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Connect and Collect:
Photosynthesis in Minutes

Experiments:

Photosynthesis and Respiration

- Go Direct CO_2 Gas

Plant Pigments

- Go Direct SpectroVis® Plus

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Photosynthesis and Respiration

(CO_2 Gas Sensor)

Plants make sugar, storing the energy of the sun into chemical energy, by the process of photosynthesis. When they require energy, they can tap the stored energy in sugar by a process called cellular respiration.

The process of photosynthesis involves the use of light energy to convert carbon dioxide and water into sugar, oxygen, and other organic compounds. This process is often summarized by the following reaction:

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

Cellular respiration refers to the process of converting the chemical energy of organic molecules into a form immediately usable by organisms. Glucose may be oxidized completely if sufficient oxygen is available by the following equation:

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

All organisms, including plants and animals, oxidize glucose for energy. Often, this energy is used to convert ADP and phosphate into ATP.

OBJECTIVES

- Use a CO_2 Gas Sensor to measure the amount of carbon dioxide gas consumed or produced by a plant during respiration and photosynthesis.
- Determine the rate of respiration and photosynthesis of a plant.

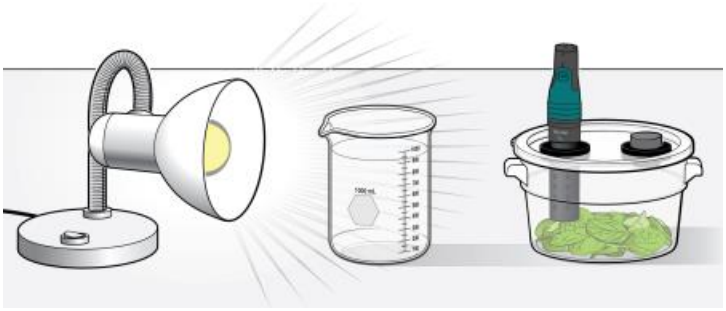

Figure 1

MATERIALS

Chromebook, computer, **or** mobile device
Graphical Analysis app
Go Direct CO_2 Gas
BioChamber 2000
600 mL beaker
aluminum foil
lamp with bulb
#6 rubber stopper
spinach leaves
goggles

PROCEDURE

1. Wrap the BioChamber with aluminum foil so that no light will reach the leaves.
   a. Wrap the outside of the chamber with foil.
   b. Cover the lid with foil, poking the holes open to insert the sensor and the rubber stopper.
2. Cover the bottom of the chamber with a one centimeter layer of fresh, turgid spinach leaves.
3. Launch Graphical Analysis. Connect the CO_2 Gas Sensor to your Chromebook, computer, or mobile device.
4. Set up the data-collection mode.
   a. Click or tap Mode to open Data Collection Settings.
   b. Change Rate to 15 samples/min and End Collection to 15 min. Click or tap Done.
5. Change the unit to ppt by clicking or tapping the CO_2 meter and choosing ppt from the Units menu.
6. Secure the lid on the chamber. Insert the CO_2 Gas Sensor into one the holes and the rubber stopper into the other.
7. Wait five minutes for the sensor to equilibrate, then click or tap Collect to start data collection. Data will be collected for 15 minutes.
8. When data collection is complete, determine the rate of respiration/photosynthesis.
   a. Click or tap Graph Tools, , and choose Apply Curve Fit.
   b. Select Linear as the curve fit. Click or tap Apply.
   c. Enter the slope of the line, *m*, as the rate of respiration/photosynthesis in Table 1.
   d. Dismiss the Linear curve fit box.
9. Make a heat sink by filling a 600 mL beaker with water.
10. Set up the lamp and heat sink as shown in Figure 1. **Important**: Do not turn the lamp on until instructed to do so.

11. Remove the aluminum foil from the respiration chamber.
12. Turn on the lamp.
13. Repeat Steps 8–10 to collect and analyze data for photosynthesis. **Note:** Data from the previous run will automatically be stored.
14. Graph both runs of data on a single graph.
    a. To display multiple data sets on a single graph, click or tap the y-axis label and select the data sets you want to display. Dismiss the box to view the graph.
    b. Use the displayed graph and Table 1 to answer the questions below.
15. Clean and dry the respiration chamber.

DATA

Table 1	
Leaves	Rate of respiration/photosynthesis (ppt/min)
In the dark	
In the light	

QUESTIONS

1. Were either of the rate values a positive number? If so, what is the biological significance of this?
2. Were either of the rate values a negative number? If so, what is the biological significance of this?
3. Do you have evidence that cellular respiration occurred in leaves? Explain.
4. Do you have evidence that photosynthesis occurred in leaves? Explain.
5. List five factors that might influence the rate of carbon dioxide production or consumption in leaves. Explain how you think each will affect the rate?

EXTENSIONS

1. Design and perform an experiment to test one of the factors that might influence the rate of carbon dioxide production or consumption in Question 5.
2. Compare the rates of photosynthesis and respiration among various types of plants.

PRELIMINARY ACTIVITY FOR

Plant Pigments

Guided Inquiry Version

Plants contain many different molecules directly or indirectly involved with photosynthesis, which may also impart color to the plant. The mixture of chlorophyll molecules found in spinach, for example, absorbs several wavelengths of visible light, with distinct absorbance peaks in the blue range (400–500 nm) and in the yellow-red range (600–700 nm). The combination of visible light that is not absorbed appears green to the human eye. Chlorophyll contains a porphyrin ring in its structure with a magnesium ion in the center. The porphyrin ring accounts for much of the molecule's light absorbance. Chlorophyll is found in the thylakoid membrane of a plant chloroplast.

Carotenoids are accessory pigments associated with many colors observed in vegetation. There are hundreds of different types of carotenoids. Carrots get their color, which is often orange but is not restricted to orange, from carotene. Carotene is a family name for several compounds that also go by the name terpene.

Another type of carotenoid plant pigment is called anthocyanin. The purplish color of a red cabbage and the rusty red of the flesh of a blood orange are a result of the presence of anthocyanins, which also have the property of changing color with changes in pH. Anthocyanins absorb UV light, which is used by plants to perform two important functions: to attract insects, which augment pollination, and as a "sunscreen" to protect the other parts of the plant cells such as DNA from harmful UV radiation.

Another characteristic of chlorophyll is it is a fluorescent substance. Fluorescent substances can absorb light of one wavelength and then reemit a new and longer wavelength of light. Chlorophyll absorbs light in the violet and blue regions of the visible spectra. If a violet or blue light is shined through a sample of spinach extract, the solution turns red in color. The intensity of the red color is an indication of how much chlorophyll is in the sample.

In this Preliminary Activity, you will use a spectrophotometer to determine and analyze the visible light absorbance spectrum of spinach extract.

After completing the Preliminary Activity, you will investigate your assigned researchable question. Use reference sources to find out more about plant pigments, visible light spectra, and fluorescence before planning and conducting your investigation.

PROCEDURE

1. Obtain and wear goggles.
2. Prepare the spinach extract.
    a. Measure out 0.5 g of fresh spinach and cut or tear the spinach into tiny pieces.

b. Grind the spinach pieces with a mortar and pestle.

c. Add 20 mL of 70% isopropyl alcohol and transfer the mixture to a small beaker. **DANGER**: *Isopropyl alcohol*: *Keep away from heat, open flames, and hot surfaces—highly flammable liquid and vapor. Do not breathe mist, vapors, or spray. Causes eye irritation. Causes damage to organs. May cause drowsiness or dizziness.*

d. Allow the mixture to sit for about 10 minutes.

e. After 10 minutes have elapsed, use a funnel and filter paper to filter the spinach extract into a clean beaker.

3. Launch Spectral Analysis. Connect the SpectroVis Plus to your Chromebook or computer. Select Absorbance vs. Wavelength.

4. Prepare a blank by filling an empty cuvette 3/4 full with 70% isopropyl alcohol. To correctly use a cuvette, remember:

- Wipe the outside of each cuvette with a lint-free tissue.
- Handle cuvettes only by the top edge of the ribbed sides.
- Dislodge any bubbles by gently tapping the cuvette on a hard surface.
- Always position the cuvette so the light passes through the clear sides.

5. To calibrate the spectrophotometer, place the blank cuvette in the spectrophotometer and select Finish Calibration. **Note**: If necessary, wait for the spectrophotometer to warm up before selecting Finish Calibration.

6. Conduct a full spectrum analysis of the spinach extract.

a. Empty the blank cuvette and rinse it twice with small amounts of the spinach extract. Fill the cuvette 3/4 full with spinach extract.

b. Place the cuvette containing the spinach extract into the spectrophotometer.

c. Start data collection. A full spectrum graph of the spinach extract will be displayed.

d. Stop data collection.

e. Examine the graph, noting the absorbance peak ranges for chlorophyll described in the introductory remarks. If any of the peak absorbance values are greater than 1.5, dilute your sample to an extent that will bring the peaks below 1.5 and repeat the data collection.

7. Print or save your experiment file as directed.

Part II Collect a Fluorescence Spectrum of Spinach Extract

8. Ensure that the cuvette containing the spinach extract is in the cuvette slot of the spectrophotometer.

9. Click or tap File, 🗋, and choose New Experiment. Select Fluorescence vs. Wavelength.

10. Set up the SpectroVis Plus to measure fluorescence.

a. Change Integration Time to 150 ms in the Collection Settings menu.

b. Change Excitation Wavelength to 405 nm. Click or tap the graph to dismiss Spectrometer Settings.

11. Start data collection. A full spectrum graph of the fluorescence of the sample will be displayed. Note that one area of the graph contains a peak at approximately 675 nm. This peak is from chlorophyll.

12. Stop data collection. The height of the peak should be between 0.6 and 1.0. If necessary, adjust the integration time to increase or decrease the size of the fluorescent peak and repeat the data collection.

13. Answer Question 2.

QUESTIONS

1. Describe the visible light absorbance spectrum of the spinach extract, identifying the absorbance peaks and other distinguishing features.

2. List five colorful plant flowers, fruits, leaves, or other parts whose extracts might contain plant pigments of interest in visible light spectra.