

BCMB 406A

Laboratory Manual

Fall 2021

Student Name: _____ Section: _____

Email address: _____

Instructors and Contact Information

| Lab | Instructor | Email | Phone | Office |
|-----|-----------------|------------------|--------------|------------|
| 1 | Stephen Redpath | sredpath@uvic.ca | 250-721-7076 | Petch 179a |
| 2 | Erika Wall | ewall@uvic.ca | 250-472-5119 | Petch 190 |

Each instructor is responsible for a different portion of the course as indicated above.
Please address any concerns or questions to the appropriate instructor.

We acknowledge and respect the ~~ONW % 3/4% Q~~ ^Ū people on whose traditional territory the university stands and the Songhees, Esquimalt D Q G : 6 È 1 (ū S H R S O H V Z K R V H K L relationships with the land continue to this day.

Department of Biochemistry and Microbiology
University of Victoria

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BCMB 406A Laboratory Schedule

| Week | Dates | Lab(s) | Day 1 (5 hours) | Day 2 (2 hours) | Due Dates |
|------|----------------|---|--|--------------------------------------|-----------|
| 1 | Sep 13-17 | Lab 1: Isolation & Identification of Peptides & Proteins | Bradford assay | 1 st dimension of 2D-PAGE | |
| 2 | Sep 20-24 | | 2 nd dimension of 2D-PAGE | Gel imaging & spot excision | |
| 3 | Sep 27 – Oct 1 | | Tryptic digestion B04 sign up for different section | Zip-tip plate spotting | |
| 4 | Oct 4-8 | | Mass spec Cell culture & cell staining | Complete cell staining | |
| | | Lab 2: Immunological Characterization of Cancer Cell Lines | | | |

Course Description

In BCMB 406A, you will develop your practical skills to prepare samples for analysis using modern methods in laboratory research. In this process, you will become familiar with current techniques in biochemistry and microbiology; specifically you will get experience running samples on a mass spectrometer, HPLC, flow cytometer and QPCR machine. These 3-4 week experiments offer an introduction to project-based learning experiences where you will be given an opportunity to plan some logistical portions of your own experiment. Once samples have been processed, you will critically examine and interpret multiple sets of data to study complex problems.

In lab 1, you will learn how mass spectrometry and HPLC are used in the field of proteomics. In two parallel experiments, *E.coli* proteins will be separated by 2D gel electrophoresis and a Glu-C digest of insulin will be used to generate peptides that will be separated using HPLC. The resulting purified proteins/peptides will be identified by mass spectrometry.

The focus of Lab 2 is tumour immunology and flow cytometry. You will work with a tumour-derived cell line and characterize its phenotype by staining with fluorescently labeled antibodies and analyzing using flow cytometry. You will also evaluate the ability of these cells to elicit a T-cell response using a functional immune assay.

In lab 3, epigenetic theory is applied to data generated by performing QPCR on ChIP-enriched DNA. The first step of this experiment is isolating chromatin from yeast. This chromatin is then fragmented into smaller pieces using a micrococcal nuclease digestion in preparation for ChIP. After specific pieces of chromatin are immuno-precipitated, the resulting DNA will be analyzed using QPCR and conventional PCR.

Intended Learning Objectives:

Upon completion of this course you will be able to:

- Describe the theory and principles of mass spectrometry, HPLC, flow cytometry and QPCR
- Develop proficiency in practical skills used for protein and DNA sample preparation, cell culture, flow cytometry, epigenetics and ChIP
- Evaluate experimental controls
- Solve typical calculations used in a biochemistry/microbiology lab
- Generate a record of your experimental work and data in a lab journal
- Evaluate data generated and summarize findings in written lab reports
- Compare and contrast data generated in the laboratory with that of relevant published research articles

UVic Grading Scheme

| | | | | | | | |
|----------------|---------|----------------|---------|----------------|---------|------|------|
| A ⁺ | 90 -100 | B ⁺ | 77 - 79 | C ⁺ | 65 - 69 | F | < 50 |
| A | 85 - 89 | B | 73 - 76 | C | 60 - 64 | N ** | < 50 |
| A ⁻ | 80 - 84 | B ⁻ | 70 - 72 | D | 50 - 59 | | |

**** N grades**

Students who have completed the following elements will be considered to have completed the course and will be assigned a final grade:

- *(list all components to be considered, e.g. assignments, midterms, final exam, etc.)*

Failure to complete one or more of these elements will result in a grade of "N" regardless of the cumulative percentage on other elements of the course. An N is a failing grade, and it factors into a student's GPA as 0. The maximum percentage that can accompany an N on a student's transcript is 49.

DEPARTMENT INFORMATION AND POLICIES

1. The Department of Biochemistry and Microbiology upholds and enforces the University's policies on academic integrity. These policies are described in the current University Calendar. All students are advised to read this section.
2. Cell phones, computers, and other electronic devices must be turned off at all times during live class sessions unless being used for the purpose of connecting and engaging with the class.
3. No recordings of live lectures are permitted without permission of the instructor. However, many courses will be recorded by the instructor for accessibility for students unable to attend. If you do not wish to be recorded, contact your instructor to determine if alternative arrangements can be made.
4. Students and instructors are expected to assess their health daily and avoid campus if they are ill.
5. Course materials, such as notes, problem sheets, quizzes, examinations, example sheets, or

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8. Deferral of a final exam must be requested with an Academic Concession form and submitted directly to Undergraduate Records. Deferred final exams for fall term courses will be arranged by the instructor. Deferred final exams or spring term courses will be arranged through Undergraduate Records and must be written before the end of the summer term as stipulated in the University Calendar.
9. Requests for review/remark of a midterm exam must be made within one week of the exam being returned.