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Application of a novel polymer-modified screen-printed gold electrode for detection of the anticancer drug doxorubicin

Abstract

Screen-printing technology has emerged as an efficient method for constructing highly sensitive and accurate electrochemical sensors, with applications across various fields. This unique combination of sensitivity, selectivity, and stability, along with miniaturization, portability, and on-site, in situ detection, makes screen-printed electrodes a valuable tool in sensor design. With these advantages, screen-printed electrode-based sensors have become increasingly popular in pharmaceutical and biological analysis, for detecting trace biomolecules. In this study, a novel, polymer-modified screen-printed gold electrode electrochemical sensor has been developed for detecting low clinical concentrations of doxorubicin. The sensor modification, based on a Nano composite of reduced graphene oxide decorated with gold nanoparticles embedded in a Nafion matrix, has not been previously reported in the literature. Using cyclic voltammetry, the sensor achieved accurate quantification of doxorubicin, with a linear concentration range of 1.5 to 22.1 µM and a detection limit of 11.462 µM under optimal conditions. The sensor's high surface area, functionalization capability, wide linear range, high sensitivity, and rapid response indicate its potential for practical applications in anticancer drug detection.

Keywords: electrochemical sensor, screen-printed gold electrode, doxorubicin, anticancer medication, monitoring

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Introduction

Screen-printed electrodes (SPEs) are versatile tools widely used in electrochemical sensor construction and are recognized as effective platforms for designing disposable electro analytical sensors. They offer numerous advantages, including rapid and reliable analysis, high sensitivity, good selectivity, ease of use, miniaturization, uniformity, portability, and cost-effectiveness.[1] The concept of screen-printing emerged to address the need for smaller, more affordable electrochemical devices, making these tools more accessible and practical. Screen-printing technology gained groundbreaking recognition and commercial success with the development of the glucose biosensor.[2] In the early 2000s, the commercialization of SPE-based devices expanded significantly across fields such as environmental monitoring, food safety, and healthcare.[3] The affordability, portability, and ease of mass production made SPEs highly attractive for diverse applications, including pharmaceutical and biological analysis.[4] SPEs have since been successfully applied for on spot, in situ detection of various analytes across a wide range of matrices, enabling the detection of pharmaceuticals and other biomolecules.[1] One of the main advantages of SPEs is their adaptability: they can be used as disposable, ready-to-use electrodes or surface-modified for specialized applications, making them suitable for trace determination of biomolecules.[5, 6] Recent advancements in SPE technology have focused on enhancing performance through innovative surface-modification strategies integrating nanomaterials.[7] The modification is employed for sensitivity enhancement, selectivity improvement, and overall stability increase.[8] Two main approaches are typically considered: first, altering the printing ink composition by incorporating polymers, metals, complexes, enzymes, and other materials to develop novel ink-based SPEs; second, modifying the surface of commercial electrodes with metal or ceramic composite films, enzymes, or polymers. The class of carbon nanomaterials, comprising carbon nanotubes (CNTs), graphene, graphene oxide (GO), reduced graphene oxide (rGO), carbon black, and carbon quantum dots, provides materials commonly used as SPEs surface modifiers. These nanomaterials offer several advantages, including a significant enhancement of the electrochemical response by increasing the electroactive surface area and improving electron transfer rates, which boosts the analytical sensitivity of the analysis. Additionally, the high electron transfer rate of these nanomaterials can help reduce fouling issues, resulting in more stable and reliable sensor performance.[9] Reduced graphene oxide, obtained by removing oxygen groups from GO, has notably improved electrical conductivity and excellent electron transfer capability compared to GO, making it ideal for electrochemical applications.[10] Its large surface area, retained after reduction, can be readily functionalized with biomolecules, polymers, and other materials, thereby enhancing the sensitivity and selectivity of SPEs.[11] Additionally, modifying SPE surfaces with polymers can offer further performance benefits.[12–16] For instance, Nafion, known for its chemical and mechanical stability, enhances the durability and longevity of modified SPEs. Beyond its good electrical conductivity, Nafion also contributes to greater analytical signal intensity and helps minimize interference effects.[17] Incorporating metal nanoparticles of varying sizes (1–100 nm) onto the SPE surface introduces unique optical, mechanical, electrical, and chemical properties, which expand the surface-active area, increase transport rates, and facilitate faster electron transfer.[18] Gold nanoparticles (Au NPs) exhibit specific chemical, physical, and electrochemical characteristics i.e. active area increase, making them ideal for improving sensitivity, conductivity, and stability.[19, 20]

Doxorubicin hydrochloride (DOX), an anthracycline anticancer drug, is widely used to treat various cancers, including breast, bladder, ovarian, and bone cancer, as well as leukemia and lymphomas.[21] Despite its effectiveness against this wide range of cancers, its use is limited by numerous side effects, the most severe being acute and chronic cardiotoxicity, which can lead to cardiomyopathy or heart failure.[22] Consequently, rigorous monitoring of DOX levels in patients following chemotherapy is crucial to ensure accurate drug dosing. The use of screen-printed electrode-based electrochemical sensors meets the need for rapid, accurate, portable, and on-site detection and analysis. Ghanbari et al.[23] reported development of an electrochemical sensor for detection of DOX, constructed by modifying a glassy carbon electrode (GCE) with a nanocomposite based on reduced graphene oxide, gold nanoparticles and a 2-amino-5-mercapto-1,3,4-thiadiazole electropolymerized film. The authors reported linear response in the 30 pM to 30 nM and 30 nM to 30 µM DOX concentration ranges, with a limit of detection (LOD) of 9 pM. Zhao et al.[24] proposed a sensitive electrochemical sensor for DOX detection based on a novel electrode material of covalent organic frameworks decorated with gold nanoparticles and multiwalled carbon nanotubes (AuNPs@COFs-MWCNTs) and reported that the sensor exhibited a better liner range for DOX from 0.08 µM to 25 µM with a low detection limit of 16 nM. A glassy carbon electrode modified with multi-walled carbon nanotubes (MWCNTs) decorated with gold nanoparticles has been proposed as an ultrasensitive electrochemical sensor for the determination of DOX by Sharifi et al.[25] The authors reported a wide linear DOX concentration range from 1×10^{-11} to 1×10^{-6} M with a very low detection limit of 6.5 pM under optimal conditions.

In this work, we propose an electrochemical sensor based on screen-printed electrodes as a platform for surface modification. A synthesized conductive ink, composed of reduced graphene oxide decorated with 0.5 wt.% gold nanoparticles (rGO + 0.5 wt.% Au NPs) and embedded into a Nafion matrix was used as a modifying material and deposited onto a commercial screen-printed gold electrode. This modification aims to enhance sensitivity for detecting trace amounts of doxorubicin in a simulated biological matrix. The components of the ink were carefully selected to optimize the sensor's performance by enhancing conductivity, stability, and sensitivity- a combination not previously reported in the literature. Reduced graphene oxide offers a large surface area and excellent electrical properties, while the gold nanoparticles increase electron transfer rates, resulting in a stronger and more reliable analytical signal. The interaction mechanism between DOX and rGO, involving π-π stacking and hydrophobic interactions, is illustrated in Figure 1.[26]

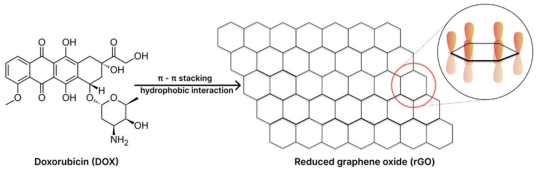

Figure 1 Mechanism of interaction between DOX and rGO.

Experimental

Screen-printed gold BT carbon electrodes (SPEs) (model DS220) were purchased from Dropsens Ltd., Llanera, Asturias, Spain. These gold BT SPEs are cured at low temperatures, resulting in a rougher surface that enhances non-covalent attachment between the electrode and the modifying material. The electrodes were then modified with a nanocomposite material embedded in a polymer matrix, serving as a polymer-based conductive ink. A two-step method described by Behravan et al. [27] was followed to synthesize the nanocomposite. As illustrated in Figure 2, this method involves reducing the graphene oxide in the first step and decorating the reduced graphene flakes with 20 nm gold nanoparticles in the second, to obtain the rGO + 0.5% wt. Au NPs nanocomposite material. All chemicals were purchased from Sigma Aldrich. Furthermore, the synthesized rGO + 0.5 wt.% Au NPs nanocomposite was incorporated into a Nafion matrix to produce a conductive ink for coating and modifying the gold BT SPEs. The ink was prepared by mixing the nanocomposite with 5% wt. Nafion solution (Ion Power, USA) as a binder, Vulcan XC-72R carbon powder (Cabot Corporation, USA), ethanol, and distilled water, followed by 1-hour sonification. The modified SPEs were then dried under infrared light. The detailed experimental scheme is given in Figure 3. All electrochemical measurements were performed in 25 mL of 0.1 M phosphate-buffered saline (PBS) with pH 7.0, used as an electrolyte. Doxorubicin hydrochloride (purchased from Ebewe a.d. as a 2 mg/mL solution) was added to the electrolyte in portions ranging from 10 to 150 µL, resulting in a linear concentration range of 1.5 to 22.1 µM. Cyclic voltammetry (CV) was used as the main electrochemical characterization technique on an Ivium Vertex One potentiostat/galvanostat/ZRA, in the potential range from –0.4 to +0.6 V, under automatic current setting, with a total of 3 scans collected at a scan rate of 25 mV/s. Two calibration curves were constructed in the domain from 1.5 to 10.3 µM and 11.8 to 22.1 µM and the LOD and limit of quantification (LOQ) were calculated based on the S/N = 3 and S/N = 10, respectively. All electrochemical measurements were performed in triplicate at 25 °C.

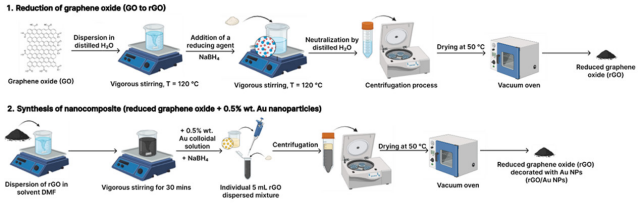

Figure 2 Two-step method synthesis of reduced graphene oxide decorated with Au nanoparticles (rGO + 0.5% wt. Au NPs).

Results and discussion

The electroanalytical behavior of the commercial (unmodified) and modified gold BT SPEs was evaluated and compared using cyclic voltammetry (CV) in 25 mL of 0.1 M PBS at pH 7.0. The analyte, DOX, was added to the solution in two linear concentration ranges: 1.5-10.3 µM and 11.8-22.1 µM. The electrochemical profile of the unmodified and modified SPEs is shown in Figure 3. Cyclic voltammograms define the peak potential (Ep), as the point where the current reaches its maximum. As shown in Figure 4, the voltammograms of the tested SPEs display two distinctive current peaks in the anodic and cathodic regions of the voltammograms, corresponding to the redox process of DOX and indicating a reversible reaction on the electrode surface. In CV, reversibility refers to the ability of an electrochemical reaction to proceed easily in both oxidation and reduction directions while maintaining equilibrium. The degree of reversibility can be quantified by the peak-to-peak separation (ΔEp), calculated as the difference between the anodic and cathodic peak potentials.[27] A smaller ΔEp value correlates with higher reversibility, while a larger ΔEp suggests lower reversibility. Although this relationship is not strictly inverse, ΔEp indicates how closely the reaction approaches ideal reversibility. In our systems, the modified SPE exhibits an average ΔEp of 80 mV, compared to 105.5 mV for the commercial SPE. This reduction in ΔEp indicates enhanced performance, demonstrating improved reversibility and more efficient electron exchange on the modified SPE's surface. All oxidation peak potentials are summarized in Table 1.

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Table 1 Oxidation peak potentials of the modified and commercial SPE in the linear concentration range from 1.5 μM to 22.1 μM

Concentration [μM]	Modified SPE's potential [mV]			Commercial SPE's potential [mV]		
	Pox	Pred	ΔEp	Pox	Pred	ΔEp
0	419	341	78	470	362	108
1.47	424	342	82	473	367	106
4.42	429	356	73	446	348	98
7.36	408	321	87	514	404	110
Average			80			105.5

Figure 3 rGO + 5% wt. Au NPs)/Nafion-based conductive ink preparation method.

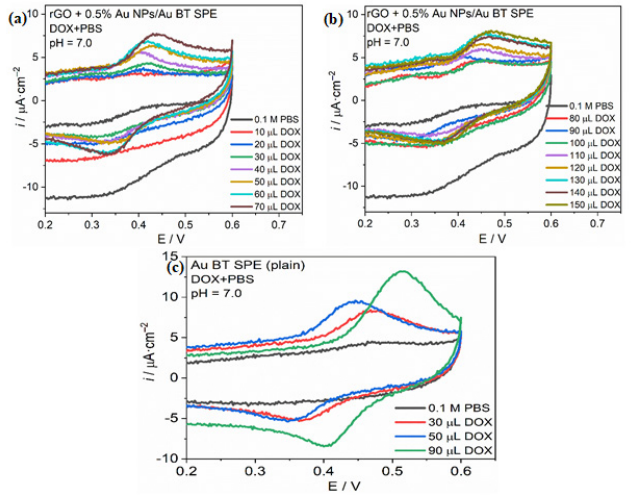

Figure 4 Cyclic voltammograms of the modified SPE tested in **(a)** low and **(b)** high concentration ranges, compared to the unmodified SPEs **(c).**

The quantitative determination of DOX was performed using the CV method under optimal conditions. Increasing concentrations of DOX were employed to determine the relationship between the concentration and the analytical signal, and this data was used to construct the calibration curve. Moreover, the sensitivity, LOD, and LOQ were calculated from the calibration curve. Figure 5 depicts the CV responses of the modified rGO + 0.5% wt. Au NPs/ Au BT SPE and commercial Au BT SPE sensor toward the different concentration ranges of DOX. It can be noticed that the correlation between the DOX's concentration and the oxidation peak current exhibits great linearity for the modified sensor, in both concentration ranges. The corresponding linear function is characterized using the equation $I_{ox}(\mu A) = 0.08505 \times C(\mu M) + 0.13128; R^2 = 0.89041$ for the modified SPE tested in the low concentration range, $I_{ox}(\mu A) = 0.02053 \times C(\mu M) + 0.36446; R^2 = 0.95295$ for the modified SPE tested in the high concentration range and $I_{ox}(\mu A) = 0.05023 \times C(\mu M) + 0.67997; R^2 = 0.84419$ for the unmodified, commercial SPE. The sensitivity of the modified electrode was found to be 0.085 μA/μM in the low linear concentration range and 0.021 μA/μM in the high linear concentration range, compared to a slope of 0.050 μA/μM for the unmodified SPE. Both linear concentration ranges were used to calculate the LOD and LOQ for the modified and commercial SPEs. The LOD for the modified electrode was estimated at 11.462 μM in the 1.5 to 10.3 μM concentration range and 15.933 μM in the 11.8 to 22.1 μM concentration range, while the LOD for the commercial SPE was calculated to be 18.141 μM. Similarly, the LOQ was determined to be 38.207 μM and 53.108 μM for the modified SPE in the low and high concentration ranges, respectively. These values are significantly lower than the LOQ of 60.471 μM calculated for the commercial electrode. These findings, which characterize the analytical performance of our modified electrochemical sensor, align with the practical requirements for a sensor intended to monitor DOX levels in cancer patients, where the expected maximum plasma concentration reaches up to 11 μM. [27]

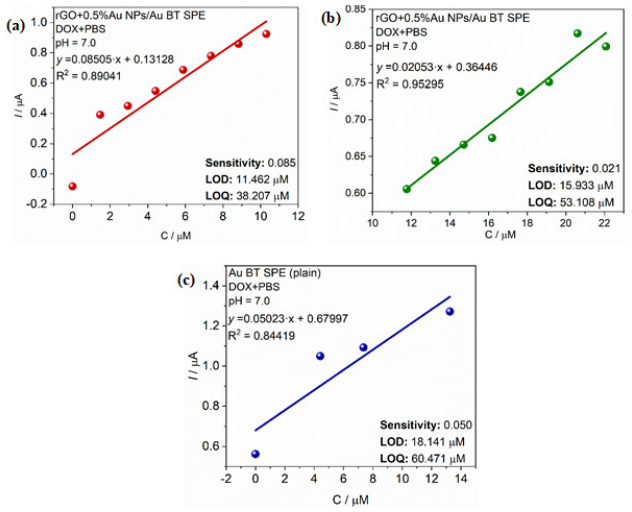

Figure 5 Linear correspondence between the current and DOX concentration for the modified SPE tested in **(a)** low and **(b)** high concentration ranges, compared to the unmodified SPEs **(c).**

Conclusion

A novel modified screen-printed electrochemical sensor has been developed for the detection and monitoring of doxorubicin, an anthrax cycline chemotherapeutic widely used in cancer treatment. The sensor is constructed on a screen-printed gold electrode, which serves as a sensing platform, surface-modified with a nano composite of reduced graphene oxide decorated with gold nanoparticles, embedded in a Nafion matrix. The conducted electrochemical characterization confirmed the superior reversibility observed on the modified electrode compared to the commercial one. The modified electrode also exhibited lower detection and quantification limits than the unmodified one. This sensor demonstrated high sensitivity, stability, and a broad linear response range, making it well-suited for detecting and monitoring traces of doxorubicin. The unique combination of

materials and modification strategies has not been previously reported, highlighting the innovation of this approach. Being characterized by superior properties, this modified sensor shows significant promise for practical applications in pharmaceutical and clinical settings, particularly in the detection of anticancer drugs like doxorubicin.

Acknowledgments

None.

Conflict of interests

The authors declare that there are no conflicts of interest.

References

1. Couto RAS, Lima JLFC, Quinaz MB. Recent developments, characteristics and potential applications of screen-printed electrodes in pharmaceutical and biological analysis. *Talanta.* 2016;146:801–814.
2. Honeychurch KC, Hart JP. Screen-printed electrochemical sensors for monitoring metal pollutants. *TrAC Trends Analyt Chem*. 2003;22(7):456–469.
3. Mohamed HM. Screen-printed disposable electrodes: pharmaceutical applications and recent developments. *TrAC Trends Analyt Chem*. 2016;82:1–11.
4. Taleat Z, Khoshroo A, Mazloum AM. Screen-printed electrodes for bio sensing: a review. *Microchim Acta.* 2014;181:865–891.
5. Ferrari GM, Foster CW, Kelly PJ, et al. Determination of the electrochemical area of screen-printed electrochemical sensing platforms. *Biosensors*. 2018;8(2):53.
6. Nazarpour S, Hajian R, Hosseini SM. A novel nano composite electrochemical sensor based on green synthesis of reduced graphene oxide/gold nanoparticles modified screen printed electrode for determination of tryptophan using response surface methodology approach, *Microchem J*. 2020;154:104634.
7. Metters JP, Banks CE. Screen printed electrodes open new vistas in sensing: application to medical diagnosis. *Modern Aspects Electrochem.* 56. Springer, Boston, MA.
8. Kelíšková P, Oleksandr M, Lenka J, et al. Recent advances in the use of screen-printed electrodes in drug analysis: A review. *Current Opinion in Electrochemistry.* 2023;42:101408.
9. Silva RM, Silva AD, Camargo JR, et al. Carbon nanomaterials-based screen-printed electrodes for sensing applications. *Biosensors*. 2023;13(4):453.
10. Kumuda S, Gandhi U, Mangalanathan U. Synthesis and characterization of graphene oxide and reduced graphene oxide chemically reduced at different time duration. *J Mater Sci: Mater Electron.* 2024;35:637.
11. Yu W, Sisicd L, Haiyana Y. Progress in the functional modification of graphene/graphene oxide: a review. *RSC Adv.* 2020;10:15328–15345.
12. Ku S, Palanisamy S, Chen SM. Highly selective dopamine electrochemical sensor based on electrochemically pretreated graphite and nafion composite modified screen printed carbon electrode. *J Colloid Interface Sci.* 2013;411:182–186.
13. García RG, Fernández A, García AC. Nafion modified-screen printed gold electrodes and their carbon nanostructuration for electrochemical sensors applications. *Talanta*. 2013;107:376–381.
14. Couto RAS, Lima JLFC, Beatriz M, Screen printed Electrode Based Electrochemical Sensor for the Detection of Isoniazid in Pharmaceutical Formulations and Biological Fluids. *Int J Electrochem Sci*. 2015;10(10):8738–8749.
15. Grozdanov, Paunović P, Dimitrievska I. PANI-based sensors: synthesis and application. *IntechOpen*. 2023.
16. Couto RAS, Quinaz MB. Development of a nafion/MWCNT-SPCE-Based portable sensor for the voltammetric analysis of the anti-tuberculosis drug ethambutol. *Sensors*. 2016;16(7):1015.
17. Rivero KT, Florido A, Arrieta JB. Recent trends in the improvement of the electrochemical response of screen-printed electrodes by their modification with shaped metal nanoparticles. *Sensors*. 2021;21(8):2596.
18. Nazarpour S, Hajian R, Sabzvari MH, A novel nanocomposite electrochemical sensor based on green synthesis of reduced graphene oxide/gold nanoparticles modified screen printed electrode for determination of tryptophan using response surface methodology approach, *Microchem J.* 2020;154:104634.
19. Ghanbari MH, Norouzi Z. A new nanostructure consisting of nitrogen-doped carbon nano onions for an electrochemical sensor to the determination of doxorubicin. *Microchem J. 2020*.
20. Pusta M, Tertis I, Bura D, et al. Electrochemical sensor for the evaluation of doxorubicin from novel pharmaceutical formulations and serum. *Chemosensors*. 2024;12(4):69.
21. Ghanbari MH, Fard SH, Salehzadeh H, et al. A nanocomposite prepared from reduced graphene oxide, gold nanoparticles and poly(2-amino-5-mercapto-1,3,4-thiadiazole) for use in an electrochemical sensor for doxorubicin. *Microchim Acta. 2019;186*:641.
22. Zhao H, Shi K, Zhang C. Spherical COFs decorated with gold nanoparticles and multiwalled carbon nanotubes as signal amplifier for sensitive electrochemical detection of doxorubicin, *Microcheml J*. 2022;182:107865.
23. Sharifi H, Fayazfar. Highly sensitive determination of doxorubicin hydrochloride antitumor agent via a carbon nanotube/gold nanoparticle based nanocomposite biosensor. *Bioelectrochemistry*. 2021;139:107741.
24. Liu Z, Liu J, Wang T, et al. Switching off the interactions between graphene oxide and doxorubicin using vitamin C: combining simplicity and efficiency in drug delivery. *J Mater Chem* B. 2018;6:1251–1259.
25. Behravan M, Aghaie H, Giahi M. Determination of doxorubicin by reduced graphene oxide/gold/polypyrrole modified glassy carbon electrode: A new preparation strategy. *Diamond and Related Materials*. 2021;117:108478.
26. Paunović P, Grozdanov A, Dimitrievska I. Voltammetric Detection of Diclofenac with Screen-printed Electrodes Based on graphene and PVDF-Modified grapheme. *Kem Ind.* 2024;73(7–8):293–302.
27. Chekin F, Myshin V, Ye R, et al. Graphene-modified electrodes for sensing doxorubicin hydrochloride in human plasma. *Anal Bioanal Chem.* 2019;411:1509–1516.