Materials Science & Engineering C 91 (2018) 696–704

Contents lists available at ScienceDirect

Materials Science & Engineering C

journal homepage: www.elsevier.com/locate/msec

Electrochemical preparation of surface molecularly imprinted poly(3-aminophenylboronic acid)/MWCNTs nanocomposite for sensitive sensing of epinephrine

Juan Zhang[a,b], Xiao-Tong Guo[a], Jun-Ping Zhou[a], Guang-Zhou Liu[c,*], Shu-Yong Zhang[a,*]

[a] School of Chemistry and Chemical Engineering, Shandong University, Jinan 250100, China
[b] Department of Chemistry, School of Basic Medical Sciences, Ningxia Medical University, Yinchuan 750004, China
[c] School of Marine Science, Shandong University, Jinan 250100, China

ARTICLE INFO

Keywords:
3-aminophenylboronic acid
Molecularly imprinted polymers
Electropolymerization
Electrochemical sensor
Epinephrine

ABSTRACT

A nanocomposite with multi-walled carbon nanotubes (MWCNTs) coated with surface molecularly imprinted polymers (MIPs) poly(3-aminophenylboronic acid) (PAPBA) was successfully prepared via potentiodynamic electropolymerization and tested as an effective electrochemical material for epinephrine (EP) detection. The morphology and properties of the sensing material were characterized with scanning electron microscopy and electrochemical impedance spectroscopy. Compared with MWCNTs or non-imprinted polymers PAPBA modified MWCNTs electrodes, the PAPBA(MIPs)/MWCNTs modified electrode showed a lower charge transfer resistance and enhanced electrochemical performance for EP detection. The improved performance can be attributed to the large amount of specific imprinted cavities with boric acid group which can selectively adsorb EP molecule and the synergistic effect between MWCNTs and PAPBA(MIPs). The effects of pH, the molar ratio between monomer and template molecule, the cycle number of electropolymerization, and the accumulation time of the modified electrode on the sensing performance were investigated. It was found that under the optimal conditions, the PAPBA(MIPs)/MWCNTs sensor could effectively recognize EP from many possible interferents of higher concentration within a wide linear range of 0.2–800 $\mu mol \cdot L^{-1}$, with low detection limit of 35 $nmol \cdot L^{-1}$, high sensitivity and good discrimination. The detection of EP in human serum and real injection samples using the PAPBA(MIPs)/MWCNTs sensor also gave satisfactory results.

1. Introduction

Epinephrine (EP) is an important catecholamine neurotransmitter produced in chromaffin cells of the adrenal medulla and in certain neurons of the central nervous system [1]. Because of its special functions, EP is commonly used as the important constituent of some medicines for the treatment of various cases like cardiac arrest, organic heart disease, bronchial asthma, anaphylaxis and superficial bleeding, and is occasionally misused as stimulant drug belonging to the prohibited list of World Anti-Doping Agency. Therefore, detection of EP concentration is important in clinical diagnosis, physiology and pharmaceutical research and doping detection. Various techniques including spectrophotometry [2], chromatography [3], fluorescence [4], capillary electrophoresis [5] and electrochemical [6] methods have been implemented for quantitative detection of EP. Among these techniques, electrochemical sensors have attracted special attention due to the simplicity, low cost, easy miniaturization and automation [7, 8]. However, the electrochemical detection of EP is still facing great challenging due to the complicated sensing environments such as low concentration of analyte and higher concentrations of interferences such as catecholamine metabolites, other neurotransmitters, uric acid (UA) and ascorbic acid (AA), etc. To develop new electrochemical sensors with enhanced selectivity and sensitivity for detecting of EP is of both theoretical and application significance.

Molecularly imprinted polymers (MIPs) are tailor-made biomimetic materials capable for recognizing target molecules with special selectivity. The procedure for synthesis of MIPs generally involves three steps: (1) the formation of a complex between functional monomers and template molecules which are the analyte itself or its analogues with similar structure; (2) polymerization of the complex; and (3) removal of the embedded template molecules, regenerating the memory cavities in the polymer matrix which endows the polymer high specificity [9, 10]. The functional monomer–template complex can be formed via reversible covalent or non-covalent interactions [11]. The covalent

* Corresponding author.
E-mail address: syzhang@sdu.edu.cn (S.-Y. Zhang).

https://doi.org/10.1016/j.msec.2018.06.011
Received 3 September 2017; Received in revised form 29 May 2018; Accepted 9 June 2018
Available online 10 June 2018

approach is superior to the non-covalent approach due to the generation of more homogeneous binding sites.

Recently, MIPs have been extensively investigated for sensing neurotransmitter [12, 13] such as dopamine [14, 15], histamine [16] and tryptamine [17]. For detection of EP with MIPs electrochemical sensors, only a few works has been reported. In 2013, Huynh et al. used prepolymerization complex with two bis(2,2′-bithienyl)methane derivatives serving as functional monomers and EP as template molecule for electropolymerization of a MIPs film on a Pt disk electrode and an Au film electrode [18]. In 2015, hemin modified EP MIPs electrochemical sensor was prepared via polymerization of 2,4,6-trisacrylamido-1,3,5-triazine functional monomer on a gold disc electrode [19]. In these studies, MIPs show many disadvantages such as poor site accessibility and low binding affinity for analyte due to the embedment of most imprinted sites. To address these issues, a surface molecular imprinting technique in which the MIPs composites was prepared on the surface of some nanomaterials was established. Owing to the better proximity between the imprinted sites and the materials' surfaces, faster association/dissociation kinetics, and better selectivity and sensitivity, the surface imprinting technique is more competitive for molecular recognition [20, 21]. MIPs composite sensors for EP detection based on zinc oxide nanorods [22], silica nanoparticles [23] and multi-walled carbon nanotubes (MWCNTs) covalently modified with *N*-hydroxyphenylmaleimide [24] have been reported. Nevertheless, there are still two main problems with these works: the procedure for template removal is tedious, and the imprinted sites were not homogeneous due to the non-covalent immobilization protocol.

Aminophenylboronic acid (APBA), a derivative of phenylboronic acid, can covalently bond with 1,2- and 1,3-diol compounds such as sugar and catecholamine to form stable cyclic boronate esters in an alkaline or neutral aqueous solution [25]. Once the pH is switched to acidic, the formed boronate esters can dissociate with ease [25]. Otherwise, as an electroactive functional monomer, APBA can be deposited on a transducer surface by oxidation electropolymerization [26] and the boronic acid groups functionally attached to polymer back bone can combine with cis-diol compounds [27]. With the special reversible binding property, APBA is possibly suitable for constructing a MIPs layer with homogeneous imprinted sites for detection of cis-diol compounds. Although APBA-based IMPs sensors for saccharide [28] and APBA-based IMPs composited with MWCNTs [29, 30] have been reported, the report for sensing catecholamine is quite rare [31]. Being a chiral compound [18], EP imprinted sites on APBA IMPs for recognition of EP molecule may be more specific. To the best of our knowledge, surface MIPs composite sensor for EP detection based on APBA has not been reported.

In this study, we intend to establish a new procedure for preparation of a MIPs electrochemical sensor featuring of facile electrodeposition, easy and complete removal of imprinting molecule, and uniform distribution of the imprinted sites. The MIPs electrochemical sensor was constructed using electropolymerization of 3-aminophenylboronic acid (3-APBA) as monomer on the surface of MWCNTs attached on a glassy carbon electrodes (GCE) covered with chitosan. The positively charged nature of the chitosan layer was taken advantage for homogeneous and stable assembly of the negatively charged MWCNTs using a layer-by-layer strategy through electrostatic interaction between MWCNTs and chitosan [32, 33]. Enhanced specific recognition ability, high sensitivity, wide linear range, low detection limit and good reproducibility and stability was achieved for this MIPs sensor for EP detection.

2. Experimental procedures

2.1. Chemicals and materials

3-Aminophenylboronic acid hydrochloride, (±)-Epinephrine hydrochloride and (*R*)-(−)-Phenylephrine hydrochloride were purchased from Aladdin (Shanghai, China). Ascorbic acid, uric acid, D-(+)-Glucose, D-(−)-Fructose, D-(+)-Mannose, and chitosan with the degree of deacetylation more than 95% and viscosity of 100–200 mPa·s were obtained from Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China). MWCNTs with outer diameter of 30–50 nm and length of 10–20 μm were purchased from Beijing Deke Daojin Co. Ltd. (Beijing, China). All reagents were of analytical grade and used without further purification. Ultrapure water (Millipore, 18.25 MΩ cm, Ulupure Science and Technology Co. Ltd., Chengdu, China) was used for solution preparation. Human serum samples were obtained from healthy volunteers. The epinephrine hydrochloride injections (the labeled concentration: 1.00 $mg \cdot mL^{-1}$; the auxiliary ingredients: sodium pyrosulfite, sodium chloride, edetate disodium, water for injection) were produced by Grand Pharma Co. Ltd. (Shanghai, China).

2.2. Apparatus

The electrochemical measurements were performed using a CHI660E Electrochemistry Workstation (CH Instrument Inc., Shanghai, China). A conventional three-electrode system was used with a glassy carbon electrode (GCE) (0.07 cm^2 geometric areas, Gaossunion Technology Ltd., China) or modified GCE serving as the working electrode, a platinum foil (1 cm^2 geometric areas, self-made) as the counter electrode and a commercial saturated calomel electrode (SCE) as the reference electrode. The reference electrode was placed with the Luggin capillary 2 mm away from the working electrode surface.

The surface morphology of the modified electrode was characterized using a JSM-6700F field emission scanning electron microscope (JEOL, Japan) at an acceleration voltage of 5.0 kV. The UV spectra were recorded on a U-4100 spectrophotometer (Hitachi, Japan). A quartz cell of 1.00 cm thick was used for the measurements.

2.3. Preparation of MWNTs-modified GCE

MWNTs were pretreated in a mixture of concentrated nitric acid and sulfuric acid to improve the surface wettability [23]. After vacuum filtration, the pretreated MWNTs were washed thoroughly with deionized water until the filtrate became neutral. Then, a 1.0 $mg \cdot mL^{-1}$ MWNTs suspension was prepared by dispersing the pretreated MWNTs in pure water with the aid of ultrasonic agitation.

Prior to electropolymerization, the surface of GCE was polished with 0.3 μm α-Al_2O_3, rinsed with deionized water, and then sonicated in 1:1 HNO_3, acetone and deionized water for 1 min, respectively. Then, the GCE was scanned using cyclic voltammetry (CV) between −1.0 V and 1.0 V in 0.5 $mol \cdot L^{-1}$ H_2SO_4 at 100 $mV \cdot s^{-1}$ for 20 cycles, followed by rinsing with deionized water. After drying, 10 μL of positively charged CS solution (1 $mg \cdot mL^{-1}$, pH 5.0, p*K*a ≈ 6.5) [34] were dripped onto the surface of GCE and half-dried in air, forming a CS precursor layer. The MWCNTs/CS/GCE electrode was prepared by adding 10 μL of negatively charged MWCNTs aqueous dispersion (1 $mg \cdot mL^{-1}$, pH 7.0) onto the surface of the CS/GCE electrode and dried in air.

2.4. Fabrication of MIPs and non-imprinted polymers (NIPs) modified electrodes

The MWCNTs/CS/GCE electrode was immersed in 10 mL phosphate buffer (pH 8.0) solution containing 0.5 mmol 3-APBA and 0.5 mmol EP. Electropolymerization was performed via CV from −0.16 V to 0.94 V for 20 cycles at a scan rate of 50 $mV\,s^{-1}$, obtaining a polymer film modified electrode. Subsequently, the embedded EP template molecule was extracted using both chemical and electrochemical methods. The EP template molecules were first extracted by immersing the electrode in 0.5 $mol \cdot L^{-1}$ H_2SO_4 aqueous solution. After rinsing with deionized water, the modified electrode was subjected to electrochemical scan between 0 and 1.5 V for 10 cycles in 0.5 $mol \cdot L^{-1}$ H_2SO_4 solution to remove the residual template molecules. The procedure for preparing the PAPBA(MIPs)/MWCNTs/CS/GCE sensor was depicted in Scheme 1.