### AGRO/HORT/BOTA 339:

# Introduction to the molecular techniques of plant biology and biotechnology

#### COURSE OBJECTIVE

To provide a practical experience in the major techniques used in plant genetic analysis at the molecular and bioinformatic levels.

#### **TEXTBOOK**

From Genes To Genomes: concepts and applications of DNA techonology. 3<sup>rd</sup> Edition. Jeremy W. Dale, Malcom Von Schantz, Nick Plant. Wiley-Blackwell. Dec. 2011. Required readings will be assigned from this textbook. Please bring your textbook to all lectures. A copy will also be on reserve at the library.

#### **PREREQUISITES**

Botany/Zoology 152 or equivalent Chemistry 104 or equivalent

CREDITS: 4CR

#### **INSTRUCTORS**

Professor TA (Lab Instructor)

Dr. Jean-Michel Ané Kevin Cope

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#### OFFICE HOURS

MF 1:30-2:30 and by appointment

#### **SCHEDULE**

LEC 11am-12:15 pm TR 136 Plant Sciences LAB1 1:20-4:20 pm W 136 Plant Sciences LAB2 1:20-4:20 pm R 136 Plant Sciences

SUPPLIES NEEDED: TEXTBOOK

## HANDOUTS: AVAILABLE ON LEARN@UW WEBSITE A FEW DAYS BEFORE THE LECTURE / LAB

#### HOW YOUR GRADE WILL BE DETERMINED

Mid-term exam 1 16% Mid-term exam 2 16%

Laboratory reports 40% (10 reports, 4% per report)

Presentation 12% Final exam 16%

#### LABORATORY EVALUATION

This module of the course is designed to familiarize each student with plant molecular biology techniques, ranging from crude DNA extraction to bioinformatics, and to hone writing skills along the way. Lab work is typically done in pairs; however, <u>assignments must be completed individually</u>.

Information on the lab reports is detailed below.

There will be 13 labs throughout the semester, 10 of which will require lab reports. For the reports, use a professional and impersonal writing style. All reports should be typed, between 2-3 pages long, and contain the following:

- *Title:* Describe the goals of the exercise without copying the lab hand out title.
- Introduction: Describe the purpose and application of the lab exercise.

  This section should be no more than three sentences.
- Materials and methods: Describe the materials for each part of the lab, and briefly (no more than *five* sentences) summarize the methods performed in each section.
- Results: This section should be well structured and organized, and include tables and graphs where appropriate. Just present the results; do not include any other information in this section. Conclusions and discussion about the results are reserved for the following section.
- Discussion: Discuss the results gathered in the lab exercise. Are the results expected? What interpretations can be made from the results? If the results are unexpected, interesting, or inconclusive in some way, discuss the potential reasons for arriving at them. This will be the longest section. Address ALL results and key points.

- *Conclusions:* Summarize results and discuss future opportunities for improvements or follow up experiments.
- Questions: Answer all the questions in the hand out. Credit is not awarded for incorrect answers. Be succinct.

Paper copies of all reports are due the week after each lab at the beginning of the lab session. Reports a day late will receive 50% credit and after that no credit is awarded. Reports should be double-spaced and maximum 12 point font (smaller fonts may be used, but make sure everything is clearly legible when printed). Handwritten reports <u>will not</u> be accepted. Labs will be graded based on completeness, inclusion of current resources, correct spelling, and grammar. Literature should be properly cited within the text and correspond to the Works Cited page. DO NOT PLAGARIZE. Proper discipline will be utilized, if necessary. Please spend sufficient time on these assignments, as they will make up a significant portion of your total laboratory evaluation. If you miss a lab, lab reports can be made up by writing a report for Lab 12 (see the course schedule).

#### **PRESENTATIONS**

At the end of the semester, each student will give a 10 minute presentation of a technical article to the class. This presentation will be followed by a 5 minute discussion with the class. The article and date of the presentation will be randomly assigned by the instructor.

#### POLICIES ON TESTING

All exams will be **closed book**. Tests will generally consist of short answer and essay questions and problems/calculations. Material covered in lectures, labs and assigned readings is considered fair game for tests. Tests outside of regularly-scheduled class period will not be given. Practice tests will be provided prior to each exam. Students are encouraged to form study groups; to facilitate this, the classroom where the lectures and lab sessions are held will be made available when needed.

#### FEEDBACK ON THE COURSE

Conversations about how to improve the course or any problems that you may have are welcome anytime.

Week	Topics	Readings/Assignments
1	LEC 1 Sept 2: Plant biotechnology Principles and Techniques: what, why, how	
	LEC 2 Sept 4: From DNA to proteins (Dr. Jayaraman)	Chapter 1
	LAB Sept 3 or 4: NO LAB	
2	LEC 3 Sept 9: Genes and expression (Dr. Jayaraman)	Chapter 1
	LEC 4 Sept 11: Genes and genomes (Dr. Jayaraman)	
	LAB 1 Sept 10 or 11: Lab safety, notebook, measurements, calculations, pipette exercise	
	and basic laboratory sterile techniques (No lab report evaluated)	
3	LEC 5 Sept 16: Recombinant DNA technology	
	LEC 6 Sept 18: The basics of cloning (Dr. Garcia)	Chapter 2.1-2.8
	LAB 2 Sept 17 or 18: Plant genomic DNA isolation and DNA quantification techniques	'
4	LEC 7 Sept 23: DNA sequencing and databases	
	LEC 8 Sept 25: Bioinformatics (Dr. Garcia)	Chapters 5.1-5.5, & 8.1-8. Lab 2 Report due
	LAB 3 Sept 24 or 25: Basic bacterial culture techniques (growth, selection, pure culture	
	isolation, cryostorage, inoculation of liquid media and plates). Plasmid DNA isolation	
5	LEC 9 Sept 30: Polymerase Chain Reaction (PCR) and applications	Chapter 4 & 6.1-6.3 Lab 3 Report due
•	LEC 10 Oct 2: Monitoring gene expression	
	LAB 4 Oct 1 or 2: DNA restriction analysis, cloning of restriction fragment and	
	transformation of $E$ . $coli$ agarose gel electrophoresis, sequence alignments	
6	LEC 11 Oct 7: Vectors and libraries (Midterm exam 1 covers up to here)	Chapter 3, & 2.9-2.13 Lab 4 Report due
	LEC 12 Oct 9: Next Generation Sequencing (NGS) and applications (Dr. Garcia)	
	LAB 5 Oct 8 or 9: Cloning techniques and bacterial transformation	
7	LEC Oct 14: MIDTERM EXAM 1 on Lectures 1 through 11	
,	LEC 13 Oct 16: Forward genetics (1/2)	NONE
	LAB 6 Oct 15 or 16: PCR and design of PCR primers	Lab 5 Report due
0	LEC 14 Oct 21: Forward genetics (2/2)	
9	LEC 14 Oct 21: Forward genetics (2/2) LEC 15 Oct 23: Reverse genetics (1/2)	Chapter 8.9 Lab 6 Report due
	LAB 7 Oct 22 or 23: RNA manipulation: purification, quantification	
9	LEC 16 Oct 28: Reverse genetics (2/2)	Chapter 7, 11.1, 11.5, 11.6 Lab 7 Report due
	LEC 17 Oct 30: Information from a cloned gene (Midterm exam 2 covers up to here)	
	LAB 8 Oct 29 or 30: RNA gels, analysis of gene expression by quantitative RT-PCR	
10	LEC 18 Nov 4: Protein expression, purification and analysis (1/3)	Chapter 7, 10 Lab 8 Report due
	LEC 19 Nov 6: Protein expression, purification and analysis (2/3)	
11	LAB 9 Nov 5 or Nov 6: Protein extraction, quantification and SDS-PAGE	'
	LEC 20 Nov 11: Protein expression, purification and analysis (3/3)	NONE Lab 9 Report due
	LEC Nov 13: MIDTERM EXAM 2 on Lectures 1 through 17	
	LAB 10 Nov 12 or 13: Western blots	
12	LEC 21 Nov 18: DNA fingerprinting (Article for presentation assigned)	Chapter 6.4, 10.4 Lab 10 Report due
	LEC 22 Nov 20: Genome mapping	Lab 10 Report due
	LAB 11 Nov 19 or 20: Reverse genetics	Lab 10 Report due
	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning	
	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)	Lab 10 Report due  Chapter 8.7, 9
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)	
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)  LEC 24 Dec 2: Student presentations (1-6)	Chapter 8.7, 9
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)  LEC 24 Dec 2: Student presentations (1-6)  LEC 25 Dec 4: Student presentations (7-12)	Chapter 8.7, 9
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)  LEC 24 Dec 2: Student presentations (1-6)  LEC 25 Dec 4: Student presentations (7-12)  LAB 12 Dec 3 or 4: Genetic mapping, proteomic and genomic tools (No lab report evaluated)	Chapter 8.7, 9
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)  LEC 24 Dec 2: Student presentations (1-6)  LEC 25 Dec 4: Student presentations (7-12)  LAB 12 Dec 3 or 4: Genetic mapping, proteomic and genomic tools (No lab report evaluated)  LEC 26 Dec 9: Student presentations (13-18)	Chapter 8.7, 9  NONE  Lab 11 Report due
13	LAB 11 Nov 19 or 20: Reverse genetics  LEC 23 Nov 25: Map-based cloning  LEC Nov 27: NO LECTURE (THANKSGIVING)  LAB NO LAB (THANKSGIVING)  LEC 24 Dec 2: Student presentations (1-6)  LEC 25 Dec 4: Student presentations (7-12)  LAB 12 Dec 3 or 4: Genetic mapping, proteomic and genomic tools (No lab report evaluated)	Chapter 8.7, 9