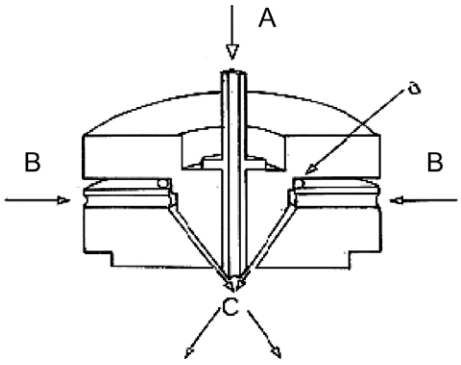

Fig. 1. Scheme of the WPN (not in scale). (A) Material aspiration (feeding), (B) air inlet, (C) atomization of the fluid, (a) o-ring.

This phenomenon takes place when a fluid (the melted mixture) is flowing through a pipe (called Venturi tube) and it is forced through a narrowing. This constriction causes a drop in pressure in the fluid flowing in the pipe; consequently, the fluid is sucked from the top of the tube (point A) towards the nozzle. At the same time the fluid velocity increases due to the conservation of energy: the gain in kinetic energy is supplied by the drop in pressure (described by Bernoulli's equation). The fluid is delivered from a thermostated reservoir, placed above the point A, to the nozzle by both the force of gravity and the effect of Venturi. The inlet air flux (B) is obtained by three holes placed one from the other at 120°; when the air comes in contact with the fluid, the atomization (C) occurs. The WPN requires modest air consumption (1–3 bar depending on the desired particle size) and it generates a uniform spray pattern; the spray stream is delivered with a 90° angle which is symmetric to the nozzle cone. The microparticles are then collected at the bottom of a cooling chamber (1.80 m in height and 75 cm in diameter). In this device, the atomization air is not heated because of the presence of two resistors connected to an inverter which heats the nozzle. The output of the inverter was feed-back regulated through a suitable electric circuit by the signal of a thermocouple embedded into nozzle, thus keeping the temperature of the nozzle at the pre-set value ±1 °C (20 °C above the melting point of the carrier).

2.2.2. Preparation of PRF-loaded microparticles (A)

Microparticles with a theoretical drug loading of 50% w/w were produced by the spray congealing process using the WPN, with a 25 g batch for each formulation. Cetearyl alcohol was heated at a temperature of 10 °C above the melting point (about 60 °C); first Aerosil® or soya lecithin (when present) and then PRF were added to the molten carrier and stirred to obtain a suspension, which was then loaded into the feeding chamber of the WPN, kept at 80 °C to avoid the solidification of the suspension in the nozzle orifice. The inlet air pressure was set at 1.5 bar. Finally, the microparticles were collected and stored in PE closed bottles at 25 ± 2 °C. The composition of the different formulations is shown in Table 1.

2.2.3. Preparation of VE-loaded microparticles (B)

Microparticles with a theoretical VE loading of 30% w/w were produced by the spray congealing process using the WPN, with a 50 g batch for each formulation. The carriers were heated at a temperature of 10 °C above the melting point and then Aerosil® (when present) and VE were added to the molten carrier and stirred to obtain a solution/suspension, which was then loaded into the thermostated feeding chamber of the WPN (kept at 80–100 °C depending on the carrier melting point). The air pressure was set at 2.5 bar. Finally, the microparticles were collected and stored

Table 1
Composition of the formulations and drug loading for the different particle sizes of microparticles containing a theoretical PRF content of 50% (w/w)

Formulation	Composition (% w/w)				Drug loading (%) ± SD for the different particle size (μm) fractions				ANOVA *F*-test
	Cetearyl alcohol	Aerosil R 812	Aerosil R 972	Soya lecithin	$75 < x < 150$	$150 < x < 250$	$250 < x < 355$	$355 < x < 500$	$F(3,8)$
1A	50.0	–	–	–	41.9 ± 0.4	51.2 ± 0.7	53.9 ± 0.3	54.8 ± 0.8	300.44*
2A	49.5	0.5	–	–	47.3 ± 0.6	52.7 ± 0.5	53.3 ± 0.3	52.0 ± 0.6	86.65*
3A	49.0	1.0	–	–	48.3 ± 0.2	51.9 ± 0.6	50.4 ± 0.6	51.6 ± 0.7	23.49**
4A	48.0	2.0	–	–	48.6 ± 0.4	51.4 ± 0.3	51.7 ± 0.2	52.9 ± 0.4	85.07*
5A	48.0	–	2.0	–	50.1 ± 0.3	51.7 ± 0.5	52.2 ± 0.3	52.3 ± 0.2	25.08**
6A	48.0	–	–	2.0	48.9 ± 0.7	55.7 ± 0.6	54.2 ± 0.7	55.3 ± 0.5	79.98*

* $p < 0.00001$.
** $p < 0.0002$.

Table 2
Composition of the formulations and particle size distribution of microparticles containing a theoretical VE content of 30% (w/w)

Formulation	Composition (% w/w)					Particle size distribution w/w (%) (mean of two determinations)					EE%
	Carnauba wax	Stearyl alcohol	PEG 4000	Aerosil 90	Aerosil R 812	$75 < x < 150$	$150 < x < 250$	$250 < x < 355$	$355 < x < 500$	$x > 500$	
1B	70	–	–	–	–	3.45	87.93	5.17	3.45	–	100
2B	–	70	–	–	–	16.83	81.57	1.60	–	–	81.4
3B	–	69	–	1	–	15.00	82.70	2.30	–	–	81.2
4B	–	69	–	–	1	13.68	79.44	5.10	1.78	–	79.4
5B	–	–	70	–	–	–	6.45	9.68	50.00	33.33	97.5
6B	–	–	69	1	–	–	–	16.67	25.81	58.06	91.0
7B	–	–	69	–	1	–	–	11.11	44.44	44.45	97.8

in PE closed bottles at 25 ± 2 °C. The composition of the different formulations is shown in Table 2.

2.2.4. Determination of viscosity

The viscosity determination was performed on the formulations mentioned in Tables 1 and 2. The measurements were carried out on about 5 g of molten mass, prepared as described in the preparation of the microparticles and placed in the small sample adapter of the viscosimeter (Visco Star-R, Fungilab S.A., Barcelona, Spain), which was previously heated to the temperature set for the spray congealing process. After some preliminary tests, the measuring elements, spindle number TR8 and TR9, were selected and the spindle rotating speed was stated at 200 rpm; the results are expressed as mPa∗s.

2.2.5. Particle size analysis

The size distribution of microparticles (A and B) was evaluated by sieve analysis, using a vibrating shaker (Octagon Digital, Endecotts, London, UK) and 5 standard sieves (Scientific Instruments s.r.l., Milano, Italy) of 75, 150, 250, 355 and 500 μm.

2.2.6. Morphological analysis

The morphological characteristics of microparticles were observed by Scanning Electron Microscopy. The samples were sputter-coated with Au/Pd using a vacuum evaporator (Edwards) and examined using a SEM (FEI Quanta 200) at 10 kV accelerating voltage.

2.2.7. Determination of bulk and tapped densities

The VE-loaded microparticles (75–500 μm) were weighted and poured into a 100 ml graduated cylinder. The bulk volume V_b and the tapped volume V_t after 1250 taps on a tap density apparatus (Erweka SVM 12) were used to calculate (in g ml^{-1}) the bulk density d and the tapped density D. Three determinations of each sample were performed and the means ± standard deviation are reported. Then, the Carr Index (or Compressibility Index, CI%) values were calculated (USP-NF, 2nd suppl. 2005) to investigate the flowability of the microparticles, as follows: $CI = 100\ (\rho_{tapped} - \rho_{bulk})/\rho_{tapped}$.

2.2.8. Determination of the drug content

The determination of the PRF content into the SLM (A) was obtained using a procedure previously reported [17–21] and conveniently modified according to the PRF properties (solubility, wavelength). Briefly, 25 mg of each fraction was poured in 200 mL of deionised water. The system was heated up at 65 °C to melt the carrier and then shaken for 4 h. Finally, the solution was filtered and the drug content was assayed spectrophotometrically (UV–vis spectrophotometer mod. UV2, Unicam, Cambridge, UK) at 250 nm. Each fraction was analysed in triplicate. The encapsulation efficiency (EE%) was then calculated. Finally, statistical analysis of the drug content was performed using one-way ANOVA F-test.

As regards VE loaded microparticles (B), 45 mg of each sample, containing 13.5 mg of VE, was dissolved in 100 ml of ethanol. The solution was filtered and the vitamin content was assayed spectrophotometrically (UV–vis spectrophotometer mod. UV2, Unicam, Cambridge, UK) at 284 nm according to EP5. Each fraction was analysed in triplicate. The encapsulation efficiency (EE%) was then calculated as follows: $EE\% = (W_a/W_t) \times 100$, where W_a = actual content and W_t = theoretical content.

2.2.9. In vitro dissolution studies

In vitro dissolution tests of microparticles A were performed using a basket apparatus (Erweka DT600) rotating at 150 rpm. As dissolution medium, 900 ml of deionised water was used at a temperature of 37 °C; each sample contained about 25 mg of PRF. The aqueous solution was filtered and continuously pumped (12.5 ml/min) to a flow cell in a spectrophotometer (UV2 Spectrometer, Unicam, Cambridge, UK). The amount of drug dissolved was analysed at 250 nm. The dissolution tests were performed at least in triplicate. The dissolution profiles were compared using f_2 similarity factor [22]. The similarity factor is a logarithmic reciprocal square-root transformation of the sum of squared error and is a measurement of the similarity in the percentage of dissolution between two curves:

$$f_2 = 50 \log \left\{ \left[1 + (1/n) \sum (R_t - T_t)^2 \right]^{-0.5} \times 100 \right\}$$

where n is the sampling number, R_t and T_t are the percent dissolved of the reference and test products at each time point t.

The similarity factor fits the result between 0 and 100. Two dissolution profiles are considered similar when the f_2 value is greater than or equal to 50. For the f_2 calculation, sampling number lower than 95% of drug released was considered.

The dissolution tests of microparticles B were performed in a paddle apparatus (Erweka DT600) rotating at 50 rpm, at 37 °C using 900 ml of deionised water containing 0.1% (w/w) of Tween 80 (HLB 15) [23]. Each sample contained about 60 mg of VE. The aqueous solution was filtered and continuously pumped (12.5 ml/min) to a flow cell in a spectrophotometer (UV2 Spectrometer, Unicam, Cambridge, UK). The amount of vitamin dissolved was analysed at 286 nm. The dissolution tests were performed at least in triplicate.

2.2.10. Differential scanning calorimetry (DSC) studies

DSC measurements were performed using a Perkin-Elmer DSC 6 (Perkin-Elmer, Beaconsfield, UK). The samples, weighting 8–12 mg, were placed into the DSC under a nitrogen flux (20 ml/min) and heated from 25 to 200 °C at a scanning rate of 10 °C/min. The PRF-loaded SLMs, were analysed 48 h after the preparation. Each analysis was car-

ried out in duplicated experiments. For comparison, the same procedure was followed for the raw materials.

3. Results and discussion

3.1. Development of the atomizer

The success and the cost of a spray congealing process mostly depend on the efficiency of the atomizer. From the view point of a lab scale to an industrial production, the main consideration concerns the spray which should produce droplets with a monodisperse size distribution and opportune fineness. In fact, small droplets have a low falling speed and consequently they require short collecting chamber, decreasing the general cost of the equipment. Moreover, an excessive falling speed of the droplets is undesirable because it can induce a partial removal of the melted material from the droplets surface due to the air attrition. Secondly, the atomizer should treat the whole batch without any occlusion and with high yields, even when the atomizer is fed with very viscous materials. To the light of these considerations, a new pneumatic nozzle was developed.

Several prototypes have been made before getting to the final realization. In particular, the air-fluid crossing angle was varied and three configurations were tested: 30°, 45° and 60°. The 30° angle produced a long narrow spray stream, while the widest angle caused a too wide atomization. A good balance between the length and the width of the spray stream and of the collecting chamber was obtained with the 45° angle. The inlet air flux was also tested: unlike other configurations, three holes placed at 120° ensured a uniform air distribution along the cone and consequently a regular and straight spray pattern.

The main advantages of using the venturimeter reported in Fig. 1 are the linear path of the fluid before the atomization, the wide orifice of the nozzle (no occlusions) and the lower kinetic energy output than that obtained by traditional nozzles (excepted for the ultrasounds devices); therefore, the microparticles congeal in a short cooling chamber.

The nozzle configuration also implies an external mixing of the fluid and of the gas after they have left the nozzle orifice. As a consequence, atomization can be varied by changing the gas pressure without affecting the liquid flow rate. In addition, high viscosity products are best atomized using external-mix nozzles. With spray congealing, usually the liquid and the gas are internal mixed and the atomization air must be heated in order to avoid excessively rapid solidification of the mass before the droplet's formation [13]. In the WPN the atomization air is not heated due to heating of the nozzle; in this way, the droplets rapidly solidify in the cooling chamber as the air output is lukewarm (the outlet air temperature varies according to the set air pressure and the nozzle temperature). Moreover, a moderate air temperature output improves the process yield due to a lower particle impact, as soft droplets can cause agglomeration or cling to the walls of the collecting chamber.

Microparticles containing a solid model drug (PRF) and a liquid nutraceutical (VE) have been prepared and characterized to evaluate the performance of the novel atomizer.

3.2. Evaluation of PRF-loaded microparticles

PRF, a class IC antiarrhythmic drug, has been widely used as treatment for ventricular and supraventricular arrhythmias since it was approved in Europe and the United States in the 1980s. Orally administered, PRF is largely absorbed and easily metabolized by the liver into active metabolites. The daily dosing regimen recommends three oral administrations with a single dose from 150 to 300 mg [24]. To reduce the side effects, minimize plasma concentration fluctuations and maintain the drug levels within a desired range, a sustained release system may be useful and microparticles could be considered. However, to swallow a reasonable dose through the oral route, microparticles should have a PRF loading not lower than 30% w/w (a PRF dose of 150 mg corresponds to 500 mg of microparticles). Obviously, a higher drug loading is preferable.

The developed WPN was able to atomize the molten mixture containing 50% w/w of PRF (formulation 1A). In the whole size fraction 75–500 μm, the theoretical and the experimental drug contents were quite similar, indicating a good encapsulation efficiency. Unfortunately, PRF was not uniformly distributed within the 1A microparticles (Table 1) as its amount increased increasing the particle size ($F_{(3,8)} = 300.44$; $p < 1.5 \cdot 10^{-8}$). This fact could be explained by hypothesizing that PRF sediments into the molten carrier and that smaller microparticles could be partially emptier than the larger ones. To improve the PRF content uniformity, colloidal silica (Aerosil®) was then added to the formulations; in fact, due to its property of suspending agent [18], it could delay the drug sedimentation in the molten mass before spraying. Two different types of hydrophobic Aerosil® were employed: Aerosil® R 812 and R 972 are hydrophobized through the bond with 3 and 2 methyl groups, respectively [25]. They have a pronounced thickening and thixotropic effect due to their large surface area (about 260 m^2/g for Aerosil® R 812 and 110 m^2/g for R 972). Hence, PRF/cetearyl alcohol microparticles containing 0.5%, 1% and 2% (w/w) of Aerosil® R 812 were prepared and compared. Table 1 reports the PRF content in each fraction of microparticles 2A, 3A and 4A and the results indicate that the difference of the drug loading in the size fractions is significantly lower than in microparticles 1A; in particular formulation 3A showed the better drug uniformity ($F_{(3,8)} = 23.49$; $p < 2.6 \cdot 10^{-4}$). When Aerosil® R 972 (2% w/w) was added to the formulation (5A), the drug distribution within the particle size fractions was uniform as well ($F_{(3,8)} = 25.08$; $p < 2.0 \cdot 10^{-4}$). An attempt to improve the mass viscosity was made adding 2% w/w soya lecithin (6A); yet the results of the drug loading

were worser than those obtained with Aerosil® ($F_{(3,8)} = 79.98$; $p < 2.63 \cdot 10^{-6}$). Therefore, formulations 3A and 5A presented a good drug loading uniformity.

For all formulations the yields were higher than 90%.

The particle size analysis (Fig. 2) evidenced that the prevalent particle size ranged from 75 to 150 μm, increasing

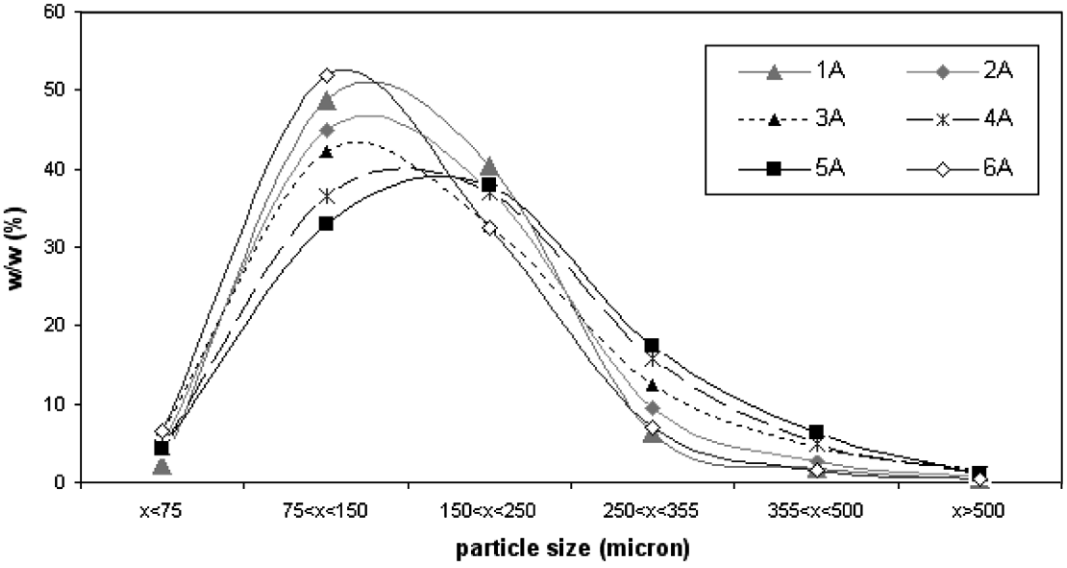

Fig. 2. Particle size distribution of the PRF-loaded SLM.

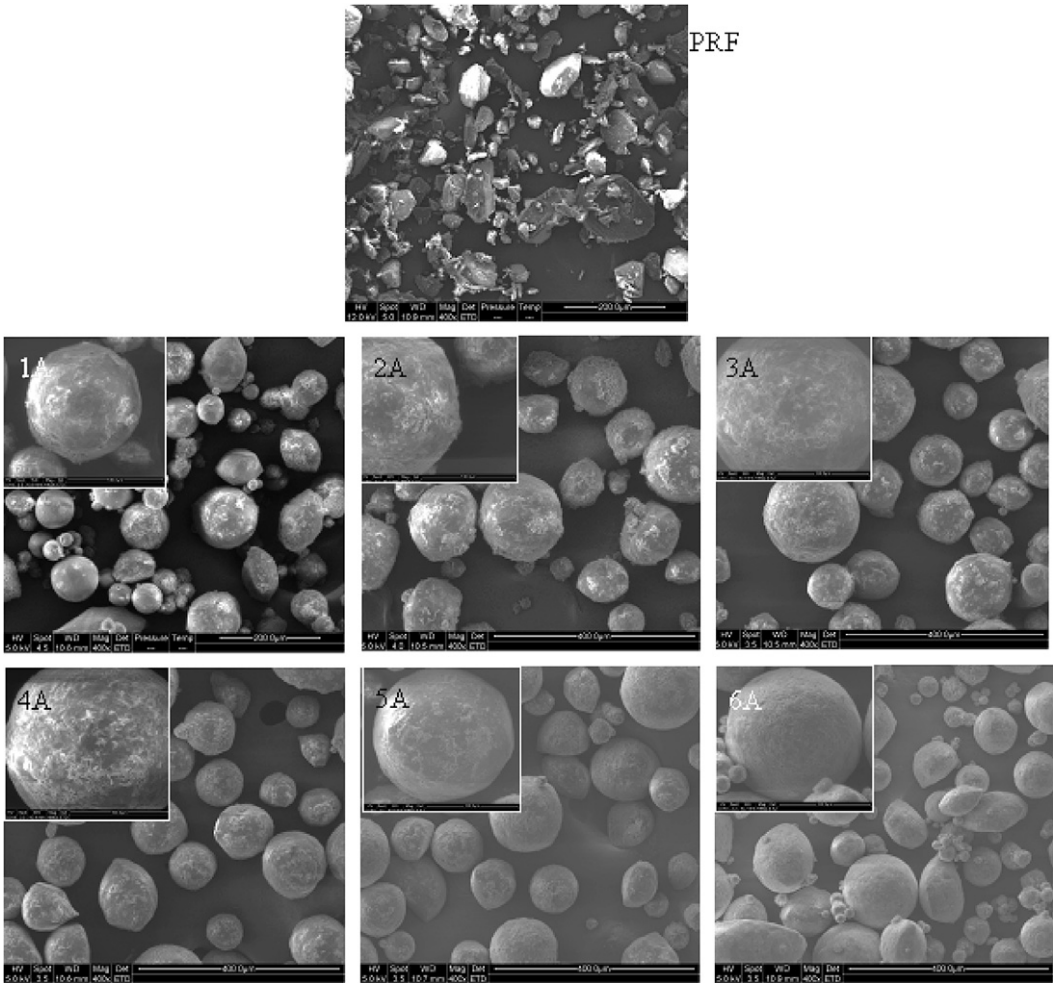

Fig. 3. SEM pictures of: pure PRF (a) and of microparticles 1A–6A at different magnifications.

the amount of Aerosil® (2A–5A), the amount of $75 < x < 150$ μm fraction decreased and the $250 < x < 355$ μm one increased. This behaviour could be correlated to the different viscosity of the molten mass; to confirm this hypothesis, viscosity measurements were performed and the viscosity values (independently from the kind or amount of the suspending and thickening agent) were then correlated to the microparticle dimensions. The results evidenced that increasing the viscosity of the suspension, the particle size lower than 250 μm decreased, while the particle size larger than 250 μm increased. In particular, the prevalent size fraction of formulations 1A and 6A was the $75 < x < 150$ μm as the viscosity was lowest (60 mPa s); while in formulations 4A and 5A the mean size fraction was $150 < x < 250$ μm as the viscosity was 400 ad 500 mPa s, respectively.

Fig. 3 shows the SEM photographs of the drug and of the PRF-loaded microparticles evidencing that PRF was in micronized form and that non aggregated microparticles with spherical shape were obtained for all the formulations (1A–6A). SEM at higher magnification revealed that the surface of microparticles 1A was quite rough due to the

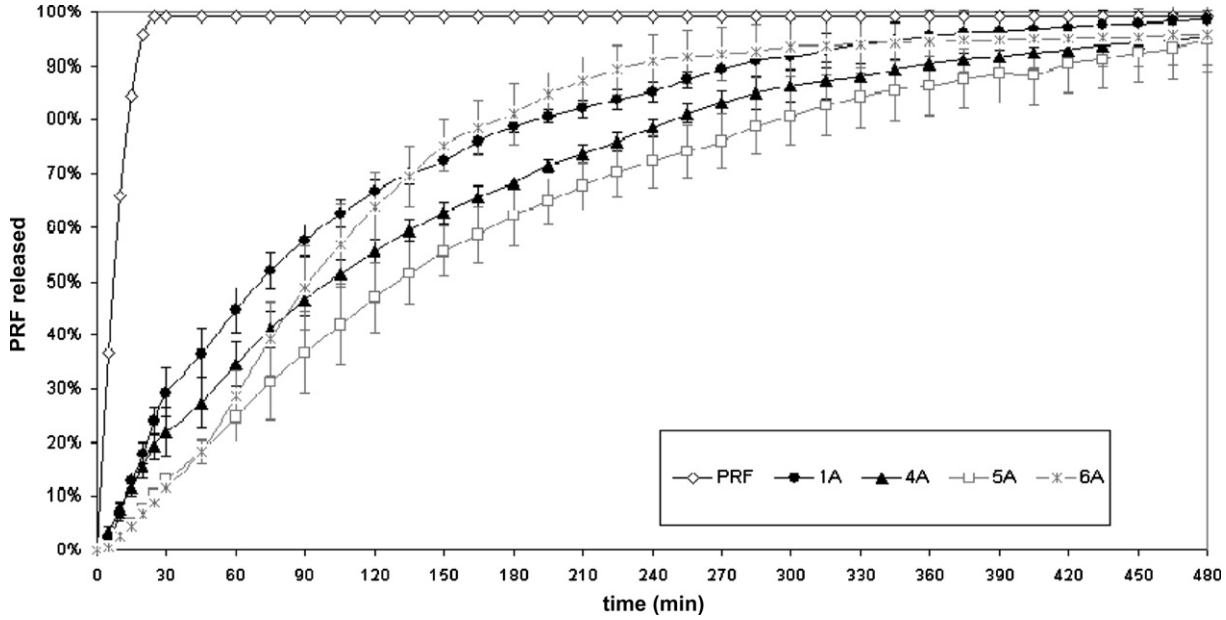

Fig. 4. In vitro dissolution profiles of PRF-loaded SLM ($75 < x < 500$ μm).

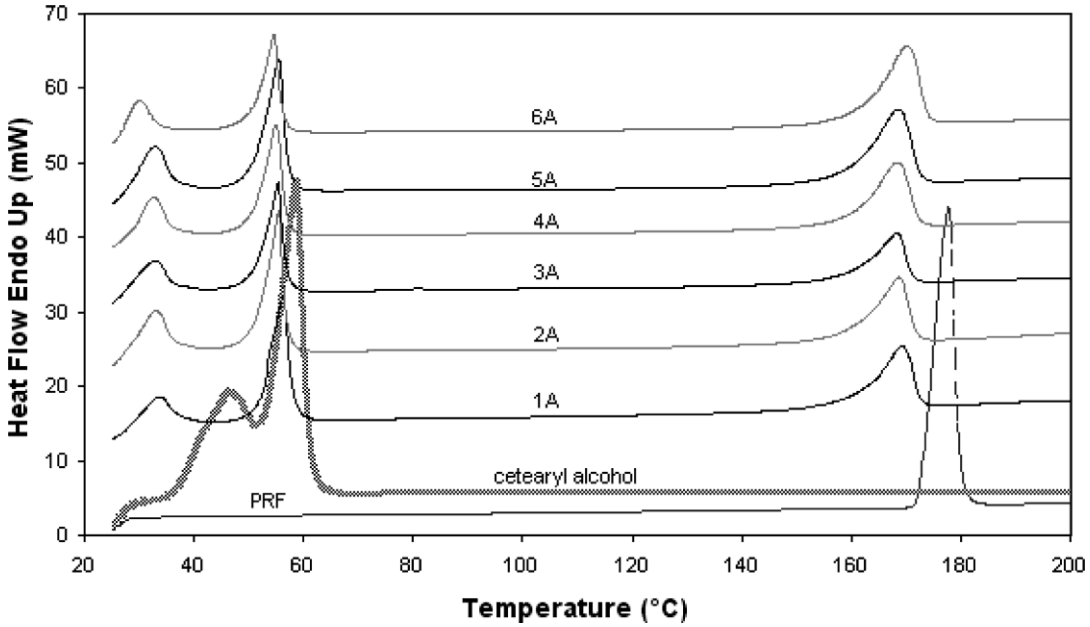

Fig. 5. DSC curves of PRF, cetearyl alcohol and of PRF-loaded SLM.

presence of the highly concentrated PRF; when Aerosil® R812 was added, the higher the amount of Aerosil® R812 in the microparticles, the rougher was the surface of the systems (2A–4A). On the contrary, the microparticles 5A, containing 2% of Aerosil® R972 had a smoother surface than microparticles 4A; this effect could be explained considering that Aerosil® R972 has a lower surface area (110 m^2/g) compared to Aerosil® R812 (260 m^2/g). Finally, the addition of soya lecithin (6A) led to a smooth surface.

Fig. 4 reports the dissolution profiles of the SLM (particle size 75–500 μm) compared to that of pure PRF. Pure PRF completely dissolved in few minutes. PRF-loaded Cetearyl alcohol microparticles (1A) showed a controlled release profile and the addition of Aerosil® R812 (4A microparticles) slightly decreased the drug release. The in vitro release data of microparticles 2A and 3A (not reported) were superimposed to the release profile of microparticles 4A demonstrating that small modifications in the Aerosil® R 812 concentration did not influence the release of the drug. Aerosil® R 972 microparticles (5A) showed a controlled release profile, while microparticles 6A exhibited a dissolution profile similar to formulation 1A.

The dissolution profiles were then compared using f_2; the results showed that using PRF as reference, the f_2 values for microparticles 1A, 4A, 5A and 6A were 9.98, 9.75, 7.86, 7.68, respectively, confirming that all microparticles significantly modified the drug release. Then, the similarity factor calculated between formulations revealed differences in the dissolution profiles only between 1A and 5A ($f_2 = 40.87$) and between 5A and 6A ($f_2 = 42.97$) microparticles.

Therefore microparticles 5A showed both good drug loading uniformity and a controlled release profile.

DSC was then used to detect possible modifications of the physico-chemical characteristics of the drug and interactions between PRF and cetearyl alcohol. DSC scans (shown in Fig. 5) indicate that PRF exhibited an endothermic peak at 176.8 ± 0.4°C ($\Delta H = 128.4 \pm 0.7$ J/g), while cetearyl alcohol showed an endotherm at 47.4 ± 0.5 °C, due to the melting of laurylic and myristic alcohols (present as a minor fraction in the excipient) and a second endotherm at 59.3 ± 0.3 °C, attributed to the cetyl- and stearyl-alcohols [26]. Aerosil® and soya lecithin did not present any phase transitions in the examined temperature range (curve not shown). The DSC scans of the microparticles (1A–6A) were very similar to each other and showed two endothermic peaks around 32–34 °C and around 56 °C, due to the carrier, and a peak at 169.7 ± 0.8 °C ($\Delta H = 50 \pm 4$ J/g), due to the drug. The lowering and broadening of the drug melting point can be due to the presence of the carrier in the molten state, as proposed by Craig [27]; therefore the results suggest the permanence of PRF in the original crystalline form.

The results presented in the first part of the study showed that WPN was able to produce solid lipid microparticles with high drug loading and good yields, upon the high viscosity of the melted mixture. The SLMs showed a relatively narrow Gaussian size distribution (considering the high viscosity of the systems), a spherical shape and a controlled release profile. Therefore, the performance of the WPN has been satisfactory in the microencapsulation of highly concentrated solid drug.

3.3. Evaluation of the VE-loaded microparticles

The ability to convert liquids to solids is an important contribution of microencapsulation to pharmaceutical products development, to the improvement of handling and to the protection from environment [2,28]. In the second part of the work, the possibility of formulating a liquid nutraceutical into a solid dosage form has been evaluated.

α-Tocopheryl acetate (Vitamin E, VE) is a clear, viscous, oily liquid, practically insoluble in water and freely soluble

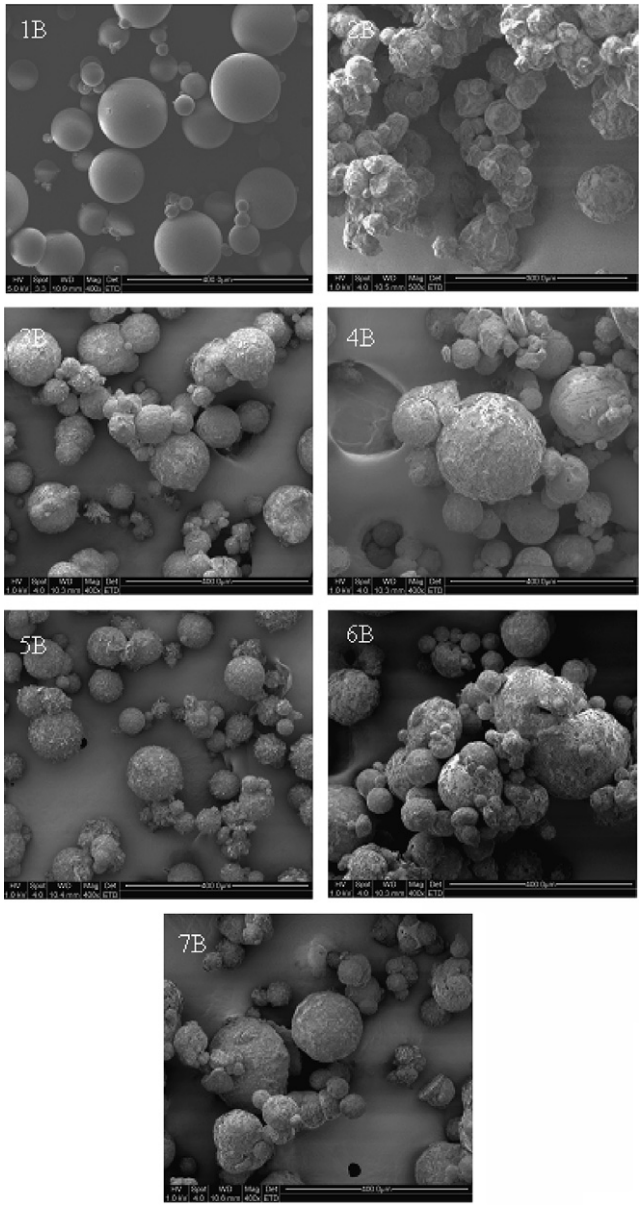

Fig. 6. SEM pictures of VE-loaded microparticles.

in acetone and in ethanol (Eur. Ph. 4th Ed.). The natural occurring form is the d (R,R,R) isomer while the synthetic analogue exists as the racemic d,l mixture. VE has a role as an antioxidant in pharmaceutical formulations and as nutraceutical product. In human supplementation, Vitamin E is available as softgels, tablets, capsules, and topical oils. Doses for oral vitamin E generally range from 50 to 1000 IU (1 mg vitamin E equals 1.5 IU). Vitamin E has also been recognized as an essential nutrient for the growth and health of all species of animals; the majority of commercially available dosage forms are oleic solution swallowed as vials or bottles for os, sc or im (150–400 IU).

Different formulations were evaluated, varying the kind of carrier and the thickening agent (Table 2); the theoretical VE content was 30% for all the formulations. The particle size distribution evidenced that the size depended on the fluid viscosity, being the air pressure fixed at 2.5 bar for all the formulations. In particular, the mean diameter of the microparticles increased increasing the viscosity of the mixture to atomized, independently by the employed carrier. For instance, the 91.38% w/w of the microparticles had diameter <250 μm when the viscosity of the fluid was 50 mPa s (formulation 1B), while 83.33% w/w of microparticles had diameter >250 μm when the viscosity was 200 mPa s (formulation 5B). The yields were higher than 90% for all formulations. The VE loading in each formulation, determined on the whole batch, is shown in Table 2. Carnauba wax microparticles (1B) exhibited the best EE (100%) despite the high nozzle temperature (about 100 °C). Microparticles prepared with PEG 4000 (5–7B), obtained setting the nozzle temperature at 70 °C, had an EE ranging from 91% to 98%, while the EE slightly decreased till 79–81% for stearyl alcohol based microparticles (2–4B), obtained at the same nozzle temperature. These results evidenced that EE depends on the affinity between VE and the carrier; when suitable carriers were employed the EE was close to 100%, indicating the absence of thermal degradation of VE during manufacturing.

Fig. 6 shows the SEM photographs of the VE-loaded microparticles. Carnauba wax microparticles (1B) appeared not aggregated, completely spherical in shape with a very smooth surface due to the high melting point of the carrier (80 °C). In contrast, aggregated microparticles with rough surfaces have been obtained using stearyl alcohol (2B); the addition of Aerosil® (3B and 4B) improved the microparticle morphology, suggesting an improvement in flowability with respect to 2B. The results of the bulk (d) and tapped (D) densities confirmed that microspheres 3B and 4B had better flowability and lower compressibility than 2B (IC% = 20.04, 15.10 and 9.08 for 2B, 3B and 4B, respectively), which are fundamental parameters for the filling of trunks. Hence, these products, especially microparticles 4B, can be simply mixed with the animal feedstuffs.

Microparticles based on PEG 4000 (5B–7B) were spherical but partially aggregated.

The in vitro dissolution profiles (Fig. 7) of the 1B–7B microparticles showed that it was possible to modify the VE dissolution profile according to the hydrophilic/hydrophobic characteristic of the carrier. In particular, carnauba wax did not improve the VE availability. Formulations 2B-4B based on stearyl alcohol exhibited the desired dissolution profiles, since they had a dissolution behaviour suggesting a zero order release kinetics. Moreover, the release data of formulations based on stearyl alcohol demonstrated that apparently little modifications regarding the Aerosil type strongly influenced the dissolution behaviour of VE. Formulation 5B, based on PEG 4000, released over 80% of VE in 1 h and the addition of Aerosil®90 further enhanced the VE dissolution; while 7B microparticles (with

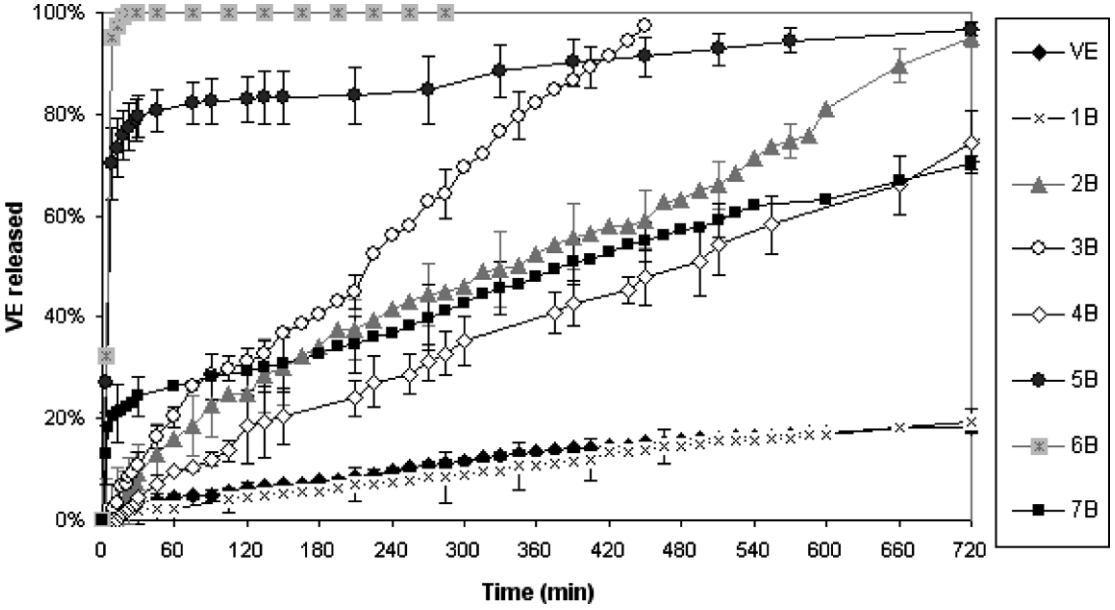

Fig. 7. In vitro dissolution profiles of VE-loaded microspheres ($75 < x < 500$ μm).

Aerosil® R 812) released about 30% and 56% of VE in 1 and 8 h, respectively, confirming that the selection of the Aerosil® type strongly influenced the drug release.

4. Conclusions

The spray congealing system equipped with the novel nozzle (WPN) is able to nebulize very viscous systems, usually not processed by conventional apparatus, producing spherical and not aggregated microparticles. The loading of the drug, either in the solid or in the liquid form, is high for microspheres (50% for propafenone hydrochloride and 30% for α-tocopheryl acetate), the yield approaches 95%, the size distribution is relatively narrow and the drug release can be easily modified according to the nature (lipophilicity and/or hydrophilicity) of the excipients.

This system is a versatile and cheap manufacturing method as it is less time consuming and has a low energy consumption (low air pressure and temperature employed). Therefore, the spray congealing system has good potentiality for the manufacture of microparticulate dosage forms in perspective of pharmaceutical industry.

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References

[1] B.-F. Gibbs, S. Kermasha, I. Alli, C.-N. Mulligan, Encapsulation in the food industry: a review, Int. J. Food Sci. Nutr. 50 (1999) 213–224.
[2] J.A. Bakan, Microencapsulation, in: J. Swarbrick, J.C. Boylan (Eds.), Encyclopedia of Pharmaceutical Technology, Vol. 9, Marcel Dekker Inc., New York, 1994, pp. 423–441.
[3] Spray cooling/spray congealing. Available online at http://www.niroinc.com/html/chemical/cspraycooler.html., accessed March 2007.
[4] S. Jaspart, G. Piel, L. Delattre, B. Evrard, Solid lipid microparticles: formulation, preparation, characterization, drug release and applications, Expert Opin. Drug Deliv. 2 (1) (2005) 75–87.
[5] T. Yajima, N. Umeki, S. Itai, Optimum spray congealing conditions for masking the bitter taste of clarithromycin in wax matrix, Chem. Pharm. Bull. 47 (2) (1999) 220–225.
[6] M. Savolainen, C. Khoo, H. Glad, C. Dahlqvist, A.-M. Juppo, Evaluation of controlled-release polar lipid microparticles, Int. J. Pharm. 244 (2002) 151–161.
[7] M. Savolainen, J. Herder, C. Khoo, K. Lövqvist, C. Dahlqvist, H. Glad, A.-M. Juppo, Evaluation of polar lipid-hydrophilic polymer microparticles, Int. J. Pharm. 262 (2003) 47–62.
[8] L.-C. Li, L. Zhu, J.-F. Song, F.-S. Deng, R. Bandipadhyay, D.-E. Wurster, Effect of solid state transition on the physical stability of suspensions containing bupivacaine lipid microparticles, Pharm. Dev. Technol. 10 (2) (2005) 309–318.
[9] Y. Akiyama, A. Yoshioka, H. Oribe, S. Hirai, N. Kitamori, H. Toguchi, Novel oral controlled-release microspheres using polyglycerol esters of fatty acids, J. Control. Release 26 (1) (1993) 1–10.
[10] T. Uchida, A. Tanigaka, Y. Miyanaga, K. Matsuyama, M. Kobayashi, H. Ikezaki, A. Taniguchi, Evaluation of the bitterness of antibiotics using a taste sensor, J. Pharm. Pharmacol. 55 (11) (2003) 1479–1485.
[11] Q. Sheng, D. Deutsch, D.-Q.-M. Craig, An Investigation into the interaction between taste masking fatty acid microspheres and alkaline buffer using thermal and spectroscopic analysis, J. Pharm. Sci. 95 (5) (2005) 1022–1028.
[12] A. Maschke, C. Becker, D. Eyrich, J. Kiermaier, T. Blunk, A. Göpferich, Development of a spray congealing process for the preparation of insulin-loaded lipid microparticles and characterization thereof, Eur. J. Pharm. Biopharm. 65 (2) (2006) 175–187.
[13] M.J. Killeen, Spray drying and spray congealing of pharmaceutics, in: J. Swarbrick, J.C. Boylan (Eds.), Encyclopedia of Pharmaceutical Technology, vol. 14, Marcel Dekker Inc., New York, 1996, pp. 207–221, 1996.
[14] M.-B. Mackaplow, I.-E. Zarraga, J.-F. Morris, Rotary spray congealing of a suspension: effect of disk speed and dispersed particle properties, J. Microencapsul. 23 (7) (2006) 793–809.
[15] M. Emas, H. Nyqvist, Methods of studying aging and stabilization of spray-congealed solid dispersions with carnauba wax. 1. Microcalorimetric investigation, Int. J. Pharm. 197 (2000) 117–127.
[16] L. Rodriguez, N. Passerini, C. Cavallari, M. Cini, P. Sancin, A. Fini, Description and preliminary evaluation of a new ultrasonic atomizer for spray-congealing processes, Int. J. Pharm. 183 (1999) 133–143.
[17] N. Passerini, B. Perissutti, B. Albertini, D. Voinovich, M. Moneghini, L. Rodriguez, Controlled release of Verapamil hydrochloride from waxy microparticles prepared by spray-congealing, J. Control. Release 88 (2003) 263–275.
[18] B. Albertini, N. Passerini, M.-L. González-Rodríguez, B. Perissutti, L. Rodriguez, Effect of Aerosil® on the properties of lipid controlled release microparticles, J. Control. Release 100 (2) (2004) 233–246.
[19] N. Passerini, B. Perissutti, M. Moneghini, D. Voinovich, B. Albertini, C. Cavallari, L. Rodriguez, Characterization of carbamazepine-Gelucire 50/13 microparticles prepared by a spray-congealing process using ultrasounds, J. Pharm. Sci. 91 (3) (2002) 699–707.
[20] C. Cavallari, L. Rodriguez, B. Albertini, N. Passerini, F. Rosetti, A. Fini, Thermal and fractal analysis of diclofenac/gelucire microparticles obtained by ultrasound assisted atomization, J. Pharm. Sci. 94 (5) (2005) 1124–1134.
[21] N. Passerini, B. Albertini, B. Perissutti, L. Rodriguez, Evaluation of melt granulation and ultrasonic spray congealing as techniques to enhance the dissolution of praziquantel, Int. J. Pharm. 318 (1–2) (2006) 92–102.
[22] J.-W. Moore, H.-H. Flanner, Mathematical comparison of dissolution profiles, Pharm. Tech. 20 (1996) 64–74.
[23] Y. Takashima, H. Yuasa, Y. Kanaya, I. Nomura, K. Shinozawa, Reduction of tablet coloration at tableting for oily medicine (tocopheryl nicotinate), Int. J. Pharm. 187 (1) (1999) 125–135.
[24] E.-L.-C. Pritchett, R.-L. Page, M. Carlson, K. Undesser, G. Fava, Efficacy and safety of sustained-release propafenone (Propafenone SR) for patients with atrial fibrillation, Am. J. Cardiol. 92 (8) (2003) 941–946.
[25] Aerosil® – Technical Bullettin N. 27, Degussa, Germany.
[26] L. Giovannelli, S. Bellomi, F. Pattarino, B. Albertini, Characterization of nifedipine microparticles prepared by hot air coating technique, Int. J. Pharm. 293 (2005) 225–234.
[27] D.-Q.-M. Craig, The mechanisms of drug release from solid dispersion in water-soluble polymers, Int. J. Pharm. 231 (2002) 131–144.
[28] S.-A. Barker, S.-P. Yap, K.-H. Yuen, C.-P. McCoy, J.-R. Murphy, D.-Q.-M. Craig, An investigation into the structure and bioavailability of a-tocopherol dispersions in Gelucire 44/14, J. Control. Release 91 (2003) 477–488.