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1. Introduction

Currently, tuberculosis (TB) is considered to be one of the most infectious fatal diseases worldwide. It is a multi-systematic disease caused by *Mycobacterium tuberculosis* bacilli and is a challenging global death threat towards the entire human community.^{1–6} According to the Global Tuberculosis Report by the World Health Organization, 10 million people (5.8 million men, 3.2 million women and 1 million children) were newly infected in 2017, and 1.3 million deaths from TB were reported.⁷ Isoniazid (ISZ) is the most extensively used antibiotic and anti-tuberculostatic drug employed for the treatment/prevention of pulmonary tuberculosis; it has been used since 1952.^{8,9} Extended treatment with ISZ leads to liver damage, epilepsy, irregular heartbeat and in some critical

Synergistic design of a tin phosphate-entrapped graphene flake nanocomposite as an efficient catalyst for electrochemical determination of the antituberculosis drug isoniazid in biological samples

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The rapid quantification of drugs in biological samples has become an important issue and ongoing challenge to protect human health. To address this issue, we designed and synthesized a tin phosphateentrapped graphene flake (SnP/GRF) nanocomposite for the trace level electrochemical detection of the antituberculosis drug isoniazid (ISZ). Moreover, the successful formation of the SnP/GRF nanocomposite was confirmed by various spectroscopic and analytical techniques. Based on its synergistic effect, the proposed electrode material exhibits excellent and superior electrochemical activity towards ISZ detection. The SnP/GRF nanocomposite-modified electrode possesses high electric conductivity and good charge transfer, which is highly desired for an advanced electrocatalyst. Benefiting from the fabricated SnP/GRF-modified electrode, the ISZ sensing exhibited a wide dynamic working range (0.01 to 348 μ M) and a low detection limit (1.0 nM) with outstanding sensitivity (1.418 μ A μ M⁻¹ cm⁻²). More importantly, the SnP/GRF-based electrochemical sensing system also has good storage stability, reliable reproducibility, rapid response time and high selectivity. Our proposed electrochemical sensor has promising potential for the determination of ISZ in biological samples. Our work represents an innovative design to fabricate multifunctional catalysts for versatile applications in emerging next-generation electrochemical fields.

> cases, even death.^{10–12} In addition, the overdosage of ISZ in the human body produces hydrazine through the metabolism of ISZ.^{13–15} Importantly, ISZ induced lung tumors in mice and received a black box warning in 1969.¹⁶ Due to these inevitable health problems, the World Health Organization considers the permissible limits of ISZ drug to be 4 to 6 mg kg⁻¹ per day. In order to mitigate this, the examination of patients treated with ISZ at periodic intervals is highly important.^{17–19}

> Currently, various analytical methods have been developed for the selective detection of ISZ, including high performance liquid chromatography, gas chromatography, gas chromatography-mass spectrometry, capillary electrophoresis, chemiluminescence and spectrophotometry.^{2–4,20,21} These detection techniques have significant advantages of high sensitivity and precise results. However, the abovementioned methods have drawbacks, such as high cost, need for qualified operators, sophisticated instruments, long processing times, and complex systems; thus, they are not suitable for real time monitoring. Compared with the aforementioned techniques, electrochemical sensors offer significant advantages, such as

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rapid response, low cost, good selectivity, high sensitivity, portability and on-site determination.^{22,23} However, un-modified electrodes show poor sensitivity, electrode fouling and high overpotential in the detection of ISZ. Therefore, it is necessary to improve the bare electrodes for selective and sensitive determination of ISZ. Due to significant exploration in nanoscience and nanotechnology, various structured nanomaterials have been commonly used to develop efficient electrochemical sensors. More recently, binary metal oxides have been recognized as a benchmark catalyst for electrochemical sensors.

Metal phosphates with different morphologies have attracted much research interest; they can potentially be used as an efficient electrode material due to their unique physicochemical properties.^{24,25} Transition metal phosphates (TMP) significantly contribute to the development of energy storage, electrochemical sensors, energy conversion, photocatalysis and optical sensors.²⁵⁻²⁷ It can be clearly observed that TMPbased nanomaterials have been paid special attention towards electrochemical sensing applications. Recently, Kokulnathan et al. reported the synthesis of nickel phosphate and evaluated its electrochemical performance towards chlorpromazine sensing.²⁶ To date, tin phosphate (SnP) has been very rarely reported in the field of electrochemical sensors. SnP possesses good electrocatalytic activity, low toxicity, environmental stability, and electronic conductivity with a high specific surface area towards energy conversion.²⁸ It is known that the performance of an electrochemical sensor strongly depends on its morphology, crystallites and size of the sensing surface. Thus, designing a nanoscale SnP-based catalyst with a unique nanostructure could provide a promising candidate for ISZ electrochemical detection. Among the investigated strategies, carbon and its derivatives can enhance electrochemical sensing activity due to their highly active sites, rapid electron transfer, good electronic distribution and good conductivity.²⁹ Graphene flakes (GRF) are an important carbonaceous nanomaterial with a two-dimensional lattice network; numerous researchers have greatly focused on GRF due to its large specific area, high electrocatalytic effects, numerous functional effects, biocompatibility, superior mechanical properties and high surface area-to-volume ratio.^{30–37} Due to these emerging properties of GRF, it is tremendously useful for different electrochemical applications. Additionally, GRF offers various functional groups and good conductivity, which have great effects on electrochemical response.38-41 Considering these facts, the fabrication of GRF combined with SnP is an ideal and promising material to modify electrodes for electrochemical sensors.

The aim of the present study is to investigate the electrochemical behavior of SnP incorporated with a GRF nanocomposite for determination of the antituberculosis drug ISZ. The as-prepared SnP/GRF nanocomposite was characterized by field emission scanning electron microscopy, transmission electron microscopy, electron dispersive X-ray spectroscopy, X-ray diffraction and Raman spectroscopy. Interestingly, the SnP/GRF modified electrode showed a wide linear range, very low-level detection, high sensitivity and excellent selectivity towards the determination of ISZ. Moreover, the proposed SnP/GRF sensor delivers an excellent recovery range for the determination of ISZ in biological samples. Our work offers new insights into the electrochemical activity of SnP/GRF nanocomposites and contributes to future clinical applications in the field of ISZ sensing.

2. Experimental section

2.1. Materials

Tin chloride (SnCl₂), disodium phosphate (Na₂HPO₄), urea (CH₄N₂O), isoniazid and all other chemicals were purchased from Sigma-Aldrich and were used without further purification. The supporting electrolyte (0.05 M phosphate buffer solution; PBS) was prepared by mixing 0.05 M Na₂HPO₄ (disodium hydrogen phosphate) and NaH₂PO₄ (monosodium phosphate) solutions and was utilized for all experiments. All other chemicals were of analytical grade, and the required solutions were prepared with de-ionized (DI) water.

2.2. Synthesis of porous tube-like SnP

In a typical synthetic procedure of porous tube-like SnP, 0.1 M SnCl₂ and 0.2 M Na₂HPO₄ were dissolved in 70 mL of DI water, and the solution was stirred continuously for 20 min. After that, 0.5 g CH₄N₂O (in 10 mL of DI water) was slowly added to the above solution with stirring for 1 h. Subsequently, the obtained mixture solution was transferred to a Teflon-lined stainless steel autoclave with a capacity of 100 mL and maintained at 160 °C for 12 h. Later, the stainless steel autoclave was cooled to room temperature, and the obtained products were washed with DI water/ethanol to remove impurities. Finally, the product was heated in a muffle furnace at 550 °C for 4 h.

2.3 Synthesis of SnP/GRF nanocomposite

The SnP/GRF nanocomposite was prepared by simple physical mixing. In a typical recipe, 5 mg of SnP and 3 mg of GRF were dispersed in 1 mL of DMF and ultra-sonicated for 15 min to prepare a homogenous suspension. The as-synthesized SnP/GRF suspension was further examined by physical and electrochemical characterizations. The overall synthesis procedure of the SnP/GRF nanocomposite is shown in Scheme 1.

2.4 Characterization

The X-ray diffraction (XRD) characterization of the as-prepared material was conducted on a PANalytical X-Pert diffractometer measured with Cu-K α radiation (λ = 1.54178 Å). The morphology of the as-prepared material was examined by field emission scanning electron microscopy (FESEM, JSM-7800F) and transmission electron microscopy (Shimadzu JEM-1200 EX) with energy dispersive X-ray spectroscopy (EDS) and elemental mapping. The electrochemical measurements (cyclic voltammetry (CV)) were performed on a CHI 1211C and 750A electrochemical workstation (CH Instruments, USA). A



conventional three-electrode cell system was used in which glassy carbon electrode (GCE) was used as the working electrode (working area = 0.07 cm^2), platinum wire was used as the auxiliary electrode and Ag/AgCl (saturated KCl) was used as the reference electrode. The amperometric measurements were carried out on a PINE instrument with a glassy carbon rotating disc electrode.

2.5 Preparation of modified electrodes

The glassy carbon electrode (GCE) was finely polished with various sizes of alumina slurry and washed with DI water/ ethanol. The as-prepared SnP/GRF nanocomposite suspension was dropped on the pretreated GCE to fabricate SnP/GRF/GCE and dried in air. Subsequently, the dried SnP/GRF/GCE was gently dipped in DI water to remove loosely attached molecules from the electrode. For comparison, SnP/GCE and GRF/GCE were fabricated in a similar way. All the electrochemical experiments were carried out in nitrogen atmosphere at room temperature.

3. Results and discussion

3.1 Physical characterization of the SnP/GRF nanocomposite

The structures and morphologies of the as-synthesized SnP, GRF and SnP/GRF nanocomposite were investigated by FESEM and TEM (Fig. 1 and 2). The FESEM image of SnP shows amazing porous tube-like structures with various sizes and smooth surfaces (Fig. 1(A–C)). The porous tube structure of SnP is favorable for rapid electron/ion transfer and electrocatalytic activity. From Fig. 1(D–F), the GRF exhibits a flake-like structure with very thin and transparent surfaces. Fig. 1 (G–I) shows FESEM images of the SnP/GRF nanocomposite with different magnifications. These images reveal that the

porous tube-like SnP structure was entrapped on the GRF surface, displaying a hierarchical architecture. Furthermore, the in-depth topographical features of the SnP/GRF nanocomposite were scrutinized by TEM. Fig. 2 shows TEM images of (A and B) SnP, (C) GRF and (D) SnP/GRF nanocomposite. As shown in Fig. 2(A and B), SnP has a porous, tube-like structure with smooth surfaces. Fig. 2C shows the TEM image of the 2D GRF nanosheets, which are transparent and wrinkled. The formation of the SnP/GRF nanocomposite is shown in Fig. 2D, which reveals that the SnP was strongly encapsulated on the GRF surface. Further clarification of the formation of the SnP/ GRF nanocomposite was studied by TEM-EDS and element mapping, as presented in Fig. 3. In Fig. 3A, the TEM image shows the SnP/GRF nanocomposite with the corresponding EDS (Fig. 3B) and elemental mapping (Fig. 3(C-F)). The EDS spectrum of the SnP/GRF nanocomposite confirms the existence of Sn, P, C and O elements without any impurities. Elemental mapping analysis of the SnP/GRF nanocomposite was performed, and the obtained results clearly reveal the uniform distributions of Sn (Fig. 3C), P (Fig. 3D), O (Fig. 3E) and C (Fig. 3F) elements in the whole region (Fig. 3A) of the SnP/GRF nanocomposite.

The phases and crystalline structures of the as-synthesized SnP, GRF and SnP/GRF nanocomposite were determined by XRD analysis, as shown in Fig. 4A. The reflection peaks in Fig. 4A(a) were in good agreement with those of SnP₂O₇ (SnP) and matched the standard data of PDF Card no. 01-075-1143.²⁸ Furthermore, no secondary peaks were identified in the XRD spectrum of SnP, which is in agreement with the elemental mapping signal results from FESEM-EDX. The sharp reflection peak at $2\theta = 26.4^{\circ}$ corresponds to the (002) plane of pristine GRF (Fig. 4A(b)).³⁰ Moreover, the reflection peaks of SnP and GRF were observed in the SnP/GRF nanocomposite (Fig. 4A(c)) with good crystallinity.



Fig. 1 FE-SEM images of (A-C) SnP, (D-F) GRF and (G-I) the SnP/GRF nanocomposite.

Fig. 2 TEM images of (A and B) SnP, (C) GRF and (D) the SnP/GRF nanocomposite.

Raman spectroscopy is a very important technique to scrutinize the disorder of carbonaceous materials. Hence, the assynthesized GRF and SnP/GRF nanocomposite were examined by Raman spectroscopy. Fig. 4B shows the Raman spectra of (a) GRF and (b) the SnP/GRF nanocomposite with the appearance of major peaks. The Raman spectrum of GRF shows a weak D band located at 1250 cm⁻¹ and a robust G band located at 1493 cm⁻¹. A strong 2D band was observed at 2647 cm⁻¹, which is attributed to the few layers of GRF.³⁰ The SnP/GRF nanocomposite shows D, G and 2D bands located at 1252, 1486 and 2597 cm⁻¹. In general, the intensity ratio of the 2D band and G band (I_{2D}/I_G) is used to assess the number of layers present in graphene. The ratios of I_{2D}/I_G for GRF and SnP/GRF were found to be 1.77 and 1.75, respectively. Thus, the decrease in I_{2D}/I_G after embedding SnP into GRF implies the place occupation by SnP. The results of the Raman spectra were highly consistent with the XRD patterns and EDX analysis results.

3.2 Electrocatalytic activity of ISZ at different GCEs

Prior to studying the electrochemical oxidation of ISZ, the effects of various suspension amounts of SnP/GRF nanocomposite on the detection of ISZ were examined by CV. Various amounts of SnP/GRF nanocomposite suspension were modified on pretreated GCE, as illustrated in Fig. 5A. It can be clearly seen that the peak current increased when the nanocomposite amount was increased from 3.0 to 5.0 μ L. After that, the anodic current response decreased for nanocomposite amounts above 5.0 μ L. At higher concentrations of SnP/GRF nanocomposite suspension, the anodic peak current of ISZ decreased due to the thickness of the nanocomposite layer and its blocking of mass transportation during the ISZ



Fig. 3 (A) TEM image of the SnP/GRF nanocomposite and its corresponding (B) EDX spectrum and elemental mapping of (C) Sn, (D) P, (E) O, (F) C.



Fig. 4 (A) XRD patterns of (a) SnP, (b) GRF, and (c) the SnP/GRF nanocomposite and (B) Raman spectra of (a) GRF, and (b) the SnP/GRF nanocomposite.

sensing. Therefore, 5.0 µL SnP/GRF nanocomposite was chosen in the GCE fabrication process for further electrochemical studies of ISZ sensing. The electrochemical behaviors of ISZ on various modified electrodes were studied by CV. Fig. 5B shows the CV response with 200 µM ISZ in 0.05 M PBS (pH 7.0) at bare GCE, SnP/GCE, GRF/GCE and SnP/GRF nanocomposite-modified GCE. It was observed that the bare GCE did not show any response to ISZ. The CV current response of SnP/GCE ($I_{pa} = 8.76 \mu A$) increased significantly compared to the bare GCE. Here, the effect of SnP decreases the electron transfer resistance and increases the anodic peak current. When GRF was introduced on the GCE, it exhibited good electrocatalytic performance in terms of increasing the oxidation peak current (I_{pa} = 12.68 µA). Remarkably, the SnP/ GRF-modified electrode displays significant enhancement of the electrochemical oxidation of ISZ, with an anodic peak current of 23.3 µA. SnP/GRF/GCE has strong electron transfer ability, an enormous number of active sites and a large surface area. Moreover, the SnP/GRF-modified electrode can specifically identify ISZ by hydrogen bonding interactions between

the amine groups of ISZ and the phosphates in the nanocomposite. Compared with the former three modified electrodes, the CV behavior of SnP/GRF nanocomposite-modified GCE showed remarkably increased anodic peak current due to its strong synergistic effect, abundant active sites and excellent accessibility towards ISZ electrochemical sensing. Additionally, the anodic peak potentials of ISZ were exhibited at +0.26 V (SnP/GRF/GCE), +0.27 V (SnP/GCE), and +0.29 V (GRF/GCE). Considering the electrochemical response presented in Fig. 5B, a possible oxidation mechanism of ISZ on SnP/GRF/ GCE can be expressed as in Scheme 2. Herein, the electrochemical oxidation of ISZ on the SnP/GRF modified electrode involves equal numbers of $2e^-$ and $2H^+$ pathways. Thus, SnP/ GRF catalyzes the oxidation of ISZ to isonicotinic acid and N₂.

Furthermore, the electrochemical activity of SnP/GRF-modified GCE was investigated in the presence of various concentrations of ISZ from 0 to 320 μ M in 0.05 M PBS (pH 7.0) at a scan rate of 50 mV s⁻¹ (Fig. 5C). From Fig. 5C, it can be obviously observed that the oxidation peak current of ISZ gradually increased with increasing concentration of ISZ from



Fig. 5 (A) The effects of the amount of SnP/GRF nanocomposite suspension loaded on GCE on the anodic peak current of the ISZ sensor. (B) CV performance at different modified electrodes in the presence of 200 μ M ISZ containing 0.05 M PBS (pH 7.0) at a scan rate of 50 mV s⁻¹. (C) CV responses of SnP/GRF/GCE to varying concentrations of ISZ (upto 320 μ M) in 0.05 M PBS (pH 7.0) at a scan rate of 50 mV s⁻¹. (D) The linear calibration plot for the peak current vs. the ISZ concentration.



40.0 to 320.0 μ M. These observations confirm that the SnP/ GRF-modified electrode has good electrocatalytic activity for the detection of ISZ. When the ISZ concentration was increased from 40.0 to 320.0 μ M, the oxidation peak current increased linearly, as shown in Fig. 5D. The line regression equation is y = 0.0754x + 6.9054 with a correlation coefficient of 0.9905. This indicates that SnP/GRF/GCE is the most promising catalyst for the electrochemical sensing of ISZ. The electrochemical characteristics of the SnP/GRF modified electrode towards the detection of ISZ, such as working linear range, limit of detection and sensitivity, are discussed extensively in section 3.3.

3.2.1 Kinetic studies of ISZ on SnP/GRF/GCE. The effects of scan rate on the CV of ISZ at SnP/GRF/GCE were evaluated.

As shown in Fig. 6A, CV measurements were performed at various scanning rates between 40 and 400 mV s⁻¹ in 200 μ M ISZ containing 0.05 M PBS (pH 7.0). The oxidation peak current of ISZ (I_{pa}) increases linearly with scan rate (ν), and this relationship can be described as I_{pa} (μ A) = 0.2367 × ν (mV s⁻¹) + 19.891 (R^2 = 0.9903) (Fig. 6B). This suggests that the electrochemical process on the proposed sensor is a surface-controlled process. For the surface-controlled process, the number of transferred electrons *n* was estimated based on the following eqn (1).

$$I_{\rm pa} = (nQF\nu)/4RT \tag{1}$$

where *Q* is the electron charge, *F* is Faraday's constant, ν is the scan rate, *R* is the gas constant and *T* is the temperature. By substituting these numbers into the above equation, the number of transferred electrons was calculated to be two. Furthermore, a shift of the anodic peak potential was observed with increasing scan rate, and its linear relation was observed by plotting the relation between the peak potential and the log of the scan rate (Fig. 6C). The linear regression equation can be expressed as y = 0.0799x + 0.1289 ($R^2 = 0.9918$) with a slope value of 0.0799, which is very close to zero. This indicates that the electro-oxidation of ISZ at SnP/GRF/GCE has very low kinetics limitations, which is favorable for good electron transfer processes during the electrochemical oxidation of ISZ.



Fig. 6 (A) CV performance of the SnP/GRF nanocomposite in the presence of 200 μ M ISZ containing 0.05 M PBS (pH 7.0) at different scan rates. (B) The linear calibration plot for the peak current of ISZ vs. the scan rate. (C) The linear plot for the anodic peak potential vs. the log of the scan rate. (D) Tafel plot for the log of the oxidation peak current vs. the anodic peak potential measured from the CV profile obtained at 40 mV s⁻¹ for ISZ (200 μ M).

The electron transfer coefficient (α) of the modified electrode can be calculated by eqn (2) for the slope value (Δ). A Tafel plot was drawn using the peak current response from the CV curve obtained at 80 mV s⁻¹ in the presence of ISZ (200 μ M), as shown in Fig. 6D.

$$\Delta = n(1-\alpha)F/2.3RT$$
(2)

where *F* is the Faraday constant, *R* is the gas constant, *n* is the number of transferred electrons and *T* is the temperature. From the Tafel plot (Fig. 6D), the slope value was measured and substituted into eqn (2). Finally, the value of α was calculated to be 0.58. The proposed electro-oxidation process of ISZ at SnP/GRF/GCE is an irreversible reaction. The adsorption amount of ISZ on the surface of SnP/GRFmodified GCE was further calculated by the following eqn (3):

$$i_{\rm p} = n^2 F^2 A \Gamma \nu / 4RT \tag{3}$$

where *n* is the number of transferred electrons, *F* is the Faraday constant, Γ is the surface concentration of the proposed electrode, *A* is the surface area, ν is the scan rate, and *T* is the temperature. The surface concentration of absorbed elec-

troactive species at SnP/GRF/GCE was estimated to be 3.64 \times $10^{-5} \mbox{ mol cm}^{-2}.$

3.2.2 Influence of pH at SnP/GRF/GCE towards ISZ. The effects of pH on the oxidation of 200 µM ISZ at SnP/GRF modified GCE were investigated with various pH values, and the corresponding CV curves are displayed in Fig. 7A. The modified electrode exhibits a peak current response of ISZ, and the anodic peak current gradually increased with increasing pH value in the range from 3.0 to 7.0. However, the maximum peak current was reached at the pH value of 7.0; afterwards, the anodic peak current gradually decreased. Notably, the oxidation peak potential of ISZ shifted towards the positive or negative direction depending on the pH value. The relationship between the peak potential and the pH value from 3.0 to 11.0 was linear, as shown in Fig. 7B. The linear regression equation can be expressed as y = -0.0473x + 0.5915 with the correlation coefficient $R^2 = 0.9965$. This electrochemical study suggests that equal numbers of protons and electrons were transferred during the electrocatalytic reaction towards ISZ detection. Therefore, 0.05 M PBS at pH 7.0 is favored as the supporting electrolyte for further electrochemical detection of ISZ.



Fig. 7 (A) CV response of SnP/GRF/GCE for varying pH values of 0.05 M PBS in the presence of 200 μ M ISZ at the scan rate of 50 mV s⁻¹. (B) The dependence of the peak potential (red) and the anodic peak current (blue) on the pH value.

3.3 Amperometric determination of ISZ

The detection sensitivity of ISZ was measured on the SnP/ GRF modified rotating disc glassy carbon electrode (RDGCE) by the amperometric technique with the addition of different concentrations of ISZ to continuously stirred 0.05 M PBS (pH 7.0) (Fig. 8A). The applied potential of SnP/GRF/RDGCE was maintained at +0.25 V, and the rotation speed was 1300 rpm. It can be clearly seen that each addition of ISZ exhibited a well-defined and sharp peak current response. The anodic peak current of ISZ linearly increases with increasing concentration from 0.01 to 547 μ M and reaches the steady state current within 4 s. The ISZ oxidation current increased linearly in the range from 0.01 to 348 μ M with a correlation coefficient of 0.9975, as shown in Fig. 8A (Inset). The limit of detection (LOD) and sensitivity were calculated to be 1.0 nM and 1.418 μ A μ M⁻¹ cm⁻², respectively. The obtained analytical parameters such as LOD, linear response range and sensitivity were compared with those of previously reported modified electrodes for the detection of ISZ, which are summarized in Table 1. It can be seen that this SnP/GRF modified electrode has better electrocatalytic activity than previously reported modified electrodes. The enhanced electrochemical activity of the SnP/GRF-modified electrode towards ISZ sensing is due to its large surface area, high electron conductivity, rapid electron/proton transfer, syngeneic effects and greater number of active sites. Hence, the SnP/GRF-modified electrode is more suitable for the sensitive electrochemical detection of ISZ.

3.3.1 Selectivity, practical application, reproducibility and stability. In on-site monitoring, selectivity is highly necessary for newly developed electrochemical sensors because many common metal ions, biologically co-active species, nitro-



Fig. 8 (A) Amperometric response of SnP/GRF-modified RDGCE to the addition of ISZ from 0.01 to 540 μ M in 0.05 M PBS (pH 7.0) with applied potential = 0.25 V; rotation speed = 1200 rpm (Inset: the calibration plot of the ISZ concentration and oxidation current). (B) Amperometric response of the proposed SnP/GRF/RDGCE to the addition of 20 μ M ISZ (a) in the presence of excess concentrations of interfering compounds (b-m).

 Table 1
 Comparison of the electrochemical characteristics of previously reported modified electrodes for the detection of ISZ

Modified electrode	LOD (µM)	Linear range (µM)	Sensitivity $(\mu A \ \mu M^{-1} \ cm^{-2})$	Ref.
Ag–P(MMA-co-AMPS) ESP	0.01	0.5 to 150	0.197	8
Co-Fe HCF/MWCNTs	0.08	0.2 to 100	0.1149	9
Au@PtNPs	0.029	0.05 to 100	_	10
PdNPs	0.47	5 to 10	_	11
C-dots@CuFe2O4	0.041	0.1 to 14	—	13
CPE	0.061	0.5 to 40	—	14
ERGO	0.17	2 to 70	3.987	15
PMel/Au	0.08	0.3 to 170	—	17
Nafion-OMC	0.0835	0.1 to 370	—	18
SnP/GRF	0.001	0.01 to 348	1.418	This work

aromatic compounds and environmental pollutants may be present in samples and interfere with the determination of ISZ. Fig. 8B shows the amperometric response of the SnP/ GRF-modified electrode to the successive addition of ISZ (a) and interferential species such as dopamine (b), glucose (c), ascorbic acid (d), uric acid (e), catechol (f), caffeic acid (g), acetaminophen (h), calcium (i), urea (j), 4-nitrophenol (k), diuron (l), and diphenylamine (m). It can be seen that upon the first addition of ISZ, the electrochemical sensor responds rapidly. However, the injection of interferences does not cause any obvious current response. These results indicate that the SnP/GRF-modified electrode is very favorable to construct an ISZ electrochemical sensor with excellent selectivity.

The practical applicability of the as-fabricated SnP/GRF modified electrode was examined using the amperometric method in biological samples. A human urine sample was collected from a healthy person and centrifuged to remove impurities. Samples of human urine, saliva and human serum albumin were determined by the standard addition method.

Table 2 Determination of ISZ in various biological samples

Sample	Spiked (µM)	Detected (µM)	Recovery (%)
Urine	0.00	_	_
	1.00	1.05	105.00
	2.00	1.98	99.30
	3.00	2.95	98.30
Saliva	0.00	_	_
	1.00	0.95	95.00
	2.00	1.90	95.00
	3.00	2.90	96.66
Human serum albumin	0.00	—	_
	1.00	1.10	110.00
	2.00	1.95	97.50
	3.00	3.05	101.70

However, the samples of human urine, saliva and human serum albumin were ISZ-free; therefore, a known concentration of ISZ was spiked into the biological samples, and these ISZ-spiked biological samples were used for the real sample analysis. The recoveries for the assay of different injections of ISZ varied in the range from 95.00% to 110.00% and are summarized in Table 2. These results indicate that the proposed SnP/GRF-modified electrode shows excellent practicability and feasibility to determine ISZ in real sample monitoring. The reproducibility of the SnP/GRF-modified GCE (Fig. 9A) was evaluated in 0.05 M PBS (pH 7.0) containing 200 µM MP at four independent SnP/GRF electrodes by means of CV profiles with a relative standard deviation of 1.53%, suggesting satisfactory reproducibility of the proposed sensor. The storage stability of the SnP/GRF-modified electrode was further explored by detecting ISZ every day for a period of 15 days. As shown in Fig. 9B, the ISZ current response lost only 7.9% of its initial current response after 15 days. These results demonstrate that the SnP/GRF-modified electrode has excellent reproducibility and storage stability towards the electrochemical detection of ISZ.



Fig. 9 (A) The reproducibility (CV response) of SnP/GRF/GCE for four different electrodes in the presence of 200 μ M ISZ containing 0.05 M PBS (pH 7.0) at a scan rate of 50 mV s⁻¹. (B) The storage stability of SnP/GRF/GCE towards detection of 200 μ M ISZ for 15 days.

4. Conclusion

In summary, an electrochemical sensor based on a SnP/GRFmodified electrode for ISZ detection has been successfully developed for the first time. The as-prepared SnP/GRF was characterized by FESEM, TEM, EDX, elemental mapping, XRD and Raman spectra. The SnP/GRF sensor exhibited a LOD of 1.0 nM, a wide linear range from 0.01 to 348 μ M, good sensitivity of 1.418 μ A μ M⁻¹ cm⁻², rapid response, excellent selectivity, good reproducibility and stability. The excellent electrochemical activity of the catalyst in our proposed sensor can be directly attributed to its large surface area, high electron conductivity, rapid electron/proton transfer and syngeneic effects. Therefore, SnP/GRF is the most promising candidate for application in electrochemical sensors to detect ISZ in biological samples.

Conflicts of interest

There are no conflicts of interest to declare.

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