Course Syllabus: Bacteriology

General Information:

Course Meetings: TR (11:00 am – 12:20 pm) Bolin Hall 213
Instructor: James Masuoka, Ph.D.
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Telephone: 397-4181
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Office Hours: MWF 9:00 – 10:00 am
TR 9:00 – 10:30 am
(Other times by appointment)

Course Description (from the catalog):

Study of bacteria and archaea cell structure and function, bioenergetics, and regulation of cellular functions; survey of adaptations to diverse environments.

For fall semester 2022: Emphasis will be placed on mechanisms of bacterial pathogenesis as it pertains to the human host. Topics will include: the relationship between bacteria and the host generally; cellular structures and products that directly or indirectly impair host function; mechanisms by which bacteria acquire virulence genes; mechanisms by which bacteria evade the immune response, antibiotics, and viral attack; how pathogenicity and virulence are studied experimentally

Required Texts:

Bacterial Pathogenesis: A Molecular Approach (Wilson, Winkler, Ho), 4th Edition (2019).ASM Press. ISBN 9781683672883 (eBook), 9781555819408 (pbk.)

Composition book (for laboratory notes), Lab coat (gloves & safety glasses will be provided)

Course Objectives (Lecture):

- Define microbiota/microbiome. Explain how the human microbiome composition is characterized.
- Explain how Koch's Postulates help us understand infectious disease. Describes cases in which Koch's Postulates cannot be satisfied.
- Describe experimental approaches to defining and characterizing virulence factors of bacteria.
- Describe bacterial cell structures and relate these structures to function, particularly those related to pathogenesis.

- Describe the mechanisms by which bacteria can acquire virulence traits through genetic modification and exchange and how expression of these traits is regulated.
- Explain how these virulence factors cause disease (damage to the host).
- Describe mechanisms by which antibacterial agents (chemicals, viruses) act and how bacteria protect themselves from these agents
- Describe strategies used by bacteria to colonize and survive within the host, including ways they evade the host immune response.
- Research and critically evaluate scientific literature to answer or address a question
- Communicate fundamental concepts of microbiology, both in written and in oral formats

Classroom Expectations and Policies:

- Students are expected to be prepared for lecture and lab by 1) reading the text, lab manual and handouts prior to coming to class; 2) having paper and pen at hand
- Students are expected to arrive a few minutes early to mentally prepare. If late arrival is unavoidable, the student should enter the class in a manner that creates as little disruption as possible.
- Points will be deducted from assignments turned in late.
- Student Conduct: Please refer to the MSU Student Handbook: (https://mwsu.edu/Assets/documents/student-life/student-handbook-2017-18.pdf) for university policies related to student responsibilities, rights, and activities. For example, see page 73 for valid grounds for an instructor drop (excessive absence, indifferent attitude, disruptive conduct, failure to meet class assignments), page 13 for the university's code of student conduct and page 55 for definitions of academic dishonesty that may be subject to disciplinary action (cheating, plagiarism, and collusion). In this class, academic dishonesty on an assignment or exam will minimally result in a score of 0 for that assignment or exam. Depending on the magnitude or frequency of these types of infractions, more severe sanctions – including being dropped from the course – will be imposed.
- **CELL PHONES (and other electronic devices): (READ THIS TWICE, PLEASE)** This class, as well as your other classes, requires your engagement, and typical cell phone use serves to detract from that engagement. While in class, your phone should be on “silent” (NOTE: vibrate is NOT silent).
- **Other electronic devices: laptops, tablets and similar devices will no longer be permitted to be used during lectures.** The ability to take good notes is a skill that university students must be able to master. Further, classroom studies have shown that taking notes by hand increases engagement in the material. Simply transcribing the lecture word-for-word is not helpful.
- Students with disabilities: It is the responsibility of the student to first contact Disability Support Services and then the instructor to determine what accommodations might be made for a disability. It will be the responsibility of the

student to plan to acquire notes. Any requests for accommodation must be made 2 weeks prior to the first exam.

- The professor considers this classroom to be a place where you will be treated with respect as a human being - regardless of gender, race, ethnicity, national origin, religious affiliation, sexual orientation, political beliefs, age, or ability. Additionally, diversity of thought is appreciated and encouraged, provided you can agree to disagree. Furthermore, guns or other weapons create a coercive environment that is neither safe nor conducive to learning. Therefore, weapons of any kind will not be permitted in my classroom. This includes guns, concealed or otherwise, regardless of licensure. Any student bringing a weapon to class or to lab will be immediately dropped from the course. It is the professor's expectation that ALL students consider the classroom a safe environment.
- The instructor reserves the right to amend these rules as needed throughout the term.

E-mail Policy:

I will respond to e-mail during regular school hours (8:30 am – 5:00 pm M-F). I will make every effort to respond to e-mail sent during the week within 24 hours. Those sent over the weekend will be attended to on Monday. Always include a subject line in your e-mail messages. It would be particularly helpful to include in the subject line the course number & section (*i.e.,* BIOL 3044). Questions regarding simple matters of class schedule or those that can otherwise be answered from information in this syllabus will be given low priority.

Attendance Policy:

Students are expected to attend all meetings of the classes in which they are enrolled. Although in general students are graded on intellectual effort and performance rather than attendance, absences may lower the student's grade where class attendance and class participation are deemed essential by the faculty member. In those classes where attendance is considered as part of the grade, the instructor should inform students of the specifics in writing at the beginning of the semester in a syllabus or separate attendance policy statement. An instructor who has an attendance policy must keep records daily. The instructor
must give the student a verbal or written warning prior to being dropped from the class. Instructor's records will stand as evidence of absences. A student with excessive absences may be dropped from a course by the instructor. Any individual faculty member or college has the authority to establish an attendance policy, providing the policy is in accordance with the General University Policies. [MSU Student Handbook, p. 61]

Absences will be excused for:

a. **Death of an immediate family member.** An immediate family member is a grandparent, parent, sibling, spouse, in-law, aunt, uncle, or child.

b. **Summons to appear in court or jury duty.** A copy of the summons is required.
c. **Call to military service.** A copy of your orders to report is required.
d. **University sponsored event.** Members of athletic teams, college bowl participants, etc. will be excused with proper notification.
e. **Debilitating illness or disability.** Illnesses will be addressed on an individual basis. If a student is affected by an illness that is not debilitating, (*i.e.,* flu, virus infection) which may result in the student missing one or more consecutive class sessions, that student will be marked as unexcused for the number of days missed **unless a doctor's note is provided**.

ROUTINE APPOINTMENTS, medical or otherwise, AND VACATION TRAVEL ARE NOT ACCEPTABLE reasons for excused absences.

Students who feel ill (esp. with fever or symptoms of COVID-19) should stay home and isolate themselves. Notify the instructor immediately.

It is the responsibility of the student to obtain notes or other information covered in class during an absence.

Exam Policies:

- No make-up exams will be given in this course. If you must miss class during a midterm exam period, and it is an excusable absence (see above), then the cumulative portion of the final exam will be used to determine the missed midterm score. You must notify the instructor of problems **prior to** the start of the exam and provide the appropriate documentation as soon as possible. Only one midterm exam will be substituted for in this manner.

- Exams are not moved for congested midterm or finals schedules.

- **All electronic devices**, including cell phones, **must be completely turned off** during exam periods. I will keep track of time and relay the information to the class. If you must keep track of time yourself, invest in a wristwatch.

Grading:

All exams and assignments count toward your final grade in the course and so it is important to do the best that you can on everything you turn in. If you find yourself having difficulties, please come to me for help early in the semester so that you give yourself time to improve.

Attendance is not a direct component of your course score. However, continual tardiness – and the subsequent class disruption due to coming in late – will be considered and may influence your final points awarded. As stated above, cell phone

use distracts from attention in class. Therefore, students who persist in using their mobile devices during class, except for designated classroom activities, will be marked as absent.

This course is not graded on a traditional curve. The course is worth 1000 points. Grade categories and equivalent percentages are as indicated: A (90-100%); B (80-89%); C (70-79%); D (60-69%); F (59% and below). Passing requires 60% of the points (unadjusted) for the course, or 600. Fractional percentages will be rounded at the end of the semester.

Lectures constitute approximately 60% of the BIOL 3044 grade. There will be **two midterm exams**, each worth 100 points. The **final exam** will be worth 200 points. Each exam, including the final, will focus on what was covered since the previous exam. However, each exam will also be cumulative in that each section of the course builds on what came before. The final will be effectively two exams. The first section will act as midterm 3 and cover the material from the final third of the semester. The second section will be cumulative in that it will cover the entire semester, focusing on material that needs to be reviewed (*i.e.,* most of the class got it wrong the first time around).

Lecture assignments:
Throughout the semester, we will read and evaluate several **research papers** taken from the primary literature. The first three will be discussed in class, with the initial paper being an unassessed introduction to the primary literature. The final three will be assessed as part of your course grade. Assignment specifics will be provided in class and on the D2L webpage.

In addition, several **video-based homeworks** will be assigned. The purpose of the assignments is to reinforce course concepts and highlight the diversity of the science and scientists in clinical microbiology. Finally, one **data-based homework assignment** will be given. This assignment will focus on working with data from the lab or taken from the literature.

Lab constitutes approximately 40% of the BIOL 2144 grade. Specifics for lab exercises, assignments and grading will be provided during the first laboratory session.

Note:

1) No regrades will be provided for exams done in pencil.
2) Misspelled words (esp. organism names) and incorrect taxonomic nomenclature will result in ¼ point deductions for each instance.

Assignment Summary:

Midterm exams:	200 points (2 x 100 points)
Research Paper Assignments:	90 points (3 x 30 points)
Video Homework Assignments:	80 points (4 x 20 points)
Data-based Homework Assignment:	30 points

Final Exam:	200 points
Laboratory:	400 points
Total:	1000 points

Important Dates (Fall 2022):

Classes begin:	August 22
Labor Day (no classes):	September 6
Midterm Exam 1:	**September 27**
Last day to drop with a "W":	October 24 (4:00 pm)
Midterm Exam 2:	**October 25**
Thanksgiving Break (no classes):	November 23 – 27
Classes end:	December 2
Final Exam*:	**December 6 (Tuesday) (1:00 – 3:00 pm)**

Tentative Lecture Schedule

Date	Week	Topic	Reading	Assignments
Aug. 23 & 25	1	Intro; Bacteria; Microbiota	Ch1, Ch5	
Aug. 30 & Sept. 1	2	Microbiota; Bacteria & Disease	Ch5, Ch6	
Sept. 6 & 8	3	Methods: Measuring Infectivity	Ch8	Paper (Intro); **Homework 1 Due (R)**
Sept. 13 & 15	4	Methods: Bacterial Factors; Host Factors	Ch9, Ch10	
Sept. 20 & 22	5	Methods: Host Factors	Ch10	**Paper 1**
Sept. 27 & 29	6	**[Exam 1]** (T); Gene Exchange	Ch7	**Homework 2 Due (R)**
Oct. 4 & 6	7	Gene Exchange; Colonization	Ch7, Ch11	
Oct. 11 & 13	8	Toxins	Ch12	
Oct. 18 & 20	9	Delivery Systems	Ch13	**Paper 2**
Oct. 25 & 27	10	[Exam 2] (T); Virulence Regulation	Ch14	
Nov. 1 & 3	11	Virulence Regulation; Antimicrobials	Ch14, Ch15	**Homework 3 Due (R)**
Nov. 8 & 10	12	Antimicrobials; Antibiotic Resistance	Ch15, Ch16	
Nov. 15 & 17	13	Antibiotic Resistance; Research Forum	Ch16	**Research Forum Paper 3 Due**
Nov. 22	14	CRISPR, Restriction Enzymes		**Homework 4 Due (T)**
Nov. 24	14	**No Classes – Thanksgiving Break**		
Nov. 29 & Dec. 1	15	Examples: Gram-positives (T); Gram-negatives (R)	Ch18, Ch19	**Data Homework Due (R)**
Dec. 6	**Final Exam (Tuesday) 1:00 – 3:00 pm**			

Bacteriology Laboratory Information

Course Description (Laboratory):

This laboratory section complements the lecture portion of the course as a study of prokaryotic structure and physiology. Students will learn and apply practical laboratory techniques to the identification and characterization of bacteria.

NOTE: Bacteriology relies on studying living organisms. As a result, time is required for organisms to grow after inoculation into growth medium. Thus, students will be required at times to come into the lab outside of the scheduled laboratory period to collect data and interpret results.

Course objectives (Laboratory):

- Practice general laboratory safety
- Practice safe handling of microorganisms, including aseptic technique
- Recognize various cellular and colony morphologies of prokaryotes
- Correctly use and maintain a compound microscope
- Use differential and selective growth media to obtain pure cultures
- Correctly perform and interpret Gram Stains and other staining techniques
- Interpret and analyze results from various assays used to identify bacteria and apply these methods to other situations
- Enrich, isolate, and characterize prokaryotes based on the various metabolic strategies by which they adapt to their environment
- Communicate fundamental concepts of microbiology, both in written and in oral format

General Laboratory Policies:

- Laboratory safety rules **must** be always followed (see following section). Safety violations will result in deductions from the laboratory participation grade (see below).
- Cell phones and pagers are to be turned off before lab begins.
- Students are expected to read the introductory material prior to each lab session.
- Cheating will not be tolerated. Anyone suspected of cheating will be subject to the consequences outlined in the University's academic honesty policy.
- All lab sessions must be attended. We will not offer make-up labs.
- Punctuality is always expected. If you are more than 10 minutes late, you will miss the quiz for the day and be counted as absent.
- Attendance in the lab is required. The attendance policy regarding excused absences is provided above.
- All quizzes, exams, or assignments missed due to unexcused absences will be recorded as zeros.

- If you have a documented disability that will impact your work in this class, please contact the TA or instructor to discuss your needs.
- Each group is responsible for proper clean up at the end of the laboratory period. This includes proper cleaning and storage of microscopes, proper disposal of contaminated materials, disinfection of benches and workspaces, etc. (see Waste Disposal Guidelines & Clean-up below)
- **Three unexcused lab absences will result in failure (receiving 0 points) of the laboratory portion of the course.**

Attendance Policy:

General and specific policies regarding attendance in BIOL 3044 Laboratory sections are provided in the sections above.

Grading:

Quizzes:	100 points (10 of 11 quizzes)
Assignments:	80 points (4 x 20 points)
Participation/safety:	70 points
Research Forum evaluation:	30 points
Research project report:	80 points
Laboratory notebook:	40 points
Total:	400 points

Lab constitutes approximately 40% of the BIOL 3044 grade.

Quizzes will be given in the first 10 minutes of the laboratory period (see General Course Policies). Quizzes will cover the exercises from the previous week and material from the upcoming exercises. Thus, it is important both to understand your previous results and to have read the exercises for the week so that you are prepared.

There will be **4 assignments** that will evaluate **key laboratory skills**. Each assignment will be worth 20 points. The first will focus on correct use and care of the compound microscope. One will focus on using correct aseptic technique – being able to transfer cultures without contaminating the culture or the environment. One will focus on successful completion of a Gram stain. One will focus on the streak plate technique of isolating colonies.

An important skill for researchers is recording procedures, observations, and other pertinent information regarding laboratory work in a **notebook**. Everyone will record experimental details each week in a lab notebook. Notebooks will be evaluated periodically, with a final check the last week of class. Information on lab notebooks will be posted on the lab D2L page. Additional instructions, including an evaluation rubric, will be provided separately. Meeting the evaluation criteria (keeping a thorough and complete notebook) will count 40 points towards your laboratory/course grade.

Each semester, the University sponsors the **Undergraduate Research and Creative Activity Forum**. In this Forum, students present their research findings or creative works. For this assignment, students will **critically evaluate** three (3) poster presentations or three (3) oral presentations (or combination thereof). This will provide experience in both how to present results and how to critically evaluate data present by others in preparation for the Case Study reports at the end of the semester. Satisfactory completion of this assignment will be awarded 30 points towards your laboratory/course grade.

The **laboratory participation/safety** grade is based on adherence to laboratory safety and attendance policies. Everyone starts the semester with 70 points. During the first two lab periods, students will be given gentle reminders regarding lab safety and attendance as needed. After that, points will be deducted for each violation. The severity of the deduction is at the discretion of the instructor. The exception to this regards appropriate dress. As stated above, students wearing open-toed shoes or short pants/skirts will not be permitted to enter the laboratory.

The major project for this semester will be isolation and identification of one-two bacterial isolates from the soil by culture-based and DNA sequence methods. The target phenotype will be production of antimicrobial agents. Isolates will be characterized using metabolic assays. In addition, genomic DNA will be extracted from each of the isolates and the 16S rDNA will be amplified by PCR and submitted for DNA sequencing. The data collected during enrichment, isolation and identification will form the basis of a **final research report** (80 points). A more detailed description of the project will be handed out later in the semester.

All data, results, observations, and other experimental notes will be recorded in a **laboratory notebook**. The notebook will also be used to record observations and results from all laboratory exercises throughout the semester. Guidelines for proper laboratory notebook keeping will be provided in the lab and on the lab D2L webpage. All notebooks will be evaluated throughout the semester, with the final assessment at the end of the semester.

Tentative Schedule of Laboratory Exercises

Week	Date	Topic
1	Aug. 22	**No Lab Meeting (Online Lab Safety Videos)**
2	Aug. 29	Intro/Check-in, Lab safety, Soil collection paperwork, Review: Aseptic technique, Microscope use
3	Sept. 5	**No Lab Meeting – Labor Day**
4	Sept. 12	Soil preparation, dilution, spread plates* & [pour plates]
5	Sept. 19	Select candidate colony, subculture (streak plate), Gram stain, prepare pour plates (if not previously)
6	Sept. 26	2nd subculture, colony morphology, Gram stain, continue to monitor growth during the week
7	Oct. 3	Set up cultures for frozen stocks, cultures for BIOLOG, cultures for antibiotic production assay
8	Oct. 10	Prepare frozen stocks, set up BIOLOG plates (read at 24 and 48 hours), Set up culture for DNA extraction, Set up antibiotic production assay
9	Oct. 17	[Read results], Colony PCR, biochemical tests
10	Oct. 24	Electrophoresis of PCR products, send for sequencing, confirmatory tests (if needed), Transformation (Bio-Rad kit)
11	Oct. 31	[Read results], DNA extraction (if colony PCR fails)
12	Nov. 7	Work on report, Conjugation
13	Nov. 14	[Read results], report draft, CRISPR (Bio-Rad kit)
14	Nov. 21	[Reports due], CRISPR day 2
15	Nov. 28	Checkout, notebook final check, CRISPR: read results, discussion

***** Note on experimental cultures: Growth or other results should be read after 24 – 48 hours (although some slow growers may require 72 hours). All members of the group should have the opportunity to observe the results. If this becomes problematic, after the required growth period the cultures can be transferred to the refrigerator (4°C) until the following Monday. Keep in mind, however, that even with the lower temperature, the organisms will continue to metabolize, and your results may change.

Laboratory Safety:

Microbes are found everywhere within the environment – in the air, on surfaces, on your body. In the Bacteriology Laboratory, we deal with microbes at higher concentrations than those found in the environment. We will also be dealing with organisms that are potentially pathogenic to humans – thus we treat EVERY organism as if it were pathogenic.

1. No food or drinks are to be taken into or consumed in the laboratory. Further, ANY activity that involves hand-to-face contact (applying cosmetics, handling contact lenses, etc.) should be avoided.
2. Disinfect the work area before starting lab, after completing lab, and after any spills that occur. Do not assume that the lab members in the previous class cleaned up after themselves.
3. Wash your hands thoroughly with soap and water before leaving the laboratory – even if you need to leave only for a brief time.
4. Open-toed shoes, sandals or similar footwear are not appropriate and should not be worn in the laboratory. Shorts and short skirts are also inappropriate in terms of laboratory safety. These regulations are for your personal safety. Students wearing inappropriate dress will not be permitted to enter the laboratory.
5. Long hair must be tied back as it is not only a potential source of contamination, but also a fire hazard.
6. Proper personal protective equipment (PPE) must be used in the lab whenever work is being done. For this laboratory, PPE includes a lab coat, safety glasses and laboratory gloves.
7. Be aware of the location of safety equipment such as fire extinguishers, eyewashes, showers, First Aid kits, etc.
8. Follow all waste disposal guidelines. (See below)
9. Refer to the Introduction section of your lab manual for additional discussion of laboratory safety issues.
10. When in doubt, ASK!

Waste Disposal and Cleanup:

Proper cleanup of the laboratory is essential to reduce contamination and to ensure that subsequent lab sections have a clean and organized work area. The following guidelines must be observed during each lab session. Each student must take an active role in proper cleanup and waste disposal. Do not leave it for someone else.

Lab benches: There are wash bottles of disinfectant (Cidecon) located on the lab benches. You must clean the lab bench before AND after each lab. If you run out of disinfectant, refill the bottle from the large carboy next to the microscope cabinet. The best technique for disinfection is to stream disinfectant over the surface, then use a paper towel to even the fluid over the surface so that a light film remains. Do not wipe to dryness but allow the disinfectant to air dry. The extended contact of the fluid to the surface increases effectiveness.

Paper towels and **soap** are located next to the sink.

Waste material: Bacterial cultures must be killed prior to disposal. Each group must dispose of their cultures once they have obtained and recorded their results. Dispose of each type of waste according to the following guidelines.

- **Liquid cultures:** add bleach to the tube. Squirt bottles containing bleach (1:2 dilution of household bleach in water, 2.5% final) are kept next to the sink. Add 1/5 the culture volume (usually about 1 cm) and place the tube into the holding racks next to the sink. The tube cap should go into the appropriately marked basket. **NEVER** pour your cultures down the drain or into the trash.
- **Solid cultures (plates):** All cultures on plastic Petri plates are disposed of in the Contaminated Material Container (CMC, large box with the red plastic bag) next to the sink.
- **Solid cultures (slants):** For cultures on agar slants – remove the cap and place the cap in the appropriate basket. The tube is disposed of in the CMC. DO NOT bleach your slants.
- **Semi-solid cultures:** Some growth media are termed semi-solid because they do not contain enough agar to completely solidify. These media are treated like agar slants.
- **Additional note on CMCs:** Only materials that are visibly contaminated are to be put into CMCs. Paper towels used for washing hands go into the trash, as do transfer pipette wrappers, sterile swab wrappers and the like. If the organisms are dead/killed – as with paper towels used to wipe up Cidecon from the benches – the material still goes into the trash.