## Priya Narasingarao

HSS 1145A

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Office hours: Tuesdays 10AM to 11AM

Lecture: MTWTH 11AM to 12:20pm in Warren Lecture Hall (WLH) 2207

**Labs**: York 2310 and 2332 MTWTH 12:30pm to 4:30pm

### **Teaching Assistants**

• Jennifer Phun - jephun@ucsd.edu

• Deron Amador - dtamador@ucsd.edu

#### **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 lab period or the first 10 minutes of the lecture period of the day on which your report is due. Check with your instructor as to where the report should be turned in. 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 results 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 25

Pop quizzes	50
Lab evaluation (continuous assessment)	25
Lab report - 1	100
Lab Report – 2	100
Midterms (I and II)	100
Final Exam (cumulative)	100
Final Total	500

## Possible assignments.

If an assignment is given, either an exam or a report will have its point value correspondingly reduced.

- Dilution assignment
- Metagenomics assignment

### **Grade Distribution**

A = 90% - 100%

B = 80% - 89.9%

C = 70% - 79.9%

D = 60% - 69.9%

F = below 60%

### **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
  - o Note who did which part of the procedure who inoculated controls, etc
  - O Note which organisms you used name and species of the controls, etc
  - o Errors
- Observations
  - o Write
  - o Draw
  - Questions and connections

- o Conclusion or summary
- o 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	6/27/2011	Mon	I. Registration, introductory remarks, safety lecture, etc.	
			II. Sterile technique.	
			<ul> <li>Microbes in the environment</li> <li><i>E.coli</i> and toilet paper experiment</li> <li>Aseptic technique, streak and spread plates</li> </ul>	
			III. Use of pipettes	
			<ul><li>Demo</li><li>Pipette calibration</li></ul>	
2	6/28/2011	Tues	I . Microscopy	
			<ul><li>Learning to focus the light microscope</li><li>Calibrating your microscope</li><li>Observing stained slides</li></ul>	
			II. Understanding dilutions:	
			Understanding dilutions- theory	
			III. Observe Results:	
			<ul> <li><i>E.coli</i> and toilet paper experiment:</li> <li>Streak and spread plates: Observe results.</li> </ul>	
3	6/29/2011	Wed	I. Microscopy:	
			Making a wet mount and Phase Contrast Microscopy: Wet mounts and phase contrast:- view, identify, and measure	
			II. Observe Results :	
			Sterile technique - Microbes in the environment	
			III. Measuring microbial growth: Yeast	
			Direct counts using a hemocytometer	
			Using a spectrophotometer	
			Counting viable cells using plating	
4	6/30/2011	Thurs	I. Measuring microbial growth: Yeast	Submit Notebook
			Counting viable cells using plating –Complete (Analyze data)	sheets to TA

			II. Soil Enumeration and Enrichment: Lab	
			Period 1	
			A. <b>Simple Enumeration:</b> Serial dilution, plating on TSA, SDA, GAA, and	
			MacConkey	
			B. Extracellular degradation:	
			o Enumeration: Serial dilution	
			and plating of soil sample on	
			starch/skim milk.	
			o Enrichment of soil	
			organisms: Inoculate minimal	
			media containing starch/skim	
			milk with soil.	
			C. Extreme conditions	
			<ul> <li>Enrichment: Inoculate medium as assigned</li> </ul>	
	7/4/2011	Mon	Holiday	
5	7/5/2011	Tues	I . Characterizing an Unknown Organism:	
			Receive a test organism: wet mount and	
			streak plate and slants.	
			MacConkey – Inoculate	
			Sticky test	
			• Endospore test – inoculate NSM	
			• TSS – Transfer	
			II. Macronutrient use – how organisms get	
			energy to survive: Introduction:	
			Hydrolysis and use of large extracellular	
			materials	
			Polysaccharides: Starch plates –     inoculate.	
			Proteins: Skim milk plates and gelatin	
			deeps - inoculate	
			Lipids: Rhodamine plates - inoculate	
	7/6/0011	*** 1	II. Bioinformatics lab Introduction	
6	7/6/2011	Wed	Microscopy: Staining	
			Smear preparation and simple staining	
			Gram stain: control organisms only	
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			Characterization of an unknown Organism:	
			Gram stain	
			Soil Enumeration/Enrichments – Examine plates for any growth	
7	7/7/2011	Thurs	<ul> <li>I. Characterizing an unknown Organism:</li> <li>streak plate and slants – Examine</li> </ul>	Submit Notebook sheets to TA
			results, make a wet mount – observe	
			by phase contrast	
			<ul> <li>MacConkey – Examine results</li> <li>Endospore test – Examine results</li> </ul>	
			Polysaccharides: Starch plates –	
			<ul> <li>complete</li> <li>Proteins: Skim milk plates and gelatin deeps – complete.</li> </ul>	
			• Lipids: Rhodamine plates – complete	
			II. Soil Enumeration and Enrichment: Lab Period 2	
			Simple enumeration: colony counts	
			Extracellular degradation:	
			Enumeration: count	
			Enrichment: subculture	
			Extreme conditions	
			Enrichment: Subculture	
			III. Nitrogen fixation: Free-living – Anabaena - Inoculate BG11 and BG11-0 with Anabaena	
8	7/11/2011	Mon	How energy is produced – aerobic vs.	Midterm -I
			anaerobic breakdown of organic	
			compounds	
			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	
			H <sub>2</sub> S production – inoculate	
			Motility – inoculate plate and deep with test	
			organism	
			Special metabolic functions: Standards only	
			Indole production from tryptophan, catabolite repression – inoculate	
			Urease test – inoculate	
			Differential utilization of citrate by enterics - inoculate	
9	7/12/2011	Tues	Soil Enumeration and Enrichment: Lab Period – 3	
			Extracellular degradation:	
			Enrichment:	
			Serial dilutions and plating to enumerate enrichment.	
			Centrifuge aliquot of enrichment and freeze pellet	
			Extreme conditions	
			Enrichment:	
			Serial dilution and plating of enriched	
			sample.	
			Centrifuge aliquot and freeze pellet	
			Characterization of an Unknown	
			Organism:	
			Oxygen requirements –complete	
10	7/13/2011	Wed	Characterization of an Unknown	
			Organism:	
			How energy is produced – aerobic vs.	
			anaerobic breakdown of organic	
			compounds	
			Nitrate reduction – complete test	
			H <sub>2</sub> S production – complete test	
			1120 production complete test	

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			Cytochrome C test – complete	
			• Catalase test – complete	
			Acid and gas from sugar fermentation - complete	
			Methyl-Red and Voges Proskauer – complete	
			• Indole production from tryptophan, catabolite repression – complete	
			Urease test - complete	
			<ul> <li>Differential utilization of citrate by enterics – complete</li> </ul>	
			• Motility – complete	
11	7/14/2011	Thurs	<b>Soil Enumeration and Enrichment</b> : Lab Period 4	1. Submit Notebook sheets to TA
			Extracellular degradation:	10 1A
			Enrichment: Complete colony counts	
			and calculations	
			Extreme conditions	
			Enrichment: Complete colony counts and calculations	
			Discuss in-class – Unknown Characteristics	
12	7/18/2011	Mon	Metagenomics: Lab Period 1	
			Step 1: Chromosomal DNA preps from frozen cell pellets from various soil enrichments	
			Step 2: Set up 16S rRNA PCR	
13	7/19/2011	Tues	I .Characterization of a Test Organism:	Submit lab
			Survival in extreme conditions: inoculate appropriate broth with Unknown organism  • pH  • Temperature	report I – SOIL Analysis
			• Salt	
			II. Metagenomics::	
			Step 2a: run gel of PCR reactions	
			Step 2a. Tull get of PCK reactions	

			Step 3: Evaluate PCR results	
			Step 4: Purify PCR product	
1.4	7/20/2011	337 1	M. 4	MIDT
14	7/20/2011	Wed	Metagenomics: Lab Period 3	MID Term II
			• Step 5: Set up ligations in pGEM-T	
			Step 6: Transform ligations and plate on selective media	
15	7/21/2011	Thurs	Metagenomics: Lab Period 3	Submit Notebook
			Step 7: Select white colonies and streak out for sequencing	sheets to TA
			• Note to TA/self: Send for sequencing by Friday	
			Characterization of a Test Organism:	
			Survival in extreme conditions:	
			Score growth/no growth in each tube	
16	7/25/2011	Mon	Screening for Antibiotic Producers: Identify	Submit lab report II –
			antibiotic producers, measure ZOI	UNKNOWN
			Evaluation of antibiotics by the Kirby  Bauer method	Test –
			Spread plates with standards efficiency of	Identification and
			antibiotics antibiotics	Characterization
17	7/26/2011	Tues	Metagenomics: Lab Period 4	
			<ul> <li>Analysis of sequence data – computer lab</li> </ul>	
			Yogurt: Inoculate milk with starter yogurt	
18	7/27/2011	Wed	Evaluation of Antibiotics by the Kirby Bauer Method	
			Measure ZOI, identify any resistant colonies	
			Growth curve experiment	
			I. Growth and graphing of <i>Vibrio</i> natriegens	
			Nitrogen Fixation	
			II. Free-living: Anabaena: check for	

			heterocysts	
19	7/28/2011	Thurs	Yogurt: measure pH, gram stain	1. Submit Notebook sheets to TA 2. Final Exam