

Microbial Cell Dynamics Laboratory

A Hub of Systems Biology Research

Systems biology research represents a holistic approach toward understanding microbial physiology and the interaction and response of microorganisms to environmental conditions. Advanced analytical technologies (e.g., microarrays, mass spectrometer-based proteomics and metabolomic profiling) are incredibly powerful research tools that are ideally suited to systems microbiology research. However, the full potential and integration of these technologies will be realized only when they are applied to samples that are reproducibly generated.

In the Microbial Cell Dynamics Laboratory (MCDL) at Pacific Northwest National Laboratory, researchers not only develop and apply highly monitored and controlled cultivation technologies that provide quality samples for downstream analyses, but also design and conduct experiments to probe the molecular dynamics of microbes—research that is relevant to DOE missions (environmental remediation, alternative energy production and carbon sequestration).

Microbes are diverse, both genetically and in terms of metabolism. They have evolved for 4 billion years and have spread to virtually every environment and corner of the planet. Microbial communities can thrive in extremes of temperature, pressure, salinity and ionizing radiation. Surviving those extremes required microorganisms to evolve to meet specific environmental challenges. The resulting enzymes and pathways in known and as-yet-undiscovered organisms could provide revolutionary answers to today's problems.



The MCDL allows careful control and manipulation of microbial growth conditions, including extreme temperature, pH, radiation, pressure and other unusual environmentally relevant conditions. The lab houses flexible experimental systems capable of making multiplexed measurements of cellular responses and processes under these conditions. Samples can be analyzed with a suite of state-of-the-art microscopic and spectroscopic instruments.

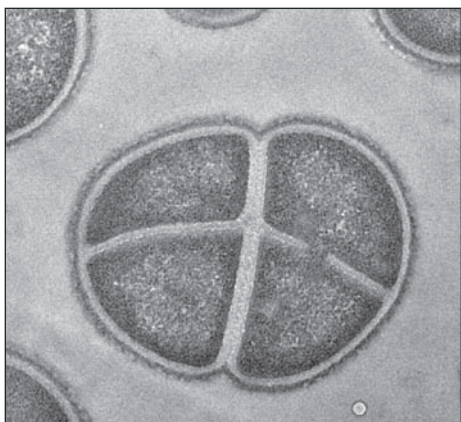
The full power of RNA, protein, and metabolite profiling and associated computational technologies is realized by using bioreactors. Bioreactors operate in a continuous mode, exactly reproduce culture conditions over multiple experiments and obtain cell populations with a minimal level of biochemical variability.

The Microbial Cell Dynamics Laboratory strengthens the link between environmental conditions, laboratory culturing, post-genomic molecular technologies and biological modeling and simulation.

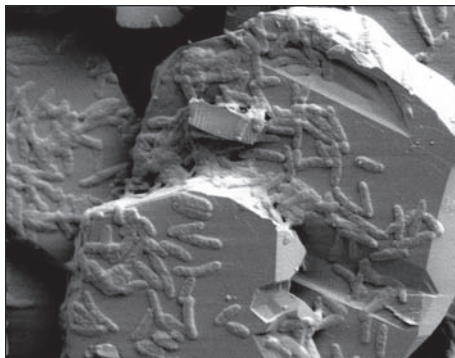
Key Capabilities

By coupling experience and expertise in environmental microbiology with capabilities in molecular and computational sciences, PNNL's systems biology program is developing capabilities for investigating the function and structure of biological macromolecules, microbial cells, mixed populations of microbial cells and microbial communities. The MCDL's core capabilities support both single-organism and microbial community studies. These include:

- Dedicated co-located laboratories and analytical stations, including mobile culturing equipment and analytical instruments
- Small reactor-scale (1-50 liter) culturing of prokaryotic cells under equilibrium conditions for generating cell populations with a minimum level of biochemical variability
- Systems for growth and analysis of planktonic cells and cells associated with surfaces or residing in biofilms
- Systems for controlling cell-cell interaction distance and rates of substrate diffusion to probe cell-signaling events



A transmission electron micrograph shows *Deinococcus radiodurans*, a bacterium that is resistant to extreme levels of ionizing radiation and to desiccation. Scientists at PNNL have isolated several strains of the bacterium from sediments collected from under a nuclear waste storage tank at DOE's Hanford Site.



A scanning electron micrograph shows *Shewanella putrefaciens* CN32 cells on the surface of hematite particles. CN32 is a metal-reducing bacterium important for cycling carbon and metals in the environment. It has potential applications for bioremediating metals and radionuclides.

- Rapid harvesting of cultures and processing and delivering cells and components to multiple analytical instruments with minimal composition alteration
- Real-time analysis of in situ biological, chemical and physical processes and parameters through automated liquid and headspace analysis
- Analysis of gene expression and signaling in individual cells and bulk populations.

Applications

Microbial cell research has diverse, far-reaching applications in areas of benefit to DOE and the nation.

Toxic waste cleanup

- In situ stabilization of metal and radio nuclide contaminants
- Biodegradation of organic contaminants
- Denitrification

Developing renewable energy sources

- H₂ from photosynthetic bacteria
- Improve conversion efficiency of plant biomass to energy
- Improve energy efficiency of chemical production

Global climate change and carbon sequestration

- Microbial CO₂ fixation
- Carbon stabilization in the geosphere
- Methanogenesis and methane oxidation

Public health and animal/crop protection

- Biofilm associated infections
- Detection of biological agents

Learning from *S. oneidensis*

Major studies are under way within the MCDL on *Shewanella oneidensis*, a metal-reducing bacterium. It can reduce mobile forms of contaminants such as uranium, technetium and chromate to immobile forms in the subsurface, ultimately preventing their migration in groundwater. *S. oneidensis* and related microbes are of global importance in the cycling of iron, manganese and carbon in the environment.

Scientists want to understand the molecular underpinnings of *S. oneidensis*—how it senses its chemical, physical and biological environment and responds to various changes in these parameters. Once scientists understand the organism and develop effective approaches and technologies, they can extrapolate these approaches to other microbes and to microbial consortia and communities.

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