

Name: _____ Band: _____ Date: _____

You Light Up My Life! Using Spectrophotometry and Beer's Law

Find the movie titled Spectrophotometric Analysis on the website below:

<http://www.oid.ucla.edu/Webcast/chemistry>

Watch the movie (14 minutes in length), and answer the questions below on a separate sheet of paper. Some outside research may be required, but most questions will be answerable after watching the movie, only.

1. Explain why an orange (the fruit) looks orange under white light.

The orange absorbs all wavelengths of light except those corresponding to orange light, which the orange reflects.

2. Explain why an orange (the fruit) looks black under blue light.

The orange absorbs all wavelengths and does not reflect any light, therefore appearing black.

3. What color of light is absorbed by a blue solution?

All colors (especially orange) are absorbed except blue, which is reflected.

4. Explain the difference between a spectrometer and a photometer.

A spectrometer consists of a light source, a wavelength control knob, a prism, and a slit. It separates the light that passes through a solution into its component colors. A photometer consists of a phototube which detects the transmitted light and changes it to a weak electrical signal which is amplified and displayed on a meter in units of absorbance and/or transmittance.

5. What is a spectrophotometer?

A spectrophotometer is an instrument that combines a spectrometer and a photometer. It is used to determine the concentration of a colored solution.

6. Distinguish between absorbance and transmittance.

Absorbance is the amount of light absorbed by a solution. Transmittance is the amount of light that passes through a solution.

7. What is the relationship between absorbance and percent transmittance?

As absorbance increases, percent transmittance decreases.

8. Every solution has a wavelength at which maximum absorbance occurs. What must be true about the value of percent transmittance at this wavelength?

Percent transmittance must be at a minimum.

9. What two factors, other than wavelength, affect the absorbance of a solution?

The other two factors are concentration of a solution and the length of the light path through the solution.

10. What is another name for “length of light path?”

Cell length

11. Write the mathematical expression known as Beer’s Law. Define each symbol and identify the units associated with each quantity. You will need to research the units.

$$A = \epsilon bc$$

A is absorbance (no units)

ϵ is molar absorptivity (L/mol•cm)

b is cell length (cm)

c is concentration (mol/L)

12. Draw a rough sketch of the expected graphical relationship between concentration and absorbance according to Beer’s Law. Make sure to label your axes.

Your graph should have concentration on the x axis and absorbance on the y axis. There should be a line indicating a direct relationship.

13. What is the most common application of Beer’s Law?

Determining the concentration of a solution

14. A blank must be used to set the limits on a spectrophotometer before collecting data. How should you choose what substance goes in a blank?

You should choose a blank that contains everything except the unknown species.

15. To what absorbance should the spectrophotometer be calibrated when the blank is in place? What about percent transmittance of the blank?

Set the spectrophotometer to read 0% absorbance (100% transmittance).

16. The first step in analyzing a colored solution is to obtain an absorbance spectrum. What are the steps for creating an absorbance spectrum, and what values are plotted?

First, obtain a spectrum of the amount of light absorbed vs. the wavelength of light. Second, obtain a Beer’s Law plot of absorbance vs. concentration. Third, measure the absorbance of the solution of unknown concentration.

17. How is maximum absorbance for a solution determined?

You should vary the wavelength of light and create a graph of wavelength vs. absorbance.

18. Write a complete procedure for gathering data for a Beer’s Law plot. Include the term “standard solution” and its definition.

Set wavelength to the wavelength corresponding to maximum absorbance. Insert a blank to check the settings. Rinse cuvette with a dilute standard solution. Add the first standard solution and record the absorbance/percent transmittance. Rinse, and then repeat with the second standard solution. A standard solution is a solution with a known concentration. When all measurements have been recorded, plot concentration vs. absorbance.

19. To what type of solutions does Beer's Law apply: dilute or concentrated?

Applies only to dilute solutions.

20. How is the concentration of an unknown solution determined using a Beer's Law plot?

After measuring the absorbance of the unknown solution, use the line on the Beer's Law plot to find the corresponding concentration.

21. What are the possible sources of error in spectrophotometric analysis?

Instrument error, concentrations of standard solutions are incorrect,

22. What are three applications of spectrophotometric analysis?

Monitoring lead concentration in gasoline, grade of tomato paste, analytical technique in medical/health sciences.

Thought/Research Questions

23. How will leaving fingerprints on the cuvette (i.e. not wiping the cuvette clean) affect the calculated concentration of an unknown? Explain.

The oils and/or dirt left behind on the cuvette may absorb some light, which would decrease the observed absorbance of the solution. This would result in a calculated solution concentration that is higher than the actual concentration.

24. Why is spectrophotometric analysis not appropriate for colorless solutions?

Colorless solutions do not absorb enough light!

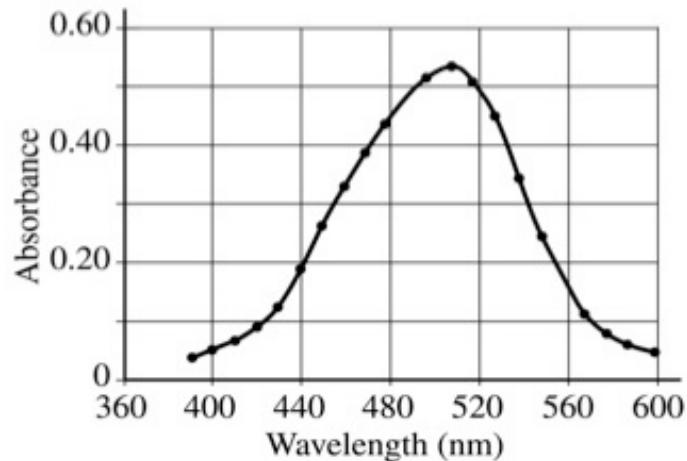
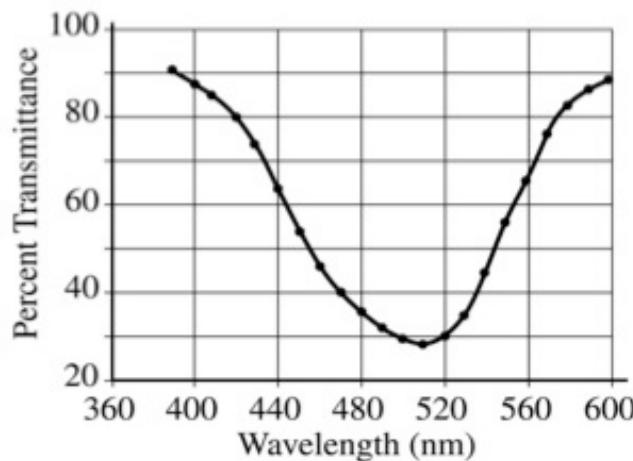
Practice

25. A student is asked to determine the concentration of a cobalt (II) chloride based on absorption of light. The student is provided with a 0.10 M solution of cobalt (II) chloride with which to prepare standard solutions with concentrations of 0.020 M, 0.040 M, 0.060 M, and 0.080 M.

- a. Describe the procedure for diluting the 0.10 M solution to a concentration of 0.040 M using distilled water, a 100 mL volumetric flask, and a pipet. Include specific amounts.

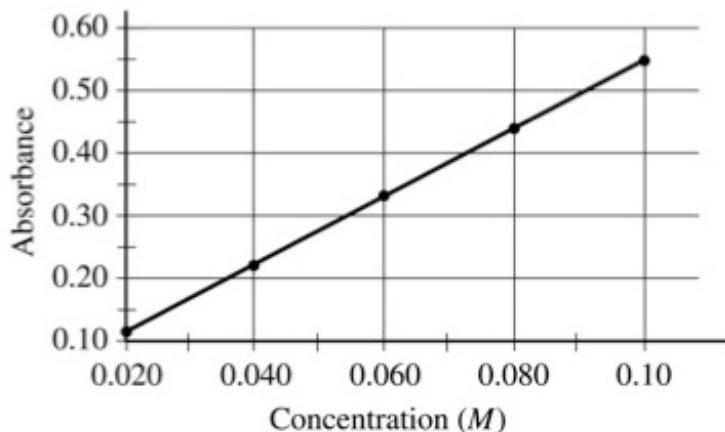
Use a pipet to transfer 40. mL of the 0.040 M solution into the volumetric flask. Add distilled water up to the line on the volumetric flask.

- b. A student finds the percent transmittance and the absorbance of the 0.10 M solution at different wavelengths. She creates the two graphs below. What is the optimum wavelength for analysis?



The optimum wavelength is approximately 510 nm.

- c. The student then uses her solutions to create the graph below.



If the absorbance of the unknown solution is 0.275, what is the concentration of the solution?

0.050 M

- d. Find the Beer's Law formula in your reference packet. Identify two factors that determine the amount of light that passes through the solution and explain how changing those factors changes the amount of light that passes through the solution.

Increasing the cell length would decrease the amount of light that passes through the solution.

Increasing the concentration would decrease the amount of light that passes through the solution.

26. The following are multiple-choice questions from the AP Chemistry exam. Choose the best answer for each question, and explain your choice.

Concentrations of colored substances are commonly measured by means of a spectrophotometer. Which of the following would ensure that correct values are obtained for the measured absorbance?

- I. There must be enough sample in the tube to cover the entire light path.
 - II. The instrument must be periodically reset using a standard.
 - III. The solution must be saturated.
- (A) I only
(B) II only
(C) I and II only
(D) II and III only
(E) I, II, and III

Choice C is correct. Option III is incorrect because the purpose of this analytical technique is to measure concentration. If a solution needed to be saturated in order to be used, then this technique would be rendered useless!

Appropriate uses of a visible-light spectrophotometer include which of the following?

- I. Determining the concentration of a solution of $\text{Cu}(\text{NO}_3)_2$
 - II. Measuring the conductivity of a solution of KMnO_4
 - III. Determining which ions are present in a solution that may contain Na^+ , Mg^{2+} , Al^{3+}
- (A) I only
(B) II only
(C) III only
(D) I and II only
(E) I and III only

Choice A is correct. Spectrophometric analysis is only useful for determining the concentration of colored solutions.