

BME Design-Fall 2019 - XAVIER FAN

Complete Notebook

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Team contact Information

• Tianxiang Zhu • Sep 13, 2019 @02:12 PM CDT

Last Name	First Name	Role	E-mail	Phone	Office Room/Building
Kinney	Millessa	Advisor			
		Client			
		Leader			
		Communicator			
		BSAC			
		BWIG			
		BPAG			



Project description

• John Puccinelli • Aug 14, 2013 @12:01 PM CDT

Course Number:

Project Name:

Short Name:

Project description/problem statement:

About the client:



2019/09/11-Client Meeting 1

• XAVIER FAN • Sep 15, 2019 @10:56 PM CDT

Title: Client Meeting

Date: 2019/9/11

Content by: Xavier Fan

Present: Xavier, TShawn, Yanbo, Salina, Courtney

Goals: To have some introductory information about this project

Content:

Required functionality:

- at least three holes, including one for seed, one for inlet and one for outlet
- device has space to storage either liquid medium or both liquid medium and sand
- device has large enough clear view for inspection under microscope
- device needs to have the ability of being dessembled after experiment for sample extraction
- device might need to be reusable

Suggested improvements:

- chamber of device won't cluster with sands at inlet port
- adding one more hole with bacteria for competing experiment

Conclusions/action items:

We have gained a genral idea about our client's vision for this device and we will start our preliminary research upon that. Also, our communicator has sent an email regarding our request to get a copy of their experiment protocol so that we can make our design more realistic and client-oriented.



2019/09/26-Client Meeting 2

- Tianxiang Zhu - Oct 09, 2019 @12:37 PM CDT

Title: Client meeting 2

Date: 2019/09/26

Content by: TShawn

Present: Team

Goals: Show our preliminary designs to our client and listen to the suggestions and ideas from client

Content: Meeting notes

Observe from below
 Small volume → can see root closely

- observe how bacteria colonize the plane root. *bacteria need to interact with sand & root.*
- set. with lid → leave proof.

May need to

- *extract a bit to do mass spec.*
- *still need base in the press to locate the bacteria*

Root between base & bottom,

look through the fabrication case

Conclusions/action items:

After talking to the client, we realized that the inspection should be performed from the below instead of the top. Thus, we need fewer layers at the bottom and a shallower inner chamber to provide a clearer view of plant roots and bacteria.



2019/09/13-Advisor Meeting 1

- XAVIER FAN - Oct 09, 2019 @03:55 PM CDT

Title: Advisor Meeting

Date: 2019/9/15

Content by: Xavier Fan

Present: Xavier, TShawn, Yanbo, Courtney, Salina

Goals: To report our first client meeting and our progress on this project

Content:

During meeting:

- we reported information learned from our first client meeting
- we brainstormed a little bit about our preliminary design

Suggestions from advisor:

- we should update our problem statement weekly on progress report so that we will have a clearer objective along the way
- we should send an email regarding requirements for materials we use for the device

Conclusions/action items:

We have gained precious suggestions from our advisor to make our progress more organized and traceable. Our communicator will sent out an email about requirements for materials soon so that we can bring that into our concerns during the process of designing.



2019/09/20

- XAVIER FAN - Oct 09, 2019 @03:57 PM CDT

Title: Advisor Meeting

Date: 2019/9/20

Content by: Xavier Fan

Present: Xavier, TShawn, Yanbo, Courtney, Salina

Goals: To report our second client meeting and our progress on this project

Content:

During meeting:

- we reported our progress on PDS
- we asked for suggestions about PDS

Suggestions from advisor:

- we should add specific dimensions into PDS
- we should try to make our numbers more quantitative
- we should try to make weekly accomplishments more specific
- we should try to have more specific goals

Conclusions/action items:

We have gained precious suggestions from our advisor about PDS and we will try to edit it based on them.

**2019/09/26**

- XAVIER FAN - Oct 09, 2019 @03:54 PM CDT

Title: Advisor Meeting**Date:** 2019/9/26**Content by:** Xavier Fan**Present:** Xavier, TShawn, Yanbo, Courtney, Salina**Goals:** To report our third client meeting and our progress on this project**Content:**

During meeting:

- we reported our progress on preliminary designs
- we asked for suggestions about preliminary designs

Suggestions from advisor:

- we should look into cost of injection molding
- we should try not to open the lid during experiment
- we should try to research on how to change the direction of plant growth
- we should try to invite our client to come on presentation

Conclusions/action items:

We have gained precious suggestions from our advisor about preliminary designs, so that we can possibly make our final design more viable.



Title: Microbial Microcosm

Date: 9/10/2019

Content by: Xavier Fan

Present: Xavier Fan

Goals: To find a brief introduction about microcosm

Content:

In the first two billion years of life on Earth, bacteria – the only inhabitants – continuously transformed the planet's surface and atmosphere and invented all life's essential, miniaturized chemical systems. Their ancient biotechnology led to fermentation, photosynthesis, oxygen breathing, and the fixation of atmospheric nitrogen into proteins. It also led to worldwide crises of bacterial population expansion, starvation, and pollution – long before the dawn of larger forms of life.

Bacteria survived these crises because of special abilities that other life forms lack and that add whole new dimensions to the dynamics of evolution. First, bacteria routinely transfer their genes to bacteria very different from themselves. The receiving bacterium can use the visiting, accessory DNA (the cell's genetic material) to perform functions that its own genes cannot mandate. Bacteria can exchange genes quickly and reversibly. Unlike other life forms, all the world's bacteria have access to a single gene pool and hence to the chemical prowess of the entire bacterial kingdom.

This extreme genetic fluidity makes the very concept of *species* of bacteria meaningless. The result is a planet made fertile and inhabitable for larger life forms by a worldwide system of communicating, gene-exchanging bacteria.

Bacteria also have a remarkable capacity to combine their bodies with other organisms, forming alliances that may become permanent. Fully 10 percent of our own dry weight consists of bacteria, some of which – like those in our intestines that produce vitamin B12 – we cannot live without.

Mitochondria live inside our cells but reproduce at different times with different methods from the rest of our bodies' cells. They are descendants of ancient bacteria. Either engulfed as prey or invading as predators, these bacteria took up residence inside foreign cells, forming an uneasy alliance that provided waste disposal and oxygen-derived energy in return for food and shelter. Without mitochondria, the nucleated plant or animal cell cannot breathe and therefore dies.

This symbiogenesis, the merging of organisms into new collectives, is a major source of evolutionary change on Earth. The results of these first mergers were protoctists, our most recent, most important – and most ignored – microbial ancestors. Protoctists invented our kind of digestion, movement, and our tactile and visual systems. They came up with speciation, cannibalism, genes organized on chromosomes, and the ability to make hard parts (like teeth and skeletons). These complex microscopic beings and their descendants even developed the first genders and our kind of cell-fusing sexuality involving penetration of an egg by a sperm.

Discovering the microcosm within and about us changes – indeed, reverses – the way we look at living things and picture their evolution on the planet. For instance, since all life on Earth evolved from bacteria, it makes more sense now to think of beetles, rose bushes, and baboons as communities of former bacteria and protoctists than as higher animals or plants.

Reference

<https://www.context.org/iclib/ic34/margulis/>

Conclusions/action items:

Now I have some general ideas about microbial microcosm and according to attributes I found, I can start looking for competing designs and pay attention to how they adapt to those attributes to get more insights.



Title: PDMS

Date: 9/19/2019

Content by: Xavier Fan

Present: Xavier Fan

Goals: To find appropriate material for making bottom of the device

Content:

PDMS is one of the most employed materials to mold microfluidic devices.

We describe here the fabrication of a microfluidic chip by soft-lithography methods.

- (1) The molding step allows mass-production of microfluidic chips from a mold.
- (2) A mixture of PDMS (liquid) and crosslinking agent (to cure the PDMS) is poured into the mold and heated at high temperature.
- (3) Once the PDMS is hardened, it can be taken off the mold. We obtain a replica of the micro-channels on the PDMS block.

Microfluidic device completion:

- (4) To allow the injection of fluids for future experiments, the inputs and outputs of the microfluidic device are punched with a PDMS puncher the size of the future connection tubes.
- (5) Finally, the face of the block of PDMS with micro-channels and the glass slide are treated with plasma.
- (6) The plasma treatment allows PDMS and glass bonding to close the microfluidic chip.

Benefits:

It is transparent at optical frequencies (240 nm – 1100 nm), which facilitates the observation of contents in micro-channels visually or through a microscope.

It has a low autofluorescence

It is considered as bio-compatible (with some restrictions).

The PDMS bonds tightly to glass or another PDMS layer with a simple plasma treatment. This allows the production of multilayers PDMS devices to take advantage of the technological possibilities offered by glass substrates, such as the use of metal deposition, oxide deposition or surface functionalization.

PDMS, during cross-linking, can be coated with a controlled thickness on a substrate using a simple spincoat. This allows the fabrication of multilayer devices and the integration of micro valves.

It is deformable, which allows the integration of microfluidic valves using the deformation of PDMS micro-channels, the easy connection of leak-proof fluidic connections and its use to detect very low forces like biomechanics interactions from cells.

It is inexpensive compared to previously used materials (e.g. silicon).

The PDMS is also easy to mold, because, even when mixed with the cross-linking agent, it remains liquid at room temperature for many hours. The PDMS can mold structures at high resolutions. With some optimization, it is possible to mold structures of a few nanometers.

It is gas permeable. It enables cell culture by controlling the amount of gas through PDMS or dead-end channels filling (residual air bubbles under liquid pressure may escape through PDMS to balance atmospheric pressure).

Reference

Elveflow. (2019). *PDMS: A review - Elveflow*. [online] Available at: <https://www.elveflow.com/microfluidic-tutorials/microfluidic-reviews-and-tutorials/the-poly-di-methyl-siloxane-pdms-and-microfluidics/> [Accessed 19 Sep. 2019].

Conclusions/action items:

I have already gained enough evidence in using PDMS as the bottom piece due to its biologically inert property and high transparency. I will start looking for a material for upper parts of the device.



Title: Polystyrene

Date: 9/19/2019

Content by: Xavier Fan

Present: Xavier Fan

Goals: To find a material for upper parts of the device

Content:

Polystyrene in Medical

Due to its clarity and ease of sterilization, polystyrene is used for a wide range of medical applications, including tissue culture trays, test tubes, petri dishes, diagnostic components, housings for test kits and medical devices. [1]

Polystyrene is typically (but not always) a homopolymer meaning that it is composed only of the monomer styrene in combination with itself. Depending on the type of PS it could be classified as a "thermoplastic" or a "thermoset" material. The name has to do with the way the plastic responds to heat. Thermoplastic materials become fully liquid at their melting point (210-249 degrees Celsius in the case of Polystyrene), but they begin to flow at their glass transition point (100 degrees Celsius for PS). A major useful attribute about thermoplastics is that they can be heated to their melting point, cooled, and reheated again without significant degradation. Instead of burning, thermoplastics liquefy, which allows them to be easily injection molded and then subsequently recycled. Thermoset plastics, by contrast, will not reliquify once they are "set" in solid form.

By contrast, thermoset plastics can only be heated once (typically during the injection molding process). The first heating causes thermoset materials to set (similar to a 2-part epoxy) resulting in a chemical change that cannot be reversed. If you tried to heat a thermoset plastic to a high temperature a second time it would simply burn. This characteristic makes thermoset materials poor candidates for recycling.

Polystyrene is most uniquely useful for its application as a foam. It is the runaway leader in the packaging industry but it also has a wide range of uses as a traditional plastic. At Creative Mechanisms, we have used Polystyrene in a number of applications across a range of industries. For many years Polystyrene, or as it is often referred to as just Styrene, was used as the go-to prototyping material - basically for the same reasons we now use ABS. It's inexpensive, readily available, white in color, and it glues, sands, cuts, and paints well. The "S" in ABS is Styrene. A lot of older engineers and designers who have been in the industry for a while will ask for a Styrene model when they're looking for a quick-down-and-dirty prototype. We still have a lot of sheets of Styrene in the shop at Creative Mechanisms. We will use them to make quick test models, paint samples, vacuum formed or thermoformed prototypes, or large models that can be created with flat sheets.

We have also seen PS used as a sort-of living hinge material (typically polypropylene fits best into living hinge applications). There are clear disposable PS containers (e.g. a hot dog container from WaWa or your neighborhood convenience store for those living outside the Northeast) that function like a clamshell with a hinge in the middle. The hinge in this case is a little different than your traditional PP living hinge. Typically the PS hinge is more of a series of bends that allow the clamshell to flex and open up. Whether it is technically a living hinge or not, it still works very well and can be easily thermoformed. [2]

Reference

[1] ChemicalSafetyFacts.org, "What is Polystyrene?: Uses, Benefits, and Safety Facts," *ChemicalSafetyFacts.org*, 17-Jun-2019. [Online]. Available: <https://www.chemicalsafetyfacts.org/polystyrene/>. [Accessed: 19-Sep-2019].

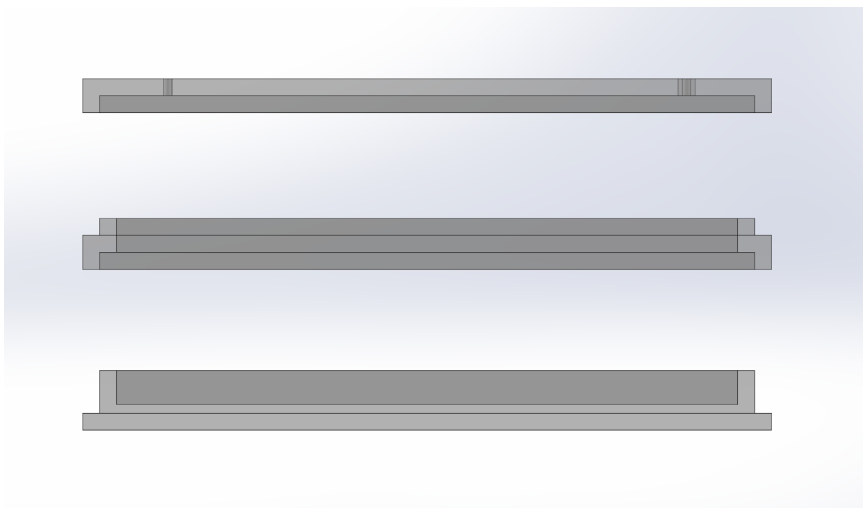
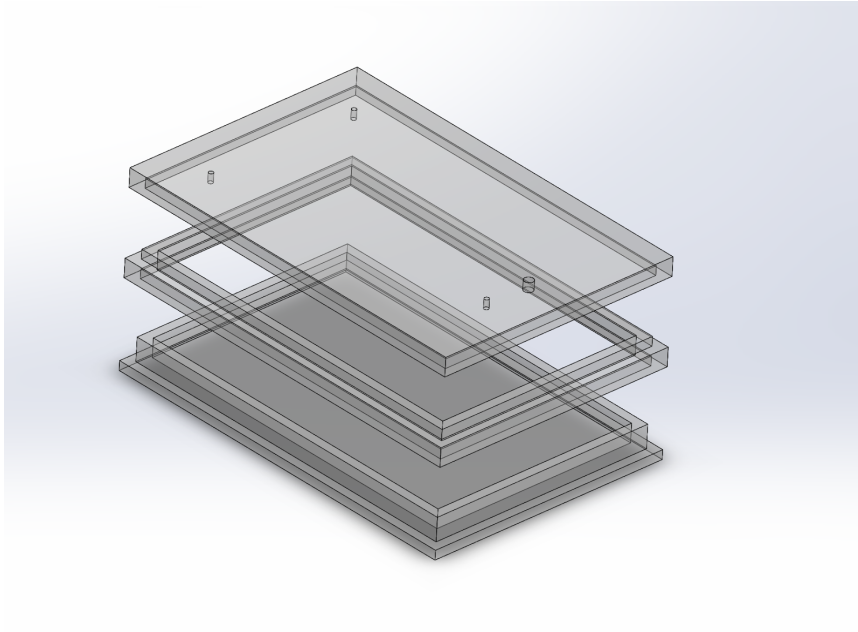
[2] T. Rogers, "Everything You Need To Know About Polystyrene (PS)," *Everything You Need To Know About Polystyrene (PS)*. [Online]. Available: <https://www.creativemechanisms.com/blog/polystyrene-ps-plastic>. [Accessed: 19-Sep-2019].

Conclusions/action items:

According to resources I found, polystyrene is a reliable material to be used in making this device. We will check with our client to make sure that our choices are viable.

 **Preliminary Design**

- XAVIER FAN - Oct 09, 2019 @04:00 PM CDT

Title: Preliminary Design**Date:** 2019/10/01**Content by:** Xavier Fan**Present:** Xavier**Goals:** To create a preliminary design**Content:**

This Modular design is composed of three parts, including a top cover with holes for loading, a middle optional layer for increasing the chamber volume, and a bottom layer for holding bacteria, root, liquid medium, and sand. The top cover has one hole for holding seed and three smaller holes for either loading bacteria or assisting airflow. The bottom layer is designed to be shallower so that root of the plant can be pushed against the bottom piece as close as possible and the middle optional layer can be put in between the other two layers to increase the chamber capacity by increasing the height if required. The bottom piece is designed to be made of very

transparent material, either glass or PDMS. The top cover and the optional layer can be made of polystyrene due to its biologically inert attribute

Conclusions/action items:

I will communicate with my teammates to try to modify this design later.



09/26/2019-THOR research paper from Handelsman Lab

▪ Tianxiang Zhu ▪ Oct 08, 2019 @12:12 AM CDT

Title: Introducing THOR, a Model Microbiome for Genetic Dissection of Community Behavior.

Date: 9/26/2019

Content by: TShawn

Present: TShawn

Goals: Get some background information from the client's lab

Content:

'The manipulation and engineering of microbiomes could lead to improved human health, environmental sustainability, and agricultural productivity. However, microbiomes have proven difficult to alter in predictable ways, and their emergent properties are poorly understood.'

The researchers found some connections and patterns of the microbiome community such as

Higher-order organization structures in an inhibitory interaction network of rhizosphere isolates.

B. cereus protects *F. johnsoniae* from *P. korensis* by modulating levels of koreenceine metabolites.

etc...

Conclusions/action items:

THOR is referring to The Hitchhikers Of the Rhizosphere.

The researchers developed a model community with representatives from three dominant rhizosphere taxa, the Firmicutes, Proteobacteria, and Bacteroidetes and their main goals are to study the manipulation and engineering of the microbiomes.

Reference:<https://mbio.asm.org/content/10/2/e02846-18>

▪ Tianxiang Zhu ▪ Oct 08, 2019 @12:13 AM CDT

View in :



09/26/2019 3D printing-Fabrication of PDMS apparatus

- Tianxiang Zhu - Oct 08, 2019 @12:21 AM CDT

Title: The upcoming 3D-printing revolution in microfluidics

Date: 9/26/2019

Content by: TShawn

Present: Tshawn

Goals: Get to know the fabrication method of the PDMS

Content:

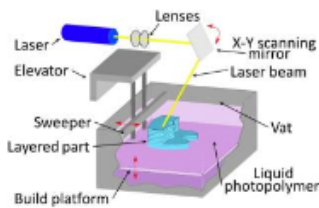
PDMS is a very promising biomaterial due to its properties of biocompatible, elastomeric, transparent, gas-permeable, water-impermeable, fairly inexpensive, copyright-free, and rapidly prototyped with high precision using simple procedures.

The authors of this article compared the salient features of PDMS molding with those of 3D-printing and they give an overview of the critical barriers that have prevented the adoption of 3D-printing by microfluidic developers, namely resolution, throughput, and resin biocompatibility.

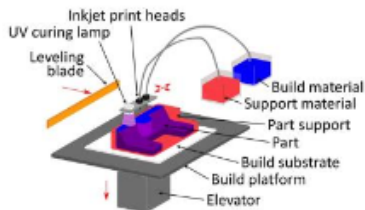
What is 3D printing:

“3D-printing” refers to a set of additive manufacturing techniques, which can create solid three-dimensional (3D) objects layer-by-layer under precise digital control.

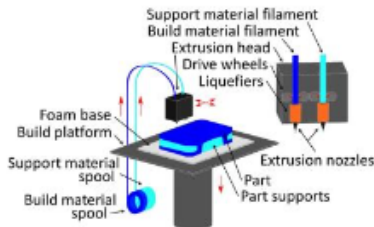
A. Stereolithography



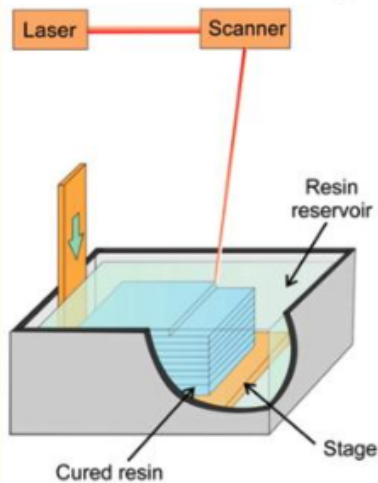
B. Multi Jet Modeling



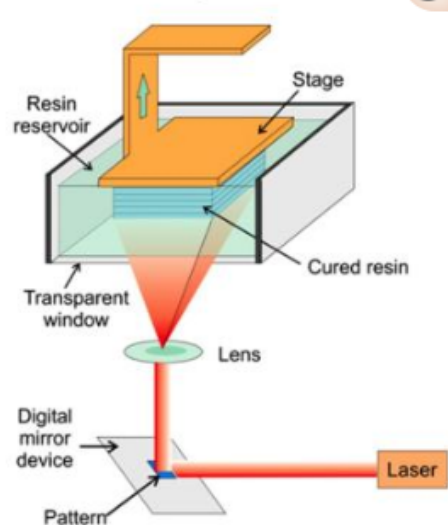
C. Fused Deposition Modeling



A. “Free Surface” Technique



B. “Bat” Configuration



Conclusions/action items:

Fabrication Methods Comparison:

	Soft lithography	Injection Molding	Paper Microfluidics	3D Printing
Setup Cost	~ \$80K ^a	> \$50K ^b	< \$1K	\$1K-20K
Cost per print/materials	High	Low	Low	High
Turn-around Time	~ 24 hrs	3 weeks ^c	< 2 hrs	< 2 hrs
3D Capability	Layered 2D designs	Layered 2D designs	Layered 2D designs	3D digital designs
Fluid Automation	Routine	Difficult	Rudimentary	Demonstrated
Throughput	Low	Very high	High	Medium
Manufacturability	Poor	Poor	Good	Good

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4862901/>



10/02/2019Permeability thickness dependence of PDMS

• Tianxiang Zhu • Oct 08, 2019 @03:49 PM CDT

Title: Permeability thickness dependence of polydimethylsiloxane (PDMS) membranes

Date: 10/02/2019

Content by: TShawn

Present: TShawn

Goals: Get to know the permeability of the PDMS Material

Content:

These articles mainly investigates the gas permeability of the PDMS membrane.

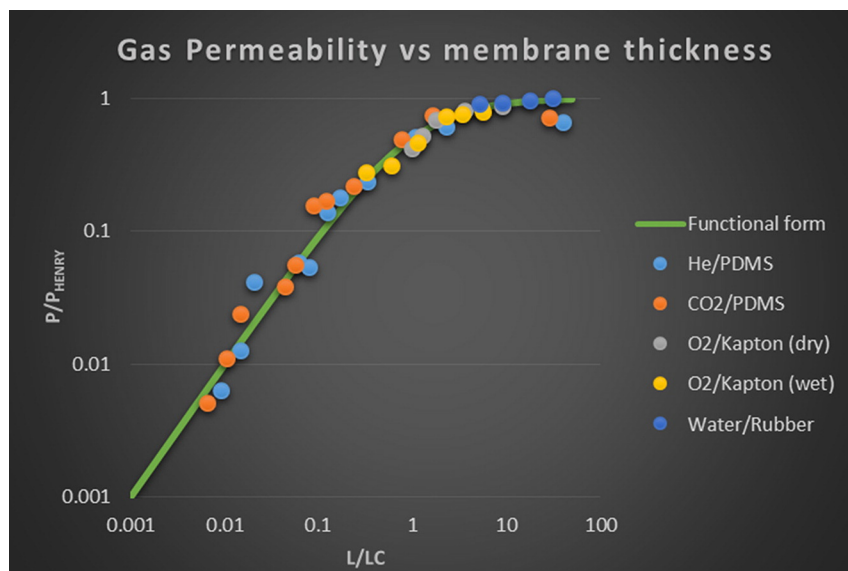


Figure 1[1]

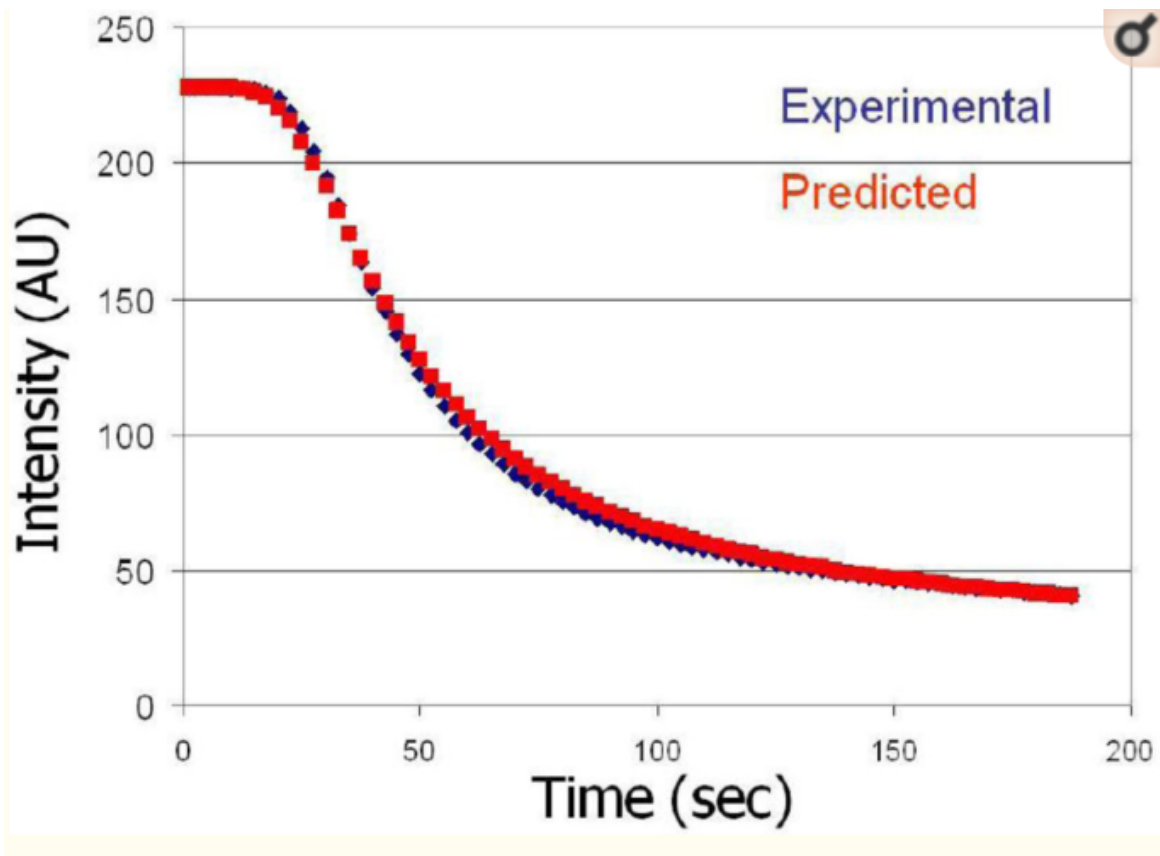


Figure 2[2]

Conclusions/action items:

According to the graph concluded from the paper, the oxygen and CO₂ have pretty high permeability on the PDMS membrane with thickness around 600nm.

O₂ and CO₂ permeability in our device (1mm PDMS) was tested by researchers in 'Variation in diffusion of gases through PDMS due to plasma surface treatment and storage conditions' as shown in figure 2

[1]<https://www.sciencedirect.com/science/article/pii/S0376738814009466#f0050>

[2]<https://ncbi.nlm.nih.gov/pmc/articles/PMC3945670/>



Title: Understanding Plant Tropisms

Date: 2019/10/07

Content by: TShawn

Present: TShawn

Goals: Get to know some basic tropisms of plant growth

Content: In an attempt to compensate for their sessile nature, plants have developed growth responses to deal with the copious and rapid changes in their environment. These responses are known as tropisms and they are marked by a directional growth response that is the result of differential cellular growth and development in response to external stimulation such as light, gravity or touch.

Conclusions/action items:

The direction of plant growth can be concluded to phototropism, gravitropism, thigmotropism, hydrotropism

Plant tropisms: providing the power of movement to a sessile organism

Int. J. Dev. Biol. 49: 605-614 (2005)
doi:10.1181/ijdb.0526036u

Plant tropisms: providing the power of movement to a sessile organism

C. ALEX ESMON, ULLAS V. PEDDALA and EMMANUEL LUSCUM*
University of Missouri-Columbia, Division of Biological Sciences, Columbia, Missouri, USA

ABSTRACT In an attempt to compensate for their sessile nature, plants have developed growth responses to deal with the copious and rapid changes in their environment. These responses are known as tropisms and they are marked by a directional growth response that is the result of differential cellular growth and development in response to an external stimulation such as light, gravity or touch. While the mechanics of tropic growth and subsequent development have been the topic of debate for more than a hundred years, only recently have researchers been able to make strides in understanding how plants perceive and respond to tropic stimulation. Thanks in large part to recent findings and recent advances in genomics, this paper focuses on the recent advances in how the level understood tropic responses and how such affects plant growth and development: phototropism, gravitropism, thigmotropism and hydrotropism. While progress has been made in deciphering the events between tropic stimulus and signal perception and such as molecular genetics responses, there are many areas that remain unclear, some of which will be discussed herein. As has become evident, such tropic response pathway exhibit a coordinating characteristic. However, these pathways of tropic perception and response also have overlapping components - a fact that is certainly related to the necessity for pathway integration given the ever-changing environment that surrounds every plant.

KEY WORDS: phototropism, gravitropism, thigmotropism, hydrotropism

When circumstances become unfavorable for optimal growth and development of animals, they can respond accordingly by moving to a more favorable environment. Plants are not afforded this luxury. Due to their sessile nature, plants are forced to make the most of their immediate surroundings, which means adapting to an ever-changing environment (Luscum, 2002). Darwin described some of these responses to environment some time a century ago in his book *The Power of Movement in Plants* (Darwin, 1880). Darwin noted that plants had a tendency to sense their environment so as to orient themselves for optimal growth and development.

Plants are constantly being bombarded with changes in their environment. Temperature fluctuations, poor light and low water content in the soil are just a few of the factors to which plants must be able to respond. Moreover, plants must respond to physical forces of nature such as gravity or touch stimulation. Over evolutionary time, plants have adapted to their surroundings with a high degree of plasticity, affording them the ability to respond to ever-changing conditions that provide constant stimulation. Plant tropisms are operationally defined as differential growth responses that orient plant organs in response to direction of physical

stimuli. Tropisms can be negative, such as a stem bending away from a gravity stimulus (Blaugauer and Mauson, 2003), or they can be positive, as in a stem bending toward a light stimulation (Luscum, 2002). Tropisms are different from basic plant movements, such as the diurnal movement of leaves on the opening and closing of flowers, in that basic growth is not directional in relation to a stimulus (Fridley, 1966). With tropic growth, the direction of the stimulus is very important.

Although it has been shown that each tropic response is governed by generally divergent genetic systems, it has become evident in recent years that at least some of the molecular features inherent to tropic responses may be shared. It is also apparent that different tropic response functions in coordinating and developing ways to give rise to adapt responses necessary for normal plant growth and development. So how are very different physical stimulations, or signals, perceived and responded to in such a way to yield outputs - differential growth responses - that are actually the same? As we see findings in recent areas of biology, nothing functions in vacuum. Much of the overlap has to do with the action of plant hormones and how such modulate cell growth. In each case it appears that it is the redistribution of plant hormones

*Address correspondence to: Dr. Emanuel Luscum, Division of Biological Sciences, 58B Life Sciences Center, University of Missouri, Columbia, MO 65211, USA. Tel.: +1-573-894-8876, e-mail: luscum@biology.missouri.edu
© 2005 by Int. J. Dev. Biol.
www.ijdb.com



Microfluidic co-cultivation platform to investigate microbial interactions

• Tianxiang Zhu • Oct 08, 2019 @12:23 AM CDT

Title: Competing Design 1

Date: 09/18-09/20/2019

Content by: TShawn

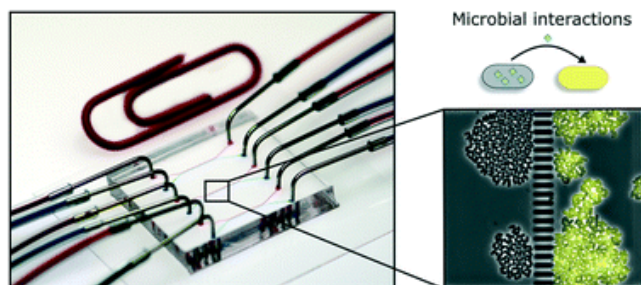
Present: TShawn

Goals: Getting to know a similar apparatus

Content:

This platform is aimed to investigate microbial interactions between two selected types of bacteria. The platform contains several microtubes that contain the culturing liquid while the different bacteria located on either side of the microtubes.

Conclusions/action items:



The presented device lays the foundation for further studies on contactless and contact-based interactions of natural and synthetic microbial communities.

<https://pubs.rsc.org/en/content/articlelanding/2019/lc/c8lc00977e#!divAbstract>



2019/09/11-Client Meeting 1

• Tianxiang Zhu • Sep 19, 2019 @10:07 PM CDT

Title: Follow-ups after the first client meeting

Date: 09/13/2019-09/15/2019

Record on 09/16-09/18

Content by: TShawn

Present: Team

Goals: Ask the client about the dimensions and the procedure of using the apparatus

Content:

During the client meeting, we talked through the basic functions of the apparatus and saw the prototype presented by Dr.Hurley. To further understand the structure and the functionality of the tool, I emailed the client for the experiment procedure and 3d modeling file. Dr.Hurley replied in 2 days and sent a detailed procedure to us with thoughts from Prof.Handelsman and her.

Conclusions/action items:

Here's the reply of our client attached

Dimensions of the Growth Chamber:

Square Base: 5 cm width x 7.6 cm length x 1 mm height

Square PDMS layer: 4.4 cm width x 6.8 cm length x 6 mm height

Oval Inner chamber: 5.4 cm width x 3.2 cm length x 3 mm height

Concluded from the file: The needs of our client are

- Change the growth chamber shape to square if possible
- At least one outlet pore for extraction of liquid and gas balance
- Detachable structures for bacteria and plant/sand extraction

9.19 update

added content to PDS under requirements

MICROCOSM FOR BACTERIA AND PLANT ROOTS
Hanselman Lab and BME Collaboration

Growth chamber dimensions:

Square Base 5 cm width x 7.6 cm length x 1.0 cm height
Square PDMS layer: 4.3 cm width x 6.8 cm length x 6 mm height
Drafflower chamber: 5.4 cm width x 3.2 cm length x 3 mm height

Growth chamber sand experiment:

1. Load chamber with 5 g sand via inlet hole. Wrap in tin foil and autoclave on a dry cycle.
2. Grow bacteria overnight. Determine bacterial concentration and dilute bacteria in fresh media at 10⁶ bacteria/mL.
3. Inoculate ~2 mL of bacteria from 1 mL pipette into the inlet.
 - a. I say ~2mL because a lot spilled out while I was normalizing.
 - b. I had to massage the sand to hydrate the bottom of the chamber.
 - c. Less sand may help with inoculation but I would worry the sand would not be static and would float in the liquid and disrupt any structures built by bacteria.
 - d. Possible options:
 - i. Include an outlet to suck out air and draw liquid down.
 - ii. Double chamber with cell-permeable membrane for equal inoculation along length.
 - iii. Include a "stopper" at the top to compress sand after inoculation.
 - iv. Is there any guys have more sand better ideas!
4. Cover inlet with breathable seal and place growth chamber in a plastic container with a loose lid. Include soaked paper towels in empty tip boxes to maintain humidity in the container.
5. Grow bacteria at 20°C for 2-5 days.
6. Every day, image bacteria at the base of the growth chamber on a Nikon microscope.
7. On that last day, use a razor to cut a horizontal line ~5-7mm below the inlet. Use the razor to separate the PDMS from the base. When open, use a sterile scoop to transfer sand into two eppendorf tubes.
8. Repeat step 7 down the chamber (twice) until there are 6 eppendorf of sand - two replicates each for top, middle, and bottom of the chamber (with respect to the inlet).
9. Weigh sand and add 1 mL liquid per 0.5 g to normalize volume harvested. Vortex and sonicate sand to remove attached bacteria. Perform serial bank dilutions and plate on appropriate antibiotics to quantify each species of bacteria.

[Microcosm_for_bacteria_and_plant_roots.docx\(120.9 KB\) - download](#)



Title: Literature Research Guideline

Date: 2019/09/16-09/19

Content by: TShawn

Present: TShawn

Goals: Come up with fields/contents to research

Content: According to client needs

Literature research can be attempted in:

- Material for growth chamber and base(Transparent, permeable to oxygen)
- Structure for pores and ports
- The manufacturing method for chamber, pores, and ports
- Distribution of bacterial community in sand/ on plant roots

Conclusions/action items:

Found articles attached

Lab on a Chip
PAPER
Check for updates
View Article Online
DOI: 10.1039/C9LC00183G

A microfluidic co-cultivation platform to investigate microbial interactions at defined microenvironments?

Alina Burmester,^{1,2*} Fabiana Häger,^{1,2*} Anika Langner,² Christoph Westendorp,^{1,2} Henric Nordhoff,^{1,2} Niklas Thielert,^{1,2} Thomas Drepper,^{1,2} Dietrich Hoffeyer,^{1,2} Eric von Lieres,^{1,2} Stephan Neack,^{1,2} and Alexander Gruber,^{1,2*}

Inter-species interactions inside microbial communities bear a tremendous diversity of complex chemical processes that are by far not understood. Even for simplified, often synthetic systems, the interactions between two microbes are barely revealed in detail. Here, we present a microfluidic co-cultivation platform for the analysis of growth and interactions inside microbial consortia with single-cell resolution. Our device allows the spatial separation of two different microbial organisms inside adjacent microenvironments, facilitating simultaneous exchange of metabolites via connecting microchannels. Inside the cultivation chambers, cell growth can be observed with high spatial-temporal resolution by time-lapse imaging. In contrast to conventional approaches, in which single-cell activity is typically fully masked by the average bulk behavior, the small dimensions of the microfluidic cultivation chambers enable accurate measurement of individual interaction of cellular interactions with full spatiotemporal resolution. Our method enables us to study site formation in microbial interactions, such as gene transfer or metabolic cross-feeding. We chose two different microbial model systems to demonstrate the wide applicability of the technology. First, we investigated commensalistic interactions between an industrially relevant *L. reuteri*-producing *Corynebacterium glutamicum* strain and an *L. reuteri* as probiotic partner of the same species. Spatially separated co-cultivation of both strains resulted in growth of the probiotic strain due to secreted lipoteichoic acid by the producer strain. As a second example, we investigated bacterial conjugation between *Escherichia coli* SH-7 and *Pseudomonas putida* ATCC 49619 cells. We could show that direct cell contact is essential for the successful gene transfer via conjugation and that bacterial strain cells were spatially separated. The presented device lays the foundation for further studies on contact- and contact-based interactions of natural and synthetic microbial communities.

Introduction
Microbial communities are complex heterogeneous systems with diverse metabolic activities and ecological dependencies. Many interactions inside these communities have not yet been analyzed in detail due to their complexity and missing experimental design for analysis. Inverse and interspecies microbial interactions are ubiquitous in nature and play a pivotal role in nearly every ecosystem. These interactions often function not only with each other but also with higher organisms. A well-known example is the symbiotic interaction of nitrogen-fixing bacteria with root nodules. These bacteria live inside the plant's roots and supply them with essential nitrogen, while they obtain carbon-phases from the plant in return.¹ Another evidence for the importance of interactions inside bacterial communities is carbon in the great discrepancy between the number of bacterial taxa present in an environmental sample and the number of bacterial species that can be cultured in vitro.² This unculturability of most bacteria is a consequence of a major problem for systematic studies of bacterial diversity,³ their resistance for the establishment of many laboratory strains is responsible for the missing knowledge on species interactions within communities.⁴ The metabolic

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RSC, 2019

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³ Leibniz Institute of Freshwater Ecology and Inland Fisheries (IGLE), 12585 Hohenheim, Germany
⁴ Institute of Microbiology and Food Technology, Technical University of Berlin (TU Berlin), Germany

Lab Chip, 2019, 19, 186–192
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[A_microfluidic_co-cultivation_platform_to_investigate_microbial_interactions_at_defined_microenvironmentspdf.pdf\(4.4 MB\) - download](#)

RESEARCH ARTICLE
Ecological and Evolutionary Science

Introducing THOR, a Model Microbiome for Genetic Dissection of Community Behavior

Gabriel L. Luzzatti,^{1,2} Juan I. Barua,^{1,2*} Manuel F. Garcia Diego,^{1,2} Hyun Song Park,^{1,2,4} Amanda Huber,^{1,2} S. Brook Peterson,^{1,2} Dik V. Stals,^{1,2} Jason W. Crawford,^{1,2,4} Michele A. Fedele,^{1,2} & Handelman^{1,2}

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5Department of Microbiology, The University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

ABSTRACT The quest to manipulate microbiomes has intensified, but many microbial communities have proven to be recalcitrant to sustained change. Developing model communities amenable to genetic dissection will undergo successful strategies for shaping microbiomes by advancing our understanding of community interactions. We developed a model community with representatives from three dominant phyla: *Proteobacteria*, *Firmicutes*, and *Bacteroidetes*. We chose *Acetivibrio* sp. as a model *Firmicutes* lineage and characterized 28 other conditions, including “killers” that coexist with it, culled from the *Acetivibrio* native analysis produced a hierarchical interaction-complexity network. We chose two *Bacteroidetes*, *Flavobacterium* lineage from the top tier of the complexity network and *Chloroflexum* lineage from the bottom of the network, to represent the *Proteobacteria* and *Bacteroidetes*, respectively. The model community has several emergent properties: induction of diazotic expression of *nif* genes occurs by either of the other members, and production of more robust biofilms by the three members together than individually. Moreover, *P. litoralis* produces a novel family of alkaloid antibiotics that inhibit growth of *P. litoralis* and production is inhibited by *A. acetivibrio*. We designate the community THOR, because the members are the grandchildren of the (N)Otophera. The genetic, genomic, and biochemical tools available for dissection of THOR provide the means to achieve a new level of understanding of microbial community behavior.

INTRODUCTION The manipulation and engineering of microbiomes could lead to improved human health, environmental sustainability, and agricultural productivity. However, microbiomes have proven difficult to alter in predictable ways, and their emergent properties are poorly understood. The history of biology has demonstrated the power of model systems to unlock complex problems such as gene expression or development. Therefore, a defined and genetically tractable model community would be useful to dissect microbial assembly, interactions, and processes. We have developed a tractable model diapaic base microbiome designated THOR, containing *Flavobacterium* lineage, *Flavobacterium* lineage, and *Acetivibrio* sp., which represent three dominant phyla in the diapaic base, as well as in soil and the mammalian gut. The model community demonstrates emergent properties, and its members are amenable to genetic dissection. We propose that THOR will be a useful model for investigations of community-level interactions.

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DOI: 10.1073/pnas.18101469

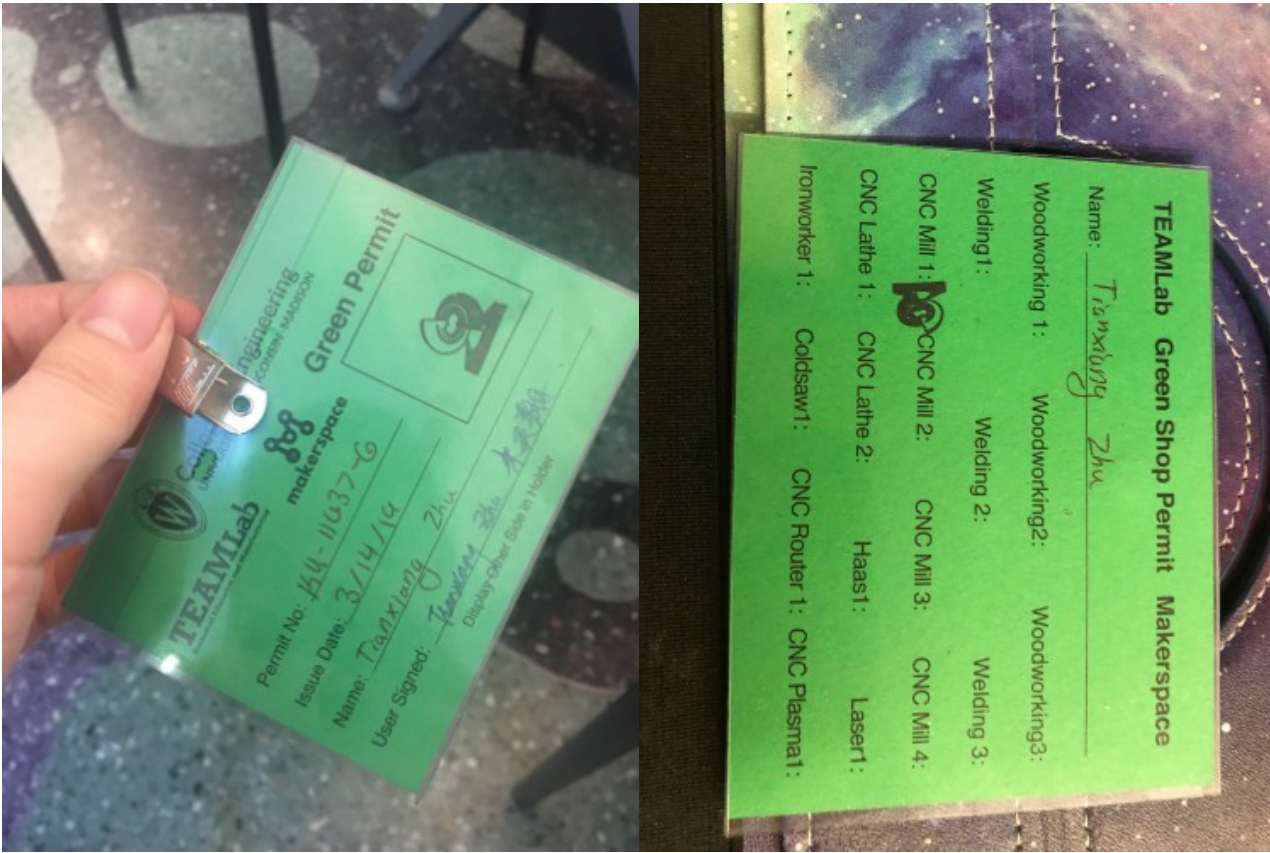
Downloaded from www.pnas.org on September 19, 2019 by guest

mBio-2019-Introducing_THOR_a_Model_Microbiome.pdf(3.3 MB) - download



Green permit and Upgrades

• Tianxiang Zhu • Oct 08, 2019 @02:09 PM CDT





- Tianxiang Zhu - Oct 08, 2019 @02:10 PM CDT

219K924 Biosafety Required Training Quiz Biosafety Required Training

Biosafety Required Training Quiz

Due No due date Points 25 Questions 25 Time Limit None
Allowed Attempts Unlimited

Instructions

You must complete the quiz with a passing score of 18 out of 25 questions correct (70%)
You may take the quiz more than once in order to achieve a passing score.
[After you submit the quiz and have a passing score, click here to finish the course.](#)
<https://courses.wisc.edu/courses/2457/tapep/2457-2019/>

Take the Quiz Again

Attempt History

Attempt	Time	Score
LATEST	Attempt 1	36 minutes 22 out of 25

Correct answers are hidden.

Score for this attempt: 22 out of 25
Submitted Mar 24 at 11:01pm
This attempt took 36 minutes.

Question 1 1 / 1 pts

Biosafety risk assessment takes into consideration the biological agent, the environment and the host.

True
 False

Question 2 1 / 1 pts

training_documents.pdf(6.2 MB) - download



Growth promotion of Xanthium italicum by application of rhizobacterial isolates of Bacillus aryabhattai in microcosm soil

- Yanbo (David) Feng - Sep 19, 2019 @10:50 PM CDT

Title: Growth promotion of Xanthium italicum by application of rhizobacterial isolates of Bacillus aryabhattai in microcosm soil

Date: 9/19/2019

Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Research some existing microcosm

Content:

In this research, the researchers studied the interaction between the Xanthium italicum and rhizobacterial isolates of Bacillus aryabhattai in the soil. This paper studies how the bacteria can promote the growth of Xanthium italicum. The study shows that isolate bacteria can facilitate the seed germination, length of roots, and shoot, allowing the plant with bacteria to grow better than the plants without the interaction from bacteria.

Conclusion:

This paper can be a reference for that the bacteria can promote the growth of bacteria. This conclusion can allow our team to figure out how to design the inner structure of the microcosm to allow the bacteria to stimulate the growth of root of our seed more effectively.

- Yanbo (David) Feng - Sep 19, 2019 @10:23 PM CDT



Growth_promotion_of_Xanthium_italicum_by_application_of_rhizobacterial_isolates_of_Bacillus_aryabhattai_in_microcosm_soil.pdf(260.9 KB) - [download](#)



Title: Plant Growth Promoting Rhizobacteria

Date: 9/19/2019

Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Research about the types of bacteria which can promote plant root growth

Content:

In this research, the researchers studied different types of bacteria that can stimulate the growth of plant root.

Conclusion:

This paper can be a source for our team to understand the types of bacteria that can possibly be the competing bacteria in the microcosm in the future.



Plant_Growth_Promoting_Rhizobacteria.pdf(71.1 KB) - [download](#)



Characterization of Polydimethylsiloxane (PDMS) Properties for Biomedical Micro/Nanosystems

• Yanbo (David) Feng • Oct 08, 2019 @10:36 PM CDT

Title: Characterization of Polydimethylsiloxane (PDMS) Properties for Biomedical Micro/Nanosystems

Date: 10/7/2019

Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Research the properties of PDMS under different conditions including sterilizing.

Content:

This work is a summary of various mechanical and material properties of PDMS under different experimental condition and with different chemicals.

Conclusion:

This work may be a very useful reference for our later testing part for our PDMS device.

• Yanbo (David) Feng • Oct 08, 2019 @10:28 PM CDT



[PDMSCharacterization-BiomedAppn.pdf\(937.6 KB\) - download](#)



A microcosm for raising plants under biotic and abiotic conditioning

• Yanbo (David) Feng • Oct 08, 2019 @09:56 PM CDT

A microcosm for raising plants under biotic and abiotic conditioning

Date: 10/7/2019

Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Research competing design

Content:

A microcosm for growing plants under biotic and abiotic conditions in order to obtain an environment in which to replicate on a laboratory scale what occurs under natural conditions in a plant or in a plant canopy when given environmental conditions are imposed and organisms intervene capable of interacting or interfering with the plant functions, said microcosm being constituted by an apparatus that comprises in combination two independent chambers, of which one is a hypogeal chamber (1) that forms the part of the apparatus or microcosm that is designed for growing the radical part of the plants and of the rhizosphere, and the other is an epigeal chamber (2) that forms the part of the apparatus or microcosm that is designed for growing the aerial part of the plants and of the phyllosphere, said chambers being assembled vertically on top of one another so as to constitute a single complex and functionally integrated structure.

Conclusion:

Unfortunately, I didn't find any PDF for this patent. As a result, the understanding about this design is just from the texts in Content.



Preliminary Design

- Yanbo (David) Feng - Oct 08, 2019 @08:44 PM CDT

Title: Preliminary Design

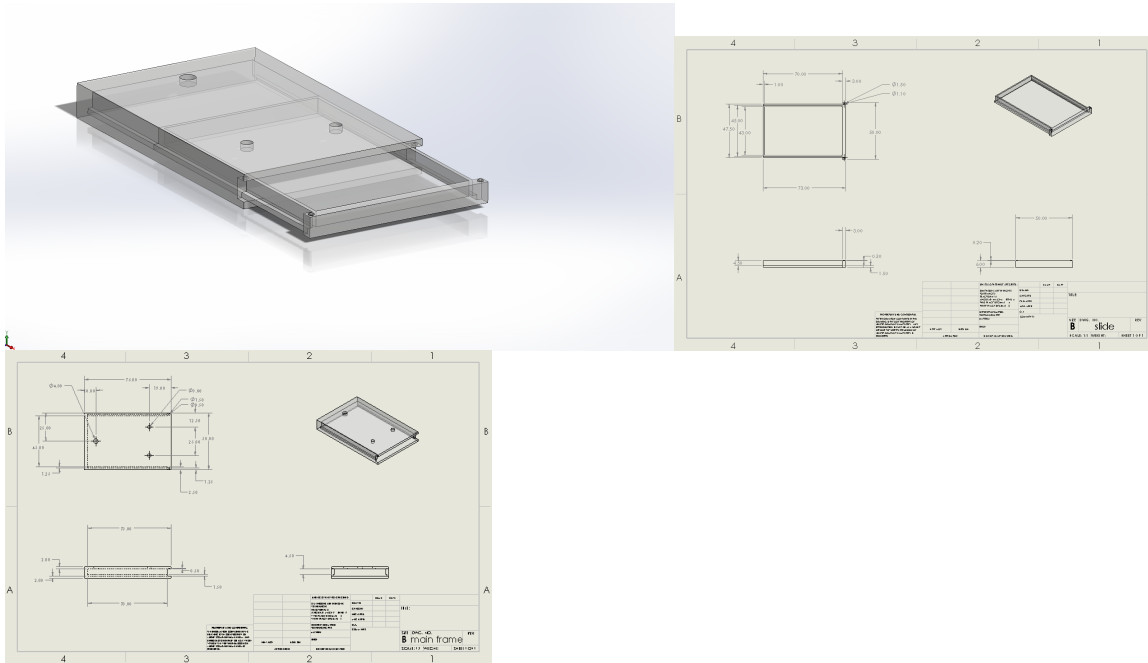
Date: 10/7/2019

Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Present and describe the Match Box design

Content:



This design contains two parts. The first part is the main frame, and the second part is the platform. The material for the main frame will be PDMS, which is biocompatible, oxygen permeable, and highly transparent. Its size is 76mm x 50mm x 10mm. The size of the device allows easy observation under microscope. On the top surface of the main frame there are three holes. The large hole has a diameter of 4mm, which is the inlet hole for seed settling and seed growth. The two small holes each has a diameter of 2mm. These two holes are the outlet holes for air flow and loading of bacteria into the device. The fixed positions of the outlet holes allow the researcher to load bacteria at fixed locations, which help keep the positions of bacteria for each experiment constantly and eliminate potential unexpected and uncertain variables. In the middle of the main frame, a chamber is designed to make sure that the platform can slide into the main frame and assemble to become the complete form of Match Box. The chamber is 70mm in depth. The height of the cavity is 6mm, which enables sufficient room for the growth of seed root, media, bacteria and sand. For the platform, the material for the slide in the middle will be 1.5mm glass slide. The material for other structure of polystyrene. The purposes of using different materials is that the glass slide has high transparency to enable clear microscopy and that the polystyrene can be more easily fabricated into detailed and complicated structures via milling than glass. The surrounding walls ensure leakproof and containment. The upper surface of the glass slide will be treated with a thin layer of PDMS for better biocompatibility. The Match Box can ease the loading process, culturing and microscopy to a large extent.

Conclusion:

This design is hard to be manufactured because the main frame part has relatively complex inner structure, which will make the injection modeling harder. Also, for the platform part, how to assemble glass slide and polystyrene together will be difficult during the manufacturing. Also, this design is not adjustable. As a result, this design needs simplification and addition of adjustability. Generally, this design is unique and effective.



Title: Green Pass

Date: 10/7/2019

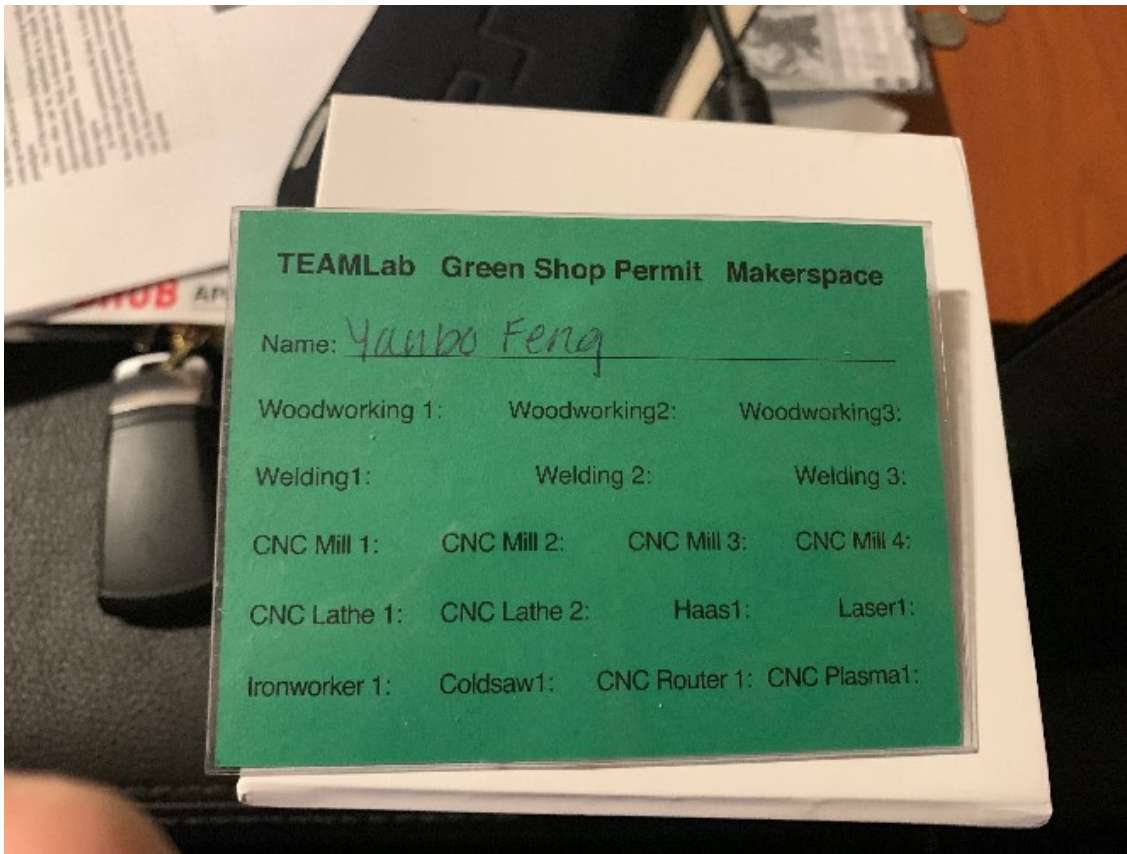
Content by: Yanbo Feng

Present: Yanbo Feng

Goals: Present Green Pass to Teamlab workshop machines

Content:





Conclusion:

The Green Pass prepare me for some possible manufactures later.



2019-09-09 First Meeting

- Yanbo (David) Feng - Oct 09, 2019 @10:23 AM CDT

Title: First Team Meeting

Date: 9/9/19

Content by: Yanbo Feng

Present: The whole team

Goals: Complete the first progress report

Content:

We discussed the work that each of us needs to complete. We also discussed that possible areas that we can do research to obtain information for the design process.

Conclusions/action items:

We will work on preliminary research.



2019-09-11 First Client Meeting

• Yanbo (David) Feng • Oct 09, 2019 @10:41 AM CDT

Title: First Client Meeting

Date: 9/11/2019

Content by: Yanbo Feng

Present: The whole team

Goals: Obtain some requirement information from the lab and take a look at the prototype that lab designed.

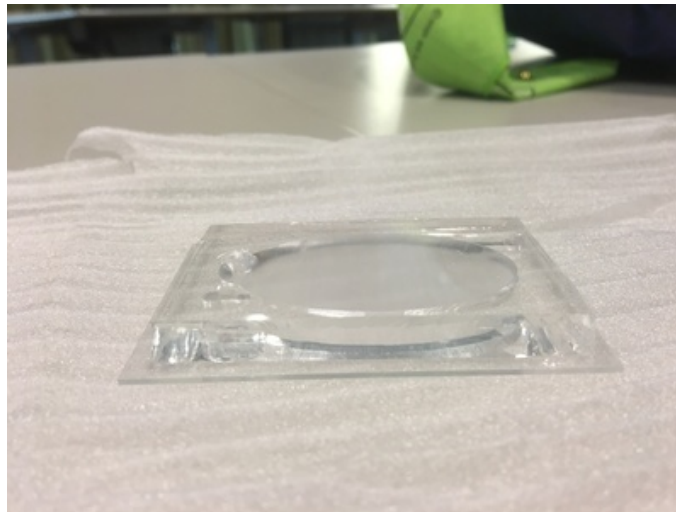
Content:

The lab expects a device made by highly transparent materials such as PDMS and glass. The size of the device needs to fit on the platform of the microscope in the lab well for inspection.

Conclusions/action items:

We need to brainstorm whether to modify the prototype or to design a new prototype.

• Yanbo (David) Feng • Oct 09, 2019 @09:55 AM CDT



IMG_7438.JPG(1.5 MB) - [download](#)

• Yanbo (David) Feng • Oct 09, 2019 @09:55 AM CDT



IMG_7437.JPG(2.1 MB) - [download](#)



2019-09-13 First Advising Meeting

- Yanbo (David) Feng - Oct 09, 2019 @10:46 AM CDT

Title: First Advising Meeting

Date: 9/13/2019

Content by: Yanbo Feng

Present: The whole team

Goals: Report and discuss the content of first client meeting with our advisor.

Content:

We reported the information from the client meeting to our advisor. One very good point our advisor gave us is that since the device needs to be able to be recycled, the impact that the autoclave on the PDMS needs to be evaluated.

Conclusions/action items:

We need to do more preliminary researches about PDMS and brainstorm the design ideas.



2019-09-25 Second Client Meeting

- Yanbo (David) Feng - Oct 09, 2019 @11:06 AM CDT

Title: Second Client meeting

Date: 9/25/19

Content by: Yanbo Feng

Present: The whole team

Goals: Present our preliminary designs to our client and get feedback for later improvement of the designs.

Content:

- Transparent glass or PDMS bottom is required for high resolution of microscopy.
- Adjustable chamber volume for multiple choices of chamber size.
- Extraction holes along the side of the top surface for extraction and observation of bacteria.
- The height should be able to be lead the root to grow along the bottom surface in the chamber.
- The height of the chamber needs to be 2-3mm for better microscopy.
- The PDMS is tolerant for autoclave for few times.

Conclusions/action items:

We need to combine the advantages from the designs and prepare for the presentation.



2019-09-26 Advising Meeting

- Yanbo (David) Feng - Oct 09, 2019 @11:19 AM CDT

Title: Advising Meeting

Date: 9/26/19

Content by: Yanbo Feng

Present: The whole team

Goals: Introduce the designs to our advisor and complete the design matrix.

Content:

We discussed that we need to do researches on the cost of materials such as PDMS and understand how root of plant would grow in a limited space

Conclusions/action items:

We need to complete the design matrix.



2019-10-03 Presentation Meeting

- Yanbo (David) Feng - Oct 09, 2019 @11:31 AM CDT

Title: Presentation Meeting

Date: 10/3/19

Content by: Yanbo Feng

Present: The whole team

Goals: To complete the slides and practice the presentation for Friday.

Content:

We assigned the different presentation parts to each team member. My assignment is the design part. I need to interpret each design to the audience during the presentation to allow the audience to understand what our expected final product looks like and the advantages of each design.

Conclusions/action items:

I need to practice the assigned part of the presentation to fluency.

Preliminary Design Idea

• SALINA LOER (loer@wisc.edu) • Oct 09, 2019 @01:23 PM CDT

Title: Preliminary Design

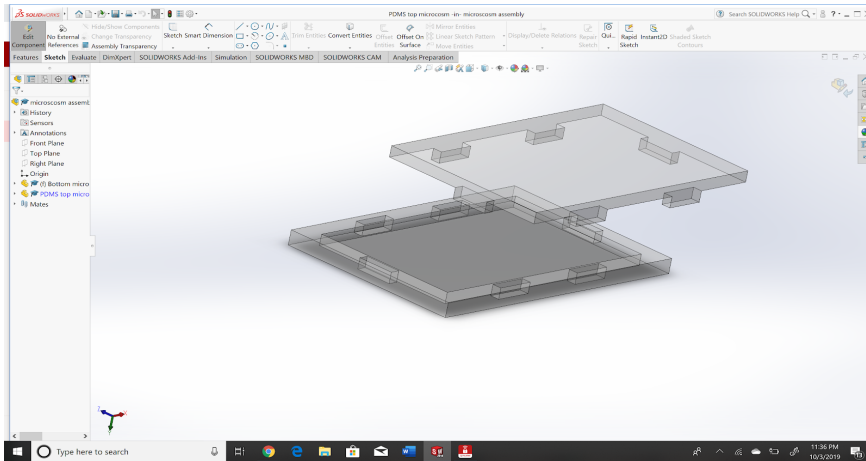
Date: 9/23

Content by: Salina Loer

Present: Salina L

Goals: Create a preliminary design idea for the microcosm to bring to team meeting

Content: Each team member has to create a preliminary design idea for the microcosm and we will bring them all to the team meeting and review and decide which ones to use in the design matrix that we will begin this week. My plan was to use the basic outline of the prototype created in Handelsman's lab and make it into two pieces that could be separated to load the device and then closed and sealed for the experiment to take place. I thought of using PDMS for the top part and glass for the bottom.



Conclusions/action items:



Team Meeting 9/9

- SALINA LOER (loer@wisc.edu) - Sep 09, 2019 @05:23 PM CDT

Title: Team Meeting

Date: 9/9

Content by: Salina Loer

Present: Salina L, Courtney M, Xavier F, TShawn Z, Yanbo F

Goals: First progress report and set up meeting with client

Content: We worked on the first progress report as well as when we would meet with our client Prof. Handelsman, which is September 25th at 3:30. We discussed when we could meet as a group to work on the project. We also emailed the client and asked if we could get information regarding the project and their initial prototype before meeting with them.

TShawn set up a google team document for the project, which we will write progress report and other documents in.

Conclusions/action items: Research project and ideas for preliminary design and finish the progress report for the week



Client meeting 9/11/23

- SALINA LOER (loer@wisc.edu) - Sep 23, 2019 @05:46 PM CDT

Title: Client Meeting

Date: 9/11/2019

Content by: Salina Loer

Present: All team members

Goals: Discuss the prototype they have with Dr. Handelsman's postdoc researcher Amanda Hurley

Content: We met with Dr. Hurley in the Wisconsin Institute for Discovery and she showed us the prototype they had come up with. We discussed what they wanted from the device and the current flaws with the one they have, which are:

- Is not reusable, it had to be cut open to study the bacteria structures
- Was hard to load with sand and media
- Was not easy to examine the bacteria structures without disturbing the sand too much
- No good way to remove the sand and media from the device

They want to use the prototype to study bacteria structures on sand and different bacteria's affinity for plant roots that they grow in the device

Conclusions/action items: Dr. Hurley will send us the specifications for their prototype and the procedure she uses for the experiments. We will begin thinking of preliminary designs and research more on the concept.



Team Meeting 9/23/2019

- SALINA LOER (loer@wisc.edu) - Sep 23, 2019 @05:39 PM CDT

Title: Team Meeting

Date: 9/23/2019

Content by: Salina Loer

Present: Salina L, Courtney M, Xavier F, Tshawn Z

Goals: Review design ideas, begin design matrix and think of questions for client meeting

Content: We each came to the meeting with our specific design ideas, either on paper or in solid works. We compared all of our designs so we could create one design idea to start our design matrix on and bring to our client meeting. Tshawn and Xavier had already created their designs on solidworks and Courtney and I had ours drawn on paper so we will create them in solidworks in the next few days. We decided on a time to meet with our advisor on Thursday and created a document to write down questions for Dr. Handelsman during our client meeting on Wednesday.

Conclusions/action items: Over the next few days with will finish our 3rd progress report and I will upload it to the website. We will meet with our client and then advisor and then finish the design matrix which i will also upload to our website. Courtney and I need to create our designs on solidworks.



Client meeting 9/25

- SALINA LOER (loer@wisc.edu) - Sep 25, 2019 @04:22 PM CDT

Title: Client Meeting

Date: 9/25/19

Content by: Salina Loer

Present: All team members

Goals: discuss preliminary designs with our client and ask any questions

Content: Prof. Handelsman was unable to come to the meeting so we showed our preliminary designs to Dr. Hurley

- Can't use polystyrene for bottom layer, must image from bottom so see through-glass.
- Potentially customizable chamber size- shorter width and larger bottom would be preferable
- PDMS can be autoclaved atleast once- client would expect to make more than one device
- alfalfa seeds would potentially be used, could fill chamber up high enough that seed would not be able to move
- likes lid idea- leak proof
- not sure what to do about inlet holes, might need a way to plug them
- height is the only thing they would want to be variable
- more holes to be able to take samples throughout chamber
- potentially want lid that would push roots closer to bottom if microscope cannot focus on them
- doesn't have to be reusable if device is cheap
- chamber could be 2 or 3 mm thick- maybe shorter walls would be better and then different lids that are taller or shorter

Conclusions/action items:



Team meeting 9/26

- SALINA LOER (loer@wisc.edu) - Sep 26, 2019 @04:04 PM CDT

Title: Team Meeting

Date: 9/26/19

Content by: Salina Loer

Present: All team members

Goals: Begin and work on design matrix

Content: After our advisor meeting today we met as a group to work on our design matrix using our preliminary design ideas. We decided on our criteria that we thought were the most important for the matrix- Transparency, ease of manufacturing, ease of loading, leakproof, adjustability, safety/contamination, cost. We weighted them from highest to lowest in that order and described what each criteria meant to us.

Conclusions/action items:



Team Meeting 9/30

• SALINA LOER (loer@wisc.edu) • Oct 09, 2019 @01:10 PM CDT

Title: Team Meeting

Date: 9/30/19

Content by: Salina Loer

Present: Salina L, Xavier F, Yanbo F, Tshawn Z

Goals: Set up preliminary presentation

Content: We met to set up the preliminary presentation slides and talk about who would work on what. We put our design matrix in the slides and then added everyone's preliminary designs as well as a few bullet points explaining the design. We moved information from the PDS to the slides and worked on editing stuff to be more concise and less text on the slides. We also started the progress report for the week.

Conclusions/action items: Continue on the preliminary presentation to be done on Wednesday so we can send it to Prof. Kinney to look over.
Finish progress report



Team Meeting 10/3

• SALINA LOER (loer@wisc.edu) • Oct 09, 2019 @01:20 PM CDT

Title: Team Meeting

Date: 10/3/19

Content by: Salina L

Present: All team members

Goals: Finish revising presentation and practice it

Content: We revised the presentation based on Prof. Kinney's comments and ideas. We edited slides to have less text and added more pictures such as our clients first prototype and our team picture. I finished my solidworks file for my design idea and added that to the slides.

We decided who would do what parts:

- Xavier- design matrix and conclusion
- Yanbo- Preliminary designs
- Courtney- Background material
- Tshawn- intro and problem statement
- Salina- Competing designs and PDS info

We practiced going through the presentation and recording ourselves. We also finished the progress report.

Conclusions/action items: Present our preliminary presentation tomorrow



Advisor meeting 9/13

- SALINA LOER (loer@wisc.edu) - Sep 23, 2019 @05:51 PM CDT

Title: Advisor meeting

Date: 9/11/19

Content by: Salina Loer

Present: All team members

Goals: Discuss first progress report and how our project is going with our advisor, Prof. Kinney

Content: We met with our advisory and she went over our first progress report. There was not a ton of information on it yet because it was the first week. She talked about how we should be constantly updating our problem statement as the project continued throughout the semester. We talked about our plan for next week and when we would be meeting with our client.

Conclusions/action items: Make sure to update problem statement in our next progress report



Advisor Meeting 9/20

- SALINA LOER (loer@wisc.edu) - Sep 23, 2019 @05:56 PM CDT

Title: Advisor meeting

Date: 9/20/19

Content by: Salina Loer

Present: All group members

Goals: Discuss our second progress report and PDS as well as our upcoming meeting with with Dr. Handelsman

Content: We went over our 2nd progress report and what we had worked on in the last week. We talked about our first design ideas and when we were going to meet as a group again to show our specific ideas and start the design matrix. We went over our PDS and she told us to update lots of specifics such as, measurements and actual tests we want it to pass to confirm what we want it to do. We will meet with our client next Wednesday the 25th to show our preliminary ideas and talk about more of what she wants from this device.

Conclusions/action items: Work on preliminary designs and start 3rd progress report and design matrix and update PDS



Advisor Meeting 9/26

- SALINA LOER (loer@wisc.edu) - Sep 26, 2019 @03:27 PM CDT

Title: Advisor meeting

Date: 9/26

Content by: Salina Loer

Present: Salina L, Courtney M, Tshawn Z, Xavier F

Goals: Discuss current project with Prof. Kinney

Content: We discussed our client meeting with Prof. Kinney and explained the new things that Dr. Hurley told us. Talked about the pros and cons of using milling vs injection molding to create it. Having holes in device so client can test many points. Discussed the issue of making sure the device is sealable if client wants to put it vertical. should decide on top three designs for preliminary design matrix

Conclusions/action items: Finish design matrix and begin presentation slides



9/9/19 Preliminary research

- Courtney Mohs - Oct 08, 2019 @02:47 PM CDT

Title: Preliminary Research

Date: 9/9/19

Content by: Courtney Mohs

Present:

Goals: To Understand the project and come up with some questions for the client meeting.

Content:

- A microcosm is a place or community that emulates the characteristics of something bigger. https://www.google.com/search?q=what+is+a+microcosm&rlz=1C1GCEA_enUS807US807&oq=what+is+a+microcosm&aqs=chrome..69i57j0l5.3723j0j7&sourceid=chrome&ie=UTF-8

Lexico Dictionaries | English. (2019). *Microcosm* | Definition of Microcosm by Lexico. [online] Available at: <https://www.lexico.com/en/definition/microcosm> [Accessed 8 Oct. 2019].

- An Alfalfa plant is a small clover-like perennial, growing 1-3 feet tall. The roots of the seedling grow rapidly, making it a good choice for the experiment because the roots will establish very quickly. <https://www.britannica.com/plant/alfalfa>

Encyclopedia Britannica. (2019). *Alfalfa* | plant. [online] Available at: <https://www.britannica.com/plant/alfalfa> [Accessed 8 Oct. 2019].

- A rhizosphere is a region close to the roots that impact the growth of the roots and the plants.
- The Apparatus

The apparatus needs to be permeable to oxygen and needs to be able to have samples taken out and have a microscope.

Some obstacles of the current design are that the sand gets packed too tightly which prohibits new bacteria and such from being mixed in.

Practice problem statement: To study the way microbes interact with plant roots and how they affect the growth of the plant an apparatus must be made that can efficiently grow the plant and study it without harming the plant. The goal is to make is an apparatus that will allow plant roots to grow and be able to sample the water surrounding them. The apparatus should also be able to introduce liquids and different bacterial cultures.

Conclusions/action items:



• Courtney Mohs • Oct 07, 2019 @10:37 PM CDT

Title: Continued Research on PDMS and materials

Date: 9/16/19

Content by: Courtney Mohs

Present: Courtney Mohs

Goals: To research how PDMS is manufactured and any possible materials that we can make.

Content:

There are multiples kinds of PDMS, not sure what kind they have been using previously. The cost seems to be around 100-170/gallon. Using PDMS you have to create a mold, pour it in and then cure it and it will set and then you can use glue to stick it to anything, or with enough time it will form a bond on its own. Also, it tends to age or slightly change color during the sterilizing process. However, PDMS seems to be the preferred choice by the clients.

Conclusions/action items:

We will most likely use milling to create the prototypes, however, if there will be a need to make multiple of these it may be a good idea to use injection molding.



Title: Transparency measurement

Date: 10/3/19

Content by: Courtney Mohs

Present: Individual research

Goals: to quantify some of our design requirements that we can test against

Content:

The transparency can be measured by transmittance, which measures the amount of light that fully passes through it. It takes into account refractions of light and things like that. For example, glass has about 94% transmittance and because it is very important for our project to have high transparency we have decided that the bottom of our device has to have at least 90% transmittance. We will use a transparency meter to determine how transparent the bottom of the device is.

[4]"Transparency Meter - Haze Gard-i | Qualitest", *Worldoftest.com*, 2019. [Online]. Available: <https://www.worldoftest.com/transparency-meter-haze-gard-i>. [Accessed: 25- Sep- 2019].

Conclusions/action items:



Procedure of the Device

• Courtney Mohs • Oct 08, 2019 @02:55 PM CDT

Title: Procedural steps Provided by Dr. Hurley

Date: 9/12/19

Content by: Dr. Hurley

Present: Courtney Mohs

Goals:

Content:

Attached

Conclusions/action items:

• Courtney Mohs • Oct 08, 2019 @02:56 PM CDT

MICROCOSM FOR BACTERIA AND PLANT ROOTS

Handwritten Lab and BME Collaboration

Growth chamber dimensions:

Square Base: 5 cm width x 7.6 cm length x 1 cm height
 Square PDMS layer: 4.8 cm width x 6.8 cm length x 4 mm height
 Oval Inlet chamber: 5.4 cm width x 3.2 cm length x 3 mm height

Growth chamber sand experiment:

- Load chamber with 5 g sand via inlet hole. Wrap in tin foil and autoclave on a dry cycle.
- Grow bacteria overnight. Determine bacterial concentration and dilute bacteria in fresh media at 10^6 bacteria/mL.
- Inoculate ~2 mL of bacteria via a 1 mL pipette into the inlet.
 - 1 day - 2 mL because a lot spilled out while I was inoculating.
 - I had to massage the sand to hydrate the bottom of the chamber.
 - Less sand may help with inoculation but I would worry the sand would not be static and would float in the liquid and disrupt any structures built by bacteria.
- Possible options:
 - Include an outlet to suck out sand and draw liquid down.
 - Double chamber with non-permeable membrane for equal inoculation along length.
 - Include a "stopper" at the top to compress sand after inoculation.
 - Be sure you guys have in case a better idea!
- Cover inlet with breathable seal and place growth chamber in a plastic container with a loose lid. Include soaked paper towels in empty tip boxes to maintain humidity in the container.
- Grow bacteria at 20°C for 2-3 days.
- Every day, image bacteria at the base of the growth chamber on a Nikon microscope.
- On the last day, use a razor to cut a horizontal line ~5-7mm below the inlet. Use the razor to separate the PDMS from the base. When open, use a sterile scoop to transfer sand into two eppendorf tubes.
- Repeat step 7 down the chamber (twice) until there are 6 eppendorf of sand - two replicates each for top, middle, and bottom of the chamber (with respect to the inlet).
- Weigh sand and add 1 mL liquid per 0.5 g to normalize volume harvested. Vortex and sonicate sand to remove attached bacteria. Perform serial bank dilutions and plate on appropriate antibiotics to quantify each species of bacteria.

[Microcosm_for_bacteria_and_plant_roots_2_.docx\(15.6 KB\) - download](#)



Title: Competing Designs

Date: 9/26/19 Competing Designs

Content by: Courtney Mohs/ Team 67

Present: Courtney Mohs

Goals: To Find any competing designs of the microcosm that we are designing.

Content:

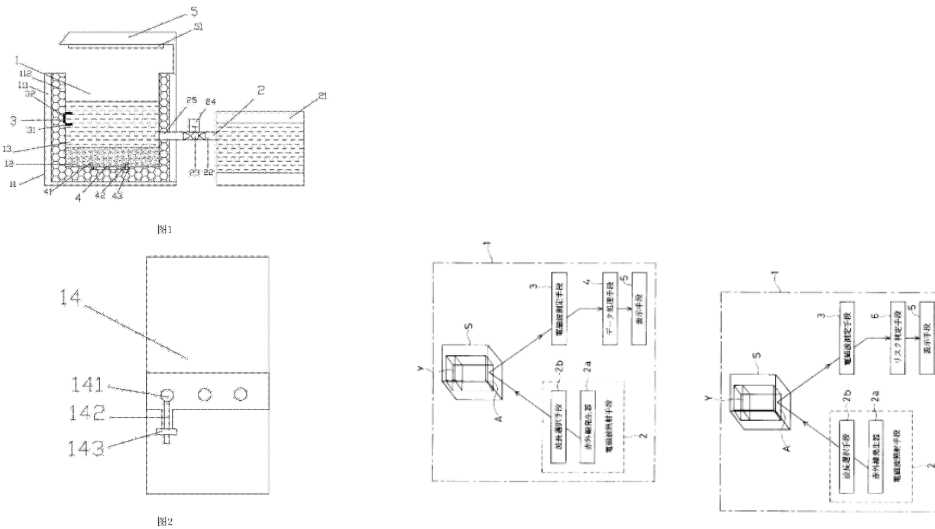
Indoor microcosmic ecological simulation experimental device and ecological simulation experimental method (CN107167564A)

A microcosm for raising plants under biotic and abiotic conditioning (EP3236741A1)

Microcosm inspection equipment (JP3891518B2)

A microfluidic co-cultivation platform to investigate microbial interactions at defined microenvironments[2]

These designs are similar to what we are doing by having a chamber for media culture, however they are way too big for what we are going to use our device for.



Conclusions/action items:

There are not many competing designs for the device we are making. This means that we may need to make many prototypes before we get a viable device. Also, we have the potential for a patent.



9/23/19- First brainstorm design

• Courtney Mohs • Sep 23, 2019 @04:52 PM CDT

Title: First Prototype designs

Date: 9/23/19

Content by: Courtney Mohs

Present: courtney, Salina, Xavier, Tshawn

Goals: To present our current ideas for the prototype and mesh them together.

Content:

Image Included below

Conclusions/action items:

• Courtney Mohs • Sep 23, 2019 @04:52 PM CDT

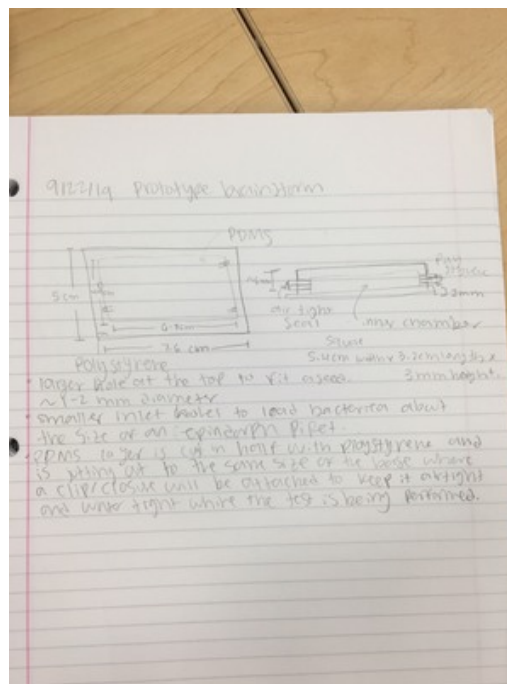


Image-1.png(1.5 MB) - [download](#)



9/9/19 w/ team

• Courtney Mohs • Sep 09, 2019 @05:22 PM CDT

Title: First Team Meeting

Date: 9/9/19

Content by: Courtney

Present: Team 67

Goals: Make the first progress report

Content:

Meeting with professor confirmed for September 25.

Made an excel sheet for expenses.

TShawn made a google doc to make the progress reports in.

Conclusions/action items:

Do some Preliminary research to get familiar with what the project is about.



9/11/19 Client meeting with Dr. Hurley

• Courtney Mohs • Sep 11, 2019 @05:29 PM CDT

Title: First client meeting to observe prototype

Date: 9.11.19

Content by: Courtney Mohs

Present: Xavier Fan, TShawn Zhu, Yanbo Feng, Salina Loer, Dr.Hurley

Goals: To observe the prototype and ask questions about what she wants fixed and to gather more information.

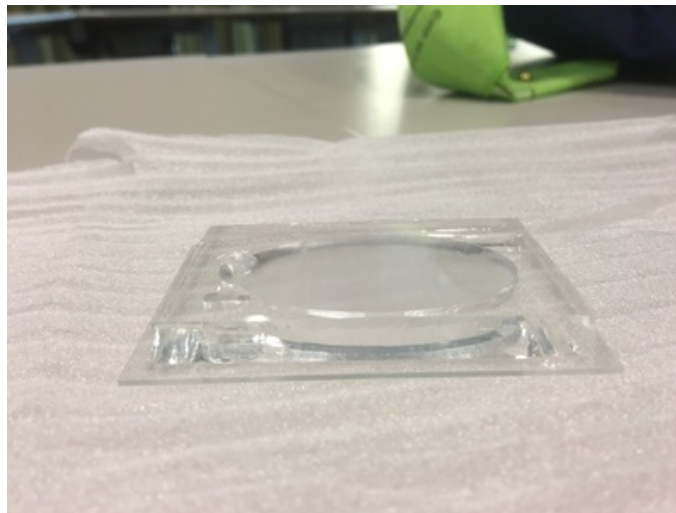
Content:

The prototype is the size and material she wants. She has had trouble loading it due to the fact there is only one hole into the chamber, so there is nowhere for the air inside the chamber to go. She also has had problems getting the bacteria out and has had to cut through the device in order to get the bacteria out. We were able to see the microscope they use, to see the bacteria and the way they have been doing the experiment before, and also to see the current device that they are trying to use. We also asked for Dr. Hurley to send us any CAD drawings of it, and also a procedure for the experiment. She would like it to be able to be taken apart so she can access the bacteria easily and scoop it out, Also a minimum of two holes on opposite ends to allow for drainage.

Conclusions/action items:

To brainstorm possible ways to fix this device.

• Courtney Mohs • Sep 11, 2019 @05:26 PM CDT



IMG_7438.JPG(1.5 MB) - [download](#)



IMG_7437.JPG(2.1 MB) - [download](#)

**Title: Advisor Meeting for Week 3****Date:** 9/20/2019**Content by:** Courtney Mohs**Present:** Professor Kinnley, Salina, TShawn, Xavier, Yenbo, Courtney**Goals:** Discuss what has been accomplished and to come up with a plan for the next week.**Content:** PDS is attached.

- References to the previous patent.
- Refined PDS, can fit onto a microscope platform- what are the dimensions of a microscope platform, what dimensions are we most constrained in.
- Polystyrene may be better used as the bottom microscope slide.
- client requirements can be used as grading for the design matrix. Need some way to quantify "transparency" and things like that.
- Talk about performance requirements that you can quantify- how much is loaded, how many bubbles, what does the loading process look like, how do you assess "works". Can be pass/fail.
- Can use food coloring to mimic the loading of bacteria.
- Be more specific in the weekly report so they can give some feedback and help guide us where we want to go and refine the objective. It goes to the client so if there is a list of questions then we can put those in them.

Conclusions/action items:

Design Matrix

Microcosm for Bacteria and Plant Roots

Date: 18 September 2019
Team Members: Yunbo Feng, Xavier Fan, TShawn Zhu, Salina Loer, Courtney Mohs
Advisor: Prof. Melissa Kinney
Client: Prof. Jo Handelsman

Function:

The microcosm apparatus should consist of large seed pore for settlement of plant seed, a top sealed chamber for root growth, an outlet pore for extracting material and assisting airflow, one or two inlet pores for media and bacteria inoculation or settlement of two different bacteria. The material for the apparatus should be crystal clear for microscopy, oxygen permeable for bacteria growth and root growth, biologically inert for plants and bacteria and immune to possible deformation caused autoclave. The apparatus should be able to be disassembled easily for studying the interaction between bacteria and plant roots under microscope.

Problem Statement:

Researchers developed an apparatus which can allow observation and study of the interaction between bacteria and plant root and possible structures formed by bacteria under the condition of culturing media and sand. However, the current prototype lacks the ability to test the effects of competing types of bacteria on the plant root, the ability to be sealed and is not friendly to users. In order to improve the functionality of the apparatus and the efficiency of the research procedure, a microcosm apparatus with inlet and outlet pores, separated parts for different bacteria and detachable structure needs to be developed by either modifying the prototype or designing a new one.

Client requirements:

- Apparatus can fit into the microscope platform
- Upper layer material permeable to oxygen
- Material transparent to observe the bacteria under the microscope
- Inner chamber for root growth and interaction between roots and bacteria
- Include an inlet for culturing liquid, bacteria, and sand/plant seed
- Include an outlet for draining out the liquid and preventing spills while loading
- Include pores for different bacteria
- Detachable structure to simplify the extraction process

[Microcosm_for_Bacteria_and_Plant_Roots_PDS_Week_3_.docx\(18.2 KB\) - download](#)



9/25/19- Second Client Meeting

- Yanbo (David) Feng - Oct 09, 2019 @10:03 AM CDT

Title: Second Client meeting

Date: 9/25/19

Content by: Courtney Mohs

Present: Yanbo, TShawn, Xavier, Salina, Courtney, Dr. Hurley

Goals: Go over our preliminary designs with Dr. Hurley and get feedback, and more information

Content:

- Alphanase seeds
- Glass bottom to work with microscopy
- The microscope objective is on the bottom of the microscope plate, so the base needs to be transparent.
- chamber with customizable chamber size, ie. it can get taller and shorter.
- For the holes at the bottom of the design, if the plate gets tipped upright 90 degrees the holes at the bottom need to be plugged in the bottom.
- The height should be able to be manipulated to allow for more or less room for the roots to grow.
- Possibly holes on the along the sides to be able to extract/ put in different liquids during the experiment.
- Inside chamber 2-3mm in height is okay. thinner height to start and then be able to add more height if needed.
- Having it be reusable is ideal, however, not expected to happen due to autoclaving.

Conclusions/action items:

To combine designs and make a design matrix. And make designs in Solidworks.



9/26/19 Meeting w Advisor

• Courtney Mohs • Sep 26, 2019 @04:03 PM CDT

Title: 9/26/19 Meeting w Advisor

Date: 9/26/19

Content by: Courtney Mohs

Present:

Goals: To go over our designs and to make a design matrix

Content:

- Looking at the cost of making molds or injection molding for PDMS.
- research how to know what way roots will grow.

Conclusions/action items:

To make the design matrix



10/3/19 Meeting with team to finish presentation slides

• Courtney Mohs • Oct 08, 2019 @03:09 PM CDT

Title: Meeting to work on and rehearse the preliminary presentation

Date: 10/3/19

Content by: Courtney Mohs

Present: Team 67

Goals: To update the slides, and to practice what we want to cover in the presentation

Content:

I will be doing the slides on the background information. ie. what we are using the device for and how it is supposed to work. Going through the procedural steps to help the audience understand why we need to make this device. Go through the current prototype likes and dislikes.

Slide 1: Mention the pore for the plant. This is the prototype that they have, they like that there is a glass slide on the bottom and that the chamber is made out of PDMS. However, it is not actually functional, it cant be loaded without the media being spilled out because there is only one hole, and it can't be opened up easily, so they have not been able to run the experiment. Currently they are studying the bacteria by putting them in a test tube, mixing them up and then taking them out of the test tube to run analysis on them, in result this disturbs any form or structure the bacteria had made. Because the prototype is not functional they haven't been able to actually put the bacteria around plant roots and

Slide 2:

The process that we are designing for is to have the device open to load the mass culture media and sand, close the device and insert the plant seed into the hole, load the bacteria into the holes. It is important these are standardized so the seed and bacteria are always loaded in the same place. Then they will allow the plant to grow and bacteria to interact with the roots. Then use microscopy to observe the plant roots and bacteria, then eventually open the lid and extract the culture media and sand and potentially run further tests on the bacteria. Next, Salina will introduce some competing designs

Conclusions/action items:

Give the presentation. Also, grade other groups while they are giving their presentations to evaluate what they did well and could do better. Also, to evaluate what we could do better.



2014/11/03-Entry guidelines

• John Puccinelli • Sep 05, 2016 @01:18 PM CDT

Use this as a guide for every entry

- Every text entry of your notebook should have the **bold titles** below.
- Every page/entry should be **named starting with the date** of the entry's first creation/activity, subsequent material from future dates can be added later.

You can create a copy of the blank template by first opening the desired folder, clicking on "New", selecting "Copy Existing Page...", and then select "2014/11/03-Template")

Title: Descriptive title (i.e. Client Meeting)

Date: 9/5/2016

Content by: The one person who wrote the content

Present: Names of those present if more than just you (not necessary for individual work)

Goals: Establish clear goals for all text entries (meetings, individual work, etc.).

Content:

Contains clear and organized notes (also includes any references used)

Conclusions/action items:

Recap only the most significant findings and/or action items resulting from the entry.



Title:

Date:

Content by:

Present:

Goals:

Content:

Conclusions/action items: