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A highly sensitive nonenzymatic glucose sensor based on NiO-modified multi-walled carbon nanotubes

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Abstract A highly sensitive and selective glucose biosensor has been constructed by using highly dispersed NiO nanoparticles supported on well-aligned MWCNTs (NiO/MWCNTs) as sensing interface. The NiO/MWCNTs nanocomposite was synthesized by magnetron sputtering deposition of NiO nanoparticles on vertically aligned carbon nanotubes. The nanocomposite electrode showed high electrochemical activity towards the oxidation of glucose in 0.20 M NaOH solution. At an applied potential of +0.50 V, it gives a fast response time (< 5 s) and a linear dependence ($R=0.997$) on the glucose concentration up to 7.0 mM with an extraordinarily high sensitivity of 1.77 mA mM^{-1} cm^{-2} and a detection limit of 2 μM. The interference by the oxidation of common interfering species such as ascorbic acid, dopamine, uric acid, lactose, and fructose is effectively avoided. The electrode was used to analyze glucose concentration in human serum samples. It allows highly sensitive, stable, and fast amperometric sensing of glucose, which is promising for the development of nonenzymatic glucose sensors.

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Introduction

Determination of dissolved glucose is of great importance in various areas [1–3]. In general, glucose oxidase (GOx) which is selected as a model enzyme in glucose detection has the advantage of high sensitivity and selectivity [4–10], which catalyzes the oxidation of glucose in the presence of O_2 to produce hydrogen peroxide for detection. However, because of the intrinsic nature of enzymes, the enzymatic glucose sensors often suffer from instability. For instance, the activity of GOx can be easily affected by temperature, pH value, humidity and toxic chemicals. In order to solve this problem, many nonenzymatic glucose sensors constructed by nanomaterials have been explored, which detect glucose based on its direct electrochemical oxidation [11–16]. Nanomaterials have become the focus of many scientific areas, including analytical science because of their unique electronic, optical, and catalytic properties [17].
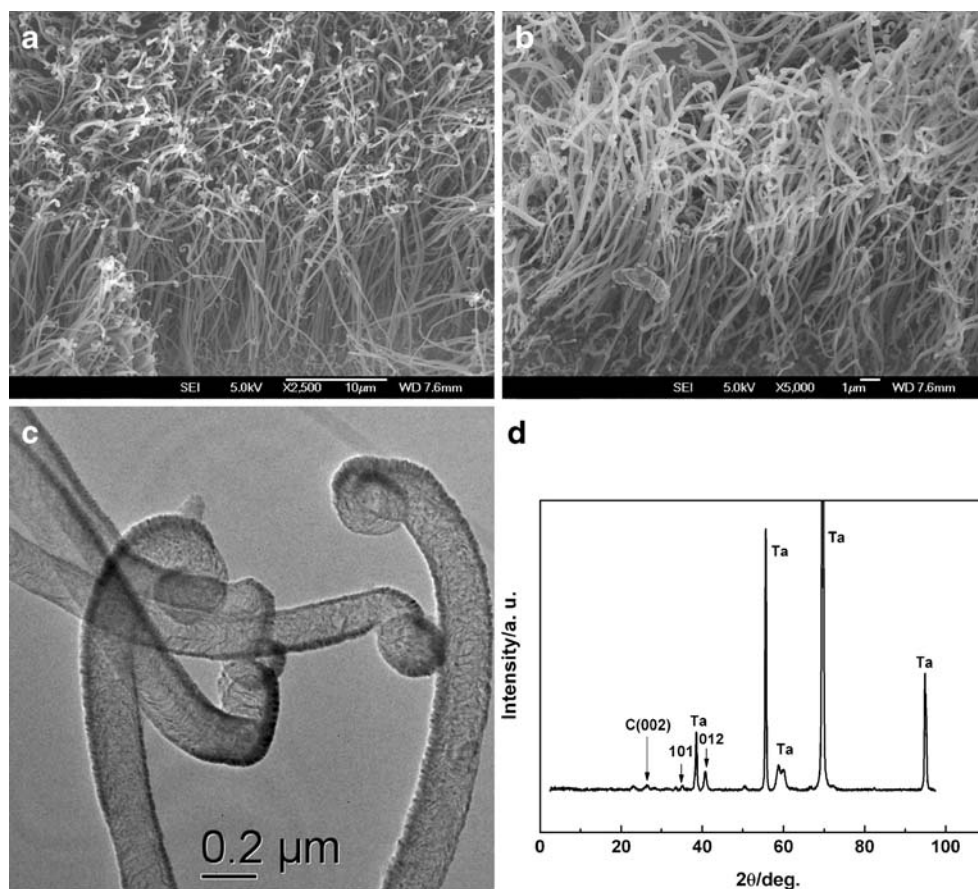
Carbon nanotubes (CNTs) have been used as functional nanomaterials because of their distinctive physical and chemical properties. Application of CNTs in biological and medical fields, particularly in electrochemical biosensors, has been expanding rapidly [18–20]. Moreover, CNTs are able to promote fast electron transfer kinetics for glucose. Therefore, nonenzymatic glucose sensors such as noble metal-MWCNTs (containing Pt [15, 21] and Au [22, 23]), alloys-MWCNTs (containing Pt, Pb and Au [24, 25]) electrodes were constructed. However, these kinds of conventional electrodes have the shortcoming of low electrocatalytic activity towards the oxidation of glucose

due to the adsorption of the interfering electroactive species such as ascorbic acid (AA), dopamine (DA), uric acid (UA) under physiological conditions, resulting in low sensitivity and poor selectivity [16, 26]. Metal oxide nanoparticles such as CuO [12, 27], ZnO [28], TiO$_2$ [29] have been successfully used for glucose sensors. In our previous work, we have successfully modified well-aligned MWCNTs with MnO$_2$ by electrodeposition for a glucose sensor [11].

In this paper, the MWCNTs are vertically aligned on Ta substrates [30–32], which efficiently simplifies the fabrication of electrode for electrochemical application. These stable, high-density, well-aligned MWCNTs with a large, electrochemically accessible surface area, high purity and high electrical conductivity, have been demonstrated to be high-performance electrode materials for electrochemical sensors [11, 25, 33, 34]. Here, we modified the well-aligned carbon nanotubes with NiO nanoparticles by magnetron sputtering deposition. The NiO/MWCNTs were directly used as an electrode to electrocatalyze the oxidation of glucose. The electrode takes the advantages of the electrocatalytic activity and high surface area of both NiO nanoparticles and carbon nanotubes. We have found that the electrode was very effective for glucose oxidation. Meanwhile, the interference from the oxidation of common

interferents such as ascorbic acid, dopamine, and uric acid was also tested. All the above results indicate the NiO/MWCNTs nanocomposite electrode can be used as a novel candidate for nonenzymatic glucose sensors.
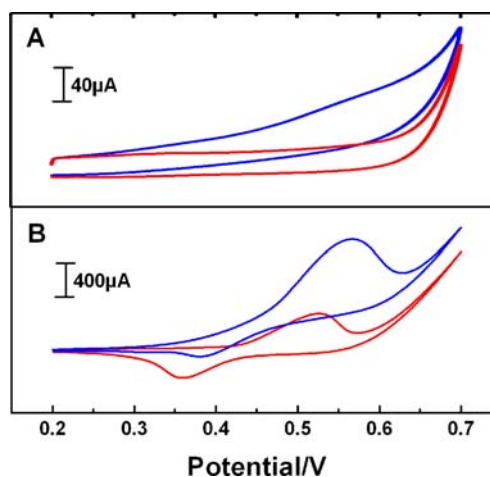
Fig. 2 CVs for 6 mM glucose at **a** MWCNTs electrode and **b** NiO/MWCNTs nanocomposite electrode in 0.10 M NaOH solution. The red lines represent the background response. Scan rate: 50 mV·s^{-1}
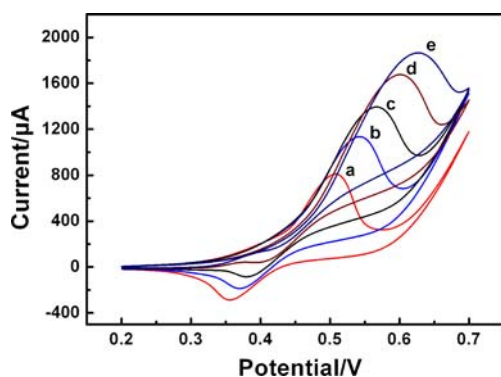
Fig. 3 CVs of the NiO/MWCNTs nanocomposite electrode in 0.10 M NaOH solution containing 2.0, 4.0, 6.0, 8.0 and 10.0 mM (**a**–**e**) glucose at 50 mV·s^{-1}

Experimental

D-(+)-glucose, L-ascorbic acid (L-AA), uric acid (UA), dopamine (DA), D-fructose and lactose were obtained from Alfa Aesar (http://www.alfa.com/china) and were used as received. All other reagents were of analytical grade. All solutions were prepared with high quality deionized water (18.4 MΩ cm^{-1}).

The growth and application of well-aligned MWCNTs for electrode have been reported elsewhere [30–34]. In the present work, the MWCNTs were modified with NiO nanoparticles by magnetron sputtering of metallic Ni in an atmosphere of O_2 and Ar (3:1) [35, 36].

A field-emission scanning electron microscope (SEM) (JEOL JSM 6700F) was used to observe the NiO modified well-aligned MWCNTs arrays. Transmission electron microscopy (TEM) observation of NiO/MWCNTs was performed with a Philips CM 300 FEG instrument operated at 300 kV. X-ray diffraction (XRD) pattern was recorded on a Bruker GADDS diffractometer with an area detector operating under a voltage of 40 kV and a current of 40 mA using Cu Ka radiation (λ=0.15418 nm). Electrochemical measurements were performed using CHI 660C electrochemical workstation (Shanghai Chenhua, China). A three-electrode system was employed with MWCNTs or NiO/MWCNTs electrode as working electrode, an Ag/AgCl

(3 M KCl) electrode and a platinum wire served as reference electrode and counter electrode, respectively. All potentials were referred to Ag/AgCl (3 M KCl) electrode.

Results and discussion

### Morphological characterization of NiO/MWCNTs nanocomposite

The typical SEM images of pristine MWCNTs and NiO/MWCNTs are shown in Fig. 1a and b, respectively. It can be seen that the surface of the MWCNTs are homogeneously coated after magnetron sputtering deposition. TEM observations were carried out for a more detailed characterization. Fig. 1c provides an overview of modified nanotubes, clearly showing that the nanotubes are uniformly coated with a thin layer of NiO nanoparticles. This is in agreement with the observation by SEM.

The crystal structure of the NiO/MWCNTs nanocomposite was studied by X-ray diffraction analysis, as shown in Fig. 1d. The XRD pattern shows diffractions of very strong peaks of the Ta substrate at 2θ of 38.5º, 55.6º, 69.6º, 94.9º and a very weak one at 2θ of 26.3º, which is graphite structure of MWCNTs ((002) planes). In addition, two small peaks at about 2θ of 43.3º and 37.3º, which belong to NiO (012), (101) planes, respectively, also appeared in the XRD pattern. The XRD result clearly indicates that NiO nanoparticles were deposited on the surface of MWCNTs.

CVs investigation of NiO/MWCNTs nanocomposite electrode

The electrocatalytic performance of both the bare MWCNTs and NiO/MWCNTs nanocomposite electrodes toward the oxidation of glucose was investigated by cyclic voltammetry (CV). Figure 2 shows the CV behaviors of the bare MWCNTs (A) and NiO/MWCNTs (B) electrodes in the absence (red lines) and presence (blue lines) of 6.0 mM glucose in 0.10 M NaOH solution at 50 mV s^{-1}. As shown in Fig. 2a, only a small current for glucose oxidation without

Fig. 4 A Current–time response obtained on increasing the glucose concentration at 1.0 mM steps at (a) MWCNTs electrode and (b) NiO/MWCNTs electrode. **B** The dependence of the current response verses the concentration of glucose at (a) MWCNTs nanocomposite electrode and (b) NiO/MWCNTs electrode. Applied potential: +0.50 V
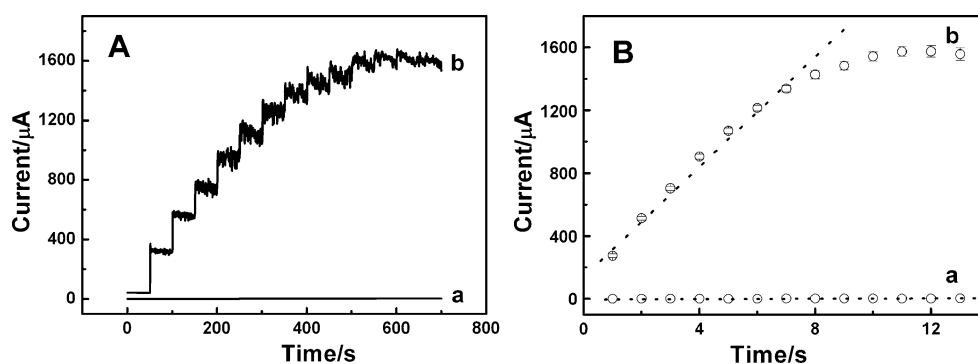
**Table 1** Comparison of the present CuO/MWCNTs electrode with different nonenzymatic glucose sensors

Electrode	Detection potential	Sensitivity	Linear range	Detection limit	Ref.
Freestanding SWCNT film	+0.6 V	248.6 μA mM^{-1} cm^{-2}	0.01-2.16 mM	0.01 mM	[37]
MnO$_2$/MWCNTs	+0.30 V	33.19 μA mM^{-1}	10 μM–28 mM	–	[11]
Mesoporous Pt	+0.4 V	9.6 μA mM^{-1} cm^{-2}	0–10 mM	–	[38]
Porous Au	+0.35 V	11.8 μA mM^{-1} cm^{-2}	2–10 mM	5 μM	[39]
MWCNTs	+0.20 V	4.36 μA mM^{-1} cm^{-2}	2 μM–11 mM	1 μM	[40]
Pt nanotubule	+0.4 V	0.1 μA mM^{-1} cm^{-2}	2–14 mM	1.0 μM	[41]
Pt-Pb/CNTs	+0.30 V	17.8 μA mM^{-1} cm^{-2}	Up to 11 mM	1.8 μM	[42]
CuO nanowires	+0.33 V	0.49 μA mM^{-1} cm^{-2}	0.4 μM–2.0 mM	0.049 μM	[12]
Ni nanowires	+0.55 V	1043 μA mM^{-1} cm^{-2}	0.5 μM–7 mM	0.1 μM	[43]
NiO/MWCNTs	+0.50 V	1768.8 μA mM^{-1} cm^{-2}	10 μM–7 mM	2 μM	This work

peak is observed at the bare MWCNTs electrode. When the NiO/MWCNTs nanocomposite electrode was used (Fig. 2b), a couple of oxidative and reductive peaks are observed in the absence of glucose. This can be attributed to the redox couple of Ni(II)/Ni(0). When adding 6.0 mM glucose, the oxidative peak current was dramatically increasing opposite to the reductive peak current. The anodic peak at +0.55 V is due to oxidation of glucose catalyzed by NiO nanoparticles. The cathodic peak at +0.37 V represents the reduction of Ni(II) to Ni. Compared to the bare MWCNTs electrode, we can see that the starting oxidative potential had no change but a dramatic increase of current signal was recorded at the NiO/MWCNTs electrode. This may result in the strong interaction between NiO nanoparticles and MWCNTs. The presence of NiO nanoparticles increased the effective surface area and active sites for glucose oxidation reaction, and also made the electrode surface more porous for facilitating electron transfer. The character of facilitated electron transfer at MWCNTs can further improve the electrochemical reaction at the NiO/MWCNTs nanocomposite electrode. Therefore, the NiO/MWCNTs nanocomposite showed high electrochemical activity to the oxidation of glucose.

The CVs of the NiO/MWCNTs nanocomposite electrode in different concentrations of glucose were also measured, as indicated in Fig. 3. The currents of the oxidative peak increased upon the increase of glucose concentration but opposite to the reductive peak current, since part of Ni(II) was reduced by glucose in the anodic process. Meanwhile, the oxidative potential shifted positively. In addition, the influence of scan rate on the current response for the anodic current (I_{pa}) of glucose oxidation was also studied (see Electronic Supplementary Material). At the NiO/MWCNTs electrode, the currents of the oxidative peak increased with the increase of scan rate, and the currents of the oxidative peak were linearly proportional to the scan rate in the range of 10–150 mV s^{-1}, indicating a surface-controlled electrode process.

Amperometric response of glucose

The optimized potential for the electrochemical oxidation of glucose at the NiO/MWCNTs electrode was determined to be +0.50 V in 0.20 M NaOH (Fig. S2 and S3 in Electronic Supplementary Material). Figure 4A displays the amperometric response (at +0.50 V) at (a) the MWCNTs nanocomposite electrode and (b) the bare NiO/MWCNTs electrode to the successive addition of 1.0 mM glucose in 0.20 M NaOH solution. One can see that the NiO/MWCNTs nanocomposite electrode shows well defined response to the change of glucose concentration, producing steady-state signals within 5 s. The regression equation is i_{pa} (μA)=152.35 + 176.88c (mM), with R=0.995. The NiO/MWCNTs nanocomposite electrode displays a linear range of 10 μM to 7.0 mM with a high sensitivity of 1,768.8 μA mM^{-1} cm^{-2}, as illustrated in Fig. 4B. The detection limit is 2 μM with a signal/noise of 3. The sensitivity is higher than that of SWCNTs, MWCNTs, and nonenzymatic glucose sensors reported in the literature [11,
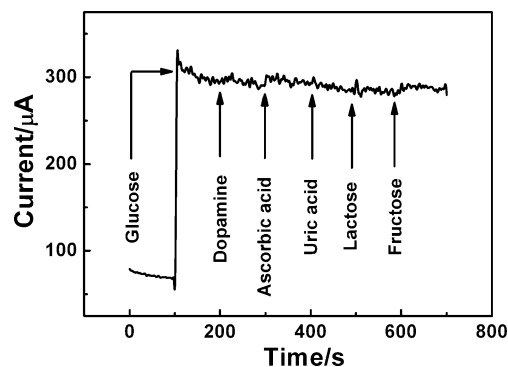
Fig. 5 Flow injection amperometric response to injections of 1.0 mM glucose, 0.05 mM DA, AA, UA, lactose and fructose at the NiO/MWCNTs electrode under +0.50 V
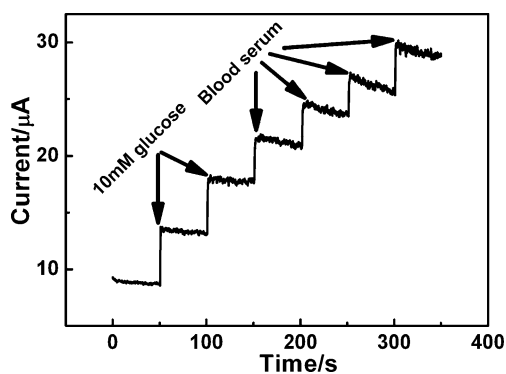
Fig. 6 The amperometric I-t curve for glucose and real serum sample

37–44], which are summarized in Table 1. The high performance of the NiO/MWCNTs electrode towards electroanalysis of glucose can be attributed to the increase of electroactive surface area and the synergistic electrocatalytic activity by combining NiO nanoparticles with well-aligned MWCNTs.

To evaluate the selectivity of the biosensor, many interfering biomolecules, AA, DA, UA, and other carbohydrates which normally co-exist with glucose in real samples (human blood) were examined. Considering that the concentration of glucose in the human blood is about 30 times that of AA, DA or UA, the amperometric response of the NiO/MWCNTs nanocomposite electrode towards the addition of 1.0 mM glucose and 0.05 mM AA, DA, UA and other carbohydrates such as D-fructose and lactose was examined in 0.20 M NaOH solution and the response currents are 227.8, 1.5, 2.7, 0.3, 0.8, and 3.3 µA, respectively (Fig. 5). The results indicate that the current response of the interferents is only 0.3–3.3% of that of glucose at the NiO/MWCNTs nanocomposite electrode. The current response of the interferents at the NiO/MWCNTs electrode is also much lower than that at the bare MWCNTs electrode. The selectivity was improved so much that the interference caused by the common interfering molecules, AA, DA, UA and other carbohydrates was negligible to the response of glucose at the NiO/MWCNTs nanocomposite electrode. Therefore, the selectivity of the NiO/MWCNTs nanocomposite electrode for glucose detection is satisfied at the presence of possible interfering reagents. The reproducibility and stability of the sensor were also evaluated. Three NiO/MWCNTs nanocomposite electrodes were identically investigated in the current response for 1.0 mM glucose at +0.50 V. The relative standard deviation (R.S.D.) was 5.2%, confirming that the sensors are highly reproducible. An electrode was measured 25 times and the yielded error is about 9%, showing that the sensor was stable without undergoing poisons by the oxidation product, and can be used for the repetitive detection of glucose. Moreover, the NiO/MWCNTs nanocomposite electrode was not affected by chlorine ion with its concentration up to 1.0 M, indicating that it can be used as a glucose sensor for many real samples.

Real serum samples measurement

The NiO/MWCNTs electrode was applied to the determination of glucose in real blood serum samples. 40.0 µL of serum sample was added to 10.0 mL of 0.20 M NaOH solution, and the current response was recorded at +0.50 V, as indicated in Fig. 6. Table 2 summarizes the result of determination of glucose in three serum samples. The experimental results indicate that the NiO/MWCNTs electrode is promising for practical application as glucose sensor.

Conclusion

In summary, a glucose biosensor based on NiO/MWCNTs nanocomposite was successfully fabricated by magnetron sputtering deposition of NiO nanoparticles on the vertically aligned carbon nanotubes. Because of synergistic electrocatalytic activity by combining NiO nanoparticles with MWCNTs, the NiO/MWCNTs nanocomposite electrode displays higher electrochemical activity for the glucose oxidation than the bare MWCNTs electrode, showing a fast response time (< 5 s) and a linear dependence ($R=0.995$) in the glucose concentration range of 10 µM to 7 mM with a very high sensitivity of 1768.8 µA mM^{-1} cm^{-2} at an applied potential of +0.50 V. The detection limit is 2 µM with a signal/noise of 3. Meanwhile, the interference from the oxidation of common interfering species such as ascorbic acid, dopamine, uric acid and other carbohydrates is effectively avoided. Thus, the NiO/MWCNTs nanomaterials are promising for the fabrication of novel nonenzymatic glucose sensors.

Table 2 Determination of glucose in human blood serum samples

Sample	Sensor (mM)	RSD (%)	Added (mM)	Recovery (%)
1	7.8	4.7	0.1	93
2	7.6	3.9	0.1	96
3	5.8	3.2	0.1	95

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