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Pharmaceutical nanotechnology

Safety and efficacy of amphiphilic ß-cyclodextrin nanoparticles for paclitaxel delivery

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Abstract

Paclitaxel is a potent anticancer agent with limited bioavailability due to side-effects associated with solubilizer used in its commercial formulation and the tendency of the drug to precipitate in aqueous media. In this study, paclitaxel was encapsulated in amphiphilic cyclodextrin nanoparticles. Safety of blank nanoparticles was compared against commercial vehicle cremophor:ethanol (50:50 v/v) by hemolysis and cytotoxicity experiments. Data revealed that nanoparticles caused significantly less hemolysis. Results were confirmed with SEM imaging of erythrocytes treated with nanospheres, nanocapsules or commercial vehicle. Cytotoxicity of the blank carriers was evaluated against L929 cells. A vast difference between the cytotoxicity of nanoparticles and cremophor:ethanol mixture was observed. Physical stability of paclitaxel in nanoparticles was assessed for 1 month with repeated particle size and zeta potential measurements and AFM imaging. Recrystallization of paclitaxel, very typical in diluted aqueous solutions of the drug, did not take place when the drug is bound to cyclodextrin nanoparticles. Anticancer efficacy of paclitaxel-loaded nanoparticles was evaluated in comparison to paclitaxel in cremophor vehicle against MCF-7 cells. Cyclodextrin nanoparticle caused a slightly higher anticancer effect than cremophor:ethanol vehicle. Thus, amphiphilic cyclodextrin nanoparticles emerged as promising alternative formulations for injectable paclitaxel administration with low toxicity and equivalent efficacy.
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Keywords: Paclitaxel; Amphiphilic cyclodextrin; Nanoparticle; Hemolysis; Cytotoxicity; Stability

1. Introduction

Paclitaxel (PCX) is a major anticancer drug first isolated form the bark of *Taxus brevifolia* with a unique mechanism of action promoting the assembly of microtubules from tubule dimers and prevents them from depolarizing. This results in the loss of normal microtubule dynamics necessary for cell division and other vital processes and consequently causes cell death. The ability of this drug to stabilize microtubules makes it significantly effective against various types of solid tumors including breast cancer, advanced ovarian carcinoma, lung cancer, head and neck carcinomas and acute leukemias (Rowinsky, 1993; Spencer and Foulds, 1994). Unfortunately, the poor water solubility of PCX impairs its bioavailability and its clinical application due to the use of solubilizer Cremophor EL in current commercial product in mixture with ethanol (50:50 v/v). Severe side effects such as hypersensitivity reactions, nephrotoxicity and neurotoxicity as well as effects on endothelial and vascular muscles causing vasodilatation, labored breathing, lethargy and hypotension along with reported incompatibility with PVC infusion sets used to administer paclitaxel during chemotherapy (Weiss et al., 1990; Kongshang et al., 1991; Dorr, 1994; Waugh et al., 1997; Gelderblom et al., 2001). To eliminate Cremophor EL from PCX administration, alternative carrier systems free of Cremophor have been suggested including parenteral emulsions, liposomes, nanoparticles, water-soluble drugs and conjugates and inclusion complexes with cyclodextrins (Sharma and Straubinger, 1994; Cserhati and Hollo, 1994; Sharma et al., 1995; Lundberg, 1997; Pendri et al., 1998; Fonseca et al., 2002; Alcaro et al., 2002).

One of the alternative PCX formulation approaches administration of poorly soluble anticancer drugs in nanoparticulate carrier systems seems to be advantageous due to reduced size of these delivery systems (<400 nm) which help escape from RES uptake and facilitate the extravasation through the leaky vasculature typical of the tumor site as well as the avoidance of Cremophor as the solubilizer. This is believed to reduce

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side-effects and increase therapeutic efficacy while altering the tissue distribution and the pharmacokinetics of the anticancer drug (Brigger et al., 2002). It has also been demonstrated that nanoparticles may overcome MDR type P-gp increasing the drug content inside the neoplastic cells. This may also help increase the therapeutic efficacy of paclitaxel since an acquired resistance to the drug has already been reported (Bennis et al., 1994; Rowinsky and Donehower, 1995). In fact, in the light of this approach it is noteworthy that Abraxane®, a Cremophor-free, albumin-bound nanoparticle paclitaxel (∼130 nm) has recently been approved by the FDA by January 7, 2005 for the chemotherapy of recurrent metastatic breast cancer (Sparreboom et al., 2005) which is reported to increase the maximum tolerated dose in different therapy regimens.

Amphiphilic cyclodextrins (CDs) are chemical derivatives of natural β- and γ-cyclodextrins by selective substitution of aliphatic chains of different structures (2–18C, linear or branched, linked with ester, ether, amide, thio, fluoro bonds (Wouessidjewe et al., 1995; Memisoglu-Bilensoy et al., 2006a). These cyclodextrin derivatives were demonstrated to yield nanospheres or nanocapsules spontaneously using the nanoprecipitation technique with or without the presence of surfactants (Memisoglu et al., 2002). Some amphiphilic β-cyclodextrin derivatives such as β-CDC6 modified on the secondary face with 6C aliphatic esters and 6-*N*-CAPRO-β-CD modified on the primary face with a 6C aliphatic amide were demonstrated to give stable nanoparticles of high drug loading capacity and reduction of burst effect during the drug release process when nanoparticles are prepared directly from pre-formed drug:amphiphilic CD inclusion complex (Memisoglu et al., 2003a, b). Amphiphilic cyclodextrins were reported to be non-hemolytic and non-cytotoxic according to studies on human blood samples and L929 mouse fibroblast cells regardless of their different chemical structures (Memisoglu et al., 2003b; Memisoglu-Bilensoy et al., 2006b).

The objective of this study was to compare the safety and efficacy of novel, non-surfactant, paclitaxel-loaded amphiphilic cyclodextrin nanoparticles as an alternative to cremophor vehicle in terms of hemolysis, cytotoxicity against L929 cells, physical stability of paclitaxel in nanoparticle form and tendency to precipitate in particular and anticancer efficacy against MCF-7 cell line. The highest possible concentrations of nanoparticles and commercial vehicle were used in equal doses for a healthy comparison representing the most exaggerated conditions.

2. Material and methods

2.1. Materials

Paclitaxel (MW: 853.9 g/mol, aqueous solubility at 37 °C: 3.5 μg/mL, solubility in ethanol: 39 mg/mL) was a kind gift from Zhejiang Pharmaceuticals (China) (EA648838320CN). Amphiphilic β-cyclodextrin, 6-O-CAPRO-β-CD, modified on the primary face with 6C aliphatic ester has been synthesized, purified and characterized as reported previously (Memisoglu et al., 2002) (MW: 1820 g/mol, solubility in alcohol at 25 °C: 3.6 mg/mL, solubility in acetone at 25 °C: 3.6 mg/mL; calculated HLB value: 11.1) Miglyol 812®, a neutral oil composed of triglycerides of the fractionated vegetable fatty acids C8 and C10, was purchased from Condea Chemie, Germany. Cremophor®EL used to solubilize PCX was purchased from BioChemika/Fluka, Buchs, Belgium. Ultrapure water MilliQ (Millipore Simplicity 185, Molsheim, France) was used in the preparation of nanoparticles. All organic solvents were of HPLC grade and were used without further purification.

2.2. Preparation of PCX-loaded nanoparticles

6-*O*-CAPRO-β-CD nanoparticles were prepared according to the nanoprecipitation technique (Memisoglu-Bilensoy et al., 2005) as follows; nanospheres were prepared by the addition of an organic phase of 1 mL consisting of amphiphilic cyclodextrin (1 mg) dissolved in acetone to an aqueous phase of 2 mL consisting only of ultrapure water under ambient temperature with constant stirring. Then the organic solvent residue is evaporated under vacuum to the desired volume (2 mL) of nanosphere dispersion.

To prepare nanocapsules of 6-*O*-CAPRO-ß-CD, the same technique was employed with difference of presence of an oil, Miglyol 812® (50 μL) in the organic phase.

Nanospheres and nanocapsules were loaded with active ingredient PCX with two different approaches previously reported (Memisoglu et al., 2003a,b). These approaches are briefly as follows;

- *High loaded* (*HL*) *nanospheres* were prepared directly from pre-formed inclusion complexes of PCX:6-*O*-CAPRO-β-CD of 1:1 molar ratio. Organic phase in this case consisted of 1 mg of PCX:6-*O*-CAPRO-ß-CD complex and excess drug solution (200 μL) in acetone added to aqueous phase of ultrapure water.
- *Conventionally loaded* (*CL*) *nanoparticles* were prepared by the addition of drug solution (200 μL) to organic phase (1 mg amphiphilic cyclodextrin in 1 mL acetone) during preparation.

2.3. Safety of blank amphiphilic cyclodextrin (6-O-CAPRO-β-CD) nanoparticles

2.3.1. Hemolysis studies

Blood samples were drawn from healthy human subjects and separated by centrifugation to isolate erythrocytes (RBC). For the isolation process, erythrocytes were separated by centrifugation, washed two times with isotonic phosphate buffer pH 7.4 and then resuspended in this buffer in a ratio of 1–5 (erythrocytes:PBS).

1 mL unloaded 6-*O*-CAPRO-β-CD nanosphere or nanocapsule dispersion prepared in isotonic PBS was obtained from varying concentration range of amphiphilic β-CD from 0.1–0.50 mM covering the actual amphiphilic β-cyclodextrin range used in nanoparticle formulations. Nanoparticle dispersion was treated with 50 μL blood sample (RBC suspension) and vortexed. Samples were incubated at 37 °C for 30 min

and centrifuged at 5000 rpm for 10 min. Supernatant was analyzed for hemoglobin content at 543 nm spectrophotometrically (Shimadzu UV 160-A, Japan). For the commercial vehicle cremophor:ethanol mixture, 1 mL of Cremophor®EL:anhydrous ethanol mixture of 50:50 v:v was used and treated with the same procedure as the nanosphere and nanocapsule dispersions. Total hemolysis (100%) was determined from the UV absorbance of samples treated with distilled water leading to total lysis of erythrocytes. Hemolysis % was the calculated as a function of UV absorbance of total hemolysis according to the following equation:

$$\text{Hemolysis}\% = \left(\frac{\text{UV absorbance of sample treated with RBC}}{\text{UV absorbance of sample treated with distilled water}}\right) \times 100$$

Hemolytic activity of amphiphilic cyclodextrin nanoparticles on erythrocytes were also confirmed with scanning electron microscopy according to the following experimental technique: the prepared nanoparticle or cremophor:ethanol vehicle was treated with 50 μL RBC suspension and vortexed. Samples were incubated at 37 °C for 30 min. Following the incubation time, samples were diluted with Sorensen's Phosphate Buffer (SPB) and fixated for 1 h in 2% glutaraldehyde solution. After the fixation period, samples were centrifuged at 3500 rpm for 5 min and the precipitate was washed with SPB. After a second centrifugation under the same conditions, resulting precipitate was spread on metal plates and dried in a dustless environment. Dry powder was coated with gold–palladium mixture with a thickness of 100 Å using a HUMMER VII Sputter Coating Device, Anatech, and further evaluated by JEOL JSM-6400 Electron Microscope, Japan.

2.3.2. Cytotoxicity of blank nanoparticles against L929 cells

Blank nanoparticles of 6-*O*-CAPRO-ß-CD were prepared according to the nanoprecipitation technique and final dispersion of nanoparticles of 2 ml volume were used for the cytotoxicity studies of blank nanoparticles on L929 mouse fibroblast cells. Commercial vehicle used in cytotoxicity studies consisted of Cremophor®EL and anhydrous ethanol mixture of 50:50 v/v. The volume ratio of sample to cell culture medium was kept to 50 μL of nanoparticle or commercial vehicle to 50 μL of cell culture medium.

L929 mouse fibroblasts (ATCC) were cultured in 25 cm^2 culture flasks containing RPMI 1640 supplemented with heat inactivated 10% fetal bovine serum, 2 mM L-glutamine, 100 units mL^{-1} penicillin G and 100 μg mL^{-1} streptomycin at 37 °C in a humidified incubator containing 5% CO_2. Confluent cell monolayers were trypsinized and cells in exponentially growing phase were used in cytotoxicity experiments.

The methlythiazolyltetrazolium (MTT) assay was used to evaluate cell viability (Hansen et al., 1989; Campling et al., 1988). Briefly, 50 μl cell suspensions containing 2×10^5 leukocytes or 10^3 L929 cells were seeded in 96-well plates (Costar, Cambridge, MA) and 50 μl of diluted material were added into each well. Flat-bottomed 96-well plates were utilized for L929 cells whereas round-bottomed plates were used for leukocytes. After 72 h of incubation, 25 μl of MTT solution (1 mg/ml final concentration) (Sigma Chemical Co., St. Louis, MO) were added to each well, and the plates were incubated for a further 4 h. The formazan precipitate was solubilized by adding 80 μl lysing buffer (pH = 4.7) composed of 23% SDS dissolved in a solution of 45% *N*,*N*-dimethylformamide. After an overnight incubation at 37 °C, the optical densities (OD) were read at 570 nm using a microplate reader (Spectramax Plus, Molecular Devices, Sunnyvale, CA, USA). Cells incubated in culture medium alone served as a control for cell viability (nontreated wells). Cell viability (%) was calculated as (OD of treated wells/OD of nontreated cells) × 100.

2.3.3. Physical stability of PCX in nanoparticles

2.3.3.1. Particle size measurements. Repeated particle size measurements were performed on fresh aqueous nanoparticle dispersion samples and after 1 day, 1 week and 1 month storage at 8 °C refrigerator temperature using Malvern Zetasizer Nano-ZS, Malvern Instruments, UK at 25 °C in triplicate based on Quasi-elastic light scattering QELS technique. Nanoparticle dispersion samples stored at 8 °C refrigerator temperature were diluted to adequate sample intensity for an efficient particle size distribution measurement. Particle size distribution was expressed as mean diameter (nm) ± standard deviation and polydispersity index.

2.3.3.2. Zeta potential measurements. Repeated zeta potential values were determined at time intervals of 0, 1, 7 and 30 days using Malvern Zetasizer Nano-ZS, Malvern Instruments, UK at 25 °C and 12° angles in triplicate. Nanoparticle dispersion samples stored at 8 °C refrigerator temperature were diluted to adequate sample intensity for an efficient zeta potential measurement. Zeta potential (mV) was expressed as the average of three measurements.

2.3.3.3. Atomic force microscopy. Nanoparticle samples stored at 8 °C refrigerator temperature were imaged after predetermined storage periods using the Q-Scope® Multimode Atomic Force Microscopy, Quesant Instruments, USA. Prior to AFM imaging, samples were concentrated by centrifugation at 3000 rpm for 5 min and washing with PBS pH 7.4. Concentrated samples were spread on a glass plate left to dry for microscopical evaluation. Images were obtained and analyzed using the Nanoflight® software.

2.4. Anticancer efficacy of nanoparticles against MCF-7 cells

MCF-7 cells were maintained in DMEM supplemented with 10% Fetal Calf Serum at 37 °C in a humidified incubator containing 5% CO_2. Confluent cell monolayers were trypsinized and cells in exponentially growing phase were used in cytotoxicity experiments. The cytotoxicity of paclitaxel loaded nanocapsules, nanospheres, and paclitaxel dissolved in cremophor:ethanol mixture (50:50 v/v) against MCF-7 cells was

assessed using MTT assay. Paclitaxel doses in nanoparticles and cremophor vehicle were equalized to 150 μg/mL for the cytotoxicity studies. MTT assay was performed as described in the previous section with the exception of application of 90 μL cell suspension of 10^4 MCF-7 cells treated with 10 μL of formulation sample.

2.5. Statistical analysis

Cytotoxicity data were analyzed by two-way ANOVA for repeated measures using the ANOVREP programme. Hemolysis curves were analyzed statistically by Kruskall–Wallis analysis of variance and differences between groups were analyzed by Tukey test.

3. Results and discussion

Nanoparticles prepared with the nanoprecipitation technique and loaded with PCX were characterized by particle size distribution, zeta potential, drug entrapment efficiency and in vitro release studies. Particle size distribution analysis confirms the formation of nanospheres and nanocapsules of appropriate size which was reported to be 150 nm for nanospheres and 500 nm for nanocapsules. Zeta potential values indicate aqueous stability of colloidal nanoparticle dispersions ranging between −18 and −30 mV. Nanoparticles were demonstrated to possess high loading properties of up to 65% entrapment efficiency for paclitaxel and controlled release profiles of biphasic nature with complete in vitro release in 24 h (Bilensoy et al., 2007).

Commercial vehicle for injectable PCX dissolved in cremophor:ethanol mixture is associated with several side effects and cremophor is reported to be responsible for the non-linear pharmacokinetics and distribution of PCX upon iv infusion and incompatibility with plastic infusion sets used for administration during chemotherapy (Goldspiel, 1994; Sparreboom et al., 1996) as well as side effects that can even be fatal to the patient. In accordance with the objective of this study, amphiphilic cyclodextrin nanospheres and nanocapsules were evaluated for their hemolysis and cytotoxicity properties in comparison to cremophor:ethanol in terms of blank vehicles. In this context, blank 6-*O*-CAPRO-β-CD nanoparticles were assessed for their safety by hemolysis and cytotoxicity studies. Hemolytic property of PCX administered in commercial cremophor vehicle has already been reported (Bitton et al., 1995; Skyes et al., 1994). The effect of blank amphiphilic cyclodextrin nanospheres, nanocapules and the cremophor:ethanol mixture was evaluated on human erythrocytes. The molar concentration range studied was between 0 and 0.50 mM which was the highest molar concentration range possible for amphiphilic cyclodextrin nanoparticles. In biological conditions, a significant dilution with blood is expected to take place. Fig. 1 displays the hemolytic activity of blank vehicles on erythrocyte suspension. It can be observed that blank nanospheres and nanocapsules were significantly less hemolytic than the blank cremophor:ethanol mixture ($p < 0.05$). Nanocapsules were found to be slightly more hemolytic than nanospheres. This difference which is not statistically significant was attributed to the presence of Miglyol 812 as oil phase

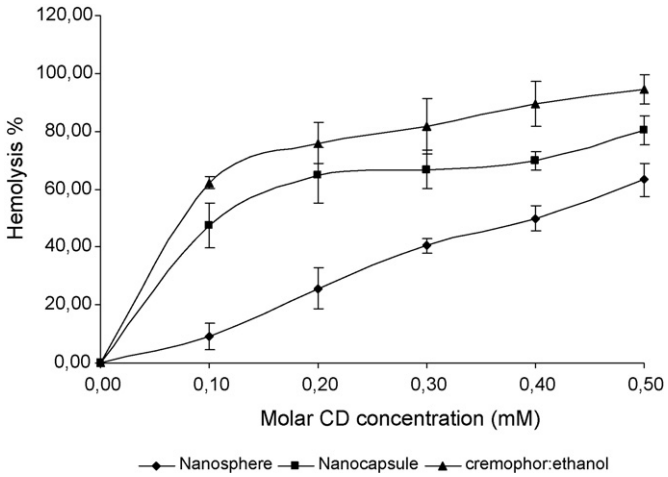

Fig. 1. Hemolysis of blank amphiphilic cyclodextrin nanospheres, nanocapsules and cremophor:ethanol mixture on erythrocyte suspension obtained from human blood ($n = 3$, S.D.).

in the nanocapsule formulations. The hemolysis caused by all vehicles was concentration-dependent with blank nanospheres yielding the most favorable hemolytic profile in all concentrations.

Hemolytic activities of blank vehicles were assessed with SEM photographs to visualize the effect of these carrier formulations on erythrocytes. It has been reported that intact erythrocytes show a typical discocyte shape that is similar to the shape of a bagel (Nagase et al., 2002). With increasing concentrations of a hemolytic agent, this shape changes to slight membrane invagination like a cup (stomatocytes) and a completely spherical shape (spherocytes). Finally, in total hemolysis, fusion-like cells may be observed upon SEM imaging. Fig. 2 represents the SEM photomicrograph of erythrocytes treated with the highest concentration of cremophor:ethanol vehicle that was used in UV spectrophotometric hemolysis studies. It can be clearly observed that all erythrocytes are in fusion-like state and

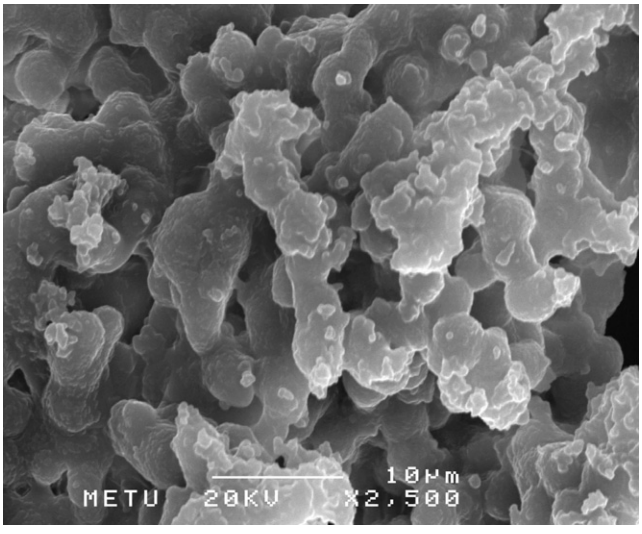

Fig. 2. SEM photomicrograph of erythrocyte suspension treated with cremophor:ethanol mixture (mag, 2500×).

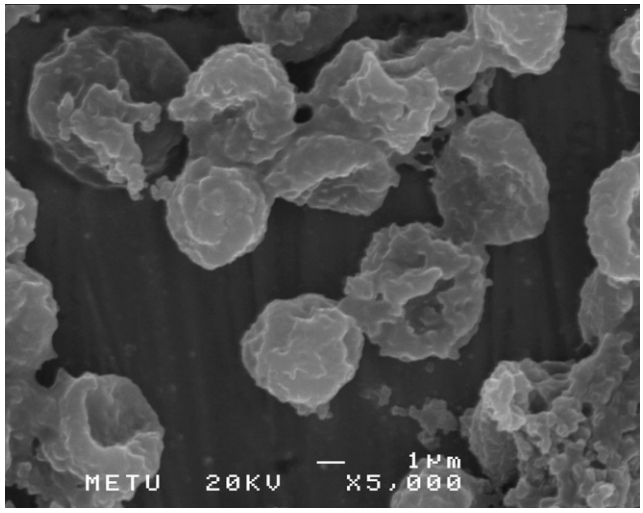

Fig. 3. SEM photomicrograph of erythrocyte suspension treated with amphiphilic cyclodextrin nanospheres (mag, 5000×).

completely disrupted. On the other hand, Figs. 3 and 4 represent nanosphere-treated and nanocapsule-treated erythrocytes respectively. Similarly, the highest concentration of nanospheres and nanocapsules used in the UV spectrophotometric study was administered to the erythrocyte suspension for SEM imaging. It is seen that discocytes are present for both nanospheres and nanocapsules. Nanocapsules on the other hand show fusion-like cells confirming the higher hemolysis found in Fig. 1 for these carrier systems. Since the highest possible concentration was used for the nanoparticles, some spherocytes are also present in Figs. 3 and 4 indicating hemolysis to a certain degree which is concentration-dependent.

Blank nanoparticles were evaluated for their cytotoxicity against L929 mouse fibroblast cell line with MTT assay. Fig. 5 represents the cell viability of L929 cells upon treatment with blank cyclodextrin nanospheres, nanocapsules and cremophor:ethanol vehicle. A major difference in the cytotoxicity

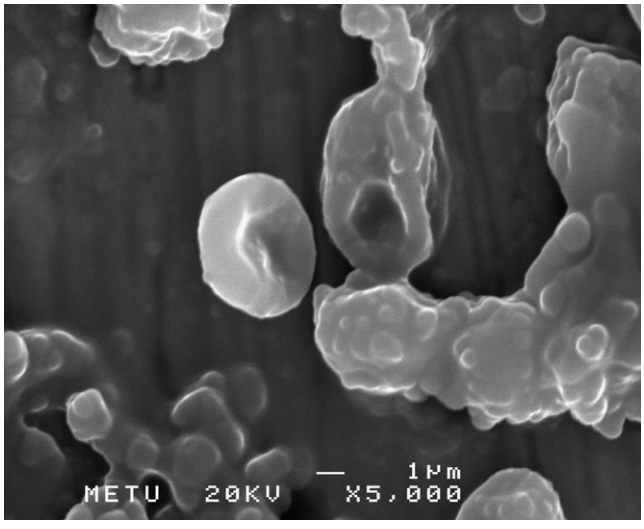

Fig. 4. SEM photomicrograph of erythrocyte suspension treated with amphiphilic cyclodextrin nanocapsule (mag, 5000×).

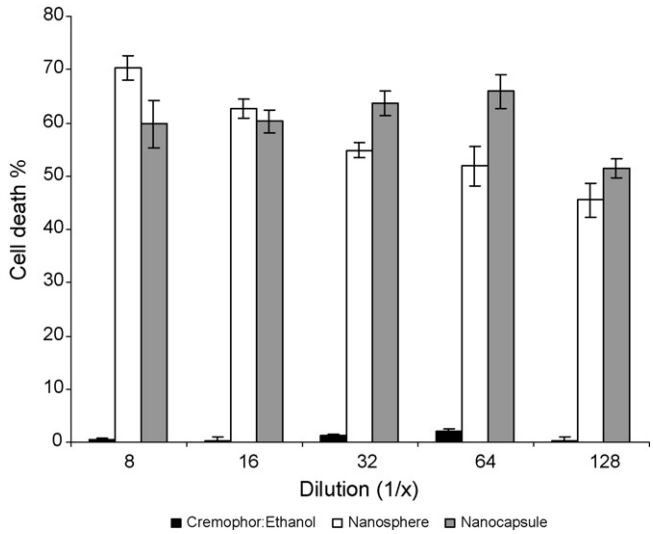

Fig. 5. Cytotoxicity of blank nanospheres, nanocapsules and cremophor:ethanol vehicle against L929 mouse fibroblast cell line ($n = 3 \pm$ S.D.).

profiles of the nanoparticulate carriers can be observed from Fig. 5. This is a significant finding in the sense that the toxicity arising from the carrier itself may be avoided by using cyclodextrin nanoparticles as carriers for PCX. No significant difference was observed between the cell viability values of nanospheres and nanocapsules.

The comparison between blank amphiphilic cyclodextrin nanospheres and nanocapsules with the commercial vehicle was performed in the highest possible concentrations for all tested samples. During a possible chemotherapy with paclitaxel, all of these three carrier systems (nanosphere, nanoparticle and cremophor vehicle) are subject to dilution with buffer solutions and blood in biological conditions.

PCX-loaded nanoparticles were evaluated for the ability to maintain the physical stability of the encapsulated PCX. When administered to cancer patients, commercially available PCX tends to re-crystallize and precipitate out in aqueous media. This leads to severe necrosis in injection site and other patient compliance problems as well as impairment of clinical efficacy of the drug and causes erratic plasma profiles. The nanoparticles loaded with PCX using two different encapsulation techniques were capable of maintaining the physical stability of PCX in aqueous dispersion for 1 month and avoid re-crystallization of the drug which could be the cause of undesired side effects. Figs. 6 and 7 show the mean diameter and zeta potential values for the amphiphilic cyclodextrin nanoparticles stored in aqueous dispersion state at 8 °C in refrigerator temperature for 7 days. It is seen that no significant changes are observed in particle size, polydispersity index (data not shown) and zeta potential during this follow-up period. Figs. 8 and 9 represent the AFM photomicrographs of amphiphilic cyclodextrin nanospheres prepared with the conventional and the high-loading techniques respectively. Nanocapsules could not be imaged by AFM due to the oily structure which does not permit the clear imaging of these delivery systems. AFM photomicrographs taken without any washing procedure or cemtrifugation for the nanospheres also show the physically stable PCX-

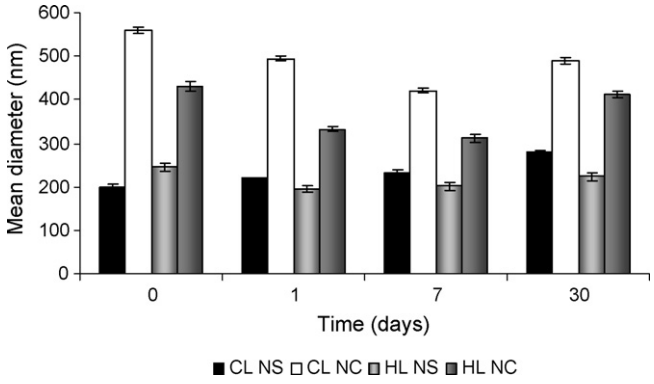

Fig. 6. Mean diameter of PCX-loaded different amphiphilic cyclodextrin nanoparticle formulations upon storage (CL NS: conventionally-loaded nanosphere, CL NC: conventionally-loaded nanocapsule, HL NS: high-loaded nanosphere, HL NC: high-loaded nanocapsule).

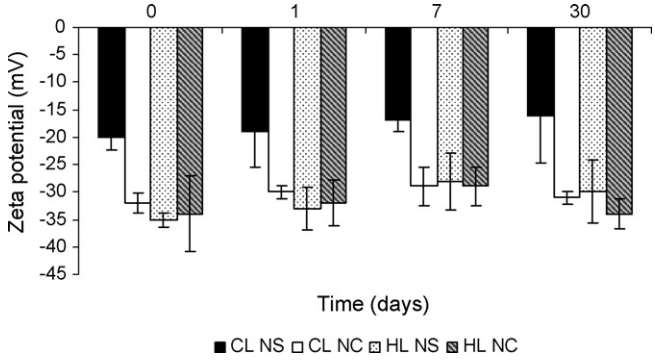

Fig. 7. Zeta potential values of PCX-loaded different amphiphilic cyclodextrin nanoparticle formulations upon storage (CL NS: conventionally-loaded nanosphere, CL NC: conventionally-loaded nanocapsule, HL NS: high-loaded nanosphere, HL NC: high-loaded nanocapsule).

loaded nanoparticles since no typical PCX crystals which have a needle-like shape were observed during atomic force microscopy.

Finally, PCX-loaded nanospheres and nanocapsules were compared to PCX dissolved in cremophor:ethanol vehicle in terms of anticancer efficacy against MCF-7 human breast cancer cell line. Cell viability after treatment with equivalent doses of PCX being 150 μg/mL associated to amphiphilic cyclodextrin nanosphere and nanocapsules and PCX solution (150 μg/mL) in cremophor:ethanol vehicle are seen in Fig. 10. It can be

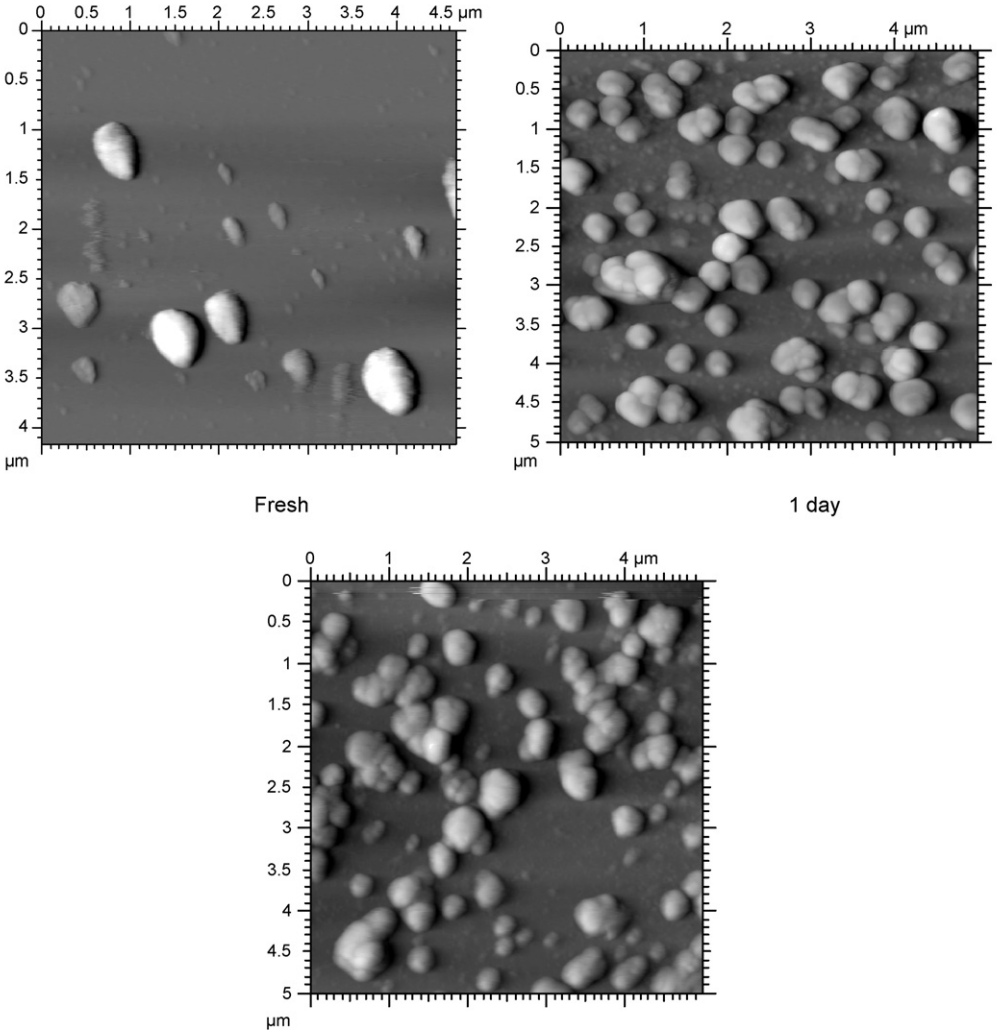

Fig. 8. AFM photomicrographs of conventionally-loaded cyclodextrin nanospheres.

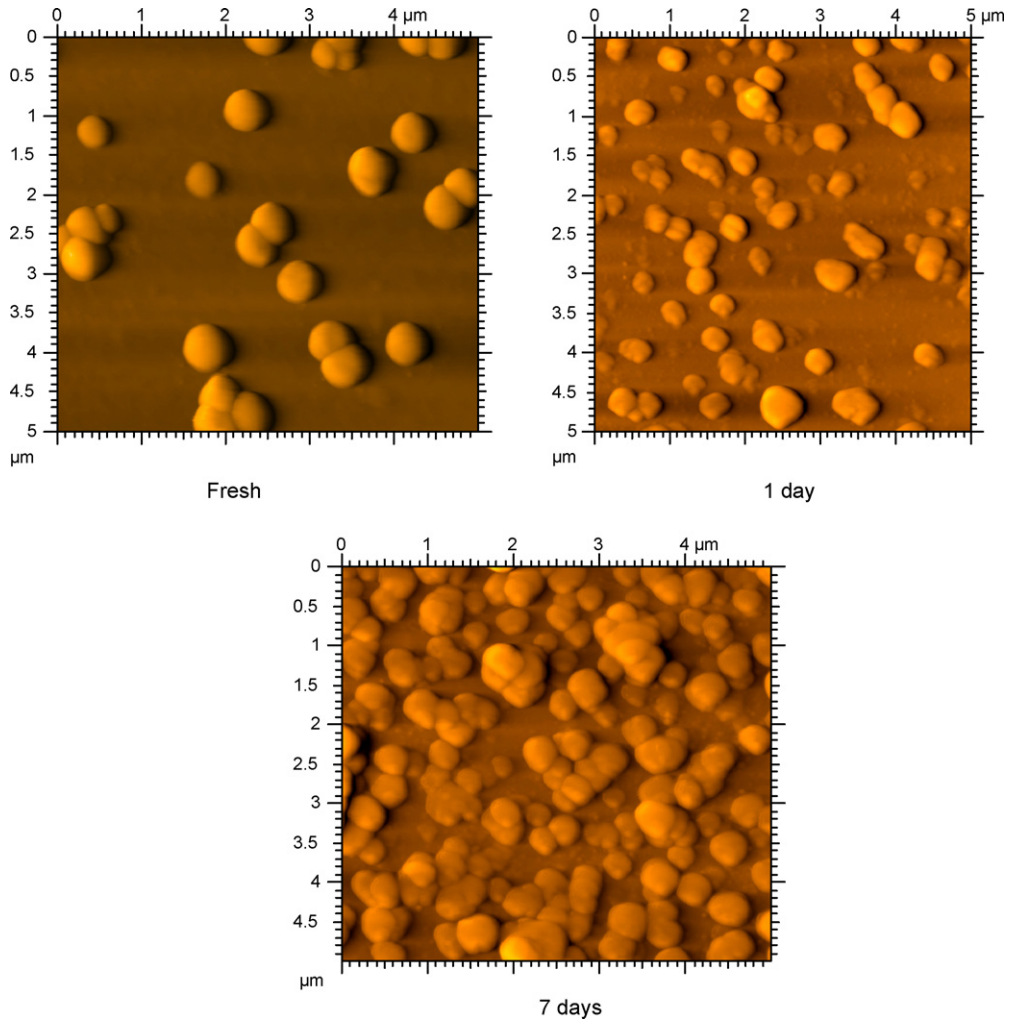

Fig. 9. AFM photomicrographs of high-loaded cyclodextrin nanospheres.

observed that all formulations exerted very similar cytotoxic effects against MCF-7 cells after a 72-h incubation time which can be considered quite short for such a study. Considering nanoparticulate formulations have slower release profiles for PCX, higher cytotoxicity can be expected from these formulations with a longer incubation time up to 120 h. The ratio of formulation sample to cell suspension was kept to 1:10 which is indicative a low dose for PCX. Regarding these facts, cell viability data are demonstrative of the anticancer efficacy of PCX-loaded amphiphilic CD nanospheres and nanocapsules which is similar to and slightly higher than that of PCX solution in cremophor:ethanol mixture.

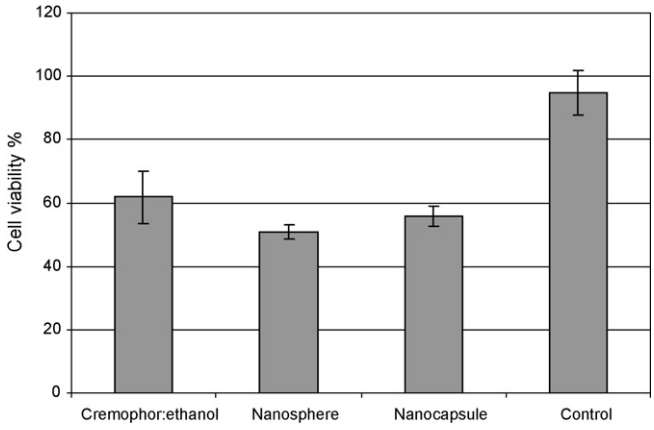

Fig. 10. Cytotoxicity of PCX-loaded amphiphilic cyclodextrin nanosphere, nanocapsule and PCX solution in cremophor:ethanol vehicle against MCF-7 cell line ($n = 4$).

4. Conclusion

In the light of the data obtained in this study, it can be concluded amphiphilic cyclodextrin nanospheres and nanocapsules loaded with paclitaxel seem to be effective alternatives avoiding the use of any surfactant or solubilizer for the injectable PCX formulation resulting in lower hemolysis and cytotoxicity for the carrier system with higher physical stability for the drug. Nanoparticle associated PCX can be considered as a promising alternative for cancer chemotherapy reducing side effects and enhancing potential bioavailability of the drug.

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