Journal of Electroanalytical Chemistry 752 (2015) 17–24

Contents lists available at ScienceDirect

Journal of Electroanalytical Chemistry

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

Simultaneous electroanalysis of norepinephrine, ascorbic acid and uric acid using poly(glutamic acid) modified carbon paste electrode

P.S. Ganesh, B.E. Kumara Swamy *

Department of PG Studies and Research in Industrial Chemistry, Kuvempu University, Jnana Sahyadri, Shankaraghatta 577451, Shimoga, Karnataka, India

ARTICLE INFO

Article history:
Received 16 March 2015
Received in revised form 31 May 2015
Accepted 1 June 2015
Available online 3 June 2015

Keywords:
Electroanalysis
Norepinephrine
Ascorbic acid
Uric acid
Electropolymerization
Voltammetry

ABSTRACT

A sensitive, selective and reproducible electrochemical method was developed for the electroanalysis of norepinephrine (NE) using poly(glutamic acid) modified carbon paste electrode. The modified electrode shows excellent electrocatalytic activity towards the oxidation of NE in phosphate buffer solution (PBS) of pH 7.4 by cyclic voltammetric and differential pulse voltammetric techniques. The lower limit of detection of NE was found to be 0.43 μM. The interference studies showed that the modified electrode exhibits excellent selectivity in the presence of large excess of ascorbic acid (AA) and uric acid (UA). The separation of the oxidation peak potentials for NE–AA and NE–UA were about 0.262 V and 0.122 V, respectively. This peak differences were large enough to determine NE, AA and UA individually and simultaneously. This work provides a simple and easy approach to selectively detect NE in the presence of AA and UA.

1. Introduction

Norepinephrine (NE) belongs to a family of catecholamine's plays multiple significant roles including as a hormone and a neurotransmitter [1]. NE is secreted in the adrenal medulla and maintains important physiological functions in the central nervous system. It affects muscle and tissue control, stimulates arteriole contraction, decreases peripheral circulation and activates lipolysis in adipose tissue [2,3]. It is also critical in the neurological disease, heart failure; DNA breaks in cardiac myoblast cells and diabetes. Recent reports have indicated that NE enhances adhesion of human immune deficiency virus-1 (HIV-1)-infected leukocytes to cardiac micro vascular endothelial cells and also accelerates HIV replication via protein kinase [4,5]. Hence, it is very necessary to develop sensitive, selective and practically reliable methods for the direct determination of the level of NE for monitoring physiological activities and diagnosing diseases [6–8].

Uric acid (UA) is the primary product of purine metabolism in the human body and a major nitrogenous compound in the urine [9]. Its abnormality in human body leads to many severe diseases, such as gout, hyperuricemia and Lesch–Nyhan disease [10,11]. Increased urate level also leads to the pneumonia and leukaemia [12]. Ascorbic acid (AA) is also known as vitamin-C and is a water soluble compound that take part in the maintaining many important life processes. It has been used as a medicine for the treatments of common cold, mental illness and cancer [13]. It can be chemically or electrochemically oxidized to dehydroascorbic acid [14]. Hence, monitoring the concentration of these biological compounds is very important in clinical diagnosis. There are so many endless reports for the voltammetric determination of these molecules either individually or in presence of probable interferences [15–22]. These biomolecules usually coexist in biological fluids and also the direct electrochemical oxidation of NE and AA at bare electrode gives poor and overlapped voltammetric response with sluggish electron transfer kinetics making their individual identification very difficult and also requires high over potential due to fouling of the electrode surface due to the adsorption of oxidized products. Therefore, the modification of bare carbon paste electrode (BCPE) is an attractive and emerging field for the electroanalytical researchers to contribute their skill to the scientific community. During the last few decades the modification of electrode by the application of electropolymerization technique is of great importance in constructing electrochemical sensors and biosensors because of their practical advantages such as selectivity, good reproducibility, electrochemical reversibility, stability, and low cost [23–25]. In support to this there are so many literatures on the usage of these modified electrodes for the qualitative and quantitative determination of catecholamine's [26–28]. The present work demonstrates the fabrication of a stable and selective electrode by electropolymerising L-glutamic acid at the BCPE using cyclic voltammetric technique. The modified poly(glutamic acid)

* Corresponding author.
E-mail address: kumaraswamy21@yahoo.com (B.E.K. Swamy).

http://dx.doi.org/10.1016/j.jelechem.2015.06.002
1572-6657/

modified carbon paste electrode (MCPE) was successfully used for the determination of NE in the presence of probable interferences like AA and UA. This work reports about sensitivity, selectivity, stability and reproducibility of an important neurotransmitter at poly(glutamic acid) MCPE at physiological pH of 7.4 for the electrochemical analysis of NE, AA and UA. The oxidation mechanism of NE, AA and UA is shown in scheme 1.

2. Experimental section

2.1. Reagents

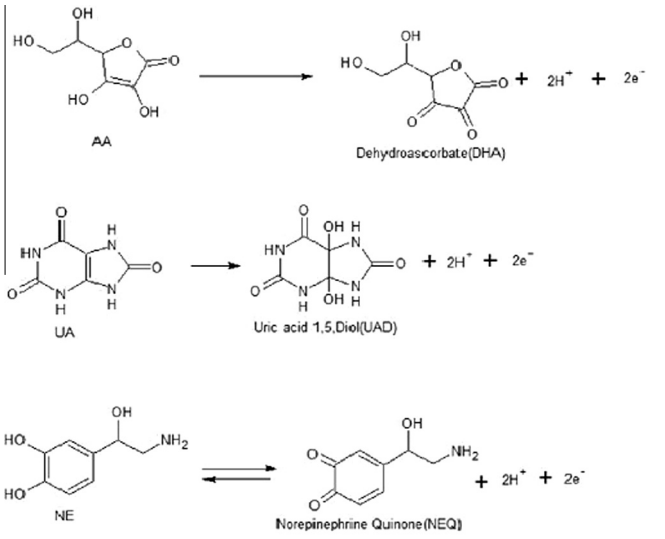
Norepinephrine (L-Noradrenaline hydrochloride) (NE) was purchased from Fluka analytical, uric acid (UA), ascorbic acid (AA), L-glutamic acid were purchased from Himedia. The stock solution 25×10^{-4} M NE, 25×10^{-3} M UA, 25×10^{-3} M AA was prepared in 0.1 M perchloric acid, 0.1 M NaOH, and double distilled water respectively. The phosphate buffer solutions (PBS) with different pH levels were prepared by mixing the same ionic strength (0.2 M) solutions of Na_2HPO_4 and NaH_2PO_4 H_2O solutions at different ratios. The pH levels were adjusted by adding 0.1 M CH_3COOH and/or 0.1 M NaOH solution and PBS of physiological pH 7.4 was used as a supporting electrolyte. Graphite powder of 50 μm particle size was purchased from Merck and silicone oil from Himedia was used to prepare carbon paste electrode (CPE). All the chemicals mentioned were all of analytical grade used as received without any further purification.

2.2. Apparatus

All electrochemical experiments including cyclic voltammetry (CV) and differential pulse voltammetry (DPV) were performed using a model CHI-660c (CH Instrument-660 electrochemical workstation). A conventional three electrode system was used in a single compartment electrochemical cell with a saturated calomel electrode (SCE) as a reference, a platinum counter electrode, and bare carbon paste electrode or poly(glutamic acid) modified carbon paste electrode as working electrode.

2.3. Preparation of the working electrode

The bare carbon paste electrode (BCPE) was prepared according to the reported literature [29]. Electrochemical polymerization of L-glutamic acid at the BCPE was carried out using cyclic voltammetric method in aqueous solution containing 1 mM L-glutamic acid monomer in 0.2 M PBS of pH 7.4. The electropolymerization was achieved by the formation of film that grew between −1.0 V and +1.3 V at the scan rate of 0.1 Vs^{-1} for 12 cycles. After that the electrode was rinsed thoroughly with double distilled water.



Scheme 1. Oxidation mechanism of AA, UA and NE.

3. Result and discussion

3.1. Electrochemical polymerization of L-glutamic acid on BCPE

The poly(glutamic acid) modified carbon paste electrode (MCPE) was prepared by placing 1.0 mM solution of L-glutamic acid monomer in 0.2 M PBS of pH 7.4 in an electrochemical cell. Over the potential range of −1.0–1.3 V with scan rate 0.1 Vs^{-1}. It can be seen from Fig. 1 the anodic peak currents enhanced gradually, indicating the formation and growth of an electroactive layer on the surface of BCPE. After the ninth cycle, the increase of this peak current tended to be almost constant and becomes more stable; suggesting that growth of polymerization was reached the level of saturation [30]. The mechanism of electropolymerization of glutamic acid is described in scheme 2.

From the obtained experimental results the thickness of the film has a significant influence on the electrocatalytic property of the carbon paste electrode. The extent of level of thickness was also calibrated. The coating was controlled by varying the number of multiple cycles on the CPE (from 4 to 16 multiple cycles) and corresponding electrocatalytic activity towards oxidation of 0.1×10^{-4} M NE in PBS of pH 7.4 was investigated. Fig. 2 shows that at 12 multiple cycles, both the anodic and cathodic peak currents were increased. Therefore twelve cycles was chosen for the electropolymerization of L-glutamic acid. With the polymerizing cycles increasing, the electrocatalytic response of NE increased at first. But when the polymerizing cycles is more than 12, the peak currents begin to decrease. This was due to the increase in thickness of the film that would prevent the electron transfer process. Therefore, 12 cycles was used for all electrochemical analysis.

3.2. Scanning Electron Microscope (SEM) analysis of poly(glutamic acid) MCPE

The SEM was employed to study the surface morphology of BCPE and poly(glutamic acid) MCPE as shown in Fig. 3. The surface of BCPE is of irregular shape (A). After the electropolymerization, the electrode surface is covered by a glutamic acid (B), which

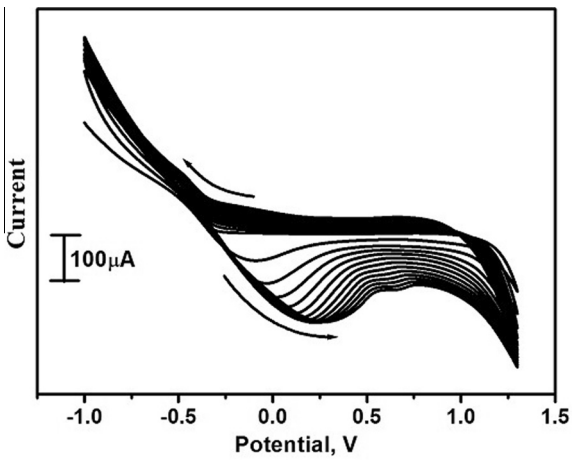


Fig. 1. Cyclic voltammograms of preparation of poly(glutamic acid) MCPE. 1 mM solution in PBS of pH 7.4 at 12 cycles with scan rate 0.1 Vs^{-1}.