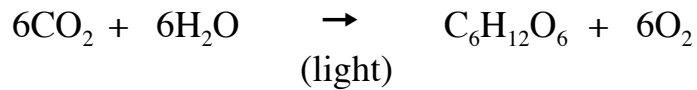


Photosynthesis and Light

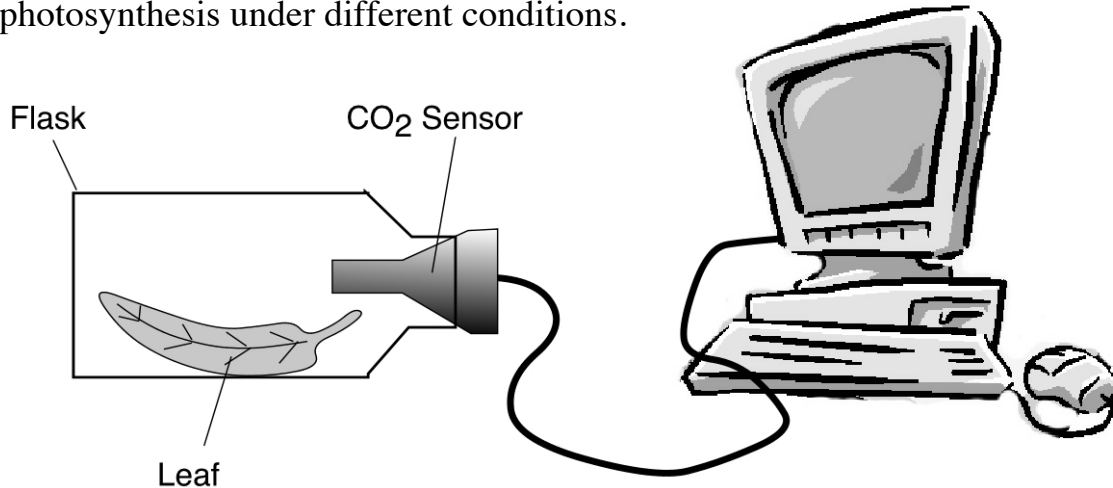
a) Introduction

You are already familiar with the photosynthesis equation:



The equation says that when light is present plants consume carbon dioxide and water to make glucose and oxygen. The plants get the water for photosynthesis from the soil. Carbon dioxide is a gas that is freely available to the plants from the air.

The rate of carbon dioxide consumption (the amount of CO_2 that the plant takes in per minute) is equal to the rate of photosynthesis. In other words, the more photosynthesis a plant is doing the faster it will remove CO_2 from the air. In today's experiment you will place leaves in a sealed flask and then insert a sensor that detects the CO_2 concentration of the air in the flask. A computer will display a graph that shows the decreasing CO_2 as the leaf uses it for photosynthesis. From the graph you will determine the rate of photosynthesis under different conditions.



You might hypothesize that increasing light intensity would increase photosynthetic rate. A related question is whether some colors of light are better than others for photosynthesis. Your lab group will investigate both of these questions today. In addition, you will familiarize yourself with leaf structures under the microscope.

b) Setting up the CO₂ Sensor and the Computer for the Photosynthesis Experiments

- 1) Obtain a white cardboard box that contains the following items:
 - a) A LabPro box (a green plastic electronic device)
 - b) A black power adaptor plug for the LabPro
 - c) A long white USB cable
 - d) A short gray adaptor cable for the CO₂ sensor

Also obtain a CO₂ sensor, a plastic flask, and a laptop computer.

- 2) Plug one end of the power adaptor into your desktop power outlet. Plug the other end into in the LabPro box (see figure below). The LabPro should make a beep a few seconds after it has received power.
- 3) Plug the CO₂ sensor into the round end of the adaptor cable, then plug the adaptor cable into the Channel 1 of the LabPro box (see figure below).
- 4) Plug the square end of the USB cable into the LabPro box (see figure below).



5) Obtain a laptop computer. Turn on the computer. After a few minutes, a log-on window will appear. Ask your instructor for the user name and password. Click the Log On button. The desktop screen should appear on the computer.

6) Plug the flat end of the USB cable into any USB port in the back of the computer. Click the Logger Pro icon on the desktop.

7) Find the File pulldown menu at the top of the screen. Starting with this menu, navigate as follows:

File>Open>Experiments>Biology with Vernier>11B Cell Resp

(If a window called “Sensor Confirmation” appears, click its Done button)

8) Once the 11B experiment is opened, you must reset the experimental run time to 3 minutes, as follows: Click on the Data Collection icon (an image of a clock next to the green button at the top of the screen). A pop up window will appear. In the pop up window, set Length for 3 minutes and Sampling Rate for 30 samples per minute. Then click Done.

9) Test that your set up is working properly by doing the following:

a) The CO₂ level is displayed in the lower right of the screen.

b) Exhale a VERY TINY puff of air into the plastic flask, then insert the CO₂ sensor firmly into the flask. The CO₂ levels in the flask should rise to above 2000 ppm. If the CO₂ level does not rise above 2000, try again with another small puff of air. But do not add so much breath that the CO₂ rises above 4000 ppm.

c) Setting up the Photosynthesis Chamber

- 1) Take the plastic flask outside to the lawn in the quad area. Pull off some grass leaves. Holding the flask horizontally, add enough grass to completely cover the floor of the flask, but don't pile grass on top of the grass that covers the floor of the flask.
- 2) Return to the lab with your flask. Carefully place the CO₂ sensor firmly back into the flask. If the CO₂ concentration is below 2000, remove the sensor and add another tiny puff of air, then replace the sensor back into the flask. But if the CO₂ goes above 4000, lower the CO₂ by opening the flask and fanning into it.
- 3) Gently slide the flask into the black cardboard chamber. The cardboard chamber has an open side on top to allow light to reach the flask. Be sure that the open side of the chamber does **not** have a colored light filter (a colored plastic sheet) on it.
- 4) Position the lamp so that the bulb is very very close (almost touching) the top of the flask. In other words, the bulb should be shining as brightly as possible onto the grass. Ask your instructor to inspect you set up.

d) Photosynthesis in White Light

- 1) Turn the light on and allow it to shine onto the grass leaves in the flask for 4 minutes. The reason for shining the light on the leaves for 4 minutes is that it takes about 4 minutes for the photosynthesis “machinery” in the leaves to reach a steady rate of photosynthesis.
- 2) After 4 minutes of light shining, start collecting data by clicking the green Collect button in the upper right of the screen. If a pop-up window appears, click “Erase and Continue” A red line on the graph should begin to appear. The line shows the CO₂ levels inside the flask.
- 3) As soon as you have clicked the Collect button, record the initial values of the experiment in the blanks below. The initial CO₂ concentration is the CO₂ concentration on the screen for time zero.
Initial CO₂ concentration = _____ ppm.

The CO₂ levels should change during the experiment. The data collection will automatically halt after 3 minutes. During the 3 minute data collection, you can work on the microscopy (part h) of this laboratory.

4) After the data collection has halted at 3 minutes, record the final CO₂ concentration:

Final CO₂ concentration = _____ ppm.

5) Lastly, calculate the photosynthesis rate of the experiment. The graph on your computer shows CO₂ concentration (in ppm) on the Y-axis and time length of the experiment (3 minutes) on the X-axis. The slope of the graph line is the rate of photosynthesis. To calculate the slope of the line (the rate of photosynthesis), divide the change in the amount of CO₂ by the change in time (3 minutes).

Final CO₂ ppm - Initial CO₂ ppm

3 minutes

Photosynthesis rate = _____ CO₂ ppm/minute

6) Although you will not repeat this experiment in dim light or in darkness, you should be aware of what would happen if you did repeat the experiment under those two conditions.

- If you had repeated the experiment under dim light, the rate of photosynthesis would have been lower. In other words, the CO₂ would still have decreased, but it would have decreased at a slower rate than it decreased in bright light.
- If you had repeated the experiment in total darkness (for example, by covering the flask with a coat), the CO₂ in the flask would have increased, not decreased. The reason why the CO₂ would increase is that in darkness plants make energy for themselves using a process called cellular aerobic respiration, which makes CO₂ and consumes O₂.

Show your instructor all the data in this section before continuing with the next section.

f) Photosynthesis in Blue, Green, and Red Light

1) Uncover the chamber with the flask inside. If the CO₂ concentration is below 2000, remove the sensor and add another very tiny puff of air, then replace the sensor into the flask. Open and fan the flask if the CO₂ is above 4000.

2) Place a green colored filter over the open top end of the chamber. Position the lamp so that the bulb is about 7 cm from the desktop/leaves. In other words, the bulb should be very very close (almost touching) the top of the flask. If the filter is placed correctly only green light will shine onto the leaves.

3) Turn the light on and allow it to shine through the colored filter onto the leaves in the flask for 4 minutes. After 4 minutes with the light on, proceed to step 4.

4) Start collecting data by clicking the green Collect button in the upper right of the screen. If a pop-up window appears, click “Erase and Continue”

5) As soon as you have clicked the Collect button, record the initial values of the experiment in the blanks below.

Color of light: _____.

Initial CO₂ concentration = _____ ppm.

6) After the data collection has halted at 3 minutes, record the final CO₂ concentration:

Final CO₂ concentration = _____ ppm.

7) Lastly, calculate the photosynthesis rate of the experiment.

Final CO₂ ppm - Initial CO₂ ppm

3 minutes

Photosynthesis rate = _____ ppm/minute

8) Remove the green filter and replace it with a red filter. Repeat steps 4 - 7 using the red filter. Record the new color and the other new data in the blanks below:

Color of light: _____.
Initial CO₂ concentration = _____ ppm.
Final CO₂ concentration = _____ ppm.
Photosynthesis rate = _____ ppm/minute

10) Remove the red filter and replace it with a blue filter. Repeat steps 4 - 7 using the blue filter. Record the new color and the other new data in the blanks below:

Color of light: _____.
Initial CO₂ concentration = _____ ppm.
Final CO₂ concentration = _____ ppm.
Photosynthesis rate = _____ ppm/minute

Show your instructor these calculations before cleaning up

g) Clean up

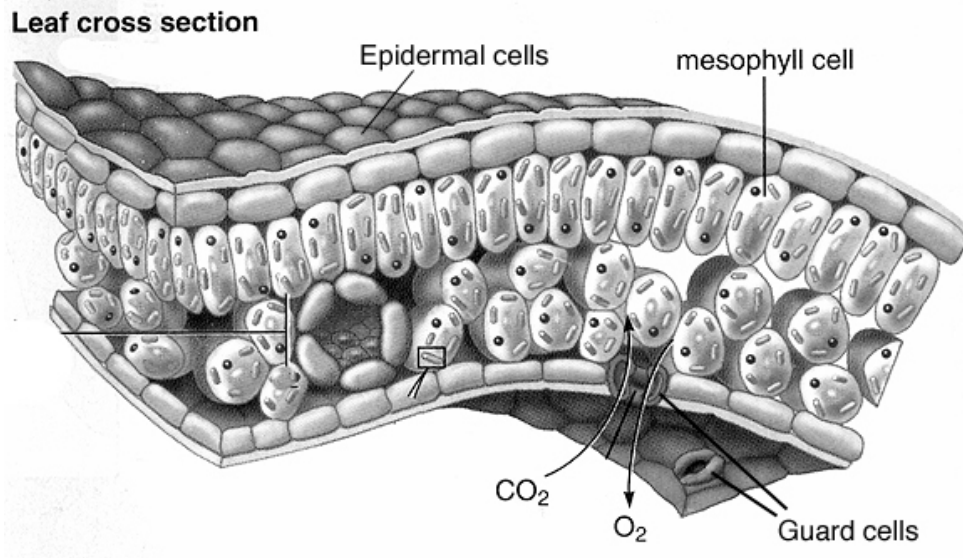
1) Quit the LoggerPro program (don't save your results). Next, shut down your computer using the Shut Down selection on the screen's lower left menu. Wait until the screen has completely turned off before folding your computer closed.

2) Unplug all cables from the blue LabPro box and from the computer. Also unplug the gray adaptor cable from the CO₂ sensor. The CO₂ sensor goes in its own box, but all other cables go into the white cardboard Vernier box.

3) Wrap the lamp cord neatly around the lamp. Empty all leaf pieces out of the flask and dispose of them into the trash.

h) Viewing Leaf Cells

The cells that form the top and the bottom surface of a leaf are called epidermal cells. Their job is to form a water-tight skin to protect the leaf from dehydration. They do not perform photosynthesis. The cells located in the middle of the leaf, called mesophyll cells, are the ones that perform photosynthesis (see the illustration below).



The mesophyll cells need carbon dioxide from the air to perform photosynthesis, and they need a way to get rid of the oxygen produced by photosynthesis. To allow CO_2 and O_2 gases to travel between the mesophyll cells and the air, there are small openings in the lower surface of the leaf. These openings are called stomata. Each stoma is surrounded by a pair of lip-shaped cells called guard cells. The guard cells can open or close the stoma in the same way that your lips can open or close your mouth. The guard cells close the stoma on hot days to protect the leaf from dehydration on hot days.

In this section you will observe mesophyll and guard cells using your microscope.

- 1) Obtain a microscope slide, a cover slip, and one leaf from the water plant elodea. Make a wet mount of one elodea leaf.

2) View the leaf using the 40X objective lens (remember to focus first on 5X and 10X). You should be able to clearly see the chloroplasts inside the mesophyll cells.

3) Sketch one mesophyll cell in the space to the right. Label the cell wall, the chloroplasts, and the cytoplasm in your drawing.

4) Estimate the number of chloroplasts in the cell you sketched. Also estimate the size of the cell you sketched. (You may need to refer back to your microscopy lab to recall how to estimate cell size). There are 1000 μm per one mm.

Chloroplast number: _____ Cell size: _____ μm .

5) Dispose of the elodea leaf. Obtain a leaf from the Mother of Thousands plant (scientific name: *Kalanchoe daigremontiana*). Bend the leaf gently until it snaps. Peel off a piece of the epidermis, which is the thin outermost layer of cells (it looks like a piece of scotch tape). You may need forceps (tweezers) to pull off the epidermis.

6) Make a wet mount of the epidermis and view it under 40X. Notice that most of the cells have irregular shape like pieces of a jigsaw puzzle. Note that, unlike the illustration on the previous page, you are viewing the leaf epidermis face on, not as a cross section.

7) Find a pair of guard cells. They look like pairs of green lips. Each pair of guard cells surrounds one stomata (the hole in the leaf where the CO_2 enters and the O_2 exits). During hot days the guard cells block the stomata to prevent water from evaporation through the opening.

8) Sketch the guard cells in the space to the right. Label the guard cells and the stomata. Include in your drawing several of the jigsaw-like epidermal cells around the guard cells.

9) Wash all slides and put them back in their boxes. The cover slips go into the glass waste box.

i) Review Questions

- 1) Write the complete photosynthesis equation:

- 2) What is the function of...
 - a) Epidermal cells?
 - b) Mesophyll cells?
 - c) Guard cells?

- 3) What gas or gases passes into the stomata? What gas or gases passes out of the stomata?

- 4) Guard cells have a bent shape (like kidney beans). If the plant needs to close the stomata, will the guard cells straighten or bend more?

- 5) At the biochemical level, photosynthesis is described as taking place in two distinct stages. What are those stages called? Which of those stages (the first or the second) was not able to function well in reduced light intensity?

- 6) If you had put the flask in total darkness, no more photosynthesis would have occurred. It might seem at first that the CO₂ should not change at all, yet the CO₂ would increase if the leaves were in darkness. Explain why plants produce CO₂ in darkness .

- 7) Which light color (red, green, or blue) was best for photosynthesis? Which was worst?

- 8) Explain, in terms of pigments and light color absorption, why the worst color light for photosynthesis was the same color as the leaves.

9) Your computer screen created a graph the CO₂ concentration in the flask versus the time of the experiment. If the slope of the graph went downward sharply, does that indicate a high rate of photosynthesis or a low rate of photosynthesis?

10) The stoma and guard cells are on the bottom/top (← circle one word) surface of the leaf.

11) The concentration of gases in the air is given in units called ppm. What does ppm stand for?

12) When you first turned the light on, the CO₂ concentration did not immediately begin to fall. Why is there a lag time before the CO₂ decreases?

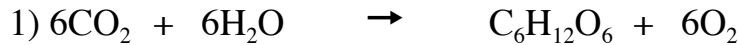
13) If you had used an oxygen sensor instead of a CO₂ sensor, what would the graph of the white light experiment look like? In black ink, sketch the curve in the square below. In blue ink, sketch the oxygen curve if the plants were in darkness.

**Oxygen
(ppm)**



Time

j) Review Question answers



2) a) To form a water-tight barrier to prevent evaporation from the leaf

b) To perform photosynthesis

c) To regulate gases (CO_2 , H_2O , and O_2) entering and exiting the leaf by opening and closing the stoma

3) CO_2 gas enters the stoma. H_2O , and O_2 gases exit the stoma.

4) Guard cells straighten to close the stoma.

5) The two stages of photosynthesis are the light dependent reaction and the light independent reaction. The light dependent reaction does not function well in reduced light intensity.

6) In darkness plants obtain energy using a process called cellular aerobic respiration. This process produces CO_2 .

7) Of the colors of light that you tested (red, blue, and green), green light is the worst (lowest rate) of photosynthesis. Of the colors of light, the highest rate of photosynthesis can be red or blue light, depending on the plant type and the conditions of the experiment.

8) Green light was the worst light for photosynthesis because it reflects off of the leaves instead of being absorbed by the leaves. The fact that green light reflects off of the leaves explains why leaves look green: Any object is the color of the light that it reflects into our eyes.

9) A sharply downward slope of a CO_2 concentration versus the time graph means that there was a high rate of photosynthesis.

10) The stoma and guard cells are on the bottom surface of the leaf.

11) Parts per million

12) There is a short delay (typically a few seconds to a few minutes) between shining the light on the plants and the decrease in CO₂ because it takes time for the enzymes of the light dependent reaction to generate enough high energy molecules for the light independent reaction to convert CO₂ into glucose.

13)

