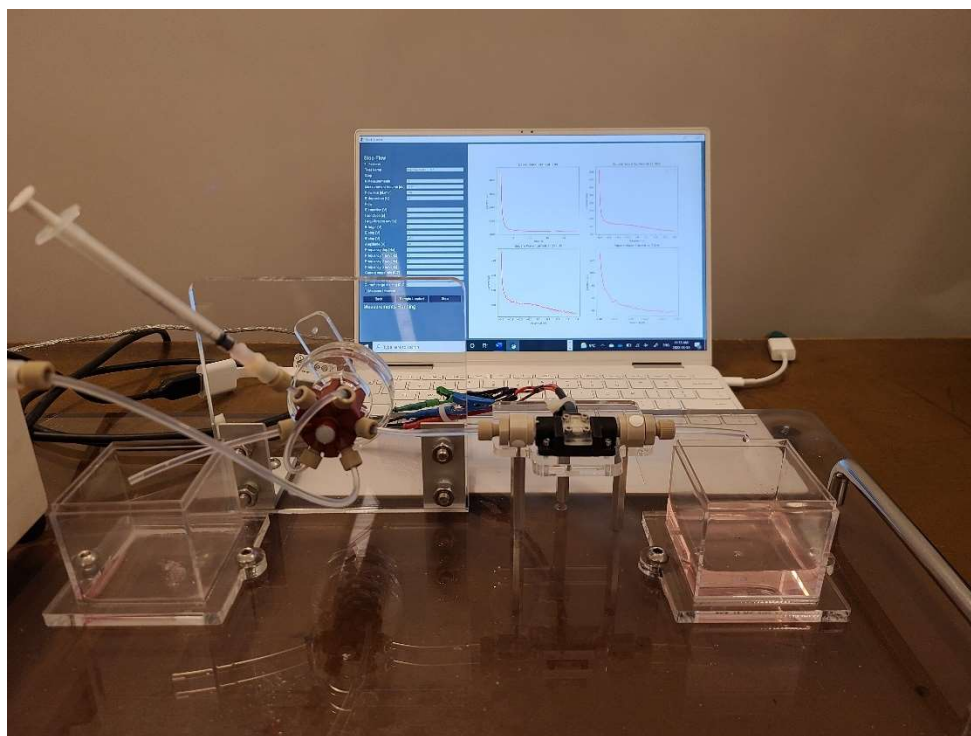


Portable Electrochemical Fentanyl Quantification Device



Project Team

Lewis Arnold
Mykal Bakker-Westeinde
Nicholas Bratvold
Holden Jones

Project Sponsors

Dr. Dan Bizzotto, Professor, UBC Chemistry
Dr. Glenn Sammis, Professor, UBC Chemistry
Dr. David Weekes, Business Innovation Manager, Stewart Blusson Institute

Project 2200
Engineering Physics 459
Engineering Physics Project Laboratory
The University of British Columbia
April 8, 2022

Executive Summary

Thousands of British Columbians are dying every year due to illicit drug overdoses and the annual death toll is increasing each year (BC Coroners Service, 2022). Chief among the causes of this worsening epidemic is fentanyl, a synthetic opioid that is extremely potent and can be up to one hundred times stronger than morphine (Juergens & Parisi, 2021). Because of its potency, fentanyl is detected in over 80% of illicit drug overdoses in British Columbia (BC Coroners Service, 2022).

In-the-field drug testing seeks to reduce the number of overdose deaths by providing information to drug users about the composition of their drugs before use. However, since fentanyl is highly addictive, many users will use a drug sample even if they know fentanyl is present. Therefore, it is essential to provide a quantitative measure of the amount of fentanyl in a drug sample.

Professor Dan Bizzotto and Professor Glenn Sammis of the University of British Columbia (UBC) Chemistry department have created an electrochemistry technique to precisely measure the concentration of fentanyl in street drug samples. They are working to implement this technique in a fast, portable and inexpensive device that can be deployed in drug testing applications.

Our team has developed a testing apparatus to enable validation and refinement of the chemical processes. We have created a custom flow cell where the drug sample can be analyzed and have automated the system control to make testing more efficient. We have also designed and built a portable physical platform that ensures repeatable, consistent testing. Our apparatus has achieved encouraging preliminary results for basic electrochemical measurements that are essential to the operation of the system. Crucially, our tests showed that the system can produce quantitative results with a test compound.

To promote further development, we recommend more testing to determine the limit of detection for fentanyl and to study the effect of contamination across measurements.

Table of Contents

Executive Summary.....	ii
Table of Contents.....	iii
Table of Figures.....	vi
Table of Equations	vii
1 Introduction	1
1.1 Sponsor Information	1
1.2 Background.....	1
1.3 Problem Statement	2
1.4 Scope and Limitations	3
2 Discussion.....	4
2.1 Theory.....	4
2.1.1 Sample Preparation and Internal Standard	4
2.1.2 Fluid Train and 6-Way Valve	5
2.1.3 Syringe Pump	6
2.1.4 Separation	6
2.1.5 Electrochemical Analysis.....	7
2.1.6 Data Analysis and Concentration Calculation.....	10
2.1.7 System Operation	11
2.2 Design, Approach and Method	12
2.2.1 Flow Cell.....	12
2.2.2 Graphical User Interface (GUI) and System Control.....	22
2.2.3 Physical Platform	23
2.3 Testing and Results.....	24

2.3.1	Flow Cell Leak Testing	24
2.3.2	Material Compatibility Testing	24
2.3.3	System Control Software Testing.....	25
2.3.4	Full System Electrochemistry Measurements	25
3	Conclusions	28
4	Recommendations	29
4.1	Accurately Quantify Fentanyl	30
4.2	Find the Lower Detection Limit	30
4.3	Prevent Contamination Across Measurements	30
4.4	Characterize and Prevent Air Bubbles.....	30
4.5	Design Custom Electrodes.....	31
5	Deliverables.....	31
	References	32
	Appendices.....	34
A.	Internal Standard.....	34
B.	Separation Column	35
C.	Flow Cell and 6-Way Valve Mounting	36
D.	Testing and Results.....	37
	Flow Cell Leak Testing	37
	Full System Electrochemistry Measurements	38
E.	Engineering Recommendations	39
	Toggle Clamp for Flow Cell	39
	Software	40
	System Control from a Raspberry Pi	41

F. User Guides	42
Flow Cell	42
Software Installation	44
Quick Start.....	46
Saving Data.....	49
Adjusting the Code.....	51
Setting Up Raspberry Pi for Remote Connection	53
G. Known Bugs and Issues	55

Table of Figures

Figure 1: System Diagram	4
Figure 2: 6-Way Valve Load and Inject Positions.....	5
Figure 3: Syringe Pump	6
Figure 4: Separation Column	6
Figure 5: Three-Electrode System.....	8
Figure 6: Electrode Chip used in Our System.....	9
Figure 7: System Diagram	11
Figure 8: Our Custom Flow Cell Design.....	13
Figure 9: Installation of the Electrode Chip in the Flow Cell	13
Figure 10: Position of the Gasket in the Flow Cell	14
Figure 11: Installation of the Lid on the Flow Cell	14
Figure 12: O-Ring Flow Cell Design Available for Purchase	15
Figure 13: Geometry of the Electrode Chip that Prohibits O-Ring Use	16
Figure 14: Sealing Mechanism of our Flow Cell.....	17
Figure 15: Function of the Hard Stops for the Flow Cell Lid	18
Figure 16: Shape of the Fluid Chamber	19
Figure 17: Inlet Hole that Dictates Fluid Chamber Depth.....	20
Figure 18: Flow Cell for Two Electrode Chips	21
Figure 19: Cut-outs to Hold Nuts	21
Figure 20: Our GUI Measurement Control Screen	22
Figure 21: Physical Layout Overview	23
Figure 22: Single Chronoamperometry Measurement.....	26
Figure 23: Current vs. Voltage Curves for a Stop-Flow Measurement.....	27
Figure 24: 3D Plot of Current vs. Voltage vs. Sweep Number for a Stop-Flow Measurement.....	28
Figure 25: Process by which a Mixed Sample is Separated in a Separation Column	35
Figure 26: Flow Cell Mounting.....	36
Figure 27: 6-Way Valve Mounting	37
Figure 28: Comparison of a Small Toggle Clamp Arm to a Larger One.....	40

Figure 29: Flow Cell Components	42
Figure 30: Electrode Chip Installation	43
Figure 31: Gasket Installation	43
Figure 32: Lid Installation	44
Figure 33: Installation of the Check Valves	44
Figure 34: Final Flow Cell Assembly	44
Figure 35: Checking Python Installation	45
Figure 36: Navigating to an Installation Location	46
Figure 37: GUI Screen for Test Selection and Port Configuration	46
Figure 38: GUI System Parameters Screen	47
Figure 39: Deposition Current Shown During the Initial Flow Through Phase	48
Figure 40: Button to Push When Injecting Sample	48
Figure 41: Example of Data Plotted During a Test	48
Figure 42: Example of a Configuration File	49
Figure 43: Example of a “measurement#.csv” File	50
Figure 44: Example of an “injecttime_total.csv” File	50
Figure 45 Example of Default Values	51
Figure 46: Initializing Serial Connection to Syringe Pump	52
Figure 47: Serial Write Function	52
Figure 48: Serial Read Function	53
Figure 49: Setting Up a Raspberry Pi	54
Figure 50: Installing VNC Viewer	54

Table of Equations

Equation 1: Fentanyl Oxidation above 0.8 V	7
Equation 2: Norfentanyl Reduction	7

1 Introduction

1.1 Sponsor Information

This project is sponsored by Dr. Dan Bizzotto and Dr. Glen Sammis, two Professors in the UBC Department of Chemistry. Dr. David Weekes, Business Innovation Manager at the Stewart Blusson Institute at UBC, is also supporting the product development.

Dr. Bizzotto and Dr. Sammis have worked together for three years to develop an electrochemical method to quantify the fentanyl concentration of a drug sample. This chemical technique constitutes the theoretical basis for this project.

1.2 Background

The opioid epidemic is a worsening health emergency in British Columbia that is claiming thousands of lives each year. The 2232 illicit drug overdose deaths that occurred in 2021 represent a 650% increase compared to ten years ago (BC Coroners Service, 2022).

The replacement of heroin and cocaine by fentanyl in street drugs is a driving factor of this worsening epidemic. As fentanyl is extremely potent, small variations in its concentration can be the difference between life and death.

Drug testing is a part of the solution to reduce the number of overdoses by informing drug users about the composition of their drugs before use. According to a 2019 report by Yau et al., there are currently two major options for fentanyl detection: immunoassay strips and infrared spectroscopy.

Immunoassay strips can detect the presence of fentanyl in drug samples before ingestion. The result is a coloured line that indicates if fentanyl is present; there is no quantitative information indicating the amount of fentanyl present (Yau et al., 2019). Since fentanyl is often present in

drug samples, this binary result provides no insight to users who already expect fentanyl to be present.

Fourier-Transform Infrared Spectroscopy (FTIR) systems provide a quantitative measure of the amount of fentanyl in a drug sample. The FTIR system measures a drug sample's absorbed infrared spectrum, which requires an expert technician to interpret and quantify the amount of fentanyl in the sample. Although quantitative, these results have a 28% false-negative rate (Yau et al., 2019).

Binary strips do not provide quantitative results and FTIR systems do not provide reliable or easily interpretable results. Our sponsors have developed the chemical basis for a system that will satisfy both these needs. The method uses an electrochemical technique to provide fast, quantitative measures of fentanyl in drug samples. Our sponsors have shown the technique is capable of a detection limit 25 times better than that of FTIR (Bizzotto & Sammis, 2020). The method implements separation techniques to ensure quality measurements when contaminants are present, as is often the case in street drugs.

Having proven that the underlying chemistry allows for a reliable quantitative measurement, our sponsors are developing a device that can be deployed in real applications. The first step, and the focus of our project, is to develop a prototype to refine the chemistry and system design.

1.3 Problem Statement

Our project goal has been to provide our sponsors with a robust data acquisition system that can be used to improve the quantification method. The system automates the manual routine that our sponsors are currently performing to acquire data. As a high-level summary, our goals have been to implement:

- automatic control of the sample flow through the system.
- a leak-free fluid path, including a flow cell where electrochemical reactions take place.

- automatic control of measurement electronics.
- a graphical user interface (GUI) to control the system.

1.4 Scope and Limitations

Our sponsors' final goal is to deploy a commercial product to be operated by an individual without expert chemistry knowledge. The ideal system has a single-use fluid path to reduce contamination between samples and an integrated analytics app to convert raw data to a meaningful fentanyl concentration. As a first step towards this long-term goal, we focused on creating a development system to be used by our sponsors.

Our project's scope centered on designing a robust system without any hidden features, such that it could be used to run a wide variety of electrochemical measurements. Specific design objectives included developing a low-volume electrochemical flow cell and a simple graphical user interface (GUI) to interface with the system hardware and record measurement data. Our scope also included the automation of the system.

Developing a single-use fluid path and an analytics app were beyond our scope, as our sponsors can clean the system and manage raw data as needed. Likewise, data collection and analytics beyond what was required for basic system testing were not part of the scope.

Ultimately, we delivered two flow cell designs and a working GUI that automates the critical system components. We also investigated, developed and partially implemented the automation for secondary system components, and gathered recommendations for further development based on our work.

2 Discussion

2.1 Theory

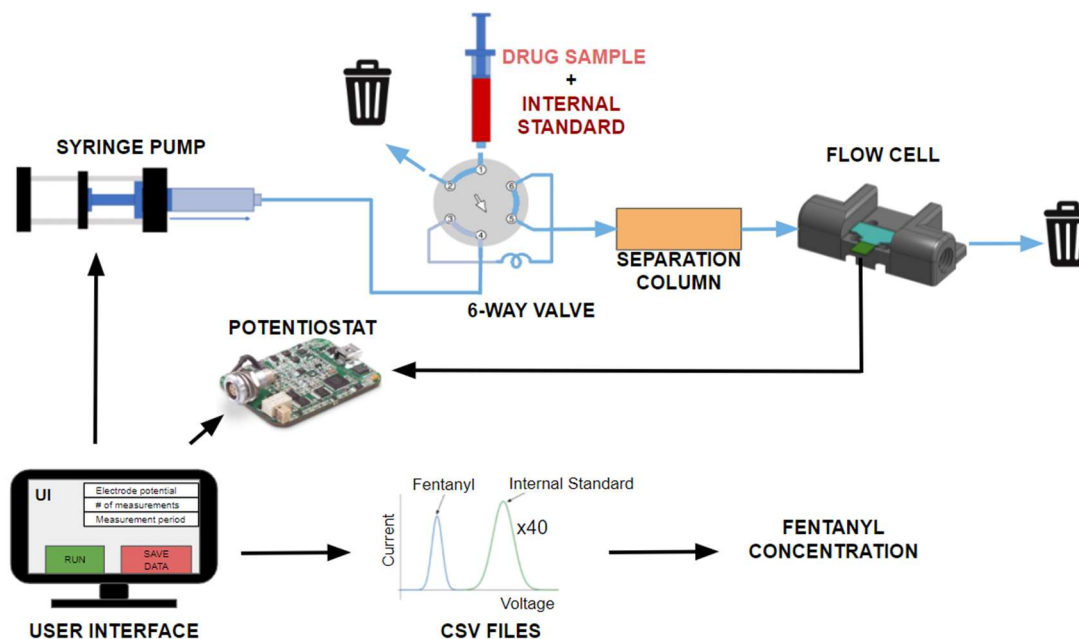


Figure 1: System Diagram¹

A diagram of the major components and connections in our system is shown in Figure 1. We describe the major steps and components in the next subsections.

2.1.1 Sample Preparation and Internal Standard

Our system requires the drug sample to be in aqueous form. To begin, the operator draws the drug sample into a syringe and mixes it with a fixed quantity of an internal standard, a chemical of known concentration designed to calibrate the system. Please refer to [Internal Standard](#) in the appendix for more details.

¹ Potentiostat photograph: PalmSens, n.d.

6-way valve diagram: modified from Valco Instruments Company Inc., n.d.

Trash icon: VectorStock, n.d.

2.1.2 Fluid Train and 6-Way Valve

To move the drug sample through the system, it must be pushed by a carrier fluid, which can be any inert substance that does not confound the measurements. We employ a 6-way valve to inject the sample in the carrier fluid.

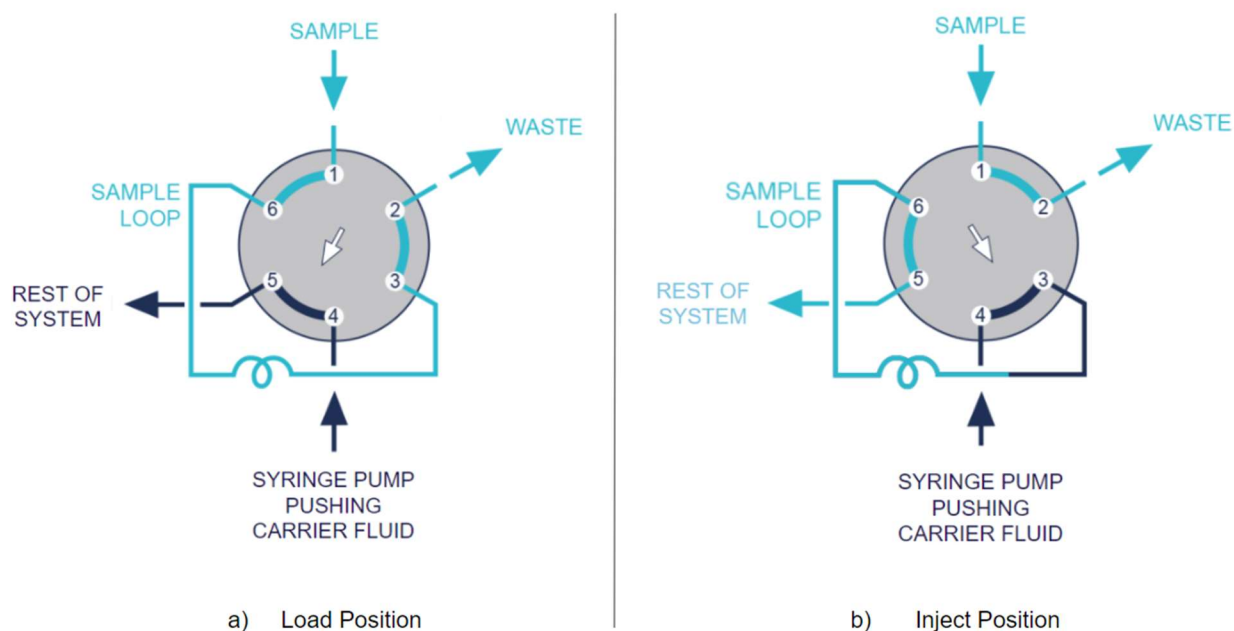


Figure 2: 6-Way Valve Load and Inject Positions²

Shown in Figure 2, the valve has six fluid ports that can be connected in two different ways. In the load position (Figure 2 a)), the syringe pump pushes carrier fluid through the 6-way valve directly to the remainder of the system. In the inject position (Figure 2 b)), the sample loop is part of the fluid path. To introduce the drug sample and internal standard mixture to our system, the operator loads it into the sample loop and then turns the knob to the inject position. Our carrier fluid then drives the sample through the rest of our system.

² Modified from Advion Interchim Scientific, n.d.

2.1.3 Syringe Pump

The syringe pump pushes a linear stage and compresses a syringe at a constant rate. We are using a production syringe pump that our sponsor provided as seen in Figure 3.



Figure 3: Syringe Pump

2.1.4 Separation

Street drug samples often contain a mixture of drugs and cutting agents. The presence of multiple substances can confound the electrochemical measurements. Therefore, before the drug sample can undergo analysis, it must be separated into its components. This is achieved using a separation column, shown in Figure 4. For a more detailed discussion of the separation column, please refer to [Separation Column](#) in the appendix.



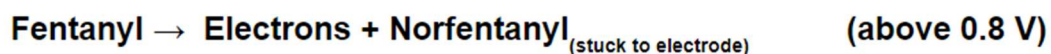
Figure 4: Separation Column

2.1.5 Electrochemical Analysis

Next, the sample is pushed into the flow cell by the syringe pump. Since the sample's components have been separated in time, there is only one component in the flow cell at a time. The electrochemical detection occurs in the flow cell to measure each component and is explained in the following sections.

Fentanyl Reactions

Fentanyl undergoes two specific reactions, oxidation and reduction, that our sponsors use to detect its presence and concentration. These reactions are commonly known as redox reactions.



Equation 1: Fentanyl Oxidation above 0.8 V

First, fentanyl undergoes an oxidation if the voltage is raised above 0.8 V. The simplified equation for this reaction is shown in Equation 1. The product, norfentanyl, is bound to the electrode after the reaction.



Equation 2: Norfentanyl Reduction

When norfentanyl has been captured on the electrode, it will undergo a reduction if the voltage is sufficiently decreased. The simplified reaction is shown in Equation 2.

Three-Electrode System and Potentiostat

We can measure the total number of electrons produced or consumed by these two reactions, and thus the total amount of fentanyl present, by measuring the current in the fluid as these reactions occur. We do this using a three-electrode system and a potentiostat.

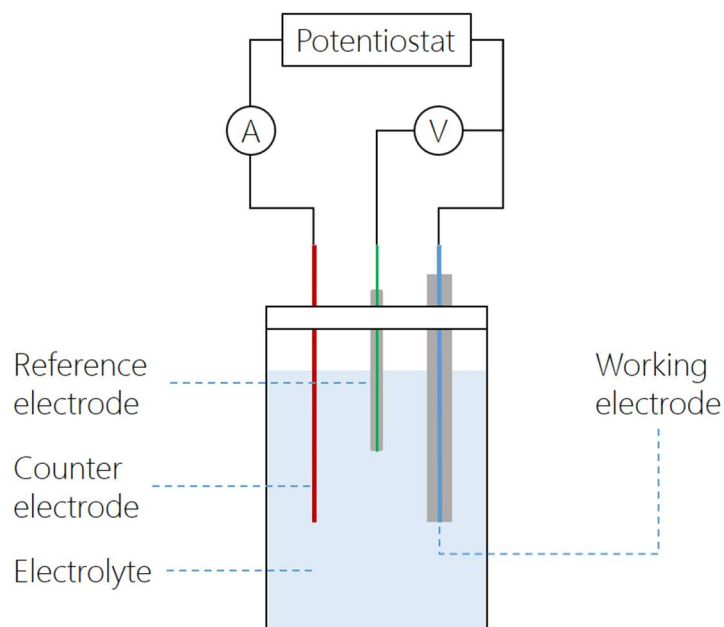


Figure 5: Three-Electrode System³

As shown in Figure 5, the three-electrode system consists of a working electrode, a counter electrode, and a reference electrode. The potentiostat measures the current flowing between the working and counter electrodes as a function of the applied voltage and as a function of time. In our system, we are using the EmStat3 potentiostat from PalmSens.

Our sponsors have selected a specific electrode chip that combines all three electrodes into one package.

³ Fundamental Electrochemistry, 2020

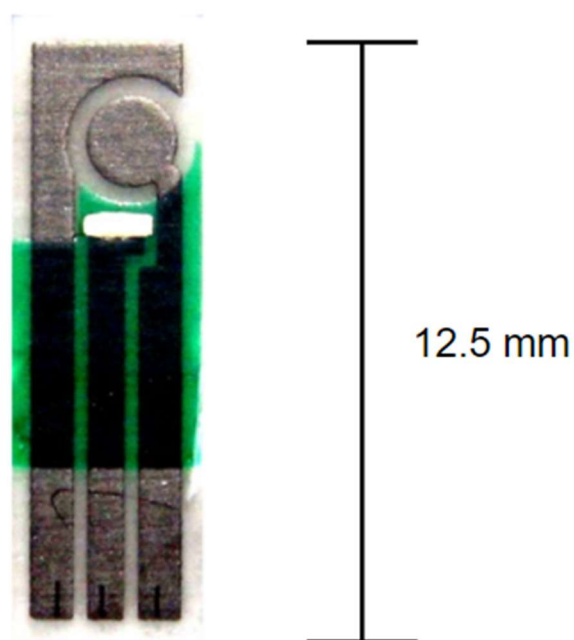


Figure 6: Electrode Chip used in Our System⁴

The electrode chip is shown in Figure 6. The active area of the electrodes is finished with carbon and cannot be touched. In the middle, green dielectric material insulates the electrodes. At the bottom, there are three contacts to connect the electrodes to the potentiostat. Our sponsors prefer this electrode chip because of its low cost and high accuracy.

Measurement Techniques

There are several different methods of catalyzing these redox reactions at different potentials. Our system can conduct chronoamperometry, voltammetry and stop-flow measurements. Chronoamperometry consists of setting a fixed potential across the electrodes and measuring the current over time. In voltammetry, the potential is decreased from 0.8 V to -0.4 V and current is measured as a function of potential. Instead of linearly decreasing the voltage, it is possible to sweep the voltage while superimposing small, square-wave oscillations. This technique, known as square wave voltammetry (SWV), allows for increased accuracy.

⁴ Modified from BioDevice Technology, Ltd., n.d.

Stop-Flow

Chronoamperometry and SWV can be combined into a repeated stop-flow sequence for targeted detection of fentanyl. First, the fluid train is pushed to fill the flow cell while chronoamperometry is executed, causing the oxidation reaction shown in Equation 1. Then, the fluid flow is stopped and voltammetry is executed, causing the reduction reaction in Equation 2. Cycles are repeated until the whole fluid train has moved through the flow cell into waste. To conduct these measurements, the syringe pump and potentiostat must be coordinated. We accomplish this by using a software control program, which we discuss in [Graphical User Interface \(GUI\) and System Control](#) below.

2.1.6 Data Analysis and Concentration Calculation

We can measure the total charge produced from the fentanyl redox reactions by integrating the current-time data from the chronoamperometry tests and by calculating the peak heights in the current-voltage data from the voltammetry. We use the same technique to measure the concentration of the internal standard, and then scale the measured fentanyl concentrations appropriately.

2.1.7 System Operation

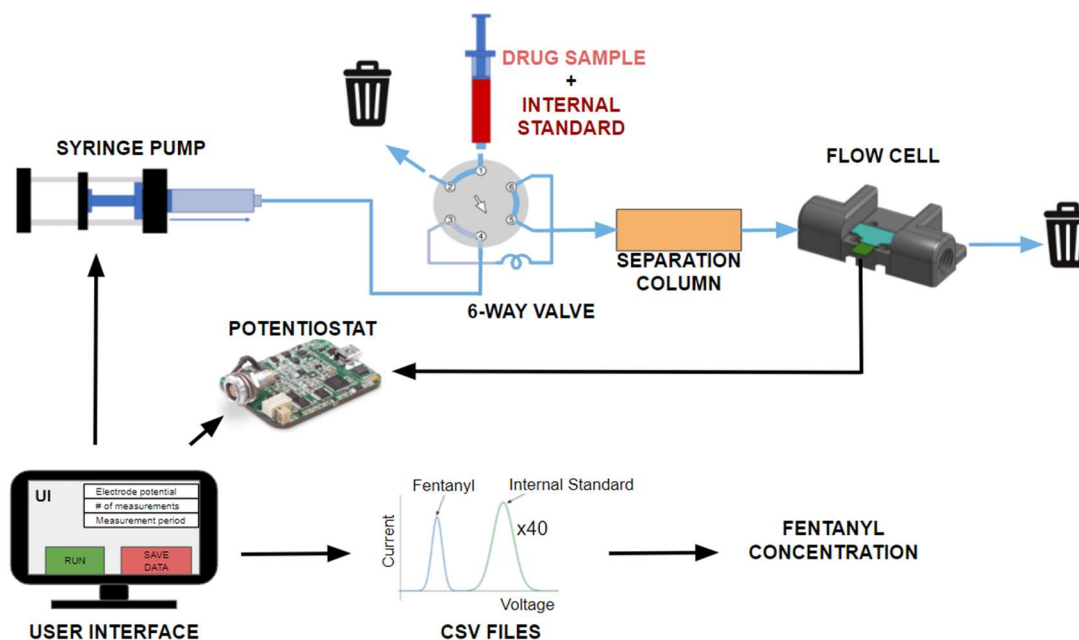


Figure 7: System Diagram⁵

Now that the system components and theory have been discussed in detail, we return to the system diagram to list the steps of the system operation.

1. To prime the system, the system control program runs the syringe pump, pushing carrier fluid through the entire system and removing air bubbles.
2. The system operator draws the drug sample and a known amount of internal standard into a syringe.
3. The operator injects the drug sample into the 6-way valve while it is in the load position.
4. The operator turns the 6-way valve to the inject position to put the sample into the fluid train.
5. The system control program runs the syringe pump to push the fluid train through the separation column, separating the constituent substances in time.

⁵ Potentiostat photograph: PalmSens, n.d.

6-way valve diagram: modified from Valco Instruments Company Inc., n.d.

Trash icon: VectorStock, n.d.

6. The constituent substances arrive in the flow cell one at a time. While the substance is flowing into the flow cell, the system control program directs the potentiostat to perform chronoamperometry.
7. The system control program stops the fluid flow and executes square wave voltammetry while the substance is in the flow cell.
8. Steps 5 and 6 are repeated until the entire fluid train has passed through the flow cell.
9. The system control program saves the data for later analysis which determines the concentrations of the constituent substances, including fentanyl.

2.2 Design, Approach and Method

2.2.1 Flow Cell

The flow cell was a major component of our engineering work. Below, we describe the purpose and requirements of the flow cell and then review its design.

Purpose and Requirements

The purpose of the flow cell is to pass the sample over the electrodes to perform electrochemical measurements. The flow cell must have a very small volume (approximately 50 μL) to improve the accuracy of the system. The flow cell must also prevent leaks to ensure that drug samples do not exit the system in an uncontrolled fashion. Finally, it is important that fluid cannot be trapped in the flow cell and linger across measurements; all fluid must be completely replaced during flow.

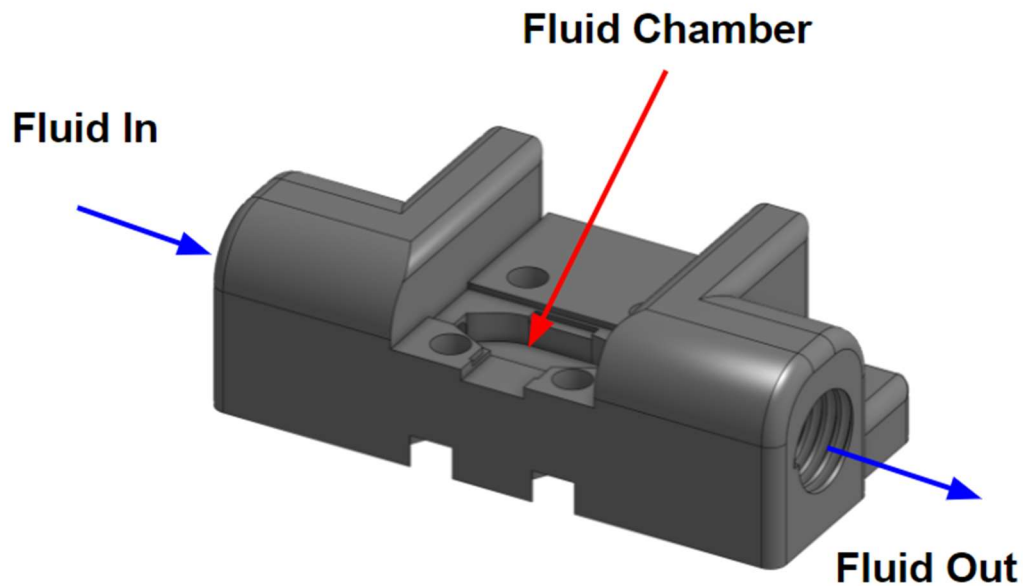


Figure 8: Our Custom Flow Cell Design

The basic design of our flow cell is shown in Figure 8. On each end of the flow cell, there is a threaded connection that allows the flow cell to be installed in the fluid path. In the middle of the device, there is a fluid chamber where the sample can interact with the active parts of the electrode chip.

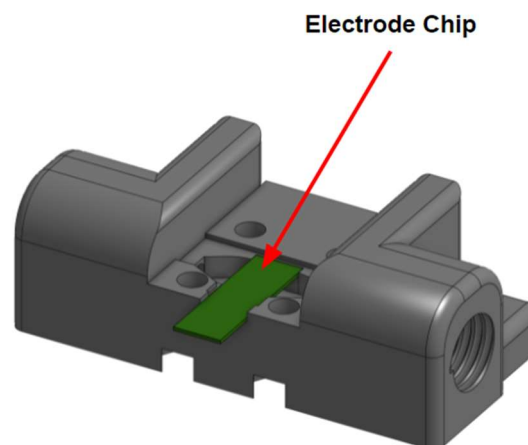


Figure 9: Installation of the Electrode Chip in the Flow Cell

Figure 9 shows how the electrode chip is installed in the flow cell. The electrode chip fits into a small slot and is oriented so that the active parts of the electrodes are facing down into the fluid chamber.

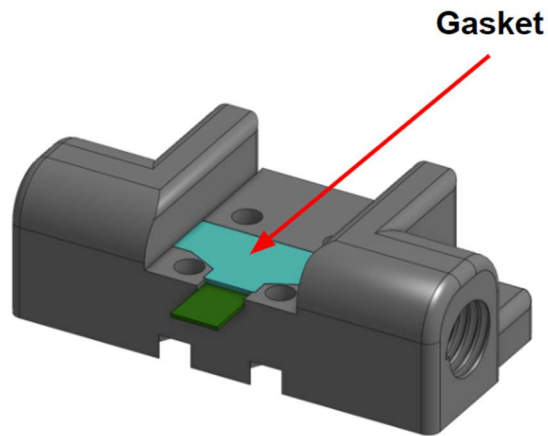


Figure 10: Position of the Gasket in the Flow Cell

Figure 10 shows how the gasket is installed in the flow cell. It sits on top of the electrode chip and extends around the edges of the fluid chamber.

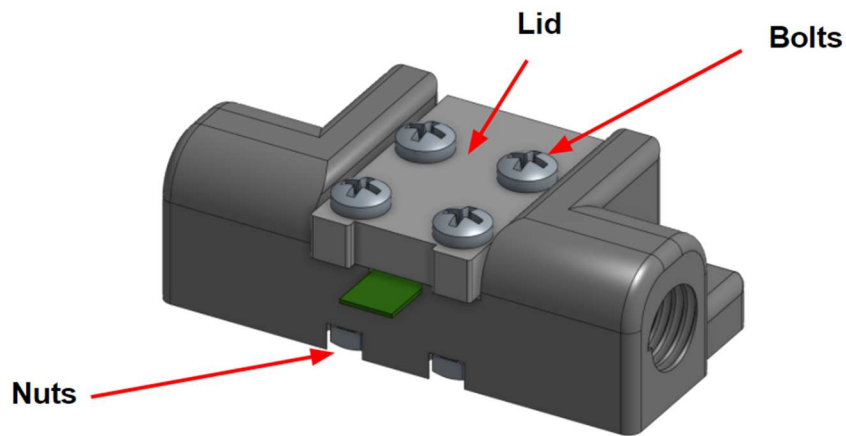


Figure 11: Installation of the Lid on the Flow Cell

In Figure 11, we see how the lid is installed on top of the flow cell. Four bolts pass through the body of the flow cell and are tightened into four nuts on the bottom. This compresses the gasket and ensures a good seal is made.

Design Alternatives

Before we developed this design, we investigated industry alternatives. We observed that many industry designs use an O-ring to seal on the surface of the electrode chip, such as in Figure 12.



Figure 12: O-Ring Flow Cell Design Available for Purchase⁶

The principal advantage of this design is that there is only one surface which must be sealed: the surface where the O-ring meets the face of the electrode. However, due to the geometry of our electrode chips, this design is not possible, and we did not pursue it.

⁶ Metrohm, n.d.

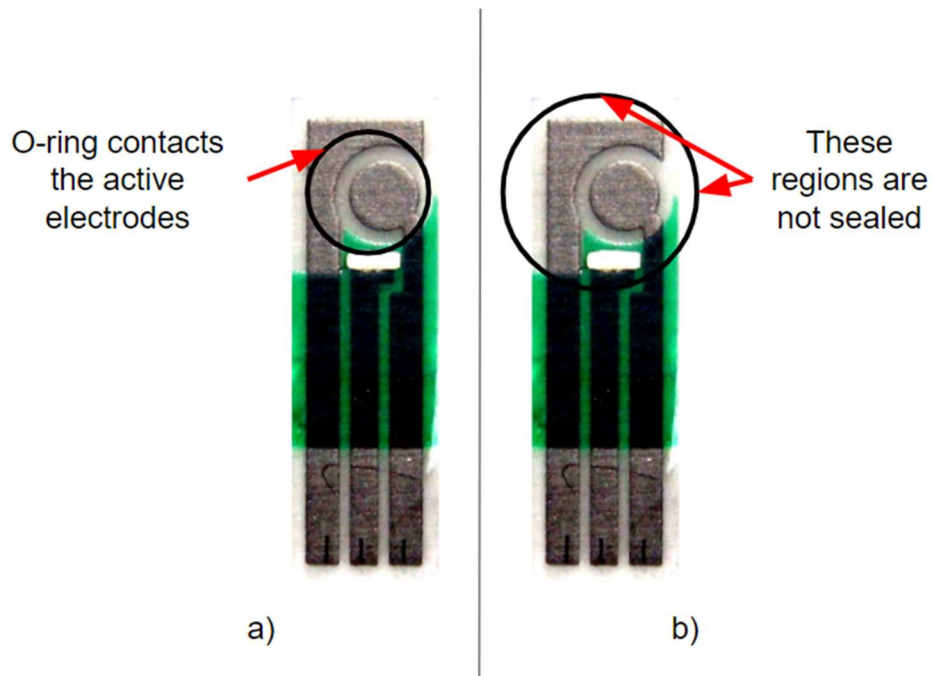


Figure 13: Geometry of the Electrode Chip that Prohibits O-Ring Use⁷

The reason an O-ring is not suitable for our electrode chips can be seen in Figure 13. To ensure correct measurements, the O-ring must not touch the active part of the electrodes. Therefore, the layout shown in Figure 13 a) where the black O-ring touches the electrodes is not acceptable. However, using a larger O-ring causes another issue. Since the active area of the electrodes extends to the edge of the chip, a larger O-ring would extend beyond the edges of the chip as seen in Figure 13 b). This would not create an effective seal due to the gaps beside the electrode chip where fluid could escape.

Instead of using an O-ring design, we developed a custom design that uses a different sealing method. A cross-section of the sealing mechanism is shown in Figure 14.

⁷ Modified from BioDevice Technology, Ltd., n.d.

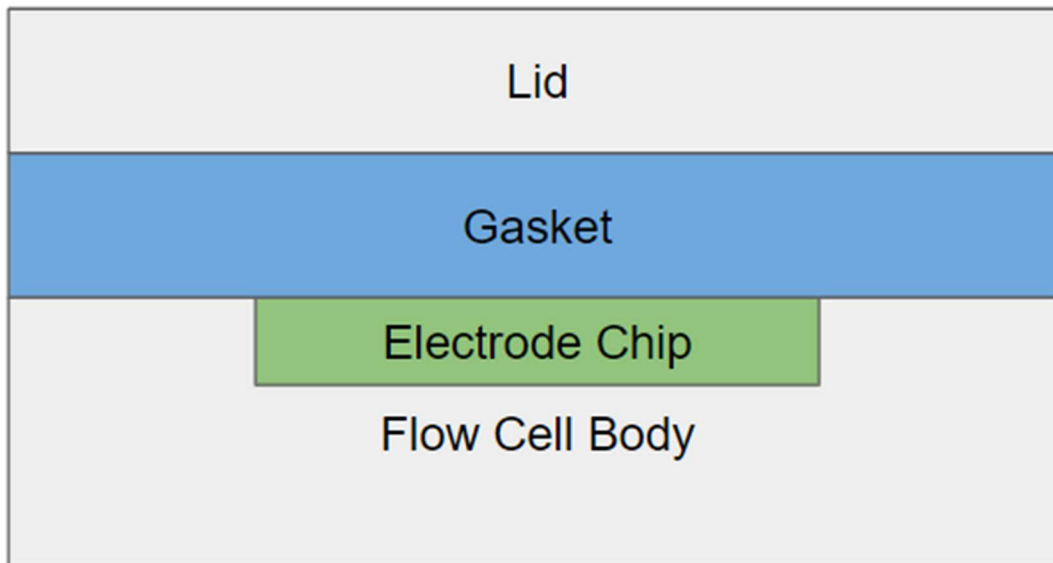


Figure 14: Sealing Mechanism of our Flow Cell

There is a recess in the body of the flow cell for the electrode chip. A gasket sits on top of the electrode. Finally, a lid sits on top of the gasket, forming the top surface. When the whole stack is compressed, the gasket forms a seal around the top of the electrode and the bottom surface beside the electrode.

The major risk of this design is that the electrode must form its own seal against the sides and bottom of its slot in the flow cell body, where there is no gasket. However, after testing multiple iterations of the flow cell, we confirmed that this method ensures adequate sealing. These tests are described in [Flow Cell Leak Testing](#) in the appendix.

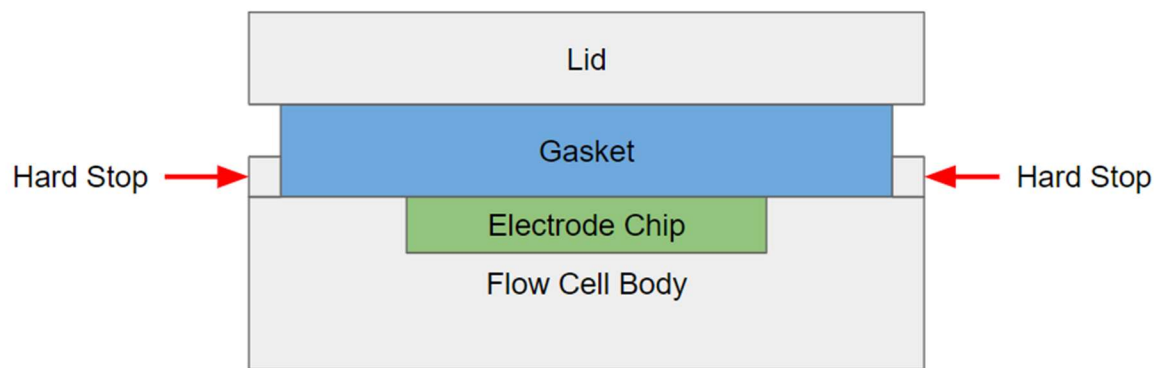
Gasket Material

We evaluated samples of PTFE and Buna-N gasket materials to determine which was more suitable for our application. We selected a 1/32" Buna-N gasket because it is softer and therefore allows for more compression and a better seal.

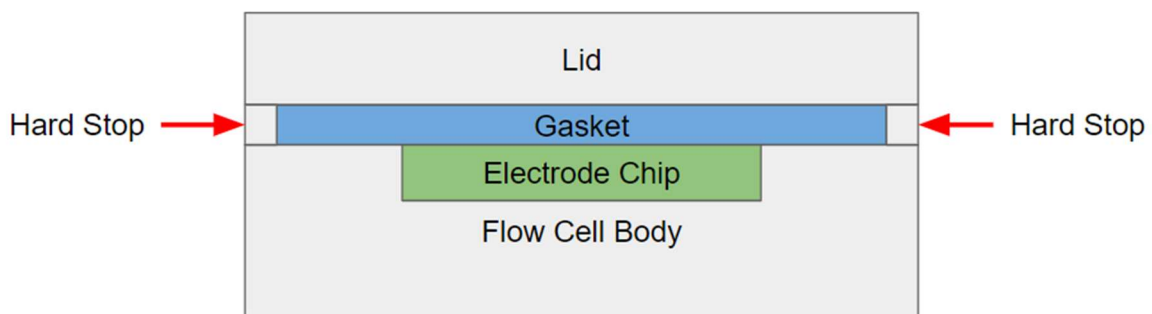
Hard Stops: Purpose, Location and Height

Since the gasket is compressed by tightening bolts, there can be variability in the amount the bolts are tightened and therefore the amount by which the gasket is compressed. To ensure repeatability and consistency across runs, it is important that the flow cell be in the same state every time it is assembled.

To achieve the desired consistency, we implemented hard stops for the lid.



a) Uncompressed State



b) Compressed State

Figure 15: Function of the Hard Stops for the Flow Cell Lid

As shown in Figure 15, the hard stops are extensions above the flow cell body. As the lid is tightened down to compress the gasket, it runs into the hard stops. Any further tightening of the bolts will not cause the gasket to be further compressed since the lid cannot move down further.

Fluid Chamber Shape and Depth

The shape of the fluid chamber was designed to prevent fluid from being retained across measurements.

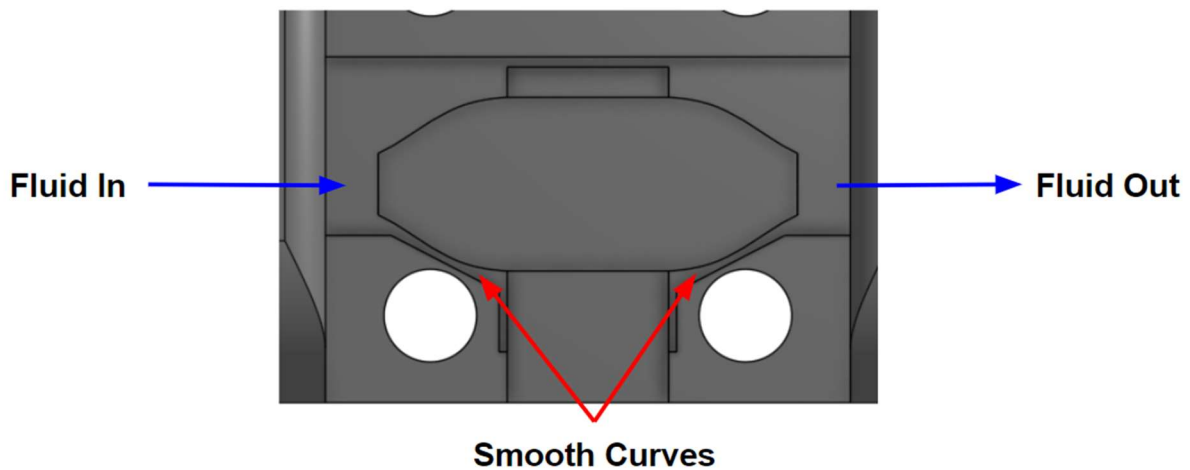


Figure 16: Shape of the Fluid Chamber

Figure 16 shows the shape of the fluid chamber from above. The smooth curves at the entry and exit of the chamber prevent contamination across measurements by ensuring that no fluid is trapped.

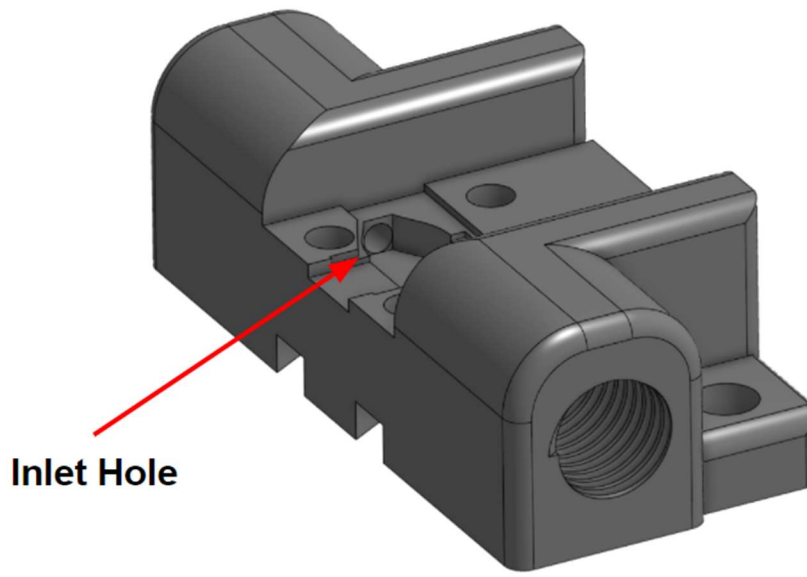


Figure 17: Inlet Hole that Dictates Fluid Chamber Depth

The fluid chamber's minimum depth is governed by the diameter of the inlet hole, which must match the fluidic connector used to connect the flow cell to the remainder of the system. As seen in Figure 17, we set the depth of the fluid chamber to be nearly as small as the inlet hole diameter while maintaining 0.3 mm wall thickness. The resulting fluid chamber volume is 67 μL .

Two-Electrode Flow Cell

To allow for more complex measurements, our sponsors requested a flow cell that could hold two electrode chips simultaneously. We modified our original flow cell to include a second slot for another electrode chip.

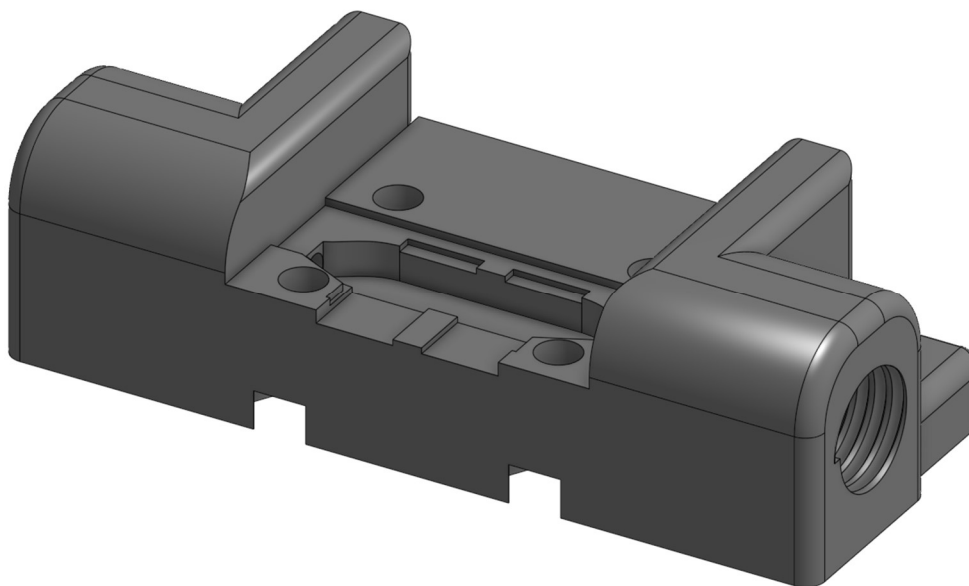


Figure 18: Flow Cell for Two Electrode Chips

Figure 18 shows the flow cell body that can accommodate two electrode chips. The design is very similar to the flow cell for one electrode chip. This flow cell has a volume of 110 μL .

Cut-outs for Nuts

To allow for easier assembly, we designed cut-outs on the bottom of the flow cell that hold the nuts.

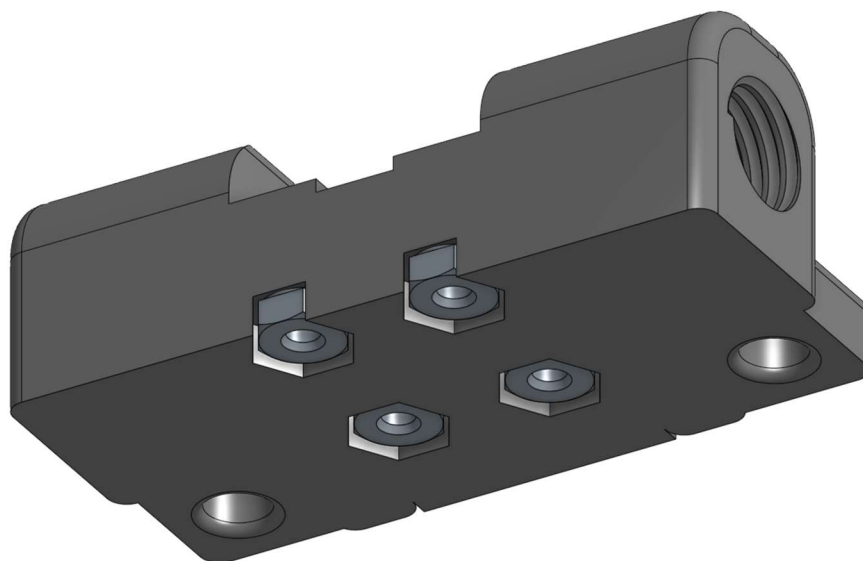


Figure 19: Cut-outs to Hold Nuts

Figure 19 shows the cut-outs in the bottom of the flow cell body. The tight fit of the cut-outs holds the nuts tightly and prevents them from spinning. This allows the bolts to be installed from the top with one hex key and relieves the need for an additional wrench.

2.2.2 Graphical User Interface (GUI) and System Control

The second major focus of this project was to develop a simple and intuitive control interface to synchronize the syringe pump and potentiostat, allowing the system to run both the constant-flow chronoamperometry and the stop-flow voltammetry previously discussed in [Measurement Techniques](#).

Operation

The user selects the parameters of the test to run and starts the test. Our program then communicates with the pump and potentiostat through a serial connection and produces a file with the current and voltage measurement data. A guide on how to complete a measurement with our script is in the [Quick Start](#) user guide of the appendix.

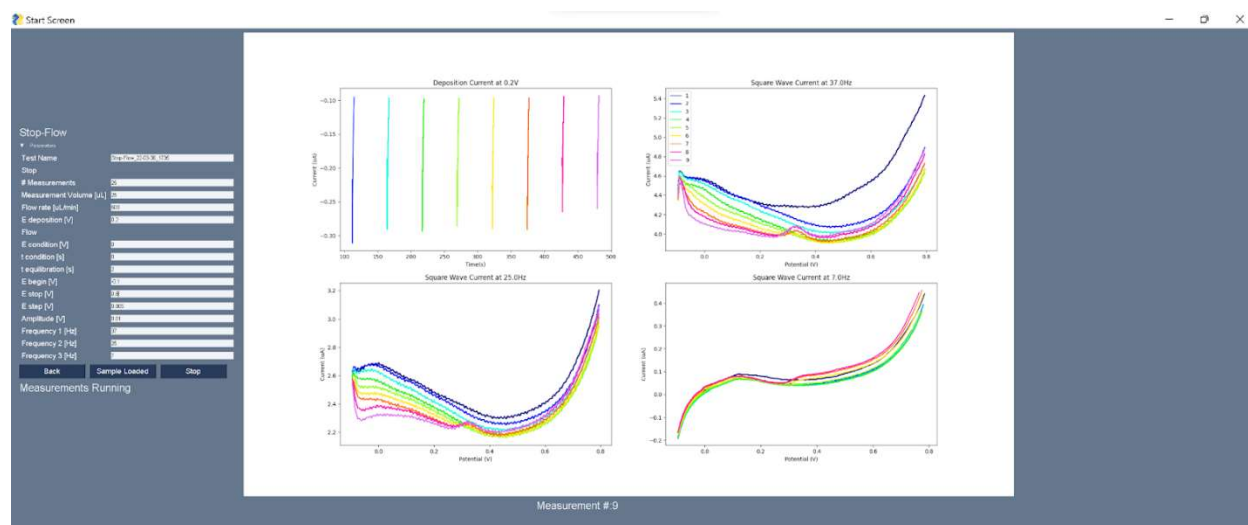


Figure 20: Our GUI Measurement Control Screen

Figure 20 shows the measurement control screen. Once the test is started, the potentiostat readings appear in real time on the graphs. We run the potentiostat and syringe pump

functions concurrently to allow for real-time plotting and better user experience. The user can iteratively modify test parameters to create the optimal fentanyl quantification test.

2.2.3 Physical Platform

Our first prototype was built using a plywood base to allow for easy removal of each component. Once the layout was finalized, we developed a portable and robust platform using a Lexan base, shown in Figure 21. We chose Lexan as it is non-absorbent, easy to waterjet cut, and rigid enough to carry the entire system. We installed a handle on each side of the setup to make it easily portable by one person.

Overview

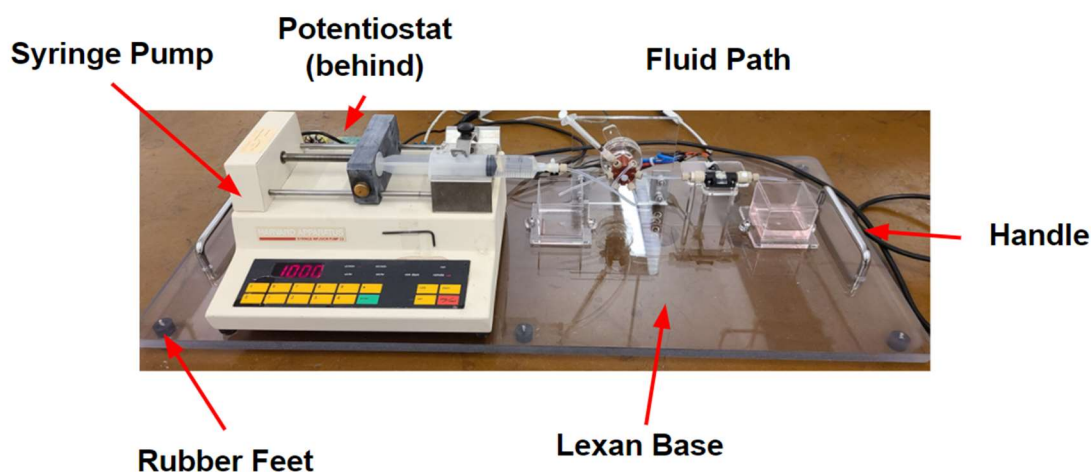


Figure 21: Physical Layout Overview

The layout is designed so that there is a logical progression: the sample passes from left to right. This design also ensures that the non-fluidic components are separated from the fluid path to minimize the risk of a spill.

For more details about the component mounting, please refer to [Flow Cell and 6-Way Valve Mounting](#) in the appendix.

2.3 Testing and Results

Our final product is a working data acquisition system capable of chronoamperometry and square wave voltammetry measurements. We designed a fully sealed electrochemical flow cell and graphical user interface (GUI) to automate both measurement routines. We integrated our design with off-the-shelf components (syringe pump, separation column, 6-way valve and potentiostat) to assemble the final working system.

To evaluate our system, first we assessed our flow cell and materials individually. We then used a comprehensive approach to test the system control and electrochemistry once the individual components were integrated.

2.3.1 Flow Cell Leak Testing

We invested considerable time testing for leaks since it is the most fundamental requirement for the flow cell. Following the testing process in [Flow Cell Leak Testing](#) in the appendix, flow rates of 16 mL/min did not produce a leak. Since the fentanyl detection measurements run at a much lower flow rate of 100-1000 $\mu\text{L}/\text{min}$, this is a strong indication that leaks will not be an issue.

2.3.2 Material Compatibility Testing

Methanol is a possible cleaning agent or mobile phase in the final system's separation column. Since it is an organic solvent, it presents a risk of material incompatibility.

To assess the flow cell and gasket material compatibility, we observed their response to a 72-hour submersion in a pure methanol solution. We looked for signs of discolouration and softening and observed no noticeable change. This is promising for the long-term reliability of our cell, although it did not rule out small amounts of leeching that could affect the sample's electrochemical signature.

2.3.3 System Control Software Testing

We evaluated the system control software by unit testing the syringe pump and potentiostat. After successfully interfacing with each component, we integrated the system and eliminated software bugs as they appeared. To ensure that we were processing data correctly, we compared our chronoamperometry and SWV results with over 20 different measurements taken with PSTrace, the software shipped with the potentiostat. The measurements were identical.

2.3.4 Full System Electrochemistry Measurements

To evaluate the entire measurement system, we used a black-box approach, where we focused on the system's inputs and outputs. Specifically, we were interested in confirming that the system can produce chronoamperometry and square wave voltammetry data correctly. We used Alizarin Red as a test compound as it has a strong electrochemical signature around 0.4 V, which is within the range of our system. To verify our measurements, we used our sponsors' lab data showing the expected response.

Chronoamperometry

We ran a single 10-minute measurement, during which we injected three different concentrations of Alizarin Red at 3-minute intervals. The detailed testing scheme is in [Chronoamperometry](#) in the appendix. The results were well-aligned with the expectations and are shown in Figure 22 a).

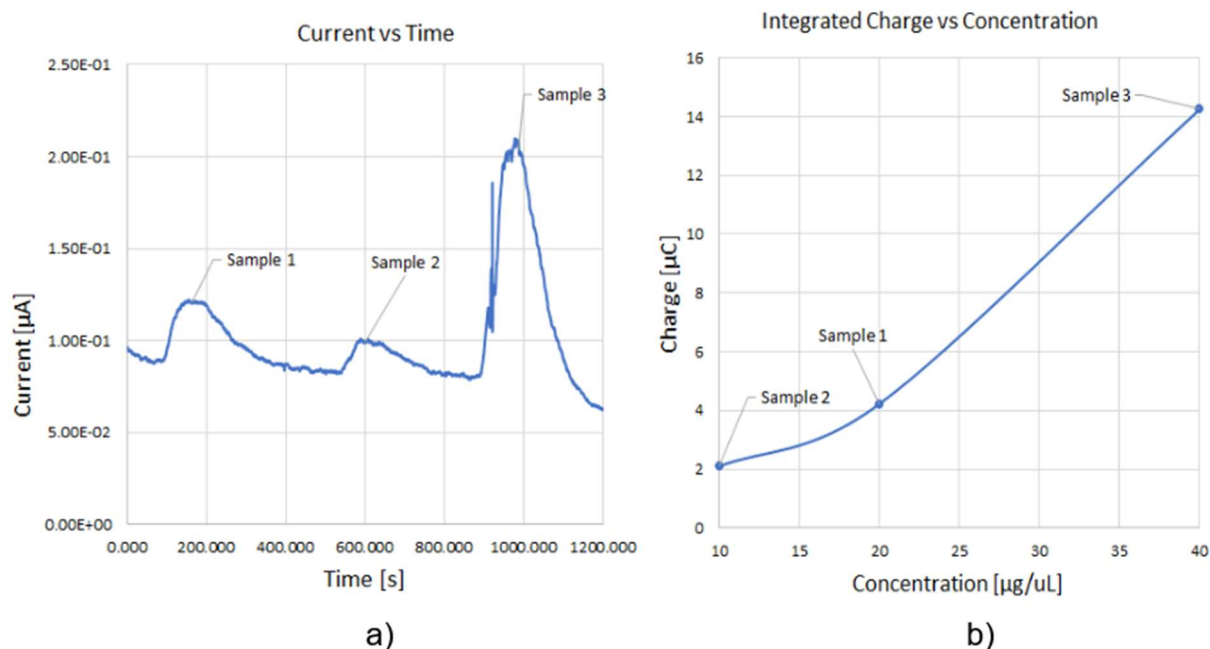


Figure 22: Current vs. Time and Charge vs. Concentration Data from a Single Chronoamperometry Measurement

The three peaks in Figure 22 a) correspond to the three samples of varying concentrations passing through the flow cell and undergoing oxidation. We integrated each peak separately, relative to its baseline, to get the total charge transferred for each sample concentration. This gave three data points of charge versus concentration, which we plotted in Figure 22 b). We expected the total charge to increase with the sample concentration, which is consistent with what we saw. The exact relationship between charge and concentration is not of interest to us, so there is no need to perform a curve fit on a larger dataset.

Another measure of our data is the width of the current peaks in time. Figure 22 a) shows how the falling edge of each peak is approximately 2 minutes longer than the rising edge of the same peak, which shows how the sample is being stretched out over time. Sample retention in the flow cell could account for this and should be further investigated with similar measurements. Repeating the measurements with lower concentrations can also give an estimate of the system's limit of detection.

Stop Flow with Voltammetry

Our sponsors have previous results for a square wave voltammetry measurement on Alizarin Red from 0.0 V to 0.8 V. Using the same parameters, we repeated their measurements in our flow cell with the goal of reproducing a current peak at a similar voltage. The full test is in [Stop-Flow with Voltammetry](#) in the appendix.

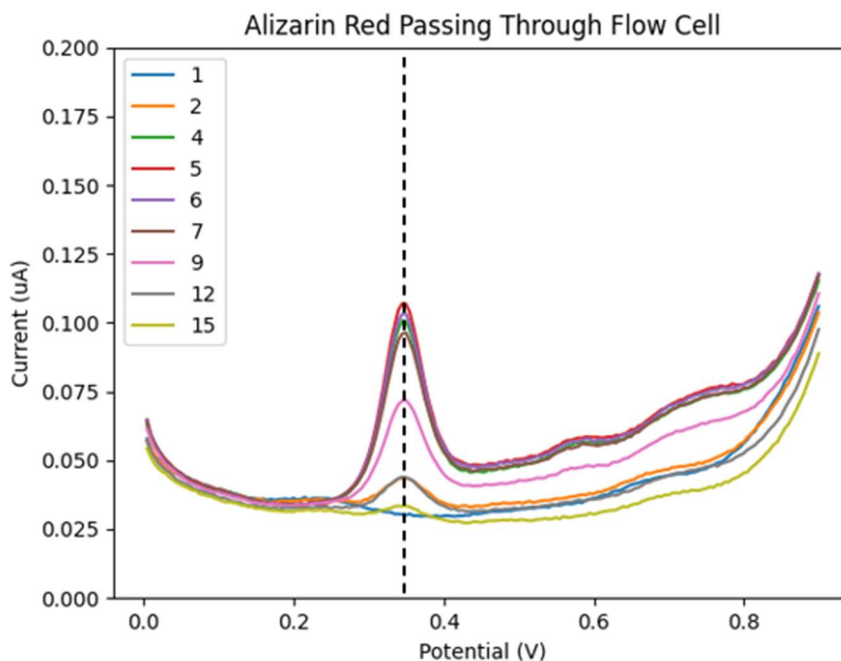


Figure 23: Current vs. Voltage Curves for 9 of 16 Voltammetry Sweeps Performed During a Stop-Flow Measurement

Figure 23 shows the results for the stop-flow measurement. Sweep 1 corresponds to the background curve before the sample arrived. The sample entered the flow cell on sweep 2 and a peak formed at 0.37 V (marked with black dashed line). The magnitude of the peak increased as the sample arrived in the flow cell (sweeps 4 through 7). Then, as the sample left the flow cell, the peak magnitude decreased (sweeps 9 and 12). The sample left by sweep 15. The peak location of 0.37 V coincides with our sponsor's previous results.

Alizarin Red Passing Through Flow Cell

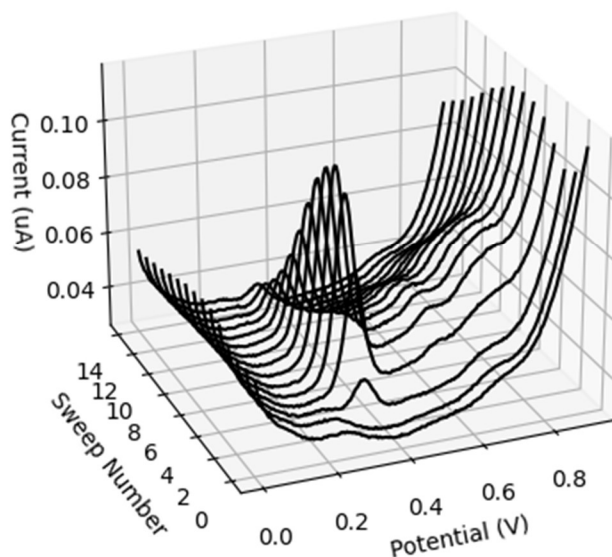


Figure 24: 3D Plot of Current vs. Voltage vs. Sweep Number for a Stop-Flow Measurement

For visualization, Figure 24 shows the same data plotted in 3D, where the peak at 0.37 V can be seen growing and shrinking as the sample enters and leaves the flow cell.

3 Conclusions

The motivation for this project was to bring our sponsors closer to their final goal of a market-ready automated fentanyl detection system. Given our 8-month period, we focused on a simplified system, only containing components that are essential to detection, so our sponsors can optimize the detection scheme before proceeding with the system.

We have developed a portable bench-top testing device that automates the electrochemistry technique our sponsors developed. We proved that this technique can be used to detect and quantify substances in our flow cell. We have also automated the system to allow our sponsors and their students to run repeatable experiments quickly.

A major result of our work is a working, 3D-printed flow cell. We confirmed that our resin and gasket materials are compatible with a pure methanol solution, which is sufficient for the system's purposes. We also showed that our design is fully sealed for the system's required flow rates, which was the most critical design requirement.

Another deliverable is our GUI's simultaneous control of sample flow and data acquisition, allowing us to run an automated stop-flow analysis. With a working flow cell and this automated routine, our sponsors can easily run hundreds of consecutive measurements during the process of optimizing the detection scheme.

A remaining unknown is the complete characterization of the system's electrochemical response, so that its results can be standardized and easily interpreted. This procedure is a continuation of the measurements described in Testing and Results, and is outlined in Recommendations.

4 Recommendations

Future work must focus on characterizing the accuracy and repeatability of the detection process, particularly with fentanyl and its analogues. There is also significant engineering work ahead to develop a commercial product that can be used outside the lab.

Our principal recommendation is that the validation and refinement of the chemistry processes take place before significant additional engineering work. The testing system we developed will allow for the refinement of the key system parameters, such as the exact flow rates, volumes, voltages and currents. Additional engineering work will be most effective if these parameters have been tuned and the system requirements are solidified, allowing for the optimal selection of components and layout. Our specific recommendations are outlined below.

4.1 Accurately Quantify Fentanyl

More testing should be done to prove that fentanyl can be accurately quantified with this system. We have completed initial testing and found that the charge transferred during oxidation increases with the concentration of Alizarin Red. However, this was not a strictly linear relationship. We recommend testing to determine if the same relationship exists with fentanyl. We also recommend running these tests with the internal standard to determine the margin of error of fentanyl quantity.

4.2 Find the Lower Detection Limit

The lower limit of detection needs to be determined for fentanyl and its analogues in the system. Testing needs to check that the system can quantify traces of fentanyl well below the lethal dose. This is critical to ensure this system will be effective. Voltammetry measurements with smaller concentrations can help find this limit.

4.3 Prevent Contamination Across Measurements

A procedure needs to be developed to quantify the contamination across measurements. The contamination needs to be small enough to ensure that fentanyl traces from past measurements do not confound later measurements. The long-term goal of implementing a single-use fluid path will alleviate this issue.

4.4 Characterize and Prevent Air Bubbles

When air bubbles move through our flow cell, they disrupt the electrode's measurement. We have seen air bubbles move through quickly so that they do not affect later measurements. We have also seen air bubbles linger and alter the baseline of measurements without compromising the variation. We recommend further testing to determine the effect of air bubbles on different measurements. We also recommend the development of data processing techniques to remove the noise from the air bubble signals if necessary.

4.5 Design Custom Electrodes

A big challenge we faced over the course of this project was designing a flow cell that could seal the electrode chips provided to us, as discussed in Design Alternatives. Designing custom screen-printed electrodes with a ring of dielectric material around the electrodes would make this flow-cell sealing challenge much easier and repeatable.

Once the above risks are addressed and the system parameters are more solidified, we recommend the engineering improvements in [Engineering Recommendations](#) of the appendix.

5 Deliverables

System Layout

1. Complete system mounted on ½" thick Lexan board

Flow cell

1. 1 double- and 1 single-electrode flow cell with gasket, lid, nuts and bolts
2. 3 double- and 1 single-electrode flow cell, unassembled
3. 3 spare lids, spare nuts and bolts
4. 8 spare gaskets, 36 in² of Buna-N gasket material and a .dwg file to laser cut more if needed
5. OnShape flow cell assembly and .stl files for all parts

Software

1. Computer with application loaded
2. GitHub repository with all code and .stl files

Unimplemented Extras

1. PCB with Raspberry Pi to run system without laptop
2. Servo motor

References

- Advion Interchim Scientific. (n.d.). [6-Way Valve Function]. Retrieved April 2, 2022, from <https://www.flash-chromatography.com/injection-loops/>
- BC Coroners Service. (2022, March). *Illicit Drug Toxicity Deaths in BC January 1, 2012 – January 31, 2022*. <https://www2.gov.bc.ca/assets/gov/birth-adoption-death-marriage-and-divorce/deaths/coroners-service/statistical/illicit-drug.pdf>
- BioDevice Technology, Ltd. (n.d.). *Round working electrode EP-N* [Photograph]. Retrieved April 2, 2022, from <https://biodevicetech.com/materials/157603883747601.pdf>
- Bizzotto, D., & Sammis, G. (2020). *Development of a Versatile Prototype for Opioid Quantification*. Grant Application.
- Fundamental Electrochemistry. (2020, May 8). *Three electrode system* [Diagram]. <https://www.facebook.com/Dore.TheExplorer/>
- Generalic, E. (2018, October 20). *Column Chromatography* [Illustration]. <https://glossary.periodni.com/glossary.php?en=column+chromatography>
- Juergens, J., & Parisi, T. (2021, November 15). *Fentanyl Addiction, Abuse And Treatment*. Addiction Center. Retrieved April 2, 2022, from <https://www.addictioncenter.com/opiates/fentanyl/>
- Metrohm. (n.d.). *Flow Cell for Screen-Printed Electrodes* [Photograph]. Retrieved April 2, 2022, from <https://www3.metrohm.com/nb-no/products-overview/electrochemistry/cells%20for%20electrochemistry/FLWCL?fromProductFinder=true>

PalmSens. (n.d.). *EmStat3 Module* [Photograph]. EmStat3 Module. Retrieved April 2, 2022, from <https://www.palmsens.com/product/oem-emstat3-module-2/>

Valco Instruments Company Inc. (n.d.). *Sample Injection – Valco 6 port valve* [Diagram].

Retrieved April 3, 2022, from

https://www.vici.com/support/app/app11j.php?fbclid=IwAR0agc2T6sFhZ3ElOLj6E_KBAcko4OzU0uzGRD6FlskIkDS6fyITRn7TWwM

VectorStock. (n.d.). *Trash icon* [Illustration]. Retrieved April 4, 2022, from

[https://www.vectorstock.com/royalty-free-vector/trash-icon-garbage-symbol-can-bin-delete-vector-](https://www.vectorstock.com/royalty-free-vector/trash-icon-garbage-symbol-can-bin-delete-vector-21894282?fbclid=IwAR09yqT4M6NML9y0mAZOf1Ket01wHd4p8qObfPrcExPKstaRhd7fsoQYyYE)

[21894282?fbclid=IwAR09yqT4M6NML9y0mAZOf1Ket01wHd4p8qObfPrcExPKstaRhd7fsoQYyYE](https://www.vectorstock.com/royalty-free-vector/trash-icon-garbage-symbol-can-bin-delete-vector-21894282?fbclid=IwAR09yqT4M6NML9y0mAZOf1Ket01wHd4p8qObfPrcExPKstaRhd7fsoQYyYE)

Yau, B. et al. (2019, September). *A field assessment of Fourier Transform Infrared (FTIR)*

Spectroscopy and fentanyl immunoassay strips as point-of-care drug checking

technologies. [https://www.bccsu.ca/wp-](https://www.bccsu.ca/wp-content/uploads/2020/08/FTIR_TestStrip_Field_Assessment-Sept2019.pdf)

[content/uploads/2020/08/FTIR_TestStrip_Field_Assessment-Sept2019.pdf](https://www.bccsu.ca/wp-content/uploads/2020/08/FTIR_TestStrip_Field_Assessment-Sept2019.pdf)

Appendices

A. Internal Standard

It is possible that the drug sample can get caught in system components or diluted before it is measured. Therefore, a method of accounting for these losses and ensuring correct calibration is necessary. This is accomplished by mixing an internal standard into the sample.

An internal standard is a chemical substance with a known electrochemical signature designed to calibrate the system. A known concentration of internal standard is mixed into the sample before the sample is introduced to the system. Since the internal standard will pass through system components and be mixed with the carrier fluid together with the drug sample, it will experience losses and dilutions in the same proportion as the sample. Then, when a quantitative measure of the drug sample is made, a quantitative measure of the internal standard is also made. The measured concentration of internal standard can be compared to the known concentration that was inserted into the system. In this way, the factor by which losses and dilutions have decreased the concentration can be calculated. By scaling the measured drug concentration by this same factor, the effect of losses and dilutions can be removed from the drug sample quantification.

As a simple example, suppose a concentration of 20 g/mL of internal standard is mixed with a drug sample of unknown composition. When the measurement is conducted, 10 g/mL of internal standard and 5 g/mL of drugs are detected. Since we know that half of the internal standard has been lost before detection, we know that half of the drug sample has also been lost. Therefore, the final drug concentration is reported as 10 g/mL.

Our sponsors are developing a custom internal standard specifically for this system and its chemical composition has not been finalized.

B. Separation Column

A separation column is a tube filled with a chemical substance known as the stationary phase. The stationary phase is selected so that it has a specific polarity, either polar or nonpolar. The constituent substances of the sample bind to the stationary phase in different amounts based on their polarity. This determines how quickly they will be moved through the separation column.

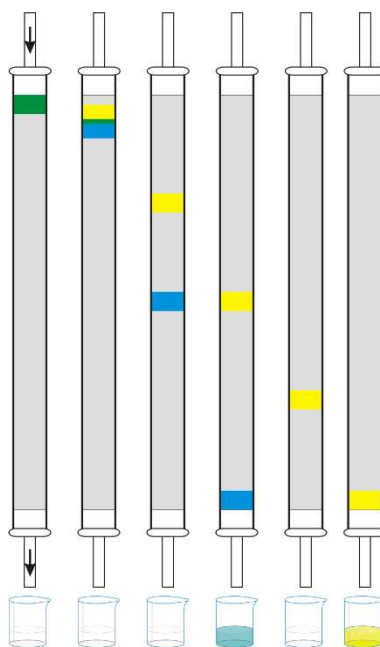


Figure 25: Process by which a Mixed Sample is Separated in a Separation Column⁸

When a sample containing multiple substances is pushed into the separation column, it is separated as shown in Figure 25. The mixed sample, which is composed of a blue substance and a yellow substance, is green when it is introduced to the separation column. The blue substance has a different polarity than the stationary phase and is therefore not bound to it. It moves through the separation column quickly and appears at the outlet early. Conversely, the yellow substance has a similar polarity to the stationary phase and is therefore bound more tightly to it. It progresses through the column more slowly and appears at the outlet after the blue sample. Altogether, the effect of the separation column is to spread the different substances

⁸ Generalic, 2018

out over time so that at any given time, only one sample is emerging from the column. This ensures that when the measurements are conducted, only one substance is present and therefore confounding results are avoided.

C. Flow Cell and 6-Way Valve Mounting

Flow Cell Mounting

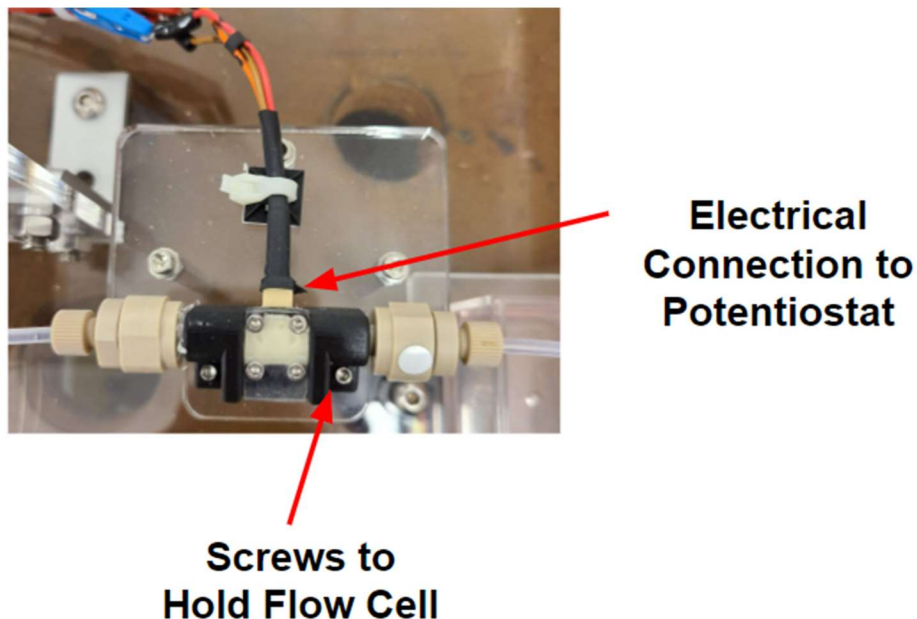


Figure 26: Flow Cell Mounting

Figure 26 shows how the flow cell is mounted to the physical platform. There is an acrylic plate mounted to the Lexan base with standoffs. The acrylic plate has threaded holes into which we installed bolts to hold the flow cell down. We also made a second set of threaded holes with the required spacing to hold the two-electrode chip flow cell if it is used instead. We mounted the electrical connector for the potentiostat to the acrylic plate and tied it down so that it does not become accidentally disconnected.

6-Way Valve Mounting

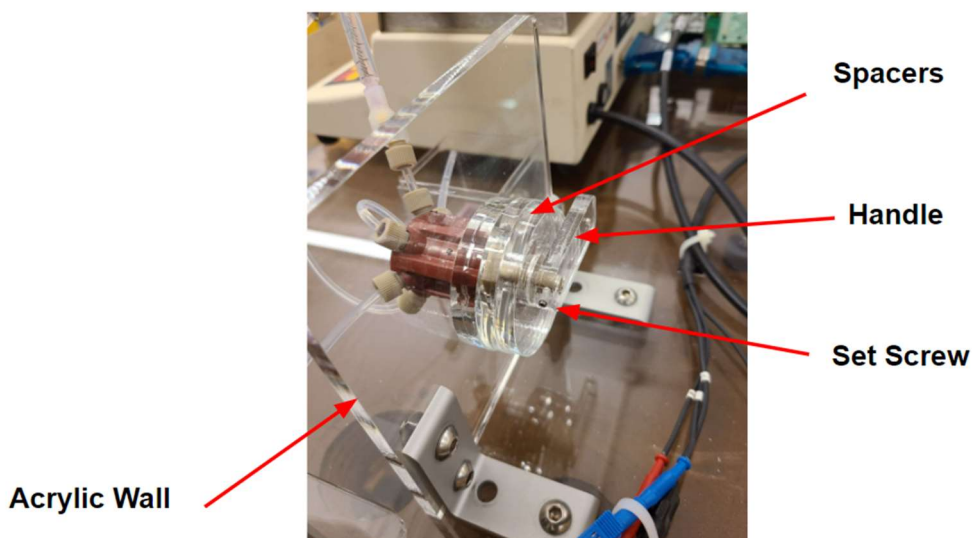


Figure 27: 6-Way Valve Mounting

Figure 27 shows how the 6-way valve is mounted. The valve does not have any convenient mounting features. Therefore, we made an acrylic wall with a cut-out that fits the shape of the valve body closely. To secure the valve, we designed spacers that fit between the valve handle and the wall, preventing it from moving out of the wall. We created a custom handle to turn the valve which can be automated by a servo motor. We used a set screw to attach the handle to the valve's shaft.

D. Testing and Results

Flow Cell Leak Testing

The single most important requirement of the flow cell is that it does not leak at the required flow rates. Increasing the flow rate increases the pressure in the cell, thereby raising the potential for leaks. Therefore, the flow cell's seal can be tested by recording the maximum allowable flow rate that does not produce a leak. Starting with 100 $\mu\text{L}/\text{min}$, we increased the flow rate in increments of 100 $\mu\text{L}/\text{min}$. At each rate we ran fluid through the flow cell for 30 seconds and monitored for leaks. We increased the flow rate to 5 mL/min, the maximum rate of the syringe pump, without observing a leak. As the measurement is expected to use a flow rate of 100-500 $\mu\text{L}/\text{min}$, this is a strong indication that leaks will not be an issue.

Full System Electrochemistry Measurements

Chronoamperometry

Each sample was roughly 200 μL and we used a 100 $\mu\text{L}/\text{min}$ flow rate. The sample was pumped directly into the flow cell, without a separation stage. The potential in the flow cell was held at 1 V. Since this is above Alizarin Red's oxidation-reduction potential, we expected it to undergo oxidation in the flow cell and produce a current. In between samples of Alizarin Red, when there was only carrier fluid in the flow cell, we expected to see a flat baseline current across time. When the samples arrived, we expected to see a current peak with an area proportional to the concentration. From the sample volume and flow rate, we expected the sample to be in our flow cell for 2 minutes.

Stop-Flow with Voltammetry

We used a 20 parts-per-million (ppm) solution of Alizarin Red, diluted with phosphate buffered solution as our sample, and ran 20 stop-flow measurement with 1 cycle of square-wave voltammetry (zero flow rate) followed by 6 second intervals of steady flow (200 $\mu\text{L}/\text{min}$) with no electrode stimulus.

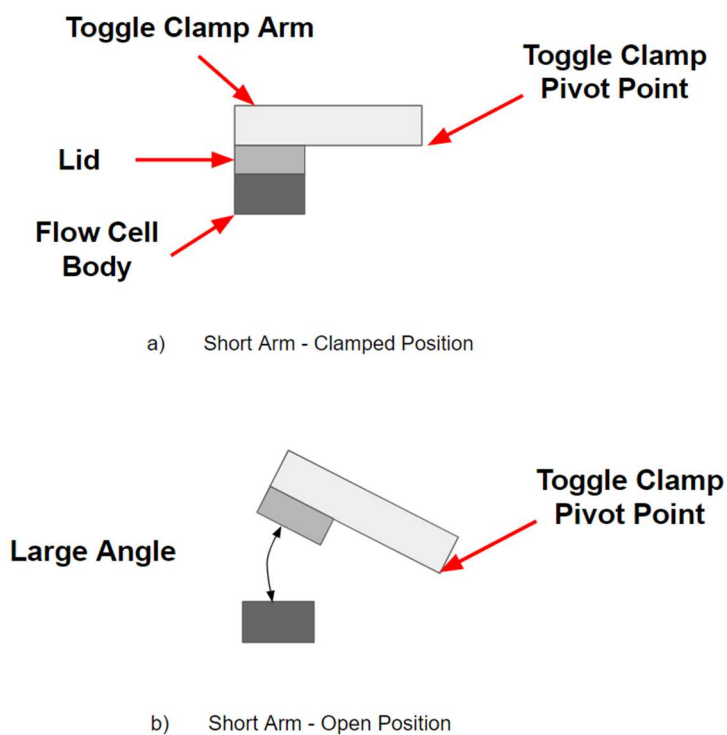
We expected to see the current versus voltage change across the 20 measurements as the sample passed into and out of the flow cell. Before the sample arrived, we expected a constant background. As the sample arrived in the flow cell and increased in concentration, we expected to see a current peak near its oxidation-reduction potential as it was being oxidized. Then, once the sample left the flow cell, we expected that the profile would return to the background we had seen before the sample arrived. The results are shown in Figure 23.

E. Engineering Recommendations

Toggle Clamp for Flow Cell

To further improve the ease of assembly, we explored a mechanism to clamp the lid instead of using nuts and bolts. We purchased a toggle clamp with a 20 mm arm length. However, the small arm length made the toggle clamp ineffective at sealing the flow cell and ensuring easy assembly. We therefore did not pursue the toggle clamp and leave it as a recommendation for further development.

Since the arm length is short, the end of the toggle clamp arm must rotate through a large angle to lift the lid and open the flow cell. However, this large angle of rotation makes it difficult to align the lid with the flow cell and disturbs the arrangement of the gasket and electrode chip.



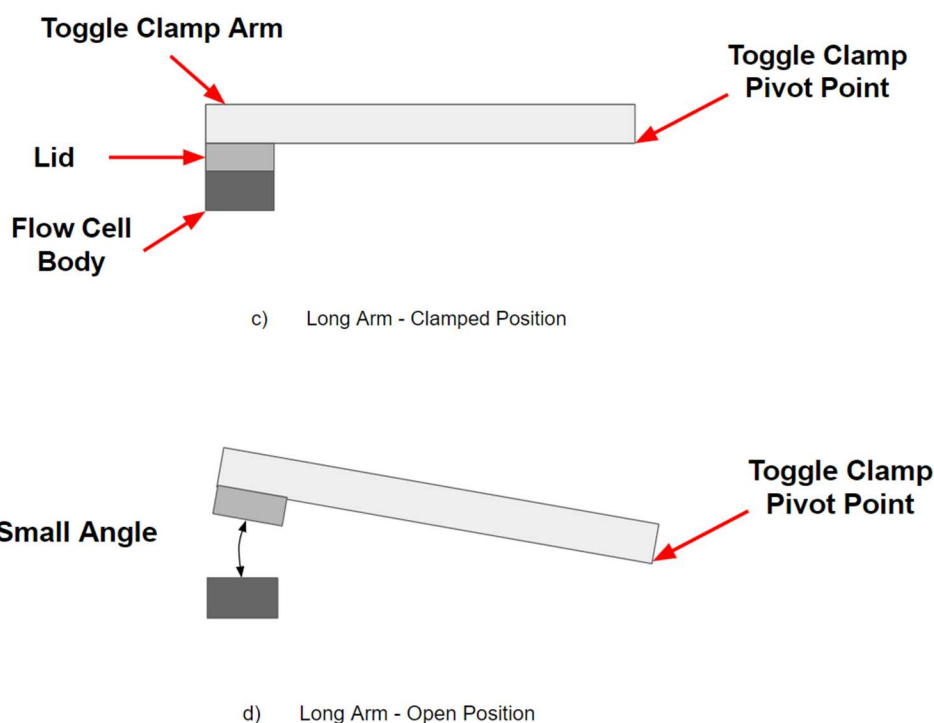


Figure 28: Comparison of a Small Toggle Clamp Arm to a Larger One

Figure 28 shows exactly how a longer toggle clamp arm makes it more difficult to align the flow cell lid and body. We recommend buying a toggle clamp with a much longer arm (~60 mm), or to use a linear toggle clamp if this is to be pursued further.

Software

There is some additional software functionality that we did not have time to time to implement but would be useful for increased ease of use and capability.

Multiplexer Implementation

We designed a flow cell with multiple electrodes but have not implemented a way of reading current from two electrodes simultaneously. There is a multiplexer in the kit for the potentiostat and we made a double-electrode connector but we couldn't get the code to successfully interface the multiplexer in time. A multiplexer would allow us to hold electrodes at different potentials. Only the deposition (flow) phase of the measurement can be

multiplexed. See page 32 of the [emstat communication protocol](#) on how to send a command to the potentiostat to start multiplexing, and page 10 on how to convert the packages sent back into current and potential measurements.

Cyclic Voltammetry Implementation

Cyclic voltammetry is another electrochemical measurement that our sponsors frequently use to check the electrodes. Currently, we can only run it through PSTrace but it would be nice to add the ability to run cyclic voltammetry to our program as well. To implement it, write the measurement parameters defined on page 31 of the [emstat communication protocol](#) to the potentiostat and then read the packages sent back from the potentiostat as done in the chronoamperometry and square wave voltammetry functions.

Live Plot Readability Improvements

We currently plot the current vs potential readings for every measurement on the same graph on top of each other. After several consecutive stop-flow measurements, it becomes difficult to differentiate the plots. It would be beneficial to be able to hide/show certain plots during the live reading, as well as be able to zoom in on specific parts of each plot.

System Control from a Raspberry Pi

We originally planned on running the system from a Raspberry Pi instead of a laptop computer. Due to stock limitations, we were not able to get a Pi in time to integrate it into our system. To increase the portability of the system, a Pi can easily run our system by following the steps in [Setting Up Raspberry Pi for Remote Connection](#).

F. User Guides

Flow Cell

Components

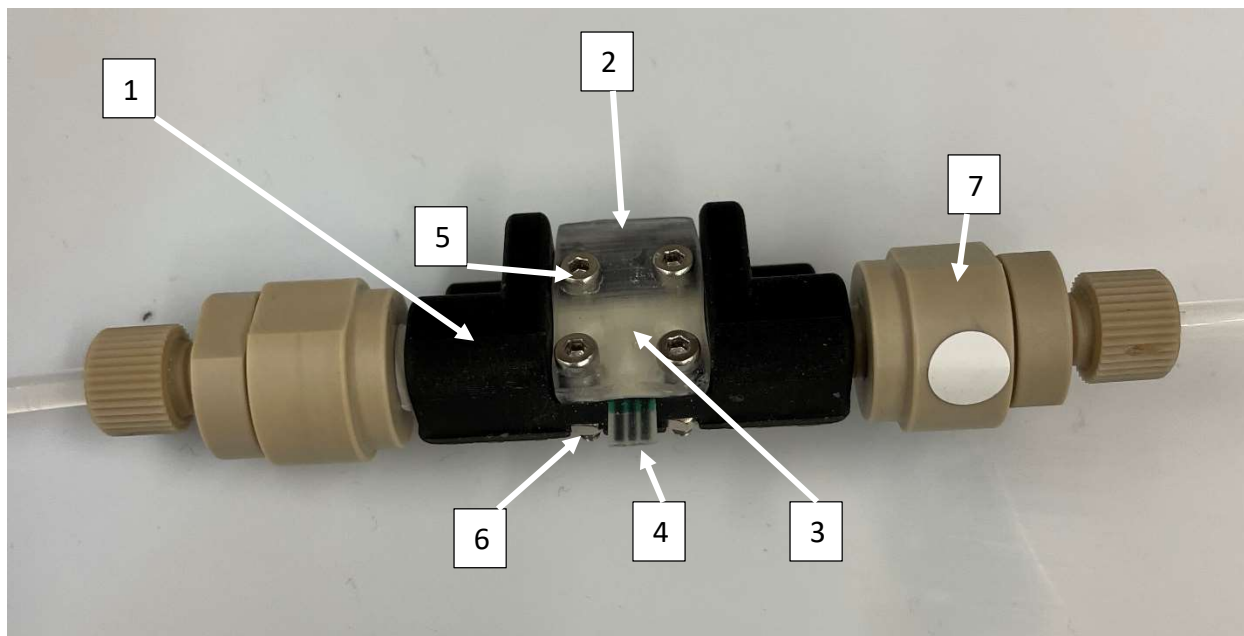


Figure 29: Flow Cell Components

Parts:

1	Flow cell body
2	Flow cell lid
3	Gasket
4	Electrode chip
5	(4) M2x10 bolts
6	(4) M2 nuts
7	Inlet and outlet check valves

Assembly Procedure

1. If the nuts are not already fitted in the bottom of the flow cell, begin by installing the nuts. To do this, put the bolt through the hole in the flow cell body from the top, without the electrode chip, gasket or lid in place. Put the nut on the bottom of the bolt and tighten it by hand until it meets the bottom of the flow cell body. Align the nut with the slot in the bottom of the body. Tighten the bolt using a hex key. The nut will get

pulled into its slot. This may require you to apply a large torque. Once the nut is in its slot, unscrew the blot. The nut will stay in place.

2. Remove the electrode chip from the sheet. We recommend leaving small stubs of the pieces connecting the electrode to the sheet and then sanding these stubs down with a small piece of sandpaper until the edge of the electrode is flat. This ensures that some of the electrode is not accidentally cut with the scissors and therefore it is precisely the right size for its slot.
3. Place the electrode chip face down in the slot. Ensure the electrode is seated on the ledge on the far side of the fluid chamber.

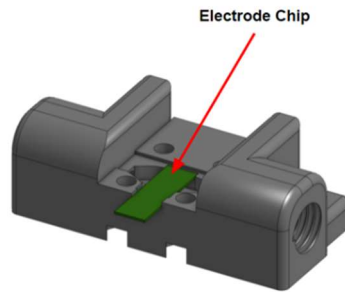


Figure 30: Electrode Chip Installation

4. Place the gasket on top of the electrode within the guides.

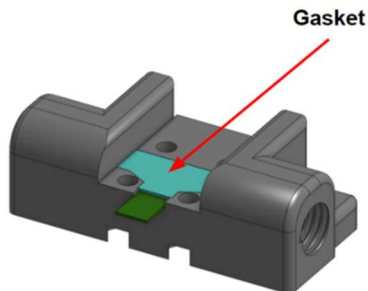


Figure 31: Gasket Installation

5. Place the lid over the gasket and align the holes with the base. Screw in the bolts until it becomes difficult to turn. Do not overtighten as it may cause the piece to break. If leaks are observed, continue tightening until no leaks are detected.

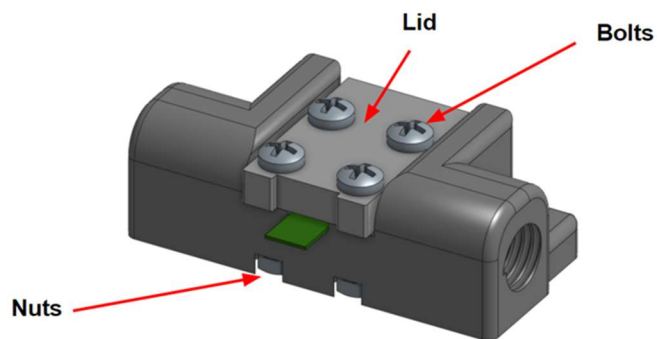


Figure 32: Lid Installation

6. Wrap the check valves in Teflon tape and screw them into the flow cell.



Figure 33: Installation of the Check Valves

7. Attach the flow cell to the acrylic mounting piece with two bolts and connect the potentiostat connector.

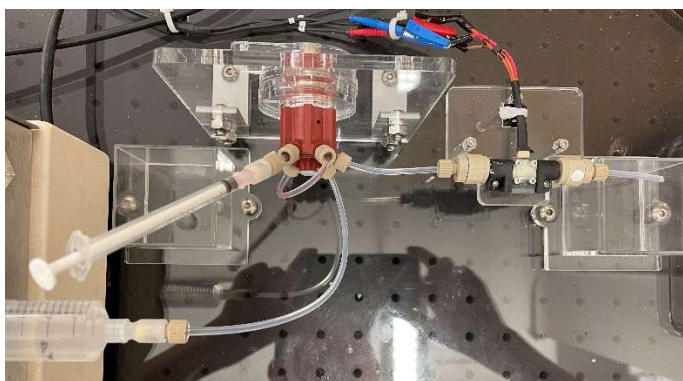


Figure 34: Final Flow Cell Assembly

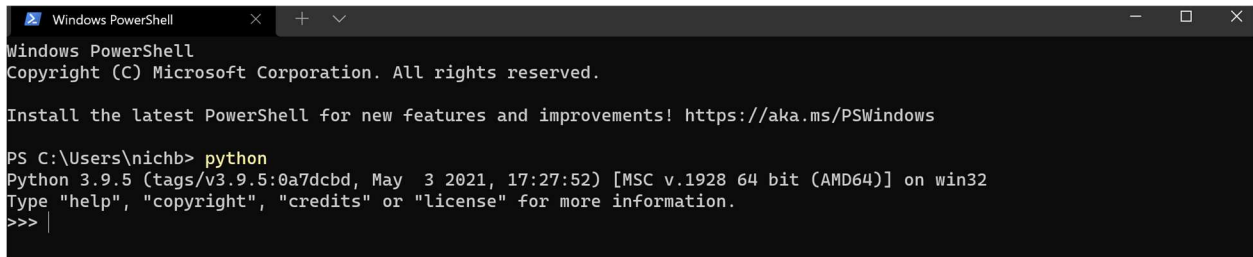
Software Installation

USB Driver Installation

A special USB driver must be installed for controlling the RS232 syringe pump. Please follow the steps on the website to install the driver properly. [Products \(prolific.com.tw\)](http://Products(prolific.com.tw))

Python Installation

Open a new terminal instance and check that Python 3.9 or newer is installed.

A screenshot of a Windows PowerShell terminal window. The title bar says 'Windows PowerShell'. The text inside shows the standard PowerShell startup messages, including the copyright notice for Microsoft Corporation. The user has entered the command 'python' at the prompt 'PS C:\Users\nichb>'. The output shows 'Python 3.9.5 (tags/v3.9.5:0a7dcdb, May 3 2021, 17:27:52) [MSC v.1928 64 bit (AMD64)] on win32'. It also lists options like 'help', 'copyright', 'credits', and 'license' for more information. The prompt '>>>' is visible at the end of the line.

```
Windows PowerShell
Copyright (C) Microsoft Corporation. All rights reserved.

Install the latest PowerShell for new features and improvements! https://aka.ms/PSWindows

PS C:\Users\nichb> python
Python 3.9.5 (tags/v3.9.5:0a7dcdb, May 3 2021, 17:27:52) [MSC v.1928 64 bit (AMD64)] on win32
Type "help", "copyright", "credits" or "license" for more information.
>>> |
```

Figure 35: Checking Python Installation

If not installed, it can be done here: <https://www.python.org/downloads/>.

Git Installation

Next, check if git is installed on your device. Type “git” into your terminal and if it gives an error, download it here: <https://git-scm.com/download/win>. Please check all the default values during installation, except change the default MinTTY terminal to the windows default console window.

Python Packages Installation

Once the latest version is installed, please install the following packages by typing the following into the terminal.

```
pip install pandas
pip install Matplotlib
pip install pyserial
pip install PySimpleGUI
pip install numpy
```

Fentanyl Quantification Code Cloning

To clone the latest version of the Fentanyl Quantification device code from github, first use the command line to navigate into the directory in which you would like to have the application.

```
cd pathtofolder
```

```
PS C:\Users\nichb> cd 'C:\Eng Phys\W2022\ENPH 359\'
PS C:\Eng Phys\W2022\ENPH 359> |
```

Figure 36: Navigating to an Installation Location

Next, install the code. In the terminal you can type:

```
git clone https://github.com/hjonesmtb/SyringePump.git
```

Quick Start

Run the system_control script by either opening the installation folder and clicking on

'system_control.py' or typing

```
python .\SyringePump\system_control.py
```

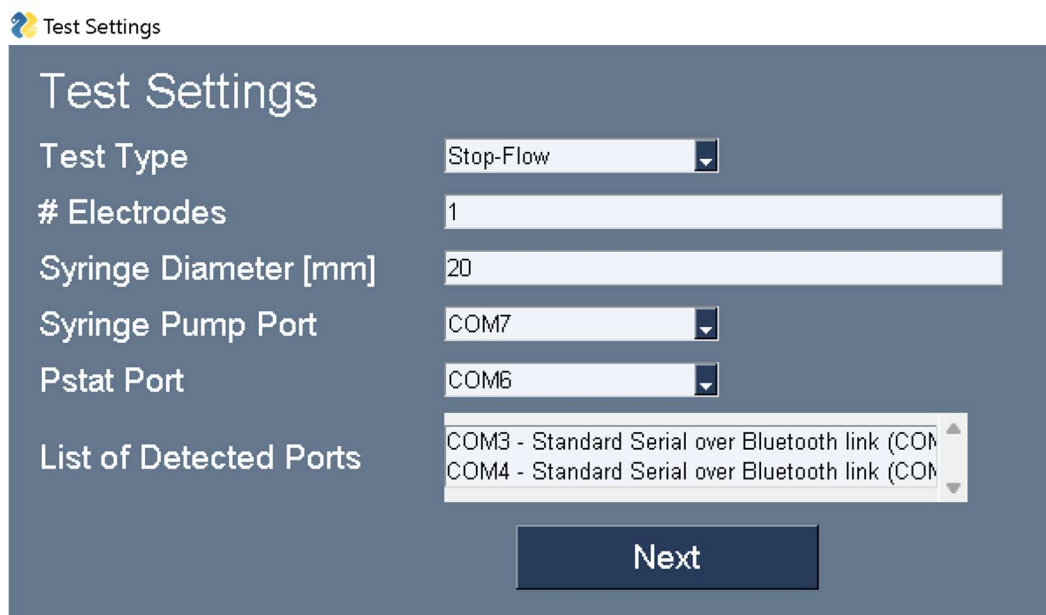
 into the terminal.

Figure 37: GUI Screen for Test Selection and Port Configuration

Select the test type and the correct ports. The syringe pump port will usually have “prolific” beside the port name in the list of detected ports. The potentiostat port can be found by trial and error or by opening the device manager and plugging in the potentiostat and seeing what port gets connected.

Once the proper ports are selected click next. The System Parameters Screen appears.

Stop-Flow

▼ Parameters

Test Name: Stop-Flow_22-04-02_1136

Stop

Measurements: 10

Measurement Volume [uL]: 100.0

Flow rate [uL/min]: 1000

E deposition [V]: 0.8

Flow

E condition [V]: 0

t condition [s]: 0

t equilibration dep [s]: 2

t equilibration swv [s]: 2

E begin [V]: -0.4

E stop [V]: 0.4

E step [V]: 0.005

Amplitude [V]: 0.01

Frequency dep [Hz]: 10

Frequency 1 swv [Hz]: 37

Frequency 2 swv [Hz]: 25

Frequency 3 swv [Hz]: 7

Current range min (0-7): 1

Current range max (0-7): 5

Current range starting (0-7): 3

☐ Measure i forward only

Back Start Stop

Press Start to Begin Test

Figure 38: GUI System Parameters Screen

Here, the system parameters can be adjusted for the test run. Only the square wave measurements without a zero frequency will run. For example, if frequencies of 37, 25 and 0 are specified, every stop phase will run a square wave measurement at 37 Hz, then 25Hz and continue. For 37, 0 and 0, only one square wave measurement (37Hz) will run per measurement sequence.

As the test runs, data will appear on the deposition graph showing the electrode being prepared for the test run. Press load sample and turn sample valve at the same time when you are ready to inject.

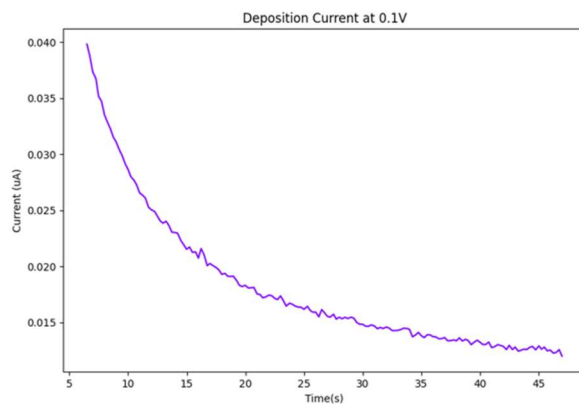


Figure 39: Deposition Current Shown During the Initial Flow Through Phase

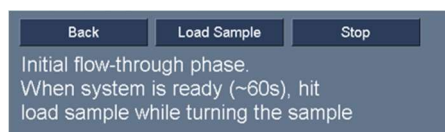


Figure 40: Button to Push When Injecting Sample

As the system runs data will be automatically plotted and saved.

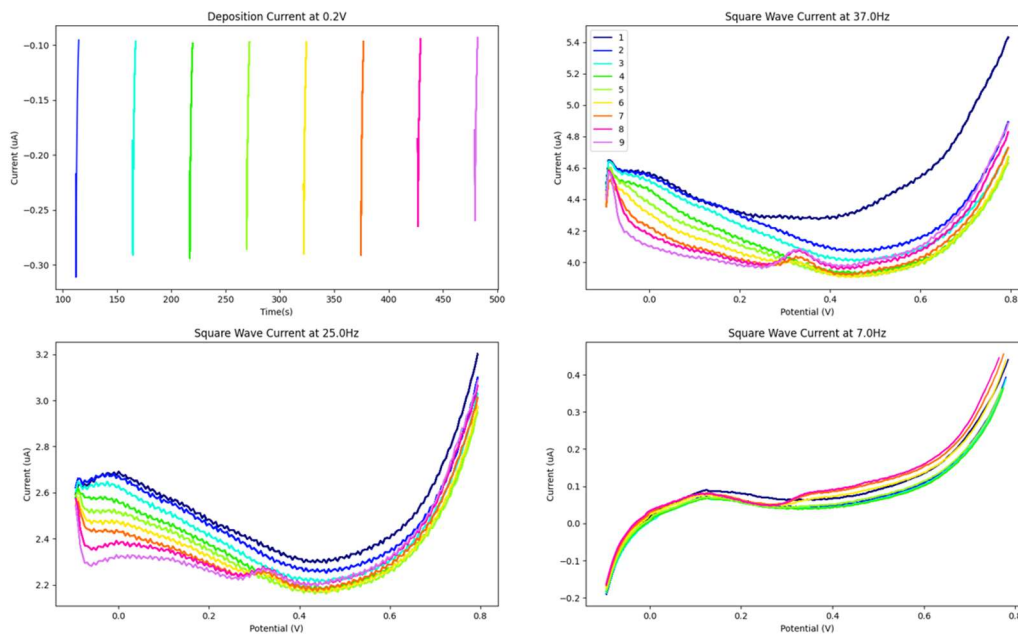


Figure 41: Example of Data Plotted During a Test

To run the syringe pump manually, independently of a measurement, use the options at the bottom of the screen. The syringe pump must be stopped before starting a measurement.

Saving Data

Data is saved in 3 ways in the path_to_installation_folder/SyringePump/data/testname folder

1) Configuration file. 'config.json'

This contains all the system parameters set by the user for future reference.

```
{
  "__System_Data__": true,
  "test_name": "Stop-Flow_22-03-30_1817",
  "test_type": "Stop-Flow",
  "Inject_time": 51.00284957885742,
  "flow_rate": 800.0,
  "infusion_volume": 1.08,
  "e_cond": 0.0,
  "e_dep": 0.0,
  "e_begin": -0.1,
  "e_end": 1.0,
  "e_step": 0.005,
  "t_cond": 0.0,
  "t_dep": 4.499999999999999,
  "t_equil": 12.0,
  "amplitude": 0.01,
  "frequencies": [
    37.0,
    25.0,
    18.0
  ],
  "n_measurements": 18.0,
  "step_volume": 0.06,
  "syringe_diam": "20",
  "n_electrodes": "1"
}
```

Figure 42: Example of a Configuration File

2) 'measurement#.csv'

A .csv of each measurement containing data on all the different measurement techniques.

A	B	C	D	E	F	G	H
Potential_dep	Current_dep	Potential_37.0	Current_37.0	Potential_25.0	Current_25.0	Potential_18.0	Current_18.0
0	-1.972	-0.095040563	3.9125	-0.095040563	2.404375	-0.095040563	1.5218125
0	-1.153125	-0.090043687	4.076875	-0.090043687	2.33875	-0.090043687	1.440625
0	-0.791875	-0.085046812	4.055625	-0.085046812	2.26	-0.085046812	1.400625
0	-0.60375	-0.080049937	3.98125	-0.080049937	2.21125	-0.080049937	1.40125
0	-0.488125	-0.075053062	3.9175	-0.075053062	2.195	-0.075053062	1.39625
0	-0.4125	-0.070056188	3.8625	-0.070056188	2.1925	-0.070056188	1.39625
0	-0.355625	-0.065059313	3.836875	-0.065059313	2.175625	-0.065059313	1.4125
0	-0.314375	-0.060062437	3.813125	-0.060062437	2.190625	-0.060062437	1.416875
0	-0.280625	-0.055065563	3.795625	-0.055065563	2.180625	-0.055065563	1.41625
0	-0.2630625	-0.050068687	3.7925	-0.050068687	2.18625	-0.050068687	1.43625
0	-0.2395625	-0.045071813	3.765625	-0.045071813	2.195	-0.045071813	1.438125

Figure 43: Example of a "measurement#.csv" File

3) 'injecttime_total.csv'

A csv containing a running collection of every measurement along with the time that measurement is taken.

1	Time	Potential	Current
2	1.719811	0	-0.09462
3	1.957231	0	-0.06975
4	2.220073	0	-0.054
5	2.469716	0	-0.04438
6	2.719377	0	-0.03825
7	2.964653	0	-0.034
8	3.212654	0	-0.03069
...			
6582	746.8228	0.109831	2.119375
6583	746.8384	0.114828	2.129375
6584	746.9165	0.119825	2.120625
6585	746.9634	0.124822	2.139375
6586	746.9634	0.129819	2.135
6587	747.0102	0.134816	2.13375

Figure 44: Example of an "injecttime_total.csv" File

Adjusting the Code

Default Parameters

Open the 'system_data.py' file in preferred IDE or as a text file to edit the following parameters to your liking.

```
17 # default values
18
19 TEST_TYPES = ["Stop-Flow", "Chronoamperometry"]
20 DATE = datetime.now().strftime("%y-%m-%d_%H%M")
21 TEST_NAME = TEST_TYPES[0] + "_" + DATE
22 PUMP_BAUD = 1200
23 PUMP_COM = "COM3"
24 PSTAT_COM = "COM7"
25 E_CONDITION = 0 # V
26 T_CONDITION = 0 # s
27 E_DEPOSITION = 0.8 # V
28 T_EQILIBRATION = 2 # s
29 T_EQILIBRATION_DEPOSITION = 2 #s
30 E_BEGIN = -0.4 # V
31 E_STOP = 0.4 # V
32 E_STEP = 0.005 # V
33 AMPLITUDE = 0.01 # V
34 FREQUENCY_DEP = 10 #Hz
35 FREQUENCIES = [37, 25, 7] # Hz
36 FLOW_RATE = 1000 # uL/min
37 INFUSION_VOLUME = 1 # mL
38 N_MEASUREMENTS = 10
39 N_ELECTRODES = 1
40 T_DEPOSITION = INFUSION_VOLUME / N_MEASUREMENTS / (FLOW_RATE * FLOWRATE_CONVERSION)
41 STEP_VOLUME = INFUSION_VOLUME / N_MEASUREMENTS
42 FIGSIZE = (20,12)
43 PATH = os.getcwd()
44 PUMP_SCALE = 20 #cuts off first few points of the first deposition during stop flow.
45 MAX_PLOTS = 5
46 INITIAL_PUMP_TIME = 240
47 CR_MIN = 1 # minimum current range, 0: 1nA 1: 10nA, 2: 100nA, 3: 1 uA, 4: 10uA, 5: 100uA, 6: 1mA, 7: 10mA, user input
48 CR_MAX = 5 # max current range, user input
49 CR_BEGIN = 3 #Starting current range, user input
50 INIT_PUMP_SPEED = 1000
```

Figure 45 Example of Default Values

Updating Syringe Pumps

To upgrade the system's syringe pump, the serial driver in 'Pump22.py' will have to be updated. The basic serial interface will work for any RS-232 pump, but the device-specific commands will change.

First, the serial connection must be instantiated as shown in Figure 46. The number of stopbits, baudrate, and parity are all standard parameters that will be specified in the pump's datasheet. Everything else should remain unchanged.

```
def __init__(self, ser, baudrate):
    #self.port = serial.Serial(port = 'COM{}', stopbits = 2, baudrate = baudrate, parity = 'N', timeout = 2)
    try:
        self.ser = serial.Serial(port = ser, stopbits = 2, baudrate = baudrate, parity = 'N', timeout = 10)
        if self.ser.isOpen():
            print("pump port opened successfully")
    except:
        print(ser)
        print("COM port is not available")
    self.ser.flushInput()
    self.ser.flushOutput()
```

Figure 46: Initializing Serial Connection to Syringe Pump

Next, identify the character sequence recognized by the pump. For the [Model 22](#) pump we used, each command is a 3 character sequence followed by a carriage return (see Appendix C of the datasheet).

```
def write(self, cmd):
    print("write: {}".format(cmd))
    command = "{}\r".format(cmd)
    self.ser.write(command.encode())

    response = self.ser.read(3).decode("utf-8") # Isolate prompt from CR LF

    return response.strip()
```

Figure 47: Serial Write Function

In 'Pump22.py', the basic serial write functionality is contained in the 'write' function. The function takes a character command (from pump datasheet), and formats it according to the required character sequence. For the Model 22, it just adds a carriage return ('\r') to the end of the command. It then encodes the command as UTF-8 and writes it over the serial connection. Change this line according to the new pump's spec.

```
def query(self, cmd):
    print("query: {}".format(cmd))
    command = "{}\r".format(cmd)
    self.ser.write(command.encode())

    value = self.ser.read(10).decode("utf-8") # Isolate value from CR LF
    prompt = self.ser.read(3).decode("utf-8") # Isolate prompt from CR LF

    return value.strip(), prompt.strip()
```

Figure 48: Serial Read Function

Similarly, the basic read functionality is in the 'query' function. The function performs a similar task to 'write', but the command will trigger a response from the pump, which is returned by the function.

The remaining functions are all wrappers for specific commands, to improve modularity. They each call 'write' or 'query' using a device-specific command. The commands should be easily available from the pump's datasheet.

Setting Up Raspberry Pi for Remote Connection

Power:

Use 2A or greater 5V power supply.

Find IP:

Ubcvisitor: 10.43.164.200

ShawOpen : 10.243.100.1

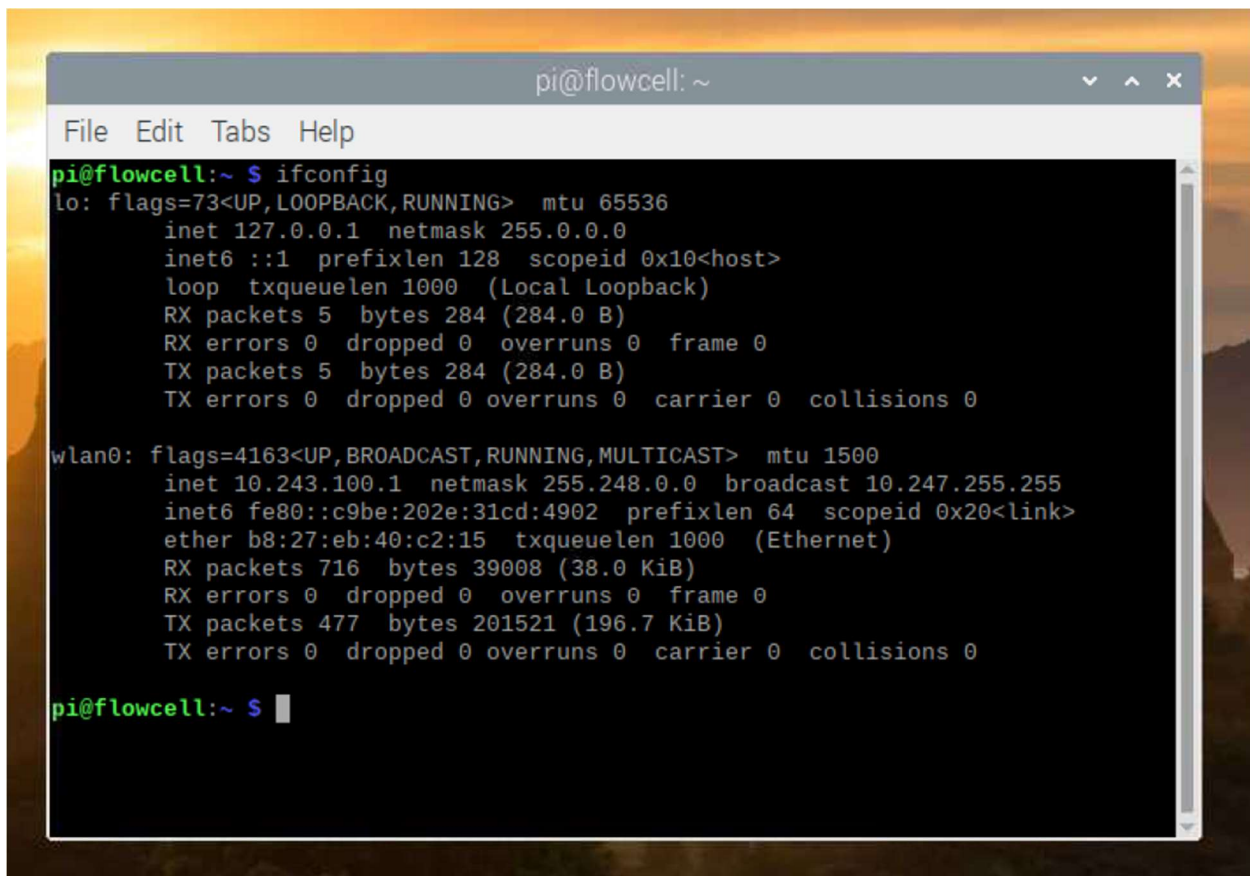
SharkTank: nurgi2021nurgi2021

If those do not work or the raspberry pi is on a different wifi.

Connect keyboard, mouse(optional), and display to raspberry PI.

Open a terminal instance (ctrl+alt+t)

Type ifconfig:

A terminal window titled 'pi@flowcell: ~' with a menu bar (File, Edit, Tabs, Help). The command 'ifconfig' has been executed, showing details for the loopback interface 'lo' and the wireless interface 'wlan0'.

```
pi@flowcell:~ $ ifconfig
lo: flags=73<UP,LOOPBACK,RUNNING> mtu 65536
    inet 127.0.0.1 netmask 255.0.0.0
    inet6 ::1 prefixlen 128 scopeid 0x10<host>
    loop txqueuelen 1000 (Local Loopback)
    RX packets 5 bytes 284 (284.0 B)
    RX errors 0 dropped 0 overruns 0 frame 0
    TX packets 5 bytes 284 (284.0 B)
    TX errors 0 dropped 0 overruns 0 carrier 0 collisions 0

wlan0: flags=4163<UP,BROADCAST,RUNNING,MULTICAST> mtu 1500
    inet 10.243.100.1 netmask 255.248.0.0 broadcast 10.247.255.255
    inet6 fe80::c9be:202e:31cd:4902 prefixlen 64 scopeid 0x20<link>
    ether b8:27:eb:40:c2:15 txqueuelen 1000 (Ethernet)
    RX packets 716 bytes 39008 (38.0 KiB)
    RX errors 0 dropped 0 overruns 0 frame 0
    TX packets 477 bytes 201521 (196.7 KiB)
    TX errors 0 dropped 0 overruns 0 carrier 0 collisions 0

pi@flowcell:~ $
```

Figure 49: Setting Up a Raspberry Pi

IP address is number after under wlan0 -> inet
I.e., 10.243.100.1

Install VNC viewer on windows machine:

[Download VNC Viewer for Windows | VNC® Connect \(realvnc.com\)](#)

Type IP address into search bar and connect.

Username: pi

Password raspberry

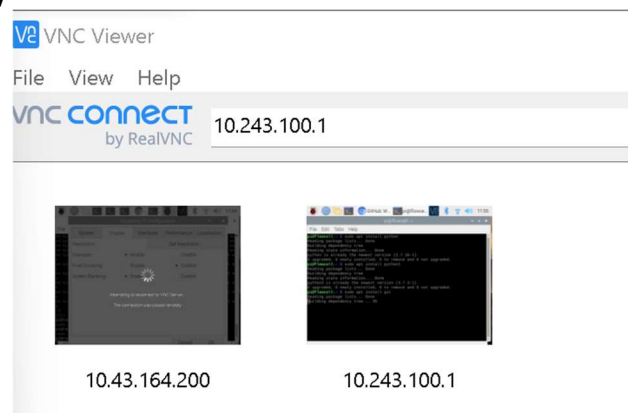


Figure 50: Installing VNC Viewer

Once the raspberry pi is set up follow the regular software instructions to install the application.

G. Known Bugs and Issues

The RS232 pump can disconnect unexpectedly from the software control program. This can't be avoided. The potentiostat will continue recording measurements but the pump will no longer move fluid.

Ports are not updated when a USB is plugged in after the system controller is open. Pressing "next" then "back" will update the values.