

# Lab 7 - An Introduction to Photosynthesis

(April 2014)



## Section 1 - Energy

[2] Hi, this is Lyn Koller again. This week we will be exploring one of the biochemical pathways organisms use to obtain and use energy. We will start with how plants use energy from sunlight.

[3] If we are going to see how plants use energy, we better learn what energy is first. Let's start by defining energy. Write the definition you see here in your lab manual in Section 1. When you are finished, we will look at some different types of energy.

[4] The first type of energy we will look at is potential or stored energy. One way energy can be stored is by virtue of position. Here you see water behind a dam. Its stored energy can be released as the water flows through electric generating turbines.

[5] Here is another example of energy stored by position. This rock, poised at the top of a hill has potential energy. You probably don't want to be standing below it when its energy is released!

[6] Another way to store energy is in structure. I hope you remember that electrons in atoms contain varying amounts of energy depending on which energy level or shell they occupy. Electrons in higher energy levels contain more energy and can readily give up or accept energy as the electrons are transferred to other atoms.

[7] You probably also remember from an earlier lab that molecules are made up of atoms which are held together by energy relationships called chemical bonds. These bonds also store energy.

[8] Food molecules store energy in electrons and chemical bonds. Be sure you have defined potential energy and listed examples in your lab book before you continue.

[9] The second type of energy is kinetic energy or the energy of motion. Anything that is moving has kinetic energy. This moving car has kinetic energy.

[10] Remember the dam and electric turbines? The electricity that moves through power lines has kinetic energy that we use – here for instance to light up our world.

[11] Light also contains kinetic energy. Light is made up of tiny moving particles called photons that travel in waves. The kinetic energy in sunlight is used by plants during photosynthesis. At this time complete Section 1 and then return to the program.

## Section 2 - Energy Molecules

[12] Now that you understand the difference between potential and kinetic energy you should be able to match these examples to the correct type of energy.

[13] In this lab we will see how plants convert light energy into chemical energy. First we need to look at how two molecules involved in the process can store energy. Here you see a molecule of ATP. ATP stands for adenosine triphosphate.

[14] As you can see in this simplified version, here and in your lab manual in Section 2, ATP is a molecule made up of one adenosine molecule and three phosphate groups.

[15] Study the reaction you see here. ATP can be broken down into ADP, adenosine diphosphate, and a phosphate group. The bonds between the phosphate groups contain energy so that when the bonds are broken, energy is released.

[16] If organisms had to make new ATP from scratch each time they needed the molecule, there wouldn't be enough resources in the world! Therefore the ADP and phosphate are bonded together again and the ATP and ADP and phosphate can be used over and over again. See if you can complete the reverse reaction in Section 2. Here is the breakdown reaction to use as a reference. When you have finished return to the program.

[17] Did you figure out the reverse reaction? Here it is so you can correct your equation if you need to.

[18] Another important molecule in photosynthesis is  $\text{NADP}^+$ . This molecule can also store energy, but this time the energy is stored in electrons.  $\text{NADP}^+$  is capable of picking up high energy electrons in one location and then giving them up in another location. As you can see the negative charge of the electron is canceled out when the molecule also picks up a positively charged hydrogen ion to become NADPH. Finish up Section 2 and then return.

[19] Now see if you can match each molecule to the correct type of energy storage.

### **Section 3 - The Laws of Thermodynamics**

[20] The Laws of Thermodynamics sound like a complicated topic, but you probably already know them in common terms. As you can see in your lab manual in Section 3, the first law states that energy can neither be created nor destroyed, but energy can be transformed from one kind of energy to another and transferred from one place to another. Let's focus on the process of photosynthesis. Plants use light energy to power the reaction and then they store the energy in the molecules they produce.

[21] The second law of thermodynamics tells us that energy transformations or transfers are not 100% efficient. That is, some of the energy is dispersed as heat. Heat is a type of kinetic energy that is not the most useful so we sometimes say that during the transfers and transformations, energy is lost as heat. Complete Section 3 applying these two laws to photosynthesis remembering that plants use sunlight to make molecules.

### **Section 4 - Photosynthesis and the Levels of Organization**

[22] Now that you know a little about energy let's see how the plant uses sunlight. We will start with plant structure. Section 4 shows you the different levels of organization involved in photosynthesis. Fill in the chart as we go through the next few program pages.

[23] The plant organ involved in photosynthesis is the leaf. We will look at this organ in more detail later in the lab.

[24] Inside the leaf there are several different tissue types, but the main one involved in photosynthesis is the parenchyma tissue of the palisade layer.

[25] In the palisade layer, individual parenchyma cells are responsible for the majority of photosynthesis although any green cell can carry on photosynthesis.

[26] Within the parenchyma cells the organelle responsible for the photosynthetic reaction is the chloroplast.

[27] At the level of the molecule, there are many involved in the entire process, but for now we will just say that the plant uses carbon dioxide to make glucose. Be sure you have the chart filled in and then we'll continue.

[28] See if you can put these plant structures in the correct order?

### **Section 5 - The Photosynthetic Reaction**

[29] Here you can see the overall reaction for photosynthesis. Do you remember that reactions that build more complex products are anabolic and those that produce less complex products are catabolic? Write this equation down in the beginning of Section 5 and answer the question that follows before returning to the program.

[30] For the next part of Section 5 you'll see a long set of instructions for the first of today's experiments. This experiment will take a ***minimum*** of 45 minutes, so don't start it unless you can stay in lab until it's finished. Before you start the experiment let me give you some hints for completing the experiment successfully.

[31] When you are filling your tubes with water, be sure you fill them  $\frac{3}{4}$  of the way full.

[32] Be sure you exhale directly into the tube. You want to supply the plants with carbon dioxide for photosynthesis. After exhaling, cap and shake the tube.

[33] Keep exhaling into the tube and shaking until the color of your solution is close to the example tube's color. Repeat this with all three tubes.

[34] Once your three solutions are the same correct color place an entire sprig of *Elodea* in tubes A and B, but not in C.

[35] Once you have your tubes set up and signed off, be sure to record the time and your block number and then set them right in front of the lights. Now if you have enough time, set up the experiment and then return to the program.

## **Section 6 - The Leaf and Photosynthesis**

[36] We can continue with Section 6 while your tubes are sitting in front of the lights. At the end of the lab, before the summary we will come back and review the experiment.

[37] In Section 6 we are going to take a closer look at a leaf. Find the slide of the leaf cross section in the slide box in your booth. Place the slide on the microscope stage and once you have found the section with scanning power, switch to low and then to high power. Look for a portion of the leaf cross section that looks similar to this photograph. In your lab manual, fill in the missing cells of the leaf cross section. You will be able to label the structures listed as we see what each structure does in the following program pages.

[38] The carbon dioxide plants need for photosynthesis enters the leaf through each stomate which is an opening between two guard cells. Guard cells can open and close the stomate to regulate gas exchange.

[39] The water used in photosynthesis is transported up from the soil through specialized vascular tissue called xylem. Phloem, another vascular tissue, transports the products of photosynthesis. The xylem and phloem together form the vascular bundle.

[40] The carbon dioxide and water travel to the palisade cells through the openings in the spongy mesophyll. Remember the cells of the palisade layer are responsible for the majority of photosynthesis.

[41] The upper and lower epidermis form protective layers and are coated by a cuticle. The cuticle is an acellular waxy-like coating that protects the leaf against excess water loss.

[42] See if you can match the leaf structures with their names. When you have them all correct continue with the program.

[43] It's not always easy to see how the guard cells are arranged from the cross section, so the next thing I'll have you do is make your own slide of the epidermis of a leaf. This photo demonstrates how you need to tear the leaf in order to obtain the whitish looking epidermis. You will only need a small amount of epidermis. Also be sure it doesn't fold or roll-up on you as you make the slide. When you have your slide ready, return to the program.

[44] When you view your slide, you should see something like this. Once you have finished drawing and labeling your leaf preparation, we'll be ready to continue. By the way, the term "stomata" is the plural of stoma or stomate – all leaves have many stomata.

## **Section 7 - Chloroplast Structure**

[45] Here is the overall equation for photosynthesis again. We are going to break down the overall equation into two major sub-reactions. The first, the light reaction, is sometimes referred to as the light dependent reaction. As you can see during the light reaction water is broken down and oxygen gas is released.

[46] The dark reaction is also known as the light independent reaction. During this portion of the overall reaction carbon dioxide is converted into glucose.

[47] Using the information given here, answer the questions in Section 7. When you get to the chloroplast, return to the program.

[48] You can see the structure of a chloroplast here and in your lab manual. I'll go over each of the structures and its function in the next few program pages, so you can label the structures as we go along.

[49] Since the chloroplast is an organelle that is imbedded in the cytoplasm it transports carbon dioxide and water into the interior and oxygen and glucose out across the double membrane surrounding the stroma. It is in the stroma that the dark or light independent reaction takes place and carbon dioxide is converted into glucose

[50] Within the stroma you see stacks of disks called thylakoid disks. Each stack of disks is called a granum and collectively all of the stacks are the grana. The thylakoid disks contain the photosynthetic pigments and are the site of the light reactions. Light reactions require water which is split into hydrogen ions and the oxygen the plant gives off.

### **Section 8 - Photosynthetic Pigments**

[51] So what are photosynthetic pigments? These pigments are molecules that are able to capture light energy. The photosynthetic pigments are different types of chlorophyll which give leaves their green color. These pigments capture light energy and use it to power the light reactions.

[52] You may have noticed that not all plants are green. They may be different colors. This is especially noticeable in the fall when the leaves turn various shades of reds and yellows. What produces these other colors?

[53] Because chlorophyll is green it reflects green wavelengths of light. Plants have accessory pigments that are other colors and can absorb green wavelengths of light. The absorbed energy is then passed on to use during photosynthesis.

[54] Your next experiment is going to demonstrate the presence of accessory pigments in a plant extract. In Section 8 you will identify two different chlorophyll pigments and two accessory pigments. This is done by separating them in a process called paper chromatography. The more soluble a pigment is the further up it will travel on the chromatography paper. Here you see the items you will need.

[55] Follow the directions in Section 8 carefully and you should get a nice chromatography paper to attach to your lab page. Be sure you place 20 drops of the extract in exactly the same place; air drying the paper between drops.

[56] When you place your paper into the chromatography tube, be sure only the tip of the paper is placed into the solution – you don't want to wash off the extract.

[57] Once your chromatography paper is in the solution, it should take about 15 minutes to see adequate pigment separation. You can continue with the lab, but just keep an eye on the process so the solution doesn't reach the top of the paper.

[58] When you can see two separate green and two yellow bands, remove the paper and air dry it. Outline the bands in pencil and staple or tape it into your lab book in the space provided. Label the pigments as you see here.

[59] As you have read, the separation of the pigments is based on their solubility in the chromatography solution. Can you match the solubility to the pigment?

[60] At this point you should be able to complete the questions at the end of Section 8. Return to the program when you have done so.

[61] Are you beginning to have an appreciation of the overall process of photosynthesis? A plant needs sunlight and the photosynthetic pigments in the chloroplasts to convert carbon dioxide and water into oxygen and glucose. We are now going to focus on the sub-reactions that make this possible.

[62] Let's start with the light reactions which are also referred to as the light dependent reactions. See if you remember where these take place.

[63] Hopefully you got that on the first try so let's find out what happens in the thylakoid disks. As the light shines on the plant some of this light energy enters the leaf and the palisade cells to strike the chloroplast. Once inside the chloroplast the light energy strikes the thylakoid disk where special molecules take the light energy and transform it into chemical energy. By the way, which law of thermodynamics supports this energy transformation?

[64] Yes, it's the First Law of Thermodynamics. This energy transformation will store the chemical energy in the two energy molecules we discussed at the beginning of the lab - ATP and NADPH. The lab will only cover these reactions in general terms and you may get more detail in lecture.

[65] ATP can be produced in two different ways. The one you see here is cyclic. The sunlight energy is converted to chemical energy when an electron from chlorophyll absorbs light energy. This high energy electron is passed through a series of molecules in an electron transport chain. As the electron is passed along, energy is released and used to produce ATP from ADP and P. Once all the gained energy is released, the electron returns to its original location.

[66] The next way ATP is generated is linked with another reaction that produces NADPH. Here is the whole process, but we'll take it one step at a time.

[67] Once again you can see in the first part of the process, an electron absorbs light energy. The electron is passed through another electron transport chain and the energy given off by the electron is used to produce ATP just like before. The difference here is that the electron is not returned to its original molecule.

[68] OK, let's look at the second half of this process and see how NADPH is produced. Here is the whole process again. Take a moment to see how the formation of ATP fits in, and then return to the program so we can continue.

[69] Here is the production of NADPH. The process starts out the same way – sunlight energy is transformed into chemical energy when electrons absorb the light energy. These high energy electrons are passed through another electron transport chain, but instead of returning to the original chlorophyll, they are picked up by  $\text{NADP}^+$ .

[70] One more thing to look at in this process. The negative charge of the electrons is cancelled out when  $\text{NADP}^+$  picks up one hydrogen ion.

[71] So where does the hydrogen ion come from? Look again at the light reaction. You will see that below the thylakoid disks water is being broken down and electrons, hydrogen ions, and oxygen gas are being produced. This process actually occurs inside the thylakoid disks, but again you don't need that much detail for now.

## Section 9 - The Stages of Photosynthesis

[72] The light reactions can be summarized in the two chemical equations you see here. Copy these into your lab manual in the first part of Section 9 and then return to the program.

[73] In order to help you answer the next questions in your lab manual, let's review the light reactions. The energy starts out as light energy and then gets transformed into chemical energy. This energy is stored in either electrons or chemical bonds. OK, now answer the questions in the first part of Section 9 and then return to the program.

[74] Did you remember that ATP stores energy in chemical bonds whereas NADPH stores energy in high energy electrons? If not, perhaps you need to briefly review Section 2. When you are ready, just continue.

[75] The dark reaction (or light independent reaction) is powered by the energy stored during the light reaction. In the stroma of a chloroplast, a complex set of chemical reactions called the Calvin cycle, takes carbon dioxide and converts it into glucose.

[76] Considering the overall equation for photosynthesis, how many carbon dioxide molecules does it take to make one molecule of glucose?

[77] Here is the equation for the dark reaction. Copy this into your lab manual in the second part of Section 9 and then come right back.

[78] Where is the energy now? The ATP molecule provides energy to bond the atoms of glucose together and the high energy electrons from NADPH are combined into the glucose molecule.

[79] At this time you should be able to answer the last questions in Section 9. Return to the program when you have finished.

[80] Before we summarize the whole process of photosynthesis, I want to make sure you have completed and understood the experiment in Section 5. By this time, you should have had your tubes signed off at the end of their exposure to light. You may have also filled in the table and answered the questions that followed, so let's see how you did.

[81] You added bromothymol blue to each tube to monitor pH. In a previous lab you learned that pH measures the hydrogen ion concentration – an excess of hydrogen ions produces a pH from 0-7 and an excess of hydroxide ions produces a pH from 7-14 with 7 being neutral with equal numbers of hydrogen and hydroxide ions.

[82] When you exhaled into the tubes, what molecule were you adding?

[83] What did the added carbon dioxide do to the pH of the solution? Remember when you added the bromothymol blue, the solution was blue and as you added the carbon dioxide the solution turned green.

[84] If carbon dioxide is removed from the solution, what color would you predict the solution should be? When you are correct continue.

[85] What was the pH of each tube at the end of the experiment? See if you can match tube and pH correctly. Be sure the answers in your lab manual reflect these correct answers.

[86] OK, if you got all of these correct it should be obvious that tube A had the least carbon dioxide at the end of the exposure to light.

[87] You actually ran two experiments at the same time with the elodea. The first experiment was between tubes A and B. What was different about the experimental conditions between tubes A and B? In other words, what variable were you testing?

[88] Therefore what is the first thing necessary for the removal of carbon dioxide from the solution? Be sure the answer in your manual is correct and then continue with the program.

[89] The second experiment was between tubes A and C. What was different about the experimental conditions between tubes A and C? Or, what variable were you testing here?

[90] Therefore what is the second thing necessary for the removal of carbon dioxide from the solution? Be sure the answer in your manual is correct.

[91] Try this one last question.

[92] Do you think you have proved that the entire that photosynthetic reaction took place? What tests did you make for the presence of glucose? If you haven't already filled in the correct answers at the end of Section 5, do so now and then return to the program.

[93] Since energy is so important, see if you can drag the energy sources or molecules into the correct order.

### **Section 10 - A Summary of Photosynthesis**

[94] Now you can summarize the process of photosynthesis in Section 10. Before you fill-in the diagram, write the general equation for photosynthesis one last time to use as a reference. If you have written the correct equation, you will notice that you have three general reactants and two general products. Complete your diagram, have it signed off, clean up your booth and have a great week.