



Electrochemical Detection of Epinephrine in The Presence of Ascorbic Acid at Poly (*p*-Aminobenzene Sulfonic Acid) Modified Sensor

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Abstract. In this study, a rapid, reliable, selective and sensitive simultaneous voltammetric determination of Epinephrine (EP) in the presence of ascorbic acid at poly (*p*-aminobenzene sulphonic acid, ABSA) modified sensor was aimed. The glassy carbon electrode was successfully modified with ABSA in 0.1 M KCl solution by the cycling voltammetry technique. The sensor, modified with a polymeric thin film, showed excellent electrocatalytic activity against the oxidation of EP and ascorbic acid (AA). The results showed that the oxidation potential and current responses of EP and AA improved significantly. The modified sensor showed excellent response with limit of detection as 50 nM in the determination of EP at the 5.0 - 53.2 µM concentration range under optimum conditions. In real sample analyzes performed in pharmaceutical preparation and blood serum, recovery values were 77.3%-98.4%. The results obtained show that the modified sensor can be applied to the detection of EP in the presence of AA. The proposed sensor is promising for routine analysis because of its high selectivity, reproducibility, reproducibility and long-term stability characteristics and high recovery values obtained in pharmaceutical and biological samples.

Keywords: Epinephrine, *p*-aminobenzene sulfonic acid, sensor, voltammetry.

Poli (*p*-Aminobenzen Sülfonik Asit) Modifiye Sensöründe Askorbik Asit Varlığında Epinefrinin Elektrokimyasal Tayini

Özet. Poli (*p*-aminobenzen sülfonik asit, ABSA) modifiye sensör ile askorbik asit varlığında epinefrinin (EP) hızlı, güvenilir ve duyarlı bir şekilde seçici ve eş zamanlı voltammetrik tayini gerçekleştirildi. Camı karbon elektrot, ABSA ile 0.1 M KCl çözeltisi içerisinde çevrimli voltametri tekniğiyle başarılı bir şekilde modifiye edildi. Polimerik ince film ile modifiye edilen sensör, EP ve askorbik asitin (AA) yükseltgenmesine karşı mükemmel elektro-katalitik etki göstermiştir. Sonuçlar, her iki türün oksidasyon potansiyeli ve oksidasyon pik akım yanıtlarının önemli derecede iyileştiğini göstermiştir. Modifiye sensör, optimum koşullar altında EP tayinine karşı 5.0–53.2 µM derişim aralığında 50 nM tayin sınırı ile mükemmel cevap göstermiştir. Farmasötik preparatta ve kan serumunda gerçekleştirilen gerçek numune analizlerinde geri kazanım değerleri % 98.4 - 77.3 di. Elde edilen sonuçlar, modifiye sensörün AA varlığında EP'in saptanmasında uygulanabileceğini göstermiştir. Önerilen sensörün, yüksek seçicilik, tekrarlanabilirlik, tekrarüretilebilirlik ve uzun-sürelilik özelliklerinden ve farmasötik ve biyolojik örneklerde elde edilen yüksek geri kazanım değerlerinden dolayı rutin analizlerde uygulanabilecek olması umut vericidir.

Anahtar Kelimeler: Epinefrin, *p*-aminobenzen sülfonik asit, sensör, voltametri.

1. INTRODUCTION

Catecholamine is a class of important compounds involved in the transfer of transmissions in the central nervous system of mammals [1]. It is released by the adrenal medulla at low blood sugar levels or psychological stress conditions [2,3]. Catecholamines are used as medicines in heart surgery, myocardial infarction, hypertension and bronchial asthma treatment [4]. Catecholamines act as neurotransmitter substances in brain tissues [5]. Sudden changes in the concentrations of these biochemical substances that provide conduction in the central nervous system affect the life [5, 6]. Epinephrine one of the most significant catecholamines, belongs to the family of excitatory chemical neurotransmitters [7, 8]. Epinephrine, also known as adrenaline, is one of the important catecholamines that play an important role in the functioning of the central nervous system, renal hormonal, physical, cardiovascular system and mental stress, and stimulate a series mechanism of the sympathetic nervous system [7, 9]. Abnormalities at EP level are the symptoms of some diseases such as Parkinson 's disease [10].

Consequently, it has been focused on the detection of such molecules which are important in biochemistry and which provide nerve conduction [5, 6]. In general, chromatographic (HPLC) [11], fluorometric [12] and spectrometric [11] methods are used for detection of such molecules. Furthermore, it is also possible to carry out the determination of the molecules responsible for neurotransmission by electrochemical methods, since EP are electrochemically active [11]. In addition, the electrochemical behavior of catecholamines was studied by differential pulse polarography a glassy carbon electrodes in pharmacologically [11]. It was also possible to determination of EP by using Au and Pt rotating disc electrodes [11, 13-15]. Quantitative analysis of catecholamines in various urine, brain and plasma specimens by flow injection technique were performed electrochemically [11]. However, AA and uric acid are overlapped when they are present together with EP [12, 13, 16, 17]. Especially AA which shows similar electrochemical behavior to

these active compounds, is oxidized at closed potentials [18]. Thereby, the determination of the level of such EP in the cerebrospinal fluid is very important in terms of the diagnosis and the treatment of the related diseases [2, 3]. The simultaneous determination of EP is very important in terms of electroanalysis, in the presence of many interfering substances in biological fluids such as blood, serum and urine.

Due to the large anodic over-potential and contamination of the electrode surface with oxidation products, the determination of EP and direct electrochemical oxidation is very difficult. Many studies have been reported on the electrocatalytic oxidation of EP to overcome these problems using various chemically modified electrodes [5, 7, 19]. Polymer-modified carbon electrodes are preferred because they have lower current at a wider potential range compared to metal electrodes, as the uncertainty in response to electrode currents is minimized [20].

In this study, selective membrane sensors were prepared with poly (ABSA) glassy carbon electrode (GCE) for determination of EP in the presence of ascorbic acid. Voltammetric determination of EP in real samples were also performed such as in drug and blood serum samples in order to demonstrate the analytical performance and catalytic ability of the modified sensor. The recovery values showed that the results were in agreement with the EP value in the drug samples. The results indicated that the proposed method could be used easily in the identification of catecholamines in drug samples and clinical analysis.

2. MATERIALS AND METHODS

2.1. Chemicals

Chemical substances used in this studies, *p*-aminobenzene sulfonic acid (Aldrich), ascorbic acid (Aldrich), KCl, Na₂S₀₄, NaClO₄, NaNO₃, NaCl, Na₂HPO₄, Epinephrine (Merck), KH₂PO₄ (Carlo Erba) were analytical purity. Adrenaline® (Drogsan) (1 mL of an ampoule contains 1 mg Epinephrine) was used for recovery studies.

2.2. Apparatus and Equipment Used in Measurement

In voltammetry experiments, BAS 100BW (Bioanalytical Systems, Inc.) electrochemical analyzer was used. The electrochemical cell used in the study was placed in the C3 cell stand, so that it was isolated from the external electrical and magnetic effects of the cell thanks to the Faraday cage. The pH measurements were measured with Orion 601A. Bandelin Sonorex brand RK 100 ultrasonic bath was used during the cleaning of the electrodes and preparing of the solutions. Mono Block AB204-S precision scale was used for weighing of the chemicals. The ultra-pure water, which obtained from Millipore Milli-Q water system, was used for preparing solutions. Ag / AgCl electrode (CHI111) was used as the reference electrode, hand-made platinum electrode in the form of a spiral disk was used as auxiliary electrode, GCE (CHI104) and poly (ABSA) electrodes were used as the working electrodes.

The reference electrodes were stored in 3 M KCl solution. The working electrode was cleaned on a velvet disk (BAS, MF-1040) using 0.3 and 0.05 μm Al_2O_3 slurries, respectively, and subsequently washed with ultra-pure water. After mechanical cleaning, these electrodes were kept in ultrasonic bath for 5 min in ultra-pure water. Prior to electrochemical modification, the bare GCE was sonicated for 5 min in 1: 1 HNO_3 and pure water solution.

2.3. Electrochemical Measurements

Films of poly (ABSA) were deposited on a GCE in a 0.1 M KCl solution containing 5.0 mM ABSA by cycling 14 times between (-1.5) V to (2.5) V using cyclic voltammetry (CV) technique (vs. Ag / AgCl). The electrochemical measurements of EP in the 5.0-53.2 μM concentration range were performed in 0.1 M PBS (pH 7.0) at the poly (ABSA) sensor at by differential pulse voltammetry (DPV) technique (vs. Ag / AgCl).

2.4. Preparation of Real Samples

A drug sample known as Adrenaline (0.5 mg EP / mL) was analyzed using the proposed method to demonstrate its analytical applicability and to validate the poly (ABSA) modified sensor. Drug samples were diluted 1:50 with 0.1 M PBS (pH 7.0) and five replicate samples were analyzed. It was observed that the results obtained using the poly (ABSA) modified sensor were in agreement with the amounts recommended in the drug samples. In blood serum application, the blood serum was diluted 1: 4 with 0.1 M PBS buffer (pH 7.0) and Adrenaline added to the sample in the concentration of 1.0 mM of EP.

3. RESULTS AND DISCUSSION

3.1. Modification of Electrodes with Poly (ABSA)

Electropolymerization of poly (ABSA) were deposited on a GCE in a 0.1 M KCl solution containing 5.0 mM ABSA monomer by using CV technique (vs. Ag / AgCl) at -1500 mV to 2500 mV at a scan rate of 50 mV/s. Prior to the polymerization, nitrogen gas was purged in the monomer solution for ten minutes. Repeatable cycles, showing that the monomer is stably polymerized, are shown in Figure 1 [22].

The anodic and cathodic peaks of ABSA can be seen in Figure 1. Poly (ABSA) has two redox pairs, which shown an irreversible reaction. Oxidation peaks were observed at about 0.50 V (peak C) and 0.82 V (peak D), respectively, reduction peaks were observed at about 0.45 V (peak B) and -0.45 V (peak A), respectively. An adherent bluish polymer formed on the GCE as the number of cycles increased. The mechanism of electropolymerization of ABSA is also found in the literature [23]. G. Jin et al. were also carried out electropolymerization of ABSA at GCE in different supporting electrolyte solution [23]. Cyclic Voltammograms of 2.0×10^{-3} M ABSA were investigated in pH 7.0 acetate buffer solution containing 0.1 M NaCl at the GCE [23]. In the first scan, anodic peak and cathodic peak were seen with peak potential value at +0.9 and -0.7V,

respectively. From the second cycle on, anodic peak appeared with potential at +0.2V [23].

The modified sensor was found to be more stable when immersed in distilled water and dried in air. In order to obtain repeatable currents, it was necessary to use the modified electrodes very carefully. When approximately 10-15 measurements were taken in 0.1 M phosphate buffer solution (PBS) by DPV, no anodic peak was detected when the measurement was taken again for the ground with a used sensor. In addition, when the modified electrode was not used stored in 0.1 M PBS and it was observed that there was a decrease of only about 5-8% in peak currents after one week. Furthermore, when DPV measurements were taken after the modified sensor was stored in PBS for one month, only 12-15 percent decrease in peak current responses was observed. These results showed that the availability and reusability of the modified sensor was very good. Consequently, it was understood that the prepared modified sensor was stable, repeatable, reproducible and repeated analyzes could be performed with the same sensor.

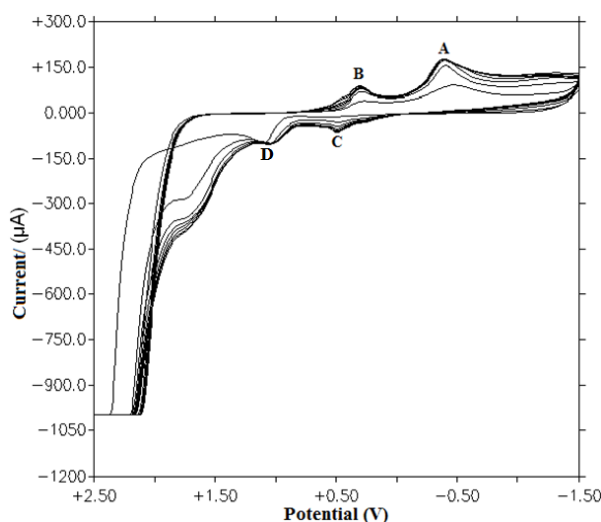
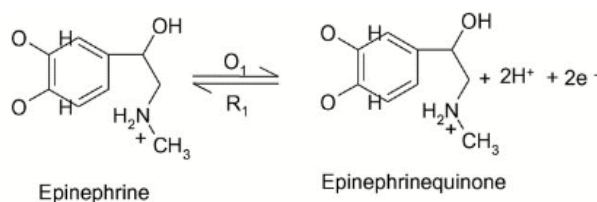


Figure 1. The CVs of 5.0 mM ABSA in 0.1 mM KCl at GCE, with 14 cycles, scanning rate: 50 mV / s.

3.2. Electrochemical Behavior of EP

One of the objectives of this work was the development of a modified sensor capable of the electrochemical oxidation of EP. In order to investigate the electrochemical activity of poly

(ABSA) film, the CVs were obtained in the presence of 0.2 mM EP, and the curves are shown in Figure 3. A cathodic reduction peak at nearly 0.17 V and two anodic oxidation peaks were observed at nearly 0.10 V and 0.20 V, respectively (Figure 2). Since the peak current intensity of the oxidation was greater than the reduction peak current intensity, EP displayed a semi-reversible reaction. In another study, EP exhibited semi-reversible nature at poly (caffeic acid) modified GCE [24]. These results suggest that two-electron transfer processes were involved in the rate-determining steps of all oxidation processes of EP by supporting the reaction in Scheme 1. The two one-electron transfer processes might corresponded to reaction (Scheme 1), where oxidation of two hydroxyl groups occur at a very similar oxidation potential, but the small potential difference was caused by different positions of these groups relative to the other substituents in the ring [25].



Scheme 1. Oxidation reaction of Epinephrine to Epinephrinequinone [26,27].

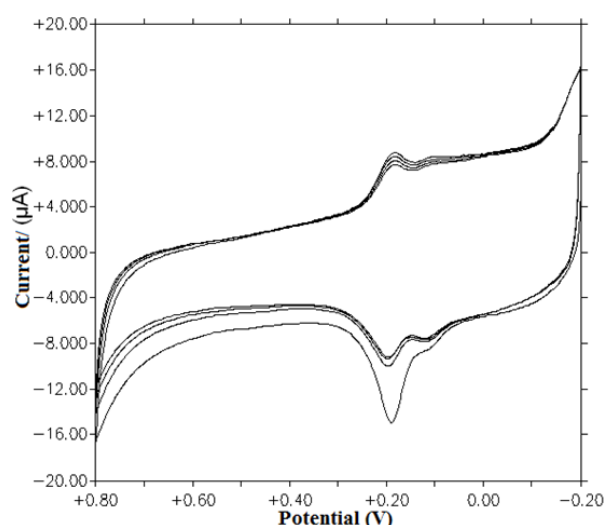


Figure 2. The CVs of 0.2 mM EP in 0.1 M PBS (pH 7.0) at poly (ABSA) sensor.

3.3. Effect of Film Thickness

In order to evaluate the film thickness of modified sensor, DPV technique was used to determination of the EP at modified sensors that had different film thickness with different cycles, and the EP was oxidized at nearly 200 mV (Figure 3). When the responses at the sensors with various film thickness (8-16 cycles) were viewed, the highest peak current was observed for the EP at the sensor, which had 14 cycled film thickness. An increase in current responses of EP was observed at increased film thickness. This indicates that the film has both an electrocatalytic property and a permselective behavior, which prevents the passage of EP with a large molecular structure by increasing the film thickness. It was found that thicker films than 14 cycles were not suitable because the durability of the films (dissolution and wiping) and homogeneity (cracking on the surface) decrease. The sensor, which had 14 cycles film thickness, was used in subsequent studies.

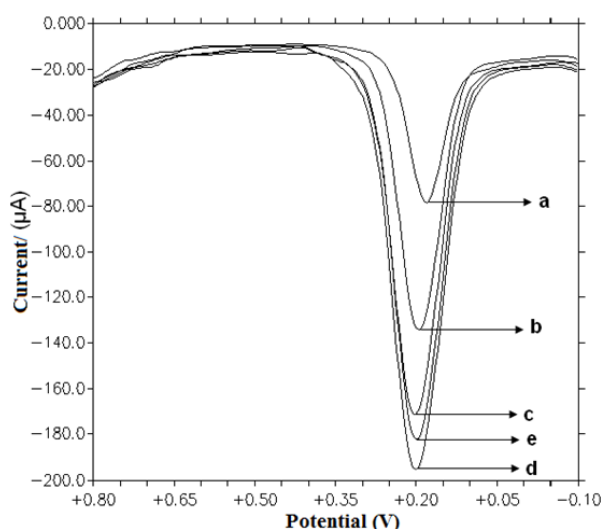


Figure 3. DPVs of 0.2 mM EP in 0.1 M PBS (pH 7.0) at poly (ABSA) sensor film thicknesses with a) 8 b) 10 c) 12 d) 14 e) 16 cycles.

3.4. Effect of Supporting Electrolyte

The type of supporting electrolyte, which is one of the parameters that can affect the voltammetric behavior of EP, plays an important role. The well-supporting electrolyte solution forms a conductive environment between the submerged electrodes. Apart from this basic purpose, the supporting

electrolyte is doped in the composed polymer. The choice of support electrolyte depends on its resolution, dissociation degree, and nucleophilic character [28]. For this purpose, the DPV responses of Epinephrine were investigated in Na_2SO_4 , PBS (pH 7.0), NaNO_3 , NaClO_4 , NaCl and KCl electrolytes. While experiments in PBS were performed at pH 7.0, the experiments in the other electrolyte species were realized at the natural pHs.

Responses of EP were examined in Figure 4, it was observed that the sharpest peak and the highest peak current were better in PBS (pH 7.0) compared to other electrolyte types. The voltammetric behavior of EP and AA at the polymeric sensor was investigated at different pHs of the selected PBS supporting electrolyte.

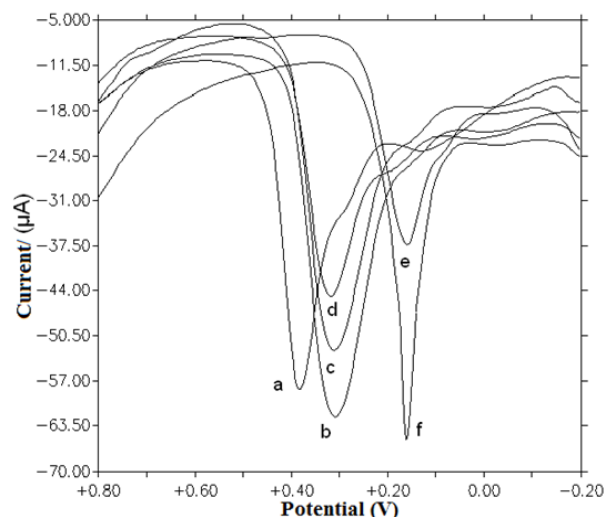


Figure 4. DPVs of 0.2 mM EP at poly (ABSA) sensor in different supporting electrolyte solutions a) 0.1 M NaNO_3 b) 0.1 M Na_2SO_4 c) 0.1 M KCl d) 0.1 M NaCl e) 0.1 M NaClO_4 f) 0.1 M PBS (pH 7.0).

3.5. Effect of pH

The poly (ABSA) film contains electronically rich nitrogen atoms and high sulphonyl groups with electron density. Therefore, poly (ABSA) film is negatively charged [23]. The relationships between the potential and current obtained from responses of EP in solutions with pH from 2.0 to 11.0 were plotted in order to observe how it behaved in relation to pH in the PBS (Figure 5). Determination of 0.2 mM EP using DPV techniques was performed in 0.1 M PBS at different pHs in the

range of 2 to 11. Figure 5 shows that as the pH of the PBS supporting electrolyte medium increases, the peak potentials of EP responses shift to lower potentials. Peak potential was found to be pH dependent. There is an increase in peak currents up to pH 7.0, but a decrease in peak currents after pH 7.0. Furthermore, the best peak shape and current of DOPAC were obtained at pH 7.0.

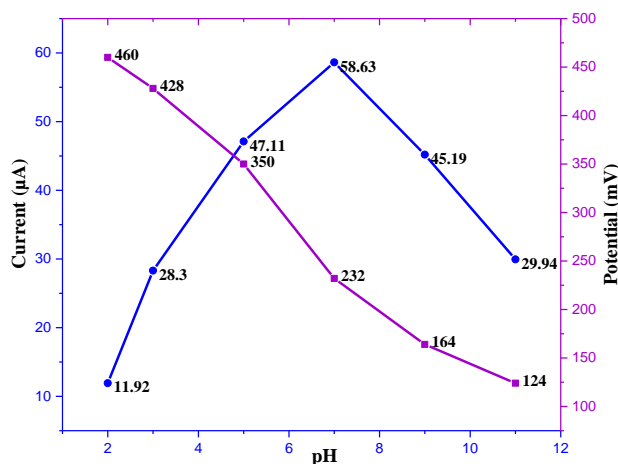


Figure 5. The effect of pH on Current and Potential responses of 0.2 mM EP from DPV results.

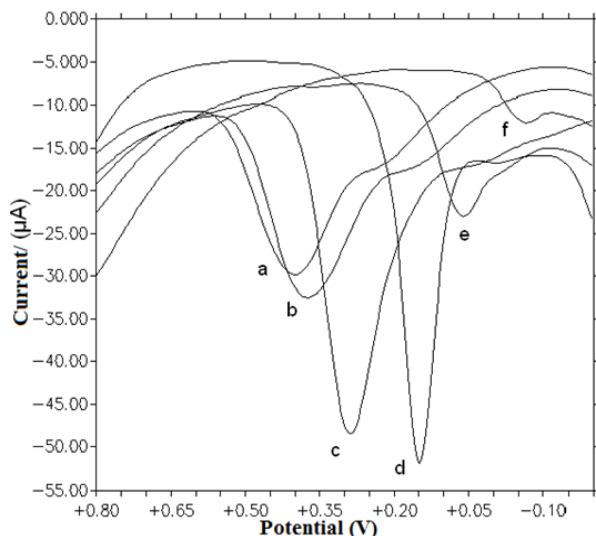


Figure 6. DPVs of 0.2 mM EP at poly (ABSA) sensor in PBS at different pHs a) 2.0 b) 3.0 c) 5.0 d) 7.0 e) 9.0 f) 11.0.

Differential pulse voltammetric responses of 0.2 mM EP in the PBS at pH ranging from 2.0 to 11.0 were shown in Figure 6. As seen in Figure 6, the oxidation peak potentials were shifted to lower potentials as the pH increased, and the oxidation peak current value was highest at pH 7.0. Furthermore, the maximum response at pH 7.0, which is physiological pH, is promising in terms of working with real samples. Determination of EP in the presence of AA was performed at the modified sensor in PBS at varying pHs using DPV technique. From the results, simultaneous determination of EP and AA, and highest peak currents were obtained at pH 7.0. Consequently, subsequent electroanalytical studies were performed at PBS pH 7.0 as the supporting electrolyte medium.

3.6. Determination of EP

Determination of EP at the poly (ABSA) sensor was performed using DPV. Differential pulse voltammograms of different EP concentrations (5.0, 9.8, 14.9, 18.9, 25.0, 29.0, 34.0, 37.9, 44.0, 48.0, 53.2 μM) in 0.1 M PBS (pH 7.0) at the poly (ABSA) sensor were shown in Figure 7. The results showed that the anodic peak currents of EP were linear at a concentration range of 5.0 μM to 53.2 μM . The calibration equation for EP was calculated as $I_{pa} (\mu\text{A}) = 0.6103C (\mu\text{M}) + 1.935$. The sensitivity, the detection limit (LOD) and quantification limit of EP were found from the slope of the calibration curve. $\text{LOD} = 3 s/m$, $\text{LOQ} = 10 s/m$; where, s is the standard deviation of the peak currents (for five runs) and m is the slope of the calibration curve. The LOD was 50 nM, LOQ was 166.7 nM and the sensitivity was 0.6103 $\mu\text{A}/\mu\text{M}$.

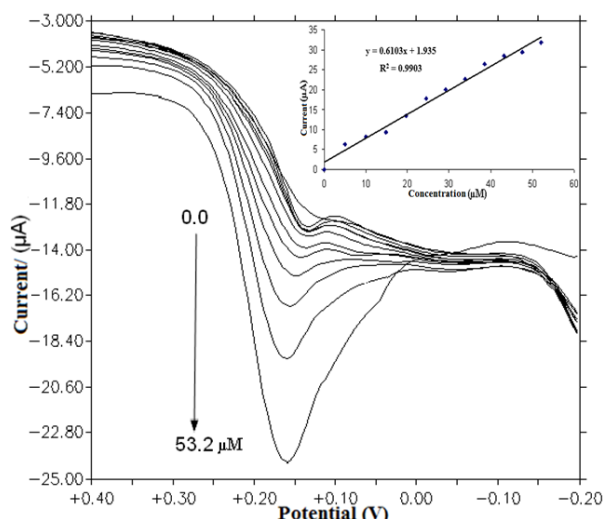


Figure 7. DPVs and calibration graph of increased EP concentrations (5.0 μM - 53.2 μM) in 0.1 M PBS (pH 7.0) at the poly (ABSA) sensor.

The electrochemical investigation of EN on the surface of GCE modified with over-oxidized PBSA film has shown a linear range from 5.0 to 53.2 μM with a detection limit of 50 nM. However, the voltammetric response properties obtained in this work (including linear range and detection limit) are better than similar studies from literature. A comparison between voltammetric response properties of various modified electrode and proposed modified sensor in this study for determination of EN was shown in Table 1.

Table 1. Voltammetric response characteristics of different modified electrodes for EN determination.

Modified electrode	Techniques	Condition	Linear range for EN (μM)	LOD (μM)	Ref.
MDWCNTPE	CV	PBS (pH 7.0)	0.7–1200	0.216	[5]
BBNBHTNMCPE	DPV	PBS (pH 8.0)	1.0–600.0	0.2	[8]
DMSA-Au	CV/DPV	PBS (pH 7.7)	0.40–3.0/ 0.40–4.0	0.054/0.053	[15]
Poly (caffeic acid)/GCE	CV	PBS (pH 7.4)	2.0–300	0.6	[24]
Au–Ag thin film/Au coated Pt plates	DPV	PBS (pH 7.0)	10–100	5.05	[25]
FePc/CPE	DPV	Acetate (pH 4.0)	1–30	0.5	[26]
BCH/GCE	DPV	PDPB (pH 5.8)	1-130	–	[29]
Caffeic acid/GCE	FIA/DPV	PBS (pH 6.0)	0.1–10	–	[30]
Poly (caffeic acid)/GCE	CV	PBS (pH 7.7)	2–80	0.6	[31]
f-MWCNT-Nf-PtAu	DPV	PBS (pH 6.75)	60–240	–	[32]
o-ABA/GCE		PBS (pH 7.0)	–	–	[33]
DMSA-PCA/Au	CV/DPV	PBS (pH 7.7)	5.0–800	0.39/0.25	[34]
Present work	DPV	PBS (pH 7.0)	5.0–53	0.05	–

3.7. Determination of EP in The Presence of AA

In order to prove the performance of the modified sensor, the DPV responses were investigated in the solutions containing increasing concentrations of EP in the presence of AA. For this purpose, DPV measurements were performed in PBS at pH 7.0 containing 3.0 mM AA and EP (0.04, 0.08, 0.12, 0.16, 0.2, 0.24, 0.28, 0.32, 0.36, 0.40 mM) at the modified sensor (Figure 8). The AA peaks remained the same while the EP peaks increased in direct proportion. The fact that EP gave this result in the presence of AA at the physiological pH value of 7.0 makes EP can be determined by this method

in real samples such as human or animal samples, possible.

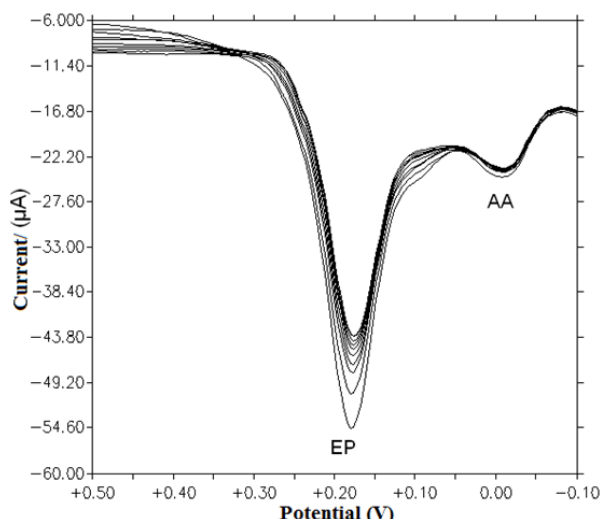


Figure 8. DPVs of increased concentrations of EP (0.04 - 0.40 mM) in the presence of 3.0 mM AA in 0.1 M PBS (pH 7.0) at poly (ABSA) sensor.

Differential pulse voltammetric responses of increased AA concentrations (1.0, 2.0, 4.0, 6.0, 8.0, 10.0 mM) in the presence of 0.2 mM EP were shown in Figure 9. As AA concentration increased, the peak current of AA also increased linearly and the peak current density of EP remained constant. This suggests that the method can be applied in samples containing even high concentrations of AA.

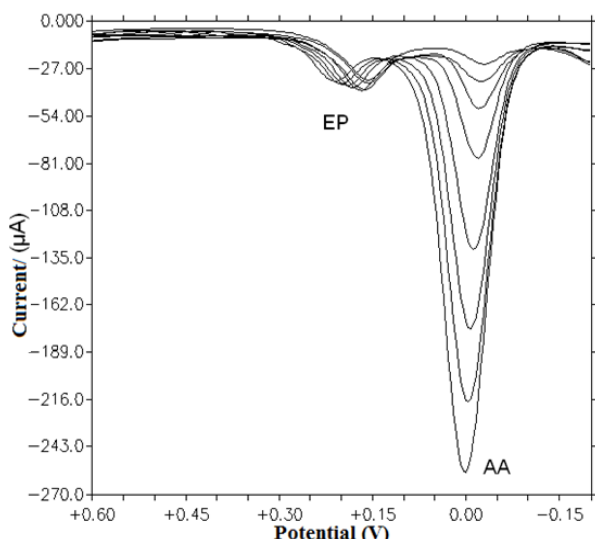


Figure 9. DPV of increased concentrations of AA (1.0 - 10.0 mM) in the presence of 0.2 mM EP in 0.1 M PBS (pH 7.0) at poly (ABSA) sensor.

Effect of analytical parameters on the peak current of EP such as film thickness, supporting electrolyte type and pH were optimized. The EP amounts in pharmaceutical preparations were analyzed at the proposed modified sensor. The modified sensor obtained by electropolymerization of ABSA on the GCE were shown that simultaneous determination of EP could be performed even in the presence of ascorbic acid. This sensor was successfully applied to pharmaceutical preparation and biological fluid samples.

3.8. Analysis of EP in Real Samples

Five replicate solutions of these drug and blood serum samples were prepared and EP responses were obtained using the DPV technique. EP concentrations in real samples were calculated by using the obtained current values from the calibration curve equation. The recovery values obtained from the EP analysis in real samples showed the accuracy and reproducibility of the sensor (Table 2). From the results found, the recovery values in the real samples were found satisfactory. This proved the applicability and accuracy of the sensor and the method.

Table 2. Analytical application of EP in real samples at poly (ABSA) sensor.

Samples	Labeled, mM	Found, mM	RSD*	Recovery, %
Adrenaline	1.000	0.984	0.163	98.4
Blood Serum	1.000	0.773	0.310	77.3

* RSD: Relative standard deviation.

4. CONCLUSIONS

A poly (ABSA) modified sensor was used for simultaneous determination of EP and AA by voltammetric methods. When CV and DPV responses of EP in the presence of AA were examined, it was observed that a single overlapped peak appeared at the bare GCE. However, AA and EP were found to be oxidized at different peak potentials at the poly (ABSA) sensor. It has been shown that AA is the most interfering species and catecholamines can be detected in the presence of AA at developed sensor. The results show that the proposed method can be used easily for identification of catecholamines in drug samples and clinical analyzes. The detection of EP was successfully and satisfactorily carried out in real samples such as human blood serum and pharmaceutical preparation at the poly (ABSA) sensor.

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REFERENCES

1. Tavana, T., Khalilzadeh, M.A., Karimi-Maleh, H., Ensafi, A.A., Beitollahi, H. and Zareyee, D., Sensitive voltammetric determination of epinephrine in the presence of acetaminophen at a novel ionic liquid modified carbon nanotubes paste electrode, *J. Mol. Liq.*, 168 (2012) 69–74.
- [2] Jackson, L., Williams, F.L.R., Burchell, A., Coughtrie M.W.H. and Hume, R., Plasma catecholamines and the counterregulatory responses to hypoglycemia in infants: A critical role for epinephrine and cortisol, *J. Clin. Endocrinol. Metab.*, 89-12 (2004) 6251–6256.
- [3] Bergquist, J., Ciubisz, A., Kaczor, A. and Silberring, J. Catecholamines and methods for their identification and quantitation in biological tissues and fluids. *J. Neurosci. Meth.* 113 (2002) 1–13.
- [4] Martinez-Quintana, E. et al. Acute myocardial infarction secondary to catecholamine release owing to cocaine abuse and pheochromocytoma crisis. *Int. J. Endocrinol. Metab.*, 11 (2013) 48–51.
- [5] Beitollahi, H., Ardakani, M.M., Ganjipour, B. and Naeimi, H. Novel 2,2'-[1,2-ethanediylbis(nitriloethylidyne)]-bis-hydroquinone double-wall carbon nanotube paste electrode for simultaneous determination of epinephrine, uric acid and folic acid. *Biosens. Bioelectron.*, 24 (2008) 362–368.
- [6] Jin, G.-P., Lin, X.-Q. and Gong, J. Novel choline and acetylcholine modified glassy carbon electrodes for simultaneous determination of dopamine, serotonin and ascorbic acid. *J. Electroanal. Chem.*, 569 (2004) 135–142.
- [7] Mazloum-Ardakani, M., Talebi, A., Naeimib, H. and Sheikh-Mohseni, M.A. Electrocatalysis of epinephrine by TiO₂ nanoparticles and 2,20-[1,7-hepthandiyylbis(nitriloethylidyne)]-bis-hydroquinone modified carbon paste electrode. *Anal. Methods*, 3 (2011) 2328–2333.
- [8] Mazloum-Ardakani, M. et al. Simultaneous determination of epinephrine and acetaminophen concentrations using a novel carbon paste electrode prepared with 2,2'-[1,2 butanediylbis(nitriloethylidyne)]-bis-hydroquinone and TiO₂nanoparticles. *Colloids Surfaces B Biointerfaces* 76 (2010)82–87.
- [9] Wong, D.L., Tai, T.C., Wong-Faull, D.C., Claycomb, R. and Kvetňanský, R. Adrenergic responses to stress: Transcriptional and post-transcriptional changes. *Ann. N. Y. Acad. Sci.*, 1148 (2008) 249–256.
- [10] Carbone, M., Nesticò, A., Bellucci, N., Micheli, L. and Palleschi, G. Enhanced performances of sensors based on screen printed electrodes modified with nanosized

- NiO particles. *Electrochim. Acta*, 246 (2017) 580–587.
- [11] Zhang, W., He, L., Gu, Y., Liu, X. and Jiang, S. Effect of ionic liquids as mobile phase additives on retention of catecholamines in reversed-phase high-performance liquid chromatography. *Anal. Lett.*, 36 (2003) 827–838.
- [12] Babaei, A., Sohrabi, M. and Afrasiabi, M. A Sensitive Simultaneous Determination of Epinephrine and Piroxicam Using a Glassy Carbon Electrode Modified with a Nickel Hydroxide Nanoparticles/Multiwalled Carbon Nanotubes Composite. *Electroanalysis*, 24 (2012) 2387–2394.
- [13] Niu, L.M., Luo, H.Q. and Li, N.B. Electrochemical behavior of epinephrine at a penicillamine self-assembled gold electrode, and its analytical application. *Microchim. Acta* 150 (2005) 87–93.
- [14] Wang, L. et al. Electrochemical behavior and determination of epinephrine at a penicillamine self-assembled gold electrode. *Int. J. Electrochem. Sci.*, 1 (2006) 238–249.
- [15] Niu, L.M., Luo, H.Q. and Li, N.B. Electrochemical behavior of uric acid at a meso-2,3-dimercaptosuccinic acid self-assembled gold electrode. *Instrum. Sci. Technol.*, 35 (2007) 59–73.
- [16] Fang, C., Tang, X. and Zhou, X. Preparation of Poly(malachite green) Modified Electrode and the Determination of Dopamine and Ascorbic Acid. *Anal. Sci.*, 15 (1999) 41–46.
- [17] Kumar, S.S., Mathiyarasu, J., Phani, K.L., Jain, Y.K. and Yegnaraman, V. Determination of uric acid in the presence of ascorbic acid using poly(3,4-ethylenedioxythiophene)-modified electrodes. *Electroanalysis*, 17 (2005) 2281–2286.
- [18] Zhao, Y., Bai, J., Wang, L., Huang, P. and Wang, H. Simultaneous Electrochemical Determination of Uric Acid and Ascorbic Acid Using L -Cysteine Self-Assembled Gold Electrode. *Int. J. Electrochem. Sci.*, 1 (2006) 363–371.
- [19] Yang, Z., Hu, G., Chen, X., Zhao, J. and Zhao, G. The nano-Au self-assembled glassy carbon electrode for selective determination of epinephrine in the presence of ascorbic acid. *Colloids Surfaces B Biointerfaces*, 54 (2007) 230–235.
- [20] Wang, H.-S., Li, T.-H., Jia, W.-L. and Xu, H.-Y. Highly selective and sensitive determination of dopamine using a Nafion/carbon nanotubes coated poly(3-methylthiophene) modified electrode. *Biosens. Bioelectron.*, 22 (2006) 664–669.
- [21] Savan, E.K. and Erdoğan, G. Determination of 3,4-dihydroxyphenylacetic acid in the presence of ascorbic acid and uric acid at poly(p-aminobenzene sulfonic acid) conducting polymer electrode. *Polym. Bull.*, 74-10 (2017) 4349-4360.
- [22] Erdoğan, G. and Mutlu, M.M. Selective Detection of Dopamine in the Presence of Ascorbic Acid at Poly (m-Aminobenzene Sulfonic Acid). *Am. J. Anal. Chem.*, 02 (2011) 582–588.
- [23] Jin, G., Zhang, Y. and Cheng, W. Poly(p-aminobenzene sulfonic acid)-modified glassy carbon electrode for simultaneous detection of dopamine and ascorbic acid. *Sensors Actuators B Chem.*, 107 (2005) 528–534.
- [24] Ren, W., Luo, H.Q. and Li, N.B. Electrochemical behavior of epinephrine at a glassy carbon electrode modified by electrodeposited films of caffeic acid. *Sensors*, 6 (2006) 80–89.
- [25] Wierzbicka, E. and Sulka, G.D. Nanoporous spongelike Au–Ag films for electrochemical epinephrine sensing. *J. Electroanal. Chem.*, 762 (2016) 43–50.
- [26] Shahrokhian, S., Ghalkhani, M. and Amini, M.K. Application of carbon-paste electrode modified with iron phthalocyanine for voltammetric determination of epinephrine in the presence of ascorbic acid and uric acid. *Sensors Actuators, B Chem.*, 137 (2009) 669–675.
- [27] Bakir C.C., Preparation and characterization of metal nano particles modified carbon nanotube and polymer film

- electrodes and their analytical applications. Ege Üniversitesi, Fen Bilimleri Enstitüsü, (2010).
- [28] Rostami, A. and Taylor, M.S. Polymers for anion recognition and sensing. *Macromol. Rap. Commun.*, 33 (2012) 21-34.
- [29] Liu, H., Zhao, G., Wen, L. and Ye, B. Simultaneous voltammetric determination of epinephrine and serotonin at a p-tetra-butyl calix [6] arene-L-Histidine chemically modified electrode. *J. Anal. Chem.*, 61 (2006) 1104–1107.
- [30] Gong, J. and Lin, X. A glassy carbon supported bilayer lipid-like membrane of 5,5-ditetradecyl-2-(2-trimethyl-ammonioethyl)-1,3-dioxane bromide for electrochemical sensing of epinephrine. *Electrochim. Acta*, 49 (2004) 4351–4357.
- [31] Ren, W., Luo, H.Q. and Li, N. B. Simultaneous voltammetric measurement of ascorbic acid, epinephrine and uric acid at a glassy carbon electrode modified with caffeic acid. *Biosens. Bioelectron.*, 21 (2006) 1086–1092.
- [32] Yogeswaran, U., Thiagarajan, S. and Chen, S.-M. Nanocomposite of functionalized multiwall carbon nanotubes with nafion, nano platinum, and nano gold biosensing film for simultaneous determination of ascorbic acid, epinephrine, and uric acid. *Anal. Biochem.*, 365 (2007) 122–131.
- [33] Cheng, W., Jin, G. and Zhang, Y. Electrochemical Characteristics of Poly (o -Aminobenzoic Acid) Modified Glassy-Carbon Electrode and Its Electrocatalytic Activity towards Oxidation of Epinephrine . *Russ. J. Electrochem.*, 41 (2005) 940–945.
- [34] Taylor, P., He, Y. Bin, Luo, H. Q. and Li, N. B. Electrochemical Behavior of Epinephrine at Two - Component Self - Assembled Monolayer of meso - 2 , 3 - Dimercaptosuccinic Acid and Penicillamine on Gold Electrode. *Instr. Sci. Technol.*, 35 (2007) 163–175.