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Planar intercalated Copper (II) complex molecule as small molecule enzyme mimic combined with  $Fe_3O_4$  nanozyme for bienzyme synergistic catalysis applied to the microRNA biosensor

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To appear in: Biosensors and Bioelectronic

Received date: 6 January 2018 Revised date: 20 March 2018 Accepted date: 20 March 2018

Cite this article as: Liang Tian, Jinxu Qi, Olayinka Oderinde, Chen Yao, Wei Song and Yihong Wang, Planar intercalated Copper (II) complex molecule as small molecule enzyme mimic combined with Fe<sub>3</sub>O<sub>4</sub> nanozyme for bienzyme synergistic catalysis applied to the microRNA biosensor, *Biosensors and Bioelectronic*, https://doi.org/10.1016/j.bios.2018.03.045

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#### **ACCEPTED MANUSCRIPT**

## Planar intercalated Copper (II) complex molecule as small molecule enzyme mimic combined with $Fe_3O_4$ nanozyme for bienzyme synergistic

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#### catalysis applied to the microRNA biosensor

Liang Tian, Jinxu Qi, Olayinka Oderinde, Chen Yao, Wei Song, Yihong Wang\* School of Chemistry and Chemical Engineering, Southeast University, Nanjing, 211189, PR China.

#### Abstract

Enzyme mimics have been developed for bioassay of nucleic acids, with some of them involving complicated labeling. Herein, we report a label-free bioassay for ultrasensitive electronic determination of microRNA at an ultralow concentration based on target-triggered long-range self-assembly DNA-based hybridization chain reaction (HCR) protocol coupled with bienzyme mimics synergistic catalysis strategy. In this work, a planar intercalation molecule, copper (II) complex, is applied for the first time as a small molecule enzyme mimic as well as intercalation molecule in microRNA biosensor for signal amplification. Fe<sub>3</sub>O<sub>4</sub> nanozyme were used as a separate and enriched target under magnetic field, and also in combination with HCR protocol detected in 3,3',5,5'-tetramethylbenzidine+hydrogen peroxide (TMB+H<sub>2</sub>O<sub>2</sub>) system to improve the sensitivity of the biosensor. Under optimal conditions, these strategies present good electrochemical behaviors for the detection of microRNA with a wide range from 100 aM to 100 nM and at relatively low detection limit of 33 aM. This remarkable sensitivity can make this proposed approach a promising scheme for development of next-generation microRNA sensors without the need of enzyme labeling or fluorophore labeling.

Graphical abstract

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