

Beer-Lambert Law:  
Measuring Percent Transmittance of  
Solutions at Different Concentrations  
*(Teacher's Guide)*

## OVERVIEW

Students will study the relationship between transmittance, absorbance, and concentration of one type of solution using the Beer-Lambert law. They will determine the concentration of an “unknown” sample using mathematical tools for graphical analysis.

## MATERIALS NEEDED

Ward’s DataHub  
USB connector cable\*  
Cuvette for the colorimeter  
Distilled water  
6 - 250 mL beakers  
Instant coffee  
Paper towel  
Wash bottle  
Stir Rod  
Balance

\* – *The USB connector cable is not needed if you are using a Bluetooth enabled device.*

## NUMBER OF USES

This demonstration can be performed repeatedly.

## FRAMEWORK FOR K-12 SCIENCE EDUCATION © 2012

\* The Dimension 1 practices listed below are called out as **bold** words throughout the activity.

Dimension 1 Science and Engineering Practices	✓	Asking questions (for science) and defining problems (for engineering)	✓	Use mathematics and computational thinking
	✓	Developing and using models	✓	Constructing explanations (for science) and designing solutions (for engineering)
	✓	Planning and carrying out investigations	✓	Engaging in argument from evidence
	✓	Analyzing and interpreting data	✓	Obtaining, evaluating, and communicating information

Dimension 2 Cross Cutting Concepts	✓	Patterns	✓	Energy and matter: Flows, cycles, and conservation
	✓	Cause and effect: Mechanism and explanation		Structure and function
	✓	Scale, proportion, and quantity		Stability and change
	✓	Systems and system models		

Dimension 3 Core Concepts	<b>Discipline</b>	<b>Core Idea Focus</b>
	Physical Science	PS1: Matter and Its Interactions
		PS1.A: Structure and Properties of Matter

NGSS Standards	<b>Middle School Standards Covered</b>	<b>High School Standards Covered</b>
	MS.PS-SPM: Structure and Properties of Matter	HS.PS-SPM: Structure and Properties of Matter

## NATIONAL SCIENCE EDUCATION STANDARDS © 2002

Content Standards (K-12)			
✓	Systems, order, and organization		Evolution and equilibrium
✓	Evidence, models, and explanation	✓	Form and Function
✓	Constancy, change, and measurement		

Physical Science Standards Middle School		Physical Science Standards High School	
✓	Properties and Changes of Properties in Matter		Structure of Atoms
	Motions and Forces	✓	Structure and Properties of Matter
	Transfer of Energy		Chemical Reactions
			Motions and Forces
			Conservation of Energy and Increase in Disorder
			Interactions of Energy and Matter

✓ Indicates Standards Covered in Activity

# LEARNING OBJECTIVES

## **Core Objectives (National Standards):**

- Develop the ability to refine ill-defined questions and direct to phenomena that can be described, explained, or predicted through scientific means.
- Develop the ability to observe, measure accurately, identify and control variables.
- Decide what evidence can be used to support or refute a hypothesis.
- Gather, store, retrieve, and analyze data.
- Become confident at communicating methods, instructions, observations, and results with others.

## **Activity Objectives:**

The purpose of this activity is to relate light absorbance and transmittance in solutions with different concentrations, create a hypothesis and proceed to test it using the Ward's DataHub colorimeter sensor.

## **Time Requirement:**

60 - 90 minutes

## VOCABULARY

**Absorbance:** The amount of light absorbed by a sample.

**Colorimeter:** An instrument that measures the amount of light that passes through a sample.

**Concentration:** The relative amount of a given substance contained within a solution or in a particular volume of space; the amount of solute per unit volume of solution.

**Incident Light:** The light that falls directly on an object.

**Molar Absorptivity:** The molar absorption coefficient is a measure of how strongly a chemical species absorbs light at a given wavelength.

**Monochromator:** An optical device that transmits a mechanically selectable narrow band of wavelengths of light or other radiation chosen from a wider range of wavelengths available at the input.

**Monochromatic Light:** Light with only one wavelength present.

**Path Length:** The overall length of the path followed by a light ray.

**Solute:** The minor component of a solution. The substance dissolved in the solvent.

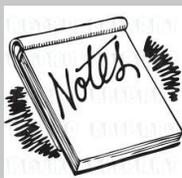
**Solvent:** A liquid capable of dissolving other substances.

**Transmittance:** The passage of light through a sample.

**% Transmittance:** The manner in which a colorimeter reports the amount of light that passes through a sample.

**UV Light:** The wavelength of light that is used to detect colorless molecules.

**Wavelength:** The distance between two successive crests or troughs in a light wave.



### Teacher Notes

## INTRODUCTION

As part of our everyday life we prepare and use different solutions. Each one is composed of two components: the solvent (most of the time this is water), i.e., the substance into which the solute is dissolved; and the solute, which is the substance that is dissolved in the solvent. A common example of a solution may be salt water for cooking pasta, which becomes more salty the more concentrated the solution is.

When you are doing scientific work, particularly in chemistry, biology, or medical sciences, it is very important to know exactly the concentration of the solutions you are working with. To measure concentration, several techniques can be used.

- **Can you think of some different solutions you have prepared at school or at home?**
- **When you are making tea, how do you know when it is ready? Describe.**

*Carry out the experiment with your class so that at the end, students will be able to answer the following question:*

- **How can a light beam that passes through a sample of a given solution help determine its concentration?**



### DID YOU KNOW?

The concept behind the Beer-Lambert law was actually discovered before 1729 by Pierre Bouguer, however it is often attributed to Johann Heinrich Lambert who cited Bouguer's *Essai d'Optique sur la Gradation de la Lumiere*. It was not until much later that August Beer extended the exponential absorption law to include concentrations of solutions in the absorption coefficient. He did this in 1852.



Johann Heinrich Lambert

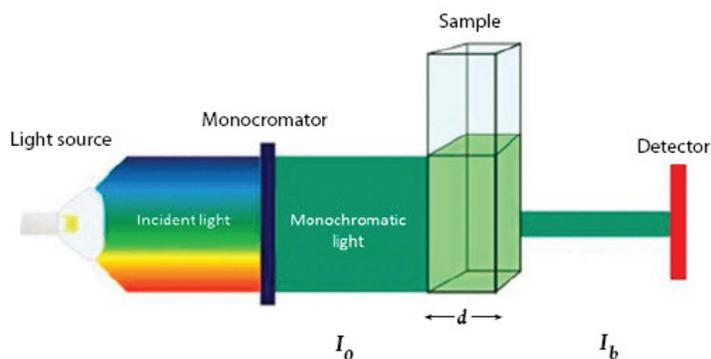
## BACKGROUND

A solution is a homogenous mixture of two or more substances that exist in a single phase and has, depending on the relationship between solute and solvent, different concentrations. To determine the concentration of a given solution, you can use a few different methods. Here we are going to use colorimetry to measure the transmittance and determine the solution's concentration, thus applying the Beer-Lambert law. This law states that the concentration of a solute is proportional to the absorbance.

The colorimeter allows light to pass through a cuvette containing a sample of the solution which absorbs some of the incoming beam. When the ray of light of a given wavelength and intensity ( $I_0$ ) comes into contact perpendicularly with the solution of a tinted chemical compound, the compound will absorb part of the light radiation ( $I_a$ ). The remaining light ( $I_b$ ) will pass through the solution and strike the detector. As such, the following equation is demonstrated.

$$I_0 = I_a + I_b$$

The absorbance of light is related to the number of molecules present in the solution (concentration of the solution).



The Beer-Lambert law defines the relationship between the concentration of a solution and the amount of light absorbed by the solution:

$$A = \epsilon dC$$

Where:

A = Absorbance

$\epsilon$  = Molar absorptivity ( $L \text{ mol}^{-1} \text{ cm}^{-1}$ )

d = Path length of the cuvette containing the sample (cm)

C = Concentration of the compound in the solution ( $\text{mol L}^{-1}$ )



### DID YOU KNOW?

The concepts of colorimetry are used daily in microbiology laboratories. Bacterial growth studies are performed using a technique called spectrophotometry. Much like colorimetry, spectrophotometry is used to determine the concentration of particles in a solution. Researchers compare the absorbance of an unknown sample of bacteria in media to a known curve that has been constructed. This allows them to determine the number of living bacteria in a sample.

Transmittance is the relationship between the amount of light that is transmitted to the detector once it has passed through the sample ( $I$ ) and the original amount of light ( $I_0$ ). This is expressed in the following formula.

$$T = I / I_0$$

Where  $I_0$  is the intensity of the incident light beam and  $I$  is the intensity of the light coming out of the sample. Transmittance is the relative percent of light that passes through the sample. Thus, if half the light is transmitted, we can say that the solution has 50% transmittance.

$$T \% = (I / I_0) \times 100 \%$$

The relationship between transmittance ( $T$ ) and absorbance ( $A$ ) can be expressed by the following:

$$A = \log_{10} \left( \frac{1}{T} \right)$$

$$A = \log_{10} \left( \frac{100}{T[\%]} \right) = 2 - \log T[\%]$$

*At this point, encourage students to formulate a hypothesis to test as part of this activity. Students may find it helpful to formulate their hypothesis as an answer to the following question:*

- **If you have one type of solution of different concentrations, how do you expect the transmittance percentages to change when you measure from the lowest to the highest concentration? Why?**

## CONNECTING THE WARD'S DATAHUB TO A COMPUTER

### If you are using a Bluetooth communication device:

Right-click on the Bluetooth icon in the lower right corner of the screen and select the Ward's DataHub you are using. The icon will change from gray to blue, as shown at right, indicating that the Ward's DataHub and the computer are now connected via Bluetooth.



### If you are using a USB communication device:

In order to use USB communication, connect the Ward's DataHub and the computer with the USB cable supplied. Click on the USB icon at the lower right corner of the screen. This icon will change from gray to blue, as shown at right, indicating that the Ward's DataHub is connected to the computer via USB.



## USING THE WARD'S DATAHUB



= Select key



= On/Off and Escape key



= Scroll key

To collect measurements with the Ward's DataHub, it must first be configured as follows:

1. Turn on the Ward's DataHub by pressing the On/Off/Esc key.		8. Press the On/Off/Esc key to return to the setup menu.	
2. Go to setup by using the Scroll key then select Setup by pressing the Select key.	 then 	9. Press the Scroll key to highlight the Number of Samples and then press the Select Key.	 then 
3. Select the Set Sensors option by pressing the Select key.		10. Press the Scroll key until "Manual" is highlighted, then press the Select key.	 then 
4. If any sensor(s) appear on the screen, press the key representing that sensor to deactivate it. Once you have a blank screen, press the <b>Colorimeter Sensor</b> key once.		11. Press the On/Off/Esc key Three times to return to the main operating screen.	 x 3
5. Press the On/Off/Esc key once to return to the setup menu.		12. <b>Press the Select key to start measuring.</b> (You are collecting data when there is an icon of a Runner in the upper left hand corner of the screen.)	
6. Press the Scroll key to highlight the Sampling Rate and then press the Select Key	 then 	13. Once you have finished measuring, stop the Ward's DataHub by pressing the Select key, followed by the Scroll key.	 then 
7. Press the Scroll key until "Manual" is highlighted, then press the Select key.	 then 		



### DID YOU KNOW?

The Beer-Lambert Law is not applicable when the concentration of a solution reaches a certain value. This is due to electrostatic interactions between the molecules in close proximity. The value will plateau at a point and thus, scientists often have to dilute concentrated samples in order to analyze them.

## PRE-ACTIVITY PREPARATION

### Preparation of the “unknown” samples:

The teacher will prepare two samples of solution for the students. The students will determine the concentrations of the samples through the experiment.

The instructions to prepare the “unknown” samples are as follows:

	Sample A	Sample B
Solute (instant coffee) (g)	1	1
Solvent (distilled water) (mL)	140	90
Concentration (g/mL)	0.0071	0.011

The teacher will give 3 mL of one of the samples to each work group. The students will be told that they have to obtain the concentration. At the end of the class, they will compare their results to the theoretical value.

Students will read a graph to obtain the concentration. To understand it, they have to know the slope-intercept form for the equation of a line.

$$y = mx + b$$

Where:

m = slope

x = x coordinate

y = y coordinate

b = y intercept (where the line crosses the y-axis)

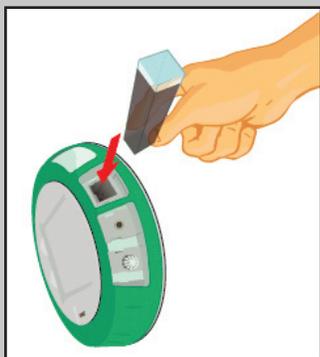
Students must:

1. Obtain the percent transmittance by using the colorimeter.
2. Divide the percent transmittance by 100 to obtain transmittance (T) of the sample.
3. Calculate absorbance (A) using the following formula:  $A = \text{LOG}_{10} T$ .
4. Obtain x by solving the slope-intercept equation, substituting the absorbance value for the y coordinate.



### DID YOU KNOW?

Transparent materials are those materials that allow one or more of the frequencies of visible light to be transmitted through them. The colors of light that are not transmitted are usually absorbed by the material. The frequencies of light that are transmitted are then picked up by our eyes and that is the color we perceive the material to be.



## ACTIVITY

**Note: Before you start recording, calibrate the colorimeter with the solvent you will use during the experiment. This step is important to minimize the experimental error of the measurement. To calibrate, add some solvent (in this case, distilled water), filling 3/4 of the sensor's cuvette and place the cuvette in the port of the colorimeter. Then, press the button on the colorimeter until you hear the Ward's DataHub signal. Once you have finished the calibration, remove the cuvette from the colorimeter.**

1. Mark each beaker with the number of the sample (1 to 6).
2. Place distilled water in beaker 1.
3. Prepare samples 2 to 6 as shown in the table below by measuring the amount of distilled water (solvent) and adding 1 gram of coffee (solute) to each beaker.

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
Coffee (g)	0	1	1	1	1	1
Distilled Water (mL)	100	150	120	100	80	60
Concentration (g/mL)	.00	0.0066	0.0083	0.0100	0.0125	0.0166

4. Use a stir rod to mix the coffee until it dissolves completely.
5. Measure the transmittance of the solution. To take the data reading, fill the cuvette until it is 3/4 full with solution from sample 1. ALWAYS hold the upper part of the cuvette to ensure the sides remain clean from fingerprints.
6. Clean and dry the outside of the cuvette with paper towels.
7. Place the cuvette in the port of the colorimeter. (See diagram at left.)  
  
Cover the cuvette, moving the rotating ring of the Ward's DataHub to cover the port and then start measuring.
8. Record the percent transmittance. Take the sample out of the port and clean the cuvette with distilled water. Repeat the steps with the other samples, including the "unknown" solution given to you by your instructor.
9. Once you have finished measuring, stop the Ward's DataHub.

## RESULTS AND ANALYSIS

1. Export the data to Microsoft Excel by clicking on  . Save the data to your computer.
2. Observe the percent transmittance values obtained by the Ward's DataHub when the samples were exposed to different light wavelengths (red, green and blue).
3. Organize the data starting with the percent transmittance given by the Ward's DataHub. Data should be organized in the following order and as seen in the chart below.

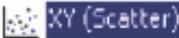
### Sample - Concentration (g/mL)

**% Transmittance red - Transmittance red - Absorbance red**

**% Transmittance green - Transmittance green - Absorbance green**

**% Transmittance blue - Transmittance blue - Absorbance blue**

Sample	Conc. [g/mL]	%Transm. Red	Transm. Red	Abs. Red	%Transm. Green	Transm. Green	Abs. Green	%Transm. Blue	Transm. Blue	Abs. Blue
1	0		=C2/100	=LOG10(D2)		=F2/100	=-LOG10(G2)		=I2/100	=-LOG10(J2)
2	0.0066									
3	0.0083									
4	0.01									
5	0.0125									
6	0.0166									
Unknown 1A or 1B										

4. Use the formulas shown in chart above, to calculate the transmittance and absorbance of each sample.
  5. Observe and compare the transmittance values for each sample. Remember that the Beer-Lambert law can be used for absorbance values between 0 and 1. Therefore, select the color where you obtained most results within this absorbance range.
  6. Create a line graph of absorbance as a function of concentration. To do this, first create a scatter plot by clicking on the  button.
  7. Select the Data Range. Select the concentration values for the x-axis and absorbance values for the y-axis.
  8. Add a regression line to the graph by right-clicking on the points and selecting "Add Trendline". Select linear regression type. Choose the options chart and select "Display equations on chart". Press OK to finish.
- **How do the results relate to your initial hypothesis? Explain.**
  - **What was the relationship between the transmittance and concentration of the solutions?**
  - **What was the relationship between the absorbance and concentration of the solutions?**

*(continued on next page)*



### DID YOU KNOW?

Water selectively scatters and absorbs certain wavelengths of visible light. Red, Yellow and Orange light have longer wavelengths and thus can penetrate up to 50 meters (164 feet) into water.

Shorter wavelengths such as the violet, blue and green light waves, can penetrate further into the water. Blue penetrates the farthest and this is why deep, clear ocean water appears blue.

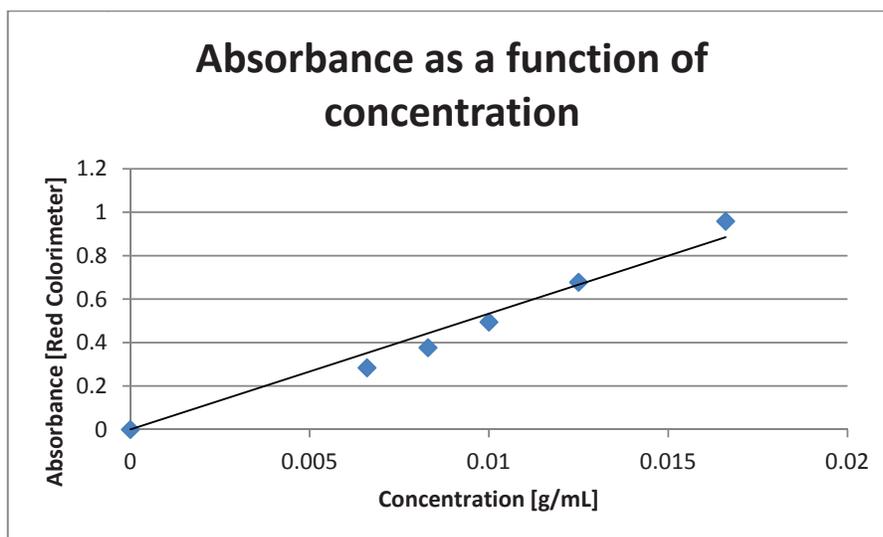
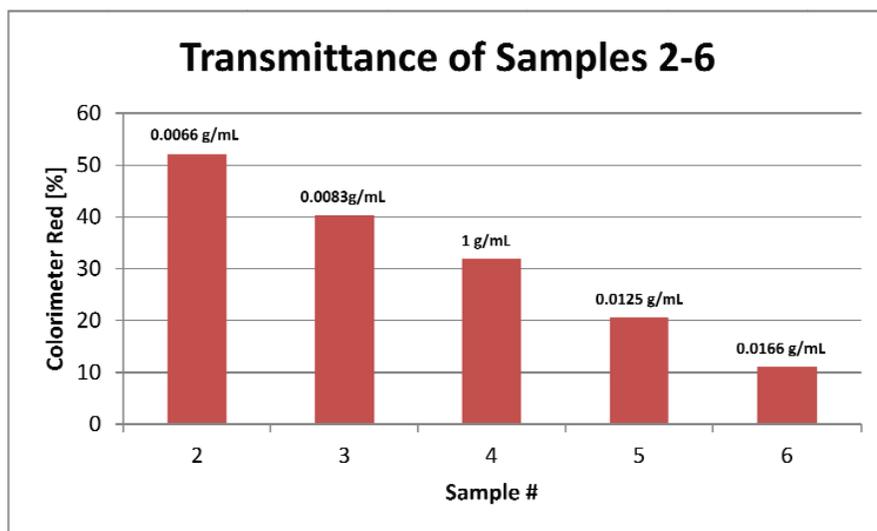


## RESULTS AND ANALYSIS

*continued*

*The graphs below should be similar to the one the students come up with for the red colorimeter. The decreasing order of the percentages can also be observed in both the green and blue colorimeters.*

*This graph is percent transmittance as a function of concentration.*



## CONCLUSIONS AND ASSESSMENTS

1. What variables are related to the Beer-Lambert law?

*Students should point out that the Beer-Lambert law relates absorbance of a solution to its concentration.*

2. If you have a solution and samples of it with different concentrations; in what range of absorbance can we use the Beer-Lambert law? **Explain.**

*Students should point out that the Beer-Lambert law is only valid for absorbance values between 0 and 1.*

3. Suppose the equation on the Excel graph of absorbance as a function of concentration is a straight line. What is the relationship between the expression and the variables related to the Beer-Lambert Law?

*Students should understand the meaning of the equation of the graph. In this case, the y coordinates represent absorbance and the x coordinates represent concentration. Therefore, we can understand the equation as the following expression:*

$$\text{Absorbance} = (m) (\text{concentration}) + b$$

4. If you have a sample of unknown concentration, how would the Beer-Lambert law be useful? **Argue** your answer with evidence from the experiment.

*Students should point out that you can obtain concentration of the unknown sample by measuring absorbance and using the Beer-Lambert's law to compare its absorbance to a known solution absorbance reference.*

5. Use the graph of absorbance vs. concentration to identify the concentration of the "unknown" solution.

*Students should obtain 0.0071 g/mL or 0.011 g/mL approximately, depending on the "unknown" sample they worked with.*

## ACTIVITIES FOR FURTHER APPLICATION

*The aim of this section is for students to extrapolate the knowledge acquired during this class and apply it to different contexts and situations. Furthermore, it is intended that students question and present possible explanations for the experimentally observed phenomena.*

1. Suppose you have an instant coffee solution of an unknown concentration which you want to calculate using the Beer-Lambert Law. However, your absorbance is higher than one. How can you calculate concentration?

*Students should point out that they will have to decrease the absorbance value to use the Beer-Lambert law, diluting the initial sample to obtain an absorbance value between 0 and 1. Starting from that point, they could calculate the concentration of the diluted solution and then the concentration of the original sample.*

Beer-Lambert Law:  
Measuring Percent Transmittance of  
Solutions at Different Concentrations  
(*Student Guide*)

**INTRODUCTION**

As part of our everyday life we prepare and use different solutions. Each one is composed of two components: the solvent (most of the time this is water), i.e., the substance into which the solute is dissolved; and the solute, which is the substance that is dissolved in the solvent. A common example of a solution may be salt water for cooking pasta, which becomes more salty the more concentrated the solution is.

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- **Can you think of some different solutions you have prepared at school or at home?**
- **When you are making tea, how do you know when it is ready? Describe.**

*After carrying out this experiment, you should be able to answer the following question:*

- **How can a light beam that passes through a sample of a given solution help determine its concentration?**

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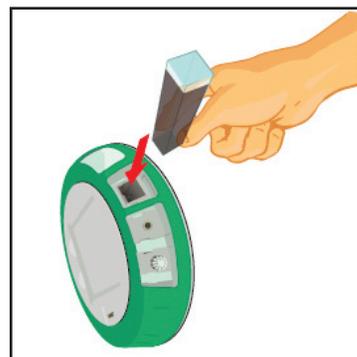
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6. Clean and dry the outside of the cuvette with paper towels.
7. Place the cuvette in the port of the colorimeter.  
(See diagram at right.)

Cover the cuvette, moving the rotating ring of the Ward's DataHub to cover the port and then start measuring.

8. Record the percent transmittance. Take the sample out of the port and clean the cuvette with distilled water. Repeat the steps with the other samples, including the "unknown" solution given to you by your instructor.
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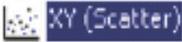
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**% Transmittance red - Transmittance red - Absorbance red**

**% Transmittance green - Transmittance green - Absorbance green**

**% Transmittance blue - Transmittance blue - Absorbance blue**

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1	0		=C2/100	=-LOG10(D2)		=F2/100	=-LOG10(G2)		=I2/100	=-LOG10(J2)
2	0.0066									
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5. Observe and compare the transmittance values for each sample. Remember that the Beer-Lambert law can be used for absorbance values between 0 and 1. Therefore, select the color where you obtained most results within this absorbance range.
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## RESULTS AND ANALYSIS

*continued*

- **How do the results relate to your initial hypothesis? Explain.**

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- **What was the relationship between the transmittance and concentration of the solutions?**

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- **What was the relationship between the absorbance and concentration of the solutions?**

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## CONCLUSIONS AND ASSESSMENTS

1. What variables are related to the Beer-Lambert law?

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2. If you have a solution and samples of it with different concentrations; in what range of absorbance can we use the Beer-Lambert law? **Explain.**

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3. Suppose the equation on the Excel graph of absorbance as a function of concentration is a straight line. What is the relationship between the expression and the variables related to the Beer-Lambert Law?

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4. If you have a sample of unknown concentration, how would the Beer-Lambert law be useful? **Argue** your answer with evidence from the experiment.

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5.. Use the graph of absorbance vs. concentration to identify the concentration of the “unknown” solution.

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