What is in the Kool Aid?
Liquid Spectroscopy Lab
Student Copy
Mr. Barry, Chemistry

Objectives:

- Determine "λ max" (wavelength of maximum absorbance)
- Create a calibration curve using standard molar concentrations of Yellow #5 Dye
- Determine an unknown concentration of Yellow #5 Dye in Tang using a calibration curve

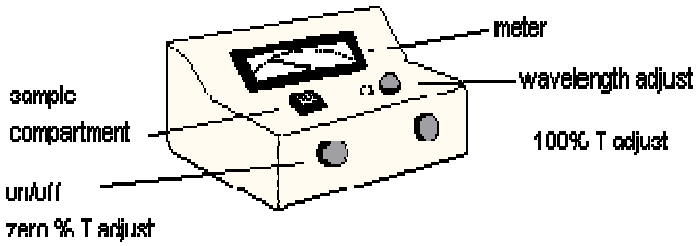
Introduction:

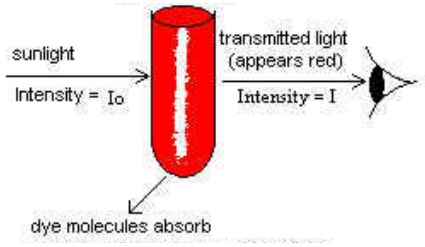
Tang has many different flavors and many different colors. Once mixed with sugar, it can provide a delicious treat. However, there is more going on with the color of Tang than meets the eye. Color can been seen with the human eye, but its intensity can also be detected. We have already learned that different colors emit different wavelengths. If one changes the wavelength, you change the color. Tang is emitting certain wavelengths and transmitting others, depending on the flavor and color. A spectrophotometer is an instrument that can be used to detect what wavelengths of light are being absorbed and what wavelengths are being emitted in a liquid solution.

In this lab students will determine what are the maximum wavelengths are being absorbed by a particular Tang solution. You will also create a calibration curve using standard molar concentrations of Yellow #5. Finally, using the calibration curve, <u>you will determine the unknown molar concentration of the Yellow #5 in Tang</u>.

Theory:

We already know that when electrons receive energy, like in the form of light, they get excited and move up on energy levels. Once they relax, they fall back down to their ground state and emit energy. If light passes through a sample of Tang, the electrons will absorb some of this light energy. Therefore some of the light is not fully transmitted through the sample. A spectrophotometer is a device that records the light that is transmitted (passes through) after being absorbed by a solution. (see pictures below)





Pictures From: http://wwwchem.csustan.edu/CHEM1102H/koolaid.htm

The amount of light that is absorbed by the Tang is expressed in terms of the absorbance, A, where

$$A = \log I_o/I$$

$\mathbf{I_o}$ is the amount of light entering the sample and **I** is the amount of light that actually traveled through the sample. (**I** would be less than $\mathbf{I_o}$ because some of the light was most likely absorbed by Tang)

The absorbance of the sample can change for the various reasons:

- ***Wavelengths***-Depending upon the color of the Tang solution, the part of the visible spectrum that will absorb light will vary. We will determine what the maximum absorbance ("λ max") before starting our calibration curve.
- ***Solution thickness***-The longer the sample, the more the solution can absorb. To eliminate this variable, we will be using the sample size cuvettes (smaller test tubes) in the spectrophotometer.
- ***Concentration***-The absorbance will increase with increased concentration. In order to determine the molar concentration of the unknown sample, you will first determine the absorbance of three known sample concentrations.

Materials:

- "Spectrophotometer 20"
- 2 small cuvettes
- 3 Tang solutions of known molar concentrations
- 1 Tang solution of unknown molar concentration
- Distilled water for the "blank"
- Goggles
- Distilled water wash bottle
- Kim wipes

Part I Procedure:

Determining the Maximum Absorbance at a Particular Wavelength. ("λ max")

1. Turn the "Spec 20" on for 15 minutes in advance.
2. Fill one cuvette ¾ full with distilled water to use as your blank. A blank is necessary to calibrate the Spec 20 at **each** wavelength.
3. Fill another cuvette ¾ full with any of the standard known molar concentration solutions provided by your teacher.
4. Record the stock solution that you have chosen and the color on your chart. Predict the color (and wavelength) that the solution will absorb and transmit. Record this data on your data chart.
5. Make sure the sample compartment is closed and empty. Adjust the % T (Transmission) using the "power switch / zero control" knob till it reads "0".
6. Set the "wavelength control" to 400 nanometers (nm).
7. Wipe off your cuvette and insert your blank (with distilled water) into the sample compartment and close the compartment. (Wiping off the cuvette **each time** is essential to remove dust and fingerprints.)
8. Adjust the %T until it reads 100% T. (all of the light should be passing through the sample and none should be absorbed by the distilled water)
9. Remove the blank and insert the second cuvette of stock solution. Close the sample compartment.
10. Read and record the %T in the data table.
11. Repeat steps 6 through 10 and record the %T and wavelength at 20 nm intervals up to 800 nm. (The second cuvette should be reused for each stock solution. Rinse the cuvette out with distilled water, dry, and reuse for the next stock solution)

Data for Part I:

Food Color analyzed: _____

Molar Concentration of Stock Solution Tested _____

Guess of what color it will absorb: _____

Guess of what color it will transmit: _____

Wavelength (nm)	***% Transmittance***	***Absorbance***
400		
420		
440		
460		
480		
500		
520		
540		
560		
580		
600		
620		
640		
660		
680		
700		
720		
740		
760		
780		
800		

"λ max" from graph _____

Data Analysis:

1. Using the formula below, convert each % Transmittance value into Absorbance:

$$\text{Absorbance} = 2 - \log(\%T)$$

2. Construct a plot of wavelength (on the x-axis) and Absorbance (on the y-axis) using Microsoft Excel. Include an appropriate Title and submit your graph with the lab.
3. Determine the "λ max" from your graph and record it on the Data Table in Part I.

Part I Questions

1. What color was your sample? What color light does it transmit? Absorb?
2. Over what wavelength range does the solution exhibit the greatest absorbance? What color(s) of light are absorbed?
3. What is the maximum wavelength (λ) that the solution absorbed? This wavelength will be used to construct a calibration curve and test the unknown solution to determine the molar concentration in Part II of the lab.
4. What wavelengths are not absorbed? What colors are transmitted?

Part II Procedure:

Determining the Unknown Concentration Using a Calibration Curve:

You will now measure the absorbance of each of the stock solutions.

1. Turn the "Spec 20" on for 15 minutes in advance
2. Fill one cuvette ¾ full with distilled water to use as your blank. A blank is necessary to calibrate the Spec 20 at the maximum wavelength. You will only need to do this once.
3. Fill another cuvette ¾ full with the first standard known molar concentration solutions provided by your teacher.
4. Make sure the sample compartment is closed and empty. Adjust the % T (Transmission) using the "power switch / zero control" knob until it reads "0".
5. Set the "wavelength control" to the maximum absorbance wavelength determined in Part I of the lab.
6. Wipe off your cuvette blank (this needs to be done each time) and insert your blank (with distilled water) into the sample compartment and close the compartment.
7. Adjust the %T until it reads 100% T. (all of the light should be passing through the sample and none should be absorbed by the distilled water)
8. Remove the blank and insert the cuvette (wipe off the cuvette) of the first stock solution. Close the sample compartment.
9. Read and record the %T in the data table.
10. Repeat steps 6 through 9 and record the %T for each of the three stock solutions. **Do not change the wavelength at any time.** Use the same cuvette for each stock solution, but be sure to carefully rinse out the cuvette and dry the cuvette between each use. Your results will be affected if you do not adhere to these directions.
11. Repeat steps 6 through 9 and record the %T for the unknown solution.

Data for Part II:

Maximum Absorbance from Part I:_____(nanometers)

Standard Sample	***Concentration of Yellow #5 (Molarity)***	***% Transmittance at λ Max***	***Absorbance at λ Max***
Standard 1			
Standard 2			
Standard 3			
Standard 4			

Standard Sample	***Concentration (Molarity)***	***% Transmittance at λ Max***	***Absorbance at λ Max***
Unknown	**(from graph)**		

Data Analysis:

1. Using the formula below, convert each % Transmittance value into Absorbance:

$$\text{Absorbance} = 2\text{-}\log(\%T)$$

2. Construct a plot of Concentration (on the x-axis) and Absorbance (on the y-axis) using Microsoft Excel. Include an appropriate Title. Right click on any data point and select "add trendline". Add your linear equation to the graph.

3. Now that you have your equation, substitute the unknown solution's absorbance to determine the unknown molar concentration. Record your work in your lab. Record the unknown's molar concentration.

Part II Questions

1. Why was it necessary to construct a calibration curve?
2. What was the unknown concentration of your solution? How did you determine this? (Be sure to show work)
3. How can this lab be used in everyday practical application?

Conclusion:

Be sure to restate the purpose of the lab and the general procedure for the lab. Give the molar concentration of the unknown solution and give a brief explanation of how you found it. **Use data (numbers) from the lab to support** your unknown molar concentration. Include any difficulties you experienced with the lab.

(See attached Rubric for Lab for complete expectations)