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Corresponding Author: Dr Nuria Serrano,

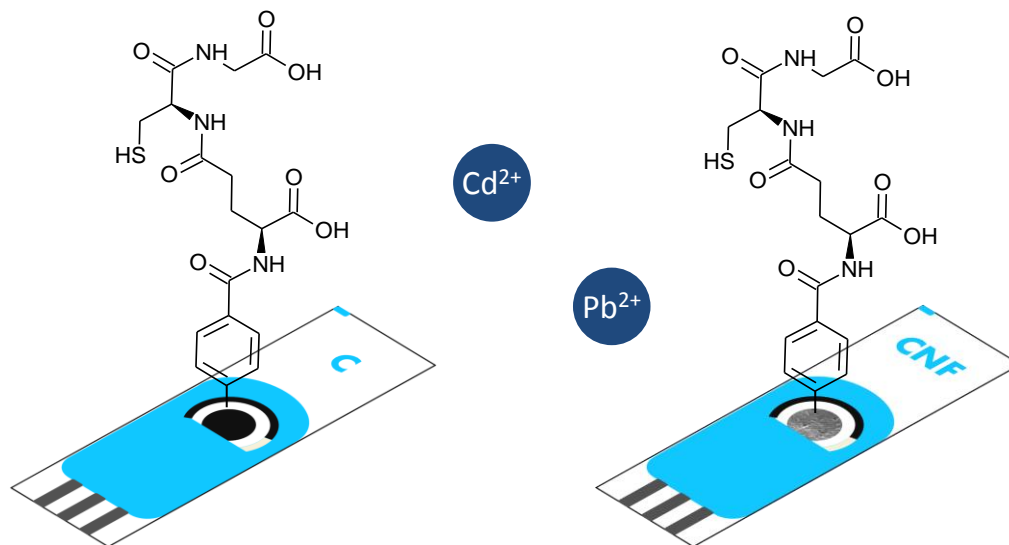
Corresponding Author's Institution: University of Barcelona

First Author: Clara Pérez-Ràfols

Order of Authors: Clara Pérez-Ràfols; Nuria Serrano; José Manuel Díaz-Cruz; Cristina Ariño; Miquel Esteban

Abstract: This work reports the development of a glutathione modified electrode via electrografting on a screen-printed carbon nanofiber substrate (GSH-SPCNFE). GSH-SPCNFE was compared to a classical screen-printed carbon electrode modified with glutathione (GSH-SPCE) for the simultaneous voltammetric determination of Cd(II) and Pb(II). Their electrochemical characterization and analytical performance suggest that SPCNFE could be a much better support for GSH immobilization. The applicability of GSH-SPCNFE for the determination of low concentration levels of Pb(II) and Cd(II) ions in environmental samples was successfully tested in a certified wastewater reference material by means of stripping voltammetry with a very high reproducibility and good trueness.

Graphical abstract



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Glutathione modified screen-printed carbon nanofiber electrode for the voltammetric determination of metal ions in natural samples

Clara Pérez-Ràfols, Núria Serrano*, José Manuel Díaz-Cruz, Cristina Ariño, Miquel Esteban

Departament de Química Analítica, Facultat de Química, Universitat de Barcelona,

Martí i Franquès 1-11, E-08028 Barcelona (Spain)

Phone: 0034934039274, fax: 0034934021233, e-mail: nuria.serrano@ub.edu

* to whom correspondence should be addressed

ABSTRACT

This work reports the development of a glutathione modified electrode via electrografting on a screen-printed carbon nanofiber substrate (GSH-SPCNFE). GSH-SPCNFE was compared to a classical screen-printed carbon electrode modified with glutathione (GSH-SPCE) for the simultaneous voltammetric determination of Cd(II) and Pb(II). Their electrochemical characterization and analytical performance suggest that SPCNFE could be a much better support for GSH immobilization. The applicability of GSH-SPCNFE for the determination of low concentration levels of Pb(II) and Cd(II) ions in environmental samples was successfully tested in a certified wastewater reference material by means of stripping voltammetry with a very high reproducibility and good trueness.

Keywords: peptide-modified sensors, electrochemical grafting, screen-printed electrodes, metal determination, stripping voltammetry, glutathione

1. INTRODUCTION

The great variety of natural and anthropogenic sources of heavy metals, as well as their inability to be degraded, results in concerning levels of contamination of the environment. This, coupled to these metals being a serious threat to human health, makes necessary to have proper methods for their determination [1]. In this sense, stripping techniques and, in particular anodic stripping voltammetry (ASV), are especially suitable for trace heavy metal ions determination in natural samples because they provide excellent detection limits and high reproducibility and sensitivity and they are capable of multielement determination with a relatively low cost [2]. Traditionally, stripping techniques were connected to the use of mercury-based working electrodes due to their high reproducibility and wide cathodic potential range [3]. However, concerns about mercury toxicity have led to focus on the development of more environmentally friendly working electrodes. In this area, chemically modified electrodes are of great interest. In the design of these electrodes, many different strategies for designing recognition interfaces can be followed. In recent years, though, monolayers strategies, dominated by alkanethiol self-assembled monolayers (SAMs), have become increasingly popular. However, alkanethiols SAMs present some limitations including a narrow potential range for metal ion determination as well as poor long term stability [4]. Another viable strategy for molecule immobilization that can overcome these limitations and has proven its usefulness in the development of metal ion sensors is based on aryl diazonium salt monolayers anchored on the electrode surface [4–6]. This strategy leads to stable electrodes that can be used for a wide number of measurements without signs of degradation [7–9] and allows to incorporate a wide range of functional groups to the electrode surface [10].

1 Peptides are known as effective, and often specific, ligands for metal ions. Their ability
2 to bind metals is a consequence of the great number of donor atoms not only in their
3 peptide backbone but also in their aminoacid side chains [11–13]. Particularly, the
4 complexation of heavy metals with thiol rich peptides like glutathione (GSH) or its
5 fragments has been largely studied by electroanalytical techniques (Table 2 in [13]).
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7 Thus, thiol rich peptides and related structures can be an interesting choice as
8 recognition molecule in metal ion sensors [7,8].
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12 The modification of electrodes with aryl diazonium salts has been applied to many sorts
13 of carbon surfaces including glassy carbon [7,14,15], graphite [8,9], diamond [16,17]
14 and screen-printed carbon electrodes [18–20] among others. Great advancements have
15 been achieved in the last years in the field of screen-printed electrodes (SPEs) allowing
16 the mass production of reproducible, economical and disposable devices. The whole
17 electrode system, including reference, auxiliary and working electrodes, is usually
18 printed on the same strip. Other important features of these SPEs are related to its
19 miniaturized size and their capability to be connected to portable instrumentation, which
20 makes them suitable for on-site analysis. Furthermore, SPEs offer the possibility to use
21 many different compositions of printing inks that can strongly influence their
22 performance [21,22]. This versatility in printing inks composition also allows the
23 incorporation of nanomaterials like carbon nanotubes or carbon nanofibers to the SPEs,
24 which are ideal for sensor application since they are conductive, easily functionalized
25 and possess very large surface areas. In particular, carbon nanofibers are unique in the
26 fact that their whole surface area can be activated [23]; recent studies have shown that
27 they can be a great choice of substrate in sensors design, enabling to achieve better
28 analytical performances [24,25].
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In this paper, all the above-mentioned benefits have been considered for the first time in the development of a glutathione modified electrode via electrografting on a screen-printed nanomaterial substrate leading to a glutathione modified screen-printed carbon nanofiber electrode (GSH-SPCNFE) which will be compared to a classical screen-printed carbon electrode modified with glutathione (GSH-SPCE) in terms of their electrochemical characterization and their analytical performance in the simultaneous determination of Cd(II) and Pb(II) as a model metal ion system. Taking advantage of the better analytical performance of the GSH-SPCNFE, its applicability has been tested through the simultaneous determination of Pb(II) and Cd(II) ions in a certified wastewater reference material.

2. EXPERIMENTAL

2.1 Chemicals

2-(N-morpholino)-ethanesulfonic acid (MES), potassium ferrocyanide $K_4[Fe(CN)_6] \cdot 3H_2O$, hydrochloride acid, sodium acetate, acetic acid and glutathione (GSH), in the reduced form, with purity greater than 99% were purchased from Merck (Darmstadt, Germany). *N*-hydroxysulfosuccinimide (sulfo-NHS), *N*-(3-dimethylaminopropyl)-*N*'-ethylcarbodiimide hydrochloride (EDC), 4-aminobenzoic acid (ABA), perchloric acid, potassium dihydrogen phosphate, sodium nitrite, sodium monophosphate, methanol and certified reference material, wastewater ERM[®]-CA713, were supplied by Sigma-Aldrich (St. Louis, MO, USA). Potassium ferricyanide $K_3[Fe(CN)_6]$ was provided by Panreac (Barcelona, Spain). All reagents were of analytical grade. Pb(II) and Cd(II) stock solutions 10^{-2} mol L⁻¹ were prepared from

1 Pb(NO₃)₂·4H₂O and Cd(NO₃)₂·4H₂O respectively and standardized complexometrically
2 [26]. Ultrapure water (Milli-Q plus 185 system, Millipore) was used in all experiments.
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5 6 7 **2.2 Apparatus**

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10 An Autolab System PGSTAT12 (EcoChemie, The Netherlands) attached to a Metrohm
11 663 VA Stand (Metrohm, Switzerland) and a personal computer with GPES version 4.9
12 data acquisition software (EcoChemie) was used for stripping measurements.
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17 Ag|AgCl|KCl (3 mol L⁻¹) and Pt wire (Metrohm, Switzerland) were used as reference
18 and auxiliary electrode respectively. The working electrode was a modified glutathione
19 electrode prepared from carbon screen-printed electrode (GSH-SPCE) or carbon
20 nanofibers modified screen-printed electrode (GSH-SPCNFE), both with 4 mm
21 diameter and provided by Dropsens (Oviedo, Spain) (ref. 110, DS SPCE and 110CNF,
22 DS SPCE respectively).
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27 Screen-printed electrodes were connected to the Autolab System by means of a flexible
28 cable (ref. CAC, DropSens).
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33 A Crison micro pH 2000 pH-meter was used for pH measurements.
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38 All measurements were carried out in a glass cell at room temperature (20 °C) without
39 oxygen removal.
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42 43 44 45 46 47 **2.3 Procedures**

48 49 50 **2.3.1 Preparation of modified SPEs**

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53 Glutathione modified SPEs were prepared based on a two-step procedure previously
54 described [7] with slight modifications.
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2.3.1.1 Diazonium salt electrografting

Aryl diazonium salt was generated *in situ* by adding 2 mmol L⁻¹ of sodium nitrite to a cooled solution of 73 mmol L⁻¹ of ABA in 1 mol L⁻¹ aqueous HCl. The resultant solution was allowed to stir in an ice bath for about 30 min before the electrochemical grafting process [27] was conducted. For this purpose, the SPE was immersed in 20 mL of the diazonium salt solution and 15 cyclic voltammetry (CV) cycles from 0 V to -1 V at 0.2 V s⁻¹ were applied. In order to remove any physisorbed compounds the functionalized electrodes were thoroughly rinsed with Mili-Q water and methanol.

2.3.1.2 Covalent immobilization of glutathione via carbodiimide coupling

10 µL of a 26 mmol L⁻¹ EDC and 35 mmol L⁻¹ sulfo-NHS solution in 100 mmol L⁻¹ MES buffer (pH 4.5) were dropped onto the functionalized SPE and left to incubate for 1 h to activate the carboxyl groups of the electrografted diazonium salt. The activated carboxyl groups reacted overnight at 4 °C with the amine terminal groups of glutathione by placing 10 µL of a 2.9 mg/100 mL glutathione solution in 0.1 mol L⁻¹ MES buffer (pH 4.5).

2.3.2 Voltammetric measurements

In stripping voltammetric measurements of Cd(II) and Pb(II) using a GSH-SPE, target metal ions were deposited for a deposition time (t_d) of 120 s at a deposition potential (E_d) of -1.4 V, applied with stirring. Deposition was followed by a rest period (t_r) of 5 s and determinations were done by scanning the potential from -1.4 to -0.45 V using pulse times of 50 ms, step potentials of 5 mV and pulse amplitudes of 50 mV.

Linear calibration plots were obtained by increasing metal ion concentrations in 0.1 mol L⁻¹ acetate buffer (pH 4.5) solution.

1 In the analysis of the certified wastewater sample, a volume of the sample in 0.1 mol L⁻¹
2 acetate buffer (pH 4.5) was placed in the cell and the scan was recorded (dilution factor
3 10/17). Calibration was performed by the standard addition method, five aliquots of
4 metal standard solutions were further added and the respective curves were recorded.
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9 In both linear calibration plots and analysis of the certified wastewater sample, a
10 cleaning step was performed before each set of measurement by applying a conditioning
11 potential (E_{cond}) of -0.3 V for 15 s in 0.1 mol L⁻¹ HClO₄.
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19 **3. RESULTS AND DISCUSSION**

20 **3.1 Electrochemical characterization**

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23 Electrochemical characterization of both GSH-SPCE and GSH-SPCNFE was performed
24 at each functionalization step by CV using ferrocyanide / ferricyanide as redox probe in
25 100 mmol L⁻¹ phosphate buffer (pH 7.4). The potential was scanned between -0.6 V and
26 0.6 V at 100 mV s⁻¹ without stirring. As expected, bare CNF resulted in more intensive
27 peaks than bare SPCE (Figure 1), which can be attributed to its enhanced surface area.
28 In both electrodes, a current decrease could be observed after electrografting (Figure 1)
29 due to the formation of a blocking layer [28]. There is not a clear agreement about the
30 number of CV cycles that should be performed during the electrografting process. In
31 fact, values ranging from 1 to 200 cycles can be found in the literature [29,30]. In this
32 paper, the optimum number of CV cycles to be applied was investigated. It was proved
33 that once the peak corresponding to the reduction of the diazonium salt disappears,
34 increasing the number of cycles does not improve the electrode performance whereas it
35 prolongs the functionalization process. For this reason, 15 cycles were selected as the
36 optimum value. For both GSH-SPCE and GSH-SPCNFE, covalent binding of
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glutathione resulted in intermediate currents (Figure 1). These modified electrodes can be stored for some weeks without signs of degradation.

3.2 Repeatability and reproducibility

Several electrochemical parameters, such as E_d , t_d and pH, were firstly optimized to ensure the detection of both Cd(II) and Pb(II) ions at each GSH-modified electrodes in the selected concentration range. DPASV measurements of a solution containing $50 \mu\text{g L}^{-1}$ of Pb(II) and Cd(II) were carried out at different E_d and t_d values ranging from -1.2 V to -1.5 V and from 30 s to 300 s respectively, looking for a compromise between peak area and analysis time. Two different media, acetate buffer (pH 4.5) and HCl (pH 2), were also tested. Well-defined peaks were obtained at pH 4.5 whereas the electrode was damaged for measurements at pH 2. In all cases, the compromise conditions were an E_d of -1.4 V applied with stirring for 120 s at pH 4.5 (acetate buffer).

Figure 2 shows a comparison of the analytical performance of the GSH-SPCE (thin line) and GSH-SPCNFE (thick line) for the simultaneous determination of a solution containing $80 \mu\text{g L}^{-1}$ Cd(II) and Pb(II). Well-defined peaks can be observed for both Cd(II) and Pb(II) ions with all electrodes. However, more intense peaks are obtained using the GSH-SPCNFE, particularly in the case of Cd(II). This fact can be attributed to CNF substrate being able to immobilize a higher amount of ligand due to its enhanced surface area [24].

Repeatability and reproducibility of both GSH-SPCE and GSH-SPCNFE were tested by measuring a solution of $80 \mu\text{g L}^{-1}$ Cd(II) and Pb(II) in acetate buffer (pH 4.5). Repeatability was calculated from 5 repetitive measurements using the same glutathione-modified electrode whereas reproducibility was calculated from two

1 different glutathione-modified units within a series of 5 repetitive measurements. In the
2 case of repeatability GSH-SPCNFE yielded an RSD % of 5.1% and 7.6 % for Cd(II)
3 and Pb(II) respectively whereas GSH-SPCE yielded an RSD% of 8.2 % for Cd(II) and
4 8.3 % for Pb(II). Good values of reproducibility were also obtained for GSH-SPCNFE
5 (6.8 % and 8.9 % for Cd(II) and Pb(II) respectively). However, reproducibility
6 calculated for GSH-SPCE yielded more than 20 % for both Cd(II) and Pb(II). This poor
7 reproducibility is probably associated with the modification procedure, particularly with
8 the covalent immobilization of GSH step. In the case of GSH-SPCNFE, whenever 10
9 μL of EDC/sulfo-NHS or GSH solutions are dropped onto the electrode, a persistent
10 well-formed drop can be observed. GSH-SPCE, however, is not able to sustain the drop
11 and the 10 μL of solution spread out of the working electrode surface without
12 guaranteeing that the working electrode is always covered by the same amount of
13 ligand, resulting in less reproducible devices.

14 Repeatability and reproducibility values achieved for GSH-SPCNFE are of the same
15 order of those reported for glassy carbon modified with penicillamine [7] or epoxy
16 graphite modified with crown ethers [9], both following the same modification strategy.

3.3 Calibration data

17 First of all, the influence of the presence of Pb(II) in the determination of Cd(II) and
18 vice versa was studied. In this sense, individual calibrations of Cd(II) and Pb(II) ions
19 ranging from 1.2 to 150.1 $\mu\text{g L}^{-1}$ and calibrations of considered metal ions in presence
20 of 100 $\mu\text{g L}^{-1}$ of either Cd(II) or Pb(II) by stripping voltammetry were carried out on
21 GSH-SPCNFE. Sensitivities (a.u. / $\mu\text{g L}^{-1}$) obtained as the slope value of the calibration
22 curves of Cd(II) and Pb(II) with and without the presence of the other metal ion were

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0.67 and 0.63 for Cd(II), and 1.05 and 3.02 for Pb(II), respectively. At the view of these results, it can be concluded that the presence of Pb(II) does not affect the determination of Cd(II), whereas the presence of high concentrations of Cd(II) slightly influence in the sensitivity (slope) towards Pb(II).

Sensitivity, limit of detection (LOD) and limit of quantification (LOQ) for the separate determination of Pb(II) and Cd(II) obtained by stripping voltammetry on GSH-SPCNFE were compared with those achieved on GSH-SPCE (Table 1). For this reason, linear calibration curves were carried out by measuring at the optimized conditions nine increasing concentrations of Pb(II) and Cd(II) ranging from 1.2 to 150.1 $\mu\text{g L}^{-1}$ in GSH-SPCNFE and from 1.1 to 153.0 $\mu\text{g L}^{-1}$ in GSH-SPCE. LOD and LOQ were calculated as 3 and 10 times the standard deviation of the intercept over the slope of the calibration curve respectively. As shown in Table 1, good linear responses of peak area *versus* metal ion concentration were obtained for both Pb(II) and Cd(II) with GSH-SPCE and a slightly higher sensitivity was observed for Cd(II) than for Pb(II). However, although LODs achieved for both Cd(II) and Pb(II) were at levels of $\mu\text{g L}^{-1}$, the LOD obtained for Pb(II) was much better than that obtained for Cd(II). In fact, in comparison with other peptide-modified electrodes reported in the literature [8,31], the LOD obtained for Pb(II) in this work is similar or even lower whereas Cd(II) LOD is quite higher. These results were remarkably improved when GSH was immobilized onto a carbon nanofiber substrate. As it can be seen in Table 1, although sensitivity for Cd(II) decreased in GSH-SPCNFE, a much lower LOD was achieved, resulting therefore in a wider linear range. The LOD of 3.2 $\mu\text{g L}^{-1}$ obtained now for Cd(II) is of the same order and in some cases better than those reported with other peptide modified electrodes [8,31] as well as with other similar electrodes modified following the same strategy like penicillamine-modified glassy carbon electrode [7] or crown ether-modified epoxy-graphite electrodes

1 [9]. It should be pointed out though, that in many of these reported LODs an unpractical
2 deposition time of 300 s or even 600 s instead of 120 s was applied. Thus, GSH-
3 SPCNFE can provide similar or even better LODs with faster determinations.
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5 Regarding to Pb(II), LOD was also slightly improved with GSH-SPCNFE, which also
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7 provided a much higher sensitivity.
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11 These improvements observed for GSH-SPCNFE could be attributed to the fact that,
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13 unlike SPCE, the whole surface area of carbon nanofibers can be activated and,
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15 therefore, a higher amount of ligand can be immobilized on the surface of the electrode
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17 [24].
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22 Therefore, the reported calibration data, coupled with the better reproducibility
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24 previously observed, suggest that SPCNFE could be a much better platform for peptide
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26 immobilization in general, and particularly for GSH, than the classical SPCE.
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28 Furthermore, GSH-SPCNFE could be fully suitable and a valuable alternative to more
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30 conventional electrodes for the simultaneous determination of Cd(II) and Pb(II) at trace
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32 levels in natural samples. It must be emphasized that unlike glassy carbon or epoxy-
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34 graphite electrodes, SPEs do not require any polishing prior to GSH immobilization. On
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36 the other hand, the durability of the GSH immobilization on every screen-printed
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38 platform for a large set of measurements (more than 20) without loss of sensitivity
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40 enables the voltammetric determination of metal ions with the same GSH-SPE unit.
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47 **3.4 Application to the analysis of a wastewater reference material**

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51 At the view of the reproducibility and the analytical performance of both GSH modified
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53 screen-printed electrodes, GSH-SPCNFE was considered for the simultaneous
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55 determination of Cd(II) and Pb(II) in natural samples and its applicability was tested by
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57 measuring a wastewater certified reference material (ERM[®]-CA713). It should be
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1 mentioned though, that the Cd(II) concentration in this available reference material was
2 below the quantification limit. For this reason, only Pb(II) was determined in the
3 presence of Cd(II).
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7 Pb(II) determination was performed by means of the standard addition method. DPASV
8 measurements were carried out under the above optimized conditions, including five
9 metal additions. Representative stripping voltammograms acquired in the analysis of the
10 wastewater reference material using GSH-SPCNFE are shown in Figure 3a. Well-
11 defined peaks that behave equally to the calibration data were obtained and, as it is
12 shown in Figure 3b, good correlation of the representative DPASV measurements
13 performed with GSH-SPCNFE was also observed for Pb(II).
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25 Three replicates of the voltammetric determination of the certified wastewater reference
26 material using GSH-SPCNFE were performed. The obtained metal concentration data
27 are reported in Table 2. Good concordance between all the replicates, as well as with the
28 certified Pb(II) ion value was obtained. It should be pointed out that the selected
29 reference material contains other metals like Cr, Cu, Fe, Mn and Ni at similar or higher
30 concentrations than Pb(II) that do not seem to interfere with the determination of Pb(II).
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41 These good results confirm the applicability of GSH-SPCNFE for the determination of
42 Pb(II) in the presence of Cd(II) in natural samples being, therefore, a valuable
43 alternative to the most conventional electrodes with the additional advantage that the
44 SPCNFEs which are the basis of the GSH-SPCNFE are commercially available and do
45 not need any polishing prior to the immobilization of the peptide.
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57 **4. Conclusions**

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1 This work reports the development of a glutathione modified screen-printed sensor in
2 which the peptide was successfully immobilized through aryl diazonium
3 electrochemical grafting onto a nanomaterial substrate electrode based on carbon
4 nanofiber (GSH-SPCNFE). The repeatability and reproducibility of GSH-SPCNFE
5 compared to a classical screen-printed carbon electrode modified with glutathione
6 (GSH-SPCE) as well as its analytical performance were much better due to both the
7 enhanced surface area attributed to CNF substrate and the modification process itself.
8 Furthermore, the modified GSH-SPCNFE can be used for a large set of measurements
9 without signs of degradation or loss of sensitivity requiring only the performance of a
10 simple cleaning stage between each set of measurements. The LODs and LOQs
11 achieved for Pb(II) and Cd(II) determination with a GSH-SPCNFE are similar or even
12 better than those reported with other peptides or chelating agents applying a t_d of 120 s
13 instead of the 300 s [8, 9] or even 600 s [31] used in earlier studies. On the other hand,
14 the basis of the GSH-SPCNFE is a commercial screen-printed support modified with
15 carbon nanofibers, it means that the developed sensor has the additional advantages of
16 the screen-printed electrodes, that is their accessibility, low-cost character and
17 suitability for on-site analysis. Moreover, unlike glassy carbon or epoxy-graphite
18 electrodes, SPEs do not require any polishing prior to peptide immobilization. Finally,
19 the applicability of the developed GSH-SPCNFE was successfully verified by the
20 determination of Pb(II) concentration in a certified wastewater reference material with
21 very high trueness and good reproducibility inferred by the relative error (0.8 %) and
22 the relative standard deviation (1.2 %). With respect to Cd(II), it was not possible to
23 quantify it since its concentration in the available certified reference material was below
24 the obtained LOQ. Nevertheless, at the sight of the results it can be concluded that
25 GSH-SPCNFE is an attractive alternative to the more conventional glutathione modified
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1 electrodes for Cd(II) and Pb(II) determination at low $\mu\text{g L}^{-1}$ levels in environmental
2 samples. Therefore, the use of SPEs modified with CNF is postulated as a promising
3 support for the immobilization of other chelating agents via electrografting for metal ion
4 determination. Finally, it should be taken into consideration that the developed electrode
5 can be also used for metal determination in combination with others forming a multi-
6 sensor array in search for a multivariate response.
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10 11 12 13 14 15 16 17 **Acknowledgments**

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Table 1 Calibration data for separate determination of Pb(II) and Cd(II) on GSH-SPCE and GSH-SPCNFE at E_d of -1.4 V, t_d of 120 s and pH 4.5

	GSH-SPCNFE		GSH-SPCE	
	Pb(II)	Cd(II)	Pb(II)	Cd(II)
Sensitivity (a.u. μg^{-1} L)	3.02 (0.04)	0.63 (0.01)	1.07 (0.02)	1.22 (0.05)
R^2	0.999	0.999	0.998	0.997
Linear range ($\mu\text{g L}^{-1}$) ^a	10.1-150.1	10.8-150.1	11.7-153.0	43.8-153.0
LOD ($\mu\text{g L}^{-1}$)	3.0	3.2	3.5	13.2

^aThe lowest value of the linear range was considered from the LOQ

Table 2 Total concentrations of Pb(II) and Cd(II) determined in certified wastewater (ERM[®]- CA713) by stripping voltammetry on GSH-SPCNFE by standard addition calibration method applying an E_d of -1.4 V and t_d of 120 s at pH 4.5

	Lead			Cadmium	
	c ($\mu\text{g L}^{-1}$)	RSD (%)	Relative error (%)	c ($\mu\text{g L}^{-1}$)	RSD (%)
GSH-SPCNFE	50.1	1.2	0.8	n.q.	—
Certified metal value	49.7	3.4	—	5.09	3.9

n=3 for RSD (%); n.q. not quantified.

Caption to figures

Figure 1. CVs plots recorded at each functionalization step for GSH-SPCE and GSH-SPCNFE. Measurements were performed in a 2 mmol L⁻¹ ferrocyanide/ferricyanide solution in phosphate buffer.

Figure 2. DPASV measurements of 80 µg L⁻¹ Pb(II) and Cd(II) on GSH-SPCNFE (thick line) and GSH-SPCE (thin line), applying a E_d of -1.4 V during 120 s at 0.1 mol L⁻¹ acetate buffer (pH 4.5).

Figure 3. a) Stripping voltammetric measurements in a wastewater sample by using a GSH-SPCNFE at pH 4.5 with E_d of -1.4 V during a t_d of 120 s and t_r of 5 s; and, b) Pb(II) standard addition plot.

Figure 1

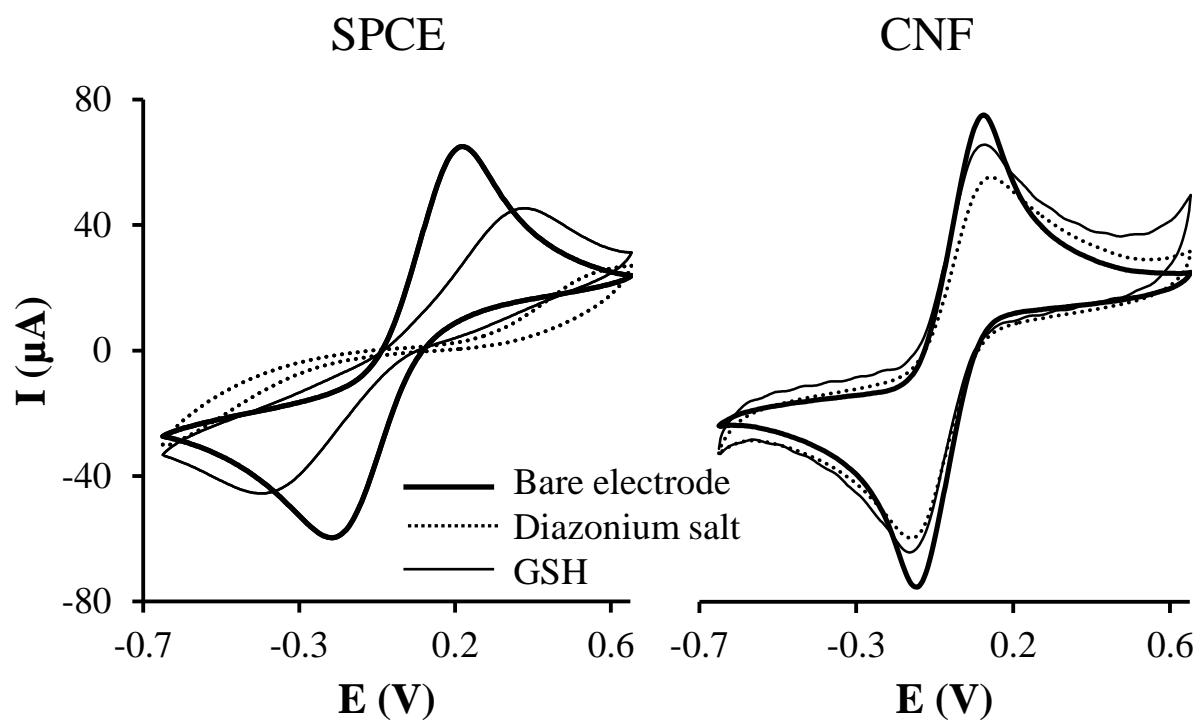


Figure 2

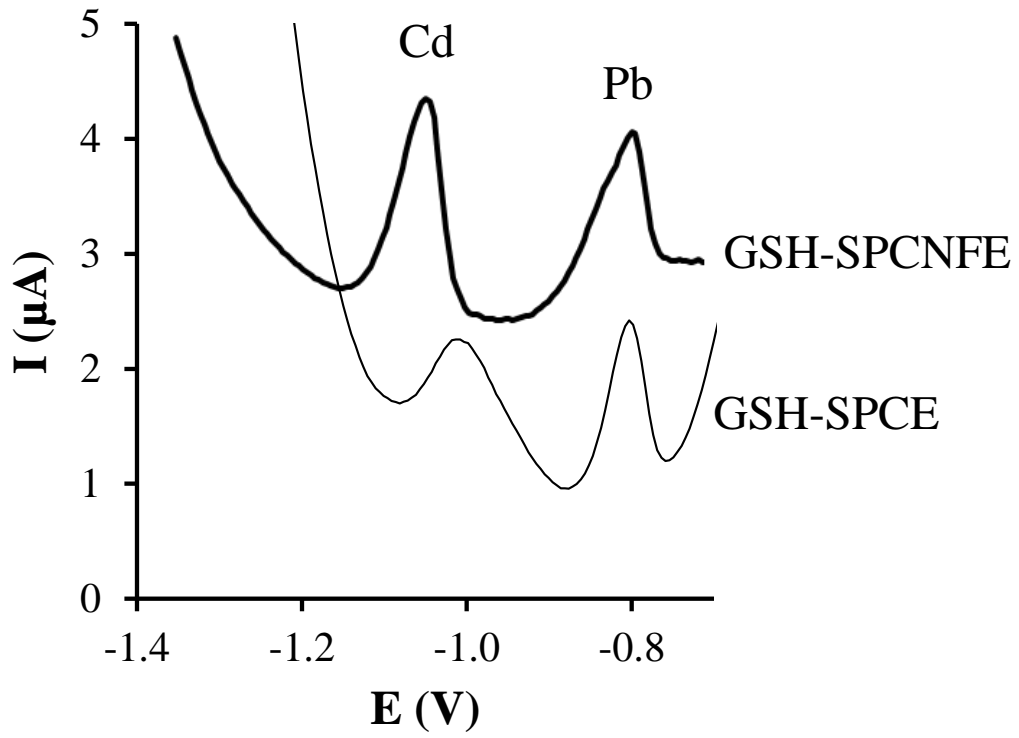


Figure 3

