

Genetics 201  
Extra *Drosophila* Problems

1a). A graduate student is studying two different *Drosophila* mutants, both of which have curly wings. The student knew that the mutations in the two strains (*Cy1* and *Cy2*) were in different genes. In addition, the student knew that each strain was homozygous for the *Cy1* or *Cy2* mutation, respectively. When female *Cy1* and male *Cy2* mutants were crossed to each other, the F1 progeny consisted of females with straight wings and males with curly wings. The F1 flies were then intercrossed to generate F2 progeny, which had the following phenotypes:

F2 Females    50% wild type  
                  50% curly wings

F2 Males        6% wild type  
                  94% curly wings

Explain the results observed in the F1 and F2. Please provide genotypes for all classes of progeny and explain their relative frequencies.

b). (This question is unrelated to part a, above).

Suppose you are given a true-breeding strain of *Drosophila* that exhibits a previously unknown abnormal mutant phenotype, *black-eyes*. You mate the Black-eyed mutant females to males from a balanced lethal strain of the genotype:

$$Cy Pm^+ / Cy^+ Pm, D Sb^+ / D^+ Sb$$

Note that the dominant markers curly wings (*Cy*) and plum eye (*Pm*) are on chromosome 2; and the dominant markers dichaete wing (*D*) and stubble bristles (*Sb*) are on chromosome 3. Homozygosity for either the curly, plum, dichaete, or stubble mutation is lethal.

i. You observe that the Black-eyed mutant phenotype does not appear in the F1. Can you conclude whether or not the *black-eyes* mutation is X-linked? Why or why not?

ii. Is the mutation that causes Black-eyes dominant or recessive? Briefly explain your reasoning.

iii. The F1 males with curly wings and stubby bristles are then backcrossed to the original mutant females.

Predict the distribution of phenotypes and their frequency in the F2 progeny if the *black-eyes* mutation is on chromosome 2.

iv. Suppose you observed that in the F2 progeny, the Black-eyed phenotype appeared in equal association with the curly and stubble phenotypes.

Which chromosome would you conclude carries the *black-eye* mutation, and why?

2). Having gotten a taste of the awesome power of fly genetics in Genetics 201, you decide to rotate in Dr. Dinerro Hughes' lab, which has just cloned the single master invertebrate gene PQE. The PQE gene, which maps to chromosome two, is upstream of all zygotically expressed fly genes and is therefore essential for viability.

To date, the only existing mutation in the PQE gene is a loss of function allele named *pqe*. *pqe/pqe* homozygous embryos die at the onset of zygotic transcription, while *pqe/+* heterozygotes are indistinguishable from wild-type flies. Impressed by your enthusiasm and solid background in genetics, Dr. Hughes asks you to design a screen to isolate additional alleles of the PQE gene.

You have available the following fly stocks:

(Note: all strains are homozygous at the *white* (*w*) locus, a marker on the X chromosome that causes white eyes.)

1. ***w; Tft / CyO*** Tufted (***Tft***) has a dominant bristle phenotype (tufted bristles) and a recessive lethal phenotype. ***CyO*** is a second chromosome balancer, which carries a recessive lethal mutation and has a dominant curly wing phenotype.
2. ***w; P[w<sup>+</sup>] / P[w<sup>+</sup>]*** Flies of this genotype have red eyes due to the ***w<sup>+</sup>*** allele. Also, their copy of the ***w<sup>+</sup>*** allele is flanked by fully functional P element inverted repeats. The P element insertion maps near the PQE locus on chromosome 2.
3. ***w; Sb Δ2-3 / TM6*** Stubble (***Sb***) is a dominant homozygous lethal mutation on chromosome 3, resulting in short bristles (this phenotype is distinguishable from ***Tft***). ***Δ2-3*** is a stable source of transposase on chromosome 3. ***TM6*** is a balancer for chromosome 3.  
Note: A P-element screen may be performed with a specially modified, immobile P-element that provides a stable source of transposase, ***Δ2-3***. Crossing the ***Δ2-3*** chromosome into a fly with a mobile P-element causes the P-element to hop.
4. ***pqe Su / CyO*** Lab stock of ***pqe*** flies, maintained as heterozygotes with ***CyO*** chromosome 2 balancer. Chromosome with the ***pqe*** allele also carries the dominant marker ***Su*** (stumpy), which

produces a short-legged phenotype. *CyO* carries a recessive lethal mutation and has a dominant curly wing phenotype.

a). Using the above reagents, design a P-element mutagenesis screen that would allow you to identify additional loss-of-function mutant alleles at the PQE locus. Be sure to specify genotypes, sex, and number of flies used in each cross. Also, indicate the phenotypes you would use to identify the desired progeny at each step.

b). Your screen is a success! Early on, you manage to pull out a candidate for a new allele of the PQE gene, which you name *pqe*<sup>2</sup>. In order to study the allele further, you must make a balanced stock of *pqe*<sup>2</sup> flies. How would you do this, using the available strains?

c). As you tally your potential *pqe* mutants, you find that you have an astonishing total of 17 new alleles. Sequence data determines that 16 of these new alleles contain a G to C transition at nucleotide 174; the 17th allele contains an A to T transition at nucleotide 55. Dr. Hughes looks at your data and asks you whether or not you are sure your mutations were independently isolated (i.e., that each of the 16 identical alleles arose from a separate P-element hop).

-Given your screen design in part a, are your mutations independently isolated? If so, justify your assertion. If not, propose how you could modify your screen to guarantee that you isolated independent mutations.

3). You are a *Drosophila* geneticist who has isolated a new mutation affecting wing size, *gne*<sup>1</sup>, on chromosome 3. This mutation has the following dosage phenotypes:

Genotype	Wing
+ / +	wild-type
+ / <i>Df</i>	half-sized
+ / <i>gne</i> <sup>1</sup>	half-sized
<i>gne</i> <sup>1</sup> / <i>Df</i>	none
<i>gne</i> <sup>1</sup> / <i>gne</i> <sup>1</sup>	none
+ / + / <i>gne</i> <sup>1</sup>	wild-type
+ / + / <i>Df</i>	wild-type
+ / <i>gne</i> <sup>1</sup> / <i>Df</i>	half-sized
+ / + / +	larger than normal

a). What type of mutation is *gne*<sup>1</sup>? What are the dosage requirements?

b). Design a simple scheme for generating and isolating more *gne* alleles on a wild-type chromosome. Include crosses you would use to put the new mutations into a permanent fly stock. For each cross, be sure to note the sex of the flies in the crosses and what phenotypes you will use to identify the desired progeny. In addition to wild-type flies, you may also use the following stocks:

*gne*<sup>1</sup> *e*<sup>1</sup> / *gne*<sup>1</sup> *e*<sup>1</sup>      A homozygous stock which has the *gne*<sup>1</sup>-bearing chromosome marked with *e*<sup>1</sup>, a recessive homozygous viable marker. Flies which are *e*<sup>1</sup> / *e*<sup>1</sup> have a darkened adult cuticle. The *e*<sup>1</sup> locus maps so near to *gne*<sup>1</sup> that recombination between the two genes is negligible.

*l*<sup>1</sup> / *Bal*, *Sb* *e*<sup>1</sup>      *Bal* is a balancer for the entire third chromosome. It carries the *e*<sup>1</sup> allele described above, as well as a dominant *Sb* (Stubble) marker that causes short bristles. It is kept in stock heterozygous with a chromosome carrying *l*<sup>1</sup>, a recessive lethal mutation.

c). Now, imagine that the *e*<sup>1</sup> marker is still on chromosome three but is located very far away from the *gne* gene. How does this affect your strategy described in part b), above?

4). As a graduate student in a *Drosophila* genetics lab, you identify a mutation that affects the head size of flies. Flies homozygous for this mutation, called Bighead (*bh*), have enlarged heads compared to wildtype flies. Homozygous *bh/bh* flies are viable and fertile.

a). You have mapped the mutant *bh* allele to chromosome II. You decide to map its position relative to two other known markers on chromosome II, cinnabar (*cn*) and brown (*bw*). To do so, you cross *cn bh bw / cn bh bw* flies to homozygous wild-type flies. You then take the phenotypically wild-type females, cross them to *cn bh bw / cn bh bw* males, and analyze the progeny.

You obtain the following data:

<u>Phenotype</u>	<u># progeny</u>
<i>cn bh bw</i>	435
+ + +	450
<i>cn</i> + +	68
+ <i>bh</i> <i>bw</i>	75
<i>cn</i> + <i>bw</i>	183
+ <i>bh</i> +	205
<i>cn bh</i> +	7
+ + <i>bw</i>	4
Total progeny	1427

Draw a map ordering the *cn*, *bh*, and *bw* genes, including the distances between them in map units.

b). To continue your project studying *Drosophila* head size, you decide to screen for recessive mutations on chromosome II that give a small head phenotype.

Design a screen to identify these mutations and generate stable lines given only the two strains listed below (and an ample supply of EMS). Be sure to indicate the sex of the flies in each cross, when you will be using single flies or mass vials, and what phenotype you will use to identify the desired progeny.

<u>Strains</u>	<u>Explanation</u>
<i>B1 / CyO</i>	<i>B1</i> is a chromosome II marker with a dominant bristle phenotype. <i>CyO</i> is a chromosome II balancer with a dominant curly-wing phenotype and a recessive lethal phenotype.
+/+	Wild-type flies

c). How would you modify your screen from part b) to identify maternal effect mutations that result in the small head phenotype (using the same starting strains)?

d). A neighboring lab which also studies head development in *Drosophila* has identified a gene, Tinyhead (*Th*), that is essential for normal head development. However, a *th* mutation is homozygous lethal. One way to study the function of the *Th* gene during eye development is use mitotic recombination.

Describe an experiment to generate clones of mutant tissue using FLP/FRT-mediated mitotic recombination. Assume you have access to any necessary FRT-marked chromosomes. Draw the chromosomes you will use, including markers, both before and after the recombinations occur. How will you interpret the data from this experiment?