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International Journal of Pharmaceutics 302 (2005) 187–196
international journal of pharmaceutics
www.elsevier.com/locate/ijpharm

Pharmaceutical Nanotechnology

Development and characterization of sub-micron poly(D,L-lactide-co-glycolide) particles loaded with magnetite/maghemite nanoparticles

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Received 6 January 2005; received in revised form 2 June 2005; accepted 14 June 2005
Available online 11 August 2005

Abstract

Purpose: The objective of this study is to develop biodegradable sub-micron poly(lactide-co-glycolide) particles loaded with magnetite/maghemite nanoparticles for intravenous drug targeting.
Method: Sub-micron magnetite/PLGA particles (also called composite nanoparticles) were prepared by a modified double emulsion method (w/o/w) or by an emulsion-evaporation process (o/w). To optimize the composite nanoparticles formulation, the influence of some experimental parameters, such as types of magnetite/maghemite nanoparticles, volume of magnetite suspension and amount of polymer were investigated. The morphology, size and zeta potential of the magnetite/PLGA nanoparticles were determined. The magnetite entrapment efficiency and magnetite content were assessed by dosing iron in the composite nanoparticles.
Results: TEM photomicrographs showed that the composite nanoparticles were almost spherical in shape with a rather monomodal distribution in size. All composite nanoparticle formulations were found to have the mean diameter within the range of 268–327 nm with polydispersity index within the range of 0.02–0.15. Magnetite nanoparticles coated with oleic acid showed more efficient entrapment (60%) as compared to uncoated magnetite nanoparticles (48%). In both cases, when the volume of magnetite suspension increased, the magnetite entrapment efficiency decreased but the magnetite content increased. In addition, the two-fold rise in the amount of polymer did not significantly affect the composite nanoparticle characteristics except the magnetite content. Finally, none modification of the mean diameter of the composite nanoparticles was observed after storage for 3 months at 4 °C.

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0378-5173/$ – see front matter
doi:10.1016/j.ijpharm.2005.06.024

Conclusions: Magnetite/PLGA nanoparticles were prepared and the influence of some process parameters have been assessed. Improvement of the magnetite entrapment efficiency are in progress and the magnetization properties of the composite nanoparticles will subsequently be tested.

Keywords: Magnetite; Poly(D,L-lactide-co-glycolide); Composite nanoparticles; Double emulsion technique; Magnetite coating

1. Introduction

Both tissue and cell distribution profiles of antitumor drugs can be controlled by their association with submicronic colloidal systems, i.e nanoparticles. The main goal of this approach is to increase antitumor efficacy and to reduce systemic side-effects. If designed appropriately, nanoparticles may act as a drug vehicule to target specifically tumor tissues or cells while reducing their toxicity towards normal tissues. However, after systemic administration, conventional nanoparticles are rapidly opsonized and massively captured by the macrophages of the mononuclear phagocytes system (MPS). Thus, the drug is mainly accumulated in the MPS organs (liver, spleen, lungs and bone marrow) (Grislain et al., 1983). This specific biodistribution can be of benefit for the chemotherapeutic treatment of only MPS-localized tumors as hepatocarcinoma, bronchopulmonary tumors or myeloma (Chiannilkulchai et al., 1989; Gibaud et al., 1994).

Due to the very short half-life (below 5 min) of conventional nanoparticles after intravenous administration (Brigger et al., 2002) targeting anticancer drug to other tumoral tissues is strongly limited. In recent years, several research groups have developped "stealth" nanoparticles which are undetectable by the MPS (Müller and Kissel, 1993; Bazile et al., 1995; Tobio et al., 1998; Gref et al., 2000). These stealth nanoparticles have shown a prolonged half-life in the blood (Gref et al., 1994). Such long-circulating nanoparticles are supposed to be able to directly target tumors located outside the MPS regions.

The aim of the drug targeting consists of carrying the desired amount of drug to the required target and releasing it at a controlled rate. Among the ways to control the targeting specificity, there is a possibility to use the magnetically-guided particles as drug carriers. By application of an external magnetic field, magnetic particles could be retained within a target organ for a given period of time limiting the spreading of the particles in the general circulation. For this purpose, the magnetic particles should be entrapped into a particulate biodegradable polymer matrix to improve the drug loading and the release profile (Chasin and Langer, 1990).

For Arias et al. (2001), a magnetic carrier must have ideally the following properties: (i) small size (below 1 μm) allowing capillary distribution at the desired target site; (ii) appropriate magnetic responsiveness to technically achievable local field in physiological systems; (iii) the ability to carry numerous active drugs at sufficient loadings avoiding administration of excessive magnetizable material; (iv) controllable release rates of the drug from the carrier; (v) minimum toxicity and immunological response; (vi) biodegradability with elimination or minimized toxicity of degradation products.

A few authors have described the preparation of polymeric micro- or nanoparticles containing certain amount of magnetite (Fe_3O_4). Magnetic microparticles consisting of magnetite (Fe_3O_4) particles entrapped in bovine serum albumin microspheres loaded with doxorubicin were first described by Widder et al. (1979). However, bovine serum albumin possess the disadvantage of a possible immune response and synthetic polymers have been progressively used. Dactinomycin (Ibrahim et al., 1982), indomethacin (Malaiya and Vyas, 1988) and methotrexate (Hung et al., 1990) have been successfully incorporated into nanoparticles of PIBCA, PMMA and polyglutaraldehyde, respectively, in presence of magnetite. After intravascular injection of drug loaded nanoparticles and placing a magnet near the target site, significantly higher concentrations of the drugs were observed at the target site (Vyas and Malaiya, 1989). More recently, in order to avoid disadvantges of a possible toxicity and lacking in vivo biodegradability for these polymers, Müller et al. (1996) produced nanoparticles from polylactide (PLA) and polylactide/glycolide (PLGA) loaded with different amounts of magnetite. The nanoparti-

cles obtained had a size comprised between 456 and 890 nm and a theoretical magnetite content up to 50% (w/w). It was shown that the cytotoxicity of these magnetite-loaded polymeric nanoparticles was relatively low qualifying them as potential formulation for intravenous injection with regard to the toxicological acceptance.

Another suitable biodegradable polymer, namely poly(ethyl-2-cyanoacrylate), was chosen by Arias et al. (2001) to be the drug carrier. The method of preparation of magnetite/PE-2-CA (core/shell) nanoparticles was based on the so-called emulsion/polymerization process, often used in the synthesis of PE-2-CA nanospheres designed for drug delivery. The results of this study show that the synthetic new material displays an intermediate behavior between that of the magnetite and PE-2-CA nanospheres. The composite particles obtained have an average diameter of 144 ± 15 nm. It was also demonstrated that all the magnetite particles was covered by a polymer shell when the amount of monomer is well in excess of that of magnetite. Unfortunatly in this case, the appearance of the composite system is that of a film of polymer with magnetic particles embedded in it which was virtually impossible to destroy by non-chemical methods.

The same research group has also produced composite particles formed by a magnetic nucleus (magnetite) coating by a biodegradable poly(D,L-lactide) polymer (Gomez-Lopera et al., 2001). In this case, the process used is the double emulsion technique widely employed to obtain polymeric nanoparticles loaded with hydrophilic drugs. The composite particles obtained exhibit a spherical shape with an average diameter of 180 ± 5 nm with a reasonably narrow size distribution. The magnetic characterization of these mixed colloidal particles show sufficient magnetization for their magnetic properties to be use as magnetic carrier.

Despite the progress of the knowledge in this field, there is few published reports on the magnetite entrapment efficiency in PLA/PLGA nanoparticles. The main problem is how to incorporate magnetite nanoparticles into PLGA nanoparticulate carrier in a high extent. Special attention must be given to the size and composition of the magnetite nanoparticles. Conventionnal magnetite particles are so big that they cannot be incorporated within the biodegradable nanoparticles. An alternative consisting of aqueous magnetite colloidal dispersion (also called ferrofluid) has been developped but the problem is the incompatibility between ferrofluid and PLGA due to the hydrophobic nature of this polymer. To increase the lipophilicity to the ferrofluid, surface modification of the magnetite nanoparticles should be obtained. The goals of the present work were as follows: (1) synthesis and characterization of surface modified superparamagnetic nanoparticles, and (2) formulation and characterization of spherical composite nanoparticles consisting of magnetite/maghemite nanoparticles incorporated in a PLGA nanoparticulate matrix. The influence of several process parameters on the characteristics of these composite nanoparticles was assessed with special attention to the effect of magnetite surface modification on its incorporation into the polymeric nanoparticles. The efficiency of the polymer coating has been studied by analyzing the infrared absorption spectra and by determining the magnetite content of the composite nanoparticles.

2. Materials and methods

2.1. Materials

Poly(D,L-lactide-co-glycolide) (Lactel® BP-0100) was provided by Sigma–Aldrich (St. Quentin Fallavier, France). The lactide:glycolide ratio is 50:50 and the average molecular weight is comprised between 40,000 and 75,000 kDa (data from the manufacturer).

Iron(II) chloride anhydrous ($FeCl_2$), iron(III) chloride anhydrous ($FeCl_3$), oleic acid, ethyl acetate, methylene chloride and polyvinyl alcohol 4–88 (Mw 31,000) were purchased from Sigma–Aldrich (St. Quentin Fallavier, France) and used as received.

2.2. Synthesis of iron oxides nanoparticles

The magnetite/maghemite nanoparticles (MagNP) were prepared according to the method of Massart as previously described (Massart, 1981). Briefly, ferrous and ferric chlorides were dissolved in 1 N HCl in order to obtain an acidic solution. This mixture was added dropwise to a solution of 1 N KOH under N_2 atmosphere leading to a black precipitate. The precipitate was isolated and washed several times with deionized water until the pH of the medium was around 8. After

magnetic decantation, the solid fraction was acidified with 2 M perchloric acid and further oxidized by ferric nitrate at 80 °C for 1 h. To remove the salts in excess, the particles were brought to flocculation by addition of a solution of tetramethylammonium hydroxide and washed with water. Then, the precipitate was stirred in diluted nitric acid, washed several times with acetone and finally peptized in water. The final iron oxides nanoparticles were kept in the dark at 4 °C until further use.

In some set of experiments, an ethanolic solution of oleic acid was added under gentle stirring to the precipitate after washing with deionized water. Finally, the oleic acid-treated magnetite/maghemite nanoparticles (OA-MagNP) were washed several times by acetone and redispersed in methylene chloride. In this case, the tendancy of the particles to agglomerate is lowered and the lipophilicity of the magnetite/maghemite nanoparticles is increased (Montagne et al., 2002; Igartua et al., 2002).

2.3. Preparation of composite nanoparticles

The preparation of the first type of composite nanoparticles made with MagNP and PLGA was performed by using the double emulsion (w/o/w) technique previously applied to the encapsulation of proteins into nanospheres with slight modifications (Blanco and Alonso, 1997). In order to stabilize the dispersed phase, a surfactant is required and poly(vinyl alcohol) is one of the most commoly used polymer surfactant. Typically, 50 μl of MagNP aqueous suspension (11.6 mg Fe/ml) were emulsified in an organic solution of the polymer (100 mg PLGA in 2 ml methylene chloride) by sonication (Branson Sonifier® 150) for 15 s (15 W) in an ice bath, to obtain an (aqueous suspension of magnetite)-in-oil emulsion. Then, 4 ml of an aqueous PVA solution (3% (w/v)) were added to this first emulsion and the mixture was sonicated again for 30 s (15 W). The resulting double emulsion was diluted in 50 ml aqueous PVA solution (0.3% (w/v)) under mechanical stirring and the solvent was rapidly eliminated by evaporation under reduced pressure.

Unloaded PLGA nanoparticles were prepared in the same way, except that the MagNP aqueous suspension was substituted by the same volume of deionized water.

The procedure followed to prepare the second type of composite nanoparticles from OA-MagNP and PLGA was the so-called simple emulsion-evaporation process. Briefly, 50 μl of OA-MagNP suspension (12.8 mg Fe/ml) in methylene chloride were mixed with an organic solution of the polymer (100 mg PLGA in 2 ml methylene chloride) by vortexing. This organic mixture was then emulsified by sonication in 4 ml of an aqueous PVA solution (3% (w/v)) in the same conditions described above. The resulting simple emulsion was diluted in 50 ml aqueous PVA solution (0.3% (w/v)) under mechanical stirring and the solvent was rapidly eliminated by evaporation under reduced pressure.

2.4. Purification of composite nanoparticles

After solvent removal, the composite nanoparticles were isolated by centrifugation at $1000 \times g$ for 1 h at 4 °C. In these conditions, free magnetic nanoparticles could be separate from the composite nanoparticles. The latter were finally washed three times with deionized water by centrifugation at $15{,}000 \times g$ for 20 min at 4 °C and kept in the dark at 4 °C until required.

2.5. Physico-chemical characterization of magnetite/maghemite and composite nanoparticles

2.5.1. Particle size and morphology

The morphological examination of magnetite/maghemite nanoparticles and composite nanoparticles were performed using an electronic transmission microscope at 88 kV (JEOL 1010, Jeol, Japan). The samples were placed on a carbon coated copper grid and stained with 3% (w/v) uranyl acetate for TEM viewing.

The mean hydrodynamic diameter and polydispersity index were determined by photon correlation spectroscopy (PCS) using a Malvern Autosizer® 4700 (Malvern Instruments Ltd., Malvern, UK) with photodetector perpendicular to the laser beam. The zeta potential measurements were determined by laser Doppler velocimetry (LDV) using a Malvern Zetasizer® 3000 (Malvern Instruments Ltd., Malvern, UK). In the two cases, all samples were sonicated and then diluted in deionized water before measure-

ment. Thus, the mean of the nanoparticle size distribution as well as the mean of the zeta potential distribution values are the average of three determinations.

2.5.2. Chemical composition of particles

Molecular composition of dehydrated iron oxide nanoparticles was studied using Raman microspectrometry (Labram microspectrometer, Jobin Yvon-Horiba, France).

Adsorption of oleic acid on the surface of iron oxides nanoparticles was examined by Fourier transform-infrared spectrometry (Bruker Vector® 22 spectrometer, Bruker, Germany). Dried MagNP and OA-MagNP were mixed and pressed with KBr to obtain small pellets. FT-IR spectrometry was also used for the chemical characterization of the two types of composite nanoparticles.

2.5.3. Iron oxides entrapment efficiency

The amount of magnetite incorporated into the composite nanoparticles was determined after dosing iron by atomic absorption spectrophotometry (AAS) (SpectrAA-10 Plus, Varian, France). Extraction of iron was achieved after the destruction of the nanoparticles by incubation with 10 ml of 6 N HCl until complete hydrolysis of the polymer and yellow coloring appeared. One gram of the resulting solution was diluted with HCl 1% (v/v) and assayed by AAS using a calibration curve. Magnetite entrapment efficiency and magnetite content in the composite nanoparticles were calculated by Eqs. (1) and (2) respectively.

Magnetic entrapment efficiency (%):

$$= \frac{\text{mass of magnetite in composite nanoparticles}}{\text{mass of magnetite used in formulations}} \times 100 \quad (1)$$

Magnetite content in composite nanoparticles (% (w/w)):

$$= \frac{\text{mass of magnetite in composite nanoparticles}}{\text{mass of magnetite + mass of PLGA used in formulations}} \times 100 \quad (2)$$

3. Results and discussion

3.1. Characterization of iron oxides and composite nanoparticles

Fig. 1 displays the physical appearance of iron oxides nanoparticles as observed in TEM. These MagNP exhibit a rather spherical shape and are relatively monodisperse with a mean diameter of 8 ± 2 nm. The hydrodynamic size distribution profiles exhibited a single sharp maximum at 20–22 nm (data not shown). According to the Raman spectrometry data, the dehydrated nanoparticles obtained from the co-precipitation step of the Massart's protocol were constituted of magnetite (Fe_3O_4) and maghemite (γ-Fe_2O_3) and were void of any detectable presence of other iron oxides or oxy-hydroxides (data not shown). The relative content of magnetite and maghemite was quantitatively established from their characteristic frequencies in Raman spectra (Chourpa et al., 2005). The results averaged over at least five independent preparations of the iron oxides nanoparticles indicated the molecular fraction of about 72% magnetite and 28% maghemite. The suggested fine structure of individual particles should include magnetite in the inner layers and maghemite in the outer layers.

Considering together the described chemical composition (magnetite/maghemite) and the small size of the iron oxides nanoparticles (<10 nm), one can suppose them to be superparamagnetic, i.e. void of mag-

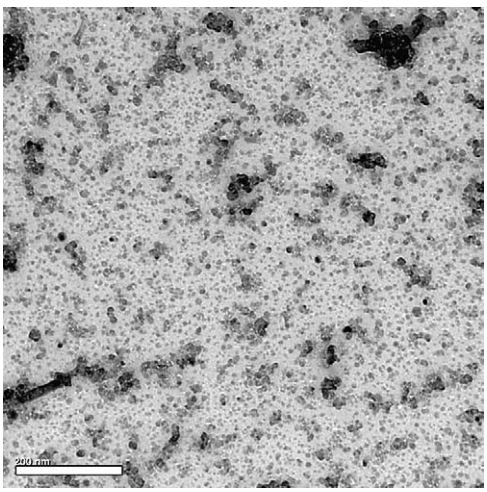

Fig. 1. TEM photomicrograph of MagNP.

netic remanence. Although the magnetic properties of these particles will be a subject of further studies, it is noteworthy that rather strong magnetisation of the ferrofluids was observed upon action of a stationary magnet.

The infrared spectrum of iron oxides nanoparticles exhibit a strong band at 588 cm^{-1} which is consistent either with magnetite (Fe_3O_4) spectrum (band, 570 cm^{-1}) or maghemite (γ-Fe_2O_3) spectrum (broad band, 520–610 cm^{-1}) (data not shown). As reported previously in the literature (Montagne et al., 2002), the adsorption of oleic acid on iron oxides surface can be evidenced by infrared spectrometry. The complexation reaction between iron atom and the carboxylate groups of oleic acid is usually characterized by two bands at 1425 and 1520 cm^{-1}. The spectrum obtained with OA-MagNP showed two bands at 1414 and 1531 cm^{-1} that could be attributed to the oleic acid adsorbed on the magnetite/maghemite nanoparticles surface (data not shown). Finally, the infrared spectra of the composite nanoparticles showed the presence of all the bands of the polymer and a weak peak at 582 cm^{-1} that could be attributed to the magnetite/maghemite nanoparticles.

The morphology of the composite nanoparticles was also studied by TEM. Fig. 2a shows a network of spherical shape composite nanoparticles with a diam-

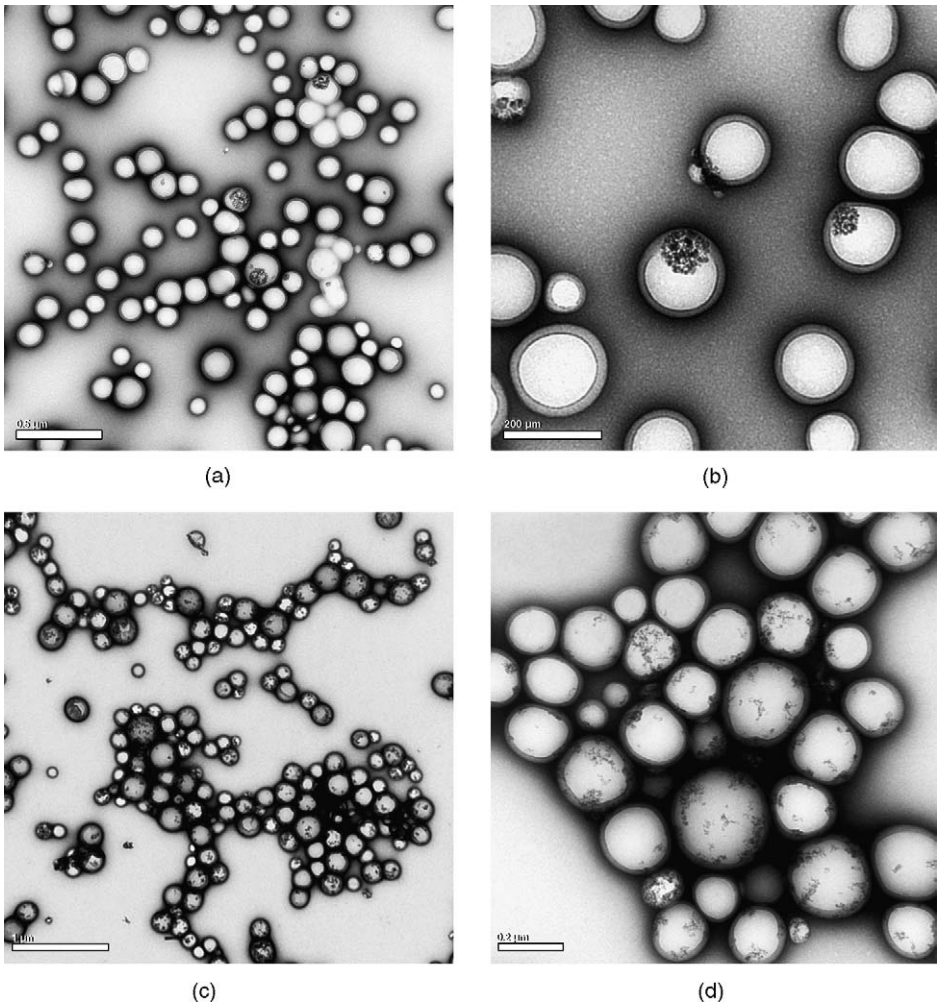

Fig. 2. TEM photomicrographs of composite nanoparticles: top, composite nanoparticles prepared with MagNP (a) lower magnification and (b) higher magnification; bottom, composite nanoparticles prepared with OA-MagNP (c) lower magnification and (d) higher magnification.

eter below 0.5 μm. It should be noticed some aggregates of MagNP in a relatively low number of composite nanoparticles but no free MagNP (Fig. 2b). The presence of such large clusters is probably due to the ease of aggregation of the magnetite nanoparticles during the encapsulation process leading to a poor incorporation of magnetic material in the polymer matrix and to make difficult the formation of individual magnetite nanoparticle covered with a PLGA shell. In contrast, in composite nanoparticles prepared with OA-MagNP and PLGA, the magnetite nanoparticles appear to be more or less uniformly distributed in the polymer matrix without such evidence of aggregates of magnetic material (Fig. 2c and d). This could be explained by the more lipophilic tendancy of the OA-MagNP allowing their better incorporation inside the PLGA nanoparticles. In addition, the diameter is not significantly different from the other composite nanoparticles.

3.2. Formulation optimisation

With the final goal to entrap a lot of magnetite/maghemite nanoparticles in PLGA nanoparticulate matrix, various formulation parameters were investigated. Two types of magnetite/maghemite nanoparticles (MagNP and OA-MagNP) with different lipophilicity were used. The volume of magnetite suspension was varied from 50 to 200 μl and the amount of polymer was increased from 100 to 200 mg for preparing the composite nanoparticles. The effect on the nanoparticles characteristics (mean diameter and polydispersity index, zeta potential and magnetite incorporation efficiency) were assessed.

The nanoparticles mean diameters from all the formulations were very close and were in the range of 268–327 nm with polydispersity index in the range of 0.02–0.15 indicating a narrow size distribution (data not shown). The results of increasing the amount of polymer did not significantly affect composite nanoparticles size distribution. Besides, an increase in the internal aqueous phase could lead to an increase in loading, which is a critical parameter in the (w/o/w) emulsion process. Thus, a four-fold rise in the volume of the magnetite aqueous suspension (i.e. 200 μl instead of 50 μl) gives particles with similar characteristics, regarding their mean diameter and polydispersity index. In addition, the type of magnetite/maghemite nanoparticles used (i.e. Mag-NP or OA-MagNP) did not alter the composite nanoparticles size distribution.

The zeta potential of the composite nanoparticles (-18.2 ± 0.7 mV) was similar to that of blank nanoparticles (-24.7 ± 1.8 mV) and was not changed by the various manufacturing conditions. This result shows that no free magnetite can be detected by zeta potentiometry.

In order to avoid the use of methylene chloride which is not recommended for the preparation of parenteral dosage form, some batches of composite nanoparticles have been prepared with ethyl acetate which is a good solvent for the polymer and much less toxic than methylene chloride (Sanchez et al., 1999). The mean diameter and polydispersity index of the nanoparticles were in the same order of magnitude (with a slight reducing of size) than those of the nanoparticles prepared with methylene chloride. This result is interesting for the preparation of composite nanoparticles for in vivo study.

When MagNP were used in preparing nanoparticle formulation, the magnetite entrapment efficiency was less than 50% (Fig. 3a). This was mainly due to the hydrophilic nature of the MagNP aqueous suspension which caused moderate magnetite retention in the polymer matrix. To increase the magnetite loading in the nanoparticles, the volume of MagNP aqueous suspension in the formulations was increased from 50 to 200 μl maintaining the amount of polymer constant. Nevertheless, it can be observed a decrease in the magnetite entrapment efficiency. Similar results have been reported by Saxena et al. (2004) for the incorporation of high hydrophilic indocyanine green dye in PLGA nanoparticles. In the double emulsion-evaporation process, the critical step is the dispersion of the aqueous phase containing the hydrophilic compound in the organic solution of the polymer material. Increasing the volume fraction of the internal aqueous phase lowered the encapsulation efficiency due to droplet coalescence and increased probability of contact between the internal compound solution and the external extraction phase resulting in compound loss. In addition, if the amount of polymer in the formulations was increased by two times, the magnetite entrapment efficiency remained in the same values.

It is well known that the hydrophobic nature of the PLGA was not favourable for the entrapment of hydrophilic compounds. In order to enhance the mag-

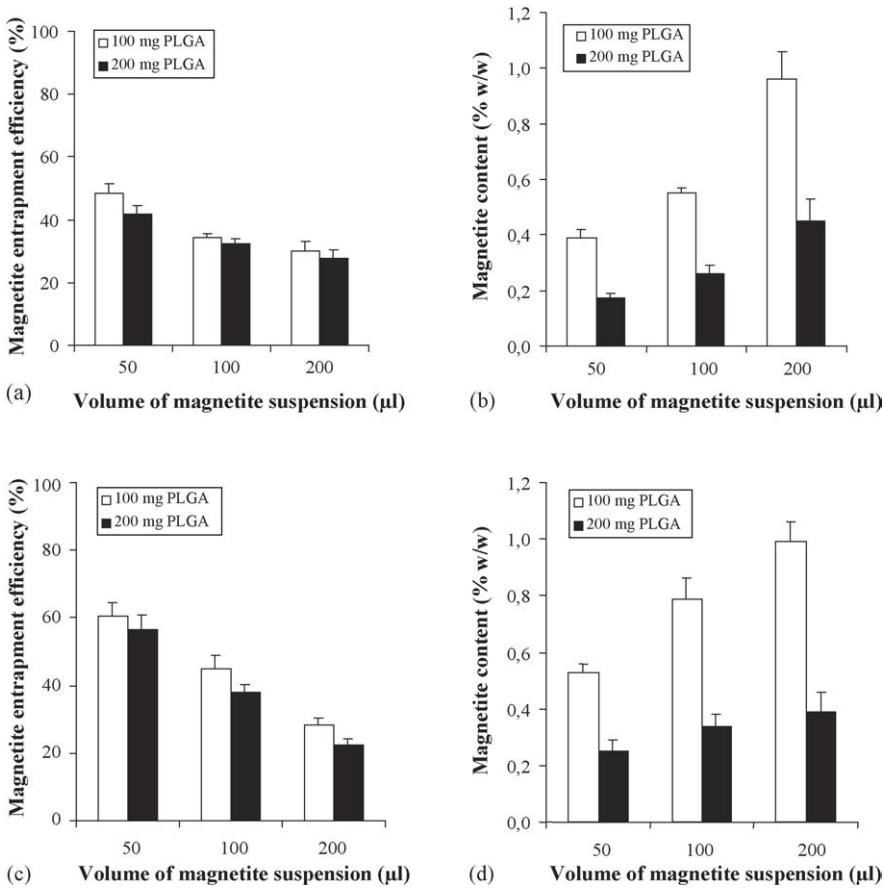

Fig. 3. Magnetite entrapment efficiency and magnetite content of composite nanoparticles as a function of the volume of magnetite suspension and the amount of polymer (a and b, formulations with MagNP; c and d, formulations with OA-MagNP).

netite/polymer affinity and consequently to increase the incorporation of the magnetite into the PLGA nanoparticles, MagNP were treated with oleic acid in order to obtain more lipophilic nanoparticles (OA-MagNP). This procedure would allow us to introduce directly the OA-MagNP into the polymer organic solution without the first sonication step. The results (Fig. 3c) showed an increase up to 60% in the magnetite entrapment efficiency but a decrease when the volume of OA-MagNP suspension increase. In addition, even with the OA-MagNP, the use of the two-fold amount of PLGA did not increase the magnetite entrapment efficiency above 60%.

Despite this fact, it is interesting to determine the magnetite content both for the composite nanoparticles made with the MagNP (Fig. 3b) or made with the OA-MagNP (Fig. 3d). It could be noted that the magnetite content increase from 0.4 to 1% (w/w) and from 0.5 to 1% (w/w) for the composite nanoparticles made with the MagNP and the OA-MagNP, respectively, when the volume of magnetite suspension increase. Similar result has been reported for the nanoencapsulation of a model protein (namely human serum albumin) in poly(lactic acid) nanoparticles (Zambaux et al., 1998). It could be concluded that a sufficient amount of magnetite suspension was needed in order to obtain high magnetite content. In contrast, when the polymer amount was increased up to 200 mg, the values of the magnetite content were approximately divided by two. This fact remains unclear but it could suggested that as the magnetite loss is the same whatever the amount of PLGA used, the same amount of

Table 1
Evolution of mean diameter and polydispersity index of various composite nanoparticles stored at 4 °C (batch I prepared with MagNP; batches II and III prepared with OA-MagNP)

Batch number	Magnetite suspension (μl)	Mean diameter ± S.D. (nm)[a] $T = 0$	Polydispersity index	Mean diameter ± S.D. (nm)[a] $T = 3$ months	Polydispersity index
I	100	270 ± 5	0.13	258 ± 6	0.01
II	50	263 ± 4	0.04	270 ± 3	0.06
III	200	272 ± 5	0.02	276 ± 4	0.07

All formulations made with 100 mg PLGA.
[a] $n = 3$.

magnetite is incorporated in two-fold more quantity of polymer.

In some experiments, the washing procedure of the composite nanoparticles was slightly modified by replacing deionized water with an aqueous solution of acetic acid (1 M) to evaluate if the magnetite nanoparticles were not associated to the PLGA nanoparticles by a simple adsorption process. The magnetite content and entrapment efficiency were not significantly affected by this procedure that is in favour of an incorporation of the magnetite nanoparticles in the polymer matrix (data not shown).

3.3. Stability study

Three batches of composite nanoparticles made with MagNP and OA-MagNP were stored 3 months at 4 °C and their physical stability was assessed by determination of the mean diameter of the nanoparticles. In all cases, there is no evolution of the size of the nanoparticles during the time of the study (Table 1). It could be deduced that no aggregation occurs during the storage and the formulations exhibit a good stability.

4. Conclusion

We have reported in this work a process to obtain the incorporation of modified magnetite/maghemite nanoparticles into PLGA nanoparticulate matrix. Morphological aspect in TEM, FT-IR spectra, zeta potential measurements and magnetite content of the produced composite nanoparticles give strong evidences of the presence of magnetite nanoparticles within the polymer matrix. These composite nanoparticles were nearly spherical in shape with a mean diameter below 300 nm. We have shown that the coating of the magnetite/maghemite nanoparticles with a fatty acid enhances the magnetite entrapment efficiency and that the magnetite content increases when increasing the amount of magnetite. The coating procedure allow us to incorporate the coated magnetite/maghemite nanoparticles into the organic polymer solution without an emulsification step. The hydrophobic nature if these nanoparticles would facilitate their incorporation in the polymer nanoparticulate matrix and could be explain by hydrophobic interactions between the two compounds.

On the basis of these reported preliminary data, the presently described composite nanoparticles might constitute a promising formulation for magnetic drug targeting and consequently could be an interesting carrier for site specific anticancer drug delivery. To check this latter point, the magnetization properties of the composite nanoparticles will subsequently be tested.

Acknowledgements

The authors are very grateful to Prof. F. Boury (INSERM U646, University of Angers, France) for his assistance with Photon Correlation Spectroscopy and Zeta potential analysis and Drs B. Arbeille and P.-Y. Sizaret (PPF Analyse des Systèmes Biologiques, University of Tours, France) for their assistance in Transmission Electronic Microscopy.

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