

Supporting information

Apoptosis-like Bacterial Death Modulated by Photoactive Hyperthermia Nanomaterials and Enhanced Wound Disinfection Application

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Table. S1. Concentration of extracellular Ca^{2+} and Mg^{2+} at different PTT temperature

Extracellular $\text{Ca}^{2+}/\text{Mg}^{2+}$ in <i>E. coli</i>	Temperatures ($^{\circ}\text{C}$)			
	30	45	55	61
Mg^{2+} ($\mu\text{g L}^{-1}$)	66.7 ± 0.65	82.3 ± 1.65	189.0 ± 1.50	193.4 ± 1.30
Ca^{2+} concentration	77.7 ± 1.15	97.9 ± 1.45	183.1 ± 1.53	191.7 ± 1.16

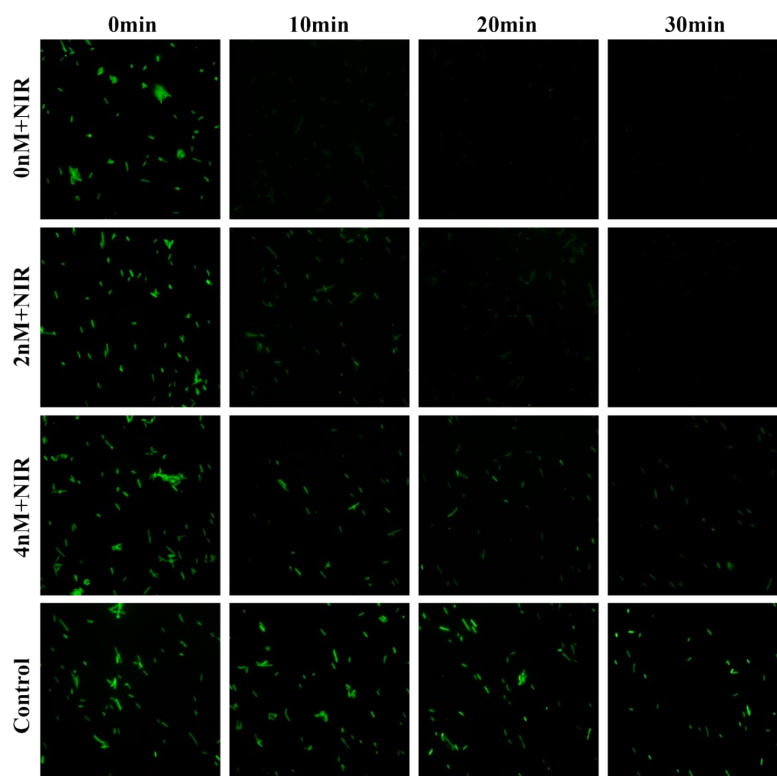


Figure S1. Changes in intracellular calcium homogeneity of bacteria treated with Amlopin at different concentrations of 0, 2, and 4 nM, and exposed to NIR radiation over time. The PTT conditions are of Au concentration = $30 \mu\text{g mL}^{-1}$, *E. coli* concentration = 10^7 CFU mL^{-1} , Total solution volume = $400 \mu\text{L}$, 808 nm NIR laser power density = 2 W cm^{-2} , $T_{\text{max}} = 50 ^{\circ}\text{C}$, irradiation time = 0-30 min.

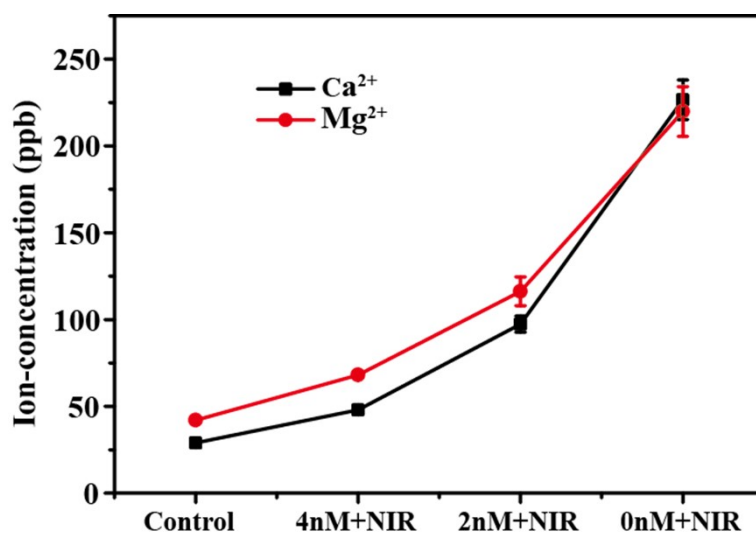


Figure S2. Corresponding extracellular calcium and magnesium ions concentration of bacteria treated with different Amlopin concentrations (0, 2, 4 nM) under PTT for 10 min. The PTT conditions are of Au concentration = $30 \mu\text{g mL}^{-1}$, *E. coli* concentration = 10^7 CFU mL^{-1} , Total solution volume = $400 \mu\text{L}$, 808 nm NIR laser power density = 2 W cm^{-2} , $T_{\text{max}} = 50^\circ\text{C}$.

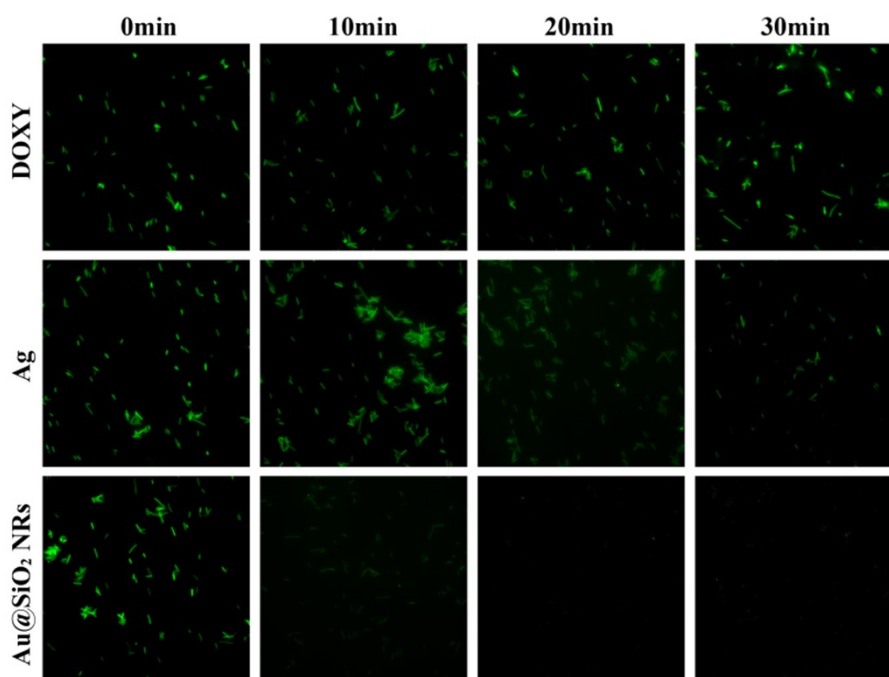


Figure S3. Changes in intracellular calcium homogeneity of bacteria treated with DOXY ($15 \mu\text{g mL}^{-1}$), Ag ($3 \mu\text{g mL}^{-1}$), and Au@SiO₂ NRs ($30 \mu\text{g mL}^{-1}$) exposed to NIR radiation (2 W cm^{-2}) over time.

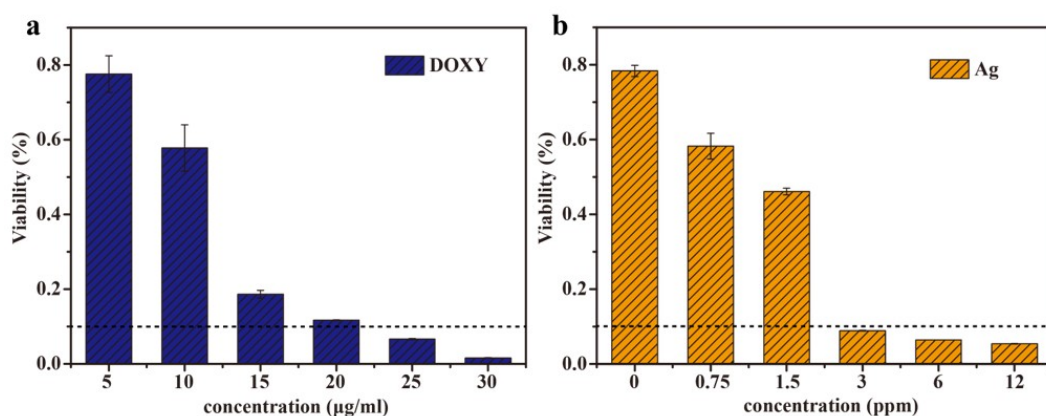


Figure S4. Antibacterial effect of different concentrations of DOXY (a) and Ag (b), which were incubated with 10^7 CFU mL⁻¹ *E. coli* for 1 h.

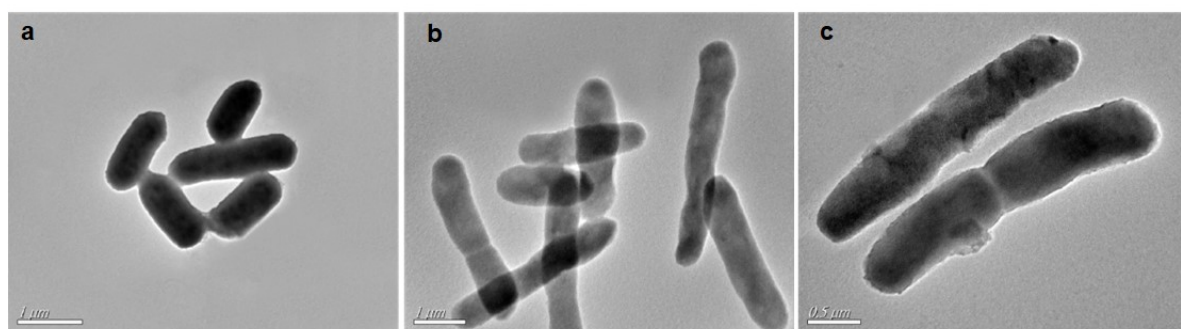


Figure S5. TEM images of *E. coli* (a), PTT treated *E. coli* at 55 °C (b) and 61 °C (c), respectively. PTT conditions: Au concentration, 30 μg mL⁻¹; *E. coli* concentration, 10^7 CFU mL⁻¹; Total solution volume, 400 μL; 808 nm NIR laser power density, 3 W cm⁻². Note: the PTT will stop immediately when reaching the desired temperature.

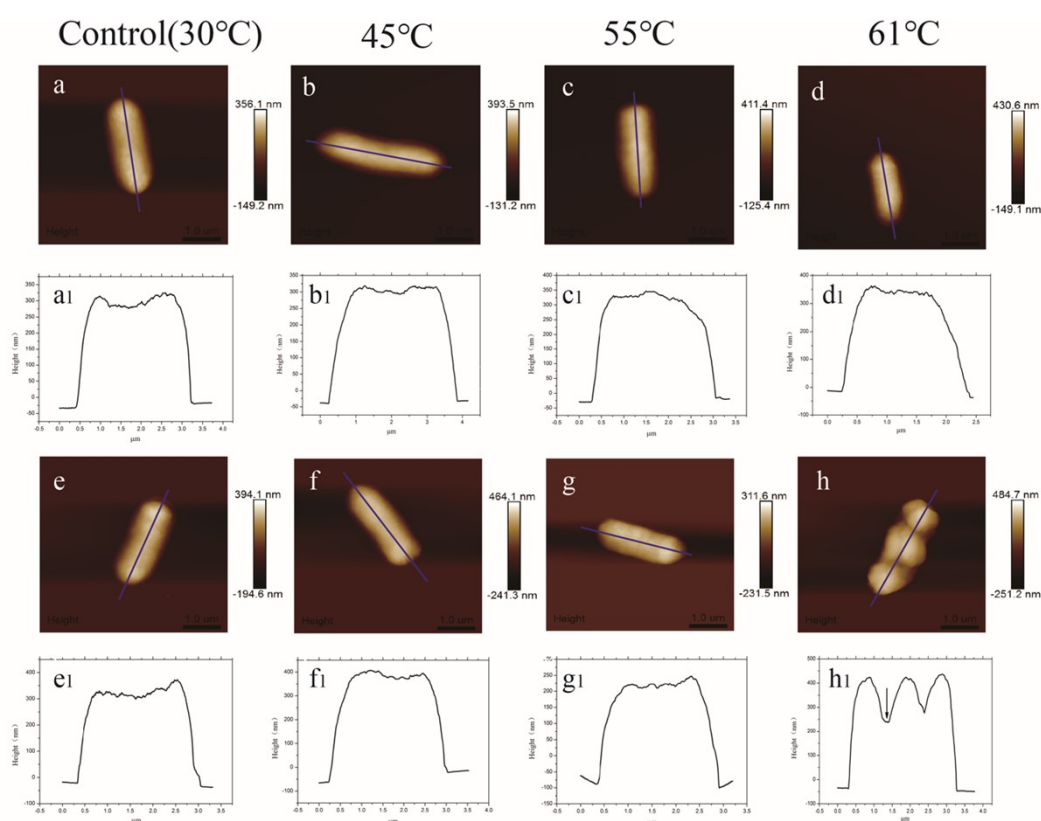


Figure S6. AFM tapping mode images of Gram negative *E. coli* in air. (a) AFM height image of individual healthy *E. coli* which mixed with PBS. (e) Height image of individual healthy *E. coli* mixed with Au@SiO₂ NRs. (b-d) Height images of *E. coli* treated by water-bath heating, when it reached 45, 55, 61 °C, respectively. (a₁-d₁) Cross-section taken along the blue line in height images. (f-h) Height images of *E. coli* treated by Au@SiO₂ NRs exposed to NIR laser irradiation (3 W cm⁻²), when temperatures are reached to 45, 55, 61 °C respectively. (e₁-h₁) Cross-section taken along the blue line in height images. **Note:** All scan range are 5 μm².

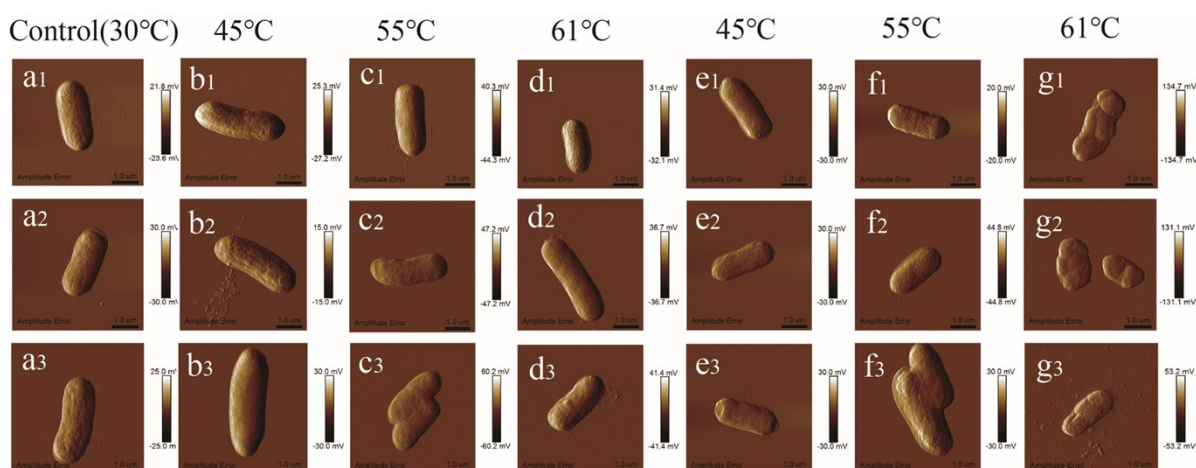


Figure S7. AFM tapping mode images of Gram negative *E. coli* in air. (a1, a2, a3) control, i.e., the

amplitude images of untreated *E. coli* at 30°C. (b1, b2, b3; c1, c2, c3; d1, d2, d3) the amplitude images of *E. coli* which are treated by water-bath heating with temperature of 45 °C, 55 °C, 61 °C, respectively. (e1, e2, e3; f1, f2, f3; g1, g2, g3) the amplitude images of *E. coli* treated by Au@SiO₂ NRs exposed to NIR laser irradiation (3 W cm⁻²). **Note:** All scan range are 5 μm².

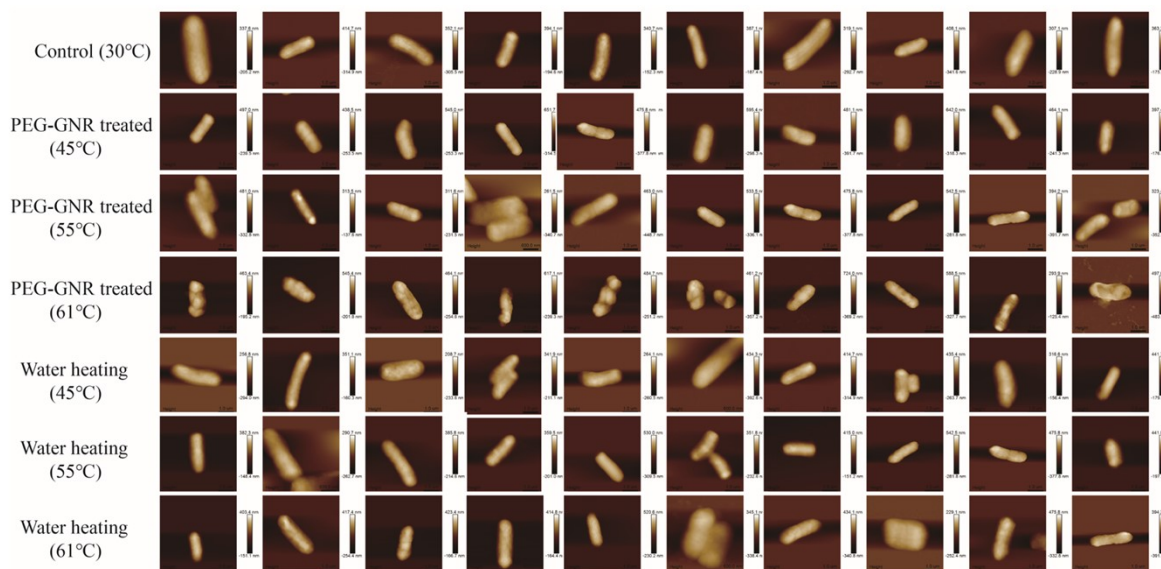


Figure S8. AFM tapping mode images of Gram negative *E. coli* in air. AFM height images at every temperature.

Table S2. W: water bath heating; A: Au@SiO₂ NRs treated under NIR. Average surface roughness of 80×300 nm² areas on 10 cells at each temperature.

Average surface roughness (nm)											
Bacteria	1	2	3	4	5	6	7	8	9	10	Average
Healthy	8.88	7.58	8.35	9.51	8.90	11.2	10.9	10.7	9.60	9.61	9.522
45 (W)	11.4	9.97	10.0	9.19	8.38	9.61	8.89	9.47	9.10	9.49	9.551
55 (W)	9.92	10.8	9.82	10.2	9.81	11.47	10.4	9.79	9.09	10.0	10.12
61 (W)	11.4	10.9	10.0	10.9	10.2	9.80	10.6	11.0	10.9	10.3	10.61
45 (A)	12.0	11.9	11.1	9.87	10.3	10.30	11.1	11.9	11.9	11.8	11.23
55 (A)	13.8	15.3	17.7	13.0	11.7	15.51	14.9	15.4	13.4	15.1	14.58
61 (A)	17.2	26.9	20.5	21.7	27.2	24.21	19.9	26.0	19.5	16.6	23.15

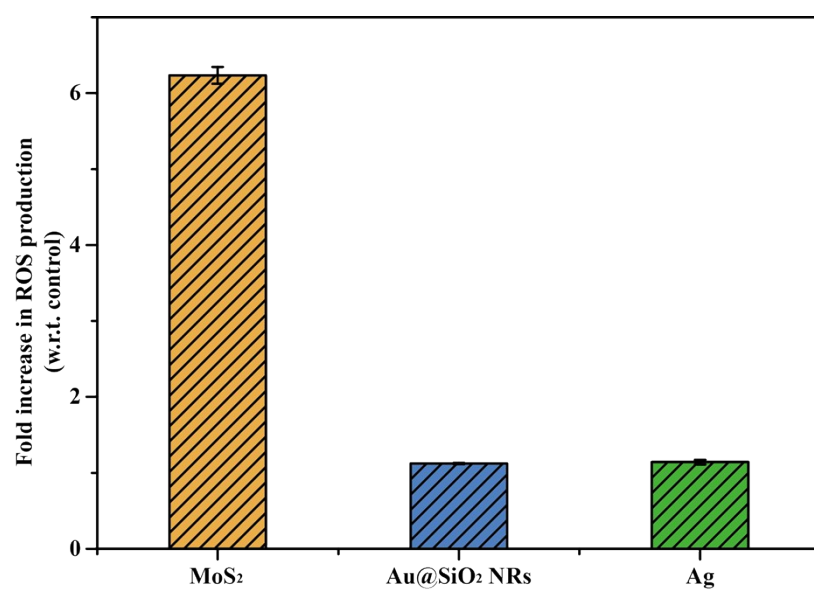


Figure S9. Compared with MoS₂, Au@SiO₂ NRs and Ag produce almost no ROS.