

Supporting Information For

A colorimetric biosensor based on enzyme-catalysis-induced
production of inorganic nanoparticles for sensitive detection of
glucose in white grape wine

Huang Dai,^a Yuqing Li,^a Qi Zhang,^a Yingchun Fu,^a Yanbin Li^{a,b,*}

^a College of Biosystems Engineering and Food Science, Zhejiang University, Hangzhou 310058, China

^b Department of Biological and Agricultural Engineering, University of Arkansas, Fayetteville, AR 72701, USA

*E-mail: yanbinli@zju.edu.cn. Tel: +86 571 88982536.

Experimental

Optimization of reaction parameters

For effects of pH, the MES solution pH was adjusted with HCl (12 M) and KOH (12 M). The mixture of 0.25 mM FeCl₃, 0.25 mM K₃Fe(CN)₆, 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES solution (pH 1.0 - pH 7.0) after 10 min was prepared. Then, the absorbance of the solution was measured at a wavelength of 706 nm against a blank without glucose.

For effects of GOx concentration, the mixture of 0.25 mM FeCl₃, 0.25 mM K₃Fe(CN)₆, and 1 mM glucose in 50 mM MES (pH 3.0) with different concentrations of GOx after 10 min was prepared. Then, the absorbance of the solution was measured at 706 nm.

For effects of the molar ratio Fe³⁺ versus Fe(CN)₆³⁻, the total volume of 0.5 mM FeCl₃ and 0.5 mM K₃Fe(CN)₆ were 200 μL while the volume ratio of two solutions varied from 0 to 19 in 50 mM MES (pH 3.0). 10 μL of 1.1 mg mL⁻¹ GOx and 22 mM glucose was added in succession. Then, the absorbance of the solution was measured at 706 nm after 10 min.

For effects of FeCl₃/K₃Fe(CN)₆ concentration, 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES (pH 3.0) with different concentrations of FeCl₃/ K₃Fe(CN)₆ was prepared. Then, the absorbance of the solution was measured at 706 nm after 10 min.

For effects of reaction time, 0.75 mM FeCl₃, 0.75 mM K₃Fe(CN)₆, 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES (pH 3.0) was prepared. Then, the absorbance of the solution was monitored every two minutes for 24 minutes at 706 nm.

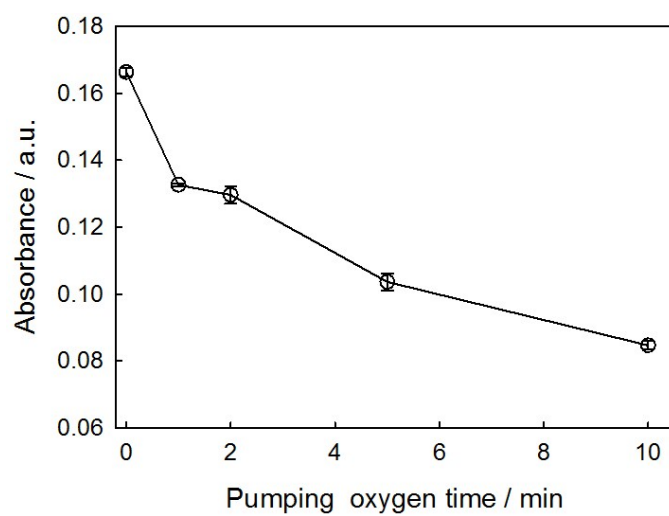


Fig. S1. The effects of oxygen on the absorbance of PBNPs. Reaction conditions: 0.25 mM FeCl₃, 0.25 mM K₃Fe(CN)₆, 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES (pH3.0) at 25 °C, reaction for 10 min.

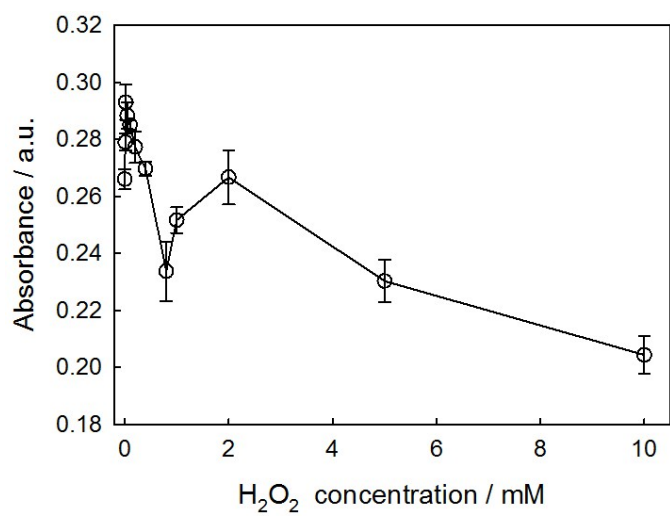


Fig. S2. The effects of H₂O₂ concentration on the absorbance of PBNPs. Reaction conditions: 0.25 mM FeCl₂, 0.25 mM K₃Fe(CN)₆, in 50 mM MES (pH3.0) at 25 °C, reaction for 10 min.

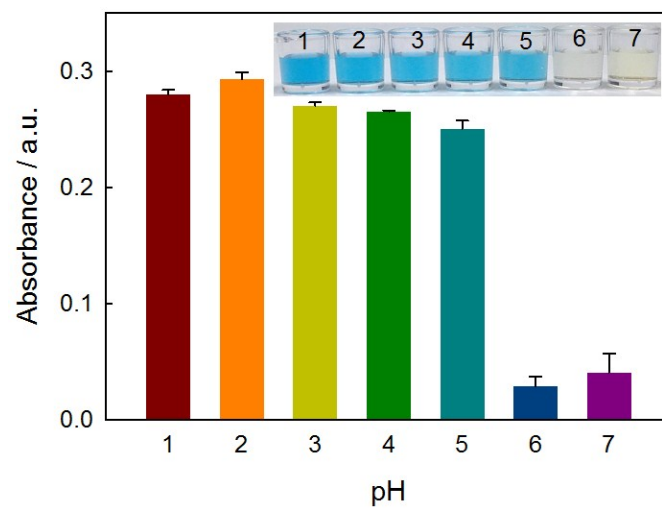


Fig. S3. The effects of pH on the absorbance of PBNPs. Reaction conditions: 0.25 mM FeCl₂, 0.25 mM K₃Fe(CN)₆, in 50 mM MES (pH 1.0 - pH 7.0) at 25 °C, reaction for 10 min.

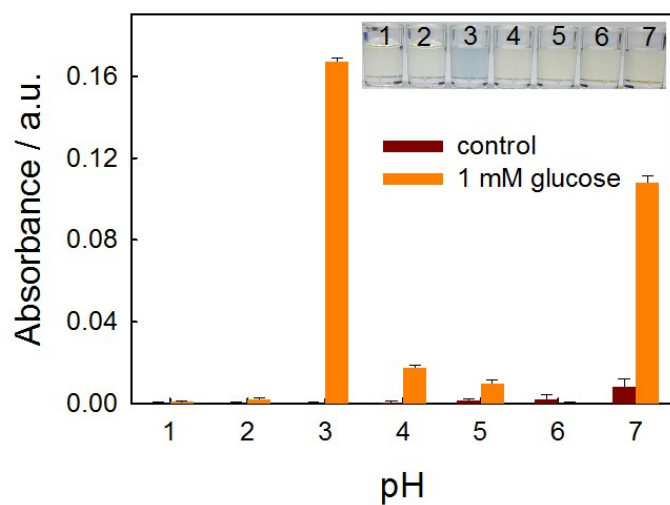


Fig. S4. The effects of pH on the absorbance of PBNPs. Reaction conditions: 0.25 mM FeCl₃, 0.25 mM K₃Fe(CN)₆, 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES (pH 1.0 - pH 7.0) at 25 °C, reaction for 10 min. The control means the system without glucose.

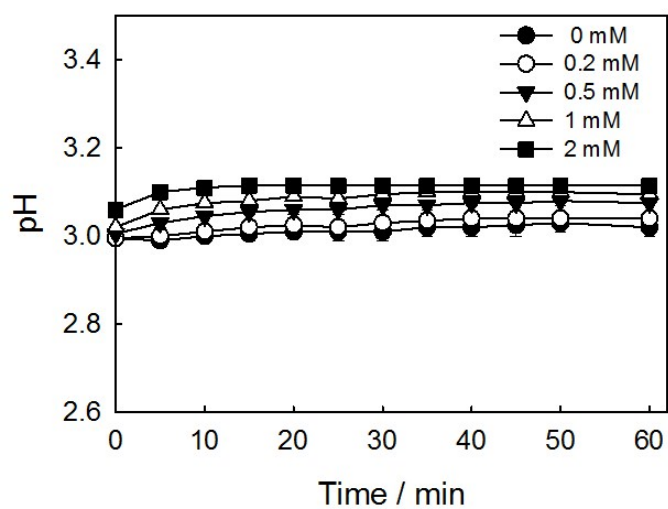


Fig. S5. The pH of the solution during the experiments. Reaction conditions: 0.25 mM FeCl_3 , 0.25 mM $\text{K}_3\text{Fe}(\text{CN})_6$, 0.05 mg mL^{-1} GOx, and different concentrations of glucose in 50 mM MES (pH 3.0) at 25 °C.

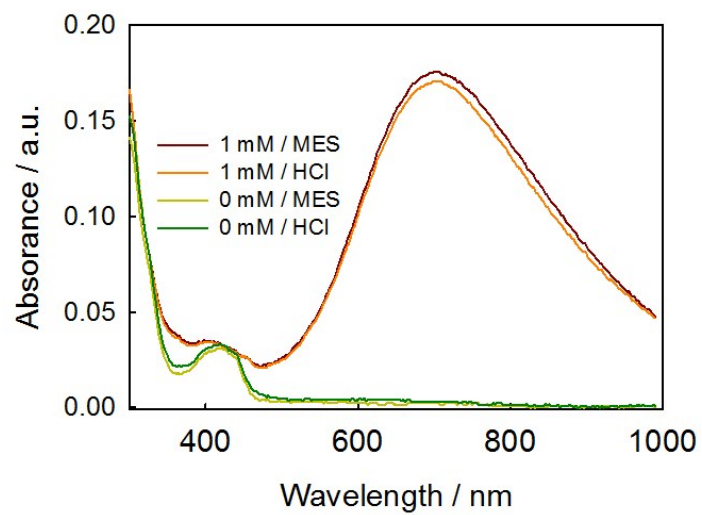


Fig. S6. The effects of medium on the absorbance of PBNPs. Reaction conditions: 0.25 mM FeCl_3 , 0.25 mM $\text{K}_3\text{Fe}(\text{CN})_6$, 0.05 mg mL^{-1} GOx, and 1 mM glucose in 50 mM MES and hydrochloric acid (pH 3.0) at 25 °C, reaction for 10 min.

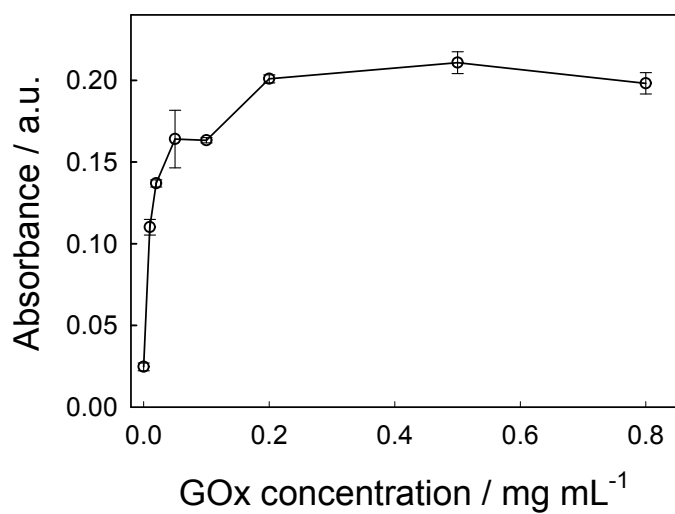


Fig. S7. Effects of GOx concentration on the formation of PBNPs. Reaction conditions: 0.25 mM FeCl₃, 0.25 mM K₃Fe(CN)₆, and 1 mM glucose in 50 mM MES (pH 3.0) at 25 °C, reaction for 10 min.

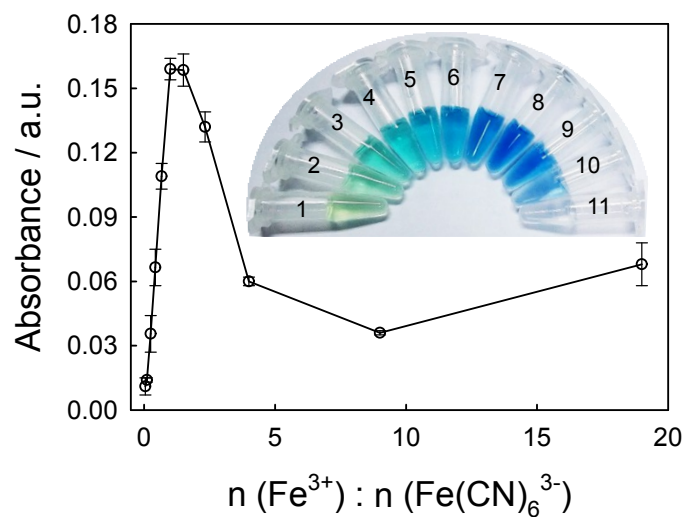


Fig. S8. Effects of the molar ratio of FeCl_3 to $\text{K}_3\text{Fe}(\text{CN})_6$ and digital image (inset). The molar ratio arranged from 0 to 19 (1 to 11). Reaction conditions: 0.05 mg mL^{-1} GOx, and 1 mM glucose in 50 mM MES (pH 3.0) at $25 \text{ }^\circ\text{C}$, reaction for 10 min.

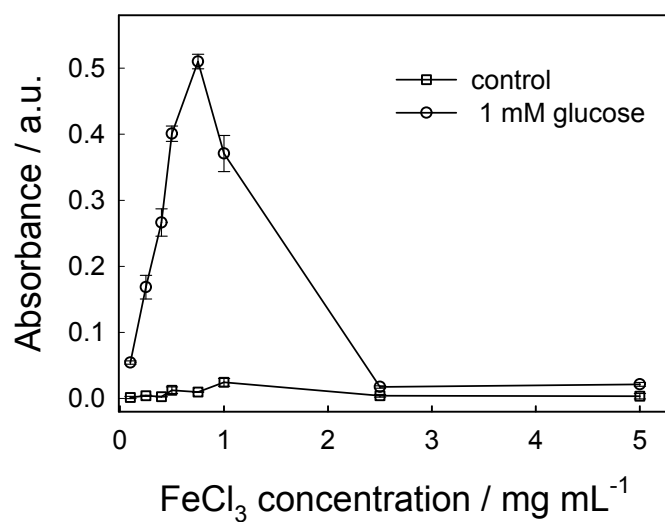


Fig. S9. Effects of FeCl₃ concentration on the formation of PBNPs. Reaction conditions: 0.05 mg mL⁻¹ GOx, and 1 mM glucose in 50 mM MES (pH 3.0) at 25 °C, reaction for 10 min.

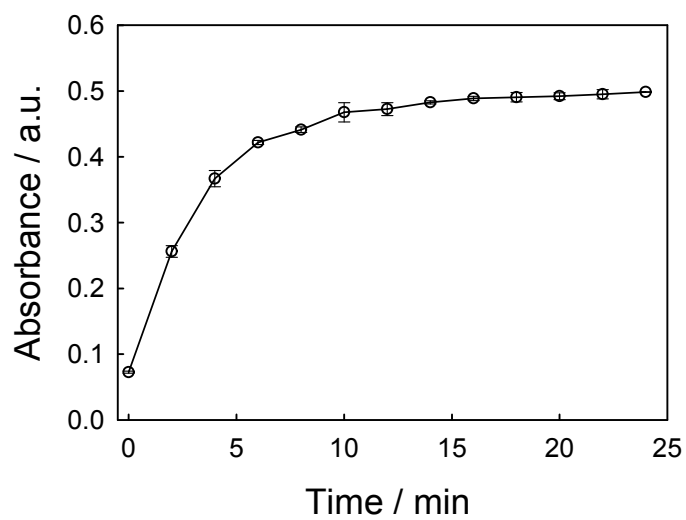


Fig. S10. Absorbance changes of PBNPs in a function of time at the wavelength of 706 nm. Reaction conditions: 0.75 mM FeCl_3 , 0.75 mM $\text{K}_3\text{Fe}(\text{CN})_6$, 0.05 mg mL^{-1} GOx, and 1 mM glucose in 50 mM MES (pH 3.0) at 25 °C.

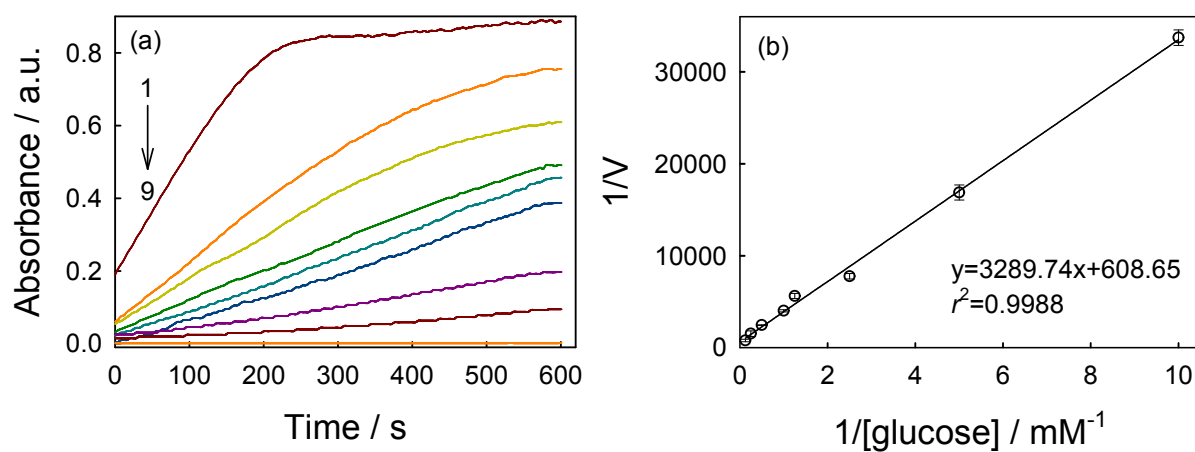


Fig. S11. UV-vis spectra (a) of 0.02 mg mL^{-1} GOx to the addition of different concentrations of glucose in the presence of 0.75 mM FeCl_3 and $0.75 \text{ mM K}_3\text{Fe(CN)}_6$ in 50 mM MES (pH 3.0). Glucose concentrations (all in mM) from top to bottom (1 to 9): 8, 4, 2, 1, 0.8, 0.4, 0.2, 0.1, 0. Lineweaver-Burk plot (b) is also given.

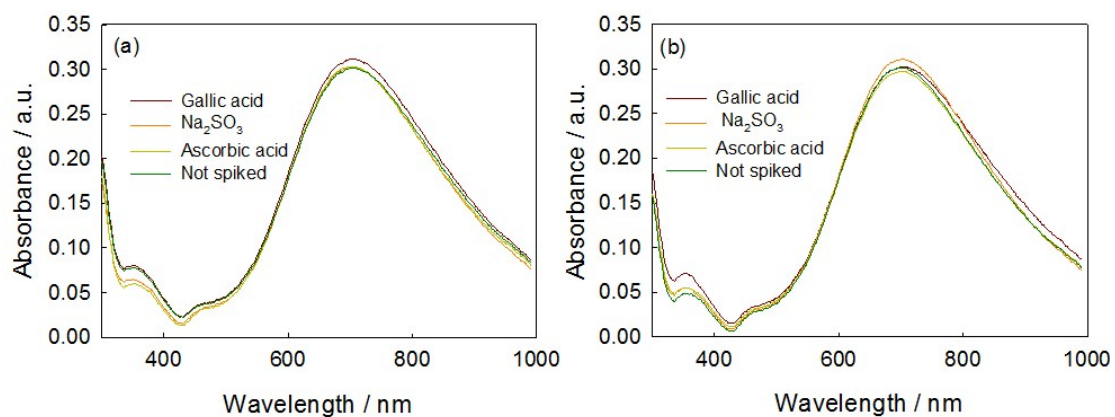


Fig. S12. UV-vis spectra for the detection of glucose in sweet wine (a) and half sweet wine (b) with or without spiked reductants (sulfite, gallic acid and ascorbic acid) in 50 mM MES (pH 3.0) at 25 °C. Concentrations: 0.75 mM FeCl₃, 0.75 mM K₃Fe(CN)₆, 0.05 mg mL⁻¹ GOx, 196.8 mg L⁻¹ Na₂SO₃, 500 mg L⁻¹ gallic acid, 12 mg L⁻¹ ascorbic acid. Reaction time: 10 min.