

Supporting Information

Visual detection of ascorbic acid via alkyne-azide click reaction using gold nanoparticles as colorimetric probe

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Synthesis of 11-(acetylthio)undecanoic acid (1) ¹

Thioacetic acid (3.0 g) and 11-bromooctanoic acid (5 g) were added into a mixture of Sodium (0.89 mg) and dry methanol (165 mL). The reaction mixture was stirred for 20 h at 70 °C in a nitrogen atmosphere, and then poured into a beaker containing ice (15 g). 6 M HCl was added for adjusting to pH 3.0. The organic layer was extracted with dichloromethane (3 × 80 mL), dried over Na₂SO₄ and concentrated to yield 11-(acetylthio) undecanoic acid(4.4 g , 95%).

Synthesis of S-11-(2-(2-(2-aminoethoxy) ethoxy) ethylamino)-11-oxoundecylethanethioate (2) ^{2,3}

To a solution of NHS (1.56 g) and DMAP (0.161 g) in 50 mL of anhydrous DCM, crude (1) (4.4 g) were added, and then DCC (2.76 g) was added to the solution. The reaction was cooled at 5 °C for the first hour and react at room temperature overnight. The solution was diluted with DCM, filtered and evaporated to the active ester.

The ester (7 g) was dissolved in 50 mL of DCM. 2, 2'-(ethylenedioxy)bis(ethylamine) (32 g) was slowly dropped into the solution within 90 min under vigorous magnetic stirring.. The reaction went on 12 h at room temperature. The solution was filtered, washed with water (3 × 200 mL), dried over anhydrous MgSO₄ and concentrated to yield

S-1-(2-(2-(2-

aminoethoxy)ethoxy)ethylamino)-11-oxoundecylethanethioate (6.56 g, 93.3%).

Synthesis of S-11-(2-(2-(2-(3-azidopropanamido)ethoxy)ethoxy)ethylamino)-11-oxoundecyl ethanethioate (3)

3-bromopropanoic acid (2.7 g) and sodium azide (3.48 g) were dissolved in 10 mL of DMF and stirred at 60 °C for 48 h. The reaction mixture was then dissolved in DCM, washed with water and NaHCO₃ (aq), dried over anhydrous MgSO₄ and concentrated by rotary evaporation to obtain 3-azidopropanoic acid as a light yellow liquid (2.1 g, 78%).

3-azidopropanoic acid (0.92 g, 8 mmol) was dropped into the 50 mL solution of DCM for a period of 20 min with stirred, which was composed of (2) (3.2 g) and EDC (1.6 g). The reaction would go on 12 h at room temperature. The solution was washed with water (2 × 50 mL), dried over anhydrous MgSO₄ and concentrated to yield crude S-11-(2-(2-(2-(3-azidopropanamido)ethoxy)ethoxy)ethylamino)-11-oxoundecyl ethanethioate (3.1 g, 77%).

Synthesis of 11-mercapto-N-(2-(2-(2-propiolamidoethoxy)ethoxy)ethyl) undecanamid (4)

To a solution of dry methanol, 2 g of 3 and 1 mL of 0.5 M sodium methoxide was added. The reaction mixture was stirred for 1 hour at

room temperature. In order to prevent hydrolysis, the solution was newly-prepared. The solution was washed with 0.2 M NaOH (3 × 100 mL) and brine (3 × 100 mL), dried over anhydrous MgSO₄ and evaporated to dryness. The crude product was purified by silica gel column chromatography using EtOAc/petroleum ether/MeOH (1:2:0.5) to obtain purified product 11-mercapto-N-(2-(2-(2-propiolamidoethoxy)ethoxy) ethyl) undecanamide (4) (1.35 g, 73.8%). ¹H-NMR (CDCl₃, 300 MHz): δ 1.1-1.5 (br m, 14H), δ 1.5-1.6 (t, 2H), δ 1.6-1.8 (t, 2H), δ 1.8-1.9 (t, 2H), δ 2.0-2.2 (m, 2H), δ 2.6-2.72 (t, 2H), δ 3.2 (s, 1H).

Synthesis of S-11-oxo-11-(2-(2-(2-propiolamidoethoxy) ethoxy) ethylamino)undecyl ethanethioate(5)⁴

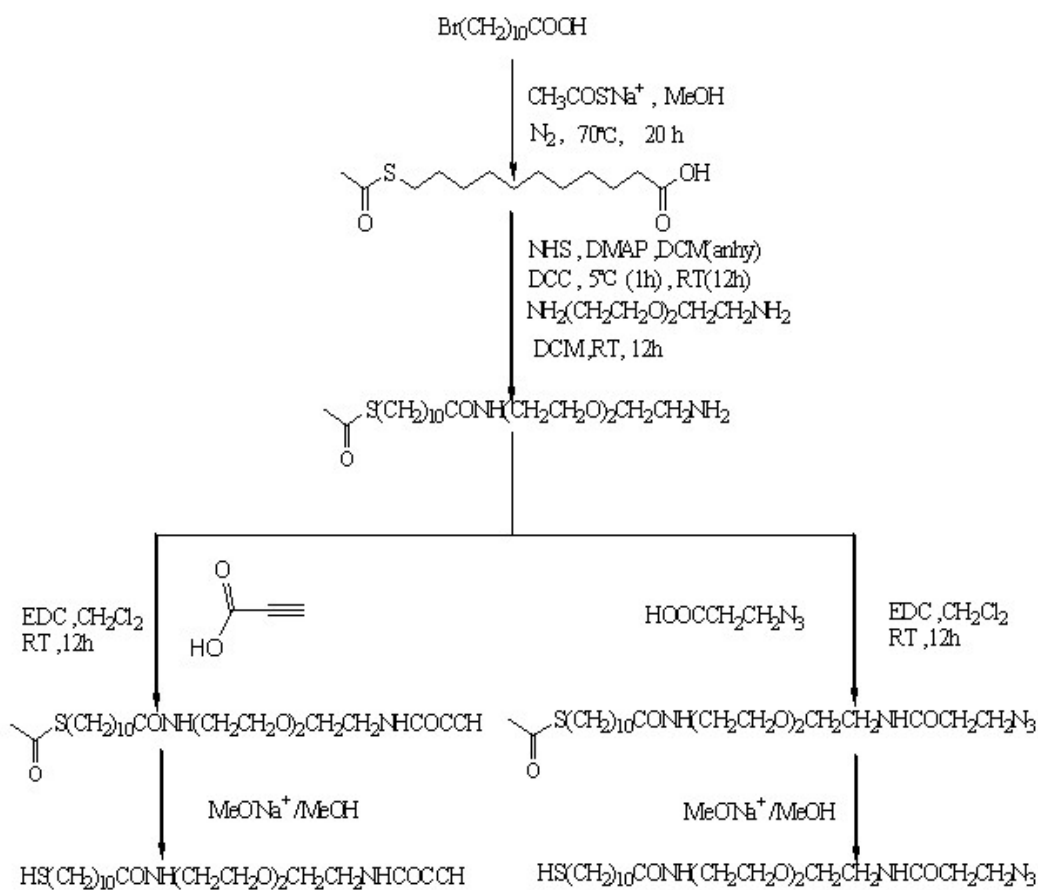
3.2 g of 2 and EDC (1.6 g, 8 mmol) were mixed in 50 mL of anhydrous DCM. The propargyl acid (0.57 g, 8 mmol) was dropped into the solution over a period of 20 minutes with stirred. The reaction was left at room temperature for 12 h. The solution was washed with water (2 × 50 mL), dried over anhydrous MgSO₄ and concentrated to yield crude S-11-oxo-11-(2-(2-(2-propiolamidoethoxy) ethoxy) ethylamino) undecyl ethanethioate (3.04 g, 76%).

Synthesis of N-(2-(2-(2-(3-azidopropanamido) ethoxy) ethoxy)

ethyl-11-mercaptoundecanamide (6)

This procedure is similar to (4). To a solution of dry methanol, 144 mg (0.4 mmol) of (5) and 1 mL of 0.5 M sodium methoxide was added. The reaction mixture was stirred for 1 hour at room temperature. The solution need newly-prepared for prevent hydrolyzed. The crude product was purified by silica gel column chromatography using DCM/EtOAc/MeOH (4:1:1) to obtain purified product (1.4 g, 77%). ¹H-NMR (CDCl₃, 400 MHz): δ1.15-1.48 (br m, 14H), δ1.6-1.75 (t, 2H), δ1.5-1.6 (m, 2H), δ1.6-1.7 (t, 2H), δ1.8-1.9 (t, 2H), δ2.1-2.23 (t, 2H), δ2.25-2.4 (t, 2H), δ2.58-2.72 (t, 2H), δ3.1-3.28 (t, 2H), δ3.3-3.5 (m, 4H), δ3.55-3.68 (m, 4H).

All reactions were carried out in aluminum foil wrapped flasks to exclude light during reaction.



Scheme 1 Synthesis of $\text{HS}(\text{CH}_2)_{10}\text{CONH}(\text{CH}_2\text{CH}_2\text{O})_2\text{CH}_2\text{CH}_2\text{NHCOCCH}$
 and $\text{HS}(\text{CH}_2)_{10}\text{CONH}(\text{CH}_2\text{CH}_2\text{O})_2\text{CH}_2\text{CH}_2\text{NHCOCCH}_2\text{CH}_2\text{N}_3$

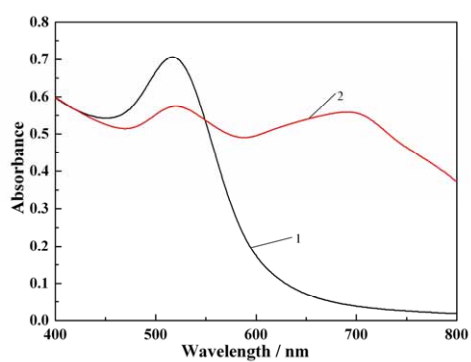


Figure S1. Absorption spectra of AuNPs (1) in the absence of cysteine, (2) in the presence of 2.5×10^{-6} M cysteine and 1 mM Cu^{2+} .

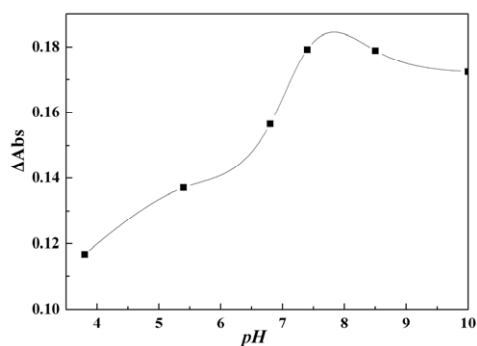


Figure S2. Effect of pH on the response. Experimental conditions: Alkyne-functionalized AuNPs (10 nM, 150 μ L); azide-functionalized AuNPs (10 nM, 100 μ L); Cu^{2+} (4.0×10^{-5} M, 150 μ L); ascorbic acid: 20 nM; incubation time: 20 min.

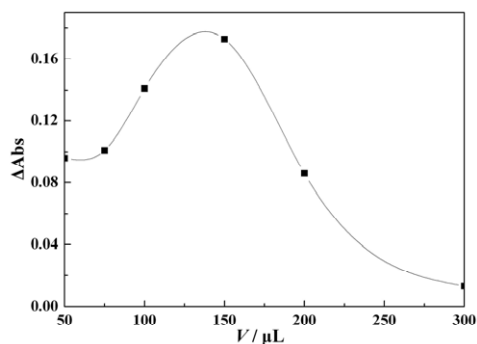


Figure S3. Effect of Cu^{2+} amount on the response. Experimental conditions: Alkyne-functionalized AuNPs (10 nM, 150 μL); azide-functionalized AuNPs (10 nM, 100 μL); ascorbic acid: 20 nM; Cu^{2+} : 4.0×10^{-5} M; incubation time: 20 min.

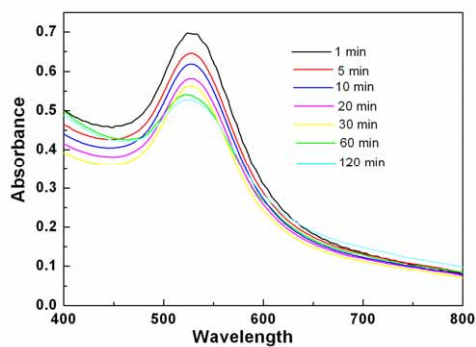


Figure S4. Effect of incubation time on the response. Experimental conditions: Alkyne-functionalized AuNPs (10 nM, 150 μ L); azide-functionalized AuNPs (10 nM, 100 μ L); ascorbic acid: 20 nM; Cu^{2+} (4.0×10^{-5} M, 150 μ L).

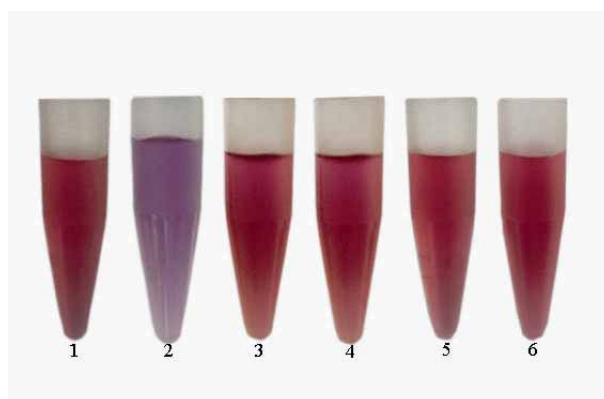


Figure S5. A photograph of the solution containing the mixtures of functionalized AuNPs with different reducing agents (1) blank, (2) ascorbic acid, (3) cysteine, (4) dopamine, (5) thiamine, and (6) uric acid. Experimental conditions: alkyne-functionalized AuNPs (10 nM, 150 μ L); azide-functionalized AuNPs (10 nM, 100 μ L); Cu^{2+} (40 μ M, 150 μ L); incubation time, 20min.

Table S1. Determination of ascorbic acid content in beverage samples

Sample	Added (mg/100 mL)	Proposed method (mg/100 mL) ^a	Official method (mg/100 mL)	Recovery (%)
Orange juice	0	22.4(±1.74)	22.2	–
	20	42.6(±2.41)	42.1	101.8
	40	61.2(±1.89)	62.3	95.1
	60	82.4(±1.13)	82.5	99.6
Grapefruit juice	0	18.6(±0.71)	18.9	–
	20	38.6(±2.31)	39.0	97.9
	40	59.4(±2.83)	59.0	102.1
	60	79.2(±1.35)	79.0	101.0

^a Average of five replicates ± SD.

References

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