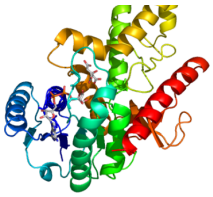
Techniques in Molecular Biology

BIOL/CHEM 330 Lecture & Lab
Fall 2017



Course Overview: Students will learn and apply molecular biology techniques (focused on nucleic acids) in the laboratory. Students will learn principles and practice of basic bacterial culture techniques, transformation, agarose gel electrophoresis, nucleic acid purification (plasmid and genomic DNA, RNA), nucleic acid quantification, DNA restriction digestion and analysis, polymerase chain reaction (PCR), and basics of computer-based DNA sequence analysis and planning of cloning strategies. In addition, students will learn about the nature and selection of DNA cloning vectors (bacterial & eukaryotic), restriction enzymes, modifying enzymes, polymerases, and other reagents used in molecular biology. We will examine aspects of bioinformatics and genomics, and newer/advanced molecular technologies such as next-generation sequencing and microarrays that are especially important in genomics, and discuss the burgeoning other '-omics' fields. Some primary literature articles illustrating standard techniques will be studied. We will apply our newly learned molecular techniques toward solving real biological research questions, and to carry out a basic DNA cloning project.

At the end of Biology/Chemistry 330, a student should be able to:

* List and explain safety issues and proper practices associated with standard molecular techniques, including bacterial culture, electrophoresis, and nucleic acid purification, detection and quantification.
* Explain, demonstrate and practice principles of sterile technique, bacterial culture, transformation, and DNA and RNA purification and quantification.
* Understand the nature of molecular biological hypothesis and testing – how molecular (genetic) analysis answers scientific questions.
* Use and explain the application of various standard bioinformatic techniques to experimental planning and analysis, including sequence accessing and manipulation, BLAST, multiple sequence alignment, PCR primer design, cloning strategies, etc.
* Explain and discuss the modern role of genomics (comparative, functional, etc.) and other '-omics' in molecular analysis.
* Understand and explain the many variations on PCR and when to use them, and how to troubleshoot a PCR protocol by selecting parameters to vary.
* Plan, execute and document a basic DNA cloning project involving PCR amplification, cloning into an appropriate DNA vector, transformation, plasmid DNA isolation, followed by restriction enzyme analysis with agarose gel electrophoresis, DNA sequencing, and sequence analysis to evaluate success of the procedure.

Work outside of the regular lab period

The practice of molecular biology cannot easily be restricted to a single four-hour lab period. It is unavoidable that students will be required on many occasions either to come to the lab briefly the day before lab to set up something for the next day, or to return to the laboratory the day following a particular experiment to examine their results.

Book *From Genes to Genomes. Concepts and Applications of DNA Technology 3rd Ed. Dale, Von Schantz and Plant. Wiley-Blackwell ISBN 978-0-470-68385-9*

Electronic Notebook (ELN) *$15. We will set up your account on the first day of lab. You will have three weeks to use the service before you will be locked out of your notebook if you haven't paid. Bring a laptop or tablet to class. https::://mynotebook.labarchives.com/*

Grading: A 100-90%, B 80-89% C 70-79% D 60-69% F 59-0%

Points	Description
150	End of Semester Lab Practical
400	Take Home Exams (two at 200 points each)
150	Lab Notebook (50 points x three reviews)
200-300	Lab Results (50 pts x each)
50	SOP Assignment
150	Cloning Assignments I & II and Serial Cloner Assignment (50 pts each)
50	Bioinformatics Assignment
100	Project Plan/Results Presentation (50 pts each)
50	Unannounced Lab Notebook Checks 5 x 10 points

Tuesday Lab 8:00 – 12:00 (Lab)		Thursday Lecture/Lab 10:45 – 12:05 (ST 130)	
		Sept 7	**Block 1 Lect:** Intro, lab safety, electronic notebooks, solutions, Lab math, intro to sterile and bacteriological tech
Sept 12	**Block 1 Lab 1:** *Pipetting and Culture Tech*. Discussion: Pipetting, bact culturing / media prep and autoclave training	Sept 14	**Block 1 Lect:** bacterial strains, antibiotics and transformation, Plasmid purification,
Sept 19	**Block 1 Lab 2:** *Transformation* Discussion: Bacterial Plasmids Promoters, DNA electrophoresis, DNA/RNA quantification	Sept 21	**Block 1 Lect:** SOP/QA/QC, working in industry
Sept 26	**Block 1 Lab 3:** *Plasmid Purification & quantification*	Sept 28	**Block 1 Lect:** Restriction digest, intro to cloning and subcloning.
Oct 3	**Block 1 Lab 4:** *Agarose Gel & Restriction Digest*	Oct 5	**Block 1 Lect:** In class project. Cloning Assignment I and II.
Oct 10	**Block 2 Lab 1***: Bioinformatics and sequence analysis workshop*	Oct 12	**Block 2 Lect:** Next-generation and advanced high throughput sequencing.
Oct 17	**Block 2 Lab 2:** *Trip to biotech/pharma company*	Oct 19	**Block 2 Lect:** Basic and Advanced PCR theory and techniques, primer design, site directed mutagenesis, sequence insertion and qPCR
Oct 24	**Block 2 Lab 3:** Continued - Advanced PCR theory and techniques, primer design, site directed mutagenesis, sequence insertion and qPCR	Oct 26	**Block 2 Lect:** *SNP analysis of human tissues*
Oct 31	**Block 2 Lab**:*SNP analysis of human tissues*	Nov 2	**Block 2 Lab**: *SNP Analysis and rtPCR Prep*
Nov 7	**Block 2 Lab:** *rtPCR expression of NHE1*	Nov 9	**Block 3 Lect:** Basic Cloning Techniques
Nov 14	**Block 3:** Advanced Cloning Techniques	Nov 16	**Block 3:** Serial Cloner Software and Cloning Project Design/Introduction
Nov 21	**Block 3:** Cloning Project Presentation and Start Project	Nov 23	*Thanksgiving*
Nov 28	**Block 4:** Cloning Project	Nov 30	**Block 4:** Cloning Project
Dec 5	**Block 4:** Cloning Project	Dec 7	**Block 4:** Cloning Project
Dec 12	**Block 4:** Cloning Project	Dec 14	**Block 4:** Cloning Project Results Presentation
Final Lab Practical Dec 18, 8:00-10:00 & 11:00-1:00			

Deadlines - The timing in this document are guidelines. Some experiments may take more time, others less. Due dates for assignments will be provided in class. The web will have target dates for all assignments as well as a description for each homework assignment.

Each laboratory experiment - from simple DNA gel running to the more project oriented experiments MUST be written into the lab book following the instructed format. There are no other formal or informal write up. The discussion section should cover the salient points and make recommendations based on your results.