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The Determination of Dopamine Using Glassy Carbon Electrode Pretreated by a Simple Electrochemical Method

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In this study, the pretreated glassy carbon electrode (GCE) was prepared by electrochemical oxidation firstly at +1.75 V for 300 s, and then by electrochemical reduction at –1.75 V for 300 s in 0.1 mol L^{-1} pH 7.0 phosphate buffer solution (PBS) and used for the determination of dopamine (DA) by cyclic voltammetry (CV). The pretreated GCE gives 100-fold greater current responses for dopamine compared with unpretreated GCE. The effect of pH, pretreated mode, scan rate and concentration of dopamine on the peak current was investigated, and the results indicated that the peak current of dopamine is the highest in 0.1 mol L^{-1} pH 7.0 PBS and the electrode reaction corresponds to a rate-controlled process. The peak current of the anodic peak and the concentration of dopamine hydrochloride is linear in the range of $1.0 \times 10^{-7} – 9.0 \times 10^{-6}$ mol L^{-1} and $1.2 \times 10^{-5} – 8.0 \times 10^{-5}$ mol L^{-1} with correlation coefficients of 0.9973 and 0.9980, and the detection limit is 3.0×10^{-8} mol/L. It has been successfully applied to the determination of dopamine in dopamine hydrochloride injection with recoveries ranging from 98 to 103%. The proposed method possesses the distinct advantages of simple, appropriate for operation, good reproducibility and cheap instrument.

Keywords: Pretreated GCE, cyclic voltammetry, dopamine, determination

1. INTRODUCTION

Dopamine (DA) is an important neuron transmitter compound widely existed in the brain for message transfer in the mammalian central nervous system [1,2]. Abnormal concentration levels of DA

may lead to several diseases, such as Parkinson's disease, schizophrenia and HIV infection [1-4]. Dopamine is currently the subject of intense research focus by chemists and neuroscientists. Therefore, it is of great significance to develop simple and rapid determination methods of dopamine.

Dopamine can be oxidized by electrochemical methods, thus electrochemical methods are usually used for the determination the concentration of dopamine in different solution. However, the rate of the electrochemical reactions is significantly influenced by the nature of the electrode surface [5]. Generally, the electron-transfer rate is slow and the electrochemical response signal of the analytes is not obvious at low concentration at the bare electrode surface, which leads to the development of several surface modification methods based on chemical [6-8], electrochemical [9,10] as well as electrochemical oxidative treatments [11-14]. The electrochemically pretreated methods are a kind of oxidative approaches for electrodes surface based on electrochemical methods. The methods holds the virtues of simple, reliable, and cheap instrument, and have been used for the determination DNA[11], guanine and adenine in DNA[12], aminophenol isomers[13] and hydroquinone and catechol [14], etc. The electrochemically pretreated electrodes are usually prepared by electrochemical oxidation at +1.75 V (vs. SCE) for 300 s in pH 5.0 phosphate buffer solution [11], +1.8 V (vs. SCE) for 300 s in pH 5.0 phosphate buffer solution [12], cyclic potential scanning from –0.6 to +1.6 V (vs. Ag/AgCl) in 0.1 mol L^{-1} phosphate buffer solution at a scan rate of 0.1 V s^{-1} for 40 cycles [13], at 2.0 V for 900 s in pH 7.0 phosphate buffer solution [14], or cyclic scan in 0.1 mol L^{-1} H_2SO_4 between 0 and 2 V (vs. Ag/AgCl) at 0.1 V s^{-1} [15].

In our work, the electrochemically pretreated GCE was prepared by electrochemical oxidation firstly at +1.75 V for 300 s, and then electrochemical reduction at –1.75 V for 300 s in 0.1 mol L^{-1} pH 7.0 PBS. The results indicated that the peak current of dopamine is increased 100-fold at the electrochemically pretreated GCE compared with on unpretreated GCE.

2. EXPERIMENTAL

2.1. Chemicals and instrumentation

Cyclic voltammetry (CV) was carried out with a CHI 660D electrochemical analyzer (Chenhua Instruments, China). Three-electrode system was used for the electrochemical experiment, containing a bare or pretreated GCE (3 mm diameter for bare GCE), a saturated calomel reference electrode (SCE) and a platinum wire counter electrode, respectively. The pH values were measured with a PHS–3C pH meter (Shanghai, China).

All chemicals were at least of analytical grade and were purchased from Shanghai Chemicals Co., Ltd. (Shanghai, China) unless otherwise stated and used as received without further purification. Dopamine hydrochloride was purchased from Sigma–Aldrich (St. Louis, MO, USA). The standard solution was prepared using dopamine hydrochloride and stocked at 4 °C. Dopamine hydrochloride injection was obtained from Jiuan Pharmaceutical Co., Ltd., Wuhan (labeled 2.0 mg mL^{-1}). The

phosphate buffer solution was prepared with potassium dihydrogen phosphate. The aqueous solutions were prepared in Millipore ultrapure water. All the experiments were performed at room temperature.

2.2. Preparation of pretreated GCE

The pretreated GCE was prepared by electrochemical oxidation firstly at +1.75 V for 300 s, and then by electrochemical reduction at –1.75 V for 300 s in 0.1 mol L^{-1} pH 7.0 PBS. After this, the pretreated GCE was scanned for 20 circles by cyclic voltammetry between –0.1 V and 0.5 V until stable cyclic voltammetric curve obtained for further use.

2.3. Procedure

20 mL of PBS and certain volume of dopamine hydrochloride stock solution or dopamine hydrochloride injection solution were added into the electrolytic cell, mixed and the cyclic voltammetric curves were recorded between –0.1 and 0.5 V at different scan rate.

3. RESULTS AND DISCUSSION

3.1 The electrochemical behavior of DA

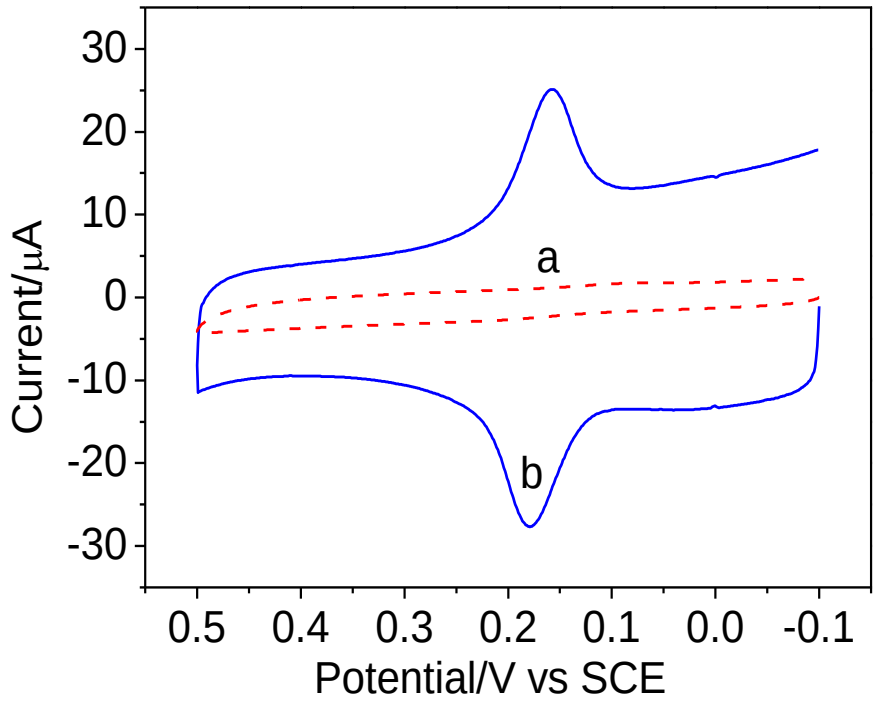
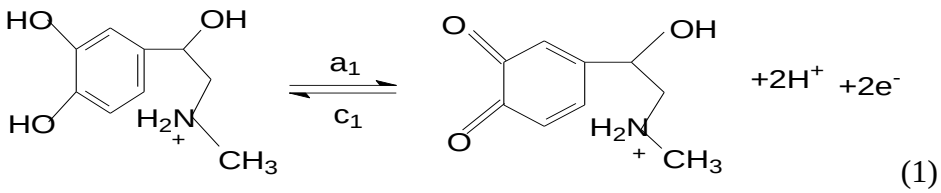


Figure 1. Cyclic voltammograms of dopamine at a bare GCE (a) and a pretreated GCE (b). Buffer solution: 0.1 mol L^{-1} pH 7.0 PBS, C_{DA}: 2.0×10^{-6} mol L^{-1}. Scan rate: 0.2 V/s.

The electrochemical behavior of dopamine was studied at a bare GCE or an electrochemically pretreated GCE. It can be seen that dopamine gives a pair of redox peaks both at a bare GCE (Fig 1 a) and a pretreated GCE (Fig 1 b) in the potential range of –0.1 V to 0.5 V. The peak current is obviously increased at the pretreated GCE compared with at the bare GCE, while the peak potential difference is decreased. The peak potentials are E_{pa}=0.078 V, and E_{pc}=0.210 V at the bare GCE, while E_{pa}=0.179 V, and E_{pc}=0.156 V at the pretreated GCE. These are likely owing to the electrocatalytic oxidation of the pretreated GCE to dopamine. The electrocatalytic oxidation mechanism of dopamine at the pretreated GCE is shown in equation (1).



(1)

Dopamine Dopamine quinone

3.2 Influence of pretreated mode on the performance of GCE

The influence of pretreated mode on the performance of GCE was studied in 0.1 mol L^{-1} pH 7.0 PBS.

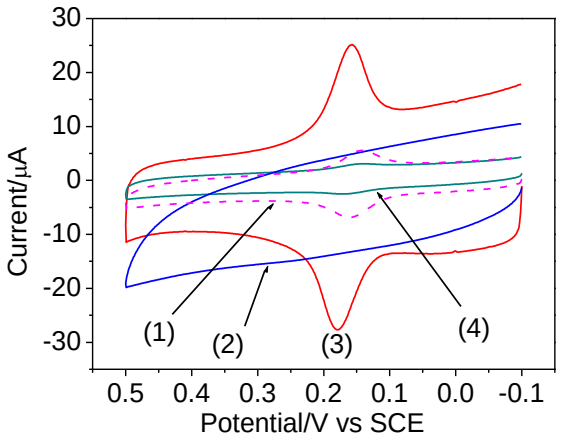


Figure 2. Cyclic voltammograms of dopamine at the different pretreated GCE. Pretreated modes: (1) +1.75 V for 300 s, (2) –1.75 V for 300 s, (3) first +1.75 V for 300 s, and then –1.75 V for 300 s, (4) first –1.75 V for 300 s, and then +1.75 V for 300 s; Buffer: 0.1 mol L^{-1} pH 7.0 phosphate solution; C_{DA}: 2.0×10^{-6} mol L^{-1}.

The pretreated modes for GCE are electrochemical oxidation only at +1.75 V for 300 s, electrochemical reduction only at –1.75 V for 300 s, first electrochemical oxidation at +1.75 V for 300

s, and then electrochemical reduction at –1.75 V for 300 s, first electrochemical reduction at –1.75 V for 300 s, and then electrochemical oxidation at +1.75 V for 300 s, respectively. After pretreated, the GCE was scanned for 20 circles by cyclic voltammetry between –0.1 V and 0.5 V until stable cyclic voltammetric curve obtained. It can be seen from Fig. 2 that the peak current of dopamine is the highest at the pretreated GCE when the pretreated mode of first electrochemical oxidation at +1.75 V for 300 s, and then electrochemical reduction at –1.75 V for 300 s is selected.

3.3 Influence of solution pH

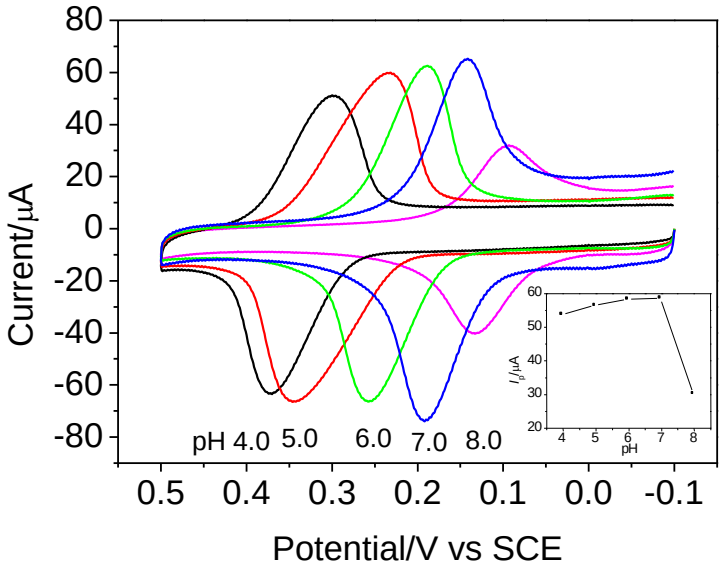


Figure 3. The effect of buffer pH on the peak current of dopamine. Buffer: 0.1 mol L^{-1} PBS. pH: 4.0, 5.0, 6.0, 7.0, 8.0. C_{VB2}: 2.0 × 10^{-5} mol L^{-1}. Inset: the relationship between the oxidation peak current and the pH.

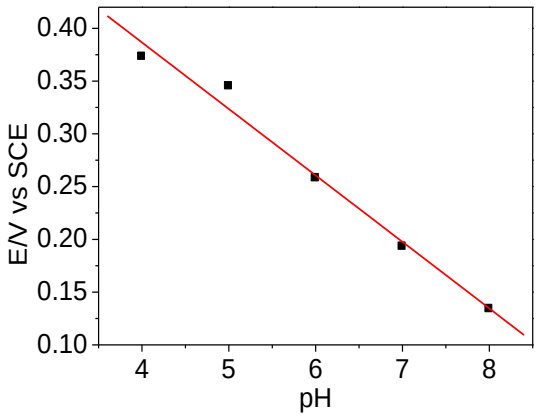


Figure 4. The relation between peak potential of dopamine and pH. Buffer: 0.1 mol L^{-1} PBS. pH: 4.0, 5.0, 6.0, 7.0, 8.0. C_{VB2}: 2.0 × 10^{-5} mol L^{-1}.

The influence of PBS pH on the peak current and peak potential was investigated. It can be seen from Fig 3 that the peak current of dopamine is increased with the increase of pH, and the peak current of dopamine reached the highest value when the pH of PBS is 7.0. Thus, the subsequent determination experiment was performed in 0.1 mol L^{-1} pH 7.0 PBS. The oxidation peak potential of dopamine is shifted to negative with the increase of pH (Fig. 4) indicates that protons take part in the electrode reaction. The regression equation is E(V)=–0.063pH + 0.64 with a correlation coefficient of 0.9917. A slope of 0.063 V/pH implies that the electrochemical reaction of dopamine at the pretreated GCE is a two-electron transfer process coupled to two-proton transfer steps [16].

3.4 Influence of scan rate

To further investigate the mechanism of the electrochemical oxidation of dopamine at the pretreated GCE, the influence of scan rate (v) on the voltammetric response of dopamine was studied in detail. Fig. 5 shows the cyclic voltammograms of dopamine at different scan rate ranging from 25 to 800 mV/s. It is clear that both the redox peak currents are enhanced with increasing of the scan rate. As can be seen from Fig. 5 (inset), both the oxidation and reduction peak currents were increased with the increase of scan rate, and they are linearly proportional to square root of the scan rate in the range from 50 to 800 mV/s. The regression equations are I_{pa}(μA) = –6.402 $v^{1/2}$ + 29.92 (r=–0.9993) and I_{pc} (μA) =7.611$v^{1/2}$ + 45.60 (r = 0.9960). It suggests that the electrode reaction corresponds to a rate-controlled process.

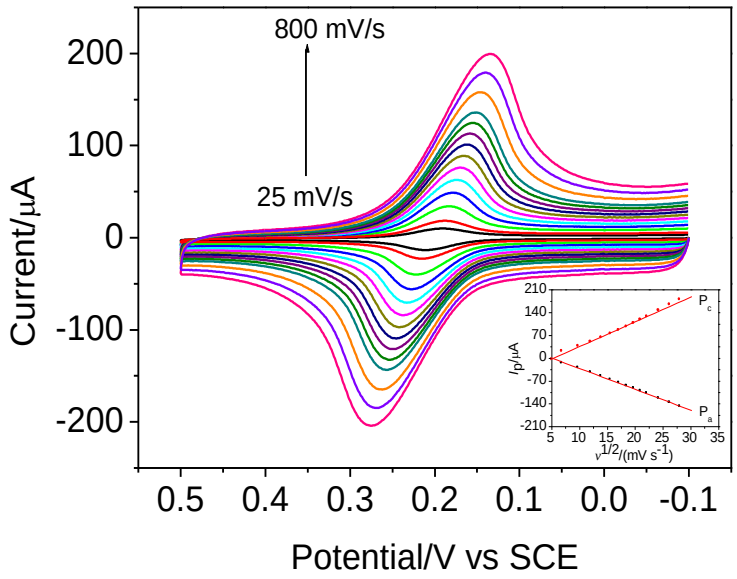


Figure 5. Effect of scan rate on the peak current of dopamine. Buffer: 0.1 mol L^{-1} pH 7.0 PBS. C_{VB2}: 2.0 × 10^{-5} mol L^{-1}. Inset: the relationship between the peak current of dopamine and the square root of scan rate.

3.5 Reproducibility

In order to inspect the reproducibility of the pretreated GCE, the precision experiment was carried out. Under the optimum conditions, the RSD (n=10) for 2.0×10^{-5} mol L^{-1} DA is 1.7%, which indicates that the pretreated GCE shows good reproducibility.

3.6 Interference experiment

The possible interference for dopamine determination was investigated including uric acid and ascorbic acid. As the peak current of dopamine is affected by ± 5% considered as interference. For 2.0×10^{-5} mol L^{-1} dopamine, 2 times of uric acid and 10 times of ascorbic acid do not disturb the determination of dopamine.

3.7 Calibration curve

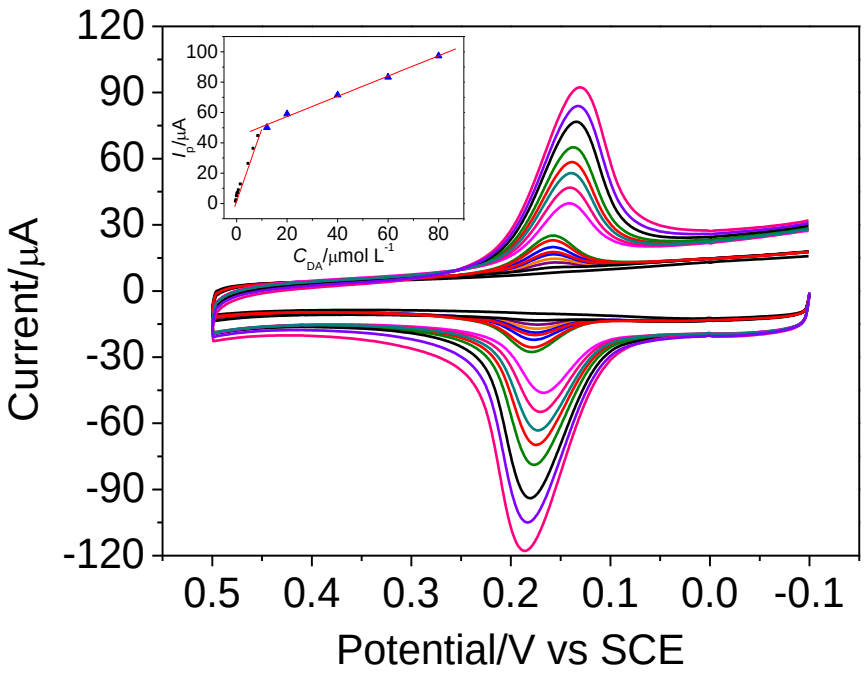


Figure 6. Cyclic voltammograms of dopamine changed with its concentration. Buffer: 0.1 mol L^{-1} pH 7.0 PBS. C_{VB2}: 2.0×10^{-5} mol L^{-1}. Inset: The relationship between peak current and concentration of dopamine. C_{DA} a→i: 0, 1.0×10^{-7}, 3.0×10^{-7}, 8.0×10^{-7}, 9.0×10^{-7}, 1.2×10^{-6}, 1.5×10^{-6}, 2.0×10^{-6}, 5.0×10^{-6}, 7.0×10^{-6}, 9.0×10^{-6}, 1.2×10^{-5}, 2.0×10^{-5}, 4.0×10^{-5}, 6.0×10^{-5} and 8.0×10^{-5} mol L^{-1}.

CV was used to investigate the relationship between the peak current and concentration of DA. Fig. 6 shows the cyclic voltammograms of pretreated GCE in 0.1 mol L^{-1} pH 7.0 PBS containing different concentration of dopamine. Obviously, the oxidation peak current of dopamine is enhanced

gradually with increasing its concentration in the range of 1.0×10^{-7}–9.0×10^{-6} mol L^{-1} and 1.2×10^{-5} – 8.0×10^{-5} mol L^{-1}. The corresponding regression equations can be expressed as I_{pa} (μA) = $4.77C_{DA}$ (μM) + 1.61 (r = 0.9980) and I_{pa} (μA) = 0.668 C_{DA} (μM) + 43.9 (r = 0.9973). The detection limit of this method for dopamine is 3.0×10^{-8} mol L^{-1} calculated as 3σ blank, which is lower than that at most of modified electrodes(Table 1), closed to that at graphite oxide bulk modified carbon paste electrode (1.5×10^{-8} mol L^{-1}) [16], multi-wall carbon nanotube-poly(3,5-dihydroxy benzoic acid) film modified electrode (1.0×10^{-8} mol L^{-1}) [17], poly(p-aminobenzene sulfonic acid)-modified glassy carbon electrode (2.0×10^{-8} mol L^{-1}) [18], and higher than at nano-Au self-assembly glassy carbon electrode (4.0×10^{-9} mol L^{-1}) [19]..

Table 1. Summary of the linear range and limit of detection for dopamine using different electrodes.

Modified electrode	Principle	Linear range(μmol L^{-1})	LD(μmol L^{-1})	Ref.
CDDA/GCE	ECO	5.0–280	0.29	1
PDBA/GCE	ECO	0.1–100	0.06	2
Tin hexacyanoferrate/CPE	ECO	200–25000		3
Oxidized GCE	ECO	19.7–98.8		15
Graphite oxide bulk/GCE	ECO	0.07–70	0.015	16
MWCNT/PDBA/GCE	ECO	0.1–70	0.01	17
PPABSA /GCE	ECO	0.1–1.0, 1.0–10, 10–100	0.02	18
Nano-Au/GCE		0.01–25	0.004	19
Nafion/choline Bi/CFE	ECO	0.38–16	0.1	20
Poly(taurine)/GCE		1.0–800	0.1	21
[Fe(pyterpy)2](SCN)2	ECO	2.0–740	1.0	22
2-MES/GE	ECO	10 –350	1.1	23
MWCNT/Chitosan/GCE	ECO	1.0–210	0.19	24
Polypyrrole/GME	ECO	25.0–1000, 0.5–10	0.1	25
CNB/GNCME	ECO		0.078±0.0065	26
Tiron/GCE	ECO	0.2–45.8	0.07	27
Polyethylene glycol/CPE	ECO		10	28
PB, PA, PEDOT, PEAVT	ECO	2.0–100		29
Fc-SWNT/GCE	ECO	5.0–30	0.05	30
Poly(sulfonazo III)/GCE	ECO	0.05–470.0	0.03	31
PICA /TCNQ/GCE	ECO	4.0–100	4.0	32
AgNP/CNTPE	ECO	0.8–64	0.3	33
PG/CPE	ECO		0.1	34
PGE		1.0–20	0.11	35
SDS/CPE		0.5–800	0.05	36
CILE	ECO	2.0–1500	2.0	37
PLA/GCE	ECO	0.8–500	0.3	38
Banana-MWCNTs /CPE		10–30	2.09	39
CoNSal/TOAB/CPE	ECO	1.0–100	0.5	40
MWCNT/GE	ECO	0.5–400	0.2	41
MWCNT/GCE	ECO	3–200	0.8	42
PdNP/CNFsE	ECO	0.5–160	0.2	43
PEDOT/PtE	ECO	0.5–25, 30–100	0.061	44
CPE/SDS	ECO	10–200	5	45

The linear range and limit of detection of dopamine at several modified electrode was summarized in table 1.

CDDA/GCE: Poly3-(5-chloro-2-hydroxyphenylazo)-4,5-dihydroxynaphthalene-2,7-disulfonic acid (CDDA) film glassy carbon electrode; ECO: Electrocatalytic oxidation; PDBA: Poly(3,5-dihydroxy benzoic acid); CFE: Carbon fiber electrode; CPE: Carbon paste electrode; GE: Gold electrode; MWCNT: Multi-walled carbon nanotube; GME: Graphene modified electrodes; CNB: carbon nanotubes; GNCME: Gold nanocomposite modified electrode; PB: Prussian blue; PA: polyazulene; PEDOT: Poly(3,4-ethylenedioxythiophene; PEAVT: poly(3-[(E)-2-azulene-1-yl)vinyl]thiophene); PICA: Poly(indole-6-carboxylic acid); PDBA: poly(3,5-dihydroxy benzoic acid); AgNP:Silver nanoparticles; CNTPE: carbon nanotube paste electrode; PG/CPE: Polyglycine modified carbon paste electrode; PGE: pyrolytic graphite electrode; SDS: Sodium dodecyl sulfate; CILE: carbon ionic liquid electrode; PLA: Poly(L-arginine); CoNSal: Cobalt salophen; TOAB: tetraoctylammonium bromide; PPABSA: Poly(p-aminobenzene sulfonic acid); PdNP: palladium nanoparticle; CNFs: carbon nano fibers; 2-MES: 2-mercaptoethanesulfunate.

3.8 Determination of dopamine in dopamine hydrochloride injection

Under the selected optimum conditions, the pretreated GCE was applied to determine DA in sample of dopamine hydrochloride injection (labeled 2.0 mg mL^{-1}, Jiuan Pharmaceutical Co., Ltd., Wuhan). The determination procedure was as follows: 20 μL of dopamine hydrochloride injection was taken using a microinjection, and added into 20 mL 0.1 mol L^{-1} pH 7.0 PBS. After the solution was mixed, the oxidation peak current of dopamine was measured. The determination results are listed in Table 2. The results show that the proposed methods could be efficiently used for the determination of DA in dopamine hydrochloride injection.

Table 2. Determination results of dopamine in dopamine hydrochloride injections (n=3).

Sample	Labeled (mg mL^{-1})	Found (mg mL^{-1})	Recovery (%)	Pharmacopoeia method (mg mL^{-1})
1	2.0	1.96	98	1.98
2	2.0	2.06	103	2.04
3	2.0	2.02	101	2.03

4. CONCLUSIONS

In this work, a simple, cheap and sensitive determination method for dopamine using electrochemically pretreated GCE was developed. The electrochemically pretreated electrode shows a

wider linear range $1.0 \times 10^{-7} – 9.0 \times 10^{-6}$ mol L^{-1} and $1.2 \times 10^{-5} – 8.0 \times 10^{-5}$ mol L^{-1} with correlation coefficients of 0.9973 and 0.9980, and the limit of detection is 3.0×10^{-8} mol L^{-1}, which was much lower than that at most of the previous reported modified electrode. The developed method had been successfully applied to determine dopamine in dopamine hydrochloride injection with satisfactory recoveries from 98% to 103%.

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