Highly sensitive nanostructure voltammetric sensor employing Pt/CNTs and 1-butyl-3-methylimidazolium hexafluoro phosphate for determination of tryptophan in food and pharmaceutical samples

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1. Introduction

Tryptophan is well known as an essential amino acid in human and herbivores bodies, and the precursors of catecholamine synthesis in human body [1]. Trp has to be taken by foods in daily diet to maintain the nitrogen balance. It also serves as serotonin and melatonin precursors which regulate several human functions such as sleep, mood and various aspects of mental health [2]. Since, Trp is a usual constituent of most protein-based foods or dietary proteins, then rapid and sensitive determination of Trp in food and medicine is of concern for scientists. Many analytical methods have been suggested for the determination of Trp, including high performance liquid chromatography [3–5], chemiluminescence [6–8], spectroscopic [9,10], and electrochemical method [11–14]. Among them, electrochemical sensors have advantages such as simple analysis, good selectivity, lower cost, high sensitivity and high speed for electroactive compounds analysis [15–25]. So, we used this strategy for trace analysis of Trp in food and pharmaceutical samples.

Chemically modified electrodes with room temperature ionic liquids and nanomaterials (CMILNME) are high quality tools for the trace analysis of electrochemical determination for biological, environmental and pharmaceutical samples in different condition [26–34]. Scientific investigations show that nanomaterials have a good quality for application in sensor field due to high surface area of nanomaterials and good electrical conductivity [35–40]. Reducing the over-potential and increasing the sensitivity, including the substantial benefits of application of CMILNME as voltammetric electroactive compound analysis [41–51]. Modification of electrodes are necessary for trace analysis of Trp due to high overvoltage of this compound at a surface of bare electrode [51].

In this study, we describe the synthesis and application of Pt/CNTs nanocomposite modified carbon ionic liquid paste electrode, with utilization of [C4mim]-[PF6] as a good conductive binder. The electrochemical behavior of Trp at [C4mim]-[PF6]/Pt/CNTs/CPE, at carbon paste electrode modified with [C4mim]-[PF6] ([C4mim]-[PF6]/CPE), at Pt/CNTs/CPE paste electrode (Pt/CNTs/CPE), and at carbon paste electrode (CPE) was investigated. The results showed the superiority of [C4mim]-[PF6]/Pt/CNTs/CPE to the other electrodes in terms of higher sensitivity. The [C4mim]-[PF6]/Pt/CNTs/CPE is sensitive enough for the determination of Trp in meat and tablet samples.
2. Experimental section

2.1. Apparatus and compounds

Mineral oil, Tryptophan, NaOH and graphite powder (<50 μm) were obtained from Merck. All of the voltammetric investigation performed using Autolab, potentiostat/galvanostat connected to a three-electrode cell, Azar Electrode, linked with a computer (Pentium IV) and with Autolab software. Three-electrode cell assembly consisting of a platinum wire as an auxiliary electrode and an Ag/AgCl (KCl_sat) electrode as a reference electrode was used.

2.2. Preparation of the [C4mim]-[PF6]/Pt/CNTs/CPE

[C4mim]-[PF6]/Pt/CNTs/CPE was prepared by mixing of 0.2 g of [C4mim]-[PF6], 0.80 g of the paraffin oil, 0.15 g of Pt/CNTs, and 0.85 g of graphite powder. Then the mixture was mixed well for 40 min until a uniformly wetted paste was obtained. A portion of the paste was filled firmly into one glass tube as described above to prepare Pt/CNTs.

2.3. Synthesis of Pt/CNTs nanostructure

Pt/CNTs nanocomposite obtained by mixing stoichiometric of purified carbon nanotubes, Pt (acac)2 (0.5 mmol) and 1,2 Hexadecanediol (5 mmol) in 40 mL phenylether at room temperature under a flow of N2 atmosphere, and then the solution was heated at 120 °C for 45 min. Reduction of Pt atoms in the presence of 1,2 Hexadecanediol have been obtained until starting the reduction of Pt salt and nucleation of Pt nanoparticles. Afterward the solution was heated to 250 °C for 100 min during the reflux process. After completion of reaction, the black product solution was cool to room temperature under flow of N2 atmosphere. Then the product was rinse with ethanol and hexane solution for 3 times and dried in 200 °C under Ar atmosphere.

2.4. Preparation of real samples

5 mL of meat sample extract was added to a volumetric flask and diluted to the mark with the supporting electrolyte (PBS pH 8.0).

50 mg of Trp tablet finely powdered and then dissolved in 50 mL water with ultrasonication. Then, 0.5 mL of the solution plus 9.5 mL of the buffer (pH 8.0) was used for the analysis with standard addition method.

3. Results and discussion

3.1. Pt/CNTs nanocomposite characterization

The morphology of the as-grown Pt/CNTs nanocomposite was characterized by TEM. Typical TEM micrograph of the synthesized Pt/CNTs nanocomposite is shown in Fig. 1. Results confirm synthesis of Pt/CNTs nanocomposite.

3.2. Voltammetric investigation

According to Scheme 1, we found that electro-oxidation of Trp is relative to pH value in aqueous solution. Therefore, the effect of pH on electro-oxidation of Trp was investigated using SWV technique (Fig. 2 inset). Result shows that the electro-oxidation peak current for Trp has a maximum sensitivity at a pH 8.0 and this condition was chosen as the best optimal experimental condition for this work (Fig. 2).

The active surface area of the using sensors was estimated according to the Randles–Sevcik equation for a known concentration of K4Fe(CN)6/K3Fe(CN)6. The results obtained were 0.24, 0.19, 0.13 and 0.09 cm2 for [C4mim]-[PF6]/Pt/CNTs/CPE, [C4mim]-[PF6]/PE, Pt/CNTs/CPE and CPE, respectively. The obtained results show that the presence of nanoparticles and ionic liquids together contributed to an increase in the active surface area of the using sensor.

Current density derived from the cyclic voltammograms of 100 μM Trp (pH 8.0) at the surface of modified and bare electrodes with a scan rate of 50 mV/s shows in Fig. 3 inset. The results confirm that the presence of Pt/CNTs and [C4min]-[PF6] together causes the increase of the electrode current density. Fig. 3 shows CV of 100 μM Trp pH 8.0 at the surface of modified and unmodified electrodes with a scan rate of 50 mV/s. [C4mim]-[PF6]/Pt/CNTs/CPE exhibited significant oxidation peak current around 785 mV with the peak current of 13.1 μA (Fig. 3, curve a). However, low electro-oxidation activity peak was observed at Pt/CNTs/CPE (Fig. 3, curve c) and at CPE (Fig. 3, curve d) over the same condition. The Trp oxidation peak potential at Pt/CNTs/CPE and at carbon paste electrode observed around 820 mV vs. the reference electrode with the oxidation peak current of 3.52 and 7.05 μA, respectively. In addition, at the surface of [C4mim]-[PF6]/CPE, the oxidation peak appeared at 790 mV with the peak current was 10.0 μA (Fig. 3, curve b), which indicated the presence of [C4mim]-[PF6] in CPE could enhance the peak currents and decrease the oxidation potential. A significant negative shift of the currents starting from
oxidation potential for Trp and dramatic increase of current of Trp indicated the catalytic ability of [C4mim]-[PF6]/Pt/CNTs/CPE to Trp oxidation. The results indicated that the presence of Pt/CNTs on [C4mim]-[PF6]/Pt/CNTs/CPE surface had great improvement with the electrochemical response, which was mainly owing to excellent characteristics of Pt/CNTs such as good electrical conductivity. The suitable electronic properties of Pt/CNTs nanocomposite together with the [C4mim]-[PF6] gave the ability to promote charge transfer reactions, good anti-fouling properties, especially when mixed with a higher conductive compound such as ILs as an electrode particle.

The effect of scan rate ($\nu$) on the electrochemical oxidation peak current of Trp was also examined (Fig. 4 inset). The results of this investigation showed that the Trp oxidation peaks current increased linearly with increasing the square root of scan rate that ranged from 5 to 60 mV/s (Fig. 4). The result shows that the electrode process for oxidation of Trp is controlled under the diffusion step [52–65]. Also, the peaks potential shifts in negative direction when the scan rate increases, meaning that the electrochemical reaction is irreversible.

To obtain further information on the rate determining step, a Tafel plot was developed for the Trp at a surface of [C4mim]-[PF6]/Pt/CNTs/CPE using the data derived from the raising part of the current–voltage curve (Fig. 5). The slope of the Tafel plot is equal to $n(1 - \alpha)F/2.3RT$ which comes up to 0.2594 V decade$^{-1}$. We obtained $\alpha$ as 0.77.

Chronoamperometric measurements of Trp at [C4mim]-[PF6]/Pt/CNTs/CPE using the data derived from the raising part of the current–voltage curve (Fig. 6A). For Trp with a diffusion coefficient of D, the current observed for the electrochemical reaction at the mass transport limited condition is described by the Cottrell equation. Experimental plots of $I$ vs. $t^{-1/2}$ were employed, with the best fits for different concentrations of Trp (Fig. 6B). The slopes of the resulting straight lines were then plotted vs. Trp concentration. From the resulting slope and Cottrell equation the mean value of the D was found to be $9.56 \times 10^{-5}$ cm$^2$/s.

3.3. Figure of merit

Limit of detection and linear dynamic range are two important analytical parameters that must be report for new analytical sensors [58–66]. The electrochemical determination of Trp has been studied at the optimum condition using SWV method. Fig. 7 shows the SW voltammograms (inset) responses and the calibration curves for Trp. The results showed that the dynamic linear range of Trp was 0.1–400.0 μM with the correlation coefficient of 0.9918. The detection limit of the Trp was 0.04 μM ($S/N = 3$) along with the sensitivity of 0.0469 μA/μM.
4.3. Stability and reproducibility

We have investigated the stability of the [C4mim]-[PF6]/Pt/CNTs/CPE in this study. It was observed that [C4mim]-[PF6]/Pt/CNTs/CPE conserved their activity for long time durations (i.e. 40 days) when they were stored in a laboratory. To investigate the reproducibility of the [C4mim]-[PF6]/Pt/CNTs/CPE, electrochemical oxidation of 20.0 μM Trp at the same conditions were performed by using 5 different [C4mim]-[PF6]/Pt/CNTs/CPE. The results showed that an acceptable reproducibility with a RSD of 3.1% was obtained. The results indicate that the [C4mim]-[PF6]/Pt/CNTs/CPE has good fabrication reproducibility.

3.5. Interference study

The possible interference for Trp determination at [C4mim]-[PF6]/Pt/CNTs/CPE was investigated. Some amino acids, including valine, alanine, threonine, isoleucine, glutamine, aspartic acid, and histidine, had no influence on the current response for 40 μM Trp. Meanwhile, the [C4mim]-[PF6]/Pt/CNTs/CPE showed an inhibiting effect on the oxidation of other electroactive species such as glucose, lactose, fructose and methanol (500 times content).

3.6. Real sample analysis

We study the analytical utility of the [C4mim]-[PF6]/Pt/CNTs/CPE sensor was assessed by applying it to the determination of Trp in a food and tablet samples using standard addition method by SWV method. The results are given in Table 1, confirm that the [C4mim]-[PF6]/Pt/CNTs/CPE retained its efficiency for the determination of Trp in real samples.

4. Conclusions

The carbon paste electrode modified with [C4mim]-[PF6] and Pt/CNTs have excellent electrocatalytic activity towards Trp. Besides the good analytical performance, [C4mim]-[PF6]/Pt/CNTs/CPE has the advantages of good reproducibility, stability and selectivity. Under the optimum conditions, the peak current was proportional to the Trp concentration in the range of 1.0 to 400.0 μM with the detection limit of 0.04 μM. The [C4mim]-[PF6] and Pt/CNTs was successfully used for the determination of Trp in food and pharmaceutical samples.

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References


