SUPPORTING INFORMATION

# Magnetic Fe-N-C nanoparticles as dual nanozyme for lable-free colorimetric detection of antibiotics

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In this study, we reported a simple one-step pyrolytic strategy for fabrication of N-C codoped Fe-based nanoparticles (Fe-N-C) and exhibited their oxidase-mimic and peroxidase-mimic activity in a chromogenic reaction with 3,3',5,5'-tetramethylbenzidine dihydrochloride (TMB) oxidation. The structure analysis revealed that Fe-N and $Fe_3C$ structures were formed with Fe-imace coordination compound as precursors and melamine as nitrogen source, which provided abundant active sites for Fe-N-C nanozyme. Interestingly, owing to the blocking effect triggered by $\pi$-$\pi$ stacking between the Tetracycline antibiotics tetraphenyl skeleton and Fe-N-C, the substrate affinity of the Fe-N-C was significantly blocked, resulting in the solution color fading with the increase of TCs concentration. Under the optimum conditions, the UV absorption intensity versus the concentration of TCs was found to be linear over the range of 0.08-90 $\mu$M for tetracycline (TC), 0.1–80 $\mu$M for chlortetracycline (CTC) and 0.09–100 $\mu$M for oxytetracycline (OTC). The detection limits were 62 nM for TC, 88 nM for OTC and 85 nM for CTC. The Fe-N-C displayed good storage and long-term stability, with 4℃ stored for 30 days. In addition, the superparamagnetic of Fe-N-C is conducive to its recycling and reuse, avoiding secondary pollution caused by the catalyst.

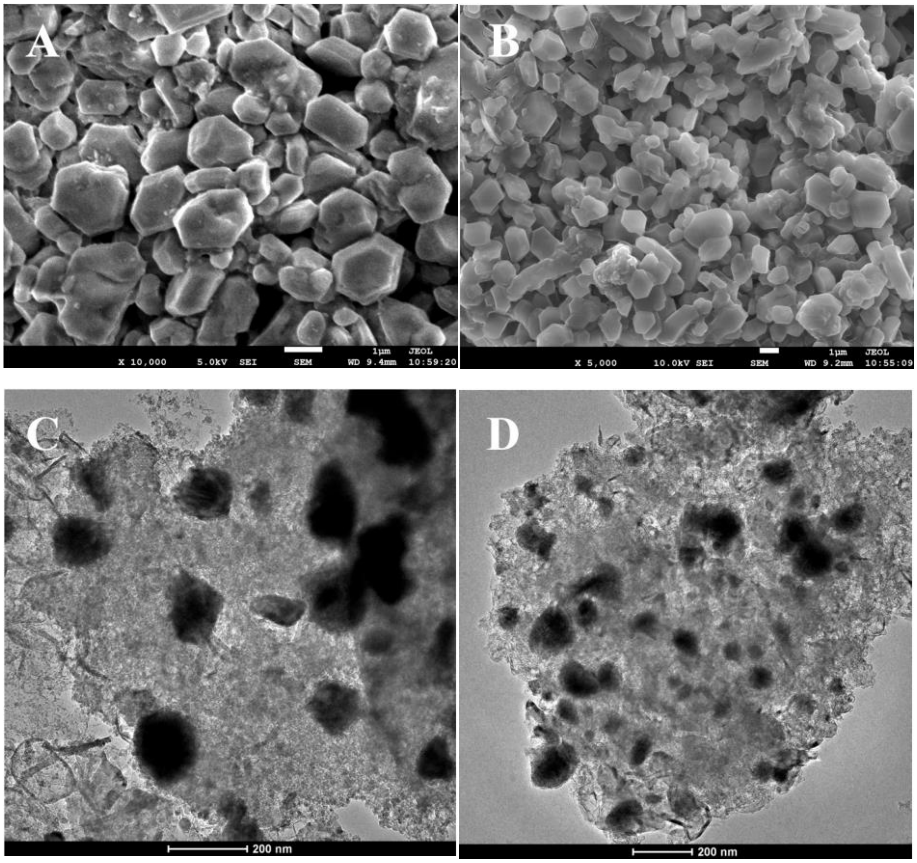


**Fig. S1** SEM images of Fe-N-C-700 (A) and Fe-N-C-800 (B); TEM images of Fe-N-C-700 (C) and Fe-N-C-800 (D).

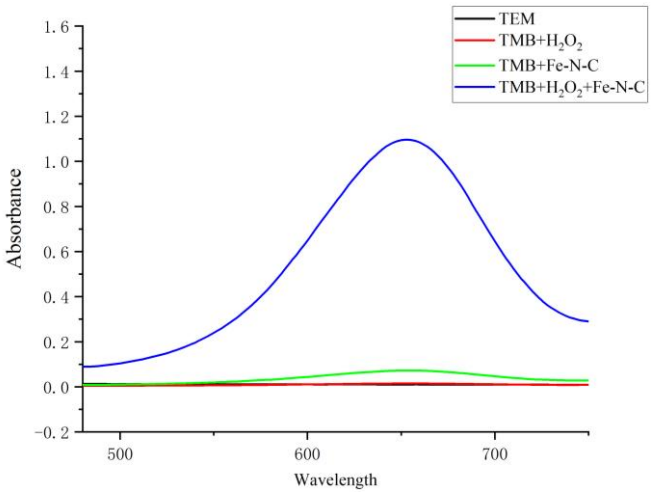


**Fig. S2** Absorption spectra of TMB in different reaction systems. (top-to-bottom: TMB, TMB+ $H_2O_2$, TMB+ Fe-N-C and TMB+ $H_2O_2$+Fe-N-C).

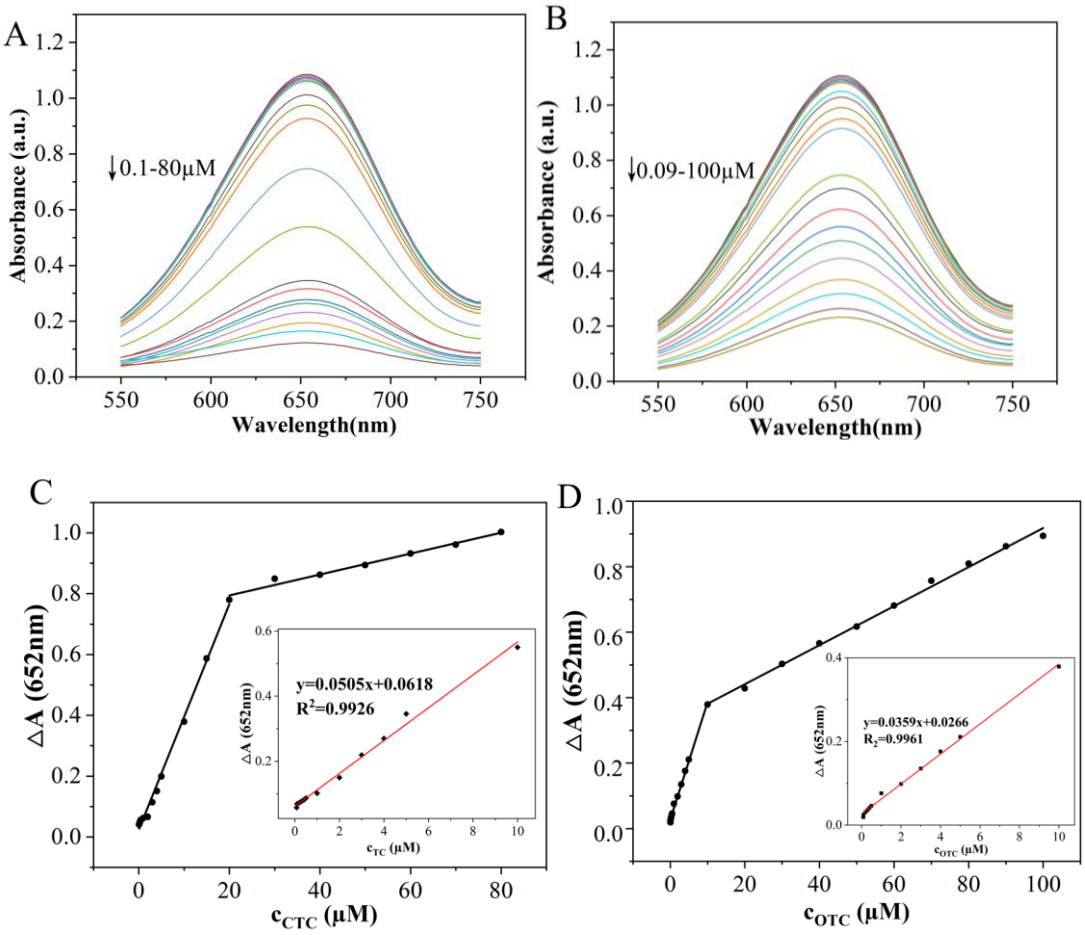


**Fig. S3** (A) UV – vis absorption spectra of this Fe-N-C nanozyme sensing system following incubation with CTC in the range from 0.1 to 80.0 μM. (B) UV – vis absorption spectra of this Fe-N-C nanozyme sensing system following incubation with OTC in the range from 0.09 to 100.0 μM. (C) Calibration curve of absorbance variations versus quantities of CTC added. (D) Calibration curve of absorbance variations versus quantities of OTC added.

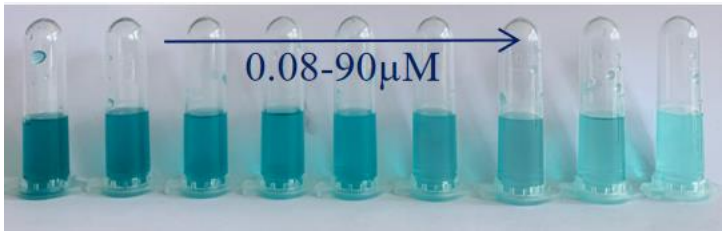


**Fig. S4** Photographs for color changes of the detection solutions after addition of various TC concentrations.

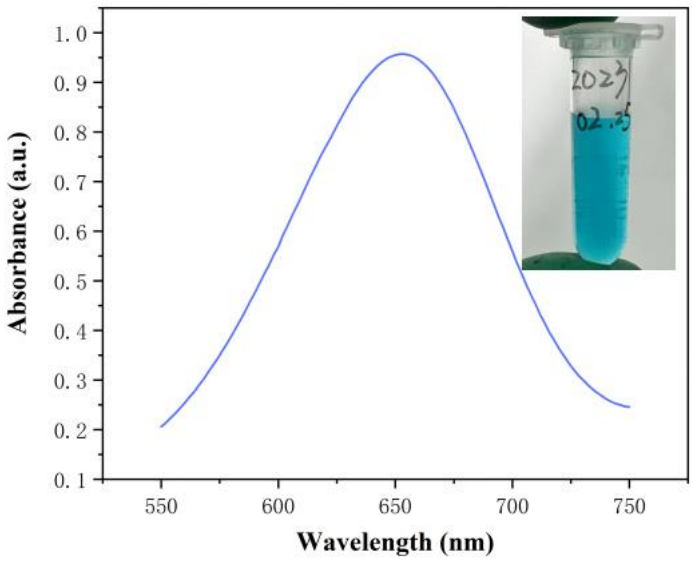


**Fig. S5** Absorption spectra and photographs (inset) of TMB.

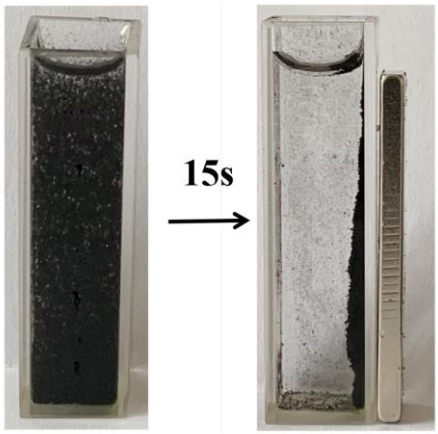


**Fig. S6** Verification of the magnetic properties of Fe-N-C. (A magnetite is placed outside the tube wall, and Fe-N-C is observed to be magnetically drawn to one side of the tube after 15 seconds.)

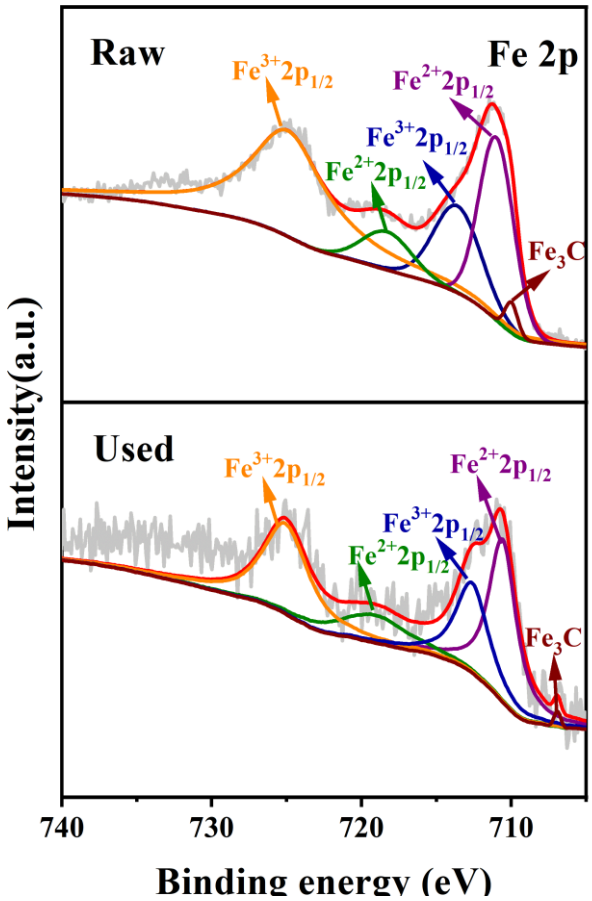


**Fig. S7** XPS spectra of Fe 2p before and after the catalytic reaction.

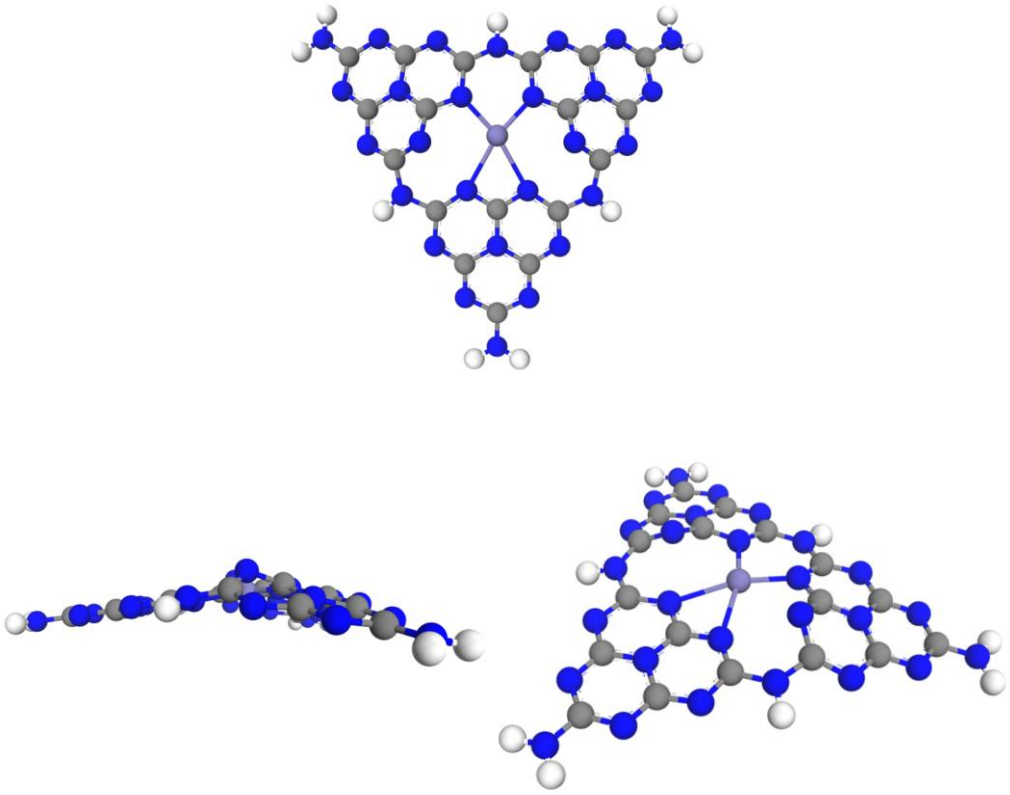

**Fig. S8** Molecular structure model of Fe-N-C.

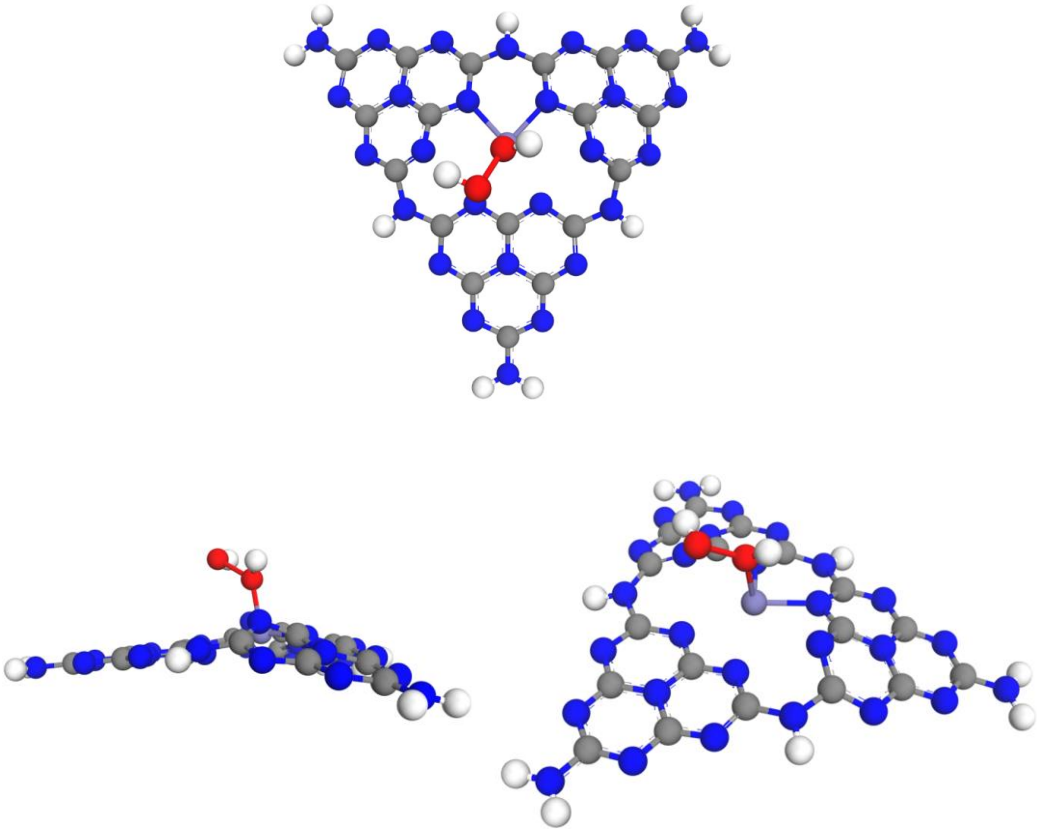

**Fig. S9** DFT calculation of the $H_2O_2$ activation of the Fe-N-C.

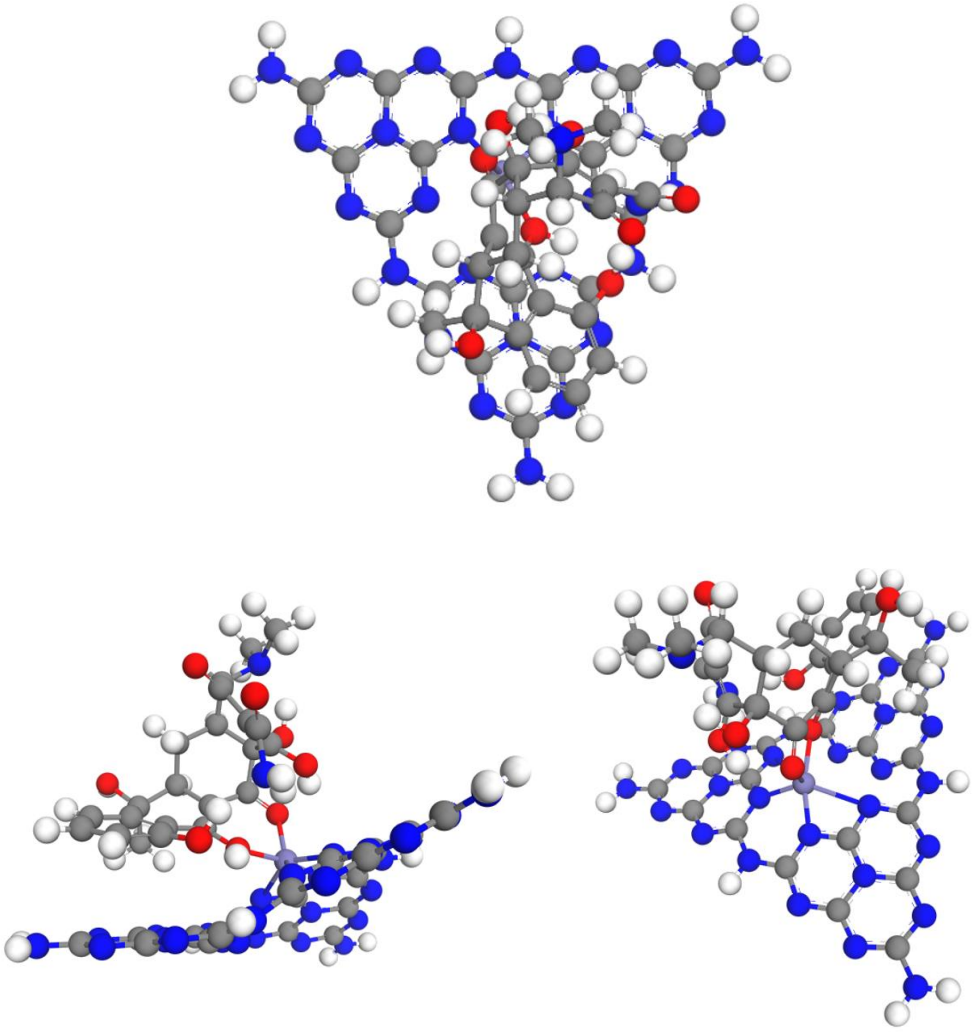

**Fig. S10** DFT calculation of the TC activation of the Fe-N-C.

**Table S1** The concentrations of core elements in the Fe-N-C-x catalysts.

| Catalysts | Fe/at.% | C/at.% | N/at.% | O/at.% |
|---|---|---|---|---|
| Fe-N-C-700 | 0.88 | 88.68 | 5.76 | 4.68 |
| Fe-N-C-800 | 5.54 | 75.12 | 3.13 | 16.22 |
| Fe-N-C-900 | 5.89 | 79.33 | 3.09 | 11.69 |

**Table S2** The $K_m$ and $V_m$ of HRP and Fe-N-C to $H_2O_2$ and TMB.

| Catalyst | Substance | $K_m$ (mM) | $V_m$ ($10^{-8}$ M $s^{-1}$) |
|---|---|---|---|
| HRP | $H_2O_2$ | 3.70 | 8.71 |
| | TMB | 0.343 | 10.00 |
| Fe-N-C | $H_2O_2$ | 0.2 | 11.22 |
| | TMB | 0.124 | 14.29 |

**Table S3** Comparison of various methods for TCs detection.

| Determinand | Method | Detection range | LOD | Real samples | Ref. |
|---|---|---|---|---|---|
| TC | Colorimetry | 0.001-4 μg/mL | 0.3333 ng/ml | Milk and beef | 1 |
| | Fluorescence | 0.32-32 μg/ml | 278 ng/ml | Environmental water samples | 2 |
| | Electrochemistry | $5 \times 10^{-7}$-$2.25 \times 10^{-4}$ M | $1.67 \times 10^{-7}$ M | Water | 3 |
| | SERS spectroscopy | $10^{-10}$-$10^{-5}$ M | $1.0 \times 10^{-11}$ M | Water and milk | 4 |
| | Electrochemiluminescence | 1-100 μM | 0.23 μM | Drug | 5 |
| | Colorimetry | 0.08-90 μM | 62 nM | Lake water | This work |
| CTC | Colorimetry | 0.001-4 μg/mL | 0.3702 ng/ml | Milk and beef | 1 |
| | Fluorescence | 0-50 μM | 0.46 μM | Tap water and milk | 6 |
| | Fluorescence | 0.12-6.0 μg/ml | 40 ng/ml | Environmental water samples | 2 |
| | Electrochemiluminescence | 1-100 μM | 0.16 μM | Drug | 5 |
| | Colorimetry | 0.1-80 μM | 88 nM | Lake water | This work |
| OTC | RP-HPLC | 0.05-50 μg/ml | 0.01μg/ml | Milk | 7 |
| | Fluorescence | 25-440 nM | 22 nM | Tap water | 8 |
| | Electrochemistry | 1.00-540 nM | 30.0 pM | Milk | 9 |
| | Photoelectrochemistry | $2 \times 10^{-4}$ -$1 \times 10^{-1}$nM | $1.2 \times 10^{-4}$ nM | Water | 10 |
| | Electrochemiluminescence | 0.1-100 μM | 0.1μM | Drug | 5 |
| | Colorimetry | 0.09-100 μM | 85 nM | Lake water | This work |

**Table S4** Practical analysis in lake water samples.

| Sample | Determinand | Added (μM) | Found (μM) | Recovery % | RSD (n = 3) |
|---|---|---|---|---|---|
| lake water | TC | 0 | 0 | - | - |
| | | 5 | 4.88 | 97.6 | 3.3% |
| | | 10 | 10.5 | 105 | 4.3% |
| | | 20 | 18.3 | 91.5 | 3.91% |
| | OTC | 0 | 0 | - | - |
| | | 5 | 4.51 | 90.2 | 2.94 |
| | | 10 | 9.3 | 93 | 4.16 |
| | | 20 | 22.1 | 110.5 | 3.2 |
| | CTC | 0 | 0 | - | - |
| | | 5 | 5.27 | 105.4 | 3.77 |
| | | 10 | 11.28 | 112.8 | 3.56 |
| | | 20 | 18.11 | 90.55 | 4.1 |

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