Supporting information for

Algorithm-assisted automated identification and enumeration system for sensitive hydrogen sulfide sensing under dark field microscopy

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Table of content

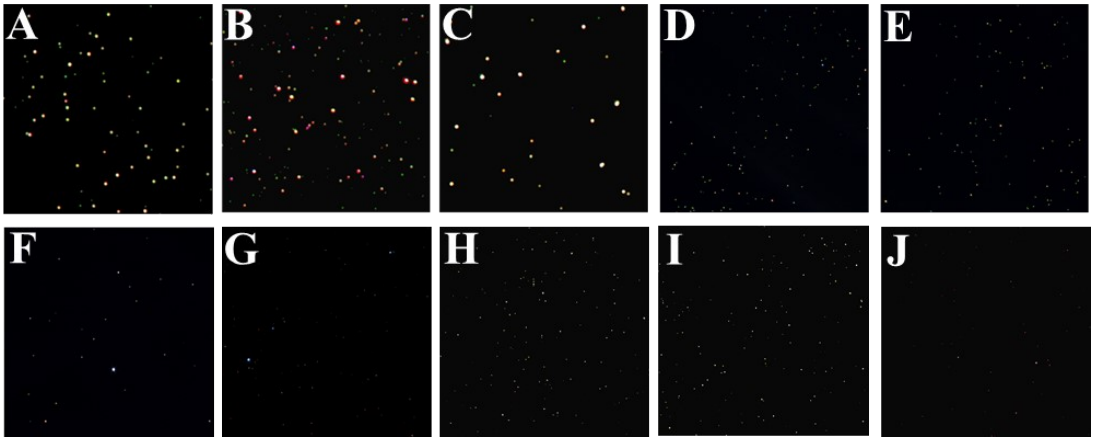

Figure S1 DFM images of AuNPs.

Table S1 The counting results of total particle number, green particle number and the ratio of green particle by two methods.

Method	Color threshold[1]			Meanshift[2]		
	Total	Green	Ratio (%)	Total	Green	Ratio (%)
A	76	42	55.26	76	19	25.00
B	155	93	60.00	151	75	49.67
C	31	6	19.35	31	2	6.452
D	119	118	99.16	130	51	39.23
E	97	99	102.1	122	61	50.00
F	31	21	67.74	44	10	22.73
G	141	44	32.21	181	52	28.73
H	167	152	91.02	168	94	55.95
I	174	165	94.83	173	62	35.84
J	66	5	7.576	65	10	15.38

1. Parameters for color threshold. For total scattering spot: Hue range was set 0 ~ 255, brightness range was set 10 ~ 255; and hue range was set 60 ~ 110, brightness range was set 10 ~ 150 for green spot. In counting part, the size (pixel^2) was set 10. Other parameters were both default.

2. Parameters for mean shift algorithm. The color size was set as 7 and bandwidth was set as 10 for both images analyzing.

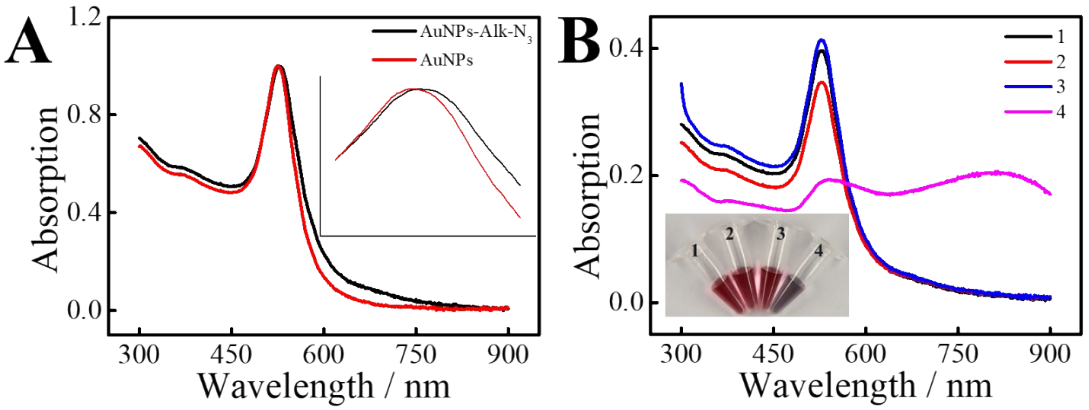

Figure S2 (A) Uv-Vis spectrum of bare AuNPs (red) and AuNPs-Alk-N_3 (black), inserted spectrum was Uv-Vis spectrum between 500 to 560 nm; (B) Uv-Vis spectrum of AuNPs-Alk-N_3 after mixed with (1) H_2O, (2) 1 mM Cu^{2+}, (3) 2 mM SA, and (4) 1 mM Cu^{2+} and 2 mM SA; inserted picture was their corresponding digital photograph.

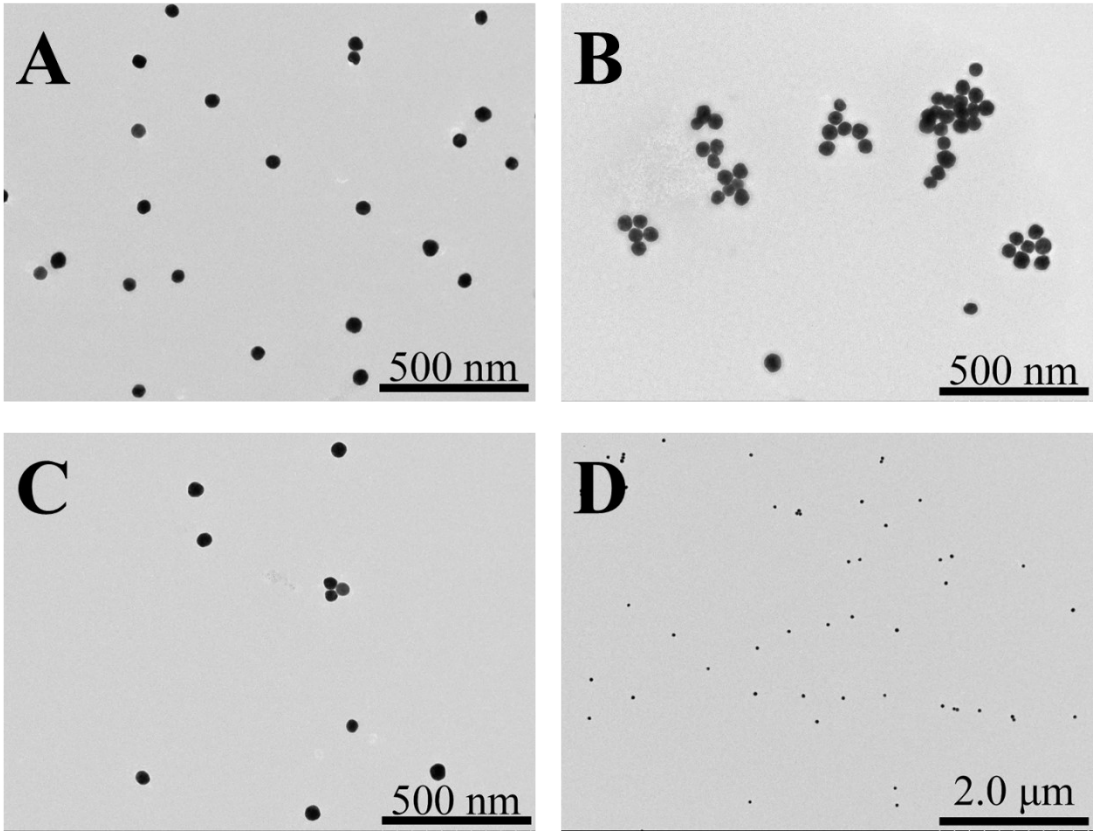

Figure S3 TEM images of (A) AuNPs-Alk-N_3; (B) AuNPs-Alk-N_3 with no H_2S treated after mixed with Cu^{2+} and SA; (C) 80 μM H_2S treated AuNPs-Alk-N_3 after after mixed with Cu^{2+} and SA and (D) the corresponding large scare images.

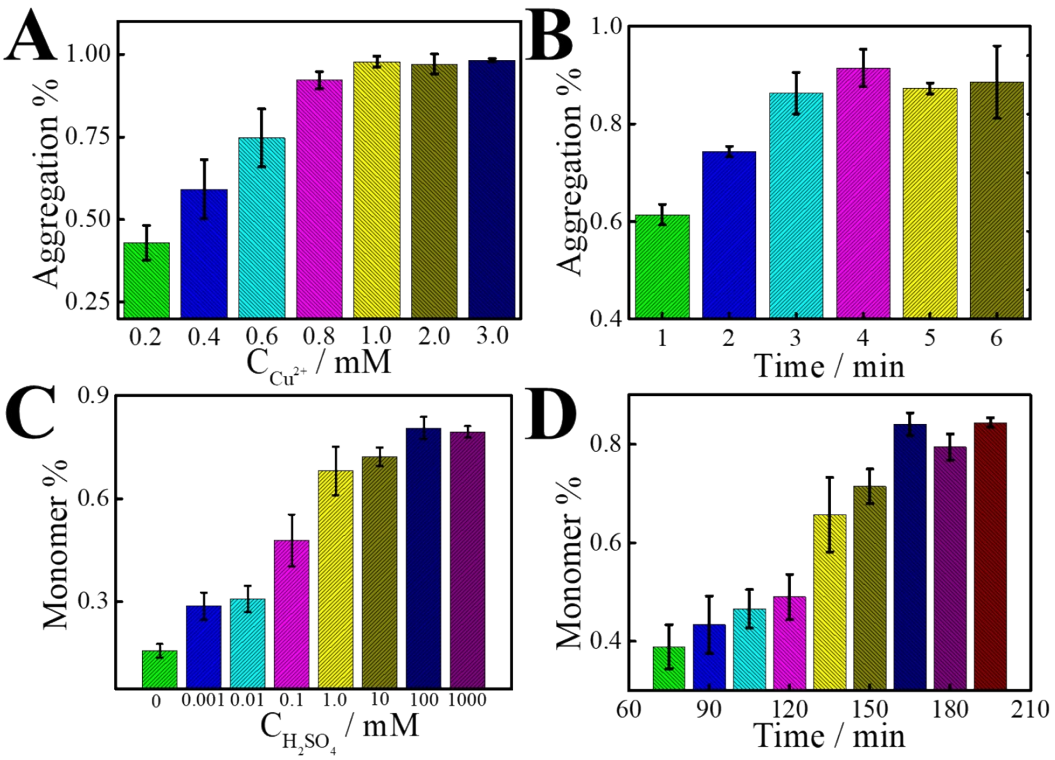

Figure S4 Optimization of experimental conditions. Effect of (A) the concentration of Cu^{2+} and (B) click chemical reaction time on the aggregation of gold nanoparticles; effect of (C) H_2SO_4 concentration and (D) incubate time between H_2S and AuNPs-N_3-Alk on the monomer ratio. The concentration S^{2-} is 80.0 μM .

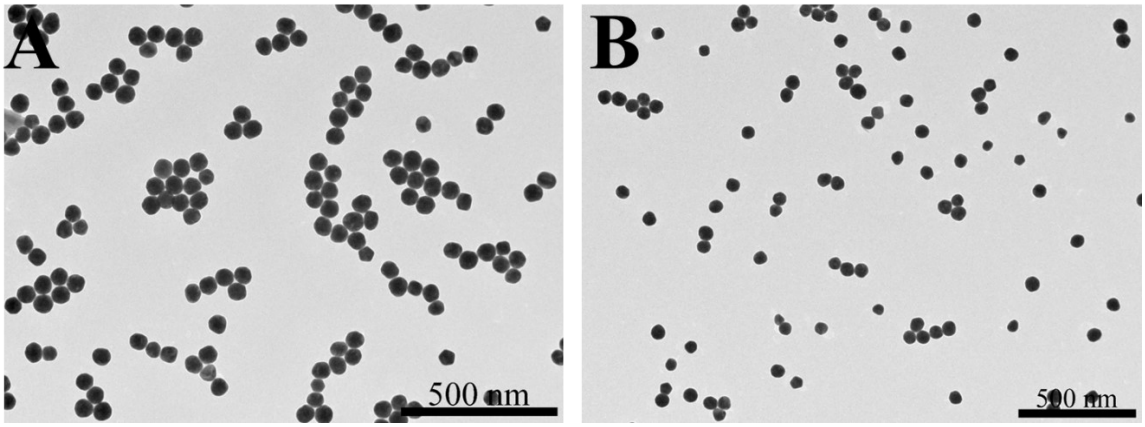

Figure S5 TEM images of AuNPs after treated with (A) 5.0 μM and (B) 50 μM H_2S.

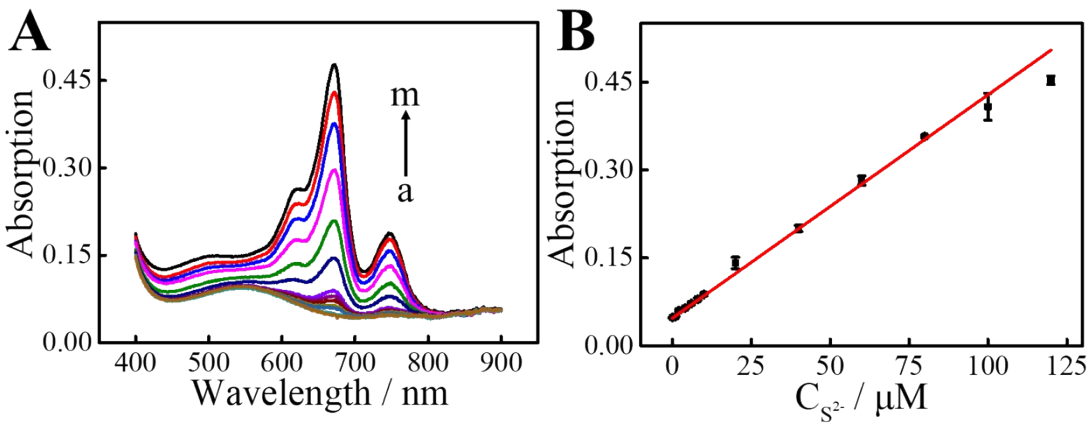

Figure S6 Result of H_2S detection by methylene blue colorimetric method. (A) The UV-visible absorption spectrum of solution, and (B) corresponding linear relationship between H_2S concentration and the absorption value at 665 nm. The concentration of H_2S concentration from a to m were 0, 1, 2, 4, 6, 8, 10, 20, 40, 80, 100, and 120 μM.

Table S2 Comparison of analytic performance of the different methods for H_2S detection.

Detection Method	Linear Range	Detection Limit	Ref.
Chemiluminescent	20 – 100 μM	4.6 ± 2.0 μM	S1
Chemiluminescent	0.78 – 40 μM	0.30 μM	S2
Electrochemiluminescence	0.05 – 100.0 μM	0.02 μM	S3
Fluorescence	0 – 100 μM	0.86 μM	S4
Fluorescence	4.1 – 110 μM	4.1 μM	S5
Fluorescence	0.10 – 80 μM	0.035 μM	S6
Fluorescence	0.5 – 5 μM	0.2 μM	S7
Colorimetric	0.05 – 50 μM	0.019 μM	S8
Colorimetric	0.01 − 5 μM	0.01 μM	S9
Colorimetric	1 − 6 μM	0.78 μM	S10
DFM	2 – 80 μM	2 μM	This work

References

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