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## Automated AFM for Large-Area Biofilm Imaging and Characterization of Antifouling Surfaces

Biofilms are complex microbial communities encased in an extracellular matrix, exhibiting structural and functional heterogeneity driven by chemical and nutritional gradients. Their architecture enhances resilience and antibiotic resistance, posing challenges in clinical and industrial contexts. Traditional analysis methods often lack the spatial resolution and contextual integrity needed to capture their full complexity.

To overcome these limitations, we developed an automated platform for large-area Atomic Force Microscopy (AFM) imaging, capable of high-resolution mapping over millimeter-scale regions. Combined with image stitching and machine learning-based analysis, our approach preserves spatial relationships and reveals key biofilm features, including cellular arrangements, and flagella.

We applied this platform to study *Pantoea* sp. YR343 biofilms on gradient-structured and antifouling surfaces, revealing how surface properties influence biofilm morphology and stability. In particular, we observed a honeycomb architecture on hydrophobic regions and demonstrated the role of flagella in structural integrity. By automating AFM imaging, we extend its applicability to mesoscale investigations, offering a powerful tool for characterizing biofilm-surface interactions and advancing the design of antifouling materials.

### Topical Area

Biology and life sciences

**Author:** Dr MILLAN-SOLSONA, Ruben (ORNL)

**Co-authors:** BROWN, Spenser R.; CHECA, Martí; MADUGULA, Sita Sirisha; ZHANG, Lance; ZHAO, Huan-Huan; WEBB, Amber; LAVRIK, Nickolay V.; VASUDEVAN, Rama; BISWAS, Arpan; RETTERER, Scott; MORRELL-FALVEY, Jennifer L; COLLINS, Liam

**Presenter:** Dr MILLAN-SOLSONA, Ruben (ORNL)