SUPPLEMENTARY INFORMATION

Novel theranostic DNA nano-scaffolds for the simultaneous detection and killing of *Escherichia coli* and *Staphylococcus aureus*

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Supplementary Experiment Details

Glutathione protected gold nanoclusters (GSH-Au NCs) synthesis and characterization

Glutathione protected gold nanoclusters (GSH-Au NCs) were prepared as described previously[1] with a slight modification. Briefly, aqueous stock solution of $NaBH_4$ (~112 mM) was prepared by introducing 43 mg $NaBH_4$ powder into NaOH (0.2 M, 10 mL), and stock solutions of 20 mM $HAuCl_4$ and 50 mM GSH were prepared with ultrapure water. In a typical synthesis of luminescent GSH-Au NCs, 150 μL of GSH (50 mM) and 125 μL of $HAuCl_4$ (20 mM) were mixed with 4.7 mL of ultrapure water under vigorous stirring, followed by slowly introducing 57 μL of $NaBH_4$ (112 mM) into the reaction mixture. The sample was collected after 10 minutes of stirring, and then subjected to incubation under sub-zero temperature (-16 °C in the freezer compartment of the refrigerator) for 5 days. Highly red-emissive GSH-Au NCs could be obtained.

The red-emissive GSH-Au NCs were then purified with a PD-10 desalting column by GE Healthcare with an exclusion limit of M_r 5000. The red-emissive GSH-Au NCs were further characterized. UV-Vis spectra and photo-emission spectra were acquired on a Shimadzu UV-1800 spectrometer and a PerkinElmer LS55 fluorescence spectrometer, respectively. Transmission electron microscopy (TEM) image was obtained on a JEOL JEM 2010. The GSH-Au NCs were also analyzed with X-ray photon spectroscopy (XPS, AXIS HIS, Kratos Analytical) with a mono Al Kα radiation source (hν = 1486.71 eV) operating at 15 kV and 5 mA.

DPAu/AMD characterization

The construction of the DP was verified with 12.5 % or 6% native polyacrylamide gel electrophoresis (PAGE). 3 μM of DP was run in TBE buffer at 150 V in 4^{o}C approximately

for 1.5 h. After electrophoresis, the gel was stained with ethidium bromide for 30 min before washing off the extra stain using ultrapure water for another 10 minutes. The gel image was then visualized using UV transilluminator Imaging System (Syngene, UK)

Meta analysis of DNase I activity in bacterial cells

E.coli K12 is reported to have DNase I with the capability to degrade 297 nmole/mg protein within 30 minutes at $37^{o}C$.[2]

The authors reported the protein content to be 0.05 g/L and the volume reaction for DNase I assay to be 0.3 mL.[2]

Thus, the protein amount could be determined to be 0.015 mg

And the DNase I activity could be determined to be 4.46 nmole of DNA being degraded in 30 min time.

Similarly, the *S. aureus* were reported to produce DNase I with activity of 41 Kunitz unit. Basing on the unit definition and taking calf thymus molecular weight as 10^6 Da, The amount of DNA which could be degraded by *S. aureus* to be 4.2 nmole within 30 min at $37^{o}C$.[3]

In comparison, the commercial DNase I enzyme of bovine pancreatic that we used in this study defines 1 U to be equal to the degradation of 1 μg of pUC19 in 10 minutes at $37^{o}C$.[4]

pUC19 is known to be in the size of 2686 bp with MW of 1.74×10^6 g/mol.[5] Thus, the rate of degradation of 1 U of DNase I is 5.74×10^{-4}nmole of DNA in 10 minutes.

Assuming the enzymatic reaction is linear in rate, therefore 10 U of DNase I enzyme would have the capability to degrade 0.017 nmole DNA in 30 minutes time.

SUPPLEMENTARY DATA

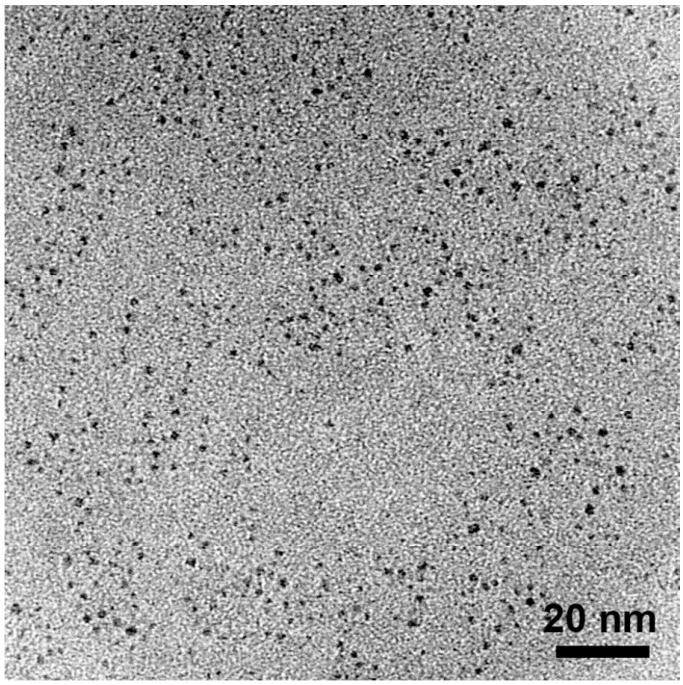

Figure S1. A representative TEM image of the luminescent GSH-Au NCs. Scale bar: 20 nm

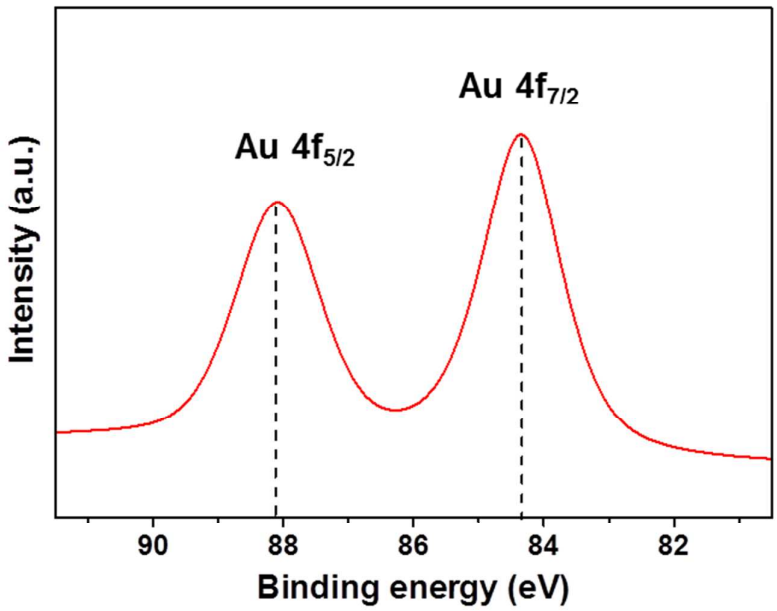

Figure S2. XPS spectrums of the luminescent GSH-Au NCs. Doublet peak signals confirm the formation of GSH-Au NCs.

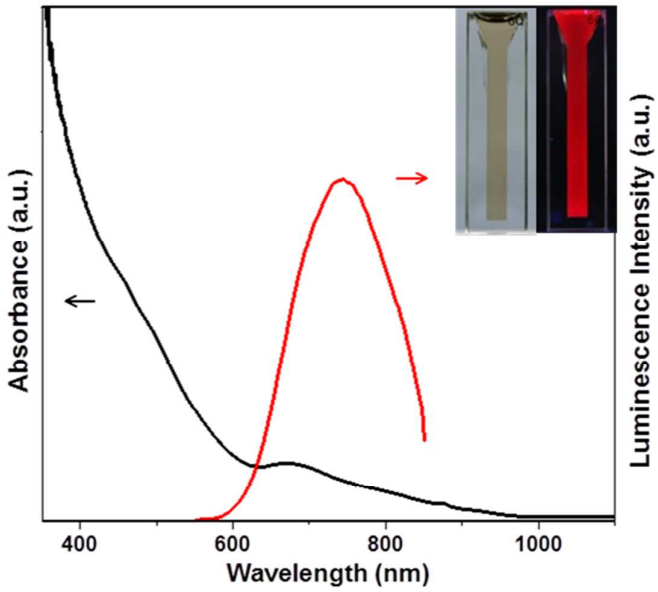

Figure S3. Optical characterization of red-emissive GSH–Au NCs. Optical absorption (black line) and photoemission (red line) spectra of the luminescent GSH–Au NCs. The inset shows photographs of the GSH–Au NCs under visible (left) and UV (right) light illumination.

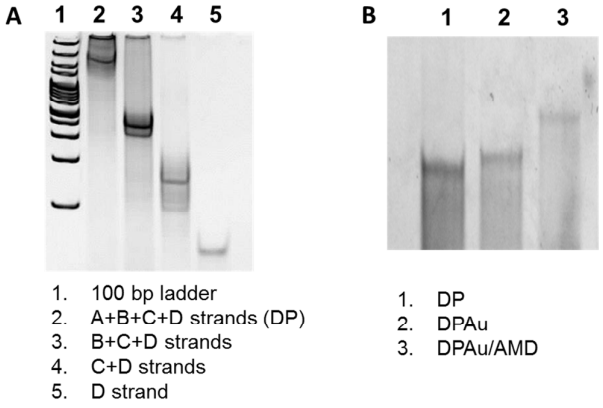

Figure S4. Native PAGE analysis shows the formation of DP, DPAu and DPAu/AMD nanostructures. (A) Separation of oligunoculetiode and DP structure on 12.5% PAGE, indicating the formation of DP. (B) Native PAGE analysis (6% PAGE) shows that DPAu and DPAu/AMD move at slower migration rate when compared to DP.

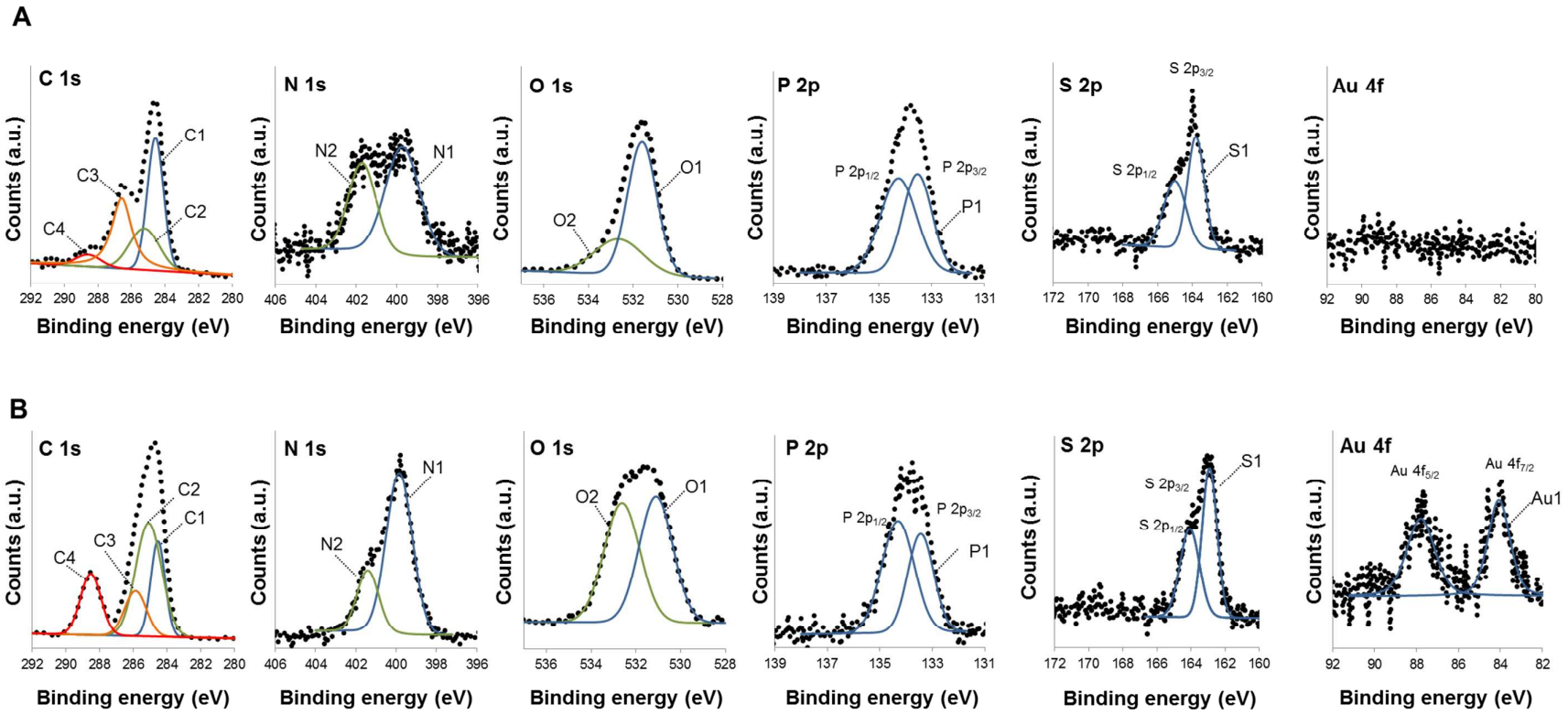

Figure S5. Deconvolution spectra of XPS spectra (A) DP and (B) DPAu/AMD. Both samples were assessed for element: C1s, N1s, O1s, P2p, S2p and Au4f. The peak association and each atomic composition ratio for each element were summarized in Table 1.

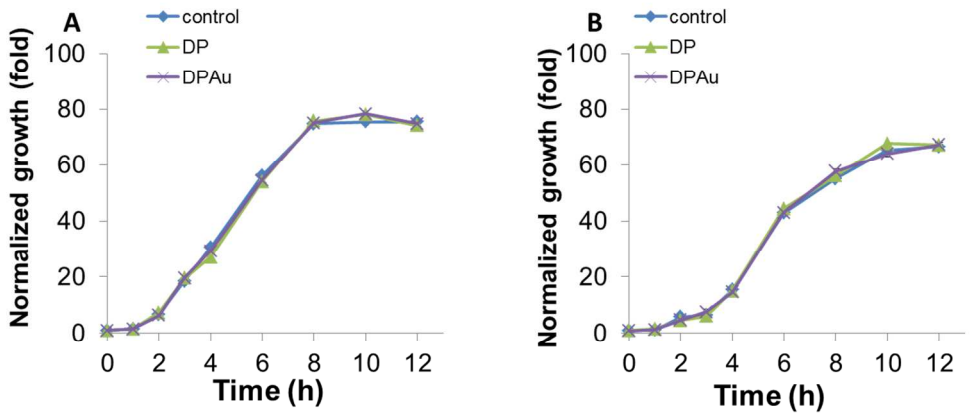

Figure S6. Bacterial growth curve control experiment show that both DP and DPAu do not exert any killing effect on the tested bacteria strains. (A) *E. coli* and (B) *S. aureus* were incubated with either 2 μM of DP or DPAu for 12 hours. Ultrapure water was used as vehicle control. Data is mean of three individual experiments.

SUPPORTING REFERENCE

(1) Yuan, X.; Luo, Z.; Zhang, Q.; Zhang, X.; Zheng, Y.; Lee, J. Y.; Xie, J., Synthesis of Highly Fluorescent Metal (Ag, Au, Pt, and Cu) Nanoclusters by Electrostatically Induced Reversible Phase Transfer. *ACS Nano* **2011,** *5* (11), 8800-8808.

(2) Shortman, K.; Lehman, I. R., The Deoxyribonucleases of *Escherichia coli*. VI. Changes in Enzyme Levels in Response to Alterations in Physiological State. *J. Biol. Chem.* **1964,** *239*, 2964-2974.

(3) Troller, J. A.; Stinson, J. V., Influence of water activity on the production of extracellular enzymes by Staphylococcus aureus. *Appl Environ Microbiol* **1978,** *35* (3), 521-526.

(4) ThermoScientific DNase I, RNase free. http://www.thermoscientificbio.com/dna-and-rna-modifying-enzymes/dnase-i-rnase-free/ (accessed April 2014).

(5) ThermoScientific pUC18, pUC19 DNA. http://www.thermoscientificbio.com/molecular-cloning/puc18-puc19-dna/ (accessed April 2014).