Anal. Bioanal. Electrochem., Vol. 9, No. 6, 2017, 718-728

Analytical & Bioanalytical Electrochemistry

2017 by CEE
www.abechem.com

Full Paper

Voltammetric Determination of Epinephrine Based on ZnO Nanoparticles Assisted Graphene Oxide Nanosheets

Mohammad Baniasadi,[1,*] Shohreh Jahani,[1,*] Hamed Maaref[1] and Reza Alizadeh[2]

[1]*Bam University of Medical Sciences, Bam, Iran*
[2]*Department of Chemistry, Faculty of Science, Qum University, Qum, Iran*

*Corresponding Author, Tel.: +98 2184234114; Fax: +98 2188948994
E-Mail: jahanishohre@gmail.com

Received: 4 June 2017 / Received in revised form: 31 July 2017 /
Accepted: 8 August 2017 / Published online: 30 September 2017

Abstract- This work reports the voltammetric determination of epinephrine (EP) using ZnO-graphene oxide nanocomposite (ZnO-GO). The electrochemical behaviors of epinephrine at ZnO-GO nanocomposite modified graphite screen printed electrodes (SPE) were studied by cyclic voltammetry and differential pulse voltammetry. The outcomes confirmed that the proposed electrode demonstrate excellent electrocatalytic activity towards the oxidation of epinephrine in phosphate buffer solution (PBS, pH 7.0). The fabricated electrode possess lowest detection limit of 0.07 µM for epinephrine with the very wide dynamic linear range of 0.5 µM to 500.0 µM. Finally, the modified sensor was successfully implemented to detect epinephrine in epinephrine injection and urine samples.

Keywords- Epinephrine, ZnO-GO nanocomposite, Graphite screen printed electrode, Voltammetry

1. INTRODUCTION

One example of neurotransmitters considered is epinephrine, also known as adrenaline. It neutrally channels the nerve impulse and is a key hormone produced by the medulla of the adrenal glands. Furthermore, it is recognized as the ‘fight’ or ‘flight’ hormone which will be transferred to the blood stream, exposing the thoughts of anger or worries and raises the blood glucose stage. It helped intermediate for carrying the nerve pulse to different body part.

Epinephrine in the medical field is used to fuel the heartbeat and to manage emphysema, bronchitis, bronchial asthma and other allergic illnesses; it is also used in the eye treatment and glaucoma. The properties of epinephrine in local anesthetics have been well established. Epinephrine is the vital constrictor of blood vessels and blood coagulation accelerator, specifically on the skin or mucous membranes for bleeding control at the procedure site. It decreases the absorption of local anesthetics into the bloodstream, causing reduced systemic toxic side effects, prolonged medical duration of action and decreased surgical blood loss. The alterations in the concentration of epinephrine lead to several diseases, such as, Schizophrenia and Parkinsonism; therefore it is important to improve quantitative means for epinephrine detection, to learn its physiological role and diagnosing certain illnesses in clinical medicine field [1-5].

During recent years the screen printing technology applied to sensor and biosensor construction has been considerably improved and a large number of papers, and recently some reviews, have appeared in the literature. Screen printed electrodes (SPEs) are in fact inexpensive, simple to prepare, rapid and versatile and this technology appears to be also the most economical mean for large-scale production of disposable electrodes [6-9]. The modification of electrodes has been widely explored by using different nanomaterials [10-23]. Nanomaterials are one of the good supporting materials with properties such as high mechanical, catalytic performance and chemical stability which support the diffusion of the reacting species [24-40].

Graphene has recently attracted tremendous interest because of its unique thermal, mechanical, and electrical properties. One of the promising applications of graphene is electrochemical sensing. Since every atom in a graphene sheet is a surface atom, molecular interaction and thus electron transport through graphene can be highly sensitive to adsorbed molecules. Graphene, which consists of a one-atom-thick planar sheet comprising and sp^2-bonded carbon structure with exceptionally high crystal and electronic quality, is a novel material that has emerged as a rapidly rising star in the field of material science. Ever since its discovery in 2004,5 graphene has been making a profound impact in many areas of science and technology due to its remarkable physicochemical properties. These include a high specific surface area (theoretically 2630 m^2/g for single-layer graphene), extraordinary electronic properties and electron transport capabilities, unprecedented pliability and impermeability, strong mechanical strength and excellent thermal and electrical conductivities. These unique physicochemical properties suggest it has great potential for providing new approaches and critical improvements in the field of electrochemistry. For example, the high surface area of electrically conductive graphene sheets can rise to high densities of attached analyte molecules. This in turn can facilitate high sensitivity and device miniaturization. Facile electron transfer between graphene and redox species opens up opportunities for sensing strategies based on direct electron transfer rather than mediation. It

is not surprising; therefore, that graphene has recently attracted great attention worldwide from the electrochemical community. Despite its short history, this 2D material has already revealed potential applications in electrochemistry, and remarkably rapid progress in this area has already been made. In recent years, many reviews covering graphene and related materials have been published. In addition, several reviews with particular emphasis on graphene-based electrochemical applications have also appeared [41-48].

Metal oxide nanoparticles such as zinc oxide (ZnO) had been extensively developed to respond to pharmaceutical formulations due to their low-cost, large specific surface area, being friendly to the environment and high sensitivity. Tremendous efforts had been devoted to create highly responsive sensors by incorporating nanoparticles metal oxide. However, these sensors had obvious electrical responses only on the premise of a high operating temperature, which resulted in power consumption and obstruction to miniaturization of the sensors [49-56].

According to the previous points, it is important to create suitable conditions for analysis of epinephrine in biological fluids. In this study, we describe application of novel ZnO-GO nanocomposite as a nanostructure sensor for voltammetric determination of epinephrine. The proposed sensor showed good electrocatalytic effect on epinephrine. Eventually, we evaluate the analytical performance of the suggestion sensor for epinephrine determination in real sample.

2. EXPERIMENTAL

2.1. Apparatus and chemicals

The electrochemical measurements were performed with an Autolab potentiostat/galvanostat (PGSTAT 302N, Eco Chemie, the Netherlands). The experimental conditions were controlled with General Purpose Electrochemical System (GPES) software. The screen-printed electrode (DropSens, DRP-110, Spain) consists of three main parts which are a graphite counter electrode, a silver pseudo-reference electrode and a graphite working electrode.

All solutions were freshly prepared with double distilled water. Epinephrine and all other reagents were of analytical grade and were obtained from Merck chemical company (Darmstadt, Germany). The buffer solutions were prepared from orthophosphoric acid and its salts in the pH range of 2.0-9.0.

Epinephrine ampoule was purchased from Caspian tamin Company, Iran, contained 200 mg in 5 ml of epinephrine and urine samples were prepared from local laboratories.

2.2. Synthesis of graphene oxide/ZnO nanorods nanocomposite

Graphene oxide nano sheets were synthesized from natural graphite flakes based on the modified Hummers and Offeman's method. The reduced graphene oxide (0.096 g) was dispersed in 40 ml water and the solution was kept in ultrasonic bath for 1 h. The prepared solution was added to 40 ml of $ZnCl_2$ (0.04 M) solution. Final solution pH was set 11.7 by ammonia solution. The solution was kept at 95 °C for 4 h. The precipitate was gathered at 15000 rpm centrifugation for 15 min. Then it was washed by distilled water three times. Finally it was dried in oven at 45 ºC for 4 h.

2.3. Preparation of modified electrode

The bare graphite screen printed electrode was coated with ZnO-GO nanocomposite as follows. A stock solution of ZnO-GO nanocomposite in 1 ml aqueous solution was prepared by dispersing 1 mg ZnO-GO nanocomposite with ultrasonication for 1 h, and a 5 µl aliquot of the ZnO-GO/H_2O suspension solution was casted on the carbon working electrodes, and waiting until the solvent was evaporated in room temperature.

2.4. Preparation of real samples

One milliliter of a epinephrine ampoule (Caspian tamin Company, Iran, contained 200 mg in 5 ml of epinephrine) was diluted to 10 ml with 0.1 M PBS (pH 7.0); then, different volume of the diluted solution was transferred into each of a series of 25 ml volumetric flasks and diluted to the mark with PBS. The epinephrine content was analyzed by the proposed method using the standard addition method.

Urine samples were stored in a refrigerator immediately after collection. 10 mL of the sample was centrifuged for 15 min at 2000 rpm. The supernatant was filtered out using a 0.45 µm filter. Then, different volume of the solution was transferred into a 25 mL volumetric flask and diluted to the mark with PBS (pH 7.0). The diluted urine sample was spiked with different amounts of epinephrine.

3. RESULTS AND DISCUSSION

3.1. Electrocatalytic oxidation of epinephrine at a ZnO-GO/SPE

The electrochemical behavior of epinephrine is dependent on the pH value of the aqueous solution. Therefore, pH optimization of the solution seems to be necessary in order to obtain the electrocatalytic oxidation of epinephrine. Thus the electrochemical behavior of epinephrine was studied in 0.1 M PBS in different pH values (2.0< pH< 9.0) at the surface of ZnO-GO/SPE by CV. It was found that the electrocatalytic oxidation of epinephrine at the surface of ZnO-GO/SPE was more favored under neutral conditions than in acidic or basic

medium. Thus, the pH 7.0 was chosen as the optimum pH for electrocatalysis of epinephrine oxidation at the surface of ZnO-GO/SPE.

Fig. 1 depicts the cyclic voltammetric responses for the electrochemical oxidation of 500.0 μM epinephrine at ZnO-GO/SPE (curve a) and bare SPE (curve b). The anodic peak potential for the oxidation of epinephrine at ZnO-GO/SPE (curve a) is about 275 mV compared with 385 mV for that on the bare SPE (curve b). Similarly, when the oxidation of epinephrine at the ZnO-GO/SPE (curve a) and bare SPE (curve b) are compared, an extensive enhancement of the anodic peak current at ZnO-GO/SPE relative to the value obtained at the bare SPE (curve b) is observed. In other words, the results clearly indicate that the ZnO-GO nanocomposite improve the epinephrine oxidation signal.

The effect of potential scan rates on the oxidation current of epinephrine has been studied (Fig. 2). The results showed that increasing in the potential scan rate induced an increase in the peak current. In addition, the oxidation process is diffusion controlled as deduced from the linear dependence of the anodic peak current (I_p) on the square root of the potential scan rate ($v^{1/2}$) over a wide range from 5 to 500 mV s^{-1}.

Further Tafel curves of both analytes were plotted using the data from the rising sections (i.e. the Tafel regions) of the current–voltage curves obtained at 10 mVs^{-1} (Fig. 3).

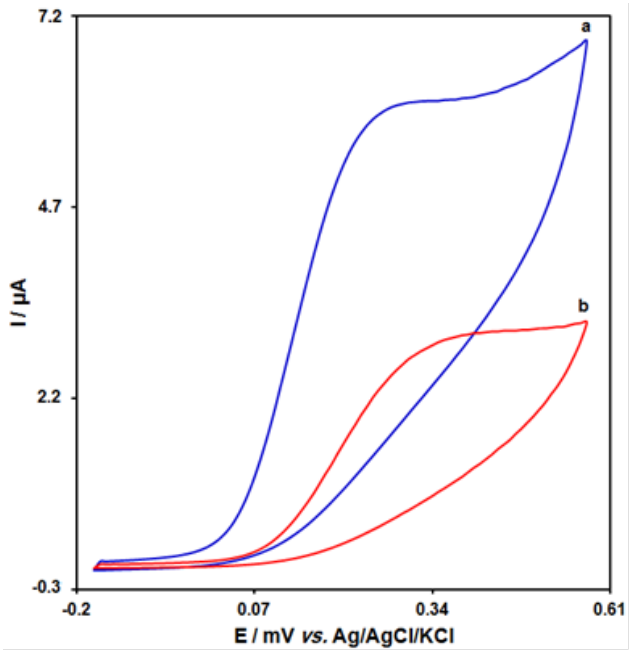

Fig. 1. Cyclic voltammograms of (a) ZnO-GO/SPE and (b) bare SPE in 0.1 M PBS (pH 7.0) in the presence of 500.0 μM epinephrine at the scan rate 50 mVs^{-1}

The Tafel regions of the current potential curves are influenced by the electron transfer kinetics of the electrode reactions.

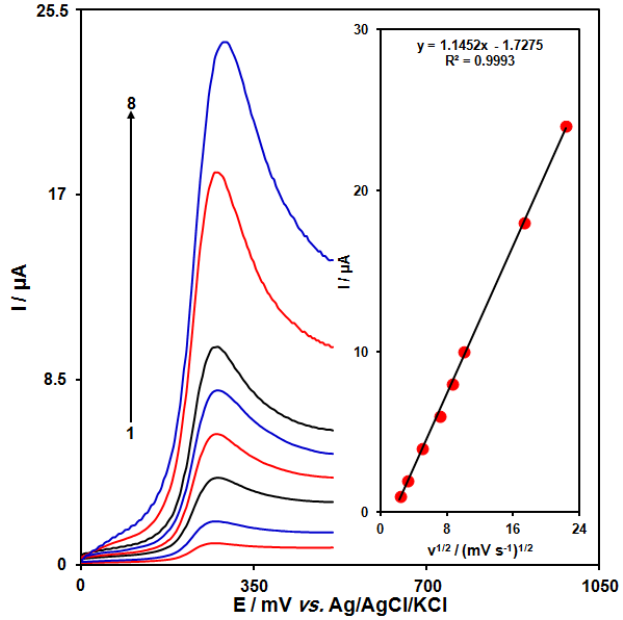

Fig. 2. LSVs of ZnO-GO/SPE in 0.1 M PBS (pH 7.0) containing 500.0 µM epinephrine at various scan rates; numbers 1-8 correspond to 5, 10, 25, 50, 75, 100, 300 and 500 mV s^{-1}, respectively. Inset: variation of cathodic peak current *vs*. $\nu^{1/2}$

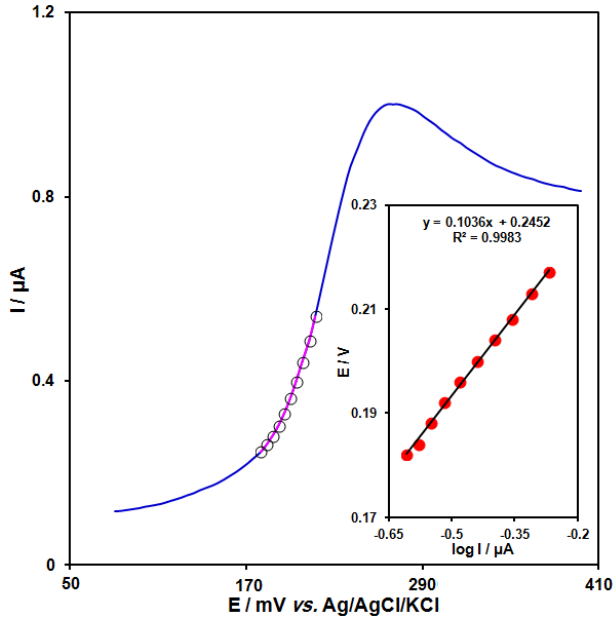

Fig. 3. LSV (at 5 mV s^{-1}) of electrode in 0.1 M PBS (pH 7.0) containing 500.0 µM epinephrine. The points are the data used in the Tafel plot. The inset shows the Tafel plot derived from the LSV

The results showed Tafel slope of 0.1036 V, which indicates one electron (Fig. 3) rate determining step (RDS) for the electrode process [57] for charge transfer coefficient (α) of 0.44.

3.2. Chronoamperometric measurements

Chronoamperometric measurements of amitriptyline at ZnO-GO/SPE were carried out by setting the working electrode potential at 0.33 V for the various concentration of epinephrine in PBS (pH 7.0) (Fig. 4). For an electroactive material (epinephrine in this case) with a diffusion coefficient of D, the current observed for the electrochemical reaction at the mass transport limited condition is described by the Cottrell equation [57].

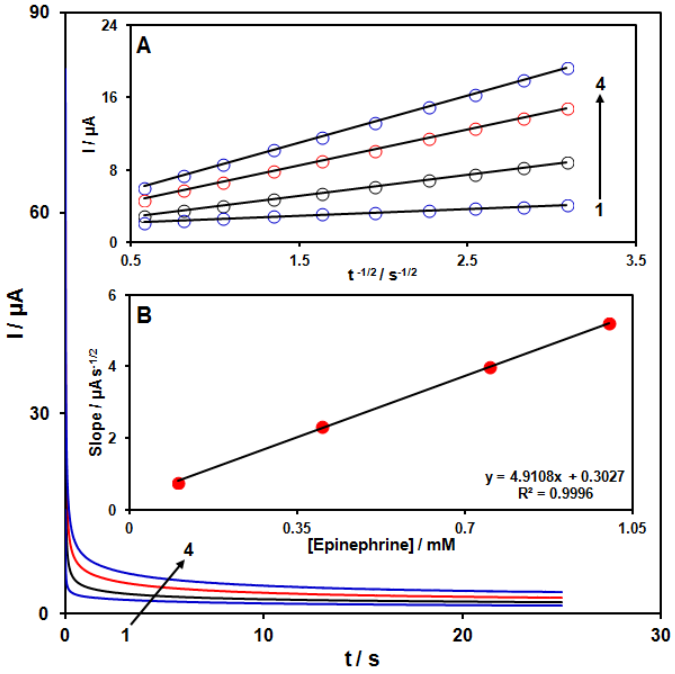

Fig. 4. Chronoamperograms obtained at ZnO-GO/SPE in 0.1 M PBS (pH 7.0) for different concentration of epinephrine. The numbers 1–4 correspond to 0.1, 0.4, 0.75 and 1.0 mM of epinephrine. Insets: (A) Plots of I *vs.* $t^{-1/2}$ obtained from chronoamperograms 1–4; (B) Plot of the slope of the straight lines against epinephrine concentration

$$I=nFAD^{1/2}C_b\pi^{-1/2}t^{-1/2}$$

where D and C_b are the diffusion coefficient (cm^2 s^{-1}) and the bulk concentration (mol cm^{-3}), respectively. Experimental plots of I *vs.* $t^{-1/2}$ were employed, with the best fits for different concentrations of epinephrine (Fig. 4A). The slopes of the resulting straight lines were then plotted *vs.* amitriptyline concentration (Fig. 4B). From the resulting slope and Cottrell equation the mean value of the D was found to be 2.0×10^{-6} cm^2/s.

3.3. Calibration plot and limit of detection

The peak current of epinephrine oxidation at the surface of the modified electrode can be used for determination of epinephrine in solution. Therefore, differential pulse voltammetry (DPV) experiments were done for different concentrations of epinephrine (Fig. 5). The oxidation peak currents of epinephrine at the surface of a modified electrode were

proportional to the concentration of the epinephrine within the ranges 0.5 to 500.0 µM. The detection limit (3σ) of epinephrine was found to be 7.0×10^{-7} M.

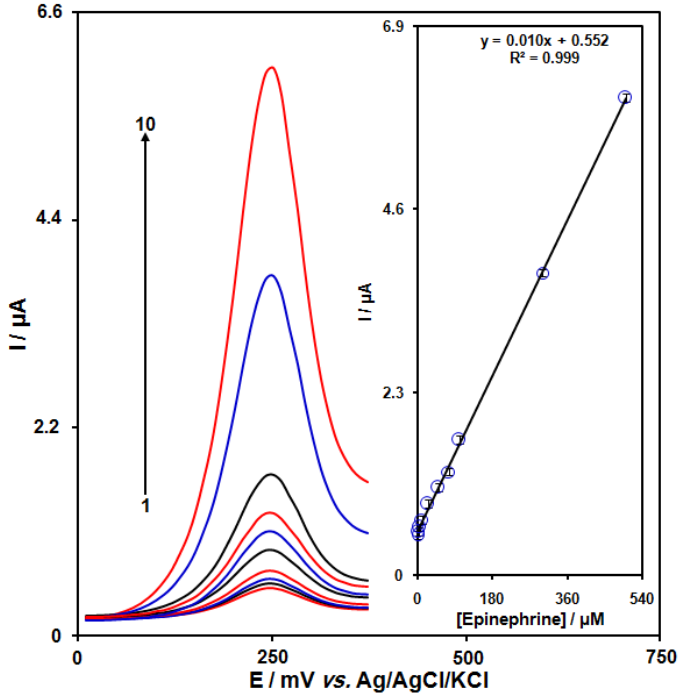

Fig. 5. DPVs of ZnO-GO/SPE in 0.1 M (pH 7.0) containing different concentrations of epinephrine. Numbers 1–15 correspond to 0.5, 2.5, 5.0, 10.0, 25.0, 50.0, 75.0, 100.0, 300.0 and 500.0 µM of epinephrine. Inset: plot of the electrocatalytic peak current as a function of epinephrine concentration in the range of 0.5-500.0 µM

3.4. Real sample analysis

In order to evaluate the analytical applicability of the proposed method, also it was applied to the determination of epinephrine in epinephrine injection. The results for determination of epinephrine in real samples are given in Table 1. Satisfactory recovery of the experimental results was found for epinephrine. The reproducibility of the method was demonstrated by the mean relative standard deviation (R.S.D.).

4. CONCLUSIONS

ZnO-GO nanocomposite modified graphite screen printed electrode acts as an efficient electrocatalyst for the determination of epinephrine in the presence of several potentially interfering substances in 0.1 M PBS (pH=7) by voltammetric method. These biomolecules were effectively catalyzed at less positive potentials yielding higher peak current. DPV studies exhibits the excellent sensitivity and selectivity towards the determination of epinephrine. The lowest detection limits of 7.0×10^{-7} M. (S/N=3) were achieved. The practical

applicability of the fabricated electrode was successfully validated by measuring the concentration of epinephrine in real samples.

REFERENCES

[1] M. D. Tezerjani, A. Benvidi, A. D. Firouzabadi, M. Mazloum-Ardakani, and A. Akbari, Measurement 101 (2017) 183.

[2] T. Kawano, F. X. Scheuermeyer, R. Stenstrom, B. H. Rowe, E. Grafstein, and B. Grunau, Resuscitation 112 (2017) 53.

[3] H. Nolte, T. B. Casale, R. F. Lockey, B. S. Fogh, A. Kaur, S. Lu, and H. S. Nelson, J. Allergy Clin. Immunol. 5 (2017) 84.

[4] A. J. Fuenmayor, M. I. Solórzano, and L. Gómez, Int. J. Cardiol. 220 (2016) 333.

[5] F. Wei, G. Xu, Y. Wu, X. Wang, J. Yang, L. Liu, P. Zhou, and Q. Hu, Sens. Actuators B Chem. 229 (2016) 38.

[6] S. Li, Q. Zhang, Y. Lu, D. Ji, D. Zhang, J. Wu, X. Chen, and Q. Liu, Sens. Actuators B 244 (2017) 290.

[7] J. Agrisuelas, M. I. Gonzalez-Sanchez, and E. Valero, Sens. Actuators B 249 (2017) 499.

[8] A. Dago, J. Navarro, C. Ariño, J. M. Díaz-Cruz, and M. Esteban, J. Chromatogr. A 1409 (2015) 210.

[9] M. Khairy, A. A. Khorshed, F. A. Rashwan, G. A. Salah, H. M. Abdel-Wadood, and C. E. Banks, Sens. Actuators B 239 (2017) 768.

[10] M. R. Akhgar, H. Beitollahi, M. Salari, H. Karimi-Maleh, and H. Zamani, Anal. Methods 4 (2012) 259.

[11] H. Beitollahi, J. B. Raoof, H. Karimi-Maleh, and R. Hosseinzadeh, J. Solid State Chem. 16 (2012) 1701.

[12] B. Jeyaraj, Anal. Bioanal. Electrochem. 5 (2013) 193.

[13] S. Mohammadi, H. Beitollahi, and A. Mohadesi, Sens. Lett. 11 (2013) 388.

[14] M. Mazloum-Ardakani, B. Ganjipour, H. Beitollahi, M. K. Amini, F. Mirkhalaf, H. Naeimi, and M. Nejati-Barzoki, Electrochim. Acta 56 (2011) 9113.

[15] R. Kurangalara, B. E. K. Swamy, M. Kumar, and C. Vishwanatha, Anal. Bioanal. Electrochem. 5 (2013) 555.

[16] M. M. Foroughi, H. Beitollahi, S. Tajik, M. Hamzavi, and H. Parvan, Int. J. Electrochem. Sci. 9 (2014) 2955.

[17] Z. Taleat, M. M. Ardakani, H. Naeimi, H. Beitollahi, M. Nejati, and H. R. Zare, Anal. Sci. 24 (2008) 1039.

[18] I. E. Mulazımoglu, E. Ozkan, and A.O. Solak, Anal. Bioanal. Electrochem. 3 (2011) 102.

[19] S. Tajik, M. A. Taher, and H. Beitollahi, Sens. Actuators B 197 (2014) 228.

[20] H. Beitollahi, J. B. Raoof, and R. Hosseinzadeh, Anal. Sci. 27 (2011) 991.
[21] P. Norouzi, S. Karamdoust, and M. R. Sohrabi, Anal. Bioanal. Electrochem. 3 (2011) 184.
[22] M. Baghayeri, H. Veisi, H. Veisi, B. Maleki, H. Karimi-Maleh, and H. Beitollahi, RSC Adv. 4 (2014) 49595.
[23] M. Mazloum-Ardakani, Z. Taleat, A. Khoshroo, H. Beitollahi, and H. Dehghani, Biosens. Bioelectron. 35 (2012) 75.
[24] Sh. Jahani, and H. Beitollahi, Anal. Bioanal. Electrochem. 8 (2016) 158.
[25] M. Khatami, S. Kharazi, Z. Kishani Farahani, H. Azizi, M.A. Lima Nobre, Tehran Univ. Med. J. 75 (2017) 72.
[26] M. M. Ardakani, Z. Taleat, H. Beitollahi, M. Salavati-Niasari, B. B. F. Mirjalili, and N. Taghavinia, J. Electroanal. Chem. 624 (2008) 73.
[27] M. P. Deepak, M. P. Rajeeva, G. P. Mamatha, Anal. Bioanal. Electrochem. 8 (2016) 931.
[28] M. Mazloum-Ardakani, H. Beitollahi, M. K. Amini, B. F. Mirjalili, and F. Mirkhalaf, J. Electroanal. Chem. 651 (2011) 243.
[29] M. Baghayeri, M. Namadchian, H. Karimi-Maleh, and H. Beitollahi, J. Electroanal. Chem. 697 (2013) 53.
[30] M. Soltani Nejad, G.H. Shahidi Bonjar, M. Khatami, A. Amini, and S. Aghighi, IET Dig. Lib. 11 (2017) 236.
[31] S. Tajik, M. A. Taher, and H. Beitollahi, Sens. Actuators B 188 (2013) 923.
[32] H. Beitollahi, M. M. Ardakani, H. Naeimi, and B. Ganjipour, J. Solid State Chem. 13 (2009) 353.
[33] G. Zhu, Z. Azizi, S. Pourseyedi, M. Khatami, and H. Mohammadi, J. Cluster Sci. 27 (2016) 1613.
[34] H.M. Moghaddam, H. Beitollahi, S. Tajik, Sh. Jahani, H. Khabazzadeh, and R. Alizadeh, Russ. J. Electrochem. 53 (2017) 452.
[35] S. Tajik, M. A. Taher, Sh. Jahani, and M. Shanesaz, Anal. Bioanal. Electrochem. 8 (2016) 899.
[36] J. Chen, P. He, H. Bai, H. Lei, K. Liu, F. Dong, and X. Zhang, J. Electroanal. Chem. 784 (2017) 41.
[37] F. Garkani Nejad, H. Beitollahi, and R. Alizadeh, Anal. Bioanal. Electrochem. 9 (2017) 134.
[38] H. Beitollahi, J. B. Raoof, H. Karimi-Maleh, and R. Hosseinzadeh, Anal. Bioanal. Electrochem. 4 (2012) 32.
[39] M. Khatami, R. Mehnipor, M.H. Sobhani Poor, and G. Salehi Jouzani, J. Cluster Sci. 27 (2016) 1061.

[40] S. Z. Mohammadi, H. Beitollahi, and H. Afzali, Anal. Bioanal. Electrochem. 8 (2016) 977.
[41] N. Y. Sreedhar, and K. Sunil, Anal. Bioanal. Electrochem. 6 (2014) 151.
[42] S. Li, Q. Zhang, Y. Lu, D. Ji, D. Zhang, J. Wu, X. Chen, and Q. Liu, Sens. Actuators B 244 (2017) 290.
[43] D. Kim, S. Lee, and Y. Piao, J. Electroanal. Chem. 794 (2017) 221.
[44] G. Yildiz, Z. Aydogmus, M. E. Cinar, F. Senkal, and T. Ozturk, Talanta. 173 (2017) 1.
[45] G. Zhu, J. Qian, H. Sun, X. Wu, K. Wang, and Y. Yi, J. Electroanal. Chem. 794 (2017) 126.
[46] J. Wang, B. Yang, J. Zhong, B. Yan, K. Zhang, C. Zhai, Y. Shiraishi, Y. Du, and P. Yang, J. Colloid Interface Sci. 497 (2017) 172.
[47] B. Sun, X. Gou, R. Bai, A. A. A. Abdelmoaty, Y. Ma, X. Zheng, and F. Hu, Mater. Sci. Eng. C 74 (2017) 515.
[48] M. Shamsipur, M. A. Tabrizi, Mater. Sci. Eng. C 45 (2014) 103.
[49] R. Jain, and A. Sinha, Appl. Surf. Sci. 369 (2016) 151.
[50] R. Sundarmurugasan, M. B. Gumpu, B. L. Ramachandra, N. Nesakumar, S. Sethuraman, U. M. Krishnan, and J. B. B. Rayappan, Sens. Actuators B 230 (2016) 306.
[51] G. Gasparotto, J. P. C. Costa, P. I. Costa, M. A. Zaghete, and T. Mazon, Mater. Sci. Eng. C 76 (2017) 1240.
[52] S. B. Khan, M. M. Rahman, A. M. Asiri, S. A. B. Asif, S. A. S. Al-Qarni, A. G. Al-Sehemi, S. A. Al-Sayari, and M. S. Al-Assiri, Phys. Low Dim. Syst. Nanostruct. 62 (2014) 21.
[53] M.R. Ganjali, F. Garkani Nejad, H. Beitollahi, Sh. Jahani, M. Rezapour, and B. Larijani, Int. J. Electrochem. Sci. 12 (2017) 3231.
[54] M. Maruthupandy, M. Anand, G. Maduraiveeran, S. Suresh, A. S. H. Beevi, and R. J. Priya, Adv. Natural Sci. 7 (2016) 045011.
[55] Y. Zheng, Z. Wang, F. Peng, and L. Fu, Braz. J. Pharm. Sci. 52 (2016) 781.
[56] E. Zare, S. Pourseyedi, M. Khatami, and E. Darezereshki, J. Mol. Struct. 1146 (2017) 96.
[57] A. J. Bard, and L. R. Faulkner, Electrochemical Methods Fundamentals and Applications, second ed, Wiley, New York (2001).