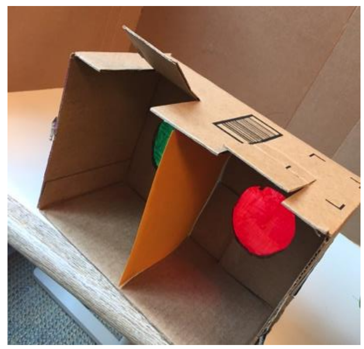
Photosynthesis: How do plants get energy?
Teacher Version

In this lab, students explore the process of photosynthesis in spinach leaves. As oxygen is produced, the density of the leaves change and they will begin floating in a sodium bicarbonate solution. The time it takes for a certain number of leaves to float can be used to calculate the rate of photosynthesis.

California Science Content Standards:

- **1. Cell Biology: The fundamental life processes of plants and animals depend on a variety of chemical reactions that occur in specialized areas of the organism's cells.**
- 1f. Students know usable energy is captured from sunlight by chloroplasts and is stored through the synthesis of sugar from carbon dioxide.

Preparation and Lab Notes:



1. Trace two circles on the underside of one shoe box, about 3 inches in diameter. Position the circles at opposite ends. Do the same for the second shoebox, but only trace one hole on one end.
2. Cut out the circles. Label the holes 1, 2, and 3. Label the area not cut out on the second shoebox 4.
3. Along the long side edge of each shoebox, cut two flaps about three inches high, spaced just as the holes. Fold the flaps outward. These will allow for observations of the cups within.
4. Lay each shoebox on its short end over a piece of cardstock. Trace and cut out the card. Insert the card as a divider such that the boxes are evenly divided in half. Fix in place with tape.

Complete List of Materials: (for one experimental set-up)

- 1 8 oz. box baking soda
- 1 L water
- Teaspoon
- Graduated beaker (200mL)
- 5 transparent cups
- single hole punch
- 7 fresh spinach leaves
- Small bottle of dish soap, clear (if available)
- 1 large desk lamp (>60 watt bright white, if possible)
- 2 sheets of cellophane: one green, one red (transparencies colored with markers work)
- 4 plastic syringes (10 mL)
- ruler
- 2 shoe boxes, pre-cut according to pre-lab directions
- 2 pieces of cardstock or heavy paper, pre-cut according to pre-lab directions
- 1 L graduated beaker

Key Concepts:

- **Photosynthesis** is the primary means by which plants get their energy. They derive this energy from a sugar called **glucose** ($C_6H_{12}O_6$).
- To make glucose, sunlight is captured in pigments like **chlorophyll**, the substance that gives leaves their green color.
- The sun's energy is passed through a chain of events that breaks **water** (H_2O) into **oxygen** (O_2) and creates a store of energy-rich molecules. These molecules will enter a continuous cycle of events to build glucose out of **carbon dioxide** (CO_2) from the air.

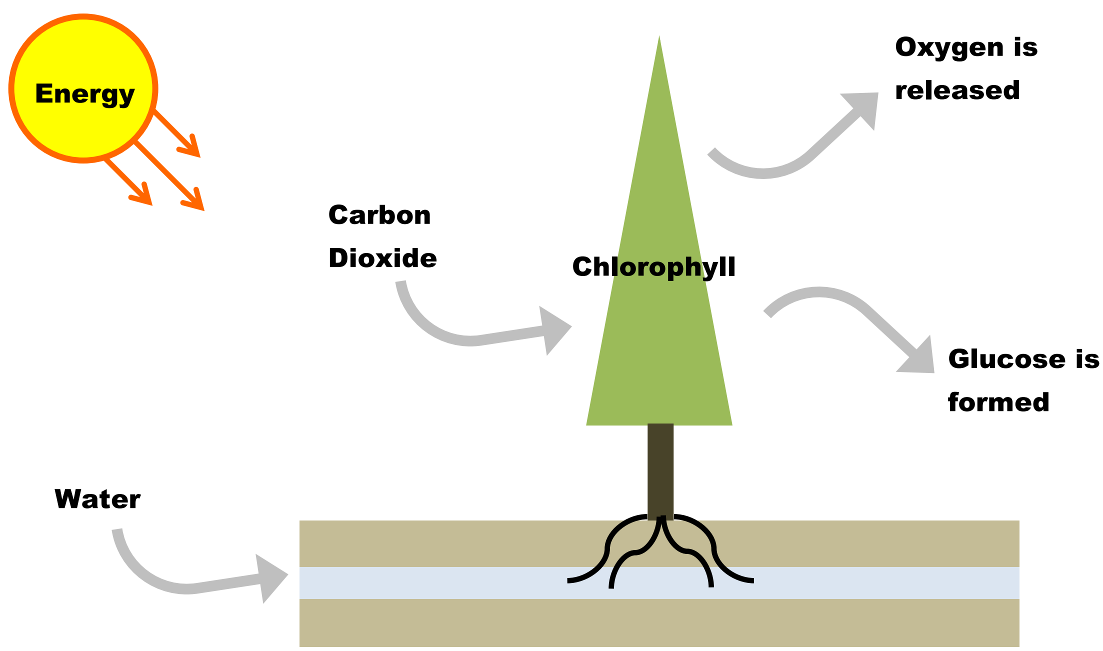
Introductory Mini-Lecture:

The chain of events that breaks apart water are called the **Light Reactions** because they only happen when light is shining on the leaf. The cycle of events that builds glucose are called the **Dark Reactions** because they are going on both day and night and don't require light to be shining. The whole process together can be summarized like this:

$$6H_2O + 6CO_2 \xrightarrow[\text{Sunlight}]{} C_6H_{12}O_6 + 6O_2$$

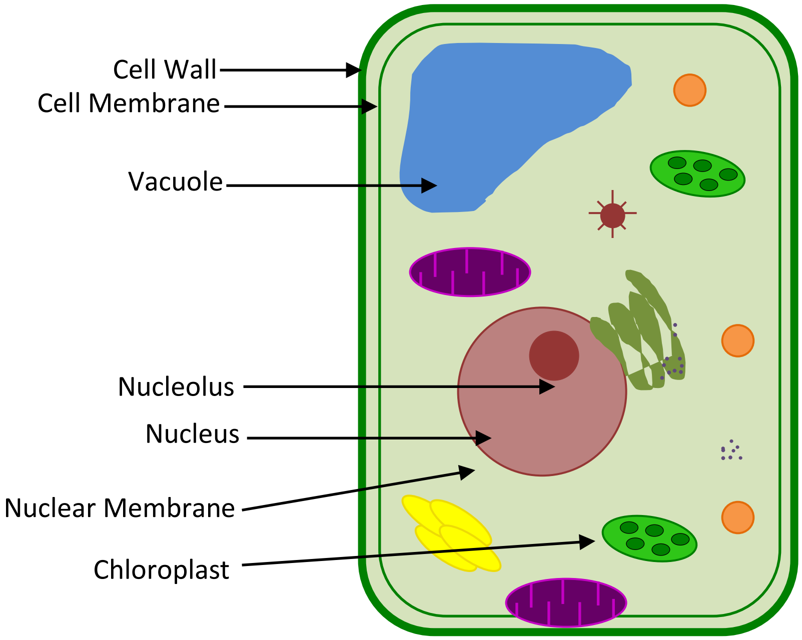
(Six parts water) and (six parts carbon dioxide) result in (one part glucose) and (six parts oxygen)

Some of the oxygen is used by the plant, but most of it is released into the atmosphere, to be breathed in by animals. In return, animals breathe out carbon dioxide, which plants use to create more oxygen (and food for the animals).



Photosynthesis

Within a plant cell are green chloroplasts, where all the events of photosynthesis take place.

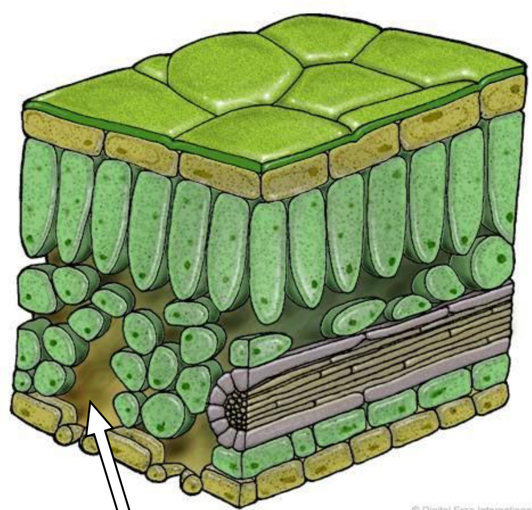


Cross-section of a plant cell

The central vacuole, shown in the center, stores the product of photosynthesis: sugar (in various forms like starch).

This lab focuses on several different factors that affect the net rate of photosynthesis. We will be comparing the rate of photosynthesis in different light intensities -including in the dark- by viewing the number of leaf disks that rise to the surface of a special solution over time. The solution contains two important ingredients:

- Sodium Bicarbonate (baking soda) – a source of carbon dioxide when dissolved in the solution
- Dish detergent – soap that breaks down the water-repellent barrier on the leaf surface so sodium bicarbonate can get inside



As the leaf disks absorb the sodium bicarbonate solution, the density of the leaf increases, it becomes heavier, and the leaf sinks. This experiment takes advantage of the fact that when oxygen leaves the Light Reactions chain, it gets trapped in the inner spaces of So as photosynthesis takes place in the leaf disk, tiny oxyge e solution from the leaf spaces and make the leaf disks light enough to rise. Since the amount of oxygen released into the leaf interior represents the excess oxygen produced after the Light Reactions, the rate that the disks rise is proportional to the net rate of photosynthesis.

The time at which half of the original number of disks are floating can be used to compare net rates of photosynthesis. The value has an inverse relationship with the net rate of photosynthesis; as the net rate increases, the time for half of the disks to float decreases.

Hypotheses:

QS1. Do you think a light green or a dark green leaf has more chlorophyll?

QS2. Will photosynthesis work in the dark?

QS3. Do you think chlorophyll works better under a certain color of light? Does chlorophyll absorb all the light that hits a leaf?

QSA1. What do you think the effects of varying the type of plant leaf we use will be on the net rate of photosynthesis (i.e. which one will have a faster rate, the cabbage or the spinach and why?)? (provide literature values and an explanation at the end) Different Plants have different amounts of chlorophyll hence their rates are different. Spinach is clearly much greener than cabbage, which shows which one has a faster rate of photosynthesis (spinach).

QSA2. What do you think the effect of light versus dark will be on the net rate of photosynthesis? Plants need light so the chlorophyll can convert it into energy, so the process will only take place when light is present.

QSA3. How do you think varying the intensity of light (i.e. distance from the light source) affect the net rate of photosynthesis?
The farther away the light source is the longer it takes for the process to occur, and vice versa; the closer the light source is, the faster the rate of photosynthesis.

Procedure:

1) **Label** 5 cups and the 5 syringes in the following manner: “No BS light”, “BS + red”, “BS + green”, “BS light”, and “BS dark”.

2) **Mix** 4 tsp of baking soda and 4 tsp of detergent with 800 mL of water in the beaker. **Label** is as “BS solution”.

3) **Mix** 1 tsp detergent with 200 mL of water into the cup labeled “No BS light”. **Label** this as “No BS solution”

4) **Place** the red cellophane over Hole 1. **Place** the green cellophane over Hole 2. **Leave** Hole 3 open.

Q4. What type of light will enter each region of the boxes? What type do you think is most favorable to plants?

Hole 1 lets in only red light. Hole 2 lets in only green light. Hole 3 lets in a broad spectrum of light of many colors. The dark chamber 4 lets in no light. The broad spectrum of light is most favorable to plants because chlorophyll absorbs light in the red and blue parts of the color spectrum, which no other cup provides.

5) **Hole punch** 70 spinach leaf disks. Note: this experiment works best with very fresh leaves.

6) **Put** 10 disks into each syringe

7) **Insert the plunger** without crushing the disks.

8) **Suck up 3-4 mL** of the BS solution in the BS syringes; **suck up 3-4mL** of the No BS solution in the No BS syringe. Be sure that the leaves are suspended and floating in the solution, not sticking to the sides.

Q5. Why should the leaves still be floating at this stage?
Because the process has not started yet –the leaves have not fully absorbed the solution yet.

9) **Point the syringe tip upward and press the plunger** to remove excess air. There should be virtually no air in the syringe – just a few bubbles.

10) **Create a vacuum** in the syringe by placing your finger on the hole at one end of the syringe and pulling the plunger up. **Hold the plunger** up like this for 10 seconds while swirling the leaf disks in the solution and then release.

Q6. What do you think the purpose of the vacuum is? What do you think the vacuum we made is "sucking"?
The purpose of the vacuum is to force out all air in the inner spaces of the leaf disks. The solution will now have room to flood in and saturate the leaf disk. By "sucking" we are actually pulling the air out of the leaf discs.

11) **Repeat this vacuum** until leaf disks are no longer floating in solution and have sunk to the bottom of the syringe. This may take several strong "tugs". If the leaves fail to sink add more detergent to the solution.

Q7. Why do you think adding more detergent will help?
Students are allowed to answer freely. The soap breaks down the molecules on the leaf's surface (because the leaf repels the solution) to allow the bicarbonate solution to penetrate the cells. That way, the leaf disk can absorb the solution.

12) **Pour the leaf disks** and their solution into the appropriately labeled cup. Fill the BS cups with the baking soda solution from the beaker until the cups are 3/4 full. Note: you already filled the "No BS" cup with the appropriate solution. Check that all disks are resting at the bottom of their cups. Prod or stir the solution with a clean stir stick or pencil to loosen any clinging disks.

13) **Arrange the cups** under the appropriate holes in the shoeboxes. Make sure the side flaps are closed to keep light out. Leave the "No BS" cup uncovered next to the boxes. Place the light so that it shines from above onto the three holes and uncovered cup.

14) Using the side flaps for viewing, **record the number of floating disks** after each minute in the data table handout for each experimental condition until all of the disks are floating. Be sure that none of the disks are stuck to the sides of the cups. (Note: data table handout is one of the attachment files)

Q8. Why do the disks start to float?

Remember that oxygen is a product of photosynthesis. When it is released, it gets trapped in the leaf and has no where to go. It pushes out the heavy solution, so the density of the cell has now changed, making the disk lighter, so it can float.

QS9. What process is at work inside the leaves when light shines on them?

Photosynthesis is using the light energy to convert water (H_2O*) and carbon dioxide (*CO_2*) into glucose (*$C_6H_{12}O_6$*, food energy for the plant) and oxygen (*O_2*).*

QS10. Did any results surprise you?

15) **Graph your results**.

QSA9. What do you think is the most effective way to graph the results? Why are we calculating a "net" rate and not just a rate? What would be the units of the rate, and thus what would be the independent variable (on the x axis) and what would be the dependent variable (on the y axis)?

We are calculating a net rate because what that means is that we are only calculating a part of a photosynthesis rate. Photosynthesis is an on-going process, so we are not able to calculate the entire rate. The net rate is simply the rate we are calculating from the moment we start our experiment.

QSA10. How do we calculate a rate using the graphs?

Finding the rate would be the same as calculating the slope on a graph. The slope is found by locating two points (with coordinates) on the graph and dividing their differences. For example, two coordinate points could be (1,3) and (2,6). The x-coordinates 2 and 1 have a difference of 1. The y-coordinates 6 and 3 have a difference of 3. Now, the new coordinates are (1,3) but these differences must be divided to give you the slope. To get the slope, the y-coordinate must be divided by the x-coordinate, so in this case 3 is divided by 1 and the slope is 3. Calculating the rate would mean taking two points on the graph and finding the slope. The rate = slope.

Concept questions

Q11. What was the best source of light for photosynthesis? Was this what you predicted?

Answers vary

Q12. Describe any sources of error in the experiment. Was there anything that made your results hard to read or gave no results? Why?

Answers vary

QSA13. What other controls would you suggest using in this experiment? We would expect that this prospective experiment would also test the rate of photosynthesis, but under different conditions.

Possible controls: changing temperature of light source, one light source versus multiple, using a different plant (comparing spinach with cabbage, lettuce, etc.).