



EYE

Extreme Yellowstone Expedition

LIMITS OF LIFE

LESSON 2

Instructor's Key



MONTANA
STATE UNIVERSITY

Thermal
Biology
Institute



BRIGHTLY COLORED *Thermochromatium* BACTERIA, found in small springs in the Mammoth area of Yellowstone National Park, are examples of extremophiles. They thrive in hot conditions that humans and other life forms would find extremely inhospitable.

Living on the Edge...

In the past few decades scientists have discovered that life can thrive in places we never imagined it could—such as hundreds of feet below ground, underneath glaciers, at the bottom of the ocean, and in boiling hot springs. Life is a lot tougher than we thought.

Life in extreme conditions

Now we know that there are many forms of life that thrive in environments that we think of as extreme: salty, acid or alkaline, very hot or cold, poisonous, high or low pressure, and even radioactive. Organisms that live in these places are called **extremophiles**, and the environments they live in—what we consider extreme on Earth—might be similar to what is normal on other planets or moons. Many scientists think that if we find life elsewhere in the universe, it may resemble these organisms living in Earth's most extreme environments, not the little green men often shown in cartoons and movies.

They're not extremely rare

When we talk about life in extreme environments, you might think these life forms are rare, because we think of extreme things as being uncommon. Remember, though, that what we call extreme just means places where we couldn't live. Humans (and most plants and animals) only thrive in a narrow range of temperature, pressure, salinity and pH.

There are a lot more extreme environments than one would think. In fact, extremophiles are present all around us. Most of them are single-celled organisms and many are in the domain Archaea.

What's a domain?

Now you are probably wondering, *What's a domain? And what's Archaea?*

When scientists arrange living things into groups that have similarities, that is called **taxonomy**. The broadest category that scientists use is called a **domain**. Scientists group all living things into three domains:

Eukaryota - plants, animals, fungi and some single-celled organisms

This is the only domain that contains life with multiple cells that work together to do different jobs.

Bacteria - single-celled organisms that lack a membrane-bound nucleus

They live all over the Earth—from deep inside rocks to inside the intestines of most eukaryotes.

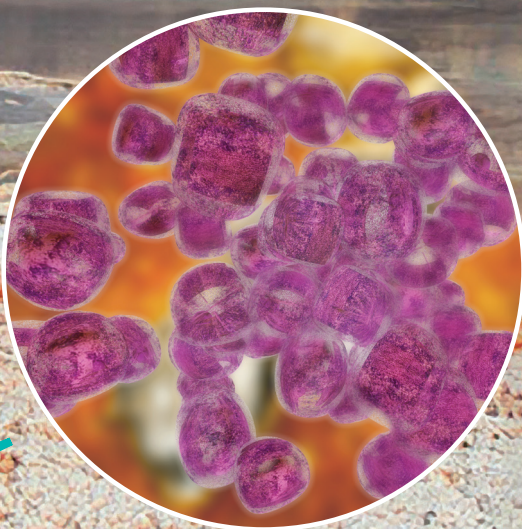
Archaea - single-celled organisms that lack a membrane-bound nucleus

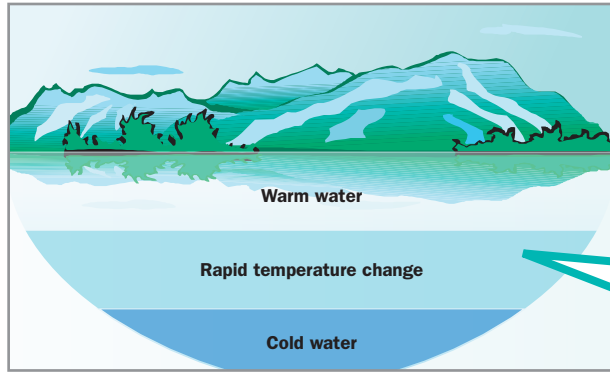
They were originally categorized as Bacteria, but now scientists realize their cells have unique properties separating them from the other two domains of life: they have a different genetic and chemical make up. Many thrive in extreme environments and they evolved in early Earth's harsh environment (hot with no oxygen).

What are the limits of life?

Researchers are working to establish what the environmental limits are for life to exist. Establishing these limits will help scientists in the search for life on other planets and in the search for as yet undiscovered life on Earth.

All this brings us to our next point: organisms function well (thrive) only within particular ranges of an environment. No single organism could survive the entire range of surface temperatures of Earth, but there are certainly those adapted to very hot climates, while others are adapted to very cold





ENVIRONMENTAL GRADIENTS

An environmental gradient can be a smooth, even change or a more sudden change. This is an example of a basic temperature gradient in a mountain lake. The water gets colder as you go deeper into the lake. Gradients of water pressure and oxygen in the lake would follow a similar pattern. As you go deeper, oxygen levels would diminish, but pressure would increase.

climates. Every species has its “preferred” temperatures, acidity, moisture levels, nutrients, and so on. Some species can tolerate a broader range of some of these **gradients**; others have a very narrow survival range.

The ranges that an organism can tolerate help define its **niche**—where it can live, and with what other organisms. Human beings are unique in that we have developed technology that lets us expand our niche and live in places that we couldn’t even visit otherwise.

Finding an organism’s niche

Here’s one way to help find an organism’s niche. When we take measurements of any **abiotic** (non living) factor along some geographic or time axis and see a change (gradual or sudden) in the value, we are measuring an **environmental gradient**.

Gradient just means “a continuous change in a value.” An environmental gradient can be a smooth, even change (for example, pressure varies predictably with water depth) or a more sudden change (like air pressure before a storm).

Gradients can be found on the land and in the air as well. At higher altitudes on a mountain, the average temperature drops significantly, creating a climate gradient on the mountain’s slopes. For some mountains, the base can be a tropical rainforest while the top resembles an Arctic tundra. Gradients can be invisible or they can be marked by visible changes such as color or vegetation.

Measuring environmental factors

We can actually measure almost anything in the environment, but let’s focus on things that affect life the most.

One obvious measurement is temperature. The Earth’s surface experiences temperatures from about -50°C up to 60°C (about -180°F to 130°F), and most life we’re familiar with can’t survive much outside that range.

Another measurement is humidity (amount of water vapor in air), ranging from nearly 0% up to 100%. In the oceans, we could measure acidity (on the pH scale, from just below 0 up to 14.0), or turbidity (how “muddy” the water is).

We could also measure how much oxygen or CO_2 is in the air. The average concentration of CO_2 was about 396 PPM (parts per million) in 2013, up from less than 320 PPM in the 1960s.

Each of these values can vary, depending on exactly where in the environment we do our measurement. For example, we might measure ocean water temperature at different depths, and we’d find that ocean water becomes colder as we go deeper.

Take a beach for example

A beach is an example of a location with many environmental gradients. In particular, we have the “wetness” of the land: Beyond the tide line is ocean environment, then we have the wet sand where the tides wash in, then the drier beach sand, and finally the dunes.

We can see distinct areas here, each with its characteristic forms of life. These identifiable areas along gradients are called **zones**, and the process of forming them is called zonation. Each zone has distinct lifeforms. For example, we would rarely find hermit crabs or clams in the dunes area, while beach grass can’t take hold in the intertidal zone.

ACTIVITY 1

Identifying Gradients

Analyze this photo looking for gradients.



ANSWER KEY

The following are some possible answers. There are no absolute correct or incorrect answers. This is an exercise to gauge your students' understanding of the concept of gradients, not assess the precision of their answers. *The italicized words in the first row of the table are given as an example in student workbooks.*

Write in the table below a few of the types of gradients represented in this landscape. (Note that some might be visible and some might be invisible.) The first row in the table shows one example answer, but the gradients, zones and life forms you identify may differ from the example.

	Kind of gradient (i.e. temperature, moisture, etc.)	How many zones of the gradient do you see?	List the different zones you see.	Name a plant or animal that could live in each zone.
1.	<i>moisture</i>	5	1. ocean 2. tidal zone 3. dry sand 4. grass dunes 5. air	1. fish, seaweed, shark, dolphin, bacteria, microbes etc. 2. clams, mollusks, fish, starfish, microbes, etc. 3. crab, insect, microbes, etc. 4. snake, tortoise, bird, microbes, etc. 5. bird, insect, microbe, etc.
2.	temperature	6	1. ocean 2. tidal zone 3. dry sand 4. grass dunes 5. shaded areas of grass dunes 6. air	1. fish, seaweed, shark, dolphin, bacteria, microbes, etc. 2. clams, mollusks, fish, starfish, microbes, etc. 3. crab, insect, microbes, etc. 4. snake, tortoise, bird, microbes, etc. 5. snake, tortoise, bird, microbes, etc., 6. bird, insect, microbe, etc.
3.	pressure	2	1. in the water 2. air	1. fish, seaweed, shark, dolphin, bacteria, microbes, etc. 2. bird, insect, microbes, etc.
4.	elevation	3	1. water 2. dry sand 3. grass dunes	1. fish, seaweed, shark, dolphin, bacteria, microbes, etc. 2. crab, insect, microbes, etc. 3. snake, tortoise, bird, microbes, etc.
5.	saltiness (salinity)	3	1. water 2. dry sand 3. grass dunes	1. fish, seaweed, shark, dolphin, bacteria, microbes, etc. 2. crab, insect, microbes, etc. 3. snake, tortoise, bird, microbes, etc.
6.	pH	3	1. water 2. dry sand 3. grass dunes	1. fish, seaweed, shark, dolphin, bacteria, microbes, etc. 2. crab, insect, microbes, etc. 3. snake, tortoise, bird, microbes, etc.

→ **TEACHERS PLEASE NOTE:** When you go over the answers with your students it would be good to mention that microbes could live in each of the zones that students find. Emphasize that microbes live everywhere.

Write in the table below a few of the types of gradients represented in this landscape. (Note that some might be visible and some might be invisible.) The first row shows one example answer, but the gradients, zones and life forms you identify may differ from the example.

Analyze this photo looking for gradients.



ANSWER KEY

The following are some possible answers. The italicized words in the third row of the table are given as an example in student workbooks.

	Kind of gradient (i.e. temperature, moisture, etc.)	How many zones of the gradient do you see?	List the different zones.	Name a plant or animal that could live in each zone.
1.	moisture	6	1. river (riparian), 2. valley floor (flood plain), 3. lower part of mountain with deciduous trees (foothills) 4. mid-mountain with evergreens (forest & sub alpine), 5. top of mountain above tree level (alpine), 6. air (atmosphere)	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. deciduous trees, squirrel, deer, bear, microbes, etc., 4. elk, evergreens, chipmunk, microbes, etc. 5. grizzly bear, pica, lichens, wolverine, etc. 6. bird, insect, microbes, etc.
2.	temperature	6	1. river, 2. valley floor, 3. lower part of mountain with deciduous trees, 4. mid-mountain with evergreens, 5. top of mountain above tree level, 6. air	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. deciduous trees, squirrel, deer, bear, microbes, etc., 4. elk, evergreens, chipmunk, microbes, etc. 5. grizzly bear, pica, lichens, wolverine, etc. 6. bird, insect, microbes, etc.
3.	human land use	2	<i>1. valley floor, 2. mountains</i>	<i>1. sage brush, yarrow, grass, insects, antelope, microbes, etc., 2. deer, bear, elk, evergreens, microbes, etc.</i>
4.	elevation	5	1. river (riparian), 2. valley floor (flood plain), 3. lower part of mountain with deciduous trees (foothills) 4. mid-mountain with evergreens (forest & sub alpine), 5. top of mountain above tree level (alpine)	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. deciduous trees, squirrel, deer, bear, microbes, etc., 4. elk, evergreens, chipmunk, microbes, etc. 5. grizzly bear, pica, lichens, wolverine, etc.
5.	pressure	3	1. water, 2. valley floor, 3. top of the mountain	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. grizzly bear, pica, lichens, wolverine, etc.
6.	saltiness (salinity)*	3	1. water, 2. valley floor, 3. mountain	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. deer, bear, elk, evergreens, microbes, etc.
7.	pH**	3	1. water, 2. valley floor, 3. mountain	1. fish, otter, duck, microbes, etc., 2. sage brush, yarrow, grass, insects, antelope, microbes, etc., 3. deer, bear, elk, evergreens, microbes, etc.

*(salt washes down with rain and snowfall off the mountains eventually ending up in the river)

** (small gradient) (pockets of minerals under the ground can change soil pH, as can fertilizers, the presence of certain plants, decaying plants or animals, etc.)

Background info for teachers to help students analyze the photos in Activity 1

A beach is an example of a location with many environmental gradients. In particular, we have the “wetness” of the land: Beyond the tide line is ocean environment, then we have the wet sand where the tides wash in, then the drier beach sand, and finally the dunes.

We can see distinct areas here, each with its characteristic forms of life. These identifiable areas along gradients are called **zones**, and the process of forming them is called zonation. Each zone has distinct lifeforms. For example, we would rarely find hermit crabs or clams in the dunes area, while beach grass can’t take hold in the intertidal zone.

Mountain slopes are another example of zonation; distinct types of vegetation and wildlife will be found at different *altitude zones*.

In the open ocean, the depth determines how much sunlight can penetrate, and we have zonation based on sunlight—the *euphotic zone* has enough sunlight to grow plants, the *dysphotic zone* is a “twilight” zone, and the *aphotic zone* is in complete darkness. Organisms thrive in all these zones but will have quite different adaptations. Creatures below the euphotic zone, for instance, often have bioluminescent features; they can “glow in the dark.”

- In the beach example, if you identify zones of a water gradient, that is an abiotic gradient but the grass on the dunes are a biotic marker that the resources available have changed. In other words, biotic features can help identify abiotic zones.
- In discussing the beach example, you may want to point out that air does not have a pH, however water moving through air (such as rain) does have a pH. In the photo the ocean would have a pH level, the drier sand out of the normal tidal range might have a slightly different pH due to minerals or decaying organic material, and the sand in the grass dunes might have a slightly different pH due to the plants growing there and mixing with non sandy soils at the boundary of the beach.
- Note: While explaining the concepts of biotic and abiotic, ask your students whether dead things should be classified as biotic or abiotic. The answer is that it is sort of in a “gray area.” We’d certainly call a living cow or tree biotic; but the minute it dies, is it abiotic? Since “recently living” things are important food sources, just like living things, scientists normally classify them as biotic. If it comes from the living world, it’s biotic. Now, once the bacteria and fungi have completely broken down that dead tree or cow into soil, and it’s no longer an important food source, we can consider it part of the abiotic world.

ACTIVITY 2 ANSWER KEY

Who’s More Extreme? You, an Insect, a Plant or a Fish?

What are the toughest organisms on our planet? What kind of life form could survive the world’s coldest or hottest temperatures? Could humans survive hotter or colder temperatures than most other life forms?

Think about all these questions as you make predictions below. Base your predictions on the temperature limits you think each type of organism could withstand, not what its optimal living conditions might be.

Vascular plants have specialized tissues for moving water and minerals throughout the plant, which includes almost all plants except for a few types such as mosses and algae.

Organism	Heat Survivors Which two types of organisms do you think can survive hotter temperatures than the rest? (Mark your first choice in the #1 column and your second choice in the #2 column.)	Cold Survivors Which two types of organisms do you think can survive colder temperatures than the rest? (Mark your first choice in the #1 column and your second choice in the #2 column.)
Mammals		#1
Insects		
Vascular Plants		
Fish		
Fungi		
Algae		
Bacteria	#2	#2
Archaea	#1	

ANSWER KEY

→ **TEACHERS PLEASE NOTE:** Instruct your students whether to use Fahrenheit or Celsius to graph and work with the data.

Now look at the temperature limits of organisms.

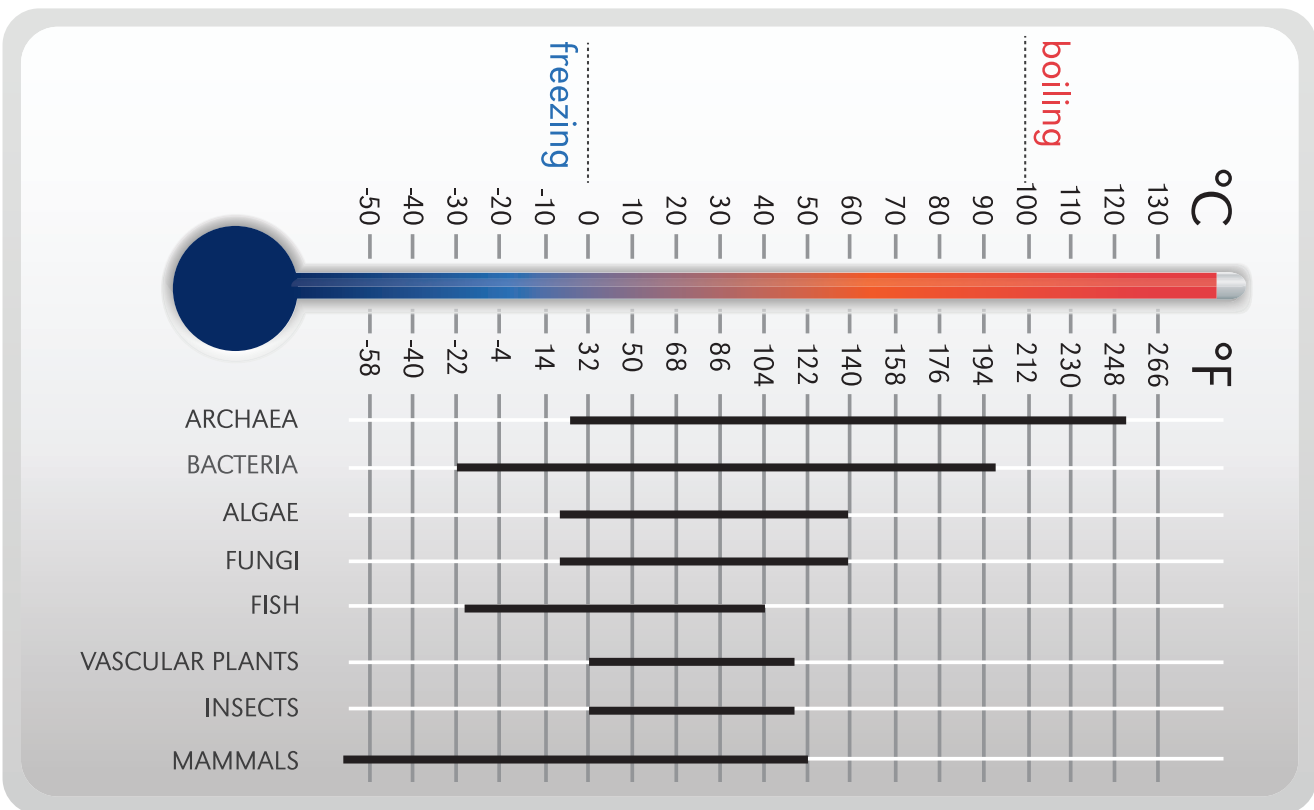
Calculate the middle temperature of the range and write it in the table below.

(Ask your teacher whether to work with the data using Fahrenheit or Celsius.)

Organism	Temperature Range	Middle Temp. of Range
Mammals	-58°F to 122°F (-50°C to 50°C)	32°F (0°C)
Insects	32°F to 118°F (0°C to 48°C)	75°F (24°C)
Vascular Plants	32°F to 118°F (0°C to 48°C)	75°F (24°C)
Fish	-20°F to 104°F (-29°C to 40°C)	62°F (16.5°C)
Fungi	-20°F to 140°F (-29°C to 60°C)	80°F (26.5°C)
Algae	-20°F to 140°F (-29°C to 60°C)	80°F (26.5°C)
Bacteria	-22°F to 203°F (-30°C to 95°C)	111.5°F (44°C)
Archaea	23°F to 250°F (-5°C to 121°C)	136.5°F (58°C)

When you have finished your graph, go back to the predictions you made and mark whether or not they were correct.

ANSWER KEY



ACTIVITY 3

ANSWER KEY

Yellowstone's Extreme Life

Use the information in this table to graph the niches of all of the microbes listed. (Your teacher will tell you whether to use Fahrenheit or Celsius values.)

Microbes that use photosynthesis	pH	Temperature range
Cyanidioschyzon	0–4	104–131°F (40–55°C)
Synechococcus	7–9	126–165°F (52–74°C)
Thermochromatium	6–9	93–135°F (34–57°C)
Zygogonium	0–4	90–131°F (32–55°C)
Microbes that don't use photosynthesis		
Hydrogenobaculum	3–5.5	131–162°F (55–72°C)
Metallosphaera	2–4	122–176°F (50–80°C)
Sulfurihydrogenibium	6–8	140–167°F (60–75°C)
Thermocrinis	7–9	131–195°F (55–91°C)

Directions:

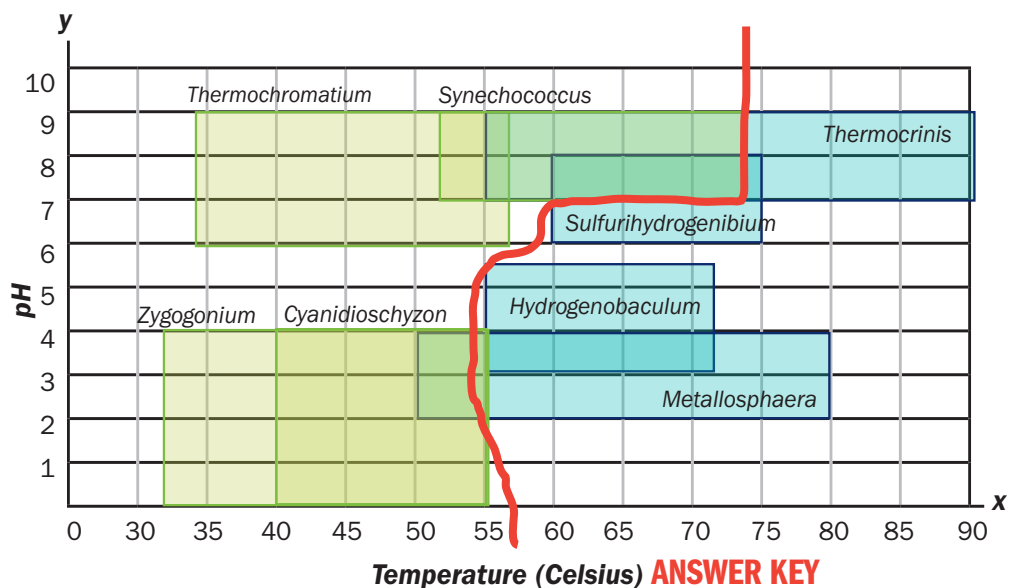
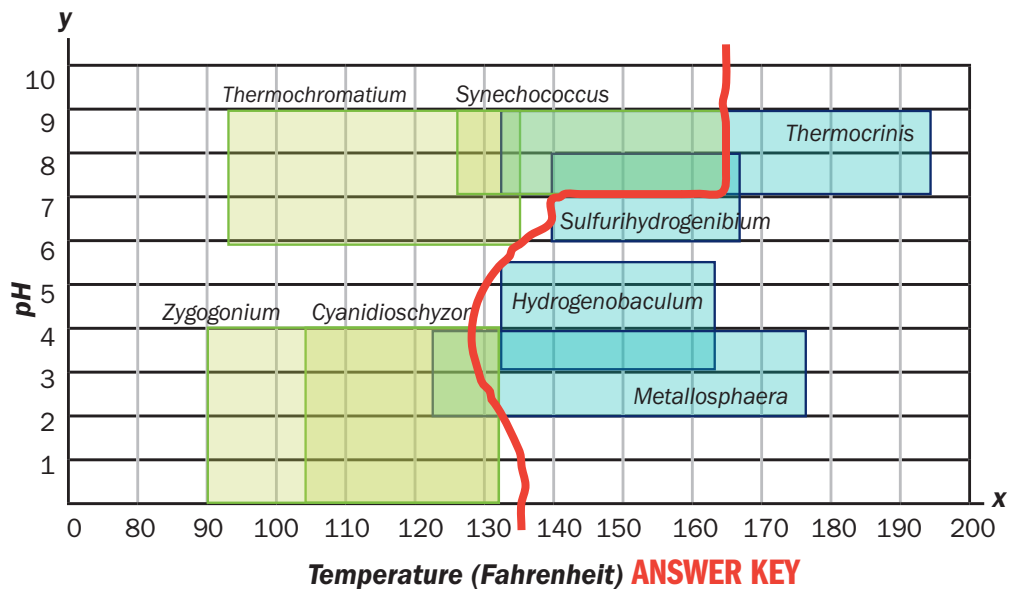
Use two colors: One to graph microbes that use photosynthesis and one to graph those that don't.

Graph each microbe's **pH range on the Y axis** and its **temperature range on the X axis**. (Since each organism can live in a range of pH and temperature, you will end up with a box that represents its niche. One is done for you as an example.)

Label each niche with the organism's name, abbreviating if necessary.

After you have plotted temperature and pH ranges for all of the organisms, draw a dividing line between those that use photosynthesis and those that don't.

Then, answer questions 1-4 on the next page.



Questions about extreme life **ANSWER KEY**

1. Now that you have finished your graphing, what pattern do you notice? (The organisms that use photosynthesis for energy are clustered on the left hand side of the graph indicating lower temperatures and the non-photosynthesizers are clustered on the right hand side indicating hotter temperatures.)
2. Based on the data you have graphed, what do you think is the upper temperature limit for organisms to live who use photosynthesis? (Around 166°F or 74°C)
3. Do you think this would be enough data for a scientist to make a prediction about the upper temperature limit of photosynthesis? (No, because this is an extremely small sample size of organisms from Yellowstone. There are thermal areas around the world that host a large diversity of microorganisms. Note that the actual upper temperature limit for photosynthesis is around 166°F or 74°C. Scientists need to be cautious about making definitive statements based on small experiments or samples)

Extreme life in Whirligig Geyser

Now look at this photo of Whirligig Geyser in Norris Geyser Basin. It is pH 3.4 and 154°F (68°C) at its source. It is full of iron and arsenic. Arsenic is very poisonous to most multicellular life. (Arsenic contamination of groundwater is a problem that affects millions of people across the world.)

Label the photo

Using the data you graphed on p.15 and the information below, find and label where each of the three following microbes are living in the photo.

- **Zygonium** is a purple or black colored type of algae and it performs photosynthesis.
- **Metallosphaera** is orange. It is resistant to many toxic metals and even uses some for energy. It oxidizes iron (turning it to rust).
- **Cyanidioschyzon** is a type of green colored algae and it performs photosynthesis.



Whirligig Geyser, Norris Hot Springs

4. For each microbe you labeled in the Whirligig Geyser photo, explain what environmental factors could be influencing where it lives. Focus on temperature and metabolism.

Zygonium (*Zygonium* is living in a side channel of water that has cooled to below 131°F (55°C). The water temperature is cool enough for photosynthesis and there is plenty of access to sunlight.)

Metallosphaera (*Metallosphaera* is living in the hot center channel where water has not cooled much. The temperature in the channel is most likely above 131°F (55°C) since no large mats of *Cyanidioschyzon* are visible. The water must be very iron rich to provide *Metallosphaera* with food.)

Cyanidioschyzon (*Cyanidioschyzon* is living along the edges of the spring in areas where the water has cooled to below 131°F (55°C). There is lots of access to sunlight and perhaps most of the iron and toxic minerals in the water have been removed by the *Metallosphaera* by the time it reaches the edges, so there less food available for *Metallosphaera* and more pure water for *Cyanidioschyzon*.)

ACTIVITY 4

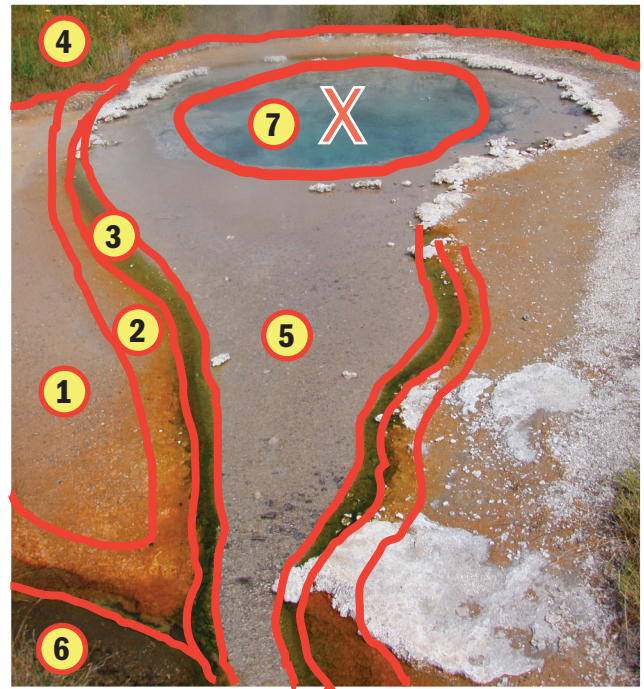
Exploring Yellowstone's Gradients

Yellowstone is one of the few places in the world where an amazing diversity of microbes can be seen without a microscope.

Large microbial **mats** and **filaments** with distinct white, yellow, green, pink, orange, brown and black colors are visible to the naked eye. Remember that Yellowstone is home to half (more than 10,000) of the world's hydrothermal features and they have a wide range of pHs, temperatures, and chemical compositions that yield a diversity of microbes that scientists are just beginning to discover.

Microbial colors can act as a living thermometer. Millions of tiny microbes band together into groups and form filaments or mats that are often distinct colors. The colors indicate where the water changes temperature, providing a giant map of temperature gradients, or occasionally chemical or pH gradients.

→ **TEACHERS PLEASE NOTE:** When discussing these images with your students, instead of “the effects of resource availability” determining where certain organisms live, it could be more of a case of tolerance. For example, sulfide inhibits photosynthesis and limits distribution of photosynthetic organisms probably because it binds to the same place water binds independent of the temperature.



“Gabby’s Spring,” Heart Lake region, was given this unofficial nickname by MSU researchers because Gabby Michel, a high school student from Big Sky helped them research the spring.

Find the gradients in “Gabby’s Spring”

1. Draw lines between the zones of the thermal gradients on the photo above.

How many zones do you think there are? (Approximately 7: 1-outer dirt area, 2-orange channel, 3-green channel, 4-grass, 5-center channel, 6-black area in lower right hand corner, 7-center blue area. The crusty white area could also be considered a zone.)

2. Draw an X where you think the hottest part of the spring is.

3. Do you think this thermal features contains life? (yes)

Why or why not? (There are microbes living in extreme environments around the Earth so there are most likely microbes living in this spring. The green and the orangish channels are probably microbial mats. The blue zone contains life but the microbes there don't form mats.)

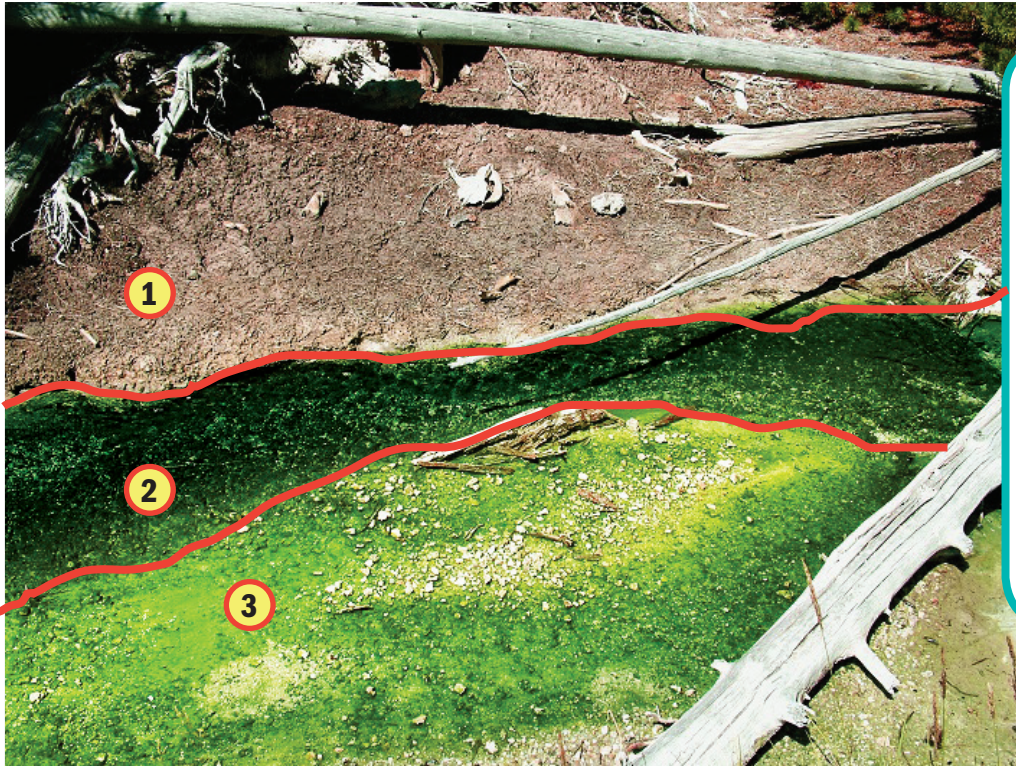
(If you think it contains life, answer the following questions. If not, skip to the photo on the next page.)

4. Describe what kind of microbes you think live in the spring and where they live. (Microbes, specifically bacteria and Archaea live in the spring.) Are there multiple zones of life? (Yes, there are multiple zones of life: 1), the blue part of the spring, 2), the green channel, 3), the orange channel, 4.)The white is a silica precipitate (abiotic) caused by biotic (living) and / or abiotic (non living) processes.)

5. How do you think the microbes get food? Do they all get food the same way? (The colors may be an indicator of the microorganisms having different metabolisms. The green may be a sign of chlorophyll and organisms that conduct photosynthesis. The organisms in the orange channel may use photosynthesis or the color could be a sign of organisms that are using iron and oxidizing it thus creating rust. The center of the spring is probably too hot for photosynthesis to occur so organisms there must use chemicals or minerals for energy.)

Hint: Finding Signs of Photosynthesis

Photosynthesis is a process used by plants and other organisms to convert light energy, normally from the Sun, into chemical energy. All photosynthesizing organisms have a substance called chlorophyll that is necessary to conduct photosynthesis. It is a green color, but it can sometimes be masked by other pigments that range in color from yellow to orange, or red to purple. If you see something green, that's usually a good indicator of photosynthesizing life.

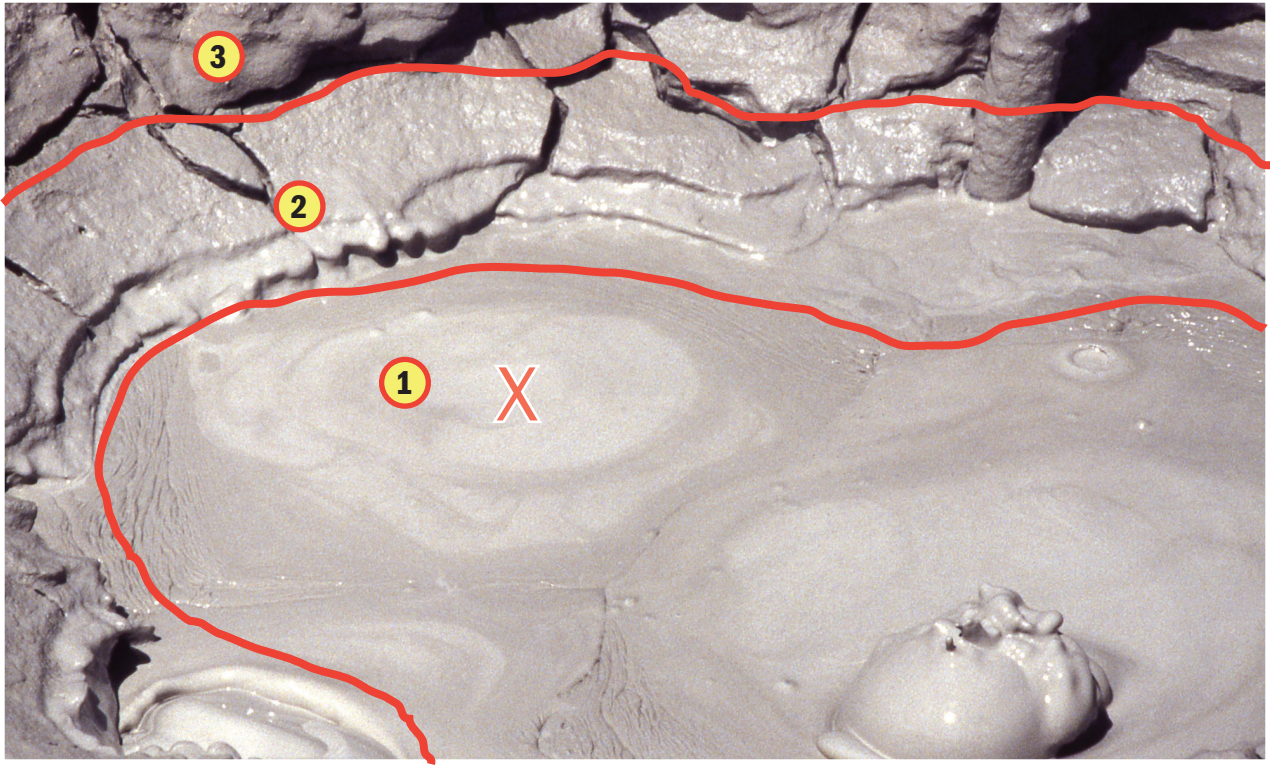


Note: Because of the high altitude of Yellowstone, water only needs to be approximately 199°F (93°C) to boil whereas at sea level water must be 212°F (100°C) to boil. Sometimes a bubbling hot spring is just an indicator of gases such as carbon dioxide escaping and not a sign of boiling from heat.

Lemonade Creek is fed by hot springs and filled with algae that eat arsenic, which is toxic to humans.

Find the gradients in Lemonade Creek

1. Draw lines between the zones of the thermal gradients on the photo above.
How many zones do you think there are? (approximately 3: 1-dirt, 2-darker green area, 3-lighter green area)
2. Do you think this thermal features contains life? (yes)
Why or why not? (The thick green growth is a sign of life.)
(If you think it contains life, answer the following questions. If not, skip to the photo on the next page.)
3. Describe what kind of microbes you think live in the spring and where they live. (Bacteria or algae live in the spring.) Are there multiple zones of life? There are at least two zones: the lighter green zone and the darker green zone.)
4. How do you think the microbes get food? Do they all get food the same way? (The green is probably a sign of chlorophyll. They probably all get food the same way due to both parts being green.)



Mud pots are created by very wet, acidic conditions that dissolve rock.

Find the gradients in this mud pot

1. Draw lines between the zones of the thermal gradients on the photo above.
How many zones do you think there are? (approximately 3: 1-the liquid part, 2-the thicker more solid mud, 3-the hard dirt at the farthest edge of the photo which has little or no water in it)
2. Draw an X where you think the hottest part of the spring is.
3. Do you think this thermal features contains life? (yes)
Why or why not? (There are no visible signs of life but microbes live in all kinds of extreme environments and probably live in this spring.)
(If you think it contains life, answer the following questions. If not, skip to the photo on the next page.)
4. Describe what kind of microbes you think live in the spring and where they live. Are there multiple zones of life? (The microbes who live in this spring do not form communities of mats. There are probably differing types of microbes in each zone of the water and/or heat gradient of this spring.)
5. How do you think the microbes get food? Do they all get food the same way? (Photosynthesizing microbes do not live in this spring since there are no green, yellow, red or purplish pigments present. The microbes who live in this spring do not necessarily all get their food the same way. They might get energy from either minerals or chemicals.)

ACTIVITY 5

RESEARCHERS SAMPLING HOT SPRING BACTERIA POPULATION IN YELLOWSTONE

Keep in mind that people and animals can die from falling in hot springs! Researchers need special permits to collect samples in Yellowstone. If you visit Yellowstone, never leave trails or boardwalks and do not touch the hot springs or microbial mats.



Sweet Population Sampling

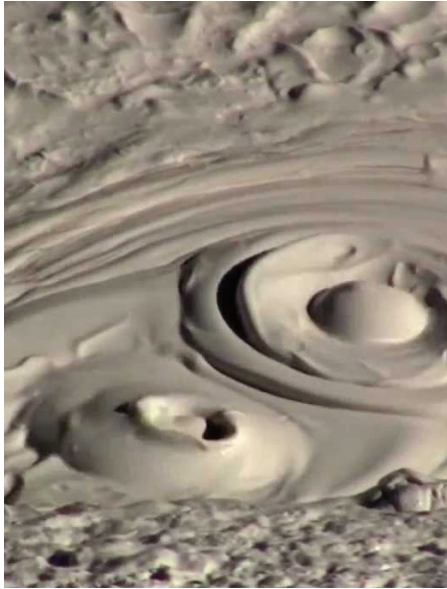
There are three simulations of the hot springs you studied in Activity 4 set up around your classroom. You are going to move around the room and count samples of microbes (that are represented by different colors of candy) from three Yellowstone thermal features.

Note that when scientists collect real data from Yellowstone hot springs there are many more types of microbes living in the hot springs than specimens they see in their sample. They may see numerous shapes and sizes of microbes in a microscope but may only be able to identify a few because of the difficulties of growing and cultivating extremophiles in a lab.

Copy in the box below what candy your teacher is using to represent each microbe.

1. Fill each square of the grid with drawings of the symbols (noted below the grid) that represent each organism (represented by a colored candy) you see.
2. Draw the symbols in the exact order and place you see them in the virtual spring, then total the numbers of each kind of microorganism you find in each box.
3. Once you have found a total number of specimens for each box, calculate the total number of specimens in the whole sample area (in other words in the whole grid).
4. Then, read the descriptions of the microorganisms found in that hot spring and answer the questions about them.

Organism	Candy Color
<i>Sulfolobus</i>	
<i>Caldisphaera</i>	
<i>Cyanidioschyzon</i>	
<i>Chlorella</i>	
<i>Thermus</i>	
<i>Chloroflexus</i>	
<i>Pseudoanabaena</i>	



Mud Pot **ANSWER KEY**

***Sulfolobus*, 65-87°C (149-188°F), pH 2-4**

This organism was first discovered in Yellowstone in 1972. Since then, species of *Sulfolobus* have been found in hot springs on Mount St. Helens, and in Italy, Russia, Chile, Japan and Papua New Guinea. It was one of the first hyperthermophilic organisms—to be discovered. It gets its energy from metabolizing sulfur or sulfur compounds.

***Caldisphaera*, 65-75°C (149-167°F), pH 2.5-5.5**

This microorganism converts sulfur into hydrogen sulfide – a gas that smells like rotten eggs and is flammable, very poisonous and corrosive. They have been found in hot springs in Yellowstone, the Philippines, Russia, and California. Hydrogen sulfide was used by the British Army as a chemical weapon during WW I. If someone is poisoned by inhaling too much hydrogen sulfide, a clue can be the discoloration of copper coins in the pockets of the victim. A bit of hydrogen sulfide is also present in the gas humans produce and is part of the cause of its smell.

Total <i>Sulfolobus</i> 7 Total <i>Caldisphaera</i> 4 Total # of microbes 11 X O O X X O O X X XX	Total <i>Sulfolobus</i> 24 Total <i>Caldisphaera</i> 1 Total # of microbes 25 O X	Total <i>Sulfolobus</i> 8 Total <i>Caldisphaera</i> 2 Total # of microbes 10 X X X X X O O X X X
O O X X X X X O O X X Total <i>Sulfolobus</i> 7 Total <i>Caldisphaera</i> 4 Total # of microbes 11	X O O Total <i>Sulfolobus</i> 22 Total <i>Caldisphaera</i> 2 Total # of microbes 24	X O O X X X X X O O Total <i>Sulfolobus</i> 6 Total <i>Caldisphaera</i> 4 Total # of microbes 10

X = *Sulfolobus*, O = *Caldisphaera*

Total of *Sulfolobus* in the grid 74

Total of *Caldisphaera* in the grid 17

1. Which of these organisms helps create smells like human gas? (*Caldisphaera*)
2. If someone dies of exposure to hydrogen sulfide gas, what might happen to any pennies in his or her pocket? (They might become discolored.)
3. Which of these organisms has been found in sites across the world? (both)
4. Does either of these organisms use photosynthesis for energy? (no)



Lemonade Creek

Cyanidioschyzon, 40-55°C (104-131°F), pH 0-4

Cyanidioschyzon is a spherical type of algae found in acidic hot springs. It uses sunlight for energy and performs oxygen photosynthesis. It is one of the most heat- and acid-tolerant algae known.

Chlorella, 20-35°C (68-95°F), pH 0.5-4

Chlorella, like *Cyanidioschyzon*, are a form of algae and they use photosynthesis for energy. They are a group of eukaryotes, meaning the interior of their cell contains both a nucleus as well as other compartments within the cell, called organelles. This makes them distinct from prokaryotic microorganisms, which lack both of these features. Interestingly, *Chlorella* are packed full of protein, fat, fiber, vitamins, and minerals, making them a very potent superfood that many people use as a daily vitamin.

<p>Total <i>Cyanidioschyzon</i> 6 Total <i>Chlorella</i> 0 Total # of microbes 6</p> <p style="text-align: center;">X X XX XX</p>	<p>Total <i>Cyanidioschyzon</i> 15 Total <i>Chlorella</i> 1 Total # of microbes 16</p> <p style="text-align: center;">XXXXXXXXXXOX XXXXX</p>	<p>Total <i>Cyanidioschyzon</i> 17 Total <i>Chlorella</i> 1 Total # of microbes 18</p> <p style="text-align: center;">XXX XXXXXXXXX XXXOXXX</p>
<p style="text-align: center;">XXXXXO</p> <p>OOOOOOOOO O OOOOO OOOOO OOOO OOOOO</p> <p>Total <i>Cyanidioschyzon</i> 5 Total <i>Chlorella</i> 29 Total # of microbes 34</p>	<p>OOOOOOOOOO OOOOO OOOOOOO OOOOOOO</p> <p>Total <i>Cyanidioschyzon</i> 0 Total <i>Chlorella</i> 28 Total # of microbes 28</p>	<p>OOOOOXO OOOOOOO OOOOOO</p> <p>Total <i>Cyanidioschyzon</i> 1 Total <i>Chlorella</i> 19 Total # of microbes 20</p>

X = *Cyanidioschyzon*, O = *Chlorella*

Total of *Cyanidioschyzon* in the grid 44

Total of *Chlorella* in the grid 78

1. Which of these organisms do some people use as a vitamin? (*Chlorella*)
2. What do both of these organisms do to get energy? (photosynthesis)



“Gabby's Spring”

Clear center channel: *Thermus*, 40-79°C (104-174°F), pH 5-9

Thermus is a rod-shaped bacterium that sometimes forms bright red or orange streamers. It contains pigments called carotenoids that act as a sunscreen and protect it from high levels of sunlight. A species of *Thermus* found in Yellowstone was the original source material for an enzyme that allows scientists to make many copies of DNA. Reproducing DNA quickly has allowed for breakthroughs in solving crimes, diagnosing diseases and identifying genes.

Green channel: *Chloroflexus*, 35-85°C (95-185°F), pH 7-9

This bacteria is rod shaped and forms filaments. It uses light for energy but does not produce oxygen as a byproduct. Scientists are studying *Chloroflexus* because they think it may shed light on the evolution of photosynthesis.

Orange channel: *Pseudoanabaena*, 30-50°C (86-122°F), pH 7-9

This microorganism is a type of cyanobacteria, an important group of bacteria that are nearly 2.8 billion years old. Cyanobacteria conduct photosynthesis, meaning they use a combination of sunlight, water, and carbon dioxide to produce food and oxygen. In fact, these ancient organisms are so good at photosynthesis they're thought to have created the oxygen atmosphere that we humans depend on to survive. Sometimes cyanobacteria can grow so fast in water bodies that they form large blooms. These blooms can threaten lakes and streams by blocking sunlight from penetrating down the water column or by producing harmful toxins that can make animals and/or people sick.

Total <i>Thermus</i> 0 Total <i>Chloroflexus</i> 0 Total <i>Pseudoanabaena</i> 6 Total # of microbes 6 V VV VVV	Total <i>Thermus</i> 13 Total <i>Chloroflexus</i> 15 Total <i>Pseudoanabaena</i> 15 Total # of microbes 43 VVVV V V OOO V V VV O O XXX O O V V V O V O XXXX O O V O O O XXXXXX O	Total <i>Thermus</i> 0 Total <i>Chloroflexus</i> 1 Total <i>Pseudoanabaena</i> 3 Total # of microbes 4 V O V V
VVV VV O O VVV O O VV O O O VV O O O	O O X O XXXXXX O XXX O O O	O O V V V O V O V V O O O V V O O V O O O V V V
Total <i>Thermus</i> 0 Total <i>Chloroflexus</i> 9 Total <i>Pseudoanabaena</i> 13 Total # of microbes 22	Total <i>Thermus</i> 10 Total <i>Chloroflexus</i> 7 Total <i>Pseudoanabaena</i> 0 Total # of microbes 17	Total <i>Thermus</i> 0 Total <i>Chloroflexus</i> 12 Total <i>Pseudoanabaena</i> 12 Total # of microbes 24

X = *Thermus*, O = *Chloroflexus*, V = *Pseudoanabaena*

Total of *Thermus* in the grid 23, Total of *Chloroflexus* in the grid 44, Total of *Pseudoanabaena* in the grid 49

1. Which one of these organisms has a pigment that it uses like sunscreen? (*Thermus*)
2. Which type of organism is similar to the ones scientists think may have been responsible for producing the oxygen in Earth's atmosphere? (*Pseudoanabaena*)
3. Which type of organism are scientists studying to try to understand the origins of photosynthesis? (*Chloroflexus*)

Extension Activity: Population Pie Charts **Answer Key**

Using your Sweet Sampling data, calculate the fraction, decimal and percent of each microbe in Mud Pot, Lemonade Creek and “Gabby’s Spring.” Color and label the pie charts to show which microbes are most prevalent in each feature.

Feature	Microbe	Fraction numerator/ denominator	Decimal numerator ÷ denominator	Percent decimal X 100	Degrees decimal X 360	Color representation on pie chart
Mud pot	<i>Sulfolobus</i>	74/91	.81	81%	292	
Mud pot	<i>Caldisphaera</i>	17/91	.19	19%	68	
Lemonade Creek	<i>Cyanidioschyzon</i>	44/122	.36	36%	130	
Lemonade Creek	<i>Chlorella</i>	78/122	.64	64%	230	
“Gabby’s Spring”	<i>Thermus</i>	23/116	.2	19%	72	
“Gabby’s Spring”	<i>Chloroflexus</i>	44/116	.38	38%	137	
“Gabby’s Spring”	<i>Pseudoanabaena</i>	49/116	.42	42%	151	

