RUTGERS
School of Environmental and Biological Sciences

COURSE SYLLABUS

Experimental Evolution
11:216:353 (4 credits)
Summer 2017
MTWTh 9am-1pm
Foran Hall 193 (wet lab), 124 (computer lab)

CONTACT INFORMATION:

Instructor(s): Siobain Duffy and Mansha Seth Pasricha
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Office Hours: by email appointment

PRE-REQS: 01:119:116 & 117 AND 01:447:380 OR permission of instructor

COURSE WEBSITE, RESOURCES AND MATERIALS:

- Sakai/Canvas site TBD
- Kassen, R. 2014. Experimental Evolution and the Nature of Biodiversity. Greenwood Village, CO: Roberts & Co.
- Primary literature as listed on schedule and subject to vary each time the course is given

COURSE DESCRIPTION:

This summer lecture-laboratory course will introduce undergraduates to the breadth of research using experimental evolution and will give them hands-on experience with viral evolution. The lecture portion of the course will follow a recent textbook, and lab assignments during the course will include readings from the recent primary literature and emphasize the interpretation of graphs. Most class days will consist of one hour of lecture and discussion and 2-3 hours of lab work, though some sessions will be entirely devoted to lab work and analysis of project results.

The laboratory portion will be one long evolution experiment and analysis of its results. We will use strains of a fast-evolving RNA virus that cannot infect humans (the bacteriophage phi6). The speed of RNA virus evolution greatly exceeds that of cellular organisms (e.g., yeast, *Drosophila*, mice), providing the perfect opportunity to see a great deal of phenotypic and molecular change within a summer course. Phage phi6 is usually grown on a particular strain of bacteria, *Pseudomonas syringae* pathovar *phaseolicola*, however host-range mutants of phi6 can infect and grow on other *Pseudomonas* strains. All of these bacterial strains are not harmful to humans and both phage and hosts can provide a safe introduction to experimental evolution with microbes.

Students will tackle a project designed to reveal how viruses adapt. Specifically, each lab group (2 students) will receive a unique phage strain that had been evolved on a novel host for 30 days (~150 passages). As these phage adapted to the novel host they accumulated mutations, some of which led the phage to be able to have more descendants more quickly than they could at the start of the 30 days. We have fully sequenced the genome of these phage strains and know where the mutations are – many of which are in the gene responsible for host range, named P3. However, some of these mutations also affected how well these phage could infect other hosts such as the ancestral, typical lab host, *P. syringae* pv *phaseolicola* – through evolution on a novel host, the phage became less effective at infecting the ancestral lab host. In the course, students will evolve one phage strain on the ancestral lab host, *P. syringae* pv *phaseolicola*. Each lab group will conduct four replicate evolution lineages. At the end of 20 days of passaging, the four lineages will be more effective at growing on the typical host, due to one or more mutations. RNA genomes will be isolated from the four lineages, students

will prepare the genomes for sequencing, and the single gene that is responsible for host range in phi6 (P3) will be sequenced to determine what changes, if any conferred increased fitness on *P. syringae* pv *phaseolicola*. We are especially interested in whether the phage will restore fitness by reversion (getting rid of mutations that had been useful when growing on the novel host) or by further genetic changes.

LEARNING GOALS:

This course furthers several of the learning goals of the Ecology, Evolution and Natural Resources major:

- Describe evolutionary origins, processes and patterns.
- Develop an understanding of software and laboratory techniques commonly used in the study of evolution.
- Communicate scientific concepts effectively in an oral presentation.

ASSIGNMENTS/RESPONSIBILITIES & ASSESSMENT:

Learning Goal assessments:
Increased understanding of evolutionary origins, processes and patterns will be assessed through performance on at least five short-answer questions on each of the four quizzes. The percentage score on these questions will determine level of mastery: >90% Outstanding, 80-89.9% Good, 70-79.9% Satisfactory, <69.9% Unsatisfactory.

Proficiency in laboratory techniques will be assessed through observation of microbiological lab technique, proper lab notebook maintenance and successful use of DNA sequence analysis software. Following instructions and making recommended corrections in technique will result in Satisfactory achievement. Anything less than a 75% score over all six lab notebook reviews, persistent difficulties in using the microbial lab techniques in the 5th week of lab, or inability to complete the DNA sequence assignment for >2 days in the computer lab will be Unsatisfactory.

Proficiency in scientific communication will be assessed during final oral presentations on each group's results. The presentation will be graded according to the attached rubric, and total score will determine level of mastery: >90% Outstanding, 80-89.9% Good, 70-79.9% Satisfactory, <69.9% Unsatisfactory.

Grades will be based on:

In-lab assignments (10 pt/lab for 11 labs, drop lowest score)	100 pt
Quizzes (15 pt/quiz for 4 quizzes)	60 pt
Final presentation	40 pt
Final project writeup	30 pt
Lab Notebook reviews (6.7 pts/review, 6 reviews)	40 pt
Attendance and participation	30 pt
Total	300 pt

Final letter grades will be based on: 90-100% (A), 85.1-89.9% (B+), 80-85% (B), 75-79.9% (C+), 70-74.9 (C), 60-69.9 (D).

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/

ABSENCE POLICY

This is an unusual lab class in that we are not conducting a series of independent activities with pre-ordained results. We conducting research that has not been done before – so we aren't sure what the results will be – and because we are conducting one long evolution experiment every day's work builds on the previous day's results. There is no way to make up work missed so absences will be very disruptive. We will accommodate absences for documented reasons in according to Rutgers' policies only. If you need to miss one or two classes, please use the University absence reporting website https://sims.rutgers.edu/ssra/ to indicate the date and reason for your absence. An email is automatically sent to us – this is the only way to officially inform us of your absence. Please note that attendance is mandatory on the final day (8/16/17) to participate in the final presentation.

THERE IS NO FINAL FOR THIS COURSE, ONLY A PRESENTATION OF EXPERIMENTAL RESULTS ON THE LAST DAY, ON WHICH ATTENDANCE IS MANDATORY.

COURSE SCHEDULE ATTACHED

ACADEMIC INTEGRITY

Rutgers has a comprehensive and thorough policy on Academic Integrity (see below, including links). In summary: don't cheat, give credit where credit is due and be forthright and honest in your lab notebooks. This course allows each of you to conduct research on an NSF-funded grant on viral evolvability that can influence our understanding of how RNA viruses host-shift and evolve. Hopefully our work will impact how public health agencies model viral emergence for their emergency management predictions. There are real and persistent consequences if research isn't conducted with the upmost integrity. Your lab notebooks must reflect what actually happened – mistakes happen, accidents happen, but attempting to cover them up is not just a violation of university academic integrity but harms the larger research project. In fact, it harms the opportunity for undergraduates to participate in research, period. So be honest, don't cheat and don't present someone's words or images without permission and giving credit.

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

School and life can get stressful, and Rutgers has many ways to get help. We can offer you a Safer Space and help you connect to these resources, but here is information you might need.

Just In Case Web App 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/
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/
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.

216:353 summer 2017 schedule

All readings should be completed /before/ the assigned class

	Lecture/Discussion	Lab Activity
Week 1 – Foran 193		
7/10	Introduction	Lab Safety
	Reading primary literature	How to use a lab notebook
		Streak a bacterial quandrant plate
		Set up o/n bacterial culture
7/11	Kassen Ch1	Dilution of a phage stock, plating phage
		Hand in Lab Assignment
7/12	-	Titer phage: Count plaques, calculate titer
		Make a phage lysate: harvest, remove bacteria
		Hand in Lab Assignment
7/13	Kassen Ch2	Titer phage, calculate volume for ~300 pfu
		Hand in Lab Assignment
		Lab Notebook Check
Week 2 – Foran 193		
7/17	Kassen Ch3	Plate ~300 pfu for D=1 of experiment
7/18	-	Make lysate, dilute and plate for D=2
		Hand in Lab Assignment
7/19	-	Make lysate, dilute and plate for D=3
		Hand in Lab Assignment
7/20	quiz	Make lysate, dilute and plate for D=4
		Lab Notebook Check
(Professors and TA passage samples from Friday-Sunday, plate for D=7 for students)		
Week 3 – Foran 193		
7/24	Kassen Ch4	Make lysate, dilute and plate for D=8
		Hand in Lab Assignment
7/25	Chao 1990 Nature	Make lysate, dilute and plate for D=9
		Hand in Lab Assignment
7/26	Kassen Ch5	Make lysate, dilute and plate for D=10
		Hand in Lab Assignment
7/27	quiz	Make lysate, dilute and plate for D=11
		Lab Notebook Check
(Professors and TA passage samples from Friday-Sunday, plate for D=14 for students)		
Week 4 – Foran 193		
7/31	Kassen Ch6	Make lysate, dilute and plate for D=15
		Hand in Lab Assignment
8/1	Kassen Ch7	Make lysate, dilute and plate for D=16
		Hand in Lab Assignment
8/2	Wichman et al 2005	Make lysate, dilute and plate for D=17
		Hand in Lab Assignment
8/3	quiz	Make lysate, dilute and plate for D=18
		Lab Notebook Check

(Professors and TA passage samples from Friday-Saturday, plate for D=20 for students. Sunday is a buffer day in case of need for a catch-up day for some groups, also Monday can be a second buffer day if needed)

Week 5 – Foran 193

8/7	Kassen Ch8	Harvest D=20 lysates, titer
8/8	Kassen Ch9	Extract RNA, cDNA conversion
8/9	Kassen Ch10	PCRs
8/10	quiz	Gel electrophoresis, Exo-SAP
		Lab Notebook Check

(Professors and TA prepare and submit PCR products for sequencing)

Week 6 – Foran 124 (computer lab)

8/14	Analyze sequences
8/15	Work on presentations, submit project writeup
	Lab Notebook Check
8/16	Final presentations, compare class results

Additional readings:

Experimental Evolution, Kaweki et al, Trends in Ecology and Evolution, October 2012, Vol. 27, No. 10

Evolution of Host Specificity Drives Reproductive Isolation Among RNA viruses, Duffy et al, *Evolution 61-11: 2614–2622*

Fitness of RNA virus decreased by Mullers Ratchet, Lin Chao, *Nature, Vol 348, 29 Nov 1990*

Frequent coinfection reduces RNA virus population genetic diversity, Dennehy et al, *Journal of Heredity, 2013, 104 (5), 704-712*

Evolutionary Genomics of host-use in bifurcating denes of RNA virus phi-6, Turner et al, *BMC Evolutionary Biology, 2012, 12:153*

Pleiotropic costs of niche expansion in the RNA bacteriophage phi6. Duffy et al, *Genetics, 2006, 172:1-7.*

Genome Dynamics during experimental evolution, Barrick and Lenski, *Nature Reviews Genetics, Dec 2013, Vol 14*

Experimental coevolution with bacteria and phage: The *Pseudomonas fluorescens*—Φ2 model system, Brockhurst et al, *Infection, Genetics, and Evolution, July 2007, Vol 7, Issue 4, 547-552*

Adaptive molecular evolution for 13,000 phage generations: a possible arms race, Wichman et al, *Genetics, May 2005, 170 (1), 19-31*

EXPERIMENTAL EVOLUTION 11:216:353
Grading Sheet for 216:353
Oral Presentations Rubric

Lab Group _____

_____ *(max) Points*

Sourcing and credits (10 pt):

Graphic impression (4) _____

Easy to read slides (3) _____

Copyright and source information for all images (incl. students' own) (3) _____

Content (20 pt):

Introduction: was the question framed well and in an interesting way?
All accurate information? (4) _____

Methods: accurate and complete without being too detailed? (4) _____

Results: complete, accurate, intelligible, well-discussed? (8) _____

Use of images, table, graphics (4) _____

Individual contribution _____ (10 pt):

Spoke for at least 40% of the time (3) _____

Audible, articulate? (5) _____

Handled questions well? (2) _____

Individual contribution _____(10 pt):

Spoke for at least 40% of the time (3) _____

Audible, articulate? (5) _____

Handled questions well? (2) _____

Total points for _____: (40) _____

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