



Welcome to Biochemistry 3LA3

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Contact information

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Class Time - Mondays 10:30 AM – 12:20 PM in MDCL 1010

Lab Times - Fridays 1:30-4:20 PM in HSC 1H1-8

Course description



Hello everyone and **welcome** to Biochemistry 3LA3 !!!

We designed this course to immerse you in several advanced biochemistry techniques that have huge biomedical implications in the research field. First you will embark on an amazing adventure in the world of antibodies and their applications: from purifying immunoglobulins to Western blotting, and ELISA techniques. Next, you'll discover the wonderful world of quantitative PCR and its applications to a plethora of research fields. Finally, we'll explore mammalian cell culture and CRISPR/Cas9 technology. The course itself is designed to provide an experiential learning opportunity into the world of research. As such, we have also implemented team-based learning and reflection skills as strong components threaded throughout the course. You will be assigned to a research team and, as a team, you will work together through the different laboratory techniques.

Here is an overview of the techniques that covered in the course:

Lab basics module – In this module you will revisit certain basic laboratory techniques such as pipetting, alkaline lysis minipreps, agarose gel electrophoresis, spectrophotometry. You will also be introduced to new basic laboratory techniques such as mammalian cell culture, passaging mammalian cells, and counting mammalian cells using a hemocytometer.

Antibodies module - In this module you will work with your team to isolate immunoglobulins (aka. Antibodies) from rabbit serum. You will be using the automated AKTA start FPLC (Fast Performance Liquid Chromatography) system with a Protein-A conjugated affinity chromatography column. Once purified, you will perform a number of downstream techniques including a Western Blot, enzyme-linked immunosorbent assay (ELISA), and bicinchoninic acid assay (BCA). The ELISA, in particular, is a powerful diagnostic technique as it can be used to determine if patients have antibodies to many infectious diseases.

CRISPR module – In this module you will use CRISPR/Cas9 technology to knock down expression of a specific gene. More specifically, you will be knocking down the expression of glucose-6-phosphate dehydrogenase (*G6pd*) using CRISPR/Cas9 technology. This module is divided into two parts:



Part 1 – transfection and cell line selection. You will use a technique called transfection to introduce a Cas9/guide-RNA plasmid into a specific mouse liver cell line. Following successful transfection of the Cas9/guide-RNA plasmid, you will select for cells that have successfully taken up the desired plasmid. You will then perform a dilution assay to isolate single cells. Each of these cells could have been successfully gene edited by the transfected Cas9/guide-RNA plasmid. The next task involves growing larger, clonal cell quantities to validate the success of the CRISPR/Cas9 gene editing technique.

Part 2 – you will design various techniques to validate which of the isolated clonal cells have succeeded in knocking down expression of glucose-6-phosphate dehydrogenase using CRISPR/Cas9 technology. You will use quantitative PCR to determine gene expression levels of the *G6pd* gene in the cells obtained from part 1 of this module.

We developed this course structure with the hopes of:

- Engaging you in advanced biochemistry techniques and their biomedical applications
- Providing an immersive lab environment conducive to collaboration, technical skill development, and knowledge translation
- Developing your technical communication skills

Materials & Fees

- **Appropriate** surgical masks must be worn by each student during the lab time.
- McMaster standard calculator, safety goggles (must be UV protective) and lab coat must be purchased by each student and brought to each lab. Lab consumables (timer, sharpies, etc.) will be provided for you in the lab.
- Lab notebook – each student must purchase a lab notebook for this course from the McMaster Campus store ([click here for more information](#)).

Safety training requirements

Available online through the Mosaic portal. Instructions on how to proceed with these requirements will be posted on the 3LA3 A2L course, "course information" folder (all safety training is due Jan 12 at 11:59 pm):

1. FHS Fire Safety (hospital locations, must be re-taken annually) - course code: FHSFSF



2. WHMIS training (Must be taken once every 5 years. If you already completed this within the last 5 years, you do not need to do it again) - course code: WHMS15
3. Biosafety core training is taken once (the BBS teaching labs are designated Risk Group 1) - course code: BSLTRA If you already took this course last year, you need to complete the biosafety update (BSUPD).

Please note: No food or drink is allowed into the lab (includes empty food containers, wrappers, bottles, gum). No cell phones, tablets, laptops allowed in the lab space.

General lab safety rules



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Safety: please ensure that when working in the lab you designate an area for your lab notebook and you do NOT work over this area so as to avoid spillage on your books. If you do spill on your books, please let your lab mentor know so they can help you clean up your books appropriately (depending on the spill).



<ul style="list-style-type: none">• Please be alert at all times while in the lab. Notify your mentor/Felicia/Cait/Vivian immediately if you observe any unsafe practices.	<ul style="list-style-type: none">• No food or drink in the lab. This means that you may NOT bring food or drink into the lab and you may NOT throw out empty food/drink containers in the lab garbage.
<ul style="list-style-type: none">• No laptops/cell phones/etc. are allowed during the lab.	<ul style="list-style-type: none">• You cannot work alone in the laboratory (a mentor/Felicia/Cait/Vivian must be present at all times).
<ul style="list-style-type: none">• You will have a storage area for your book bags and jackets that is not in the actual wet-lab space. You must leave your pencil case, hats, etc. in this area.• You may NOT eat or drink in this area. In addition, you cannot store any food or drink in this area. All water bottles/drinks and any food MUST be stored in your bag so that they are not visible to anyone standing in this area.	<ul style="list-style-type: none">• Please note, if you forget your lab coat or goggles you will be asked to purchase them from the bookstore prior to attending the labs. We do not provide you with lab coats or goggles.• Dangling jewelry is a hazard in the laboratory and should be secured.
<ul style="list-style-type: none">• You need to carry your lab coat in a separate plastic bag. Please do NOT wear your lab coat outside the lab space. The hallway is NOT an appropriate place for you to put on your lab coat.	<ul style="list-style-type: none">• You may not eat or drink anything from the lab. You may not take anything home from the lab (test tubes, gels, reagents, Petri dishes, pipette, etc.) or bring any outside experiments into the lab.
<ul style="list-style-type: none">• Please keep your hands away from your face, eyes, mouth, and body while using chemicals or lab equipment. Wash your hands with soap and water in the designated hand washing sink after performing all experiments and prior to leaving the lab.	<ul style="list-style-type: none">• Please make sure that you do not walk around the lab and distract other students during the lab period. Please focus on your own work and on conducting the lab in a safe manner.
<ul style="list-style-type: none">• Please make sure that you take notes during the lab safety walk through and you know the location of safety features in the lab. Please make sure that you know the proper procedures in case of emergencies. These will all be discussed during the labs and your safety training.	<ul style="list-style-type: none">• Never return unused chemicals to their original container.• All chemicals/biologicals in the laboratory are to be considered dangerous. Avoid handling chemicals without gloves. Always read the SDS prior to handling any chemicals/biologicals and follow the proper safe handling instructions. Do not taste or smell any chemicals/biologicals.
<ul style="list-style-type: none">• Please maintain good housekeeping practices. Work areas should be kept clean and tidy at all times. Any area in the lab left untidy will result in a mark of zero on the day's participation sheet for the students (individual, pairs or entire team). This includes communal lab areas, like weigh scales and fume hoods.	<ul style="list-style-type: none">• Report any accident (spill, breakage, etc.), injury (cut, burn, etc.) or broken equipment to your mentor immediately. <u>Do not panic</u>. If you or your lab partner is hurt, immediately (and loudly) yell out your mentor's name to get their attention. Do not panic.
<ul style="list-style-type: none">• Dispose of all chemical waste properly. Never mix chemicals in sink drains. Check with your mentor for disposal of chemicals and solutions. Never dump any chemicals down the hand washing sink.• Please do not touch any of the equipment without proper training and supervision by your mentor.	<ul style="list-style-type: none">• Perform only those experiments authorized by your mentor. Carefully follow all instructions, both written and oral. Unauthorized experiments are not allowed. If you do not understand a direction or part of a procedure, ask your lab mentor before proceeding with the activity.

Course Schedule

This schedule is based upon current university and public health guidelines and may be subject to changes during the term. Any changes to the schedule or course delivery will be communicated on the course announcements section on Avenue to Learn. Please check the announcements prior to attending class.

Wk	Monday class 10:30am-12:20pm MDCL 1010	Description	Friday lab 1:30-4:20pm HSC 1H1-8	Description
1	Jan 9	Welcome to 3LA3! Team flowcharts	Jan 13	Lab basics module Teams A and B – molecular cloning basics Teams C and D – tissue culture basics
2	Jan 16	Tissue culture basics (all teams) and passing cells for Team C and D	Jan 20	Lab basics module Teams A and B – tissue culture basics Teams C and D – molecular cloning basics
3	Jan 23	Antibodies module – overview and passing cells for Team A and B	Jan 27	Antibodies module – lab 1 (IgG purification, column chromatography)
4	Jan 30	Antibodies module - techniques	Feb 3	Antibodies module – lab 2 (SDS-PAGE, Coomassie stained gels, transfer, BCA assay)
5	Feb 6	Antibodies module - techniques	Feb 10	Antibodies module – lab 3 (develop western blot, ELISA)
6	Feb 13	Test-1 review	Feb 17	Test 1
	Feb 20	Recess	Feb 24	Recess
7	Feb 27	CRISPR module - overview	Mar 3	CRISPR module Teams A and B – lab 1 (transfections) Teams C and D – lab 1 (CRISPR workflow)
8	Mar 6	Teams A and B – please come to the BBS teaching labs to check your cells and start antibiotic selection Teams C and D – no lecture on this day	Mar 10	CRISPR module Teams A and B – lab 1 (CRISPR workflow) and check on transfected cells Teams C and D – lab 1 (transfections)
9	Mar 13	Teams A and B – no lecture on this day Teams C and D – please come to the BBS teaching labs to check your cells and start antibiotic selection.	Mar 17	CRISPR module Teams A-D – lab 2 (Single cell dilutions) Teams C and D – check on transfected cells
10	Mar 20	CRISPR validation techniques	Mar 24	CRISPR module Teams A-D– lab 3 (Cell lysis/cDNA/qPCR)
11	Mar 27	Test 2 review	Mar 31	Test 2
12	Apr 3	No class	Apr 7	Good Friday, no lab
13	Apr 10	No class		



Course Assessment

Assessment Type	Percent (%)
Lab preparedness and participation (this mark includes weekly lab notebooks, class attendance, lab preparedness)	10
Team Flowcharts (module flowchart (2.5%), CRISPR workflow (5%))	7.5
Tissue culture aseptic technique checklist	2.5
Test 1	30
Test 2	30
CRISPR module data analysis report	10
Antibodies module data analysis report	10

Assessment due dates

Assessment Type	Due date
Lab preparedness and participation (individual assessment)	throughout the course
Module Flowchart (2.5%) - team assessment	Mon Jan 9, end of class time
Tissue culture aseptic technique checklist (2.5%) – individual assessment	Mon Jan 23, 11:59 pm
Test 1 (30%) – individual assessment	Fri Feb 17
Antibodies module data analysis report (10%) – individual assessment	Mon Feb 27, 11:59 pm
CRISPR workflow (5%) – team assessment	Teams A and B – Fri Mar 3 Teams C and D – Fri Mar 10
Test 2 (30%) – individual assessment	Fri Mar 31
CRISPR module data analysis report (10%) – individual assessment	Mon Apr 10, 11:59 pm



Lab notebook requirements

A well-maintained laboratory notebook is vital to your success in this course. You are required to keep an up-to-date lab notebook as part of this course. This component will be checked on a regular basis by your lab mentor. This is part of your lab participation mark.

Your lab notebook serves two important purposes. In the first place, it helps ensure that you are prepared for the week's experiments and have familiarized yourself with the appropriate protocols. Second, your lab notebook helps keep track of your work and stands as a record of your activities in the lab. It will help you stay organized and assist you as you prepare your lab reports! The more details you include in your lab notebook-the better. Even small changes, modifications, or observations can make a BIG difference to the outcome of an experiment. The lab notebook will also help us, as instructors, follow your progress over the semester. The contents of the notebook should be brief and concise, yet descriptive. It should be written in enough detail that another person with no knowledge of your experiment could reconstruct your study and reproduce your results. Maintaining an effective notebook will also facilitate the future writing of a good quality lab report or scientific research paper, or act as a starting point for future experiments.

One lab entry should encompass the entire week and needs to include the following sections. Your mentor will check the lab notebook entry at the beginning of each lab.

Purpose: Concise but informative. You should include information on what your team is working on at the same time. You cannot work in isolation:

- ✓ Why are we doing this experiment?
- ✓ What came before that led us to this line of questioning?
- ✓ What might the expected results be?
- ✓ What will the next logical steps be?

Safety and Reagents: Always have the following "Wear lab coat, gloves, and goggles." Review all reagents you are using and list them in a concise table. If dangerous briefly describe why and proper handling procedures. Here's an example for acrylamide:

Acrylamide: Neurotoxin, carcinogen. Avoid skin exposure and inhalation. Use in fume hood, wear gloves, lab coat, goggles. See SDS (reference, APS citation style).



Nice and simple. Do not write an essay but make sure you understand the safety considerations of each lab (this includes the reagents your team members are working on, since you will be working in the same space).

Protocols/Procedures: Please write down all step-by-step protocols required for the day. Record all changes to the protocol.

Observations/data/results/discussion: Please write out all observations/results.

Each week the lab notebook will be checked by your lab mentor at the beginning of the lab. This is part of the lab preparedness and participation mark. The marking scheme is as follows:

Lab preparedness (/3)		
Please assign mark (/3) based on the following criteria:		
<ul style="list-style-type: none"> Has proper personal protective equipment (i.e. goggles, lab coat) and has brought the lab notebook, a pen/pencil, and a calculator to the lab. Has completed the lab notebook entry. If the lab notebook entry has not been attempted for the day's lab (or the lab notebook was not brought to the lab), please assign a mark of zero for the entire day's participation (0/7). 		
Assign a mark of 1/3 if the student: has not completed more than 50% of the criteria specified for this section.	Assign a mark of 2/3 if the student: has completed almost all of the criteria specified for this section.	Assign a mark of 3/3 if the student: has completed all the criteria specified above.
Lab work (/3)		
Please assign mark (/3) based on the following criteria:		
<ul style="list-style-type: none"> Using equipment properly and safely Following the protocol properly and listening to instructions (and keeping proper observations in the lab notebook) Actively participating in lab work Maintaining a clean work environment Understanding and following lab safety rules 		
Assign a mark of 1/3 if the student: does not follow more than 50% of the criteria stated above.	Assign a mark of 2/3 if the student: follows most, but not all, of the criteria stated above.	Assign a mark of 3/3 if the student: follows all of the criteria stated above.
Lab progress (/1)		
Please assign mark (/1) based on the following criterion:		
<ul style="list-style-type: none"> Student shows progress with respect to understanding and performing lab work based on feedback received 		
TOTAL	(/7 possible marks)	



Course policies

Requests for Relief for Missed Academic Term Work

[McMaster Student Absence Form \(MSAF\)](#): Lab times are mandatory. 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". Even with submission of an MSAF, a minimum of 9 lab weeks must be attended (i.e., attendance to lab and completion of all assessments) to be considered for completion of this course.

MSAF Course Specific Information

- ✓ The lab times are extremely important for this course and so they are mandatory. However, if the lab time must be missed due to unforeseen circumstances (such as illness), proper documentation (such as an MSAF) must be provided. Once proper documentation is provided, we will accommodate the missed lab on a case-by-case basis. Additionally, you must complete all requirements of the missed assessment component.
- ✓ Missed assessment components also require supporting documentation. We will tackle these on a case-by-case basis, but we typically ask that you complete the assessment at a later time.
- ✓ Try not to be late when handing in your assignments. Late penalties are usually 10%/day unless otherwise specified.

Academic Accommodation of Students with Disabilities

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

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 [RISO](#) 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 or to the Registrar's Office prior to their examinations. Students should contact their instructors as soon as possible to make alternative arrangements for classes, assignments, and tests.



Courses with An On-Line Element

Some courses may use on-line elements (e.g. e-mail, Avenue to Learn (A2L), LearnLink, web pages, capa, Moodle, 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, usernames 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.

Academic Integrity

The [Academic Integrity Policy](#) states that students are responsible for being aware of and demonstrating behaviour that is honest and ethical in their academic work. Breaching of academic ethics is ultimately destructive to the values of the University; it is, furthermore, unfair and discouraging to those students who pursue their studies with integrity. Academic dishonesty is to knowingly act or fail to act in a way that results or could result in unearned academic credit or advantage. It can result in serious consequences such as a grade of zero, loss of credit or even expulsion from the university.

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 team 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. A2L, 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 the [McMaster Office of Academic Integrity's](#) webpage.

Conduct Expectations

As a McMaster student, you have the right to experience, and the responsibility to demonstrate, respectful and dignified interactions within all our living, learning and working communities. These expectations are described in the [Code of Student Rights & Responsibilities \(the "Code"\)](#). 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 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.

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, A2L and/or McMaster email.