## AP BIOLOGY LAB: PHOTOSYNTHESIS

(adapted from AP College Board Photosynthesis lab)

## Background and Pre-lab:

Photosynthesis fuels ecosystems and replenishes the Earth's atmosphere with oxygen. Like all enzyme-driven reactions, the rate of photosynthesis can be measured by either the disappearance of substrate, or the accumulation of product.

The equation for photosynthesis is:

$$
6 \mathrm{CO}_{2}+6 \mathrm{H}_{2} \mathrm{O} \rightarrow \mathrm{C}_{6} \mathrm{H}_{12} \mathrm{O}_{6}+6 \mathrm{CO}_{2}\left(+\mathrm{H}_{2} \mathrm{O} \text { technically }\right)
$$

To determine the rate of photosynthesis one could measure the production of oxygen, which is released when photosynthesis occurs or the consumption of carbon dioxide.

## Leaf Structure and Function

In this investigation, you will use a system that measures the accumulation of oxygen in the leaf. Consider the anatomy of the leaf. The leaf is composed of layers of cells. The spongy mesophyll layer is normally infused with gases, oxygen and carbon dioxide. Leaves (or disks cut from leaves) will normally float in water because of these gases. If you draw the gases out from the spaces, then the leaves will sink because they become more dense than
 water. If this leaf disk is placed in a solution with an alternate source of carbon dioxide in the form of bicarbonate ions, then photosynthesis can occur in a sunken leaf disk. As photosynthesis proceeds, oxygen accumulates in the air spaces of the spongy mesophyll. The leaf becomes buoyant and floats.

While this is going on, the leaf is also carrying out cellular respiration. This respiration will consume the oxygen that has accumulated and possibly cause the plant disks to sink. In other words, the buoyancy of the leaf disk is actually an indirect measurement of the NET rate of photosynthesis occurring in the leaf tissue.

## Pre-lab Questions:

1. What is the function of the stoma (or stomata)?
2. What will happen to the leaf disks if you remove the air from the spongy mesophyll?
3. How will air return to these air spaces?
4. Instead of carbon dioxide, what will be used to as the reactant in this lab?

## Materials:

Baking soda
Liquid soap
Plastic syringes ( 10 mL or larger)

Leaves (spinach or ivy)
Timer
Hole punch Light source

## Procedure:

1. Collect leaf disks by punching holes in the dark green portion of a leaf (try to get them between the veins). You will a total of 20 disks.
2. Make a solution of sodium bicarbonate by mixing 300 mL of water to 3 g of baking soda.
3. Make a diluted solution of liquid detergent ( 3 drops of soap to 70 mL of water). This may be already done for you.
4. Add one drop of this dilute soap solution to our bicarbonate solution - do not generate suds.
5. Place 10 leaf disks into the syringe and pull in a small volume of the bicarbonate and soap solution. Replace the plunger and push out most of the air, but do not crush your leaves.
6. Create a vacuum by covering the tip of the syringe with your finger. Draw back the plunger.
7. Release the vacuum so that the solution will enter the disks. It may take a few times to get the disks to sink.
8. Once they have sank, you can put them back into the sodium bicarbonate solution and expose the disks to light.
9. Repeat steps $5-7$ with another set of 10 disks. Place this set into a control beaker that contains only water and expose to light.
10. Record the number of disks floating at the end of each minute, swirl the disks to dislodge any that may get stuck to the beaker or to each other. The experiment should continue until all the disks are floating.
11. Repeat steps 1-10 with "light green" leaf disks.

## Data Table:

| Time <br> (min) | \# Dark disks floating in <br> baking soda | \# Dark disks floating in <br> water (control) | \# Light disks floating in <br> baking soda | \# Light disks floating in <br> water (control) |
| :---: | :---: | :---: | :---: | :---: |
| 1 |  |  |  |  |
| 2 |  |  |  |  |
| 3 |  |  |  |  |
| 4 |  |  |  |  |
| 5 |  |  |  |  |
| 6 |  |  |  |  |
| 7 |  |  |  |  |
| 8 |  |  |  |  |
| 10 |  |  |  |  |
| 11 |  |  |  |  |
| 12 |  |  |  |  |
| 13 |  |  |  |  |
| 14 |  |  |  |  |
| 15 |  |  |  |  |
| 17 |  |  |  |  |
| 18 |  |  |  |  |
| 19 |  |  |  |  |
| 20 |  |  |  |  |

## Analysis:

To make comparisons between experiments, a standard point of reference is needed. Repeated testing of this procedure has shown that the point at which $50 \%$ of the disks are floating $\left(\mathrm{ET}_{50}\right)$ is a reliable and repeatable point of reference.

Graph your data for the experiment. X-axis is Time (min) and Y -axis is \# floating disks. Be sure to include a key for each sample set. There should be four lines on your graph.

Determine the $\mathrm{ET}_{50}$ for each set of leaf disks.

Write a summary of your data. Was there a difference in the rate of photosynthesis between the light and dark green leaf disks? Describe this difference (if any).


## Conclusion:

Answer the following in your conclusion: What would account for your results? Summarize photosynthesis and relate to the differences between the leaf samples. What environmental factors might affect the rate photosynthesis? How and why do you think these environmental factors would affect the rate of photosynthesis?

