Determination of an Unknown Measuring Acetaminophen using Cyclic Voltammetry

This experiment is designed as an approachable introductory experiment for students to learn the linear relationship between current and concentration in cyclic voltammetry experiments.

ESTIMATED TIME

We estimate that this experiment can be completed in one three-hour class period.

TIPS

1. In the Electronic Resources you will find PDF and word-processing files of the student experiment. You can print the PDF, distribute it to students electronically, or post the file to a password-protected class web page or learning management system. Edit the word-processing file if you would like to tailor the experiment to suit your equipment and students. Sign in to your account at **www.vernier.com/account** to access the Electronic Resources.

2. Due to the pH-dependent electrochemical behavior of acetaminophen, it is important to consider the buffer provided to the students. Because acetaminophen maintains its electrochemical reversibility at higher pH, use a slightly alkaline buffer solution for best results. Additionally, use a buffer that is free of significant redox peaks in the potential region where acetaminophen undergoes redox reactions (from –0.5 V to +1.0 V). Borate buffers meet this criteria well and are recommended for this experiment.
   - Contact lens rinsing and storage solution serves as a consumer product for borate buffer solutions. This ready-to-use borate buffer is available in most drug stores. Look for brands that contain the ingredients: boric acid, sodium borate, and sodium or potassium chloride. A small amount of preservative such as edetate disodium has negligible effects, but other additives, such as polymers containing ethylene diamine, tend to give interfering oxidation peaks located in the same potential region as that for acetaminophen (e.g., Equate™ Lens Cleaner: Multi-Purpose Solution, purchased from Walmart, and most solutions sold as a "multi-purpose solution").
   - Alternatively, you can prepare a 0.1 M borate buffer of approximately pH 8.5.

3. To prepare the sample with an unknown concentration of children's acetaminophen medication suspension follow these steps:
   a. Due to the viscosity of the sample, it is recommended that you weigh the medication sample during the preparation of the stock solution and calculate the sample volume using its density. The density of the children's Tylenol® is typically 1.31 g/mL. The acetaminophen concentration in the suspension should be written on the bottle. In the case of the sample data presented below, it was 160 mg per 5 mL.

b. To prepare 100 mL of a 10 mM children's medication stock solution, weigh out 6.2 g of the children's medication suspension into a 100 mL volumetric flask and dilute to the mark with buffer. Make sure you record the exact mass, you will want to give the value to students so they can compare their experimentally determined value with the known value.

c. A 10 mM stock solution is too concentrated for optimal measurements using cyclic voltammetry. You will need to prepare a 1:10 dilution of this stock solution and give it to students. Each student needs about 30 mL of the diluted sample. To prepare 500 mL of this sample, dilute 50 mL of the stock solution to 500 mL with buffer solution in a volumetric flask.

4. It is prudent to try this experiment with the solutions you have prepared prior to giving the solution to your students to ensure proper concentrations and experimental parameter setting. Modify the student procedure if necessary.

5. If students get a signal greater than 2 μA at approximately 500 mV for their 0.0 mM buffer solution, replace the SPE with a new one.

6. Here are some generic troubleshooting tips if the voltammogram has additional peaks or if signal saturation occurs:
   - While testing your solution before giving it to the students, make sure the recommended parameters in the student instructions are still appropriate. You may want to modify the Switching Potentials.
   - Particularly when using acetaminophen suspension samples, it is common for the electrode to get fouled due to additives like colors, flavors, and suspension stabilizers. A fouled electrode cannot accurately quantify the acetaminophen concentration because the effective electrode area decreases. If you or your students start to see irregular peaks, it is best to replace the SPE with a new, unused SPE. Alternatively, you can activate the carbon surface of the electrode according to the user manual for the SPE.
   - Students have a tendency to wipe the SPE surface too hard. This can also lead to issues with the voltammogram. If students are measuring solutions from least concentrated to most concentrated, they can skip the cleaning step in the procedure.
   - If the current setting is set to High and the voltammogram stops plotting in the middle of a run, it is likely that your sample is too concentrated. Dilute your sample and repeat data collection.
   - If the voltammogram is too noisy or stops plotting during a run and is not set on High, consider increasing the current setting in the parameters.

HAZARD ALERTS

The chemical safety signal words used in this experiment (**DANGER** and **WARNING**) are part of the Globally Harmonized System of Classification and labeling of Chemicals (GHS). Refer to the Safety Data Sheet (SDS) that came with the chemical for proper handling, storage, and disposal information. SDS can also be found online from the manufacturer.

Acetaminophen, solid, $CH_3CONHC_6H_4OH$: **WARNING**: Acute toxicity, oral. Harmful if swallowed. Do not eat, drink or smoke when using this product. Skin and serious eye damage, corrosion or irritation. Causes skin and serious eye irritation. May cause respiratory irritation.

Product should be treated as a chemical and is not for consumption as it has been stored with other non-food-grade chemicals. Store in a locked poison cabinet.

ANSWER TO PRE-LAB ACTIVITY

Students should use $M_1V_1 = M_2V_2$ and present their plan for how to prepare (via serial dilution) 25 mL each of the required solutions.

M_1 (mM)	V_1 (mL)	M_2 (mM)	V_2 (mL)
1.0	20.00	0.8	25.0
0.8	18.75	0.6	25.0
0.6	16.67	0.4	25.0
0.4	12.50	0.2	25.0

1. To prepare 25 mL of a 0.8 mM solution from the 1.0 mM acetaminophen stock solution, pipet 20.0 mL of 1.0 mM stock solution into a 25 mL volumetric flask. Fill to the line with buffer and mix.

2. To prepare 25 mL of a 0.6 mM solution from the 0.8 mM acetaminophen solution, pipet 18.75 mL of 0.8 mM acetaminophen solution into a 25 mL volumetric flask. Fill to the line with buffer and mix.

3. To prepare 25 mL of a 0.4 mM solution from the 0.6 mM acetaminophen solution, pipet 16.67 mL of 0.6 mM acetaminophen solution into a 25 mL volumetric flask. Fill to the line with buffer and mix.

4. To prepare 25 mL of a 0.2 mM solution from the 0.4 mM acetaminophen solution, pipet 12.50 mL of 0.4 mM acetaminophen solution into a 25 mL volumetric flask. Fill to the line with buffer and mix.

DATA TABLE

Acetaminophen concentration in solution (mM)	Peak current (μA)
0.00	0.002
0.20	15.042
0.40	27.689
0.60	41.796
0.80	52.967
1.00	67.621
unknown	49.950

SAMPLE VOLTAMMOGRAMS

Figure 1 CV of 1.0 mM acetaminophen solution

Figure 2 CV of all acetaminophen standard solutions

Figure 3 Calibration curve of acetaminophen standards

ANSWERS TO ANALYSIS QUESTIONS

1. See Figure 3.

2. From Figure 3, the calibration curve is $y = 66.57x + 0.901$

   The observed peak current of the children's Tylenol sample is 35.004 µA. Using this value for x, the concentration of the unknown sample is calculated to be 0.512 mM.

3. The observed peak current is 67.621 µA. By definition, 1 ampere = 1 coulomb/second. Thus, the observed peak current can equivocally be written as 67.621 µC/s. With Faraday's constant, 96,485 C/mol, Avogadro's number, 6.022×10^{23} electrons/mol, and careful attention to units, the current flow is calculated to be 4.22×10^{14} electrons/s.

   67.621×10^{-6} C/s × 1/(96,485•C·mol^{-1}) × 6.022×10^{23} electrons/mol
   = 4.22×10^{14} electrons/s

   This is chemically equivalent to oxidizing 2.11×10^{14} molecules of acetaminophen per second (since two electrons are removed per acetaminophen molecule).

4. Answers will vary based upon experimental data and concentration of acetaminophen in the commercial medication solution. The first step is to convert the manufacturer label to units of molarity. Our sample data was collected with 160 mg acetaminophen per 5 mL.

   0.160 g × (1 mol / 151.163 g) × (1 / 0.005 L)
   = 0.211 mol/L

   We then weighed out 0.657 g of this 0.211 mol/L solution, which, using the density value of 1.31 g/mL, is equivalent to 0.501 mL of solution. This solution then underwent a few serial dilutions to make it in the appropriate measurement range:

   (0.501 mL)(0.212 mol/L) = (10 mL) M_2 M_2 = 0.0106 mol/L
   (1.0 mL)(0.0106 mol/L) = (10 mL) M_2 M_2 = 0.00106 mol/L
   (5.0 mL)(0.00106 mol/L) = (10 mL) M_2 M_2 = 0.000531 mol/L = 0.531 mM

   From question 2 above, we determined 0.512 mM as the experimental concentration of acetaminophen. Therefore, we can calculate the percent error:

   ((0.512 mM – 0.531 mM) / 0.531 mM) × 100%
   = 3.5% error

   Depending on the number of trials collected in this laboratory, this is an excellent opportunity to introduce some statistical analyses, such as confidence intervals.

EXTENSION

Figure 4 CV with varied scan rates of 1.0 mM acetaminophen standard

Sweep rate (mV/s)	Peak current (μA)
0	0.50
25	35.50
50	47.85
100	65.39
200	92.68
300	118.64

The relationship between sweep rate and peak current is described via the Randles-Sevcik equation:

$$i_{pc} = 2.69 \times 10^8\ n^{3/2}\ A\ D^{1/2}\ v^{1/2}\ C$$

where, i_{pc} = peak current in A, n = number of electrons involved, A = electrode area in m^2, D = diffusion coefficient in m^2/s, v = scan rate in V/s, and C = concentration in mol/L.

In Graphical Analysis app, students can graph the sweep rate *vs*. peak current and fit a power equation to the data to discover the square root relationship as shown in Figure 5.

Figure 5 Sweep rate vs. *peak current with power equation fit of acetaminophen*