Determining the Concentration of a Solution: Beer's Law

The primary objective of this experiment is to determine the concentration of an unknown copper (II) sulfate solution. The $CuSO_4$ solution used in this experiment has a blue color, so Colorimeter users will be instructed to use the red LED. Spectrometer users will determine an appropriate wavelength based on the absorbance spectrum of the solution. A higher concentration of the colored solution absorbs more light (and transmits less) than a solution of lower concentration.

You will prepare five copper (II) sulfate solutions of known concentration (standard solutions). Each solution is transferred to a small, rectangular cuvette that is placed into the Colorimeter or Spectrometer. The amount of light that penetrates the solution and strikes the photocell is used to compute the absorbance of each solution. When you graph absorbance *vs*. concentration for the standard solutions, a direct relationship should result. The direct relationship between absorbance and concentration for a solution is known as *Beer's law*.

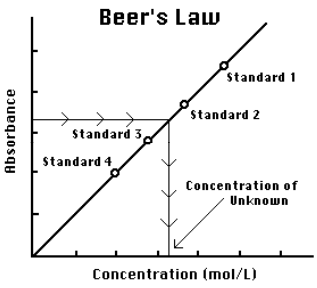

Figure 1

You will determine the concentration of an unknown $CuSO_4$ solution by measuring its absorbance. By locating the absorbance of the unknown on the vertical axis of the graph, the corresponding concentration can be found on the horizontal axis. The concentration of the unknown can also be found using the slope of the Beer's law curve.

MATERIALS

LabQuest
LabQuest App
Vernier Colorimeter or Spectrometer
one cuvette
five 20 × 150 mm test tubes
two 10 mL pipets or graduated cylinders
two 100 mL beakers
0.40 M copper (II) sulfate, $CuSO_4$, solution
copper (II) sulfate, $CuSO_4$, unknown solution
pipet pump or pipet bulb
distilled water
test tube rack
stirring rod
tissues (preferably lint-free)

PROCEDURE

1. Obtain and wear goggles.

2. Obtain small volumes of 0.40 M $CuSO_4$ solution and distilled water in separate beakers.

3. Label five clean, dry, test tubes 1–5. Use pipets to prepare five standard solutions according to the chart below. Thoroughly mix each solution with a stirring rod. Clean and dry the stirring rod between uses.

Test Tube	0.40 M $CuSO_4$ (mL)	Distilled H_2O (mL)	Concentration (M)
1	2	8	0.080
2	4	6	0.16
3	6	4	0.24
4	8	2	0.32
5	~10	0	0.40

4. Prepare a *blank* by filling a cuvette 3/4 full with distilled water. To correctly use cuvettes, remember:
   - Wipe the outside of each cuvette with a lint-free tissue.
   - Handle cuvettes only by the top edge of the ribbed sides.
   - Dislodge any bubbles by gently tapping the cuvette on a hard surface.
   - Always position the cuvette so the light passes through the clear sides.

5. Connect the Colorimeter to LabQuest and choose New from the File menu.

6. Calibrate the Colorimeter.
   a. Place the blank in the cuvette slot of the Colorimeter and close the lid.
   b. Press the < or > button on the Colorimeter to select the wavelength of 635 nm (Red). Then calibrate by pressing the CAL button on the Colorimeter. When the LED stops flashing, the calibration is complete.

7. Set up the data-collection mode.
   a. On the Meter screen, tap Mode. Change the mode to Events with Entry.
   b. Enter the Name (Concentration) and Units (mol/L). Select OK.
   c. Proceed to Step 8.

8. You are now ready to collect absorbance-concentration data for the five standard solutions.
   a. Start data collection.
   b. 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 device. (Close the lid of the Colorimeter.)
   c. When the value displayed on the screen has stabilized, tap Keep and enter **0.080** as the concentration in mol/L. Select OK. The absorbance and concentration values have now been saved for the first solution.
   d. Discard the cuvette contents as directed. Using the solution in Test Tube 2, rinse and fill the cuvette 3/4 full. Wipe the outside and place the cuvette in the device (close the lid of the Colorimeter). Wait for the value displayed on the screen to stabilize, and tap Keep. Enter **0.16** as the concentration in mol/L.
   e. Repeat the procedure for Test Tubes 3 and 4. Trial 5 is the original 0.40 M $CuSO_4$ solution. **Note:** Do not test the unknown solution until Step 11.
   f. When you have finished testing the standard solutions, stop data collection.
   g. To examine the data pairs on the displayed graph, tap any data point. As you tap each data point, the absorbance and concentration values are displayed to the right of the graph.

9. Write down the absorbance values, for each of the five trials, in your data table.

10. Display a graph of absorbance *vs.* concentration with a linear regression curve.
    a. Choose Graph Options from the Graph menu.
    b. Select Autoscale from 0 and select OK.
    c. Choose Curve Fit from the Analyze menu.
    d. Select Linear as the Fit Equation. The linear-regression statistics for these two data columns are displayed for the equation in the form: $y = mx + b$ where x is concentration, y is absorbance, 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.
    e. Select OK. The 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.

11. Determine the absorbance and concentration values of the unknown $CuSO_4$ solution.
    a. Tap the Meter tab.
    b. Obtain about 5 mL of the *unknown* $CuSO_4$ in another clean, dry, test tube. Record the number of the unknown in your data table.
    c. Rinse the cuvette twice with the unknown solution and fill it about 3/4 full. Wipe the outside of the cuvette and place it into the device (close the lid of the Colorimeter).
    d. Monitor the absorbance value. When this value has stabilized, record it in your data table.
    e. Tap the Graph tab.
    f. On the Graph screen, choose Interpolate from the Analyze menu. Tap any point on the regression curve (or use the ◄ or ► keys on LabQuest) to find the absorbance value that is closest to the absorbance reading you obtained in Step 10. Determine the concentration of your unknown $NiSO_4$ solution and record the concentration in your data table.

DATA TABLE

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