Microbial Planet Lab (MPL)

General Information

- Course: GEOL-3181 Microbial Planet Lab (MPL)
- Instructor: Sebastian Kopf (he/him, sebastian.kopf@colorado.edu)
- Teaching assistant: TBD
- Class times & location: Tuesday/Thursday 1:00-3:50 pm in Benson 355
- Office hours: on slack and by appointment

What is this course about?

This class is a course-based undergraduate research experience (CURE) class that teaches how to conduct original scientific research from hypothesis to experimentation, evaluation and reporting. In this class we study non-pathogenic microorganisms that you yourself isolate from soil. Hands-on research topics covered in class include how to isolate & culture new microbes; how to observe, describe and classify them; and how to examine their metabolic capabilities such as the production of lipids and antibiotics. The overall learning goals for this course are:

- 1. Discover the diversity and physiological breadth of microbial life
- 2. Understand the extent of the antibiotic resistance crisis.
- 3. Learn how science is conducted and how to engage in the scientific process.
- 4. Master key biological concepts in the context of an original research project.
- 5. Analyze, present and communicate original research results.
- 6. Collaborate with each other and the instructor/TA.
- 7. Contribute to the cultivation and characterization of new microorganisms.

Laboratory Skills

The main focus of the first half of the semester is learning the following laboratory skills while working with real life samples and new environmental isolates together as a class. Lab safety will be the highest priority throughout the semester. See **Safety** section below for details.

- working safely with microorganisms
- disposing safely of laboratory waste
- keeping a detailed lab notebook
- using variable volume pipettors
- making stock solutions for media components
- making liquid and solid media both rich media and defined modular media
- streaking microbes on solid media plates
- dilution plating environmental samples

Microbial Planet Lab

- quantifying colonies by colony forming units (CFU counts)
- describing colony morphology and diversity
- pick & patch plating isolated colonies
- amplifying the 16S ribosomal RNA gene by PCR
- visualizing amplified DNA by gel electrophoresis
- purifying, quantifying, and sequencing (Sanger) amplified DNA
- evaluating 16S data and using BLAST to identify close relative of isolates
- evaluating isolate safety & preparing freezer stocks of BSL1 strains
- growing BSL1 microbes in liquid culture and evaluating growth curves
- light microscopy and gram staining of isolates
- lipid extraction and fatty acid quantification by gas chromatography
- testing antibiotic production of isolates against BSL1 relatives of common pathogens

Semester Project

The main focus of the second half of the semester are individual student-designed and executed research project for which each student will formulate hypotheses, predictions, and experimental design as part of a proposal to isolate and characterize bacteria that can make use of a specific compound of interest as C, N, S, and/or P source. Each student may pick any non-gaseous molecule they are curious about except for controlled substances, anything acutely toxic, explosive or carcinogenic.

These projects will be student-driven and discovery oriented, and aim to teach and reinforce scientific practices & ethics, overcoming obstacles and the importance of iteration in scientific research, as well as collaboration with peers and the instructor. The instructor will provide feedback and guidance for each step along the way.

Schedule

The following is a tentative week-by-week schedule for this class. This schedule is likely to change as we will adapt weekly activities to fit with the growth stages of our isolates and cultures.

Week	Topics
1	Introduction, lab safety, making LB plates, streaking plates
2	Scientific method & ethics, building blocks of life, plating soil, pipetting practice
3	Modular medium, making defined media plates, dilution plating of soil samples, pick & patch plating of colonies
4	Colony morphology & 16S colony PCR of LB plate colonies, colony quantification (CFU counts) of dilution plated samples
5	Gel, electrophoresis, PCR purification + quantification + sanger sequencing; colony morphology & pick/patch plating of defined media plate colonies
6	16S data evaluation, BLAST, streak plates and liquid culture of BSL1 strains

7	Preparation of freezer stocks, PCR of defined media plate colonies
8	Lipid extraction and derivatization; continuation of PCR sequencing and 16S evaluation (BSL1 strains into liquid culture)
9	Light microscopy and gram staining
10	Students' individual isolation projects (enrichment plates, sample collection, dilution plating)
11	Evaluation of growth curves and lipid data; CFU counts, colony morphology, pick & patch plates for students' project plates
12	Testing antibiotic production against ESKAPE pathogen relatives
13	Student experiment with their individual isolates (BSL 1 strains only!)
14	Student experiment with their individual isolates (BSL 1 strains only!)
15	Wrapping up experiments, cleanup, final presentations

Education Research

As a course requirement, you will complete 3 course evaluation surveys (pre-survey, mid-point survey, and post-survey). We appreciate your candid responses to the surveys to help us understand how we might improve the course. Please keep in mind that we are evaluating the course and not you as an individual student. All survey responses will be collected by the course evaluator, Christine Okochi, Associate Scientist and education evaluator at the University of Colorado Boulder. Your course instructor will only know if you completed the surveys (completing all three is worth 5% of the course grade), but your name will not be connected to your survey responses. Your responses will be de-identified and only reported to the instructor in aggregate.

You are also invited to have your responses included in an educational research study on how the lab component of the course might impact students. Participating in the study means you give permission for your de-identified pre- and post-survey responses to be included in the study and possibly shared beyond this class. Participation in the study is voluntary and your instructor will not know if you chose to participate. Your responses are valuable for understanding the impact of courses like this one. If you have any questions about the course evaluation or educational research study, please contact Christine at christine.okochi@colorado.edu.

Safety

The lab exercises in this course involve the use of living organisms. Although the microorganisms typically found in soil are not considered to be highly virulent (risk group 1) and we will exclude fungi using a fungicidal compound in all isolation plates (cycloheximide), even benign soil bacteria can evolve pathogenicity and some of the species we will encounter in this class are considered opportunistic pathogens (risk group 2). This means that even though they may occur in or on our bodies and are members of our normal microflora, given the right

Microbial Planet Lab

circumstances (e.g., a compromised immune system or a point of entry into the body such as cuts or scrapes), these microorganisms may abnormally spread, multiply, and cause disease. It is thus imperative to treat all soil microorganisms as potential pathogens and follow lab safety protocols. Different organisms are categorized into different risk groups (RG1 – 4) and individual labs are cleared to work at the corresponding biosafety levels (BSL1 – 4). Generally, we will work exclusively with organisms in RG1 (those *NOT associated with disease in healthy adult humans or animals*) but pathogenic organisms that exist in soil might grow on some of our isolation plates, and, therefore, might be encountered in this class. We will stop work with any strain identified or suggested to be above RG1 by 16S Ribosomal RNA gene DNA sequencing. However, if you or someone you live with has a compromised or suppressed immune system (this includes those who are pregnant), please speak to me about any necessary additional precautions.

Resources

Canvas: all course materials, including readings, protocols, assignments, and supplemental materials will be managed from here. Please check in regularly to keep on top of course prep and upcoming deadlines.

Slack: the course has a slack channel for communicating directly with the instructor, asking questions about the class, syllabus, assignments, etc. Installing the <u>slack desktop app</u> or phone app is highly recommended, especially if you are already using other slack spaces and want to keep it all organized in one place. Please contact me ASAP if you're having trouble accessing the course slack.

Benchling: one of the most important skills to learn in scientific research is keeping a notebook. Because the world is going digital and you are likely to find that wherever you go you will be keeping your notebook electronically, we will be doing the same thing. We will be using the electronic lab notebook Benchling and set it up together in class during the first experiment. Building on the protocols you start with for each experiment, you will keep a detailed log of what you do and discover in the lab. You will be expected to keep day to day notes about exactly how you performed your experiments, how they are progressing and what the results are. Your notebook contains not only the details about what you did and what happened but why you did it and what it means.

Textbooks: we will be using some of the Research Protocols and readings from the Research Guide to Microbial and Chemical Diversity booklets from the Small World Initiative. You can borrow copies of each from me for the semester or buy your own copies if you'd like to have them. The bundle is \$25 on Amazon (link). Other than this, there is no required textbook for this course. However, if you want to dig deeper, there are a few **optional** books that you may find useful:

• A **General Microbiology** book: you may find it worthwhile to have a textbook that can be used to dive deeper into concepts that you find interesting along the way. An inexpensive option is: Through the microscope: A look at all things small by Timothy Paustian, which is available <u>online</u> as an ebook for \$30. A more comprehensive background reference is

Brock Biology of Microorganisms (<u>a few hard copies available @ CU Boulder library</u>). Although the current edition is 15th ed.; older ones are still very useful and beautifully illustrated

- A General Chemistry book: depending on how comfortable you feel about your general chemistry, you may find it useful to have a chemistry textbook at the ready. The most important concepts from general chemistry for this class are the chemical elements, covalent vs. ionic bonds, solubility and molecular weight, concentration (molarity) and acid/base chemistry.
- A Visual Guide for Microbiology Lab Procedures: visual learners may find A Photographic <u>Atlas for the Microbiology Laboratory, 5th Edition</u> (4th edition works too), by Leboffe and Pierce very helpful in the lab for knowing what to expect for various experimental tests and procedures.
- A Student Handbook for Writing in Biology 6th Ed. by Karen Knisely

Mental Health: If you are feeling stressed, worried, or down during the semester, or if you notice signs of emotional distress in someone else, please feel free to stop by my office (Benson 322A) or consider reaching out for support. CU Boulder's Counseling and Psychological Services (CAPS) also provide walk-in services that are available at several on-campus locations including the C4C and the Wardenburg Health Center. If you or someone you know needs to talk to someone right now, call 303-492-2277 (CU) or the national hotline 1-800-273-TALK (8255) at any time (24/7) to talk to a trained counselor. These services are free and confidential. For more information: https://www.colorado.edu/counseling/

Evaluation

The overall course grade will be determined by the following components. Detailed instruction for each assignment will be posted on Canvas.

Participation (20%)

Active participation; lab safety; self evaluation of learning at the end of the semester; filling out the education research surveys (see details in the **Education Research** section above - inclusion of your responses in the education research study is completely up to you).

Research Projects (40%)

Lab notebook maintenance throughout the semester (see **Benchling** section in the **Resources**); research proposal & presentation + poster on research results + report on the student's individual semester project (see details in the **Semester Project** section).

Prep Quizzes (20%)

Chemistry refresher quizzes and regular short quizzes on the reading materials in preparation of a new topic or experiment.

Exams (20%)

Two take home midterms to examine the students' understanding of key concepts and techniques covered in this class. There is no final.

Late Work Policy

Students and teams are responsible for handing in work on time. Work that is turned in one day late (i.e. within 24 hours after the deadline) will be marked down by 25%. Work turned in up to a week late will be marked down by 50% but you must let me know when to expect your work or I will reserve the right to deduct points. If you are having trouble with an assignment, please talk with me ahead of time. No late work will be accepted more than a week after an assignment deadline or after the last day of classes.

Attendance

Attendance is mandatory. This is a hands-on research experience class that meets each week specifically to conduct experiments. You are expected to be present the entire class period. Everyone is expected to help set up for experiments and clean up afterwards. Research is a team effort and everyone brings valuable perspectives and different levels of prior experience into the classroom - if you finish your own experiments early, please use the time to work with others on theirs or help tidy up. Excused absences for health reasons (including covid, see detailed policies in the covid section below), religious holidays, and research-related travel are acceptable but you must inform me by email **as soon as possible** if you need an excused absence and I will work with you on arranging either remote attendance or a plan for how to approach the missed material. More than 4 <u>unexcused</u> absences are not acceptable given the nature of this class and will result in automatic failure of the course.

General Policies

Classroom Behavior

Both students and faculty are responsible for maintaining an appropriate learning environment in all instructional settings, whether in person, remote or online. Those who fail to adhere to such behavioral standards may be subject to discipline. Professional courtesy and sensitivity are especially important with respect to individuals and topics dealing with race, color, national origin, sex, pregnancy, age, disability, creed, religion, sexual orientation, gender identity, gender expression, veteran status, political affiliation or political philosophy. For more information, see the policies on classroom behavior and the Student Conduct & Conflict Resolution policies.

Requirements for COVID-19

As a matter of public health and safety, all members of the CU Boulder community and all visitors to campus must follow university, department and building requirements and all public

health orders in place to reduce the risk of spreading infectious disease. CU Boulder currently requires COVID-19 vaccination and boosters for all faculty, staff and students. Students, faculty and staff must upload proof of vaccination and boosters or file for an exemption based on medical, ethical or moral grounds through the MyCUHealth portal.

The CU Boulder campus is currently mask-optional. However, if public health conditions change and masks are again required in classrooms, students who fail to adhere to masking requirements will be asked to leave class, and students who do not leave class when asked or who refuse to comply with these requirements will be referred to Student Conduct and Conflict Resolution. For more information, see the policy on classroom behavior and the Student Code of Conduct. If you require accommodation because a disability prevents you from fulfilling these safety measures, please follow the steps in the "Accommodation for Disabilities" statement on this syllabus.

If you feel ill and think you might have COVID-19, if you have tested positive for COVID-19, or if you are unvaccinated or partially vaccinated and have been in close contact with someone who has COVID-19, you should stay home and follow the further guidance of the Public Health Office (contacttracing@colorado.edu). If you are fully vaccinated and have been in close contact with someone who has COVID-19, you do not need to stay home; rather, you should self-monitor for symptoms and follow the further guidance of the Public Health Office (contacttracing@colorado.edu). In order for me to excuse a medically-related absence from class due to illness or quarantine, you must alert me right away (you do not need to state the nature of your illness) and I will work with you on arranging either remote attendance (if you are well enough) or a plan for how to approach the missed material.

Accommodation for Disabilities

If you qualify for accommodations because of a disability, please submit your accommodation letter from Disability Services to your faculty member in a timely manner so that your needs can be addressed. Disability Services determines accommodations based on documented disabilities in the academic environment. Information on requesting accommodations is located on the <u>Disability Services website</u>. Contact Disability Services at 303-492-8671 or <u>dsinfo@colorado.edu</u> for further assistance. If you have a temporary medical condition, see <u>Temporary Medical Conditions</u> on the Disability Services website.

Preferred Student Names and Pronouns

CU Boulder recognizes that students' legal information doesn't always align with how they identify. I will gladly honor your request to address you by an alternate name or gender pronoun. Please advise me of this preference early in the semester by updating your preferred name and pronoun via the student portal (as those preferred names and pronouns are then listed on the class roster) or by letting me know in person.

Honor Code

All students enrolled in a University of Colorado Boulder course are responsible for knowing and adhering to the <u>Honor Code</u>. Violations of the Honor Code may include, but are not limited to: plagiarism, cheating, fabrication, lying, bribery, threat, unauthorized access to academic materials, clicker fraud, submitting the same or similar work in more than one course without permission from all course instructors involved, and aiding academic dishonesty. All incidents of academic misconduct will be reported to Student Conduct & Conflict Resolution (honor@colorado.edu); 303-492-5550). Students found responsible for violating the <u>Honor Code</u> will be assigned resolution outcomes from the Student Conduct & Conflict Resolution as well as be subject to academic sanctions from the faculty member. Additional information regarding the Honor Code academic integrity policy can be found on the <u>Honor Code website</u>.

Sexual Misconduct, Discrimination, Harassment and/or Related Retaliation

CU Boulder is committed to fostering an inclusive and welcoming learning, working, and living environment. University policy prohibits sexual misconduct (harassment, exploitation, and assault), intimate partner violence (dating or domestic violence), stalking, protected-class discrimination and harassment, and related retaliation by or against members of our community on- and off-campus. These behaviors harm individuals and our community. The Office of Institutional Equity and Compliance (OIEC) addresses these policies, and individuals who believe they have been subjected to misconduct can contact OIEC at 303-492-2127 or email cureport@colorado.edu. Information about university policies, reporting options, and support resources can be found on the <u>OIEC website</u>.

Please know that faculty and graduate instructors have a responsibility to inform OIEC when they are made aware of any issues related to these policies regardless of when or where they occurred to ensure that individuals impacted receive information about their rights, support resources, and resolution options. To learn more about reporting and support options for a variety of concerns, visit <u>Don't Ignore It</u>.

Religious Holidays

Campus policy regarding religious observances requires that faculty make every effort to deal reasonably and fairly with all students who, because of religious obligations, have conflicts with scheduled exams, assignments or required attendance. In this class, you must inform me by email as early as possible in the semester (at least 1 week before any scheduled exams or assignments) about any religious observance conflicts. See the <u>campus policy regarding religious observances</u> for full details.