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Low-Cost
Controllable
Hypoxia Chamber
for Exploring Stem
Cell Behavior

Objective & Motivation

Design and develop a low-cost hypoxia chamber for stem cell research.

- Stem cells naturally grow in a low-oxygen environment
- Simulating this environment during experiments allows:
 - Efficient cultivation of stem cells
 - Research regarding the effect of low-oxygen environments on the cells' development



Value Proposition

- A hypoxic environment allows for more efficient cultivation of stem cells
- Current commercially available hypoxia chambers are expensive
- The aim is to develop a simple, low-cost hypoxia chamber for stem cell research
- The chamber design and manufacturing plan will be included in a paper which will be published in an open access journal
- This will allow other researchers to utilize our technology and ideas

Product Requirements

Functionality

Maintain oxygen levels between 1% and 21%
Maintain CO₂ levels at 5%

Durability

Maintain airtight hypoxic environment for 21 days
Work for multiple experiments

Working Environment

Function in temperature and humidity conditions in incubators

Logistics & Construction

Have a simple user interface
Simple and reproducible construction process
Low-cost

Physical Properties

Large enough to fit at least 4 well plates
Transparent
Fit inside incubator

Risk Management



Pressure buildup



Contamination



Gas leaks



Manufacturing risks



Response time



Sensor accuracy

Control System



Wiring

Exhaust Tubing

User Interface

Solenoids

Input tubing

Incubator

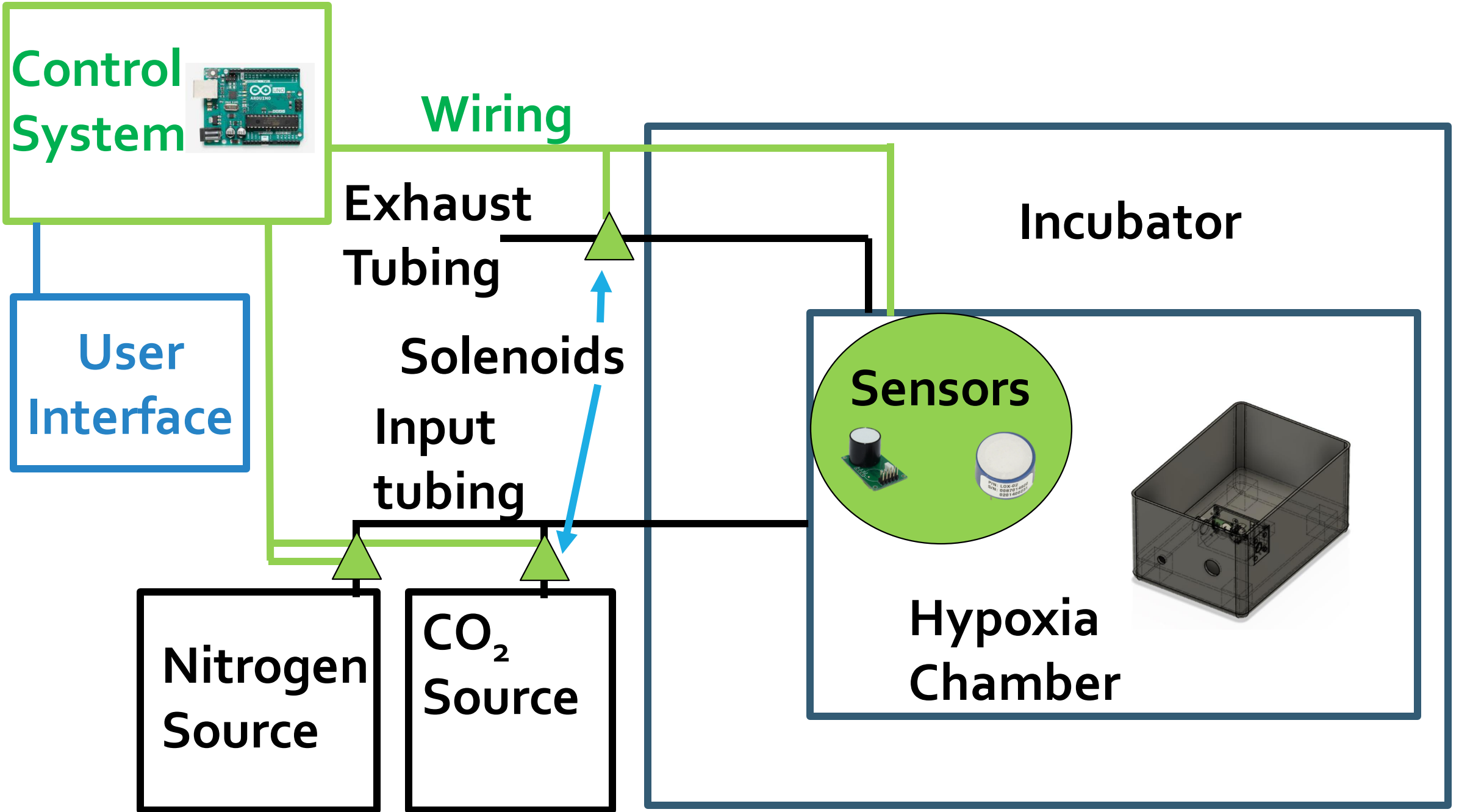
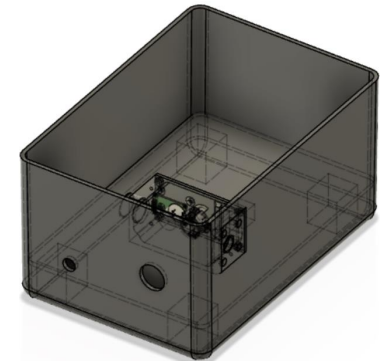
Sensors



Nitrogen Source

CO₂ Source

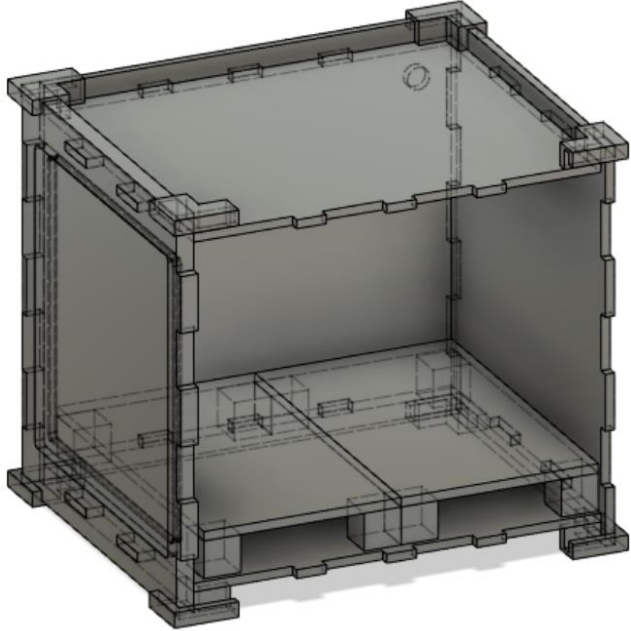
Hypoxia Chamber





Chamber Design Solutions

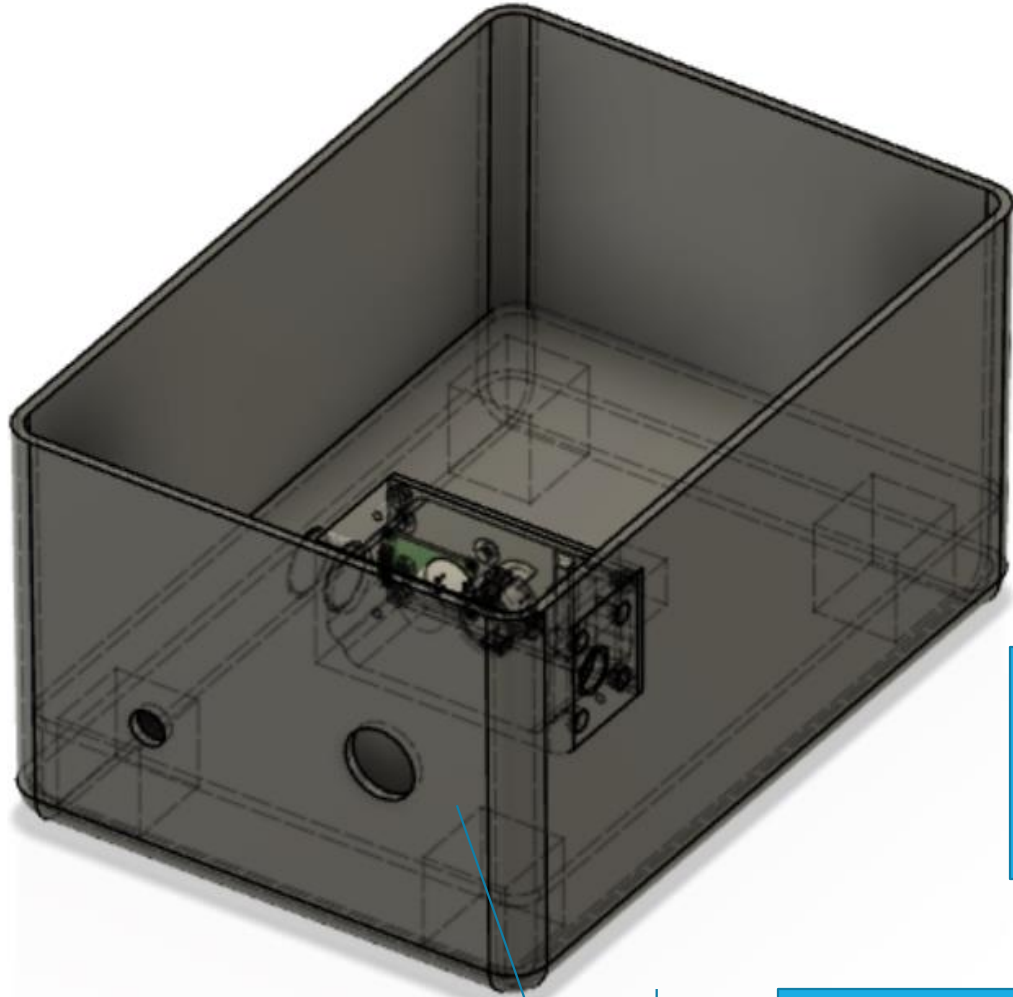
Custom Chamber Prototype 1



- Acrylic piece was laser cut
- Ethanol, the solvent to be used for sterilization, was used to clean the piece post-cutting
- We discovered that ethanol causes crazing within the acrylic, which affects the structural integrity



Final Chamber Design



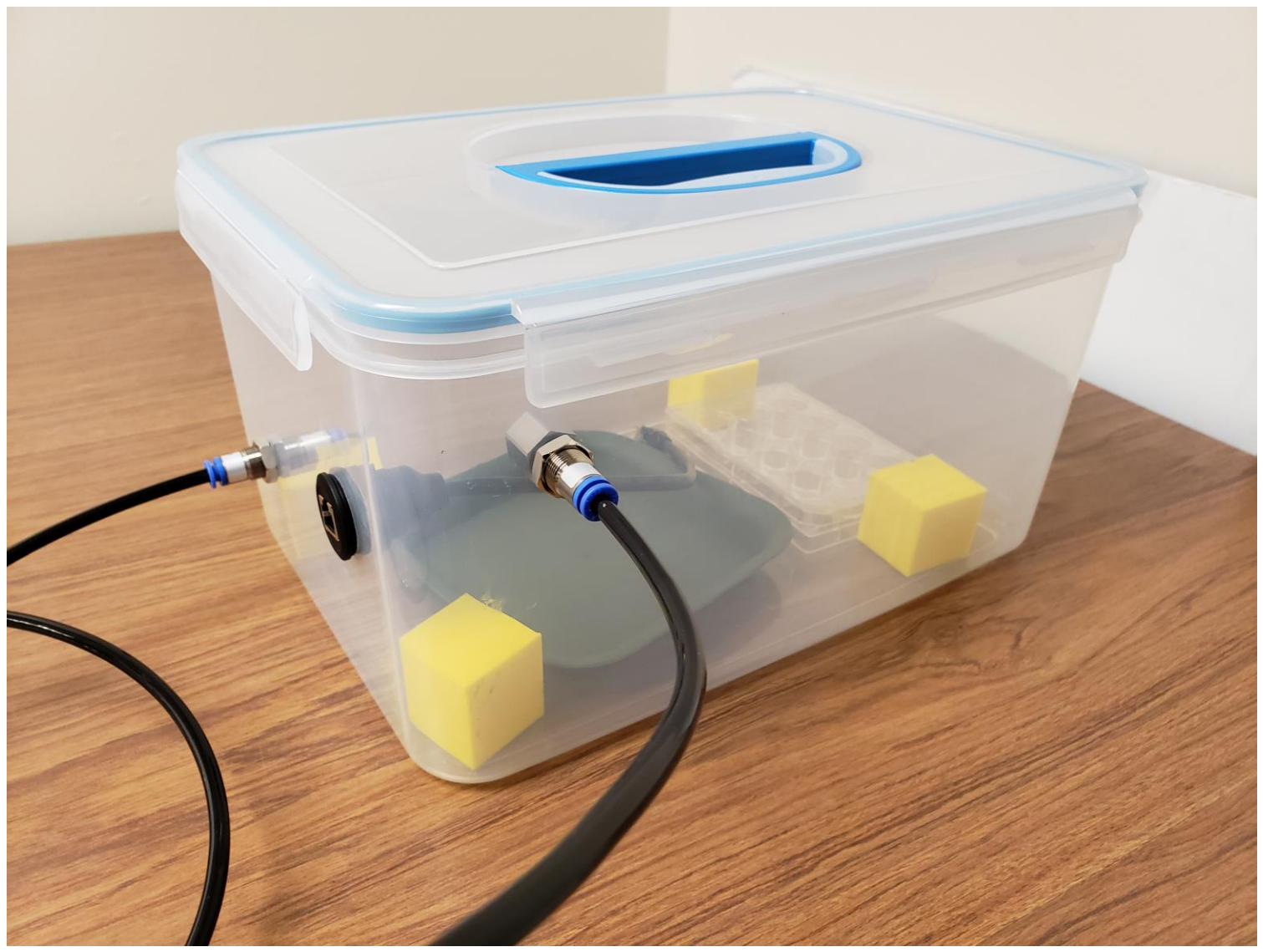
Handle and
door seal



Guaranteed
airtight

Ample space for several well plates,
humidity control system, and sensors

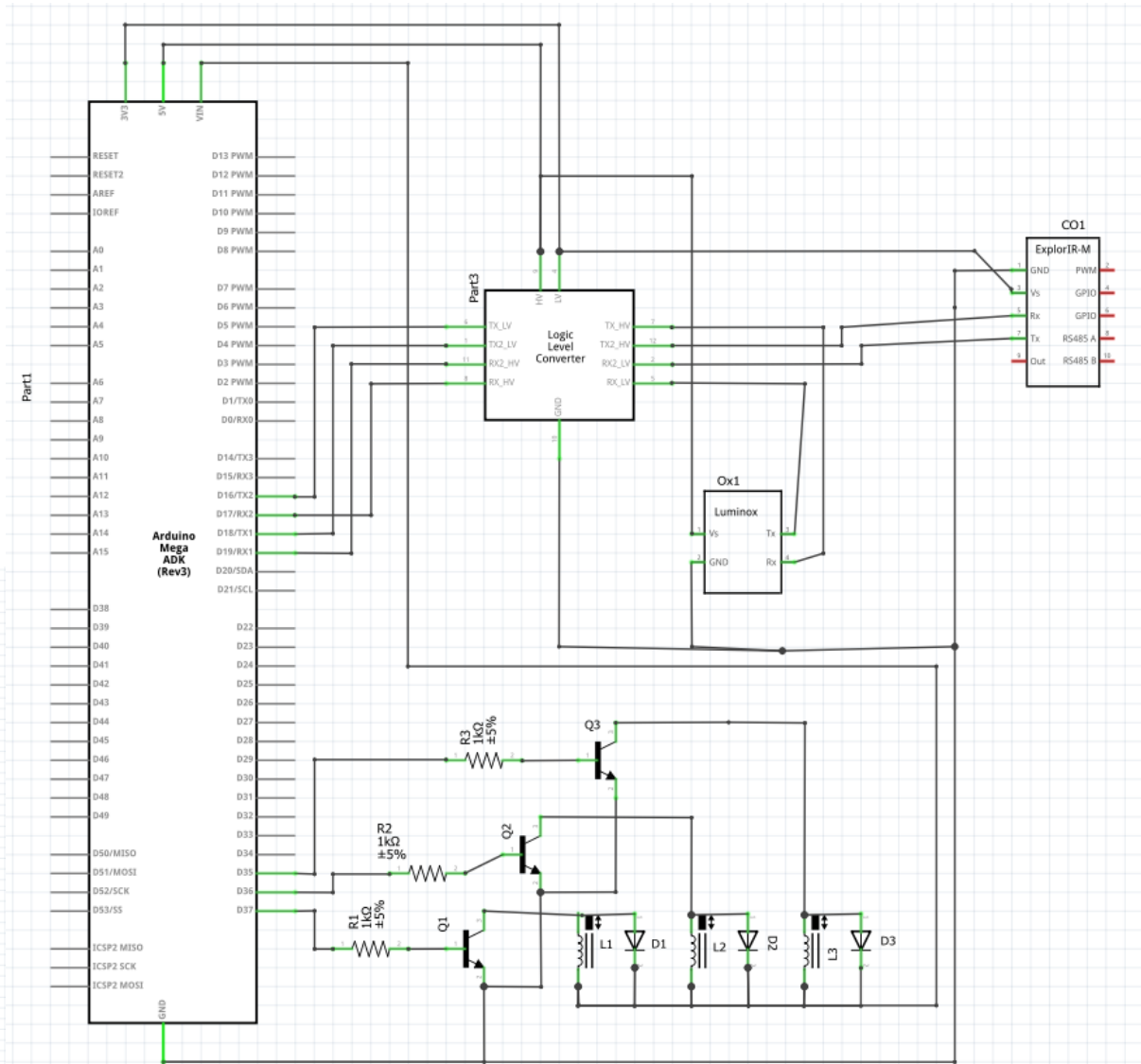
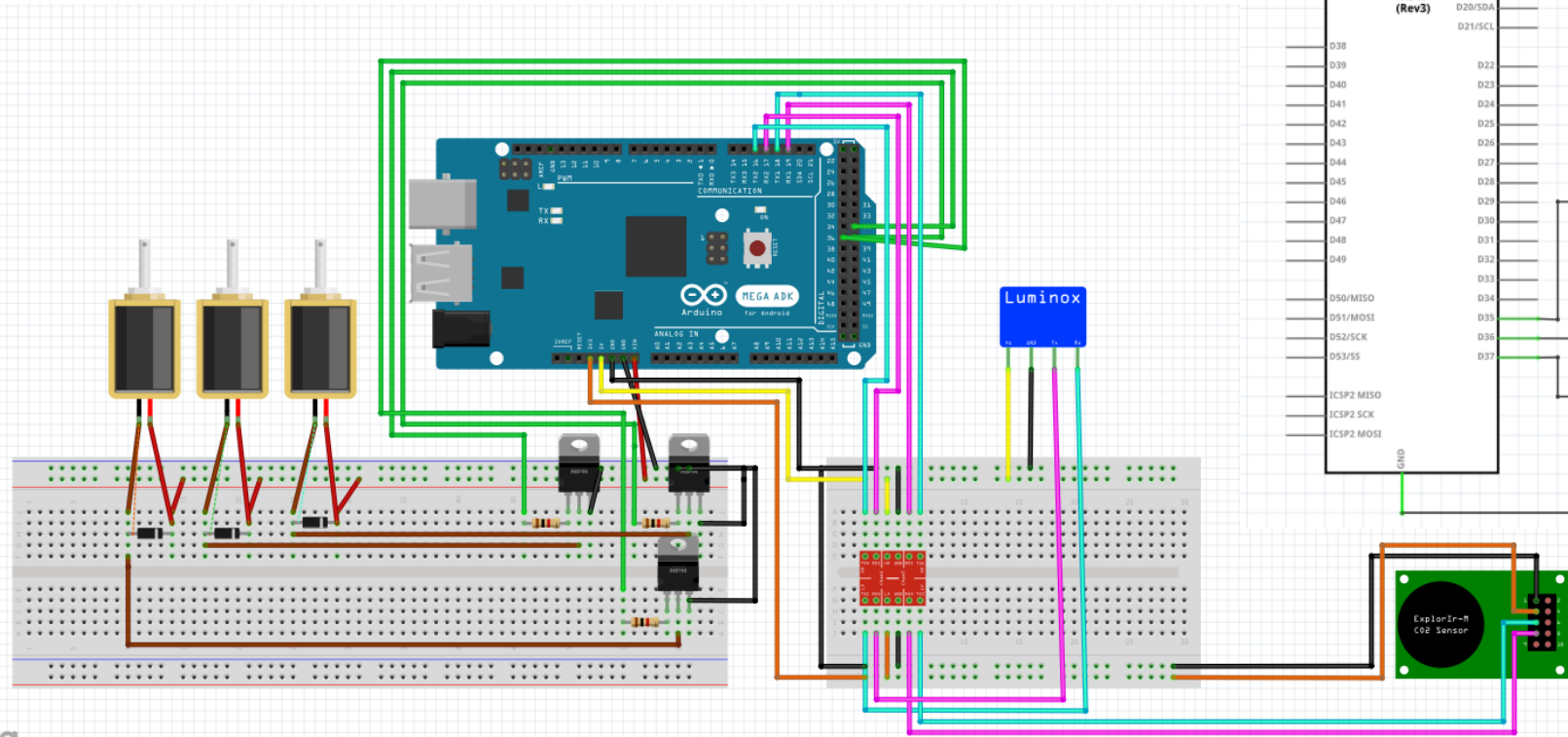






Control System

- Arduino Mega with multiple serial ports
- 3 Solenoids to control airflow, with diodes, resistors, and BJTs
- O₂ and CO₂ sensors connected through level converter



Sensors



O₂ Sensor

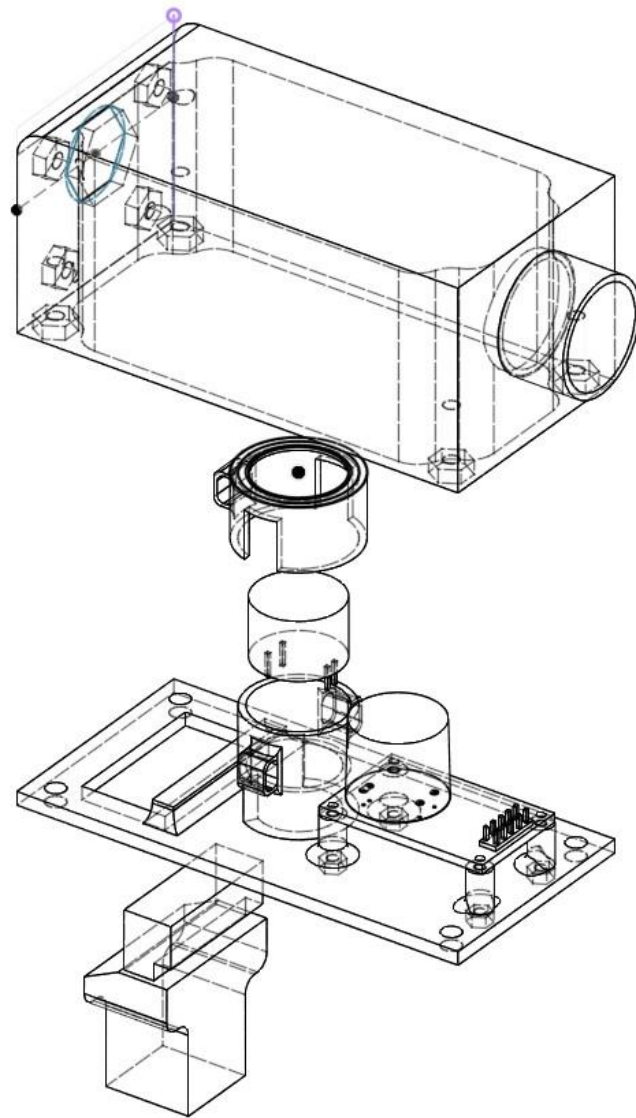
- Measurement Range: 0 to 25% O₂
- Response Time: <60s
- Resolution: 0.01%
- Lifetime: >5 years
- Voltage: 4.5-5.5VDC
- Interface: UART



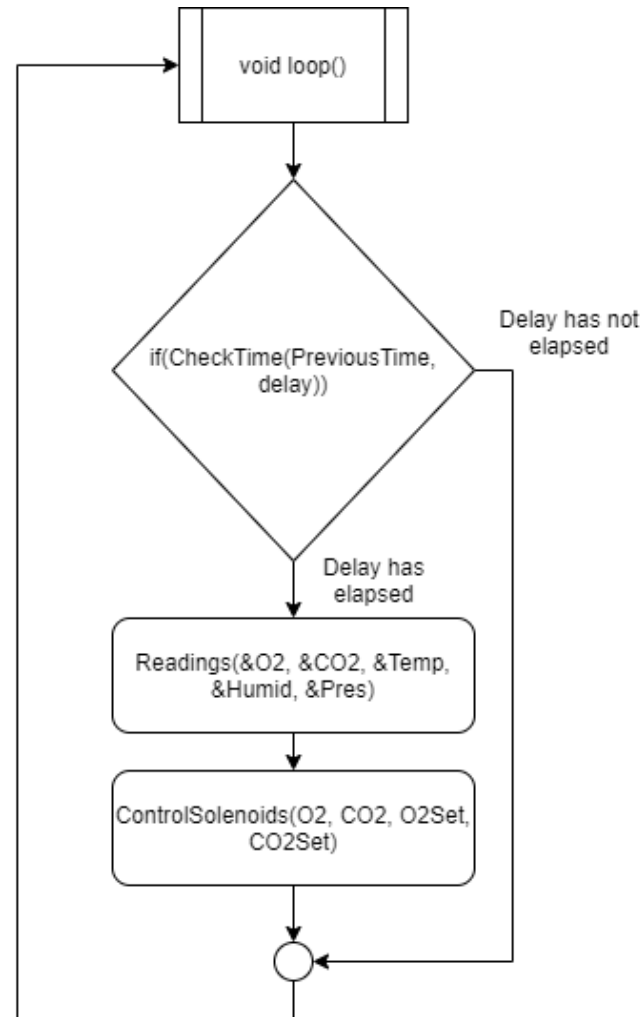
CO₂, Temp, Pressure, Humidity Sensor

- Measurement Range: 0 to 20% CO₂
- Response Time: 10s to 3min (Configurable)
- Resolution: 10ppm
- Lifetime: >15 years
- Voltage: 3.3v
- Interface: UART

Sensor Housing



Control Algorithm



CheckTime()

Utilizes "millis()" function to delay a set amount of time from the last saved reading

Readings()

Receives and Parses readings from both serial sensors

ControlSolenoids() Math

- O2: % Value read
- set: Desired O2 setpoint
- x: Time to open the solenoid

$$\frac{25 - O2}{O2 - set} = \frac{100 - x}{x - 10}$$

$$x = \frac{100 * (O2 - set) + 10 * (25 - O2)}{(O2 - set) + (25 - O2)}$$

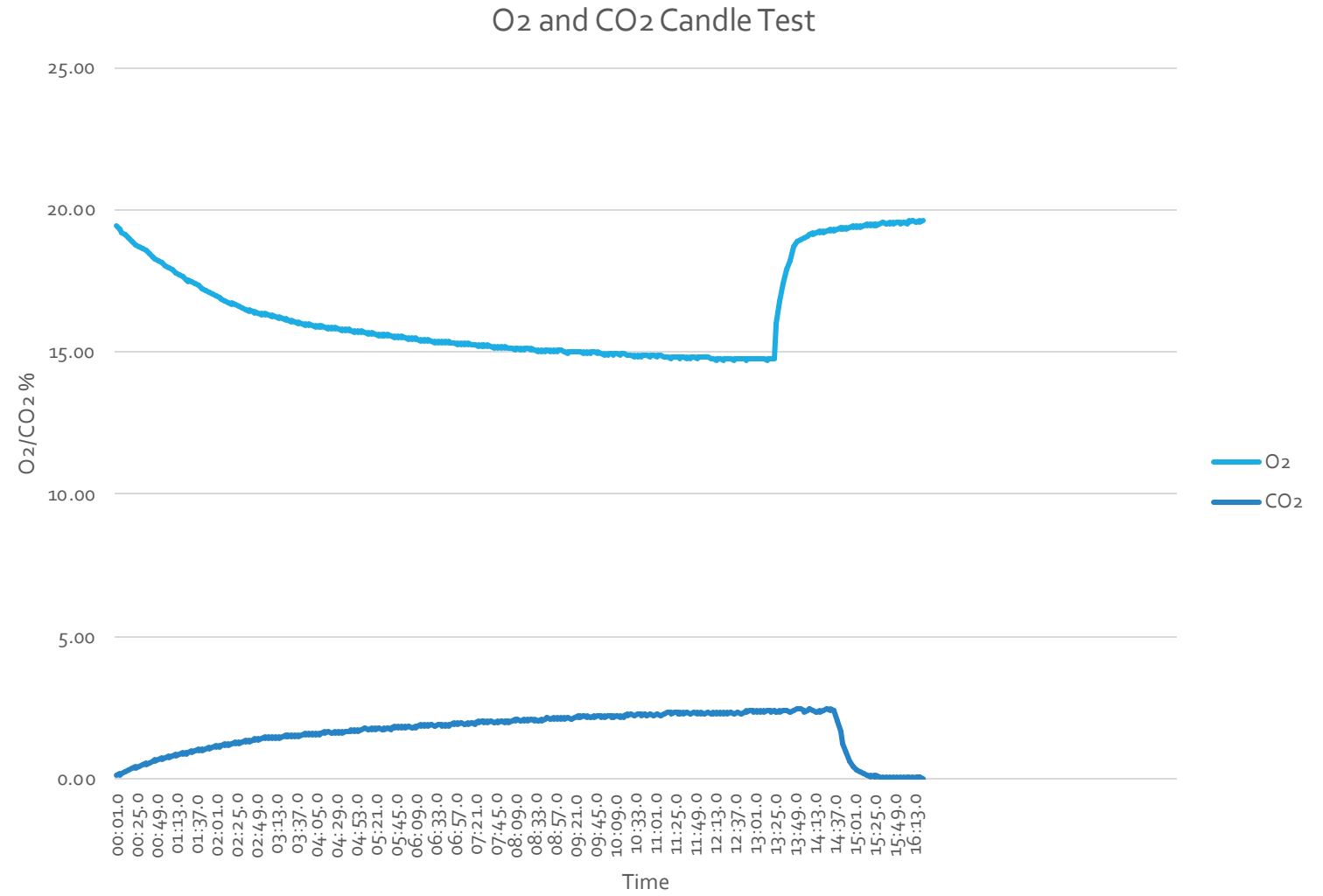


Design Validation and Testing

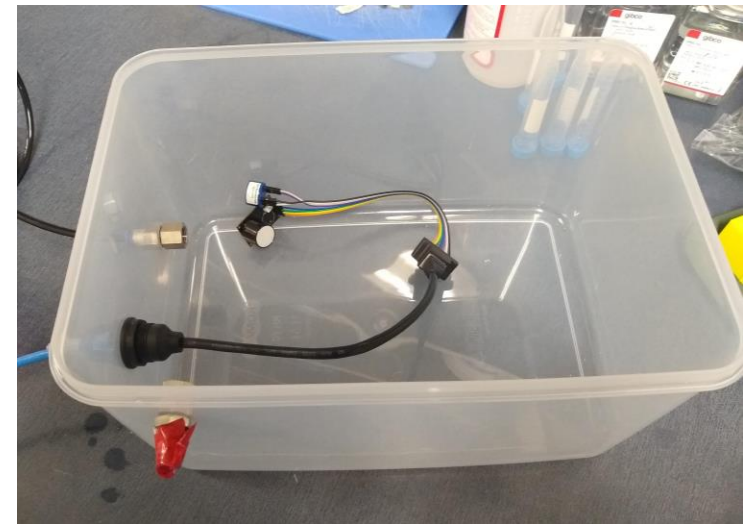
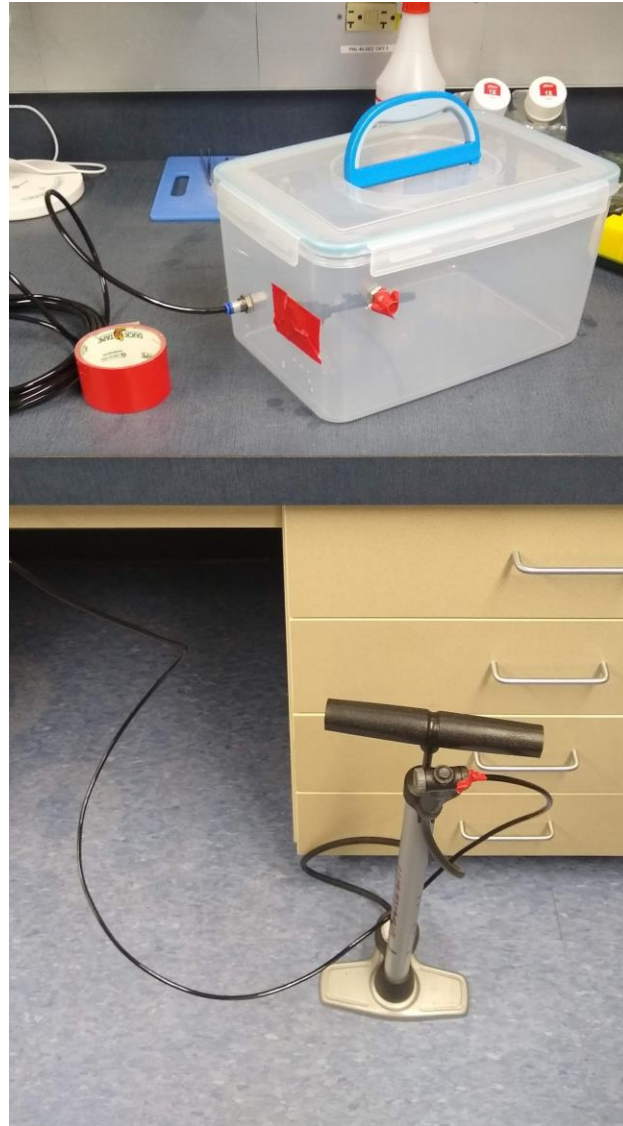
Design Validation

Requirement	Testing
Maintain a hypoxic environment (1% O₂ and 5% CO₂) for at least 21 days	<ul style="list-style-type: none">• Run control system to pump the correct % of gas in to create hypoxic environment• Use sensors for daily measurements for 5 days• Run 21-day test to confirm durability
Airtight/Pressure Test	<ul style="list-style-type: none">• Slightly pressurize chamber and use soapy water to check for bubbles on all edges
Reach hypoxic conditions within 15 minutes	<ul style="list-style-type: none">• Create a setpoint on the GUI and check if it reaches that level within 15 minutes.
Product must be durable to sterilization procedures	<ul style="list-style-type: none">• Review the properties of the components used
Sensors read accurate data to the Arduino	<ul style="list-style-type: none">• Connect the O₂ and CO₂ sensors to the Arduino and try and get sensors values
Disable functionality when door is opened	<ul style="list-style-type: none">• Create a switch on the gui to alert when the door is open and disable input of gas
Achieve 95% humidity within the chamber	<ul style="list-style-type: none">• Place a well of water or saturated sponge in the chamber and measure humidity over a day period

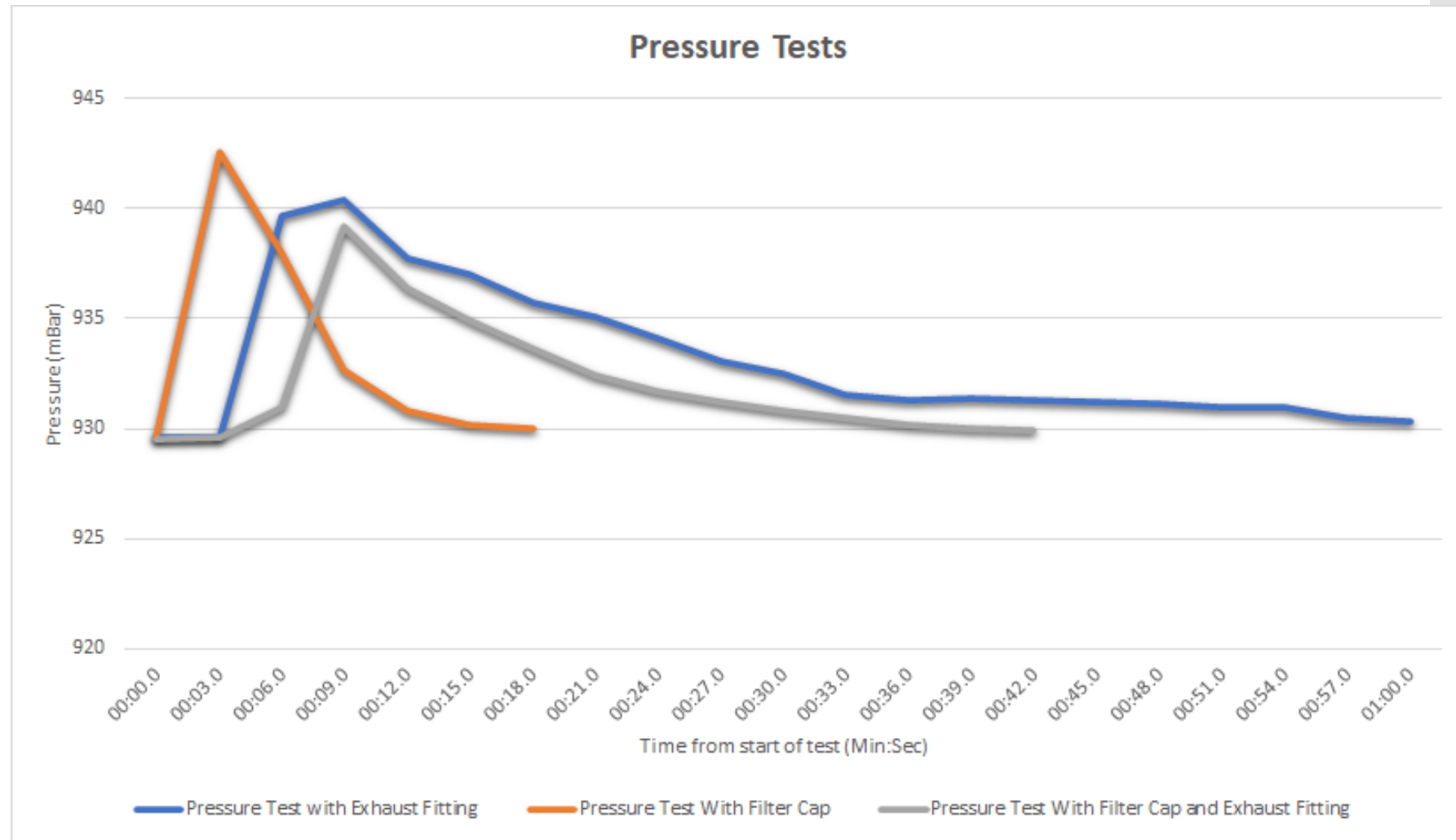
Sensor Testing



First Pressure Test



Results

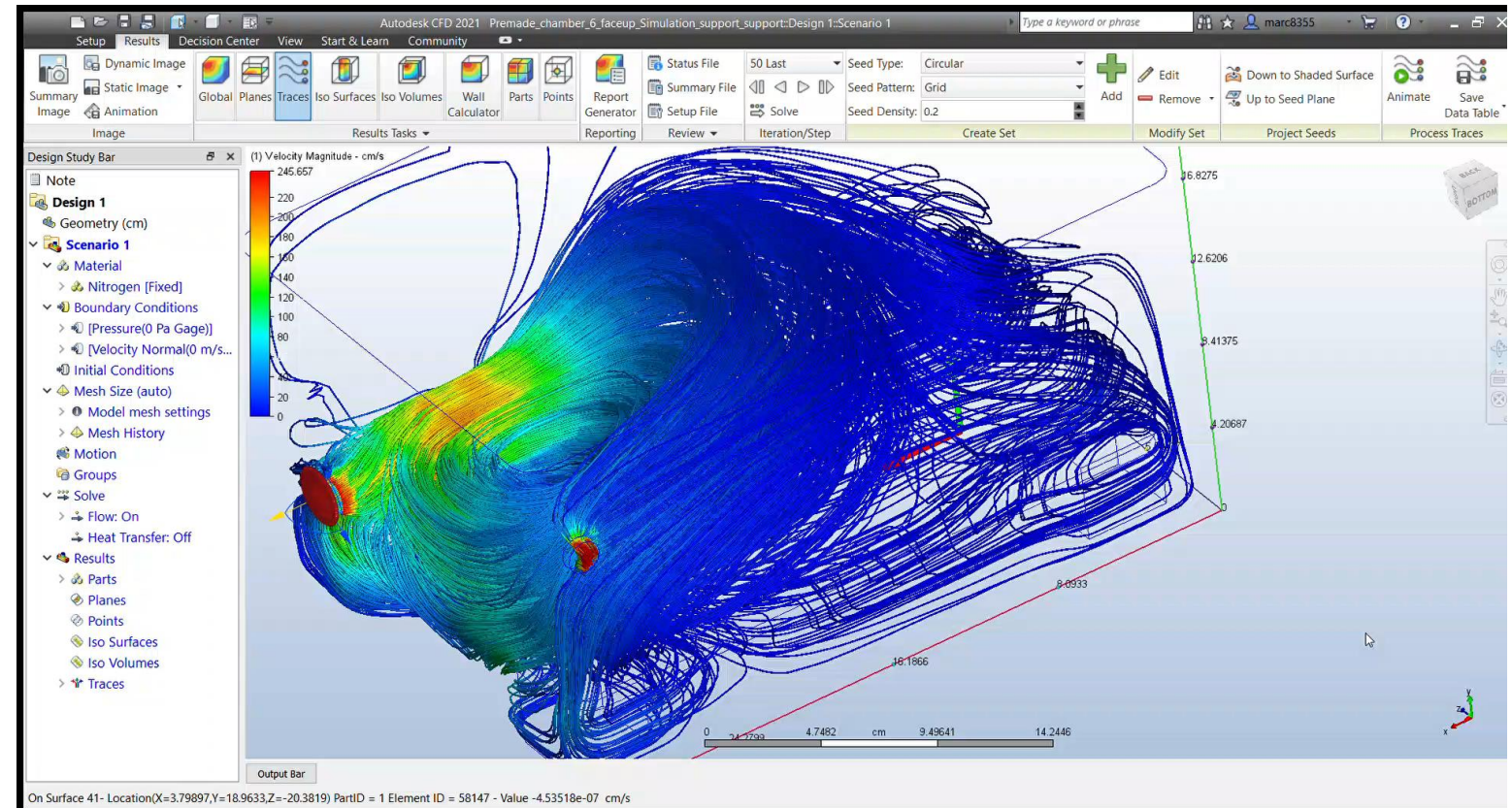


18 seconds

42 seconds

60 seconds

Fluid Simulation Validation



Budget

We are working with a budget of \$1000

Expense Items	Price	Expense Items	Price
Arduino Mega Equivalent	\$ 15.86	Epoxy	\$ 12.15
Oxygen Sensor	\$ 99.00	Tubing	\$ 10.00
O2 adapter board	\$ 7.94	3D -printing filament	\$ 25.50
Breadboard	\$ 8.59	Pre-made Container	\$ 21.99
Wires	\$ -	Logic converter	\$ 7.29
Input Solenoids x2	\$ 26.00	Power Supply	\$ 12.50
Exhaust Solenoid	\$ 13.00	O-rings	\$ -
Nitrogen	\$ 10.00	Through wall ethernet cable	\$ 10.05
CO2 Sensor	\$ 180	Ethernet adaptor	\$ 9.59
Through wall Tubing connector x2	\$ 15.30	Water Trays	\$ 10.00
Transistors	\$ 9.99	Misc.	\$ 100.00
Acrylic	\$ 19.98		
O-ring	\$ 7.29	Total:	\$ 644.95

Looking Forward

	Sept	Oct	Nov	Dec	Jan	Feb	March	April
Concept design & Brainstorming	Yellow							
Modeling, Design, & Prototype Development		Yellow						
Prototype Development, Manufacturing, and Modifications			Yellow	Yellow	Yellow	Green		
Product Testing & Modifications						Green	Green	
Final Product Preparation							Green	Green

Immediate Tasks:

- Finish constructing and testing the control system
- Continue product testing