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Original Article

Fabrication of a novel electrochemical sensor for determination of hydrogen peroxide in different fruit juice samples



Navid Nasirizadeh ^{a,*}, Zahra Shekari ^a, Ali Nazari ^c,
Masoumeh Tabatabaee ^b

^a Scientific Society of Nanotechnology, Yazd Branch, Islamic Azad University, Yazd, Iran

^b Department of Chemistry, Yazd Branch, Islamic Azad University, Yazd, Iran

^c Department of Art and Architectural, Yazd Branch, Islamic Azad University, Yazd, Iran

ARTICLE INFO

Article history:

Received 19 February 2015

Received in revised form

30 May 2015

Accepted 15 June 2015

Available online 26 July 2015

Keywords:

amperometry

electroreduction

hydrogen peroxide

reactive blue 19

ABSTRACT

A new hydrogen peroxide (H_2O_2) sensor is fabricated based on a multiwalled carbon nanotube-modified glassy carbon electrode (MWCNT-GCE) and reactive blue 19 (RB). The charge transfer coefficient, α , and the charge transfer rate constant, k_s , of RB adsorbed on MWCNT-GCE were calculated and found to be 0.44 ± 0.01 Hz and 1.9 ± 0.05 Hz, respectively. The catalysis of the electroreduction of H_2O_2 by RB-MWCNT-GCE is described. The RB-MWCNT-GCE shows a dramatic increase in the peak current and a decrease in the over-voltage of H_2O_2 electroreduction in comparison with that seen at an RB modified GCE, MWCNT modified GCE, and activated GCE. The kinetic parameters such as α and the heterogeneous rate constant, k' , for the reduction of H_2O_2 at RB-MWCNT-GCE surface were determined using cyclic voltammetry. The detection limit of $0.27 \mu M$ and three linear calibration ranges were obtained for H_2O_2 determination at the RB-MWCNT-GCE surface using an amperometry method. In addition, using the newly developed sensor, H_2O_2 was determined in real samples with satisfactory results.

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1. Introduction

Hydrogen peroxide (H_2O_2) is an essential mediator used in many fields of practice such as food, pharmaceutical, clinical, diagnostic, environmental protection, and in industries [1]. Indeed, H_2O_2 has wide applications in industrial

processes as a universal oxidant and is a very important intermediate agent in environmental and biological reactions. It has also emerged as an important by-product of enzymatic reactions in biosensing processes, because it is released during the oxidation of substrates in the presence of oxygen [2,3]. Monitoring H_2O_2 with a reliable, rapid, and economical method is of great significance for numerous

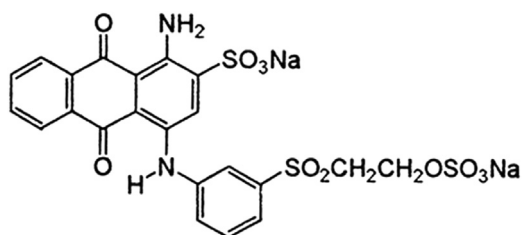
* Corresponding author.

E-mail address: nasirizadeh@iauyazd.ac.ir (N. Nasirizadeh).

<http://dx.doi.org/10.1016/j.jfda.2015.06.006>

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processes. The determination of H_2O_2 has been an important project over the past century. This is still something to accomplish, especially with the recent realizations about the existence of H_2O_2 in environmental samples. Various techniques including fluorimetry [4], titrimetry [5], chemiluminescence [6], spectrophotometry [7], photometry [8], and electrochemistry [9–11] have been developed for this purpose. Among these techniques, electrochemistry based on simple and low-cost electrodes has been extensively used to determine H_2O_2 [12]. It has been a preferred technique due to its quick response, low cost, simple instrumentation, high sensitivity, and possibility of miniaturization [13]. A number of enzyme electrodes reported for H_2O_2 determination are used due to their simplicity, high sensitivity, and selectivity [14]. Although, many enzymatic H_2O_2 assays possess good sensitivity and selectivity, native enzymes gradually lose their catalytic activity after repeated measurements and they are comparatively expensive. Therefore, the development of enzyme-free H_2O_2 sensors with a low detection limit and a wide responding range is preferred. However, the direct electrochemistry of H_2O_2 requires a higher over-potential and slow electrode kinetics on many electrode materials. Electrodes modified with different electroactive materials have the ability to detect H_2O_2 at low potentials. In recent years, various kinds of electrochemical sensors based on detection of different compounds in the food industry have been developed [15–18]. The study of electrochemical biosensors has been in progress to improve the speed, selectivity, sensitivity, and cost of producing chemical compounds. Immobilization of mediators in charge of electron transfer to electrode surfaces is a key step for the design, fabrication, and performance of sensors and biosensors [19]. Quinones are excellent electron mediators that efficiently reduce the working potentials and eliminate the effects of various electroactive substances in real samples; they have been successfully used in numerous biosensors to detect H_2O_2 [20,21]. Reactive blue 19 (RB) is a derivative of quinone (see its structure in Scheme 1). Owing to the good reactivity of quinone derivative mediators, it seems that the use of RB as a modifier could be important to yield some new information about the catalyzation of slow reactions. In this work, for the first time, RB is introduced as a catalytic compound for electroreduction of H_2O_2 , then its electrochemical behavior and kinetic parameters are investigated. Cyclic voltammetry and amperometry were used to investigate the electrochemical properties and electrocatalytic activity of the modified electrode for determination of H_2O_2 .



Scheme 1 – Structure of reactive blue 19.

2. Methods

2.1. Reagents and apparatus

H_2O_2 , dimethyl formamide, and the other chemicals used to produce the buffer solution were obtained from Merck Company (Darmstadt, Germany) and used as received. RB was obtained from Sigma–Aldrich (St Louis, MO, USA). Multi-walled carbon nanotubes (MWCNTs; with a diameter of 10–20 nm, length of 5–20 μm , and purity of 95%) were purchased from NanoLab Inc. (Waltham, MA, USA). All chemical reagents were of analytical grade. Phosphate buffer solutions (0.1M) were prepared with H_3PO_4 , the pH was adjusted with 2.0M NaOH. All solutions were prepared with doubly distilled water.

Electrochemical experiments were carried out with an Autolab modular electrochemical system (ECO Chemie, Utrecht, The Netherlands) equipped with a PGSTA 30 module and driven by GPES 4.9 software, in conjunction with a three-electrode cell. The cell used was equipped with an RB-MWCNT-modified glassy carbon electrode (GCE) as a working electrode, a platinum electrode (Azar Electrode Co, Tabriz, Iran) as an auxiliary electrode, and a saturated calomel electrode as a reference electrode. All potentials in the text are quoted versus this reference electrode. The pH was measured with a pH/mV meter model 827 (Metrohm, Riverview, FL, USA).

2.2. Preparation of modified electrodes

The procedure of fabricating various modified electrodes was as follows. Prior to modification, a bare GCE was polished successively with 0.05 μm Al_2O_3 slurry on a polishing cloth and then rinsed with doubly distilled water. To be electrochemically activated, the cleaned electrode was immersed in a 0.1M sodium bicarbonate solution and activated by a continuous potential cycling from -1.45 to 1.7 V at a sweep rate of 100 mV/s. To prepare a RB modified GCE (RB-GCE) the activated GCE (AGCE) was rinsed and placed in a 0.10mM solution of RB in a 0.1M phosphate buffer (pH 7.0). It was modified by eight cycles of potential sweep between -0.4 and 0.5 V at 20 mV/s. To fabricate MWCNT-GCE, 3 μL of dimethyl formamide-MWCNT solution (1 mg/mL) was placed directly onto the AGCE surface and dried at room temperature for 30 minutes to form a MWCNT film at the GCE surface. An RB-MWCNT-GCE was prepared by immersing the MWCNT-GCE in a 0.10mM solution of RB in a 0.1M phosphate buffer (pH 7.0; Scheme 2). It was modified using the same procedure as for RB-GCE.

3. Results and Discussion

3.1. Electrochemical behavior of RB-MWCNT-GCE

In recent years, the mechanism of electrodeposition of *o*- or *p*-hydroquinone derivatives, as modifiers, on the surface of an activated GCE has been a matter for discussion [22–25]. Fig. 1 shows the cyclic voltammograms of the

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