Name: _____

Lab Instructor:_____

PREPARATION FOR CHEMISTRY LAB: SPECTROSCOPY

1. Why do you think some solutions appear colorless?

2. Using the following data points, graph absorbance versus concentration (absorbance on the y-axis and concentration on the x-axis) using the piece of graph paper included with the lab.

Concentration (%, by volume)	10	30	50	70
Absorbance	0.131	0.394	0.657	0.920

   a) By visually examining your graph, determine the concentration of a solution that has an absorbance of 0.49 A. Mark this point on your graph.

   b) Calculate the slope, m, for the line on your graph.

   c) Assuming that your y-intercept is zero, use the slope to find the concentration of the solution assigned above. {$A = m \cdot c$}

3. A sample made up of solvent only (and reactants other than the compound of interest, if any) is called a **blank.** If the blank has a measurable absorbance, then the curve has a non-zero y-intercept. The equation for the absorbance of a solution as a function of concentration then becomes: $A = (m \cdot c) + b$ Calculate the concentration of a solution with an absorbance of 1.275 (A). The slope is 0.0184 (A)/% and the y-intercept is 0.037 (A). Show your work.

4. Using Figure 8-2 in your textbook, determine the color of the light associated with each of the four wavelengths of light used by the LabQuest colorimeter. Enter these below as well as in the table where it also occurs in the lab.

	430 nm	470 nm	565 nm	635 nm
Color				

SPECTROSCOPY

INTRODUCTION

Spectroscopy is the study of the interaction of electromagnetic radiation with matter. All substances interact with electromagnetic radiation in a unique way. Our eyes act as fairly sensitive detectors of electromagnetic radiation that falls in the visible region of the electromagnetic spectrum. Electromagnetic radiation that contains all the wavelengths of visible light is known as white light. Objects appear to be a particular color because they selectively absorb some of the components of white light and reflect and/or transmit the others. The light that is not absorbed registers in the human eye as color. In spectroscopy, the detector is not your eye but rather an instrument that recognizes and measures electromagnetic radiation that is absorbed, transmitted, or emitted by a sample. Such an instrument is called a spectrophotometer.

The spectrophotometer that you will be using is called a colorimeter. A colorimeter sends a beam of light of a specific wavelength through a colored solution in a cuvette. The chemical compound responsible for the color absorbs light of certain wavelengths and transmits other wavelengths. The detector "sees" only the percentage of light that is not absorbed when it passes through the sample. The detector changes this energy into a measurable electrical current which shows up on a scale that is calibrated in absorbance (A) units. The more concentrated the solution, the more light is absorbed.

For substances that produce colored solutions, there is a linear relationship between the concentration, c, of the substance in solution and the amount of light that is absorbed by the solution, A. Mathematically, A is related to c by the equation: $A = (m \cdot c) + b$; where k is a constant and b is the absorbance of the blank (a sample made up of solvent and reactants other than the compound of interest). A is unitless and c can have any units appropriate to concentration: %, moles of solute per L of solution, ppm, etc.

Spectroscopy is not a new topic. The technique was used in Measurement: Part II when you generated a calibration curve to check your skill in using dilution to prepare solutions of specified concentrations. In this experiment, you will again use the LabQuest Colorimeter. You will use food coloring to make standard solutions and prepare a calibration curve. You will then use the calibration curve to determine the concentration of an unknown solution. The use of a calibration curve to determine concentrations of unknown solutions is a common quantitative technique.

PROCEDURE

1. The LabQuest Colorimeter needs to be powered for about 5 minutes before using so do this step before preparing your solutions. Plug the LabQuest Colorimeter into one of the LabQuest sensor ports and turn on the instrument.

2. Arrange seven clean, dry, large test tubes (TT) in your TT rack. Number them 1 through 6. Label the seventh tube - BLANK.

3. Set up and load two 25 mL burets; one with deionized (DI) water and the other with your assigned stock dye solution (concentration: 100% dye).

4. Deliver the volumes (mL) of dye stock solution and DI water shown in the table below. Mix the contents of each tube well.

TT	mL stock dye solution	mL DI H_2O	% v/v
6	10.0	0	100
5	8.0	2.0	
4	6.0	4.0	
3	4.0	6.0	
2	2.0	8.0	
1	1.0	9.0	
BLANK	0	10.0	0

5. Determine the wavelength of maximum absorbance.

   a) Be sure one of the four green wavelength lights is on.

   b) Only touch the ridged faces of the cuvette, never touch the clear faces. Rinse and fill one cuvette (a cuvette should be filled so that it is about ¾ full) with the blank. In this lab the blank is deionized water. Rinse and fill (about ¾ full) a second cuvette with the most dilute of the prepared solutions. **Gently blot** (don't scratch the sides of the cuvette) off any drips on the outside of the cuvette with a Kimwipe.

   c) Place the cuvette containing the blank in the colorimeter. Place cuvettes in the colorimeter so that one of the clear faces is toward the arrow at the top of the cuvette slot. Close the lid.

   d) Using the front panel arrow keys on the colorimeter choose a wavelength. Press the **CAL** button on the front of the colorimeter. Release it when the red LED begins to flash. When the red light stops flashing, the colorimeter is calibrated and ready to use.

   e) Place the cuvette containing the solution to be tested (not the blank) in the colorimeter. Wait for the reading to stabilize and record the absorbance of the solution at that wavelength in your lab report.

   f) Repeat steps c, d, and e until the absorbance is recorded for each of the four available wavelengths.

g) The wavelength that gave the maximum absorbance is the one that will be used for the remainder of the experiment.

6. Calibrate the colorimeter. Place the cuvette containing the blank in the colorimeter set at the wavelength of maximum absorbance. Press the **CAL** button on the front of the colorimeter. Release it when the red LED begins to flash. When the red light stops flashing, the colorimeter is calibrated and ready to use.

7. Using the stylus (never touch the LabQuest screen with your fingers): touch Sensors, Data Collection, choose Events with Entry in the Mode box. Highlight the word Event in the Name: box and replace it with Concentration. Put percent in the Units box. OK.

8. Place the cuvette containing the prepared solution in the colorimeter. Touch the begin data collection button (triangle) on the bottom left of the screen. When the absorbance reading stabilizes, touch the **KEEP** button located just to the right of the data collection button. Don't touch the stop data collection button (square). Type in the concentration of the solution that is in the cuvette (don't include units). OK.

9. Pour your samples back into the original TT after you have measured each absorbance and discard them in the waste container only after you have acceptable data.

10. Working in order of most dilute to most concentrated of the remaining solutions, rinse and then fill a cuvette with the solution that will be analyzed. Place the cuvette in the colorimeter. When the absorbance reading stabilizes, touch the KEEP button, and enter the concentration of the analyzed solution. Repeat until the absorbance of all solutions has been determined.

11. When the absorbance of all prepared solutions has been determined, touch the stop data collection button (square) located on the bottom left of the screen and **save** your data.

12. Your calibration curve should be linear.

    Use the LabQuest to determine the equation of the line. To receive a passing grade for this lab your calibration curve must be a good, straight-line graph, with a correlation coefficient of 99% or better (Corr: on the screen reads 0.9900 or greater). Make fresh solutions if you do not get this level of proficiency. Work carefully, you must have this proficiency and be done with the experiment, write up, and post-lab questions by the end of the lab period.

13. Rinse and fill the cuvette with your assigned unknown dye solution. Place the cuvette in the colorimeter. Touch the Sensor icon on the upper left of the screen. Record the absorbance in your lab report once the absorbance reading has stabilized.

14. Print out a copy of the calibration curve for each lab partner. Each lab report must have a LabQuest generated printout of the calibration curve attached to it. The printout must show the graph, axis labels, the information needed to generate the equation (slope and intercept) for the line, and the correlation reading. This should all show up automatically on the printout. Be sure that the graph title and the names of all lab partners are entered in the footnote and that the date box is checked.

15. Delete all saved files before returning your LabQuest to Lab Services.

DATA AND ANALYSIS SHEET: SPECTROSCOPY

Name: _____

Date _____ Lab Partner _____

Enter the color associated with each of the following wavelengths into the table below and the absorbance from Step 5 at each wavelength.

	430 nm	470 nm	565 nm	635 nm
Color				
Absorbance				

Wavelength of maximum absorbance: _____

Equation of line for the calibration curve: _____

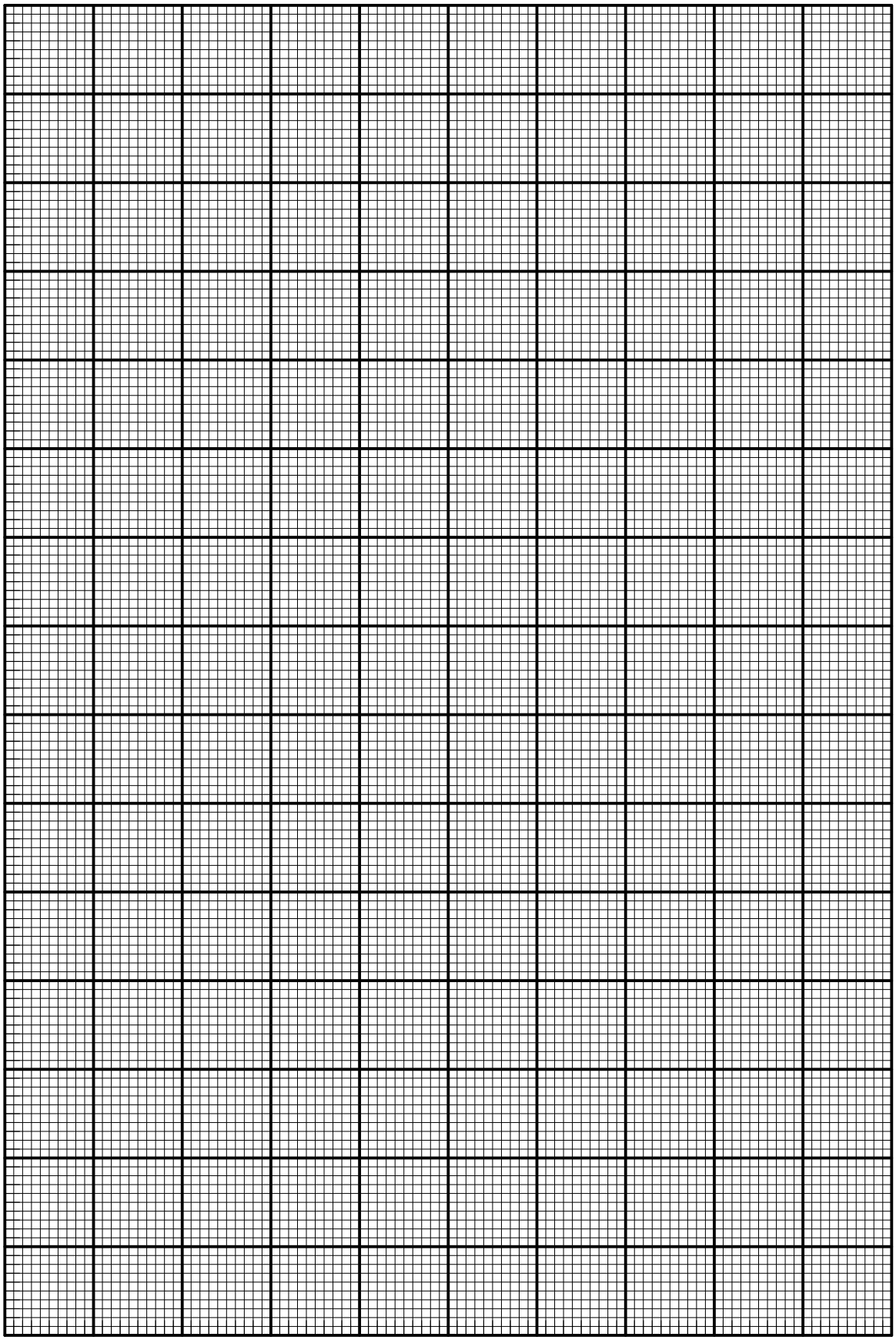
Correlation: _____

Unknown Assignment: _____

Absorbance of your assigned unknown dye solution: _____

Determine the concentration of your unknown using the calibration curve (not the equation). Circle the point on the graph.

Determine the concentration of your unknown using the equation for the line for **your** calibration curve (show work):



Name: _____

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QUESTIONS ABOUT THIS LAB: SPECTROSCOPY

1. Examine your calibration curve. What is the absorbance of a solution in which the concentration of food dye is 0%.. How do you account for this absorbance?

2. 53.6 mL of a pure dye and 18.7 mL of water are mixed together. What is the percent, by volume, of dye in the solution?

3. Using **your** calibration curve, what is the absorbance of a 45.8% (by volume) solution?

   Mark this point with an X on your calibration curve.

   What would be the absorbance of the same solution using **your** equation for the calibration curve?

4. The color of a solution is the color of the light that is transmitted or passes through the solution.

   a) What was the color of your assigned solution? _____

   b) You ran your experiment using the wavelength of maximum absorbance. What was this wavelength?

   c) Compare the color of your solution with the colors associated with the four wavelengths of light available on the colorimeter. Why did the wavelength you used for your experiment give the greatest absorbance?