CBL Laboratory to Determine The Concentration of Blue 1 in Scope Cool Peppermint

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Revision 0

NOTE! In this document I have included explanatory information in italic type. You should omit the italic text to produce a procedure for your students. If you find any mistakes in the procedure or calculations, please email me.

INTRODUCTION

Since manufacturers want to know as much as possible about their competitors' products, they often perform qualitative and quantitative analyses of the products.

Colorants, often food dyes, are added to products to make them more attractive to the consumer. A common colorant added to consumer products is FD&C Blue 1. If you are assigned the task of determining the concentration of Blue 1 in a product such as Scope Cool Peppermint, you might consider applying a spectrophotometric technique.

Spectrophotometry *[for an explanation of spectrophotometry you can access standard analytical chemistry texts or go to http://www.chem.ubc.ca/courseware/121/tutorials/exp3A/colorimetry or http://www.biology.ucsc.edu/classes/bio20L/content/basics/dyes.pdf or http://dl.clackamas.cc.or.us/ch105-05/colorime.htm]* is a quantitative analytical technique based upon Beer's Law: A=abC, where A=absorbance, a=absorptivity, b=path length of the cuvette, and C= concentration. When the apparatus is designed to measure only electromagnetic radiation in the visible range, the apparatus is known as a colorimeter and the technique is called colorimetry.

There are several experimental designs that might be applied to this type of analysis. In this lab you will prepare a three point standard curve (also known as a working curve), and using this curve, determine the concentration in grams/liter of Blue 1 in Scope Cool Peppermint.

OBJECTIVE

Using the analytical technique of colorimetry, the objective of the lab is to determine the concentration, in g/L, of Blue 1 in Scope Cool Peppermint. You will construct a calibration curve by making multiple dilutions of a working solution of known concentration provided by your teacher. *[To prepare the working solution, add exactly 2.00g of Blue 1 (Aldrich Product Number 86114-6, Aldrich's website is http://www.sigmaaldrich.com) to a 500mL volumetric flask (or 600mL beaker or Erlenmeyer flask) and then dilute to the 500mL mark with water. The concentration of this solution is 4.00g/L. By pipet, transfer 7.5mL of this solution to a 500mL volumetric flask (or beaker or Erlenmeyer flask) and dilute to the 500mL mark with distilled water. This working solution is 0.06g/L. It should be noted that the Blue1obtained from Aldrich (or any other source) is not 100% pure. Information from the manufacturer described the tartrazine used to develop this lab procedure to be 94% colorant. I do not know what technique was used to determine the % colorant. I have not used the purity in the calculations below. You should consider whether this factor should be included in the calculations. Modifying the equations to include the % purity might be an extension or extra credit problem for your students.]*

SAFETY

Wear goggles. It is advisable to wear latex gloves and a lab apron to protect your hands and clothes from stains.

MATERIALS

- Colorimeter
- Cuvette
- TI 83Plus calculator
- CBL
- 100mL graduated cylinder
- Wash bottle
- Distilled (or deionized) water
- Pipet bulb
- 10mL serological pipet
- 1 dropper *[or Beral pipet]*
- Distilled water
- Working solution of FD&C Yellow 5 (tartrazine)
- Cepacol
- Five 16mmX150mm test tubes *[You can use inexpensive and reusable plastic cups available at any grocery store if you do not have enough test tubes.]*
- 1 #6 rubber stopper

[Volumetric flasks would produce more accurate results but the graduated cylinder will suffice. Consider discussing the issue of which glassware would produce the most accurate and precise results.
If you are using other calculators or the CBL2 or LabPro, you will have to make some modifications in the procedure.]

PROCEDURE

If you have the time, see if the students can devise their own experimental design. It is assumed that the students are already familiar with dilution and concentrations calculations.

Preparation of the Working Curve Solutions

You will prepare three "standards" by diluting a working solution of known concentration provided by your teacher. Since you know how the standards were prepared, you will be able to calculate the concentration of the standards. After measuring the absorbance of these standards, you will construct a "working curve", that is, a graph of absorbance vs. concentration. Use of the data and/or curve to determine the concentration of the Blue 1 in the Scope will be described in the "Calculations" section below.

Your teacher will supply you with a test tube containing 20mL-30mL of a FD&C Blue 1 at a concentration of 0.06g/L. Your teacher will also supply you with a test tube containing Scope with a concentration of C_{Sco}. (*If you use volumetric flasks to prepare your working solution, your precision will be better than 2 significant figures*).

3. Transfer by pipet exactly 2mL of the working solution to the 100mL graduated cylinder and fill to the mark with distilled water. Stopper and agitate to produce a homogeneous solution. Transfer this solution to a test tube and label the test tube WS_1. The concentration of this solution is 0.00120g/L. The symbol for this concentration is C_{WS1}. *[Consider having the students perform the concentration calculation instead of supplying them with the value]*
4. Transfer by pipet exactly 5mL of the working solution to the 100mL graduated cylinder and fill to the mark with distilled water. Stopper and agitate to produce a homogeneous solution. Transfer this solution to a test tube and label the test tube WS_2. The concentration of this solution is 0.00300g/L. The symbol for this concentration is C_{WS2}.
5. Transfer by pipet exactly 10mL of the working solution to the 100mL graduated cylinder and fill to the mark with distilled water. Stopper and agitate to produce a homogeneous solution.

Transfer this solution to a test tube and label the test tube WS_3. The concentration of this solution is 0.00600g/L. The symbol for this concentration is C_{WS3}.

Set Up of CBL

6. Plug the colorimeter into CH1 and connect the CBL to the calculator.
7. Turn on both the calculator and the CBL.
8. Press APPS on the calculator and start the ChemBio application.
9. Press ENTER when you see the "VERNIER SOFTWARE" screen.
10. Select #1, "SET UP PROBES".
11. Type 1 for "ENTER NUMBER OF PROBES:" and then press ENTER.
12. Select #4 for the colorimeter sensor.
13. Type 1 for the "ENTER CHANNEL NUMBER:" and then press ENTER
14. Use the arrows on the colorimeter to select the 635nm wavelength and press ENTER on the calculator. *[This set up assumes you have the second generation colorimeters. If you have the original colorimeters, you must modify the set up.]*
15. Rinse the cuvette and fill ¾ full with distilled water. Insert cuvette into the colorimeter and press ENTER.
16. Press CAL on the colorimeter and wait for the red light to stop blinking. Press ENTER.
17. Select #2 for "COLLECT DATA".
18. Select #1 for "MONITOR INPUT". The absorbance of the water should be 0.000-0.002

Absorbances of Working Curve and Cepacol Samples

19. Empty the cuvette and using the dropper, rinse several times with WS_1. Fill the cuvette ¾ full with WS_1. Place the cuvette into the colorimeter and record the absorbance, A_{WS1}.
20. Empty the cuvette and using the dropper, rinse several times with WS_2. Fill the cuvette ¾ full with WS_2. Place the cuvette into the colorimeter and record the absorbance, A_{WS2}.
21. Empty the cuvette and using the dropper, rinse several times with WS_3. Fill the cuvette ¾ full with WS_3. Place the cuvette into the colorimeter and record the absorbance, A_{WS3}
22. Empty the cuvette and using the dropper, rinse several times with Scope. Fill the cuvette ¾ full with Scope. Place the cuvette into the colorimeter and record the absorbance, A_{Cep}.
23. Press the "+" key on the calculator to stop the data collection and then select #7 to quit the program. Turn off the CBL and calculator.

CALCULATIONS

[1 Method III below assumes that there is a linear relationship between absorbance and concentration. It should be noted that the TI83Plus has sophisticated statistical functionality that allows a student to obtain accurate results form non-linear data. Depending upon the time available and the interest or prior knowledge of your students, you might consider discussing how to investigate the linearity of data and how to address non-linear relationships.

2 There are several ways to determine the concentration of the "unknown". You might construct a graph using a piece of graph paper (Method II), you might construct a graph on a calculator or computer (Method I), you might select the working curve concentration closest to the unknown (Method I), you can use a statistical method (Method III), or you can probably find several alternative approaches I have not mentioned. Select the one you think is best.]

Method I-The Single Standard Method

In this method, it is assumed that if one of the standard solutions and the unknown solution exhibit absorbances that are numerically close, Beer's Law can be used to determine a valid value for the concentration on the unknown by comparing the absorbance and concentration of the "known" with the absorbance and concentration of the "unknown". You are to select the standard that exhibits an absorbance closest to the absorbance of Scope.

[If the "standard" solution and the "unknown" solution exhibit absorbances that are numerically close, it is possible to determine an accurate value for the concentration of the unknown even if the absorbance vs. concentration data is non-linear. Care must be taken in the decision as to the limits placed on what is considered "numerically close"]

If A_1=abC_1 and A_2=abC_2 and ab is constant, then $\frac{A_1}{C_1} = \frac{A_2}{C_2}$ *rearranging* $C_2 = A_2 \bullet \frac{C_1}{A_1}$

If A_{Sco} is the absorbance of Cepacol and if it happens that WS_2 exhibits an absorbance, $A_{WS2,}$ closest to A_{Cep}, then the concentration of Scope is given by:

$$C_{Sco} = A_{Sco} \bullet \frac{C_{WS2}}{A_{WS2}}$$

Method II-The Graphical Method

1. Either on a piece of graph paper or a calculator or a computer, plot the concentration-absorbance data pairs, $C_{WS1,}$ A_{WS1}; C_{WS2}, A_{WS2}; C_{WS3}, A_{WS3}, and draw a straight line through the points.

2. Using the absorbance of the Scope solution, A_{Sco}, draw a horizontal line from the y-axis (absorbance) to the straight line and then drop a perpendicular to the concentration axis. Read the concentration value at the point the perpendicular crosses the concentration axis. This is C_{Sco}.

Method III-The Statistical Method

[Inspection of the graph of the data points, an by inspection of the correlations coefficient, suggests the curve is non-linear. Instead of LinReg suggested in #2 below, you might consider QuadReg as a more appropriate data treatment.]

1. Press the STAT key on the calculator and select EDIT. Key in the absorbance values in L1 and the concentration values in L2.

2. Press the STAT key, cursor right to CALC, cursor down to LinReg and press ENTER twice.

3. Press the Y= key and press CLEAR.

4. Press the VARS key, cursor down to Statistics, press ENTER, cursor right to EQ and press ENTER.

5. Press TBLSET key (that is 2nd and then the WINDOW key), and then key in absorbance of Scope to the right of TblStart=.
6. Key in .001 to the right of ΔTbl=, and then press the TABLE (that is, 2nd GRAPH) key.

7. Record the concentration, C_{Sco}, of the Scope solution in the Y_1 column.

RESULTS and CONCLUSIONS

[Included below are some suggestions as to thought provoking items for discussion. You must decide which are appropriate for your students or you might devise your own questions.]

1. What is the concentration of the Scope?
2. How would you determine if your results are correct? *[Answers such as "compare results with other students", or "contact the manufacturer of Scope", or prepare an aqueous solution of Blue 1 at the same concentration as the experimental value and determine the absorbance of this "known" solution, are possible answers]*
3. Which calculation method yields the most accurate results? How do you know? *[Examples: The efficacy of Method III can be evaluated by reference to the R or R^2 values. The experimental values can be compared with the "true" values provided by the manufacturer of Scope.]*
4. There are numerous materials in Scope besides Blue 1. How would you determine if these additives might interfere or change the absorbance of Blue 1? *[One approach is to read the label, buy the ingredients listed and by mixing them with Blue 1, see if the absorbance changes.]*
5. Provide an explanation for the fact that when you contact the manufacturer, the value for the concentration of Blue 1 they provide is lower than the value you determined. *[Besides the simple explanations that the student made a mistake, possible explanations are: The sample of Blue 1 used to prepare the working solution contained non-absorbing impurities. There are materials in the Scope, other than Blue 1, that absorb radiation at 635nm.]*
6. How much solid Blue 1 must be added to a 1L bottle of colorless mouthwash to produce a product with the same color intensity as the sample of Scope you analyzed? *[The equation for the calculation is given below:*

$$\begin{aligned}&\textit{Since}:\\&\textit{Concentration}\left(\frac{grams}{L}\right)\bullet \textit{Volume}(L)=\textit{Mass}(grams)\\&\textit{Therefore}:\\&C_{Sco}\left(\frac{grams\ Blue\ 1}{L\ Scope}\right)\bullet 1L\ Scope = grams\ Blue\ 1\end{aligned}$$