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New solid lipid microparticles for controlled ibuprofen release: Formulation and characterization study

Laurent Perge, Mike Robitzer*, Coralie Guillemot, Jean-Marie Devoisselle, Françoise Quignard, Philippe Legrand**

Institut Charles Gerhardt Montpellier UMR 5253 CNRS-UM2-ENSCM-UM1, Matériaux Avancés pour la Catalyse et la Santé, ENSCM, 8 rue de l'Ecole Normale, 34296 Montpellier Cedex 5, France

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ABSTRACT

A hot melt dispersion method was used to prepare new sustained release ibuprofen composite microparticles of a solid lipid at ambient temperature, cetyl alcohol. The dispersion of colloidal silicon dioxide nanoparticles (hydrophilic Aerosil® 200 or hydrophobic Aerosil® R974) either in the oily phase or in the aqueous phase led to the preparation of large (about 400 μm diameter) surfactant free free-flowing particles. Mapping-scanning electronic microscopy using silicon probe revealed that silicon was in the oily core in all cases. The nature of silica nanoparticles and the way used for their dispersion influenced the internal structure of the composite microparticles and the aggregation of nanoparticles in the core of the microparticles. Hydrophobic Aerosil® R974 allowed the formation of homogeneous microparticles. Although silica nanoparticles had no influence on thermic profile, crystalline state of ibuprofen and lipid, they had an influence on the kinetics drug release related to the increase of the size of the composite solid lipid microparticles prepared.

1. Introduction

According to continuous demand of controlled drug release, recently many researches were devoted to the development of inorganic solids based on silica precursors able to control the kinetics of release of associated drugs. These materials were obtained by a combination of controlled porosity owing to templated organic compounds in the framework of pharmaceutical materials (Manzano et al., 2009). Unfortunately, the main disadvantage of this kind of silicate precursors is their lack of pharmaceutical acceptance. However, colloidal silica nanoparticles, which are amorphous raw material of pharmaceutical grade (Aerosil®), are present for 60 years in many pharmaceutical dosage forms. They are mainly used as glidants (Lerk et al., 1977), adsorbents (Mani et al., 2004) or either disintegrants in solid dosage forms (El-Shanawany, 1993), but also in semi-solid and liquid dosage forms as viscosity or strength controlling agents (Sherriff and Enever, 1979; Kotsiomiti and McCabe, 2008), freeze-dried nanocapsules stabilizer or as emulsion stabilizer for the preparation of Pickering emulsions (Aveyard et al., 2003; Schaffazick et al., 2003; Frelichowska et al., 2009). Nevertheless, only few studies by pharmaceutically accepted colloidal silica nanoparticles focused on the preparation of pharmaceutical dosage form to control the kinetics drug release. In the case of the Pickering emulsions, the colloidal silica nanoparticles adsorbed at the oil/water interface of these O/W or W/O emulsions can form a rigid coating. Chambin et al. (2002) and Ito et al. (2010) showed that these silica nanoparticles were responsible for a decrease in the kinetics of release of a water soluble encapsulated drug (Chambin et al., 2002; Ito et al., 2010). The idea of controlling the release of water insoluble drug encapsulated in W/O Pickering emulsion by silica nanoparticles was further developed by Prestidge and Simovic (2006). They observed that the coating of emulsions with silica nanoparticles could influence the kinetics of release of encapsulated drug. Fast release was achieved with water insoluble drugs in emulsified systems stabilized by hydrophilic colloidal silica whatever the pH and the ionic strength of the aqueous continuous phase. At the opposite hydrophobic colloidal silica nanoparticles were able to reduce the kinetics of release of water insoluble drugs at high pH and ionic strength of the aqueous continuous phase (Simovic and Prestidge, 2007). Albertini et al. (2004), Chauhan et al. (2005) and Cheboyina and Wyandt (2008) proposed an alternative to the Pickering emulsion. They observed that introduction of hydrophilic colloidal silica nanoparticles inside solid lipid microparticles (SLM) could also modulate the kinetics of release through the formation of a silica network inside the solid lipid microparticles. The amount of silica and its surface area were the main factors influencing the drug release.

* Corresponding author. Tel.: +33 467163494; fax: +33 467163470.
** Corresponding author. Tel.: +33 411759445; fax: +33 411759465.
E-mail addresses: mike.robitzer@enscm.fr (M. Robitzer), philippe.legrand@univ-montp1.fr, philippe.legrand@enscm.fr (P. Legrand).

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These composite microparticles present a major interest to promote applications of SLM. In fact, SLM are biodegradable, chemically stable microparticles able to incorporate high amounts of lipophilic drugs. SLM can be administered by subcutaneous, oral, intramuscular, topical or pulmonary ways, but SLM suffered from a low resistance to heat, and show some physical instability and early burst releases of encapsulated drugs as proteins or water insoluble drugs as ibuprofen (zur Mühlen et al., 1998; Ahlin et al., 2000; Jaspart et al., 2005; Trotta et al., 2005; Long et al., 2006a; Almeida and Souto, 2007). This phenomenon could be due to the location of a part of the associated drug at the outer surface of the particles (Pietkiewicz et al., 2006) or by the presence of surfactants at the interface.

In this article we report the formulation of a new composite SLM (CSLM) obtained by hot melt homogenization, encapsulating ibuprofen, a poorly water soluble model drug and based on cetyl alcohol, a lipid solid at room and body temperature known to reduced drug release (Long et al., 2006b) in the presence of different kinds of silica nanoparticles.

The CSLM were prepared as a function of the nature of Aerosil®, hydrophilic or hydrophobic colloidal silica nanoparticles (Aerosil® 200 or Aerosil® R974, respectively) and the way of its dispersion (directly dispersed in melted lipid phase or in the aqueous phase). They were characterized by scanning electron microscopy (SEM), dynamic quasi-elastic light scattering (DQELS), thermal and spectral analyses to evaluate the size, the morphology, the surface and the inner core of the CSLM, the physical state of lipid and drug, the silica and drug loading and finally drug release.

2. Materials and methods

2.1. Materials

The production of solid lipid microparticles (SLM) and composite solid lipid microparticles (CSLM) required the following materials: cetyl alcohol, Sipol C16 (Henkel, Düsseldorf, Germany); polyvinylic alcohol Gohsenol EG05, (saponification degree, 86.5–89%) (Nippon Gohsei, Osaka, Japan); two kinds of fumed silica nanoparticles, Aerosil® 200 and Aerosil® R974 (see Table 1 for physical properties) (Evonik, Frankfurt, Germany); ibuprofen (Global bulk drugs and fine chemicals limited, Hyderabad, India). All were used without further purification.

Potassium chloride, potassium dihydrogen, disodium hydrogen phosphate, sodium chloride, hydrochloric acid and sodium hydroxide were supplied by Prolabo Normapur (Darmstadt, Germany) to prepare dissolution media.

2.2. Particles preparations

2.2.1. Solid lipid microparticles (SLM)

Cetyl alcohol was heated to 65 °C, above its melting temperature with or without ibuprofen (5–15 wt% of lipid). The surfactant PVA (0.5%, w/v) was dissolved in water and heated to 90 °C to achieve dissolution before cooling to 65 °C. The emulsion was obtained by stirring the aqueous phase with the lipid phase in a ratio 30:1 (w/w), using a turbine Ika Eurostar (IKA® Werke GmbH & Co. KG, Germany), with a 3-blade Teflon® propeller at 750 rpm. Finally the emulsion was cooled at 20 °C after 8 min mixing. That allowed lipid crystallization and SLM formation. Microparticles were separated by filtration, washed with distilled water before being freeze-dried.

Table 1
Physical properties of silica nanoparticles.

Product name	Contact angle (°) air/water	BET surface area (m^2/g)	Average primary particle size (nm)
Aerosil® 200	14	200 ± 25	12
Aerosil® R974	117 ± 4	170 ± 20	12

2.2.2. CSLM associated with silica nanoparticles into the lipid phase

Cetyl alcohol (CA) was heated to 65 °C with one type of fumed silica nanoparticles (3 wt% of CA) with ibuprofen (5 wt% of CA). The aqueous phase (0.1 M NaCl, pH 9 adjusted with NaOH 1 M) was heated at 65 °C to avoid premature decrease of temperature of the organic phase during the emulsion process. The emulsion was obtained at 65 °C by stirring the aqueous phase with the organic phase in a ratio 30:1 (w/w), using a turbine Ika Eurostar (IKA® Werke GmbH & Co. KG, Germany), with a 3-blade Teflon® propeller at 750 rpm. The emulsion was cooled at 20 °C after 1 h mixing. Microparticles were separated by filtration, washed with distilled water before being freeze-dried.

2.2.3. CSLM associated with Silica nanoparticles suspension in the aqueous phase

CA was heated to 65 °C, above its melting temperature with ibuprofen (5 wt% of CA). The fumed silica nanoparticles suspension was obtained by dispersing Aerosil® 200 or 974 (0.1%, w/v) in the aqueous phase (0.1 M NaCl, pH 9 adjusted with NaOH 1 M) and heated at 65 °C to avoid premature decrease of temperature of the organic phase during the emulsion process. The emulsion was obtained by stirring the aqueous phase with the lipid phase in a ratio 30:1 (w/w), using a turbine Ika Eurostar (IKA® Werke GmbH & Co. KG, Germany), with a 3-blade Teflon® propeller at 750 rpm. The emulsion was cooled at 20 °C after 1 h mixing. Microparticles were separated by filtration, washed with distilled water before being freeze-dried.

Composite microparticles are labelled according to their theoretical charge in silica and ibuprofen. For example MP-S_3-I_5 is attributed to composite microparticles containing a theoretic charge of 3% of silica and 5% of ibuprofen per mass of particles. The actual charges in silica and ibuprofen were determined by thermogravimetric and UV–vis analysis, respectively. The labels and compositions of the different samples used in this work are reported in Table 2.

2.3. Particles characterizations

2.3.1. Size and morphology of microparticles

Particle-size measurements were determined using the laser Mastersizer 2000 (Malvern instruments Ltd., UK) equipped with an optical bench of two lasers (helium-neon laser, λ = 543 nm and

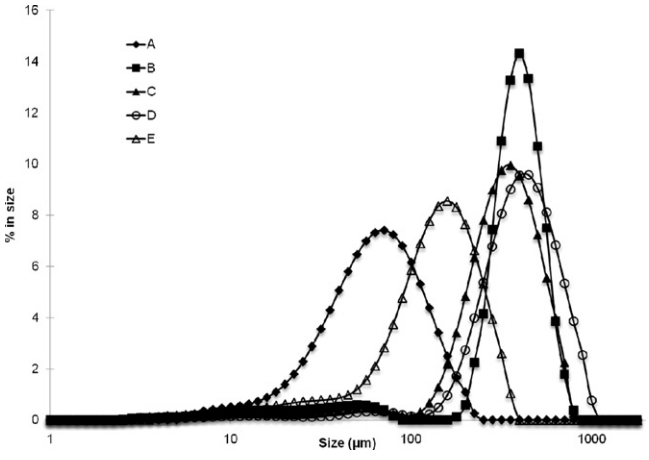

Fig. 1. Size distribution of SLM and CSLM A, B, C, D, E which are defined in Table 2.

Table 2
Labels and composition of the samples.

Theoretic charge in silica and ibuprofen	Text label	Type of fumed silica nanoparticles	Location of fumed silica nanoparticles	Experimental silica loading (mg/g of SLM)	Silica Encapsulation efficiency (%)	Experimental ibuprofen loading (mg/g of SLM)	Ibuprofen Encapsulation efficiency (%)
MP-S_0-I_5	A	None		0		50 ± 4	100
MP-S_3-I_5	B	Aerosil® R974	Organic	35	100	56 ± 3	100
MP-S_3-I_5	C	Aerosil® R974	Aqueous	27	90	53 ± 2	100
MP-S_3-I_5	D	Aerosil® 200	Organic	32	100	52 ± 3	100
MP-S_3-I_5	E	Aerosil® 200	Aqueous	28	93	49 ± 4	98

diode laser, $\lambda = 405$ nm). Measurements were achieved on aqueous dispersions of SLM, under stirring (3500 rpm).

To compare the size distributions, the terms $d(10)$, $d(50)$ and $d(90)$ respectively represent the diameter where 10%, 50% and 90% of all particles are finer than this size. The span (S) was defined as the ratio $[d(90) - d(10)]/d(50)$ and reflected the polydispersity of the sample.

2.3.2. Scanning electron micrographs (SEM)

Scanning electron micrographs (SEM) of the microparticles were obtained using a S-4800 I (Hitachi, Japan) after platinum metallization. The elemental mapping was recorded on a cross-section of the particles using a S-4500 I SEM apparatus (Hitachi, Japan) with a resolution of 1 μm^2.

2.3.3. Differential scanning calorimetry (DSC)

The DSC enthalpy peaks correspond to the melting temperatures of organic components. The system used was DSC Perkin-Elmer 7 apparatus (Perkin-Elmer, USA). The sample (8 mg) was placed in an aluminium tank. An empty aluminium tank was used as a reference. The pressure of nitrogen was 2 bar. The temperature profile was first 2 min at 25 °C, then heating at 5 °C min^{-1} from 25 °C to 90 °C, with a rate of 5 °C min^{-1}, followed by 1 min at 90 °C and finally cooling from 90 °C to 25 °C, with a rate of 5 °C min^{-1}. All the samples were sieved between 125 and 250 μm to avoid thermal properties from the diffusion of heat due to the surface developed by the particles.

Enthalpy of fusion of Sipol C16 was obtained through direct proportionality to the Sipol content in the samples.

2.3.4. X-ray diffraction (XRD)

X-ray diffractograms were obtained in a D8 Advance Brüker Theta Theta (Brüker, Germany) with a power of 40 kV and 40 mA for the analysis at wide angles, and 40 kV and 35 mA for the analysis at small angles.

2.3.5. IR spectroscopy

Transmission infrared spectra of samples were done with pellets using anhydrous KBr (Sigma–Aldrich, USA). They were recorded in the range 7200–450 cm^{-1} on Brüker Vector 22 Fourier transformed spectrometer (Brüker, Germany) equipped with a DTGS detector.

2.3.6. Determination of silica loading

Thermogravimetric analysis was performed with Perkin-Elmer STA 6000 apparatus (Perkin-Elmer, USA) sample 10 mg, air flow of 40 mL/min, 25–900 °C with a rate of 5 °C min^{-1}. The percentage of silica in the composition of the microparticles was determined by the residual mass at 900 °C.

2.3.7. Determination of ibuprofen loading

To measure the actual amount of ibuprofen loaded in the microparticles, a sample of 20 mg of microparticles was placed in a flask. It was then filled with ethanol to achieve complete dissolution of the organics and destructuration of the particles. The solution was diluted to 1/10th before being analyzed by UV spectroscopy (Spectrometer UV/Visible Lambda 35, Perkin-Elmer, USA) at $\lambda = 222$ nm. The molar extinction coefficient of ibuprofen in ethanol at 222 nm determined from a standard range was equal to: $\varepsilon_{222\,nm} = 0.04$ L mol^{-1} cm^{-1}. The loading of ibuprofen was determined from the molar extinction coefficient and the absorbance of the sample. The determination of drug loading was made in triplicate.

$$\text{Drug loading } (\%) = \frac{\text{amount of drug in SLM or CSLM}}{\text{amount of SLM or CSLM}} \times 100$$

$$\text{Encapsulation efficiency} = \frac{\%\ \text{of experimental drug loading}}{\%\ \text{of theoretical drug loading}} \times 100$$

2.3.8. In vitro release studies

The release kinetics of ibuprofen encapsulated in microparticles are studied using a USP/EP flow-through method (dissolutest CE 1, SOTAX, Switzerland). Cells have an internal diameter of 22.6 mm. For laminar flow conditions, the conical part of the cell was filled with 1 mm diameter glass beads. A ruby bead of diameter 5 mm in the tip of the cell cone prevented solvent escaping when the cell was removed from the unit at the end of the test. For laminar operation, the sample was inserted in the cell above the bed of glass beads. The dissolution medium entered the cone through a capillary bore situated on the bottom and flows upwards. The cone was separated from the cylindrical portion by a 40-mesh screen. The amount of particles beads used per experiment was adapted to contain 25 mg of ibuprofen. The flow rate of dissolution medium across the cell was set at 8 mL/min and the cell was thermostated at 37 °C. The dissolution fluid was a simulated intestinal medium (PBS pH 7.4). Experiments were carried out in a closed loop setup and samples were collected over time during enough hours to allow the nearly complete release of ibuprofen. Each sample was analysed at least in duplicate.

Ibuprofen dissolved was analyzed by UV spectroscopy at 222 nm. We used the molar extinction coefficient of ibuprofen in PBS previously committed to $\varepsilon_{222\,nm} = 0.0455$ L mol^{-1} cm^{-1}. The determination of ibuprofen in PBS was made in triplicate.

3. Results and discussion

3.1. Preparation and physical characterization of SLM and CSLM

3.1.1. Preparation

Different composite solid lipid microparticles based on fumed silica nanoparticles, either highly hydrophilic Aerosil® 200, or highly hydrophobic Aerosil® R974 are prepared by two different one-pot hot emulsification of cetyl alcohol (CA) at a temperature above the melting point of CA. In all cases, constant and high speed agitation (750 rpm) of composite melted emulsion until cooling generates physically stable surfactant free microparticles. After freeze-drying, CSLM are spherical and free flowing microparticles with a non negligible tensile strength as compared with agglomerated sticky and soft PVA stabilized SLM. It can be noticed the physical instability of CSLM particles before cooling, when the high

speed rotation is stopped at 65 °C, as already reported by Aveyard et al. (2003).

The ibuprofen and silica loading efficiencies, reported in Table 2, are close to 100% whatever the way of preparation and the nature of silica nanoparticles with yields of preparations close to 100%.

3.1.2. Particle size measurements

According to particle size measurements (Fig. 1), all the formulations of CSLM have larger size than the SLM (10 times larger, about 400 μm as compared with 40 μm respectively for their median size) and a reduced span index (from 1.8 to 0.9 for A and B, respectively) in agreement with previous studies by Aveyard et al. (2003). The exception concerned CSLM containing Aerosil® 200 dispersed in the aqueous phase measuring about 150 μm and a span index of 1.5. The increase of size of CSLM seems to be due to both silica and ibuprofen loading because median size of unloaded CSLM prepared with Aerosil® R974 in aqueous phase is about 200 μm (data not shown). The size of these CSLM are slightly higher than the size of other unloaded or drug loaded SLM produced also by hot melt emulsion (Jaspart et al., 2005).

3.1.3. Microscopy analyses

The investigation by SEM shows that all batches of loaded or unloaded CSLM present irregular surface (Figs. 3–4) as compared with relatively smooth surfaces of ibuprofen loaded or unloaded SLM (Fig. 2).

Core of CSLM are based on lamellar phase of cetyl alcohol as observed in Figs. 3d and 4f. It can be noticed that it is impossible to localise drug crystals anywhere associated with these particles. At the opposite aggregated silica nanoparticles can be observed not only on the surface of CSLM (Fig. 3b) but also on the cross-section of CSLM owing to SEM investigations coupled or not with Si element mapping (coloured dots, Figs. 3–5). Thus a composite material of silica and organic phase (lipid and ibuprofen) is obtained whatever the ways of preparation of CSLM and whatever the kind of silica nanoparticles used. SEM microscopy of cross-sections of microparticles coupled with EDX microprobe analyses of different atomic element (as silicon, carbon, oxygen, chloride and sodium) mainly reveals the differences of localization of silicon atoms as a function of the nature of Aerosil® and the way of process on microparticles. The main difference between the various preparations of CSLM

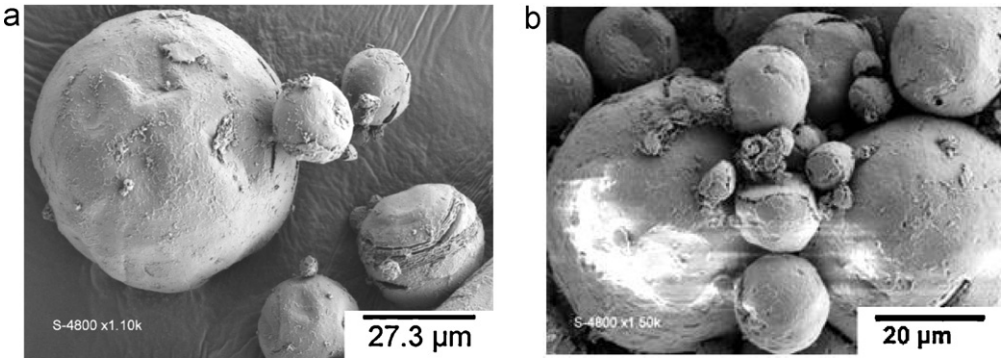

Fig. 2. SEM micrograph of SLM: (a) unloaded; (b) 5% weight of ibuprofen loaded.

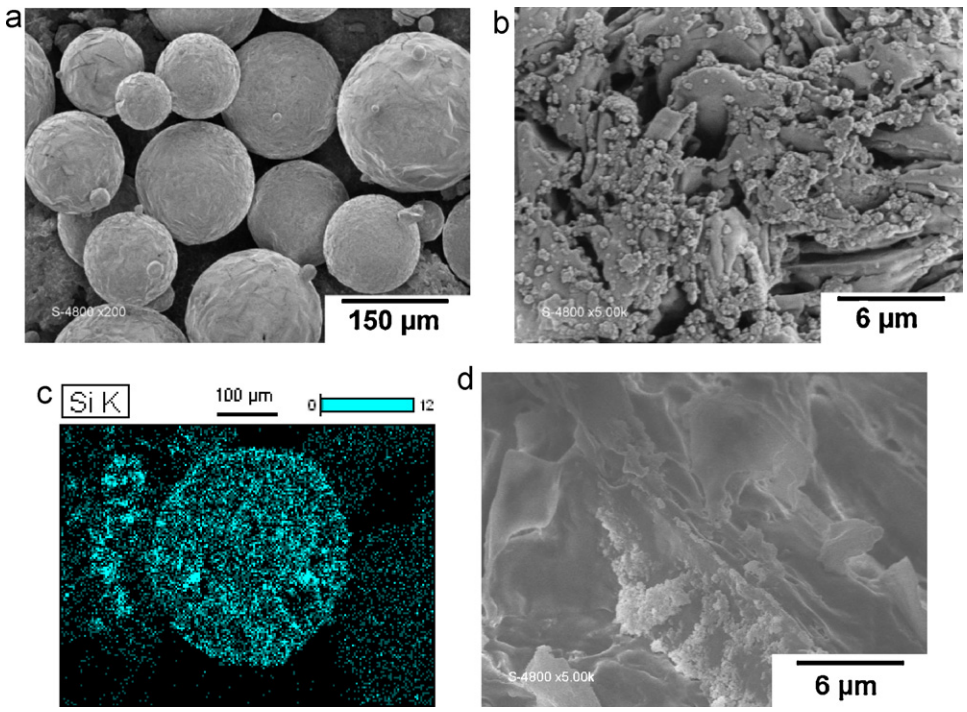

Fig. 3. SEM micrographs of ibuprofen free CSLM (MP-S_3-I_0) obtained by dispersion of Aerosil® R974 in the aqueous phase: (a) entire microparticles; (b) focus on the surface of entire microparticles; (c) Si element mapping with EDX microprobe analysis coupled with SEM technique of microparticles cross-section; (d) focus on cross-section of microparticles.

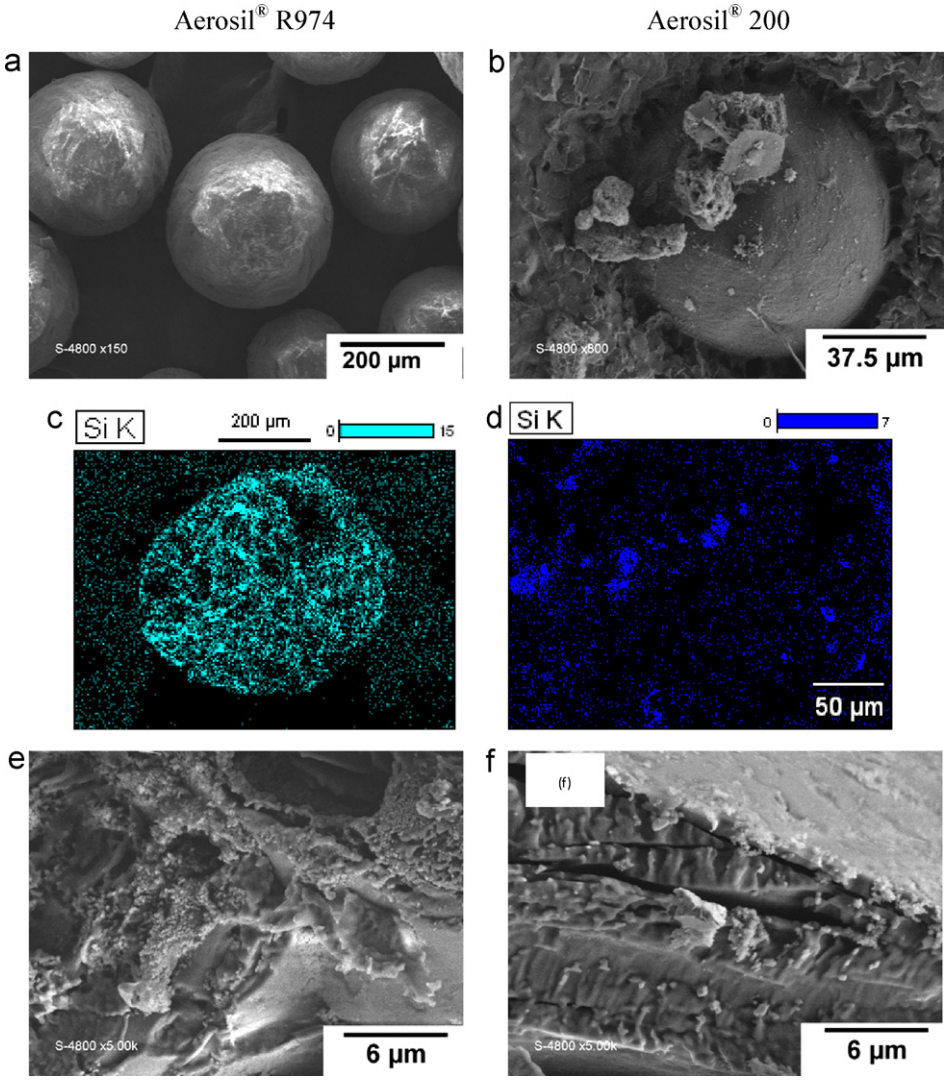

Fig. 4. SEM micrographs of ibuprofen loaded CSLM (MP-S_3-I_5) obtained by dispersion of Aerosil® in the aqueous phase. Formulation C with Aerosil® R974 (left) Formulation E with Aerosil® 200 (right): (a and b) entire microparticles; (c and d) Si element mapping with EDX microprobe analysis coupled with SEM technique of microparticles cross-section; (e and f) Focus on cross-section of microparticles. C and E are defined in Table 2.

concerns the homogeneity of the dispersion of silicon on cross-section of CSLM.

When silica nanoparticles are dispersed in the aqueous phase (Fig. 4) the differences of location of silicon dispersed in all the core of CSLM is observed for the first time on cross-section of particles owing to EDX microprobe. The understanding of these differences of internal structure of CSLM by the use of hydrophilic or lipophilic silica nanoparticles depends firstly on the knowledge of the interaction of particles dispersed in water at pH 9 and 0.1 M NaCl before emulsification and secondly on the interaction of these particles with the oily phase.

Dispersion of *Aerosil® 200* in the aqueous phase during emulsification was not favourable for the preparation of homogeneous CSLM. In fact, two populations of microparticles were present in the case of the formulation E: one population of hydrophilic silica stays in the aqueous phase to form small (below 100 μm) CSLM (irregular-shape microparticles highly silica loaded which have not diffuse through the lipid phase in Fig. 4b) while another population is in interaction with lipid phase and diffuse in the core of lipid phase to form large composite microparticles. Using Aerosil® 200 Fig. 4d shows few intense areas of silicon dispersed in lipid core. It was confirmed on focus of cross-section of these CSLM (Fig. 4f), where lamellar lipid represents the main material of the core with only few silica particles. These observations can explain the high span index of these microparticles of reduced size as compared with the other CSLM. These results are in agreement with the value of the contact angle (air/water) of Aerosil® 200, below 90° ($\theta = 14°$) which is in favour for their localization at the interface of O/W emulsions and in the water phase as described by Simovic and Prestidge (2003).

In this case the interparticular silica interactions are driven by hydrogen bounding and lead to the formation of agglomerates related to their high capacity to adsorb water. Nevertheless the specific presence of sodium and chloride atoms inside the core of these CSLM (Fig. 6b and c) shows that ions participate in the hydrophilic silica nanoparticles coagulation in the aqueous phase before interaction with melted emulsion. As Amiri et al. (2009) observed at high pH and high ionic strength it means that cations of electrolytes displaced protons of silanol at the surface of nanoparticles in place of water molecules and provoke coagulation of nanoparticles as bridging agent. High energy of mixing and high temperature can promote and accelerate the diffusion of a fraction of hydrophilic silica nanoparticles in the oil phase through the interface water/fatty alcohol.

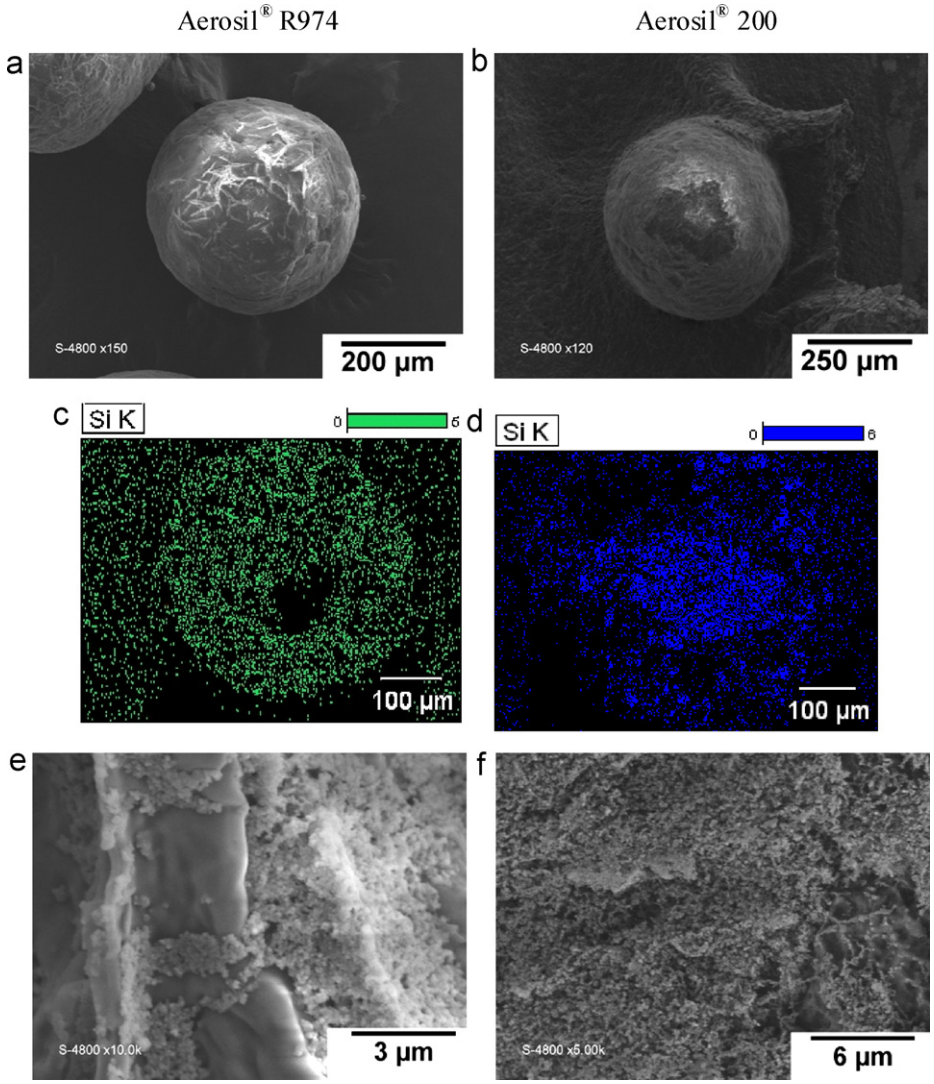

Fig. 5. SEM micrographs of ibuprofen loaded CSLM (MP-S_3-I_5) obtained by dispersion of Aerosil® in the oily phase. Formulation B with Aerosil® R974 (left) Formulation D with Aerosil® 200 (right): (a and b) entire microparticles; (c and d) Si element mapping with EDX microprobe analysis coupled with SEM technique of microparticles cross-section; (e and f) Focus on cross-section of microparticles. B and D are defined in Table 2.

Only hydrophobic *Aerosil® R974* dispersed in the water phase can promote the formation of monodispersed CSLM based on homogeneous silica loaded lipidic core (Fig. 4c and e). In the case of Aerosil® R974 the hydrogen bounding is limited by the low density of silanol reported. The replacement of silanol groups by methyl ones are in favour for interactions particle–particle driven by very weak Van der Waals strength to form in the aqueous phase silica network of hydrophobic silica as described by Forny et al. (2007). The same methyl groups are also in favour for strong interactions with cetyl alcohol (CA) as reported by Simovic and Prestidge (2004). High shear and high temperature lead to total diffusion of aggregated hydrophobic nanoparticles in the oily core of the emulsion. It is interesting to notice that equivalent ibuprofen and silica loading are present in each fraction of different size obtained by sieving CSLM prepared with Aerosil® R974. These data are in favour of homogeneous diffusion of hydrophobic nanoparticles whatever the size of oily droplets (data not shown).

When silica nanoparticles are dispersed in the oily core of the hot emulsion (Fig. 5), it can be noticed that Si elements are more heterogeneously dispersed in CSLM prepared with hydrophilic Aerosil® 200 as compared with formulations containing hydrophobic Aerosil® R974. In fact, silicon spots are concentrated in a large area in the centre of CSLM (Fig. 5d and f) presenting an homogeneous cluster containing only aggregated silica particles. This aggregation of hydrophilic silica nanoparticles in an organic phase as long chain fatty alcohol which is poorly hydrogen bounding liquid, is in agreement with a creation of a network through inter particles hydrogen bonds by the silanol groups on their surface already reported by Sherriff and Enever (1979). At the opposite, this network seems to not exist with Aerosil® R974 at 3% silica loading due to a low density of silanol (0.39 groups nm^{-2}) as compared with a density of 2.6 groups nm^{-2} for Aerosil® 200. In fact, even if there is an aggregation of silica nanoparticles of Aerosil® R974 (Fig. 5c and e) silicon spots are less connected each other.

3.2. Physico-chemical characterization of SLM and CSLM

DSC and X-ray diffraction were used to detect possible modification of the physico-chemical properties of one component of the formulation related to their interactions. DSC thermograms in Fig. 7 compare different thermal characteristics of ibuprofen and cetyl alcohol free species and incorporated in CSLM. The thermogram of ibuprofen (Fig. 8f) gives a sharp single peak at 75 °C with an enthalpy of fusion $\Delta H = 25\ kJ\ mol^{-1}$. Thermogram of CA

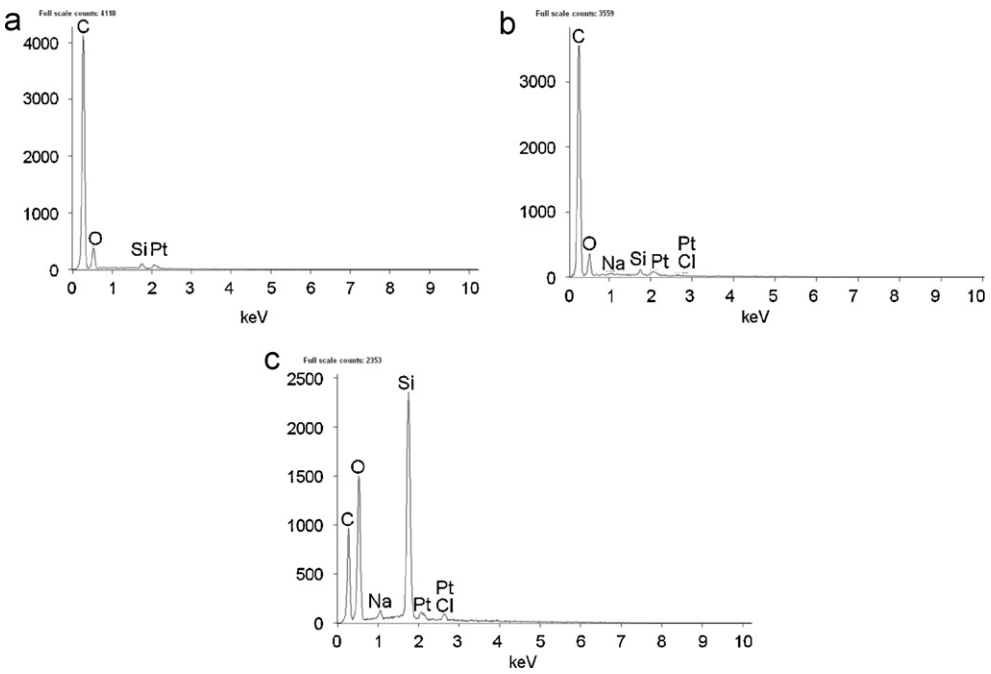

Fig. 6. EDX spectra obtained from formulation D (a) and formulation E (b and c): (b) from cross-section of regular-shape MP; (c) from irregular-shape microparticles. D and E are defined in Table 2.

(Fig. 7a) shows a broad transition between 38 and 50 °C with a double peak characteristic of the two polymorphs of CA already described by Schmid et al. (2000) and a global enthalpy of fusion $\Delta H = 58\ \mathrm{kJ\,mol^{-1}}$. These two values of enthalpy are in agreement with values of the literature (Xu et al., 2004; Xing et al., 2008). As we can see in Fig. 8b, incorporation of ibuprofen in SLM leads to decrease the melt temperature of CA to $\Delta H = 48\ \mathrm{kJ\,mol^{-1}}$ (20% reduction of ΔH value) with a broad transition between 38 and 47 °C in agreement with an interaction drug/lipid. It is not the case with the incorporation of silica nanoparticles, whatever their grade or their way of introduction, on thermograms of ibuprofen loaded or unloaded composite CSLM. In fact, no change in thermograms happens between ibuprofen loaded SLM and CSLM (Fig. 8c–e). These data confirm that in these conditions and at these drug and silica loading the main interaction inside microparticles concerns the drug and the lipid. As it can also be observed, when ibuprofen is melted in presence of CA, its peak disappears from the thermogram of drug loaded microparticles. Finally, the crystallization peak of CA when cooling is at 43 °C as compared with the peak of ibuprofen loaded SLM or CSLM present at 41 °C whatever the grade of silica nanoparticles and the way of process. IR spectra of drug loaded microparticles with or without silica were very similar and interactions concerning silica nanoparticles cannot be ascertain (data not shown).

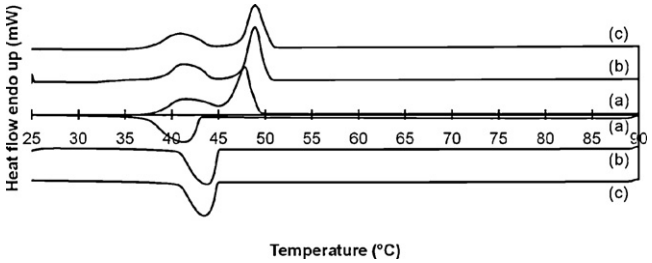

Fig. 7. DSC thermograms of (a) cetyl alcohol; (b) MP-S_0-I_0; (c) MP-S_3-I_0.

Secondly, the X-Ray diffraction spectra (Figs. 9 and 10) give information about the crystalline state of ibuprofen and CA in SLM and CSLM.

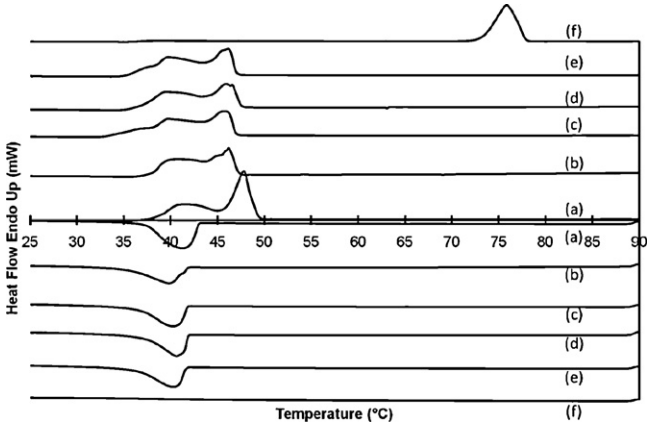

Fig. 8. DSC thermograms of (a) cetyl alcohol; (b) A; (c) C; (d) D; (e) E; (f) pure ibuprofen.

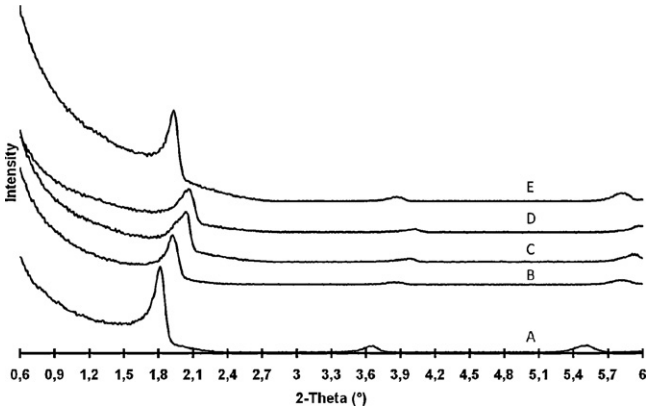

Fig. 9. Small angle X-ray diffraction patterns.

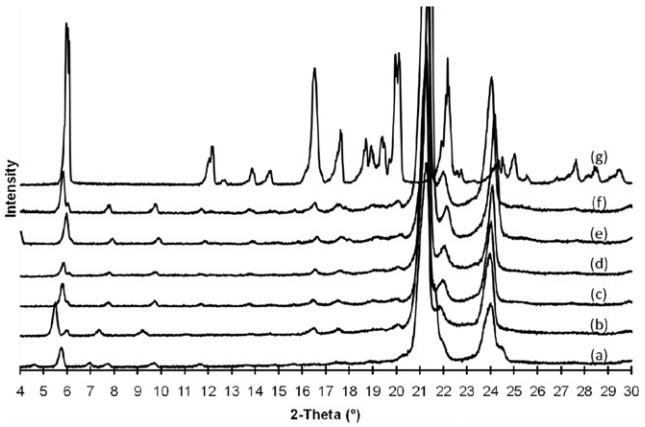

Fig. 10. Wide angle X-ray diffraction patterns of (a) cetyl alcohol; (b) A; (c) B; (d) C; (e) D; (f) E; (g) pure ibuprofen.

At small angle (Fig. 9), XRD spectra exhibit the major harmonic of CA between 1.8° and 2° 2Θ, corresponding to (h; k; l) = (2; 0; 0) and a distance of 46.5 Å. The three peaks of CA are also in good agreement with the EVA software database, corresponding to d(Å) = 46.5, 23.1 and 15.3 respectively for the three harmonics with d_l values in the ratio 1:2:3, suggesting that the CA is in a lamellar phase. Moreover, these three peaks also appear whatever the way of preparations of microparticles. Less intense peaks of their reflection are observed when SLM are drug and silica loaded, corresponding to less ordered structures.

At wide angle, the CA spectrum (Fig. 10a) presents three peaks, one close to 6° 2Θ, one close to 21° 2Θ and the other at 24° 2Θ with shoulders at the right of these last two peaks. The ibuprofen diffractogram (Fig. 10g) presents numerous peaks between 4° and 30° 2Θ, in particularly an intense one close to 6° 2Θ and two peaks between 16° and 18° 2Θ, one double at 20° 2Θ, and an intense but not-well defined one at 22° 2Θ. Crystallinity of ibuprofen was characterized owing to the presence of specific peaks at 16 and 18° 2Θ which could be attributed to crystals of ibuprofen present in all formulations tested. This also supports the hypothesis that for 5 wt% ibuprofen loading the disappearance of the drug melting peak in DSC seems to be due to the dissolution of ibuprofen in the molten lipid. Moreover, the use of PVA or silica nanoparticles to prepare SLM or CSLM respectively (Fig. 10b–f) leads to the disappearance of the shoulders at 21° and 24° 2Θ and the peaks have higher intensities than the respective ones in CA. In addition, a low-intensity peak appears at 22° 2Θ, whatever the sample of CSLM, certainly due to ibuprofen.

3.3. Kinetics of ibuprofen release from SLM and CSLM

Characterization of these microparticles shows us that there is only difference in their size when using different grade of silica. However, different kinetics of ibuprofen release are obtained depending on the presence of silica, the grade of silica and the introduction phase of silica nanoparticles (Fig. 11). The dissolution tests are achieved in continuous flow cells below the limit of solubility of ibuprofen in PBS, pH 7.4 at 37 °C, at a concentration of 25 μg mL^{-1}. In this study the ibuprofen crystals are nearly completely dissolved in 5 h in the PBS ($t_{50\%}$ = 1 h; data not shown). No difference was observed on the release rate of ibuprofen when encapsulated in SLM or encapsulated in CSLM made with Aerosil® 200 introduced in the aqueous phase (Fig. 11a-A and E). With these preparations, the release is total, with a "burst" release kinetic effect ($t_{50\%}$ = 1 h). With the other preparations (B, C and D) there is no more "burst". With incorporation of Aerosil® 200 by the organic phase and Aerosil® R974 by aqueous or organic phase, the release of ibuprofen is total

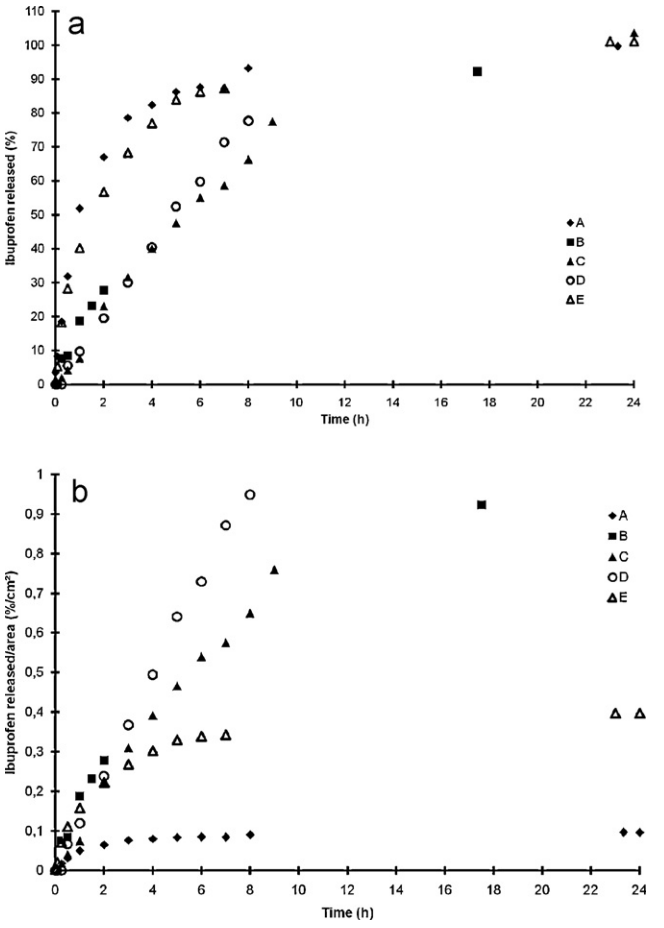

Fig. 11. Kinetics of ibuprofen release from SLM (MP-S_0-I_5) and CSLM (MP-S_3-I_5): (a) expressed as relative rate of release; (b) expressed as a function of geometric area of particles.

and the kinetics is decreased ($t_{50\%}$ = 5 h), compared to silica free microparticles.

The main difference of kinetics of release can be explained by an increase of size of CSLM as compared with SLM and consequently by a reduction of their surface in contact with the aqueous phase (Fig. 11b). The absence of interaction silica nanoparticles – whatever the physico-chemical properties tested – with cetyl alcohol either with ibuprofen clearly shows that Aerosil has no direct action on the drug release from CSLM. Expressed as a function of a fixed surface of microparticles the percentage rate of ibuprofen release is faster with hydrophilic silica and hydrophobic silica loaded CSLM as compared with SLM. In the first hours, it seems that the rate of release does not depend on the nature of Aerosil® or on the way of preparation either. Even hydrophobic Aerosil® homogeneously dispersed aggregated silica nanoparticles does not create hydrophobic barrier able to reduce the kinetics of diffusion of ibuprofen from the core of microparticles to their surface. The better access to water owing to irregular surface or the presence of silica nanoparticles containing silanol on the surface of CSLM could be sufficient to promote an acceleration of water diffusion in the core of microparticles. Similar surface area of Aerosil® 200 and Aerosil® R974 could be the major parameter controlling the release as suggested by Albertini et al. (2004). The exception concerns the reduction of the kinetics of ibuprofen release after 4 h from CSLM obtained by dispersion of Aerosil® 200 in the aqueous phase. It can be explained by the coexistence of two kinds of populations of CSLM.

It is interesting to notice that all the CSLM were spherical and intact after 24 h in dissolution medium PBS; at the opposite we

observed the fusion of SLM in the aqueous phase after 24 h in dissolution medium PBS (data not shown).

4. Conclusion

Composite solid lipid microparticles prepared by hot melt process in conditions of pH and ionic force which promote heterocoagulation of silica nanoparticles represent an original drug delivery system in term of structuring, physical properties (free-flowing and easy to handle particles) and in term of reduction of ibuprofen release from lipidic microparticles.

For the first time surfactant free solid lipid microparticles were obtained by Pickering emulsion route.

We show there that it is possible to prepare surfactant free solid lipid microparticles by hot melt process owing to fumed silica nanoparticles as described with Pickering emulsions. However, CSLM obtained differ from Pickering emulsions because silica, particularly by using Aerosil® R974 is not only adsorbed onto the interface between CA and water, but diffuse inside the lipidic core of microparticles owing to hydrophobic forces to form a homogeneous composite core. The main influence of the incorporation of Aerosil® dispersion in hot emulsion concerns its ability to stabilize the interface melt cetyl alcohol/water and to modulate the surface, the size of CSLM and indirectly the kinetics of ibuprofen release. On the other hand interparticular interactions between silica particles and ibuprofen or cetyl alcohol were not detected on thermic and crystalline state of ibuprofen and lipid profile. Further experiments are needed to know the influence of the ibuprofen and silica loading on the structure of composite microparticles and on the drug release.

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