Anal. Bioanal. Electrochem., Vol. 11, No. 1, 2019, 1-18

Analytical &
Bioanalytical
Electrochemistry

2019 by CEE
www.abechem.com

Full Paper

Fe_3O_4@MCM-48-SO_3H/ Multi-Wall Carbon Nanotubes Composite Modified Glassy Carbon Electrode: an Efficient Sensor for Sensitive and Selective Simultaneous Determination of Serotonin and Sertraline in the presence of Uric Acid

Ali Babaei,[1,2,*] Mohammad Afrasiabi[1] and Akbar Yousefi[1]

[1]*Department of Chemistry, Faculty of Science, Arak University, Arak, I.R. Iran*
[2]*Institute of Nanosciences & Nanotechnology, Arak University, Arak, I.R. Iran*

*Corresponding Author, Tel.: +98 863 4173401; Fax: +98 863 4173406
E-Mail: ali-babaei@araku.ac.ir

Received: 22 May 2018 / Received in revised form: 15 December 2018 /
Accepted: 21 December 2018 / Published online: 31 January 2019

Abstract- In this study a novel electrode was developed to simultaneous determination of serotonin (5-HT) and sertraline (ST) in the presence of uric acid based on a glassy carbon electrode modified by a composition of Fe_3O_4@MCM-48-SO_3H and multi-wall carbon nanotubes (MWCNTs) (Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE). The results showed that the modified electrode is a promising one for the simultaneous determination of 5-HT and ST in the presence of uric acid (UA) at optimized circumstances. Cyclic voltammetry (CV) and differential pulse voltammetry (DPV) techniques were used to study behavior of the modified electrode. The DPV results showed a linear relationship between 5-HT concentration and the oxidation peak current in the concentration ranges from 0.05 µM to 100 µM with a detection limit of 0.015 µM. For ST, a linear relationship between the concentration and the oxidation peak current in the ranges from 0.1 µM to 85 µM was obtained. The corresponding detection limit for ST was 0.025 µM. The Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE was used for determination of 5-HT and ST in real samples like human blood serum and urine with reasonable outcomes.

Keywords- Serotonin, Sertraline, Multi-Walled Carbon Nanotubes, Fe_3O_4@MCM-48-SO_3H, Electrochemical Sensor

---

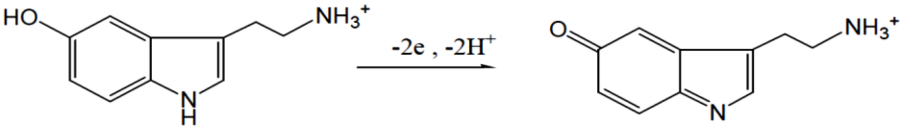
1. INTRODUCTION

Carbon nanotubes (CNTs), was detected in 1991, present in two forms of single-walled CNTs (SWCNTs) and multi-walled CNTs (MWCNTs). These compounds have recently attracted the attention of a great deal of scientists in various fields of nanotechnology because they have their sole properties [1]. The outstanding properties of CNTs led to the wide usage of these compounds in our life [2,3]. In addition, regarding the mentioned properties, CNTs have widely been utilized for the creation of various electrochemical sensors [4-10] with elevated sensitivity, high chemical permanence, wide potential window with option of compact size and small background current.

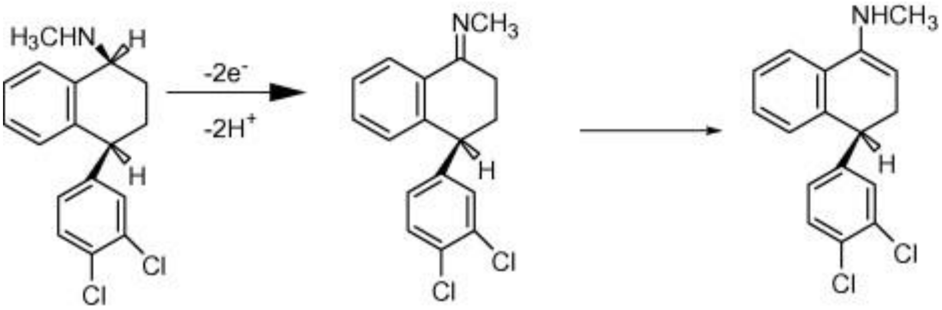
MCM are series mesoporous materials that were first synthesized by Mobil's research center. Two popular porous adsorbents can be found in these materials. The first one is defined as MCM-41 and it has a hexagonal arrangement of unidirectional pores. On the other hand, the second one is named MCM-48 and its structure is cubic indexed in the space group Ia3d (recently modeled as a gyroid minimal surface) [11]. Both MCM-41 and MCM-48 have shown some specific properties; like great surface area, pore size with controllability, high porous surface and arranged consistent hole structure; which have made them suitable materials for constructing electrochemical sensors [12,13]. However, MCM-48 seems to be a better absorbent in division methods (like supercritical fluid chromatography) and a better catalyst carrier than MCM-41 better activity because of having higher specific area (up to 1600 cm^2 g^{-1}), higher specific pore volume (up to 1.2 cm^3 g^{-1}) and higher thermal stability [14]. Application of Fe_3O_4@MCM-48-SO_3H nanoparticles has two advantages as catalytic effect of Fe_3O_4 and selectivity of negatively charged sulfonic acid group in the nanoparticles.

Serotonin (5-HT) which is produced in serotonergic neurons (located in the central nervous system) is a very important neurotransmitter for clinical purposes. Along with other monoamine transmitters, this neurotransmitter shows a major contribution in the emotional systems like regulation of mood, sleep, emesis (vomiting), sexuality and appetite. In other word, low levels of 5-HT can lead to several disorders (notably depression, migraine, bipolar disorder and anxiety) [15,16]. Regarding the information mentioned above, it is crucial for us to introduce practical methods for determining the amount of 5-HT. Several methods have been reported for this purpose in the literature. Some of these methods can be mentioned as high performance liquid chromatography coupled to electrospray mass spectrometry (HPLC- MS) [17], solid surface-room temperature phosphorescence (SS-RTP) [18], spectrophotometry [19], fluorometry [20], enzyme immunoassay [21], radioimmunoassay [22] and capillary electrophoresis (CE) [23]. Besides, since 5-HT can be oxidized electrochemically (Scheme 1), electrochemical determination of 5-HT have been reported in some articles [24-33].

Sertraline (ST) is a drug from 5-HT reuptake inhibitor (SSRI) family. SSRIs act as antidepressant drugs and it works by restoring the balance of 5-HT in the brain. ST can increase the level of energy and feelings of well-being in people's body [34]. Therefore simultaneous determination of 5-HT and ST could be important issue. According to the literature, several analytical methods have been reported for measuring the amount of ST; including gas chromatography, GC–MS, HPLC, capillary electrophoresis and some assembled techniques [35–41]. ST is also oxidized electrochemically (Scheme 2) so a few electrochemical studies have been reported on determination of ST [42-44].



Scheme 1. Oxidation mechanism of Serotonin (5-HT)



Scheme 2. Oxidation mechanism of sertraline (ST)

The aim of this work was to develop a simple, sensitive and selective electrochemical sensor for the simultaneous determination of 5-HT and ST in presence of UA. Therefore GCE modified with Fe_3O_4@MCM-48-SO_3H/MWCNTs nanocomposite was used. To the best of our knowledge this is the first report on simultaneous electrochemical determination of 5-HT and ST. Finally the modified electrode was successfully applied for determination of 5-HT and ST in human blood serum and urine.

2. EXPERIMENTAL

2.1. Chemicals and solutions

The most of chemicals were analytical grade and were utilized without further purification. 5-HT and Sertraline hydrochloride (ST) were bought from Sigma-Aldrich Chemical Corporation. MWCNTs (purity > 95%) were bought from Plasma Chem GmbH Company. The stock standard solutions of 5 mM 5-HT and 1 mM ST were freshly prepared in 0.1 M phosphate buffer solution (PBS) with pH equal to 7. Different concentrations of analytes were prepared via diluting the stock solutions with 0.1 M PBS (pH=7). All electrochemical experiments on 5-HT and ST were carried out in 0.1 M PBS (pH=7), otherwise is stated. In order to prepare solutions for experiments, triply distilled water was used.

2.2. Instrumentation

AUTOLAB 30V device (ECO CHEMIE) was used as the potentiostat. Three electrode system include modified GCE as the working electrode, an Ag/AgCl/3M KCl electrode as the reference electrode and a platinum wire as an auxiliary electrode were used. The potentiostat was controlled by GPES software version 4.9. In order to measure pH, Metrhom 744 instrument was used. MIRA3-TESCAN field emission scanning electron microscope (FESEM) was applied to measure morphological features of the nanocomposite.

2.3. Synthesis of Fe_3O_4@MCM-48-SO_3H nanoparticles

Fe_3O_4@ MCM-48- SO_3H nanoparticles was synthesized as previously reported [45]. Briefly, 1.5 g of MNPs.Fe_3O_4 and 5 ml of ammonia solution were added to 50 ml of distilled water in a glass beaker, and sonicated for 2 min at 40 °C. After obtaining a stable suspension, 10 ml of TEOS, 0.9 g of NaOH and 0.19 g of NaF were added to it and the mixture was stirred for 2 h at 25 ºC. Then, 7.0 g of acetyltriamonium bromide was added to the mixture and it was stirred for another 2 h at 40 °C. Next, the obtained product was placed in an autoclave at 120 °C under hydrothermal conditions for two days. The resulting solid was filtered and washed with distilled water and dried at 60 °C. Finally, the calcination of the synthesized magnetic particles was carried out for 3 h at 300 °C to remove the template. In the next step, Fe_3O_4@MCM-48 was functionalized with SO_3H groups by adding 20 ml of aqueous solution of $NaHSO_4.H_2O$ to 1.5 g of mesoporous Fe_3O_4@MCM-48 nanoparticles, sonicating the resulting mixture for 1 min and then stirring it for 30 min at 25 °C. Finally, the nanoparticles were washed with water and put into the oven (at 90 °C) for 2h. The obtained brown solid was Fe_3O_4@MCM-48-SO_3H.

2.4. Preparation of Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE

DPV analysis was used to obtain the optimum composition ratio of MWCNTs to Fe_3O_4@MCM-48-SO_3H nanoparticles for the modification of a GCE. The results showed that not only the sensitivity of the sensor was greatly affected by the amount of Fe_3O_4@MCM-48-SO_3H in the modifier matrix but also the amount of these nanoparticles could improve the selectivity of the modified electrode respect to interference of UA. Thus, different composition ratios of MWCNTs to Fe_3O_4@MCM-48-SO_3H were prepared and the optimum composition ratio of Fe_3O_4@MCM-48-SO_3H was obtained to be 12% for the fabrication of the modified electrode (Fig. 1). In order to prepare a stock solution of Fe_3O_4@MCM-48-SO_3H/MWCNTs in dimethyl formamide (DMF), 1.76 mg of MWCNTs and 0.24 mg of Fe_3O_4@MCM-48-SO_3H (12%:88% w/w) were sonicated in 2 mL of DMF until a homogeneous suspension was achieved. The modified electrode was obtained by casting 20 µL of the obtained dissolution on the bare electrode with a micro syringe. Then, the fresh modified electrode was left to dry at ambient temperature. Finally, the fabricated Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE was put in the electrochemical compartment including 0.1 M of PBS and then 10 cycles in the potential window of 0.0 to 1.0 V were applied using CV technique to gain stable responses.

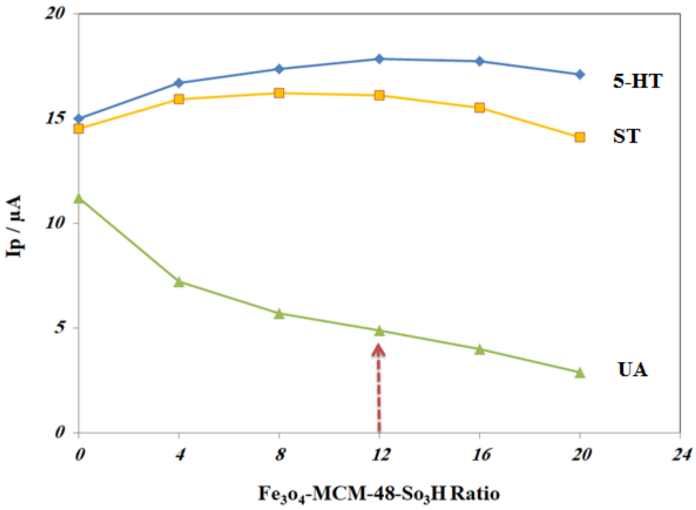


Fig. 1. Effect of the ratio of Fe_3O_4@MCM-48-SO_3H nanoparticles for modification of a GCE for simultaneous determination of 20 µM 5-HT and 20 µM ST in the presence of 20 µM of UA at Fe_3O_4@MCM-48-SO_3H/ MWCNTs/GCE in 0.1M PBS (pH=7.0).

2.5. General procedure

The following method was typically employed for obtaining the voltammograms. 10 ml of the sample solutions were poured in a voltammetric cell to carry out electrochemical experiments. The sample solutions contained 0.1 M of PBS with pH of 7 and then appropriate amounts of 5-HT and ST were added to the solution. The oxidation of 5-HT and ST were performed on the surface of the modified electrode at +0.1 and +0.8 V, respectively. The corresponding oxidation peak current peaks were proportional to the concentration of 5-HT and ST, so these peaks were used to obtain calibration curves for 5-HT and ST. The surface of the electrode should be cleaned before each experiment. Thus, after finishing each experiment, the surface of the electrode was washed thoroughly with triply distilled water and NaOH solution (0.2%) to remove all adsorbates from its surface. Fresh human serum samples were available from Razi Institute of vaccine and serum Company (Tehran, Iran). In order to determine spiked 5-HT and ST in real samples, human serum and urine samples were filtered and diluted 50 times using a 0.1 M PBS with pH of 7.

3. RESULTS AND DISCUSSION

3.1. Characterization of Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE

Figs. 2a and 2b illustrate the SEM images of the MWCNTs and Fe_3O_4@MCM-48-SO_3H NPs, respectively.

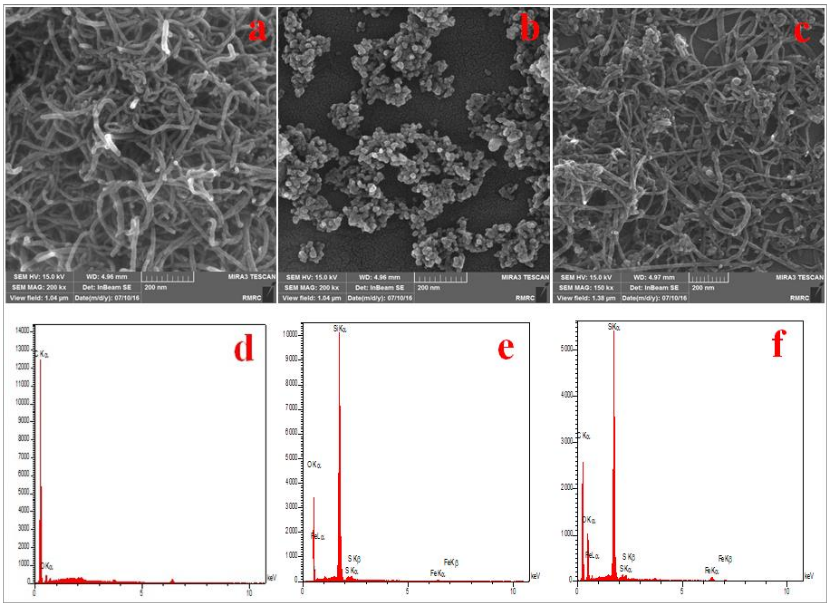


Fig. 2. FESEM micrographs of the samples: (a) MWCNTs; (b) Fe_3O_4@MCM-48-SO_3H NPs and (c) Fe_3O_4@MCM-48-SO_3H/MWCNTs and SEM-EDX spectrum of the samples: (d) MWCNTs; (e) Fe_3O_4@MCM-48-SO_3H NPs and (f) Fe_3O_4@MCM-48-SO_3H/MWCNTs

According to the SEM results, the size of Fe_3O_4@MCM-48-SO_3H nanoparticles was between 20 to 50 nm. SEM image of the Fe_3O_4@MCM-48-SO_3H/MWCNTs composite (Fig. 2c) showed that the surfaces of Fe_3O_4@MCM-48-SO_3H NPs were covered uniformly with MWCNTs. The porosity of Fe_3O_4@MCM-48-SO_3H/MWCNTs results in an increase in the surface area of the composite so the electrode modified with this composite was supposed to show higher electrical response than the unmodified electrode. The EDX analysis of the MWCNTs (Fig. 2d) verified the attendance of C in the construction of the compound. The peaks related to presence of Fe, O, S and Si elements were seen in the EDX analysis of Fe_3O_4@MCM-48-SO_3H NPs (Fig. 2e). As can be seen in Fig. 2f, the peaks corresponding to

Fe, O, S, C and Si elements have appeared in the EDX analysis of Fe_3O_4@MCM-48-SO_3H/MWCNTs, proving that the components were successfully mixed in the nanocomposite.

Electrochemical impedance spectroscopy (EIS) can be mentioned as another technique used for characterizing modified electrodes. EIS technique can provide some information about charge transfer resistance (R_{ct}) of the studied electrode surface. Fig. 3 shows the Nyquist plots obtained from the bare GCE (Fig. 3a), MWCNTs/GCE (Fig. 3b) and Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE (Fig. 3c) electrodes. To obtain EIS results the electrodes were immersed in the solution of 5 mM of $K_3[Fe(CN)_6]$ and 5 mM of $K_4[Fe(CN)_6]$ which contain 0.1 M of KCl. The R_{ct} for the electrodes were obtained from the Corresponding Nyquist diagrams. According to the results R_{ct} were obtained to be 0.19 and 0.37 kΩ for Fe_3O_4@MCM-48-SO_3H/MWCNTs/ GCE and MWCNTs/GCE, respectively. These amounts are much lower than that of the unmodified GCE (3.03 kΩ).

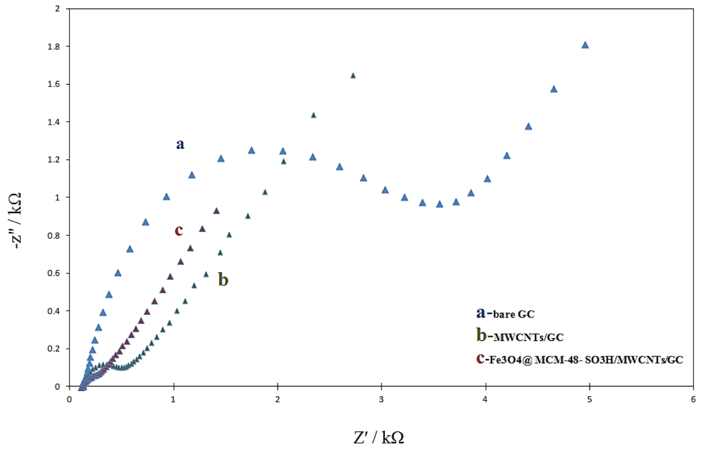


Fig. 3. Nyquist plots for (A) bare GCE, (B) MWCNTs/GCE and (C) Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE obtained in solution of 0.1 M KCl, 5 mM $K_3[Fe(CN)_6]/K_4[Fe(CN)_6]$

The results showed that the application of Fe_3O_4@MCM-48-SO_3H/MWCNTs nanocomposite led to lower charge transfer resistance on the electrode surface in comparison with MWCNTs. Therefore the use of Fe_3O_4@MCM-48-SO_3H/MWCNTs composite at the surface of GCE is preferred due to its lower electrochemical resistance.

The effect of modifying the electrode on the active surface area was investigated by CV of 4 mM of potassium hexacyanoferrate(II) ($K_4[Fe(CN)_6]$) in 0.1 M of PBS (pH 7.0) at GCE, MWCNTs/GCE and Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE (Fig. 4). $K_4[Fe(CN)_6]$ showed a couple of reversible redox peaks at bare and modified GCEs. In addition, the redox peak currents for the modified electrodes were bigger than those for the unmodified GCE. When the unmodified GCE was employed, the potential of the redox peak related to $K_4[Fe(CN)_6]$ shifted by increasing scan rates so we can say that the redox behavior of this compound was like a quasi-reversible system. In contrast, when the analysis was carried out at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE, increasing scan rates didn't lead to shifting the potential of the corresponding redox peaks.

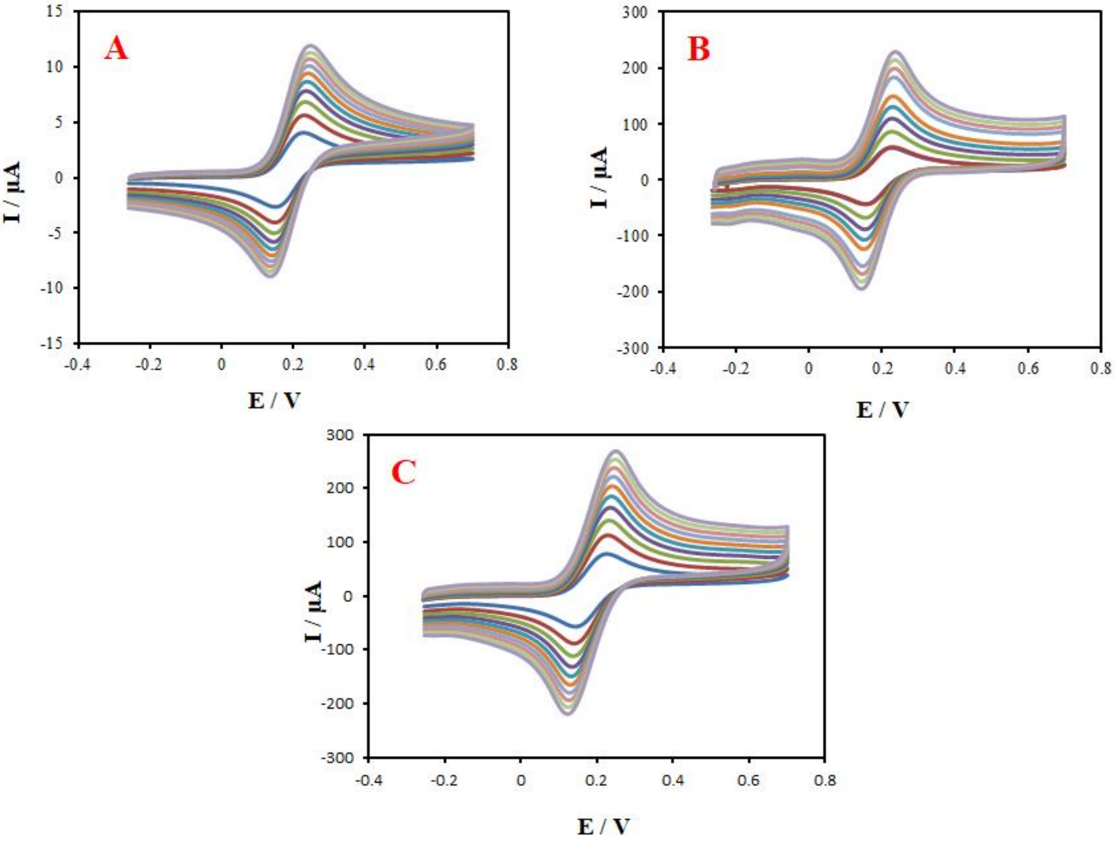


Fig. 4. Cyclic voltammograms of 4 mM $K_4Fe(CN)_6$ in phosphate buffer solution (pH=7.0) at (A) GCE, (B) MWCNTs/GCE, (C) Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE at scan rate = 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mV s^{-1}.

These observations confirmed that the higher electron transfer rates were obtained by performing the analysis at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE respect to unmodified GCE. Investigation on variation of scan rate showed that the anodic peak currents had a linear relationship with the square root of scan rate in the range of 10-100 mVs^{-1} on the GCE and the modified GCE. The results indicated that the electrode processes were diffusion controlled. The regression equations for the electrodes are obtained as follows:

I_{pa} (µA)=29.48 $v^{1/2}$ (V $s^{-1})^{1/2}$+2.76 (R^2=0.998) GCE (1)
I_{pa} (µA)=570.0 $v^{1/2}$ (V $s^{-1})^{1/2}$+6.68 (R^2=0.999) MWCNTs/GCE (2)
I_{pa} (µA)=688.4 $v^{1/2}$ (V $s^{-1})^{1/2}$+13.14 (R^2=0.995) Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE(3)

Also, it should be noticed that a reversible system with diffusion controlled redox processes should satisfy the Randles-Sevcik equation [46]:

$$I_p=(2.69 \times 10^5)\ n^{3/2}\ A\ C_o\ D^{1/2}\ \nu^{1/2} \quad (4)$$

Comparing the slopes of equations 2 (MWCNTs/GCE) and 3 (Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE) with that of equation 1 (the bare GCE) revealed that, the apparent areas of the Fe_3O_4@MCM-48-SO_3H/MWCNTs/GC and MWCNTs/GC modified electrodes were 23.4 and 19.3 times larger than that of the unmodified GCE, respectively. It can be concluded that the application of the Fe_3O_4@MCM-48-SO_3H/MWCNTs composite led to the highest electrochemically active surface area than that of others.

3.2. Optimization of experimental variables

3.2.1. Effects of solution pH

In order to study the effect of pH values on the voltammetric behavior of 5-HT, ST and UA at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE, the pH of the solution was changed from 4.0 to 9.0 in PBS (0.1 M). The results showed that the pH of the samples could largely affect the oxidation currents of all three compounds (Fig. 5).

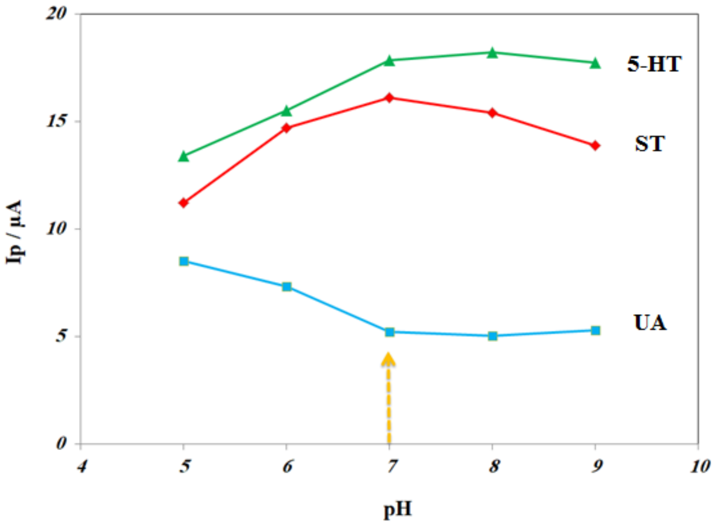


Fig. 5. Effects of the pH of solution for modification of a GCE for simultaneous determination of 20 µM 5-HT and 20 µM ST in the presence of 20 µM of UA at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE in 0.1M PB solution.

At pH of 7, 5-HT and ST showed the highest peak currents. However UA as a interfering compound, showed the lowest peak current at such pH value. Therefore pH of 7 was selected as an optimal solution pH for next experiments. The plots of the oxidation peak potentials (E_{pa})

versus pH (in the range of 4.0 to 9.0) were linear and their slopes were equal with −0.054 V and −0.055 V for 5-HT (equation 5) and ST (equation 6), respectively.

$$E_{pa} = -0.054pH + 0.738;\ R^2=0.9921 \quad (5)$$
$$E_{pa} = -0.055pH + 1.065;\ R^2=0.9967 \quad (6)$$

It can be seen in the above equations, the slopes of E_p versus pH plots equal close to the Nernstian slope of 0.059 so it can be suggests that an equal number of protons and electrons were involved in the oxidation reactions of 5-HT and ST (Scheme 1 and Scheme 2).

3.2.2. Effects of accumulation time

Accumulation time can affect the response the Fe_3O_4@MCM-48-SO_3H/ MWCNTs/GCE towards oxidation of 5-HT and ST. Therefore DPV experiments in solution of 25 µM 5-HT and 20 µM ST were carried out in PBS (pH=7). The results indicated that the responses for 5-HT and ST rose sharply up to 50 s and then they smoothed out at extended time (Fig. 6). Consequently, the accumulation time of 50 s was used as the optimal time for next experiments.

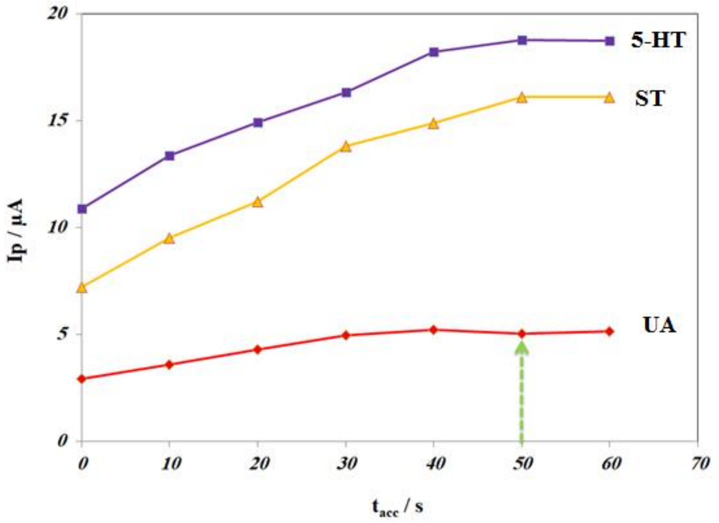


Fig. 6. Effects of the accumulation time for modification of a GCE for simultaneous determination of 20 µM 5-HT and 20 µM ST in the presence of 20 µM of UA at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE in 0.1M PB solution (pH=7.0).

3.3. Electrochemical studies of 5-HT and ST on Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE

CV method was employed to investigate the electrochemical performance of a mixture of 5-HT (20 µM) and ST (20 µM) at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GC electrodes (Fig. 7).

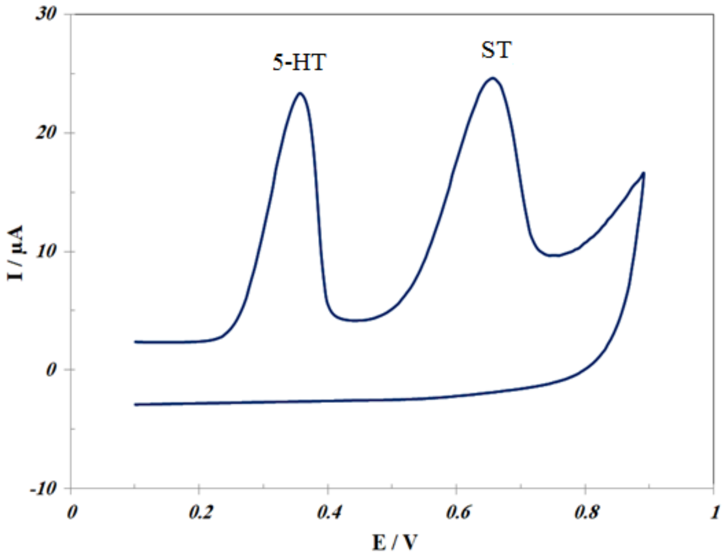


Fig. 7. Cyclic voltammogram of simultaneous determination of 20 µM 5-HT and 20 µM ST at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE in 0.1M PBS (pH=7.0) at scan rate of 50 mVs^{-1}.

Two irreversible oxidation peaks were seen for 5-HT and ST at pH=7.0. In addition proton release from Fe_3O_4@MCM-48-SO_3H at pH of 7 can impose negative charge on this compound and this property would increase the selectivity of the modified electrode respect to interfere chemicals (e.g. UA). In this work, a solution of 5-HT (25 µM) and ST (30 µM) in the presence of UA (20 µM) was prepared for recording the differential pulse voltammograms at GCE (curve a in Fig. 8), Fe_3O_4@MCM-48-SO_3H/GCE (curve b in Fig. 8), MWCNTs/GCE (Fig. curve c in Fig. 8) and Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE (curve d in Fig. 8). It is clear from Fig. 8(a) that small oxidation peak currents were obtained for 5-HT and ST at the unmodified GCE. The anodic peak currents of 5-HT and ST increased higher and somewhat shifted in the direction of less positive potentials at the Fe_3O_4@MCM-48-SO_3H/GCE in comparison to GCE. This fact is an indication for which confirms the electro-catalytic effects of Fe_3O_4@MCM-48-SO_3H (See curve b in Fig. 8). In comparison to the bare GCE, the MWCNTs/GCE showed a significant increase in the oxidation peak currents related to 5-HT and ST due to the high surface area of MWCNTs (See curve c in Fig. 8). It is clear from curve d in Fig. 8, the anodic peak currents of 5-HT and ST indicated the larger values at the Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE with negative shift of potential due to the synergistic electrocatalytic effect of Fe_3O_4@MCM-48-SO_3H/MWCNTs nanocomposite on the electrode surface and its high surface area. The results obtained in this section are in agreement with those obtained in section 3.1. On the other hand, a decrease was observed in the oxidation peak currents of UA at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE compared to MWCNTs/GCE. This fact clearly confirms the negative charge of Fe_3O_4@MCM-48-SO_3H at pH of 7 which could enhance the selectivity of the proposed modified electrode as the sensor for simultaneous determination of 5-HT and ST in solution.

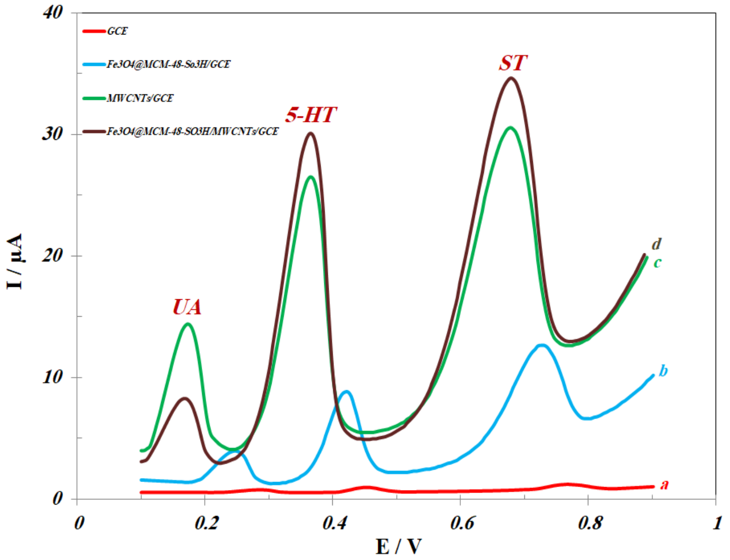


Fig. 8. Differential pulse voltammograms of 25 µM 5-HT and 30 µM ST at (a) GCE, (b) Fe_3O_4@MCM-48-SO_3H/GCE, (c) MWCNTs/GCE and (d) Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE in 0.1 M PB solution (pH=7.0) contain 20 µM UA. Other conditions: Open circuit, t_{acc}=50 s, pulse amplitude=50 mV, scan rate=10 mV s^{-1}, interval time 0.5 s, modulation time=0.2 s and step potential=5 mV

The relationship between E_p and ν was describes by Laviron’s theory for an irreversible anodic reaction as shown in below:

$E_p = E^0 - (RT/\alpha_a nF) \ln (RTK_s / \alpha_a nF) + (RT/\alpha_a nF) \ln \nu$

Application of scan rate studies on 5-HT and ST solution showed the values of α_a of 0.46 and 0.43 respectively.

3.4. Linear dynamic range and detection limit of the method

Figs.9 and 10 show the electrochemical results for simultaneous determinations of 5-HT and ST in a solution of PB (0.1 M) with pH=7.0 at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE. Differential pulse voltammograms were recorded for different concentrations of 5-HT and ST in the presence of UA (20 µM) at Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE (See Fig. 9). Linear dynamic ranges from 0.05 µM to 100 µM and 0.1 µM to 85 µM with calibration equations of I_p (µA)=0.954C (µM)+0.1412 (R^2=0.9984) and Ip (µA)=0.758C (µM)+0.358 (R^2=0.9991); and detection limits 15 nM of (S/N=3) and 25 nM (S/N=3) were obtained for 5-HT and ST, respectively.

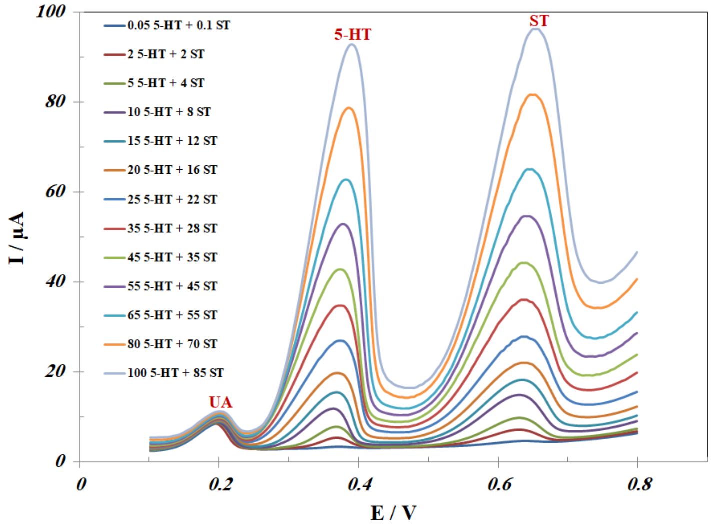


Fig. 9. Differential pulse voltammograms for different concentrations of 5-HT and ST mixtures in 0.1 M PB solution (pH=7.0) contain 20 µM UA at optimum conditions

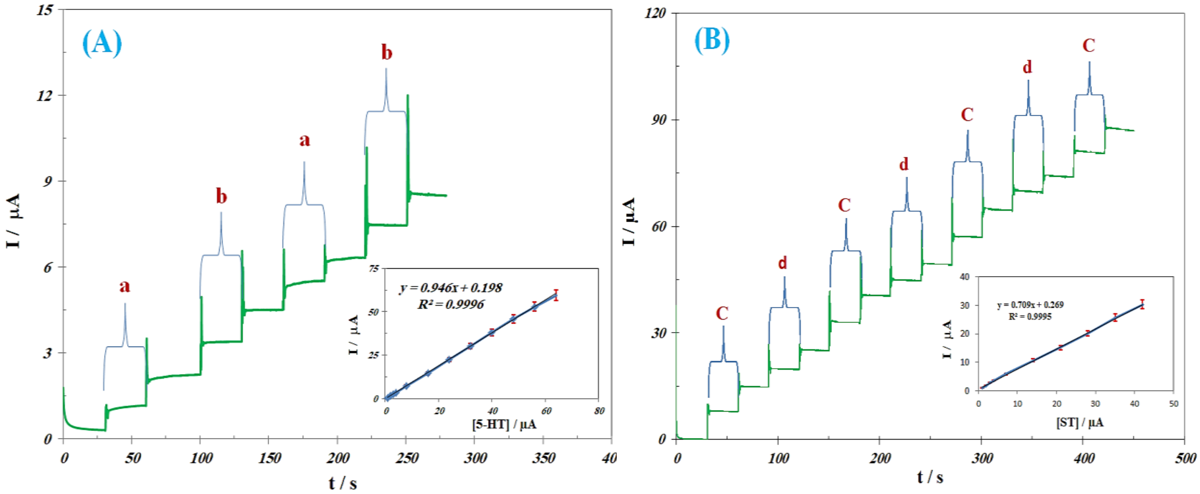


Fig. 10. Hydrodynamic amperometric response at rotating Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE (rotating speed 3000 rpm) held at 0.8 V in PB solution (pH=7.0) contain 20 µM UA for successive additions in µM of (a) 1 µM 5-HT, (b) 1 µM ST, (c) 8 µM 5-HT and (d) 7 µM ST

According to further studies, the linear ranges mentioned above were kept invariable in the combined solutions of 5-HT and ST so it can be concluded that the fabricated modified electrode possessed highly efficiency in the simultaneous determinations of these compounds in mixed pharmaceutical samples.

Fig. 10 illustrates the chronoamperograms related to the response of a rotated modified electrode (3000 rpm) going after the consecutive insertion of 5-HT and ST at an practical potential of 0.8 V in PB solution at pH of 7.0, including UA (20 µM). The results related to 5-HT showed a linear dynamic range from 1 µM to 64 µM with a calibration equation of I_p (µA)=0.946C (µM)+0.198 (R^2=0.9996) and a detection limit of 0.15 µM (S/N=3).

Alternatively, the results obtained from ST showed the linear dynamic range of 1 µM to 42 µM, with a calibration equation of Ip(µA) = 0.709C (µM) + 0.269 (R^2=0.9995) and a detection limit of 0.18 µM (S/N = 3).

Table 1. Maximum tolerable concentration of interfering species

Interfering species	**5-HT**	**ST**
	C_{int} / (µM)	**C_{int} / (µM)**
Ascorbic acid	200	250
Uric acid	150	200
L-Dopa	100	180
Caffeine	300	250
Tyrosine	250	220
L-Glutamic acid	400	350
L-Alanin	300	200
Aspartic acid	350	400
L-histidine	500	600
Dopamine	100	120
Tryptophan	200	250
Vitamin B_1	350	400
Vitamin B_6	180	150
Vitamin B_{12}	500	450

C_{int} refers to interfering compound concentration

3.5. Stability and repeatability of the electrode

One of the significant advantages of our modified electrode is its superior long-term permanence. The long-term stability of this electrode was examined by measuring the amount of decrease in the voltammetric current during the repetitive DPV measurements of 5-HT and ST solutions at the Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE which had already been either stored in wet conditions or exposed to dry conditions for a definite episode of time. For example, when the electrode was kept under wet conditions for 36 h, a reduction less than 2.2 and 2.8 % was observed in the voltammetric oxidation peak currents of 5-HT (10 µM) and ST (10 µM) in 0.1 M PBS (pH=7.0), respectively.

Also, when the electrode was kept in dry conditions for 10 days, the amount of reduction in the oxidation peak currents of 5-HT and ST (in solution) was less than 2.9 and 3.2%, respectively. The results verified the outstanding long-standing constancy of the modified electrode as a sensor for determination of 5-HT and ST.

DPV experiments were repeated in a series of measurements in a solution of 5-HT (10 μM) and ST (10 μM) to estimate the repeatability of the suggested electrode. According to the results obtained from ten successive measurements, the electrode showed excellent repeatability with RSD equivalents to 1.7% and 1.9% for 5-HT and ST, respectively. The results showed that the proposed electrode possessed good repeatability under the optimum conditions.

3.6. Interference studies

In order to examine the causes of some general interfering species on the oxidation peak currents, a solution of 10 µM 5-HT and 10 µM ST was analyzed under the optimum analytical conditions. Table 1 shows the tolerance limit measurements for each interfering potential, which is defined as the concentration upon which the interfering gives an error less than 10% for determination of 5-HT and ST before and after interfering is added.

Table 2. Determination of 5-HT and ST in human serum and urine with Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE

Sample	**Spiked/ μM**		**Found / μM**		**RSD^a/%**		**Recovery**	
	5-HT	**ST**	**5-HT**	**ST**	**5-HT**	**ST**	**5-HT**	**ST**
Blood	1.00	1.00	1.01±0.03	1. 02±0.03	3.7	3.8	101.0	102.0
	5.00	5.00	4.98±0.09	4.97±0.10	2.5	2.7	98.4	101.6
	10.00	10.00	10.11±0.14	10.43±0.16	1.9	2.0	98.1	103.2
Urine	1.00	1.00	1.02±0.03	1.03±0.04	4.1	4.2	98.4	104.6
	5.00	5.00	4.91±0.10	4.97±0.11	3.2	3.4	99.6	97.4
	10.00	10.00	10.12±0.17	10.08±0.19	2.3	2.5	100.7	97.9

3.7. Real sample analysis

Human serum and human urine were chosen to examine the applicability of the Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE to the determination of 5-HT and ST in real samples. The prepared real solutions were spiked with appropriate samples and Fe_3O_4@MCM-48-SO_3H/MWCNTs/GCE was applied under the optimum conditions to record the differential pulse voltammograms. Also, the concentrations of 5-HT and ST were calculated by standard addition method to avoid any matrix effect. The results have been summarized in Table 2. The fine recoveries revealed that both the precision and repeatability of the suggested method are perfectly satisfactory. The experimental data verified that under the optimal circumstances, the suggested electrode has excellent potential for the determination of 5-HT and ST in biological systems or pharmaceutical preparations.

4. CONCLUSION

In this paper, a novel and simple method was introduced to prepare a new modified glassy carbon electrode based on Fe_3O_4@MCM-48-SO_3H/MWCNTs. Then, the applicability of the modified electrode as a sensor for the simultaneous determination of 5-HT and ST in aqueous was extensively studied. When the electrode was modified with a composite resulted from the combination of MWCNTs and Fe_3O_4@MCM-48-SO_3H, the results showed very high sensitivity for the simultaneous determination of 5-HT and ST under the optimum conditions. This enhancement in sensitivity can be due to the outstanding electro-catalytic performance and elevated electro-active surface area of the composite. In addition, the negative charge of the modified electrode surface at pH of 7.0 can increase the selectivity of the proposed method by decreasing negatively charged interfering compound like UA. The studies carried out on some common interfering species showed no significant interference in the simultaneous determination of 5-HT and ST. This sensor showed some excellent benefits such as the facility of production procedure, high stability and sensitivity and broad linear range. Therefore, the suggested sensor can be an good applicant for other realistic purposes.

Acknowledgments

The authors appreciatively acknowledge the research council of Arak University for providing financial support (No. 96.77) for this work.

REFERENCES

[1] S. Wang, J. Yang, X. Zhou, J. Xie, L. Ma, and B. Huang, J. Electroanal. Chem. 722 (2014) 141.
[2] D. Vairavapandian, P. Vichchulada, and M. D. Lay, Anal. Chim. Acta 626 (2008) 119.
[3] C. Hu, and S. Hu, J. Sensors 2009 (2009) 1.
[4] A. Babaei, M. Babazadeh, and M. Afrasiabi, Chin. J. Chem. 29 (2011) 2157.
[5] A. Asghari, S. Kianipour, M. Afrasiabi, and M. Rajabi, Sensor Lett. 11 (2013) 545.
[6] A. Babaei, M. Afrasiabi, and G. Azimi, Anal. Methods, 7 (2015) 2469.
[7] M. Fouladgar, and H. Karimi-Maleh, Ionics 19 (2013) 1163.
[8] A. A. Ensafi, and H. Karimi-Maleh, Int. J. Electrochem. Sci. 5 (2010) 1484.
[9] H. Karimi-Maleh, M. Keyvanfard, Kh. Alizad, M. Fouladgar, H. Beitollahi, A. Mokhtari, and F. Gholami-Orimi, Int. J. Electrochem. Sci. 6 (2011) 6141.

[10] S. Cheraghi, M. A. Taher and H. Karimi-Maleh, J. Food Composition. Anal. 62 (2017) 254.
[11] M. W. Anderson, Zeolites 19 (1997) 220.
[12] A. Babaei, A. Yousefi, M. Afrasiabi, and M. Shabanian, J. Electroanal. Chem. 740 **(**2015) 28.
[13] Y. Wang, L. Sun, T. Jiang, J. Zhang, C. Zhang, C. Sun, Y. Deng, J. Sun, and S. Wang, Drug Dev. Ind. Pharm. 40 (2014) 819.
[14] J. C. Vartuli, K. D. Schmitt, C. T. Kresge, W. J. Roth, M. E. Leonwicz, S. B. McCullen, S. D. Hellring, J. S. Beck, J. L. Schlenker, D. H. Olson, and E. W. Sheppard, Chem. Mater. 6 (1994) 2317.
[15] M. Afrasiabi, and S. Kianipour, Anal. Bioanal. Electrochem. 7 **(**2015) 331.
[16] Z. H. Wang, Q. L. Liang, Y. M. Wang, and G. A. Luo, J. Electroanal. Chem. 540 **(**2003) 129.
[17] J. M. Zen, I. L. Chen, and Y. Shih, Anal. Chim. Acta, 369 **(**1998) 103.
[18] E. Weisskop, A. Panchaud, K. A. Nguyen, D. Grosjean, J. M. Hascoët, Ch. Csajka, Ch. B. Eapa, and N. Ansermot, J. Chromatogr. B 1057 (2017) 101.
[19] T. Ramon-Marquez, A. L. Medina-Castillo, A. Fernandez-Gutierrez, and J. F. Fernandez-Sanchez, Biosens. Bioelectron. 82 (2016) 217.
[20] Q. Jin, L. Shan, J. Yue, and X. Wang, Food Chem. 108 **(**2008) 779.
[21] M. Mumtaz, N. Narasimhachari, R. O. Friedel, G. N. Pandey, and J. M. Davis, Res. Commun. Chem. Pathol. Pharmacol. 36 **(**1982) 45.
[22] I. Hammel, Y. Naot, E. Ben-David, and H. Ginsburg, Anal. Biochem. 90 **(**1978) 840.
[23] F. Engbaek, and B. Voldby, Clin. Chem. 28 (1982) 624.
[24] Z. D. Peterson, M. L. Lee, and S. W. Graves, J. Chromatogr. B 810 **(**2004) 101.
[25] A. Babaei, and M. Babazadeh, Electroanalysis 23 (2011) 1726.
[26] J. Li, and X. Lin, Sens. Actuators B 124 **(**2007) 486.
[27] A. Babaei, and A. R. Taheri, Sens. Actuators B 176 **(**2013) 543.
[28] A. Babaei, A. R. Taheri, and M. Aminikhah, Electrochim. Acta 90 (2013) 317.
[29] A. L. Sanati, F. Faridbod, and M. R. Ganjali, J. Mol. Liq. 241 (2017) 316.
[30] O. E. Fayemi, A. S. Adekunle, and E. E. Ebenso, Sensing and Bio-Sensing Res. 13 (2017) 17.
[31] A. Özcan, and S. Ilkbas, Sens. Actuators B 215 **(**2015) 518.
[32] H. S. Han, H. K. Lee, J. M. You, H. Jeong, and S. Jeon, Sens. Actuators B 190 **(**2014) 886.
[33] I. Cesarino, H. V. Galesco, and S. A. S. Machado, Mater. Sci. Eng. C 40 **(**2014) 49.
[34] D. Y. Chen, S. M. Jiang, Y. Y. Chen, Y. Z. Hu, J. Pharm. Biomed. Anal. 34 (2004) 239.
[35] C.B. Eap, G. Bouchoux, M. Amey, N. Cochard, L. Savary, P. Baumann, J. Chromatogr. Sci. 36 (1998) 365.

[36] 36 E. Lacassie, J. M. Gaulier, P. Marquet, J. F. Fabatel, and G. Lachâtre, J. Chromatogr. B 742 (2000) 229.

[37] J. Patel, E. P. Spencer, and R. J. Flanagan, Biomed. Chromatogr. 10 (1996) 351.

[38] M. M. Schultz, and E. T. Furlong, Anal. Chem. 80 (2008) 1756.

[39] A. L. Batt, M. S. Kostich, and J. M. Lazorchak, Anal. Chem. 80 (2008) 5021.

[40] A. Lajeunesse, C. Gagnon, and S. Sauvé, Anal. Chem. 80 (2008) 5325.

[41] A. J. Ramirez, M. A. Mottaleb, B. W. Brooks, and C. K. Chambliss, Anal. Chem. 79 (2007) 3155.

[42] M. H. Vela, M. B. Quinaz Garcia, and M. C. Montenegro, Fresen. J. Anal. Chem. 369 (2001) 563.

[43] S. Dermis, and H. Y. Cay, Pharmazie 65 (2010) 182.

[44] H. Cheng, J. Liang, Q. Zhang, and Y. Tu, G. Electroanal. Chem. 674 (2012) 7.

[45] H. Kefayati, M. Golshekan, S. Shariati, and M. Bagheri, Chinese J. Catalysis 36 **(**2015) 572.

[46] A. J. Bard, and L. R. Faulkner, Electrochemical Methods. Fundamentals and Applications, Wiley, New York, 2nd edn (2001).