Herbert Wertheim College of Engineering
UNIVERSITY of FLORIDA

CAMbox

Compact Autonomous Microbioreactor

Section 22695, Group 14

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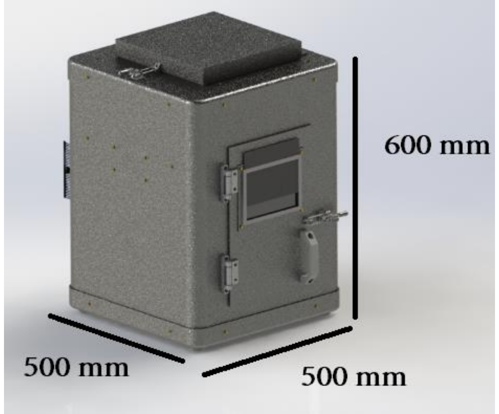
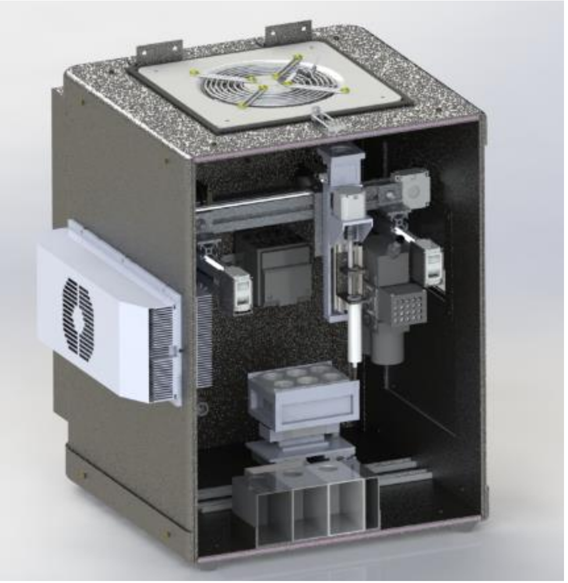
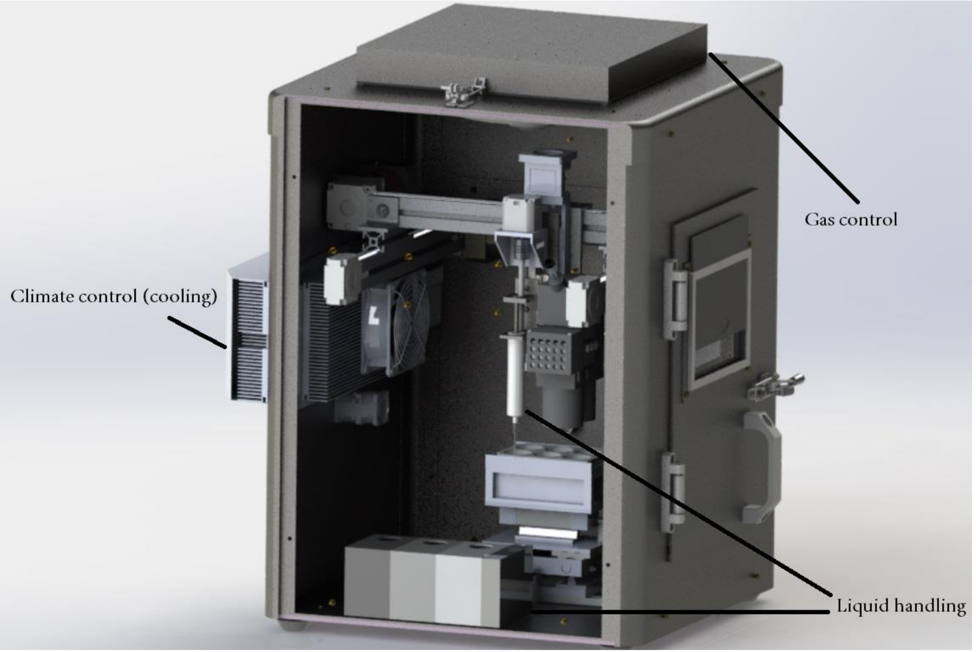
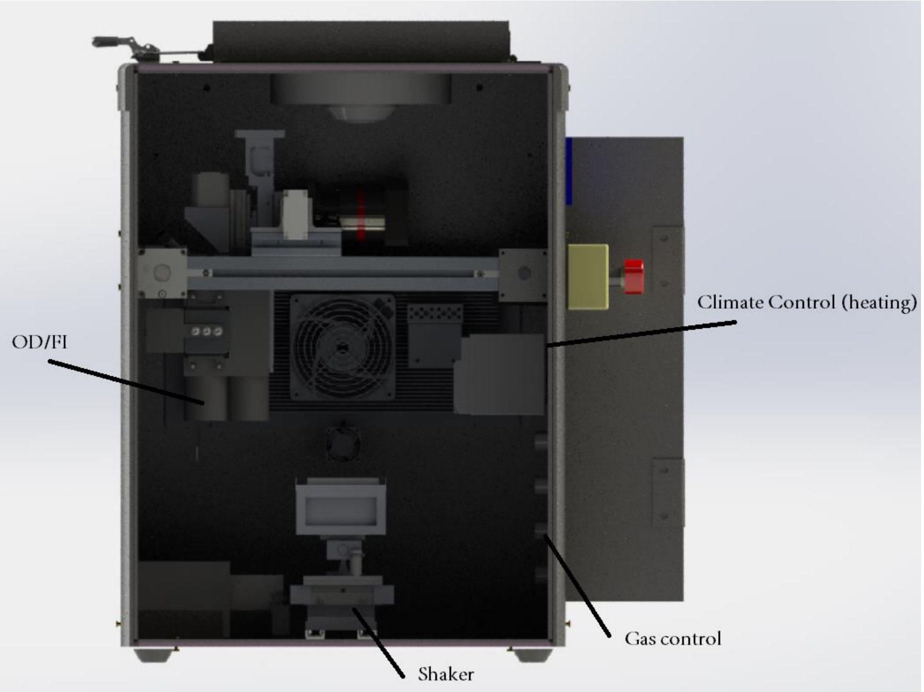
POWERING THE NEW ENGINEER TO TRANSFORM THE FUTURE

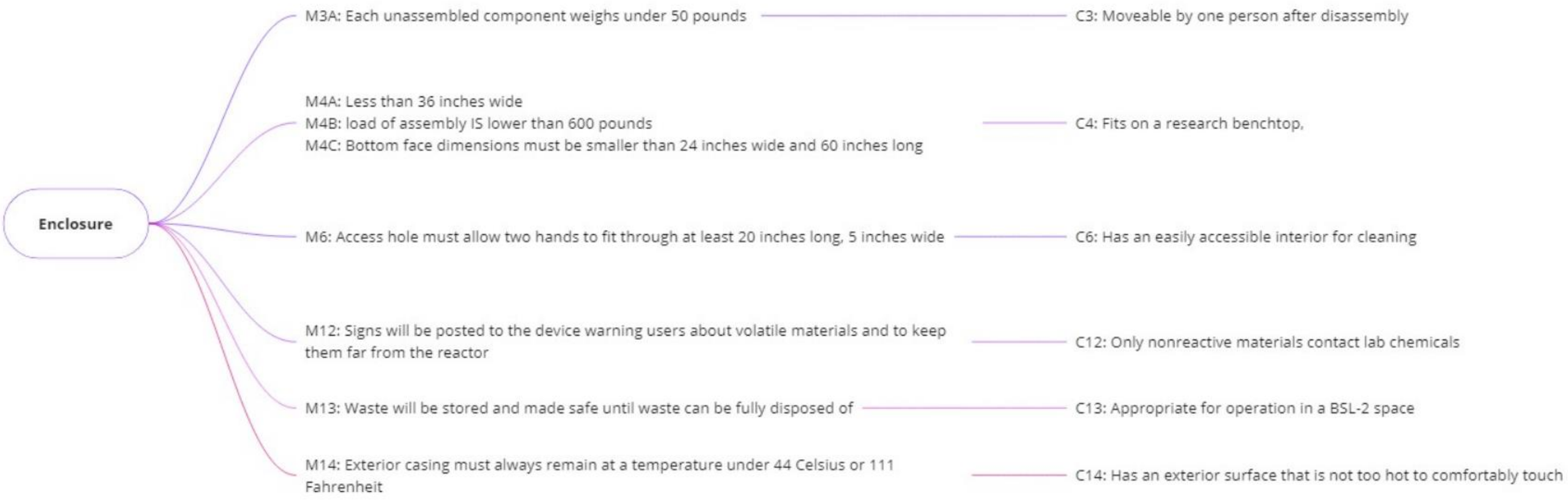
- **Compact:** Smaller footprint allows for easier placement.
- **Lower cost:** Fewer materials used enabled use of higher quality OTS parts.
- **Marketable:** Compact, cost efficient, design makes it available to a larger audience.





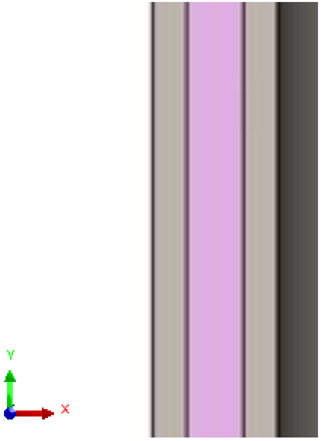

Gas control
Climate control (cooling)
Liquid handling

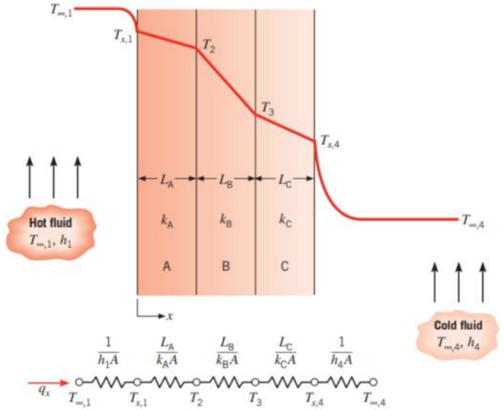

Climate Control (heating)
OD/FI
Gas control
Shaker


Enclosure
M3A: Each unassembled component weighs under 50 pounds
C3: Moveable by one person after disassembly
M4A: Less than 36 inches wide
M4B: load of assembly IS lower than 600 pounds
M4C: Bottom face dimensions must be smaller than 24 inches wide and 60 inches long
C4: Fits on a research benchtop,
M6: Access hole must allow two hands to fit through at least 20 inches long, 5 inches wide
C6: Has an easily accessible interior for cleaning
M12: Signs will be posted to the device warning users about volatile materials and to keep them far from the reactor
C12: Only nonreactive materials contact lab chemicals
M13: Waste will be stored and made safe until waste can be fully disposed of
C13: Appropriate for operation in a BSL-2 space
M14: Exterior casing must always remain at a temperature under 44 Celsius or 111 Fahrenheit
C14: Has an exterior surface that is not too hot to comfortably touch

Enclosure

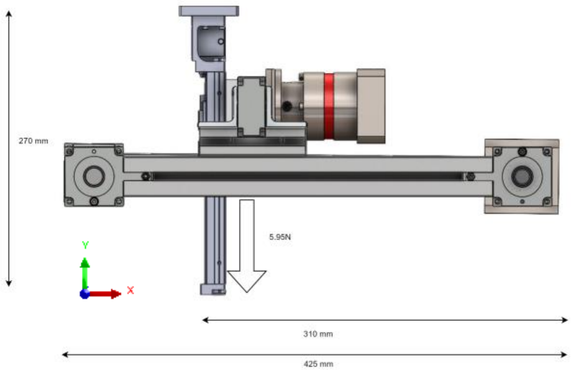
- **Material Selection**
  - Constructed from ASTM A36 Steel
  - Layer of Polyurethane Foam Rigid
    - Insulating R-value ≈ 5
    - Maintains internal temperatures

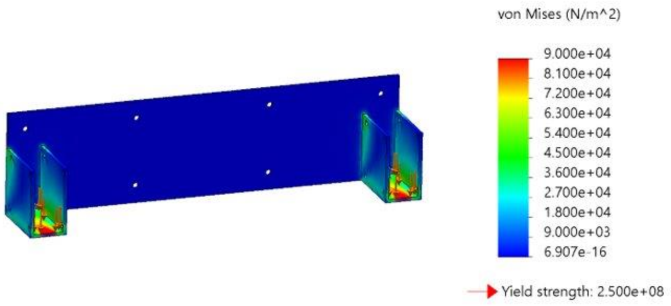


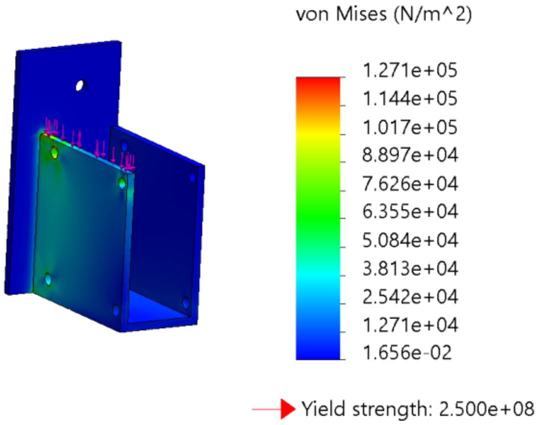


Enclosure

- Material Analysis
  - ASTM A36 Steel has a yield strength of 2.5e8 Pa
  - Maximum stress stress from gantry on supports ≈ 1.271e5 Pa



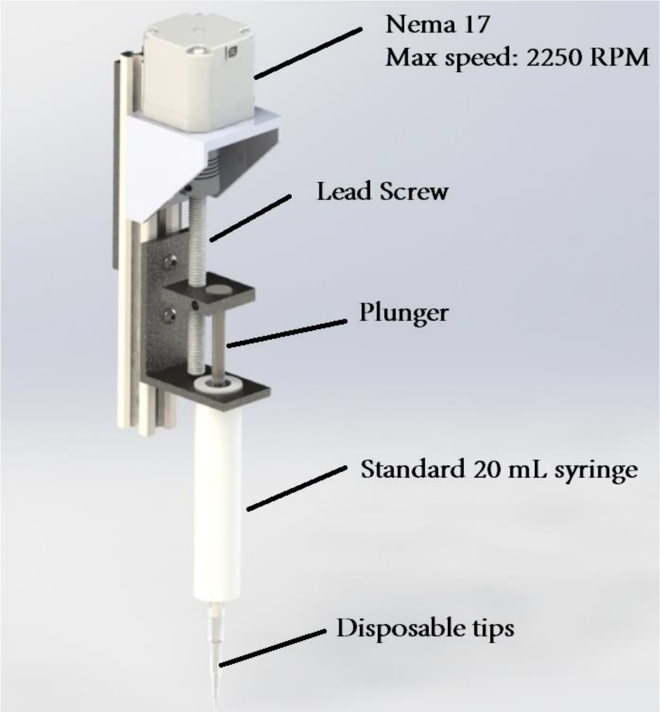


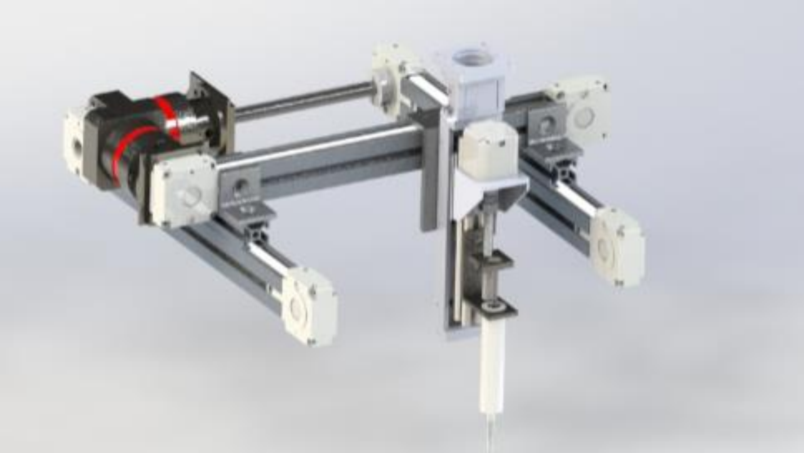


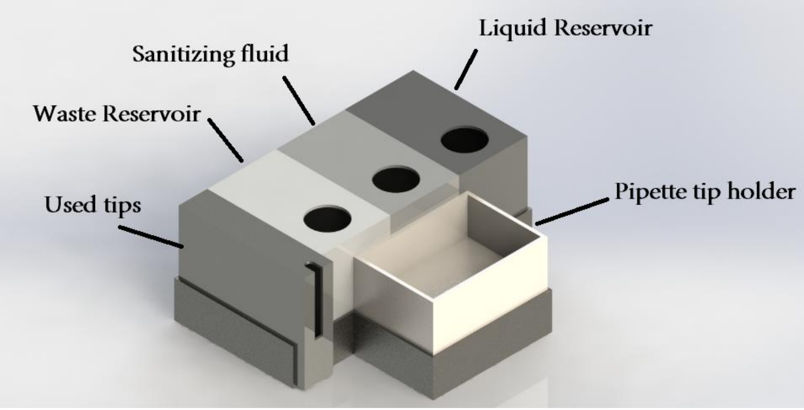
Liquid Handling

- M11: Disposable tips will be made of nonporous materials. 1 disposable tip per liquid being used. — C11: Only nonporous materials contact cell cultures
- M15: Waste will be neutralized by being placed in a reservoir containing 0.10 of the volume in bleach. — C15: . Is capable of sequestering and neutralizing its own liquid and solid waste
- M34: Quantified by the number of steps the subsystems require to complete the process of adding or subtracting liquid. The number of steps this subsystem takes will be between 3 and 6. — C34: Is capable of automated liquid handling with fluid addition/subtraction from each well or tube
- M35: To determine whether aerosols will be created the Reynolds number will be calculated. The Reynolds number will have a maximum of 2300 in order to avoid creating aerosols — C35: Dispenses fluid without creating aerosols
- M36: Dispenses fluid from 225 uL/s to 300 uL/s — C36: Achieves dispense rates from 225 uL/s to 300 uL/s
- M37: Deposit from 5-20000 uL of aliquot fluid volume — C37: Deposits a minimum/maximum aliquot fluid volume from 5–20,000 μL
- M38: Volume accuracy 0.01 uL — C38: Achieves dispensing volume accuracy of ±0.1 μL
- M39: Volume accuracy 0.01 uL — C39: Achieves dispensing volume precision ±0.01 μL
- M40: Cross contamination will be avoided by using disposable tips. The number of tips used will depend for the number of pipettes, a minimum of 1. — C40: No cross contamination between individual wells/tubes during liquid handling

Liquid Handling







Liquid handling

- Flow rate: 375 μL/s

$$\bar{V} = \frac{2\pi}{60} \times 2250 \times (9.566 \times 10^{-3}) = 2.25\ m/s$$

$$Q = 2.25 \times (1.66 \times 10^{-7}) = \frac{3.75 \times 10^{-7} m^3}{s} = 375\ {}^{\mu L}/_{s}$$

- No Aerosols, confirmed with Reynolds Number:

$$Re = \frac{997 \times 2.25 \times 4.6 \times 10^{-4}}{1.08 \times 10^{-3}} = 957$$

- Dispense accuracy: ±0.03 μL

$$\pm 0.05° \rightarrow 7200\ steps\ per\ revolution$$

$$screw\ lead: 1.5\ mm$$

$$\frac{1.5}{7200} = 0.000208\ mm/step$$

$$Cross\ sectional\ area\ of\ syringe: \frac{\pi(9.57 \times 10^{-3})^2}{2} = 0.00014\ m^2$$

$$0.000208\ \times 0.00014 = \pm 3 \times 10^{-11}\ m^3$$

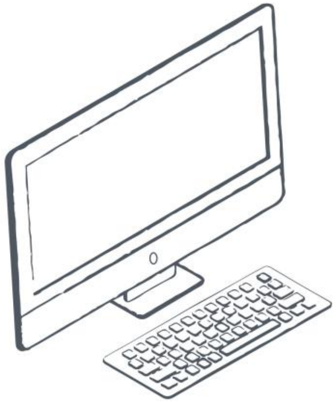
$$\pm 0.03\ \mu L$$

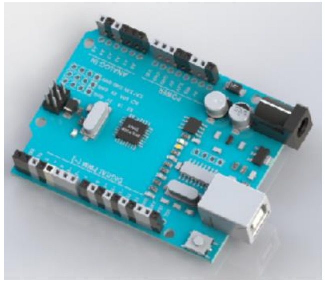
User Interface

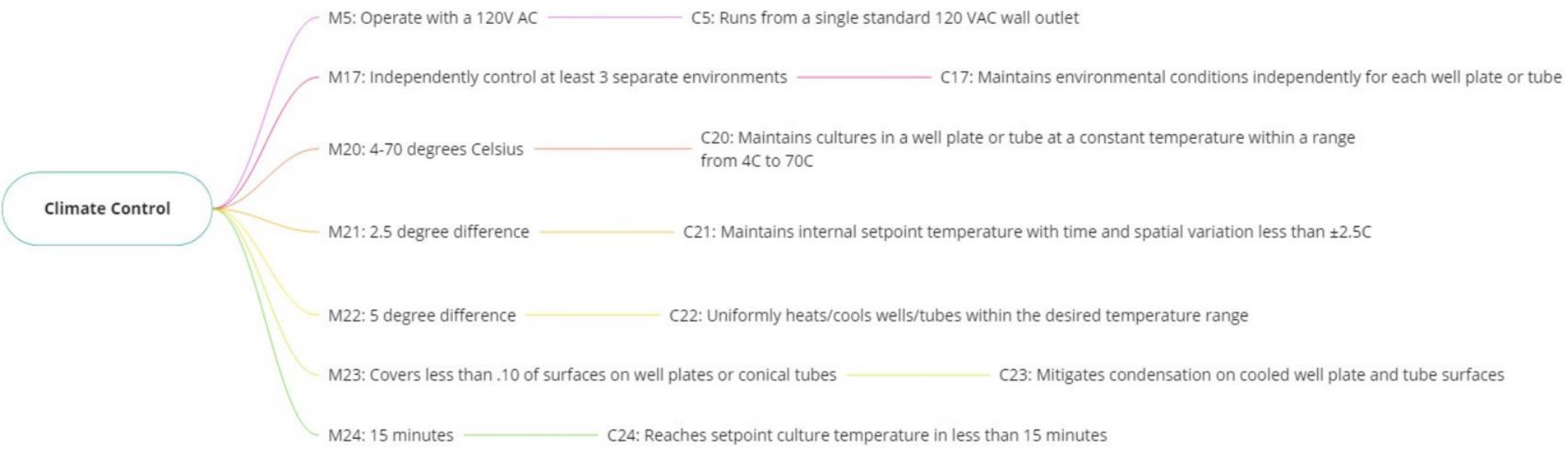
- M7: Quantified by how long to takes to stop all function, a minimum of 0.5 seconds will be used and a maximum of 1 second — C7: Includes an easily actuated emergency shut-off that safely that stops all functions
- M8: There will be a minimum of 1 setting per number of subassemblies of the system and a maximum of 2 settings per number of subassemblies, making the minimum settings on the interface 8 and the maximum 16 — C8: Has an intuitive user interface
- M9: This need will be quantified by the brightness of the interface display or flashing of the buttons on a scale of 1-5, 1 being the dimmest and 5 being the brightest — C9: Has a visual indicator that is easily seen by the user and nearby personnel
- M10: Each subsystem can be controlled individually, quantified by the number of controls on the interface, a minimum of 8 to allow control of different subsystems — C10: Is programmable: control parameters can be changed by the user, and more complex processes can be added

User interface

- Arduino will be inside the control box.
- Device connects to laboratory computer
- Sensors are compatible with Arduino.

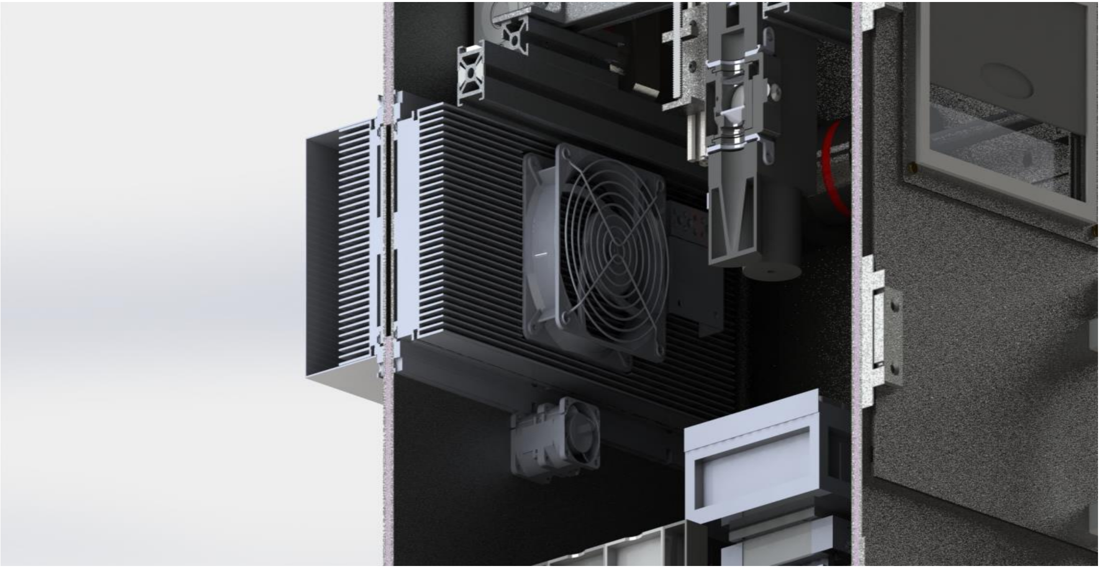





Climate Control
M5: Operate with a 120V AC
C5: Runs from a single standard 120 VAC wall outlet
M17: Independently control at least 3 separate environments
C17: Maintains environmental conditions independently for each well plate or tube
M20: 4-70 degrees Celsius
C20: Maintains cultures in a well plate or tube at a constant temperature within a range from 4C to 70C
M21: 2.5 degree difference
C21: Maintains internal setpoint temperature with time and spatial variation less than ±2.5C
M22: 5 degree difference
C22: Uniformly heats/cools wells/tubes within the desired temperature range
M23: Covers less than .10 of surfaces on well plates or conical tubes
C23: Mitigates condensation on cooled well plate and tube surfaces
M24: 15 minutes
C24: Reaches setpoint culture temperature in less than 15 minutes

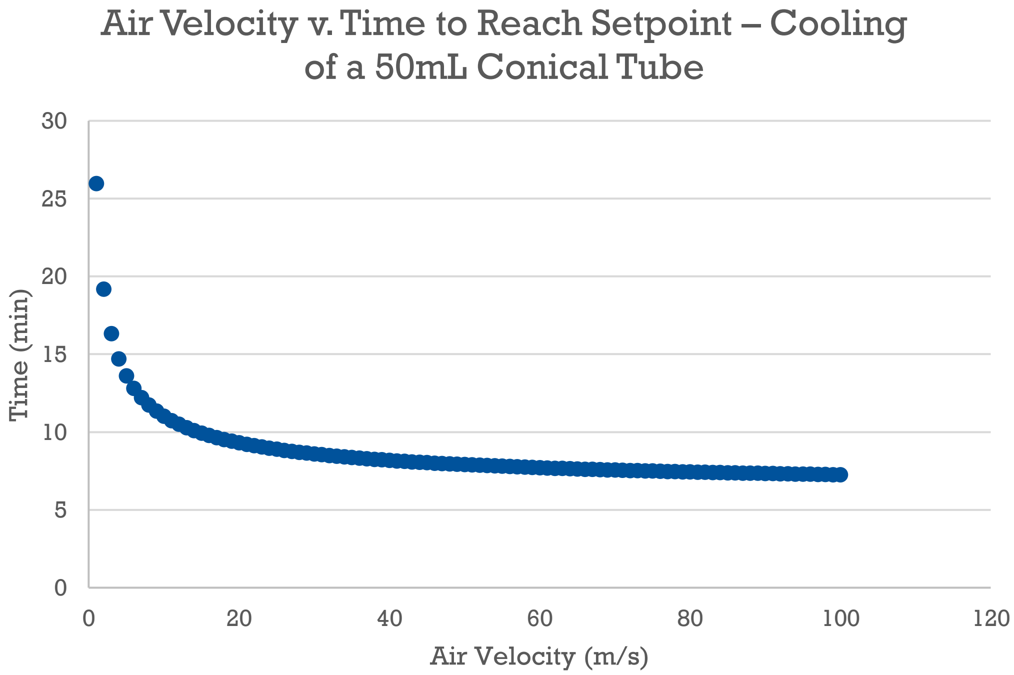
Climate Control - Cooling

- **Seifert 3152303 thermoelectric cooler**
  - More compact
  - Better longevity
  - No moving parts
  - 200W cooling capacity
  - Temperatures as low as -10 C



Climate Control - Cooling

- Compact enclosure enables rapid heating & cooling.
- Variable-speed fan offers greater user control.
- 20C to 4C in under 12 minutes.



Climate Control – Circulation

- System utilizes forced convection.
- Variable-speed Supermicro fan.
- 25.2 CFM with an outlet of 1.57in x 1.57in.

Maximum output air speed calculations:

Outlet area:

$$1.57[\text{in}] * 1.57[\text{in}] * \left(\frac{2.54[\text{cm}]}{1[\text{in}]}\right)^2 * \left(\frac{1[\text{m}]}{100[\text{cm}]}\right)^2 = 1.5903 * 10^{-3}[\text{m}^2]$$

Volumetric flow rate conversion:

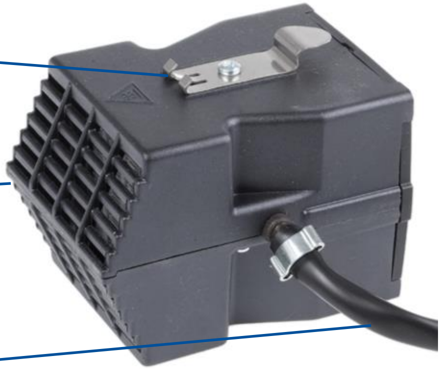
$$25.2\left[\frac{\text{ft}^3}{\text{min}}\right] * \left(\frac{1}{3.28}\right)^3 \left[\frac{\text{m}^3}{\text{ft}^3}\right] * \frac{1}{60}\left[\frac{\text{min}}{\text{s}}\right] = 0.0119\left[\frac{\text{m}^3}{s}\right]$$

Maximum output air speed:

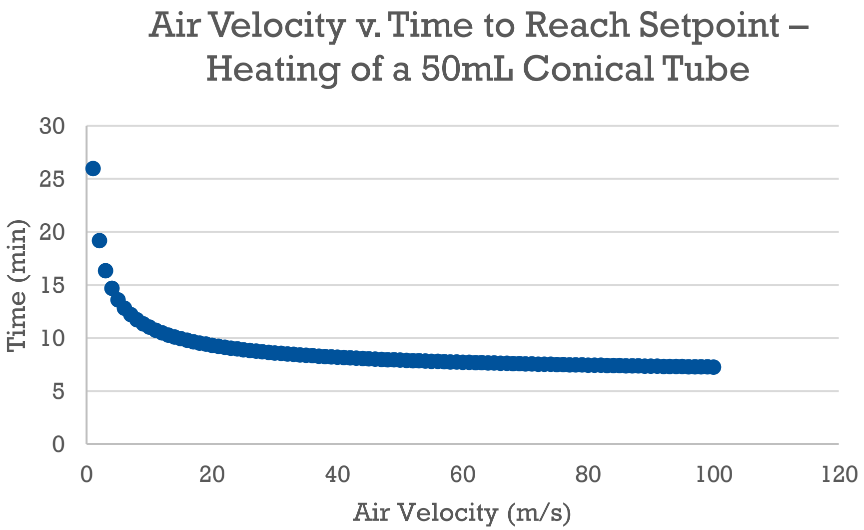
$$\frac{0.0119\left[\frac{\text{m}^3}{\text{s}}\right]}{1.5903 * 10^{-3}[\text{m}^2]} = 7.48\left[\frac{\text{m}}{\text{s}}\right]$$

Climate Control - Heating

- 200W DBK FGC3000 Series Fan Heater.
- Built in DIN rail clip.
- Maximum temperature of 70 C.
- 300,000h+ lifetime.
- Secondary function as defogger during cooling

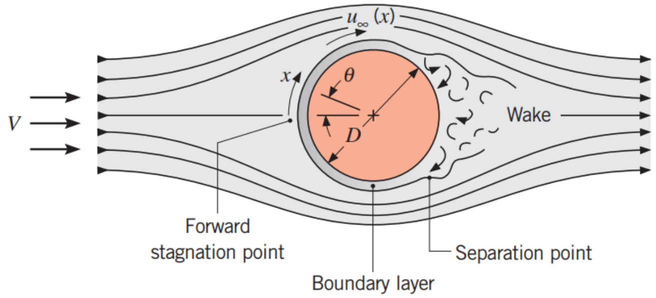


Climate Control - Heating



- With selected fan: 20C to 70C in under 14 minutes.

- Analysis based on forced convection: cylinder in cross-flow



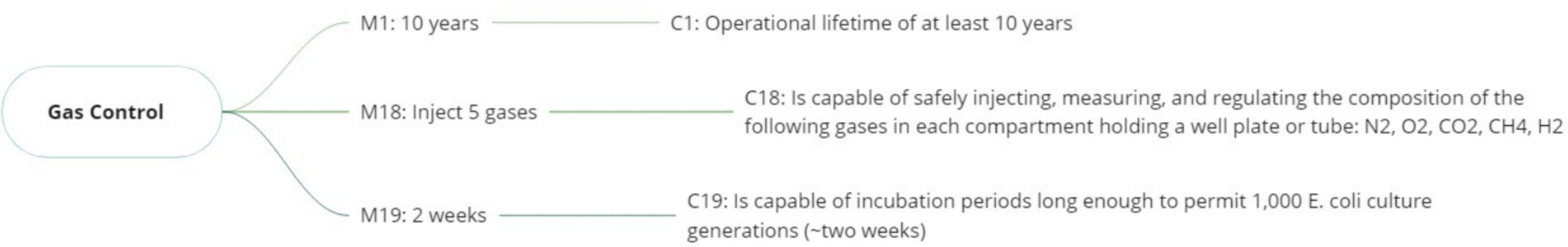
Climate Control - Heating

$T_{amb} = 70\ C$ $\quad T_{s,ext}$ $\quad T_{s,int}$

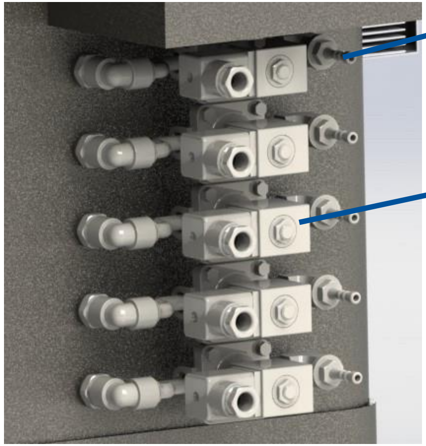
$T_{culture} = 20\ C$

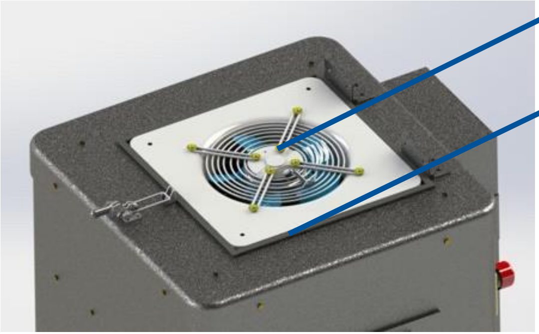
$R_{conv} = \frac{1}{\bar{h}A_1}$ $\quad R_{cond,t} = \frac{\ln\left(\frac{r_2}{r_1}\right)}{2\pi L_2 k}$ $\quad R_{cond,c} = \frac{L_3}{kA_3}$

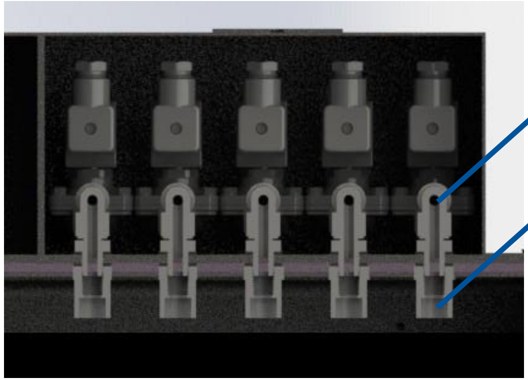
- Areas, radii, lengths from conical tube dimensions:
  - $A_1 = \pi d_o h = \pi * 30\ [mm] * 115\ [mm] = 10{,}838.49\ [mm^2]$
  - $A_3 = \pi d_i h = \pi * 26\ [mm] * 115\ [mm] = 9393.36\ [mm^2]$
  - $r_1 = 13\ [mm], r_2 = 15\ [mm], L_2 = 115\ [mm], L_3 = 13\ [mm]$
- Thermo properties for polypropylene (conical tube) surface at 20 C, air at 70 C, water at 20 C:
  - $v_{air} = 20.92 * 10^{-6}\left[\frac{m^2}{s}\right], k_{water} = 598.03\left[\frac{W}{m*K}\right], k_{air} = 30 * 10^{-3}\left[\frac{W}{m*K}\right]$
  - $Pr = 0.7,\ \mathrm{Pr_s} = 0.707, \mathrm{k_{PP}} = 0.165\left[\frac{\mathrm{W}}{\mathrm{m*K}}\right]$
  - $For\ 1000 < Re < 2 * 10^5:\ C = 0.26, m = 0.6$
  - $For\ Pr < 10 : n = 0.37$
- Modified convective heat transfer coefficient:
  - $\bar{h} = \frac{\overline{Nu_d}k}{D}$
  - $\overline{Nu_d} = CRe_D^m Pr^n (Pr \backslash Pr_s)^{\frac{1}{4}}$
  - $Re_D = \frac{VD}{v}$
- For fan speed 7.48 m/s:
  - $Re_D = \frac{7.48\left[\frac{m}{s}\right]*0.03[m]}{20.92*10^{-6}\left[\frac{m^2}{s}\right]} = 10{,}727$
  - $\overline{Nu_d} = 0.26(10727)^{0.6}(0.700)^{0.37}(0.700 \backslash 0.707)^{\frac{1}{4}} = 59.695$
  - $\bar{h} = 59.695 * \frac{30*10^{-3}\left[\frac{W}{m*K}\right]}{0.03[m]} = 59.695\left[\frac{W}{m^2*K}\right]$
- Heat transfer rate:
  - $q = \frac{\Delta T}{R_{tot}} = \frac{343\ [K]-293\ [K]}{2.748[\frac{K}{W}]} = 18.197[W]$
- Time required to reach setpoint:
  - $t = \frac{Q}{q} = \frac{14974[J]}{18.197[J/s]} = 789.31[s] = 13.16[min]$


Gas Control
M1: 10 years
C1: Operational lifetime of at least 10 years
M18: Inject 5 gases
C18: Is capable of safely injecting, measuring, and regulating the composition of the following gases in each compartment holding a well plate or tube: N2, O2, CO2, CH4, H2
M19: 2 weeks
C19: Is capable of incubation periods long enough to permit 1,000 E. coli culture generations (~two weeks)

Gas Control





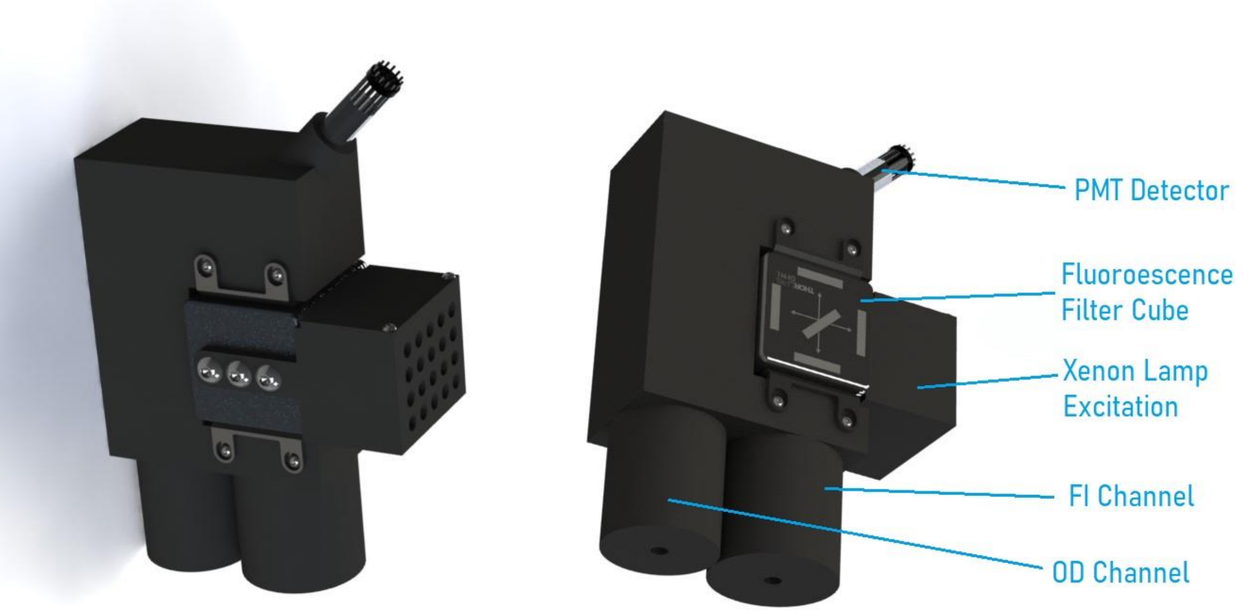


- Solenoid valves ASCO Red Hat
- Long life Equipment (5-20 million cycles)
- Two weeks of incubation

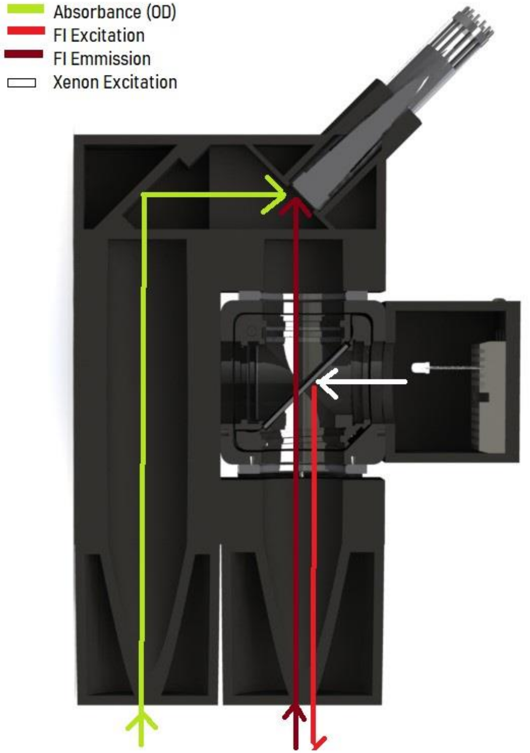
OD/FI

- M2: $10,000 — C2: Prototype production cost does not exceed $10,000
- M27: Illuminate from 400nm640nm at 1kW/cm3 — C27: Includes a photobioreactor mode to illuminate photosynthesis-capable cell cultures (e.g., cyanobacteria) with white light
- M30: Measure OD at 600nm — C30: Measures optical density (OD) in all individual wells and conical tubes
- M31: Excite fluorescence up to 640nm and detect emission up to 740 nm — C31: Measures fluorescent intensity (FI) in all individual wells and conical tubes
- M32: Maintain Light intensity of 640nm — C32: . For OD/FI measurement, sustains adequate light intensity to make measurements at wavelengths not lethal to cells
- M33: 6.5 Minutes — C33: Processes a 384 well plate through OD/FI measurements in less than 6.5 minutes

OD/FI Reader



- Includes:
  - PMT Detector
  - Filter Cube Chassis
  - Excitation
    Filter glass
  - Emission
    Filter Glass
  - Dichroic Mirror
  - Standard Mirror
  - Xenon Lamp



- **Analysis Speed**

$$\frac{Well\ Plate\ Dia. * (columns * rows + 2)}{Gantry\ Velocity\ Speed * Load\ Percentage} + (Analysis * well\ plates)$$
$$= \frac{0.0031 * (24 * 16 + 2)}{.513 * .6} + (0.15 * 384) = \mathbf{61.95}\ Seconds$$

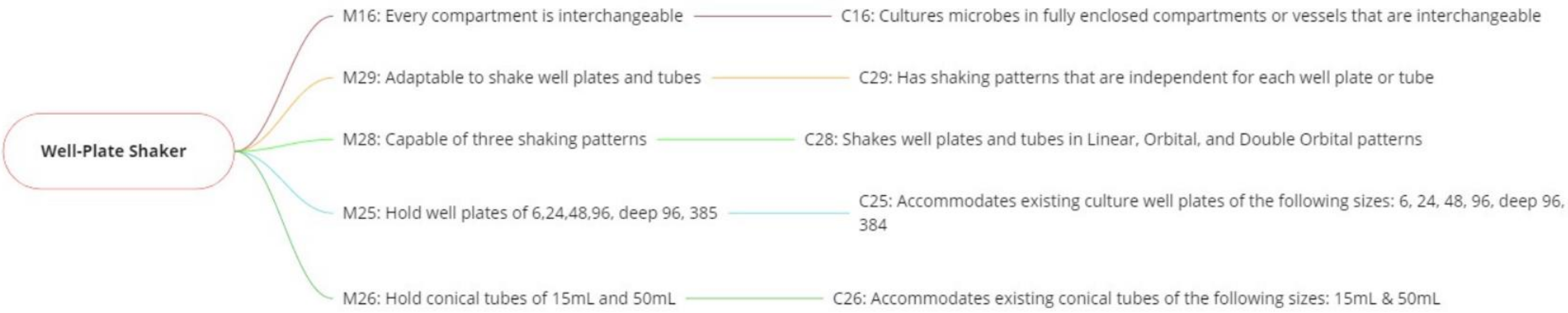
- **Cost**
  - OTS Parts: $1507.02
  - Material: $53.68
  - Manufacturing: $60.30

OD Capability
- Measures absorbance at 600 nm

FI Capability
- Measures Texas Red Fluorescence

White Light
- Illuminates well plate sample at 640nm 0.2 Kw/cm2

Lethality
- The 600nm featured ranges ensure all cells will live!


Well-Plate Shaker
M16: Every compartment is interchangeable
C16: Cultures microbes in fully enclosed compartments or vessels that are interchangeable
M29: Adaptable to shake well plates and tubes
C29: Has shaking patterns that are independent for each well plate or tube
M28: Capable of three shaking patterns
C28: Shakes well plates and tubes in Linear, Orbital, and Double Orbital patterns
M25: Hold well plates of 6,24,48,96, deep 96, 385
C25: Accommodates existing culture well plates of the following sizes: 6, 24, 48, 96, deep 96, 384
M26: Hold conical tubes of 15mL and 50mL
C26: Accommodates existing conical tubes of the following sizes: 15mL & 50mL

Shaker

- Linear, orbital, and double orbital shaking patterns
  - Uses springs and two motors to achieve a maximum velocity of 14.4963 m/s
- Interchangeable between well plates and tubes
- Tray designed with non-porous material

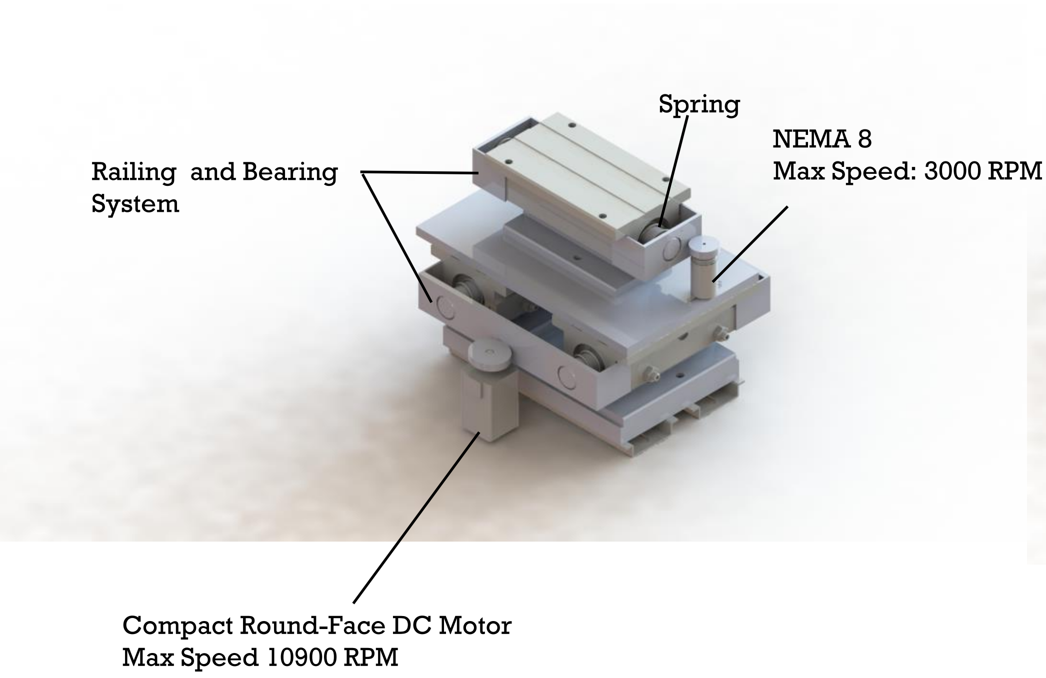
Governing Equations

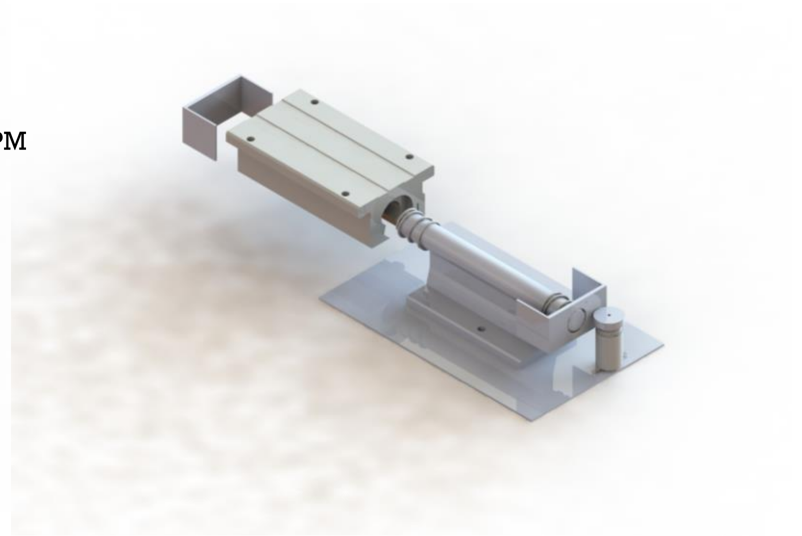
$$\omega = \frac{RPM}{60\ s/min} * 2\pi \frac{rad}{rev}$$

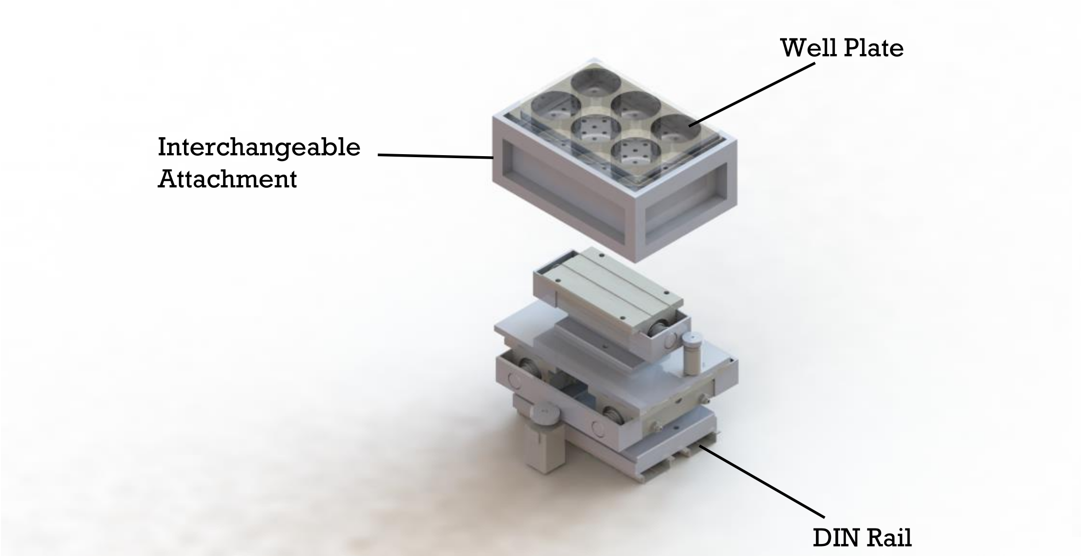
$$\omega = \frac{600\ RPM}{60\ s/min} = 62.83 \frac{rad}{s}$$

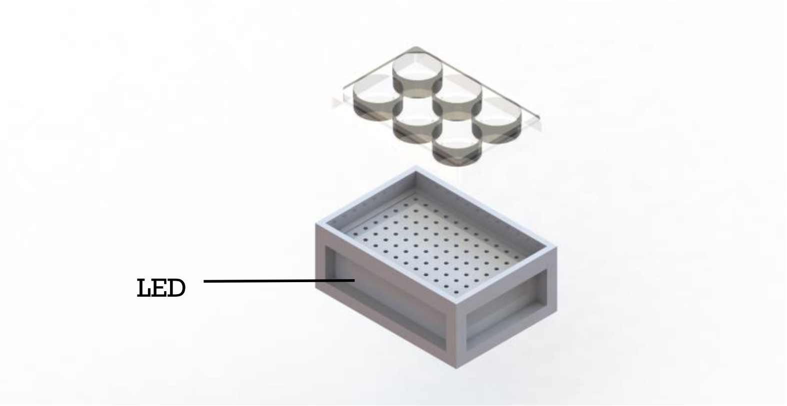
$$v = \omega r = 62.83 \frac{rad}{s} * 0.0381\ m = 2.39 \frac{m}{s}$$

$$v = \frac{10900\ RPM}{60} * 2\pi * 0.012\ m = 14.4963 \frac{m}{s}$$

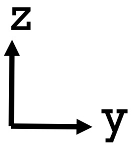

Spring
NEMA 8
Max Speed: 3000 RPM
Railing and Bearing System
Compact Round-Face DC Motor
Max Speed 10900 RPM

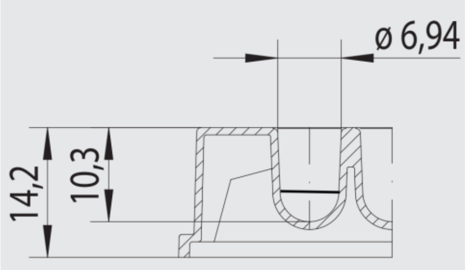


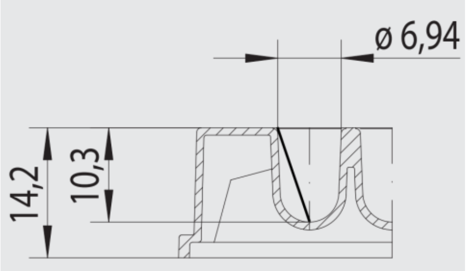

Well Plate
Interchangeable
Attachment
DIN Rail


LED

Shaker – Acceleration







Acceleration to cause spillage and cross contamination of cell cultures

$$\frac{\partial P}{\partial x} = -\rho \cdot a_x = 0 \rightarrow No\ acceleration\ in\ the\ x\ axis$$

$$\frac{\partial P}{\partial y} = -\rho \cdot a_y$$

$$\frac{\partial P}{\partial z} = -\rho \cdot (a_z + g) = -\rho \cdot g \rightarrow No\ acceleration\ in\ the\ z\ axis$$

$$dP = \frac{\partial P}{\partial x} d_x + \frac{\partial P}{\partial y} d_y + \frac{\partial P}{\partial z} d_z = -\rho \cdot a_y \cdot d_y - \rho \cdot g \cdot d_z = -\rho \cdot (a_y \cdot d_y + g \cdot d_z)$$

$$\frac{\partial P}{\partial z} = -\rho \cdot g \rightarrow \int_{P_2}^{P_1} \partial P = \int_{Z_2}^{Z_1} -\rho \cdot g \cdot \partial z \rightarrow P_1 - P_2 = -\rho \cdot g (Z_1 - Z_2)$$

Apply equation along surface of the liquid, since pressure is constant

$$dP = 0 \rightarrow Constant\ pressure\ in\ the\ surface$$

$$0 = -\rho \cdot (a_y \cdot d_y + g \cdot d_z)$$

$$a_y \cdot d_y = -g \cdot d_z \rightarrow -\frac{a_y}{g} = \frac{d_z}{d_y} \frac{z}{y}$$

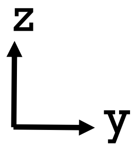
Shaker – Acceleration (Cont.)

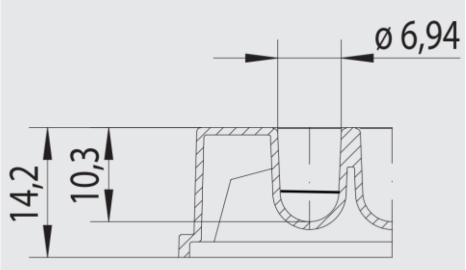
Relationship of y since volume is constant

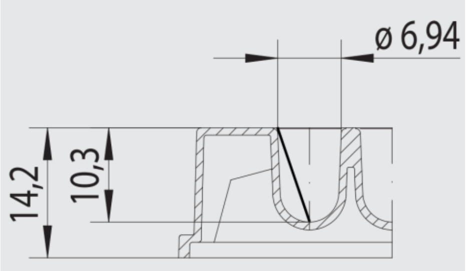
$$y = 2\frac{h_o L}{H}$$

$$a_y = -g \cdot \frac{Z_2 - Z_1}{Y_2 - Y_1} = -g \cdot \frac{z}{y} = -g \cdot \frac{z \cdot H}{2h_o L}$$

$$P_2 = \rho g(Z_1 - Z_2)$$







Thus, maximum acceleration before the cell cultures overspill is

$$y = 2\frac{h_o L}{H} = 2\frac{(4\ mm)(6.49\ mm)}{10.3\ mm} = 5.39\ mm$$

$$a_y = -g \cdot \frac{z}{y} = -9.81\frac{m}{s^2}\left(\frac{10.3\ mm}{5.39\ mm}\right) = 18.745\ m/s^2$$

Shaker – Acceleration (Cont.)

Maximum acceleration of the shaker:

Linear:

$$v = \frac{10900\ RPM}{60} * 2\pi * 0.012\ m = 14.4963\frac{m}{s}$$
$$\alpha = \frac{\pi(10900\ RPM)}{1(30)} = 1141.445\frac{rad}{s^2}$$
$$a = \alpha r = 1141.445\frac{rad}{s^2} * 0.012\ m = 13.69\frac{m}{s^2}$$

Orbital and Double Orbital:

$$\alpha = \frac{\pi(13900\ RPM)}{1(30)} = 1455.6$$
$$a = \alpha r = 1455.6\frac{rad}{s^2} * 0.012\ m = 17.47\frac{m}{s^2}$$

Cost

Subsystem	Total cost
Enclosure	$1468.54
Liquid handling	$323.47
User interface	$243.99
Climate control	$1903.44
Gas control	$1071.85
OD/FI	$1580.61
Shakers	$343.20
Total	**$6074.39**

Why CAMbox should be selected for prototyping

- Efficient use of space and funds.
- Possibility to market outside of universities.
- All customer needs were met.

Conclusion

- CAMbox is a compact, cost efficient design.
- The autonomous microbioreactor is a single environment and can house cell cultures for 2 weeks.
- This design is meant to be targetted at an audience outside of the University.

Thank you!

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