# Microbiology Lab Students' Manual

BIOL4040/5040/7040, 1 credit Spring 2013 Wolfe Hall 1214

Wednesday 9:00-11:50 am (section 003)

2:00-4:50 pm (section 001)

6:30-9:50 pm (section 002)

Thursday 1:00-3:50 pm (section 004)

#### **INSTRUCTOR:**

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#### **TEACHING ASSISTANTS:**

Jeffrey Zahratka (section 003) Matt Clark (section 001) Lindy Lutz (section 002) Jun Zhou (section 004)

#### **OFFICE HOURS**

Contact the instructor by email or by appointment. Consult with TAs for their individual office hours.

#### **COURSE DESCRIPTION**

This lab course examines basic microbiological techniques. Students will be exposed to different methods to identify, isolate and culture microorganisms, protocols to study basic biochemical and molecular biological properties of microorganisms, and approaches to control microorganisms. The goal is to provide basic trainings in common microbiological techniques that are up-to-date and are widely used in clinical and research labs. This laboratory manual was reorganized and rewritten based on previous manuals by Drs. Scott Leisner, John Gray and Louis Glatzer at the Department of Biological Sciences of the University of Toledo. Your input for any possible further modifications is welcome.

**Please note** although the hands-on time for each lab is usually less than 1 hr, the students sometimes need arrange time before or after the lab day to do certain preparations or observe results. The instructor and the TAs make great efforts to reduce these extra visits but some are unavoidable due to the nature of a microbiology lab.

2013 Spring Laboratory Schedule

Please note: this is a tentative syllabus and it may be changed during the course of the semester.

Lab

Week	Lab No.	Date	Lab Title					
Part I Basic Techniques								
1	1	1/9-10	The rules, the media, the tools, and aseptic techniques					
2	2	1/16-17	Isolation of A Pure Culture					
Part II Microbial Physiology and Characterization of Microorganisms								
3	3	1/23-24	Bacterial Growth Curve and liquid culture					
4	4	1/30-31	Introduction To Staining, basic microscopy					
5	5	2/6-7	Bacterial Growth On Specialized Media					
6	6	2/13-14	Bacterial Enzymes					
7	7	2/20-21	Immunological and Molecular characterization of microorganisms					
8		2/27-28	Mid-term exam					
9		3/6-7	Spring Break, No Lab					
Part III Bacterial Genetics								
10	8	3/13-14	Induction Of Bacterial Mutants					
11	8 9	3/20-21	Mutant Characterization Bacterial Responses To Antibiotics					
12	10	3/27-28	Molecular Basis Of Antibiotic Resistance					
13	11	4/3-4	DNA transformationGreen Bacteria!					
Part IV Control of Microorganisms								
14	12 13	4/10-11	Microbes And Man Mammalian Defenses Against Microbial Attack					
15	14	4/17-18	Effects Of Environmental Conditions On Bacterial Growth					
16		4/24-25	Lab Practical Final Exam					
17		5/1-5/2	Finals Week, No Lab					

### Just for fun:

There are two optional experiments if any student volunteers to perform extra experiments (sorry, no extra credits!). This can be scheduled after week 14. Directly contact the instructor so we can make arrangements if there is enough interest.

- 1. Fermentation of wine or beer: Volunteers need to bring in grape juice and malt extract and commercial yeast. Utensils, tools and incubators will be provided.
- 2. Making yogurt: Volunteers need to bring yogurt (the varieties containing live and active cultures) and milk.

## Grading

Grades for this course will be based upon:

Quizzes, twelve worth 10 pts each		100
Lab Notebook, worth 140 pts, 10 points each experiment		140
Lab Performance, worth 140 pts, 10 points each experiment		140
Lab Practical mid-term exam worth 60 points		
Lab Practical final exam worth 60 points		60
	Total points:	500

The total points will be converted into % and letter grades will be given according to the following table.

% of available marks	Grade	Standard		
<u>≥</u> 90	A	Achievement of outstanding quality		
≥88	A-	Achievement of slightly less than outstanding		
quality				
≥85	B+	Achievement of slightly more than high quality		
≥78	В	Achievement of high quality		
≥75	B-	Achievement of slightly less than high quality		
≥72	C+	Work of slightly more than acceptable quality		
≥64	C	Work of acceptable quality		
≥62	C-	Work of slightly less than acceptable quality		
≥61	D+	Work slightly above the quality expected		
≥52	D	Work below the quality expected		
≥50	D-	Work slightly below the quality expected		
< 50	F	Fail		

### **Notes on Grading**

#### I. Quizzes

There will be one quiz each lab week except the first meeting and exam weeks. This is a total of 12 quizzes but your quiz grade will be based on your best 10 scores. In other words, you get to drop your lowest 2 quiz scores. Quizzes will be based on materials from the previous lab and the materials for the lab of that day. Quizzes will be given at the beginning of the class and no make-up quiz will be given if you are more than 5 minutes late for class.

#### II. Lab Notebook

A notebook with a sewn binding (no spiral binding or loose-leaf paper) that should catalog what you have done in the lab. It should be organized in the following manner:

- -Lab title and date
- -Any changes in the protocols for each experiment---You should read the lab manual before starting any experiment. Note down how you exactly did the experiment.
  - -Data for each experiment. Any observations you made during the lab.
- -Conclusions for each experiment. Include discussions when necessary (i.e. in case of failed experiments).

The entries for the notebook should be finished in class after each experiment unless your instructor/TA tells you otherwise (Results for some experiments need wait for 24~48 hrs or longer). After each experiment, you should let the TA check your notebook before you leave the lab. No make-up is allowed. See Appendix A for assistance with organizing your notebook. **Use** 

of new technology to record your results (digital images, videos etc) is encouraged but should not interfere with the order in the class and should be consulted with the TAs. The digital results may be emailed to the TA and/or Dr. Song-Tao Liu for future teaching references (you will be acknowledged but no copyright claims please).

#### III. Lab Performance

Because this is a laboratory course, you must have a practical working knowledge of various techniques such as proper sterile technique, the Gram stain, labeling your plates correctly, as well as cleaning up your laboratory area. It is essential that each person must pull their weight even though you will be working in groups of two or four. **Individual performance will be evaluated by your instructor/TA during every experiment**. For obvious reasons no make-up is allowed if you miss experiments.

#### IV. Lab Practical Exams

Two lab practical exams will test your understanding of the various techniques and organisms examined during the semester. The mid-term exam will be scheduled in Week 8 (2/27-28, 2013), and the final exam will be in week 16 (4/24-4/25, 2013). The final exam will be comprehensive. The exams will be based on quizzes and lab practices, so make sure you review the answers to quizzes and get familiar with proper lab techniques.

#### V. Attendance

Laboratory experiments will begin promptly and most require the full class time. Instructions and advice will be given at the outset of the lab as well as the quizzes. Therefore, it is critical for you to be in class on time. If you must miss a lab, make sure to notify both your lab instructor/TA and your lab partner prior to the lab. You can get a make-up quiz if you have legitimate excuses for absence. Again, since this is a hands-on lab course, no make-up is allowed for lost points on lab notebook and lab performance.

#### VI. Preparation

In most experiments you will be working with live bacteria and viruses and certain of these organisms can be pathogenic. It is important for you to understand the purpose of the experiments, the sequence of steps and operation of equipment before you begin the experiment. Therefore, it is essential that you read through the lab materials prior to attending the lab. The quizzes will contain questions pertaining to the procedure that you are about to perform in order to encourage you to understand the lab before you begin. There are also a number of basic rules that you must follow to allow the course to be run safely and smoothly. These rules are on the next two pages.

### VII. Group work

All experiments are to be done in groups of two unless otherwise specified. Be sure to come prepared for the lab for your own benefit and for that of your partner. Even though the students will be working in groups of two and the results of experiments will be shared, the performance of each student will be evaluated individually. Therefore, it is important to earn your own "hands-on" performance points during each experiment. Specifically, each student must demonstrate their **competence to isolate a pure culture** and **to develop a good Gram stain** and your instructor will note this information in their records. Failure to accomplish this task will cost you a letter grade.

### **Microbiology Laboratory Rules**

Most of these rules are common sense. Since you are working with microorganisms, and some of which could be pathogenic (i.e., disease causing), it is very important to take several precautions. Be careful working with these organisms for both yourself and for others that may have contact with you! How well you follow these rules is an important part for your lab performance scores.

- 1. Under **NO** circumstances is eating, drinking, or gum chewing allowed in the laboratory.
- 2. Coats and other clothing accessories should be left on the coat rack or designated area only.
- 3. You are required to have the following lab supplies available for each lab:
  - 1 "Sharpie" type permanent marking pen.
  - 1 Lab notebook

A lab coat is suggested but not mandatory (You are required to keep it clean though. If necessary, you need to wash it at home).

4. A drawer will be assigned to each group. The instructor will unlock and lock the drawers for each lab section. You should keep the following lab supplies in you drawer.

Lab coat

"Sharpie" type permanent marking pen.

Gram staining kit (Supplied)

Inoculating loops (Supplied)

Glass spreaders (Made by each student or supplied)

- 5. Purses, Book bags, textbooks, and notebooks may be placed under the counters or in your assigned lab drawers but never on the counter or shelf above the counter.
- 6. Because this course is MICRObiology, it is essential that proper care of <u>microscopes</u> be adhered to. All microscopes should be cleaned, covered, and placed in the appropriate location. Abuse of microscopes will not be tolerated!
- 7. <u>Labeling</u>: All labeling is to be done with an organic solvent-based (non-water based) "Sharpie" type permanent marking pen.

Petri plates: Should be labeled on the <u>bottom</u> with your name, dates, and other necessary information.

Test tubes: May be labelled on the glass surface of the tubes or on a piece of tape put onto the glass surface of the tube but **NEVER** on the cap.

Test tube racks: Are to be labelled with masking tape but no markings should be made directly on the rack itself with any form of marker.

- 8. <u>Contaminated material</u> is to be placed in the proper containers at the completion of the experiment: **NEVER** put contaminated material in regular waste baskets or sinks or on the countertop.
- 8a. Slides containing microbes are to be put into a slide morgue or glass container labeled "contaminated" after the experiment is completed.

- 8b. Plastic petri plates inoculated with microbes should be discarded in the **orange** biohazard bags upon completion of experiments.
- 8c. Contaminated/used pipette tips (except Pasteur Pipettes) are to be placed into pipette jars labeled "Bleach Morgue."
- 8d. Contaminated Pasteur pipettes are to be placed tip down in glass containers labeled "Lysol Morgue."
- 8e. All other contaminated materials should be discarded on the metal cart provided for this purpose.
- 8f. **NEVER** return opened or used materials to the supply bench without telling your lab instructor.
- 9. Hands should be washed before and after lab work.
- 10. Lab countertops must be cleaned with disinfectant (Roccal) both before and after lab work.
- 11. You will be using some containers filled with bleach. This chemical kills bacteria very quickly. However, it can also stain your clothes so be careful when using it.
- 12. For many experiments, you will be using a flame to sterilize materials. NEVER LEAVE A FLAME UNATTENDED!!!!
- 13. Immediately report any accidental spill of microbial cultures to your lab instructor.

### STATEMENT ON ACADEMIC DISHONESTY

### **Department of Biological Sciences**

Academic dishonesty by students enrolled in undergraduate or graduate courses and programs offered by the Department of Biological Sciences will not be tolerated. Academic dishonesty includes but it not limited to:

- 1. Obtaining assistance from another individual during an examination.
- 2. Giving assistance to another individual during an examination.
- 3. The unauthorized use of study material or textbooks during an examination.
- 4. Changing answers on an examination after it has been returned and then submitting it for re-grading.
- 5. Plagiarizing written assignments. Plagiarizing includes but is not limited to: a) Copying laboratory reports from previous years, b) copying or paraphrasing reports, term papers, or theses prepared by other students, c) unauthorized collaboration in the preparation of reports, term papers, or theses, and d) use of another author's materials without appropriate acknowledgment through quotation and citation.
- 6. Attempting to bribe or otherwise induce an instructor to alter either a grade or examination score.
- 7. Obtaining or attempting to obtain a copy of an examination prior to its administration.

In accordance with policies presented in The Student Handbook and The University Catalog, Instructors have the responsibility and right to report cases of alleged dishonesty to departmental, college, and university administrative units. Students involved in academic dishonesty may expect to receive a grade of F on specific assignments as well as in the course where the assignment was made. In addition, disciplinary action may be recommended through appropriate college and university disciplinary committees. Please consult your instructor for instructions on the implementation of this policy.