

SHEAR STRESS BIOREACTOR TO STUDY MECHANOBIOLOGY OF STEM CELLS

Brian Penney, Lola Bangudu, Nikhil Nayar and Sonja Tollefson

Objective

Design, build, and evaluate an inexpensive bioreactor system that can apply a constant shear stress to cultured stem cells within a standard cell incubator environment.

Background

- ❖ Stem cells have the potential to differentiate into any type of cell in the body
- ❖ Tenogenesis, the differentiation of stem cells to tendon cells, comes from many unknown factors
- ❖ Naturally, tendon cells have collagen fibers slide past them, inducing a shear stress on the cells
- ❖ Dr. Schiele's lab currently investigates how shear stress affects tenogenesis using an orbital shaker which is not ideal as shear is approximated from velocity and cells experience inconsistent stresses
- ❖ Current shear stress bioreactors on the market are too expensive and incompatible

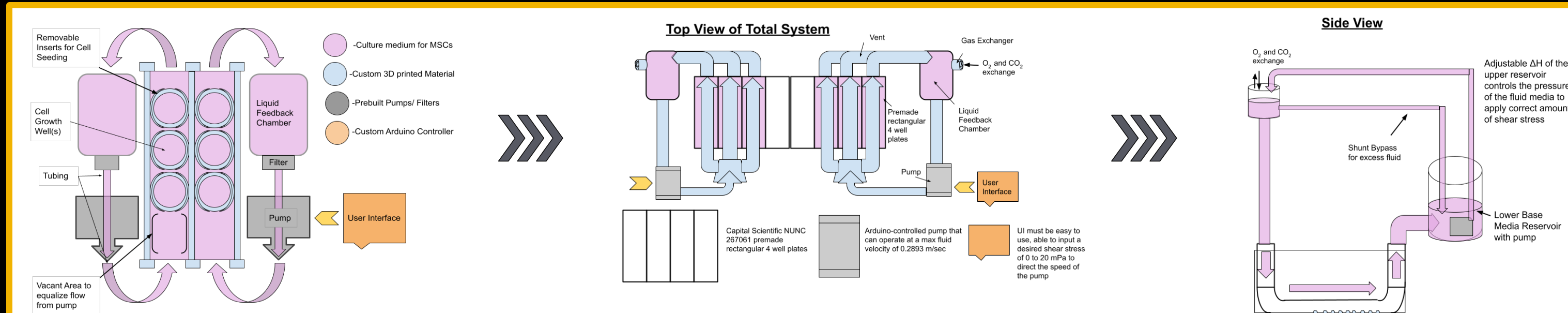


By further studying tenogenic factors, tendon can be engineered and produced from stem cells to serve as medical transplants for one of the most injured tissues in humans.

Requirements

- Apply a maximum of 20mPa of shear stress
 - Using flow of culture medium
- Fit within the dimensions of a standard incubator (36cm x 30cm x 30cm)
- Withstand incubator environment
 - 37 °C and 95% humidity
- Minimum of 2 treatment groups
- Autoclavable or able to be sterilized in 70% ethanol
- Function independently or with little supervision
 - Experiments last up to 21 days
- User-friendly interface
- Budget of \$1500

Conceptual Development



Initial designs utilized custom 3D-printed channel plates and seeding inserts to run fluid over cultured cells. To minimize custom parts, pre-existing 4 well culture plates and custom inserts were chosen. Due to the extremely low volumetric flow in the system, no commercially available pump was viable for moving the fluid accurately. A valve-controlled gravity-fed system was then chosen to provide an adjustable array of shear stresses.

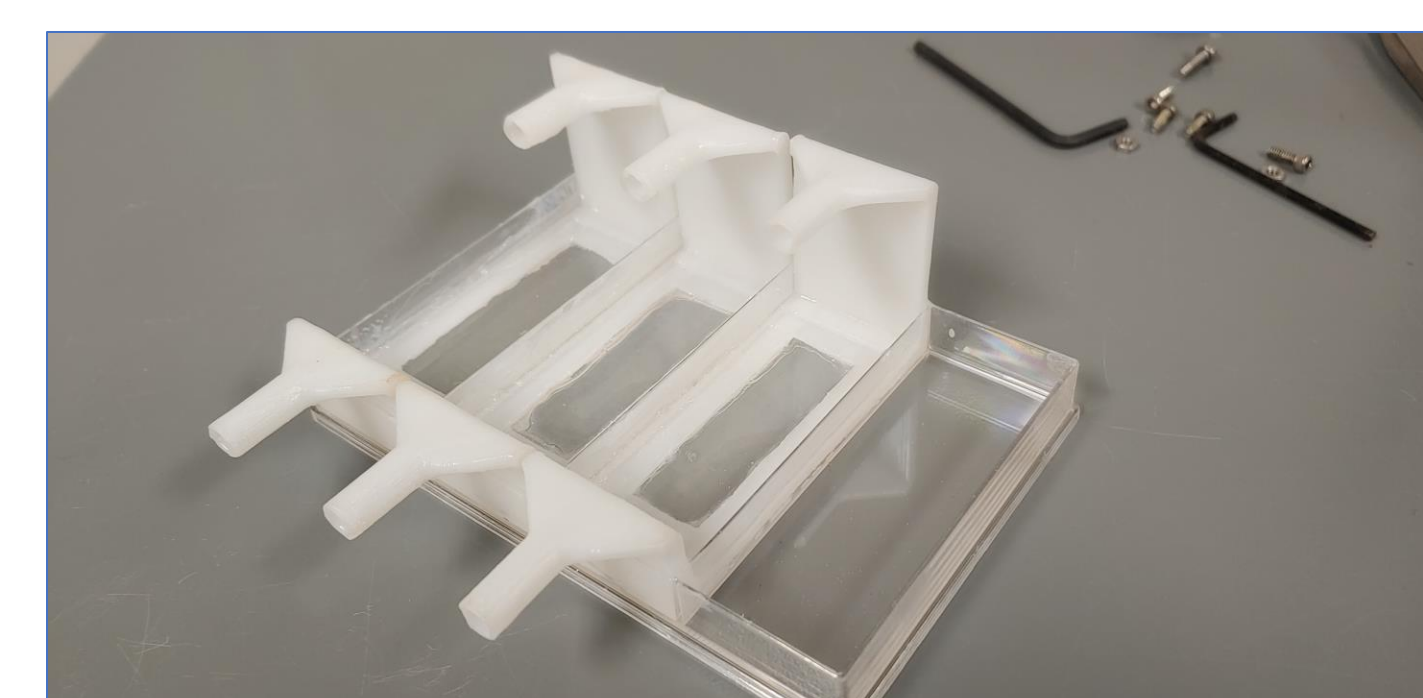
Finalized Design



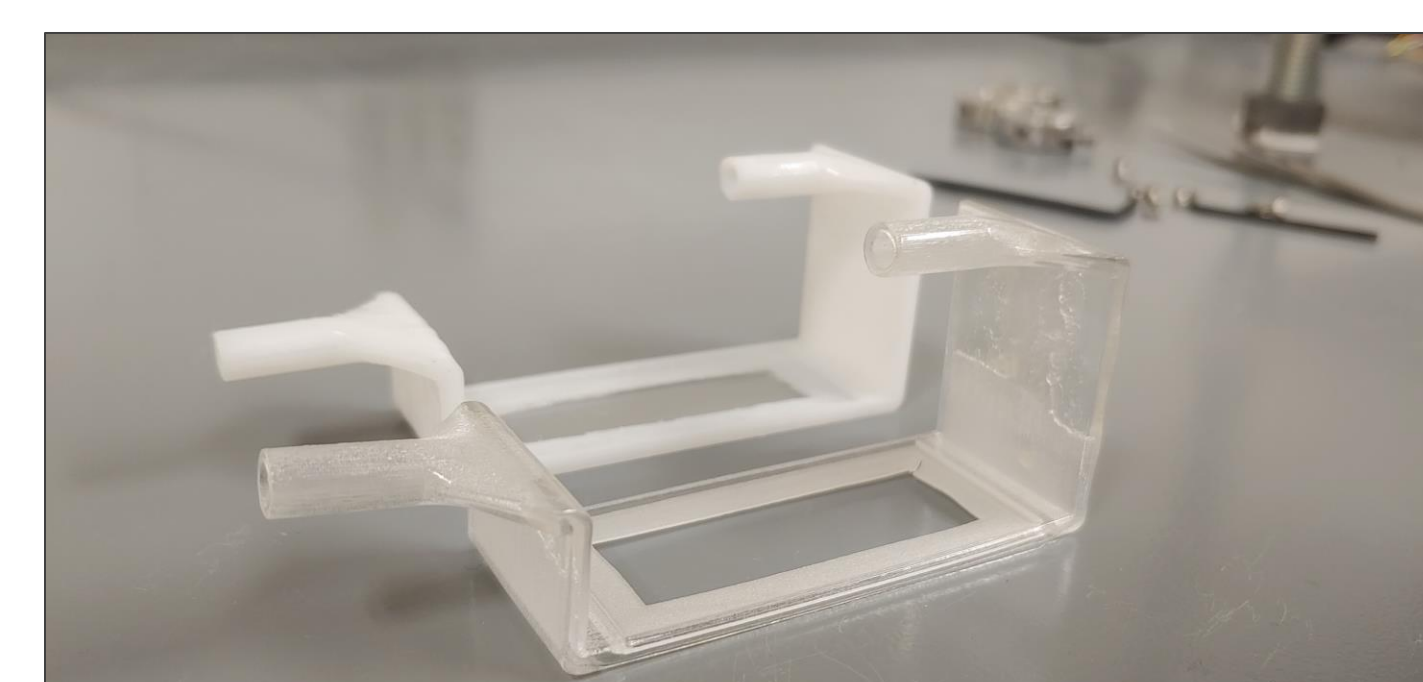
Self-sustaining Bioreactor system designed to apply fluid shear stress to stem cells

Successful Design Criteria Met

- Inexpensive and minimal custom parts
- Able to provide needed shear stresses
- Self-sustaining and semi-independent
- Allows for good visualization of cells which is important for cell culture and imaging during experiments
- System meets size restrictions
- Cell collection possible through breakage of glass cover slip



3D printed Flow Inserts in 4 well culture plate



Flow Inserts, Resin print (front) vs PLA standard print (back)

Known Issues

- Large cell media usage and subsequent treatment volumes leads to high expensive running the system
- Current 3D printers cannot print at the resolution needed to prevent leaks. The high-res *Stratasys Objet J850 Pro* was tested; however, geometry was deformed in post-print recovery.
- Is a one-time use system.

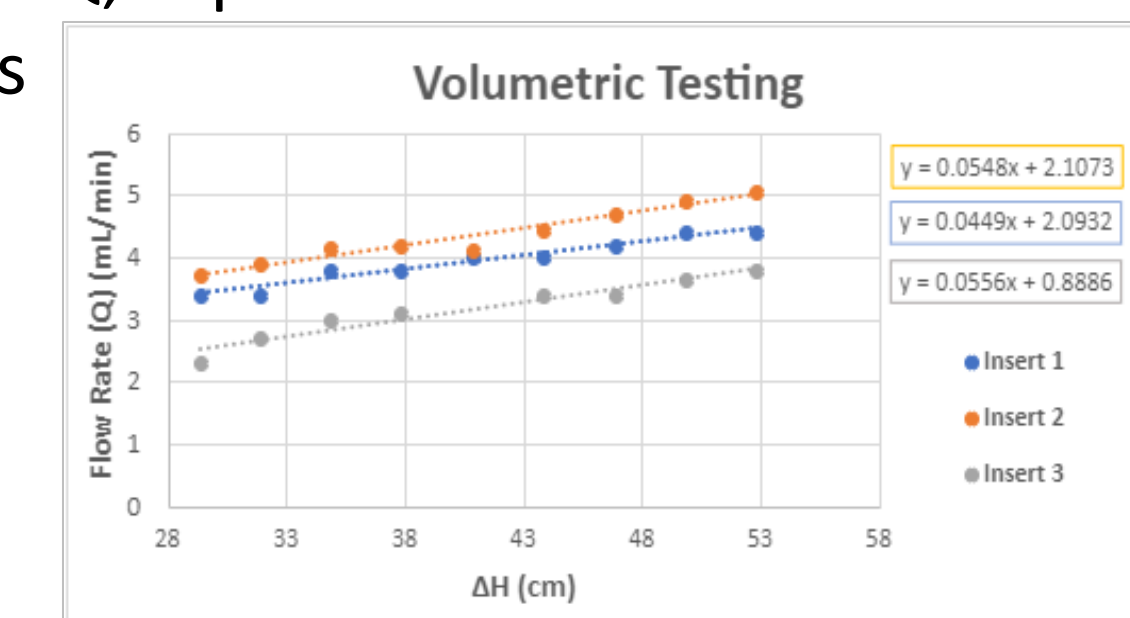
Design Validation

Mathematical Modeling

Using the desired shear stress, we were able to mathematically determine the necessary flow rate Q , expected velocities and our required head including losses

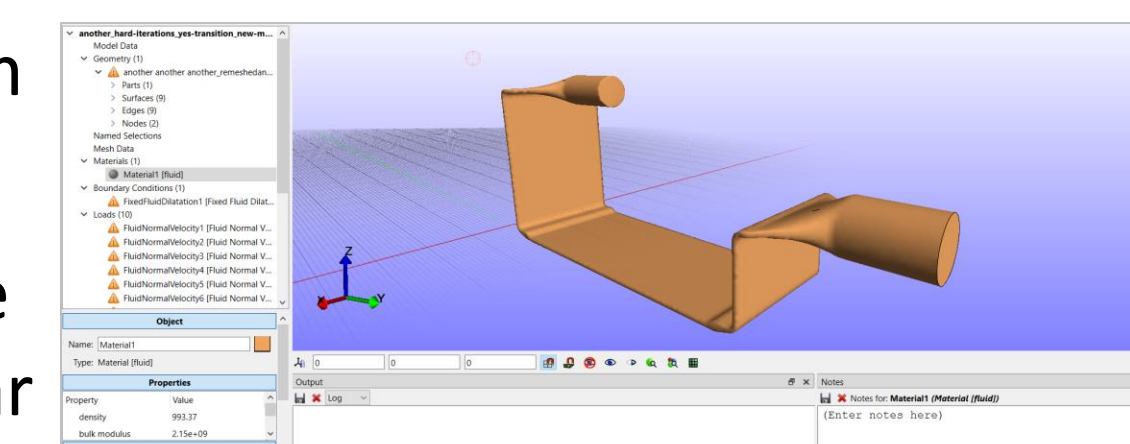
Volumetric Testing

Compares the expected inverse relationship between height of pressure head and flow rate



FE BIO CFD Modeling

Computational fluid dynamics to model the fluid for clearer visualization of shear forces and velocities in the system



Dye Test

Visual Examination to confirm the flow through the system is laminar

Conclusion

Reflection:

The design of our shear stress bioreactor is able to deliver shear stresses from 0 – 20 mPa. Adjusting the height of the upper reservoir changes the shear stress applied to the stem cells. This design is inexpensive and requires only one custom part which allows it to be utilized by smaller labs like Dr. Schiele's tendon tissue engineering lab which was one of the main goals of this project.

Future work:

In the future, engineers should work to downscale the system, reducing the overall volume of media needed. More designing is needed to create an option for pulsatile and intermittent flow. This could be accomplished with an automated mechanical action that adjusts the height of the upper reservoir.

Acknowledgements

This project was funded by Dr. Nathan Schiele and his tendon tissue engineering lab at the University of Idaho. The mentor for our project was Dr. Russell Qualls and Colin Marchus was our graduate mentor.