MICROBIAL GENETICS (BIO-375/575)
Fall 2016 (3 credits)

Place and Time: Art & Communications S118: 9:40 – 11:10 Tuesday & Thursday

Instructor: Dr. Toivo Kallas

Office: Halsey 245 (phone 424-7084; e-mail: kallas@uwosh.edu)
webpage: http://www.uwosh.edu/faculty_staff/kallas

Office hours: 1:50 – 3:50 Monday, 3:00 – 5:00 Tuesday, -- other times by appointment. Anytime by phone or e-mail. If I am not in, please leave a message or check the lab rooms (HS 238, 240, or 163/145 Labs)

Textbooks and Resources

Required

1. Snyder, L., Peters, L., Henkin, T.M. and Champness, W. 2013. *Molecular Genetics of Bacteria*, 4th edition, American Society for Microbiology, Washington, D. C.
2. Much of the reading material for the course will come from journals such as *Nature, Science, Proceedings National Academy Sciences, J. Bacteriology* and others. These and other reading materials will be posted on the class D2L site. Required readings will be indicated.

Recommended

McMillan, Virginia E. 2012. *Writing Papers in the Biological Sciences,* 5th edition, Bedford/St. Martin's.

Some Classical Bacterial Genetics References

1. Brock, T.D. 1990. *The Emergence of Bacterial Genetics,* Cold Spring Harbor Lab Press.
2. Ptashne, M. 2002. *Genes and Signals*, Cold Spring Harbor Laboratory Press.
3. Miller, J.R. 1992. *A Short Course in Bacterial Genetics: Lab Manual*, Cold Spring Harbor Laboratory Press.

Desire2Learn (D2L) Site

Powerpoint presentations, copies of literature discussion and reference articles, and other materials will be available via the class D2L site (**Microbial Genetics Bio-375/575**).

Some Microbial and Genetics Resources, Websites

1. **Class D2L site**, described above.
2. **American Society for Microbiology (ASM)** home page: http://www.asmusa.org.
3. **DOE Joint Genomics Institute (JGI)**: http://www.jgi.doe.gov/JGI_microbial/html/index.html (Microbial genome databases and a great resource for genome analysis including BLAST searches.)
4. **ExPASy Molecular Biology Server:** http://www.expasy.ch/. (A very useful site for molecular biology, genomics, and proteomics included predicted peptide mass fingerprints.)
5. **NATIONAL CENTER FOR BIOTECHNOLOGY INFORMATION (NCBI):** http://www.ncbi.nlm.nih.gov/. (This site includes the GenBank and other DNA, protein, and genomic databases and extremely useful search programs such as "BLAST." Includes the PubMed, MEDLINE literature database.)
6. Within **NCBI**, note for example **PubMed** (http://www.ncbi.nlm.nih.gov/pubmed/) for literature database searches and **PubChem** (http://pubchem.ncbi.nlm.nih.gov/) for structures and information about small molecules including metabolites, antibiotics, and inhibitors.
7. **TIGR** (The Institute for Genomic Research): http://www.tigr.org.
8. **Kazusa** Genome Research Institute: http://www.kazusa.or.jp.
9. ***E. coli* Genetics Stock Center**: http://cgsc.biology.yale.edu/. (a nice site for gene names, maps, etc.)
10. ***E. coli* Genome Center**: http://www.genome.wisc.edu

11. **The RCSB Protein DATA Bank**: http://www.rcsb.org/pdb/. (Site from which to download ".pdb" files of coordinates for viewing and manipulating protein and DNA sequence 3D structures).
12. **PyMOL:** http://pymol.sourceforge.net/ (Site for downloading the PyMOL program for very nice viewing and manipulation of protein and molecular 3D structures on Mac and Windows platforms.)
13. **Webcutter** (a site for on-line restriction site analysis): http://www.firstmarket.com/cutter/cut2.html
14. **Net Primer** (a site that allows downloaded or on-line design of PCR primers. They also carry "Plasmid Premier" a program for plasmid design): http://www.premierbiosoft.com/netprimer.html
15. **BioBIKE** (Biological Integrated Knowledge Environment): http://ramsites.net/~biobike/ (Provides integrated databases and access to a 'non-expert' programming language for bioinformatics investigation of biological databases).
16. **CyanoBIKE** (Cyanobacterial Biological Integrated Knowledge Environment): http://cyanobike-community.csbc.vcu.edu/ (graphical interface programming environment for access to integrated cyanobacterial genome databases, manipulation and data mining).
17. **KEGG** (Kyoto Encylopedia of Genes and Genomes): http://www.genome.jp/kegg/ (A very useful bioinformatics resource for linking genomes to biological systems and environments.)
18. **New England Biolabs**, Restriction Enzyme Database (NEB-REB): http://rebase.neb.com.
19. **Promega** Corporation (Madison, WI): http://www.promega.com/
20. **UWO (Polk) Library:** http://www.uwosh.edu/library/ (Polk Library provides access to a variety of useful literature databases such as NCBI PubMed and carries on-line, full-text subscriptions to several relevant journals including *Science, the Nature Journals,* Elsevier Journals via Science Direct, and the *American Chemical Society (ACS) Journals*. Follow on-screen instructions or see me.)

Course Overview and Objectives

Understanding microbial genes, genomes, and gene expression is essential for understanding the biology and evolution of microorganisms and their interactions with the environment. Since the discovery of genes in microorganisms, the structure of DNA, and DNA as the molecule of heredity (1940's and 50's), microbes have been used extensively to explore the structure, function, regulation, and evolution of genes and genomes. Microbial genetics is also important for understanding molecular techniques used to modify genes and proteins, manipulate bacteria, archaea, and eukaryotic organisms for fundamental research as well as practical applications in diverse areas of medicine and biotechnology.

We are in an exciting era of 'genomics,' 'metagenomics,' and 'post-genomics.' Complete genetic blueprints (genome sequences) of organisms and environments are being determined at amazing rates and these hold enormous potential for expanding our understanding of living organisms. We will discuss the structure, function, expression, and evolution of microbial genes and methods for their study and manipulation. Topics include microbial genomes and their evolution; gene discovery, identification, and mapping; mutation; DNA repair; gene transfer among organisms; plasmids; transposable genetic elements; recombination; and gene regulation. We will also discuss concepts and strategies of molecular genetics including gene cloning, polymerase chain reaction (PCR) and quantitative PCR, hybridization techniques, global gene expression studies, 'proteomics,' 'metabolomics,' uses of gene expression, directed mutagenesis, gene fusions, 'reporters,' probes, and emerging technologies such as 'Next Generation' DNA sequencing. Throughout the semester, we will discuss research and review articles related to both fundamental concepts and emerging topics in microbial genetics.

Learning objectives – students will gain:

1) Understanding of fundamental concepts in microbial genetics.
2) Insight into genetic methods used to investigate interesting biological problems.
3) Insight into current, exciting topics in microbial genetics and related fields.
4) Experience in reading and evaluating scientific articles.
5) Understanding of how microbial genetics has advanced science and society.

Undergraduate Grading and Requirements

Assignment	Due date	Points
Lac and GFP genotype tests & predictions	September 15	20
Journal article reports	6 reports @ 10 points each. May submit 2 additional reports for extra credit.	60
Genome analysis, gene manipulation, gene expression exercises	2 assignments due: Sept. 29 and Nov. 3. (50 points each). An additional exercise may be done for extra credit.	100
Midterm exam 1	October 7-14	150
Midterm exam 2	November 11-18	150
Final exam	December 9–16 (due December 16)	150
Total points		630

*Parts of exams may be given independently in the form of separate assignments.

Journal Article Reports: To encourage exploration of current topics, students are required to read journal articles related to microbial genetics and write brief reports on these **(*no more than 1 page each*)**. Six reports are required with up to two additional for extra credit. Reports should describe the **objective** of the study, the **methods** used, and the main **conclusions** of the work. Further instructions will be given.

Literature Discussion/Analysis: Usually one or more papers per week (from *Nature*, *Science*, *Journal of Bacteriology*, *Molecular Microbiology,* or other sources) will be assigned for class discussion. ***Students are expected to read these papers ahead of class and should be prepared to summarize and discuss them in class.*** Students may not fully understand these papers prior to class but grades can be improved by participating actively in discussions and asking questions.

Undergraduate Grading Policy and Exams: 90-100% =A, 80-90% =B, 70-80% = C, 60-70% = D, less than 60%=F. Grades of A^+, A^-, B^+, B^-, C^+, C^-, D^+, and D^- will be used, at the discretion of the instructor, for borderline scores. For example, scores within 2% of a grade cutoff will be designated minus or plus grades (e.g. 90-92 = A^- and 88-89 = B^+). If the class scores on particular exams or assignments are uniformly low, grades may be adjusted accordingly. Exams will consist of definition, problem, and discussion questions. Exams will typically be 'open-book' and 'take-home.' Undergraduates will be graded separately if graduate students consistently score higher.

Late Work: Late work will receive no more than 90% of full credit unless arranged in advance.

Attendance Policy: Students are responsible for obtaining class materials, completing exercises, and meeting requirements. Because this is an advanced course with a small class size, regular attendance is expected to maintain class progress and discussion. Advance notification of absences is expected.

Academic integrity: We operate under the principle of "academic integrity" expected at this university. UW System guidelines state: "*Students are responsible for the honest completion and representation of their work, for the appropriate citation of sources and for respect of others' academic endeavors*." (s. UWS 14.01, Wis. Adm. Code). Cheating or obstruction of the efforts of others will not be tolerated in any form. Students caught cheating will receive an F grade on the exam or assignment and may be subject to further disciplinary action. ***Note in particular that this honor system applies during take-home exams and assignments. Please do not be tempted to represent the work of others as your own. This constitutes cheating (plagiarism) and will be treated as described above.***

Additional Information and Requirements for Graduate Students

Graduate students are expected to complete all assignments required of undergraduates as well as additional assignments as described below.

Graduate Student Grading and Requirements

Assignment	Due date	Points
Lac and GFP genotype tests & predictions	September 15	20
Journal article reports	6 reports @ 10 points each. May submit 2 additional reports for extra credit.	60
Genome analysis, gene manipulation, gene expression exercises	3 assignments due: Sept. 29, Nov 3, and Dec 8 (50 points each). An additional exercise may be done for extra credit.	150
Midterm exam 1	October 7-14	170
Midterm exam 2	November 11-18	170
Graduate student presentations	December 8	50
FINAL EXAM	December 9–16 (due December 16)	170
Total points		790

*Parts of exams may be given independently in the form of separate assignments.

Graduate Student Grading Policy and Exams: 92-100% =A, 90-92% =A-, 88-90% =B+, 82-90% =B, 80-82% =B-, 75-80% =C+. Grades of C or lower are considered failing for graduate students. Grades may be curved if necessary. Exams will consist of definition, problem, and discussion questions. Exams will typically be 'open-book' and 'take-home.' Graduate students will answer two or more additional questions on exams. These will represent an additional ~10 - 20% of exam material relative to undergraduates.

Oral Presentations. Graduate students will give a 20 minute presentation on a selected topic.

Graduate Student Objectives

1) Data Analysis and Critical Thinking. Special emphasis will be placed on the ability of graduate students to understand and interpret data and think analytically and critically about information that has been presented in class. To assess accomplishment of this objective, all **exams will contain 2 or 3 additional questions** for graduate students. These will be data analysis problems or questions that require critical assessment of new information related to topics covered in class.

2) Clear and Logical Expression in Writing. Graduate students will be expected to write more clearly, logically and correctly than undergraduates. To assess accomplishment of this objective, graduate students will be held to a higher standard on exams and written assignments. Answers to exam questions should be clearly written and show a more detailed understanding of the topic than those by undergraduates. Assignments, such as genome analysis assignments, should be logically organized, use vocabulary correctly and show a superior understanding of the topic. **An additional genome analysis assignment will be assigned.**

3) Clear and Logical Oral Presentation. Graduate students will be expected to clearly and logically present material orally. To assess accomplishment of this objective, graduate students will be expected to give an **oral presentation on a selected topic** in Microbial Genetics at the end of semester. This presentation should be well organized and logically presented. Graduate students are expected to show a good depth of understanding of the topic presented, field questions, and provide thoughtful answers.

4) Ability to Synthesize Information and Develop Creative Work. Graduate students will be expected to use information presented in class to synthesize and develop new and creative work or genetic

strategies to tackle important and interesting questions in biology. To assess accomplishment of this objective, graduate students will complete an **additional genome analysis assignment** and give an **oral presentation** at the end of the semester. In these assignments, they will be expected to synthesize existing information (e.g. from scientific journals) and explore and/or present new or emerging findings or methods that may be used to address interesting biological problems and expand understanding.

5) Leadership. Graduate students will be expected to take a leadership role in the class. They will be expected to **participate actively in class discussions**, ask questions and show a deeper understanding of the material, and **help undergraduates understand fundamental as well as challenging concepts and assignments.** To assess accomplishment of this objective, I will keep track of graduate student participation and work with undergraduates. Assessment of such leadership activities may elevate or lower the final grade of graduate students, particularly those at the borderline between grades.

Topics and Schedule – Undergraduate and Graduate Students

Week	Topic	Text chapters
1-3 Sept 8>	**Introduction & historical perspective** ***A central theme: How do we identify genes & their function?*** Genetic nomenclature – *How not to get confused by gene names!* Introduction to and review of DNA structure Introduction to genome sequences, genetic & genome databases, & genome analysis. ***The crucial role of bioinformatics!*** Polymerase chain reaction (PCR) & gene cloning via "5'-add-on" PCR. ***Genome assignment no. 1: Gene identification & cloning via PCR*** DNA & genome sequencing strategies. ***Revolutions in DNA sequencing: '454' pyrosequencing, Illumina, & other 'Next Generation' sequencing technologies*** Review of classical genetic concepts in microbial genetics: complementation, recombination, & mapping.	Introduction Ch 1 Box 1.2 Ch 2 Box 2.5 Ch 3
3-5 Sept 20>	Structure and replication of DNA. ***How do we know that DNA is the genetic material?*** The basis for molecular genetics: ***DNA duplexes, melting, reannealing, & the activity of enzymes that bind to DNA*** Review of molecular genetic techniques: restriction analysis, gel electrophoresis, DNA & RNA hybridizations, melting curves, cutting & joining DNA, & gene cloning & construction strategies. *Emerging techniques – 'Gibson assembly' for cloning & gene constructs?*	Ch 1
	(RNA, transcription, translation, protein folding, & membrane proteins. Review mostly on your own)	Ch 2
5-6 Oct 4>	'Post-genomic' analyses: ***global gene expression studies via microarrays*** Spotted vs. oligonucleotide synthesis and 'tiling' arrays Emerging technologies: ***Global gene expression studies via 'deep mRNA sequencing'*** ***Real-Time, quantitative PCR (qPCR) & reverse transcriptase quantitative PCR (RT-qPCR) for gene expression studies***	Parts of Ch13 Box 13.7
	MIDTERM EXAM 1 (take-home: Oct 7 – Oct 14)	

6-7 Oct 13>	***Mass spectrometry and 'proteomics'*** as a way to identify gene products and study gene function *(Possible introduction to 'metabolomics' as a way of assessing the impacts of gene function)*	Parts of Ch13 Box 13.7
7-8 Oct 20>	***Mutation, DNA repair, and evolution*** Mutation & DNA repair Mutagenesis ***Mechanisms of genome and microbial evolution***	Ch 3 Ch 11 Box 11.1, 11.2, 11.3
8-9 Oct 27>	***Extra-chromosomal and moveable elements:*** Plasmids: gene cloning and *in vitro* mutagenesis	Ch 4 Box 4.1, 4.2
10 Nov 8>	***Gene Transfer:*** Impact on microbial evolution & basis for classical mapping and mutation analysis, ***Conjugation and conjugative plasmids***	Ch 5 Box 5.1
	MIDTERM EXAM 2 (take-home: Nov 11 – Nov 18)	
11 Nov 15>	***Gene Transfer:*** ***Transformation:*** physiological and artificial ***Transduction and bacteriophages***	Ch 6 Ch 7, 8
	Thanksgiving Break! (no classes Tues, Nov 22 – Sun, Nov 27)	
12 Nov 29>	***Moveable genetic elements*** Transposons, 'illegitimate' recombination, & site-specific recombination Plasmids and transposons as tools Microbial introns, retrons, and inteins	Ch 9 Box 9.2 Box 2.6
Dec 6	***Homologous recombination***	Ch 10 Box 10.1,10.2
13-14 Dec 8> with related topics throughout semester	***Regulation of gene expression & responses to changing environments*** Operons, repressors, activators, & paradigms of gene regulation Global regulatory mechanisms Regulatory cascades, two component sensors, sensor-kinases & response regulators, enhancers & silencers ***Regulatory RNAs*** *Global gene expression studies, further discussion of microarrays, proteomics & new technologies?*	Ch 12 Box 12.1-12.3 Ch 13 Box 13.1-13.7 Ch 14
13-14 Dec 8> & throughout semester	Special topics: Genetic analysis of bacteria, strain construction, gene fusions & ***genetic reporters***. Synthetic genes & genomes, *in vitro* genetic manipulations, ***final discussions, & late-breaking news!***	
13 (Dec 8)	***Graduate student presentations on selected topics***	
14	**FINAL EXAM (take-home: Dec 9 – Dec 16)**	
	End of semester celebration, Fratello's outing! (Dec 16*)*	