

Supporting Information

Mechanically Stabilized UiO-66-NH₂/MB Screen Printed Carbon Electrode for High Performance Electrochemical Ratiometric of Quantification miR-21-5p

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Materials and reagent. DNA Marker (25-500 bp), 4SGelblue, Acryl/Bis 40% Solution (37.5:1), 50X TBE Buffer, 5X TBE Buffer, 6X DNA Loading Dye, TBE buffer, TAE buffer, Premixed Powder (1X), 1,2-bis(dimethylamino)-ethane (TEMED) and all DNA sequences were synthesized and purified by Sangon Biotechnology Co., Ltd. (Shanghai, China). The chloroauric acid ($\text{HAuCl}_4 \cdot 4\text{H}_2\text{O}$) was obtained from Beijing Huawei Ruike Chemical Co., Ltd. (Beijing, China), Sodium phosphate dibasic dodecahydrate ($\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$) and sodium phosphate monobasic dihydrate ($\text{NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O}$) were obtained from Xilong Chemical Industry Co., Ltd. (Shanghai, China). Benzoic acid ($\text{C}_7\text{H}_6\text{O}_2$) was purchased from Lingfeng Chemical Reagent Co., Ltd. (Shanghai, China). Zirconium tetrachloride (ZnCl_4) was bought from Yi En Chemical Technology Co., Ltd. (Shanghai, China). N, N-dimethylformamide ($\text{C}_3\text{H}_7\text{NO}$) was gotten from Sinopharm Chemical Reagent Co., Ltd. (China). 2-amino-terephthalic acid ($\text{C}_8\text{H}_7\text{NO}_4$) was acquired from Anhui Zesheng Technology Co., Ltd. (Anhui, China). Methylene blue ($\text{C}_{16}\text{H}_{18}\text{ClN}_3\text{S} \cdot 3\text{H}_2\text{O}$) was supported by Tianjin Institute of Chemical Reagents. (Tianjin, China). Methyl alcohol (CH_4O) was ordered from Wuxi Yasheng Chemical Co., Ltd. (Wuxi, China). 6-mercapto-1-hexanol (MCH) was bought from TCI Chemical Industry Development Co., Ltd. (Shanghai, China).

Apparatus. The data of XRD was acquired by a Zelss Sigma300 (German) diffractometer. The polyacrylamide gel electrophoresis (PAGE) and agarose gel electrophoresis (AGE) were imaged with a BIO-RAD ChemiDoc XRs (Tanon, China). The electrode surface conductivity and electrochemical signal were determined on CHI660D (Chen-Hua, China). Fourier transform infrared (FT-IR) spectra were recorded on a Nicolet iS5 spectrophotometer using the KBr pellet technique. UV-vis spectra were performed on a LAMBDA950 spectrophotometer (PerkinElmer, USA). Scanning electron microscopic (SEM) images were obtained from a Zelss Sigma300 scanning electron microscope.

Polyacrylamide gel electrophoresis

12% polyacrylamide gel electrophoresis was employed to verify the feasibility of the signal amplification strategy. Electrophoresis was performed in 1×TBE buffer at 150 V for 45 min. Preparation of samples by mixing 12 μ L reaction product with 3 μ L 6×loading buffer. After staining with 4SGelblue, the gel was imaged using the Tanon Imager (Tanon MINI SPACE 1000).

Electrochemical detection settings

All electrochemical experiments were performed on a CHI660D electrochemical workstation for cyclic voltammetry (CV), electrochemical impedance spectra (EIS), amperometric *i-t* and differential pulse voltammetry (DPV) detections. Electrochemical experiments were performed on a three electrodes system with a UiO-66-NH₂-MB/SPCE as working electrode, a platinum electrode as the counter electrode, and a saturated calomel reference electrode (SCE) as the reference electrode. The CV used for verification of Au electrode surface modification was scanned at a rate of 100 mV/s from -0.6 V to +0.8 V. The DPV used for characterization of I_{FC}/I_{MB} signal was scanned from -0.4 V to 0.4 V. Error bars showed the standard deviation of three individual tests.

Optimization of the ratio of H₁: miR-21-5p (T), H₁: miR-21-5p (T):H₂ and H₁: miR-21-5p (T): H₂: H₃

CHA, the simplification strategy of this research, plays an important role in the performance of the constructed biosensor. To improve the sensitivity of the electrode, the concentration of DNA and miR-21-5p was optimized in this study. In which, H₁: T=5:1, 2:1, 1:1, 1:2, 1:5, H₁+T:H₂=2:1, 1:1,1:2, 1:3, H₁+T+H₂:H₃=2:1, 1:1, 1:1.5, 1:2, 1:3 respectively. The reaction was carried out at 37 °C for 2 h, and then terminated in a refrigerator at 4 °C for 1 h.

Fabrication the electrode for miR-21-5p detection

In order to explore the construction of UiO-66-NH₂-MB/AuNPs/MCH/H₁/SPCE and its detection ability of miR-21-5p, CV curves of [Fe(CN)₆]^{3-/4-} was used to study the characteristics of the modified electrode.

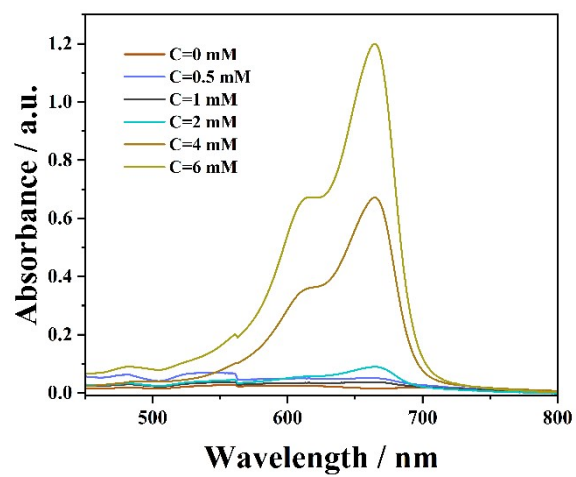


Figure S1. UV-vis spectra of the optimization of the concentration of MB encapsulated in UiO-66-NH₂.

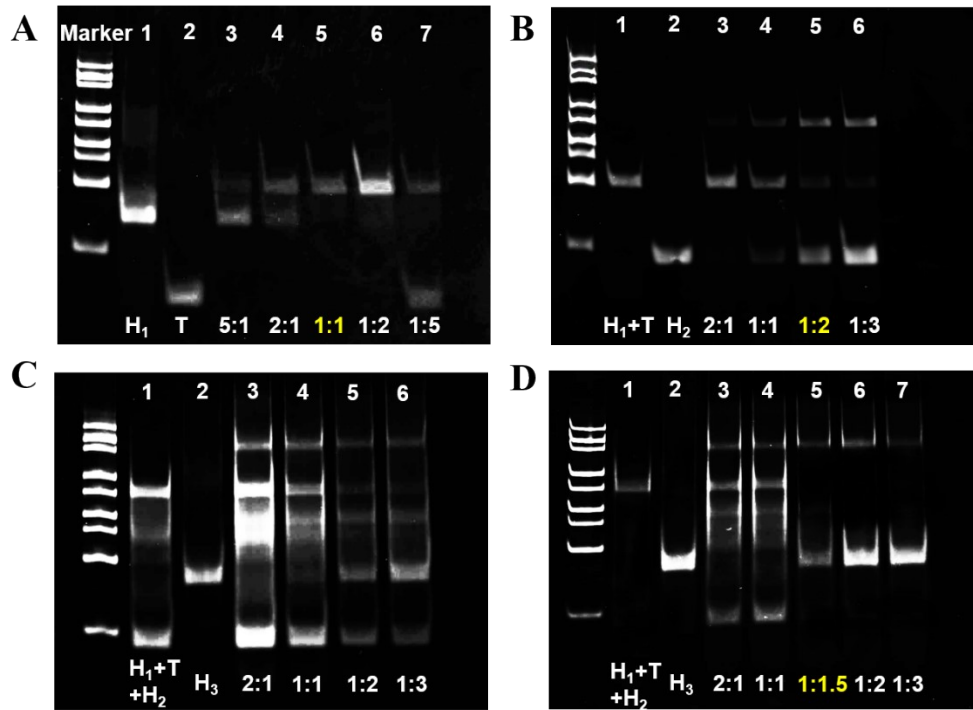


Figure S2. 12% native-PAGE images of the concentration ratios of H_1/T (A), $H_1/T/H_2$ (B), $H_1/T/H_2/H_3$ (C-D).

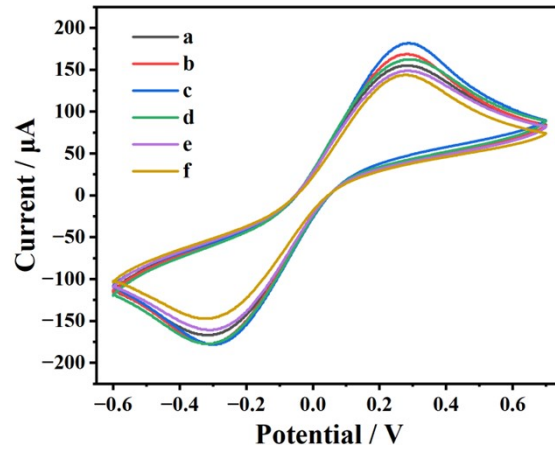


Figure S3. CV curves of $[\text{Fe}(\text{CN})_6]^{3-/4-}$ at the bare screen printed carbon electrodes (SPCE) (a), UiO-66-NH₂-MB/SPCE (b), UiO-66-NH₂-MB/AuNPs/SPCE (c), UiO-66-NH₂-MB/AuNPs/MCH/SPCE (d), UiO-66-NH₂-MB/AuNPs/MCH/H₁/SPCE (e), UiO-66-NH₂-MB/AuNPs/MCH/H₁/SPCE+ miR-21-5p+H₂-Fc+H₃-Fc (f) in 0.1 M KCl solution.

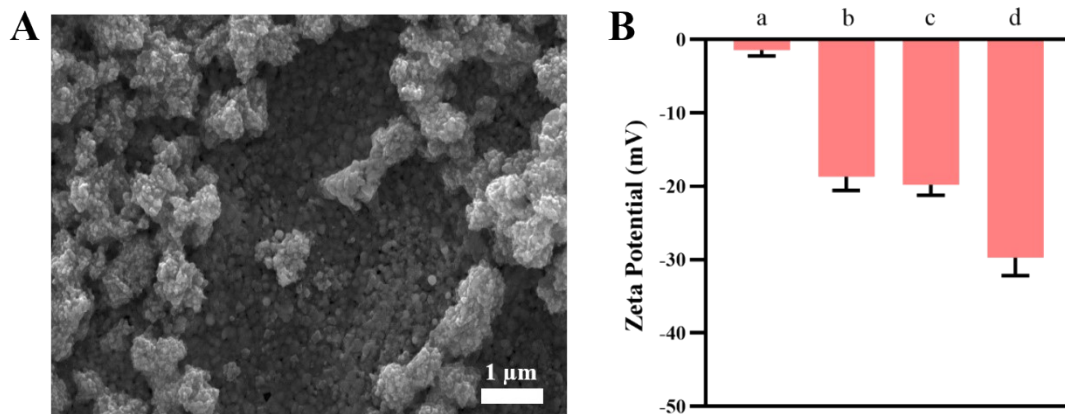


Figure S4. A) SEM image after deposition of AuNPs on UiO-66-NH₂-MB SPCE surface. B) Zeta potential measurements of MB (a), UiO-66-NH₂ (b), UiO-66-NH₂-MB (c) and UiO-66-NH₂-MB/AuNPs (d).

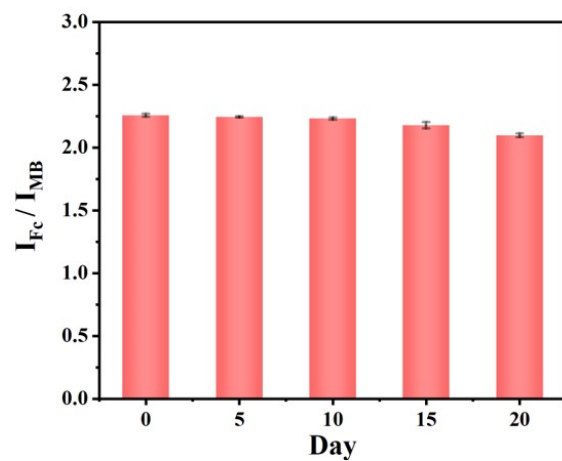


Figure S5. Stability testing of mechanically stabilized UiO-66-NH₂-MB screen-printed carbon electrodes for the detection of miR-21-5p on different days.