

Biofilms:

Stuck Like Glue, Stuck On You

Lesson #	Lesson Title	Suggested Time
1	What's On Our Teeth?	2-3 class periods
2	What is a Biofilm?	2 class periods
3	Biofilms Are Everywhere!	1 class period
4	Why Do Bacteria Form Biofilms?	1-2 class periods
5	Are All Biofilms Bad?	2 class periods

Biofilms- Stuck Like Glue, Stuck On You- Grades 6-8 (Life Science)

Students explore a variety of phenomena caused by bacteria living on moist surfaces. The most common biofilm we encounter on a daily basis, and the unit phenomenon, is dental plaque. Students begin by investigating animal and plant cells, and modeling the similarities and differences between them. Bacterial cells are then introduced, before students explore the parts of a biofilm, as well as the important steps in development of dental plaque. The unit then introduces biofilms found in a range of environments, and challenges students to sort bacterial species by which biofilm is most likely to satisfy their genetically encoded needs. Next, students design and carry out an experiment to determine why biofilms are so hard to destroy. Finally, students explore examples of “good” biofilms and ways in which biofilms are being used by scientists and engineers to help solve problems. Then, students collaborate in an engineering design proposal aimed at combating a real-world biofilm-associated concern.

Biofilms- Stuck Like Glue, Stuck On You

The phenomenon of dental plaque drives student exploration in this five-lesson introduction to biofilms. In the first lesson, students investigate the similarities and differences between plant and animal cells, as well as develop models to describe the function of cells. Additionally, students explore how parts of cells contribute to the overall function of the cell. Finally, bacteria are introduced for students to compare/contrast with plant and animal cells. Overall, students learn that some living organisms, like bacteria, are made of a single cell. Though humans are made of many cells, there can be smaller living things (such as bacteria) living in and/or on us... such as in our dental plaque.

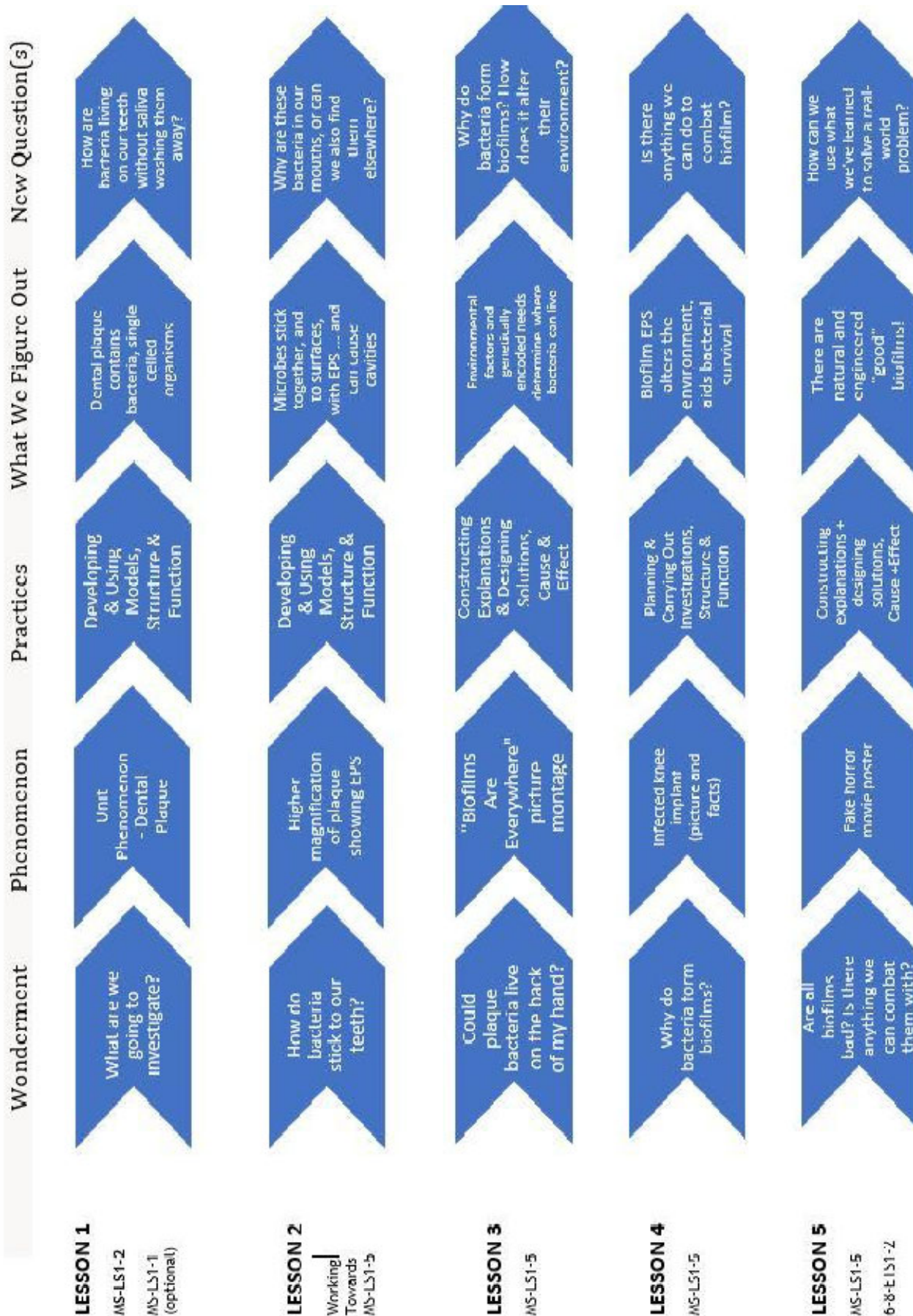
Lesson Two promotes an awareness of oral hygiene methods, and how they pertain to the fight against microbial biofilms in the human mouth. Initially, students self-document oral hygiene methods and investigate cultural differences (and similarities) in the fight against cavities. Then, students explore biofilms and the process by which a biofilm develops through game play. Ultimately, groups collaborate to choreograph a play/skit/dance that models and explains the components of a biofilm, as well as the steps involved in biofilm development. Overall, students learn that bacteria are a major component of dental biofilms (plaque). The microbes of a biofilm collaborate, finding safety in numbers and the sticky extracellular substances they many of them secrete. It can be extremely hard to remove a biofilm completely, even with scraping, and what remains quickly reforms a vibrant new biofilm community.

Students discover, in *Lesson Three*, that biofilms occur just about everywhere. After considering cause and effect relationships between environmental factors (or *available resources*) and genetically encoded factors (*needs*), students engage in discourse during a card sort activity while determining the most likely biofilm environment for supporting the survival of various bacterial species. Overall, students learn that whether an organism like a bacterium thrives, survives, or dies in any environment depends on their needs, encoded within the DNA of their cell(s). If the environmental factors present support the genetically encoded needs of an organism, then it will likely survive in that environment.

Lesson Four focuses on a biofilm infected knee implant introduced in *Lesson Three*, and the fact that antimicrobial substances (like antibiotics) usually fail to clear these infections. Students are challenged to design and carry out an investigation using a model EPS (using one or more widely available polysaccharides) and antimicrobial (food dye) system. The goal is to gather data as evidence in the construction of an explanation for why antimicrobial treatment often fails to cure infected knee implants. Overall, students discover that bacteria in a biofilm alter their environment to help ensure their continued survival. The exclusion of antimicrobial products from the deepest regions of the biofilm is one example.

Students begin *Lesson Five* by observing a fake horror movie poster (*The Biofilm*) and flip through their *Lesson One - Four* notes collecting evidence to use in determining whether they believe all biofilms are bad, or not. Students then share beliefs and evidence with each other before engaging in an activity highlighting examples of beneficial biofilms and biofilm associated engineering solutions. Students then collaborate in the design of an object, tool, process, or system for combating a real-world biofilm-associated concern.

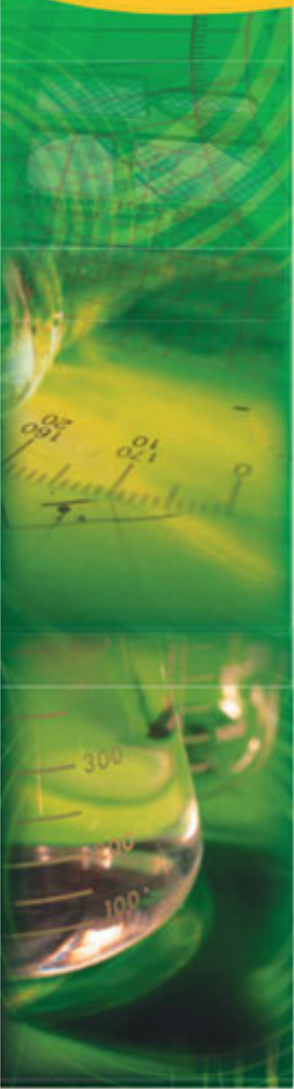
Biofilms- Stuck Like Glue, Stuck On You



Lesson	Standards
1	(Optional) MS-LS1-1 Conduct an investigation to provide evidence that living things are made of cells; either one cell or many different numbers and types of cells. MS-LS1-2 Develop and use a model to describe the function of a cell as a whole and ways parts of cells contribute to the function. Science and Engineering Practice (SEP): - Developing and Using Models- Develop and/or use a model to predict and/or describe phenomena. Crosscutting Concept (CCC): - Structure Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function.
2	Working Towards MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): - Developing and Using Models- Develop and/or use a model to predict and/or describe phenomena. Crosscutting Concept (CCC): - Structure and Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function.
3	MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): - Constructing Explanations and Designing Solutions- Construct an explanation that includes qualitative or quantitative relationships between variables that predict(s) and/or describe(s) phenomena. Crosscutting Concept (CCC): - Cause and Effect- Cause and effect relationships may be used to predict phenomena in natural or designed systems.
4	MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): - Planning and Carrying Out Investigations- Collect data to produce data to serve as the basis for evidence to answer scientific questions. Crosscutting Concept (CCC): - Structure and Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function.
5	MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. 6-8-ETS1-2 Evaluate competing design solutions using a systematic process to determine how well they meet the criteria and constraints of the problem. Science and Engineering Practice (SEP):

	- Constructing Explanations and Designing Solutions- Apply scientific ideas or principles to design, construct, and/or test a design of an object, tool, process or system. Crosscutting Concept (CCC): - Cause and Effect- Cause and effect relationships may be used to predict phenomena in natural or designed systems.
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Science and Engineering Practices

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- 1** Asking questions (science) and defining problems (engineering).
 - 2** Developing and using models.
 - 3** Planning and carrying out investigations.
 - 4** Analyzing and interpreting data.
 - 5** Using mathematics and computational thinking.
 - 6** Constructing explanations (science) and designing solutions (engineering).
 - 7** Engaging in argument from evidence.
 - 8** Obtaining, evaluating, and communicating information.

A Framework for K-12 Science Education: Practices, Crosscutting Concepts, and Core Ideas (NRC 2011)

Materials List

➤ Teacher-Provided

Lesson	Materials Needed
Prior to Lesson 1	➤ Materials to create science notebooks, or to paste notebook scaffold pages into existing science notebooks
1	For teacher: ➤ Projector and speakers For each student: ➤ Lesson One Notebook Scaffold pages (on USB in teacher guide) • Copy of unit phenomenon (dental plaque) • (Optional) Copy of <i>Part A</i> phenomenon (cork) • Bacteria Information Sheet For each group of 3: • Jigsaw resources (A-C)
2	For teacher: ➤ Projector (speakers optional) For each student: ➤ Science notebook scaffold pages • Copy of lesson phenomenon ➤ Copy of blog post How Different Countries And Cultures Handle Oral Health Traditions And Trends ➤ Photocopy of <i>Step Cards</i> (Cavity Creator Game) ➤ Photocopy of <i>Character Cards</i> (Cavity Creator Game) • <i>Tooth Decay And How To Prevent It</i> (for assessment) For each group of 3: • <i>Cavity Creator</i> game board and supply bag
3	For teacher: ➤ Projector For each student: ➤ Science notebook scaffold pages • (Optional) Copy of <i>Lesson Three Phenomenon</i> ➤ Photocopy of <i>Biofilm Descriptions</i> sheet

	➤ Photocopy of <i>Bacterial Species</i> sheet ➤ Photocopy of <i>Lesson Three Assessment</i> habitat and organism cards For each pair: ● <i>Biofilm Bacteria Card Sort</i> supplies
4	For teacher: ➤ Projector and speakers ● (3) Empty 2-liter bottles ● (1) Bottle dextrin (Equate Clear Soluble Fiber Powder) ● (1) Bottle psyllium (Konsyl Fiber Original) ● (1) Bottle guar gum (Benefiber Healthy Balance) ● (1) Box food coloring (red, yellow, blue, green) For each student: ➤ Science notebook scaffold pages ● Copy of <i>Infected Knee Implant Phenomenon</i> ➤ (optional) <i>Presentation Cue Cards</i> for non-presenting students For each group of 2-3: ● (5) Clear containers ● (1) Dropper ➤ Any (teacher approved) classroom supplies
5	For teacher: ➤ Projector and speakers For each student: ➤ Science notebook scaffolds ● Copy of <i>Lesson Five Phenomenon</i> (movie poster) For each group of 4: ● Set of <i>New Biofilm Example Sheets</i> (Labeled A-D) ➤ (1) Large poster paper or <i>Lesson Five Summary Sheets</i> (A-D)

Important Note	
Science Notebooks	You may wish to have students set up their <i>Science Notebooks</i> prior to the first lesson. Science notebooks are an important part of solidifying your students' learning. Student scientists record their observations, questions, evidence and ideas in a running record of their thinking. Science notebooks are also a great way to practice writing skills! As students develop their understanding of science concepts, they may use drawings and other illustrations- such as charts, tables, models, and graphs. Additionally, the science notebook can be used by the teacher to assess the development of student understanding. Science notebooks may be a printed booklet prepared by the teacher, a bound composition, or a spiral notebook. <i>Biofilms- Stuck Like Glue, Stuck On You</i> includes science notebook scaffold pages designed to be cut and pasted onto the left-hand pages of a bound composition or spiral notebook so that the right-hand pages are blank for student responses, notes, sketches and such. There is also a file for creating stand-alone student notebooks that can be printed and stapled, where the right half of each page is blank. The printable science notebook scaffold pages specific to this unit are available on the unit USB located in the front of the <i>Teacher Guide</i> (3-ring binder).

Overview Standard(s) & Objectives	Overview In this lesson, students investigate the similarities and differences between plant and animal cells, as well as develop models to describe the function of cells. Additionally, students explore how parts of cells contribute to the overall function of the cell. Finally, bacteria are introduced for students to compare/contrast with plant and animal cells. <i>Please note that this lesson will likely take more than one class period.</i> Standards (Optional Part A) MS-LS1-1 Conduct an investigation to provide evidence that living things are made of cells; either one cell or many different numbers and types of cells. MS-LS1-2 Develop and use a model to describe the function of a cell as a whole and ways parts of cells contribute to the function. Science and Engineering Practice (SEP): - Developing and Using Models- Develop and/or use a model to predict and/or describe phenomena. Crosscutting Concept (CCC): - Structure and Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function. Objectives Students - Make observations and ask questions about dental plaque. - Compare/contrast plant, animal and bacterial cells. - Develop a model to describe the function of the cell, and how parts of cells contribute to the overall function. - Use a model to describe the unit phenomenon.
Background Information (for teachers)	Adapted from :https://www.nature.com/scitable/topicpage/what-is-a-cell-14023083/ Cells Cells are considered the basic unit of life in part because they come in discrete and easily recognizable packages. This is because all cells are surrounded by a structure called the cell membrane — which, much like the walls of a house, serves as a clear boundary between the cell's internal

and external environments. The cell membrane is sometimes also referred to as the plasma membrane.

Within this membrane, a cell's interior environment is water based. Called cytoplasm, this liquid environment is packed full of cellular machinery and structural elements. In fact, the concentrations of proteins inside a cell far outnumber those on the outside — whether the outside is ocean water (as in the case of a single-celled alga) or blood serum (as in the case of a red blood cell). In plants (as well as fungi and many bacteria), the cell membrane is surrounded by an additional structure called the cell wall. Cell walls provide rigidity and structure.

As previously mentioned, a cell's cytoplasm is home to numerous functional and structural elements. These elements exist in the form of molecules and organelles — picture them as the tools, appliances, and inner rooms of the cell. Major classes of intracellular organic molecules include nucleic acids, proteins, carbohydrates, and lipids, all of which are essential to the cell's functions.

Some cells also feature orderly arrangements of molecules called organelles. Similar to the rooms in a house, these structures are partitioned off from the rest of a cell's interior by their own intracellular membrane. Organelles contain highly technical equipment required for specific jobs within the cell. One example is the mitochondrion — commonly known as the cell's "power plant" — which is the organelle that holds and maintains the machinery involved in energy-producing chemical reactions. Plants have an additional organelle called the chloroplast. Since plants don't typically eat food the way animals do, they need chloroplasts to produce their own food utilizing energy from the sun.

Bacteria

Rather than grouping cells by their size or shape, scientists typically categorize them by how their genetic material is packaged. If the DNA within a cell is not separated from the cytoplasm, then that cell is a prokaryote. All known prokaryotes, such as bacteria and archaea, are single cells. In contrast, if the DNA is partitioned off in its own membrane-bound room called the nucleus, then that cell is a eukaryote. Some eukaryotes, like amoebae, are free-living, single-celled entities. Other eukaryotic cells are part of multicellular organisms. For instance, all plants and animals are made of eukaryotic cells — sometimes even trillions of them.

	Dental plaque Adapted from: https://www.healthline.com/health/dental-and-oral-health/plaque Plaque is a sticky film that forms on your teeth every day: You know, that slippery/fuzzy coating you feel when you first wake up. Scientists call plaque a “biofilm” because it’s actually a community of living microbes surrounded by a gluey polymer layer. The sticky coating helps the microbes attach to surfaces in your mouth so they can grow into thriving microcolonies. Therefore, your mouth is a thriving ecosystem. Bacteria and other organisms come in when you eat, drink, and breathe. Most of the time, a delicate balance is maintained in your oral ecosystem, but problems can arise when certain strains of bacteria become overabundant. When you eat carbohydrates, like sugary foods and drinks, bacteria feed on the sugars and produce acids. These acids can cause problems like cavities, gingivitis, and other forms of tooth decay. When plaque isn’t regularly removed, it can accumulate minerals from your saliva and harden into an off-white or yellow substance called tartar. Tartar builds up along your gumline on the fronts and backs of your teeth. Although an attentive flossing may dislodge some tartar buildup, it typically requires a visit to the dentist to rid yourself of all of it.
Materials	For teacher: ➤ Projector and speakers For each student: ➤ Lesson One Notebook Scaffold pages (on USB in teacher guide) • Copy of unit phenomenon (dental plaque) • (Optional) Copy of <i>Part A</i> phenomenon (cork) • Bacteria Information Sheet For each group of 3: • Jigsaw resources (A-C)
Prior Knowledge	Students should understand that cells are the basic building block of all living things. <i>There is a formative assessment at the beginning of the exploration. If any students do not answer correctly, please consider utilizing the (optional) Part A activity to address this prior knowledge.</i>
Launch (10-15 minutes)	
Phenomenon	Handout copies of the <i>Lesson One Notebook Scaffolds</i> for students to cut in half along the mid-line and paste into their notebooks on the left hand pages, leaving the right hand pages blank. Stress to students that the right hand pages are blank for writing notes, answering prompts, sketching their

	thinking/ideas, and any other visual models they may require through the course of the unit. Project and pass out copies of the unit phenomenon, but do <u>not</u> tell students what it is (there is a picture of a human mouth on it). Ask students to take a few minutes to <u>silently</u> observe the unit phenomenon, and then note the following in their science notebooks (the prompt is found near the top of page 1 of the science notebook scaffold). <i>Please note that the silent think time is essential, as it allows every student time to contemplate and process at their own pace.</i> • What do you notice? • Write down two questions that you have. Tell students that they are going to have an opportunity to discuss, one at a time, what they observed and wondered with a partner. This is called a “Stand and Talk” (for more information about the <i>Stand and Talk</i> strategy, follow this hyperlink). Explain that they will stand face-to-face with a partner (you may arrange this however you like, but choosing partners originally positioned on opposite sides of the classroom to mix it up is strongly encouraged). You will choose which partner shares first (by stating that the partner with the longest hair, or darkest colored hair, etc. goes first), and they will get exactly 1 minute to share what they noticed and wondered. If someone finishes early, both partners stand silently and wait for time to be called (this allows the speaker time to think of something new to share). When time is called, the speaker stops, and then it will be time for the second partner to share for 1 minute (when you say to begin). If time permits, allow an additional minute or so for partners to freely discuss and ask questions of one another. After the <i>Stand and Talk</i>, <u>randomly</u> choose 3-4 students and ask them to share one of the questions or wonderments from their discussion. Make sure that you, the classroom teacher, do not answer any questions at this point (except questions pertaining to the unit as a whole, or the general strategy, if you desire). You may want to tell your students that the following lessons aim to answer most of their questions.
Explore (60 - 90 minutes)	
Exploration	Formative Assessment of prior knowledge Before starting the exploration, check your students understanding of cell theory (MS-LS1-1). When you call “show me”, students should hold up the number of fingers that correspond with the answer of their choice (the prompt and choices are in the notebook scaffolds on page 1). Make sure to give students plenty of silent think time before calling <i>show me</i>.

Question 1: What are all living things made of?

1. Blood
2. Cells
3. Muscle
4. All of the above
5. None of the above

Observe answers without indicating anything to students about the correct answer. Then proceed to the second question:

Question 2: What do all living things “do”?

1. Move around
2. Use energy
3. Breath oxygen
4. All of the above
5. None of the above

Again, observe student answers without indicating which answer was correct. If every student held up two fingers in response to both questions, proceed to *Part B*. If there was a variety of answers for either question (or both), consider beginning with *Part A*. **However, if you plan to begin with *Part A*, DO NOT tell students which answers were correct!**

(Optional) Part A

Display and hand out copies of the cork board phenomenon and direct students to the top of page 3 of the *Lesson One Notebook Scaffold- Cork Phenomenon*. Alternatively, if you have access to microscopes, have students observe the prepared cork slides provided with the kit. Give students a few minutes to silently observe the phenomenon, and then note the following in their science notebooks.

- What do you notice?
- Write down two questions that you have:

Tell students that you are about to show them a 6 minute video. While watching, you’d like them to:

- Listen for answers to their questions
- Write down 2 NEW questions they have after watching the video

Show the TED Talk “[The Wacky History of Cell Theory](#)” (6 min 11 sec).

Conduct another Stand and Talk, but with new partners this time. Use a different determining characteristic to decide which partner shares first. One minute per partner to address the following:

- One of your original questions and the answer you discovered
- One new unanswered question you have

Again, if time permits, allow a minute or so for open discussion between partners.

Randomly select 3-4 students to share one of the answered questions their group discussed, and then select another 3-4 to share a yet unanswered question.

If any unanswered questions pertain to cell theory ([MS-LS1-1](#)) that you feel are critical to answer before continuing, feel free to add whatever activities you desire.

Part B

Ask students to think silently for a moment about what these three things have in common (besides having the word “system” in each). Students should also consider the overall function of each system, the parts of each, and how the parts work together to perform their unique function. *There is a slide on the unit USB to project, if desired. It displays visuals for the systems as well as the questions for students to consider.*

- School System
- Circulatory System
- Justice System

Note: Feel free to alter any of the provided examples, as appropriate, for your students. For instance, maybe your students are more familiar with weather systems, the Dewey Decimal system, or computer operating systems.

Students may write their thinking in their science notebook or hold it in working memory. Then, have students pair-up to share their thinking (2-3 minutes). This can be done through another *Stand and Talk* with new partners, or as a seated “*Dyad*” where partners sit knee-to-knee. Otherwise, the basic structure of the *Dyad* is the same as that for *Stand and Talk*.

Randomly choose a few groups to share what they discussed. Keep calling on groups or hold a larger group discussion if necessary. *A system is a group of related parts that make up a whole and can carry out functions its individual parts cannot.*

Tell students that the basic building block of all living things, the cell, is a system as well. But what is the overall function of the cell? Provide silent think time again, before having partners discuss and share. The task card for this activity is on page 5 of the student notebook scaffold.

After a few minutes of silent think/write time, form groups of three for the discussion. It is acceptable to have a few groups of 4, if necessary. Each student gets about a minute, one at a time, to share his or her thoughts. *To facilitate equitable sharing, consider the use of a [talking stick](#). Any student may begin, but only the student holding the talking stick may speak... while the rest of the group listens. The talking stick is passed around the table (to the left) so that every student has a chance to share. In order to prevent a high-status student from grabbing the talking stick to begin every discussion, consider having any student pick up the stick, and then tell them to hand it to the student on their left. Alternatively, you could specify that the student wearing the darkest colored shirt picks up the stick to begin, and alter the identifying feature at the start of each discussion.*

After every team member has shared his or her ideas, allow 3-5 minutes for open discussion. *To facilitate equitable group discussion, we recommend using [talking chips](#). Almost anything can be used for talking chips; pencils, erasers, poker chips, small colored pieces of paper, small craft pom-poms, etc. Each student needs 3 talking chips. Any student may start the discussion. A talking chip is placed in the center of the table each time a student contributes to the discussion- 1 chip per statement/thought/question. When there are no more chips to be used in the group, students can refresh their chips to continue the discussion. This ensures that every group member contributes to the discussion, and no student is allowed to dominate.*

Be sure to listen-in on student conversations, ready to provide probing questions as necessary. If students are having trouble accessing the question, here are some suggestions for rephrasing the prompt:

- If cells are the basic “building block” of living organisms, and you may be familiar with building things out of blocks, what does this tell you about the purpose of all those cells in a plant or animal made out of many-many cells?
- What are the things that all living organisms do? (If your students are familiar with any single celled organisms, you might remind them that those single cells are still living things!)

Wrap-up by randomly asking 2-4 groups to share what they believe are the overall function(s) of the cell, and why.

Cell Parts Jigsaw

Explain that next they will consider the individual parts of the cell, and how each contributes to the overall function. Continue using groups of three, these will be the “home groups” for the jigsaw activity. Hand each group a set of jigsaw resources, labeled A-C, and direct students to the task card on page 5 of the notebook scaffold. Each student will choose one of the resources (Nucleus, Cell Wall/Membrane, or Mitochondria/Chloroplast) to become the “expert” on that resource for the group.

We recommend printing copies of the A-C resources so students can interact with the text through underlining, highlighting, making notes in the margins, And such..

Students should take 5-8 minutes to silently read the resource and contemplate the questions provided. Note taking is encouraged.

Next, form “expert groups”. Every student who analyzed resource “A” should gather for discussion, and every student who analyzed resource “B” should gather in a different location, etc. (*Expert group sizes of 4-5 students are optimal, but not always achievable. Consider forming two of each expert group if you have a large class*). Each expert group should attempt to come to consensus as they consider the associated questions. The use of a [talking stick](#) and/or [talking chips](#) is highly encouraged. Circulate among the expert groups, listening for any that might need you to ask guiding or probing questions to help them draw logical, truthful, conclusions.

Reassemble the original “home groups”. Each student (or “expert”) will, one at a time, share their findings/learning with their team members. When not sharing, students should be listening attentively and making notes... as every student is responsible for every piece of learning, even if they were not an “expert” on it to begin with.

Groups should then collaborate on crafting, in their science notebooks, two cell models: a plant cell model, and an animal cell model. Each cell part should be labeled somehow, with both name and function.

Creativity and originality are encouraged! Every student should craft his or her cell models in whatever manner desired, though the general information should be similar.

	Finally, group members will collaborate as they compare and contrast plant and animal cells.
Questions	Driving Question - What's on our teeth? Probing Questions - What is the overall function of the cell, as a whole? - How does each part of the cell contribute to the overall function? - How are plant cells similar to animal cells? - How are plant cells different from animal cells? - What are bacteria? - How do bacteria compare with plant and animal cells? - What evidence is there that dental plaque is/is not living "alive"?
Summarize (20-30 minutes)	
Communicate	Continue with the same groups of three for this activity. Hand each student a copy of the <i>Bacteria Information Sheet</i>. The task card for this activity is on page 7 of the notebook scaffold. Remind students to honor the silent <i>read/think time</i> their team members require before beginning discussion and model creation. A printed copy of the bacteria resource sheet is recommended so that students may interact with the text. Groups will collaborate during the discussion and planning phase but will individually create their own bacterial cell models. This allows each student to express his or her creativity and originality. Finally, students will collaborate on comparing/contrasting bacterial cells with plant and animal cells and deciding what ALL cells have in common. Wrap-up with groups sharing one thing they noticed that all cells have in common, and/or something they are wondering. Homework! Encourage students to consider the ways that they care for their teeth before the next lesson. Additionally, students should consider what they believe to be the purpose of each strategy. They should also ask family and friends, outside of class time, about their own strategies and ideas. The task is found on page 7 of the notebook scaffold. Consider having students self-document their strategies for posting in the classroom. STEM Teaching Tool #31 provides information on self-documentation and is hyperlinked here.

Terminology and Concepts to Solidify	System- a group of related parts that make up a whole and can carry out functions its individual parts cannot. Cell- the smallest structural and functional unit of an organism. Plant- a living organism of the kind exemplified by trees, shrubs, herbs, grasses, ferns, and mosses, typically growing in a permanent site. Plants absorb water and inorganic substances through their roots and synthesize food in their leaves by photosynthesis in their chloroplasts. Animal- a living organism that feeds on organic matter (plants and/or other animals), typically having specialized sense organs and nervous system, and able to respond rapidly to stimuli. Bacterium- a member of a large group of unicellular microorganisms, many of which have cell walls, but generally lack membrane-bound organelles or an organized nucleus. Nucleus- a dense organelle presents in most eukaryotic cells, typically a single rounded structure bounded by a double membrane, containing the genetic material. Chloroplast- an organelle found in green plant cells that contains chlorophyll and enables photosynthesis to take place, generating food for the plant. Mitochondria- an organelle in the cytoplasm of eukaryotic cells (not in bacteria) that functions in energy production. Cell Membrane- the semipermeable membrane surrounding the cytoplasm of a cell. Cell Wall- a rigid layer lying outside the cell membrane of the cells of plants, fungi, and most bacteria.
Connection to Big Ideas (Phenomena)	All living things are made of cells. Some living organisms, like bacteria, are made of a single cell. Other living things are made of many cells. Though humans are made of many cells, there can be smaller living things, like bacteria, living in and/or on us... such as in our dental plaque.
(Optional) Follow Up/Practice	Cell Theory Song: https://www.youtube.com/watch?v=KuJqqiATlgw If you have access to microscopes, consider having students make and observe slides of their • cheek cells: https://learning-center.homesciencetools.com/article/microscope-experiments/#SmearCheekCells

	Dental plaque: How to Prepare Microscope Slides: A Step by Step Guide » Microscope Club Prepared slides of cheek, liver, cork, onion and privet leaf are also included in the kit for microscopic observation.
Assessment	<i>Lesson One Assessment (prompt on page 7 of the notebook scaffold):</i> Observe the lesson phenomenon pictures again. Scientists call this substance found on teeth “dental plaque”. The small round objects in the 17,500x magnified picture are bacteria. Using what you have learned in <i>Lesson One</i>, decide if you believe that dental plaque is “alive”. Why or why not? What evidence do you have? Be certain to refer to your cell models in support of your argument! <i>Note: Accept all well supported answers, as this question can be argued either way.</i>

Lead-in to Lesson 2- what about the non-round and not-so-small things in the 17,500x magnified picture? How do bacteria stick to tooth surfaces... why don't they get washed away by saliva?

Lesson 2	What Is A Biofilm?
Overview Standard(s) & Objectives	Overview This lesson promotes an awareness of oral hygiene methods, and how they pertain to the fight against microbial biofilms in the human mouth. Initially, students self-document oral hygiene methods and investigate cultural differences (and similarities) in the fight against cavities. Then, students explore biofilms and the process by which a biofilm develops through game play. Ultimately, groups collaborate to choreograph a play/skit/dance that models and explains the components of a biofilm, as well as the steps involved in biofilm development. <i>This lesson is expected to take 2 class periods.</i> Standards Working Towards MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): ● Developing and Using Models- Develop and/or use a model to predict and/or describe phenomena. Crosscutting Concept (CCC): ● Structure and Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function.

	Objectives Students • Make observations and ask questions about Extracellular Polymeric Substance (EPS) in dental plaque. • Consider a variety of oral hygiene methods and hypothesize about how/why they work. • Utilize a game-based model to learn about biofilms, their components and the steps of development. • Model the parts and development of biofilms through acting/dance. • Relate oral hygiene strategies to biofilms, explaining how each strategy effects dental plaque (thus working to prevent cavities).
Background Information (for teachers)	Biofilms Adapted from https://www.livescience.com/57295-biofilms.html Biofilms are a collection of one or more types of microorganisms that can grow on many different surfaces. Microorganisms that form biofilms include bacteria, fungi and protozoa. One common example of a biofilm is dental plaque, a slimy buildup of bacteria that forms on the surfaces of teeth. Pond scum is another example. Biofilms have been found growing on minerals and metals. They have been found underwater, underground and above the ground. They can grow on plant tissues and animal tissues, and on implanted medical devices such as catheters and pacemakers. Each of these distinct surfaces has a common defining feature: they are wet. Biofilms have been around for a very long time. Fossil evidence of biofilms dates to about 3.25 billion years ago. For example, biofilms have been found in the 3.2 billion-year-old deep-sea hydrothermal rocks of the Pilbara Craton in Australia. Similar biofilms are found in hydrothermal environments such as hot springs and deep-sea vents. Biofilm formation begins when free-floating microorganisms such as bacteria come in contact with an appropriate surface and begin to put down roots, so to speak. This first step of attachment occurs when the microorganisms produce a gooey substance known as an extracellular polymeric substance (EPS). An EPS is a network of sugars, proteins and nucleic acids (such as DNA). It enables the microorganisms in a biofilm to stick together, as well as stick to a surface. Attachment is followed by a period of growth. Further layers of microorganisms and EPS build upon the first layers. Ultimately, they create a bulbous and complex 3D structure. Water channels crisscross biofilms and allow for the exchange of nutrients and waste products.

Multiple environmental conditions help determine the extent to which a biofilm grows. These factors also determine whether it is made of only a few layers of cells, or many. For instance, microorganisms that produce a large number of EPS can grow into thick biofilms, even if they do not have access to a lot of nutrients. On the other hand, for microorganisms that depend on oxygen, the amount of oxygen able to penetrate the biofilm limits how thick they can grow. Another environmental factor is the concept of "shear stress". If there is a very high flow of liquid across a biofilm, like in a creek, the biofilm is usually thin. If you have a biofilm in slow flowing water, like in a pond, it can become thick.

Finally, the cells within a biofilm can leave and establish themselves on a new surface. Either a clump of cells breaks away, or individual cells burst out of the biofilm and seek out a new home.

Living as part of a biofilm comes with certain advantages, as communities of microbes are usually more resilient to stress as compared to those living alone. Potential stressors include the lack of water, high or low pH, or the presence of substances toxic to microorganisms such as antibiotics, antimicrobials or heavy metals.

There are many possible explanations for the hardness of biofilms. For example, the slimy EPS covering can act as a protective barrier. It can help prevent dehydration or act as a shield against ultraviolet (UV) light. Also, harmful substances such as antimicrobials, bleach or metals are either bound or neutralized when they come into contact with the outermost layers of EPS. Thus, they are diluted to concentrations that aren't lethal well before they reach cells deep in the biofilm.

Still, it is possible for certain antibiotics to penetrate the EPS and make their way through a biofilm's layers. Here, another protective mechanism can come into play: the presence of bacteria that are physiologically dormant. In order to work well, all antibiotics require some level of cellular activity. So, if bacteria are physiologically dormant to begin with, there is not much for an antibiotic to disrupt. The dormant bacteria will be around to begin a new biofilm community when the antibiotics are no longer present.

Modeling Through Movement

There are many different ways to *model* scientific concepts, but the use of physical movement through acting and/or dancing...physically experiencing and expressing the concepts... can be particularly beneficial.

	Additionally, choreographing and performing an original model creates an avenue for student creativity, which can lead to increased student engagement. This video about the 2018/2019 <i>Dance Your PhD</i> winner in the social sciences category explains using a variety of physical science examples: https://www.youtube.com/watch?v=oLCTJQ-GCLQ
Materials	For teacher: ➤ Projector (speakers optional) For each student: ➤ Science notebook scaffold pages ➤ Copy of blog post How Different Countries And Cultures Handle Oral Health Traditions And Trends ➤ Photocopy of <i>Step Cards</i> (Cavity Creator Game) ➤ Photocopy of <i>Character Cards</i> (Cavity Creator Game) • <i>Tooth Decay And How To Prevent It</i> (for assessment) For each group • Copies of lesson phenomenon • <i>Cavity Creator</i> game board and supply bag
Prior Knowledge	Students should understand that cells are the basic building block of all living things (<i>Prior Knowledge/Lesson One</i>) There are multiple cell types (such as animal, plant and bacteria), yet all cells share some common core parts and functions. Dental plaque on our teeth contains bacterial cells (<i>Lesson One</i>).
Launch (10-15 minutes)	
Phenomenon	As students enter the classroom, remind them about their <i>Lesson One</i> “homework” (asking friends and family what ways they care for their teeth, as well as considering their own habits). Ask students to post these methods in the location of your choice, grouping similar strategies together. Handout copies of the <i>Lesson Two Notebook Scaffolds</i> for students to paste into their notebooks on the left hand side, leaving the right hand pages blank. Project and pass out copies of the <i>Lesson Two</i> phenomenon (there is a picture of <u>really</u> thick dental plaque in the lower left corner). Ask students to take a few minutes to <u>silently</u> observe the lesson phenomenon, and then note the following in their science notebooks.

	• What do you notice? • Write down two questions that you have. Form groups of 3 students (a few groups of 4 is acceptable). Groups should use talking chips to share ideas and discuss interesting observations and/or wonderments. <u>Randomly</u> choose 3-4 students and ask them to share one of the questions or wonderments from their discussion. Make sure that you, the classroom teacher, do not answer any questions at this point. The desire is for student wonderments to drive their engagement and learning through the lesson.
Explore (30-40 minutes)	
Exploration	Students should remain in their groups of 3. Students should take a few minutes to consider the ways that they care for their teeth, as well as any different methods posted on the self-documentation board, carefully considering what they believe to be the purpose of each strategy. The prompt is located on page 9 of the notebook scaffold. Students should write their thoughts on the blank notebook page to the right of the prompt. Group members then share their thoughts, one at a time, before an open discussion for 2-3 minutes. Talking stick and/or chips are recommended. <i>Note: At this point, most ideas about how oral hygiene methods work will likely focus on cleaning and/or disinfecting. It is important to note that there are really no wrong answers... the rest of Lesson Two should provide a clearer picture of what is happening in the human mouth and why methods such as brushing and/or flossing (methods of scraping) are vital.</i> Have students read the blog post How Different Countries And Cultures Handle Oral Health Traditions And Trends, or supply the article to read outside of class time (the article is available on the unit USB, as well as the above hyperlink). Printed copies of the article are recommended so that students can interact with the text using pen, pencil and/or highlighter. As students read, they should be looking for various methods different cultures around the world use for oral hygiene. Additionally, they should watch for common themes- what do some methods have in common, both in terms of desired outcome as well as method of action. Students will find these prompts on page 9 of the science notebook scaffold. Cavity Creator Game Explain that your students are about to use a model to explore the parts of a biofilm, as well as the steps in development of a biofilm. The instructions

begin on page 11 of the notebook scaffold. The model they will be using is in the form of a game. As they play, they will be collecting “Step Cards”. Additionally, they will collect “Character Cards” upon completion of the game. These cards are designed to facilitate note taking during game play, but not replace it. Students should record anything that aids their understanding of biofilm development because they will be responsible for choreographing a skit, play, dance (etc.) to perform at the end of the lesson. Specifically, they need to be looking closely for:

- The parts of a biofilm (in this case, dental plaque)... what needs to be present for a biofilm to develop?
- The steps of biofilm development (and, maybe, dental cavities)... is there a specific order in which events must take place? Why?

Pass out the *Cavity Creator* game boards and supply bags. Each supply bag should contain a die, a token for the game board, a card deck, and “EPS” tokens. Also, provide each student with a sheet of “Step Cards” to cutout and *collect* as they play. The collection of *Step Cards* during game play models, in order, the steps of biofilm development. It is the order of *Step Card* collection that matters most, and *Step Cards* should be fixed in place in student notebooks once they have been properly ordered. It will most likely take about 5 minutes for groups to read the directions, cut out the *Step Cards* and prepare the board and supplies.

5 minute Check-In: As groups begin actual game play, pause the class for a “check-in”. Randomly call on students to share something they need to be paying attention to (or “looking for”) as they play the game (see page 11 of the student notebook scaffold). Answer questions the students may still have, and then resume game play.

Allow about 10 minutes for game play.

Cycle around the groups watching for any that may need encouragement moving on to planning and choreographing their skit, play, or dance (etc.). An understanding of how cavities form is desirable, but not essential.

When groups come to you for their final “check-in”, look for 1) a basic understanding of the components of a biofilm (bacteria, a surface, EPS, etc.), 2) the steps involved in biofilm formation (correctly ordered *Step Cards*), and 3) logical explanations for why our tooth care methods work (what effect they have on biofilms in the human mouth).

Note: Groups do not necessarily need to reach the “Finish” space on the game board to pass the final “check-in” and progress toward choreographing their presentation.

	When groups pass the final “check-in”, hand each student a copy of the “<i>Character Card</i>” sheet. If desired, students may cut these out and fix them into their notebook next to the <i>Step Card(s)</i> they believe each character most likely to influence. The ordered <i>Step Cards</i> and <i>Character Cards</i> are to aid students in the development of their skit/play/dance. As students are choreographing their presentations, remind them that they should be ready to explain how their work models the parts and development of biofilms. At a minimum, presentations should clearly model all five steps of biofilm development represented with the <i>Step Cards</i>, and utilize all three characters provided by the <i>Character Cards</i>. Group presentations will likely occur on another day. Encourage students to continue planning as needed until the next class period. This also provides time for groups to find and/or develop props. <i>Note: Consider allowing multiple groups to join forces if more “actors” are desired.</i>
Questions	Driving Question • What are biofilms, and how do they develop? Probing Questions • What are the important components of a biofilm? • What are the key steps of biofilm development? • How are cavities in teeth created?
Summarize (Time depends on class size)	
Communicate	Group Presentations As students watch each presentation, they should use their ordered <i>Step</i> and <i>Character Cards</i> to keep track of the number of parts/steps that each presentation clearly portrays. When you call “Show Me”, students hold-up the number of fingers that match their assessment. <i>A 100% would be eight fingers, but students may show more than eight fingers if they believe the presentation went above and beyond to portray more than the 8 required parts/steps.</i> Depending on the responses, call on students for clarification and/or elaboration. Potential prompts include: • What do you believe was best about the presentation? • What questions do you have for the presenters?

	• How was step _____ portrayed? How do you know? • How was the _____ bacteria/character portrayed? What is your evidence? • What do you wonder? Why? Think and Share (Part 2) Ask students to look back at page 9 of their science notebook and recall the <i>Think and Share</i> group discussion. Allow students a few minutes to reflect on how their understanding of how/why our dental hygiene methods work may have changed. Now that they understand a bit more about the biofilm that grows on our teeth, how do our dental hygiene methods affect that biofilm? Group members then share their thoughts, one at a time, before an open discussion for 2-3 minutes (prompts on notebook page 15). Talking stick and chips are recommended.
Terminology and Concepts to Solidify	Biofilm- a community of microorganisms living together within a sticky substance called extracellular polymeric substance (EPS). Biofilms tend to form on moist surfaces. Extra Polymeric Substance- a sticky network of sugars, proteins and nucleic acids (such as DNA) secreted by some bacteria. It enables the microorganisms in a biofilm to stick together, as well as stick to a surface. Species- a group of living organisms consisting of similar individuals. The species is the principal natural taxonomic unit, denoted by a Latin binomial name, e.g. <i>Homo sapiens</i> (<i>Homo</i> is the genus, and <i>sapiens</i> is the species).
Connection to Big Ideas (Phenomena)	Microbes like bacteria are a major component of dental biofilms (plaque). The microbes of a biofilm collaborate, finding safety in numbers and the sticky extracellular substances they many of them secrete. It can be extremely hard to remove a biofilm completely, even with scraping, and what remains quickly reforms a vibrant new biofilm community.
(Optional) Follow Up/Practice	<i>Stronger and Clearer Every Time</i> Strategy- What is a biofilm? Use this strategy to empower your students as they develop working definitions of the term “biofilm”. This strategy comes from the <i>Stanford Math Language Routines</i> hyperlinked here, beginning on page 9. Interesting article about dental plaque destroying micro-sized robots: https://penntoday.upenn.edu/news/army-microrobots-can-wipe-out-dental-plaque The American Dental Association has a variety of resources for the classroom: https://www.mouthhealthy.org/en/resources/lesson-plans Video of plaque removal, portion showing demineralization of a tooth: https://www.youtube.com/watch?v=8lqeW6_QhtU

	Science Friday Ask A Dentist: <i>Brushing Up On Tooth Science</i> (33 min 45 sec audio only): https://www.sciencefriday.com/segments/ask-a-dentist-tooth-science/ Consider acquiring “plaque disclosing tablets” and let your students design their own experiments aimed at testing the performance of various tooth cleaning methods. For ideas: https://www.dentaly.org/us/oral-hygiene/dental-plaque-tartar/plaque-disclosing-tablets/
Assessment 	Refer to your biofilm notes and the poster entitled <i>Tooth Decay and How to Prevent It</i>. Choose two of the methods presented on the poster and discuss how they help to limit/prevent human tooth decay. Pay particular attention to the structures found in biofilms, and their functions in aiding the microbial cells within. <i>Note: Accept all logical and well-supported answers. Examples include brushing to physically remove biofilm, dental visits to more thoroughly remove biofilm, and maybe even brushing to expose more biofilm bacteria to an antimicrobial mouthwash. Other acceptable answers may focus on eating a low sugar diet and/or brushing immediately after meals to inhibit biofilm formation by essentially “starving” the bacteria.</i>

Lead-in to Lesson 3- Where else can we find biofilms? Are they only on our teeth?

Lesson 3	Biofilms Are Everywhere!
Overview Standard(s) & Objectives	Overview In this lesson, students will learn that biofilms occur just about everywhere. Using a biofilm habitat and bacterial species card sort, students explore the relationships between environmental and genetic factors. It is the genetic factors encoded within an organism’s DNA (it’s “needs”) that determine which environments are capable of supplying those needs (the “availability”). In the end, students analyze the environmental factors available in three South Dakota habitats as they determine where two grasses are most likely to thrive. Standards MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): Constructing Explanations and Designing Solutions- Construct an explanation that includes qualitative or quantitative relationships between variables that predict(s) and/or describe(s) phenomena.

	Crosscutting Concept (CCC): ● Cause and Effect- Cause and effect relationships may be used to predict phenomena in natural or designed systems. Objectives Students ● Ask questions about biofilms. ● Consider cause and effect relationships between environmental and genetic factors in influencing the growth of organisms. ● Engage in discourse while determining the most likely biofilm environment for supporting the survival of various bacterial species. ● Construct a qualitative explanation for the relationships between environmental and genetic factors in determining where a species is likely to survive and thrive.
Background Information (for teachers)	Environmental and Genetic Factors Bacteria exist just about everywhere on Earth, even nearly a mile underground at the Sanford Underground Research Facility in Lead, South Dakota. Yet, each particular environment has specific factors associated with it that can either help, or hinder, living organisms attempting to exist there. Examples of environmental factors include the availability of food, light, space, and water. Bacteria are no different from other living organisms. Different types (or <i>species</i>) of bacteria contain different genetic factors in their DNA, and these determine what each bacterium needs to survive, grow and reproduce. It is a combination of the genetic and environmental factors that determine whether an organism survives, thrives, or dies. The contributions of specific factors to organism growth can only be described using probability (e.g., not every fish in a large pond grows to the same size). Biofilm A- 4,850 feet underground at the Sanford Lab Nearly a mile underground in Lead, South Dakota, scientists are studying the biofilms of the Sanford Underground Research Facility (Sanford Lab). Home to some of the world's most cutting-edge physics and astrophysics experiments, the lab also provides a unique environment for a large variety of microbial species. Cut-off from the rays of the sun, where the rock temperature is about 96 degrees Fahrenheit and the air temperature in the 70's due to ventilation from the surface, many wet surfaces host biofilms of various colors. Many of the bacteria found here are new to science. The species found in various biofilms appear to relate to the water source for each biofilm. Those with similar water sources seem to host similar species. Geologists are even studying the microbes found in

different parts of the underground spaces in an effort to map the various water sources seeping slowly through cracks and fissures present in the rock.

Biofilm B- Infected wound

Adapted from <https://www.woundsource.com/blog/what-are-biofilms>

Biofilms can have a significant impact on wound healing; by contributing to bacterial infection, inflammation, and delayed wound healing. These issues make reducing biofilm presence a critical component of effective wound care. 60% of chronic wounds contain a biofilm! As the body tries to fight the biofilm through an inflammatory response, the body may actually help the biofilm by providing nutrition in the form of exudate (secretions). This creates a situation in which the body is ineffectively fighting biofilms while damaging healing tissue and delaying wound healing. Initially, health care practitioners can reverse the development of the biofilm through cleaning and scraping. After a period of time, though, the biofilm forms a more permanent attachment and begins producing extracellular polymeric substance (EPS). EPS creates a protective barrier that makes it significantly more difficult to sanitize the wound and remove the bacteria. Even after debriding (scraping), a well-established biofilm can form again in as little as 24 hours.

Biofilm C- Knee implant (biofilm stained black)

In reflecting upon the (rare, yet increasing) infection of implanted devices, A. G. Gristina coined the idea of a “race to the surface”. Implanted biomaterials act as fresh, uncolonized environments. This “race to the surface” pertains to the concept of healthy host tissues competing against microorganisms in a race to attach to (or “inoculate”) available surfaces like biomaterials (plates, screws, wires, joint prosthesis, etc). If host tissues attach first, the surface can be defended and is less available for bacterial colonization. However, if the microorganisms win out, then biofilms can form. Biofilm formation takes place in steps. First is the formation of a conditioning film on the surface of the biomaterial, occurring seconds after exposure. This film allows for microorganisms to adhere to the biomaterial surface. At this point, the microorganisms are reversibly attached. Next is the production of the extracellular polymeric substance (EPS) and more permanent adherence. The EPS not only acts as the anchor, but is also a protection against antibiotics. These organisms also have a lowered metabolic state, reducing antibiotic sensitivity and creating a difficulty in detection through microbiological culturing.

Biofilm D- Streambed rock

Adapted from <https://stroudcenter.org/news/getting-slimed/>

	Slippery, shiny, and available in varying shades of green, the slime covering rocks in streams is a biofilm. This matrix of freshwater microorganisms (often including algae), sand, and silt makes up much of the biomass covering underwater surfaces in streams and rivers. The slower water flows, the thicker the biofilm grows. If you have ever seen a sign warning against climbing on the rocks near a waterfall, it is because the slippery biofilm coating is dangerous. Every year, people sustain injuries (or die) when trying to get just a little too close for that perfect photograph. Biofilm E- Used bathtub rubber ducky (cut open) Adapted from https://www.discovermagazine.com/planet-earth/rubber-ducky-youre-the-onewhos-filled-with-nasty-biofilms Kids' bath toys can get gross, especially the ones with a small hole that allows water to get in and never come out. Scientists observing a black residue on the inside of these bath toys decided to investigate. They collected bath toys (e.g., rubber ducks) from households and proceeded to cut the toys in half and use a variety of methods to characterize the biofilms growing inside. They found fungi (mold), as well as "dense biofilms with complex bacterial and fungal communities." The communities were diverse, with many rarely identified bacteria. The scientists suggest that bath toy biofilms are influenced by (1) the carbon leaching from the flexible plastic material, (2) the chemical and biological tap water quality, (3) additional nutrients from personal care products and human body fluids in the bath water, as well as, (4) additional bacteria from dirt and/or the end-users' microbiome.
Materials	For teacher: ➤ Projector For each student: ➤ Science notebook scaffold pages • (Optional) Copy of <i>Lesson Three Phenomenon</i> ➤ Photocopy of <i>Biofilm Descriptions</i> sheet ➤ Photocopy of <i>Bacterial Species</i> sheet ➤ Photocopy of <i>Lesson Three Assessment</i> habitat and organism cards For each pair: • <i>Biofilm Bacteria Card Sort</i> supplies
Prior Knowledge	Students should understand that cells are the basic building block of all living things (<i>Prior Knowledge/Lesson One</i>)

	There are multiple cell types (such as animal, plant and bacteria), yet all cells share some common core parts and functions. Dental plaque on our teeth contains bacterial cells (<i>Lesson One</i>). Communities of bacteria working together for survival create Biofilms. The production of Extracellular Polymeric Substance (EPS) helps the bacteria stick together, helps adhere the biofilm to the surface on which it forms, and protects the bacteria found within. The most effective method of removing a biofilm is scraping it off (<i>Lesson Two</i>).
Launch (10 minutes)	
Phenomenon	Display and/or hand out copies of the <i>Lesson Three Phenomenon</i> picture montage. Give students 2-3 minutes to silently observe and contemplate these new biofilm pictures. Students should then write down two questions they have about biofilms <i>in general</i>. Each question should be on a separate scrap of paper, and they should be anonymous (do not write names on the scraps of paper). Have students separately ball-up each scrap of paper in preparation for <u>randomly</u> tossing them to different parts of the classroom. You might split students so that half are on one side of the room, and the other half on the opposite side. When you call “toss it”, have students throw their questions (paper balls). Each student then picks-up two new questions written by other students. Give students 2-3 minutes to silently read and contemplate these new questions, written by their fellow classmates. Then randomly call on 3-4 students to share the question they find most interesting (either one of their own questions or one anonymously written by a classmate). The rest of this lesson, as well as the final two lessons, aim to answer as many student questions as possible.
Explore (25-35 minutes)	
Investigation	Form groups of 3-4 students. Direct students to page 17 of the notebook scaffold. This task card will help direct students as they collaborate in their groups. Explain that in order to understand these new biofilms, they will need to begin contemplating how environmental and genetic factors contribute to the growth of organisms. Allow 2-3 minutes for students to consider (silently) the differences and similarities between the inside of their mouth, and the skin on the back of their hand. They should pay particular attention to <i>environmental factors</i> in these two places. Examples of environmental factors include the

availability of food, light, space, water and other such characteristics of the places that organisms might live.
Each group member will then share, one at a time, their thoughts about the similarities and differences between the environmental factors present in a human mouth as opposed to the back of a hand. Talking sticks recommended.

Groups should produce a Venn Diagram, T-chart, or other visual representation of the similarities and differences between the environmental factors “available” in each habitat (mouth versus hand).

Groups should then discuss whether they believe the same bacteria that live in their mouths would be able to exist (long-term) on the backs of their hands, as well as their reasoning (talking chips recommended). As you walk around the classroom, listen for groups that might require prompting to ensure they are explaining the reasoning behind their ideas. Also, listen for any struggling groups that would benefit from leading questions:

- What evidence do you have for that?
- What would be the effect on your health and wellbeing if you suddenly found yourself on the moon, where there is little atmosphere?
- Which similarities, or differences, between the two places make you think that?
- Would a fish be able to live, grow and reproduce on land? Why or why not?

Wrap-up by randomly calling on 3-4 students to share an interesting conclusion, insight, or “A-HA!” from their group’s discussion.

Biofilm Bacteria Card Sort (Pairs)

The task card for this activity is on page 19 of the notebook scaffold.

Provide a photocopy of the *Biofilm Descriptions* sheet and the *Bacterial Species* descriptions sheet to every student. Provide at least 5 minutes for students to work, silently and individually, on which biofilm environment they believe each bacterial species might thrive. Photocopies of these

	sheets enable students to interact with the text: make notes, draw connecting lines, and note reasons for their choices. <u>The Challenge:</u> Sort the bacteria according to which biofilm you believe is most likely to support its needs (where will each bacterium thrive?). Explain WHY you chose each placement. Remember to consider the environmental factors present at each biofilm's location (the "availability"), and what genetic factors ("needs") encoded in the bacterial DNA would make that an appropriate location. Hand out the <i>Biofilm Bacteria Card Sort</i> materials and create student pairs. Partners will begin by sharing their thoughts, one at a time, and then comparing. Students will use discourse as they justify their placements, and consider the pros and cons of each potential biofilm environment for the various bacterial species. In the end, partners should come to consensus on where to place each bacterial species, and why they believe that is the best biofilm environment. The card sort materials are there to help as students work to find a mutually agreed upon arrangement. <i>Note: there is really no clear-cut "correct" way to sort the bacteria in this activity. What matters most is that students support their choices with logical reasoning based on the information provided. The fact that there are multiple possible placements for most bacterial species encourages student discourse, and the use of logical reasoning, in supporting various points of view.</i>
Questions	Driving Question: - How do environmental and genetic factors influence the growth of organisms? Probing Questions - What is an environmental factor? - What is a genetic factor? - How do genetic factors determine the species capable of growing in a particular environment? - What environmental factor(s) are most important for bacterial biofilm formation? - What are the potential effects on organisms when placed into an environment that does not support their genetically encoded needs?
Summarize (10-15 minutes)	

Communicate	Pairs should join to form groups of 4. Each pair will share their <i>Biofilm Bacteria Card Sort</i> groupings and reasoning, and then the larger group of 4 will compare, discuss and attempt to reach consensus (page 19 of the notebook scaffold). Prompts for clarification and refining student thinking/reasoning: • Why did you place _____ in biofilm __ and not Biofilm __? • If bacterium _____ requires ____, how does biofilm __ provide that environmental factor? • Couldn't _____ also find what it needs in biofilm __? Finally, groups should construct a qualitative explanation for the relationships between environmental and genetic factors in determining where a species is likely to survive and thrive. (Optional) Have groups create a poster displaying their <i>Biofilm Bacteria Card Sort</i> groupings <u>and reasoning</u> for a gallery walk. As students contemplate the work of other groups, they should leave a peer feedback question at each poster. Suggested prompts:
Terminology and Concepts to Solidify	Environmental factor- any factor, living or non-living, that influences the growth and development of other living organisms. Abiotic factors include ambient temperature, amount of sunlight, and pH of the water or soil in which an organism lives. Biotic factors include all the other organisms in an ecosystem. Genetic factor- a factor influencing the growth and development of an organism that is contained within that living organism. The genetic information contained within the nucleus of the cell(s) of an organism.
Connection to Big Ideas (Phenomena)	Bacteria exist just about everywhere on Earth. Yet, each environment has specific factors associated with it that can either help, or hinder, living organisms attempting to exist there. Whether an organism like a bacterium thrives, survives, or dies in any particular environment depends on their needs, encoded within the DNA of their cell(s). If the environmental factors present support the genetically encoded needs of an organism, then it will likely survive in that environment.
(Optional) Follow Up/Practice	Bring the Sanford Underground Research Facility into your school or classroom! Invite an Education and Outreach team member to visit your school. They will talk about the lab, share pictures and video, as well as answer questions from students. Our programs are available to schools

	across South Dakota, free of charge. https://sanfordlab.org/educators/school-presentations A short article about the use of deep underground microbes for mapping water flows can be found in this Sanford Lab Deep Thoughts. Snap and Share: Challenge students to discover and document interesting biofilms growing in their area. Consider creating a space for students to share pictures and descriptions of the biofilms that they find.
Assessment	Carefully analyze the two grasses and three environments found in South Dakota. Based on the environmental factors available in each location, and the genetic needs of the grasses, decide where each grass would most likely survive and thrive. Clearly state where you placed each grass, as well as your reasoning. Make sure to address both environmental availability and genetic needs in your explanation. Additionally, explain what you believe would happen (effect) to each grass if it were placed into one of the <i>other</i> (not-best) environments (cause).

Lead-in to Lesson 4- As students begin to understand that biofilms are just about everywhere, they may begin asking questions. What makes biofilms so successful? Why do bacteria form biofilms? Why are biofilms so hard to eliminate?

Lesson 4	Why Do Bacteria Form Biofilms?
Overview Standard(s) & Objectives	Overview This lesson focuses on the infected knee implant biofilm of <i>Lesson Three</i>, and the fact that antimicrobial substances (like antibiotics) usually fail to clear such infections. Students are challenged to design and carry out an investigation using a model EPS (biofilm) and antimicrobial (food dye) system. The goal is to gather data as evidence in the construction of an explanation for why biofilm infections are resistant to antibiotics. <i>This lesson will likely take 2 class periods, due to poster creation and presentations.</i> <i>Note: Please prepare EPS solutions ahead of time. More information provided in the Exploration section of this lesson.</i> Standards MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. Science and Engineering Practice (SEP): - Planning and Carrying Out Investigations- Collect data to produce data to serve as the basis for evidence to answer scientific questions. Crosscutting Concept (CCC): - Structure and Function- Complex and microscopic structures and systems can be visualized, modeled, and used to describe how their function depends on the shapes, composition, and relationships among its parts; therefore, complex natural and designed structures/systems can be analyzed to determine how they function. Objectives Students - Identify scientific questions about biofilms. - Design and carry out an experiment investigating why biofilm bacteria resist the action of antibiotics. - Present experimental results to the class. - Utilize experimental results of the class to construct an explanation for why antimicrobial treatment often fails to cure infected knee implants.

Background Information (for teacher)	Extracellular Polymeric Substance (EPS) Extracellular polymeric substances (EPSs) are natural polymers of high molecular weight secreted by microorganisms into their environment. EPSs establish the functional and structural integrity of biofilms, and so determine their physical properties. EPSs are mostly composed of polysaccharides (long chains of carbohydrates) and proteins, but also macromolecules like DNA, lipids and other organic substances. Some of the common polysaccharides found in nature are starch, cellulose and chitin. Common polysaccharides of bacterial origin are xanthan gum and dextran... commonly found in foods, lotions, cleaners and paints. EPSs are the sticky construction material of bacterial biofilms and either remain attached to the cell's outer surface, and/or are secreted into its surroundings. These compounds are important in biofilm formation and cell attachment to surfaces. EPSs constitute 50% to 90% of a biofilm's total organic matter. Biofilm Antimicrobial Resistance Once a biofilm is established (with plenty of EPS surrounding the bacterial cells) it can be virtually impossible to eliminate. Yet why this occurs is complicated. It is the combination of the bacterial species involved (and their unique EPS molecules), as well as the type of antimicrobial substance utilized, that appears to determine whether the biofilm is able to resist destruction. Additionally, the biofilm bacteria and antimicrobial substance involved appear to determine <u>how</u> the resistance comes about. For instance, there are biofilms that prevent some antimicrobials from entering the biofilm at all. If the EPS shields the bacteria from the antimicrobial substance, the biofilm bacteria remain unharmed. On the other hand, some antimicrobials have no trouble penetrating an entire biofilm, and yet there are still bacteria within that survive to reform the biofilm. In these instances, it may be that the bacteria “deep” within the biofilm are in a dormant state (much like <i>hibernating</i>) due to limitations in nutrients, oxygen, etc. Alternatively, these resistant bacteria may turn on expression of stress response genes that aid in their survival. Scientists continue to investigate biofilm antimicrobial resistance, as well as novel techniques for treatment of biofilm infections.
Materials	For teacher: ➤ Projector and speakers ● (3) Empty 2-liter bottles ● (1) Bottle dextrin (Equate Clear Soluble Fiber Powder) ● (1) Bottle psyllium (Konsyl Fiber Original)

	• (1) Bottle guar gum (Benefiber Healthy Balance) • (1) Box food coloring (red, yellow, blue, green) For each student: ➤ Science notebook scaffold pages • Copy of <i>Infected Knee Implant Phenomenon</i> ➤ (optional) <i>Presentation Cue Cards</i> for non-presenting students For each group: • (5) Clear containers • (1) Dropper ➤ Any (teacher approved) classroom supplies
Prior Knowledge	Students should understand that cells are the basic building block of all living things (<i>Prior Knowledge/Lesson One</i>) There are multiple cell types (such as animal, plant and bacteria), yet all cells share some common core parts and functions. Dental plaque on our teeth contains bacterial cells (<i>Lesson One</i>). Communities of bacteria working together for survival create Biofilms. The production of Extracellular Polymeric Substance (EPS) helps the bacteria stick together, and helps adhere the biofilm to the surface on which it forms. The most effective method of removing a biofilm is scraping it off (<i>Lesson Two</i>). Biofilms are found just about everywhere, but it's the interplay between genetic factors ("needs") and environmental factors ("availability") that determine where an organism (like a bacterium) can survive (<i>Lesson Three</i>).
Launch (10-15 minutes)	
Phenomenon	Handout copies of the <i>Lesson Four Notebook Scaffolds</i> for students to paste into their notebooks, as well as copies of the <i>Lesson Four "Infected Knee Implant" phenomenon</i>. Ask students to make note of things that they <u>notice</u>, and things they <u>wonder</u>, as they watch the video and observe the infected knee implant phenomenon. Play the <i>Lesson Four Phenomenon Video</i> (1 min 56 sec) found on the unit USB. Provide students with a few minutes to think, and write notes, after watching the video.

	Form groups of 3-5 students. Groups should use a talking stick (one time around), and each student gets ~1 minute to share what they noticed and wondered. Display the <i>Lesson Four Scientific Questions</i> slide, and ask students to consider (silently) which of the displayed questions is “scientific”. Do not explain what constitutes a scientific question! Have students turn to a partner to share their thoughts, one at a time, or hold a dyad discussion. <u>Randomly</u> choose 1-2 students and ask them to share one of the questions they believe is scientific, as well as why. Then choose 1-2 students to share a question they believe is NOT scientific, as well as their reasoning. Finally, ask groups to think back to what they wondered about the lesson phenomena. Groups should discuss which wonderments are scientific (or could easily be rephrased scientifically), and why (talking chips recommended). Students will write the scientific question about infected knee implants they find most interesting, and then groups will craft a definition of what it means for a question to be “scientific”. Use the interesting questions and group definitions in determining when students are ready to move to the exploration.
Explore (30-40 minutes)	
Exploration	Explain that the class will be designing and carrying out experiments, as well as sharing results, in an effort to explain why biofilm bacteria resist treatment with antibiotics. Scientists are actively investigating this question, hoping that a better understanding of why bacteria are resistant will lead to effective treatments for biofilm-infected implants. Form groups of 2-3 students (supplies for 10 groups are provided with the kit). Each group will choose one variable to investigate, or you can form groups of students with similar interests. There is a list of suggested variables in the student notebook scaffold, but students should not be limited to these. Encourage creativity! <i>All groups will be observing the movement of a model antibiotic (food coloring) into a model biofilm (food grade polysaccharide as “EPS”).</i> Potential variables to test: • Depth (or “thickness”) of model biofilm layer • Concentration (or “amount”) of EPS within biofilm

- Different EPS substances
- Different antibiotics (different colors of food dye)
- Different concentrations (or amounts) of antibiotic

The kit includes 3 water soluble polysaccharides (EPS) to choose from, psyllium, guar gum, and dextrin. Additionally, there are 4 food dye colors to choose from (red, yellow, green and blue).

The kit contains supplies for up to 10 groups of 2-3 students.

Once each group has a variable chosen, they should work on designing their investigation. If your students are less familiar with designing experiments, consider showing the video “Control and Variable Groups” hyperlinked here: <https://www.youtube.com/watch?v=aLesk8fujH8>
After the video, you may want to discuss the use of more than two experimental set-ups/groups. *For instance, if you were conducting a hypothetical experiment on whether the addition of energy drinks effect plant growth, it would be advisable to set-up 3-4 plants that get different amounts of energy drink. What if ¼ cup a day supports growth, but ½ cup a day inhibits growth? Amounts can matter!*

Experimental Notes/Suggestions (for teachers)

Using food coloring to model antibiotic:

- Student groups dilute 1 drop of food coloring in 15 ml (1 Tbsp) water, then mix gently (swirling works fine).
- Layer a pre-determined amount of the diluted food coloring over each biofilm slowly to limit mixing.
- Droppers provided for layering food coloring (recommended). *Place the dropper tip on the side of the container, just above the polysaccharide, to minimize mixing.*
- White paper underneath/behind experiment aids visualization.
- Consider allowing groups to experiment with more concentrated food dye solutions, as needed.

Handling EPS (polysaccharide):

- All 3 polysaccharides are water soluble.
- If testing EPS concentration, diluting the stock solution is strongly recommended.
- Stock solution recipes (to be made ahead of time):

	○ Dextrin: 16.9 g (8.5 tsp) in 2 liters water ○ Psyllium: 50.7 g (8.5 tsp) in 2 liters water ○ Guar Gum: 21.2 g (5 Tbsp) in 2 liters water ● Cool tap water works fine, mix by shaking. ● Some settling occurs, encourage students to mix/shake stock solutions before removing what they need. It is highly recommended that you approve each group's design before proceeding to experimental set-up. Suggested items to check: ● Variable being tested ● Appropriate control(s) ● Experimental plan ● Data collection plan (what to measure and how) There is a place to initial each student's science notebook upon approval, if desired. Encourage students to record their plans, observations, data, and results in their science notebooks. Also discuss the fact that students will be presenting their results to the class and will need to refer to the poster and presentation rubric found on page 27 of the notebook scaffold.
Questions	Driving Question ● Why do bacteria form biofilms? Probing Questions ● How do biofilms help bacteria survive? ● Why are biofilms resistant to antibiotics? ● What benefits do bacteria experience by altering their environment with the production of EPS? ● Why is scraping the best way to remove dental biofilm from teeth?
Summarize (20 minutes)	
Communicate	Groups should display the results of their experiment on poster paper, clearly stating the variable tested as well as the control, and prepare for a short presentation to the class. The rubric for the poster and presentation are on page 29 of the notebook scaffold. Non-presenting students should take notes and ask questions. Specifically, students should be listening for the items in the check-off list located on page 27 of the notebook scaffold. Most importantly, students should

	record new data/results that would help in explaining why biofilm bacteria are typically resistant to antibiotics. Potential prompts for students include (and are available in the <i>Print Masters</i> for printing as “cue cards”): • Why did you choose to use ____ in your experiment? • What do you think ____ means, and why? • What are the implications of your results for biofilms? • Would you clarify ____ for me, please? • How did you test for (or analyze) ____? Posters should remain visible and accessible as students work on the assessment (below).
Terminology and Concepts to Solidify	Antimicrobial- an agent that kills microorganisms or stops their growth. Antibacterial- an antimicrobial agent that primarily targets bacteria. Concentration- the amount of a substance per defined space/volume. Dilution- when something is watered down or weakened. Diffusion- the net movement of anything (for example, molecules) from a region of higher concentration to a region of lower concentration. Variable- a factor(s) that differ between the test and the control samples of an experiment. Control- often referred to as a "control group", is a population or set of objects that is similar to the set being tested, but on which no changes are implemented.
Connection to Big Ideas (Phenomena)	When bacteria build a biofilm, they alter their environment (i.e.: by producing EPS) in order to ensure their continued survival. The exclusion of antimicrobial products from the deepest regions of the biofilm is one example.
(Optional) Follow Up/Practice	Borax-based “slime” is another interesting EPS model for investigation: https://littlebinsforlittlehands.com/how-to-make-borax-slime-easy/ This experiment challenges students to identify and test various coatings to see if they are capable of preventing or slowing the growth of biofilm on a surface. Probably best attempted when the weather is warm. https://thehomeschoolscientist.com/biofilm-experiment/#.VK6t0MIHCS0
Assessment	Look at the <i>Infected Knee Implant</i> lesson phenomenon again. Construct an explanation for why antimicrobial treatment often fails to cure infected

	knee implants. Include in your discussion how the bacteria benefit from building a biofilm... what's in it for them? Use the experimental results of your group, and the rest of your class, as evidence to support your answer. The use of labeled pictures, graphics, and other visual representations in your explanation are encouraged. Rubric on page 29 of the notebook scaffold.
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Lead-in to Lesson Five- At this point, students are likely wondering if ALL biofilms are bad. They may also be wondering if there are any tools, other than antibiotics, for treating/curing biofilm infections.

Lesson 5	Are All Biofilms Bad?
Overview Standard(s) & Objectives	Overview Students begin by observing a fake horror movie poster (<i>The Biofilm</i>) and flip through their <i>Lesson One</i> through <i>Four</i> notes collecting evidence to use in determining whether they believe all biofilms are bad, or not. Students then engage in an activity highlighting examples of beneficial biofilms and biofilm associated engineering solutions. Finally, students collaborate in the design of an object, tool, process or system addressing a real-world problem where either the problem, or the solution, relates to biofilm. Standards MS-LS1-5 Construct a scientific explanation based on evidence for how environmental and genetic factors influence the growth of organisms. 6-8-ETS1-2 Evaluate competing design solutions using a systematic process to determine how well they meet the criteria and constraints of the problem. Science and Engineering Practice (SEP): - Constructing Explanations and Designing Solutions- Apply scientific ideas or principles to design, construct, and/or test a design of an object, tool, process or system. Crosscutting Concept (CCC): - Cause and Effect- Cause and effect relationships may be used to predict phenomena in natural or designed systems. Objectives Students - Assess evidence from Lessons 1-4 before stating a claim in response to the driving question: <i>Are all biofilms "bad"?</i> - Investigate examples of beneficial biofilms and biofilm-associated engineering efforts, and then reassess the driving question. - Analyze engineering design solutions for cause and effect relationships, as well as relationships between genetic (<i>needs</i>) and environmental factors (<i>availability</i>). - Design an object, tool, process or system based on scientific ideas or principles addressing a real-world problem where either the problem, or the solution, relates to biofilm.
Background Information (for teachers)	Beneficial Biofilms Adapted from: https://www.cs.montana.edu/webworks/projects/stevesbook/contents/chapters/chapter001/section005/green/page001.html

Biofilms form and grow in practically every possible environment on earth. Obviously, then, not all biofilms are harmful. Many play an important role in the ecology of the earth and the sustainability of life in general. Microbes comprise the bulk of living biomass on Earth and play a major role in the cycling of elements. Most of those microbes live in biofilms on surfaces.

It is important to recognize that microorganisms, such as bacteria, that live in biofilms have evolved along with other organisms, including humans. While some bacteria produce effects that are bad for other organisms, most bacteria are harmless or even beneficial. When it comes to bacteria, higher organisms (like us) are just another environment to colonize.

Bioremediation

One of the best examples of a beneficial application of biofilms is in the treatment of wastewater. Engineers long ago developed a system that uses biofilm to process wastewater and sewage. They figured out that if you provide a surface (like a sand filter) and the desired bacteria, they will attach, form a biofilm, and eat the "bad" stuff in the water.

One of the less obvious beneficial applications of biofilms is in cleaning up oil and gasoline spills. The fact that some of the naturally occurring bacteria in soil and water can clean-up contamination (by using it as a carbon and energy source) is called bioremediation.

Bioremediation using biofilms is also used for cleaning up sites contaminated with other hazardous wastes. Making it work requires an understanding of biofilm processes. Engineers assess the genetic *needs* of the biofilm species they want to utilize and the environmental factors (or *available* resources) present at the contaminated site. If any environmental factors are missing at the contaminated site, engineers develop a process for incorporating them so that the beneficial biofilm bacteria flourish.

For more information, pictures and a short video about the Sanford Lab's Waste Water Treatment Plant, which uses biofilm:

<https://www.sanfordlab.org/feature/waste-water-treatment-plant>

South Dakota EPSCoR

The National Science Foundation created the Established (formerly Experimental) Program to Stimulate Competitive Research (EPSCoR) in 1979 because Congress recognized the uneven distribution of federal research and development grants. After World War II, federally funded

academic research grew dramatically, but national science policy at the time tended to funnel resources to a small number of centers of excellence.

This status quo ignored the dramatic growth in regional educational and research institutions. In every state, talented young people aspired to careers in science, technology, engineering and mathematics, but the nation wasn't profiting fully from the wealth of ingenuity and skill embedded across the country. EPSCoR provided a solution and is now a federal-wide initiative spanning five agencies including NASA, DOE, DoD, NIH and NSF.

Current goals of South Dakota EPSCoR (SD EPSCoR) include:

- Increase South Dakota's Science and Technology Research Capacity
- Provide educational opportunities for k-12, undergraduate, and graduate students
- Promote Science Based Economic Development for South Dakota

Legumes

A legume is a plant in the family Fabaceae (or Leguminosae); also called a pulse, especially in the mature, dry condition. Legumes are grown agriculturally, primarily for human consumption, for livestock forage and silage, and as soil-enhancing green manure. Well-known legumes include alfalfa, clover, beans, peas, chickpeas, lentils, soybeans, and peanuts.

Legumes are notable in that most of them have symbiotic nitrogen-fixing bacteria in structures called root nodules. The nitrogen-fixing bacteria convert atmospheric nitrogen (which most living organisms cannot utilize), and convert it into forms that are useful to other living things. In return for usable nitrogen (a major component of fertilizer), the plant shares food, in the form of sugars, with the bacteria. The structure of the root nodule also protects the nitrogen-fixing bacteria from predators in the soil that would otherwise prey on the bacteria.

A focus of the SD EPSCoR project, begun in 2020, focuses on increasing the nitrogen fixing capability of soybeans. Soybean is the second largest crop in South Dakota (5.6M acres planted in 2018). The first step in forming root nodules is the production of a biofilm on the soybean root. However, not all root nodules contain bacterial species capable of supplying high levels of nitrogen. Some are better than others are. Evidence suggests that soybeans and other legumes may supply more food to the root nodules providing the most nitrogen, and/or limit food to the inefficient nodules. Researchers with SD EPSCoR are working on increasing the number of highly efficient soybean root nodules to decrease the amount of fertilizer

	required for high crop yield. A focus of this research is increasing the ability/specificity of efficient nitrogen fixing species to form root biofilms, and thus root nodules. Biocorrosion Another facet of the 2020 SD EPSCoR research has to do with biofilm-based corrosion of pipes transporting oil, gas and water. The ability of some bacterial species to cause corrosion is called biocorrosion. Sulphate-reducing bacteria (SRB) are particularly problematic. SRB utilize sulphate instead of oxygen, and so are sometimes said to “breathe” sulphate. Organisms that can carry out respiration in the absence of oxygen are called “anaerobic”, and the liquid within gas, oil and water pipes is typically low in oxygen. Additionally, the bacteria deep within a biofilm experience an even lower oxygen environment. In fact, scientists have demonstrated that biocorrosion increases 10,000 fold when the responsible bacteria are in a biofilm. When bacteria like the SRB “breathe” sulphate, they excrete acid as a waste product. The acidic waste appears to cause the corrosion. The U.S. spends nearly \$4 billion a year to address these challenges. Though various coatings can delay the onset of corrosion, they typically fail. SD EPSCoR researchers hope to better understand the bacteria causing biocorrosion, and use Machine learning techniques to discover a range of potential pipe coatings (as well as predict the effect each coating will have on the bacteria) in hopes of lowering the cost associated with biocorrosion in gas, oil and water pipes. To learn more about South Dakota EPSCoR: http://sdepscor.org/
Materials	For teacher: ➤ Projector and speakers For each student: ➤ Science notebook scaffolds ● Copy of <i>Lesson Five Phenomenon</i> (movie poster) For each group of 4: ● Set of <i>New Biofilm Example Sheets</i> (Labeled A-D) ➤ (1) Large poster paper or <i>Lesson Five Summary Sheets</i> (A-D)
Prior Knowledge	Students should understand that cells are the basic building block of all living things (<i>Prior Knowledge/Lesson One</i>)

	There are multiple cell types (such as animal, plant and bacteria), yet all cells share some common core parts and functions. Dental plaque on our teeth contains bacterial cells (<i>Lesson One</i>). Communities of bacteria working together for survival create Biofilms. The production of Extracellular Polymeric Substance (EPS) helps the bacteria stick together, helps adhere the biofilm to the surface on which it forms, and protects the bacteria found within. The most effective method of removing a biofilm is scraping it off (<i>Lesson Two</i>). Biofilms are found just about everywhere, but it's the interplay between genetic factors (<i>needs</i>) and environmental factors (<i>availability</i>) that determine where an organism (like a bacterium) can survive (<i>Lesson Three</i>). Mature biofilms with plenty of extracellular polymeric substance (EPS) can be incredibly resilient and resistant to antimicrobial products. This is why physical removal of biofilms, coupled with antimicrobials, is most efficient for combating unwanted biofilms. Unfortunately, such as in the case of implanted devices, physical removal of a biofilm may not be possible, or may be highly undesirable (<i>Lesson Four</i>).
Launch (10 minutes)	
Phenomenon	Form groups of 4. A few groups of 3 are acceptable, if necessary. Display and pass out copies of the <i>Lesson Five Phenomenon</i> (displays a fake horror movie poster). Also, hand out the <i>Lesson Five</i> notebook scaffolds. Allow about 5 minutes for students to <u>silently</u> read the task found at the bottom of the phenomenon sheet (also on page 31 of the notebook scaffold), and flip back through <i>Lessons One-Four</i> in their science notebook ("touch every page"). Walk around the class watching for written responses supported with evidence. When students are ready, explain that when you call "show me", they should hold up the number of fingers that correspond with the answer of their choice: Prompt: Are all biofilms "bad"? 1. Yes 2. No Randomly call on a few students to share the evidence for their reasoning. Make sure to call upon students answering both ways, if applicable. Most importantly, do <u>not</u> assign "correct" or "incorrect" designations to student answers. Rather, allow students to develop their own understandings as the lesson unfolds.
Explore (20-30 minutes)	
Investigation	Biofilm Jigsaw Activity

	Continue using groups of four, these will be the “home groups” for the jigsaw activity. Hand each group a set of <i>New Biofilm Example</i> sheets, labeled A-D. Each student will choose one of the resources, and will serve as the group “expert” for that resource. Students should take 8-10 minutes to silently read the resource and contemplate the questions provided at the bottom of the resource. Note taking is encouraged. Next, form “expert groups”. Every student who analyzed resource “A” should gather for discussion, and every student who analyzed resource “B” should gather in a different location, etc. (<i>Expert group sizes of 4-5 students are optimal, but not always achievable. Consider forming two of each expert group if you have a large class</i>). Each expert group should attempt to come to consensus as they consider the associated questions. The use of a talking stick and/or talking chips is highly encouraged. Circulate among the expert groups, listening for any that might need you to ask guiding or probing questions to help them draw logical, truthful, conclusions. Reassemble the original “home groups”. Each student (or “expert”) will, one at a time, share their findings/learning with their team members. When not sharing, students should be listening attentively, and making notes... as every student is responsible for every piece of learning, even if they were not an “expert” on it to begin with. Groups should move to the summary (notebook scaffold page 33) when ready.
Questions	Driving Question - Are all biofilms bad? Probing Questions - How has your understanding of biofilms changed, if at all? - What are ways that scientists and engineers can use biofilms for “good”? - What is it that determines whether a biofilm is “good” or “bad”? Explain your reasoning!
Summarize (10-15 minutes)	
Communicate	Hand each group a large sheet of poster paper. If necessary, the <i>Lesson Five Summary</i> Sheets (A-D) may be printed and utilized instead. Have groups divide the poster paper into four quadrants: A. Examples of <i>bad</i> biofilms B. Examples of <i>good</i> biofilms

	C. Examples of biofilms <i>neither good nor bad</i> D. Questions we still have about biofilms Each student should use a different colored marker and silently, no talking yet, write a response to the prompt closest to them (4 quadrants, 4 students). Groups may either rotate the paper so that a new question is available for each student or move to the next prompt (i.e.: move the people, or move the paper). Students respond to this new prompt, and the process continues until the group has run out of responses, or time is called. Now groups may discuss, ask questions of one another, etc. Then, group members collaborate on responding to the summary prompts located on page 33 of the notebook scaffold: • What surprised you, if anything? • How has your understanding of biofilms changed, if at all? • Are all biofilms bad, or just some of them? Explain your reasoning! <u>Randomly</u> choose 3-4 students and ask them to share an “A-ha”, conclusion, or interesting observation from their group discussion. (Optional) Before showing the video, direct students to watch/listen for ways that bacteria are important for life on earth. Show the video Nitrogen Fixation - Seven Wonders of the Microbe World (4/7) (3 min 33 sec). Facilitate a discussion about the importance of certain “beneficial” bacterial biofilms. Ask students to think of other ways that humans rely on bacteria/biofilms for our health, wellbeing, and/or survival. <i>[for instance, food digestion relies on bacterial biofilm within our intestines, and human skin is coated with bacteria that compete with potential pathogens to help prevent skin infections. Students may also wish to discuss beneficial bacteria from the jigsaw resources, such as in degrading waste/recycling nutrients]</i>
Terminology and Concepts to Solidify	Bioremediation- the use of either naturally occurring or deliberately introduced microorganisms or other forms of life to consume and break down environmental pollutants, in order to clean up a polluted site. Legume- a type of plant. Most have symbiotic bacteria in structures called root nodules. The bacteria supply nitrogen in a form that most plants and animals can use. For that reason, they play a key role in crop rotation. Nitrogen-Fixation- the chemical processes by which atmospheric nitrogen is assimilated into organic

	compounds, especially by certain microorganisms as part of the nitrogen cycle. Enzyme- a substance produced by a living organism that acts to bring about a biochemical reaction. Can break a substance apart or join substances. Biocorrosion- corrosion caused by or enhanced by bacteria or other microorganisms, biologically induced corrosion. Machine Learning- the study of mathematical models that computer systems use to perform a specific task without explicit instructions, relying on patterns and inference instead; a subset of artificial intelligence (AI).
Connection to Big Ideas (Phenomena)	Not all biofilms are bad. There are multiple examples of beneficial biofilms that help humans and the environment. Additionally, engineers have designed some interesting solutions to problems involving biofilms, and there is currently much research in this area.
(Optional) Follow Up/Practice	Watch this short video (1 min) about deep underground biofilm research happening at the Sanford Underground Research Facility: https://vimeo.com/357614335 Students research other examples of beneficial biofilms and/or engineering efforts for solving biofilm related problems to share with the class.
Unit Assessment	Engineering Design Brainstorming The goal is to develop an object, tool, process or system addressing a real-world problem where either the problem, or the solution, relates to biofilm. First, reorganize groups of 3 or 4 based upon individual student interest. Potential real-world problems include: ● Dental plaque/cavities ● Knee implant infections ● Waste water treatment ● Pipe corrosion ● Crops/nitrogen requirements ● Any other teacher-approved issue Students will begin with about 10 minutes of individual think time as they attempt to imagine as many potential solutions/ideas as possible. Then, students will analyze and rank their ideas using the <i>Questions to Consider</i> found on page 35 of the notebook scaffold. One at a time, students will then share their top 1 or 2 ideas with their group. Each idea shared should

	include: • Description of the object, tool, process, or system • Brief explanation of why you chose this idea above any other(s) • Expected outcome (effect) when implemented (the cause) • Discussion of how genetic (needs) and environmental factors (available resources) are involved Next, groups will discuss which ideas are most promising, as well as how parts of proposed solutions could be combined to create an even better object, tool, process or system. Talking chips recommended. Final engineering design proposal Page 37 of the science notebook scaffold guides students as they compose their final design proposal. Final design proposals should be individual, to allow for individuality. Consider allowing non-written, alternative, presentation options to enhance student creativity! Final proposals should include: • Detailed description of the object, tool, process or system. • Brief comparison with at least 1 other idea highlighting the reason(s) you chose the proposed solution. • Explanation of how it works, including: • Expected outcome (the <i>effect</i>) when implemented (the <i>cause</i>). • Discussion of the relationships between genetic (<i>needs</i>) and environmental factors (<i>available resources</i>) involved.
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Print Masters

Biofilms- Stuck Like Glue, Stuck On You

Lesson One- What's On Our Teeth?

Note to students: The right-hand pages are blank for your notes, observations, answers to questions, sketches, etc.

Unit Phenomenon:

Driving Question

What's on our teeth?

Your Task

- 1- Closely observe the Lesson One phenomenon pictures.
- 2- Record things you notice.
- 3- Write down two questions that you have.
- 4- Share with a partner (follow your teacher's directions)

Assessment of Prior Knowledge:

When your teacher says "show me", hold up the number of fingers that match your answer choice:

Question 1: What are all living things made of?

- Answer choices:
- 1) Blood
 - 2) Cells
 - 3) Muscle
 - 4) All of the above
 - 5) None of the above

Question 2: What do all living things "do"?

- Answer choices:
- 1) Move around
 - 2) Use energy
 - 3) Breathe oxygen
 - 4) All of the above
 - 5) None of the above

Biofilms- Stuck Like Glue, Stuck On You

Lesson One- Exploration

Cork Phenomenon:

Driving Question

What is cork made of?

Your Task

- 1- Closely observe the magnified cork phenomenon picture.*
- 2- Record things you notice.*
- 3- Write down two questions that you have.*

As you watch the video- listen for answers to your questions.

- 4- Write down two new questions that you have after watching the video.*

Follow your teacher's directions for sharing with a new partner.

Exploration- Part B:

Driving Question

What is a system?

Your Task

- 1- Think silently for a moment:*

- What do the following 3 examples have in common?*
- What is the overall function of each?*
- What are the various parts of each?*
- How do the parts work together to carry out the function?*

****School System***

****Circulatory System***

****Justice System***

- 2- Follow your teacher's directions for sharing your thinking.*

- 3- After class discussion, write a final definition for "system".*

Biofilms- Stuck Like Glue, Stuck On You

Lesson One- Exploration (continued)

Driving Question

What is the overall function(s) of the cell?

Your Task

- 1- *Silent think time (1-2 minutes).*
- 2- *Your teacher will form groups.*
- 3- *Listen carefully to your teacher's directions for sharing and discussion!*
- 4- *One at a time, each group member shares his or her thoughts (~1 min).*
- 5- *Using talking chips, discuss and ask questions within your group.*
- 6- *Be ready to share your group's thinking with the larger class, and why!*
- 7- *Write in your science notebook what your class believes to be the overall function(s) of the cell.*

Jigsaw Activity:

Driving Question

How do the parts of cells contribute to their overall function?

Your Task

- 1- *Remain in your groups. This is your "home group" for the jigsaw activity.*
- 2- *Each group member selects a resource, there are 3 labeled A, B, and C.*
- 3- *Begin by silently reading your resource and contemplating the questions.*
- 4- *Follow your teacher's directions for forming temporary "expert groups".*
- 5- *Discuss your resource and decide how to answer the questions- ensure that EVERY student in the expert group contributes to the discussion!*
- 6- *Go back to your original home group.*
- 7- *One at a time, share your findings with the rest of your home group.*
When not sharing, listen attentively... taking notes is encouraged.

Product:

Collaborate with the other experts in your group as you craft, in your science notebook, two cell models: a plant cell model, and an animal.

Each cell part should be labeled, somehow, with name AND function...
Be creative!

Finally, compare/contrast plant and animal cells (written in your science notebook). How are they similar? In what ways do they differ?

Biofilms- Stuck Like Glue, Stuck On You

Lesson One- Summary

Driving Question

What do ALL cells have in common?

Your Task

- 1- *Silently read the "Bacteria" information sheet and compare bacterial cells with your plant and animal cell models.*
- 2- *Collaborate with your group members:*
 - *Create individual bacterial cell models. Be creative!*
 - *Compare/contrast bacteria, plant and animal cells.*
 - *Make a list of what ALL cells have in common.*
- 3- *Be prepared to share with the larger class.*

Homework

- *Consider all the ways you care for your teeth.*
- *What do you believe might be the purpose of each strategy?*
- *Ask your family and friends, outside of class time, about their own strategies and ideas before the next lesson!*

Assessment:

Driving Question

Is dental plaque "alive"??

Your Task

- 1- *Observe the lesson phenomenon pictures again. Scientists call this substance found on teeth "dental plaque". ... the round objects in the 17,500x magnification are bacteria!*
- 2- *Using what you have learned, do you consider dental plaque "alive"? Why or why not?*

What evidence do you have?

Be certain to refer to your cell models in support of your argument!

Product:

Convey your answer and supporting evidence in any format your teacher approves. Have fun and be creative!

Biofilms- Stuck Like Glue, Stuck On You

Lesson Two- What Is A Biofilm?

Remember to use the blank pages to the right for your notes →

Unit Phenomenon:

Driving Question

What is found in human dental plaque? A closer look!

Your Task

- 1- Closely observe the new dental plaque phenomenon pictures.*
 - 2- Record things you notice.*
 - 3- Write down two questions that you have.*
 - 4- Share with a group (following your teacher's directions)*
-

Think and Share:

Driving Question

How do people take care of their teeth? How/Why do these methods work?

Your Task

Think silently

- 1- How do people in your community care for their teeth?*
- 2- How/Why do you think these methods might work?*
- 3- Write your ideas to the right.*

Share with a group (according to your teachers directions).

Reading:

As you read the blog post, watch for:

- Various methods of dental care.
- Similarities among the different methods.

Look for similarities in 1) desired outcomes
 2) how they work.

Biofilms- Stuck Like Glue, Stuck On You

Lesson Two- Cavity Creator Game

Learning Intention: **Model the steps of biofilm development**

Your Task

1- Discover the steps of biofilm formation by collecting "Step Cards" as you pass the yellow spaces. Paste these into your notebook in the proper order as notes.

2- Learn about three biofilm bacteria commonly found in the human mouth when you collect the "Character Sheet" from your teacher. What step(s) in biofilm formation would each "character" be most likely to participate?

3- Record information obtained during game play that helps in explaining:

- The parts of a biofilm (bacterial characters and what they need/produce). What needs to be present for a biofilm to develop?*
- The steps of biofilm development (and, maybe, cavities). Is there a specific order in which events must take place? Why?*

*4- As soon as your group feels confident you understand the **parts** of a biofilm and the **steps** in biofilm development, check-in with your teacher to obtain the "Character Sheet" and begin developing your skit/play/dance (etc.) for modeling biofilm formation!*

Note: You do NOT need to reach the "Finish" space on the game board to check-in with your teacher and obtain the *Character Sheet*!

As You Play:

1- Make note of what helps you form a biofilm (moves you forward).

2- Notice the things that interfere with your biofilm's development (things that make you move backwards toward the "Start" space).

3- **Think:** Why do these things move your biofilm backward or forward?
How do they work? What might be the effect(s) on the parts of the biofilm?

Turn to the next page for game play instructions!

Biofilms- Stuck Like Glue, Stuck On You

Lesson Two- Cavity Creator (continued)

Cavity Creator Instructions:

Objective: *You and your team are playing bacteria found in the human mouth. You will need to work together if you are to achieve your goal: the formation of a protective biofilm... and maybe even a cavity.*

- 1- Place the game play token on the "Start" space.*
- 2- Shuffle the card deck and place it face down nearby.*
- 3- Cut out the "Step Cards", and place nearby to collect during game play.*
- 3- Place the "EPS tokens" nearby. Collect a token whenever your team lands on an  space. **EPS tokens may be used to lower the number of spaces moved backwards.** 1 token = 1 less space moved backwards. You may use more than 1 at a time.*
- 4- Take turns rolling the die and moving the specified number of spaces. Follow any directions found on the space where you land.*
- 5- Only collect "Step Cards" and "Cleaning Card" the first time you land on or pass over each yellow space on the game board.*
- 6- Play until your teacher calls time, **or you clearly understand:***
 - A- The parts of a biofilm (bacteria and things they need/produce)*
 - B- The steps of biofilm formation*
 - C- How dental hygiene methods (or lack of) effect biofilms*
- 7- Check-in with your teacher to obtain the "Character Sheet".*

Product:

Prepare a skit/play/dance (etc.) that models at least the **3 biofilm bacteria** (characters) and **5 steps** (Step Cards) of biofilm formation.

Be prepared to perform for your classmates next class period!

Be creative, but make sure you can explain everything you are showing, how it pertains to biofilms, and why you chose to show it that way.

Biofilms- Stuck Like Glue, Stuck On You

Lesson Two- Summary

Group Presentations:

- *As you watch each presentation, use your ordered Step Cards (5) and Character Cards (3) to track of the number of parts/steps portrayed.*
- *When your teacher calls "Show Me", hold up the number of fingers that equal the number of parts/steps that you believe were clearly shown.*
- *Eight fingers represents all required steps observed.*
- ***You may show more than eight fingers** for groups that go above and beyond to portray more than the minimum required parts/steps!*
- *Be prepared to justify your score and explain your reasoning.*

Think and Share (Part 2):

Driving Question

How has your understanding changed about how various tooth care methods work?

Your Task

Think back on your original ideas about how tooth care methods work:

- 1- How has your understanding changed, if at all?*
- 2- How do various tooth care methods affect biofilm?*
- 3- What do you think is the best way to avoid cavities? Why?*

Share with a group (according to your teachers directions).

Assessment:

Driving Question

Why do we care for our teeth the way that we do?

Your Task

- 1- Observe the Tooth Decay and How to Prevent It poster.*
- 2- Choose 2 of the 3 methods presented to analyze.*
- 3- Explain how each method limits/prevents human tooth decay (cavities).*

Pay particular attention to the structures found in biofilms, and their functions in aiding the microbial cells within.

Biofilms- Stuck Like Glue, Stuck On You

Lesson Three- Biofilms Are Everywhere!

Lesson Phenomenon:

Driving Question

Where else do biofilms exist?

Your Task

- 1- Closely observe the new biofilm phenomenon pictures.
- 2- Consider two questions that you have "in general" about biofilms.
- 3- Write each question on a separate scrap of paper (no name).
- 4- Follow your teachers directions for randomly sharing your questions...
You will get two new (anonymous) questions to read and contemplate.
- 5- Choose the one question (of your 4) that you consider most interesting.

Group Discussion:

Driving Question

What are the potential effects on organisms when placed into an environment that does not support their genetically encoded needs?

Your Task

- 1- Think silently- What are the environmental similarities and differences between the inside of your mouth, and the back of your hand?
(Environmental factors are things like light, water, temperature, etc.)
- 2- Each group member, one at a time, will share their thoughts.
- 3- Create a Venn Diagram or T-chart (etc.) to compare/contrast the factors "available" in each environment (mouth versus hand).
- 4- Next, discuss within your group whether you believe a bacterium from a human mouth would be able to survive (long-term) on the back of someone's hand. Support your ideas with evidence and reasoning, and ensure that **every** group member's ideas are heard!

Note: Each species of bacteria has it's own unique genetic material (DNA)... thus each species has unique genetic factors or "needs".

- 5- Write down your answer and reasoning: Would a bacterium from your mouth survive on the back of your hand? Support your answer!

Biofilms- Stuck Like Glue, Stuck On You

Lesson Three- Card Sort

Biofilm Bacteria Card Sort:

Driving Question

How do genetic factors determine the species capable of growing in a particular environment?

Your Task

- 1- Silently observe the Biofilm Descriptions and Bacterial Species sheets. Make notes as you consider which biofilm each bacterial species would most likely survive and thrive within.*
- 2- With a partner and a set of Card Sort supplies, attempt to come to consensus on where each species would thrive. Remember to consider the genetic factors ("needs") listed on the Bacterial Species cards- Which environment best provides what each species needs?*
- 3- Remember to explain the reasoning behind your ideas! There are many different ways to sort the cards- there is no wrong answer as long as you can back up your reasoning with logical arguments!*

Share and Compare:

- 1- You and your partner will share your sorting with a second pair.
- 2- Work with this second group to come to consensus. Focus on logical reasoning to ensure that each species is placed into the environment that best supplies it's genetically encoded "needs".

Suggested prompts for discussion:

- Why did you place _____ in biofilm __ and not Biofilm __?
 - If bacterium _____ requires _____, how does biofilm __ provide that environmental factor?
 - Couldn't _____ also find what it needs in biofilm __?
- 3- Generate an explanation: How do the genetically encoded "needs" of an organism determine where it is likely to survive and thrive?
 - 4- Follow your teachers directions for recording your final sorting.

Biofilms- Stuck Like Glue, Stuck On You

Lesson Three- Assessment

Assessment Prompt:

Driving Question

How do environmental and genetic factors contribute to the growth of organisms?

Your Task

- 1- *Carefully analyze the two grasses and three environments presented. Based on the environmental factors available in each location, and the genetic needs of the grasses, decide where each grass would most likely survive and thrive.*
- 2- *Clearly state where you placed each grass, as well as your reasoning. Make sure to address both environmental availability and genetic needs in your explanation.*
- 4- *Additionally, explain what you believe would happen (effect) to one of the grasses if it were placed into one of the other environments (cause).*

Clearly state which "other" environment and grass you choose, as well as what you believe would happen to the grass. Explain your reasoning!

Rubric: Check off when present

- ☐ *Best environment/location chosen for each grass clearly stated*
- ☐ *Reasoning for each placement supported with evidence (discussion includes genetic factors of grasses and environmental factors of locations)*
- ☐ *Choice of 1 grass and 1 "other" (not-best) environment stated*
- ☐ *What you believe would happen to this grass if placed into the "other" environment is clearly stated*
- ☐ *Reasoning behind what you believe would happen when this grass is moved to the other environment clearly explained.*

Biofilms- Stuck Like Glue, Stuck On You

Lesson Four- Why Do Bacteria Form Biofilms?

Lesson Phenomenon:

Driving Question

Why are biofilm bacteria resistant to antibiotics?

Your Task

- 1- As you watch the video and observe the phenomenon pictures, make note of things that you: a) Notice b) Wonder
- 2- Share with a group (following your teacher's directions)
- 3- Think: What is a "scientific question"? What makes scientific questions different from other questions?
- 4- Follow your teacher's directions as you consider which of the following is/are "scientific". Be prepared to explain your reasoning!
 - a) What is the relationship between sun exposure and skin cancer?
 - b) What is the best flavor of ice cream?
 - c) What factors cause dandelion flowers to bloom?
 - d) What is the effect of poor sleep on student test scores?
 - e) Should students who litter be suspended?
- 5- Group Discussion: Look back at your "I Wonder" statements about infected knee implants. Which seem "scientific", or could easily be rephrased as a "scientific question"? Why?
- 6- Which scientific question about infected knee implants is most interesting? Write that question below (or to the right):
- 7- Create a group definition for "scientific question":

Biofilms- Stuck Like Glue, Stuck On You

Lesson Four- Biofilm Experiment

Design Your Own Biofilm Experiment:

Driving Question

Why are biofilm bacteria resistant to antibiotics?

Your Task

1- Choose a variable to investigate. Some suggestions are listed below:

- Depth of EPS biofilm layer (modeled with polysaccharide in water)
- Concentration (or "amount") of EPS within biofilm layer
- Type of antibiotic (modeled with food coloring diluted in water)

2- Design an experiment to test how your chosen variable affects how far an antibiotic travels into a biofilm, and/or how quickly it travels.

Available Materials: Containers (4 for biofilm, 1 for food coloring)
Variety of polysaccharides to model EPS (3)
Food dye colors to model antibiotics (4)
Classroom supplies (as approved by your teacher)

3- Remember to include at least 1 control in your experiment.

4- Record your plan on the page to the right. Include in your plan:

- ☐ Variable being tested (the 1 thing that differs)
- ☐ Control
- ☐ How each experimental group will be treated (the plan)
- ☐ What you plan to measure, and how

5- Obtain teacher approval before beginning your experiment.

Teacher Initials

6- Carefully record all observations as you work.

Experimental Notes

Using food coloring to model antibiotic:

- Dilute 1 drop food coloring in 15 ml (1 Tbsp) water, mix gently.
- Layer diluted food coloring over biofilm *slowly* to limit mixing.
- Droppers provided for layering food coloring (recommended).
- White paper underneath/behind experiment aids visualization.

Biofilms- Stuck Like Glue, Stuck On You

Lesson Four- Biofilm Experiment

Biofilm Experiment (continued):

After running your experiment:

1- Sketch a visual representation of your results/data to the right.

2- What do your results tell you? Do you have information useful in explaining why biofilm bacteria are typically resistant to treatment with antibiotics? If so, then great! You are ready to create your poster and prepare your class presentation (rubric below).

If not, look back at your experimental design. Based on your results, how could you alter your experiment to obtain useful information?

Poster Presentation:

- Prepare a group poster and presentation to present to the class.
- Use the rubric below as a guide:

Poster:

- ☐ Variable and control(s) clearly stated
- ☐ Experimental plan/set-up sketched-out
- ☐ Data/results clearly displayed
- ☐ Statement explaining your result... what did you learn?

Presentation:

- ☐ Explain why you chose this variable and control(s) to test
- ☐ Explain why you ran the experiment the way you did (your plan)
- ☐ What do the results tell you? How do you know?
- ☐ If you were to run another experiment, what would you try?
Why?

Biofilms- Stuck Like Glue, Stuck On You

Lesson Four- Assessment

Assessment Prompt:

Driving Question

Why do bacteria form biofilms?

Your Task

- 1- Look back at the infected knee implant lesson phenomenon.*
- 2- Construct an explanation for why antimicrobial treatment often fails to cure infected knee implants.*
- 3- Include in your explanation why the bacteria are forming a biofilm... how do they benefit from living within a biofilm?*
- 4- Use the experimental results of your group, and the rest of your class, as evidence to support your answer.*
- 5- The use of labeled pictures, graphics, and other visual representations in your explanation is encouraged!*

Rubric: Check off when present

- ☐ *Explained why implant infection is so difficult to cure*
- ☐ *Explained why bacteria benefit from living in a biofilm*
- ☐ *Supported reasoning with data from class experiments*
- ☐ *Utilized visual representations where appropriate*

Biofilms- Stuck Like Glue, Stuck On You

Lesson Five- Are All Biofilms Bad?

Lesson Phenomenon:

Driving Question

Are all biofilms "bad"?

Your Task

- 1- Observe the Lesson 5 Phenomenon (fake movie poster).*
- 2- Think: Does this movie poster accurately reflect biofilms?*
- 3- Look back through your Lesson 1-4 notes for evidence.*
- 4- Compose a response to the driving question: Are all biofilms Bad?
What evidence do you have to support your claim?*
- 5- Be prepared to share your reasoning with the class.*

Jigsaw Activity:

Your Task

- 1- Each group member selects a resource, there are 4 labeled A, B, C and D.*
- 2- Begin by silently reading your resource and contemplating the questions.*
- 3- Follow your teacher's directions for forming temporary "expert groups".*
- 4- Discuss your resource and decide how to answer the questions- ensure that EVERY student in the expert group contributes to the discussion!*
- 5- Write notes about your resource (what you learned, what you find interesting, and answers to the questions).*
- 6- Go back to your original home group.*
- 7- One at a time, share what you have each learned. When not sharing, listen attentively and take notes... **Every student is responsible for all 4 resources!***
- 8- As a group, proceed to the Summary when ready (next page).*

Biofilms- Stuck Like Glue, Stuck On You

Lesson Five- Are All Biofilms Bad?

Summary:

1- Silent Analysis:

- *Each group will get a large sheet of paper.*
- *Divide the paper into 4 sections: Good Biofilms, Bad Biofilms, Neither Good Nor Bad Biofilms, and Questions We Still Have.*
- *There should be **no talking**.*
- *Each student writes ONE response in the section nearest them, then rotates so that students all have a new prompt.*
- *Look back through your notebook for ideas!*
- *Continue until your group runs out of responses to add.*

2- Group Discussion:

- *Now you can talk, ask questions of one another, and discuss the questions below:*

1- What surprised you, if anything?

2- How has your understanding of biofilms changed, if at all?

2- Are all biofilms bad, or just some of them? Explain your reasoning!

Biofilms- Stuck Like Glue, Stuck On You

Lesson Five- Assessment (Part 1)

Engineering Design Brainstorming:

Goal

Develop an object, tool, process or system addressing a real-world issue where either the problem, or the solution, relates to biofilm.

Your Task

1- Choose a problem to address:

- *Dental plaque/cavities*
- *Knee implant infections*
- *Waste water treatment*
- *Pipe corrosion*
- *Crops/nitrogen requirements*

2- Follow your teacher's directions for forming new groups.

3- Brainstorm potential solutions for addressing your chosen issue:

A- Individually, imagine as many potential solutions/ideas as you can.

*B- Use the **Questions to Consider** (below) to rank your ideas.*

C- One at a time, group members should share their top 1 or 2 ideas.

- *Describe the object, tool, process or system you envision*
- *Explain how you believe it would work*

D- Take notes as you listen to the ideas of your groupmates. Be considering which idea(s) might best address the issue.

4- Discussion:

*As a group, discuss which idea(s) are most promising and WHY. Also look for opportunities to combine parts of different proposals in the creation of an even better "new" idea. You do not need to come to consensus. **In the end, every student will have the opportunity to create and support their own, individual, design.***

Questions to Consider:

- ☐ *When implemented (the "cause"), what effect is expected?*
- ☐ *How will the genetic needs of the biofilm involved be met, or not met?*
- ☐ *Is it practical? Would it be easy to use?*
- ☐ *Would it be affordable, and therefore widely available?*
- ☐ *Does it have promise above and beyond current solutions, and so be worthy of development and testing?*

Biofilms- Stuck Like Glue, Stuck On You

Lesson Five- Assessment (Part 2)

Engineering Design Proposal:

Goal

Develop an object, tool, process or system addressing a real-world issue where either the problem, or the solution, relates to biofilm.

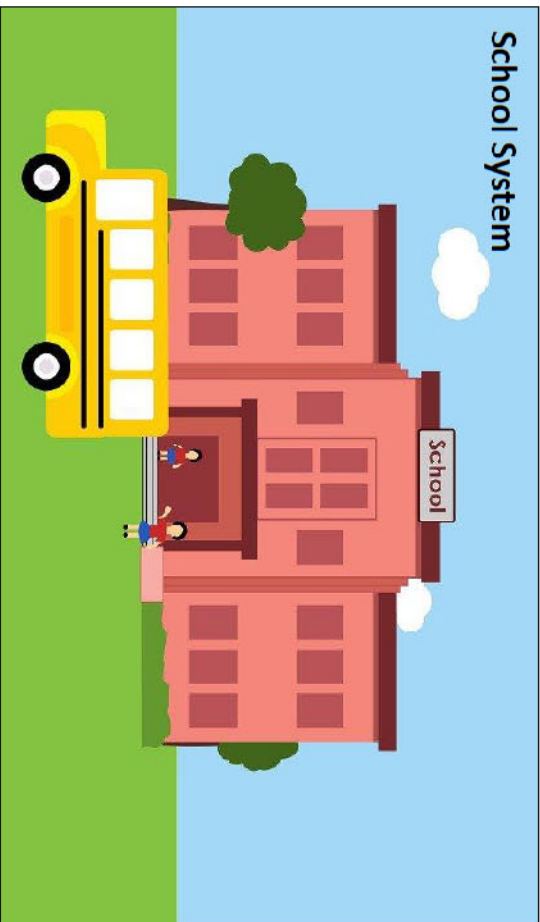
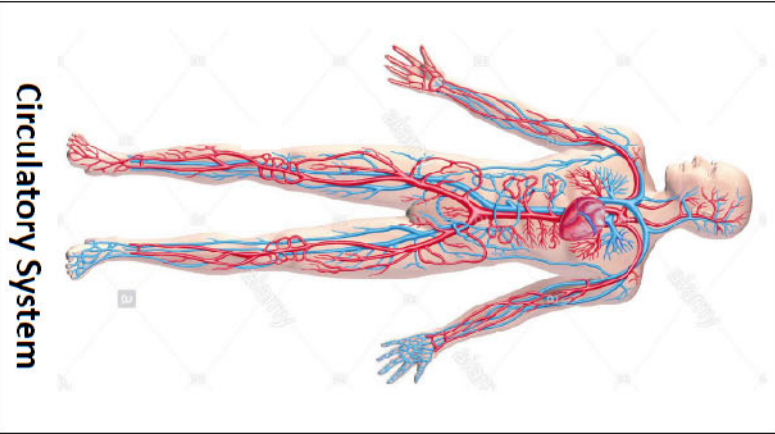
Your Task

- 1. After your initial brainstorming, sharing, and discussion, think back on the goal once more (above).*
- 2. Consider what YOU believe is the best solution for your chosen issue.*
- 3. Look back at the **Questions to Consider** (page 35) as you decide on ONE object, tool, process or system to propose and support.*
- 4. Use the checklist below as a guide while composing your response.*

Final Engineering Design Proposal Checklist:

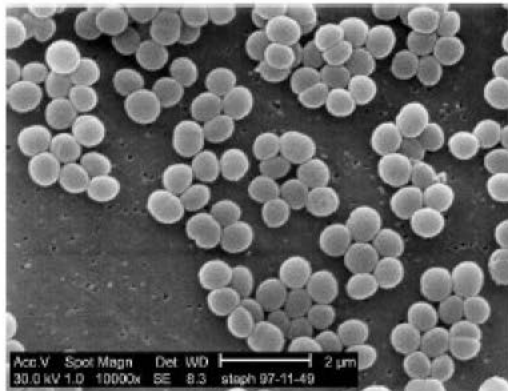
- ☐ *Detailed description of the object, tool, process or system*
- ☐ *Brief comparison with at least 1 other idea, explaining the reason(s) why you chose the proposed solution*
- ☐ *Expected outcome (effect) when implemented (the cause)*
- ☐ *Discussion of the relationships between genetic (needs) and environmental factors (available resources) involved in how your proposed solution works*

1. Overall function?
2. Parts?
3. How do the parts work together?

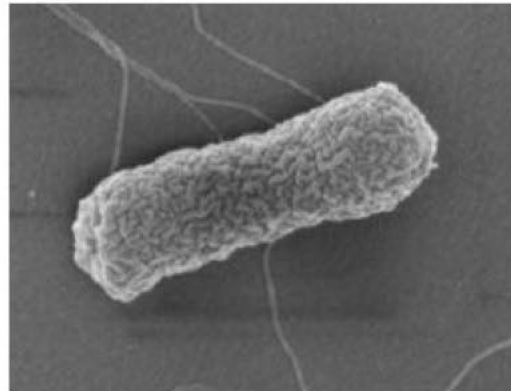


Bacteria

- Single celled organisms
- About the size of a mitochondrion and/or chloroplast
- Genetic material (DNA) NOT enclosed within a nucleus, enclosed by cell membrane
- Genetic material acts as “instructions” for making everything the cell needs
- Lacks mitochondria
- Uses cell membrane for energy generation- uses sugar for fuel
- Most have a cell wall beyond the cell membrane
- Many have special parts used for moving around called flagella or cilia
- 3 main shapes: rods, spirals and spheres



Magnified spherical *Staphylococcus* cells

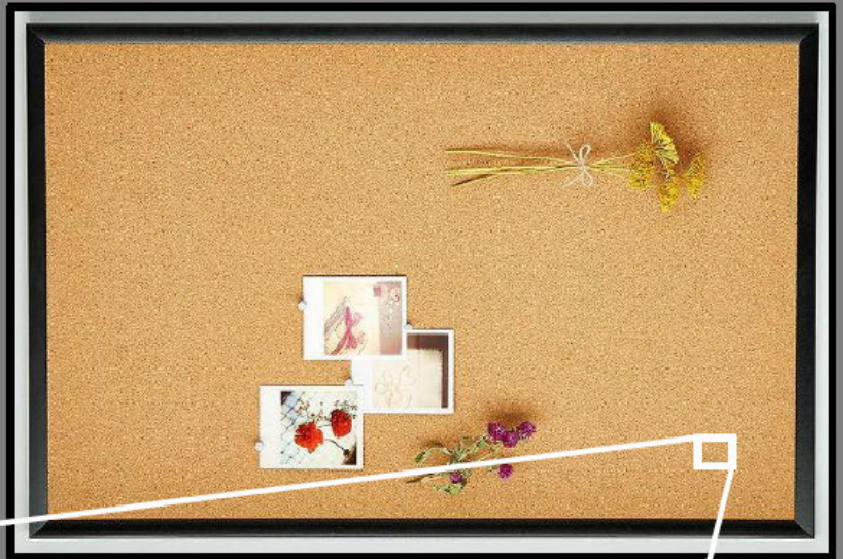


Magnified image of rod shaped *E. Coli* cell



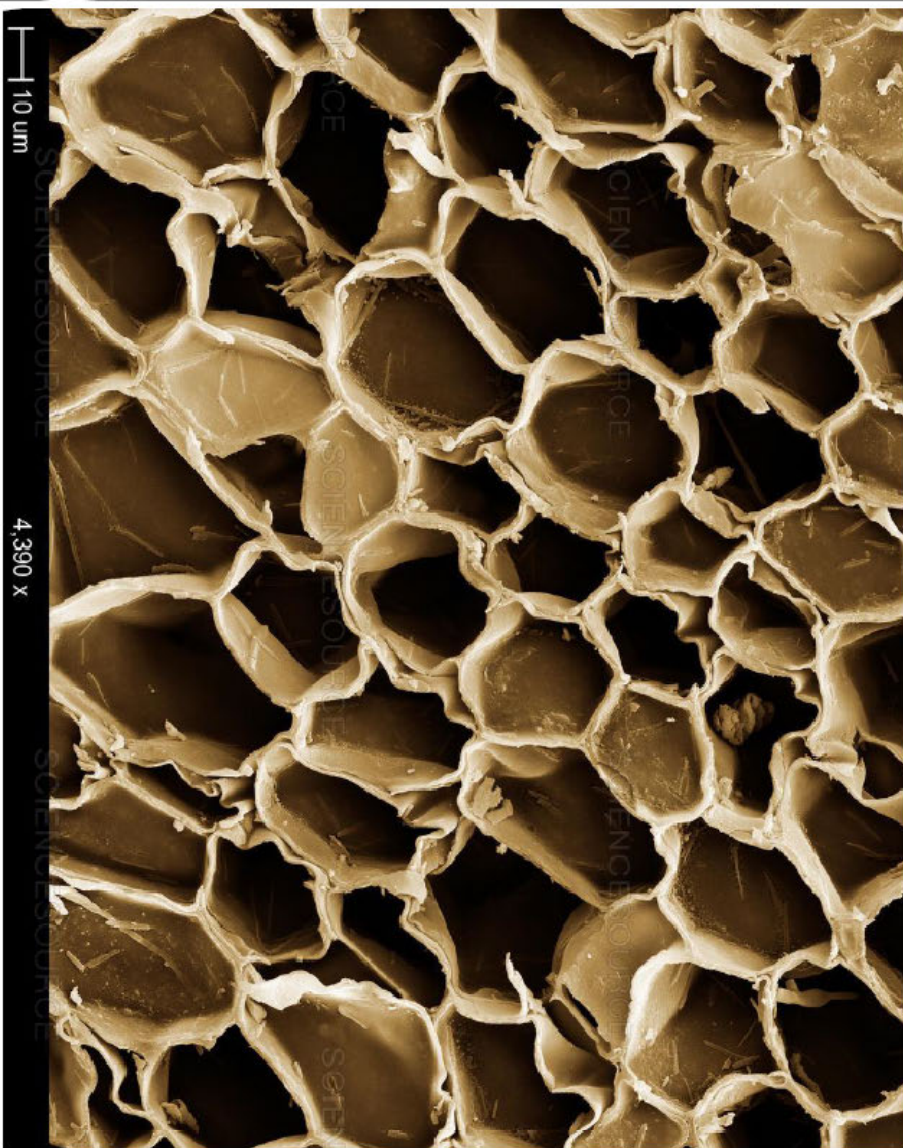
Magnified image of spiral *Treponema* cell

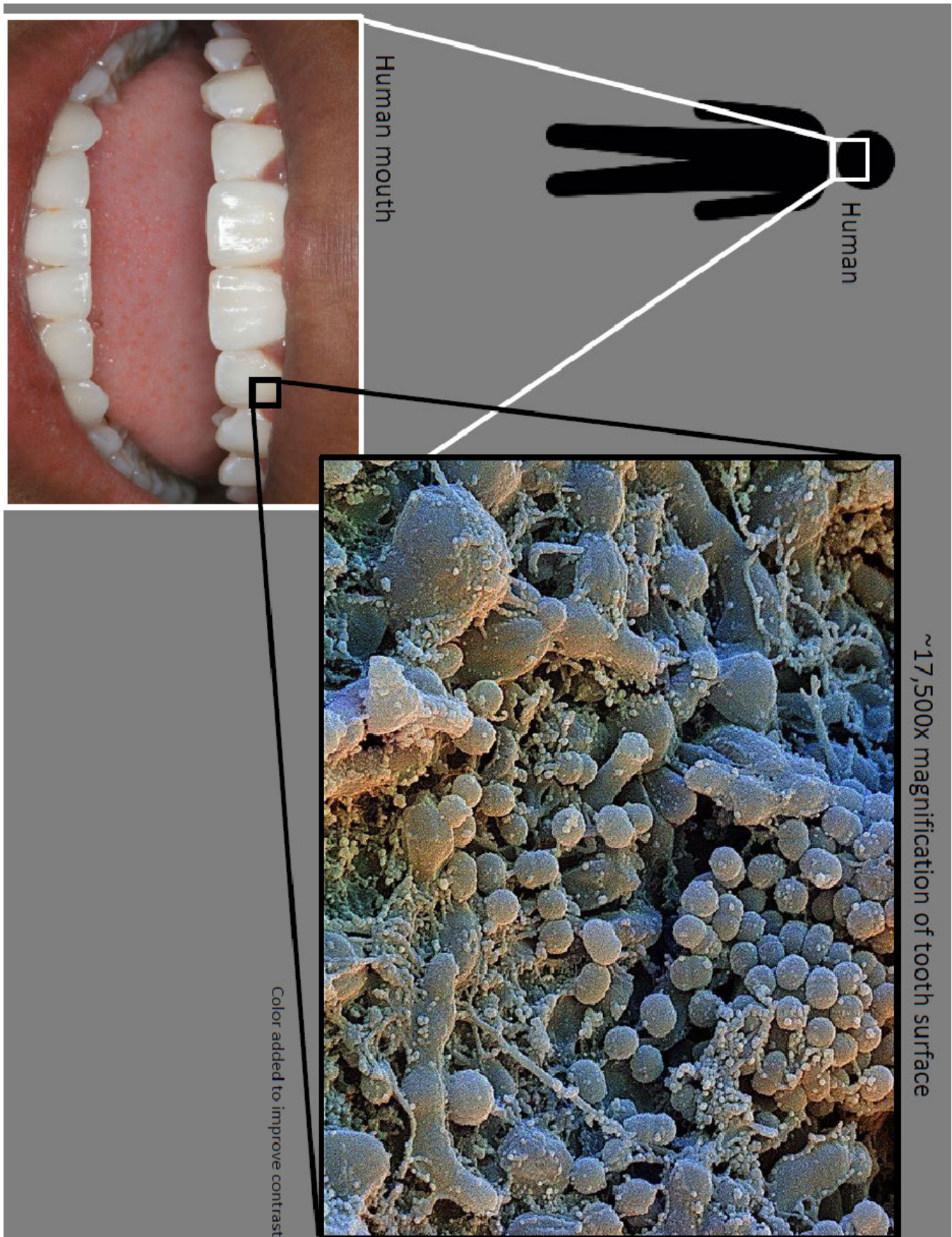
Cork Board



Observing cork with a microscope

4,390x magnification

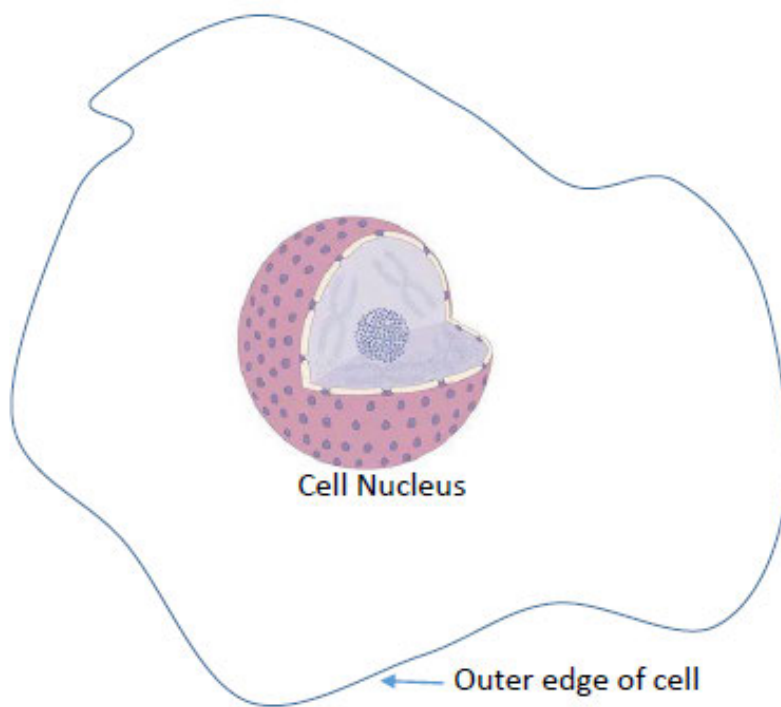




Nucleus



- Found inside both plant and animal cells
- Large spherical organelle (with a smaller sphere inside)
- Contains the genetic material (DNA)
- Genetic material acts as “blueprints” or “instructions” for the cell
- DNA determines what a cell can do, and also what it needs to “live”
- The largest of the organelles found inside plant and animal cells



Magnified image of nucleus
(color added for contrast)

Questions:

- What cell types contain a nucleus?
- What does the nucleus “do” that contributes to the overall function of the cell?
- Generate 2-3 new analogies to help explain the purpose of the cell nucleus.

Mitochondria vs. Chloroplast

Key Differences and Similarities

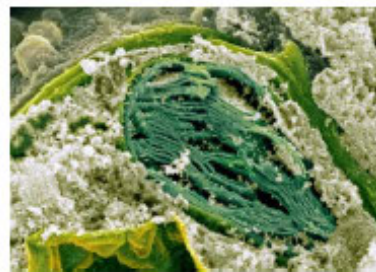
B



- | | |
|---|--|
| <ul style="list-style-type: none">• Found inside both plant and animal cells• Makes energy from sugar• Much smaller than the overall cell• Range in number from between 1 per cell, up to thousands• How living things get energy from food | <ul style="list-style-type: none">• Found inside plant cells• Produces sugar• Much smaller than the overall cell• Range in number from between 0 per cell, up to ~100• Where photosynthesis happens• Makes plants green |
|---|--|



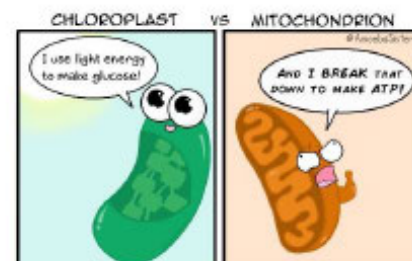
Magnified image of mitochondrion
(color added for contrast)



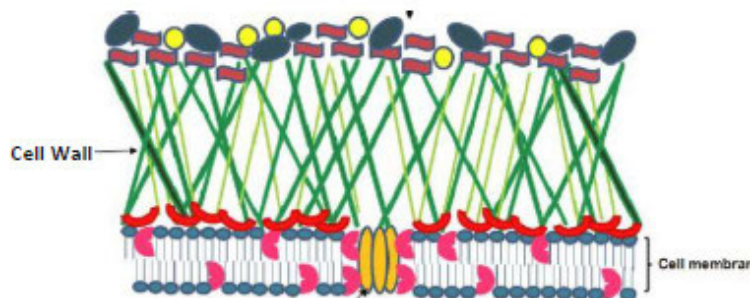
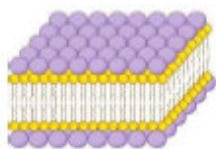
Magnified image of chloroplast
(color added for contrast)

Questions:

- What do these cell parts “do” that contributes to the overall function of the cell?
- How are these cell parts similar?
- How are these cell parts different?

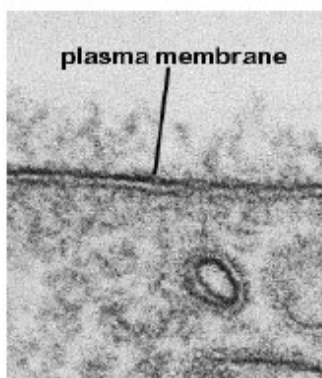


Cell Membrane -vs- Cell Wall

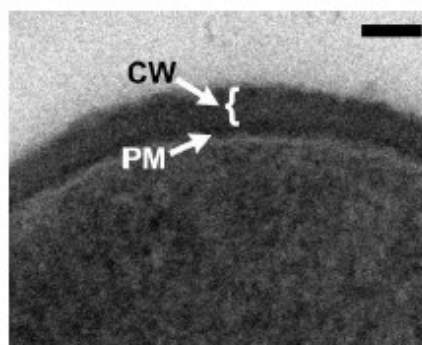


- Found surrounding both plant and animal cells
- Provides outer “boundary” of all cells
- Allows only certain things in/out of the cell
- Also called the “plasma membrane”

- Found surrounding plant cells
- Beyond (outside) the cell membrane
- Rigid structure, provides shape and support



Magnified image of cell membrane



Magnified image of cell wall (CW)
And cell membrane (PM)

Questions:

- What do these cell parts “do” that contributes to the overall function of the cell?
- How are these cell parts similar?
- How are these cell parts different?
- What might happen if these cell parts were absent?

C

Your host ate lots of
sugar today

**Move Forward
2 Spaces**

Your host didn't
scrape their teeth

**Move Forward
2 Spaces**

Your host didn't clean
between their teeth

**Move Forward
1 Space**

Your host scraped
their teeth extra well

**Move Backward
2 Spaces**

Your host used an
anti-microbial

**Move Backward
1 Space**

Your host had a
professional cleaning

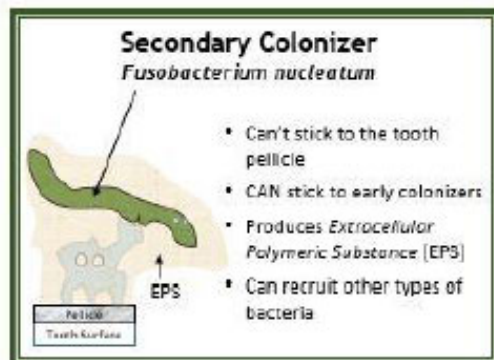
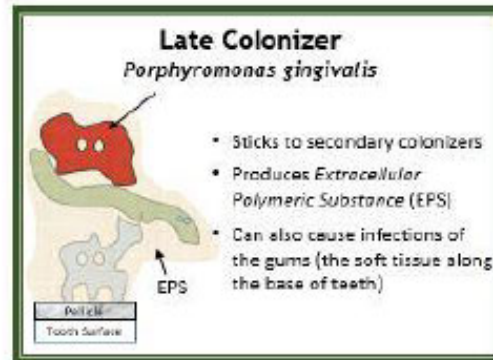
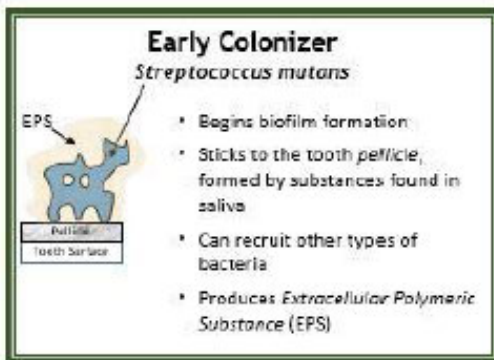
Go Back To Start

Your host ate little to
no sugar today

**Move Backward
2 Spaces**

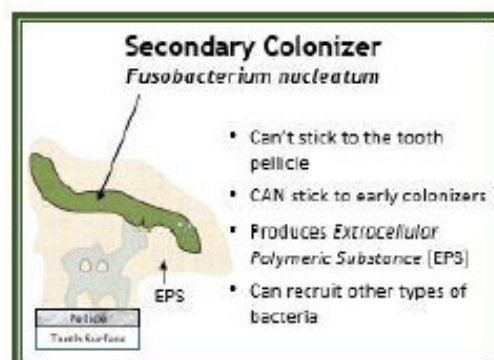
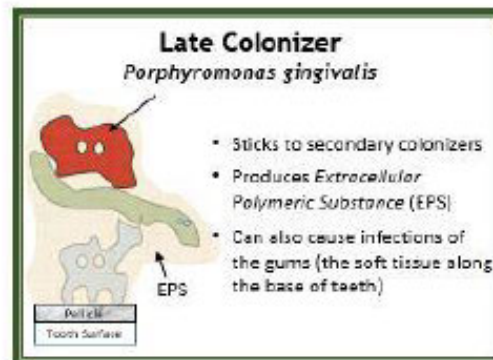
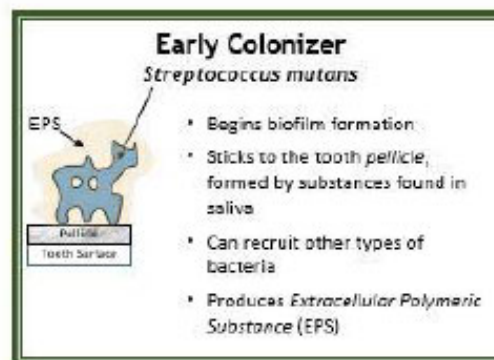
Your host cleaned
between their teeth

**Move Backward
2 Spaces**



Character Cards:

- Use these characters in your skit/play/dance
- Consider where in biofilm development each character is likely to participate
- How will you demonstrate each character's abilities in your skit/play/dance?



Character Cards:

- Use these characters in your skit/play/dance
- Consider where in biofilm development each character is likely to participate
- How will you demonstrate each character's abilities in your skit/play/dance?

Saliva Coating

The **pellicle** is a thin coating of saliva that coats teeth.

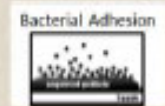
The pellicle contains compounds that some bacteria can stick onto!



Bacterial Adhesion

Some bacteria specialize in sticking, or **adhering**, to the tooth pellicle (saliva coating)...

They begin the biofilm!



EPS Production

Some bacteria excrete an **Extracellular Polymeric Substance (EPS)**... it is a sticky goo that holds the biofilm together, keeps it attached to the tooth, and protects the bacteria.



Cleaning Card

Currently, the only reliable way to remove a biofilm is to scrape it off. Professional tooth biofilm scrapers are called **dentists** and/or **dental hygienists**.

Professionals can scrape nearly ALL the biofilm from teeth!

Return to the START!

Mature Biofilm

Mature biofilms have LOTS of **Extracellular Polymeric Substance (EPS)**, and often contain many different types of bacteria living together in a **community**.

Some bacteria join the community by sticking to the bacteria attached to the tooth pellicle.



Biofilm Dispersal

When a biofilm gets large enough, bits break-off (or **disperse**) to start other biofilms in the mouth.





How Different Countries and Cultures Handle Oral Health Traditions and Trends

Your oral health is the window to your overall health. Hence, it's vital that you take good care of your mouth, teeth and gums. This way, you will be able to prevent tooth decay, bad breath and gum disease. But do people around the world have the same way of understanding the importance of dental health? In this post, let's explore how different countries and cultures take care of their oral health, and if they have unique oral health practices and traditions.

Replacing toothbrushes with the Miswak

You might be surprised to know that in many regions of the world, there are individuals who clean their teeth with twigs. These twigs are often obtained from neem and oak trees. The twig will then be broken into half, splayed and softened at the broken end. The "modified" twig is now ready for use, and the user will proceed to rub it on their teeth.

For Muslim and African cultures, the twigs they use naturally contain a high concentration of cavity-fighting fluoride. Also known as Siwak or the "Chewing Stick", the twig substitutes the toothbrush we are familiar with in the Western world. While the idea of not using a toothbrush may sound sketchy to some, studies have shown that the Miswak is better than toothpaste and can even prevent gum diseases by killing the bacteria that causes it. What's more, it helps eliminate bad breath and odors from the user's mouth, leaving a pleasant fragrance behind.

Different cultures have different perceptions of the tooth fairy

American Indian tribes like the Chippewa tribe, have parents that use charcoal to make their child's loose tooth turn black. This is done before the extraction, and once the tooth is removed, the tooth is thrown to the west. The child's grandmother will pray for the permanent tooth to grow in strong.

In China, Chinese children will place their lower teeth on the roof and upper teeth at the foot of their beds before going to sleep. They have the belief that this practice will make their permanent teeth grow in faster and stronger. Instead of a tooth fairy, children in El Salvador believe that a rabbit comes into their rooms during the night to take their tooth instead (leaving money too!).

TOOTH DECAY

AND HOW TO PREVENT IT

i Did you Know?
3 in 10 FIVE-YEAR OLDS
Have visible signs of tooth decay



So how can we prevent it?

Eat less sweets!
REDUCE OUR SUGAR INTAKE



No. 1

No. 2 Brushing our teeth
TWICE A DAY
MORNING AND NIGHT



No. 3 Regular Check-ups
VISIT OUR DENTIST



Brought to you by **BATTLE ROAD DENTAL PRACTICE**

Check out our site at www.battleroaddental.co.uk

Your host ate lots of
sugar today

**Move Forward
2 Spaces**

Your host didn't
scrape their teeth

**Move Forward
2 Spaces**

Your host didn't clean
between their teeth

**Move Forward
1 Space**

Your host scraped
their teeth extra well

**Move Backward
2 Spaces**

Your host used an
anti-microbial

**Move Backward
1 Space**

Your host had a
professional cleaning

Go Back To Start

Your host ate little to
no sugar today

**Move Backward
2 Spaces**

Your host cleaned
between their teeth

**Move Backward
2 Spaces**

Lesson 3 Assessment



Badlands National Park, South Dakota



- Elevation: 721-1,018 m (2,365-3,340 feet)
- Average Yearly Precipitation: 41 cm (16 inches)
- Temperature: ~12-33°C (11-92°F)
- Soil pH: 6.3-7.5 (slightly acidic to neutral)



Waubay, South Dakota

- Elevation: 554 m (1,818 feet)
- Average Yearly Precipitation: 61 cm (24 inches)
- Temperature: ~16-28°C (4-83°F)
- Soil pH: ~7 (neither acidic nor alkaline)



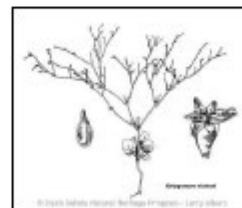
Rochford, South Dakota

- Elevation: 1,616 m (5,302 feet)
- Average Yearly Precipitation: 52 cm (20 inches)
- Temperature: ~11-28°C (12-82°F)
- Soil pH: 3.3-5.5 (acidic)



Grass #1: *Eriogonum visherii* (Dakota Buckwheat)

- Elevation: 575-825 m (1,886-2,707 feet)
- Yearly Precipitation: 36-43 cm (14-17 inches)
- Temperature: ~14-32°C (6.8-90°F)
- Soil pH: 5-8 (slightly acidic to slightly alkaline)
- Other: Tolerates salty soil



Grass #2: *Deschampsia cespitosa* (Tufted Hairgrass)

- Elevation: 0-4,300 m (Sea Level to 14,108 feet)
- Yearly Precipitation: ≥51 cm (at least 20 inches)
- Temperature: ~25-20°C (~13-68°F)
- Soil pH: 5.2-5.5 preferred (acidic), but tolerates 3.3-8.4
- Other: Tolerates toxic heavy metals and standing water



Bacterial Species

1

Acidithiobacillus ferrooxidans (*A. ferrooxidans*)

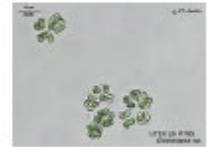
- May not tolerate sunlight well
- Fine with or without oxygen
- Tolerates high temperatures (up to 42°C/108 °F)
- Can use minerals like iron to make food



2

***Synechocystis* sp. PCC6803**

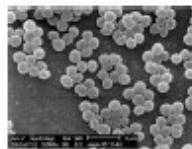
- Makes own food in daytime (photosynthesis)
- Produces oxygen
- Grows best ~23-38°C/73-100 °F
- Tolerates fresh and slightly salty water



3

Staphylococcus aureus (*S. aureus*)

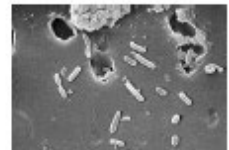
- Tolerates sunlight
- Fine with or without oxygen
- Grows best at 37°C/98.6 °F
- High salt tolerance
- Tolerates low nutrient availability



4

Pseudomonas aeruginosa (*P. aeruginosa*)

- Usually killed by sunlight (but not every cell)
- OK with or without oxygen
- Grows best ~37°C/98.6°F (OK up to 42°C/108 °F)
- High salt tolerance
- Tolerates low nutrient availability



5

Escherichia coli (*E. coli*)

- Can be killed by sunlight
- Fine with or without oxygen
- Grows best at 37°C/98.6 °F
- Tolerates both fresh and salty water, but does not tolerate high salt levels well



6

Vibrio cholera (*V. cholera*)

- Tolerates sunlight
- Fine with or without oxygen
- Grows best around 20-45 °C/68-113°F
- Prefers salty waters, but tolerates fresh when a little salt is present



Biofilm Descriptions



4,850 feet underground at the Sanford Lab (in man-made tunnel)

- No sunlight
 - Exposed to atmosphere (ventilation system)
 - Warm surface (rock ~36°C/96°F)
 - Fresh water with dissolved nutrients (from rock)
- Note: Water can be seen in picture squirting from the rock*



Infected wound

- Sunlight
- Salty water (occasionally cleaned with fresh water)
- Exposed to atmosphere
- High nutrients
- Warm surface (~37°C/98.6°F)



Infected Knee implant (biofilm stained black)

- No sunlight
 - Salty water
 - Not exposed to atmosphere
 - Nutrients limited
 - Warm environment (~37°C/98.6°F)
- } when implanted
Inside body



Streambed rock

- Sunlight
- Fresh water with dissolved nutrients (natural)
- Occasionally exposed to atmosphere
- Variable temperature



Used bathtub rubber ducky (cut open)

- No sunlight (until cut open)
- Fresh water (with nutrients from bath products and body oils)
- Variable temperature
- Variable exposure to atmosphere (fluctuates)

Biofilms Are Everywhere!



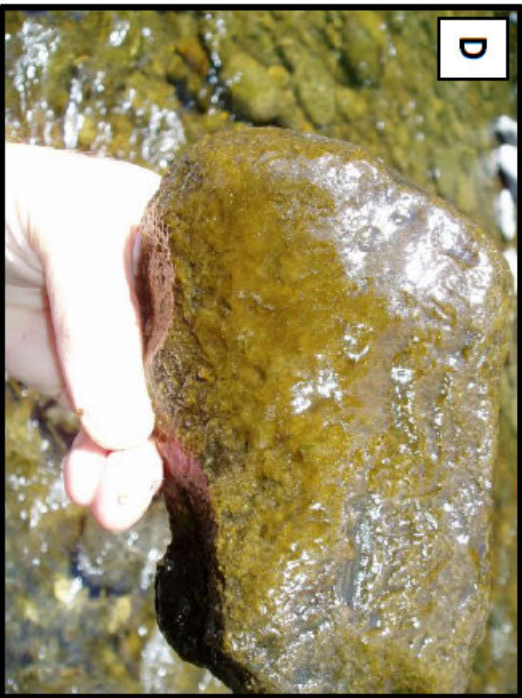
4,850 feet underground at the Sanford Lab



Infected wound



Infected Knee implant (biofilm stained black)



Used bathtub rubber ducky (cut open)

- Why did you choose to use ____ in your experiment?
- What do you think ____ means, and why?
- What are the implications of your results for biofilms?
- Would you clarify ____ for me, please?
- How did you test for (or analyze) ____?

- Why did you choose to use ____ in your experiment?
- What do you think ____ means, and why?
- What are the implications of your results for biofilms?
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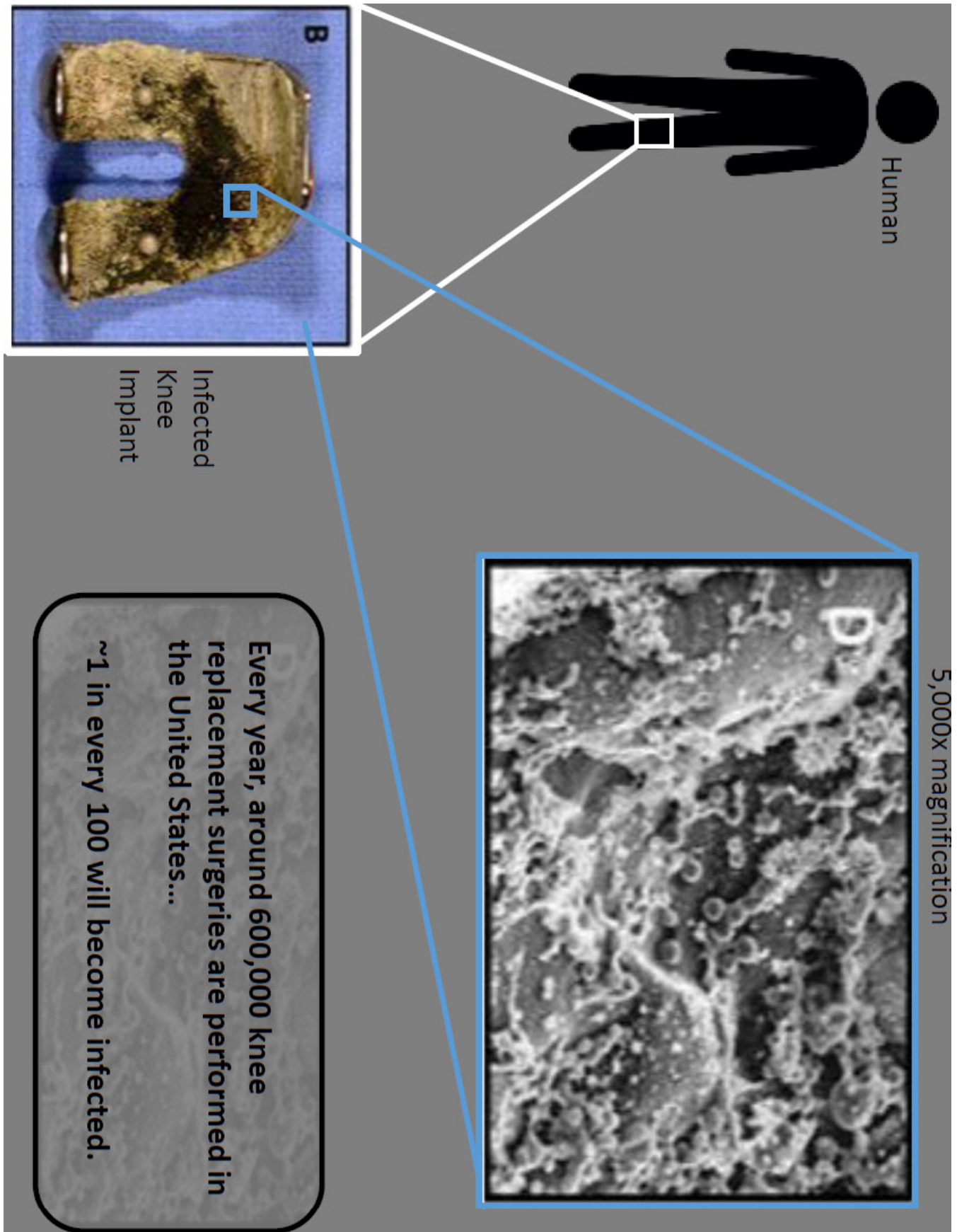
- Why did you choose to use ____ in your experiment?
- What do you think ____ means, and why?
- What are the implications of your results for biofilms?
- Would you clarify ____ for me, please?
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- What are the implications of your results for biofilms?
- Would you clarify ____ for me, please?
- How did you test for (or analyze) ____?



Which are “scientific questions”? Why, or why not?

- a)What is the relationship between sun exposure and skin cancer?*
- b)What is the best flavor of ice cream?*
- c)What factors cause dandelion flowers to bloom?*
- d)What is the effect of poor sleep on student test scores?*
- e)Should students who litter be suspended?*

Waste Water Treatment

A

The Sanford Underground Research Facility, or Sanford Lab, is a deep underground science facility in Lead, SD. The lab hosts science research in a variety of disciplines, and is home to the deep underground biofilm you observed in *Lesson Three*. Located almost a mile underground, in hot, dark, wet passageways created when the site was home to the Homestake gold mine, these bacteria are called *extremophiles*. Scientists hope to learn more about these organisms because of their unique characteristics: some “eat” methane (natural gas), others generate their own electricity, and they are all surviving in an extreme environment that scientists consider similar to places on other planets where bacterial life might be possible.

The Sanford Lab is also home to a unique waste water treatment plant (WWTP), constructed by the Homestake gold mine in the 1980’s. The WWTP was the first of its kind to use biofilm to clean up mining waste. These biofilm bacteria were found growing along the edge of a pond containing mining waste, so engineers designed the WWTP in such a way as to supply the environmental factors they required (i.e.: their *genetic needs*). The WWTP bacteria require a surface to grow on, water with dissolved minerals and other energy sources, warmth and oxygen. Therefore, engineers created rotating biological contactors (RBCs), to grow the waste-eating biofilm. The RBC’s are half submerged in warm water that contains contaminants (i.e.: food), and they slowly rotate. Because the RBCs rotate, the biofilm spends half of its time in the atmosphere (for oxygen), and the other half in the warm water absorbing minerals/contaminants/food. As the water flows from one RBC to the next (inside the long white buildings in the images below), it becomes cleaner and cleaner. This is an example of “bioremediation”, the use of living organisms to clean-up contaminants.

Because the RBCs are very large, heavy, and costly to operate, engineers are investigating ways to replace the RBCs with surfaces submerged in oxygen rich waste water. If successful, their efforts could greatly reduce the plant’s operating cost.

Image A- Sanford Lab’s WWTP



Image B- RBC housing and uncovered RBCs



Questions:

- Is this an example of a “good” or “bad” biofilm? What is your evidence?
- How are the biofilm bacteria’s needs met in the WWTP system; what do they require and how do engineers supply those requirements?
- When engineers developed the WWTP system (the “cause”), what effect was produced?
- How are scientists and engineers utilizing biofilm in current investigations? What do they hope to accomplish?

Soybeans

B

Soybeans are the second largest crop in South Dakota (5.6 million acres were planted in 2018). Soybean is a special type of crop called a “legume”, legumes have a special relationship with bacteria that take nitrogen from the atmosphere (unusable by most living things), and turn it into a form that plants and animals can use. These bacteria are called “nitrogen-fixers”. Legumes like soybean allow these nitrogen-fixing bacteria to live within their roots in “nodules”. The nodules protect the bacteria, and the legume even provides food for the bacteria. In return, the bacteria provide the plant with much of the nitrogen (fertilizer) it requires.

The first step in forming root nodules is the production of a biofilm, the nitrogen-fixing bacteria need to ‘stick’. The bacteria then enter the root (infect it), and the plant forms a nodule around them. Not all root nodules supply high levels of nitrogen, though, some are better than others. Evidence suggests that legumes may share more food with the “best” root nodules, and/or limit food to those providing less nitrogen.

South Dakota’s Established Program to Stimulate Competitive Research (SD EPSCoR) is a federally funded program designed to increase South Dakota science and technology research capacity, provide educational opportunities for students, as well as promote science based economic development within South Dakota. SD EPSCoR researchers hope to discover and/or create a substance that will increase biofilm development by the best nitrogen-fixers. If successful, their work could decrease the amount of fertilizer that South Dakota farmers apply to these crops... potentially saving significant amounts of money.

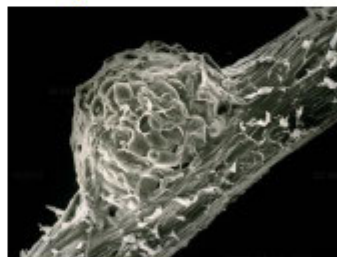
Image A- Soybean



Image B- Root Nodules



Image C- Inside Root Nodules



Questions:

- Is this an example of a “good” or “bad” biofilm? What is your evidence?
- How are the root nodule bacteria’s needs met in this system; what do they require and how do legumes/nodules supply these requirements?
- When legumes and nitrogen-fixing bacteria developed this association (the “cause”), what effect(s) was produced?
- How are scientists and engineers utilizing biofilm in their current research? What do they hope to accomplish?



EPS Cutting Enzymes

C

Adapted from: <https://www.scientificamerican.com/article/sticky-bacterial-biofilms-dissolve-on-contact-with-sugar-cutting-enzymes/>

To build a biofilm, bacteria secrete extracellular polymeric substance (EPS) into what becomes a defensive barrier around the bacteria- producing the microbial world's equivalent of "walled cities," according to biologist Perrin Baker of Toronto's Hospital for Sick Children. He and his colleague Lynne Howell have been working on methods to disassemble these defensive structures. "What we want to do is create chinks in the wall to allow the invading army in," Howell explains.

To make these chinks, the researchers turned to *Pseudomonas aeruginosa*—a bacterium known for building biofilms in the lungs of cystic fibrosis patients. This bacterium produces several different enzymes (substances that can "chew-up" other substances) that remove unwanted EPS so the bacteria can move around inside the biofilm. Baker and Howell wanted to see if they could turn these enzymes to their advantage. They first isolated two EPS-cutting enzymes from the bacteria, then added them to other biofilms to see what would happen. The researchers found that the enzymes destroyed most of the EPS produced by several different types of related bacteria. Even though the enzymes destroyed the EPS structure of the biofilms, the bacteria within were left unharmed. This leaves the majority of the biofilm bacteria exposed and vulnerable to antibiotics, and/or the host body's immune system.

Next, the researchers will test whether the enzymes can be used to coat hospital equipment, perhaps preventing the formation of antibiotic-resistant biofilm infections in the first place.

Image A- Petri dish Biofilms



Image B- Cartoon representation of research



Questions:

- Is this an example of a "good" or "bad" biofilm component/product? What is your evidence?
- What environmental "needs" of bacteria are removed from the biofilm environment by EPS-cutting enzymes?
- If EPS-cutting enzymes were placed on the surface of a knee implant before surgery (the "cause"), what effect(s) might you expect?
- How are scientists and engineers utilizing biofilm in this research? What do they hope to accomplish in future research?

Pipeline Corrosion

D

The U.S. spends nearly \$4 billion each year fighting biofilm-caused corrosion in metal pipes transporting oil, gas and water. This includes many drinking water lines for human consumption. Though the bacteria responsible do not necessarily need to be in a biofilm to cause corrosion, the formation of a biofilm increases corrosion 10,000 fold. This ability of a living organism to cause corrosion is called “biocorrosion”.

Currently, nonmetallic coatings inside metal pipes can delay this corrosion. Yet, even when corrosion is delayed, the nonmetallic pipe coatings eventually fail. One reason for this failure is the ability of the biofilms to degrade the coatings, and/or their ability to invade tiny cracks within the coatings. Though researchers are still investigating how/why various bacteria cause corrosion of metal pipes, it appears to be at least partially due to bacterial waste products. Specifically, acids the bacteria excrete (think microbial “poop”). Some of these biocorroding bacteria produce acid in the presence of sulfur when there is little to no oxygen. These conditions can be found inside pipes filled with sulfur-containing liquid, and/or deep within a biofilm.

South Dakota’s Established Program to Stimulate Competitive Research (SD EPSCoR) is a federally funded program designed to increase South Dakota science and technology research capacity, provide educational opportunities for students, as well as promote science based economic development within South Dakota. SD EPSCoR researchers hope to better understand the biofilm bacteria involved in this corrosion, as well as understand how they interact with different pipe coatings. They also plan to use Machine learning techniques (“artificial intelligence”) to identify potential coatings, even predict the effect each will have on biocorrosion. In this manner, they hope to discover new and improved coatings to lower the economic burden of biocorrosion on our oil, gas and water pipelines.



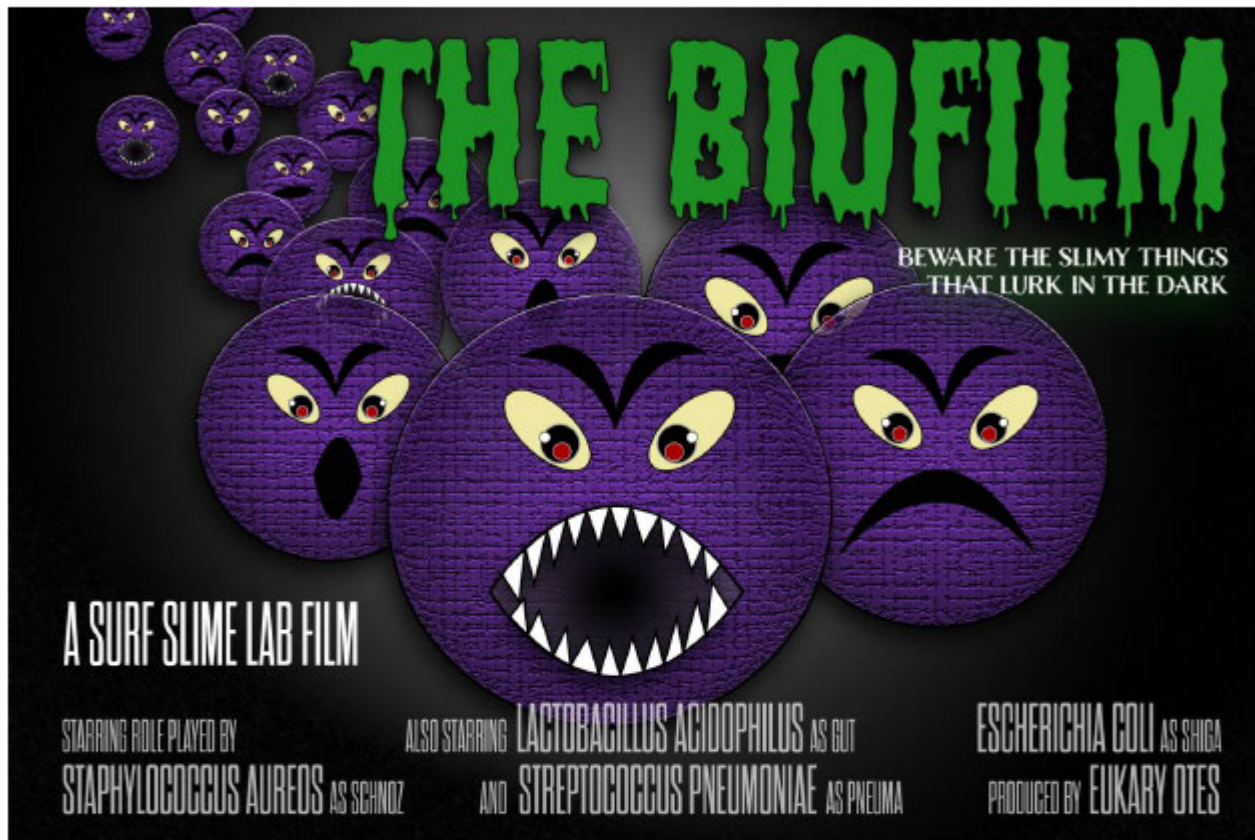
Questions:

- Is this an example of a “good” or “bad” biofilm? What is your evidence?
- How are biocorroding bacterial needs met in this system; what do they require and how does the pipe environment supply those needs?
- When engineers coat metal pipes with non-metallic substances, what effect(s) is produced?
- What are scientists and engineers hoping to accomplish with this research? How do they plan to gather the information they require?



Biofilms- Stuck Like Glue, Stuck On You

Lesson Five



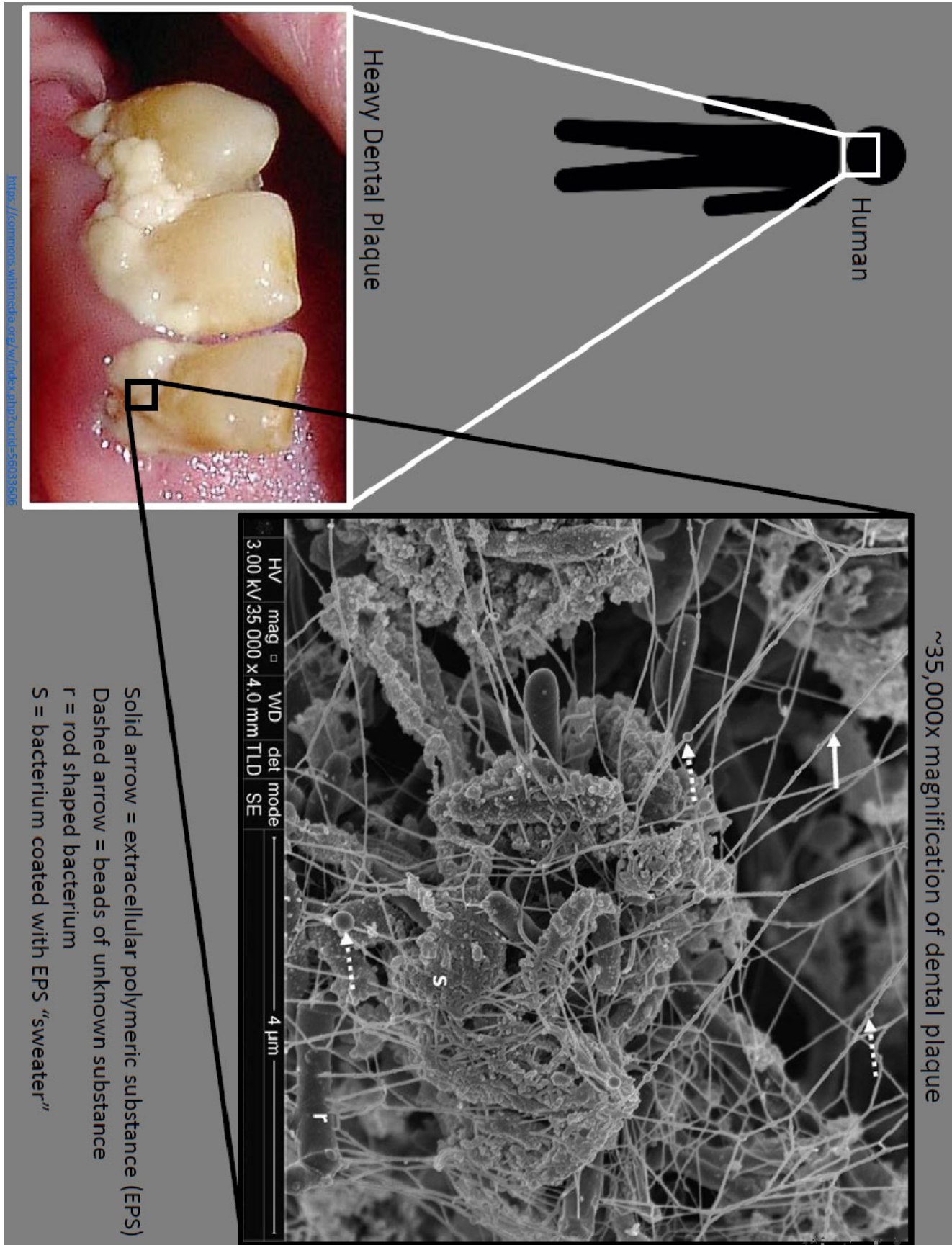
Artwork by Erica Popelka (BHSU Advanced Graphic Arts student) 2020

Driving Question:

Are all biofilms “bad”?

Your Task:

- Observe this fake horror movie poster
- Think: Does this movie poster accurately reflect biofilms?
- Look back through your Lesson 1-4 notebook pages for evidence
- Compose a response to the driving question: *Are all biofilms bad?*
- What evidence do you have to support your claim?



Biofilms – Stuck On You, Stuck Like Glue Refurbishment Inventory

[illegible]

NOTICE: THE HEADER/FOOTER AND REVISION HISTORY OF THIS ATTACHMENT IS NOT INCLUDED IN THE PRINTED DELIVERABLE. ANY BLANK PAGES ARE LEFT SO INTENTIONALLY.

Revision History

Rev	Date	Section	Paragraph	Summary of Change	Authorized by
01	2/27/2024	NA	NA	Initial Release	CCR 911