Honors Chemistry Lab 14: **Colorimetry and 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, 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.

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.

Objectives:

•Prepare and test the absorbance of five standard copper(II) sulfate solutions.

•Calculate a standard curve from the test results of the standard solutions.

•Test the absorbance of a copper(II) sulfate solution of unknown molar concentration.

•Calculate the molar concentration of the unknown $CuSO_4$ solution.

Materials: LabQuest; 0.40 M copper(II) sulfate solution, ? M copper(II) sulfate solution, Vernier Colorimeter or Spectrometer, pipet pump or pipet bulb, one cuvette, distilled water, five 20 × 150 mm test tubes, test tube rack, two 10 mL pipets or graduated cylinders, stirring rod, two 100 mL beakers, Kimwipes

Procedure: Both Colorimeter and Spectrometer Users

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. You will need to calculate the concentration (M) column on the table. 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	
2	4	6	
3	6	4	
4	8	2	
5	~10	0	

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.

Spectrometer Users Only (Colorimeter users proceed to the Colorimeter section)

5. Calibrate the Spectrometer.

a. Place the blank cuvette in the Spectrometer.

6. Determine the optimal wavelength for creating this standard curve and set up the data collection mode.

a. Remove the blank cuvette, and place the 0.40 M standard into the cuvette slot.

b. Start data collection.

Colorimeter Users Only (Spectrometer users proceed to the next section)

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.

Both Colorimeter and Spectrometer Users

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 (Colorimeter or Spectrometer). Close the lid of the Colorimeter.

c. After 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. After the value displayed on the screen has stabilized, tap Keep. Enter 0.16 as the concentration in mol/L.

e. Repeat the procedure for Test Tubes 3 – 5. 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. Spectrophotometer users will have to generate their graph using Excel or manually in their notebooks.

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. Select OK. The best-fit linear regression line will be shown on the graph for your five data points.

11. Determine the absorbance and concentration values of the unknown CuSO4 solution.

a. Tap the Meter tab.

b. Obtain about 5 mL of the unknown CuSO4 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.

d. Monitor the absorbance value. After 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. Find the absorbance value closest to the absorbance reading you obtained in Step 11d. Determine the concentration of your unknown CuSO4 solution and record it in your 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 ____	

Data Analysis:

1. What is the molar concentration of your unknown sample of copper(II) sulfate solution?

2. What factors are included in the Beer's law expression for determining how much light passes through a liquid solution?

3. How would your test results be affected if you left fingerprints on the sides of the cuvette in line with the light path of the spectrometer (or colorimeter)?

4. Could this method of testing be used to determine the concentration of a NaCl solution?