Supplementary Materials

A Novel Label-Free Fluorescent Strategy for Methyltransferase Activity Assay based on dsDNA-Templated Copper Nanoparticles Coupled with Endonuclease-Assisted Signal Transduction System

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Table S1. Sequences of Oligonucleotides (5'→ 3') Used in This Work

Probes	**Sequences**	
Probe **1**	GTATTCTGATCTCTCTAC	18-mer
Probe **2**	GTAGAGAGATCAGAATAC	18-mer
Probe **3**	GTATTCTGA	9-mer
Probe **4**	TCTCTCTAC	9-mer
Probe **5**	CGTATTCTGATCTCTCTACA	20-mer
Probe **6**	TGTAGAGAGATCAGAATACG	20-mer
Probe **7**	ACGTATTCTGATCTCTCTACAC	22-mer
Probe **8**	GTGTAGAGAGATCAGAATACGT	22-mer
Probe **9**	TATTCTGATCTCTCTA	16-mer
Probe **10**	TAGAGAGATCAGAATA	16-mer
Probe **11**	ATTCTGATCTCTCT	14-mer
Probe **12**	AGAGAGATCAGAAT	14-mer

Fig. S1. Fluorescence spectra of CuNPs by dsDNA with different numbers of base pairs (bp) as templates. P1/P2 (18 bp); P1 (ssDNA, 18 bp); P2 (ssDNA, 18 bp), P3/P4 (9 bp). The dsDNA-templated fluorescent CuNPs was obtained in the reaction solution containing 100 μM Cu^{2+}, 2 mM ascorbate, and 1μM dsDNA for 10 min.

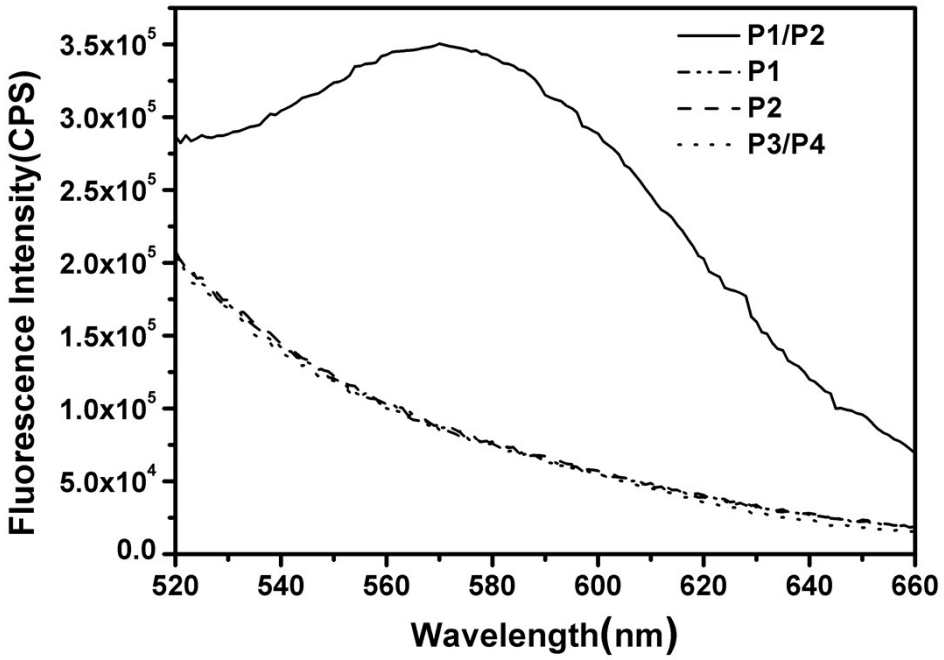

Fig. S2. TEM images of the dsDNA-templated fluorescent CuNPs for different lengths dsDNA with (A) 14 bp; (B) 18 bp; (C) 22 bp. Other reaction conditions are as Fig. **2**.

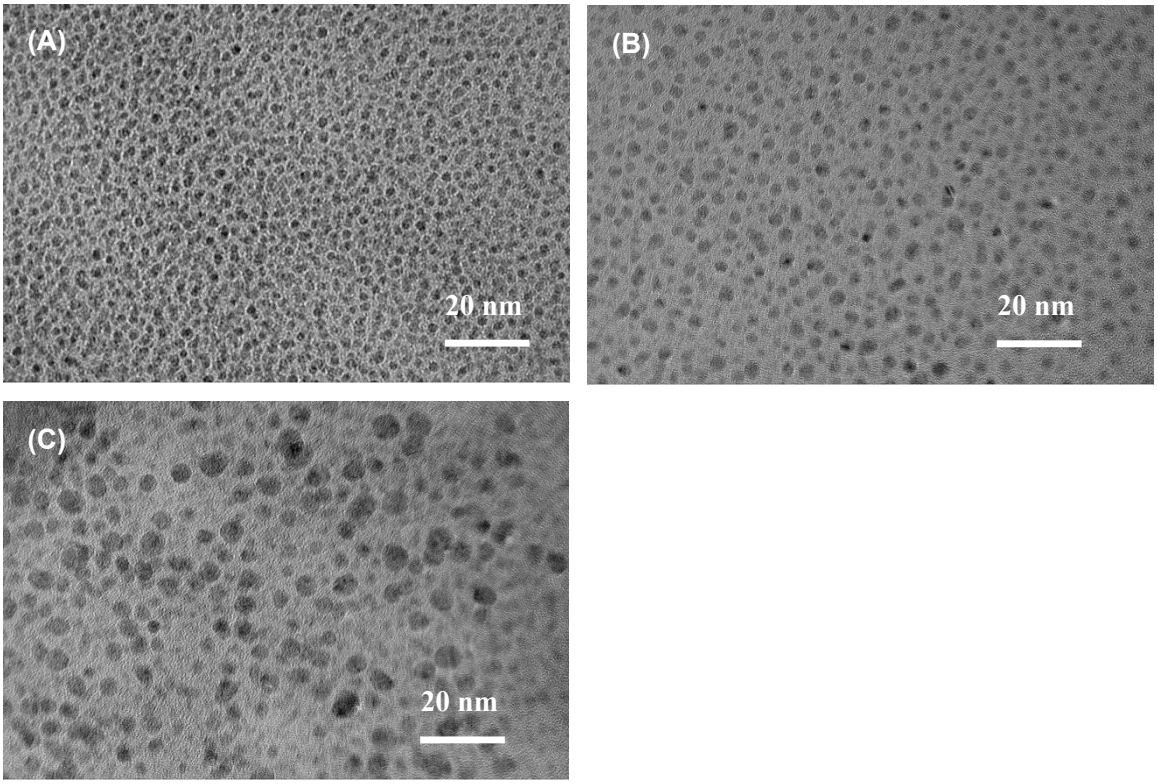

Fig. S3. Impact of (A) the concentration of Cu^{2+} and (B) the reduction time of Cu^{2+} by ascorbate on the fluorescent response of CuNPs; TEM images of dsDNA-templated CuNPs under different concentrations of Cu^{2+} with (C) 50 μM, (D) 100 μM, (E) 400 μM. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments. The average diameter scales of CuNPs under Cu^{2+} with 50 μM, 100 μM, and 400 μM are around 2.0 nm, 4.3 nm, 3.2 nm. The corresponding fluorescent quantum yields are 0.5%, 1.2%, 0.8%, respectively.

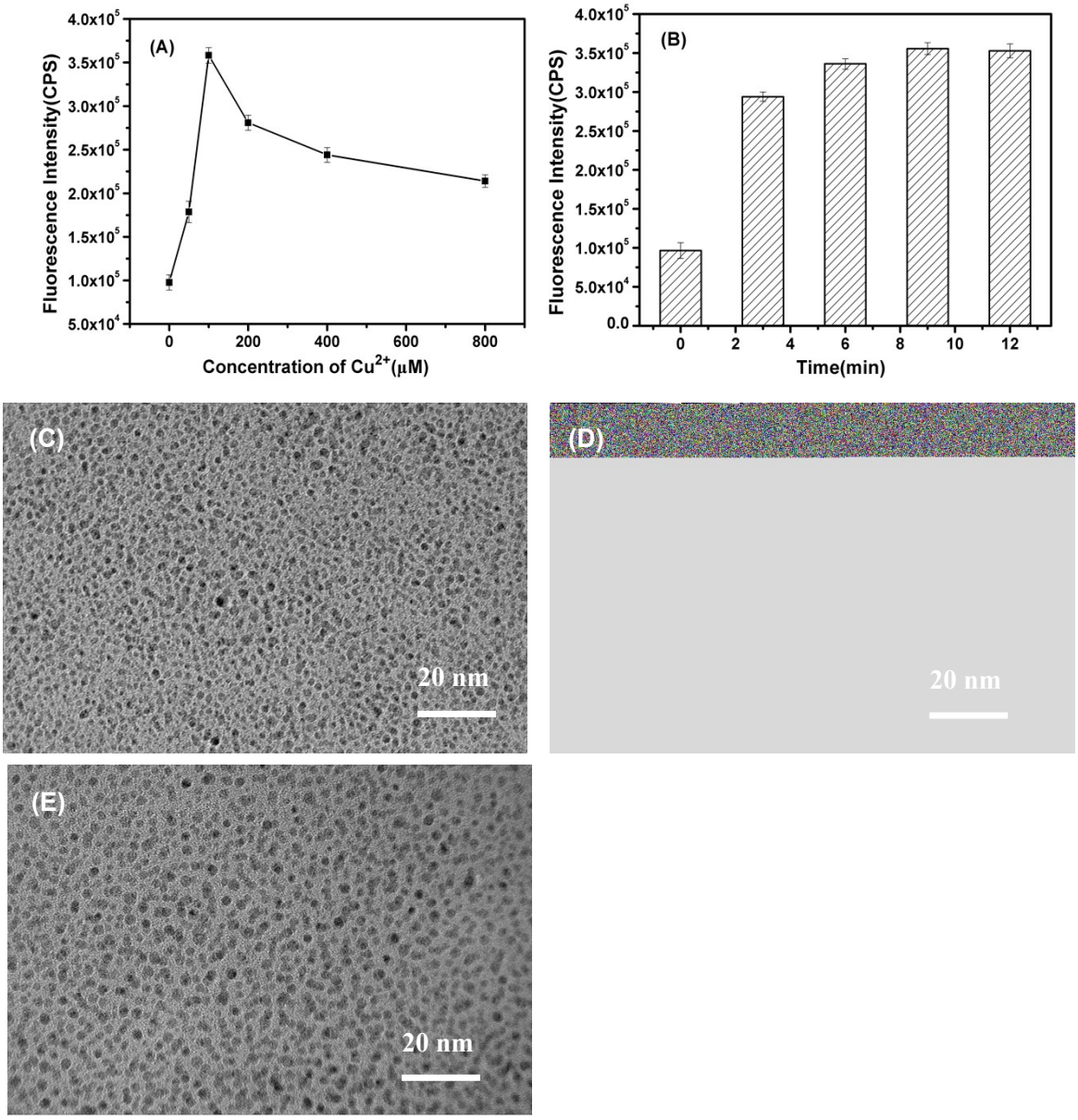

Fig. S4. Impact of the enzyme reaction time on the fluorescent response of the dsDNA-templated fluorescent CuNPs. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments.

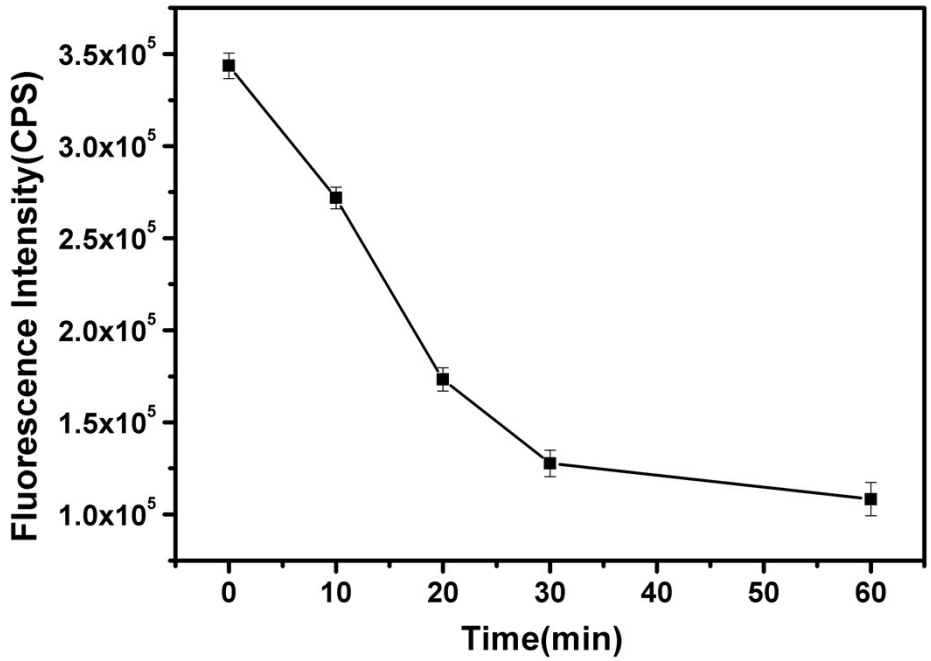

Fig. S5. Fluorescent responses for the blank sample, 20 U mL^{-1} M.Sss I MTase, 20 U mL^{-1} AluI MTase, and 20 U mL^{-1} Dam MTase. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments.

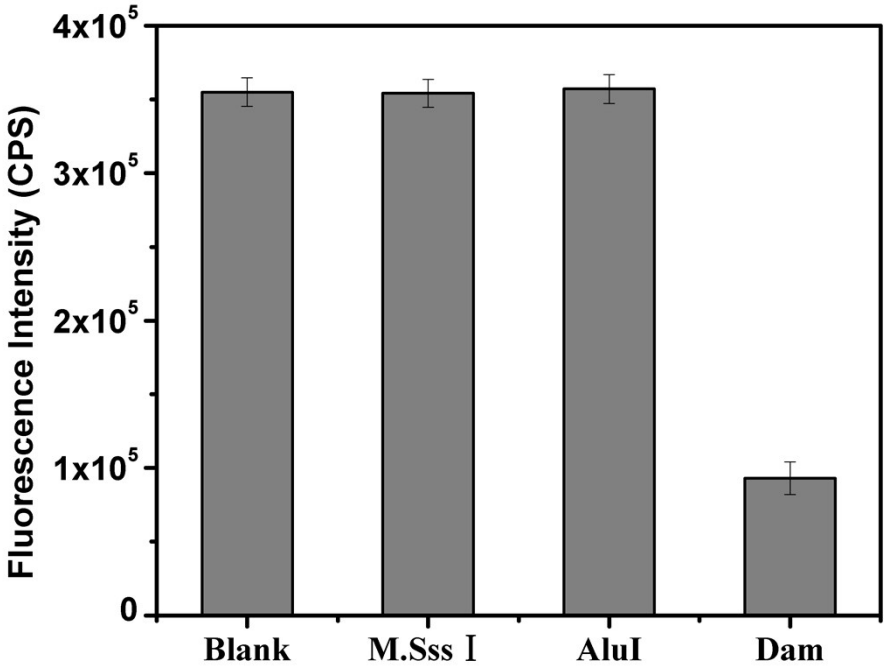

Fig. S6. Calibration curve for Dam MTase detection in diluted serum sample. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments.

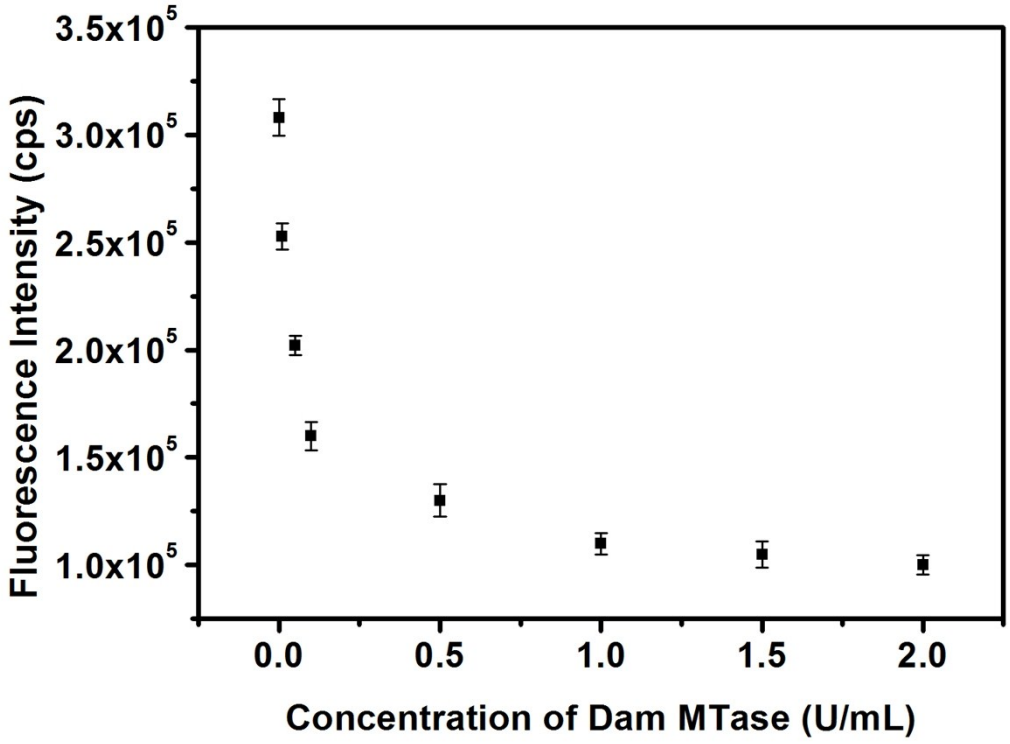

Fig. S7. The effect of various concentrations of 5-flurouracil on the relative activity of

DpnI. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments.

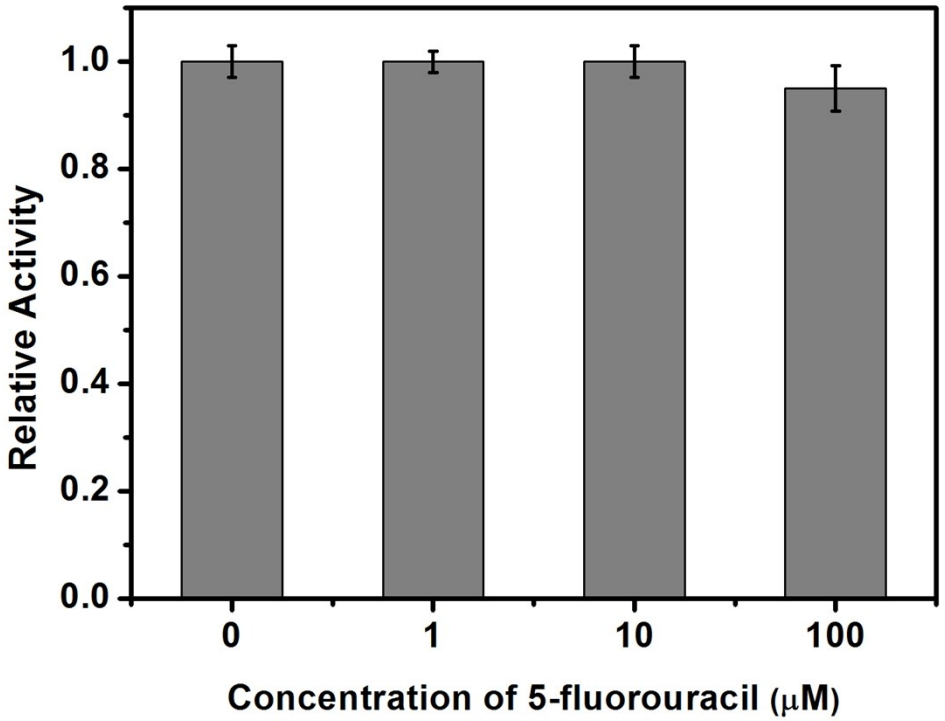

Fig. S8. Influence of different drugs on the activity of Dam MTase. (1) No drug, (2)

gentamycin, (3) benzylpenicillin, (4) ampicillin sodium, and (5) 5-fluorouracil. The concentration of all drugs was 0.5 μM. Other reaction conditions are as Fig. **2**. Error bars show the standard deviation of five experiments.

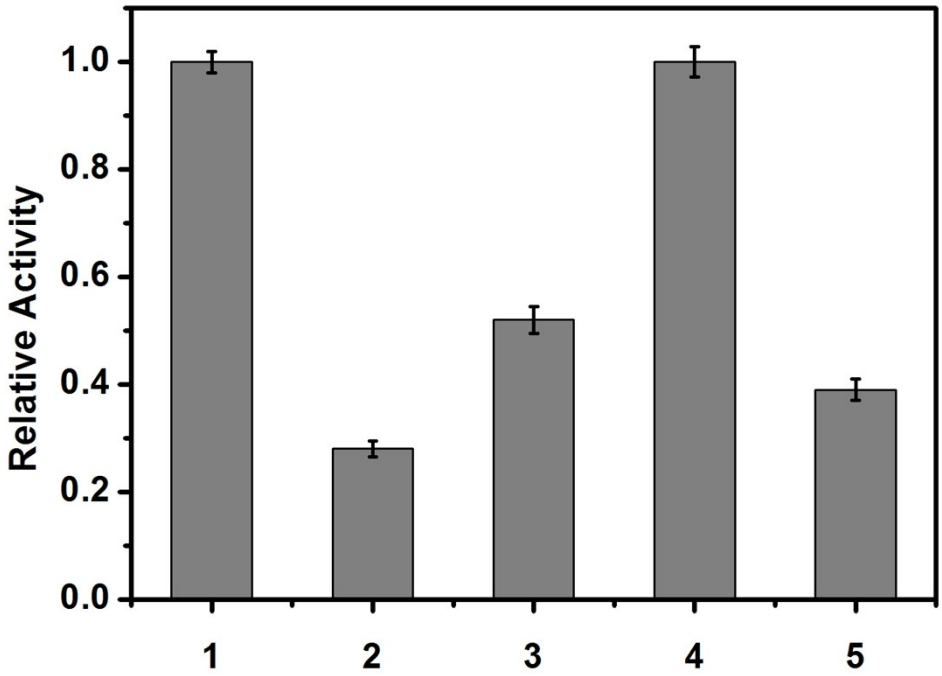

Table S2. Comparison of the performances of the developed strategy for MTase activity assay with the reported literatures.

Detection method	Linear range (U/mL)	LOD (U/mL)	Ref
A Novel Label-Free Fluorescent Strategy for Methyltransferase Activity Assay based on dsDNA-Templated Copper Nanoparticles Coupled with Endonuclease Assisted Signal Transduction System	0.01 to 5	0.01	Present work
DNA-AuNPs based signal amplification for highly sensitive detection of DNA methylation, methyltransferase activity and inhibitor screening	0.075–30	0.02	Jing et al. 2014
Highly sensitive fluorescence assay of DNA methyltransferase activity via methylation-sensitive cleavage coupled with nicking enzyme-assisted signal amplification	—	0.06	Zhao et al. 2013
A bioluminescence assay for DNA methyltransferase activity based on methylation-resistant cleavage	0.2–100	0.08	Jiang et al. 2012
Electrochemical strategy for sensing DNA methylation and DNA methyltransferase activity	0.25–10	0.18	Wang et al. 2013
An electrochemical one-step system for assaying methyltransferase activity based on transport of a quantum dot signaling tracer	1.0–128	0.79	Baek et al. 2013

Table S3. Recovery experiments of Dam MTase in diluted serum samples

Samples	Added Dam MTase (U mL^{-1})	Detected Dam MTase[#] (U mL^{-1})	Recovery (%)
1	0.1	0.095+±0.01	95.0
2	0.5	0.49±0.03	98.0
3	1.0	1.02±0.06	102.0
4	1.5	1.48±0.06	98.7

[#] Average of five experiments ± standard deviation.