

Mol Biol 3V03 Techniques in Molecular Genetics Winter 2020 Course Outline

Course Details

Instructor: Prof. Herb Schellhorn, LSB433, 905-525-9140 x 27316

Office hours: Tuesdays 10:00am-12:00pm (by Microsoft Teams)

Lab/TAs: TBA

Lectures: Thursdays, 9:30am-10:20am, Location To Be Determined

Labs: Scheduled as 2 x 3.0 h labs each week. Refer to your MOSAIC timetable for details on

dates & times

Course Description

Calendar course description

A laboratory course providing hands-on experience in experimental design and molecular biology and molecular genetic techniques.

One lecture, two labs (three hours each); one term

Prerequisite(s): <u>BIOLOGY 2EE3</u>; and credit or registration in <u>MOLBIOL 3003</u>; and registration in Level III or above of an Honours program in Biology or Molecular Biology and Genetics. If not already completed, <u>BIOSAFE 1BSO</u> (or HTHSCI 1BSO) and <u>WHMIS 1A00</u> must be done prior to the first lab.

Antirequisite(s): BIOLOGY 3VV3

Course Scope

The goal of this course is for students to gain experience in performing diverse molecular genetic techniques, and hone their critical thinking and research skills.

Materials and Fees

Lab coat: \$26 from the Campus Store, if you do not already have one.



Course Learning Objectives

By the end of this course students will:

Lectures

- Understand molecular biology objectives.
- Be able to develop a genetic model to explain experimental phenotypic results.
- Understand bacterial genetic nomenclature.
- Gain and appreciation for the history of Molecular Biology
- Understand how to apply reductionist principles to scientific problems (and understand their limitations.
- Use scientific reasoning to resolve experimental outcomes.
- Understand current techniques in Molecular Biology.
- Learn how Molecular Biology will impact future societal goals.

Laboratories

- Plan and execute experiments.
- Quantify protein, DNA and determine enzyme activity.
- Use mutagenesis to generate loss of function mutants to identify gene/proteins relationships.
- Maintain a laboratory notebook.
- Write a scientific report.

Course Schedule

The schedule (below) used in 2019—the 2020 schedule will be very similar. The final schedule will be posted one week before the first scheduled class.

Lectures

One lecture per week, where we will discuss the theory behind the up-coming labs, the techniques that will be used (along with alternative strategies that could be employed), and controls that will be important for experimental interpretation.

Two labs will be held twice per week in LSB 102 and LSB 103. Students will also be required to occasionally check on results outside of lab time.

Instructor Lectures (synchronous) will be given live with a designated time at the end of each lectures to answer questions arising from the material.

This is the weekly plan for the course. Because of the nature of lab work, it may change if circumstances dictate. Note the schedule is given for the 2019 offering. Dates will be modified for the 2021 offering.



	Lecture	Lab Period 1	Lab Period 2	Evaluations
Week 1			Introduction (1A) Plate streaking General: pipetting review and solution preparation	
Week 2	Gene deletion (general), PCR primer design and phenotypic/genotypic assessment	(1B) PCR of 'knockout cassette' for csgD deletion (1A) Evaluate plates from Thurs	(1B) PCR check and clean- up	Accurate pipetting (lab)
Week 3	Gene deletion – PCR targeting; anticipating & predicting results; mutation	(1B) Create mutant (introduce knockout cassette into <i>E. coli</i>)	(1B) Test mutation success using colony PCR	
Week 4	DNA: in silico investigation, and in- lab isolation	(1B) Evaluate mutation success (1B) Complement mutant	(1B) Test effect of complementation (1C) Isolate plasmids for reporter fusion construction	Streak for single colonies (lab)
Week 5	Cloning and ligations	(1B) Assess complementation success (1C) Amplify promoter for cloning (2A/B) Test <i>S. venezuelae</i> spore concentration	(1C) Clean up PCR, and digest product (plus plasmid) in preparation for cloning	Aseptic technique (lab)
Week 6	Transformations and choice of bacterial strain.	(1C) Ligate products to create a csgD-lacZ reporter fusion	(1C) Transform product into <i>E. coli</i>	Hand in 1 st lab report (Oct 9 th) Hand in lab notebooks (Oct 11 th)
Week 7		Semester break		
Week 8	Protein overexpression and purification; different gel options (Bradford assay for Protein quantitation)	(1C) Test success of fusion construction (1D) Transform fusion construct into <i>E. coli</i> mutants	(1D) Assess success of transformations (1D) Pour SDS-PAGE	



	Lecture	Lab Period 1	Lab Period 2	Evaluations
Week 9	EMSAs and mutagenesis	(1D) Compare phenotypes of reporter-containing strains (1D) Lyse cells and run extracts on SDS-PAG	(1D) Perform Bradford assay (with std. curve) (1D) Pour EMSA gel (PAGE)	
Week 10	CRISPR	(1D) Run EMSA (2A) Conjugate CRISPR construct into <i>S.</i> venezuelae	(2B) Plan mutagenesis, and conduct expt. (2A/2B) Practice streaking out S. venezuelae for single colonies	
Week 11	After the mutagenesis	(2A) Streak out CRISPR exconjugants for single colonies (2B) Analyze mutagenesis outcomes (2B) Set up screen for mutants of interest	(2A) Patch out a selection of candidate mutants (2B) Patch out mutants of interest	Hand in 2 nd lab report (Nov 14 th)
Week 12		(2A) Colony PCR on candidate mutants (2B) Test candidate genes by PCR	(2A) Assess CRISPR success (2B) Clean up PCR reactions and check on gel (will be sent for sequencing, and results will be emailed to you)	
Week 13				
Week 14				Hand in 3 rd lab report (Dec 6 th) Hand in lab notebooks (Dec 6 th)

Late Work, Missed Work, and Extensions

Late submissions will be penalized with a 10% per day deduction on the grade of the assignment. Students will be able to make up missed work following the guidelines under the Request for Relief for Missed Academic Work policy. Missed work cannot be made up without a valid MSAF or permission from the Faculty Office, as appropriate. Extensions may be given for medical or compassion reasons, or during extreme circumstances.



IMPORTANT NOTE

The instructor and University reserve the right to modify elements of the course during the term. The university may change the dates and deadlines for any or all courses in extreme circumstances. If any modification becomes necessary, reasonable notice and communication with the students will be given with explanation and the opportunity to comment on changes. It is the responsibility of the student to check their McMaster email and course websites weekly during the term and to note any changes. Changes will be communicated through regular McMaster communication channels, such as McMaster Daily News, Avenue to Learn and/or McMaster email.

Course Materials

Reference textbook:

There is no text for this course. Registered students will be provided with a suggested reading list one week before the start of the course.

Course Evaluation

The course grading will be based on the following weighting.

Evaluation

15% Lab notebook

35% Lab reports (3)

15% Online activities/completion of case studies:

20% Final exam

15% Lab performance (competence, pre-lab quizzes, peer evaluation)

Lab notebook (15%)

It is critical that you take notes in the lab **while you are doing the work**. These notes should be written into a laboratory notebook as you do the work. They should contain:

- a sentence on the objective of the experiment
- the date when each experiment was performed (this can often be one of the most useful pieces of information in a lab notebook)
- reagents added (and volumes of each) for enzyme reactions/assays performed
 (e.g. PCR, restriction enzyme digestions, ligations, EMSAs)
- other technical numerical information such as incubation times (start, end, duration), temperatures, PCR programs, antibiotic concentrations in the plates, agarose gel percentages, etc.
- any modifications of the procedure or ways in which you deviated from the manual
- expectation of experimental outcomes (e.g. anticipated PCR product size)
- the results you obtained (in the form of the data as you actually obtained them), with



everything very well labelled (e.g. what is shown in each lane of your gel picture?)

- a conclusion, stating what the experiment showed (and whether it matched what you predicted the outcome would be)

These sorts of notes are incredibly important when trying to troubleshoot experimental problems. Getting comfortable with this now will help you tremendously in future lab work (co-op terms, thesis positions, etc.). To encourage you to follow these recommendations, your lab notes will be signed at the end of each class by your TA, and will be evaluated twice during the term (once after the midterm break, and once at the end of term). Lab notebooks will be submitted at the end of the labs. A sample page has been provided for you in the 'Helpful Information' section on Avenue.

Lab reports (35%)

There will be three full lab reports. The relative percentage associated with each of these may be subject to change.

10% 1st report covering Experiment 1A,B 10% 2nd report covering Experiment 1C,D 15% 3rd report covering Experiment 2

You can find more detail about the lab reports, and what is expected to go into them (along with excellent examples) in the 'Helpful Information' section of Avenue. Lab reports will be submitted in person to your TAs at the beginning of the labs.

Quizzes/activities/case studies (15%)

Many of the lecture classes will be associated with online quizzes on the theory behind the week's labs. These are intended to provide you with an opportunity to practice different skills (both technical and critical-thinking types), and to give me feedback on

areas where some additional discussion might be useful. Case studies will also be provided on a relatively regular basis, to help you work through some of the more challenging concepts. Online quizzes will be submitted online. Prelab quizzes will be handed out and submitted within the first 10 min of each lab to the TAs. Case studies will be submitted to your TAs during lab time.

Exam (20%)

The Final Written Exam will deal with both theory and practice of the material covered in the lectures and laboratory exercises.

Lab performance (15%)

This mark will encompass a number of different factors/components, including (but not limited to):

- performance on competence exercises
- performance on self-directed aspects of the labs
- lab citizenship (e.g. cleaning up after yourself)
- pre-lab quizzes/activities
- peer evaluation on your contribution to lab work

This course may use online proctoring software for tests and exams. This software may require students to turn on their video camera, present identification, monitor and record their computer activities,



and/or lock/restrict their browser or other applications/software during tests or exams. This software may be required to be installed before the test/exam begins.

Academic Integrity

You are expected to exhibit honesty and use ethical behaviour in all aspects of the learning process. Academic credentials you earn are rooted in principles of honesty and academic integrity. It is your responsibility to understand what constitutes academic dishonesty.

Academic dishonesty is to knowingly act or fail to act in a way that results or could result in unearned academic credit or advantage. This behaviour can result in serious consequences, e.g. the grade of zero on an assignment, loss of credit with a notation on the transcript (notation reads: "Grade of F assigned for academic dishonesty"), and/or suspension or expulsion from the university. For information on the various types of academic dishonesty please refer to the <u>Academic Integrity Policy</u>, located at https://secretariat.mcmaster.ca/university-policies-procedures- guidelines/

The following illustrates only three forms of academic dishonesty:

- plagiarism, e.g. the submission of work that is not one's own or for which other credit has been obtained.
- improper collaboration in group work.
- copying or using unauthorized aids in tests and examinations.

Authenticity/Plagiarism Detection

Some courses may use a web-based service (Turnitin.com) to reveal authenticity and ownership of student submitted work. For courses using such software, students will be expected to submit their work electronically either directly to Turnitin.com or via an online learning platform (e.g. Avenue to Learn, etc.) using plagiarism detection (a service supported by Turnitin.com) so it can be checked for academic dishonesty.

Students who do not wish their work to be submitted through the plagiarism detection software must inform the Instructor before the assignment is due. No penalty will be assigned to a student who does not submit work to the plagiarism detection software. All submitted work is subject to normal verification that standards of academic integrity have been upheld (e.g., on-line search, other software, etc.). For more details about McMaster's use of Turnitin.com please go to www.mcmaster.ca/academicintegrity.

Courses with an On-line Element

Some courses may use on-line elements (e.g. e-mail, Avenue to Learn, LearnLink, web pages, capa, Moodle, Echo360, Microsoft Teams, ThinkingCap, etc.). Students should be aware that, when they access the electronic components of a course using these elements, private information such as first and last names, user names for the McMaster e-mail accounts, and program affiliation may become



apparent to all other students in the same course. The available information is dependent on the technology used. Continuation in a course that uses on-line elements will be deemed consent to this disclosure. If you have any questions or concerns about such disclosure please discuss this with the course instructor.

Online Proctoring

Some courses may use online proctoring software for tests and exams. This software may require students to turn on their video camera, present identification, monitor and record their computer activities, and/or lock/restrict their browser or other applications/software during tests or exams. This software may be required to be installed before the test/exam begins.

Conduct Expectations

As a McMaster student, you have the right to experience, and the responsibility to demonstrate, respectful and dignified interactions within all of our living, learning and working communities. These expectations are described in the <u>Code of Student Rights & Responsibilities</u> (the "Code"). All students share the responsibility of maintaining a positive environment for the academic and personal growth of all McMaster community members, **whether in person or online.**

It is essential that students be mindful of their interactions online, as the Code remains in effect in virtual learning environments. The Code applies to any interactions that adversely affect, disrupt, or interfere with reasonable participation in University activities. Student disruptions or behaviours that interfere with university functions on online platforms (e.g. use of Avenue 2 Learn, WebEx, Echo360, Microsoft Teams or Zoom for delivery), will be taken very seriously and will be investigated. Outcomes may include restriction or removal of the involved students' access to these platforms.

Academic Accommodation of Students with Disabilities

Students with disabilities who require academic accommodation must contact <u>Student Accessibility Services</u> (SAS) at 905-525-9140 ext. 28652 or <u>sas@mcmaster.ca</u> to make arrangements with a Program Coordinator. For further information, consult McMaster University's <u>Academic Accommodation of Students with Disabilities</u> policy.

Requests for Relief for Missed Academic Term Work

<u>McMaster Student Absence Form (MSAF):</u> In the event of an absence for medical or other reasons, students should review and follow the Academic Regulation in the Undergraduate Calendar "Requests for Relief for Missed Academic Term Work".

View the McMaster Student Absence Form (MSAF) for more information.



Academic Accommodation for Religious, Indigenous or Spiritual Observances (RISO)

Students requiring academic accommodation based on religious, indigenous or spiritual observances should follow the procedures set out in the <u>RISO</u> policy. Students should submit their request to their Faculty Office *normally within 10 working days* of the beginning of term in which they anticipate a need for accommodation <u>or</u> to the Registrar's Office prior to their examinations. Students should also contact their instructors as soon as possible to make alternative arrangements for classes, assignments, and tests.

Copyright and Recording

Students are advised that lectures, demonstrations, performances, and any other course material provided by an instructor include copyright protected works. The Copyright Act and copyright law protect every original literary, dramatic, musical and artistic work, **including lectures** by University instructors

The recording of lectures, tutorials, or other methods of instruction may occur during a course. Recording may be done by either the instructor for the purpose of authorized distribution, or by a student for the purpose of personal study. Students should be aware that their voice and/or image may be recorded by others during the class. Please speak with the instructor if this is a concern for you.

Extreme Circumstances

The University reserves the right to change the dates and deadlines for any or all courses in extreme circumstances (e.g., severe weather, labour disruptions, etc.). Changes will be communicated through regular McMaster communication channels, such as McMaster Daily News, Avenue to Learn and/or McMaster email.