LAB 3 (FINAL LAB):

In this lab, you will fabricate an entire device from beginning to end using all the steps you learned this quarter using the mask that you designed. The most important part of this lab report is the *careful* and *thorough* reporting of everything that you did, so that we can inspect the chips afterwards, recreate your steps, and assign you the proper grade. Please keep track and write down everything that you did, every step along the way, and finally document with as many figures as possible.

Process Overview:

Step 1: Hard mask deposition, lithography, metal etch, microchannel etch, and hard mask removal

1. Use E-Beam evaporator to deposit 500 Å of Chromium and 500 Å of Gold following the detailed procedure below. **Bob will help you with the E-beam evaporator – Make sure you are properly trained.**
2. Solvent clean your samples and spin positive tone photoresist, following the procedure described below. Expose photoresist with the microchannel pattern and develop.
3. Use Gold etchant to etch the Gold then Chrome etchant to etch the Chrome.
    a. From Etch Rates for Micromachining Processing- Part II (Williams, Gupta, Wasilik 2003)
        i. Gold Etch: AU-5 etches gold at ~660nm/min
        ii. Chrome Etch: CR-7 etches chrome at ~170nm/min
        iii. Chrome Etch: CR-14 etches chrome at ~93nm/min

    Ask Bob for further detailed instructions and safety precautions.
4. Inspect wafer (and take pictures) to ensure that you have etched all the way through the metal.
5. Leaving the photoresist on the sample, etch your sample approximately 10 microns in Buffered HF (BHF). The etch rate of BHF for glass slides is ~1.13 μm/min. Immerse the chip in DI for 1 minute before you place it in HF. This prevents bubble formation and ensures uniform etching in HF. PLEASE READ SAFETY INFORMATION REGARDING HF ETCHING (on web) PRIOR TO THIS STEP.
6. Strip off Au using the Gold etch and Cr using Chrome etch.

Step 2: Lithography using the metal mask pattern, metallization and lift-off

1. Solvent clean samples and spin positive tone photoresist (See below for process details).
2. Align the metal mask pattern (darkfield) to transfer the electrode pattern to the samples. See Below for the detailed process steps. Remember that there is an additional toluene soak step involved here. Also the exposure and develop times required are longer.
3. Prepare the evaporator to evaporate 500 Å of Titanium and 500 Å of Gold. **Bob will help you with the E-beam evaporator.**
4. Lift-off the undesired metal by placing the samples in Acetone. Be extremely gentle or all the metal may come off.
5. Strip off the photoresist in acetone and then rinse the samples in isopropyl alcohol and DI H_2O.

Step 3: PDMS preparation and channel sealing

1. Prepare a 10:1 mixture of PDMS base to curing-agent in a disposable metal tray. Prepare enough to cover a glass slide by 3-5mm.
2. Place a blank glass slide in another disposable tray and pour PDMS onto it.
3. Put into vacuum chamber to remove bubbles.
4. Place a cover to reduce evaporation and cure on the hot plate or oven at 80C for 10 minutes
5. Allow it to cool to room temperature.
6. Use a razor blade to cut out a rectangular piece larger than the fluidic pattern.
7. Use the biopsy punch to cut access holes in the PDMS above each of the microchannel reservoirs.
8. Descum with O_2 plasma, align access holes to the reservoirs and bond.

Step 4: Experiments within channels and efficient reporting
See below for details.

Detailed processing steps:

Chip handling

1. Use gloves while handling the chip.
2. Using the diamond tipped scribe, make a small 'G' scratch at the edge on the back side of the chip. This is done because the chips are transparent and a mark helps in distinguishing the top side during processing.
3. Avoid scratching the front surface of the chip - always place the chip on a clean wipe.
4. It might take some time in getting used to holding the chips.

Chip Cleaning

1. Place the chips to be cleaned in a chip holder and then immerse in a beaker containing Acetone(ACE). Keep the beaker in the ultrasonic cleaner for 3 minutes.
2. Repeat for 3 minutes in beaker of Isopropyl alcohol (ISO) in the ultrasonic cleaner.
3. Rinse the chip in deionized (DI) water.
4. Blow dry with N2 .
5. Dehydrate in 125 deg C oven if going directly to lithography.

Note:

1. The cleaning solutions can be reused several times. All the groups will share the same set of cleaning solvents, so be careful not to contaminate the solutions.
2. In case the existing cleaning solvents need to be replaced, dispose them in the appropriate container- **Do not pour solvents down the drain**.
3. All beakers in the lab must be labeled correctly. Beakers are color coded to prevent cross-contamination, please do not use for other purposes.

Optical lithography

The following detailed procedures given here are for optical lithography using AZ 4110, which is a positive photoresist.

Spin on Photoresist

1. Place cleaned chips on the aluminum tray, place the tray on the aluminum block in the furnace. Dehydration bake for 3 minutes at 120 °C.
2. Place chips under the HMDS vapor deposition hood for 1-3 minutes.
3. Place a dummy chip on the spinner and set the spinner speed at 4000 RPM. This is to check if the spinner vacuum is on and other things are working as expected.
4. Place the actual sample on the spinner and put on a few drops of the resist till all of the chip is covered.
5. Spin the chip at 4000 RPM.
6. Pre-bake for 1 minute at 85 °C on the hot plate.

Exposure using MJB-3 mask aligner

1. Turn on <POWER>
2. Slide out mask holder and put the correct mask in with the chrome side facing the chip.
3. Put your chip on the chuck and slide tray under mask
4. Rotate lever to raise chuck. Make sure you actually are in <CONTACT>.
5. Put chip in <SEPERATION>and align mask to your pattern, (if needed)
6. Set exposure <TIMER> (UV=7.5 mW/cm2) recommended for resist in use (17-25 seconds). For AZ 4110, this time is 20 seconds.
7. Bring chip and mask in contact and press <EXPOSE>
8. Lower chuck and remove your chip.

Developing (AZ 400K)

1. Place the chip in the holder and immerse into a beaker containing 4:1 H_2O: AZ400K with mild agitation for 60 seconds.
2. Rinse in DI H2O for 60 seconds.
3. Blow dry with N2 gas.
4. Inspect for photoresist residue under the optical microscope and insure that the patterns have been transferred accurately.

Etching (50:1)

1. Read the instructions on working with HF and be very careful when working with it.
2. Immerse the chip in DI for 1 minute before you place it in HF. This prevents bubble formation and ensures uniform etching in HF.
3. Dip the chip in BHF for the calculated time. (You have to etch down by xx nm. Etch rate of buffered HF is about 1000Å/min. but you should verify the etch rate.)
4. Strip off the photoresist in acetone and then rinse the samples in isopropyl alcohol and DI water.

Caution:

* Spinner will not start unless the vacuum chuck is turned ON.
* Always **wear eye protection** in the lithography room.
* Do not touch any parts of the MJB-3 Aligner with bare hands.
* Do not use plastic tweezers while handling hot chips.
* Do not throw any Developer down the drain. Put it in the container marked "Waste Developer".

Notes on handling Hydrofluoric Acid

General Information:

Synonyms: Hydrogen fluoride
Descriptions: Clear, colorless, fuming corrosive liquid or gas
Formula: HF
Constants: Mol. wt. 20.01, mp: -92.3°C, bp: 19.4°C, density: 0.987 (liquid)

Hazard Analysis:

Toxic hazard rating:

Acute local:	Irritant 3; Ingestion 3; Inhalation 3.
Acute systemic:	Ingestion 3; Inhalation 3.
Chronic local:	Irritant 2.
Chronic systemic:	Ingestion 2; Inhalation 2.

Toxic hazard rating code:

0: None
1: Slight
2: Moderate May involve both irreversible and reversible changes; not severe enough to cause death or permanent injury.
3: High: May cause death or permanent injury after very short exposure to small quantities.

Toxicology:

It is extremely irritating and corrosive to the skin and mucous membranes. Inhalation of the vapors may cause ulcers of the upper respiratory tract. Concentrations at 50-250 ppm are dangerous, even for brief exposures. Hydrofluoric acid produces severe skin burns which are slow in healing. The subcutaneous tissues may be affected, becoming blanched and bloodless. Gangrene of the affected areas may follow.
(All of the above information was taken from Dangerous Properties of Industrial Materials by N. Sax.)

1. Always **wear eye protection, face shield** and **gloves** when using HF.
2. **Work under the HF hood at all times**.
3. Do not bring open containers or cotton swabs with HF out into the room. This applies equally to any other solutions containing HF.
4. HF should be kept in polyethylene or Teflon containers, beakers and dishes ONLY**. No glassware is allowed in the HF hood.**
5. If gloves become wet or otherwise come into contact with HF, do not touch on any valves or switches. Have someone else do it.

6. If there is any doubt about skin contact with HF, flood area for several minutes (especially under fingernails). HF causes severe burns to skin and eyes that are not immediately painful or visible. Therein lies a substantial part of the danger.
7. **It is not advisable nor permissible to work alone in the lab when handling HF.**

CAUTION: The Hydrofluoric Acid or buffered hydrofluoric-acid etch is dangerous. Use utmost caution! A suggested procedure to follow is to wash or rinse your hands following the performance of an etch or when moisture that might even remotely contain HF is contacted in the HF etch area. If HF is contacted, immediately quench with water and use the ACID-AID that can be found on the shelf in the HF-etch room. Note the eye-wash sink in the laboratory.

Metal Evaporation and Lift-Off Process

Metallization in a vacuum evaporator system is a line of sight process- metal is deposited over all exposed areas. Lift off is a common technique to pattern the metal. The sample is first patterned using standard lithographic techniques such that photoresist covers regions where metal is not desired. The next step is to cover the entire sample with the metal. The metal only contacts the substrate in developed regions. The photoresist is then stripped away removing with it the undesired metal. An important feature of lift processes is that the side wall profile of the photoresist must be vertical or with an overhang. This causes a break in the deposited metal film and ensures easy lift-off. After normal lithography the side walls are sloping and in order to get an overhang profile the lithographic process is altered slightly. A toluene (formerly chlorobenzene) soak step is included after the UV exposure but before development. The toluene hardens the top layers of the photo resist making them harder to develop away. In general, the exposure time and the development time need to be changed from the optimal conditions to account for the alteration of the resist properties due to the toluene soak.

1. Clean the sample using the standard procedure given earlier.
2. Spin positive resist on your samples and expose it to UV light on the MJB-3 aligner. Remember to use the light field mask- the desired features are dark and the background is light. The detailed steps for the lithography were given earlier. Do not develop the photoresist at this stage.
3. Use the procedure that you had success with in lab 2 for the Toluene step. Blow dry the sample with N2. Do not rinse the sample in DI H2O after the toluene soak.
4. Now develop the photoresist using DI:AZ400K 4:1 using the standard procedure.
5. Never postbake a sample that is intended for lift-off. Rinse and dry.
6. Evaporate the desired thickness of metal in the vacuum evaporation setup. **Ask Bob for help you with metal evaporation.**
7. Lift off the photoresist
    a. Place the sample in a beaker containing acetone for about 15-20 minutes.
    b. The acetone should strip off the photoresist thus removing the undesired metal. In case some metal does not come off, you may have to squirt some acetone over these regions to dislodge the metal. Be gentle!
    c. Dip the sample in Isopropyl alcohol for 1 minute to remove the acetone.
    d. Rinse in running DI H2O for 1 minute.
    e. Blow dry with N2.

PDMS Preparation

In this step you will use your silicon mold to make a PDMS version of your channel geometry. You will be mixing together the base and curing agent that comprise PolyDiMethylSiloxane, removing bubbles from the mixture, and then curing it in an oven. Below are the detailed process steps and questions to answer in the lab report.

Procedure:

1. Mix PDMS together in a tin foil tray. The weight ratio of base to cross-linker is 10:1, ie 10g PDMS base mixed with 1g of the curing agent. Make only enough appropriate for the number of chips needed. Mix thoroughly with mixing stick. Weigh using scale provided.
2. Place a clean glass slide face up in a second, new, tinfoil tray.
3. Pour the PDMS mixture over the glass slide, at a thickness of about 3-5mm above the mold.
4. Place the PDMS in a vacuum chamber, for about 20 minutes or until all bubbles have left the PDMS. In order to make sure the chamber works, make sure that the arrow on the top of the top is lines up with the hole out to the vacuum pump.
5. Place the entire tray in the oven and cure for about 10 minutes at 80ºC.
6. After curing, take out of the oven and let it stand for several minutes until room temperature.
7. After it has cooled, carefully cut out PDMS with an exacto knife provided by Bob and peel off the glass slide. Be careful not to rip or damage the PDMS.
8. Punch access holes in your PDMS mold in each reservoir using the Biopsy punch.
9. CLEAN your area. PDMS is very sticky and messy. Please make sure things are clean for the next group.

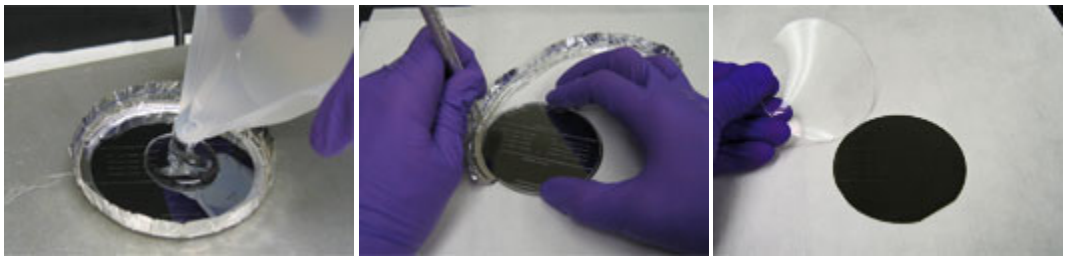


Figure 2: Pictures showing process of pouring PDMS mold.

Bonding PDMS layer to seal microchannels

Overview:

Here, you will be using the O_2 plasma descum machine to aide in bonding the PDMS channel onto a glass slide. First you need to clean the slide properly and after bonding you will punch access holes into the device. Below are the detailed process steps.

Procedure:

1. Clean the glass slide using standard wafer cleaning techniques, acetone, isopropanol, water. Dry the glass slide with N2 and then placing it on a hotplate for several minutes. Keep the side to be bonded face up throughout this process. Cool the slide to room temperature
2. Place the glass slide and PDMS, both boding sides face-up, side by side. Place in the O_2 descum machine to make both surface hydrophilic.
3. O_2 Plasma instructions:
    1. Turn on the AC power toggle switch on the ICP power supply located on top of the reaction chamber enclosure.
    2. Open the reaction chamber door and load samples on the flat sample tray. Return the tray with samples and close the door. Note: The door guides are notched and need to be adjusted upwards to close the door.
    3. Ensure the vacuum purge switch is in the down/off position. NOTE: this switch is located in the lower left position on the front of the chamber enclosure.
    4. Plug in the mechanical pump to one of the wall outlets at the back of the system.
    5. Move the 3 position vacuum fast/slow toggle switch from the middle/off position to the up/fast position to evacuate has and create a vacuum in the reaction chamber. Note: This switch is located next to the purge switch.
    6. Turn on the AC power for the RF power supply by placing the AC line toggle switch in the up/on position. The switch is located on the left side of the front panel. Note: This power supply is located on the right side of the reaction chamber on the bench top.
    7. Wait for the vacuum level to reach 300 millitorr on the analog vacuum gauge located on top of the reaction chamber enclosure
    8. Place gas #3 (O_2) toggle switch in the up position and wait for the vacuum to reach 300 millitorr again.
    9. Turn on RF power to the reaction chamber by moving the RF power toggle switch from the down off/remote position to the up/n position.
    10. Wait 1 minute and then turn off the RF power by placing the RF power toggle switch back in the down/off remote position.
    11. Turn off the O_2 gas flow by placing the gas #2 toggle switch in the down/off position.
    12. Stop pulling vacuum in the chamber immediately after turning off O_2 gas flow by moving the 2 position vacuum fast/slow toggle switch to the middle/off position.
    13. Turn off the AC line power to the RF power supply on the bench top by placing the AC line toggle switch in the down/off position.
    14. Place the vacuum purge toggle switch in the up/on position to vent the reaction chamber. Note: this will take about 30 seconds.
    15. Open the chamber door and remove the sample tray and samples.
    16. Return the sample tray to the chamber and close the door.
    17. Place the vacuum purge switch in the down/off position.
    18. Unplug the mechanical pump.
    19. Turn off the AC power line to the IC power supply on the top of the reaction chamber.
4. Use the biopsy punch to make access holes in the PDMS at the ends of the channels (at any point in this process).
5. Attach the PDMS and glass slide together firmly so that they bond.

In this lab, it is extremely important that you address all the issues that were brought up in the first lab report. Please make sure to write a detailed introduction, outlining the entire process and what exactly you will be doing in this lab. Make sure all figure captions are extremely detailed, with appropriate equipment used, etc… and that figures all have dimensions that are LEGIBLE. Also, please be as quantitative and descriptive as possible on all your processes. I would rather that you write bullet points of everything you did than omit things altogether for lack of wanting to write it up.

Following the same procedure as Lab Report I, please write all procedures that you performed succinctly. Please do not leave any detail out, so that if you came back to the lab 10 years from now, you would be able to exactly re-create what you did. Please append any handwritten notes, etc… to the lab report if necessary. In the process of writing this report, please make sure to address the following items/questions:

1. Analysis of Transparency Mask
    1. Study your printed masks under the microscope and comment on the quality of the print. Make sure to include at least one picture of the mask (with the appropriate figure caption in your lab report). The more pictures and commentary, the better. After analyzing this mask, please comment on why you think we are using the chrome mask instead of the film mask for this lab. Give at least two reasons.
    2. Measure the thickness of the ink using the Dektak. Measure the thickness in at least three different places, and comment on both the accuracy of the Dektak as well as the roughness of the film. Discuss on what you think the source or error comes from and why.
    3. Describe any differences from the mask that we fabricated and the one you designed, if any. Pay close attention to the alignment marks. Is this design going to work for a final microfluidic device? If so, can you outline any differences in your procedure with that of the ones outlined in this lab? If not, what needs to be changed in order for your design to be achievable?
2. Evaporation
    1. What is the density and melting temperature of Cr and Au?
    2. Why are we depositing two metals-Cr and Au as opposed to using just one of them?
    3. Do you think we could have use Al? Give reasons.
3. Lithography #1
    1. Discuss the possibility of using a negative resist instead of the positive. What changes would be required to be made on the mask and in the lithography process? Which process (positive resist vs. negative resist) is a better for this particular process why? What reasons do you think we chose to use a positive resist?
    2. Make sure to discuss this process in detail, all misalignment, all steps and equipment used, etc.. Please take as many pictures as necessary to get the point across, and label all figures. Any schematic that you can draw to establish a point will help, especially with quantitative dimensions.
    3. Did you have to re-do the lithography step at all? Why or why not? What determines whether the lithography is **good enough** to proceed to the next step?
4. Metal Etch

1. Make sure to detail the type of etchants used for the gold and chrome etch.
2. Comment on the quality of the results.
3. Note the parts that have not etched all the way, and explain why. Did the etch undercut the resist at all? Were there pinholes? Make sure to explain why or why not.
4. Provide a DekTak measurement of the etch profile and comment.

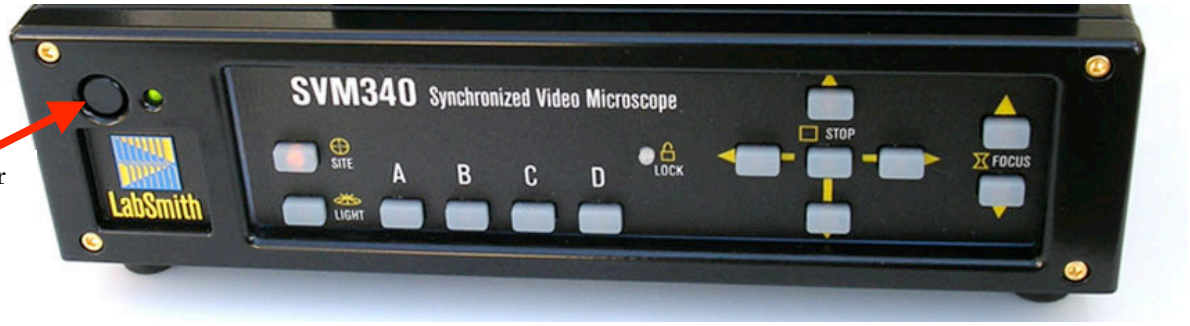
5. Glass Etch
    1. What is BHF? Give the chemical composition and use. Discuss etch rate characterization. Compare BHF etch rate with dilute HF. Find out the etch rates for Cr and Au in BHF and dilute HF.
    2. Exactly how deep did you etch the channels? Take multiple dektak measurements and quantify both the uniformity and the roughness of the channels. Why did the profiles come out the way that they did?
    3. Compare the uniformity and roughness measurements between the oxide etch in laboratory 1 and this microchannel etch (dilute HF vs. BOE). Why do you think that you have these results?
6. Lithography #2
    1. Describe the quality of the PR over the etched features. Take a DekTak measurement over non-critical feature and include the plot in your report.
    2. Describe the quality of the features after development. Are there problems? Explain what problems you have, the causes, and what you could do to try to fix them.
7. Lift off
    1. Why is the toluene soak needed before lift-off? What are the other common methods in use to perform lift-off?
    2. Discuss the results. Compare these results with your metal etch procedure. Which worked better? Why? What would you recommend for the next time you run this procedure? Why or why not? Why do you think we chose the procedure we chose.
    3. Take a DekTak measurement of an electrode inside the channel. What does the profile look like? Why?
8. PDMS
    1. How long did it take to remove all the bubbles from the PDMS? Why do you think this is important to the process?
    2. What were the exact proportions that you mixed to make the PDMS? How would changing these ratios affect the curing, formation, and structure of the PDMS?
    3. What were the major problems and issues encountered in this step, if any? What suggestion do you have to mitigate these problems in the future?
    4. Comment on the quality of the PDMS mold – is it thick enough and strong enough? Did it adhere well enough What would you do differently to improve your PDMS mold for the next run?
9. Bonding
    1. How important is it that the glass slide is clean before bonding to the PDMS? What can happen if the glass is not so clean?
    2. How well did the O_2 descum work? Did you get a perfect bond between the PDMS and glass? How can you tell whether the PDMS and glass is bonded perfectly?
10. Device characterization

1. Were you able to create a successful device? What were the problems, if any, with the device? Carefully write about what steps could be optimized, what you would have done differently, what steps should be re-done and why, etc…
2. If your device is successful, put a drop of DI water on one end of the channel and watch the channel fill. Take as many picture as you can of the filling process. Did any air bubbles occur during the filling process? What did you observe during filling? Were there any leaks through the device?

Device Testing

In this section, you will use the LabSmith SVM340 portable fluorescent microscope and LabSmith HVS448 6000 High Voltage Sequencer to image and apply voltages for electrokinetic experiments.

Procedure:



ıe Portable Microscope, recording, and saving

Turn on the SVM 340 by pressing the button furthest on the left of the front panel

f prompted for Communications properties, set the serial port: to "COM1" and press ok.

3. On the top bar, click on Video -> Process Live Video to activate the microscope camera.
4. To record a video, press the **record** button to start the recording, and the **stop** button to end the recording. The software should prompt you to save the files after pressing stop. Save the files in the folder on the desktop labeled ME141B, create a folder for your group and save all of your files there.

Controlling the camera, focus, and LEDs

1. To move the objective, press the **SITE** button then use the arrow keys to move in the XY plane.
2. To focus the microsope, press the up/down arrows labeled focus on the right side of the front panel
3. You can adjust the LED intensity inside the UScope software or on the front panel.
    a. UScope Software: On the top of the screen there is a box labeled LEDs with letters A,B,C,D. Adjust the sliders of each letter to change the brightness of the LEDs in each bank. For the LEDs in this lab, A,B, and D are blue, and C is white.
    b. On the front panel: press the **Light** button, next press the ABCD buttons to specify which banks of LEDs you want to manipulate (the letters that are illuminated), finally press the up/down arrows (previously used to move the microscope in the Y-direction) to adjust the LED brightness.

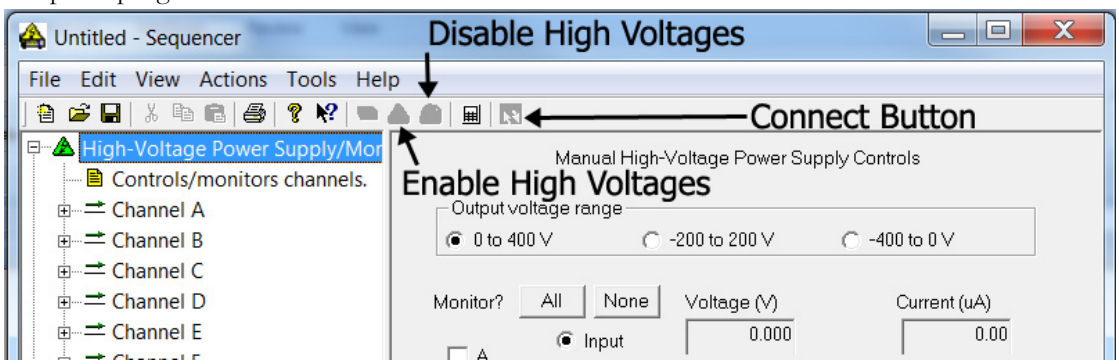
Filling the Channel

1. Use the 1 mL syringe with a 30 gauge needle, and fill the East channel well with **buffer**. NOTE: One syringe and needle should be used for all your experiments. Do not cross-contaminate this syringe with that for fluorescent sample. Use only one eppendorf tube with buffer.
2. The fluid should fill by capillary action, if it stops filling you can apply a pressure by pressing on the East well with your thumb (wear gloves). Refill the well and keep pressing until liquid **completely** fills all the channels. Make sure there are no bubbles in the channels. This is important for the electrokinetic flow to work correctly.

3. Once the channels are completely filled with buffer, use the 1mL syringe and fill the remaining 3 wells.

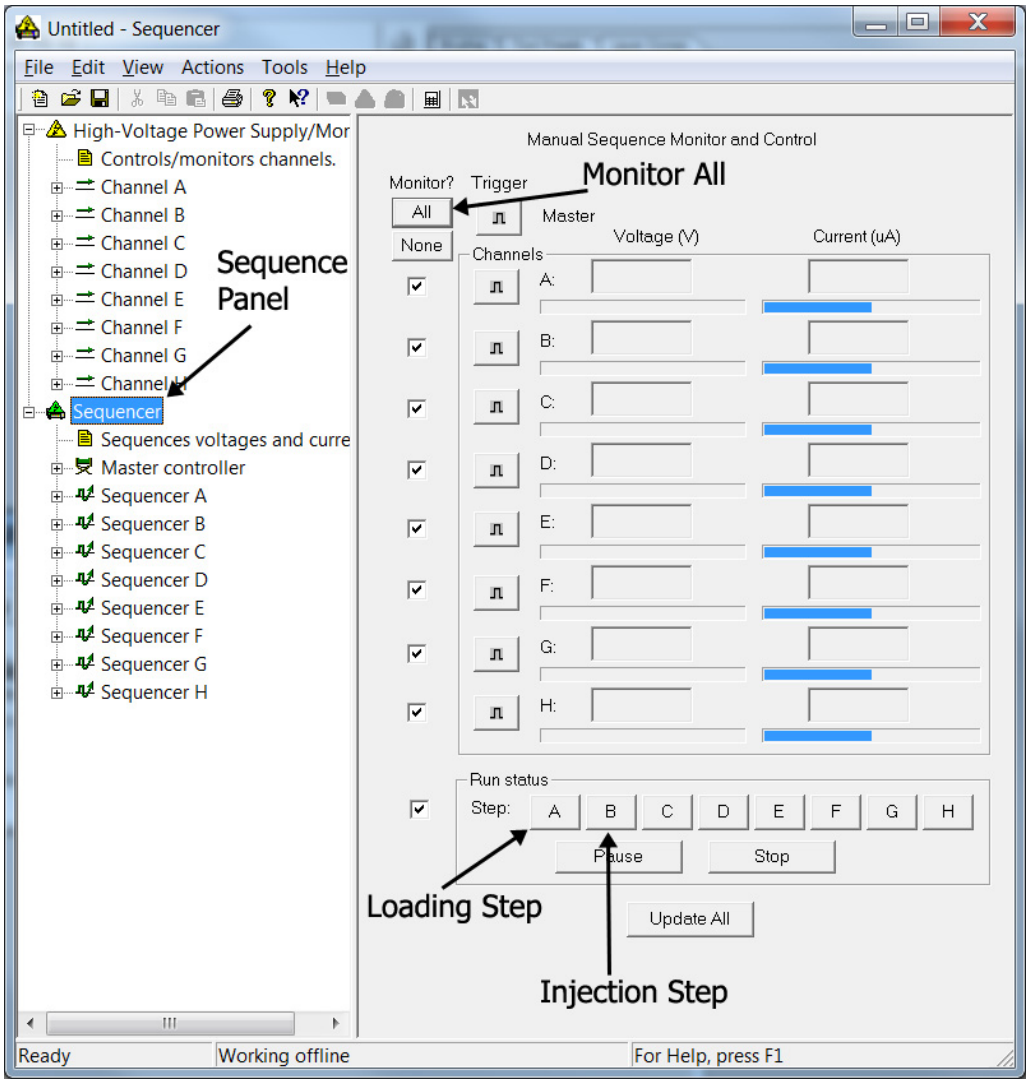
Placing Electrodes and using the Voltage Sequencer

1. Place the electrodes in the wells of the PDMS/glass chip, and connect them to the terminals of the LabSmith voltage sequencer. Make sure that Terminal A goes to North, B to East, C to South, and D to West.
2. Turn on the LabSmith HVS, open the Sequencer software and press the 'connect' button on the computer program.



3. Go to Tools -> Simple Sequence Wizard. Under the tab, 'Step A,' rename the Step name to "Loading," and enter the following voltages in the DC Voltage box, for A, B, C, and D, respectively; 492, 500, 0, and 345. Go to the Step B tab, and rename the Step name to "Injection." Enter the following voltages in the DC Voltage box, for A, B, C, and D, respectively; -381, 0, 156, and 2000. Press Apply, and then close the Simple Sequence Wizard.
4. Press the 'Enable high voltages' button.

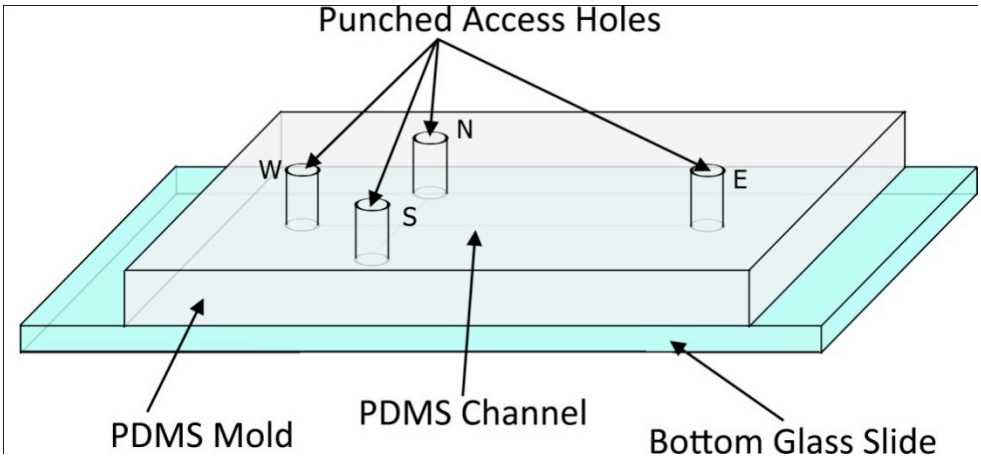
Press 'Sequencer' to open the Sequence Panel.



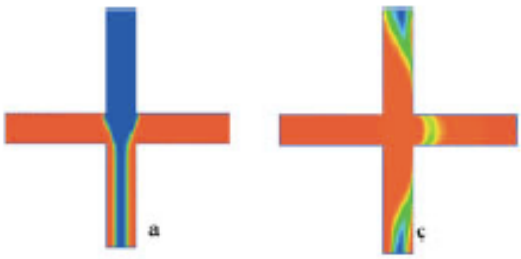
5. Press 'All' to monitor all the voltage and currents of the electrodes. Be sure that you are not getting a short in the device, and that all voltages are correct. Also note what currents are typical.
6. Finally, Press 'A' to run the Loading voltage sequence. NOTE: The wires are now live, exposed wires at high voltage. There is a current limiter, helping to ensure that no serious injury can occur, but avoid the eye opening shock and possible damage to the LabSmith and do not touch the wires (with anything) while the High Voltage Button is selected. At anytime, you need to adjust the positioning of the wires, click the Disable High Voltage button.

Experiments: Cross Channel

1. Fill the channel will buffer using procedures listed above. Next, Fill the north well with fluorescein. Put the electrodes in the wells using the following schematic: Terminals A goes to North, B to East, C to South, and D to West.



2. Next, apply the "loading" voltage to the system, and focus on the intersection between NSWE. You should see fluid flow from the north to the south well. Record this loading step in the ME141B folder. Try to get a video where the liquid is perfectly flowing from north to south, with no leaking.
3. Next, press the "injection" step on the sequence panel, you should see a small pug of fluorescein inject down into the east channel. After seeing this, click back on the "loading step", and you may see the plug move back into the loading channel.
4. Once you get a few successful injections, your goal is to record an injection at the intersection, and then record the plug propagating some distance downstream from the injection point. For the former, press record right before you press the "injection step" on the sequence panel. For the latter, move the stage to a point downstream, then press "injection" and "record". You may need to play with the timings a bit before you get a good recording.
5. How do you think your fabrication affected the results of your experiment?
6. You have at this point successfully injected a bolus of florescent analyte into a channel. You job is now to analyze this plug as it moves downstream with your videos. First, figure out the electrophoretic velocity of the plug. You can do this by measuring the time is takes a plug to travel downstream and the distance your measurement location is at. Next, take a single picture of a plug downstream and plot the resulting Gaussian of this plug. Figure out the diffusivity of the analyte, assuming that the sigma of the Gaussian is equal to 2*Diffusivity*time (where time is the time it took for the plug to reach the point that you are analyzing the plug). Finally, solve for the mobility of fluorescein, you can do this by assuming that Diffusivity = mobility * R *T, where R is the universal gas constant and T is the temperature (make sure you use the right units!!). Compare this with literature values of the diffusivity and mobility of fluorescein, which you can find off the web, or in relevant papers.



False color image of loading step and injection step at the intersection. Your video of injection at the intersection should look like this.

7. Clean you device and repeat the experiments with fluorescent nanoparticles

Experiments: Custom Design

1. Next, if you have chosen to use your own design, you need to characterize the channel using both conventional methods described above, as well as through experimental characterization. You are responsible for designing your own experiment using the dye and the buffer available. After being trained on both the microscope and high voltage power supply, you will have a range of experiments that you can perform. Design an experiment for your particular channel and write it out succinctly. Examples include, an injection like above, flow of dye through a particular region, investigating the effect of different flow rates in your channel with a dye tracer, etc…
2. Record your results with both the video as well as written observations and potentials. You may need to adjust the potentials in the sequencer specifically for your design
3. Explain what modifications you made to the voltage sequence, what is happening in your device and why it is interesting/useful. Suggests methods of improvement, and future designs for future students.

Channel Cleaning

1. Remove the fluorescein and buffer solutions from each of the wells and replace with DI water. Apply the loading voltage sequence to drive DI water through the channels. Replace the DI water every few minutes. Continue flushing until you cannot see any fluorescein in the channel.

End of Lab

1. Once you have finished with your lab, I would like to come in and inspect both your custom chip and the chip that you fabricated for lab 2. Please put your name on the chip in a holder where I can inspect. If you would like to keep your chip, please let me know and I'll examine it before the end of finals. Finally, please write in your lab report whether this chip is reusable and why or why not.