

## ***Winogradsky Columns***

### **For YSP Volunteers: How to Run this Activity**

#### **Logistics**

**Number of Volunteers:** 1 – 2 volunteers for every 10 students

**Amount of Set-up Time:** 20 – 30 min (depends on when the soil is collected)

**Amount of Running Time:** 2 hours

#### **Module Overview**

This module begins with an overview of biogeochemical processes that take place on Earth. Students learn how nutrients are cycled within the ecosystem. The best way to learn about these biogeochemical processes over time is by studying it in a closed system via a winogradsky column. Within this column, stratifications are produced over time and a multitude of bacteria grow in specific areas due to the availability of certain nutrients.

#### **Curriculum Links**

##### **Missouri Science Standards**

**4.2.B.a** - Explain the processes involved in the recycling of nitrogen, oxygen, and carbon through an ecosystem

**4.2.B.b** - Explain the importance of the recycling within an ecosystem

##### **Next Generation Science Standards**

**HS-ESS2-6.** Develop a quantitative model to describe the cycling of carbon among the hydrosphere, atmosphere, geosphere, and biosphere.

#### **Goals**

1. Define the biogeochemical cycle.
2. Understand the role of microorganisms in ecosystems.

#### **Materials**

##### **For each group:**

- 2-liter plastic soda bottle or small vase about 6.5” cylinder vase
  - Try purchasing this from Walmart, if not then try the dollar stores, they are expensive on Amazon
  - Try to go with smaller and longer rather than wider since that will require more mud if you want each student to have one
- 5 cups of mud/sand from a forest, garden, lake, pond, marsh or ocean
- 5 cups of water from each mud/sand location used
- 1 small bucket
- 1 measuring cup
- 1 paint stirrer or large spoon
- 1 sheet of newspaper
- 1 tablespoon powdered chalk
- Hard boiled egg yolk
- 1 measuring spoon
- Rubber band

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### **For the entire classroom:**

- Aluminum foil or plastic wrap
- Pencil sharpener
- Mortar and pestle
- Lamp with 40- to 60-watt light bulb
- Hand wipes or sanitizer
- Paper Towels or Clorox Wipes
- Lots of Newspaper
- Gloves

### **What to Expect**

**Based on past experience running this activity, you can expect...**

This to be a very messy demonstration. It could be less messy if you decide to gather the mud and water before hand or messier if you incorporate it into the demonstration by renting the Washington University Biology vans. Make sure you have lots of newspaper underneath the bucket the students will be using to scoop the mud out of. Also line the desks with newspapers or the plastic wrap before the demonstration so it's a lot easier to clean up the desks. It's also best if the students have done the pond ecosystem module before doing this module so that you don't have to review what an ecosystem is.

### **Other Resources/Citations:**

- <http://www.scientificamerican.com/article/bring-science-home-soil-column/>
- [http://quest.arc.nasa.gov/projects/astrobiology/fieldwork/lessons/Winogradsky\\_5\\_8.pdf](http://quest.arc.nasa.gov/projects/astrobiology/fieldwork/lessons/Winogradsky_5_8.pdf)
- <http://www.sumanasinc.com/webcontent/animations/content/winogradsky.html>

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### Introduction

**Ecosystems** are sets of living organisms (plants, animals, and microorganisms) that interact with each other and the environment in which they live (soil, climate, water and light). For all living components of an ecosystem to survive, all chemical elements that make up living cells must be recycled continuously. Biogeochemical cycles (which humans interact with the most) can be classified as **gaseous**, in which the reservoir is the air or the ocean, or **sedimentary**, in which the reservoir is the Earth's crust.

**Gaseous cycles** include those of nitrogen, oxygen, carbon and water. These tend to move more rapidly than do sedimentary ones to adjust more readily to changes in the biosphere because of the large atmospheric reservoir. Extraordinary disturbances (such as global warming) and more frequent local disturbances (such as wildfires and storm-driven elements) can seriously affect the capacity for self-adjustment.

**Sedimentary cycles** include those of iron, calcium, phosphorus, sulfur and other earthbound elements. These vary from one element to another, but each cycle consists fundamentally of a solution phase and a rock phase. In the solution phase, weathering (alteration, disintegration of rock) releases minerals from Earth's crust in the form of salts, some of which dissolve in water, pass through a series of organisms, and ultimately reach the deep seas, where they settle out of circulation indefinitely. In the rock phase, other salts deposit out as sediment and rock in shallow seas eventually to be weathered and recycled.

We will now go through some pretty important gaseous cycles.

**Carbon cycle:** Carbon moves from the atmosphere to plants. In the atmosphere, carbon is attached to oxygen in a gas called carbon dioxide ( $\text{CO}_2$ ). Through **photosynthesis**, carbon dioxide is pulled from the air to make food from the carbon component. Then carbon moves from producers (plants) to consumers (animals) through food chains, when consumers eat producers and other consumers. Finally, when plants and animals die their bodies **decay**, releasing carbon to the soil. Carbon can also move from living things to the atmosphere. For example, every time you breathe you release carbon dioxide into the atmosphere. This process is called **respiration**. Additionally, carbon can move from fossil fuels to the atmosphere when the fuels are burned. Carbon can move from the atmosphere to the ocean, which can serve as a carbon reservoir because it is able to soak up carbon from the atmosphere. Carbon dioxide is a greenhouse gas and traps heat in the atmosphere. Without it and other greenhouse gasses, Earth would be a frozen world.

**Nitrogen cycle:** Nitrogen can be fixed from the atmosphere to ammonium ( $\text{NH}_4^+$ ) by nitrogen-fixing organisms, such as bacteria in the genus *Rhizobium*. Ammonium is then used by plants, other bacteria, or other soil organisms. Ammonium is incorporated into proteins and other organic nitrogen compounds such as DNA. Nitrogen can also be fixed by high-energy natural events such as lightning, forest fires and hot lava. Nitrogen ( $\text{N}_2$ ) is broken into individual N atoms. Organic nitrogen can be converted back to inorganic nitrogen (ammonium) through **nitrogen mineralization** or **decomposition**. The ammonium can then be taken up by plants or converted into nitrate ( $\text{NO}_3^-$ ) through **nitrification**. Then denitrifying bacteria convert nitrate to dinitrogen ( $\text{N}_2$ ) through **denitrification**. This is the only nitrogen transformation that removes nitrogen from ecosystems.

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**Water cycle:** During warmer temperatures, water is **evaporated** from oceans and freshwater bodies into the atmosphere. During **condensation**, when the temperature has decreased the gas form of water is released from the atmosphere and is returned to the Earth in the form of millions of tiny droplets which form clouds in the sky or fog at ground level. When the temperature and atmospheric pressure is just right, the small droplets of water in clouds form larger droplets and fall to the ground as raindrops. This is called **precipitation**. When water has returned to the ground, it flows across the landscape and down hills into streams, rivers, ponds, and lakes. Small streams flow into larger streams, which flows into rivers and eventually into the oceans. This process is called **surface runoff**. Once the water is in the ocean it can then be evaporated and returned to the atmosphere. When rain water soaks into the ground it is called **infiltration**. The water can either remain underground or return to the surface at springs or in low spots downhill. If the water remains in the soil, it can be taken up by plants through their roots and can transferred to their leaves. Water can then evaporate from the leaves, a process called **transpiration**.

**Oxygen cycle:** Plants produce oxygen as a byproduct of **photosynthesis** when they use energy from sunlight to release carbon from carbon dioxide. The sunlight frees up oxygen from the atmosphere by the process of **photolysis**. Animals breathe in oxygen through **respiration**. When water runs over rocks oxygen can enter water and persists as dissolved oxygen. Also oxygen can be fixed in minerals and freed by chemical weathering.

Biogeochemical cycles are **closed systems**, which means nutrients are never lost or created; they are continuously reused and recycled usually by soil microbes. Sergei Winogradsky invented a system in order to study the biogeochemical recycling process: a long, sealed-column of muddy so, water and shredded paper, called a **Winogradsky column**. You can think of the Winograsky column as a small ecosystem. As oxygen diffuses downward from the surface of the water, fermentation products from the breakdown of cellulose and hydrogen sulfide move up to the top of the column. Oxygen conditions remain very poor at the bottom of the column. Bacteria that are present at the bottom of the column are mainly responsible for the decomposition of the cellulose found in the shredded paper. As the chemical products are released from bacteria, they diffuse upward and are responsible for groups of organisms to accumulate based upon the stratification of nutrients and their required conditions for survival.

### **Materials**

- 2-liter plastic soda bottle (or you can choose to use
- 5 cups of mud/sand from a forest, garden, lake, pond, marsh or ocean
- 5 cups of water from each mud/sand location used
- 1 small bucket
- 1 measuring cup
- 1 paint stirrer or large spoon
- 1 sheet of newspaper
- 1 tablespoon powdered chalk
- Hard boiled egg yolk
- 1 measuring spoon
- Rubber band

## Winogradsky Columns

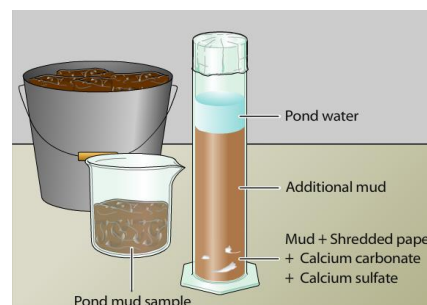
### Protocol

#### Prior to building the column:

1. Gather mud or sand from a forest, garden, lake, pond, marsh, or ocean
2. Gather water from each mud or sand location used.
3. Carefully cut off the top of the 2-liter bottle to use as a funnel
4. Use a pencil sharpener to powder the chalk.
5. Use a mortar and pestle to mash the hard-boiled egg yolk. (1 egg yolk is approximately 4 teaspoons of egg yolk powder)

#### Directions for building the column:

1. In a small bucket, add 5 cups of mud or sand. Remove any sticks, leaves or rocks
2. Stirring the mud or sand with a large spoon, slowly add water until the mixture is like thick cream. Be careful not to add too much water.
3. Shred a full sheet of newspaper into very small pieces. Add the newspaper shreds to the mixture.
4. Then add 1 tablespoon of powdered chalk to the mixture.
5. Add 1 teaspoon of mashed hard-boiled egg yolk to the mixture
6. Stir the mixture gently using a large spoon. Make sure the mixture is fluid so it will flow through the funnel.
7. Remove any labels from your bottle. Make a new label with the names of the students in your group and source of the mud or sand.
8. Set the funnel into the mouth of the bottle. Secure the funnel with tape or have a group member hold the funnel in place.
9. Pour or scoop a small amount of the mixture into the base of the bottle.
10. Place your hand over the top of the bottle and tap the bottom of the bottle firmly on the table. This helps the mixture settle and removes oxygen that is trapped in the mixture.
11. Repeat the two previous steps of adding a small amount of mud and settling the mixture until the bottle is about 90% full
12. Stir the mixture in the bottle to remove any air bubbles.
13. Let the bottle sit for 30 minutes. Add 2cm of pond water. Add/remove the water in your bottle as needed.
14. Cover the bottle with foil or plastic wrap and a rubber band.
15. Place the completed column in a well-lit place away from direct sunlight. Or you can place the bottle about 60 cm from a 40- to 60-watt lamp.



### Data and Analysis

Every week in your notebook:

1. Record the temperature in the room
2. Record visual observations of the amount of light in the room
3. Draw, label, color a picture of your column
4. Measure the thickness of the layers

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5. Use the table below to identify the bacteria that are growing in the column.

### Discussion Questions

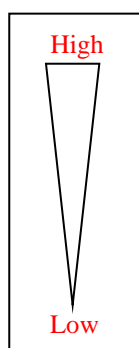
1. How are the elements carbon, hydrogen and oxygen cycled through the column?
2. Draw the oxygen gradient in the column and explain why there is a gradient.
3. Is the Winogradsky column a closed system?
4. How do you think the oxygen gradient in a Winogradsky column will affect where different microbes are living in it?

Column Position	Bacteria	Color
Top	Cyanobacteria	Green
	Purple non-sulfur bacteria	Red, purple, orange, or brown
	Purple sulfur bacteria	Red/purple
	Green sulfur bacteria	Green
Bottom	Sulfate reducing bacteria	Black

5. Draw the sulfur gradient in the Winogradsky column. What is the difference between purple and green sulfur bacteria? What are the similarities?

### Discussion Questions—ANSWER KEY

1. How are the elements carbon, hydrogen and oxygen cycled through the column?  
 Through photosynthesis and cell respiration.
2. Draw the oxygen gradient in the column and explain why there is a gradient.



The shredded newspaper and sulfate triggers microbial activity and the microbes deplete the oxygen in the mud. The loss of oxygen creates a gradient with low oxygen at the bottom and high oxygen at the top.

3. Is the Winogradsky column a closed system?

Yes, it is a mini ecosystem which contains microbes that are contributing to the biogeochemical cycles.

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4. How do you think the oxygen gradient in a Winogradsky column will affect where different microbes are living in it?

Microbes that can tolerate oxygen or need oxygen in order to live will be found at the top of the column while the microbes that can grow in the absence of oxygen will be found at the bottom of the column.

5. Draw the sulfur gradient in the Winogradsky column. What is the difference between purple and green sulfur bacteria? What are the similarities?

Green sulfur bacteria are able to tolerate higher concentrations of sulfur than purple sulfur bacteria. They both use carbon dioxide as a carbon source and sulfur as an electron donor.

