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ORIGINAL PAPER

Enzyme-free glucose sensor using a glassy carbon electrode modified with reduced graphene oxide decorated with mixed copper and cobalt oxides

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Abstract We describe a binary porous catalyst consisting of a reduced graphene oxide (rGO) support decorated with mixed Cu-Co oxides. It was synthesized electrochemically and characterized by scanning electron microscopy, energy dispersive X-ray spectroscopy and cyclic voltammetry. The nanocomposite was deposited on a glassy carbon electrode (GCE) where it exhibits better electrocatalytic activities for glucose oxidation compared to GCEs modified with CuO_x or CoO_x only on an rGO support. The improved electrocatalytic activities are believed to result from the synergistic effect of CuO_x-CoO_x binary catalyst, the high conductivity of rGO support, and the porous scaffold. The amperometric sensor, operated in 0.1 M NaOH at a working potential of +0.5 V (vs. SCE), displays a calibration plot for glucose that is linear in the 5 to 570 μM concentration range, and the detection limit is 0.5 μM. The performance of the sensor was evaluated by determination of glucose in (spiked) human urine.

Keywords Nanocomposite · Nanoporous material · Nonenzymatic sensing · Binary catalyst · Electrocatalytic oxidation · Cyclic voltammetry · Scanning electron microscopy · Energy dispersive X-ray spectroscopy

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Introduction

Glucose is the primary source of energy for the body and its determination has important significance in clinical diagnosis, health protection and control of food production processes [1]. The increasing demand for glucose sensors with high sensitivity and stability, fast response, low cost, excellent selectivity and reproducibility has driven tremendous research efforts from analytical scientists. Among various glucose sensors, the commercial enzymatic sensors are promising due to their simple equipment, convenient operation, good selectivity and high sensitivity for glucose detection [2]. However, owing to the intrinsic nature of enzymes, enzymatic glucose sensors are often limited by their instability, high cost, complicated immobilization procedures, and critical operating conditions [3, 4]. As an alternative strategy to avoid the above-mentioned drawbacks, enzyme-free glucose sensors have aroused enormous interest and several reviews have summarized the recent advances in this field [5, 6].

The advancement for enzyme-free glucose sensors strongly depend on research into high performance electrode material. So far, noble metals (Pd, Pt, Au) and their alloys [7–10], transition metal or their oxides (Ni or NiO, CuO or Cu_2O, Co_3O_4, MnO_2, Fe_3O_4) [11–23] have been reported as electrode modifications. The high-cost noble metals (such as Pt) can perform in neutral pH values for glucose sensing, which is fascinating for blood glucose monitor. However, they often suffer from very slow kinetics, and the surfaces of which are easily poisoned by adsorbed chloride ions and chemisorbed intermediates that originate from the glucose oxidation process, resulting in decreased sensitivity and poor operational stability [5]. Therefore, increasing attention has been focused on fabricating high-performance enzyme-free devices using inexpensive and resourceful transition-metal catalysts due to their high electrocatalytic activity, good stability, low cost, abundance and high anti-poisoning resistance toward intermediate compounds and

chloride ions. Graphene have been widely used as scaffolds for loading of monometallic electrocatalysts for biosensing applications [24–28]. Another alternative is binary electrocatalyst prepared from two different metals, which are also drawing much research interest because of their better catalytic activities and anti-interference ability for glucose detection than their corresponding monometallic counterparts due to the synergistic enhancement effect of two metallic materials. Recently, Awad's group developed a binary catalyst of NiO_x/MnO_x as an anode for an amplified electrochemical oxidation of glucose in alkaline solutions [29]. Yuan et al. reported Cu_2O/NiO_x/graphene oxide modified electrode for the enhanced electrochemical oxidation of reduced glutathione and nonenzymatic glucose sensor [30]. Wang et al. developed Ni-Co nanostructures coated reduced graphene oxide nanocomposites electrode for nonenzymatic glucose biosensing [31]. Dong et al. prepared graphene foam loaded nickel-cobalt hydroxides nanoflakes ($Ni_xCo_{2x}(OH)_{6x}$/graphene foam) and showed its superior capability for applications in supercapacitor and electrochemical sensor [32]. These results indicate that the rational design and optimal component for preparation of binary catalyst is important in achieving high-performance electrochemical sensors.

We present a binary catalyst of Cu-Co oxides decorated reduced graphene oxide (CuO_x-CoO_x/rGO) composites synthesized by a facile two-step electrochemical approach and demonstrated its application in enzyme-free glucose sensors. In the first step, the bimetallic Cu-Co/rGO composites was firstly fabricated by solution-casting of graphene oxide (GO) nanosheets on glassy carbon electrode, followed by simultaneous electrochemical reduction of GO and electrodeposition of Cu-Co binary nanostructures on it. In the second step, the resultant Cu-Co/rGO composites were subjected to be oxidized in situ into CuO_x-CoO_x/rGO composites by the cyclic potential sweep technique. The present strategy for the synthesis of CuO_x-CoO_x/rGO composites by direct electrochemical method endows the intimate contact between the electroactive materials and the current collector, which facilitates the diffusion of active species and the transport of electrons. The electrocatalytic properties of the fabricated CuO_x-CoO_x/rGO composites modified electrode for glucose oxidation were systematically assessed. Our results showed that the CuO_x-CoO_x/rGO composites demonstrated better electrocatalytic activities toward glucose oxidation in comparison with those of only CuO_x or CoO_x on rGO sheets. Therefore, due to the synergistic effect of CuO_x or CoO_x and the highly electrical conductivities of rGO, a CuO_x-CoO_x/rGO nanocomposites based enzyme-free glucose sensor was fabricated, which showed good performances for glucose detection, including high sensitivity, good stability, fast response, and excellent selectivity. Importantly, the simple and facile fabrication method of the present sensor makes it competitive to other glucose sensors.

Experimental

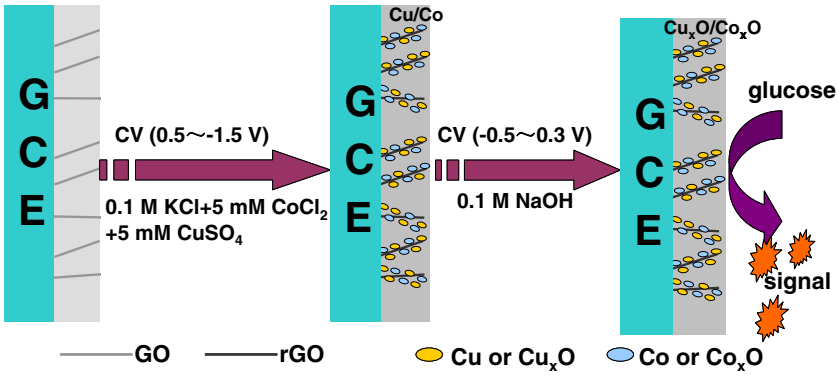
Reagents and apparatus

GO was purchased from Nanjing XFNANO Materials Tech Co., Ltd. (China, http://www.xfnano.com/). Glucose and interference species were purchased from Sigma-Aldrich (http://www.sigmaaldrich.com/china-mainland.html). $CuSO_4 \cdot 5H_2O$, Cobalt (II) chloride anhydrous, KCl, NaOH and H_2O_2 were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China, http://www.sinoreagent.com.cn/). All of these reagents were of analytical grade and used as received. Unless otherwise stated, ultrapure water (18.2 MΩ cm) produced by a Milli-Q system was used as the solvent throughout this work.

Scanning electron microscopy (SEM) and energy dispersive X-ray spectroscopy (EDS) were conducted by EDS-integrated JSM-6701F (Japan, http://www.jeol.cn/) for surface morphology observations and surface elemental composition analysis. Electrochemical experiments were performed on a CHI 660D electrochemical station (Shanghai Chenhua, China, http://chi.instrument.com.cn) with a conventional three-electrode system. The CuO_x-CoO_x/rGO modified glassy carbon electrode (CuO_x-CoO_x/rGO/GCE) was used as the working electrode. A Pt wire and a saturated calomel electrode (SCE) acted as the counter and reference electrodes, respectively.

Fabrication of CuO_x-CoO_x/rGO modified electrode

The obtained GO was dispersed in water, giving a yellow-brown dispersion with a concentration of 1 mg mL^{-1} by an ultrasonic technique. A 10 μL portion of the resulting GO dispersion was dropped onto a pretreated bare GCE and dried at room temperature to obtain the GO modified GCE (GO/GCE). Here, prior to the surface modification, GCE was polished with 0.3 and 0.05 μm alumina slurries respectively, and then ultrasonically cleaned with ethanol and double distilled water for 10 min to remove the physically adsorbed substance. Next, the electrochemical reduction of GO on electrode surface and the simultaneous electrodeposition of Cu and Co nanostructures were performed in a 0.1 M KCl aqueous solution containing 5 mM $CoCl_2$ and 5 mM $CuSO_4$ in the potential range from 0.5 to −1.5 V (vs. SCE) for 10 cycles at a scan rate of 50 mV s^{-1} by cyclic voltammetry (CV). The electrode prepared in this step was denoted as Cu-Co/rGO composites modified electrode. The electrode was then rinsed several times with water and dried with a flow of N_2 before it was repeatedly scanned in a 0.1 M NaOH with CV under the potential range of −0.5 to 0.3 V at 50 mV s^{-1} for 60 cycles, allowing the Cu-Co nanostructures to be oxidized into CuO_x-CoO_x nanostructures. The whole process for preparation of CuO_x-CoO_x/rGO/GCE is illustrated in Scheme 1. For



Scheme 1 Schematic illustration for the fabrication and application of CuO_x-CoO_x/rGO modified glassy carbon electrode (GCE) for glucose sensor. GO and rGO in the scheme are the acronyms of graphene oxide and reduced graphene oxide

comparison, CuO_x/rGO/GCE and CoO_x/rGO/GCE were prepared in deposition solutions of 0.1 M KCl + 10 mM $CuSO_4$ and 0.1 M KCl + 10 mM $CoCl_2$, respectively, by the similar procedure as described above. The total electrolyte concentration was also maintained as 10 mM to minimize the difference about deposition conditions but highlighted the component difference between monometallic oxides/rGO/GCE and bimetallic oxides/rGO/GCE (deposition solutions of 0.1 M KCl + 5 mM $CuSO_4$ + 5 mM $CoCl_2$).

Electrochemistry measurements

Cyclic voltammetry (CV) and amperometric experiments were carried out at room temperature. A certain volume of stock solution of glucose and 10 mL supporting electrolyte were added into an electrochemical cell, and then the three-electrode system was inserted into the cell. The CV was carried out to investigate the electrochemical response of the modified electrode toward glucose. The amperometric experiment was performed to achieve the quantitative analysis.

Results and discussion

Formation mechanism and characterization of CuO_x-CoO_x/rGO composites

The Cu-Co binary catalyst was prepared on GO/GCE by CV technique in the first step. Figure 1 shows the cyclic voltammograms (CVs) of the GO/GCE scanned in 0.1 M KCl solution containing 5 mM $CoCl_2$ and 5 mM $CuSO_4$ at a potential range from 0.5 to −1.5 V for 10 cycles. As observed, there is a large cathodic peak at −1.36 V in the first cycle due to reduction of the high number of oxygen-containing functional groups in GO. With further increasing the scanning cycles, the cathodic currents decrease considerably and disappear after several potential cycles, indicating that GO has been electrochemically reduced to rGO [33–35]. The obvious reduction peak at −0.88 V in the first cycle is resulted from the reduction of Co^{2+} to Co(0), which is accordance with previous results [31, 36]. The two redox peaks located at −0.20/0.17 V and −0.01/0.43 V are corresponded to the conversion between three different valence state of copper of Cu(0), Cu(I) and Cu(II). The current decrease of both characteristic peak of Co and Cu with increasing cycle number indicated that Co^{2+} and Cu^{2+} have been reduced and deposited on the surface of rGO/GCE slowly. After the resultant electrode was subjected to be scanned in NaOH with CV for oxidizing binary Cu-Co to form CuO_x-CoO_x/rGO composites, the obtained CuO_x-CoO_x/rGO/GCE was scanned in 0.1 M NaOH solution under the potential range from −1.5 to 0.5 V to characterize the successful deposition of binary CuO_x-CoO_x on rGO surface. As shown in Fig. 1b, it is clear that several anodic and cathodic peaks appeared, among which the anodic peaks at −0.40 and −0.15 V represent the conversion of Cu(0) to Cu(I) and of Cu(I) to Cu(II), respectively. The corresponding cathodic peaks at −0.38 and −0.84 V are attributed to the transition of Cu(II) to Cu(I) and of Cu(I) to Cu(0), respectively. This typical redox peaks of copper is very similar to those of previous results [37, 38]. Furthermore, the other two anodic peaks at 0.04 and 0.18 V and the cathodic peak at 0.15 V corresponds to the conversion between different cobalt phases, such as Co(0), CoO(II) and CoOOH(III) [36, 39]. The CV results demonstrate that CuO_x-CoO_x binary catalyst has been successful deposited on rGO support using the present method.

The morphological characterization of the modified electrodes is disclosed by SEM imaging and shown in Fig. 2. Image of Fig. 2a and b show the typical SEM micrographs of CuO_x/rGO/GCE at different magnifications. It reveals that the CuO_x is deposited as nanoparticles in a uniform distribution with an average particle size about 50 nm on wrinkled rGO surface. This average particle size of CuO_x was measured from about 100 nanoparticles in Fig. 2a. Figure 2 c and d show SEM images of CoO_x/rGO/GCE. A flower-like shape constituted by several CoO_x nanoflakes with thickness of about 20 nm can be deposited on rGO surface. In image of Fig. 2e (CuO_x-CoO_x/rGO/GCE sample), a three-dimensional (3D) nanocomposites composed with wrinkled rGO, nanoparticles and nanoflakes were obtained. The CoO_x nanoflakes can

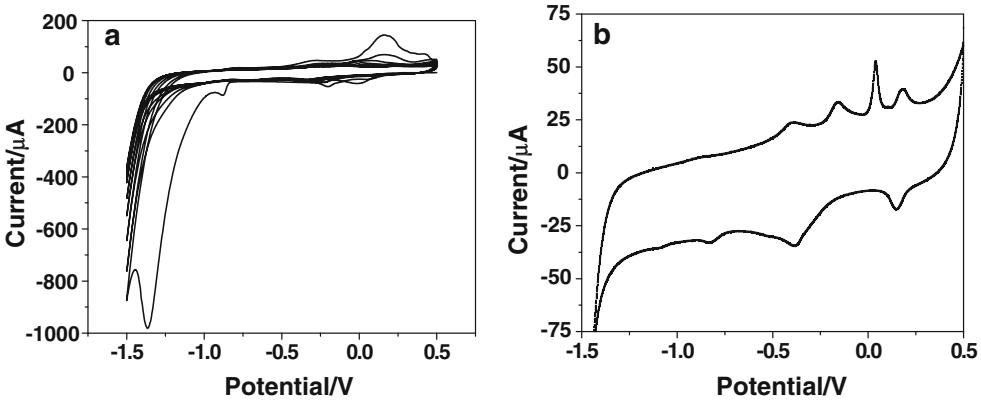


Fig. 1 **a** CVs of GO/GCE in 0.1 M KCl solution containing 5 mM $CoCl_2$ and 5 mM $CuSO_4$ for 10 cycles, **b** CV of CuO_x-CoO_x/rGO/GCE in 0.1 M NaOH solution. The scan rate is 50 mV s^{-1}

induce the formation of CuO_x nanoparticles and thus some aggregated nanoparticles cover with the nanoflakes to form a porous and interconnected network. The obtained 3D porous structures provide probability for enhancing glucose adsorption and facilitating transport of analyte species. The EDX spectra shown in Fig. 2f reveal the presence of the elements oxygen, Cu and Co. This proves the successful deposition of the composite on the surface of rGO.

Electrocatalytic oxidation of glucose on CuO_x-CoO_x/rGO composites modified electrode

To evaluate the catalytic properties of the binary catalyst in the oxidation of glucose, CVs were recorded for the rGO/GCE (a), CoO_x/rGO/GCE (b), CuO_x/rGO/GCE (c) and CuO_x-CoO_x/rGO/GCE (d) in 0.1 mol L^{-1} NaOH in the absence (black curve) and the presence (red curve) of 5 mM glucose

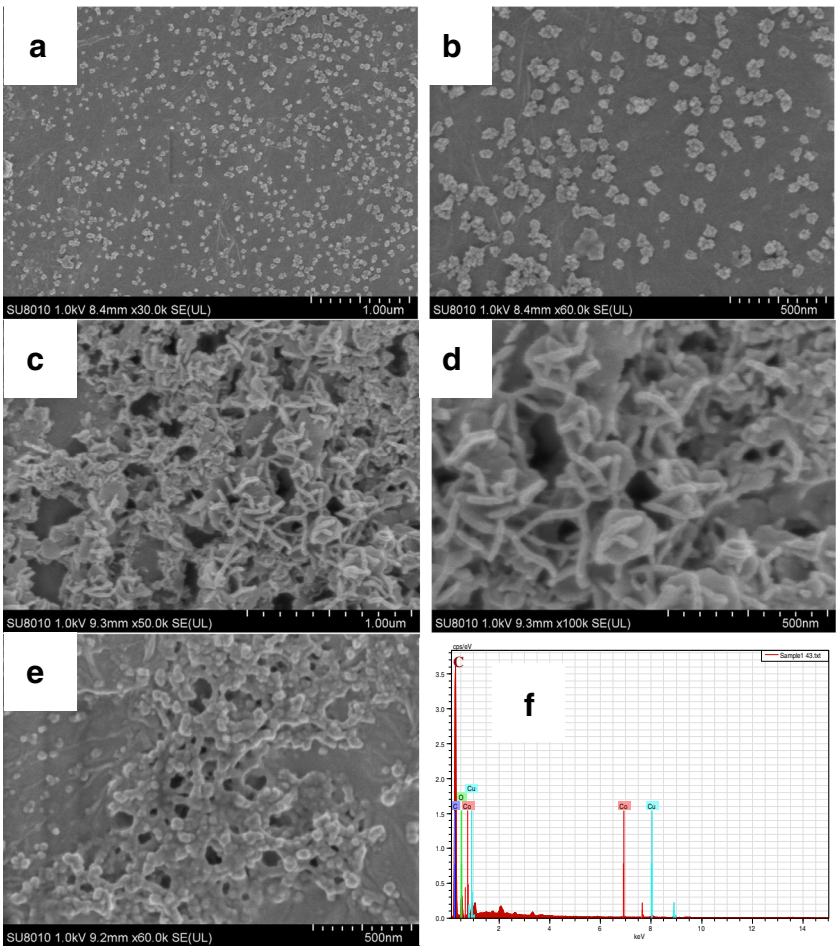


Fig. 2 SEM images of CuO_x/rGO/GCE (**a**, **b**), CoO_x/rGO/GCE (**c**, **d**), CuO_x-CoO_x/rGO/GCE (**e**) at different magnification and the EDX spectra of CuO_x-CoO_x/rGO/GCE (**f**)

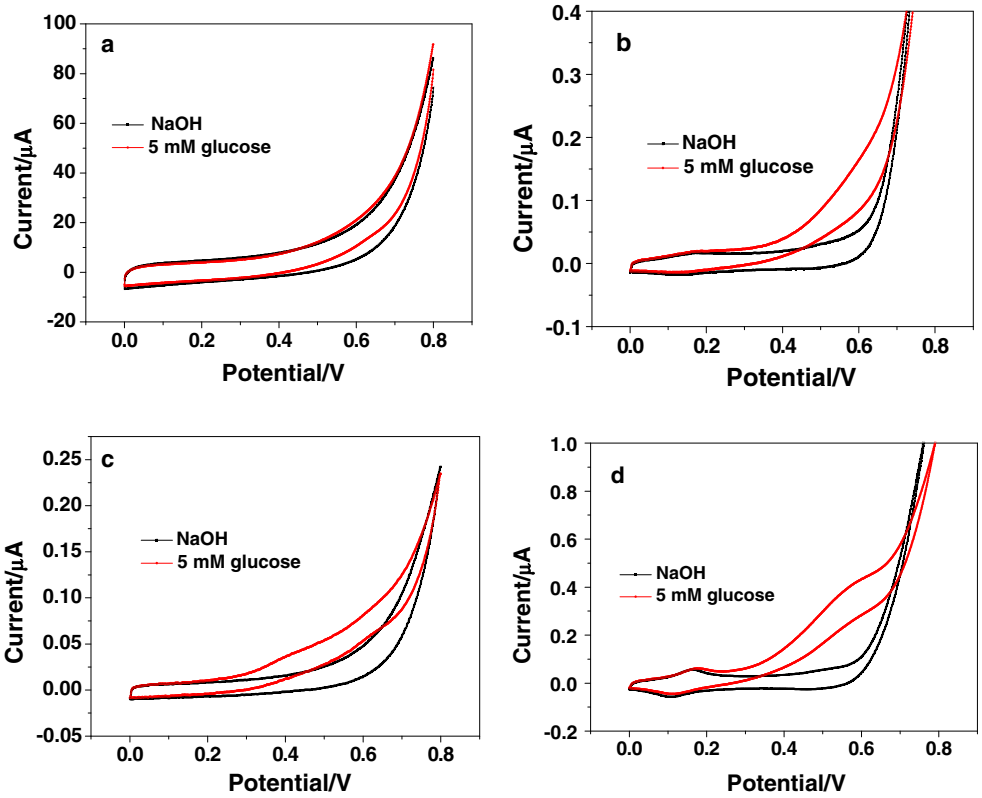


Fig. 3 CVs of rGO/GCE (**a**), CoO_x/rGO/GCE (**b**), CuO_x/rGO/GCE (c) and CuO_x-CoO_x/rGO/GCE (d) in 0.1 mol L^{-1} NaOH in the absence (*black curve*) and the presence (*red curve*) of 5 mM glucose at a scan rate of 50 mV s^{-1}

at a scan rate of 50 mV s^{-1} (Fig. 3). As observed from Fig. 3a, no obvious current change upon addition of glucose into blank solution, indicating that the electrocatalytic activities of rGO toward glucose oxidation is limited. The CVs of CoO_x/rGO/GCE in blank solution shows a redox peak at +0.15/+0.17 V, which corresponds to the redox couples of Co itself. Glucose addition causes an obvious anodic current increase starting at about 0.30 V due to the irreversible glucose oxidation catalyzed by CoO_x nanoflakes. An observable increase in anodic current, starting at about +0.23 V for CuO_x/rGO/GCE after the addition of glucose also shows efficient electrocatalytic activities of CuO_x nanoparticles toward glucose oxidation. When the two electrocatalysts were integrated together to form a binary catalyst at CuO_x-CoO_x/rGO/GCE, a noticeable and significant increase in anodic current, corresponding to glucose oxidation, starts at about +0.19 V, which is more negative than the monometallic oxides on graphene surface (+0.23 V for CuO_x/rGO/GCE and +0.30 V CoO_x/rGO/GCE). In addition, the anodic current of 5 mM glucose at CuO_x-CoO_x/rGO/GCE is the largest among the four electrodes. All these results demonstrate that the electrocatalytic activity of CuO_x-CoO_x/rGO composites is greatly enhanced in comparison with those of CuO_x or CoO_x on rGO sheets due to synergistic enhancement effect.

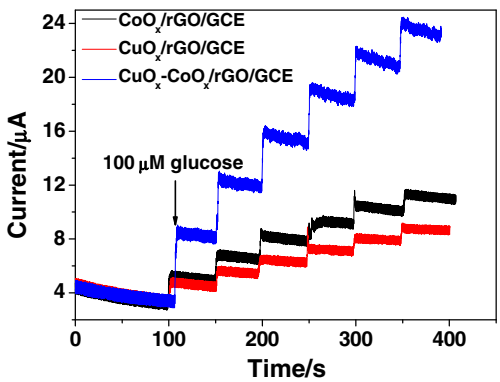


Fig. 4 The amperometric responses of CoO_x/rGO/GCE, CuO_x/rGO/GCE, CuO_x-CoO_x/rGO/GCE upon the successive additions of 100 μM glucose at applied potential of +0.5 V

The oxidation of glucose at the Cu and Co electrodes in an alkaline medium is generally considered a multi-step process, where strongly oxidizing Cu(III) and Co(IV) species (CuOOH and CoO_2) are responsible for its electrocatalytic behavior [28, 37]. According to previous results, CuO and CoOOH is electrochemically oxidized to Cu(III) and Co(IV) species.

$$CuO + OH^- \rightarrow CuOOH + e^-$$
$$CoOOH + OH^- \rightarrow CoO_2 + H_2O + e^-$$

Then, glucose is oxidized by Cu(III) and Co(IV) species to produce glucolactone that finally hydrolyses in aqueous medium to form gluconic acid.

$$CuOOH + \text{glucose} \rightarrow CuO + OH^- + \text{glucolactone}$$
$$2CoO_2 + \text{glucose} \rightarrow 2\ CoOOH + \text{glucolactone}$$

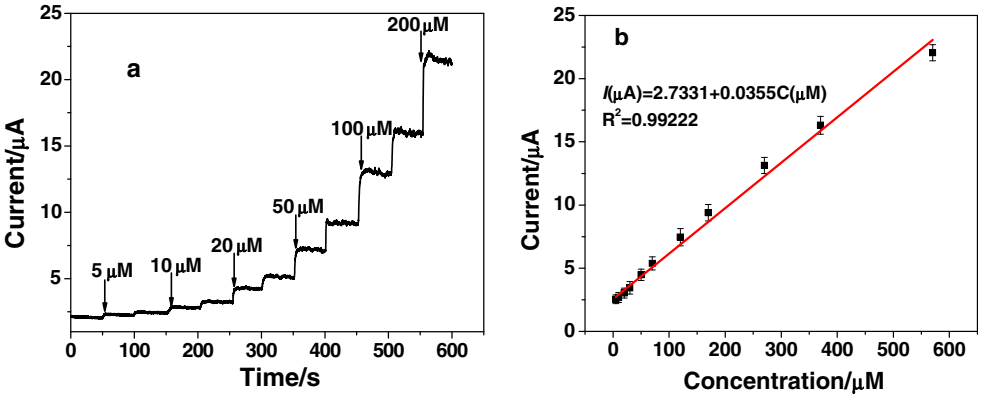


Fig. 5 **a** Amperometric response of the CuO_x-CoO_x/rGO/GCE (holding at +0.50 V) upon addition of glucose to increasing concentrations in 0.1 M NaOH, **b** The corresponding calibration curve

To further validate the synergistic enhancement effect of electrocatalytic behavior of CuO_x and CoO_x, amperometric responses of CoO_x/rGO/GCE, CuO_x/rGO/GCE and CuO_x-CoO_x/rGO/GCE upon successive additions of 100 μM glucose in 0.1 M NaOH at a constant potential of +0.50 V were recorded in Fig. 4. As observed, the detection sensitivity of glucose at CoO_x/rGO/GCE, CuO_x/rGO/GCE and CuO_x-CoO_x/rGO/GCE, was calculated to be 15.98, 10.07 and 42.03 μA μM^{-1}, respectively. The largest response sensitivity for glucose was obtained at CuO_x-CoO_x/rGO/GCE, which is consistent with the CV results. Adding the current of CoO_x/rGO/GCE and CuO_x/rGO/GCE together, it is found that this value (26.05 μA μM^{-1}) is still smaller than that of CuO_x-CoO_x/rGO/GCE (42.03 μA μM^{-1}). This further indicates that a synergistic enhancement effect between CoO_x and CuO_x occurs toward the electrochemical oxidation of glucose. Such enhanced electrocatalytic performance of the CuO_x-CoO_x/rGO composites may be ascribed to a synergistic effect between CuO_x nanoparticles and CoO_x nanoflakes, which includes more catalytic active sites for the glucose oxidation. Furthermore, the porous and interconnected network of composites and high conductivity of rGO provide convenient electron transfer channel and highly effective catalytic sites to facilitate the electrooxidation of glucose correspondingly.

Amperometric sensing of glucose and interference test

Before quantitative analysis, we investigate the effect of applied potentials (+0.3, +0.4, +0.5, +0.6, +0.7 V) on amperometric currents of 100 μM glucose (data not shown). The results demonstrated that +0.5 and +0.6 V have equal amperometric currents for 100 μM glucose and the current is the largest among all of the above potentials. Considering that the high detection sensitivity as well as the fact that serious interference at high applied potentials, +0.5 V was chosen as the optimum applied potential for glucose detection. Figure 5a presents the amperometric response at the CuO_x-CoO_x/rGO composites electrode upon addition of glucose to increasing concentrations in 0.1 M NaOH at an applied potential of +0.5 V, from which rapid response to the addition of glucose (within 3 s) can be obtained. The response to glucose shows a good linear range from 5 μM to 570 μM with a correlation coefficient of 0.9922 and a slope of 35.5 μA mM^{-1} (as shown in Fig. 5b). Based on the previous work described by Shi et al. [40], the electrode surface area of the CuO_x-CoO_x/rGO composites electrode can be calculated to be about 0.07 cm^2. The sensitivity of the sensor is calculated to be 507 μA mM^{-1} cm^{-2}, which is higher than the reported values at Fe_3O_4 nanorod arrays (Fe_3O_4 NRAs) electrode (406.9 μA mM^{-1} cm^{-2}) [23], CoO_xNPs/ERGO modified electrode (79.3 μA mM^{-1} cm^{-2}) [28], RGO-$Ni(OH)_2$ modified

Table 1 Comparison of enzyme-free glucose sensing performances based on various nanomaterials

Electrode materials	Response time (s)	Potential (V)	Sensitivity (μA mM^{-1} cm^{-2})	Linear range (mM)	LOD (μM)	Reference
Fe_3O_4 NRAs	<8	+0.6	406.9	0.0005–0.766	0.1	23
CoO_xNPs/ERGO	<5	+0.60	79.3	0.01–0.55 mM	2	28
RGO-$Ni(OH)_2$	<7	+0.54	11.43	0.002–3.1	0.6	41
CuO nanorods	<10	+0.60	371.43	0.004–8 mM	4	42
CoOOH nanosheets	<4	+0.40	341	0.03–0.7 mM	30.9	43
FeOOH nanowires	–	+0.4	12.13	0.015–33 mM	15	44
CuONPs-CSs	<5	+0.55	2981	0.0005–2.3	0.1	45
CuO_x-CoO_x/graphene	<3	+0.50	507	0.005–0.570	0.5	This work

electrode (11.43 μA mM^{-1} cm^{-2}) [41], CuO nanorods modified electrode (371.43 μA mM^{-1} cm^{-2}) [42], CoOOH nanosheets modified electrode (341 μA mM^{-1} cm^{-2}) [43], and FeOOH nanowires modified electrode (12.13 μA mM^{-1} cm^{-2}) [44], but lowers that the CuONPs-CSs modified electrode with a sensitivity of 2981 μA mM^{-1} cm^{-2} [45]. The detection limit at a signal to noise ratio of 3 is estimated to be 0.5 μM. The performance of the prepared CuO_x-CoO_x/rGO catalyst for glucose sensing is compared with some of existing enzyme-free glucose sensors based on various nanomaterials. As shown in Table 1, it can be concluded that the prepared CuO_x-CoO_x/rGO catalyst is among the top list of sensors with a faster response speed, a higher sensitivity, and a lower detection limit.

Other species often present along with glucose in biological samples, such as H_2O_2, NaCl, dopamine (DA), ascorbic acid (AA), uric acid (UA) and fructose, may interfere. In the physiological sample, glucose concentration (4.4–6.6 mM) is generally much higher than those of interfering species [7–9]. Therefore, the influence of 0.1 mM interference species on the current response of 1 mM glucose was evaluated. From the current response in Fig. 6, it can be observed that a remarkable glucose signal was obtained comparing to the other six interfering species. Compared to glucose, all the interfering species yield current response less than 9.3 %. The results indicate that an acceptable selectivity has been obtained for the present sensor.

Repeatability, reproducibility and stability of the sensor

The inter-electrode reproducibility investigation was conducted by comparing the response currents of six independent CuO_x-CoO_x/rGO modified electrodes prepared under the same conditions. The relative standard deviation (RSD) of response for amperometric determination of 100 μM glucose at 0.50 V was 5.3 %. In addition, four measurements of 100 μM glucose using the same electrode yielded a RSD of 4.6 %. These results indicated excellent intra-electrode and inter-electrode reproducibility. The long-term stability of electrode was investigated by analyzing its amperometric response after one-month storage. The results showed only 7.4 % decrease in the current response to 100 μM glucose. The good stability and repeatability make the CuO_x-CoO_x/rGO modified electrode feasible for practical applications.

Determination of glucose in urine

The CuO-CoO/rGO composites modified electrode was applied for the analysis of glucose spiked in the urine. The urine spiked with 10 mM and 20 mM standard solutions of glucose were signed as sample 1 and 2, respectively. The amperometric detection was carried out at the applied potential of 0.5 V in 10 mL 0.1 mol L^{-1} NaOH solution under stirring condition with the injection of 10 μL urine. The quantitative determination of samples was performed using the standard addition method and the results are shown in Table 2. The recoveries indicate that the sensor can be used for the determination of glucose in urine.

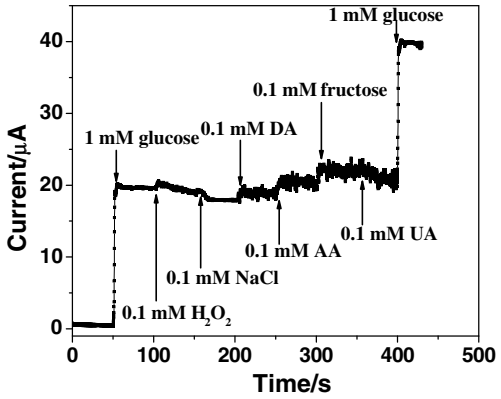


Fig. 6 Amperometric responses of the CuO_x-CoO_x/rGO/GCE to successive additions of 1 mM glucose, 0.1 mM H_2O_2, 0.1 mM NaCl, 0.1 mM DA, 0.1 M AA, 0.1 M fructose, 0.1 mM UA and 1 mM glucose in 0.1 M NaOH at applied potential of 0.5 V

Conclusions

In summary, an efficient enzyme-free glucose sensor based on a binary catalyst CuO_x-CoO_x/rGO was prepared by a direct two-step electrodeposition on electrode surface. The present sensor for glucose possessed good analytical performance, such as low detection limit, high detection sensitivity and reproducibility, acceptable selectivity and low cost. But there are some drawbacks when applied to sensing glucose in real samples. The first is the limited linear range of the present sensor (5 ~ 570 μM), which is far from the normal blood glucose concentration. To match the analytical ranges of sensor to real samples, simulated conditions using buffers (0.1 M NaOH) have to be used in the laboratory. The second is the CuO_x-CoO_x/rGO based sensor unable to be used in solutions with pH equal to or less than 7 because the electrocatalysis is highly dependent on the presence of OH^- anions. Therefore, there is still a long way for the low-cost nonnoble metals based enzyme-free glucose sensor to be used for practical applications. However, we envision that this work will open a new way to fabricate high performance binary nanocomposites as electrode materials and promote their multifunctional applications in biosensor, catalysis and bioengineer related areas.

Table 2 Amperometric determination of glucose spiked in human urine samples (n = 4)

Urine samples	Spiked (μM)	Found (μM)	R.S.D. (%)	Recovery (%)
Sample 1	10.0	9.68	2.64	96.8
Sample 2	20.0	19.8	2.08	99.0

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Compliance with Ethical Standards The author(s) declare that they have no competing interests

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