Sensors and Actuators B 209 (2015) 391–398

Contents lists available at ScienceDirect

Sensors and Actuators B: Chemical

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

Enhancement of amperometric response to tryptophan by proton relay effect of chitosan adsorbed on glassy carbon electrode

Verónica I. Paz Zanini [a,*], Rodrigo E. Giménez [a], Omar E. Linarez Pérez [b], Beatriz A. López de Mishima [a], Claudio D. Borsarelli [a,*]

[a] *Centro de Investigaciones y Transferencia de Santiago del Estero (CITSE-CONICET), Universidad Nacional de Santiago del Estero, RN9, km 1125, CP4206 Santiago del Estero, Argentina*

[b] *INFIQC-CONICET Departamento de Fisicoquímica, Facultad de Ciencias Químicas, Universidad Nacional de Córdoba, Haya de la Torre y Medina Allende, 5000 Córdoba, Argentina*

ARTICLE INFO

Article history:
Received 29 August 2014
Received in revised form
10 November 2014
Accepted 14 November 2014
Available online 26 November 2014

Keywords:
Tryptophan
Chitosan
Modified glassy carbon electrodes
Electro-oxidation
Proton relay

ABSTRACT

A simply prepared, low-cost, and sensitive electrochemically activated glassy carbon electrode (GCE_a) modified with absorbed chitosan (CHIT) film for quantification of tryptophan (Trp) is reported. Combination of cyclic voltammetry (CV), differential pulse voltammetry (DPV) and electrochemical impedance spectroscopy (EIS) techniques were used for characterization of the electro-oxidation of the amino acid and the electro-analytical performance of the CHIT-modified electrode. The electro-oxidation of Trp involves an irreversible two-electron and two-proton transfer process in both bare and modified electrodes, but the adsorption of CHIT as a polycation onto GCE_a produces a ≈4-fold increase of the oxidation current of Trp without changing both the oxidation potential and the heterogeneous reaction rate constant, suggesting that the biopolymer behaves as a proton relay species, probably due to hydrogen bonding/proton acceptor capability of hydroxyl and ether groups of CHIT. Finally, the electro-analytical features of the CHIT-modified electrode as Trp sensor were also evaluated, obtaining a linear response range up to 130 μM Trp, sensitivity of 0.68 $\mu A\,\mu M^{-1}$ and detection limit of 0.04 μM Trp, with almost no interference of other amino acids.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

Tryptophan (Trp) is an essential amino acid, with a significant role in cell metabolism as a protein building block and in the synthesis of neurotransmitters, such as serotonin [1]. Therefore, depletion of Trp in the human body would cause low levels of serotonin and this may be involved in rapid fluctuations in mood, depression, aggression and in blocking the analgesic effect of morphine [2]. Moreover, Trp deficiency has also been associated with Alzheimer Disease (AD), since it was shown that increasing Trp intake would decrease pathological plaque in AD [3]. On the other hand, toxic products generated in the brain by improper metabolism of Trp could cause hallucinations and delusions [2]. Also in animals the absence of Trp produces significant effects, such as delayed growth and maturation of the central nervous system in rats [4], as well as affecting the thyroid gland and energy waste in chickens [5].

Therefore, the development of sensitive, rapid and reliable analytical methods for the quantification of Trp is a current issue for its application in both health and dietary aspects of humans and animals. Several conventional analytical techniques such as high-performance liquid chromatography (HPLC) [6], liquid chromatography-tandem mass spectrometry [7], spectrophotometry [8], spectrofluorometry [9] and capillary electrophoresis [10] have been widely used for the detection and quantification of Trp in different type of samples. However, most of these techniques have the disadvantages of requiring expensive and not always available equipment and in some cases a complex and time-consuming pretreatment of the sample is needed.

On the other hand, electrochemical techniques have been widely used for the detection and quantification of a variety of standard and real samples including amino acids, and are characterized by its simplicity, precision and sensitivity [11,12]. However, the electrochemical response of Trp is not always satisfactory, mainly because of the slow rate of heterogeneous electron transfer at the electrode surface [13]. In order to overcome this disadvantage, several kinds of nano-structured modified electrodes have been used to

* Corresponding authors. Tel.: +54 385 4509528x1797.
E-mail addresses: vipzanini@hotmail.com (V.I. Paz Zanini), cborsa@unse.edu.ar (C.D. Borsarelli).

http://dx.doi.org/10.1016/j.snb.2014.11.087
0925-4005/© 2014 Elsevier B.V. All rights reserved.

improve the electrochemical response of Trp, such as the incorporation of multi-walled carbon nanotube/cobalt salophen [14], electrospun carbon nanofibers [12], or silver nanoflakes deposited on the surface of molybdenum sulfide [15]. Albeit these modified electrodes exhibit a good performance for electrochemical detection of Trp, most of them require laborious procedures to modify the electrode surface, e.g. synthesis of nanoparticles or nanocomposites.

As a different approach, in the recent years the natural biopolymer Chitosan (CHIT), which is obtained from deacetylated chitins, has begun to be used for the modification of electroactive surfaces [16–18]. This biomolecule is a primary aliphatic amine derivative [19], and under specific conditions it is able to form highly water swellable hydrogels [20]. Recently, Seng et al. [17] have shown that acetylene black paste electrodes (ABPE) modified with either CHIT or salicylaldehyde modified chitosan (s-CHIT) increased the anodic stripping current of Trp compared with both ABPE and carbon paste bare electrodes. However, despite the detection improvement, the role of CHIT on the reaction mechanism of electro-oxidation of Trp remained elusive.

In this work, we describe the electro-analytical response of a low-cost CHIT-modified glassy carbon electrode prepared by a simple and quick procedure. The electro-oxidation of Trp was characterized with both bare and CHIT-modified electrodes as function of pH, and it was confirmed that the single voltammetric oxidation peak is produced by a two electron and two proton irreversible reactions [21], independently of the presence of adsorbed CHIT. However, the presence of the biopolymer produces an almost 4-fold increment of the anodic peak current without modification of both the oxidation potential and the heterogeneous reaction rate constant, suggesting that adsorbed CHIT acts as a proton relay species rather than as a catalyst. Additionally, the electro-analytical parameters for detection of Trp with the CHIT-modified electrode were characterized in detail, including the interference effect of several amino acids, and its analytical performance for quantification of Trp in a commercial pharmaceutical formula.

2. Experimental

2.1. Materials

Chitosan (CHIT) from crab shells (minimum 85% deacetylated), tryptophan (Trp) and potassium ferricyanide $K_3[Fe(CN)_6]$ (99%) were purchased from Sigma–Aldrich (Argentina S.A.). Potassium ferrocyanide $K_4[Fe(CN)_6]$ and acetic acid were from Cicarelli (Argentina). Phosphate buffer solution ($2 < pH < 12$) was made by mixing reagent grade phosphoric acid (H_3PO_4), dibasic (Na_2HPO_4) and/or monobasic (NaH_2PO_4) sodium phosphate salts from J.T. Baker (Mexico D.F., Mexico). All other reagents (analytical grade) were from Parafarm (Argentina) and used without further purification. Dietary supplement capsules of 500 mg of L-tryptophan (Vitabay®, VB 1057) were purchased in a local pharmacy. Triply distilled water was used for all solutions, which were deoxygenated by bubbling for at least 15 min with high-purity nitrogen (99.99% Indura, Argentina) prior to electrochemical measurements and keeping the gas flow over the solution during the experiments.

2.2. Methods

2.2.1. UV–vis absorption and fluorescence measurements

UV–vis absorption spectra were registered using an Agilent 8453 diode array spectrophotometer (Palo Alto, CA, USA). Fluorescence emission and anisotropy measurements were achieved with a Hitachi F-2500 spectrofluorometer (Kyoto, Japan). Excitation of Trp solutions was performed at 280 nm and both emission spectrum and anisotropy was measured in the 300–500 nm spectral range. All measurements were performed with continuous stirring using a magnetic bar and at constant temperature of 25 °C. Anisotropy measurements were performed using the classical L-format and calculated as described before [22].

2.2.2. Electrochemical measurements

Cyclic voltammetry (CV), differential pulse voltammetry (DPV) and electrochemical impedance spectroscopy (EIS) studies were carried out with an Autolab PGSTAT 30 potentiostat using the software package GPES and FRA 4.9 (Eco-Chemie, Utrecht, Netherlands) [23]. Electrochemical experiments were performed in a three-compartment electrochemical cell with standard taper joints thus all compartments could be hermetically sealed with Teflon® adapters. Working electrodes were prepared with glassy carbon disks, diameter 2 mm (Metrohm, 6.1204.600). A large-area platinum wire was used as a counter electrode. The potentials were measured against a reference electrode $Ag|AgCl|Cl^-$ (3 M) and all measurements were performed at room temperature (25° C). For the EIS experiments, the sine wave potential amplitude applied was 5 mV at a bias potential of 200 mV and frequency range 0.05 Hz–10 KHz. Experimental EIS curves were fitted by the non-linear least squares method provided by Autolab FRA 4.9® software comparing the experimental system to a Randles equivalent circuit (Fig. S1 of Supplementary Information). DPV measurements were performed from 0.60 to 1.2 V at 0.020 V s^{-1} with potential pulse amplitude of 0.050 V and data sampling width of 0.010 V.

2.2.3. Preparation of the CHIT-modified electrode

The surface of the glassy carbon electrodes (GCE) was polished sequentially with alumina powder (Buehler, USA) of decreasing particle size of 1.0, 0.3 and 0.05 μm, and thoroughly rinsed with triply distilled water and sonicated for 1 min between polishing stages. Prior to the adsorption of the chitosan layer, electrochemical activation of the polished GCE was performed as follows: the electrode was cleaned in 0.1 M sodium phosphate buffer solution at pH 7 by cyclic voltammetry between −0.50 and +1.2 V at 0.050 V s^{-1} until a stable voltammogram profile was observed. Subsequently, an oxidation potential of 2.0 V for 60 s followed by a pulse reduction at −1.1 V for 30 s also in a solution of 0.1 M phosphate buffer at pH 7 was applied. Afterwards, the electrode surface was rinsed with triply distilled water and dried under nitrogen flow. The electrochemical activated electrode was immediately used for its modification with the chitosan film.

The adsorption of CHIT film on the previously activated glassy carbon electrode (GCE_a) was formed by dipping the activated electrode in a 0.5 wt.% CHIT dissolved in 2 wt.% acetic acid solution. The optimal time for chitosan film adsorption was ≥20 min, as tested by the constant value of the oxidation peak current of a 100 μM Trp standard solution in 0.1 M phosphate buffer pH 6.0 (±0.1) obtained by cyclic voltammetry at 0.050 V s^{-1}. The CHIT-modified electrode (CHI/GCE_a) also was immediately used after preparation.

2.2.4. Real sample preparation for Trp quantification

Commercial tablets formulation containing 500 mg Trp (Vitabay® VB 1057) were purchased in a local pharmacy. Tablets were triturated and mixed thoroughly during 1 h with 10 ml of 0.1 M phosphate buffer (pH 2.0). Then, the suspension was filtered, and the collected solution was diluted to 100 ml with the same buffer. Subsequently, a 15 μl aliquot of the diluted solution was added to the electrochemical cell containing 10 ml of the phosphate buffer.

Download English Version:

https://daneshyari.com/en/article/7146157

Download Persian Version:

https://daneshyari.com/article/7146157

Daneshyari.com