BME Design-Spring 2020 - Jason Wang
Complete Notebook

PDF Version generated by

Nicholas Pauly

on

Apr 29, 2020 @03:43 PM CDT

Table of Contents

Team contact Information

Nicholas Pauly - Apr 27, 2020, 9:14 PM CDT

Last Name	First Name	Role	E-mail	Phone	Office Room/Building
		Advisor			
Campagnola	Paul	Client			
Wang	Jason	Leader	jwang889@wisc.edu	920-313-0320	
Pauly	Nicholas	Communicator	npauly@wisc.edu	414-379-0137	
Meuler	Robert	BSAC	rmeuler@wisc.edu		
Koesser	Kevin	BWIG	kkoesser@wisc.edu	262-945-0271	
Beckman	Jiacomo	BPAG	jpbeckman@wisc.edu		

Project description

Nicholas Pauly - Apr 27, 2020, 9:17 PM CDT

Course Number: BME 301

Project Name: Nanofabrication of Microfluidic Device

Short Name: Microfluidic Device

Project description/problem statement:

Dr. Campagnola's lab has designed and constructed several microscope-based instruments for creating 3D nano/microstructure tissue-engineered scaffolds. These are used for studying cell-extracellular (ECM) interactions in cancers and fibrosis as well as for general biology applications. Currently, the method of introducing and eliminating excess protein solution is done by hand with a micropipette. This process is rather tedious and prolongs the fabrication process. Our client seeks an automated system capable of introducing a protein solution, filtering excess precipitate, and recovering unpolymerized protein solutions for re-use. Ideally, the device will be controlled by pumps and integrated into the current LabVIEW framework.

About the client:

Dr. Campagnola received his BA in chemistry at Colgate University in 1986 and his Ph.D. in physical chemistry at Yale University in 1992. His lab focuses on studying the alterations of the extracellular matrix (ECM) in epithelial cancers as well as in connective tissue disorders. His lab utilizes Second Harmonic Generation imaging microscopy along with two-photon excited fluorescence for the imaging of structural aspects of tissues. These approaches will ultimately be implemented as clinical diagnostics. Dr. Campagnola’s lab also uses 3D nano/microfabrication approaches to create biomimetic models of the ECM to study signaling pathways associated with cancer and provide insight into the design of tissue engineering scaffolds.

1/30/2020 - Adviser Meeting

Jason Wang - Feb 23, 2020, 4:39 PM CST

Title: Adviser Meeting

Date: 1/30/2020

Content by: Team

Present: Team

Goals:

Content:

- Gave adviser an overview of what was accomplished during the previous semester
-

Conclusions/action items:

2/7/2020 - Adviser Meeting

Jason Wang - Feb 23, 2020, 4:41 PM CST

Title: Adviser Meeting

Date: 2/7/2020

Content by: Team

Present: Team

Goals:

Content:

- Discussed different possible design ideas
    - clamp, weights, magnets
- Filtering methods
    - micropipette filtering
    - Cell strainers
- Fluorescence testing

Conclusions/action items:

2/14/2020 - Adviser Meeting

Jason Wang - Feb 23, 2020, 4:43 PM CST

Title: Adviser Meeting

Date: 2/14/2020

Content by: Team

Present: Team

Goals:

Content:

- Struggles with powering pump
    - Voltage amplifier
- Discussed general project deadline materials
- Filtering
    - Cell Strainer
- Discussed results of material testing
-

Conclusions/action items:

04/17/2020

KEVIN KOESSER - Apr 29, 2020, 3:41 PM CDT

Title: Advisor meeting - poster recommendations

Date: 04/17/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To summarize what needs to improve on the poster.

Content:

Intro:

mention cancer mortality in abstract

say cons of animal and in plastico models and pros of synthetic ECM model

less text

Design:

3 colors of Eppendorf tubes to represent different protein solutions

pillar filter pictures

check off which criteria are met and explain why while presenting

Testing:

swap the position of the flow chart and adsorption data

add a pressure diagram from Solidworks

Preliminary Designs

JIACOMO BECKMAN - Apr 29, 2020, 12:37 PM CDT

Title: Preliminary Designs

Date: 2/8/2020

Content by: Team

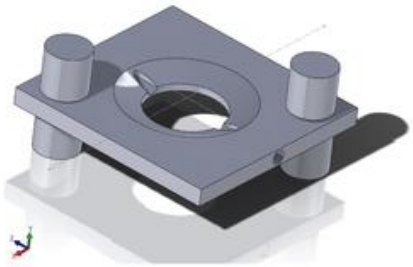
Present: Jiacomo, Jason, Kevin

Goals: Showcase and describe preliminary designs

Content: see below

Conclusions/action items: Create a design matrix to determine a final design

Jason Wang - Feb 23, 2020, 4:48 PM CST

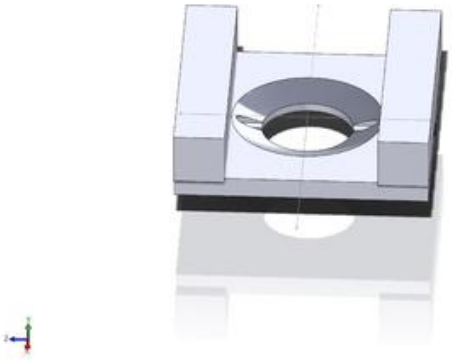


Magnet_Design_1_-page-001.jpg(212 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:37 PM CDT

The magnet design deploys two oppositely charged magnets being placed above and below the glass slide of the hybridization chamber. When this is done, the chamber would be effectively sealed. The magnets can also be moved to different corners of the glass slide to whichever combination that provides the best seal. More magnets can also be used on this design if two magnets are determined to not be enough.

Jason Wang - Feb 23, 2020, 4:48 PM CST

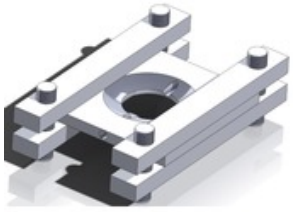


Weight_Design_1_-page-001.jpg(196 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:37 PM CDT

The weights design uses simple weights to seal the hybridization chamber. The weights will be in the shape of rectangles, to seal the hybridization chamber. The weights can be placed at multiple different places, allowing this design to be amended for the best possible seal to occur. The weights would be placed on top of the glass slide, and by their weight, would hold down the glass slide.

KEVIN KOESSER - Feb 23, 2020, 6:24 PM CST



clamp.png(252.1 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:38 PM CDT

The clamp design incorporates the same size hybridization chamber as does the others. The method for maintaining a tight seal uses two sets of clamps to hold and sandwich together all components. These clamps would be manually tightened with a screw in order to keep all of the parts in place.

Design Matrix

JIACOMO BECKMAN - Apr 29, 2020, 12:43 PM CDT

Title: Design Matrix

Date: 2/8/2020

Content by: Team

Present: Jiacomo

Goals: Determine a final design

Content: see below

Conclusions/action items: Develop the full, augmented system using the clamp design

JIACOMO BECKMAN - Apr 29, 2020, 12:43 PM CDT

Explanation for criteria

- Ease of fabrication
  - If the component is too difficult to create, then the design would be useless.
- Sealing capabilities
  - In order for protein solution to flow in and out of the chamber, a tight seal should be met.
- Ease of use
  - This refers to how each design would be operated, considering both time and amount of labor for the user.
- Cost weighs
  - This category refers to how much money each component would be to purchase.
- Durability
  - Durability takes into account how long each design could last over repeated uses.

JIACOMO BECKMAN - Apr 29, 2020, 12:43 PM CDT

Weight	Criteria	Weights		Clamp		Magnets	
35	Ease of Fabrication	4	28	4	28	4	28
25	Sealing Capabilities	1	5	5	25	3	15
20	Ease of Use	4	16	3	12	4	16
15	Cost	4	12	4	12	3	9
5	Durability	5	5	5	5	5	5
100	Total (100)	66		81		74	

Design_Matrix.jpg(64.9 KB) - download

Revised Microfluidic Device

JIACOMO BECKMAN - Apr 29, 2020, 12:46 PM CDT

Title: Revised Microfluidic Device

Date: 4/17/2020

Content by: Kevin

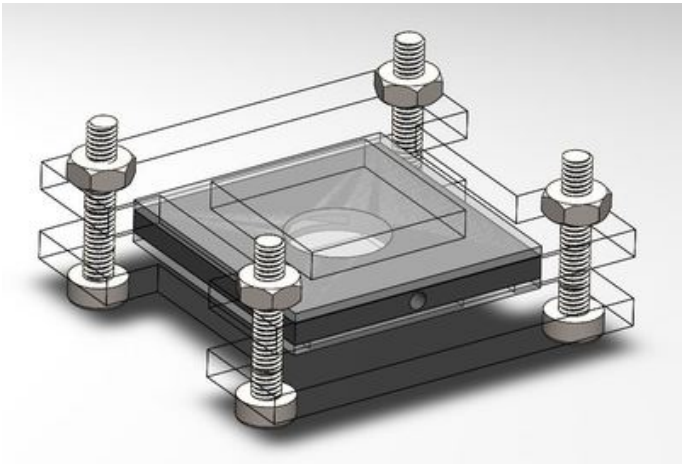
Present: Jiacomo

Goals: A modified version of the clamp design

Content: see below

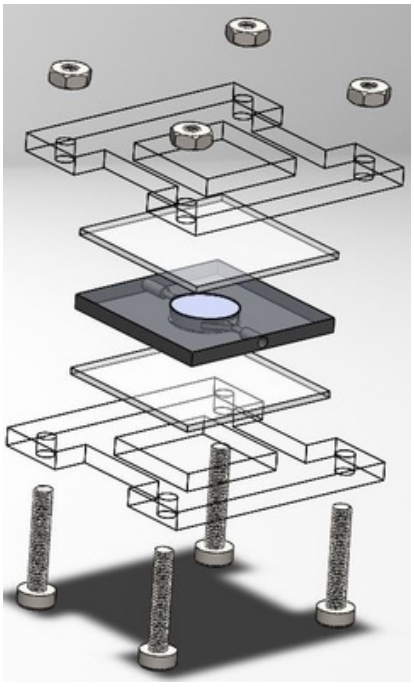
Conclusions/action items: Develop the full, augmented system using the clamp design

JIACOMO BECKMAN - Apr 29, 2020, 12:46 PM CDT



Sandwich.JPG(90.2 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:46 PM CDT



unnamed-2.jpg(61.9 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:49 PM CDT

Assembly view version and fully manufactured Solidworks models of the microfluidic device. Four screws at each corner of the clamp will subsequently hold the microfluidic device (made of EPU 40) to the glass slides there promoting a tight seal.

Full Proposed System

JIACOMO BECKMAN - Apr 29, 2020, 12:51 PM CDT

Title: Fully Proposed System

Date: 4/17/2020

Content by: Team

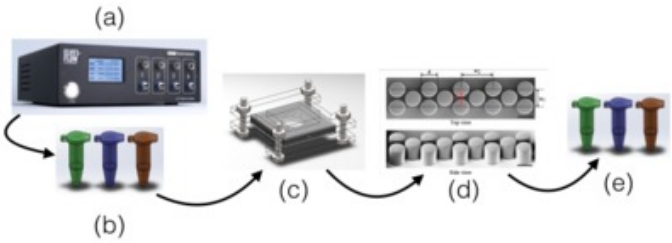
Present: Jiacomo

Goals: Propose full, augmented system

Content: see below

Conclusions/action items: While purely theoretical, the team feels confident that the system will work

JIACOMO BECKMAN - Apr 29, 2020, 12:52 PM CDT



Full_System.tiff(145.8 KB) - download

JIACOMO BECKMAN - Apr 29, 2020, 12:54 PM CDT

- To connect each segment, PTFE tubing will be used
- (a): OB1 multi-channel microfluidic flow controller exerts pressure into individual capsules (b)
    - (b) contains the separate protein solutions or cleaning agent
    - Upon optimal pressure induced, solution will flow out of the capsule and into the microfluidic device
- After ECM synthesis, the OB1 pump will continue to push unsynthesized solution through a built-in filtering mechanism in the microfluidic device (d)
    - Pillars allow broken pieces of ECM scaffold to be captured while allowing the rest of the unpolymerized solution to pass through
    - Collected in individual capsules (e)

Expenses from Fall 2019

JIACOMO BECKMAN - Feb 25, 2020, 10:45 PM CST

Title: Expenses from Fall 2019

Date: 2/25/20

Content by: Jiacomo

Present: n/a

Goals: Keep track of what monetary budget is left based on previous expenses.

Content:

Below is a table that contains everything that was purchased from last semester and the allocated cost. We did confirm with Dr. Campagnola that the budget is still $1500 and includes the expenses from last semester.

Conclusions/action items: We have about half of our total budget remaining for purchases.

JIACOMO BECKMAN - Feb 25, 2020, 10:47 PM CST

Item	Manufacturer	Part Number	Date	QTY	Cost Each	Total	Link
PTFE Tubing 1/16" OD x 1/32" ID (20 m)	Darwin Microfluidics	n/a	11/1/2019	1	$60.30	$60.30	LINK
Upchurch Precolumn MicroFilter	Sigma Aldrich	502685	11/11/2019	1	$128.00	$128.00	LINK
Piezoelectric Pump	Dolomite Microfluidics	3200138	11/11/2019	1	$340.00	$340.00	LINK
Microfluidic Reservoir	Darwin Microfluidics	n/a	11/11/2019	1	$130.00	$130.00	LINK
Custom 3D Printed Microfluidic Devices	Midwest Prototype	n/a	11/20/2019	1	$52.00	$52.00	n/a
Poster Print	n/a	n/a	12/5/2019	1	$48.00	$48.00	n/a
Poster Tube	n/a	n/a	12/5/2019	1	$3.10	$3.10	n/a
TOTAL:						**$761.40**	

Spring 2020 Expenses

JIACOMO BECKMAN - Apr 28, 2020, 9:01 PM CDT

Title: Spring 2020 Expenses

Date: 4/28/20

Content by: Jiacomo

Present: n/a

Goals: Have a written account of this current semester's expenses

Content:

As an official confirmation, there were no expenses made this current semester.

Conclusions/action items: n/a

Protein Adsorption Materials and Methods Procedure

JIACOMO BECKMAN - Feb 25, 2020, 10:54 PM CST

Title: Protein Adsorption Materials and Methods Procedure

Date: 2/25/20

Content by: Jiacomo

Present: Jiacomo, Nick, Bob

Goals: Do protein adsorption testing on the materials provided by Midwest Prototype

Content: Below is all of the materials used and the overall procedure that was conducted for the protein adsorption experiment. This was pulled directly from the report that I wrote for Midwest Prototype.

Conclusions/action items: See experimental results

JIACOMO BECKMAN - Feb 25, 2020, 10:51 PM CST

Materials Used

- Elastomeric polyurethane
- Silicone 30
- (6%-8%) Bovine serum albumin
- Eppendorf pipette
- 250mL glass beakers
- 96 well plate
- Biotek Synergy HT plate reader

Testing Methods

EPU-40 and SIL-30 were obtained and placed individually into a glass beaker. Approximately 4 mL of BSA was added to each of the two beakers containing the material and another empty beaker to serve as a control. After a 5 minute interval, an Eppendorf pipette was used to extract three samples from each beaker and placed into the well plate. The Biotek Synergy HT reader measured the maximum optical density of the samples at a wavelength of 280 nm. This process was repeated at a total elapsed time period of 10 minutes, 30 minutes, 1 hour, 2 hours, and 3 hours.

Testing Protocol for Luminescence

JIACOMO BECKMAN - Apr 29, 2020, 12:55 PM CDT

Title: Testing Protocol for Luminescence

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To explain the procedure for the luminescence test

Content:

1. **Pour luminescent liquid into a beaker**
2. **Place the Sil 30 and EPU 40 into the beaker**
3. **Make sure the materials are covered with liquid**
4. **Have a third beaker of just the luminescent liquid be a control**
5. **Take Sil 30 and EPU 40 out of liquid at different time intervals and examine with an electron microscope.**
6. **The time intervals will be 1 min, 5 min, 15 min, 30 min, 1 hour, 2 hours, and 3 hours.**
7. **Save the images of the materials under the electron microscope**
8. **Use ImageJ to measure the intensity of the luminescence**
9. **The material with a higher intensity of light allows proteins to adhere to it more**
10. **The material with the less intense light will be used in the final design.**

Conclusions/action items: Fabricate the pump so that testing can occur.

Testing protocol for flow rate test

JIACOMO BECKMAN - Apr 29, 2020, 12:56 PM CDT

Title: Testing protocol for flow rate test

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To explain the testing protocol for the flow rate test

Content:

1. **Turn the pump on**
2. **Run 5mL of water through the pump**
3. Start a timer when the pump is turned on
4. Stop the time when all of the water is about of the tubing
5. Divide the 5mL by the time to get the flow rate of the device
6. Alter the power to the pump to see the pump rate at different voltages
7. Use a voltage divider to lower the V out of the microcontroller
8. Repeat steps 1-6
9. Use an op-amp to amplify the signal
10. Repeat steps 1-6

Conclusions/action items: Build the pump so that this testing can occur.

Optical Density Graphs

JIACOMO BECKMAN - Feb 25, 2020, 10:57 PM CST

Title: Optical Density Graphs

Date: 2/2/20

Content by: Jiacomo

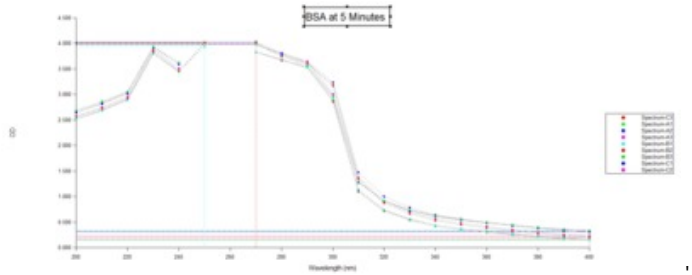
Present: Jiacomo, Bob, Nick

Goals: Attain the optical density of the samples in a graphical format.

Content: Below are the graphs of all the optical densities of each sample at their respective time points.

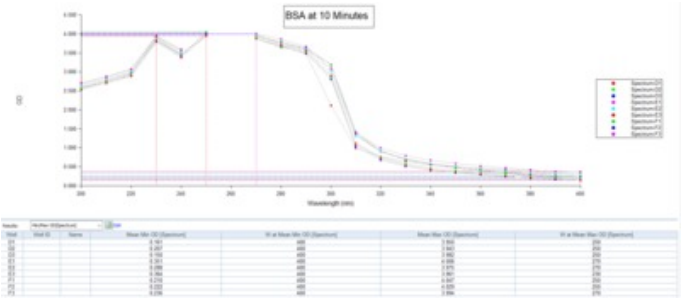
Conclusions/action items: See data analysis for more clear numbers and statistical calculations

JIACOMO BECKMAN - Feb 25, 2020, 10:58 PM CST



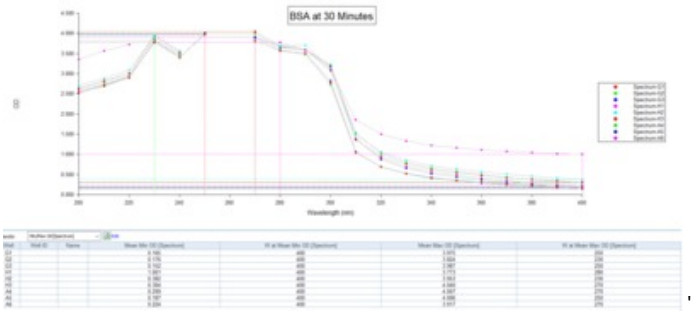
5_Min_Graph.PNG(31.2 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 10:58 PM CST



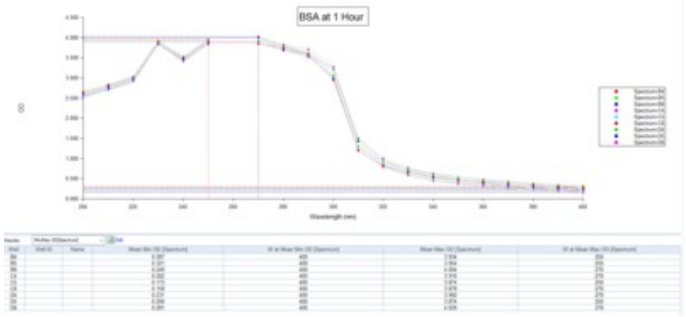
10_Min_Graph.PNG(43.8 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 10:59 PM CST



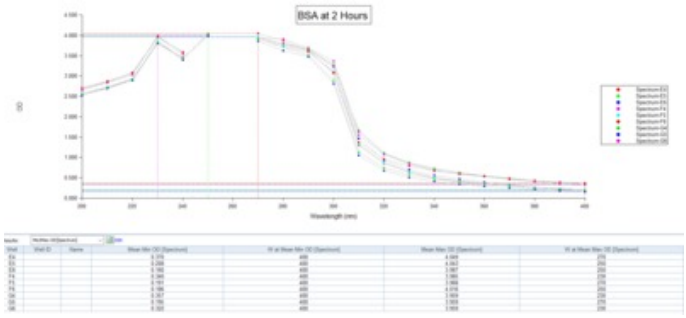
30_Minute_Graph.PNG(46 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 10:58 PM CST



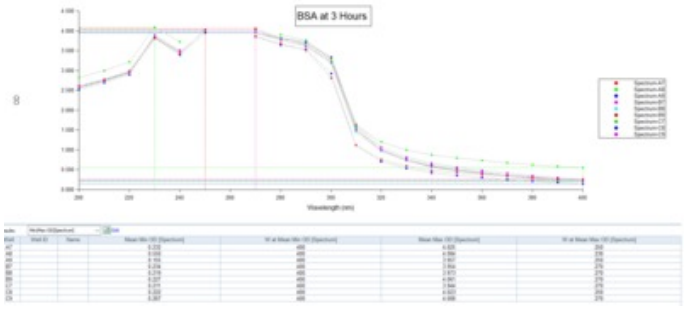
1_hour_Graph.PNG(43.4 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 10:58 PM CST



2_Hour_Graph.PNG(44.7 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 10:58 PM CST



3_Hour_Graph.PNG(44.1 KB) - download

Data Compilation and Analysis

JIACOMO BECKMAN - Feb 25, 2020, 11:02 PM CST

Title: Data Compilation and Analysis

Date: 2/8/20

Content by: Jiacomo

Present: Jiacomo

Goals: Compile the data into a readable format and analyze it.

Content: Below is all of the data obtained from the experiment. Also included is a table of statistical analysis. Given a significance level of 0.05, all of the t-tests fail to reject the null hypothesis. This thereby means that there is no significant difference between EPU-40, SIL-30, and the control BSA solution.

Conclusions/action items: We will most likely use EPU-40 anyway due to its rigidity compared to SIL-30

JIACOMO BECKMAN - Feb 25, 2020, 11:05 PM CST

Table of acquired optical densities of each sample at each time point

JIACOMO BECKMAN - Feb 25, 2020, 11:05 PM CST

Time Elapsed	Material/Control	Sample Maximum Optical Density		
		1	2	3
5 minutes	SIL 30	3.981	3.984	3.992
	EPU 40	3.94	4.021	4.001
	Control	4.028	3.99	4.01
10 minutes	SIL 30	3.95	3.943	3.982
	EPU 40	4.006	3.975	3.961
	Control	4.047	4.029	3.994
30 minutes	SIL 30	3.975	3.824	3.987
	EPU 40	3.773	3.953	4.04
	Control	4.007	4.006	3.917
1 hour	SIL 30	3.934	3.954	4.004
	EPU 40	3.91	3.874	3.878
	Control	3.992	3.874	4.029
2 hours	SIL 30	4.049	4.043	3.987
	EPU 40	3.985	3.968	4.016
	Control	3.959	3.959	3.959
3 hours	SIL 30	4.025	4.084	3.957
	EPU 40	3.954	3.973	4.061
	Control	3.944	4.023	4.006

Data.jpg(118.4 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 11:05 PM CST

Table of statistics

JIACOMO BECKMAN - Feb 25, 2020, 11:06 PM CST

Time Elapsed (min)	SIL-30 OD Averages	EPU-40 OD Averages	Control OD Averages
5	3.985666667	3.987333333	4.009333333
10	3.958333333	3.980666667	4.023333333
30	3.928666667	3.922	3.976666667
60	3.964	3.887333333	3.965
120	4.026333333	3.988966667	3.959
180	4.022	3.996	4.022
AVG	3.980833333	3.960383333	3.992555556
Standard Deviation	0.05549165279	0.06877435398	0.04452776911
Variance	0.003079323529	0.004729911765	0.001982722222
T-test (control)	0.6984126497	0.1743441487	n/a
T-test (comparing EPU 40 with SIL30)		0.3361640262	n/a

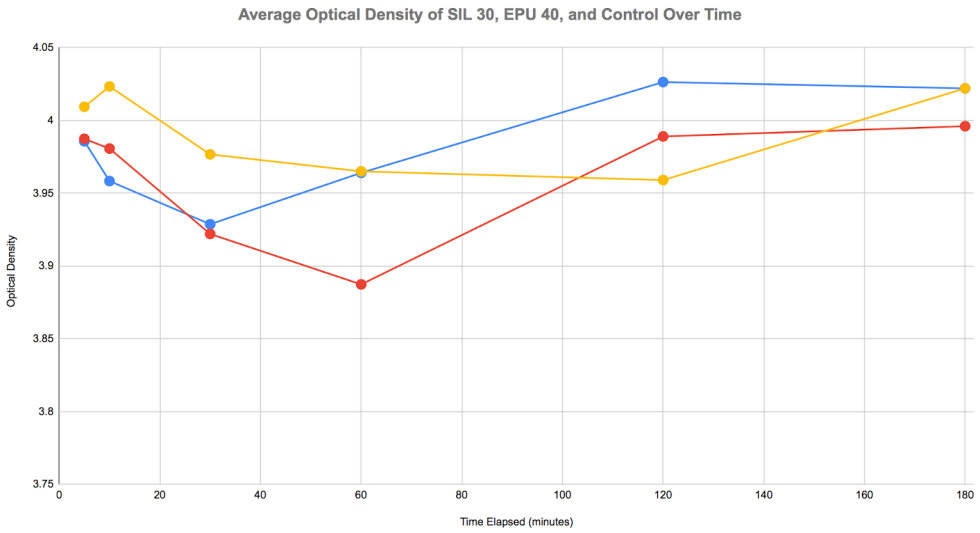
,

Stats.jpg(122.8 KB) - download

JIACOMO BECKMAN - Feb 25, 2020, 11:08 PM CST

Graph of average optical densities over time. Yellow, blue and red corresponds to control, SIL-30, and EPU-40 respectfully.

JIACOMO BECKMAN - Feb 25, 2020, 11:08 PM CST



Report Written for Midwest Prototype

JIACOMO BECKMAN - Feb 25, 2020, 11:12 PM CST

Title: Report Written for Midwest Prototype

Date: 2/15/20

Content by: Jiacomo

Present: Jiacomo

Goals: Analyze the results obtained from the experiment conducted and report them to Midwest Prototype.

Content: Below is an attachment to the document I wrote for Midwest Prototype. It contains essentially everything already included here in Lab Archives.

Conclusions/action items: n/a

JIACOMO BECKMAN - Feb 25, 2020, 11:12 PM CST

Protein Adsorption

An analysis of EPU-40 and SIL-30

Biomedical Engineering Design 301
Department of Biomedical Engineering
University of Wisconsin
February 15, 2020

Team Members:
Jason Wang
Kevin Koosar
Nicholas Pauly
Jiacomo Beckman

EPU_vs_SIL_Midwest-Prototype.pdf(759.9 KB) - download

Pressure Simulation

JIACOMO BECKMAN - Apr 29, 2020, 12:59 PM CDT

Title: Pressure Simulation

Date: 4/27/20

Content by: Nicholas Pauly, Kevin

Present: Nicholas Pauly

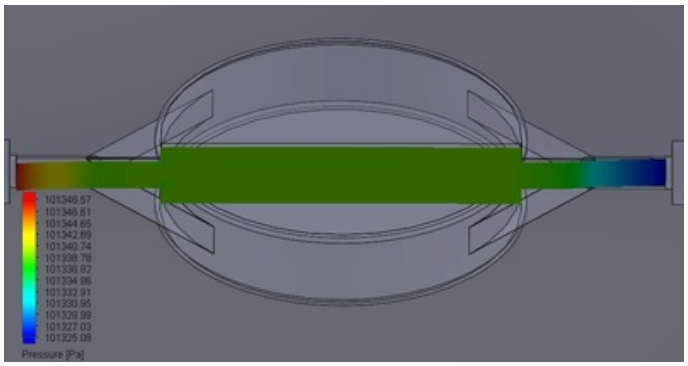
Goals: Theoretically determine pressure inside the microfluidic device if there is significant leakage

Content:

The pressure flow simulation shows pressure distribution inside the team's fabricated microfluidic device. The inlet hole of the device, on the left, has the greatest pressure because the pump will be pressurizing the tube at the beginning of the tubing. The outlet hole, on the right side of the image, will have the least amount of pressure because the majority of the pressure is dissipated within the microfluidic device. The center of the microfluidic device, where the reaction will take place, is at a constant pressure throughout. It shows that there are no high pressure areas inside the device that would cause leaking to occur. This allows the team to conclude that there will be no leaks in the fabricated clamp.

Conclusions/action items: Put on the poster and present to audience

JIACOMO BECKMAN - Apr 29, 2020, 12:59 PM CDT



unnamed.jpg(42.9 KB) - download

Fall 2019 - Final Notebook

Jason Wang - Jan 24, 2020, 5:12 PM CST

Title:

Date:

Content by:

Present:

Goals:

Content:

Conclusions/action items:

Jason Wang - Jan 24, 2020, 5:12 PM CST


BME Design-Fall 2019 - Jason Wang
Complete Notebook

PDF Version generated by

Jason Wang

on

Jan 24, 2020 @ 5:09 PM CST

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2020_01_24_notebook_82772.pdf(15.6 MB) - download

Original Paper

Jason Wang - Apr 29, 2020, 3:32 PM CDT

Title: Original Paper

Date:

Content by: Jason

Present:

Goals:

Content:

The link to the original paper that Dr. Campagnola provided to the group. https://www.osapublishing.org/oe/abstract.cfm?uri=oe-21-21-25346

Notes were taken on this in the previous semester's notebook

Conclusions/action items:

Soft Lithography - Fabrication of Microfluidic devices

Jason Wang - Oct 18, 2019, 7:40 PM CDT

Title: Soft Lithography - Fabrication of Microfluidic Devices

Date: 10/13/2019

Content by: Jason Wang

Present: Individual

Goals:

Content:

- Can be made of glass, silica, or polymer (PDMS)
- Simplest current design is a collection of micro-channels molded in a polymer that is bonded to a flat surface (glass slide)
    - Most common polymer is PDSM
        - transparent, biocompatible, deformable, and inexpensive
        - easy to mold and bond onto glass
- Design of Micro-channels
    - design made with dedicated software (AUTOCAD, Illustrator, LEDIT, ...)
    - it is transferred on a photomask: chrome coated glass plates or plastic films for the most common templates
    - The micro-channels are thus printed with UV opaque ink (if the substrate is a plastic film) or etched in chromium (if the substrate is a glass plate)
- Fabrication of microfluidic mold - Photolithography
    - Transfers microchannel patterns from the photomask into a real mold
    - (1) Resin is spread on a flat surface (often a silicon wafer) with the desired thickness (which determines the height of microfluidic channels)

      (2) The resin, protected by the photomask with the microchannel pattern, is then partially exposed to UV light. Therefore, in the case of a negative resin like SU-8 type, only the parts representing the channels are exposed to UV light and cured, the other parts of the mold being protected by the opaque areas of the mask.

      (3) The mold is developed in a solvent that etches the areas of resin that were not exposed to UV light.

      (4) We then obtain a microfluidic mold with a resin replica of the patterns from the photomask (future micro-channels make “reliefs” on the mold). The height of the channels is determined by the thickness of the original resin spread on the wafer. Most of the time, the mold is then treated with Silane to facilitate the release of microfluidic devices during molding steps (see next paragraph).

- Molding of the microfluidic chip
    - (1) The molding step allows mass production of microfluidic chips from a mold.

      (2) A mixture of PDMS (liquid) and cross-linking agent (to cure the PDMS) is poured into the mold and heated at a high temperature.

      (3) Once the PDMS is hardened, it can be taken off the mold. We obtain a replica of the micro-channels on the PDMS block.

      + Microfluidic device completion:

      (4) To allow the injection of fluids for future experiments, the inputs and outputs of the microfluidic device are punched with a PDMS puncher of the size of future connection tubes.

      (5) Finally, the face of the block of PDMS with micro-channels and the glass slide are treated with plasma.

      (6)The plasma treatment allows PDMS and glass bonding to close the microfluidic chip.

- Source:
    - “The Basics of Microfluidic Tubing & Sleeves,” Elveflow. [Online]. Available: https://www.elveflow.com/microfluidic-tutorials/microfluidic-reviews-and-tutorials/microfluidic-fittings-and-tubing-resources/the-basics-of-microfluidic-tubing-sleeves/. [Accessed: 19-Oct-2019].

Conclusions/action items:

- The method of fabrication of microfluidic devices using soft lithography. This could be used as a way to eliminate the separate well and incorporate the well into the structure.

PEG - Adsorption of Proteins

Jason Wang - Oct 21, 2019, 8:38 PM CDT

Title: Binding characteristics between polyethylene glycol (PEG) and proteins in aqueous solution

Date: 10/18/2019

Content by: Jason

Present: Individual

Goals:

Content:

- Abstract
    - Polyethylene Glycol (PEG)
        - well known antifouling material
        - little known about PEG-protein interactions
        - MW of PEG and mass ratio of PEG is observed to determine the effect
    - Results show that PEG with optimal MW is more capable of interacting with proteins
        - induces a conformational change of proteins through more stable binding sites and stronger interactions with long chain PEG
- Intro
    - Poly(ethylene glycol) (PEG) and poly(ethylene oxide) (PEO) are the most prevalent antifouling materials used for many biomedical and industrial applications against fouling processes
    - PEG-coated surfaces demonstrate superlow fouling ability to resist protein adsorption, cell adhesion, and even bacteria attachment
    - The resistance of PEG to the adsorption of proteins is strongly dependent on the formation of densely packed PEG brushes with high surface coverage and optimal brush thickness

Conclusions/action items:

- Stopped reading after introduction because the research being done was not relevant to our project. Just the material properties were relevant.

Methods for Studying Protein Adsorption

Jason Wang - Apr 29, 2020, 3:00 PM CDT

Title: Methods for Studying Protein Adsorption

Date: 2/26/2020

Content by: Jason

Present: Me

Goals: Learn methods for determining protein adsorption

Content:

- Spectroscopic Techniques
    - Fluorescence spectroscopy, infrared absorption (IR), Raman scattering, and circular dichroism (CD)
    - Fluorescence emission spectroscopy
        - Great specificity and sensitivity in protein studies
        - utilize both intrinsic protein fluorescence and a variety of extrinsic fluorophores attached to protein molecules.
        - most common way to achieve a surface sensitivity is to excite adsorbed protein fluorescence with an evanescent surface wave generated by the total internal reflection of the excitation beam.
        - Total internal reflection fluorescence spectroscpoy (TIRF) often used to study kinetics of protein adsorption
    - IR spectroscopy
        - region between 1100 and 1700 cm-1 can provide info on global prop. of the polypeptide conformation
        - amide I band located in wavenumber region between 1600 and 1700 cm-1
            - most frequently studies parameter in protein IR studies
- https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2664293/

Conclusions/action items:

Piezoelectric pump controller

Jason Wang - Feb 24, 2020, 10:01 PM CST

Title: Piezoelectric pump controller

Date: 2/12/2020

Content by: Jason

Present: Jason

Goals:

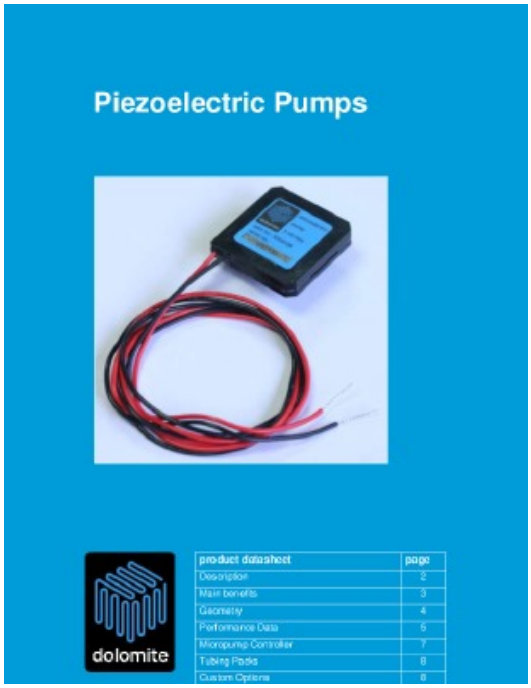
Content:

https://www.takasago-fluidics.com/products/products_pump/transfer/

- The company of the website above carries an AC voltage amplifier that is able to produce the Vpp and Hz signals necessary to power the piezoelectric pump.
    - The specifications needed to power the pump are located within the datasheet.

Conclusions/action items:

Jason Wang - Feb 24, 2020, 9:59 PM CST



Dolomite___PiezoelectricPumpsDatasheet___pdf.pdf(189.2 KB) - download

Possible pump controllers

Jason Wang - Apr 29, 2020, 3:16 PM CDT

Title: Possible pump controller information

Date:

Content by: Jason

Present:

Goals:

Content:

- Piezoelectric Micropump
  - Using an ac voltage amplifier produced by takasago-fluidics, We would be able to manually modulate the flow rate through our control of the outgoing waveform
- Elveflow
  - provided the team with a quote and recommendations for pumps/vacuum controller
  - AF1
    - standalone system
    - single pressure outlet
    - 200mb, 1600mb, -700 to 1000mbar
  - OB1
    - Upgradable
    - 1-4 pressure outlets
    - 5 ranges from -900mb to 8000 mb
    - requires and external pressure source
  - Vacuum vs pressure
    - Can use either or
  - Connection Kit
    - A starter pack containing luer connections/tubing that are needed to connect the OB1 to the external pressure source
  - Software
    - SDK is provided for LabVIEW, MATLAB, python, and C++
    - provided SDK and ESI software user guide
  - Shipping
    - 3 weeks after purchase order
  - Dimensions
    - OB1
      - 240x233x80 mm
    - AF1
      - 220x130x130 mm
  - Pressure source for OB1
    - The OB1 needs an external pressure source to be operational
      - Lab air supply, compressor, nitrogen tank, etc.
      - comprised between 1.5 and 10 bar

Conclusions/action items:

Filtering Method

Jason Wang - Apr 29, 2020, 3:27 PM CDT

Title: Microfluidic Pillar Filters

Date:

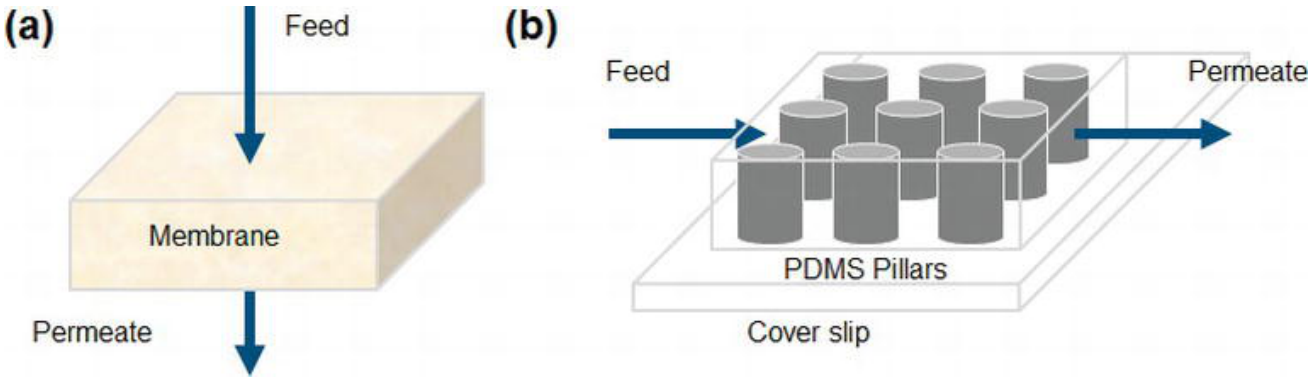
Content by: Jason Wang

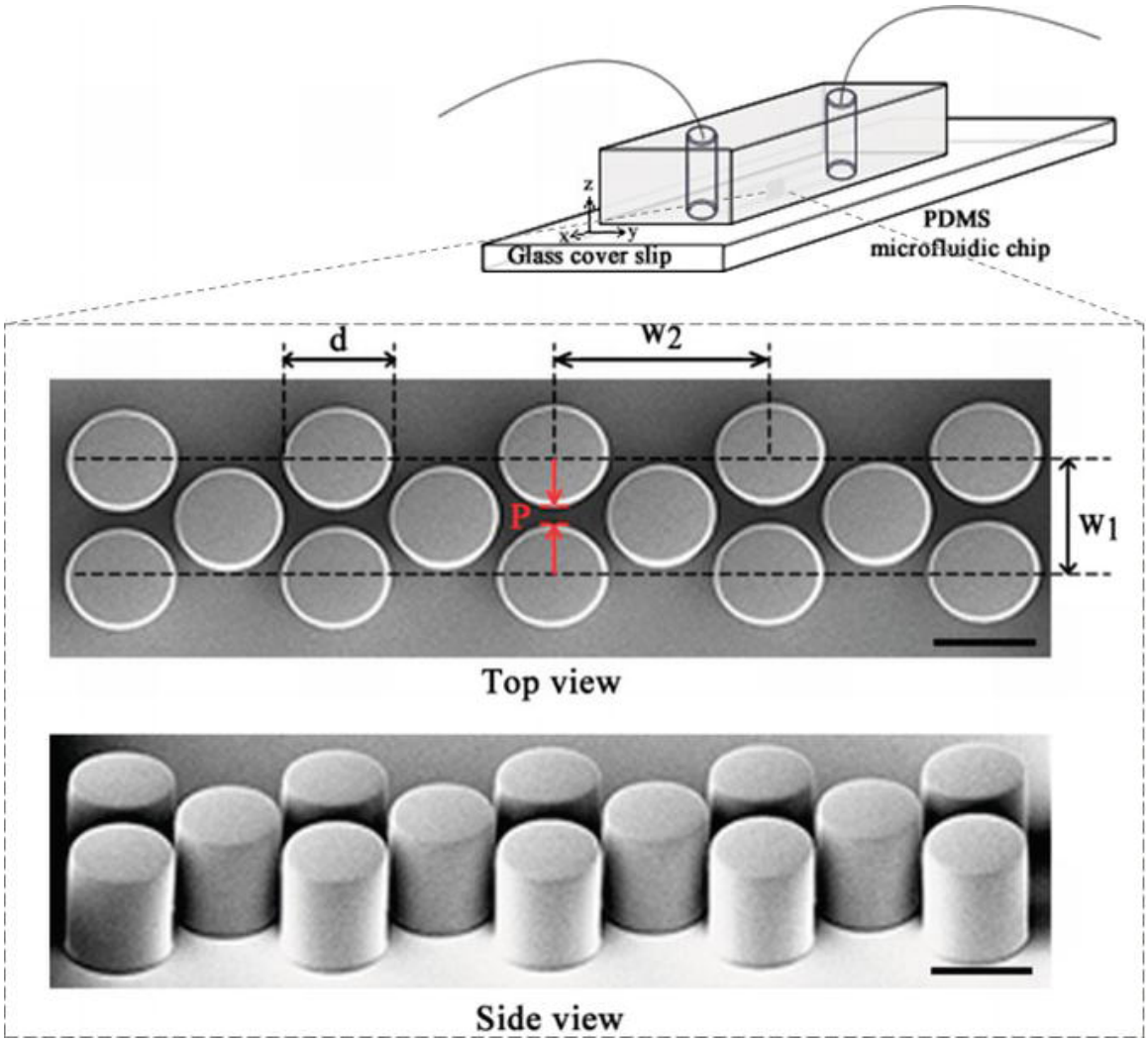
Present: Me

Goals: Learn about the pillar filters that could be implemented into the well chamber to help filter the excess proteins.

Content:

- Pillars are solid structures made out of PDMS
- Gap between the pillars are considered the pore
- Pillars are used as a microfluidic-based membrane mimic to counteract biofouling
- Possible Fabrication techniques:
    - photolithography
        - common technique when feature sizes are larger than 1um are desired
    - electron lithography
        - nanoscale features can be fabricated where the minimum resolution could go down to 10 nm
    - hot embossing
    - injection molding
- Actual fabrication techniques unclear





https://www.intechopen.com/books/microfluidics-and-nanofluidics/microfluidic-membrane-filtration-systems-to-study-biofouling

Conclusions/action items:

Preliminary Designs

Jason Wang - Feb 24, 2020, 10:02 PM CST

Title: Preliminary Designs

Date: 2/8/2020

Content by: Team

Present: Team

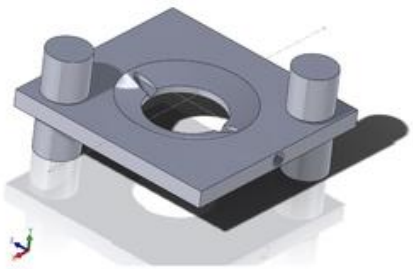
Goals:

Content:

- Figure 1:
  - Magnet Design
- Figure 2:
  - Weight Design
- Figure 3:
  - Clamped Design

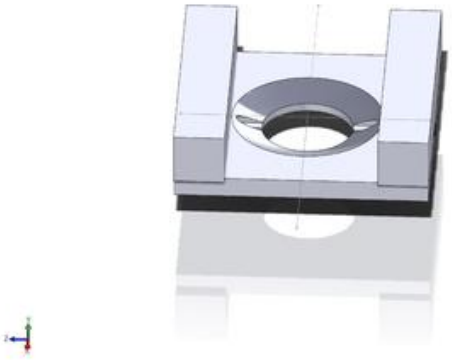
Conclusions/action items:

Jason Wang - Feb 24, 2020, 10:01 PM CST



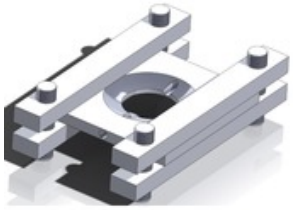
Magnet_Design_1_-page-001.jpg(212 KB) - download

Jason Wang - Feb 24, 2020, 10:01 PM CST



Weight_Design_1_-page-001.jpg(196 KB) - download

Jason Wang - Feb 24, 2020, 10:01 PM CST



clamp.png(252.1 KB) - download

Microfluidic design

Jason Wang - Apr 29, 2020, 3:18 PM CDT

Title: Revised Microfluidic Device

Date: 4/17/2020

Content by: Kevin, Jason

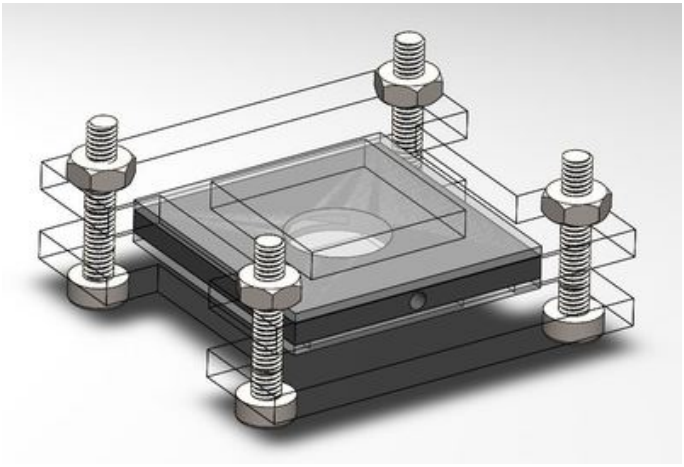
Present:

Goals: A modified version of the clamp design

Content:

Conclusions/action items:

Jason Wang - Apr 29, 2020, 3:17 PM CDT



'

Sandwich.JPG(90.2 KB) - download

Green Shop Pass

Jason Wang - Mar 08, 2019, 7:25 PM CST

Title: Green Shop Pass

Date: 03/08/2019

Content by: Me

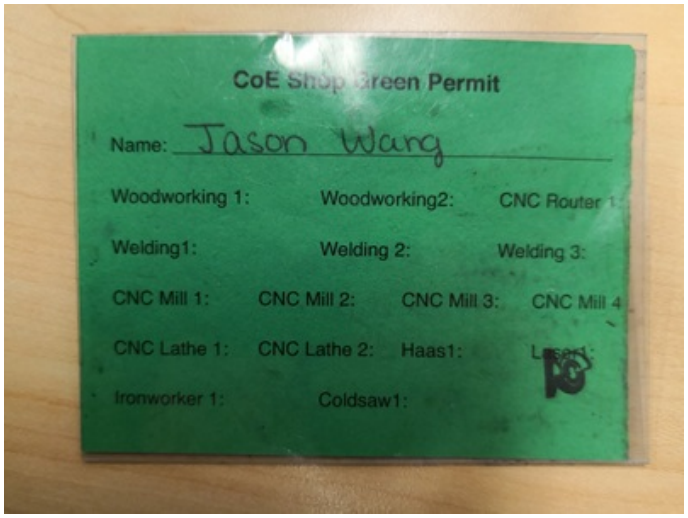
Present: Me

Goals:

Content:

Conclusions/action items:

Jason Wang - Mar 08, 2019, 7:29 PM CST



IMG_20190308_192752.jpg(2.5 MB) - download

Jason Wang - Mar 08, 2019, 7:29 PM CST

IMG_20190308_192817.jpg(2.6 MB) - download

BioSafety Training

Jason Wang - Mar 08, 2019, 7:42 PM CST

Title: Biosafety Training

Date: 03/08/2019

Content by: Me

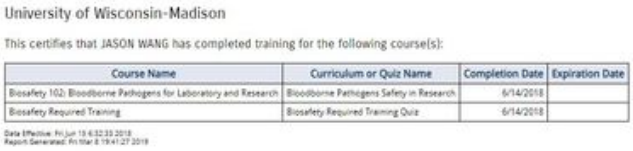
Present: Me

Goals:

Content:

Conclusions/action items:

Jason Wang - Mar 08, 2019, 7:43 PM CST



University of Wisconsin-Madison

This certifies that JASON WANG has completed training for the following course(s):

Course Name	Curriculum or Quiz Name	Completion Date	Expiration Date
Biosafety 102: Bloodborne Pathogens for Laboratory and Research	Bloodborne Pathogens Safety in Research	6/14/2018	
Biosafety Required Training	Biosafety Required Training Quiz	6/14/2018	

Data Effective: Fri Jun 15 6:32:33 2018
Report Generated: Fri Mar 8 19:41:27 2019

Capture.JPG(40.8 KB) - download

Microfluidic Pump

ROBERT MEULER - Feb 25, 2020, 10:01 PM CST

Title: Microfluidic Pump

Date: 2/25/2020

Content by: Bob Meuler

Present: Me

Goals: Research pump specs and find alternative

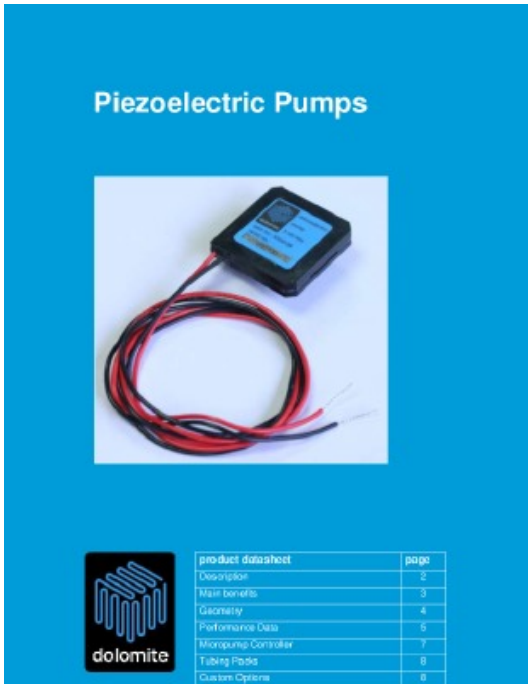
Content:

The dolomite microfluidic pump was purchased last semester for this project. Along with it was the suggested controller for $1000 that is out of the scope of our budget. The pump needs to work within a range of 60-250 Vp-p and an applied frequency range of 10-60 Hz. One controller that Dr. Nimunkar recommended to our team was the Driver Board MPD-200A. This board requires an input voltage of only 5 V DC. It outputs the required frequency from 1-60 Hz and voltage of 50-340 Vp-p. It is small and compact, weighing only 9.0 g which will be useful for our project.

Conclusions/action items:

Purchase Driver Board MPD-200A and begin testing

ROBERT MEULER - Feb 25, 2020, 9:51 PM CST



Dolomite___PiezoelectricPumpsDatasheet___pdf.pdf(189.2 KB) - download

ROBERT MEULER - Feb 25, 2020, 9:51 PM CST

Driver Board MPD-200A

The MPD-200A is a thin, compact and lightweight driver board. It is a high voltage circuit board, specifically designed for the piezoelectric micro pumps. From a regular 5 VDC input, it readily generates approximately the 250 Vp-p, 40 Hz necessary for driving the pump.

Item	Specifications
Input voltage	5 VDC ±5%
Output frequency	1 ~ 60 Hz
Output voltage	50 ~ 340 Vp-p
The maximum number of pumps	2 pieces
External dimensions	30 x 30 x 18 mm
Weight	Approximately 9 g

MPD_200A.png(376.2 KB) - download

Filters

ROBERT MEULER - Feb 25, 2020, 10:33 PM CST

Title: Filters

Date: 2/25/2020

Content by: Bob Meuler

Present: Me

Goals: Find possible filters for project

Content:

Currently the plan is to directly filter excess protein solution/debris from the output line. This would need a pre-filter before the main filter, which would need replacement often. Alternative ideas include a filter system post extraction. One option is a cell strainer, which is very easy to use. These are relatively inexpensive at around $138 for 50. These would be a one time use/disposable item.

https://www.fishersci.com/us/en/browse/90184183/cell-strainers

Micropipette filters can also be used in a similar method that the cell strainers are used. These are one time use and easily disposable filters that are able to filter out our proteins. These are inexpensive in bulk, at around $172-$190 for 960 filters.

https://www.thomassci.com/Laboratory-Supplies/Pipettor-Tips/_/Filter-Pipette-Tips1?q=Pipette%20Filter

Conclusions/action items:

These will be good for easy/quick testing if a more permanent filter is not found.

Flow Rate

ROBERT MEULER - Apr 29, 2020, 10:22 AM CDT

Title: Flow Rate within tubing

Date: 4/29/2020

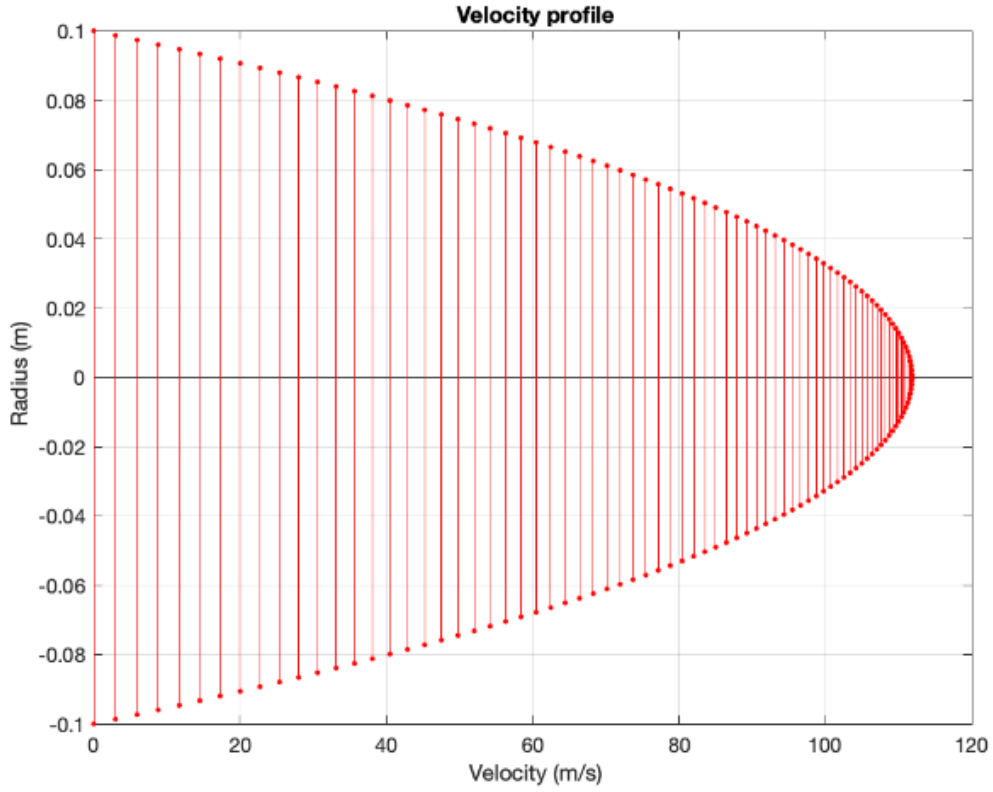
Content by: Bob Meuler

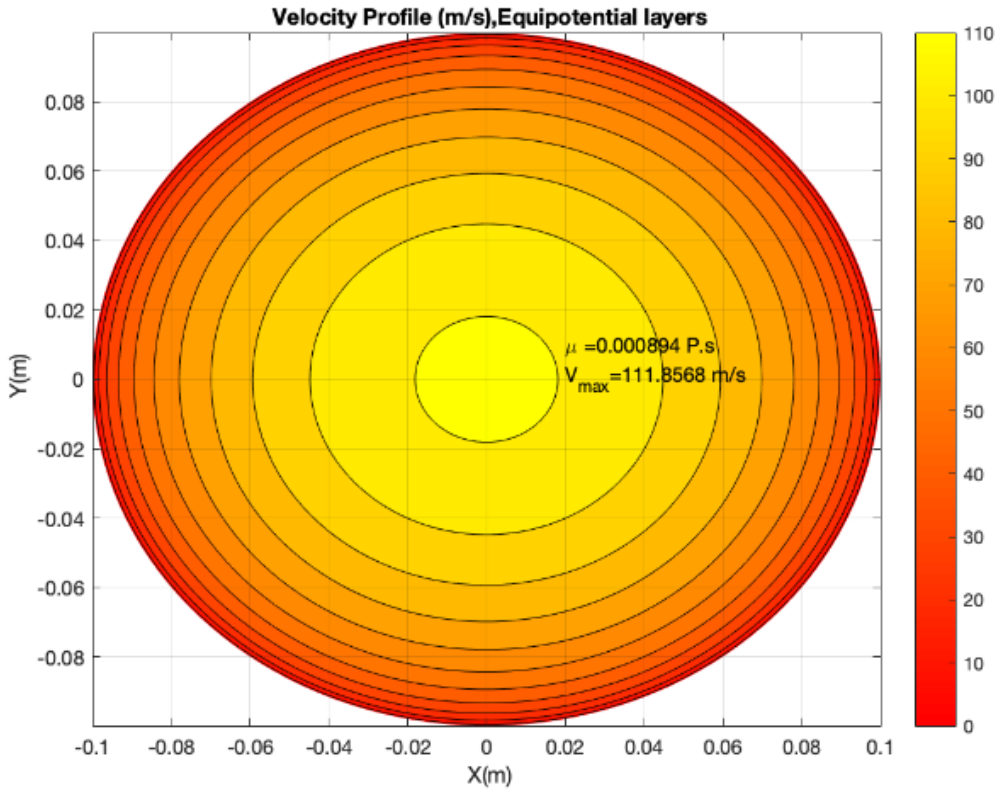
Present: Me

Goals: Look at velocity profile within tubing for the microfluidic device

Content:

This MatLab program calculates the velocity profile through a tubing with a few given inputs. Given the radius of the tubing, pressure at two points of the tube, viscosity of solution, and length of the tube, the velocity profiles can be calculated. These velocity profiles, such as max and mean velocity, can help to provide some knowledge of what the proteins are like going through the tubing into the chamber. It can tell us if they are of laminar flow or turbulent flow. This can help us with deciding the rate of the pump to be used in order to get the best movement into the well. Below are two of the figures that this code also displays, showing the velocity profiles from a side view as well as a head on view.





https://www.mathworks.com/matlabcentral/fileexchange/42186-viscous-flow-in-pipe-velocity-profile#overview_tab

Conclusions/action items:

This code is helpful for deciding the rate at which the pump will run, and provides an intuitive display for the velocity profiles of the proteins solution from within the tubing into the well chamber.

Pillar Filters

ROBERT MEULER - Apr 29, 2020, 11:33 AM CDT

Title: Microfluidic Pillar Filters

Date: 4/29/2020

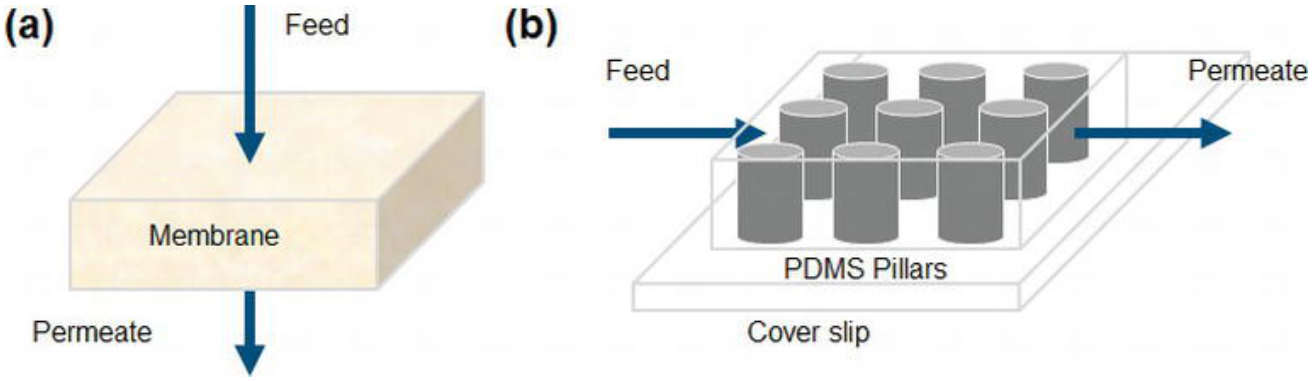
Content by: Bob Meuler

Present: Me

Goals: Learn about the pillar filters that could be implemented into the well chamber to help filter the excess proteins.

Content:

These pillar filters are made of PDMS and can be fabricated using photolithography techniques, and can then be bonded to our device easily. This filter system would be placed immediately after the well chamber where it can efficiently contain the extra proteins. It will effect the overall flow rate going out of the system, but this should not be a problem if a vacuum is implemented to help suck out the proteins on the output portion.



https://www.intechopen.com/books/microfluidics-and-nanofluidics/microfluidic-membrane-filtration-systems-to-study-biofouling

Conclusions/action items:

This pillar filter system looks promising for our design. A post-filtration collection system would still be required to gather the re-usable proteins, but that was of our original plan anyways.

Green Pass

ROBERT MEULER - Feb 25, 2020, 10:23 PM CST

Title: Bob Meuler

Date: 2/25/2020

Content by: Bob Meuler

Present: Bob Meuler

Goals: Acquire Green Permit

Content:

Acquired

Conclusions/action items:

ROBERT MEULER - Mar 11, 2019, 9:58 PM CDT

IMG_1743.HEIC(1.7 MB) - download

Biosafety

ROBERT MEULER - Mar 24, 2019, 10:52 PM CDT

Title: Biosafety Training

Date:

Content by: Bob Meuler

Present:

Goals:

Content:

Conclusions/action items:

ROBERT MEULER - Mar 24, 2019, 10:51 PM CDT

University of Wisconsin-Madison

This certifies that ROBERT MEULER has completed training for the following course(s):

Course Name	Curriculum or Quiz Name	Completion Date	Expiration Date
Biosafety Required Training	Biosafety Required Training Quiz	3/7/2019	

Data Effective: Fri Mar 8 7:26:55 2019
Report Generated: Sun Mar 24 22:48:15 2019

Biosafety.Training.Meuler.png(72 KB) - download

2020/2/12 Luminescent materials

Nicholas Pauly - Feb 12, 2020, 9:18 PM CST

Title: Luminescent materials

Date: 2/12/2020

Content by: Nicholas Pauly

Present: Nicholas Pauly

Goals: Describe materials that could be used in luminescent tests for the materials

Content:

ZInc Sulfite

https://cosmeticsinfo.org/ingredient/luminescent-zinc-sulfide-0

- a colorant that glows in the dark
- may not even need a microscope
- will produce a yellow green light

Strontium Aluminate

- glow in the dark compound
- greenish yellow color
- is expensive on sigma aldrich
- but can be found for cheap on amazon

https://www.thoughtco.com/what-glows-under-a-black-light-607615

Vitamin A and the B vitamines thiamine, niacin, and riboflavin are strongly fluorescent.

Chlorophyll makes plants green, but it also fluoresces a blood-red color. Grind some spinach or swiss chard in a small amount of alcohol (e.g., vodka or Everclear) and pour it through a coffee filter to get chlorophyll extract

Other possibilities are to ask Sam from the lab if they have any fluorescent materials, and then run them through the microfluidic device

Conclusions/action items: Talk to Sam and the team about using these materials after the microfluidic devices is functional

2020/2/24 Possible AC/DC converters

Nicholas Pauly - Feb 24, 2020, 9:11 PM CST

Title: Possible AC/DC converters

Date: 2/24/20202

Content by: Nick Pauly

Present: Nick Pauly

Goals: To research alternatives for the microcontroller

Content:

https://www.digikey.com/product-detail/en/sl-power-electronics-manufacture-of-condor-ault-brands/GSC20-5G/271-2362-ND/1036615

- Reaches the desired voltage peak to peak
- is an AC to DC converter
- unsure about modified sine wave
- only 64$

https://www.digikey.com/product-detail/en/mean-well-usa-inc/HRP-600-48/1866-2736-ND/7704324

- AC to DC converter
- 150$ for one
- reaches the desired voltage
- 12V
- unsure about sine wave

https://www.digikey.com/product-detail/en/mean-well-usa-inc/RSP-1000-48/1866-4219-ND/7706283

- AC to DC converter
- more expensive (-200$)
- 48 V
- reaches the desired peak to peak

Conclusions/action items: Speak with Dr. Nimunkar about these possibly working for the project

2020/2/25 Possible Filters for output

Nicholas Pauly - Feb 25, 2020, 10:07 PM CST

Title: Possible Filters for the output

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To look for some microfilters to be used in the outout

Content:

https://scientificfilters.com/nylon-syringe-filters-sf13829?gclid=Cj0KCQiAqNPyBRCjARIsAKA-WFymWzEEA-wC6Gf0nthnyXgWT-eRCWNt3RecA2Cl40v0MUNV8BXorSoaAnKxEALw_wcB

- nylon syringe filter
- hydrophilic
- used for solvent-based filtration
- $83 for 100
- 0.22 micrometers and larger filtered out

https://shop.pall.com/us/en/laboratory/0-2-m-gamma-irradiated-3/pkg--zid12201?gclid=Cj0KCQiAqNPyBRCjARIsAKA-WFzJCx6Izx9RgH29kw3d5-fINa689ZFczxwZfBK4NFd6AWhEMUESzpUaAldCEALw_wcB

- compatible with aqueous and organic solvents
- suitable for viscous liquids
- 0.1 to 0.2 micrometer pore size
- 20 filters
- sterile

https://scientificfilters.com/polytetrafluoroethylene-ptfe-disk-filters-df15274?gclid=Cj0KCQiAqNPyBRCjARIsAKA-WFyE58V86pltXOb5PjCWTM-9ltks0ZHYm8A_T97-hulYLcohS6WbYaYaAv1sEALw_wcB

- PTFE disk filters
- 0.22 micrometer pore size
- 10 per pack
- hydrophilic
- nonsterile
- $38 per pack with $16 shipping

Conclusions/action items: Talk with the team about these filters before purchasing. Talk with an advisor to see if they would work.

2020/4/27- Pillar Filters

Nicholas Pauly - Apr 27, 2020, 8:53 PM CDT

Title: Pillar FIlter Research

Date: 4/27/20

Content by: Nicholas Pauly

Present: Nicholas Pauly

Goals: Find pillar filters that could work in our final product

Content:

https://www.intechopen.com/books/microfluidics-and-nanofluidics/microfluidic-membrane-filtration-systems-to-study-biofouling

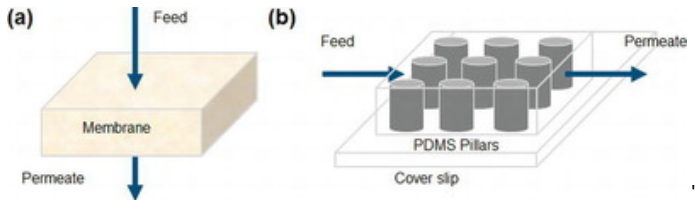
- pores in between filters
- has a membrane on top of the filters
- help filter out the solution
- flow rate affected
- can be assembled based on what we need

https://link-springer-com.ezproxy.library.wisc.edu/article/10.1007/s10544-007-9085-z

- different shapes of pillars matters
- in this paper cells lost are diminished
- increased the distances between the filters
- does not say how to make, just about the effectiveness

Conclusions/action items: Discuss with the team the best way to implement the pillar filter design

Nicholas Pauly - Apr 27, 2020, 8:53 PM CDT



BME_301_Pillar_Filter.png(91.4 KB) - download

2020/2/25- Beebe Paper on Microfluidics

Nicholas Pauly - Feb 25, 2020, 10:52 PM CST

Title: Beebe Paper on Microfluidics

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To discuss the role of microfluidics and some competing designs

Content:

https://www-nature-com.ezproxy.library.wisc.edu/articles/nature13118.pdf

- allows for rapid sample processing
- a popular candidate to replace traditional procedures
- aim to replace traditional assays
- microscale properties differ a lot from the macroscale properties
- mostly used for immunology and blood sample work
- chemotaxic assays are becoming popular
    - this creates a concentration gradient
- new materials are being used PDMS
    - thermoplastic materials are also being researched
- low cost helps this field
- allows for more relevant in vitro models

Conclusions/action items: Use these ideas when trying to develop our own microfluidic device

2020/4/27- Possible pumps or Microcontrollers

Nicholas Pauly - Apr 27, 2020, 9:06 PM CDT

Title: Possible pumps or microcontrollers

Date: 4/27/20

Content by: Nicholas Pauly

Present: Nicholas Pauly

Goals: To show research pumps and microcontrollers

Content:

https://www.dolomite-microfluidics.com/product/piezoelectric-pump-controller/

- dolomite piezoelectric pump controller
- works with our pump that has already been purchased
- 1100$
- would put the project over budget, but if absolutely needed will talk with the client about price

https://www.elveflow.com/microfluidic-products/microfluidics-flow-control-systems/ob1-pressure-controller/

- whole new pump
- is a controller as well as a pump
- has multiple input ports
- can be integrated with LabVIEW and easily programmable
- needs a flow sensor
- a compressor for pressure
- Elveflow's starter kit
- Everything needed was quoted at 5,400 euros
    - this is exactly what is needed the team cannot afford it

Conclusions/action items: Discuss with the team the options and then talk to our client.

Nicholas Pauly - Apr 27, 2020, 9:10 PM CDT

BME_301_Dolomite_Pump.png(29.3 KB) - download The first image is the microcontroller from Dolomite. The second image is the OB1 pump/controller from Elveflow

Nicholas Pauly - Apr 27, 2020, 9:10 PM CDT

,

BME_301_OB1_Pump_Elveflow.jpg(55 KB) - download The first image is the microcontroller from Dolomite. The second image is the OB1 pump/controller from Elveflow

2020/02/25 Testing Protocol for Luminescence

Nicholas Pauly - Feb 25, 2020, 10:23 PM CST

Title: Testing Protocol for Luminescence

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To explain the procedure for the luminescence test

Content:

1. **Pour luminescent liquid into a beaker**
2. **Place the Sil 30 and EPU 40 into the beaker**
3. **Make sure the materials are covered with liquid**
4. **Have a third beaker of just the luminescent liquid be a control**
5. **Take Sil 30 and EPU 40 out of liquid at different time intervals and examine with an electron microscope.**
6. **The time intervals will be 1 min, 5 min, 15 min, 30 min, 1 hour, 2 hours, and 3 hours.**
7. **Save the images of the materials under the electron microscope**
8. **Use ImageJ to measure the intensity of the luminescence**
9. **The material with a higher intensity of light allows proteins to adhere to it more**
10. **The material with the less intense light will be used in the final design.**

Conclusions/action items: Fabricate the pump so that testing can occur.

2020/2/25 Testing Protocol for Flow Rate Test

Nicholas Pauly - Apr 09, 2020, 8:14 PM CDT

Title: Testing protocol for flow rate test

Date: 2/25/2020

Content by: Nick Pauly

Present: Nick Pauly

Goals: To explain the testing protocol for the flow rate test

Content:

1. **Turn the pump on**
2. **Run 5mL of water through the pump**
3. Start a timer when the pump is turned on
4. Stop the time when all of the water is about of the tubing
5. Divide the 5mL by the time to get the flow rate of the device
6. Alter the power to the pump to see the pump rate at different voltages
7. Use a voltage divider to lower the V out of the microcontroller
8. Repeat steps 1-6
9. Use an op-amp to amplify the signal
10. Repeat steps 1-6

Conclusions/action items: Build the pump so that this testing can occur.

2020/4/27- Velocity and Pressure Equations

Nicholas Pauly - Apr 27, 2020, 8:29 PM CDT

Title: Velocity and Pressure Equations for inside the tubing

Date: 4/27/20

Content by: Nicholas Pauly

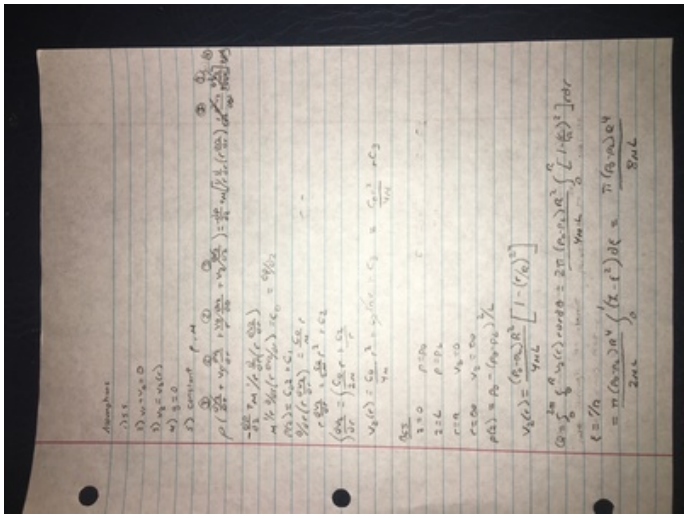
Present: Nicholas Pauly

Goals: Give theoretical results of what the fluid is doing inside the tubing

Content:

Conclusions/action items: Discuss with the team to put on final poster

Nicholas Pauly - Apr 27, 2020, 8:29 PM CDT



IMG_1120.JPG(2.5 MB) - download

2020/4/27- Pressure Flow Diagram

Nicholas Pauly - Apr 27, 2020, 8:33 PM CDT

Title: Pressure Flow Diagram

Date: 4/27/20

Content by: Nicholas Pauly

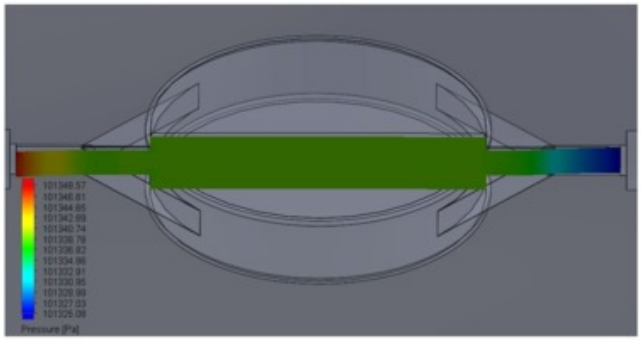
Present: Nicholas Pauly

Goals: Display the pressure-flow diagram generated off of the velocity and pressure equations

Content:

Conclusions/action items: Put on the final poster and present the team's findings

Nicholas Pauly - Apr 27, 2020, 8:36 PM CDT



BME_301_Pressure_Flow_Diagram.PNG(233.1 KB) - download

2020/4/27- Final Design

Nicholas Pauly - Apr 27, 2020, 8:38 PM CDT

Title: Final Design

Date: 4/27/20

Content by: Nicholas Pauly

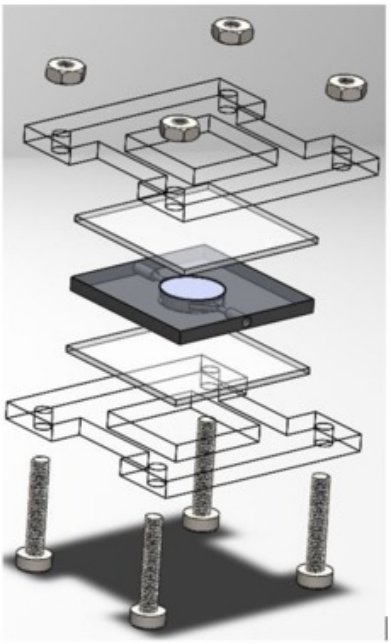
Present: Nicholas Pauly

Goals: Present the final design

Content:

Conclusions/action items: Put on the poster and present to audience

Nicholas Pauly - Apr 27, 2020, 8:39 PM CDT



BME_301_Final_Design.PNG(178.3 KB) - download

2020/4/27- Testing Results for Protein Adsorption

Nicholas Pauly - Apr 27, 2020, 9:27 PM CDT

Title: Testing Results for Protein Adsorption

Date: 4/27/20

Content by: Nicholas Pauly

Present: Nicholas Pauly

Goals: To display the results from the protein adsorption testing

Content:

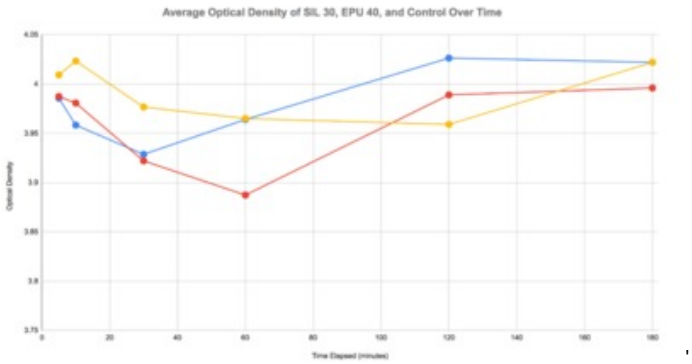
- EPU-40 and SIL-30 were obtained and placed individually into a glass beaker.
- Approximately 4 mL of BSA was added to each of the two beakers containing the material and another empty beaker to serve as a control.
- After a 5 minute interval, an Eppendorf pipette was used to extract three samples from each beaker and placed into the well plate.
- The Biotek Synergy HT reader measured the maximum optical density of the samples at a wavelength of 280 nm.
- This process was repeated at a total elapsed time period of 10 minutes, 30 minutes, 1 hour, 2 hours, and 3 hours

A t-test was done at the end to determine if there was any statistical significance between the two

There was no statistical significance

Conclusions/action items: Report the findings to Midwest Prototype and display the results

Nicholas Pauly - Apr 27, 2020, 9:27 PM CDT



BME_301_Protein_Adsorption_Graph.PNG(67.1 KB) - download

Nicholas Pauly - Apr 27, 2020, 9:27 PM CDT

Time Elapsed (min)	SIL-30 OD Averages	EPU-40 OD Averages	Control OD Averages
5	3.985666667	3.987333333	4.009333333
10	3.958333333	3.980666667	4.023333333
30	3.928666667	3.922	3.976666667
60	3.964	3.887333333	3.965
120	4.026333333	3.988966667	3.959
180	4.022	3.996	4.022
AVG	3.980833333	3.960383333	3.992555556
Standard Deviation	0.05549165279	0.06877435398	0.04452776911
Variance	0.003079323529	0.004729911765	0.001982722222
T-test (control)	0.6984126497	0.1743441487	n/a
T-test (comparing EPU 40 with SIL30)		0.3361640262	n/a

BME_301_Protein_Adsorption_Table.PNG(84.6 KB) - download

2020/2/25 Green Pass

Nicholas Pauly - Feb 25, 2020, 10:10 PM CST

Title: Green Pass

Date: 2/25/2020

Content by: Nick Pauly

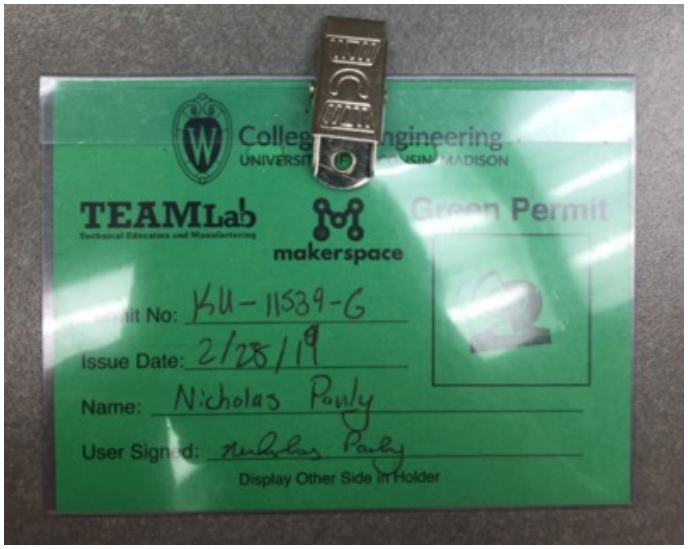
Present: Nick Pauly

Goals: To display my green pass

Content:

Conclusions/action items:

Nicholas Pauly - Feb 25, 2020, 10:13 PM CST



Green_Pass.png(746.4 KB) - download

2020/26/02-Client Purpose - Cancer ECM

KEVIN KOESSER - Feb 26, 2020, 3:11 PM CST

Title: Cancer ECM

Date: 2/26/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To understand why Dr. Campagnola images and builds ECM

Content:

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5998835/

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5294710/

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5073303/

Fabrication of three-dimensional multi-protein microstructures for cell migration and adhesion enhancement

While it is largely irrelevant to the specifications of the device the client has commissioned, the biological background of the project is interesting and provides extra motivation to manufacture an effective prototype. The four articles above provide a comprehensive survey of the client's research using second harmonic generation microscopy over the past several years. In general, this form of microscopy has elucidated differences between healthy and cancerous ovarian extracellular matrix.

Conclusions/action items:

By understanding the nature of the cancerous ECM, scientists can look into treatments for cancer by manipulating the ECM. The client believes that such differences between healthy and cancerous tissue are likely found in other cancers besides just ovarian cancer.

2020/26/02-Pump Controller

KEVIN KOESSER - Feb 26, 2020, 2:47 PM CST

Title: Piezoelectric Pump controller

Date: 02/26/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To find a controller for the piezoelectric pump and document the website source.

Content:

https://www.takasago-fluidics.com/products/products_pump/transfer/

Last semester, the group tried to find a pump and controller system that was accurate, affordable, and compatible with Labview. Unfortunately the team ran into budget constraints for the controller and began looking into other options. After discussing with Dr. Nimunkar last semester and this semester we have decided to purchase the driver board pictured above to simply create our own controller (without paying thousands of dollars).

The driver board generates a highly-amplified AC square wave to power the pump. It contains RC circuits which controls the rate of current applied to the pump. Purchasing this driver board would be significantly cheaper than building our own, but it may be difficult to operate within the preexisting Labview framework.

Conclusions/action items:

In the future, we hope to integrate this controller into the client's current Labview framework using an Arduino microcontroller.

2020/26/02-Filtering

KEVIN KOESSER - Feb 26, 2020, 3:01 PM CST

Title: Filtering options

Date: 2/26/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To compile all the options for filtering post hoc

Content:

Last semester, the team considered several filtration methods to remove chunks of ECM that break off during fabrication. The design that the team chose incorporated a thin slide with a circular filter opening within it to filter precipitate out of the protein solution downstream of the reaction chamber. This design included a gap in the structure for the slide to move across so that the slide can line up the opening with individual output channels. The filter within the slide apparatus would be removable via an insert similar to that of a SIM card tray of a phone, permitting simple filter replacement. Despite having a relatively simple method of filtering out the precipitate from the protein solution, the production of the slide apparatus would be difficult due to the scale that the piece would have to be built to (especially with the inner removable tray that would house the filter). On these grounds, the team never pursued fabrication of this device but rather bought a filter that could attach to the output tubing. The team has since reconsidered these options, since including the filter as part of the microfluidic system makes the device prone to clogging and developing back pressure that could harm the pump.

Therefore, a post hoc filtration procedure is a more attractive design for this device. However, there are also limitations to this technique: primarily, the volumes of fluid that the client needs to filter are typically between 30 and 50 microliters, so a significant amount of the fluid would be 'held up' on the filter membrane after use. To limit hold-up volume, researchers may use smaller filters, low-surface area membranes, and aid filtration by adding positive pressure. The following product uses centrifugation to force small volumes of liquid through the filter.

https://www.sigmaaldrich.com/catalog/product/sigma/cls8160?lang=en&region=US

Additionally, one could use a syringe filter, as shown below. This method is the most realistic because it is cheaper and the manufacturer included a procedure describing how to limit hold-up volume to about 5 microliters.

https://shop.pall.com/us/en/laboratory/analytical-qc/lcms-sample-preparation/acrodisc-ms-syringe-filter-zidMS-3301

https://www.pall.com/content/dam/pall/laboratory/literature-library/non-gated/13-8478_Acrodisc_13mm_AN.pdf

Conclusions/action items:

The team will suggest using the cheaper Pall syringe filter (adhering to the manufacturer's guidelines for filtering small volumes); the centrifuge filter manufacturer does not provide information on hold-up volume. If one attains 5 microliter hold-up volume on a 50 microliter protein solution, then that is a 10% loss, which is better than the client's current loss of about 70%.

2020/04/10-More filtering options

KEVIN KOESSER - Apr 29, 2020, 12:59 PM CDT

KEVIN KOESSER - Apr 29, 2020, 1:33 PM CDT

Title: Filtering options - a second look

Date: 04/10/2020

Content by: Kevin Koesser

Present: Kevin Koesser

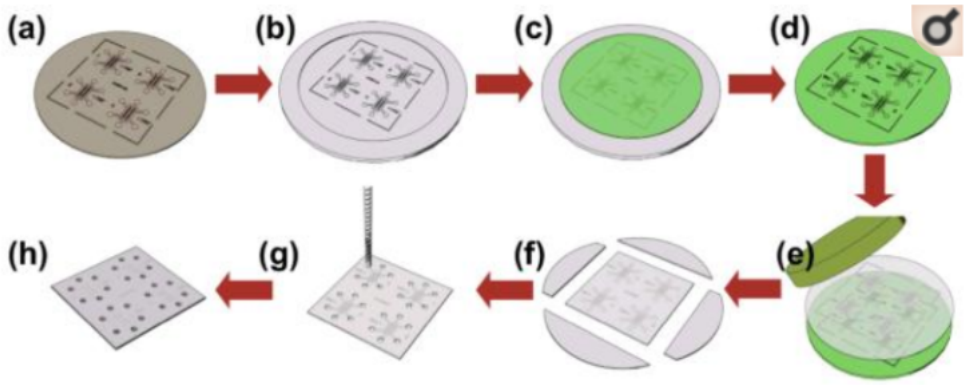
Goals: To compile all the options for filtering in real time

Content:

In the last filtering entry, I summarized the potential use of post hoc filtration methods to remove chunks of scaffold in the protein solution upon exiting the polymerization chamber. The pros of post hoc filtration include 1) the filter does not clog the microfluidic system after repeated use 2) the proposed filters (syringe (preferred) and centrifuge) are commercially available and do not involve accessing a fabrication lab. The cons of post hoc filtration include: 1) it removes automation from the system, which is one of the primary goals of the project, 2) filters are expensive and disposable.

To mitigate the cons, I looked into real time filtration again. There are examples in the literature that prove the efficacy of microfluidic filtration using micrometer-scale PDMS pillars. In the linked article, Biswas et al. build and test the pillar system and mimicked a 10 um pore size. The device was fabricated using the hot embossing technique, described by Jeon, et al. (see fig below).

Fig. 1



Hot embossing generated the devices. Patterned SU8 photoresist on a silicon wafer (a) served as a mold to create a negative replica in PDMS (b) to permit pouring and curing of epoxy (c) to create the durable epoxy master mold (d). The master mold formed the microfluidic features in the COC plate under load and elevated temperature through hot embossing (e). The resulting embossed COC plate was trimmed (f), drilled for fluid connections (g), then sealed with a thin COC laminate layer to complete the array of finished devices (h)

The photoresist wafer was created using standard photolithography techniques. Because our desired effective pore size is greater than 1 um, we would not need the resolution granted by electron beam lithography. After each use, the device could be sonicated and rinsed with back-flowed PBS to remove any residual scaffold debris and reused thereafter. This retains some manual input but it is minimal. Overall, this method is cheaper than post hoc filtration since only one device is needed, however none of the team members are trained in such fabrication methods.

Conclusion:

After a second look at filtration methods, I think the pillar method would be better, since the client really desires automation, and this filter could be effectively sonicated and rinsed to prevent clogging after repeated use. None of the team are trained to fabricate using the hot embossing method, and we do not have access to the photolithography lab space in ECB, but due to COVID-19, the project became more theoretical, so suggesting this method to our client (who can fabricate such a structure) is reasonable.

2020/04/20-Multiple inputs/outputs

KEVIN KOESSER - Apr 29, 2020, 3:17 PM CDT

Title: Multiple input and output methods

Date: 04/10/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To compile some options for implementing multiple inlets and outlets to the microfluidic chamber.

Content:

In the future, it would be advantageous to have multiple inlets and outlets so that protein solutions are not contaminated and automation is maintained. We would hold to our plan of a positive pressure inflow assisted by negative pressure outflow when polymerization is finished. "Microfluidic manifold" Products exist that merge or split multiple channels. This product could be attached to the "mouth" of the microfluidic device channel so that multiple protein solutions and a PBS solution could flow into the device. One concern I have with this product is the potential for backflow: if the inputs are not all flowing simultaneously, then there is a potential for some protein solution of one inlet flowing through another inlet instead of the mouth of the microfluidic channel. This may be ameliorated if the system were entirely driven by negative pressure on the outlet end. Otherwise there are several methods to introduce resistance to the inlet channels such that backflow is less likely. 1) increasing the inlet tube length and adding a T intersection to the merger, similar to the figure 1 below [1], or 2) adding a Tesla valve to each inlet, as demonstrated (simulations and tests) by Lam and Li (2012) (see fig 2) and crudely seen in this YouTube video.

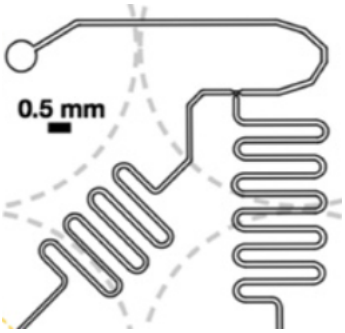


Fig 1

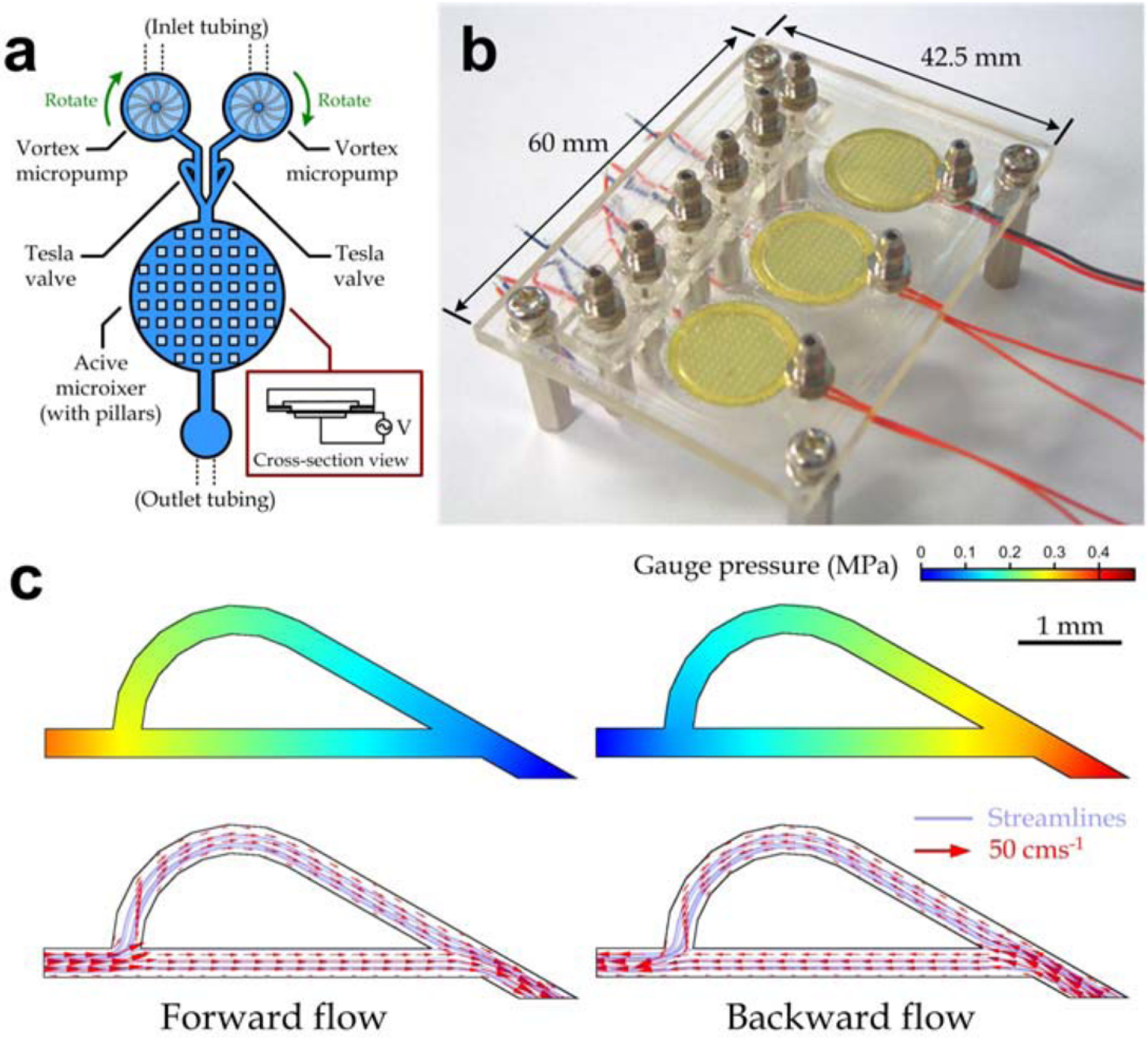


Fig 2

Multiple outputs presents a more challenging problem if protein solutions are to remain pure after polymerization. Splitting the outflowing solution in the correct collection reservoir is not feasible. Additionally, adding multiple outlet channels may permit flow through the wrong exit channel if it is not fully blocked (i.e. flow must only be permitted through one outlet at a time). The multiple input design by Precigene could solve this problem if used in reverse on the outflowing channel (see fig 3). In this design, a microcontroller controls the rotation of a rotary valve with holes connected to each of the reservoirs and negative pressure from the pump draws out protein solution through one channel at a time.

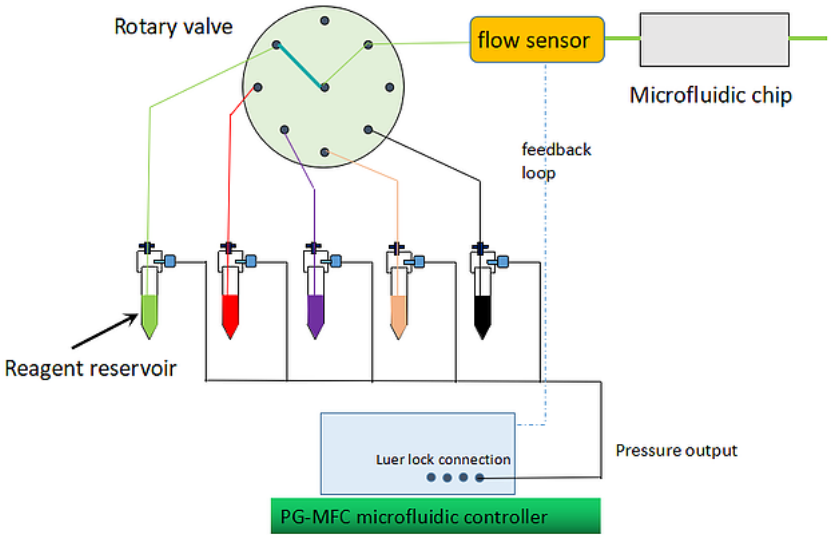


Fig 3: Reversing the direction of flow could allow this mechanism to work as a multiple-outlet selector

Conclusions:

The practicality of manufacturing or purchasing any of the proposed components is not feasible, but it is rather the subject of future work. Due to COVID-19, the theoretical benefit is emphasized over practicality.

I think the commercially available microfluidic manifold would work for an input method, but we would need to test it to determine if backflow would occur or if negative pressure driven system would be necessary. The rotary valve device would be difficult to integrate with the Labview software, however a proficient bioinstrumentation student with prior experiance with microcontrollers could handle it.

2020/04/21-Purity Calculations

KEVIN KOESSER - Apr 29, 2020, 3:37 PM CDT

Title: Purity/concentration of exiting peptide solutions

Date: 04/21/2020

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To get a rough idea of how much protein solutions are diluted after polymerization.

Content:

Initial Conditions:

inlet protein concentration: 1 mg/mL stock - 30 ug minimum dissolved in 200 uL total solution = .15g/L solution

max scaffold size: 2400x2400x50 um

scaffold density: 2 mg collagen/mL scaffold *varies depending on desired ECM structure

===> max scaffold volume * scaffold density = mass of collagen per scaffold = .576 ug

Therefore need at least .576/.15 = 3.84 uL solution per scaffold. if all input protein is consumed. Because about 35-40 uL is used per scaffold, the protein concentration is sufficient to maintain the continuity assumption of the Brownian distribution of technically discrete protein molecules in the solution, so there is enough protein in the solution to properly print a scaffold. Additionally, the protein solution is diluted about 10% per scaffold.

Biosafety Training

KEVIN KOESSER - Oct 09, 2019, 12:50 PM CDT

Title: Biosafety training

Date: 10/09/2019

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To document the completion of the biosafety training.

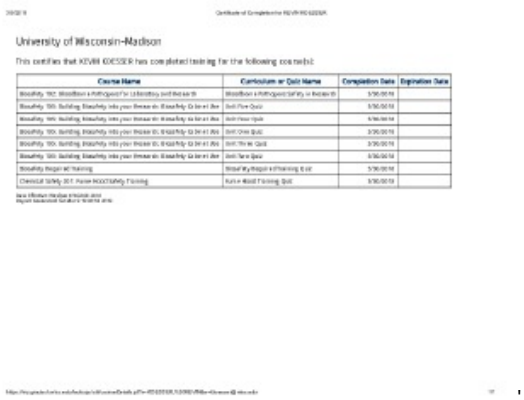
Content:

see attached document

Conclusions/action items:

As of October 9, 2019, I am certified for working at biosafety level 2.

KEVIN KOESSER - Oct 09, 2019, 12:57 PM CDT



University of Wisconsin-Madison

document.pdf(147.9 KB) - download

2019/12/11-Shared Lab Access

KEVIN KOESSER - Dec 11, 2019, 2:20 PM CST

Title: Permission to use the shared BME teaching labs in ECB

Date: December 11, 2019

Content by: Kevin Koesser

Present: Kevin Koesser

Goals: To document that I have access to the BME shared labs.

Content:

Current Access

Request Data	Expires	Actions
Groups: Shared Research Labs: 2005 External Doors: No	5/30/2021	Request Extension

Conclusions/action items:

Lab access lasts until May 2021

Types of Protein Assays

JIACOMO BECKMAN - Feb 26, 2020, 1:43 PM CST

Title: Types of Protein Assays

Date: 1/27/20

Content by: Jiacomo

Present: Jiacomo

Goals: Find a protein assay that may be more effective than spectrophotometry

Content: Below is a set of notes taken during my BME 545 lecture on protein assays.

Conclusions/action items: If given the chance, a histological stain may prove to be more likely to use.

JIACOMO BECKMAN - Feb 26, 2020, 1:51 PM CST

RNA - In situ hybridization

- visualize gene expression
- design a probe that will sequence w/RNA of interest
    - probe has tag that colors the RNA
- VERY specific
- can detect multiple genes at once
- spatial information
- cons
    - depends on good probes
    - finicky

Protein - Immunofluorescence, Immunohistochemistry, ICC

- in-place antibody detection of protein
    - add primary antibody to protein - anti-fibronectin
    - add secondary antibody to amplify the signal
    - add colorimetric label or fluorescent factor
- qualitative and semi-quantitative data

Protein Western Blot

- essentially IHC on a membrane
- separate protein by MW
- use histochem technique to detect proteins
    - size of band corresponds to how much of protein is present

Protein - ELISA

- takes immunohistochem approach and makes it quantitative
- darkness of solution is proportionate to amount of protein
- primary and secondary antibodies

Histological Stains

- nonspecific charged dyes - attracts opposite charge
    - very trial and error
- very qualitative data
- resort to this for 1st pass differentiation evaluation

Collagen

JIACOMO BECKMAN - Feb 26, 2020, 1:57 PM CST

Title: Collagen

Date: 1/27/20

Content by: Jiacomo

Present: Jiacomo

Goals: Understand further one of the ECM components used in Dr. Campagnola's lab

Content:

- versatile building block
    - most abundant fibrous protein in mammals
- provides mechanical integrity, scaffolding, organization, and strength
    - is the main load carrying component of soft tissue
- COL1 is the most common that is used (this is probably what Dr. Campagnola uses)
- triple helix
    - variation in length of non-helical parts, length of helical parts, and number of carbohydrate attachments to helix

Conclusions/action items: This may be one of the cheaper proteins that our client uses. If there is a higher emphasis in recollecting other protein solutions, collagen may not be the most prioritized.

Elveflow Research

JIACOMO BECKMAN - Mar 04, 2020, 6:45 PM CST

Title: Elveflow Research

Date: 3/4/20

Content by: Jiacomo

Present: Jiacomo

Goals: Look into Elveflow pumps since we are having issues with the piezoelectric pump

Content:

MULTI CHANNEL PRESSURE & VACUUM CONTROLLER

- Pressure stability 0.005 % FS
- Response time 9 ms
- Pressure resolution 0.006 % FS
- Settling time down to 35 ms
- flow regulation down to 7.5 nL/min
- compatible with LabView
- 5 pressure ranges are available

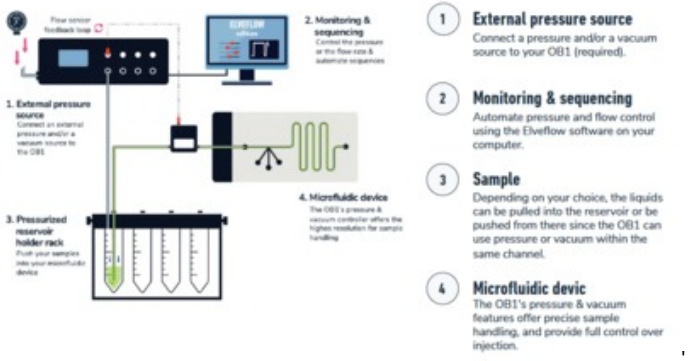
SINGLE CHANNEL AUTONOMOUS PUMP

- The AF1 is a high performance autonomous pressure and flow controller
- It comes in three different ranges and embeds pressure and vacuum sources
- It is compatible with Elveflow software

Really these two pumps seem to be the only ones that would be most relevant to the project. The other pumps involve an absurd number of outlets that we would not need whatsoever.

Conclusions/action items: Converse with the team about this and email Elveflow about pricing. Hopefully they are not insanely expensive

JIACOMO BECKMAN - Mar 04, 2020, 6:33 PM CST



How_to_Use_OB1.tiff(389.2 KB) - download

Green Shop Pass

JIACOMO BECKMAN - Feb 26, 2020, 1:36 PM CST

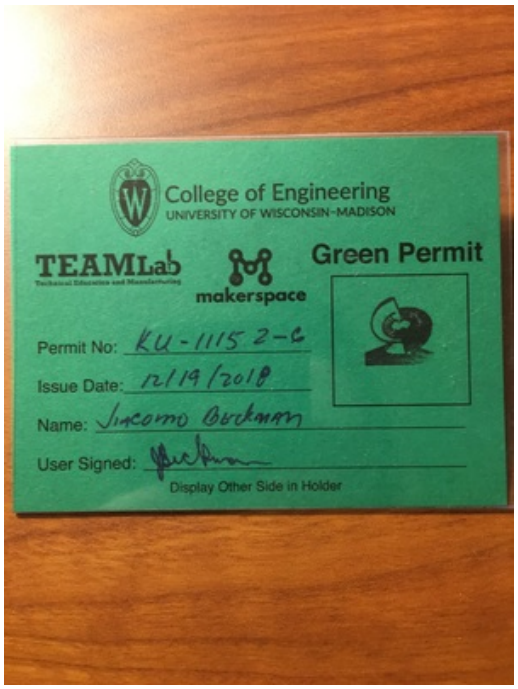


image1.jpeg(2.9 MB) - download

Lab Safety Verification

JIACOMO BECKMAN - Feb 26, 2020, 1:36 PM CST

University of Wisconsin-Madison

This certifies that JIACOMO BECKMAN has completed training for the following course(s):

Course Name	Curriculum or Quiz Name	Completion Date	Expiration Date
Biosafety Required Training	Biosafety Required Training Quiz	3/24/2019	

Data Effective: Sun Mar 24 16:03:05 2019
Report Generated: Sun Mar 24 19:24:31 2019

Lab_Safety_Training.jpg(63.1 KB) - download

2014/11/03-Entry guidelines

John Puccinelli - Sep 05, 2016, 1:18 PM CDT

Use this as a guide for every entry

- Every text entry of your notebook should have the **bold titles** below.
- Every page/entry should be **named starting with the date** of the entry's first creation/activity, subsequent material from future dates can be added later.

You can create a copy of the blank template by first opening the desired folder, clicking on "New", selecting "Copy Existing Page...", and then select "2014/11/03-Template")

Title: Descriptive title (i.e. Client Meeting)

Date: 9/5/2016

Content by: The one person who wrote the content

Present: Names of those present if more than just you (not necessary for individual work)

Goals: Establish clear goals for all text entries (meetings, individual work, etc.).

Content:

Contains clear and organized notes (also includes any references used)

Conclusions/action items:

Recap only the most significant findings and/or action items resulting from the entry.

2014/11/03-Template

John Puccinelli - Nov 03, 2014, 3:20 PM CST

Title:

Date:

Content by:

Present:

Goals:

Content:

Conclusions/action items: