Imaging and Monitoring the Stages of Biofilm Formation

Research Team: Shane Addleman, George Bonheyo, Erin Miller, Marvin Warner
Key Collaborators: Matthew Marshall, Robby Robinson, Kristyn Roscioli, Christine Mahoney, Zihua Zhu, Galya Orr, Tony Cinson, Mike Larche, Aaron Diaz

Pacific Northwest NATIONAL LABORATORY
Proudly Operated by Battelle Since 1965

Project 1.11; March 2013 – September 2015

Purpose

Fundamental Enabling Science

- Develop and apply analytical methods to characterize the molecular and cellular processes that enable cell adhesion and colonization, and ultimately lead to biofilm formation
- Examine biofilms and biofouling processes from medical and marine environments to understand and compare the basic mechanisms for each type

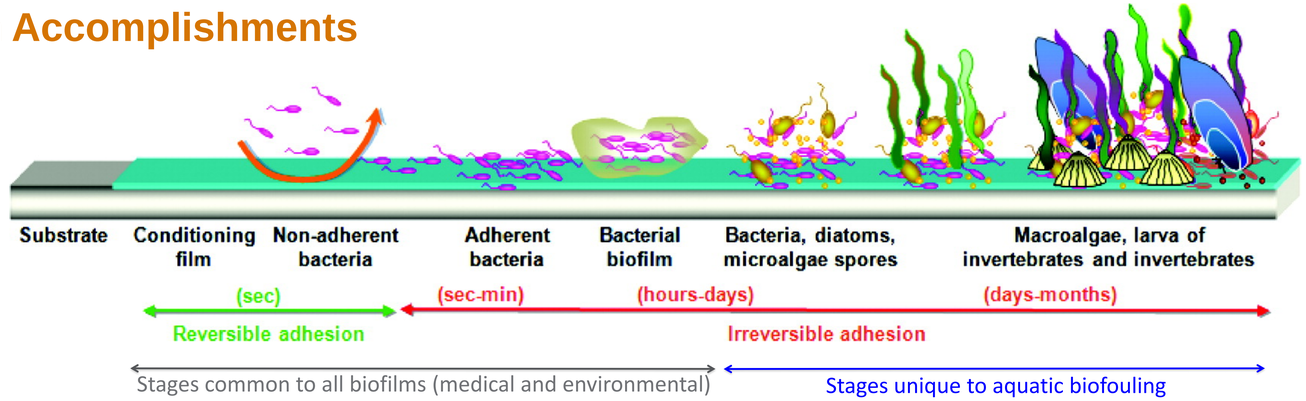
Applied Science

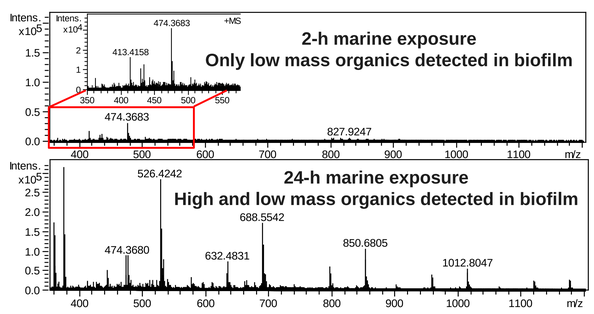
- New analytical tools will enable effective measurement of biofilms, which will support the rational development of better antifouling materials

Expected Outcomes and Impact

- Improved and integrated analytical methods to study biomolecular processes at fluid-surface interfaces
- Identify how composition and distribution of conditioning film impacts cellular adhesion and colony formation
- Understand how surface features impact sequential and spatial deposition of biomolecules and initial cellular growth processes
- Improved mechanistic understanding of how surface chemistry and features impact biofilm formation to enable development of new antifouling materials/solutions
- Better antifouling surfaces to improve shipping fuel efficiency, reduce transport of invasive species, and reduce infections from medical devices

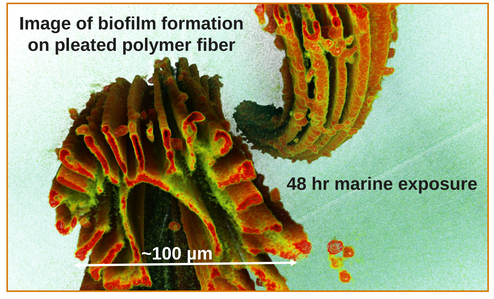
Research Accomplishments





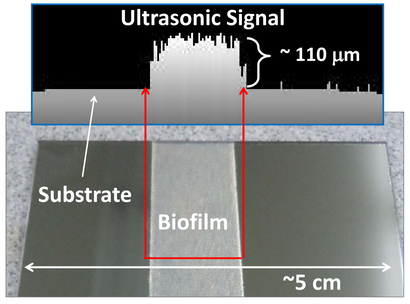
MALDI-TOF

Horizontal resolution ~5 µm (stage dependent)
Spatial chemical analysis of biologically active surfaces with QTOF and moderate sample prep



X-ray Microtomography

Resolution ~1.4 µm
Excellent structural imaging; extensive prep



Ultrasonic Imaging

Resolution ~4 µm vertical, ~20 µm horizontal;
In situ nondestructive monitoring; no sample prep

Other Methods to Be Evaluated

- TOF-SIMS
- AFM-Raman
- Nano-DESI
- E-STEM
- X-ray and electron spectroscopy
  - Energy-dispersive X-ray spectroscopy, Auger, and X-ray photoelectron spectroscopy
- Optical imaging (STORM, SIMS)

Next Steps

- Demonstrate methods and publish results
- Apply methods to the mechanistic study of antifouling materials
- Write proposals for follow-on funding to Office of Naval Research, National Institutes of Health, and Special Programs clients

PNNL-SA-96656