

**MBG\*3350**  
**Laboratory Methods in Molecular Biology**  
**Fall 2016**

**Department of Molecular and Cellular Biology**  
**University of Guelph**

## **COURSE OBJECTIVES**

This course involves laboratory based instruction in the basic methodologies of Molecular Biology. Students will have the opportunity to develop technical skills and practical knowledge sufficient to perform basic procedures independently, and to diagnose and analyze experimental results obtained with these techniques.

## **COURSE PERSONNEL**

**INSTRUCTOR:** Dr. A. Nassuth  
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Ext. 58787  
Email: [anassuth@uoguelph.ca](mailto:anassuth@uoguelph.ca)

**LAB  
DEMONSTRATOR:** Amanda van der Vinne  
Office: SSC 3519  
Ext: 56189  
Email: [avander@uoguelph.ca](mailto:avander@uoguelph.ca)

## **COURSE SCHEDULE**

**Laboratory:** Either Monday & Wednesday **or** Tuesday & Thursday  
1:30pm – 5:20pm  
SCIE 4108/4109

**Lecture:** Mo/Tue in the weeks of Sept 12 and 19 only: 1:30-2:30 pm in SSC2315  
Friday 1:30pm – 2:20pm ANNU 156

# LEARNING OUTCOMES

By the end of this course, students should be able to:

1. Explain the fundamental principles of practical molecular biology
2. Recognize and interpret experimental results
3. Implement the theoretical principles and apply them in the execution of lab experiments
4. Plan, design, monitor, troubleshoot and optimize experiments
5. Use online tools to research a particular topic, and read primary research articles in molecular genetics

# COURSE RESOURCES

## Lab Requirements:

**MBG\*3350 Laboratory Manual:** purchased from **SSC 2302** the first three days of the semester (Sept 8, 9 and 12; between 9:30-12 and 1- 3:30)

**A bound Laboratory Notebook**

**Lab Coat**

**Indelible (“Sharpie”) marker:** ultra-fine point

**ImageLab** (PC and Mac compatible) and **CFX Manager** (PC compatible): software provided by the lab demonstrator for download on your computer

## Courselink:

This course will make use of the University of Guelph’s course website on D2L (via Courselink). Consequently, you are responsible for all information posted on the Courselink page for MBG\*3350. Please check it regularly.

## Undergraduate Calendar:

The Undergraduate Calendar is the source of information about the University of Guelph’s procedures, policies and regulations, which apply to undergraduate programs. It can be found at: <http://www.uoguelph.ca/registrar/calendars/undergraduate/current/>.

# COURSE FORMAT

## Lecture

One lecture per week: Friday 1:30pm – 2:20pm ANNU 156. Note that in addition some lectures will be presented on Mo/Tue from 1:30 - 2:20pm in SSC2315 (i.e. during the first hour of the first lab periods) in weeks 1 to 2, to ensure that you receive relevant information before you do a lab.

## Laboratory

Two lab sessions per week: Monday/Wednesday or Tuesday/Thursday 1:30pm – 5:20pm  
SCIE 4108/4109

## Progress Reports

During the course of the semester you will be required to complete and hand in progress reports (see course outline for due dates). These reports are meant to assist you in continually monitoring the outcomes of your experiments. The reports are designed to have you analyze your results and perform calculations so your formal lab reports are a compilation of results already obtained.

## Literature Review

Before research is conducted one should have a good grasp of what is currently known for the topic/area of study. As such each student will be required to complete a literature review on His-tagged proteins and green fluorescent protein (GFP). Specific details will be presented in lab and on Courselink.

## Formal Lab Report

You are required to write one formal lab report for this course, covering the cloning and analysis of GFP. Although the report will be written in the form of a scientific manuscript, you must remember that the **audience** and **purpose** of a formal report is somewhat different than that of a scientific paper. The aim is to show that you understand the **principles** and **significance** of the experiments you performed. Remember your data will have been marked already. What is of importance here is your ability to discuss and interpret **cumulative** data in a manner that demonstrates an **understanding** of what you have accomplished in the lab and the relevance of the experiments. Further information and guidelines for your lab report will be available in lab and on Courselink.

## Assignments

One web-based assignment in the first week (to be completed during lab time) will have you use several web-based sequence analysis programs. Three other assignments will be due towards the end of the semester for the PCR optimization, qPCR GMO Analysis, and the qPCR *Arabidopsis* experiment. Instructions will be given in lab and posted to Courselink.

## Tentative Lecture Topics and Schedule

### Week 0

Lecture 1, Sept 9: Course introduction, DNA synthesis

### Week 1

Lecture 2, Sept 12/13: Plasmids, *E. coli*, spec, RE digest

Lecture 3, Sept 16: Electrophoresis, cloning

### Week 2

Lecture 4, Sept 19/20: Polymerase chain reaction (PCR) principles, primers

Lecture 5, Sept 23: Preparing vectors with desired sequences

**Week 3**

Lecture 6, Sept 30: Cloning and expression vectors

**Week 4**

Lecture 7, Oct 7: qPCR

**Week 5**

**TERM TEST 1 in class Oct 14, covering lectures & labs of weeks 0-4 (15%)  
Combination of short answer, multiple choice and problem solving questions**

**Week 6**

Lecture 8, Oct 21: Protein expression and purification

**Week 7**

Lecture 9, Oct 28: Protein quantification, analyses by gel electrophoresis

**Week 8**

Lecture 10, Nov 4: Western, Northern and Southern blotting

**Week 9**

Lecture 11, Nov 11: Nucleic acid isolation, qRT-PCR

**Week 10**

No lecture

**Week 11**

**TERM TEST # 2 in class Nov 25 covering lectures 1-10 & Labs 1-10 (15%)  
Combination of short answer, multiple choice and problem solving questions**

**Week 12**

No Lecture

**Laboratory Schedule****Week 1 (Sept 12-16)**

## Day 1

- Introduction to the lab
- Lab safety
- Molecular Biology Review
- PubMed Search
- Molecular Biology Web Tools

## Day 2

- Web exercise assignment (bring your computer!)

**Week 2 (Sept 19-23)**

## Day 1

- Micropipetting
- Plating Cultures
- Inoculating Cultures

Day 2  
Purification of Plasmid DNA  
Restriction Enzyme Digestion of pET-28a  
pET-28a Quantification

**Week 3** (Sept 26-30)

Day 1  
Agarose Gel Electrophoresis of DNA products  
Polymerase Chain Reaction of *gfp*  
Day 2  
Analysis of PCR products  
Purification of *gfp* PCR Product  
*gfp* Quantification

**Week 4** (Oct 3-7)

Day 1  
Preparing pET28a and *gfp* for Ligation  
Ligation of *gfp* into pET28a  
Day 2  
Transformation of Ligation Reactions into *E. coli* DH5 $\alpha$   
PCR Primer Design

**Week 5** (Oct 10-14)

Day 1  
*Thanksgiving break*  
Day 2  
qPCR – Isolation of DNA from a Soy sample

**Week 6** (Oct 17-21)

Day 1  
qPCR – GMO detection  
Day 2  
Isolation of pET28*gfp*  
Restriction Enzyme Digest and gel

**Week 7** (Oct 24-28)

Day 1  
Amplification of your *E. coli* gene and gel analysis  
Day 2  
Isolation of His-GFP  
His-GFP Purification Using Ni-NTA Column

**Week 8** (Oct 3-Nov 4)

Day 1  
Amplification of your *E. coli* gene (continue until successful)  
Day 2  
SDS-PAGE and Coomassie Stain

**Week 9** (Nov 7-11)

Day 1  
SDS-PAGE and Western Immunoblotting

Day 2  
SDS-PAGE and Western Immunoblotting

**Week 10** (Nov 14 -18)

Day 1  
qPCR – RNA Isolation from *Arabidopsis*  
Day 2  
qPCR – *Arabidopsis* Gene Expression Assay

**Week 11 &12** (Nov 21 – Dec 2)

Day 1 No Lab Scheduled  
Day 2 No Lab Scheduled

**Key Dates (where two dates are listed: the first applies to sections 1 and 2, the second applies for sections 3 and 4)**

Sept 14/15:	Web exercise assignment
Sept 28/29:	Literature Review
Oct 3/4:	Progress Report #1
Oct 12/13:	Progress Report #2
Oct 14:	Term test #1
Oct 24/25:	Progress Report #3
Oct 31/Nov1:	qPCR GMO Assignment
Nov 4:	40th class day--Last day to drop the course
Nov 7/8:	Progress Report #4
Nov 14/15:	Progress Report #5
Mar 16/17:	PCR Report
Nov 23/24:	Lab Report
Nov 25:	Term test #2
Nov 30/Dec1:	qPCR <i>Arabidopsis</i> Assignment

## METHODS OF ASSESSMENT

Assessment				
Form of Assessment	Weight of Assessment (% of final)	Due Date of Assessment	Course Content /Activity	Learning Outcome (see above)
Web-based Assignment	3%	Sept 14/15	Lab Weeks 1-11	3 and 5
Literature Review*	10%	Sept 28/29	Lab Weeks 1-11	1 and 5
Progress Reports (5)	20% (4% each)	Various (see key dates)	Lab Weeks 1-9	2, 3 and 4
Lab Report*	10%	Nov 23/24	Lab Weeks 1-9	1, 2, 3, 4 and 5
PCR Report*	4%	Nov 16/17	Lab Weeks 4-11	1,2, 3 and 4
GMO Assign.*	4%	Oct 31/Nov1	Lab Week 10	1, 2, 3, 4 and 5
<i>Arabidopsis</i> Assign.*	4%	Nov 30/Dec1	Lab Week 11	1, 2, 3, 4 and 5
Lab Performance	15%	Sept 9 - Nov 17	Lab Weeks 2-11	2, 3 and 4
Term test 1	15%	Oct 14	Lectures and Labs weeks 1-4	1,2, 3 and 4
Term test 2	15%	Nov 25	Lectures and Labs weeks 1-11	1,2, 3 and 4

\*Assignments must be typed, double-spaced, 12-point font.

You will receive information for the week 1 Web-based assignment in the lab.

The three later Assignments are due at 1:30pm on your scheduled lab day. Late assignments will be accepted without penalty only for medical or compassionate reasons with documentation. Late assignments without documentation will be penalized 10% per day up to 50%. A grade of zero is assigned after 5 days late.

Progress Reports are submitted on your first lab day of the week and are returned on the second lab day of the week. This is to provide you with immediate feedback as to whether your analysis, interpretation and conclusion of your experimental results are correct.

The lab performance grade is determined by your performance in the lab. Of this, 10% is based on your actual results (success of your experiments). The other 5% is based on your day to day performance in the lab: punctuality, attendance, attitude, preparedness, independence etc.

**Term test 1 and 2 will be held during regular lecture time; if you fail to write the quiz 1 a grade of 0% will be assigned unless an acceptable cause such as sickness or family emergency is documented. In the situation where academic consideration is given the Term test 2 will be adjusted to 30%. For missed Term test 2 an Incomplete grade will be submitted with a recommendation of 0% unless academic consideration is granted for a deferred exam.**

## **COURSE AND UNIVERSITY POLICIES**

You must come to lab prepared and ready to start working by 1:30

It is disrespectful to arrive late as this interrupts the TA, your partner and your fellow classmates. Additionally you will miss out on specific announcements for the day that the TA is not obligated to repeat. If you miss specific safety announcements you may be asked to leave. During the course of the lab there may be times where you can get a coffee as you have a gel running. Feel free to do so, however, if any announcements or discussions take place during your absence you will be responsible for obtaining the information from a fellow classmate.

Lab Attendance is mandatory

This is a lab based course where the majority of your final grade is assigned based on the laboratory component rather than the lecture component of the course. The nature of the lab exercises also build on one another. As such there is no opportunity for make-up labs. Lab absence is only acceptable for medical or compassionate reasons.

You must keep a lab notebook

Before coming to lab you must record in your lab notebook: What are you doing in lab today? What are the expected results? You must have completed all calculations that are required to carry out the experiments.

Additionally you should record the variables of the experiment (reaction conditions), insert the actual results you obtained, in table format or gel image (labelled) and a statement of whether or not the experiment was successful. Your lab notebook will be graded for the PCR assignment.



### All Assignments Have to be Completed

ALL lab assignments are an important part of the course. Failure to complete an assignment will lead to an “incomplete” at the end of the semester.

### When You Cannot Meet a Course Requirement

When you find yourself unable to meet an in-course requirement because of illness or compassionate reasons, please advise the course instructor (or designated person, such as a teaching assistant) in writing, with your name, id#, and e-mail contact. See the undergraduate calendar for information on regulations and procedures for Academic Consideration: <http://www.uoguelph.ca/registrar/calendars/undergraduate/current/c08/c08-ac.shtml>

### Accessibility

The University of Guelph is committed to creating a barrier-free environment. Providing services for students is a shared responsibility among students, faculty and administrators. This relationship is based on respect of individual rights, the dignity of the individual and the University community's shared commitment to an open and supportive learning environment. Students requiring service or accommodation, whether due to an identified, ongoing disability or a short-term disability should contact the Centre for Students with Disabilities as soon as possible.

For more information, contact CSD at 519-824-4120 ext. 56208 or email [csd@uoguelph.ca](mailto:csd@uoguelph.ca) or see the website: <http://www.uoguelph.ca/csd/>

### Academic Misconduct

It is the nature of undergraduate labs to complete experiments with a partner. Your results should be discussed with your partner as this is expected in all scientific research. However, **ALL** assignments must be completed **INDEPENDENTLY**.

The University of Guelph is committed to upholding the highest standards of academic integrity and it is the responsibility of all members of the University community – faculty, staff, and students – to be aware of what constitutes academic misconduct and to do as much as possible to prevent academic offences from occurring. University of Guelph students have the responsibility of abiding by the University's policy on academic misconduct regardless of their location of study; faculty, staff and students have the responsibility of supporting an environment that discourages misconduct. Students need to remain aware that instructors have access to and the right to use electronic and other means of detection.

Please note: Whether or not a student intended to commit academic misconduct is not relevant for a finding of guilt. Hurried or careless submission of assignments does not excuse students from responsibility for verifying the academic integrity of their work before submitting it. Students who are in any doubt as to whether an action on their part could be construed as an academic offence should consult with a faculty member or faculty advisor.

The Academic Misconduct Policy is detailed in the Undergraduate Calendar: <http://www.uoguelph.ca/registrar/calendars/undergraduate/current/c08/c08-amisconduct.shtml>

### E-mail Communication

As per university regulations, all students are required to check their <uoguelph.ca> e-mail account regularly. E-mail is the official route of communication between the University and its students.

### Drop Date

The last date to drop one-semester courses, without academic penalty, is the 40<sup>th</sup> class day. To confirm the actual date, please see the schedule of dates in the Undergraduate Calendar. For regulations and procedures for Dropping Courses, see the Undergraduate Calendar: <http://www.uoguelph.ca/registrar/calendars/undergraduate/current/c08/c08-drop.shtml>

### Copies of out-of-class assignments

Keep paper and/or other reliable back-up copies of all out-of-class assignments: you may be asked to resubmit work at any time.

### Recording of Materials

Presentations which are made in relation to course work—including lectures—cannot be recorded or copied without the permission of the presenter, whether the instructor, a classmate or guest lecturer. Material recorded with permission is restricted to use for that course unless further permission is granted.

### Resources

The Academic Calendars are the source of information about the University of Guelph's procedures, policies and regulations which apply to undergraduate, graduate and diploma programs:  
<http://www.uoguelph.ca/registrar/calendars/index.cfm?index>

### Grading

All assignments are due at 1:30pm and a hard copy is to be submitted in lab.

Students who wish to have their assignments re-graded must submit them to the Lab Demonstrator within 5 class days of their return. The entire assignment will be re-graded so the mark may go up, down or remain unchanged.

### Turnitin

In this course, your instructor will be using Turnitin, integrated with the CourseLink Dropbox tool, to detect possible plagiarism, unauthorized collaboration or copying as part of the ongoing efforts to maintain academic integrity at the University of Guelph.

All submitted assignments will be included as source documents in the Turnitin.com reference database solely for the purpose of detecting plagiarism of such papers. Use of the Turnitin.com service is subject to the Usage Policy posted on the Turnitin.com site.

A major benefit of using Turnitin is that students will be able to educate and empower themselves in preventing academic misconduct. In this course, you may screen your own assignments through Turnitin as many times as you wish before the due date. You will be able to see and print reports that show you exactly where you have properly and improperly referenced the outside sources and materials in your assignment.

Note: The final copy has to be a hard copy.

## CAMPUS RESOURCES

If you are concerned about any aspect of your academic program:

- make an appointment with a program counsellor in your degree program.  
<http://www.bsc.uoguelph.ca/index.shtml> or  
<https://www.uoguelph.ca/uaic/programcounsellors>

If you are struggling to succeed academically:

- There are numerous academic resources offered by the Learning Commons including, Supported Learning Groups for a variety of courses, workshops related to time management, taking multiple choice exams, and general study skills. You can also set up individualized appointments with a learning specialist.  
<http://www.learningcommons.uoguelph.ca/>

If you are struggling with personal or health issues:

- Counselling services offers individualized appointments to help students work through personal struggles that may be impacting their academic performance.  
<https://www.uoguelph.ca/counselling/>
- Student Health Services is located on campus and is available to provide medical attention.  
<https://www.uoguelph.ca/studenthealthservices/clinic>
- For support related to stress and anxiety, besides Health Services and Counselling Services, Kathy Somers runs training workshops and one-on-one sessions related to stress management and high performance situations. <http://www.uoguelph.ca/~ksomers/>

If you have a documented disability or think you may have a disability:

The Centre for Students with Disabilities (CSD) can provide services and support for students with a documented learning or physical disability. They can also provide information about how

to be tested for a learning disability. For more information, including how to register with the centre please see: <https://www.uoguelph.ca/csd/>