

A fast and simple ozone-mediated method towards highly activated screen printed carbon electrodes as versatile electroanalytical tools

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Abstract

A facile and effective electrochemical activation method of screen printed carbon electrodes (SPCEs) has been performed using ozone gas. Activated SPCEs showed relevant improvements in the electrochemical properties such as an impedance reduction and better electroanalytical outcomes. Such improved properties were attributed to the increase of the electroactive surface area and the functionalization of the electrode surface with carbon-oxygen groups onto the carbonaceous ink surface. The optimized activation method consisted in the performance of a voltammetric cycle between -2 and 2 V at 10 mVs⁻¹ in 0.1 M NaOH solution with constant ozone gas bubbling. This activation procedure takes 12 min, which allows its use routinely prior to the electrode modifications and electroanalytical measurements. The resulting activated SPCEs exhibited superior sensitivities towards hydrogen peroxide, acetaminophen, hydroquinone and dopamine. This methodology might be considered as a strategy to attain SPCEs with improved electroanalytical properties for multiple applications.

KEYWORDS: Activation, hydrogen peroxide, ozone, phenolic compounds, screen-printed carbon electrodes.

1. Introduction

Ozone, a triatomic form of oxygen, has been used for years in applications such as treatment of waters ^[1], food disinfection ^[2] and agriculture ^[3]. Since ozone survives for only a matter of minutes before decomposing into oxygen, it is considered as an environmentally friendly disinfectant ^[4]. This oxidant molecule is generally produced on-site via the generation of an electrical discharge from molecular oxygen ^[5]. Ozone has also been used for producing activated carbon surfaces with enhanced electron-transfer properties. In this sense, Zhou et al. (2001) published an effective UV/ozone pretreatment able to produce clean and active glassy carbon surfaces ^[6]. The same authors correlated this high activity directly to an increase in oxygenated functionalization groups on the surface of the electrode. Similarly, flexible carbon nanotube electrodes were exposed to UV/ozone for neuronal recording ^[7]. This electrode surface treatment provided a low-cost route of carbon nanotubes functionalization by introducing oxygen-carbon moieties on the surface of electrodes, enhancing their electrochemical properties.

Screen printed electrodes (SPEs) are commercial devices made of conductive inks based on some metals (usually silver, platinum, gold) or carbon. Among them, screen printed carbon electrodes (SPCEs) are the most widely used due to their versatility and cheapness ^[8]. Conductive carbon inks have been employed for many years in screen printing technology for a variety of sensor/biosensor applications ^[9]. In the production of these devices, mineral binders, insulating polymers and other additives are added to electroactive species to improve the characteristics of the ink onto the ceramic or polymeric substrates ^[10]. All these non-electroactive additives are detrimental in electrochemistry providing slow electrode electrochemical kinetics ^[11, 12]. For this reason, pretreatments of electrodes are mandatorily required in many cases to enhance their electro-transfer properties ^[8, 13]. Over the years, researchers have developed methods to activate carbon based electrodes surface including electrochemical pretreatments ^[11, 12], mechanical activation ^[14], heat treatment ^[15] and oxygen plasma treatment ^[16]. Changes in the carbon surface chemistry, such as the introduction of carbon-containing functional groups ^[6] or the introduction of edge plane sites or defects ^[17] exhibit significant impact on various electrocatalytic reactions. Very recently, the improvement of the electrochemical performance of SPCEs by UV/ozone modification ^[18] and the renewal mechanism of UV/ozone cleaning process on contaminated SPCEs have been reported by Wang et al. ^[19]. Even though the non-electrochemical UV/ozone pretreatment used by

Wang's works takes 5 min, the chemical cleaning only provides slight improvements in the analytical signals of nicotinamide adenine dinucleotide and $K_3[Fe(CN)_6]$, as well as the reusability of contaminated SPCEs for further usage.

Our group has reported recently an alternative method based on the electrochemical activation of SPCEs using hydrogen peroxide ^[20] and sulfuric acid/hydrogen peroxide ^[21] solutions through the use of cyclic voltammetry (CV) in a certain potential range. In doing those pretreatments, the introduction of oxygenated groups on the carbon working electrode improved remarkably the electro-transfer properties and enhanced sensitivity to hydrogen peroxide and phenolic compounds. Even though these activation protocols are highly effective, they are time-consuming procedures, thereby limiting their use in large-scale pretreatments.

The present work evaluates for the first time the electrochemical activation of SPCEs based on CV combined with the presence of ozone gas in aqueous solution. According to the optimization of the experimental conditions in terms of number of voltammetric cycles and scan rate, electrode potential window and pH solution, the best electroanalytical outcomes for the electrooxidation of H_2O_2 and some phenolic compounds (namely acetaminophen, dopamine and hydroquinone) resulted from the performance of one CV in 0.1 M NaOH, under continuously ozone bubbling through the electrochemical cell. The combination of the oxidizing power of ozone with the application of high cathodic and anodic electrode potentials exerts a synergistic effect on the SPCEs activation. This improvement in the electrochemical properties of the activated electrodes (aSPCEs) is evident in the electrooxidation of H_2O_2 and certain important electroactive phenolic compounds.

2. Experimental section

2.1. Chemical and solutions

Acetaminophen (APAP), dopamine (DP), hydrogen peroxide (35%) hydroquinone (HQ), potassium chloride, sodium phosphate (monobasic and dibasic) and sodium ferrocyanide decahydrate were purchased from Sigma-Aldrich (Madrid, Spain). Sodium hydroxide was purchased from Merck (Madrid, Spain) and sulphuric acid (98%) from Panreac (Barcelona, Spain). All reagents were purchased at their highest available purity and used without further purification. Solutions were prepared with deionized water (resistivity $\approx 18.2 \text{ M}\Omega\cdot\text{cm}$ at 25°C) (Millipore, Watford, UK) and were freshly prepared every day.

An ozone generator (OzoGas, Barcelona, Spain, <http://www.ozogas.com/>) was used to continuously bubble ozone gas with a flow rate of 2 g/h into the solutions during the activation pretreatment.

2.2. Electrochemical measurements

Electrochemical experiments were performed using a potentiostat μStat 300 from Dropsens (Asturias, Spain, <http://www.dropsens.com/>). The electrodes used were also from Dropsens: SPCEs (DRP-110), which a carbon working electrode; graphene (GPH) modified SPCEs (GPH-SPCEs, DRP-110GPH), with a GPH/carbon working electrode; and carboxyl functionalized multi-walled carbon nanotubes (MWCNT-COOH) modified SPCEs (CNT-SPCEs, DRP-110CNT), with a MWCNT-COOH/carbon working electrode. All these electrodes contain a working electrode with 4 mm of diameter, a carbon counter electrode and a pseudo-silver reference electrode.

The voltammetric activation method was optimized by changing the experimental activation conditions such as number of cycles, scan rate, potential window and pH. The optimized protocol was 1 CV between -2 and +2 V in 0.1 M NaOH at 10 mVs^{-1} with constant ozone gas bubbling of 2 g/h-always performed in a fume hood. After activation, the activated electrodes (aSPCEs) were rinsed with deionized water and dried in air.

The electrooxidation of H_2O_2 was used to check the effectiveness of the activation process. Electroanalytical measurements were taken in sodium phosphate buffer (PB, 0.1 M, pH 7) by chronoamperometry by successive additions of micromolar amounts of H_2O_2 under constant stirring at 0.7 V at 25°C . APAP, DP and HQ were analyzed by measuring their oxidative current intensity peaks by lineal sweep voltammetry and CV in the same

buffer at 50 mVs^{-1} . The value of pH 7 was chosen for phenolic compounds assays for physiological reasons.

Electroactive areas of the non-activated electrodes (nSPCEs) and aSPCEs were calculated by cyclic voltammetry at different scan rates in the presence of $1 \text{ mM Na}_4\text{Fe}(\text{CN})_6$ plus 0.1 M KCl , previously deoxygenated with $\text{N}_2(\text{g})$. The Randles-Sevcik equation was used for this purpose ^[22]. The $\text{Na}_4\text{Fe}(\text{CN})_6$ diffusion coefficient used was $6.5 \times 10^{-6} \text{ cm}^2\text{s}^{-1}$ at 25°C ^[23].

Electrochemical impedance spectroscopy (EIS) measurements were performed in an AUTOLAB PGSTAT128N potentiostat with EIS analyzer (Eco Chemie B.V., The Netherlands) using NOVA 2.0 software. EIS were recorded at 0.12 V (vs. Ag) in $1 \text{ mM Na}_4\text{Fe}(\text{CN})_6$ plus 0.1 M KCl . Experimental conditions: stabilization time 60 s , amplitude 5 mV rms , frequencies 65 kHz - 10 mHz , with five points per decade.

2.3. Physico-chemical measurements

Scanning electron microscopy (SEM, HITACHI S-3000N microscope), working at 30 kV with X-ray detector Bruker Xflash 3001 for microanalysis, was employed for the analysis of the morphology of the electrodes.

XPS experiments were recorded on a K-Alpha Thermo Scientific spectrometer using $\text{Al-K}\alpha$ (1486.6 eV) radiation, monochromatized by a twin crystal monochromator, yielding a focused X-ray spot with a diameter of $400 \text{ }\mu\text{m}$ mean radius. The alpha hemispherical analyzer was used as electron energy analyzer, operating in fixed analyzer transmission mode, with survey scan pass energy of 200 eV and 40 eV narrow scans. The angle between X-ray source and the analyzer (magic angle) was 54.7° . Processing of the XPS spectra was performed in Avantage software, with energy values referenced to the C1s peak of adventitious carbon located at 284.6 eV , and a Shirley-type background.

3. Results and Discussion

3.1. Optimization of activation method

In this paper, we propose for the first time the simultaneous combination of the voltammetric activation method and the use of ozone gas in aqueous solution as a strong oxidizing agent to improve the electrochemical properties of SPCEs. The optimization of the pretreatment conditions was firstly carried out according to the sensitivities reached at the aSPCEs towards the electrooxidation of H_2O_2 at 0.7 V (*vs* Ag) (Supporting Information, Figure 1S). It was demonstrated that an increase in the number of cycles (Figure 1SA) or a decrease in the scan rate (Figure 1SB) favoured the activation of the electrodes, and therefore, the electroanalytical outcome towards H_2O_2 . Furthermore, the change of the electrode potential limits applied also produced an important effect on the electroanalytical response: the higher electrode potential window the larger increase in activation, as shown in Figure 1SC. Moreover, the excursion through positive electrode potentials yielded better activation of the electrode than that performed to negative ones.

In addition, similar electrochemical sensitivities towards H_2O_2 were seen when comparing aSPCEs pre-treated either by performing one voltammetric cycle between -2 and +2 V or 25 voltammetric cycles between -1 and +1 V (Figure 1SD). The increase of the electrode potential window considerably reduced the activation time, thereby achieving highly activated SPCEs in only 12 minutes at this scan rate (10 mV s^{-1}). This is of relevance since this activation time is much lower than that previously reported by us ^[20] preparing aSPCEs, which provided a reduction time of 92 % with sensitivities to H_2O_2 electrooxidation in the same order of magnitude. A higher potential window (-3 V to 3 V, Figure 1SB) and a lower scan rate (5 mV s^{-1} , Figure 1SD) yielded higher sensitivities, although with a considerable increment in the time of activation.

Therefore, the performance of 1 cyclovoltammogram between -2 and +2 V in 0.1 M PB at 10 mVs^{-1} with continuous ozone gas bubbling was chosen for the generation of activated electrodes (aSPCEs-PB). In this regard, the chronoamperometric profiles with respect to H_2O_2 electrooxidation using both the nSPCE and aSPCEs are compared in Figure 1. The nSPCE exhibited a very low signal to H_2O_2 additions with a sensitivity of $3.2 \pm 0.05 \text{ nA}\mu\text{M}^{-1}\text{cm}^{-2}$, whereas the use of aSPCEs achieved a great sensitivity enhancement ($0.180 \pm 0.003 \mu\text{A}\mu\text{M}^{-1}\text{cm}^{-2}$), i.e. a sensitivity increase ca. 60-fold with this activation treatment.

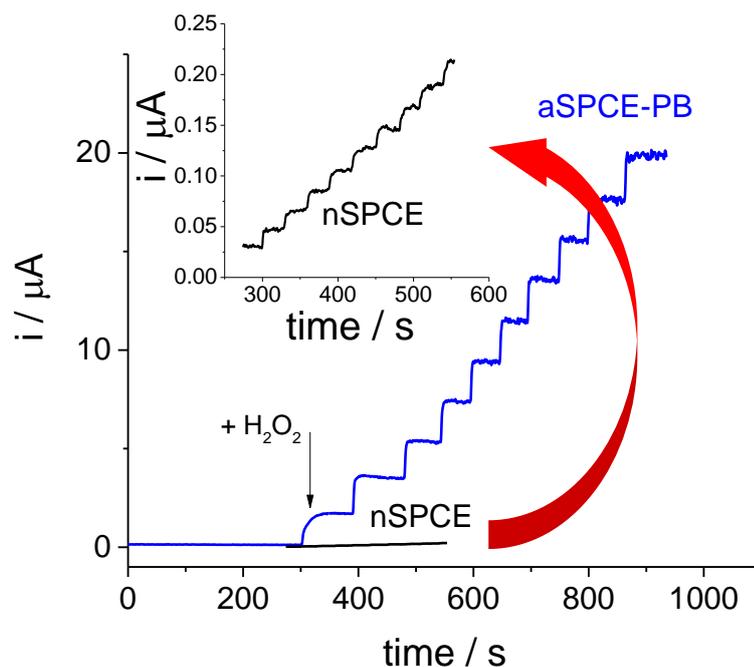


Figure 1. Amperometric profiles at 0.7 V to 80 μM additions of H_2O_2 using nSPCE and aSPCE-PB. The aSPCE-PB was pretreated by performing one cyclic voltammogram in 0.1 M PB pH 7 between -2 and +2 V at 10 mVs^{-1} in the presence of ozone. The inset shows an amplification of the black plot. The amperometric measurements were taken in 0.1 M PB, pH 7.

Figure 2 shows the effect of pH on the electrochemical activation of the electrodes using as supporting electrolytes NaOH (pH 12.7, aSPCEs-NaOH), PB (pH 7, aSPCEs-PB) and H_2SO_4 (pH 0.6, aSPCEs- H_2SO_4) solutions. Higher electrochemical output towards H_2O_2 electrooxidation was obtained with aSPCEs-NaOH whereas the lowest electroanalytical signal was achieved for that one using aSPCEs- H_2SO_4 . The sensitivities for the aSPCEs were 0.103 ± 0.001 , 0.180 ± 0.003 and $0.333 \pm 0.004 \mu\text{A} \mu\text{Mcm}^{-2}$ for aSPCE- H_2SO_4 , aSPCE-PB and aSPCE-NaOH, respectively. The pH is one of the most important factors to be studied, as demonstrated by the ozone decomposition rate with pH [24]. Therefore, the higher activity obtained at alkaline pH could very likely be related with an increase in the number of hydroxyl radicals generated during O_3 decomposition, as a consequence of its interaction with hydroxide ions in water [24, 25]. Thus, even though the radicals generated are very short-living species, they get an even stronger oxidizing power than that of ozone itself [25]. Furthermore, the application of high electrode potentials result in driving force of diffusion of the above radical intermediates from the bulk

solution to the electrode surface, thereby, changing the electrochemical properties of the carbon ink.

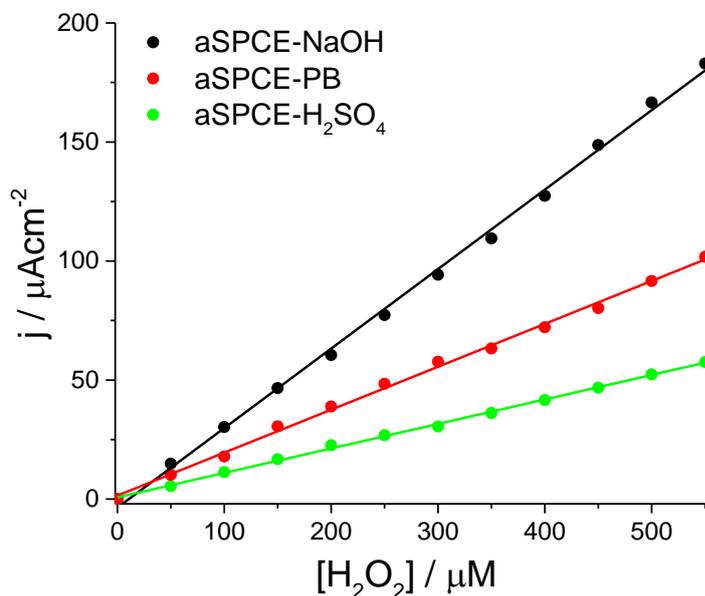


Figure 2. Calibration plots of H₂O₂ electrooxidation obtained at aSPCE-NaOH, aSPCE-PB and aSPCE-H₂SO₄. The points stand for experimental data, and the lines for the results obtained by lineal regression to a first-order equation. The amperometric measurements were taken in 0.1 M PB, pH 7.

The synergistic effect between the application of the electrode potential and use of ozone is explored in Figure 2S. In this regard, the amperometric profile towards H₂O₂ was measured by using electrodes after the complete ozonation treatment (aSPCEs-NaOH), electrodes treated in the absence of ozone, and electrodes treated with ozone but without applied potential. The electrochemical treatment in the absence of ozone produced electrodes with a sensitivity of 0.036 μAμM⁻¹, whereas a sensitivity of 0.056 μAμM⁻¹ was achieved for electrodes activated using the complete treatment. Therefore, the application of ozone has improved the analytical signal approximately 35%, which corroborates that both the applied potential and ozone are required to obtain greater analytical outcomes.

Carbon surfaces can be activated by applying anodic and/or cathodic current densities using constant potentials [12, 26]. In order to know if these protocols are appropriate for the activation of SPCEs, the O₃-based voltammetric activation using

NaOH was compared with other methods of activation based on controlled potential pretreatments (under bubbling ozone in 0.1 M NaOH). The results obtained in the measurement of H₂O₂ using electrodes activated by this kind of pretreatments are shown in Figure 3S (Supporting Information). Results revealed that the maximum response to H₂O₂ was achieved by the voltammetric method.

It has also been documented that it is possible the electrochemical activation of other carbon surfaces such as carbon nanotube (CNT)/polymer composite [27] and GPH electrodes [28]. Commercial GPH- and CNT-SPCEs were subjected to the O₃-based activation treatment in 0.1 M NaOH and the amperometric signal to H₂O₂ was evaluated. The results are shown in Figure 4S, which probe that the above protocols are able to activate both GPH- and CNT-SPCEs but in lesser extent than nSPCEs (Figure 3S, black points).

3.2. Characterization of aSPCEs

We determined the electroactive surface areas, the percentages of electroactive areas and the roughness factors for the nSPCE and the different aSPCEs, which are compiled in Table 1S. All the aSPCEs showed higher percentages of electroactive area and roughness factors. The aSPCEs-NaOH exhibited the highest electroactive area, in the order of twice that of the nSPCEs. This indicates that the activation process might have produced defects in the carbonaceous inks due to the loss of non-electroactive material, thereby increasing its porosity. Nevertheless, the SEM analysis shown in Figure 5S revealed scarcely noticeable changes in the electrode surface topologies, although a slightly more porous surface could barely be observed for the aSPCEs-NaOH which correlates with the higher electroactive surface area.

The XPS analysis proved certain modifications in the carbonaceous inks of aSPCEs (Figure 3) after the activation pretreatment. The analysis of the C1s and O1s spectra showed a decrease of atomic content (at.%) of carbon and an increase of the at.% of oxygen in the aSPCEs compared to the nSPCE (Figures 3A and B). These additional oxygen functionalities are related with the enhancement of the electrochemical response of SPCEs, as previously reported in [14, 20]. XPS deconvolution of the C1s spectra showed the presence of peaks at 284.6, 285.5, 286.4 and 288.6 eV, assigned to graphitic C-C, C-C and/or C-H and/or C-N, C-O and C=O, respectively [29]. Figure 3C shows that the at.% of graphitic C-C decreased in all the aSPCEs while C=O increased, compared to the nSPCEs. With regards to aSPCEs-NaOH, XPS showed a higher at.% of C-C and/or C-H

and/or C-N. Figure 3C also shows the increment of the ratio C=O/C-O as the pH increases. On the other hand, the XPS deconvolution of O1s showed energy peaks at 532.4, 533.7 and 534.8 eV, which were ascribed to C=O, C-O and C-OH groups, respectively ^[30]. In all the cases, the oxygenated species were higher for the aSPCEs, which corroborates that the increment of the electroactivity is related with the introduction of oxygenated functionalities on the surface. This results in an increase of the hydrophilicity and wettability of the electrode surface. The aSPCEs-PB and aSPCEs-H₂SO₄ showed a clear increase in the amount of C=O whereas an increase in the amount of C-O was observed in all the aSPCEs (Figure 3D). The existence of C-OH was only demonstrated in the aSPCEs-PB and aSPCEs-NaOH, being the ratio C-OH/C=O higher at the higher pH (Figure 3D). These aspects, together with the increment in percentage of electroactive area, could be the cause of the improvement in the electrochemical properties of aSPCEs. As a matter of fact, these issues have been linked by other authors to defects in carbonaceous materials as important promoters of electron transfer ^[17, 31].

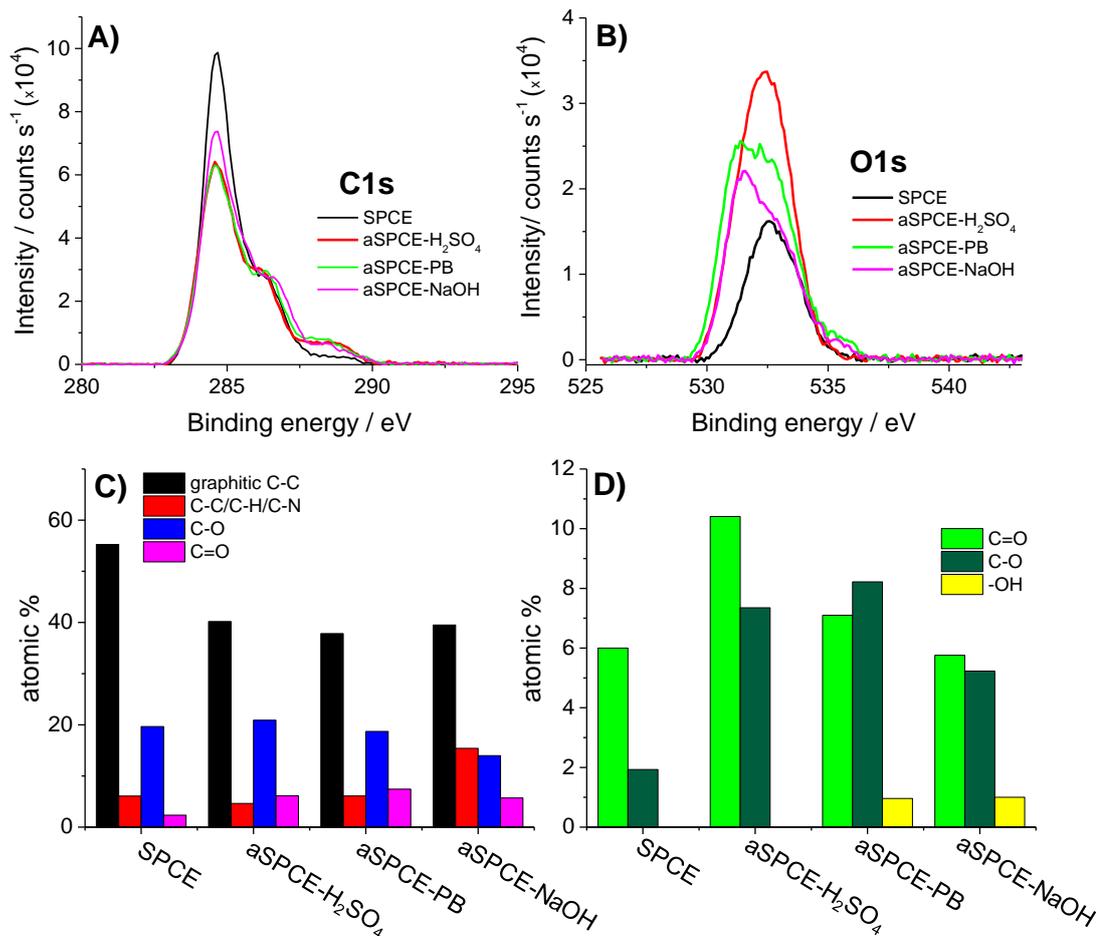


Figure 3. XPS C1s (A) and O1s (B) spectra for the nSPCE, aSPCE-H₂SO₄, aSPCE-PB and aSPCE-NaOH. Atomic % of functional groups after deconvolution of C1s (C) and O1s (D) core level spectra, respectively.

Figure 4 shows the Bode plots of the impedance modulus $|Z|$ and the phase angle for an nSPCE and the aSPCEs. The plot of the modulus $|Z|$ vs log frequency gives some qualitative information about the electrode interfacial processes before and after the activation process. Figure 4A shows that the impedance values decreased after the ozone treatment at lower frequencies in all aSPCEs, whose data agree with the enhanced electrochemical properties found for the electrooxidation of H₂O₂ when using aSPCEs, especially in aSPCEs-PB and aSPCEs-NaOH. This fact was mainly attributed to the increment in oxygen functionalities on the electrode surface ^[16] (Figure 3) and the increment of electroactive area ^[32] (Table 1S).

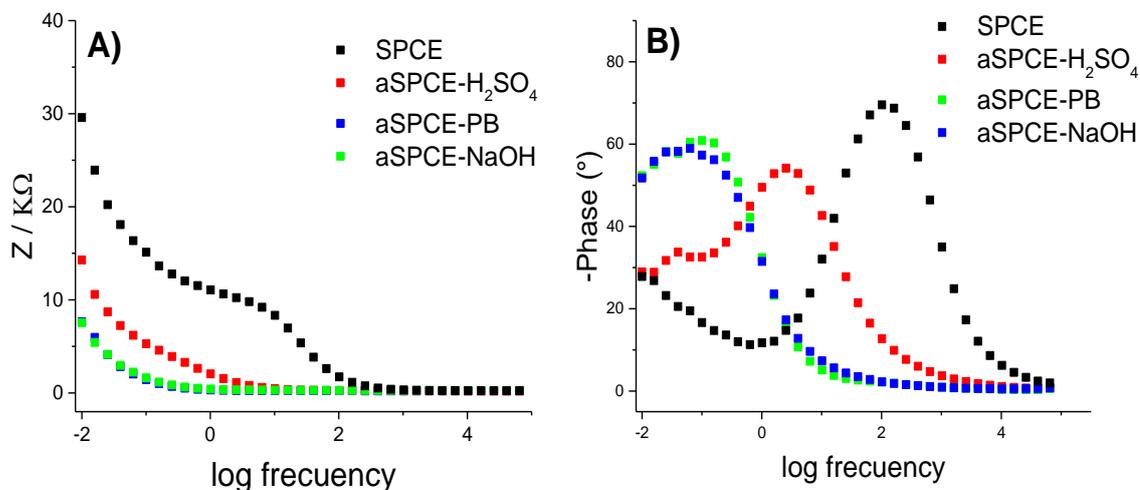


Figure 4. Bode plots of the impedance modulus $|Z|$ (A) and the phase angle (B) of an nSPCE (black dots), and the different aSPCEs as a function of log frequency.

The constant phase angle impedance is a measure of the non-faradaic impedance arising from the interface capacitance or polarization^[33]. The maximum phase angle of the nSPCE seen on the Bode Plots is about 75° (Figure 4B), less than the value of 90° expected for an ideal capacitor. In addition, Figure 4B shows that the maximum phase angles in all the aSPCEs decreased towards 55-60° and shifted to lower frequencies. The higher deviation from the ideal capacitive behavior in aSPCEs could be attributed to surface inhomogeneity, roughness effects, and variations in properties or composition of surface layers^[34].

3.3. Analytical application of aSPCEs-NaOH

Activation pretreatment of nSPCEs at alkaline media was chosen to get the electroanalytical figures of merit to H_2O_2 and phenolic compounds electrooxidation. The nSPCEs showed a sensitivity to H_2O_2 of $3.2 \pm 0.05 \text{ nA}\mu\text{M}^{-1}\text{cm}^{-2}$ (see Figure 1), while this parameter was $0.33 \pm 0.05 \mu\text{A}\mu\text{M}^{-1}\text{cm}^{-2}$ after the pre-treatment with ozone in NaOH (see Figure 2), with a sensitivity increase of more than 100 times. The sensitivity obtained for the electrooxidation of H_2O_2 was in the same order of magnitude than that previously reported with the electrochemical treatment using H_2O_2 as the activating agent^[20]. However, of even greater importance is the fact that the activation time resulted considerably reduced with respect to that treatment with a time saving of up to 92% -

from 2.5 h to 12 min. Moreover, calibrations plots to H₂O₂ sensing were performed at 0, 10, 20 and 30 days after the activation process, as depicted in Figure 6S (Supporting Information), with sensitivities of 0.0566 $\mu\text{A}\mu\text{M}^{-1}$ and 0.0543 $\mu\text{A}\mu\text{M}^{-1}$, at 0 and 30 days of storage, respectively, which means a decrease in the signal of only 4.1 %. In addition, aSPCEs-NaOH exhibited a good stability when taking measurements of 80 μM H₂O₂, as shown in the inset of Figure 6S (Supporting Information).

To extend the applicability of the aSPCEs-NaOH, their electroanalytical response towards several important phenolic compounds was explored. APAP, DP and HQ were chosen as model species accordingly the great importance in chemical and pharmaceutical industry [35]. The CVs of 1 mM of each of these compounds measured by an nSPCE and an aSPCE-NaOH are shown in Figure 5. In all the cases, the aSPCE-NaOH achieved higher peak current intensities and lower oxidation potentials than the nSPCE (Figures 5A, B and C). Furthermore, a lower peak-to-peak separation (ΔE_p) was observed in all the cases compared to that one using the nSPCE. In particular, ΔE_p was 0.361, 0.414 and 0.367 V for HQ, APAP and DP, respectively, using nSPCEs, and 0.180, 0.236 and 0.231 V using aSPCEs-NaOH. This means that the oxidation-reduction process of these compounds was significantly more reversible when using aSPCEs-NaOH. Moreover, the aSPCEs-NaOH can strikingly detect these three compounds simultaneously by just one CV, depicting all three oxidation and reduction peaks unlike when using nSPCEs where only two oxidative and one reductive peaks could be depicted, (Figure 5D).

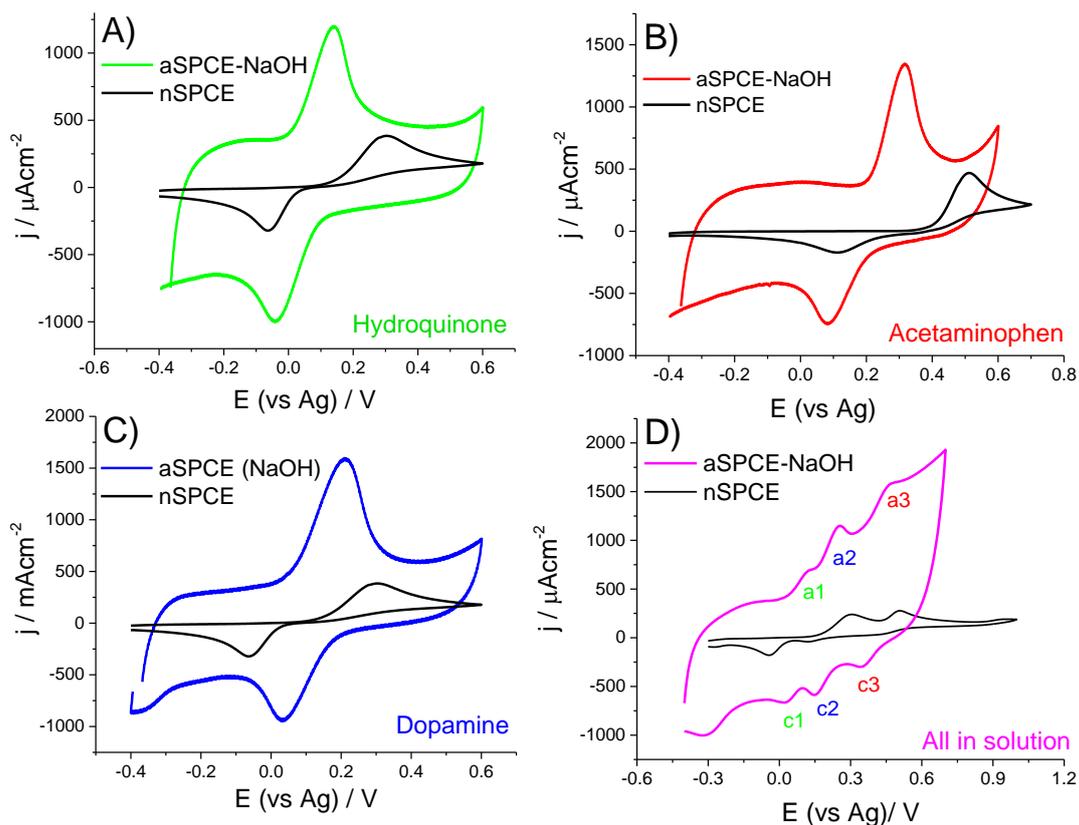


Figure 5. Voltammetric responses (first cycle) of nSPCEs and aSPCEs-NaOH towards 1 mM of: A) HQ, B) APAP, C) DP and D) all together. In Figure D, a1 and c1: anodic and cathodic peaks for HQ, a2 and c2 for DP; and a3 and c3 for APAP, respectively. The voltammetric measurements were taken in 0.1 M PB, pH 7.

Similarly to what happened in the electrooxidation of H_2O_2 , it was also demonstrated that HQ, APAP and DP have better electroanalytical signals using aSPCEs-NaOH than aSPCEs-PB or aSPCEs- H_2SO_4 (Table 2S). This higher signal could be related with the significant content of C-OH groups on the surface (Figure 3), which could have formed hydrogen bonds with the hydroxyl groups of phenols, facilitating their electro-oxidation [36].

Scan rate effect on the electrochemical behavior of the phenolic compounds was performed using the aSPCEs-NaOH. Tables 3S, 4S and 5S (Supporting information) compile the anodic and cathodic peaks electrode potential values (E_{p_a} and E_{p_c}), the ΔE_p and the anodic to cathodic ratios of peak current intensity (I_{p_a}/I_{p_c}) as a function of the scan rate for HQ (Table 3S), APAP (Table 4S) and DP (Table 5S). As expected, the ΔE_p increased with the scan rate for all the compounds associated to a quasi-reversible

process. Furthermore, the I_{p_a}/I_{p_c} ratio values were higher than the unity for APAP and DP, which can be attributed to complex coupled chemical reactions involving APAP, DP and/or its oxidation products^[37]. In the case of HQ, in general, we obtained I_{p_a}/I_{p_c} near the unit, which agrees with a more reversible redox behavior^[38]. Similar behavior was also reported by these compounds when using SPCEs activated with a combined H_2SO_4/H_2O_2 activation method^[21].

Double logarithmic plots of I_{p_a} vs scan rate using the nSPCEs and aSPCEs-NaOH is shown in Supporting information (Figure 7S). Before activation, a slope of ~ 0.5 for the three compounds was found, in agreement with the slope for diffusion controlled processes^[22]. For HQ and APAP, the slopes also showed a diffusion controlled process when using the aSPCEs-NaOH. However, this slope changed to 0.69 for DP when using the aSPCEs-NaOH, which means that its redox process is only partially controlled by diffusion^[22]. This result indicate that the changes achieved on the electrode after the activation may favor the adsorption of DP molecules on the electrode surface, without being adsorption the determining step for its electrooxidation.

A proof of concept of the electroanalytical determination of these phenolic compounds by using the aSPCEs-NaOH is depicted in Figure 6. The nSPCEs gave sensitivities of 0.37 ± 0.02 , 0.40 ± 0.02 and $0.32 \pm 0.01 \mu A \mu M^{-1} cm^{-2}$ and limits of detection (LODs) of 5.6, 3.7 and 3.7 μM for HQ, APAP and DP, respectively. These analytical parameters were improved using the aSPCEs-NaOH. In that case, the sensitivities were 1.12 ± 0.02 , 0.63 ± 0.02 and $0.94 \pm 0.02 \mu A \mu M^{-1} cm^{-2}$ with LODs of 1.3, 3.2 and 1.3 μM for HQ, APAP and DP, respectively. Therefore, the aSPCEs-NaOH improve analytical outcomes and they are preferable to the analytical determination of these compounds.

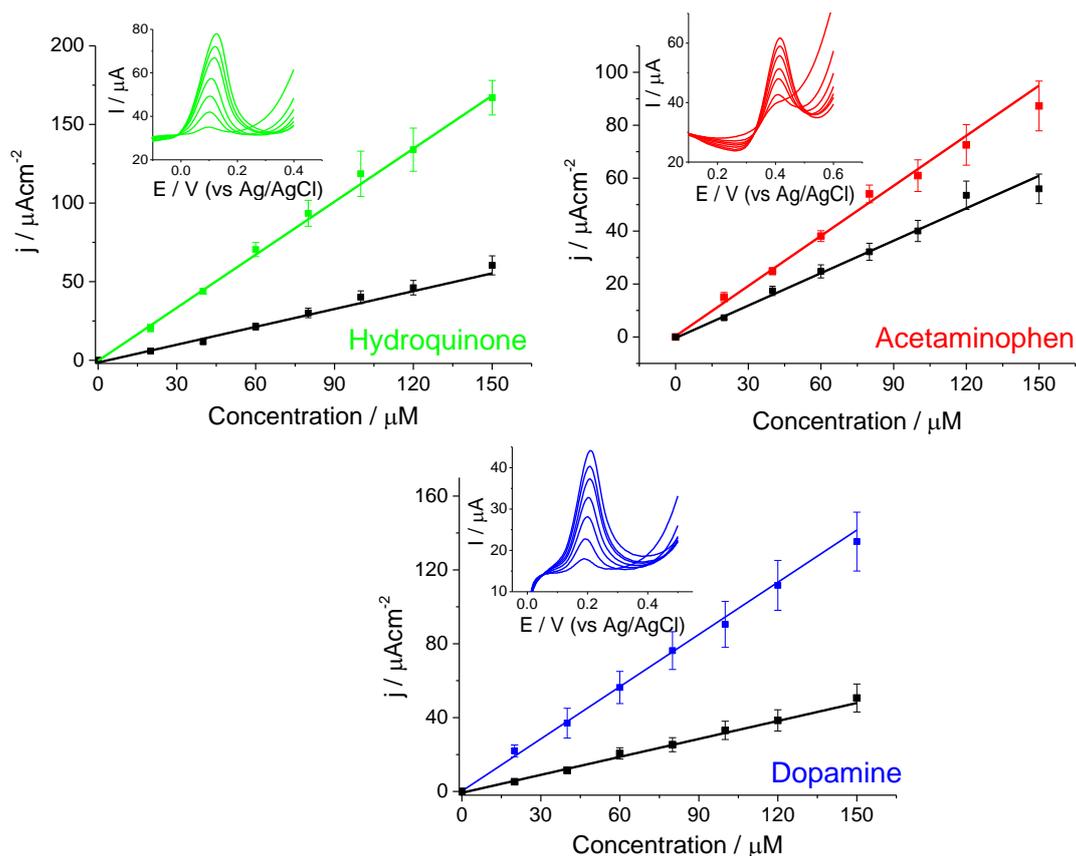


Figure 6. Plots of current density vs. HQ, APAP and DP concentrations using aSPCEs-NaOH (coloured plots) and nSPCEs (black plots). The measurements were taken in 0.1 M PB, pH 7 and were taken in triplicate. Error bars represent the standard deviation of uncertainty. Insets: LSVs of the aSPCEs-NaOH to increasing phenol derivative compound concentrations.

Some analytical parameters of the aSPCEs-NaOH to the HQ, APAP and DP electrooxidation were compared with other sensors reported in the literature (Table 1). The sensitivities and the LODs obtained with the aSPCEs-NaOH are similar or sometimes better than those given for other electrodes previously modified with different materials. Therefore, our simple electrochemical activation method combined simultaneously with ozone gas can produce highly activated carbonaceous electrodes with similar electroanalytical achievements than other electrodes with more sophisticated or time consuming modified protocols.

Table 1. Electroanalytical parameters towards HQ, APAP and DP for a variety of sensors.

Electrode	Phenol	Sensitivity ($\mu\text{A}\mu\text{M}^{-1}\text{cm}^{-2}$)	Area (cm^2)	LOD (μM)	Reference
Fe-doped ZnO NRs	HQ	1.80×10^{-2}	0.130	0.5	[39]
Graphene modified GCE	HQ	1.38	0.070	12.0	[40]
Pt-graphene modified GCE	HQ	3.56	0.070	6.0	[40]
aSPCE-NaOH	HQ	1.12	0.168	1.3	This work
HRP-poliacrylamide microgel-GCE	APAP	0.07	0.031	3.1	[41]
CeO ₂ -SPCE	APAP	6.62 (0.09-7 μM); 0.09 (16.4-1160 μM)	0.067	0.05	[42]
aSPCE-NaOH	APAP	0.63	0.168	3.2	This work
ND-SPEs	DP	0.27	0.075*	0.6	[43]
NiO-RGO/ITO electrode	DP	1.48	0.700	1.0	[44]
GME	DP	0.93	0.071*	2.6	[45]
aSPCE-NaOH	DP	0.94	0.168	1.3	This work

*These areas were calculated as πr^2 , being r the electrode radius.

GCE: glassy carbon electrode, GME: graphene modified electrodes, HRP: horseradish peroxidase, ITO: indium tin oxide, MWCNT: multiwall carbon nanotubes, ND: nanodiamond, NRs: nanorods, PANI: polyaniline, RGO: reduced graphene oxide.

4. Conclusions

The activation protocol of SPCEs based on a simple voltammogram in the presence of ozone gas in alkaline medium provided highly improved electrochemical properties and promising analytical applications for the sensing of H₂O₂ and certain phenolic derivatives. The activation protocol consists of just one voltammetric cycle between -2 and +2 V (vs Ag) at 10 mVs⁻¹ in 0.1 M NaOH while ozone gas is continuously bubbled through the solution. After this pretreatment, the electrodes achieved a sensitivity more than 100-fold higher than the nSPCEs (for the electrooxidation of H₂O₂). Such improvement was attributed to the increment in roughness and to the modification of the carbonaceous surface with the incorporation of oxygen functionalities. Impedance measurements demonstrated that the electrochemical activation increased the

conductivity in aSPCEs. The proposed method resulted very effective, not only for H₂O₂ electroanalysis, but also for the determination of APAP, HQ and DP. This facile protocol is a promising procedure for the activation of carbonaceous inks at SPCEs. Moreover, by taking into account the short duration of the activation pretreatment (only 12 minutes), it could be routinely used prior to any electrode surface modification conducted for electroanalytical applications.

Acknowledgements

This work was funded by the Spanish Ministry of Economy and Competitiveness (MINECO, <http://www.mineco.gob.es/portal/site/mineco/idi>), Projects No. BFU2016-75609-P (AEI/FEDER, UE) and CTQ2016-76231-C2-2-R, and by the Junta de Comunidades de Castilla-La Mancha (Spain), Project No. SBPLY/17/180501/000276/2 (cofunded with FEDER funds, EU). BGM is a post-doctoral research fellow of the Youth Employment Initiative (JCCM, Spain, cofunded with ESF funds, EU). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

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