Research Article

Synthesis and Cytotoxicity of POSS Modified Single Walled Carbon Nanotubes

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Received 4 December 2014; Revised 24 January 2015; Accepted 3 February 2015

Academic Editor: Andrew R. Barron

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Single walled carbon nanotubes (SWNTs) decorated with polyhedral oligomeric silsesquioxane (POSS) were synthesized via the amide linkages between the acid treated SWNTs and amine-functionalized POSS. The successful modification of SWNTs with POSS was confirmed by Fourier transform infrared spectroscopy (FTIR), X-ray photoelectron spectroscopy (XPS), and UV-Vis spectra. The resulting SWNTs-POSS can be dispersed in both water and organic solutions. The biocompatibility and cytotoxicity of the SWNTs and SWNTs-POSS were evaluated by CCK-8 viability assays, which indicated that SWNTs-POSS exhibit very extremely low toxicity. The low toxicity of the POSS modified SWNTs leads to more opportunities for using carbon nanotubes in biomedical fields.

1. Introduction

Carbon nanotubes (CNTs) have drawn considerable attention for many years due to their excellent electrical, mechanical, thermal, and optical properties. The unique structure and excellent properties allow carbon nanotubes to be suitable for many applications [1–6], such as catalysts [2], dry adhesives [3], sensors [4], and devices [5]. Recently, carbon nanotubes have shown promising applications in biomedical systems [7–12]. For instance, carbon nanotubes can be used for imaging [8, 10], drug delivery [9], tissue engineering [12, 13], and so on. The wide usage of carbon nanotubes in biomedical systems and human daily life raises urgent questions about the safety of its use and toxicity assessment in various systems.

The biocompatibility and toxicity of carbon nanotubes have been researched in some medical fields. Recent studies have shown that the toxicity of CNTs is determined by their morphology, number of the walls, and functional groups. Lam et al. [14] found that single walled carbon nanotubes are more toxic than carbon black in lungs, and SWNTs can induce dose-dependent epithelioid granulomas and interstitial inflammation in the animals. When carbon

nanotubes were induced into the abdominal cavity of mice, they would result in asbestos-like, pathogenic behavior [15]. Manna et al. [16] found that SWNTs can induce oxidative stress and activate nuclear transcription factor- κ B in human keratinocytes. Bai et al. [17] found that repeated administrations of carbon nanotubes in male mice can cause reversible testis damage without affecting fertility. Recent studies show that surface modification can decrease the cytotoxicity of carbon nanotubes [18, 19].

Polyhedral oligomeric silsesquioxane (POSS), a well-defined organic/inorganic hybrid molecule, has received much interest due to its unique cage-like structure and high performance when combining with polymers. Our previous work has shown that POSS modified polymethylmethacrylate (PMMA) exhibited superhydrophobic properties [20] and POSS modified Fe_3O_4 nanoparticles showed both superhydrophobic and magnetic properties [21]. POSS modified graphene has also shown amazing properties for multiple applications [22]. As POSS is constituted by Si–O and Si–C bonds, which is similar to the low toxic silicone, POSS is thought of as a potential building block for biomaterials [23–26].

FIGURE 1: Schematic representation of the preparation route to SWNTs-POSS.

FIGURE 2: (a) FTIR spectra of OapPOSS, SWNTs-COOH, and SWNTs-POSS. (b) UV absorption spectra of OapPOSS, SWNTs-COOH, and SWNTs-POSS in water solution.

In this paper, we decorated single walled carbon nanotubes (SWNTs) with POSS via covalent bond. The cytotoxicity of POSS, SWNT, and POSS modified SWNTs on human retinal pigment epithelial (RPE) cells was assessed by using CCK-8 methods. POSS modified SWNTs shows extremely low cytotoxicity, which may greatly enlarge applications of carbon nanotubes in biomedical field.

2. Materials and Methods

2.1. Synthesis of Octaaminopropyl Polyhedral Oligomeric Silsesquioxane (OapPOSS). The OapPOSS was synthesized according to the literature method [27]. Typically, to a flask equipped with a magnetic stirrer, 360 mL of methanol, 27 mL of concentrated hydrochloric acid (37%), and 15 mL

FIGURE 3: Photos of (a) SWNTs dispersed in water and (b–e) SWNTs-POSS dispersed in water and organic liquids.

FIGURE 4: TGA curves of OapPOSS, SWNTs, SWNTs-POSS, and SWNTs-COOH.

(0.0838 mol) of γ -aminopropyltriethoxysilane were charged. The hydrolysis and rearrangement reaction was carried out for 6 weeks at room temperature and the microcrystalline precipitates were obtained. The product was collected (3.7 g, 30% yield) after filtration, washed with methanol, and dried in a vacuum oven. FTIR (cm^{-1}): 3200–2800 (N–H), 3000–2800 (C–H), 1105 (Si–O–Si); ^{29}Si NMR (ppm): 67.0 (s).

2.2. Synthesis of SWNTs-COOH. SWNTs were purchased from Nanjing XFNANO materials Tech Co., Ltd., China. 50 mg of SWNTs was added into a 100 mL flask containing a mixture of H_2SO_4 (30 mL, 98%) and HNO_3 (10 mL, 60%). The mixture was then ultrasonicated for 8 hours. The resulting solution was diluted with 200 mL of deionized water, followed by a vacuum-filtering through a $0.22\ \mu\text{m}$ polycarbonate film. The solid product was washed with deionized water three times. SWNTs functionalized with carboxyl groups were obtained after vacuum drying.

2.3. Preparation of SWNTs-POSS. In a 100 mL round bottom flask, 50 mg of SWNTs-COOH, 2 g of OapPOSS, and 50 mg of dicyclohexylcarbodiimide (DCC) (as catalyst) were dispersed

FIGURE 5: Raman spectra of SWNTs, SWNTs-COOH, and SWNTs-POSS.

in 40 mL ethanol, followed by ultrasonication for 10 minutes. The mixture was then refluxed for 24 hours at the temperature of 100°C . When the reaction finished, the resulting solution was cooled down to room temperature and then vacuum filtered through a $0.22\ \mu\text{m}$ polycarbonate film. The solid product was thoroughly washed with distilled water for three times; then the product was washed with chloroform and dried in vacuum oven.

2.4. Characterization. Fourier transform infrared spectroscopy (FTIR) spectra were recorded on a PerkinElmer spectrum GX FTIR system at room temperature. The samples and KBr crystal were ground together using a mortar and pestle. The resulting ground power was pressed into small flakes for FTIR measurement. UV-Vis spectra were collected on an Agilent Cary 100 UV-Vis spectrophotometer; all the samples were dissolved in water solution for UV-Vis measurements. The thermogravimetric analysis was taken on a TA instrument with a heating rate of 10°C . The Raman measurement was carried out on a Raman spectroscopy (Renishaw) using 514 nm laser. X-ray photoelectron spectroscopic (XPS) spectra were recorded on a PHI 5000 VersaProbe.

3. Results and Discussion

Figure 1 illustrates the synthesis process of POSS functionalized SWNTs. Firstly, SWNTs were converted into SWNTs-COOH by surface modifying SWNTs with a mixture of H_2SO_4 and HNO_3 . POSS was then incorporated into SWNT via covalent bond between the amine groups in POSS and acid groups in SWNTs-COOH.

The structure of the as-prepared SWNTs-POSS was characterized by FTIR and UV. Figure 2(a) shows the FTIR spectra of SWNTs, OapPOSS, and SWNTs-POSS. The spectrum of OapPOSS shows a very sharp peak at $1110\ \text{cm}^{-1}$, which is corresponding to the Si–O–Si stretching band. This peak was also found in the spectrum of SWNTs-POSS, indicating POSS has been incorporated into SWNTs.

FIGURE 6: TEM images of (a) SWNTs and (b and c) SWNTs-POSS.

FIGURE 7: XPS spectrum of SWNTs-POSS.

FIGURE 8: Vitality of RPE cell incubated with OapPOSS at 24, 48, and 72 hours.

The synthetic results also characterized UV absorption spectra. As is shown in Figure 2(b), the spectrum of OapPOSS has a strong absorption peak at 200 nm, while SWNTs-COOH has a broad absorption band at 225 nm. After being modified with POSS, the resulting SWNTs-POSS have a broad adsorption peak at about 250 nm.

Figure 3 shows the photographs of SWNTs and SWNTs-POSS dispersing in water and organic solutions. On dispersing the neat SWNTs in water, the black powder quickly deposited at the bottom of the vial. However, when SWNTs were surface modified with OapPOSS, the resulting SWNTs-POSS can be stably dispersed not only in water, but also in organic solutions, such as N,N-dimethylformamide (DMF), dimethyl sulfoxide (DMSO), and tetrahydrofuran (THF).

The thermal stability of OapPOSS, SWNTs, SWNTs-COOH, and SWNTs-POSS was studied by TGA. As is presented in Figure 4, the pristine SWNTs have very high thermal stability and the initial decomposition temperature (T_d) is as high as 600°C, while OapPOSS has a T_d at 347°C. The char yields of SWNTs and OapPOSS at 800°C are 79.71% and 40.23%, respectively. The initial decomposition temperatures of SWNTs-POSS and SWNTs-COOH are greatly decreased when comparing with pristine SWNTs because of the functional groups of POSS and COOH. The thermal stability of SWNTs-POSS was better than SWNTs-COOH, because the unstable COOH groups in SWNTs-COOH were reacted with NH_2 groups in POSS. TGA results also confirmed that OapPOSS has been successfully incorporated into SWNTs.

Figure 5 is the Raman results of SWNTs, SWNTs-COOH, and SWNTs-POSS. In the Raman spectrum of SWNTs, there is a small peak at 195 cm^{-1} , which is the characteristic peak of SWNTs. However, this peak disappears in the Raman spectra of both SWNTs-COOH and SWNTs-POSS. Moreover,

the ratio of D band to G band of SWNTs-POSS is much higher than that of SWNTs and SWNTs-COOH, because the structure of SWNTs was destroyed during the acids treatment and POSS functionalization process. All these have indicated that POSS has been successfully synthesized onto SWNTs' surface.

Figure 6 shows TEM images of SWNTs and SWNTs-POSS with different magnifications. As is shown in Figure 6(a), the SWNTs were aggregated together. After being treated with acid and reacted with POSS, SWNTs-POSS become short and separated (Figure 6(b)). In Figure 6(c), some black dots, which were thought as POSS, were found on the surface of carbon nanotubes. X-ray photoelectron spectroscopy (XPS) has also been used to characterize the SWNTs-POSS. As is shown in Figure 7, the atom percent of the carbon, oxygen, silicon, and nitrogen is 62.77%, 28.66%, 5.20%, and 3.37%, respectively. The appearance of silicon and nitrogen peaks in XPS curve of SWNTs-POSS (Figure 7) came from POSS, which can also confirm that SWNTs-POSS were synthesized.

The cytotoxicity and biocompatibility of carbon nanotubes are very important when one introduces these materials into biomedical applications. OapPOSS was used to modify the surface of SWNTs in order to designing biocompatible nanohybrids. CCK-8 viability assays were used to assess the cytotoxicity of OapPOSS on human retinal pigment epithelial (RPE) cells firstly. As is shown in Figure 8, the cell survival is more than 85% even at the high concentration (500 µg/mL) of OapPOSS after 72 hours. OapPOSS demonstrates extremely low toxicity levels.

When combining POSS with SWNTs, POSS formed a protective layer surrounding carbon nanotubes. As is presented in Figure 9, The RPE cell viability decreased with increasing the concentration of SWNTs, SWNTs-COOH, and

FIGURE 9: Vitality of RPE cell incubated with SWNTs, SWNTs-COOH, and SWNTs-POSS at (a) 24 hours, (b) 48 hours, and (c) 72 hours.

SWNTs-POSS. However, the highest cell viability was found in those culture solutions with SWNTs-POSS, which indicated that the cytotoxicity of SWNTs was greatly decreased when being functionalized with OapPOSS.

4. Conclusion

In summary, we have successfully incorporated POSS onto the surface of single walled carbon nanotubes via covalent bond. The results from FTIR, XPS, UV, and TGA clearly indicated that SWNTs-POSS were obtained successfully. CCK-8

assays demonstrate that OapPOSS has very low cytotoxicity and high biocompatibility. When combining OapPOSS and SWNTs, POSS formed a protective layer surrounding carbon nanotubes, and the cytotoxicity was greatly decreased. It will greatly extend the biomedical applications of carbon nanotubes.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

Acknowledgment

This work was supported financially by NSFC (51202167 and 81271703).

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