Topic 12. Genetics

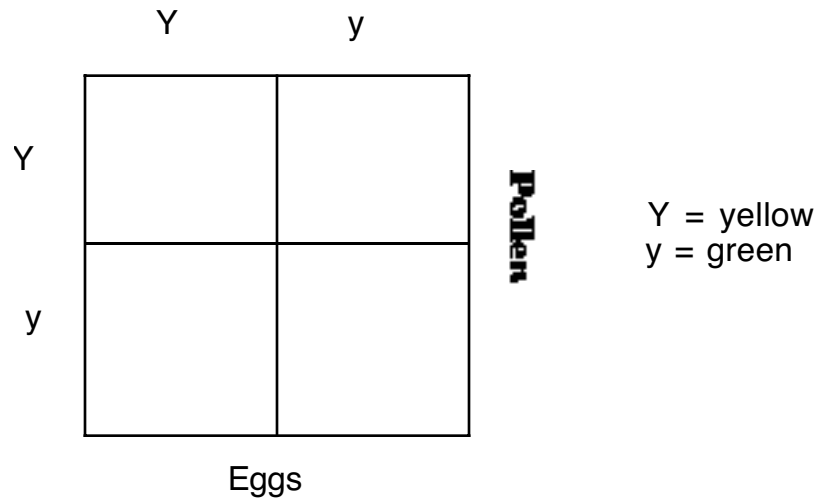
Introduction. Genetics is the study of how the biological information that determines the structure and function of organisms is passed from one generation to the next. It is also concerned with how that information is expressed in individual organisms. While breeders have developed varieties of plants and animals through selective breeding for thousands of years, the science of genetics dates back only to the nineteenth century.

I. Patterns of Inheritance: One Trait Considered

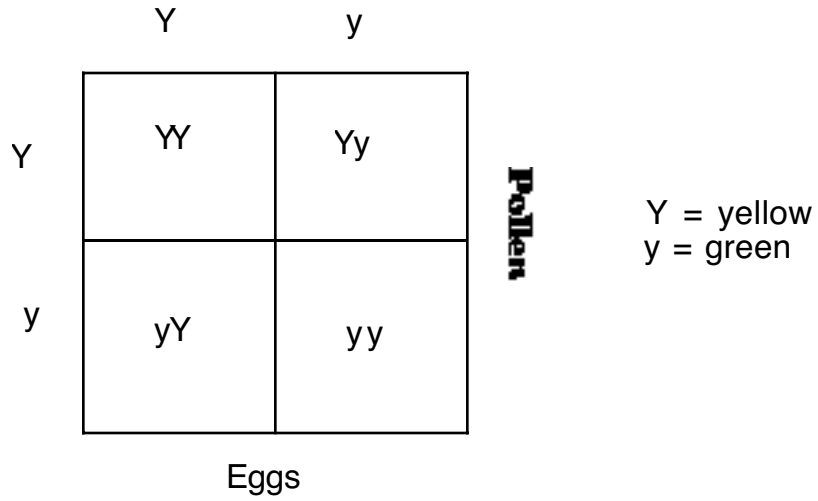
The Law of Segregation: Gregor Mendel was a monk who studied inheritance of traits in peas from 1859 to 1863. For the trait, seed color, for example, he observed that some pea plants bore seeds that were green while others bore yellow seeds. In his experiments he crossed plants from a true-breeding line with yellow seeds to plants from a different true-breeding line that had green seeds. The resulting progeny all exhibited yellow seed color. It was as if the information for the trait, "green seed color" had been destroyed. However, when he self-pollinated these progeny, their progeny included plants that exhibited both green and yellow seeds. From this experiment and others he hypothesized that the information that determined traits in organisms was carried in non-dilutable packets (genes). Further, for any given trait, these genes were carried by organisms two at a time, and that the presence of one gene could mask the presence of the alternate gene for that trait. During sexual reproduction, these genes separate (segregate) into gametes which end up with either one or the other of the two genes carried for each trait. These gametes, in turn, recombine randomly again resulting in plants each with two genes for each trait. By his hypothesis, the line of yellow-seeded peas was made up of a population of individuals all of whom had two genes for yellow seeds. The plants from the line with green-colored seeds each had two genes for green color (hence they bred true). When yellow-seeded plants from one line were crossed with green-seeded plants from the other, all the resulting progeny had both a gene for yellow seed color and one for green. By Mendel's hypothesis, the gene for yellow color masked the presence of the gene for green. When one of these offspring was selfed, however, it produced two kinds of gametes, those with the gene for yellow seed color and those with the gene for green seed color. Further, these two types of gametes should be produced in equal numbers, and they should recombine randomly. i.e. Given an egg with the gene for yellow seed color, there should be an equal chance for it to be combined with pollen bearing either a yellow or green seed color gene. Mendel's hypothesis not only explains why green seed color reappears in the third generation, but, as with any good hypothesis, it makes predictions that can be tested. Specifically, if half the gametes produced by the second generation include the gene for yellow seeds and the other half the gene for green seeds, and if they recombine randomly, we should expect the resulting progeny to manifest a 3 yellow to 1 green ratio.

To understand this consequence we need to create a contingency table called a

Punnet square. On the y axis of this square we include a partition for all the possible types of pollen grains. On the x axis we do the same for all the possible types of eggs.



We then exhaust all the possible combinations by filling in boxes at the points of intersection.



Because there are two routes to derive Yy (Yy = yY), for the third generation (F_2), we should expect the following ratio for the offspring.

1 YY: 2 Yy: 1 yy

Since YY = yellow seed color, and Yy = yellow seed color, our expected ratio of

yellow-seeded to green-seeded offspring is

3 Yellow: 1 Green

In the case of seed color, this hypothesis predicts observed outcomes well enough that the Law of Segregation is accepted. Further, this law is now universally accepted for the inheritance of all traits that are determined by a single pair of alleles.

Some Important Definitions.

Alleles. Genes that are carried in pairs and which separate during the production of gametes. These typically affect a given trait in different ways. i.e. red vs, white flower color, round vs. wrinkled seeds etc.

Genotype. The specific combination of two alleles carried by an organism.

Phenotype. The observable expression of the genotype.

Trait. Is a character of an organism that can manifest alternate forms (phenotypes) which is determined by a pair of alleles (flower color, seed coat, stature...).

Dominant. If one allele masks the presence of another it is said to be dominant. Its symbol is often capitalized.

Recessive. When one allele is masked by he presence of another it is said to be recessive. Its symbol is often lowercase.

Incomplete Dominance. This is the situation where one allele doesn't completely mask the presence of the other, and we see a blending of phenotypes. In this case, the heterozygote has its own phenotype (see co-dominance).

Co-dominance. This is the situation where both alleles are fully expressed such as with the 'A' and 'B' antigens for human blood types. In this case the heterozygote also has its own phenotype (see incomplete dominance).

Homozygous. The condition where an organism carries copies of the same allele.

Heterozygous. The condition where an organism carries different alleles.

P generation. "P" comes from "parental. This is the generation from which we make our first cross.

F_1 generation. "F" comes from filial. The first set of progeny. These result from the cross made from the two parents of the P generation.

F_2 generation. The second set of progeny. These result from the cross made from the F_1 generation where two members of the F_1 are crossed.

Monohybrid Problems

Seed color in pea is determined by a pair of alleles,

Y = yellow seed color and **y = green seed color**.

1. What genotypes are possible for an unknown plant with yellow seeds?

_____

2. Given the following cross

Yy x yy

what will be the genotypes of the resulting offspring and in what ratio?

_____

Note. In problem two, each type of gamete produced by the heterozygous individual results in a unique genotype in the next generation. A cross made between an individual with the dominant phenotype, but with an unknown genotype (like in problem "1"), and an individual with a recessive phenotype is termed a **test cross.** The results of the cross indicated the genotype of the unknown plant.

Monohybrid problems where the heterozygote has a unique phenotype.

The presence of one allele does not always mask the presence of the other. If the phenotype of the heterozygote is distinctly different from the phenotypes of the two homozygotes, and this phenotype is a blend of the phenotypes of the two homozygotes, then the condition is termed **incomplete dominance** (also see co-dominance in the list of definitions)**.**

The phenotype for flower color of a certain plant in Iceland is determined by a pair of alleles

R = red flower color and **r = white flower color**

3. A homozygous plant with red flower color is crossed with a homozygous plant with white flowers. All the progeny are pink.

3a. What is the genotype of the progeny?

_____

3b. One of these pink-flowered progeny is self pollinated. Determine the genotypes and their ratios resulting from this cross.

_____

3c. Determine the phenotypes and their ratios resulting from the cross.

_____

Monohybrid Problems Involving Multiple Alleles

In the examples above, the phenotype of each trait was determined by the combination of only two possible alleles. However, sets of segregating genes in a population can include more than two alleles. but only two alleles are carried at a time in a diploid organism. An example is the "**A**", "**B**" and "**O**" blood types in humans. In this case there are three alleles, "**A**", "**B**", and "**O**". "**A**" together with "**B**" demonstrate **co-dominance** (the "**AB** "blood type). "**A**" and "**B**" are dominant when paired with "**O**" ("**AO**" = "**A**" blood type, and "**BO**" = "**B**" blood type).

4. If you have "B" blood type and your partner has "A" blood type what possible phenotypes can your children have?

_____

5. If you have "A" blood type, your sister has "B" blood type, and your mother has "O", what is the phenotype of your father?

_____

6. An anthropologist studying a population in a remote area of the Philippines discovers that everyone has "AB" blood type. Is it possible that this represents a people undisturbed by the modern culture around it?

   Speculate about why or why not ?

_____

_____

_____

_____

_____

II. Patterns of Inheritance. More than One Trait Considered

<u>The Law of Independent Assortment</u>. Another trait in peas studied by Gregor Mendel was seed texture. Some pure-breeding varieties of pea produced smooth seeds while others produced wrinkled seeds. In this case, he discovered that the allele for 'smooth' was dominant over that for 'wrinkled'. Further, he discovered that the alleles for 'seed texture', assorted themselves independently from the way the alleles for 'seed color' assorted themselves.

For example.

The alleles for seed color are **Y = yellow** **and** **y = green**

Those for seed texture are **W=smooth** **and** **w=wrinkled**

He observed that a plant with the following genotype

YyWw

produces four different gametes in equal numbers. During meiosis, a pollen grain or an egg cell with a **Y** it is equally likely to receive a **w** or a **W.** The same reasoning applies if a pollen grain or an egg cell receives a **y** gene, hence, this plant should produce eggs and pollen grains with the following genotypes in equal numbers.

YW: Yw: yW: yw

This can be determined by means of a **testcross.** In this case, the heterozygous plant is crossed with another that is homozygous for the recessive character of both traits. Every gamete produced by the heterozygous plant will be reflected in the phenotypic ratio of the resulting progeny - for example if we conducted the following cross

YyWw x yyww

and if we assume independent assortment, we would build the following punnet square.

	Yw	YW	y w	yW
y w	Yyww	YyWw	yyww	yyWw

We should expect the following phenotypic ratio of the resulting offspring.

1 yellow-wrinkled. 1 yellow-round. 1 green-wrinkled. 1 green-round

This prediction is consistent with Mendel's observations and we accept the rule in this case.

Dihybrid Problems

7. Consider the following cross.

YyWw x YyWw

a. Using the two Mendelian laws determine the possible gametes from each parent.
b. Use that information to set up a punnet square to determine the possible genotypes and their ratio for the resulting progeny.
c. Use that information to determine the possible phenotypes and their ratio.

7a. What gametes can be produced from an individual with the genotype **YyWw**?

_____

7b. What are the possible genotypes of the next generation and in what expected ratio?

_____

7c. What are the possible Phenotypes and in what expected ratio?

_____

Violations of the law of independent assortment

Mendel's second law only applies if the loci of the traits under consideration are on different chromosomes. If this so, then the genes from the different allelic groups sort themselves independently from one another because the chromosomes on which they reside sort themselves in a similar fashion during meiosis. If, however, the loci reside on the same chromosome, then the genes from the different allelic groups are physically bound together. This condition is called linkage. This usually results in the over representation of certain phenotypes in a frequency greater than expected.

In peas if **T**= tall shoots and **W** = red flowers
t = dwarf shoots **w**= white flowers

and if we were to make the following cross

P **TTWW x ttww**

All of the **F_1 =** **TtWw**

and, if we do an F_1 cross. **TtWw x TtWw**

We should expect a 9:3:3:1 phenotypic ratio in the F_2 generation.

That is a **9** dominant-dominant: **3** dominant-recessive: **3** recessive-dominant: **1** recessive-recessive

or

9 Tall-red: 3 Tall-white: 3 Dwarf-red: 1 Dwarf-white

This prediction assumes that the two allelic groups segregate themselves independently consistent with Mendel's second law.

One hypothetical outcome if these traits were linked with no crossing over, would be....

F_2 = 30 Tall-red: 10 Dwarf-white

Note. The predominance of the original parental phenotypes in a ratio similar to what would be expected by a monohybrid cross.

Redoing the **F_1** cross where we indicate linkage by drawing a line between genes carried on the same chromosome.....

F1 $\frac{T \quad W}{t \quad w}$ **X** $\frac{T \quad W}{t \quad w}$

It is clear that each **F_1** will not produce four different types of gametes as expected by Mendel's second law. Each **F_1** plant will produce only two types of gametes as we are working with only one pair of homologues. One with the genes TW and the other with the genes tw. Without crossing over, no other chromosomes are possible, and we must consider the assortment of these two chromosomes.

Now using a punnett square.

Pollen \ Eggs	TW	t w
TW	TTWW	TtWw
t w	tTwW	t t w w

It is clear why we are skewed towards a 3 .1 ratio.

In actual practice, one usually finds other phenotypic combinations because of crossing over between loci on homologous chromosomes.

So if we again consider the cross

F1 $\frac{T \quad W}{t \quad w}$ **X** $\frac{T \quad W}{t \quad w}$

A more typical outcome than

30 Tall-Red. 10 Dwarf-white,

would be.

25 Tall-red: 4 Tall-white: 3 Dwarf-red: 8 Dwarf-White

In this case, even though all four phenotypic combinations occur in the F_2, the results above should lead us to reject the assumption of independent assortment. With independent assortment, we anticipate a 9:3:3:1 phenotypic ratio, and, these numbers don't fit. Which leads us to conclude that the traits are linked. We can determine which progeny resulted from crossovers by considering the original parental genotypes, and deducing what phenotypes will appear in the F_2 in the absence of crossing-over. In this case, those progeny in the F_2 <u>not of the parental phenotypes</u> must have resulted from crossing over (tall-white and dwarf-red).

Problem 8. Consider the following cross

P TTww x ttWW

<u>Assuming linkage with no crossing over</u>, determine what gametes are produced from each parent, then, determine the genotype and phenotype of the F_1 population. Consider an F_1 cross TtWw x TtWw. Using a punnet square determine what genotypes will appear in the F_2 and in what ratio. Use this information to determine the expected phenotypic ratio for the F_2.

8a. What is the genotype and phenotype of the F_1 population?

8b. Consider the F_1 generation in the example problem (see page 8) and in this problem. Are they genetically different? If so how?

8c. What gametes can be produced by the F_1 plants in this problem assuming complete linkage and, hence, no crossing over.

8d. What are the expected genotypes and their ratio of the F_2 population assuming complete linkage and, hence, no crossing over.?

8e. In the F_2, for this problem, which phenotypes are impossible without crossing over? How is this different from the example problem?

III. Genetics Simulation Activity.

Introduction. In a perfect world we would conduct genetics experiments by crossing plants and raising progeny. In a fifteen week course, however, this is impractical. In this activity you will use a computer to simulate crosses between organisms. These choices will immediately provide you with progeny that can then be used for further crosses. The software (GCK toolkit) first provides you with a hypothetical population where you can observe some sort of variation. It is then your task to develop hypotheses, experiments (based on crosses) and conclusions about how these traits are passed on from one generation to the next. In some ways you will be repeating the process undertaken by Gregor Mendel in the nineteenth century.

Getting Started

Observable traits determined by a single pair of alleles are rare. Most traits are derived from the interactions of multiple allelic groups. These characters often display a continuous variation between extremes. One example of such a trait is height in humans. Traits determined by single allelic groups display a discrete variation. either red or white flowers, A, B, AB, or O blood type, wrinkled or smooth seeds. Mendel's success in determining the **Law of Segregation** was due to his good fortune in choosing traits that are derived from simple allelic groups. To understand this good fortune, reflect on the fact that in 1906 it took T.H. Morgan, working with fruit flies at Columbia University, over a year to discover one such trait, and Morgan was working with a team of graduate students. With our computer simulation you can assume that every trait is derived from a single set of alleles. The individuals provided represent a sub-sample of a simulated wild population. It is your task to develop and test a hypothesis that best explains the pattern of inheritance displayed.

Part One. Monohybrid Crosses - Problems 1 & 2

Procedure.

1. Produce pure breeding populations from your wild sample.

This is best done by crossing like members of the wild sample, then crossing like members of their progeny, and then crossing like members of theirs, and continuing the process until an alternate trait is eliminated.

The difficulties of developing a pure breeding strain should provide clues as to which allele is dominant (if either). Record the number of generations required to develop each strain and check off a preliminary hypothesis on the data sheet.

Once you achieve a generation all with the same phenotype, do you have a pure breeding strain?

_____

At this stage, continue the process for three more generations to ensure that the final generation is pure breeding.

2. Make a cross between members of the two pure-breeding strains. These two organisms are your Parental (P) generation.

The result of this cross will confirm if you are working with complete or incomplete dominance and if not which allele is dominant.

3. Based on the phenotypes of the F_1 generation you should form a final hypothesis. Using this hypothesis predict the phenotypes of the F_2 generation when a members of the F_1 are crossed.

Your hypothesis should include.

a. Statement that observed characters are determined by a single set of alleles (given). Assign symbols to represent the different genes.

b. Whether complete or incomplete dominance is displayed, and if complete, which allele is dominant, if incomplete what are phenotypes for each of the three genotypes.

c. Predict the phenotypes resulting from the F_1 cross and their relative ratios.

4. Cross members of the F_1 generation to generate an f_2 generation. Using the X^2 test given as part of the analysis menu of the program, determine if the outcome of this cross is close enough to your expected such that $p \geq .05$. If so you need not reject your hypothesis.

Note. Chi square is simply a method of quantifying how badly your observed outcome differs from your predicted results. For each phenotype we take the difference between what we predict and what we observe, square that value and divide by the expected. We take the resulting value for each phenotype and add them to arrive at a sum which is our chi square. Symbolically it is expressed as

$$\mathbf{X^2 = \sum (O-E)^2/E}$$

O = observed
E = Expected

This a chi square value can be converted into a "p" value using a table. For our purposes, the program has a built in table and will make this determination for you.

Part 2. Dihybrid Problem - No Linkage - Problem 3.

Procedure

1. Produce pure breeding populations from your wild sample.

You need to develop two strains that differ from each other for both traits.

Again this is best done by crossing like members of the wild sample, then crossing like members of their progeny, and then crossing like members of theirs, and continuing the process until alternate traits are eliminated. Once you achieve a generation all with the same phenotype, you should continue the process three more generations before assuming that you have a pure breeding strain?

2. Make a cross between members of the two different strains. These two strains are your Parental (P) generation.

The result of this cross will confirm which alleles are dominant and if you were successful in generating pure-breeding strains.

3. Based on the phenotypes of the F_1 generation you should form a final hypothesis. Using this hypothesis predict the phenotypes of the F_2 generation when a members of the F_1 are crossed.

- Your hypothesis should include.

a. Statement that each of the observed characters are determined by a single set of alleles (given). Assign symbols to represent the different genes.

b. Whether complete or incomplete dominance is displayed, and if complete, which alleles are dominant.

c. Predict the phenotypes resulting from the F1 cross and their relative ratios.

4. Cross members of the F1 generation to generate an f_2 generation. Then, using the X^2 test given as part of the analysis menu of the program, determine if the outcome of this cross is close enough to your expected such that $p \geq .05$. if so you need not reject your hypothesis.

Part 3. Dihybrid Problems with the Possibility of Linkage
(problems 4 and 5)

Procedure

1. Produce pure breeding populations from your wild sample.

You need to develop two strains that differ from each other for both traits.

Again this is best done by crossing like members of the wild sample, then crossing like members of their progeny, and then crossing like members of theirs, and continuing the process until alternate traits are eliminated. Once you achieve a generation all with the same phenotype, you should continue the process three more generations before assuming that you have a pure breeding strain?

2. Make a cross between members of the two different strains. These two strains are your Parental (P) generation.

The result of this cross will confirm which alleles are dominant and if you were successful in generating pure-breeding strains.

3. Based on the phenotypes of the F_1 generation you should form a final hypothesis. Using this hypothesis predict the phenotypes of the F_2 generation when a members of the F_1 are crossed.

- Your hypothesis should include.

a. Statement that each of the observed characters are determined by a single set of alleles (given). Assign symbols to represent the different genes.

b. Whether complete or incomplete dominance is displayed, and if complete, which alleles are dominant.

c. Predict the phenotypes resulting from the F1 cross and their relative ratios.

4. Cross members of the F1 generation to generate an f_2 generation. Then, using the X^2 test given as part of the analysis menu of the program, determine if the outcome of this cross is close enough to your expected such that $p \geq .05$. if so you need not reject your hypothesis.

5. If you reject your hypothesis, formulate a new hypothesis where you assume linkage.

6. Determine which phenotypes in the F_2 must have resulted from crossing over (looking back at the P phenotypes/genotypes will help you make this determination). Show how you determine the answer on the back of the work sheet!

Monohybrid Work Sheet **Problem # 1**

Trait _____ Fill in all observed phenotypes **_____ vs. _____ vs. _____**

True-Breeding Strain 1. Vial/Generation # _____
True-breeding Phenotype _____
Number of like-crosses required to develop strain ____

True-Breeding Strain 2. Vial/Generation # _____
True-breeding Phenotype _____
Number of like-crosses required to develop strain ____

Preliminary hypothesis. Check one. Incomplete dominance _____
Dominance _____

P cross. _____ X _____
Phenotype x Phenotype

F1 phenotype/s

Final Hypothesis

Expected phenotypes and ratio for the F_2 generation

Observed F_2 phenotypes

Phenotype	**# Present**	**# Expected**
_____	_____	_____
_____	_____	_____
_____	_____	_____

X^2 = _____ **P = _____**

Reject Hypothesis. **yes _____ no _____**

Monohybrid Work Sheet **Problem # 2**

Trait _____ Fill in all observed phenotypes **_____ vs. _____ vs. _____**

True-Breeding Strain 1. Vial/Generation # _____
True-breeding Phenotype _____
Number of like-crosses required to develop strain _____

True-Breeding Strain 2. Vial/Generation # _____
True-breeding Phenotype _____
Number of like-crosses required to develop strain _____

Preliminary hypothesis. Check one. Incomplete dominance _____
Dominance _____

P cross. _____ X _____
Phenotype x Phenotype

F_1 phenotype/s

Final Hypothesis

Expected phenotypes and ratio for the F_2 generation

_____

Observed F_2 phenotypes

Phenotype	**# Present**	**# Expected**
_____	_____	_____
_____	_____	_____
_____	_____	_____

X^2 = _____ **P = _____**

Reject Hypothesis. **yes _____ no _____**

Dihybrid Work Sheet **Problem # 3**

Trait _____ Fill in all observed phenotypes **_____ vs. _____ vs.**

Trait _____ Fill in all observed phenotypes **_____ vs. _____ vs.**

True-Breeding Strain 1. Vial/Generation # **_____**

Phenotype **_____ _____**

True-Breeding Strain 2. Vial/Generation # **_____**

Phenotype **_____ _____**

P cross. **_____** X **_____**
Phenotype **x** **Phenotype**

F1 phenotypes

<u>Final Hypothesis</u>

Expected phenotypes and ratio (assuming no linkage) for the F_2 generation

9 _____ : 3 _____ : 3 _____ : 1 _____

Observed F_2 phenotypes

Phenotypes	**# Present**	**# Expected**
_____	_____	_____
_____	_____	_____
_____	_____	_____
_____	_____	_____

X^2 = _____ **P = _____**

Reject Hypothesis. yes _____ no _____Dihybrid Work Sheet

Problem # 4

Trait _____ Fill in all observed phenotypes _____ **vs.** _____ **vs.**

Trait _____ Fill in all observed phenotypes _____ **vs.** _____ **vs.**

True-Breeding Strain 1. Vial/Generation # _____

Phenotype _____ _____

True-Breeding Strain 2. Vial/Generation # _____

Phenotype _____ _____

P cross. _____ X _____
Phenotype **x** **Phenotype**

F1 phenotypes

Final Hypothesis

Expected phenotypes and ratio (assuming no linkage) for the F2 generation

9 _____ : **3** _____ : **3** _____ : 1 _____

Observed F_2 phenotypes

Phenotypes	**# Present**	**# Expected**
_____	_____	_____
_____	_____	_____
_____	_____	_____
_____	_____	_____

X^2 = _____ **P =** _____

Reject Hypothesis. **yes** _____ (See the back page) **no** _____

If you reject your hypothesis, a compelling alternate hypothesis is that there is linkage. **Assume that the traits are linked** (that their loci are on the same chromosome), and deduce which phenotypic combinations are impossible in the F_2 without crossing-over. This is best done by determining which combinations are possible without crossing over.

1. What are the genotypes of the parents?

   $Parent_1$ _____ $Parent_2$ _____

2. What meiotic products can be produced by $parent_1$ _____

3. What meiotic products can be produced by $parent_2$ _____

4. Using a punnet square determine the genotypes of the F_2 generation.

4. What phenotypes do we expect in the F_2 generation without crossing over?

5. What phenotypes are impossible without crossing over?

Dihybrid Work Sheet **Problem # 5**

Trait _____ Fill in all observed phenotypes _____ **vs.** _____ **vs.**

Trait _____ Fill in all observed phenotypes _____ **vs.** _____ **vs.**

True-Breeding Strain 1. Vial/Generation # _____

Phenotype _____ _____

True-Breeding Strain 2. Vial/Generation # _____

Phenotype _____ _____

P cross. _____ X _____
Phenotype **x** **Phenotype**

F_1 phenotypes

Final Hypothesis

Expected phenotypes and ratio (assuming no linkage) for the F2 generation

9 _____ : **3** _____ : **3** _____ : 1 _____

Actual F_2 phenotypes resulting from the F1 cross

Phenotypes	**# Present**	**# Expected**
_____	_____	_____
_____	_____	_____
_____	_____	_____
_____	_____	_____

X^2 = _____ **P =** _____

Reject Hypothesis. **yes** _____ (see next page) **no** _____

If you reject your hypothesis, a compelling alternate hypothesis is that there is linkage. **Assume that the traits are linked** (that their loci are on the same chromosome), and deduce which phenotypic combinations are impossible in the F_2 without crossing-over. This is best done by determining which combinations are possible without crossing over.

1. What are the genotypes of the parents?

   $Parent_1$ _____ $Parent_2$ _____

2. What meiotic products can be produced by $parent_1$ _____

3. What meiotic products can be produced by $parent_2$ _____

4. Using a punnet square determine the genotypes of the F_2 generation.

4. What phenotypes do we expect in the F_2 generation without crossing over?

5. What phenotypes are impossible without crossing over?