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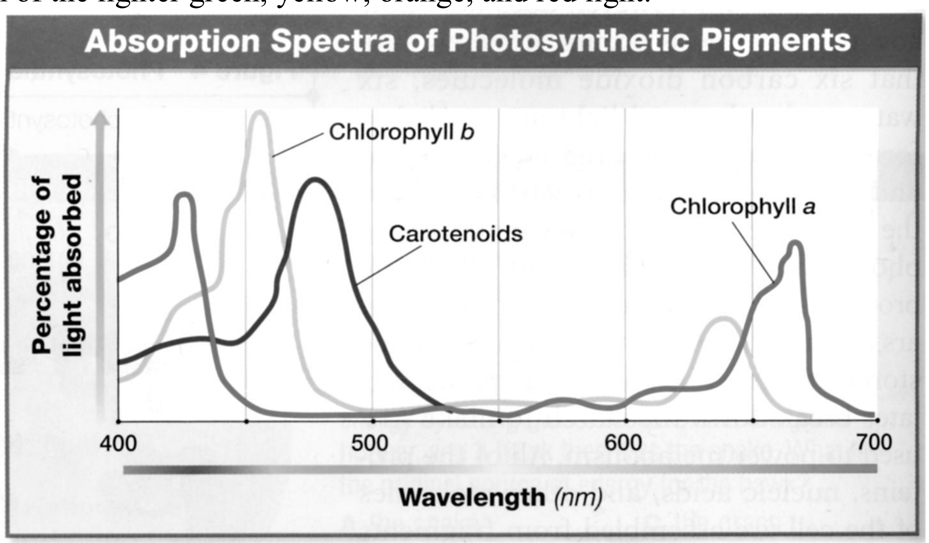
Why are some plants green and others not?

Adapted from: Why are plants green?

White light from the sun or from a light bulb contains all the colors of the rainbow: red, orange, yellow, green, blue, indigo, and violet. To see all the colors of the rainbow you need to split up white light. Use a prism to split white light into the separate colors.

Organisms absorb light using light-absorbing molecules called **pigments**. Not all pigments can absorb every color of light. **Chlorophylls**, the primary pigments in photosynthesis, absorb violet, blue and red light very readily but not much of the lighter blue, green and yellow light. The lighter blue, green and yellow light that is not well absorbed by chlorophylls is reflected back to your eye and that is why chlorophyll looks green to us.

Besides chlorophyll plants have other pigments, called accessory pigments, which collect the energy from different colors of light and transfer that energy to chlorophyll. Carotenoids are one group of these accessory pigments. Carotenoids absorb light blue and green light very readily but not much of the lighter green, yellow, orange, and red light.



In the figure above **absorption spectra** for three types of photosynthetic pigments are graphed. An absorption spectrum is a graph of what wavelengths of light are absorbed by a pigment.

Part 1: Measuring photosynthesis surface area

Objective: Estimate surface area of leaf

You know that plants absorb energy in the form of light from the sun and use it to run the chemical reactions of photosynthesis and make sugar. Photosynthesis can be summarized by the following equation.

$$6\ \mathbf{CO_2} + 6\ \mathbf{H_2O} \xrightarrow{\text{light}} \mathbf{C_6H_{12}O_6} + 6\ \mathbf{O_2}$$

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carbon dioxide	water	sugar	oxygen gas

Obtain two leaves, a spinach and a coleus. Measure the length and width (1/2bh) to estimate surface area of the leaf, this will be the estimated area of photosynthetic activity.

Leaf type	Length (cm)	Width (cm)	Surface area (cm^2)

Part 2: Viewing photopigments

Objective: To observe the different photopigments plants use for photosynthesis.

You will use a technique called chromatography to separate the pigments in spinach and Coleus leaves so you can see the colors of the different types of pigments plants use for photosynthesis.

Chromatography is a technique used to separate mixtures of compounds. Paper chromatography a spot of mixture is put onto a chromatography paper. The end of the paper is put into a solvent. As the solvent creeps up the paper and past the spotted mixture of pigments, some of the pigments dissolve in the solvent more quickly than others. After a period of time the different pigments in the mixture end up spread out between the original spot and the point the solvent reaches.

Hypothesis: What colors do you think will be separated in the spinach and Coleus leaves?

Safety First: You must wear your goggles and gloves throughout the experiment. Work under the fume hood!

1. Obtain a 250 mL beaker, a piece of chromatography paper, 5-10 mL acetone, 2 spinach leaves and one Coleus.
2. First you will transfer the photopigments from spinach leaves onto your chromatography paper. Take your piece of chromatography paper, fold it in half and place a spinach leaf across the bottom of the one half of the paper. Roll a quarter over the spinach 1.0 cm from the bottom creating a straight green line of pigment on the filter paper. **Allow your paper to dry**. Repeat this 5-7 times using a fresh portion of leaf and allowing the paper to dry a little in between rolls so you don't rip your paper. It is **VERY** important to make all of your spinach lines on top of one another so the green line gets very dark.

3. Repeat with Coleus leaf on the other half of the paper.

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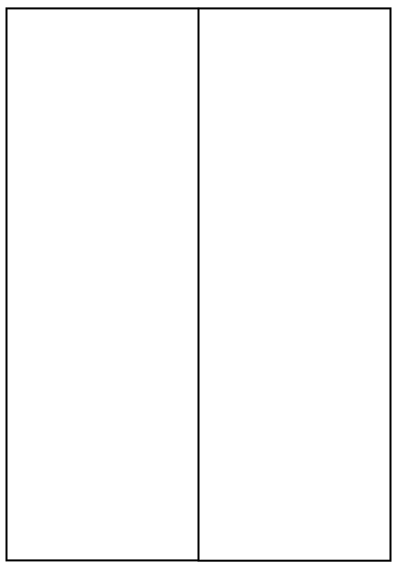
4. Add 5-10 mL of acetone to the 250 ml beaker.

5. Now you will use acetone to separate the different photopigments. Place the chromatography paper into the container and cover the tube loosely with a paper towel. Make sure the green line of spinach is not in the acetone. Place under the fume hood.

6. Watch the acetone and pigments run up the filter paper. Remove the paper chromatogram from the test tube after about 10 minutes.

7. Examine and draw the chromatogram for the presence of different bands of color. Each color band is a different pigment. What color(s) are carotenoids (the bands of color at the top and bottom of the paper)?



8. Why do you think plants have photopigments that absorb different colors of light?

9. If plants have both chlorophylls and carotenoids why do you think leaves are usually green?

10. Explain how your observations relate to the change in leaf color that takes place in the fall.

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Part 3. Determine % Absorbance of Spinach and Coleus

Objective: Generate an absorption spectrum for spinach and coleus leaves by using a spectrophotometer. From these results determine the best pigments for photosynthesis.

In this section we will generate the absorption spectrum of a spinach chloroplast suspension and compare it to the absorption spectrum of a coleus suspension. Before generating the spectra make a hypothesis below outlining your prediction of the results. The color of the spinach and the coleus should help.

Hypothesis:

1. Obtain 5ml of spinach suspension and add 5 mL of distilled water, mix and pour into a cuvette (3 in test tube).

2. Obtain 5 mL of Coleus suspension and add 5 mL of distilled water, mix and pour into a cuvette.

3. Fill a third cuvette with 10 mL of distilled water.

4. Set spectrophotometer to A (absorbance) and change the wavelength to 350 nm.

5. Zero the machine by placing your blank in the chamber, (remember to face the white side of the test tube towards the marker), close the cover and hit the 0A/%T button. Why is zeroing a type of control?

6. Once you have zeroed the spectrometer, determine the absorption spectrum as follows:

   a. Place the cuvette with spinach into the spectrophotometer and record absorbance. Repeat for the Coleus.

   b. Measure the absorbance of the suspensions at 25nm intervals. (You **MUST zero** the spectrophotometer with the blank at each wavelength BEFORE measuring the suspension.)

7. What “blank” did you use to calibrate the spectrophotometer?

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8. Record absorbance data of your extract at 25 nm intervals from 350 - 700 nm:

Wavelength (nm)	Spinach	Coleus
350		
375		
400		
425		
450		
475		
500		
525		
550		
575		
600		
625		
650		
675		
700		

12. Graph an absorption spectrum plot of your spectrometry data on the graph paper provided. (wavelength on the x axis and absorbance on the y axis).

13. Do different leaves (and pigments) have different absorption spectra? Explain…

14. Using your graph, discuss which light wavelengths are best absorbed by plant pigments. What does your absorption spectrum tell you? Can you relate this to photosynthesis?

15. Why do tropical plants have large colorful leaves vs. a cactus which the leaves are reduced to spines?

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Why are plants green?

Pre lab questions must be completed before the start of lab!!!

1. What structure in a plant cell captures sunlight?

2. What is the name of the chemical substance that is found in the above structure?

3. What is the chemical equation for photosynthesis?

4. What type of energy is responsible for the yield of sugar and oxygen in this equation?

5. How does this relate to the Law of Conservation of Energy?

6. Draw a chloroplast and include the following: stroma, grana, thylakoid