Electronic Supplementary Material (ESI) for ChemComm.

Evidence of Single-Nanoparticle Translocation through Solid-State Nanopore by Plasmon Resonance Energy Transfer

Yue Cao[a], Yao Lin[a], Ruo-Can Qian[a]*, Yi-Lun Ying[a]*, Wei Si[b], Jingjie Sha[b]*, Yunfei Chen[b], Yi-Tao Long[a]*

Key Laboratory for Advanced Materials & Department of Chemistry, East China University of Science and Technology, 130 Meilong Road, Shanghai, 200237, P. R. China.

Fax: +86 021-64250032

E-mail: ytlong@ecust.edu.cn

Index

Experimental Section

1. Reagents and materials.

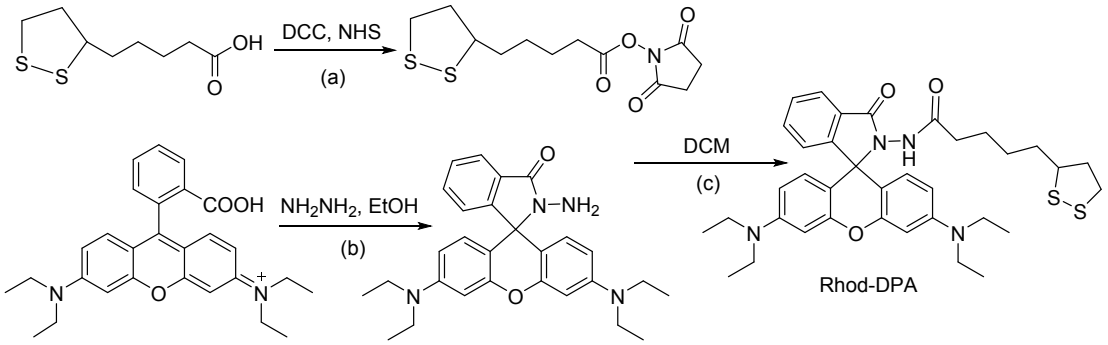
All reagents were of analytical grade. Gold chloride trihydrate ($HAuCl_4 \bullet 3H_2O$, >99.0%), sodium citrate were purchased from Sigma (USA). Absolute ethanol, acetone, $CuSO4 \bullet 5H_2O$, $Zn(NO_3)_2$, $Pb(NO_3)_2$, $Cd(NO_3)_2$, $CaSO_4$ and other materials were purchased from the Sinopharm Chemical Reagent Company (Shanghai, China).The indium tin oxide (ITO) slides were purchased from the Wuhan Geao Chemical Technology Co. Ltd (Wuhan, China). Polydimethylsiloxane (PDMS) was purchased from Dow Corning (Wiesbaden, Germany) which was curing with a mix ratio 1:10 at 100 °C for 30 min. All other chemical reagents and materials are of analytical grade, and solvents were purified by standard procedures. All solutions were prepared by Milli-Q ultrapure water with resistance of 18.2 M Ω cm at 25 °C (EMD Millipore, Billerica, USA) and were filtered with 0.02 mm Anotop filter (Whatman, Maidstone, UK).

2. Experimental setup and data acquisition.

The scanning electron microscope (SEM) observations of the AuNPs were performed with a field-emission scanning electron microscope (Ultra 55, Carl Zeiss Ltd., Germany). The transmission electron microscopic (TEM) images were obtained on a JEM-2010 high-resolution transmission electron microscope (JEOL Ltd., Japan) equipped for analysis at an accelerating voltage of 200 kV. UV-vis spectra were obtained from Ocean Optic. Time-of-flight secondary ion mass spectrometry (ToF-SIMS V, IONTOF GmbH, Germany). The chip with a nanopore was sandwiched between two silicone elastomer gaskets, and this assembly was positioned between two polytetrafluoroethylene (PTFE) flow cells using screws. A thin polydimethylsiloxane (PDMS) layer was painted and cured to both sides of silicone elastomer gasket. Before the assembly of the device, the chip is treated in oxygen plasma for 30s on both sides to increase the hydrophilicity of the nanopores. The electrolyte used was 10 mM potassium chloride (KCl) solution at pH 8.5. Two Pt electrodes were immersed into two electrolyte chambers respectively to apply a bias voltage and detect ionic current through the pore. The current trace was amplified and measured via a ChemClamp (Dagan Corporation, Minneapolis, USA) instrument with a 3 kHz low-pass Bessel filter. Data were acquired at a sampling rate of 100 kHz by using a DigiData 1440A converter and a PC running PClamp 10.4 (Axon Instruments, Forest City, USA). Data

analysis was performed using a home-designed software (http://people.bath.ac.uk/yl505/nanoporeanalysis.html)[1, 2] and Origin 9.2 (OriginLab Corporation, Northampton, USA). An inverted microscope (eclipse Ti-U, Nikon, Japan) with an external triple channel optical system was equipped with a 40× plan objective (NA=0.8), which was used to obtain the various spectra (the fluorescence spectra were obtained in a technological channel and the Raman spectra were measured in an external channel optical system). The dark-field condenser (0.8 < NA < 0.95, Nikon, Japan) with a 100 W halogen tungsten lamp was used for obtain the scattering light.

3. Preparation of Rhod-DPA.



(a) Synthesis of Rhodamine hydrazide

The Rhodamine hydrazide was synthesized according to the literature[3].

(b) Synthesis of N-Lipoyloxy succinimide

Lipoic acid (0.5 g, 2 mmol) and N-hydroxysuccinimde (0.23 g, 2 mmol) were dissolved in 4 mL dichloromethane, cooled at 0°C, then added dropwise to DCC solution (0.5 g, 2.5 mmol) in 3 mL dichloromethane. The mixed solution was stirred for 1 h, and then kept in refrigerator overnight to fully precipitate dicyclohexylurea (DCU). The precipitate was filtered and the filtrate was evaporated in vacuo. The residue was treated with diethyl ether and the formed yellowish solid was washed with ether to give the pure product (98%). The product 5 was used in the next step without further purification.

(c) Synthesis of Rhod-DPA

To an ice cold solution of Rhodamine hydrazide (0.45 g 1.0 mmol) in dichloromethane was added drop-wise N-lipoyloxy succinimide (0.35 g, 1.1 mmol) in 10 mL dichloromethane. The mixture was stirred at room temperature for 3 h, over then diluted with 30 mL CH_2Cl_2 and saturated Na_2CO_3. The organic layer was washed with saturated brine, and dried with anhydrous Na_2SO_4. The solvent was evaporated in vacuo and the residue was purified by silica gel columnchromatography to afford 2 (0.33 g,

89%) as white solid. 1H NMR (400 MHz, CDCl3) δ = 7.94 (dd, J = 4 Hz, 1H), 7.45 (t, J = 4 Hz, 2H), 7.11 (m, 1H), 6.44 (dd, J = 12, 4 Hz, 4H), 6.29 (dd, J = 8 Hz, 2H), 3.34 (dd, J = 8 Hz, 8H), 3.16 (m, 2H), 2.63 (t, J = 8 Hz, 2H), 2.48 (m, 1H) , 1.93 (m, 1H), 1.77 (m, 2H), 1.70 (m, 2H), 1.57 (m, 2H), 1.26 (t, J = 8 Hz, 1H), 1.17 (t, J = 6 Hz, 12H). 13C NMR (100 MHz, CDCl3) δ =169.18, 168.44, 166.17, 153.85, 151.57, 148.87, 132.54, 128.14, 128.11, 123.83, 108.02, 104.51, 97.94, 65.94, 56.10, 44.38, 40.17, 38.54, 34.43, 30.79, 28.34, 25.61, 12.62. MS (TOF-SIMS) m/z calcd for (M+H) $C_{36}H_{44}N_4O_3S_2$ 645.2888; found 645.2928.

4. Preparation of the AuNPs.

All the glassware was immersed in an aqua regia solution (HNO_3/HCl, 1:3) for 12h (caution: aqua regia is strong acid and is highly corrosive and should be handled with care) and then rinsed several times with ultrapure water before use. Seed AuNPs with diameters of 13nm were prepared according to a procedure which has been described previously.[4] In brief, 50 mL of 0.01% $HAuCl_4$ was added to a 100 mL round-bottomed flask that was equipped with a condenser. The solution was brought to a rolling boil under vigorous stirring, and 5 mL of 38.8 mM sodium citrate was rapidly added to the vortex of the solution. The addition of the sodium citrate changed the color of the solution from pale yellow to red. The solution was heated for 15 min and then stirred for an additional 15 min after the heating mantle had been removed. The resulting solution of seed particles was used to prepare the larger gold particles using a procedure that has been described previously.[5] In brief, 25 mL of water, 1 mL of the solution of seed particles and 100 μL of 0.2 M NH_2OH• HCl were combined in a 50 Ml beaker, and 3.0 mL of a 0.1% $HAuCl_4$ solution was added dropwise under vigorous stirring. As the $HAuCl_4$ solution was added, the color of the mixture gradually changed to dark red. The addition of the $HAuCl_4$ was completed within 5 min. The nanoparticle solutions were stored in dark bottles at 4 °C.

5. Preparation of the samples.

The AuNPs-functionalized were then modified with Rhod-DPA on the surface of the AuNPs via the Au-S covalent bond after immersing in 500μL of 80 μm Rhod-DPA solution for 1h. The AuNPs was centrifuged for several times to remove unbound Rhod-DPA. After the AuNP probes were added the nanopore cell to the left side. The cleaned ITO slides was added the nanopore cell to the right side. The ITO was modified with AuNPs via electrostatic adsorption by placing them translocation of the AuNPs probes by for the end of experiments. The ITO was rinsed with pure water for several times to remove unbound Cu^{2+}, and then dried under a stream of ultrapure

nitrogen prior to the dark-field measurements.

6. Nanopore fabrication.

Nanopores were drilled in 100 nm thick low-stress silicon nitride membranes. And the low-stress membranes was deposited on silicon substrate by low-pressure chemical vapor deposition (LPCVD). Next, 100 μm by 100 μm windows were fabricated through the wafer using photolithography and wet-etching methods. Then the silicon nitride membrane is milled to reduce the film thickness from 100 nm to about 20 nm by focused ion beam (FIB) operated in a dual beam microscope (Helios 600i NanoLab, FEI Company, Hillsboro, USA). Finally, a 100 nm nanopore is drilled in the chip using focused ion beam as describe in previous studies.[6]

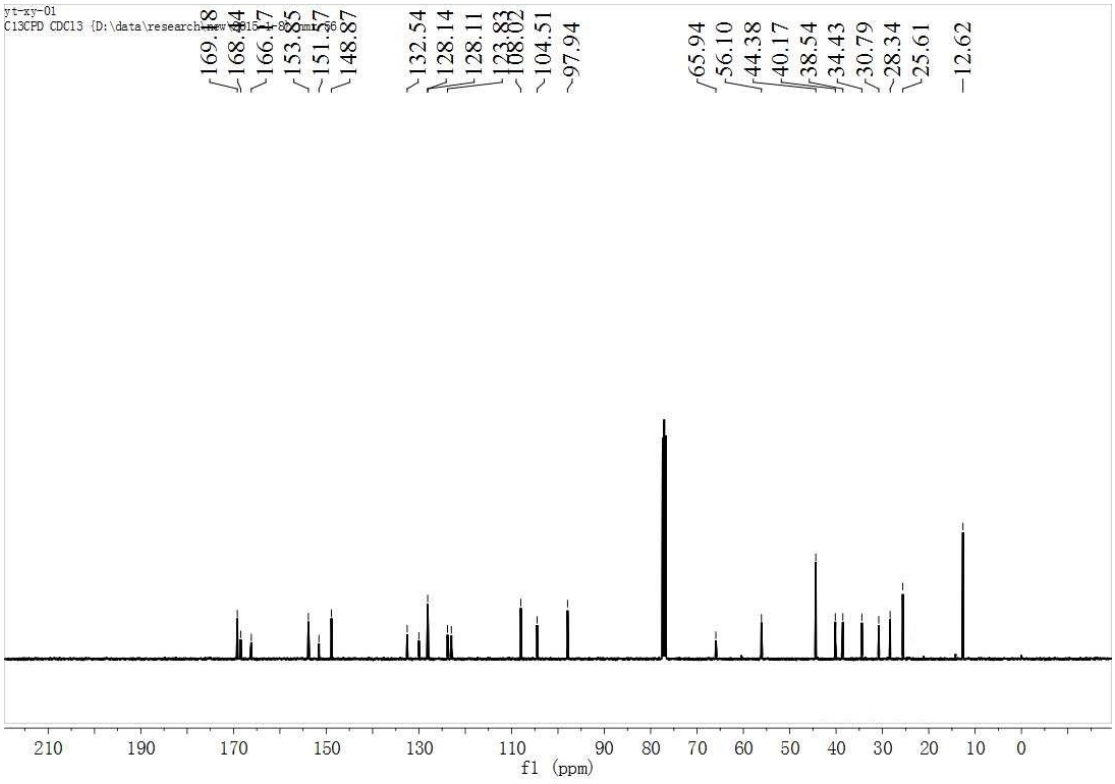


Figure S1. ^{13}C NMR spectrum of Rhod-DPA.

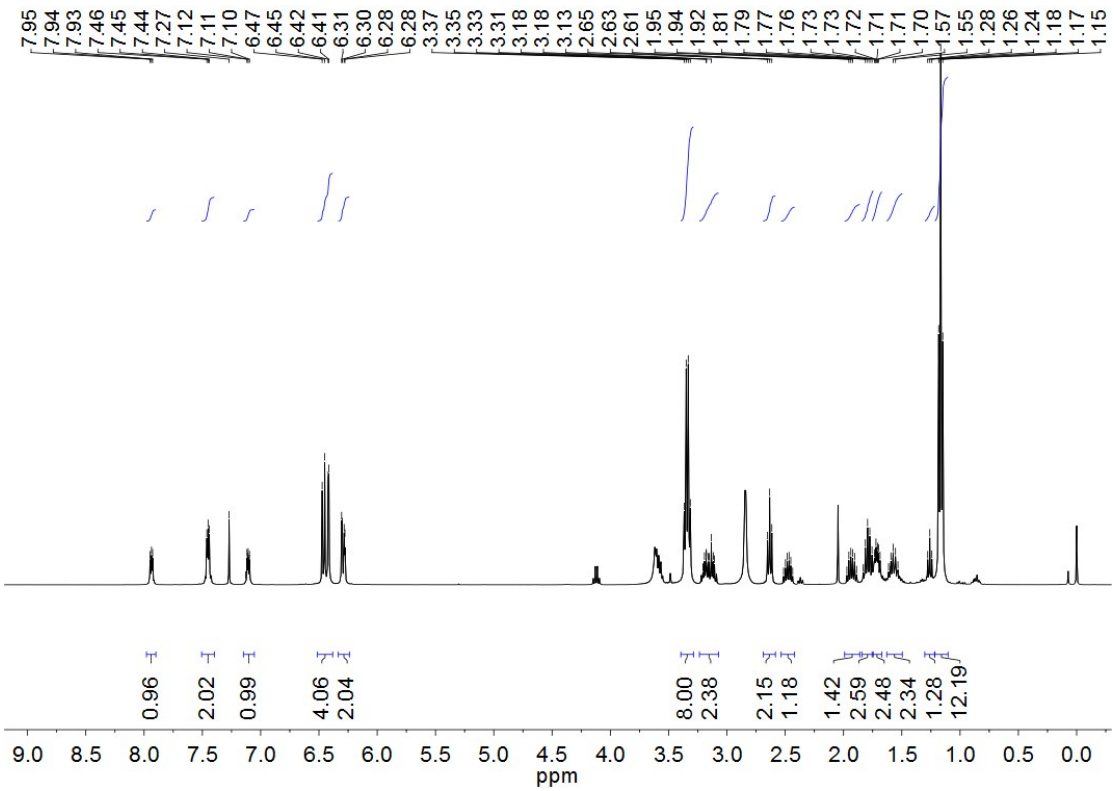


Figure S2. ^{1}H NMR spectrum of Rhod-DPA.

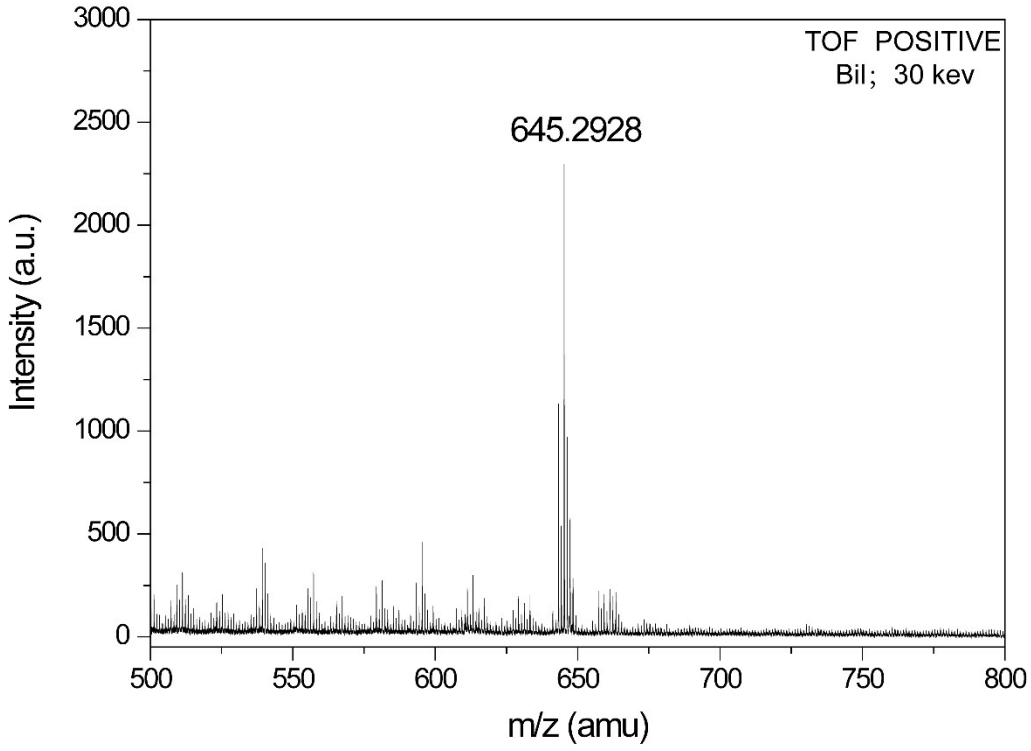


Figure S3. Mass spectrum of Rhod-DPA.

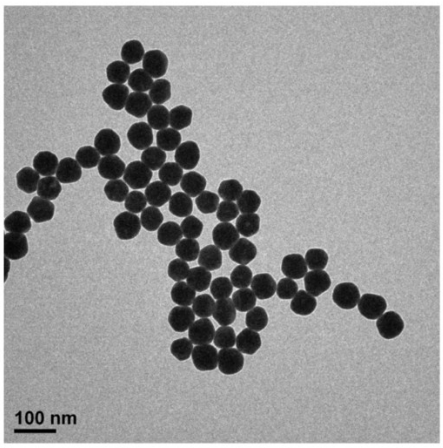


Figure S4. TEM image of Rhod-DPA modified AuNP probes

Results

		Mean (mV)	Area (%)	St Dev (mV)
Zeta Potential (mV):	10.0	**Peak 1:** 10.0	100.0	9.70
Zeta Deviation (mV):	9.70	**Peak 2:** 0.00	0.0	0.00
Conductivity (mS/cm):	0.143	**Peak 3:** 0.00	0.0	0.00

Result quality Good

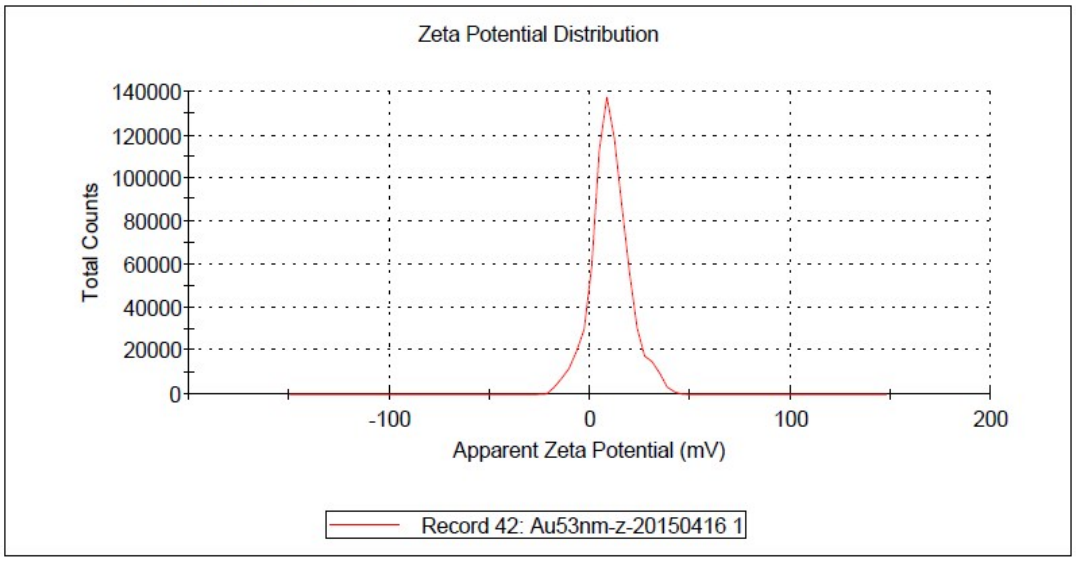


Figure S5. Zeta potential distribution of AuNP probes.

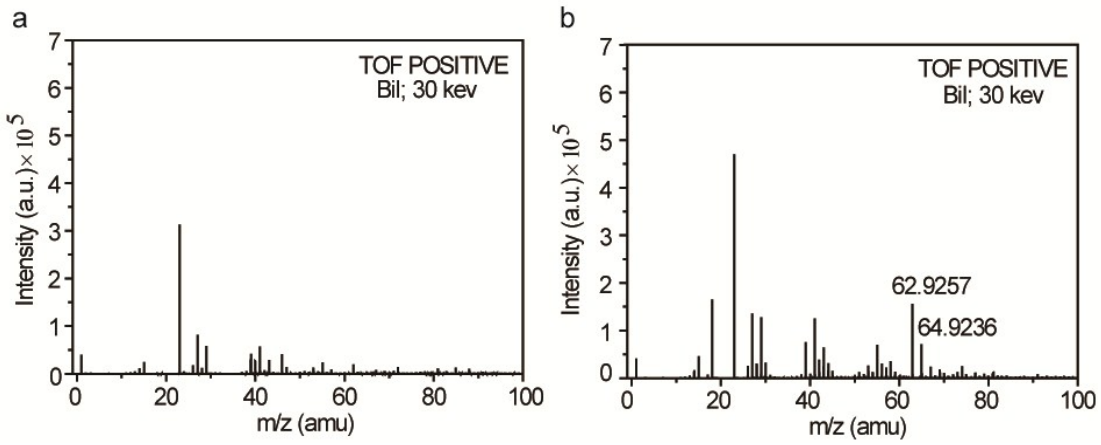


Figure S6. Time-of-flight secondary ion mass spectrometry (ToF-SIMS) of in the absence (a) and presence (b) of 1 mM Cu^{2+} Rhod-DPA modified AuNP probes surface in ITO.

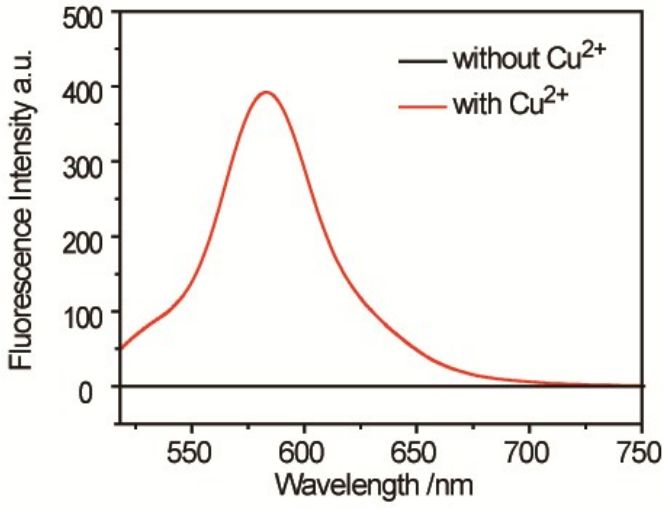


Figure S7. Fluorescence spectra of Rhod-DPA before (black line) and after (red line) the conjugation of Cu^{2+}

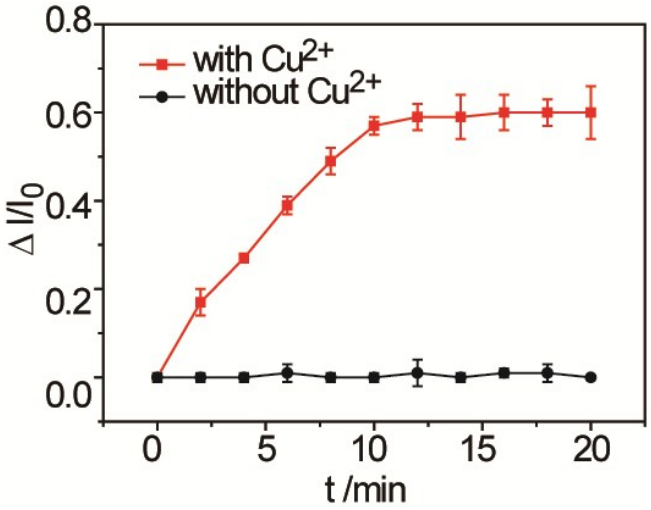


Figure S8. Representative time-dependent scattering spectra intensity changes of single Rhod-DPA-modified AuNPs in the absence (black line) and presence (red line) of 1 mM Cu^{2+}.

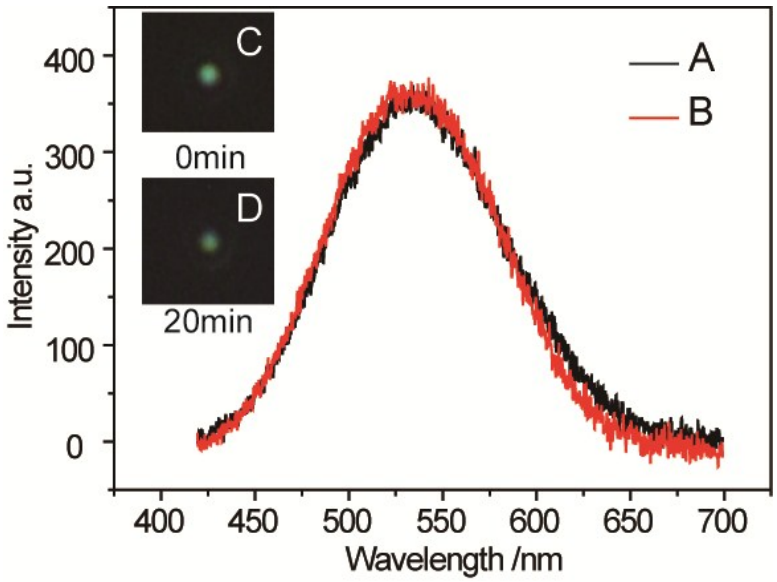


Figure S9. Scattering spectra of single bare AuNPs before (A) and after (B) the addition of 1mM Cu^{2+}. Corresponding dark-field images before (C) and after (D) the addition Cu^{2+}.

References

[1] Z. Gu, Y. L. Ying, C. Cao, P. He and Y. T. Long, Anal. Chem., 2005, 87, 907-913.
[2] Z. Gu, Y. L. Ying, C. Cao, P. He and Y. T. Long, Anal. Chem., 2005, 87, 10653-10656.
[3] N. R. Chereddy and S. Thennarasu, *Dye. Pigment.,* **2011**, 91, 378–382.
[4] K. C. Grabar, R. G. Freeman, M. B. Hommer and M. J. Natan, *Anal. Chem.*, **1995**, 67, 735–743.
[5] S. Link, M. A. El-Sayed, *J. Phys. Chem. B*, **1999**, 103, 4212–4217.

[6] Y. Zhang †, G.S. Wu †, J. Ma, Z.S. Yuan, W. Si, L. Liu, J.J. Sha, Y. F. Chen. *Sci China Tech Sci*, **2015**, 58,519-525.