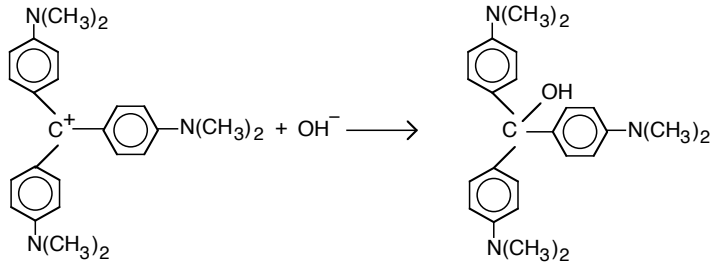
CRYSTAL VIOLET KINETICS

INTRODUCTION

In this experiment, you will observe the reaction kinetics between crystal violet and sodium hydroxide. The net ionic equation for the reaction is shown here:



A simplified version of the equation is:

$$CV^+(aq) + OH^-(aq) \rightarrow CVOH(aq)$$

The rate law for this reaction is in the form:

$$\text{rate} = k[CV+]^m[OH-]^n \qquad (1)$$

Where *k* is the *true rate constant* for the reaction, *m* is the order with respect to crystal violet (CV^+), and *n* is the order with respect to the hydroxide ion (OH^-). In this experiment the crystal violet is the limiting reactant and the rate will depend only on the concentration of crystal violet. The initial hydroxide ion concentration is more than 2500 times as large as the concentration of crystal violet and will not change appreciably during the reaction. By combining the true rate constant k and the initial concentration of hydroxide ion, $[OH^-]_o^n$, into a new *pseudo rate constant, k'*:

$$k' = k[OH^-]^n \qquad (2)$$

We can rewrite equation (1) as:

$$\text{rate} = k'[CV^+]^m \qquad (3)$$

The new constant k' is called a *pseudo rate constant* since it is a product of the true rate constant, k, and the initial concentration of a reactant *that is in large excess*.

As the reaction proceeds, the violet-colored crystal violet will be slowly changing to a colorless product. Using a spectrometer (see figure at left), you will monitor the absorbance of the crystal violet reactant in solution with time. We will assume that absorbance, Abs, is proportional to the concentration of crystal violet according to Beer's Law.

$$\text{Abs} = m[CV^+] + b \qquad (4)$$

Where m is a proportionality constant, (slope) and b is the y-intercept.

To determine the order of the reaction with respect to crystal violet the integrated rate laws are used. A plot of $[CV^+]$ versus time will give a concentration profile that will ***fit ONLY ONE*** of the following integrated rate laws since the order for crystal violet is either 0, 1 or 2.

0. *Zero order, m = 0*: $[CV^+]_t = -k't + [CV^+]_0$

1. *first order, m = 1*: $\ln[CV^+]_t = -k't + \ln[CV^+]_0$

2. *second order, m = 2*: $1/[CV^+]_t = +k't + 1/[CV^+]_0$

In these equations the subscript *t* means the concentration at any time t and the subscript *o* means the concentration at time zero. Once the order, m, with respect to crystal violet has been verified, you will also find the order with respect to the hydroxide ion, n, then the true rate constant, k, and the reaction half-life, $t_{1/2}$.

MATERIALS

LabQuest	0.10 M NaOH	waste beaker or flask
3 small beakers or flasks	2.0×10^{-5} M crystal violet	Automatic Pipet (5 mL) with tips
Vernier SpectroVis Spectrometer	distilled water	one plastic cuvette

SAFETY

Crystal violet is a biological stain. Avoid spilling it on your skin or clothing.
0.10 M sodium hydroxide is corrosive. Avoid contact with skin and eyes. Cleanup immediately if spilled.

ABSORBANCE STANDARDS GUIDE

Three standards will be prepared and their absorbance measured to obtain a Beer's Law graph. **Write this table on your DATA PAGE in your notebook. GIVE THE REQUIRED VOLUMES in your PROCEDURE PAGE.**

Crystal Violet Standards Data Table

Standard	Crystal Violet (mL)	Distilled Water (mL)	Calculated CV^+ Molarity	Measured CV^+ Absorbance
1	0.70	2.30		
2	1.10	1.90		
3	1.50	1.50		

STEP 1: CALIBRATING THE SPECTROMETER FOR ABSORPTION MEASUREMENTS

1. Obtain ≈ 10 mL of crystal violet and distilled water in small beakers or flasks.
2. Plug the spectrometer into the standard USB port of the LabQuest. The LabQuest should default to absorbance measurements.
3. Insert a cuvette about ¾ full with distilled water into the spectrometer. Align the cuvette so the clear sides of the cuvette are facing the light source (east-west).
4. **Choose Sensors ➤ Calibrate ➤ USB:Spectrometer**. There may be a slight delay before the calibrate screen appears. When warm-up is complete, select **Finish Calibration**. After the message "Calibration Completed" appears, **tap OK**. Remove the blank cuvette.

STEP 2: GENERATING A FULL ABSORBANCE SPECTRUM – SETTING THE WAVELENGTH OF MAXIMUM ABSORBANCE

1. The data collection mode should be Full Spectrum. If not, tap the **Mode:** window and set to **Full Spectrum**. Use the default parameters as listed.
2. Shake dry your cuvette. Prepare the THIRD absorbance standard by pipetting 1.50 mL of crystal violet and then 1.50 mL of distilled water directly into the cuvette. **If the cuvette is more than ¾ full you have pipetted incorrectly**. Please call your instructor over to correct this issue. Use the appropriately labeled pipet tips. Cap the cuvette and invert twice to mix. *This standard will first be used to find the wavelength of maximum absorbance*.
3. Insert the standard into the spectrometer. **Tap the green start arrow** to collect a spectrum. The Spectrometer will display a real-time absorbance spectrum of crystal violet. **Tap the red Stop button to stop**. **DO NOT REMOVE THE STANDARD**.
4. The LabQuest will automatically identify the wavelength of maximum absorbance, λ_{max}. **Record this wavelength in your notebook.** This wavelength will be used by the LabQuest to measure the absorbance of the standards and the kinetic data.

STEP 3: MEASURING THE THREE ABSORBANCE STANDARDS

1. Before proceeding, determine the molarity of each standard solution through dilution. **Enter your values into the Crystal Violet Standards Data Table in your notebook DATA PAGE.** Use the actual molarity given on the stock solution of crystal violet in your dilution calculations.
2. **Tap the Meter tab**. On the meter screen tap **Mode.**
3. Change the mode to **Events with Entry**. Enter a **Name** (e.g. *Concentration*) and **Units** (*M*).
4. **Select Average over 10 seconds**. **Select OK**.
5. A message will appear warning you to either save or discard the Full Spectrum run. Select Discard.
6. Using standard 3, still in the cuvette and spectrometer, start data collection by **tapping the green start arrow**. A graph should appear with the real-time absorbance being shown on the graph and to the right. After the absorbance reading stabilizes, **tap KEEP**. After the ten-second averaging is complete enter the molarity of this absorbance standard into the LabQuest. **Select OK**. You have just stored your first point for the Beer's Law plot.

7. Empty the cuvette contents into your waste container. Shake dry as completely as possible your cuvette. Prepare the second absorbance standard and insert into the spectrometer. After the absorbance reading stabilizes, **tap KEEP**. After the ten-second averaging is complete enter the molarity of this second absorbance standard. **Select OK**.
8. Repeat the previous step using standard 1.
9. **Tap the red Stop button to stop.** Empty the cuvette contents into your waste container.

STEP 4: ANALYSIS OF THE STANDARDS– BEER'S LAW ANALYSIS

1. A Beer's Law graph of absorbance versus concentration should now be displayed on the LabQuest.
2. **Tap the List icon** to see a list of the molarities and absorbance values. If any molarities were entered incorrectly, correct them now. **Record the absorbance values in the Crystal Violet Standards Data Table on your notebook DATA PAGE.**
3. **Tap the Graph icon. Choose File ➤ Save…** Enter a name for the data set (e.g. **Standards**) and **select SAVE**. The data set is now saved on the LabQuest flash memory. The graph should reappear on the LabQuest after the file is saved.
4. To fit the standards graph to a straight line **Choose Analyze ➤ Curve Fit**. Select the **Abs check-box**. From the **Fit Equation** menu select **Linear**. The slope and y-intercept values should be displayed. **Select OK**.
5. **Record slope and y-intercept values in your notebook.** The slope value should be around $5x10^4$ and the y-intercept close to zero (±0.030). Check with your instructor if your values seem incorrect. Units of the slope are M^{-1} while the y-intercept is unit less.

STEP 5: ACQUIRING THE CRYSTAL VIOLET KINETIC DATA – RUN 1 AND RUN 2.

1. Tap the **Meter icon** and then tap **Mode**. Select **Time Based**. Set the parameters as follows:
   Rate: **0.5 samples/s** Interval: **2 s/sample** Duration: **180 s** Advanced: **Oversampling** is checked.
   Select OK.
2. A message will appear warning you to either save or discard the previous data (Beer's Law data). Select **Discard**. (You already should have saved the data. If not, save it now.)
3. Obtain ≈ 5 mL of sodium hydroxide in small beaker or flask.
4. **Run 1**:
   a) Shake dry as completely as possible your cuvette. Pipet 1.50 mL of crystal violet into the cuvette. Pipet 1.50 mL of sodium hydroxide into the cuvette. Cap the cuvette and invert twice to mix.
   b) Place the cuvette into the spectrometer and **Tap the green start arrow** to collect the Run 1 absorbance versus time curve.
   c) When data collection is complete, **tap the file cabinet icon**. This prepares the graph for Run 2.
   (Note: Run 1 is stored in RAM memory but not in the LabQuest flash memory.)
5. **Run 2**:
   a) Shake dry as completely as possible your cuvette.
   b) **FIRST**: Pipet 0.75 mL of distilled water and 0.75 mL of sodium hydroxide into the cuvette.
   SECOND: Pipet 1.50 mL of crystal violet into the cuvette. Cap the cuvette and invert twice to mix.
   c) Place the cuvette into the spectrometer and **Tap the green start arrow** to collect the Run 2 data.
   d) When data collection is complete, **Choose File ➤ Save**… **Enter YOUR LOCKER NUMBER** as the name for the data set (e.g. 24C) and select **SAVE**. The data set is now saved on the LabQuest flash memory.
6. Have your instructor check the kinetic runs on the LabQuest before cleaning up.

STEP 6: TRANSFERRING YOUR DATA TO LOGGER PRO AND CLEAN UP

1. Your two kinetics runs in the LabQuest need to be transferred to Logger Pro via a USB cable for analysis. Start Logger Pro, attach the LabQuest with a USB cable (available from your instructor/stockroom) and follow the transfer instructions. If you do not have a laptop – your instructor will store the data on their computer and then distribute the Logger Pro file to you via email or a website download.
2. When you are finished DO NOT FORGET to remove the sample cuvette from the spectrophotometer.
3. Pour all remaining solutions into your waste beaker. Dispose of them in the appropriate waste bottle.

CRYSTAL VIOLET KINETICS: DATA ANALYSIS

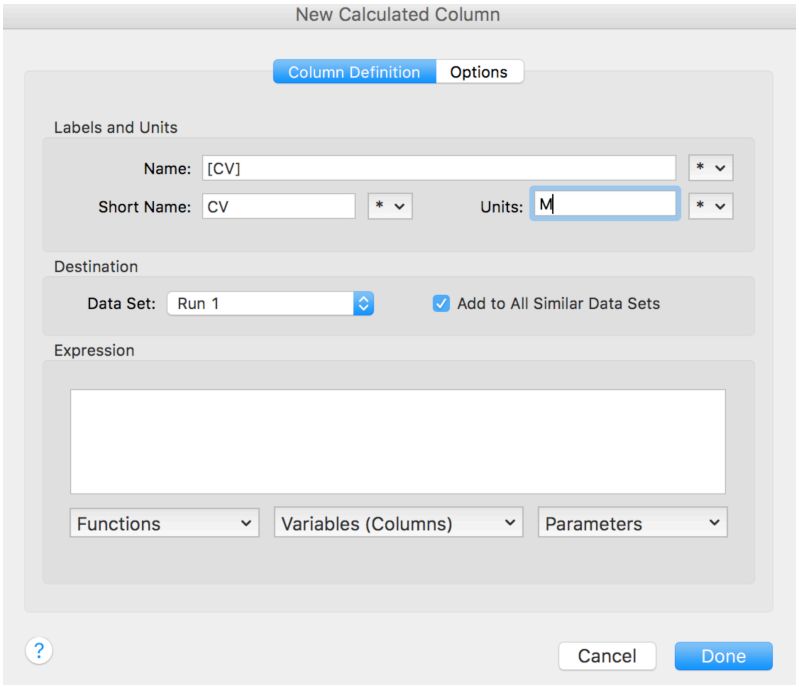
The kinetic data analysis will be done in lab using a computer. Either you or your lab partner should have a laptop with your kinetic data loaded into Logger Pro for analysis.

CRYSTAL VIOLET KINETICS DATA ANALYSIS – LOGGER PRO

1. Open the Logger Pro program, and then import your data by opening the "File" dropdown menu and selecting "Open". Then, find your downloaded Logger Pro data from the experimental day of lab (if you got it off the class website, it might be in your download folder).

2. Once your two runs have been opened in Logger Pro, delete the first two blank columns labeled "X" and "Y". To do this, click the title box for a column, right click, then select "delete column".

3. **Use the "New Calculated Column" function** to create a column **in each data set** (Run 1 and Run 2) to convert the absorbance values into absolute concentration of crystal violet. **The column name should be "[CV]", the units are molarity, M**. As shown below, use ***CV*** as the short name for the new calculated column, and make sure the "Data Set" drop down menu is changed to "Run 1", and the box "Add to All similar data sets" is checked. The slope and y-intercept from the Beer's Law plot made in lab from the standards is used to convert the absorbance readings for each run to absolute crystal violet concentrations. You need to enter the formula: ("Abs" – b)/m. Where "Abs" is the column name for your absorbance data for Run 1 and Run 2, b is the value of the y-intercept and m is the value of the slope from your Beer's Law standards graph. These should be recorded in your notebook. **The actual values of b and m are typed in by you, they are not typed as the letter "b" and the letter "m".** If your value of b from the Beer's law graph is negative, you need to *ADD* the positive value of b to the absorbance in the equation above.



GRAPH 1: ZERO ORDER GRAPH

Make a graph of [CV] vs time for both runs on the same graph. Fit each run to a LINEAR regression line, the zero order rate law: $[CV^+]_t = -k't + [CV^+]_0$.

GRAPH 2: FIRST ORDER GRAPH

Make a new **PAGE** in Logger Pro. Make a new calculated column for **ln[CV]** to make the first order graph. Make sure the same box and data set are selected when making this new column. Graph both run 1 and run 2 of ln[CV] vs. time from the function: $\ln[CV^+]_t = -k't + \ln[CV^+]_0$ on the same graph. Fit a linear regression line to this data.

GRAPH 3: SECOND ORDER GRAPH

Make a new PAGE in Logger Pro. Make a new calculated column for **1/[CV]** to make the second order graph. Make sure the same box and data set are selected when making this new column. Graph both run 1 and run 2 of 1/[CV] vs. time from the function: $1/[CV^+]_t = k't + 1/[CV^+]_0$ on the same graph. Fit a linear regression line to this data.

CRYSTAL VIOLET KINETICS – REPORT SHEET

PRELIMINARY CALCULATIONS:

Use the molarities of the crystal violet and sodium hydroxide stock solutions provided on the bottles to calculate the concentrations of crystal violet and sodium hydroxide **at the time of mixing (dilution calculation)**. These are the concentrations of reactants in the system when mixed, but before they start to react with each other. Show your work clearly! THESE DILUTION CALCULATIONS COME FROM THE RUN 1 and RUN 2 KINETICS PIPET VOLUMES. NOT THE BEER'S LAW STANDARDS.

Run 1

Run 2

CRYSTAL VIOLET GRAPHS: ATTACH GRAPHS OF YOUR ZERO, FIRST AND SECOND ORDER FITS TO THIS REPORT.

1. The data you obtained should show the crystal violet fits the first-order integrated rate law. The crystal violet – hydroxide reaction is first order with respect to crystal violet. If your data shows otherwise, consult your instructor!
2. The values of ***k*** for Run 1 and Run 2 determined from the linear fit to your data are actually ***the pseudo rate constants (k'$_1$ and k'$_2$)*** from the crystal violet first-order integrated rate law as defined by eqn 3. (**Report these values to 2 sig figs only and do not forget correct units**).

   Run 1: ***k'$_1$*** _____ Run 2: ***k'$_2$*** _____

3. **The order of reaction with respect to the hydroxide ion, n,** can be determined by comparing the pseudo rate constant, k' from each run. The ratio of the two pseudo rate constants will give an expression for determining n. In this equation, the OH^- concentrations are the initial concentrations of hydroxide ion in solution for run 1 and run 2. From the procedure, this should be a ratio of 2 to 1. (See your preliminary dilution calculations for these experimental values.) The pseudo rate constants will differ for each run if the order for the hydroxide ion is not 0 as seen by the following ratio.

$$\frac{k_1'}{k_2'} = \frac{k[OH^-]_1^n}{k[OH^-]_2^n} = \left(\frac{[OH^-]_1}{[OH^-]_2}\right)^n = 2^n$$

n = _____ (The correct experimentally determined order for the hydroxide ion is 1.)

4. Now that the order of the reaction for the hydroxide ion has been determined, the true rate constant, ***k***, can be determined from equation 2 for each run: ***k'*** = ***k***[OH^-]. Solve this equation algebraically for the true rate constant ***k***. You need to use the initial hydroxide concentration in your calculation of each *k*. (THINK! Should these be the same? Why?)

k*$_1$** _____ ***k*$_2$** _____ average ***k _____

5. Finally, write the complete rate law expression for the crystal violet sodium hydroxide reaction using the average value for ***k*** and correct orders. **Don't forget the units!**

   Rate =

6. Looking at your orders, how would the rate of reaction change for the crystal violet if the initial hydroxide concentration were doubled while the crystal violet concentration was decreased by a factor of 3?

EXTENDED ANALYSIS OF RUN #1.

Your RUN 1 data line and points are represented by the first-order integrated rate law: $\ln[CV]_t = -kt + \ln[CV]_0$

Here, *k* is really *k' for your RUN 1 data set.*

Use the values obtained by the Logger Pro linear fit from RUN 1 for all the following calculations. Write these parameters below, rounded to 2 significant figures with units!

$[CV^+]_0$ = _____ ***k*** = _____

1. Calculate the instantaneous rate of crystal violet consumption at the beginning of the data collection, t = 0 seconds.

   $Rate_0 = k[CV^+]_0$ =

2. Calculate the crystal violet concentration at the end of data collection, t = 180 seconds. Use $\ln[CV^+]_t = -kt + \ln[CV]_0$.

   $[CV^+]_{180}$ =

   a. Now, using this concentration for crystal violet, calculate the instantaneous rate of crystal violet consumption at t = 180 seconds.

      $Rate_{180} = k[CV^+]_{180}$ =

b. How does this rate (t = 180 seconds) compare to the instantaneous rate of crystal violet consumption at the beginning of the data collection (t = 0 seconds)? Why do they differ?

c. Over this 180 seconds of reaction time:

i. What was the change in crystal violet concentration? ($\Delta[CV^+] = ?$)
(this will be a negative number)

ii. What was the change in sodium hydroxide concentration over this same time period? Use the reaction stoichiometry. ($\Delta[OH^-] = ?$) (this will also be a negative number)

iii. What is the remaining concentration of sodium hydroxide in solution at t = 180 s?
($[OH^-]_{180} = [OH^-]_0 + \Delta[OH^-]$)

iv. Did the $[OH^-]$ change significantly compared to the initial concentration? Why or why not?

3. Estimate the time required, t, to reach a concentration of 1.0×10^{-6} M for the crystal violet, $[CV^+]_t = 1.0 \times 10^{-6}$ M.

4. How much time would be needed to react 99% of the initial crystal violet?

5. Calculate the ½-life for Run 1, $t_{1/2}$. The ½-life is the time for ½ of the initial concentration of crystal violet to react, leaving ½ of the crystal violet in solution.

The first-order ½-life eqn: $t_{1/2} = \dfrac{\ln(\mathbf{2})}{k}$ =

a. On your first-order graph, draw a vertical line at the ½-life time that intersects your concentration curve for Run 1. Check, at this time, is there ½ of the crystal violet remaining in solution?

6. The concentration of crystal violet at the time of mixing (preliminary calculation) should be slightly greater than when you begin data collection, why?

   a. Using the concentration of crystal violet at the time of mixing (preliminary calculation), calculate how much time elapsed from the time of mixing until you collected the first data point, $[CV^+]_0$ for Run 1.