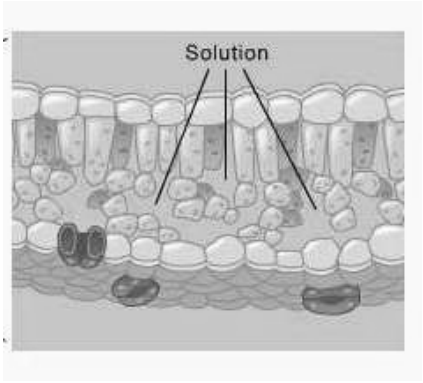
Photosynthesis in Leaf Disks
Teacher Preparation and Background Information

General Information: **Safety:** Goggles should be worn during the experiment. Solutions may be handled without gloves and may be disposed of in sink drains.

In this experiment, students will use a syringe to vacuum the air from the spaces in the spongy mesophyll of leaf disks (do not use thick leaves such as holly). The spaces will then be infiltrated with a sodium bicarbonate ($NaHCO_3$) solution, which contains a tiny amount of detergent to break down the waxy leaf coating (cuticle).
Note: The amount of sodium bicarbonate is approximate. You may wish to test this prior to the experiment, or measure it for the students. The sodium bicarbonate solution adds carbon dioxide to the solution to stimulate photosynthesis.



As solution enters the leaf spaces and forces the air out, the additional mass causes the disks to sink in the solution. The solution should be vacuumed through the leaf disks several times, shaking the syringe after each attempt.

Caution the students to grasp the syringe firmly and keep one finger over the tip to prevent water from spraying out the opening. If the disks do not sink add a little more detergent to the solution.

The leaf disks are then divided equally into 2 cups, the remaining solution is added equally to each. One cup is exposed to light while the other serves as the control, and is covered. As photosynthesis occurs, oxygen is produced which fills the intercellular spaces. The decrease in density causes the leaf discs to rise and float. Observations are made as the disk cells undergo photosynthesis and students will record the time it takes for leaf disks to rise to the surface.

As an extension you may propose that the floating disks be placed in the dark to determine if the disks will sink after photosynthesis ceases.

After mastering the basic techniques in the controlled lab, each pair of students will design and carry out an Experimental Lab with one variable introduced. You may need to provide suggestions and provide equipment: concentration of sodium bicarbonate solution, pH of solution, temperature of solution, intensity of light, color (wavelength) of light, distance from light, type of leaf, and presence of other chemicals such as herbicides that interfere with photosynthesis. (Use safety precautions with chemicals)

Materials: Per Group

Sodium bicarbonate (baking soda)
1 large plastic cup or beaker (app. 500 mL)
2 clear plastic cups or beakers (app. 250 mL)
Hole punch (can be shared by groups)
Fresh Leaves (spinach, ivy, pansy, other of your choosing)
Large plastic syringes (35-65 mL, 1 per group)
Light sources, 60 watt bulb or higher
Liquid detergent
Plastic spoon or straw
Eyedropper
Timers or a clock with second hand
(Other materials for experimental labs)

Photosynthesis in Leaf Disks

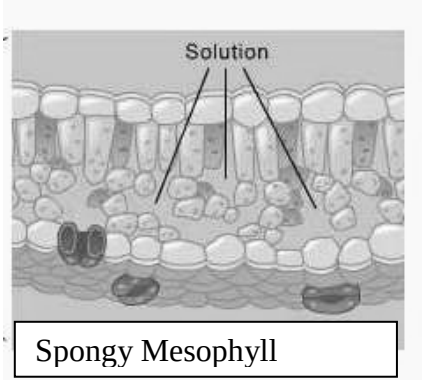
Name_____ Group_____ Date_____

Photosynthesis in Leaf Disk

Introduction:
Photosynthesis is a process in which plants convert light energy (sunlight) into usable chemical energy (carbohydrates). Photosynthesis involves two simultaneous processes: the light dependent reactions and the light independent reactions (Calvin Cycle). In the light dependent reactions, light energy is captured and converted to high energy ATP and NADPH molecules. In the light independent reactions these high-energy molecules are used to reduce CO_2 and eventually form carbohydrates such as glucose.

Overall reaction (unbalanced): $CO_2 + H_2O$ + light energy → Glucose + O_2

In this experiment, the spaces in the spongy mesophyll of leaf disks are filled with a sodium bicarbonate solution, which causes them to sink in the solution. The leaf disks are then exposed to light and observations are made as the cells undergo photosynthesis.



Objectives:
By the end of this activity you should be able to:

- Describe the reactants and products of photosynthesis and the source of reactants from the environment.
- Explain the relationship of photosynthesis to the observations made during the experiment.
- Identify another variable that might affect photosynthesis and design an experiment that uses leaf disks to test your ideas.
- Create hypotheses about the effects of environmental variables on the rate of photosynthesis.

Materials:

- 1.5 g sodium bicarbonate (baking soda)
- Liquid dish soap
- Eyedropper
- Plastic syringe (20-65 mL)—no needle!
- Plastic spoon or straw (for stirring)
- Leaf material
- Hole punch
- 1 large beaker or plastic cup
- 2 small beakers or plastic cups
- Timer or clock with second hand
- Light source
- Paper towels

Photosynthesis in Leaf Discs

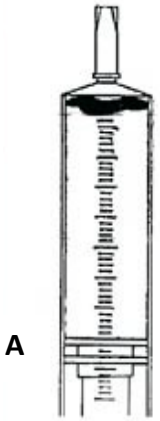
Procedure: **Solutions are safe to handle without gloves.** **Wear Goggles!**

1. Using a one-hole punch, cut 20 leaf disks from young actively growing leaves.

2. Prepare a 0.2% solution of sodium bicarbonate ($NaHCO_3$) and water in the large beaker or clear plastic cup, by adding approximately 1.5 g sodium bicarbonate to 300 mL of water. Stir until dissolved. Use an eyedropper to add about 2 drops of dish detergent to the solution and stir gently. There should be no bubbles afterward.

3. Remove the plunger from a large clean syringe (no needle). Place 20 leaf disks into the body of the syringe. Be sure the leaf disks are near the tip of the syringe as you re-insert the plunger so as not to damage the disks. **(A)**

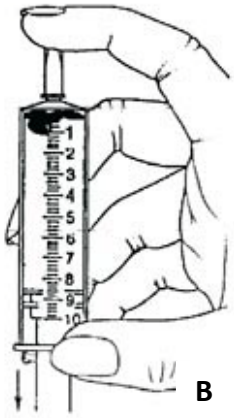
4. Insert the tip of the syringe into a beaker of 0.2% sodium bicarbonate solution and draw 15-20 mL into the syringe. The leaf disks should be floating at this time. If your syringe is smaller than 60ml fill it about one third full.



A

5. Hold the syringe tip upward and expel the air by depressing the plunger carefully. **Stop** before solution comes out the tip.

6. Seal the tip of the syringe using the index finger of your left hand and hold tightly. Pull back on the plunger creating a partial vacuum within the syringe. If you have a good seal it should be hard to pull on the plunger and you should see bubbles coming from the edge of the leaf disks. Hold for a count of ten. **(B)**



B

7. Simultaneously, release your index finger and the plunger. Some of the leaf disks should start to sink. Tap the side of the tube or shake gently to break any bubbles on the edges of the disks.

8. Repeat steps 6 and 7 until all the disks sink. Do not overdo these steps!! You have been successful if the disks sink to the bottom. Don't repeat "just to be sure" as it is possible to damage the cells of the leaves.

9. Remove the plunger from the syringe and pour the solution containing the disks into 2 plastic cups or beakers and **add the remainder** of your solution equally to both beakers. There should be 10 disks per cup. Make sure they sink to the bottom.

10. Cover ONE of the beakers to block light from the leaf disks. Place the second beaker under a light source, approximately 6-8 inches below the light. The lights are best held in a clamp on a ring stand. Begin timing the experiment as soon as the light is turned on. Record your observations on page 4.

11. Notice what is happening to the leaf disks as photosynthesis proceeds. Continue to record your observations in the chart on page 4. After each time check, tap the side of the beaker to make sure the

disks are not "sticking" to the container walls. ***Note:*** *Check the covered beaker quickly to avoid light exposure. When instructed, clean the lab equipment and dispose of solutions in the sink drains.*

Photosynthesis in Leaf Disks **_____Name**

Data Table: Number of Leaf Disks Floating

Time (Minutes)	Number of Disks Floating (Light)	Number of Disks Floating (Dark)
0		
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
11		
12		
13		
14		
15		

(After 15 minutes consider the experiment over and that no more disks will rise.)

Lab Analysis and Questions:

1. Use the graphing grid on page 6 and graph the results from the light and dark treatments.

2. What is the variable in the experiment?

What problem/question did you answer in this experiment?

3. Why was detergent added to the solution?

Why was sodium bicarbonate ($NaHCO_3$) added to the solution?

Photosynthesis in Leaf Disks _____Name

4. Explain why it was important to keep one beaker covered during the experiment.

5. Describe and explain the relationship between the number of disks floating and time, as shown on the graph.

6. Did any leaf disks float in the dark treatment? If so, what may explain this result?

7. What process cannot occur in the dark treatment?

8. Explain the changes that occurred within the leaf tissue that allowed the leaf disks to rise to the surface.

9. You will now design and carry out an "Experimental Lab" using a variable that you select. Think about another factor you could test to determine its affect on photosynthesis. Set up a lab notebook page that lists your hypothesis, materials, procedures, data charts and conclusions. Ask your teacher for any extra materials you might need to be sure they are available. Be sure to follow all safety guidelines.

Photosynthesis in Leaf Disks _____Name

Question 1 –Graph

In the area below create a **double line graph** to display the results of the experiment. Provide a title, label the X and Y axis, and label each line appropriately or use a color key.