Studying the patterns of inheritance when the mutation is lethal, when the phenomenon of epistasis is occurring between two gene pairs and when the mutation is linked to a sex chromosome (X chromosome)

Devina Chaudhary
7333619

Lab partner
Savannah Turon

Section C6

March 6th,2015

Lab demos
Cloe and MaryAnn

Department of Biology

University of Ottawa

INTRODUCTION

Part 1: Lethal mutations

Question: What is the pattern of inheritance in a monohybrid cross with a lethal mutation?

Hypothesis: A cross between two parents that have different alleles (one that expresses the mutated trait and one that expresses the wild type trait) at one gene will produce offspring following Mendel's first law, the law of segregation.

Assumption: Since the homozygous genotype of the lethal mutation would lead to the death of fruit flies, any parents that carry the lethal mutation have to be heterozygous for the studied character. Both male and female flies have different lethal mutations. The parents are true-breeding individuals (pure-bred).

Ar= aristapedia antennae

1) **Predicted phenotypic ratio of the F_1 generation (parental cross)**

Phenotype of parents: ♀ AR x ♂ +
Genotype of parents: ♀ARAR x ♂++

♀ ♂	+
AR	AR+

Predicted phenotypic ratio of F_1: 1+ : 1AR

2) **Predicted phenotypic ratio of the F_2 generation (F_1 cross)**

F_1 cross: ♀ + x ♂ + (♀ AR+ x ♂ AR+)

♀ ♂	**AR**	+
AR	ARAR	AR+
+	AR+	++

Predicted phenotypic ratio of F_2: 1+ : 2AR

L2:

Question: What is the pattern of inheritance in a dihybrid cross with a lethal mutation?

Hypothesis: A cross between two parents that have different alleles (one that expresses the mutated trait and one that expresses the wild type trait) at two genes will produce offspring following Mendel's first law the law of segregation and second law, law of independent assortment.

Assumption: One gene does not exert an influence on the other gene and the parents are pure-bred homozygous for both traits. The two genes are not sex-linked (. not linked to a sex chromosome).

ST=star eyes SB=stubble bristle

1) Predicted phenotypic ratio of the F_1 generation (parental cross)

Phenotype of parents: ♀ ST,+ x ♂ +, SB
Genotype of parents: ♀STST, ++ x ♂++, SBSB

♀ ♂	**+, SB**
ST, +	ST+, +SB

Predicted phenotypic ratio of F_1: 1+ : 1SB : 1 ST : 1SB;ST

2) Predicted phenotypic ratio of the F_2 generation (F_1 cross)

F_1 cross: ♀ STSB x ♂ STSB (♀STST, SBSB x ♂STST, SBSB)

♀ ♂	**ST, SB**	**ST, +**	**+, SB**	**+, +**
ST, SB	STST, SBSB	STST, SB+	ST+, SBSB	ST+, SB+
ST, +	STST, SB+	STST, ++	ST+, SB+	ST+, ++
+, SB	ST+, SBSB	ST+, SB+	++, SBSB	++,SB+
+, +	ST+, SB+	ST+, ++	++, SB+	++, ++

Predicted phenotypic ratio of F_2: 1+ : 2SB : 2ST : 4SB;ST

Part 2: Epistasis

Question: What is the pattern of inheritance in a dihybrid cross with epistatic and hypostatic genes?

Hypothesis: A cross between two parents that have different alleles (one that will be expressed, the epistatic genes and the other genes will be repressed and therefor hypostatic).

Assumption: One gene does exert an influence on the other gene and the parents are pure-bred homozygous for both traits. The two genes are not sex-linked (not linked to a sex chromosome).

EY=eye-less eyes BW= brown eyes

1) **Predicted phenotypic ratio of the F_1 generation (parental cross)**

Phenotype of parents: ♀ EY,+ x ♂ BW,+
Genotype of parents: ♀EYEY, ++ x ♂BWBW, ++

♂ ♀	**+, BW**
EY, +	EY+, BW+

Predicted phenotypic ratio of F_1: 100%+

2) **Predicted phenotypic ratio of the F_2 generation (F_1 cross)**

F_1 cross: ♀ ++ x ♂ ++ (♀+EY, +BW x ♂+EY, +BW)

♂ ♀	**EY,BW**	**EY, +**	**+, BW**	**+, +**
EY, BW	EYEY, BWBW	EYEY, BW+	EY+, BWBW	EY+, BW+
EY, +	EYEY, BW+	EYEY, ++	EY+, BW+	EY+, ++
+, BW	EY+, BWBW	EY+, BW+	++, BWBW	++, BW+
+, +	EY+, BW+	EY+, ++	++, BW+	++, ++

Predicted phenotypic ratio of F_2: 3+ : 1BW : 1.3EY

Part 3: Sex-linked

Question: What is the pattern of inheritance in a monohybrid cross with a sex-linked mutation?

Hypothesis: A cross between two parents that have different alleles (one that expresses the mutated trait on the X chromosome and one that expresses the wild type trait) on the X chromsome at one gene will produce offspring following Mendel's first law, the law of segregation.

Assumptions: Genes are sex-linked. The mutated gene is on the X chromosome. Both males and females are potential carriers.

S= sable body

1) **Predicted phenotypic ratio of the F_1 generation (parental cross)**

Phenotype of parents: ♀ S x ♂ +
Genotype of parents: ♀ $X^s X^s$ x ♂ $X^+ Y$

♀ ♂	**X^+**	**Y**
X^s	$X^s X^+$	$X^s Y$
X^s	$X^s X^+$	$X^s Y$

Predicted phenotypic ratio of F_1: ♀ 1+ : ♂ 1S

2) **Predicted phenotypic ratio of the F_1 generation (parental cross)**

F_1 cross: ♀ ++ x ♂ ++ (♀X^+ , X^s x ♂ X^s ,Y)

♀ ♂	**X^s**	**Y**
X^+	$X^+ X^s$	$X^+ Y$
X^s	$X^s X^s$	$X^s Y$

Predicted phenotypic ratio of F_1: ♀ 1+ ; ♀ 1S : ♂ 1+; 1S

S1 Reciprocal:

Question: What is the pattern of inheritance in a monohybrid cross with a sex-linked mutation?

Hypothesis: A cross between two parents that have different alleles (one that expresses the mutated trait on the X chromosome and one that expresses the wild type trait) on the X chromsome at one gene will produce offspring following Mendel's first law, the law of segregation.

Assumptions: Genes are sex-linked. The mutated gene is on the X chromosome. Both males and females are potential carriers.

1) **Predicted phenotypic ratio of the F_1 generation (parental cross)**

Phenotype of parents: ♀ + x ♂ S
Genotype of parents: ♀ $X^+ X^+$ x ♂ $X^s Y$

♀ ♂	**X^s**	**Y**
X^+	$X^s X^+$	$X^+ Y$
X^+	$X^s X^+$	$X^+ Y$

Predicted phenotypic ratio of F_1: ♀ 1+ : ♂ 1+

2) Predicted phenotypic ratio of the F$_1$ generation (parental cross)

F$_1$ cross: ♀ + x ♂ + (♀X^+ , X^s x ♂ X^+ ,Y)

♀ ♂	**X^s**	**Y**
X^+	X^+ X^s	X^+ Y
X^s	X^s X^s	X^s Y

Predicted phenotypic ratio of F$_1$: ♀ 2+ : ♂ 1+; 1S

Summary of experimental design: Using the Flylab online program and chi-squared goodness of fit test we designed two flies and crossed them together. One of their offspring were selected and crossed with a designed heterozygous fly.

RESULTS

Table 1- Chi-Square Analysis Results for Mono-, Di-, and Tri-hybrid Crosses

Cross Type	**Cross (♀ x ♂)**	**n**[a]	**Predicted ratio**	**Observed ratio**	**Total x^{2b}**	**DF**[c]	**p**[d]	**H_o**[e]
Part 1: Monohybrid (L1)	P(AR) x P(+)	10053	100% +	100% +	-	-	-	-
	F_1(AR) x F1(AR)	10038	-	-	0.016	1	0.99	Non rejected
Part 1: Dihybrid (L2)	P(ST,+) x P(+,SB)	9983	1+ : 1SB : 1 ST : 1SB;ST	1+ : 1SB : 1 ST : 1SB;ST	-	-	-	-
	F_1(STSB,STSB) x F_1(STSB,STSB)	10183	-	-	3.53	3	0.32	Non rejected
Part 2: Epistasis	P(EY,+) x P(+,BW)	9932	100%+	100%+	-	-	-	-
	F_1(+,+) x F_1(+,+)	9973	-	-	0.804	2	0.67	Non rejected
Part 3: Sex-linked Mutation (S1)	P(S) x P(+)	9925	100%+	100%+	-	-	-	-

	F_1(+) x F_1(S)	9983	-	-	2.76	3	0.43	Non rejected
Part 3: Sex-linked Mutation (Reciprocal)	P(+) x P(S)	10033	100%+	100%+	-	-	-	-
	F_1(+) x F_1(+)	9863	-	-	0.454	2	0.78	Non rejected

a- Sample size
b- Chi-squared value
c- Degrees of freedom
d- Probability
e- Null hypothesis

DISCUSSION

In Part 1: monohybrid cross (L1) the parental phenotype was P(AR) x P(+) and the F1 phenotype was F_1(AR) x F1(AR). The predicted ratio and observed ratio for the parent's generation was 100% +. The sample size of the parents was 10053 and for the F1 generation it was 10038. The chi-squared value was 0.016 and the degrees of freedom being 1. The statistical significance was 0.99 and we accepted the hypothesis. In the dihybrid cross (L2), the parental phenotype was P(ST,+) xP(+,SB) and the F1 phenotype was F_1(STSB,STSB) xF_1(STSB,STSB). The predicted ratio and observed ratio for the parent's generation was 1+ : 1SB : 1 ST : 1SB;ST. The sample size of the parents was 9932 and for the F1 generation it was 10183. The chi-squared value was 3.53 and the degrees of freedom being 3. The statistical significance was 0.32 and we accepted the hypothesis. He classic Mendelian ratio for a dihybrid cross is 9:3:3:1, our discovered ratios did not follow the Mendelian principles (Carlsson, 1996).

In Part 2: Epistasis the parental phenotype was P(EY,+) xP(+,BW) and the F1 phenotype was F_1(+,+) xF_1(+,+). The predicted ratio and observed ratio for the parent's generation was 100% +. The sample size of the parents was 9932 and for the F1 generation it was 9973. The chi-squared value was 0.804 and the degrees of freedom being 2. The statistical significance was 0.67 and we accepted the hypothesis. In this part of the lab we looked to observe the ratios of the mutated genes. The genes that were expressed would be considered epistatic and the ones repressed are classified as hypostatic (Cordell,2002).

In Part 3: Sex-linked Mutation (S1) the parental phenotype was P(+) xP(S) and the F1 phenotype was F_1(+) xF_1(S). The predicted ratio and observed ratio for the parent's generation was 100% +. The sample size of the parents was 9925 and for the F1 generation it was 9983. The chi-squared value was 2.76 and the degrees of freedom being 3. The statistical significance was 0.43 and we accepted the hypothesis. We observed the ratio of offspring that inherited this sex-linked trait when the trait was on the father's X-chromosome. A sex-linked trait that gets inherited by the male offspring will express the mutation whereas on a female she may be a carrier as she has 2

X chromosomes and if it is on only 1 she's classified as a carrier if the mutated gene is on both chromosomes that she will express the mutation also (Lyon,1961).

In Part 3: Sex-linked Mutation (Reciprocal) the parental phenotype was P(+) xP(S) and the F1 phenotype was F_1(+) xF_1(+). The predicted ratio and observed ratio for the parent's generation was 100% +. The sample size of the parents was 10033 and for the F1 generation it was 9863. The chi-squared value was 0.454 and the degrees of freedom being 2. The statistical significance was 0.78 and we accepted the hypothesis.

Overall conclusion:

The genotypic and phenotypic ratios of these flies can be predicted using an online simulation of fruit fly breeding where we observe the effects of a dominant wild type gene and mutant genes on 2 generations of offspring (Kelsell, 1987). We observed these mutations in a monohybrid and dihybrid. And we calculated the possible genotypic ratios of the parents who had a sex-linked (X-chromosome) mutation.

Appendix

Sample chi-squared calculation done on a dihybrid cross L2:

$$X^2 = \sum \frac{(Obs - Exp)^2}{Exp}$$

Phenotype	+	SB	ST	SB,ST	Total
Observed Value	1136	2269	2269	4509	10183
Expected Value	1170.48	2223.87	2223.87	4564.75	10183
Obs-Exp	-34.48	45.13	45.13	-55.75	...
$(\text{Obs-Exp})^2$	1188.87	2036.72	2036.72	3108.06	...
$(\text{Obs-Exp})^2$ /Exp	1.02	0.92	0.92	0.68	$3.53=X^{2a}$

[a] Values based on the degrees of freedom: 4-1=3, the p value: 0.3171 and that the hypothesis was accepted.

References

Carlsson, C., Jonsson, M., Norden, B., Dulay, M. T., Zare, R. N., Noolandi, J., ... & Zielenski, J. (1996). Screening for genetic mutations [1]. *Nature*,*380*(6571), 207-207.

Cordell, H. J. (2002). Epistasis: what it means, what it doesn't mean, and statistical methods to detect it in humans. *Human molecular genetics*, *11*(20), 2463-2468.

Kelsell, D. P., Dunlop, J., Stevens, H. P., Lench, N. J., Liang, J. N., Parry, G., ... & Leigh, I. M. (1997). Connexin 26 mutations in hereditary non-syndromic sensorineural deafness. *Nature*, *387*(6628), 80-83.

Lyon, M. F. (1961). Gene action in the X-chromosome of the mouse (Mus musculus L.).