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Chitosan Modified Screen-Printed Carbon Electrode for Sensitive Analysis of Heavy Metals

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The chitosan-modified screen-printed carbon electrode was used as a working electrode for the determination of selected metals: Pb, Cu, Cd and Hg. In 0.1 M HCl/KCl medium. The chlorocomplexes of these elements form ion-associates with the protonated amino group in the chitosan molecule at the electrode surface, then the resulting accumulated metals were stripped off using differential pulse anodic stripping voltammetry. The experimental conditions were optimized with respect to homemade printing ink composition, chitosan content, preconcentration time and scan rate. With 30 s preconcentration time, limit of detection values were 3.4, 5, 5 and 2 ng mL^{-1}, for Pb, Cu, Cd and Hg, respectively. The relative standard deviation for measuring Pb^{2+} was 4.1 % (n = 5) using the same electrode and 7 % (n=15) with different electrodes in the same batch. The screen-printed electrodes exhibit adequate shelflife (6 months**)**. There was no interference from Cd, Tl, Zn, Cu, Bi, Sb, Fe, Co, Au, Al, As, Pt, and Mn in Pb^{2+} determination while Sn showed significant interference. The developed screen-printed carbon electrodes modified with chitosan were successfully employed for the determination of Pb^{2+} in spiked tape water sample, holding a great promise for decentralized testing of heavy metals**.**

Keywords: Screen-printed carbon electrode; Chitosan; Stripping voltammetry; Heavy metals

1. INTRODUCTION

Heavy metals are important environmental pollutants, second only to pesticides in terms of environmental impact. They are a threat to the environment and to human health, because they are not biodegradable as they are retained indefinitely in the ecological systems and in the food chain; therefore, monitoring of heavy metals at trace levels is usually an increasingly important issue [1]. Many techniques are employed for heavy metal determination such as atomic absorption spectrometry, atomic emission spectrometry with inductively coupled plasma excitation, electroanalytical methods, X-ray fluorescence, neutron activation analysis, etc. Electroanalytical methods, particularly stripping analysis, are the most widely used competition for atomic spectroscopy, as far as trace metal determination are concerned. The remarkable sensitivity of stripping analysis is attributed to the unique coupling of effective preconcentration steps and advanced measurement procedures in which the metals are stripped away from the electrode during an appropriate potential scan. Such a combination offers convenient quantization of trace metals down to the sub-nanomolar concentration levels [2-5]. The acceptance of stripping methods has however suffered from the use of mercury working electrodes, which lack of market penetration compared to other techniques. The beauty of electrochemical techniques is to utilize a chemically modified electrode (CME) tailor made for sensitive and selective analytical applications [6-9] avoiding application of such toxic mercury element.

Chitosan, poly-[β-□1-4]-2-amino-2-deoxy-D-glucopyranose (CTS), is one of the most abundant natural polymers [10] that can form stable chelates with many transition metal ions through the hydroxyl and amino groups. CTS and its derivatives have been applied for electrochemical determination of silver, platinum, palladium, gold [11], lead, copper [12-14] and mercury [15] through coating of the pretreated GCE with thin CTS film. However, the coated film dissolved and leached from the electrode surface and therefore the durability of the electrode ranged between 10 days to one month. Nevertheless, the aforementioned method involves several manipulation steps for electrode fabrication including ultrasonication, drying and proper polishing of GCE itself. To overcome such difficulties and to simplify the procedure of electrode modification, the carbon paste electrode modified with CTS was exploited for determination of lead in tap water using differential pulse voltammetry [16]. However, designs and shapes of GCE and CPE are not suitable for every purpose as in the case of measurements in flowing streams or field monitoring with portable analyzers where the respective detection units require electrodes of particular constructions. Usually, smaller dimensions and planar configuration of electrodes are inevitable, which was documented on recent trends in the development and applications of screen-printed electrodes (SPEs) and can be proposed as a solution to the lack of commercial viability of electrochemical sensors.

There is a growing need for on-site tests in environmental analysis, which is related both to the elimination of errors associated with sampling, storage, and long transportation times to the main laboratory, and to the need for rapid monitoring of heavy metals for the characterization of contaminated sites. The coupling of disposable SPEs with stripping techniques is more favorable in comparison with conventional stripping analysis [17] as the design and operation are greatly simplified, in accordance with the requirements of a decentralized assay [8, 18-22]. Screen-printed carbon electrodes (SPCEs) were ideally used for the determination of various elements using different commercial printing inks [23-28]. The main disadvantage of commercial ink formulations is that their compositions are usually unknown as an intellectual property of the producer and some of the ink components may affect the electrochemical properties of the working electrode [29].

The present work aims to prepare homemade printing carbon ink consisting of carbon powder with PVC as binding material and to test the performance of such ink in the fabrication of simple CTS-SPCEs for the voltammetric determination of heavy metals.

2. EXPERIMENTAL PART

2.1. Reagents

All the reagents were of the analytical grade and bidistilled water was used throughout the experiments. Relative high molecular weight polyvinylchloride (PVC), graphite powder (synthetic 1–2 μm) and cellulose acetate (CA) were purchased from Fluka. Commercial carbon ink (Gwent Electronic Material Ltd, United Kingdom) was also used for printing of the electrodes. Low viscous chitosan (Fluka) was used as electrode modifier.

2.2. Apparatus

A Metrohm model 693 VA processor and 694 VA stand equipped with Ag/AgCl/3M KCl reference electrode and platinum counter electrodes were employed. The modified SPCEs described below were used as the working electrode for electrochemical measurements. The concentration of heavy metals solutions was measured with a Perkin-Elmer Model 372 atomic absorption spectrophotometer.

2.3. Sensors preparation

SPCEs (each 5×35 mm) were printed following the procedures described in details elsewhere [30] using homemade carbon ink prepared by mixing 0.75 g CTS, 5 g PVC solution (8 % in acetone-cyclohexanon 1:1) and 3 g carbon powder . The printed electrodes were cured at 50 ^{o}C for 30 min and covered with a layer of a PVC insulator, leaving a defined rectangular shaped (5×5 mm) working area and a similar area on the other side for electrical contact.

2.4. Procedure

The electrode surface was activated by five replicate direct current sweeps from –1000 to –400 mV with scan rate 50mVs^{-1} in 0.1M HCl/KCl solution. The solution was then exchanged by a sample solution containing the same supporting electrolyte purged by pure nitrogen gas for 2 min. Analytes were preconcentrated for 30 s at –1000 mV with stirring at 2000 rpm. After resting for 10 s, the differential pulse anodic stripping voltammetry (DPASV) was performed from –1000 to –400 mV with scan rate 50 mV/s, pulse amplitude 50 mV, pulse duration 20 ms and measurement time 10 ms. The experiment was triplicated without electrode regeneration and two standard additions of metals were put sequentially. The average peak heights were used for construction of the calibration curve.

3. RESULTS AND DISCUSSION

In our previous work, CPE modified with CTS was successfully applied in the voltammetric determination of lead in different samples. However, the prepared paste (composed of carbon powder and paraffin oil) was soft, non-compatible with the PVC substrate and had to be packed preferably into a special piston-like shaped electrode holder, which is not always available or suitable for decentralized application. Screen-printing seems to be the most suitable technique of choice for the low cost and fast mass production of small size, simple and disposable sensors. Commercially available carbon inks used for the printing of carbon working electrodes are usually composed of graphite particles, polymer binders and other proprietary additives for promotion of dispersion, printing and adhesion of the layer to the substrate [8, 31]. It is well known that ink composition as well as printing and curing processes greatly affect the electrochemical properties of the fabricated sensor [22], so the selection and use of particular ink is not a simple task as its exact composition may not be available. In addition, some of these printing inks need high curing temperature, which could destroy chemical modifiers added to the base ink and require special ceramic substrates.

The above-mentioned consideration led us to the preparation of homemade printing inks with known formula, enabling the deposition of the films containing modifier under mild conditions non destructive for ink components and support.

3.1. Principle of the method

The principle of the proposed method involves accumulation of MCl_4^{2-} at the CTS-SPCE where the metal ions are then reduced to the corresponding metals by application of sufficiently negative potential. In the next step, the metals are reoxidized back into the solution and the resulting stripping peak constitutes the analytical signal. Selective enhancement could be explained by the electrostatic attraction between the negatively charged metal chlorocomplexes and the protonated amino groups of CTS molecule.

3.2. Optimization of printing ink formulation

The first part of the present work was oriented to the optimization of printing ink composition containing carbon particle, suitable binder and modifier. The height of peaks current obtained for lead were used to compare the performance of the ink when varying its different chemical and physical parameters. Each parameter was analyzed three times with the same sensor and the corresponding average value of peak height was plotted.

The carbon ink for screen-printing purpose is usually composed of graphite particles and polymer matrix which solidify after curing process [9, 31], and the overall properties of the printed electrodes depend on the modifier and printing conditions. In the present work, different polymer binders were applied for the preparation of ink matrices such as cellulose acetate (CA), PVC, polyacrylic acid and epoxy resins dissolved in a proper organic solvent. Cellulose, which has a molecular structure similar to CTS, has the ability of chelating heavy metals, but the printed electrodes with CA as binder showed no voltammetric peak probably due to the strong binding between CA and metal cations. The electrode fabricated using polyacrylic acid or epoxy resins had relatively high background current which reduced the sensitivity of the method. In contrast, PVC is known to be chemically inert and the printed electrodes worked only in presence of the CTS. The concentration of the binder was changed from 0.5 to 14 %, and it was found that both thickness and ohmic resistance of the printed electrodes increased by increasing the PVC content. It was also noticed that PVC concentration more than 14% gave a very viscous ink while lower content rendered a poor printing quality; therefore, 8 % PVC solution was selected. The ink solvent should have suitable evaporation rate to achieve ink homogeneity during preparation and printing. Several relatively high boiling point solvents (e.g. cyclohexanone, cyclohexanone-acetone mixtures, or commercial thinner) were used as substitute for terahydrofuran and it was found that the mixture of cyclohexanone-acetone in 1:1 ratio was the most suitable solvent for the proper printing process and appropriate curing time.

The carbon content within the printing ink was varied from 15 to 80 % and the carbon ratio of 72.2 % was selected as the printed electrodes have a moderate resistance and good adhesion to the substrate. Despite the ohmic resistance of 1.5 k Ω, there was finally no need to print a conductive silver track underneath the carbon layer which may be subjected to oxidative corrosion and in addition it could be chemically or potentiometrically sensitive to various substances present in the analyzed solution. The proposed procedures will simplify the printing of the working electrodes to one-step process.

As the accumulation of metal ions chlorocomplexes at the electrode is driven by the interaction with CTS, its content within the electrode matrix was expected to have a significant influence on the voltammetric response. A study was carried out to establish the relationship between the CTS loading and DPASV current (Fig. 1). The maximum stripping current was obtained with CTS loading 0.75 g. Further loading of the CTS may be related to the more dense packing of CTS on the orientation of the complex molecule which then affects on the electron transfer reaction to the complex metal ion.

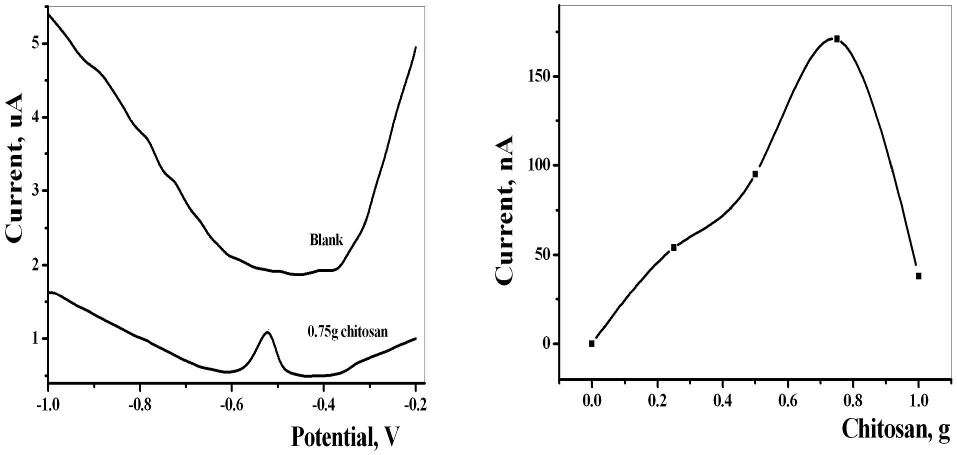

Figure 1. Effect of CTS on peak current of 50 ng mL^{-1} lead after 30 s accumulation and DPASV.

SPEs were also fabricated using commercial carbon ink (Gwent C2010517D4) modified with CTS (0.75 g) and applied for DPASV determination of Pb^{2+}. The obtained results showed higher base line than that of the homemade ink, which raise the detection limit of the method to 60 ng mL^{-1}. The final homemade ink composition was as follows: 18 % CTS, 72.2 % carbon and 9.8 % binder. The homemade ink showed shelf life similar to the commercial one with the advantages of easier preparation and modification.

3.3. Optimization of experimental conditions

To establish the optimum conditions for the determination of heavy metal by means of DPV technique, various instrumental variables were studied. The response of the CTS-SPCE was strongly dependent on the concentration of chloride necessitated for tetrachlorocomplexes formation and it was found experimentally that a mixture solution of 0.1 M HCl and 0.1 M KCl gave optimum peak current. The effect of preconcentration time on the peak current was also investigated. Figure 2 shows that the peak current of Pb increased with increasing preconcentration time within 120 s, which indicates that Pb on the modified electrode surface was rapidly adsorbed while further prolongation accumulation did not improve the peak height. A preconcentration time of 30 s was chosen as a compromise between length of analysis time and sensitivity.

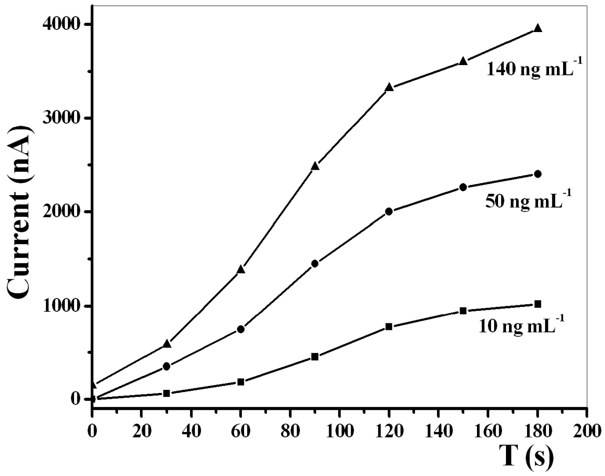

Figure 2. Effect of preconcentration time at different concentration of Pb.

Furthermore, the peak current increases with increasing the scan rate and remain stable between 30 - 60 mV s^{-1} (Fig. 3). In the same time, the peak potential becomes constant over the range of scan rate between 50 to 120 mV s^{-1} indicating the reversibility of the electrode reaction. Thus, scan rate of 50 mV/s was chosen as optimum parameter.

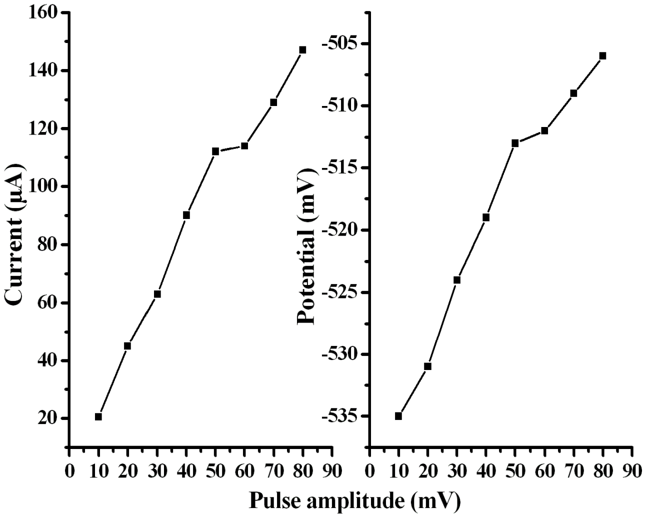

Figure 3. Effect of scan rate after 30 s accumulation time in 0.1M HCl/KCl solution.

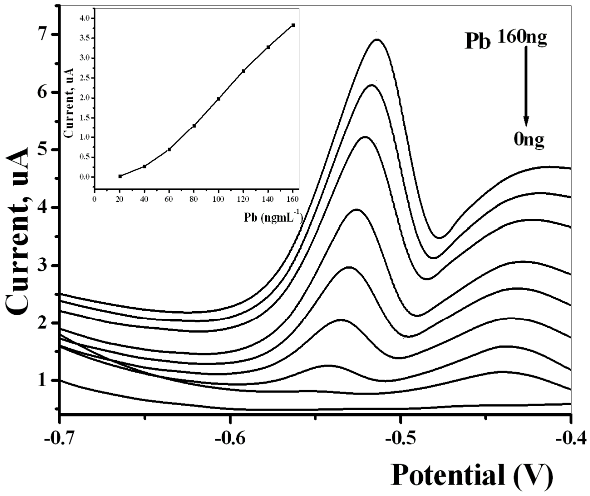

Figure 4. Calibration curve of Pb using DPASV based on CTS-SPCE and after 30 s accumulation time in 0.1 M HCl/KCl solution.

3.4. Calibration plot, limit of detection and precision

Under the preceding optimum conditions of detection, the anodic peak currents were proportional to Pb concentrations in the range of 20 – 160 ng mL^{-1} with regression coefficient of 0.9984 (Fig. 4). The detection limit was 3.4 ng mL^{-1} in terms of signal to noise ratio of 3:1 (s/n= 3). The repeatability was evaluated through the relative standard deviation (RSD) of five replicate measurements of 50 ng mL^{-1} Pb^{2+} and found to be 4.1%. It is noteworthy to mention that the same electrode can perform 15 successive measurements with RSD 6.5 %. The performance of CTS-SPCE fabricated in the same batch and in different batches was also tested for Pb determination. The obtained relative standard deviation reflects the high reproducibility of the printing process as the RSD values range between 5 – 7 % only. The useful lifetime of the printed electrodes was tested by performing day to day measurement of 50 ng mL^{-1} Pb. CTS-SPCE showed useful lifetime of more than 6 months during which the peak height did not change significantly (decreased by 6%).

CTS has the ability to chelate several metal cations [11-14]. In addition to Pb, it was found that Cu^{2+} and Cd^{2+} gave well-defined stripping peaks (Fig. 5) and can be determined simultaneously by standard addition method. The calibration graphs were linear in the range 10 – 200 ng mL^{-1} with limit of detection of 5 ng mL^{-1}.

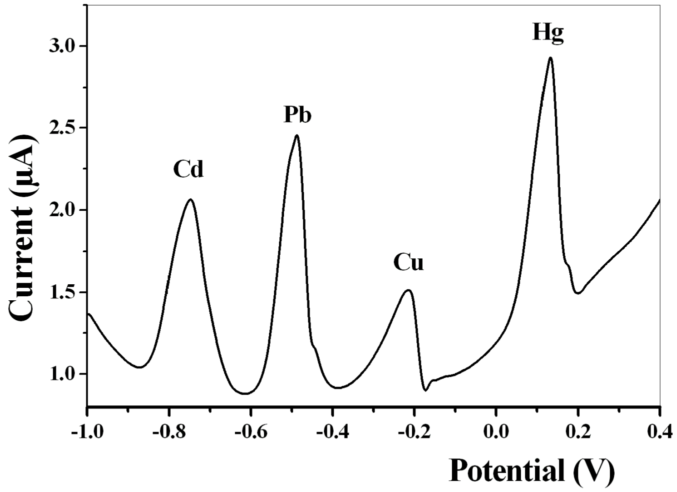

Figure 5. Simultaneous determination of Cd, Pb and Cu after 30 s accumulation time and DPASV in 0.1 M HCl/KCl solution.

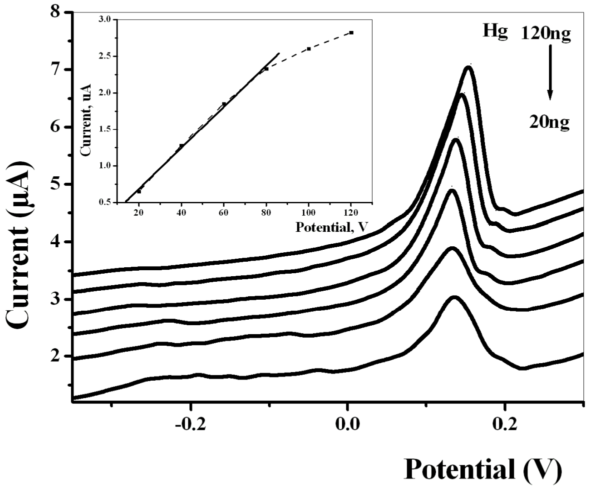

Figure 6. Calibration curve of Hg (II) at CTS-SPE using DPASV after 30 s accumulation time in 0.1 M HCl/KCl solution.

Rassaei et al [32] reported that the amine groups in the CTS biopolymer were used to bind effectively Hg(II) ions from an aqueous KCl solution. In the present work, the CTS-SPCE was successfully applied for the determination of the Hg(II) in the concentration range 20 – 80 ng mL^{-1} with regression coefficient 0.9983 (Fig. 6). The detection limit of Hg was 2 ng mL^{-1} in terms of signal to noise ratio of 3:1 (s/n = 3).

3.5. Interferences

The interfering effect of some cations on the determination of lead with CTS-SPCE was studied by addition the interfering ions to the supporting electrolyte containing 50 ng mL^{-1} Pb. The presence of 100 fold excess of Cd, Tl, Zn, Cu, Bi, Sb, Fe, Co, Au, Al, As, Pt, and Mn did not cause interference while the same concentration of Sn display noticeable interference.

3.6. Sample analysis

The proposed method was applied for the determinations of lead in tap water samples and the Pb content was found to be 34.4 ± 0.2 ng mL^{-1} (n = 5) which is in accordance with results obtained by atomic absorption spectroscopy (33.9 ± 0.5 ng mL^{-1}).

4. CONCLUSIONS

The experiments carried out in the present study showed that the SPCEs can be successfully applied in the voltammetric determination of heavy metals using the procedures adopted from the application of the ordinary glassy carbon electrode (GCE). However, SPCEs have several advantages, namely mass production, reproducibility, and ease of the preparation process. The bulk modification of the electrode was very simple compared with GCE, which needs several preparation and refreshment steps. Moreover, this sensor was able to detect simultaneously Pb, Cd and Cu with good sensitivity and reproducibility and could serve as a tool to aid environmental decision making.

These results may be the base for further research leading to improvement of the analytical parameters of prepared sensors and expanding the spectrum of analyzed heavy metals with commercialization of such electrodes for various voltammetric purposes. Further studies will be done for fabrication of three electrode strip containing the working electrode, pseudo silver/silver chloride reference electrode and auxiliary plain carbon electrode using a homemade PVC ink as well as a portable measuring system for the field measurement of heavy metals.

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