Using Color to Identify an Unknown
Sample Data and Answers to Questions

Pre-lab Questions

1. In this lab, water soluble dyes will be used. These dyes are used to color drinks such as Kool-Aid. How are these different from the pigments that color paints?

Dyes can uniformly distribute in a polar solvent such as water. When measuring the absorbance of a dye, the ratio of light that transmits through the solution can be compared to that of pure water. Pigments are finely ground insoluble particles that will absorb and scatter light. The light that is reflected off these particles is useful to relate back to absorbance.

2. A colored stock solution has a concentration of 1.50 microMolar (µM). If 2.0 mL of the stock solution is used to make a 10.0 mL dilute aqueous solution, what is the new concentration?

\[
\frac{M_1 V_1}{V_2} = \frac{M_2 V_2}{V_1}
\]

\[
1.50 \, \mu \text{M} \times 0.0020 \text{L} = M_2 \times 0.0100 \text{L}
\]

\[
M_2 = 0.30 \mu \text{M}
\]

3. What is meant by calibrating the spectrophotometer by running a “blank”? Why is it a necessary step?

In order to compare the amount of light transmitting through the solution in question to the amount of light transmitted when no solute is present.

4. A solution made with FD&C Yellow Dye #5 has an absorbance measured to be 0.299. Using Beer’s Law, determine the concentration of the dye if the molar absorptivity constant is 27,300 M⁻¹ cm⁻¹.

\[
A = abc
\]

\[
0.299 = 27300 \, \text{M}^{-1} \text{cm}^{-1} \times 1.0 \, \text{cm} \times c
\]

\[
c = 1.10 \times 10^{-5} \, \text{M}
\]

Sample Absorbance Spectra
Sample Beer's Law Curve

\[
f(x) = 0.1321479353x - 0.0050770433
\]
\[
R^2 = 0.9994514326
\]

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<th>Concentration (µM)</th>
<th>Absorbance</th>
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<tbody>
<tr>
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<tr>
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Answers to Extensions

1. A student attempts to make an orange mixture using FD&C Red #40 and FD&C Yellow #1. When attempting to determine the wavelengths of maximum absorbance in the mixture, the student obtains a graph that looks like this:

   ![Mixture of Red + Yellow](image)

   If the student was not aware that this was a mixture of Red and Yellow, would this graph be useful in identifying the unknowns in the mixture? Explain.

   The data appears to have a peak at approximately 425 nm (the wavelength of maximum
absorbance for yellow dye) but then levels off. This would not conclusively point to a mixture of yellow and red.

2. Using the information you have acquired, give a possible explanation as to what may be occurring in the student's graph and a possible solution as to how to determine the components of the mixture.

Since the absorbance spectra for the yellow dye and red dye have overlap near the wavelengths of maximum absorbance, this curve results. This can also be a result of differences in the concentrations of the yellow and red—one may be more dilute than the other which effects the absorbance.

A possible solution is to calibrate the spectrophotometer using one of the dyes in question as the “blank”. When measuring the unknown, the other component in question should have a clear visible peak at the predicted wavelength where absorbance is largest. For example, if yellow is used as the blank, when the absorbance spectrum is collected for this mixture, a peak should appear at approximately 505 nm as is characteristic of the red dye.

3. In your group, design an experiment and perform the experiment to test your hypothesis.

A procedure based off the ideas in question 2 should be designed and performed.

Conclusion

Paint is a mixture of an **insoluble** pigment, a solvent, and a binder (a material that helps the particles stick to each other. The lab that you just completed worked with water **soluble** dyes and analyzed the absorbance of these dyes when exposed to different wavelengths of light.

When light moved through the solutions in this lab, specific wavelengths were absorbed. Those wavelengths not absorbed, were transmitted.

1. Draw a particle diagram to model the transmittance of some wavelengths of light and the absorbance of others.

Diagram should include particles in an aqueous solution. Some light should be pictured transmitted (moving completely through the solution to the other side); some light should be absorbed (stopping at a particle).

2. Draw a particle diagram showing the difference between transmittance of light in a solution and reflection of light off of a pigment (like that found in paint).

Diagram should include particles more closely packed like those of a solid. Some light should be pictured as reflected (hitting a particle then off the solid); some light should be absorbed (stopping at a particle).

4. Using the information you have acquired, propose a method the paint store may be able to use to match an unknown color as closely as possible.

Possible methods may include the use of a spectrophotometer measuring reflected light versus transmitted light.