Electronic Supplementary Information

Random dsDNA-templated formation of copper nanoparticles as novel fluorescence probes for label-free lead ions detection

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Experimental Section

Oligonucleotides and chemicals

Random dsDNA with different length and base composition were synthesized by Shanghai Sangon Biotechnology Co. Ltd. (Shanghai, China). The oligonucleotide sequences were listed in Table S1. All DNA templates were prepared with MOPS buffer (10 mM, pH 7.5; NaAc, 150 mM). Sodium ascorbate, $Cu(Ac)_2$, $Pb(Ac)_2$, NaAc and other used metal salts were commercially available and at least analytical grade. All solutions were prepared with Milli-Q water (18.25 MΩ cm^{-1}) from Millipore system.

Table S1 Sequences of the DNA templates used in this study

Name	Sequences	Length
DNA1	5'-ATGAACGTATGAGCG-3'	15-mer
DNA2	3'-TACTTGCATACTCGC-5'	15-mer
DNA3	5'-AGTTGCAAGAAGATGACAGAGAAGT-3'	25-mer
DNA4	3'-TCAACGTTCTTCTACTGTCTCTTCA-5'	25-mer
DNA5	5'-TACTCATACGCTCATACGTTCATCACGACTACACA-3'	35-mer
DNA6	3'-ATGAGTATGCGAGTATGCAAGTAGTGCTGATGTGT-5'	35-mer

Synthesis of fluorescent copper nanoclusters (CuNPs) in dsDNA templates

Fluorescent CuNPs were prepared according to a reported method with some modifications.[1,2] Aqueous solution (500 μL) containing 500 nM dsDNA (DNA1-DNA2, DNA3-DNA4, or DNA5-DNA6) in the buffer consisting of MOPS (10 mM, pH 7.5), NaAc (150 mM), and sodium ascorbate (1 mM) was heated to 90 ^{o}C and slowly cooled down to room temperature. Then, 100 μM $Cu(Ac)_2$ was added. The formation of fluorescent CuNPs was completed within 20 min as judged by fluorescence spectroscopy: increase of the fluorescence intensity at 585 nm (λ_{ex} = 340 nm). The concentration of the as-prepared dsDNA-CuNPs was denoted as "1X".

Fluorescence assay of Pb^{2+} ions

Pb^{2+} ions of different concentrations were mixed with the dsDNA-CuNPs solution. After a 5 min incubation, the fluorescence spectra of the dsDNA-CuNPs-Pb^{2+} system were recorded by a Perkin-Elmer LS-55 Fluorescence Spectrometer (Perkin-Elmer, USA) using a 1-cm path length quartz cell at room temperature (λ_{ex} = 340 nm, λ_{em} = 585 nm).

Preparation of real samples

Different concentrations of Pb^{2+} ions in human urine samples were certified using the standard inductively coupled plasma/mass spectroscopy (ICP/MS) method by Guangzhou 12th Hospital, China. After centrifugation at 3000 g for 15 min, the supernatant containing Pb^{2+} ions was

collected and appropriately diluted with MOPS buffer (10 mM, pH 7.5) for analysis using the present sensing approach. River water samples collected from the Zhujiang River (Guangzhou, China) were filtered through a 0.2 μm membrane. Aliquots of the river water (40 μL) were spiked with different concentration of standard Pb^{2+} solution (10 μL), and then mixed with dsDNA-CuNPs solution (450 μL; final concentration, "1X"). The mixture was incubated at room temperature for 5 min and then fluorescence spectra were recorded under excitation at 340 nm.

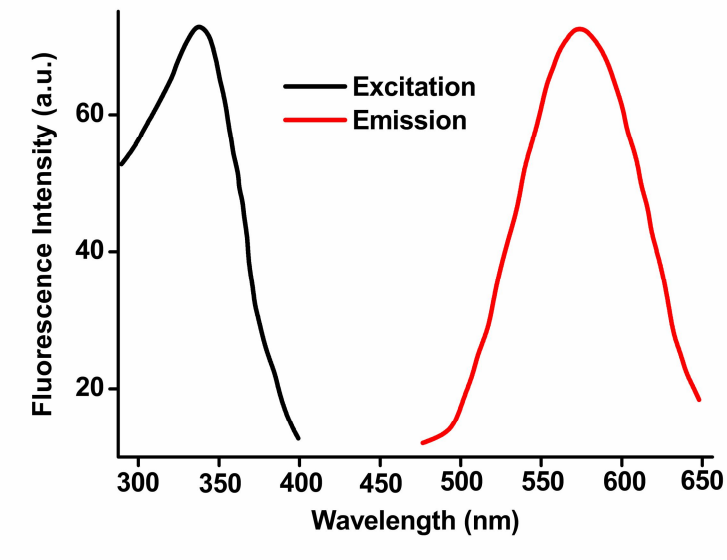

Fig. S1 Fluorescence excitation (black line) and emission (red line) spectra of the dsDNA-CuNPs.

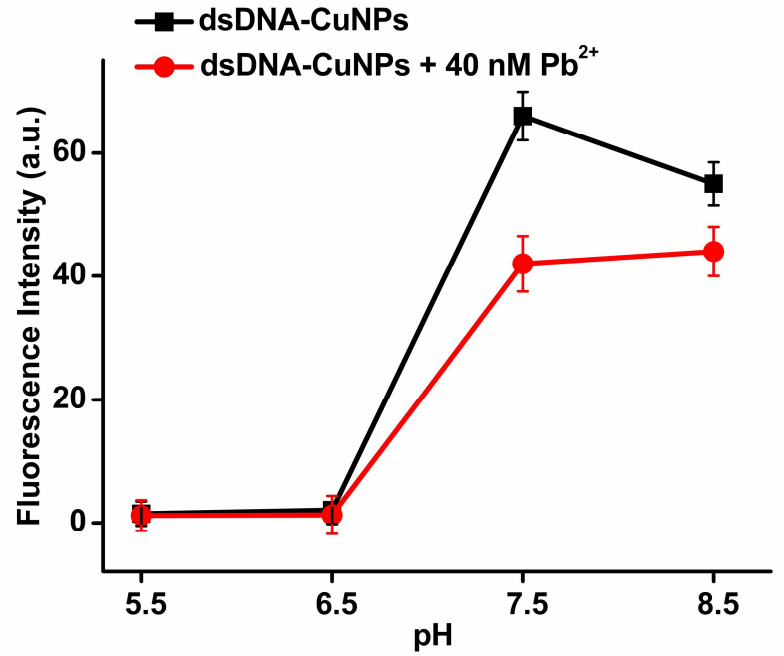

Fig. S2 Effects of pH on the fluorescence intensity of the dsDNA-CuNPs in the absence (black line) and presence of 40 nM Pb^{2+} (red line), respectively. (dsDNA, 500 nM; Cu^{2+}, 100 μM; sodium ascorbate, 1 mM; 10 mM MOPS buffer with various pH was adjusted by hydrochloric acid or NaOH; excitation wavelength, 340 nm; emission wavelength, 585 nm).

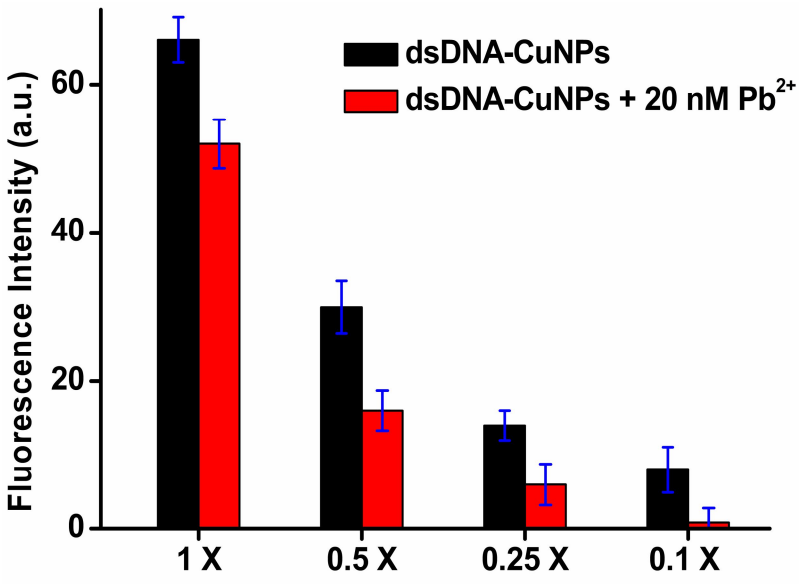

Fig. S3 The fluorescence intensity of different concentration of the dsDNA-CuNPs in the absence (black column) and the presence (red column) of 20 nM Pb^{2+}. The concentration of the as-prepared dsDNA-CuNPs is denoted as "1X".

Table S2 Determination of Pb^{2+} (nM) in human urine samples

Urine samples	Certified value[a]	dsDNA-CuNPs probe[b]	Relative error (Re)[c] (%)
1	12.5	13.2 ± 1.6	+5.6
2	34.7	33.2 ± 2.1	-4.3
3	56.2	59.7 ± 4.2	+6.2
4	83.8	91.5 ± 5.3	+9.2

[a]The concentration of Pb^{2+} in human urine samples was certified using the standard inductively coupled plasma/mass spectroscopy (ICP/MS) method by Guangzhou 12th Hospital, China.

[b]Average of five determinations ± standard deviation.

[c]Re: dsDNA-CuNPs probe *vs.*ICP/MS method.

Table S3 Recovery experiments of Pb^{2+} in river water samples

River water	Pb^{2+} spiked (nM)	Pb^{2+} founded[a] (nM)	Recovery (%)
1	10	10.8 ± 1.3	108
2	20	17.2 ± 1.8	86
3	40	38.5 ± 3.4	96.3
4	80	84.1 ± 5.7	105.1

[a]Average of five determinations ± standard deviation.

References

1 A. Rotaru, S. Dutta, E. Jentzsch, K. Gothelf and A. Mokhir, *Angew. Chem. Int. Ed.*, 2010, **49**, 5665.

2 Z. Zhou, Y. Du and S. Dong, *Anal. Chem.*, 2011, **83**, 5122.