

# Quantitative Genetics Lab

## Lab 3

### Background:

When Darwin published *On the Origin of Species* in 1859, a major gap in the theory of evolution was the lack of a mechanism for inheritance. At the time, one generally accepted explanation was 'blending inheritance.' According to the blending inheritance explanation, the phenotype of the offspring of a mating is always intermediate between the parental phenotypes. Without an understanding of inheritance, it was unclear to many whether evolution could proceed through the gradual process Darwin postulated. At the time, many believed that the variation natural selection acted on could not be the continuous variation seen in populations because in each successive generation any progress made by natural selection would be blended away.

An alternate theory, popularized by Francis Galton, was that evolution proceeded not by slow, gradual changes but instead happened quickly, driven by mutations with large effects. Although the rediscovery of Gregor Mendel's experiments on peas clarified the inheritance of simple Mendelian traits, it did not explain the mechanism of inheritance for quantitative traits. Indeed, many thought Mendel's laws of inheritance were in conflict with Darwin's theory of evolution. A controversy between so-called Biometricians (who believed in slow, gradual evolution and mutations with small effects) and Mendelians (proponents of evolution by jumps and mutations with large effects) arose that was not resolved until 1918.

In this lab, you will investigate the mechanism of inheritance that puzzled biologists for nearly 60 years. By looking at the results of crosses between plants with different phenotypes, you will determine the genetic basis of two important plant traits.

To determine the genetics of these traits, you will first need several statistical tools to describe populations of individuals. One description of a population is a 'mean.' The mean is the average value of some measurement of individuals within a population. Another statistical method used to describe a population is its 'variance'. Variance describes how far values lie from the mean.

The equation for a mean ( $\bar{x}$ ) is:

$$\bar{x} = \frac{\sum_{i=1}^n x_i}{n}$$

This can be said as "the mean equals the sum ( $\Sigma$ ) of all values ( $x_i$ ), divided by the number of individuals in the population ( $n$ )."

The equation for variance ( $V_x$ ) is:

$$V_x = \frac{\sum_i^n (x_i - \bar{x})^2}{n}$$

This can be said as "the variance equals the sum of the squares of differences between each value and the mean, divided by the number of individuals in the population."

To understand the mean and variance, we will look at two, very small, non-random populations: five people from your class and the starting five from the Duke 2010 Championship basketball team.

Smith 6'2" = 74"  
Scheyer 6'5" = 77"  
Singler 6'8" = 80"  
Zoubek 7'1" = 85"  
Thomas 6'8" = 80"

Calculate the following values by hand.

Mean = \_\_\_\_\_

Variance = \_\_\_\_\_

Have your TA try to 'randomly' pick 5 people from your class and record their height in inches.

Mean = \_\_\_\_\_

Variance = \_\_\_\_\_

After you've calculated these values, plot them on a scale on the board using different colors.

How do the means and variances differ between these populations?

**What is the genetic basis of trichome and anthocyanin production?**

To study the genetic basis of these plant traits, you will receive plants from several generations of controlled matings (“crosses”). The plant species you are receiving is *Brassica rapa*. It is a close relative of *Arabidopsis thaliana*, the lab rat of the plant world.

Your lab will receive:

24 green hairless parents

24 purple hairy parents

48 F<sub>1</sub>

96 F<sub>2</sub>

The F<sub>1</sub> (1<sup>st</sup> filial) generation is the offspring from a mating between the purple hairy plants and the green hairless plants. The F<sub>2</sub> generation is the offspring from self-fertilizations of many F<sub>1</sub> plants. In a plant that cannot self-pollinate the way *B. rapa* does, the F<sub>2</sub> generation would come from **random** crosses between different F<sub>1</sub> individuals.

*The genetic basis of anthocyanin production:*

Anthocyanins are stress-induced compounds made by plants.

In your group, determine how you want to measure the trait and then compare your methods to the rest of the class.

(One method could be to look at the variation in color and assign numbers 0-6 to the plants. 0 could be completely green and 6 could be the darkest plants.)

After your whole lab has agreed on how you will measure the trait, fill in the chart below.

Plant	Trait Measure	Plant	Trait Measure	Plant	Trait Measure	Plant	Trait Measure
G&HL P-1		P&HR P-1		F1 P-1		F1 P-25	
G&HL P-2		P&HR P-2		F1 P-2		F1 P-26	
G&HL P-3		P&HR P-3		F1 P-3		F1 P-27	
G&HL P-4		P&HR P-4		F1 P-4		F1 P-28	
G&HL P-5		P&HR P-5		F1 P-5		F1 P-29	
G&HL P-6		P&HR P-6		F1 P-6		F1 P-30	
G&HL P-7		P&HR P-7		F1 P-7		F1 P-31	
G&HL P-8		P&HR P-8		F1 P-8		F1 P-32	
G&HL P-9		P&HR P-9		F1 P-9		F1 P-33	
G&HL P-10		P&HR P-10		F1 P-10		F1 P-34	
G&HL P-11		P&HR P-11		F1 P-11		F1 P-35	
G&HL P-12		P&HR P-12		F1 P-12		F1 P-36	
G&HL P-13		P&HR P-13		F1 P-13		F1 P-37	
G&HL P-14		P&HR P-14		F1 P-14		F1 P-38	
G&HL P-15		P&HR P-15		F1 P-15		F1 P-39	
G&HL P-16		P&HR P-16		F1 P-16		F1 P-40	
G&HL P-17		P&HR P-17		F1 P-17		F1 P-41	
G&HL P-18		P&HR P-18		F1 P-18		F1 P-42	
G&HL P-19		P&HR P-19		F1 P-19		F1 P-43	
G&HL P-20		P&HR P-20		F1 P-20		F1 P-44	
G&HL P-21		P&HR P-21		F1 P-21		F1 P-45	
G&HL P-22		P&HR P-22		F1 P-22		F1 P-46	
G&HL P-23		P&HR P-23		F1 P-23		F1 P-47	
G&HL P-24		P&HR P-24		F1 P-24		F1 P-48	

You will add your data to a master table of data for your whole lab that your TA will set up.

Collect the F2 data here:

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E												
F												
G												
H												

Now make histograms of the data (fill in the number of each type in the space provided—we assume you used the 0-6 color measuring scale- please adjust chart if necessary):

	GP	PP	F1	F2
0	_____	_____	_____	_____
1	_____	_____	_____	_____
2	_____	_____	_____	_____
3	_____	_____	_____	_____
4	_____	_____	_____	_____
5	_____	_____	_____	_____
6	_____	_____	_____	_____

Draw the distribution of each generation from the combined data on graph paper (provided) on three different graphs (one for each generation). Use the same scale for each generation.

Calculate these values from the combined data.

G&HL parent            mean\_\_\_\_\_ variance\_\_\_\_\_

P&HR parent            mean\_\_\_\_\_ variance\_\_\_\_\_

F<sub>1</sub>                            mean\_\_\_\_\_ variance\_\_\_\_\_

F<sub>2</sub>                            mean\_\_\_\_\_ variance\_\_\_\_\_

What do these means and variances mean? Are there any problems with using means and variances? (Is there a single distribution or is it bimodal?)

In the space below, can you think about a better way to assess the phenotypes? Does the distribution of the F<sub>2</sub> look like something you've seen before?

What can you say about the genetics of anthocyanin production from the distributions that you described above?

Can the results of these crosses be explained by variation at one locus? Why or why not?

What does the mean of the  $F_1$  tell us about dominance?

*The genetic basis of trichome production*

Trichomes are small hair-like structures on the plant surface. Observe the trichomes produced on the leaf margin of the first true leaf. How will you measure this trait? Is this trait binary (would presence and absence suffice) or do you need to count the trichomes? To investigate the trichomes, it is easiest to look at the first true leaf through a magnifying glass with a piece of black construction paper behind the leaf. Record your observations in the table below.

**IMPORTANT:** Label each of the F<sub>2</sub> plants with their trait measures!  
After you've decided how you will measure the trait, fill in the charts below.

Plant	Trait Measure	Plant	Trait Measure	Plant	Trait Measure	Plant	Trait Measure
G&HL P-1		P&HR P-1		F1 P-1		F1 P-25	
G&HL P-2		P&HR P-2		F1 P-2		F1 P-26	
G&HL P-3		P&HR P-3		F1 P-3		F1 P-27	
G&HL P-4		P&HR P-4		F1 P-4		F1 P-28	
G&HL P-5		P&HR P-5		F1 P-5		F1 P-29	
G&HL P-6		P&HR P-6		F1 P-6		F1 P-30	
G&HL P-7		P&HR P-7		F1 P-7		F1 P-31	
G&HL P-8		P&HR P-8		F1 P-8		F1 P-32	
G&HL P-9		P&HR P-9		F1 P-9		F1 P-33	
G&HL P-10		P&HR P-10		F1 P-10		F1 P-34	
G&HL P-11		P&HR P-11		F1 P-11		F1 P-35	
G&HL P-12		P&HR P-12		F1 P-12		F1 P-36	
G&HL P-13		P&HR P-13		F1 P-13		F1 P-37	
G&HL P-14		P&HR P-14		F1 P-14		F1 P-38	
G&HL P-15		P&HR P-15		F1 P-15		F1 P-39	
G&HL P-16		P&HR P-16		F1 P-16		F1 P-40	
G&HL P-17		P&HR P-17		F1 P-17		F1 P-41	
G&HL P-18		P&HR P-18		F1 P-18		F1 P-42	
G&HL P-19		P&HR P-19		F1 P-19		F1 P-43	
G&HL P-20		P&HR P-20		F1 P-20		F1 P-44	
G&HL P-21		P&HR P-21		F1 P-21		F1 P-45	
G&HL P-22		P&HR P-22		F1 P-22		F1 P-46	
G&HL P-23		P&HR P-23		F1 P-23		F1 P-47	
G&HL P-24		P&HR P-24		F1 P-24		F1 P-48	

You will add your data to a master table of data for your whole lab that your TA will set up.

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E												
F												
G												
H												

Now make histograms of the data:

	GP	PP	F1	F2
0	___	___	___	___
1-3	___	___	___	___
4-6	___	___	___	___
7-9	___	___	___	___
10-12	___	___	___	___
13-15	___	___	___	___
16-18	___	___	___	___
19-21	___	___	___	___
22-24	___	___	___	___
25-27	___	___	___	___
29-31	___	___	___	___
32-35	___	___	___	___
>36	___	___	___	___

**IMPORTANT!!!** From *all* the F2s, record the following values:

# trichomes for the four least hairiest plants:    \_\_\_    \_\_\_    \_\_\_    \_\_\_  
 # trichomes for the four median hairy plants:    \_\_\_    \_\_\_    \_\_\_    \_\_\_  
 # trichomes for the four hairiest plants:    \_\_\_    \_\_\_    \_\_\_    \_\_\_

Now, label those plants! No one leaves lab today until everyone has recorded this data and the plants that correspond to that data are transplanted into fresh pots that are **well labeled**. On the label, please put L, M or H for least, median or hairiest, respectively. Also include the number of trichomes, section number, and date on the label. Write clearly.

Draw the distribution of each generation from the combined data on graph paper (provided) on three different graphs (one for each generation). Use the same scale for each generation.

Calculate these values from the combined data. **This data is also crucial!!!!**

G&HL parent            mean\_\_\_\_\_ variance\_\_\_\_\_

P&HR parent            mean\_\_\_\_\_ variance\_\_\_\_\_

F<sub>1</sub>                            mean\_\_\_\_\_ variance\_\_\_\_\_

F<sub>2</sub>                            mean\_\_\_\_\_ variance\_\_\_\_\_

Does using means and variances accurately represent your data in this case?

How does this trait differ from anthocyanin production?

Can the results of these crosses be explained by variation at one locus? Why or why not?

What can we say about the genetic basis of anthocyanin variation and trichome variation in these populations of *B.rapa*?

### **The contributors to phenotypic variation**

Both of these traits display phenotypic variation. What factors, besides genetic variation, might be contributing to the phenotypic variation?

How can we determine how much of the phenotypic variation is due to genetic differences versus other factors?

Looking back at your distribution of anthocyanin production, assign genotypes to the different phenotypes assuming one locus is causing the variation (i.e. PP or Pp is purple, and pp is green). If we assume that all the purple parent are PP and we know all the colorless parent are pp, what is the genotype of the F<sub>1</sub>s? What are the genotypes of the F<sub>2</sub>s? Label them on your distribution.

Now look at the distribution of trichome production. First we will assume one locus is causing the variation. If AA is hairy and aa is hairless, label your distributions above.

Now, assume four loci of equal effect are causing the variation. A capital letter at any of the four loci causes production of more trichomes and a lower case letter causes the production of fewer. So the genotype of the hairless parent

could be aabbccdd and the genotype of the hairy parent might be AABBCCDD. What is the genotype of the  $F_1$  in this case? What are some examples of the genotypes in the  $F_2$  generation?

What are the contributors to phenotypic variation in the Parental,  $F_1$  and  $F_2$  generations?

Parental:

$F_1$ :

$F_2$ :

**Why is genetic variation important to natural and artificial selection?**

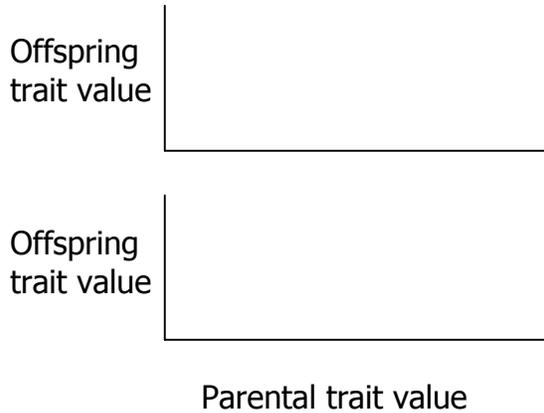
For the *next* plant genetics lab later in the semester, we will be examining the offspring of crosses between the hairiest, medium hairy and least hairy  $F_2$  individuals. (That generation will be called the *selected offspring of the  $F_2$* .)

Predict how the results will look given the following genetic bases for the trait using a regression. A regression shows the relationship between two variables. For example, the regression of water consumption given temperature might look something like this:

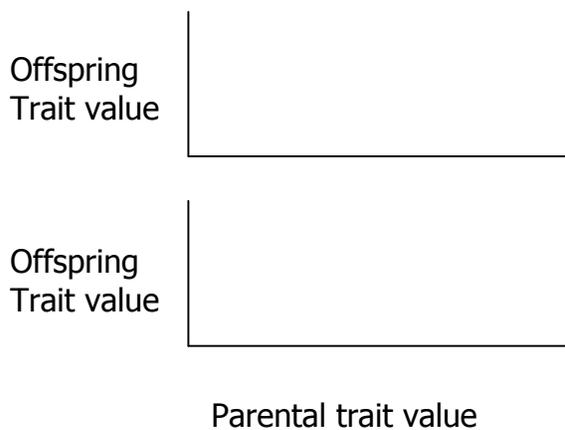


In the first graph, sketch what you think the relationship between the selected  $F_2$  parents and their offspring will be if there is only genetic variation. In the second graph, draw the relationship if there is only environmental variation.

1. One locus controlling the trait



2. Multiple loci controlling the trait



Reference: W. B. Provine, *The origins of theoretical population genetics*. (University of Chicago Press, 2001).