# Exams

# Fall 2001

# 7.03 Exam 1

**Name:** _____

**Section:** **TA:** _____

**Exam starts at 11:05 and ends at 11:55**

**There are six pages including this cover page**
**Please write your name on each page.**

Also...

- Look over the entire exam so you don't spend too much time on hard questions leaving easy questions unanswered.

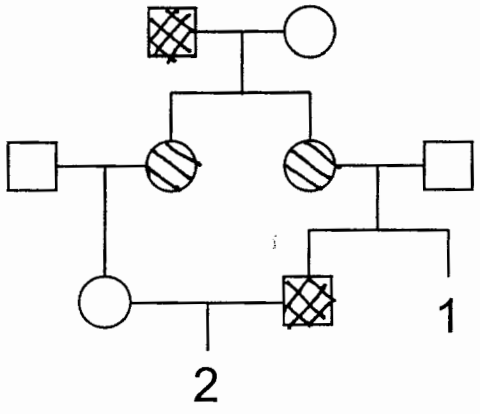
- Check your answers to make sure that they make sense.

- To help us give partial credit, show your work and state any assumptions that you make.

| | |
|---|---|
| **Question 1** | **25 points** |
| **Question 2** | **25 points** |
| **Question 3** | **25 points** |
| **Question 4** | **25 points** |

**Name:**

**1.** Consider the following mouse pedigree showing the segregation of two **X-linked** traits. (Assume all phenotypes are completely penetrant and that no new mutations arise).




= female with trait **A**

= male with trait **A** and trait **B**

Trait **A** is caused by a dominant X-linked allele. Trait **B** is caused by a recessive X-linked allele. The genes for trait **A** and trait **B** are 10 cM apart on the X chromosome. Two mice in this pedigree labeled 1 and 2 are of unknown gender and phenotype.

(**a** 5 pts.) If mouse 1 is a male, what is the probability of it exhibiting trait **A**?

(**b** 5 pts.) If mouse 1 is a female, what is the probability of it exhibiting trait **B**?

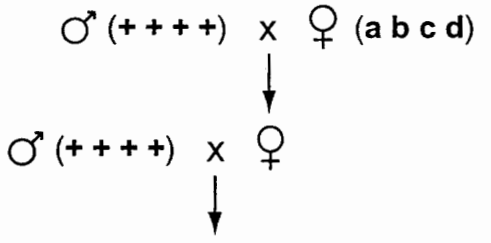
(**c** 5 pts.) If mouse 2 is a male, what is the probability of it exhibiting trait **A**?

(**d** 5 pts.) If mouse 2 is a female, what is the probability of it exhibiting *both* trait **A** and trait **B**?

(**e** 5 pts.) If mouse 2 is a male, what is the probability of it exhibiting *neither* trait **A** or trait **B**?

**Name:**

**2.** A true-breeding *Drosophila* line with four different recessive traits(**a**, **b**, **c**, and **d**) is crossed to wild-type. The $F_1$ females that result from this cross are then crossed to wild-type males.




**(a** 5 pts.) Many flies of both sexes from this second cross are examined and none show the recessive **d** trait. What does this tell you about the chromosome on which the **d** gene resides?

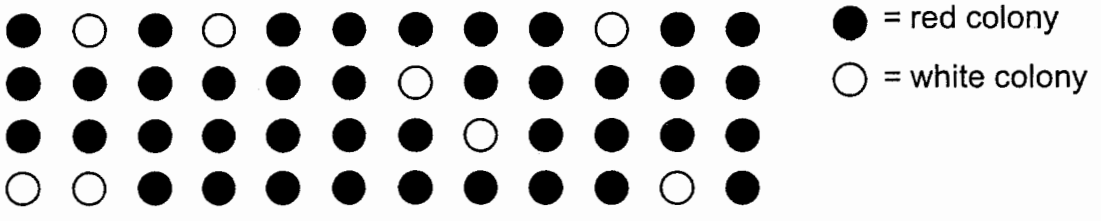
A total of 200 progeny from the second cross are evaluated for each of the three remaining traits. The 100 females among the progeny all appear as wild-type (i.e. none exhibit any of the recessive traits). For the 100 males among the progeny, eight different phenotypic classes are observed. The phenotypes and number of each of the phenotypic classes are given below. For simplicity, phenotypes of the three recessive traits are designated **a**, **b**, and **c,** while the corresponding wild-type phenotypes are designated with a "**+**".

| Phenotype | Number |
|---|---|
| + + + (females) | 100 |
| + + + (males) | 18 |
| **a b c** (males) | 22 |
| **a b** + (males) | 21 |
| + + **c** (males) | 19 |
| **a** + **c** (males) | 6 |
| + **b** + (males) | 4 |
| + **b c** (males) | 7 |
| **a** + + (males) | 3 |

**Name:**

**2b** 20 pts.) Give as much information as you can about the chromosomal positions of the three markers **a**, **b**, and **c**. Include in your answer any relevant map distances in cM.

**3.** Wild type yeast make white colonies. You have isolated two mutants that make red colonies that you call **red1** and **red2**. A **red1** mutant is crossed to a **red2** mutant and twelve of the resulting tetrads are dissected and analyzed as shown below:



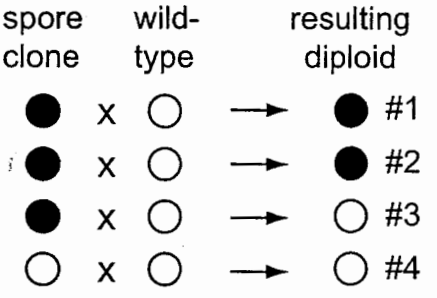

(**a** 6 pts.) How many tetrads of each type are there?

PD NPD T

(**b** 7 pts.) Are the **red1** and **red2** mutations linked? If so, how far apart are they in cM?

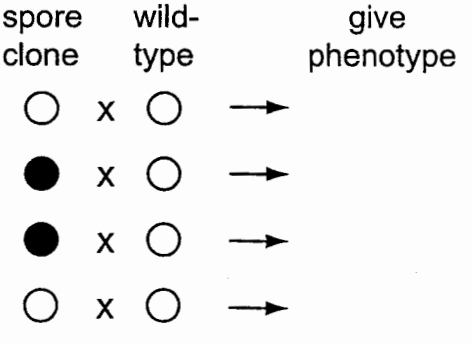
**Name:**

**3** cont.) One of the tetrads from above is selected for tests of dominance and recessivity. To do this, each of the four spore clones is mated to wild-type. The phenotypes of the resulting diploids are shown below:




**(c** 5 pts.) When diploid #3 is sporulated, what will the tetrads look like with respect to red and white phenotypes? Give a short explanation of your reasoning.

**(d** 7 pts.) A second tetrad from part (a) is chosen, and each of the four spore clones is mated to wild-type. In the diagram below, fill in the expected phenotypes of the resulting diploids. State any ambiguities that may exist.

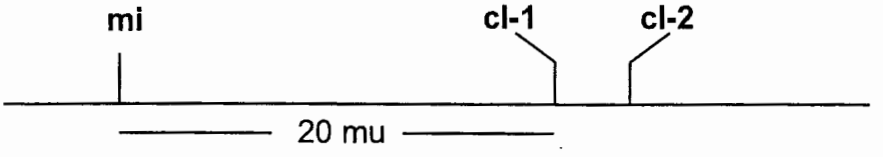


**Name:**

**4.** You have isolated two different mutants of phage λ in the repressor gene that make clear plaques rather than the normal turbid plaques. These mutants are called **cI-1⁻** and **cI-2⁻**. You cross a **cI-1⁻** phage with a **cI-2⁻** phage by coinfecting *E. coli* with phage of both types. Of 1000 plaques that result from the cross, only 20 form turbid plaques while the rest are clear.

**(a** 10 pts.) What is the distance between the **cI-1⁻** and the **cI-2⁻** mutations in map units?

Mutants that are **mi⁻** are easily detected because they form small plaques. The distance between the **mi** gene and the **cI** gene is about 20 m.u.. Assume that the genetic order of mutations is as follows:




**(b** 15 pts.) For a cross of a **mi⁻ cI-1⁻** double mutant to a **cI-2⁻** mutant a total of 1000 plaques are examined. In the table below fill in the expected number of plaques of each phenotypic type:

| Plaque Phenotype | Number |
|---|---|
| clear, large | |
| clear, small | |
| turbid, large | |
| turbid, small | |

# 7.03 Exam 1

Name: KEY

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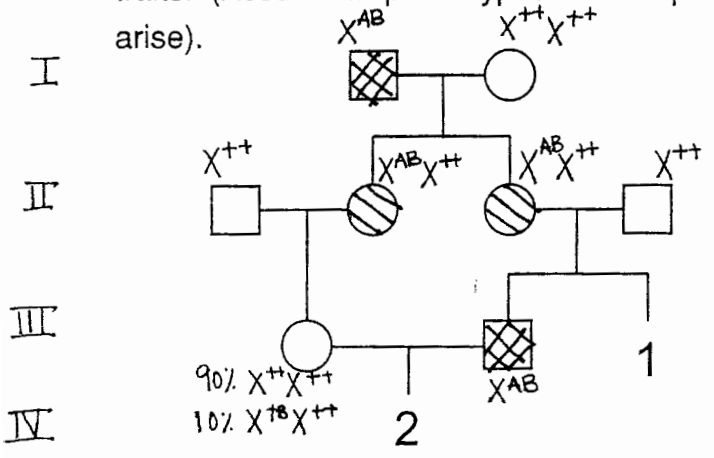
**Name:**

**1.** Consider the following mouse pedigree showing the segregation of two **X-linked** traits. (Assume all phenotypes are completely penetrant and that no new mutations arise).




= female with trait **A**

= male with trait **A** and trait **B**

Trait **A** is caused by a dominant X-linked allele. Trait **B** is caused by a recessive X-linked allele. The genes for trait **A** and trait **B** are 10 cM apart on the X chromosome. Two mice in this pedigree labeled 1 and 2 are of unknown gender and phenotype.

(**a** 5 pts.) If mouse 1 is a male, what is the probability of it exhibiting trait **A**?

p(A from mom) = $\boxed{\frac{1}{2}}$

(**b** 5 pts.) If mouse 1 is a female, what is the probability of it exhibiting trait **B**?

p(B from mom and B from dad) = $\frac{1}{2} \times 0 = \boxed{0}$

(**c** 5 pts.) If mouse 2 is a male, what is the probability of it exhibiting trait **A**?

p(A from mom) = $\boxed{0}$

A is dominant, mom can't have A

(**d** 5 pts.) If mouse 2 is a female, what is the probability of it exhibiting *both* trait **A** and trait **B**?

II-B meiosis: ($X^{AB}$ 45%), $X^{++}$ 45%, ($X^{A+}$ 5%), $X^{+B}$ 5%

not possible — III-A doesn't have A

III-A: 90% $X^{++}X^{++}$, 10% $X^{+B}X^{++}$

p(A and B from dad) × p(B from mom) × p(mom has B)

$1 \times \frac{1}{2} \times \frac{1}{10}$

$\boxed{\frac{1}{20} = 5\%}$

(**e** 5 pts.) If mouse 2 is a male, what is the probability of it exhibiting *neither* trait **A** or trait **B**?

p(getting $X^{++}$ from mom) = $\boxed{95\%}$

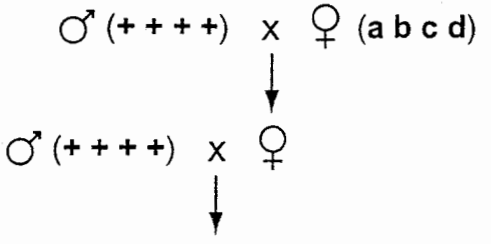
90% $X^{++}X^{++}$ → will always give $X^{++}$ = 90%

10% $X^{+B}X^{++}$ → will gives $X^{++}$ half the time = 5%

95%

**Name:**

**2.** A true-breeding *Drosophila* line with four different recessive traits(**a**, **b**, **c**, and **d**) is crossed to wild-type. The $F_1$ females that result from this cross are then crossed to wild-type males.




(**a** 5 pts.) Many flies of both sexes from this second cross are examined and none show the recessive **d** trait. What does this tell you about the chromosome on which the **d** gene resides?

d resides on an autosomal chromosome

A total of 200 progeny from the second cross are evaluated for each of the three remaining traits. The 100 females among the progeny all appear as wild-type (i.e. none exhibit any of the recessive traits). For the 100 males among the progeny, eight different phenotypic classes are observed. The phenotypes and number of each of the phenotypic classes are given below. For simplicity, phenotypes of the three recessive traits are designated **a**, **b**, and **c,** while the corresponding wild-type phenotypes are designated with a "**+**".

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| **a** + + (males) | 3 |

**Name:**

**2b** 20 pts.) Give as much information as you can about the chromosomal positions of the three markers **a**, **b**, and **c**. Include in your answer any relevant map distances in cM.

| | ab | bc | ac |
|---|---|---|---|
| parental | 80 | 50 | 50 |
| recombinants | 20 | 50 | 50 |

① c is unlinked → number of parentals and recombinants are equal

② a, b, c are all on X-chromosome; all are X-linked traits

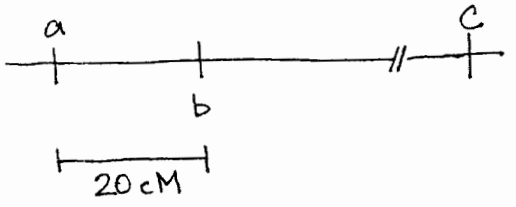
③ a,b distance cM = $100 \times \frac{20}{100}$

= 20 cM

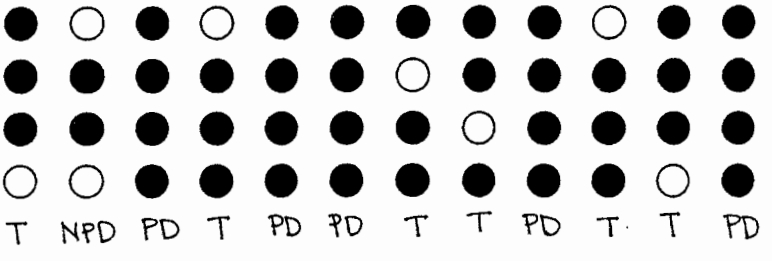
b,c distances cM = $100 \times \frac{50}{100}$

a,c = 50 cM

confirms that c is unlinked




**3.** Wild type yeast make white colonies. You have isolated two mutants that make red colonies that you call **red1** and **red2**. A **red1** mutant is crossed to a **red2** mutant and twelve of the resulting tetrads are dissected and analyzed as shown below:



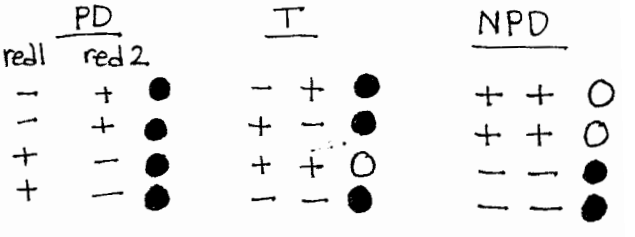

● = red colony

○ = white colony

red1⁻ red2⁺ × red1⁺ red2⁻

(**a** 6 pts.) How many tetrads of each type are there?

| PD | NPD | T |
|---|---|---|
| 5 | 1 | 6 |




(**b** 7 pts.) Are the **red1** and **red2** mutations linked? If so, how far apart are they in cM?

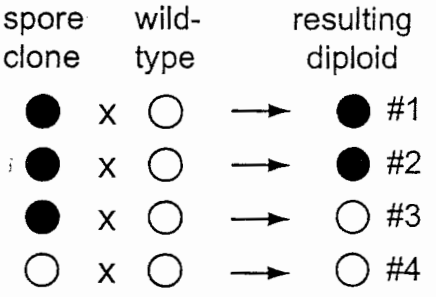
PD ≠ NPD ⟹ linked

$$\text{Distance} = \frac{T + 6NPD}{2\Sigma} \times 100 = \frac{6 + 6(1)}{2(12)} \times 100$$

$$= 50\ \text{cM}$$

**Name:**

**3** cont.) One of the tetrads from above is selected for tests of dominance and recessivity. To do this, each of the four spore clones is mated to wild-type. The phenotypes of the resulting diploids are shown below:

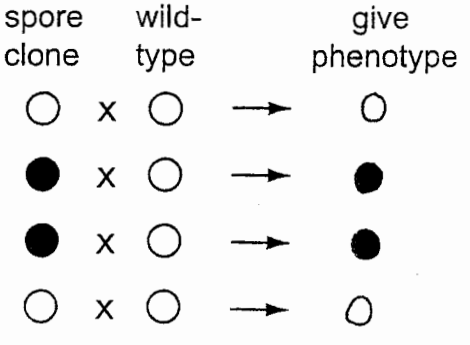



(**c** 5 pts.) When diploid #3 is sporulated, what will the tetrads look like with respect to red and white phenotypes? Give a short explanation of your reasoning.

2 red : 2 white

The spore was either $red1^{-}red2^{+}$ or $red1^{+}red2^{-}$ and the mutation was recessive.

(**d** 7 pts.) A second tetrad from part (a) is chosen, and each of the four spore clones is mated to wild-type. In the diagram below, fill in the expected phenotypes of the resulting diploids. State any ambiguities that may exist.



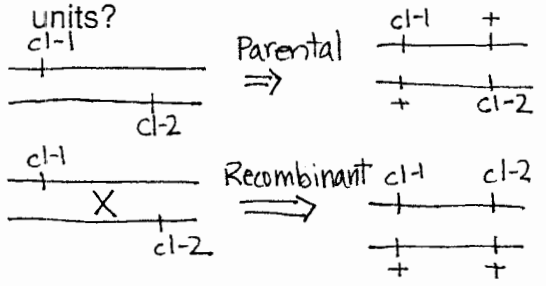

NPD from part a, so spores are either $red1^{+}red2^{+}$ or $red1^{-}red2^{-}$. Since either $red1^{-}$ or $red2^{-}$ is dominant (known from part c) the diploid is red.

Ambiguity = whether $red1^{-}$ or $red2^{-}$ is dominant to wt

**Name:**

**4.** You have isolated two different mutants of phage λ in the repressor gene that make clear plaques rather than the normal turbid plaques. These mutants are called **cI-1⁻** and **cI-2⁻**. You cross a **cI-1⁻** phage with a **cI-2⁻** phage by coinfecting *E. coli* with phage of both types. Of 1000 plaques that result from the cross, only 20 form turbid plaques while the rest are clear.

**(a** 10 pts.) What is the distance between the **cI-1⁻** and the **cI-2⁻** mutations in map units?



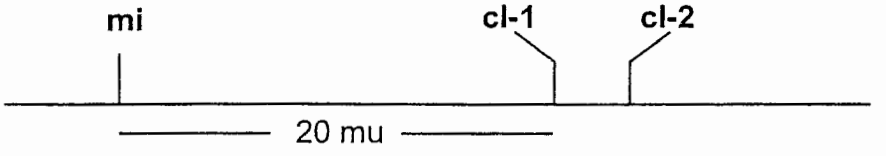

hidden recombinants

20 = ½ recombinants

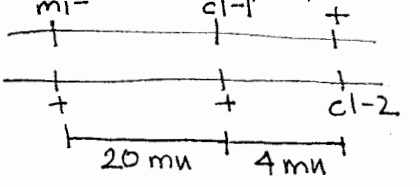
$$\text{Distance} = \frac{2 \times 20}{1000} \times 100$$

$$= 4 \text{ mu}$$

Mutants that are **mi⁻** are easily detected because they form small plaques. The distance between the **mi** gene and the **cI** gene is about 20 m.u.. Assume that the genetic order of mutations is as follows:




**(b** 15 pts.) For a cross of a **mi⁻ cI-1⁻** double mutant to a **cI-2⁻** mutant a total of 1000 plaques are examined. In the table below fill in the expected number of plaques of each phenotypic type:




| | Plaque Phenotype | Number |
|---|---|---|
| | clear, large | |
| Parental: No | + + cI-2 | (.80)(.96)/2 x1000 = 384 |
| Cross-overs 1 | + cI-1 + | (.20 x .96)/2 x 1000 = 96 |
| 2 | + cI-1 cI-2 | (.2 x .04)/2 x 1000 = 4 |
| | clear, small | |
| No | mi⁻ cI-1 + | (.80 x .96)/2 x 1000 = 384 |
| 1 | mi⁻ + cI-2 | (.20 x .96)/2 x 1000 = 96 |
| 2 | mi⁻ cI-1 cI-2 | (.80 x .04)/2 x 1000 = 16 |
| | turbid, large | |
| 1 | + + + | (.80 x .04)/2 x 1000 = 16 |
| | turbid, small | |
| 2 | mi⁻ + + | (.20 x .04)/2 x 1000 = 4 |

= 484

= 496

General Calculation For Single CO = [% chance rec. (A-B) x % chance no rec. (B-C)] / 2 (# products formed) x 1000 (total #)

# 7.03 Exam 2

Name: _____

Section: _____ TA: _____

**Exam starts at 11:05 and ends at 11:55**

**There are five pages including this cover page**
**Please write your name on each page.**

| | |
|---|---|
| Question 1 | 25 points |
| Question 2 | 27 points |
| Question 3 | 28 points |
| Question 4 | 20 points |

Name: _____

**1.** **(a** 5 pts.) The codon for tryptophan is $^{5'}$UGG$^{3'}$. Write out the RNA sequence of the anti-codon portion of tRNA$^{trp}$, with the 5' and 3' ends labeled.

**(b** 5 pts.) Write out the double stranded DNA sequence for the anti-codon portion of the gene for tRNA$^{trp}$ (label the 5' and 3' ends).

**(c** 5 pts.) The sequence of the amber stop codon is $^{5'}$UAG$^{3'}$. Say that you have isolated an amber suppressing allele of tRNA$^{trp}$. Write out the RNA sequence of the anti-codon of this suppressing tRNA (label the 5' and 3' ends).

**(d** 5 pts.) You have a mutagen that can chemically modify the base guanine so that it can form base pairs with thymine. Will treatment of *E. coli* with this mutagen increase the probability of generating amber suppressing alleles of tRNA$^{trp}$? Why or why not?

**(e** 5 pts.) The sequence of the ochre stop codon is $^{5'}$UAA$^{3'}$. Is it more probable for tRNA$^{trp}$ to be mutated to become an ochre suppressor or an amber suppressor? Why?

Name: _____

**2.** **(a** 5 pts.) You have isolated a **Tn5** insertion in an otherwise wild type *E. coli* strain that is linked to the **Lac** operon. You grow **P1** phage on the strain with the **Tn5** insertion and use the resulting phage to infect a **LacZ⁻** strain. Among the resulting Kan$^r$ transductants, 60% have no ß-galactosidase activity and 40% express ß-galactosidase normally. What is the distance between the **Tn5** insertion and **LacZ**, expressed as a cotransduction frequency?

You grow **P1** phage on one of the Kan$^r$ transductants isolated in part (a) that is **LacZ⁻**. You use the resulting phage to infect a **LacI⁻** mutant and then isolate 1,000 Kan$^r$ transductants. For each transductant you assay both ß-galactosidase activity (**LacZ**) and Lac permease activity (**LacY**) in the presence or absence of inducer.

**(b** 12 pts.) In the table below fill in the **Lac** genotypes of the different classes of transductants.

| number | ß-galactosidase | permease | Genotype |
|---|---|---|---|
| 578 | uninducible | regulated | _____ |
| 400 | constitutive | constitutive | _____ |
| 20 | uninducible | constitutive | _____ |
| 2 | regulated | regulated | _____ |

**(c** 5 pts.) What is the distance between the **Tn5** insertion and the **LacI** gene expressed as a cotransduction frequency?

**(d** 5 pts.) Draw a genetic map showing the relative order of **Tn5**, **LacZ**, and **LacI**. Also give any relevant distances expressed as cotransduction frequencies.

**Name:** _____

**3.** **(a** 16 pts.) For the following merodiploid strains, determine the level ß-galactosidase expression in either the presence or absence of the inducer IPTG. Assume that when no repressor is bound to DNA, 100 units of ß-galactosidase activity is produced from each functional copy of the **LacZ** gene and when repressor is fully bound to DNA only 1 unit of enzyme is produced for each functional copy of **LacZ**. The presence of **Lac $I^{-d}$** protein will fully prevent other forms of the repressor in the same cell from binding to DNA. The **Lac $I^{s}$** protein binds to DNA but not to the inducer.

| | ß-galactosidase activity | |
|---|---|---|
| | –IPTG | +IPTG |
| Lac $O^+$ $Z^+$ / F' Lac $O^c$ $Z^-$ | _____ | _____ |
| Lac $I^+$ $O^c$ $Z^+$ / F' Lac $I^{-d}$ $O^+$ $Z^+$ | _____ | _____ |
| Lac $I^+$ $O^c$ $Z^+$ / F' Lac $I^s$ $O^+$ $Z^+$ | _____ | _____ |
| Lac $I^{-d}$ $O^c$ $Z^+$ / F' Lac $I^s$ $O^+$ $Z^-$ | _____ | _____ |

**(b** 12 pts.) For the following merodiploid strains, determine the level maltase activity in either the presence or absence of the inducer maltose. Assume that when the activator (**MalT**) is bound to DNA, 100 units of maltase activity is produced from each functional copy of the **MalQ** gene and when no activator is bound to DNA only 1 unit of enzyme is produced for each functional copy of **MalQ**. The **$MalT^C$** protein binds DNA regardless of whether maltose is present.

| | maltase activity | |
|---|---|---|
| | –maltose | +maltose |
| $MalT^-$ $Q^+$ / F' $MalT^+$ $Q^-$ | _____ | _____ |
| $MalT^C$ $Q^+$ / F' $MalT^+$ $Q^-$ | _____ | _____ |
| $MalT^C$ $Q^-$ / F' $MalT^-$ $Q^+$ | _____ | _____ |

**Name:** _____

**4.** An *E. coli* enzyme encoded by gene E is expressed in response to an inducer molecule. You have isolated regulatory mutations in two genes A and B that are not linked to each other and are not linked to the gene for enzyme E. Both the $A^-$ and $B^-$ mutations give uninducible expression of enzyme E. You construct the following merodiploids to learn more about the $A^-$ and $B^-$ mutations.

| Genotype | Phenotype |
|---|---|
| $A^-E^+$ / F' $A^+E^+$ | regulated |
| $B^-E^+$ / F' $B^+E^+$ | regulated |

**(a** 10 pts.) On the basis of these results, diagram two different possible regulatory pathways that can explain the functions of the A and B gene products in the regulation of enzyme E. Include a role for the inducer molecule in your answer.

Next, you isolate an allele of the B gene that you call $B^C$ that gives constitutive expression of enzyme E. The genotype and phenotype of strains carrying the $B^C$ mutation are as follows:

| Genotype | Phenotype |
|---|---|
| $B^C$ $E^+$ / F' $B^+E^+$ | constitutive |
| $B^C$ $A^-$ $E^+$ | constitutive |

**(b** 5 pts.) Draw out the model from part (a) that is consistent with these new results?

**(c** 5 pts.) Propose a molecular mechanism for the $B^C$ mutation that explains its behavior in the regulatory pathway.

# 7.03 Exam 2

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5'-CCA-3'

**(b** 5 pts.) Write out the double stranded DNA sequence for the anti-codon portion of the gene for $tRNA^{trp}$ (label the 5' and 3' ends).

5'-CCA-3'
3'-GGT-5'

**(c** 5 pts.) The sequence of the amber stop codon is 5'UAG3'. Say that you have isolated an amber suppressing allele of $tRNA^{trp}$. Write out the RNA sequence of the anti-codon of this suppressing tRNA (label the 5' and 3' ends).

5'-CUA-3'

**(d** 5 pts.) You have a mutagen that can chemically modify the base guanine so that it can form base pairs with thymine. Will treatment of *E. coli* with this mutagen increase the probability of generating amber suppressing alleles of $tRNA^{trp}$? Why or why not?

5' CCA 3' → (mutagen) → 5'CTA 3' / 3'GGT 5' → (replication) → 5'CTA 3' / 3'GAT 5'

Yes, this mutagen will cause an increase in probability of changing $tRNA^{trp}$ (5'CCA3') to an amber suppressor (5'CUA3')

**(e** 5 pts.) The sequence of the ochre stop codon is 5'UAA3'. Is it more probable for $tRNA^{trp}$ to be mutated to become an ochre suppressor or an amber suppressor? Why?

Amber is more probable since only one base needs to be mutated. Two bases need to be mutated to create an ochre suppressor.

$tRNA^{trp}$ 5'CCA3'

5' CUA 3'  5' UUA 3'

Name: _____

**2.** (**a** 5 pts.) You have isolated a **Tn5** insertion in an otherwise wild type *E. coli* strain that is linked to the **Lac** operon. You grow **P1** phage on the strain with the **Tn5** insertion and use the resulting phage to infect a **LacZ⁻** strain. Among the resulting Kan$^r$ transductants, 40% have no ß-galactosidase activity and 60% express ß-galactosidase normally. What is the distance between the **Tn5** insertion and **LacZ**, expressed as a cotransduction frequency?

60%

You grow **P1** phage on one of the Kan$^r$ transductants isolated in part (a) that is **LacZ⁻**. You use the resulting phage to infect a **LacI⁻** mutant and then isolate 1,000 Kan$^r$ transductants. For each transductant you assay both ß-galactosidase activity (**LacZ**) and Lac permease activity (**LacY**) in the presence or absence of inducer.

(**b** 12 pts.) In the table below fill in the **Lac** genotypes of the different classes of transductants.

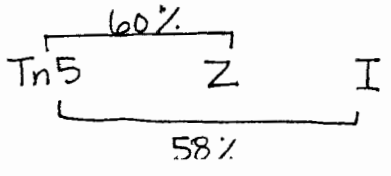
| number | ß-galactosidase | permease | Genotype |
|---|---|---|---|
| 578 | uninducible | regulated | $I^+ Z^-$ |
| 400 | constitutive | constitutive | $I^- Z^+$ |
| 20 | uninducible | constitutive | $I^- Z^-$ |
| 2 | regulated | regulated | $I^+ Z^+$ |

(**c** 5 pts.) What is the distance between the **Tn5** insertion and the **LacI** gene expressed as a cotransduction frequency?

$$\frac{578 + 2}{1000} = 58\%$$

(**d** 5 pts.) Draw a genetic map showing the relative order of **Tn5**, **LacZ**, and **LacI**. Also give any relevant distances expressed as cotransduction frequencies.

rarest class (4 cross-overs) = $I^+Z^+$ donor: Tn5 $Z^- I^+$ recipient: $Z^+ I^-$



Name: _____

**3.** (**a** 16 pts.) For the following merodiploid strains, determine the level ß-galactosidase expression in either the presence or absence of the inducer IPTG. Assume that when no repressor is bound to DNA, 100 units of ß-galactosidase activity is produced from each functional copy of the **LacZ** gene and when repressor is fully bound to DNA only 1 unit of enzyme is produced for each functional copy of **LacZ**. The presence of **Lac $I^{-d}$** protein will fully prevent other forms of the repressor in the same cell from binding to DNA. The **Lac $I^{s}$** protein binds to DNA but not to the inducer.

half credit was awarded if you used +/- scale, question asked for level

| | ß-galactosidase activity | |
|---|---|---|
| | –IPTG | +IPTG |
| Lac $O^+$ $Z^+$ / F' Lac $O^c$ $Z^-$ | 1 | 100 |
| Lac $I^+$ $O^c$ $Z^+$ / F' Lac $I^{-d}$ $O^+$ $Z^+$ | 200 | 200 |
| Lac $I^+$ $O^c$ $Z^+$ / F' Lac $I^s$ $O^+$ $Z^+$ | 101 | 101 |
| Lac $I^{-d}$ $O^c$ $Z^+$ / F' Lac $I^s$ $O^+$ $Z^-$ | 100 | 100 |

(**b** 12 pts.) For the following merodiploid strains, determine the level maltase activity in either the presence or absence of the inducer maltose. Assume that when the activator (**MalT**) is bound to DNA, 100 units of maltase activity is produced from each functional copy of the **MalQ** gene and when no activator is bound to DNA only 1 unit of enzyme is produced for each functional copy of **MalQ**. The **$MalT^C$** protein binds DNA regardless of whether maltose is present.

| | maltase activity | |
|---|---|---|
| | –maltose | +maltose |
| $MalT^-$ $Q^+$ / F' $MalT^+$ $Q^-$ | 1 | 100 |
| $MalT^C$ $Q^+$ / F' $MalT^+$ $Q^-$ | 100 | 100 |
| $MalT^C$ $Q^-$ / F' $MalT^-$ $Q^+$ | 100 | 100 |

Name: _____

**4.** An *E. coli* enzyme encoded by gene E is expressed in response to an inducer molecule. You have isolated regulatory mutations in two genes A and B that are not linked to each other and are not linked to the gene for enzyme E. Both the $A^-$ and $B^-$ mutations give uninducible expression of enzyme E. You construct the following merodiploids to learn more about the $A^-$ and $B^-$ mutations.

| Genotype | Phenotype | |
|---|---|---|
| $A^-E^+$ / F' $A^+E^+$ | regulated | trans-acting, recessive, uninducible ⇒ activator |
| $B^-E^+$ / F' $B^+E^+$ | regulated | trans-acting, recessive, uninducible ⇒ activator |

**(a** 10 pts.) On the basis of these results, diagram <u>two</u> different possible regulatory pathways that can explain the functions of the A and B gene products in the regulation of enzyme E. Include a role for the inducer molecule in your answer.

① inducer ⟶ A ⟶ B ⟶ E

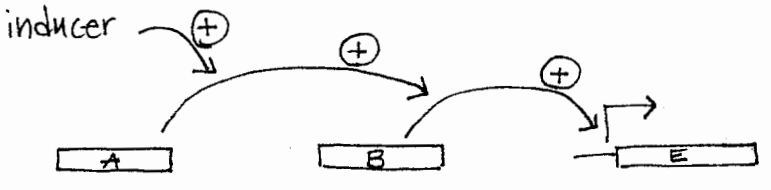
② inducer ⟶ B ⟶ A ⟶ E

Next, you isolate an allele of the B gene that you call $B^C$ that gives constitutive expression of enzyme E. The genotype and phenotype of strains carrying the $B^C$ mutation are as follows:

| Genotype | Phenotype |
|---|---|
| $B^C$ $E^+$ / F' $B^+E^+$ | constitutive |
| $B^C$ $A^-$ $E^+$ | constitutive |

dominant, constitutive, trans-acting
↓
Activator$^S$

**(b** 5 pts.) Draw out the model from part (a) that is consistent with these new results?




**(c** 5 pts.) Propose a molecular mechanism for the $B^C$ mutation that explains its behavior in the regulatory pathway.

$B^C$ is an $Act^S$ that always binds the promoter for E and always activates transcription of E.

# 7.03 Exam 3

Name: _____

Section: _____ TA: _____

Exam starts at 11:05 and ends at 11:55

There are 7 pages including this cover page
Please write your name on each page.

| | |
|---|---|
| Question 1 | 36 points |
| Question 2 | 34 points |
| Question 3 | 30 points |

**Name:** _____

**1.** Consider an autosomal gene at which a rare allele (call it allele **a**) results in homozygotes (**aa**) having 20% as many offspring as average individuals in the population.

**(a** 3 pts.**)** What is the value S for **aa** homozygotes?

Heterozygotes (**Aa**) have a 50.1% chance of surviving an infectious disease that afflicts all children in the population. On average, individuals in this population have a 50.0% chance of surviving this infectious disease.

**(b** 5 pts.**)** What is the value h associated with allele **a**?

**(c** 5 pts.**)** What is the frequency of allele **a** in the population? Show your calculations and state any simplifying assumptions that you make.

**(d** 3 pts.**)** What is the frequency of heterozygotes among newborn children?

**(e** 10 pts.**)** Inbreeding is rare in this population, but some second-cousin marriages (marriages between children of first cousins) do occur. What is the probability that offspring of second-cousin marriages will be **aa** homozygotes?

**Name:** _____

**(f** 10 pts.**)** Suppose that the organism causing the infectious childhood disease is completely eradicated. Estimate the frequency of allele **a** 50 generations later.

**2.** The DNA sequence of a 0.4-Mb region of mouse chromosome 7 has not yet been determined, but you have available 6 BACs (order unknown) and 11 STSs (order unknown) that derive from the region. By PCR, you test each of the 6 BACs for the presence (+) or absence (-) of each of the 11 STSs, and you obtain the following results:

| | BACs | | | | | |
|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 |
| STS1 | - | - | - | + | + | - |
| STS2 | - | - | + | - | - | + |
| STS3 | + | - | - | - | - | + |
| STS4 | - | + | - | - | + | - |
| STS5 | + | - | + | - | - | + |
| STS6 | + | - | - | - | - | - |
| STS7 | - | - | + | + | - | - |
| STS8 | - | - | - | + | - | - |
| STS9 | - | + | - | - | - | - |
| STS10 | - | + | - | + | + | - |
| STS11 | - | - | + | - | - | - |

**(a** 10 pts.**)** Using this STS content data, construct a physical map of the 0.4-Mb region in which you indicate the order of the BACs and the overlaps among them, and the order of the STSs.

**Name:** _____

You identify two protein-coding genes (gene A and gene B) within the 0.4-Mb region. Gene A maps between STS2 and STS7. Gene B maps between STS4 and STS9. Through genetic linkage analysis, you map a recessive eye-color mutant (called *pinkeye*) to the 0.4-Mb region. The *pinkeye* gene has not yet been defined at a molecular level, but you are confident that either gene A or gene B must be the site of the *pinkeye* mutation. Your 6 BACs all derive from a library prepared using genomic DNA from wild-type (brown-eye) mice. Recall that recessive phenotypes often reflect the loss or absence of gene function.

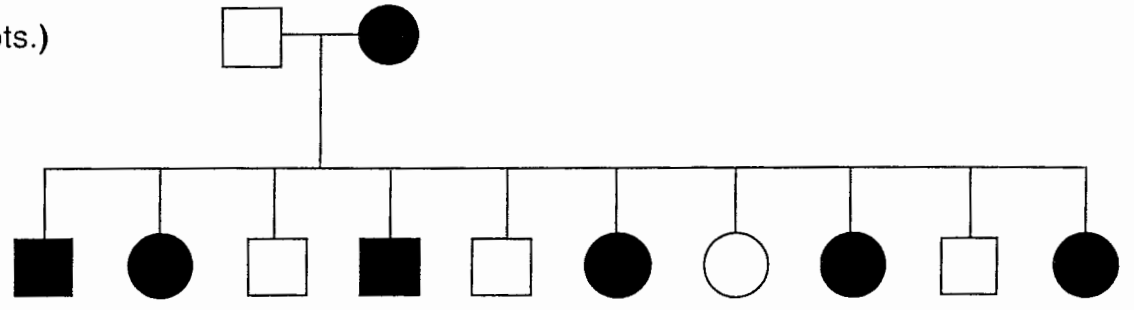
**(b** 12 pts.**)** Propose an experiment involving one or more gene knockouts (but no transgenes) that would test whether the **phenotypically defined** *pinkeye* mutation is in the **molecularly defined** gene A or whether it is in the **molecularly defined** gene B. Diagram your targeting construct(s), how it (or they) would integrate into the mouse genome, any crosses required, and the resulting genotypes.

**(c** 12 pts.**)** Propose experiments involving BAC transgenes (but no knockouts) that would test whether the *pinkeye* mutation is in gene A or whether it is in gene B. Diagram your transgene(s), how it (or they) would integrate into the mouse genome, any crosses required, and the resulting genotypes.

**Name:** _____

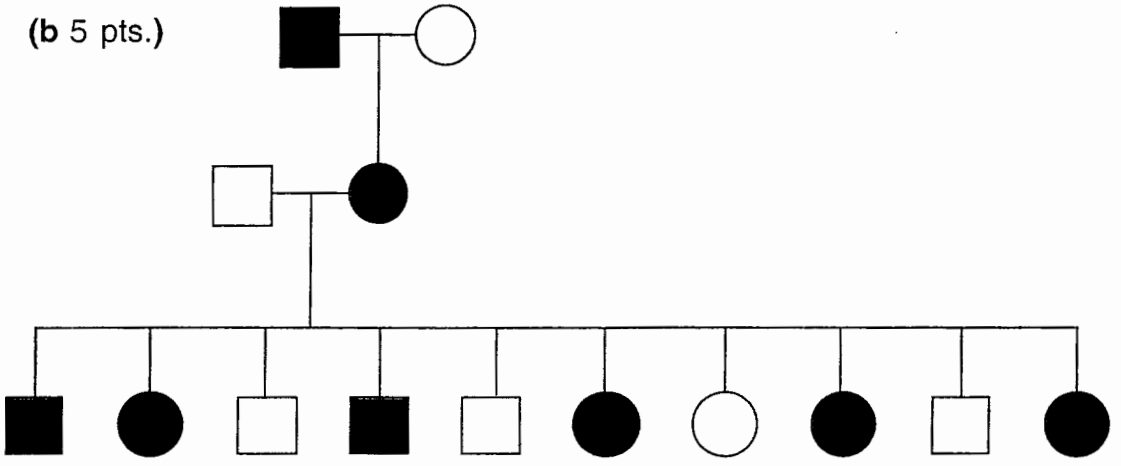
**3.** When setting out to determine the chromosomal location of a human disease gene by genetic linkage analysis (LOD scores), it is useful to calculate the theoretical maximum LOD score that a family of a given size and structure might contribute. For each of the families shown below, calculate, for $\theta = 0$, the LOD score that could be obtained using an SSR that is genetically inseparable from (shows no recombination with) the disease, which is autosomal dominant. Also indicate the minimum number of such families that would be required to achieve a publishable composite LOD score for $\theta = 0$. Assume that DNA samples are available for all living individuals.

**(a** 5 pts.)

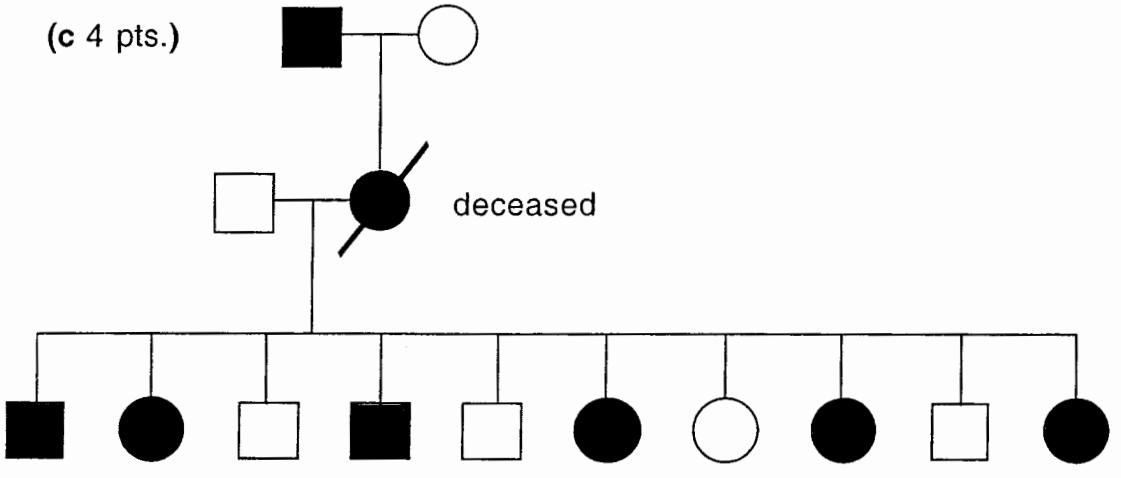


**Name:** _____

(**b** 5 pts.)

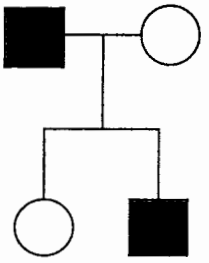


(**c** 4 pts.)

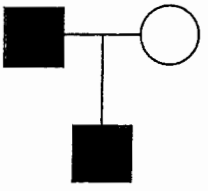


**Name:** _____

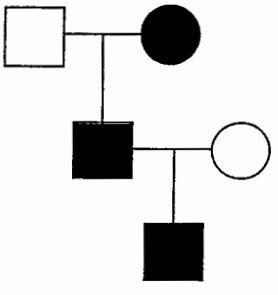
**(d** 4 pts.)



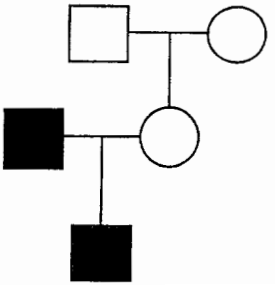
**(e** 4 pts.)



**(f** 4 pts.)



**(g** 4 pts.)



# 7.03 Exam 3

Name: KEY

Section: _____ TA: _____

Exam starts at 11:05 and ends at 11:55

There are 7 pages including this cover page
Please write your name on each page.

| | |
|---|---|
| Question 1 | 36 points |
| Question 2 | 34 points |
| Question 3 | 30 points |

Name: _____

**1.** Consider an autosomal gene at which a rare allele (call it allele **a**) results in homozygotes (**aa**) having 20% as many offspring as average individuals in the population.

(**a** 3 pts.) What is the value S for **aa** homozygotes?

$S = 0.8$

Heterozygotes (**Aa**) have a 50.1% chance of surviving an infectious disease that afflicts all children in the population. On average, individuals in this population have a 50.0% chance of surviving this infectious disease.

(**b** 5 pts.) What is the value h associated with allele **a**?

$1 - \frac{.501}{.50} = .002$

(**c** 5 pts.) What is the frequency of allele **a** in the population? Show your calculations and state any simplifying assumptions that you make.

$\Delta q = -Sq^2 + hq$

$q = \frac{h}{S} = .0025$

(**d** 3 pts.) What is the frequency of heterozygotes among newborn children?

$2pq = 2(.0025)(.9975)$

$= .005$

(**e** 10 pts.) Inbreeding is rare in this population, but some second-cousin marriages (marriages between children of first cousins) do occur. What is the probability that offspring of second-cousin marriages will be **aa** homozygotes?

$F = \frac{1}{64}$

$P = \frac{1}{64}(.0025)$

$= .9 \times 10^{-5}$

**Name:** _____

**(f** 10 pts.) Suppose that the organism causing the infectious childhood disease is completely eradicated. Estimate the frequency of allele **a** 50 generations later.

$\Delta q_{/gen} = -Sq^2 = -5\times10^{-6}$

$\Delta q_{50} = 2.5\times10^{-4}$

$q_{50} = .0025 - 2.5\times10^{-4} = 225\times10^{-3}$

**2.** The DNA sequence of a 0.4-Mb region of mouse chromosome 7 has not yet been determined, but you have available 6 BACs (order unknown) and 11 STSs (order unknown) that derive from the region. By PCR, you test each of the 6 BACs for the presence (+) or absence (-) of each of the 11 STSs, and you obtain the following results:

| | BACs | | | | | |
|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 |
| STS1 | - | - | - | + | + | - |
| STS2 | - | - | + | - | - | + |
| STS3 | + | - | - | - | - | + |
| STS4 | - | + | - | - | + | - |
| STS5 | + | - | + | - | - | + |
| STS6 | + | - | - | - | - | - |
| STS7 | - | - | + | + | - | - |
| STS8 | - | - | - | + | - | - |
| STS9 | - | + | - | - | - | - |
| STS10 | - | + | - | + | + | - |
| STS11 | - | - | + | - | - | - |

STS

BAC 4 - 3, 5, 6

2 - 4, 9, 10

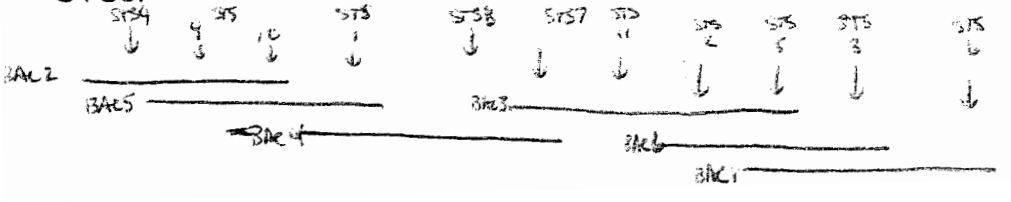
3 - 2, 5, 7, 11

4 - 1, 7, 8, 10

5 - 1, 4, 10

6 - 2, 3, 5

**(a** 10 pts.) Using this STS content data, construct a physical map of the 0.4-Mb region in which you indicate the order of the BACs and the overlaps among them, and the order of the STSs.

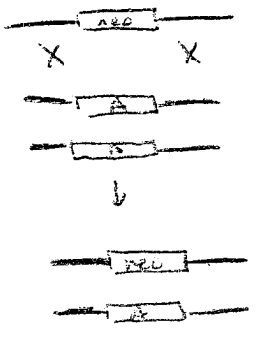


**Name:** _____

You identify two protein-coding genes (gene A and gene B) within the 0.4-Mb region. Gene A maps between STS2 and STS7. Gene B maps between STS4 and STS9. Through genetic linkage analysis, you map a recessive eye-color mutant (called *pinkeye*) to the 0.4-Mb region. The *pinkeye* gene has not yet been defined at a molecular level, but you are confident that either gene A or gene B must be the site of the *pinkeye* mutation. Your 6 BACs all derive from a library prepared using genomic DNA from wild-type (brown-eye) mice. Recall that recessive phenotypes often reflect the loss or absence of gene function.

**(b** 12 pts.) Propose an experiment involving one or more gene knockouts (but no transgenes) that would test whether the **phenotypically defined** *pinkeye* mutation is in the **molecularly defined** gene A or whether it is in the **molecularly defined** gene B. Diagram your targeting construct(s), how it (or they) would integrate into the mouse genome, any crosses required, and the resulting genotypes.

Use sequence flanking gene A or gene B (i.e. around STS2 and STS7 or around STS4 and STS9 respectively) to create a knockout cassette of neomycin resistance. Electroporate into ES cells derived from a black mouse. ② Homologous recombination will replace parts of either gene A or B with the neo cassette. ③ Neomycin selection for heterozygous ES cells and then injection of these ES cells into a blastocyst from a white mouse. ④ Both of the chimera and the cross chimera to white mice to test for germline transmission. ⑤ Black mice from that mating have a ½ chance of being a heterozygous mouse, test by PCR. ⑥ Cross two heterozygous mice to get a homozygous knockout mouse (¼ chance).




Knockout mice will be generated for gene A and gene B on its own. The resulting knockout mice will be crossed to homozygous pinkeye mice to test function by complementation. Whichever gene is the phenotypically defined pinkeye gene will fail to complement.

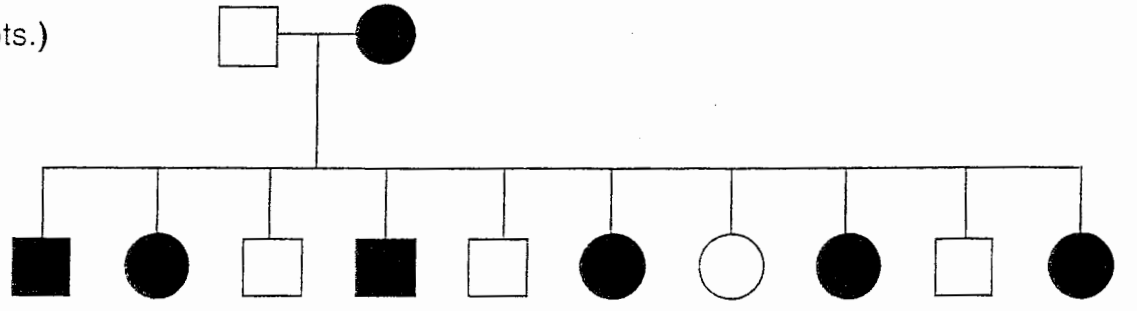
**(c** 12 pts.) Propose experiments involving BAC transgenes (but no knockouts) that would test whether the *pinkeye* mutation is in gene A or whether it is in gene B. Diagram your transgene(s), how it (or they) would integrate into the mouse genome, any crosses required, and the resulting genotypes.

BAC transgene rescue of pinkeye phenotype: Pronuclear injection to ♂ pronucleus of BAC3 to test for rescue with gene A or BAC2 to test for rescue with gene B. The injection into the ♂ pronucleus will be done in a pinkeye mutant, therefore you can test for rescue of pinkeye phenotype to give brown-eyes.

**Name:** _____

**3.** When setting out to determine the chromosomal location of a human disease gene by genetic linkage analysis (LOD scores), it is useful to calculate the theoretical maximum LOD score that a family of a given size and structure might contribute. For each of the families shown below, calculate, for $\theta = 0$, the LOD score that could be obtained using an SSR that is genetically inseparable from (shows no recombination with) the disease, which is autosomal dominant. Also indicate the minimum number of such families that would be required to achieve a publishable composite LOD score for $\theta = 0$. Assume that DNA samples are available for all living individuals.

(a 5 pts.)



For all these problems, we want to calculate the LOD score if we see no recombination between disease allele and SSR in offspring. In these families, the only informative meiosis is affected parent → offspring.

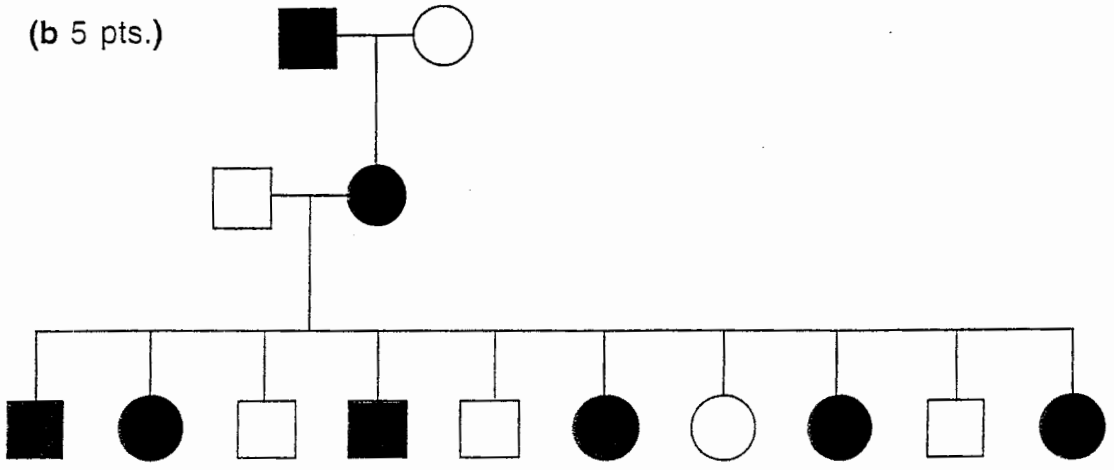
a) $LOD_0 = \log \frac{P(\text{data if linked})}{P(\text{data if unlinked})} = \log \frac{\frac{1}{2}(P \text{ if phase 1}) + \frac{1}{2}(P \text{ if phase 2})}{P(\text{data if unlinked})}$

$= \log \frac{\frac{1}{2}(\frac{1}{2})^{10} + \frac{1}{2}(0)^{10}}{(\frac{1}{4})^{10}} = 2.7$

2 such families would be needed.

**Name:** _____

(**b** 5 pts.)

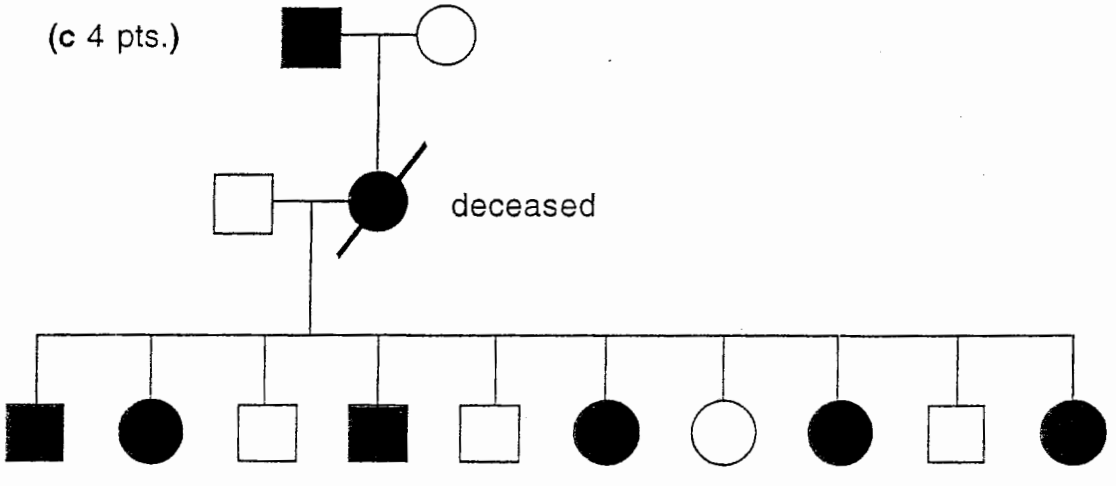


Phase is now known  P(data if linked) = P if phase 1

$$LOD_0 = \log \frac{(\frac{1}{2})^{10}}{(\frac{1}{4})^{10}} = 3.01$$

only 1 family is needed.

(**c** 4 pts.)

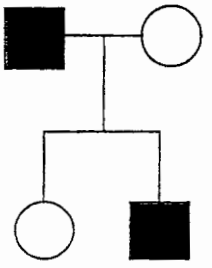



This is the same as above. With the grand parental genotype we can determine the phase of the mother.

$LOD_0 = 3.01$  only one family is needed

**Name:** _____

**(d** 4 pts.)

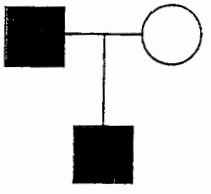


Phase is unknown.

$$LOD_0 \overset{\log}{=} \frac{\frac{1}{2}\left(\frac{1}{2}\right)^2 + \frac{1}{2}(0)^2}{\left(\frac{1}{4}\right)^2} = .301$$

10 such families would be needed

**(e** 4 pts.)

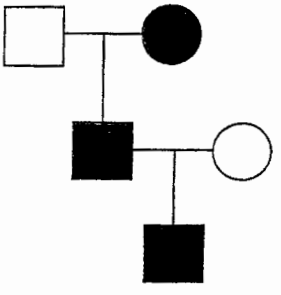


Phase is unknown

$$LOD_0 \overset{\log}{=} \frac{\frac{1}{2}\left(\frac{1}{2}\right) + \frac{1}{2}(0)}{\left(\frac{1}{4}\right)} = 0$$

This type of family cannot help prove the hypothesis.

**(f** 4 pts.)

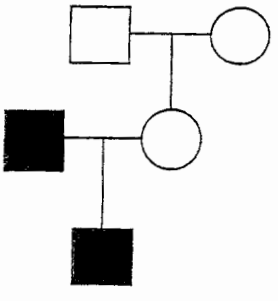


Phase is known.

$$LOD_0 = \log \frac{\left(\frac{1}{2}\right)}{\left(\frac{1}{4}\right)} = .301$$

10 such families would be needed.

**(g** 4 pts.)



Phase of the informative parent is unknown.

$$LOD_0 \overset{\log}{=} \frac{\frac{1}{2}\left(\frac{1}{2}\right) + \frac{1}{2}(0)}{\left(\frac{1}{4}\right)} = 0$$

This type of family cannot help prove the hypothesis.

# 7.03 Final Exam

Name: _____

Section: _____ TA: _____

There are 14 pages including this cover page.

Verify that you have all 14 pages.

Please write your name on each page.

| | |
|---|---|
| Question 1 | 25 points |
| Question 2 | 25 points |
| Question 3 | 30 points |
| Question 4 | 20 points |
| Question 5 | 32 points |
| Question 6 | 24 points |
| Question 7 | 24 points |
| Question 8 | 20 points |

**Name:** _____

**1.** **(a** 8 pts.**)** You are studying DNA replication in phage λ and you have isolated two different mutations in the **P** gene that will not form plaques on wild-type (**Su⁻**) *E. coli*, but will form plaques on an *E. coli* strain carrying an amber suppressor mutation (**Su⁺**). The two mutants are infected together into a **Su⁺** host so that each cell receives at least one phage of each type. The phage produced from this mixed infection form $10^9$ plaques/ml when plated on a **Su⁺** host, but only form $1.2 \times 10^7$ plaques/ml when plated on a **Su⁻** host. What is the distance between the two mutations in map units?

**(b** 10 pts.**)** The genome of phage λ is $5 \times 10^4$ bp long in physical length, and 200 map units long in genetic length. Say that the normal **P** protein is 40 kDa in size, and that the **P** protein produced by one of the mutants is 35 kDa. Using the map distance calculated in part **(a)** and the general approximation that the average mass of an amino acid 0.11 kDa, estimate the size of the P protein that will be produced by the other mutant.

**(c** 7 pts.**)** The mutagen hydroxylamine causes C•G to T•A mutations. If phage λ that carries an amber mutation in the **P** gene is treated with hydroxylamine will there be an increase in the frequency of revertant phage that can form plaques on a wild-type (**Su⁻**) *E. coli* host? Explain why or why not. (The amber stop codon is $^{5'}$UAG$^{3'}$, the ochre stop codon is $^{5'}$UAA$^{3'}$, and the opal stop codon is $^{5'}$UGA$^{3'}$).

**Name:** _____

**2.** (**a** 7 pts.) You have isolated an *E. coli* strain that carries a **Tn5** insertion that you think might be located close to the **Lac** operon (this insertion strain is kanamycin resistant (**Kan$^r$**)). You grow **P1** phage on this insertion strain and then infect a **LacZ$^-$** mutant (which does not express ß-galactosidase). Out of 100 **Kan$^r$** transductants, 30 are **Lac$^-$** and 70 express ß-galactosidase normally. What is the distance between the **Tn5** insertion and **LacZ$^-$** expressed as a cotransduction frequency?

(**b** 10 pts.) In order to determine on which side of the **Lac** operon the **Tn5** insertion resides, you grow **P1** on one of the **Kan$^r$ LacZ$^-$** transductants isolated in part (**a**). You then use this phage to infect a **LacY$^-$** recipient strain and isolate 100 **Kan$^r$** transductants. Each of the transductants is then tested for either **LacZ$^-$** (no ß-galactosidase) or **LacY$^-$** (no Lac permease). The following results were obtained:

| Genotype | Number of transductants |
|---|---|
| **LacZ$^-$ LacY$^+$** | 70 |
| **LacZ$^+$ LacY$^-$** | 28 |
| **LacZ$^+$ LacY$^+$** | 2 |

On the basis of these results, draw a map showing the relative positions of the **Tn5** insertion, the **LacZ$^-$**, and the **LacY$^-$** mutations. Also give any relevant distances expressed as cotransduction frequencies.

**Name:** _____

**(c** 8 pts.) You introduce an **F'** factor that carries the **Lac** operon into a strain with the **Tn5** insertion. You next isolate an **Hfr** strain that arises by recombination between the **Lac** operon on the **F'** factor and the **Lac** operon on the chromosome. You find that this **Hfr** strain will transfer **Kan$^r$** early and efficiently. On the basis of these results, draw a diagram of the original **F'** factor showing the relative positions of the **LacZ** and **LacY** genes to the orientation of the origin of transfer (**OriT**).

**3.** Yeast cells have a set of enzymes that can synthesize the amino acid histidine. You select one of these enzymes, histidinol dehydrogenase, as a reporter to study the regulation of the histidine pathway. First, you learn that the **His4** gene is the structural gene for histidinol dehydrogenase, and that recessive **His4$^-$** mutations don't express histidinol dehydrogenase. You also find that the **His4** gene is regulated; it is expressed when histidine is absent from the medium, but is not expressed when histidine is present. These results are summarized below:

| | Histidinol dehydrogenase activity | |
|---|---|---|
| | + histidine | – histidine |
| Wild type | – | + |
| **His4$^-$** | – | – |
| **His4$^-$ / His4$^+$** | – | + |

**(a** 5 pts.) You isolate a mutant, designated **His10$^-$**, which shows constitutive histidinol dehydrogenase expression. A cross of a **His4$^-$** mutant to a **His10$^-$** mutant gives diploids that show wild type expression of histidinol dehydrogenase. What does this result allow you to conclude about the **His10$^-$** mutant?

**Name:** _____

**(b** 7 pts.**)** When the diploids from part (**a**) are sporulated, the resulting tetrads are of three types with respect to the expression/regulation of histidinol dehydrogenase. Out of a total of 50 tetrads, 30 are Type 1, 16 are Type 2, and 4 are Type 3.

| Type 1 | Type 2 | Type3 |
|---|---|---|
| constitutive | uninducible | uninducible |
| constitutive | uninducible | uninducible |
| uninducible | constitutive | regulated |
| uninducible | regulated | regulated |

Are the **His4⁻** and **His10⁻** mutations linked? If so, how far apart are they in cM?

**(c** 8 pts.**)** Next, you isolate a mutation, designated **His11⁻**, which has uninducible histidinol dehydrogenase expression. A cross of a **His4⁻** mutant to a **His11⁻** mutant gives diploids that show wild-type expression of histidinol dehydrogenase. On the basis of this result and the results for parts (**a**) and (**b**), diagram two different possible regulatory pathways that can explain the functions of the **His10** and **His11** gene products in the regulation of **His4**. Be sure to include a role for the regulatory action of histidine in your answer.

**Name:** _____

**(d** 5 pts.) A cross of a **His10⁻** mutant to an **His11⁻** mutant gives the following tetrad types. Out of a total of 50 tetrads, 35 are Type 1, 8 are Type 2, and 7 are Type 3.

| Type 1 | Type 2 | Type3 |
|---|---|---|
| uninducible | uninducible | constitutive |
| uninducible | uninducible | constitutive |
| constitutive | regulated | uninducible |
| regulated | regulated | uninducible |

Is a **His10⁻ His11⁻** double mutant regulated, constitutive, or uninducible?

**(e** 5 pts.) On the basis of your answer for part (**d**) and from the rest of the information given in this problem, diagram the regulatory model that best explains the functions of the **His10** and **His11** gene products and histidine.

**Name:** _____

**4.** As discussed in 7.03, HNPCC is an autosomal dominant predisposition to cancer of the colon and other organs. As discussed, HNPCC is the result of mutation in one or another component of the mismatch repair system.

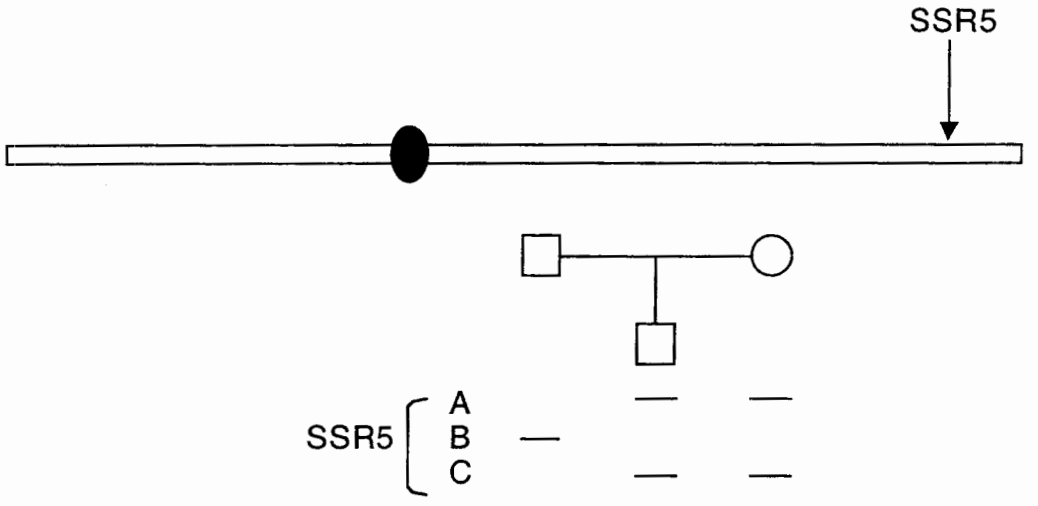
**(a** 5 pts.**)** In HNPCC individuals, do non-cancerous tissues display SSR instability? Why or why not?

**(b** 5 pts.**)** You plan to perform genetic linkage studies (using SSRs) in families with HNPCC. For the affected individuals in these families, you have access to genomic DNA samples from normal white blood cells and from cancerous tumors. For purposes of genetic linkage analysis, would you prefer to type SSRs in tumor DNAs or in blood DNAs? Why?

**(c** 10 pts.**)** The yeast **MSH2** gene encodes a component of the mismatch repair system. You obtain a haploid yeast strain that is mutant in **MSH2** and that exhibits the "mutator phenotype." In yeast, would you expect this mutator phenotype to be dominant or recessive? Briefly justify your prediction, and briefly outline an experiment to test your prediction.

**Name:** _____

**5.** While home on winter break, an uncle who is a physician asks you to interpret genetic findings in a Klinefelter (47,XXY) boy. Your uncle explains that he has typed the boy and his parents for one X-linked SSR, whose location on the X chromosome is shown here:



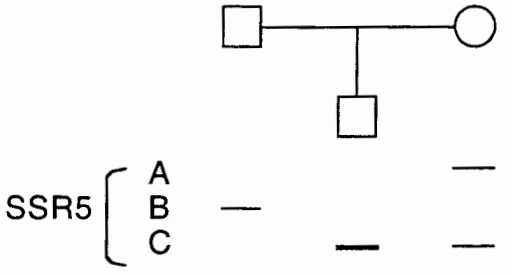

**(a** 2 pts) Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer.

**(b** 5 pts) Could nondisjunction have occurred in meiosis I? If so, sketch the meiotic event in which nondisjunction occurred; include SSR5 in your sketch.

**(c** 5 pts) Could nondisjunction have occurred in meiosis II? If so, sketch the meiotic event in which nondisjunction occurred; include SSR5 in your sketch.

**Name:** _____

Your uncle then tells you of a second couple with a Klinefelter (XXY) son:

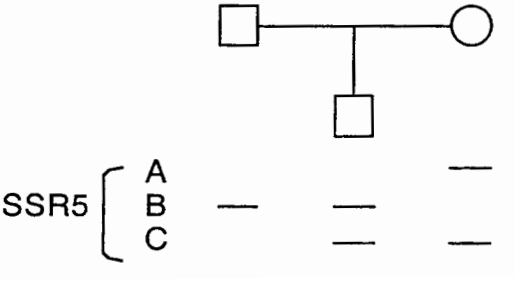



**(d** 2 pts**)** Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer.

**(e** 4 pts**)** Could nondisjunction have occurred in meiosis I? Briefly explain your answer (no sketch required).

**(f** 4 pts**)** Could nondisjunction have occurred in meiosis II? Briefly explain your answer (no sketch required).

Finally, your uncle tells you of a third couple with a Klinefelter (XXY) son:



**Name:** _____

**(g** 2 pts**)** Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer.

**(h** 4 pts**)** Could nondisjunction have occurred in meiosis I? Briefly explain your answer (no sketch required).

**(i** 4 pts**)** Could nondisjunction have occurred in meiosis II? Briefly explain your answer (no sketch required).

**6.** Red-green color blindness is an X-linked recessive trait, and it maps very close to SSR5 as shown in the preceding question. In the United States, 5% of males are red-green colorblind. **Assume random mating** and Hardy-Weinberg equilibrium.

**(a** 4 pts**)** What is the frequency of red-green colorblindness among females in the US?

**(b** 4 pts**)** What is the frequency of red-green colorblindness among US females whose fathers are red-green colorblind?

**(c** 4 pts**)** What is the frequency of red-green colorblindness among US females who have one or more brothers who are red-green colorblind but whose mothers have normal color vision?

**Name:** _____

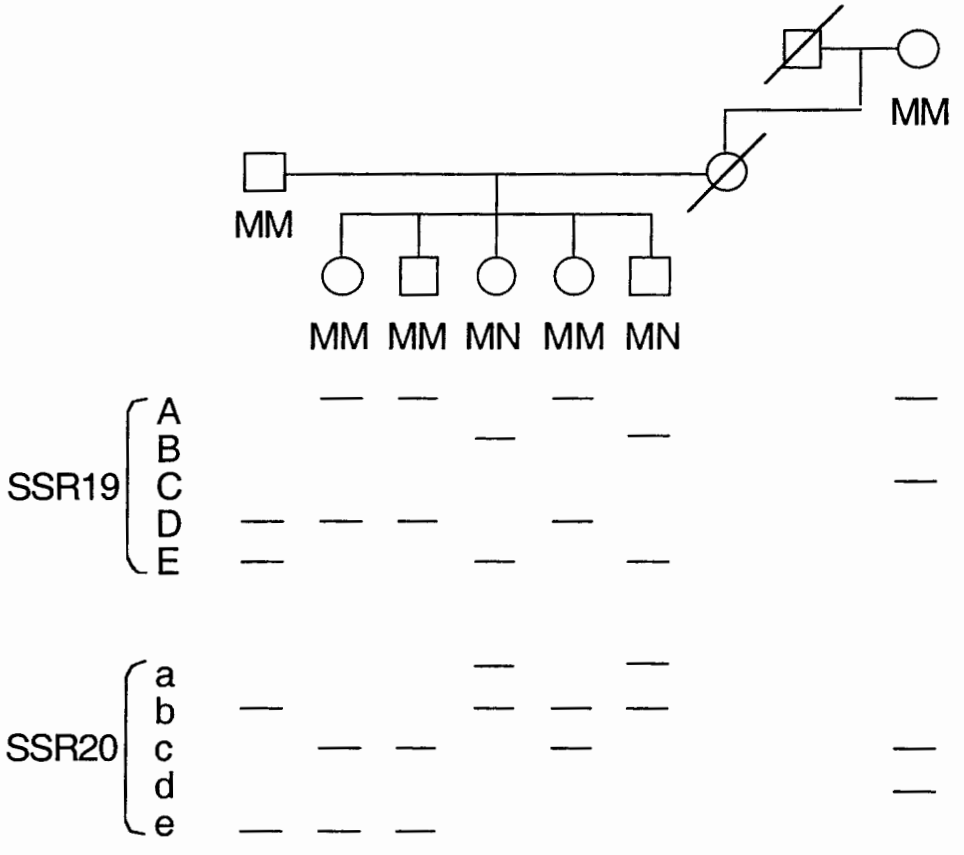
**(d** 4 pts) If all XXY males were due to meiotic nondisjunction in fathers, what would the frequency of red-green colorblindness be among XXY males in the US?

**(e** 4 pts) If all XXY males were due to mitotic nondisjunction after fertilization, what would the frequency of red-green colorblindness be among XXY males in the US?

**(f** 4 pts) Using a different SSR (that is tightly linked to the centromere of the X chromosome), you find that a particular XXY boy inherited two X chromosomes from his mother as a result of nondisjunction in meiosis I. Both the mother and the father have normal color vision, but the XXY son is red-green colorblind. How can you explain the XXY boy's red-green colorblindness (without appealing to new mutations)?

**Name:** _____

**7.** As described in 7.03, M and N are different red-blood-cell antigens specified by alleles of the same gene, which we will call **RBCA**. The M and N antigens are codominant, so a simple blood test can distinguish among the three possible genotypes. You set out to genetically map the **RBCA** gene by linkage analysis in families like the one shown here, where the **RBCA** genotypes of the living individuals are indicated:




**(a** 2 pts.**)** What allele at **RBCA** did the mother (deceased) inherit from the grandfather (deceased)?

**(b** 2 pts.**)** What allele at SSR19 did the mother (deceased) inherit from the grandfather (deceased)?

**Name:** _____

**(c** 2 pts.**)** Diagram the phase relationship(s) between the **RBCA** and SSR19 alleles in the mother (deceased).

**(d** 6 pts.**)** Calculate the LOD score for linkage at $\theta = 0$ between **RBCA** and SSR19 in this family.

**(e** 3 pts.**)** Diagram the phase relationship(s) between the SSR19 and SSR20 alleles in the mother (deceased).

**(f** 3 pts.**)** Diagram the phase relationship(s) between the SSR19 and SSR20 alleles in the father (living).

**(g** 6 pts.**)** Calculate the LOD score for linkage at $\theta = 0.1$ between SSR19 and SSR20 in this family.

**Name:** _____

**8.** Recall our discussion of the disease retinitis pigmentosa (RP), an example of nonallelic heterogeneity, where at least 66 genetic loci have been implicated. As we discussed, RP exhibits autosomal recessive inheritance in 84% of families with affected individuals.

Let's now simplify this complex situation. Let's suppose that you've discovered an isolated human population in which RP affects one in 8000 individuals and in which RP always exhibits autosomal recessive inheritance. The RP phenotype is 100% penetrant. In this isolated population, only two genes are implicated in RP: the **RP6** gene on chromosome 6, and the **RP12** gene on chromosome 12. DNA sequencing of the genomes of 100 RP patients selected at random from this population reveals that 80 patients are homozygous for a mutation in the **RP6** gene, while the remaining 20 patients are homozygous for a mutation in the **RP12** gene. In answering the following questions, assume random mating and Hardy-Weinberg equilibrium unless specified otherwise.

**(a** 2 pts.**)** What rate of concordance for the RP phenotype would you expect among monozygotic twins in this isolated population?

**(b** 3 pts.**)** Among dizygotic twins?

**(c** 5 pts.**)** What is the frequency of the mutant allele at **RP6** in this isolated population?

**(d** 5 pts.**)** What is the frequency of the mutant allele at **RP12** in this isolated population?

**(e** 5 pts.**)** What would the frequency of RP be among individuals in this population whose parents are first cousins?

# 7.03 Final Exam

Name: KEY

Section: _____ TA: _____

There are 14 pages including this cover page.

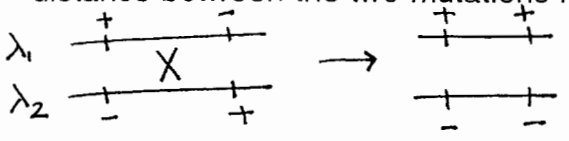
Verify that you have all 14 pages.

Please write your name on each page.

| | |
|---|---|
| Question 1 | 25 points |
| Question 2 | 25 points |
| Question 3 | 30 points |
| Question 4 | 20 points |
| Question 5 | 32 points |
| Question 6 | 24 points |
| Question 7 | 24 points |
| Question 8 | 20 points |

**Name:** _____

**1.** (**a** 8 pts.) You are studying DNA replication in phage λ and you have isolated two different mutations in the **P** gene that will not form plaques on wild-type (**Su⁻**) *E. coli*, but will form plaques on an *E. coli* strain carrying an amber suppressor mutation (**Su⁺**). The two mutants are infected together into a **Su⁺** host so that each cell receives at least one phage of each type. The phage produced from this mixed infection form $10^9$ plaques/ml when plated on a **Su⁺** host, but only form $1.2 \times 10^7$ plaques/ml when plated on a **Su⁻** host. What is the distance between the two mutations in map units?




$1.2 \times 10^7$

Total Recombinants: $2.4 \times 10^7$
Total Phage : $10^9$

$$mu = 100 \times \frac{2.4 \times 10^7}{10^9} = 2.4 \text{ mu}$$

(**b** 10 pts.) The genome of phage λ is $5 \times 10^4$ bp long in physical length, and 200 map units long in genetic length. Say that the normal **P** protein is 40 kDa in size, and that the **P** protein produced by one of the mutants is 35 kDa. Using the map distance calculated in part (**a**) and the general approximation that the average mass of an amino acid 0.11 kDa, estimate the size of the P protein that will be produced by the other mutant.

Distance between mutations : $2.4 \text{ mu} \times \frac{5 * 10^4 \text{ bp}}{200 \text{ mu}} = 600 \text{ bp} = 200 \text{ aa} = 22 \text{ kDa}$

Mutant protein must be < 40 kDa

size = 35 kDa − 22 kDa = 13 kDa

(**c** 7 pts.) The mutagen hydroxylamine causes C•G to T•A mutations. If phage λ that carries an amber mutation in the **P** gene is treated with hydroxylamine will there be an increase in the frequency of revertant phage that can form plaques on a wild-type (**Su⁻**) *E. coli* host? Explain why or why not. (The amber stop codon is 5'UAG3', the ochre stop codon is 5'UAA3', and the opal stop codon is 5'UGA3').

5'-UAG-3' 5'UAA3' ochre stop

DNA 5' TAG → 5' TAA
ATC 5' ATT 5'

No, the nonsense amber mutation can only be changed to ochre stop. Therefore, no revertants are created.

**Name:** _____

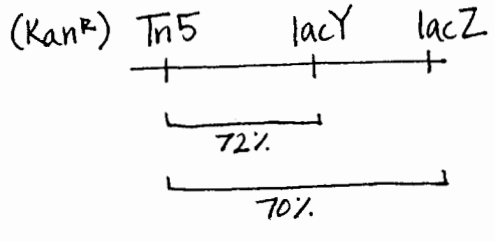
**2.** (**a** 7 pts.) You have isolated an *E. coli* strain that carries a **Tn5** insertion that you think might be located close to the **Lac** operon (this insertion strain is kanamycin resistant (**Kan$^r$**)). You grow **P1** phage on this insertion strain and then infect a **LacZ$^-$** mutant (which does not express ß-galactosidase). Out of 100 **Kan$^r$** transductants, 30 are **Lac$^-$** and 70 express ß-galactosidase normally. What is the distance between the **Tn5** insertion and **LacZ$^-$** expressed as a cotransduction frequency?

The LacZ$^+$ gene was co-transduced with the Kan$^R$ on Tn5 70% of the time.

(**b** 10 pts.) In order to determine on which side of the **Lac** operon the **Tn5** insertion resides, you grow **P1** on one of the **Kan$^r$ LacZ$^-$** transductants isolated in part (**a**). You then use this phage to infect a **LacY$^-$** recipient strain and isolate 100 **Kan$^r$** transductants. Each of the transductants is then tested for either **LacZ$^-$** (no ß-galactosidase) or **LacY$^-$** (no Lac permease). The following results were obtained:

| Genotype | Number of transductants |
|---|---|
| **LacZ$^-$ LacY$^+$** | 70 |
| **LacZ$^+$ LacY$^-$** | 28 |
| **LacZ$^+$ LacY$^+$** | 2 |

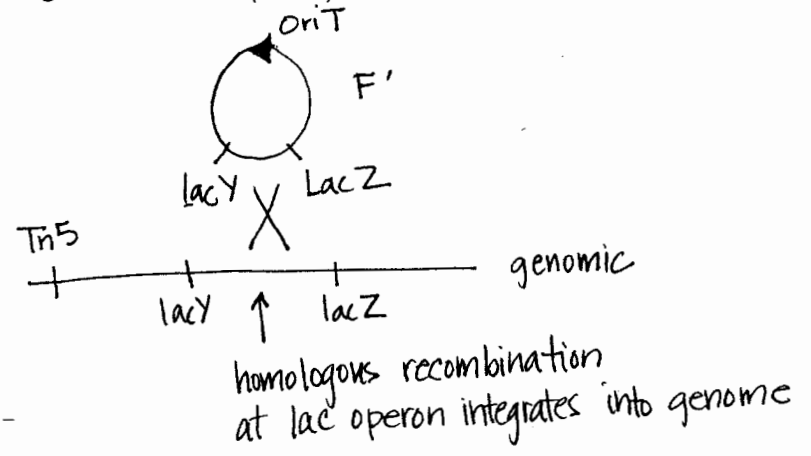
On the basis of these results, draw a map showing the relative positions of the **Tn5** insertion, the **LacZ$^-$**, and the **LacY$^-$** mutations. Also give any relevant distances expressed as cotransduction frequencies.




Rare class is 4 cross-over class ($lacZ^-Y^-$)

**Name:** _____

**(c** 8 pts.) You introduce an **F'** factor that carries the **Lac** operon into a strain with the **Tn5** insertion. You next isolate an **Hfr** strain that arises by recombination between the **Lac** operon on the **F'** factor and the **Lac** operon on the chromosome. You find that this **Hfr** strain will transfer **Kan$^r$** early and efficiently. On the basis of these results, draw a diagram of the original **F'** factor showing the relative positions of the **LacZ** and **LacY** genes to the orientation of the origin of transfer (**OriT**).




**3.** Yeast cells have a set of enzymes that can synthesize the amino acid histidine. You select one of these enzymes, histidinol dehydrogenase, as a reporter to study the regulation of the histidine pathway. First, you learn that the **His4** gene is the structural gene for histidinol dehydrogenase, and that recessive **His4$^-$** mutations don't express histidinol dehydrogenase. You also find that the **His4** gene is regulated; it is expressed when histidine is absent from the medium, but is not expressed when histidine is present. These results are summarized below:

| | Histidinol dehydrogenase activity | |
|---|---|---|
| | + histidine | – histidine |
| Wild type | – | + |
| **His4$^-$** | – | – |
| **His4$^-$ / His4$^+$** | – | + |

**(a** 5 pts.) You isolate a mutant, designated **His10$^-$**, which shows constitutive histidinol dehydrogenase expression. A cross of a **His4$^-$** mutant to a **His10$^-$** mutant gives diploids that show wild type expression of histidinol dehydrogenase. What does this result allow you to conclude about the **His10$^-$** mutant?

his10 ⟶ histidinol

$\frac{his4^- \; his10^+}{his4^+ \; his10^-}$ his10 and his4 are recessive

his10 is constitutive, recessive

**Name:** _____

**(b** 7 pts.**)** When the diploids from part (**a**) are sporulated, the resulting tetrads are of three types with respect to the expression/regulation of histidinol dehydrogenase. Out of a total of 50 tetrads, 30 are Type 1, 16 are Type 2, and 4 are Type 3.

| Type 1 | Type 2 | Type3 |
|---|---|---|
| constitutive | uninducible | uninducible |
| constitutive | uninducible | uninducible |
| uninducible | constitutive | regulated |
| uninducible | regulated | regulated |
| PD | T | NPD |

Are the **His4⁻** and **His10⁻** mutations linked? If so, how far apart are they in cM?

His4 and His10 are linked because PD >> NPD

$$\frac{T + 6NPD}{2\Sigma} \times 100 = cM$$

$$\frac{16 + 6(4)}{2(50)} \times 100 = 40\,cM$$

**(c** 8 pts.**)** Next, you isolate a mutation, designated **His11⁻**, which has uninducible histidinol dehydrogenase expression. A cross of a **His4⁻** mutant to a **His11⁻** mutant gives diploids that show wild-type expression of histidinol dehydrogenase. On the basis of this result and the results for parts (**a**) and (**b**), diagram <u>two</u> different possible regulatory pathways that can explain the functions of the **His10** and **His11** gene products in the regulation of **His4**. Be sure to include a role for the regulatory action of histidine in your answer.

his11 ⟶ histidinol

his10 ⊣ histidinol

histidine ⊣ histidinol

Model 1

histidine ⟶ his10 ⊣ his11 ⟶ histidinol

Model 2

histidine ⊣ his11 ⊣ his10 ⊣ histidinol

**Name:** _____

**(d** 5 pts.) A cross of a **His10⁻** mutant to an **His11⁻** mutant gives the following tetrad types. Out of a total of 50 tetrads, 35 are Type 1, 8 are Type 2, and 7 are Type 3.

| Type 1 | Type 2 | Type3 |
|---|---|---|
| uninducible | uninducible | constitutive |
| uninducible | uninducible | constitutive |
| constitutive | regulated | uninducible |
| regulated | regulated | uninducible |

Is a **His10⁻ His11⁻** double mutant regulated, constitutive, or uninducible?

Uninducible inferred from Type 2 (NPD)

**(e** 5 pts.) On the basis of your answer for part (**d**) and from the rest of the information given in this problem, diagram the regulatory model that best explains the functions of the **His10** and **His11** gene products and histidine.

his11 is epistatic to his10

histidine ⟶ his10 ⟶| his11 ⟶ histidinol

**Name:** _____

**4.** As discussed in 7.03, HNPCC is an autosomal dominant predisposition to cancer of the colon and other organs. As discussed, HNPCC is the result of mutation in one or another component of the mismatch repair system.

**(a** 5 pts.) In HNPCC individuals, do non-cancerous tissues display SSR instability? Why or why not?

No, because non-cancerous cells are normal

**(b** 5 pts.) You plan to perform genetic linkage studies (using SSRs) in families with HNPCC. For the affected individuals in these families, you have access to genomic DNA samples from normal white blood cells and from cancerous tumors. For purposes of genetic linkage analysis, would you prefer to type SSRs in tumor DNAs or in blood DNAs? Why?

Blood DNA because tumor DNAs show SSR instability

**(c** 10 pts.) The yeast **MSH2** gene encodes a component of the mismatch repair system. You obtain a haploid yeast strain that is mutant in **MSH2** and that exhibits the "mutator phenotype." In yeast, would you expect this mutator phenotype to be dominant or recessive? Briefly justify your prediction, and briefly outline an experiment to test your prediction.

Recessive, it is a loss of function mutation.

Mate $MSH2^-$ with $MSH2^+$ (opposite mating type)

↓

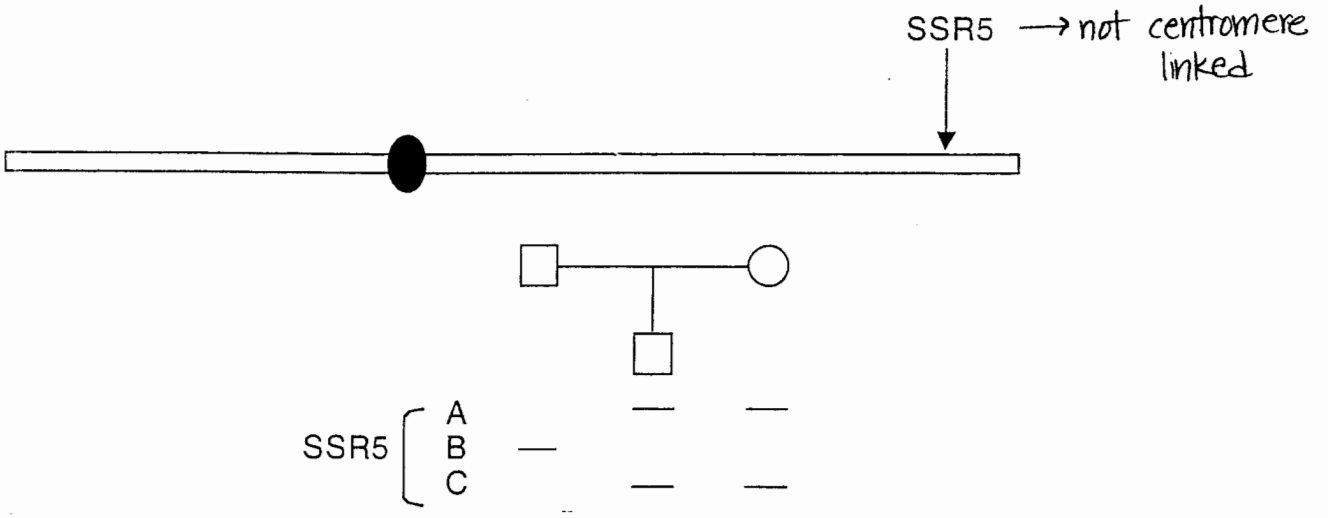
$MSH2^-/MSH2^+$ observe phenotype of diploid

Wild-type ⇒ recessive

"mutator" ⇒ dominant

**Name:** _____

**5.** While home on winter break, an uncle who is a physician asks you to interpret genetic findings in a Klinefelter (47,XXY) boy. Your uncle explains that he has typed the boy and his parents for one X-linked SSR, whose location on the X chromosome is shown here:

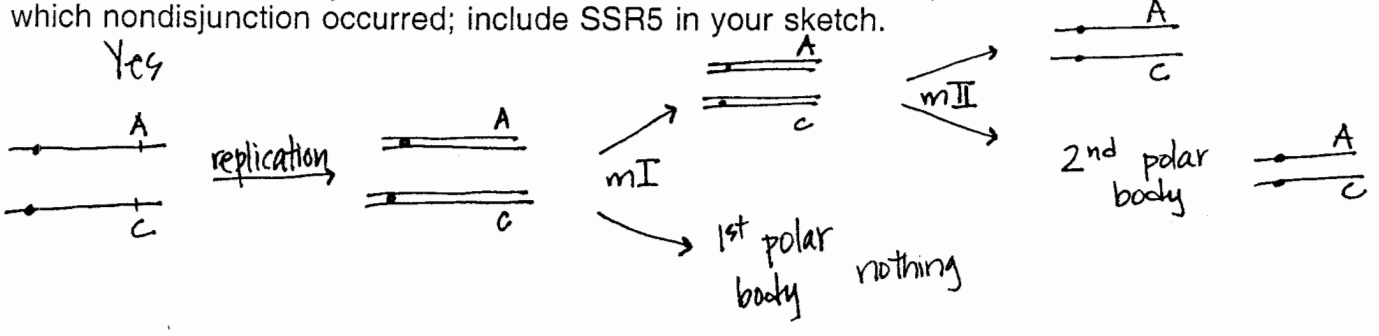



**(a** 2 pts) Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer.

No. The child received 2 different SSR alleles, meaning 2 X-chromosomes came from the mom.

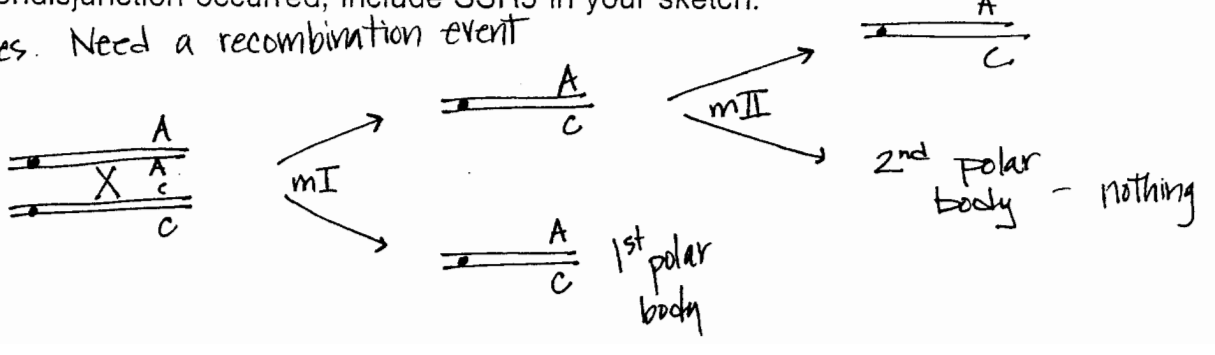
**(b** 5 pts) Could nondisjunction have occurred in meiosis I? If so, sketch the meiotic event in which nondisjunction occurred; include SSR5 in your sketch.

Yes



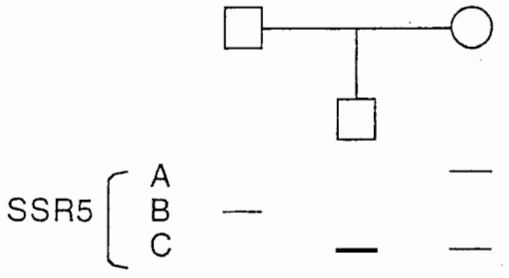

**(c** 5 pts) Could nondisjunction have occurred in meiosis II? If so, sketch the meiotic event in which nondisjunction occurred; include SSR5 in your sketch.

Yes. Need a recombination event



**Name:** _____

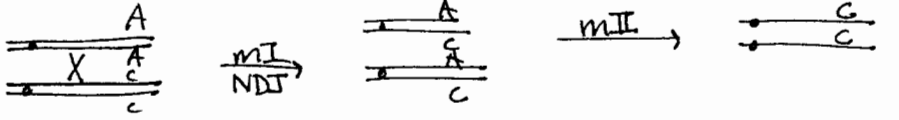
Your uncle then tells you of a second couple with a Klinefelter (XXY) son:



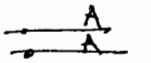

**(d** 2 pts) Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer.

Yes. After normal meiotic events, the child could have received one copy of the c allele from mom. A nondisjunction in mitosis caused the duplicated C allele.

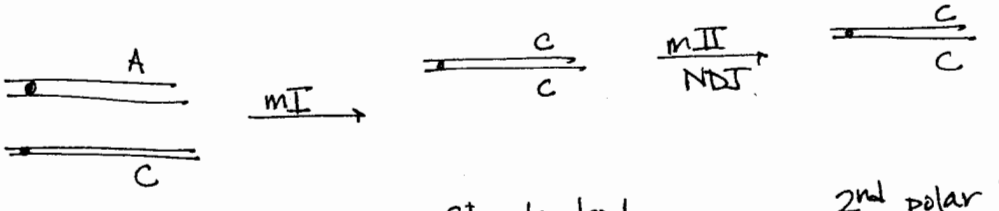
**(e** 4 pts) Could nondisjunction have occurred in meiosis I? Briefly explain your answer (no sketch required). Yes. Need a recombination event.

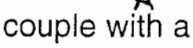



1st polar body – nothing

2nd polar body 

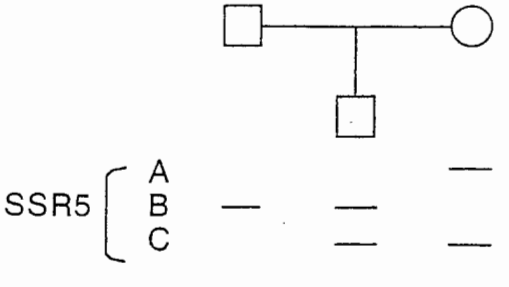
**(f** 4 pts) Could nondisjunction have occurred in meiosis II? Briefly explain your answer (no sketch required). Yes.




1st polar body 

2nd polar body – nothing

Finally, your uncle tells you of a third couple with a Klinefelter (XXY) son:

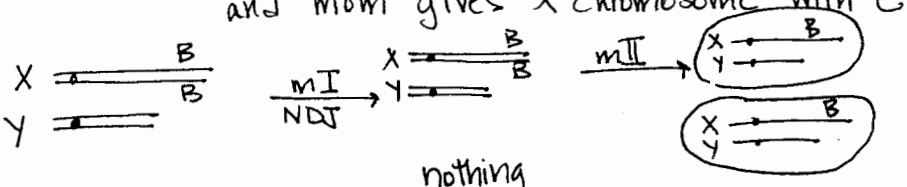



Nondisjunction in Dad

**Name:** _____

(g 2 pts) Could nondisjunction have occurred after fertilization, in mitosis? Briefly explain your answer. No. The child received an X and a Y from dad, therefore NDJ must have happened in father's meiosis

(h 4 pts) Could nondisjunction have occurred in meiosis I? Briefly explain your answer (no sketch required). Yes. Dad is giving Y chromosome and X chromosome with B allele and mom gives X chromosome with C allele ⇒ $X^{B}X^{B}Y$




nothing

(i 4 pts) Could nondisjunction have occurred in meiosis II? Briefly explain your answer (no sketch required). No. Homologs separate in meiosis I. so there is no way to get X and Y together in meiosis II. Meiosis II NDJ would give either two Xs or two Ys.

(No recombination between X & Y before meiosis would cause this. If B translocated to Y chromosome, Dad's gamete would be $Y^{SSR5}$ which doesn't lead to a XXY child.)

**6.** Red-green color blindness is an X-linked recessive trait, and it maps very close to SSR5 as shown in the preceding question. In the United States, 5% of males are red-green colorblind. **Assume random mating** and Hardy-Weinberg equilibrium.

(a 4 pts) What is the frequency of red-green colorblindness among females in the US?

$f(X^{cb}) = q = .05$

$f(X^{cb}X^{cb}) = q^2 = .0025$

(b 4 pts) What is the frequency of red-green colorblindness among US females whose fathers are red-green colorblind?

$P(X^{cb}X^{cb}) = p(\text{dad gives } X^{cb}) \times p(\text{mom gives } X^{cb})$

$= 1 \times [2pq(\tfrac{1}{2}) + q^2(1)]$

$= .0475 + .0025$

$= .05$

(c 4 pts) What is the frequency of red-green colorblindness among US females who have one or more brothers who are red-green colorblind but whose mothers have normal color vision?

$p(X^{cb}X^{cb}) = p(\text{mom het})\, p(\text{mom gives } X^{cb})\, p(\text{dad gives } X^{cb})$

$= (1)(.5)(1)(.05)$

$= .025$

**Name:** _____

**(d 4 pts)** If all XXY males were due to meiotic nondisjunction in fathers, what would the frequency of red-green colorblindness be among XXY males in the US?

$P(X^{cb}X^{cb}Y) = P(\text{dad } X^{cb}Y)\ P(\text{mom gives } X^{cb})$ → from part b

$= (.05)(.05)$

$= .0025$

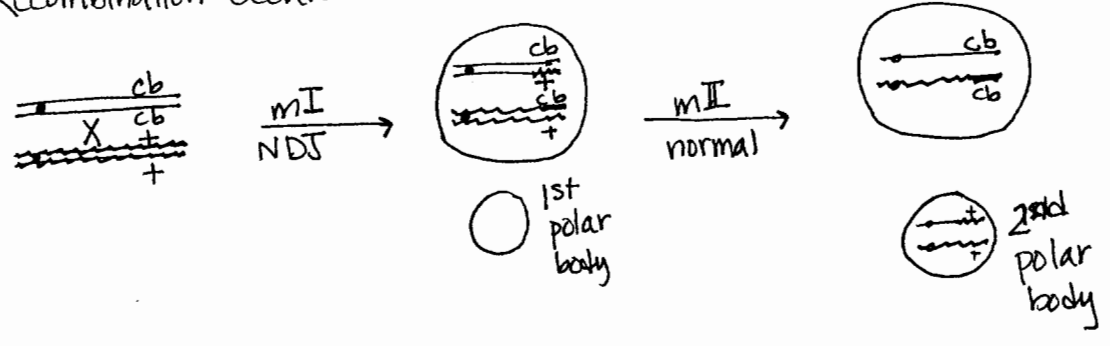
**(e 4 pts)** If all XXY males were due to mitotic nondisjunction after fertilization, what would the frequency of red-green colorblindness be among XXY males in the US?

$P(\text{mom gives } X^{cb}) = .05$

**(f 4 pts)** Using a different SSR (that is tightly linked to the centromere of the X chromosome), you find that a particular XXY boy inherited two X chromosomes from his mother as a result of nondisjunction in meiosis I. Both the mother and the father have normal color vision, but the XXY son is red-green colorblind. How can you explain the XXY boy's red-green colorblindness (without appealing to new mutations)?

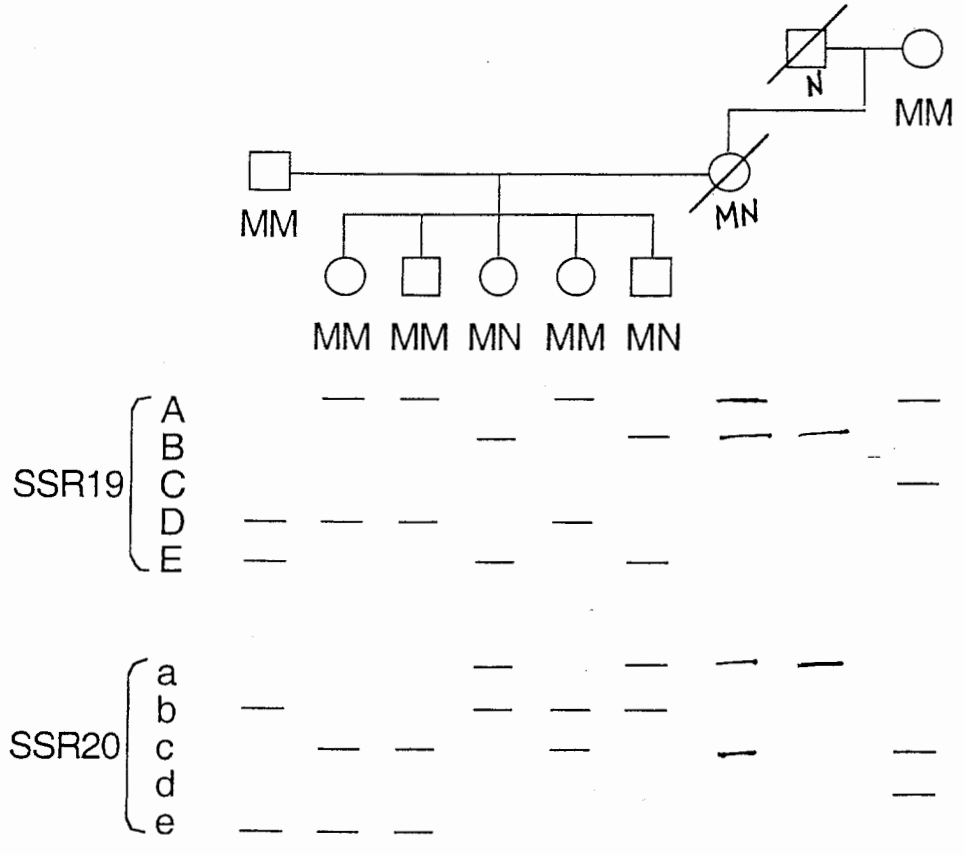
Mom is a heterozygote $X^{cb}X^{+}$

Recombination occurs in mother before meiosis I



**Name:** _____

**7.** As described in 7.03, M and N are different red-blood-cell antigens specified by alleles of the same gene, which we will call **RBCA**. The M and N antigens are codominant, so a simple blood test can distinguish among the three possible genotypes. You set out to genetically map the **RBCA** gene by linkage analysis in families like the one shown here, where the **RBCA** genotypes of the living individuals are indicated:




**(a** 2 pts.) What allele at **RBCA** did the mother (deceased) inherit from the grandfather (deceased)?

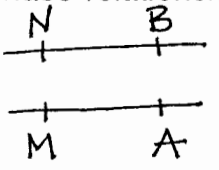
N

**(b** 2 pts.) What allele at SSR19 did the mother (deceased) inherit from the grandfather (deceased)?

B

**Name:** _____

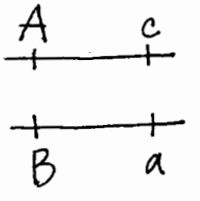
**(c** 2 pts.) Diagram the phase relationship(s) between the **RBCA** and SSR19 alleles in the mother (deceased).




**(d** 6 pts.) Calculate the LOD score for linkage at $\theta = 0$ between **RBCA** and SSR19 in this family.

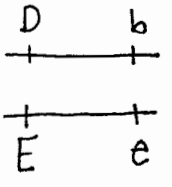
$$LOD_{\theta=0} = \log_{10} \frac{(\frac{1}{2})^5}{(\frac{1}{4})^5} = 1.505$$

**(e** 3 pts.) Diagram the phase relationship(s) between the SSR19 and SSR20 alleles in the mother (deceased).

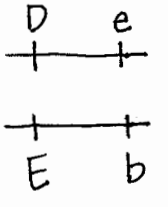



**(f** 3 pts.) Diagram the phase relationship(s) between the SSR19 and SSR20 alleles in the father (living).

Phase 1




Phase 2




**(g** 6 pts.) Calculate the LOD score for linkage at $\theta = 0.1$ between SSR19 and SSR20 in this family.

♀ $LOD_{\theta=.1} = \log \frac{(.45)^5}{(.25)^5} = 1.276$

♂ $LOD_{\theta=.1} = \log \frac{\frac{1}{2}(.05)^4(.45) + \frac{1}{2}(.45)^4(.05)}{(.25)^5} = .022$

$LOD_{\theta=.1} = 1.276 + 0.022$

$= 1.298$

**Name:** _____

**8.** Recall our discussion of the disease retinitis pigmentosa (RP), an example of nonallelic heterogeneity, where at least 66 genetic loci have been implicated. As we discussed, RP exhibits autosomal recessive inheritance in 84% of families with affected individuals.

Let's now simplify this complex situation. Let's suppose that you've discovered an isolated human population in which RP affects one in 8000 individuals and in which RP always exhibits autosomal recessive inheritance. The RP phenotype is 100% penetrant. In this isolated population, only two genes are implicated in RP: the **RP6** gene on chromosome 6, and the **RP12** gene on chromosome 12. DNA sequencing of the genomes of 100 RP patients selected at random from this population reveals that 80 patients are homozygous for a mutation in the **RP6** gene, while the remaining 20 patients are homozygous for a mutation in the **RP12** gene. In answering the following questions, assume random mating and Hardy-Weinberg equilibrium unless specified otherwise.

**(a** 2 pts.) What rate of concordance for the RP phenotype would you expect among monozygotic twins in this isolated population?

100%

**(b** 3 pts.) Among dizygotic twins?

25%

**(c** 5 pts.) What is the frequency of the mutant allele at **RP6** in this isolated population?

$$q_{RP6} = \sqrt{q^2 \times p(RP6)}$$

$$= \sqrt{\frac{1}{8000} \times 0.8} = .01$$

**(d** 5 pts.) What is the frequency of the mutant allele at **RP12** in this isolated population?

$$q_{RP12} = \sqrt{q^2 \times p(RP12)}$$

$$= \sqrt{\frac{1}{8000} \times 0.2} = .005$$

**(e** 5 pts.) What would the frequency of RP be among individuals in this population whose parents are first cousins?

$$q_{RP} = F q_{RP6} + F q_{RP12}$$

$$= \frac{1}{16}(.01) + \frac{1}{16}(.005)$$

$$= 9.375 \times 10^{-4}$$