In situ construction of a cobalt oxyhydroxide loaded pyrene-based fluorescent organic nanoprobe for bioimaging of endogenous ascorbic acid in living cells

Huijuan Yan,*[a] Shuanghui Liu,[a] Shuo Yang,[a] Wu Ren,[b] Jingfang Shangguan,[a] Jieli Lv,[a] Mengzhen Zhang,[a] Juan Tang[a] and Ying Zhao[a, c]

[a] *School of Pharmacy, Xinxiang Medical University, Xinxiang, Henan 453003, P. R. China;*

[b] *School of Medical Engineering, Xinxiang Neurosense and Control Engineering Technology Research Center, Xinxiang Key Lab of Biomedical Information Research, Xinxiang Medical University, Xinxiang, Henan 453003, P. R. China;*

[c] *Xinxiang Key Laboratory of Clinical Psychopharmacology, Xinxiang Medical University, Xinxiang, 453003, China.*

* Corresponding author. Tel.: +86-373-3029879

E-mail address: yanhuijuan2013@126.com (H. J. Yan)

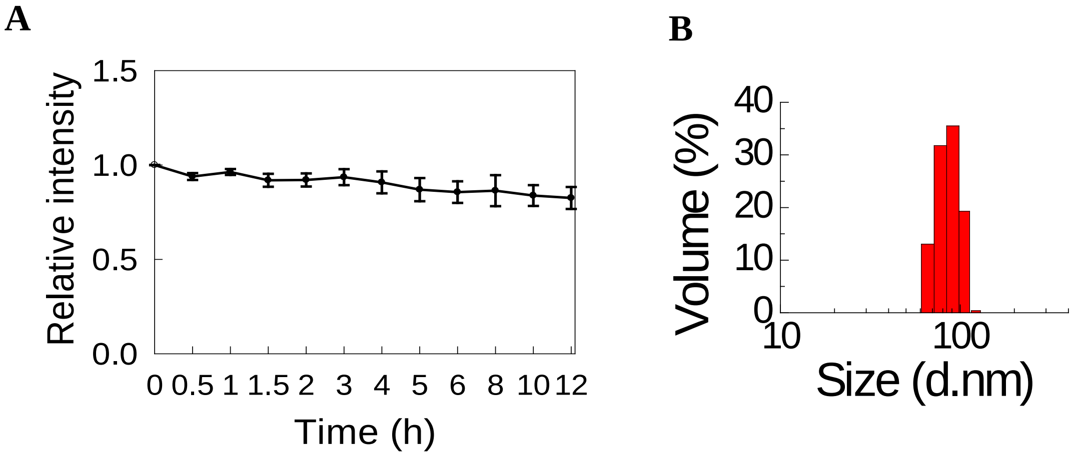
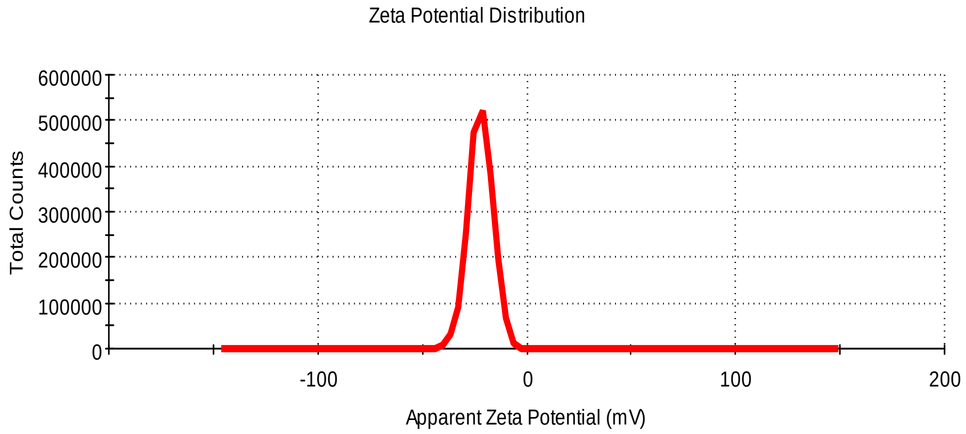


Fig. S1 (A) Relative intensity of PyFONs during 12 h, respectively. (B) DLS of PyFONs.

A



B

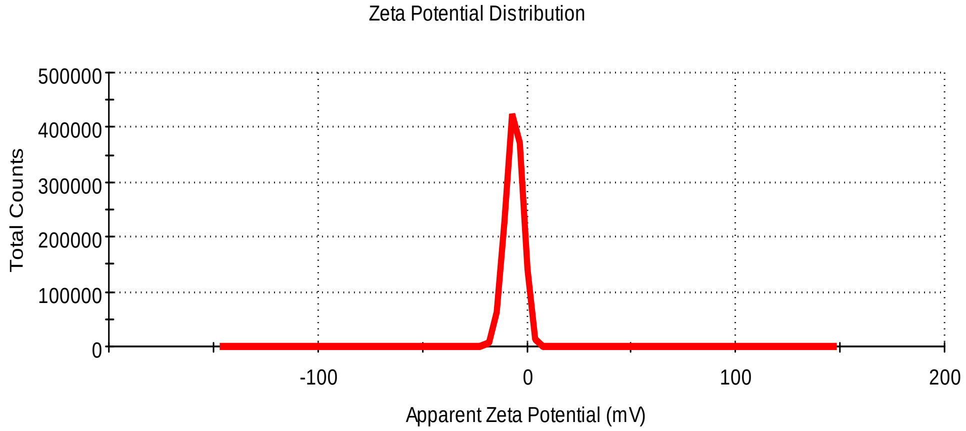


Fig. S2 (A) Zeta potential of PyFONs (-22.1mV) and CoOOH-modified Py nanoconjugate (-6.07 mV).

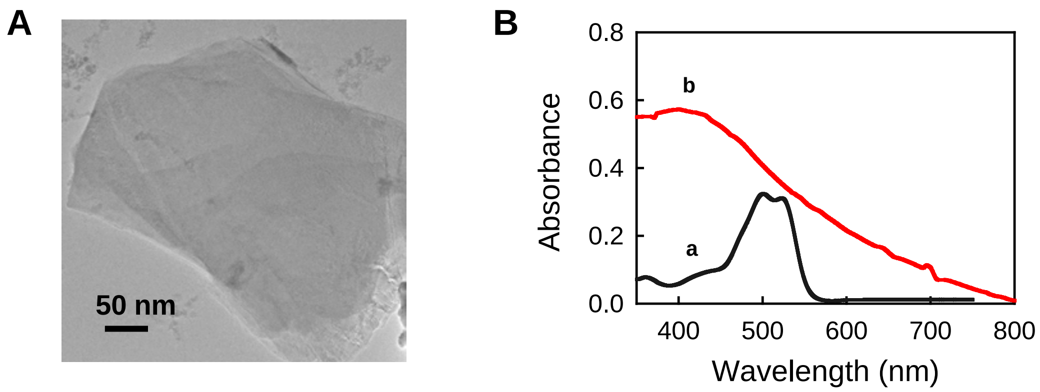


Fig. S3 (A) TEM image of CoOOH nanoflakes; (B) Absorption spectra of aqueous solutions of $CoCl_2$ (a) and CoOOH nanoflakes (b).

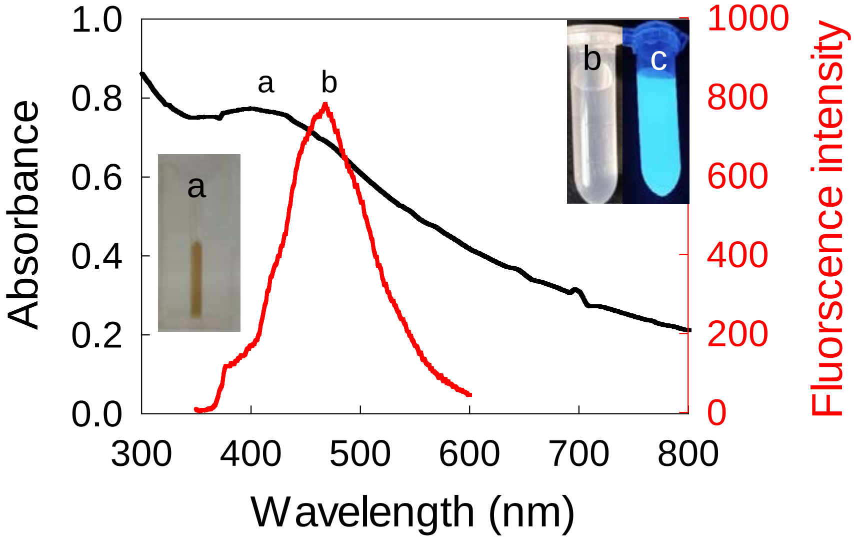


Fig. S4 UV-vis spectrum of aqueous solutions of CoOOH nanoflakes (a) and fluorescence spectrum of PyFONs (b). Inset: photographs of the CoOOH nanoflakes (a) and PyFONs under visible light (b) and irradiated by a laser pointer of 365 nm (c).

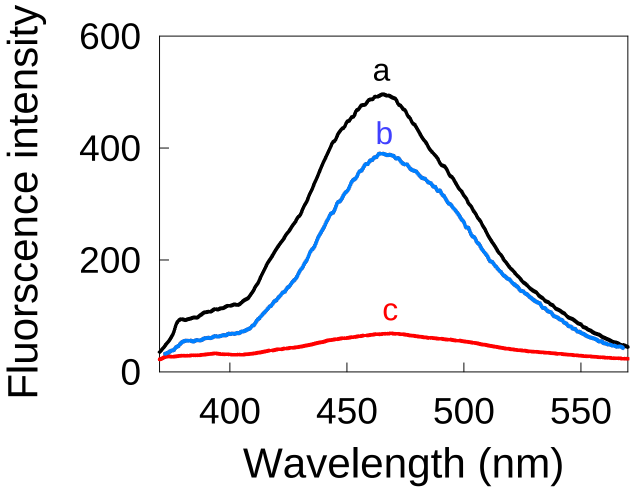


Fig. S5 FL spectra of aqueous solutions of PyFONs (line a), physical mixture of CoOOH and PyFONs (line b), and CoOOH-modified PLNPs (line c).

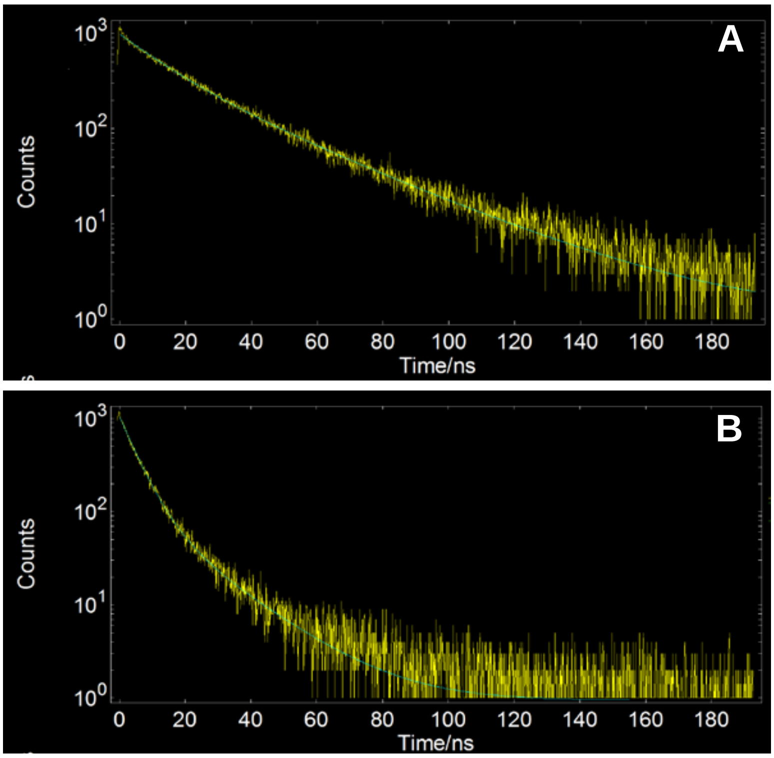


Fig. S6 Fluorescence decay dynamics of PyFONs (A) and in situ grown of CoOOH nanoflakes on the surface of PyFONs (B).

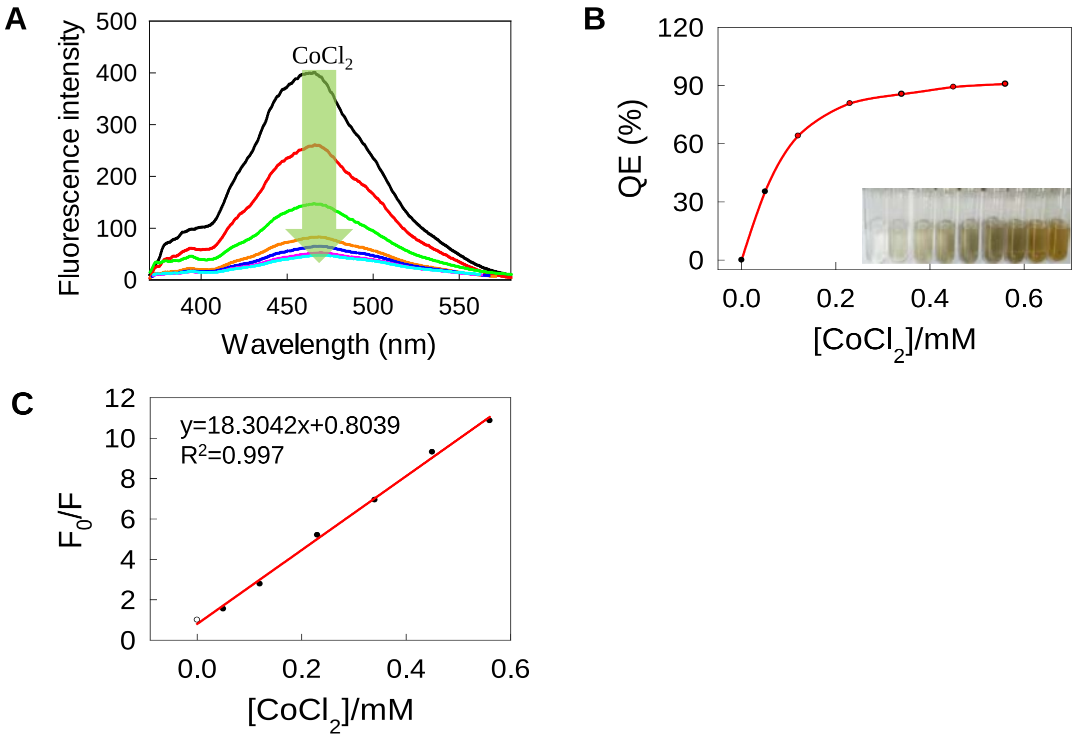


Fig. S7 (A) FL spectra of PyFONs with different $CoCl_2$ feeding amounts (from bottom to top: 0, 0.05, 0.12, 0.23, 0.34, 0.45, 0.56 mM), with the excitation wavelength at 345 nm. (B) Relationship between quenching efficiency (QE %) and the contents of $CoCl_2$. Inset: photographs of PyFONs at a series of different $CoCl_2$ feeding amount. (C) The corresponding Stern-Volmer plot of F_0/F versus the concentrations of $CoCl_2$ feeding. F_0 and F correspond to the fluorescence intensity of the PyFONs at 468 nm in the absence and presence of $CoCl_2$, respectively. All above solutions include NaClO (0.2 M) and NaOH (0.8 M).

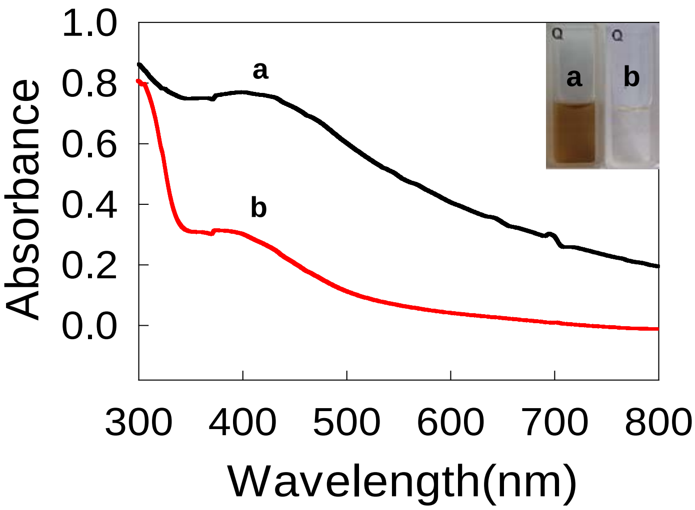


Fig. S8 UV-Vis absorption spectrum of CoOOH nanoflakes in absence (curve a) and present (curve b) of AA. The inset displays the photographic images of CoOOH nanoflakes solutions in absence (a) and present (b) of AA under broad daylight.

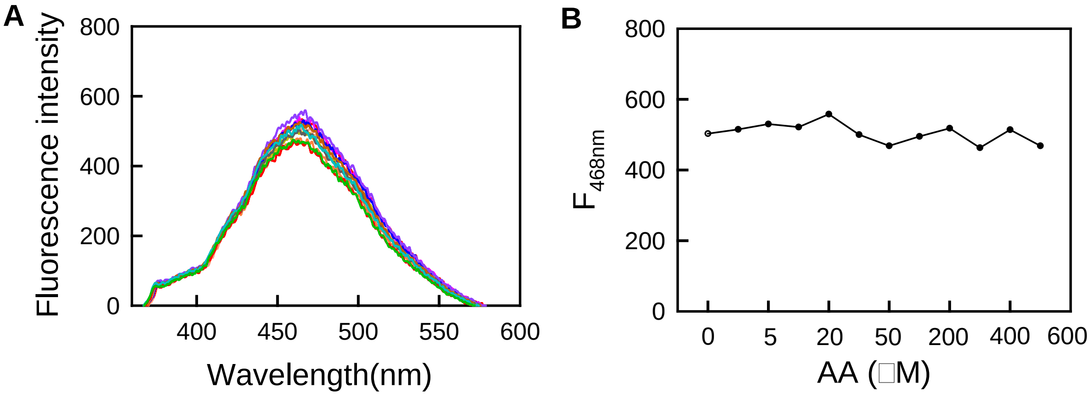


Fig. S9 The effect of AA on PyFONs. (A) FL spectra of PyFONs with different AA (0, 2, 5, 10, 20, 30, 50, 100, 200, 300, 400 and 500 μM, respectively). (B) Fluorescence at 468 nm vs. AA concentration. λex/λem = 345 nm/468 nm.

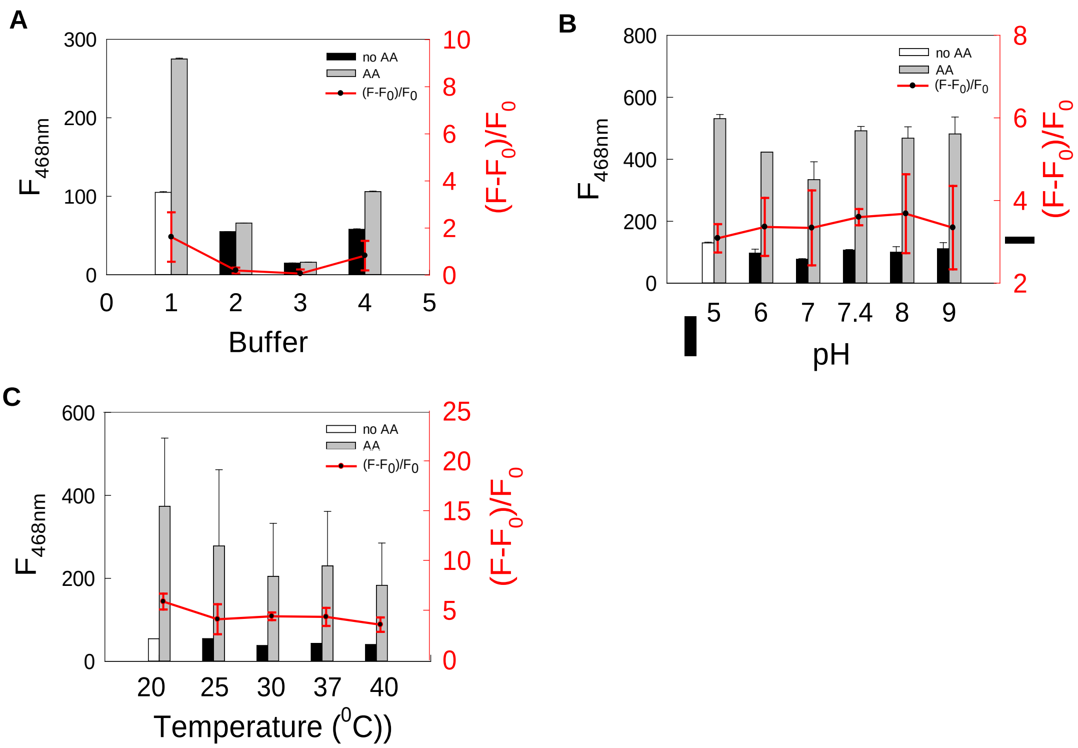


Fig. S10 Effects of buffer (A), pH (B), and temperature (C) on the fluorescence responses to different concentrations of AA. Buffer 1-4: 1. 137 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4, and 1.8 mM KH_2PO_4; 2. 0.2 M Na_2HPO_4 and 0.2 M NaH_2PO_4; 3. 0.2 M Na_2HPO_4 and 0.1 M citric acid; 4. 0.2 M KH_2PO_4 and 0.2 M NaOH; pH = 5.0-9.0; [Py@CoOOH] = 90 μg·mL^{-1}; [AA] = 400 μM AA; $\lambda_{ex}/\lambda_{em}$ = 345 nm/468 nm.

Table S1. An overview on recently reported nanomaterial-based fluorometric methods for determination of ascorbic acid

Materials used	Method applied	LOD	Response range	Reference
NIR GQDs (NGs)	Fluorometric (TPEM)	270 nM	1 ~ 30 μM	[1]
$NaYF_4$:Yb/Tm@$NaYF_4$	Fluorometric (upconversion approach)	0.2 μM	0 ~ 60 μM	[2]
$NaYF_4$:Gd/Yb/Tm/Ho@ $NaYF_4$	Fluorometric (UCL images)	0.63 μM	0 ~ 40 μM	[3]
MoS_2 quantum dots (QDs)	Fluorometric (ratiometric detection)	0.21 μM	0.8 ~ 32μM	[4]
CdTe quantum dots (QDs)	Fluorometric (OPE)	1.3 μM	10 ~ 250 μM	[5]
DEASPI/βCDP nanomicelle	Fluorometric (TPEM)	0.27 μM	2 ~ 50 μM	[6]
Persistent luminescence nanoparticles (PLNPs)	Fluorometric (PLI)	0.59 μM	1 ~ 100 μM	[7]
Fluorescent Polydopamine (PDA) nanoparticles	Fluorometric (OPE)	4.8 μM	0 ~ 500 μM	[8]
PyFONs@CoOOH	Fluorometric (OPE)	0.21 μM	2 ~ 500 μM	This work

Notes: One-photon excited (OPE), Persistent luminescence imaging (PLI), Time-gated luminescence microscopy (TGLM), Two-photon excited microscopy (TPEM), Upconversion Luminescence images (UCL images).

Table S2. Analytical results of AA in human plasma using PyFONs@CoOOH nanoprobe.

Samples	Added AA (μM)	Found AA (μM)	Recovery (%)	RSD (%, n=3)
1	20	19.3	96.5	3.7
2	80	81.9	102.4	6.1
3	160	158.2	98.9	5.4

References

[1] L. L. Feng, Y. X. Wu, D. L. Zhang, X. X. Hu, J. Zhang, P. Wang, Z. L. Song, X. B. Zhang, W. H.Tan, *Anal. Chem*, 2017, 89, 4077-4084.

[2] Y. Cen, J. Tang, X. J. Kong, S. Wu, J. Yuan, R. Q. Yu and X. Chu**,** *Nanoscale*, 2015, 7, 13951-13957.

[3] Q. X, Han, Z, Dong, X. L, Tang, L, Wang, Z. H, Ju, W. S, Liu, *J. Mater. Chem. B,* 2017, 5, 167-172.

[4] Z. H. Wu, D. Y. Nan, H. Yang, S. Pan, H. Liu, X. L. Hu, *Anal. Chim. Acta*, 2019, 1091,59-68.

[5] J. Zhu, Z. J. Zhao, J. J. Li, J. W. Zhao, *J. Lumin*, 2017, 192, 47-55.

[6] H.J. Yan, Y. F. Liu, W. Ren, J. F. Shangguan, X. Yang, *Microchim. Acta*, 2019, 186, 201.

[7] N. Li, Y. H. Li, Y. Y. Han, W. Pan, T. T. Zhang, B. Tang, *Anal. Chem*, 2014, 86, 3924-3930.

[8] Y. Y. Zhao, L. Li, R. Q. Yu, T. T. Chen, X. Chu, *Anal. Methods*, 2017, 9, 5518-5524.