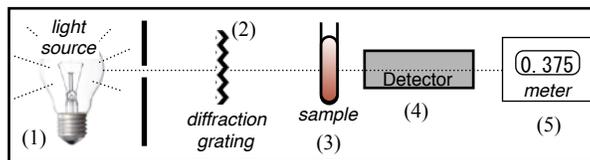


The Spectrophotometer

Any solution that is colored absorbs light at a particular wavelength of visible light. An instrument that is used to measure the absorbance of light by a sample is called a spectrophotometer. A general schematic diagram for a spectrophotometer is shown to the right.



The process begins (1) with a light source or a light bulb. For the instrument that is used in this lab, the light bulb emits light of wavelengths ranging from 340 nm to 750 nm. The light then travels to a monochromator (diffraction grating) (2). This device as the name indicates, separates the light into its individual wavelengths so that light of only a single particular wavelength shines towards the sample. Next the light of a particular wavelength and intensity, is passed through the sample (3). As the light passes through the sample, some of the light may be absorbed. This absorption lowers the intensity of the light. The detector (4) measures this decreased intensity. The readout of the spectrophotometer is called absorbance, which is a measure of the amount of light absorbed. In this lab, we will measure absorbance to determine the amount of copper ions in solution. At absorbance measurements between ~1.5 – 0.50, absorbance is directly proportional to the molar concentration of the substance in a solution that is absorbing the light. This linear relationship is called Beer's law:

$$A = \epsilon bc$$

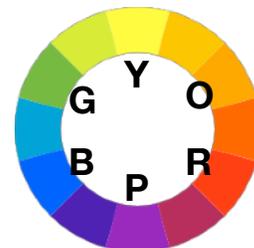
- In this law the term "A" is the absorption of light by the sample.
- The term " ϵ " is molar absorptivity which is a constant for a substance at each particular wavelength.
- The term "b" is the cell path. This is the width of the test tube (aka cuvette) and will be held constant throughout the experiment.
- The term "c" is the molarity of the substance absorbing the light.

Pre PreLab

- Let's go to the Phet simulation about Beer's Law (you can use the url below or find a direct link on the homework page):
- http://phet.colorado.edu/sims/html/beers-law-lab/latest/beers-law-lab_en.html
- Choose the Beer's Law simulation, NOT the Concentration simulation.
- Setup: Click the red button to turn the light source on and click the Variable wavelength button to show choices in colors of light. On the light sensor to the right, choose Absorbance (not Transmittance). Absorbance just what it sounds like – a measure of the amount of light absorbed by the solution.

Answer the following questions below.

1. To do/notice: Click on the yellow arrow to vary the width of the solution container (the length that the light travels through solution is called *path length*). Write a correlation statement comparing the amount of light absorbed as related to path length?
2. Use the blue slider at the bottom to vary the concentration of the solution. Write an correlation statement comparing the solution concentration as related to the absorbance of light as it passes through the sample.
3. Click the "variable" button under the light source, then slide the tab below the rainbow spectrum to change the color of light that you are sending through the solution. For the red drink mix, what color of light absorbs the most light? What color is least absorbed by the solution?
4. Change the solution to a different color solution and determine which color of light is most absorbed by each solution, and what color of light is least absorbed. Try this for at least two different solutions.
5. Considering the color wheel to the right, write a brief comment about what you may have noticed from questions #3 & #4?



Introduction

Brass is a mixture made of copper and zinc. In this lab we will determine the percentage of copper in a sample of brass using analysis with a spectrophotometer.

The Reaction

Up until now, you may have learned that copper will not react with acid, however, there is more to nitric acid than just free floating H^+ ions. The balanced (molecular) equation for the reaction that occurs in this lab is represented below. Apply oxidation numbers to each element on both sides of the equation.



6. Which *element* is oxidized, and which *element* is reduced? How can you tell? (2)

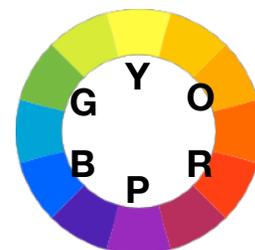
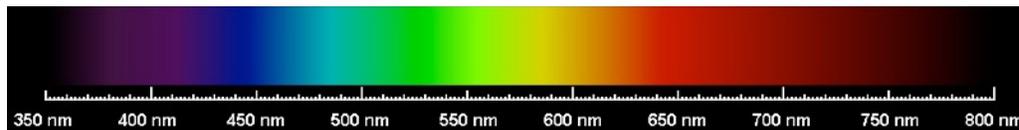
Procedure Overview

As you may know, Cu^{2+} ions in solution are blue, and the intensity of the blue solution, it is an indication of the concentration of Cu^{2+} ions. First we must measure the absorbance of several solutions of known concentrations, from we will prepare by careful dilution of a stock solution. The absorbances of those known solutions will be measured and a graph of concentration vs absorbance will be constructed. The copper in the brass sample that was reacted into solution will be diluted to a particular volume, and then the absorbance of that brass sample solution will be measured. Using the equation of the line from the graph of concentration of the known solutions vs absorbance, and the measured absorbance of the brass sample solution, will allow the determination of the concentration of copper ions in the brass sample solution. Knowing the concentration and the brass sample's volume will allow you to back calculate into the percentage of copper in the brass.

PreLab – Please show computational work clearly.

7. Concentrated nitric acid has a molarity of 15.8 M. Determine the volume of nitric acid required to completely react with 1.00 g of copper. (2)
8. Calculate the mass of copper that would be present in 250.0 ml of solution that has a 0.0375 M of Cu^{2+} ions. (2)
9. The solution in the previous question was produced from a 1.00 g sample of brass. Calculate the mass percentage of copper in the brass. (1)

Observe the continuous spectrum and a complementary color wheel to help understand color and wavelengths as well as complementary colors as we proceed through the remainder of the PreLab on the next pages.



Lab B1 (pg 3 of 6) **Spectrophotometer: % Cu in Brass**

10. What color is cobalt(II) chloride solution? (1)

- What color (and approximate wavelengths) of light do you suspect a cobalt(II) chloride solution most strongly absorb?
- Sketch the graph of the absorption spectrum.

11. What color is potassium permanganate solution? (1)

- What wavelengths would you expect this solution to most strongly absorb?
- On the same permanganate graph sketch the spectrum of second sample of permanganate and explain on the graph why it looks the way that it does.

12. What color is the zinc nitrate solution? (1)

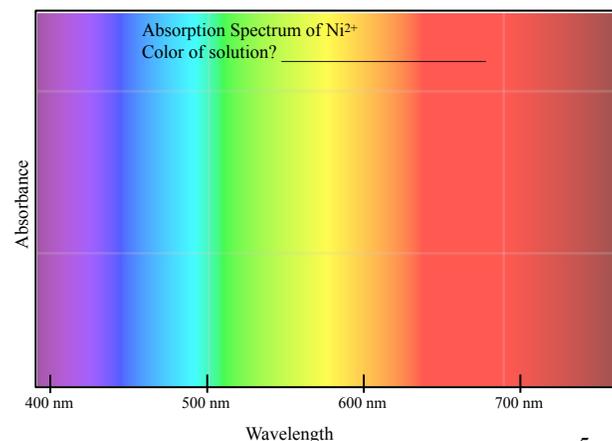
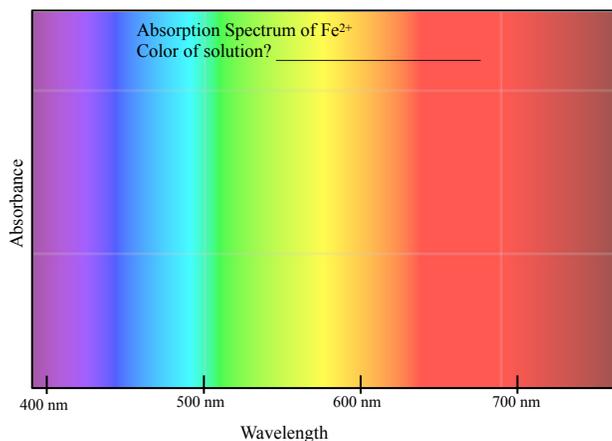
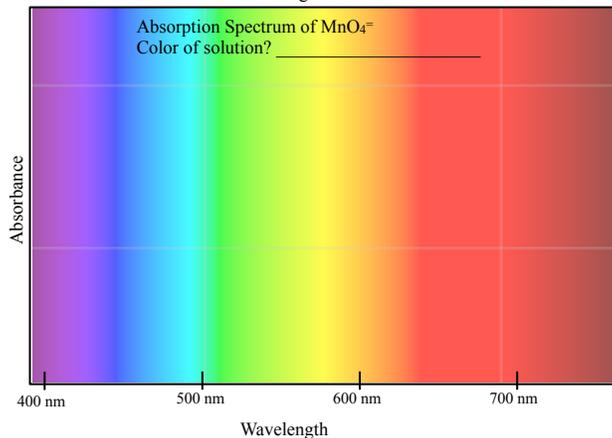
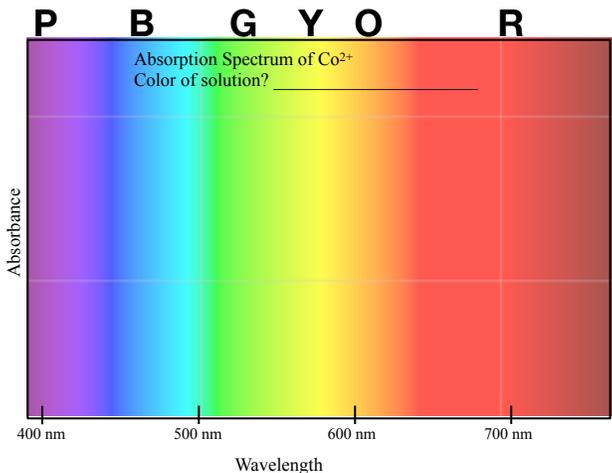
- What wavelengths within the visible spectrum would zinc nitrate absorb?
- This could be problematic for an experiment like this when testing for the concentration of zinc ions. Explain.

13. What color is iron(II) nitrate solution? (1)

- What color (and approximate wavelengths) of light will iron(II) nitrate most strongly absorb?
- Sketch the graph of the absorption spectrum.

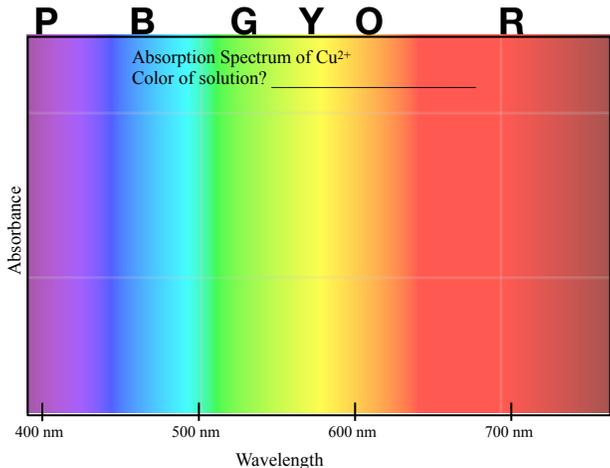
14. What color is nickel(II) nitrate solution? (1)

- What color (and approximate wavelengths) of light will nickel(II) nitrate most strongly absorb?
- Sketch the graph of the absorption spectrum.



Lab B1 (pg 4 of 6) **Spectrophotometer: % Cu in Brass**

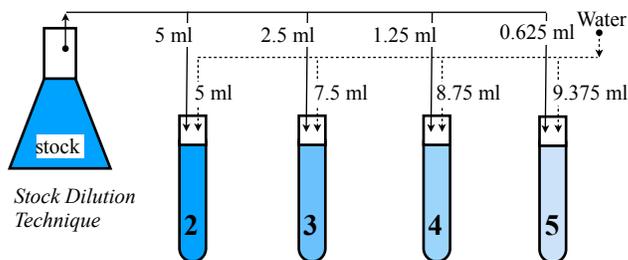
15. It is important to determine the wavelength that we will need to measure the absorbances for the Cu^{2+} ions that we will be testing.
- What color is a copper(II) solution?
 - What wavelength will you expect Cu^{2+} ions' maximum absorbance?
 - Make a sketch of the absorption spectrum of copper(II) ions.
16. What would be the optimal wavelength setting to measure absorbances for the Cu^{2+} solutions in this lab? How can you tell from the graph?



17. Your team will dilute a copper(II) nitrate solution to various concentrations. We will use the *Stock Dilution Technique*. Calculate the concentration that will result from the dilutions as demonstrated, using the formula and showing your work to the right:

$$M_{\text{stock}} V_{\text{stock}} = M_{\text{diluted}} V_{\text{diluted}}$$

trial #	Measured Absorbance	Concentration (M)
A1 (stock sol'n)		0.10
B1 (stock sol'n)		0.075
A2		
B2		
A3		
B3		
A4		
B4		
A5		
B5		
brass solution		??
mass of brass sample (g)		



Materials (per lab group)

- rack for cuvet and beaker to mix dilution
- 5 ml pipet for stock solution
- flask with stock
- 10 ml pipet for water
- beaker with water
- pipet pumps or bulbs

Procedure *Eyewear is not optional, it's a must..*

- Measure the mass of the brass sample, label a 50 ml beaker, then go to the hood and just cover the sample with 15.8 M nitric acid. Leave the beaker in the hood until all of the brass has reacted and the poisonous colored gas has evolved.
- Dilute the reacted brass solution by pouring all of the solution into a 250 ml volumetric flask and diluting to the mark.
- First PRACTICE with the pipets and pumps using the stock solution in the 5 ml pipet and practice with the water in the 10 ml pipet. Look carefully at the markings on the pipet so that you are clear on how to accurately measure the appropriate amount of solution.
- Prepare the known concentration solutions that you have been assigned using the "Stock Dilution Technique."
- Pour (do NOT pipete) in enough of prepared dilution solution into a cuvette just three quarters full.
- The brass sample solution will be put into a cuvette. Record all the measured absorbances.

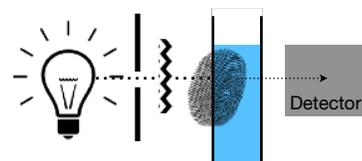
Lab B1 (pg 5 of 6) Spectrophotometer: % Cu in Brass

Process the Data (on your Google Lab Sheet)

1. Make a graph of known concentration and absorbance data for the stock and dilutions.
 - a. Graph concentration vs absorbance. (concentration on the y-axis and absorbance on the x-axis)
 - b. Put in a trend line and display the equation for the line.
2. Use the trendline equation to calculate the concentration of the brass sample solution.
3. From the calculated concentration of the brass solution and the volume of your brass solution, calculate the moles and mass of copper in the solution.
4. Calculate the mass percent of copper in our brass sample.

Post Lab Questions

1. Why was it important not to touch the cuvette with your fingers? Would this problem cause the percentage of copper in brass to be larger, smaller, or no effect? Justify.
(Be sure and start your justification by first commenting on any effect on the absorbance.)

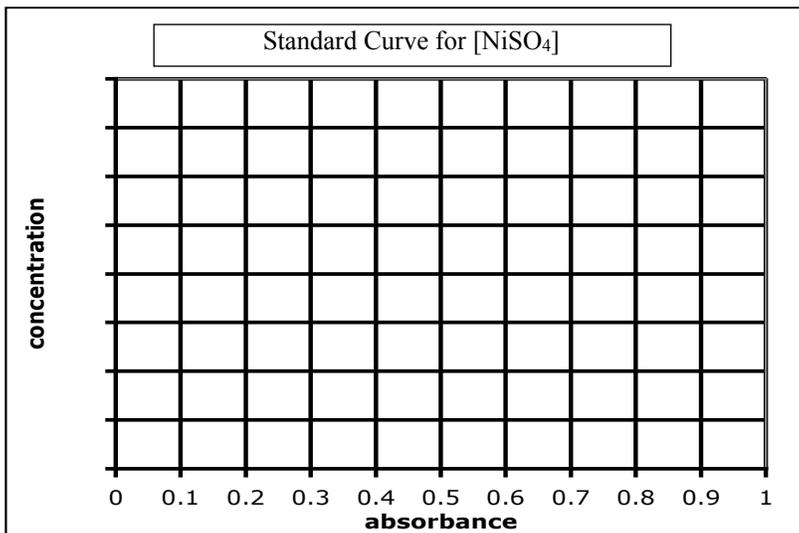


2. While making up his solution from the brass shell, Ferdinand topped off the volumetric flask to the 250 ml line and then immediately poured some solution into the cuvette for analysis **without** inverting and mixing the solution in the flask. Would this cause the percentage of copper in brass to be larger, smaller, or no effect? Justify.
(Be sure and start your justification by first commenting on any effect on the absorbance.)
3. If you were to do this same lab but testing for the percentage of nickel in 5 cent pieces (nickels). How would you need to adjust the procedure with respect to testing for nickel instead of copper? Refer back to the spectral information in the prelab.

Lab B1 (pg 6 of 6) **Spectrophotometer: % Cu in Brass**

5. Calvin was given the task of determining the molar concentration for a sample of nickel(II) sulfate solution of “unknown” concentration. In order to establish a standard or calibration curve, the student prepares a set of five standard solutions of nickel(II) sulfate. The student collects the data shown in the following table. Use the below to sketch a graph of the data.

#	Concentration (mol/L)	Absorbance
1	0.0008	0.205
2	0.0016	0.404
3	0.0024	0.599
4	0.0032	0.789
5	0.0040	0.982



- Hobbs then measured the absorbance for the unknown sample as 0.669
- Determine the concentration of the unknown NiSO₄ solution directly from the graph and record it on the line below. **Clearly mark the graph to indicate how you determined the concentration.**
- _____

6. Minnie measured the absorbance of a 0.350 M solution of cobalt(II) nitrate as 0.728 in a cuvette with a 1.0 cm width.

- Use Beers Law to determine the concentration of another cobalt(II) nitrate solution that had an absorbance of 0.539

7. Later, Mickey measured the absorbance of the 0.350 M solution of cobalt(II) nitrate again in a different cuvette and this time found the absorbance to be 0.473

a. Mickey noticed that the cuvette seemed to be a different size. Do you suppose the cuvette was larger or smaller than Minnie’s cuvette? Justify.

b. Calculate the width of Mickey’s cuvette.

Scoring Rubric	
5	PRE
35	Questions
35	Data Table
<i>Out of 75</i>	
25	Lab Quiz