

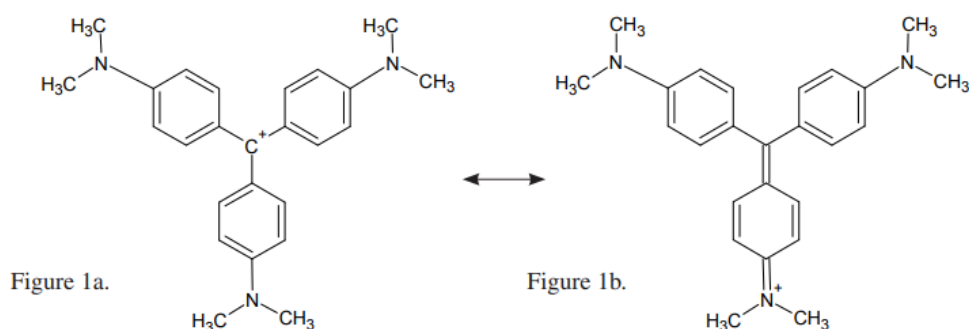
# KINETICS OF CRYSTAL VIOLET FADING

## Introduction

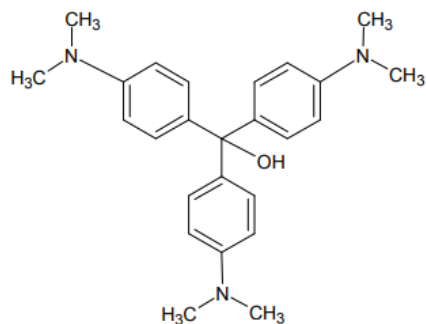
Crystal violet is a common, beautiful purple dye. In strongly basic solutions, the bright color of the dye slowly fades, and the solution becomes colorless. The kinetics of this “fading” reaction can be analyzed by measuring the color intensity or absorbance of the solution versus time to determine the rate law.

## Background

Crystal violet belongs to a class of intensely colored organic compounds called triphenylmethane dyes. The structure and color of crystal violet depend on pH, making it a valuable acid–base indicator as well as an excellent dye. The major structural form of crystal violet is the monovalent cation, abbreviated  $CV^+$ , which is shown in Figure 1a.  $CV^+$  is the predominant form of crystal violet in the solid state and in aqueous solution across a broad range of pH values from pH 1 to 13. The positive charge shown on the central carbon atom in Figure 1a is delocalized via resonance to the three nitrogen atoms. See Figure 1b for one of the three additional resonance forms with the positive charge on a nitrogen atom. Delocalization of the charge across the system of double bonds in the benzene rings stabilizes the carbocation and is responsible for the vibrant purple color of the dye.



**Figure 1.**



**Figure 2.**

# KINETICS OF CRYSTAL VIOLET FADING

In strongly basic solutions the purple  $\text{CV}^+$  cation slowly combines with hydroxide ions to form a neutral product,  $\text{CVOH}$ , which is colorless (see Figure 2). The rate of this reaction (Equation 1) is slower than typical acid–base proton transfer reactions and depends on the initial concentration of both crystal violet and hydroxide ions.



Exactly how much the rate changes as the reactant concentration is varied depends on the rate law for the reaction. In the case of the reaction of  $\text{CV}^+$  with  $\text{OH}^-$  ion, the rate law has the general form

$$\text{Rate} = k [\text{CV}^+]^n [\text{OH}^-]^m \quad \text{Equation 2}$$

The exponents  $n$  and  $m$  are defined as the order of reaction for each reactant and  $k$  is the rate constant for the reaction at a particular temperature. The values of the exponents  $n$  and  $m$  must be determined by experiment. If the reaction is carried out under certain conditions then Equation 2 will reduce to the form

$$\text{Rate} = k' [\text{CV}^+]^n \quad \text{Equation 3}$$

where

$$k' = k [\text{OH}^-]^m \quad \text{Equation 4}$$

The constant  $k'$  is a new “pseudo” rate constant incorporating both the “true” rate constant  $k$  and the  $[\text{OH}^-]^m$  term. Equation 3 is referred to as a pseudo-rate law because it is a simplification of the actual rate law, Equation 2.

The pseudo-rate law is valid when the concentration of  $\text{OH}^-$  ions is much greater than the concentration of  $\text{CV}^+$  ions. Under these conditions the  $[\text{OH}^-]^m$  term in Equation 2 will not change much over the course of the reaction and may be treated as a constant in the rate equation.

Recall that the absorbance for a specific concentration of a solution with a fixed path length varies directly with the absorptivity coefficient of the solution. This relationship is known as Beer’s law.

$$A = abc \quad \text{Equation 5}$$

where  $A$  is absorbance,  $a$  is the molar absorptivity coefficient,  $b$  is the path length in cm, corresponding to the distance light travels through the solution, and  $c$  is the concentration of the solution. Beer’s law provides the basis of using spectroscopy in quantitative analysis. Using this relationship, concentration and absorbance may be calculated if one variable is known while keeping  $a$  and  $b$  constant. This relationship is also extremely valuable in kinetics experiments, making it possible to follow the rate of disappearance of a colored substance by measuring its absorbance as a function of time.

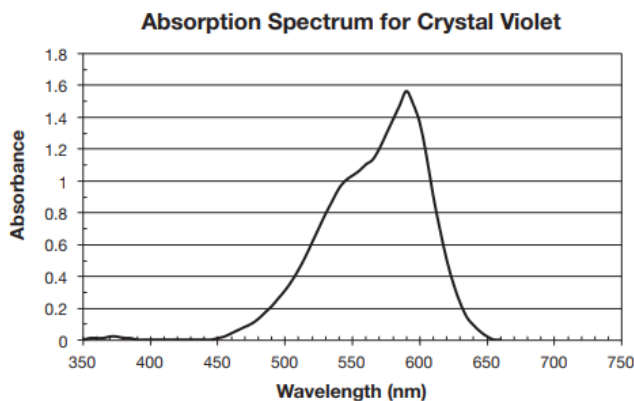
## Experiment Overview

The purpose of this advanced inquiry lab activity is to use spectroscopy and graphical analysis to determine the rate law for the color-fading reaction of crystal violet with sodium hydroxide. The lab begins with an introductory activity (laboratory technique) for constructing a calibration curve of absorbance versus concentration for the dye crystal violet. A series of known or standard solutions is prepared from a stock solution of crystal violet and the absorbance of each solution is measured at an optimum wavelength. A Beer’s law plot of absorbance as a function of concentration may be used to calculate the concentration of any “unknown” solution of the dye in a rate law experiment. The procedure provides a model for guided-inquiry design of experiments to determine the order of reaction with respect to both crystal violet and sodium hydroxide. Additional triphenylmethane dyes, malachite green and phenolphthalein, are also available for optional extension or cooperative class studies.

# KINETICS OF CRYSTAL VIOLET FADING

## Pre-Lab Questions for the Crystal Violet Standard (Part A)

The visible absorption spectrum for crystal violet,  $\text{CV}^+$ , is shown in Figure 3. The concentration of the dye was  $12.5 \mu\text{M}$  ( $12.5 \times 10^{-6} \text{ M}$ ).



**Figure 3.**

1. What would be the optimum wavelength for generating a Beer's law calibration curve for crystal violet and measuring absorbance versus time for the reaction of  $\text{CV}^+$  with  $\text{OH}^-$ ? Recall that absorbance measurements are most accurate and sensitive in the range of 0.2-1.0.
2. A calibration curve requires the use of several concentrations of the test solution. Using  $25 \mu\text{M}$   $\text{CV}^+$  solution as the stock solution, complete the following table to show how you would prepare 2.5, 5, 7.5, 10, and  $12.5 \mu\text{M}$  solutions of  $\text{CV}^+$ . Assume that the final solution volume should be 10.0 mL in all cases.

**Table 1**

	<b>CV Stock Solution</b>	<b>A</b>	<b>B</b>	<b>C</b>	<b>D</b>	<b>E</b>
Concentration (micromolar, $\mu\text{M}$ )	25 $\mu\text{M}$	2.5 $\mu\text{M}$	5.0 $\mu\text{M}$	7.5 $\mu\text{M}$	10.0 $\mu\text{M}$	12.5 $\mu\text{M}$
Water (mL)	0					5.0 mL
Stock Solution (mL)	10.0					5.0 mL

# KINETICS OF CRYSTAL VIOLET FADING

## Crystal Violet Standard Laboratory (Part A)

1. Turn on the colorimeter and allow it to warm up for 10-20 minutes before use. Adjust the wavelength setting to the optimum wavelength determined in the *Pre-Lab* section.
2. Construct an appropriate data table to record measurements.
3. Using a 10 mL graduated cylinder for accuracy, prepare the series of standard dilutions of the crystal violet stock solution. Use the amounts calculated in the *Pre-Lab* assignment. *Lab Hint:* To avoid contaminating with stock solution, use separate graduated cylinders and disposable pipets with the stock solution and the distilled water.
4. Measure and record the absorbance of the stock solution and each standard solution (dilution) concentration at the selected wavelength.
5. Prepare a Beer's law calibration curve of absorbance versus concentration for crystal violet and report the equation in your laboratory notebook.

# KINETICS OF CRYSTAL VIOLET FADING

## Pre-Lab Questions for the Kinetics of a Chemical Reaction (Part B)

1. Chemical Reactions all take a certain amount of time to react. The reaction rate of a chemical reaction is defined as the change in concentration of a reactant per unit time. If the rate of the reaction does not depend upon the concentration of the reactant, it will be a zero-order chemical reaction. If the rate of the reaction is proportional to the concentration of the reactant (or to the first power), it will be a first-order chemical reaction. If the rate of the reaction is proportional to the concentration of the reactant squared, it will be a second-order chemical reaction.

Match the order of the kinetics of a chemical reaction with its rate law order below:

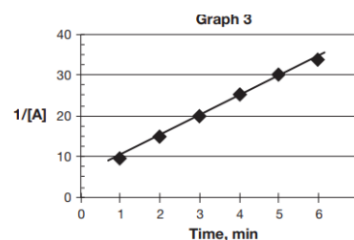
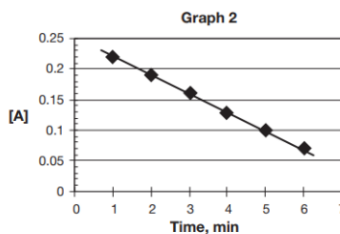
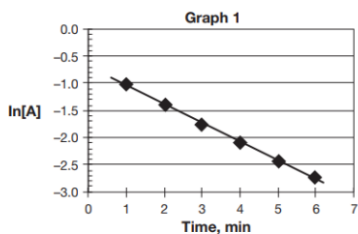
(a) Zero-Order, (b) First-Order, or (c) Second-Order with respect to [A]. (Note: [A] is used to represent the concentration of the reactant):

$$\text{Rate} = k [A]^1$$

$$\text{Rate} = k [A]^2$$

$$\text{Rate} = k$$

2. Consult an online resource or chemistry textbook for the mathematical treatment and graphical analysis of experimental data of concentration versus time for the disappearance of a reactant [A] in a reaction. Match the linear graph shown below with that expected if the reaction is (a) Zero-Order, (b) First-Order, or (c) Second-Order with respect to [A].



3. When a student was conducting a laboratory to determine the kinetics or order of a specific chemical reaction involving substance A, they first did a standard curve to know how the concentration of substance A relates to its absorbance (Part A). Then the student took a specific concentration of substance A and mixed it with an excess amount of substance B. They efficiently mixed the substance, placed an aliquot in a cuvette, placed the cuvette in a colorimeter, and pressed collect. The absorbance was recorded every 20 seconds for 3 minutes. Using the calibration curve from Part A, the absorbance values were converted over into concentration.

## KINETICS OF CRYSTAL VIOLET FADING

Fill out the table below using a calculator to determine the  $\ln [A]$  and  $1/[A]$  values.

Time (seconds)	[A]	$\ln [A]$	$1/[A]$
20	0.1003		
40	0.0743		
60	0.0550		
80	0.0408		
100	0.0302		
120	0.0224		
140	0.0166		
160	0.0123		
180	0.0091		

- Using a computer graphing device, graph each of the following and sketch the graphs in your laboratory notebook:
  - Time versus [A]
  - Time versus  $\ln [A]$
  - Time versus  $1/[A]$
- Using your graphical relationship of each, determine the order of the rate law of substance [A].

### Laboratory to Determine the Kinetics of a Crystal Violet (Part B)

- Set the colorimeter to the proper wavelength (Part A) and calibrate the instrument using a “blank” of equal volumes distilled water and 0.10 M NaOH.
- Measure 10.0 mL of 25  $\mu\text{M}$  crystal violet stock solution using a 10 mL graduated cylinder and add it to a clean 50 mL beaker.
- Measure 10.0 mL of 0.10 M NaOH in a different 10 mL graduated cylinder.
- Important:** Once you mix the two solutions, the chemical reaction has begun, so do not mix these two solutions until you are absolutely prepared and ready to test the solution in the colorimeter.
- Get a cuvette ready and set the colorimeter to collect for at least 600 seconds (about 10 minutes).
- Very efficiently mix both solutions in the beaker, swirl to mix uniformly, add a sample to the cuvette, place the cuvette inside the colorimeter, and press collect.
- Record absorbance readings every 15 seconds. (0.066666666 samples/sec)
- Using your calibration curve equation from Part A, convert the absorbance of each crystal violet reading into a concentration, [CV].
- Calculate the  $\ln[\text{CV}]$  and  $1/[\text{CV}]$  in the same manner that Pre-Lab Part B was calculated.
- Using a computer graphing device, graph each of the following and sketch the graphs in your laboratory notebook:
  - Time versus [CV]
  - Time versus  $\ln [\text{CV}]$
  - Time versus  $1/[\text{CV}]$

## KINETICS OF CRYSTAL VIOLET FADING

11. Using your graphical relationship of each, determine the order of the rate law of substance [CV].