
2010 Annual Report:

Biofilm concepts realized

The Center for Biofilm Engineering: 1990–2010
Montana State University College of
Engineering

CONCEPT

“The long-term goal of the Research Program is to institute a national resource to satisfy the entrepreneurial needs of a U.S. industry related to interfacial microbial processes.” –from the Center’s Year One report

“Montana State University was awarded today its largest grant ever—\$7.5 million from the National Science Foundation to help establish a one-of-a-kind engineering research center in Bozeman.” –Bozeman Daily Chronicle, Monday, January 29, 1990

FROM THE DIRECTOR: Phil Stewart

This year marks the 20th anniversary of our founding as a major center in the biofilm field. It was in the early spring of 1990 that the National Science Foundation (NSF) launched the Center for Biofilm Engineering with an Engineering Research Center award. I am proud to say that we still actively engage in all three pursuits that the NSF initially charged us with: interdisciplinary cutting-edge research, innovative education, and technology transfer. You can see for yourself, in the pages that follow, that we have been busy in these areas.

The bar chart running alongside this message makes the CBE’s impact visible. The length of each red bar is the total number of papers indexing to the keyword ‘biofilm’ from the ISI Web of Science database for each U.S. state, divided by the population of that state. Crossing the CBE—world’s largest and oldest biofilm center—with a sparsely populated state sure makes Montana stand out!

How did an international center of excellence sprout in the wide open space of the American West? It was through the prescient vision of our founder, Bill Characklis, and by a culture of creativity, teamwork, dedication, inclusiveness, and industrial interaction. On behalf of the current CBE participants I express our gratitude to all of the CBE alumni—students, staff, and faculty—whose hard work over two decades helped build the Center that we enjoy today.

Our 20th anniversary is a perfect occasion to revisit some of the foundational concepts in the biofilm field—the CBE team has made important contributions to the science and dissemination of each of these concepts over the years. In this year’s report, alongside highlights of our activities of the past year, we are pleased to bring you a selection of essential concepts and phenomena that underpin the special properties of microorganisms in biofilms. More discoveries await us, and as new concepts of biofilm function are realized, we look forward to bringing them to you.

RESEARCH

Center Research Areas

1990 (from the Center's Year One report)

Environmental biotechnology
Bioremediation

2010

Biofilm control strategies
Bioremediation
Environmental biotechnology
Energy solutions
Healthcare and medical biofilms
Industrial systems & processes
Standardized methods
Water systems

OVERVIEW

Major grants this year from the National Science Foundation and U.S. Department of Energy reinforced emerging strength at the CBE in sustainable energy solutions. The two NSF grants focus on fungal biofuels and computational modeling of biofilm mediated mineral deposition, respectively. The DOE award continues our work to apply bacterial biomineralization as a means to enhance geologic sequestration of carbon dioxide. Industrially sponsored research projects were an important part of our research portfolio, with 40 projects funded by 30 companies. Application areas of these projects range from medical devices to oilfield corrosion and water purification to microbes on textiles.

PUBLICATIONS

Sponsored research at the CBE helps move products to market. One example is research that was featured in the past year on the cover of the *Journal of Clinical and Translational Science*.

Journal of Clinical and Translational Science, Volume 2, Issue 3, June, 2009; The Society for Clinical and Translational Science and The Association for Patient-Oriented Research; Wiley-Blackwell.

CBE researchers authored 44 publications in the reporting period, including book chapters as well as articles in these 29 peer-reviewed journals:

American Biology Teacher
Anti-Infective Drug Discovery
Antimicrobial Agents and Chemotherapy
Applied and Environmental Microbiology
Bioelectromagnetics
BMC Systems Biology
Chemosphere
Desalination
Ecological Engineering
Environmental Science and Technology

Environmental Toxicology and Chemistry
FEMS Microbiology Letters
Gene
IEEE Transactions on Information Theory
Journal of Applied Microbiology
Journal of AOAC International
Journal of Bacteriology
Journal of Basic Microbiology
Journal of Membrane Science
Journal of Microbiology
Journal of Otolaryngology–Head and Neck Surgery
Microbial Ecology
Microbiology
Nature ISME Journal
Nucleic Acids Research
Proceedings of the National Academy of Sciences
Water, Air, and Soil Pollution
Wound Repair and Regeneration
Wounds

CONCEPT

“The Center. . . focuses on interfacial phenomena in ENVIRONMENTAL BIOTECHNOLOGY.” –from the Center’s Year One report

RESEARCH HIGHLIGHT

From the thermal pools of Yellowstone to the forests of Patagonia, the study of environmental biofilms at the CBE continues to provide new ideas for industrial solutions. Two examples are highlighted here.

Yellowstone National Park is a veritable hotbed of microbial discoveries right in MSU’s back yard. Brent Peyton, a professor in MSU’s Department of Chemical and Biological Engineering, has worked in the Heart Lake area of the park for the past five summers, collecting samples and recording water temperatures and pH levels in out-of-the-way geothermal pools. Peyton worked alone for the first two years, originally looking for heat-loving organisms that could break down explosives, as well as others that could be used in a biofuel production process.

Recently Peyton has been joined by several CBE graduate students and two other MSU faculty members—Matthew Fields, an assistant professor in the Department of Microbiology and the CBE, and Robin Gerlach, associate professor in the Department of Chemical and Biological Engineering.

Fields wants to identify organisms that have a particular function, such as producing fuel or converting chemicals into other useful products. “If we can I.D. a microorganism that will work in those extremes,” Fields said, “it might be something you can use in an industrial project or biotechnology.”

Montana State University professors are taking the next step in research that could make it possible to produce biofuel from wood chips using a fungus discovered in South America.

In the fall of 2009, MSU received a four-year, \$2 million grant from the National Science Foundation to allow faculty members at MSU and collaborators at Yale University to conduct a detailed study of the fungus *Ascooryne sarcoides*, which naturally produces gases that contain many of the same hydrocarbon compounds found in petroleum-based diesel fuel.

Brent Peyton, professor of chemical and biological engineering and the grant's principal investigator; Gary Strobel, professor of plant sciences; and Ross Carlson, assistant professor of chemical and biological engineering, will coordinate their research with Yale professors who are mapping the fungus's genes to learn just how it produces its hydrocarbon-rich vapors. MSU researchers will use that genetic information to experiment with the fungus's growing conditions, attempting to optimize the growth of *A. sarcoides* and get it to produce hydrocarbons as fast as possible.

EDUCATION

CONCEPT

"The Center will create an integrated program of interdisciplinary research, course work, and projects which offer an applied systems approach to industrial problems." –from the Center's Year One report

Summary of graduate students: 2009-10

44 graduate students from 9 disciplines:

- Biochemistry
- Cell Biology & Neuroscience
- Chemical & Biological Engineering
- Civil/Environ Engineering
- Health Services
- Land Resources & Environ Sciences
- Mathematics
- Mechanical & Industrial Engineering
- Microbiology

24 female / 20 male

33 PhD / 11 Masters

GRADUATE HIGHLIGHT

Stronger relationships, better water

CBE researchers are demonstrating that collaborative work can link laboratories and communities to produce relevant results.

Two CBE doctoral researchers in microbiology are investigating Montana water quality issues with the cooperation of community residents. Mari Eggers and Crystal Richards—both EPA STAR Fellowship grant awardees under the direction of Anne Camper, professor of civil engineering—work with residents collecting data in their communities to identify potential water quality problems in rural Montana water supplies.

Richards targets three known human pathogens: *Helicobacter pylori* (associated with stomach ulcers), *Legionella pneumophila* (which causes a severe respiratory ailment), and *Mycobacterium avium* (implicated in respiratory infections in immunocompromised people). *M. avium* was the only pathogen detected in Gallatin County (33% positive samples), while all three pathogens were found in Big Horn County: *H. pylori*—14% positive samples, *L. pneumophila*—41% positive samples, and *M. avium*—50% positive samples.

While Richards studies pathogens, Eggers performs complementary research on chemical exposures. She has been working with the dedicated members of the Crow Environmental Health Steering Committee, Tribal member and MSU graduate Crescentia Cummins, and a dozen Tribal College science majors over the past six years to carry out a community-based risk assessment of exposure to chemical and bacterial contaminants in water on the Crow Reservation in south central Montana. Their data have identified a number of concerns, including pollution in three rivers and bacterial and inorganic contamination of wells on the reservation. The data are being used by the community to seek funding to upgrade the water and wastewater treatment systems.

Richards credits Eggers' work with tribal community representatives for facilitating her own research experience. Said Richards, "I have met many friends along the way and have learned a lot about conducting research in a community in a culturally appropriate way."

Summary of undergraduate students: 2009-10

34 undergraduate students (12 female/22 male) from 9 disciplines:

- Biochemistry
- Cell Biology & Neuroscience
- Chemical & Biological Engineering
- Environmental Sciences (LRES)
- Physics
- Mechanical & Industrial Engineering
- Mechanical Engineering Technology
- Microbiology
- Nursing (Bridges program)

UNDERGRADUATE HIGHLIGHT

More interdisciplinary collaboration, better design solutions

The CBE goes to great lengths to emphasize the importance of collaborative research, but two senior undergraduates from opposite sides of the globe recently demonstrated how far they were willing to go to get successful results. Mechanical Engineering Technology (MET) majors Salman Adam, from Pakistan via Dubai, UAE, and Steven Anderson, of Gardiner, MT, recently combined their efforts in an internship with the CBE—and in the process gained valuable experience in teamwork, communication skills, and tool design skills. Their MET capstone project involved developing a set of testing tools, including a tweezer-like biofilm coupon holder that can be manufactured and sold by Biosurface Technologies (BST), a Bozeman-based company owned by Bryan Warwood. The project was supported by a grant from the Montana Board of Research and Commercialization Technology.

Adam and Anderson quickly discovered that their first challenge was to understand what biofilm is and how scientists test biofilms in a laboratory setting. Their goal was to design and construct a prototype tool set to make it easier and more efficient for laboratory technicians to perform the ASTM standard method for biofilm sampling. Helping them in the lab were members of the CBE's Standardized Biofilm Methods Laboratory: Darla Goeres, Diane Walker, Lindsey Lorenz, and Kelli Buckingham-Meyer. Their project advisor was MET instructor Kevin Cook. The development process required over 30 iterations in tool design before landing on the best option.

When asked what they learned by doing the project, Steven responded, "It was a very good interdisciplinary learning experience working with the CBE. Typically you would not see a mechanical engineer working in a microbiology lab, but understanding the laboratory procedure for the standard method was essential to the design process." To which Salman happily added, "This week when I appeared for the FE (Fundamentals of Engineering) Exam, I aced the biology section of the test!"

INDUSTRY

CONCEPT

"The Center will promote strong interaction among industry, university, and government agencies through information exchange activities such as publications, seminars, workshops, short courses, personnel exchanges, and review meetings."

—from the Center's Year One report

INDUSTRIAL HIGHLIGHTS

In 1990 the Center's list of Industrial Associates numbered 17 companies, the majority of them big names in the oil business. Over the past 20 years, the CBE's membership has grown and diversified significantly. In FY 2010, the Industrial Associates numbered 33 subscribing members (28 full members and 5 small business members) distributed among a variety of interests: consumer products, specialty chemicals, medical products and processes, testing laboratories, and government entities (*e.g.*, NASA and Sandia National Laboratories). As in the past several years, the fastest growing industry segment continues to be healthcare and biomedical companies. Recently, the energy companies have demonstrated a re-kindled interest in biofilm research. Annual membership fees provide a significant source of support for research continuity that keeps the CBE functioning as a true Center. In FY 2010, the CBE conducted 40 industry sponsored testing and research projects for 30 different companies with a total budget of over \$900,000.

Industrial Associates 2009–10

3M
Alcon Research
BASF
Bausch & Lomb
Baxter Healthcare
Bayer MaterialScience
BD Medical
Bridge Preclinical Testing Services
CareFusion
Church & Dwight
Colgate-Palmolive
Covidien
Dow Microbial Control
Embro Corporation
Ethox International
Glanbia Nutritionals
GlaxoSmithKline

ICU Medical
Johnson & Johnson
Kane Biotech
Kimberly-Clark
Masco Corporation
Mölnlycke Health Care AB
NASA
Novozymes A/S
Procter & Gamble
Quiescence Technologies
Sandia National Laboratories
Semprus BioSciences
Sherwin-Williams
Unilever
W.L. Gore & Associates
Whirlpool

REGULATORY OUTREACH UPDATE

EPA

An EPA-funded multiyear project with the CBE's Standardized Biofilm Method Laboratory (SBML) supports the development of a standard method for testing the efficacy of liquid disinfectants against biofilm. The method under development uses the CDC biofilm reactor to grow a *Pseudomonas aeruginosa* biofilm. The proposed efficacy method has gone through an initial two-lab collaborative study, and the SBML continues to work with the Microbiology Laboratory Branch of the EPA's Office of Pesticide Programs: Biological & Economic Analysis Division (BEAD) to refine and validate the method.

FDA

CBE staff (Drs. Phil Stewart, Garth James and Paul Sturman) delivered a workshop titled "Biofilms in Medicine" at the FDA's Silver Spring, MD, campus on April 6, 2010. The workshop was attended by over 30 FDA scientists and decision makers and received very high marks for quality and usefulness. The workshop covered current basic and applied work in oral biofilms, wound biofilms, implant-related biofilms and the role of biofilms in healthcare associated infections. The objective of this workshop was to assist the FDA in addressing the biofilm-related data gaps identified in the recently published Action Plan to Prevent Healthcare-Associated Infections (U.S. Department of Health and Human Services, 2009) and to establish the CBE as a resource for future biofilm-related questions.

MONTANA BIOFILM MEETINGS

The Montana Biofilm Science & Technology Meeting (MBM) continues to be a showcase for CBE research, but has also increasingly become a highly sought venue for

the presentation of industrially relevant work from outside the CBE. Over the past several years, we have increased the presence of high caliber outside speakers, seeking to develop the MBM into the single most useful biofilm-centered meeting available to our member company representatives. Record attendance at the February 2010 meeting suggests that our efforts have been successful. The MBM has become a forum not just for learning the latest in biofilm research, but also a premiere venue for networking, problem solving, and accessing biofilm technology.

The CBE recently initiated a program to include outstanding young biofilm researchers from around the world in our industrial meetings. Starting with the February 2010 meeting, the Young Investigator Program offers a \$750 travel reimbursement to two researchers for each meeting. In its initial announcement, 15 abstracts were received and travel awards were made to allow these relatively new biofilm researchers the opportunity to present their work to our industrial audience.

Young Investigator Inaugural Year Awardees

Gregory Anderson, Indiana University-Purdue University, Indianapolis
Joao Xavier, Sloan Kettering Institute, NY
Ge Alice Zhao, University of Washington, Seattle
Anne Han, Johns Hopkins Medical Institutions, Baltimore, MD

OUTREACH

CONCEPT

"The Center must maintain a leadership role in environmental biotechnology, which requires persistent interaction with researchers around the world as well as providing a forum for conveying research products in a timely manner to industry and the research community at large." –from the Center's Year One report

REACHING THE WORLD

The Center's initial proposal (1989) and Year One report outlined its intent to spread the biofilm news via symposia, workshops, short courses, publications—and an "electronic bulletin board." Little did any of us realize how quickly the idea of electronic information dissemination would catch on, in the form of the World Wide Web.

In 2010 the CBE rolled out a new version of its popular web site that includes all of the features and content previously available, as well as plenty of new material,

within a more adaptable, dynamic content management system.

Development also continues on the first interactive, electronic biofilm textbook—*BIOFILMS: The Hypertextbook*—an initiative that was funded by the National Science Foundation.

“Outstanding job; you guys are the best in the world at advancing biofilm knowledge and appreciation. My students at all levels know you well from illustrations that I have used in classroom and lab presentations. My congratulations to you for this splendid effort.”

– Recent unsolicited comment about the CBE web from a professor at the University of Massachusetts

VISITING RESEARCHERS

Anozie Ebigbo, graduate student,
University of Stuttgart, Germany

Mariana Fittipaldi, graduate student,
Laboratorio de Microbiologia Sanitaria y
Medioambiental, Universitat Politècnica de Catalunya,
Barcelona, Spain

Marion Fontagneu, graduate student,
University de Pau, France

Anna Heinkel, undergraduate,
University of Duisburg-Essen, Germany

Mijeong Jang, postdoctoral researcher, Seoul, Korea

Danielle Kinsey, undergraduate,
Fort Belknap College, MT

Emma Mean, undergraduate,
Notre Dame University, IN

Maria Nikolopoulou, graduate student,
Technical University of Crete, Greece

Patricia Peters, undergraduate,
University of Duisburg-Essen, Germany

Esther Rosenbrand, graduate student,
University of Stuttgart, Germany

Mary Schweitzer, faculty, and **Elizabeth Johnson**,
postdoctoral researcher, North Carolina State University,
NC

Federica Villa, graduate student,
University of Milan, Italy

Yi Wang, faculty,
Xi’an University of Architecture & Technology, China

Kenichi Yoshida, administration,
Ibaraki Prefecture, Japan

PEOPLE

Nancy Characklis presented the 2010 W.G. Characklis Outstanding Student Award to two CBE doctoral students. Sabrina Behnke, doctoral student in microbiology, was awarded in recognition of her successful pursuit of industrial funding for her thesis project, facilitation of the participation of multiple international visiting students at CBE, contributions to revitalizing the Center’s TGIF social hour, and consistent and enthusiastic volunteer assistance at Montana Biofilm Meetings and workshops. Erin Field, also a doctoral student in microbiology, was recognized for her successful interdisciplinary research interactions, quality instruction in the CBE “Microbes in the Environment” course, contributions to a collaborative proposal with an industrial associate, contributions to organization of the CBE seminar series, and enthusiastic participation in Montana Biofilm Meetings.

Erin Field, doctoral student in microbiology, also received the Ferguson Graduate Student Fellowship Award, given by the Department of Microbiology for her outstanding contributions both in research and in teaching.

Heidi Smith, doctoral student in Land Resources and Environmental Sciences (LRES), received a competitive NASA Earth and Space Science Fellowship (NESSF) for 2010/2011, with the possibility for two additional years of renewal support (\$30K each year). Heidi’s project is “The role of microbes in microbial synthesis and transformation of dissolved organic matter in glacial environments.” Heidi’s advisor and mentor is Christine Foreman, associate research professor in LRES.

Senior in chemical and biological engineering Trevor Zuroff was awarded a graduate research fellowship from the National Science Foundation. He will use the \$90,000 three-year graduate research award to investigate using different organisms to break down cellulose. He begins working on his doctorate in the fall at Penn State, where he was also awarded a McWhirter fellowship granting him \$90,000 for two more years after the NSF grant expires, plus \$4,000 in research funds and funding to hire an undergraduate assistant.

ESSENTIAL BIOFILM CONCEPTS AND PHENOMENA

ADHESION

Bacteria preferentially attach to surfaces when favorable environmental conditions are available. Microbes stick to surfaces. They will stick to plastic, glass, or metal—as well as plant or animal tissues. Adhesion is the initial step of biofilm formation that allows microbes to associate with each other and to establish residence in a particular environment.

SELECTED PUBLICATIONS

1. Camper AK, Hayes JT, Sturman PJ, Jones WL, Cunningham AB, “Effects of motility and adsorption rate coefficient on transport of bacteria through saturated porous media,” *Appl Environ Microbiol*, 1993; 59(10):3455–3462.
2. Hall-Stoodley L, Costerton JW, Stoodley P, “Bacterial biofilms: From the environment to infectious disease,” *Nat Rev Microbiol*, 2004; 2(2):95–108.

EXTRACELLULAR MATRIX

Microbes in a biofilm secrete extracellular polymeric substances (EPS) that form a sticky, hydrated gel that holds the biofilm together. EPS constituents include polysaccharides, proteins, and extracellular DNA.

The formation and development of a biofilm occurs in a predictable manner. Initially the attachment of a bacterium to a surface is tenuous and reversible. Within a matter of minutes, the bacterium becomes irreversibly attached and begins to secrete the anchoring EPS.

As bacteria begin to multiply, EPS holds cells in close proximity, which is necessary for cell-cell communication. It also allows the formation of three-dimensional structures that give the bacteria increased access to nutrients and the advantages of multicellular living.

SELECTED PUBLICATIONS

3. Clark ME, Edelmann RE, Duley ML, Wall JD, Fields MW, “Biofilm formation in *Desulfovibrio vulgaris* Hildenborough is dependent upon protein filaments,” *Environ Microbiol*, 2007; 9(11):2844–2854.
4. Hornemann JA, Lysova AA, Codd SL, Seymour JD, Busse SC, Stewart PS, Brown JR, “Biopolymer and water dynamics in microbial biofilm extracellular polymeric substance,” *Biomacromolecules*, 2008; 9(9):2322–2328.

SPECIES DIVERSITY

An astonishing diversity of microbial species often coexist in real-world biofilms. Hundreds of phylogenetically and metabolically distinct species have been found in environments ranging from the human mouth (dental plaque) to hot springs in Yellowstone National Park.

SELECTED PUBLICATIONS

5. James GA, Swogger E, Wolcott R, deLancey Pulcini E, Secor P, Sestrich J, Costerton JW, Stewart PS, “Biofilms in chronic wounds,” *Wound Repair and Regeneration*, 2008; 16(1):37–44.

6. Taffs R, Aston JE, Brileya K, Jay Z, Klatt CG, McGlynn S, Mallette N, Montross S, Gerlach R, Inskip WP, Ward DM, Carlson RP, “In silico approaches to study mass and energy flows in microbial consortia: A syntrophic case study,” *BMC Systems Biology*, 2009; 3:114.

OXYGEN GRADIENTS

Mature biofilms are criss-crossed by gradients in the concentrations of metabolic substrates and products. Oxygen is the foremost example. Local depletion of oxygen allows anaerobic bacteria to thrive in the depths of a biofilm bathed in aerated water.

SELECTED PUBLICATIONS

7. de Beer D, Stoodley P, Roe F, Lewandowski Z, “Effects of biofilm structures on oxygen distribution and mass transport,” *Biotechnol Bioeng*, 1994; 43(11):1131–1138.
8. Xu KD, Stewart PS, Xia F, Huang C-T, McFeters GA, “Spatial physiological heterogeneity in *Pseudomonas aeruginosa* biofilm is determined by oxygen availability,” *Appl Environ Microbiol*, 1998; 64(10):4035–4039.

PHENOTYPIC HETEROGENEITY AND DIFFERENTIATION

Cells of a given species can occupy a wide variety of phenotypic states in the same biofilm, from rapidly growing to dormant to expressing a unique activity. Mechanisms of diversification include nutrient gradients, mutation and natural selection, and genetic regulatory switches and signaling pathways.

SELECTED PUBLICATIONS

9. Sauer K, Camper AK, Ehrlich GD, Costerton JW, Davies DG, “*Pseudomonas aeruginosa* displays multiple phenotypes during development as a biofilm,” *J Bacteriol*, 2002; 184(4):1140–1154.
10. Stewart PS, Franklin MJ, “Physiological heterogeneity in biofilms,” *Nat Rev Microbiol*, 2008; 6(3):199–210.

DIFFUSION LIMITATION

Diffusion is the predominant solute transport process within biofilm cell clusters and is often rate-limiting. Many biofilm phenomena (e.g., oxygen gradients, antimicrobial tolerance, species diversity) can be explained in part by reaction-diffusion interactions.

SELECTED PUBLICATIONS

11. Costerton JW, Lewandowski Z, de Beer D, Caldwell D, Korber D, James G, “Biofilms, the customized microniche,” *J Bacteriol*, 1994; 176(8):2137–2142.
12. Stewart PS, “Diffusion in biofilms,” *J Bacteriol*, 2003; 185(5):1485–1491.

ANTIMICROBIAL TOLERANCE

Microbes in biofilms are hard to kill with biocides or antibiotics. Protective mechanisms include poor penetration of reactive agents, non-growing cells whose inactivity makes them less vulnerable, and implementation of adaptive responses.

SELECTED PUBLICATIONS

13. de Beer D, Srinivasan R, Stewart PS, "Direct measurement of chlorine penetration into biofilms during disinfection," *Appl and Environ Microbiol*, 1994; 60(12):4339–4344.
14. Stewart PS, Costerton JW, "Antibiotic resistance of bacteria in biofilms," *Lancet*, 2001; 358(9276):135–138.

GENETIC & BIOCHEMICAL BASES

The activities and interactions of microorganisms in biofilms have specific genetic and biochemical bases. Analyses of mutants, gene expression patterns, and quorum sensing pathways linked to conditions of an organism's microenvironment reveal complex spatial and temporal changes in the biological activities of microorganisms in a biofilm.

SELECTED PUBLICATIONS

15. Davies DG, Parsek MR, Pearson JP, Iglewski BH, Costerton JW, Greenberg EP, "The involvement of cell-to-cell signals in the development of a bacterial biofilm," *Science*, 1998; 280(5361):295–298.
16. Mah T-F, Pitts B, Pellock B, Walker GC, Stewart PS, O'Toole GA, "A genetic basis for *Pseudomonas aeruginosa* biofilm antibiotic resistance," *Nature*, 2003; 426(6964):306–310.

PERSISTENT INFECTIONS

Biofilms that form on implanted devices or damaged tissue establish slow-moving but difficult to resolve infections. Periodontitis, cystic fibrosis pneumonia, and catheter-associated infections are examples.

SELECTED PUBLICATIONS

17. Costerton JW, Stewart PS, Greenberg EP, "Bacterial biofilms: A common cause of persistent infections," *Science*, 1999; 284(5418):1318–1322.
18. Jesaitis AJ, Franklin MJ, Berglund D, Sasaki M, Lord CI, Bleazard J, Duffy JE, Beyenal H, Lewandowski Z, "Compromised host defense on *Pseudomonas aeruginosa* biofilms: Characterization of neutrophil and biofilm interactions," *J Immunol*, 2003; 171(8):4329–4339.

DETACHMENT & DISPERSAL

The release of biofilm-associated microbes into the fluid surrounding the biofilm is a natural phenomenon. It can occur by multiple pathways, from hydrodynamic shearing to concerted activation of motility and matrix degradation. In a mature biofilm, detachment is the primary process balancing growth by limiting biofilm accumulation.

SELECTED PUBLICATIONS

19. Peyton BM, Characklis WG, "A statistical analysis of the effect of substrate utilization and shear stress on the kinetics of biofilm detachment," *Biotech and Bioeng*, 1993; 41(7):728–735.
20. Sellam A, Al-Niemi T, McInerney K, Brumfield S, Nantel A, Suci P, "A *Candida albicans* early stage biofilm detachment event in rich medium," *BMC Microbiol*, 2009; 9(1):25.

VISCOELASTICITY

Biofilms stretch, deform, and oscillate when subjected to an applied force, such as flowing water. They exhibit a combination of elastic and viscous behavior that makes them resilient to physical challenges.

SELECTED PUBLICATIONS

21. Klapper I, Rupp CJ, Cargo R, Purevdorj B, Stoodley P, "A viscoelastic fluid description of bacterial biofilm material properties," *Biotech Bioeng*, 2002; 80(3):289–296.

HYDRODYNAMICS

Biofilms interact with the flowing fluid around them. In many industrial and household systems this results in fouling that reduces the performance of piping or equipment. Nuclear Magnetic Resonance imaging can help us visualize the interactions of biofilms and water in conduits.

SELECTED PUBLICATIONS

22. Cunningham AB, Characklis WG, Abedeen F, Crawford D, "Influence of biofilm accumulation on porous media hydrodynamics," *Environ Sci and Tech*, 1991; 25(7):1305–1311.
23. Seymour JD, Codd SL, Gjersing EL, Stewart PS, "Magnetic resonance microscopy of biofilm structure and impact on transport in a capillary bioreactor," *J Magnetic Resonance*, 2004; 167(2):322–327.

MINERAL TRANSFORMATIONS

The metabolic activity and extracellular matrix polymers of a biofilm can facilitate the precipitation or dissolution of a mineral phase. Algal biofilms raise pH and produce CO₂, sometimes resulting in deposition of calcium carbonate. Dental plaque lowers pH, which may result in dental caries.

SELECTED PUBLICATIONS

24. Sani RK, Peyton BM, Amonette JE, Geesey GG, "Reduction of uranium (VI) under sulfate-reducing conditions in the presence of Fe(III)-(hydr)oxides," *Geochimica et Cosmochimica Acta*, 2004; 68(12):2639–2648.
25. Mitchell AC, Dideriksen K, Spangler LH, Cunningham AB, Gerlach R, "Microbially enhanced carbon capture and storage by mineral-trapping and solubility-trapping," *Environ Sci Technol*, 2010; 44(13):5270–5276.

PHOTO CREDITS

Cover credits:

SEM image: K Kirker, MSU-CBE. Confocal scanning laser microscopy: S Sánchez and B Pitts, MSU-CBE (cover of *Chemical & Engineering News*, 86:23, June 9, 2008, ACS)

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Journal of Clinical and Translational Science, Volume 2, Issue 3, June, 2009; The Society for Clinical and Translational Science and The Association for Patient-Oriented Research; Wiley-Blackwell.

SEM micrograph at 9,000 X magnification of *Staphylococcus aureus* biofilm formation on an exposed titanium coupon. Imaged by Alessandra Marçal Agostinho, MSU-CBE. Related article: "Inhibition of *Staphylococcus aureus* biofilms by a novel antibacterial envelope for use with implantable cardiac devices," Agostinho A, James G, Wazni O, Citron M, Wilkoff BD. pp.193-198.

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Patagonia photos, above right, courtesy of G Strobel (pictured); photos of Yellowstone Park and PhD student Jacob Valenzuela courtesy of K Gorham, MSU News Services.

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SEM imaging of cells and extracellular filaments by ME Clark. TEM imaging provided courtesy of G Geesey. From: Costerton JW, Geesey GG. 1979. "Microbial contamination of surfaces," In: KL Mittal (ed.) *Surface Contamination*, 1979; Vol 1:211-221. Plenum Press, New York, NY. Used with permission of author.

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Photo courtesy of K Gorham, MSU News Services.

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Pyrosequencing and clone library imaging reveal genus diversity using color to designate different genera and bandwidth to indicate representation in the community sampled; data imaging provided by K De León.

Plot of microsensor-detected gradients of dissolved oxygen in a biofilm: D de Beer, P Stoodley, Z Lewandowski

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Photograph by C Abernathy; NMR imaging by E Gjersing

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Calcite crystal stereo microscope image, top, by L Schultz

Throughout report:

Biofilm conceptual graphics by P Dirckx.

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