Supporting Information

Reagents and Materials: Pyrrole and horseradish peroxidase (HRP) were purchased from Sigma-Aldrich. Tetramethylbenzidine (TMB) was obtained from BBI (Ontario, Canada). Ferric chloride hexahydrate ($FeCl_3$•$6H_2O$, Beijing Chemical reagent Corporation, China), Polyvinyl alcohol (PVA-124, Guangzhou Medicine Corporation, China) and other reagents were used directly without further purification. Nanopure water (18.2 MΩ; Millpore Co., USA) was used throughout the experiment.

Instruments: Scanning electron microscopic (SEM) image was recorded using a Hitachi S-4800 Instrument (Japan). FTIR characterization was carried out on a BRUKE Vertex 70 FTIR spectrometer. UV-vis absorbance measurements experiments were carried out on a Jasco-V550 UV/Vis spectrophotometer. EDS was carried out using a HITACHI S-4500 instrument.

Synthesis of polypyrrole nanoparticles: 1.5 g PVA was dissolved in 20 mL de-ionized water at 60 °C, and then cooled to room temperature. $FeCl_3$•$6H_2O$ (0.23 M) was added to the stirred PVA solution, the system changed from a clear state to a yellow one. After 1 hour equilibration, pyrrole monomer (0.1M) was introduced and the temperature was controlled at 4 °C. The mixture solution was stirred for 4 hours. After the completion of polymerization, the resulting nanoparticles were separated from the dispersion solution by centrifugation and washed at least five times with hot water to remove impurities. The obtained PPy nanoparticles were resuspended with water by ultrasonication.

Bioassay: In a typical test, chemicals were added into buffer solution (25 mM Na_2HPO_4, pH 4.0) in an order of certain amounts of the PPy, TMB, and H_2O_2. Kinetic measurements of the peroxidase reactions of PPy were performed using a Jasco-V550 UV-Vis spectrophotometer in time course mode at 652 nm.

Cell Culture: The murine macrophage-like RAW264.7 cells were grown at 37 °C in an atmosphere of 5 v/v% CO_2 in air, in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% heat-inactivated FBS, 1.5 g L^{-1} $NaHCO_3$, 100 units

mL^{-1} penicillin, 100 mg mL^{-1} streptomycin, 4.5 g L^{-1} glucose and 4 mM glutamine. The media was changed every three days, and the cells were digested by trypsin and resuspended in fresh complete medium before using.

Measuring the Flux of H_2O_2 Releasing from macrophages: The RAW264.7 cells were washed, resuspended in PBS (pH = 7.4) and counted with a hemacytometer. Cells were incubated in a 1.5 mL EP tube at a density of 3×10^5 cells per 20 μL at 37 °C. Then, PMA at different concentrations (5 μL) were added to the EP tubes and incubated for 20 min. After that, 200 μL PPy (8 μg PPy) in pH 4.0 PBS and 10 μL TMB (80 mM) were added into the above solutions. And the time-dependent absorbance changes at 652 nm of TMB from PBS solution containing PPy/TMB kit and RAW264.7 cells (3×10^5 cells) in the presence of different concentrations of PMA and LPS were measured. The calibration curve was obtained under similar conditions in the presence of different concentrations of H_2O_2 in PBS media.

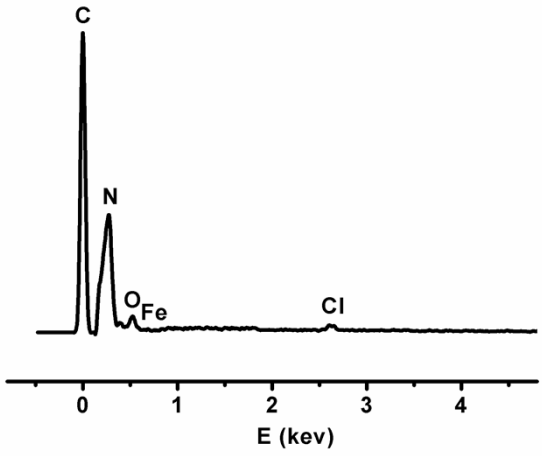

Fig. S1 EDS spectrum of the PPy nanoparticles.

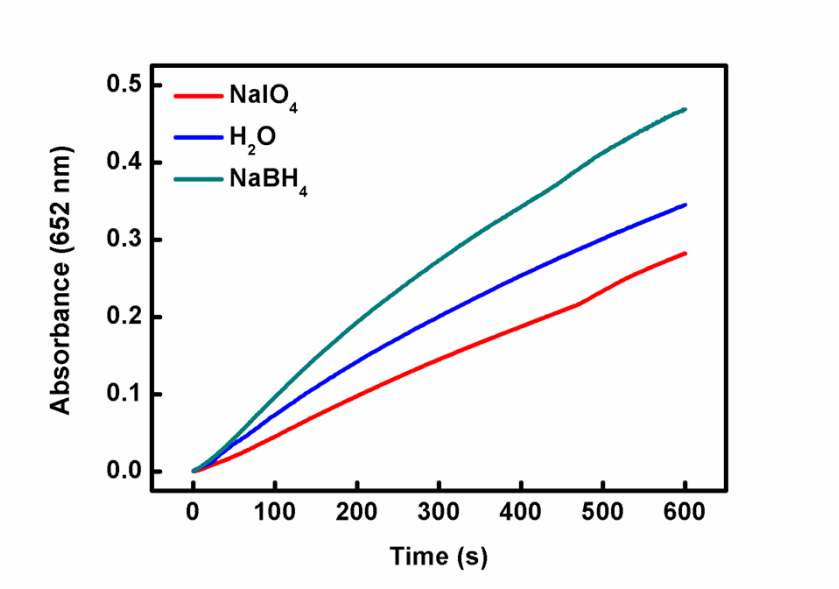

Fig. S2 The peroxidase-like activity of PPy after the nanoparticles were treated with 2% $NaBH_4$, 2% $NaIO_4$ or H_2O.

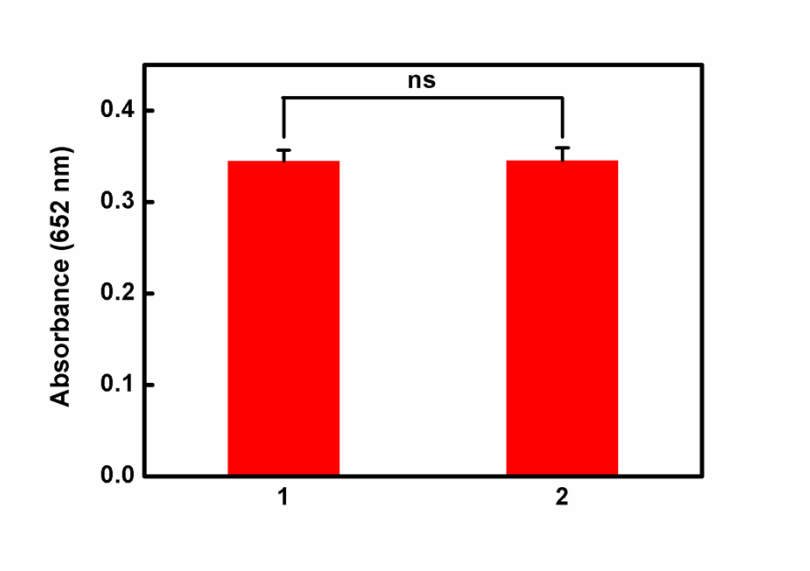

Fig. S3 The peroxidase-like activity of PPy in different batches. Error bars represent standard deviation of three independent measurements; ns=no significant difference ($P > 0.05$).

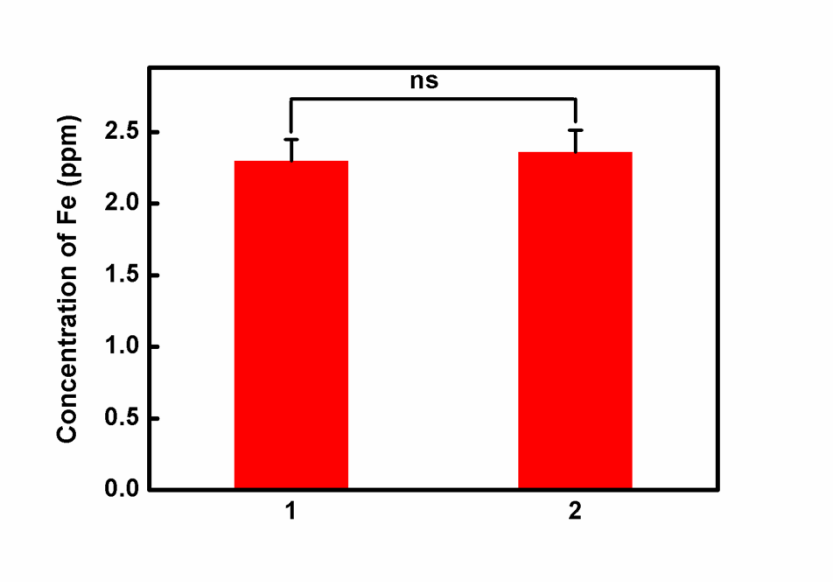

Fig. S4 The mass of Fe was measured using ICP-MS in 1 mg/mL PPy in different batches. Data represent mean values for n = 3, and the error bars represent standard deviation of the means; ns=no significant difference ($P > 0.05$).

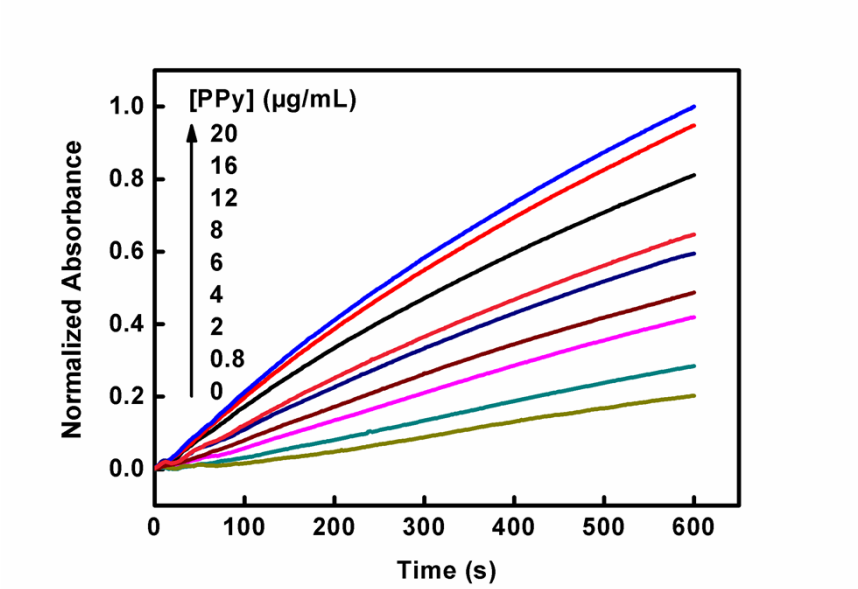

Fig. S5 The time-dependent absorbance changes at 652 nm in the presence of different concentrations of PPy nanoparticles in phosphate buffer (25 mM Na_2HPO_4, pH 4.0).

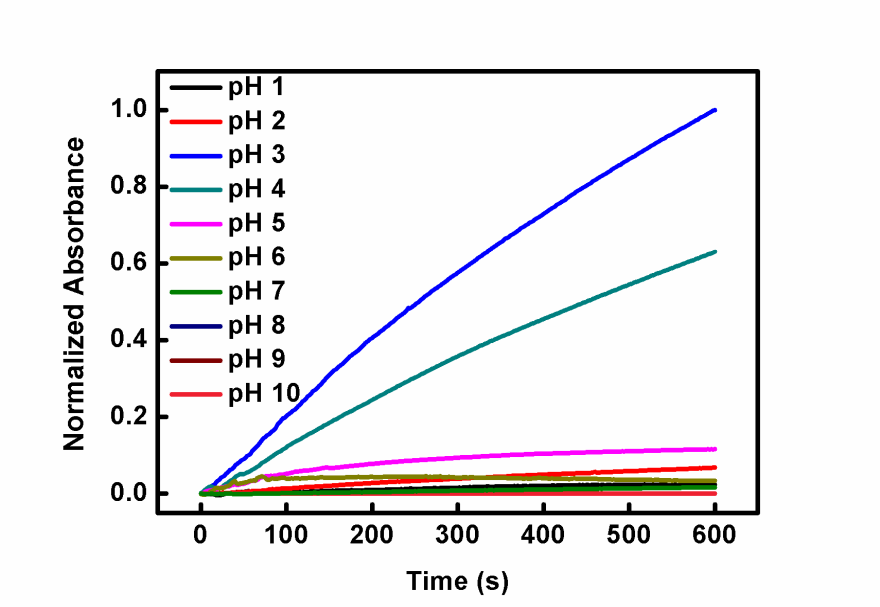

Fig. S6 The time-dependent absorbance changes at different pH.

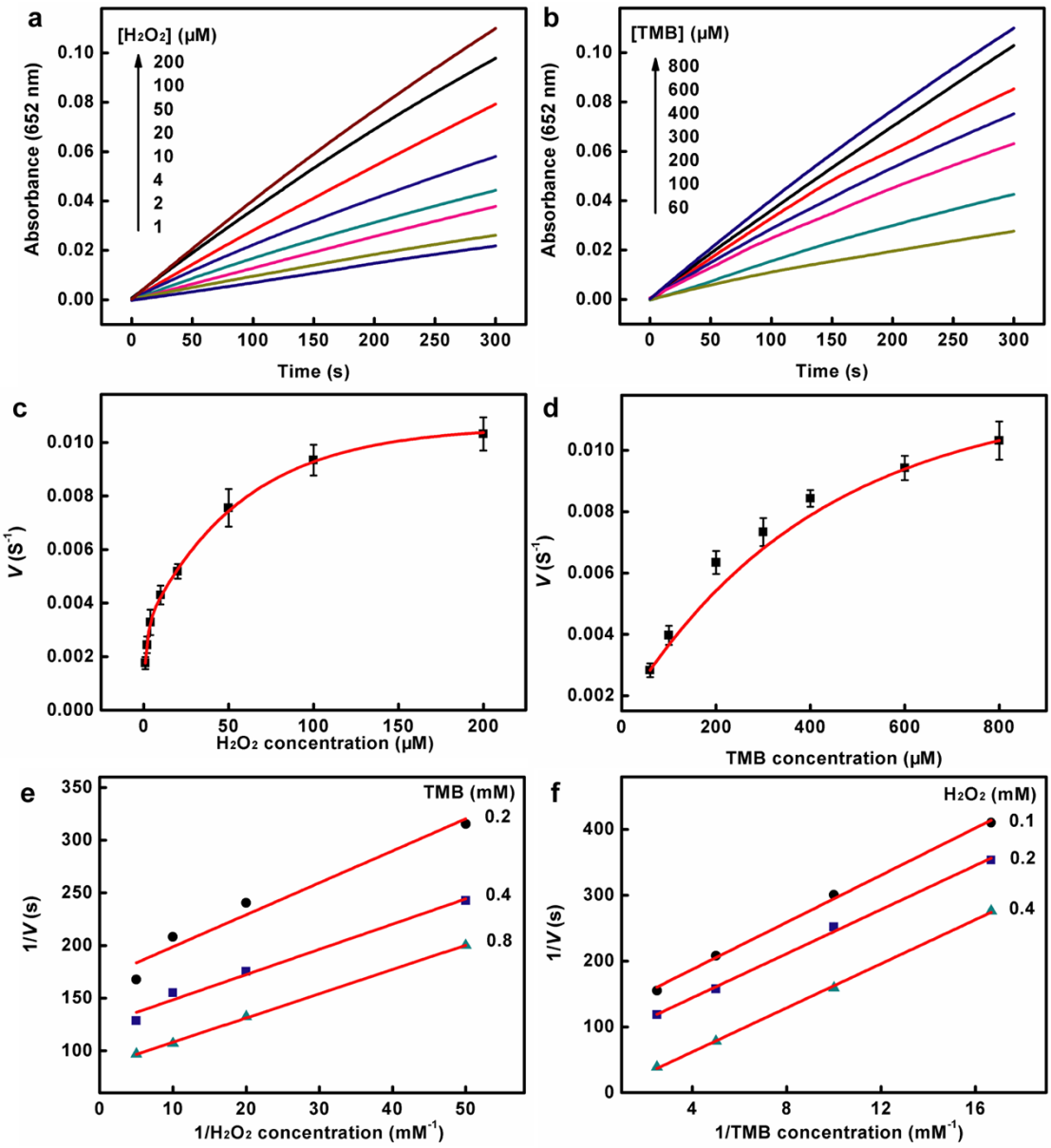

Fig. S7 (a, b) Time-dependent absorbance changes at 652 nm of TMB reaction solutions catalyzed by the PPy nanoparticles in the presence of different concentrations of H_2O_2 or TMB. (c-d) Steady-state kinetic assay of the PPy nanoparticles. Experiments were carried out in 25 mM phosphate buffer (pH 4.0) using 20 μg mL^{-1} PPy nanoparticles. (a, c) TMB concentration was fixed at 800 μM and the H_2O_2 concentration was varied. (b, d) H_2O_2 concentration was fixed at 0.2 mM and the TMB concentration was varied. Error bars shown represent the standard error derived from three repeated measurements. (e, f) Double-reciprocal plots of activity of PPy at a fixed concentration of one substrate versus different concentration of the second substrate for H_2O_2 or TMB.

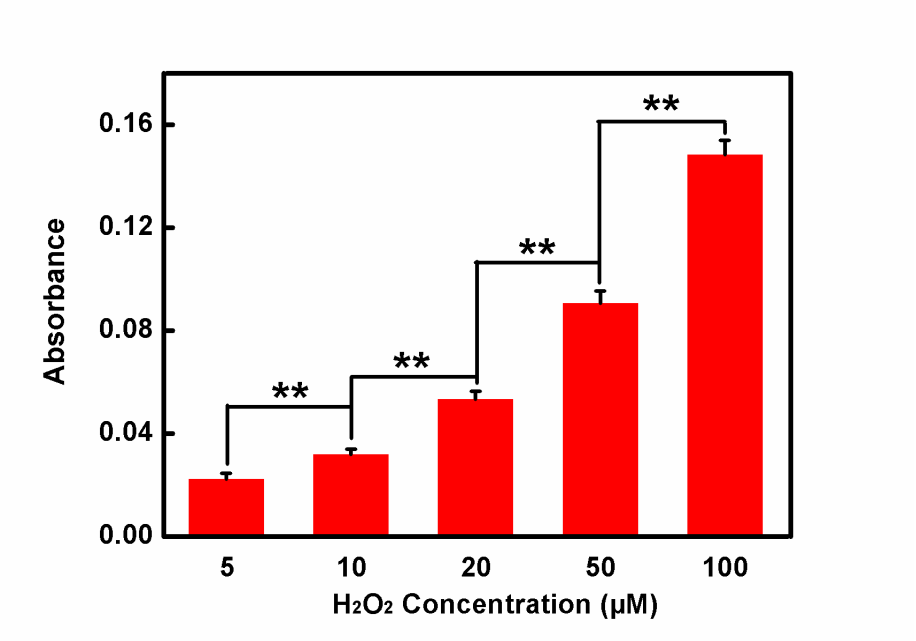

Fig. S8 H_2O_2 concentration dependent absorbance containing the PPy/TMB kit**.** Asterisks represents significant differences between the absorbance values in the presence of different concentrations of H_2O_2 (*$P < 0.005$, **$P < 0.001$).

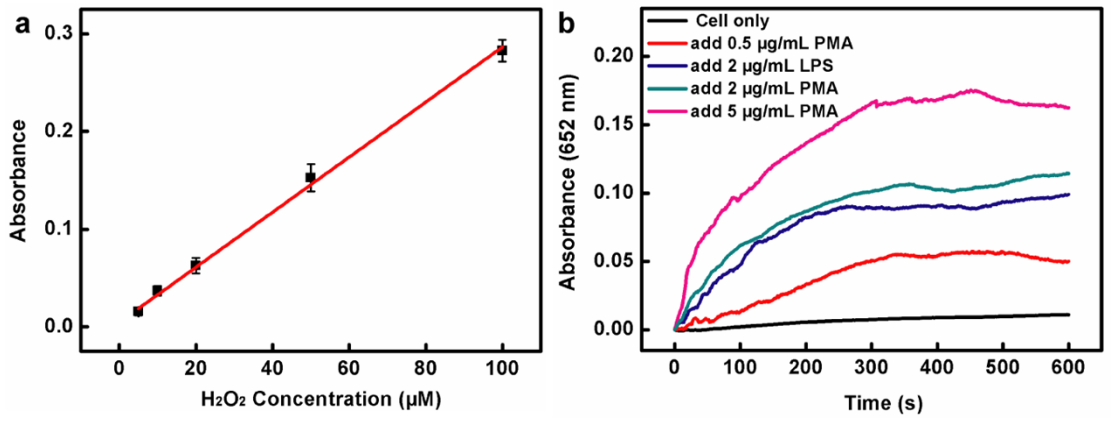

Fig. S9 (a) H_2O_2 concentration dependent UV absorbance containing the HRP/TMB kit. The background absorbance from the same solutions but without H_2O_2 have been subtracted. Error bars represent standard deviation of three independent measurements. (b) Time-dependent absorbance changes at 652 nm of TMB from PBS solution containing HRP/TMB kit and RAW264.7 cells (3×10^5 cells) in the presence of different concentrations of PMA and LPS.