

RECOMBINANT DNA TECHNIQUES

BI 410 and BI 510
MWF 11:30 -12:35
UTS Room 507

INSTRUCTOR	COURSE DESCRIPTION
Justin Courcelle 725-3866 justc@pdx.edu	A study of techniques and approaches used in DNA manipulation genetic engineering.

Textbook: The “Course Readings” (CR) packet is available at Smart Copy, 1915 SW 6th Ave. Additional readings will be handed out in class and/or posted on the course website.

Websites: Course homepage <http://web.pdx.edu/~justc/courses>

Office hours: M 1:00-3:00 SB2 Rm 432 or by appointment

Problem sets: 2 problem sets will be assigned and can be downloaded from the course website on the dates indicate in the syllabus. Homework will be graded, and points will be deducted if they are handed in late.

Exams: Exams will be in the form of short answer and multiple choice questions looking to determine your understanding of the material as well as your ability to interpret data

Grading:	<u>BI410</u>	<u>BI510</u>
Exam I	35%	25%
Exam II	35%	25%
Problem Sets	30%	25%
Presentation		25%

There are NO makeup exams. You must take both exams or you cannot earn a passing grade. If caught in an act of academic dishonesty, you will receive a zero for the assignment and be reported to student affairs.

If you are a student with a documented disability and registered at the Disability Resource Center, please contact me immediately to facilitate arranging academic accommodations.

Tentative Schedule

Week	Date	Topic	Course Readings
1	Jan 07	Overview, DNA manipulation and safety, intro to <i>Molecular Cloning</i> manual	1-3
	Jan 09	Visualization and detection of DNA, RNA, and protein	4-6
	Jan 11	Detection of DNA, RNA, and protein	7-9
2	Jan 14	Isolating DNA, isolating RNA	10-11
	Jan 16	PCR and its applications	12-13
	Jan 18	DNA sequencing—methodology	14
3	Jan 21	No Class	
	Jan 23	DNA sequencing—bioinformatics (PROB SET 1 HANDED OUT)	15
	Jan 25	Genomics and Proteomics: massively parallel measurements	15
4	Jan 28	Manipulating DNA—cutting and pasting	16-18
	Jan 30	Manipulating DNA—DNA enzymes and their utility (PROB SET 1 DUE)	19-20
	Feb 01	Mobilizing DNA: Plasmids and transformation	21-22
5	Feb 04	Mobilizing DNA: Phages, strain construction, large DNA	23-24
	Feb 06	Mobilizing DNA: Specialized vectors	25-27
	Feb 08	EXAM I	
6	Feb 11	Gene cloning: Genomic DNA and “library” construction	28-29
	Feb 13	Gene cloning: Screening for genes	29
	Feb 15	Mutagenesis, Protein engineering and altering the genetic code	30
7	Feb 18	Cloning in bacteria other than <i>E. coli</i>	
	Feb 20	Manipulation of bacterial genomes, genome shuffling	
	Feb 22	Metabolic engineering, development of antimicrobials	
8	Feb 25	Cloning in eukaryotes: 2-hybrid system (PROB SET 2 HANDED OUT)	31
	Feb 27	Transformation: higher eukaryotes, baculovirus expression	32
	Feb 29	Genetic manipulation of plant cells	
9	Mar 03	Genetic manipulation of animal cells (PROB SET 2 DUE)	
	Mar 05	Advances in transgenics	
	Mar 07	Embryonic stem cells and organismal cloning	
10	Mar 10	Medical diagnostics and development of therapeutics, gene therapy	
	Mar 12	Molecular anthropology/archaeology	
	Mar 14	DNA nanotechnology	
*	Mar 20	FINAL 12:30-2:20	

Readings and resources for Bi430/530 **(“MC” = Molecular Cloning)**

1. “Unnatural Selection” by Allison Snow
2. Contents of the “Molecular Cloning” (MC) lab manual: MC v–xx.
3. National Institutes of Health Directive on Recombinant DNA Safety
4. Quantitation of DNA: MC A8.19–A8.24
5. Electrophoresis: MC 5.2–5.17 (agarose), 5.40–5.42 (polyacrylamide), 8.40–8.49 (protein gels)
6. Visualizing DNA (and protein) in gels: MC A9.3–A9.8
7. Southern blots: MC 6.33–6.38
8. Northern blots: MC 7.21–7.26, 7.45
9. Western blots: MC A9.28, A8.52–A8.55
10. DNA purification: MC 6.61–6.62, 1.16–1.19, A8.9–A8.18
11. RNA purification: MC 7.2–7.5, 7.8, 7.13–7.14, 7.20, 7.30, 7.82–7.84
12. General PCR: MC 8.4–8.29 (introduction, basic method, troubleshooting, processing), 8.107–8.113 (PCR facts)
13. Specialized PCR: MC 8.66–8.68 (MOPAC), Quantitative-RT PCR review
14. DNA sequencing: MC 12.3–12.12 (introduction), 12.51–12.52 (cycle sequencing), 12.106 (sequencing PCR products), 12.94–12.100 (automated sequencing), The “\$1000 Genome”
15. Bioinformatics: MC A11.1–A11.2 (intro), A11.21–A11.24 (databases), DNA microarrays: MC A10.1–A10.19
16. Cloning DNA in plasmids: MC 1.19–1.24 (intro), 1.84–1.85 (directional cloning protocol), 1.90 (blunt-ended cloning), 1.93–1.95 (vector dephosphorylation)
17. Restriction/modification systems: MC A4.3–A4.9
18. DNA ligases: MC 1.157–1.159
19. Molecular biology buffers and reagents: MC A1.2–A1.12
20. Enzymes for DNA manipulation: MC A4.12–A4.16 (DNA polymerases), A4.24–A4.27 (reverse transcriptase, terminal transferase), A4.35–A4.37 (kinase, alkaline phosphatase), A4.40–A4.48 (nucleases)
21. Plasmids: MC 1.2–1.16 (plasmid biology), 1.24–1.29 (transformation with plasmids), 1.162 (electroporation), 1.149–1.150 (alpha complementation)
22. Antibiotic selection: MC 1.143–1.148
23. Bacteriophage lambda: MC 2.2–2.24 (lambda biology, lambda vectors, cloning in lambda), 2.110 (minimizing damage to large DNA molecules)
24. Bacteriophage M13: MC 3.1–3.11 (M13 biology, cloning in M13), 3.42–3.44 (phagemids)
25. High-capacity vectors (introduction): MC 4.1–4.10
26. Cosmids: MC 4.11–4.16
27. Expression of cloned genes in *E. coli*: MC 15.2–15.13
28. DNA libraries: MC 12.10–12.12 (random libraries), 11.1–11.36, 11.112, 11.107 (cDNA libraries)
29. Gene expression libraries: MC 14.1–14.3, 14.47–14.49
30. Mutagenesis: MC 13.2–13.10
31. Protein interaction technologies: MC 18.6–18.13
32. Introducing DNA into mammalian cells: MC 16.2–16.6, 16.47–16.53