Quantifying the structural evolution of Pseudomonas putida biofilms under flow

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Biofilms are surface-associated bacterial communities embedded in an extracellular polymeric substance (EPS) matrix, whose structure governs transport, mechanical stability, and function. However, quantitative, time-resolved characterization of biofilm architecture under controlled flow conditions remains limited.
Here, we combine time-lapse confocal microscopy with microfluidic cultivation to track the three-dimensional evolution of *Pseudomonas putida* biofilms under well-defined flow, nutrient, and temperature conditions [1]. Image data are analysed using a convolutional neural network for robust segmentation. From the segmented data, we extract a comprehensive set of structural descriptors capturing local density, orientational order, and spatial correlations. These metrics enable a quantitative description of biofilm architecture across scales, from single-cell organization to collective structure. Preliminary results indicate a partial alignment of cells with the flow, and an initial increase in local density and coordination, after which a dynamical equilibrium between growth and shear-induced removal sets in.
We show that flow conditions and nutrient availability systematically modulate biofilm architecture, affecting both local cell organization and long-range structural correlations during growth. This approach establishes a quantitative framework to link environmental conditions to biofilm structural evolution, and highlights the potential of integrated microfluidics and imaging for probing biofilm structure.

Keywords:
Biofilms, microfluidics, confocal microscopy, image segmentation, structural descriptors, multi-scale architecture

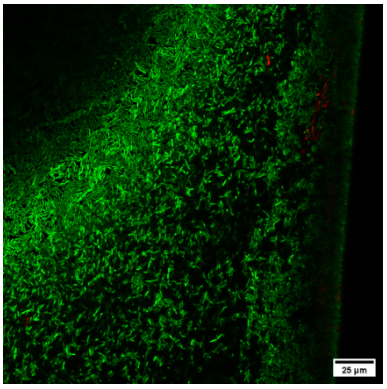

Figure: confocal micrograph of a P. putida biofilm 3h after start of the experiment (left). The live cells are visible in green, the dead cells in red. Image taken 5 µm below the PDMS surface of the microfluidic channel.

References:

[1] Steffen Geisel, Eleonora Secchi, Jan Vermant. eLife. 2022.

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