SQUARE WAVE ANODIC STRIPPING VOLTAMMETRIC DETERMINATION OF HG (II) USING CARBON PASTE ELECTRODE MODIFIED WITH –RGO@AU

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Abstract

Heavy metals (HM) are identified as critical environmental pollutants. characterized by their extreme toxicity, ability to accumulate in ecosystems, and lack of degradability. Mercury, in its ionic form, is one of the most toxic pollutants, posing severe risks to the immune system, nervous system, and cellular structures. The electrochemical method for detecting heavy metals has attracted considerable attention due to its ability to produce accurate results. perform analyses faster, and achieve higher sensitivity levels. The main goal of this study is to develop a carbon-based sensor, suitable for the determination of mercury Hg (II). Here, based on the advantages of graphene oxide and gold nanoparticles, we developed the carbon sensor modified with -rGO@Au. The obtained nanomaterial (rGo@Au) is fully characterized using Transmission Electron Microscopy (TEM) and Energy-Dispersive X-ray The Electrochemical characterization of Spectroscopy (EDS). the *CPE/rGOAu sensor was performed via Cvclic Voltammetry (CV), and Square* Wave Anodic Stripping voltammetry (SWASV) was used as a typical technique for the determination of Hg (II). The oxidation peak currents of Hg (II) were proportional to concentration in the range of 0.66-1.96 ppm with a limit of detection of 0.31 ppm. In pursuit of practical applications, the sensor underwent additional testing to measure Hg(II) concentration in water samples.

Key words: Carbon paste electrode, Gold nanoparticles, Reduced Graphene Oxide, Square Wave Anodic Stripping Voltammetry (SWASV), Mercury Hg(II).

Përmbledhje

Metalet e rënda (HM) janë konsideruar ndotës krvesorë të mjedisit për shkak të toksicitetit të lartë, aftësisë për t'u grumbulluar dhe paaftësisë për t'u degraduar në mjedis. Merkuri në formën e tij jonike është një nga ndotësit më të rrezikshëm, duke shkaktuar dëmtime të rënda në sistemin imunitar, atë neurologjik dhe në qelizat e tjera. Metodat elektrokimike për identifikimin e metaleve të rënda kanë tërhequr vëmendje për shkak të saktësisë, shpejtësisë në analiza dhe ndjeshmërisë së lartë. Oëllimi krvesor i këtij studimi është krijimi i një sensori me bazë grafeni për përcaktimin e joneve të mërkurit Hg (II). Bazuar në avantazhet e oksidit të grafenit dhe nanogrimcave të arit, është zhvilluar një sensor karboni i modifikuar me rGO@Au. Kompozita e sintetizuar (rGO@Au) u karakterizua duke përdorur mikroskopinë elektronike të transmetimit (TEM) dhe spektroskopinë me rreze X për shpërndarjen e energjisë (EDS). Karakterizimi elektrokimik i sensorit EPK/rGO@Au u realizua përmes Voltametrisë Ciklike (CV), ndërsa Voltametria me Zhveshje anodike (SWASV) u përdor për përcaktimin e Hg (II). Intensitetet e rrymës oksiduese janë proporcionale me përqendrimin brenda intervalit 0.66-1.96 ppm, me një kufi detektimi prej 0.31 ppm. Në kuadër të aplikimeve praktike, sensori u nënshtrua testeve të tjera për të përcaktuar përgendrimin e Hg(II) në mostra uji.

Fjalë kyçe: Elektroda me paste karboni, Nanogrimcat e Arit, Oksidi i Grafenit i reduktuar, Voltametria me zhveshje anodike, Merkurit Hg (II).

1 Introduction

Heavy metals represent a considerable category of environmental contaminants. Their hazardous nature stems from their non-biodegradable properties, which adversely impact numerous biological and chemical processes, (Aragay et al., 2011), (Iqbal et al., 2023), (Park et al,2017). Mercury Hg (II), found extensively in air, water, and soil, is regarded as one of the most toxic heavy metal ions. It can cause severe health issues, including neurological damage and kidney failure, especially when it bio-accumulates in the food chain. In response to these risks, the urgency for efficient and

accurate detection of Hg(II) is intensifying. (Gong et al., 2020), (Rahman et al., 2019), (Sakthi Priya et al., 2022).

The swift and precise advancement of detection methodologies is crucial for safeguarding human health and preserving ecological balance. In this context, analytical chemistry is essential for the formulation and implementation of more effective methods and techniques for the quantitative assessment of these metals. Traditional analytical methods, including atomic absorption spectroscopy (AAS), atomic emission spectroscopy (AES), etc, demand significant time and financial resources, require skilled personnel, and are limited to ex-situ analysis. (Cao, et al., 2024), (Jency Feminus et al., 2019), (Lateef Ahmad et al., 2019).

The obstacles presented by low concentrations, species distribution, and complex matrices can only be surmounted through the introduction of new, efficient, and cost-effective methods that produce trustworthy results. As a viable option, analytical techniques grounded in electrochemical methods provide an economical, user-friendly, and highly sensitive approach, (Cheng, Y et al., 2019), (Lateef Ahmad et al., 2019), (Zheng, et al., 2018).

Graphene oxide (GO) is a derivative of graphene, which consists of a single layer of carbon atoms arranged in a hexagonal lattice formation. Unlike pure graphene, graphene oxide is characterized by the presence of various oxygencontaining functional groups, such as hydroxyl, epoxy, carbonyl, and carboxyl groups. These groups endow graphene oxide with unique properties that affect its hydrophilicity, conductivity, mechanical strength, and chemical reactivity, (Festinger, N., 2022). However, these functional groups can also lead to issues related to aggregation and π - π bond interactions, (Rahman et al., 2019). To mitigate these challenges, metal nanoparticles are often incorporated into the graphene structure, (Scroccarello et al., 2022). The small size of these nanoparticles enhances the electrode's surface area, increases its reactivity, and facilitates improved electron transfer kinetics (Aragay et al., 2011), (Cheng, Y et al., 2019), (Gong et al., 2020), (Rahman et al., 2019), (Sawan et al., 2020).

In this work, we developed a carbon paste sensor modified with graphene oxide functionalized with gold nanoparticles, aimed at detecting mercury Hg (II). Square Wave Anodic stripping voltammetry (SWASV) served as the fundamental technique for trace metal detection. The Hg²⁺ ions are initially concentrated and then reduced to elemental mercury (Hg⁰) on the

CPE/rGO@Au electrode surface by applying a negative potential. They are subsequently re-oxidized to Hg^{2+} through the application of a different potential in the form of square pulses, (Yang et al., 2021). The current generated is proportional to the concentration of Hg^{2+} ions that have been accumulated on the CPE/rGO@Au electrode, which in turn reflects the concentration in the solution.

2-Materials and methods

2.1 Apparatus and Chemical

The standard mercury solution (Hg at 1000 ppm, AAS grade) was purchased from Merck. 0.1 M acetate buffer pH=4.5 was used as an indifferent electrolyte. All solutions were prepared using distilled water. The material was characterized using Transmission Electron Microscopy (TEM) and Energy-Dispersive X-ray Spectroscopy (EDS). For the acquisition of transmission electron microscopy (TEM) images, a JEOL JEM-2100 electron microscope was used. The PalmSens4 (potentiostat-galvanostat) was employed to carry out electrochemical experiments.

2.2 Preparation of graphene oxide (GO)

Graphene oxide was synthesized through the modified Hummer method by combining 1.5 g of graphene, 1.5 g of sodium nitrate (NaNO₃), and 69 ml of sulfuric acid (H₂SO₄) in a 500 ml beaker. The mixture was stirred until homogeneous, and the beaker was then placed in an ice-water bath. After 15 minutes, 9 g of potassium permanganate (KMnO₄) was added gradually, and the mixture was stirred for 1 hour at room temperature. The beaker was then returned to the ice-water bath, and 100 ml of distilled water was added before heating at 90°C for 1 hour. Afterward, 300 ml of distilled water and 10 ml of 30% hydrogen peroxide (H₂O₂) were mixed in, and the solution was sonicated for 30 minutes at 25°C. The solution was then centrifuged, rinsed with 10% hydrochloric acid and distilled water, and dried at 40°C for 24 hours before being stored in a dry container, (Mnyipika et al., 2021).

2.3 Preparation of reduced graphene oxide functionalized with gold (-rGO@Au)

0.102 g of graphene oxide, 20 ml of distilled water, and 11.9 ml of 0.0249 M gold solution were mixed in a 75 ml beaker. The solution was stirred for about two hours at room temperature using a magnetic stirrer. Afterward, the beaker was placed in an ultrasonic bath for four hours to enhance mixing. The solution was then centrifuged at 5000 revolutions per minute for five minutes. After centrifugation, the precipitate was dried in a thermostat at 80°C for a specified time.

2.4 Preparation of modified carbon paste electrodes.

2.4.1 Preparation of CPE/rGO

A carbon paste electrode modified with reduced graphene oxide (CPE/rGO) was fabricated by adding 1.5 g of carbon powder, 0.150 g of graphene oxide, and 450 μ l of paraffin oil in an agate mortar. The mixture was blended for one hour until a consistent and compact texture was obtained. Then, approximately 1 g of the mixture was placed into a Teflon tube with a copper wire to ensure electrical connectivity. (Yang Z et al., 2024)

2.4.2 Preparation of CPE/rGO@Au

The carbon paste electrode modified with gold functionalized graphene oxide (CPE/rGO@Au) was prepared by combining 1.51 g of carbon powder, 0.153 g of gold functionalized graphene oxide, and 450 μ l of paraffin oil in an agate mortar, mixing gently for 1 hour until reaching a uniform consistency. About 1 g of the mixture was then placed into a Teflon tube with a copper wire to secure electrical connectivity. (Yang Z et al.,2024)

2.5 Electrochemical measurement

The electrochemical analysis was carried out using the PalmSens4 device, which utilizes a three-electrode system. The system included a platinum auxiliary electrode, an Ag/AgCl reference electrode, and a CPE/rGO@Au as the working electrode. The electrochemical characterization of the sensors was performed using cyclic voltammetry in 5 mM Fe(CN)₆ ^{2-/3-}solution containing 0.1 M KCl. Square wave anodic stripping voltammetry (SWASV) was used to quantify Hg (II) in 0.1 M acetate buffer pH=4.5.

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Figure 1. Schematic preparation of CPE/rGO@MNps along with electrochemical measurement

2.6 Sample preparation

Water samples were collected from the decantation unit of the plant (Fushë-Arrëz, Shkodër, Albania) and filtered through 47 mm glass microfiber filters to remove any suspended particles. The samples were acidified with nitric acid (HNO₃), adjusting the pH to 2, and then stored at 4°C. Following storage, the samples were mixed with a 0.1 M acetate buffer at pH 4.5 for further analysis.

3 Results and discussion

3.1 Characterization of the rGo and rGO@Au

Characterization of the gold nanoparticle-functionalized reduced graphene oxide was carried out using Transmission Electron Microscopy (TEM) and Energy-Dispersive X-ray Spectroscopy (EDS). TEM images are presented in Figure 2. (A) for -rGO and in Figure 2.(B) for -rGO@Au. Based on the TEM images, it can be concluded that gold nanoparticles are embedded into the graphene sheets. The darker areas correspond to graphene oxide sheets, while the brighter spots in Figure.2 (B) are attributed to the gold nanoparticles. (Sahoo et al., 2017)



Figure 2. TEM images of A) reduced graphene oxide (-rGO) B) reduced graphene oxide functionalized with gold nanoparticles (-rGO@Au)

EDS was employed to verify the presence and distribution of elements, identify impurities, and examine the material's composition at a microstructural level. The EDS data confirmed the existence of gold, demonstrating that gold nanoparticles were successfully integrated into the -rGO structure. (Cheng, Y et al.,2019) Carbon and oxygen were identified as defining components of graphene oxide. Furthermore, the spectra in Figure 3.(A) and (B) showed the presence of additional elements (Fe, Si, Cl, etc.), suggesting possible impurities from the modifier synthesis and technical interferences, such as copper (Cu) from the supporting tape.





Figure 3. EDS spectra of **A**) reduced graphene oxide rGO **B**) reduced graphene oxide functionalized with gold nanoparticles -rGO@Au

3.2 Electrochemical characterization of carbon-based sensors

Cyclic voltammetry measurements of bare and modified CPE were performed in a solution of potassium ferricyanide. The $Fe(CN)_6^{2^-/3^-}$ interacts closely with the sensor surface, and electron transfer takes place directly through the adsorption of the redox probe onto the material. A quasi-reversible behavior was observed for all the sensors, with a notable decrease in ΔE from bare CPE ($\Delta E = 289$ mV) to modified CPE.

The best improvement was obtained with the CPE/rGO@Au with ($\Delta E = 160$ mV), followed by CPE/rGO (($\Delta E = 190$ mV). The anodic/cathodic peak intensity ratio for CPE/rGO@Au is 1.2, for CPE/rGO 1.6, and for CPE 2.1. The obtained results show that the CPE/rGO@Au displays enhanced electrochemical properties.



Figure 4. CVs of CPE (grey curve), CPE/rGO (yellow curve), and CPE/rGO@Au (blu curve) in 5 mM Fe(CN)6^{2-/3-} solution containing 0.1 M KCl. Scan rate: 100 mV s⁻¹

3.3 Electrochemical behavior of Hg (II) at bare and modified carbon paste electrode

The electrocatalytic performance of CPE, CPE/rGO, and CPE/rGO@Au was evaluated using Square-Wave Anodic Stripping Voltammetry (SWASV) in a 6.6 ppm Hg(II) solution, with a 0.1 M acetate buffer at pH 4.5. As shown in the Figure 5, the peak current increased significantly with the CPE/rGO@Au (yellow curve) compared to CPE/rGO (grey curve) and CPE (blue curve), indicating that the Au nanoparticles strongly adsorb Hg(II).

This enhancement can be attributed to the favorable physicochemical properties of Au nanoparticles, such as their effective catalytic activity, excellent conductivity, and large available surface area.



Figure 5. SWAS Voltammograms of Hg (II) recorded in 0.1M acetate buffer pH=4.5+ 6.6 ppm Hg (II) using CPE (blu curve), CPE/rGO (grey curve), and CPE/rGO@Au (yellow curve).

3.4 Influence of deposition time, amplitude, and frequency

The CPE/rGO@Au was tested in 0.1 M acetate buffer pH=4.5+1.3 ppm Hg²⁺ for evaluation of SWASV operational parameters; deposition time, amplitude, and frequency. The deposition time was examined in the range of 60s - 300s.

(Figure 6. (A)). The stripping peak current gradually increased with increasing deposition time, primarily due to the increased metal ion deposition on the CPE/rGO@Au surface.

In deposition time 300 s stripping peak current was decreased. The reason for this could be the saturation of metal ion concentration on the modified electrode surface as the deposition time increases. Based on the results obtained, the optimal deposition time was 240s. The impact of amplitude, ranging from 5 to 70 mV, on the current for 1.3 ppm Hg (II) is shown in Figure 6. (B), with the optimal amplitude found to be 60 mV. Figure 6. (C) illustrates the effect of frequency on the stripping current peaks, where the optimal frequency was determined to be 25 Hz.



Figure 6. Effect of **A**) deposition time **B**) amplitude **C**) frequency on the anodic stripping peak current of 1.3ppm Hg (II) using CPE/rGO@Au

3.5 Analytical performance of Hg (II)

Figure 7. (A) presents the SWAS voltammogram recorded under optimized conditions. As illustrated, the peak current response was gradually amplified as the concentration of Hg (II) increased. The corresponding calibration plot is shown in Figure 7. (B). The plot demonstrates a linear correlation between the current and concentration, with the following fitting parameters: an intercept of 138.16, a slope of 265.33 μ A/ppm, and a correlation coefficient of 0.9907. The detection limit was identified as 0.31 ppm and the limit of quantification was 1.02 ppm.



Figure 7. **A)** SWAS Voltammograms recorded in different concentrations of Hg (II) in 0.1M acetate buffer pH=4.5 using CPE/rGO@Au. (Ed = -0.1 V, td=240s, f=25 Hz, A=60 mv) **B)** A linear calibration plot between the intensity(μ A) and concentration (ppm)

To evaluate both the repeatability and reproducibility of the CPE/rGO@Au sensor, measurements were performed in 0.39 ppm Hg(II). No significant decrease in the Hg(II) response (less than 5%) was observed after consecutive measurements on the same electrode, indicating satisfactory repeatability. The relative standard deviation (RSD) was 3.29% when different electrodes were evaluated. These results confirm that the CPE/rGO@Au sensor demonstrates both good reproducibility and repeatability.

| Electrode | Analyte | Method | Sensitivity uA /ppm | <i>R</i> ² | Linear range | | LOQ ppm | RSD % |
|----------------|---------|--------|------------------------|-----------------------|-----------------|-----------|------------|----------|
| | | | <i>F</i> / F F | | ppm | FF | FF | |
| CPE/rGO @Au | Hg (II) | SWASV | 265.33 | 0.9907 | 0.66- 1.96 | 0.31 | 1.02 | 3.29 |

Table 1. Analytical performance parameters of CPE/rGO@Au

3.6 Interference study

The interference experiment was investigated by mixing the Hg(II) solution with Cu (II). Cu (II) was selected as the interfering ion for this study because its electrochemical properties closely resemble those of mercury Hg (II), resulting in overlapping peaks during detection. In order to assess the impact of interfering heavy metal (Cu II) on the response (signal) of CPE/rGO@Au, the normalized response percentage was employed, (Lee, J et al., 2021). Formula to calculate Normalized Response Percentage:

Normalized Response $\% = \frac{\text{Response without interference}}{\text{Response with interference}} X 100$

Figure 8. illustrates the impact of different concentrations of Cu (II) on the signal of Hg (II). The graph indicates that a Cu(II) concentration of 0.33 ppm decreases 13% the response for Hg(II). When the concentration increases to 1.98 ppm, this reduction intensifies to 30.51%.



Figure 8. Interference effect on Hg (II) detection of CPE/rGO@Au: **A**)1.98 ppm Hg(II) without Cu(II) **B**) 1.98ppm Hg(II) +0.66 ppm Cu(II) **C**) 1.98ppm Hg(II) +1.98ppm Cu(II)

To verify that elevated levels of Cu(II) influence the current response of Hg(II), various concentrations of Hg(II) were added to a solution containing 0.1 M acetate buffer (pH = 4.5) +0.66 ppm Cu(II). The sensitivity of Hg has decreased slightly from 265.33 μ A/ppm to 182.99 μ A/ppm due to amalgamation and competition for active surface sites between two ions. (Figure 9) The correlation coefficient for Hg is notably high, remaining unaffected by the interference of cations.

The peak current of Cu (II) increased with the addition of Hg due to film formation. The Hg(II) ion tends to reduce at the electrode surface and form a film, which can promote the reduction and ri-oxidation of Cu(II) at the modified electrode. The formation of the film makes it challenging to analyze mercury alongside Cu (II) and other heavy metals simultaneously. (Z.G. Wu et al., 2021)



Figure 9. A) SWAS Voltammograms recorded in different concentrations of Hg (II) in 0.1M acetate buffer pH=4.5 + 0.66 ppm Cu (II) using CPE/rGO@Au.(Ed= -0.1 V, td=240s, f=25 Hz, A=60 mv) B) A linear calibration plot between intensity(μA) and concentration (ppm)

To conclude, elevated concentrations of copper (II) (>0.33 ppm) significantly obstruct the detection of mercury, as demonstrated in Figures 8 and 9. This interference poses challenges for precise mercury measurement, whereas lower copper (II) concentrations have a negligible impact on mercury analysis.

3.7 Real samples analysis

To evaluate the proposed sensor's application, the CPE/rGO@Au was tested with a real water sample by spiking a known concentration of Hg (II). The treated samples were analyzed using the sensor under optimal conditions; the results are provided in Table 2. These results clearly demonstrate that the analytical performance of this sensor for Hg(II) can be accurately quantified with reliability and good precision.

Table 2. Results of the proposed method for the determination of Hg(II) in water samples

| Sample | Analyte | Spiking concentration (ppm) | Calculated concentration (ppm) | Recovery % |
|--|---------|-----------------------------------|--------------------------------------|---------------|
| Water from the decantation unit of the plant | Hg (II) | 8.9 | 9.5 | 107 |

Conclusions

The CPE/rGO@Au sensor, in conjunction with the SWASV technique, presents an innovative, efficient, rapid, and cost-effective electroanalytical approach for the detection of mercury Hg (II) in real water samples. The current observed for Hg(II) detection is significantly higher with the modified electrode compared to the unmodified CPE.

This enhanced response is attributed to the synergistic properties of rGO and AuNP. The proposed sensor exhibits good voltammetric performance, achieving a detection limit of 0.31 ppm, along with notable sensitivity, and reproducibility. When tested on real water samples, the modified electrode demonstrated a recovery rate of 107%, indicating satisfactory performance at the assessed concentration. In areas with elevated copper concentrations >0.33ppm, the sensor may experience significant interference, leading to challenges in accurately determining mercury (II) levels.

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The effectiveness of this electrochemical approach underscores the potential of AuNP and rGO as valuable materials for the development of new nanocomposite-modified electrodes, which can greatly enhance portable analyzers for the sensitive detection of Hg (II) in real water samples.

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