Supporting Information

Long-term cell culture and electrically *in-situ* monitoring of living cells based on polyaniline hydrogel sensor

Rong Wu,^{†,a} Lanlan Li,^{†,b,c} Lijia Pan,^{*,c} Ke Yan, ^c Yi Shi,^c and Liping Jiang^{*,a} and Jun-Jie Zhu^a

a. State Key Laboratory of Analytical Chemistry for Life Science, Chemistry and Biomedicine Innovation Center (ChemBIC), School of Chemistry and Chemical Engineering, Nanjing University, Nanjing 210023, China

b. College of Mechanical and Electrical Engineering, Henan Agricultural University, Zhengzhou 450002, China

c. School of Electronic Science and Engineering, Collaborative Innovation Center of Advanced Microstructures, Nanjing University, Nanjing 210093, China

[†] These authors contributed equally to this work.

Corresponding Authors

* E-mail: jianglp@nju.edu.cn; ljpan@nju.edu.cn

1. Pretreatment of substrate electrodes.

Before use, CC should be cleaned by ultrasonic treatment for 15min in ethanol and water in succession to remove dust, some organic matter, and by-product carbon sphere. Although the purchased CC was hydrophilic, the shown performance was still poor and needed to undergo hydrophilic treatment once again by immersed in a nitric acid solution, just like the CA results shown. Next, CC was rinsed thoroughly by DI water and dried under nitrogen. Besides, the ITO slices were cleaned by ultrasonic treatment in acetone, 1 M NaOH of water/ethanol mixture (1:1, v/v), and water in succession for washing and hydrophilic treatment. The glassy carbon electrode (GCE) was prepared by traditional treatment.

2. SEM characterization of CC/PAni/Pt NPs/cell biointerfaces.

For exploring the hydrogels/cell interfaces, SEM characterization was performed. After captured cells, the electrodes were dipped into glutaraldehyde solution for cell immobilization. Then, the dehydration was reached with the different concentration alcohol aqueous solution (dipped into 0, 25%, 50%, 75%, 95%, 100% alcohol aqueous solution for 10min in succession). Before SEM testing, the electrode must be pretreated with electron beam evaporation for enhancing the conductivity.



Figure S1. The EC performance and mechanical stability of PAni hydrogel on three kinds of substrate electrodes. (a-c) CVs of CC/PAni electrode, GCE/PAni electrode and ITO/PAni electrode. (d) Photographs of the electrodes after multiple purification and EC tests.



Figure S2. (a) Photographs of the CP/PAni electrode. (b, c) SEM images of CP at different dimensions.



Figure S3. SEM image shows the unique 3D structure of CC. Compared with the flat structure, it is favorable for the formation and adsorption of PAni hydrogel on the electrode surface.



Figure S4. Morphological characteristics. SEM images of PAni hydrogels synthesized on a) CC with bigger gap, b) CP, and c) flat base. The former exhibits more flexibility and micron size pores, suitable for sensor fabrication and mass transfer. The insets show the photograph of different CC/PAni electrodes.



Figure S5. Contact angle analysis. (a) CC/PAni. (b) ITO/PAni. (c) CC with hydrophilic treatment. (d) ITO with hydrophilic treatment.



Figure S6. Size distribution of the Pt NPs deposited on the PAni nanofibers.



Figure S7. Feasibility evalution. (a) EIS of PAni purified in different solutions. (b) SEM image of PAni purified in 0.01 M PBS. (c) SEM image of PAni purified in RPMI 1640 medium. (d) CVs of CC/PAni/Pt NPs electrode in 0.01 M PBS and 0.1 M PBS. (e) Cell Viability of MCF-7 cells cultured with PAni, PAni/Pt NPs and PAni/AuNPs for 24 h. The scale bar is 1 µm.



Figure S8. Nitrogen adsorption–desorption isotherm of PAni hydrogels purified in (a) DI water and (b) 0.01 M PBS. The insets show the pore size distribution of PAni hydrogel.



Figure S9. EC performance evaluation of the CC/PAni/Pt NPs electrode after bending. a) CV responses of CC/PAni/Pt NPs electrode in 0.01 M PBS with different bending degrees (θ =0°, 30°, 60°, 90°, 120° and 150°). b) CV responses of CC/PAni/Pt NPs electrode before and after bending treatment when bending degree is zero. C) Amperometric responses of the CC/PAni/Pt NPs electrode to different bending treatment in 0.01 M PBS at an applied potential of -0.7 V.



Figure S10. Recovery capability evaluation of a) non-optimized and b) optimized CC/PAni/Pt NPs electrode in H_2O_2 solution. (c) High-resolution XPS of non-optimized CC/PAni/Pt NPs electrode. for Pt 4f with just Pt⁰ (Pt 4f_{7/2} at 71 eV, Pt 4f_{5/2} at 74.3 eV), and partially oxidized Pt²⁺ (Pt 4f_{7/2} at 72.5 eV, Pt 4f_{5/2} 75.8 eV), which limited the EC performance of H_2O_2 detection.



Figure S11. The relationship between current responses and H_2O_2 concentration. The inset shows the magnified view with H_2O_2 concentration from 10 μ M to 10 mM.



Figure S12. CLSM images of MCF-7 cells and K562 cells cultured with CC/PAni/Pt NPs electrode for 24 h stained by calcian-FAM (green) and PI (red).



Figure S13. CLSM images of NIH-3T3 cells cultured on CC/PAni/Pt NPs electrode for (a) 24 h, (b) 48 h, and (c) 72 h stained by Calcein-AM and PI.