The Gatorade Lab: What Is the Relationship Between the Concentration of a Solution and the Amount of Transmitted Light Through the Solution?

■Central Challenge

“How can light be used to study color and determine concentrations of chemical species in solutions?”

■Context for This Investigation

Measuring how much of which wavelengths of light are absorbed by a substance, and getting useful information about that substance from the results, is the scientific discipline of spectroscopy. The visible spectrum is the only part of the electromagnetic spectrum that we can access with the equipment found in a typical high school chemistry laboratory. The basic principles of spectral analysis that you learn in school can also be applied to the more sophisticated instrumentation required to access the ultraviolet, infrared, and X-ray regions.

What you learn by performing the lab will help you to understand more sophisticated instruments that you may encounter at a college or university. In a visible spectrophotometer, students shine a beam of light into a solution containing the sample, and detect how much of it comes out of the other side of the solution. By comparing the amount of light transmitted by the pure solvent to the amount transmitted when the sample is dissolved in it, we can calculate a quantity called the absorbance. Spectrophotometers can report measurements as percent transmittance (%T) or directly as absorbance. In this investigation, you will discover the relationship between transmittance and concentration and ultimately the relationship between transmittance, absorbance, and concentration of a solution.

Safety and Disposal

All of the food dyes can be flushed down the sink with plenty of water.

■Prelab Guiding Questions

Step 1: You will receive a stock solution of the blue #1 (Brilliant Blue) molecule dissolved in water with a known concentration. The chemical species in the solution for which you will be measuring transmittance is shown in Figure 1 .



Step 2: Explore how light, transmitted by a solution, is related to the concentration of the solution. The appropriate wavelength to take transmittance measurements using the SPEC 20 for this dye is 630 nm.

Step 3: Gather data as to the percent transmittance of the molecule in solution at various concentrations. Each group should perform a range of dilutions and then collect transmittance data and report such data back to the entire group.

Step 4: Record the % transmittance of their diluted solutions of known concentration using the SPEC 20 (or colorimeter) in a data table in a central location in the lab.

Step 5: Determine the relationship between transmittance and molarity of the solution. Students can do this by graphing the data in the data table (plot μM on the x-axis versus transmittance as a decimal on the y-axis).



Step 6: It would be really useful if the class got a straight line that goes through zero. Scientists try to find linear relationships because such relationships make it easier to identify unknowns and predict outcomes of investigations. It is often helpful to have the slope of the relationship between the dependent and independent variable be positive. What mathematical routine could we choose to plot to get a linear relationship between transmittance and molarity that has a positive slope, and which line goes through zero? Try out some of these techniques and report back to the whole group:

1/T versus [dye]

1x10T versus [dye]

logT versus [dye]

–logT versus [dye]

■Explanation to Strengthen Student Understanding

Transmittance and Absorbance Relationship

The relationship between transmittance and absorbance is Abs = – log10 (T) where T is the transmittance, rather than the %T (i.e., 0.50, not 50%). When the absorbance is 1.0, only 10% of the light beam is reaching the detector. When Abs = 2, only 1% of the light beam is reaching the detector and when Abs = 3, only 0.1% of the light is reaching the detector. It should be no surprise that the accuracy and sensitivity of low-cost instruments start to suffer at absorbance values higher than 1.5. If a sample shows an absorbance higher than 1.5, it is a good idea to dilute the sample by a factor of 5 or 10 and remeasure. The extra work is paid back in better accuracy in your measurement. A useful rule is don’t accept measurements of >1.5Abs. If you get such an absorbency value, dilute the sample and measure it again.

■Investigation

Many common sports drinks contain blue #1 dye. Students will use the relationship between transmittance, absorbance, and concentration (as well as their calibration line from the prelab) to determine the concentration of this dye in the sports drink.

Procedure

Obtain a sample of the blue-colored sports drink. Use the skills and information gained in the prelab to design a data-collection and data analysis procedure to determine the molarity or concentration of blue #1 dye in the sports drink.

Data Collection and Computation

1. Determine the molar concentration of blue #1 dye in the sports drink. Show all work.
2. Determine the mass of blue #1 dye found in 500 mL of the drink. Show all work.

Argumentation and Documentation

In the conclusion of lab, have students justify the procedure they chose, the instrumentation they used, and the selection of the kind of data they decided was needed to determine the concentration of blue #1 dye in a blue-colored sports drink containing only this dye.

■Postlab Assessment

1. Suppose a solution was too concentrated for an accurate reading with the spectrophotometer. The concentrated solution was diluted by placing 1.00 mL of the concentrated solution in 4.00 mL of water. The solution was then placed in the spectrophotometer and an absorbance was obtained and after a few calculations the molar concentration was calculated to be 3.5 Å~ 10–6 M. What was the concentration of the original stock solution before dilution?

2. If a 0.10 M solution of a colored substance has a maximum absorbance at 500 nm and an absorbance of 0.26 M at this wavelength, what will be the measured absorbance of a 0.20 M solution at 500 nm?

3. The spectrophotometer really measures the percent of light that is transmitted through the solution. The instrument then converts %T (transmittance) into absorbance by using the equation you determined in the prelab section. If the absorbance of a sample is 0.85, what is the percent of light transmitted through the colored sample at this collected wavelength?

How Can Color Be Used to Determine the Mass Percent of Copper in Brass?

■Central Challenge

What are the relationships between color, wavelength, absorbance, and concentration? Design an experiment that can quantitatively measure the absorption of light by a colored solution in order to determine the concentration of the absorbing species in that solution.

■Context for This Investigation

Spectrophotometry is an extremely important tool used in forensic science to determine the detailed chemical composition of evidence obtained from a crime scene. It can be used to determine the concentration of either a single chemical species in solution or even the concentration of a species within a mixture of species in solution. For example, it can be used to determine the mass percent of copper in brass shell casings collected by the crime scene investigator (CSI), and then match the brass composition to a particular manufacturer.

Safety and Disposal

Concentrated nitric acid is corrosive and will attack and destroy metals, proteins, and most plastics. Avoid skin contact and neutralize any spills with baking soda, then rinse with copious amounts of water. The acid will discolor the skin for days after contact, so be sure to wear rubber gloves. The NO gas that forms quickly oxidizes in air to produce a toxic, reddish-brown gas of NO2. For more information, read the Material Safety Data Sheet (MSDS) for nitric acid found at <http://www.ehso.com/msds.php> Perform the addition of nitric acid to the brass solid under a fume hood. If you do not have a fume hood, you should not perform this lab. Take normal laboratory precautions, including wearing splash-proof goggles and chemical-resistant gloves and apron at all times. The remaining brass solution should be neutralized by adding small amounts of solid baking soda until the bubbling has subsided and the pH is 5–9. Then the waste solution can be safely disposed of following standard procedures as directed by your instructor.

Prelab Preparation

Ideally, each lab group should have a spectrophotometer or colorimeter to determine the absorbance of their solutions. However, by first preparing all solutions, then measuring the absorbance values of one solution after another, it is possible to complete this lab with only a few of these instruments in the classroom. The brass sample should have a mass of 1–2 grams, and ensure that there is an excess of nitric acid available to dissolve the brass sample completely.

■Explanation to Strengthen Student Understanding

This experiment involves the relationships between color, wavelength, absorbance, and concentration. These factors are summarized by the Beer-Lambert law, which states that the amount of light absorbed, A, is related to the concentration of the Cu2+(aq), c, by the equation A = abc, where “a” is the molar absorptivity constant whose value depends upon the wavelength used and substance in solution, and b is the thickness (or depth) of the sample. When a graph of absorbance against concentration is plotted for the standard solutions, at one wavelength, a direct relationship should result, as shown in Figure 6.

You must determine which wavelength of visible light is most appropriate to provide a maximum range of absorbances as the concentration varies. The concentration of an unknown Cu(NO3)2 solution is then determined by measuring its absorbance with the spectrophotometer. By locating the absorbance of the unknown brass solution on the vertical axis of the graph, the corresponding concentration can be found on the horizontal axis (follow the arrows in Figure 6). The concentration of the unknown can also be found using the slope of the Beer’s law curve. Then, you will determine the procedure to prepare standard solutions with known concentrations.

The mass percent of copper in brass can be determined by first reacting it with concentrated nitric acid that will dissolve the zinc and copper metals in the brass. Zinc nitrate solution is colorless, but the copper (II) nitrate has a deep blue color.

The unbalanced ionic equation for the copper reaction is:

Cu(s) + NO3 - (aq) → Cu2+(aq) + NO(g) in an acidic solution

A second reaction occurs when the colorless NO(g) reacts with oxygen in the air to form the observed brown-orange NO2(g) according to the equation:

2NO(g) + O2(g) → 2NO2(g).

The Cu2+ ions in the unknown aqueous solution form the complex ion, [Cu(H2O)6]2+, which causes the blue color. This means that when “white” light (all wavelengths) passes through the solution, the dominant emerging color is blue. A spectrophotometer is used to analyze the color intensity of the copper (II) nitrate solution that forms. For this analysis, it will be necessary to determine which color, with a specific wavelength, will be most strongly absorbed by the copper ions. The concentration of the unknown brass solution will then be determined by comparing its absorbance with that of solutions having known concentrations of Cu(NO3)2.

■Investigation

Procedure

This part of the lab is an application of the concepts the students learned during the Gatorade Lab. You can use any solid brass item, such as brass screws purchased at a hardware store, .22 caliber rifle shells, or brass shot available from a chemical supply company. U.S. pennies could also be used, even though post-1982 pennies are not a brass alloy, but a zinc core encased in a copper shell. Post-1982 pennies have a mass of 2.5 grams and contain 2.5 percent copper and 97.5 percent zinc (pre-1982 pennies are 95 percent copper and 5 percent zinc with a mass of 3.11 grams). If a penny is used, in order to obtain a molarity within the range of the five standards, you should dilute the penny solution to 250 mL using a volumetric flask, instead of the 100 mL flask described in the lab procedure.

1. Measure the mass of a 1–2 gram sample of brass to ± 0.001 g. Place the sample in a small beaker. The brass sample should have a mass of 1–2 grams to ensure that the concentration of the solution falls within a range that can be measured by the spectrophotometer, and that there is an excess of nitric acid available to dissolve it completely.
2. Assuming your brass sample is 100 percent copper by mass, calculate the minimum volume of concentrated 15.8 M HNO3(aq) that needs to be added to react completely with the brass. (The reaction produces a solution of copper (II) nitrate, along with nitrogen monoxide gas and water.) Under the fume hood, have your teacher add approximately 2 mL more than this volume of 15.8 M HNO3(aq) so that the acid is in excess, and then your teacher will cover the beaker with a watch glass. For safety reasons, only you as the teacher should handle the concentrated nitric acid!
3. After the metal dissolves completely, add 50 mL of distilled water to the beaker (again, your teacher will perform this part of the investigation). Then you will remove the beaker from the fume hood and transfer the solution to a 100 mL volumetric flask. Rinse the beaker 3–4 times with 5 mL of distilled water and add the washings to the flask. Dilute to a final volume of 100.0 mL. The excess nitric acid will dissolve the zinc and copper metals in the brass.
4. Obtain 10.0 mL of 0.400 M Cu(NO3)2(aq) stock solution in a 10 mL graduated cylinder. Determine what volume is required to make 10.00 mL of 0.200 M Cu(NO3)2(aq). Use a volumetric pipette to transfer this volume of the stock solution into a clean test tube. Then add a sufficient amount of distilled water to reach 10.00 mL. Thoroughly mix the solution. Repeat the dilution process to make 10.0 mL each of three more additional dilute solutions that are 0.100 M, 0.0500 M, and 0.0250 M, respectively. Have your instructor verify your dilution calculations!
5. Based upon the results of the prelab experimentation, set the wavelength of the SPEC 20 to that which is strongly absorbed by the blue-colored Cu2+ solutions. Ideally, the maximum absorbance value should be ≤ 1. Since absorbance is a logarithmic function of the percent transmittance, the instrument is in a nonlinear region to measure the light passing through when the absorbance value is at a range of 1-2. When selecting a wavelength for measurement, keep in mind that a wavelength at maximum absorbance provides maximum sensitivity, but the smallest concentration range, while a wavelength with a smaller absorbance would provide less sensitivity, but a larger concentration range to be measured in the experiment. Have your teacher approve of your selected wavelength before you continue with the next step.
6. Calibrate the SPEC 20 to read 0% transmittance with no cuvette in the instrument, and100% transmittance with a blank inside (cuvette filled with distilled water).
7. Empty the water from the “blank” cuvette. Using the most dilute Cu(NO3)2 standard solution, rinse the cuvette twice with ~1-mL amounts and then fill it ¾ full. Wipe the outside with a tissue, place it in the SPEC 20, and close the lid. Read and record the absorbance value. Discard the cuvette contents back into the original test tube.
8. Continue testing the other solutions, starting with the most dilute Cu(NO3)2 to the most concentrated (0.400 M). Finally, determine the absorbance value of the unknown Cu(NO3)2 solution from your brass sample. Using the absorbance and concentration values for the five standard solutions, prepare a graph of the absorbance (Y) versus the concentration (X) values. Draw a best-fit straight line for your data and calculate the slope and y-intercept for Beer’s plot. Then determine the concentration of your unknown brass solution.
9. Dispose of all solutions as directed by your teacher. Rinse the cuvettes with distilled water. Return all equipment to your teacher for final approval.

Data Collection and Computation

1. Prepare a data table to record all of your measured data and calculated values.
2. Show the calculations used to prepare the Cu(NO3)2(aq) with known molarities by diluting the 0.400 M Cu(NO3)2(aq) standard solution.
3. Determine the molarity of the Cu(NO3)2(aq) found in 100.0 mL of the brass solution using both the SPEC 20 (or colorimeter) analysis AND the visual comparison method. Support your answers with the appropriate calculations.
4. Determine the mass of Cu dissolved in the brass solution based upon the molarities calculated in Step 3, and use these values to calculate the mass % of Cu in the brass sample using both experimental techniques.

Argumentation and Documentation

Use a shared Google Form spreadsheet to analyze the precision of the class data. Enter your group’s values for the calculated mass percent of copper in brass based upon the colorimeter. Calculate the averages and standard deviations using the compiled data.

■Postlab Assessment



1. Based upon the Beer’s law plot shown below for the absorbance of CuSO4(aq) versus its concentration, *what mass of copper* could be produced by a complete reaction of excess zinc metal with 50.0 mL of a CuSO4(aq) that has a measured absorbance of 0.685?
2. In 2011, a Tsunami in Japan crippled a nuclear power plant complex and caused a release of radioactive iodine across the country. As a precautionary measure against radiation exposure, the Japanese distributed 230,000 units of potassium iodide tablets to evacuation centers in the area around the nuclear power plant. Potassium iodide has been shown to protect the thyroid gland from the radioactive form of iodine released by nuclear accidents or emergencies that could lead to thyroid cancer. (Thyroid cancer ended up being the biggest negative health impact caused by the 1986 Chernobyl nuclear reactor disaster.) Using a spectrophotometer to create a Beer’s law plot on can determine the mass percent of potassium iodide present in tablets that are readily available in the United States through vitamin and drugstores, or online. Once the molarity of the pill solution is determined based upon the color intensity, then the mass of KI and its mass percent in the pill can be calculated. The dosage value can be used to determine the accuracy of the experimental procedure. (Pills are available in 130- and 65-milligram doses, and smaller 30-mg doses that are given to children.)

One pill should be dissolved in distilled water in a volumetric flask to make 100.0 mL of solution. Then the iodide ions can be reacted with KNO2(aq) and 6 M HCl(aq) to oxidize the colorless I–(aq) to a yellow–orange I2(aq). The net ionic equation for this reaction is:

2I- (aq) + 2NO2- (aq) + 4H+ (aq) → I2(aq) + 2NO(aq) + 2H2O(aq)

*Describe how the procedure for determining the percent of copper in brass could be modified to measure the mass percent of KI in commercially available tablets found in US vitamin or drugstores.*