UNIVERSIDADE FEDERAL DO MARANHÃO

CENTRO DE CIÊNCIAS EXATAS E TECNOLOGIA

CURSO DE QUÍMICA LICENCIATURA

SENSOR ELETROQUÍMICO PARA DETECÇÃO DO ANTIDEPRESSIVO IMIPRAMINA EM BAIXO POTENCIAL BASEADO EM NANOTUBOS DE CARBONO OXIDADO, ÁCIDO FERROCENOCARBOXILICO, CICLODEXTRINA: APLICAÇÃO EM DROGAS PSICOTRÓPICAS E AMOSTRAS DE URINA

Antônio Gomes dos Santos Neto

São Luís

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Monografia apresentada ao Curso de Química Licenciatura da Universidade Federal do Maranhão para obtenção do grau de Licenciatura em Química.

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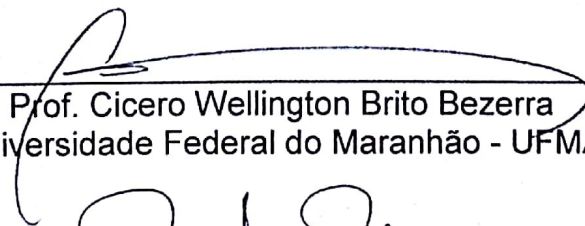
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RESUMO

Imipramina(IMP), uma droga antidepressiva tricíclica é comumente prescritra para tratamento de pacientes psiquiátricos que sofrem de diferentes formas de depressão. A dose apropriada da ingestão da droga é crucial para garantir os ótimos efeitos terapêuticos minimizando os efeitos colateráis e toxicidade. Por isso, o monitoramento da IMP é essencial para suas aplicações clínicas. Neste trabalho, nós reportamos um sensor eletroquímico baseado em um composto de ácido ferrocenocarboxilico(AFC), ciclodextrina(CD) e nanotubos de carbono de multiparedes oxidado(CNT) modificado em eletrodo de carbono vítreo para a detecção de IMP em baixos potenciais. O comportamento eletroquímico do sensor proposto foi caracterizado por microscopia eletrônica de varredura, espectroscopia RAMAM, e voltametria cíclica. Os resultados mostram que a determinação de IMP usando o sensor proposto ocorre em torno de 0 V vs Ag/AgCl em tampão fosfato pH 7.0. As curvas de calibração foram obtidas por voltametria cíclica e voltametria de pulso diferencial, com faixa linear de 10 a 350 µmol L^{-1} e 0.1 a 10 µmol L^{-1}, respectivamente. Um limite de detecção de 0.03 µmol L^{-1} foi obtido para a detecção de IMP. O sensor foi aplicado para a determinação de IMP em drogas psicotrópicas e amostras de urina e os resultados mostram um porcentagem de recuperação entre 99 e 101% para o analito.

Palavras-Chave: Antidepressivo. Imipramina. Sensor Eletroquímico. Ácido Ferrocenocarboxilico. Nanotubos de carbon oxidado.

ABSTRACT

Imipramine (IMP), a tricyclic antidepressant drug, is commonly prescribed for treatment of psychiatric patients suffering from different forms of depression. The appropriate amount of drug intake is crucial to ensure the optimum therapeutic effects minimizing severe collateral effects and toxicity. Therefore, the monitoring of imipramine is essential for its clinical applications. Herein, we report an electrochemical sensor based on a composite of ferrocenecarboxylic acid (FCA), β-cyclodextrin (CD), and oxidized multi-walled carbono nanotubes (f-CNT) modified glassy carbon electrode for detection of IMP at low potential. The electrochemical behavior of the proposed sensor was characterized by scanning eléctron microscopy, Raman spectroscopy, and cyclic voltammetry. The results show that imipramine determination using the proposed sensor occurs around 0 V vs Ag/AgCl in phosphate buffer pH 7.0. The calibration curves were obtained by cyclic voltammetry and differential pulse voltammetry, with linear ranges of 10 to 350 μmol L−1 and 0.1 to 10 μmol L−1, respectively. A detection limit of 0.03 μmol L−1 was obtained for the detection of IMP. The sensor was applied for IMP determination in psychotropic drugs and urine samples and the results show a recovery percentage between 99 and 101% for the analyte.

Keywords: Antidepressant. Imipramine. Electrochemical sensor.Ferrocenecarboxylicacid. Oxidized carbono nanotubes.

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Electrochemical sensor for detection of imipramine antidepressant at low potential based on oxidized carbon nanotubes, ferrocenecarboxylic acid, and cyclodextrin: application in psychotropic drugs and urine samples

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Abstract Imipramine (IMP), a tricyclic antidepressant drug, is commonly prescribed for treatment of psychiatric patients suffering from different forms of depression. The appropriate amount of drug intake is crucial to ensure the optimum therapeutic effects minimizing severe collateral effects and toxicity. Therefore, the monitoring of imipramine is essential for its clinical applications. Herein, we report an electrochemical sensor based on a composite of ferrocenecarboxylic acid (FCA), β-cyclodextrin (CD), and oxidized multi-walled carbon nanotubes (f-CNT) modified glassy carbon electrode for detection of IMP at low potential. The electrochemical behavior of the proposed sensor was characterized by scanning electron microscopy, Raman spectroscopy, and cyclic voltammetry. The results show that imipramine determination using the proposed sensor occurs around 0 V vs Ag/AgCl in phosphate buffer pH 7.0. The calibration curves were obtained by cyclic voltammetry and differential pulse voltammetry, with linear ranges of 10 to 350 μmol L^{-1} and 0.1 to 10 μmol L^{-1}, respectively. A detection limit of 0.03 μmol L^{-1} was obtained for the detection of IMP. The sensor was applied for IMP determination in psychotropic drugs and urine samples and the results show a recovery percentage between 99 and 101% for the analyte.

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Introduction

Psychotropic drugs are an important family of compounds whose application in therapy requires methods for the monitoring of their pharmaceutical dosage and amount in body biological fluids. Typical tricyclic psychotropic drugs are characterized by a tricyclic rings and presence of sulfur and nitrogen atoms. Tricyclic antidepressant drugs, which have a characteristic three-ringed nucleus structure, are commonly used to treat endogenous and reactive depression [1–4]. Additionally to the treatment of various forms of depression, tricyclic antidepressants also have been used to treat patients with chronic pain, anxiety, post-traumatic disorders, attention deficit disorders, and obsessive-compulsive disorders. Despite of their therapeutic actions, tricyclic antidepressants intake can cause side effects as such as blurred vision, chest pain and panic attacks [5]. An overdose from this kind of drugs can lead to more serious consequences including life-threatening arrhythmias or cardiac disturbances or even it can be lethal [6, 7]. Imipramine (IMP), sold as Tofranil and also known as melipramine, a tricyclic

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antidepressant acts through synaptic reuptake inhibition of the neurotransmitters serotonin and noradrenalin. It has a therapeutic range of 150–250 ng mL^{-1} (or ~ 535–1039 nmol L^{-1}), and it becomes toxic at concentrations higher than 450 ng mL^{-1} (or ~ 1605 nmol L^{-1}) [8]. This relatively narrow therapeutic range of IMP requires clinical monitoring for an effective and safe treatment. In this sense, biological fluids such as urine are promising samples for drug monitoring due to its ready availability and non-invasive collection in comparison to blood, serum, amniotic fluids, gastric contents, or specific tissues, among others.

A variety of analytical methods have been developed for the analysis of IMP in biological fluids or pharmaceutical preparations samples, such as liquid chromatography [4, 9, 10], capillary electrophoresis [11–13], spectrophotometry [14–16], gas chromatography-mass spectrometry [17, 18], and electrochemical [19–31].

The application of electrochemical methods in the analysis of drugs or pharmaceuticals and biological samples has increased greatly over the last few years. The renewed interest in electrochemical techniques can be attributed in part for the low cost, simplicity of operation, and ease control of the physical-chemical characteristics of the working electrode through its modification with different materials.

In this paper, an easy to fabricate electrochemical sensor for the quantitative determination of IMP in pharmaceutical and biological samples is presented. Herein, we demonstrate that a glass carbon electrode (GCE) modified with a composite based on ferrocene carboxylic acid (FCA), β-Cyclodextrin (CD), and oxidized multi-walled carbon nanotubes (f-CNT) is able to determine IMP in a very low potential, as low as 0.02 V, due mainly to its synergistic properties. The motivation of using this composite is to combine the well know exquisite electronic properties of the multi-walled carbon nanotubes [21–23] with a FCA molecule encapsulated in the cavity of CD. Ferrocene-based compounds are commonly used in electrochemistry due to the rapid response to electroactive species. However, ferrocene and its derivatives are not adsorbed in a stable form on electrode surfaces, in particular their oxidized forms, which presents high solubility in aqueous solutions. One approach to overcome this challenge is the formation of an inclusion complex between the ferrocene and cyclodextrin at the electrode surface [32]. An inclusion complex is formed when the guest molecule, in this case ferrocene carboxylic acid, is incorporated into the cavity of a host molecule such as cyclodextrin as result of chemical and steric factors. Therefore, the main aim of the simultaneous use of these three materials (FCA, CD, and f-CNT) is the development of an electrochemical sensor for the determination of imipramine at a low potential and with wide concentration range, thereby increasing the selectivity and sensitivity of the system for application for different samples.

Experimental

Reagents and solutions

β-Cyclodextrin (CD), ferrocenecarboxylic acid (FCA), Nafion® (Nf), imipramine (IMP), magnesium stearate, titanium dioxide, ascorbic acid, uric acid, dopamine, levodopa, sodium chloride, calcium nitrate, nickel nitrate, zinc sulfate, glucose, potassium ferricyanide, and potassium ferrocyanide⁻ were purchased from Sigma-Adrich. Potassium phosphate and sodium hydroxide were purchased from ISOFAR. Potassium chloride and aluminum oxide were purchased from FORTEL. All reagents were of analytical grade and their solutions were prepared with deionized water obtained using a GEHAKA ultrapurifier (OS100LXE GEHAKA Company).

Electrochemical and spectroscopic measurements

Electrochemical measurements were performed with an Autolab® PGSTAT 128 N potentiostat/galvanostat (Utrecht, the Netherlands) linked to a computer with GPES 4.9 software for data acquisition. Electrochemical responses of peak current and peak potential were obtained by cyclic voltammetry and differential pulse voltammetry techniques. Electrochemical impedance spectroscopy measurements were performed in 0.1 mol L^{-1} KCl solution containing 5 mmol L^{-1} of $[Fe(CN)_6]^{3-/4-}$. The EIS measurements were performed in the frequency range of 10^{-2} to 10^5 Hz under AC amplitude of 10 mV at the formal potential of redox probe. A three electrode electrochemical cell was employed for all electrochemical measurements. The working electrode was a modified glassy carbon electrode (GCE). The counter and reference electrodes were a platinum electrode and Ag/AgCl$_{(sat.)}$, respectively.

Synthesis, functionalization of multi-walled carbon nanotubes, and electrode surface modification

The multi-walled carbon nanotubes (CNT) were synthetized according to previous works described in the literature [33–36]. Briefly, a mixture of camphor and ferrocene was vaporized in the antechamber of a modified thermal chemical vapor deposition (CVD) furnace. The vapor was carried by a nitrogen gas flow to the chamber of the CVD furnace set at 850 °C and atmospheric pressure being converted into CNT powders [33]. The CNT powders were sonicated for 5 h in HCl and they were copiously washed with water. The iron-based catalytic particles were removed from the CNT powder by acid etching [34]. The introduction of oxygen-containing groups (–OH, –COOH, =O) and tip exfoliation of nanotubes were performed in a pulsed-DC plasma reactor with an oxygen flow rate of 1 sccm, at a pressure of 150 mTorr, − 700 V, at a frequency of 20 kHz. After this functionalization of the CNT it was designated as f-CNT.

The composite material was prepared from two stock solutions. The first one was prepared mixing 0.0010 g of f-CNT to 1 mL of deionized water, and the second one was prepared dissolving 0.0011 g of β-Cyclodextrin (CD) and 0.0021 g of ferrocenecarboxilic acid (FCA) in a mixture of 500 μL of ethanol and 500 μL of deionized water.

In addition, prior to the electrode modification, the glassy carbon electrode surface was polished and then cleaned by sonication to remove any adsorbed species. Then, 10 μL of previous prepared solution was placed on the electrode surface in order to modify the electrode, and the set was allowed to dry at 50 °C until the formation of a thin film on the electrode surface. Finally, 10 μL of Nafion (Nf) was placed over the film and allowed to dry again at 25 °C which was further designated as FCA-CD/f-CNT.

Results and discussion

Characterization of synthesized carbon nanotubes by SEM microscopy and Raman and FTIR spectroscopies

Figure 1a, b shows first- and second-order Raman spectra from as-grown CNT and f-CNT samples. Curve fitting was carried out using Lorentzian functions for the D and G bands and 1622 cm^{-1} (D' shoulder). On the other hand, curve fitting with Gaussian functions were performed for the bands around 1250 (#) and 1480–1520 (*). The D band can be associated to disorder in carbon crystalites stemming from a double resonance process involving phonon and defect. The G-band has E_{2g} symmetry corresponding to stretching vibrations in the basal plane associated to sp^2 domains of single-crystal graphene or nanocrystalline graphite. The high intensity G' band indicates that the synthetized material presents a high crystallinity of sp^2 carbon [37]. Two Gaussian peaks centered at around 1250 and 1480 cm^{-1} were added in the Raman first order in order to closely curve-fit the data. The origin of the 1480 cm^{-1} band is suggested to be correlated with the polar

groups attached on CNT walls, while the shoulder at 1250 cm^{-1} has its origin in double resonance process on graphene phonon dispersion curves [38]. In summary, Raman analyses indicate slight increase on defects of CNTs after oxygen plasma treatment and the attachment of oxygen-containing groups on their surfaces.

Figure 2a shows the scanning electron micrography of the as-grown CNTs. As can be seen, the as-grown CNTs presented diameters ranging from 20 to 50 nm. On the other hand, Fig. 2b shows defects caused by exfoliation and oxidation of CNT, which causes etch pits. FTIR spectra of the materials are shown in Fig. 2c. For the FCA monomers (spectrum c1), the peaks at 1654 cm^{-1} and 1286 cm^{-1} may be assigned to the C=O and C–O stretching vibrations of the carboxyl moieties, respectively. The ferrocenyl ring skeleton (C=C) vibrations can be found at 1476 and 1400 cm^{-1}, the cyclopentadienyl C–H vibrational stretch at about 3100 cm^{-1}, and bending vibration at 1160 cm^{-1} [39–41]. Figure 2c (spectrum c2) shows the FTIR spectrum of pure CD. The FTIR spectrum of pure cyclodextrin presented a broad band centered at about 1650 cm^{-1} due to the stretching vibrations of water molecules existing in the cavities of cyclodextrin [42]. The O–H stretching region (~ 3400 cm^{-1}) in FTIR spectrum of CD is highly complex since the primary and secondary hydroxyl groups of cyclodextrin may be bonded intramolecularly or intermolecularly as well as hydrogen bridges may also be formed between the water molecules complexed in the cavity of cyclodextrin [42]. In addition to previous peaks and bands, the frequencies for pure cyclodextrin were observed at about 2920 cm^{-1}, 1157 cm^{-1}, and 1030 cm^{-1} which correspond to the symmetric and antisymmetric stretching of $v(CH_2)$, $v(C-C)$, and bending vibration of $v(O-H)$, respectively [43]. Moreover, the FTIR spectrum of pure cyclodextrin presented high number of peaks and bands between 1460 and 440 cm^{-1} due to stretching and bending modes of skeletal vibrations of cyclodextrin structure, which has been discussed elsewhere [44].

Fig. 1 First and second-order Raman spectra from **a** as-grown CNT and **b** f-CNT
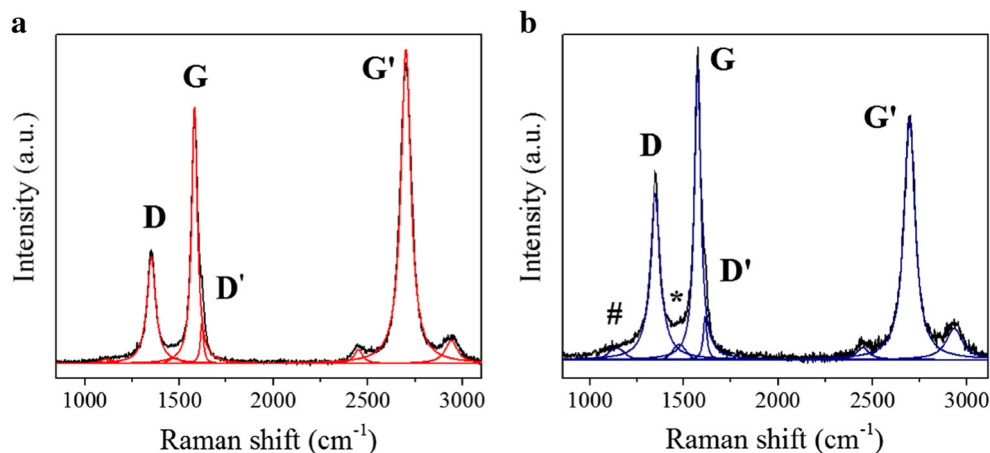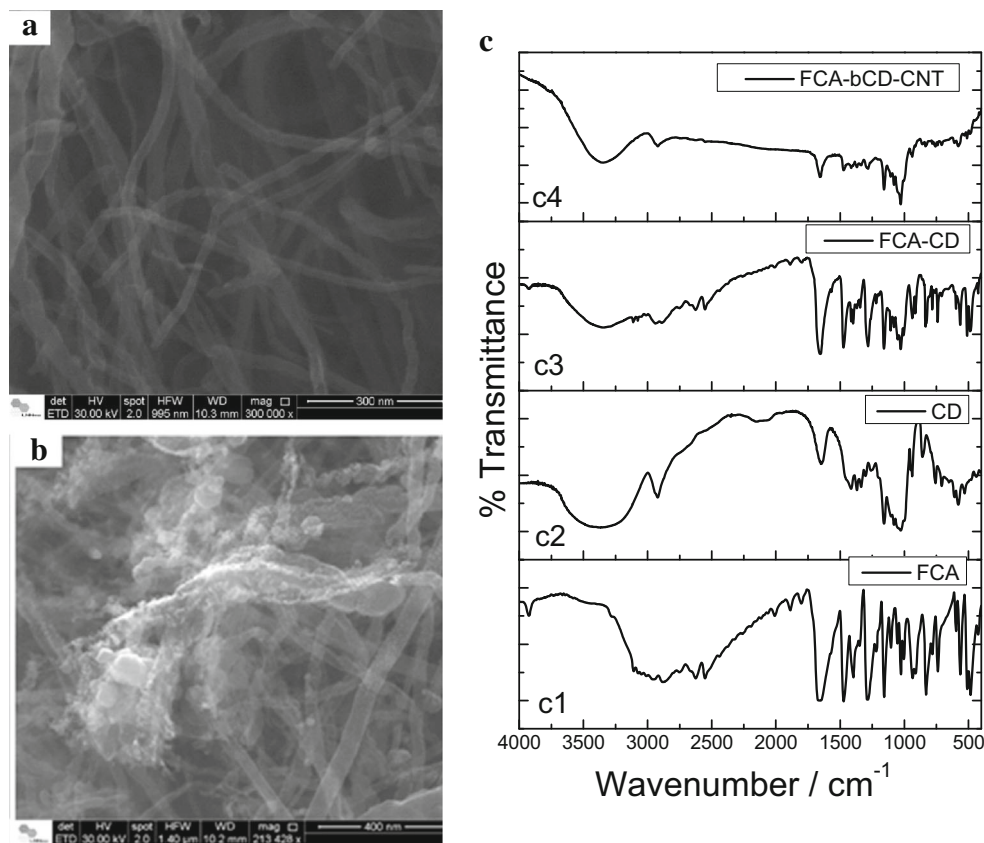
Fig. 2 SEM micrographs of **a** synthetized CNT and **b** synthetized/functionalyzed f-CNT; **c** FTIR spectra of FCA (spectrum c1), CD (spectrum c2), FCA-CD (spectrum c3), and FCA-CD/f-CNT (spectrum c4)

The IR spectra of FCA/CD (Fig. 2c—spectrum c3) and FCA/CD/f-CNT (Fig. 2c—spectrum c4) do not show some FCA absorption peaks upon complexation suggesting that the inclusion complex also involve the interaction between the hydroxyl groups of the host cavities and the FCA carbonyl group. Furthermore, the broad band at about 1650 cm^{-1} associated to the stretching vibrations of water molecules existing in the cavities of CD were not verified in FTIR spectra of FCA/CD and FCA/CD/f-CNT as consequence of formation of the inclusion complex between FCA and cyclodextrin reducing the amount of water molecules into the CD cavity. The FTIR spectrum of FCA/CD/f-CNT presented peaks of both compounds FCA and CD indicating that FCA molecules successfully entered the cavities of cyclodextrin in FCA/CD/f-CNT composite. The O–H stretching band originally strongly associated to included water in CD (3400 cm^{-1}) shift to about 3335 cm^{-1}, which can be associated to presence of hydroxyl moieties at the carbon surface.

Electrochemical characterization of the FCA-CD/-CNT sensor

Figure 3 shows the cyclic voltammograms of the glassy carbon electrode (GCE) modified with ferrocenecarboxilic acid and Nafion (FCA/Nf) (black line); ferrocenecarboxilic acid, cyclodextrin, and Nafion (FCA-CD/Nf) (blue line); and

ferrocenecarboxilic acid, cyclodextrin, oxidized carbon nanotubes, and Nafion (FCA-CD/f-CNT/Nf) (red line). The experiments were carried out in phosphate buffer solution (PBS) at pH 7 ranging the potential applied to the electrode from −0.1 to 0.7 V. As can be seen in Fig. 3, the cyclic voltammograms of all materials presented a well-defined redox couple related to the FCA electrochemical processes since cyclodextrin and f-CNT do not present faradaic processes in the investigated
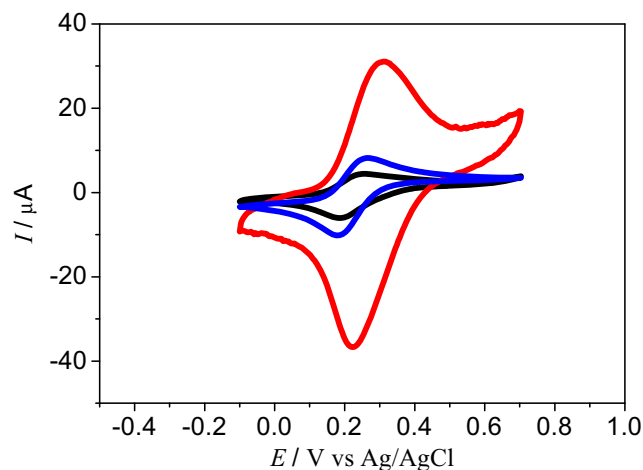

Fig. 3 Cyclic voltammograms of the GCE modified with FCA/Nf (black line), FCA-CD/Nf (blue line), and FCA-CD/f-CNT/Nf (red line). Experiments carried out in 0.1 mol L^{-1} PBS (pH 7). $v = 0.05$ V s^{-1}

potential range applied to the modified electrode. The FCA-CD/f-CNT/Nf modified electrode presented the highest faradaic peak currents when compared to the electrode modified only with FCA/Nf or FCA-CD/Nf. The higher peak current observed with FCA-CD/f-CNT/Nf modified electrode can be related to the high porosity, surface area to volume ratio, and electrical properties of the f-CNT as well as it can also be associated to the capability of the cyclodextrin to form an inclusion complex with FCA.

Stability of the FCA/Nf, FCA-CD/Nf, and FCA-CD/f-CNT/Nf materials

In order to verify the stability of each proposed material, the GCE was modified with FCA/Nf, FCA-CD/Nf, and FCA-CD/f-CNT/Nf, respectively. Thus, successive cyclic voltammograms were performed for each material in 0.1 mol L^{-1} PBS at pH 7 and scan rate of 0.05 V s^{-1}. Figure 4 shows 20 cyclic voltammograms of the GCE modified with (i) FCA/Nf (Fig. 4a), (ii) FCA-CD/Nf (Fig. 4b), and (iii) FCA-CD/f-CNT/Nf (Fig. 4c) in the potential range of −0.1 to 0.7 V vs Ag/AgCl. As can be seen in Fig. 4, the cyclic voltammograms presented a redox couple referring to the FCA/FCA^{+} redox couple. However, after 20 cycles, a significant exponential decrease in the peak currents was observed to CGE modified with FCA/Nf (inset of Fig. 4a) and FCA-CD/Nf (inset of Fig. 4b), which suggests that the electroactive species leach out from the electrode surface.

In addition, Fig. 4b shows that the value of the peak current of the first cyclic voltammogram (22.7 μA) was about fourfold higher than that observed for the first cyclic voltamogram presented in Fig. 4a. However, the peak current gradually decreases for both cases until the 11th cycle and then they remain stable at low values of peak currents (1.34 μA in Fig. 4a and 0.41 μA in Fig. 4b). Although the cyclodextrin has contributed to the increase of the peak, it is probable that inclusion complex between cyclodextrin and the FCA leach out from the surface of the GCE.

On the other hand, Fig. 4c shows that the use of f-CNT combined to FCA-CD inclusion complex on the electrode surface resulted in a very stable electrochemical system. As can be seen, the peak current remains very stable after 20 cycles, demonstrating that the synergistic effect resulting from combined raw materials is of high importance to the stability of the film (inset of Fig. 4c). The mean value of the anodic and cathodic peak currents of last cyclic voltammogram presented in the insets of Fig. 4 were 0.16, 0.85, and 25.38 μA to FCA/Nf, FCA-CD/Nf, and FCA-CD/f-CNT/Nf, respectively. These results suggest that the use of FCA-CD inclusion complex combined to f-CNT results in higher peak currents probably due to the high adsorption capability of the oxidized carbon nanotubes to cyclodextrin molecules, which can be provided
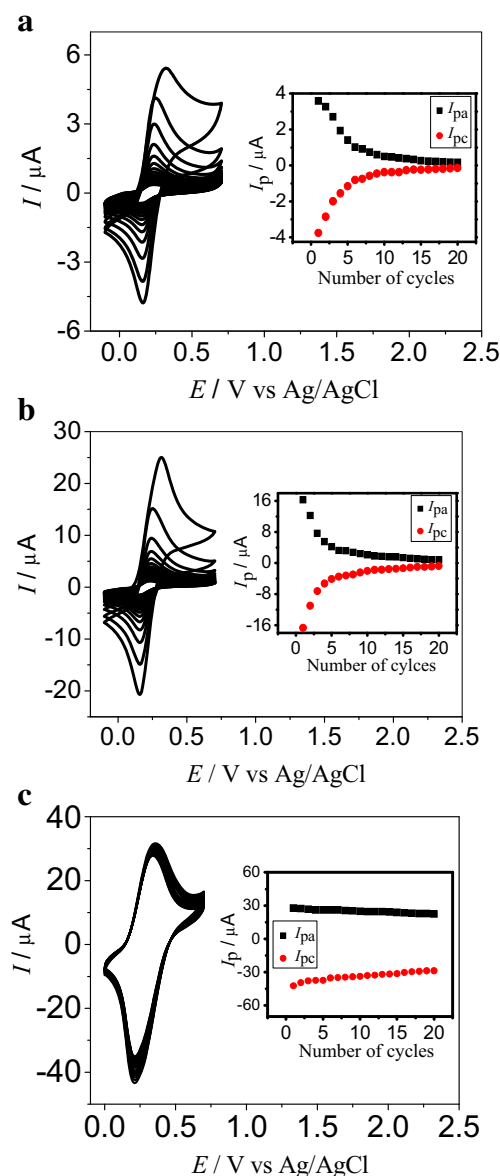
Fig. 4 Cyclic voltammograms of the GCE modified with **a** FCA/Nf, **b** FCA-CD/Nf, and **c** FCA-CD/f-CNT/Nf. Experiment were carried at 0.1 mol L^{-1} PBS, pH 7.0. $v = 0.05$ V s^{-1}

by van der Waals forces [45, 46]. Therefore, the FCA-CD/f-CNT/Nf was chosen for further experiments.

For a better understanding of the redox processes on the kinetic properties of the FCA-CD/f-CNT/Nf, the electronic transfer coefficient, α, and the apparent heterogeneous electron transfer rate constant, $k_{app}°$, referring to electron transfer between the electrode and the redox species were determined. Fig. S1a, b shows cyclic voltammograms obtained at several scan rates for FCA-CD/f-CNT/Nf modified GCE in 0.1 mol L^{-1} phosphate buffer at pH 7.0. As can be seen, the peak current increases linearly with the scan rate from 0.01 to 0.8 V s^{-1} indicating that the redox processes are typical of surface-confined processes.

Thus, the Laviron's theory [47] was applied to voltametric data in order to evaluate the values of α and k_{app}^o. Fig. S1c shows the plot of the peak potential (E_p–E_m) versus the log of the scan rate (v). For high scan rate values, it is observed a linear dependence between (E_p–E_m) and log scan rate, which allows the determination of the kinetic parameters k_{app}^o and α, from the curve slopes.

According to Laviron's theory, the slope of the linear segment is equal to $2.303RT/\alpha_c nF$ for cathodic process and $2.303RT/\alpha_a nF$ for the anodic process. The values calculated for α_c and α_a were 0.39 and 0.38, respectively. Therefore an average value of 0.385 for α was used in the subsequent study. Although the value of the electron transfer coefficient in reversible systems is 0.5, quasi-reversible systems present values of α between 0.3 and 0.7 [48]. Taking into account that the apparent heterogeneous electron transfer rate constant gives the magnitude of the electron transfer between the electrode and the redox probe, it was determined with the following equation [47]:

$$\log\left(k_{app}\right) = \alpha\log(1-\alpha) + (1-\alpha)\log(\alpha) \qquad (1)$$
$$-\log\left(RT\big/nFv\right) - \alpha nF(1-\alpha)\left(\frac{\Delta Ep}{2.3RT}\right)$$

where α is the mean value of the charge transfer coefficient, v is the scan rate and all other symbols have their conventional meaning. The k_{app}^o value obtained for the proposed sensor was 2.96 ($\pm$ 0.10) s^{-1}, which was similar to those previously reported [49–51].

## Electrochemical behavior of IMP on the modified electrode

Figure 5a shows the electrochemical behavior of the FCA-CD/f-CNT/Nf modified electrode in the absence (black line) and presence (red line) of IMP. As can be seen, the faradaic current of the peak at about 0.3 V decreases in presence of IMP, and a new redox couple appears at 0.04 V which currents increase in presence of the analyte. The decreasing of the anodic and cathodic peak currents in presence of IMP can be attributed to the high affinity of the CD to IMP, such as the drug can replace FCA in cavity of the host compound. On the other hand, the redox couple arising at 0.04 V can be attributed to the oxidation/reduction process of some FCA molecules released from CD cavity, which can probably be directly adsorbed on the surface of f-CNT. For the bare glassy carbon electrode in the absence and presence of the analyte, no change was observed upon analyte addition (blue and green voltammograms in Fig. 5a. Figure 5b, c shows a schematic representation of the FCA-CD/f-CNT/Nf modified electrode in the absence (b) and presence (c) of IMP.

Electrochemical determination of imipramine

Cyclic voltammograms were performed in order to investigate the behavior of the FCA-CD/f-CNT modified GCE under successive additions of IMP in the electrochemical cell. Under optimized operational conditions, the analytical performance of the FCA-CD/f-CNT modified GCE for the determination of IMP was evaluated by cyclic voltammetry (Fig. 6) and differential pulse voltammetry (Fig. 7). Figure 6a shows the cyclic voltammograms for the GCE modified with FCA-CD/f-CNT/Nf obtained to successive additions of IMP to electrochemical cell in the following concentrations: 10, 20, 30, 40, 70, 100, 150, 200, 250, 300, and 350 µmol L^{-1}. As can be seen, the anodic and cathodic peak currents around 0.04 V increased with the increase of the concentration of IMP, which is related to the FCA redox reaction (redox couple I) due your release from the ciclodextrin cavity. On the other hand, it was observed a decrease of the anodic and cathodic peak currents for the second redox couple (at about 0.3 V) with the increase of the analyte concentration (Fig. 6b). The decrease in peak currents of redox couple II is according to proposed scheme in Fig. 5, since the increase in concentration of IMP can reduce the amount of FCA within the cavity of the CD. A linear relationship between the concentration of the analyte and the cathodic and anodic currents was observed only for the redox couple I (Fig. 6b). For this redox couple the linear equations obtained were $I_{pa}/\mu A$ = 1.60 ($\pm$ 0.068) + 0.010 ($\pm$ 0.004) [IMP]/µmol L^{-1} (R^2 = 0.997) for the oxidation process and $I_{pc}/\mu A$ = −1.20 ($\pm$ 0.060) − 0.010 ($\pm$ 0.003) [IMP]/µmol L^{-1} (R^2 = 0.998) for the reduction process. On the other hand, for the redox couple II, the peak currents were not linear with the IMP concentration (Fig. 6c). So, taking into account the couple redox I, the detection and quantification limit obtained by cyclic voltammetry were 3 and 10 µmol L^{-1}, respectively.

Figure 7a shows the differential pulse voltammograms (DPV) for the GCE modified with FCA-CD/f-CNT/Nf obtained for successive additions of IMP (0.10, 0.74, 1.30, 2.20, 4.20, 6.70, 8.40, and 10.00 µmol L^{-1}). As can be seen, the anodic currents around 0.02 V (peak I) increase (Fig. 7b), and the anodic currents around 0.20 V (peak II) decrease (Fig. 7c). A linear relationship between the peak current and the concentration of IMP was observed for the peak I ($I_p/\mu A$ = 0.014 ($\pm$ 0.003) + 0.096 ($\pm$ 0.006) [IMP]/µmol L^{-1}) with a correlation coefficient of 0.999. The calibration curve for determining IMP presented a sensitivity of 0.096 µA L mol^{-1}. This value is greater than or similar to many previously reported (Table 1). The good sensitivity was due to the efficient interaction between the film present on the electrode surface and the analyte in solution due to low resistance to electron transfer of the proposed platform combined to the

Fig. 5 **a** Cyclic voltammograms of GCE in the absence (blue line) and presence (green line) of IMP and GCE modified with FCA-CD/f-CNT/Nf in the absence (black line) and presence (red line) of IMP. Measurements carried out in 0.1 PBS, pH 7.4, scan rate = 0.05 V s^{-1}. Schematic representation of the FCA-CD/f-CNT/Nf modified electrode surface in **b** absence and **c** presence of IMP molecules
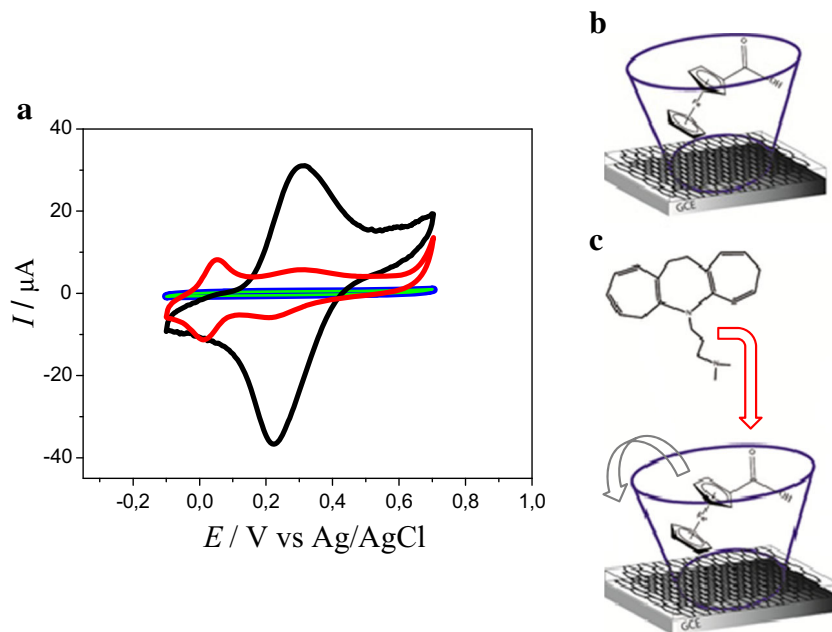

capability of the sensor to encapsulate the analyte. The detection and quantification limits were estimated from peak I. The detection and quantification limits were 0.03 and 0.1 μmol L^{-1}, respectively. In Table 1, the linear ranges of concentration for determination of the analyte are presented showing that the sensor can be applied for the determination of IMP in high (10 to 350 μmol L^{-1}) or low (0.1 to 10 μmol L^{-1}) concentrations in the samples.

Finally, the accuracy of the proposed sensor for the determination of IMP was investigated under optimized

experimental conditions. Addition and recovery experiments were performed by adding a known amount of the analyte to samples (Table S1). The results from the addition and recovery tests presented in Table 1 show that the proposed method has a good accuracy with recovery percentages between 98 and 101%.

The selectivity of the sensor was evaluated by examining the influences of several possible interfering substances on the detection of IMP in pharmaceutical formulation and biological fluids samples. Solutions of these compounds were freshly

Fig. 6 **a** Cyclic voltammograms of the FCA-CD/f-CNT/Nf modified electrode in presence of different concentrations of IMP (10, 20, 30, 40, 70, 100, 150, 200, 250, 300, and 350 μmol L^{-1}). **b** and **c** Plots of I_p (redox couples I and II) versus concentration of IMP. Both plots (b and c) were obtained from Fig. 6a. Experiments were carried out in 0.1 mol L^{-1} PBS, pH 7.0. $v = 0.05$ V s^{-1}
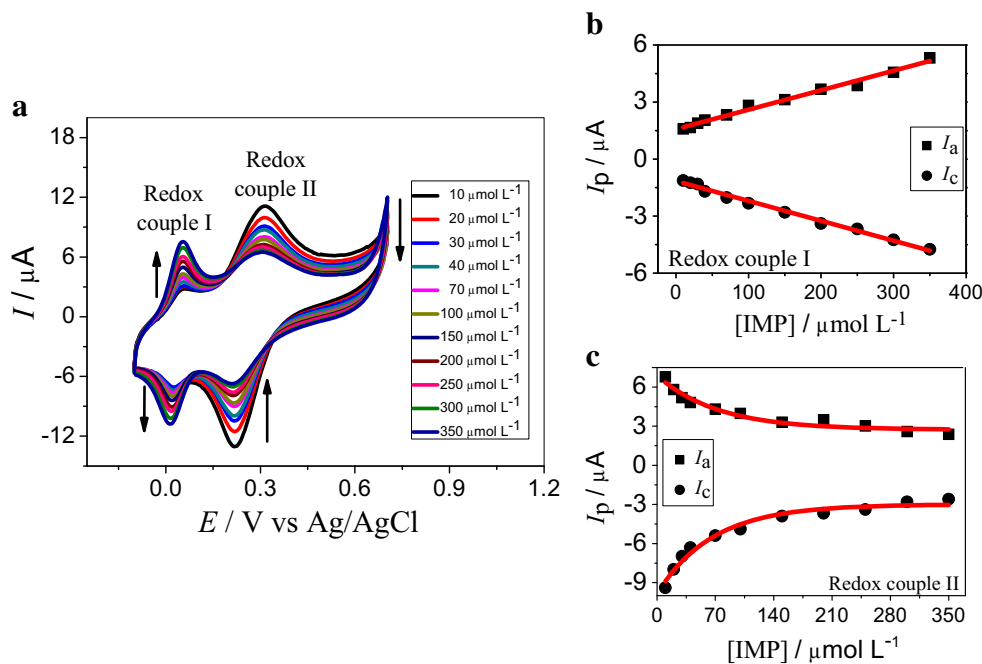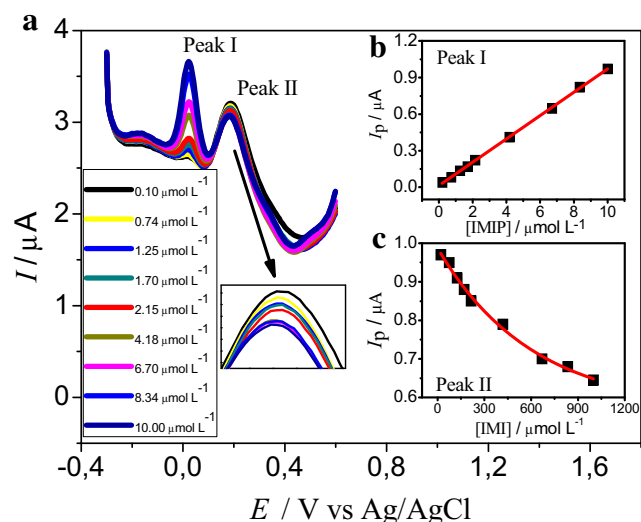
Fig. 7 a Differential pulse voltammograms for IMP oxidation in the concentration range of 0.1–10 μmol L^{-1}. **b** and **c** Plots of I_p versus [IMP] for the peaks I and II, respectively. Measurements performed under the optimized conditions. v = 0.05 V s^{-1} and pulse amplitude = 0.05 V

f-CNT/Nf modified electrode was able to provide good selectivity for detection of imipramine, without any interference from commonly coexisting substances.

Conclusions

A simple and efficient procedure was proposed for adsorption FCA-CD on f-CNT. This work shows that a GCE modified with FCA-CD on f-CNT is a feasible alternative for analytical determination of IMP in psychotropic drugs and urine samples. The results suggest that the presence of IMP in the electrolytic solution promote the release of FCA from the inclusion complex allowing the detection of this analyte at low potential due to the direct oxidation/reduction of FCA molecules on the carbon nanotube surface. Optimization of the experimental conditions resulted in a detection limit and sensitivity for IMP better than those described in the literature. This sensor showed good repeatability for both the measurements and electrode preparation, evaluated in terms of relative standard deviations. The proposed sensor was applied to determination of IMP in psychotropic drugs and the results were in agreement with nominal values. Moreover, the method showed a good percentage of recovery for the samples; therefore, it can be concluded that the FCA-CD/f-CNT/GCE is a sensitive, robust, and stable sensor for IMP determination in psychotropic drugs and urine samples.

prepared in the same conditions of IMP, 0.1 mol L^{-1} PBS, pH 7.0. The electrochemical results indicated that 100-fold higher concentrations of different species such as magnesium stearate, titanium dioxide, ascorbic acid, uric acid, dopamine, levodopa, Na$^+$, K$^+$, Ca^{2+}, Ni^{2+}, Zn^{2+}, and glucose did not interfere in the IMP determination. It was therefore apparent that by exploiting the oxidation of these species, the FCA-CD/

Table 1 Comparison of analytical parameters of the IMP oxidation on the modified electrode with FCA-CD/f-CNT/Nf

Electrode	Technique	E_{oxi}/V	Electrolyte/pH	Range linear (mol L^{-1})	LOD (mol L^{-1})
MIP/Au-NP/ITO electrode [24]	Differential pulse voltammetry	0.87 (vs SCE)	PBS/6.8	5.0 to 1000 × 10^{-6}	1.0 × 10^{-9}
PdNPs/GCE [25]	Electrochemicluminescence	0.75 (vs Ag/AgCl)	PBS/pH 7	1.0 to 100 × 10^{-12}	0.1 × 10^{-12}
Au-DNA [26]	Differential pulse voltammetry	0.75 (vs Ag/AgCl)	PBS/pH 7.5	5.0 to 50 × 10^{-9}	5 × 10^{-10}
FFT/Au [27]	Stripping cyclic voltammetry	0.8 (vs Ag/AgCl)	H$_3$PO$_4$/pH 2	50 × 10^{-12} to 8 × 10^{-8}	16.25 × 10^{-12}
NC-CILE [28]	Stripping voltammetry	0.65 (vs Ag/AgCl)	PBS/pH 7.2	0.1 to 2.0 × 10^{-6} 0.1	19 × 10^{-9}
β-CD-CPE [20]	Differential pulse voltammetry	0.8 (vs Ag/AgCl)	PBS/pH 6.6	0.1 to 1.0 × 10^{-6}	2.0 × 10^{-8}
BDD [29]	HPLC with electrochemical detection	0.85 (vs Ag/AgCl)	PBS/pH 6.9	0.05 to 100 × 10^{-6}	3 × 10^{-9}
GPU [30]	Square wave voltammetry	0.04 (vs Ag/AgCl)	BR buffer/pH 7.0	0.3 to 2.3 × 10^{-6}	2.0 × 10^{-8}
XAD2-TNP-GCPE [31]	Stripping differential pulse voltammetry	0.83 (vs Ag/AgCl)	PBS/pH 6.0	1.3 × 10^{-9} to 6.23 × 10^{-6}	3.93 × 10^{-10}
FCA-CD/f-CNT/Nf [This work]	Differential pulse voltammetry cyclic voltammetry	0.02 (vs Ag/AgCl) 0.04 (vs Ag/AgCl)	PBS/pH 7.0	0.1 to 10 × 10^{-6} 10 to 350 × 10^{-6}	1 × 10^{-9}

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