

Biology 3326F (Cell Biology Laboratory) Course Outline

1. General Course Information

Course Information

Biology 3326F (Cell Biology Laboratory), **Fall 2022**

Lectures/Tutorials Mondays, 1:30 pm-2:30 pm, BGS-1056

Lab sections: section 002 (Wednesdays) and section 003 (Thursdays), BGS-3077, 1:30pm-5:30 pm

List of Prerequisites

The prerequisites for this course are Biology 2290F/G, Biochemistry 2280A; a minimum mark of 70% in Biology 2382A/B.

Unless you have either the prerequisites for this course or written special permission from your Dean to enroll in it, you may be removed from this course and it will be deleted from your record. This decision may not be appealed. You will receive no adjustment to your fees in the event that you are dropped from a course for failing to have the necessary prerequisites.

2. Instructor and Contact Information

Instructor: Dr. Alexander Timoshenko

Office hours: Mondays, 3:00 pm to 4:00 pm, BGS-3032

Tel.: 519-661-2111 ex. 88900, E-mail: atimoshe@uwo.ca

Teaching Assistants: Ryley Yost (ryost@uwo.ca) and Stefan Grahovac (sgrahov@uwo.ca)

Laboratory Technician: Chunhui Li (cli93@uwo.ca)

Students must use their Western (@uwo.ca) email addresses and include Bio3326 in the subject line when contacting their instructor or TAs.

3. Course Syllabus, Schedule, Delivery Mode

Course description: Biology 3326F/G is a laboratory course which provides hand-on training in current cell biological methods such as animal cell culture, microscopy, karyotype analysis, PCR, SDS-PAGE, fluorescence staining of cellular organelles, and bioimaging. A discovery-based component of this course focuses on application of the mentioned methods to study changes in protein expression and cytoskeleton organization in cells exposed to microenvironmental stress stimuli.

Course Content and Learning Outcomes: The work in this course is divided into two modules, outlined below. **The first module** is intended to give students an understanding of and experience with the basic animal cell culture techniques and cell line characterization and authentication. **The second module** is based on application of SDS-PAGE and fluorescence microscopy methods to study protein expression

and remodeling of cytoskeleton systems in cultured cells in response to microenvironmental stress stimuli. The major training objectives are:

Module I. Basics of Animal Cell Culture: • Examining the microscope potential (upright vs inverted; magnification; numerical aperture; resolution; field of view; working distance) • Proper alignment of a bright-field light microscope (Köhler illumination) and a phase contrast microscope • Identification of cells and cellular components on slides and in cell culture flasks • Harvesting methods for adherent cell lines • Cell counts using hemacytometer • Cell density and confluency • Test for cell viability • Monitoring morphology of animal cells in culture • Bioimaging using an inverted microscope • Chromosome spreads from monolayer cultures • Staining procedure for chromosome spreads with Giemsa and DAPI • Karyotype analysis of a cell line • Modal number of chromosomes and heteroploidy • Genetic instability in cell culture • Genomic DNA isolation • PCR assay for identifying cell lines • Agarose gel electrophoresis.

Module II. Cell Stress Biology and Bioimaging: • Preparation of cell lysates using RIPA buffer • Spectrophotometrical determination of protein concentration using Bradford assay • SDS-PAGE • Staining proteins in gels using Coomassie blue • Gel analysis using ImageLab software (Bio-Rad) • Fluorescence microscopy • Fluorochromes • Fluorescence microscope operation • Image capture • Direct fluorescence staining of actin microfilaments with phalloidin • Preparation of slides for fluorescence microscopy • Fixation procedure • Monoclonal and polyclonal antibody • Primary and secondary antibody • Indirect immunofluorescence staining • Blocking solution • Observation of microfilament, microtubules and intermediate filament modifications in response to stress stimuli • Bioimaging using fluorescence microscope • Northern Eclipse and ImageJ software.

By the end of the course, successful students will be able to:

- ☒ Demonstrate a knowledge of animal cell culture models in cell biology.
- ☒ Perform animal cell line characterization using regular light, inverted and fluorescence microscopy, karyotyping, SDS-PAGE, and PCR assay.
- ☒ Isolate and quantify genomic DNA and proteins in animal cells.
- ☒ Run horizontal and vertical electrophoresis to analyze biological molecules.
- ☒ Prepare chromosome spreads, stain chromosomes, and analyze cell karyotype.
- ☒ Stain the cytoskeleton and cell nucleus using fluorescent probes.
- ☒ Operate with inverted and fluorescence microscopes and perform bioimaging of live and stained animal cells.
- ☒ Understand the complexity and nature of the cell cytoskeleton and its remodeling under stress conditions.
- ☒ Analyze data using professional software such as ImageLab and ImageJ.
- ☒ Organize and analyse research projects by formulating hypotheses for each laboratory experiment and providing an interpretation of collected data in laboratory notebooks.
- ☒ Demonstrate competence with reading primary research articles, scientific writing, data assessment, and critical thinking by reporting their experimental results in a form of research paper manuscript and poster presentation.
- ☒ Develop good teamwork habits by working in pairs and discussing the results with lab partners and in the class.

Lectures/Tutorials Schedule: Lectures/Tutorials will be delivered in-person and held on Mondays at 1:30pm in **BGS-1056**. We will discuss the results collected in the lab, statistics, software for data analysis, assignment requirements, and upcoming lab classes.

Weeks	Dates (Mondays)	Tutorial topics
Week 1	September 5	No tutorial this week (classes begin September 8)
Week 2	September 12	Orientation and the first lab overview
Week 3	September 19	Animal cell culture methods
Week 4	September 26	Authentication of animal cell lines
Week 5	October 3	Midterm paper requirements
Week 6	October 10	Thanksgiving Day
Week 7	October 17	In-class Test #1
Week 8	October 24	Animal cell lysis, protein assay, and SDS-PAGE
Week 9	October 31	Reading Week (October 31 – November 6), no classes
Week 10	November 7	Methods of fluorescence staining in cell biology
Week 11	November 14	Data analysis; ImageJ
Week 12	November 21	Poster project requirements
Week 13	November 28	In-class Test #2
Week 14	December 5	Student's questions

Laboratory Schedule: In-person lab classes (4 h) are held on Wednesdays (section 002) and Thursdays (section 003) from 1:30pm to 5:30pm in **B&GS 3077**.

Weeks	Dates (Wed/Thu)	Laboratory classes
Week 1	September 8	Classes begin, no labs this week
Week 2	September 14/15	Light microscopy in cell biology
Week 3	September 21/22	Animal cell culture methods (trypsinization, cell counts, viability assay)
Week 4	September 28/29	Cell line characterization by chromosomal staining
Week 5	October 5/6	Genomic DNA isolation and PCR
Week 6	October 12/13	Agarose gel electrophoresis and gel imaging (ImageLab)
Week 7	October 19/20	Quantification of total proteins in animal cell lysates
Week 8	October 26/27	SDS-PAGE, protein staining and quantification on gels
Week 9	November 2/3	Reading Week (October 31 – November 6), no classes
Week 10	November 9/10	Rotation labs: Fluorescence staining of the cytoskeleton and nuclei, microscopy, and bioimaging
Week 11	November 16/17	
Week 12	November 23/24	
Week 13	Nov 30/Dec 1	Poster project discussion with individual groups
Week 14	December 7/8	Poster Presentations

Contingency plan for an in-person class pivoting to 100% online learning: In the event of a COVID-19 resurgence during the course that necessitates the course delivery moving away from face-to-face interaction, affected course content will be delivered entirely online, either synchronously (i.e., at the times indicated in the timetable) or asynchronously (e.g., posted on OWL for students to view at their convenience). The grading scheme will **not** change. Any remaining assessments will also be conducted online as determined by the course instructor.

4. Course Materials

Required materials from the Book Store at Western:

1. *Life Sciences Student Lab Notebook* (ISBN: 9781930882355, Publisher: Hayden-McNeil)
2. Student laboratory manual "**Biology 3326F/G Cell Biology Laboratory**", version 2021-22, # M12141.

Additional readings (available on reserve in the Taylor Library):

1. Freshney, R.I. (2016) **Culture of animal cells: a manual of basic technique and specialized applications, Seventh Edition**, Willey-Blackwell. (Call No. Q525.F885c 2016). This textbook is also available online.
2. Lodish, H., et al. (2016) **Molecular Cell Biology, 8th Edition**, W.H. Freeman and Company, NY (**Chapter 4**, sections 4.1, 4.2; **Chapters: 8**, sections 8.5, 8.6; **Chapters 17, 18**).
3. Alberts, B. et al. (2015) **Molecular Biology of The Cell, 6th Edition**, Garland Science, NY. (Call No. QH581.2.M64 2015).
4. Knisely, K. (2013) **A Student Handbook for Writing in Biology, 4th Edition**, Sinauer Associates (Call No. QH304.K59 2013). (New edition of this textbook is also available in the Book Store at Western).

Lecture notes, lab results, assignments, and grades will be available through OWL: All course material will be posted to OWL: <http://owl.uwo.ca>. Any changes will be indicated on the OWL site and discussed with the class. Students are responsible for checking the course OWL site (<http://owl.uwo.ca>) on a regular basis for news and updates. This is the primary method by which information will be disseminated to all students in the class. If students need assistance with the course OWL, they can seek support on the [OWL Help page](#). Alternatively, they can contact the [Western Technology Services Helpdesk](#). They can be contacted by phone at 519-661-3800 or ext. 83800. [Google Chrome](#) or [Mozilla Firefox](#) are the preferred browsers to optimally use OWL; update your browsers frequently.

Copyright and Audio/Video Recording Statement: Course material produced by faculty is copyrighted and to reproduce this material for any purposes other than your own educational use contravenes Canadian Copyright Laws. You must always ask permission to record another individual and you should never share or distribute recordings.

Student Responsibilities and General Conduct in Teaching Laboratory: All persons working in a laboratory are required to follow the Western University Safety Policy:

1. Laboratory dress code (shirt, long pants, socks and close-toed shoes).
2. Proper lab attire including safety glasses and lab coats.
3. Disposable gloves are to be worn in the laboratory.
4. No food or drinks are allowed in the laboratory.
5. Medical-grade (ASTM level 3) masks are to be worn in indoor instructional spaces.

Technical Requirements: When offered in online mode all students must have access to a stable internet connection, and a computer with working microphone and/or webcam. During in-person learning it is highly recommended that students bring a portable computer to lab and tutorial that can be used for data analysis.

5. Methods of Evaluation

Click [here](#) for a detailed and comprehensive set of policies and regulations concerning examinations and grading. The table below outlines the University-wide grade descriptors.

A+	90-100	One could scarcely expect better from a student at this level
A	80-89	Superior work which is clearly above average
B	70-79	Good work, meeting all requirements, and eminently satisfactory
C	60-69	Competent work, meeting requirements
D	50-59	Fair work, minimally acceptable
F	below 50	Fail

The overall course grade - **out of 100** - will be calculated as specified below.

Component	Notes	Value
Class participation	Based on tutorial attendance (1 point) and lab work (4 points)	5
Lab notebook	Marked 2 times by TAs (Module I and Module II)	15
Module tests	Two in-class tests (40 min), 20 multiple-choice questions each time, Mondays: October 17 and November 28	25
Midterm assignment	Written report due Monday, October 24	30
Poster Project	Final presentations in this class, December 7/8	25

Class Participation

Class participation includes tutorial attendance (1 point) and lab participation (4 points). Records of tutorial attendance will be taken during each session. The lab participation will be evaluated by your TAs and the average grade will be submitted to the instructor based on following four grading criteria, each is worth of 1 point:

1. **Lab Safety** (lab coat, safety glasses, long pants, socks and close-toed shoes, confine long hairs, no food/drinks in the lab).
2. **Punctuality and Readiness** (late to class, no lab notebook, did not read lab notes).
3. **Performance and Professionalism** (extra time, not doing right, proper labelling on slides, fail to clean lab benches, accuracy of lab reports, teamwork, cell phones, inappropriate behaviour).
4. **Engagement** (knowledge of protocols and workflow, proactive contribution, offering ideas, enthusiasm, ask appropriate questions).

Lab Notebook

Complete and accurate record keeping is one of the most important elements in any piece of laboratory work. There are many acceptable forms of record keeping, depending on the nature of the

study. In this course, you will be required to use *Life Sciences Student Lab Notebook* (ISBN: 9781930882355, Publisher: Hayden-McNeil). Your lab notebook **must** be with you whenever you are in the lab. Here are a few guidelines to keep in mind:

(a) Make entries in the notebook as and when you do things. Do not transcribe records for the sole purpose of making the book "look neat". It is important to note exactly what you did.

(b) The notebook should contain everything that is relevant to your project including literature, methods, protocols, images, etc. Keep notes or copies of your literature searches, relevant papers, ideas for solving problems in your lab notebook.

(c) All raw data must be entered in the notebook. There will be some breaks between exercises in the lab; use them to enter all required information in your notebook. Tape in all images you collect. Also use your OWL **Drop Box** to save files with raw data, images, and other relevant materials.

(d) Lab notebooks should be sufficiently complete to allow any laboratory-literate person to duplicate your experiments and analyses. When it comes time to prepare your manuscript and poster presentation, your lab notebook will be your primary source of information.

The lab notebooks will be checked 2 times by TAs. The grades will be averaged and contribute to **15%** of your final grade. Grade zero will be assigned for each missing submission because we expect that you consistently take care about records in your lab notebook. The following **Lab Notebook Markers** will apply:

1. TOC, pages and entries dated (1 point).
2. Purpose of experiment, hypothesis if applied (2 points).
3. Experimental procedure, protocols (3 point).
4. Raw data, drawings, photos, calculations, figures, tables, etc (5 points).
5. Interpretation, ideas (2 points).
6. Overall clarity and readability of your records (2 points).

Module tests

Two module tests (12.5 points each, based on 20 multiple-choice questions format) will be given during tutorial time at the end of each lab module. The length of each test is 40 min. Test #1 (Monday, October 17) will cover module I (microscopy, animal cell culture, karyotyping, PCR) and Test #2 (Monday, November 28) will cover module II (SDS-PAGE, cytoskeleton, fluorescence microscopy, cell staining and bioimaging).

Midterm Assignment

Bio3326F assignments are designed to introduce you to scientific writing and to provide you with proper understanding of formal requirements to **research paper manuscripts** for scientific journals. The assignments will be based on your original experimental results obtained in the module I of the lab course (i.e. animal cell culture, PCR, and karyotype of animal cell lines) and their critical analysis using the literature data. The midterm assignments are allowed to be prepared by two students working together on the same bench, who can share their results, implying that the same grade will be applied to co-authors. We expect that such format will encourage the teamwork and discussion of the collected data. The midterm assignment is to be organized as a formal manuscript for **The Journal of Cell Biology** as per the Instructions to Authors of this journal (separate handout) and must be submitted by Monday, October 24, 11:50pm as an electronic copy on OWL (late submissions will be penalized at a rate of 5% per day). **Midterm assignment must be handed in to pass the course.**

Poster Project

BIO3326F/G posters are group projects of 3-4 students working with same stress stimuli, usually on the same bench. The electronic version of posters (no hard copies) will be presented as a team work by group members during special sessions covering lab hours (1:30-5:30pm) on December 7 and December 8. The poster project will be based on the results of your lab work over the module II including effects of stress on protein expression (SDS-PAGE) and on the cytoskeleton in cultured cells. The poster presentations will be your final report in this class. **Please note that the PowerPoint file of your poster should be submitted on OWL one day in advance of your presentation, i.e. by 1:30pm on the preceding day.** The peer evaluation may change the portion of your poster group mark by up to 10%. A separate handout provides you with the guidelines and instructions for the preparation and peer evaluation of BIO3326F/G Poster. **Poster project and presentation must be completed to pass the course.**

6. Student Absences

Attendance in the lab and completion of all lab exercises is mandatory. If you miss a lab class due to a valid reason confirmed by the Dean's office, your average grade will be applied to that class and the case will be dealt with on an individual basis. It is the student's responsibility to make alternative arrangements with their instructor once the accommodation has been approved and the instructor has been informed. If you do not have the confirmation from Dean's office, there will be 5% deduction from your final mark (i.e. 1 unexcused lab absence results in a maximum possible final grade of 95%). If a student misses **more than two labs**, and therefore do not demonstrate sufficient mastery of those technical skills to progress and data collection for reports, they may not receive credit for the course or receive a grade of Incomplete (INC) with subsequent academic outcomes.

Tutorial attendance is mandatory, and attendance will be taken. There will be a deduction of 5% from the final tutorial mark for each unexcused absence.

Late submissions of the take-home assignment will be penalized at a rate of 5% per day (including the weekend) until the percentage of the assignment is used up.

Missed assignments, tests, presentations will not be reweighted to other parts of the course.

For work totaling less than 10% of the final course grade, extensions will be considered by the instructor.

For work totaling 10% or more of the final course grade, you must provide valid medical or supporting documentation to the Academic Counselling Office of your Faculty of Registration as soon as possible. For further information, please consult the University's medical illness policy at

https://www.uwo.ca/univsec/pdf/academic_policies/appeals/accommodation_medical.pdf.

The Student Medical Certificate is available at

https://www.uwo.ca/univsec/pdf/academic_policies/appeals/medicalform.pdf.

Students should note that individual instructors are not permitted to receive documentation directly from a student, whether in support of an application for consideration on medical grounds, or for other reasons.

7. Accommodation and Accessibility

Religious Accommodation

When a course requirement conflicts with a religious holiday that requires an absence from the University or prohibits certain activities, students should request accommodation for their absence in writing at least two weeks prior to the holiday to the course instructor and/or the Academic Counselling office of their Faculty of Registration. Please consult University's list of recognized religious holidays (updated annually) at <https://multiculturalcalendar.com/ecal/index.php?s=c-univwo>.

Accommodation Policies

Students with disabilities are encouraged to contact Accessible Education, which provides recommendations for accommodation based on medical documentation or psychological and cognitive testing. The policy on Academic Accommodation for Students with Disabilities can be found at [https://www.uwo.ca/univsec/pdf/academic_policies/appeals/Academic Accommodation_disabilities.pdf](https://www.uwo.ca/univsec/pdf/academic_policies/appeals/Academic_Accommodation_disabilities.pdf).

8. Academic Policies

The website for Registrar Services is <http://www.registrar.uwo.ca>.

In accordance with policy, <http://www.uwo.ca/its/identity/activatenonstudent.html>, the centrally administered e-mail account provided to students will be considered the individual's official university e-mail address. It is the responsibility of the account holder to ensure that e-mail received from the University at his/her official university address is attended to in a timely manner.

Participants in this course are not permitted to record the sessions, except where recording is an approved accommodation, or the participant has the prior written permission of the instructor.

Scholastic offences are taken seriously and students are directed to read the appropriate policy, specifically, the definition of what constitutes a Scholastic Offence, at the following Web site: http://www.uwo.ca/univsec/pdf/academic_policies/appeals/scholastic_discipline_undergrad.pdf.

All required papers may be subject to submission for textual similarity review to the commercial plagiarism detection software under license to the University for the detection of plagiarism. All papers submitted for such checking will be included as source documents in the reference database for the purpose of detecting plagiarism of papers subsequently submitted to the system. Use of the service is subject to the licensing agreement, currently between The University of Western Ontario and Turnitin.com (<http://www.turnitin.com>).

Professionalism & Privacy

Western students are expected to follow the [Student Code of Conduct](#). Additionally, the following expectations and professional conduct apply to this course:

- ☒ Students are expected to be professional and scholarly in all online postings. Use proper grammar and spelling. Cite the ideas of others appropriately.
- ☒ All course materials created by the instructor are copyrighted and cannot be sold/shared
- ☒ Recordings are not permitted (audio or video) without explicit permission

- ☒ Permitted recordings are not to be distributed
- ☒ Students will be expected to take an academic integrity pledge before some assessments
- ☒ All recorded sessions will remain within the course site or unlisted if streamed

9. Support Services

Please visit the Science & Basic Medical Sciences Academic Counselling webpage for information on adding/dropping courses, academic considerations for absences, appeals, exam conflicts, and many other academic related matters: <https://www.uwo.ca/sci/counselling/>

Students who are in emotional/mental distress should refer to Mental Health@Western (<https://uwo.ca/health/>) for a complete list of options about how to obtain help.

Western is committed to reducing incidents of gender-based and sexual violence and providing compassionate support to anyone who has gone through these traumatic events. If you have experienced sexual or gender-based violence (either recently or in the past), you will find information about support services for survivors, including emergency contacts at

https://www.uwo.ca/health/student_support/survivor_support/get-help.html.

To connect with a case manager or set up an appointment, please contact support@uwo.ca.

Please contact the course instructor if you require lecture or printed material in an alternate format or if any other arrangements can make this course more accessible to you. You may also wish to contact Accessible Education at

http://academicsupport.uwo.ca/accessible_education/index.html

if you have any questions regarding accommodations.

Learning-skills counsellors at the Student Development Centre (<https://learning.uwo.ca>) are ready to help you improve your learning skills. They offer presentations on strategies for improving time management, multiple-choice exam preparation/writing, textbook reading, and more. Individual support is offered throughout the Fall/Winter terms in the drop-in Learning Help Centre, and year-round through individual counselling.

Western University is committed to a thriving campus as we deliver our courses in the mixed model of both virtual and face-to-face formats. We encourage you to check out the Digital Student Experience website to manage your academics and well-being: <https://www.uwo.ca/se/digital/>.

Additional student-run support services are offered by the USC, <https://westernusc.ca/services/>.

This course is supported by the Science Student Donation Fund. If you are a BSc or BMSc student registered in the Faculty of Science or Schulich School of Medicine and Dentistry, you pay the Science Student Donation Fee. This fee contributes to the Science Student Donation Fund, which is administered by the Science Students' Council (SSC). One or more grants from the Fund have allowed for the purchase of equipment integral to teaching this course. You may opt out of the Fee by the end of September of each academic year by completing the online form linked from the Faculty of Science's Academic Counselling site. For further information on the process of awarding grants from the Fund or how these grants have benefitted undergraduate education in this course, consult the Chair of the Department or email the Science Students' Council at ssc@uwo.ca.

10. How to Be Successful in This Class?

Students enrolled in this class should understand the level of autonomy and self-discipline required to be successful outside the lab hours:

1. Invest in a planner or application to keep track of your courses. Populate all deadlines at the start of the term and schedule time at the start of each week to get organized and manage your time.
2. Make it a daily habit to log onto OWL to ensure you have seen everything posted to help you succeed in this class.
3. Take notes as you go through the lecture/tutorial material. Keeping handwritten notes or even notes on a regular Word document will help you learn effectively.
4. Connect with others. Try forming an online study group and try meeting on a weekly basis for study and peer support.
5. Do not be afraid to ask questions. If you are struggling with a topic, check the online discussion Forums or contact your instructor and or teaching assistant.
6. Reward yourself for successes. It seems easier to motivate ourselves knowing that there is something waiting for us at the end of the task.
7. Take advantages of office hours with your instructor on Mondays (3:00 pm to 4:00 pm).

11. Equal Opportunity and Evaluation Policy

All individuals involved in the offering of Biology 3326F/G were, at one time, undergraduate students themselves. Accordingly, your professor and your teaching assistants thoroughly understand the importance of course grades and the hard work that you will invest into this course. They are there to help you achieve your goals. We want you to do well in the course, but we also have to be fair. The university is committed to academic integrity and has high ethical and moral standards. All students will be treated equally and evaluated using the criteria presented in this course outline and their respective weights. The evaluation criteria are based strictly on actual achievement, not on effort or how hard the student tried. Claims of an excellent academic history, of attendance in the course components, or of personal issues (family, relationship, financial, etc.) cannot be used to justify a higher grade in the course because they are not criteria for evaluation. There is no extra work available for extra credit or to “make up” another grade. We do not offer any extra assignments, essays, or other work of any kind to any student. The requirement for a higher grade in order to, for example, maintain a scholarship, enter a program, or obtain a higher GPA for various reasons, is not a justifiable reason for increasing your grade. If we increased or “bumped” your grade (*i.e.* gave you a grade that you did not legitimately earn), it would be unfair to the other students and also a great disservice to the scholarships and programs who are evaluating all students on the basis of their grades.

12. Land acknowledgment

We acknowledge that Western University is located on the traditional lands of the Anishinaabek, Haudenosaunee, Lūnaapēwak and Attawandaron peoples, on lands connected with the London Township and Sombra Treaties of 1796 and the Dish with One Spoon Covenant Wampum. This land continues to be home to diverse Indigenous peoples (e.g. First Nations, Métis and Inuit) whom we recognize as contemporary stewards of the land and vital contributors of our society.

Good Luck with your Studies!