Electronic Supplementary Materials

Boosting the performance of iontophoretic biosensing system with graphene aerogel and Prussian blue for highly sensitive and noninvasive glucose monitoring

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S1: Preparation of the electrode system

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S1: Preparation of the electrode system

The geometry of the electrode system was designed with AutoCAD (Autodesk, San Rafael, CA) and the screen printing formes (Fig. S3) were outsourced for fabrication (Chuangzan Printing Equipment, Hangzhou, China) with the size of 15×25 cm^2. Sequentially, silver ink, carbon ink, silver chloride ink and insulator ink were printed on PET film utilizing a small semi-automatic screen-printing machine-SPC-3050 (HF-Kejing, Hefei, China). After each printing step, the printed PET was placed in drying oven curing for 20 min at 90 ℃, 90 ℃, 70 ℃, 90 ℃ respectively. Hereinto, silver, silver chloride, carbon and insulator ink were purchased from Shenzhen Baojiayi, Jinan Refreshing Electronic, Shenzhen Shengtianfeng Technology Co., Ltd, respectively. Plythylene terephthalate (PET) film was bought from Shanghai Feixia rubber & Plastic Hardware Co., Ltd.

S2: Preparation of the GA@PB composites solution

Firstly, 0.02475 mmol $K_3Fe(CN)_6$ was added into 1mL deionized water stirring 10 min and then 3 mL graphene oxide was dispersed to above-mentioned solution with stirring 1 h to prepare the mixture solution A. Secondly, dissolving 0.3 g LA and 0.05 mmol $FeCl_3 \cdot 6H_2O$ in 3.5 mL deionized water obtained homogeneous mixture solution B. Finally, the GA@PB solution was compounded by adding the mixture solution A to B and then stirring for 10 min.

S3: Preparation of the CTS/GO_x composites solution

Firstly, 0.12 g acetic acid was mixed into 20 mL deionized water by sonic oscillation to prepare 0.1 M homogeneous acetic acid solution and then dissolving 0.1011 g chitosan in above-mentioned acetic acid solution was obtained 0.5 wt.% chitosan solution. Secondly, GO_x solution (30 mg/mL) containing 10 mg/mL BSA stabilizer and chitosan solution was mixed in a 1:1 v/v ratio to prepare CTS and GO_x mixture solution. Finally, 1% glutaraldehyde was added into the mixture solution as cross-linking agent in a 1:10 v/v ratio to obtain CTS/GO_x composites solution.

S4: Calculation method of sensitivity and limit of detection (LOD)

The sensitivity and limit of detection (LOD) was calculated by the following equation (1) and (2), respectively.

$$\text{Sensitivity} = \frac{\Delta I}{\Delta c \cdot \mathrm{A}} = \frac{\mathrm{S}}{\mathrm{A}} \quad \text{* MERGEFORMAT (1)}$$

$$\mathrm{LOD} = \frac{3.3\sigma}{\mathrm{S}} \quad \text{* MERGEFORMAT (2)}$$

where Δc is the change value of the glucose concentrations, ΔI is the change value of the current density with Δc, A is the area of the working electrode of the SPE, $\mathrm{S} = \frac{\Delta I}{\Delta c}$ is the slope value of the calibration curve, σ is the standard deviation of the current response value in PBS without glucose.

S5: Supporting Figure: Fig. S1~S14

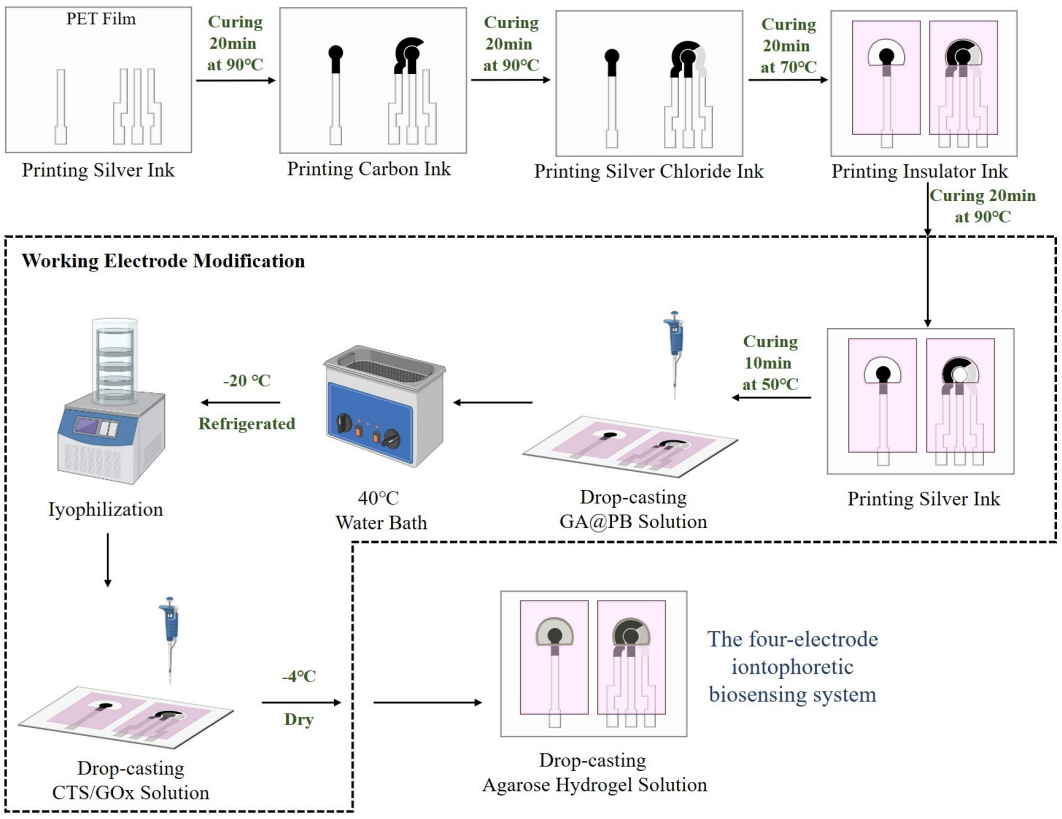

Fig. S1. Fabrication and chemical modification process of the four-electrode iontophoretic biosensing system.

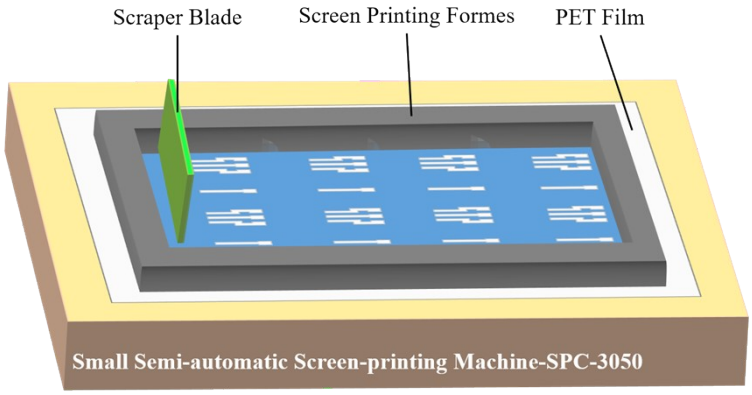

Fig. S2. Schematic diagram of screen-printing technique.

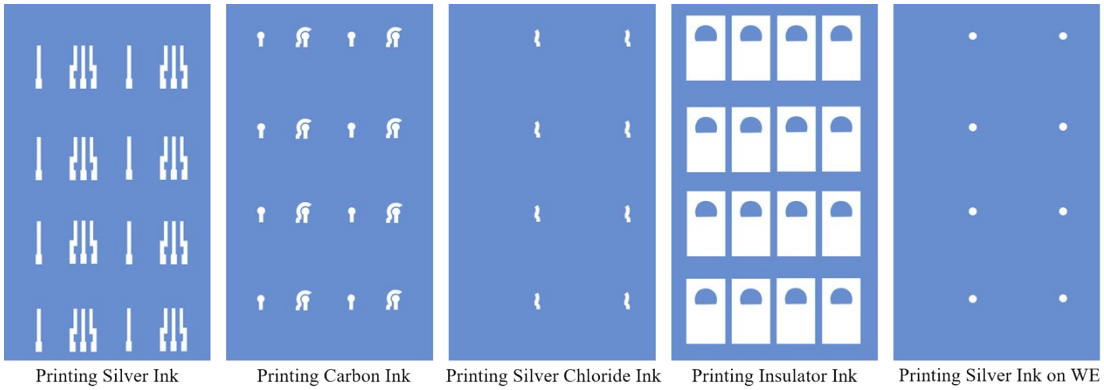

Fig. S3. Schematic diagram of the screen printing formes.

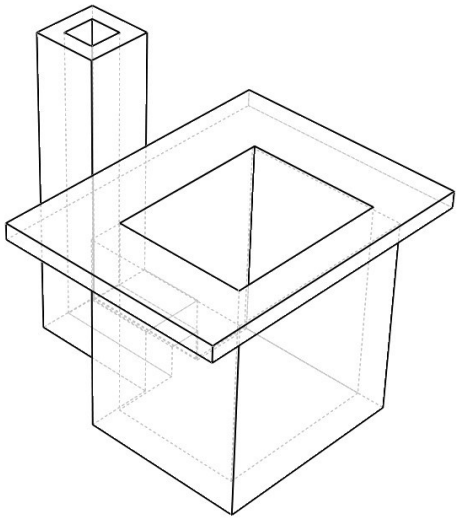

Fig. S4. The diffuse cell of the reverse iontophoretic experimental platform.

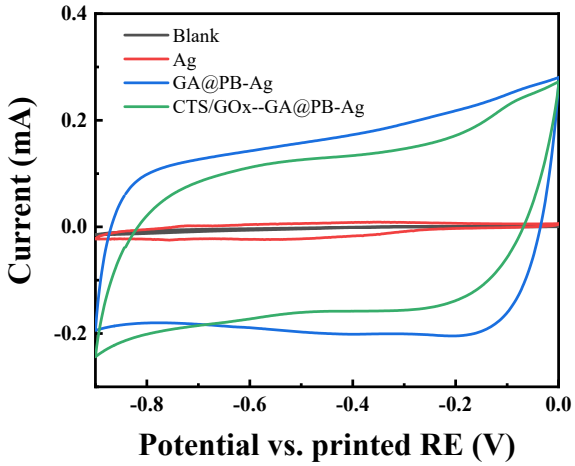

Fig. S5. CV curves of blank electrode (black line), printed Ag electrode (red line), GA@PB on Ag (denoted as GA@PB, blue line) and CTS/GOx-GA@PB on Ag (denoted as CTS/GOx-GA@PB, green line) at a scan rate of 50mV/s in 10 mM PBS.

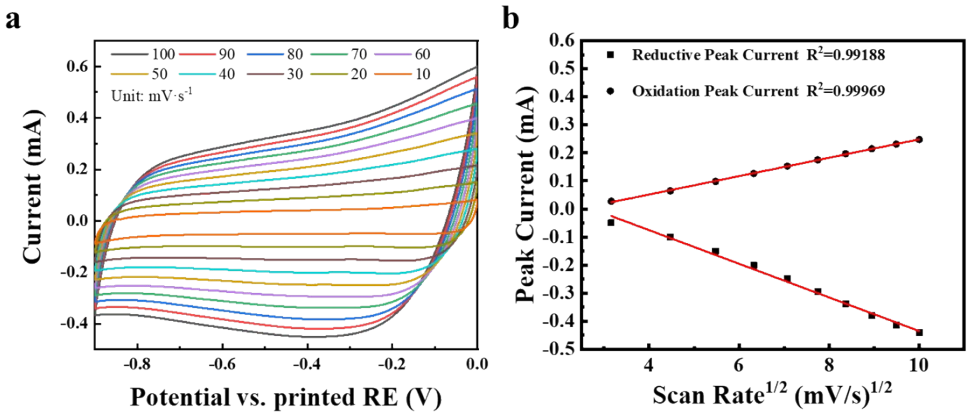

Fig. S6. (a) CV curves of printed Ag electrode modified with GA@PB at different scan rates in 10mM PBS. (b) Calibration curves of anodic and cathodic peak currents versus scanning velocity.

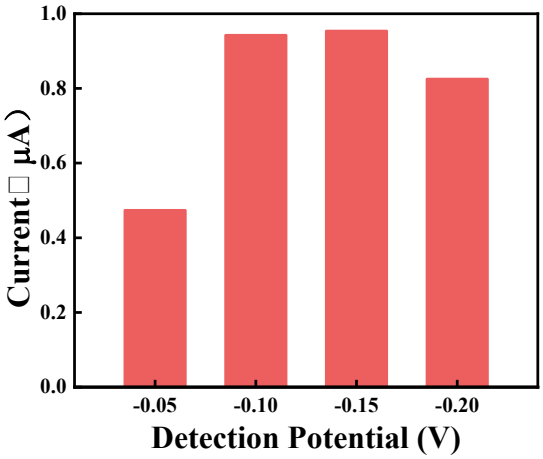

Fig. S7. Current responses of the analytical electrodes modified with CTS/GOx-GA@PB at different detection potential.

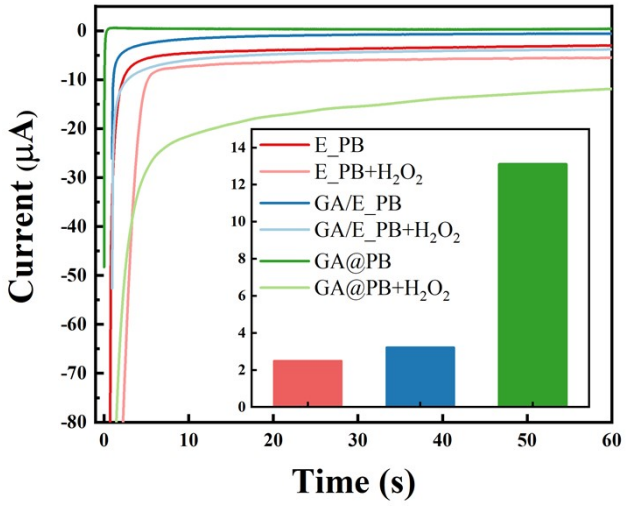

Fig. S8. Current responses of the analytical electrodes modified with electrodeposited PB (E_PB, red line), GA with electrodeposited PB (GA/E_PB, blue line) and GA@PB (green line) over the

same H_2O_2 concentration (1 mM).

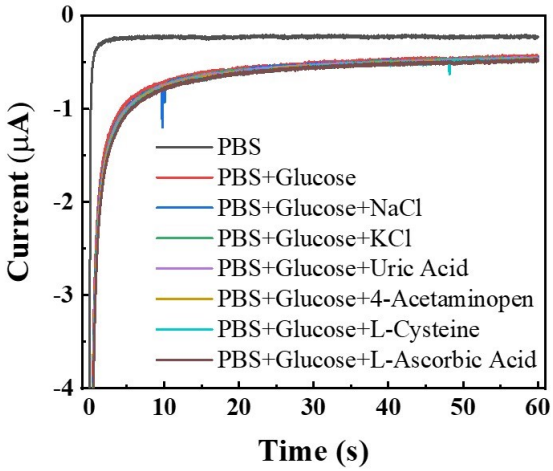

Fig. S9. Interference study. Chronoamperometric response in PBS with the presence of 50 μM glucose and other common coexisting electroactive species.

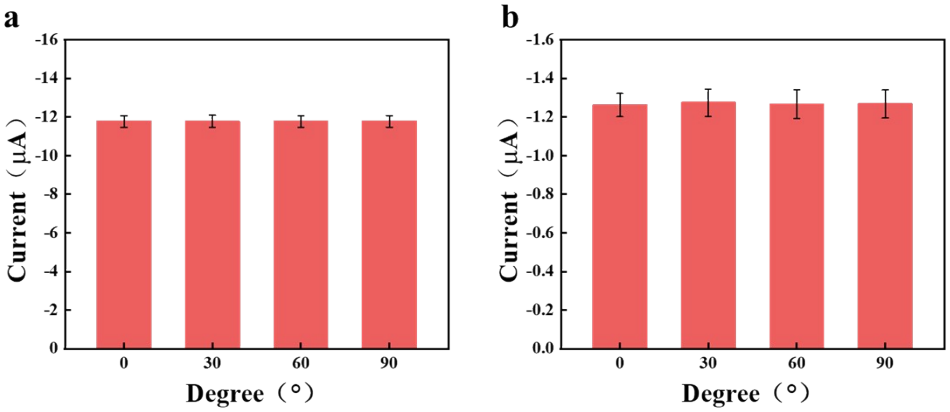

Fig. S10. The flexibility experiment about the analytical electrodes. (a) current responses of the analytical electrodes modified with GA@PB in 1 mM H_2O_2; (b) current responses of the analytical electrodes modified with CTS/GOx-GA@PB in 300 μM glucose.

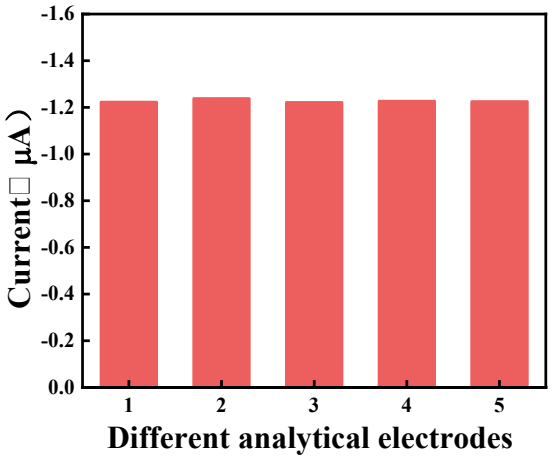

Fig. S11. Current responses of different analytical electrodes in the same glucose concentration (300 μM) by chronoamperometry in 10 mM PBS (pH 7.4)

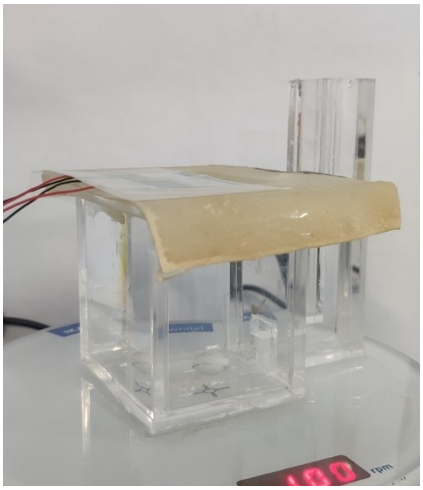

Fig. S12. Digital photograph of the self-developed reverse iontophoresis experimental platform.

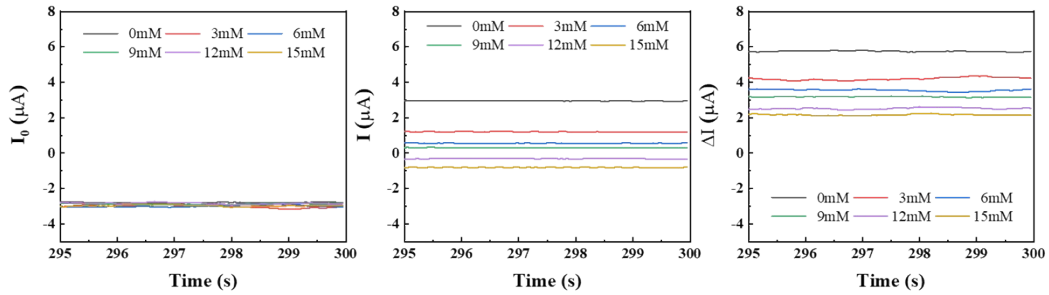

Fig. S13 I, I_0 and ΔI of the iontophoretic biosensing system on glucose sensing ranging from 0-15 mM..

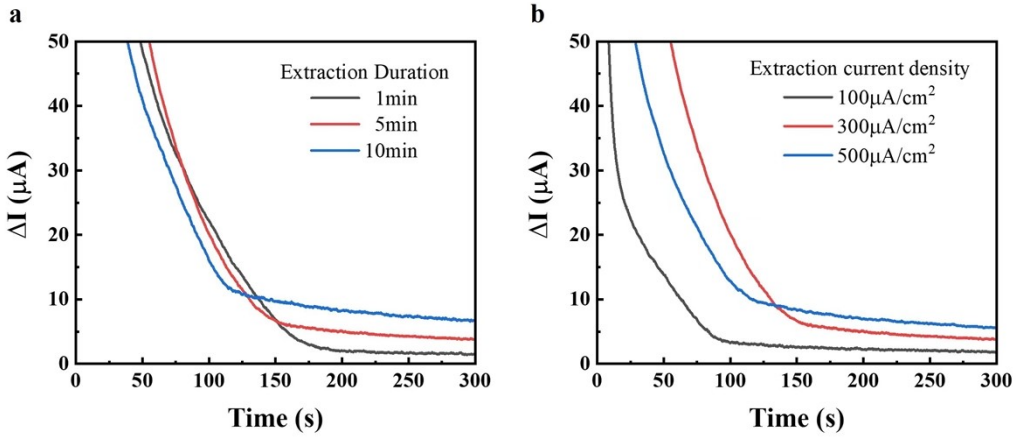

Fig. S14. Influence of extraction duration and current density during reverse iontophoresis on current responses. (A) Influence of extraction duration (extraction current density: 300 μA·cm^{-2}). (B) Influence of extraction current density (extraction duration: 5 min).

Before Detection **After Detection**

Fig. S15. The skin temperature before and after the detection on health volunteer.

S6: Supporting table: Table S1~S2

Table S1 Comparison of previous H_2O_2 biosensors

Modified electrode	Sensitivity ($\mu A\ mM^{-1}\ cm^{-2}$)	Potential (V)	Ref.
GA@PB	**80.30**	**-0.1**	**This work**
PB–FCNF/GCE	35.94	0.12V	[1].
Ni–Fe PBA HNCs	36.13	0.65	[2].
PB cube ink	27.25	-0.05	[3].
PB@Au	39.72	0	[4].

Table S2 Comparison of previous glucose biosensors based on reverse iontophoresis

Modified electrode	Sensitivity of Glucose Detection ($\mu A\ cm^{-2}\ mM^{-1}$)	Sensitivity of ISF Glucose Detection ($\mu A\ cm^{-2}\ mM^{-1}$)	Extraction Current Densities ($\mu A\ cm^{-2}$)	Extraction Durations (min)	Additional auxiliary	Ref.
CTS/GO_x-GA@PB	**15.11**	**1.11**	**300**	**5**	**none**	**This work**
Pt NPs/graphene/Au Nafion/GO_x	2.29	0.38216	150	480 (incessantly)	thermal activated	[5].
PB Ink Nafion/ glutaraldehyde/GO_x	5.66	0.2866	270	480 (incessantly)	none	[6].
PB Ink HEMA/ DMAEMA/EG/GO_x	9.4	0.094	130	480 (incessantly)	none	[7].
PB Ink Tetrahydronaphthalene/ Glutaraldehyde/GO_x	4.635	0.04635	250	30	none	[8].
Ferrocene/GO_x	0.037	0.00545	300	90	none	[9].
MWCNT epoxy/GO_x	0.694	0.021	300	90	none	[10].

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