

BEAN BEETLE MICROBIOME: INTRODUCTION TO LABORATORY RESEARCH

INSTRUCTOR: Lawrence Blumer
Office: 302 Hope Hall
lawrence.blumer@morehouse.edu
470 639-0283

Laboratory: Tuesday 13:00-16:50, 212 NMM (CRN 43362)

COURSE TEXTS: Bean Beetle Microbiome student guide. (pdf on course BlackBoard website)

A Student Handbook for Writing in Biology, 3rd edition (2009) or 4th edition (2013) by Karin Knisely; Sinauer and Freeman Publishers (available from Amazon)

Research papers will be posted to BlackBoard

Bound composition notebook and a 3-ring binder for laboratory handouts

CREDIT HOURS: This laboratory course will count toward 40% of your grade in BIO 111.

DESCRIPTION AND ORGANIZATION

In this course, you will be conducting original research on the microbial communities found in the digestive tract of bean beetles, *Callosobruchus maculatus*. The course-based research experience (CURE) that we will share is a different method of learning and teaching in a laboratory course than you may have experienced in the past. CUREs have been found to yield significant improvements in the learning and performance of students compared to control groups. We look forward to starting each of you on a very successful academic program.

COURSE OBJECTIVES

1. The ability to perform the basic techniques of Molecular Biology, such as PCR.
2. Keep detailed and accurate laboratory notes
3. Prepare and present concise research seminars using PowerPoint slides
4. The ability to determine appropriate controls to validate the experiment.
5. Demonstrate the skills needed to mine bioinformatics data to produce meaningful results.
6. Produce clear and concise written assignments and a Laboratory Research Paper.
7. The ability to interpret data and scientific charts, and to create tables and graphs that meaningfully conveys information.
8. Describe and interpret the importance of microbial communities in and on animals including humans.

Laboratory Pre-test

Please take the laboratory pre-test on-line at:

This link also is located on the BlackBoard site for the course.

Week	Activities	Readings PRIOR to class
1	Laboratory safety and Pre-Course Assessments	Introduction, Lab Basics (BlackBoard) Preventing Contamination and Aseptic Technique, and Notebook Protocols
27 Aug	Introduction to insect microbiomes, bean beetles, experimental design, & culturing of microbes How to use a micro pipettor Aseptic Technique Proper notebook protocol	Microbiome Culturing Protocol (BlackBoard)
2	Discuss Sevim et al., 2016 paper	Read Sevim et al., 2016 paper and complete worksheet (BlackBoard)
3 Sep	Design and Start Experiment Review microbiome biology Review Scientific Method	Visit Woodruff Library, find 3 Microbiome research studies, and complete worksheet (BlackBoard) Scientific Method Readings (BlackBoard)
	Micropipettor quiz	
3	Review microbiome biology	Take practice Microbiome Biology quiz
10 Sep	Microbiome quiz Practice microbiome isolation	Microbiome Culturing Protocol (BlackBoard)
4	Write Introduction draft	Student Handbook for writing
17 Sep	Bacterial phenotypic assessment practice	Colony Phenotype Traits (BlackBoard)
5 - 6	DNA extraction for microbial community sequencing (repeat as needed)	DNA Extraction Protocol (BlackBoard)
24 Sep 1 Oct	Microbiome culturing	Microbiome Culturing Protocol

7	Colony-based PCR and colony archiving with stab cultures, repeat PCR as needed Electrophoresis of PCR products and submission for colony sequencing	Colony PCR protocol (BlackBoard)
8 Oct	Bacterial phenotypic assessment and data entry Photograph colonies on plates	Colony Phenotype Traits (BlackBoard) Taking Microbiome Culture Photographs
8 15 Oct	No Classes – Fall Break	
9 22 Oct	Bacterial phenotypic assessment and data entry Photograph colonies on plates	Colony PCR protocol (BlackBoard) Taking Microbiome Culture Photographs
10 29 Oct	Introduction to bioinformatics Analysis of colony sequence data Introduction to community analysis, phenotype & colony sequencing analysis	nBLAST protocol (BlackBoard) Microbial Community Colony Phenotype and Colony-based Sequencing Database Analyses (BlackBoard)
11 5 Nov	Introduction to DNA Subway and analysis of microbial community sequencing data	
12 12 Nov	Bioinformatic analysis of microbial community sequencing data	
13 19 Nov	Community analysis of sequencing data	

14 26 Nov	Community analysis of sequencing data
Prep Time for Presentations	

15 3 Dec	Final Presentation and Final Report due in class
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EVALUATION

Your grade will consist of the following parts:
 30% - Class Attendance
 20% - Laboratory Notebook Completeness
 10% - Oral presentations
 25% - Writing Assignments, Homework and Quizzes
 15% - Final Laboratory Report

GRADING SYSTEM OUTLINE

A = 90 - 100	C+ = 75 - 77
A- = 88 - 89	C = 70 - 74
B+ = 85 - 87	C- = 68 - 69
B = 80 - 84	D+ = 65 - 67
B- = 78 -79	D = 60 - 64

CLASS ATTENDANCE

Students are expected to attend each class meeting. Students with more than 3 unexcused absences will be referred to the Office of Student Success and may be administratively withdrawn from the course. Failure to meet minimum attendance requirements may result in the loss of the student’s financial aid in accordance with federal financial aid requirements.

If you are absent from class, it is your responsibility to make up scheduled work missed because of an officially excused class absence. Any unexcused absence will result in a reduction of 1 letter grade in the course. Official excuses from the Dean for absences must be presented to your instructor within five days of returning to class.

Inclement Weather Policy

In the event of inclement weather, the College will announce any closures via the emergency notification system and/or through local news outlets. Absent an official closure, students are not excused from attending class due to weather and any absences will be considered unexcused.

WRITING ASSIGNMENTS

There will be frequent short writing assignment in this course that will count for 25% of the course grade. These assignments will be made in class on Wednesday and will be due before class the following Monday each week. Unless otherwise instructed, all writing assignments must be submitted via the Turn-It-In website as a typed, double-spaced document using 12pt font. You must register on the Turnitin.com website prior to submitting assignments.

CLASSROOM ACCOMMODATIONS

Morehouse College is an equal opportunity employer and educational institution. Students with disabilities or those who suspect they have a disability must register with the Office of Disability Services (“ODS”) in order to receive accommodations. Students currently registered with the ODS are required to present their Disability Services Accommodation Letter to faculty immediately upon receiving the accommodation. If you have any questions, contact the Office of Disability Services, 104 Sale Hall Annex, Morehouse College, 830 Westview Dr. S.W., Atlanta, GA 30314 (404) 215-2636.

ACADEMIC HONESTY

Morehouse College students are expected to conduct themselves with the highest level of ethics and academic honesty at all times and abide by the terms set forth in the Student Handbook and Code of Conduct. Instances of academic dishonesty, including, but not limited to plagiarism and cheating on examinations and assignments, are taken seriously and may result in a failing grade for the assignment or course and may be reported to the Honor and Conduct Review Board for disciplinary action.

Although much of the work we do in this course will require that we pool data and construct a single class data set, each of you is expected to do your own work on all assignments, in-class quizzes, take-home quizzes, and all writing assignments. You will be expected to make your own figures and tables and write your own prose for these assignments.

Copying or paraphrasing someone else’s prose (from a fellow student or a published reference), using someone else’s figure or table (even if it is based on the same data as a figure or table you could make) or submitting someone else’s work as your own is plagiarism. Giving a literature citation is not sufficient. We require that you submit work that you have written yourself in your own words. Papers with long quotations (even if fully referenced) will not be accepted. Leaving your work on a laboratory computer hard-drive so other students may freely copy that work is not advised, as it will result in accusations of plagiarism against both the honest and dishonest students. At a minimum, plagiarism will result in a failing grade and a report to the Dean of Students.

A syllabus is not a contract between instructor and student, but rather a guide to course procedures. The instructor reserves the right to amend the syllabus when conflicts, emergencies or circumstances dictate. Students will be duly notified.

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Office: 302 Hope Hall (office hours are via BlackBoard)
lawrence.blumer@morehouse.edu
Office Hours MWF 10:00-10:50 EST and by appointment
Office Zoom Link

LABORATORY: **Tuesday 13:00-16:50 EST (CRN 45771)** meeting on Zoom:
Synchronous laboratory meeting Zoom Link
Class meeting will begin promptly at 13:00 EST.

COURSE TEXTS: **On-line tutorial Understanding Experimental Design** (see page 5 on SimBio)
Link to SimBio tutorial

Bean Beetle Microbiome student protocols. (pdfs on BlackBoard)
Readings and worksheets will be posted to BlackBoard

Thumb drive (flash drive) to save all handouts and written work.

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DESCRIPTION AND ORGANIZATION

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COURSE OBJECTIVES

1. **Read and interpret scientific publications and suggest new research questions.**
2. Describe the importance of microbial communities in and on animals including humans.
3. **Design a new experiment to test alternative hypotheses and predictions**
4. Determine appropriate controls for the experiment.
5. Keep detailed and accurate laboratory notes.
6. Evaluate bioinformatics data to address the question posed by your new experiment.
7. Write a Laboratory Research Paper.
8. Interpret data to create tables and graphs to describe experimental results.
9. Prepare and present research seminar using PowerPoint or Google slides.

Laboratory Pre-test

Please take the laboratory pre-test on-line at:

Week	Activities	Readings and work PRIOR to class
1 2 Feb	Laboratory Safety and Pre-Course Assessments Introduction to insect microbiomes, bean beetles, experimental design, & culturing of microbes Units of measure and using a micro-pipetter (Demo) Aseptic technique (Demo) Laboratory Notebook protocol	Introduction, Lab Basics Preventing Contamination and Aseptic Technique, and Notebook Protocols (BlackBoard) Start Understanding Experimental Design (SimBio)
2 9 Feb	Discuss research papers Design and Start Experiment Review microbiome biology Review scientific method Notebook check (Journal in BlackBoard) Microbiome quiz (submit) Pronouncing important names	Read two research papers and submit reading worksheets (BlackBoard) Post one experimental question to Discussion Board and respond to two other students. Scientific Method Readings (BlackBoard) Take practice Microbiome Biology quiz Complete Understanding Experimental Design (SimBio)
3 16 Feb	Surface sterile and isolate beetles from experiment (Demo) DNA extraction for microbial community sequencing (Demo). Extracted DNA to be sent for NextGen Sequencing. Review Research Report format Microbiome culturing (Demo)	Review Research Report rubric and outline (BlackBoard) Visit Woodruff Library at www.auctr.edu, find 3 microbiome research studies, and submit 3 worksheet (BlackBoard) DNA Extraction Protocol (BlackBoard) Microbiome Culturing Protocol (BlackBoard)

Week	Activities	Readings and work PRIOR to class
4 23 Feb	View media plates of bacteria from whole beetle microbiome culturing Bacterial Colony Phenotype results (submit data) Identify colonies to pick for PCR (one per student). Prepare Colony-based PCR and colony archiving with stab cultures (Demo - PCR is run after class) Notebook check	Colony PCR protocol (BlackBoard) Colony Phenotype Traits (BlackBoard) Report Draft 1, Introduction (submit via BlackBoard)
5 2 Mar	View electrophoresis of PCR products (identify samples to be submitted for Sanger Sequencing of individual colonies) Colony Phenotype analysis (submit results)	Colony PCR protocol (BlackBoard) Colony Phenotype Traits (BlackBoard) Review Research Report rubric and outline (BlackBoard)
6 9 Mar	Introduction to bioinformatics Analysis of picked colony Sanger sequences from Colony-based Sequencing data Introduction to community analysis, phenotype & colony sequencing analysis Notebook check Submit nBlast results for Colony-based Sequencing	nBLAST protocol and video tutorial (BlackBoard) Microbial Community Colony Phenotype and Colony-based Sequencing Database Analyses and video tutorial (BlackBoard) Report Draft 2, Introduction and Results (submit via BlackBoard)
7 16 Mar	Introduction to Community Ecology Evaluating Diversity and Comparing Communities Submit community analysis of picked colony data	Taxonomic classification worksheet (BlackBoard) Community Ecology protocol (BlackBoard)
8 23 Mar	Introduction to DNA Subway and analysis of microbial community sequencing data Submit level-5 table formatted for analysis Notebook check	CyVerse Tutorial (BlackBoard) DNA Subway protocol and video tutorial (BlackBoard) Preparing Files for Downstream Analyses (BlackBoard) Report Draft 3, Introduction Results and Discussion (submit via BlackBoard)
30 Mar	No Laboratory This Week – Mini-Break	

Week	Activities	Readings and work PRIOR to class
9 6 Apr	Bioinformatic analysis of microbial community sequencing data Submit a Taxonomy Bar Graph (using rarified data)	Community Analysis in R-Shiny app and video tutorial (BlackBoard)
10 13 Apr	Bioinformatic analysis of microbial community sequencing data Discuss Seminar Format Role of Figures and Tables in Results Submit Alpha Diversity results and Notebook check	Research Seminar rubric (BlackBoard) Preparing graphs and tables (BlackBoard)
11 20 Apr	Bioinformatic analysis of microbial community sequencing data Preparation for Research Seminars Pronouncing scientific names with confidence	Research Seminar rubric (BlackBoard) Research Seminar Draft (ten slides) (submit via BlackBoard)
12 27 Apr Moved to 29 Apr	Research Seminar Presentations Submit Final Seminar slides	Your seminar will be presented to the class this week via BlackBoard Collaborate Ultra or Zoom. Have your slides ready to present from your computer.

EVALUATION

Your laboratory score will consist of the following parts:

- 10% - Laboratory Notebook (50)
- 30% - Research Seminar slides and presentation (150)
- 30% - Writing Assignments, Homework and Quizzes (150)
- 30% - Mid-semester Research Report (150)

GRADING

There is no grade for the laboratory part of BIO 111. Your work in the laboratory will contribute to 40% of the grade you receive in the whole BIO 111 course, lecture and laboratory combined. The Pass/Fail Option that you may choose at the end of the semester in S2021 will apply to the entire course, since the lecture and laboratory has one combined grade.

Your laboratory assignments will be graded and all scores will be posted on BlackBoard.

CLASS MEETINGS and ATTENDANCE

Our regular lecture class meeting will be held via Zoom on W 13:00-16:50pm EST and will be recorded. Every class meeting recording will be posted on the course BlackBoard site and will be available for the entire semester. **However, if you are unable to regularly attend the scheduled**

meetings of the laboratory, past experience suggests that you should not take this course. Success in this course, whether on-line or in-person, requires regular interactions with the instructor and the materials. The following attendance policy is from the Office of the Provost, Morehouse College, Academic Affairs:

Students enrolled in online courses are expected to maintain ongoing course engagement in order to uphold positive academic standing with Morehouse and in order to comply with Federal Regulations. Maintaining an ongoing online presence requires timely participation in course related activities that may include, but is not limited to reading announcements, taking exams online, participating in group work, posting to discussion forums, submitting assignments, watching lectures and responding to prompts, and carrying out the requirements set forth by the instructor. It is a good practice to login to online courses several times a week to stay informed of news, announcements, grades, assignments, and other important course information.

WRITING ASSIGNMENTS

All writing assignments will be submitted via the BlackBoard website for our course.

SIMBIO SimUText ON-LINE TUTORIAL

It is important that you review the information below *before* you subscribe to the SimUText for **General Biology Laboratory** at **Morehouse College**. **To avoid possible problems, do not wait until the last minute. The cost of this subscription is \$6.00.**

1. CHECK YOUR TECH!

Visit <https://simutext.zendesk.com/hc/en-us/categories/200170134-Check-Your-Tech-> to confirm that the SimUText application will work on your computer, and/or to explore your options if there is a problem.

2. SimUText Voucher Code (optional)

If you purchased a SimUText Voucher from your bookstore, be sure to have it with you when subscribing, as you will need to enter your voucher code.

3. Registration Link

When you are ready to subscribe and download installers, follow this link to initiate the process: link to SimUText for this course

4. SimUText Application Installers

After you have completed the subscription process, if you need to download the SimUText application installers again, you will be able to access them by logging into the [SimUText Student Portal](https://simutext2.com/student/) (<https://simutext2.com/student/>).

Save this email! Should you encounter problems, you may need your course-specific Access Key. It is: **access key code**

Problems or questions? Visit [SimUText Support](http://simbio.com/support/simutext) (<http://simbio.com/support/simutext>)

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ViABLE mini Workshop
2021

Larry Blumer, Morehouse College

Teaching the Bean Beetle Microbiome
CURE in an Online Format

Bean Beetle Microbiome Project

www.beanbeetles.org

with Chris Beck, Nicole Gerardo, Anna
Zelaya, Sinead Young



Supported by awards
DUE-1821533 and DUE-1821184



MOREHOUSE
COLLEGE



EMORY
UNIVERSITY

Both a **Biological** Research and
Education Research Project

Insect-Microbiome Interactions

Course-Based Undergraduate
Research Experience (CURE)

1. Colony Phenotypes >>> ID different Taxa
2. Picked Colony, PCR, Sanger sequence >>>
Taxa ID (Genus)
3. NextGen Sequence >>> Taxa ID (Family)

Taxa ID permits community comparisons

Taxa ID permits community comparisons

Alpha Diversity

- Taxon Richness

- Simpson Index

- Shannon-Weaver Index

Beta Diversity

- Community composition similarity

Full Semester Implementation at Morehouse

Fall 2019: In-person laboratories
first-semester Biology Majors (BIO 111L)

Spring 2020: Started in-person
shifted to online at midsemester

Fall 2020 and Spring 2021:
fully online laboratories (BIO 111L)

Online Implementations

No kits sent to students

No biological materials handled by students

Simulation tutorial at the start of the course

Introduce experimental design concepts

What is lost in online format?

What is maintained?

Is something gained?

What is lost?

Ability to handle tools and materials

Measuring and following protocols

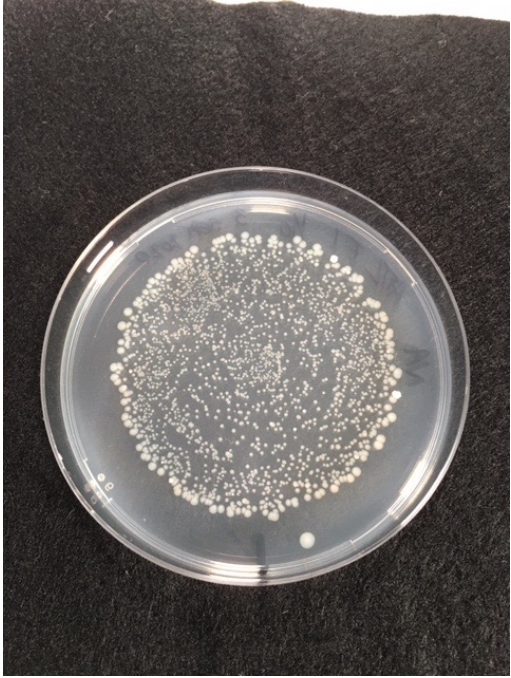
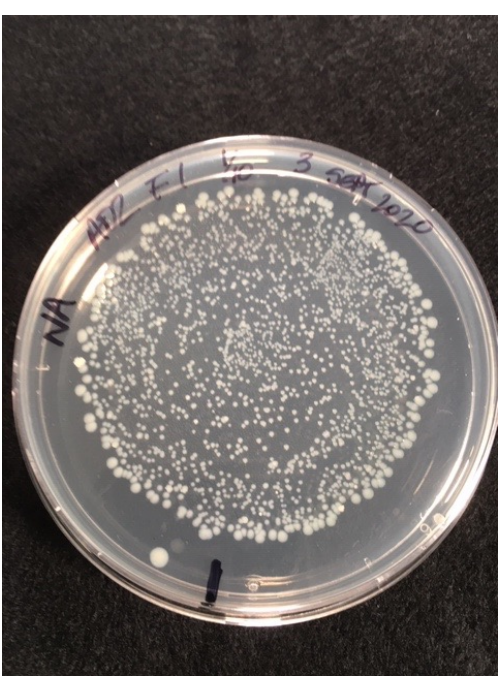
Conducting experiment

What is maintained?

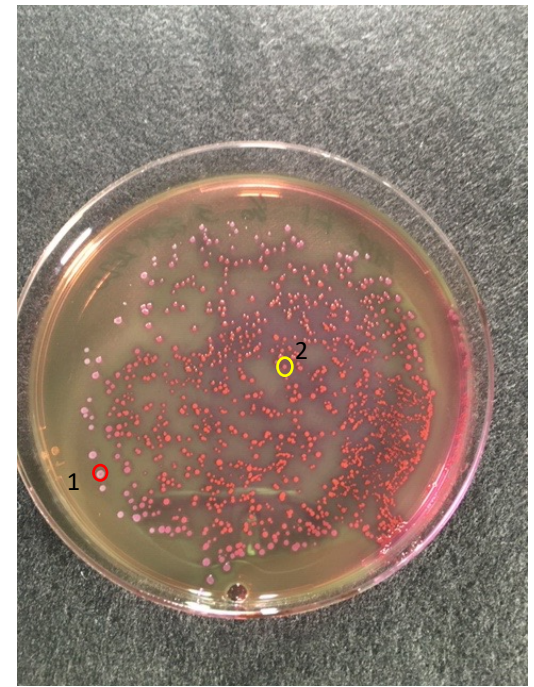
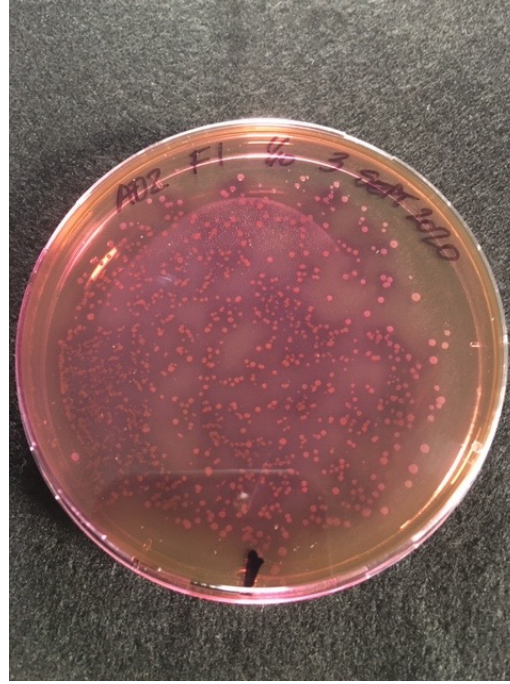
Scoring microbiome cultures

Observing PCR products of picked colonies

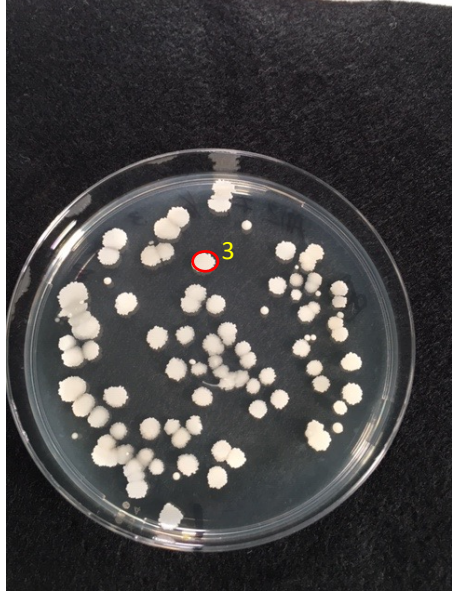
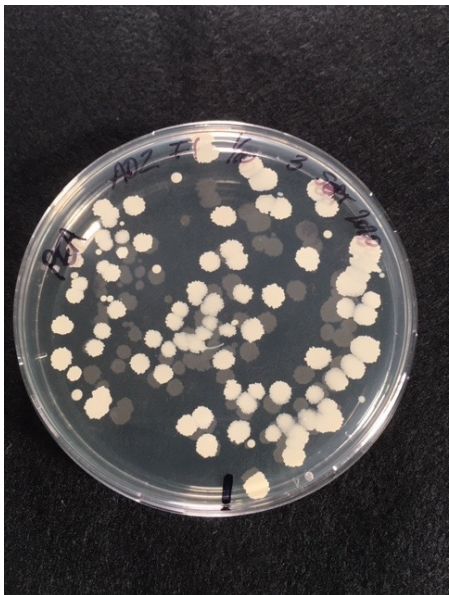
Conducting nBLAST searches



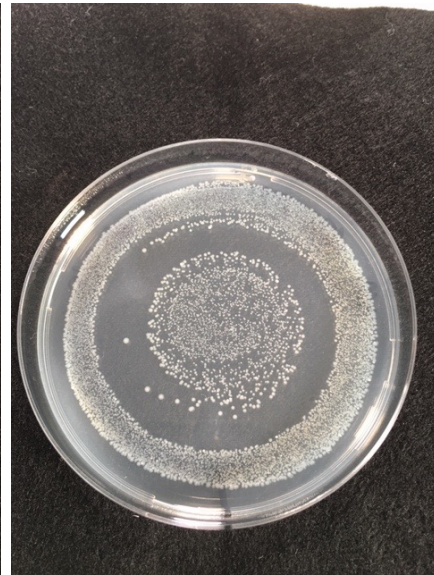
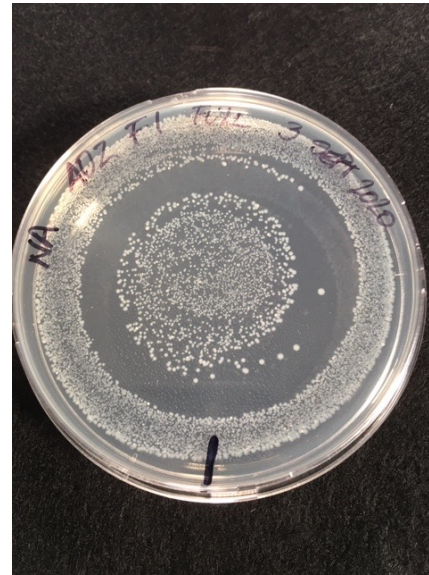
ADZ Female 1 Nutrient Agar 0.1 concentration



ADZ Female 1 EMB Agar 0.1 concentration



ADZ Female 1 PEA Agar 0.1 concentration



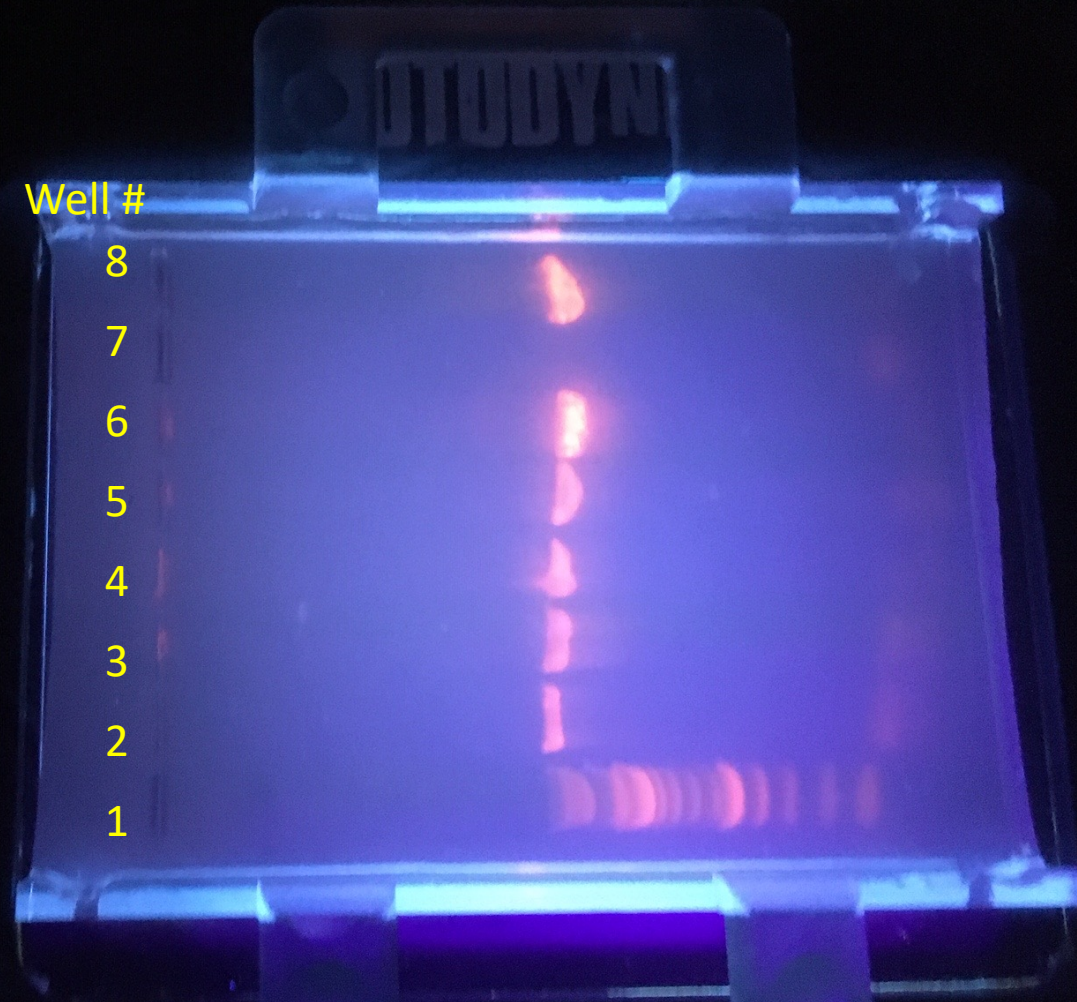
ADZ Female 1 Nutrient Agar full concentration

Well #

- 1: 100bp ladder
- 2: picked colony 1
- 3: picked colony 2
- 4: picked colony 3
- 5: picked colony 4
- 6: picked colony 5
- 7: Negative Control
- 8: Positive Control

Well #

8
7
6
5
4
3
2
1



Sanger Sequencing

>T1-ADZ-1429R_5uL__A09.ab1

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NNNNNNNNTNNGTAGCGCCCTCCCGAAGGTTAAGCTACCTACTTCTTTTTGCAACCCACTCCCATG
GTGTGACGGGCGGTGTGTACAAGGCCCGGGAACGTATTCACCGTGGCATTCTGATCCACGATTAC
TAGCGATTCCGACTTCATGGAGTCGAGTTGCAGACTCCAATCCGGACTACGACGCACTTTATGAGG
TCCGCTTGCTCTCGCGAGGTCGCTTCTCTTTGTATGCGCCATTGTAGCACGTGTGTAGCCCTACTCT
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GTTTACGGCGTGGACTACCAGGGTATCTAATCCTGTTTGCTCCCCACGCTTTCGCACCTGAGCGTC
AGTCTTTGTCCAGGGGGCCGCCTTCGCCACCGGGTATTCCTCCAGATCTCTACGCATTTACCCGC
TACAC
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Similarities: In-person and Online Laboratories

- Read and discuss research literature
- Introduction to animal-microbiome interactions
- Introduction to bean beetle lifecycle
- Design experiment
 - Set question (student autonomy)
 - Develop hypotheses
 - Make predictions
 - Discuss controls

Similarities: In-person and Online Laboratories

- Maintain laboratory notebook
- Prepare and revise written research report
 - Text revisions
 - Results Figures and Tables
 - Use of references for context
- View-tabulate colony phenotype results, prepare taxonomy table
- Select bacterial colonies for PCR amplification

Similarities:

In-person and Online Laboratories

- View and interpret electrophoresis gel results
- Perform BLASTn to ID picked colonies and prepare taxonomy table
- Perform community ecology analyses on colony phenotype and picked colony data

Similarities:

In-person and Online Laboratories

- Process NextGen Sequencing output to prepare taxonomy table
- Perform community ecology analyses on NextGen sequence data
- Prepare and present seminar on research results

Is online less expensive than
in-person laboratories?

Time

Materials

Replication

Is online sufficient authentic to achieve
the promise of CURE learning-teaching?

Student self-efficacy

Student skill gains

Experimental design skills

Practical laboratory skills

Student knowledge gains

Opportunities for iteration and collaboration diminished in online laboratories?

Should online laboratories be used as a scaffolding for in-person laboratories?

What do we lose in online?

Are online CUREs more efficient means of learning-teaching?