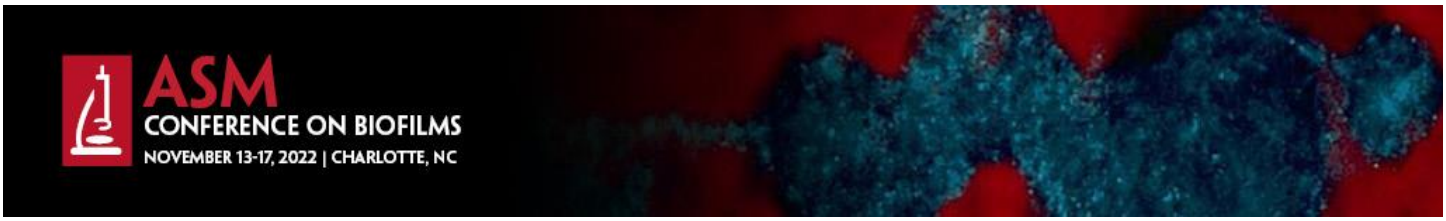

Surveying the Battlefield: Using *In Situ* Information To Recreate Infection-relevant Biofilms *In Vitro*

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We lack fundamental knowledge about biofilm structure and function in native environments. My lab works with nontuberculous mycobacteria (NTM), specifically the opportunistic pathogen *Mycobacterium abscessus* and the model strain *Mycobacterium smegmatis*, to determine how aspects of the infection environment affect biofilm dynamics *in vitro*. In Synthetic Cystic Fibrosis Medium (SCFM), carbon abundance enhances *M. abscessus* aggregation while nitrogen abundance favors dispersal and growth as planktonic cells. Intracellular glutamine serves as a nitrogen indicator in many bacteria, and we have evidence that the glutamine pool impacts biofilm formation in *M. smegmatis*. We are exploring the mechanism through which *M. smegmatis* translates intracellular glutamine to a biofilm phenotype and whether *M. abscessus* has similar regulatory pathways. In addition, we have found that low oxygen conditions, which are common in parts of the CF lung, prevent *M. smegmatis* from either forming or dispersing biofilms. We are investigating how the NTM anoxic dormancy response, specifically the DosSR two-component system, is involved in this process.

In parallel, we are utilizing the tissue-clearing/bacterial labeling technique, MiPACT-HCR, to characterize *M. abscessus* biofilms directly in human sputum samples. We have found that *M. abscessus* can form mono-species biofilms and exist in polymicrobial communities with an unidentified partner(s) *in situ*. We are utilizing transcript labeling via HCR to determine whether and to what degree the biofilm-relevant pathways identified in our *in vitro* work correlate with biofilm formation *in situ*. Ultimately, we hope to build tractable model systems in which infection-relevant biofilms can be recapitulated and probed in the laboratory.