Electronic Supplementary Information

for

Insight into a reversible energy transfer system

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CONTENT

Experimental and Apparatus

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Reagents and chemicals. Chloroauric acid tetrahyrate ($HAuCl_4 \bullet 4H_2O$) was purchased from Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China). Carbon disulfide, Propargylamine and Tris-(2-carboxyethyl)-phosphine hydrochloride (TCEP) were purchased from Sigma-Aldrich Co. LLC. (USA). Tetramethylrhodamine (TAMRA) azide was purchased from Life Technologies (Carlsbad, USA). DNAs were purchased from Sangon Biotech Co., Ltd. (Shanghai, China). All chemicals were of analytical reagent grade and used as received unless otherwise statements. Ultrapure water (18.2 MΩ) which prepared with a Milli-Q system (Millipore, Bedford, USA) was used throughout the experiments.

Characterization. Scanning electron microscopy (SEM) were recorded with S-4800 Scanning electron microscopy (Hitachi, Japan). Absorption and PL spectra were measured at room temperature with a 3600 UV-Vis-NIR spectrophotometer (Shimadzu, Japan) and a 2500 fluorescence spectrophotometer (Hitachi, Japan), respectively. ^1H and ^{13}C NMR spectra were recorded with AV-300 spectrometer (Bruker, German). Mass spectra were recorded by Bruker esquire HCT (Bruker, German). Fluorescence lifetimes were measured using a fluorolog-3 fluorescence spectrometer (Horiba Jobin Yvon Inc., France). Dark-field light scattering images were obtained through BX51 optical microscope (Olympus, Japan) equipped with dark-field condenser (U-DCW, 1.2-1.4) and DP72 single chip true-color CCD camera (Olympus, Japan), which was controlled by IPE software (MediaCybernetics, USA). The scattering lights from AuNPs were collected by a 100× object lens (adjustable numerical aperture from 0.6 to 1.3) and photographed by a 2070 × 1548 pixel true-color digital CCD camera. The acquired images were all 24-bit TIFF picture files.

Synthesis of *N*-propynyl dithiocarbamate (*N*-PDTC). *N*-PDTC was synthesized according to the report by Stevens' group. Briefly, 0.600 g (10.9 mmol, 1 equiv) propargylamine was dissolved in 30 ml of hexane in a dry 50 ml flask. To this 11 g (109mmol, 10 equiv) Et_3N was added. The flask was placed under N_2 atmosphere and was cooled to 0 ^oC in order to buffer the exothermic reaction. Slowly 1.26 g (16.4 mmol, 1.5 equiv) carbondisulfide was added by syringe. After that, the cooling bath was removed and the reaction was kept stirring at room temperature for 1 h. The white product was formed. **Chromatography:** PE/EA 65/35. **Yield:** 80%.

^{1}H-NMR (300MHz, $CDCl_3$): δ 8.08 (br, 1H); 5.24 (d, J_1 = 3Hz, 1H); 5.14 (d, J_1 = 3Hz, 1H); 4.67 (t, J = 3Hz, 2H). **^{13}C-NMR (75MHz, $CDCl_3$): δ** 198.95; 140.92; 105.64; 56.98. **ESI-MS (**m/z): $[M]^-$ calcd. for $C_4H_5NS_2$, 129.99; found, 129.8**.**

Synthesis of gold nanoparticles. Gold nanoparticles (Au NPs) were synthesized according to a traditional method by reducing $HAuCl_4$ with sodium citrate. Briefly, 0.5 mL of 1 % (w/w) HAuCl4 solution was added into 50 mL H2O and brought to boiling. Then, 0.2 mL of 2 % (w/w) citrate sodium was added under vigorous stirring. After the solution turned to violetred, 1 mL of 2 % (w/w) citrate sodium was added to stabilize the Au NPs. The mixture was kept boiling and refluxed for 5 minutes, and then cool down to room temperature under continuous stirring.

Fabrication of Au NPs-decorated glass slides. The procedures for modifying glass slides was given as follow: 1) glass slides were cleaned with chromic acid for 24 h; 2) the cleaned glass slides were modified with APTMS by incubating with ethanol solution containing 1% (w/w) APTMS for 12 h to present thiols (-SH) on glass slide surface; 3) AuNPs were immobilized on the glass slide surface through Au-S bonds by dipping thiolated glass slide into AuNPs colloid solution for 12 h.

Preparation of N - PDTC - Au NPs conjugates

In aqueous solution. Preparation of the conjugates referred to our previous methods. 1 mL Au NPs was treated with 2×10^{-6} M N-PDTC. 0.25 M NaOH was added in the solution to adjust the pH value to 9. Then, the mixture was kept shaking for 24 h (120 rpm, 25 ^oC). The obtained conjugates were centrifuged once to remove excess N-PDTC, and the precipitates were resuspended for further use. The decoration of N-PDTC on the surface of AuNPs makes the localized surface plasmonic resonance absorption spectra of the 50 nm AuNPs bathochromic-shifted from 526 nm to 532 nm owing to the surface dielectric constant change of AuNPs.

On glass slide surface. The AuNPs-decorated glass slide was firstly immersed in N-PDTC solution to producing N-PDTC–AuNPs conjugates on the whole surface of the glass slide (Fig. S9). Briefly, the previous AuNPs modified glass slide was totally dipped into 100 mL aqueous solution containing 5×10^{-6} M N-PDTC for 12 h and 0.25 M NaOH was added in the solution to adjust the pH value to 9. Then the conjugated glass slide was washed with ethanol and water and dried with N_2.

"Click" reactions between alkynyl Au NPs conjugates and TAMRA

In aqueous solution. 500 μL alkynyl Au NPs conjugates were treated with 10 μL 1×10^{-4} M TAMRA, 10 μL 1×10^{-4} M ascorbic acid and different concentrations of $CuCl_2$ varying from 0 M to 10^{-4} M. The mixture was kept shaking for 12 h (120 rpm, 25 ^{o}C). The obtained products were centrifuged once to remove excess N-PDTC, and the precipitates were resuspended for fluorescence measurement.

On glass slide surface. The conjugates-embellished glass slide was then transferred to a bottle containing the TAMRA solution, only half of the slide was immersed in the solution (Fig. S10). Generally, the N – PDTC - Au NPs conjugates modified glass slide was half dipped into 50 mL aqueous solution containing 5×10^{-6} M TAMRA, 1.0×10^{-4} M ascorbic acid and 1.5×10^{-4} M $CuCl_2$. After 12 h reaction, the glass slide was washed with water and dried with N_2. Then the glass slide was transferred to take dark-field light scattering images. Click reaction was only occurred in the bottom half slide that was submerged in it, while the top half of the slide did not participate the reaction and acted as a comparison. This synthesis strategy was employed to assure the same observation conditions, including light intensity and distance between the slide and the condenser lens, before and after the reaction Control experiment was processed by following the above-mentioned procedure but without TAMRA. The result indicated that simple immersion could not influence the scattering intensity of AuNPs (Fig. S11)

Conjugation of AuNPs with organic dyes by DNA

HS-DNA Sequence: 5' HS GCT CGG AAT TCG TCG

TAMRA-DNA Sequence: 5' TAMRA CGA CGA ATT CCG AGC

FAM-DNA Sequence: 5' FAM CGA CGA ATT CCG AGC

DNA was coupled to the citrate-capped AuNPs (for both 13 nm and 50 nm) through a 5' C_6 alkanethiol functionality with the method provided by Liu and his co-workers.[1] Generally, 100 μM TECP was incubated with 7.5 μM HS-DNA to activate the thiol groups. Then, the HS-DNA was coupled to the citrate-capped AuNPs in 10 mM citrate·HCl buffer of pH 3.0. The mixture was incubated for 12 h at room temperature to form DNA-AuNPs. The solution was centrifuged for 20 min (9,711 *g* for 13 nm AuNPs and 4,316 *g* for 50 nm AuNPs). The supernatant was discarded and the precipitate was resuspended in water. TAMRA-DNA or FAM-DNA was conjugated with the DNA-AuNPs at 37 ^{o}C for 6 h. Then the solution was transferred to fluorescence measurements.

Computational details. The Lorentz and quasi-static approximations were performed on MATLAB and the FDTD simulations were performed on FDTD solutions 8.11.422 by Lumerical Solutions, Inc.

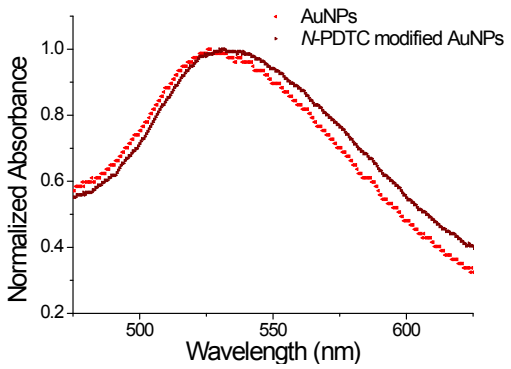


Fig. S1. The absorption spectra of AuNPs before and after modified with N-PDTC. The 4 nm bathochromic shift indicated the successful attachment of N-PDTC on AuNPs.

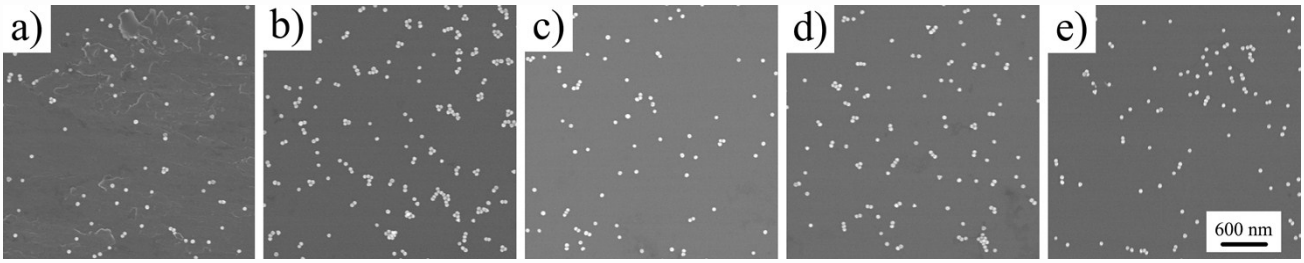


Fig. S2. Morphology of alkynyl-decorated AuNPs before (a) and after (b, c, d, e) CuAAC click reaction at respective Cu^{2+} concentrations of: 0, 10^{-6}, 10^{-5} and 10^{-4} M.

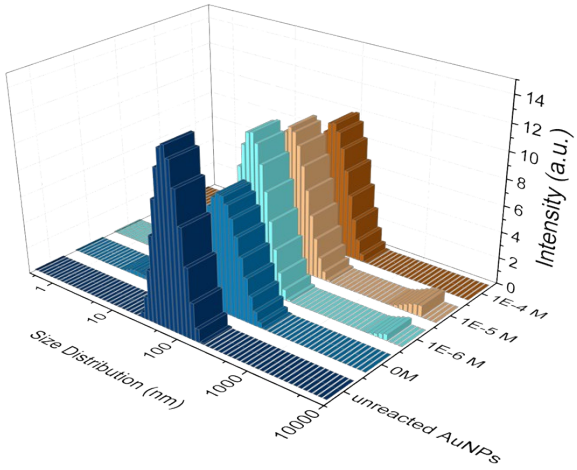


Fig. S3. The DLS data of Au NPs before and after CuAAC click reaction with different concerntration of Cu(II).

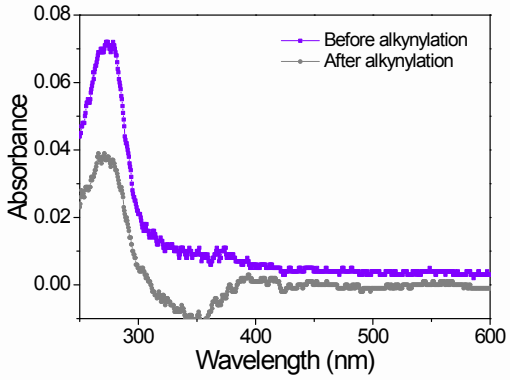


Fig. S4 Absorption of N-PDTC solution before and after the AuNPs alkynylation. By comparing the absorbance at 273 nm, we could calculate the amount of reacted N-PDTC. Concentration: $[\text{N-PDTC}]_{\text{before alkynylation}}$: 4.2×10^{-6} M, [AuNPs]: 4.8×10^{-11} M.

Table S1. Fluorescence lifetime of TAMRA which reacted with Au NPs via different concentration of Cu^{2+}.

Entry	$[Cu^{2+}]$ (M)	τ_1 (ns)	α_1	τ_2 (ns)	α_2	τ_3 (ns)	α_3	$<\tau>$ (ns) [a, b]
1	10^{-6}	1.284	0.0747	0.1568	0.7723	2.955	0.1530	0.6691
2	10^{-5}	1.004	0.1008	0.1267	0.7738	2.562	0.1254	0.5205
3	10^{-4}	0.1124	0.8136	0.1741	0.7879	2.565	0.0122	0.1879
TAMRA-N_3	-	0.7317	0.1279	2.227	0.8510	6.941	0.0211	2.135

[a] PL average lifetime was calculated according to the formula of $<\tau> = (\sum \alpha_i \tau_i) / \sum \alpha_i$.

[b] To avoid the interfere from the scattering light of AuNPs, the lifetime was collected under the pulsed laser excitation at 454 nm.

Table S2. The parameters of the marked AuNPs.

	d_1 (nm)	d_2 (nm)	S (nm^2)
Particle 1	**64**		**3217**
Particle 2	**52**	**70**	**4457**
Particle 3	**42**	**70**	**3435**

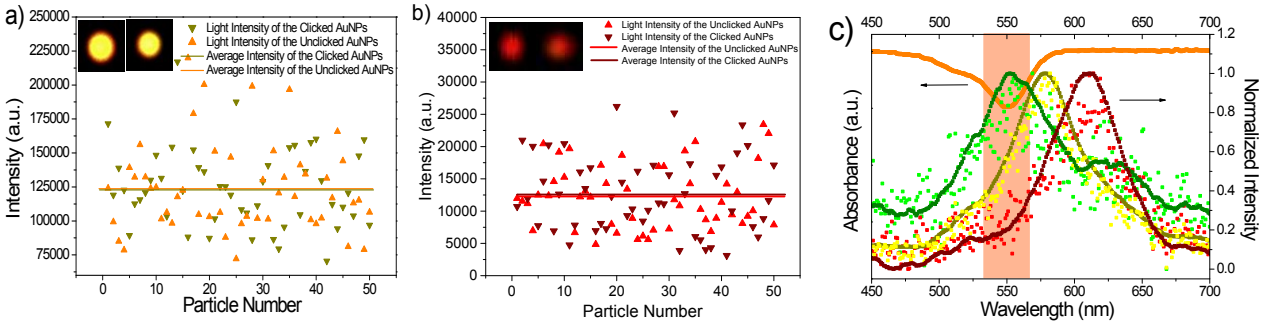


Fig. S5. Light scattering intensities and the average light scattering level from yellow (a) and red (b) particles. The insets show visual comparison of a randomly selected unclicked AuNP (left) and clicked AuNP (right) via *i*DFM at the same magnification. The significant differences were 0.95 and 0.83, respectively. This data implied that the difference of the yellow and red scattering phenomena between the clicked and unclicked was not significant. This was because that the PRET process required a resonant interaction between the D-A pair and the cross-section of

the TAMRA-N_3 absorption spectrum with the AuNPs scattering spectrum directly. c) Spectral overlap between the absorbance of TAMRA-N_3 and single nanoparticle light scattering spectra (green, yellow and red). This spectra indicated the requirement of the cross-section between the D-A pairs directly influenced energy transfer efficiencies.

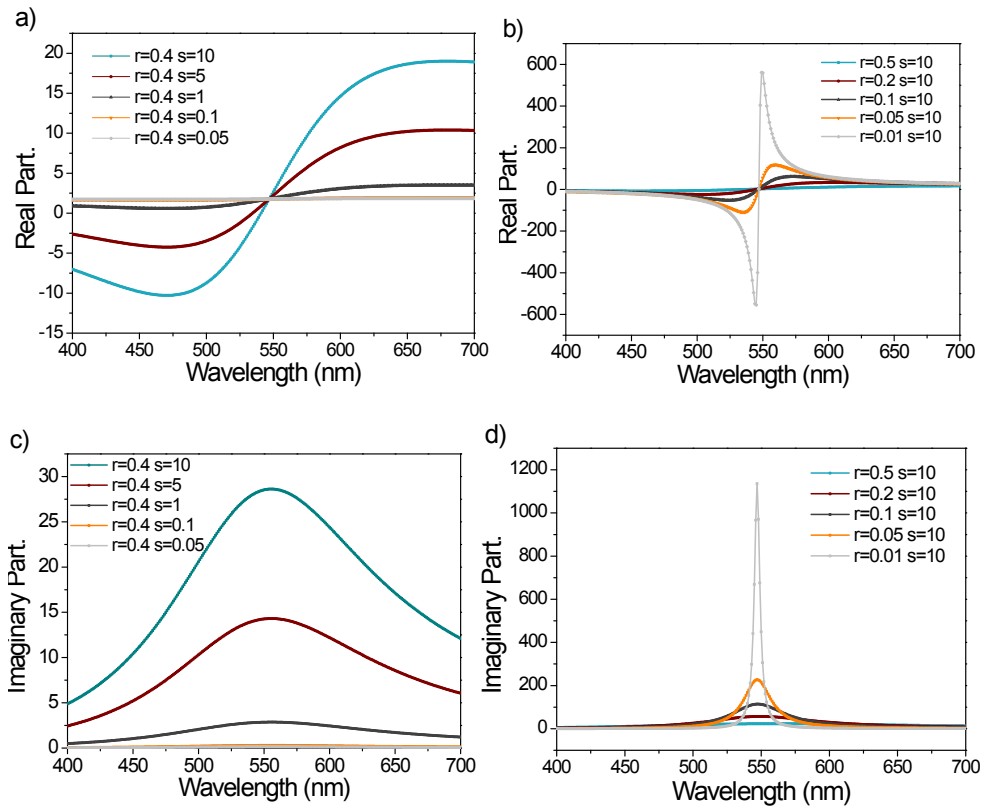


Fig. S6 The value of *s* and *γ* effected the width and the magnitude of the complex dielectric function. By increasing *s* could have a stronger intensity of dielectric function for both real part (a) and the imaginary part (c). By increasing *γ* could have a wider distribution of dielectric function for both real part (c) and the imaginary part (d).

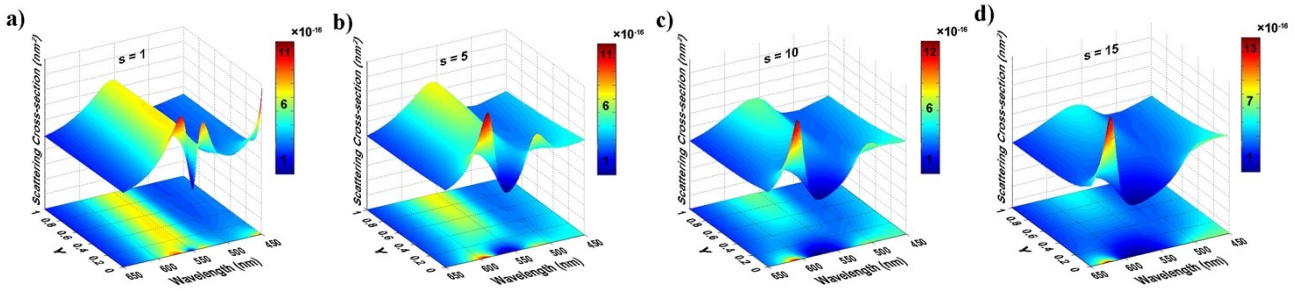


Fig. S7 the calculation of scattering cross-section diagrams with vary of *γ* under different *s*. The x-axis referred to the wavelength and the y-axis referred to the *γ*. *S* in each diagram was set at 1, 5, 10, 15. The simulations indicated the smaller *γ* could arouse a deeper quenching dip.

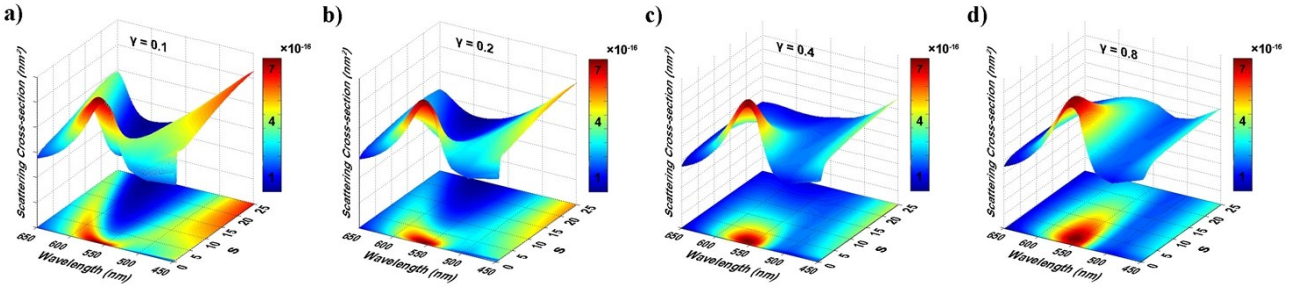


Fig. S8 the calculation of scattering cross-section diagrams with vary of *s* under different *γ*. The x-axis referred to the wavelength and the y-axis referred to the *s*. *γ* in each diagram was set at 0.1, 0.2, 0.4, 0.8. The simulations indicated the bigger *s* has a stronger quenching magnitude.

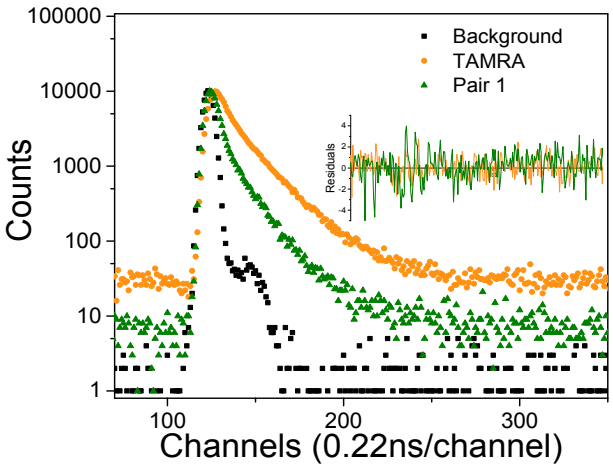


Fig. S9 Fluorescence lifetime of TAMRA-DNA and TAMRA-DNA-AuNPs (pair 1). The inset was the residuals of the fitting data.

Table S3. Fluorescence lifetime of TAMRA-DNA and TAMRA-DNA-AuNPs (pair 1).

Entry	τ_1 (ns)	α_1	τ_2 (ns)	α_2	τ_3 (ns)	α_3	$<\tau>$ (ns) [a]
TAMRA-DNA	3.208	0.4781	1.137	0.3430	5.389	0.1790	2.888
TAMRA-DNA-AuNPs	0.8521	0.0633	0.1382	0.7300	2.814	0.2067	0.7363

Fig. S10. Photographs of the reaction strategy and overall comparison between the unclicked and clicked AuNPs.

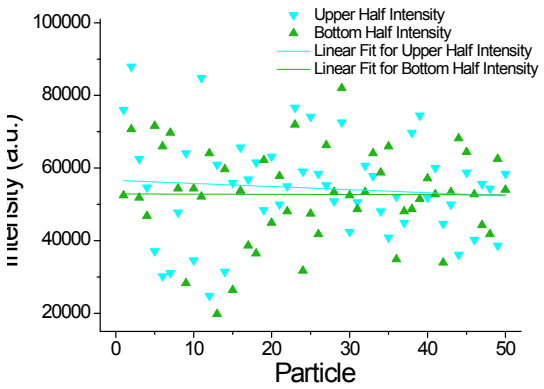
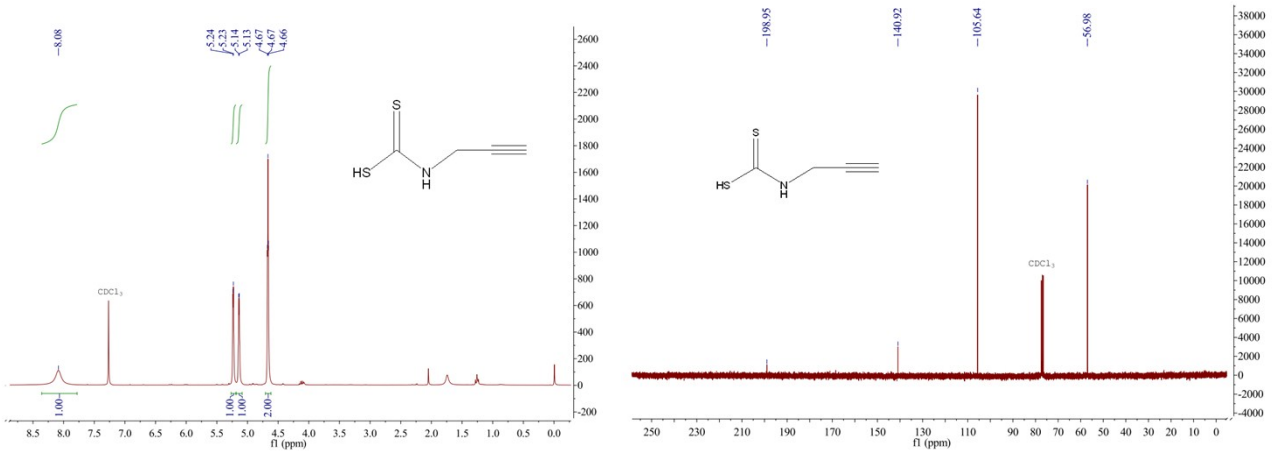
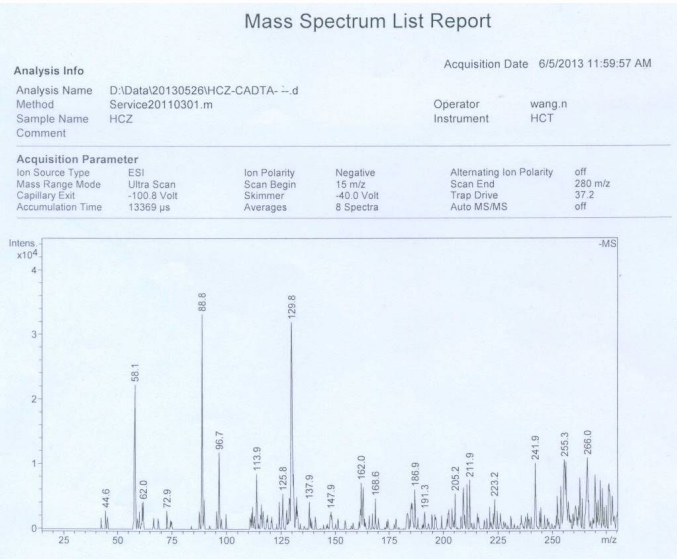


Fig. S11. Scattering light intensity and linear fit for the upper and bottom half of unclicked Au NPs prepared in the control experiment. The probability was calculated to be 0.5299, which was far bigger than the set significance level (α) 0.05, indicated no significant difference between the upper and bottom unclicked Au NPs.

^{1}H-NMR, ^{13}C-NMR and mass spectrum reports of N-PDTC





1. X. Zhang, M. R. Servos and J. Liu, *J. Am. Chem. Soc.*, 2012, **134**, 7266-7269.