

BIOC23H: Practical Approaches to Biochemistry Winter 2016

Description

A lecture and laboratory course that introduces students to experimental approaches used in biochemical research. Topics include practical and theoretical aspects of: spectrophotometry; chromatography, electrophoresis; radioisotopes; enzyme assay and protein purification. Students will be expected to solve numerical problems involving these and related procedures.

Prerequisites

Prerequisite: BIOB12H & BIOC12H Corequisite: BIOC13H

Lecture section

Monday, 12:00-14:00, SW319

Instructor

Dr. Rongmin Zhao, Office: SY248, Email: rzhao@utsc.utoronto.ca Please use E-mail ONLY when it is critical for you to get in touch with me and use BIOC23-Biochemistry in the subject line. Your student name and student number must be included in your email.

Instructor office hours

Monday 3:00pm-5:00pm

Laboratory teaching assistant

There is a teaching assistant for each lab section. Different TA may run the lab slightly differently and have different requirement for a particular lab. Students are required to follow his/her own TA's requirement and contact his/her own TA in case of any questions.

Course contents

Lectures --- BIOC23 is a laboratory course. The materials covered in the lecture will relate to the laboratory techniques carried out throughout the course. In the lecture, we will discuss methodologies, theories behind the methodology and relevant applications for the particular techniques used. Specific aspects of protocols and results of some labs will also be discussed. Lectures also give you a chance to ask questions. Therefore, attending lectures are highly recommended and please come with questions. Lecture notes will be posted before the lecture day.

There is no Textbook for this course. The lectures come from a number of sources including primary methodologies. The principles for some lab components have been discussed in the BIOC12 course.

Laboratories --- Attendance to labs is mandatory. If you miss a lab, you must provide a UTSC medical certificate or other acceptable reason cleared by Dr. Zhao to be excused from the lab and to be able to hand in any assignments related to that lab. There is no possibility to make up labs. Each missed lab will result in a loss of 4% of your grade. If you miss more than 3 laboratories without proper documentation, you will not be given credit for the lab component of the course.

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!!! Before coming to the laboratory, students are responsible for preparing the following items:

1. Lab coat (no exception) and closed toe shoes
2. Safety glasses for most labs
3. A permanent black marker (Sharpie fine point)
4. A book for recording your work (your lab log book). This book can be hard or soft cover, or a binder. This book has to be used only for the BIOC23 lab and will be collected at the end of the course.
5. A pen to record your lab and it is prohibited to use pencil to record your lab.

Supporting platform

We will use the UofT Blackboard system as the supporting platform. All course related materials, announcements will be posted in the Blackboard. You need to log in with your UTORid at the website <https://portal.utoronto.ca/webapps/portal/frameset.jsp>. You are responsible for checking the course website regularly for important information.

Laboratories are posted on the blackboard a week before the lab is scheduled. The emphasis of the course is the laboratory. The lecture complements the laboratory. **It is your responsibility** to come prepared each week to carry out your laboratory. To this end, before coming to the lab, you need to complete **an introduction** to each lab (a paragraph) explaining the purpose, and to write down your own **flow chart** based on the posted labs (this is what you should refer to when doing the lab). When carrying out experiments, you must take complete notes.

Therefore, as you do the experiment, you will write down in pen exactly what you did, what you have observed, such as times of incubations and any specific volumes, **AND ANY MISTAKE** you have done by accident.

You will have **data analysis** (including standard curve construction) **summary / conclusion**, where you analyze the data, do any graphs etc, which are required components of your lab assignment/report. To ensure you are prepared for the lab, there will be **quizzes** before each lab topic. The quiz will test whether you have adequately prepared for the lab and will be an essential component for the final grade.

Equipment in the lab is expensive. You are responsible for your equipment. Accidents can happen but most “accidents” are avoidable with proper preparation and attention to the task.

Course Evaluation

Midterm (15%)

It will test the lab and lecture materials. There is no makeup term test. If you miss the midterm because of illness and provide me with the UTSC medical certificate filled in by your doctor, you will be eligible to combine the term test mark to the final exam. You must contact me within 48 hours in case that you miss the term test. The format of the midterm will be multiple choice and short answer questions.

Final exam (35%)

It will test the lab and lecture materials. The format of the final exam will be multiple choice and short answer questions. The final exam is cumulative.

Quizzes 12% (6X 2%)

Date of quizzes: Weeks of Jan 11, Jan 18, Jan 25, Feb 1, Feb 8, March 7, 2016, essentially at the beginning of each lab module.

Lab performance (10%)

Preparation (introduction and flow chart) 4% - The TA is going to check the introduction and flow chart.

Laboratory log / summary/data analysis and technical performance 6% (4% will be particular on ion exchange and gel filtration chromatography sessions)

Lab assignments (28%)

Some lab reports will need to be uploaded to Turnitin: "Normally, students will be required to submit their course essays to Turnitin.com for a review of textual similarity and detection of possible plagiarism. In doing so, students will allow their essays to be included as source documents in the Turnitin.com reference database, where they will be used solely for the purpose of detecting plagiarism. The terms that apply to the University's use of the Turnitin.com service are described on the Turnitin.com web site". If you would like to opt out of Turnitin, you are required to submit a request to the instructor in writing by January 18 and be prepared to submit only the hard copies of your lab reports.

- 1. Mitochondrion isolation: (5%)** (Maximum of 2 pages of text double spaced, not including figures/figure legends, tables and references and cover page). This report will be submitted only as hard copy.
- 2. Alkaline phosphatase, (6%)** will not require materials and methods requires brief introduction, graphs/with appropriate legends and data analysis (maximum 2 pages of text double spaced, not including graphs, legends or references and cover page). This report will be submitted only as hard copy.
- 3. Formal report 1: IgG purification (8%)** (maximum 7 double spaced pages of text not including figures/figure legends/ reference page and cover page). This report will need to be submitted as hard copy AND uploaded to Turnitin.
- 4. Formal report 2: GST-fusion protein isolation and Western blotting (9%)** (maximum 7 pages of double spaced pages of text not including figures/figure legends/ reference page and cover page). This report will need to be submitted as hard copy AND uploaded to Turnitin.

* The class ID and password for log into Turnitin will be announced later in Blackboard. Please also visit the website <http://teaching.utoronto.ca/ed-tech/teaching-technology/turnitin/a-guide-for-students/> for instructions.

The content required for each assignment will be explained during the appropriate lecture and laboratory class. It is also addressed at the end of each laboratory manual.

Due dates:

The formal lab reports are due at the beginning of the lab indicated in the following weeks:

1. Mitochondrial isolation/differential centrifugation: week of Jan 18, 2016
2. Alkaline phosphatase: week of Feb 8, 2016
3. IgG purification: week of March 14, 2016
4. GST and Western blotting with lab logbook: week of March 28, 2016

Lecture schedule

Lectures relevant to the particular lab will be given either the week before or the week of the laboratory. Some topics will last more than one week.

- Topic 1: pH and buffers
- Topic 2: Cellular fractionation/
- Topic 3: Enzyme kinetics
- Topic 4: Protein isolation/purification
- Topic 5: Chromatography
- Topic 6: Protein quantification and gel electrophoresis
- Topic 7: Immunochemistry/Immunoblotting analysis
- Topic 8: Mass spectrometry and bioinformatics
- Topic 9: Use of radioisotopes in biochemical studies

Laboratory Safety

1. **Eating or drinking is prohibited in the laboratory.** Breaking this rule will lead to expulsion from the laboratory and loss of marks associated with the laboratory for which you have been asked to leave
2. **Please do not bring** coats, hats, etc. into the laboratory.
3. **You must Always wear a lab coat** (done up) in the laboratory with the sleeves rolled down and **closed shoes**
4. Wear gloves or safety glasses when instructed to do so.
5. Keep paper, pencils, fingers, etc. out of your mouth.
6. At the beginning and end of the laboratory, follow procedure 12 and 13
7. Discard chemical waste in appropriate containers
8. Discard bacterial culture material in to autoclave bags
9. Discard pipettes point-down, in the upright plastic pipet holders. Make sure the pipet tips are covered with disinfectant.
10. Place all *test tubes* containing living cells in the designated racks/pans;
11. Discard pipette tips in the appropriately marked container
12. **Wash hands thoroughly with soap and water** once or twice during the lab, at any time you come in contact with 1 chemicals/live cells and also just before leaving the laboratory.
13. When leaving the lab, REMOVE YOUR LAB COAT. However, DO NOT UNDER ANY CIRCUMSTANCES wear your lab coat in any public area (hall way and washroom etc.)

Laboratory schedule

(Monday of the week is indicated)

Jan 4, 2016: Week 1

Find groups and lab benches. Lab safety issues. NO real lab components.

Jan 11, 2016: Week 2:

Experiment 1: Differential centrifugation/mitochondrial isolation

Jan 18, 2016: Week 3

Differential centrifugation assignment due

Experiment 2: Enzyme kinetics: alkaline phosphatase

Jan 25, 2016: Week 4:

Experiment 2: Data analysis

Experiment 3: Ion exchange chromatography

Feb 1, 2016: Week 5

Finish experiment 3: Thin layer chromatography

Experiment 4: Gel filtration

Feb 8, 2016: Week 6:

Enzyme kinetics: alkaline phosphatase lab report due

Experiment 5 part 1: IgG: salting out via ammonium sulfate/dialysis

Feb 15, 2016 reading week - no lab

Feb 22, 2016: Week 7:

Experiment 5 part 2: IgG: DEAE Sephadex and protein quantification

Mar 29, 2016: Week 8

Experiment 5 part 3: IgG: SDS gel electrophoresis and staining (**picture posted by the following day**).

Mar 7, 2016: Week 9

Experiment 6 part 1: Purification of His₆-GST fusion proteins from *E. coli* (isolation and affinity chromatography)

Experiment 5 discussion of IgG data

Mar 14, 2016: Week 10

IgG report due

Experiment 6 part 2: His6-GST fusion protein: gel electrophoresis and Western transfer

Mar 21, 2016: week 11

Experiment 6 part 3: His6-GST fusion protein immunoblotting with anti-GST antibody/dark room

March 28, 2016: Week 12

GST lab report due along with your note books

Information Regarding AccessAbility Services at UTSC

Students with diverse learning styles and needs are welcome in this course. In particular, if you have a disability/health consideration that may require accommodations, please feel free to contact with me and/or the AccessAbility Services Office as soon as possible. The UTSC AccessAbility Services staff (located in Room SW-302) are available by appointment to assess specific needs, provide referrals and arrange appropriate accommodations. You can contact AccessAbility Services at 416-287-7560 or ability@utsc.utoronto.ca.

Academic Integrity

Academic integrity is essential to the pursuit of learning and scholarship in a university, and to ensuring that a degree from the University of Toronto is a strong signal of each student's individual academic achievement. As a result, the University treats cases of cheating and plagiarism very

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seriously. The University of Toronto's Code of Behaviour on Academic Matters (<http://www.governingcouncil.utoronto.ca/policies/behaveac.htm>) outlines the behaviours that constitute academic dishonesty and the processes for addressing academic offences. Potential offences include, but are not limited to:

- **IN PAPERS AND ASSIGNMENTS:** Using someone else's ideas or words without appropriate acknowledgement; submitting your own work in more than one course without the permission of the instructor; making up sources or facts. Obtaining or providing unauthorized assistance on any assignment.
- **ON TESTS AND EXAMS:** Using or possessing unauthorized aids; looking at someone else's answers during an exam or test; misrepresenting your identity.
- **IN ACADEMIC WORK:** Falsifying institutional documents or grades; Falsifying or altering any documentation required by the University, including (but not limited to) doctor's notes.

All suspected cases of academic dishonesty will be investigated following procedures outlined in the Code of Behaviour on Academic Matters. If you have questions or concerns about what constitutes appropriate academic behaviour or appropriate research and citation methods, you are expected to seek out additional information on academic integrity from your instructor or from other institutional resources (see <http://www.utoronto.ca/academicintegrity/resourcesforstudents.html>).