Biology 322 Fall 2012 aha experiment

➔ *This work-up is worth 30 pts and is due on* ***Tuesday 11/6*** *at the beginning of lab. Please start working on this report ASAP so if your have questions about the instructions we can sort them out in class.*
➔ *This report MUST be word-processed and in paragraph form with appropriate subheadings*
➔ *I encourage you to discuss this work-up with your fellow classmates but* ***the product that you submit must absolutely be your own work.***
➔*You will likely need to review plant reproduction and some plant terminology: alternation of generations, sporophyte and gametophyte*
➔ *For each chi square analysis, clearly state the genetic principles that you used to generate the expected numbers in each category*
➔

Title of Lab Report: *Figure out something appropriate based on your exploration of this assignment*
Introduction: *A couple of nicely crafted sentences stating the researcher's rationale for generating the aha3-1 mutation. Use proper terminology and be sure to describe the type of mutation.*
Methods*: OMIT*
Data analysis/presentation:

1. *Briefly describe what you observed with respect to the phenotypes of the seedlings examined in the first part of the experiment. 2-3 sentences. Include a table with your data but don't do any statistical analysis.*
2. *Carefully examine the class genotyping data included at the end of the handout. [A word file is also available on the web site if you want to include the whole table in your lab report (your choice here).]*
3. *Examine the genotype ratios. First discuss the outcome you would expect if assuming no complications to Mendel. State your hypothesis and do the appropriate chi square analysis. NOTE, your report just needs to include the chi square and p values (not the arithmetic). State explicitly what the p value tells you.*
4. *Next consider the data in the context of a complication: sporophyte (zygotic) lethality. State the hypothesis and do the appropriate chi square analysis. State explicitly how the p value helps you assess the validity of your hypothesis. Then, assuming that your hypothesis of sporophyte lethality is correct, discuss possible explanations for the skew in the genotype data include one hypothetical explanation relating to the PCR experiment and one related to the growth/selection of seedlings (you may need to tinker with your underlying genetical assumptions for the latter).*
5. ***Additional info from the literature****: Review the data shown in Table 1 on the next page. Are they consistent with sporophyte (zygotic) lethality? Defend your answer.*
6. ***Again, inspect these data****. What conclusion can you draw from these reciprocal crosses (Table 3) about transmission of the aha3 mutation? Is the class data consistent with this transmission? Do the appropriate chi square analysis. Be very explicit about you how you arrived at the predicted genotypic ratios of the F2s. In other words, explain how you arrived at your expected categories.*

7. *Examine the class data. Which genotype is surprising? How would you explain this data point?*
8. *Take some time to inspect the individual data from each of your fellow classmates and note how it nicely illustrates the principle of chance in small sample sizes. Inspect the data and eyeball the frequency of specific outcomes: 3:1, 1:3, 4:0 etc. Are they more or less consistent with what you would expect from the basic rules of probability. Calculate the likelihood of each possible outcome and include this info in your discussion. You can ignore the data set with just 3 genotypes and the set with the outlier.*
9. *And, finally, At the end of your report attach a labeled print-out of your agarose gel photo. Start with the digital image and make sure that your printed version reveals all of the bands seen in the digital version. Label sizes of HiLo bands (as many as you can confidently assign) , each gel lane, identify products from wild-type and mutant alleles, etc. For each plant indicate genotype (or ND if no PCR products were obtained). Identify and label PCR artifacts.*

DATA from Genetics **168:** 1677–1687 (November 2004)
Feel free look at the original paper, but it should not really be necessary. It would be better to work through the analysis on your own and then check the paper if you are feeling insecure.

AHA3 = wildtype allele

aha3-1 and *aha3-5* = two different T-DNA mutations in the aha3 gene

TABLE 1

PCR genotyping of progeny of seedlings heterozygous for *aha3*

	Progeny genotype			
Parent genotype	*AHA3/AHA3*	*AHA3/aha3*	*aha3/aha3*	*P*
AHA3/aha3-1	126	121	0	0.9
AHA3/aha3-5	110	115	0	0.9

TABLE 3

Determination of gametophytic lethality by reciprocal crosses

Female parent genotype	Male parent genotype	Progeny	
		AHA3/AHA3	*AHA3/aha3*
AHA3/aha3-1	*AHA3/AHA3*	23	25
AHA3/AHA3	*AHA3/aha3-1*	52	0
AHA3/aha3-5	*AHA3/AHA3*	55	52
AHA3/AHA3	*AHA3/aha3-5*	101	0

# / name	Wt ++	Het +-	Homo - -	Not determined (no PCR Products in either reaction}
Jessica Brooks	1	3	0	0
Greg Krause	3	1	0	0
Kevin Hager	3	1	0	0
Nathan Brickett	2	2	0	0
Katy Swift	3	0	0	1
Jonathan York	3	1	0	0
Miranda O'Donnell	2	1	1	0
Briana Kinash	2	2	0	0
Greg Pennington	3	1	0	0
Kevin Hope and Russell Kato	2	2	0	0
Audriana Gonzalez	1	3	0	0
Isadora	3	1	0	0
Teresa	0	4	0	0
Kenny	4	0	0	0
Billy	1	3	0	0
Totals				