Experiment
11

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.

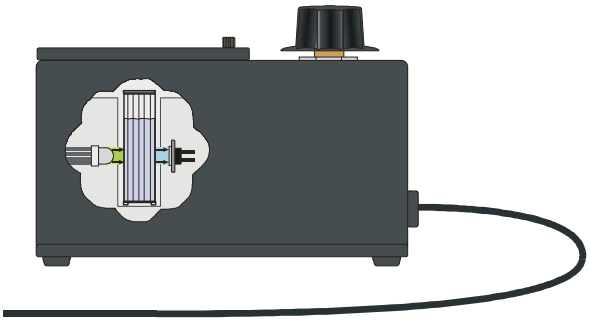


Figure 1

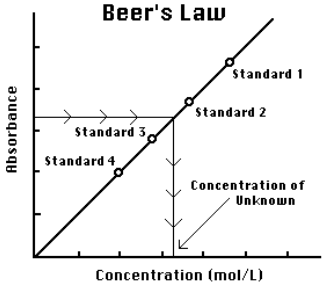


Figure 2

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.

MATERIALS

LabPro or CBL 2 interface
TI Graphing Calculator
DataMate program
Vernier Colorimeter
one cuvette
five 20 x 150 mm test tubes
30 mL of 0.40 M $NiSO_4$
5 mL of $NiSO_4$ unknown solution
two 10-mL pipets (or graduated cylinders)
two 100-mL beakers
pipet or pipet bulb
distilled water
test tube rack
stirring rod
tissues (preferably lint-free)

PROCEDURE

1. Obtain and wear goggles! **CAUTION:** *Be careful not to ingest any $NiSO_4$ 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 $NiSO_4$ 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 $NiSO_4$). Pipet 2, 4, 6, and 8 mL of 0.40 M $NiSO_4$ 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 $NiSO_4$ 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_2O (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. Plug the Colorimeter into Channel 1 of the LabPro or CBL 2 interface. Use the link cable to connect the TI Graphing Calculator to the interface. Firmly press in the cable ends.

5. Prepare a *blank* by filling an empty cuvette ¾ full with distilled water. Seal the cuvette with a lid. 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 right of the cuvette slot on the Colorimeter.

6. Turn on the calculator and start the DATAMATE program. Press CLEAR to reset the program.

7. Set up the calculator and interface for the Colorimeter.
   a. Place the blank in the cuvette slot of the Colorimeter and close the lid.
   b. Select SETUP from the main screen.
   c. If the calculator displays COLORIMETER in CH 1, set the wavelength on the Colorimeter to 635 nm (Red). Then calibrate by pressing the AUTO CAL button on the Colorimeter and proceed directly to Step 8. If the calculator does not display COLORIMETER in CH1, continue with this step to set up your sensor manually.
   d. Press ENTER to select CH 1.
   e. Select COLORIMETER from the SELECT SENSOR menu.
   f. Select CALIBRATE from the SETUP menu.
   g. Select CALIBRATE NOW from the CALIBRATION menu.

First Calibration Point

h. Turn the wavelength knob of the Colorimeter to the 0% T position. When the voltage reading stabilizes, press ENTER. Enter "0" as the percent transmittance.

Second Calibration Point

i. Turn the wavelength knob of the Colorimeter to the Red LED position (635 nm). When the voltage reading stabilizes, press ENTER. Enter "100" as the percent transmittance.

j. Select OK to return to the setup screen.

8. Set up the data-collection mode.

   a. To select MODE, press ▲ once and press ENTER.

   b. Select EVENTS WITH ENTRY from the SELECT MODE menu.

   c. Select OK to return to the main screen.

9. You are now ready to collect absorbance-concentration data for the five standard solutions.

   a. Select START from the main screen.

   b. 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, place it in the Colorimeter, and close the lid.

   c. When the value displayed on the calculator screen has stabilized, press ENTER. Enter "0.080" as the concentration in mol/L. The absorbance and concentration values have now been saved for the first solution.

   d. Discard the cuvette contents as directed by your instructor. Using the solution in Test Tube 2, rinse the cuvette twice with ~1-mL amounts, and then fill it 3/4 full. After closing the lid, wait for the value displayed on the calculator screen to stabilize and press ENTER. Enter "0.16" as the concentration in mol/L.

   e. Repeat the procedure for Test Tube 3 (0.24 M) and Test Tube 4 (0.32M), as well as the stock 0.40 M $NiSO_4$. **Note:** Wait until Step 10 to do the unknown.

   f. Press STO▶ to stop data collection. The absorbance and concentration values have now been saved for the standard solutions.

   g. Examine the data points along the curve on the displayed graph. As you move the cursor right or left, the concentration (X) and absorbance (Y) values of each data point are displayed below the graph. Record the absorbance values in your data table (round to the nearest 0.001).

   h. Press ENTER to return to the main screen.

10. Determine the absorbance value of the unknown $NiSO_4$ solution. To do this:

    a. Obtain about 5 mL of the *unknown* $NiSO_4$ in another clean, dry, test tube. Record the number of the unknown in your data table.

    b. 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.

    c. Monitor the absorbance value displayed on the calculator. When this value has stabilized, record it in your data table (round to the nearest 0.001).

    d. Dispose of the remaining solution as directed by your instructor.

11. Discard the solutions as directed by your instructor. Proceed directly to Steps 1–2 of Processing the Data.

PROCESSING THE DATA

1. To determine the concentration of the unknown $NiSO_4$ solution, plot a graph of absorbance *vs*. concentration with a linear regression curve displayed, then interpolate along the regression line to convert the absorbance value of the unknown to concentration. To do this:
   a. Select ANALYZE from the main screen.
   b. Select CURVE FIT from the ANALYZE OPTIONS menu.
   c. Select LINEAR (CH 1 VS ENTRY) from the CURVE FIT menu. The linear-regression statistics for these two lists are displayed for the equation in the form

      $$y = ax + b$$

      where *y* is absorbance, *x* is concentration, *a* is the slope, and *b* is the y-intercept.
      Note: One indicator of the quality of your data is the size of *b*. It is a very small value if the regression line passes through or near the origin. The correlation coefficient, *r*, indicates how closely the data points match up with (or *fit*) the regression line. A value of 1.00 indicates a nearly perfect fit.
   d. To display the linear-regression curve on the graph of absorbance *vs*. concentration, press ENTER. This graph should indicate a direct relationship between absorbance and concentration, a relationship known as Beer's law. The regression line should closely fit the five data points *and* pass through (or near) the origin of the graph.
   e. To interpolate along the curve, press ▲. A cursor is displayed on the regression line, along with its X and Y coordinates below the graph. Use ► or ◄ to move the cursor to an absorbance value (Y value) that is closest to the absorbance reading you obtained in Step 10. The $NiSO_4$ concentration, in mol/L, will be equal to the corresponding X value. Record this value in your data table.

2. Print a graph of absorbance *vs*. concentration, with a regression line and interpolated unknown concentration displayed.

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

TEACHER INFORMATION

Determining the Concentration of a Solution: Beer's Law

1. The light source for the nickel (II) sulfate solution is the red LED (635 nm). Since the $NiSO_4$ is green in color, the nearly monochromatic red light is readily absorbed by the solution.

2. The 0.40 M $NiSO_4$ solution can be prepared by using 10.51 g of $NiSO_4 \cdot 6H_2O$ per 100 mL. **HAZARD ALERT:** Toxic; avoid dispersing this substance; dispense with care; alleged carcinogen. Hazard Code: B—Hazardous.

   The hazard information reference is: Flinn Scientific, Inc., *Chemical & Biological Catalog Reference Manual, 2000*, P.O. Box 219, Batavia, IL 60510. See *Appendix F* of this book, *Chemistry with Calculators*, for more information.

3. Solutions of $Ni(NO_3)_2$ also work well, and can be prepared by using 11.63 g of solid $Ni(NO_3)_2 \cdot 6H_2O$ per 100 mL of solution.

4. Unknowns can be prepared by doing dilutions starting with the 0.40 M $NiSO_4$ stock solution. For example, to prepare a 0.22 M unknown, use 55 mL of the standard plus 45 mL of water:

$$(55 \text{ mL}/100 \text{ mL})(.40 \text{ M}) = 0.22 \text{ M}$$

5. This experiment works well using solutions of green food coloring. A solution with an absorbance similar to 0.40 M $NiSO_4$ can be prepared by dissolving 8-9 drops of green Schilling Food Coloring in 1 liter of water. Check to see that this stock solution has an absorbance in the range of 0.35 to 0.55. Assign this solution a concentration of 100%. Students will follow the same procedure to dilute the stock solution to 80%, 60%, 40%, and 20%.

6. The cuvette must be from 55% to 100% full in order to get a valid absorbance reading. If students fill the cuvette 3/4 full, as described in the procedure, they should easily be in this range. To avoid spilling solution into the cuvette slot, remind students not to fill the cuvette to the brim.

7. Since there is some variation in the amount of light absorbed by the cuvette if it is rotated 180°, you should use a water-proof marker to make a reference mark on the top edge of one of the clear sides of all cuvettes. Students are reminded in the procedure to align this mark with the reference mark to the right of the cuvette slot on the colorimeter.

8. The use of a single cuvette in the procedure eliminates errors introduced by slight variations in the absorbance of different plastic cuvettes. If one cuvette is used throughout the experiment by a student group, this variable is eliminated. The two rinses done prior to adding a new solution can be accomplished very quickly.

9. As an alternative to the rinses suggested above, we also recommend the use of cotton swabs, or Q-tips, to dry a cuvette after water or a solution has been emptied from it. The cotton swab easily removes any remaining droplets in the cuvette. After completely drying the cuvette using both ends of the cotton swab, it is no longer necessary to rinse with the new solution.

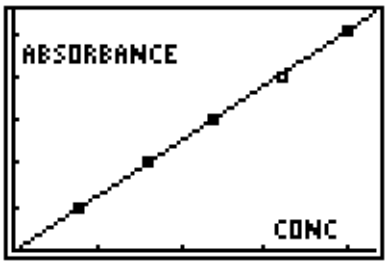
10. Satisfactory results can be obtained if a different cuvette is used for each solution of the experiment. For best results, the cuvettes for one student lab team should be *matched*. Each cuvette in a matched set absorbs light (when empty) at approximately the same level. To match a set of cuvettes, first calibrate the colorimeter using the method described in the section on calibration. Use a clean, dry cuvette for the 100% calibration instead of a distilled water blank. Put a reference mark on one of the clear sides of the cuvette so it is always oriented the same way in the cuvette slot. Place each cuvette in the batch into the colorimeter and record percent transmittance values for each. When you are finished, group cuvettes according to similar %T values. Each of these groups represents a set of matched cuvettes.

11. In addition to absorbance (L2) and concentration (L1), percent transmittance data is stored in L5 when using EVENTS WITH ENTRY mode in the DataMate data-collection program.

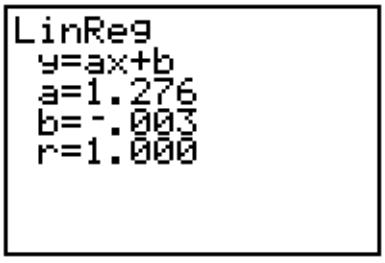
SAMPLE RESULTS

Trial	Concentration (mol / L)	Absorbance
1	0.080	0.100
2	0.16	0.201
3	0.24	0.302
4	0.32	0.403
5	0.40	0.510
6	Unknown number 1	0.360

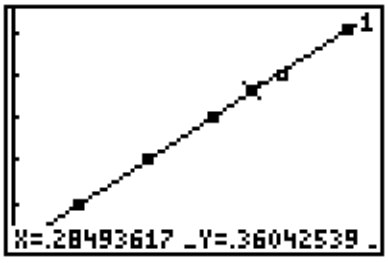
Concentration of the unknown	0.282 mol/L



Absorbance vs. Concentration



Regression Statistics



Interpolating an Unknown