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Electronic Supplementary Information (ESI†)

Gold Nanopopcorn Attached Single-Walled Carbon Nanotube Hybrid for Rapid Detection and Killing of Bacteria

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Contents:

1. Additional Experimental details (S1)

Attachment of Popcorn-shaped AuNP to f_3-SWCNTs: The attachment of the Popcorn-shaped gold nanoparticles onto the thiol-terminated SWCNTs was carried out by adding an excess of AuNP colloids in order to achieve the highest possible surface attachment onto the SWCNTs. Briefly, 10 mg of f_3-SWCNTs were dispersed in 10 mL of nanopure water via sonication for 5 min. AuNP colloids was added to the f_3-SWCNTs dispersion dropwise, and the mixture was agitated until the mixture retained the bluish color, characteristic of the popcorn shaped AuNP. The mixture was left at rest for 12 hrs. The clear supernatant was removed and the process was repeated until the supernatant retained the bluish color. The mixture was centrifuged at 3500 rpm for 45 minutes to remove the excess unbound AuNP from the mixture and finally re-dispersed in 10 mL of nanopure water.

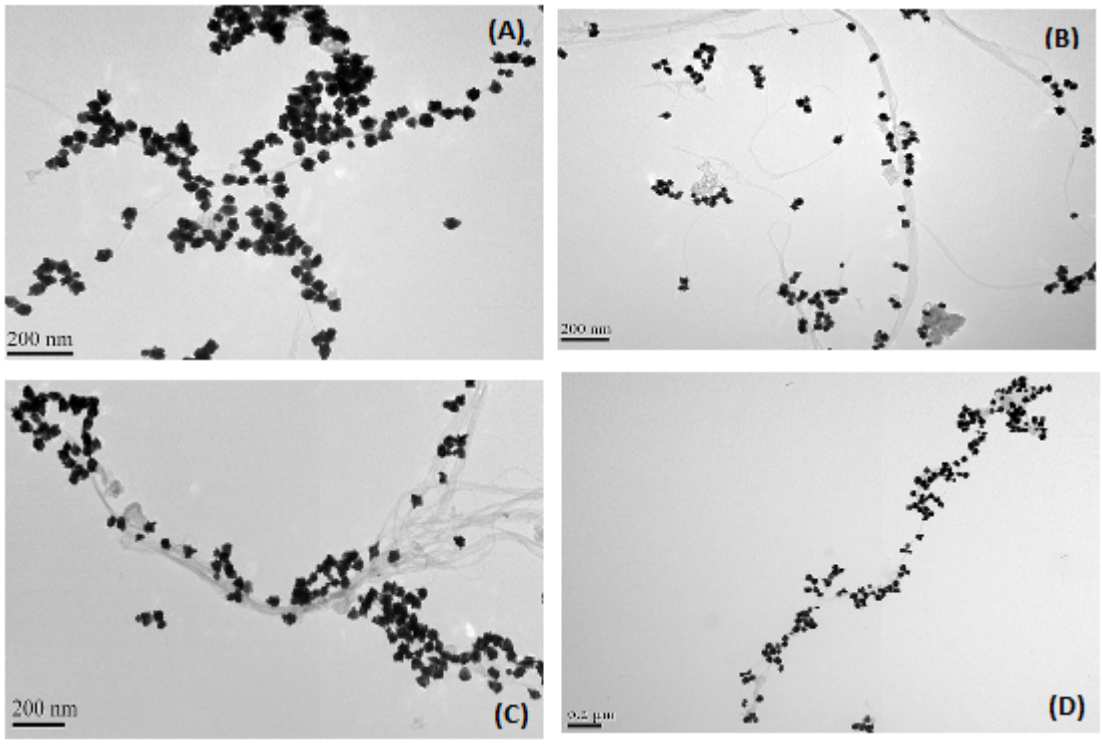
Microwave irradiation experiments: Microwave-assisted reactions were carried out in Biotage initiator Eight EXP 2.0 single-mode microwave cavity producing controlled irradiation at 2450 MHz (400 W max magnetron power output) in 20-mL microwave process vials for sealed-vessel reactions (Biotage AB, Uppsala)

Transmission electron microscopy (TEM) measurements 5μL portion of the sample solutions was dropped on carbon-coated copper grid (Cat. # 01801, Ted pella Inc.CA). The grid was then air-dried. The samples were examined using Transmission electron microscope (TEM) JEM-2100F (JEOL, Tokyo Japan) at 200 kV.

UV/Vis spectroscopic measurements UV/Vis absorption spectra were obtained using UV-Shimadzu 2600/2700 UV/Vis/NIR spectrophotometer, which is run by Varian's Cary Win UV software version 2.0.

OD measurements. The spectrophotometer was blanked using water (suspending fluid). The spectrophotometer was set at 600 nm. Bacterial cultures at OD_{600} were centrifuged and washed twice to yield a final bacterial concentration of approximately $1.3X10^8$ CFU/mL. Turbidity measurements were carried out to monitor the bacteria capturing selectivity of mAb conjugated-AuNP@f_3-SWCNTs. OD_{600} was used to estimate the bacteria concentration in the medium[1]. Typically, 0.5 mL of mAb conjugated-AuNP@f_3-SWCNTs was added to 1.5 mL $1.3X10^8$ CFU/mL of bacterial suspension. Samples were mechanically agitated using a pipette for 40 seconds and absorbance acquired 30 seconds after. mAb-conjugated-AuNP@f_3-SWCNTs along with the captured bacteria aggregated at the bottom of the cuvette. Turbidity of the suspension was determined after every 70 seconds for 16 minutes.

2. Supporting figures for the controlled decoration of gold nanoparticles onto SWCNTs



Supplementary Figure S2

TEM image showing antibody conjugated popcorn-shaped gold nanoparticles attached on: (A) previously ester modified and 1,3-cycloaddition functionalized SWCNTs (AuNP@f_3SWCNT) after 12 hours incubation, (B) 1,3-cycloaddition functionalized SWCNTs (AuNP@SWCNT), (C) Dilute acid modified SWCNT, (D) previously ester modified and 1,3-cycloaddition functionalized SWCNTs (AuNP@f_3-SWCNT) 3 weeks later.

3. Tables for cytotoxicity studies and viability of bacteria after photothermal treatment.

Cytotoxicity studies

1.3 X 10^3 CFU/mL of *E. coli* samples were treated with mAb-AuNP@f_3-SWCNT hybrid nanomaterial. These samples were incubated for 30 mins at room temperature as per the table below. 50µL of the treated samples were plated in triplicate on agar plates and incubated at 37 °C for 18 hours.

Table S3-a. Viability rate of bacteria in different concentrations of the hybrid nanostructure probe

Viability Rate (N/No)[a] %

		Bacteria: probe ratio			
Bacteria	**Bacteria Only**	**1:2**	**1:1**	**2:1**	**3:1**
***E. coli* 49979**	**100**	**86.3±5.2**	**94.8±4.2**	**98.0±2.9**	**98±2.5**

[a](N= number of viable bacteria, N_o= number of original bacteria). These are averages from three measurements.

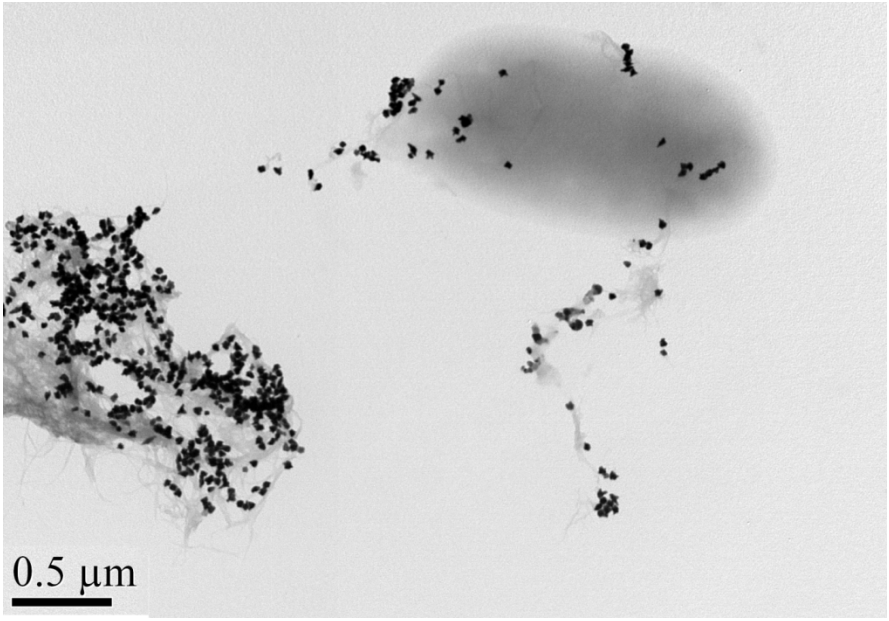
Photothermal Laser treatment, General procedure $1.3x10^3$ CFU/mL of samples without and with antibody-conjugated -hybrid nanomaterial and -gold nanopopcorns were exposed in an optical cuvette to a 670 nm laser at 1.5-2.5 W/cm^2 power. Use of a 2-mm-square pinhole in front of the cuvette provided a relatively uniform beam-intensity profile and there was a relatively uniform exposure of the entire sample in the cuvette. 50µL of the treated samples were plated in triplicate on agar plates and incubated at 37 °C for 18 hours.

Table S3-b. Viability rate of bacteria after photothermal treatment

	Viability Rate (N/No)[a] %						
			Bacteria + probe +NIR (ratio 3:1)				
Bacteria	Bacteria + NIR (15mins)	Bacteria+ Au NP+NIR (15mins)	t=3mins	6	9	12	15
E. coli 49979	96.33±0.88	56.67±3.21	89.33±4.04	60.67±4.16	21.0±3.6	6.3±1.53	2.67±0.58
Salmonella DT104	95.47±0.86	86.14±2.14	94.2±2.8	91±4	85.08±1.80	84±2.06	84.56±1.86

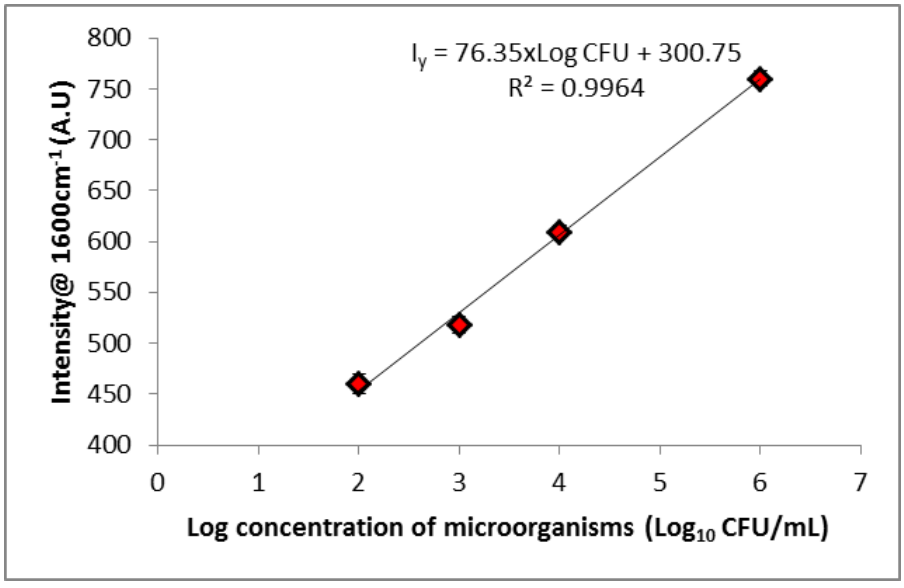
[a](N= number of viable bacteria, N_o= number of original bacteria). These are averages from three measurements.

4. Additional Data



Supplementary Figure S4: TEM for Salmonella DT104 in mAb-AuNP@f_3-SWCNT showing poor binding.

Supplementary Figure S5



A Logarithmic plot of *E. coli* concentration vs the signal intensity for the band at 1600 cm^{-1}.

Upon addition of different concentrations of E. coli, a linear relationship between the Raman intensity (I_y) value (at 1600 cm^{-}) and the log value of the concentration of the bacteria cells could be obtained, where the fitted equation could be expressed as I_y=76.35 X Log CFU + 300.75, and R^2= 0.9964.

5. REFERENCES

1. E.W. Nester, D.G. Anderson, C.E. Roberts Jr. and M.T. Nester, *Microscopy and Cell Structure In Microbiology: A Human Perspective,* Peterson, K. A. Ed.; McGraw-Hill, New York, 2007, 55-82