Pre-lab Homework Lab 6: Photosynthesis & Cellular Respiration

After reading over the lab and the topics of photosynthesis and cellular respiration from your textbook, answer these questions to be turned in at the beginning of the lab!

1. Recall chemical equations like that from last week’s lab:
   This week’s lab covers the metabolic processes of photosynthesis and cellular respiration. Both are complex sequences of chemical reactions that can be simplified into an overall summary chemical equation. Within the lab manual, find the summary chemical equations for photosynthesis and for cellular respiration and write them in the space below. The Cellular Respiration and Photosynthesis chapters of your textbook describes the complexity of the events.
   - Photosynthesis summary chemical equation (don’t forget light input):
   - Cellular Respiration summary chemical equation (don’t forget energy output):

2. During exercise 1, you will be testing for starch in plant leaves. Relate the following to the summary equation of photosynthesis above:
   a. What product formed from photosynthesis is the building blocks of starch?
   b. Why are we testing for starch instead of the product glucose?

3. A. Look over the experiment in exercise 1: Last week, the lab prep staff put different colored light filters on plant leaves (blue, green and red). You will be comparing the amount of starch produced under each treatment. They also put black paper which blocks all light to parts of the leaf. What would you expect happen here?
   B. Based on the components of experimental design on page 5, what is the purpose of measuring photosynthesis in leaves that are not getting any light?

4. Fill in the blanks based on exercise 2: You, blowing CO₂ from your breath through a straw into water forms ____________ acid. This added acid will cause a pH indicator to change color. You will measure the amount of this dissolved CO₂ by adding drops of the base ____________ until the pH indicator returns to its original color.
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Lab 6: Photosynthesis & Cellular Respiration

LAB SYNOPSIS:
We will cover two topics; photosynthesis and cellular respiration.
- Photosynthesis: we will do an experiment to determine the affect of different wavelengths of light (colors of light) on plants ability to produce starch.
- Cellular respiration: you and your group will design and perform an experiment to test situations affecting the rate of CO\(_2\) that you breathe out.

OBJECTIVES: After successfully completing this lab, a student will be able to:
- Explain the connection between light and energy storage in plants.
- Describe the colors of light that are most affective for photosynthesis and why.
- Explain the connection between photosynthesis and cellular respiration.
- Describe the affects of some common activities on cellular respiration.

Overview:

Photosynthesis:
Summary chemical equation

\[
6\text{CO}_2 + 6\text{H}_2\text{O} \rightarrow \rightarrow 6\text{O}_2 + \text{C}_6\text{H}_{12}\text{O}_6
\]

The above is the summary chemical equation of photosynthesis. Plants use light energy to convert carbon dioxide and water \(\rightarrow\) oxygen gas and the sugar glucose. This is a complex, multistep series of reactions that occur in the plant’s chloroplasts (a plant cell organelle). You should be able to recognize the substrates and products on the above reaction.

Cellular Respiration:
Summary chemical equation

\[
\text{C}_6\text{H}_{12}\text{O}_6 + 6\text{O}_2 \rightarrow \rightarrow 6\text{CO}_2 + 6\text{H}_2\text{O}
\]

ADP + Pi \rightarrow ATP

The above is the summary chemical equation of cellular respiration. This is the reason you need to breath. In the presence of oxygen gas, cellular respiration converts the energy of sugar into ATP. ATP couples the energy released from burning sugar to reactions that require energy (e.g. walking, building macromolecules, growing hair, growth and development, etc.). The products of the reactions of cellular respiration are carbon dioxide and water. Cellular respiration is a multistep series of reactions that occur partly within in the fluid of the cytoplasm, then within the cell’s mitochondria. You should be able to recognize the substrates and products on the above reaction.
Note: when comparing photosynthesis to cellular respiration.

- The chemical equations are just the reverse of each other!
  - However, the process is very much not the reverse. Different enzymes and pathways are used.
- The carbon cycle and oxygen cycle are interrelated!
  - Oxygen from photosynthesis is responsible for all the oxygen in our atmosphere. This is oxygen we need to breathe.
  - Carbon dioxide from cellular respiration will be used by plants in photosynthesis.

**Exercise 1: Photosynthesis and Light Wavelengths**

Light energy occurs in discrete wavelengths. These wavelengths determine the color of light. A pigment is a substance that absorbs light energy. Plants use green pigments called chlorophylls to absorb light energy. The summary chemical equation for photosynthesis is:

\[
\text{Light} \quad 6\text{CO}_2 + 6\text{H}_2\text{O} \rightarrow 6\text{O}_2 + \text{C}_6\text{H}_{12}\text{O}_6
\]

The product of photosynthesis is the sugar, glucose (C₆H₁₂O₆). It is not easy to test for glucose in leaf. Instead we will test for starch. When plants are busy photosynthesizing, they temporarily store sugars as the polysaccharide starch (Glucose $\rightarrow$ Starch).

Recall starch is a polysaccharide made up of 1000’s of glucose monomers.

We will be measuring starch as an indirect measurement of photosynthesis in leaves that have been given different wavelengths of light.

**OBSERVATION:**

Chlorophyll, the major pigment plants use to absorb light energy, is green. Review the experiment below to determine the steps of the scientific method used to explore this observation.

**Experiment:**

Filters of different colors were placed on some of the leaves of a geranium plant (*Geranium sp.*). We used four types of filters (see plant set-up in the room).

- **Black** (blocks all light)
- **Blue** (blocks all wavelengths except for blue)
- **Green** (blocks all wavelengths except for green)
- **Red** (blocks all wavelengths except for red)
- **Unfiltered** (note that parts of the leaf are not covered with a filter)

These plants were then grown for 1 week under growth lights at room temperature. Using the procedure below, we will determine the amount of photosynthesis under each filter by measuring the amount of starch produced.
Experimental variables.

Review the above experiment and determine the experimental variables.

a. What is the independent variable in this experiment? What are the treatments? _____________________

b. What is the dependent variable in this experiment? i.e. what is being measured? __________________

c. Review the experiment and list at least 3 things that are being kept constant? (standardized variables). These could be affecting the amount of photosynthesis.

QUESTION: based on the experiment, what is the question we are trying to answer?

Control groups

Recall: good experiments have one or more control groups. These serves as a comparison for the experimental group(s). Since ideally the only difference between these groups is a change in the independent variable, any difference in the two tests should be due to the affect of the independent variable.

- **Experimental group(s):** This is the trial you are exposing to your independent variable.
- **Positive control(s):** This is a control where you know the outcome should show expected results. From the experiment, which treatment would you expect to form starch?
- **Negative control(s):** This is a control where you know the outcome should not show expected results. From the experiment, which treatment would you expect not to form starch?

HYPOTHESIS and RESULTS table: based on the experiment, hypothesis/predict the amount of starch produced

<table>
<thead>
<tr>
<th>Filter</th>
<th>Prediction (amount of starch you think)</th>
<th>Results after staining (amount of starch seen)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Black No light</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Blue</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Green</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Red</td>
<td></td>
<td></td>
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<tr>
<td>Unfiltered Full light</td>
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</tbody>
</table>

Which of the above is the negative control? Which is the positive control?
READ ALL INSTRUCTIONS BEFORE BEGINNING THIS PROCEDURE. Because this procedure involves heating up a highly flammable liquid, you need to be very careful and follow instructions carefully!

Procedure for Testing for Starch in Leaves:
This is not the experiment. This is just how we will measure for photosynthesis.

1. Record color filter placement on your leaves:
Obtain the leaf or leaves assigned to your group and take them back to your desk. If you are using more than one leaf, make sure that you can tell the leaves apart (cutting notches in them or cutting their stems works best; don't rely on the color as they will all come out the same color!).

<table>
<thead>
<tr>
<th>Table 1: Before and after leaf drawings</th>
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</thead>
<tbody>
<tr>
<td>Before you remove any of the filters, draw your leaf, showing the placement of the filters. Leave space to sketch your “after” leaf as well.</td>
</tr>
<tr>
<td>Leaf with filters</td>
</tr>
<tr>
<td>Make sure leaf is same side up</td>
</tr>
</tbody>
</table>

2. Decolor leaves:
In order to stain for starch, all the color in the leaf must first be removed. Wear goggles as you proceed.

a) You will set-up a double boiler system. To do this add ~200ml tap water to a 1000-ml beaker, add a few boiling chips and heat on a hot plate. Turn the hot plate to high. The hot plate will get HOT—please be careful!

b) Add ~100 ml of 80% ethanol into another, 250-ml beaker. Carefully set this beaker into the boiling water beaker. When the water returns to boiling, turn down the hot plate until the water maintains a slight boil.

c) After sketching your leaf, carefully remove the filters and submerge the leaf in the boiling ethanol.

d) After the leaf is a very pale green or white color, remove the leaf, rinse it with tap water and put it into a Petri dish.

e) Make sure you turn off your hot plate!

f) Spread your leaf out flat within the Petri dish and add a little water so that the leaf is just covered.
3. Staining leaf for starch:

a) Now add 4-5 whole **droppers full** of Lugol's iodine solution to the dish and swirl it to cover the leaf.

b) In about 5-10 minutes, the leaf will begin to turn black where the starch is. This is where photosynthesis was happening!

4. Record results: Table 1 drawing of leaf after staining for starch

a) In table 1 above, record by drawing the areas of your leaf that have stained black/dark brown. The dark color indicates the presence of starch and thus where photosynthesis happened. Illuminate your Petri dish with leaf on the light table will help in determining where/how much starch there is. (putting your Petri dish onto white paper will also make it easier to see).

5. Clean-up

a) After your double boiler has cooled, ethanol goes into the labeled alcohol waste container (in the fume hood)! You can put the water down the drain. DO NOT let boiling chips go down the drain. They can be reused.

**CONCLUSIONS:** Compare whole class results to the expected results from your hypotheses (review your predictions).

| a. Did you get your expected results?  
| Why? Why not? |
| b. Based on class data; What **color** of light was best for photosynthesis? ( Red, Green, Blue ) |
| b. Based on class data; What **color** of light was worst for photosynthesis? (Red, Green, Blue ) |

Recall: the green filter is blocking all wavelengths of light **except** for green. What do your results suggest about your hypothesis on photosynthesis and starch production. i.e.

| c. How does this relate to the **color** of chlorophyll? |

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Spectroscope- device used to analyze the spectrum produced by a light source. Essentially a way to look at how a prism to separates light.

Now that you know what wavelengths (colors) of light plants can and cannot use for photosynthesis, use the spectroscope to determine if they are good, efficient sources of light to grow plants. Especially check out the florescent lights. Note how very inefficient they are.

Note: there are lots of spectroscope phone apps.
Exercise 2: Cellular Respiration

OBSERVATION:
You need to breathe to stay alive!

You breathe in oxygen and breathe out carbon dioxide.

Your cells need oxygen to convert the energy found in the sugar glucose into the cellular energy of ATP. This is the process of cellular respiration. Because these reactions use oxygen, it is also referred to as aerobic respiration. The summary chemical equation for cellular respiration is:

\[ C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O \]

How can we measure the CO2 you produced during cellular respiration? Your cells use the energy in ATP to power many reactions (making macromolecules, cell division, muscle contractions, active transport, etc.).

In the following experiment, we will measure cellular respiration by measuring the product, CO\(_2\). You will exhale through a straw, bubbling air into a beaker of water with a pH color indicator in it (bromothymol blue). As you breathe CO\(_2\) into the water, the solution will become more acidic (CO\(_2\) dissolves in water forming carbonic acid)

\[ H_2O + CO_2 \rightarrow H_2CO_3 \text{ (Carbonic acid)} \rightarrow H^+ + HCO_3^- \text{ (Bicarbonate)} \]

As pH decreases, the pH indicator will change color. You will then add drops of a base, NaOH, to increase the pH, until the solution returns to its original color. The number of drops of the base, NaOH, needed is an approximation of the amount of CO\(_2\) your body produced.

Procedure

Make a stock solution of pH indicator:
Supplies: bromothymol blue, 100ml graduated cylinder, 1 large beaker (600ml), 2 medium beakers (250ml), straws, NaOH

1. Obtain one 600 ml beaker and add ~400 ml of dH\(_2\)O (deionized water).

2. Add ~20 drops of the pH indicator, bromothymol blue, to the water. Add one drop NaOH. Bromothymol blue is yellow at pH below 7 (an acidic solution). It has a blue color at pH of 7 and above (neutral and alkaline pH).

3. Get two clean 250ml beakers and, using a graduate cylinder, measure 100 ml of stock solution into each. One beaker will be used for your experiment, the other will be used as a standard, a control to be used to compare color change. (Figure 3). Retain your stock solution for your designed experiment below.

Measuring resting CO\(_2\):

4. To collect your data, blow into the experiment beaker through a straw for 15 seconds. To ensure accurate measurements, be very careful to blow with equal force for each reading.

5. After blowing into your beaker, add one drop of base (NaOH) at a time and mix gently but thoroughly. Add (and count) the number of drops of the base until the solution returns to its original color (compare to the standard beaker). This indicates that the CO\(_2\) you added has been neutralized (brought back to a pH ~7). The number of drops of base you add is then an indirect measure of the amount of CO\(_2\) that you added to the beaker. Record results in the table below.

To gain familiarity with the experimental set-up and to create a positive control, each member of the group should measure their rate of cellular respiration while sitting down. Record the number of drops of base, NaOH, needed to return the solution to its original color.
Table 1. Resting CO₂

<table>
<thead>
<tr>
<th>Subjects name</th>
<th>Number of drops NaOH to return solution to original color</th>
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Now that you know the amount of cellular respiration happening while you are just sitting, you can use that information as a basis for comparison to some sort of experiment. i.e. your table 1 is the control for the experiment your group is developing below.

**Design your own cellular respiration experiment**

Recall: **Observation:** You need to breathe to stay alive!
**Question:** What kinds of things that you do to live have an affect on cellular respiration?

1. What are some variables you think may affect your rate of cellular respiration and thus the amount of CO₂ produced in people? (Brainstorm as many as you can think of!)

2. Circle one of these variables that you will test. Explain why you think this variable will change the rate of cellular respiration.

3. What do you prediction will happen?
Use your prediction to answer the following.

**Designing Your Experiment:** *(based on your prediction/hypothesis)*

*(if you are having one group member act as secretary to record information, inform your instructor and acknowledge who is recording info here __________________*)

1. What is your independent variable?

2. Your dependent variable is the number of drops of base you need to add to neutralize the CO₂ dissolved into the beaker as you blew.

3. What are some other possible independent variables that could affect CO₂ production? (i.e. standardized variables. You will need to try to control or standardize these).
3. In the space below, outline the experiment you will perform. Include how you will attempt to control for any other possible independent variables. Explain how you will alter the one independent variable you wish to investigate.

4. Predict the results that will support your hypothesis.

5. Predict the results that will NOT support your hypothesis.

Before performing the experiment, get your *instructor's initials. ______________

RESULTS: Conduct your experiment and analyze your results (you will probably be comparing your data with that in table 1). Develop a table and/or graph that best displays your results

CONCLUSIONS:
What conclusions can you make?

Error analysis/troubleshoot: How could you improve your experiment and clarify your results? Are there any doubts or alternative explanations of the results you got?