

Biofilms and Biodiversity

An Interactive Exploration of Ecology and Environmental Biotechnology

Pre-visit Preparation

Welcome to the University of Maryland Biotechnology Institute's (UMBI) SciTech Education Program. We hope that these materials are useful and will help prepare your students for a unique and exciting "hands-on" lab experience. We encourage you to review these pre-visit materials. The teacher background sheets are designed to increase your understanding of this topic. Students will have a richer experience with us if you go over the pre-visit materials with your class before your visit. For more information regarding SciTech, visit our website: www.umbi.umd.edu/~scitech. We encourage you to make copies of the pre-visit information for students and visit the Maryland Sea Grant biofilms and biodiversity website <http://www.mdsq.umd.edu/Education/biofilm/index.htm> for more information about biofilms and an interactive lesson on biodiversity.

Summary of Student Experience

After a guided inquiry about biofilm communities and their formation with the SciTech staff, students will generate questions about the biofilms in Baltimore's Inner Harbor. Students will receive background and demonstrations on the use of water sampling equipment and the collection of biofilm discs. The SciTech staff will lead student groups to the pier and each group will test the water quality and collect water and biofilm samples for analysis in the SciTech Education Program Laboratory. Students will work with live biofilm communities to observe and understand their development, identify key invertebrate species, and investigate the impact of water quality on biofilm communities in the Inner Harbor. *Because students will be working with living organisms, we ask that they treat them with respect and follow the appropriate guidelines defined by our staff on the day of the trip.* Upon request, a Center of Marine Biotechnology (COMB) scientist will discuss his or her research, personal science career path and respond to student questions about possible careers in science.

Tips for a Successful SciTech Experience

The Biofilms and Biodiversity lab offers some unique opportunities for students and teachers. The Olympus America, Inc., microscope equipment is set-up so that you can videotape and photograph your favorite species and biofilm communities in the lab while the students are at work. To take advantage of this opportunity, please bring:

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A VHS videotape for recording,
A roll of 35 mm film, print or slide film, 400 or 800 speed, and
Some blank computer discs for digital pictures, Mac or PC format.

Experimental Design Background

Stating the Question



Every experiment begins with a question that the experiment will be designed to answer. Formulating this question is often the most difficult part of setting up a research project. There are many questions that could be investigated during the exploration of the biofilms, ranging from structure and function of organisms to their ecological niche. One question about biofilms could be “How does poor water quality affect biofilms?” Hopefully, many questions, including the one above, will be answered during the lab experience.

Hypothesis

The clearest way to write a hypothesis is to use “if...then” statements. For example: “If low oxygen conditions harm biofilm organisms, then biofilms in low oxygen will have fewer organisms or decreased diversity compared to biofilms in high oxygen.” The most common hypothesis is the *null hypothesis*, which simply states that the variable or experimental situation being tested will exhibit no significant difference from the controls. For example, one null hypothesis could state, “There will be no difference in the abundance or diversity of biofilm organisms between discs in high versus low oxygen conditions.”

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Controls

When designing an experiment, it is important to plan ahead so that the method you are testing is compared against a standard. To investigate how water quality affects biofilms we need to establish communities in Baltimore's Inner Harbor under good, moderate, and poor water quality conditions. In our experiment we place racks of biofilm discs at varying depths in the Inner Harbor, because different depths experience different water quality conditions.

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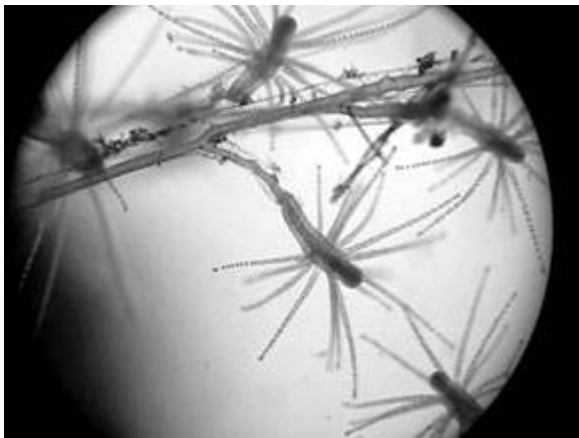


Biofilms and Biodiversity Background

What is That Stuff?

Have you ever run your tongue across your teeth in the morning? The slime on your teeth is a deposit (a sticky sugary substance) that bacteria have produced overnight. These bacteria, and the substances they produce, are affectionately known as biofilms. What other areas of your body, inside or outside, have biofilms? Are they beneficial or harmful?

What are Biofilm Communities?



Biofilms are a hot topic in microbiology today. Scientists are studying the ways bacterial colonies form these slimy layers, which can be resistant to antibiotics and the immune system, in hopes that new information will help us understand how the layers form, adhere to surfaces, and how they can be prevented. The biofilm time-line is a summary of important events linking biofilms and disease as described in Science, Vol 283, 19 March 1999.

Mid-1960s	Mid-1970s	1976	Mid-1980s
Dental researchers discover relationship between bacteria in the mouth and sticky plaque substances	Researcher Thomas Marrie of Nova Scotia reveals a biofilm layer on a heart pacemaker via electron microscopy	29 members of the American Legion were killed by infections from chunks of Legionella biofilm from air conditioners at a convention in Philadelphia, PA	Joseph Lam of Univ of Calgary confirmed that biofilms are present in the lungs of cystic fibrosis patients

Biofilms moved to the forefront of microbiology after a 1994 case involving the infection of hundreds of asthmatics. It was found that all the asthmatics used the same inhalant contaminated with a bacterium known as *Pseudomonas aeruginosa*. This bacterium was able to survive the routine disinfection of the inhalant during manufacturing by forming a biofilm comprised of many colonies. The contaminated inhalers contained pieces of the biofilm, which were transported directly to the lung tissue. In the lung tissue the *Pseudomonas* biofilm was able to flourish. One hundred people died from the biofilm infection, a dramatic example of the danger posed by some bacterial biofilms.

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Biofilms can be found in many areas of the human body and the environment. They are found on teeth, in intestines, on medical devices, on contact lenses, in drainage pipes, and on the bottoms of ships. The common denominator is that all of the biofilms are comprised of a primary layer of bacteria that create an attractive environment for other bacteria and larger organisms to live. Biofilms on the hull of a ship contain large organisms like barnacles and mussels. These biofilms slow ships and are expensive to remove and prevent. Current methods of preventing biofilm formation on ships include painting the hull with a wide variety of toxic marine paints. However, these paints wear off and sometimes the biofilms continue to form regardless of the toxins in the paint.

An Introduction To Biofilms & Biodiversity in Chesapeake Bay

The Chesapeake Bay is the largest, most productive estuary in the United States and provides a habitat for some 2,700 species of plants and animals. Good water quality is essential for all of these species to remain abundant.



Many of us have heard about or participated in water quality monitoring throughout the Chesapeake Bay region and we recognize the importance of making these measurements for understanding the health of Chesapeake Bay. Water quality can be directly related to the biodiversity within the Bay ecosystem. Studying the connection between water quality and biodiversity can help us understand how abiotic (nonliving)

factors impact the biotic (living) factors in an aquatic environment. One way to measure biodiversity is to examine the communities of biofilm invertebrates colonizing substrates such as acrylic discs.

In the lab, you will evaluate biofilms grown on acrylic discs that were suspended in the Baltimore Inner Harbor for various periods of time. These discs were suspended at three depths in the Inner Harbor – shallow water (0-1 m), middle level depth (2-3 m) and deep water (3-4 m). Each of these depths experiences distinct water quality conditions that we can monitor. The biofilm discs provide a fascinating look at Chesapeake Bay invertebrates and an introduction to what these creatures can tell us about the relationship between bacterial biofilms and invertebrate biofouling.

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How Do Biofilm Communities Develop in the Inner Harbor?



Colonization of biofilm organisms in the Inner Harbor area can be compared to colonization in an old abandoned farm field. In the field, colonization is first characterized by a great diversity of weeds, wildflowers, small seedlings, and grasses. In time, seedlings of small shrubs and trees begin to dominate. These plants will compete for space more efficiently and "weed out" their neighbors. After a few seasons, larger trees will dominate the landscape and the diversity will decline to a small number of well-

adapted species. This process is known as succession and occurs in many ecosystems. Succession begins rapidly on biofilm discs in the Inner Harbor, but for the first few days consists primarily of bacteria and organic molecules. After this rapid colonization, invertebrate animals begin to attach. Protozoa and small invertebrate larvae soon begin to compete for the limited space on the discs. Invertebrates such as mussels, barnacles, mud worms, and anemones gradually come to dominate the biofilm discs, and these animals limit the space available to other invertebrates. In Baltimore's Inner Harbor, this process takes about 2-3 months and is most rapid in spring and summer, while succession in the old farm field will take many years.



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Glossary

Abiotic - The physical and chemical non-living factors in an environment.

Annelids - A phylum that includes segmented terrestrial and aquatic worms.

Bacteria - Single celled prokaryotic organisms that form the first layer of a biofilm.

Biodiversity - The number of different species of organisms in a particular environment.

Biofilm - A coating or covering on the surface of a living or nonliving substrate composed of organisms like bacteria, protozoa, algae, and invertebrate animals.

Biotic - The living factors in an environment.

Cnidaria - An animal phylum that includes hydra, sea anemones, jellyfish, and hydrozoan colonies.

Crustacean - A class of arthropod with varying numbers of legs, antennae, and a hard exoskeleton.

Entoprocta - An animal phylum that includes organisms with tentacles on a cup shaped body supported by a single stalk.

Evenness (E) - A measure of how similar the abundance of different species are in the community.

Invertebrates – Animals without backbones.

Larvae - Immature forms of organisms, that typically look different from the fully-grown adult and are usually smaller than the adult or even microscopic. Many attached biofilm invertebrates have planktonic larvae.

Mollusks - An animal phylum that includes bivalves (such as mussels and clams), snails, slugs, squid, and nudibranchs.

Nematoda - An animal phylum that includes all roundworms.

Phytoplankton - Drifting microscopic plants that trap the energy from the sunlight and are primary organisms in a marine food chain.

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Platyhelminthes - A phylum of animal that includes all flatworms.

Protozoa - A Kingdom of single-celled organisms such as amoeba, stentor, vorticella, and colonial ciliates.

Rotifers - A phylum of animals with ciliated mouths and a retractable "foot" for anchoring.

Sessile Organisms - Organisms that remain attached to a substrate.

Species - Organisms that are genetically related, similar physically, and can reproduce viable offspring.

Species Richness - The number of different species found in a particular environment.

Succession – The process of community change over time.

Zooplankton - Microscopic aquatic organisms, including larvae, which are the first consumers in a marine food chain.

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Books/Articles References

Begon, Michael, John L. Harper and Colin R. Townsend. 1986. Ecology, Individuals, Populations and Communities. Sinauer Associates, Inc. Publishers, Sunderland, MA.

Lippson, Alice Jane and Robert L. Lippson. 1984. Life in the Chesapeake Bay. Johns Hopkins University Press, Baltimore, MD.

National Audubon Society. 1981. Field Guide to North American Seashore Creatures. Alfred A. Knopf, Inc., Publishers, New York.

Odum, Eugene P. 1983. Basic Ecology. Saunders College Publishing, New York.

Science. Volume 283, 19 March 1999. Forging a Link Between Biofilms and Disease.

Web References

Maryland Sea Grant, Interactive Marine Education Material
<http://www.mdsg.umd.edu/Education/biofilm/index.htm>

Microbial World:Biofilms
<http://helios.bto.ed.ac.uk/bto/microbes/biofilm.htm#crest>

Microbe Zoo
<http://commtechlab.msu.edu/sites/dlc-me/zoo/>

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Maryland State Department of Education Core Learning Goals

The following Core Learning Goals link directly to the Biofilms and Biodiversity Lab. Take a few moments to review the specific goals, expectations, and indicators below so that you may prepare your students appropriately in conjunction with the pre-visit materials that will be sent when you book your field trip. If you do not have a Core Learning Goals document or CD, talk to your department chair, contact your science supervisor or visit the website http://www.mdk12.org/mspp/high_school/what_will/index.html.

Core Learning Goal – Science

Goal 1 - Skills and Processes

Expectation 1.2 - The student will pose scientific questions and suggest investigative approaches to provide answers to questions.

Indicator 1.2.1 -

- The student will identify meaningful, answerable scientific questions.

Indicator 1.2.2 -

- The student will pose meaningful, answerable scientific questions.

Indicator 1.2.3 -

- The student will formulate a working hypothesis.

Indicator 1.2.4 -

- The student will test a working hypothesis.

Indicator 1.2.5 -

- The student will select appropriate instruments and materials to conduct an investigation.

Expectation 1.3 - The student will carry out scientific investigations effectively and employ the instruments, systems of measurement, and materials of science appropriately.

Indicator 1.3.4 -

- The student will learn the use of new instruments and equipment by following instructions in a manual or from oral direction.

Expectation 1.4 - The student will demonstrate that data analysis is a vital aspect of the process of scientific inquiry and communication.

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Indicator 1.4.9 -

- The student will use analyzed data to confirm, modify, or reject a hypothesis.

Expectation 1.5 - The student will use appropriate methods for communicating in writing and orally the processes and results of scientific investigation.

Indicator 1.5.1 -

- The student will demonstrate the ability to summarize data (measurements/observations).

Indicator 1.5.2 -

- The student will explain scientific concepts and processes through drawing, writing, and/or oral communication.

Expectation 1.7 - The student will show that connections exist both within the various fields of science and among science and other disciplines including mathematics, social studies, language arts, fine arts, and technology.

Indicator 1.7.5 -

- The student will investigate career possibilities in the various areas of science.

Goal 3 - Concepts of Biology

Expectation 3.2 - The student will demonstrate an understanding that all organisms are composed of cells which can function independently or as part of multicellular organisms.

Indicator 3.2.2 -

- The student will conclude that cells exist within a narrow range of environmental conditions and changes to that environment, either naturally occurring or induced, may cause changes in the metabolic activity of the cell or organism.

Expectation 3.5 - The student will investigate the interdependence of diverse living organisms and their interactions with the components of the biosphere.

Indicator 3.5.3 -

- The student will investigate how natural and man-made changes in environmental conditions will affect individual organisms and the dynamics of populations.

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