Fabrication of high-performance cell-imprinted polymers based on AuNPs/MXene composites via metal-free visible-light induced ATRP

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Fig. S1 The microscope image of yeast cells on a cell counting plate. The red box was the counting area

Fig. S1 was the microscopic image when the yeast cell solution was dripped onto a cell counting plate. There were two cells counting chambers on the cell counting plate. One counting chamber was composed of 25 large squares, and each large square was divided into 16 small squares, such as the red box in Fig. S1, thus, each counting chamber was made up of 400 small squares. The side length of the counting chamber was 1 mm, so the area of the counting chamber was 1 mm². After the cover glass was covered on the surface of the counting plate, the height of the counting chamber was 0.1 mm³. Typically, five large squares (80 small squares) were selected as the counting areas, as shown in Fig. S1. The amount of cells in these squares need to be counted carefully under the ordinary optical microscope. By using the followed formula, the amount of cells per milliliter could be calculated.

$$A = \frac{B}{80} \times 400 \times 10 \times 1000C$$

Where A was the amount of cells per milliliter, B was the number of cells in the counting areas, C was the multiple of cells diluted. For example, 6 μ L 0.4 mg/mL yeast cell solution was dripped onto the counting

plate, the value of C was 1, by counting the number of the cells on the image three times, the average B was 200 cells, A was 1.0×10^7 cells/mL by calculating. That was to say, the number concentration of 0.4 mg/mL yeast cell was 1.0×10^7 cells/mL.



Fig. S2 The microscope image of yeast cells after staining by methylene blue

Methylene blue (MB, 0.01g) and trisodium citrate dihydrate (2 g) were dissolved in ultrapure water (100mL) to achieve the staining solution. Then the solution was mixed with an equal volume of the yeast sample ($\sim 2 \times 10^7$ cells/mL in PBS). After dying for five minutes, the mixture was loaded into a glass slide. The samples were examined microscopically under visible light. As displayed in Fig. S2, the dead yeast cells were dyed blue, active yeast cells were pale yellow. This might be attributed to a sort of reductase in active yeast cells, which could reduce methylene blue. However, the reductase in dead yeast cells was inactivated and did not decolorize MB, so the dead yeast cells was dyed blue.¹



Fig. S3 The photographs (A) of electrode (For observation, the concentration of fluorescein in the polymerization solution was magnified 10 times) and the preparation scheme (B) of microreactor.



Fig. S4 The EIS (A) characterization of the stepwise modified electrodes (1-bare GCE, 2-AuNPs/Ti₃C₂T_x/GCE, 3-Br/AuNPs/Ti₃C₂T_x/GCE, 4-Polymer/AuNPs/Ti₃C₂T_x/GCE, 5-CIPs/AuNPs/Ti₃C₂T_x/GCE) in PBS containing 5 mmol/L [Fe (CN)₆]^{3-/4-} and 0.1 mol/L KCl. (B) The "on-

off" experiment. (Yellow regions were the visible light "on" and white regions were the visible light "off").



Fig. S5 The CV curves of bare GCE (A) and AuNPs/Ti₃C₂T_x/GCE (B) at different scan rates (curves 1 to curve 7 were 10, 20, 40, 60, 80, 100 and 120 mV/s). (C) Plots of peak current with square root of scan rates (red-GCE; black-AuNPs/Ti₃C₂T_x/GCE)

In order to explore the effect of AuNPs/Ti₃C₂T_x composites on the surface area of the electrode, bare GCE electrode and AuNPs/Ti₃C₂T_x/GCE electrode were characterized by CV at different scanning rates in Fig. S5. According to the Randles-Sevcik equation, $I_p = 2.69 \times 10^5 n^{3/2} AD^{1/2} Cv^{1/2}$, when *n*, *D*, and *C* were the same, the slope of the linear equation ($I_p vs. v^{1/2}$) was proportional to the effective area (*A*) of the electrode.⁴² By calculation, the slope of the AuNPs/Ti₃C₂T_x/GCE electrode was approximately 1.75 times that of the bare GCE, further proving that the actual effective surface area of GCE was enlarged by AuNPs/Ti₃C₂T_x composites.



Fig. S6 The DPV signal response of CIPs/AuNPs/Ti₃C₂T_x/GCE (A), CIPs/AuNPs/GCE(B) and AuNPs/Ti₃C₂T_x/GCE (C). 1-before, 2-after incubating in yeast cells solution $(1.00 \times 10^4 \text{ cells/mL})$



Fig. S7 The effect of illumination time (A) on the performance of Polymer/AuNPs/Ti₃C₂T_x/GCE, the ratio of MAA to MBA (B) and the cells concentration (C) on the performance of CIPs/AuNPs/Ti₃C₂T_x/GCE.

Materials	Analyte	Detection	Linear range	Detection	References	
		method	(cells/mL)	(cells/mL)	Kelefences	
Marine-mussel-inspired			1 1 0 2			
polydopamine (PDA)	yeast cells	(PDA)	Chronopotentio	1×10 ² ~	50	2
coating		metry	1.5×10 ³			
MIPs based on		Impedance	$3 \times 10^{1} \sim$	30	3	

Table S1 Comparison of CIPs/AuNPs/Ti₃C₂T_x/GCE and similar sensors *

polyurethane (PU)	spectroscopy	1×10 ⁵		
MIPs based on screen-	Thermal			
printed carbon	resistance	$1.0 \times 10^2 \sim 1.0 \times 10^7$	22	4
electrodes (SPCEs)	method			
MIP coated quartz crystal	Fluoroscopco	$1 \times 10^4 \sim 1 \times 10^5$	1×10 ⁴	5
microbalances (QCM)	Thorescence			
CIPs/AuNPs/Ti ₃ C ₂ T _x /GCE	DPV	$1.00 \times 10^2 \sim$ 1.00×10^9	20	This paper

*For facilitate comparison, the data units were converted





Analyze	Original sample concentration [*] (cells/mL)	Added (cells/mL)	Detection concentration	Recovery (%)	RSD
			(cells/mL)		(70)
Yeast cells	9.00×10 ³	9.00×10 ³	1.79×10 ⁴	99.6	3.71
		1.80×10^{4}	2.66×10 ⁴	96.9	3.61
		4.50×10 ⁴	5.44×10 ⁴	104.8	4.97

Table S2 Detection of yeast cells in real samples (n=3)

*The concentration of yeast cells in SBS was detected by cell counting method.

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