Materials Science and Engineering C 69 (2016) 292–300

Contents lists available at ScienceDirect

Materials Science and Engineering C

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

Gated magnetic mesoporous silica nanoparticles for intracellular enzyme-triggered drug delivery

Na An, Huiming Lin *, Chunyu Yang, Ting Zhang, Ruihan Tong, Yuhua Chen, Fengyu Qu *

College of Chemistry and Chemical Engineering, Harbin Normal University, Harbin 150025, PR China

ARTICLE INFO

Article history:
Received 30 March 2016
Received in revised form 26 May 2016
Accepted 26 June 2016
Available online 29 June 2016

Keywords:
Mesoporous silica
Enzyme-triggered
Drug-delivery
Targeting
Core-shell nanoparticles

ABSTRACT

The targeting drug release is significant to the anticancer treatment. In this context, the redox-responsive drug delivery has attracted most attention owing to the intracellular reductive environment, such as the high concentration of glutathione reductase in many cancer cells. Herein, a glutathione sensitive drug delivery nanoplatform was constructed by using core-shell mesoporous silica nanocomposite (Fe_3O_4@$mSiO_2$) as carrier. By a simple silane coupling reaction, the glutathione cleavable diselenide linker has been prepared and grafted on to Fe_3O_4@$mSiO_2$ to insure the encapsulation of anticancer drug doxorubicin. The detail release kinetics studies reveal the glutathione triggered drug release, which could be further adjusted by varying the amount of diselenide linker. To improve the tumor-targeting, folic acid was grafted. The cellular uptake and drug release investigation was carried out using HeLa (cervical cancer cell line) as the model cancer cell and L02 and HUVEC (human hepatic cell line and human umbilical vein endothelial cells, non-cancerous cell lines) as control, indicating the enhanced cytotoxicity toward HeLa cells that benefits from the fast endocytosis and enhanced cellular drug release owing to their overexpressing folic acid receptors and high concentration of glutathione. Associating with the magnetic targeting, these novel nanomaterials are expected to be promising in the potential application of tumor-targeting therapy.

1. Introduction

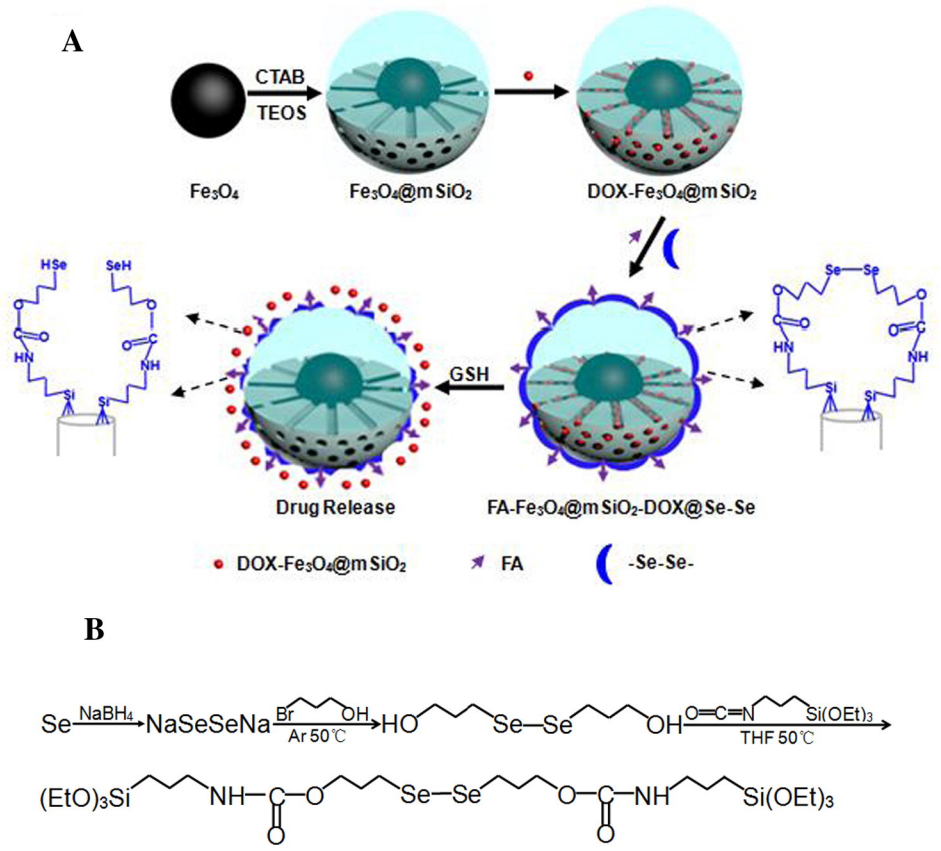
Currently, cancer has been considered as a major health concern worldwide. Chemotherapy always adopts particular cytotoxic drugs to induce the apoptosis of cancer cells and has been thought of the feasible and effective treatment modality, while the lack of specific discrimination between normal cell and cancer cell further limits its development. To further manipulate the release process, various nanomaterials have been developed as the vehicles for controlled drug release, including polymers [1–5], micelles [6–10], liposomes [11–15], inorganic materials [16–21] and so on. Among them, mesoporous silica nanoparticles (MSNs) have been considered as the one of most candidates due to the tunable pore size, high pore volume/surface area, prominent biocompatibility, and accessible surface functionalization [20,22–31]. To realize the on-demand drug release, the functional MSNs were constructed to reveal the drug release in response to various triggers. For example, Zhong et al. prepared the hollow mesoporous silica nanoparticles modified with poly(2-(diethylamino)-ethyl methacrylate), revealing the triple sensitive release to pH, reduction and light [32]. Zhu et al. developed a intelligent architecture by incorporating ZnO QDs to clog nanochannels of mesoporous silica, which in response to extracellular and/or intracellular acidic environ of tumor [33].

It is known that the distinct concentration difference of glutathione reductase (GSH) between the tumor tissues (2–10 mM) and the normal tissues (below 0.002 mM) has presented a novel trigger for the design of reduction-responsive nanovesicles. Currently, the preparation of GSH responsive almost focuses on the incorporation of disulfide bond (—S—S—) materials. Hu et al. have developed a chitosan based glycolipid-like nanocarrier (CSO-s-s-SA) which selectively responded to the reducing environment in tumor cells, showing the fast degradation and drug release in 10 mM levels of GSH [34]. The development of GSH-responsive delivery system based on disulfide modified poly(ethylene glycol) to cap MSNs and to deliver safranin O and doxorubicin in a controlled manner [16]. Based on the previous reports, the alternative Se—Se bond with more reactive and sensitive to the reduce environment and also has attracted many attentions. Currently, the utilization of Se—Se bond mainly was constructed into polymers or micelle systems, while the preparation of Se—Se modified mesoporous nanoparticles for sensitive drug release is few to be present [35–37].

In this paper, diselenide modified MSNs nanocarrier was constructed for the enzyme sensitive drug release in cancer cells. The methodology adopted to prepare the nanocomposites is illustrated in Scheme 1A. Firstly, Fe_3O_4 nanoparticles were introduced as a magnetic-targeting

* Corresponding authors.
E-mail addresses: linhuiming@hrbnu.edu.cn (H. Lin), qufengyu@hrbnu.edu.cn (F. Qu).

http://dx.doi.org/10.1016/j.msec.2016.06.086
0928-4931/



Scheme 1. (A) Schematic illustration of the synthesis and the controlled-release process. (B) Synthetic route of the diselenide linker.

core and coated with a mesoporous silica shell to obtain Fe_3O_4@mSiO_2 drug carriers. Doxorubicin hydrochloride (DOX), a typical anticancer drug was doped as the model drug to evaluate the controlled release-kinetics. In order to introduce —Se—Se— linker into the nanovehicles, the diselenide silane coupling agent was design and prepared as shown in Scheme 1B. After drug loading, the diselenide linker was grafted onto the surface of nanocomposites as the "plug" by a simple silane coupling reaction to insure the few prematurity. In addition, folic acid (FA) was introduced to enhance the cancer targeting performance. When these nanocomposites (named as Fe_3O_4@mSiO_2-DOX@Se-Se-FA) were taken up by objective cells via pinocytosis or phagocytosis, the "gate" is expected to be broken down owing to the high expression of GSH in the cancer cell, inducing the DOX release. Conclusively, the magnetic and FA targeting associates with the specific sensitive DOX release further improve the goal cytotoxicity to cancer cell.

2. Experimental section

2.1. Materials

Unless specified, all of the chemicals were analytical grade and were used without further purification. Iron chloride ($FeCl_3 \cdot 6H_2O$), sodium oleate, oleic acid, hexadecyltrimethyl ammonium bromide (CTAB), tetraethyl orthosilicate (TEOS), selenium powder, sodium borohydride, 3-bromo-1-propanol, 3-(triethoxy silyl) propyl isocyanate, doxorubicin hydrochloride (DOX), glutathione (reduced), folic acid (FA), *N*-(3-Dimethylaminopropyl)-N′-ethylcarbodiimide hydrochloride (EDC), *N*-Hydroxysuccinimide (NHS) and aminopropyltriethoxysilane (APTES) were obtained from Aladdin (Shanghai, China). 3-[4,5-dimethylthialzol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) and 1-octadecene were purchased from Sigma Aldrich (St. Louis, MO, USA). Dimethyl sulfoxide (DMSO), hexahydrate, ethanol, n-hexane, dichloromethane, ethyl acetate and tetrahydrofuran (THF) were purchased from Tianjin Chemical Corp. of China.

2.2. Method

2.2.1. Synthesis of Fe_3O_4 magnetic nanoparticles

Fe_3O_4 nanoparticles were prepared using a previously reported [38]. 36 g (40 mmol) of the iron–oleate and 5.7 g of oleic acid (20 mmol, 90%) were dissolved in 200 g of 1-octadecene (90%) at room temperature. The reaction mixture was heated to 320 °C with a constant heating rate of 3.3 °C min^{-1}, and then kept at that temperature for 30 min. When the reaction temperature reached 320 °C, a dramatic reaction occurred and the initial transparent solution became turbid and brownish black. The resulting solution, containing the nanocrystals, was then cooled to room temperature, and 500 mL of ethanol was added to the solution to precipitate the nanocrystals, which were collected by centrifugation and then dispersed in chloroform.

2.2.2. Synthesis of Fe_3O_4@mSiO_2nanoparticles

0.5 mL of the Fe_3O_4 nanocrystals in chloroform (10 mg mL^{-1}) were dropped into 8 mL of 0.2 M aqueous CTAB solution and the resulting solution was stirred vigorously for half an hour. The formation of an oil-in-water microemulsion resulted in a turbid brown solution. Then, the mixture was heated up to 60 °C for 30 min to volatilize the chloroform, resulting in a transparent black Fe_3O_4/CTAB solution. Next, 20 mL of distilled water was added to the obtained black solution and the pH value of the mixture was adjusted to 9–10 by using 0.1 M NaOH. After that, 100 mL of 20% TEOS in ethanol was injected six times at 30 min intervals. The reaction mixture was reacted for 24 h under violent stirring. The obtained Fe_3O_4@mSiO_2 NPs were centrifuged and rinsed with ethanol repeatedly to remove the excess precursors and CTAB molecules, and they were then dispersed in ethanol (8 mL).