Supplementary Information for:

"Using bacterial cell growth to template catalytic asymmetry"

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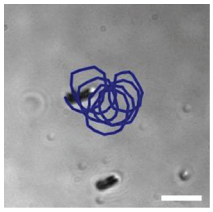
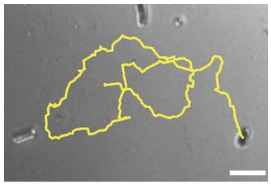
Captions for Supporting Movies

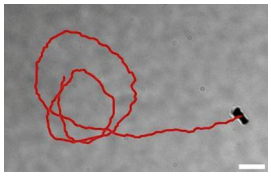
(All movies replay at 5× real time. Scale bars for movie stills are 5μm.)



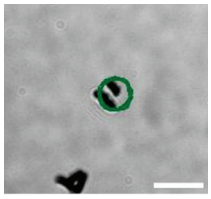
Movie S1: Video showing clockwise (left) and counterclockwise (right) movement of bacterial-templated Pt/Au bimetallic particles. The image above (from Fig. 3B, upper right panel) depicts the trajectory of the clockwise moving particle.



Movie S2: Video showing motion of a bacterial-templated Pt/Au particle turning both left and right (Fig 3B, upper left panel).



Movie S3: Video showing counter-clockwise biased motion of a bacterial-templated Pt/Au particle (Fig 3B, lower left panel).



Movie S4: Video showing counter-clockwise rotation of a bacterial-templated Pt/Au double particle (Fig 3B, lower right panel).

Materials

Positively charged gold nanoparticles (2022) and gold enhancement solution (2112) were purchased from Nanoprobes (Yaphank, NY), Potassium tetrachloroplatinate (K_2PtCl_4 99.99%), L-ascorbic acid (AA) (99.0%), and phosphate buffered saline (PBS1) were used as received from Sigma-Aldrich (St. Louis, MO).

Bacterial cell culture and metallization

E. coli strains RP437 (wild type, kindly supplied by J.S. Parkinson, University of Utah) were streaked on 1.5% agar (214050, Becton Dickinson) containing T-broth (1% Tryptone [211705, Becton Dickinson], 0.5% wt/vol NaCl) and grown at 37 °C. Single-colony isolates were used to inoculate a solution of T-broth, which were grown to stationary phase on a rotary shaker at 37 °C. RP437 carrying the pS2-GFP vector were used occasionally to enable visualization. Typically 2 mls of saturated liquid cell culture was centrifuged (2 min at 5000 rpm) and the pellet was rinsed/resuspended three times (in phosphate buffered saline, PBS; pH 7.0), with a final resuspension in 1 ml of a 1:1 mixture of PBS:H_2O. To this solution was added 40 μL of positively charged 1.4 nm gold nanoparticles (AuNPs) in H_2O (60 μM) and the solution was shaken gently for ~20 mins. Cells were centrifuged, rinsed (3x in PBS) and AuNPs were incubated (immediately or following incubation in TB growth media at 10-100x dilution of cells) in gold enhancement solution (10 min) or a platinum solution (~20 min) containing 20 mM K_2PtCl_4 dissolved in H_2O to which was added solid L-ascorbic acid to a final concentration of 80 mM. Metallized bacteria were rinsed (H_2O) and deposited on glass (for optical analysis) or silicon (for SEM analysis) substrates.

Cells undergoing sequential metallization reactions (interceded by a cell growth step) were prepared as described above followed by 1) incubation in 10 mM cystamine (C8707, Sigma) for ~ 60 min to coat metallized surfaces with positive charge, thereby directing AuNPs (incubated for ~ 60 min) to the nascently formed cell envelope; 2) development of AuNP seeds as described above. Additionally, in the absence of cystamine, large aggregates of particles formed during Pt metallization, indicating a stabilizing role of cystamine for Pt/Au particles. All incubations in this study were performed at room temperature unless otherwise noted.

Zeta potential measurements

For zeta potential measurements, 10 μL were collected from an *E. coli* growth culture (2 mL TB broth at 37°C) at time points noted in Figure 1D, dispersed in 0.6 mL H_2O and transferred to a disposable cuvette for zeta-potential measurement. Measurements were performed on a Zetasizer Nano dynamic light scattering instrument (Malvern) at 25 °C.

Imaging

Optical microscopy images and movies were acquired on a Nikon Eclipse Ti inverted microscope with a 100X objective and an iXon 885 EMCCD camera. Image analysis was performed using Image J. A Hitachi S-5200 scanning electron microscope (SEM) equipped with an Energy Dispersive Spectrometer (EDS) from Princeton Gamma Tech (PGT) was used in spotlight, linescan, and mapping modes to image metallized cells and perform elemental analysis.

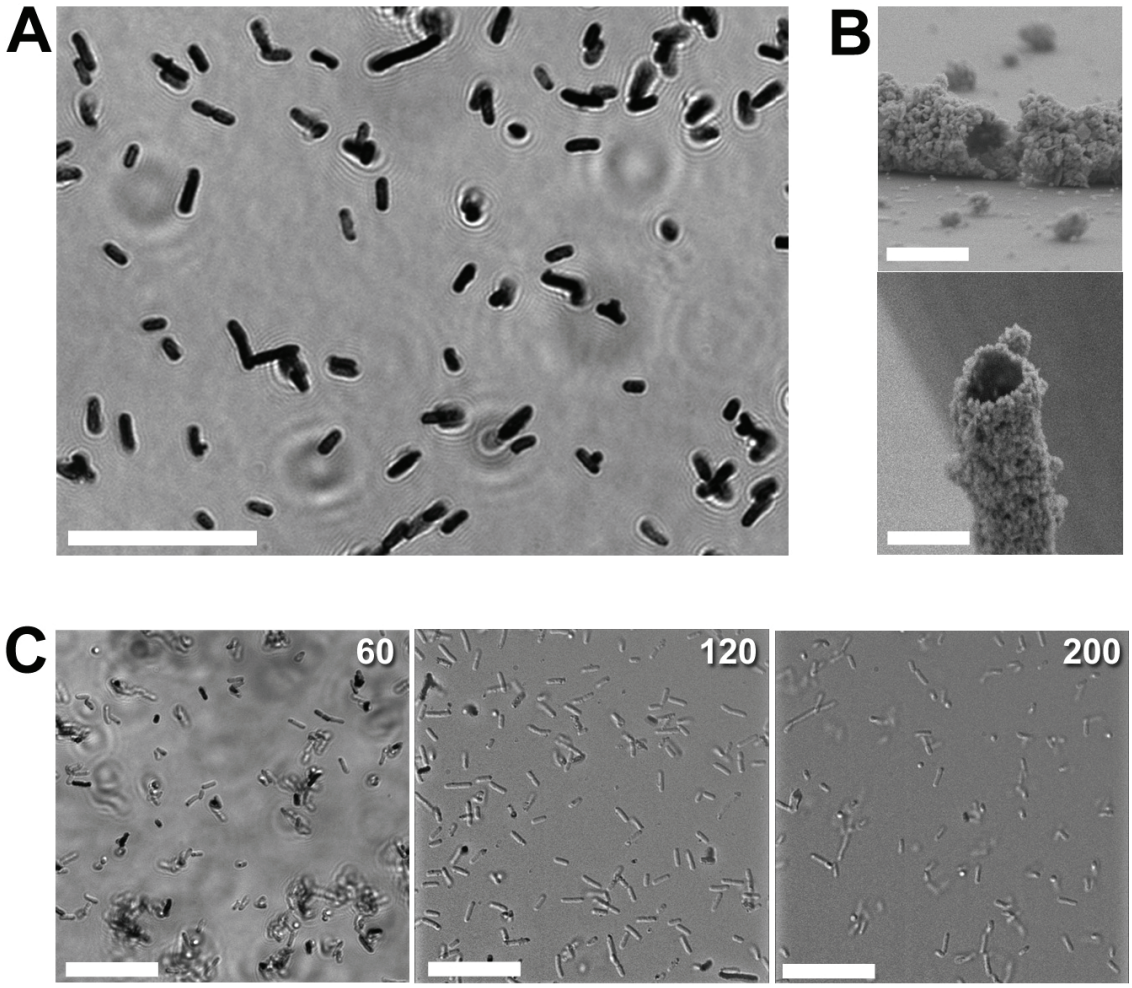


Figure S1: Optical and SEM characterization of *E. coli* cells following incubation with positively charged gold nanoparticles (AuNPs). (A, optical micrograph; scale bar, 10 μm) Autometallographic development of AuNPs bound to stationary phase cells indicates AuNP binding occurs homogenously over the whole cell envelope across the population, resulting in hollow metallic shells (B, SEM image; scale bars, 1 μm) following prolonged drying of particles (desiccation >72 hrs). (C, optical micrographs; scale bars, 10 μm) Development of AuNPs following growth of *E. coli* cells in liquid culture shows heterogeneous coverage (left and middle panel) and dissipation (all panels) of AuNPs from the population over time. Numbers indicate elapsed time (in minutes) in nutrient conditions (TB-broth, 37°C).

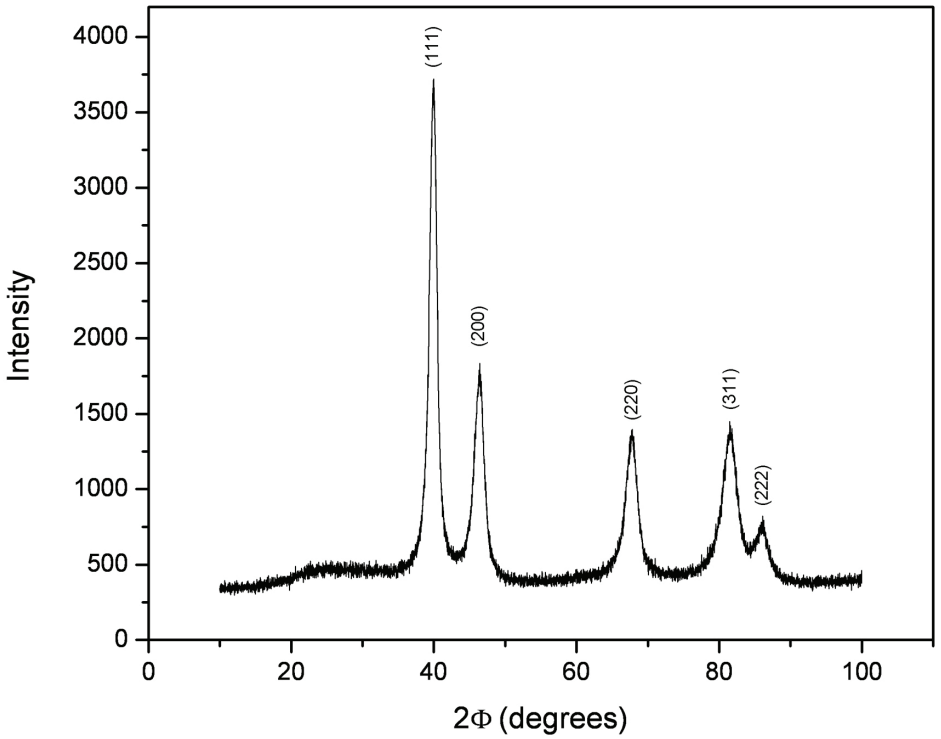


Figure S2: X-ray diffraction (XRD) of *E. coli* templated platinum. Platinized cells were prepared as described in the methods section above (without a cell growth step) and kept under desiccated conditions for ~ 80 hrs. The peaks were fit to face-centered cubic (fcc) platinum (space group: *Fm*3*m*) and the average size of single-crystalline particles (τ) was estimated using the Scherrer equation $\tau = K\lambda/\beta\cos\theta$, where K is the shape factor (0.9), λ is the x-ray wavelength (1.54 Å), β is the full width at half max (FWHM), and θ is the Bragg angle. This analysis gave a minimum size of ~5 nm thickness for the platinum single-crystalline particles.

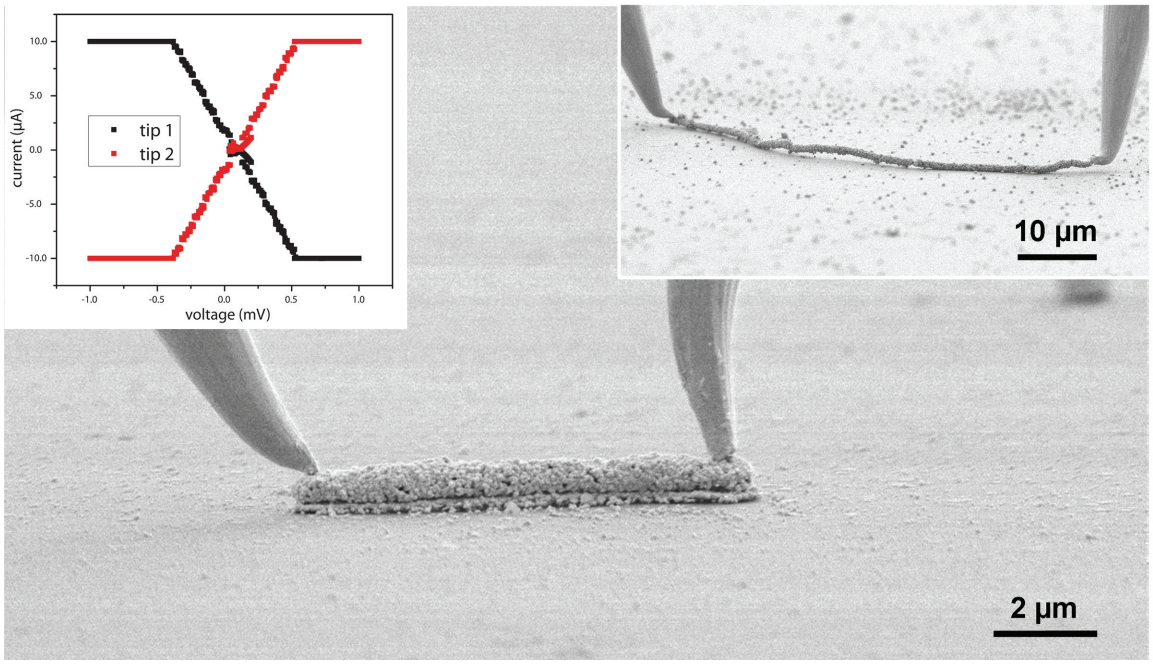


Figure S3: The conductivity of *E. coli* templated gold wires was investigated using W nanoprobes in direct contact with metallized cells (above). A typical *IV* response is shown in the upper left inset. The voltage sweep (-1.0 to +1.0 mV at a limit of ± 10.0 μA) from tip 1 is recorded by tip 2 to determine the resistance of the ohmic wire (R = 1/slope). The resistivity (ρ) of the above wire was 8.0×10^{-7} Ωm (bulk gold = 2.44×10^{-8} Ωm). The upper right inset shows a metallized cell that was elongated by first exposing the bacterial template to 20 μg/mL cephalexin for two hours under growth conditions (TB broth at 37 °C) before metallization procedures. Elongated wires displayed similar ohmic behavior with resistivities on the order of 10^{-6} - 10^{-7} Ωm.

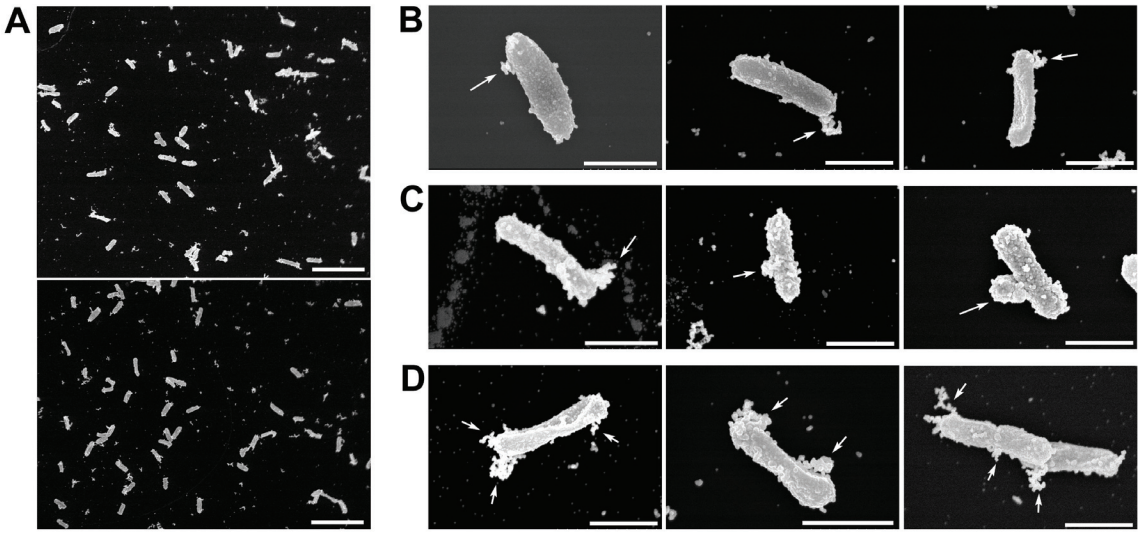


Figure S4: Pt metallization following cell growth and gold deposition. (A) Representative backscatter SEM images of Pt/Au particles. Particles display single dendrite-like (B) nodule (C) or multiple (D) platinum outgrowths (arrows) extending from the cell envelope. Scale bars: A, 10μm; B, C, D, 2 μM).