Original scientific paper

Benzimidazole-modified polyaniline micro-shells for electrochemical detection of cadmium in aqueous solution

Eldhose V. Varghese[1,2], Bejoy Thomas[1], Carsten Schwandt[3], Praveen C. Ramamurthy[4] and Alex Joseph[1,✉]

[1]*Department of Chemistry, Newman College, Thodupuzha 685585, India*

[2]*Department of Medicinal and Applied Chemistry, Kaohsiung Medical University, Kaohsiung 807, Taiwan*

[3]*Department of Materials Science and Metallurgy, University of Cambridge, Cambridge CB3 0FS, United Kingdom*

[4]*Department of Materials Engineering, Indian Institute of Science, Bangalore 560012, India*

Corresponding author: ✉*alex.chiramattel@gmail.com*

Abstract

Benzimidazole-functionalized polyaniline (BMPANI) was synthesized by interfacial poly-merization technique and used for electrochemical sensing of cadmium ions in an aqueous solution. The material was characterized for its structural and morphological features using Fourier-transform infrared spectroscopy (FTIR), X-ray diffractometry (XRD), and scanning electron microscopy (SEM). The BMPANI has a micro-shell structure produced from the self-assembly of the monomer units in solution before the polymerization reaction. The material was trialed for cadmium ion sensing using a BMPANI-modified carbon paste electrode (BMPANI-CPE). Electrochemical techniques, i.e., cyclic voltammetry (CV) and differential pulse anodic stripping voltammetry (DPASV), were performed to assess the sensing characteristics of the material. Various electrode preparation parameters, i.e., deposition potential, pH of deposition solution, and thickness of the active layer, were optimized to achieve the highest level of sensitivity. The selectivity towards cadmium ions, interference from other ions, as well as stability and reusability of the BMPANI-CPE, were also examined and found to be satisfactory.

Keywords

Polyaniline; electrochemical biosensor; cyclic voltammetry; differential pulse anodic stripping voltammetry; sensor stability; sensor reusability

Introduction

The Agency for Toxic Substances and Disease Registry (ATSDR) and Environmental Protection Agency (EPA) positioned cadmium at the top seventh in the latest hazardous substances priority list

(ATSDR-2022), with 1317 against arsenic with 1675 total points. Cadmium is detrimental and poses potential threats to human health due to its known and suspected toxicity, causing issues such as cancer and organ system toxicity. The latter regards the skeletal, respiratory, cardiovascular, urinary and reproductive systems, as well as the central and peripheral nervous systems [1]. For instance, it affects the mitochondrial respiratory chain, which plays an essential role in maintaining energy homeostasis through oxidative phosphorylation (OXPHOS), generating energy in the form of adenosine triphosphate (ATP) which is the energy mandate for life systems [2]. The maximum permissible concentration of Cd(II) ions in drinking water asserted by the World Health Organization (WHO) is 3 µg/L [3]. Several reliable techniques are used for the detection of Cd(II) ions, including atomic absorption spectrometry (AAS) [4], flame atomic absorption spectrometry (FAAS) [5,6], inductively coupled plasma mass spectrometry (ICP-MS) [7], inductively coupled plasma optical emission spectrometry (ICP-OES) [8], and fluorescence spectrometry (FS) [9]. The prerequisites for these methods are tedious and corrosive sample preparation protocols in addition to the requirement of highly experienced personnel and expensive instruments, all of which limit their use.

An alternative to the aforementioned methods is electrochemical sensors, employing measuring techniques such as cyclic voltammetry (CV), differential pulse anodic stripping voltammetry (DPASV) and chronoamperometry. DPASV is considered to be one of the most sensitive electrochemical techniques in electroanalysis. The electrodes used are usually based on carbon paste and modified with various active materials like polymers, inorganic compounds or composites. Conducting polymers are considered one of the best choices to modify electrodes because they can often be synthesized in nanostructured forms possessing large surface area, high porosity, fast electron mobility, and possibilities for surface modifications to tune the electrochemical properties [10,11]. The sensitivity can be enhanced further by chemical modification through covalently-bound chelating functional groups on the polymer backbone [12,13] or by physical modification mediated through Van der Waals forces. To mention specific examples, polyaniline with backbones modified with either imidazole [14] or iminodiacetic acid [15] was reported for heavy metal ion sensing with improved sensitivity compared to the unmodified form. In another study by Mustafin *et al.* [16], polyaniline modified with alkenyl side chains was used for resistive sensing of humidity. All said above shows that chemical modification is a preferred strategy to modify the transduction properties of polyaniline.

The present work reports an electrochemical Cd^{2+} ion sensor based on poly(2-(1H-benzimidazol--2-yl) aniline) with enhanced sensitivity. The polymer was synthesized by polymerizing 2-(1H-benzimidazol-2-yl) aniline monomer using the interfacial polymerization technique. The resultant polymer micro-shells (BMPANI) were then used to modify a carbon paste electrode (CPE), and this modified electrode was trialed for the quantitative determination of Cd^{2+} ions in aqueous media. The electrochemical techniques used to investigate the interaction of Cd^{2+} ions with the electrode surface were CV and DPASV. The potential of Cd deposition, pH of deposition solution, and thickness of the active layer were all optimized to ensure a high sensing efficacy. Selectivity and interferences, as well as stability and reusability of sensor BMPANI-CPE, were also investigated.

Experimental

Materials and methods

All chemicals and solvents used for the synthesis of poly(2-(1H-benzimidazol-2-yl) aniline) were of analytical grade and used without any further purification unless specified. Cadmium nitrate, lead nitrate, sodium acetate and acetic acid were obtained from Merck India Pvt. Ltd. Zinc nitrate, nickel nitrate and cobalt nitrate were acquired from Spectrochem India Ltd. Ammonium peroxodisulfate (APS),

sulfuric acid, hydrochloric acid, nitric acid and chloroform were obtained from Nice Chemicals Pvt. Ltd. India. Oxalic acid and phosphoric acid were acquired from SD Fine Chemicals Ltd. India. 2-(1H-benzimidazol-2-yl) aniline was purchased from AstaTech Inc. USA. Chloroform was double distilled over 4 Å molecular sieve and preserved under an inert atmosphere prior to use. Deionized (D.I.) water was used throughout.

The chemical structure of the synthesized polymer was analyzed by FTIR using a Thermo Nicolet iS5 spectrometer in the ATR mode with 64 scans in the range of 400 to 4000 cm^{-1}. The crystalline properties of the polymer were studied by XRD using a Rigaku Mini Flex 600 diffractometer in the 2θ range of 5 to 80 degrees. Its morphology was analyzed by SEM using a JEOL JSM-6490LA field emission scanning electron microscope. All electrochemical studies were performed with a Metrohm Autolab PGSTAT302N workstation. A three-electrode configuration was used, comprising an inhouse-prepared chemically modified CPE as the working electrode, a saturated calomel electrode (SCE) from CH Instruments Inc. as the reference electrode, and a platinum wire as the counter electrode.

Interfacial polymerization of 2-(1H-benzimidazol-2-yl) aniline (BMPANI)

The polymerization of 2-(1H-benzimidazol-2-yl) aniline was carried out by interfacial polymerization, following a procedure reported previously for the synthesis of poly(anthranilic acid) [17-19]. Briefly, two solutions consisting of (a) 0.43 g of ammonium peroxodisulfate in 100 mL 0.25 M H_2SO_4 and (b) 0.36 g of 2-(1H-benzimidazol-2-yl) aniline in 100 ml chloroform were prepared separately. Then, the former solution was added to the latter dropwise without mixing the solutions. The interface between the two solutions served as the site where the polymerization reaction was initiated, as thereafter indicated by the appearance of a reddish-brown coloration. The reaction was left for 24 h in order to ensure complete polymerization, and the polymer formed gradually filled up the entire aqueous phase. The black precipitate was filtered out, washed with D.I. water, and finally dried in a vacuum oven at 50 °C for 24 h. The yield was 0.124 g, corresponding to 36 %.

### Preparation of BMPANI-modified carbon paste electrode (BMPANI-CPE)

The carbon paste required for CPE was prepared by mixing graphite powder and paraffin oil in a 7:3 ratio in a ball mill for 72 h. The carbon paste thus prepared was then packed in a glass tube of 4 mm inner diameter and 60 mm length. A thin silver wire was inserted into the paste and served as the electrical contact. The exposed carbon paste was polished on plain printer paper to provide a smooth surface.

Uniform dispersion of the polymer in water was achieved by stirring 1 mg of poly (2-(1H-benzimidazol-2-yl) aniline) in 1 mL of D.I. water for 24 h followed by sonication for 20 min. The polymer-modified carbon paste electrode (BMPANI-CPE) was then made by drop casting the desired volume of this dispersion onto the CPE surface and subsequent drying in air.

Results and discussion

Characterization of the reaction product

A schematic representation of the interfacial polymerization reaction of 2-(1H-benzimidazol-2--yl) aniline is shown in Scheme 1. The polymerization occurs through the aniline moiety, forming a 1,4-polymerized polyaniline backbone that is substituted with benzimidazole at its ortho position (BMPANI). The purpose of the benzimidazole side chain is to serve as the metal-coordinating ligand in the envisioned sensing application.
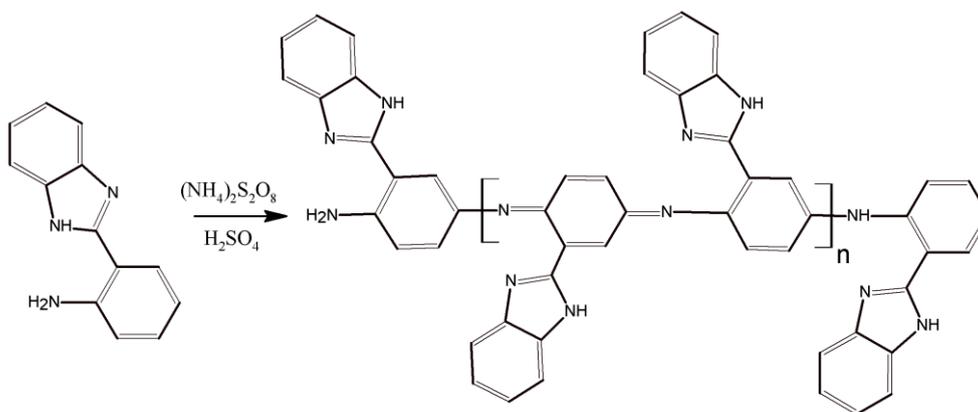
Scheme 1. *Illustration of the interfacial polymerization reaction for the production of 1,4-polymerized polyaniline*

The structures of BMPANI and PANI were determined using FTIR spectroscopy. Figure 1 shows the FTIR spectrum of the synthesized BMPANI along with that of PANI synthesized under similar conditions as reference material. In the case of PANI (curve A), the main peaks observed at 1502 and 1560 cm^{-1} correspond to the characteristic C=C stretching vibrations of the quinoid and benzenoid rings of polyaniline, respectively, and the peak at 1289 cm^{-1} is due to the typical C–N stretching vibration of the polyaniline backbone [20]. The band around 3230 cm^{-1} results from the N–H stretching vibration, and the weak band at 795 cm^{-1} is from the C–H out-of-plane bending vibration of the aromatic ring [20]. In the case of BMPANI (curve B), the spectrum shows the characteristic features of PANI, indicating that the BMPANI is based on the polyaniline backbone. The benzenoid and quinoid characteristic C=C stretching vibrations are now observed at 1506 and 1570 cm^{-1}, respectively, and the typical C–N stretching vibration of the polyaniline backbone now appears at 1302 cm^{-1}. The peak at 1670 cm^{-1} corresponds to the characteristic C=N stretching vibration of benzimidazole [21], and the peaks at 1459 and 1220 cm^{-1} are due to the C=C [22] and C–N [23] stretching vibrations, respectively, of benzimidazole. Altogether, these findings prove the presence of the benzimidazole entity as the side chain in the synthesized polymer.
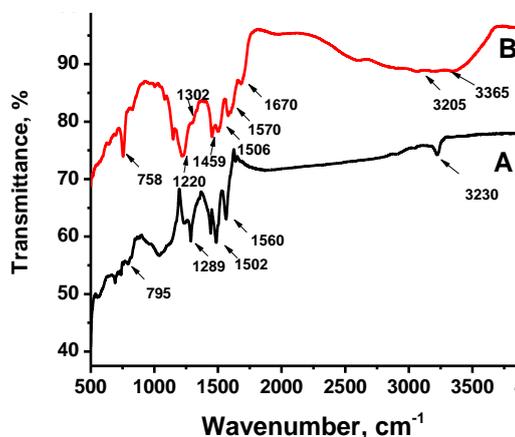

Figure 1. *Fourier-transform infrared spectra of: A – PANI and B – BMPANI*

The crystallinity of BMPANI and PANI was investigated by XRD, and the patterns for both are shown in Figure 2. Broad absorption bands, as opposed to sharp peaks, are observed for both materials, indicating their amorphous nature. In the case of PANI, these bands are centered around 2θ = 13.4, 19.5 and 26.2° and correspond to the (011), (020) and (200) crystal planes, respectively [24]. BMPANI exhibits these three bands equally, with the angles shifted slightly to 2θ = 13.8,

18.8 and 26.2°, corresponding to the above three planes. The shift in band positions is ascribed to the presence of the benzimidazole group, intervening sterically between the polyaniline backbone and altering the interlayer spacings.
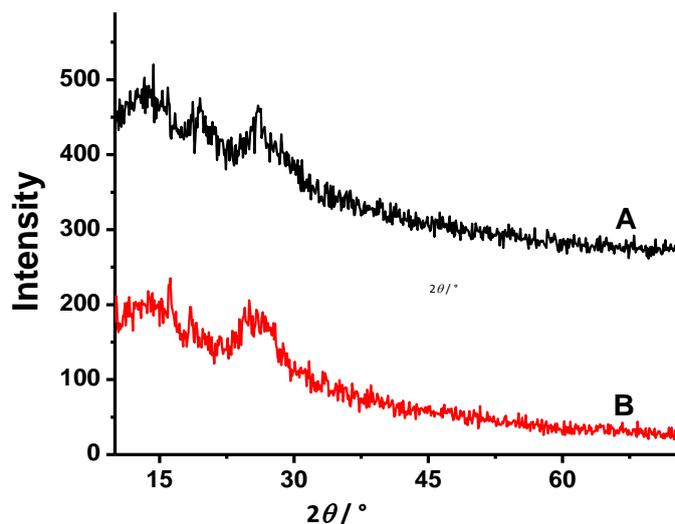

Figure 2. XRD patterns of: A – PANI and B – BMPANI

The measured diffraction angles and the calculated *d*-spacings of BMPANI and PANI are tabulated in Table 1, showing that BMPANI and PANI are similar in properties regarding their backbones and amorphous nature.

Table 1. *Angles of diffraction and corresponding d-spacings for BMPANI and PANI*

PANI	2θ / °	13.4	19.5	26.2
	d spacing, nm	0.66	0.46	0.34
BMPANI	2θ / °	13.8	18.8	26.2
	d spacing, nm	0.64	0.47	0.34

The morphology of BMPANI was analyzed by SEM, and the images recorded are shown in Figure 3. It is observed that BMPANI has a unique microstructure of spherical shells with hollow spaces inside. The formation of this distinct morphological feature is explained on the basis of the self-assembly of the monomer at the interface before polymerization in such a way that it forms hollow micelles that are then converted into the spherical shells of the resultant polymer. The average diameter of the shells is measured to be in the range of 1 to 5 μm.
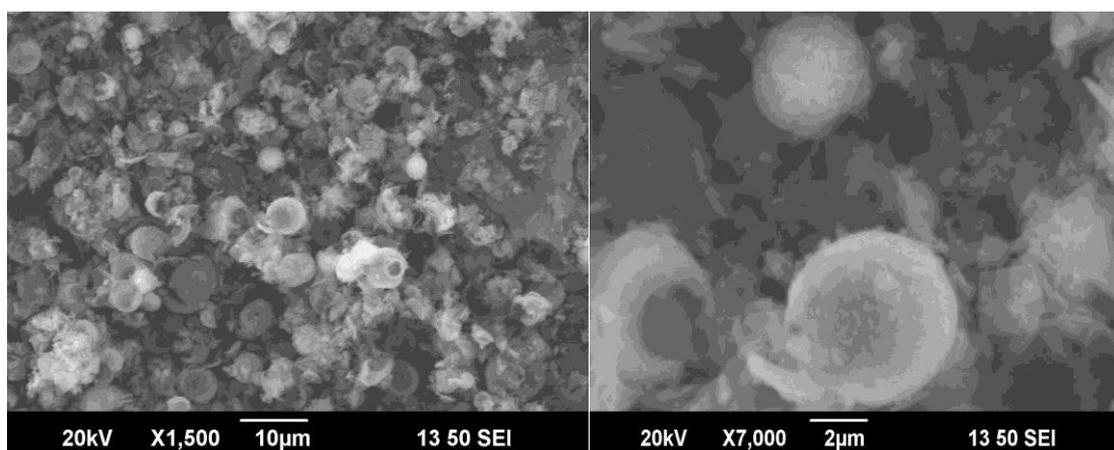

Figure 3. SEM images of BMPANI at two different magnifications

Electrochemical detection of Cd²⁺ with BMPANI-CPE

A series of electrochemical studies were performed in order to validate the efficacy of BMPANI-CPE for the detection of cadmium ions in aqueous media.

Cyclic voltammetric studies were carried out to understand the interaction of the BMPANI-CPE with Cd²⁺ ions. The CVs recorded for the BMPANI-CPE and, for comparison, the bare CPE, both in the absence and presence of Cd²⁺ ions, are shown in Figure 4. The experimental conditions were an aqueous acetate buffer of pH 5, a potential scan window from 0.5 to -1.8 V, and a scan rate of 50 mV/s. It is observed that, in the plain buffer solution, both the CPE and the BMPANI-CPE display no characteristic features. In contrast, in the presence of 10 mM Cd²⁺ ions in the buffer solution, both the CPE and the BMPANI-CPE exhibit a characteristic redox peak couple. The presence of two peaks in these voltammograms results from a reduction of Cd²⁺ ions to Cd metal in the cathodic direction and oxidation of Cd back to Cd²⁺ in the anodic direction. For the CPE, the reduction and oxidation peaks are observed at -1.21 and -0.97 V, respectively, and for the BMPANI-CPE, they are seen at -1.16 and -0.89 V, respectively.
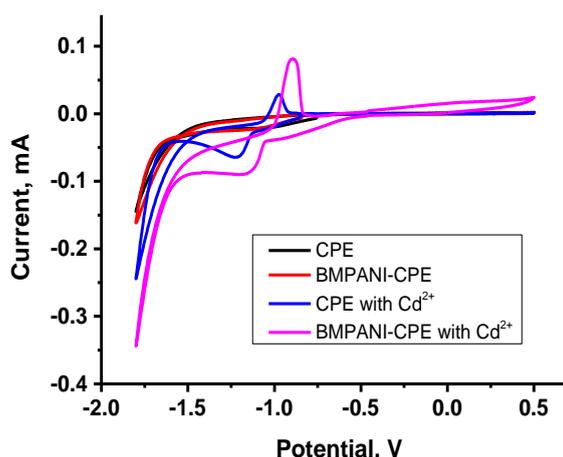

Figure 4. *Cyclic voltammograms showing the interaction of BMPANI-CPE and CPE with Cd²⁺ ions (Cd²⁺ ion concentration is 10 mM and potential is* vs. *SCE)*

It is further observed that the BMPANI-CPE has an improved signal intensity compared to the bare CPE. This indicates an enhanced interaction of the BMPANI-CPE with Cd²⁺ ions, which can readily be attributed to the benzimidazole functional group on the BMPANI chelating the ion. Overall, the CV results suggest that the BMPANI-CPE can sense cadmium ions in an aqueous solution.

Stripping voltammetric studies were carried out for the quantitative determination of Cd²⁺ ions in an acetate buffer solution. The DPASV analyses were done in two steps. In the first step, the pre-concentration of Cd²⁺ ions from the solution onto the electrode surface was induced by applying an appropriate cathodic deposition potential. In this step, Cd²⁺ is coordinated by the BMPANI functional group and then reduced to Cd on the CPE surface, as represented in Equation (1).

$$Cd^{2+} \rightarrow [Cd\text{-}BMPANI]^{2+} \rightarrow Cd_{sorbed} \tag{1}$$

In the second step, the stripping of the Cd analyte from the electrode surface back into the solution was effected by sweeping the potential in the anodic direction from −1.2 to 0 V. In this step, the reactions leading to the deposition are reversed, as represented in Equation (2).

$$Cd_{sorbed} \rightarrow Cd^{2+} \tag{2}$$

In each measurement, the anodic stripping current peak, due to the oxidation of the deposited Cd, was observed at around -1.0 V during the potential sweep.

In order to achieve maximum sensitivity of BMPANI-CPE for sensing Cd^{2+} ions, the DPASV electrode parameters were optimized by trial-and-error experiments. This included the potential of Cd deposition, pH of the deposition solution, and thickness of the polymer layer coated onto the electrode surface, all as summarized in Figures 5A to 5C. The Cd^{2+} ion concentration was kept at 1 µM in all cases.

First, the effect of the Cd deposition potential on the anodic stripping current was analyzed by varying the deposition potential from -1.4 to -0.8 V (Figure 5A). From the stripping currents in the figure, it is clear that the amount of Cd^{2+} ions deposited onto the electrode surface firstly increases on lowering the deposition potential from -1.4 to -1.2 V and then decreases on further lowering the potential. Therefore, the deposition potential of -1.2 V was selected as the optimum one in further analysis.

Second, the effect of the pH of the deposition solution on the anodic stripping current was investigated by changing the pH of the acetate buffer from 3.5 to 5.5 (Figure 5B). It is seen that the stripping current increases steadily with the increase of pH, attains a maximum at pH 5, and declines thereafter. This phenomenon is ascribed to the pH-dependent coordination ability of the BMPANI [25], which then affects the subsequent electrochemical reduction of the Cd^{2+} ions and, thereby, the Cd stripping current. Acetate buffer of pH 5 was hence chosen for further analysis.

Finally, the impact of the thickness of the active polymer layer coated onto the CPE surface was assessed by varying the volume of the drop-casting solution from 5 to 40 µL (Figure 5C). Here, the stripping current increases with the increase in layer thickness corresponding to solution volumes from 5 to 20 µL, and then decreases with a further increase in layer thickness for solution volumes from 20 to 40 µL. The optimum thickness of polymer coated onto the electrode surface was therefore taken as that produced by 20 µL of deposition solution.
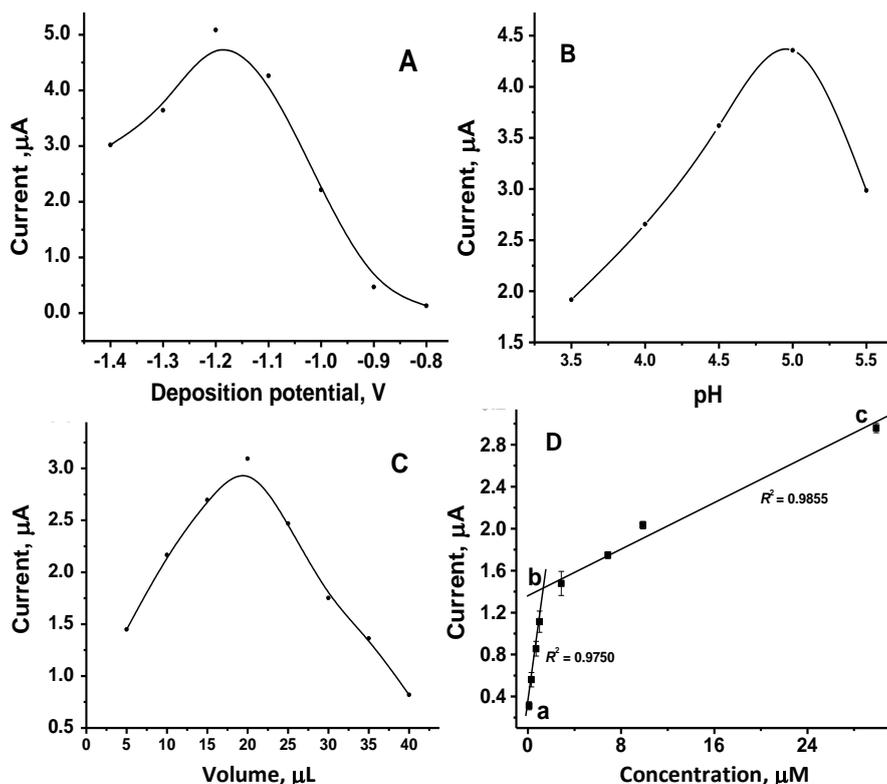

Figure 5. *Stripping current as a function of: A – deposition potential; B – pH of deposition solution; C – thickness of polymer layer (Cd^{2+} ion concentration is 1 µM); D – stripping current as a function of Cd^{2+} ion concentration in the form of two linear correlations*

Table 2 lists the optimized electrode preparation parameters obtained from the DPASV analysis.

Table 2. *Optimized electrode preparation parameters for DPASV studies with Cd^{2+} ions*
(Cd^{2+} ion concentration is 1 μM)

Parameter	Optimized value
Deposition potential, V	-1.2
pH of deposition solution	5
Volume of deposition solution controlling thickness of polymer layer coated onto CPE, μL	20

The pre-set default instrumental parameters used in the individual DPASV experiments were a scan rate of 50 mV/s, pulse amplitude of 100 mV, and pulse period of 40 ms.

Applying the above-optimized electrode preparation parameters and the pre-set DPASV instrumental parameters, a graph was constructed to plot the recorded anodic stripping currents as a function of the Cd^{2+} ion concentration in the range from 1 μM to 0.1 nM, as shown in Figure 5D. It is evident that the anodic current increases with the Cd^{2+} ion concentration. More specifically, this occurs in two linear regions, a first linear regime between the concentrations from 1 to 15 nM with R^2 value of 0.9855 and a second linear regime over the concentrations from 15 to 300 nM with R^2 value of 0.9750. Consequently, with knowledge of these calibration lines, BMPANI-CPE has the potential to be utilized for the determination of Cd^{2+} ions within the specified concentration range with a high degree of accuracy. The limit of detection (LOD) for Cd^{2+} ion sensing was calculated using the equation LOD = $3S_a/b$, where S_a is the standard deviation of the lowest concentration measured, and b is the slope of the linear regression line [26]. For the regions ab and bc, as marked in Figure 5D, the LOD values are calculated as 0.139 and 25.8 nM, respectively.

Selectivity, stability, and reusability of BMPANI-CPE

Selectivity, stability, and reusability are three important qualifying parameters for the commercial application of any sensor. Selectivity determines how the electrode behaves in the presence of interfering species, stability defines the shelf-life of the electrode prior to use, and reusability specifies the number of times a single electrode can be used for the same analysis.

The selectivity of the BMPANI-CPE sensor towards Cd^{2+} ions was tested by recording DPASV signals in aqueous solutions of 100 nM concentration of Cd^{2+} ions in the presence of 0.1 equivalent concentrations of potentially interfering ions, *i.e.,* Zn^{2+}, Pb^{2+}, Ni^{2+}, Co^{2+}, SO_4^{2-}, Cl^-, NO_3^-, $C_2O_4^{2-}$ and PO_4^{3-}, under the experimental conditions previously optimized. Figure 6 represents the effects of these ions on the anodic stripping current. It is found that 0.1 equivalents of Zn^{2+}, Pb^{2+}, Ni^{2+}, Co^{2+} and SO_4^{2-} ions have no marked effect on the Cd^{2+} ion sensing, whereas Cl^-, NO_3^-, $C_2O_4^{2-}$ and PO_4^{3-} ions cause noticeable interferences and thus affect the quantitative analysis of Cd^{2+} ions. Cl^- and NO_3^- ions decrease the anodic stripping current, while $C_2O_4^{2-}$ and PO_4^{3-} ions increase the stripping current, indicating opposing impacts of these ions on the sensing mechanism. Altogether, the results prove that the BMPANI-CPE possesses a good selectivity towards Cd^{2+} ions in the presence of Zn^{2+}, Pb^{2+}, Ni^{2+}, Co^{2+} and SO_4^{2-} ions and should be useable in practical applications where these ions occur in small amounts.

In order to test the selectivity towards Cd^{2+} ions in the presence of higher concentrations of Ni^{2+}, Zn^{2+}, Co^{2+}, Pb^{2+} and SO_4^{2-} ions, DPASV studies were carried out in the presence of both 1 and 10 equivalent concentrations of these ions. Figure 7 presents the results. It is seen that 1 equivalent of Ni^{2+}, Zn^{2+} and SO_4^{2-} ions and 10 equivalents of Zn^{2+} and SO_4^{2-} ions do not interfere with the quantitative analysis of Cd^{2+} ions. Therefore, the proposed sensor should be useable for the determination of Cd^{2+} ions even in the presence of enhanced concentrations of these ions. Only 10 equivalents of Ni^{2+} ions and 1 and 10 equivalents of Co^{2+} and Pb^{2+} ions cause visible interferences.
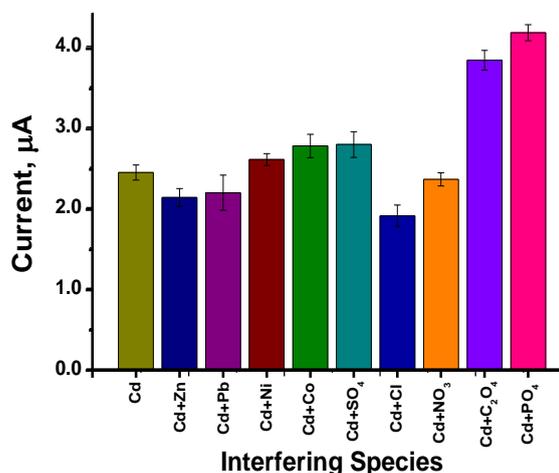
Figure 6. *Effect of 0.1 equivalent concentrations of interfering ions on the determination of Cd²⁺ ions (100 nM) under optimized experimental conditions*
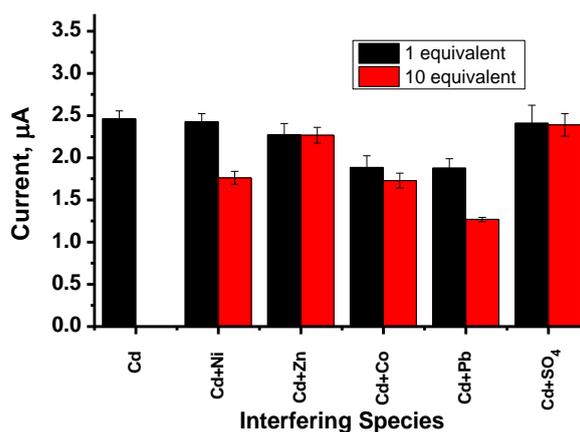

Figure 7. *Effect of 1 and 10 equivalent concentrations of interfering ions on the determination of Cd²⁺ ions (100 nM) under optimized experimental conditions*

The long-term stability, or shelf-life, of the BMPANI-CPE sensor was assessed by recording the DPASV currents for a number of sensors after storage periods between 1 and 15 days. Cd²⁺ ion solutions of concentration 100 nM were used under the optimized experimental conditions, and sensors were kept in a closed vessel prior to analysis. The sensor responses are shown in Figure 8A, in which each current value is the average of three different sensors. The results indicate that more than 80 % of the initial electrode response is retained up to a storage time of 11 days. This demonstrates that the BMPANI-CPE sensor has an acceptable shelf-life for applications with a time gap between sensor preparation and sensor utilization.
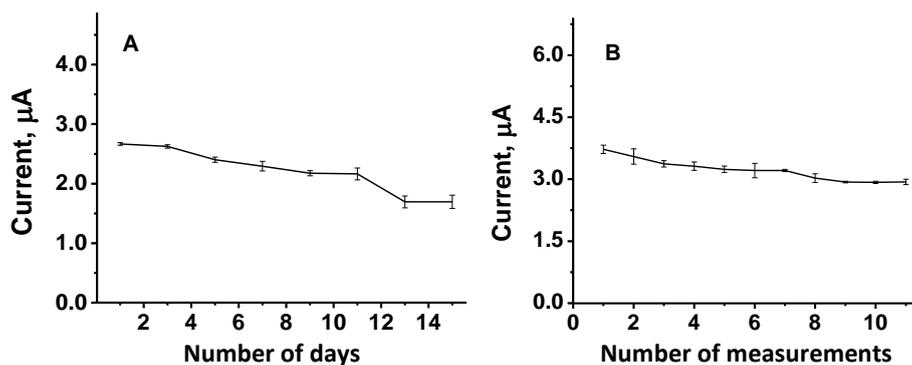

Figure 8. *A – stability over time of BMPANI-CPE for Cd²⁺ ion sensing (100 nM); B – reusability of BMPANI-CPE for Cd²⁺ ion sensing (1 μM)*

The reusability of the BMPANI-CPE sensor was evaluated in a series of successive DPASV analyses of the same sensor in a Cd^{2+} ion solution of concentration 1 μM. Each measurement was followed by leaching in 0.1 M HNO_3 solution to ensure the complete removal of Cd from the electrode surface, as confirmed by DPASV analysis in pure acetate buffer solution. The sensor responses are shown in Figure 8B, with each current reading being the average from three different sensors. The results indicate that more than 85 % of the initial electrode response is retained in up to 7 successive measurements. This highlights the excellent reusability of the BMPANI-CPE sensor, which is hugely beneficial for its practical use.

Conclusions

Polyaniline modified with a side chain of benzimidazole (BMPANI) has been synthesized by interfacial polymerization and characterized with regard to its structure and morphology. The synthesized BMPANI contains the benzimidazole entity as an ortho substituent on its polyaniline backbone and has a unique microstructure of spherical shells resulting from the applied polymerization technique. The synthesized BMPANI has been used to modify the surface of a carbon paste electrode (CPE), and the BMPANI-CPE obtained has been employed as the working electrode for cadmium ion sensing in aqueous media. The electrode preparation parameters have been optimized for maximum sensor sensitivity, and the parameters empirically arrived at are deposition potential of -1.2 V *vs*. SCE, pH 5 for the deposition solution, and a volume of 20 μL for the deposition solution that controls the thickness of the polymer layer coated onto the CPE. Sensor measurements using DPASV have provided a calibration plot exhibiting two linear regions, one in the range from 1 to 15 nM and the other from 15 to 300 nM, with the corresponding LODs of 0.139 and 25.8 nM, respectively. The BMPANI-CPE has been found to possess a good selectivity towards Cd^{2+} ions in the presence of 0.1 equivalents of Zn^{2+}, Pb^{2+}, Ni^{2+}, Co^{2+} and SO_4^{2-} ions. It also displays excellent stability, reusability, and repeatability for Cd^{2+} ion sensing under optimum experimental conditions.

Conflict of interest

The authors declare no conflict of interest.

References

[1] G. Genchi, M. S. Sinicropi, G. Lauria, A. Carocci, A. Catalano, The effects of cadmium toxicity, *International Journal of Environmental Research and Public Health* **17** (2020) 3782. https://doi.org/10.3390/ijerph17113782

[2] G. Kazantzis, Cadmium, osteoporosis and calcium metabolism, *BioMetals* **17** (2004) 493–498. https://doi.org/10.1023/B:BIOM.0000045727.76054.f3

[3] Status of trace and toxic metals in Indian rivers, Ministry of Jal Shakti Department of Water Resources, River Development and Ganga Rejuvenation, Central Water Commission, New Delhi, India, August, 2019.

[4] I. M. M. Kenawy, M. A. H. Hafez, M. A. Akl, R. R. Lashein, Determination by AAS of some trace heavy metal ions in some natural and biological samples after their preconcentration

using newly chemically modified chloromethylated polystyrene-PAN ion-exchanger, *Analytical Sciences* **16** (2000) 493–500. https://doi.org/10.2116/analsci.16.493

[5]   T. Daşbaşı, Ş. Saçmacı, N. Çankaya, C. Soykan, A new synthesis, characterization and application chelating resin for determination of some trace metals in honey samples by FAAS, *Food Chemistry* **203** (2016) 283–291. https://doi.org/10.1016/j.foodchem.2016.02.078

[6]   N. Goudarzi, Solvent microextraction–flame atomic absorption spectrometry (SME–FAAS) for determination of ultratrace amounts of cadmium in meat and fish samples, *Journal of Agricultural and Food Chemistry* **57** (2009) 1099–1104. https://doi.org/10.1021/jf8027057

[7]   Z.-W. Zhang, S. Shimbo, N. Ochi, M. Eguchi, T. Watanabe, C.-S. Moon, M. Ikeda, Determination of lead and cadmium in food and blood by inductively coupled plasma mass spectrometry: a comparison with graphite furnace atomic absorption spectrometry, *The Science of the Total Environment* **205** (1997) 179–187. https://doi.org/10.1016/S0048-9697(97)00197-6

[8]   A. M. Massadeh, A. A. Alomary, S. Mir, F. A. Momani, H. I. Haddad, Y. A. Hadad, Analysis of Zn, Cd, As, Cu, Pb, and Fe in snails as bioindicators and soil samples near traffic road by ICP-OES, *Environmental Science and Pollution Research* **23** (2016) 13424-13431. https://doi.org/10.1007/s11356-016-6499-2

[9]   L. Ebdon, P. Goodall, S. J. Hill, P. B. Stockwell, K. C. Thompson, Ultra-trace determination of cadmium by vapour generation atomic fluorescence spectrometry, *Journal of Analytical Atomic Spectrometry* **8** (1993) 723–729. https://doi.org/10.1039/JA9930800723

[10] W. A. El-Said, M. Abdelshakour, J. H. Choi, J. W. Choi, Application of conducting polymer nanostructures to electrochemical biosensors, *Molecules* **25** (2020) 307. https://doi.org/10.3390/molecules25020307

[11] H. F. Alesary, H. K. Ismail, M. Q. Mohammed, H. N. Mohammed, Z. K. Abbas, S. Barton, A comparative study of the effect of organic dopant ions on the electrochemical and chemical synthesis of the conducting polymers polyaniline, poly(o-toluidine) and poly(o-methoxyaniline), *Chemical Papers* **75** (2021) 5087-5101. https://doi.org/10.1007/s11696-020-01477-8

[12] J. M. S. Alshawi, M. Q. Mohammed, H. F. Alesary, H. K. Ismail, S. Barton, Voltammetric determination of Hg^{2+}, Zn^{2+}, and Pb^{2+} ions using a PEDOT/NTA-modified electrode, *ACS Omega* **7** (2022) 20405-20419. https://doi.org/10.1021/acsomega.2c02682

[13] M. Q. Mohammed, H. K. Ismail, H. F. Alesary, S. Barton, Use of a Schiff base-modified conducting polymer electrode for electrochemical assay of Cd(II) and Pb(II) ions by square wave voltammetry, *Chemical Papers* **76** (2022) 715-729. https://doi.org/10.1007/s11696-021-01882-7

[14] A. Joseph, P. C. Ramamurthy, S. Subramanian, Imidazole functionalized polyaniline: synthesis, characterization, and Cu (II) coordination studies, *Journal of Applied Polymer Science* **123** (2012) 526-534. https://doi.org/10.1002/app.34561

[15] A. Joseph, S. Subramanian, P. C. Ramamurthy, S. Sampath, R. V. Kumar, C. Schwandt, Iminodiacetic acid functionalized polypyrrole modified electrode as Pb(II) sensor: synthesis and DPASV studies, *Electrochimica Acta* **137** (2014) 557–563. https://doi.org/10.1016/j.electacta.2014.06.040

[16] A. G. Mustafin, L. R. Latypova, A. N. Andriianova, I. N. Mullagaliev, S. M. Salikhov, R. B. Salikhov, G. S. Usmanova, Polymerization of new aniline derivatives: synthesis, characterization and application as sensors, *RSC Advances* **11** (2021) 21006-21016. https://doi.org/10.1039/D1RA02474D

[17] B. Gupta, R. Prakash, Interfacial polymerization of polyanthranilic acid: morphology controlled synthesis, *Macromolecular Chemistry and Physics* **213** (2012) 1457-1464. https://doi.org/10.1002/macp.201200152

[18]　S. Ranganathan, P. Raju, V. Arunachalam, G. Krishnamoorty, M. Ramadoss, S. Arumainathan, N. Vengidusamy, Poly(anthranilic acid) microspheres: synthesis, characterization and their electrocatalytic properties, *Bulletin of the Korean Chemical Society* **33** (2012) 1919-1924. https://dx.doi.org/10.5012/bkcs.2012.33.6.1919

[19]　A. A. Khalil, A. F. Shaaban, M. M. Azab, A. A. Mahmoud, A. M. Metwally, Synthesis, characterization and morphology of polyanthranilic acid micro- and nanostructures, *Journal of Polymer Research* **20** (2013) 142. https://doi.org/10.1007/s10965-013-0142-4

[20]　E. T. Kang, K. G. Neoh, K. L. Tan, Polyaniline: a polymer with many interesting intrinsic redox states, *Progress in Polymer Science* **23** (1998) 277-324. https://doi.org/10.1016/S0079-6700(97)00030-0

[21]　B. Sarma, S. Mohan, J. Saravanan, S. Deka, P. Kalita, N. Talukdar, B. Nimavat, Syntheses, characterization and antimicrobial screening of some novel benzimidazoles, *Hygeia Journal for Drugs and Medicines* **7** (2015) 28-37. http://dx.doi.org/10.15254/H.J.D.Med.7.2015.141

[22]　F. Alam, B. K. Dey, A. Chakraborty, Synthesis, characterization and pharmacological screening of some novel benzimidazole derivatives, *International Research Journal of Pharmacy* **5** (2014) 325-331. https://dx.doi.org/10.7897/2230-8407.050469

[23]　A. J. Kadhim, Synthesis and characterization benzimidazole ring by using o-phenylinediamine with different compounds and using Mannich reaction for preparation of some derivatives, *Oriental Journal of Chemistry* **34** (2018) 473–481. http://dx.doi.org/10.13005/ojc/340152

[24]　A. G. Sonkusare, S. Tyagi, R. Kumar, S. Mishra, Room temperature ammonia gas sensing using polyaniline nanoparticles based sensor, *International Journal of Materials Science* **12** (2017) 283-291. ISSN 0973-4589

[25]　J. B. Christensen, T. H. Christensen, The effect of pH on the complexation of Cd, Ni and Zn by dissolved organic carbon from leachate-polluted groundwater, *Water Research* **34** (2000) 3743-3754. https://doi.org/10.1016/S0043-1354(00)00127-5

[26]　A. Shrivastava, V. B. Gupta, Methods for the determination of limit of detection and limit of quantitation of the analytical methods, *Chronicles of Young Scientists* **2** (2011) 21-25. https://doi.org/10.4103/2229-5186.79345