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# β-Sonogel-Carbon electrodes: A new alternative for the electrochemical determination of catecholamines

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ABSTRACT

In this work, a new alternative for the electrochemical determination of catecholamines based on β-cyclodextrin-Sonogel-Carbon electrodes is reported. The incorporation of β-CD and graphite in the preparation of the Sonogel-Carbon material leads to a modification of the electrode surface properties which causes a significant increase in the oxidation peak current of biomolecules such as dopamine, L-epinephrine, D,L-norepinephrine and catechol. This phenomenon might be attributed to the formation of an inclusion complex between β-CD and the catecholamines. The amount of β-CD necessary to form the Sonogel electrode was studied and optimization of electrochemical parameters, perm selectivity and mechanical stability of the sensor are discussed. Scanning electron microscopy and electrochemical impedance spectroscopy measurements were employed to characterize the electrical parameters and the structural properties of the new electrode surface, respectively. Cyclic voltammetry (CV) and Adsorptive differential pulse voltammetry (AdDPV) measurements were also used to explore the electrochemical behaviour of the electrode versus the quoted catecholamines. The β-CD-Sonogel-Carbon electrode offers fast and linear responses towards dopamine, norepinephrine, epinephrine and catechol, with good and low detection limits: 0.164, 0.294, 0.699 and 0.059 μmol $L^{-1}$, respectively.



## 1. Introduction

In literature, there are many papers reporting the manufacture process and analytical applications involving Sonogels. The Sonogel-Carbon materials were patented and described first by Hidalgo-Hidalgo de Cisneros et al. [1,2]. The fabrication procedure of Sonogel-Carbon materials is based on the use of sonocatalysis. Sonocatalysis consists of applying high-energy ultrasound directly to the precursors. In this way, ultrasonic cavitation is achieved, promoting hydrolysis with acidic water in the absence of any additional solvent. As a consequence of this, the time necessary to get a unique phase is reduced drastically, with regard to the classical sol–gel processes [3,4]. The mixture of Sonogel with spectroscopic grade graphite leads to the Sonogel-Carbon electrodes, which possesses especially favourable electrochemical properties.

Moreover, a good deal of modifiers has been included in this type of electrodes: polyethylenglycol (PEG), C-18, thioureas, hydrotalchyte, polythiophene [5–14], or has been used to recover their surface: glutaraldehyde, alumine, PEG, nafion, and several enzymes, such as tyrosinase, laccase, peroxydase and acetylcholinesterase [15–18], in order to test their electrochemical and structural behaviour. In this paper, we have agglutinated β-cyclodextrins into the matrix of the Sonogel-Carbon electrodes.

Cyclodextrins (CDs) are cyclic oligomers composed of six, seven or eight glucopyranose units (α, β or γ-cyclodextrins, respectively), linked by α(1–4) bonds [19]. They are widely studied in aqueous media as examples of host molecular receptors owing to their high affinity for hydrophobic molecules [20]. It is well known that CDs form inclusion complexes with a great variety of analytes having a diameter of 5–8 Å [21,22]. The ability to build inclusion complexes has been widely used in pharmaceuticals [23], as well as in analysis of organic and inorganic materials [24]. Current methods for the determination of these inclusion complexes include UV/vis spectrophotometry [25], IR spectroscopy [26], spectrofluorimetry [27], and electrochemical methods. Considerable attention has been paid to the electrochemical studies of inclusion complexes between cyclodextrins and electroactive species. Electrochemical techniques such as cyclic voltammetry [28], polarography [29,30],

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and pH determination [31], are particularly useful in the study of guest molecules lacking of a chromophore group for spectrophotometric measurement, but able to develop electroactivity.

Cyclodextrins, spread on an electrode surface, have proved to be effective and selective binding agents to form inclusion complexes with various compounds fulfilling structural requirements of the CDs cavity. This is important for electrochemical and electroanalytical applications [32].

As far as we know, the use of CDs to modify Sonogel-Carbon electrodes is a totally new challenge in the field of the chemically modified electrodes. The employment of β-cyclodextrin in electroanalysis is very frequent; particularly, it is used as modifier in carbon paste electrodes [33–36]. There is also a review about immobilization of cyclodextrins, their complexation abilities and analytical applications [32]. In this paper, we have carried out the incorporation of the monomeric form of β-CD into the electrode, instead of on its surface; in this way, no polymerization process was required.

In order to explore these possibilities further, in this paper we propose a new alternative, based on β-cyclodextrin-Sonogel-Carbon electrodes, for the electrochemical determination of several neurotransmitters belonging to catecholamines: catechol, dopamine (DA), norepinephrine (NE), and epinephrine (EN). The study was performed using two electroanalytical techniques: cyclic voltammetry (CV), and adsorptive differential pulse voltammetry (AdDPV). The objective was focused on the capability of the β-cyclodextrin-Sonogel-Carbon electrodes to detect better this neurotransmitters and thus to develop an original electroanalytical method for the determination of these substances. The optimization of all the analytical conditions are described and close attention is paid to the interferences due to ascorbic and uric acid, and some anions.

## 2. Experimental

### 2.1. Reagents and materials

Methyltrimethoxysilane (MTMOS) was from Merck (Darmstad, Germany) and hydrochloric acid was from Panreac (Barcelona, Spain).

β-Cyclodextrin (>99%) was obtained from Fluka (Switzerland). L-ascorbic acid (99%) was purchased from Sigma (Barcelona, Spain). L-dopamine, D,L-norepinephrine, epinephrine and catechol were all purchased from Aldrich (Milwaukee, USA) and used as received. Potassium phosphate dibasic-anhydrous and potassium phosphate monobasic used to obtain the phosphate buffer solution (PBS, 0.1 mol $l^{-1}$, about pH 7) were also purchased from Fluka. All reagents were of analytical grade or higher and used as received without further purification.

Graphite powder (spectroscopic grade RBW) was from SGL Carbon (Ringsdorff, Germany). Nanopure water was obtained by passing twice-distilled water through a Milli-Q system (18 MΩ cm, Millipore, Bedford, MA).

Glassy capillary tubes, i.d. 1.15 mm, were used as the bodies for the composite electrodes.

### 2.2. Instrumentation

The experimental work was realized in the Faculties of Sciences of Cadiz (Spain) and Tetouan (Morocco).

The electrochemical measurements were performed in two equipments, depending on the electrochemical technique employed. On one hand, the AdDPV and CV measurements were performed with an AutoLab PGSTAT20 (Ecochemie, Utrecht, The Netherlands) potentiostat/galvanostat interfaced with a personal computer, using the AutoLab software GPES for waveform generation, data acquisition and elaboration. The experiments were carried out in a single-compartment three-electrode cell, at room temperature (25 ± 1 °C). The counter electrode was a platinum wire, and a silver/silver chloride/3 M KCl electrode was used as the reference. The composite-filled glass capillary tubes were used as the working electrode.

On the other hand, the electrochemical impedance spectroscopy (EIS) measurements were performed with a Voltalab® 10, type PGZ 100, from Radiometer (Villeurbanne, France). The impedance spectra were recorded using the same three-electrode cell setup described above. The initial frequency used was 100 kHz and the final frequency was 10 mHz, with an AC amplitude of 10 mV. A potential of 0 V was chosen in order to insure the stability of the films on the electrodes during the experiments.

The synthesis of the Sonogel-Carbon materials was carried out sonicating with a high power ultrasonic generator, SONICATOR 3000, from MISONIX (MISONIX, Inc. Farmingdale, NY, USA) (equipped with a 13 mm titanium tip), that provides a maximum power of 600 W.

Scanning electron microscopy (SEM) studies were carried out on a QUANTA 200 (FEI Company, Hillsboro, Oregon, USA) operating at 20 keV and equipped with a microanalyzer (EDAX) to perform energy dispersive spectroscopy (EDS).

### 2.3. Electrode preparation procedure

To prepare the Sonogel-Carbon materials modified with β-cyclodextrin, the following steps were carried out. On one hand, a mixture of 500 μl of MTMOS and 100 μl of 0.2 M HCl was insonated for 5 s, obtaining the Sonosol. On the other hand, different amounts of β-cyclodextrin and graphite powder (until 1 g) were homogenized, in order to obtain different percentages of modification. Afterwards, this mixture was added and adequately dispersed into the Sonosol, resulting in the β-cyclodextrin-Sonosol-Carbon. After several minutes, the resulting material acquires enough consistency to filled the capillary tubes, giving place to the β-cyclodextrin-Sonogel-Carbon electrodes. 24 h later, the surface of the electrodes could be polished and a copper wire inserted as the electrical contact into the electrodes, being ready to be used.

The electrochemical measurements were carried out using a phosphate buffer solution as the supporting electrolyte. The optimal instrumental parameters for AdDPV scans were as follows: potential range from +0.1 to +0.8 V; accumulation potential: 0.4 V; accumulation time: 120 s; modulation time: 0.06 s; interval time: 0.6 s; scan rate: 10 mV $s^{-1}$; pulse amplitude: 100 mV. The optimal instrumental parameters for CV sweeps were as follows: range from 0 to +1 V; scan rate: 50 mV $s^{-1}$. Measurements were carried out under $N_2$ atmosphere when required.

## 3. Results and discussion

The preparation mode of the Sonogel Carbon electrodes is different from the one usually used with other solid electrodes and similar to the one used in carbon paste electrodes.

Comparing with carbon paste electrodes, the preparation of Sonogel-Carbon electrodes, although easy and fast, is a bit less easy. Nevertheless Sonogel-Carbon electrodes have several advantages versus carbon paste electrodes: the robustness against the presence of organic solvents is bigger; the amounts of reagents required to prepare an electrode are lesser; the regeneration of the surface is easy and allows that the same Sonogel-Carbon electrode is utilized for many times (a longer life time), with respect to carbon paste electrodes that are usually used only once. These advantages are complemented with their good reproducibility.

### 3.1. Influence of β-cyclodextrin in the responses of the Sonogel-Carbon electrodes

Different percentages of β-cyclodextrin (β-CD) were added into the Sonosol matrix in order to study the influence of the modifier proportion in the β-CD-Sonogel-Carbon electrodes. The modification percentages tested were: 2.5%, 5% and 7%, w/w β-cyclodextrin:graphite. The AdDPV responses of the modified electrodes were compared with an unmodified Sonogel-Carbon electrode. The analyte employed was cathecol ($5 \times 10^{-4}$ M in the electrochemical cell).

The results obtained showed that the unmodified Sonogel-Carbon electrode had a poor and low response with respect to the modified electrodes. The presence of β-CD in the composite caused a spectacular increase in the response. The maximum response was for 5% of β-CD modification. For higher proportions, the current peak values were lower, the signal loosing resolution.

Furthermore, the increase up to 7% β-CD presented gelification troubles, affecting the structure of the basis material and, subsequently, the mechanical and electrochemical behaviour. This circumstance could explain the loss of signal resolution. The gelification troubles depend on the nature of the modifier and its modification percentage included in the graphite powder, as it has previously been reported [11].

From the previous discussion, the 5% modification of β-CD into the Sonogel-Carbon electrodes was kept as optimal for the subsequent experiments.

### 3.2. Influence of the supporting electrolyte

Several types of 0.1 M supporting electrolytes were also tested: phosphate buffer solution (PBS), $H_2SO_4$, KCl and Britton–Robinson, at different pH values depending on the chemical nature of the reagents used. The AdDPV responses of the modified electrodes versus cathecol ($5 \times 10^{-4}$ M) were compared using the different buffers. The results obtained were very similar in all cases; nevertheless, the PBS was chosen due to the following reasons: (1) it is very easy to manipulate their components and to prepare it, since the initial pH value of the solution is always approximately 7; (2) this pH value is the most appropriate to analyze biological samples as neurotransmitters, so there is no need to modify the pH of the buffer; (3) the use of buffers at very acid or basic pH values may deteriorate the analytes or the composite electrodes.

### 3.3. Influence of pulse amplitude, accumulation time and accumulation potential

Different values of pulse amplitude, accumulation time and accumulation potential were tested in order to determine their influence on the peak intensity values when analyzing the neurotransmitters: L-dopamine (DA), D,L-norepinephrine (NE), epinephrine (EN) and catechol (CA), $5 \times 10^{-4}$ M in all cases. The electrochemical technique employed was AdDPV.

The effect of varying pulse amplitude and its influence on the peak height was studied. This electrochemical parameter ranged from 75 to 200 mV, the maximum having been found at 100 mV.

On one hand, the accumulation time ranged from 40 to 400 s. The peak intensity increased with the accumulation time, as Fig. 1 shows. However, the greatest increment occurs up to 80 s, after which a very slight increase is observed. For future analyses, 120 s were considered the optimum value of accumulation time, since saturation was observed for higher values.

On the other hand, the accumulation potential ranged from 0 V to 500 mV. The maximum peak intensity value was found at 400 mV.

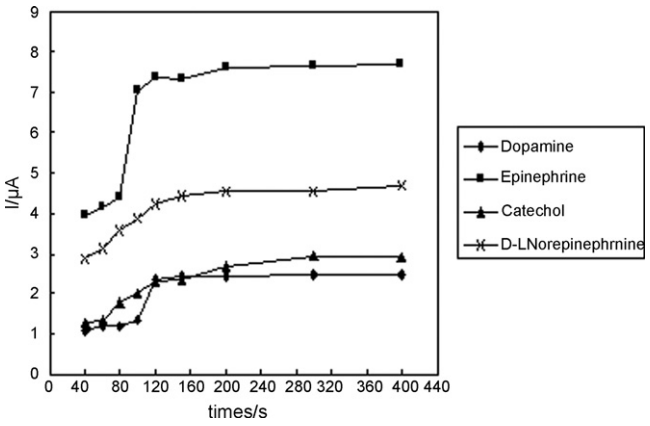


**Fig. 1.** Effect of the accumulation time on the peak height for $5 \times 10^{-4}$ M of epinephrine, dopamine, D,L-norepinephrine and catechol in PBS buffer at pH 7.4 using a 5% β-CD-Sonogel-Carbon electrode.

Consequently, for successive experiments the optimal values reported here were utilized.

### 3.4. Studies of repeatability, reproducibility and stability

The repeatability was established carrying out six consecutive determinations of the different catecholamines in order to determine the peak intensity values in AdDPV measurements. The electrochemical parameters used were the optimized ones previously. The concentration of the analytes was $4 \times 10^{-5}$ M. The relative standard deviations for DA, EN and NE were 1.8%, 0.9% and 3.9%, respectively. These results showed that the 5% β-CD-Sonogel-Carbon electrode has good repeatability.

In order to study both the reproducibility and the stability, a 5% β-CD-Sonogel-Carbon electrode was used, measuring daily $4 \times 10^{-5}$ M solutions of DA. After the measurements, the electrode was stored at the lab environment. A relative standard deviation of 2.7% was obtained after 30 days of consecutive measurements.

### 3.5. Study of the electrochemical behaviour of neurotransmitters by cyclic voltammetry

Fig. 2 shows the cyclic voltammograms of 5 mM DA, EN and NE, measured at an unmodified Sonogel-Carbon electrode (a) and at a 5% β-CD-Sonogel-Carbon electrode (b). As it can be seen there are clear differences between the responses obtained from the two types of electrodes. No peak is observed with the unmodified electrode; nevertheless, the presence of the β-cyclodextrin in the structure of the electrodes causes the appearance of the redox activity of the neurotransmitters tested.

The results reported here for DA, EN and NE are better than those obtained by some of us using other type of electrodes [37]. The electrochemical peak parameters are summarized in Table 1. From these data, it can be concluded that, under these conditions, the 5% β-CD-Sonogel-Carbon electrode shows the following electrochemical kinetics: reversible or quasi-reversible behaviours with $\Delta E_p$ of 346, 374, and 432 mV for DA, NE and EN, respectively.

Excepting for the borderline case of NE, the currents ratio of the oxidation and the reduction peaks is less than 1; this means that these compounds do not undergo complex reactions and they are stable at the modified electrode.

### 3.6. Influence of scan rate on the electrochemical responses

The scan rate has great influence on the peak current of cyclic voltammograms. In this paper, the effect of varying the scan rate for

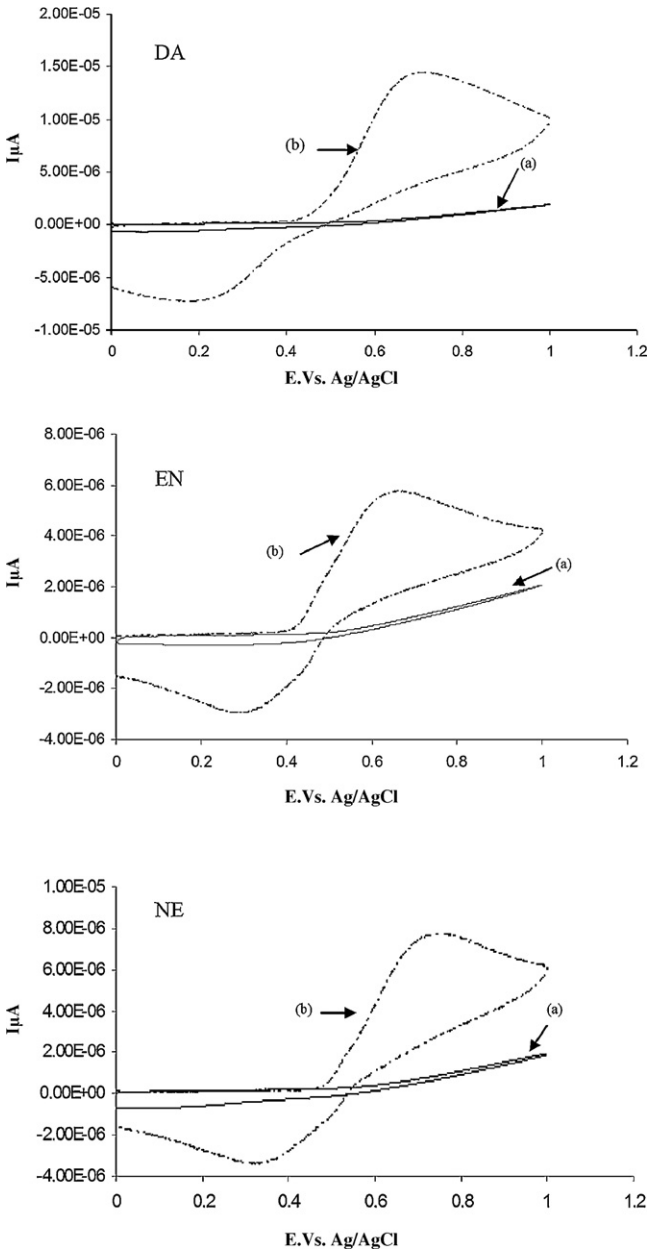


**Fig. 2.** Cyclic voltammograms for 5 mM of D,L-norepinephrine (1), epinephrine (2), and dopamine (3) at: (a) an unmodified Sonogel-Carbon electrode, and (b) a 5% β-CD-Sonogel-Carbon electrode; scan rate = 50 mV $s^{-1}$; PBS buffer at pH 7.4.

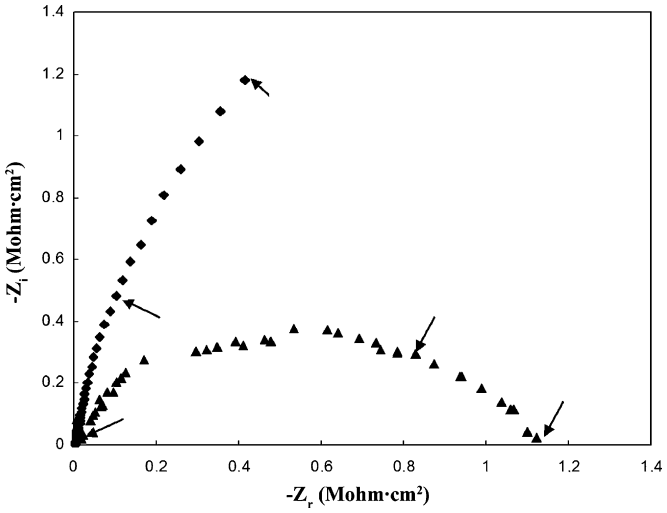


**Fig. 3.** Nyquist plots for impedance data obtained at 0 V for unmodified Sonogel-Carbon electrodes (▲) and 5% β-CD-Sonogel-Carbon electrodes (♦). Frequency range: 100 kHz–10 mHz.

the determination of the neurotransmitters selected was studied in the optimized conditions described above. The anodic peak currents $I_{pa}$ obtained with a 5% β-CD-Sonogel-Carbon electrode had a linear dependence with respect to the square root of the scan rate ($v^{1/2}$) in the range from 5 to 200 mV $s^{-1}$. The correlation coefficients ($R^2$) were 0.9997, 0.9997, and 0.9975 for DA, NE, and EN, respectively. The linear regression equations obtained were $I_{pa}(DA) = 0.3764 + 0.4121 \times v^{1/2}$; $I_{pa}(NE) = -0.7293 + 0.3873 \times v^{1/2}$; and $I_{pa}(EN) = -0.0724 + 0.1211 \times v^{1/2}$, where the units are $I_{pa}$(μA) and $v^{1/2}$ (mV $s^{-1}$)$^{1/2}$. Our results indicate that the electron transfer reaction is diffusion controlled in the case of the neurotransmitters studied.

### 3.7. Electrochemical impedance spectroscopy (EIS) studies

Electrochemical impedance spectroscopy (EIS) is an effective method to explore the interfacial properties of modified electrodes [38]. It is based on the perturbation of a system at equilibrium by a small AC potential amplitude wave from 5 to 10 mV. The interaction of an analyte with the electrode surface is indicated by a shift in the impedance or a change in capacitance of the bulk electrode [39–41]. In our case, the impedance experiments were performed in order to characterize the electrode/electrolyte interface. Fig. 3 shows EIS spectra of an unmodified Sonogel-Carbon electrode (▲) and a 5% β-CD-Sonogel-Carbon electrode (♦) in 0.1 M $H_2SO_4$, recorded at 0 V versus a saturated calomel electrode (SCE). The frequency range and the AC potential amplitude have been previously described in the Section 2.

**Table 1**
Electrochemical peak parameters obtained from the cyclic voltammograms for dopamine, epinephrine, and D,L-norepinephrine. Scan rate: 50 mV $s^{-1}$.

| | Dopamine | Epinephrine | D,L-Norepinephrine |
|---|---|---|---|
| $E_{pa}$ (V)[a] | 0.674 | 0.755 | 0.663 |
| $E_{pc}$ (V)[a] | 0.328 | 0.323 | 0.289 |
| $I_{pa}$ (A)[b] | $1.060 \times 10^{-06}$ | $1.502 \times 10^{-06}$ | $1.511 \times 10^{-06}$ |
| $I_{pc}$ (A)[b] | $-1.276 \times 10^{-06}$ | $-1.871 \times 10^{-06}$ | $-1.463 \times 10^{-06}$ |
| $\Delta E$ (V)[c] | 0.346 | 0.432 | 0.374 |
| $I_{pa}/I_{pc}$ (A) | 0.832 | 0.804 | 1.034 |
| $E_p - E_p/2$(V, oxidation) | 0.074 | 0.077 | 0.076 |
| $E_p - E_p/2$(V, reduction) | −0.087 | −0.099 | −0.091 |

[a] $E_{pa}$ and $E_{pc}$ are the oxidation and reduction potential peaks.
[b] $I_{pa}$ and $I_{pc}$ are current intensity of oxidation and reduction peaks.
[c] $\Delta E$(V): is the difference between oxidation and reduction potential (or the peak separations for the molecules at 5% β-CD-Sonogel-Carbon electrode). Scan rate = 50 mV $s^{-1}$; buffer = PBS at pH 7; concentration of the analytes = 5 mM.

**Table 2**
Electrical parameters calculated from the impedance spectra in 0.1 M of $H_2SO_4$ for an unmodified-Sonogel-Carbon electrode and a 5% β-CD-Sonogel-Carbon electrode.

| | $R_e$ ($\Omega$ cm$^2$) | $R_{ct}$ (M$\Omega$ cm) | $C_{dl}$ (nF cm$^2$) |
|---|---|---|---|
| Sonogel-Carbon | 4.909 | 5.537 | 177 |
| 5% β-CD-Sonogel-Carbon | 5.012 | 1.006 | 287.3 |

$R_e$ = electrolyte resistance; $R_{ct}$ = charge transfer resistance; $C_{dl}$ the double layer capacitance at the electrode/electrolyte interface.

The EIS spectra are characterized by a perfect semicircle corresponding to the charge transfer process at the electrode surface with a correlation of 0.999 for the two complexes plot. The presence of cyclodextrin in the Sonogel-Carbon electrode seems to influence the interfacial impedance values. Electrical parameters were calculated using a Voltamaster® 4.0 software. Fitting results are presented in Table 2. $R_e$ is the electrolyte resistance, $R_{ct}$ is the charge transfer resistance, and $C_{dl}$ is the double layer capacitance at the electrode/electrolyte interface.

On one hand and from these data, we can notice a decrease in the charge transfer resistance value for the 5% β-CD-Sonogel-Carbon system with respect to the unmodified Sonogel-Carbon system. On the other hand, the higher value calculated for the electrical double layer, $C_{dl}$, was observed at the 5% β-CD-Sonogel-Carbon/electrolyte interface. This result could be probably attributed to an increase in the electrode surface area [42]. This change in the capacitance suggested that cyclodextrin was successfully incorporated into the Sonogel-Carbon electrode. The reproducibility of the measurements from several 5% β-CD-Sonogel-Carbon electrodes was good and this could be explained by an exhaustive control of the experimental conditions of the electrodes preparation.

### 3.8. Interference studies

In order to explore possible analytical applications of the sensors described in this paper, the effect of ascorbic acid (AA), which causes severe interference, and several inorganic ions, was studied during the determination of the neurotransmitters selected.

In the literature, it has been already reported the catalytic effect of DA on the AA oxidation at the surface of different types of electrodes [43,44]. In these papers, the strong AA interference on the electrochemical determination of DA and other catecholamines is confirmed.

With the aim of investigating this issue, we have carried out a careful study of this catalytic effect employing a 5% β-CD-Sonogel-Carbon electrode in solutions containing DA (analyte of reference) and AA; AdDPV was the electrochemical technique employed. On one hand, Fig. 4a shows that the peak height of DA remained constant when the concentration of AA in the mixture was varied from 0 to $8 \times 10^{-4}$ M, while peak height of ascorbic acid increased linearly with concentration. On the other hand, when varying the concentration of DA from 0 to $2 \times 10^{-7}$ M instead of AA, the peak height of ascorbic acid remained constant (Fig. 4b). These results demonstrate that there is no homogeneous catalytic coupling between AA and DA at the 5% β-CD-Sonogel-Carbon electrode. Moreover, although the peaks of ascorbic acid and dopamine are partially overlapped, it seems that the modified electrode has the ability to resolve the system allowing the determination of DA even in the presence of high amounts of ascorbic acid (250-fold higher).

To achieve this purpose, we have adopted as a criterion to evaluate the peak height of DA taking the distance between the maximum of the peak and the tangent to the baseline of the DA peak with no interferents [45]. The linear relation between these heights and the concentration of DA can be verified with the following equation and correlation coefficient: $I_p$ (nA) = 0.0264 × $C_{DA}$ (μM) + 0.1318; $r^2$ = 0.9954.

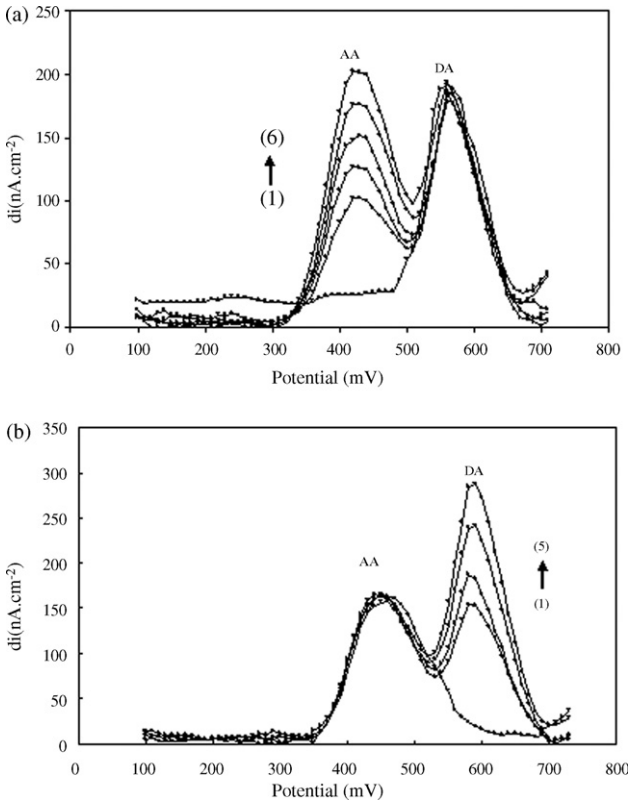


**Fig. 4.** AdDPV voltammograms corresponding to the interference studies when dopamine (DA) is determined in the presence of ascorbic acid (AA) at a 5% β-CD-Sonogel-Carbon electrode. (a) Concentration of DA is constant ($2 \times 10^{-7}$ M), and concentration of AA is varied: (1) 0 M; (2) $3 \times 10^{-4}$ M; (3) $4 \times 10^{-4}$ M; (4) $5 \times 10^{-4}$ M; (5) $6 \times 10^{-4}$ M; (6) $8 \times 10^{-4}$ M. (b) Concentration of AA is constant ($10^{-4}$ M), and concentration of DA is varied: (1) 0 M; (2) $1 \times 10^{-7}$ M; (3) $2 \times 10^{-7}$ M; (4) $4 \times 10^{-7}$ M; (5) $6 \times 10^{-7}$ M.

Our modified electrode is also able to allow the selective determinations of other catecholamines in the presence of ascorbic acid and in the presence of uric acid as well, which has a similar behaviour to AA.

With respect to the interference studies using inorganic ions, the concentration of all the neurotransmitters chosen was kept at $1 \times 10^{-5}$ M. After the measurements, no significant interference was obtained from the following species: $K_2SO_4$ (150), KCl (400), KBr (400), where the values in parentheses are the interferent/analyte concentration ratios.

### 3.9. Analytical applications to the determination of neurotransmitters

The quantitative determination of dopamine (DA), norepinephrine (NE) and epinephrine (EN) at the 5% β-CD-Sonogel-Carbon electrode was performed using adsorptive differential pulse voltammetry. In our case, the oxidation peaks, obtained by this voltammetric technique, for these neurotransmitters, were taken as the analytical signal.

Under the optimal experimental conditions obtained above, linear relationships between the concentrations and peak currents for DA, EN, and NE were obtained. The linear regression equations were: DA, $I$ (μA) = −0.0438 + 0.276 × C (μM); EN, $I$ (μA) = 0.2279 + 0.0119 × C (μM); NE, $I$ (μA) = 0.0039 + 0.002 × C (μM). The $r^2$ correlation coefficients were 0.9939, 0.9989 and 0.9992, respectively. The detection limit was considered as the concentration whose intensity value equals the blank intensity plus three times the standard deviation of the blank; for the determi-

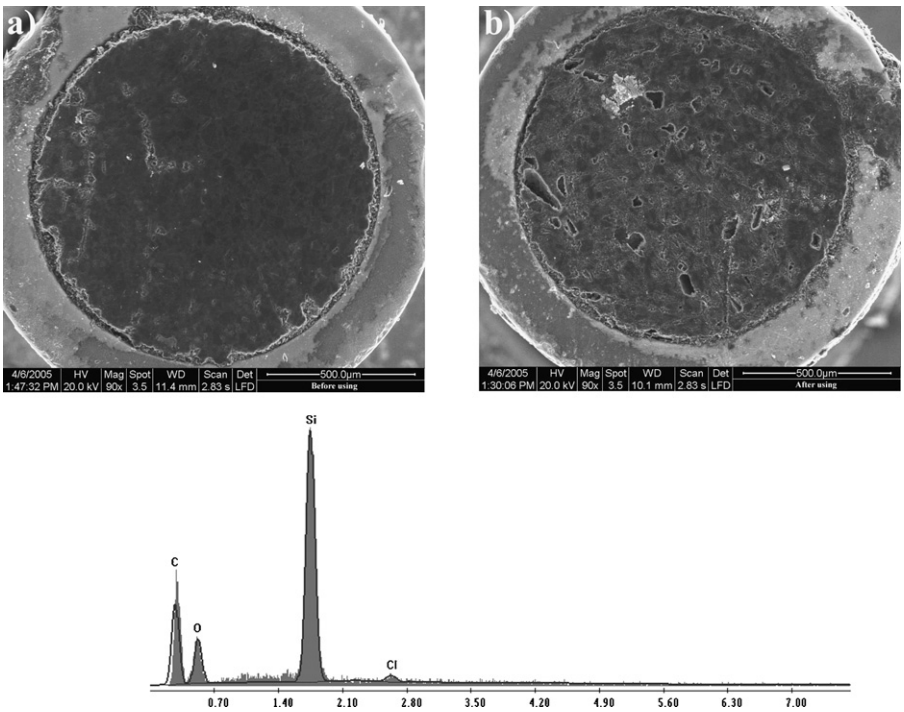


**Fig. 5.** Micrographs corresponding to the 5% β-CD-Sonogel-Carbon electrode polished: (a) before using, and (b) after using. The EDS spectrum corresponding to both micrographs is also included.

nation limit 10 times the standard deviation of the blank was used [46]. The detection limits were 0.164 μM for DA, 0.294 μM for NE, and 0.699 μM for EN. The latest LOD (Epinephrine) was much lower than the one obtained recently using a conducting polymer glassy carbon modified electrode [37]. These LODs are good enough to be exploited for the detection of these neurotransmitters in pharmaceutical products and/or biological fluids. The determination limits were 0.546 μM for DA, 0.980 μM for NE and 2.328 μM for EN.

The determination of catechol was also studied. A linear relationship between the anodic peak current and the concentration of catechol, ranging from $2 \times 10^{-7}$ to $2.2 \times 10^{-5}$ M, was obtained with a linear regression equation $I\,(\mu A) = 0.1467 + 0.2356 \times C\,(\mu M)$, and $r^2 = 0.9992$. The LOD: 0.059 μM, was lower than the value obtained in the literature with a polymer glassy carbon electrode [37]. The determination limit for this compound was 0.197 μM.

### 3.10. Scanning electron microscopy (SEM) studies

Six different samples were studied: 2.5% β-CD-Sonogel-Carbon electrode polished and not used, 2.5% β-CD-Sonogel-Carbon electrode polished and used, 5% β-CD-Sonogel-Carbon electrode polished and not used, 5% β-CD-Sonogel-Carbon electrode polished and used, 7% β-CD-Sonogel-Carbon electrode polished and not used, 7% β-CD-Sonogel-Carbon electrode polished and used. For each sample, the SEM and EDS studies were performed on the same equipment and at the same time.

Since SEM studies were carried out at low vacuum, it was not necessary a previous step of coating the samples with gold. The micrographs were always taken at 20 kV.

In all used electrodes, it can be observed the presence of certain type of erosion, in the form of a marked widening of the separation material/capillary tubes, detected in their surface. The fact of using an electrode on measurements in aqueous solution increases highly the quoted separation, as well as the appearance of holes and fissures in their surface. When comparing the erosion suffered by the three different electrodes, it can be concluded that 2.5% β-CD modified samples had a greater separation material/capillary tubes due to erosion, while the other two types of materials: 5% and 7% β-CD modified samples, present a minor separation. The greatest quantity of fissures and holes was presented by 2.5% β-CD modified samples, although electrodes with 7% of modification had quite a lot of them as well.

This fact could be used to corroborate the worst electrochemical results obtained with 2.5% and 7% β-CD modified materials, and the better electrochemical results for 5% β-CD modified material. This material presented smaller erosion and less number of fissures and holes, as well as better mechanical behaviour.

As an example to illustrate the surface of the materials studied, Fig. 5 shows two micrographs corresponding to a 5% β-CD-Sonogel-Carbon electrode polished: (a) before using and (b) after using. With regard to the data obtained by means of EDS, Fig. 5 also gives the composition for the materials studied. Furthermore, from all the spectra collected, it can be concluded that no differences were observed in the composition of the materials that denote the influence of the quantity of β-cyclodextrin.

## 4. Conclusions

In this paper, a new alternative for the electrochemical determination of catecholamines has been reported. Our choice is based on the use of β-cyclodextrin-Sonogel-Carbon electrodes. Besides, this is the first time that this electroanalytical performance has been carried out with this type of formulation: β-cyclodextrin included in Sonogel-Carbon matrices.

The modification percentage of β-cyclodextrin, the supporting electrolyte and the pH, as well as the electrochemical parameters were optimized in order to obtain the best responses for the electrodes. The results showed that β-cyclodextrin incorporated in Sonogel-Carbon electrodes gives better responses towards catecholamine molecules than unmodified Sonogel-Carbon electrodes. This is probably due to the formation of inclusion complexes between catecholamines and the β-CD. When comparing the cyclic

voltammetry responses with those obtained at modified glassy carbon electrodes, the results were also improved. This type of electrodes also showed excellent reproducibility, repeatability and stability. The good electroanalytical and structural properties of this type of electrodes have been corroborated by EIS and SEM studies.

Furthermore, the negligible effect of ascorbic and uric acids on the measurements together with the very encouraging LODs obtained for the different cathecolamines make the new electrodes very stable and allow easy operation life time during more than 4 weeks. This life time of the electrode can be explained too by a weak leaching of the cyclodextrin in the solution. Our preparation method is sensitive enough to be applied to single tablet assay which may open new horizons for their use in the medical and biochemical analysis field.

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