

## Supplementary Information

### Enzymatic Biofuel Cells based on Protective Hydrophobic Carbon Paste Electrodes: Towards Epidermal Bioenergy Harvesting in the Acidic Sweat Environment

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## 1. Experimental section

### 1.1. Chemicals, reagents and solutions

Lactate oxidase (LOx) (EC 1.1.3.2) was acquired from Toyobo. 1,4-Naphthoquinone (NQ), mineral oil (MO), lactic acid, potassium phosphate monobasic, platinum black (Pt), potassium phosphate dibasic and Nafion<sup>®</sup> were purchased from Sigma-Aldrich. Carboxylic acid functionalized multi-walled carbon nanotubes (MWCNT-COOH) (purity >95%, diameter = 10–20 nm, length = 10–30 µm) were obtained from Cheap Tubes Inc. All chemicals were used without purification. Linear scanning voltammetry was performed in phosphate buffer 1.0 M in pH 7.5, 6.5, 5.5 and 4.0. The pH was adjusted using hydrochloric acid obtained from EMD Millipore Inc. (Massachusetts, USA). Ultra-pure deionized water (18.2 MΩ cm) was employed for all solutions.

### 1.2. Electrochemical measurements

Electrochemical measurements were conducted by a µAutolab Type II controlled by NOVA software (version 1.11) in a single electrochemical cell at room temperature.

The power curves were obtained firstly by performing LSV from open circuit voltage (OCV) to 0 V, at a scan rate of  $5 \text{ mV s}^{-1}$ , and then the current was multiplied by potential. The geometric area of  $\sim 0.071 \text{ cm}^2$  was used to calculate the areal power density (P.D.). All electrochemical measurements for the blank were performed in PBS 1M and the power curves were obtained by adding 30 mM lactic acid in the 1M PBS solution.

### **1.3. Preparation of the carbon-paste bioanode and cathode**

First, 25 mg of MWCNT was added in 50 mL of a concentrated solution containing  $\text{HNO}_3$  and  $\text{H}_2\text{SO}_4$  in the ratio 1:3 (v/v). The suspension was homogenized with magnetic stirring for 12 h at room temperature and filtered with a  $0.45\text{-}\mu\text{m}$  Millipore nylon membrane. The treated MWCNT was washed with DI water, until neutral pH, and dried in a furnace at  $100^\circ\text{C}$  for 3 h<sup>1</sup>.

The lactate bioanode was made by thoroughly grinding 7.5 mg of the acidic treated MWCNT with 3.1 mg NQ in an agate mortar. After well-mixed, 3.3 mg of LOx were added and homogenized, by grinding, followed by addition of  $14 \mu\text{L}$  MO. All grinding steps were performed during 20 min. The resulting paste was manually filled compactly into the cavity of a plastic tube (3.0 mm inner diameter, 7 mm depth). Subsequently, the electrode surface was smoothed on a weighing paper. Only the tip of the carbon-paste tube was exposed to the test solution. Electrical contact was made by using a 0.8 mm diameter conductive stainless-steel wire. The cathode was prepared with a similar protocol used for anode. 7 mg of MWCNTs (non-treated) were mixed with 4 mg of Pt black followed by addition of  $100 \mu\text{L}$  of 2% Nafion<sup>®</sup> in ethanol solution. For a better and uniform distribution, automated mixing of the individual components is recommended to ensure their uniform distribution throughout the paste.

### **1.4. Preparation of the screen-printed bioanode**

Screen-printed carbon electrodes were prepared accordingly with previous work<sup>2</sup>. After printing, three  $1 \mu\text{L}$  layers of a suspension containing  $2 \text{ mg mL}^{-1}$  of acidic treated MWCNT-COOH in 0.2 M of NQ in an organic solution 9:1 (vol/vol) of ethanol/acetone was dropped on the SPE surface (3 mm diameter). Followed by  $5 \mu\text{L}$  of a  $40 \text{ mg mL}^{-1}$

LOx in 10 mg mL<sup>-1</sup> BSA solution dissolved in 0.1 M phosphate-buffer solution (pH 7.4). Finally, 5 µL of glutaraldehyde at 1% in 0.1 M phosphate-buffer solution (pH 7.4) was dropped on LOx layer. Each drop-casting layer was made after the previous casted layer had dried at room temperature. The device was washed with 0.1 M phosphate-buffer solution (pH 7.4).

## References

1. T. C. Canevari, P. A. Raymundo-Pereira, R. Landers, E. V. Benvenutti and S. A. S. Machado, *Talanta*, 2013, **116**, 726-735.
2. A. Barfidokht, R. K. Mishra, R. Seenivasan, S. Liu, L. J. Hubble, J. Wang and D. A. Hall, *Sensors and Actuators B: Chemical*, 2019, DOI: <https://doi.org/10.1016/j.snb.2019.04.053>.