J. Braz. Chem. Soc., Vol. 23, No. 6, 1070-1077, 2012.
Printed in Brazil - ©2012 Sociedade Brasileira de Química
0103 - 5053 $6.00+0.00

Article

Fabrication, Characterization and Analytical Performance of the Hydroxylamine Sensor Based on an Oracet Blue Multi-Walled Carbon Nanotubes Film Deposited on an Electrode Surface

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Neste trabalho, a oxidação eletrocatalítica de hidroxilamina em um eletrodo de carbono vítreo modificado com "oracet blue" e nanotubos de carbono de paredes múltiplas (OBMWCNT-GCE) foi estudada. O potencial de pico de hidroxilamina em superfície OBMWCNT-GCE desloca em cerca de 320 e 335 mV para valores negativos, em comparação com aqueles em superfície MWCNT e superfície GCE ativada. Os parâmetros cinéticos para a oxidação de hidroxilamina na superfície modificada do eletrodo foram determinados por medidas de voltametria cíclica. A detecção amperométrica da hidroxilamina foi realizada em 170 mV em uma solução tampão de fosfato (pH 7,0), resultando em dois intervalos de resposta linear de 4,0-102,4 e 102,4-5820,9 µmol L^{-1} e limite de detecção de 0,7 µmol L^{-1}. A resposta amperométrica para a determinação de hidroxilamina provou ser reprodutível, rápida e estável. Finalmente, OBMWCNT-GCE foi aplicado com sucesso à determinação de hidroxilamina em duas amostras de água natural.

In this work, the electrocatalytic oxidation of hydroxylamine on a glassy carbon electrode modified with oracet blue and multi-walled carbon nanotubes (OBMWCNT-GCE) has been studied. The peak potential of hydroxylamine at OBMWCNT-GCE surface shifts by about 320 and 335mV toward negative values compared with those at a MWCNT and activated GCE surface. Kinetic parameters for the oxidation of hydroxylamine at the modified electrode surface were determined by cyclic voltammetry measurements. The amperometric detection of hydroxylamine was carried out at 170 mV in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) resulting in two linear response ranges of 4.0-102.4 µmol L^{-1} and 102.4-5820.9 µmol L^{-1} and the limit of detection of 0.7 µmol L^{-1}. The amperometric response for hydroxylamine determination proved to be repeatable, fast and stable. Finally, OBMWCNT-GCE was successfully applied to the hydroxylamine determination in two water natural samples.

Keywords: hydroxylamine, multi-walled carbon nanotubes, oracet blue, amperometry, electrocatalytic oxidation

Introduction

In recent years, intensive investigations have been carried out on chemically modified electrodes (CMEs), by virtue of their fast, selective, repeatable, sensitive response and low cost for a chemical analysis.[1-5] Many different strategies have been employed for the modification of the electrodes.[6-12] Due to the unique properties of carbon nanotubes (CNTs) such as high surface area, chemical stability, and high electrical conductivity, the application of CNTs in the fabrication of electrochemical sensors and biosensors has been studied by many research groups.[13-17] Indeed, these compounds are used in an electrode structure so as to promote the rate of electron transfer reactions. The electrocatalytic activity of important chemical and biochemical species, such as hydrazine,[18] epinephrine,[19] amino acids,[20] oxygen,[21] paracetamol,[22] hemoglobin,[23] acetylsalicylic acid,[24] rutin,[25] cholesterol,[26] NADH and hydrogen peroxide,[27] ascorbic acid,[28] uric acid,[29] nitric oxide,[30] glucose,[31] and hydroxylamine,[10,14] have been investigated at the surface of different electrodes modified with CNTs.

Hydroxylamine, NH_2OH, as a derivative of ammonium, is an intermediate in two important microbial processes of nitrogen cycle which are nitrification and anaerobic ammonium oxidation.[10] Hydroxylamine belongs to an important class of reducing agents, which are commonly

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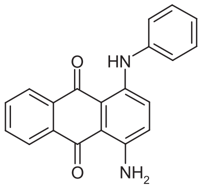
used as industrial raw materials. It is one of the intermediate products of nitrogen-cycle and holds an important place in biological studies.[32] Hydroxylamine is a natural product found in mammalian cells and bacteria. In the former, some hydroxylamine derivatives also constitute a great part of anticancer drugs.[33] In addition, hydroxylamine has been shown to inactivate or inhibit a number of cellular enzymes and some viruses *in vitro*. It is also a skin irritant and sensitizer. Therefore, various methods have been reported for the determination of low levels of hydroxylamine from industrial, environmental, and health viewpoints.[34-44] These include chromatography,[34,35] spectrophotometric[37] and electrochemical methods.[10,11,14,16,32,38-44] In this regard, we had reported the preparation of a rutin multi-walled carbon nanotubes modified glassy carbon electrode (RMWCNT-GCE),[14] a coumestan derivative modified carbon paste electrode,[32] an indenedione derivative MWCNT-GCE (IMWCNT-GCE),[41] a chlorogenic acid MWCNT-GCE (CGA-MWCNT-GCE),[42] a 4-hydroxy-2-(triphenylphosphonio)phenolate MWCNT-GCE (HTP-MWCNT-GCE),[43] a ruthenium oxide nanoparticles,[44] and their applications in hydroxylamine determination. The modified electrodes for hydroxylamine measurement have advantages and limitations. Therefore, it is of interest to have further efforts to fabricate certain modified electrodes that can improve the electrocatalytic characteristics and analytical parameters of hydroxylamine quantification. Oracet blue (OB) is an anthraquinone derivative which can be viewed as a quinone derivative. It is accepted generally that anthraquinone derivatives can be quite active in the electrocatalytic redox reactions of different analytes.[45-49] Preparation of OB modified glassy carbon electrode and its use in the simultaneous determination of dopamine, ascorbic acid and uric acid were reported by our group.[45] Owing to the importance of anthraquinone derivatives in mediated electron transfer reactions, in this study, the fabrication of oracet blue MWCNT-GCE (OBMWCNT-GCE) and its application for the electrocatalytic oxidation of hydroxylamine have been reported. The findings indicate that OBMWCNT-GCE has advantages such as potent and persistent electrocatalytic activity, good repeatability, low limit of detection, high sensitivity, good stability and wide linear concentration range for hydroxylamine determination.

Experimental

Electrochemical apparatus and chemicals

An Autolab potentiostat-golvanostat PGSTAT 30 (Eco Chemie, Ultrecht, the Netherlands) equipped with GPES 4.9 software, in conjunction with a three-electrode system and a personal computer, was used for electrochemical measurements. A saturated calomel reference electrode (SCE), a platinum wire counter electrode, an oracet blue electrodeposited on a GCE (OBMGCE), multi-walled carbon nanotubes modified GCE (MWCNT-GCE), and an oracet blue electrodeposited on multi-walled carbon nanotubes modified GCE (OBMWCNT-GCE) were employed as working electrodes for the electrochemical studies. The pH was measured with a Metrohm model 691 pH/mV meter.

The multi-walled carbon nanotubes with a diameter of 10-20 nm, a length of 5-20 µm, and a purity > 95% were purchased from Nanolab Inc. (Brighton, MA). The oracet blue (OB), 1-amino-4-anilinoanthraquinone, (see Scheme 1 for structure), hydroxylamine, and other reagents had analytical grades from Merck and were used as received. Doubly distilled water was used to prepare all the solutions. Buffer solutions (0.1 mol L^{-1}) were prepared from H_3PO_4, and the pH was adjusted with 0.1 mol $L^{-1}$of H_3PO_4 or 2.0 mol $L^{-1}$of NaOH. Hydroxylamine solution was freshly prepared just prior to use and all the experiments were carried out at room temperature.



Scheme 1. Structure of oracet blue (OB).

Electrodes preparation

The procedure of preparing the working electrodes was as follows. At first, GCE was carefully polished mechanically with 0.05 µm of Al_2O_3 slurry on a polishing cloth and then rinsed with doubly distilled water. For the electrochemical activation of the electrode, it was immersed in a 0.1 mol L^{-1} sodium bicarbonate solution and was activated by a continuous potential cycling from –1.4 to 1.7 V at a sweep rate of 100 mV s^{-1} until a stable voltammogram was obtained. For the preparation of OB modified GCE (OBMGCE), the activated GCE (AGCE) was rinsed with doubly distilled water and was modified by 18 cycles of potential sweep between 0.0 and 230 mV at 25 mV s^{-1} in a 1.0 mmol L^{-1} solution of OB in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) and a small amount of ethanol in order to dissolve OB in an aqueous solution. For the preparation of MWCNT modified GCE (MWCNT-GCE), a 3 µL of MWCNT-DMF suspension (1 mg mL^{-1}) was placed directly onto the activated GCE surface and dried at room temperature to form a MWCNT film at the GCE surface. OBMWCNT-GCE was prepared by immersing MWCNT-GCE in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) containing 0.1 mmol L^{-1} of OB by 18 continuous potential cycles from 0.0 to 230 mV at 25 mV s^{-1}.

Results and Discussion

The optimum conditions for preparation of OBMWCNT-GCE and stability of the modified electrode

The effect of number of potential cycles and MWCNT value, used for GCE surface modification, on the current response of OBMWCNT-GCE were investigated to optimize the testing performance. The current response of the modified electrode is expected to be affected by the amount of OB on the surface of MWCNT modified GCE, which can be controlled by a number of potential cycles during the modification of the electrode. The results show that with the increment in the number of potential cycles, the current response increased and the highest current was found around 18 cycles of potential. However, 18 cycles of potential is considered as the optimum since, for more than 18 cycles, a decrease is observed in the current response. This is probably due to the formation of a thick and compact film, which disturbs the electron transfer. The current response of OBMWCNT-GCE is also affected by the amount of MWCNT on the surface of GCE, which can be controlled by using the same concentration of MWCNT with different volumes (1-20 µL) of the suspension. After an OB film was formed on MWCNT modified GCE, the current responses of the modified electrode were recorded. The relationship between the current response and the MWCNT value indicated that an increment of MWCNT value caused the current response to increase. This implies that a higher MWCNT value results in a higher sensitivity. In the experiment, however, it is observed that the background current also increased with the increase of the MWCNT value, which would not facilitate the determination of an analyte. Therefore, in this work, a moderate MWCNT value of 3 µL of MWCNT-DMF solution (1 mg mL^{-1}) was selected for the fabrication of OBMWCNT-GCE. This value of MWCNT-DMF solution is significantly less than 15 µL that were used for fabrication of CGA-MWCNT-GCE,[42] but the sensitivity of hydroxylamine determination at OBMWCNT-GCE surface increases compared with that at a CGA-MWCNT-GCE. This result indicates that OB is an excellent mediator for electrocatalytic oxidation of hydroxylamine in comparison with CGA.

Also, the stability of OBMWCNT-GCE was tested by repetitive scans in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) for a period of time and recording the voltammograms at different time intervals. The experimental results show that although a significant decline in the current response of the modified electrode was observed during the initial minutes, the rate of the current decline was then slowed significantly. The measurement of the current response of the modified electrode during continuous potential cycling indicates that the loss of the current response is about 8% after 50 potential cycles at 50 mV s^{-1}. Also, only a 9% decrease was observed in the current response of the modified electrode when it was kept in a 0.1 mol L^{-1} phosphate buffer solution of pH 7.0 for 5 days. The above results indicate that the chemical stability of the modified electrode is acceptable.

Electrocatalytic oxidation of hydroxylamine at OBMWCNT-GCE

Figure 1 shows the cyclic voltammetric responses of a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) containing 0.5 mmol L^{-1} of hydroxylamine at OBMWCNT-GCE (curve b),OMGCE (curve d), MWCNT-GCE (curve e) and an activated GCE (curve f). In the absence of hydroxylamine, a well-behaved redox response can be observed corresponding to the redox reaction of *p*-hydroquinone ring of the electrodeposited OB on a MWCNT-GCE (Figure 1, curve a) or an activated GCE (Figure 1, curve c). Upon the addition of 0.5 mmol L^{-1} of hydroxylamine, there is an enhancement in the anodic peak currents, and a small current is observed in the cathodic peak (Figure 1, curves b and d). This is indicative of a very strong electrocatalytic effect. As illustrated, the anodic peak potential for hydroxylamine oxidation at OBMWCNT-GCE (curve b) is about 135 mV which is close to that of the

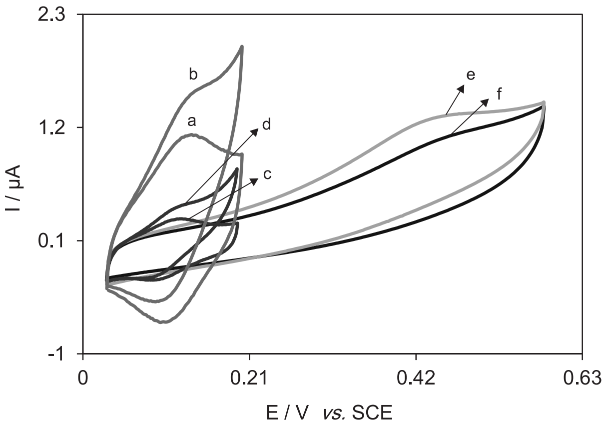


Figure 1. Cyclic voltammograms of OBMWCNT-GCE in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) at scan rate 20 mV s^{-1} in (a) the absence and (b) the presence of 0.50 mmol L^{-1} hydroxylamine. (d), (e) and (f) as (b) for OBMGCE, MWCNT-GCE and activated GCE respectively. (c) as (a) for OBMGCE.

Table 1. Comparison of the electrocatalytic oxidation characteristics of hydroxylamine (0.5 mmol L^{-1}) on various electrode surfaces at pH 7.0

Type of electrode[a]	Oxidation peak potential / mV	Oxidation peak current / µA
AGCE	470	0.12
MWCNT-GCE	455	0.25
OBMGCE	135	0.07
OBMWCNT-GCE	135	0.36

[a]AGCE: activated glassy carbon electrode, MWCNT-GCE: multi-walled carbon nanotubes modified glassy carbon electrode, OBMCCE: oracet blue modified glassy carbon electrode, OBMWCNT-GCE: oracet blue multi-walled carbon nanotubes modified glassy carbon electrode.

surface-confined mediator anodic peak potential in the absence of hydroxylamine. But at MWCNT-GCE (curve e) and activated GCE (curve f), the peak potentials are about 455 and 470 mV respectively. At the bare GCE, no current is observed in the presence of hydroxylamine. Table 1 shows the electrochemical characteristics of hydroxylamine oxidation on various electrode surfaces at pH 7.0. From Table 1, it is concluded that the best electrocatalytic effect for hydroxylamine oxidation is gained at OBMWCNT-GCE surface. For example, according to the results, there is a dramatic enhancement of the anodic peak current at OBMWCNT-GCE(curve b) relative to the value obtained at OBMGCE (curve d), MWCNT-GCE (curve e), and activated GCE (curve f). Also, the peak potential of hydroxylamine oxidation at OBMWCNT-GCE (curve b) shifts by about 320 mV and 335 mV toward negative values compared with that at MWCNT-GCE (curve e) and activated GCE (curve f) respectively. The improvement of the hydroxylamine determination sensitivity at OBMWCNT-GCE is due to the enhancement of the effective surface area of the modified electrode as well as the electrocatalytic effect of OB in the E_rC_i catalytic ($E_rC'_i$) reaction mechanism. Therefore, it is logical to conclude that MWCNT can be used as a new material for the immobilization and electron transfer reactions of OB. Also, a combination of MWCNT and a mediator (OB here) definitely improves the characteristics of hydroxylamine oxidation. Also, the peak potential of hydroxylamine oxidation at the OBMWCNT-GCE is less positive, in most cases, in comparison with those reported for electrodes modified with other mediators such as rutin,[14] nickel hexacyanoferrate,[16] a coumestan derivative,[32] gold nanoparticle-polypyrrole nanowire,[40] an indenedione derivative,[41] chlorogenic acid,[42] 4-hydroxy-2-(triphenylphosphonio)phenolate,[43] and ruthenium oxide nanoparticles.[44] The oxidation potentials of hydroxylamine on different modified electrodes are shown in Table 2.

Table 2. Comparison of some of the electrocatalytic characteristics of the hydroxylamine oxidation at various modified electrode surfaces

Modifier	Linear range / (µmol L^{-1})	Sensitivity / (µA µmol^{-1} L)	Limit of detection / (µmol L^{-1})	E_p / mV	Ref.
ZnO nanofilm on to carbon nanotubes	0.4-19000	0.0075	0.12	80	10
rutin MWCNT	1.0-33.8 33.8-81.7	0.0288 0.025	1.0	190	14
nickel hexacyanoferrate	1.0-50.0	0.00046	0.024	460	16
coumestan derivative	60-1000	-	10.75	320	32
gold nanoparticle-polypyrole nanowire	1-500 500-18000	0.0639 0.0104	0.21	-	40
indenedione derivative	1.0-10.0 10.0-100.0	0.1955 0.0841	0.8	140	41
chlorogenic acid MWCNT	11.8-74.1 74.1-758.6 758.6-1939.8 1939.8-2900.7	0.0024 0.0014 0.0011 0.0008	1.4	150	42
4-hydroxy-2-(triphenylphosphonio)phenolate MWCNT	2.0-10.0 10.0-1000.0 1000.0-8000.0	0.0227 0.0042 0.0016	0.16	200	43
ruthenium oxide nanoparticles	4.0-33.8 33.8-78.3	-	0.45	450	44
oracet blue MWCNT	4.0-102.4 102.4-5820.9	0.005 0.0004	0.7	135	This work

The effect of the potential scan rate on the electrocatalytic oxidation of hydroxylamine at OBMWCNT-GCE surface was studied to get information about the rate determining step. The linear sweep voltammograms of the modified electrode in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) containing 1.0 mmol L^{-1} of hydroxylamine were obtained at different scan rates varying from 6 to 16 mV s^{-1} (not shown). Tafel plots were drawn (Figure 2A), from points of the Tafel region of the linear sweep voltammograms. The results of polarization studies for the electrooxidation of hydroxylamine at OBMWCNT-GCE show that, for all potential sweep rates, the average Tafel slope is 10.8 V^{-1}. Referring to equation 1,[50] the average Tafel slope of 10.8 V^{-1} agrees well with the involvement of one electron in the rate-determining step of electrode process, assuming a charge transfer coefficient of $\alpha = 0.36$.

$$\text{Tafel slope} = (1 - \alpha)n_\alpha F/2.3RT \qquad (1)$$

In addition, the exchange current density, j_0, appears to be readily accessible from the intercept of the Tafel plots.[50] The average value of the exchange current density, j_0, for hydroxylamine oxidation at the modified electrode surface is found to be 2.4 µA cm^{-2}. The value obtained for j_0 of hydroxylamine at OBMWCNT-GCE is higher than that of the exchange current density at a coumestan-derivative modified electrode (0.084 µA cm^{-2})[32] but it is lower than the j_0 value of hydroxylamine at the indenedione modified MWCNT-GCE (16.9 µA cm^{-2}).[41] Figure 2B shows that a plot of the catalytic peak current versus the square root of scan rate is linear. This result indicates that, at a sufficient overpotential, the process is diffusion rather than surface-controlled, which is the ideal case for quantitative applications.[50] Also, from this plot, one can calculate the approximate total number of electrons in the overall oxidation of hydroxylamine (n) using the following equation for diffusion-controlled electrochemically irreversible reaction:[51]

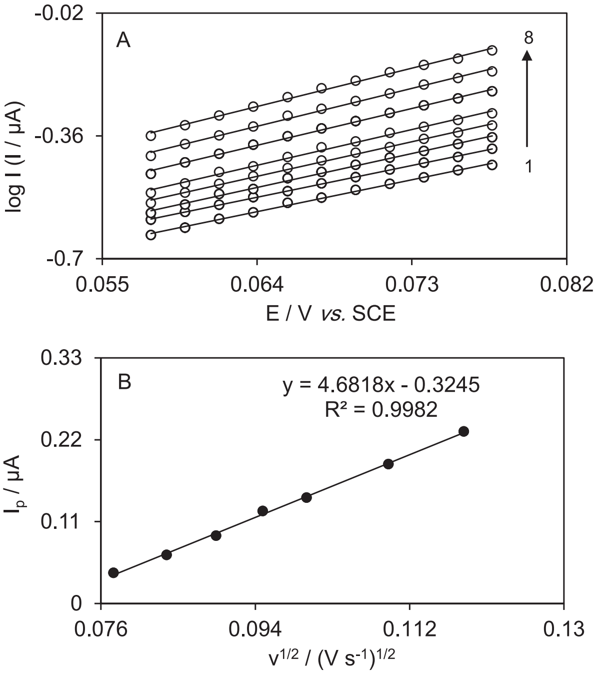


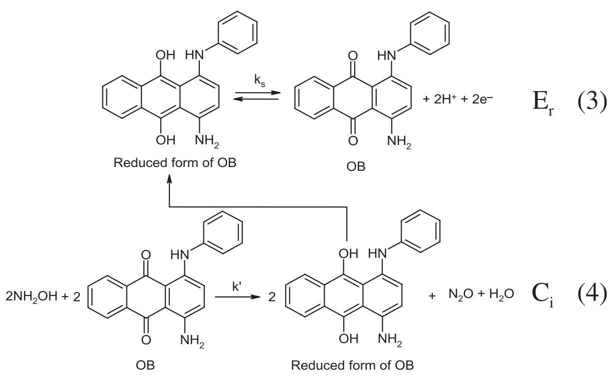
Figure 2. (A) The Tafel plots derived from the linear sweep voltammograms of the OBMWCNT-GCE in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) containing1.0 mmol L^{-1} hydroxylamine at the different scan rates. Numbers of 1-8 correspond to 6.0, 7.0, 8.0, 9.0, 10.0, 12.0, 14.0 and 16.0 mV s^{-1}. (B) Variation of the electrocatalytic peak currents *vs.* the square root of scan rates.

$$I_p = 3.01\times10^5 n[(1-\alpha)n_\alpha]^{1/2} A C_b D^{1/2} \nu^{1/2} \qquad (2)$$

where D_{app} is the apparent diffusion coefficient of hydroxylamine ($D = 1.2\times10^{-7}$ $cm^2\ s^{-1}$ obtained by chronoamperometry), C_b is the bulk concentration of hydroxylamine (1.0 mmol L^{-1}), and A is the electrode surface area (0.0314 cm^2). The values of α and n_α which are deduced from Tafel plots are 0.36 and 1, respectively. Also, it is concluded that the total number of electrons involved in the anodic oxidation of hydroxylamine at the modified electrode is $n = 1.8 \cong 2$. Based on the above results, one can describe the E_rC_i catalytic ($E_rC'_i$) reaction mechanism of hydroxylamine at OBMWCNT-GCE as shown in equations 3 and 4. For an $E_rC'_i$ mechanism, the value of catalytic reaction rate constant, k′, between the electrodeposited oracet blue (OB) and hydroxylamine can be estimated using the Andrieux and Saveant theoretical model.[52] Based on this theory, the mean value of the catalytic rate constant, k′, between hydroxylamine and OB is calculated to be $(1.7 \pm 0.3) \times10^{-4}$ cm s^{-1}. It is expected the k′ and α values are pH dependent. Therefore, the hydroxylamine oxidation at the modified electrode surface was studied in different pHs and the results are shown in Table 3. The data given in Table 3 revealed that the k′ value is increased and α value is decreased with increasing of pH. Also, the results show that the catalytic rate constants, k′, obtained for hydroxylamine oxidation in this work are in good agreement with 5.51×10^{-4},[32] 4.0×10^{-4},[41] 2.8×10^{-4},[42] and 6.9×10^{-4} cm s^{-1},[43] which were previously reported for hydroxylamine oxidation at the other modified electrode surfaces.

Table 3. The charge transfer coefficient, α, and the catalytic rate constant, k', between the electrodeposited OB and hydroxylamine at different pHs

pH	3	5	7	9
α	0.54	0.51	0.36	0.33
k' / (cm s^{-1})	1.02×10^{-4}	1.26×10^{-4}	1.7×10^{-4}	1.86×10^{-4}



The overall oxidation reaction of hydroxylamine at the modified electrode surface is given in equation 5:

$$2NH_2OH \longrightarrow N_2O + H_2O + 4H^+ + 4e^- \qquad (5)$$

Chronoamperometric studies of hydroxylamine oxidation at OBMWCNT-GCE

The electrocatalytic oxidation of hydroxylamine at OBMWCNT-GCE surface was also studied by chronoamperometry. Chronoamperograms were obtained at different concentrations of hydroxylamine at a potential step of 170 mV (Figure 3A). For an electroactive material (hydroxylamine in this case) with an apparent diffusion coefficient, D_{app}, the current corresponding to the electrochemical reaction (under diffusion control) can be described by Cottrell equation,[50]

$$I = nFAD_{app}^{1/2}C/\pi^{1/2}t^{1/2} \qquad (6)$$

where D_{app} and C are the apparent diffusion coefficient (cm^2 s^{-1}) and the bulk concentration (mol cm^{-3}) of the analyte, respectively. Figure 3B shows the experimental plots of I *versus* $t^{-1/2}$ with the best fits for different concentrations of hydroxylamine employed. The slopes of the resulting straight lines were then plotted *versus* the hydroxylamine concentration (Figure 3C), from whose slope (3.713 μA s$^{1/2}$ mmol L^{-1}) and using the Cottrell equation[50] the apparent diffusion coefficient ($D_{app.}$) of hydroxylamine was calculated as 1.2×10^{-7} cm^2 s^{-1}. The calculated apparent diffusion coefficient is in a good agreement with some of those previously reported forhydroxylamine,[32,41,42] but is lower than the values which are reported in some other published papers.[14,40,43]

Amperometric studies of electrocatalytic oxidation of hydroxylamine at the OBMWCNT-GCE surface

Amperometry under a stirred condition has a much higher current sensitivity than cyclic voltammetry, and it can be used to determine the linear range and to estimate the lower limit of detection of hydroxylamine at the OBMWCNT-GCE surface. Figure 4A depicts the amperograms obtained for different concentrations of 4.0-5820.9 μmol L^{-1} of hydroxylamine at the potential step of 170 mV, while OBMWCNT-GCE rotates at the speed of 2000 rpm. As shown in the Figure, during the addition of even 1.0 and then 10.0 μmol L^{-1} of hydroxylamine, certain well-defined responses are observed. For each addition of hydroxylamine, a sharp rise in the current was observed within a response time less than 2 s. Also, Figures 4B and 4C clearly show that the plot of the peak current *versus* the hydroxylamine concentration is constituted of two linear segments of 4.0-102.4 and 102.4-5820.9 μmol L^{-1} with different slopes. According to the method mentioned in the references,[53] the lower limit of detection, C_m, was obtained to be 0.7 μmol L^{-1} by using the equation $C_m = 3s_{bl}/m$, where s_{bl} is the standard deviation of the blank response and m is the slope of the calibration plot in the range of 4.0-102.4 μmol L^{-1} of hydroxylamine (0.005 μA μmol^{-1} L). In Table 2, the analytical parameters of electrocatalytic determination of hydroxylamine in this work are compared with the corresponding values previously reported for some other modified electrodes.[10,14,16,32,40-44] It can be seen that the responses made by the proposed modified electrode are in most cases superior to the previously reported modified electrodes. The average current measured and the precision estimated in terms of relative standard deviation (RSD%) for fifteen repeated measurements (n = 15) of 10.0 μmol L^{-1} of hydroxylamine at the applied potential of 170 mV on OBMWCNT-GCE were 0.107 ± 0.003 μA and 2.9%

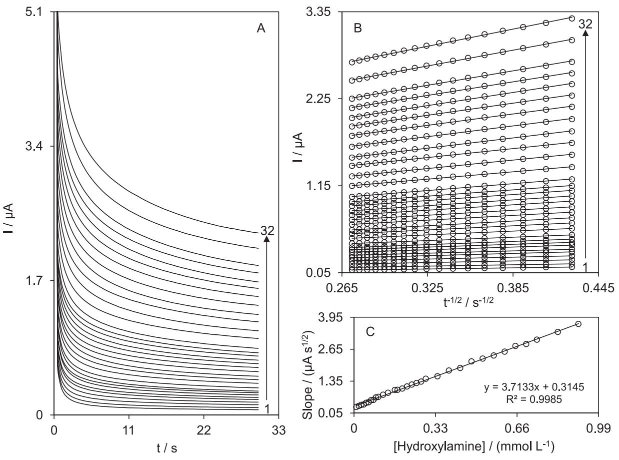


Figure 3. (A) Chronoamperometric responses of OBMWCNT-GCE in a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0) at potential step of 170 mV for different concentrations of hydroxylamine. Numbers of 1-32 correspond to hydroxylamine concentrations of 0.009-0.909 mmol L^{-1}. (B) Plots of I *vs.* $t^{-1/2}$obtained from the chronoamperograms of (A). (C) Plot of the slopes of straight lines of (B) against the hydroxylamine concentrations.

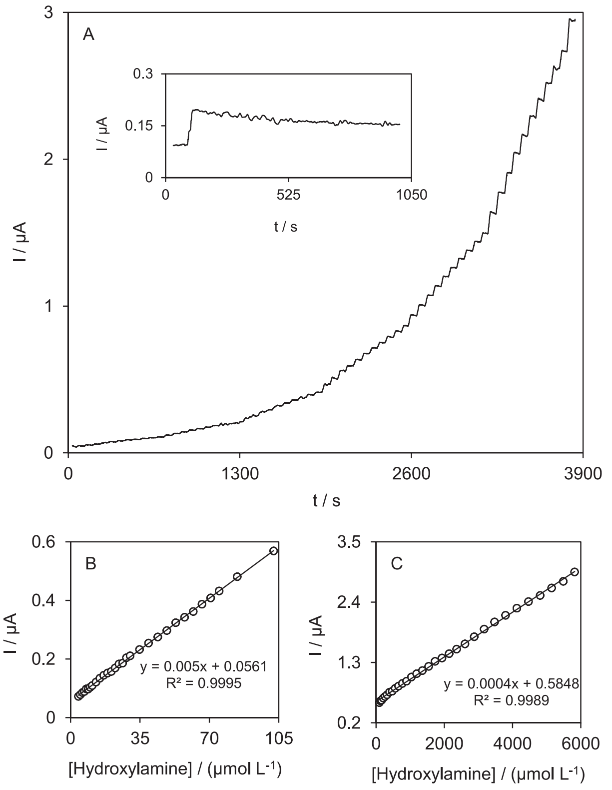


Figure 4. (A) Amperometric responses at a rotating OBMWCNT-GCE (rotation speed 2000 rpm) held at 170 mV in different concentrations of 4.0-5820.8 µmol L^{-1} hydroxylamine. Inset shows the stability of the response of OBMWCNT-GCE to 17.0 µmol L^{-1} hydroxylamine during 1000 s. (B) and (C) show variations of the amperometric currents *vs.* hydroxylamine concentrations in the two ranges of 4.0-102.4 µmol L^{-1} and 102.4-5820.9 µmol L^{-1}.

respectively. The amperometric response of 17.0 µmol L^{-1} of hydroxylamine over a period of 1000 s (inset of Figure 4A) indicates that after an initial decrease of current, the response of OBMWCNT-GCE has remained almost stable through out the experiment. This is proof that there is no inhibitory effect of hydroxylamine and its oxidation product(s) on the modified electrode surface. Thus OBMWCNT-GCE was found to have merits such as fast response time, high sensitivity, a good limit of detection, and a wide linear range for hydroxylamine determination.

Determination of hydroxylamine in tap and well water samples

From the results that are mentioned in the previous section, it is apparent that OBMWCNT-GCE possesses a high sensitivity and a good limit of detection to determine hydroxylamine in real samples. In order to test its practical application, the modified electrode was used to determine hydroxylamine in two natural water samples. To do the experiment, 5.0 mL of a natural water sample was diluted to 10 mL with a 0.1 mol L^{-1} phosphate buffer solution (pH 7.0). Then, certain amounts of hydroxylamine were added and their recoveries were determined using amperometry measurements as well as the calibration plot which is shown in Figure 4B. The results in Table 4 show that the RSD% and the recovery rates of the spiked samples are acceptable. Thus, OBMWCNT-GCE can be efficiently used for hydroxylamine determination in different water samples.

Table 4. Determination and recovery of hydroxylamine in two water samples using OBMWCNT-GCE

Samples	Added / (µmol L^{-1})	Found / (µmol L^{-1})	RSD / %	Recovery / %
Drinking water	-	Not found	-	-
	15.0	15.2	3.1	101.3
	30.0	29.8	2.5	99.3
Tap water	-	Not found	-	-
	20.0	20.4	2.6	102.0
	40.0	40.9	2.7	102.2

Conclusions

The results obtained here show that oracet blue can be electrodeposited easily at the surface of a multi-walled carbon nanotubes (MWCNT) modified glassy carbon electrode (GCE). It is also shown that electrodeposited OB at the MWCNT-GCE surface (OBMWCNT-GCE) exhibits a potent and persistent electrocatalytic behavior toward hydroxylamine oxidation. According to the data in this study, the kinetic parameters of electron transfer coefficient, α, and the heterogeneous catalytic reaction rate constant, k', between OBMWCNT-GCE and hydroxylamine have been estimated as 0.36 and $(1.7 \pm 0.3) \times 10^{-4}$, respectively. The apparent diffusion coefficient ($D_{app.}$) of hydroxylamine is also determined as 1.2×10^{-7} $cm^2\ s^{-1}$ using a chronoamperometry technique. In amperometric measurements, a limit of detection of 0.7 µmol L^{-1} and two linear ranges of 4.0-102.4 µmol L^{-1} and 102.4-5820.9 µmol L^{-1} have been obtained for hydroxylamine at the proposed modified electrode. Finally, it has emerged that amperometry method can be used as an analytical method to determine hydroxylamine in two water samples.

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Submitted: June 26, 2011