

Porphyrin functionalized Co(OH)₂/GO nanocomposite as an excellent peroxidase mimic for colorimetric biosensing

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1. Steady-state kinetic study

The steady-state kinetic assays were monitored at 652 nm with a 3 min interval in a time-scan mode. A series of experiments were carried out as follows: 200 μL H₂TCPP/Co(OH)₂/GO solution (0.4 mg·mL⁻¹) in HAc-NaAc buffer solution (1.4 mL, pH 4) treated with 0.2 mL different concentration of TMB (0.05-0.25 mM) and 0.2 mL of H₂O₂ (25 mM), or 0.2 mL different concentration of H₂O₂ (20-100 mM) and TMB (0.1 mM) with a total reaction volume of 2 mL at room temperature. The apparent kinetic parameters were calculated according to the Line weaver-Burk plot: $1/v = K_m/V_{max} (1/S + 1/K_m)$, where v is the initial velocity, S represents the concentration of the substrate, K_m is the Michaelis-Menten constant, and V_m is the maximal reaction velocity ¹.

2. Colorimetric detection of glutathione

Colorimetric detection of glutathione was established based on the excellent peroxidase-like activity of H₂TCPP/Co(OH)₂/GO. The mixed solution containing 1.2 mL HAc-NaAc buffer solution, 200 μL of H₂O₂, 200 μL of H₂TCPP/Co(OH)₂/GO NPs and 200 μL of TMB was incubated for 2 min at room temperature. Then, the

various concentrations of GSH were added the above mixture, the absorbance of different reaction systems was recorded. Notably, the blue color of the reaction systems started to fade gradually with the concentration of GSH increased.

Table S1. Peak area in O1s.

Position (eV)	Area	Affiliation
529.3	6072.924	Co-O
530.7	165497.4	Co-OH
531.6	35141.480	Co-O

Reference

- 1 D.-F. Chai, Z. Ma, H. Yan, Y. Qiu, H. Liu, H.-D. Guo, et al., RSC Advances, 2015, **5**, 78771-9.