

Screen-printed electrodes for the determination of iridium in drugs

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Abstract

Among the impurities that must be controlled in drugs and pharmaceuticals, metal ions are considered. These metal impurities appear in those products because of their use as catalysts. Although Pharmacopeias recommend ICP based methods for their determination, the use of electroanalytical techniques can be considered as a sensible, selective and non-expensive alternative for the determination of heavy metal ions in general and metal impurities in particular. In the present work, we propose a differential pulse voltammetric method based on the use of carbon based-screen-printed electrodes (graphite, nanotubes or nanofibers) that allows the determination of iridium in drugs at the concentration limits recommended by official organisms. Carbon screen-printed electrode modified with nanotubes (CNT-SPCE) is the sensor that provided the best LOD and LOQ (0.03 and 0.10 mg L⁻¹ respectively). Then, the proposed method was applied to the voltammetric determination of iridium in spiked drug samples purchased in local chemists using CNT-SPCE.

Keywords: metal impurities, drugs, iridium, voltammetry, screen-printed electrodes

Introduction

Organic and inorganic compounds and residual solvents are the three types of impurities that must be controlled in drugs and in their ingredients, and by extension in certain foods and dietary supplements because of their potential toxic effects in humans [1-6]. Inside inorganic impurities, metals like iridium, rhodium, ruthenium and osmium must be considered. These metals, classified as Class 1B by the European Agency for the Evaluation of Medicines (EMA), can be introduced during the manufacturing process and their concentration must be controlled in the final product as impurities. Table 1 shows the exposure and the concentration limits of different metals according EMA [2].

To control their presence in drugs and pharmaceutical, methodologies with high sensitivity and selectivity must be developed to substitute the traditional procedures described in all international pharmacopoeias for metal impurities determination that have serious limitations. In this sense, a method used for monitoring inorganic contaminants in drugs is based on a classic colorimetric test described in the USP General Chapter <231> [7]. This method is based on the precipitation of sulfide compounds after reaction with thioacetamide, in which the resulting colored precipitate is compared visually to a 10 mg L⁻¹ Pb(II) standard solution to determine compliance with the heavy metal limit concentration. Besides the associated error with the subjective visual comparison, this method is only useful for Ag, As, Bi, Cd, Cu, Hg, Mo, Pb, Sb and Sn. Thus, this method can neither provide selectivity nor give individual concentration of each particular element. In addition, it cannot be used to determine elements like chromium or platinum group elements that are commonly used as catalysts. To solve this problem, USP General Chapter <233> [3] recommends the use of multi-element techniques like inductively coupled plasma mass spectrometry (ICP-MS) and inductively coupled plasma atomic emission spectroscopy (ICP-AES) instead of the colorimetric test described above. Relevant papers devoted to the determination of elemental impurities in pharmaceutical products by ICP based techniques have been published in the last years [6,8-11]. However, the high cost of the required instrumentation appears as the main drawback of these methods. In this point, electroanalytical techniques can be considered as a sensible, selective and non-expensive option for the determination of heavy metals in general and metal impurities in particular. In the literature, a great number of papers devoted to the analysis of some of these analytes have been published [12-16]. In these papers as well as in the works cited therein, a lot of metal ions, matrices and electrodes have been considered. However, the number of works devoted to the determination of elements of platinum group and in particular to iridium was very scarce. Locatelli [17-19] proposes a voltammetric determination method for iridium in natural samples by square wave

adsorptive-catalytic stripping voltammetry (SWAdCSV) in a glassy carbon electrode at pH 4.8 media containing NaBrO_3 and L-1-cetyltrimethylammonium bromide (CTAB) allowing very favorable detection limits. Svancara *et al.* [20] consider a carbon paste electrode modified with a surfactant for the determination of platinum group elements both in model solution and in real industrial wastewaters. In any case, the use of screen-printed electrodes (SPE) has not been considered for the analysis of iridium. On the other hand, the number of publications considering the use of electroanalytical techniques for the analysis of metal impurities in drugs is very limited, as far as we know, only a work by Guna *et al.* [21] aims on the development of a potentiometric sensor for the analysis of some metal ions in drugs and pharmaceuticals. In this work, although a great number of metal ions have been considered no reference to iridium appears.

In the present work, a simple voltammetric methodology using carbon based screen-printed electrodes has been proposed for the analysis of iridium in drugs at the levels recommended by official organisms. The screen-printed electrodes considered were a graphite screen-printed electrode (SPCE), a carbon screen-printed electrode modified with carbon nanotubes (CNT-SPCE) and a carbon screen-printed electrode modified with nanofibers (CNF-SPCE). The proposed differential pulse voltammetric methodology was applied to the determination of iridium in spiked commercial drugs using a CNT-SPCE as the optimal sensor.

2. Experimental

2.1 Reagents

Ir(IV) working solutions were prepared by the appropriate dilutions of an atomic standard solution of 1.000 g L^{-1} of Ir(IV) from Sigma-Aldrich (St. Louis, USA). Ultrapure sulphuric acid (Merck, Darmstad, Germany) was used to control the pH and as supporting electrolyte.

All solutions were prepared in ultrapure water obtained from a Milli-Q plus 185 system, Millipore.

Adolonta[®] and Trigon[®] drug samples were purchased in local chemists.

2.2 Apparatus

Cyclic voltammetric (CV) and differential pulse voltammetric (DPV) experiments were performed in a μ Autolab Type III (Ecochemie, The Netherlands) coupled to a Metrohm 663VA Stand

(Metrohm, Switzerland) and a personal computer with GPES version 4.9 data acquisition software (Ecochemie).

The electrochemical experiments were carried out using different working electrodes: i) a graphite screen-printed electrode (SPCE) referenced as DRP-110 (DropSens, Oviedo, Spain); ii) a carbon screen-printed electrode modified with carbon nanotubes (CNT-SPCE) referenced as DRP-110CNT (DropSens); and iii) a carbon screen-printed electrode modified with carbon nanofibers (CNF-SPCE) referenced as DRP-110CNF (DropSens). As reference and auxiliary electrodes, external Ag/AgCl/KCl (3 mol L⁻¹) and carbon electrodes were respectively used unless otherwise indicated. It should be pointed out that internal reference and auxiliary electrodes of the screen-printed devices were also considered in some of the performed experiments.

2.3 Voltammetric measurements

Voltammetric measurements were performed by introducing in the voltammetric cell a certain volume of standard or sample solution in 0.016 mol L⁻¹ H₂SO₄ media. Instrumental parameters were 50 mV of pulse amplitude with a modulation time of 50 ms applied at interval times of 0.5 s. The cathodic scan rate applied was from 1.1 V to 0.35 V. After each measurement, a conditioning potential of 1.1 V was applied during 30 s to remove possible residues of iridium from the electrode surface. When measurements were performed in drop (using the three-electrode cell of the SPE), the measuring solution was prepared in a volumetric flask and a drop of the solution was deposited on the surface of the CNT-SPCE covering the area of the three electrodes. Instrumental parameters were equal to those considered in cell measurements.

3.- Results and discussion

To characterize the electrochemical redox processes of Ir(IV) solution, cyclic voltammetric experiments were performed using a SPCE. Figure 1 shows a cyclic voltammogram of 60 mg L⁻¹ Ir(IV) solution in 0.016 mol L⁻¹ H₂SO₄ media using a scan rate of 100 mV s⁻¹. Scans were performed from 1.2 V to -0.5 V and returned to 1.2 V. Two well-defined cathodic peaks close to 0.8 V (peak 1) and 0.63 V (peak 2) were observed together with their respective anodic peaks close to 0.83 V (peak 1') and 0.70 V (peak 2'), pointing out the reversibility of the redox processes. The cathodic peaks can be attributed to the reduction of Ir(IV) to Ir(III) (peak 1) and of Ir(III) to Ir(0) (peak 2), and the respective anodic ones to the reverse processes (peak 1' and peak 2'). The analysis of the dependence of the peak current intensity in front of the square root of the scan rate (from 10 mV s⁻¹ to 1000 mV s⁻¹) was considered showing a linear relation between both

parameters which indicates that all the processes are controlled by diffusion (results not shown). Moreover, at potentials from -0.3 V and 0.15 V some small signals are also observed (peaks 3, 4 and 3') that can be associated to catalytical signals related with protons [22,23].

Figure 2 shows the differential pulse voltammogram of 40 mg L⁻¹ Ir(IV) solution in 0.016 mol L⁻¹ H₂SO₄ media using a SPCE in which two peaks corresponding to the reduction of Ir(IV) to Ir(III) (peak 1) and of Ir(III) to Ir(0) (peak 2) can be observed. With the aim to establish which of both peaks is the most appropriate for analytical purposes, different studies were performed considering the three screen-printed electrodes above indicated: SPCE, CNT-SPCE and CNF-SPCE. The obtained results are shown in Table 2. Reproducibility for each electrode was computed from 5 independent measurements of a 60 mg L⁻¹ Ir(IV) solution using 5 different units; whereas repeatability was obtained from 10 consecutive measurements of the same solution carried out using the same SPE unit. As it can be seen in this table, reproducibility and repeatability results obtained for each peak at each considered SPE pointed out that peak 1, corresponding to the reduction process of Ir(IV) to Ir(III), performs better than peak 2 (reduction process of Ir(III) to Ir(0)). Therefore, peak 1 will be further considered for analytical purposes. Moreover, according to the obtained results, it can be concluded that CNT-SPCE would be the more convenient sensor in term of reproducibility and repeatability.

With the aim to establish the optimal concentration range for the determination of Ir(IV), DPV measurements of different solutions containing Ir(IV) from 0.1 mg L⁻¹ to 100 mg L⁻¹ in 0.016 mol L⁻¹ H₂SO₄ media using the three considered SPEs were performed. Figure 3a shows the obtained voltammograms using a SPCE. Again, it can be observed the presence of two overlapped peaks corresponding to peak 1 and peak 2. The obtained calibration plot is shown in Figure 3b. This plot presents two different lineal ranges from 0.1 to 10 mg L⁻¹, and around 10 mg L⁻¹ to 100 mg L⁻¹, being 100 mg L⁻¹ the highest concentration considered. Similar results were observed for the other studied SPEs (results not shown). Taking into account that the interest is related to the determination of Ir(IV) at low concentrations, in the following the lower linear range will be considered. Thus, linear calibration curves were carried out by DPV using SPCE, CNT-SPCE and CNF-SPCE by measuring increasing concentrations of Ir(IV) ranging from 0.1 to 10 mg L⁻¹ (Figure 4). The obtained voltammograms confirm that the peak 1 is the most appropriate for analytical purposes because the height of peak 2 is lower and appears at higher Ir(IV) concentrations.

Calibration data for the determination of Ir(IV) using SPCE, CNT-SPCE and CNF-SPCE are reported in Table 3. Sensitivities were calculated as the slope of the calibration curves and limits of detection (LOD) and quantification (LOQ) were computed as 3 and 10 times, respectively, the

standard deviation of the intercept over the slope of the calibration plot. The LOQ was considered as the lowest value of the linear concentration range.

As it can be seen in Table 3, the lowest LOD was achieved using a CNT-SPCE. Although CNF-SPCEs usually provide better LODs [23], it must be pointed out that in this study, the baseline obtained with CNF-SPCE shows a great contribution of capacitive currents in comparison to that of the other SPEs considered that leads to higher LODs. From these studies, it can be concluded that taking into account that the objective is to determine low concentrations of Ir(IV) in drugs and the reproducibility and repeatability results (Table 2), the optimal sensor for this aim is a SPCE modified with CNTs.

In order to test the applicability of this methodology to a real drug, the analysis of Ir(IV) was attempted in drug samples purchased in local chemists. The considered drugs were Adolonta[®] that contains, as active pharmaceutical ingredient (API), tramadol hydrochloride that it is used for the treatment of moderate to severe pain, and Trigon[®] that contains triamcinolone acetonide as API and it is used as anti-inflammatory drug. For their analysis these samples were previously spiked with Ir(IV). The level of Ir(IV) added was evaluated taking into account the recommended dose of the API per day, the recommended limit concentration of iridium per day, and the sample dilution required to obtain the measuring solution in 0.16 mol L⁻¹ H₂SO₄ media. According to these points, samples of Adolonta[®] and Trigon[®] were spiked with an Ir(IV) concentration that results in 0.666 mg L⁻¹ and 1.333 mg L⁻¹ m respectively in the measuring solution. The external calibration method was applied for quantification and the obtained results are shown in Table 4. Proper recoveries were achieved pointing out that this methodology is appropriate for the analysis of Ir(IV) at these levels of concentration in drugs by DPV in CNT-SPCE. Moreover, this determination was also performed considering the particular properties of SPEs that allow carrying out the determination using only a drop sample since the three electrodes of the electrochemical cell are integrated in the device. The good results obtained considering the drop analysis are also shown in Table 4.

From these studies it can be concluded that CNT-SPCEs are very suitable for the determination of Ir(IV) in drug samples by DPV at the concentration levels between 0.10 and 10 mg L⁻¹, which has allowed the detection of concentration levels lower than the maximum concentration level recommended per day.

Conclusions

In this work the voltammetric determination of Ir(IV) in drugs using carbon based- SPEs (SPCE, CNT-SPCE or CNF-SPCE) was studied as a sensible, selective and non-expensive alternative to ICP based methods. Cyclic voltammetric measurements show that the reversible and diffusion controlled reduction process of Ir(IV) in a 0.016 mol L⁻¹ H₂SO₄ media takes place in two steps, first the reduction of Ir(IV) to Ir(III) and later the reduction of Ir(III) to Ir(0). Studies performed by DPV show that the peak related to the Ir(IV) to Ir(III) reduction process is the most appropriate for analytical purposes. Figures of merit of the procedure show that the best sensor is that modified with nanotubes (CNT-SPCE) providing both the lower LOD and LOQ values (0.03 and 0.10 mg L⁻¹ respectively), and the best repeatability and reproducibility. Then, the proposed method was successfully applied to the voltammetric determination of iridium in spiked drug samples purchased in local chemists using a CNT-SPCE, providing good reproducibility and trueness inferred by the RSD and the relative error respectively.

Acknowledgements

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Table 1. Classification and recommended limits of concentration of elemental impurities [2].

Classification	Oral exposure		Parental exposure		Inhalat exposi
	PDE*** (µg per day)	Concentration (mg L ⁻¹)	PDE (µg per day)	Concentration (mg L ⁻¹)	PDE (ng per
Class 1A: Pt, Pd	100	10	10	1	Pt: 70
Class 1B: ** Ir, Ru, Rh, Os	100	10	10	1	
Class 1C: Mo, Ni, Cr, V Metals with significant safety control	250	25	25	2.5	Ni: 10 Cr(IV):
Class 2: Cu, Mn Metals with low safety concern	2500	250	250	25	
Class 3: Fe, Zn Metals with minimal safety concern	13000	1300	1300	130	

*Pt as hexachloroplatinate

**The total amount of listed metals should not exceed the indicated limit

***PDE: Permitted daily exposure

Table 2.- Reproducibility and repeatability obtained measuring a solution of 60 mg L⁻¹ of Ir(IV) in 0.016 mol L⁻¹ H₂SO₄ media by DPV using the SPCE, CNT-SPCE and CNF-SPCE.

	SPCE		CNT-SPCE		CNF-SPCE	
	Peak 1	Peak 2	Peak 1	Peak 2	Peak 1	Peak 2
Reproducibility (RSD %)	6	10	1	4	5	10
Repeatability (RSD %)	5	8	2	1	2.5	4

Table 3.- Calibration data, obtained by DPV using SPCE, CNT-SPCE and CNF-SPCE.

Working electrode	Calibration plot ^a	R ²	LOD (mg L ⁻¹)	LOQ (mg L ⁻¹)	Concentration range (mg L ⁻¹)
SPCE	$y = -0.1332 x + 0.0072$	0.9945	0.05	0.15	0.15 - 10
CNT-SPCE	$y = -0.1136 x + 0.0214$	0.9939	0.03	0.10	0.10 - 10
CNF-SPCE	$y = -0.1321 x + 0.0374$	0.9997	0.14	0.48	0.48 - 10

^a y peak current in μA and x Ir(IV) concentration in mg L^{-1}

Table 4.- Results obtained in the analysis of drugs spiked with Ir(IV). Measurements performed by DPV using CNT-SPCE as working electrode in cell or in drop. Standard deviation are denoted by parenthesis (n=3).

Adolonta®		Expected	Found
In cell	c (mg L ⁻¹)	0.666	0.68
	RSD (%)	--	6.1
	Relative error (%)	--	1.7
In drop	c (mg L ⁻¹)	0.666	0.68
	RSD (%)	--	3.4
	Relative error (%)	--	1.6
Trigon®		Expected	
In cell	c (mg L ⁻¹)	1.333	1.32
	RSD (%)	--	6.5
	Relative error (%)	--	1.3
In drop	c (mg L ⁻¹)	1.333	1.32
	RSD (%)	--	5.05
	Relative error (%)	--	1.07

Figure 1.- Cyclic voltammogram of a 60 ppm Ir(IV) solution in a 0.016 mol L⁻¹ H₂SO₄ media measured with a SPCE. Scan rate 100 mV s⁻¹.

Figure 2.- Differential pulse voltammogram of a 20 ppm Ir(IV) solution in a 0.016 mol L⁻¹ H₂SO₄ media measured with a SPCE.

Figure 3.- a) Peak current vs. Ir(IV) concentration plot, and b) voltammograms measured at increasing Ir(IV) concentration from 0.04 to 100 ppm in a 0.016 mol L⁻¹ H₂SO₄ media measured with a SPCE.

Figure 4.- Differential pulse voltammograms of Ir(IV) solutions from 0.04 to 10 ppm measured with SPCE, CNT-SPCE and CNF-SPCE in a 0.016 mol L⁻¹ H₂SO₄ media. Insets show the corresponding calibration plots.

Figure 1

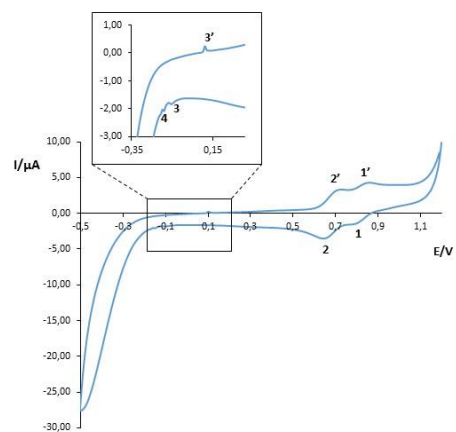


Figure 2

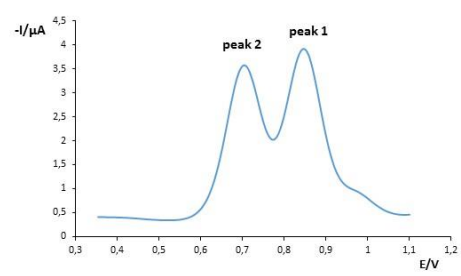


Figure 3

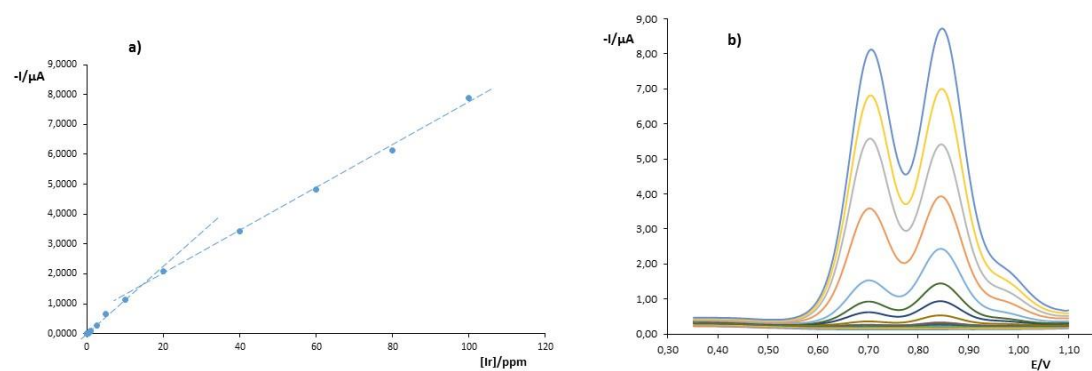


Figure 4

