

## Project Title

Structural mapping of the *in vivo* *Pseudomonas aeruginosa* biofilm matrix exopolysaccharide, Psl, does it change with virulence?

### Summary

Quantitative 3D mapping of Psl exopolysaccharide in 72 murine tissue samples using IHC, CLSM and automated ZEISS Intellesis segmentation to correlate biofilm structure with genotypic virulence.

**My Role:** Lead Researcher - designed wet-lab pipeline, imaging workflow and automated image-analysis.

### Methods

Sample prep	Tissue fixation, embedding, H&E, IHC staining, PA01 antibody, HHL-TRITC, POPO1.
Imaging	Confocal Microscopy (CLSM), ZEISS Axioscan Slide scanner, fluorescent, brightfield & pol
Image Analysis	ZEISS Intellesis machine-learning segmentation pipeline; and metrics quantification
Stats and Plots	Data cleaning and statistics in R and GraphPad Prism (Two-way RM ANOVA, post-hoc tests, correlation tests).

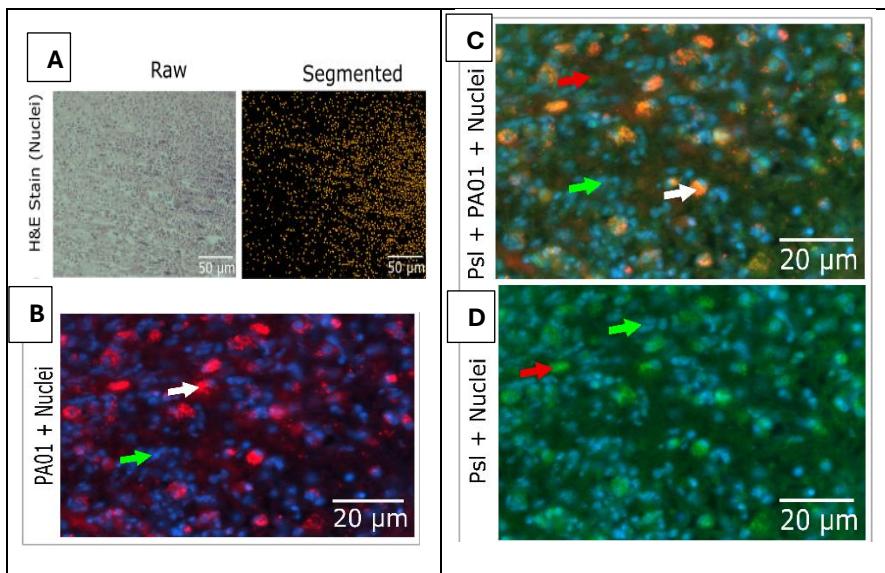
### Key Results

- Processed and analysed **72 tissue samples** with a consistent imaging pipeline. Automated segmentation reduced manual processing time by ~70% and produced reproducible volume metrics (CV < 8%).
- Statistically significant correlation between Psl volume and virulence genotype ( $p < 0.01$ ). Biofilm/aggregate is not solely dependent on Psl expression levels.

### Impact / Takeaway

Provided a reproducible, high-throughput imaging + analysis pipeline enabling data-driven comparisons across genotypes - directly translatable to assay validation or phenotypic screens.

### Images and Plots



**Figure X. (A)** Representative ROIs stained with H&E and acquired using ZEISS Axioscan 7 slide scanner 20× objective. Raw images were segmented and analysed in ZEISS Intellesis.

**(B-D)** Representative image (CE group) showing bacterial aggregates (white arrows), Psl clusters (red arrows) and host nuclei (POPO-1, green arrows). Acquired on ZEISS Axioscan 7, 20×; scale bar = 20 μm.

