NGRI Phage Course, Fall Semester, at Washington University in St. Louis: Washington University in St. Louis Washington University in St. Louis Washington University Washington University Washington University Washington University Washington University Matter Dial What We Did Men We Did It

At Washington University (WU), we had 18 students in the fall semester. They were paired by the instructor according to biology background and schedule. Each pair isolated, purified and characterized a putative novel mycobacteriophage by the middle of November.

Just

learning





Plaques of BEEST (above) And J-Gladiator (below)





EM work (left), and results Buckbeak (above) and Uncle Howie (below)

First time gel loading (left and center), and J-Gladiator gel (right)





Our class met 5 hours a week on Tuesday and Thursday afternoons. During phage purification, students were expected to work briefly on Wednesdays and Fridays.

class	Tuesday (1-4 PM)	Thursday (1-3 PM)	Reading
1		 Intro to overall experiment (KH) 	 All the World's a Phage: discussion
		 Intro to "how class works" (KH) 	9/2/08
		 "getting to know you" and surveys 	 Manual: Capture A1-A6 for 9/2/08
2	 Intro to phage and bacteria growth (KII) 		 Manual: Capture D1-D4 for 9/4/00
	 HHMI lab safety video (30') 		
	 discuss lab notebooks (KH) 		
	 Demo/Lab:Aseptic technique (KH) 		
~	 Lab: learn pipetting/micropipetung 	show the state second contract (1077)	- Coderes essential destructions
3		 phage/bacteria growth, continued (KT) how to collect soil (KH) Note: collection 	 Sadava pages and Hersney chapter: discussion and PR due 9/9/09
		outside of scheduled lab time	Enrichment handout for 9/9/08
		 Lab: preparing/plating top agar 	- Entremment nandoac for 5/5/00
4	 Discuss/review central dooma, including 	- car, proparing pauling top agai	 Manual: Tame A1-A15 for 9/11/08
	Hershey/Chase reading (KH)		
	 Lah: process soil samples 		
	 Wednesday: Start enrichment 		
5		 Lab: Examine plates; proceed (spot test) 	 Kornberg article: discussion and readin
		Lab: continue enrichment procedure	response due 9/16/08
		 Wed and Fri purification as needed 	 Manual: Tame B1-B9 for 9/16/08
6	 Central Dogma and discussion of Kornberg 		
	reading (KH and SE)		
	 Lab: purify plaques 		
	 Lab: calculate/test phage for web plate 		
	 Lab: plate lysate from enrichment 	tak a Washington	Martine A New York and All and
1		 Lab: purity plaques 	 Maniatis and Ptasnne article: discussio
		 Lab: examine/punity plaques from cariabaset 	Manualy Camburg C1 C6 fee 0/23/08
		- Wed and Fri swifesting as and d	 Mailual: Capture C1*C6 101 9/23/08
8	Lecture: intro to phage life cycle and reading	- wea and rin partication as needed	
	discussion (PL)		
	 Lah: plate for flood/web plates 		
	 Wednesday: flood plates 		
9		 Lab: harvest phage lysate 	 Manual: Dissect B1-C8 for 9/30/08
		Quiz 1	
10	 Lecture: Restriction enzymes (KT) 		
	 Lab: titer phage lysate 		
11		Catch-up day!	
		 plan digests 	
12	Lecture: Gel electrophoresis (KH)		
	 Lab: purify phage DNA 		1

13		 Lab: pour gels 	
		 Lab: set up DNA digests 	
14	Lab: run gel electrophoresis Lab: photograph/analyze gel results talk with Dr. Barker from HHM1		Manual: Dissect D1-D4 for 10/16/0
15		 Discuss each phage results: compare/contrast size, etc. 	 Manual Dissect A1-A3 for 10/21/08 microscopy reading assignment
16	 Lecture: microscopy (KT) Lab: run required JGI gel on selected samples 		
17		Prepare for EM Quiz 2	
18	 Lab: Electron microscopy OR discussion of reading responses (with Kelsey and Ryan) 		
19		 Lab: Electron microscopy OR discussion of reading responses (with Kelsey and Ryan) 	
20	 Lab: share/discuss EM results and phage choice run gels as needed 		
22	-	 Finalize phage choice run gels as needed 	 Mediaville, et al article: discussio and reading response due 11/13 (Note this article replaces Pedulla al)
23	Celebrate success! Discuss reports Intro to in silico big ideas (CS)		
24		 discussion of Mediavilla article reading response due report work 	 Hugenholtz and Tyson article and Turnbaugh et al.: discussion and reading response due 11/18/08
25	 Jeff Gordon talk and paper discussion intro to BLAST (CS) 		
26		 Computer lab: practice with BLAST 	
!7 !8	Report work day individual consultations Final Presentations Final Reports due		
29	The report of	Archiving	

Acknowledgements: Thanks to our great TA's Kelsey Tinkum, Ryan Lee , Anthony Tubbs and Matt Dothager. Thanks to guest lecturers Petra Levin, Jeffrey Gordon, Douglas Berg, Swaine Chen and Roger Beachy. We could not have done the EM work without the the excellent guidance of R. Howard Berg at the Danforth Plant Science Center. Thanks to the SEA staff, and to Deborah Jacobs-Sera for excellent M. smeg hints. Students were surveyed at the end of the semester.

Selected Student Survey Comments:

• I think the time commitment for the class was great. It was enough to make sure people were dedicated, but not too much to turn us away from research.

- I thought the time commitments were very reasonable, and the way partners were paired based on their schedules worked out really well.
- I liked having a partner, and the fact that we didn't pick our own. Coming in as a freshman, I didn't know anyone in the class.
- . I like having partners as we can discuss problems and share ideas.

• I really like how the readings all coalesced by the end of the semester to give us a broad background of the importance of phages and how our work in the lab fits into the broader picture.

This was a terrific class! The only thing I regret is not getting to do more
experiments like observing plaque morphology over time, testing temperate

phages, calculating burst size, or trying to find the phage receptor

• I have very much enjoyed this semester and am eager for the next...I'm sure it will be a "BLAST". Sorry for the pun.



Lessons Learned:

Pairing partners by schedule worked well. With one exception the groups
worked well together, and contributed equally to the work.

• We need to take better quality plaque photos, and be more organized in storing and sharing photos.

 Working more than two days a week is necessary to get DNA ready for the JGI deadline.

• The DNA isolation protocol needs to be improved. We felt we lost a lot of yield.

Students need more in-class time to work on notebooks, or notebooks need to be allowed out of the lab.

We need to give better instructions for what is expected on writing
assignments.

Always have a back-up M. smeg culture ready!

This class was a great experience for students, TA's and instructors. The more help the better!