Chapter-5

PRETREATED/CARBON PASTE ELECTRODE BASED VOLTAMMETRIC SENSORS FOR THE DETECTION OF DOPAMINE IN PRESENCE OF ASCORBIC ACID AND URIC ACID

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5.1. Introduction

A voltammetric resolution for the determination of dopamine using a Pretreated/Carbon paste electrode was developed. The Pretreated/Carbon paste electrode showed excellent electrocatalytic activity towards the oxidation of dopamine in phosphate buffer solution (pH 7.0). From the electrochemical studies of scan rate, the overall electrode process was diffusion and adsorption-controlled. The pH effect suggested that equal number of protons and electrons were involved in the electrochemical detection of dopamine. Detection limit (LOD) was calculated in phosphate buffer solutions at pH 7.0 and the interference studies showed that the modified electrode exhibited excellent selectivity in the presence of large excess of ascorbic acid and uric acid. The separation of the oxidation peak potentials for dopamine–ascorbic acid and dopamine–uric acid was found to be 0.187 V and 0.121 V, respectively. These differences were large enough to determine dopamine, ascorbic acid and uric acid individually and simultaneously by using cyclic voltammetry and differential pulse voltammetric techniques.

5.2. Chemistry and Biological Relevance of Dopamine

The chemistry and biological relevance of dopamine has been explained in details in chapter 3 section 3.2.

5.3. Chemistry and Biological Relevance of Ascorbic Acid and Uric Acid

The chemistry and biological relevance of Ascorbic acid and Uric acid has been explained in details in chapter 4 section 4.3. and section 4.4.

5.4. Review of Cyclic Voltammetry of Dopamine, Ascorbic Acid and Uric Acid

Dopamine is a simple organic chemical in the catecholamine family and a monoamine neurotransmitter which played a number of important physiological roles in the bodies of animals. Dopaminergic neurons of the midbrain are the main source of dopamine in the brain [1]. Dopamine has been shown to be involved in the control of movements, the signaling of error in prediction of reward, motivation, and cognition. Cerebral dopamine depletion has been the hallmark of Parkinson's disease [1]. Abnormalities in dopaminergic neurotransmission demonstrated in painful clinical conditions, including burning mouth syndrome [2], fibromyalgia [3,
4] and restless legs syndrome [5]. Other pathological states have also been associated with dopamine dysfunction, such as schizophrenia, autism, and attention deficit hyperactivity disorder, as well as drug abuse. Several important diseases of the nervous system had been associated with dysfunctions of the dopamine system. Parkinson's disease, an age-related degenerative condition caused tremor and motor impairment. It was caused by loss of dopamine-secreting neurons in the substantia nigra. Schizophrenia, increased heart rate, renal failure and blood pressure was shown to involve elevated levels of dopamine activity in the mesolimbic pathway and decreased levels of dopamine in the prefrontal cortex [6, 7]. Attention deficit hyperactivity disorder (ADHD) was believed to be associated with decreased dopamine activity. Therefore, the determination of concentration of dopamine in body fluids was of great significance in the field of pharmacology. A major problem in DA determination is the resolution between DA and coexisting species such as uric acid (UA) and ascorbic acid (AA). UA was a primary product of purine metabolism in the human body [8]. Its abnormal concentration level caused many diseases, such as gout, hyperuricaemia and Lesch-Nyhan disease [9]. Therefore, the research of UA determination has been of great importance in reality also [10]. However, AA and UA has been oxidized at potentials close to that of DA, resulting in an overlapping voltammetric response [11-13]. To overcome from this problem, various chemically modified electrodes have been used for the sensitive and selective determination. Till now there are many techniques have been used and however, it is an important task among researchers to improve the sensitivity and selectivity of the electrode towards the detection of dopamine. Earlier reported literature found that the activation of the CPE was carried out by electro-chemical moderately fast scanning in basic solutions (e.g., NaHCO₃ or NaOH, 0.5 M) within a given potential interval. The response of the glucose oxidase and the hydroquinone was tested before and after surface activation, either in solution or coimmobilized on the CPE. The studies were conducted in quiescent solutions and under hydrodynamic conditions. The results demonstrated an improvement in the biosensor response, as determined by its dynamic linear range and sensitivity at low redox potential, after surface activation [14]. The development of reagentless biosensor was a great challenge for researchers, therefore, in the present work developed the carbon paste
electrode was activated in 0.1 M NaOH under potential between 0.6 to 2.0 V at 5 Vs\textsuperscript{-1} for a 200 cycles in an unstirred solution. The developed one held high stability and good electrocatalytic activity towards the electrocatalytic oxidation of DA in presence of excess of AA and UA at a neutral pH. Characterisation of the electrode was carried by both cyclic voltammetry and differential voltammetric techniques. The activated electrode not only was having sensitivity but also it separate voltammetric signals and showed lower detection limit when compared to traditional carbon paste electrode. The same method was utilized for applying injection sample and human body fluids for the biomedical application.

5.5. Experimental Section

5.5.1. Reagents

Fine graphite powder (50μm particle size) purchased from Merck and silicon oil (Himedia) was used to prepare modified carbon paste electrode. 25×10^{-4} M stock solution of DA (Himedia) was prepared in 0.1 M perchloric acid solution. 25×10^{-4} M UA (Himedia) stock solution was prepared in 0.1 M NaOH and 25×10^{-3} M AA (Himedia) in double distilled water. Phosphate buffer (0.2 M pH 7.0) (Merck) solution was prepared by mixing the appropriate quantity of 0.2 M aqueous sodium dihydrogen phosphate monohydrate and 0.2 M aqueous disodium hydrogen phosphate. Sodium hydroxide and acetic acid was used for increasing and decreasing the pH of the buffer. Chemicals mentioned above were all of analytical grade used as received. Water used in the preparation of solutions was double distilled water.

5.5.2. Apparatus

The electrochemical experiments was carried out using a model CHI-660c (CH Instrument-660 electrochemical workstation). The electrode system contained the working electrode was BCPE and pretreated/CPE, a platinum counter electrode and saturated calomel as reference electrode.

5.5.3. Preparation and Pretreatment of CPE

The mixture of fine graphite powder and silicon oil of ratio 80%:20% was mixed by hand grinding in an agate mortar for about 45min. The resulting paste was packed in the home made cavity of carbon paste electrode of 3mm in diameter. The
surface of the packed carbon paste was smoothed on tissue paper. The CPE was pretreated by applying a linearly varying potential between 0.6 and 2.0 V at 5 Vs⁻¹ for 200 cycles in an unstirred solution of 0.1 M NaOH [14]. Pretreatment was achieved by oil depletion from the surface with a concomitant increase in the surface hydrophilicity using CV technique [15]. After surface pretreatment, the electrode was thoroughly washed with distilled water and it was applied for further analysis.

The surface area available for reaction of species in the solution can be estimated by the Randles–Sevcik equation (1) [16-18]. This equation relates the peak current for an electron-transfer-controlled process to the square root of the scan rate:

\[ i_p = 2.69 \times 10^5 \times n^{3/2} \times A \times D_0^{1/2} \times C_0^* \times v^{1/2} \]  

(1)

where \( i_p \) is the peak current, \( A \) is the electroactive area (cm²), \( C_0^* \) is the concentration of the electroactive species (mol cm⁻³), \( n \) is the number of exchanged electrons, \( D_0 \) is the diffusion coefficient (cm² s⁻¹), and \( v \) is the scan rate (Vs⁻¹). The value of the diffusion coefficients for DA in buffer solution is obtained from the literature [19].

5.6. Results and Discussion

5.6.1. Electrochemical characterization of pretreated/CPE using standard potassium ferricyanide system

The freshly prepared 1mM potassium ferricyanide and 1M potassium chloride solutions was placed in the electrochemical cell. The fig. 5.1 showed the cyclic voltammograms recorded for the 1 mM potassium ferricyanide at both BCPE (solid line) and at pretreated /CPE (dashed line) at scan rate of 0.1 Vs⁻¹. The low redox peak current signal was observed at BCPE and the anodic and cathodic peak potentials was located at 0.258 V and 0.186 V, respectively and the redox peak potentials difference (\( \Delta E_p \)) was 0.072 V. But the pretreated/CPE showed significant improvement in the redox peak current signals. The anodic and cathodic peak potentials was found at 0.252 V and 0.0191 V. The \( \Delta E_p \) was 0.061 V with increase in peak current. It showed the electrocatalytic property of pretreated /CPE.
5.6.2. Effect of solvent and multiple cycles for pretreatment of electrode

From the experimental results, the molarity of NaOH affected the electrocatalytic property of that electrode. The CPE was pretreated by varying the molarity of NaOH from 0.05 M to 1 M. At 0.1 M NaOH solvent pretreated electrode exhibited higher surface area of the electrode when compared to others and for the corresponding electrode, the variation of cycles in the potential between 0.6 and 2 V at 5 Vs\(^{-1}\) from 50 cycles to 300 cycles in that 200 cycles exhibited significant surface area with minimization in over potential when compared to bare carbon paste electrode. Higher surface area confirmed the high electrocatalytic activity has been shown in fig. 5.2 and recorded in the Tables 5.1a and 5.1b.

5.6.3. Electrochemical response of DA at pretreated/CPE

Fig. 5.3 shows the cyclic voltamograms of 10 μM DA at BCPE and pretreated/CPE in 0.2 M phosphate buffer solution at pH 7.0 and scan rate 0.05 Vs\(^{-1}\). At the BCPE the cyclic voltammogram of DA (solid line) showed an oxidation peak potential at 0.176 V and reduction peak potential at 0.133 V with low current signals. The peak potential separation (ΔEp) was found to be 0.043 V. The electrochemical response of DA at pretreated/CPE showed well defined redox waves of DA with strong increase of the redox peak currents (dashed line). The oxidation peak potential occurred at 0.154 V and reduction peak potential at 0.130 V respectively, with the peak potential separation (ΔEp) 0.024 V. The modified electrode exhibited strong promoting effect and high stability towards the electrochemical oxidation of DA. It was observed that the peak currents enhanced at the pretreated/CPE, which provided more evidence for asserting that the pretreatment in the CPE possessed high electrocatalytic activity towards the DA detection.

5.6.4. Effect of scan rate on DA

According to Randles-Sevick’s equation, the scan rate was directly proportional to peak current. Fig. 5.4A reveals the cyclic voltammogram of DA at pretreated/CPE for 10 μM DA in 0.2 M phosphate buffer solution and scan rates from 0.05 to 1 V s\(^{-1}\) at pH 7.0. The effect of scan rate on the anodic peak current of DA was studied by cyclic voltammetry and increase in the scan rate from increase
was anodic peak current ($I_{pa}$). The graph of anodic peak current ($I_{pa}$) vs. square root of scan rate ($v^{1/2}$) was plotted in the range from 0.05 to 0.2 Vs$^{-1}$. The graph obtained was linearly straight line shown in Fig. 5.4B. A good linearity of the anodic peak currents were proportional to the square root of scan rate ($v^{1/2}$) with correlation coefficient 0.995. It indicated that the electrode transfer reaction was diffusion-controlled process on the pretreated electrode surface. The graph of anodic peak current versus scan rate ($v$) was plotted in the range from 0.2 to 1 Vs$^{-1}$. The graph obtained straight line has been shown in Fig. 5.4C and the good linearity of the anodic peak currents was proportional to the scan rate ($v$) with correlation 0.997. It indicated that the electrode transfer reaction was adsorption-controlled process on the pretreated electrode surface. For a reversible wave, the potential was independent of scan rate and $I_{pa}$ (as well as the current at any other point on the wave) was proportional to $v^{1/2}$. The property of the electrode indicated the diffusion-controlled which depended on “$n$” and the constant could be used to estimate “$n$” by using eq. (2), and recorded in Table 5.2 from the range of 0.05 to 0.2Vs$^{-1}$ [20].

$$E_p - E_{p/2} = 56.5/n \text{ mV at } 25^0 \text{C}$$ (2)

5.6.5. Effect of pH

The effect of pH on the determination of DA in PBS solution at the pretreated/CPE was carefully investigated in the pH range of 5.8–7.8. The cyclic voltammograms of 10 \( \mu \text{M} \) DA was recorded at 0.2 M PBS of 5.8–7.8 solutions. The anodic and cathodic peak potentials shifted to less positive potentials with increasing the pH from 5.8 to 7.8 with increasing anodic peak current of DA increased with increasing pH values 5.8–7.8. Graphs of $\Delta E_p$ versus the pH of the solution and $E^0$ (V) versus the pH of the solution for pretreated/CPE has been shown in Fig. 5.5. The formal potential ($E^0$) of DA decreased with an increase in the pH value. A linear regression equations obtained was $E^0$ (V) = $-0.06374 \text{ pH } + 0.5921$ ($n = 6$, $r^2 = 0.9993$) for the pretreated/CPE. This results confirmed that the equal number of protons and electrons were involved in the electrochemical oxidation of DA [21] and $\Delta E_p$ decreases indicated that the electrode showed good electrocatalytic activity at pH 7 compared to other pH solutions which clearly shown in Fig. 5.5 and for further studies pH 7 was selected for DA.
5.6.6. Electrochemical oxidation of AA at pretreated/CPE

Fig. 5.6 showed the cyclic voltammograms of $4.9 \times 10^{-4}$ M AA at the BCPE (solid line) and pretreated/CPE (dashed line) with pH 7.0 PBS. At the bare CPE the oxidation peak occurred at around 0.22 V. The oxidation of AA at bare electrode was generally irreversible and required high over potential due to fouling of the electrode by the adsorption of oxidized product of AA [22, 23]. However, at the pretreated/CPE, the oxidation peak potential of AA was obtained at around -0.02 V which shifted to negative potential when compared to that of bare CPE, which indicated that pretreatment of the electrode lowers the over potential and favoured the oxidation process of AA. This activated electrode prevented the fouling of the electrode surface, hence faster electron transferred kinetics of AA at the pretreated/CPE. Since the oxidation peak of AA was shifted to negative potential leads to absence of interfere with the measurement of DA.

Fig. 5.7A shows the cyclic voltammogram of AA at pretreated /CPE for $4.9 \times 10^{-4}$ M AA in 0.2 M phosphate buffer solution and scan rates from 0.05 to 1 V s$^{-1}$ at pH 7.0. The effect of scan rate on the anodic peak current of AA was studied by cyclic voltammetry and increase in the scan rate increase was anodic peak current ($I_{pa}$). The oxidation peak potential was observed to shift positively with the increase in scan rate [24], in addition, the graph of anodic peak current ($I_{pa}$) versus scan rate ($v$) was plotted in the range from 0.05 to 1 Vs$^{-1}$. The graph obtained was linearly straight line shown in Fig. 5.7B. A good linearity of the anodic peak currents was proportional to the scan rate ($v$) with correlation coefficient 0.9921 indicated that the electrode transfer reaction was adsorption-controlled process on the pretreated electrode surface.

5.6.7. Electrochemical oxidation of UA at pretreated/CPE

Fig. 5.8 shows the cyclic voltammograms of $2.96 \times 10^{-5}$ M UA for BCPE (solid line) and pretreated/CPE (dashed line) at pH 7.0 PBS. It could be seen that voltammetric peak appeared at about 0.273 V for BCPE, the peak was rather broad suggesting slow electron transfer kinetics, presumably due to the fouling of the electrode surface by the oxidation product [25]. However, at pretreated/CPE, the UA showed well defined redox peak with the oxidation peak potential at 0.261V, which was negatively shifted by 0.012V and reduction peak potential was at 0.238 V. By
this pretreated/CPE, a remarkable increase in anodic peak current and the above results indicated that electrocatalytic reaction occurred between the pretreated/CPE and UA.

Fig. 5.9A shows the cyclic voltammogram of UA at pretreated /CPE for $2.96 \times 10^{-5} \text{ M UA}$ in 0.2 M phosphate buffer solution and scan rates from 0.05 to 1 V s$^{-1}$ at pH 7.0. The effect of scan rate on the anodic peak current of UA was studied by cyclic voltammetry and increase in the scan rate from increase was anodic peak current ($I_{pa}$). The graph of anodic peak current ($I_{pa}$) versus scan rate ($v$) was plotted in the range from 0.05 to 1 V s$^{-1}$. The graph obtained was linearly straight line shown in Fig. 5.9B. A good linearity of the anodic peak currents was proportional to the scan rate ($v$) with correlation coefficient 0.9948. It showed that the electrode transfer reaction was adsorption-controlled process on the pretreated electrode surface [26].

**5.6.8. Differential pulse voltammetry**

**5.6.8a. Individual concentration variation**

The DPV plots recorded in the pretreated/CPE/0.1 M NaOH at pH 7 at concentrations of DA was analyzed in the range from $1 \times 10^{-7}$ to $1 \times 10^{-4}$ M (Fig. 5.10A & 5.10B). There was a dramatic enhancement in the anodic current. The anodic peak current was proportional to concentration of DA and but the better linearity occurred in the range from $2 \times 10^{-6}$ to $1 \times 10^{-5}$ M and the linear regression equation was $I_{pa}(\mu A) = 0.01004 (C_{i} \mu M/L) + 0.1053 (\mu A)$, ($r^2 = 0.9993$ n=11) (Fig. 5.10C) the limit of detection (LOD) for DA in the lower concentration range was $0.198 \times 10^{-6}$ M for pretreated/CPE. The LOD was calculated according to the equation $LOD = KS^0/S$, where K was a constant related to the confidence level. In accordance with the suggestion of the IUPAC, the value of K was 3 at the 99% confidence level, $S^0$ was the standard deviation of Six blank-solution measurements (no added DA). S was the slope of the calibration graph [30]. Similarly, in the Fig. 5.11A and 5.11B showed the concentration effect of AA from $1 \times 10^{-6}$ to $1 \times 10^{-3}$ M but the better linearity occurred in the range from $3 \times 10^{-6}$ to $2 \times 10^{-5}$ M the anodic peak current was proportional to concentration of AA and its linear regression equation was $I_{pa}(\mu A) = 0.00109(C_{i} \mu M/L) + 0.2039(\mu A)$, ($r^2 = 0.9982$ n=11) (Fig. 5.11C). The limit of detection (LOD) for AA in the lower
concentration range was $1.0316 \times 10^{-6}$ M for pretreated/CPE. Fig. 5.12A and Fig. 5.12B shows the concentration effect of UA analyzed in the range from $1 \times 10^{-7}$ to $1 \times 10^{-4}$ M but the linearity occurred in the range from $1 \times 10^{-5}$ to $9 \times 10^{-5}$ M the anodic peak current was proportional to concentration of UA. The linear regression equation was $I_{pa} (\mu A) = 0.0021 (C \mu M/L) + 0.1951 (\mu A)$ ($r^2 = 0.9997$ n=12) (Fig. 5.12C) the limit of detection (LOD) for UA in the lower concentration range was $1.73 \times 10^{-5}$ M for pretreated/CPE. In agreement with the results obtained, it could be observed that the pretreated/CPE was more sensitive toward the DA, when compared with the UA and the AA, and very low detection limit occurred for DA. Moreover, the CPE showed similar analytical features towards DA, AA and UA as those found using different electrodes, namely vitreous carbon for AA [27], Bare Carbon paste Electrode from a 0.1 M NaCl aqueous solution at pH 7 for UA [28] and Triton X-100-modified CPE for DA [29].

5.6.8b. Interference investigation

In order to examine the sensitivity and selectivity of pretreated/CPE, the electrochemical behavior of a mixture of $9.96 \times 10^{-6}$ M DA, $4.9 \times 10^{-4}$ M AA and $2.96 \times 10^{-5}$ M UA was investigated using cyclic voltammetric technique. Fig. 5.13 shows the cyclic voltammograms obtained for DA, AA and UA coexisting at bare CPE, and pretreated/CPE. As shown in Fig. 5.13, bare CPE (solid line) unable to separate the voltammetric signal of DA, AA and UA. Only one broad voltammetric signal for DA, AA and UA was observed at approximately 0.207 V. The fouling of the electrode surface by the oxidation products resulted in a single voltammetric peak for DA, AA and UA. Therefore, it was impossible to use bare electrode for the voltammetric determination of DA, UA in the presence of AA. Moreover, the pretreated/CPE resolved the voltammetric signal into three-well defined voltammetric peaks at -0.051 V, 0.136 V and 0.257 V corresponding to AA, DA and UA respectively (dashed line). The separation between the oxidative peaks of DA and AA was approximately 0.187 V and the separation between the oxidative peaks of DA and UA was approximately 0.121 V. The differential pulse voltammetric technique was used for analysis of DA, AA and UA in their mixture was also performed when the concentration of one species remained constant at pretreated/CPE. From the Fig. 5.14A it can noticed that the concentration of DA
increased from $9.96 \times 10^{-6}$ to $2.38 \times 10^{-5}$ M when keeping the concentration of UA was $2.96 \times 10^{-5}$ M and AA was $4.9 \times 10^{-4}$ M. The anodic peak current was proportional to concentration of DA and there was no change in the peak current and peak potential occurred for AA and UA. The linear regression equation was 

$$I_{pa} (\mu A) = 0.0139 (C \mu M/L) + 0.269 (\mu A), \quad (r^2 = 0.995 \ n=7)$$

(Fig. 5.14B) the limit of detection (LOD) for DA in the lower concentration range was $1.4 \times 10^{-6}$ M for pretreated/CPE. The LOD was calculated according to the equation 

$$LOD = KS^0/S,$$

where $K$ was a constant related to the confidence level. In accordance with the suggestion of the IUPAC, the value of $K$ was 3 at the 99% confidence level. $S^0$ was the standard deviation of Six blank-solution measurements (no added DA), and $S$ was the slope of the calibration graph. The proposed electrode exhibited a relatively lower detection limit than those recently reported elsewhere [31-33] (Table 5.3). Similarly in the Fig. 5.15A showed the concentration effect of AA from $1 \times 10^{-5}$ to $1 \times 10^{-4}$ M at constant $9.96 \times 10^{-6}$ M DA and $2.96 \times 10^{-5}$ M UA. The anodic peak current was proportional to concentration of AA and there was no change in the peak current and peak potential occurred for DA and UA. The linear regression equation was 

$$I_{pa} (\mu A) = 0.0007 (C \mu M/L) + 0.4262 (\mu A), \quad (r^2 = 0.9961 \ n=7)$$

(Fig. 5.15B) the limit of detection (LOD) for AA in the lower concentration range was $5.41 \times 10^{-6}$ M for pretreated/CPE and Fig. 5.16A shows the concentration effect of UA from $4.99 \times 10^{-6}$ to $3.99 \times 10^{-5}$ M at constant $9.96 \times 10^{-6}$ M DA and $4.9 \times 10^{-4}$ M AA. The anodic peak current was proportional to concentration of UA and there was no change in the peak current and peak potential occurred for DA and AA. The linear regression equation was 

$$I_{pa} (\mu A) = 0.001 (C \mu M/L) + 0.0301 (\mu A), \quad (r^2 = 0.9873 \ n=5)$$

(Fig. 5.16B) the limit of detection (LOD) for UA in the lower concentration range was $4.3 \times 10^{-6}$ M for pretreated/CPE. These results showed that the DA, AA and UA existed independently in their sample mixture at pH.7.

5.6.9. Analytical applications

The pretreated/CPE was applied in the analysis of DA containing injection samples. The DA injection sample was purchased from sterile specialities India Private Ltd with a specified content of DA 40.0 mg/mL and the sample was used after suitable dilution. 0.2 M phosphate buffer was used for diluting the injection
samples. The results have been shown in Table 5.4. The recovery and R.S.D. was acceptable, showing that the proposed methods could be efficiently used for the detection of DA in injections with recovery in the range 97.02–99.02%.

5.6.10. Real Sample Analysis

Abnormal concentrations of DA in body fluids influenced the function of central nervous system. The pretreated/CPE was applied to the determination of DA content in healthy human blood serum samples applying the differential pulse voltammetric procedure. Using the proposed method, the dopamine concentration was analyzed in healthy human blood serum and the results obtained have been listed in Table 5.5. Procedure for real sample analysis has been: 1 ml of healthy human blood serum sample without any pretreatment was diluted to 100 ml with buffer and diluted solutions pipetted into each of series of 10 ml volumetric flasks. To this, different known standard concentration of DA solution was added and diluted to the mark with pH 7.0 phosphate buffer. The obtained results was shown in Table 5.5 and the recovery and relative standard deviation (RSD) was acceptable, which showed that the proposed methods could be efficiently used for the determination of DA in real blood serum sample with recovery in the range ~97.26 - 99.34% for DA analysis.

5.7. Conclusions

The Pretreated/Carbon paste electrode was fabricated and electrochemical parameters was studied. The Pretreated/Carbon paste electrode was prepared by 0.1 M NaOH in the potential window between 0.6 and 2 V at 5 Vs⁻¹ for 200 cycles exhibited stable enhanced electrocatalytic activity, sensitivity, selectivity, surface area of electrode, lower detection limit and higher linear range in 0.2 M phosphate buffer solution at pH 7.0 compared with bare CPE. The proposed method can be applied to the detection of DA both in injection samples and blood serum samples. Therefore, the present method can be extended to various bases to their modified electrode in the field of electroanalytical chemistry and electrochemical sensors.
Surface area of CPE for different concentration of NaOH at scan rate 5 Vs\(^{-1}\) for 200 cycles

<table>
<thead>
<tr>
<th>Concentration of NaOH (M)</th>
<th>Surface area in cm(^2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>0.030 ± 0.0011</td>
</tr>
<tr>
<td>0.1</td>
<td>0.0447 ± 0.0018</td>
</tr>
<tr>
<td>0.2</td>
<td>0.0421 ± 0.0025</td>
</tr>
<tr>
<td>0.3</td>
<td>0.0349 ± 0.0019</td>
</tr>
<tr>
<td>0.4</td>
<td>0.0366 ± 0.0009</td>
</tr>
<tr>
<td>0.5</td>
<td>0.0427 ± 0.0016</td>
</tr>
<tr>
<td>0.6</td>
<td>0.0422 ± 0.0032</td>
</tr>
<tr>
<td>0.7</td>
<td>0.0414 ± 0.0014</td>
</tr>
<tr>
<td>0.8</td>
<td>0.0426 ± 0.0023</td>
</tr>
<tr>
<td>0.9</td>
<td>0.0431 ± 0.0017</td>
</tr>
<tr>
<td>1.0</td>
<td>0.0433 ± 0.0012</td>
</tr>
</tbody>
</table>

Table 5.1a - Variation of surface area of CPE at different molarity of NaOH concentration.
Table 5.1b - Variation of surface area of CPE at different multiple cycles.

<table>
<thead>
<tr>
<th>Number of multiple cycles</th>
<th>Surface area in cm²</th>
</tr>
</thead>
<tbody>
<tr>
<td>50</td>
<td>0.0299 ± 0.0016</td>
</tr>
<tr>
<td>75</td>
<td>0.0312 ± 0.0024</td>
</tr>
<tr>
<td>100</td>
<td>0.0341 ± 0.0031</td>
</tr>
<tr>
<td>150</td>
<td>0.0363 ± 0.0013</td>
</tr>
<tr>
<td>200</td>
<td>0.0447 ± 0.0018</td>
</tr>
<tr>
<td>300</td>
<td>0.0336 ± 0.0017</td>
</tr>
</tbody>
</table>

Table 5.2 - Number of electron transferred for different scan rate in the range 0.05 to 0.2 Vs⁻¹.

<table>
<thead>
<tr>
<th>Scan Rate (Vs⁻¹)</th>
<th>Eₚₐ (mV)</th>
<th>Eₚ/2 (mV)</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>147.5</td>
<td>119</td>
<td>1.98</td>
</tr>
<tr>
<td>0.06</td>
<td>144</td>
<td>115</td>
<td>1.95</td>
</tr>
<tr>
<td>0.07</td>
<td>146</td>
<td>116</td>
<td>1.88</td>
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<tr>
<td>0.08</td>
<td>147.5</td>
<td>115.6</td>
<td>1.76</td>
</tr>
<tr>
<td>0.09</td>
<td>147.5</td>
<td>115.6</td>
<td>1.76</td>
</tr>
<tr>
<td>0.1</td>
<td>149</td>
<td>115.6</td>
<td>1.74</td>
</tr>
<tr>
<td>0.15</td>
<td>149</td>
<td>115.6</td>
<td>1.74</td>
</tr>
<tr>
<td>0.2</td>
<td>149</td>
<td>115.6</td>
<td>1.74</td>
</tr>
</tbody>
</table>
Chapter 5

Electrode | Sensitivity | Detection limit(µM) | Techniques | Reference |
---|---|---|---|---|
SDS micelles at pH 7/CPE | 106.58 ± 0.99 µA mM⁻¹ | 3.70 ± 0.01 | DPV | [31] |
CTAB/CPE | 7.390 ± 0.003 µAmM⁻¹ | 11 ± 0.2 | DPV | [32] |
SDS micelles as masking agent/CPE | 0.0481 ± 0.004 µA µM⁻¹ | 5.0 ± 0.5 | DPV | [33] |
Pretreated/CPE | 0.1390 ± 0.0008 µA µM⁻¹ | 1.4 ± 0.03 | DPV | This paper |

Table 5.3 - Comparison of the detection limit for different modified electrodes.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Content (mg/mL)</th>
<th>Found (mg/mL)</th>
<th>RSD (%)</th>
<th>Recovery (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.2</td>
<td>0.198 ± 0.0008</td>
<td>1.85</td>
<td>99.02</td>
</tr>
<tr>
<td>2</td>
<td>0.4</td>
<td>0.3938 ± 0.0107</td>
<td>2.12</td>
<td>98.45</td>
</tr>
<tr>
<td>3</td>
<td>0.6</td>
<td>0.5875 ± 0.0251</td>
<td>2.46</td>
<td>97.91</td>
</tr>
<tr>
<td>4</td>
<td>0.8</td>
<td>0.7803 ± 0.0047</td>
<td>1.69</td>
<td>97.54</td>
</tr>
<tr>
<td>5</td>
<td>1.0</td>
<td>0.9702 ± 0.0023</td>
<td>2.37</td>
<td>97.02</td>
</tr>
</tbody>
</table>

Table 5.4 - Detection of DA in injection samples (n=5).
Table 5.5 - Detection of DA in real blood serum samples (n=5).

<table>
<thead>
<tr>
<th>Sample</th>
<th>DA added (µM)</th>
<th>Found (µM)</th>
<th>Recovery (%)</th>
<th>RSD (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>10</td>
<td>9.934</td>
<td>99.34</td>
<td>1.37</td>
</tr>
<tr>
<td>2.</td>
<td>20</td>
<td>19.721</td>
<td>98.60</td>
<td>1.8</td>
</tr>
<tr>
<td>3.</td>
<td>30</td>
<td>29.18</td>
<td>97.26</td>
<td>1.52</td>
</tr>
</tbody>
</table>

Fig. 5.1 - Cyclic voltammograms of BCPE (solid line) and at Pretreated/CPE (dashed line) in the presence 1 mM potassium ferricyanide at scan rate of 0.1 Vs⁻¹.
Fig. 5.2 - Graph of anodic peak current ($I_{pa}$) vs peak potential ($E$) in 0.1M NaOH solution for multiple cycles.

Fig. 5.3 - Cyclic voltamogramms of 10 µM DA at BCPE (solid line) and pretreated/CPE (dashed line) in 0.2 M phosphate buffer solution at pH 7.0 and scan rate 0.05 V s$^{-1}$. 

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Fig. 5.4 - (A) cyclic voltammograms of $10 \times 10^{-6}$ M DA with different scan rates (0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.15, 0.2, 0.25, 0.3, 0.35, 0.4, 0.45, 0.5, 0.6, 0.7, 0.8, 0.9 and 1 Vs$^{-1}$) for the pretreated/CPE in PBS at pH 7.0. (B) Graph of the redox peak current versus the square root of the scan rate for DA in the range from 0.05 to 0.2 Vs$^{-1}$. (C) Graph of the redox peak current versus the scan rates for DA in the range from 0.2 to 1 Vs$^{-1}$.

Fig. 5.5 - Graph of DA potential difference two peaks versus PBS Solution and formal potential versus PBS Solution pH (5.8-7.8) at a Scan rate 0.05 Vs$^{-1}$ for pretreated/CPE.
Fig. 5.6 - Cyclic voltamograms of $4.9 \times 10^{-4}$ M AA at BCPE (solid line) and pretreated/CPE (dashed line) in 0.2 M phosphate buffer solution at pH 7.0 and scan rate 0.05 V s$^{-1}$.

Fig. 5.7 - (A) cyclic voltammograms of $4.9 \times 10^{-4}$ M AA with different scan rates (0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.15, 0.2, 0.25, 0.3, 0.35, 0.4, 0.45, 0.5, 0.6, 0.7, 0.8, 0.9 and 1 V s$^{-1}$) for the pretreated/CPE in PBS at pH 7.0. (B) Graph of the redox peak current versus the scan rates for AA in the range from 0.05 to 1 V s$^{-1}$. 
Fig. 5.8 - Cyclic voltamograms of 2.96×10⁻⁵ M UA at BCPE (solid line) and pretreated/CPE (dashed line) in 0.2 M phosphate buffer solution at pH 7.0 and scan rate 0.05 Vs⁻¹.

Fig. 5.9 - (A) cyclic voltammograms of 2.96×10⁻⁵ M UA with different scan rates (0.05, 0.06, 0.07, 0.08, 0.09, 0.1, 0.15, 0.2, 0.25, 0.3, 0.35, 0.4, 0.45, 0.5, 0.6, 0.7, 0.8, 0.9 and 1 Vs⁻¹) for the pretreated/CPE in PBS at pH 7.0. (B) Graph of the redox peak current versus the scan rates for UA in the range from 0.05 to 1 Vs⁻¹.
Fig. 5.10 - (A) Differential pulse voltammograms from $1 \times 10^{-7}$ to $1 \times 10^{-4}$ M DA individually in PBS at pH 7.0 for pretreated/CPE, (B) Graph of the anodic peak current versus the concentration of DA in the analyzed range $1 \times 10^{-7}$ - $1 \times 10^{-4}$ M and (C) Graph of the anodic peak current versus the concentration of DA in the better linearity range $2 \times 10^{-6}$ - $1 \times 10^{-5}$ M.

Fig. 5.11 - (A) Differential pulse voltammograms from $1 \times 10^{-6}$ to $1 \times 10^{-3}$ M AA individually in PBS at pH 7.0 for pretreated/CPE, (B) Graph of the anodic peak current versus the concentration of AA in the analyzed range $1 \times 10^{-6}$ M - $1 \times 10^{-3}$ M and (C) Graph of the anodic peak current versus the concentration of AA in the better linearity range $3 \times 10^{-6}$ - $2 \times 10^{-5}$ M.
Fig. 5.12 - (A) Differential pulse voltammograms from $1 \times 10^{-7}$ to $1 \times 10^{-4}$ M UA individually in PBS at pH 7.0 for pretreated/CPE, (B) Graph of the anodic peak current versus the concentration of UA in the analyzed range $1 \times 10^{-7}$ - $1 \times 10^{-4}$ M and (C) Graph of the anodic peak current versus the concentration of UA in the better linearity range $1 \times 10^{-5}$ - $9 \times 10^{-5}$ M.

Fig. 5.13 - Cyclic voltammograms for $9.96 \times 10^{-6}$ M DA, $4.9 \times 10^{-4}$ M AA and $2.96 \times 10^{-5}$ M UA at pH 7.0 PBS at a scan rate of 0.05V s$^{-1}$ in BCPE (Solid line) and pretreated/CPE (dotted line).
Fig. 5.14 - (A) Differential pulse voltammograms from $9.96 \times 10^{-6}$ to $2.38 \times 10^{-5}$ M, DA in 0.2M phosphate buffer solution at pH 7.0 in the presence of $2.96 \times 10^{-5}$ M UA and $4.9 \times 10^{-4}$ M AA at the pretreated/CPE. (B) Graph of the anodic peak current versus the concentration of DA in the presence of AA and UA.

Fig. 5.15 - (A) Differential pulse voltammograms from $1 \times 10^{-5}$ to $1 \times 10^{-4}$ M, AA in 0.2M phosphate buffer solution at pH 7.0 in the presence of $2.96 \times 10^{-5}$ M UA and $9.96 \times 10^{-6}$ M DA at the pretreated/CPE. (B) Graph of the anodic peak current versus the concentration of AA in the presence of DA and UA.
Fig. 5.16 - (A) Differential pulse voltammograms from $4.99 \times 10^{-6}$ to $3.99 \times 10^{-5}$ M, UA in 0.2 M phosphate buffer solution at pH 7.0 in the presence of $4.9 \times 10^{-4}$ M AA and $9.96 \times 10^{-6}$ M DA at the pretreated/CPE. (B) Graph of the anodic peak current versus the concentration of UA in the presence of DA and AA.
5.8. References


