

Glucose sensors made of novel carbon nanotube-gold nanoparticle composites

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Abstract. Carbon nanotube and metal particle composites have been exploited to fabricate high performance electrochemical devices. However, the physical and chemical procedures to synthesize the composites are labor intensive and inefficient. Our study reveals an one-step wet chemistry method to accomplish fast and controllable production of gold nanoparticle (AuNP) and carbon nanotube (CNT) composites. Such a process is sensitive to the surface charge. Especially, when functionalized with carboxyl groups, the CNTs carried negative charges and showed low level association with negatively charged AuNPs. Thermal treatment was employed to decompose the carboxyl groups and render each CNT a charge-free surface thereby achieving a high level AuNP-CNT association. The fabricated glucose sensors demonstrated dependence of their sensitivities to the amount of AuNPs on the CNTs. The enhancement of sensitivity can be attributed to an accelerated electron transfer rate from glucose oxidase Gox to the electrode. The Michaelis-Menten kinetics also indicated improved performance in the glucose sensor made of AuNP-CNTs. Therefore, our research revealed a novel approach to produce metallic nanoparticle and CNT composite for fabricating high performance electrochemical sensors.

Keywords: Carbon nanotube, sensor, nanoparticle, glucose oxidase

1. Introduction

Carbon nanotubes (CNTs) have promising properties in electrochemical applications, such as sensors and fuel cells [1,4,7,11,13,15,19,20,22]. The large surface area of the CNTs offers abundant reactive sites to generate Faradic currents. Also, the reactions on CNT modified electrodes have shown direct electron transfer property [5,14,15,24]. Recently, metallic nanoparticle-CNT composites were used to enhance the sensitivity of bio/chem-sensors [6,10,12,17,21]. However, the method used to prepare these electrodes is labor-intensive. Herein, a one-step method is described to fabricate CNT and gold nanoparticle (AuNP) composites with minimal physical or chemical modifications. Glucose sensors made of such composites were used to evaluate the performance of this electrochemical material.

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2. Nanomaterial preparation

CNT produced by chemical vapor deposition was obtained from NanoLab, Inc. (Newton, MA, USA). The nominal parameters are 30 nm in diameter, 5–20 μm in length, and 95% in purity.

A process for CNT functionalization is to use oxidative acid to etch CNTs and generate carboxylic groups on the surface [3,16,18]. Briefly, CNTs were mixed with acids composed of 3:1 (v:v) of H_2SO_4 (98%) and HNO_3 (67%) at 3 g/L. The mixture was heated at 80°C and kept overnight. The CNTs were then recovered by filtration through a glass fiber filter twice. The CNT biscuits were sonicated to resuspend in solution. We found that the functional groups made the suspension very stable. NaCl was added to the suspension at 1 M to trigger salt-induced-aggregation and facilitate the precipitation of functionalized CNT (fCNT) by centrifugation at 3000 rpm. The CNT pellet was washed four times in diH_2O and the final solution was titrated with NaOH until the pH is between 3 and 7. The concentration of CNT was adjusted to 3 g/L and remained stable for at least 3 months. The defunctionalization of fCNTs was carried out by thermal treatment as reported [23]. fCNTs 100 mg were extracted from the suspension and transferred to a quartz container followed by vacuum-drying in a tube furnace without heating overnight. The furnace was then heated to 95°C (for 2 hours) and then temperature was gradually raised to 500°C. The thermal treatment was maintained for 1 hour in vacuum to decompose the carboxyl groups thereby yielding the thermal CNT (tCNT). The tCNTs maintained the same structural features as the fCNTs.

Negatively charged AuNPs were obtained from Nanoprobes (Yaphank, NY).

CNTs were dispersed in diH_2O at 1 mg/ml by sonication. The sonicating (Fisher Scientific) parameters were 20% power, 30 s duration with 1 s (on)/1 s (off) cycling. 1 ml CNT dispersion was removed and mixed with 200 μL colloidal AuNP. The mixture was then shaken and kept at room temperature for 10 min. The CNT precipitated by itself. The colorless supernatant indicated the adsorption of AuNP on CNTs. Following centrifugation, the supernatant was removed and the AuNP-CNTs were resuspended in diH_2O . The centrifugation-suspension cycle was repeated four times to remove the unadsorbed AuNP. Finally, the CNTs were brought to 1 mg/ml.

The adsorption of AuNPs to CNTs was evaluated by transmission electron microscopy (TEM) (Fig. 1). The high resolution transmission electron microscopic (HRTEM) images were obtained with JEOL 2010F; samples were suspended in ethyl alcohol and cast (4 μL) on a carbon film coated Cu grid and left to dry.

According to TEM imaging, the linear density of AuNPs on CNTs was found in the order: CNTs > tCNTs > fCNTs. The fCNTs are negatively charged due to the presence of carboxyl groups. Probably, it was the electrostatic force that prevented the fCNTs from binding to the negatively charged AuNPs. As for tCNTs, the barrier was removed after the thermal treatment. We observed high level AuNP adsorption on the CNTs. On average, there were 57 AuNPs along every 100 nm length of CNT, which were more than that on regular CNTs, i.e. 11 AuNPs per 100 nm. Although more characterization is required to elucidate the mechanisms of adsorption, it is clear that AuNP can directly attach to CNTs through a one-step process, which requires no additional chemical modifications and can yield uniform coating of the CNTs with AuNPs.

3. Glucose sensor fabrication

The AuNP-CNT composites were used to fabricate glucose sensors (Scheme 1). Following the AuNP-CNT composite formation (Step 1), the AuNPs surface was modified with cysteamine based on thiol-gold

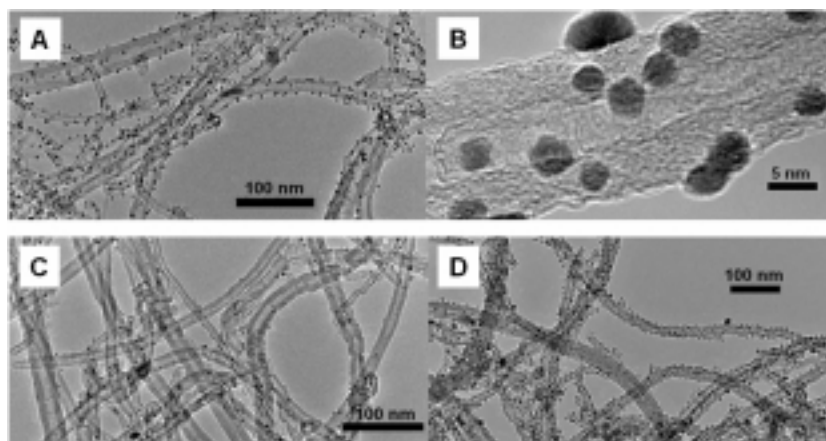
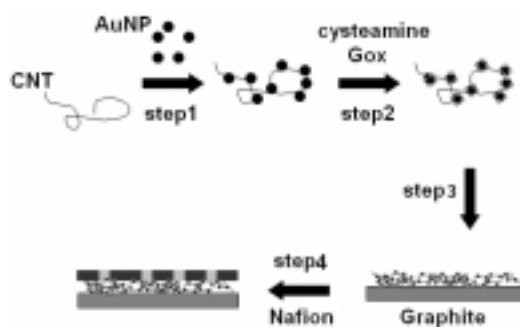


Fig. 1. TEM characterization of the AuNP-CNT composites. (A) CNTs, (C) fCNTs and (D) tCNTs with including 3 nm AuNPs. (B) AuNP-CNT at high magnification of A.



Scheme 1. Glucose sensor made of the AuNP-CNT composite.

interaction. The amine group in cysteamine positively charged the composite surface and attracted the negative glucose oxidase (Gox) at neutral pH [2] (Step 2). The AuNPs-CNT composite with Gox was then dispersed on fine polished graphite disc and dried at 4 °C (Step 3). Finally, the whole structure of the electrode was covered by Nafion (Step 4) as reported [17].

Ferrocenecarboxylic acid (FCA) and Nafion (5 wt.% in lower aliphatic alcohols) were purchased from Aldrich (Milwaukee, WI). Glucose, glucose oxidase (Gox) and cysteamine were obtained from Sigma (Saint Louis, MO). Phosphate buffered saline (PBS) was obtained from Fisher Scientific (Suwanee, GA). High grade graphite rods were purchased from Poco graphite (Decatur, TX). They were cut into 0.5 mm thick discs. After being polished, one side of the graphite disc was connected to an insulated electrical wire with silver paste, then sealed with Epon Resin 828 (Miller-Stephenson Chemical Co., Sylmar, CA). The other side was left for CNT deposition.

4. Electrochemical detection and performance evaluations

The fabricated glucose sensors were used to evaluate the performance with respect to the sensitivity, electrotransfer rate and enzymatic kinetics. Electrochemistry experiments were performed with a Gamry

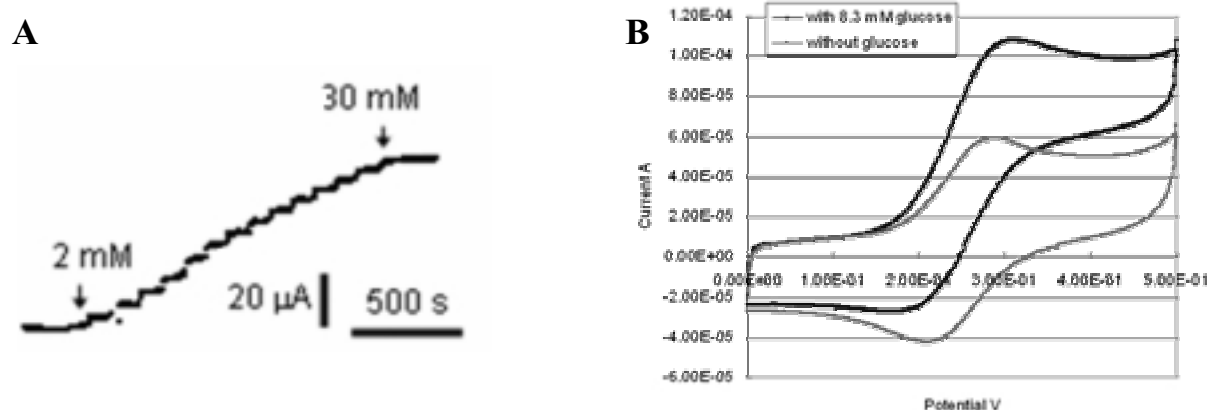


Fig. 2. Electrochemistry recording with the AuNP-tCNT based glucose sensors. (A) Cyclic voltammogram with 1 mM FCA. (B) Amperometric current corresponding to successively addition of 2 mM glucose every 200 s.

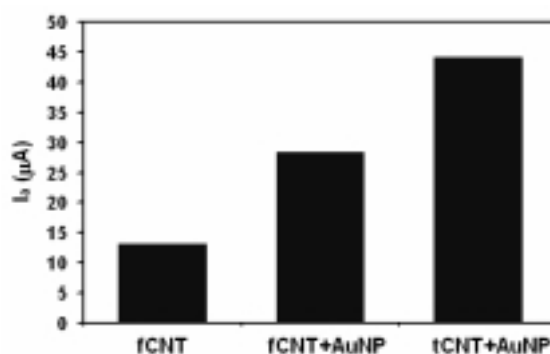


Fig. 3. Anodic currents of sensors made of three kinds of CNT materials in response to 10 mM glucose.

PC400 that was plugged in a Pentium III computer system. Gamry Instruments Framework and Gamry Echem Analyst software were used for recording and data analysis. The glucose sensor, the Ag/AgCl reference electrode, and platinum wire counter electrode were inserted into the Dr. Bob's cell (Gamry Instruments, Warminster, PA). Usually, we injected 5 ml N_2 purged buffer in the cell for recording. A magnetic stirrer provided the convective transport during the amperometric measurement. As shown in Fig. 2, for amperometric recording, the sensor worked at 500 mV with the magnetic stirrer on; for voltammetry recording, the voltage scanned from 0 to 500 mV at variable scanning rates with respect to the experimental design. Before each new experiment, the containers and electrodes were rinsed and refreshed. All solutions were prepared freshly.

Anodic currents, I_a , in response to 10 mM glucose were obtained from glucose sensors made of three kinds of different materials, i.e. fCNT, fCNT+AuNP and tCNT+AuNP. As mentioned above, the materials for the sensors were of same origin and held the similarity in their physical properties, such as electrical conductance and morphological parameters. Their primary differences exist in the amount of AuNPs adsorption. As shown in Fig. 3, the sensor sensitivities exhibited a correlation to the amount of AuNPs involved in the sensors, i.e. more AuNPs correlated to higher sensitivity.

The cyclic voltammetric (CV) curves of Gox redox at various scan rates, ν , were also studied. The anodic peak current, I_{pa} , increases linearly with ν (Fig. 4A), indicating that the redox reaction is a surface

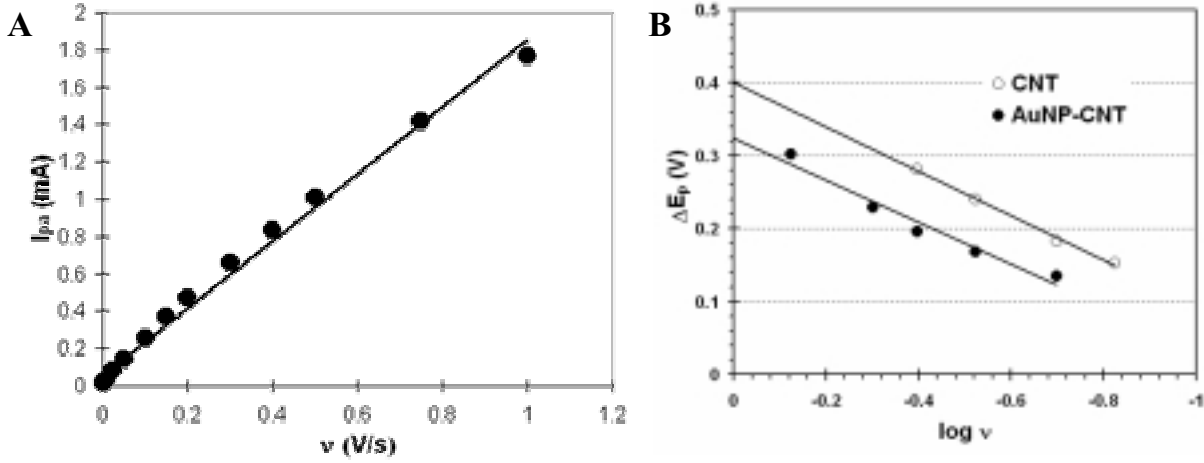


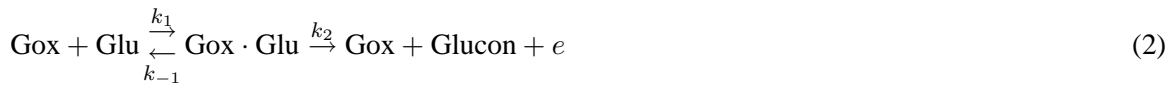
Fig. 4. Electron transfer rate analysis. (A) The anodic peak current I_{pa} vs. the scanning rate, ν , indicates the reaction on the sensor surface was surface-controlled quasi-reversible process. (B) Electron transfer analysis based on Laviron's equation. ΔE_p is the voltage difference between the redox peak currents in CV. The sensors that were made of CNT and AuNP-CNT were compared with respect to the electron transfer parameters indicated in the charts.

controlled reversible process. The slope of the straight line ($r^2 = 0.9924$) of I_{pa} versus ν is $1.7963 \times 10^{-3} \text{ A s/V}^{-1}$. ΔE_p showed linear dependence vs. $\log \nu$, when the scan rate was larger than 100 mV/s (see Fig. 4B). For a surface-controlled electrode reaction, the Laviron's equation [8,9] describes the relationship between ΔE_p and $\log n$:

$$\Delta E_p = \frac{2.3RT}{(1-\alpha)\alpha nF} \times \left[\alpha \log(1-\alpha) + (1-\alpha) \log \alpha - \log \left(\frac{RT}{nF} \right) - \log k \right] + \frac{2.3RT}{(1-\alpha)\alpha nF} \log \nu \quad (1)$$

where α is the electron-transfer coefficient and k is the heterogeneous electron transfer rate constant. Both parameters can be derived from the slope and interception of fittings in Fig. 4B. For sensors made of CNT and AuNP-CNT, the reactions exhibited same α , i.e. 0.73, but k of the AuNP-CNT (1.17 s^{-1}) was larger than the CNT alone (0.63 s^{-1}), which was considered as the evidence of AuNP facilitated electron transfer.

The reaction on a glucose sensor can be simplified as:



where Glu and Glucon stand for glucose and gluconolactone. The Michaelis-Menten equation:

$$\frac{1}{I} = \frac{1}{I_{a,\max}} + \frac{K_m}{I_{a,\max}} \cdot [\text{Glucose}] \quad (3)$$

describes the Gox mediated glucose oxidation kinetics. The steady state anodic currents ($I_{a,\max}$) and Michaelis-Menten constants (K_m) are defined as:

$$I_{a,\max} = [\text{Gox}]_0 \cdot k_2 \quad (4)$$

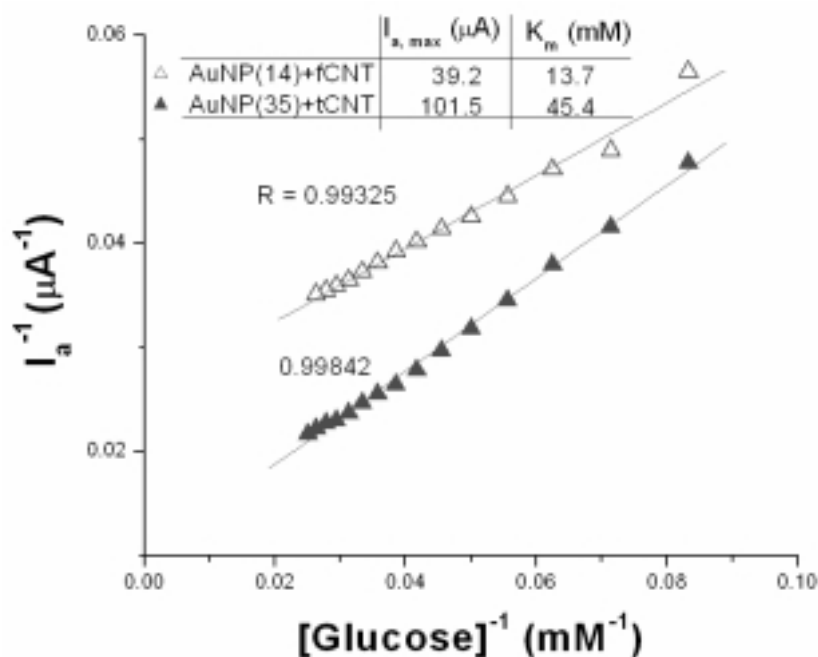


Fig. 5. Lineweaver-Burk plot to elicit the Michaelis-Menten kinetics parameters. The two kinds of sensors were made of AuNP-CNT composites containing 14 or 35 AuNPs per 100 nm length. More AuNPs resulted in higher Michaelis-Menten parameters.

$$K_m = \frac{k_{-1} + k_2}{k_1} \quad (5)$$

Both are correlated with the turnover rate k_2 . The Lineweaver-Burk plots of two different sensors were shown in Fig. 5. The increase of the two parameters was observed corresponding to the enrichment of AuNPs in the sensors. One interpretation is that AuNPs provide more reactive sites, facilitate faster electron transfer from enzyme to CNTs, then may reduce the electron accumulation and consequently lead to higher k_2 .

5. Conclusion and discussion

We report a novel process to associate AuNPs with CNTs without complicate chemistry. It is convenient and efficient, however, sensitive to surface charges. The AuNP-CNT composite can be used as high performance materials for electrochemical applications. According to the measurement with the fabricated glucose sensors, higher sensitivity was obtained with AuNP-CNT composite containing more AuNPs. Also, examined with Laviron method, the improved electron transfer was confirmed in the glucose sensor made of AuNP-CNTs in contrast to those with CNTs only. The Michaelis-Menten kinetics also indicated promoted enzymatic activity facilitated by the AuNP-CNT composite. The discovery reported here may contribute to the fabrication of novel biosensor and biofuel cells.

Acknowledgements

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