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Coupling of NiO Nanoparticles and Room Temperature Ionic Liquid for Fabrication of Highly Sensitive Voltammetric Sensor in Tryptophan Analysis

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Abstract- A NiO nanoparticle (NiO/NPs) ionic liquid (n-hexyl-3-methylimidazolium hexafluoro phosphate) modified carbon paste electrode (NiO/NPs/HMIHP/CPE) was developed for the analysis of tryptophan. The electrochemical behavior of tryptophan was investigated using cyclic voltammetry, chronoamperometry and square wave voltammetry (SWV). NiO nanoparticles synthesized by direct chemical precipitation method and characterized with scanning electron microscopy (SEM) and X-ray diffraction (XRD) methods. These studies revealed that the oxidation of tryptophan is facilitated at NiO/NPs/HMIHP/CPE. Using SWV, the method allowed determination of tryptophan in the linear working range of 0.08-350 μM with detection limit of 0.04 μM (S/N=3). The NiO/NPs/HMIHP/CPE was used for the determination of tryptophan in urine and water samples.

Keywords- Tryptophan analysis, NiO nanoparticle, Ionic liquid, Modified electrode

1. INTRODUCTION

Tryptophan is an essential amino acid needed for normal growth in infants and for nitrogen balance in adults and also an essential amino acid to help make niacin and serotonin in human body [1]. Some of food sample such as fish, milk, sesame seeds and etc. are rich of

tryptophan and necessary for body. Therefore, fabrication of novel sensor for fast and simple analysis of tryptophan is very important in food and biological sample. There are some analytical methods for measurement of tryptophan, including high performance liquid chromatography [2,3], fluorescence [4], flow-injection chemiluminescence [5] and electrochemical methods [6-10]. Electrochemical methods have more attention compare to other analytical methods due to high sensitivity, low cost, good selectivity and fast response [11-20].

Room temperature ionic liquids are the most rapidly growing in fabrication of electrochemical sensors in electrochemical analysis [21-25]. The structure of room temperature ionic liquids and its conductivity is extremely important for evaluating and selecting room temperature ionic liquids for electrochemical sensor modification [26-30]. Nanostructured materials and especially metal based nanoparticles are promising in the development of electrochemical sensors and biosensors due to highly specific surface area and high electrical conductivity [31-35]. According to the above points, it is predictable that coupling of room temperature ionic liquid and metal based nanoparticles can be help to improving electrical conductivity of modified electrode for trace level analysis [36-38].

Electrochemical voltammetric sensors based on these NiO nanostructures ionic liquid carbon paste electrode have been fabricated to detect tryptophan through electrochemical measurement. The electrochemical oxidation of tryptophan has been studied by cyclic voltammetry (CV), chronoamperometry (CA), and square wave voltammetry (SWV) in comparison with that of a carbon paste electrode. The proposed sensor was used for determination of tryptophan in urine and water samples.

2. EXPERIMENTAL SECTION

2.1. Apparatus and compounds

All of the voltammetric investigation performed using Autolab, potentiostat/galvanostat connected to a three-electrode cell, Azar electrode, linked with a computer (Pentium IV). Three-electrode cell assembly consisting of a platinum wire as an auxiliary electrode and an Ag/AgCl (KCl satˈd) electrode as a reference electrode was used.

Nickel(II) nitrate hexahydrate, tryptophan and HMIHP were procured from Sigma-Aldrich. Mineral oil and graphite powder (<50 µm) were procured from Merck. All solutions were prepared using double distilled water having a specific conductivity of 0.4–0.9 µS. Phosphate buffer solutions (0.1 M) (PBS) with different pH values were used.

2.2. Synthesis of NiO/NPs

To prepare the NiO/NPs, a 1.0 M of nickel nitrate hexahydrate and a 2.0 M aqueous solution of NaOH were prepared in distilled water. Then, the beaker containing NaOH

solution was heated at the temperature of about 30°C. The nitrate salt solution were added drop wise to the above heated solution under high-speed stirring. The beaker was sealed at this condition for 2 h. The precipitated $Ni(OH)_2$ were cleaned with deionized water and ethanol then calcined at 450 °C for 2.0 hours for synthesis of NiO/NPs.

2.3. Preparation of the electrode

NiO/NPs/HMIHP/CPE was prepared by mixing of 0.15 g of HMIHP, 0.85 g of the liquid paraffin, 0.05 g of NiO/NPs, and 0.95 g of graphite powder. Then the mixture was mixed well for 65 min until a uniformly wetted paste was obtained. A portion of the paste was filled firmly into one glass tube as described above to prepare NiO/NPs/HMIHP/CPE.

3. RESULTS AND DISCUSSION

3.1. NiO/NPs characterization

Fig. 1A shows XRD patterns of the synthesized NiO nanoparticle by chemical precipitation method.

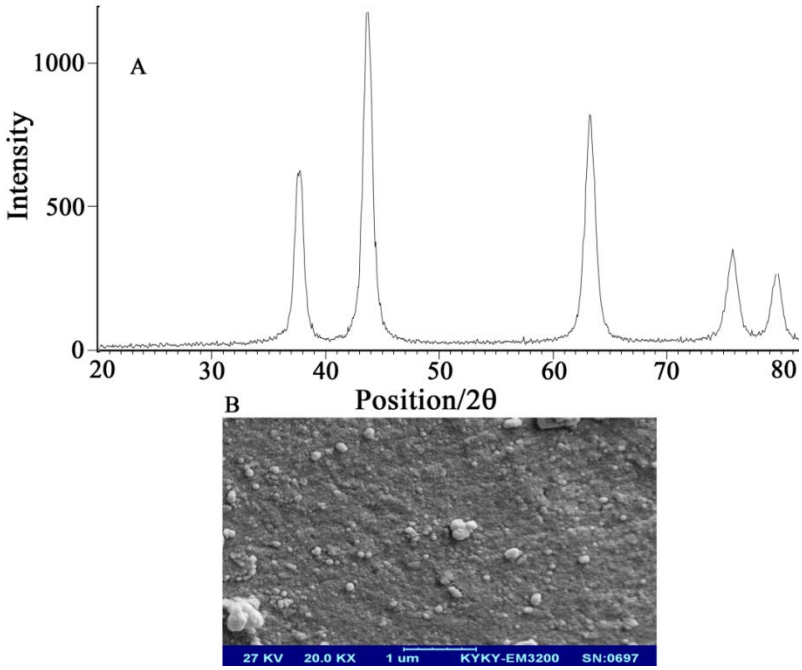

Fig. 1. (A) XRD patterns of as-synthesized NiO nanoparticles; (B) SEM image of as-synthesized NiO nanoparticles

The diffraction peaks relative to (111), (200), (220), (311) and (222) planes of NiO nanoparticle are presence in this figure, respectively. The mean particle size of NiO nanoparticles calculated by the Scherrer equation is about 20.1 nm. NiO (101) is used to

calculate the size of NiO nanoparticles. For more investigation, we used SEM micrographs for morphological characterization (Fig. 1B). The presence data confirm synthesis of NiO nanoparticle with recommend procedure.

3.2. Electrochemical investigation

The effect of pH value on the electrochemical behavior of tryptophan is performed in the range from 3.0 to 7.0 in 0.1 M PBS solutions (Fig. 2 insert). The maximum oxidation peak currents was obtained in pH=3.0.

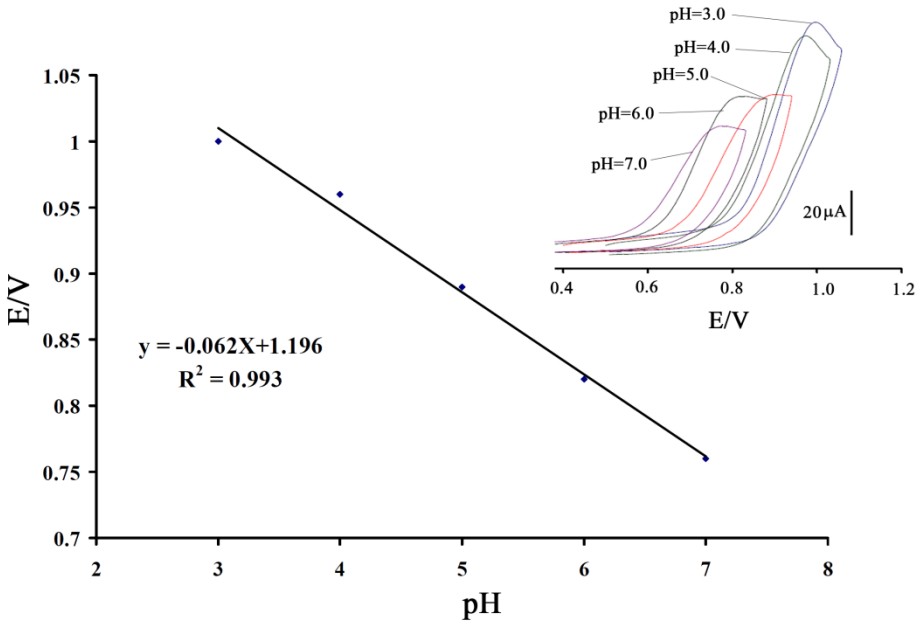

Fig. 2. Plot of potential, E, *vs*. pH for the electro-oxidation of 500 µM tryptophan at a surface of NiO/NPs/HMIHP/CPE. Inset: influence of pH on cyclic voltammograms of tryptophan at a surface of the modified electrode (pH 3–7, respectively)

Therefore, 0.1 M PBS with pH 3.0 is used as the supporting electrolyte in all subsequent experiments. The relationship between the oxidation peak potential of tryptophan and pH was also constructed. A linear shift of E_{pa} towards negative potential with an increasing pH can be obtained and obeyed the regression equation of E_{pa} (V)=-0.062 pH+1.196 (R^2=0.993), which indicates that protons are directly involved in the oxidation of tryptophan (Fig. 2). A slope of 62 mV/pH suggests that the number of electron transfer is equal to the proton number involved in the electrode reaction.

The electrochemical oxidation of tryptophan investigated at a surface of different electrodes in the presence of 800.0 µM tryptophan (Fig. 3). As shown in Fig. 3, (a) NiO/NPs/HMIHP/CPE, (b) HMIHP/CPE, (c) NiO/NPs/CPE, (d) CPE. Strong dependency

upon the increasing of dropped different materials are shown peak current for the tryptophan oxidation processes.

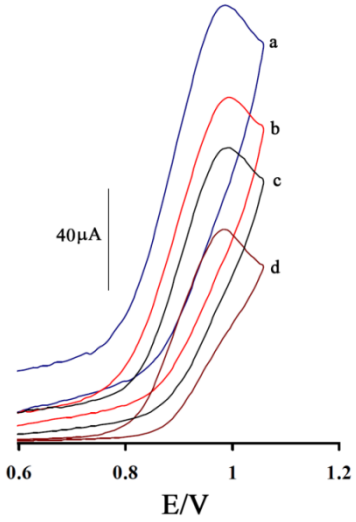

Fig. 3. Cyclic voltammograms of (a) NiO/NPs/HMIHP/CPE, (b) HMIHP/CPE, (c) NiO/NPs/CPE and (d) CPE in the presence of 800 μM tryptophan at pH 3.0, respectively

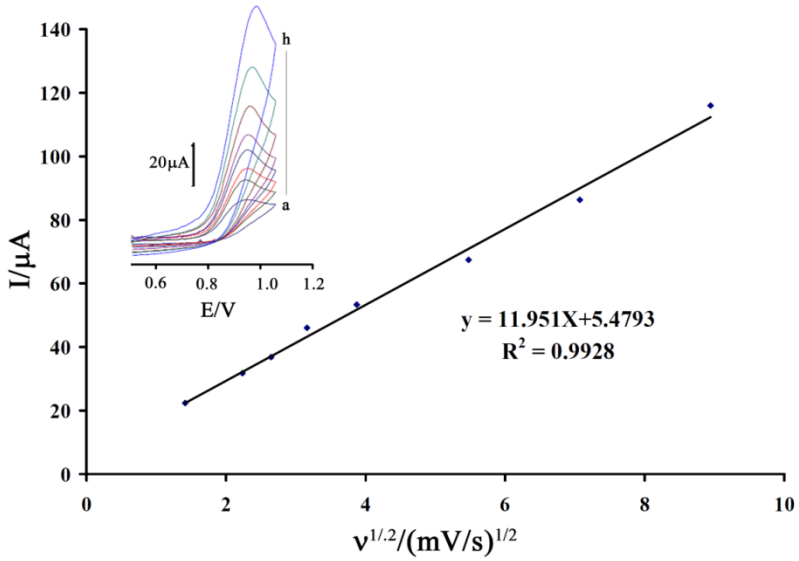

Fig. 4. Plot of I_{pa} versus $\nu^{1/2}$ for the oxidation of tryptophan at NiO/NPs/HMIHP/CPE. Inset shows cyclic voltammograms of tryptophan at NiO/NPs/HMIHP/CPE at different scan rates (from inner to outer) of 2, 5, 7, 10, 15, 30.0, 50.0 and 80.0 mV s^{-1} in 0.1 M phosphate buffer, pH 3.0

The small anodic peak can be observed at CPE (curve d). After modification with NiO/NPs, it is found that the oxidation current increased (curve c), indicating the NiO/NPs can increase the conductibility of electrode and then increase the oxidation current for

tryptophan. Upon adding HMIHP in the electrode surface, the cyclic voltammogram exhibits a stable oxidation peak and the oxidation current increased (curve b). The reason is that HMIHP as a conductive binder can increase the effective surface area and enhance the oxidation peak current for tryptophan.

The effect of scan rate on the electro-oxidation of tryptophan (550.0 µM) was also studied. The peak currents (I_p) increased and the anodic peak potentials shifted to more positive directions by an increase in the scan rate (Fig. 4).

It can be proposed that a kinetic limitation in the reaction between the redox sites of the NiO/NPs/HMIHP/CPE and tryptophan. Moreover, the linear dependency of anodic oxidation peak currents versus the square root of the scan rate ($v^{1/2}$) indicated that the electrode reaction is a diffusion controlled process [39-41].

A Tafel plot (Fig. 5) was developed for obtaining the information on the rate-determine step according to the data from the rising part of the current-voltage curve at a scan rate of 7 mV/s for 550 µM tryptophan.

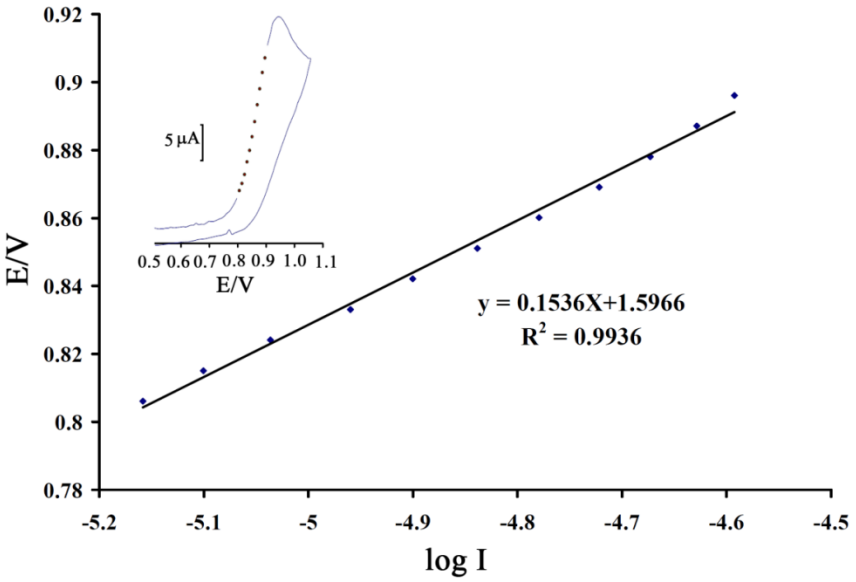

Fig. 5. Tafel plot for NiO/NPs/HMIHP/CPE in 0.1 M PBS (pH 3.0) with a scan rate of 7 mV s^{-1} in the presence of 550 µM tryptophan

The slope of the Tafel plot was equal to n(1-α) F/2.303 RT, and in this work, the Tafel slope was found to be 0.1536 V $decade^{-1}$. Then, n(1-α) was calculated to be 0.38. Chronoamperometry method was used for the determination of the diffusion coefficient (D) of tryptophan, (Fig. 6 A). By employing chronoamperometry, the plot of I_p *vs*. the $t^{-1/2}$ shows a linear relationship. According to the integrated slop Cottrell equation, the diffusion

coefficient can be estimated (Fig. 6 B). The value of D was found to be 4.9×10^{-5} cm^2/s using Cottrell equation.

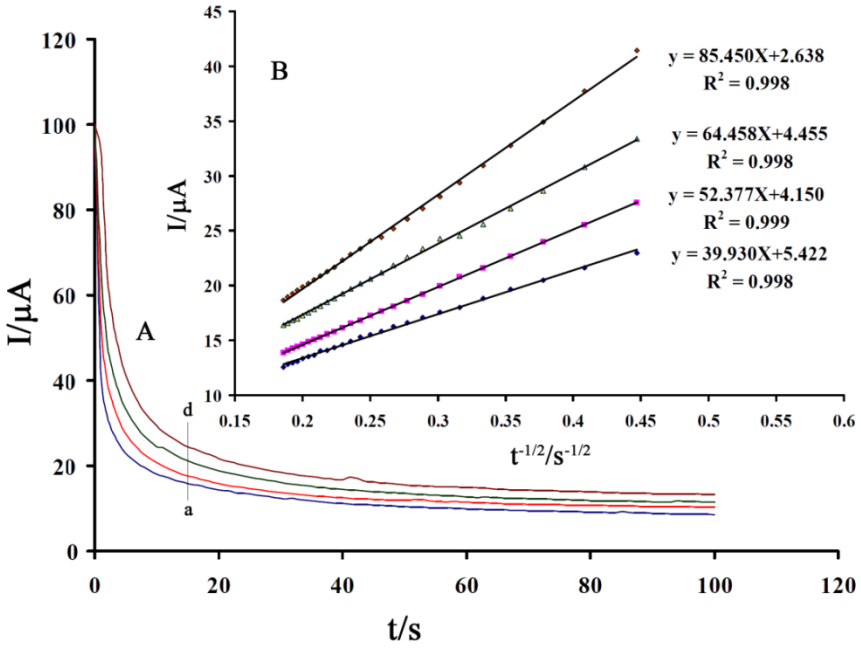

Fig. 6. (A) Chronoamperograms obtained at NiO/NPs/HMIHP/CPE in the presence of (a) 400; (b) 500; (c) 600 and (d) 700 μM tryptophan in the buffer solution (pH 3.0). (B) Cottrell's plot for the data from the chronoamperograms

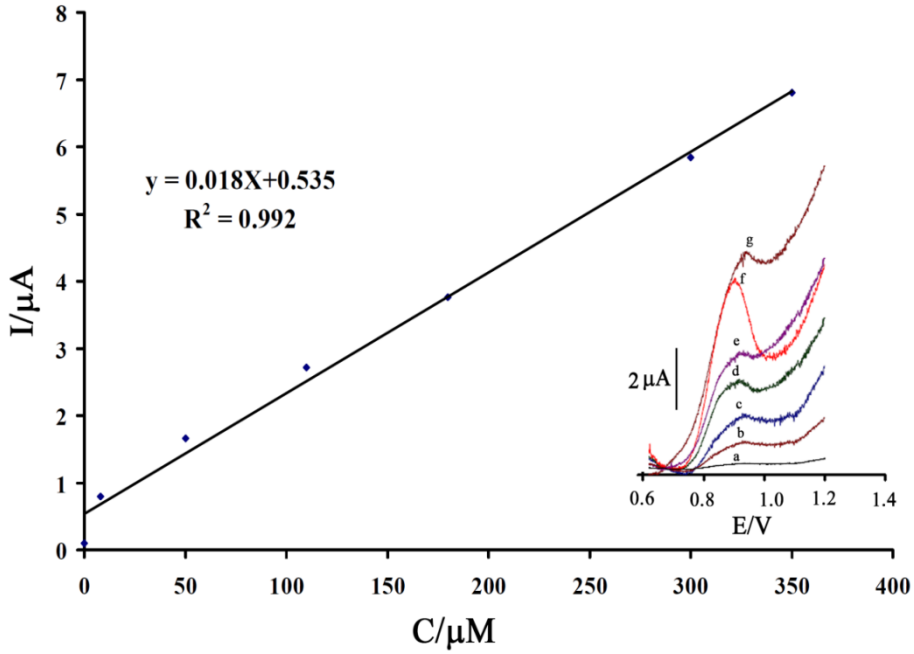

Fig. 7. The plots of the electro-oxidation peak current as a function of tryptophan concentration. Inset; SWVs of NiO/NPs/HMIHP/CPE in 0.1 MPBS (pH 3.0) containing different concentrations of tryptophan in μM; a–g: 0.08; 8.0; 50.0; 110.0; 180.0; 300.0 and 350.0

3.3. Linear dynamic range and limit of detection for tryptophan

The peak currents of the electrochemical sensor increases with the increasing of tryptophan concentrations, and the resulting calibration plots are a good linear over the range from 0.08 µM to 350 µM using square wave voltammetric sensor (Fig. 7). The detection limit is estimated to be 0.04 µM at a signal-to-noise ratio of 3σ (where σ is the standard deviation of the blank, n=10).

3.4. Selectivity of the propose voltammetric sensor

In continuous to evaluate the selectivity of NiO/NPs/HMIHP/CPE in the determination of tryptophan the influence of different foreign species on the determination of 50.0 µM tryptophan was investigated. Tolerance limit was taken as the maximum concentration of foreign substances that caused an approximate relative error of ±5%. The obtained results are shown in Table 1. The results in Table 1 demonstrate that NiO/NPs/HMIHP/CPE has a good selectivity for tryptophan analysis.

Table 1. Interference study for the determination of 50.0 µM tryptophan under optimized conditions (pH 3.0)

Species	**Tolerance limits** ($W_{species}/W_{GSH}$)
Alanine, Phenyl alanine, L-Threonine, and L-Isoleucine, Sucrose, Lactose and Fructose	1000
Urea,	500
Starch	Saturation

3.5. Determination of tryptophan in spiked urine and water samples

To prepare the spiked water and urine samples, known concentration of tryptophan solution was added into water and urine samples. The recovery and relative standard deviation of tryptophan was determined and presented in Table 2. The excellent recovery of the samples indicated that the proposed sensor can be successfully applied in the detection of tryptophan concentration in water and urine sample.

4. CONCLUSION

In this study, the HMIHP modified NiO/NPs carbon paste electrode was used to investigate the electrochemical behaviors of tryptophan. The NiO/NPs/HMIHP/CPE showed

great improvement to the electrode process of tryptophan compare to CPE. The electrode was successfully used for the determination of tryptophan in water and urine samples.

Table 2. Determination of tryptophan in water and urine samples (n=5)

Sample	Added (µM)	Expected (µM)	Founded (µM)	Recovery (%)
Urine samples	––	––	<LOD	<LOD
	15.00	15.00	14.85±0.74	99.0
Water samples	––	––	<LOD	<LOD
	20.00	20.00	20.68±0.78	103.4

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REFERENCES

[1] J. B. Raoof, R. Ojani, and H. Karimi-Maleh, Electroanalysis 20 (2008) 1259.
[2] M. E. Soto, A. M. Ares, J. Bernal, M. J. Nozal, and J. L. Bernal, J. Chromatogr. A 1218 (2011) 7592.
[3] J. Yang, S. Wang, J. Liu, and A. Raghani, J. Chromatogr. A 1156 (2007) 174.
[4] H. Iizuka, K. Ishii, Y. Hirasa, K. Kubo, and T. Fukushima, J. Chromatogr. B 879 (2011) 3208.
[5] Y. D. Liang, and J. F. Song, J. Pharm. Biomed. Anal. 38 (2005) 100.
[6] C. Li, Y. Ya, and G. Zhan, Coll. Surf. B 76 (2010) 340.
[7] G. P. Jin, and X. Q. Lin, Electrochem. Commu. 6 (2004) 454.
[8] S. Shahrokhian, and L. Fotouhi, Sens. Actuators B 123 (2007) 942.
[9] H. Karimi-Maleh, M. Salimi-Amiri, F. Karimi, M. A. Khalilzadeh, and M. Baghayeri, J. Chem. 2013 (2013) 7.
[10] R. Moradi, S. A. Sebt, H. Karimi-Maleh, R. Sadeghi, F. Karimi, A. Bahari, and H. Arabi, Phys. Chem. Chem. Phys. 15 (2013) 5888.
[11] H. Karimi-Maleh, P. Biparva, and M. Hatami, Biosens. Bioelectron. 48 (2013) 270.
[12] H. Karimi-Maleh, F. Tahernejad-Javazmi, A.A. Ensafi, R. Moradi, S. Mallakpour, and H. Beitollahi, Biosens. Bioelectron. 60 (2014) 1.
[13] A. A. Ensafi, and H. Karimi-Maleh, J. Electroanal. Chem. 640 (2010) 75.

[14] M. R. Shahmiri, A. Bahari, H. Karimi-Maleh, R. Hosseinzadeh, and N. Mirnia, Sens. Actuators B 177 (2013) 70.
[15] A. A. Ensafi, H. Karimi-Maleh, S. Mallakpour, and M. Hatami, Sens. Actuators B 155 (2011) 464.
[16] H. Beitollah, M. Goodarzian, M. A. Khalilzadeh, and H. Karimi-Maleh, J. Mol. Liq. 173 (2012) 137.
[17] E. Afsharmanesh, H. Karimi-Maleh, A. Pahlavan, and J. Vahedi, J. Mol. Liq. 181 (2013) 8.
[18] H. Yaghoubian, H. Karimi-Maleh, M. A. Khalilzadeh, F. Karimi, Int. J. Electrochem. Sci. 4 (2009) 993.
[19] M. Elyasi, M. A. Khalilzadeh, and H. Karimi-Maleh, Food Chem. 141 (2013) 4311.
[20] M. Najafi, M. A. Khalilzadeh, and H. Karimi-Maleh, Food Chem. 158 (2014) 125.
[21] M. Bijad, H. Karimi-Maleh, and M. A. Khalilzadeh, Food Anal. Methods 6 (2013) 1639.
[22] Y. Zhao, Y. Gao, D. Zhan, H. Liu, Q. Zhao, Y. Kou, Y. Shao, M. Li, Q. Zhuang, and Z. Zhu, Talanta 66 (2005) 51.
[23] T. Y. Kim, H. W. Lee, M. Stoller, D. R. Dreyer, C. W. Bielawski, R. S. Ruoff, and K. S. Suh, ACS Nano 5 (2011) 436.
[24] G. C. Zhao, M. Q. Xu, J. Ma, and X. W. Wei, Electrochem. Commun. 9 (2007) 920.
[25] W. Sun, M. Yang, and K. Jiao, Anal. Bioanal. Chem. 389 (2007) 1283.
[26] T. Jamali, H. Karimi-Maleh, and M. A. Khalilzadeh, LWT-Food Sci. Technol. 57 (2014) 679.
[27] A. Baghizadeh, H. Karimi-Maleh, Z. Khoshnama, A. Hassankhani, and M. Abbasghorbani, Food Anal. Methods 8 (2015) 549.
[28] T. Tavana, M. A. Khalilzadeh, H. Karimi-Maleh, A. A. Ensafi, H. Beitollahi, and D. Zareyee, J. Mol. Liq. 168 (2012) 69.
[29] A. Safavi, N. Maleki, O. Moradlou, and F. Tajabadi, Anal. Biochem. 359 (2006) 224.
[30] W. Sun, Y. Li, M. Yang, S. Liu, and K. Jiao, Electrochem. Commun. 10 (2008) 298.
[31] H. Karimi-Maleh, M. Moazampour, M. Yoosefian, A. L. Sanati, F. Tahernejad-Javazmi, and M. Mahani, Food Anal. Methods 7 (2014) 2169.
[32] B. J. Sanghavi, and A. K. Srivastava, Electrochim. Acta 56 (2011) 4188.
[33] B. J. Sanghavi, S. Sitaula, M. H. Griep, S. P. Karna, M. F. Ali, and N. S. Swami, Anal. Chem. 85 (2013) 8158.
[34] B. J. Sanghavi, and A. K. Srivastava, Electrochim. Acta 55 (2010) 8638.
[35] M. L. Yola, N. Atar, T. Eren, H. K. Maleh, and S. Wang, RSC Adv. 5 (2015) 65953.
[36] R. Bavandpour, H. Karimi-Maleh, M. Asif, V. K. Gupta, N. Atar, and M. Abbasghorbani, J. Mol. Liq. 213 (2016) 369.
[37] S. Cheraghi, M. A. Taher, and H. Karimi-Maleh, Electroanalysis 28 (2016) 366.

[38] W. Sun, R. Gao, and K. Jiao, J. Phys. Chem. B 111 (2007) 4560.
[39] H. Karimi-Maleh, K. Ahanjan, M. Taghavi, and M. Ghaemy, Anal. Methods 8 (2016) 1780.
[40] V. Arabali, M. Ebrahimi, M. Abbasghorbani, V.K. Gupta, M. Farsi, M.R. Ganjali, and F. Karimi, J. Mol. Liq. 213 (2016) 312.
[41] S. Gheibi, H. Karimi-Maleh, M. A. Khalilzadeh, and H. Bagheri, J. Food Sci. Technol. 52 (2015) 276.