

Name: \_\_\_\_\_

Chemistry 117 Laboratory  
University of Massachusetts Boston

## CONCENTRATIONS

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In this experiment you will make a series of dilute solutions from a more concentrated stock solution. You will calculate molarities ( $M = \text{mol/L}$ ) of these solutions, and you will determine their absorbance of light as a function of concentration. From this you will obtain a straight-line calibration plot. Using this plot and its known linear equation, you will be able to determine the concentration of an unknown sample by measuring its light absorbance.

A transparent, colored substance, such as a solution, absorbs certain wavelengths of light and transmits the non - or weakly - absorbed wavelengths. It is these transmitted wavelengths that produce the color we see. For example, in this experiment we will use an aqueous solution of  $\text{CoCl}_2$ , which has a reddish purple color, not unlike dilute red-grape juice. This color is observed because  $\text{CoCl}_2(aq)$  absorbs the middle wavelengths of visible light, which correspond to the colors orange, yellow, and green. The transmitted wavelengths, then, correspond to blue and red, which together make purple.

As this suggests, a colored solution generally transmits light at many wavelengths, but the intensity of transmitted light usually is not the same at each wavelength. For a given wavelength of light, if we compare the intensity of the light shining on the sample,  $I_o$ , to its intensity after it has passed through the sample,  $I$ , we can define the *transmittance* ( $T$ ) as the ratio

$$(1) \quad T = I/I_o$$

The amount of light that was absorbed at the same wavelength is measured by the *absorbance* ( $A$ ), defined as the negative of the logarithm of the transmittance; i.e.,

$$(2) \quad A = -\log T = -\log(I/I_o) = \log(I_o/I) = \log(1/T)$$

For a given solute, there is often one wavelength (or a small range of wavelengths) at which the absorbance is greatest (and therefore the transmittance least). This wavelength is sometimes referred to as  $\lambda_{\text{max}}$ . For  $\text{CoCl}_2$ ,  $\lambda_{\text{max}}$  occurs at 510 nm.

For a solution, the actual value of the absorbance,  $A$ , at any wavelength such as  $\lambda_{\text{max}}$  depends upon the following three factors:

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$\epsilon$ , the *molar absorptivity*, a constant characteristic of the absorbing solute species;

$b$ , the *path length*, the distance the light travels in passing through the sample; and,

$c$ , the *concentration* in molarity (mol/L) of the absorbing solute species.

The relationship between these factors is called *Beer's Law* :

$$(3) \quad A = \epsilon bc$$

You will use a spectrophotometer (Spectronic 70) to measure absorbances of  $\text{CoCl}_2(aq)$  solutions at  $\lambda_{\text{max}} = 510 \text{ nm}$  using a particular sample tube placed in a certain instrument's beam of light. Therefore, both  $\epsilon$  and  $b$  will be constants throughout your measurements. This being the case, we can combine these constants into a single experimental constant,  $k$ , and rewrite Beer's Law as

$$(4) \quad A = kc$$

Absorbance is a unitless number, and  $c$  has units of mol/L, so the units on  $k$  must be L/mol or  $\text{M}^{-1}$ .

Note that equation (4) is a straight-line function of the form  $y = mx + b$ , where  $y$  is the dependent variable,  $x$  is the independent variable,  $m$  is the slope of the line, and  $b$  is the  $y$ -intercept. In equation (4), the constant  $k$  is the slope of the line, which can be determined from a plot of the absorbance ( $A$ , the dependant variable) versus concentration ( $c$ , the independent variable). When the concentration of the absorbing species is zero the absorbance must also be zero. Therefore, the  $y$ -intercept ( $b$  in the straight-line equation  $y = mx + b$ ) must be zero.

In constructing a Beer's Law plot, absorbances are measured for a series of solutions of different known concentrations. The straight line that best fits the data is drawn through the plotted data points, and the slope of the line is determined to obtain  $k$ . Once the value of  $k$  is known, it can be used to calculate the concentration of a sample with unknown concentration, using equation (4) and the measured absorbance of the solution under identical conditions to those used to obtain the Beer's Law plot.

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### IN THE LABORATORY

#### *Making a Beer's Law Plot of Absorbance vs. Concentration*

1. Obtain five clean, dry 18 x 150-mm test tubes, and label them 1 through 5. Fill test tube No. 1 approximately one-half to two-thirds full with 0.150 M  $\text{CoCl}_2$  stock solution.
2. Obtain a clean 5-mL Mohr pipet. Draw up about 1 mL of stock solution into the pipet from test tube 1. With your finger on the top of the pipet to hold the liquid in, tip the pipet on its side and roll the liquid around to wet the inside of the pipet with stock solution. Drain this wash liquid into a 100-mL or larger beaker, which you will continue to use as a temporary waste receptacle until you have finished with this experiment. At the end of the experiment, empty the contents of this beaker into the waste container for the laboratory.
3. Using the solution-rinsed Mohr pipet, measure the amounts of stock solution indicated in the table below into test tubes 2 through 5:

Test Tube No.	Volume 0.150 M $\text{CoCl}_2$	Volume deionized water	Molarity of $\text{CoCl}_2$
1	>5	0 mL	0.150 M
2	4.00 mL	1.00 mL	
3	3.00 mL	2.00 mL	
4	2.00 mL	3.00 mL	
5	1.00 mL	4.00 mL	

4. Rinse out the Mohr pipet with deionized water. Fill a small beaker with deionized water, and using the freshly rinsed pipet add the amounts of water indicated in the table above to the test tubes. The level of liquid in test tubes 2 through 5 should look the same (5 mL), unless you have made an error. Mix the solution in each test tube thoroughly by swirling it for about a minute. **Do not cover the test tube with a finger to shake the contents.** The level of the remaining solution in test tube 1 should be at least as high as the level in the other test tubes. If it is not, add additional stock 0.150 M  $\text{CoCl}_2$  solution, as needed.
5. Turn on the Spectronic 70 and set the wavelength to 510 nm, using the hand crank on the right side of the case. Rinse out the sample tube with deionized water, and then fill it to the horizontal mark. Place the water-filled sample tube in the sample chamber on top of the instrument, making sure to align the white perpendicular mark on the tube with the reference mark at the front of the hole of the chamber. Using the knob on the top of the instrument, adjust the

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absorbance reading (red scale) to read exactly zero. This sample is the blank sample. **The instrument settings from the blank must not be changed throughout the rest of the measurements.** Remove the sample tube.

6. You are now ready to measure the absorbances of your five samples. For each sample, using a clean and dry Pasteur pipet (long medicine dropper), add a small amount of the solution to be measured to the sample tube, swirl it around to wet the sides, and empty this wash solution into your temporary waste beaker. Then, with the same Pasteur pipet, fill the sample tube to the horizontal mark with the solution to be measured. Measure the absorbance. (Take care in reading the red absorbance scale -- it increases right to left and does not have the same scaling throughout.) Repeat the process for all five samples, and record your results in the following table:

Sample No.	Molarity of $\text{CoCl}_2$	Absorbance
1	0.150 M	
2		
3		
4		
5		
blank	0	0

7. Plot absorbance vs. molarity on the graph paper provided. **Use pencil, not pen, for graphing.** Absorbance should appear on the vertical axis (ordinate), and concentration in mol/L should appear on the horizontal axis (abscissa). Each data point should be a small, blackened circle. Also, drawing a small circle around each data point increases visibility. Using a ruler, draw a straight line that passes more or less equally between all the data points and which also passes through the origin (zero absorbance at zero concentration). This is your Beer's Law calibration graph.
8. Determine the slope of the line you have drawn through your data points. What is your experimentally determined value of  $k$  in the Beer's Law equation  $A = kc$ ?

$$k = \text{_____} \text{ M}^{-1}$$

**Do not change any instrument settings. Use the same sample tube for the next part of this experiment.**

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*Determining the Concentration of An Unknown Sample of  $\text{CoCl}_2(\text{aq})$*

1. From your instructor, obtain a sample of the  $\text{CoCl}_2$  solution that has an unknown concentration. The unknown solution is more concentrated than any of the solutions you used in making the Beer's Law plot. In fact, it is too concentrated to obtain an accurate absorbance reading with the Spectronic 70. Therefore you will have to make a dilute solution that gives an absorbance reading in the range of your Beer's Law plot.
2. Using the Mohr pipet, measure a portion of your unknown solution into a clean, dry 18 x 150-mm test tube. Record the volume of unknown solution used.

Volume of unknown = \_\_\_\_\_

3. Now, successively add measured amounts of water to the sample, taking absorbance readings after each dilution, until you make a solution with an absorbance in the range of your Beer's Law plot. In the table below, record the amount of water added to make each successive dilution and the absorbance reading of the solution at each stage, using as many lines as needed to get to the successful solution. If the absorbance is "off scale" at any point, enter ">2" for the absorbance reading.

Dilution No.	mL $\text{H}_2\text{O}$ added	Absorbance

Total volume of water added to make a successful solution = \_\_\_\_\_

4. Successively adding measured amounts of water results in accumulated errors in determining the final solution composition. Therefore, make up a new test solution with the same composition, using a single pipetting of the required amount of water. Measure the absorbance of this solution, and determine its concentration by using the value of  $k$  from your Beer's Law plot. Record your data and calculated values below.

Volume of unknown  $\text{CoCl}_2$  solution taken = \_\_\_\_\_

Volume of  $\text{H}_2\text{O}$  added to make dilute solution = \_\_\_\_\_

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Absorbance of dilute solution = \_\_\_\_\_

Concentration of dilute solution = \_\_\_\_\_

Concentration of original unknown solution = \_\_\_\_\_

5. Compare your results for the concentration of the original unknown solution with those of other students working around you.

Students' Names	Calculated concentration, (M)

Including your result with those shown above, calculate the mean and standard deviation of the concentration of the unknown solution.

Mean concentration = \_\_\_\_\_

Standard deviation =  $\pm$  \_\_\_\_\_