USING THE SPECTROPHOTOMETER TO ANALYZE A MIXTURE
INTRODUCTION

Description
This experiment is designed to introduce students to the spectrophotometer and the decisions that must be made in using it as a means of gaining information about a system. Because students need to learn the skills, this experiment is very guided in design.

Goals for This Experiment
Students should leave this experiment understanding the following:
1. the relationships between absorbance and wavelength,
2. the relationships between absorbance and concentration,
3. the uniqueness of light absorbance by ions in solution,
4. the ways in which the presence of more than one colored ion can affect the absorbance values obtained by the mixture in solution,
5. the kinds of questions and decisions that must be made in collecting and making sense of data collected by a spectrophotometer, and
6. the way in which experimental data is used as *evidence* to support or refute a question.

Students will also learn to use a Spectronic 20 correctly and have another round of practice using a buret and pipets for measuring volumes.

Recommended Placement in the Curriculum
In our curriculum, we implemented the lab "Using the Spectrometer to Analyze a Mixture" about midway through the first semester General Chemistry course. At this point, students will have had experience pipetting and using a buret.

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PURPOSE
The purpose of this experiment is to determine the degree of usefulness of spectroscopic techniques as a means of analyzing a mixture of cobalt (II) and nickel (II) ions.

BACKGROUND
Chemicals that are toxic have the potential to cause injury when they come into contact with or gain entry into the body. The mechanism for the toxicity varies from interfering with a single function within the body to affecting multiple functions. There are Federal, state, and sometimes local laws requiring the labeling of chemicals to indicate the nature and degree of hazard, including toxicity. In particular, chemicals cannot be transported without packaging that is appropriate to the nature of the chemicals. There are additional regulations regarding the disposal of toxic and hazardous wastes. The options for toxic or hazardous waste disposal include one or more of the following: incineration, physical treatment such as encapsulation into cement blocks, chemical treatment to convert the materials into less harmful products, and storage in a secure landfill. Chemical treatments include acid/base neutralization, oxidation or reduction, and precipitation of toxic ions as insoluble salts. These treatments transform the ions into a material less likely to gain entry into or be absorbed by the body.

SCENARIO
A Chemistry Department has been collecting solutions of Co^{2+} and Ni^{2+} left over from student experiments. It is both illegal and irresponsible to put such solutions down the drain because of the toxicity of these metal ions. The solutions have been stored in a large carboy for several years, and at this point, no one knows the concentration of these two ions in the solution. The Chemistry Department must pay to have the solution disposed of, but in order to do so, they must know the amount of these metal ions in the waste. Also, the cost of disposal depends on the mass (and volume) of the waste as well as toxicity; thus the department is investigating the options listed in the previous paragraph in hopes of reducing the cost of disposal.

The first step is to determine the concentration of the Co (II) and Ni (II) ions in the solution. Because each of these ions produces a colored solution, one potential method for the determination of their concentrations involves use of a spectrophotometer. A solution containing a colored substance absorbs visible light in a predictable and measurable amount that allows one to determine the concentration of the colored substance in the solution. If a solution contains several colored substances, the colored components may interact in such a way that the light-absorbing property of the mixture is not a simple sum of the properties of the individual components. However, for some mixtures, there is little or no interfering interaction and a quantitative measure of the concentrations of the individual components can be made.

YOUR TASK

Your task in this experiment is to determine whether or not a spectrophotometric method can be used to provide the concentrations of the Co^{2+} and Ni^{2+} in the waste solution.

You will have available solutions containing:

- a known concentration of cobalt (II) ion,
- a known concentration of nickel (II) ion, and
- a mixture of the cobalt and nickel ions, for which the individual concentrations are the same as in the separate solutions.

The following procedure is the accepted method for doing analyses with a spectrophotometer. You should work in groups of three, *having each member of the team responsible for collecting information for one of the solutions.*

CAUTION: The ions you will use are toxic. Be careful not to get them on your skin. Should you get some on your skin, wash the area with large amounts of water. If you should spill any, please ask your instructor to clean up the mess. (You may watch.) **Do not** dispose of any of these solutions down the drain. Labeled waste containers will be provided for you to use.

INFORMATION CONCERNING THE SPECTROPHOTOMETER:

A spectrophotometer is used to make absorbance measurements. Light passes into a monochromator where only the desired wavelength, or a very narrow range of wavelengths, can pass through. From there, light passes through the sample, and on to a phototube where the light energy is converted to an electrical current that is registered on a meter. The instruments we will use are capable of quite precise measurements, so it will be essential that solutions are made carefully and the directions for making the measurements are followed precisely. You need to be sure that you use the <u>same</u> instrument for all of your work. Directions for using the spectrophotometers are included in the "Real-World Projects: Challenges from the Polymer Industry" scenario.

PROCEDURES AND ANALYSES:

A. Absorbance as a Function of Wavelength for a Colored Solution

- Use a solution of known concentration.
    - ⇌ Record the identity and concentration data.
- Fill one cuvette about 40% full with the sample solution. Fill a second cuvette with the same amount of distilled water to be used as a blank.

Note: Cuvettes look like regular test tubes, but they are <u>not</u>. The tubes have been carefully and precisely made to have a composition that is transparent to light of the visible range and a perfectly round shape that is a constant 1 cm diameter. Cuvettes must be handled with care; be sure to touch them <u>only near the top</u>, and to wipe off the outside with a piece of lintless paper, such as Kimwipes, <u>every time</u> before you put them into the instrument. To prevent scratching, cuvettes should be stored in test tube racks when not in use. Students who break cuvettes will be

responsible for the cost of replacing them, so **be careful!**

Preparation of an absorbance spectrum
1. Record the absorbance and/or the percent transmittance of your solution for every 20 nm throughout the range 370–600 nm.
2. Review your results. In the range of greatest absorbance, repeat the scanning every 5 nm.
3. Determine the optimum wavelength for use with each of the solutions containing a single ion.
    ☞ Compare the colors and optimum wavelengths of these solutions.

Q 1: What relationship exists between the *wavelength of light absorbed* by a solution of a particular color and the *color you see* for that solution?

   ☞ Compare the data for the mixture with the data for the single solutions.

Q 2: What relationship appears to exist between the spectrum for the mixture and the spectra for the individual ion solutions? NOTE: you may want to graph the data for the three solutions to decide the answer for this question.

Q 3: For this particular pair of ions in the mixture, is spectrophotometry a useful method of analysis? Support your choice with evidence.

You can test your decision by the procedure in Part B, which you will carry out on **the solutions containing only one colored ion.**

B. Absorbance as a Function of Concentration for a Single Component Solution
1. Label 4 clean, dry containers that will hold at least 10 mL of solution.

2. Make the dilutions listed in the table below from your stock solution. Use pipets to measure the stock solution and a buret to measure water. As each solution is made, stir it well with a clean, dry stirring rod to ensure thorough mixing.

SOLUTIONS FOR STUDY

solution label	volume stock solution (mL)	volume water (mL)
A	2.0	3.0
B	1.0	4.0
C	1.0	9.0
D	5.0 mL of solution # C	5.0

3. Set the wavelength on the spectrophotometer to the wavelength that you have chosen as the optimum absorbance for your solution.

4. Using water as a blank, determine the absorbance of the <u>four dilutions</u>.

C. Identification of the Concentration of a Solution
<u>Each</u> student should obtain an unknown sample of the solution of the mixture. Measure the absorbance of the sample at each of the two wavelengths chosen as optimum.

D. Data Manipulation and Analysis
1. Calculate the concentrations of the diluted sample.
   (vol conc • M conc) = (vol dil • M dil)
2. Prepare a graph with concentration on the x-axis, absorbance on the y-axis.
3. Draw the "best straight line" through the data points.
4. Use the graphs to determine the concentration of each of the components in the unknown sample.

Once you have your calculations completed, ask your instructor for the actual concentrations of ions in your mixture. Compare your concentration values for the ions of the mixture with the concentrations actually prepared. Compare your pattern of results with that of the others in your group.

E. Now Think about Your Original Task
Q 4: Does the data you have collected confirm or refute the decision you made earlier about the feasibility of using the spectrophotometric method to analyze this mixture?

⇒ Write a short summary statement defending your conclusion, including the reasoning and observable evidence you used to come to your decision.

F. Back to the Department's Disposal Problem
Once the concentration of each of the toxic metal ions is known, their amount can be calculated from the concentration and volume of the solution. This information will aid the Chemistry Department in the determination of an appropriate method for disposal. The idea here is to convert the metal ions into a form which is either 1) less toxic so it is no longer considered a hazard and/or 2) less likely to be leeched into the environment surrounding the disposal site.

⇒ Based on the information provided in the introduction and your prior experience in this laboratory class, suggest a potential method for such a conversion. You do not need to provide extensive details, just a brief description of the technique which you think would be worth further investigation.

References

Understanding Chemical Hazards: A Guide for Students; Task Force on Occupational Health and Safety, Department on Government Relations and Science Policy, American Chemical Society, 1155 Sixteenth Street, NW, Washington, DC, 20036; 1995, p 22.

Lewis, Sr., Richard J, Editor. *Rapid Guide to Hazardous Chemicals in the Workplace; 3rd Edition;* Van Nostrand Reinhold, New York, 1993.

Armour, M.A. *Hazardous Laboratory Chemicals, Disposal Guide*, CRC Press, Boca Raton, 1991.

Waste Disposal in Academic Institutions, James A. Kaufman, Editor, Lewis Publishers, 1990.

Time Required
The approximate time required to complete this activity is 3 hours.

Group Size
Students can work in groups of three.

Materials Needed
per group:
- spectrophotometer
- 4 cuvettes
- box of Kimwipes
- 1-, 2-, and 5-mL pipets with bulbs
- burets, ring stands, clamps, etc.

per class:
- 250 mL of stock solution of 0.20 M Ni^{2+} from $Ni(NO_3)_2 \cdot 6H_2O$
- 250 mL of stock solution of 0.15 M Co^{2+} from $Co(NO_3)_2 \cdot 6H_2O$
- 250 mL of a mixture that is 0.20 M in Ni^{2+} and 0.15 M in Co^{2+}
- 1 unknown solution per group to use in Part C

Safety, Disposal, and Special Handling
Review the Material Safety Data Sheets of any chemical used in the experiment for information regarding safety and handling. Dispose of waste according to your local ordinances.

Pre-Lab Discussion
1. Before students go into the lab, you should make sure they understand the nature of the task they are given. It will be very easy for them to get so caught up in the procedure that they fail to think about what the experiment means. Ask several questions about what is going on and what the data means as it unfolds.

2. Show students the cuvettes and the Kimwipes. Remind them how different a cuvette is from a test tube and how cuvettes should be handled to prevent fingerprints and scratches.

3. I find it easier to show students how to work the Spec 20 while they are in the lab working with their own instrument as I repeat the directions. Encourage students to use the directions and for groups to watch and critique each other as they work so that the proper steps are taken. Students often let one person do all of the thinking unless they are specifically told to do some of their own.

4. Remind students about heavy metal ion solutions. Give directions for disposal of the solutions. Remind them to wash their hands with soap before leaving lab.

Likely Play-Out of Lab

Each student will have a solution for which he/she is responsible. The point of having students work in groups of three is so they can see, wavelength by wavelength, the uniqueness of absorbance by the different solutions. You need to be alert to each group to make sure they are following the procedure necessary to get good data.

During the dilution phase, students need to be careful in making their dilutions so their concentration/absorbance graph makes sense. Be sure to have students include the value for the stock solution as one of their points. You might remind them to put this information in the table they make for recording these data. This will give students additional practice with measuring from pipets and burets. The stock solution should be measured from pipets and the water from a buret. Have students watch and check each other's technique.

Data Analysis

For Part A

Students should see the difference in optimum wavelength for the different solutions. Each of the single component solutions has one good peak and the mixture has two, one corresponding to each of the single components.

For Part D

1. Watch that students get the right <u>initial</u> concentration for point D. Many will forget that the value is what they calculate for point C.

2. & 3. Their graph should
   • NOT show the points joined dot-to-dot,
   • include the most concentrated solution value, and
   • be reasonably linear.

4. The students should use their graphs to determine the concentration of each of the ions in the mixture. They will have problems with this part if they have not realized that the point for the mixture is the <u>sum</u> of the values for each of the ions at that particular wavelength.

For Part E

One would expect the final response to suggest that the ions be precipitated from solution; the supernatant can be checked for absence of these ions.

Possible Answers to Questions

1. **What relationship exists between the *wavelength of light absorbed* by a solution of a particular color and the *color you see* for that solution?**
   The wavelength of absorbed light is the complement of the color we see. Students will probably not know this unless a chart of light/wavelength absorbed is given to them.

2. **What relationship appears to exist between the spectrum for the mixture and the spectra for the individual ion solutions?**
   The students should see the two peaks and notice that the minimum values for the mixture are never as low as those for either of the single solutions. They may or may not realize the additive nature of the value at each particular wavelength.

3. **For this particular pair of ions in the mixture, is spectrophotometry a useful method of analysis? Support your choice with evidence.**
   Students may answer yes or no to this question. It is important that their reason be a reasonable justification. Either way they answer, they can test this assertion by doing the final part of the experiment.

4. **Does the data you have collected confirm or refute the decision you made earlier about the feasibility of using the spectrophotometric method to analyze this mixture?**
   Students' answers here depend on what they said before and how well they understand what the data meant. It is known that cobalt and nickel ions do not interact in solution. Thus, a mixture of the two should show a simple additivity of absorbencies. The students' response should include some explanation of why the answer was chosen and a listing of the actual data that supports his/her position.