

Priya Narasingarao

HSS 1145D

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Office hours – Wednesday 10 AM in HSS1145D

Lecture: TWThF 11AM to 12:20pm in Warren Lecture Hall (WLH) 2111

Labs: York 2310 and 2332

TWThF 12:30pm to 4:30pm

Teaching Assistants

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Course Structure:

This course will introduce you to the fundamentals of microbiology and allow you to explore the many ways in which microbes affect and are used in our lives. We begin the course with a foundation in basic techniques such as sterile techniques, microscopy, methods of quantitating microbes, and preparing and examining stained slides. The remaining duration of the course will comprise four main units: a comprehensive look at bacterial physiology, understanding the complex microbial community of soil, metagenomics as a tool in exploring complex communities, and the use of microbes in various aspects of our lives. Each of these units comprises several multi-day experiments and there will be considerable overlap in the execution, methodology, and analysis of data from each of these units. Throughout the course, you will also receive training in accurate data entry and analysis, scientific reasoning, and in clear and concise scientific writing.

Equipment:

For this lab you will need to purchase:

- A lab notebook WITH carbon copies.
- A lab coat; and,
- Eye protection (you may wear either safety glasses or goggles, but standard prescription eye glasses are not sufficient).

Attendance and Absences:

1. Your attendance is required at EVERY lab and through the entire lab period, until all the experimental work for the day is completed.
2. Absences will NOT be treated lightly. The labs are set up for groups of two or more and your absence will place an unnecessary burden on your partner. There are no make up labs and you will not be allowed in the lab on non-lab days or in the other Micro lab sections, although you may be asked to make up the work from the day you missed.
3. Documentation will be required for all unavoidable absences.
4. If you are likely to have interviews for graduate school, etc., please schedule them on non-lab days.
5. All absences without prior notification/permission and the appropriate paperwork will be considered unauthorized.
6. 50-point penalty for the first unauthorized, unexplained absence from the lab. If there is a second such absence, you will be asked to drop the course.
7. If you are ill on a lab day or have an emergency, e-mail or call (instructor or lab partner) before the start of the lab. If you are ill enough to miss lab you must go to the student health center and provide documentation of your illness.

Assignment Deadlines and Submission:

1. A hard copy of each lab report is due in the first 10 minutes of the lecture on which your report is due. Reports turned in more than 10 minutes after the start of class will be considered late. Penalty for late reports will be 10% for each day late.
2. In addition to the hard copy of the report, you are required to submit an electronic copy to Turnitin.com. A link to the e-submission website will be provided on WebCT. Failure to submit on Turnitin.com will result in 0 (zero points) recorded for that report. Check the deadline of the Turnitin.com submission and make sure you adhere to it. Students agree that by taking this course all required papers would be subject to review for textual similarity by Turnitin.com for the detection of plagiarism. All submitted papers will be included as source documents in the Turnitin.com reference database solely for the purpose of detecting plagiarism of such papers. Use of the Turnitin.com service is subject to the terms of use agreement posted on the Turnitin.com site.
3. Additional points may be taken for late electronic submissions.

Regrade Requests:

All regrade requests should be submitted in writing within one week of receiving the graded material.

Grading scheme:

Note Book (10*3)	30
Pop quizzes (5*4)	25
Lab evaluation (continuous assessment)	25
Home Work (20, 10)	30
Lab Report - 1	100
Lab report - 2	100
Midterms I	75
Midterm II	75
Final Total	460

Grade Distribution

97+ = A+
93-96 = A
90-92 = A-
87-89 = B+
83-86 = B
80-82 = B-
76-79 = C+
70-75 = C
68-70 = C-
60-68 = D
Less than 60 = F

Notebook:

You must have a lab notebook with carbon copies. All notebooks should have a table of contents (handwritten OK) so on the first lab day leave several blank pages at the beginning of your notebook. Number your pages. Entries should be made in chronological order and EVERY day. Each day's entries on each experiment should begin with a brief (1 – 2 sentences) summary of work done on the same experiment the previous day.

How to use your notebook

Table of contents

Start a new page each day. For each experiment:

- Purpose of experiment
- Procedure
 - Outline or page from which protocol was taken
 - Note any changes
 - Note who did which part of the procedure – who inoculated controls, etc
 - Note which organisms you used – name and species of the controls, etc
 - Errors
- Observations
 - Write
 - Draw

- Questions and connections
- Conclusion or summary
- Answer any questions in the manual or that were raised in class.

Lab Performance and Participation

In addition to quizzes, midterms, lab reports and assignments, student evaluations will be based on the following criteria:

1. Lab techniques will be evaluated in class
2. Subjective student evaluations will be based on the following criteria:
 - Pre-lab preparation
 - Careful management of lab procedures (e.g., sterile technique, proper waste disposal, experimental procedures, etc.)
 - Ability to adapt to unforeseen procedural changes
 - Caliber of thinking before asking questions
 - Scientific approach (e.g., proper use of notebooks, controls, experimental design)
 - Accuracy
 - Independence
 - Safety consciousness
 - General neatness in lab

Please note: **You will be expected to get into the habit of methodical, well-planned and organized work by the mid-term. This will help you with the experiments in the second half of the course.**

Course Website

This course is on WebCT (<https://webctweb.ucsd.edu>) and should automatically appear on your WebCT account as soon as you register for the class. We will use WebCT to post information on experiments, exams, schedules, readings and practice material, experimental data, report guidelines, etc. We strongly encourage you to use the Discussion board to post questions or answers to questions and to use it as a forum for exploring the material. The TAs and I will routinely check this website and answer any questions but feel free to respond as well. This website will also be used to post any announcements that pertain to the entire class. Please check the site regularly and update yourself on the information provided.

University Policy on Integrity of Scholarship

The principle of honesty must be upheld if the integrity of scholarship is to be maintained by an academic community. The University expects that both faculty and students will honor his principle and in so doing protect the validity of University grading. This means that all academic work will be done by the student to whom it is assigned, without unauthorized aid of any kind. Instructors, for their part, will exercise care in planning and supervising academic work, so that honest effort will be encouraged.

Student Responsibility:

Students are expected to complete the course in compliance with the instructor's standards. No student shall engage in any activity that involves attempting to receive a grade by means other than honest effort; for example:

- No student shall knowingly procure, provide, or accept any unauthorized material that contains questions or answers to any examination or assignment to be given at a subsequent time.
- No student shall complete, in part or in total, any examination, or assignment for another person.
- No student shall knowingly allow any examination or assignment to be completed, in part or in total, for himself or herself by another person.
- No student shall plagiarize or copy the work of another person and submit it as his or her own work.
- If any work is plagiarized from that of another student, both students will be reported to the Office of Academic Integrity, even if one of the students has graduated already. Remember that most graduate schools check the undergraduate records for any indications of dishonesty before awarding a degree.
- No student shall alter graded class assignments or examinations and then resubmit them for regrading.
- No student shall submit substantially the same material in more than one course without prior authorization.

Lab No:	Date	Day	Laboratory Experiments	Remarks
1	July 3 rd	Tue	<p>I. Registration, introductory remarks, safety lecture, etc.</p> <p>II. Sterile technique.</p> <ul style="list-style-type: none"> • Microbes in the environment • <i>E.coli</i> and toilet paper experiment • Aseptic technique, streak and spread plates <p>III. Use of pipettes</p> <ul style="list-style-type: none"> • Demo • Pipette calibration 	
	July 4 th	Wed	Holiday	
2	July 5 th	Thurs	<p>I . Microscopy</p> <ul style="list-style-type: none"> • Learning to focus the light microscope • Calibrating your microscope • Observing stained slides <p>II. Understanding dilutions:</p> <ul style="list-style-type: none"> • Understanding dilutions- theory <p>III. Observe Results:</p> <ul style="list-style-type: none"> • <i>E.coli</i> and toilet paper experiment: • Streak and spread plates 	
3	July 6 th	Fri	<p>I. Microscopy:</p> <p>Making a wet mount and Phase Contrast Microscopy: Wet mounts (hay infusion) and phase contrast:- view, identify, and measure</p> <p>II. Observe Results :</p> <p>Sterile technique - Microbes in the environment</p> <p>III. Measuring microbial growth: Yeast</p> <ul style="list-style-type: none"> • Direct counts using a hemocytometer • Using a spectrophotometer • Counting viable cells using plating 	

4	July 10 th	Tue	<p>I. Measuring microbial growth: Yeast</p> <p>Counting viable cells using plating –Complete (Analyze data)</p> <p>II. Soil Enumeration and Enrichment: Lab Period 1</p> <p>A. Simple Enumeration: Serial dilution, plating on TSA, SDA, GAA, and MacConkey, high temp, casein and High salinity</p> <p>B. Enrichment</p> <ul style="list-style-type: none"> ○ Enrichment: Inoculate medium as assigned 	<p>HW1 = <i>E. coli</i> and toilet paper experiment</p> <p>Lab Report presentation by TAs</p> <p>Data Analysis workshop</p>
5	July 11 th	Wed	<p>I. Characterizing an Unknown Organism:</p> <p>Receive an unknown organism: wet mount and streak plate and slants.</p> <ul style="list-style-type: none"> • MacConkey – Inoculate • Sticky test • Endospore test – inoculate NSM • TSS – Transfer <p>II. Microscopy: Staining</p> <p>Smear preparation – Simple staining</p> <p>III. Macronutrient use – how organisms get energy to survive: Introduction: Hydrolysis and use of large extracellular materials</p> <ul style="list-style-type: none"> • Polysaccharides: Starch plates – inoculate. • Proteins: Skim milk plates and gelatin deeps - inoculate • Lipids: Rhodamine plates – inoculate <p>IV. Bioinformatics lab Introduction</p>	
6	July 12 th	Thurs	<p>Microscopy: Staining</p> <ul style="list-style-type: none"> • Smear preparation and simple staining • Gram stain: control organisms only <p>Characterization of an unknown Organism:</p> <ul style="list-style-type: none"> • Gram stain 	

			Soil Enumeration/Enrichments – Examine plates for any growth – Especially high temp Enrichment – Sub culture	
7	July 13 th	Fri	I. Characterizing an unknown Organism: <ul style="list-style-type: none"> • streak plate and slants – Examine results, make a wet mount – observe by phase contrast • MacConkey – Examine results • Endospore test – Examine results • Polysaccharides: Starch plates – complete • Proteins: Skim milk plates and gelatin deeps – complete. • Lipids: Rhodamine plates – complete II. Soil Enumeration/Enrichments – Examine plates for any growth III. Nitrogen fixation: Free-living – Anabaena - Inoculate BG11 and BG11-0 with <i>Anabaena</i>	HW2 – Dilution problem set
8	July 17 th	Tue	How energy is produced – aerobic vs. anaerobic breakdown of organic compounds <ul style="list-style-type: none"> • Oxygen requirements – inoculate thioglycollate tube • Acid and gas production from sugar fermentation – inoculate • Methyl-Red and Voges-Proskauer – inoculate • T-streak plate for fresh isolated colonies • Nitrate reduction - inoculate • H₂S production – inoculate Motility – inoculate plate and deep with unknown organism	Midterm – I (in lab)

			Special metabolic functions: Standards only <ul style="list-style-type: none"> • Indole production from tryptophan, catabolite repression – inoculate • Urease test – inoculate • Differential utilization of citrate by enterics - inoculate 	
9	July 18 th	Wed	Soil Simple Enumeration Count colonies on all plates -calculate Soil Enumeration and Enrichment: <ul style="list-style-type: none"> • Serial dilutions and plating to enumerate enrichment. • Centrifuge aliquot of enrichment and freeze pellet Characterization of an Unknown Organism: <ul style="list-style-type: none"> • Oxygen requirements –complete • H₂S production – Kligler - examine and reincubate Characterization of a Unknown organism: Survival in extreme conditions: inoculate appropriate broth with Unknown organism <ul style="list-style-type: none"> • pH • Temperature • Salt 	
10	July 19 th	Thurs	Characterization of an Unknown Organism: How energy is produced – aerobic vs. anaerobic breakdown of organic compounds <ul style="list-style-type: none"> • Nitrate reduction – complete test • H₂S production – complete test • Cytochrome C test – complete • Catalase test – complete 	Data Analysis – Computer Lab

			<ul style="list-style-type: none"> • Acid and gas from sugar fermentation - complete • Methyl-Red and Voges Proskauer – complete • Indole production from tryptophan, catabolite repression – complete • Urease test - complete • Differential utilization of citrate by enterics – complete • Motility – complete 	
11	July 20 th	Fri	<p>Characterization of an Unknown organism:</p> <p>Survival in extreme conditions: Observe results</p> <p>Soil Enumeration and Enrichment:</p> <p>Enrichment: Complete colony counts and calculations</p> <p>Metagenomics: Lab Period 1</p> <ul style="list-style-type: none"> • Step 1: Chromosomal DNA preps from frozen cell pellets from various soil enrichments <p>Step 2: Set up 16S rRNA PCR</p> <p>Discuss in-class – Unknown Characteristics</p> <p>Complete ALL DATA collection</p>	Data analysis – Computer lab Dichotomy tree
12	July 24 th	Tue	<p>I. Metagenomics::</p> <ul style="list-style-type: none"> • Step 2a: run gel of PCR reactions • Step 3: Evaluate PCR results • Step 4: Purify PCR product <p>Screening for Antibiotic Producers: Identify antibiotic producers</p> <p>Evaluation of antibiotics by the Kirby Bauer method</p> <p>Spread plates with standards ---- efficiency of antibiotics</p>	Lab report – I submission

13	July 25 th	Wed	Metagenomics: Lab Period 3 <ul style="list-style-type: none"> • Step 5: Set up ligations in pGEM-T • Step 6: Transform ligations and plate on selective media Screening for Antibiotic Producers: Identify antibiotic producers, measure ZOI Evaluation of antibiotics by the Kirby Bauer method – Observe results	
14	July 26 th	Thurs	Metagenomics: Lab Period 3 <ul style="list-style-type: none"> • Step 7: Select white colonies and streak out for sequencing 	Computer Lab Practice sequence analysis Organize data for lab report II
15	July 27 th	Fri	Growth curve experiment Growth and graphing of <i>Vibrio natriegens</i> <ul style="list-style-type: none"> • Note to TA/self: Send for sequencing by Friday 	
16	July 31 st	Tue	Transposon Mutagenesis: Lab period I Step I: Set up conjugation of <i>E. coli</i> and <i>Salmonella</i> Metagenomics: Lab Period 4 <ul style="list-style-type: none"> • Analysis of sequence data – computer lab 	Computer lab
17	Aug 1 st	Wed	Metagenomics: Lab Period 4 (contd...) <ul style="list-style-type: none"> • Analysis of sequence data – computer lab Transposon Mutagenesis: Lab period II Step 2: Plate exconjugants for selection and counter selection Save LB recipient control plates for later use Yogurt: Inoculate milk with starter yogurt	

18	Aug 2 nd	Thurs	Transposon Mutagenesis: Lab period III Count colonies and calculate transposition efficiency Nitrogen Fixation I. Free-living: <i>Anabaena</i> : check for heterocysts II. Symbiotic Rhizobium/Clover Observe nodules	Lab Report II
19	Aug 3 rd	Fri	Yogurt: measure pH, gram stain	Midterm II