

Determining the Concentration of a Solution: Beer's Law

The primary objective of this experiment is to determine the concentration of an unknown nickel (II) sulfate solution. You will be using the Colorimeter shown in Figure 1. In this device, red light from the LED light source will pass through the solution and strike a photocell. The NiSO_4 solution used in this experiment has a deep green color. A higher concentration of the colored solution absorbs more light (and transmits less) than a solution of lower concentration. The Colorimeter monitors the light received by the photocell as either an *absorbance* or a *percent transmittance* value.



Figure 1

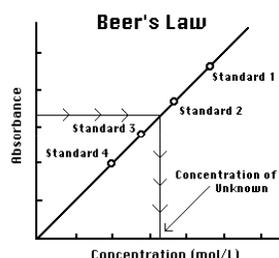


Figure 2

You are to prepare five nickel sulfate solutions of known concentration (standard solutions). Each is transferred to a small, rectangular cuvette that is placed into the Colorimeter. The amount of light that penetrates the solution and strikes the photocell is used to compute the absorbance of each solution. When a graph of absorbance vs. concentration is plotted for the standard solutions, a direct relationship should result, as shown in Figure 2. The direct relationship between absorbance and concentration for a solution is known as Beer's law.

The concentration of an *unknown* NiSO_4 solution is then determined by measuring its absorbance with the Colorimeter. By locating the absorbance of the unknown on the vertical axis of the graph, the corresponding concentration can be found on the horizontal axis (follow the arrows in Figure 2). The concentration of the unknown can also be found using the slope of the Beer's law curve.

OBJECTIVES

In this experiment, you will

- Prepare NiSO_4 standard solutions.
- Use a Colorimeter to measure the absorbance value of each standard solution.
- Find the relationship between absorbance and concentration of a solution.
- Use the results of this experiment to determine the unknown concentration of another NiSO_4 solution.

MATERIALS

computer	30 mL of 0.40 M NiSO ₄
Vernier computer interface	5 mL of NiSO ₄ unknown solution
LoggerPro	two 10 mL pipets (or graduated cylinders)
Vernier Colorimeter	pipet pump or pipet bulb
one cuvette	distilled water
five 20 × 150 mm test tubes	test tube rack
tissues (preferably lint-free)	two 100 mL beakers
stirring rod	

PROCEDURE

1. Obtain and wear goggles! **CAUTION:** Be careful not to ingest any NiSO₄ solution or spill any on your skin. Inform your teacher immediately in the event of an accident.
2. Add about 30 mL of 0.40 M NiSO₄ stock solution to a 100 mL beaker. Add about 30 mL of distilled water to another 100 mL beaker.
3. Label four clean, dry, test tubes 1-4 (the fifth solution is the beaker of 0.40 M NiSO₄). Pipet 2, 4, 6, and 8 mL of 0.40 M NiSO₄ solution into Test Tubes 1-4, respectively. With a second pipet, deliver 8, 6, 4, and 2 mL of distilled water into Test Tubes 1-4, respectively. *Thoroughly* mix each solution with a stirring rod. Clean and dry the stirring rod between stirrings. Keep the remaining 0.40 M NiSO₄ in the 100 mL beaker to use in the fifth trial. Volumes and concentrations for the trials are summarized below:

Trial number	0.40 M NiSO ₄ (mL)	Distilled H ₂ O (mL)	Concentration (M)
1	2	8	0.08
2	4	6	0.16
3	6	4	0.24
4	8	2	0.32
5	~10	0	0.40

4. Connect the Colorimeter to the computer interface. Prepare the computer for data collection by opening the file “11 Beer’s Law” from the *Chemistry with Computers* folder of LoggerPro.
5. You are now ready to calibrate the Colorimeter. Prepare a *blank* by filling a cuvette 3/4 full with distilled water. To correctly use a Colorimeter cuvette, remember:
 - All cuvettes should be wiped clean and dry on the outside with a tissue.
 - Handle cuvettes only by the top edge of the ribbed sides.
 - All solutions should be free of bubbles.
 - Always position the cuvette with its reference mark facing toward the white reference mark at the top of the cuvette slot on the Colorimeter.
6. Calibrate the Colorimeter.
 - a. Open the Colorimeter lid.
 - b. Holding the cuvette by the upper edges, place it in the cuvette slot of the Colorimeter. Close the lid.

- c. If your Colorimeter has a CAL button, Press the < or > button on the Colorimeter to select a wavelength of 635 nm (Red) for this experiment. Press the CAL button until the red LED begins to flash. Then release the CAL button. When the LED stops flashing, the calibration is complete. Proceed directly to Step 7. If your Colorimeter does not have a CAL button, continue with this step to calibrate your Colorimeter.

First Calibration Point

- d. Choose Calibrate ► CH1: Colorimeter (%T) from the Experiment menu and then click .
- e. Turn the wavelength knob on the Colorimeter to the “0% T” position.
- f. Type “0” in the edit box.
- g. When the displayed voltage reading for Reading 1 stabilizes, click .

Second Calibration Point

- h. Turn the knob of the Colorimeter to the Red LED position (635 nm).
- i. Type “100” in the edit box.
- j. When the displayed voltage reading for Reading 2 stabilizes, click , then click .
7. You are now ready to collect absorbance data for the five standard solutions. Click . Empty the water from the cuvette. Using the solution in Test Tube 1, rinse the cuvette twice with ~1 mL amounts and then fill it 3/4 full. Wipe the outside with a tissue and place it in the Colorimeter. After closing the lid, wait for the absorbance value displayed on the monitor to stabilize. Then click type “0.080” in the edit box, and press the ENTER key. The data pair you just collected should now be plotted on the graph.
8. Discard the cuvette contents as directed by your teacher. Rinse the cuvette twice with the Test Tube 2 solution, 0.16 M NiSO₄, and fill the cuvette 3/4 full. Wipe the outside, place it in the Colorimeter, and close the lid. When the absorbance value stabilizes, click , type “0.16” in the edit box, and press the ENTER key.
9. Repeat the Step 8 procedure to save and plot the absorbance and concentration values of the solutions in Test Tube 3 (0.24 M) and Test Tube 4 (0.32 M), as well as the stock 0.40 M NiSO₄. Wait until Step 12 to do the unknown. When you have finished with the 0.40 M NiSO₄ solution, click .
10. In your Data and Calculations table, record the absorbance and concentration data pairs that are displayed in the table.
11. Examine the graph of absorbance vs. concentration. To see if the curve represents a direct relationship between these two variables, click the Linear Fit button, . A best-fit linear regression line will be shown for your five data points. This line should pass near or through the data points *and* the origin of the graph. (Note: Another option is to choose Curve Fit from the Analyze menu, and then select Proportional. The Proportional fit has a y-intercept value equal to 0; therefore, this regression line will always pass through the origin of the graph).
12. Obtain about 5 mL of the unknown NiSO₄ in another clean, dry, test tube. Record the number of the unknown in the Data and Calculations table. Rinse the cuvette twice with the unknown solution and fill it about 3/4 full. Wipe the outside of the cuvette, place it into the Colorimeter, and close the lid. Read the absorbance value displayed in the meter. (**Important:** The reading in the meter is live, so it is **not** necessary to click to read the absorbance value.) When the displayed absorbance value stabilizes, record its value in Trial 6 of the Data and Calculations table.

13. Discard the solutions as directed by your teacher. Proceed directly to Steps 1 and 2 of Processing the Data.

PROCESSING THE DATA

1. Use the following method to determine the unknown concentration. With the linear regression curve still displayed on your graph, choose Interpolate from the Analyze menu. A vertical cursor now appears on the graph. The cursor's concentration and absorbance coordinates are displayed in the floating box. Move the cursor along the regression line until the absorbance value is approximately the same as the absorbance value you recorded in Step 12. The corresponding concentration value is the concentration of the unknown solution, in mol/L.
2. Print a graph of absorbance vs. concentration, with a regression line and interpolated unknown concentration displayed. To keep the interpolated concentration value displayed, move the cursor straight up the vertical cursor line until the tool bar is reached. Enter your name(s) and the number of copies of the graph you want.

DATA AND CALCULATIONS

Trial	Concentration (mol/L)	Absorbance
1	0.080	
2	0.16	
3	0.24	
4	0.32	
5	0.40	
6	Unknown number ____	
Concentration of unknown		mol/L

QUESTIONS FOR DISCUSSION

1. Why was a wavelength of light in the "red range" used in this lab, when you were dealing with a solution that was green in color?
2. In the previous lab, you determined concentration of an unknown by mass (density). Which method, absorbance or density appears to be more reliable for determining the concentration of unknowns? Give evidence to support your answer.