

LABORATORY 4, EXERCISE 3. CHROMOSOME LOCATION

Purpose

The purpose of this Laboratory is to use what you have learned up to this point to determine the chromosome location of three mutant alleles. Be sure to keep careful records as you perform this exercise. You will be expected to turn in a Lab Report based on the location of your mutant alleles defining not only their chromosomal location, but the logic in how that was determined.

Introduction

Fruit Flies are ideal models for learning the basics of Chromosome Location Analyses. Because their genome is divided into only 4 Chromosome Pairs ($2N = 8$; 3 autosome and 1 sex chromosome pairs), locating Fruit Fly genes is a matter of simple elimination using basic Mendelian crosses.

First, we need markers to uniquely identify 3 of the 4 chromosomes. Fortunately, so many loci have been mapped for Fruit Flies that selecting markers is an easy task. For Chromosome I, we will use the knowledge that this is the Sex Chromosome. If a locus is found to be Sex-Linked (*Reciprocal Crosses give different results*), then by definition the locus is located (Linked) on Chromosome I. For Chromosomes II and III, we will use autosomal loci with easy to detect phenotypes (Color, Wing Shape etc.). As in Exercise 1, if the expected Dihybrid Ratio (1:1:1:1) is rejected then we have evidence that the Mutant is Linked on the Same Chromosome as the Marker. Failure to detect a departure from the expected indicates independent assortment and that the trait is not closely linked to the Marker.

For this exercise we will take this as evidence that the two loci are not on the same chromosome. If a Mutant locus is not Sex-linked and Assorts Independently from both Markers, we will assume that it is located on the last chromosome pair (Chromosome IV). Remember that Chromosome IV is a very small chromosome in Fruit Flies and thus carries only a few genes. You will use the Dichotomous Key below to find the location of assigned mutants.

Chromosome Location Key

A. Evidence of Sex-linkage

→ YES = Chromosome 1

→ NO = Go to B

B. Assorts Independently from Black Body

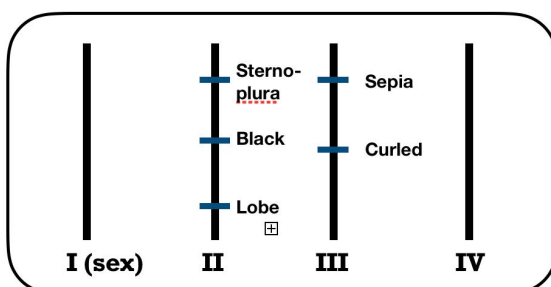
→ NO = Chromosome 2

→ YES = Go to C

C. Assorts Independently from Curled Wing

→ NO = Chromosome 3

→ YES = Chromosome 4



Exercise Protocol

1. Launch the Fly Lab Colony (<https://cws.auburn.edu/FlyLab>) and open a LINKAGE cross. You will use both the ADVANCED and the LINKAGE MODULES. For Sex-Linkage, use the ADVANCED module. Use the LINKAGE module for all other crosses.

2. Obtain a MUTANT TRAIT ASSIGNMENT SHEET from your TA. Record your unknown traits below.

Trait 1:		Location:	
Trait 2:		Location:	
Trait 3:		Location:	

3. Select the number of offspring you wish to produce and determine the Mode of Inheritance for your first mutant. Keep in mind it is always a good idea to CONFIRM your results! You may not want to simply rely on a single cross. If your first cross indicates SEX-LINKAGE, maybe do the second and third crosses just to confirm the results?!

4. Conduct the appropriate matings to determine if your mutant is Sex Linked or Autosomal (*Cross 1*).

- Assign your mutant phenotype to one parent and generate F2 data
- Perform the RECIPROCAL CROSS for your mutant.

5. Perform a dihybrid cross using your mutant and a marker for Chromosome #2 (*Cross 2*).

- Determine the expected Assortment Ratio.
- Use a Chi-Squared analysis to test for assortment.

6. Perform a dihybrid cross using your mutant and a marker for Chromosome #3 (*Cross 3*).

- Determine the expected Assortment Ratio.
- Use a Chi-Squared analysis to test for assortment.

7. Use the Chromosome Location Key provided in the Introduction to identify the location for each of the three mutants you were assigned. Keep in mind, it may be necessary to select an alternate marker for any particular chromosome than your first choice. (**Note – before you walk out of this lab, be able to answer WHY that may be case!! If you and lab mates cannot reason it out, check with your TA.**). You may also make use of traits that may have already been located by other groups. So don't be afraid to interact with others in your lab. If all else fails, ask your TA for advice.

8. Once you have localized your first trait to its specific chromosome, repeat Steps 4-6 with your other two traits and make a note of where they are located. (HINT – you may find the location information of your unknown traits useful in succeeding laboratory assignments!!)

9. Write and submit a Laboratory Report detailing your results for this exercise. Be sure to follow the format and instructions for laboratory reports you have done previously this semester. Be complete but be concise.

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 1:

Parentals

Male

Female

Phenotype

x

Genotype

X

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Parentals

Male

Female

Phenotype

x

Genotype

X

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

Chi-Squared Test Reciprocal Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 1:

CROSS 2 DIAGRAM

Parentals		Male		Female
Phenotype		x		
Genotype		x		
F1 Generation				
	Total	Male	Total	Female
Phenotype				
Genotype				
F2 Generation				
	Total	Sex	Phenotype	Genotype

Chi-Squared Test Cross 2

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value =

Degrees of Freedom (*df*) =

Table Value (0.05) =

Overall Conclusion =

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 1:

CROSS 3 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

x

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 3

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

TRAIT 1 CHROMOSOME LOCATION:

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

NAME:

TRAIT 2:

CROSS 1 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

x

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

RECIPROCAL CROSS 1 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

x

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

Chi-Squared Test Reciprocal Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 2:

CROSS 2 DIAGRAM

Parentals

	Male		Female
Phenotype		x	
Genotype		x	

F1 Generation

	Total	Male		Total	Female
Phenotype					
Genotype					

F2 Generation

Total	Sex	Phenotype	Genotype

Chi-Squared Test Cross 2

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value =

Degrees of Freedom (*df*) =

Table Value (0.05) =

Overall Conclusion =

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 2:

CROSS 3 DIAGRAM

Parentals		Male			Female	
Phenotype			x			
Genotype			x			
F1 Generation		Total	Male		Total	Female
Phenotype						
Genotype						
F2 Generation		Total	Sex	Phenotype		Genotype

Chi-Squared Test Cross 3

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value	=	
Degrees of Freedom (<i>df</i>)	=	
Table Value (0.05)	=	
Overall Conclusion	=	

TRAIT 2 CHROMOSOME LOCATION:

LAB 4 EXERCISE 3 - DATA SHEET

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X

X

Genotype

Chi-Squared Test Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

Chi-Squared Test Reciprocal Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 3:

CROSS 2 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

x

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 2

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET

TRAIT 3:

CROSS 3 DIAGRAM

Parentals	Male			Female	
Phenotype		x			
Genotype		x			

F1 Generation	Total	Male	Total	Female
Phenotype				
Genotype				

F2 Generation	Total	Sex	Phenotype	Genotype

Chi-Squared Test Cross 3

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value	=	
Degrees of Freedom (<i>df</i>)	=	
Table Value (0.05)	=	
Overall Conclusion	=	

TRAIT 3 CHROMOSOME LOCATION:

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET (if needed)

NAME:

TRAIT X:

CROSS 1 DIAGRAM

Parents

Male

Female

Phenotype

X

Genotype

X

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

RECIPROCAL CROSS 1 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

X

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

Chi-Squared Test Reciprocal Cross 1

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET *(if needed)*

TRAIT X:

CROSS 2 DIAGRAM

Parentals		Male		Female
Phenotype		x		
Genotype		x		

F1 Generation	Total	Male	Total	Female
Phenotype				
Genotype				

F2 Generation	Total	Sex	Phenotype	Genotype

Chi-Squared Test Cross 2

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value =

Degrees of Freedom (*df*) =

Table Value (0.05) =

Overall Conclusion =

GENETIC CROSS

LAB 4 EXERCISE 3 - DATA SHEET *(if needed)*

TRAIT x:

CROSS 3 DIAGRAM

Parentals

Male

Female

Phenotype

x

Genotype

x

F1 Generation

Total

Male

Total

Female

Phenotype

Genotype

F2 Generation

Total

Sex

Phenotype

Genotype

Chi-Squared Test Cross 3

Phenotype	Observed	Expected	O - E	(O - E) ²	(O - E) ² / E
TOTAL					

Observed Chi – Squared Value

=

Degrees of Freedom (*df*)

=

Table Value (0.05)

=

Overall Conclusion

=