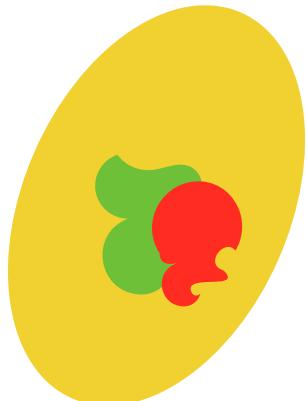


Bio393: Genetic Analysis
Problem Set #5
Due on Friday, February 16, 3 PM

Name: _____

Question 1:

Draw a cross to differentiate between maternal-effect inheritance and cytoplasmic inheritance? Hint: think about multiple generations.

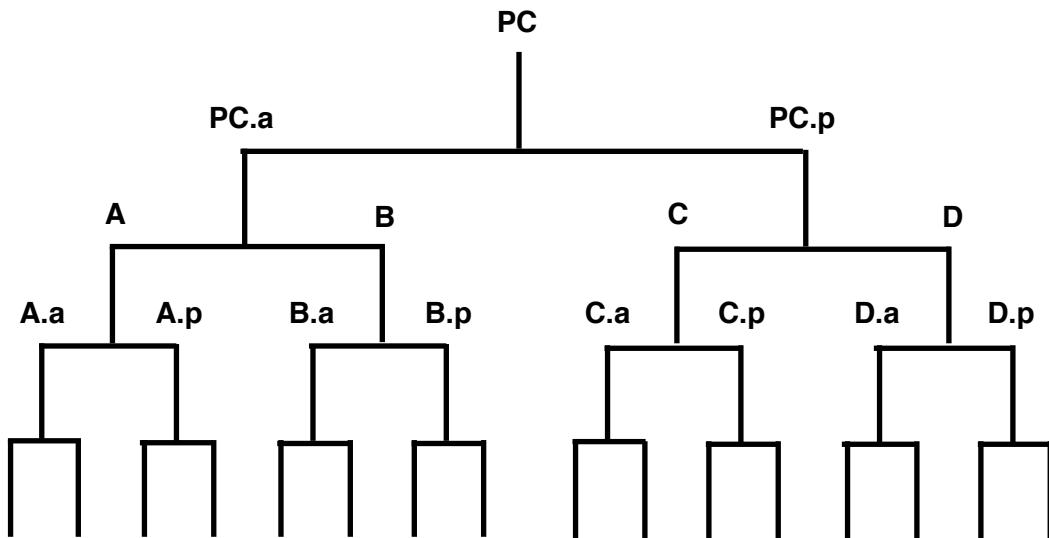


Question 2:

In *Drosophila*, you can generate twin spots using cell-specific markers. In the example below, red ommatidia are homozygous for the mCherry gene, green ommatidia are homozygous for the GFP gene, and yellow ommatidia are heterozygous for mCherry and GFP. Draw out the diploid homologous chromosomes with centromeres demarcated as open and closed circles and locations of the GFP and mCherry insertions that would lead to this mitotic recombination result.

Question 3:

You are studying a relative of *C. elegans* named *C. horvitzii*. Just like Bob Horvitz and John Sulston, you want to generate a lineage map of the organism from the zygote to the adult animal. Below is the lineage you have constructed so far.



You need to place cells 1-16 on the lineage. Using ablation to generate the following data:

Ablated cell	Cells that are present
PC.a	2, 4, and 9-14
A	2, 4, 5, 6, and 9-16
A.a	2, and 4-16
B.p	1,2,3,4,7,8, and 9-16
D	1,3,4,5,6,7,8,9,10,13,15, and 16
D.p	1,3,4,5,6,7,8,9,10, and 12-16
C.p	1-8, 10, 11, 12, 14-16

(a) Label the lineage above with cells 1-16.

You have several lineage defective mutants that fail to form certain cells and their direct descendants. Using these mutants, you attempt to discern which cells are needed for the development of particular components of the adult body.

In mutants for lineages A, B, C, and D.a, you always see the tail formed correctly. Mutants for lineage D never have a proper tail.

In mutants for lineages A.p, B, and PC.p, you always see a proper vulva. However, mutants for lineages A and PC.a never have a proper vulva.

In mutants for lineages A, B.p, C.a, and D, you always see proper formation of the gonad. However, mutants for lineages B and C do not have gonads.

(b) Which cells are responsible for tail, vulva, and gonad formation?

Question 4:

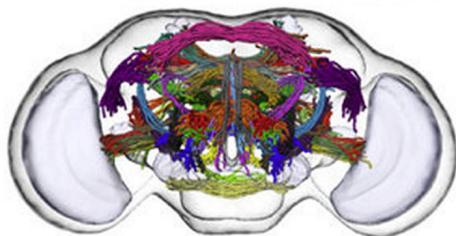
You are interested in the development of a set of cells that make the opening of the secretory duct to the intestine of *Drosophila*. Fourteen cells are arranged in two rings with seven cells in each ring. The cells of the first ring are numbered 1-7 and the second ring 8-14. To investigate the developmental origin of these cells, you induce mitotic recombination in flies heterozygous for a somatic cell marker that you can score. The time at which you induce mitotic recombination is prior to when all 14 cells are formed. When you score for clones homozygous for your somatic cell marker in a large number of flies, you observe the following numbers and patterns of clones:

Cells expressing somatic marker	Number of cases
1,3,5,10	7
8,14	10
2,7,8,9,13,14	4
1,3,4,5,6,10,11,12	4
4,6,11,12	6
8,9,13,14	7
9,13	12
1,10	11
4,11	13
6,12	12
3,5	12
2,7	14
All	2

Say as much as you can about the developmental relationships of the cells that makeup this duct.

Question 5:

Drosophila extend their proboscis to eat or drink. This behavior is controlled by a complex system of connected neurons and muscles. The four images below show the process of proboscis extension (temporally from left to right).



(a) You identified a mutant with a recessive phenotype of much reduced proboscis extension that you named *hungry*, and you are interested to figure out in which set of neurons shown in different colors (left) the gene regulates proboscis extension. Using a series of promoters that express gene products in these regions, please describe how you would determine in which set of neurons the *hungry* gene acts. Also, describe any caveats.

(b) You determine that *hungry* acts in the purple set of neurons to regulate proboscis extension. Using the same promoter and optogenetic tools, describe how you would use specific tools to determine if those neurons inhibit or promote proboscis extension.

Question 6 (15 points):

Topoisomerases are essential for proper DNA replication, transcription, and chromosome segregation. In *Drosophila melanogaster*, topoisomerase II (encoded by the *Top2* gene) is required for proper development to adulthood. Animals that are homozygous for a *Top2* null allele die during early larval development. You would like to get additional alleles of *Top2*.

Describe a screen to identify *Top2* but not mutations in other genes that cause larval lethality. You have a bottle of EMS and three fly stocks: the wild type, *Sp/Cyo* (Sternal plural dominant mutant with the Curly of Oster balancer chromosome), and a *Top2(Δ)/In(2LR)Gla* stock (*Top2(Δ)* is a null allele of *Top2* balanced by the *In(2LR)Gla* balancer chromosome that causes recessive lethality and a dominant increase in eye pigmentation). Note, *Cyo/In(2LR)Gla* flies are viable.

Question 7:

You are interested in understanding the genetics of congenital heart defects, so you perform an ENU-mutant screen in mice. After lots of work and heavy mutagenesis, you identify a mutant with a recessive heart defect and generate a pure-breeding stock. You notice that this stock also has a patchy hair loss phenotype, and your classmate (a self-purported mouse expert) points out that the stock is more aggressive than the parental stock.

You decide to cross wild-type parental males to your mutant stock. All of the resulting male offspring from that cross are patchy, but the mice have normal hearts and are less aggressive. After crossing siblings from that cross, you can identify males and females with heart defects and aggressiveness (but no patchiness) and use them to establish a stock that lacks patchy but has heart defects and aggressiveness.

(a) Write out the cross and genotypes along with a brief explanation of why you performed this cross.

(b) Provide a brief explanation for whether (or not) the same gene could be causing all three abnormal traits (heart defects, patchiness, and aggressiveness).

Question 8:

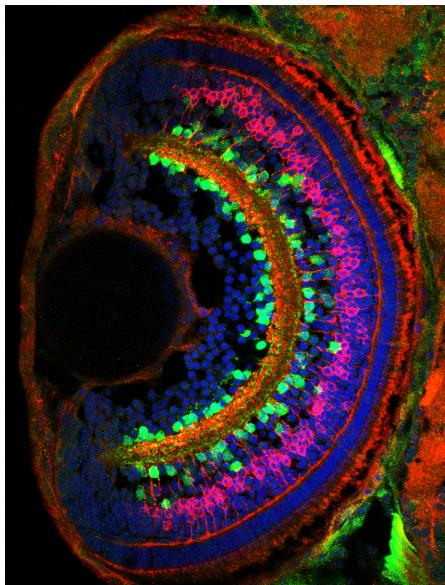
Edward B. Lewis, Christiane Nüsslein-Volhard, and Eric Weischaus shared the 1995 Nobel Prize in Physiology or Medicine for their work on the developmental genetics of *Drosophila*. In their screen for developmental genes, Nüsslein-Volhard and Weischaus initially identified 20 lines bearing maternal-effect mutations that produced embryos lacking anterior structures but with the posterior structures duplicated. When Nüsslein-Volhard mentioned this result to a colleague, he was astonished to hear that mutations in 20 genes could give rise to this phenotype. Explain why his astonishment was completely unfounded and showed a failure to understand genetics.

Question 9:

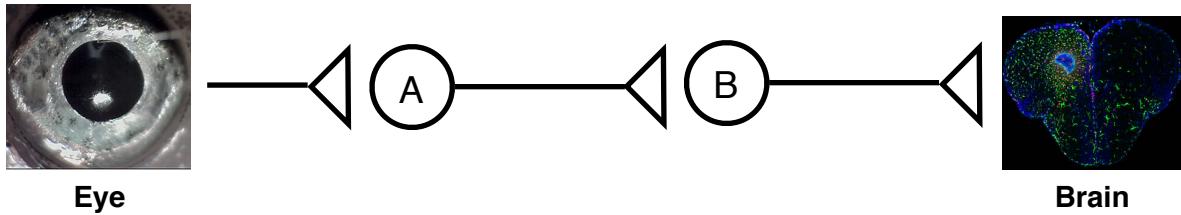
You are interested in vertebrate eye development and function. Therefore, like any good geneticist, you find the most tractable system to study your question – zebrafish.

(a) You perform a mutant screen for fish that are unable to see certain colors and obtain three mutants in three separate complementation groups. Explain whether your screen is saturated and how you would know.

(b) Over the next three years, you map and clone the three genes. You chose zebrafish because of the plethora of genetic tools available. Using promoters that express your gene of interest in the different parts of the zebrafish retina, describe an experiment to test for the function of the *cb* gene in the blue, magenta, red, orange, or green neurons shown in the retina below.



(c) Using channelrhodopsin to manipulate neuronal activity in specific parts of the zebrafish, you would like to determine where and how your three genes act. You perform the following experiments and measure visual activity. Write out where *cb* and *rb* act in the rudimentary neuronal circuit below and the reasoning for your conclusions.



Channelrhodopsin stimulation	Genetic background	Visual activity
None	WT	YES
A neuron	WT	YES
B neuron	WT	YES
None	<i>colorblind</i>	NO
A neuron	<i>colorblind</i>	NO
B neuron	<i>colorblind</i>	YES
None	<i>red-blind</i>	NO
A neuron	<i>red-blind</i>	YES
B neuron	<i>red-blind</i>	YES

Channelrhodopsin stimulation	Genetic background	Visual activity
None	WT	YES
A neuron	WT	YES
B neuron	WT	YES
None	<i>colorblind</i>	NO
A neuron	<i>colorblind</i>	NO
B neuron	<i>colorblind</i>	YES
None	<i>red-blind</i>	NO
A neuron	<i>red-blind</i>	YES
B neuron	<i>red-blind</i>	YES