Supplementary Information

Histone–DNA interaction: an effective approach to improve the fluorescence intensity and stability of DNA-templated Cu nanoclusters

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Experimental details

Reagents and Materials: All DNA molecules designed in this study were commercially synthesized by Sangon Biological Engineering Technology and Services Co., Ltd (Shanghai, China). Histone (from calf thymus, type III-S), bovine serum albumin (BSA), thrombin,single-stranded DNA binding protein (SSB), protamine sulfate were purchased from Sigma-Aldrich Chemical company (St. Louis, MO, USA). $CuSO_4$ and ascorbic acid were obtained from Sinopharm Chemical Reagent Company (Beijing, China). All other chemicals were of analytical-reagent grade and obtained from standard reagent suppliers. Millipore Milli-Q water (18 M Ω cm) was used in all experiments.

Apparatus: Fluorescence measurements were carried out using an F-4600 Fluorescence spectrophotometer (Hitachi, Japan). Ultraviolet spectra were performed on a U-3900H UV-Visible spectrophotometer

(Hitachi, Japan). Transmission electron microscopy (TEM) was conducted using a Tecnai G2 F20 electron microscope at an acceleration voltage of 200 kV with a CCD camera. X-ray photoelectron spectroscopy (XPS) measurements were performed by using an AXIS ULTRA Spectrometer (Kratos Analytical Ltd.). The quantum yield measurement was carried out by C9920-02G QY measurement system (Hamamatsu, Japan). Circular dichroism (CD) measurements were performed on a MOS-450 circular dichroism spectrometer. Dynamic light scattering (DLS) measurements were performed with an ALV-5000/E DLS instrument (Nano-ZS90, Malvern) at 90°. Time-correlated single-photon counting measurements (FLS920, Edinburgh Instruments) were used to study the lifetime of the DNA-templated CuNCs and histone/DNA-templated CuNCs on quartz substrates. All pH measurements were made with Thermo Scientific Orion Star A111 desktop pH meter.

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Synthesis of DNA-templated CuNCs and hestone/DNA-templated CuNCs: Firstly, in order to prepare dsDNA templates, ssDNA and the

complementary strand were mixed at a 1:1 molar ratio in morpholineopropanesulfonic acid (MOPS) buffer (10 mM MOPS, 150 mMNaCl, pH 7.6). The mixture was annealed at 90 °C for 10 min, and then slowly cooled down to room temperature (25 °C). Before use, diluted to the desired concentrations with the same MOPS buffer. In a typical synthesis experiment, 40 μL ascorbic acid (10 mM) was added into the 20 μL DNA templates, and 20 μL ultrapure water to give a total volume of 80 μL. After the mixtures were stirred for 1 min, 20 μL $CuSO_4$ (2.5 mM) was added to the mixture solution. Then the mixed solution was kept for 10 min at room temperature, and then the fluorescent CuNCs were formed. For histones/DNA template, 20 μL histones (5 mg/mL) was mixed with DNA and allowed to react for 1 min at room temperature (25 °C). Other steps were the same as the preparation of DNA-templated CuNCs described above.

Polyacrylamide gel electrophoresis analysis: A 15 % denaturing polyacrylamide gel was prepared using 1 × TBE buffer (89 mMTris-boric acid, 2.0 mM EDTA, pH 8.3). 20 μL of the DNA solution (DNA, histones/DNA) along with 2 μL 6 × loading buffer was loaded onto a 15 % denaturing polyacrylamide gel and then run in 1 × TBE buffer at 70 V for 90 min. The gels were stained with silver for 30 min. The visualization and photography were performed using a molecular imager with a Gel Doc system.

Table S1.Sequences of the used oligonucleotides

Oligonucleotide	Sequence (5'to 3')
dsDNA-2-(AT)50%	5'-*T*GCC*TA*CG*A*GG*AATT*CC*ATA*-3' 5'-*TAT*GG*AATT*CC*T*CG*TA*GGC*A*-3'
dsDNA-3-(AT)90%	5'-*AATAATAA*GC*TATAATAATT*-3' 5'-*AATTATTATA*G*CTTATTATT*-3'
dsDNA-4-(AT)8	5'-ATATATATATATATAT-3'
dsDNA-5-(AT)10	5'-ATATATATATATATATATAT-3'
dsDNA-6-(AT)12	5'-ATATATATATATATATATATATAT-3'
dsDNA7-(AT)15	5'-ATATATATATATATATATATATATATATAT-3'
ssDNA-1-(T)20	5'-TTTTTTTTTTTTTTTTTTTT-3'
ssDNA-2-(T)30	5'-TTTTTTTTTTTTTTTTTTTTTTTTTTTTTT-3'

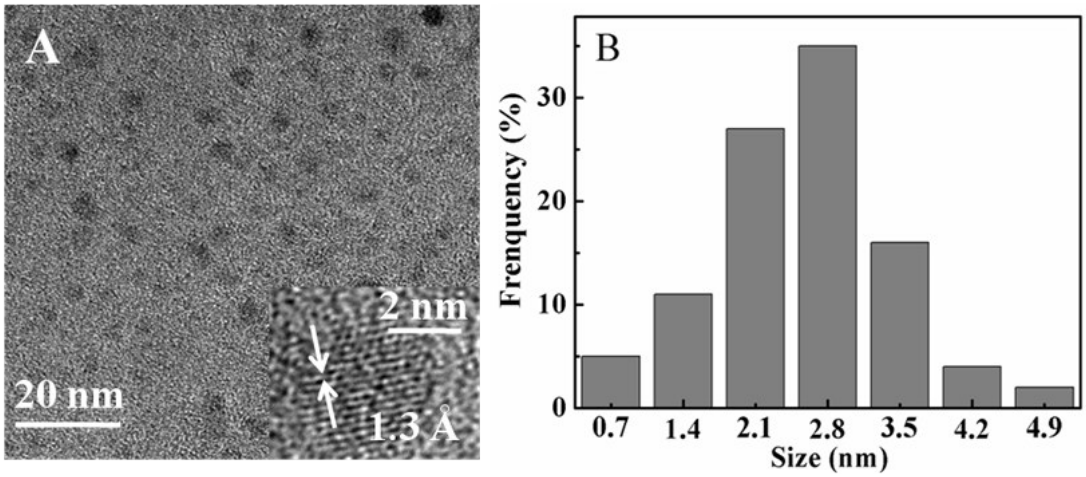

Figure S1 (A) TEM characterization of DNA-templated CuNCs, HR-TEM image of a single CuNCs (inset). (B) The particle size distribution histogram of histones/DNA-CuNCs.

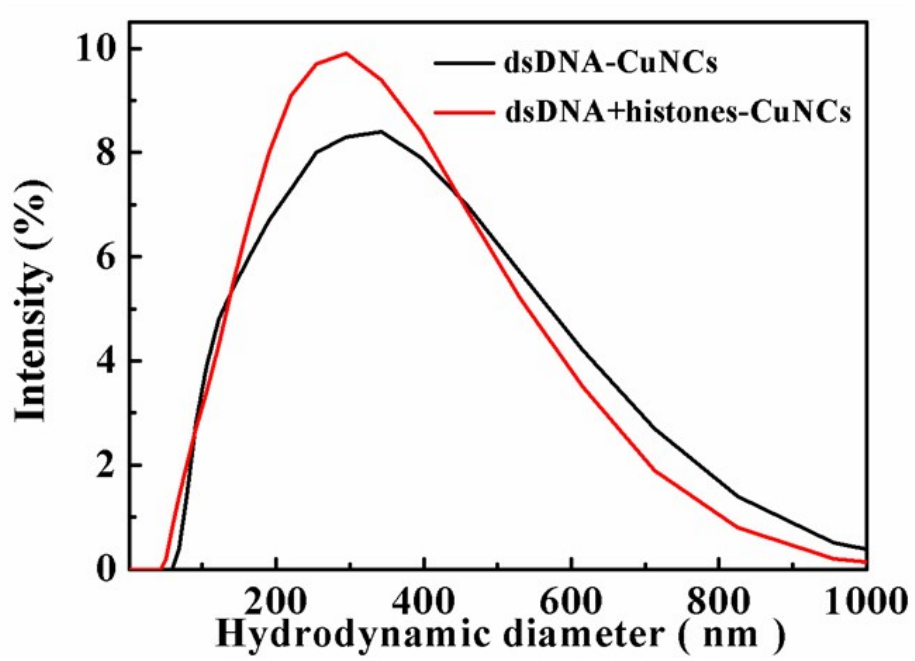

Figure S2 DLS results of CuNCs.

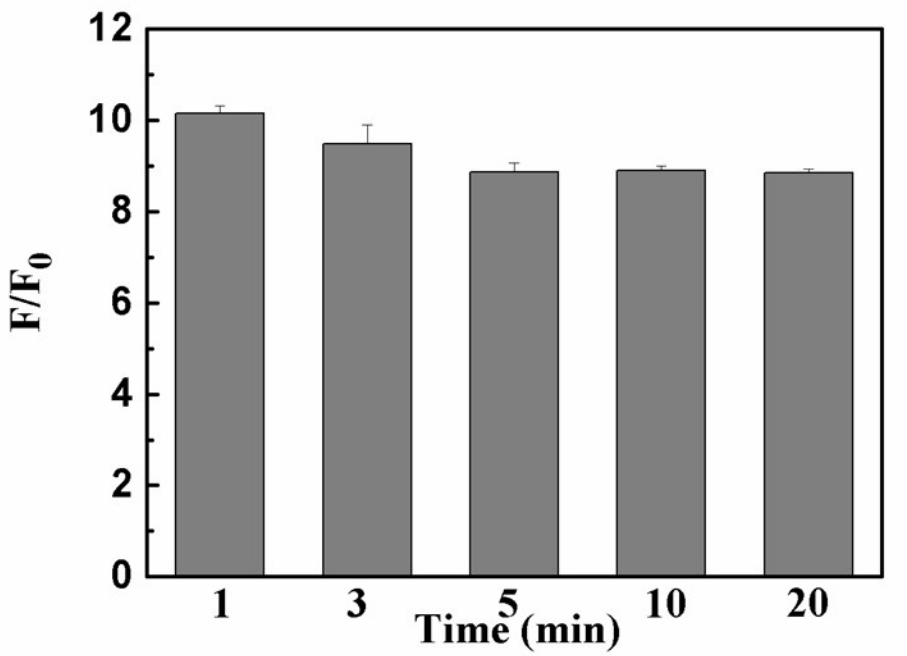

Figure S3 Effect of histone-DNA interaction time on the fluorescence intensity of CuNCs. The error bars represent the standard deviation of three independent measurements.

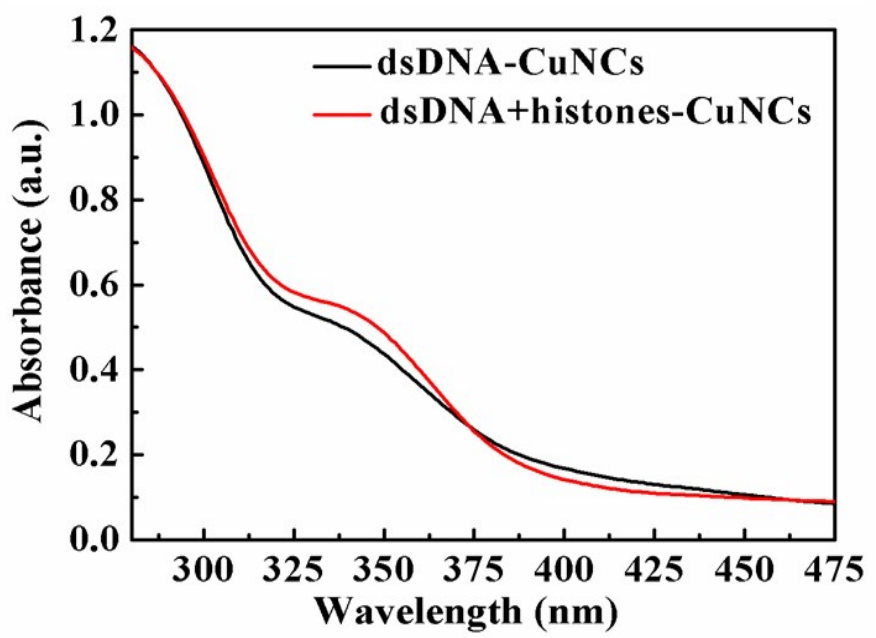

Figure S4 Absorption spectra of the CuNCs synthesized under the same conditions.

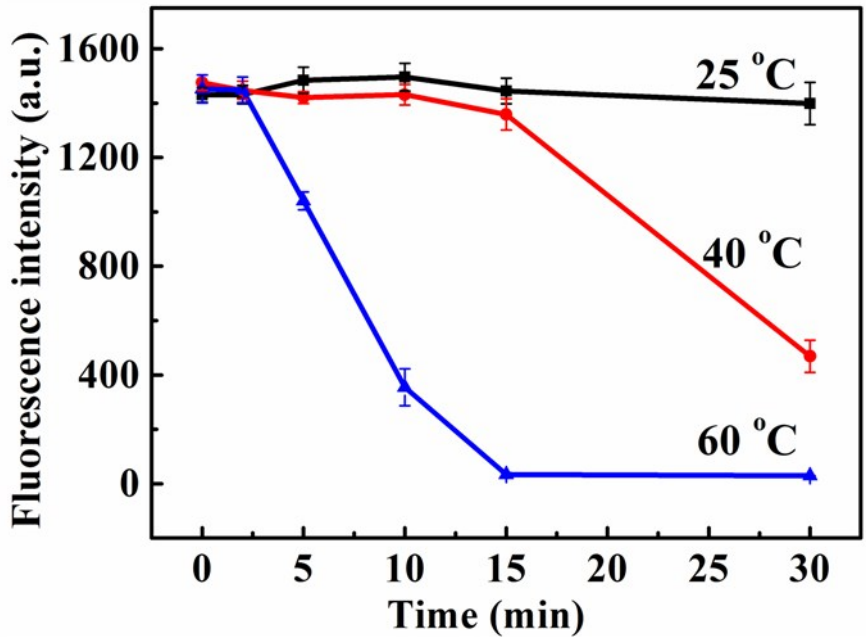

Figure S5 Effect of temperature on the fluorescent stability of histone/DNA-templated CuNCs. The error bars represent the standard deviation of three independent measurements.

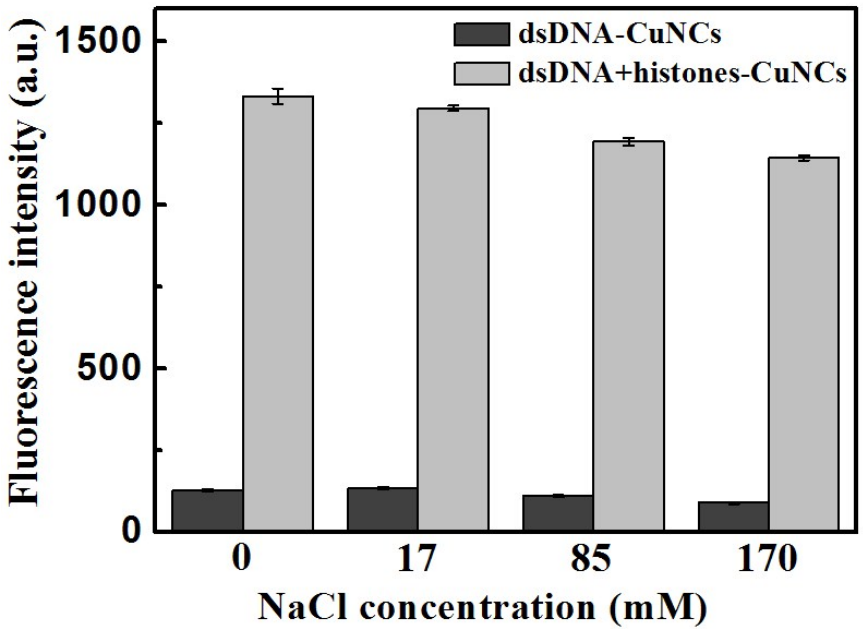

Figure S6 Fluorescence intensity of this CuNCs in high concentration of NaCl. The error bars represent the standard deviation of three independent measurements.

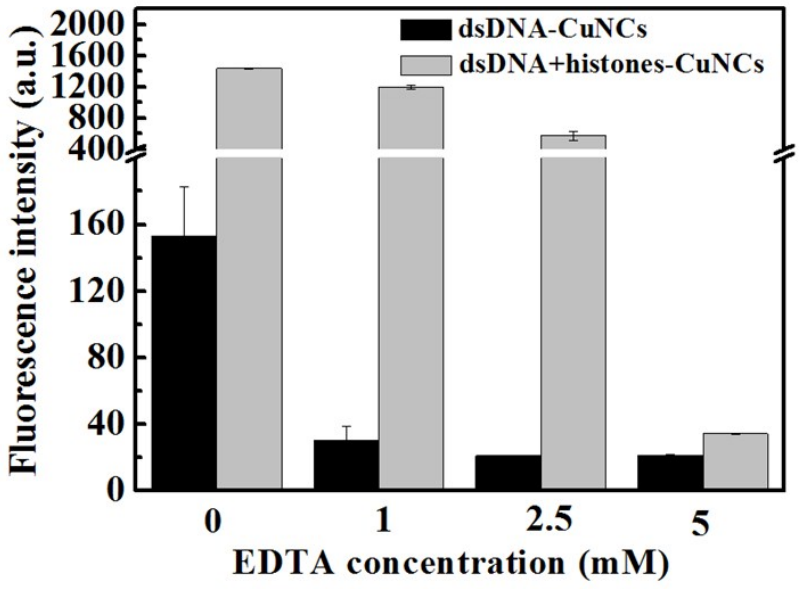

Figure S7 Effect of EDTA on the fluorescence of CuNCs. The error bars represent the standard deviation of three independent measurements.

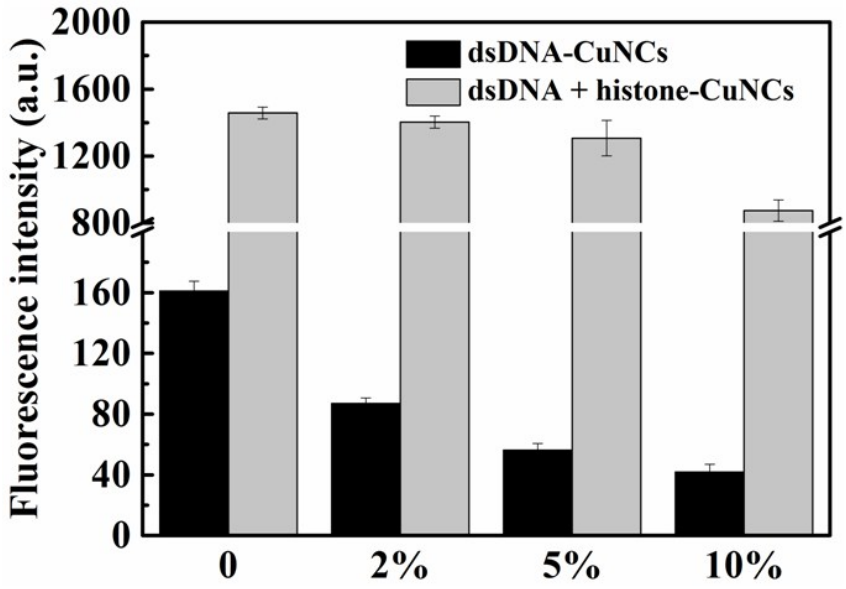

Figure S8 Fluorescence intensity of these CuNCs in the diluted serum with different concentration (v/v). The error bars represent the standard deviation of three independent measurements.

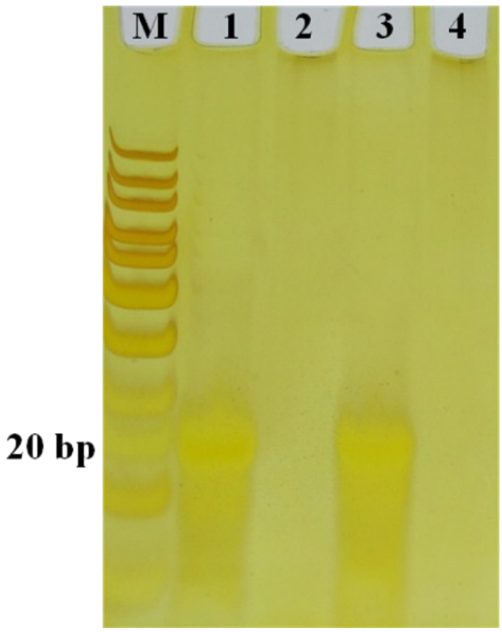

Figure S9 Gel electrophoresis verification of DNA-histone interactions. Lane 1: dsDNA 5; Lane 2: dsDNA5+ histones; Lane 3: DNA5-CuNCs; Lane 4:DNA5+histones-CuNCs

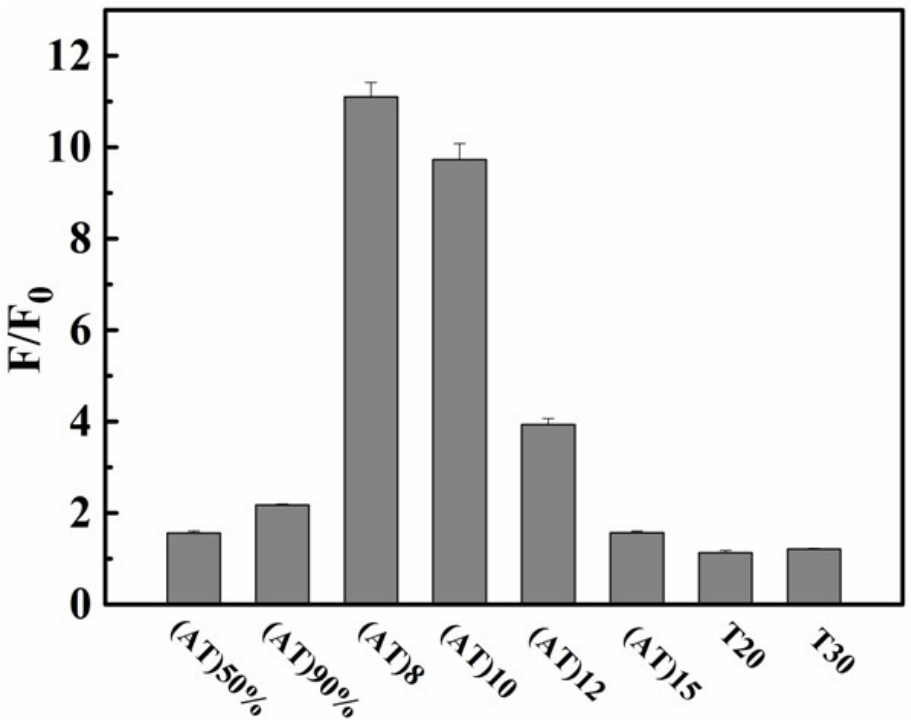

Figure S10 Effect of histones on the CuNCs with different DNA templates. The error bars represent the standard deviation of three independent measurements.

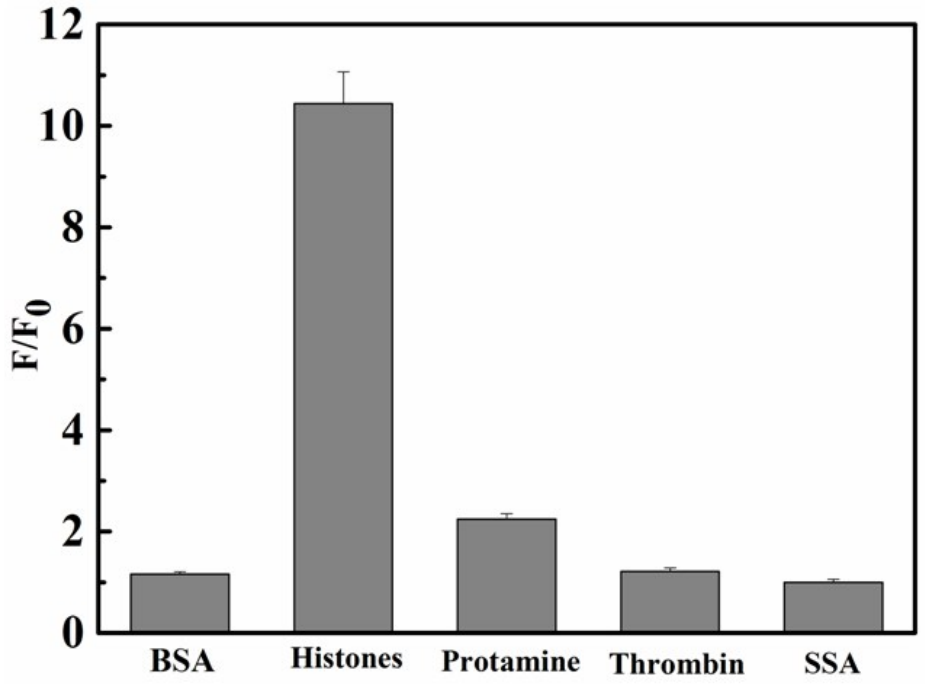

Figure S11 Effect of protein on the fluorescence intensity of DNA-templated CuNCs. Histones concentration was 5 mg/mL, and the concentrations of other proteins were 20 mg/mL. For BSA, thrombin or SSA, 20 μL protein (20 mg/mL) was mixed with DNA and allowed to react for 1 min at room temperature (25 °C), and other steps were the same as the preparation of DNA-templated CuNCs. For protamine, after DNA-templated CuNCs formed, 20 μL protamine (20 mg/mL) was added. The error bars represent the standard deviation of three independent measurements.

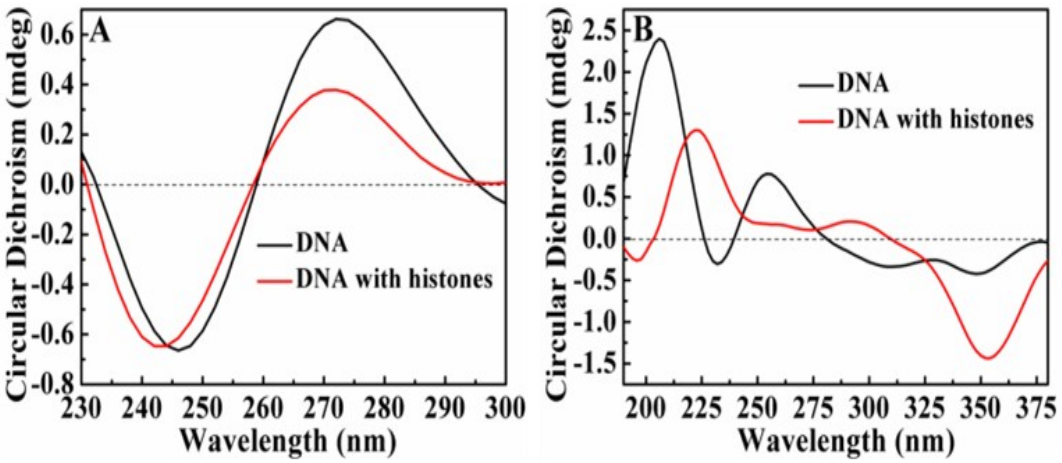

Figure S12 CD spectra of DNA in the absence (A) and presence (B) of ascorbic acid and Cu^{2+}. Experimental conditions: 30 μL dsDNA (5 μM), 30 μL histones (5 mg/mL), 60 μL sodium ascorbate (10 mM), 30 μL $CuSO_4$ (2.5 mM).

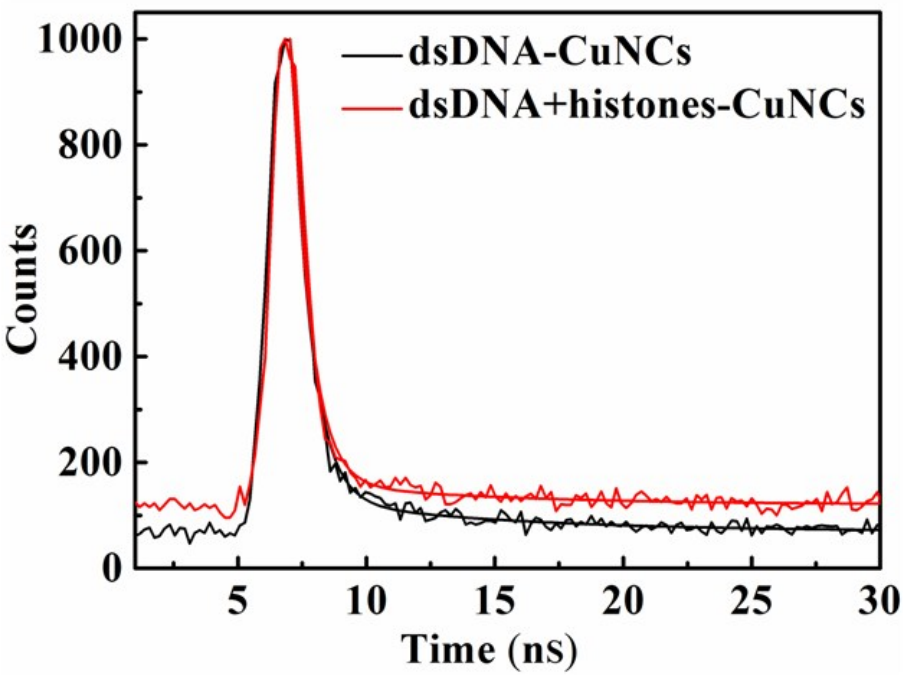

Figure S13 Fluorescence lifetime decay of CuNCs

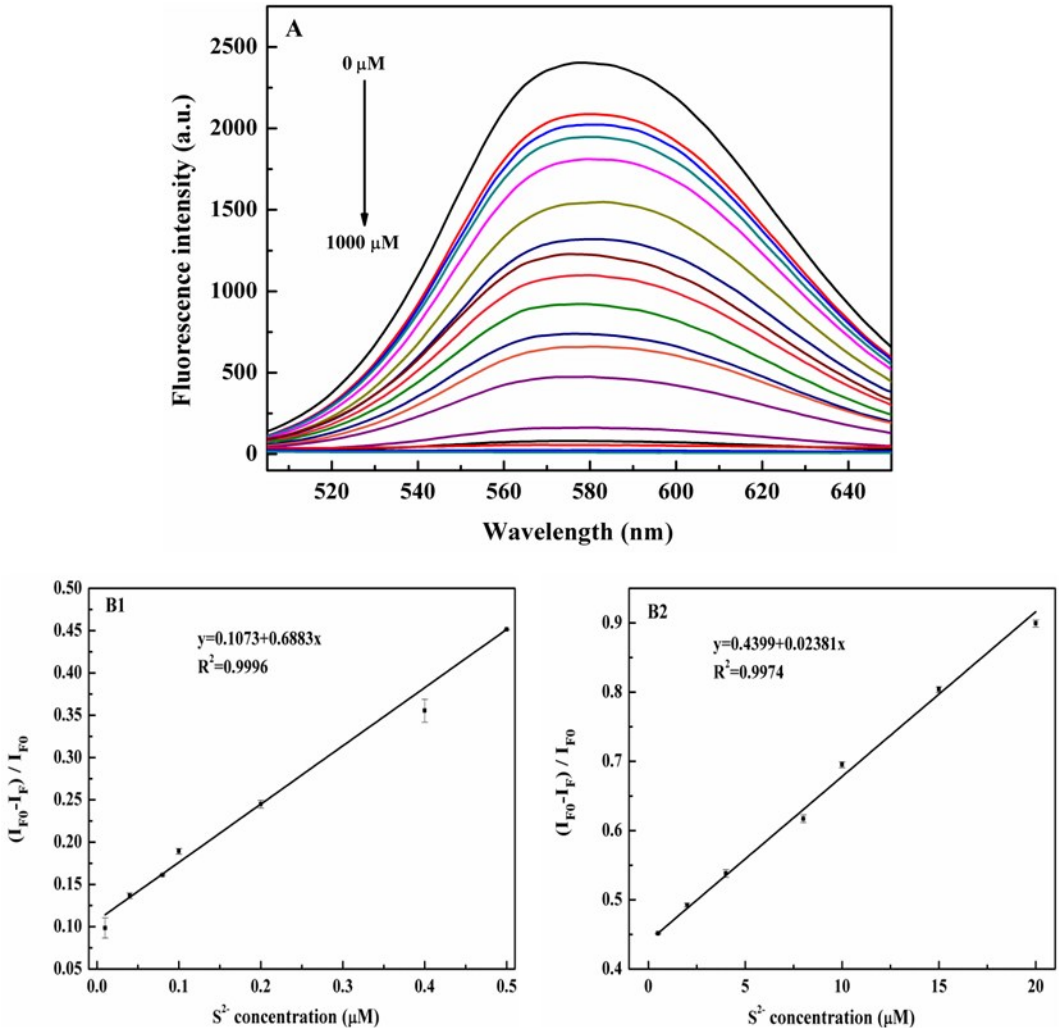

Figure S14 (A) Fluorescence emission spectra of the DNA+histone-CuNCs with the addition of different concentrations of S^{2-} ranging from 0 to 1000 μM (0, 0.01, 0.04, 0.08, 0.1, 0.2, 0.4, 0.5, 1.0, 2.0, 4.0, 8.0, 10, 15, 20, 50, 100, 200, 500, 1000 μM). (B1-B2) Linearity of relative fluorescence intensity $[(I_{F0}-I_F)|/I_{F0}]$ of the sensor against the S^{2-} concentrations. Experimental conditions: 20 μL 5 μM dsDNA (DNA 5), 20 μL 5mg/mL histones, 40 μL sodium ascorbate (10 mM), 20 μL $CuSO_4$ (2.5 mM).

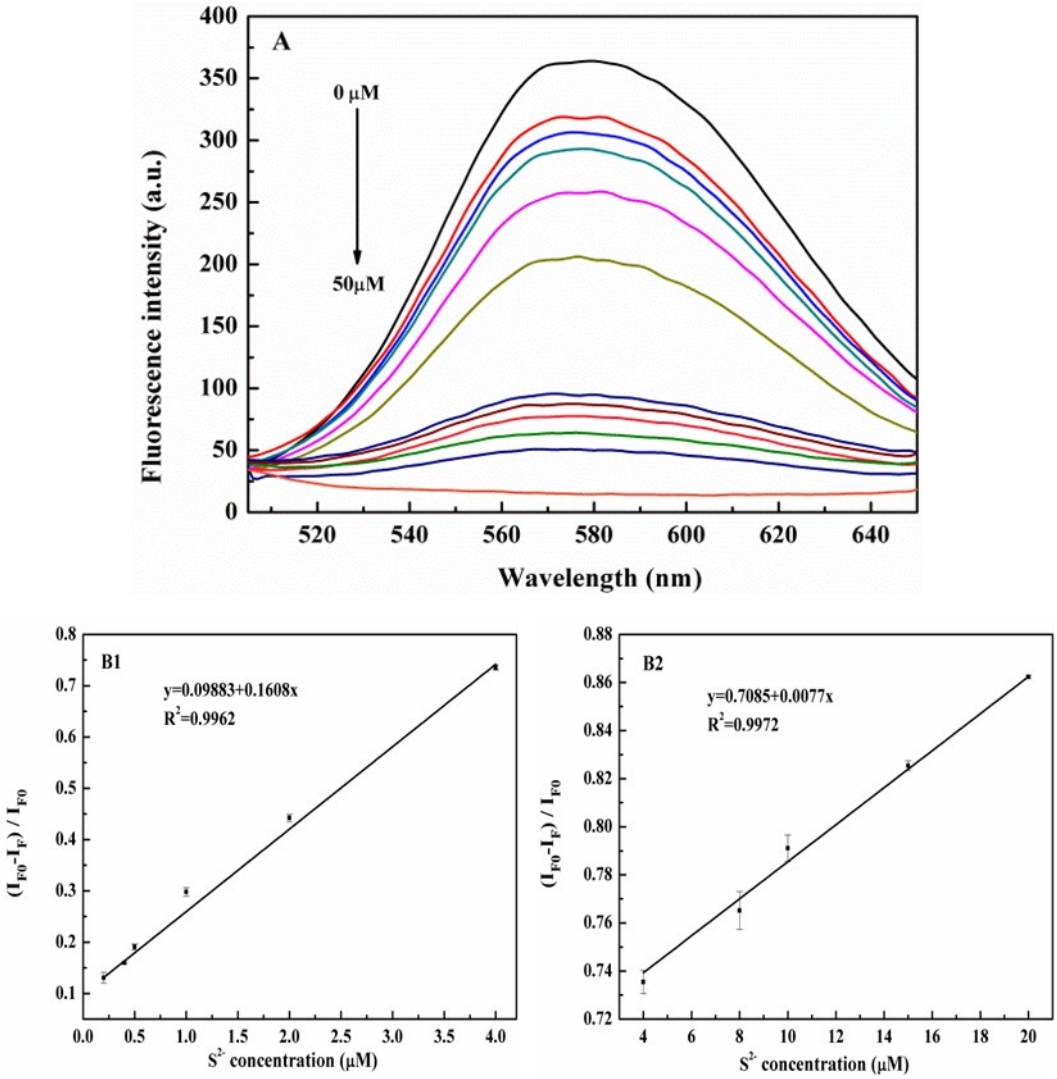

Figure S15 (A) Fluorescence emission spectra of the DNA-CuNCs with the addition of different concentrations of S^{2-} ranging from 0 to 50 mM (0, 0.2, 0.4,0.5, 1.0, 2.0, 4.0, 8.0, 10, 15, 20, 50 mM). (B1-B2) Linearity of relative fluorescence intensity $[(I_{F0}-I_F)|/I_{F0}]$ of the sensor against the S^{2-} concentrations. Error bars were estimated from three independent measurements. Experimental conditions: 20 μL 5 μM dsDNA (DNA 5), 40 μL sodium ascorbate (10 mM), 20 μL $CuSO_4$ (2.5 mM).