Support information

Synthesis of PB@FePt hybrid nanoparticles with peroxidasemimicking activity for colorimetric determination of hydrogen peroxide in living cells

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Supplemental Methods

Preparation of PB NCs

Typically, 0.5 mmol citric acid was added into 20 mL FeCl₃ aqueous solution (1.0 mM) under stirring at 60 °C. Then, 20 mL K₄[Fe(CN)₆] aqueous solution (1.0 mM) containing 0.5 mmol citric acid was dropwise added into the above solution under stirring at 60 °C. A clear bright blue dispersion formed immediately during the mixing process, and was allowed to be cooled down to room temperature with stirring continued for another 30 min. To collect as-made PB NCs, 40 mL acetone was added into the dispersion. The mixture was then centrifuged at 14800 rpm for 15 min, which were repeated for three times. Finally, the obtained PB NCs were dried in vacuo for future use.

Synthesis of FePt-DMSA

 $0.5 \text{ mmol Pt}(\text{acac})_2$, 1 mmol Fe $(\text{acac})_3$, and 3 mmol 1,2-hexadecanediol was added into 30 mL dioctyl ether in a 200 mL three-necked flask. Nitrogen was bubbled into the three-necked flask for 20 min to exclude oxygen in it. The temperature was increased to 100 °C, and held for 20 min; 0.17 mL oleylamine and 0.16 mL oleic acid were added under a nitrogen atmosphere. The temperature was raised to 295 °C rapidly, then cooled down to room temperature before heating for 60 min. 30 mL ethanol and 70 mL of nhexane were added into the flask, and the mixture was centrifuged at the speed of 11 000 rpm. The precipitate was washed and centrifuged for three times. The remaining product was FePt.

30 mg FePt and 50 mg DMSA were added into 2 mL DMSO before shaking at room temperature for 1 h; the mixture was centrifuged for 10 min, and the precipitate was washed with anhydrous ethanol before the FePt-DMSA was obtained.

Kinetic analysis

The kinetics data, Km and Vmax, could be obtained according to the typical double-reciprocal Lineweaver-Burk plot and the Michaelis-Menten equation listed as follows: 1/v = Km/Vmax (1/[S] + 1/Km), where v is the initial velocity, Km is the Michaelis constant, Vmax is the maximal reaction velocity, and [S] is the concentration of the substrate.

Detection limit (DL) of H₂O₂

A typical colorimetric analysis was realized as follows: 200μ L of TMB (1mM), 200μ L PB@FePt HNPs (10 µg mL⁻¹), and 200 µL H₂O₂ with various concentrations were added into 1.4 mL CPBS buffer. Then, the mixed solution was used for absorption spectroscopy measurement in time course mode.



The lipophilic FePt NPs were prepared according to a simple polyhydric alcohols method. As shown in Figure S1A, the FePt NPs were found to be monodisperse and uniform in size (3–4nm in diameter). The lattice fringes of the FePt were clearly observed with adjacent fringe spacing of 0.224 nm, which were in accordance with {111} lattice planes for face-centered cubic (fcc) phase of FePt nanoparticles (Figure S1B).



Figure S2. (A) Dynamic light scattering of as-prepared PB NCs, the inset is the as-prepared PB NCs; (B) Dynamic light scattering of as-prepared PB@FePt HNPs, the inset is the as-prepared PB@FePt HNPs.



Figure S3. (A) Dynamic light scattering of as-prepared PB@FePt HNPs; (B) Dynamic light scattering of as-prepared PB@FePt HNPs after staying for 12 days; (C) The images of the obtained PB@FePt HNPs dispersed in water at different times.



Figure S4. The absorbance-wavelength relationships of peroxidase-like activity of PB@FePt HNPs at different reaction time.

The absorbance of PB@FePt-TNB- H_2O_2 system increases quickly at 652 nm with proceeding of reaction, shown in Figure S2. Go beyond 80 seconds, the absorbance of the system barely increased, which means that the decomposition of H_2O_2 almost



finished.

Figure S5. The UV absorbance of different concentration of TMB.

Table S1. Comparison of Km and Vmax between PB@FePt HNPs and other systems.

Catalyst	Km/mM		Vmax	10 ⁻⁸ Ms ⁻¹	Ref.
	TMB	H_2O_2	TMB	H_2O_2	-
PtAg-MoS ₂	25.71	0.386	7.290	3.22	1
PtPdNPs	1.780	0.053	36.40	9.26	2
Cu-hemin	1.420	2.180	26.22	116	3

PS@Au@PB	1.220	0.170	59.00	38.9	4
Cu NCs	0.648	29.16	5.960	4.22	5
PB@FePt	0.426	0.015	21.220	0.205	This work

Table S2. Comparison of the linear range and the LOD of H₂O₂ by means of different sensors.

Catalysts	Linear Range	Detection Limit	Ref.
Cu ₂ (OH) ₃ Cl-CeO ₂	20-50 µM	10 µM	6
$Fe_3O_4@Cu@Cu_2O$	4-50 mM	2000 μΜ	7
GO-FeTPyP NCs	20-500 μM	72 μΜ	8
CoS	50-800 µM	20 µM	9
N-G-Fe ₃ O ₄	0-10 mM	17.1 μΜ	10
Fe ₃ O ₄ /MIL-101(Fe)	5-150 nM	3.7 nM	11
PB@FePt HNPs	20-800 µM	8.24 μM	This work



Figure S6. The cell viabilities of L02 and MCF-7 treated with different amount of PB@FePt-PEG-FA.

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