

**BB 315/BI 315: Molecular Biology Laboratory,
A WRITING INTENSIVE COURSE (WIC)**

Spring 2016 3 credits

Students will participate in one Lecture/Discussion Forum session and two 3-hour lab sessions each week:

Lecture: Tuesday 4:00-4:50 (ASL 2018)

Lab Sections (12 students): Tuesday and Thursday 1-3:50, ALS 0023

Prerequisites: BB 314, BI 314, BB 314H, BI 314H, or by instructor permission.

Instructor:

Dr. Kari van Zee, Biochemistry-Biophysics Department ALS 2141
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Office hours: MW 11am-12pm ALS 2141 or by appointment

TA:

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Office hours: T 10 am-12pm ALS 2071 or by appointment

Course Information:

Molecular Biology Laboratory, BB 315/BI 315 (WIC) is an intermediate-level laboratory and writing intensive course designed for life science majors. By completing guided-research projects focusing on fundamental molecular biology concepts and essential technologies, participating students will explore the functional relationship between DNA sequence and gene products and the transmission of genetic information from storage through expression to function. Through laboratory projects, lectures, and journal-club style discussion forums of selected primary research papers, the course will introduce students to the use of recombinant proteins and how they are used in advancing the field of molecular biology and biochemistry. Students will attend two three-hour lab sessions each week and one lecture/discussion forum.

This course is designed to expose students to how research is performed, presented, and analyzed in the academic world. The class will collaborate with the Johnson lab in the BB department to investigate the role of two muscle proteins, myoferlin and dysferlin, in membrane trafficking events. Students will then take their results and

produce a manuscript that is written to the standards and format of a real research publication.

Learning Outcomes

Students will:

1. Recall and relate foundational molecular biology concepts and laboratory techniques to recent advances in basic research, medicine, and industrial applications.
2. Design experiments including the proper controls to analyze gene expression, construct, express, and characterize recombinant proteins.
3. Compare different types of protein expression and purification methods, and identify strengths and limitations of each method.
4. Demonstrate quantitative skills by preparing accurately and reproducibly reagents and solutions for experiments.
5. Operate safely molecular biology laboratory equipment including micropipettes, thermocyclers, centrifuges, gel electrophoresis chambers, power supplies, incubators, and autoclaves.
6. Interpret and evaluate scientific papers related to the research project, analyzing both scientific methods as well as writing style
7. Develop an awareness of the major issues at the forefront of the discipline and discuss ethical issues in the molecular life sciences.
8. Communicate and present their work to both a science literate and science non-literate audience through formal and informal writing projects, including a manuscript presenting their data from lab.

WIC Learning Outcomes

Students will:

1. Develop and articulate content knowledge and critical thinking in molecular biology through frequent practice of informal and formal writing.
2. Demonstrate knowledge of audience expectations, genres, and conventions appropriate to communicating in the discipline of molecular biology.
3. Demonstrate the ability to compose a document of at least 2000 words through multiple aspects of writing, including brainstorming, drafting, using sources appropriately, and revising comprehensively after receiving feedback on a draft.

Advice

This course is a writing intensive class as well as a laboratory class. This means that it will be time consuming and labor intensive. Our goals for this class are high, and we

commit to working closely with each and every one of you, to produce publication quality writing. Through the many review and workshop processes, the quality of your writing will improve with each step. The quality and therefore grade of your final manuscript will be directly related to how well you prepare for and utilize these review and workshop processes.

Lab Project: Molecular characterization of C2 domains from the human dysferlin and myoferlin proteins. Developed in partnership with Dr. Colin Johnson and Chelsea Holman (PhD Candidate), Department of Biochemistry and Biophysics.

Myoferlin and Dysferlin are two proteins known to be crucial in muscle development and repair. The C2 domains found in these proteins are the structural units responsible for this function. This project aims to find a deeper understanding of which amino acids are most crucial in the functionality of these C2 domains. Using recombinant cloning techniques in *E. coli*, students will create individual point mutations of a C2 domain-MBP fusion protein. Molecular biology techniques required will be plasmid DNA isolation, PCR mutagenesis, bacterial transformation, expression of recombinant protein, protein purification, SDS-polyacrylamide gel electrophoresis. After expression and purification, these proteins will be characterized for their ability to aggregate membrane vesicles. The change in aggregation ability will demonstrate the importance of specific amino acids in the binding surface of the protein to membranes.

Resources: Experimental protocols, reading lists, and resources will be posted on Canvas.

Readings

Abdullah, Nazish, Muruges Padmanarayana, Naomi J. Marty, and Colin P. Johnson. "Quantitation of the Calcium and Membrane Binding Properties of the C2 Domains of Dysferlin." *Biophysical Journal* 106.2 (2014): 382-89.

Fuson, Kerry. "Alternate Splicing of Dysferlin C2A Confers Ca²⁺-Dependent and Ca²⁺-Independent Binding for Membrane Repair." *Biophysical Journal* 106.2 (2014).

Jiménez, José L., and Rumaisa Bashir. "In Silico Functional and Structural Characterisation of Ferlin Proteins by Mapping Disease-causing Mutations and Evolutionary Information onto Three-dimensional Models of Their C2 Domains." *Journal of the Neurological Sciences* 260.1-2 (2007): 114-23.

Lek, Angela, Monkol Lek, Kathryn N. North, and Sandra T. Cooper. "Phylogenetic Analysis of Ferlin Genes Reveals Ancient Eukaryotic Origins." *BMC Evol Biol BMC Evolutionary Biology* 10.1 (2010): 231.

Lek, Angela, Frances J. Evesson, R. Bryan Sutton, Kathryn N. North, and Sandra T. Cooper. "Ferlins: Regulators of Vesicle Fusion for Auditory Neurotransmission, Receptor Trafficking and Membrane Repair." *Traffic* 13.2 (2011): 185-94.

Marty, Naomi J., Chelsea L. Holman, Nazish Abdullah, and Colin P. Johnson. "The C2 Domains of Otoferlin, Dysferlin, and Myoferlin Alter the Packing of Lipid Bilayers." *Biochemistry* 52.33 (2013): 5585-592.

Evaluation of Student Performance will be based on the following components:

Lab Participation (10%): Students are expected to attend every lab session, observe safety policies while working in the lab, demonstrate proper care of equipment and reagents, and be responsible for moving their research project forward. Points will be deducted for students who fail to follow safety guidelines, leave messes behind for TAs or another group to clean-up, or who do not display proper lab etiquette.

Pre lab assignments (10%): Students are required to complete the pre-lab assignment on Canvas before attending the first lab of each week. These assignments will test your knowledge on the protocol that will be performed, as well as concepts that will be utilized in the lab.

Notebook (15%): Each student will maintain a scientific laboratory notebook recording experimental design notes, procedures, calculations, data, and conclusions. Guidelines for keeping a scientific notebook will be provided in class. Notebooks will be submitted at the end of the term for grading.

Manuscript drafts and final copy (40%):

- Manuscript draft 1 (10%)
- Manuscript draft 2 (10%)
- Final Manuscript (20%)

Each student will submit two drafts and a final manuscript based on the experimental data they collect and analyze during the term. The reports will be in the format of a scientific paper with the second draft building on the first. Students will be expected to identify and read scientific literature relevant to the project, including peer-reviewed articles and reviews from leading molecular biology journals. Students will integrate and cite these outside sources in their writing to provide background information for the project and compare and contrast their findings to the existing scientific literature.

Students will receive instructor feedback on drafts of components (Abstract, Methods, Introduction, Results and Discussion, Conclusions, and References) of both reports and are expected to incorporate this feedback as revisions in the subsequent draft or final submission. Draft 1 should have a minimum word count of 1,000 words, Draft 2 2,000

words, and the final submission 2,750 words. Deadlines for submission of drafts and the revised final report will be announced in class at the beginning of the term and posted on Canvas. Late submissions will be penalized 20% for each day beyond the posted submission.

Peer Review (10%):

For each of the two drafts, you will be reviewing a peer's manuscript. Using the provided rubric and guidelines, you are expected to provide quality feedback on both experimental methodology as well as writing style and language.

Journal Club (10%):

An important component of the "practice of everyday science" is reading scientific articles published in peer-reviewed journals. The goals of journal club in this course are to help you 1) develop skills in critical thinking, 2) practice scientific journal article-reading and learn how to extract information from a scientific paper, 3) make connections between the research presented in an article and its significance in the broader world, and 4) have fun learning about research advances in a variety of fields.

Depending on class size, teams of 3-4 students will be formed at the beginning of the term and assigned a date in which to lead a journal club style discussion forum. All students are expected to read all of the articles and resources and participate actively in the discussions. To prepare for the journal club discussions, each student will need to complete a journal club discussion guide ahead of the discussion and post this to Canvas before the discussion. Students must attend the journal club discussion session in order to receive points for the submitted journal club discussion guide

All members of the presenting team are responsible for working together to select with instructor's approval a primary article relevant to the experimental project underway and published within the last 5 years from a leading, peer reviewed journal. Each team should also select 1-2 additional resources such as a review, newspaper article, animation, Ted Talk, that will help us all better understand the research and its broader implications.

The presenting team should read the article, 1-2 pertinent background papers, and resources well enough to walk/guide the group through highlights of the paper, explain the paper's data and figures, and be well-versed in any necessary background knowledge and/or supplemental info.

Research Paper Critique (5%): Each student will write a critical analysis of one of several recent molecular and cellular biology research papers selected by the instructor from leading scientific journals. The analysis should summarize the main findings of the paper, highlight relevant background literature that paved the way for the experimental questions proposed by the authors, describe the model and experimental strategies used by the authors, critically evaluate the results provided in the paper, and discuss

the significance of these results to advancing this area of research. Specific guidelines and links to the papers will be provided early in the term on the course web portal. The evaluation should be 1000-1500 words, 1.5-spaced, Times New Roman 11 pt. font. References to other scientific articles should be properly cited.

University Policies – A reminder:

Statement Regarding Students with Disabilities

Oregon State University is committed to student success; however, we do not require students to use accommodations nor will we provide them unless they are requested by the student. The student, as a legal adult, is responsible to request appropriate accommodations. The student must take the lead in applying to Disability Access Services (DAS) and submit requests for accommodations each term through DAS Online. OSU students apply to DAS and request accommodations at our [Getting Started with DAS](#) page.

The University student conduct code can be found at:

<http://oregonstate.edu/studentconduct/offenses-0>

Cheating or plagiarism by students is subject to the disciplinary process outlined in the Student Conduct Regulations. Students are expected to be honest and ethical in their academic work. Academic dishonesty is defined as an intentional act of deception in one of the following areas:

- ◆ Cheating-use or attempted use of unauthorized materials, information or study aids
- ◆ Fabrication-falsification or invention of any information
- ◆ Assisting-helping another commit an act of academic dishonesty
- ◆ Tampering-altering or interfering with evaluation instruments and documents
- ◆ Plagiarism-representing the words or ideas of another person as one's own

Behaviors disruptive to the learning environment will not be tolerated and will be referred to the Office of Student Conduct for disciplinary action.

Use of cellular phones is not permitted in the classroom or laboratory during lectures or exams.

“The goal of Oregon State University is to provide students with the knowledge, skill and wisdom they need to contribute to society. Our rules are formulated to guarantee each student’s freedom to learn and to protect the fundamental rights of others. People must treat each other with dignity and respect in order for scholarship to thrive. Behaviors that are disruptive to teaching and learning will not be tolerated, and will be referred to the Student Conduct Program for disciplinary action. Behaviors that create a hostile, offensive or intimidating environment based on gender, race, ethnicity, color, religion, age, disability, marital status or sexual orientation will be referred to the Affirmative Action Office.”

BB 315/ BI 315 Weekly Schedule

Note: This schedule should be considered a guide and is subject to change depending on experimental progress.

Week	Tuesday	Lecture	Thursday	Readings/ Assignments
1	Navigating in a Molecular Biology Lab---Laboratory Safety and Equipment Orientation. Using Micropipets. Preparing media.	Foundations of Molecular Biology: Central Dogma and Scientific Practices	Presentation by Dr. Colin Johnson, on the importance of Myoferlin and Dysferlin. Introduction to Protein Data Bank and Pymol.	Prelab for Thursday Read Lek et al. 2012 for Thursday
2	Using protein modeling to choose point mutation. Purifying plasmid from <i>E. coli</i> .	Primer design for quick change PCR.	Agarose gel electrophoresis, preparing LB+agar selection plates.	Read Jimenez, J. Bashir, R. 2007 for Tuesday Prelab due Thursday (including primers)
3	Quick change PCR for site-directed mutagenesis; set up overnights for Wt and MBP	Methods of cloning and understanding plasmid features	DpnI treatment and transformation of mutated plasmid into <i>E.coli</i> ; induce and pellet Wt and MBP	Prelab due Tuesday Prelab due Thursday Critique of Paper due Friday
4	Miniprep mutant plasmids and submit for sequencing; make purification solutions	Bioinformatics and DNA sequencing	Pour and run SDS-PAGE gel of Wt and MBP expression	Prelab due Tuesday Prelab due Thursday
5	Purify and desalt Wt and MBP	Protein Purification Methods	Run gel on purified protein, quantitate by Bradford assay, dilute and freeze.	Prelab due Tuesday Prelab due Thursday Draft 1 due Friday
6	Run aggregation assay with Wt and MBP; express mutants and Wt	Other cloning methods	finish running aggregation assay on Wt and MBP	Prelab due Tuesday Peer review due Thursday
7	Purify mutants and Wt	Journal Club -team 1	Run gel, quantitate by Bradford assay; freeze	Prelab due Tuesday
8	Assay mutants and controls again	Journal Club – team 2	Repeat expressions, purifications, gels as needed	Prelab due Tuesday
9	ITC; draft 2 writing workshop	Journal Club- Team 3	ITC; Final lab meeting with Dr. Johnson	Draft 2 due Monday Prelab due Tuesday
10	finish any final aggregation assays	Journal Club-Team 4	Laboratory Clean-up	Prelab due Tuesday
finals				Final version of scientific manuscript and lab notebooks due Monday at 4 pm