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Original scientific paper

Voltammetric determination of dopamine in the presence of uric acid using a 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)--naphthalin-4-sulfonic acid modified glassy carbon electrode

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Abstract: A polymerized film of 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid (HHNANSA) was prepared at the surface of a glassy carbon electrode by electropolymerization. The modified electrode was used for the simultaneous determination of dopamine (DA) and uric acid (UA). The electrochemical behaviors of the compounds at the surface of the modified electrode were studied using cyclic voltammetry, chronoamperometry, and square wave voltammetry (SWV). The experimental results indicated that the modified electrode exhibited an efficient electrocatalytic activity towards the oxidation of DA and UA, with a peak separation of about 140 mV at pH 5.0. Using chronoamperometry, the catalytic reaction rate constant was measured and found to equal to 1.23×10^4 mol^{-1} L s^{-1}. At pH 5.0, the catalytic peak currents linearly depended on the DA and/or UA concentrations in the range of 1.0–300 μmol L^{-1} DA (two linear segments with different slopes) and 6.7–20 μmol L^{-1} UA, using SWV. The detection limits for DA and UA were 0.25 and 1.17 μmol L^{-1}, respectively. The *RSD* % for 40.0 and 140.0 μmol L^{-1} DA were 1.9 and 2.2 %, respectively, whereas for 10.0 and 20.0 μmol L^{-1} UA, they were 1.8 and 1.2 %, respectively. The modified electrode showed good sensitivity, selectivity, and stability. It was successfully applied for the determination of DA and UA in real samples, such as drugs and urine.

Keywords: 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid; simultaneous determination; dopamine and uric acid; voltammetry.

INTRODUCTION

Dihydroxyphenylethylamine, also commonly known as dopamine (DA), is one of the most significant catecholamines and is an important neurotransmitter in the mammalian central nervous system. It is currently the subject of intensive research by neuroscientists and chemists and rapid and simple methods for the

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determination of its concentration are sought.[1–3] It has also been suggested that DA plays a role in drug addiction and some manifestations of HIV.[4–6] Dopamine is not just a precursor of norepinephrine (noradrenaline) and epinephrine (adrenaline) but a neurotransmitter as well. Therefore, the determination of this compound is very necessary.

Uric acid (2,6,8-trihydroxypurine, UA) is a compound of great biomedical interests and has important roles in human metabolism. UA, the end metabolic product of purine[7] *via* the liver, is present in blood and urine. Monitoring UA in blood or urine can be used as an indicator for an early warning sign of kidney diseases. Abnormal UA level in a human body could be caused by several diseases, such as hyperuricemia, gout, cardiovascular disorder, Lesch–Nyan syndrome, and chronic renal disease. DA and UA are co-present in biological fluids, such as urine and blood. Therefore, it is important to develop new techniques for the selective detection of DA in the presence of UA in a routine assay. The direct electro-oxidation of DA and UA at bare electrodes requires high overpotentials. In addition, they have overlapped signals because the oxidation peaks of DA and UA at a bare electrode are at nearly the same potential, making their discrimination very difficult.[8,9]

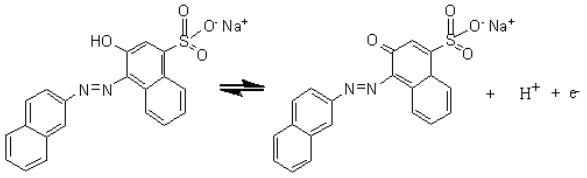
Several papers have reported using modified electrodes for the simultaneous determination of DA and UA.[10–17,21] Recently, polymer film modified electrodes have been used as electrochemical sensors in the wide application fields of chemical sensors and biosensors.[18–21] Comparisons of the proposed method with the similar electrochemical methods are presented in Table I. Although, previous studied based on poly(3-(5-chloro-2-hydroxyphenylazo)-4,5-dihydroxynaphthalene-2,7-disulfonic acid[17] and sulfonazo III film[21] modified electrodes have better limits of detection, the proposed modified electrode based on 2-hydroxy-1-(1-

TABLE I. A comparison of the efficiency of some modified electrodes in the determination of DA and UA

Sensitivity μA μmol^{-1} L		Limit of detection μmol L^{-1}		Linear dynamic range μmol L^{-1}		Ref.
DA	UA	DA	UA	DA	UA	
0.3316	0.0929	0.075	0.021	0.2–45.8	0.06–166.0	10
1.0728	0.0910	0.02	1.0	0.1–200	10.0–130.0	11
Not reported	17.00	0.0027	0.2	0.0–6.0	0.5–100.0	12
Not reported	Not reported	1.0	1.0	2.0–1500	2.0–220	13
1.0	Not reported	0.5	Not reported	2.0–10.0	Not reported	14
1.741	0.7360	0.12	0.6	0.2–80.8	1.2–100.0	15
Not reported	Not reported	Not reported	5.0	Not reported	5.0–53.0	16
0.0808	0.1013	0.03	0.11	0.05–470	0.2–100	17
0.0572	0.3533	0.29	0.016	5.0–280	0.1–18.0	21
1.5659	1.1707	0.25	1.17	1.0–20	6.7–20	This work
0.0856	–	–	–	20–300	–	This work

-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid has a longer linear dynamic range and is free from interference by aspirin.

To the best of our knowledge, no work has been reported on the use of 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid (HHNANSA, Scheme 1) modified glassy carbon electrode (HHNANSA–GCE) for the electrocatalytic determination of DA and UA. Consequently, in this study, a GCE was modified with a HHNANSA polymer film and then the electrochemical behavior of DA and UA at the surface of the modified electrode was studied. Using the modified electrode, a sensitive and selective method was established for the simultaneous determination of DA and UA.



Scheme 1. Structure of 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid.

EXPERIMENTAL

Chemicals

All chemicals were of analytical reagent grade and used without further purification. Doubly distilled water was used throughout the experiments. Uric acid was purchased from Sigma-Aldrich. Dopamine was obtained from Merck.

Phosphate buffer solutions (PBS) with different pH values were prepared by mixing 0.10 mol L^{-1} Na_2HPO_4 and 0.10 mol L^{-1} NaH_2PO_4. The pH values were adjusted by addition of 1.0 mol L^{-1} H_3PO_4 and/or NaOH solution.

A stock solution of dopamine (0.010 mol L^{-1}) was prepared daily by dissolving dopamine in water. Uric acid solution (0.010 mol L^{-1}) was prepared by dissolving the solid in a small volume of 0.1 mol L^{-1} NaOH solution and diluted with water to obtain the desired concentration. Other dilute standard solutions were prepared by appropriate dilution of the stock solutions in PBS, pH 5.0.

Apparatus

A Corning pH-meter, Model 140, with a glass electrode (conjugated with an Ag/AgCl reference electrode, Model 6.0232.100) was used to determine the pH of the solutions.

Voltammograms were obtained using an EG & G instrument, Model 384B processor, with three electrodes consisting of a platinum wire as the auxiliary electrode, a HHNANSA––GCE as the working electrode and Ag/AgCl (3 mol L^{-1} KCl) as the reference electrode.

Preparation of the poly-2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid film-modified GC electrode

Prior to each experiment, the glassy carbon electrode (GCE) was polished with 0.05 μm alumina in a water surrey using a polishing cloth. The GCE was subsequently sonicated in a mixture of water/ethanol (90/10 v/v) after each polishing step to be electrochemically pretreated later by cycling at a scan rate of 100 mV s^{-1} 10 times in 0.1 mol L^{-1} H_2SO_4 solution in the potential range of –0.40 to 1.50 V, to obtain a stable background current. Subsequently,

the electrode was placed in a solution containing 0.2 mol L^{-1} NaOH and 10.0 mmol L^{-1} 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid (HHNANSA) and a cyclic potential sweep was applied in the range of –0.10 to 1.00 at a scan rate of 33 mV s^{-1} for 30 times (the anodic peak potential and current tended to be stable after 25 scans). After the polymerization, the modified electrode was washed with water and scan cycled 5 times at pH 5.0 (PBS) between 0.00 and 1.00 V to eliminate untreated HHNANSA monomer, and thus increase its reproducibility and stability.

Procedure

Five milliliters of the buffer solution (pH 5.0) were transferred into the electrochemical cell using the three-electrode system. Then, the SW voltammogram was recorded from 0.00 to 1.00 V at a frequency of 100 Hz and a pulse height of 20 mV with a potential scan rate of 33 mV s^{-1}. The peak current was measured and recorded as the blank signal (I_{pb}). After the background voltammogram had been obtained, aliquots of the sample solution containing DA, and/or UA were introduced into the cell. Then, the SW voltammogram was recorded as described above to give the sample peak current. The peak current was measured and recorded as the sample signal (I_{ps}). All the data were obtained at room temperature. The difference in the currents ($I_{ps} - I_{pb}$) was considered as a net signal (ΔI_p) for each of the experiments. Calibration graphs were prepared by plotting the net peak currents *vs.* the DA and/or UA concentrations in the solution.

Real sample preparation

Dopamine hydrochloride injection solution (40 mg mL^{-1}) was analyzed directly after diluting 100 times with water. 0.10 mL of the diluted solution was injected into a 10-mL volumetric flask and made up to volume with buffer solution (pH 5.0). The test solution was then transferred into the electrochemical cell and the DA content was measured according to the recommended procedure.

Urine samples were analyzed directly after diluting 25 times with buffer solution (pH 5.0) without any further pretreatment. Then, 5 mL of this test solution was transferred into the electrochemical cell and the DA and UA contents were determined according to the recommended procedure.

RESULTS AND DISCUSSION

Electrochemical properties of the poly (2-hydroxy-1-(1-hydroxynaphthyl-2-azo)--naphthalin-4-sulfonic acid) film-modified GCE

The behavior of poly(2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4--sulfonic acid) (PHHNANSA) at the surface of a GCE is shown in Fig. 1. As can be seen in the cyclic voltammograms, an anodic peak appeared at about 0.10 V due to the oxidation of HHNANSA monomer with cycles, whereas on the reversed scan, a cathodic wave formed at a potential of –0.10 V. These anodic and cathodic peak potentials tended to be stable after 25 scans. This suggest that the initially-formed PHHNANSA film underwent a leaching process during the scan cycles up to 25 cycles, which may imply a self-adjustment of the polymer film thickness at the GCE.

The electrochemical properties of the modified electrode were studied by cyclic voltammetry (CV) in the buffer solution (pH 5.0). The CVs of the mo-

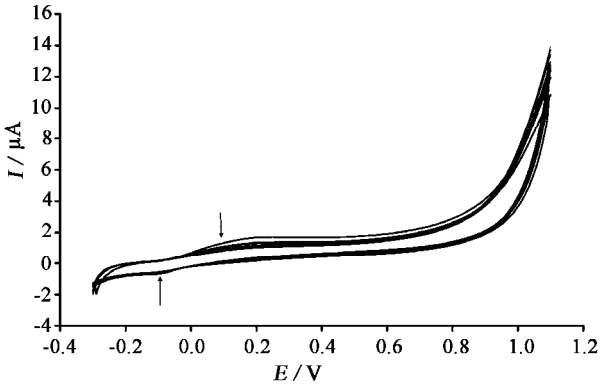


Fig. 1. Electropolymerization graph of HHNANSA in 0.2 mol L^{-1} NaOH and 10 mmol L^{-1} HHNANSA at a scan rate of 100 mV s^{-1}.

dified electrode at various scan rates (10–100 mV s^{-1}) are shown in the inset to Fig. 2. The experimental results show well-defined and reproducible anodic and cathodic peaks, with a peak separation potential of about ΔE_p ($E_{pa} - E_{pc}$) = 200 mV. These CVs were used to examine the variation of the peak currents *vs.* potential scan rates. The plot of the anodic peak currents was linearly dependent on υ with a correlation coefficient of 0.9966 at all scan rates (Fig. 2A). Therefore, the peak current must be related to the surface concentration of electroactive species, Γ, by:

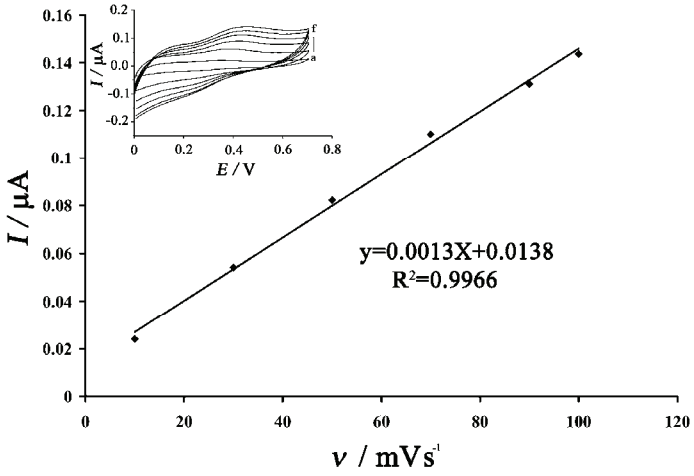
$$I_P = n^2F^2A\Gamma v/4RT \quad (1)$$

where n represents the number of electrons involved in the reaction (n = 1), A is the surface area of the electrode (0.031 cm^2), I_P is the peak current, Γ represents the surface coverage concentration (mol cm^{-2}), and v is the scan rate. From the slope of the anodic peak currents *vs.* the scan rate, the calculated surface concentration of 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid was 4.46×10^{-8} mol cm^{-2}. The cyclic voltammograms of the bare GCE in PBS (pH 5.0) at various scan rates (between 10 to 110 mV s^{-1}) are shown in Fig. 2B.

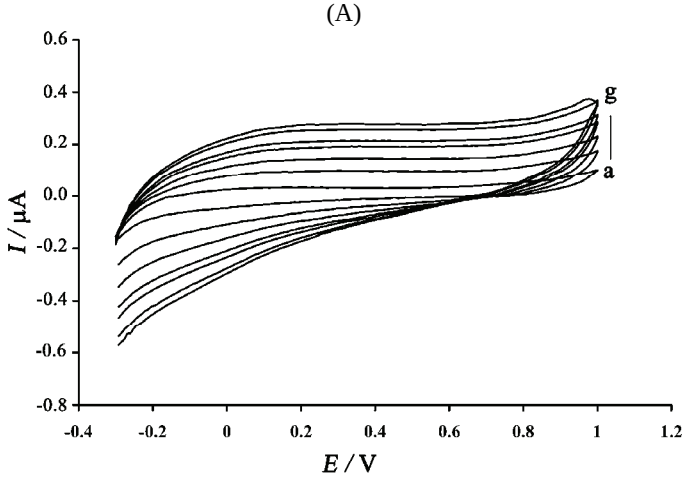
The electrochemical response of the HHNANSA–GCE depended on the pH value of the supporting electrolyte solution. By increasing the pH of the supporting electrolyte (from 2.0 to 7.0), the redox and oxidation peak potentials shifted negatively and the anodic peak potential (E_{pa}) depended linearly on the pH value.

Electro-oxidation of DA and UA at the surface of the HHNANSA–GCE

The oxidations of DA and UA at the surface of a bare GCE and HHNANSA––GCE in two different concentrations are shown in Fig. 3. The results showed that both compounds were oxidized with well-defined and distinguishable sharp peaks potential at the HHNANSA–GCE. On the other hand, the indistinguishable



(A)



(B)

Fig. 2. A) Dependence of peak current on the scan rate for the poly(HHNANSA)-modified GCE in PBS (pH 5.0). Inset: cyclic voltammograms of the poly(HHNANSA)-modified GCE in PBS (pH 5.0) at various scan rates: a) 10; b) 30; c) 50; d) 70; e) 90; f) 100 mV s^{-1}; B) Cyclic voltammograms of the bare GCE in PBS (pH 5.0) at various scan rates: a) 10; b) 30; c) 50; d) 70; e) 80; f) 100; g) 110 mV s^{-1}.

and broad peak potentials at the bare GCE indicate slow kinetics of electron transfer. The oxidation peak potentials of DA and UA at the modified electrode separated completely into two well-defined peaks at 0.32 and 0.46 V *vs.* Ag/AgCl, respectively, whereas at the bare GCE, the oxidation peak potentials were at about 0.40 and 0.50 V for DA and UA, respectively at pH 5.0. In addition, both the two peaks potential at the HHNANSA–GCE exhibited negative potential shifts. These shifts in the oxidation peaks potential and enhanced currents of the

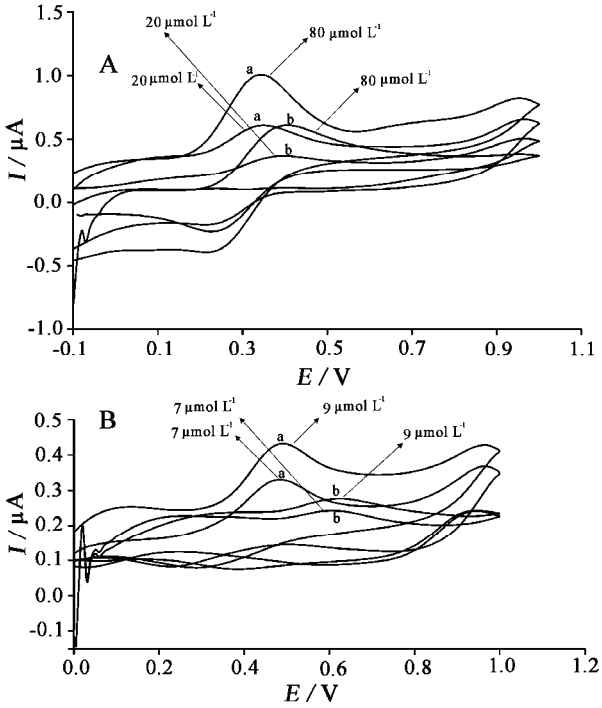


Fig. 3. Cyclic voltammograms of: A) 20.0 and 80.0 μmol L^{-1} DA; B) 7.0 and 9.0 μmol L^{-1} UA at the surface of a) the modified and b) a bare glassy carbon electrode.

oxidation peaks potential with the HHNANSA–GCE indicate that the modified electrode had a catalytic effect on the oxidation of DA and UA. Moreover, the oxidation current of DA increased linearly with the square root of the scan rate, which demonstrates a diffusion controlled electrochemical process (Fig. 4).

The influence of solution pH on the DA and UA peaks current were studied with 50.0 μmol L^{-1} DA and 50.0 μmol L^{-1} UA. The results showed that the UA peak current increased when the solution pH increased from 2.0 to 5.0, whereas at higher pH values, the peak current decreased. For DA, the peak current increased sharply on increasing the pH to 5.0, then leveled off up to pH 7.0 and then decreased at higher pH values. As is known, the pK_a values of R–SO_3H (R = aryl group) are usually about 4; therefore, the –SO_3Na of the poly(2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid) film could dissociate favorably into the negatively charged –SO_3^- group under alkaline conditions. The alkaline –NH_2 group of DA (pK_a 8.9) could obtain a proton and form positive DA ions. These have a great affinity toward the poly(2-hydroxy-1--(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid) film.

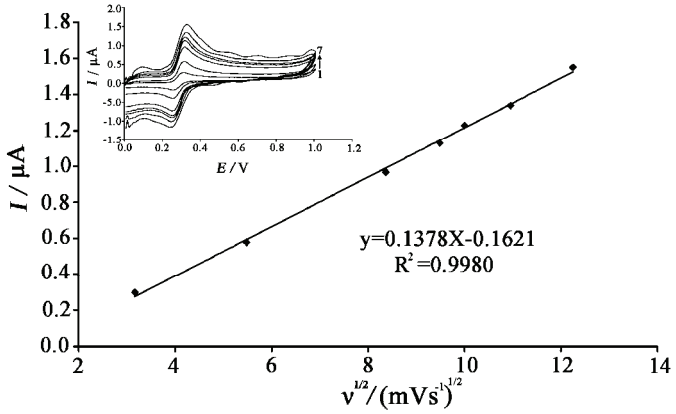


Fig. 4. Variation of electrocatalytic current (I_p) with square scan rate. Inset: cyclic voltammograms of poly(HHNANSA) modified GCE in PBS (pH 5.0) containing 100.0 μmol L^{-1} DA at different scan rates. The number of 1 to 7 corresponds to 10, 30, 70, 90, 100, 120 and 150 mV s^{-1}, respectively.

Chornoamperometric studies

For the determination of the diffusion coefficient of DA, single potential step chronoamperometry was used with the HHNANSA–GCE. The current–time curves of the HHNANSA–GCE obtained by setting the electrode potential at 270 mV (*vs.* Ag|AgCl|KCl_{sat}) for different concentrations of DA are shown in Fig. 5. The linearity of the electrocatalytic current *vs.* $v^{1/2}$ showed that the current is controlled by diffusion of DA from the bulk solution toward the surface of the

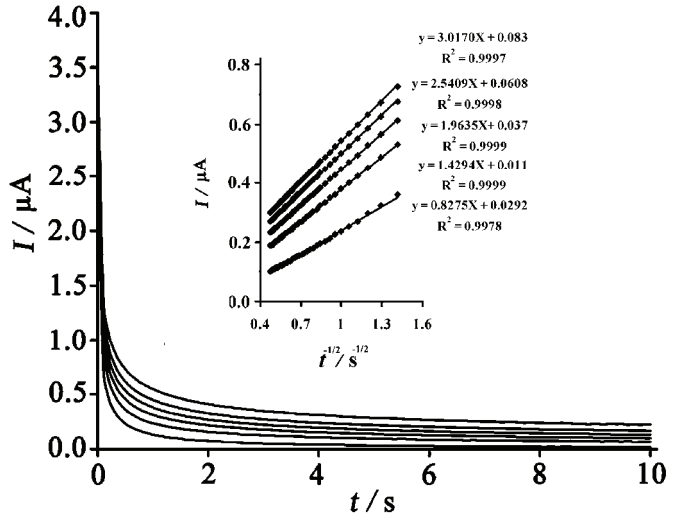


Fig. 5. Chronoamperograms of the poly(HHNANSA) modified GCE in PBS (pH 5.0) in the presence of (in direction of *I*-axis): 0.0; 60.0; 100.0; 160.0; 260.0 and 300.0 μmol L^{-1} of DA. Inset shows Cottrell plots derived from the chronoamperometric data.

electrode, which caused near Cottrellian behavior. The slope of the linear region of a Cottrell plot can be used to estimate the diffusion coefficients. A plot of I *vs.* $t^{-1/2}$ at the HHNANSA–GCE in the presence of DA gave a straight line, the slope of which can be used to estimate the diffusion coefficient (D) of DA in the range of 60 to 300 μmol L^{-1}. The mean value of the D for DA was found to be 1.30×10^{-5} cm^2 s^{-1} (Fig. 5, inset A). This value is different to the previously obtained value of 6.40×10^{-6} cm^2 s^{-1}).[17] This is due to difference in the solution pH, which affects the net charge of DA in the solutions and hence affects the diffusion coefficient.

In addition, chronoamperometry can be performed to evaluate the rate of an electrocatalyzed oxidation. The rate constant for the chemical reaction between DA and redox cites of 2-hydroxy-1-(1-hydroxynaphthyl-2-azo)-naphthalin-4-sulfonic acid can be evaluated according to the method of Galus:[22]

$$I_C/I_L = \gamma^{1/2}(\pi^{1/2} \text{ erf } (\gamma^{1/2}) + \exp(-\gamma)/\gamma^{1/2}) \quad (2)$$

where I_C is the catalytic current of the HHNANSA–GCE in the presence of DA, I_L is the limited current in the absence of DA and $\gamma = k_h C_b t$ (C_b is the bulk concentration of DA, mol L^{-1}) is the argument of the error function. In cases where γ exceeds 2, the error function is almost equal to 1 and Eq. (2) can be reduced to:

$$I_C/I_L = \pi^{1/2}\gamma^{1/2} = \pi^{1/2} (k_h C_b t)^{1/2} \quad (3)$$

where k_h and t are the catalytic rate constant (mol^{-1} L s^{-1}) and time elapsed, respectively. Equation (3) can be used to calculate the rate constant of the catalytic process, k_h. From the slope of I_C/I_L *vs.* $t^{1/2}$, the value of k_h can be simply calculated for a given concentration of the substrate. The calculated value of k_h was equal to 1.23×10^4 mol^{-1} L s^{-1}. This value of k_h explains the sharp feature of the catalytic peak observed for the catalytic oxidation of DA at HHNANSA–GCE. These methods have already been used for an estimation of D and k_h for some compounds.[23–26]

Simultaneous determination of DA and UA

As the obtained results showed, the HHNANSA–GCE possessed excellent electrocatalytic activity for the oxidation of DA and UA. The difference in the oxidation peak potentials for DA–UA was 140 mV, which is large enough to allow for the simultaneous determination of DA and UA in a mixture. The electro-oxidation processes of DA and UA in the mixture were evaluated by varying the concentration of the individual analytic species. The result showed two linear segments with different slopes for the DA concentration; namely, for 1.0–20.0 μmol L^{-1} DA with a regression equation of I_p (μA) = (1.566±0.050)c_{DA} – – (0.4211±0.0080) (r^2 = 0.9944, n = 8) (Fig. 6A), while for 20.0–300.0 μmol L^{-1} DA, the regression equation was I_p (μA) = (0.0856±0.0050)c_{DA} + (31.019±

±0.090) (r^2 = 0.9897, n = 7) (Fig. 6B). In addition, for 6.7–20.0 μmol L^{-1} UA, the regression equation was I_p (μA) = (1.1707±0.05021)c_{UA} – (3.3652±0.07010) (r^2 = 0.9920, n = 5) (Fig. 6C), where c is the concentration of the substance (μmol L^{-1}) and I_p is the net peak current (the sample peak current minus the blank peak current).

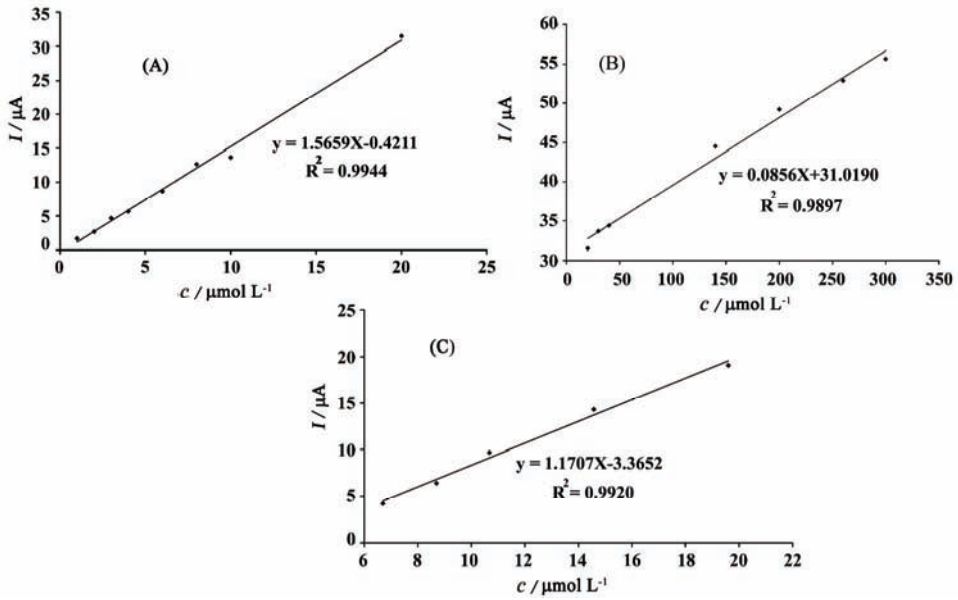


Fig. 6. A) Plot of the oxidation current *vs.* the concentration of DA in the range of 1.0–20.0 μmol L^{-1}; B) as a (A), in the range of 20–300 μmol L^{-1} using SWV; C) calibration curve for UA in the range of 6.7–20.0 μmol L^{-1} using SWV.

The detection limits were determined at 0.25 and 1.17 μmol L^{-1} for DA and UA, respectively, according to the definition of $Y_{LOD} = Y_B + 3\sigma$.[27]

In order to check for the presence of any intermolecular effects between DA and UA, two different experiments were performed under the optimum conditions at pH 5.0. In each experiment, the concentration of one of the two compounds was changed while the concentration of the other compound was kept constant. The results are shown in Figs. 7A and 7B. The results showed no any intermolecular interactions during the oxidation of the compounds at the surface of the HHNANSA–GCE.

The sensitivities towards DA in the absence and presence of UA were found to be 0.0856±0.005 μA μmol L^{-1} (in the absence of UA) and 0.0960±0.006 μA μmol L^{-1} (in the presence of UA) (Fig. 8A). However, for UA, the obtained sensitivities were 1.1707±0.0901 (in the absence of DA) and 1.1691±0.0240 μA μmol L^{-1} (in the presence of DA) (Fig. 8B). It is interesting to note that the

sensitivities of the HHNANSA–GCE to DA and UA in the absence and presence of the other compound were very similar, which indicates that the oxidation processes of DA and UA at the HHNANSA-GCE were independent. Therefore, the simultaneous or independent measurements of the two analytes are possible without any interference.

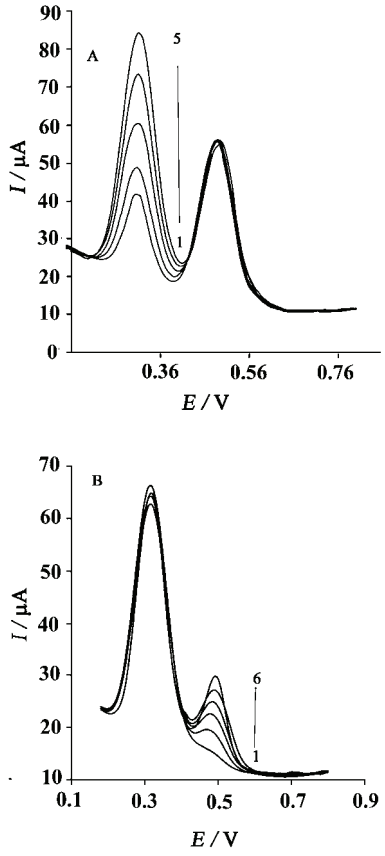


Fig. 7. A) SWV of: 1) 20.0; 2) 50.0; 3) 110.0; 4) 160.0; 5) 220.0 μmol L^{-1} DA in the presence of 60.0 μmol L^{-1} UA; B) SWV of: 1) 8.0; 2) 10.0; 3) 12.0; 4) 15.0; 5) 17.0; 6) 19.0 μmol L^{-1} UA in the presence of 100.0 μmol L^{-1} DA.

Interference study

The influence of various substances as compounds that could potentially interfere with the determination of DA and UA were studied under the optimum conditions with 100.0 μmol L^{-1} DA and 14.0 μmol L^{-1} UA at pH 5.0. The potentially interfering substances were chosen from substances commonly found with DA and UA in pharmaceuticals and/or in biological fluids. The tolerance limit was defined as the maximum concentration of the interfering substance that caused an error of less than ±5 % for the determination of DA and UA. The results are presented in Table II. These results showed the high selectivity of the HHNANSA–GCE for the determination of DA and UA.

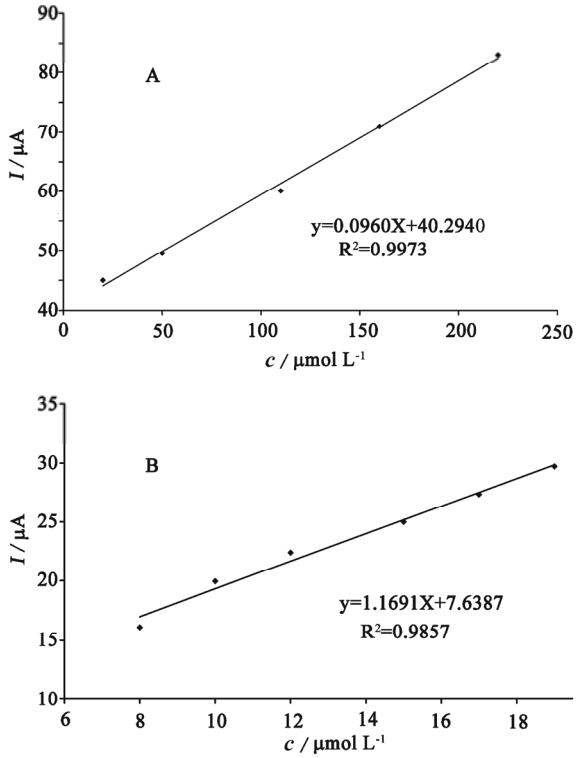


Fig. 8. A) Plot of different concentrations of DA in the presence of a fixed amount of UA; B) Plot of different concentrations of UA in the presence of a fixed amount of DA.

Table II. Interference of some foreign substances on the determination of 100.0 μmol L^{-1} DA and 14.0 μmol L^{-1} UA under the optimized conditions

Species	*w*(substance)/*w*(DA)	*w*(substance)/*w*(UA)
Glucose, sucrose, urea, fructose	400	1000
Citric acid	100	1000
Ca^{2+}, Mg^{2+}, F^{-}, Cl^{-}, Na^{+}, K^{+}, SO_4^{2-}	100	800
Carbonate	200	600
Tryosin, aspirin	20	100
Histidine, cysteine, ascorbic acid	2	100

Real sample analysis

In order to evaluate the applicability of the proposed method for the determination of DA and UA in real samples, the utility of the developed method was tested by analysis of these compounds in mixed synthetic and in real samples using standard addition methods. The results are summarized in Table III. The good recoveries of the mixture samples indicate the successful application of the proposed method for the simultaneous determination of DA and UA. For further

investigation, the recovery of DA was determined for dopamine injection. The dopamine injection solution (specified content of DA was 40.0 mg mL^{-1}) was diluted to 100 mL with water, then a different amount of the diluted solution was transferred into each of a series of 10-mL volumetric flasks and diluted to the mark with phosphate buffer. Then, 10 mL aliquot of this test solution was placed in the electrochemical cell and the DA content was measured by the proposed method. This procedure was repeated five times and the relative standard deviation was found as 1.6 %. Different standard concentrations of DA were added to the diluted DA injection solution, with recoveries between 96.5 and 103.2 % for five measurements (Table III).

Table III. Determination of DA and UA in human urine and DA injection samples

Sample	Added, μmol L^{-1}		Found, μmol L^{-1}		Recovery, %	
	DA	UA	DA	UA	DA	UA
1	20.0	10.0	20.9±0.8	9.9±0.2	104.6	99.0
2	50.0	14.0	51.6±0.6	13.4±0.5	103.1	96.1
3	70.0	20.0	67.9±0.7	19.4±0.4	97.1	97.1
DA injection						
4	10.0	–	9.6±0.5	–	96.5	–
5	15.0	–	15.1±0.7	–	100.8	–
6	50.0	–	51.6±0.6	–	103.2	–

CONCLUSIONS

In this study, the electrochemical behavior of DA and UA at a glassy carbon electrode modified with a polymerized film of 2-hydroxy-1-(1-hydroxynaphthyl--2-azo)-naphthalin-4-sulfonic acid was investigated using cyclic voltammetry and square wave voltammetry. The modified electrode separated the anodic oxidation peak potential of DA and UA with a well-defined peak separation in the presence of each other to measure DA and/or UA separately or simultaneously without any intermolecular effects. The catalytic reaction rate constant, k_h was calculated (1.23×10^4 mol^{-1} L s^{-1}) using chronoamperometry. The sensitivity of the proposed material was higher than those of reported results.[10,11,14,17,21] Moreover, the proposed method was successfully applied for the determination of these compounds in real samples.

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ИЗВОД

ВОЛТАМЕТРИЈСКО ОДРЕЂИВАЊЕ ДОПАМИНА У ПРИСУСТВУ МОКРАЋНЕ КИСЕЛИНЕ НА ЕЛЕКТРОДИ ОД СТАКЛАСТОГ УГЉЕНИКА МОДИФИКОВАНОГ 2-ХИДРОКСИ-1(1-ХИДРОКСИ НАФТИЛ-2-АЗО)-НАФТАЛИН-4-СУЛФОНСКОМ КИСЕЛИНОМ

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Полимерни филм 2-хидрокси-(1-хидроксинафтил-2-азо)-нафталин-4-сулфонске киселине је формиран на електроди од стакластог угљеника поступком електрохемијске полимеризације. Тако модификована електрода је коришћена за истовремено одређивање допамина (ДА) и мокраћне киселине (МК). Електрохемијско понашање ових једињења на модификованој електроди је испитивано цикличном волтаметријом, хроноамперометријом и волтаметријом са правоугаоним сигналом. Експериментални резултати показују да модификована електрода представља ефикасан катализатор за оксидацију ДА и МК уз сепарацију пикова од око 140 mV при pH 5,0. Константа брзине катализоване реакције одређена је методом хроноамперометрије и износи $1{,}23\times10^4$ mol^{-1} L s^{-1}. У мерењима волтаметријом са правоугаоним сигналом у раствору pH 5,0 струјни пикови су показали линеарну зависност од концентрације ДА и/или МК у опсегу 1,0–300 μmol L^{-1} ДА (две линеарне области са различитим нагибима) и 6,7–20 μmol L^{-1} МК. Границе детекције за ДА и МК су биле 0,25 и 1,17 μmol L^{-1}, редом. Стандардна девијација за 140,0 μmol L^{-1} ДА је износила 1,9 и 2,2%, а за 10,0 и 20,0 μmol L^{-1} МК 1,8 и 1,2%, редом. Модификована електрода је показала високу осетљивост, селективност и стабилност. Такође је успешно примењена и за одређивање ДА и МК у реалним узорцима као што су лекови и урин.

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REFERENCES

1. N. Q. Jia, Z. Y. Wang, G. F. Yang, H. B. Shen, L. Z. Zhu, *Electrochem Commun*. **9** (2007) 233
2. J. W. Mo, B. Ogorevc, *Anal. Chem.* **73** (2001) 1196
3. R. M. Wightman, C. Amatorh, R. C. Engstrom, P. D. Hale, E. W. Kristensen, W. G. Kuhr, L. J. May, *Neuroscience* **25** (1998) 513
4. R. M. Wightman, L. J. May, A. C. Michael, *Anal. Chem.* **60** (1988) 769
5. A. Heinz, H. Przuntek, G. Winterer, A. Pietzcker, *Nervenarzt* **66** (1995) 662
6. F. M. Benes, *Trends Pharmacol. Sci.* **22** (2001) 46.
7. J. K. Baillie, M. G. Bates, A. A. Thompson, W. S. Waring, R. W. Partridge, M. F. Schnopp, A. Simpson, F. Gulliver-Sloan, S. R. Maxwell, D. J. Webb, *Chest* **131** (2007) 1473
8. Y. Y. Sun, K. B. Wu, S. S. Hu, *Chem. J. Chin. Universities* **23** (2002) 2067
9. M. Mazloum-Ardakani, H. Beitollahi, B. Ganjipour, H. Naeimi, M. Nejati, *Bioelectrochem.* **75** (2009) 1
10. A. A. Ensafi, M. Taei, T. Khayamian, *Int. J. Electrochem. Sci.* **5** (2010) 116
11. H. Yao, Y. Sun, X. Lin, Y. Tang, L. Huang, *Electrochim. Acta* **52** (2007) 6165
12. J. M. Zen, P. J. Chen, *Anal. Chem.* **69** (1997) 5087
13. A. Safavi, N. Maleki, O. Moradlou, F. Tajabadi, *Anal. Biochem*. **359** (2006) 224
14. A. Balamurugan, S. M. Chen, *Anal. Chim. Acta* **596** (2007) 92
15. P. Wang, Y. Li, X. Huang, L. Wang, *Talanta* **73** (2007) 431

16. P. Ramesh, S. Sampath, *Electroanalysis* 16 (2004) 866
17. A. A. Ensafi, M. Taei, T. Khayamian, A. Arabzadeh, *Sens. Actuators B* **147** (2010) 213
18. J. B. Raoof , R. Ojani, S. Rashid-Nadimi, *Electrochim. Acta* **50** (2005) 4694
19. H. Zhao, Y. Zhang, Y. Yuan, *Analyst* **126** (2001) 358
20. T. Selvaraju, R. Ramaraj, *Electrochem. Comm.* **5** (2003) 667
21. A. A. Ensafi, M. Taei, T. Khayamian, *J. Electroanal. Chem.* **633** (2009) 212
22. Z. Galus, *Fundumentals of Electrochemical Analysis*, Ellis Horwood, New York, 1999
23. E. Mirmomtaz, A. A. Ensafi, H. Karimi-Maleh, *Electroanalysis* **20** (2008) 1973
24. H. Karimi-Maleh, A. A. Ensafi, H. R. Ensafi, *J. Braz. Chem. Soc.* **20** (2009) 880
25. H. Karimi-Maleh, A. A. Ensafi. A. R Allafchian, *J. Solid State Electrochem.* **14** (2010) 9
26. H. Yghoubian, H. Karimi-Maleh, M. A. Khalilzadeh, F. Karimi, *J. Serb. Chem. Soc.* **74** (2009) 1443
27. J. N. Miller, J. C. Miller, *Statistics and Chemometrics for Analytical Chemistry*, 4th ed., Pearson Education Ltd., Harlow, Essex, 2000.