Background Concepts:

Plants can carry out both photosynthesis and respiration simultaneously. During photosynthesis, plants are using the energy of the sun to build molecules which effectively store this energy (glucose). Chemically, the photosynthetic reaction looks like this:

$$6CO_2 + 6 H_2O$$
 \rightarrow $C_6H_{12}O_6 + 6 O_2$

During respiration, plants are using this stored energy (glucose), to fuel their metabolic processes. Chemically, the respiratory process looks like this:

$$C6H12O6 + 6O2 \rightarrow 6CO2 + 6H2O + energy$$

Remember that plants respire all the time.

Among other things, the converted energy from respiration is used to synthesize molecules, move materials around within the organism, grow (create new cells) and reproduce. Notice that in **photosynthesis**, CO₂ (carbon dioxide) is being used up as it is "fixed" into glucose molecules. During **respiration** the opposite is true. As the plant releases the energy stored in glucose by breaking it down, CO₂ is being given off into the surrounding water or atmosphere. The relationship between these two processes is special in that it allows plants to recycle some of their by-products. (While CO₂ is being given off during respiration, it can be re-utilized during photosynthesis.)

In this lab, you will try to demonstrate the net change in carbon dioxide when the common fresh water plant *Elodea* is placed under different conditions. You will be using a **chemical indicator**, **bromthymol blue**, as a means of determining the presence or absence of CO₂. This solution changes color when CO₂ is introduced. Bromthymol blue changes color due to a change in pH. When CO₂ is dissolved in water, it forms carbonic acid. This lowers the pH of the solution and causes the bromthymol blue to change its appearance.

Purpose:

Your lab group is asked to design, execute, and analyze an experiment that tries to accomplish the following two tasks:

- A. Demonstrate that environmental CO₂ is used during photosynthesis in *Elodea*.
- B. Demonstrate that there is a net production of CO₂ when *Elodea* respires in the absence of photosynthesis.

Supplies and Equipment Available:

- *Elodea* plants
- Aluminum foil
- Large clean test tubes
- Bromthymol blue working solution
- Tape and marking pen or wax pencil
- Straws
- Large Test tube racks
- Flasks: 250 ml (1/group) 500 ml
- Corks or Parafilm for sealing test tubes
- 100 ml graduated cylinder
- Safety goggles
- light source

Getting Started:

1. Your group should obtain a solution of bromthymol blue. This solution changes colour when CO2 is introduced. Bromthymol blue changes colour due to a change in pH. When CO2 is dissolved in water, it forms carbonic acid. This lowers the pH of the solution and causes the bromthymol blue to change its appearance.

Remember that you are a living generator of CO2. Put on your **safety goggles** for this part. Use a straw to gently exhale into a 250 ml flask containing 20 ml of bromthymol blue. Continue exhaling into bromthymol blue solution for about one minute.

CAUTION: Be careful not to swallow any bromthymol or splash it in your face. It is toxic if swallowed.

Record your observations on your Lab Report Sheet. Do not discard this solution. You will need it in Step 3.

What happened when you exhaled into the bromthymol solution? Why?

- 2. When CO₂ is removed from bromthymol blue solution, the solution turns back to its original color (blue).
- 3. To confirm this, label the 250 ml flask into which you have just exhaled CO₂. Set it aside for 24 hours with the top uncovered. The CO₂ in solution will eventually achieve equilibrium with atmospheric CO₂. The air around us contains relatively little CO₂. Therefore most of the CO₂ molecules bubbled into the solution should leave.

Your group will use sprigs (pieces) of *Elodea*, a water plant. The bromthymol blue will not interfere with respiration or photosynthesis in *Elodea*. You have test tubes and corks at your disposal, as well as other materials, including aluminium foil (which is an excellent way to provide a plant with a totally dark environment). You may use as many test tubes as are needed for you to design a controlled experiment.

Procedure:

Day 1: Now that you are familiar with the behaviour of bromthymol in relation to dissolved CO2, you are ready to use it in designing and executing your lab. Go back and read the original "Purpose." The *Elodea* sprigs can be cut to fit into the test tube provided. This allows you to give the *Elodea* a variety of controlled conditions. The test tubes may be filled with solutions containing abundant CO2 or very little. (Where can you get some CO2 gas?) You may choose to include *Elodea* in some but not all test tubes. The aluminium foil can be used to control light. This allows you to set up different test tubes for comparison. Remember to include Controls. You may need more than one test tube for your controls. You may use corks or Parafilm to seal off each of your tubes. Discuss within your group what design might best accomplish the tasks described under "Purpose."

NOTE:

Make sure you use the same amount of bromthymol and CO₂ in your experiment test tubes.

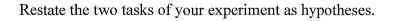
- 1. Find the total volume you will need for all the test tubes that will contain bromthymol with or without CO_2 , and fill a 500 ml flask with this volume.
- 2. Divide this volume into two flasks.
- 3. Carefully blow CO₂ into one of the flasks until it turns yellow. This will ensure that an equal amount of CO₂ is in each test tube

As you set up your experiment, label the test tubes. On your Data Table Design Sheet design a data table to record the contents and appearance of each tube. Remember to include space to record any changes you may observe on Day 2.

Start by recording the original appearance of the tubes. Use a label or tag to identify your test tube rack. Your test tube rack needs to be exposed to a light source wherever your teacher indicates.

Day 2: The test tubes of your experiment have now had approximately 24 hours to carry on respiration and perhaps photosynthesis. The original CO2 concentration of the test tubes has been altered by these two processes. Today you will make observations of your test tubes, record changes, and interpret the data you have collected. Fill in the Data Table with your 24 hour observations and your explanations for the colour changes.

Experimental Design Sheet



Task A -

Task B -

Draw the design of your experiment. Include "controls" in your design. Also include:

- 1. the test tubes
- 2. whether there is a plant in the test tubes or not
- 3. the starting colour of the bromthymol blue solution
- 4. whether the plants will be exposed to light or not

Team Members:

After you have filled out your Experimental Design Sheet, show it to your instructor.

Data Table Design

Your Data Table must include space for each test tube showing:

- 1. the presence or absence of light
- 2. the presence or absence of *Elodea*
- 3. the original colour of the bromthymol blue solution
- 4. a space for the colour of the bromthymol blue solution after 24 hours
- 5. an explanation of why the test tube changed colour

Related Material/Extending the Concepts

1. Plants in the presence of light carry on both photosynthesis and respiration. Write out the equations for Photosynthesis and Respiration (given on the first page of this handout). From the results of your investigation, which process is occurring more in a plant that is being supplied with sunlight? What evidence have you used to come to this conclusion?

2. One of the implications of this investigation is that plants can recycle some of their "waste" products. CO2 is clearly an example of a material that can be recycled by plants. What other gas might plants generate as a "waste" through one metabolic process, but re-use in a subsequent process?

3. Choose one control test tube and one experiment test tube. Use these two test tubes to discuss the role of a "control" in scientific investigations.

Putting it all together:

Related Material/Extending the Concepts

After completion of the lab, as part of your discussion, please answer the following questions:

- 1. Plants in the presence of light carry on both photosynthesis and respiration. Write out the equations for Photosynthesis and Respiration (given on the first page of this handout). From the results of your investigation, which process is occurring more in a plant that is being supplied with sunlight? What evidence have you used to come to this conclusion?
- 2. One of the implications of this investigation is that plants can recycle some of their "waste" products. CO2 is clearly an example of a material that can be recycled by plants. What other gas might plants generate as a "waste" through one metabolic process, but re-use in a subsequent process?
- 3. Describe what was in each of your control test tubes.
- 4. Choose one control test tube and one experimental test tube. Use these two test tubes to discuss the role of a "control" in your scientific investigation.

Lab Report Sheet (to hand in)

Restate the two tasks of your experiment as hypotheses.	
Tasl	k A -
Tasl	k B -
Answer the following questions:	
1.	What happened when you exhaled into the bromthymol blue solution? Why?
2.	Explain which tubes provide evidence for Task A.
3.	Explain which tubes provide evidence for Task B.
4.	Explain the control tubes.
5	If your lab results and analysis did not allow you to prove both Task A and Task B.
then you need to redesign a follow-up experiment that will provide the missing data. Explain and illustrate this follow-up experiment in your report, and explain how it	
will provide additional information.	