Spring 2-1-2019

BIOM 411.01: Experimental Microbial Genetics Lab

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BIOM 411
Experimental Microbial Genetics Laboratory
Course Syllabus
Spring 2019
Class Hours: 1:00 – 2:50 TR
Class Location: HS 405

Instructor: Bill Holben, Professor
Teaching Assistant: Sam Pannoni, PhD Candidate
TA Office: HS501
TA Office Hours: TR 12:00-1:00, or by appointment. Walk-ins also welcome unless busy or absent
TA E-mail Address: sam.pannoni@umontana.edu

A. DESCRIPTION
This laboratory course provides an overview of, and hands-on exploration of, several techniques employed in modern microbial genetics, recombinant DNA technology, and state-of-the art genomic and metagenomic analyses. Emphases are on purification of metagenomic DNA from environmental samples, PCR amplification of 16S genes, high-throughput DNA sequencing, phylogenetic classification and comparative bioinformatic analysis of microbial communities. The goal is to introduce students to the research potential of genetic, genomic and metagenomic analyses.

B: OUTCOMES
Through processing of environmental samples for next generation sequencing and bioinformatic analysis, students will learn the workflow, skills, and mindset needed to address questions in modern microbial genetics, biology, and ecology. Student outcomes from this class include:
1. Further develop student skills in hypothesis creation and scientific writing
2. Practice methods for processing environmental samples to obtain metagenomic DNA and create PCR amplicons
3. Conceptually understand and practically apply next generation sequencing (NGS) technology
4. Gain an appreciation for bioinformatic data processing and its value for research

C. ORGANIZATION
Students will have the option of working individually, or in groups. Whichever working format is selected, each student will be responsible for writing and submitting individual versions of the draft and final lab reports (described below) based on shared data as appropriate. Each class will begin with a 10-15 minute overview from the Instructor and/or TA regarding the day’s activities, where the experimental design, underlying mechanistic basis of techniques and main goals will be discussed. The remaining class time will be spent on execution of the laboratory experiments. The experiments can be complicated and the time is short, so each student is responsible for being FULLY PREPARED AT THE BEGINNING of each class and to maintain a DETAILED LABORATORY NOTEBOOK based on instructions giving at the beginning of the course and below in section E.
D. COURSE TOPICS
1. Metagenomic DNA purification from environmental samples
2. Polymerase Chain Reaction (PCR) to add barcoded primers for sequencing
3. Gel electrophoresis, DNA quantification and amplicon purification
4. Next Generation Sequencing (ultra-deep 16S gene sequencing using MiSeq)
5. Phylotype Classification and Bioinformatic Analysis

E. REQUIRED SUPPLIES
1. Laboratory Notebook (must have a fixed binding, no loose-leaf binders allowed)
2. Ultra-Fine Sharpie (one or two)
3. Lab Coat (if desired)
4. All other supplies and equipment will be provided

F. GRADING AND COURSE REQUIREMENTS
Each student will be required to maintain their own laboratory notebook and carefully record all details, calculations, observations, variations in protocol and all other details for each lab. Lab notebooks will be graded twice during the lab course. A 1-2 page Project Proposal, an interim lab report and a final lab report will be submitted during the semester to fulfill the requirements of the 2/3 writing component of this course. To enable these writing exercises, maintaining a well-organized laboratory notebook is not only required for its portion of the final grade and as good general laboratory practice, but will be essential for writing your lab reports. Note that scientific writing is an essential skill for any successful scientist. Thus the writing exercises for the course will cumulatively account for 60% of your final grade. Also note the key elements of successful modern scientific research include interaction, collaboration, and sharing of ideas. Thus, each student’s grade for this lab course will be broken down as follows:

- Lab Notebook 20% (10% at each inspection)
- Project Proposal 15%
- Interim Lab Report 15%
- Final Lab Report 30%
- Attendance/Participation 20%

*Late assignments – deduct one letter grade equivalent per day (at the discretion of the instructor/TA).

G. LAB NOTEBOOKS
A good lab notebook should have a description of the exact procedure, reagents used, amounts of reagents with proper units, and observations/results for each day’s work. Each new day’s entry should be indicated by the day’s date and a brief title for the activities (e.g. agarose gel of PCR1 products) Any competent scientist should be able to go into a student’s lab notebook and repeat the student’s experiment. Completeness is also much more important than being concise or than neatness, because anything written down could be helpful in future experiments. Erroneous entries should be left in the notebook, but crossed through with a single or double line.
H. PROJECT PROPOSAL FORMAT

Acceptable research proposals will be single-spaced in an easily readable 12-point font with each page numbered at the bottom. The proposal for research during the semester should be in the range of 3–4 pages.

A. Introduction/Overview – Background information regarding the system you have chosen to study and why it’s of interest/importance.

B. Hypotheses – Describe your proposed research in the form of testable hypotheses. You must have at least one hypothesis, but you are welcome to address more than one hypothesis with your experiment if it seems appropriate or feasible.

C. Experimental Design – Describe any experimental manipulations that you will make with your samples or study system in order to address the hypotheses and the rationale that underlies those manipulations.

D. Anticipated Outcomes – Describe what you anticipate your data will reveal and how it will be interpreted in the context of your hypotheses.

E. Optional Bibliography – You are not required to include citations in this short proposal, but if you do, refer to #6 in Section G below regarding how to cite them.

I. LABORATORY REPORT FORMAT

The purpose of the laboratory report is to communicate your scientific research (experimental work) in writing. This is a necessary skill, regardless of the field of science you are in. The goal is to help each student practice expressing their ideas and communicating their results in a scientifically professional manner. Thus, the format of the laboratory reports will be similar to that in peer-reviewed scientific literature or in applications for federal research funding. Briefly:

1. The interim and final lab reports must be submitted to the TA via e-mail as a .doc file via email by midnight on the due date.

2. The lab reports will have a title page that includes the course number and name, the student’s name, ID number, date, and a descriptive title for their work.

3. Acceptable lab reports will be single-spaced in an easily readable 12-point font with each page numbered at the bottom. The two interim lab reports should be in the range of 3–4 pages, while the final report should be 5–7 pages.

4. Reports must be in grammatically correct English without spelling errors (use your spell-checker!) as such errors will count against your grade. Please note that you will receive both the grade and constructive feedback on each of the interim lab reports that will provide valuable information and guidance for you in preparation for your final lab report.

5. Figures will be labeled as “Figure 1”, etc. and Tables as “Table 1”, etc. and should be embedded in the text in appropriate locations if possible. Each figure will have a legend that describes the figure in sufficient detail. All tables will have a title at the top of the table. Font size for legends and descriptions can be smaller but a minimum of 10-point font.

6. References cited will be indicated by sequential numbers e.g. (1), (2,3) in the body of the report, with a bibliography of references cited at the end of the report using ASM format (this will be provided on the Moodle site). Students are to rely primarily on peer-reviewed publications and
not on Internet sites, blogs, Wikipedia, etc. such that there are at minimum 2-3 citations from the primary literature for each one from the Internet.

7. The sections of the reports will be as follows:
   
   A. **Introduction** – Background information on the theory behind the experiment, system of study, questions/hypotheses that will be investigated, and general approach to answering them.
   
   B. **Materials and Methods** – Step by step (in prose) description of the experimental methods. We will discuss in class where it is appropriate to simply cite materials from handouts versus writing up your own materials and methods and protocols.
   
   C. **Results** – Report the results of the experiments with descriptive figures and tables and accompanying explanation. Do not analyze the results in terms of how they relate to the questions being asked in the study, just state the results.
   
   D. **Discussion** – Discuss the results with regard to how well they address the hypotheses or research questions that you were addressing. Draw conclusions, state strengths and limitations the techniques employed and how these qualify or strengthen your conclusions. Discuss how you would overcome such limitations in future experiments as well as the next logical steps to further your investigation.

J. ACADEMIC HONESTY

Appropriate ethical behavior in the classroom is required of every University of Montana student. Academic misconduct is subject to an academic penalty by the course instructor and/or a disciplinary sanction by the University. All written assignments in this class must be completely original.

Definition: **Academic Dishonesty** “cheating” and “plagiarism”, the theft of ideas and other forms of intellectual property – published or unpublished.

Definition: **Plagiarism** is the use of another writer’s words or ideas without acknowledging the source. Plagiarism also means “passing off a source’s information, ideas, or words as your own by omitting to cite them, which makes it an act of lying, cheating, and stealing.”

Definition: **Cheating** is defined as obtaining or attempting to obtain, or aiding another to obtain credit for work, or any improvement in evaluation of performance, by any dishonest/deceptive means. All students need to be familiar with the *Student Conduct Code*, which is available for review at [life.umt.edu/vpsa/student_conduct.php](http://life.umt.edu/vpsa/student_conduct.php)

**Accommodations**

We are happy to work with students and Disability Services for Students (DSS) to make accommodations that facilitate all students’ class participation and learning. Please see me on the first day of this class to plan for any such accommodations.
<table>
<thead>
<tr>
<th>DATE</th>
<th>DAY</th>
<th>TOPIC</th>
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<tbody>
<tr>
<td>Jan 10</td>
<td>Thur</td>
<td>Discussion of potential projects and proposal assignment + workflow from samples to sequences to data analysis</td>
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<tr>
<td>Jan 15</td>
<td>Tue</td>
<td>Discussion of samples/resources and potential hypotheses</td>
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<td>Jan 17</td>
<td>Thur</td>
<td>(NO LAB)</td>
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<td>Jan 22</td>
<td>Tue</td>
<td>Initial submission of proposals (NO LAB)</td>
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<tr>
<td>Jan 24</td>
<td>Thur</td>
<td>Proposals returned for editing, discuss projects further, begin DNA extractions</td>
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<tr>
<td>Jan 29</td>
<td>Tues</td>
<td>Extraction of DNA from environmental samples</td>
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<tr>
<td>Jan 31</td>
<td>Thurs</td>
<td>PCR to test 16S amplification ability/efficiency; <strong>Proposal 2nd Submission due</strong></td>
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<tr>
<td>Feb 5</td>
<td>Tues</td>
<td>Agarose gel electrophoresis of test-PCR amplicons; barcoded PCR to generate 16S amplicons for sequencing</td>
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<tr>
<td>Feb 7</td>
<td>Thurs</td>
<td>Magnetic bead purification of 16S/18S amplicons</td>
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<td>Feb 12</td>
<td>Tues</td>
<td>Quantify and plate DNA for sequencing; <strong>Submit notebooks for 1st grading</strong></td>
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<td>Feb 14</td>
<td>Thurs</td>
<td>Retrieve Lab Notebooks (NO LAB)</td>
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<td>Feb 19</td>
<td>Tues</td>
<td>Bioinformatics overview; download sequence data, prepare mapping files,</td>
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<td>Feb 21</td>
<td>Thurs</td>
<td>Continue bioinformatic analysis; <strong>Submit Interim Lab Report</strong></td>
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<td>Feb 26</td>
<td>Tues</td>
<td>Continue bioinformatic analysis</td>
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<tr>
<td>Feb 28</td>
<td>Thurs</td>
<td>Continue bioinformatic analysis</td>
</tr>
<tr>
<td>Mar 5</td>
<td>Tues</td>
<td>Continue bioinformatic analysis; Interim Lab Reports returned with comments and suggestions to be addressed in the final report</td>
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<tr>
<td>Mar 7</td>
<td>Thurs</td>
<td>Continue bioinformatic analysis</td>
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<td>Mar 12</td>
<td>Tues</td>
<td>Bioinformatic analysis continued as needed</td>
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<td>Mar 14</td>
<td>Thurs</td>
<td>Bioinformatic analysis continued as needed</td>
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<td>Mar 19</td>
<td>Tues</td>
<td>Bioinformatic analysis continued as needed</td>
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<tr>
<td>Mar 21</td>
<td>Thurs</td>
<td>Bioinformatic analysis continued as needed</td>
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<tr>
<td>Mar 26</td>
<td>Tues</td>
<td>Spring Break (NO LAB)</td>
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<tr>
<td>Mar 28</td>
<td>Thurs</td>
<td>Spring Break (NO LAB)</td>
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<td>Apr 2</td>
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<td>Apr 16</td>
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<td>Apr 18</td>
<td>Thur</td>
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<tr>
<td>Apr 23</td>
<td>Tue</td>
<td>In- Class Teaching evaluations; capstone discussion; <strong>Final Lab Report and notebooks due Thursday, April 25</strong></td>
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<td>Apr 25</td>
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