

Spectrophotometric Analysis of a Two-Component System with Overlapping Spectra by Walter Rohr

A number of methods have been developed to determine the composition of a binary mixture spectrophotometrically. Most of these are directed at mixtures where one component can be isolated from the other or they require a Beer's law experiment to measure the molar absorptivity of each of the substances in the mixture. However, Blanco¹, et. al. described a method of resolving mixtures with overlapping spectra, called Multi-Wavelength Linear Regression Analysis or MLRA, without determining molar absorptivities or complicated mathematics. Using Blanco's method, the composition of a binary mixture with overlapping spectra can be resolved with only three measurements, the absorbance of a standard solution for each component, and the unknown mixture itself. Vernier's Logger *Pro* software is ideally suited for this experiment with the ease at which one can manipulate data and its ability to prepare a graph even when the data are out of order.

Here's how MLRA works: Assuming additivity, the absorbance of a mixture is the sum of the absorbances of its components. If we have a mixture consisting of two components, #1 and #2, with an unknown concentration of #1x and #2x, then:

$$\text{Absorbance of the unknown mixture, } A(\text{mixture}) = A(\text{\#1x}) + A(\text{\#2x})$$

$$\text{but applying Beer's law: } A(\text{\#1x}) = \epsilon(\text{\#1}) \cdot b \cdot C(\text{\#1x}) \text{ and } A(\text{\#2x}) = \epsilon(\text{\#2}) \cdot b \cdot C(\text{\#2x})$$

$$\text{Substituting: } A(\text{mixture}) = \epsilon(\text{\#1}) \cdot b \cdot C(\text{\#1x}) + \epsilon(\text{\#2}) \cdot b \cdot C(\text{\#2x})$$

However, the absorbances of standard solutions of the same substances will follow the same Beer's law relationship and have the same molar absorbance, ϵ , and one centimeter path length, b , as the unknown solutions under the same conditions. Therefore, we can write:

$$A(\text{\#1s}) = \epsilon(\text{\#1}) \cdot b \cdot C(\text{\#1s}) \text{ and } A(\text{\#2s}) = \epsilon(\text{\#2}) \cdot b \cdot C(\text{\#2s})$$

$$\text{Rearranging these relationships: } \epsilon(\text{\#1}) \cdot b = \frac{A(\text{\#1s})}{C(\text{\#1s})} \text{ and } \epsilon(\text{\#2}) \cdot b = \frac{A(\text{\#2s})}{C(\text{\#2s})}$$

$$\text{Substituting: } A(\text{mixture}) = \frac{A(\text{\#1s})}{C(\text{\#1s})} \cdot C(\text{\#1x}) + \frac{A(\text{\#2s})}{C(\text{\#2s})} \cdot C(\text{\#2x})$$

$$\text{or: } A(\text{mixture}) = \frac{C(\text{\#1x})}{C(\text{\#1s})} \cdot A(\text{\#1s}) + \frac{C(\text{\#2x})}{C(\text{\#2s})} \cdot A(\text{\#2s})$$

Dividing through by $A(\#1s)$ and simplifying we obtain:

$$\frac{A(\text{mixture})}{A(\#1s)} = \left(\frac{C(\#1x)}{C(\#1s)} \right) + \left(\frac{C(\#2x)}{C(\#2s)} \right) \cdot \left(\frac{A(\#2s)}{A(\#1s)} \right)$$

Since the relationship follows the form: $y = mx + b$, then a plot of $\frac{A(\text{mixture})}{A(\#1s)}$ versus

$\frac{A(\#2s)}{A(\#1s)}$ will give a line with a slope of $\frac{C(\#2x)}{C(\#2s)}$ and an intercept of $\frac{C(\#1x)}{C(\#1s)}$.

That is, the concentration of the unknown component in the mixture $\#2x$, equals the slope times the concentration of the standard solution for component $\#2$. Likewise, the concentration of the unknown component in the mixture $\#1x$, equals the intercept times the concentration of the standard solution for component $\#1$, or simply $C(\#2x) = m \cdot C(\#2s)$ and $C(\#1x) = b \cdot C(\#1s)$

Let's apply this method to the analysis of the metals in a United States five-cent coin.

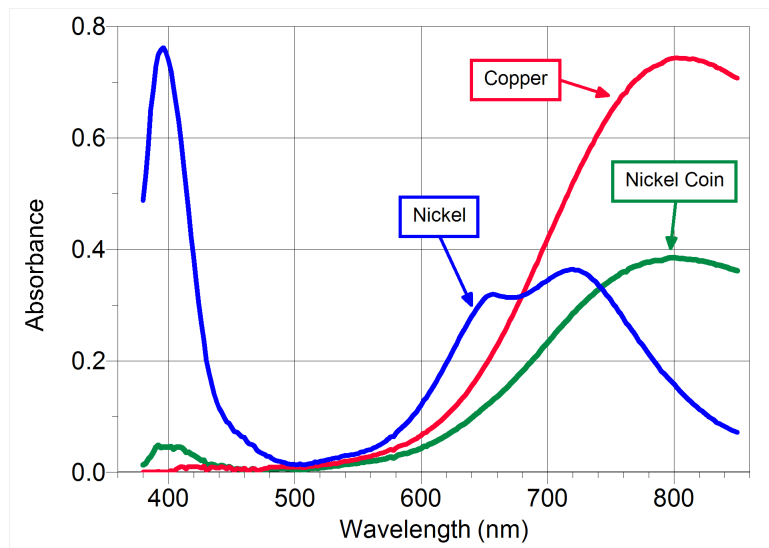


Figure 1 - Absorbance spectra of nickel, copper and five-cent coin

To this end, the absorption spectra of the standard solutions with known concentrations and that of the coin are obtained as three overlapping curves on a single graph. (See Figure 1.)

In order to apply this method, data columns for $\frac{A(\text{mixture})}{A(\#1s)}$ and $\frac{A(\#2s)}{A(\#1s)}$ will be created and plotted where the vertical axis becomes the absorbance of the mixture divided by the absorbance of one of the components, and the horizontal axis becomes the ratio of

absorbances of the two standard solutions. The divisor is constant in both calculations, and should be the standard substance with the maximum absorbance, lambda max, closest to the optimum absorbance² of 0.434. In this example, nickel was chosen as the divisor. Figure 2 is an example of the experiment at the end of the process.

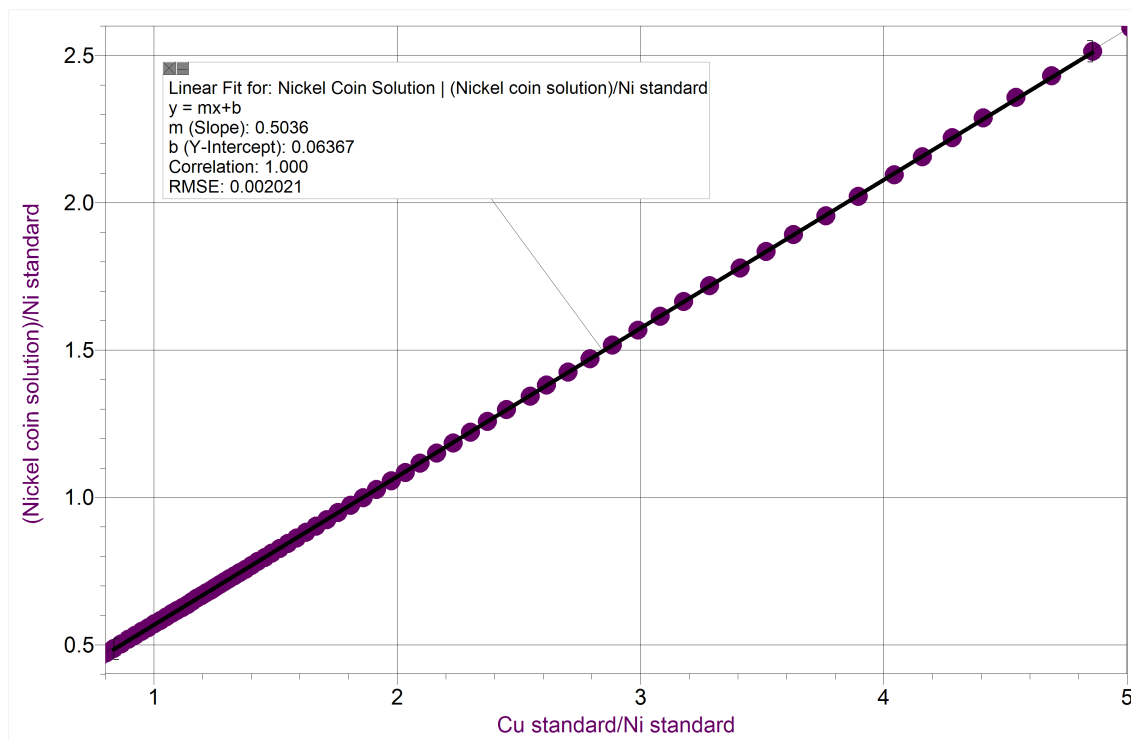


Figure 2 - Linear regression analysis of absorbance ratios

Procedure

1. The components of an unknown mixture are identified by qualitative analysis, chromatography, or another method.
2. A solution of the unknown is prepared and diluted quantitatively until the maximum absorbance is in the optimum 0.15 to 0.70 range (20% to 70% transmission). Likewise, standard solutions of each component are prepared having absorbances in the optimum 0.15 to 0.7 range.
3. The spectrophotometer, calibrated using a distilled water blank, is used to measure the absorbance vs. wavelength for each of the three solutions prepared above.
4. Now that the absorbance of each solution has been measured, remove from consideration absorbance values which are the most unreliable (those at the extreme ends of the spectra or those outside the optimum 0.15 - 0.70 absorbance range³). This is achieved by highlighting the number of the column on the far left of the data table and scrolling through the unreliable data. All three data sets will be selected by this process even though the data in only one column is suspect.

The data are then eliminated from consideration by choosing Strike Through Data Cells from the Edit menu. The selected data in those data sets will no longer be graphed or figured in when calculating the absorbance ratios. Continue this process of highlighting data and striking through unwanted data until the entire table has been processed.

5. The next step in the process is to calculate the absorbance ratios which are the key to this method. Using the Vernier Logger *Pro* software, choose New Calculated Column from the Data menu. Type in your description of the calculation that will also be used as the label for the vertical axis. In this example, Absorbance(coin))/ Absorbance(Ni) was entered. Next, enter a short name for your data column. "Coin/Ni" was entered in this example. Note: You can uncheck the box which states, "Add to Similar Data Sets."
6. Enter the equation for the absorbance of the mixture divided by the absorbance of one of the standards. This is accomplished by clicking the "Variables (Columns)" menu, followed by "Choose specific column." Next, check the "Nickel coin solution | Absorbance," followed by "OK." The "Nickel coin solution | Absorbance" should appear in the equation box. Type in a "/", the symbol for divide. Finally, check the "Variables (Columns)" option again, and select "0.166M Ni | Absorbance," then "OK." The equation will now read, "Five-cent coin solution | Absorbance"/ "0.166 M Ni | Absorbance." Click "Done." (Note: In this example, the absorbance of the standard nickel solution is used as the divisor.)
7. This process is repeated for the second calculated column, the ratio of the absorbance of the 0.0595 M Cu solution to the absorbance of the 0.0166 M nickel standard. The absorbance of the nickel solution being chosen as the standard will again be the divisor.
8. Once the absorbance ratios are calculated, they must then be plotted. To select the data to be plotted on the y-axis, click on the vertical axis, select more, then check the absorbance(nickel coin solution)/ absorbance(standard nickel). Uncheck all other data sets.
9. To select the data to be plotted on the x-axis, click on the horizontal axis checking absorbance(Cu standard)/ absorbance(Ni standard). Autoscale the graph.
10. Click on the Linear Fit icon, . The best linear fit will appear for the data selected (See Figure 2).
11. Calculate the concentration of the components of the mixture using the slope and intercept of the line of best fit found in step 10. Use this information to determine the percentage composition of the metals in a U.S. five-cent coin based on the MLRA.

Our Results

In our trial, 0.488 grams of nickel wire was dissolved in 15 mL of 1:1 nitric acid and diluted to 50 mL. The copper standard solution consisted of 0.189 grams of copper dissolved in 5 mL of the 1:1 nitric acid solution and again, diluted to 50 mL. The U.S. five cent coin solution was prepared by dissolving 0.125 grams of a coin in 5 mL of the same nitric acid solution and diluted to 50 mL. The resulting standard solutions were 0.166 M Ni^{2+} (aq) and 0.0595 M Cu^{2+} (aq). The best fit for this data gave a slope of 0.5021 and intercept of 0.06590.

Using the slope, we found the concentration of copper in the coin solution to be $0.5021 \cdot 0.0595 \text{ M} = 0.0298 \text{ M Cu}$. Since we dissolved 0.125 g of the coin in nitric acid and diluted to 50 mL, the number of moles of copper in the solution equals $0.0298 \text{ M Cu} \cdot 0.05 \text{ liter} = 0.00149 \text{ mol Cu}$ which gives us 0.095 g Cu ($0.00149 \text{ mol Cu} \cdot 63.546 \text{ g/mol}$). The percent of copper in the coin becomes $0.095 \text{ g Cu} / 0.125 \text{ g coin} \cdot 100 = 75.8\% \text{ Cu}$.

Likewise, using the intercept, we found the concentration of nickel in the coin solution to be $0.06367 \cdot 0.166 \text{ M Ni} = 0.0109 \text{ M Ni}$. Again, since the coin was diluted to 50 mL, the number of moles of Ni in the coin solution would equal $0.0109 \text{ M} \cdot 0.050 \text{ liter} = 0.000545 \text{ mol Ni}$, which gives us 0.032 gram of Ni ($0.000545 \text{ mol} \cdot 58.69 \text{ g/mol}$). Therefore, the percent of copper in the coin is $0.031 \text{ g Ni} / 0.125 \text{ g of five-cent coin} \cdot 100$ or 25.7% Ni.

Conclusion

The U.S. mint claims a five-cent coin is composed of 75% copper and 25% nickel. Our data are within acceptable limits of error for absorption spectroscopy, especially considering that both of the metal standards used are of unknown purity. Moreover, the experiment demonstrates the ease at which *Logger Pro* can collect, manipulate, and plot data.

A five-cent coin was analyzed in this example but this technique has also been successfully applied to red food dye, a mixture of red #3 and red #40, as well as Pina Colada Kool-Aid, a mixture of yellow #5 and yellow #6.

References

¹ Blanco, *J. Chem. Ed.*, **66**, 189 (February, 1989)

² Willard, Hobart H., Lynne L Merritt, and John A. Dean, *Instrumental Methods of Analysis*, 4th Ed., D. Van Nostrand Company, 1965, p. 90.

³ *ibid.*, p. 91