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**Mechanics governs single-cell signaling and multi-cell robustness in biofilm infections.<sup>1</sup>**

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In biofilms, bacteria and other microbes are embedded in extracellular polymers (EPS). Multiple types of EPS can be produced by a single bacterial strain - the reasons for this redundancy are not well-understood. Our work suggests that different polymers may confer distinct mechanical benefits. Our model organism is *Pseudomonas aeruginosa*, an opportunistic human pathogen that forms chronic biofilm infections associated with increased antibiotic resistance and evasion of the immune defense. Biofilms initiate when bacteria attach to a surface, sense the surface, and change their gene expression. Changes in gene expression are regulated by a chemical signal, cyclic-di-GMP. We find that one EPS material, called "PEL," enhances surface sensing by increasing mechanical coupling of single bacteria to the surface. Measurements of bacterial motility suggest that PEL may increase frictional interactions between the surface and the bacteria. Consistent with this, we show that bacteria increase cyclic-di-GMP signaling in response to mechanical shear stress. Mechanosensing has long been known to be important to the function of cells in higher eukaryotes, but this is one of only a handful of studies showing that bacteria can sense and respond to mechanical forces. For the mature biofilm, the embedding polymer matrix can protect bacteria both chemically and mechanically. *P. aeruginosa* infections in the cystic fibrosis (CF) lung often last for decades, ample time for the infecting strain(s) to evolve. Production of another EPS material, alginate, is well-known to tend to increase over time in CF infections. Alginate chemically protects biofilms, but also makes them softer and weaker. Recently, it is being increasingly recognized that bacteria in chronic CF infections also evolve to increase PSL production. We use oscillatory bulk rheology to determine the unique contributions of EPS materials to biofilm mechanics. Unlike alginate, increased PSL stiffens biofilms. Increasing both PSL and alginate expression increases the energy cost to break the biofilm. We compare the elastic moduli of biofilms to estimated stresses exerted by phagocytotic immune cells, and infer that increased PSL could confer a mechanical fitness benefit.

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