

Genome Editing Workshop Syllabus

BSC 6926 Workshop Bio: Genome Editing

Lectures: Tuesday GL 245, Thursday GL 245

Laboratory: AHC1 229

T/TH 3-4:45pm (additional lab time TBD)

Dr. Matthew DeGennaro

Fall 2018

NOTE: SCHEDULE IS PRELIMINARY AND SUBJECT TO CHANGE

Summary:

This workshop provides the tools you need to edit a genome. We will be using CRISPR/Cas9 to mutagenize genes in three organisms, *Aedes*, *Symbiodinium*, and *Aiptasia*. If you are interested editing the genome of another organism, that can be arranged.

As detailed below, most of the class will be hands on. Each participant will pick a gene. A list of genes to target will be provided for you to choose from. As an alternative you can pick your own gene that is related to your current research. We will identify CRISPR sites to generate a null mutation in the gene and clone the gene region to check for polymorphisms. Once the gene sequence is verified, we will proceed to make the CRISPR RNA.

During the last meeting of the course, I would like everyone to prepare a short PowerPoint presentation about the gene they targeted and the CRISPR that was made. Please send me the PowerPoint after it is presented.

Papers to discuss at first class:

1. Barrangou R, Fremaux C, Deveau H, Richards M, Boyaval P, Moineau S, et al. CRISPR provides acquired resistance against viruses in prokaryotes. *Science*. American Association for the Advancement of Science; 2007 Mar 23;315(5819):1709–12.
2. Kistler KE, Vosshall LB, Matthews BJ. Genome engineering with CRISPR-Cas9 in the mosquito *Aedes aegypti*. *Cell Rep*. 2015 Apr 7;11(1):51–60.
3. Tebas P, Stein D, Tang WW, Frank I, Wang SQ, Lee G, et al. Gene editing of CCR5 in autologous CD4 T cells of persons infected with HIV. *N Engl J Med*. 2014 Mar 6;370(10):901–10.
4. Bassett AR, Tibbit C, Ponting CP, Liu J-L. Highly Efficient Targeted Mutagenesis of *Drosophila* with the CRISPR/Cas9 System. *Cell Rep*. 2013 Jul 11;4(1):220–8.

Lecture/discussion meetings on Tuesdays and Thursdays at 3 to 4:45pm:

8/21	Intro to Genome Editing (lecture & paper discussion)	GL 245
8/23	Choosing a site in the genome to target (primer design)	GL 137
8/28-31	LAB: Gene locus sequence verification (PCR)	AHC1 229
9/11-13	LAB: Cloning amplified region for sequencing (TA cloning)	AHC1 229

9/25, 9/27	Sequence analysis, CRISPR primer design	GL 245, GL 245
10/2-4	LAB: CRISPR PCR and purification	AHC1 229
10/16-18	LAB: CRISPR RNA transcription	AHC1 229
10/30, 11/1	Presentation of data	GL 245, GL 245