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Building spatially-structured biofilms with single-cell control using laser trapping CHRISTOPHER RODESNEY, JAIME HUTCHISON, KARISHMA KAUSHIK, HENRY LE, DANIEL HURWITZ, Univ of Texas, Austin, YASUHIKO IRIE, Univ of Bath, VERNITA GORDON, Univ of Texas, Austin — Biofilms are sessile communities of microbes adhered to each other and to an interface. Biofilm infections are notoriously difficult to eradicate, and this arises in part from phenotypic changes due to the spatial structure of the biofilm. Spatial structure controls the microenvironment and intercellular associations, which in turn controls gene expression, virulence, and antibiotic resistance. There are few tools available for elucidating the role of spatial structure in biofilms. We present a method for controlling the positions of bacteria on a surface using optical trapping without impinging cell viability. Initial positions propagate into the developing biofilm, creating spatial structure. The native growth, motility, and surface adhesion of positioned cells are preserved, as shown for model organisms *Pseudomonas aeruginosa* and *Staphylococcus aureus*. We demonstrate statistically-significant effects of spatial structure on the growth of monoculture *P. aeruginosa* biofilms and for co-culture biofilms of *P. aeruginosa* and *S. aureus*. Because the laser trap we use is very basic and the other equipment required is inexpensive and standard, we believe that our technique will be a widely-usable tool for biological and physical collaborators at many types of institutions.

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