**COURSE NAME; NUMBER; SEMESTER; MEETING DAYS, TIMES, AND PLACE.**

Molecular Genetics Laboratory (4 credits)

11:126:482Spring 2022 (Index sec 1 - 12464; sec 2 - 12465; sec 3 - 12466)

**Monday 10:20-11:40 (lecture), and 2:00-7:00/10:20-11:40 on designated laboratory section days**

Location: Cook Campus, Lecture-Foran 138A; Laboratory-Foran 193

**CONTACT INFORMATION:**

Instructor: Dr. Faith Belanger

Office Location: 304 Foran Hall

Phone: 848 932-6389 Email: belanger@sebs.rutgers.edu

Instructor: Dr. Donald Kobayashi

Office Location: 263 Foran Hall

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Instructor: Dr. Ning Zhang

Office Location: 201 Foran Hall

Phone: 848 932-6348 Email: ningz@rutgers.edu

**COURSE MATERIALS:**

* Online (internet) access to course content through Canvas

**COURSE DESCRIPTION:**

Students are introduced to both chemical and transposon mutagenesis as approaches to gene identification in prokaryotic organisms. Students also investigate gene identification in bacteria by direct cloning for phenotype expression in heterologous hosts. Advantages and disadvantages of the various approaches are discussed. Students are also introduced to yeast as a model system to provide an introduction to laboratory methods used to investigate the genetics of eukaryotic organisms. The labs illustrate: 1) the use of genetic crosses to create individuals with particular genetic characteristics; 2) cloning a gene by complementation; and 3) deletion of a gene from the yeast chromosome.

**LEARNING GOALS:**

1. Mastery of basic methods and applications for molecular genetic studies, and comparing and contrasting these methods and applications between prokaryotic and eukaryotic systems
Assessment through exams, quizzes, oral presentations, and evaluation of lab performance
2. Understanding the purpose of appropriate and adequate controls and knowing how to establish these controls when designing experiments
Assessment through exams, quizzes, oral presentations and lab reports
3. Development of skills related to effective teamwork
Assessment of lab performance of the entire team and individual members of the team
4. Development of effective communication skills
Assessment of writing skills by short answer/essay questions on exams, lab reports and speaking skills by oral presentations
5. Mastery of the operation of standard equipment used in molecular biology laboratories
Assessment by evaluation of lab performance and lab reports
6. Ability to analyze data and present results effectively
Assessment by specific questions on exams, lab quizzes, lab reports and oral presentations
7. Development of organizational skills
Assessment by weekly review of flow charts written in lab notebooks, data entry in lab notebooks and evaluation of ability to conduct weekly experiments in a timely manner
8. Understanding the use of formulas for quantitative purposes in experimental procedures
Assessment by quizzes, exams and evaluations of lab performance during experimental procedures
9. Understanding hypothesis-driven experimentation
Assessment by exams, quizzes, lab reports, oral presentations, inspection of data recorded in lab notebook, and evaluation of lab performance

**ASSIGNMENTS/RESPONSIBILITIES & ASSESSMENT:**

**Course Grading**

 25%: Exam 1 (midterm)

            25%: Exam 2 (non-comprehensive final)

 25%: Written Laboratory Reports

 10%: Quizzes

            10%: Oral presentations

 5%: Lab notebook/attendance/lab participation and performance

            Total = 100%

**Learning Goal Assessment by evaluation of:**

1. Performance on specific questions exams
2. Performances on specific questions on quizzes
3. Performance on oral presentation

**ACCOMODATIONS for Students with disabilities**

Please follow the procedures outlined at <https://ods.rutgers.edu/students/registration-form>. Full policies and procedures are at <https://ods.rutgers.edu/>

**COURSE SCHEDULE:**

 **Week Topic\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ \_\_\_\_\_\_\_\_\_\_**

**Week 1** No Class

(Jan 18)

**Week 2** Laboratory 1 **P. 6 CANCELLED**

(Jan 24) **Growth curves of *Serratia marcescens***

Follow-up lab: count colonies on plates for growth curve

**Week 3** Laboratory 2 **P. 9**

(Jan 31) **Chemical mutagenesis**: generation of kill curves

Follow-up lab: count colonies on plates for kill curves

**Week 4** Laboratory 3 **P. 14**

(Feb 7) **Chemical mutagenesis (cont.)**:

 generation of NTG mutants for selection of *lipA* activity loss

dilution plating of mutagenized cells

**Transposon mutagenesis**:

mating of donor and recipient cells

plating of matings

Follow-up lab: Plating of matings 3,4 and 5 of transposon mutagenesis

**Week 5** Laboratory 4 **P. 21**

(Feb 14) **Chemical mutagenesis (cont.)**:

 Select bacterial colonies and replica-plate onto LB agar and LB agar supplemented with 1% Tween 80.

**Transposon mutagenesis (cont.)**:

Selection of mutants and replica plating

Follow-up lab: Observation of mutants and restreaking of *lipA* mutants

**Week 6** Laboratory 5 **P. 23**

(Feb 21) **Direct cloning**

 Isolation of plasmid DNA from lipase expressing clones

Follow-up lab: none

**Week 7** Laboratory 6 **P. 25**

 (Feb 28) **Direct cloning (cont.):**

 Restriction analysis of isolated plasmid DNA from lab 5

 Follow-up lab: none

**Week 8** **MIDTERM EXAM: Monday, Mar 7**

(Mar 7) First week of yeast labs (Belanger)

Yeast Genetics Laboratories

**INTRODUCTION**

 The yeast *Saccharomyces cerevisiae* (bakers yeast) is a single‑celled eukaryotic organism that is widely used in molecular genetics studies. During the last half of the course we will be using yeast to illustrate some of the principles of molecular genetics. We will be carrying out three distinct projects. These projects will be carried out simultaneously.

 1) Reassortment of yeast mutant genes

 Goal: Crossing yeast mutant strains to illustrate complementation,

segregation of alleles, and independent assortment of chromosomes.

 2) Cloning a higher plant gene using yeast

 Goal: To clone a higher plant gene by complementing a yeast mutant strain.

 3) Deleting a gene from the yeast chromosome

Goal: To create a mutation in a specific yeast gene by deleting it from the chromosome. This technique is also known as "targeted mutagenesis".

 **SCHEDULE**

Week 8

 **General Introduction**, Page 3

 **Reassortment of Yeast Mutant Genes**

 Introduction, Pages 4-7

 Mate Yeast, Pages 8-9

 **Cloning of a Plant Gene Using Yeast**

 Introduction, Pages 20-24

 Yeast transformation with *A. thaliana* cDNA library, Pages 25-26

Week 9 Spring Break

Week 10

 **Reassortment of Yeast Mutant Genes**

 Evaluation of Matings, Page 10

 **Cloning of a Plant Gene Using Yeast**

 Replica plating of yeast transformants, Page 27

 **Deletion of a Gene From the Yeast Chromosome**

 Introduction, Pages 35-44

Preparation of pRS403 plasmid and PCR amplification of HIS3 gene from pRS403, Pages 45-46

 Follow-up lab:

 **Reassortment of Yeast Mutant Genes**

 Sporulation of diploids, Page 11

 **Cloning of a Plant Gene Using Yeast**

 Replica plating of yeast transformants – selection, Page 28

Week 11

 **Reassortment of Yeast Mutant Genes**

 Meiospore enrichment, Page 11

 **Cloning of a Plant Gene Using Yeast**

Observe plates, Page 28

 **Deletion of a Gene from the Yeast Chromosome**

 Gel analysis of PCR reaction, Page 47

 Follow-up lab:

 **Reassortment of Yeast Mutant Genes**

 Plate meiospores, Page 13

Week 12

 **Reassortment of Yeast Mutant Genes**

 Plating of germinated spores onto YPD, page 14

 **Cloning of a Plant Gene Using Yeast**

 Selection of a positive yeast colony, Page 29

 **Deletion of a Gene from the Yeast Chromosome**

 Yeast transformation, Page 48

Week 13

 **Reassortment of Yeast Mutant Genes**

 Replica plating onto dropout media, Page 15

 **Cloning of a Plant Gene Using Yeast**

 Rescue of a positive *A. thaliana* clone from yeast, Page 30

 Transformation of electrocompetent *E. coli* cells, Page 31

 **Deletion of a Gene from the Yeast Chromosome**

 Replica plating yeast colonies, Page 49

 Follow-up lab:

 **Cloning of a Plant Gene Using Yeast**

Identification of *E. coli* transformed with the positive clone, Page 32

Week 14

 **Reassortment of Yeast Mutant Genes**

 Record results of replica plating, Page 16

 **Cloning of a Plant Gene Using Yeast**

Mini-prep and restriction digest of positive *E. coli* clone, Page 33-34

 **Deletion of a Gene from the Yeast Chromosome**

 Colony PCR of red and white yeast colonies, Page 50-51

 Follow-up lab

 **Deletion of a Gene from the Yeast Chromosome**

Agarose gel analysis of the PCR products, Page 52

Week 15

 **Laboratory Presentations**

Follow-up lab

 Review for exam

**Final Exam: Exam week**

**ABSENCE POLICY**

All course material will be presented in lecture and laboratory classes. Presence in laboratory class meeting times, including follow-up laboratories, is mandatory. The makeup exam for the missed midterm exam will be given during finals week.

**Final Exam/Paper Date and Time**

Online Final exam Schedule: <http://finalexams.rutgers.edu/>

**ACADEMIC INTEGRITY**

The university's policy on Academic Integrity is available at <http://academicintegrity.rutgers.edu/academic-integrity-policy>. The principles of academic integrity require that a student:

* properly acknowledge and cite all use of the ideas, results, or words of others.
* properly acknowledge all contributors to a given piece of work.
* make sure that all work submitted as his or her own in a course or other academic activity is produced without the aid of impermissible materials or impermissible collaboration.
* obtain all data or results by ethical means and report them accurately without suppressing any results inconsistent with his or her interpretation or conclusions.
* treat all other students in an ethical manner, respecting their integrity and right to pursue their educational goals without interference. This requires that a student neither facilitate academic dishonesty by others nor obstruct their academic progress.
* uphold the canons of the ethical or professional code of the profession for which he or she is preparing.

Adherence to these principles is necessary in order to ensure that

* everyone is given proper credit for his or her ideas, words, results, and other scholarly accomplishments.
* all student work is fairly evaluated and no student has an inappropriate advantage over others.
* the academic and ethical development of all students is fostered.
* the reputation of the University for integrity in its teaching, research, and scholarship is maintained and enhanced.

Failure to uphold these principles of academic integrity threatens both the reputation of the University and the value of the degrees awarded to its students. Every member of the University community therefore bears a responsibility for ensuring that the highest standards of academic integrity are upheld.

**STUDENT WELLNESS SERVICES**

[**Just In Case Web App**](http://m.appcreatorpro.com/m/rutgers/fda9f59ca5/fda9f59ca5.html) <http://codu.co/cee05e>

Access helpful mental health information and resources for yourself or a friend in a mental health crisis on your smartphone or tablet and easily contact CAPS or RUPD.

**Counseling, ADAP & Psychiatric Services (CAPS)**

(848) 932-7884 / 17 Senior Street, New Brunswick, NJ 08901/ [www.rhscaps.rutgers.edu/](http://www.rhscaps.rutgers.edu/)

CAPS is a University mental health support service that includes counseling, alcohol and other drug assistance, and psychiatric services staffed by a team of professional within Rutgers Health services to support students’ efforts to succeed at Rutgers University. CAPS offers a variety of services that include: individual therapy, group therapy and workshops, crisis intervention, referral to specialists in the community and consultation and collaboration with campus partners.

**Violence Prevention & Victim Assistance (VPVA)**

(848) 932-1181 / 3 Bartlett Street, New Brunswick, NJ 08901 / [www.vpva.rutgers.edu/](http://www.vpva.rutgers.edu/)

The Office for Violence Prevention and Victim Assistance provides confidential crisis intervention, counseling and advocacy for victims of sexual and relationship violence and stalking to students, staff and faculty. To reach staff during office hours when the university is open or to reach an advocate after hours, call 848-932-1181.

**Disability Services**

(848) 445-6800 / Lucy Stone Hall, Suite A145, Livingston Campus, 54 Joyce Kilmer Avenue, Piscataway, NJ 08854 / <https://ods.rutgers.edu/>

Rutgers University welcomes students with disabilities into all of the University's educational programs. In order to receive consideration for reasonable accommodations, a student with a disability must contact the appropriate disability services office at the campus where you are officially enrolled, participate in an intake interview, and provide documentation: <https://ods.rutgers.edu/students/documentation-guidelines>. If the documentation supports your request for reasonable accommodations, your campus’s disability services office will provide you with a Letter of Accommodations. Please share this letter with your instructors and discuss the accommodations with them as early in your courses as possible. To begin this process, please complete the Registration form on the ODS web site at: <https://ods.rutgers.edu/students/registration-form>.

**Scarlet Listeners**

(732) 247-5555 / <http://www.scarletlisteners.com/>

Free and confidential peer counseling and referral hotline, providing a comforting and supportive safe space.