

# Carbon Cycle

## 1. Pre-lab Reading

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Chapter 44 Section 5 “Climate and the Effects of Global Climate Change” Biology2e,  
OpenStax College

## 2. Purpose and Objectives

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The purpose of this lab is to

- explore ecological roles of different organisms in the carbon cycle.
- review types of metabolism including aerobic respiration and photosynthesis.

Upon completion of this lab, you should be able to:

1. Describe metabolic processes in **autotrophs** and **heterotrophs**.
2. Trace the pathway of carbon in a land ecosystem or marine ecosystem starting and ending with carbon dioxide (CO<sub>2</sub>).
3. Tell which metabolic processes add and remove CO<sub>2</sub> from the atmosphere.
4. Explain why burning **fossil fuels** can increase atmospheric CO<sub>2</sub> and describe the effect of increased atmospheric CO<sub>2</sub> on global temperature.
5. Use a **pH indicator** to measure changes in CO<sub>2</sub> level in a solution.

## 3. Safety Guidelines

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1. Wear safety goggles

## 4. Background Information

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### The Carbon Cycle

An **ecosystem** includes the organisms of a particular habitat together with the physical environment in which they live (Hillis). The viability of organisms in an ecosystem depends on their ability to obtain energy and the necessary materials to sustain their structures and reproduce. All organisms require sources of carbon, nitrogen, phosphorus, sulfur and other elements. These elements (or molecules containing them) are taken up by some organisms in an ecosystem from inorganic sources, passed along to other organisms in the food chain, and, typically, recycled to an inorganic form when organisms die. This exercise will examine some aspects of the **carbon cycle** and ask which organisms take up carbon in the form of  $\text{CO}_2$  and which convert organic forms of carbon back to  $\text{CO}_2$ .

Carbon is essential to all forms of life. All organic molecules, including carbohydrates, lipids, proteins and nucleic acids, contain carbon. Carbon enters the biosphere from  $\text{CO}_2$ , which is found in the atmosphere and also dissolved in water. **Autotrophs** are able to “fix” carbon, that is remove it from the atmosphere (or from water) and bind it to organic molecules. These organisms then use this carbon to make sugars and other organic molecules. Most autotrophs are **phototrophs**, which use light energy to power this process. A few prokaryotes are **chemoautotrophs**; organisms that use inorganic chemical sources of energy to produce organic carbon from  $\text{CO}_2$ .

The **photoautotrophs** are the **primary producers** of organic carbon in most ecosystems. These organisms use light energy from the sun to convert  $\text{CO}_2$  to glucose and other sugars in a process called **photosynthesis**. This group of organisms includes plants, algae, and photosynthetic prokaryotes. These primary producers are consumed by **heterotrophs** such as animals, which use organic forms of carbon as an energy source and as building material to manufacture their own cell parts. When organisms die, the **decomposers** break down their tissues. They, too, use organic carbon compounds as energy sources and as building material.

The heterotrophs carry out **aerobic respiration** and some other types of energy-releasing (catabolic) metabolism, which break down organic forms of carbon and produce

CO<sub>2</sub>, returning carbon to the atmosphere. Plants and other phototrophs also release some CO<sub>2</sub> when photosynthesis is not occurring, at night, for example.

Organisms in an ecosystem depend on other organisms to supply their metabolic needs. If the photoautotrophs are removed, then most organic carbon will be converted to CO<sub>2</sub> and organic forms of carbon will not be available for animals and other heterotrophs. If the decomposers (such as fungi and bacteria) are removed, then much of the organic carbon cannot be recycled to the atmosphere when organisms die, and the autotrophs will not be able to carry out photosynthesis.

An ecosystem includes both **biotic** (living) and **abiotic** (nonliving) components. Both biotic and abiotic factors affect global CO<sub>2</sub> levels. Much of the earth's carbon is bound up in sedimentary rock, buried in the form of coal and other "fossil fuel" deposits, or dissolved in deep water. These forms of carbon are returned to the atmosphere very slowly. Some carbon is stored in living organisms, and some exists as hummus, dead organic matter in the soil. These forms of carbon can be returned to the atmosphere over a much shorter time frame.

### **Atmospheric CO<sub>2</sub> levels and climate change**

Global CO<sub>2</sub> levels have been intensively studied, in part because CO<sub>2</sub> acts as a greenhouse gas and increasing atmospheric CO<sub>2</sub> levels may be causing global warming. Carbon dioxide, methane (CH<sub>4</sub>) and other gasses in the atmosphere act like the glass ceiling of a greenhouse. Sunlight passes through the atmosphere, warms the earth's surface and radiates back toward space as longer wavelength radiation in the infrared range, also known as heat. The so-called greenhouse gasses block the passage of these wavelengths, trapping heat at the surface. Without a greenhouse effect the average global temperature on earth would be -18 °C (about 0 °F), well below the freezing point of water. With this planetary "blanket" in place the average global temperature is about 15 °C (59 °F), making liquid water available to living organisms.

Atmospheric CO<sub>2</sub> levels have been measured directly since 1958 at Mauna Loa in Hawaii by scientists from the Scripps Institute of Oceanography and since 1959 by the National Oceanic and Atmospheric Administration (NOAA). The level measured in 1959 was 315.97 parts per million (ppm, measured as the dry air mole fraction). Atmospheric

CO<sub>2</sub> levels from earlier time periods (up to about 800,000 years ago) can be measured by examining air trapped in Antarctic ice. These levels fluctuated over long periods of time and were lowest between 15,000 and 30,000 years ago during the last ice age. More recently, preindustrial levels (from the 1700s) are estimated to have been approximately 280 ppm. Since then, the level has increased more than 40 % and now averages over 400 ppm. During the same time period average global temperatures have increased, with 2022 having an average global temperature 1.55°F greater than the 20<sup>th</sup> century average.

For time periods more than 800,000 years ago estimation of atmospheric CO<sub>2</sub> levels is more difficult and results using various methodologies do not always agree. These studies depend on comparing levels of radioactive isotopes in marine sediments and sedimentary rocks from various locations. During some time periods both CO<sub>2</sub> levels and temperatures were likely much higher than today.

## **5. Detection of Carbon Dioxide with Bromothymol Blue**

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When carbon dioxide dissolves in water, it combines with water to form carbonic acid (H<sub>2</sub>CO<sub>3</sub>), a weak acid.



Bromothymol blue is a dye that is blue when the pH of a solution is 7.6 or above and yellow when the pH is 6.0 or below. It can be used to detect changes in the pH of a solution. At alkaline pH the dye loses hydrogen ions (protons) and is described as being in its deprotonated state. This form of bromothymol blue is blue in color. At lower pH bromothymol blue gains a hydrogen ion and is described as being in its protonated state. This form of bromothymol blue is yellow in color.

In this experiment, we will use bromothymol blue to detect the presence of carbon dioxide in a solution. When carbon dioxide is added to a solution the pH should decrease. If carbon dioxide is removed from a solution the pH should increase.

**Table 1: Bromothymol Blue pH Indicator**

pH	acidic	alkaline
State	protonated	deprotonated
Color	yellow	blue
CO <sub>2</sub>	high	low

## **6. Metabolism Review**

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### *Procedure*

1. Write the summary reaction for aerobic respiration in your lab book. Does this process produce or eliminate carbon dioxide?
2. Write the summary reaction for photosynthesis in your lab book. Does this process produce or eliminate carbon dioxide?
3. Write the summary reaction for alcoholic fermentation in your lab book. Does this process produce or eliminate carbon dioxide?
4. Write the reaction that occurs when carbon dioxide is dissolved in water. Will the pH of a solution increase or decrease when carbon dioxide is added to a solution? What color should the pH indicator bromothymol blue be in a solution that has a large amount of carbon dioxide dissolved?
5. Set up a table in your lab book to record sample color and whether CO<sub>2</sub> is high or low for: water + glucose, yeast, water, plant in light, plant in the dark, and animal.

## 7. Determining the role of fungi (yeast) in the carbon cycle

The fungi are decomposers which are also known as **detriovores**. Together with some types of bacteria these organisms use plant and animal remains for food and play an important role in recycling nutrients in most ecosystems. The yeasts are unicellular fungi. *Saccromyces cerevisiae* is commonly used to bake bread. This organism uses aerobic respiration to metabolize sugars when oxygen is available. In the absence of oxygen *Saccromyces* switches over to alcoholic fermentation which converts glucose to alcohol and CO<sub>2</sub>.

### ***Procedure***

1. Write a hypothesis concerning what type(s) of metabolism you expect to see occurring in yeast and whether yeast will produce or use carbon dioxide. Write a prediction about the color that you expect a solution with growing yeast to turn when you add bromothymol blue.
2. Obtain one 15 mL tube with a perforated lid. Mark your tube so that you can identify it. Fill half full with 5% glucose solution in dechlorinated tap water. Fill the remainder of the tube with 7% yeast suspension in dechlorinated tap water. Fill all the way to the top so that there is no air space.
3. Incubate the tube right side up in a 35°C water bath for 30 minutes.
4. Without resuspending the yeast cells remove 1 ml of liquid from the tube and place in a labeled microfuge tube. Balance with a tube from another group or with a tube containing water. Centrifuge on high for 5 minutes. Remove 900 µL of **supernatant** into a second labeled microfuge tube without resuspending the **pellet** (yeast cells).
5. Add 100 µL Bromothymol blue to the yeast supernatant. Vortex to mix.

6. Pipette 450  $\mu\text{L}$  of dechlorinated tap water and 450  $\mu\text{L}$  5% glucose into another tube. Add 100  $\mu\text{L}$  Bromothymol blue and mix. Label this sample glucose negative control.
7. Visually inspect each sample and record the color in your table.

## **8. Determining the role of Plants in the carbon cycle**

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*Elodea* is a water plant that is commonly found in aquariums. As mentioned above, plants are the primary producers in many ecosystems and use photosynthesis to convert  $\text{CO}_2$  to sugars and other forms of organic carbon. When light is absent (at night, for example) plants must switch from **photosynthesis** to other types of metabolism, usually **aerobic respiration**, to produce energy.

### *Procedure*

1. Write a hypothesis concerning what type of metabolism you expect to see occurring in a plant **exposed to light** and whether this plant will produce or use carbon dioxide. Write a prediction about the color that you expect a solution with a plant exposed to light to turn when you add bromothymol blue.
2. Write a hypothesis concerning what type of metabolism you expect to see occurring in a plant **placed in the dark** and whether this plant will produce or use carbon dioxide. Write a prediction about the color that you expect a solution with a plant placed in the dark to turn when you add bromothymol blue.
3. Remove 1 mL of water from each plant tube into labeled microfuge tubes (label 'dark' and 'light'). Centrifuge on high for 1 minute. Remove 900  $\mu\text{L}$  of each supernatant into a second labeled microfuge tube.
4. Add 100  $\mu\text{L}$  Bromothymol blue to each tube. Vortex to mix.

5. Pipette 900  $\mu\text{L}$  of dechlorinated tap water into another tube. Add 100  $\mu\text{L}$  Bromothymol blue and mix. Vortex. Label this sample “water negative control.”
6. Visually inspect each sample and record the color in your table.

## **9. Determining the role of Animals in the carbon cycle**

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1. Write a hypothesis concerning what type of metabolism you expect to see occurring in humans and whether a human will produce or use carbon dioxide. Write a prediction about the color that you expect a solution that a human has blown into to turn when you add bromothymol blue.
2. Fill a 100 mL flask to the 25 ml mark with de-chlorinated tap water.
3. With a straw, blow gently into the solution for one minute.
4. Remove 900  $\mu\text{L}$  of the solution into a clean microfuge tube.
5. Add 100  $\mu\text{L}$  Bromothymol blue to this tube. Vortex to mix.
6. Visually inspect the sample and record the color in your table.

## **10. Conclusion and Summary**

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1. What color is Bromothymol blue at acidic pH? At basic pH?
2. If a sample is yellow what does that tell you about the carbon dioxide concentration? Which samples likely contained the most carbon dioxide?
3. Did the results support your hypothesis for the yeast experiment? Why or why not?

4. Did the results support your hypotheses for the plant experiment? Why or why not?
5. Did the results support your hypothesis for the animal experiment? Why or why not?
6. Which organisms are autotrophs (produce their own food such as glucose)? Which organisms are heterotrophic and can break down but not produce organic forms of carbon such as glucose? Which organisms are considered decomposers?
7. Based on the type of metabolism seen in the organisms tested what can you conclude about whether these organisms remove carbon dioxide from the environment or add carbon dioxide to the environment?
8. What are two possible sources of error that could have affected the reliability of your results?
9. Describe three ways the investigation could be extended.

### **Sources**

Global Warming: Frequently Asked Questions  
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