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Electrochemistry and voltammetry of procaine using a carbon nanotube film coated electrode

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Abstract

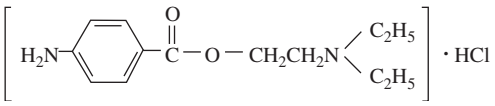
A new rapid, convenient and sensitive electrochemical method is described for the determination of procaine in pharmaceutical preparations, based on the unique properties of a multi-wall carbon nanotube (MWNT) thin film. The electrochemical behavior of procaine at the MWNT film-coated glassy carbon electrode (GCE) was investigated in detail, showing that the MWNT-coated GCE exhibits electrocatalytic activity to the oxidation of procaine because of the significant peak current enhancement and the lowering of oxidation overpotential. Furthermore, the mechanism for the oxidation of procaine at the MWNT-coated GCE was also studied. Finally, various experimental parameters such as solution pH value, the amount of MWNT, accumulation conditions and scan rate were optimized for the determination of procaine, and a new method with detection limit of 2×10^{-7} mol/L was developed for procaine determination. This newly proposed method was successfully demonstrated with procaine hydrochloride injection.

Keywords: Procaine; Electrochemical determination; Carbon nanotube-modified electrode

1. Introduction

The drug analysis, an important branch of analytical chemistry, has extensive impact on public health. Therefore, the establishment of simple, rapid, sensitive and reliable method for the determination of active ingredient is welcomed and necessary.

Procaine was first synthesized in 1905, and was the first injectable man-made local anesthetic. It was introduced into medical use by surgeon Heinrich Braun. The proper chemical name for procaine hydrochloride is 2-diethylaminoethyl 4-aminobenzoate hydrochloride and the chemical structure is:



To date, numerous methods have been reported for the determination of procaine and its salts in pharmaceutical preparations or biological samples. The present United States Pharmacopoeia method is an extration-spectrophotometric method based on the absorbance of procaine hydrochloride at 280 nm [1]. The Chinese Pharmacopoeia method is dead-stop titration [2]. Additionally, many other methods including spectrophotometry [3–5], high-performance liquid chromatography [6,7], gas chromatography [8], fluorimetry [9], ion-pairing flow injection analysis with piezoelectric detection [10], and chemiluminescence [11], were also reported.

Recently, much effort has been made to the electrochemical determination of procaine, since electroanalytical method possesses many advantages such as high sensitivity, rapid response and extreme simplicity. For instance, a pumice-modified glassy carbon electrode [12] and a modified polymetric electrode [13] were employed to detect procaine, respectively. Wang and coworkers [14] reported a method of amperometric detection using liquid chromatography with a metal-oxide dispersed glassy carbon electrode. Ion-selective-

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electrode (ISE) was also exploited to detect procaine [15], and unfortunately, its detection limit is very poor (just 10^{-5} mol/L). Otherwise, polarography [16,17], coulometric titration [18] and potentiometric titration [19] were also reported. However, voltammetric determination of procaine using a carbon nanotube-modified electrode has not been reported.

The objective of the current work is to develop a convenient and sensitive method for the determination of procaine, based on the unusual properties of carbon nanotubes such as strong adsorptive ability, huge specific area, subtle electronic properties as well as excellent electrocatalytic activity. The electrochemical behavior of procaine on the multi-wall carbon nanotube (MWNT)-coated glassy carbon electrode (GCE) strongly revealed that the electrochemical oxidation of procaine was facilitated and the determination sensitivity of procaine was significantly improved. At the MWNT-coated GCE, the remarkable peak current enhancement and negative shift of oxidation peak potential occurred to procaine, compared with that of a bare GCE. Consequently, a voltammetric method based on the carbon nanotube-modified electrode was first developed for the determination of procaine. This newly proposed method possesses following advantages such as high sensitivity, rapid response, low cost and simplicity.

2. Experimental

2.1. Reagents

A 1×10^{-3} mol/L stock solution of procaine hydrochloride (Sigma) was prepared in redistilled water and then stored in the dark at 0 °C. Standard solutions of procaine were prepared by dilution of the stock solution with redistilled water. All reagents were of analytical grade and used directly without purification. Redistilled water was used throughout.

The multi wall carbon nanotube (MWNT) with an average diameter of 30 nm was obtained from Chengdu Organic Chemicals Co., Chinese Academy Sciences, and then refluxed in concentrated HNO_3 for 10 h to cause segmentation, purification and carboxylation [20].

2.2. Apparatus

All the electrochemical measurements were carried out with a CHI 660A Electrochemical Workstation (CH Instruments, Austin, USA). A conventional three-electrode system, including a MWNT-coated GC working electrode, a saturated calomel reference electrode (SCE) and a Pt wire counter electrode, was employed.

2.3. Fabrication of MWNT-coated GCE

In our previous work [21], it was found that insoluble carbon nanotubes can be dispersed into water in the presence of a kind of surfactant — dihexadecyl hydrogen phosphate (DHP), to form a stable and well-distributed suspension. In the present work, an amount of 5 mg MWNT and 5 mg DHP were added into 5 mL of redistilled water, and then sonicated for about 30 min with an ultrasonicator (55 kHz) to get a stable and homogeneous MWNT–DHP suspension. Prior to modification, the GCE was polished successively with alumina pastes of 0.5 and 0.1 μm to a mirror finish, rinsed and sonicated (3 min) in redistilled water. Finally, the GCE was coated with 7.5 μL of the resulting MWNT–DHP suspension and allowed to evaporate water at room temperature in air. The DHP-modified GCE was prepared by the same procedure as explained above, but without MWNT.

2.4. Analytical procedure

The MWNT-coated GCE was first activated in pH 7.0 phosphate buffer by cyclic voltammetric sweeps between 0.2 and 1.0 V until stable cyclic voltammograms were obtained, and then transferred into another 10 mL of pH 7.0 phosphate buffer containing a certain concentration of procaine. After 4 min of open-circuit accumulation, the linear sweep voltammograms from 0.3 to 0.9 V at 100 mV/s were recorded for procaine. The oxidation peak current at 0.75 V was measured. After every measurement, the MWNT-modified GCE was retransferred into the pure phosphate buffer (pH 7.0) to remove the adsorptive substances and give a reproducible electrode surface by successive cyclic voltammetric sweeps until the voltammograms were kept unchanged.

3. Results and discussion

3.1. Electrochemical behavior of procaine

The electrochemical behavior of procaine on the MWNT-coated GCE was examined by cyclic voltammetry (CV). Fig. 1 shows the cyclic voltammograms of a MWNT-modified GCE in phosphate buffer at pH 7.0 in the absence and presence of procaine. Within the potential window from 0.2 to 1.0 V, no redox peak was observed (dotted line). However, in the case of 2×10^{-5} mol/L procaine, a well-defined oxidation peak with very high current is observed at 0.75 V (solid line). Nevertheless, the oxidation peak current of procaine shows a remarkable decrease during the successive cyclic voltammetric sweeps. After the second sweep, the peak current decreases slightly and finally remains unchanged. This phenomenon may be caused by the fact that the adsorption of procaine or its oxidative product occurs at the MWNT-modified GCE surface. Thus, the oxidation peak current in the first anodic sweep was recorded for procaine analysis in the following studies. Fig. 1 also shows that there is no corresponding reduction peak during the reverse potential scan from 1.0 to 0.2 V.

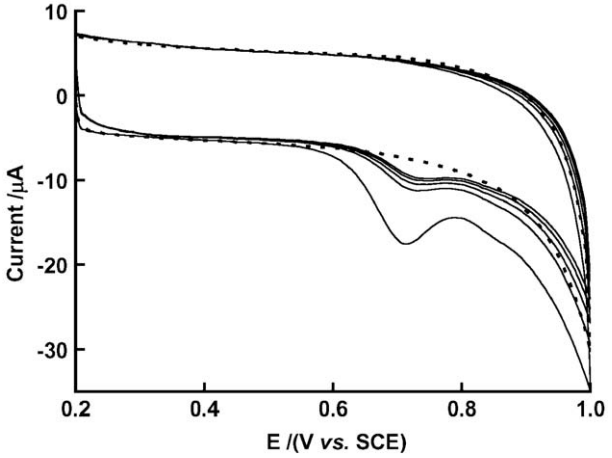


Fig. 1. Cyclic voltammograms of a MWNT-coated GCE. Dotted line: in 0.1 mol/L phosphate buffer at pH 7.0; solid line: dotted line $+2\times10^{-5}$ mol/L procaine. Scan rate: 100 mV/s.

Moreover, the electrochemical behaviors of procaine at different scan rates of 10, 25, 50, 150, 200 mV/s were also investigated by CV (not shown), showing much similarity with that at 100 mV/s. Only an oxidation peak was observed even at low scan rate (10 mV/s), suggesting that the electrode reaction of procaine under these conditions is totally irreversible.

In order to further elucidate the fascinating properties and potential of MWNT-coated GCE in the determination of procaine, the electrochemical behaviors of procaine at three different working electrodes (i.e. bare GCE, DHP-modified GCE and MWNT-modified GCE) were compared by linear sweep voltammetry (LSV), and the results are shown in Fig. 2. At a bare GCE, a poorly defined oxidation peak with very low current was observed at 0.82 V for 1×10^{-6} mol/L procaine after 4-min open-circuit accumulation (curve a). Under identical conditions, the oxidation peak of procaine almost vanishes at a DHP-modified GCE (curve b). Dihexadecyl hydrogen phosphate (DHP) can form a perfect thin film on GCE surface, and thus blocks the mass transportation and electron transfer of procaine, therefore, the peak current conversely decreases compared with that of bare GCE. However, the oxidation peak current of procaine at the MWNT-coated GCE increases significantly, and its oxidation peak potential concurrently shifts negatively from 0.82 V to 0.75 V, in comparison with that of bare GCE. Both the remarkable peak current enhancement and the negative shift of oxidation peak potential are undoubtedly attributed to the unique characteristics of MWNT, and strongly verify that MWNT-coated GCE exhibits electrocatalytic activity to the oxidation of procaine. In conclusion, a MWNT-modified GCE greatly improves the sensitivity of determination of procaine.

3.2. Influence of pH value

In 0.1 mol/L phosphate buffer, the electrochemical behaviors of procaine at different pH values were examined. It was found that the oxidation peak current gradually increases as pH increases from 5.0 to 7.0, and then shows a slight decline as pH value continuously increases to 8.0. Otherwise, the oxidation peak of procaine at pH 7.0 is best-shaped. Therefore, 0.1 mol/L phosphate buffer at pH 7.0 was used in all cases.

In addition, the relationship between pH value and the oxidation peak potential (E_{pa}) has also been studied. The pH value strongly affects E_{pa} of procaine. It shows that the E_{pa} shifts toward more negative potentials as pH increases from 5.0 to 8.0, and a good linear relationship was observed between pH value and E_{pa} with a slope of -56 mV/pH. The slope of -56 mV/pH indicates that the number of protons and electrons involved in the oxidation of procaine is equal.

3.3. Effect of the amount of MWNT–DHP suspension

Generally speaking, the thickness of the MWNT–DHP cast film on GCE surface, which is determined by the amount of MWNT–DHP suspension, has certain effects on the current response of procaine. There is no exception in this work. Fig. 3 depicts the variation of oxidation peak current of procaine as a function of the amount of MWNT–DHP suspension. When the amount of MWNT–DHP suspension increases from 0 to 5 μL, the oxidation peak current increases notably, but with further increase of the amount of MWNT–DHP suspension to 12.5 μL, the oxidation peak current increases slightly. If the amount of MWNT–DHP suspension exceeds 12.5 μL, the oxidation peak current conversely shows gradual decline. DHP, an insulator, lowers the electrical conductivity of MWNT film and the electron-transfer rate of procaine. Hence, the peak current decreases when the MWNT–DHP film is too thick.

3.4. Accumulation conditions

Accumulation, either open-circuit or close-circuit accumulation, is a common and effective tool to improve

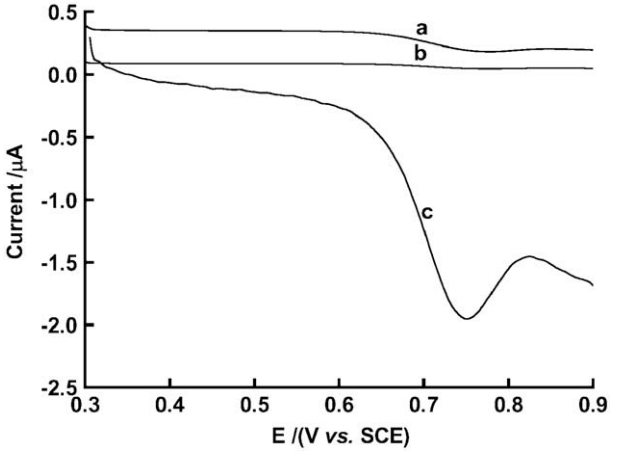


Fig. 2. Linear sweep voltammograms of 1×10^{-6} mol/L procaine in pH 7.0 phosphate buffer at three different electrodes after 4-min open-circuit accumulation. Curve (a): bare GCE; curve (b): DHP film-modified GCE; curve (c): MWNT–DHP film-modified GCE. Scan rate: 100 mV/s.

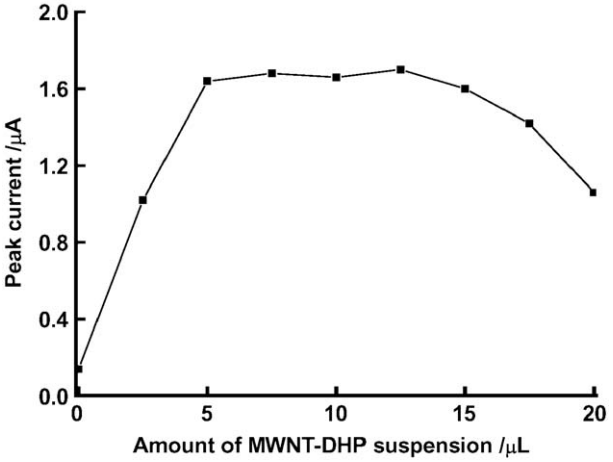


Fig. 3. Influence of the amount of MWNT–DHP suspension on the oxidation peak current of 1×10^{-6} mol/L procaine. Other conditions are the same as in Fig. 2.

determining sensitivity. The influence of accumulation potential on the oxidation peak current of procaine was examined on a MWNT-coated GCE. The oxidation peak current of 1×10^{-6} mol/L procaine was compared after 4-min accumulation under different potentials from -0.40 to 0.40 V, and the peak current was almost kept unchanged, implying that the accumulation potential had no influence on the oxidation peak current of procaine. Thus, an open-circuit accumulation was employed.

Unlike the accumulation potential, the accumulation time has obvious effects on the oxidation peak current of procaine. Fig. 4 illustrates the relationship between the accumulation time and the oxidation peak current. The oxidation peak current increases greatly within the first 4 min and then levels off, revealing that MWNT cast film exhibits effective accumulation for procaine.

3.5. Electrode reaction mechanism of procaine

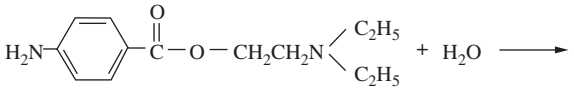
In order to get the electrode process of procaine at the MWNT-coated GCE, the electrochemical behaviors of 1×10^{-5} mol/L procaine in pH 7.0 phosphate buffer without accumulation were investigated by LSV at different scan rates from 25 to 400 mV/s. It was found that the oxidation peak current varies linearly with the scan rate, indicating that the electrochemical oxidation of procaine is a typical surface adsorption process.

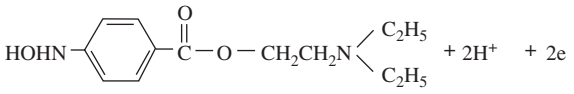
As to an irreversible electrode process, the oxidation peak potential (E_{pa}) shifts with scan rate (v) in LSV. Based on this, the number of electrons involved in the oxidation of procaine can be evaluated. Form the slope of E_{pa} with $\log(v)$, the αn_α is calculated to be 0.98. Generally, the electron transfer coefficient (α) is about 0.5 in totally irreversible electrode process. Therefore, the value of n_α is 2. Combining the former result that the identical number of protons and electrons were transferred in the oxidation process, the oxidation of procaine belongs to a two-electron and two-proton process.

In order to testify this, the procaine solution was exhaustively electrolyzed at 0.80 V until the electrolytic current was close to zero, and the quantity of consumed electric charge (Q_1) was recorded. Similarly, the blank solution was also electrolyzed under the same conditions, and the quantity of consumed electric charge (Q_2) was achieved. Then, the charge difference ($\Delta Q = Q_1 - Q_2$) represents the quantity of charge consumed in the oxidation of procaine.

$$\Delta Q = nFCV. \quad (1)$$

Here, C and V represent the concentration and volume of the procaine solution, respectively. F and n have their conventional meanings. From Eq. (1), the number of electrons (n) involved in the oxidation of procaine is calculated to be 1.92. Therefore, the possible half-reaction of procaine is as follows:





3.6. Calibration curve

The calibration curve for procaine was measured by LSV. The linear segment increases from $5.0\times10^{-7}-1\times10^{-4}$ mol/L with a regression equation of $i_p = 0.12 + 1.52\times10^6\ C$ ($r = 0.998$, C in mol/L, i_p in μA). Experiments showed that this method can detect 2×10^{-7} mol/L procaine after 4-min accumulation. The relative standard deviation (RSD) of

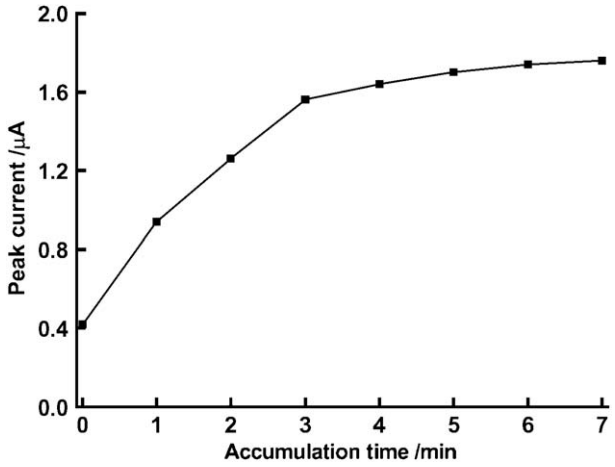


Fig. 4. Variation of the oxidation peak current of 1×10^{-6} mol/L procaine versus the accumulation time. Other conditions are the same as in Fig. 2.

Table 1
Determination of procaine in procaine hydrochloride injections

Samples	Declared procaine content (mg/mL)	Detected by literature [1] (mg/mL)	Detected by this method (mg/mL)	Recovery of this method (%)
A	20.00	20.06	19.88	102.6
B	20.00	19.92	19.94	99.6
C	20.00	20.08	20.10	101.4
D	10.00	10.06	9.84	99.2
E	10.00	9.94	10.20	102.8

5.0% for 1×10^{-6} mol/L procaine ($n=8$) showed good reproducibility.

The long-term stability of a MWNT-coated GCE was estimated by measuring the current response at a fixed procaine concentration of 1×10^{-6} mol/L over a period of 3 weeks. The MWNT-coated GCE was used daily and stored in air. The experimental results show that the current response only deviates 5.4%, suggesting that the MWNT-coated GCE reported in this work possesses long-term stability.

3.7. Interference

To evaluate the interferences of some foreign species on the determination of procaine at 1×10^{-6} mol/L level, systematic studies were performed. It was found that this method has good selectivity to the determination of procaine. For example, 20-fold concentrations of vitamin B_6, ascorbic acid (AA), uric acid (UA), xanthine (XA), dopamine (DA), vitamin A, 50-fold concentrations of vitamin E, vitamin B_1, progesterone, caffeine, and 200-fold concentrations of Zn^{2+}, Al^{3+}, Fe^{3+}, Cd^{2+}, Pb^{2+}, did not interfere with the current response of 1×10^{-6} mol/L procaine (signal change below 5%).

3.8. Determination of procaine in procaine hydrochloride injection

The MWNT-coated GCE was directly used to determine the content of procaine in procaine hydrochloride injections (The 7th Pharmaceutical Corporation of Shanghai, China). The procaine hydrochloride injection was just diluted by 20 times with the pH 7.0 phosphate buffer. After that, 10 μL of the diluted procaine hydrochloride injection was added into an electrochemical cell containing 10 mL pH 7.0 phosphate buffer, and the voltammograms were recorded as in standard procaine solutions described in analytical procedure. The content of procaine was calculated by the standard addition method, and the results shown in Table 1. Otherwise, typical method [1] was also used to detect procaine content in order to prove the accuracy of this proposed method. The results obtained by the MWNT-modified GCE are in good agreement with the results obtained by extration-spectrophotometric method and the declared procaine content. Furthermore, in order to establish the suitability of the proposed method, known amounts of the standard procaine were added into the analytical solution, and the same procedure was applied. The recoveries indicate that the accuracy of this proposed voltammetric method is excellent. From the experimental results, it is very clear that this novel MWNT-coated GCE has great potential for the determination of procaine in practical sample analysis.

4. Conclusion

In this work, a novel, chemically modified electrode, MWNT-film coated GCE, was easily fabricated for the determination of procaine. Owing to the unique properties of MWNT, such as high specific surface area, subtle electronic properties and strong adsorptive ability, the MWNT-modified GCE greatly enhances the oxidation peak current of procaine, and lowers the oxidation overpotential. As a result, a very sensitive and simple electrochemical method was developed for procaine determination.

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