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Washington University in St. Louis

SCHOOL OF ENGINEERING & APPLIED SCIENCE

Executive Summary

This tissue loading project consists of designing an apparatus that has two main functions. The first, and most obvious of these, being creating a device that will deform a band or disk of tissue with cells pre-seeded on them uniaxial while recording the stress and strain, specifically strain rate, that is applied to the tissue. While the deformation aspect of this device will be key, an equally important part of this project will be designing a part of our tissue loader that wither mimics the environmental chamber that the lab already uses or is compatible with the environmental chamber that already uses. This is key when dealing with cells as the slightest perturbation in environment can lead to cell death and therefore inconclusive research results.

MEMS 411: Senior Design Project

Tissue Stretcher

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1 INTRODUCTION AND BACKGROUND INFORMATION

1.1 INITIAL PROJECT DESCRIPTION

The idea for this senior design product is a tissue manipulator to be used while examining different tissues or gels with cells seeded on them. Being able to manipulate the environment the cells are attached to is of great interest to tissue researchers and mechanobiologists. One of the main attributes of the apparatus that is to be designed is that it must be compatible with existing microscopes that are in use for this type of research. Commonly confocal microscopes are used which employ lasers instead of the typical lenses to contrast images of tissues and cells on scales commonly in the micrometer range. Therefore, it is imperative that whatever device that is created is compatible with these sophisticated imaging technologies.

1.2 EXISTING PRODUCTS

This system by TA instrument most closely matches the scale of the device that is trying to be created. Using calibrated motors the apparatus places the material into various states of stress and strain. The loading state can be specifically specified and will output useful data on how the tissue is loaded. This could be especially useful in research as it would allow the image taken to be associated with a whole host of information about the state the sample is in, clearly allowing to see how cells in these environments are affected.



Figure 1. TA Instruments Tissue Stretcher

1.3 RELEVANT PATENTS

Patent #: US6322563B1

This patent is for a surgical tissue pinning device. It was developed as a way to pin down tissues and membranes during surgery as a faster and less intensive way than the traditional method of using sutures. This method of fixing tissue could prove useful when trying to attach the tissue our product is trying to load to the surface that it is mounted on and to the loading apparatus itself. This is especially true when a large amount of samples will have to be quickly mounted and then imaged.

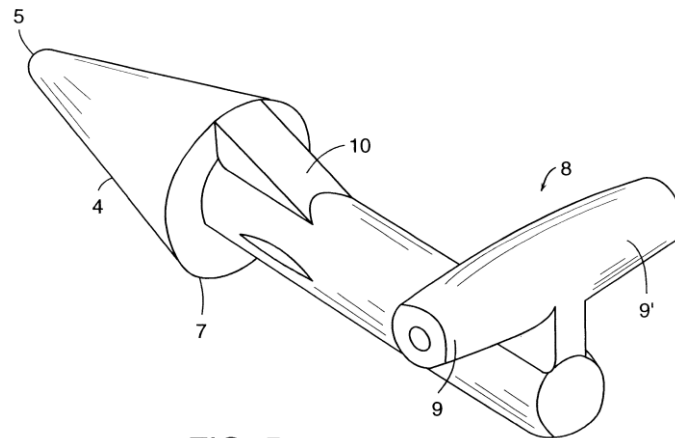


FIG. 5

Figure 2. Patent For Tissue Pin Concept

1.4 CODES & STANDARDS

As the project was designed to be used in a laboratory setting, many of the usual codes and standards that apply to a design that is to be manufactured for the general public do not apply. However, there are still guidelines and requirements that must be followed when dealing with a device that interacts with biological tissue.

Because the design incorporated an environmental chamber into the design, certain standards that apply to cell ovens apply to the design. For example, one code that was followed in the design making process was ASTM E95-68 (4.3). This standard states that any "oven" (or in this case an environmental chamber with a testing fence) that has air circulating in its chamber, as the environmental chamber in this design does, must keep the temperature of the chamber within 1° Celsius of the target temperature.

Another standard adhered to when designing this tissue stretcher was ASTM E95-68 (4.5), which involves preventing the cross-contamination of tissue samples in order to promote best biological practices. When designing a tissue stretcher prototype, efforts must be made to make sure that different samples being tested do not come in contact.

1.5 PROJECT SCOPE

1. The goal of this product is to create a loading machine that will not only deform the material while being able to fit in existing imaging equipment, but also to do it efficiently and quickly. The machine should also be able to keep track of the loading state so as to be useful to researchers who are trying to connect certain mechanical environments with certain cellular deformations and behaviors. The system must be configured so to create a loading situation that can be measured but also be held constant for long periods of time while the imaging is taking place. Also, the possibility can be explored to make the system portable and compatible with cell culture environments so as to make the seeding of cells onto the tissue being tested easier, although this is not imperative to the design.

2. The customer for this product would be tissue researchers. Specifically, for this product Professor Genin at Washington University in St. Louis. The device will help them characterize the cells overtime.
3. The value to the customer is that instead of viewing the cells in different static states of in different discrete loading conditions the researcher can know track the change in the cells environment and their reaction to these changes in environment in real time. This will allow them to better characterize and study how cells react to their environment.
4. The goals of the project are to create a tissue loading prototype that will exert a predetermined or measurable amount of stress or strain upon the material being tested while keeping the material intact. This device must be compatible with the imaging technologies and techniques that the researcher/customer currently uses. As this project is limited to one semester the goal is to deform the material axially in one to two directions. Any polar deformations would be unreasonable to complete in the time allotted.
5. The scope of this product is creating a machine that will
 - a. Deform the material in a measureable way; either having a distinct stress or strain value for the material.
 - b. Devise a way to connect the material with the load device.
 - c. Allow the sample to be imaged while it is being loaded.
 - d. Have two-dimensional deformation.
6. Things that are out of scope for this project are:
 - a. Having polar directions of deformation
 - b. Applying harmonic loads
 - c. Three-Dimensional deformation.
 - d. Creating Extensive software for the device (possible and software at all).
7. Critical Areas of Success for our project are
 - a. The device must consistently apply the determined/measured value of stress and strain. In other words, the stress and strain that the user specifies must actually be the loading state of the material.
 - b. The device does not interfere with or damage the microscope that is being used.
 - c. The device should not significantly alter the tissue that it is testing in the mounting process, i.e. it cannot degrade the material in ways that are not specified.
 - d. The device should remain connected to the loading apparatus throughout the imaging process.
 - e. The product should be reasonably durable. It can't fail after just a few uses. It should be in line with standard laboratory equipment.
8. Some assumptions that are being made in order to plan this project are:
 - a. The device itself will not be too large or heavy as it has to fit onto the microscope stage. However, the device will still be able to generate a range of different stresses and strains that will be useful for the researcher. If this assumption proves to be incorrect it will impact the effectiveness of the device as it may not adequately deform the tissue to provide useful results in the lab.
 - b. Materials that will not affect the microscope will be used. We are currently assuming that materials that will lead to effective construction of the device exist that will also not affect the images generated will be used. Such materials may not exist or materials that will distort the images somewhat might be used. If this assumption is false than it will also affect the viability of the research conclusions drawn from using this equipment.
 - c. In all likelihood motors will be used to deform the material. Motors provide control on how to deform the material so they are likely to be used somehow in this project. If they are not used than the budget of this project could be off if a less expensive or more expensive method is found to be more effective.
9. Constraints on this project are:

- a. Budget. Depending on the budget for the project the type of motor or force application device will be affected. The more expensive the device used the more control we will have over the device and how much force it will apply to the material. As such budget could affect how fine our stress/strain values that are applied to the material can be.
 - b. Programing abilities: The group members of this project have very little programing knowledge. As such the device that is made will probably not have extensive software to go along with it. This affects the project as the user interface to control the device maybe archaic or underwhelming affecting the options that the customer will have with this project.
 - c. Customer constraints. The device may be limited by what can be used in a laboratory setting or how the researcher wants the device to work.
10. Some Key deliverables for this product are:
- a. A working prototype that will perform in the way outlined in the scope of the project.
 - b. A device that can be replicated in a reasonable manner.

1.6 PROJECT PLANNING

In order to effectively use the time provided to make a working prototype and to make sure that the design created was effective, a Gantt Chart was used to organize the time used during the design process. Also, to gain more information about the project, an Interview was conducted with Dr. Genin, the principal investigator that the tissue stretcher was designed for. This allowed for there to be an adequate base of knowledge about the desired prototype so that an effective prototype could be created.

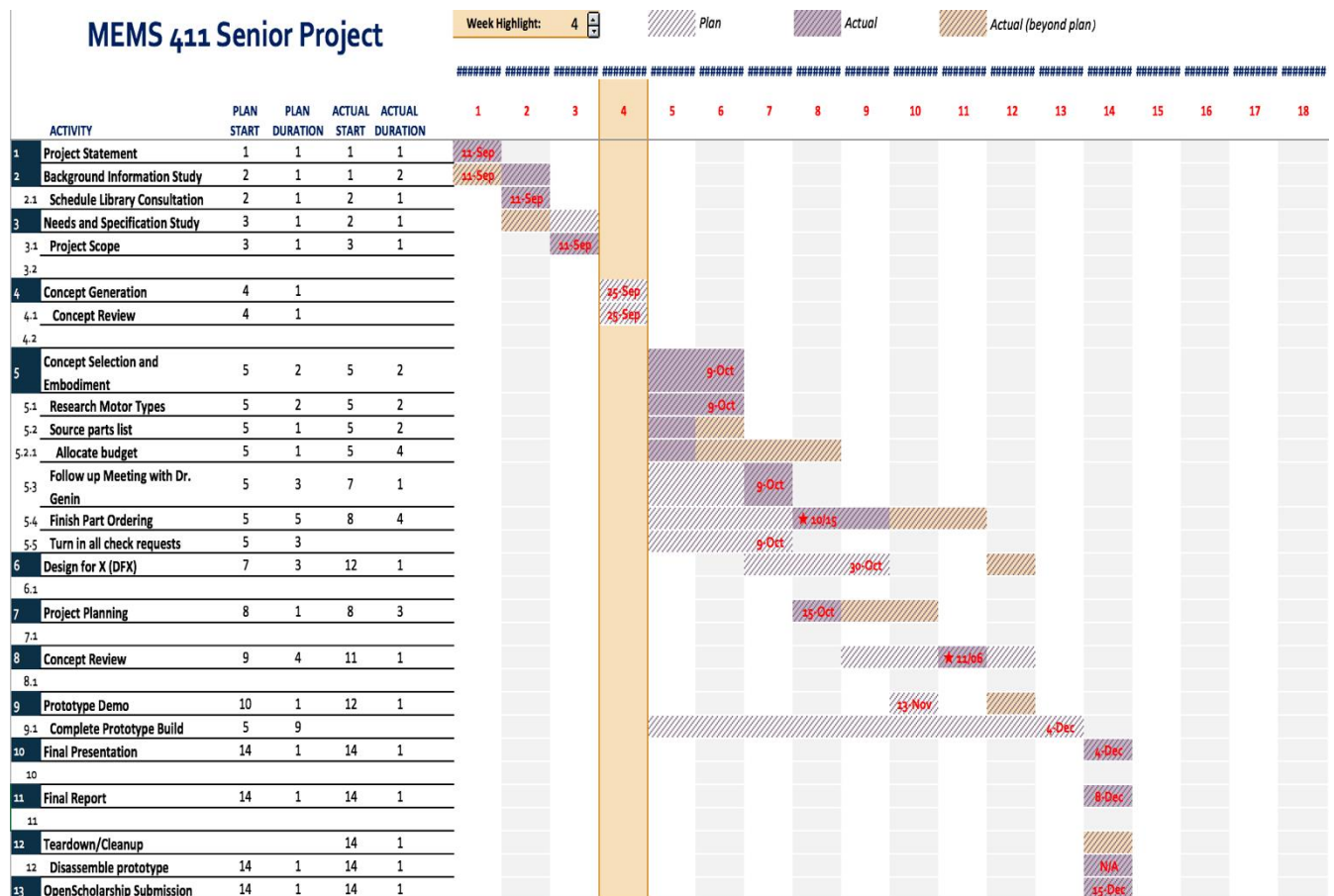


Figure 3. Gantt Chart

1.7 REALISTIC CONSTRAINTS

When designing the concepts discussed later in this report, as well as when constructing the final prototype several realistic constraints had to be followed. These constraints exist outside of specific codes and standards set out by any specific scientific body or engineering body. Instead, these constraints were applied due to budget, time, ability, as well as specifications requested by the customer or needed to help ensure smooth operation of the prototype.

1.7.1 Functional

A main functional constraint that was applied to the design of the prototype was the size of the tissue samples that were to be stretched. In Dr. Genin's lab the tissue that is manufactured is made using standard procedures and specific equipment, and have a diameter of only 5mm. Any stretching device that would be made had to keep the small scale of the tissue in mind. Additionally, the device that was created had to fit in the current environmental chamber and on the confocal microscope.

1.7.2 Safety

The main safety concern with the device is overheating due to the power source. If the power source used is incorrect it could lead to burns if hot equipment is touched as well as damage the Arduino and motor.

1.7.3 Quality

Because the environmental chamber was to be airtight so that it could maintain the temperature and CO₂ parameters that were set in the environmental chamber, it was crucial that all machining processes that were used to manufacture the device were done so at fine tolerances. Because of this, all manufacturing was done with CNC machines so that feed rate, RPM, and finishing cuts could be controlled in a consistent manner for all parts.

1.7.4 Manufacturing

Because this prototype was manufactured for a lab setting and was to be used exclusively for research purposes, usual constraints that are used for manufacturing, especially those that pertain to mass production, were not as important in the design process. If in the future there was a need to manufacture the prototype in bulk, the constraints that would most need to be minded would be with the stretcher arm and designing it so that injection molding could be used for production. Constraints would include part geometry and draft angle.

1.7.5 Timing

As the start of each test was user controlled, there was no real need to take into account the timing of any part in the device. Neither the environmental chamber used nor the confocal microscope had any timing constraints as well.

1.7.6 Economic

Perhaps the biggest constraint that this project has was economic through the budget provided to create the prototype. Because of the budget provided, an adequate force transducer was not able to be purchased. Thus, the old force transducer that was used in Dr. Genin's lab had to be used which limited

the placement of where tissue was to be stretched and the orientation of the fence in relation to the force transducer and the stretcher arm.

1.7.7 Ergonomic

There were no real ergonomic constraints with the project as there is very little physical interaction between the device and the user.

1.7.8 Ecological

The only ecological constraints that exists with this device lies with possible contamination of the surfaces by cells or other chemicals. However, if this were to occur it is the lab members responsibility to properly clean the device and dispose of any waster. To aid them in this however, the device should be able to be disassembles to be able to be cleaned.

1.7.9 Aesthetic

No aesthetic constraints exist as this device will be used in a laboratory setting and will not be marketed to the general public. The primary concern with the device is function not looks.

1.7.10 Life Cycle

While they are no specific constraints about how many cycles or years this device has to last, the device should be able to last a reasonable amount of time and should serve the lab well in its function without needing constant repair or redesign.

1.7.11 Legal

As this is a novel device requested by a research lab no Legal constraints exist with this type of device.

1.8 REVISED PROJECT DESCRIPTION

This tissue loading project consists of designing an apparatus that has two main functions. The first, and most obvious of these, being creating a device that will deform a band or disk of tissue with cells pre-seeded on them uniaxially while recording the stress and strain, specifically strain rate, that is applied to the tissue. While the deformation aspect of this device will be key, an equally important part of this project will be designing a part of our tissue loader that mimics the environmental chamber that the lab already uses or is compatible with the environmental chamber that is already in use. This is key when dealing with cells as the slightest disturbance in environment can lead to cell death and therefore inconclusive research results.

2 CUSTOMER NEEDS & PRODUCT SPECIFICATIONS

2.1 CUSTOMER INTERVIEWS

Table 1: Customer Interview Response

Customer Data: Dr. Guy Genin Address: Green Hall Room 3120 D, St. Louis, MO 63130 Date: 09/13/17			
Question	Customer Statement	Interpreted Need	Importance
Likes about current device	How the Machine can control the force on the material or the displacement to keep the force constant.	TS can vary stress and strain on the material.	5
	The current TS can send data straight to the Computer	TS is efficient at logging data for use in Analysis.	5
Dislikes About Current Device	Cell Media has to be exposed to the atmosphere to allow access to the device	TS can operate while keeping the cells in a controlled environment	4
	Data logging and control system software is outdated.	TS operates using the most recent usable software	3
	The objective can get out of focus while the machine is moving.	TS can be used in conjunction so that the microscope stays in focus	3
	Current device does not allow the use of the 40x oil immersion objective	TS can be used with the 40x oil immersion objective	2
Specific Requirements	The Tissues being tested are stretched up to 10%/s strain	TS applies stresses for up to 10% deformation of material.	4
	Add drugs and other solutions to tissue while it is being stretched	Has the ability to add other materials into the testing environment.	1
	The device must be able to test specimens 5mm in diameter		5

	The TS allows the cell environment to stay close to 37 degrees Celsius.	TS must be able to stretch specimens at least 5mm long TS can operate while keeping the cells in a controlled environment	4
Standards Required	TS keeps temperature of environment within 1 degree Celsius of set temperature. ASTM E95-68 (4.3)	TS can operate while keeping the cells in a controlled environment	4
	TS keeps the tissue specimens adequately separated to avoid cross-contamination of samples ASTM E95-68 (4.5)	TS keeps the specimens far enough apart so they are not touching.	3

2.2 INTERPRETED CUSTOMER NEEDS

Table 2: Customer Needs

Need Number	Need	Importance
1	TS can vary Stress and Strain on the material being tested.	5
2	TS is efficient at logging data for use in Analysis.	5
3	TS can operate while keeping the cells in a controlled environment	4
4	TS operates using the most recent usable software	3
5	TS can be used in conjunction so that the microscope stays in focus	3
6	TS can be used with the 40x oil immersion objective	2
7	TS applies stresses for up to 10% strain per second deformation of material.	4
8	Has the ability to add other materials into the testing environment.	1

9	TS must be able to stretch specimens at least 5mm long	5
10	TS keeps the specimens far enough apart so they are not touching.	3

2.3 TARGET SPECIFICATIONS

Table 3: Target Product Specifications

Metric Number	Associated Needs	Metric	Units	Acceptable	Ideal
1	7,9,10	Length	mm	<10	>5
2	2,4	Operating System	Year	>2010	>2015
3	3	pH	Unitless	6.0-8.0	7.4
4	3	Temperature	Celsius	35-38	37
5	5,6	Specimen Distance From Objective	mm	< 25	<.0254
6	7,1	Strain Rate	% per second	<10	<15
7	8	Volume of Containment Well	mL	>20	>30

3 CONCEPT GENERATION

3.1 FUNCTIONAL DECOMPOSITION

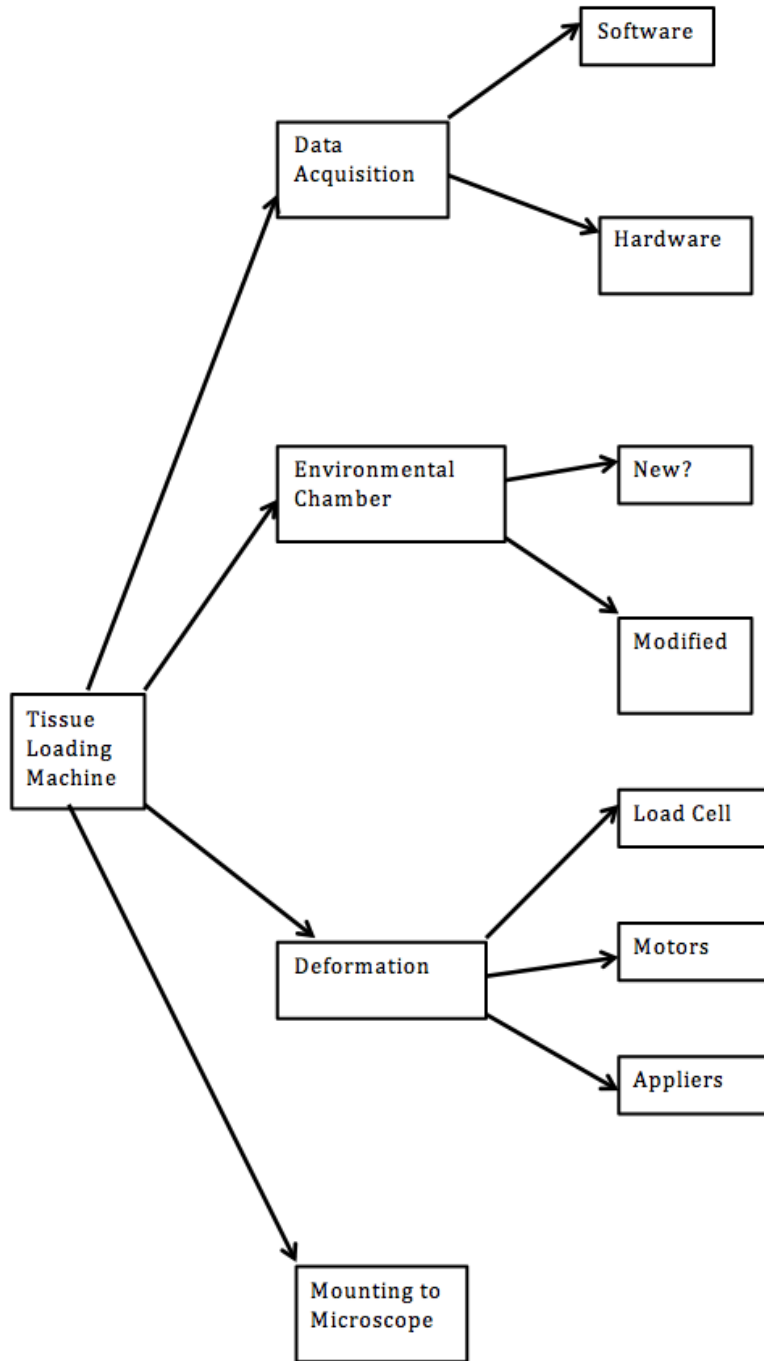


Figure 4 Function Tree Tissue Stretcher

3.2 MORPHOLOGICAL CHART

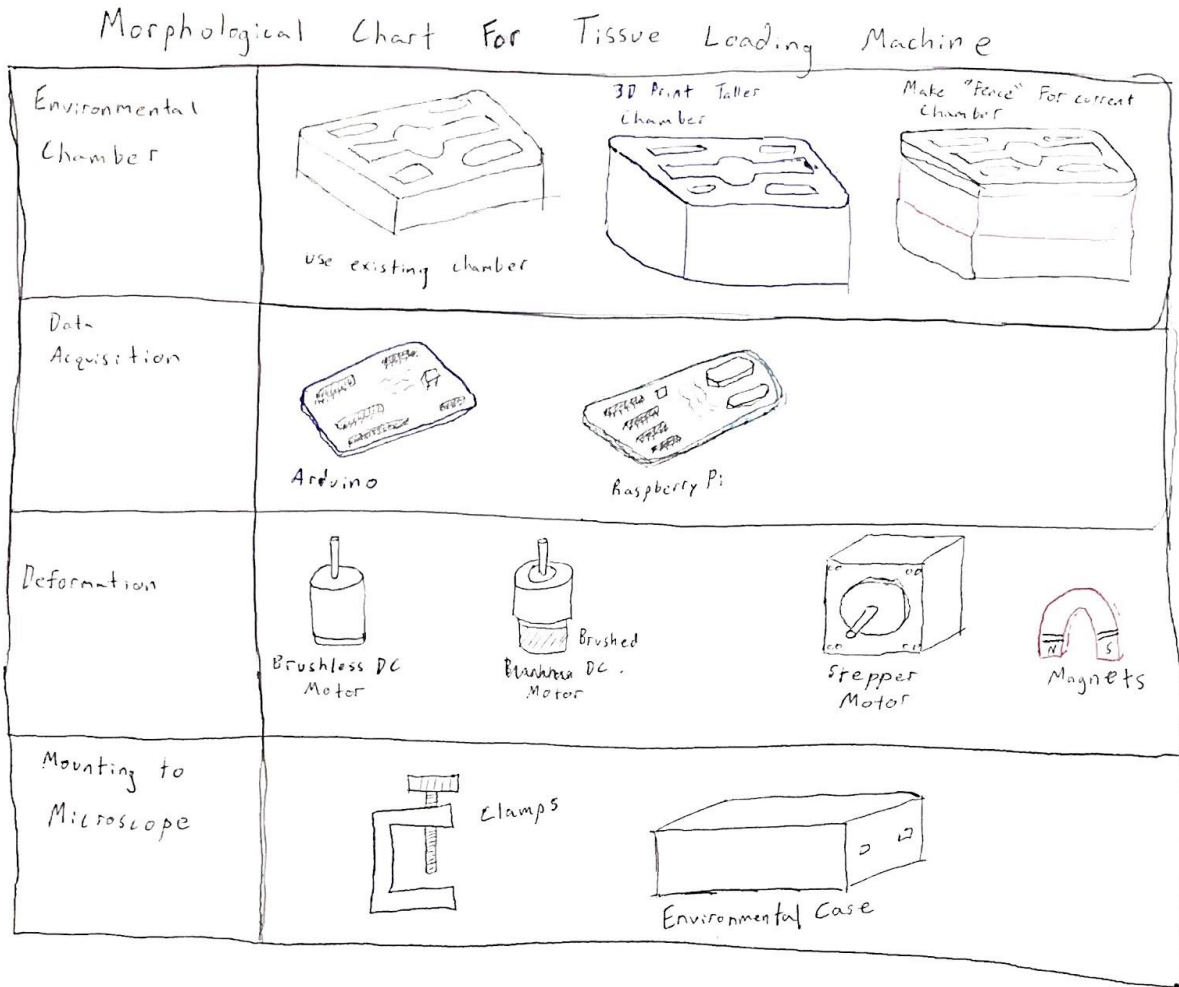


Figure 5 Morphological Chart

3.3 CONCEPT #1 – “NEW ENVIORNMENTAL LID ”

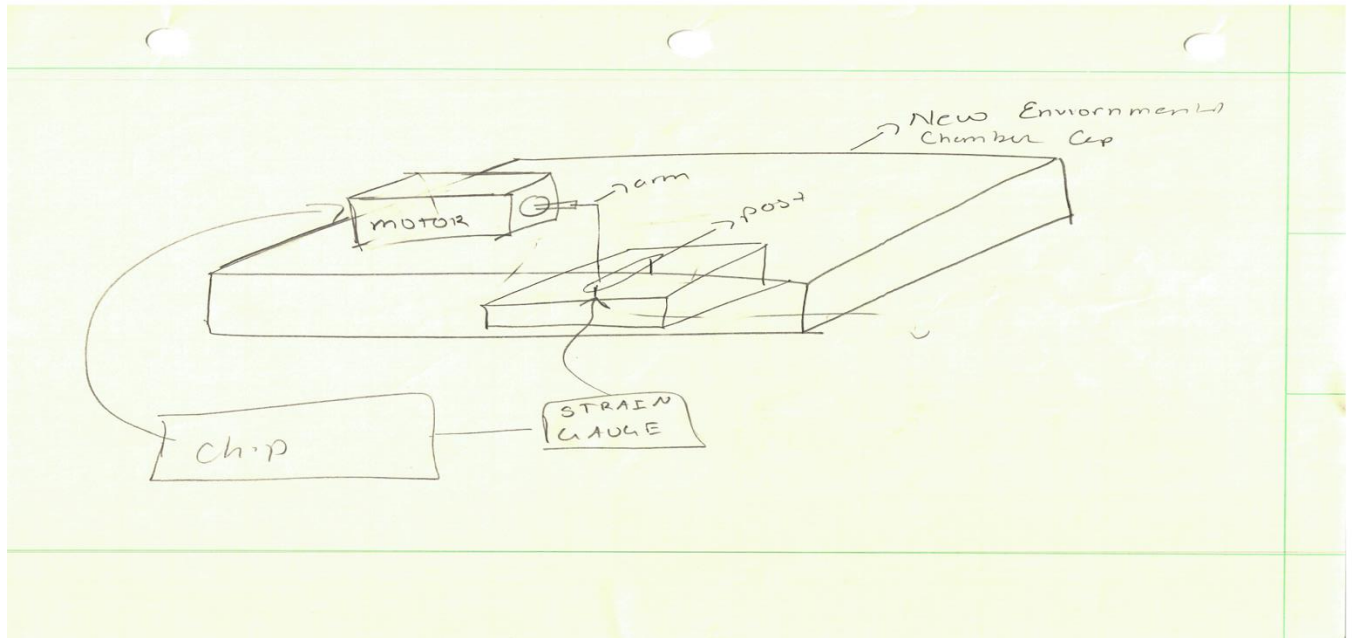


Figure 6 Environmental Lid Concept

This concept employs the construction of a new environmental chamber lid that will fit on top of the existing bottom section of the environmental chamber. Mounted on top of the lid will be a stepper motor connected to an arduino and a force transducer that can be used to apply certain strains on the tissue below. The lid also will have gate of some sorts to allow the arm connected to the motor to move the tissue back and forth inside of the chamber.

Solutions(from Morph Chart)

1. Uses new environmental chamber lid
2. Uses an Arduino
3. Uses a Stepper Motor
4. Is part of the environmental case.

3.4 CONCEPT #2 – “ENVIRONMENTAL FENCE”

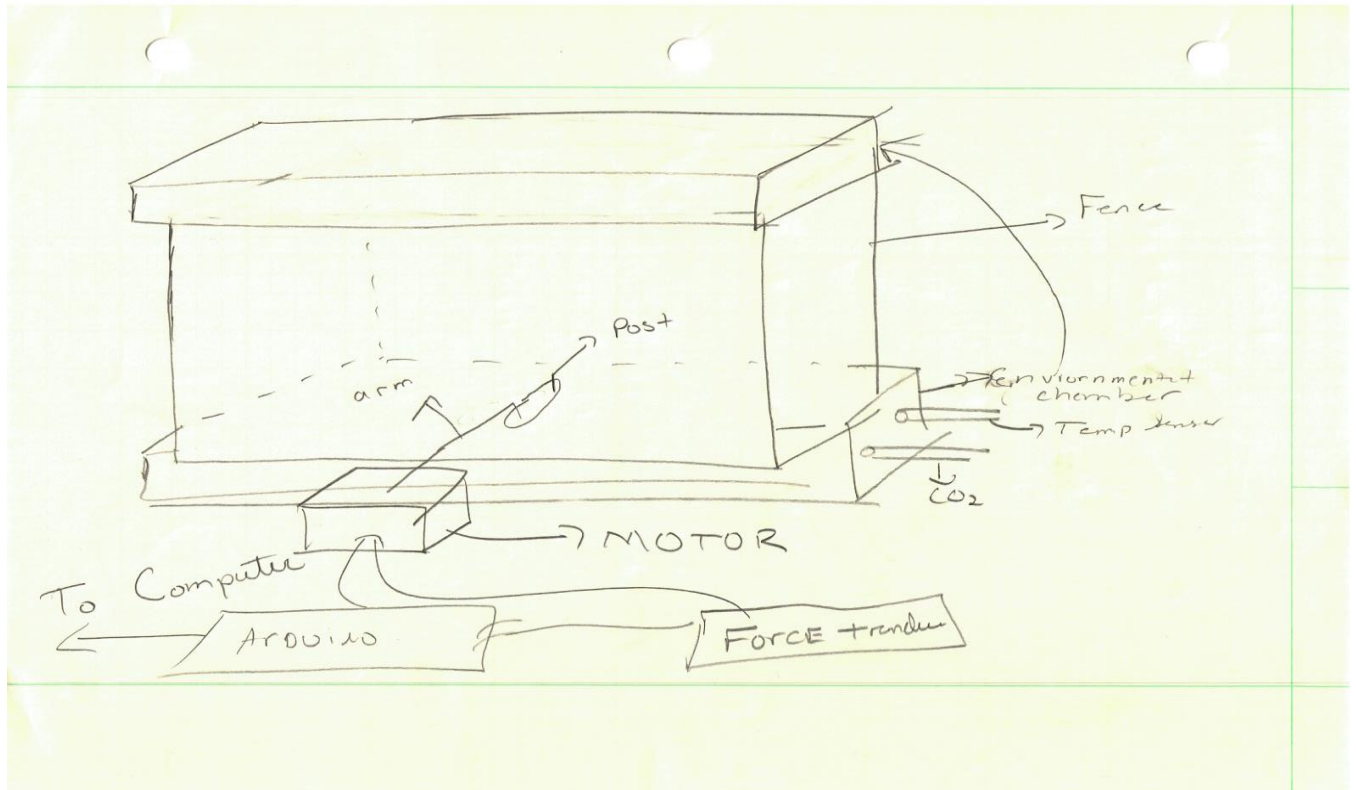


Figure 7 Environmental Fence Concept

This concept would utilize a fence that would sit in between the existing environmental chamber's top and bottom sections. This would serve to help preserve the hermetic seal without having to recalibrate and engineer the heaters and thermometers inside the environmental chamber. A stepper motor would be used to deform the tissue and its force would be controlled and recorded by an arduino and a force transducer.

Solutions:

1. Fence
2. Arduino
3. Stepper Motor
4. Clamps

3.5 CONCEPT #3 – “EXTENSOMETER”

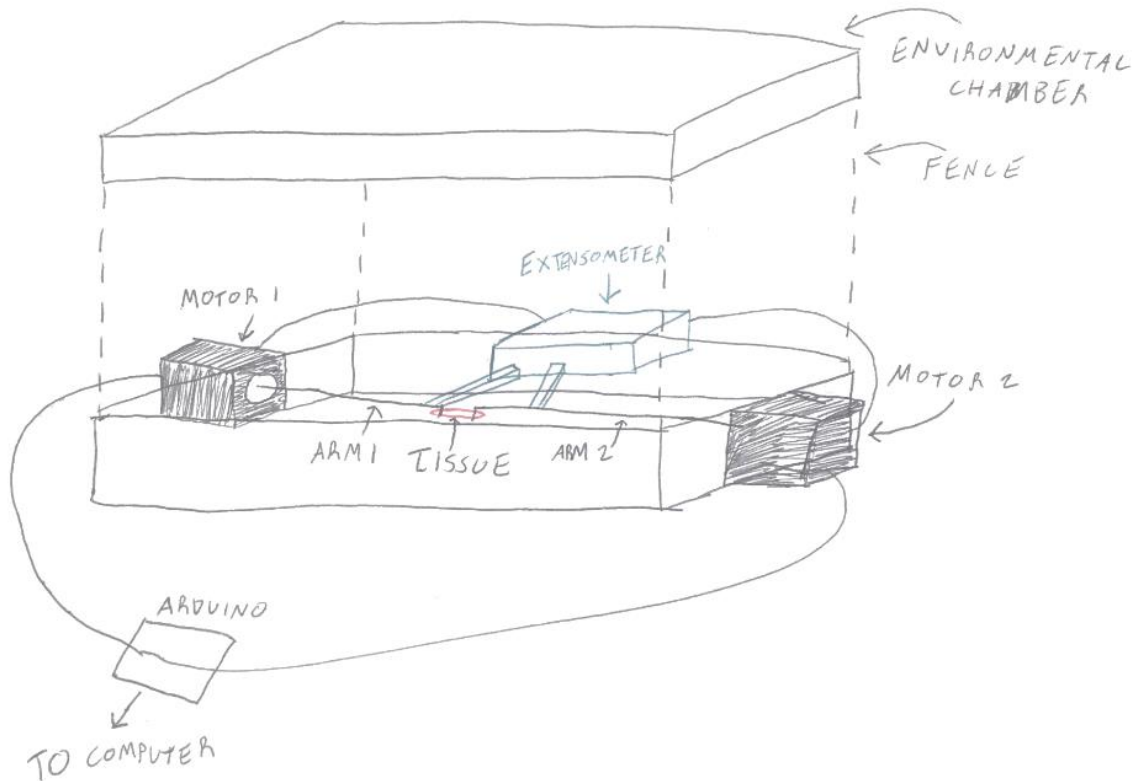


Figure 8 Extensometer Concept

Description: Just like the Environmental fence design, this concept would utilize a fence that would sit in between the existing environmental chamber’s top and bottom sections. This would serve to help preserve the hermetic seal without having to recalibrate and engineer the heaters and thermometers inside the environmental chamber. 2 stepper motors would be used to stretch the tissue and the extensometer would measure the distance that the two arms move apart. The force would be controlled and recorded by an arduino.

Solutions:

1. Fence
2. Arduino
3. Stepper Motors
4. Extensometer

3.6 CONCEPT #4 – “SERVO MOTOR”

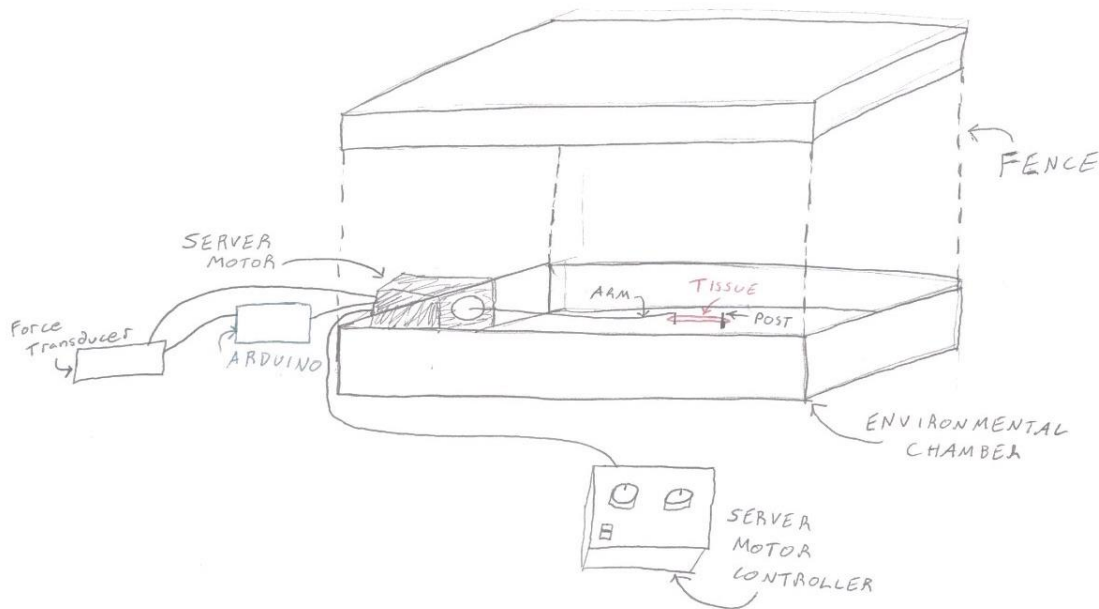


Figure 9 Servo Motor Concept

This concept would utilize a fence that would sit in between the existing environmental chamber’s top and bottom sections. This would serve to help preserve the hermetic seal without having to recalibrate and engineer the heaters and thermometers inside the environmental chamber. A servo motor would be used to deform the tissue and its force would be controlled and recorded by an Arduino and a force transducer. The use of a servo motor would enhance the ability to manipulate the tissue in a very specific manner. It might add unwanted complexity, though.

Solutions:

1. Fence
2. Arduino
3. Servo Motor

3.7 CONCEPT #5 – “HAND CRANK”

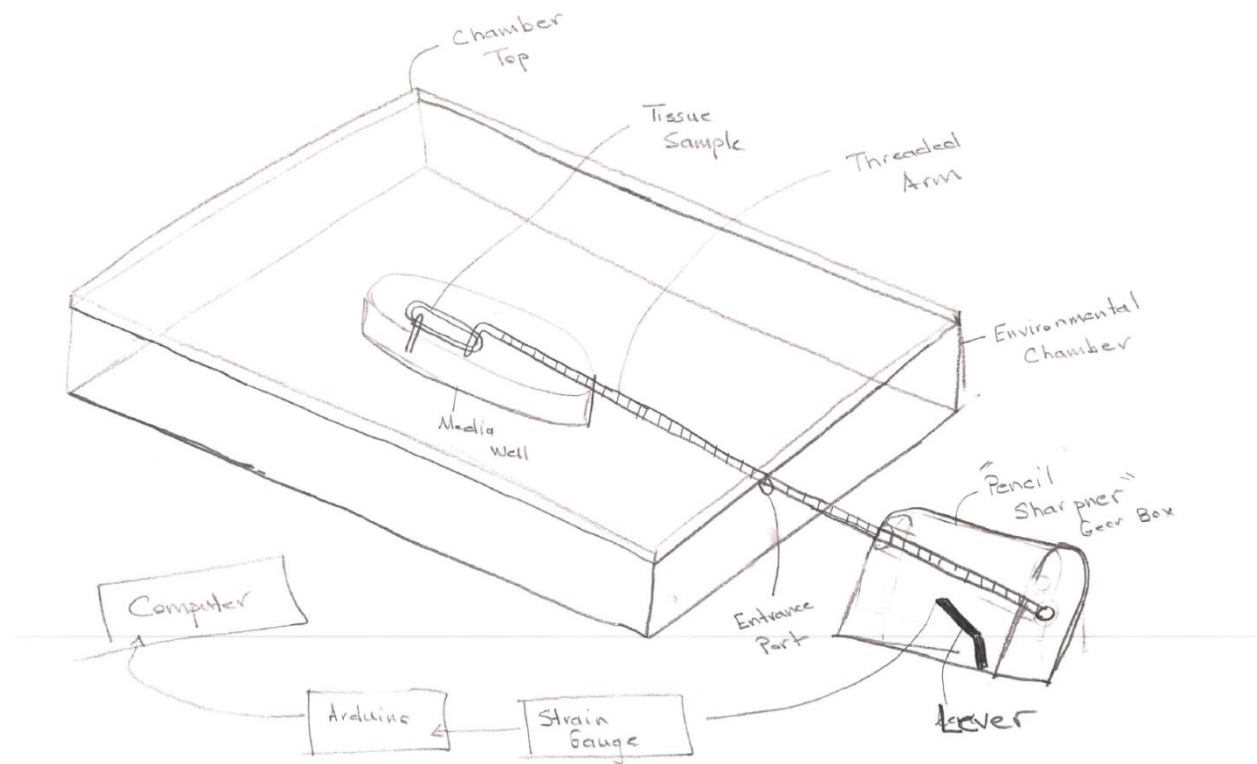


Figure 10 Hand Crank

Description: This design would utilize a port cut into the side of the preexisting environmental chamber to utilize a threaded stretcher arm to apply strain to the biological tissue in question. Movement of the stretcher arm would be controlled by a hand crank connected to the pencil sharpener like box where the lever would turn a screw that would in turn the arm. An strain gauge would be be attached to the stretcher arm to to measure mechanical properties of the tissue that would be sent to a computer via arduino.

Solutions:

1. Arduino
2. Use existing chamber
3. Hand crank
4. No clamp

3.8 CONCEPT #6 – “MAGNETS”

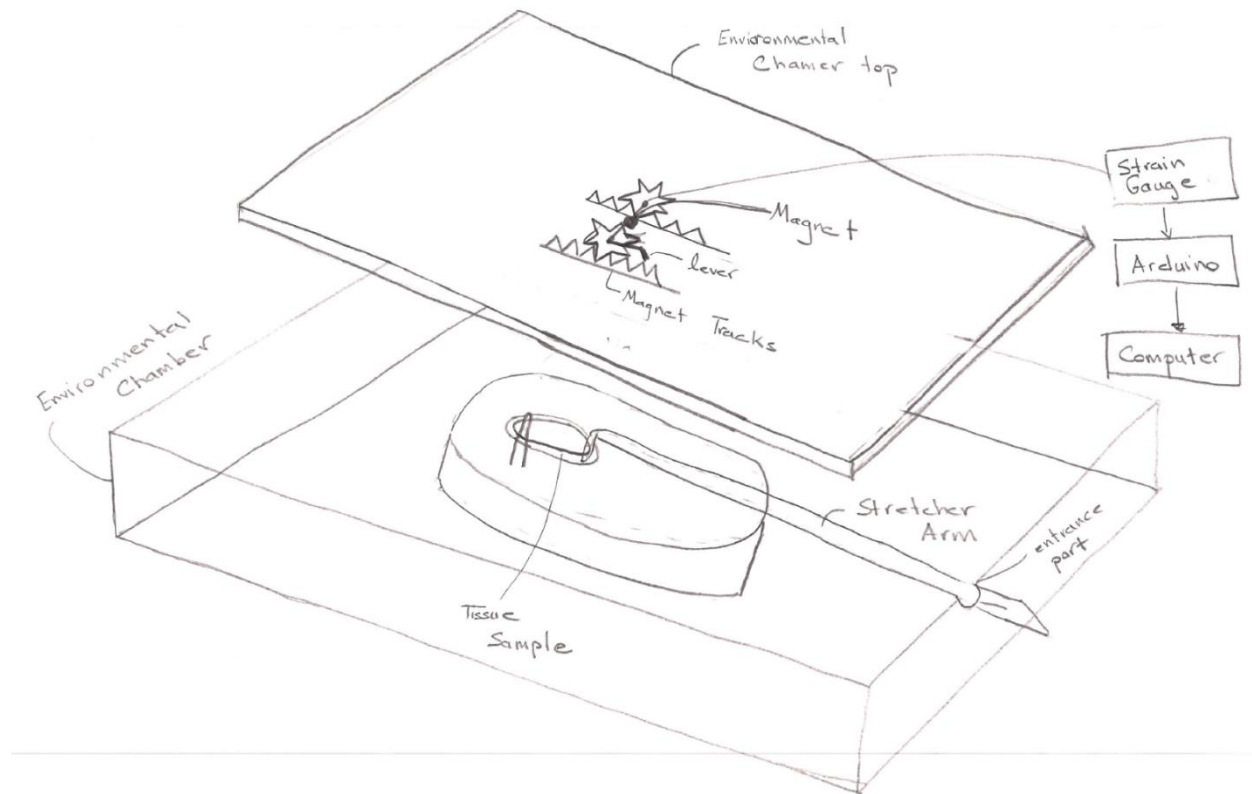


Figure 11 Magnetic Concept

This concept would use magnets that are placed on top of the environmental chamber using a rail system to force the stretcher arm to move and thus stretch the tissue sample. This design is unique that the stretch arm could be encapsulated entirely by the current environmental chamber if it was controlled by magnets on top (however, the concept is shown with an entrance port as another configuration option). A strain gauge could be connected to the magnet or its rail system to measure strain and thus force on the tissue. This could be sent to computer via arduino.

Solutions:

1. Current environmental chamber
2. Arduino
3. Magnets
4. No clamp

4 CONCEPT SELECTION

4.1 CONCEPT SCORING MATRIX

To determine what concept was the best for the needs and applications desired by the customer, a concept selection matrix was used to rank the six concepts against each other. Before that could be completed however, the weight of each need needed to be determined. This was completed using an analytical hierarchy process show below.

Table 4:Analytical Hierarchy Chart

	Mechanical Safety	Cost of components	Strain Rate Variability	Ease of Logging data	Compaibility with Enviornmental Chamber	Compatibility with Lab Tissue	Age of Software	Addition of other Materials	Compatibility with Microcope Objective	Avalibility of Parts	Row Total	Weight Value	Weight (%)
Mechanical safety	100%	20%	14%	20%	11%	11%	33%	33%	25%	13%	281%	1%	1%
Cost of components	500%	100%	33%	50%	20%	20%	50%	200%	100%	33%	1107%	5%	5%
Strain Rate Variability	700%	300%	100%	25%	50%	50%	500%	700%	200%	300%	2925%	14%	14%
Ease of Logging data	500%	200%	400%	100%	20%	20%	300%	500%	500%	200%	2740%	13%	13%
Compaibility with Enviornmental	900%	500%	200%	500%	100%	100%	500%	800%	400%	200%	4200%	20%	20%
Compatibility with Lab Tissue	900%	500%	200%	500%	100%	100%	200%	500%	100%	300%	3400%	16%	16%
Age of Software	300%	200%	20%	33%	20%	50%	100%	300%	25%	14%	1063%	5%	5%
Addition of other Materials	300%	50%	14%	20%	13%	20%	33%	100%	14%	33%	598%	3%	3%
Compatibility with Microcope Objective	400%	100%	50%	20%	25%	100%	400%	700%	100%	100%	1995%	10%	10%
Avalibility of Parts	800%	300%	33%	50%	50%	33%	700%	300%	100%	100%	2467%	12%	12%

Then the concept selection was completed as sown below and the concepts were ranked from best to worst.

Table 5: Concept Scoring Matrix

		Concept #1		Concept #2		Concept #3		Concept #4		Concept #5		Concept #6	
Selection Criterion	Weight (%)	Rating	Weighted	Rating	Weighted	Rating	Weighted	Rating	Weighted	Rating	Weighted	Rating	Weighted
Mechanical safety	1.35	3	0.0405	3	0.04	3	0.04	3	0.04	3	0.04	1	0.01
Cost of components	5.33	3	0.16	3	0.16	1	0.05	2	0.11	5	0.27	1	0.05
Strain Rate Variability	14.08	3	0.42	3	0.42	3	0.42	3	0.42	1	0.14	2	0.28
Ease of Logging data	13.19	3	0.40	3	0.40	3	0.40	3	0.40	3	0.40	3	0.40
Compatibility with Environmental Chamber	20.22	3	0.61	4	0.81	4	0.81	4	0.81	3	0.61	3	0.61
Compatibility with Lab Tissue	16.37	3	0.49	3	0.49	3	0.49	3	0.49	3	0.49	3	0.49
Age of Software	5.12	3	0.15	3	0.15	3	0.15	3	0.15	2	0.10	3	0.15
Addition of other Materials	2.88	3	0.09	3	0.09	3	0.09	3	0.09	3	0.09	3	0.09
Compatibility with Microcone Objective	9.6	3	0.29	3	0.29	3	0.29	3	0.29	3	0.29	3	0.29
Avalibility of Parts	11.87	3	0.36	3	0.36	1	0.12	2	0.24	1	0.12	1	0.12
	Total score	3.000		3.203		2.859		3.031		2.537		2.489	
	Rank	3		1		4		2		5		6	

4.2 EXPLANATION OF WINNING CONCEPT SCORES

This concept ended up being the winner. It scored exactly the same as the reference concept except for one category --Compatibility with the environmental chamber. The reason this concept earned a better score than the reference is because this concept will be using a “Fence” around the environmental chamber. The fence would sit in between the existing environmental chamber’s top and bottom sections. This would serve to help preserve the hermetic seal without having to recalibrate and engineer the heaters and thermometers inside the environmental chamber. It would also make it easier to put a motor arm in the chamber and still keep the cells alive.

4.3 EXPLANATION OF SECOND-PLACE CONCEPT SCORES

The servo motor concept narrowly finished ahead of the reference concept. This concept earned a 2 on the “cost of components” criterion vs the 3 of the reference. This is because this concept uses a servo motor instead of a stepper motor. This motor is more expensive and requires a controller to be operated which would increase the price. Like the Environmental Fence concept, the servo motor concept will also be using a fence for the environmental chamber so there is more room to work with inside the chamber. Lastly, this concept earned a 2 on the “availability of parts” criterion vs the 3 of the reference concept. This is due to the difficulty we might encounter in finding a servo motor that is the proper size and a controller that is easy to use.

4.4 EXPLANATION OF THIRD-PLACE CONCEPT SCORES

Our reference concept finished third. This concept employs the construction of a new environmental chamber lid that will fit on top of the existing bottom section of the environmental chamber. As for the construction of the lid, the most viable option would be to design it in Solidworks and have it 3D printed. The problem with this concept, and why it finished third, is that making a new lid that allows the environmental chamber to maintain proper conditions for the cells would be a much harder task than just making a “fence” to put between the top and bottom of the chamber. A fence would just enclose the inside of the chamber without adding any unwanted challenges.

4.5 SUMMARY OF EVALUATION RESULTS

The method used to determine how the various criteria in the concept selection matrix would be weighted relative to one another came largely from the analytical hierarchy chart that was made before completing the concept selection chart. The criteria in the hierarchy chart were weighted against one another by assigning ratios against one another collectively as a group. These criteria were either picked from or were largely influenced by the customer needs interview. Overall, the concepts were close to each other in score except the hand crank concept and magnetic concept. These two provided no real way to record data or accurately control how much the tissues were being deformed. As a result these two concepts scored lower than the other four. The results of the selection process reaffirmed our group's belief that the best way to move forward is to develop a technology that will coexist with the lab's existing technology and the best way to do that is to use the fence concept.

5 EMBODIMENT & FABRICATION PLAN

5.1 ISOMETRIC DRAWING WITH BILL OF MATERIALS

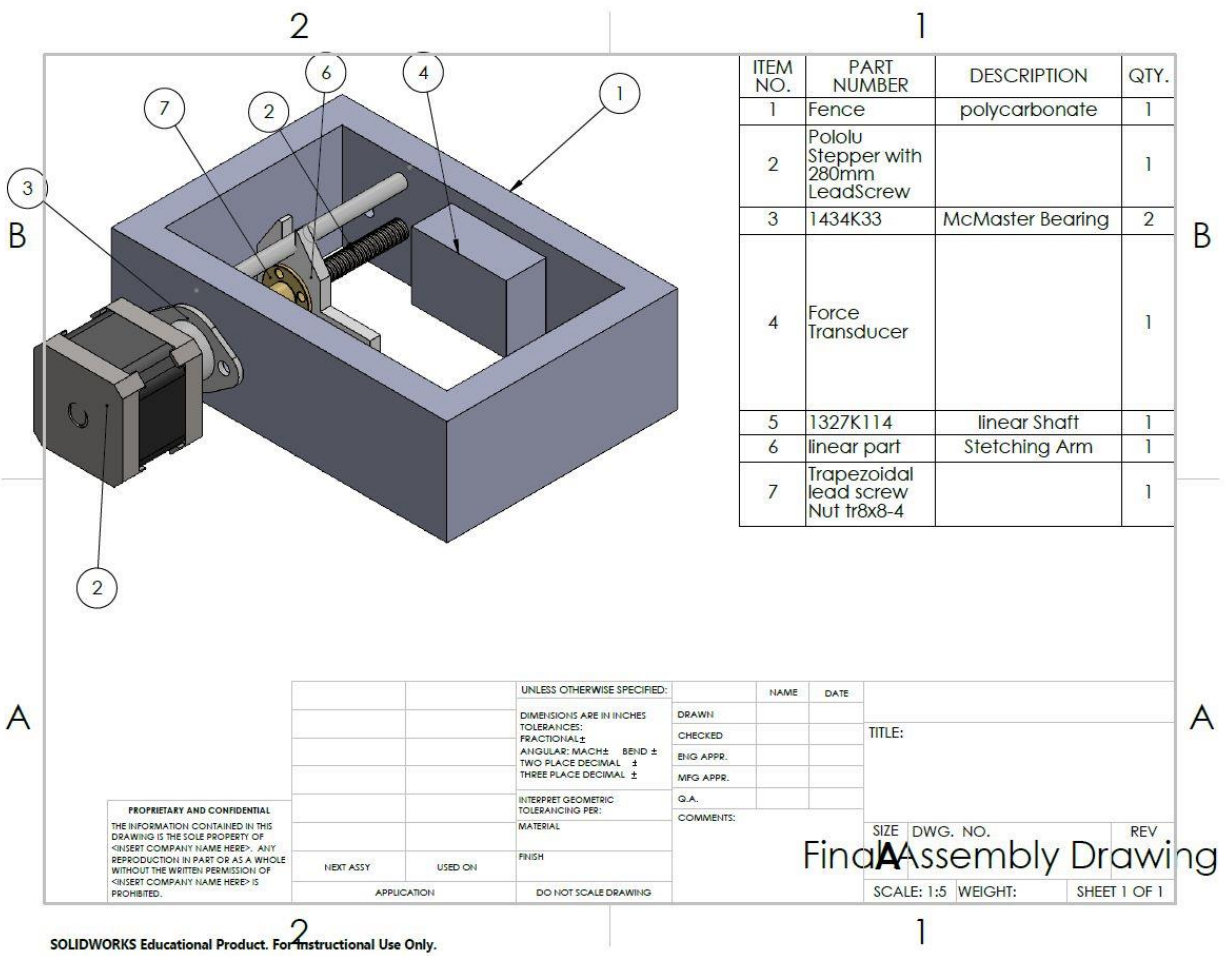
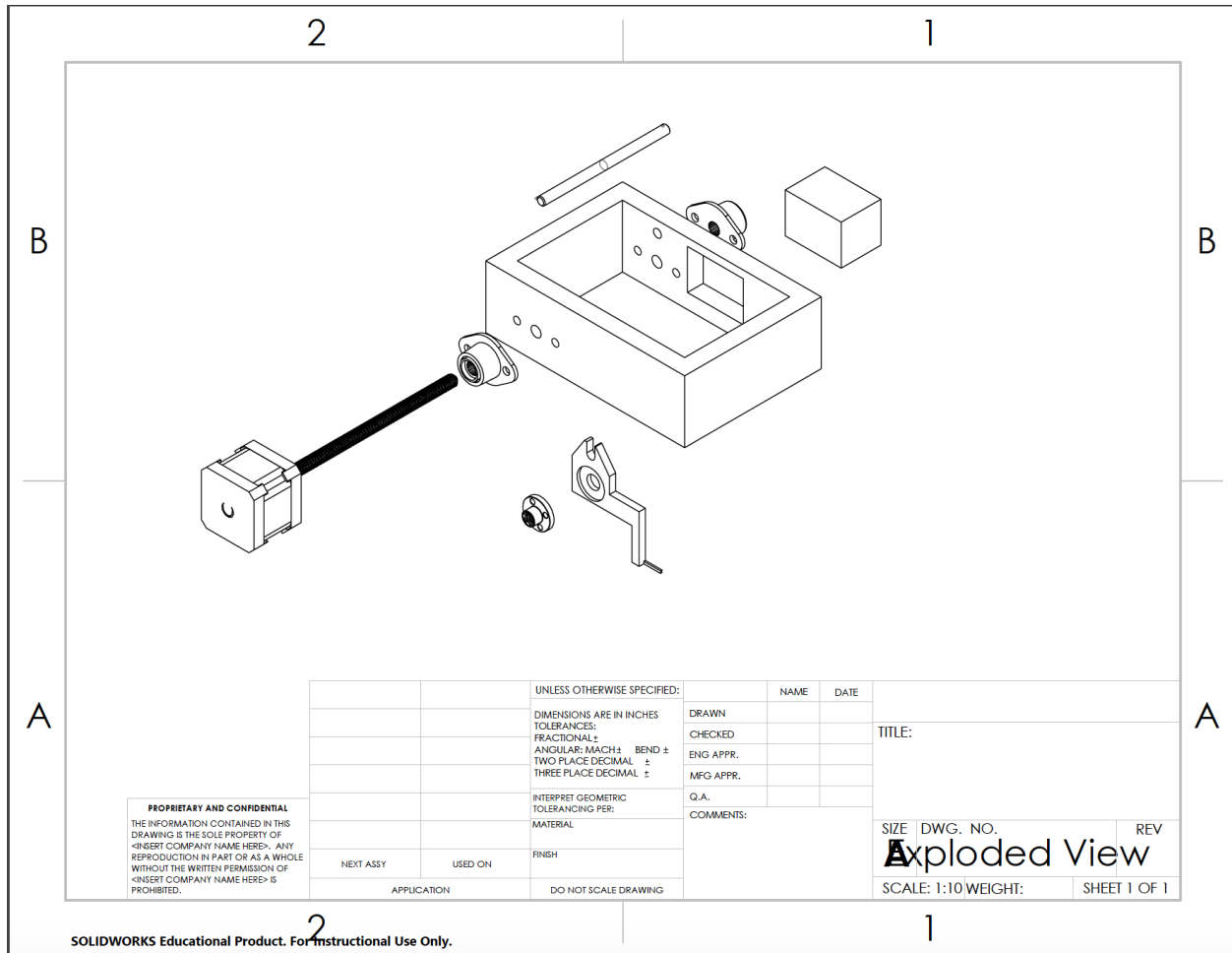


Figure 12 Isometric View of Chosen Concept

5.2 EXPLODED VIEW



6 ENGINEERING ANALYSIS

6.1 ENGINEERING ANALYSIS RESULTS

6.1.1 Motivation

The motivation for this design analysis is not rooted in a specific code or standard stems rather, from ensuring that the displacements that are recorded and used to calculate modulus and strain rate are accurate. While the forces that will be used to deform the tissues are not expected to be large in magnitude they still have the ability to deform whatever arm is used to stretch the tissue. This analysis is critical to do in the stages before creating the arm as it helps to save material and also helps to create an idea about how the arm will be made. As this part will have to fit in a confined space knowing how to make it will be key to manufacturing it.

6.1.2 Summary Statement of the Analysis

The mechanics behind the analysis is fairly simple and can be understood by approximating the arm that will stretch the tissue as a cantilever beam. Using Euler Beam Theory, the displacement of the arm that will stretch the arm can expressed as a function of the force applied to the beam

(P), the length of the beam (l), the moment of inertia of the beam (I), and the elastic modulus of the beam (E). This can be expressed by the equation shown below.

$$\delta_{\text{max}} = \frac{Pl^3}{3EI}$$

However, a computational approximation of this method is preferred over the hand calculated method for a few reasons. First, the connection between the beam arm and the rest of the apparatus is so thin that the cantilever connection of the beam is questionable. Secondly, a simulation helps to account for the whole stretcher arms geometry to help and visualize if the applied loads on just a single part of the part has unforeseen or uncalculated effects that will affect the part.

6.1.3 Methodology

The first step in carrying out the simulation was modeling the part in Solidworks. Using the dimension constraints of the environmental chamber and of the tissue sample itself the part was modeled. After modeling, its material was selected to be PLA plastic as it was anticipated to be 3D printed out of this material. After the stretcher arm was modeled in Solidworks, the Simulation plugin was used to simulate the force that would be applied to the part of the arm that would be attached to the tissue. Initially, the stretcher arm was modeled so that the tissue would be slipped around a cone-like tip to be stretched, this was done for the sake of reducing the amount of deflection that would be felt in the tip of the tissue stretcher as shown below.

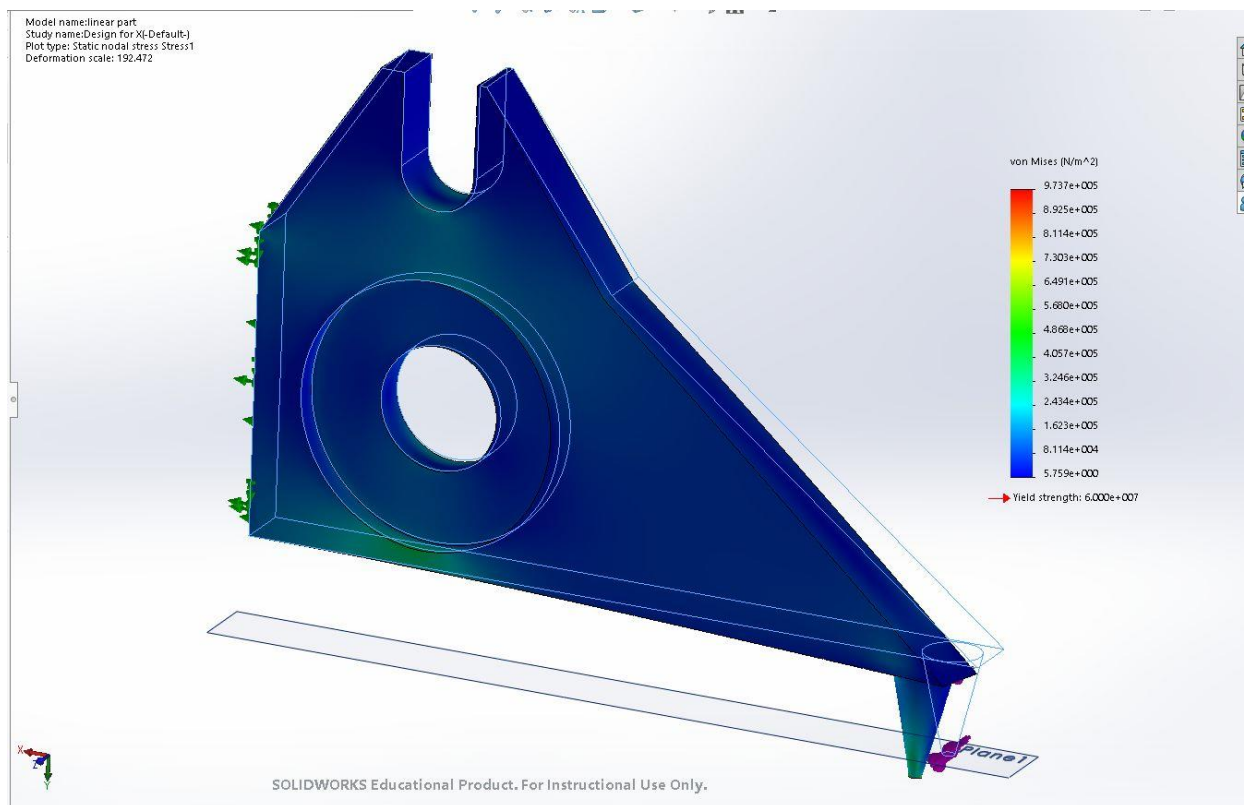


Figure 13- Simulation example

After testing the design with a prototype in the tissue stretcher, it was realized that the shape of the nib was not correct and that changes to the design were necessary to make sure that the tissue round could be attached to the stretcher arm and that it could be stretched without slipping off. After the design changes were made, the simulations were run again with the new shape.

6.1.4 Results

The results of the simulation studies for the displacement and the stress felt of the nib of the stretcher before and after the design change were recorded. Before the nib designed was changed and was still conical in nature, the max displacement seen of the nib was around .03 mm and the max von-mises stress was seen to be around 6 MPa. After the redesign to ensure that the tissue sample would not slip off the end of the nib, something that was seen when the conical shaped nib was tested. As it can be seen from the displacement simulation of the nib of the stretcher arm, when a force of 0.1 N (10 grams- the maximum force that is applied to the tissue samples) a maximum displacement of the nib was seen to be about 0.055 mm. When looking at the stress simulation, the maximum stress seen at the tip of the nib was seen to be 1.5 MPa. To make sure that any added stress from the redesign could not be alleviated from the use of ABS as the plastic used to 3D print the part, the part was also simulated with ABS as the material of choice. When ABS was used, the max displacement was seen to be .097 mm and the max Von Mises Stress was to be seen as 1.5 MPa.

6.1.5 Significance

The results of the studies completed above influenced the design of the stretcher in many ways and revealed much about the constraints that the actual required geometry of the tissue stretcher

placed of using the most effective design. When dealing with forces that are as low as 10 grams, and with strains of 10% for a sample that is only 5mm to begin with, displacements in the nib of up to .09 mm are very significant (.09 mm is almost 2% strain on a 5mm sample). The one positive form the redesign is that the max Von Mises stress was cut by 75% to 1.5 MPa ensuring that with a yield stress of 6×10^7 Pa, the part is not going to fail from stress of the applied load. Below are embodiment drawings of the part before and after simulation analysis was done (with both parts being made of PLA and not ABS). The before embodiments with max displacement and stress are shown below.

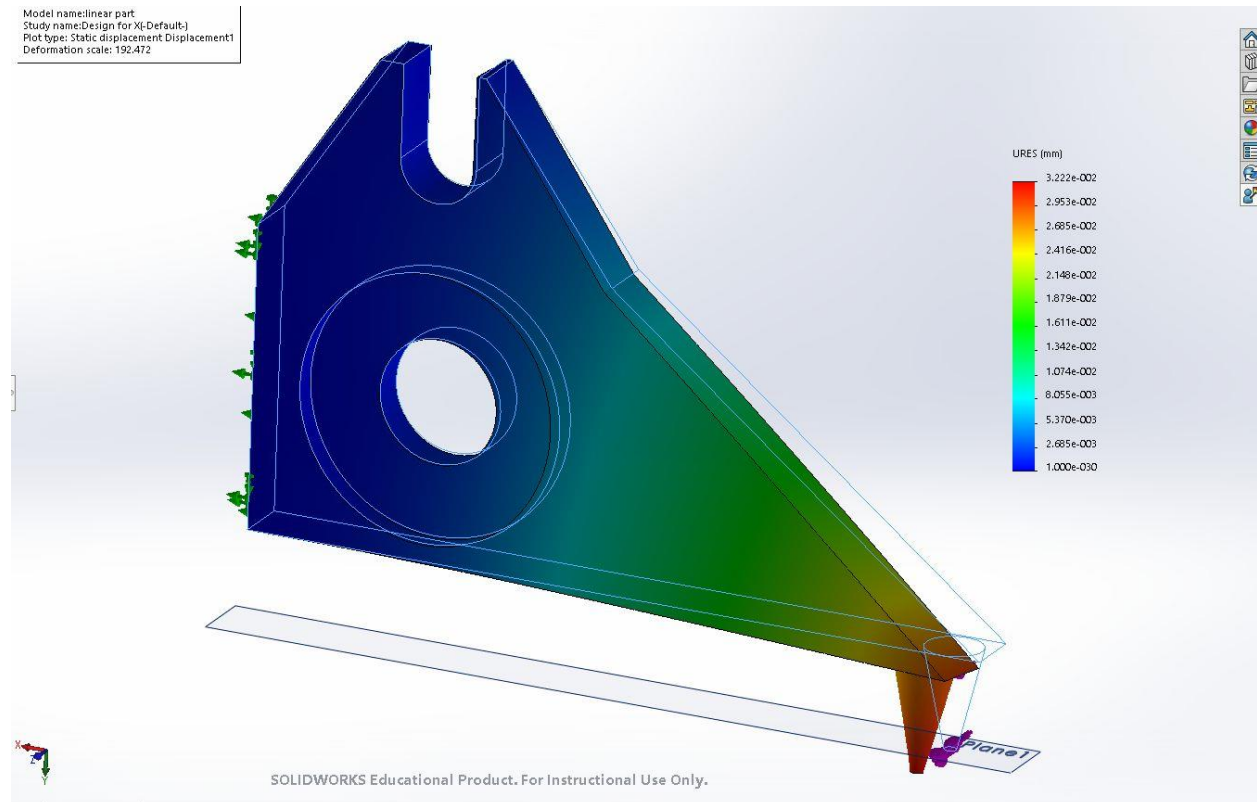


Figure 14-Displacement Simulation Before

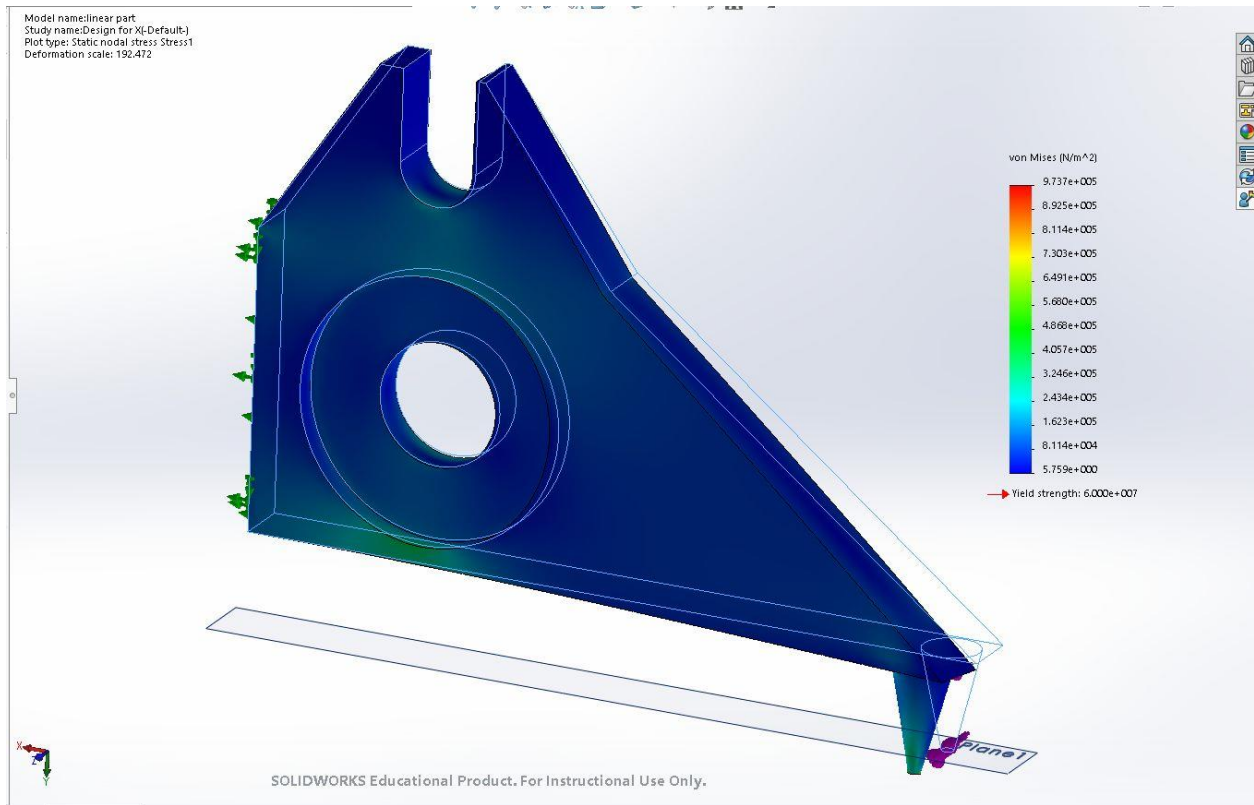


Figure 15- Von Mises Stress Before

The embodiments of the redesign are also shown below with displacement and stress values.

Model name: linear part Draft Analysis
Study name: Design for XI-Default-1
Plot type: Static displacement Displacement1
Deformation scale: 121.584

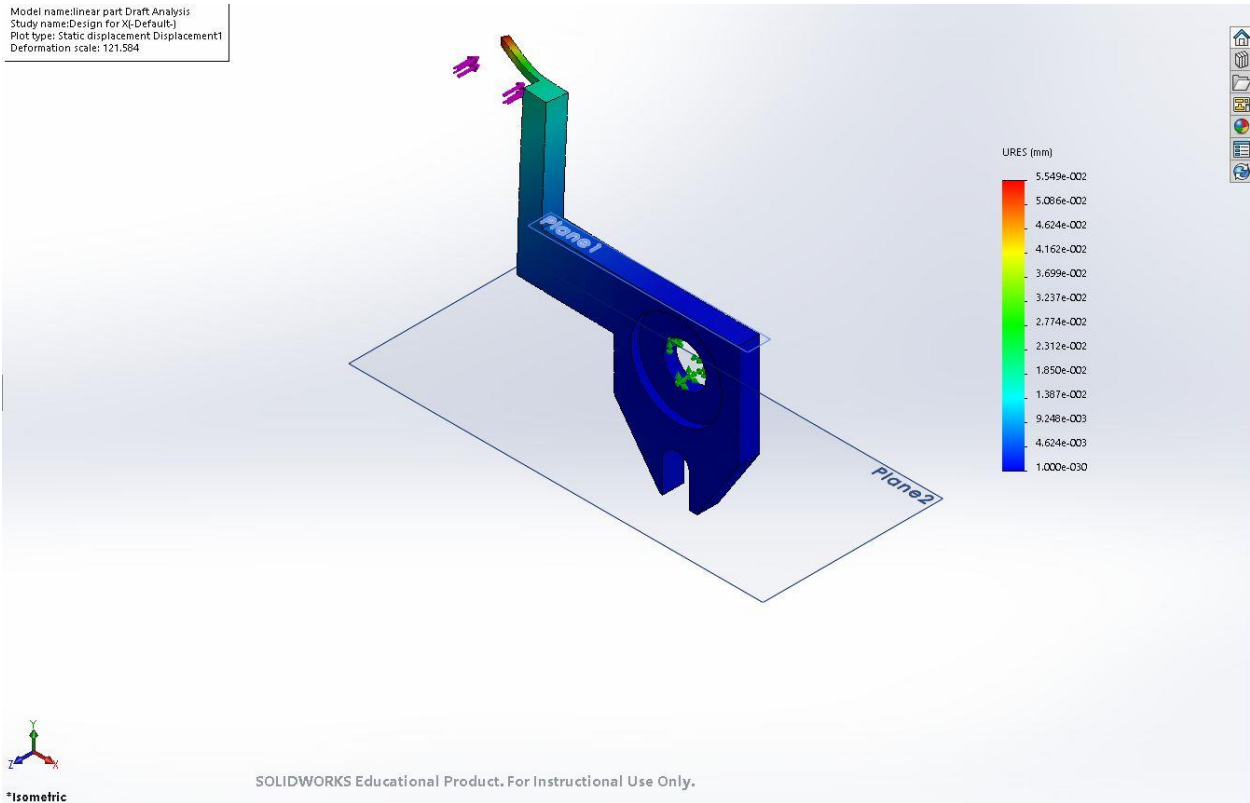


Figure 16- Displacement after redesign

Model name: linear part
Study name: Design for XI-Default-1
Plot type: Static nodal stress Stress1
Deformation scale: 121.584

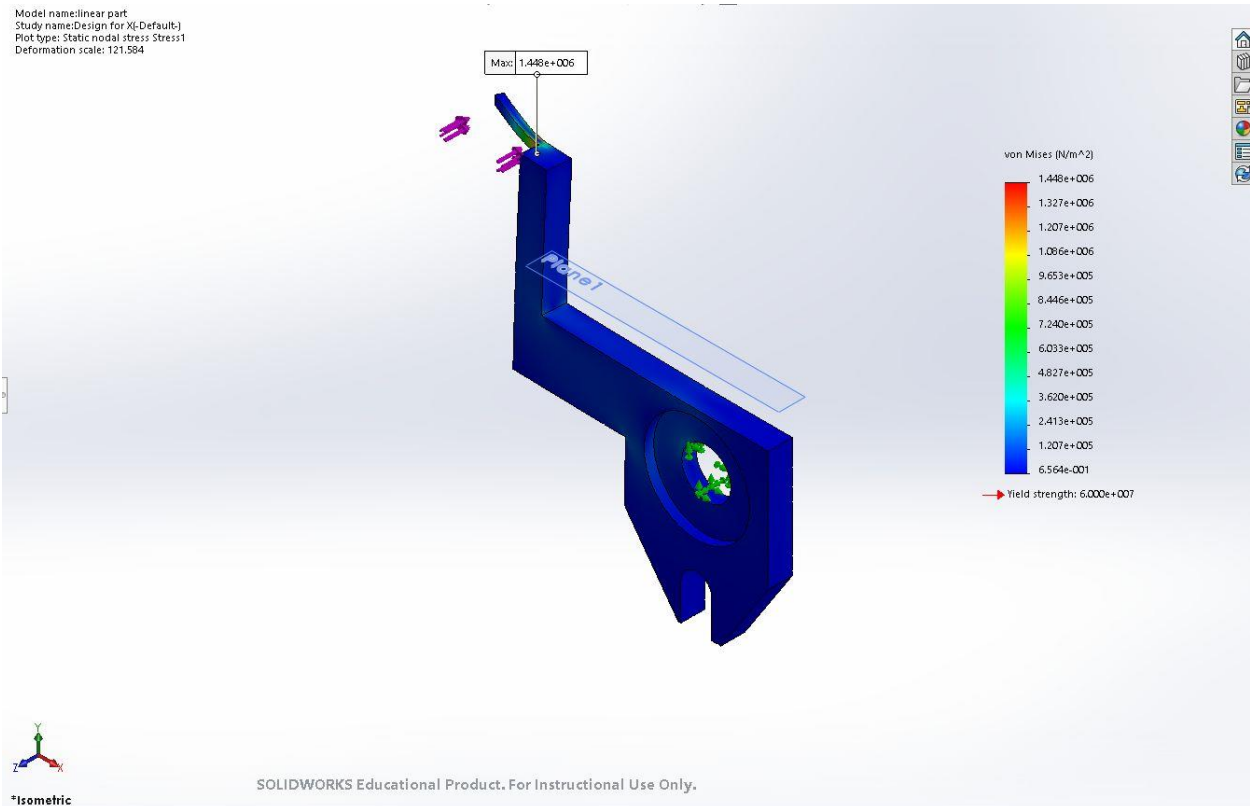


Figure 17- Von Mises Stress after redesign

As it can be seen from the before and after embodiments of the piece, the redesign does not make much sense when considering the amount of displacement that would be felt on the nib. An increase in max displacement by about .03 mm is not insignificant when dealing with displacements of about 1mm. However, the max stress seen in the redesign is 25% of the stress seen before the redesign. This is significant because it ensures that the arm would not break under load. Although it may not seem that the redesign is a logical step, it was undertaken anyway do to the fact that the first design was not able to securely stretch the tissue without the tissue falling off. Thus, it was deemed trivial that the first design displaced less under load because it would never be able to be used to stretch tissues effectively. Because of this, the embodiment after redesign was the one used in the final embodiment. Also, because of the numbers seen in the Results section, PLA was used over ABS for the final embodiment as it minimized displacement and maximum stress.

6.2 PRODUCT RISK ASSESSMENT

6.2.1 Risk Identification

Risk Name: Overheat

Description: Occasionally when testing our device, we have had instances when the either the motor, Arduino, or battery pack get really hot. This seems to be caused by the device being in use for too long, or by the 12V battery pack supplying too much voltage to the Arduino.

Impact = 3: If someone were to touch a part that was over heating then they could experience pain or a burn depending on how long they touch the hot part. If a part is too hot for too long then it may break which would require a new part to be purchased.

Likelihood = 2: We have only had this happen twice during testing.

Risk Name: Force Transducer Break

Description: A force transducer must be placed into a hole in our polycarbonate fence so that data can be collected. The force transducer is secure once in the hole but if the user is not careful when putting it in, then they could drop it which would break it instantly because the transducer is designed to measure very small forces (10 N in magnitude).

Impact = 4: The particular force transducer that our device uses costs upwards of \$500 so breaking it would not be good.

Likelihood = 1: Anyone who uses this device will have knowledge of how delicate the force transducer is, so they will be extra careful not to drop it.

Risk Name: Gap in Fence

Description: Our device is designed to be used with an existing environmental chamber to keep the artificial cells at 37 degrees Celsius as well as other important atmospheric conditions. If the device is not placed between the environmental chamber properly then the chamber cannot keep the cells at the desired conditions and the cells will die and not be able to be viewed.

Impact = 1: Dr. Genin's lab makes artificial cells frequently so obtaining new ones to test is a very small problem.

Likelihood = 1: Our device fits easily between the environmental chamber lids so the user would need to be very careless in order to place it wrongly.

Risk Name: Dropping Tissue Loader on Microscope or Environmental Chamber

Description: Our device weighs around 5lbs. If it was dropped while being placed on the microscope then it could damage the microscope and/or the environmental chamber.

Impact = 5: Confocal microscopes are very expensive, so doing any damage to that would be devastating. Dr. Genin's lab has been using the same environmental chamber for a decade and it is no longer produced. If that were to be damaged then they would need to buy a new one and have to familiarize themselves with a new chamber all over again.

Likelihood = 1: A device that weighs 5 lbs. would need to be dropped from 10+ feet in order to seriously damage the microscope and/or the environmental chamber. That has no chance of occurring.

Risk Name: Motor Lead Screw

Description: The lead screw of our stepper motor is sticking out from our fence by 3 inches. If someone were to walk by and clip the screw then our device would fall over and break. This would also result in damage to the environmental chamber.

Impact = 3: If our tissue loader were to be dropped from the height of where it is mounted to the microscope, then it would probably break. Also, as stated before, the environmental chamber is an important part to Dr. Genin's lab so damaging would be significant.

Likelihood = 1: The protrusion of the lead screw out of the fence is too small to hang over the edge of the microscope so the chances of clipping it on clothing is very minimal.

Risk Name: Motor Misuse

Description: Our device uses a MATLAB code to run the stepper motor. The code is set up to spin the lead screw a certain RPM and to stretch the tissue a certain distance. If the user inputs commands carelessly then the cells could be damaged.

Impact = 1: Dr. Genin's lab makes artificial cells frequently so obtaining new ones to test is a very small problem.

Likelihood = 3: We have already played with the motor in order to find the best settings for it, but there is still a chance the user wants to edit the motor commands. On top of that, when inputting the stretching distance the user might accidentally type 5 mm instead of 0.5mm.

6.2.2 Risk Heat Map

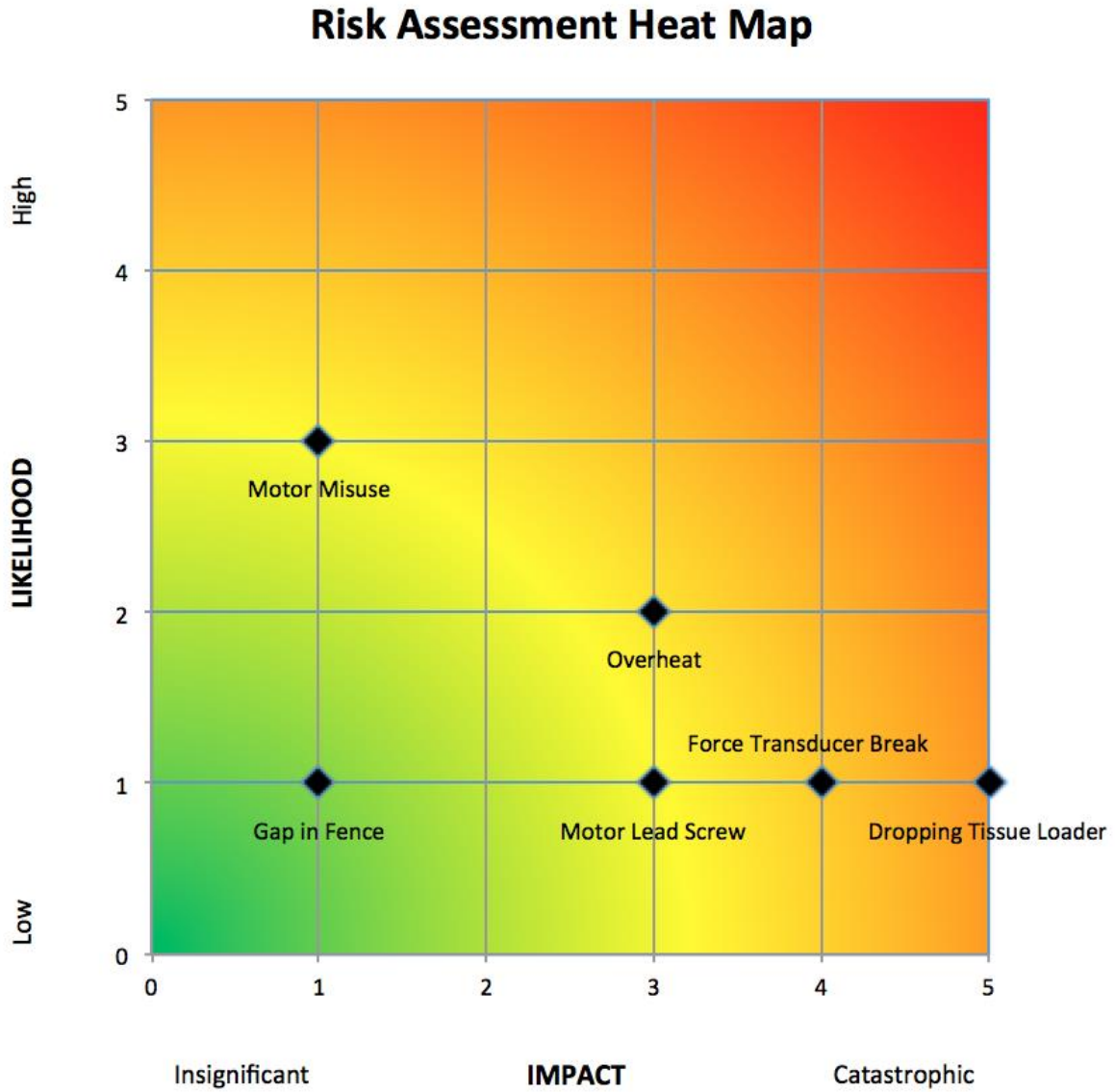


Figure 18. Risk Assessment Map

6.2.3 Risk Prioritization

1. Overheat
2. Dropping Tissue Loader
3. Force Transducer Break
4. Motor Lead Screw and Motor Misuse
5. Gap in Fence

It makes sense that the risk that involves user safety is the risk that we should prioritize the most. In order to decrease the likelihood of this risk, we will try a reducing our battery voltage from 12V to 3V to see if we were overpowering our motor which resulted in our system over heating.

7 DESIGN DOCUMENTATION

7.1 PERFORMANCE GOALS

- Strain rate of up to 10% per second for 1 second
- Maintain constant temperature setting of 37 degrees Celsius in chamber as well as 5% CO2
- 10x objective is still able to be used with system
- User input is controlled through MATLAB and Data is exported as text file and can be analyzed with MATLAB or Excel
- Components can be easily moved from microscope (less than 1 minute)

7.2 WORKING PROTOTYPE DEMONSTRATION

7.2.1 Performance Evaluation

We were able to accomplish all of our performance goals

7.2.2 Working Prototype – Video Link

https://www.youtube.com/watch?v=vL6jq_5HaKw

7.2.3 Working Prototype – Additional Photos

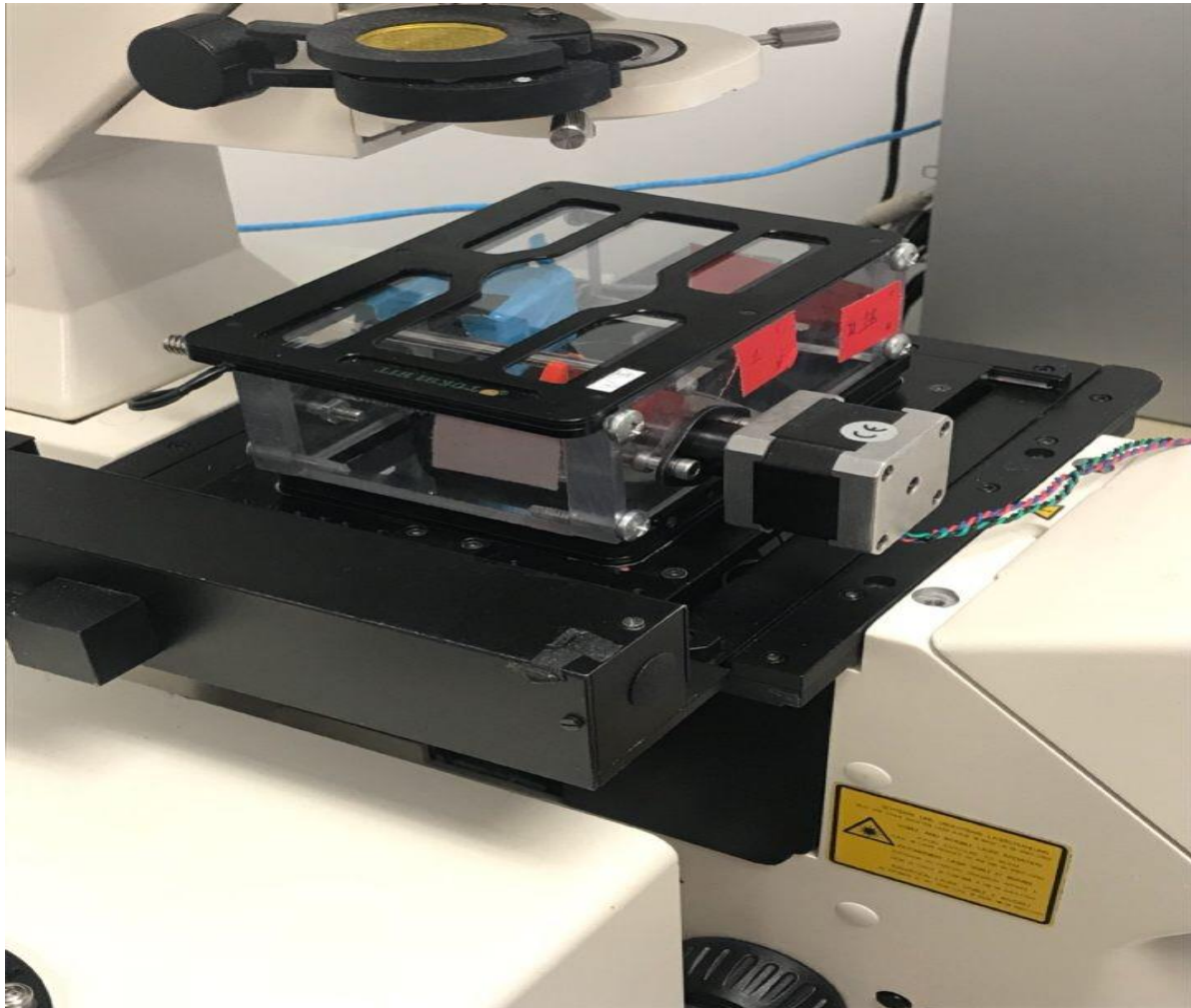


Figure 19. Prototype on Microscope

8 DISCUSSION

8.1 DESIGN FOR MANUFACTURING – PART REDESIGN FOR INJECTION MOLDING

8.1.1 Draft Analysis Results

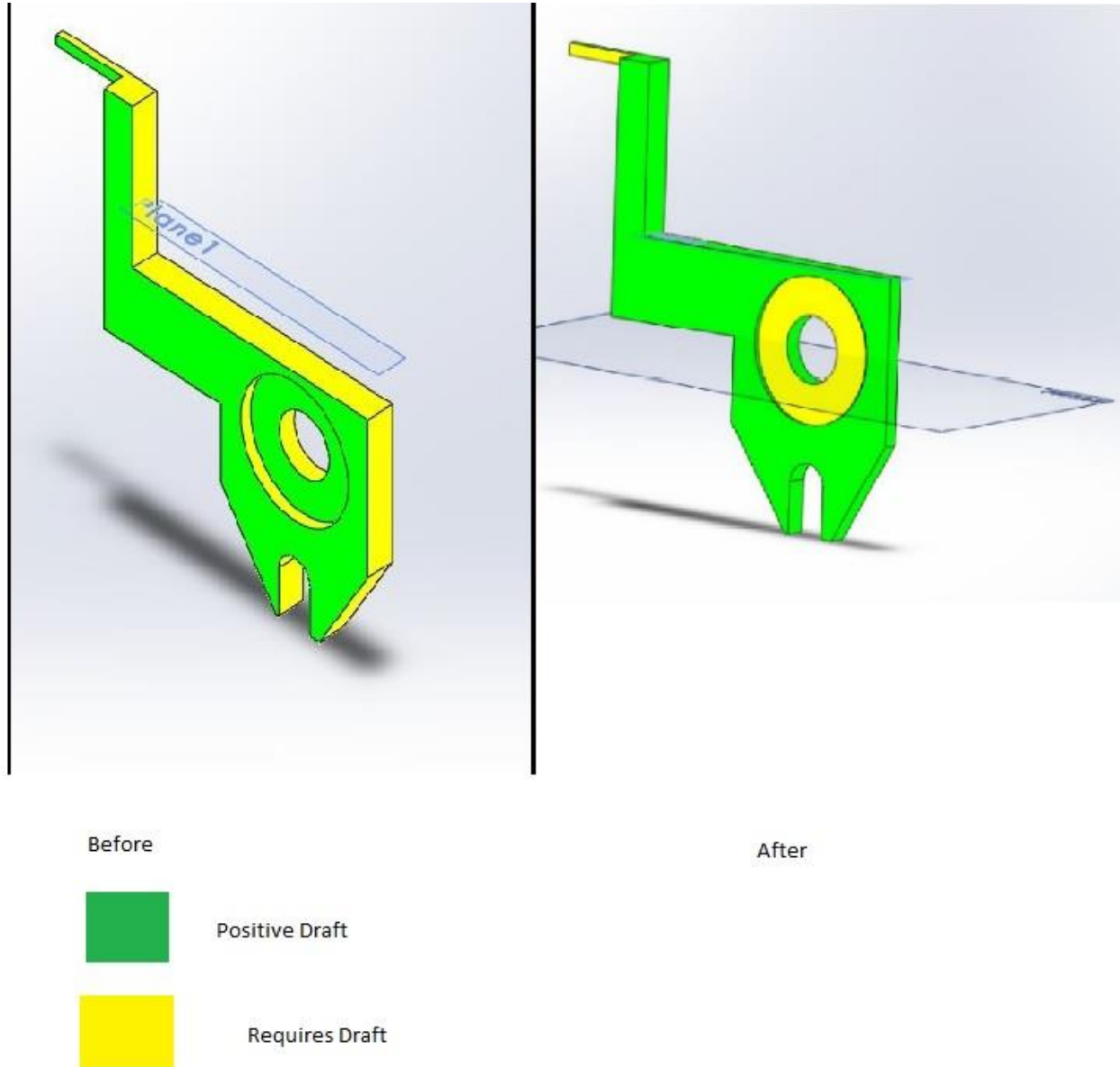


Figure 20. Draft Analysis

8.1.2 Explanation of Design Changes

The part was changed by drafting the front face of the part by 3 degrees. This allowed all of the yellow vertical sections in the before photo to become slightly slanted and by extension allowed them to become drafted. The only section that cannot be drafted is the middle circle section. This

is because the traveling nut for the lead screw of the motor fits into that section and thus it must remain vertical so that it can interface with the nut.

8.2 DESIGN FOR USABILITY – EFFECT OF IMPAIRMENTS ON USABILITY

8.2.1 Vision

- If someone suffers from presbyopia then they may have some trouble using our tissue loading device. It all depends on how severe their vision has worsened. Our device uses an arduino so in order to function properly, the motor must be connected to the arduino. The wires for the motor must be screwed into the arduino using a tiny screwdriver. This maybe be a problem for someone with very poor vision. This issue can be fixed if the user uses a magnifying glass.
- The wires for the motor are color coded and need to be hooked up to the arduino in a certain order. This would be a problem for someone who suffers from color blindness. This problem can be fixed by labeling the wires by their specific color.

8.2.2 Hearing

- Our tissue loading machine does not require the user to need to be able to hear. The computer, arduino, tissue loader, and confocal microscope do not make any command noises so hearing is not required.

8.2.3 Physical

- Our tissue loader machine weighs about 5 pounds. If the user has a physical condition, such as muscle weakness or other conditions, that inhibits them from lifting 5 pounds and placing it on the confocal microscope then they will have issues using our device. This issue can be fixed by the user seeking assistance from a lab partner or friend. To help place the device on the microscope.

8.2.4 Language

- Our tissue loading machine is used in conjunction with MATLAB. Our program is written in English so the user would need to know English in order to customize the way the motor runs and stretches the tissue. MATLAB is however available in other select languages. The user would need to translate our code into the language they prefer and then they would be able to run and customize the way the tissue is stretched efficiently.

8.3 OVERALL EXPERIENCE

8.3.1 Does your final project result align with the initial project description?

Yes, we were able to make a device that can be used with a confocal microscope to stretch a tissue while the tissue is being viewed.

8.3.2 Was the project more or less difficult than you had expected?

It was about as difficult as we expected. We knew it would be hard to build a device that could be used with the existing environmental chamber but by going through the steps in the engineering process and getting help from professor Potter we were able to accomplish our design goal.

8.3.3 In what ways do you wish your final prototype would have performed better?

We wish the batteries wouldn't get hot after using the device for over 10 minutes. If we had more time we could accomplish this by powering the motor from a wall outlet.

8.3.4 Was your group missing any critical information when you evaluated concepts?

We were not missing any critical information when we evaluated concepts.

8.3.5 Were there additional engineering analyses that could have helped guide your design?

One additional analysis we could have done was to calculate how much longer it takes for the environmental chamber to heat up with the added volume of the fence that we created.

8.3.6 How did you identify your most relevant codes and standards and how they influence revision of the design?

We looked for standards that dealt with the testing of tissues in "ovens." One standard that applied to our project was ASTM E95-68 (4.3). This standard said that the chamber must be kept within 1 degree Celsius of the desired conditions. This influenced our design by having to make a fence that was airtight.

8.3.7 What ethical considerations (from the Engineering Ethics and Design for Environment seminar) are relevant to your device? How could these considerations be addressed?

There are no ethical considerations relevant to our device.

8.3.8 On which part(s) of the design process should your group have spent more time? Which parts required less time?

We should have spent less time making our fence. It took us about a week to make it since none of us had a lot of machine shop experience. If we were skilled machinists we could have made the fence in a couple of hours. We should have spent more time finding out how the force transducer worked at the beginning of our design process. We were a little taken back when we found out the sensor could only collect force applied to it perpendicularly. We were able to adjust for this, but if we spent more time at the beginning working with the force transducer it could have saved us the trouble.

8.3.9 Was there a task on your Gantt chart that was much harder than expected? Were there any that were much easier?

Creating 6 concepts was actually a pretty hard task at first. But once we thought of one concept we were able to come up with another 5 relatively quickly. Choosing the best concept was much easier than expected. Our feedback from the class as well as our concept review assignment helped us out with this task.

8.3.10 Was there a component of your prototype that was significantly easier or harder to make/assemble than you expected?

The polycarbonate fence was much harder to make than we expected. The fence had to be airtight and precise so when we made it we had to make sure all of our holes aligned perfectly and all of our sides were the same height. Making the arm was very easy since we just 3D printed it.

8.3.11 If your budget were increased to 10x its original amount, would your approach have changed? If so, in what specific ways?

Our approach would not have changed, but our final design would have changed. With a greater budget we would be able to purchase a force transducer that would fit better with our design. With our current budget we just used Dr. Genin's force transducer. This force transducer did not work perfectly with our design because force data was only collected by pulling on the sensor in a perpendicular direction. If we

bought a force transducer that could collect data by being pulled in a parallel direction to the sensor, then our device would collect more accurate data.

8.3.12 [If you were able to take the course again with the same project and group, what would you have done differently the second time around?](#)

We would design our fence so it fit the force transducer more appropriately.

8.3.13 [Were your team member's skills complementary?](#)

Yes. Our team worked very well together. Anytime one of us had a problem another one of us could step up and help solve the problem.

8.3.14 [Was any needed skill missing from the group?](#)

None of us had a lot of programming experience with MATLAB and Arduinos before this project. If we had someone who was a master at programming in our group then I am sure we would've been able to improve our device in some way.

8.3.15 [Has the project enhanced your design skills?](#)

This project has definitely enhanced our design skills. We were able to create a design that successfully performed the way we wanted.

8.3.16 [Would you now feel more comfortable accepting a design project assignment at a job?](#)

We would feel a lot more comfortable accepting a design project assignment at a job. We learned a lot doing this design project that we could apply to the real world.

8.3.17 [Are there projects you would attempt now that you would not have attempted before?](#)

We would all feel more comfortable attempting a project that involved coding after doing this senior design project.

9 APPENDIX A - PARTS LIST

Table 6: Part List

	Part	Source Link	Supplier Part Number	Color, TPI, other part IDs	Unit price	Tax (\$0.00 if tax exemption applied)	Shipping	Quantity	Total price
1	Adafruit Motor/Stepper/Servo Shield for Arduino v2 Kit - v2.3	adafruit	1438		\$19.95	\$0.00		1	\$19.95
2	Arduino Uno R3 (Atmega328 - assembled)	adafruit	50		\$24.95	\$0.00		1	\$24.95
3	9 VDC 1000mA regulated switching power adapter	adafruit	60		\$6.95	\$0.00		1	\$6.95
4	USB Cable - Standard A-B - 3 ft/1m	adafruit	62		\$3.95	\$0.00	\$9.16	1	\$3.95
5	NEMA 17 Integrated TR8x8 Lead Screw Stepper Motor 300mm-350mm	RepRap	N/A		\$42.99	\$0.00	\$7.91	1	\$42.99
6	Clear Polycarbonate 12"x12"x1/2"	McMaster	8574K32	Clear	\$32.34	\$0.00		1	\$32.34
7	Light Duty Two-Bolt Flange-Mounted Roller Bearing	McMaster	1434K33	black	\$11.14	\$0.00		1	\$11.14
8	Clear Polycarbonate 6x12x1/2	McMaster	8574K322	clear	\$17.78	\$0.00		1	\$17.78
9	Dewalt Alan Wrench Set	Home Depot	76174027583		\$10.97		\$0.00	1	\$10.97
10	Wire	Home Depot	49223501147		\$3.67		\$0.00	1	\$3.67
12	magnetic tape	Home Depot	95421070534	black	\$3.24		\$0.00	1	\$3.24

13	AA batteries	Home Depot	41333048642		\$8.98		\$0.00	1	\$8.98
14	3M screws	Home Depot	8.8748E+11		\$0.75		\$0.00	1	\$0.75
16	10/32 Screws Button Head	Home Depot	30699027583		\$1.05		\$0	2	\$2.10
17	10/32 Screws cap screw	Home Depot	8.8748E+11		\$1.85	\$3.74	0	4	\$7.40
18	Recharchable Li-ion battery	Microcenter	1917		\$8.99		0	1	\$8.99
19	Charging Cable	Microcenter	929448		\$14.99	\$2.42	0	1	\$14.99
20	Rotary Shaft	McMast er	1327K114	Silver	\$3.93	\$0.00	0	1	\$3.93
Total:						\$6.16	\$17.07		\$248.30

10 APPENDIX B - CAD MODELS

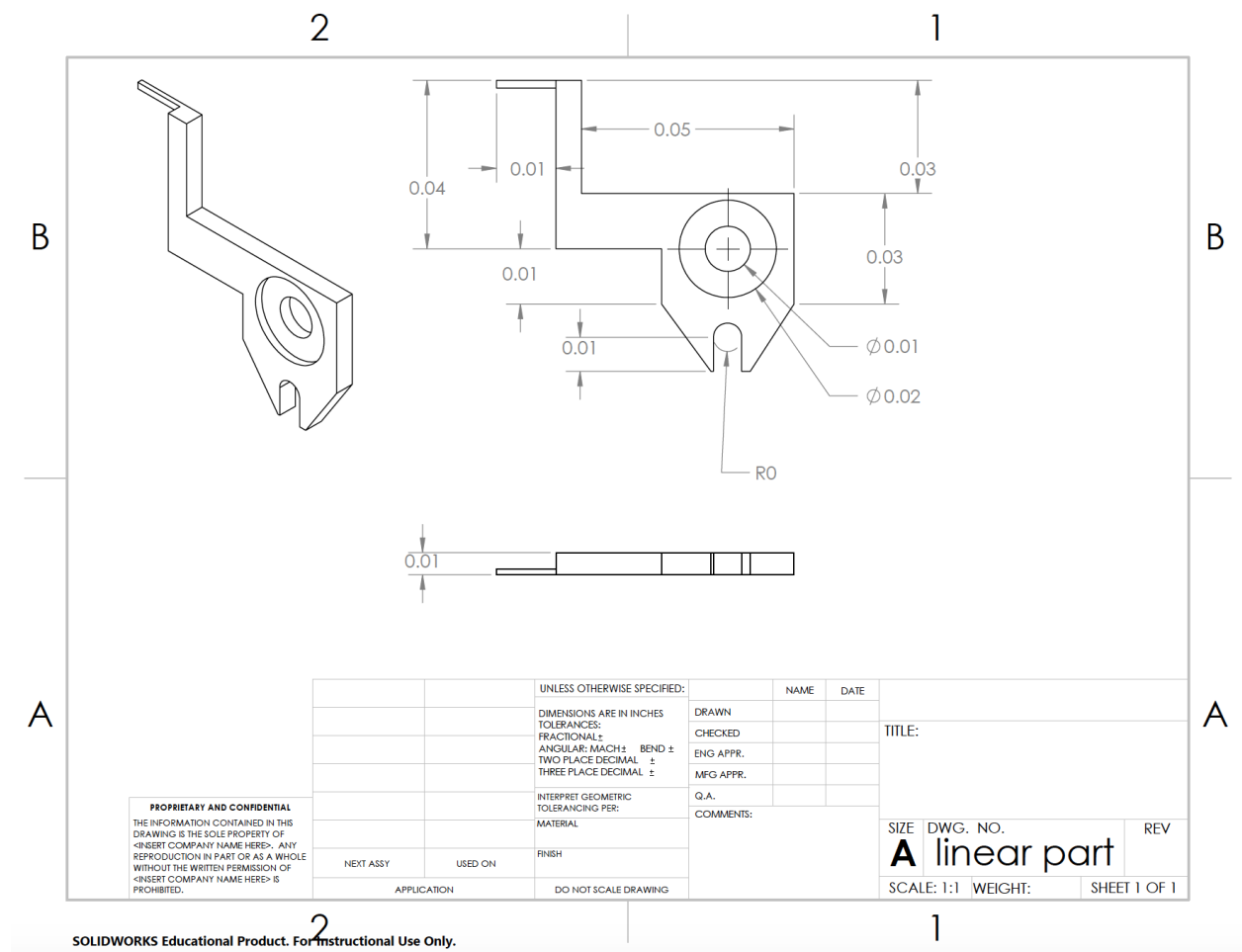


Figure 21 Stage Part Dimensioned Drawing

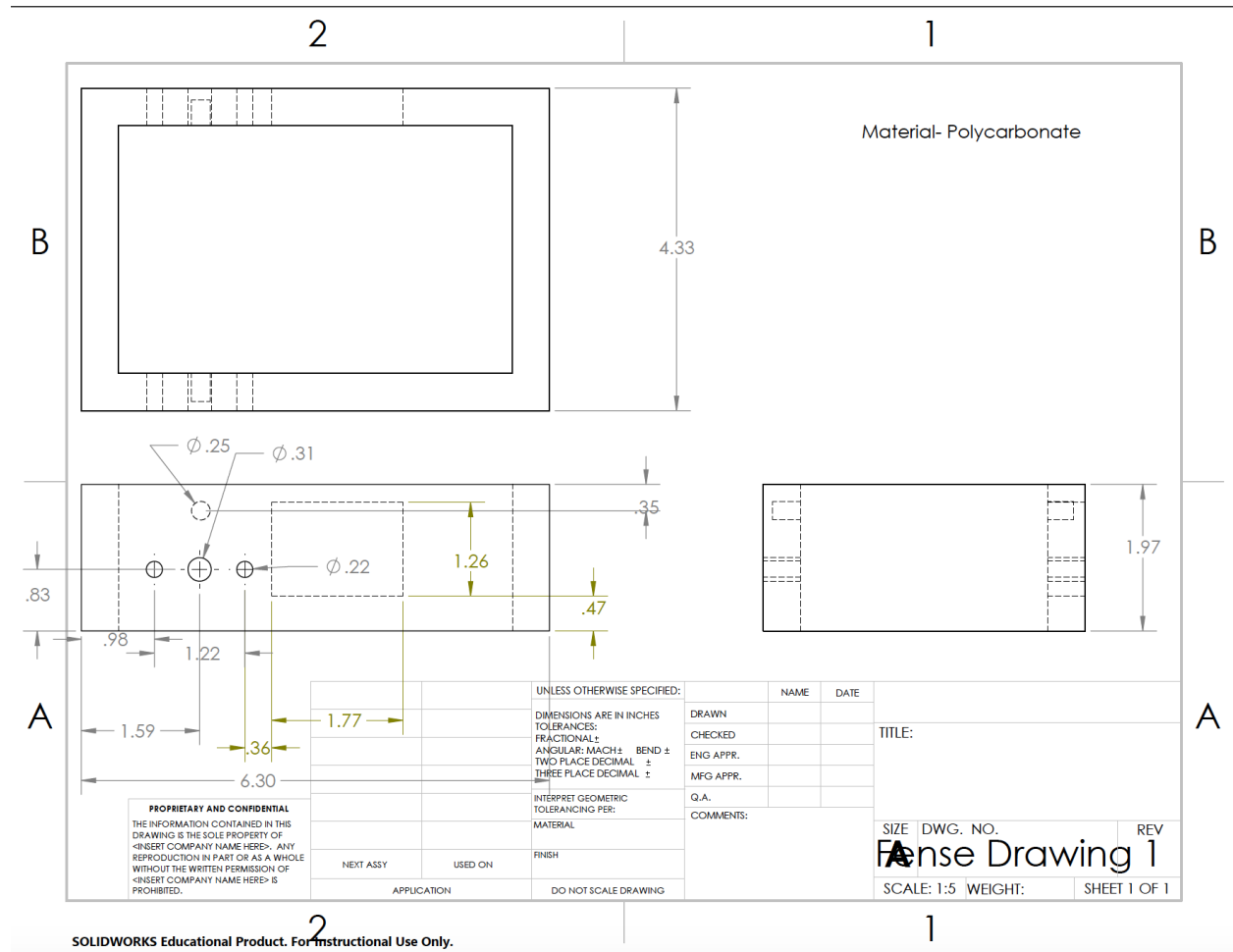


Figure 22 Fence Dimensioned Drawing

11 ANNOTATED BIBLIOGRAPHY

ASTM E95 - 68(2016). Standard Specification for Cell-Type Oven with Controlled Rates of Ventilation. Active Standard ASTM E95 | Developed by Subcommittee: E41.06

This specification covers the general and design requirements for two types of cell-type ovens based on their rates of ventilation, for determining loss in weight or changes in properties of materials on heating at elevated temperatures. This specification takes into account the fact that chamber geometry, rate of ventilation, and temperature each affect the rate of loss of volatile constituents from a material, or the rate of change in other properties. Hence, this oven is recommended whenever the results are dependent on the time and temperature of heating, the amount of ventilation, or both.

ASTM E211 - 82(2016). Standard Specification for Cover Glasses and Glass Slides for Use in Microscopy. Active Standard ASTM E211 | Developed by Subcommittee: E41.01

This specification describes the required properties and corresponding test methods for glass covers and slides for use in routine microscopy. The covers and slides comply to specified requirements as to dimension, planeness and parallelism, corrosion resistance, and workmanship. They shall also be tested for their conformance to other properties such as index of refraction, clarity, resistance to boiling, solubility, and wettability.

Cummings, Joel W, and Roland Deangelis. *SMALL TISSUE AND MEMBRANE FIXATION APPARATUS AND METHODS FOR USE THEREOF*. 27 Nov. 2001.

This patent is for a surgical tissue pinning device. It was developed as a way to pin down tissues and membranes during surgery as a faster and less intensive way than the traditional method of using sutures. This method of fixing tissue could prove useful when trying to attach the tissue our product is trying to load to the surface that it is mounted on and to the loading apparatus itself. This is especially true when a large amount of samples will have to be quickly mounted and then imaged.

“Planar Biaxial.” *TA Instruments*, 2017, www.tainstruments.com/planar-biaxial-2-motor/.

The ElectroForce Planar Biaxial TestBench instrument offers superior control and unparalleled performance in material and soft tissue characterization. Using this instrument, you can assess mechanical anisotropy and stress-strain relationships in samples that range from engineered devices, including wearable sensors and wound repair meshes, to tissues such as skin, pericardium, and heart valve leaflets. This instrument can be configured with either two or four ElectroForce linear motors mounted on a horizontal baseplate and load cell options for each axis of loading