Electronic Supplementary Information

Target-Initiated Synthesis of Fluorescent Copper Nanoparticles for Sensitive and Label-Free Detection of Bleomycin

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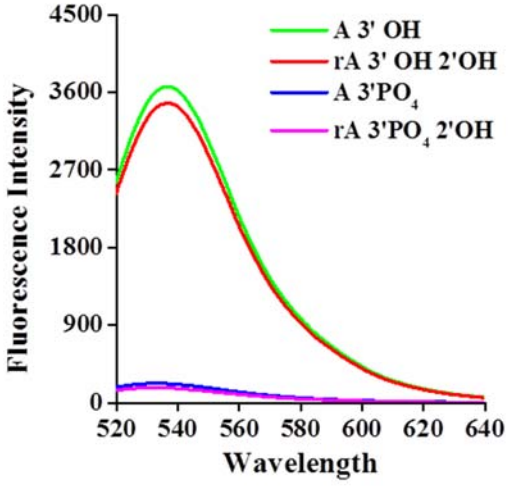

Fig. S1 Measurement of fluorescence emission spectra in response to 1 μM synthetic sequence. The synthesized sequence is used to mimic the DNAzyme-catalyzed cleavage product and performs the TdTase-assisted base extension. The sequence is 5'-GCG CCG CCG CAA AAT TCA CCA ACT AT X-3'. The underlined base (X) indicates the deoxyribonucleoside adenosine (A, green line), the ribonucleoside adenosine (rA, red line), the A

modified with PO_4 at the 3' terminal (A 3'-PO_4, blue line), and the rA modified with PO_4 at the 3' terminal (rA 3'-PO_4, magenta line), respectively. In the presence of the synthetic sequence without 3'-OH ends (blue line and magenta lines), low fluorescence signals are observed. In contrast, the enhanced fluorescence signals are detected in the presence of the synthetic sequences with 3'-OH ends (green line and red line) as a result of the staining of poly-T products by SYBR Gold. Moreover, the same fluorescence signals are observed in the presence of A (green line) and rA (red line). These results demonstrate that TdTase can incorporate a number of dNTPs to the 3'-OH ends of DNA and RNA.

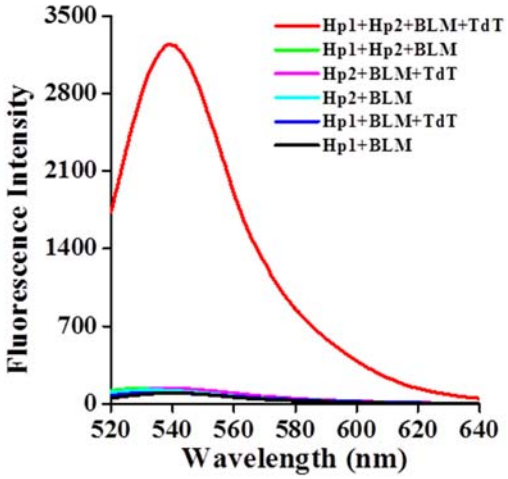

Fig. S2 Measurement of fluorescence emission spectra under different experimental conditions. The BLM concentration is 100 nM. The 1 μM Hp1, 4 μM Hp2 and 10 U of TdTase were used in the experiments. In the presence of Hp1 + BLM (black line), Hp1 + BLM + TdTase (blue line), Hp2 + BLM (cyan line), Hp2 + BLM + TdTase (magenta line), Hp1 + Hp2 + BLM (green line), low fluorescence signals are observed as a result of the staining of Hp1 and Hp2 by SYBR Gold. In contrast, an enhanced fluorescence signal is detected in the presence of Hp1 + Hp2 + BLM + TdTase (red line) as a result of the staining of poly-T products by SYBR Gold. These results demonstrate that the TdTase-assisted base extension reaction can only be triggered by target BLM in the presence

of specific Hp1 and Hp2.

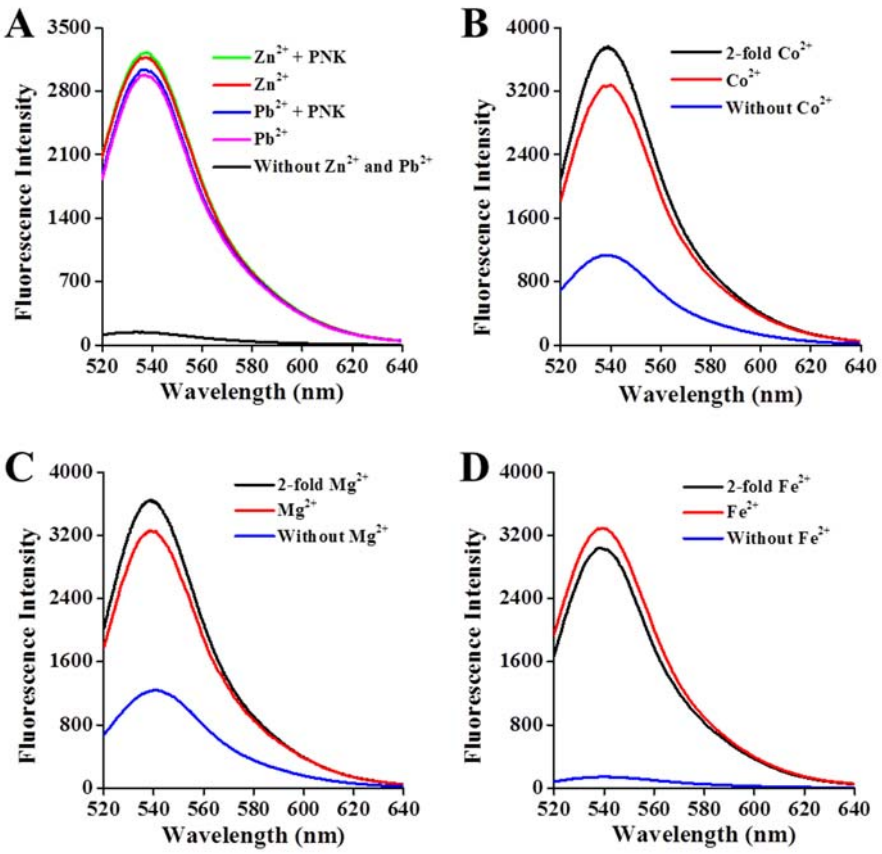

Fig. S3 (A) Measurement of fluorescence emission spectra in the absence of Zn^{2+} and Pb^{2+} (black line) and presence of 20 μM Pb^{2+} (magenta line), 20 μM Pb^{2+} + 2 U of PNK + 1 mM ATP (blue line), 10 μM Zn^{2+} (red line), and 10 μM Zn^{2+} + 2 U PNK + 1 mM ATP (green line), respectively. (B) Measurement of fluorescence emission spectra in the absence of Co^{2+} (blue line) and presence of 0.25 mM Co^{2+} (red line) and 0.5 mM Co^{2+} (black line), respectively. All buffer contains 10 μM Zn^{2+}, 10 mM Mg^{2+} and 100 nM Fe^{2+}. (C) Measurement of fluorescence emission spectra in the absence of Mg^{2+} (blue line) and presence of 10 mM Mg^{2+} (red line) and 20 mM Mg^{2+} (black line), respectively. All buffer contains 10 μM Zn^{2+}, 0.25 mM Co^{2+}, and 100 nM Fe^{2+}. (D) Measurement of fluorescence emission spectra in the absence of Fe^{2+} (blue line) and presence of 100 nM Fe^{2+} (red line) and 200 nM Fe^{2+} (black line). All buffer contains 10 μM Zn^{2+}, 0.25 mM Co^{2+}, and 10 mM Mg^{2+}. The BLM concentration is 100

nM. The 1 μM Hp1, 4 μM Hp2 and 10 U of TdTase were used in the experiments.

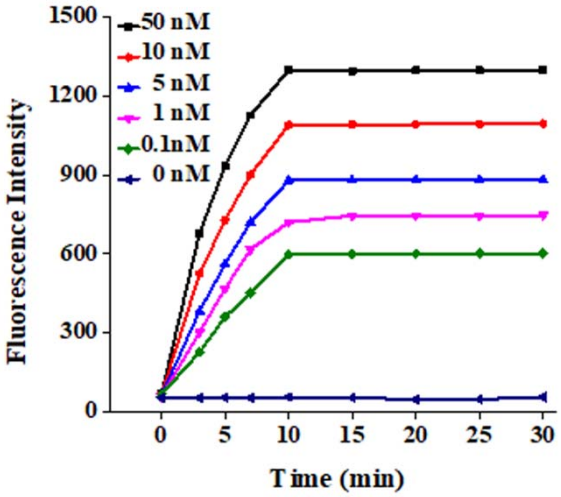

Fig. S4 Real-time monitoring of the oxidative cleavage of Hp1 induced by different-concentration BLM.

Table S1. Comparison of the proposed method with the reported methods for BLM assay

strategy	assay time*	requirement of labels	read out	linear range	LOD	ref.
electrochemical assay	~8 h	electrochemical label (ferrocene)	Turn-off	100 pM - 100 μM	100 pM	1
electrochemical assay based on dual-amplification	over 16h	electrochemical label(MBA-AuNPs/DA-AuNPs)	Turn-off	0.07-910 nM	0.02 nM	2
electrogenerated chemiluminescence assay	~12 h	no	Turn-off	0.1-50 pM	0.03 pM	3
colorimetric assay	~20 min	no	Turn-on	25 nM - 1 μM	16 nM	4
fluorescence quenching	no data	no	Turn-off	0.09-2.0 μg/mL	40 ng / mL	5
exo III-aided DNA recycling amplification-based fluorescent assay	~40 min	fluorescent label (FAM and BHQ)	Turn-on	0.1 nM - 1 μM	23 pM	6
WS_2 nanosheet quencher-based fluorescent assay	~23 min	fluorescent label (FAM)	Turn-on	0.5 nM - 1 μM	0.3 nM	7
perylene derivative quencher-based fluorescent assay	~12 h	fluorescent label (FAM)	Turn-on	5-500 nM	0.2 nM	8
silver nanocluster-based fluorescent assay	~83 min	no	Turn-off	100-500 nM	54 nM	9
enzymatic polymerization-mediated synthesis of CuNPs-based fluorescent assay	100 min	no	Turn-on	1 fM - 5 nM	8.1×10^{-16} M	this work

*Assay time includes the preparation time. MBA-AuNPs/DA-AuNPs: 4-mercaptophenyl boronic acid-capped gold nanoparticles and dopamine-capped gold nanoparticles. WS_2: tungsten disulfide. LOD: limit of detection.

References

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