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Electronic Supplementary Information

Luminescent silver nanoclusters anchored by oligonucleotides: detect human telomerase ribonucleic acid template sensitively†

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S1 Materials and Synthesis of AgNC-S0

Silver nitrate (≥99.9%, ACS reagent) and sodium borohydride (98%) were purchased from Alfa Aesar. HPLC-purified oligonucleotides were synthesized by TaKaRa Biotechnology (Dalian, China). Hairpin oligonucleotides (5'-CCCCCCCC-ATGCA-GTTAGGGTTAG-TGCAT-3') were termed S0. The HR (5'-CUAACCCUAAC-3') is the model target. A random sequence of oligonucleotides (5'-CCAUCGCCAUC-3', termed RO) was also synthesized to test the specificity of AgNC-S0. The high-purity deionized water (>18.3 MΩ cm) was produced by Millipore A10 Milli-Q. Phosphate buffer (analytical reagent, pH 7.0) was purchased from Sigma–Aldrich Ltd. All reagents were used without further purification.

Oligonucleotide S0 (30 μM) and silver nitrate (1 mM) were obtained by dissolving into the phosphate buffer (20 mM phosphate, 1 mM magnesium nitrate, pH 7.0). Magnesium ions were utilized to stabilize the oligonucleotides conformation. The concentration of S0 was quantitated by reading the absorbance at 260 nm based on the extinction coefficient of 266900 M^{-1} cm^{-1}, calculated from the website of Integrated DNA Technologies, Inc. (http://www.idtdna.com/analyzer/Applications/OligoAnalyzer/). The silver nitrate solution (180 μL) was added into S0 solution (450 μL) at 0 ^{o}C. After vigorously stirring for 15 minutes, fresh sodium borohydride solution (450 μL, 0.5 mM in deionized water) was added to reduce silver ions. To remove excessed silver nitrate, Amicon Ultra-0.5 mL centrifugal filter with the nominal molecular weight limit of 3K was used to purify the mixture with centrifugal speed of 4000 rpm for 6 hours. Then the unreacted S0 were further removed *via* size-exclusion chromatography (SEC). The purified sample was kept in the dark at 4 ^{o}C prior to further measurements.

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S2 Details of Fluorescence, UV-vis, MALDI-TOF and FTIR experiments

The luminescence measurements were carried out using Fluorolog-3 (FL3-21, HORIBA, Ltd.). The slit width is 5nm for original AgNC-S0 and 10nm for diluted one. The integration time is 0.1 second. Since free ions and unreacted S0 were both removed, the concentration of AgNC-S0 was represented by the concentration of S0. UV-vis spectrum was acquired with UV-1800 (Shimadzu Scientific Instruments) with slit width of 1 nm and quartz cuvette of 1 mm path length. The absorption spectrum of AgNC-S0 was simply fitting by the function of "Fitting Multi-peaks" with peak type of "Lorentzian" in the software of Origin. Matrix-assisted laser desorption/ionization (MALDI) mass spectra were measured by 4800 Plus MALDI TOF/TOF^{TM} Analyzer (AB Sciex) at positive ion mode. FTIR spectra were recorded using Nicolet 6700 FT-IR (Thermo Fisher Scientific Inc.) with spectral resolution of 2 nm. The Thermo Scientific MicroCL 21R centrifuge was used for centrifugation of AgNC-S0 to condense the solutions to the appropriate concentration for the FTIR and MALDI mass spectra with the centrifugal speed of 4000 rpm.

S3 Reproducibility and long-term stability of AgNC-S0

The synthesis of AgNC-S0 was repeatedly, as shown in Fig. 1a. Fig. 1b showed the long-term stability of purified AgNC-S0. The fluorescence of the purified AgNC-S0 was stable for 2 weeks.

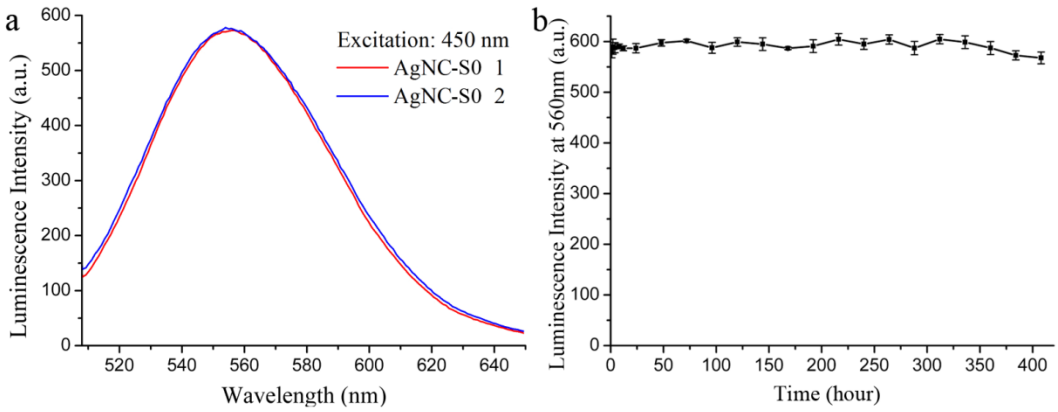

Fig. 1 (a) The luminescence spectra of two AgNC-S0 samples. (b) The luminescence intensity of purified AgNC-S0 at 560 nm plotted with time.

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S4 Linear relation of AgNC-S0

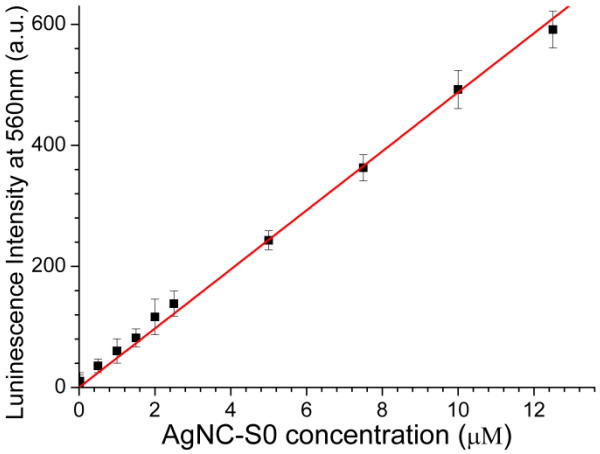

Fig. 2 Linear relation between the fluorescence intensity and concentration of AgNC-S0. The linear range was from 25nM to 12.5μM.

S5 Specificity of AgNC-S0

The sample matrix was prepared by dissolving the oligonucleotides into the phosphate buffer (20 mM phosphate, 1 mM magnesium nitrate, pH 7.0). A new sequence of oligonucleotides (5'-C**C**A**U**C**G**C**C**A**U**C, termed RO)with five similar base of HR was used to examine the specificity of AgNC-S0. The concentrations of random sequence oligonucleotides and HR were both 12.5 μM, and their volumes were both 150 μL.

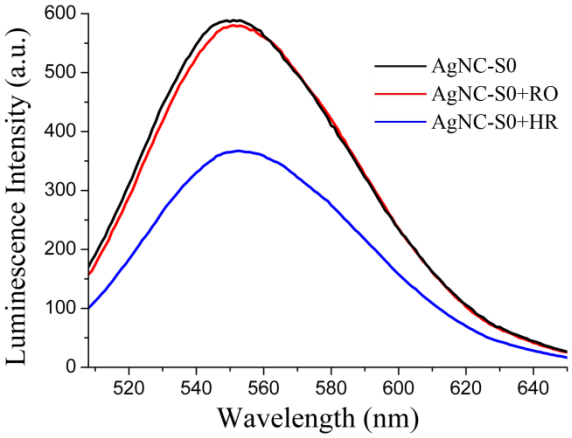

Fig. 3 The spectra of AgNC-S0 before and after adding RO and HR respectively. HR induced the notable decrease whereas RO didn't.

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S6 Sensitivity of AgNC-S0

The dose-effect relationship between the luminescence and HR was detected ranging from 25 nM to 250 nM (n=6). The concentration of AgNC-S0 was 250nM (20 mM phosphate, 1 mM magnesium nitrate, pH 7.0). The standard deviations of these measurements were used for the calculation of limit of detection (LOD), which is three times of the standard deviation divided by the slope of the linear regression curve.

S7 Lifetimes of AgNC-S0 before and after the addition of HR

The photoluminescence decays of AgNC-S0 (12.5 μM, 150 μL) before and after the addition of HR (12.5 μM, 150 μL) were measured on a Lifespec Lifetime Spectrometer (Edinburgh Instruments Ltd.) with emission wavelength of 560 nm and time resolution of 0.02 ns. Both samples showed three lifetimes, which did not change after the addition the HR, respectively.

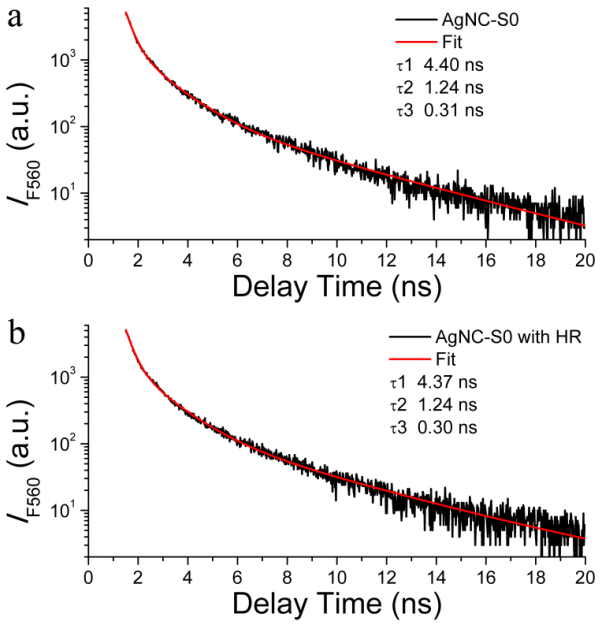

Fig. 4 The photoluminescence decays of AgNC-S0 before and after adding HR. AgNC-S0 showed three lifetimes: 4.40 ns, 1.24 ns and 0.31 ns, and AgNC-S0 with HR showed three lifetimes: 4.37 ns, 1.24 ns and 0.30 ns. All three lifetimes did not change after the addition of HR.