

# Biosphere in a Bottle

Below is an overview of the activity Biosphere in a Bottle (MicrobeWorld Experiments) to incorporate information learned from Dr. Kerkhof's presentation and subsequent discussion.

## Lesson Overview

Students investigate microbes that exist in a column of mud and the role of light on their survival.

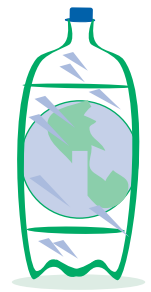
## Lesson Rationale

Microorganisms can be found everywhere. But like plants and animals, different kinds thrive in different places. Though difficult to see, pigmented photosynthetic bacteria are found in soil. Light provides the energy they need to grow. Photosynthetic microorganisms live in specific light intensities and different photosynthetic bacteria have different pigments that absorb different colors of light. Creating a Winogradski Column enables students to learn these principles of necessary resources and photosynthesis with different model organisms.

## Key Concept

Students will learn about the diversity of microbes that are present within mud and the resource requirements of different microbes for their survival by recreating Winogradski Columns in their classrooms.

# BIOSPHERE IN A BOTTLE



Have you ever dug a hole to plant a tree or bury something? Did you notice differences in the color of the soil? Why would the soil be different below the surface?

## Goal

To investigate microbes that exist in a column of mud and the role of light in their survival.

## Activity Time

3 to 8 weeks

## Time to Get Ready

Six weeks before the activity: 1 hour

One day before the activity: 1 hour

Day of the activity: 30 minutes

## What You Need

### Have the following for the group:

1 lamp with 40- or 60-watt light bulb (optional)

1 roll of masking tape

### Have the following for each team of 4:

4 wide-mouth, 2-quart jars or 2-L soda bottles

4 10 × 10-cm aluminum foil squares

1 permanent marking pen

1 trowel

1 small bucket

1 funnel

1 8-ounce measuring cup

1 small scoop or shovel

5 cups mud or sand from 4 different mud sources such as a pond, marsh, lake, garden, or forest

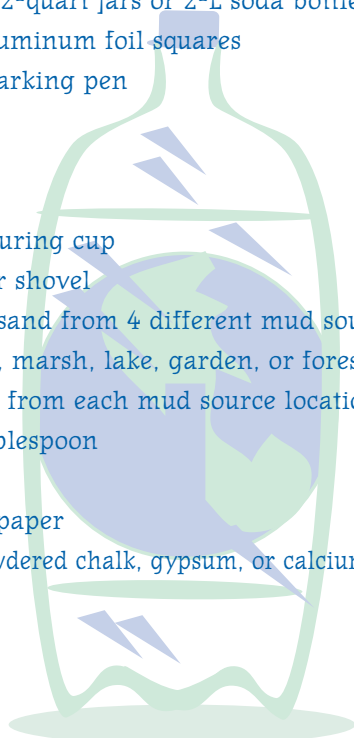
5 cups of water from each mud source location used

1 measuring tablespoon

1 paint stirrer

1 sheet of newspaper

1 tablespoon powdered chalk, gypsum, or calcium sulfate



## Getting Ready

### Six weeks before the activity

• Assemble a bottle or jar of mud as indicated in the **Participant Page**. Set it in a well-lit area until the group meets.

• Plastic soda bottles can be used instead of jars. Remove the top with scissors. See Figure 1. The bottles can be sealed using foil or plastic wrap and an elastic band. The remaining top can be used as a funnel.

### One day before the activity

- Gather soil from a variety of local areas such as a forest, garden or sediment from lake or pond.
- Use a pencil sharpener to powder the chalk. Gypsum or calcium sulfate may be substituted for powdered chalk.
- If time permits, shred the newspaper into strips no greater than 10-cm (4-inches) wide.
- Set out materials for each group.

### The day of the activity

• Assemble a second bottle or jar of mud as indicated in the **Participant Page**.

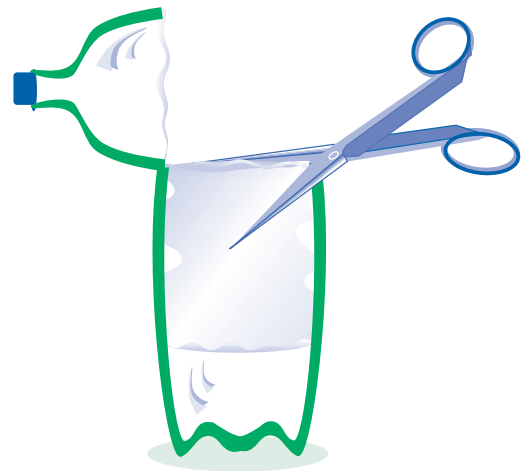


Figure 1. Diagram of a cut soda bottle.



## Useful Information

The biosphere is the Earth's life support system. Many organisms play important roles in the system. Microorganisms can be found everywhere. But like plants and animals, different kinds thrive in different places. Though difficult to see, pigmented photosynthetic bacteria are found in soil. Light provides the energy they need to grow. Two very important features of light that affect the growth of microbes are intensity and wavelength or color. Photosynthetic microorganisms live in specific light intensities. Too much light is as bad as no light. High light intensities retard photosynthesis and may cause organisms to overheat. White light is made up of many colors. A rainbow or prism reveals these colors. Different photosynthetic bacteria have different pigments that absorb different colors of light. As a result, they require different colored light to grow. For example, green algae do not grow well in green light. Oxygen requirements vary from microorganism to microorganism. An oxygen layer found at the top of mud is called an oxic zone. Further from the surface, the mud lacks oxygen, in an anoxic zone. Bacteria that require oxygen are called aerobic and live in the oxic zone, while bacteria that cannot tolerate oxygen are called anaerobic and live in the anoxic zone. Photosynthetic cyanobacteria produce rather than require oxygen. Other photosynthetic green and purple bacteria often produce sulfur or sulfur-containing compounds in place of oxygen.

In this activity, the biosphere column also is called a Winogradski Column. It is named after the Russian microbiologist Sergei Winogradski. Different substances added to the column change the microbe growth. For instance, ground chalk is a source of carbonate that starts photosynthesis. Egg shells have sulfate that makes hydrogen sulfide gas and anaerobic conditions. This is the gas that smells like rotten eggs.

## 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

- Allow participants to touch and describe the textures of the different soils (such as moist, dry, dense, and porous) gathered for a better understanding of the variable. This will make it easier for the participant to develop independent hypotheses and conclusions.

- Place all measuring devices to the right and the marked soils to the left for consistency in the setup.
- Replace group or class discussions with daily observations recorded in a journal. Discuss specific observations. Make repeated references to color, texture, size, and moisture. Individuals who are blind have a good understanding of color and will appreciate the detailed observations. Use the other senses for clarification with statements such as, "This is the gas that smells like rotten eggs."

### Deaf or Hard-of-Hearing

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

### Mobility Impaired

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

### Physically Impaired

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

### Cognitively Impaired

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

## For More Information

- Broad, W.J. (1998). Paradise lost: Biosphere retooled as atmospheric nightmare. *The New York Times*, CXLVI-II(51,346).
- Hampton, C.H., et al. (1994). Collecting and observing algae. *Classroom Creature Culture*. Virginia: NSTA Publications, 12-14.
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- Neimark, J. (1998). A conversation: Using flows and fluxes to demythologize the unity of life. *The New York Times*, CXLVII(51,246).
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## How to Start the Activity

Show the participants a fully-developed biosphere in a bottle made 6 weeks earlier and a freshly-made bottle. Tell them the bottles contain exactly the same materials. Ask them to compare the bottles.

## Let's Make a Hypothesis

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

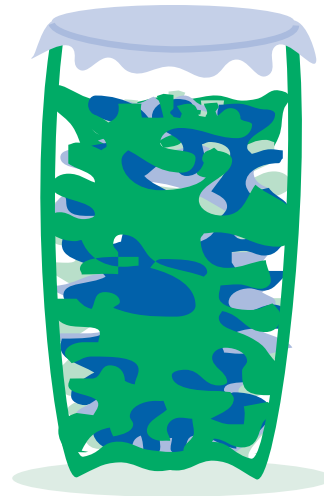
- Do the bottles look like they contain the same materials?
- If they contain the same materials, why do they look different?
- What do you think the different colors in the bottles represent?
- What is the difference between the 2 bottles?
- What is the same?

Ask the following questions at the end of the first session. How would you test the hypothesis that light is essential for the growth of photosynthetic microbes? How would you test the hypothesis that photosynthetic microorganisms can be found everywhere even if you can't see them?



## What the Data Mean

Explanations for results of the 6-week column growth in Figure 2 are listed below.



**Figure 2. Sample column showing areas of bacterial growth after 6 weeks.**

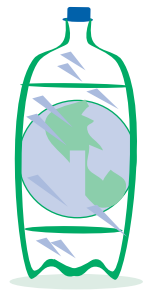
- The different colors in the mud represent the presence of different photosynthetic microbes.
- Red and orange patches are purple phototropic bacteria. Green patches in the oxic zone are cyanobacteria and algae. Olive-green patches in anoxic zones are green sulfur bacteria. The white color is sulfur produced from hydrogen sulfide by bacteria that do not produce oxygen when they photosynthesize and other sulfide-oxidizing bacteria. Hydrogen sulfide is produced by sulfate-reducing bacteria. The black patches are iron sulfide formed by sulfate-reducing bacteria.
- The patchiness is the result of the formation of microenvironments. The microenvironment is the environment right next to a microbial cell or group of cells.

### Explanations for results of possible participant-designed experiments.

- A bottle kept in the dark will not grow photosynthetic bacteria because light energy is critical to the development of photosynthetic organisms. However, other types of bacteria may grow. Even some phototrophs can produce energy for growth by other means in the dark.
- A bottle kept in direct sunlight may not show any growth because high light intensity retards photosynthesis, and the extreme heat can stifle growth.
- A bottle kept in extreme heat will not show any growth unless the soil comes from a hot spring, because most organisms cannot survive temperatures greater than 50°C (120°F).
- If the bottles are covered with colored cellophane, different microbes will grow, as they require different light colors. For instance, purple sulfur bacteria need red to near infrared light, green sulfur bacteria need green or red light, and cyanobacteria and green algae need red light.



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## Questions to Think About

Think about your neighborhood. How many different living things do you see? Where are they located? Why are some in one place, but not in others? What determines where living things can survive? Have you ever noticed patches of different colors in the soil when you dug a hole? What causes those colors? Why are they not distributed evenly throughout the hole?

## Safety Notes

- Wash hands before and after the activity.
- Use caution when working with soil and glass bottles.
- Latex or rubber gloves should be worn if a participant has an open cut or wound.
- Use chalk where possible. Gypsum or calcium sulfate can irritate the skin.

## What to Do

1. If it has not already been done, shred a full sheet of newspaper into very small pieces. Set it aside.
2. In a small bucket, add 5 or 6 cups of soil. Pick out all the sticks, leaves, and rocks. Stirring with the paint stirrer, slowly add water to the soil until it becomes the consistency of thick cream. The amount of water needed will depend on how moist the soil was at first. Add the shredded newspaper and 1 tablespoon of powdered chalk to the mud slurry. Mix the contents gently. Make sure the mixture is fluid so it will flow easily through the funnel.
3. Remove any existing labels from your bottle. Make a new label for your bottle with the name of the mud source on it. Set the funnel into the mouth of the bottle. Tape it securely in place. Scoop approximately 1 centimeter (cm) of the mud mixture into the bottle. With one hand covering the opening of the bottle, and the other holding the base of the bottle, gently tap the base on a table to settle the mixture evenly. Continue to fill the bottle, gently tapping every few centimeters until it is filled to within 4 or 5 cm of the top. Cover with foil. See Figure 1.

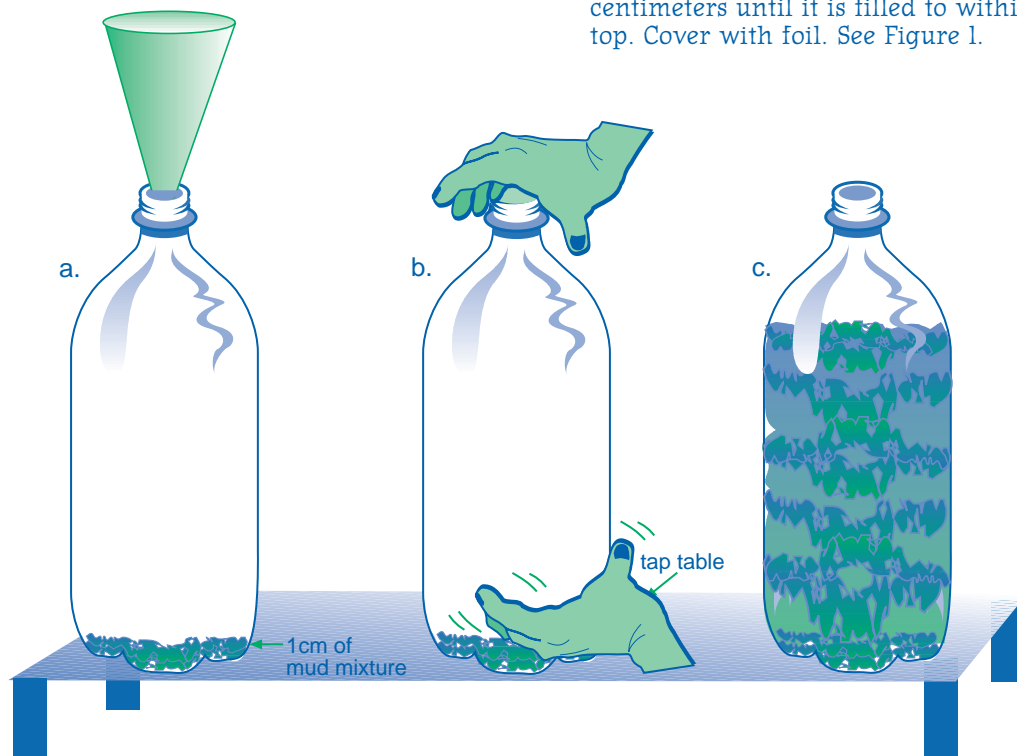


Figure 1. Biosphere setup. Figure 1a and b show how to add the mud to the column. Figure 1c shows the final setup.



4. Repeat the above process to fill each of your bottles. Use mud from different places for each bottle. Be sure each is properly labeled.

5. Take your biosphere bottle home and place it in a well-lit place away from direct sunlight. A window with a northern exposure works best. If you do not have a window, place the bottle about 60 cm (24 inches) from a 40- to 60-watt lamp. For best results, don't expose the bottle to direct sunlight or intense heat.

6. Keep the bottle in one position. Do not move it. Observe the bottle daily, looking for color or other changes in the mud. Be patient. It takes about 6 weeks to notice any color changes, but you should see other changes sooner, such as the formation of gas bubbles. What causes this? What gas could it be? Write your observations in a journal or notebook. Draw, label, and color a picture of the column at the end of each week. Why are there different colors in the bottle? What makes the red, orange, green, white, and black colors? Why do some colors appear in one part of the bottle and not another?

7. What other questions come from your results? To what other topics is this activity related? What did you learn from this activity? How is this activity related to your life? What did your results show?

8. How can you learn more about photosynthetic microorganisms? What steps would you use? What if you used different colored bottles or wrapped the clear bottles in colored cellophane? What if you added different nutritional sources like straw, grass, filter paper, baking soda, crushed vitamins, carbonated beverages, or yeast extract to the mud? What if you put the bottle in the dark? What if you put the bottle under intense heat?

9. Design a new experiment based on data you gathered or questions you asked during this activity. Develop a hypothesis that can be tested in a controlled experiment. Write a procedure in a numbered list. What is your control? What variables are important? How many trials have you included? What will you measure? How can you show your results?

## What Did You Find Out By Doing the Activity?

**Before doing "Biosphere in a Bottle," did you know:**

- that there are living organisms in water?
- that there are living organisms in mud?
- what the basic requirements for life are?

**From this activity, did you discover:**

- that living organisms survive in water? in mud?
- why the number of living organisms appearing in the bottles is different as time passes?
- why the mud layers appear to be different colors?
- how to find signs of living organisms in the bottles?
- how to find out why organisms grow in certain places in the bottle and not in other places?
- that life can exist in the slime found on a sidewalk?
- that different slimes can be good or bad for the environment?
- that organisms which are able to survive in a bottle require many of the same basic nutrients that they need to live?

