Supporting Information

Molecularly Imprinting on PtPd Nanoflowers for Selective Recognition and Determination of Hydrogen Peroxide and Glucose

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Method for kinetic analysis

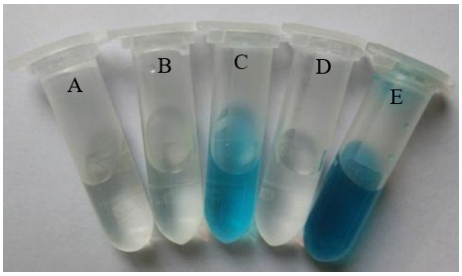
The steady-state kinetic assays with TMB or ABTS as the substrate were performed using 50 μL (0.1 mg/mL) of nanozymes (PtPd NFs, NIP-PtPd NFs and T-MIP-PtPd NFs) containing various concentrations of TMB (0.1875, 0.2, 0.25, 0.3, 0.35, 0.375, 0.5 and 0.75 mM) or ABTS (0.0125, 0.025, 0.05, 0.0625, 0.125, 0.2, 0.25 and 0.3 mM) and a constant concentration of 10 μL H_2O_2 (100 μM), the above solution were added to 940 μL acetate buffer solution (0.1 M, pH 4) The apparent kinetic parameters were calculated using Lineweaver-Burk plots of the double reciprocal of the Michaelis-Menten equation,

$$\frac{1}{v}=\frac{K_m}{V_{max}}\ (\frac{1}{[S]}+\frac{1}{K_m})$$

where V is the initial velocity, Vm represents the maximal reaction velocity, [S] corresponds to the concentration of substrate, and K_m is the Michaelis-Menten constant. The catalytic constant value (k_{cat}) is used to measure the enzymatic catalytic activity.

Real sample analysis

The human serum samples were obtained by Tianjin institute of hematology (Tianjin, China). For glucose analysis in serum samples, a standard addition experiment was performed. The serum sample was diluted by PBS (0.1 M, pH 7.0), the different concentration of glucose were spiked to each 5.0 mL of diluted serum samples and then analyzed.



F

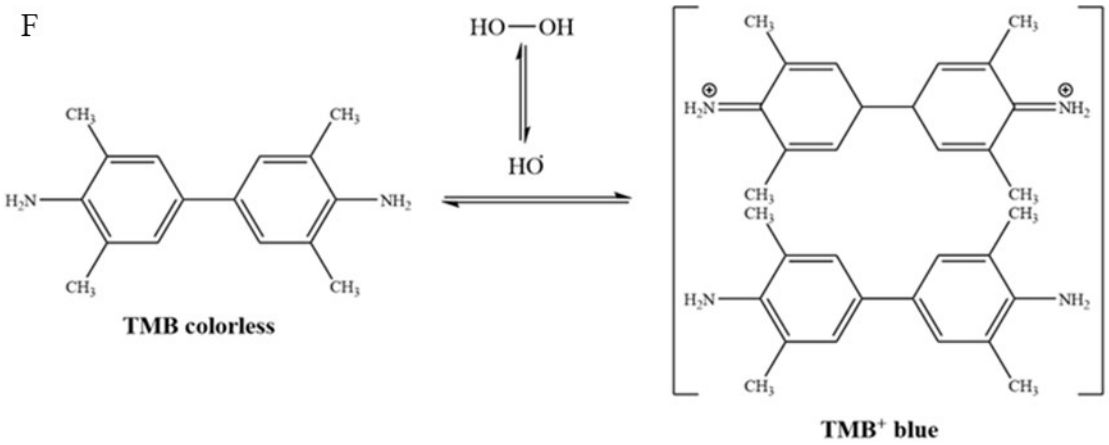
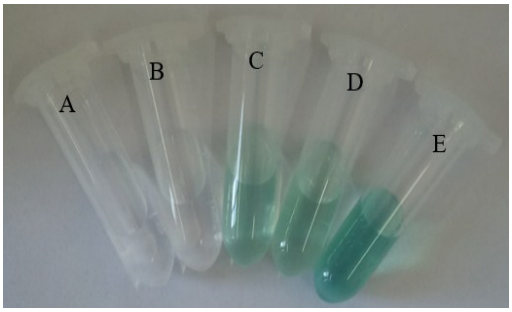


Fig. S1 Color evolution of TMB in different reaction systems: (A) HAc-NaAc buffer, (B) HAc-NaAc buffer + H_2O_2 + T-MIP-PtPd NFs, (C) HAc-NaAc buffer + H_2O_2 + TMB + PtPd NFs, (D) HAc-NaAc buffer + H_2O_2 + TMB, (E) HAc-NaAc buffer + H_2O_2 + TMB + T-MIP-PtPd NFs at 25℃ for 30 min (the pH of HAc-NaAc-HAc buffer is 4.0), and (F) Corresponding reaction for H_2O_2 reduction with TMB.



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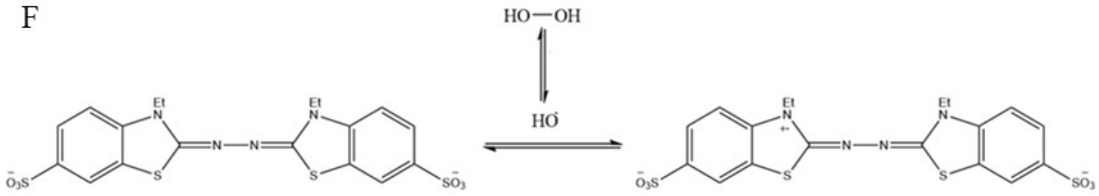


Fig. S2 Color evolution of ABTS in different reaction systems: (A) HAc-NaAc buffer, (B) HAc-NaAc buffer + H_2O_2 + T-MIP-PtPd NFs, (C) HAc-NaAc buffer + ABTS + H_2O_2 +T-MIP-PtPd NFs, (D) HAc-NaAc buffer + H_2O_2 + ABTS+NIP-PtPd NFs, (E) HAc-NaAc buffer + H_2O_2 + ABTS+ PtPd NFs at 25℃ for 30 min (the pH of HAc- NaAc buffer is 4.0), and (F) Corresponding reaction for H_2O_2 reduction with ABTS.

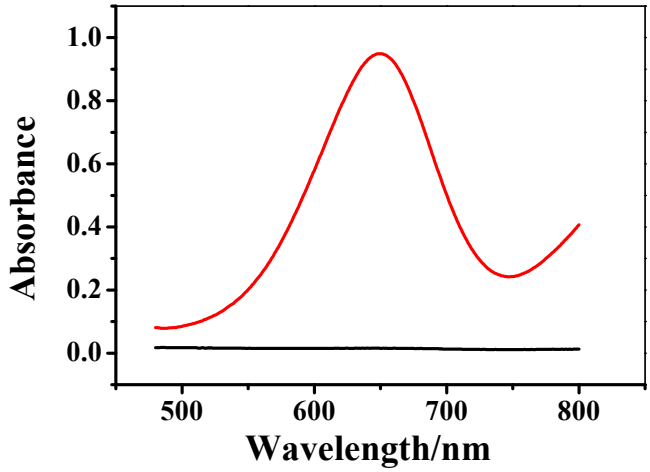
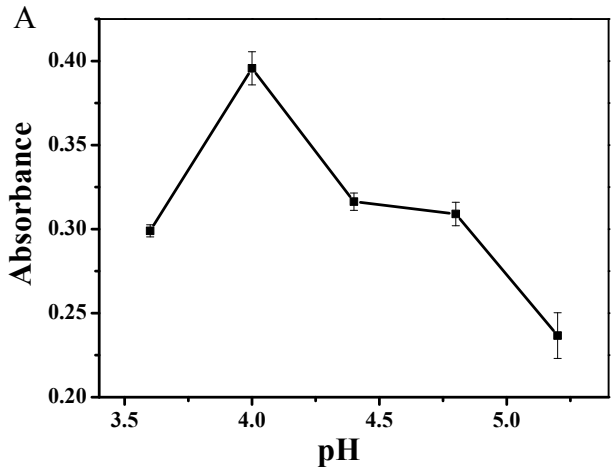
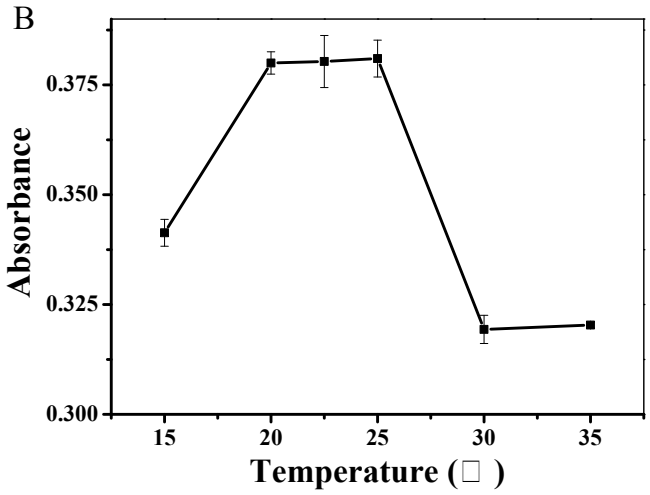


Fig. S3 The UV-vis absorption spectrum of different reaction systems: (red line) HAc-NaAc buffer +TMB+ H_2O_2 + T-MIP-PtPd NFs and (black line) HAc-NaAc buffer + H_2O_2 + T-MIP-PtPd NFs.

Optimization of experimental parameters





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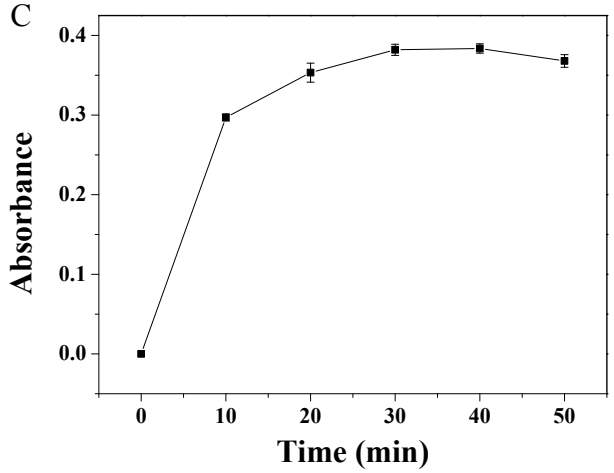
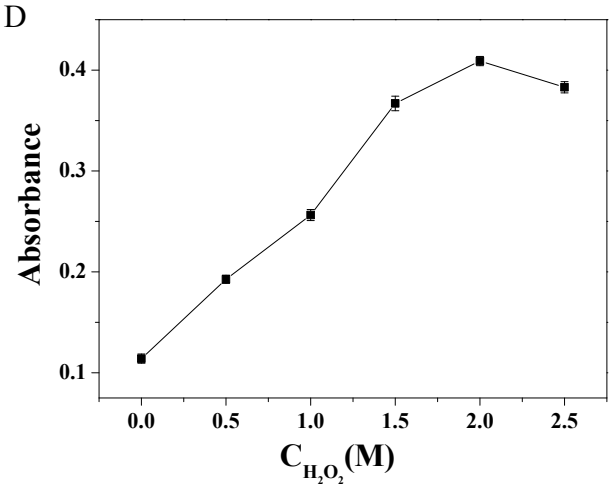
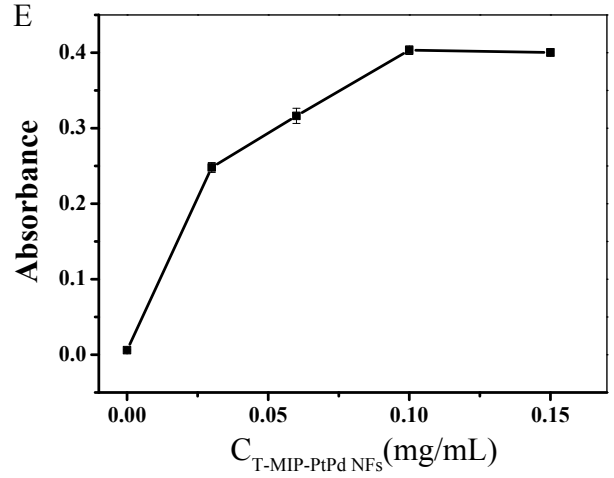

Absorbance
0.0
0.1
0.2
0.3
0.4
0
10
20
30
40
50
Time (min)

D


Absorbance
0.1
0.2
0.3
0.4
0.0
0.5
1.0
1.5
2.0
2.5
$C_{H_2O_2}$(M)

E


Absorbance
0.0
0.1
0.2
0.3
0.4
0.00
0.05
0.10
0.15
$C_{T\text{-}MIP\text{-}PtPd\ NFs}$(mg/mL)

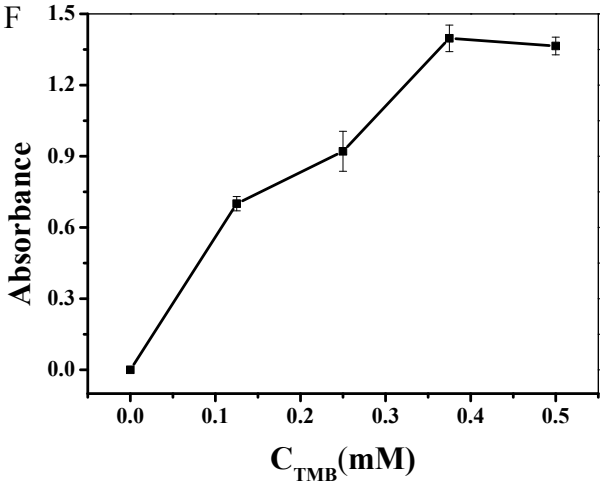


Fig. S4 Effect of (A) pH, (B) temperature, (C) time, (D) H_2O_2 concentration, (E) quantity of T-MIP-PtPd NFs and (F) TMB concentration on peroxidase-like activity of T-MIP- PtPd NFs.

In order to achieve an optimal colorimetric effect, the effect of pH, temperature, reaction time, H_2O_2 concentration, and quantity of T-MIP-PtPd NFs on relative activity were investigated. The optimal catalytic activity of T-MIP-PtPd NFs was obtained under the following optimal conditions: pH 4.0, 25 °C, 30 min, 0.1 mg/mL of T-MIP- PtPd NFs, 0.35 mM of TMB, 2.0 M of H_2O_2.

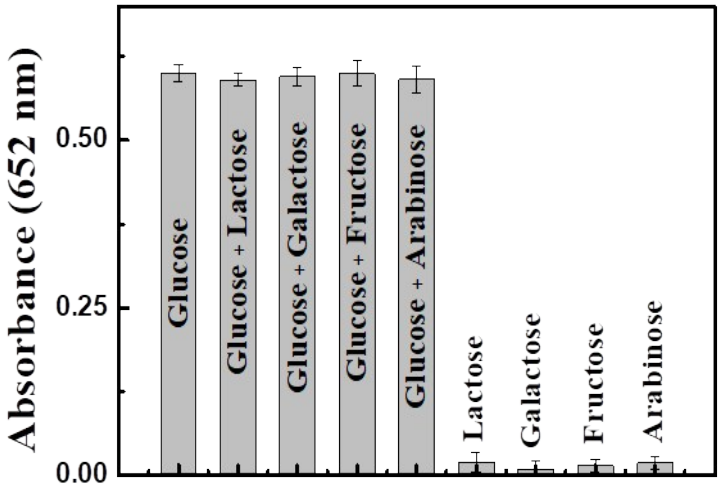


Fig. S5 The selectivity of the colorimetric method for glucose detection (from left to right: 3 mM glucose, 3 mM glucose containing 5 mM lactose, 3 mM glucose containing 5 mM galactose, 3 mM glucose containing 5 mM fructose, 3 mM glucose containing 5 mM arabinose, 10 mM lactose, 10 mM galactose, 10 mM fructose, 10 mM arabinose after incubation with glucose oxidase and T-MIP-PtPd NFs. (Experimental conditions: catalysts: 300 μL of PBS buffer solution (pH 7.0) containing 3 mM of glucose and 100 μL of PBS (pH 7.0) containing 1mg/mL glucose oxidase were mixed at 37°C for 30 min. Then 540 μL of HAc-NaAc buffer solution, 50 μL of TMB (0.35 mmol/L) and 10 μL of (0.1 mg/mL) T-MIP-PtPd NFs were added to the above mixture, and incubated at 25°C for 10 min to measure the absorbance at 652 nm.)

Table S1 Application of the T-MIP-PtPd NFs to detect glucose in human serum samples

Sample	**Added (μM)**	**Found (μM)**	**Recovery (%)**	**RSD (%)**
1#	2000	1876	93.8	3.2
2#	500	461.5	92.3	1.6
3#	50	47.4	94.8	2.6
4#	5	4.78	95.6	3.3
5#	0.05	0.049	98.7	5.2