

AP Biology Virtual Lab: Genetics of *Drosophila*

Overview

In this lab you will be doing virtual genetic crosses of fruit flies (*Drosophila melanogaster*). You will learn how to collect data from F1 and F2 generations and analyze the results from a monohybrid, dihybrid, or sex-linked cross.

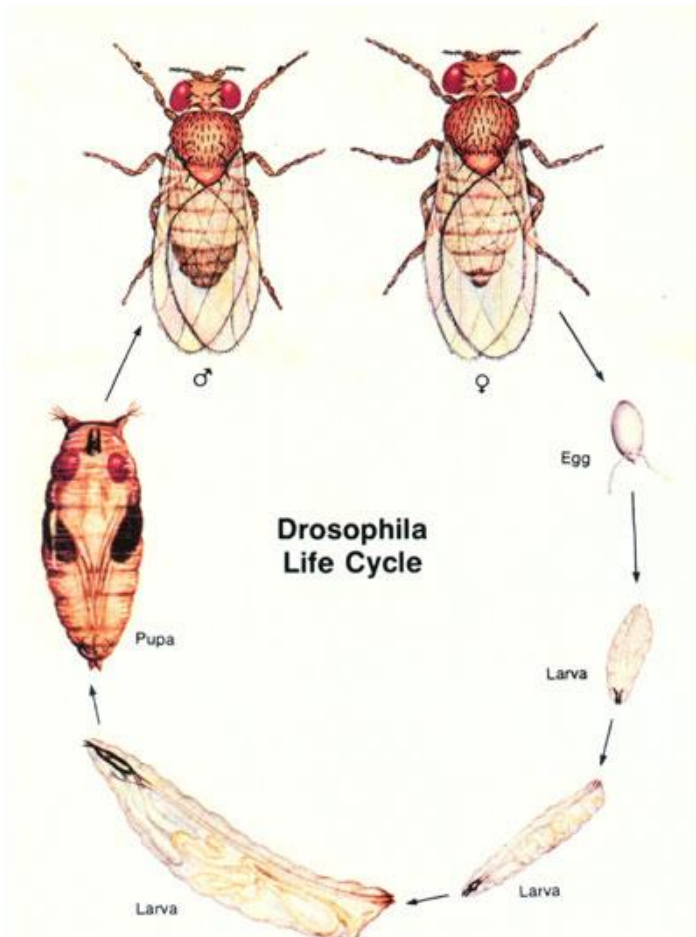
Objectives

- Use chi-square to analyze data
- Understand the life cycle of fruit flies
- Investigate the independent assortment of two genes and determine whether the two genes are autosomal or sex-linked using a multi-generation experiment

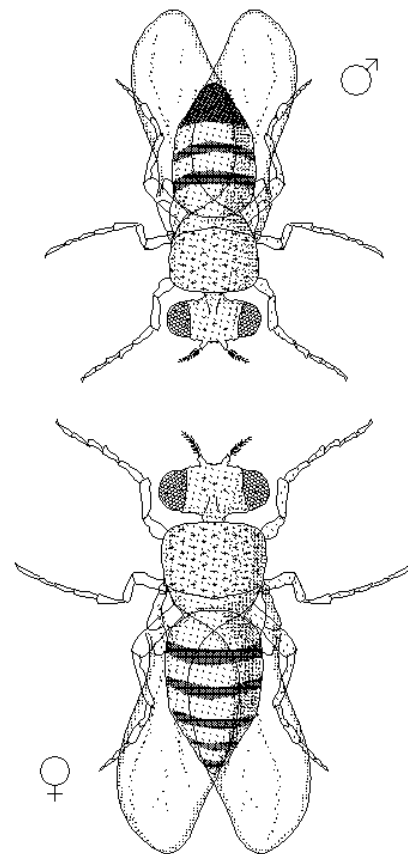
Introduction

The fruit fly is an excellent organism for genetic studies because it has simple food requirements, occupies little space, is hardy, completes its life cycle in about 12 days at room temperature, produces large numbers of offspring, can be immobilized easily for examination, and has many types of hereditary variations that can be observed with low-power magnification. Fruit flies have a small number of chromosomes (four pairs). Much research about the genetics of fruit flies during the last 50 years has resulted in a wealth of reference literature and knowledge about hundreds of its genes.

The *Drosophila* life cycle:



Male vs. female fruit flies:



Despite all of their advantages in genetics research, flies are an enormous pain to work with – especially if you are new at it. So, let’s use the benefits of modern technology and perform our lab on the computer using virtual flies.

Directions

1. Go to <http://www.sciencecourseware.org/vcise/drosophila/>
2. Click the “Enter as a Guest” option
3. Do not back out of this page until you are completely done – or you will have to start over!
4. Order your fruit flies. Use the “Order Flies” icon. We are going to perform three different crosses. For each one, you will need to order the correct flies:
 - a. Cross 1: Cross a sepia eye male with a wild type female
 - b. Cross 2: Cross a sepia eye, normal wing male with a wild type eye, vestigial wing female
 - c. Cross 3: Cross a wild type male with a white eye female
5. After ordering your flies, you will have to work through the lab until you eventually have sorted your F1 progeny. Make sure you record all of the necessary data in your data table.
6. After you have recorded the data, you will need to establish a cross between members of your F1 generation. To do this, click on the piles of sorted flies to zoom in. Then click the “Use in New Mating” button. Do this for both a male and a female. Then return to your lab. Click on the mating jar.
7. Sort your progeny and record the necessary data in the F2 data table.
8. After you collect F2 data, clean out your incubator by clicking on the small trash can next to it. This resets your lab for the next cross.
9. Now that you have the F2 data, you can begin to formulate some conclusions about the mode of inheritance for the traits you are looking at.
10. Perform the other two crosses. To order new flies, click on the computer and repeat.
11. When you have performed all of the crosses, move to the analysis portion of the lab.

DATA TABLES

Cross 1: P generation = sepia male * wild type female

Table 1: Phenotypes of F1 Generation

Phenotype	Number of Males	Number of Females

Table 2: Phenotypes of F2 Generation

Phenotype	Number of Males	Number of Females

Hypothesize the mode of inheritance of the sepia eye trait: _____
 Is this a monohybrid or dihybrid cross? _____

Cross 2: P generation = sepia-eye, normal-wing male * wild type, vestigial wing female

Table 1: Phenotypes of F1 Generation

Phenotype	Number of Males	Number of Females

Table 2: Phenotypes of F2 Generation

Phenotype	Number of Males	Number of Females

Hypothesize the mode of inheritance of the sepia eye trait: _____

Hypothesize the mode of inheritance of the vestigial wing trait: _____

Do you think the two traits are linked? _____

Is this a monohybrid or dihybrid cross? _____

Cross 3: P generation = wild type male * white eye female

Table 1: Phenotypes of F1 Generation

Phenotype	Number of Males	Number of Females

Table 2: Phenotypes of F2 Generation

Phenotype	Number of Males	Number of Females

Hypothesize the mode of inheritance of the white eye trait: _____

Is this a monohybrid or dihybrid cross? _____

DATA ANALYSIS

1. In the space below, construct two Punnett squares for each cross to predict the expected results of both the parental and F1 crosses from your null hypothesis.

Cross 1: P generation = sepia male * wild type female

Parental Cross	F1 Cross

Cross 2: P generation = sepia-eye, normal-wing male * wild type, vestigial wing female

Parental Cross	F1 Cross

Cross 3: P generation = wild type male * white eye female

Parental Cross	F1 Cross

2. Refer to the Punnett Squares above. In the boxes below, record the **expected** ratios for the genotypes and phenotypes of the F1 and F2 generations in the experiment.

Cross 1: P generation = sepia male * wild type female

	Expected Genotype Ratio	Expected Phenotype Ratio
F1		
F2		

Cross 2: P generation = sepia-eye, normal-wing male * wild type, vestigial wing female

	Expected Genotype Ratio	Expected Phenotype Ratio
F1		
F2		

Cross 3: P generation = wild type male * white eye female

	Expected Genotype Ratio	Expected Phenotype Ratio
F1		
F2		

3. Do the observed results deviate from your expected results? If so, explain how:

CHI-SQUARE ANALYSIS

Statistics can be used to determine if differences among groups are **significant**. The statistical test most frequently used to determine whether data obtained experimentally provide a good fit to the expected data is the chi-square test. This test can be used to determine if deviations from the expected values are due to chance alone, or to some other circumstance.

To determine if the observed data fall within acceptable limits, a chi-square analysis is performed to test the validity of the **null hypothesis** (that there is no statistically significant difference between the observed and expected data). If the chi-square analysis indicates that the observed data vary too much from the expected data, an **alternate hypothesis** is accepted.

The formula for chi-square is:

$$\chi^2 = \sum \frac{(o-e)^2}{e}$$

where o = observed number of individuals

e = expected number of individuals

Σ = the sum of the values (in this case, the differences, squared, divided by the number expected)

How to Use the Chi-Square Distribution Table

1. Determine the **degrees of freedom (df)** for your experiment. It is the number of observed categories minus one. For example, if there are two phenotypes, the would be one degree of freedom.
2. To use the table (at the end of this lab), we have to determine the α (acceptable error rate, α). For the purposes of these experiments, assume that α equals 0.05 (5%). In other words, we will be 95% confident in our conclusion and we accept that there is a 5% chance that the conclusion will be wrong.
3. Find the p value. Find the number under your df column and $p = 0.05$ row. For example, for 1 df, the critical value is 3.84. What does this mean? **If the calculated chi-square value is less than the critical value, then we fail to reject our null hypothesis and conclude that there were no differences in the observed and predicted numbers. If the calculated chi-square value is greater than or equal to the critical value from the table, then we reject our hypothesis and conclude with 95% certainty that there was a statistically significant difference between expected and observed numbers.** If you reject your null hypothesis, that means that chance alone cannot explain the difference between observed and expected results and there is reason to doubt your original hypothesis (or to question data collection accuracy.)

Calculate the chi-square value for your data and determine whether to reject or fail to reject your null hypothesis:

Cross 1: P generation = sepia male * wild type female

Phenotype	Observed	Expected	(o-e)	(o-e) ² /e

$$\chi^2 = \underline{\hspace{2cm}}$$

How many degrees of freedom are there? _____

What is the critical value? _____

Do you accept or reject your H_0 ?

Cross 2: P generation = sepia-eye, normal-wing male * wild type, vestigial wing female

Phenotype	Observed	Expected	(o-e)	(o-e) ² /e

$$\chi^2 = \underline{\hspace{2cm}}$$

How many degrees of freedom are there? _____

What is the critical value? _____

Do you accept or reject your H_0 ? _____

Cross 3: P generation = wild type male * white eye female

Phenotype	Observed	Expected	(o-e)	(o-e) ² /e

$$\chi^2 = \underline{\hspace{2cm}}$$

How many degrees of freedom are there? _____

What is the critical value? _____

Do you accept or reject your H_0 ? _____

POST-LAB QUESTIONS

1. Why are fruit flies excellent organisms for genetic studies?
2. Describe the fruit fly life cycle.
3. What are the differences between male and female fruit flies?
4. Please describe the following mutations (how are they different from wild type?):
 - a. White eye
 - b. Vestigial wing
 - c. Sepia eye
5. What is the null hypothesis?
6. Why is the chi-square test needed?
7. What is the equation for the chi-square test?
8. How do you determine the degrees of freedom?
9. What p-value did you use, and what does it mean?