BIOL/CHEM 4XX Advanced Laboratory in Cell and Molecular Biology
3 Credits

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Time, days: TBA

Teaching Assistants: TBA

Pre-requisite: BIOL/CHEM 360 Cell and Molecular Biology (or equivalent; may be taken concurrently)

Required items: *Essential Cell Biology* (3rd or 4th edition) by Alberts *et al*. Garland Publishing, 3-ring binder for lab manual. The manual will be available for purchase in the Biology & Wildlife office at the start of the semester.

Course description: An advanced laboratory in cell and molecular biology. Students will learn modern molecular biological techniques including, protein and nucleic acid gel electrophoresis, western blotting, cell fractionation, cellular respiration, enzymology and fluorescence microscopy. Lectures will be supplemented with reading from the primary literature.

Course goals: Students will master lab techniques commonly used in cell and molecular biology, learn how to write a scientific paper, sharpen critical thinking skills, and practice working with others to solve problems. The focus of the laboratory will be on the physiological, biochemical and molecular responses to exercise, using rats as a model organism. A central goal of this course is to prepare students for working independently in a cell, molecular biology or biochemistry laboratory.

Student learning outcomes: Students will design and conduct experiments to determine the impacts of exercise on cellular metabolism, gene expression and oxidative stress. Students will become proficient in quantifying protein concentration, measuring maximal activity of key metabolic enzymes, quantifying changes in gene expression (including isolating and quantifying RNA, and designing gene-specific primers), quantifying DNA damage caused by oxidative stress, and measuring mitochondrial respiration rates. Students will become proficient in writing a scientific paper. Students will also become proficient in searching, reading and discussing primary literature.

Instructional methods: This course will be taught through a combination of lectures, laboratories, and discussions of the primary literature. The laboratories will be centered around understanding how muscles are remodeled in response to exercise. We will examine how protein and gene expression changes, how mitochondrial function changes, and how

parameters of oxidative stress change in response to exercise in rates. Prior to each laboratory, we will read and discuss 1-2 articles from the scientific literature in which similar techniques have been used to address similar questions. Two students will be assigned to lead the discussion on a paper each week.

Policies: Students are expected to attend class and complete reading assignments prior to coming to class in preparation for group discussions and/or activities.

Lab assignments are due at the start of each lab period. No late assignments will be accepted unless you have a medical excuse and a doctor's note, explaining your illness. You must attend the lab to earn credit for the assignment. There will be no make-up labs.

Exams: Exams will be based on material covered in both the lecture and lab. If you anticipate missing an exam for family or work commitments, please let us know in advance so that we can make other arrangements. If you must miss an exam because of unexpected, extenuating circumstances (ie; family death, medical excuse) then you must contact me as soon as possible.

Blackboard: Blackboard will be used to post grades, announcements, lab materials, and reading assignments. Please check the Blackboard site on a regular basis. If you have a smartphone, you can download the Blackboard App, which will notify you immediately when new announcements have been posted.

Email etiquette: We will use UAF email accounts to contact students. Please check your UAF account on a regular basis. If you use an alternate account, please have your UAF email forwarded to that account. We will do our best to respond to your email inquiries within 24 hrs. Please be considerate in your letters and use proper English grammar. Think before you send and never write anything you would feel uncomfortable saying to us (or anyone else!) in person. Please use a greeting and sign your letter; addresses don't always reveal the identity of the writer.

Disabilities: Please let us know if you have a disability. We will work with the Office of Disabilities Service (203 WHIT, 474-7043) to provide accommodations in both the classroom and laboratory to provide equal access to all materials in this course to all students.

Support services: Writing Center 801 Gruening

Grading: Your final grades will be based on the following:

1. **Exams (200 pts)**: There will be two exams during the semester (a mid-term and final), each worth 100 points. The purpose of these exams is to assess your understanding of the material, interpret data from the primary literature, and to develop your written communication and critical thinking skills.

2. **Laboratory assignments (425 points):**

a. **Short assignments (140 pts).** A short assignment, worth 10 points (140 points total), will be given following each laboratory. Some of the questions will cover material in the lab for the following week. It is extremely important for you to read the lab before coming to lab. Many of the labs we will do are complex. You will enjoy the lab more, understand it better, and are more likely to obtain good results if you come prepared. Please answer your homework questions using complete sentences and neat handwriting. Points will be deducted for incomplete sentences, misspellings, poor grammar and illegible writing.

b. **Lab notebook (60 pts).** Each student will maintain a lab notebook. These will be checked 3 times during the semester for completeness. The first notebook check will be work 10 pts, the second, 20 pts, and the third, 30 pts (60 pts total). Requirements for the lab notebook will be explained during the first lab.

c. **Lab reports (225 pts).** The labs in this course build upon one another to assess how muscle becomes remodeled in response to exercise. You will write one complete lab report, detailing the results from all experiments that will be due at the end of the semester. Following each set of labs (ie; proteomics, enzymology, etc.), you will write a mini-lab report that will include figures, figure legends and a summary of the results and appropriate statistical analyses. Each of these mini-lab reports will be worth 25 pts (125 pts total). Your final lab report, which will include all figures, figure legends, results, a title, abstract, introduction and discussion, will be worth 100 pts.

3. **Class discussions (75 pts).** You will be tasked with leading the discussion on a scientific paper at least once during the semester and will be required to participate in discussions.

In summary your grade will be based on the following:

ASSIGNMENT	**POINTS**
Exams:	2 X 100 = 200
Short lab assignments	140
Lab notebook	60
Lab report	225
Class discussion	75
	700 points total

Final grades will be calculated based on the percentage of points earned out of the total as follows:

Grade	% of Total Points
A+	97-100
A	90-96
A-	88-89
B+	86-87
B	80-85
B-	78-79
C+	76-77
C	70-75
C-	68-69
D+	66-67
D	60-65
D-	58-59
F	0-57

LABORATORY AND LECTURE SCHEDULE

WEEK OF:	TOPIC :
Week 1	Experimental design
Week 2	Lab 1 Proteomics part 1: protein assay Lecture: Introduction to exercise physiology
Week 3	Lab 1 Proteomics part 2: protein gel electrophoresis Lecture/ Discussion: Egan and Zierath (2013) Exercise metabolism and the molecular regulation of skeletal muscle adaptation. *Cell Metabolism* 17: 162-184.
Week 4	Lab 1 Proteomics part 3: western blotting- quantify myoglobin levels Lecture: Structure and function of oxygen-binding proteins
Week 5	Lab 2 Enzymology- maximal activity of citrate synthase and catalase Lecture: Cellular respiration and oxidative stress I
Week 6	Lab 3 DNA damage/repair part 1: Comet assay 1, Running experiment Lecture/Discussion: Cellular respiration and oxidative stress II Powers et al., (2011). Exercise-induced oxidative stress in humans: Cause and consequences. *Free Rad Biol Med* 51: 942-950.
Week 7	Lab 3 DNA damage/repair part 2: Comet assay 2, Image acquisition and analysis. Lecture/Discussion: DNA stability and health, DNA repair and cancer; Cha (2013) The accumulation of DNA repair defects is the molecular origin of carcinogenesis.
Week 8	Lab 3 DNA damage/repair part 3: Comet assay 3, Image acquisition and analysis. Lecture/Discussion: DNA repair and aging process. Vijg (2014) Somatic mutations, genome mosaicism, cancer and aging.
Week 9	Lab 3 DNA damage/repair part 3: Comet assay 4, Final analysis. **Mid-term exam**
Week 10	Lab 4 Gene Expression part 1: primer design & isolate RNA Lecture: Regulation of gene expression

Week 11	Lab 4 Gene Expression part 2: agarose gel electrophoresis Discussion: Exercise and the brain. Wrann et al. (2013) Exercise induces hippocampal BDNF through a PGC-1α/FNDC5 pathway. *Cell Metab* 18: 649-59.
Week 12	Lab 4 Gene Expression part 3: synthesize cDNA Lecture: Epigenetics
Week 13	Lab 4 Gene Expression part 4: quantitative real-time PCR Discussion: Rönn et al., (2013). A six months exercise intervention influences the genome-wide methylation pattern in human adipose tissue. *PLOS Genetics* 9 (6):e1003572.
Week 14	Lab 5 Cellular respiration part 1: cell fractionation Discussion: Menshikova et al., (2006). Effects of exercise on mitochondrial content and function in aging human skeletal muscle. *J Gerontol A Biol Sci Med Sci* 61: 534-40.
Week 15	Lab 5 Cellular respiration part 2: cellular respiration
Week 16	**Final exam**