

PUDDLES



The last time you saw a puddle of water on the sidewalk, did you realize it was teeming with life? Bacteria, algae, fungi, and even tiny insects and crustaceans make their homes in puddles and other temporary bodies of water.

Goal

To discover the abundance and diversity of microbial communities in puddles.

Activity Time

60 minutes

Time to Get Ready

120 minutes over a 3-week period

What You Need

Have the following for each team of 3:

- 1 vial of pH paper or pH indicator
- 1 eye dropper
- 1 magnifying glass
- 1 microscope (optional)
- 3 glass slides and 3 cover slips (optional)
- 1 graduated cylinder or baby food jar marked in 1-milliliter increments
- brown, green, and yellow paint color charts
- 4 artificial puddles
- 1 measuring teaspoon
- distilled water
- organic carbon sources such as potato flakes; sugar; fallen leaves; dry, fresh-cut grass

Getting Ready

Three weeks before

- Gather empty tennis ball cans, 1- or 2-L soda bottles with the tops cut off, plastic peanut butter jars, or quart-sized canning jars for the lab.
- Create an artificial puddle for each group in 2-L containers as shown in Figure 1. Fill each with distilled water. Add a handful of soil and some partially decayed leaves, grass, or hay. Label each container with the date. Store in a warm, well-lit location.

Two weeks before

- Create a second artificial puddle for each group.

One week before

- Create a third artificial puddle for each group.
- Locate a puddle or create one outdoors several days before the first meeting. If possible, use puddles in different locations or with different physical characteristics.

The day of the activity

- Create a fourth artificial puddle for each group.
- If a pH meter or pH paper is not available, make a pH indicator solution from red cabbage. Liquefy 2 cups of chopped red cabbage leaves and 1 cup of water in a food processor or blender. Strain through cheesecloth or a coffee filter. To use, add 10 drops of cabbage juice to 1 tablespoon of a sample of water to be tested. Color changes correspond to the following pH values as shown in Figure 1 on the **Participant Page**.

| Observation | Probable community |
|---|--------------------|
| Gold or brown mat on bottom | Diatoms |
| Dark green mat with bluish coat | Cyanobacteria |
| Thin grass green threads | Green algae |
| Green spherical net of threads | Green algae |
| Uniformly green water | Green algae |
| Pink water (particularly in bird baths) | Flagellated algae |
| Slimy, colorless patches on bottom | Protozoans |

Table 1. Puddle organisms.

Useful Information

Puddles are often teeming with life. The types of organisms present depend on many things. Size, sun, temperature, and food all help determine what can live in a puddle. Thirty to 80 different kinds of protozoans, bacteria, algae, and fungi may be found in one puddle. The numbers and types of organisms change continually. Some can be identified with microscopes. Others, like bacteria, require special equipment and tests. A few can be identified by closely observing the puddle. See Table 1.

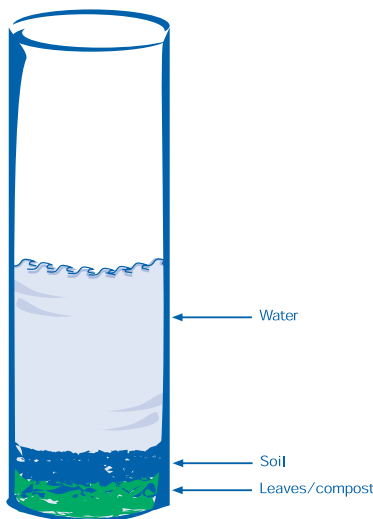


Figure 1. Puddle setup.



Suggestions to Modify the Activity for Those Who Are Exceptional

Specific modifications for this activity are found here. For common considerations when modifying activities for exceptional participants, see page V of the **Introduction**.

Blind or Visually Impaired

- Emphasize the discussions that are outlined in **Useful Information** and **Introducing the Activity**. They will be very beneficial to the participant for comprehension of the material and purpose.
- Emphasize that group discussions should include specific and detailed observations that use terms such as "fiery red" or "green grass." When discussing pH levels, refer often to the colors. Individuals who are blind have a good understanding of color and will appreciate the detailed observations.
- Construct tactile diagrams of organisms found in the puddles. Include detailed descriptions of each organism.

Deaf or Hard-of-Hearing

- See the **General Modifications** for *Blind or Visually Impaired* listed in the **Introduction**, page V.

Mobility Impaired

- Locate puddles outside in accessible areas.
- Show slides of the microscopic view of organisms if the participant cannot comfortably work at the microscope station. A magnifying glass is also an excellent adaptation.

Physically Impaired

- Wrap an elastic band around the jar to provide the participant better grasp. The larger the mouth is on the jar, the easier it will be to manipulate and transfer material into it.
- Show slides of microscopic views of the organisms if the microscope station is not comfortable for the participant. A magnifying glass is also an excellent adaptation.

Cognitively Impaired

- See the **General Modifications** for *Cognitively Impaired* listed in the **Introduction**, page V.

For More Information

Hampton, C.D. & C.H. (1994). Collecting and observing Protozoa. *Classroom Creature Culture*. Arlington, VA: NSTA Publications, 23-25.

Ingram, M. (1993). *Bottle Biology*. Department of Plant Pathology, College of AgriScience Institute: University of Wisconsin-Madison.

Kelly, S.G. & Post, F.J. (1989). *Basic Microbiology Techniques*. Belmont, CA: Star Publishing Company, 12.

Niederlehner, B.R. & Cairns, J. *Puddles* (Unpublished manuscript). MINTS. Virginia Tech Museum of Natural History: Virginia Tech, Blacksburg, VA.

Palmer, S. & Cairns, J. *Microbes in the World Around Us: A Laboratory Guide for the Study of Microbes in Environmental Samples* (Unpublished manuscript). MINTS. Virginia Tech Museum of Natural History, Virginia Tech, Blacksburg, VA.

How to Start the Activity

Take the group to an outdoor puddle. Have them list the features of the puddle. How did the water get there? Why is it still there? What lives in a puddle? What evidence of living things do they see? How many different kinds of living things do they think can be found in this puddle? What limits the variety or number of organisms found in a puddle? How will this puddle change over time? Is this puddle different from others? What factors affect living things or communities in puddles? What could they do to learn more about puddle organisms and how puddles change over time?

Let's Make a Hypothesis

Discuss the following questions to help guide the participants to make hypotheses.

- What might affect the growth of microorganisms in our artificial puddles?
- What do plant-like microorganisms need to live?
- What might bacteria feed on in natural environments?
- What are some sources of carbon that we can provide for bacteria?
- How do microorganisms get into puddles?

What the Data Mean

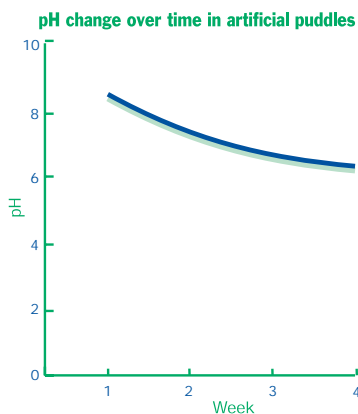


Figure 2. Graph of pH of artificial puddles over 3 weeks. pH decreased, possibly due to microbial activity. As microbes respire, they release carbon dioxide that reacts with water to form carbonic acid, lowering pH. Acidic waste products from microbial decay may also build up from decaying vegetation.

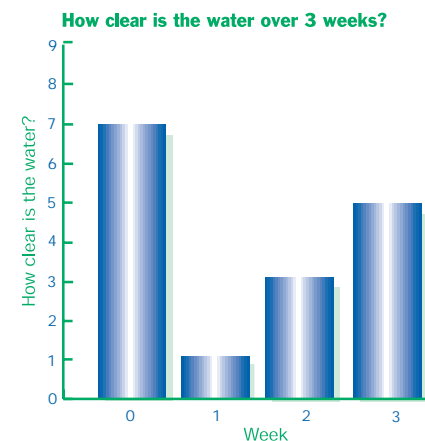


Figure 3. Graph of turbidity or clearness in artificial puddles over 3 weeks. Turbidity was high after adding soil. As material settled, turbidity decreased in Week 1. As microorganisms grew and reproduced, turbidity increased.

PUDDLES



Questions to Think About

Is there life in puddles? We have all splashed through puddles after rainy days. Some are temporary and last only a few hours while others last much longer. Does anything live in these puddles? If so, what? What environmental factors might affect what kind or how many organisms can live in a puddle? How do puddles change over time? How do organisms colonize, or end up in puddles? How can you design artificial puddles to answer these questions?

Safety Notes

- Food, drinks, and gum are not allowed.
- Wash hands at the completion of the activity.
- Avoid touching your mouth or eyes after handling water from puddles.

What to Do

1. Study your artificial puddles. Line them up according to the dates they were created. How are they similar to the puddle you observed outdoors? How are they different?
2. Use the paint color chart to record the colors of each artificial puddle. Record the color at the bottom as well as the middle of each water column. What do the colors suggest about the number or variety of organisms in the puddles? Can you determine what causes the colors on the bottom? Did the puddles' colors change over time?
3. Use pH paper or the red cabbage indicator to determine the pH of each puddle. See Figure 1 for red cabbage pH indicator colors. Graph your results. What might affect the pH in a puddle?

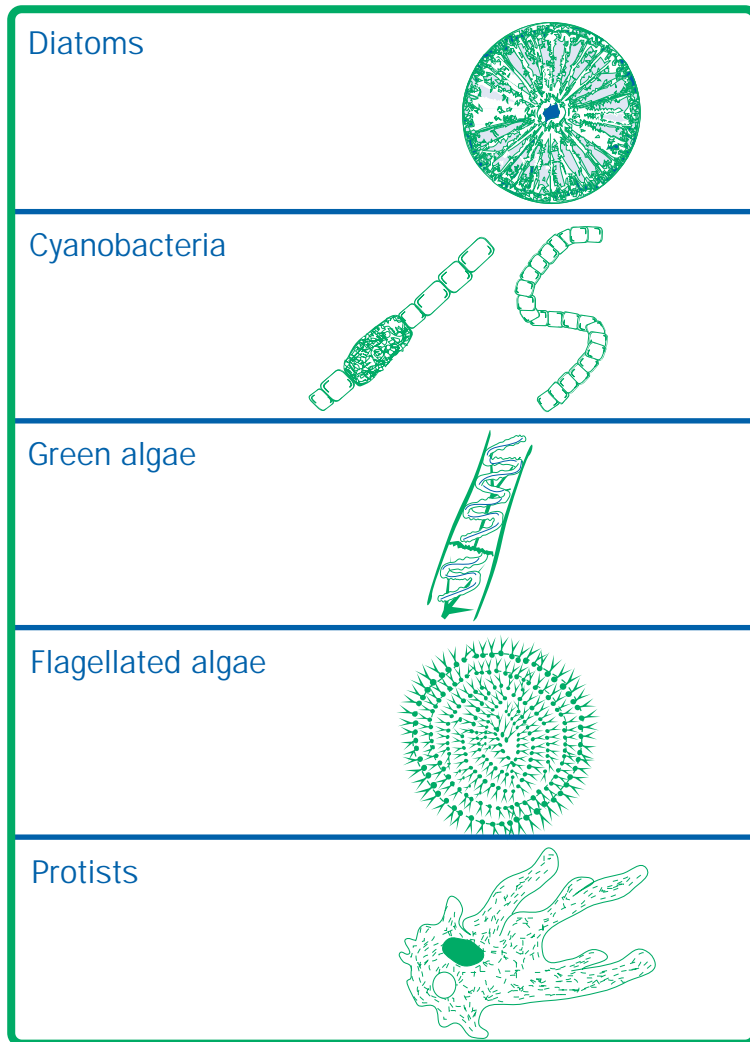
4. Collect a sample of water from the middle of each artificial puddle. If the sample is very turbid, or cloudy, do serial dilutions until the water appears clear. The number of times the solution needs to be diluted before it appears clear is your measure of turbidity. Use a graduated cylinder or baby food jar. Remove 1 part water from the puddle and add 1 part distilled water and shake to mix. If the solution is still cloudy pour off half and add another 1 part distilled water. Repeat until the solution is clear. If the sample requires 3 dilutions before it becomes clear, record a "3" as a measure of turbidity. Graph your results. What factors might cause the water to appear cloudy? Did the puddles' turbidity change over time? Graph your results.

5. Use an eye dropper to remove a sample of water from each puddle. If a microscope is available, place a drop of water on a slide and count the number of organisms and/or different kinds of organisms in the field of view at low power. Figure 2 shows some organisms that you may see. If no microscope is available, count the number of larger organisms you observe in a sample of known volume. Put this sample in the bottom of a baby food jar or a dish for closer examination. Be sure to examine and sample different parts of each puddle—the surface, middle, and bottom of the water column. Did the populations change over time? What factors caused these changes? If available, use keys and field guides to identify the organisms. If you do not have keys, sketch each kind of organism and make your own classification guide.



Figure 1. Red cabbage pH indicator colors.





What Did You Find Out By Doing the Activity?

Before doing "Puddles," did you know:

- what can be found in puddles besides water?
- how water gets into puddles?
- how puddles can support life?

From this activity, did you discover:

- what different types of living organisms exist in puddles?
- what factors affect an organisms survival in puddles?
- how the living organisms in puddles can be identified?
- if the living organisms in different puddles vary?
- how living organisms in the same puddle can change over time?
- how you could create a community of living organisms in a puddle?
- what products you use that could reach living organisms in puddles and alter their ability to survive?

Figure 2. Sketches of common organisms found in puddles.

6. What questions come from your results? To what other topics is this activity related? What did you learn from this activity? How does this activity relate to your life? What factors influence the populations of microorganisms?

7. How can you learn more about puddles? How can you learn more about what affects the populations of microorganisms in them? Could you study puddles outdoors? What factors could you manipulate to alter microbial populations? What factors might favor the growth of bacteria? of algae? of protozoans? Could you design an experiment to test a new hypothesis? What procedure would you use? What would you use for a control? What variables are important? How many trials would you include? What will you measure? Could you show your results in a graph?

