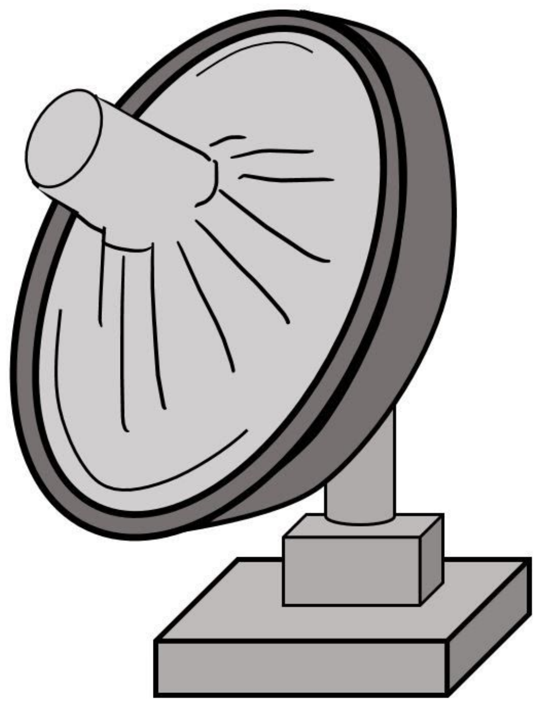
Microgravity Mushrooms



BSE 4125 - Comprehensive Senior Design Project

25 November 2020

Advisor: Dr. Clay Wright

Client: NASA, Jacob Torres

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List of Acronyms

APH	Advanced Plant Habitat
COTS	Commercial Off The Shelf
ISS	International Space Station
KSC	Kennedy Space Center
LEO	Low Earth Orbit
NASA	National Aeronautics & Space Administration
PONDS	Passive Orbital Nutrient Delivery System
PPTNDS	Passive Porous Tube Nutrient Delivery System
RH	Relative Humidity
USGS	U.S. Geological Survey
VSGC	Virginia Space Grant Consortium

Cover Letter

We have neither given nor received unauthorized assistance on this assignment

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Our advisor has had an opportunity to review this document.

Dr. Clay Wright

1.0 Introduction

1.1 Background

One challenge National Aeronautics & Space Administration (NASA) scientists face today is developing food products for astronauts that maintain their nutrient value and flavor over the course of five or more years. For food sent from Earth, a shelf life of five years is the minimum due to the time it takes to travel to and from Mars and the cost of launching resupply missions deep into space. Growing fresh food in space, reduces, if not eliminates, this need for food resupply missions.

Scientists and doctors who focus on astronaut physical and mental health also remark on the importance of maintaining weight, bone density, and muscle mass, in space. In order to maintain a healthy body in space, astronauts must eat nutritious meals full of grains, vegetables and fruits just like people on Earth. Despite a wide variety of freeze-dried food currently available on the International Space Station (ISS), doctors continue to see weight loss in astronauts. This indicates astronauts are not eating adequate amounts and suggests displeasure with current food options; a condition known as menu fatigue. Scientists and mental health professionals believe fresh food grown in space could help reduce menu fatigue and increase mental health; thus, increasing body mass and improving physical health.

With this challenge in mind, the Kennedy Space Center (KSC) recently started conducting research on oyster mushroom cultivation under a grant provided by the Virginia Space Grant Consortium (VSGC). Mushrooms are full of important vitamins, and act as a great addition to crops already grown on the ISS. Other NASA research, conducted at Ames Research Center, focuses on mushroom cultivation from a synthetic biology standpoint, in hopes to one day grow martian habitats from the mycelium. NASA is not the only one looking into mushroom cultivation and mushroom synthetic biology. According to National Geographic, mushroom research is being conducted with applications related to building materials, medicine, cleaning products, textiles, biofuels, and packaging, to name a few. With so many potential uses for mushrooms, it is easy to understand why this versatile crop is of importance in deep space travel.

1.2 Client Information

The product of this senior design project is intended for use by the Kennedy Space Center, where Biosystems Engineer, Jacob Torres, will use it to conduct research on the development of Passive Porous Tube Nutrient Delivery System (PPTNDS) for use in microgravity plant and mushroom cultivation. The PPTNDS, as indicated by its acronym, is a passive system that utilizes capillary action to deliver water and nutrients to plant roots. Seeds are glued to the porous tube so that the water and nutrients are easily absorbed. Water and nutrients are circulated through the porous tube and the reservoir, which are connected via non-porous tubing. This technology is beneficial to NASA because it is (1) low maintenance, (2) eliminates the most common source of error in active systems (the pump), and (3) has been shown to use less water than other systems. On-going research at KSC continues to show successful growth of tomato plants using the PPTNDS. For example, research conducted by Torres compares tomato growth in an NFT system and PPTNDS. Specifically he looks at water requirements, edible yield, system area and crew interaction (NASA Technology Transfer Program, 2020) (Table 1).

Other research, sponsored by the Virginia Space Grant Consortium, looks into hydroponic mushroom cultivation on Earth and in Microgravity. The experiment compares mushrooms grown using the Nutrient Film Technique (NFT), the Ebb/Flow system, a commercial off the shelf (COTS) mushroom kit purchased from Back to the Roots, and the PPTNDS.

Table 1: Comparison between the NFT and the PPTNDS

	NFT	PPTNDS
Water consumption (L)	316.7	76.32
Edible mass (g)	3511.40	2084.40
Area taken up by system supporting 6 plants (cm^2)	105.63	29.01
Number of crew interactions	78	62

1.3 Problem Statement & Design Deliverables

Though results for the hydroponic mushroom growth experiment are still being analyzed, current data indicates the need for a redesign of the PPTNDS adapted for use with mushroom hydroponics. Torres believes “the PPTNDS is not supplying enough moisture to the substrate.” While the control system yielded 27 grams of edible mushrooms, the PPTNDS produced only one very small clump of mushrooms, about the size of a pinky finger nail (Figure 1).

Figure 1: On the left is the control system using Hoagland's solution. It yielded about 27 grams. On the right is the PPNTDS using Hoagland's solution. The yield was not measured. Due to COVID-19, the facility was shut down and further growth was not recorded.

Torres suggests that this issue appears to be gravity driven. In other words, the movement of water through the PPTNDS is driven by capillary action and while this occurs in both gravity and microgravity environments, the height of the porous tube above the water reservoir is expected to play a larger role in a gravity environment than it is in space or a reduced gravity environment, such as the moon. Improving the design of the PPTNDS for use in gravity, will enable KSC scientists to perform the mandatory ground research before launching the system to space. For this purpose, our client has asked us to design and produce an adjustable stand for use in gravity that enables increased control over the capillary action taking place within the PPTNDS to better match microgravity conditions.

An additional requirement of the stand is that it include or support a pressure-increasing mechanism. Though water loss from the PPTNDS is minimal, as water is lost the pressure within the reservoir is reduced. Reduced pressure prevents proper delivery of water to the plant roots. Torres has shared his interest in developing a mechanism that will passively increase the reservoir pressure when water is low, in both gravity and microgravity environments.

An additional concern is the low-humidity atmosphere in which the PPTNDS is currently operating, as noted by our advisor: Virginia Tech Biological Systems Engineering professor, Dr. Clay Wright. The substrate (a thin strip glued on top of the porous tube) can potentially lose water via evaporation in open atmosphere conditions. Humidity levels aboard the ISS, and subsequently laboratories, are maintained on average to 60% which induces conditions for further water loss (Hamed Aghajani, et al. 2018).

Oyster mushrooms are capable of growing in a multitude of environmental conditions, but the optimal yield requires 80-85% humidity (Hamed Aghajani, et al. 2018). Thus, a proposed housing apparatus containing the substrate and mycelia within the PPTNDS will improve mushroom yields. This apparatus should better sustain the desired humidity levels, lighting, and temperature, and will be equipped with additional instrumentation; specifically micro-controller units, to help NASA's research personnel record environmental conditions to study how they affect crop yields. The housing apparatus will also help prevent contamination of the inoculated spawn during the growth phase of the mycelia, as well as reduce the level of spores spreading out to other units.

Also addressed by Torres, was the lack of research focused on the optimization of porous tube design for mushroom cultivation. Even with adequate funding, ordering supplies and equipment is difficult due to administration protocols. So far, the PPTNDS for mushroom cultivation has been studied using only tubes on-hand at KSC. The tubes currently used have an assumed pore size of 5 microns and are approximately 12 inches long with a 2 inch outer diameter. Arbitrarily selected based on availability, these porous tubes may not support optimal growing conditions. Torres expects that pore size and tube geometry play a role in not only mushroom cultivation, but also the cultivation of all crops. Under this notion, our team has agreed to design a Pore Size and Geometry Model that will help NASA determine the best tube for a specific crop based on factors such as desired yield and expected water requirements. It is our goal that this model will help NASA save time and money when ordering new porous tubes and know that predictive capabilities of this type will contribute to the future PPTNDS research Torres has in mind.

1.4 Constraints & Criteria

The table below includes general constraints that limit the solution model (left column) as well as design criteria that directly apply to these constraints along with why they belong in these categories (right column).

1.4.1 Mimicking Microgravity

Design Criteria	**Scientific Constraints**
Adjustable between 5-10 cm	Must be very close to the ground in order to reduce gravitational-induced issues with the capillary action. A stand that is too tall would allow gravity to play a larger role in counteracting the capillary action in the porous tubes.
3D printable	ABS plastic should be used because of the ties to NASA's Additive Manufacturing Facility and the commonality of this material.
Easily scalable for tubes of other geometry	The design must be sturdy enough to comfortably support the PPTNDS (for a future determined weight) while not hindering the porous tube system.

1.4.2 Predicting Moisture Levels

Design Criteria	**Scientific Constraints**
Applicable for use with a variety of plants and varying tube materials	Moisture requirements can vary greatly across plant and fungi species. The model will only be as accurate as the inputted values. Moisture requirement values will either come from literature review or researcher observation and estimates.
Applicable for use with products easily manufactured by industry	Mycelia of mushrooms can not grip to conventional tube-shapes like plant roots. Unlike other plants cultivated by seeds, the mushrooms require substrate.
Easily accessible to NASA engineers via free software, software supported by NASA, or a public access, user friendly application	It is nearly impossible to mathematically express how each variable affects desired moisture level, but to be useful, the predictive model must account for as many as possible, as accurately as possible.

1.4.3 Maintaining Environmental Conditions

Design Criteria	Scientific Constraints
Low energy and maintenance	The normal RH (relative humidity) aboard the ISS is approximately 60%, whereas the RH preferred by oyster mycelium is 80-85%, so it is important to develop a system that allows harvesting with minimal to no impact on RH in either system (Hamed Aghajani, et al. 2018)
Allows for easy cultivation and harvest of mushrooms	With the PPTNDS it will be quite difficult to make a supported cylindrical casing that is able to be easily dismantled at any interval for harvesting or maintenance.
Limits environmental contamination via spores	It will be difficult to ensure complete isolation of mushroom spores during harvesting or airflow circulation. Failure to do so could lead to possible contamination of the ISS air system.

1.5 Scope of Report

The purpose of this report is to give an introduction to the design needs and standards as they relate to the PPTNDS for use in mushroom cultivation by NASA. Also discussed is relevant past, present, and future research conducted by NASA as well as a review of existing mushroom cultivation technologies. Preliminary design ideas and design alternatives will be discussed at a basic level in section 3. In section 5 we present highlights from our brainstorming sessions, expected challenges and mitigation plans, and our project timeline.

2.0 Review of Technology

2.1 Scientific Literature Review

2.1.1 Understanding Microgravity

We know that Earth's Gravitational Acceleration Constant (g), commonly referred to as just gravity, equals (to three significant figures) 9.81 $\frac{m}{s^2}$. This value is responsible for and affects the forces that affect our everyday life. In Low Earth Orbit (LEO), this value differs, meaning a change in the behavior of certain substances such as water. Capillary action, a behavior of water, occurs within the PPTNDS and is limited by gravity. Microgravity, as experienced on the ISS, is about 90 percent of Earth's gravity. To put this into perspective, NASA explains that a person weighing 100 lbs on Earth, would weigh 90 lbs at an altitude of around 250 miles above Earth. The reason objects 'float' on the ISS, is due to the fact that the station, along with everything in it, is in perpetual free-fall around the surface of the Earth. As such, the effective gravitational force felt on the ISS is essentially none ("What Is Microgravity NASA," 2017). Similarly, gravity experienced on the moon and mars is about 16 and 30 percent of Earth's gravity, respectively. Given this, it is expected that capillary action, as necessary within the PPTNDS, will function more effectively on the ISS and other planetary bodies than in the lab on Earth. In other words, the water in the PPTNDS will travel a longer vertical distance on ISS than on Earth, given non-porous tubes of the same diameter.

An important part of crop production research, and mushroom research, at KSC depends on ground and microgravity testing in similar conditions. NASA's "Plant Habitat-01" experiment tested how *Arabidopsis* responds in microgravity environments aboard the ISS. The series of tests analyzed photosynthetic efficiencies, gravity sensing, metabolic, and genetic effects ("Plant Habitat-01," n.d.). In following studies, *Arabidopsis thaliana* demonstrated decreased cell proliferation, likely due to alterations in cell cycle regulation, in microgravity-grown samples (Matía et al., 2010). Further studies are important for understanding both long-term plant growth in space and how gravity affects plants on the ground. While the PPTNDS cannot be utilized in current ground microgravity simulators at the KSC Microgravity Simulation Support Facility, such as the Slow Rotating Clinostats, Rotating Wall Vessel Bioreactors, and the Random Positioning Machines, researchers can ensure that both ground and ISS systems provide the crops with the same amount of water and nutrients. One way of doing this is by controlling the capillary action.

It is commonly understood that there is a relationship between capillary action, more specifically adhesion, and tube diameter. As described by the U.S. Geological Survey (USGS, n.d.), capillary action is responsible for moving water and nutrients throughout trees. It is stated that the force of gravity is too great to allow for adequate movement of water from the roots of a tree to the leaves. To account for this, xylem, very small tubes within the plant tissue, increase adhesion by decreasing their diameter to help to transport the water. Of course, xylem are on the micro-scale, meaning tubes of their size are incompatible with the current PPTNDS system. As such, the height of the porous tube above the reservoir, fed by non-porous tubes, will help to regulate the amount of water delivered to the mushrooms (Figure 2).

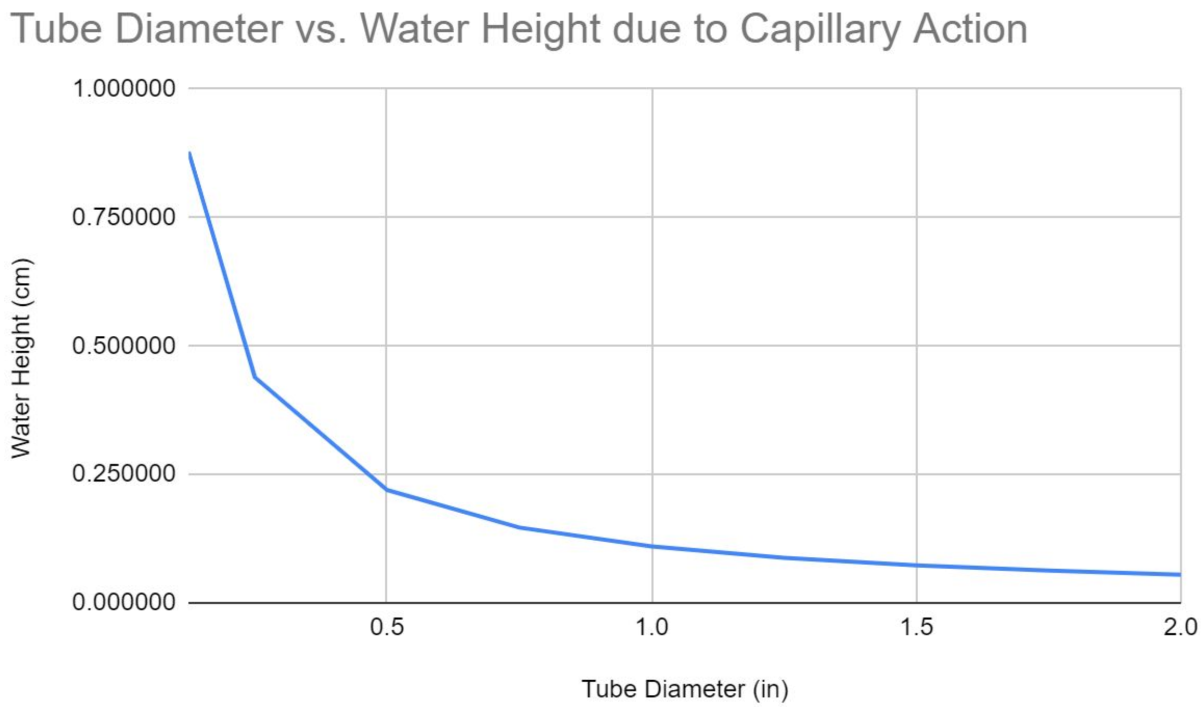

Figure 2: Height water is able to travel due to capillary action per tube diameter in normal gravity

2.1.2 Understanding Moisture Requirements

In order to design the PPTNDS for optimal delivery and control of moisture levels for various crops, specifically Oyster Mushrooms, a baseline range of moisture requirements must be established. According to researchers (Chang & Miles, 2004), the moisture in the substrate should range between 50% and 75% by mass. That said, other research shows that diseases and molds tend to grow at a substrate moisture level above 70% (Moonmoon et al., 2010). With the use of sterile substrate in the sterile environment of the ISS, disease and mold growth is less of a threat, so for the purpose of our design, the 70% moisture level will not be a limitation, but is a constraint when testing our apparatus in non-sterile areas.

With this moisture range established, the next step to understanding moisture requirements is being able to control and measure the amount of moisture delivered via the PPTNDS. A "pore size and tube diameter model for predicting moisture levels", described in section 2.4.2 will be developed and used to determine both the amount of water supplied by existing tubes and to decide the optimal moisture levels that should be delivered to different plants and fungi based on their specific individual requirements. Even though the water delivery is a passive process, there are variables we can adjust to alter the rate and amount of water delivered to the roots of plants or mycelia of mushrooms. The effect of tube diameter on water height by capillary action was discussed in section 2.1.1. Another factor contributing directly to the amount of moisture emitted is the porosity of the tubes and size of pores. To be more exact, the pores will affect the Hydraulic Conductivity of the tubes: the intrinsic permeability of a porous media to any fluid. This property has a large influence over the amount of moisture released and its wetting pattern of water release; most importantly the range of water seepage through another media such as the substrate - also known as the "wetting front".

A study conducted by Neelkanth and Gatesaniya analyzes the effect of porosity and pore size on cotton plant growth using passive porous clay pipe irrigation. The results are discussed in their article published in 2017: *Experimental Investigations on Localized Porous Clay Pipe Irrigation Technique for Sustainable Agriculture: A new Paradigm for Soil and Water Management in Rained Areas.* The researchers compared a number of variables between the passive porous clay pipe irrigation system and the control rainfed fields, including crop yield. It was found that the porous clay pipe irrigation system approximately doubled the yield from the crops. The researchers indicated the importance of controlling tube porosity and size, which is directly relevant to the development of our Pore Size and Tube Geometry Model. The study made use of handmade porous clay pipes which have a tube porosity of 59.80%. The saturated hydraulic conductivity was calculated as 1.598x10-5 cm/s using the constant head method, meaning it was calculated when the outflow was at a constant rate, using the following equation:

$$K = \frac{V*L}{A*h*t}$$ **……………………….. (1)**

Where K = hydraulic conductivity (cm/s)

V= Volume of collected water (cm^3)

L= Average wall thickness of pipe (cm)

A= surface area of clay pipe (square cm)

h= constant head (cm)

t = time (s)

We can use this methodology and equation to estimate how different qualities of our tubes affect its hydraulic conductivity.

This study also related the rate of water circulation with pore size; two components relevant to our design project. Due to ever changing weather conditions, it was important for researchers to test the ease of which flow rate of water to the crops could be adjusted. It was found that for clay piping, less time was required for plants to receive sufficient water when the pores were larger. This accelerated water delivery did have a downfall, however. Research showed that when the pipes remained dry for a long period of time, pores clogged due to salt accumulation and precipitation. With this blockage, the pipes did not function properly (Bhatt and Gatesaniya, 2017). It is unlikely we will experience an issue of this sort with our PPTNDS in a controlled environment, but a constraint to be aware of nonetheless, especially if we wish to scale up the system.

2.1.3 Understanding Environmental Conditions

The life cycle for mushrooms consists of phases for decomposition, vegetative development, and reproduction. To optimize the growth yields for mushrooms, temperature, humidity, and airflow have to be adjusted throughout the different phases (Grant, 2002).

A key differentiating factor for growing mushrooms versus traditional plant growth is the maintenance of moisture within the substrate and humidity in the surrounding air pocket. This high humidity and moisture level are necessary for the mycelium since they lack a root system that can both actively search out water sources and provide foundational support. For example woody plants have roots which penetrate substrate in search of water and subsequently do not necessarily need a substrate foundation, whereas mushrooms only have mycelium which are analogous to a root system in function, but can only work in areas where there is a solid foundation to grow on or inside. The type of foundational substrate also is significant to overall growth yield even before the environmental factors are considered; for example, sugarcane bagasse promotes a higher overall production yield as compared to corn cobs (Ha Thi Hoa et al., 2015).

The type of substrate will determine initial mycelium colonization rate, but the fruiting and expansion quality rely upon the overall environmental control. Although mycelia will develop and push into the budding phase in an open air environment, optimal growth conditions will require an enclosure until the mycelia decomposes a majority of the nutrients in a given substrate. Airflow is necessary in the initial phase to exchange heat from all metabolic processes (Grant, 2002). The conditions for airflow, as well as evaporation, are critical prior to the reproduction phase. The fruiting phase is initiated once the mycelia enters a phase of shock where temperature, and carbon dioxide levels are dropped followed by increased airflow to support evaporation (Grant, 2002). This period of oxygenation is an indicator for the mycelia to begin fruiting to spread further spores, likewise the airflow prevents oversaturation of the mushroom caps and bacterial growth.

As such, it becomes valuable to monitor humidity levels in a contained environment with a goal of optimizing the mushroom yield and biological efficiency of the substrate. There exists an abundance of studies attempting to figure out the ideal substrate for mushroom yield, and therefore ideal conditions for water supply. The intended range for moisture within the substrate via the PPTNDS supply is from 80-85%

moisture (Islam et al., 2009). The goal for the housing apparatus is to maintain a similar humidity level of 85% within the surrounding environment at a temperature of 25˚ C. This goal may be achieved through similar technology already used on the ISS: the Temperature and Humidity Control Subsystem, but scaled down to a level for the PPTNDS (Tesfay et al., 2020).

2.2 Evaluation of Existing Technology

2.2.1 Mimicking Microgravity

Several devices have been developed by NASA already for nutrient delivery in microgravity environments. This includes the Astroculture system, and the Zeoponics system, and Porous Tube Plant Nutrient Delivery System (PTPNDS), from which the PPTNDS is based (Goins, 1997). Each of these systems are built for use in microgravity, however our stand is intended for testing in gravity. For systems on earth, there have been several patents for adjusting the moisture level in hydroponic systems. Although they do not use adjustable stands, some use adjustable stand-pipe siphons (Goins, 1999) to adjust moisture levels and nutrient delivery.

Other hydroponic devices have been developed to address similar issues. One example is a 1998 patent that develops a device that adjusts to various growth methods and can be easily constructed from off-the-shelf materials. The device also relies on capillary action and it’s method of both adjustability and pressure controls are unique (Auer, 1998). Their use of a pressure equalizing liquid harness is novel and could be built on for our PPTNDS stand in gravity. A 2015 patent developed a unique method for adjusting vertical hydroponic growing systems. Although the adjustment method is relevant, this design relies on gravity flow within growth tubes for nutrient and water delivery which would not be relevant to a design such as the PPTNDS, and would not be relevant in microgravity (Collins and Hertel, 2015).

2.2.2 Predicting Moisture Levels

Putting our predictive methods into practice will require the incorporation of software that when inputted data from the PPTNDS, will output projected system properties like tube porosity and diameter. With repetition, the program's code can be improved to output predictions with higher accuracy across a range of desired moisture levels and system parameters. Data such as "V, the volume of collected water" can be collected manually, but it is ideal that we automate its collection using sensor(s).

Many different softwares exist that could be useful in designing our model. MATLAB is being considered given the team's experience in and free access to the program. MATLAB is a multi-use tool that functions as a numerical computing and modeling environment. The program allows the plotting of functions and data, the implementation of algorithms and the creation of user interfaces. This will be useful in analyzing and organizing data, and improving our model as more trials are run. Additionally, the team is interested in one of two design alternatives (1) creating a graphic user interface, such as an easy to use application or (2) using software that is available to NASA or compatible with the software used with NASA. MATLAB allows for both of these alternatives as it also has the ability to interface with other programming languages.

2.2.3 Maintaining Environmental Conditions

One of the most notable applications of enclosed housing for microgravity agricultural growth is NASA's PONDS (Passive Orbital Nutrient Delivery System) technology. The system follows a similar design mechanic to passively fuel water to plant roots. Although the delivery system functions differently from PPTNDS, the casing provides a top cover to the seed chamber (Sempsrott, 2019).

The PONDS layout is structured into three main compartments. The first layer is a water reservoir which can be filled via an attached tubing port. The following layer is a soil compartment which is encased in a plastic mold to prevent full submersion into the water. The final layer is a seeding chamber encased by an upper plastic mold that retains humidity during the initial growth phase. The casing is a design implemented by Tupperware Brands, and Techshot to replace NASA’s seed pillow plant studies (Sempsrott, 2020).

Furthermore, PONDS is built to be utilized in sequence with NASA’s Advanced Plant Habitat (APH). The APH system is a fully enclosed, automated growth chamber that is equipped with a complex unit of inputs, and outputs to control oxygen, carbon dioxide, water, temperature, and humidity. The APH system has a core component known as the Environmental Control System (ECS) which adjusts the temperature, humidity, and air flow. Coupled with an enclosed growth chamber for plants, this unit provides the fundamental outline to which the PPTNDS housing apparatus will be adjusted to (McAllister, 2018).

Although the above applications are complex in design, there are simple applications to contained mushroom production with minimal components. There are a series of independent mushroom cultivation fruiting chambers known as monotubs (NS, 2020). Monotub fruiting chambers are essentially sterilized plastic tubs with air sockets for exchange of carbon dioxide. Alternative minimalist designs include plastic bags encasing the substrate, and inoculated mycelia as a fruiting block (NS, 2020).

The bags are then suspended or stored within a growth chamber which is adjusted for temperature, and humidity conditions. There are various other at home applications for mushroom growth much of which is dependent on the type of mushroom, and the desired growth yield. A common addition to the design kits is often the use of Node MCU (micro-controller units) such as Arduino boards. The boards are typically equipped and programmed with components to monitor temperature (LM35), or humidity (DHT-11) (Subedi et al. 2019). There are further series of components that can be adapted to the Arduino system as well as standalone products specifically designed to monitor a unique trait. The data collected from these design components can then be analyzed for modifications on stand height, casing airflow, and tubing porosity dimensions.

Table 2: Types of growth chambers

Type:	***Application:***	***Description:***
PONDS:	Commercial	A passive water delivery system encased for both capillary action, and initial seeded growth phase for root based plants
APH:	Industrial	A growth facilitation chamber designed with an ECS unit to monitor environmental inputs, and outputs in a closed loop unit
Alternative Fruiting Chambers :	Common	A minimalistic box approach to APH using either plastic bins, or plastic bags as a fruiting chamber, typical designs monitor using environmental micro-controller units

2.3 Design Standards

ISO/ASTM WD 52933

Although most standards for additive manufacturing are set by the industry they are used in, there are some ISO standards already in place. This includes **ISO/ASTM WD 52933** which lists environmental health and safety concerns for the reduction of hazardous substances emitted during the operation of typical 3D printers in the workplace (“Standards by ISO/TC 261”, 2020). This standard would be especially important to NASA, who is conducting research in space on their Additive Manufacturing Facility (Werkheiser). Emitting hazardous particles into an enclosed environment such as a space station would be very dangerous. NASA’s Additive Manufacturing facility is leading the charge on additive manufacturing research in space. Their experiments test Acrylonitrile Butadiene Styrene (ABS) plastic, a common filament material. Although the stand is being designed specifically for on-ground testing of the PPTNDS in gravity, it is useful to look forward to how NASA could adapt the same materials for use in microgravity environments.

ISO 13636: 2012

Standards are set to regulate the materials that come in contact with food. Since the PPTNDS is used by astronauts to grow food, including oyster mushrooms, it will be important to ensure all components meet **ISO 13636:2012** (“ISO - ISO 13636:2012 - Plastics — Film and sheeting — Non-oriented poly(ethylene terephthalate) (PET) sheets,” 2012). This standard is applicable to non-oriented polyethylene terephthalate sheets less than 2.0 mm in thickness. There is some thought that this could lead to microplastic interference with the mushroom growth, which should be as limited as possible due to immune concerns regarding plastic consumption (Wright and Kelly, 2017). All materials in the PPTNDS that come in contact with the crops or with the water and nutrient solution delivered to the crops, should be tested according to this standard.

NOSB 205.203(c) & NOSB 205.209(a)(5)

According to the USDA National Organic Standards Board, regulation **NOSB 205.203(C)** (Giacomini, 2010), requires that the producer manage plant matter in a way that does not contribute to contamination of soil. While we are not growing in a terrestrial (outdoors) environment, the mushroom cultures used in NASA's research are considered organic. In order to maintain the crop's organic classification, it will be important to ensure that the PPTNDS manages plant matter in a fashion that meets this standard. Should a different source of mushroom cultures that are not organic, be found more compatible with the PPNTDS, this and other NOSB standards can be ignored. Furthermore, **NOSB 205.209** (Giacomini, 2010). , requires that ventilation systems eliminate the drift of prohibited materials into the organic growth enclosure. This will be relevant to the design of our housing.

NASA-STD-3001, 6.2.3

NASA's ongoing goal through their plant habitat systems is to maintain a closed loop environmental system. Through developing closed loop systems, research components are able to avoid changing environmental conditions in the surrounding compartments aboard the ISS. NASA's set standards on humidity pertain to human comfort, health factors, and evaporation conditions ("NASA-STD-3001 VOL 2 | NASA Technical Standards System (NTSS)," 2019). In relation to the PPTNDS, the issue involves potentially increasing the amount of humidity in the surrounding compartments. There is a lower level (25%) by which the humidity has to be controlled within, but the upper limit (75%) is of more concern ("NASA-STD-3001 VOL 2 | NASA Technical Standards System (NTSS)," 2019). As water is fed through the PPTNDS to the mycelia, there is a factor of concern as the water eventually exchanges into the air. Initial plans thus far will have minimal exchange but the amount by which the humidity increases is incremental and still requires attention. Further concerns pertain to electrical components in the surrounding compartments ("NASA-STD-3001 VOL 2 | NASA Technical Standards System (NTSS)," 2019). Although the design concern for housing is related to the substrate, and mycelium growth, the exchange to the surrounding compartment could potentially damage equipment.

2.4 Design Considerations and Alternatives

2.4.1 Stand for Mimicking Microgravity

Engineer Jacob Torres indicated that the stand they are using for testing the PPTNDS in gravity is insufficient. Currently, the PPTNDS sits above the reservoir on blocks that hold the system in place. However in gravity they pose issues working against the capillary forces that drive the PPTNDS. Because capillary action works against gravity, they can reach a point where the pull of gravity is too much to overcome. A stand must be designed specifically for the PPTNDS to replace these blocks and fix the gravitational issues. The stand should be easily adjustable to change the height of the device over the reservoir to study the effect on nutrient delivery.

The ideal material for developing and building the adjustable stand will be 3D printed plastic. Additive manufacturing has been a growing technique for the manufacturing of such devices, offering benefits such as rapid prototyping and very specific dimensions and geometries. Another advantage of using 3D printed plastic for manufacturing of the PPTNDS stand includes the ability to adjust and make changes very easily both on the ground and in space. Limitations to additive manufacturing include a relatively weak strength attributed to most plastic filaments and concerns over end-of-usage waste (Allen, 2016).

The stand will not only be utilized in his research, but will also be useful for adjusting moisture levels within the system to accommodate different plant/fungi preferences and to adjust to different gravitational conditions.

Another requirement is that the stand supports a passive method of pressure application to the water reservoir bag. The application of pressure to the reservoir will help the capillary action overcome gravity, thus allowing the system to better mimic the microgravity conditions. Torres mentions that this passive pressure system could also be of use in microgravity conditions on the ISS. As such, design alternatives must accommodate both earth and space environments and must take into account the change in reservoir pressure by evaporation. Ideally, this pressure system could connect to and make use of the stand design. This would eliminate the number of parts and pieces within the system and hopefully reduce the mass of the system.

2.4.2 Pore Size and Tube Geometry Model for Predicting Moisture Levels

The Pore Size and Tube Geometry Model will be designed with NASA researchers in mind. This product is intended to be a tool used during the preliminary stages of research in the Space Crop Production Laboratory at KSC. Researchers will be able to input variables such as “Plant and Mushroom Water Requirements” and model ‘the ideal tube’ for the desired crop and maximum yield. Conversely, researchers will be able to use the model to also input variables such as “known pore size and tube geometry”, to determine the amount of water supplied by existing tubes. This information could be used to place custom tube orders with a manufacturer or to identify and troubleshoot errors within existing PPTNDS(s).

The Pore Size and Tube Geometry Model will depend on a number of factors within the system including the different system components, their dimensions, and mass-balance variables. These factors are summarized in table 3 & 4 below.

Table 3: System components for porous tubes

System Components	Description
Porous Tube	Largest tube in system. Runs horizontally above the reservoir and is supported with stands. Provides moisture and root space to plants. Substrate will be glued on to this tube. Porous tube provided by KSC. Tube diameter, length, and pore size vary depending on plant/fungi type.
Non-Porous Tubes	Two tubes run water up from reservoir to porous tube via capillary action. Current design features platinum-cured silicone pump tubing from MasterFlex. Diameter = ¼ inch. Length = cut to user preference.
Reservoir	HTI's OsMemTM Forward Osmosis membrane technology. Similar to IV bag, but has two inlet/out holes. Traditionally used to purify urine and waste water on the ISS. Filter is taken out for use with the PPTNDS. Reservoir is translucent and placed in an opaque pouch to reduce algae growth via photosynthesis (Mansfield, 2011). Reservoir capacity is currently under investigation as the team could not find dimensions. See Figure 3.

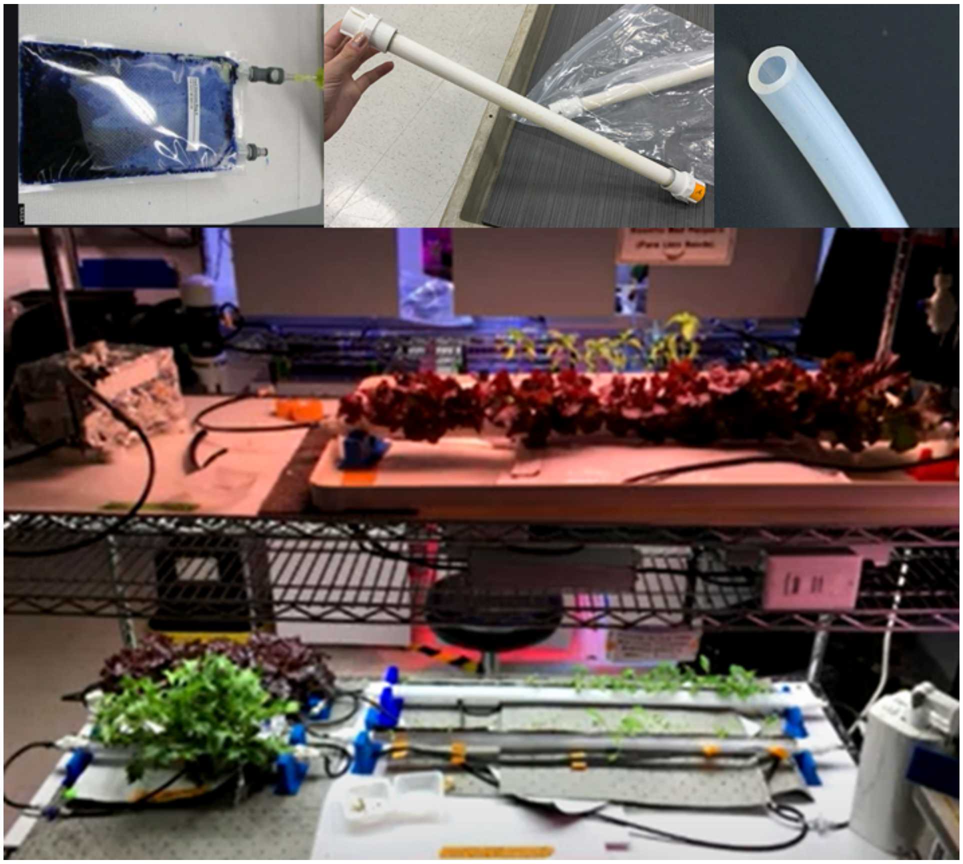

Figure 3:Top Left: Forward Osmosis Bag by HTI (Smith, 2011). Top Middle: KSC Porous Tube. Top right: Silicone Piping (Smith, 2011). Bottom: Fully set up PPTNDS in KSC Space Crop Production Laboratory (NASA Technology Transfer Program, 2020).

Table 4: Variables for mass balance of porous tubes

Variables	**Units**
Porous Tube Length	cm
Porous Tube Surface Area	cm^2
Mushroom Water Requirement	mL
Capillary Pressure	Pa
Pore Size	μm
Flow Rate	cm^3/s
Assumed Variables	
Substrate Saturation (100%)	g water / g substrate
Evapotranspiration (0%)	mm/h

The success of the model will depend heavily on these factors functioning as predicted and it will be critical to balance the complexity and utility of this predictive model. As there are many important factors, more than we could possibly account for within the scope of this course, special attention will be given to narrowing down the most significant ones. Moving forward, we will employ a decision matrix to weigh the importance of the different variables. This decision making tool will help ensure the model accounts for only the variables of highest significance and is not complicated by minor factors. This should keep the model from being inconvenient or burdomson to use.

2.4.3 Housing Apparatus for Maintaining Environmental Conditions

The studies, and plant growth systems currently account for traditional rooted plant growth. Mycelia grow through non-traditional conditions such that the immediate environment around the spawns has to be maintained for optimal yields. The housing unit would maintain the necessary humidity levels through the use of sensors to monitor levels and give real time feedback. The apparatus itself would also ideally secure the substrate along the tubing in a way that it still receives moisture and is able to support the growing mushrooms. Along their fruiting stage, these heads for each mushroom require an opening big enough for them to then grow and begin exchanging carbon dioxide. In a natural setting, this would look like mycelia growing into mushroom heads through the bark of a tree.

Likewise, the amount and type of substrate material available for the mycelia directly correlates with the final yield. Therefore, it is important to both provide the proper moisture conditions in the surrounding environment and to provide enough substrate and housing space for the mushrooms to grow fully. A non-traditional design element for monotubs are drilled holes within the tub. The openings provide two different benefits to the system; one of which is airflow, and the other is an opening by which mushrooms can grow through. By adding these openings to a potential housing design, the fruiting stage would have space to grow through.

3.0 Summary & Conclusions

3.1 Lessons Learned from Review

In summary, there are three major constraints to the design of an Alternative Fruiting Chamber: mimicking microgravity, predicting moisture levels, and maintaining environmental conditions. Of these three major constraints, there are three sub parts of the whole AFC that can be focused on optimizing to account for the listed constraints which are, an adjustable stand, the porous tube, and the casing for the environment. We understand that for this AFC to be successful, it must operate in a microgravity environment, which changes the behavior of water that we are used to on Earth.

By making the stand adjustable, we are allowing for the optimization of height for growing the mushrooms. Making the design of the stand available for 3D printing, we negate the need for shipping requirements, but add the time and accuracy for a proper set up. The porous tube is the most variable part of our setup as it includes the need for a strong understanding of how the listed variables in Table 4 interact with each other, the substrate, and the mushrooms. The goals obtained from this portion include a relationship to determine the amount of water needed for specific conditions. A housing section will be required to maintain the level of humidity, light, and temperature of the mushrooms for optimal growth as well. Being able to keep these environmental factors in a preferred range will allow for a harvest that will satisfy the needs of the astronauts.

Finally, an understanding of the substrates used to promote the growth of oyster mushrooms or the chemicals needed for optimal growth was researched. While oyster mushrooms can grow on a few substrates as talked about in section 2.1.3, there are preferred substrates that promote faster mycelium colonization.

Together with an adjustable stand, a porous tube, and a casing for our mushroom environment, we will create an advanced plant habitat that can handle the constraints of a microgravity environment, fluctuating moisture levels, and varying environmental conditions to produce fresh grown mushrooms and take a step towards self dependence on the ISS.

3.2 Topics for Further Review

Some topics that need further research and testing include the requirement of a passive method of pressure application to a water reservoir. There will also be a requirement on instruction for a proper set up of the stand in the environment. On the topic of the porous tube, there needs to be further review into a decision matrix to determine the most important factors within the system. Through thorough testing of these factors, we will be able to give an accurate system of determining the optimal growth conditions. This also applies to the housing apparatus. Further testing will be necessary to determine the best ranges for humidity, type of light, and temperature range. This will be difficult to determine without the proper equipment and time, but we plan to at least give a basis to which others can build from. A final topic for further review involves substrate for growth. While there are many articles on the best type of substrate, we understand that access to these materials on a space station is not practical. Research and testing will have to be done with known substrates that can work with the design set up.

3.3 Global Context & Contemporary Issues

Although the team is designing the PPTNDS for NASA's use in microgravity, aspects of the design can have very important applications on earth as well. According to the United Nations, current global issues, among others, involve water, food, climate change, gender equality, population, health, and peace and security (United Nations, 2017). When considered in a global context, the development of a passive porous tube nutrient delivery system has the potential to help alleviate some of these injustices and environmental stressors today, on earth, and intergalactically in the future.

Populations that are most affected by water issues include those whose livelihoods depend on agricultural practices; more specifically those who are obligated to seek and gather water sources. As discussed in the article “building equality on the roof of the world” (UN Development Programme, 2019), it is often women who assume the important duty of retrieving water when the small local supplies run out, as is the case in the Pakistan village of Siska. By doing so, the women report missing out on cultural events in the village. Additionally, because the crops are watered at night to retain maximum water, the women often lose sleep. The solution enacted by the UN in Siska was to redirect glacial water to a holding tank in the village via pipeways. While this solution is beneficial for improving the villagers' access to water, it does not completely solve issues pertaining to gender equality and food security because the women must still spend large amounts of time and energy watering the crops at night.

In a village like Siska where water and sources of electricity are not plentiful, a passive porous tube nutrient delivery system could automate agricultural watering systems, making them more efficient in water use and improving crop yield. This concept is further supported by the study outlined in section 2.1.2 which measured the success of a passive porous clay tube irrigation system in India. Researchers Neelkanth and Gatesaniya attributed the improved crop yield largely to the fact that the irrigation system delivered water directly to the roots of the plants. If designed properly, both the placement and amount of water by our PPTNDS should be highly controllable and ideal for implementation in agriculturally based societies in need.

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5.0 Appendix

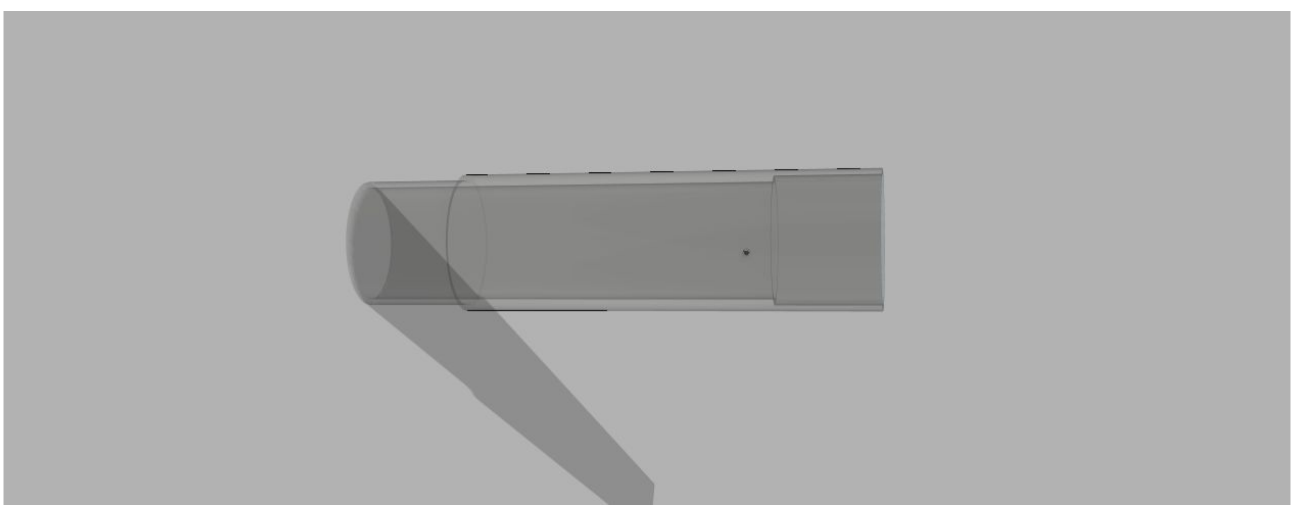
5.1 Brainstorming Results

The team's design solutions ultimately all derive from brainstorming. The method in which we brainstorm is collaborative, but also fosters a judgement free environment conducive to individual thought, thereby producing many good ideas that can be researched and evaluated further by subteams. Some of the results of our brainstorming include the following:

Related to meeting moisture requirements: some of the ideas we talked about include varying the composition of nutrients that are delivered and using liquid carbon to optimize growth without a solid substrate. One constraint to consider is the availability of tubing that can sustain both the myelia growth and liquid carbon delivery. Our idea for using liquid Carbon is tricky to put into practice because, unlike root plants, it may be hard to grow the mushrooms directly on the tubing. That said, it is an idea we will continue to look into incorporating into the final design.

Related to mimicking microgravity: some ideas include a stand that can be taken apart and put back together at the desired height like LEGO blocks. Another idea included suspending the PPTNDS with strings from the top of the housing. It could be on a pulley system to adjust the height, however this would not work in a microgravity setting. Ideas related to the pressure system include a rubber band, however as the reservoir loses water, the band will become loser, thus defeating the purpose. Ideally, we could utilize the adjustment of the stand to adjust the pressure setting, however there may or may not be a correlation between needed stand height and required pressure setting.

Related to maintaining environmental conditions: The main goal with any housing containment unit around the PPTNDS and substrate is to maintain the environmental conditions surrounding the pipe. Likewise heat exchange requires airflow so there is this balance point to reach where relative humidity is stable, and air flow is consistent. The design also has to follow guidelines to be minimal, and less invasive while also providing enough complexity to harvest the mushroom, and replace drained substrate manner. Initial ideas were to wrap the pipe with a secondary plastic tube, or create a box-like compartment around the whole unit. Further ideas involved the use of a premade unit that could be fitted into or around the pipe. Such a premade unit would hold mycelium spawn and substrate matter as well as have pore openings for heat exchange. This form of housing apparatus would make it manageable to continuously cycle through substrates while gathering fruiting mushrooms. Part of the investigation requires further understanding of spores, and filtering them out. Since the ISS plant habitat areas particle on contamination, it’s important to manage potential spore release. Spores can be collected for future spawn growth inorder to continue the growth cycle. Pore opening around the housing can be wrapped in an extra film material that can then be scrapped for spores. Current CAD designs include the implementation of a pipe casing around the PPTNDS with point of entry for microcontroller units, and potentially airflow.



Preliminary Cylindrical Housing with Humidity Monitor Chamber

5.2 Challenges and Mitigation

One challenge the team faces is limited access to KSC's systems and equipment. Any system we set up, prototype, and test will be according to dimensions provided by NASA and parts available to us through local resources. For example, the PPTNDS reservoir bag is made from the ISS Forward Osmosis Bag, which is not commercially available. KSC is unable to send us one, thus we will have to find a suitable replacement for the part should we choose to do testing on our own. Complications caused by this challenge can be mitigated and overcome in-part by constant and clear communication with our mentor, Jacob Torres. For instance, we have already talked to Torres about suitable reservoir alternatives that work similarly to the ISS bag and will allow us to test in a comparable manner. Prototype CAD drawings may also be sent to KSC for testing in their lab. Design improvements and recommendations can then be sent back to the team for adjustment.

Challenge associated with stand: The largest challenge facing the stand can be seen in the Tube Diameter vs. Water Height graph on page 14, where we discovered that even for a porous tube with an inner diameter of ⅛ of an inch, the water height is still not able to reach above 1 centimeter for successful capillary flow in 1g conditions. Originally, Jacob had indicated that he would like the stand to be 5-10 inches. So this drastically changed our ideas for stand design, and limited our options. One idea to help address this problem is to design a mechanism that exerts a constant pressure on the ISS reservoir bag, whether it is full or nearly empty. The mechanism would ideally keep the system passive, and not exert more pressure on a full bag compared to when the bag is nearly empty. Additionally, this would likely be built into the stand system itself.

Challenge associated with model: As is the case with most mathematical models, the largest challenges we face are finding equations to base our model off of and weighing each variable appropriately. These aspects are further complicated by the fact that in the end, we hope to make the model easily adaptable for predictive use in microgravity situations. To lessen the impact of these challenges, extensive and thorough research into studies that use modelling with similar capabilities is being done by the team. Additionally, we hope to ensure the validity of the model by working closely with our mentors.

Challenge associated with housing: The main challenge faced when designing the housing is keeping a completely isolated environment for both the humidity and ventilation, along with an easy method of harvesting and nearly completely passive monitoring system. These four factors create one big challenge and all factors must be considered when developing the housing, if one is left out then the overall ergonomics and viability of the design are greatly reduced. Another problem is keeping the housing enclosed around the substrate, but also providing enough flexibility to remove the house and replace drained substrate matter or harvest fruiting mushrooms. The ideal design goal would be to incorporate replaceable units for substrates, such as prepacked substrates, and spawn.

5.3 Project Timeline

September
(9/6) Project Selection
(9/6) Strengths Finder and Discussion
(9/20) Problem Statement
(9/27) Gantt Chart

October
(10/4) Website Outline
(10/9) Design Standards Research
(10/25) CATME #1

November
(11/2) Compile Technology Review Options
(11/20) Timesheets
(11/20) Notebook Check
(11/20) Contemporary Issues and Engineering Design Assessment
(11/20) CATME #2
(11/30) Technology Review

December
(12/4) Design Competition Selection
(12/10) Advisor Evaluation
(12/10) Website Updates
(12/17) Analysis of Potential Solutions/Alternatives
(12/17) Cost Analysis of Supplies
(12/17) Create Decision Matrix of Design Potentials

January
(1/1) Start CAD models for Stand, Housing
(1/1) Narrow down variables for model
(1/15) Start MATLAB code/algorithm for model
(1/26) Progress Report Presentations

February
(2/10) Document Organization/Compilation
(2/15) Prototype/analyze designs and make adjustments
(2/22) Report Outline

March
(3/1) Complete 25% of Report
(3 /8) Complete 50% of Report
(3/22) Complete 75% of Report
(3/26) Draft of Report

April
(4/29) Poster Submission

May (5 /8) Presentations (5/10) Final Reports

5.4 Team Member Responsibilities

The team has been broken into sub-teams as seen below. Members work in their sub-teams to discuss and develop design alternatives. Next semester, members will work in their teams to design, prototype and test their components. Throughout the duration of the project however, members may work across multiple teams and will contribute to full-team discussions and will work together to integrate their sub-team components into the full system. All members will help complete class deliverables.

Housing Apparatus for Maintaining Environmental Conditions:
Simon: CAD models, Constraints and Criteria for Housing, Research into humidity effects and maintenance, adding to content and edits, and appendices

Refat: Substrate considerations research, evaluated applied technology regarding plant housing, evaluation of mycology and optimal methods for mycelium growth, organized potential micro-controller units to monitor humidity

Stand for Mimicking Microgravity:
Tom Kasputis: Design considerations and alternatives, standards, evaluation of current technology, edits to the graph for the stand height based on porous tube diameter, looking into porous tubes, criteria, some NASA research on microgravity

Morgan: Act as a liaison between team and NASA, organize document and write content, act as technical expert on KSC crop science lab and PPTNDS

Robert: Source evaluation, determining and gathering of relevant information from sources, capillary action calculations, capillary action graph creation, design alternatives and consideration of additional design desires, and cover letter.

Pore Size and Tube Geometry Model for Predicting Moisture Levels:
Emma Givens: Researched and wrote on effect of porosity on water delivery and relevant units/equations (understanding moisture requirements, constraints, brainstorming), researched and wrote global context/contemporary issues section, edited paper for grammar, style, readability.

Morgan Re: Write content, conduct literature review, organize document, act as technical expert on PPTNDS and system functions

Christian White: Citations, overall organization of report, grammatical editing, lessons learned, and topics we need to review