Name:	
Date:	Pd:

Objectives

Before doing this lab you should understand:

Chi-square analysis of data & the life cycle of diploid organisms useful in genetics studies.

After doing this lab you should be able to:

Investigate the independent assortment of two genes and determine whether the two genes are autosomal or sex-linked using a multigenerational experiment, and analyze the data from your genetic crosses using chisquare analysis techniques.

Statistical Analysis Section - Example 1

Statistics can be used to determine if differences among groups are significant, or simply the result of predictable error. The statistical test most frequently used to determine whether data obtained experimentally provide a good fit or approximation to the expected or theoretical 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. For example, consider corn seedlings resulting from an F1 cross between parents that are heterozygous for color.

A Punnett square of the F1 cross Gg x Gg would predict the expected proportion of green: albino seedlings would be 3:1. Use this information to fill in the Expected (e) column and the (o-e) column in Table 7.1.

Table 7.1

Phenotype	Genotype	Observed (o)	Expected (e)	(o-e)
Green	GG or Gg	72		
Albino	gg	12		
	Total:	84		

There is a small difference between the observed and expected results, but are these data close enough that the difference can be explained by random chance or variation in the sample?

To determine if the observed data fall within the acceptable limits, a chi-square analysis is performed to test the validity of a **null hypothesis** (that there is no statistically significant difference between the observed and expected data). If the chi-square analysis indicates that the data vary too much from the expected 3:1, an **alternative hypothesis** is accepted (or you fail to reject the null).

The formula for chi-square is

$$\chi^2 = \sum \left(o - e \right)^2$$

where o = observed number of individuals e = expected number of individuals

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

Critical Values Table

		De	grees of Freedom (df)	
Probability (p)	1	2	3	4	5
0.05	3.84	5.99	7.82	9.49	11.1
0.01	6.64	9.21	11.3	13.2	15.1
0.001	10.8	13.8	16.3	18.5	20.5

- 1. This statistical test will examine the null hypothesis, which predicts that the data from the experimental cross above will be expected to fit the 3:1 ratio.
- 2. Use the data from Table 7.1 to complete table 7.2.

Table 7.2

Phenotype	Observed (o)	Expected (e)	(o-e)	(o-e) ²	(o-e) ² e
Green	72				
Albino	12				

Degrees of Freedom:	X ² value =

Conclusion:

Example 2

In a study of incomplete dominance in tobacco seedlings, the counts in table 7.3 were made from a cross between two heterozygous plants. Draw the Punnett square in the box to the right.

Table 7.3

Phenotype	Observed (o)	Expected (e)	(o-e)	(o-e) ²	(o-e) ²
Green	22				
Yellow Green	50				
Albino	12				

Degrees of Freedom: _____ X² value = _____

A Punnett square for this cross indicates an expected ratio of 1 green:2 yellow green:1 albino. Calculate the expected for each and fill out the Table 7.3.

According to the critical value of χ^2 , can you accept or reject the null hypothesis (does the data fit the expected 1:2:1 ratio?)

Practice Problem

An investigator observes that when pure-breeding, long-winged fruit flies are mated with pure-breeding, short-winged flies, the F_1 offspring have an intermediate wing length. When several intermediate-winged flies are allowed to interbreed, the following results are obtained:

230 long-winged flies, 510 intermediate winged flies, 260 short winged flies.

- a. What is the genotype of the F₁ intermediate-wing-length flies?
- b. Write a hypothesis describing the mode of inheritance of wing length in fruit flies (this is your null hypothesis).
- c. Complete Table 7.4

Table 7.4

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

Degrees of Freedom:	X ² value =	

g. Can you accept or reject the null hypothesis? Explain why.

Procedure: Monohybrid Cross

- 1. Go to the website: http://www.sciencecourseware.com/vcise/drosophila/ and enter as a guest.
- 2. "Order" a pair of flies: a male wild type for wing size and a female with vestigial wing size. Put them in your "Shopping Cart". Check out.
- 3. Breed the flies, click on the "Mating Jar" and record the Data in Table 7.5.
- 4. Sort flies and then send data to computer.
- 5. Click on analyze results and record your data below. Do not click ignore sex!

Table 7.5-F₁ Generation

Phenotype and Symbol	# Females	# Males

- 6. Return to Menu, then return to lab, click on the incubator, and then click on the F1 generation mating jar.
- 7. Zoom in on the pile of females and then click on use in new mating, and then zoom out.
- 8. Zoom in on the pile of males and then click on use in new mating, and then zoom out.
- 9. Click return to lab.
- 10. Click the mating jar to mate the flies.
- 11. Breed these flies and record the F₂ generation data in Table 7.6.

Table 7.6-F₂ Generation

Phenotype and Symbol	Females	Males

- 12. Write a hypothesis that describes the mode of inheritance of the trait you studied. This is your null hypothesis.
- 13. Construct Punnett squares to predict the expected results of both parental and F₁ generational crosses from your null hypothesis.

F ₁ Cross	
-	F ₁ Cross

		Expected Genotypi	c Ratio	Expected Phenoty	ypic Ratio
F ₁ generation					
F ₂ generation					
5. From the result	ts, describe your cro	DSS.			
	ition sex-linked or a				
	ition dominant or re				
10 1110 111010					
6. Perform a chi-s	quare test on your	F ₂ results to see if the	e deviations are	within limits expecte	d by chance.
Phenotype	Observed (o)	Expected (e)	(o-e)	(o-e) ²	(o-e) ²
					е
	Degrees of Fre	edom:	X ²	² value =	
7. Can you accept	or reject your null l	hypothesis? Explain	why.		
			-		
rocedure: Monoh	vbrid Cross				
	-	sh" to clear the lab o	of flies. (Click on	the trash can to do th	nis).
. Return to the la	ab and click the "tra		•	the trash can to do the vellow body. Check o	-
. Return to the la . "Order" a pair	ab and click the "tra of flies: a male wild	type body color and	a female with a	yellow body. Check o	-
. Return to the la . "Order" a pair	ab and click the "tra of flies: a male wild		a female with a	yellow body. Check o	-
Return to the la "Order" a pair of Breed the flies,	ab and click the "tra of flies: a male wild click on the "Matin	type body color and	a female with a	yellow body. Check o	-
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin ation	type body color and	a female with a e Data in Table	yellow body. Check o	-
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin	type body color and g Jar" and record the	a female with a e Data in Table	yellow body. Check o	out
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin ation	type body color and g Jar" and record the	a female with a e Data in Table	yellow body. Check o	out
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin ation	type body color and g Jar" and record the	a female with a e Data in Table	yellow body. Check o	out
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin ation	type body color and g Jar" and record the	a female with a e Data in Table	yellow body. Check o	out
 Return to the land "Order" a pair of the flies, Breed the flies, 	ab and click the "tra of flies: a male wild click on the "Matin ation	type body color and g Jar" and record the	a female with a e Data in Table	yellow body. Check o	out
. Return to the la . "Order" a pair of . Breed the flies, able 7.7-F ₁ General Phenotype	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol	type body color and g Jar" and record the Fema	a female with a e Data in Table i	yellow body. Check of 7.7. Ma	ales
. Return to the la . "Order" a pair of the flies, . Breed the flies, able 7.7-F ₁ General Phenotype	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol	type body color and g Jar" and record the Fema	a female with a e Data in Table i	yellow body. Check o	ales
. Return to the la . "Order" a pair of the flies, . Breed the flies, able 7.7-F ₁ General Phenotype	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol	type body color and g Jar" and record the Fema	a female with a e Data in Table i	yellow body. Check of 7.7. Ma	ales
. Return to the late of the late of the flies, able 7.7-F1 General Phenotype . Add one female the F2 generation	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table i	yellow body. Check of 7.7. Ma	ales
. Return to the la . "Order" a pair of the flies, able 7.7-F ₁ General of the flies, able 7.7-F ₂ Generation able 7.8-F ₂ Generation	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
. Return to the la . "Order" a pair of the flies, able 7.7-F ₁ Generation of the flies of the flies of the F ₂ generation of the F ₂	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales
. Return to the la . "Order" a pair of the flies, able 7.7-F ₁ General Phenotype . Add one femals the F ₂ generation able 7.8-F ₂ Generation able 7.8-F ₂ Generation in the F ₂ Generation able 7.8-F ₂ Generation in the F ₂ Generation able 7.8-F ₂ Generation in the F ₃ Generation in the F ₄ Generation in the F	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
Return to the land of the land of the flies, and the flies, and the flies, and the F ₂ generation of the F ₂	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
Return to the land of the land of the flies, and the flies, and the flies, and the F ₂ generation of the F ₂	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
. "Order" a pair of the flies, able 7.7-F ₁ Generally able 7.8-F ₂ Generally able 7.8-F ₂ Generally able 7.8-F ₂ Generally	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
Return to the land of the land of the flies, and the flies, and the flies, and the F ₂ generation of the F ₂	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and
. Return to the la . "Order" a pair of the flies, able 7.7-F ₁ General of the flies, able 7.7-F ₂ Generation able 7.8-F ₂ Generation	ab and click the "tra of flies: a male wild click on the "Matin ation and Symbol e and one male from on data in Table 7.8	type body color and g Jar" and record the Fema	a female with a e Data in Table in Tabl	yellow body. Check of 7.7. Ma to a new mating jar. E	ales Breed them and

6. Construct Punnett squares to predict the expected results of both parental and F₁ generational crosses from your null hypothesis.

Parental Cross			F ₁ Cross	·	
Pofor to the Di	innott causros shov	vo. In the table below	rocard the ave	acted ratios for the	sonotypes and
	•	ve. In the table below es in the experiment.	, record the exp	ratios for the g	genotypes and
p		so in the experiment			
		Expected Genotypi	c Ratio	Expected Phenot	ypic Ratio
1 generation					
generation					
	ts, describe your cr				
	ation sex-linked or a				
Is the muta	ation dominant or r	ecessive?			
Perform a chi-s	square test on your	F ₂ results to see if the	e deviations are	within limits expecte	ed by chance.
Phenotype	Observed (o)	Expected (e)	(o-e)	(o-e) ²	(o-e) ²
					е
	Dograas of Er	andom:	V	² value =	
	Degrees of Fre	eedom:	^	value –	

Procedure: Dihybrid Cross

- 1. Return to the lab and click the "trash" to clear the lab of flies.
- 2. Order a wild type female and order a male with sepia eyes and black body.
- 3. Breed the flies, click on the "Mating Jar" and record the Data in Table 7.9.

Table 7.9-F₁ Generation

Phenotype and Symbol	Females	Males

4. Add one female and one male from the F₁ Generation and put them into a new mating jar. Breed them and record the F_2 generation data in Table 7.10

Table 7.10-F₂ Ge	eneration		
_			

Phenotype and Symbol	Fem	ales	N	1ales
Write a null hypothesis that des	cribes the mode of inh	neritance of the t	raits you studied.	
Construct Punnett squares to pr	edict the expected res	sults of both pare	ental and F ₁ generati	onal crosses from
null hypothesis.		F ₁ Cross		
Pofor to the Duppett squares ab	ovo. In the table below	w record the eve	acted ratios for the	gonotypos and
			ected ratios for the	genotypes and
Refer to the Punnett squares ab phenotypes of the F_1 and F_2 cross			ected ratios for the particles	
phenotypes of the F ₁ and F ₂ cross	ses in the experiment			
phenotypes of the F ₁ and F ₂ cross generation	ses in the experiment			
phenotypes of the F ₁ and F ₂ cross generation	ses in the experiment			
phenotypes of the F ₁ and F ₂ cross generation generation From the results, describe your	Expected Genotype			
generation generation From the results, describe your Are the mutations sex-linker	Expected Genotype cross.			
phenotypes of the F ₁ and F ₂ cross generation generation From the results, describe your	Expected Genotype cross.			
generation generation From the results, describe your Are the mutations dominant	Expected Genotype cross. d or autosomal?	cic Ratio	Expected Pheno	typic Ratio
generation generation From the results, describe your Are the mutations dominant	Expected Genotype cross. d or autosomal?	cic Ratio	Expected Pheno	typic Ratio
generation generation From the results, describe your Are the mutations sex-linked Are the mutations dominant	Expected Genotype Expected Genotype cross. d or autosomal? c or recessive?	ne deviations are	Expected Pheno	typic Ratio
phenotypes of the F ₁ and F ₂ cross generation generation From the results, describe your Are the mutations sex-linked Are the mutations dominant Perform a chi-square test on you	Expected Genotype Expected Genotype cross. d or autosomal? c or recessive?	ne deviations are	Expected Pheno	typic Ratio
phenotypes of the F ₁ and F ₂ cross generation generation From the results, describe your Are the mutations sex-linked Are the mutations dominant Perform a chi-square test on you	Expected Genotype Expected Genotype cross. d or autosomal? c or recessive?	ne deviations are	Expected Pheno	typic Ratio

Degrees of Freedom: _____ X² value = _____

10. Can you accept or reject your null hypothesis? Explain why?