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Original scientific paper

Modification of screen-printed carbon electrodes by $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures to design and fabrication of a new electrochemical sensor for determination of epirubicin

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Abstract

In this work, a screen-printed carbon electrode (SPCE) modified with a new $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanocomposite was used to create a highly sensitive electrochemical sensor for the trace-level detection of epirubicin (EPI). To improve the electron transport kinetics and electrocatalytic oxidation of EPI at the electrode surface, the $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructure was created. Cyclic voltammetry and differential pulse voltammetry were used to thoroughly examine the electrochemical performance of the modified SPCE ($\text{Sb}_2\text{S}_3/\text{MoS}_2/\text{SPCE}$). With a low limit of detection of $0.002\ \mu\text{M}$ and a broad linear concentration range of 0.005 to $220.0\ \mu\text{M}$, the sensor demonstrated exceptional analytical performance for EPI detection under ideal experimental conditions. Additionally, the sensor showed superb repeatability, high selectivity, and outstanding stability. With recovery percentages ranging from 97.1 to 103.3 %, the practical utility of the $\text{Sb}_2\text{S}_3/\text{MoS}_2/\text{SPCE}$ was validated by measuring EPI concentrations in pharmaceutical formulations and human urine samples. These findings demonstrate the $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanocomposite's potential as an excellent electrocatalytic platform, offering a reliable, affordable, and quick analytical tool for therapeutic drug monitoring and clinical diagnostics.

Keywords

Cancer; therapeutic drug monitoring; chemotherapeutic drugs; modified electrode; nanomaterials

Introduction

After cardiovascular illnesses, cancer is the top cause of death globally and continues to be one of the most difficult public health issues [1]. Chemotherapy remains a key component in the treatment of many malignant illnesses despite notable advances in diagnostic tools, surgical techniques, radiation, immunotherapy, and targeted therapeutic approaches [2]. Chemotherapeutic drugs are frequently used alone or in conjunction with other therapies to prevent tumour growth, suppress uncontrolled cell division, reduce the risk of metastasis, and improve patient survival [3]. Because of their broad-spectrum antitumor activity and proven effectiveness against a number of solid tumours and haematological malignancies, anthracycline antibiotics have garnered significant clinical attention among the numerous classes of anticancer medications [4-6].

Epirubicin (EPI), a stereoisomer of doxorubicin and a semisynthetic anthracycline derivative, is frequently used in cancer chemotherapy, especially to treat lung, bladder, ovarian, breast, and other cancers [7]. The therapeutic activity of EPI is mainly attributed to its ability to interact with nucleic acids and interfere with crucial intracellular processes required for cancer cell survival. Like other anthracyclines, EPI can intercalate between DNA base pairs, inhibit topoisomerase II activity, disrupt DNA replication and RNA transcription, and induce the formation of reactive oxygen species [8,9]. These processes eventually lead to DNA strand breaks, oxidative stress, mitochondrial dysfunction, cell-cycle arrest, and apoptotic cell death in rapidly proliferating tumour cells [5].

Although EPI is an effective anticancer drug, several adverse effects limit its clinical use. These side effects include nausea, vomiting, alopecia, mucositis, myelosuppression, and immunosuppression [10-13]. More importantly, anthracycline-induced cardiotoxicity remains a major concern and is strongly associated with the cumulative dose administered to the patient. Therefore, accurate determination of EPI in pharmaceutical formulations, biological fluids, and clinical samples is essential for quality control, pharmacokinetic studies, therapeutic drug monitoring, and prevention of toxic side effects. Reliable monitoring of EPI concentration can help optimize dosage regimens, improve therapeutic efficacy, and reduce the risk of dose-dependent toxicity [4].

Various analytical techniques have been reported for the determination of EPI and related anthracycline drugs. These methods include high-performance liquid chromatography, liquid chromatography coupled with mass spectrometry, UV-visible spectrophotometry, fluorescence spectroscopy, capillary electrophoresis, chemiluminescence, and Raman spectroscopy [8,14-18]. Although these techniques generally provide acceptable sensitivity and selectivity, they often require expensive instruments, highly trained operators, complicated sample preparation, large volumes of organic solvents, and relatively long analysis times [19]. In addition, many of these methods are not suitable for rapid, portable, or on-site detection. Therefore, developing simple, sensitive, low-cost, and miniaturized analytical platforms for EPI determination remains an important objective in pharmaceutical and biomedical analysis [20].

Electrochemical methods have emerged as powerful alternatives to conventional analytical techniques due to their inherent advantages, including high sensitivity, rapid response, operational simplicity, low cost, miniaturization capability, and compatibility with portable devices [21]. Electrochemical sensors are particularly attractive for drug analysis because many pharmaceutical compounds possess electroactive functional groups and can undergo oxidation or reduction at the electrode surface [22]. The analytical performance of an electrochemical sensor depends strongly on the nature of the working electrode, surface chemistry, active electrochemical area, and the rate of electron transfer between the analyte and the electrode interface. Therefore, rational design and

modification of electrode surfaces are key strategies for improving sensitivity, selectivity, stability, and reproducibility [23].

Among different electrode platforms, screen-printed electrodes (SPEs) have gained significant attention in recent years. SPEs are planar, disposable, inexpensive, lightweight, and easy-to-use electrochemical devices that can be mass-produced with high reproducibility [24,25]. A typical SPE contains working, reference, and counter electrodes integrated on a single substrate, simplifying the electrochemical setup and reducing sample consumption. In particular, screen-printed carbon electrodes (SPCEs) are widely used because of their suitable potential window, chemical stability, low background current, and compatibility with a wide range of surface modifiers. These features make SPCEs excellent candidates for the development of portable and point-of-care sensing systems in pharmaceutical, environmental, food, and clinical analysis [26,27].

Despite these advantages, bare SPCEs may exhibit limited sensitivity, slow electron-transfer kinetics, weak electrocatalytic activity, and insufficient selectivity toward target analytes. To overcome these limitations, modification of the working electrode surface with nanostructured materials has become a widely applied approach [26,28]. Nanomaterials can significantly enhance the electrochemical response by increasing effective surface area, providing abundant active sites, improving analyte adsorption, accelerating charge transfer, and lowering the overpotential of redox reactions [29-33]. Accordingly, the fabrication of modified SPCEs using functional nanomaterials is a promising strategy for the sensitive determination of anticancer drugs such as EPI [34].

In this context, two-dimensional and low-dimensional nanostructured materials have received considerable attention because of their unique physicochemical properties. Molybdenum disulfide (MoS_2), a typical transition-metal dichalcogenide, has attracted considerable interest in electrochemical sensing because of its layered structure, high surface area, tuneable electronic properties, and abundant edge-plane active sites [35,36]. The weak van der Waals interactions between MoS_2 layers expose active sites and promote interactions with target molecules. Moreover, MoS_2 exhibits favourable catalytic behaviour and can improve electron-transfer processes when incorporated into electrode interfaces [37]. These characteristics have made MoS_2 a promising material for sensors, electrocatalysis, energy storage, and biomedical applications [38,39].

Antimony trisulfide (Sb_2S_3) is another important metal sulphide semiconductor that has recently been explored in various fields, including photodetectors, energy conversion, photocatalysis, supercapacitors, and electrochemical sensing [40-43]. Sb_2S_3 possesses a narrow band gap, good chemical stability, anisotropic crystal structure, and tuneable morphology [43]. It can be synthesized in different forms such as nanorods, nanowires, nanosheets, and hierarchical nanostructures, which can provide many electroactive sites. Because of these properties, Sb_2S_3 can improve electrochemical activity and enhance interfacial electron transfer [44]. However, like many individual semiconductor nanomaterials, its practical electrochemical performance may be limited by restricted conductivity or aggregation when used alone.

Combining Sb_2S_3 with MoS_2 can be an effective strategy for constructing a hybrid nanostructured sensing interface with improved electrochemical performance [45]. In such a composite system, MoS_2 may provide a layered conductive and catalytic platform with accessible active edges [46], while Sb_2S_3 may contribute additional electroactive sites and favourable semiconductor properties. The integration of these two sulphide-based nanostructures can generate synergistic effects, including enhanced surface area, improved charge-transfer pathways, better analyte adsorption, and increased electrocatalytic activity. Such synergism is particularly beneficial for the oxidation or reduction of

electroactive drug molecules at the electrode surface [47]. Therefore, $\text{Sb}_2\text{S}_3/\text{MoS}_2$ -modified SPCEs are expected to provide a sensitive and efficient platform for EPI determination.

From an electroanalytical perspective, EPI is a suitable target molecule for voltammetric detection because its anthracycline structure contains electroactive quinone/hydroquinone and phenolic moieties that can participate in redox reactions [48]. When the electrode surface is properly engineered, the oxidation signal of EPI can be amplified, improving analytical sensitivity. In this regard, modified SPCEs based on $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures can facilitate electron transfer between EPI and the electrode surface and may reduce interfacial charge-transfer resistance. Consequently, such a sensing system can potentially provide a broad linear concentration range, low detection limit, good reproducibility, and acceptable selectivity in the presence of interfering species [48].

Therefore, a SPCE modified with $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures was used in the current study to develop a novel electrochemical sensor for measuring EPI. The proposed $\text{Sb}_2\text{S}_3/\text{MoS}_2/\text{SPCE}$ combines the advantages of disposable screen-printed electrode technology with the favourable electrochemical characteristics of sulphide-based nanomaterials. The modified electrode was developed to improve the electrocatalytic reaction of EPI and to provide a quick, easy, sensitive, and affordable way to measure it. Under ideal experimental settings, voltammetric techniques can be used to assess the sensor's analytical performance. Additionally, measuring EPI in pharmaceutical formulations and biological samples can evaluate the practical application of the proposed sensor. This work enables the creation of portable electrochemical platforms for pharmaceutical quality control and anticancer medication monitoring.

Experimental

Chemicals and instruments

All chemicals and reagents used in this investigation were of analytical grade and were purchased directly from commercial vendors (Sigma-Aldrich and Merck) without any purification. All solutions were prepared using ultrapure deionized water. In every electrochemical experiment, the supporting electrolyte was phosphate buffer (PB) solution (0.1 M), made from orthophosphoric acid. By gradually adding NaOH solution under pH-meter control, the pH was brought to the specified values. A digital pH meter (Metrohm, model 713, Switzerland) was used for all pH readings and buffer solution formulations. A PGSTAT302N potentiostat/galvanostat (Metrohm, Netherlands) was used for electrochemical tests. A Dropsens DRP-110 screen-printed carbon electrode (SPCE) was used for all electrochemical tests. The working and counter electrodes of the three-electrode system had carbon-based interfaces, whereas a silver pseudo-reference electrode was used. As described in literatures, $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures were produced [49].

Modification process of screen-printed carbon electrodes with $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures

A homogenous suspension was achieved by ultrasonically dispersing 1.0 mg of the composite powder in 1.0 mL of deionized water for 20 minutes to create a thin layer of $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures on an SPCE. A thin coating of $\text{Sb}_2\text{S}_3/\text{MoS}_2$ nanostructures was then applied to the SPCE surface by drop-casting 2.0 μL of the resultant dispersion onto the electrode, which was then allowed to dry. $\text{Sb}_2\text{S}_3/\text{MoS}_2/\text{SPCE}$ is the name of the modified electrode.

Preparing real samples

Ten millilitres of human urine sample were used, and they were centrifuged at 2000 rpm for 600 seconds. Next, various volumes of the treated urine samples were collected, placed in a flask, and

diluted with phosphate-buffered (PB) solution at pH 7.0. These samples were treated with varying concentrations of EPI. The standard addition method is used to determine the EPI concentrations.

1.0 mL of an ampoule containing 2 mg of EPI was mixed with 10.0 mL of PB at pH 7.0. Different volumes of the resulting diluted solution were then transferred into a series of 25 mL volumetric flasks. The investigation was conducted using the standard addition method.

Results and discussions

Electrochemical behaviour of epirubicin on the surface of Sb₂S₃/MoS₂/SPCE

One crucial factor in electrochemical processes is the buffer solution's pH. Studies with 0.1 M PB containing EPI at different pH values (2.0 to 9.0) investigated the effect of pH on EPI determination (Figure 1). Since pH 7.0 showed the largest anodic peak current in differential pulse voltammetry (DPV) experiments for EPI, it was chosen as the solution pH in this work. Scheme 1 shows the mechanism for the redox reaction of EPI.

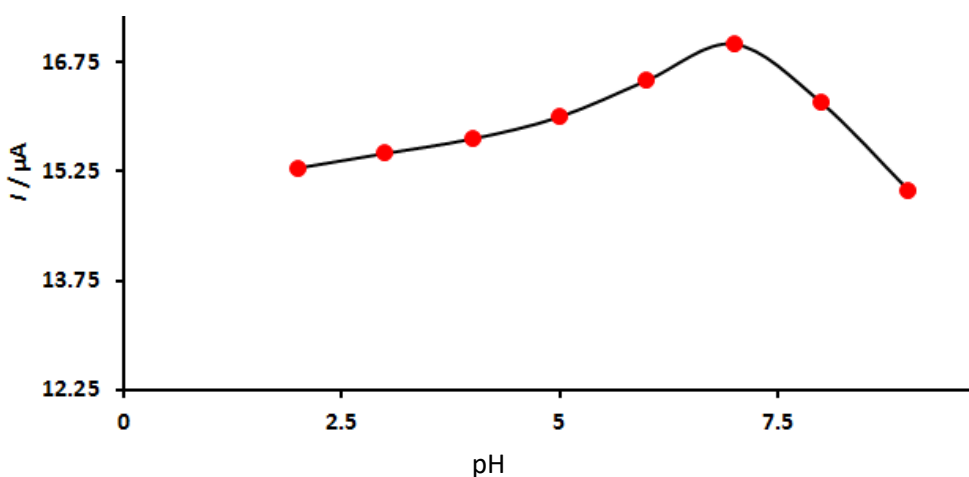
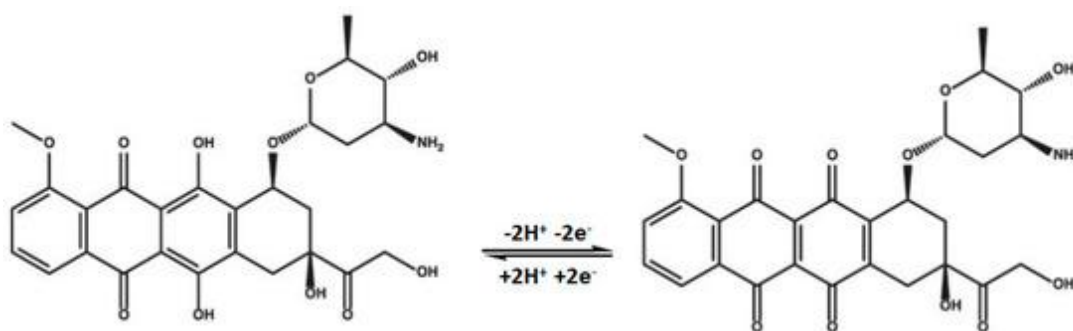


Figure 1. Variation of anodic current of 100.0 μM EPI vs. various pHs of PB (0.1 M)



Scheme 1. The mechanism for the redox reaction of EPI

In 0.1 M PB at pH 7.0 with 100.0 μM EPI, the CV responses of the Sb₂S₃/MoS₂/SPCE (Figure 2, curve b) and an unmodified SPCE (Figure 2, curve a) were measured. Both the Sb₂S₃/MoS₂/SPCE and unmodified electrodes showed clear anodic/cathodic redox peaks for EPI. However, a notable effect on EPI detection was observed on the Sb₂S₃/MoS₂/SPCE. The bare SPCE showed a poor voltammetric response to the EPI redox reaction, resulting in relatively low current values. The Sb₂S₃/MoS₂/SPCE had a greater voltammetric response to EPI than bare electrodes.

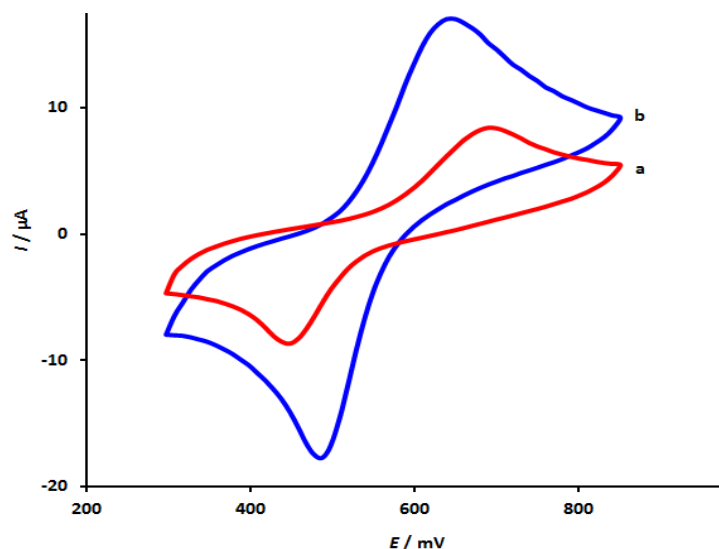


Figure 2. CVs of (a) bare SPCE and (b) Sb₂S₃/MoS₂/SPCE at a scan rate of 50 mV s⁻¹ with 100.0 μM EPI

Additionally, the redox peaks of EPI were observed at lower potentials in the Sb₂S₃/MoS₂/SPCE compared to the unmodified SPCE. Comparing these CVs suggests that the increased sensitivity of Sb₂S₃/MoS₂/SPCE to EPI compared to bare SPCE may be due to the advantageous properties of the Sb₂S₃/MoS₂ nanostructures and their synergistic effects.

Influence of scan rate

CV was used to examine the impact of scan rate on the redox reaction of EPI (100.0 μM) at Sb₂S₃/MoS₂/SPCE (Figure 3). As the scan rate increased, the redox peak potentials of EPI shifted slightly toward more positive values, suggesting kinetic constraints in the redox reaction. Additionally, the square root of the scan rate ($v^{1/2}$) showed a linear relationship with the anodic peak currents (I_{pa}) and cathodic peak currents (I_{pc}) of EPI (Figure 3-Inset). These findings imply that the diffusion process is the main mechanism governing the oxidation and reduction of EPI.

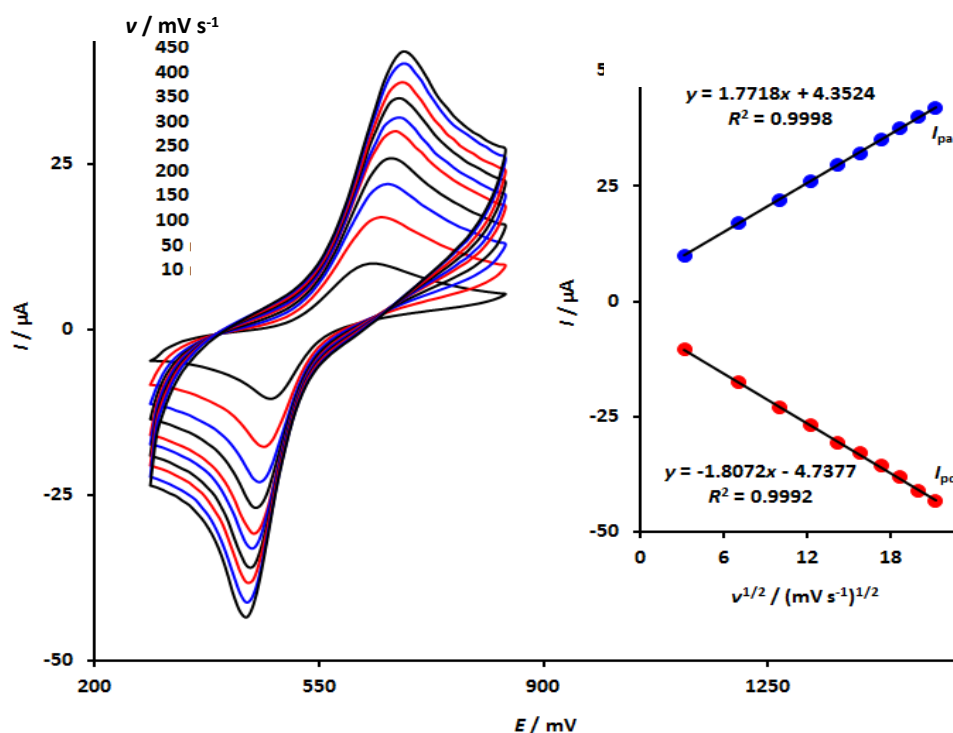


Figure 3. CV responses of Sb₂S₃/MoS₂/SPCE to 100.0 μM EPI at different scan rates. Inset: Plot of peak currents (I_{pa} and I_{pc}) vs. $v^{1/2}$

Chronoamperometric studies

For the EPI investigation, chronoamperometric analysis was also used at concentrations ranging from 0.1 to 1.0 mM at Sb₂S₃/MoS₂/SPCE (Figure 4). A constant voltage of 675 mV was applied to the Sb₂S₃/MoS₂/SPCE (as the working electrode) to perform the analysis. The Cottrell equation (1) defines the chronoamperometric current for an electroactive species with a diffusion coefficient (D) under these mass-transport-controlled conditions:

$$I = nFACD^{1/2}\pi^{-1/2}t^{-1/2} \quad (1)$$

where $D / \text{cm}^2 \text{s}^{-1}$ is the diffusion coefficient, A / cm^2 is the electroactive surface area of Sb₂S₃/MoS₂/SPCE and $C / \text{mol cm}^{-3}$ is the bulk concentration of the EPI. The chronoamperometric current values were plotted against $t^{-1/2}$ for different EPI concentrations (Figure 3A). The slopes of the resulting linear fits were then plotted against the corresponding EPI concentrations, as shown in Figure 4B. Using the slope from this latter plot in conjunction with the Cottrell equation, the average diffusion coefficient for EPI was calculated to be $6.5 \times 10^{-5} \text{ cm}^2 \text{s}^{-1}$.

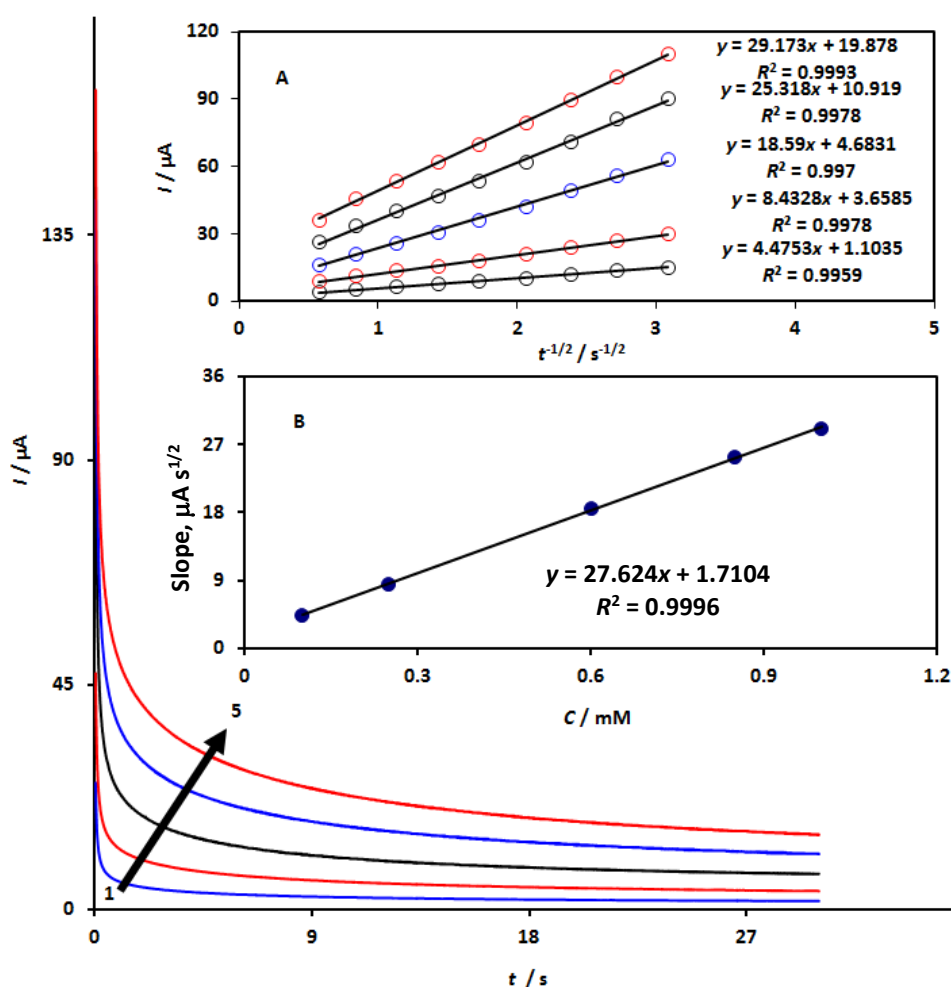


Figure 4. Chronoamperometric responses of Sb₂S₃/MoS₂/SPCE to EPI at different concentrations (1 to -5 correspond to 0.1, 0.25, 0.6, 0.85 and 1.0 mM). Linear dependence of I on $t^{-1/2}$ (inset A) and linear dependence of the slope of the linear fits on EPI concentration (inset B)

Quantitative analysis of the epirubicin by differential pulse voltammetry

Under ideal conditions, the quantitative measurements of EPI were carried out using DPV. The DPV responses of the Sb₂S₃/MoS₂/SPCE in 0.1 M phosphate buffer solution pH 7.0 to various EPI concentrations are shown in Figure 5. The I of EPI progressively increased as the EPI concentration increased, as shown in Figure 5. The I showed a linear response to EPI concentration, according to

the calibration curves from DPV measurements in the Figure 4 inset. A concentration range from 0.005 to 220.0 μM shows this linear dependency.

Using the formula $LOD = 3S/m$, the limit of detection (LOD) was found to be 0.002 μM, where m and S are the calibration curve slope and blank signal standard deviation, respectively.

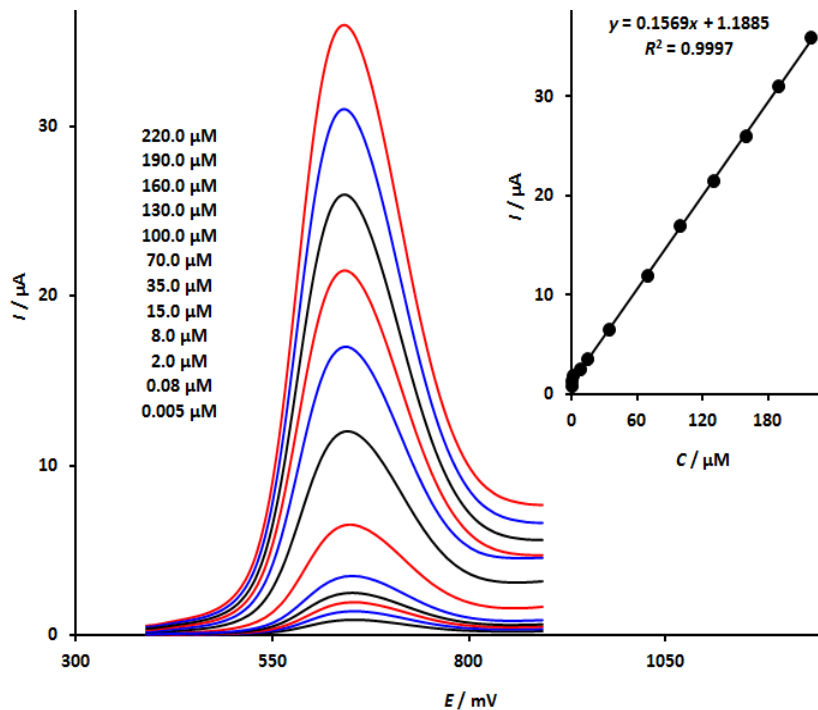


Figure 5. DPVs of Sb₂S₃/MoS₂/SPCE in PB (0.1 M; pH 7.0) with varying EPI concentrations. A plot of the I_{pa} vs. EPI concentrations is shown in the inset

Practical application Sb₂S₃/MoS₂/SPCE sensor in real specimens

To illustrate the usefulness of the developed sensor, the Sb₂S₃/MoS₂/SPCE was used to measure EP in urine samples and injection using the standard addition method. Table 1 presents the results. Recoveries ranging from 97.1 to 103.3 % were satisfactory. Additionally, the relative standard deviations (RSDs) ($n = 5$) were less than 3.3%, indicating the remarkable accuracy of the developed sensor. Therefore, the Sb₂S₃/MoS₂/SPCE sensor can be used to analyse EPI in real specimens.

Table 1. Utilizing the Sb₂S₃/MoS₂/SPCE sensor to measure EPI in actual samples

Real sample	Amount, μM		Recovery, %	RSD, % ($n = 5$)
	Spiked	Detected		
EPI injection	0	3.1	-	3.0
	2.0	5.0	98.0	2.8
	4.0	7.3	102.8	2.5
	6.0	9.4	103.3	1.9
	8.0	11.0	99.1	2.6
Urine	5.0	5.1	102.0	3.3
	7.0	6.8	97.1	2.9
	9.0	9.3	103.3	3.0
	11.0	10.8	98.2	2.1

Conclusion

In conclusion, an Sb₂S₃/MoS₂/SPCE electrochemical sensing platform was effectively created for the detection of the anticancer medication EPI. The modified electrode showed a significantly higher electrochemical response and greater electrocatalytic activity toward EPI oxidation than the bare

SPCE. Advantageous semiconducting characteristics of Sb_2S_3 , layered structure and plentiful active sites of MoS_2 and the synergistic interaction of two metal sulphide nanostructures promoted charge transfer and raised the effective electroactive surface area responsible for this improvement.

The $\text{Sb}_2\text{S}_3/\text{MoS}_2/\text{SPCE}$ sensor demonstrated exceptional sensitivity for EPI determination over a linear concentration range of 0.005 to 220.0 μM under optimum experimental circumstances, with a low detection limit of 0.002 μM . Additionally, the suggested sensor showed adequate repeatability, stability, and selectivity. Ultimately, EPI analysis in real samples yielded satisfactory recovery results, demonstrating the usefulness of the suggested approach for pharmaceutical analysis and therapeutic medication monitoring.

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