Journal of Drug Delivery Science and Technology 36 (2016) 68–74

Contents lists available at ScienceDirect

Journal of Drug Delivery Science and Technology

journal homepage: www.elsevier.com/locate/jddst

Albumin coated arginine-capped magnetite nanoparticles as a paclitaxel vehicle: Physicochemical characterizations and *in vitro* evaluation

N. Sattarahmady [a, b, *], N. Azarpira [c], A. Hosseinpour [a, d], H. Heli [a], T. Zare [a]

[a] Nanomedicine and Nanobiology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran
[b] Department of Medical Physics, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran
[c] Transplant Research Center, Shiraz University of Medical Sciences, Shiraz, Iran
[d] Department of Nanomedicine, School of Advanced Medical Sciences and Technologies, Shiraz University of Medical Sciences, Shiraz, Iran

ARTICLE INFO

Article history:
Received 31 March 2016
Received in revised form
9 June 2016
Accepted 24 July 2016
Available online 30 September 2016

Keywords:
Arginine coated magnetite nanoparticles
Paclitaxel
MRI contrast agent
Albumin
Core/shell structure

ABSTRACT

Today, magnetic nanoparticles (MNPs) were investigated for utilizing as a candidate for theranostic approaches in medicine. Because of the low solubility of paclitaxel (PTX) in aqueous media, finding a suitable delivery system for tumor treatment is desirable. Here, L-arginine-capped magnetite nanoparticles (Arg-MNPs) were synthesized and characterized. Arg-MNPs were superparamagnetic with an average size of 19.5 ± 5.3 nm obtained by field emission scanning electron microscopy and 22.1 ± 4.8 nm obtained by transmission electron microscopy. The effectiveness of Arg-MNPs on contrast enhancement in magnetic resonance imaging (MRI) was confirmed in agar phantoms. Then, Arg-MNPs were coated in a core/shell structure with human serum albumin (HSA/Arg-MNPs) and were investigated as a carrier for PTX. The data indicated that this magnetically targeted drug delivery system provides an effective treatment for breast cancer cells. Also, the carrier did not show any side effects on cell viability.

1. Introduction

Paclitaxel (PTX) is a poor water-soluble anti-cancer drug isolated from the bark of Pacific Yew (taxus brevifolia), and has a good therapeutic effect on a wide range of malignancies [1,2]. PTX binds to tubulin and disrupts the tubulin-microtubule equilibrium and microtubules de-polymerization, which arrest the cellular mitosis and lead to apoptosis [1,2]. Currently, PTX has been administrated using a formulation of 1:1 (w/w) mixture of polyethoxylated castor oil (Cremophor EL) and ethanol to increase water solubility [3,4]. However, this commercial formulation causes several side effects, such as hypersensitivity, cardiotoxicity, neurotoxicity, neuropathy and nonspecific distribution in the healthy cells; these limit the clinical application of PTX [4–7]. There have been proposed delivery systems to reduce these side effects [8,9]. Human serum albumin (HSA) has a high affinity to PTX and has been applied as a nanosized carrier for PTX, with a commercial name of Abraxane and Albunex in the market [10,11]. Nanodelivery systems can improve the permeation and retention of PTX in cancer cells and escape the normal cells [12]. Therefore, fabrication of stable nanovehicles with high loading efficiency for PTX is attracted substantial interest.

Magnetic nanoparticles (MNPs) have attracted considerable attentions in nanomedicine in theranostic goals, hyperthermia, drug targeting, magnetic resonance imaging (MRI) contrast agents, cell separation (bio-recognition), magnetofection for gene delivery, and biosensors [13–18]. Among MNPs, ferrimagnetic magnetite (Fe_3O_4) with superparamagnetic property is the most suitable for *in vivo* applications, due to biocompatibility [19,20], low toxicity [21], strong magnetic properties, remote controllability with an external magnetic field [22], cell homeostasis ability in iron balance [22], and surface reactivity [23]. In addition, MNPs with a size of <40 nm escape from the reticuloendothelial system [24] and surface modification of MNPs with active amine or carboxyl groups is useful for biomedical applications [25]. Some studies described surface functionalization and stabilization of MNPs with amines

* Corresponding author. Nanomedicine and Nanobiology Research Center, Shiraz University of Medical Sciences, Shiraz, Iran.
E-mail addresses: sattarahmady@yahoo.com, nsattar@sums.ac.ir (N. Sattarahmady).

http://dx.doi.org/10.1016/j.jddst.2016.07.004
1773-2247/

Table 1
The parameters for T_2 and T_2^* determination.

Imaging parameter	Time of repetition (TR)/ms	Times of echo (TE)/ms	Flip angle (FA)	Field of view (FOV)	Matrix size
T_2	2600	22, 44, 66, 88, 110, 132, 154, 176	180	160 × 160	256 × 168
T_2^*	40	24	20	160 × 160	256 × 192

and amino acids [26–29] and their applications in magnetic separation and immunoassays [27]. Amine-functionalized MNPs improve conjugation of magnetite with biological macromolecules such as proteins and oligonucleotides [26]. These surface modifications have important effects on binding of MNPs with serum proteins such as albumin [30]. HSA exists plentifully in the blood plasma with major roles in normal osmolality maintenance, and molecular transport in the blood and interstitial fluids [31]. Interaction ability of drugs with HSA causes a suitable delivery in target tissues with considerable effect on pharmacokinetics of drugs [32].

In this study, L-arginine (Arg)-capped magnetite nanoparticles (Arg-MNPs) were coated in a core-shell structure with HSA. Arg-MNPs were synthesized with a simple and one-step method from an iron precursor and L-Arg, and applied as a contrast agent in MRI. HSA coated Arg-MNPs (HSA/Arg-MNPs) were employed as a delivery system for PTX. The therapeutic efficiency of the PTX-loaded HSA/Arg-MNPs was evaluated using viability assessment of the breast cancer cells.

2. Materials and methods

2.1. Materials

HSA, PTX and L-Arg were purchased from Sigma (USA). Glutaraldehyde, iron(III) nitrate, sodium acetate, ethylene glycol, hydrochloric acid and acetonitrile of HPLC grade were purchased from Merck (Germany) or Scharlau (Spain) and used without further purification.

For cell culture studies, Roswell Park Memorial Institute-1640 (RPMI-1640) growth medium, fetal bovine serum (FBS), trypsin-EDTA and penicillin-streptomycin mixtures were from Gibco (USA). Dimethyl sulfoxide (DMSO) and 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) (MTT) reagent were purchased from Sigma (USA). Phosphate buffer saline (PBS) was supplied from Invitrogen (USA). MCF7 cells (NCBI C135, typical breast cancer cell line) were obtained from the Pasteur Institute Cell Bank of Iran. The PTX commercial form (PTX Stragen, 6 mg mL^{-1}) was purchased from a local drugstore. Deionized (DI) water was used throughout the study.

2.2. Synthesis of Arg-MNPs

A mixture containing 3.7 mmol iron(III) nitrate, 30 mmol L-Arg and 4.0 g anhydrous sodium acetate was added to 30 mL ethylene glycol and stirred vigorously at 50 °C to give a transparent solution. The obtained solution was then transferred into a Teflon lined autoclave and heated hydrothermally at 200 °C for 6 h. The resultant magnetite nanoparticles were then isolated by a magnet and rinsed with DI water and ethanol three times to eliminate ethylene glycol and unbound L-Arg, and then dried at room temperature.

2.3. Characterization of Arg-MNPs

Size and morphology of Arg-MNPs were evaluated using transmission electron microscopy (TEM, Zeiss, EM10C, Germany) with an accelerating voltage of 80 kV, and field emission scanning electron microscopy (FESEM, Zeiss, Sigma-IGMA/VP instrument, Germany). For TEM sample preparation, a drop of the diluted and horn-ultrasonicated sample in water was dropped on a copper grid

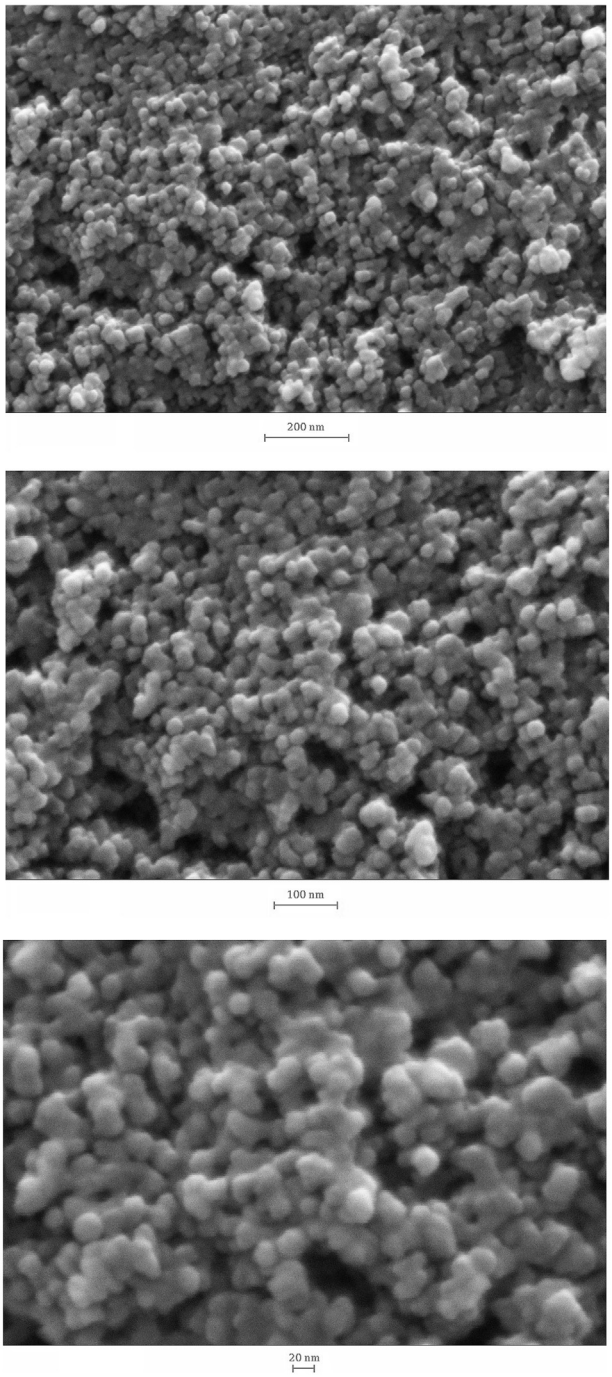

Fig. 1. FESEM images of Arg-MNPs with different magnifications.