SUPPLEMENTARY INFORMATION

Surface Plasmon Resonance Imaging (SPRi) for Multiplexed Evaluation of Bacterial Adhesion onto Surface Coatings

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Surface Coating Stability

To better understand the stability of the coatings on the surface, we carried out a control experiment coating the sensor surface with all three coatings used in this study, and flowing sterile LB without bacteria over the coatings. Since there should be no bacterial growth in this experiment, any variations in reflectivity over time would be due to the coatings changing on their own. Fig. S1 shows that the brightness of the three regions in the images does not change over the course of 10 hours, indicating no change in surface coating properties and supporting our conclusion that the proteins affect bacterial adhesion.

The center region of the sensor, coated with penicillin/streptomycin, is brighter than the BSA and casein coated regions throughout the entire experiment. This was observed from the start of the experiment, and indicates that the antibiotic coating is thicker than the casein and BSA coatings [1].



Fig. S1 SPRi difference images generated from the surface coated with BSA (left), penicillin/streptomycin (middle) and casein(left) after a) 2, b) 6, c) 10 hours of flowing sterile LB over the surface.

Viability of Bacterial Cells

To confirm that the bacteria remain viable after attaching to the coatings, we performed a series of additional experiments. First, after the prism and chip were removed from the device at the end of experiments with CFP-PA01, the sensor surface was covered with a microscope cover slip. Inspection of the surface with a fluorescence microscope revealed many fluorescing bacterial cells on the surfaces of all 3 regions, signifying viability (Fig. S2a). Inspection of the biofilms with reflected light bright field microscopy showed many moving cells (Fig. S2b). We also collected samples of the solution above the sensor and scraped biofilm samples from each of the coating regions on the sensor surface with a sterile plastic toothpick. We spread the collected samples onto culture plates and incubated them at room temperature and 37 °C to test for growth. New growth was observed on all of the plates, indicating viable cells on the sensor surface both with and without coatings (Fig. S3).



Fig. S2 Micrographs of CFP-PA01 bacteria on the sensor surface obtained after the end of the SPRi experiment using with (a) 40X florescence and (b) 100X reflected light bright field microscopy.



Fig. S3 CFP-PA01 cultures grown from samples collected after SPRi experiments from: (a,e,h) solution inside the PDMS chamber; (b,d,i) biofilm on the casein coated region; (c,f,j) biofilm on the bare gold region; and (d,g,k) biofilm on the penicillin/streptomycin coated region. (a-d) at time 0, which refers to 24 hours after starting SPRi experiment. (e-g) after 24 hours of incubation at room temperature. (h-k) after 72 hours of incubation at 37 °C.

References

 C. Haoyuan, C. Dafu and Z. Lulu, *IEEE J Nanotechnol*, 2012, 12th IEEE Nanotechnology Conference Proceedings (IEEE-Nano), 1-3.