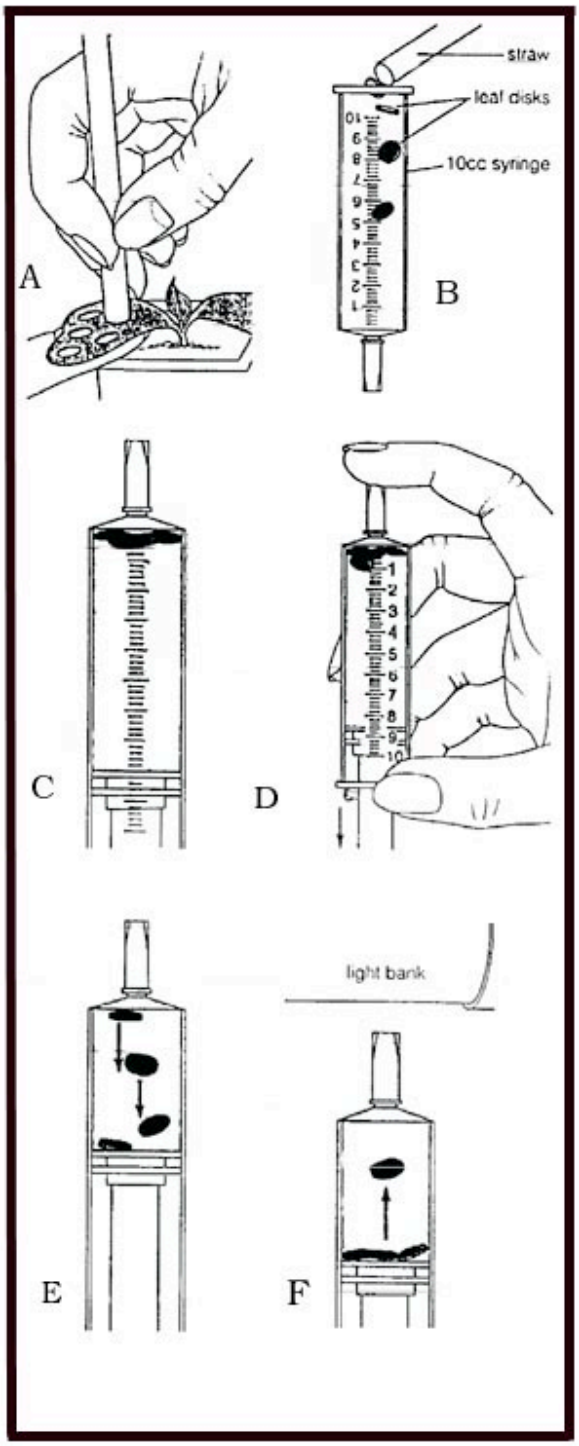
Photosynthesis: Measuring Photosynthetic Rate in Spinach Leaf Disks

Photosynthesis is a process in which primary producers are able to convert light energy (sunlight) into usable chemical energy (carbohydrates). Photosynthesis involves two interlinked processes: the light dependent reaction and the light independent reaction. In the light dependent reaction, light energy is captured and converted to high energy ATP and NADPH molecules. In the light independent reaction these high-energy molecules are used to reduce CO_2 to carbohydrates like glucose.

Overall reaction:

$CO_2 + H_2O$ + light energy → Glucose + O_2



In this experiment, the intercellular spaces of spinach leaf disks are infiltrated with a sodium bicarbonate solution, which causes them to sink in the solution. As photosynthesis occurs, oxygen is produced and collects in the intercellular spaces causing leaf disks to re-float.

Goals

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.
- Understand the relationship between the light dependent and light independent reactions and explain how ATP and NADPH are generated.
- Create hypotheses about the effects of environmental variables on the rate of photosynthesis.

Procedure

1. Using a punch made from a small diameter soda straw, cut 30 leaf disks from young actively growing spinach leaves by supporting the leaf with your index finger while pressing and using a twisting motion of the straw. **(A)**

2. Remove the plunger from 3 clean 10-ml syringes. Using the straw, place the 10 disks into the body of each 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. **(B)**

3. Insert the tip of each syringe into a beaker of 0.2% sodium bicarbonate solution and draw about 8 ml into the syringe. The leaf disks should be floating at this time. **(C)**

4. Hold each syringe tip upward and expel the air by depressing the plunger carefully.

5. Seal the tip of the syringe using the index finger of your left hand. 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. **(D)**

6. Simultaneously, release your index finger and the plunger. Some of the leaf disks should start to sink. Tap the side of the tube to dislodge bubbles on the edges of the disks. **(E)**

7. Repeat steps 5 and 6 until all 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.

8. Empty the syringes into a small beaker in front of the flood lights or other light source selected. Be sure to place the beakers at the three distances from the light- 10 cm, 30 cm, & 50cm.

9. If photosynthesis proceeds, accumulating oxygen will cause the disks to float. At 1 minute intervals, count the number of disks floating in both the light exposed and covered (dark) syringes. Keep recording for 15 minutes. Record this information in Data Table 2.

10. Choose a second variable—besides light intensity to research.

Hypothesis:

Procedure:

Data Table: Number of Spinach Disks Floating

Time (minutes)	Number of Disks Floating (10cm)	Number of Disks Floating (30cm)	Number of Disks Floating (50cm)	Number of Disks Floating	Number of Disks Floating	Number of Disks Floating
0						
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						

Lab Analysis and Questions:

1. Using the graph below graph the results from all treatments.

   a. What is the independent variable?

   b. What is the dependent variable?

2. Describe and explain the relationship between the number of disks floating and time.

Recall that rate = $\Delta y / \Delta x$

Determine the initial photosynthesis rate of spinach leaf disks during the experiment and the rates between each of the time points. Record the rates in the table below. Show your calculations!

	Initial 0 to 5	**5-10**	**10- 15**	**Overall**	**ET50**
Rates (10cm)					
Rates (30cm)					
Rates (50cm)					

3. When is the rate the highest? Explain why.

4. When is the rate the lowest? For what reasons is the rate low?

5. Did the rate change with the light intensity treatment? If so, what may explain this result?

6. Was your hypothesis correct for your selected variable? Explain your results in terms of the process of photosynthesis.

7. **Predict:** What process (the light dependent or the light independent) cannot occur in the dark treatment? Why does this stop the leaf disks from floating?