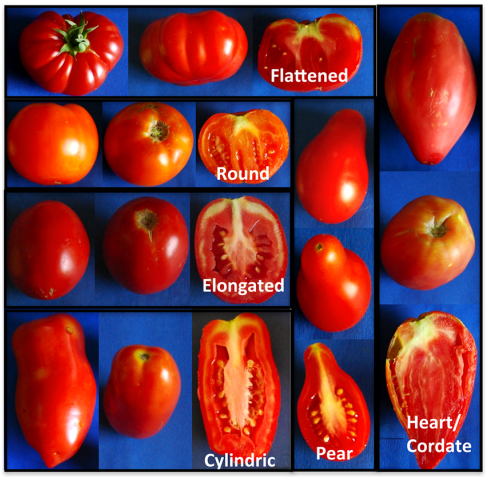
Bio393: Genetic Analysis **Name:**_____
Midterm (6 pages, 64 points)

Question 1 (10 points):
Your farmer parents are so proud that their child goes to Northwestern that they want to make a purple pear-shaped tomato variety (to be named the *wildcat* strain). Pears come in six different shapes and ten different colors, as shown below. As a budding geneticist, you decide to help them out.



They order two true-breeding strains from an heirloom seed company: a round, purple variety and a pear, red variety. You cross the two strains to get all round, red tomatoes in the next generation. Just like peas, it's easy to self tomatoes, so you set up a large number of self crosses from that generation. In your entire garden, you only see tomatoes that are round or pear-shaped and red or purple. The majority of tomatoes are round. You search through 768 tomatoes to find 12 pear, purple ones. When these pear, purple tomatoes are grown and selfed, they give rise to all pear, purple offspring. You've got your *wildcat* strain!

(a, 6 points): Propose a hypothesis for the phenotypic ratio of pear, purple tomatoes you see.

12/768 = 1/64 That ratio indicates that three independently assorting alleles are present in this cross. The rare (pear, purple) tomato class is the triply recessive class, so one of the two traits (fruit shape or color) is controlled by two loci that when both homozygous for the allele that cause the recessive trait are either pear-shaped or purple. Because most tomatoes are round, the fruit shape trait must be controlled by two loci so that only 1/16 of the tomatoes will be pear-shaped.

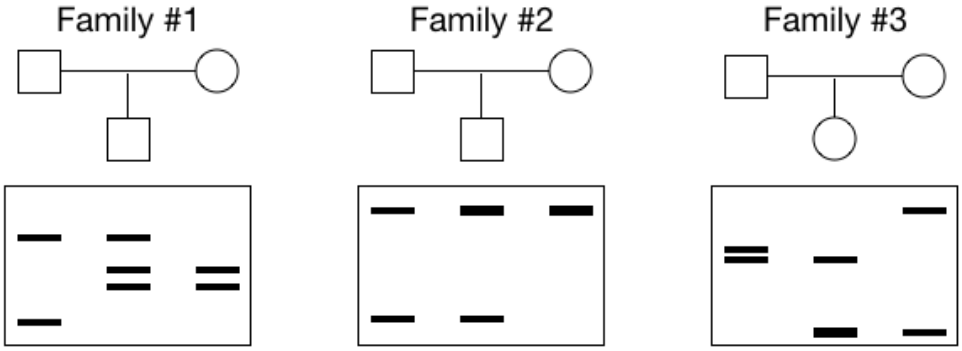
(b, 4 points) Given that hypothesis, how many pear, red tomatoes do you think you would have found if you kept track of them?

*3/4 of the tomatoes will be red and you have to get both pear-shape alleles to express the pear-shape trait, so 3/4 * 1/16 = 3/64 will be pear, red tomatoes*

Question 2 (5 points):
Using linkage mapping, you determined the genetic distance between A and B is 100 cM. What is the minimum number of intermediate markers between A and B that you would need to make this estimate possible? Please explain your reasoning.

You need at least two intermediate markers to estimate the recombination distance between A and B. Because 50 cM is the maximum recombination distance (it looks like independent assortment), two markers gives an average distance of 33 cM if they each are equally spaced between A and B. With one marker, the distance between any two markers would be 50% or appear to be independently assorting.

Question 3 (6 points):
You are interested in discovering genes that influence nondisjunction in human mothers. In order to select embryos with successful *in vitro* fertilization, clinics often look at the products of the union of sperm and egg. Some embryos have obvious visual defects and upon karyotype analysis have triploidy for one chromosome pair (an autosome). You obtain DNA from Mom, Dad, and the abnormal embryo to measure the genotype at a DNA marker on the triploid chromosome. The data from three trios (Mom, Dad, and embryo) are shown as agarose gels. Remember that Mom and Dad are diploid for the chromosome in question. The DNA bands show the different genotypes of the chromosomes by their different length products.



Describe the nondisjunction in each family and any ambiguities.

In Family #1, the heterozygous markers from Mom are both seen in the son with the triploid autosome. That means that NDJ must have occurred in meiosis I when homologs failed to separate. Homologs have different markers because Mom is heterozygous.

In Family #2, the mother is homozygous for the marker on the autosome, so we can't tell if NDJ occurred in meiosis I or II.

In Family #3, only one of the heterozygous markers from Mom are seen in the son with the triploid autosome. That means that NDJ must have occurred in meiosis II when sisters failed to separate. Sisters have the same markers because they are replicated chromosomes.

Question 4 (14 points):
Your roommate does not believe all this mumbo jumbo about genetics. He says that there is no way for you to predict the phenotypes of offspring from the following cross.

You look up on wikipedia that the x, y, and z genes are all linked, and they all cause recessive phenotypes that can be all be scored together with no viability issues. Also, x is 30 map units from y, and y is 10 map units from z. The gene order is x, y, z. Fill out the table below for the numbers of offspring with each phenotype (Total offspring = 1000). Label which phenotypes represent parental or recombinant gametes.

Phenotype	Number of offspring
x y z	**315**
+ + +	**315**
x y +	**35**
+ + z	**35**
x + +	**135**
+ y z	**135**
x + z	**15**
+ y +	**15**

Recombination between x and y should occur with a frequency of 0.3 and between y and z with a frequency of 0.1.

*However, recombination will also occur in both intervals in double recombinants with a frequency of 0.3 * 0.1 = 0.03*

Therefore, if we account for double recombinants:

freq(xy+) = 0.3 - 0.03 = 0.27
freq(+yz) = 0.1 - 0.03 = 0.07

Multiply 1000 by expected freq. and divide by 2 to account for both classes:

*n(xy) = (1000 * 0.27) / 2 = 135*
*n(yz) = (1000 * 0.07) / 2 = 35*

n(double recombinants) =
*1000 * 0.03 = 30.*

Finally, we can calculate the number of parentals:

*Probability of not having a recombinant between x and y (0.7), and probability of not having a recombinant between y and z (0.9). Therefore, the probability of both is 0.7*0.9=0.63 or 630/1000*

Question 5 (18 points):

As an avid SCUBA diver, you are concerned about the loss of coral habitats caused by ocean acidification through rising carbon dioxide levels. You get a huge NSF grant to identify acid-tolerant staghorn coral. You can do coral crosses (once per year) and have a population of acid-sensitive coral. Coral are diploid, and this species is dioecious (males and females).

(a, 10 points) Write out a selection crossing scheme that will allow you to identify mutations in genes that recessively promote acid sensitivity. You can use a mutagen. Describe which crosses are single-pair vs. bulk, when you apply the acidic ocean water selection, and what generation you score for your mutants.

For this selection scheme, we will use acidic ocean water to kill (or severely reduce the growth rate) of acid-sensitive coral strains. Only acid-resistant m/m mutants will survive. There are two ways to do this selection.

Method #1:
1. Mutagenize wild-type +/+ male corals and cross to wild-type +/+ female corals.
2. If mutations have occurred in genes that promote acid sensitivity, then m/+ corals will be produced.
3. Mate m/+ male corals with wild-type wild-type +/+ female corals in single-pair matings.
4.Take the female offspring of this cross, which are m/+ or +/+, and then mate them back to the father male coral who is m/+. This cross can be bulk cross because the mutations are all the same in these individuals.
5. In the next generation, expose all of the offspring to acidic ocean water. Only m/m acid-resistant corals will survive.

Method #2:
1. Mutagenize wild-type +/+ male corals and cross to wild-type +/+ female corals.
2. If mutations have occurred in genes that promote acid sensitivity, then m/+ corals will be produced.
3. Mate m/+ male corals with wild-type wild-type +/+ female corals in single-pair matings.
4.Take the female offspring of this cross, which are m/+ or +/+, and then mate them to their male siblings, which are m/+ or +/+. This cross can be bulk cross because the mutations are all the same in these individuals.
5. In the next generation, expose all of the offspring to acidic ocean water. Only m/m acid-resistant corals will survive.

(b, 4 points) Why are you happy that you can measure acid tolerance in millions of mutagenized coral offspring?

The long generation time makes it difficult to do many crosses each year. You are happy that you can sample the mutagenesis of millions of gametes in one year, otherwise your project would take much longer.

(c, 4 points) A colleague in Australia finds a rare staghorn coral that seems to survive in the highly acidic ocean waters near his lab. His mutant has a dominant phenotype, and one of your mutants also has a dominant phenotype. You think his allele could be an allele of one of the same genes you mutated in your selection. How would you determine if the Australian acid-tolerant true-breeding strain and your true-breeding strain are mutated in the same gene? This coral species has 14 pairs of chromosomes.

Because both alleles confer dominant phenotypes, we can not use complementation to test if they share the same function. We must cross the two true-breeding strains to get a heterozygote at both loci. Then, after crossing heterozygous males and females, we will look for linkage of the two loci. If we never see any sensitive corals from the cross, then we know the two alleles must be tightly linked and possibly be alleles of the same gene.

Question 6 (5 points):
You identify a very rare mutant in a screen for diatoms that fail to create radially symmetric shells. You map the mutation to a chromosome and genomic interval where you have deficiencies (large deletions) and duplications. When you make heterozygotes with your mutant and either deficiencies or duplications, you get the same mutant phenotype with the same penetrance. Please explain why you are happy or unhappy to find this rare mutant and why.

This genetic relationship means that we have identified a neomorph. The mutant phenotype does not get modified by extra or loss of wild-type copies of the gene. We are happy that we got a mutant in the first place. Gain-of-function alleles are rare. However, we are mostly unhappy because neomorphs do not tell us anything about the wild-type function of the gene in the creation of radially symmetric shells.

Question 7 (6 pts):
Your advisor would like you to isolate an amber suppressor (UAG nonsense codon suppressor) using the yeast *S. cerevisiae*. She gives you the following strains. Please select which strain would offer the most efficient way to screen for amber suppressor mutants and explain why. The *ACT1* and *IRA2* mutant phenotypes are different and do not interfere (suppress or enhance) with each other.

act1-6	Missense allele of *ACT1*
ira2-12	Missense allele of *IRA2*
act1-1	Amber nonsense allele of *ACT1*
ira2-87	Amber nonsense allele of *IRA2*
act1-6; ira2-12	*ACT1* and *IRA2* double mutant
act1-6; ira2-87	*ACT1* and *IRA2* double mutant
act1-1; ira2-12	*ACT1* and *IRA2* double mutant
act1-1; ira2-87	*ACT1* and *IRA2* double mutant

The most efficient strain to screen for amber suppressor mutants is the act1-1; ira2-87 strain. The only way to suppress both the ACT1 and the IRA2 mutant phenotypes is through the isolation of an amber suppressor. If we chose either single mutant or the double mutant with only one (or none) of the amber nonsense mutations, then we could get gene-specific suppressors of either gene. Because the two mutant phenotypes can be scored separately, we can screen for informational suppressors that suppress both traits simultaneously.

Please fill out the post-midterm survey at bio393.andersenlab.org