

Novel Photo-Thermal Controlled Drug-releasing Vector for Cancer Detection and Treatment

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ABSTRACT

Cancer is always the leading cause of deaths in worldwide. The aim of this research is to develop a new generation of chemotherapeutic drugs with both photothermal activated drug release and MRI diagnostic function. Carbon nanotubes will be used as the vehicle to deliver platinum-based chemotherapeutic drugs such as oxaliplatin or cisplatin. With the intrinsic photothermal property of carbon nanotubes, oxaliplatin would be released controllably. And with the surface of carbon nanotubes being modified with super paramagnetic nanoparticles as contrast agents, this new generation of chemotherapeutic drugs would have MRI diagnostic function. The therapeutic and diagnostic efficacies of this new generation of drugs for colorectal cancer will be evaluated using both in vitro and in vivo animal studies. The associated neuronal side effects will also be assessed due to the control of drug release. The ultimate goal is to make such drugs to be commercially available.

Keywords: carbon nanotubes, oxaliplatin, drug release, colorectal cancer, photothermal

1 INTRODUCTION

Carbonnanotube (CNT) was discovered at 1991 by Sumio Iijima [1]. They started to become popular in all fields of science because of excellent physical and chemical properties, such as strong optical absorbance, thermal conductivity and electronic property. With the electronic property, several antistatic products have been produced by compounding with CNT. Because of property of nanosize, scientists already used it as a novel drug carriers in living systems with no significant toxicity to normal cell.

As a result of a cylindrical nanostructure of CNT, some clinical cancer drug was modified on its surface[2] or stuffed in its hollow center[3]. Besides the applications of cancer drugs, nonorganic nanoparticles were also modified on surface of CNT as MRI contrast agents[4].

Oxaliplatin, a widespread chemotherapeutic drug, has been used in clinical for several years. Because of

disadvantage of neural side effect, drug dosage of prescriptions is always limited[5].

Our team has been set up a simple drug controlled-release system by using CNT. Oxaliplatin-loaded magnetic nanovector (OMNV) is a vector that was filled with Oxaliplatin in the center and modified with superparamagnetic iron oxide (SPIO) on the surface.

According to design of OMNV, the OMNV could be a contrast agent for MRI and as a photo-thermal controlled drug-releasing system. OMNV would be a successful drug delivery for cancer therapy.

2 MATERIALS AND METHODS

2.1 Materials

Oxaliplatin was purchased from TTY Biopharm Co., Ltd. MWCNTs were purchased Golden Innovation Business Co., Ltd. All other chemicals were purchased from Sigma-Aldrich. TEM images were obtained by using Hitachi H-7100 Transmission Electron Microscope. MRI was carried out by using BRUKER 7T MRI. AAS was measured using Perkin Elmer PinAAcle 900 Atomic Absorption Spectrometer.

2.2 Synthesis of OMNV

Oxidative Short MWNT MWNT was mixture with concentrated H₂SO₄/HNO₃ (3/1, v/v) and stirred for 24 hours. The resultant suspension was then diluted with 250 mL of water, and the SWMTs were collected on a 100-nm-pore membrane filter and washed with deionized water. The obtained MWNTs were further redispersed in water, centrifuged at 10000g for 20min to remove the water immiscible carbon nanotubes or other residuals, lyophilized to dryness at room temperature.

Magnetic MWNT The resulting particles were dissolved in water with sodium oleate and sonicated for 30 min. Then the solution refluxed for 1 h after mixing with FeCl₃•6H₂O in 30ml-ethanol. Subsequently, the mixture was removed from the flask and filtered. Then the filter cake was dispersed into ethylene glycol by sonication to be stable. The suspension was transformed into a Teflon-lined

stainless steel autoclave and maintained at 300 °C for 6 h, and then cooled down to room temperature.

Oxaliplatin-loaded magnetic nanovector (OMNV) Magnetic MWNT were dissolved in DMF with oxaliplatin and sonicated for 1 h. After sonicating, the solution was kept at 70°C for 24 hours. The solution was slowly cooled down to crystallize the drug in the center of MWNT. Then the solution was filtered and the solid was washed with ethanol several times. The products was dried for use.

2.3 Cell Culture

HT29 human colorectal cancer cells was maintained in a humidified 5% CO₂ incubator at 37 °C in DMEM (GIBCO BRL, Gaithersburg, MD, USA) supplemented with 10% heat-activated fetal bovine serum (FBS) and 1% antibiotics (Antibiotic–Antimycotic).

2.4 Phtothermal Effect in Vitro

After the cells incubated for 24h, the culture medium was replaced by only DMEM. A laser with a wavelength of 808 nm (power, 1W) was used to irradiate the cells for different times. After irradiation, we incubated the cells for 24h. The cell viability was estimated by using a cell counter- Cellometer[®] Auto T4.

3 RESULTS AND DISCUSSION

3.1 Preparation of OMNV

A schematic illustration of an OMNV is shown in Fig.1 a). The loading efficiency of oxaliplatin of OMNVs is about 48 wt%. The average size of OMNVs is about 250 nm by DLS. The TEM micrographs of OMNVs in Fig.1 b) demonstrated that SPIO indeed attached to OMNVs and its diameter was about 6.6 nm.



Figure 1: a) schematic illustration of the structure and composition of an OMNV. b) TEM micrographs of OMNVs.

3.2 In vitro cytotoxicity

Photothermal effect of CNT alone : Each sample was treated with a total energy of 300 J/cm². After treatment, the maximum temperature increase was about 9°C (Fig.2 a)). At 37°C, the temperatures of samples treated with light quickly reached that for Photothermal Therapy. Samples treated with higher CNT concentrations had greater increase in temperature than samples treated with lower CNT concentrations. In Fig.2 b) , there were no significant differences for the cell viabilities among samples treated with light doses below 200 J/cm². This was due to the little increase in temperatures for samples treated with lower light doses. Samples treated with 50 ug/mL of CNT with a light dose of 300 J/cm² lost 30% of cell viability due to the photothermal effect.

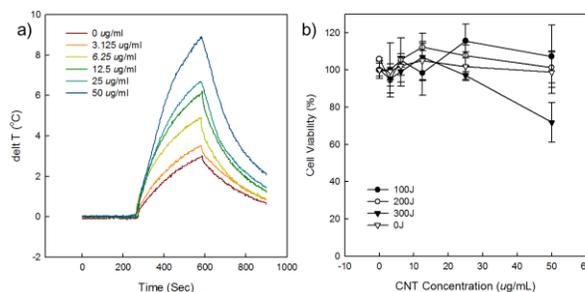


Figure 2: a) photothermal effect of different light doses with different concentrations of CNT. b) Cell viability of HT-29 cells treated with different light doses and concentrations of CNT.

Photo-thermal controlled drug-releasing effect : The cell viability of HT-29 cells treated with only OMNVs without light was almost the same as that of HT-29 cells treated with oxaliplatin only. Even though there is no significant difference in their ID50s, OMNVs still have advantages for clinical use due to their Enhanced Permeability and Retention Effect (EPR effect). The cell viabilities of HT-29 cells treated with lower OMNVs concentrations and light doses had no significant difference due to the slower increase in temperature.

Photo-thermal controlled drug-releasing effect was observed at samples treated with high OMNVs concentrations and 200 J/cm². With the greater increase in temperature, oxaliplatin could be released quickly from OMNVs. Thus, the cell viability of HT-29 cells was decreased dramatically.

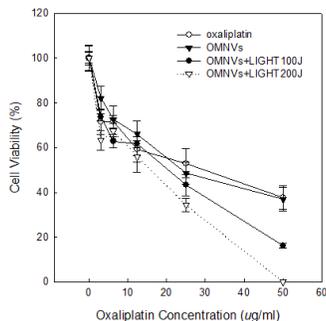


Figure 3: Cell viability of HT-29 cells treated with different light doses and concentrations of OMNVs.

3.3 Magnetization of OMNV

OMNVs were compared with normal magnetite (Fig.4 a)). OMNVs showed their magnetisms from VSM measurements (Fig.4 b)). MRI image proved OMNVs would be a contrast agent candidate (Fig.4 c)).

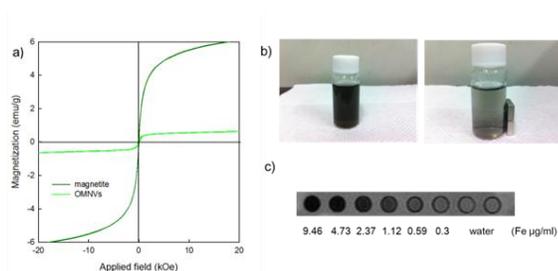


Figure 4: a) VSM measurements for the magnetizations of magnetite and OMNVs. b) Photographs of OMNVs dispersed in water before and after applying a magnet. c) MRI image of OMNVs.

4 CONCLUSIONS

A new generation of drug-controlled release system has been developed. Carbon nanotubes could be efficient vectors for Photothermal Therapy and Photo-Controlled Chemotherapy. With SPIO, both of the biodistribution and pharmacokinetics of OMNVs could be detected by MRI. The new efficient drug would be helpful for cancer therapy in the future.

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