Electrochemical Fabrication of Polypyrrole/Poly(pyrrole-3-carboxylic acid)/Graphene Oxide Composite Thin Film for Biosensor Application

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ABSTRACT

An electrochemical fabrication of functionalized conducting polymer/graphene oxide (GO) composite thin film for the detection of biomolecule were reported. Polypyrrole/ poly(pyrrole-3-carboxylic acid)/graphene oxide (PPy/PP3C/GO) composite thin films were synthesized by electropolymerization on an indium tin oxide (ITO) coated glass slide. The effect of concentration and composition of precursor solution on the electrochemical behaviour of the composited material were investigated. The PPy/PP3C/GO composite thin film synthesized under the optimized experimental conditions was characterized by cyclic voltammetry (CV). In addition, the electrochemical behavior and doping/dedoping properties of the obtained composite thin film in a neutral phosphate-buffered saline (PBS) solution were also studied. Finally, the PPy/PP3C/GO composite was used to detect ascorbic acid (AA). It was found that PPy/PP3C/GO showed good electroactivity property in neutral PBS solution and could be applied for the detection of AA. Therefore, it can be concluded that an electrochemically fabricated PPy/PP3C/GO composite material is a promising candidate as smart material for biosensing applications.

Keywords: *graphene oxide, polypyrrole, pyrrole-3-carboxylic acid, biosensor*

INTRODUCTION

Conducting polymers (CPs) such as polythiophene, polyaniline, poly(phenylene vinylene) and polypyrrole (PPy) and their derivatives have received considerable attention because of their unique electrical, optical, chemical and biochemical properties (Vidal *et al.*, 2003; Guimard *et al.*, 2007; Peng *et al.*, 2009; Singh *et al.*, 2009; Yang *et al.*, 2015). In recent years, CPs have found potential applications in various fields such as fuel cells, electrochromic displays, field effect transistors (FETs) drug release system, affinity chromatography, chemical sensor and biosensors (Adhikari and Majumdar, 2004; Demos *et al.*, 2006; Baba *et al.*, 2010; Jiang *et al.*, 2012; Yue *et al.*, 2015). Nowadays, CPs and their derivatives-based biological sensors have been extensively investigated to detect biomolecules for example glucose, urea, enzymes, hormones, neurotransmitters, antibodies and antigens (Sriwichai *et al.*, 2010; Janmanee *et al.*, 2011; Lin *et al.*, 2013; Chu *et al.*, 2015; Soares *et al.*, 2015). Among these CPs, polypyrrole (PPy) and their derivatives are especially promising as chemical and biochemical sensors due to their good electrical conductivity, environmental stability, good biocompatibility, facile

synthesis and versatility compared to the other CPs (Jiang *et al.*, 2012; Lin *et al.*, 2012; Palod *et al.*, 2012). PPy films usually are prepared by electrochemical polymerization techniques because the synthetic procedure is relatively straightforward with the ability to control thickness, shape and morphology (Guimard *et al.*, 2007; Ravichandran *et al.*, 2010; German *et al.*, 2012).

It is well known that nanomaterials have been extensively utilized in electrochemical biosensor due to their conductivity, chemical stability, flexibility and low cost. Some nanomaterials, including gold nanoparticles, carbon nanotubes (CNTs) and graphene (GR) have been successfully integrated into CPs in the field of biosensor (Bai and Shiu, 2014; Hwa and Subramani, 2014; Xue *et al.*, 2014; Devasenathipathy *et al.*, 2015; Nia *et al.*, 2015; Peng *et al.*, 2015). GR is a promising nanomaterial currently being employed for the fabrication of electrochemical biosensors due to its unique properties including fast electron transportation, excellent thermal conductivity, rich surface chemistry and good biocompatibility (Hwa and Subramani, 2014; Xue *et al.*, 2014). Considering the advantages of the CPs and nanomaterials, CPs composite nanomaterials designed for biosensors could benefit from the good conductivity, enhanced binding ability by covalent immobilization of CPs and the superior stability and conductivity of nanomaterial.

Therefore, this study aims to fabricate functionalized polypyrrle/graphene oxide composites for the detection of biomolecule in electrochemical biosensor application. The fabrication and characterization of polypyrrole/poly(pyrrole-3-carboxylic acid)/graphene oxide (PPy/PP3C/GO) composites thin film are reported. Their electrochemical behavior and doping/dedoping properties were monitored by cyclic voltammetry (CV) in a neutral phosphate-buffered saline (PBS) solution. Finally, the PPy/PP3C/GO composite was used to fabricate a sensitive amperometric sensor for the determination of ascorbic acid (AA), which is an important biomolecule in the biological fluids is related to several diseases such as cancer, diabetes and hepatic disorder. It was found that the materials designed in this work are promising candidates as smart materials for the biosensor applications.

MATERIALS AND METHODS

Chemicals and Materials

The pyrrole (Py) and pyrrole-3-carboxylic acid (P3C) were purchased from Sigma-Aldrich Co. Graphene oxide (GO) was prepared by a modified Hummers method (Hummers and Offeman, 2014). All other chemicals were analytical grade with highest purity and used as received.

Electrochemical experiments were performed with a Digi-Ivy model DY2000 potentiostat with conventional three-electrode cell. An indium tin oxide (ITO) coated glass slide was used as the working electrode. Ag/AgCl electrode and a platinum wire were used as the reference and counter electrodes, respectively.

Synthesis of PPy/PP3C/GO composite thin film

PPy/PP3C/GO composite material was deposited on ITO substrate via electrochemical method. The precursor solutions containing 0.2 g of GO in a monomer solution of α mM Py and β mM P3C with constant 10 mM monomer concentration in 0.5 M H_2SO_4 solution, the simple notation of α:β Py/P3C, such as 9:1 Py/P3C 5:5 Py/P3C, 1:9 Py/P3C etc., were used for fabricating the PPy/PP3C/GO thin films. In this study, 0.5 M sulfuric acid acted not only as electrolyte solution but also solvent in the fabrication process.

For the electrochemical system, platinum wire, Ag/AgCl electrode and ITO were used as counter, reference and working electrodes, respectively.

Characterization

The fabrication of PPy/PP3C/GO composite thin film was monitored by CV. The effect of experimental conditions such as concentration and composition of precursor solution on the electrochemical behaviour of the composited material were also investigated. In addition, the electrochemical behavior and doping/dedoping properties of the obtained composite thin film under the optimized experimental conditions in a neutral phosphate-buffered saline (PBS) solution (pH 7.4) containing 10.0 mM $K_3[Fe(CN)_6]/K_4[Fe(CN)_6]$ (1:1 mixture as redox probe) ($[Fe(CN)_6]^{3-/4-}$) were studied.

Electrochemical measurement

All electrochemical measurements were carried out in a three-electrode electrochemical cell at room temperature of 25 ^oC. CVs were performed by cyclic voltammetry in a potential range from -0.1 to 0.7 V at a scan rate of 50 mV/s for the detection of various concentrations of AA (10, 20, 30, 40 and 50 mM).

RESULTS AND DISCUSSION

Fabrication of PPy/PP3C/GO composite thin film

The electrochemical property during the electrochemical fabrication of PPy/PP3C/GO composite thin film was monitored using an electrochemical measurement. Figure 1 shows the cyclic voltammograms of 5:5 PPy/PP3C copolymer composited with GO. The current in the anodic scan increased about 0.6 V, indicating the beginning of the formation of polymer film on ITO electrodes. The current slightly decreased about 0.3 V in the cathodic scan, which indicated the dedoping process of the deposited copolymer films (Janmanee *et al.*, 2011). The electroactivity property of the PPy/PP3C/GO composite material was compared with PPy. It was found that the oxidation potential exhibited by PPy/PP3C/GO thin film was lower than that of PPy. However, the effect of experimental conditions such as concentration and composition of precursor solutions on the electrochemical behaviour of the composited material were also investigated.

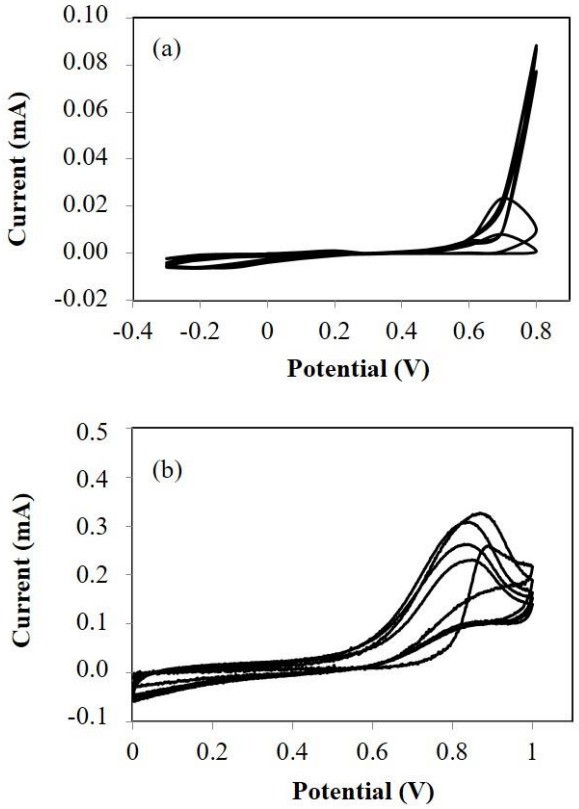

Figure 1 Cyclic voltammograms of the electrochemical fabrication of (a) PPy/PP3C/GO and (b) PPy.

The effect of concentration and composition of precursor solutions

Cyclic voltammograms of the PPy/PP3C/GO thin film fabricated on an ITO glass substrate with various ratio of monomer concentration are shown in Figure 2. As shown in the cyclic voltammogram of 1:9, 5:5 and 9:1 of PPy/PP3C/GO composite thin films, the material prepared with 5:5 PPy/PP3C/GO exhibited higher current than that of the other copolymers due to the large amount of deposited film on the electrode. These results indicate that the 5:5 PPy/PP3C/GO has lower oxidation potential which facilitates the fabrication of functionalized polypyrrole composited with GO compared to other combinations (Peng *et al.*, 2005). Based on these results, the 5:5 PPy/PP3C/GO was employed to represent the properties of PPy/PP3C/GO composite material.

The effect of GO on the electrochemical behaviour the PPy/PP3C/GO composite material was studied by electrochemical measurement. Cyclic voltammograms of 5:5 PPy/PP3C/GO and 5:5 PPy/PP3C were recorded in 0.2 M KCl containing 10.0 mM of $[Fe(CN)_6]^{3-/4-}$ at a scan rate of 20 mV/sec are shown in Figure 3. It can be clearly seen that the cyclic voltammogram of PPy/PP3C thin film

shows a well-defined reversible redox behaviour attributed to highly electron transfer from $[Fe(CN)_6]^{3-/4-}$ to the electrode. After the deposition of PPy/PP3C/GO on the surface of the electrode, the redox current of PPy/PP3C/GO thin film was higher than that of PPy/PP3C thin film due to excellent electrocatalytic property of the composite material was higher than that of the PPy/PP3C copolymer. This would improve the efficiency of the electrode (Ruiyi *et al.*, 2013).

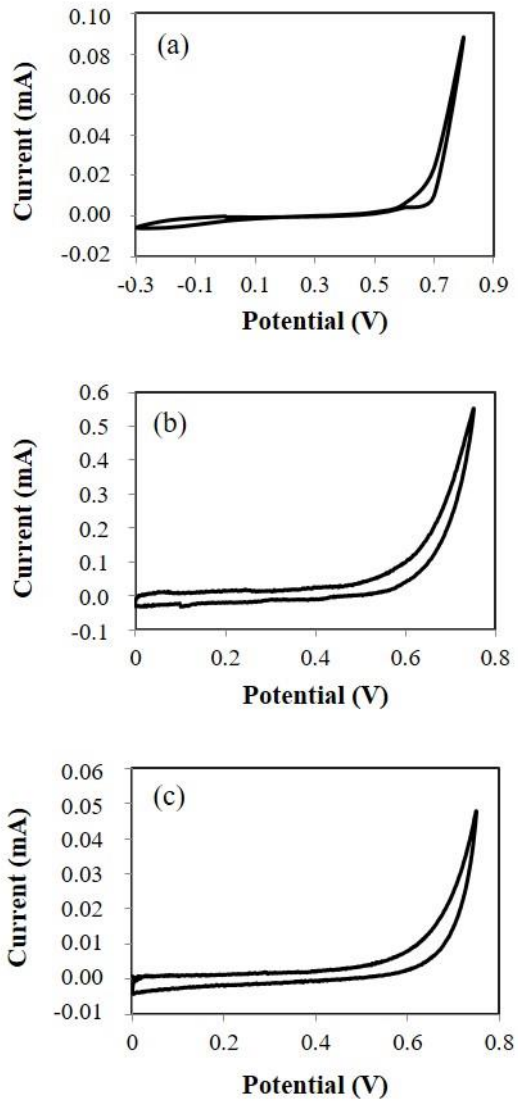

Figure 2 Cyclic voltammograms of the electrochemical fabrication of PPy/PP3C/GO with various ratio of monomer concentration of $\alpha:\beta$ Py/P3C at (a) 1:9 (b) 5:5 and (c) 1:9.

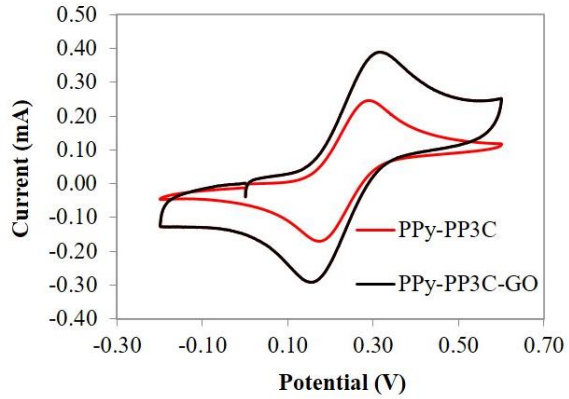

Figure 3. Cyclic voltammograms of 5:5 PPy/PP3C/GO and 5:5 PPy/PP3C in 0.2 M KCl containing 10.0 mM of $[Fe(CN)_6]^{3-/4-}$ at a scan rate of 20 mV/sec.

Electrochemical property of PPy/PP3C/GO composite thin film

The effect of the scan rate on the electrochemical performance was studied in this work. Figure 4 shows the cyclic voltammograms of the 5:5 PPy/PP3C/GO thin film deposited on an ITO glass substrate in PBS solution pH 7.4 containing 10.0 mM $[Fe(CN)_6]^{3-/4-}$ at different scan rates of 5, 10, 20, 50 and 100 mV/sec. The anodic and cathodic current of the composite thin film increased with the increasing scan rate, which indicated that the PPy/PP3C/GO can be electroactive in neutral PBS solution. It can be concluded that the functionalized polypyrrole composited graphene oxide showed good electroactivity in neutral solution which may possess potential applications in various systems of the biosensors.

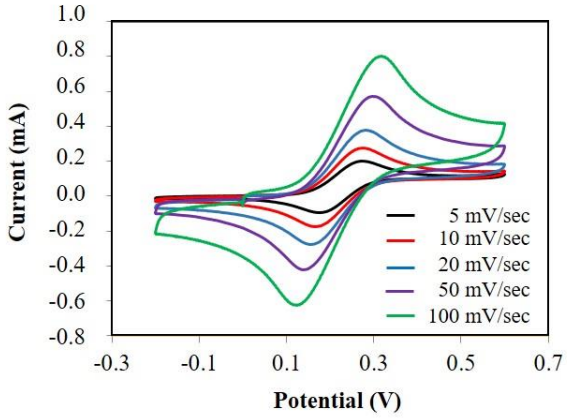

Figure 4 Cyclic voltammograms of 5:5 PPy/PP3C/GO composite thin film in PBS solution pH 7.4 containing 10.0 mM $[Fe(CN)_6]^{3-/4-}$ at different scan rate of 5, 10, 20, 50 and 100 mV/sec.

Performance of PPy/PP3C/GO composite thin film for the detection of AA

Figure 5 shows the cyclic voltammograms of the PPy/PP3C/GO thin film at different concentrations of AA in PBS solution at a scan rate of 50 mV/sec. It can be seen that the anodic and cathodic current of the composite thin film increased as the concentration of AA increased, indicating that the redox reaction of AA at the surface of PPy/PP3C/GO electrode increased with the increasing concentration of AA. In addition, the current of the oxidation peak is apparently higher than that of the reduction peak. This may attribute to the redox reaction of AA at PPy/PP3C/GO thin film is irreversible (Yue *et al.*, 2015).

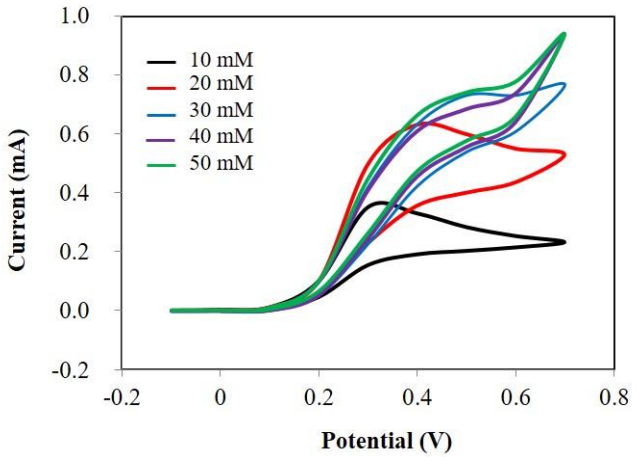

Figure 5 Cyclic voltammograms of PPy/PP3C/GO composite thin film for the determination at various concentrations of AA at the scan rate of 50 mV/sec.

CONCLUSIONS

The fabrication of functionalized polypyrrole/graphene oxide composite thin films was successfully prepared by electrochemical method. The PPy/PP3C/GO composite thin film remarkably improve electron transfer of the redox in aqueous solution to the electrode. Moreover, the CV measurement of PPy/PP3C/GO showed good electroactivity in neutral PBS solution, which would improve the sensitivity of the electrode for the detection of biomolecule in biosensing applications. The PPy/PP3C/GO composite was used to detect AA as a sample of biological molecule. It was found that PPy/PP3C/GO could be applied for the detection of AA. Therefore, it can be concluded that an electrochemically fabricated functionalized pyrrole/graphene oxide composite thin film is a promising candidate as smart materials for biosensors application in the future.

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