L-Cysteine/Graphene oxide for electrochemical determination of ascorbic acid in the presence of dopamine and uric acid

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Abstract. Ascorbic acid (AA), also known as vitamin C, has many roles in biological systems, such as prevention of cancer and common cold. Determination of AA is essential for food and pharmaceutical industries. Nowadays, electrochemical method is a potential method for detecting AA. However, detection of AA in presence of biological compounds, e.g. dopamine (DA) and uric acid (UA), is big challenge because these compounds are commonly found in biological samples and interfere the oxidation peak potential of AA. To overcome this problem, electrochemical coating of cysteine on graphene oxide decorated screen-printed electrode (Cys/GO/SP) was newly used for determination of AA. Field emission scanning electron microscope, energy dispersive x-ray and EIS were used to characterize the composites. AA of different concentrations were determined in phosphate buffer saline that has 10-µM DA and 10-µM UA using by differential pulse voltammetry (DPV). Cys/GO/SP presented three oxidation peaks corresponding to AA, DA and UA, whereas GO/SP and SP provided single peak. The oxidation peak currents of AA showed good linear relationship with AA concentrations of 0.5 - 5 mM with the detection limit of 0.26 mM. The results suggest that Cys/GO/SP can be used for electrochemical determination of AA in the interference of DA and UA.

1. Introduction

Vitamin C, additionally known as ascorbic acid (AA), Thevitamin is soluble water and observed in agricultural product and constituent of biological systems. The suitable amount of AA in body could aid to impede diseases for example cancer, tumors and Parkinson’s disease. Moreover, a lack of AA might cause many disorders such as Alzheimer’s disease, atherosclerosis and cancer [1]. Thus, the quantity of AA in foods and environment is interesting.

Recently, electrochemical approach is inexpensive, quick procedure. The detection of AA because the molecule is easily observed via oxidation reaction on electrode surface [2]. Nevertheless, oxidation peaks of dopamine (DA) and uric acid (UA) which intermingleas biological compounds in biological samples disturb electrochemical signal for each other at bare electrodes. To deal with this problem, Graphene oxide (GO) is promising modified electrode because hydrophilicity containing -
OH, -COOH and epoxides group within the basal plane and the sheet edge which is strong affinity with biological particles [3].

For our knowledge, we prepared a progressive sensor by electrochemical coating a Cys onto the GO modified screen-printed electrode (SP). The fabricated Cys/GO/SP was performed using Cyclic voltammetry (CV). The electrical properties of electrode surface were studied by electrochemical impedance spectroscopy (EIS). The investigation of morphology and chemical component field of modified electrodes were carried out by emission scanning electron microscopy (FeSEM) and energy dispersive x-ray (EDX). The quantitative detection of AA in mixture solution of DA and UA were performed by differential pulse voltammetry (DPV) techniques.

2. Experimental setup

2.1. Fabrication of Cys/GO/SP
Screen-printed electrode (SPE) was purchased from Quasense (CI1703OR). Graphene oxide was prepared based on Hummers and Offeman [4]. For preparing GO-modified working electrode (WE) (GO/SP), 2 µL of as-prepared GO suspension (3.13 mg/mL) was coated on WE and dried in air oven at 40 °C for 10 min. The GO/SP was performed as the WE and potential range of CV was set for -1.0 to 1.0 V at 50 mV·s⁻¹ for 10 cycles in 0.01 M Cys solution. The Cys/GO/SP was dried in hot air oven and kept at room temperature.

3. Results and discussion

3.1. Preparation and characterization of Cys/GO/SP
CV is a useful procedure to coat cysteine onto the WE surface. Figure 1(a) showed CV curves for each cycle when electrochemical coating of Cys was occurred at the GO/SP electrode. As exemplified, the increment of oxidation current was linearly proportional with an increment of CV cycles. This mechanism was similar to electrochemical formation of conductive polymers by CV [5]. This result suggested that Cys had been successfully coated onto the GO/SP surface.

![Figure 1](image_url)

Figure 1. (a) CV for electrochemical coating of Cys in 0.01 M PBS (pH 7.4) onto GO/SP after 10 cycles. Scan rate: 50 mV·s⁻¹. (b) The Nyquist plot of SP, GO/SP and Cys/GO/SP 0.01 M K₃[Fe(CN)]₆ in 0.1 M KCl solution.

EIS was investigated for the formation of Cys onto the GO/SP by CV and coating GO on SP surface. Figure 1(b) depicted the impedance spectra of SP, SP-GO and SP-Cys/GO in Fe(CN)₆³⁻/⁴⁻. The diameter of the semicircle in figure 1(b) indicated to charge transfer resistance (Rct) of each electrode) [6]. The Rct value of SP (5125 Ω) was smaller than those obtained at GO/SP (8520 Ω) and Cys/GO/SP (102350 Ω). These results revealed that SP surface was modified both of GO and Cys.

The surface morphology of SP, GO/SP and Cys/GO/SP were depicted by FeSEM images. Figure 2(a) displayed SP surface which is an amorphous carbon and wrinkled and crumpled surface of GO/SP indicated to graphene oxide morphology is shown in figure 2(b). After electrochemical
coating Cys on the GO/SP (figure 2(c)), the surface morphology was anchored by Cys on the wrinkled GO surface. EDX of Cys/GO/SP showed element of Nitrogen and Sulphur atom which is component of Cys as shown figure 2(d).

**Figure 2.** FeSEM image of the working electrode surface: (a) SP, (b) GO/SP and (c) Cys/GO/SP. The acceleration voltage was 10 kV and resolution of all FeSEM image was 1 µm. EDX (d) showed element components of Cys/GO/SP.

### 3.2. Study of electrochemical oxidation of AA in presence of DA and UA

Figure 3(a) compared the electrochemical oxidation for mixture solution of 2 mM AA, 10 µM DA and 10 µM UA at SP, GO/SP and Cys/GO/SP. Both bare SP and GO/SP showed a broad oxidation peak. On the contrary, Cys/GO/SP, the oxidation peak potential of AA is 0.22 V, which is separated for compared to DA (0.36 V) and UA (0.53 V). These oxidation peak was reported for AA, DA and UA by previous work [6]. Moreover, oxidation peak current of Cys/GO/SP is larger than that of SP and GO/SP which effect from functional group of Cys. Cys/GO/SP was used to gain the electrochemical differentiation of AA in presence of DA and UA because effect of Cys’s charge provide accumulative and exclusion properties. In this work, optimized pH for study electrocatalytic oxidation of AA in presence of DA and UA was 7.4 because this pH value is variety of physiological pH value which can be most relevant to electrochemical oxidation of AA study [7].

### 3.3. Determination of AA in presence of DA and UA

At suitable condition, DPV was used to analyze AA in presence of DA and UA. The concentration of AA was varied, while both DA and UA concentrations were kept for 10 µM, respectively. Figure 3(b) showed clearly separation oxidation peak of AA, DA and UA, that are located at 0.02, 0.23 and 0.37 V, respectively. The oxidation peak of AA showed good linear relationship with its concentrations from 0.5 to 5 mM. The DPV peak currents ($I_{ox}$) of AA linearly increased with increasing concentration. The linear equation is $I_{ox} = 3.189C_{AA} + 9.076 (r^2 = 0.979)$ as shown in figure 3(c). The limit of detection (LOD) was calculated to be 0.26 mM.
Figure 3. CV for 2 mM AA, 10 µM DA and 10 µM UA recorded at SP, GO/SP and Cys/GO/SP. Supporting electrolyte: 0.01 M PBS (pH 7.4) at scan rate 50 mV·s\(^{-1}\). (b) DPV of 0.5, 1, 1.5, 2.5, 3, 4, 5 mM of AA in presence of 10 µM DA and 10 µM DA on Cys/GO/SP in 0.01 M PBS (pH 7.4). (c) The plot of peak current of AA against concentration of AA.

4. Conclusions
The GO/SP interface covalently anchored with Cys via electrochemical coating. Although charge transfer resistance of Cys/GO/SP was more than that of SP and GO/SP because of containing abundant functional group of Cys, these functional group provided clearly separation of oxidation peak of AA in presence of DA and UA. By CV and DPV techniques, oxidation peak currents of AA, DA and UA were clearly separated suggesting that Cys/GO/SP is promising electrode for electrochemical simultaneous determination of AA, DA and UA.

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